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Annex

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International Conference on Sustainable Agriculture and Food Security: *Challenges and Opportunities*

Bandung-Indonesia, 27-28 September 2011

Proceeding

(Oral Papers)

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UNIVERSITAS PADJADJARAN

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Universitas Padjadjaran
Indonesia
2012

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FOREWORD

Agriculture as one of leading economic sectors in some countries, is currently facing many problems. This situation could be overcomed by policy and institutional environment which is condusive to increase agricultural productivity while maintaining a sustainable agriculture development and food security. According to this, it is required to develop strategies, a new paradigm, and holistic approach to support the agricultural growth continuum.

In order to make a significant contribution to the better understanding of sustainable agriculture for meeting food security needs and addressing climate change challenges, an International Conference on Sustainable Agriculture and Food Security was held in Bandung Indonesia on 27-28 September 2011. This conference was organized by collaboration of four faculties in Universitas Padjadjaran: Faculty of Agriculture, Faculty of Animal Husbandry, Faculty of Fishery and Marine Science, and Faculty of Agricultural Industrial Technology. Ministry of Agriculture of Republic Indonesia and internationally well-known experts from USA, Finlandia, Singapore, Germany, Malaysia, Romania, Republic of Serbia, China as well as Indonesia were invited as resource speakers.

More than 250 participants from 15 countries attended the conference. The conference shared experiences and views regarding agricultural production in a changing environment towards sustainable agriculture development to maintain food security, and stimulated cooperative research among participating institutions.

About 180 papers are presented and the committee hopes that these papers will be a lsating record of the contributions to this conference and a useful reference for all practitioners in the fields of agriculture in general. Some of the topics presented include critical issues dealing with sustainable agriculture and food security, agrosocio-economy, agritechnology, plant sciences, animal production, and food technology. The committee would like to thank the many reviewers of the papers for their contribution to these proceedings.

The conference and proceeding would have not been accomplished without the support of many individuals, groups and academic units. We owe our gratitude to those who commit and dedicate their self to this conference.

Benny Joy
Chair of ICSAFS

CONTENTS

INVITED SPEAKERS

New Technologies for The Improvement of Yield and Quality of Beef of Domestic Spotted Breed

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Introduction

The future of beef meat export in Serbia lies in the first place in the production of meat of such quality that meets the demands of a target market. In this regard, on the basis of our previous experience and production of beef for already known customers and according to their demands, the markets could be divided into two following groups: Italian and Greek market. A prerequisite for any kind of beef export is: identification and registration of all heads of cattle on the territory of the Republic of Serbia, application of international standards of control and monitoring of the production process and implementation of EU regulations relating to animal welfare (Official Journal C 340, 1997). Besides these mentioned prerequisites it is also necessary to provide high quality fattening animals, that is, female calves for the production of the meat type called "baby beef". Baby beef fattening means that the duration of fattening is 11.5 months and that during that time realized body weights of females should be 455 kg, and of males 530 kg. For Greek market both females and males are produced, and as regards the way of processing a standard medial dissection is demanded. For Italian market, especially, the meat from female cattle not older than 11,5 months is required. This market requires the cutting of carcass to be performed by so called Milanese cut. Contrary to increasing demands for animals intended for fattening, there is a decline in number of calves and young cattle (Republic Bureau of Statistics, 2010). One of the ways of fast and efficient production of quality calves for fattening is the application of the method of industrial crossing of Domestic Spotted breed of lower production traits (Aleksic *et al*., 1996 and 1999). With aim of improvement of production and quality of meat the trend of crossing of existing genotypes of cattle with fattening bulls is more and more present. Application of method of crossing enables demonstration of heterosis effect of fattening and meat quality traits in offspring (F_1 generation). Results of the crossing with fattening breeds (Aleksić *et al*., 2005 and 2006.) indicate that considerable effects in production of calves, fattening of young cattle and quality of meat can be realized in this way. In Serbia, production of beef is based on Domestic spotted breed, which is breed of combined production traits. Factors influencing the quality of meat can be divided into pre mortal Dikcman M.E *et al*.2005 (genetic basis, housing, preparation for slaughtering, etc.) and post mortal (slaughtering procedure and cooling of meat) Thompson J. 2002. Research indicates small differences in sensory traits of meat from different genotypes, and authors explanation is that they are mainly caused by the physiological age, since different breeds of the same age don't have the same slaughter maturity (Harrington, 1985, Bucher, 1985).

Breed Composition

Dominant breeds are Domestic Spotted and Simmental cattle, participating by about 70% in total number, then crossbreds of Domestic Spotted and Simmental and Busha cattle participating with about 25%, and about 5% goes to Black and Red-White cattle of European White-Black and Holstein breeds. Breeding of animals of combined production

capacities on small farms in a small number corresponds to various and moderately intensive conditions which are dominant on these farms. Production capacities of animals vary, from weak and moderate to good on smaller number of farms. The cattle quality is improved on farms which use quality bulls and insemination. Production capacities of cattle are not completely utilized, both for milk and meat, because of a number of limitations and weaknesses either in production or in the marketing of final products.

Materials and Methods

In Serbia there are several big companies engaged in production and export of beef. Technology of production is the same in all of them.Buying up of the cattle was organized in the way that the prices per kg of live weight were variable and depended on market supply and demand. The prices of fattened young cattle and carcasses were relatively stable.

Production of fattening calves is organized on farms of their subcontractors. After calving, at the age of 50 – 70 days, the calves are being bought up from subcontractors and placed into fattening facilities. Young cattle are fattened in the free housing system of the capacity of 500 – 1500 animals per cycle. Feeding and health care of fattening young cattle is under the constant supervision of experts of the companies engaged in meat export. For Greek market both male and female animals are used and as regards the way of processing, a standard medial dissection is demanded. Cutting of carcass along spine is a classical, long-standing internationally accepted (Codex Committee on Meat and Meat Products, 1965) method of carcass cutting into two halves. The halves are beheaded and spinal cord removed. As regards quality, the carcass of good conformation and low content of fat tissue is in demand. Especially for Italian market the meat of young female cattle not older than 11,5 months is required as well as the cutting of carcass by so called Milanese cut. The cutting of carcass »baby beef Milanese« is performed in two phases. The first phase is standard cutting of beef carcass along spinal column into two halves. Second phase of cutting is production of a commercial unit of »baby beef Milanese«. For the production of »baby beef Milanese« conventionally obtained halves of beef carcasses are used. A hind part of carcass half is separated along 5. or 6. rib at a right angle onto the spine, then cutting parallel to the spine, belly of the carcass is separated without hind shank. Regarding the meat quality, light red colour, equal marbling, fat tissue of white to light yellow colour and good fat covering of the carcass is required.

Research was carried out on three groups of young bulls of different genotypes (G_1 – Domestic spotted x Charolais, DS x CH; G_2 – Domestic spotted x Limousine, DS x LI and G3 – Domestic spotted breed of Simmental type). Comparative research included 25 heads of young cattle of DS genotype, 18 crosses DS x CH and 21 crosses of DS and LI. Breeding of cattle, slaughtering and primary processing were carried out on experimental farm and slaughterhouse of the Institute for Animal Husbandry. After cooling (+4^{o}C) and within 24 hours, dissection of three rib cut was carried out. Cut of the 9-10-11 ribs was separated from the left cooled carcass sides cutting along the cranial line of the 9th and 11th rib, and with cut parallel to the spine (vertical to the ribs) and 1/3 of the upper rib remained on the cut. Examination of physical-chemical and organoleptic traits of meat was performed on sample of *M.longissimus dorsi* (MLD) from the region of the 10th and 12th vertebrae. Evaluation was performed by three evaluators, and final scores (from 1 to 5) were given after consideration of opinions and reaching of consensus. Sensory analysis was done on samples of *longissimus dorsi*, of size of 2 x 2 x 2 cm subsequent to cooking with boiling water for 10 minutes. System of 5 points was used for scoring: 1-exceptionally

unacceptable to 5-exceptionally acceptable. Following meat attributes were determined/scored: softness, succulence, taste and aroma. Five (5) semi-trained scorers participated in scoring.

The carcass yield was calculated in relation to animal live weight. The yield of Milanese and front quarter was calculated in relation to animal live weight.

Statistical analysis of data obtained in the trial was carried out using the method of Least Squares (MSMLMW – Harvey, LSMLMW and MIXMDL, PC-2version,1990) according to fixed model: $y_{ijk} = \mu+G_1+b_1(x_1-x_2)+e_{ijk}$ where Gi is fixed effect of the genotype and b_1 linear regression effect of the pre-slaughter mass prior.

Results

Table 1 shows the values of studied traits of Simmental male cattle. On the basis of economic parameters presented in the table it can be seen that market value of pre-slaughter live weight is EUR 954,00. After slaughtering market value of carcass processed by standard cut increases by 1,5%. The transport and slaughter costs are covered by the income realized from skin and giblets. Due to small difference (1,5%) the state supports the export by 10% per kg of carcass.

Table 1. Average values and prices per kg for male cattle (n=30)

Male cattle	Average values	Price per kg (EUR)	Total (EUR)
Live weight on 65 day, kg	147.50	2.95	435.13
Live weight on 337 day, kg	530.00	1.80	954.00
Daily gain during fattening, g	1406.00	-	
Warm carcass weight, kg	307.40	3.15	968.31
Carcass yield, %	58.00	-	

In Table 2 we can see that the market value of pre-slaughter female cattle of EUR 773,50 increased after slaughtering by 2,98%. By further processing of the same carcass into “baby beef Milanese”, this value increased by 5,43%. The state supports the export of Milanese by 10%.

Table 2. Average values and prices per kg for female cattle (n=30)

Female cattle	Average value	Price per kg (EUR)	Total (EUR)
Live weight on 65 day, kg	130.50	2.61	340.61
Live weight on 337 day, kg	455.00	1.70	773.50
Daily gain during fattening, g	1171.00	-	
Warm carcass weight, kg	252.80	3.15	796.32
Carcass yield, %	55.47		
Weight of Milanese, kg	96.62	4.40	425.13
Yield of Milanese, %	21.20		
Weight of front quarter, kg	156.16	2.50	390.40
Yield of front quarter, %	34.27		
Milanese +front quarter, kg	252.78		815.53

Due to increasing pressure on the producers to produce the meat from heifers called "baby beef" farmers are forced to fatten quality female heads instead of using them in further reproduction aimed to improve genetic potential of Simmental breed. Because of the reduced supply of fattening calves, as well as lower quality, there is a need for new ways and methods for improvement of the production of beef. Based on obtained research results we can recommend the method of industrial crossing with French fattening breeds which can include 20-25% breeding females of total population of Domestic Spotted breed (Aleksic *et al*., 1997). In the same time, the quality of meat of crossbreds of F1 generation can fully meet all demands of consumers, especially those (Italy and Greece) who are traditionally the importers of our beef (Aleksic *et al*., 2000).

Daily gain and meat yield of carcass sides as the major slaughter traits of the carcass of young bulls, showed statistically highly significant (P<0,01) deviations between genotypes (Table 3).

The effect of genotype on traits of MLD as well as LSM deviations (C_i) is presented in Table 4. Obtained results on effect of genotype on traits of MLD show considerable deviations from the general average in regard to colour and value of pH_{24}MLD. The highest negative deviation for marbling of MLD was obtained in group G_1, and the highest positive deviation was established in trial group G_2. Obtained results of evaluation of the structure of MLD obtained from crossbreds of Domestic spotted and French fattening breeds indicate fineness of the muscle tissue, i.e. tenderness of this meat. Colour of meat from crosses with French breeds is lighter red compared to meat of Domestic spotted cattle. Established differences in meat colour are confirmed by measured pH_{24} values.

Table 3. Influence of genotype on slaughter characteristics of young cattle [#]

TRAITS	μ	SE	G_1	G_2	G_3	F exp.
Daily gain, g	1291.1	9.4	63.9	35.1	-98.9	**
Carcass weig. with fat, kg	397.2	1.9	6.0	9.0	-14.2	*
Killing out, %	59.8	.5	.9	1.8	-3.1	**
Kidney fat, kg	4.9	.3	-.9	.9	.1	NS
Half side meat, kg	139.1	1.9	8.1	6.3	-13.4	**
Half side bones, kg	26.1	.9	-.9	-.7	1.9	*
Half side fat, kg	5.9	.2	-.9	-.6	1.9	**

#The average value (μ), standard error (SE) and deviation from the population average (C1) per genotypes

Table 4. Genotype influence on MLD and pH_{24} value

TRAITS	μ	SE	G_1	G_2	G_3	F exp.
Colour (1-5)	4.5	.1	.3	.4	-.7	*
Marbling (1-5)	4.7	.1	-.2	.2	.1	NS
Structure estimate.(1-5)	4.4	.1	.2	.5	-.8	NS
pH_{24} value	5.9	.0	-.1	-.1	.1	*

[#]The average value (μ), standard error (SE) and deviation from the population average (C_1) per genotypes

The highest negative deviation from the average for tenderness and muscle fibre thickness was established in crosses of Domestic spotted breed with French fattening breeds (Table 5). Results indicate that meat of cattle from this genetic group in their physical traits had the most tender structure. Cross section of MLD was the greatest in cattle of group G_1. Tenderness ("firmness" or texture), as well as succulence ("dryness") of cooked or roasted meat, and to some extent also aroma and taste, are all important parameters of meat quality and they were evaluated by sensory method (Table 6).

Table 5. Genotype influence on physical characteristics of meat [#]

TRAITS	μ	SE	G_1	G_2	G_3	F exp.
Firmness (Volotkiewitsch)	5.8	.1	-.1	-.1	.1	**
Muscle fibre diameter, μm	46.6	.1	-.1.4	-1.8	3.2	**
Total pigment, ppm	86.2	1.2	.5	3.3	-3.8	*
Muscle eye MLD, cm^2	105.4	.7	3.5	-.5	-.1	**

[#]The average value (μ), standard error (SE) and deviation from the population average (C_1) per genotypes

Table 6. Genotype influence on organoleptic traits [#]

TRAITS	μ	SE	G_1	G_2	G_3	F exp.
Cooking test – firmness (1-5)	4.5	.1	.4	.5	-.9	**
Cooking test – dryness (1-5)	3.6	.1	.3	.4	-.7	**
Cooking test – taste (1-5)	4.0	.1	.1	.1	-.1	**
Cooking test – aroma (1-5)	4.0	.1	-.1	.2	-.1	NS
Roasting test – firmness	4.6	.1	.1	.3	-.4	*
Roasting test – dryness	3.5	.1	.1	.1	-.1	*
Roasting test – taste	4.1	.1	-.1	.2	-.1	*
Roasting test – aroma	4.2	.1	-.01	.05	.05	NS

The average value (μ), standard error (SE) and deviation from the population average (C1) per genotypes

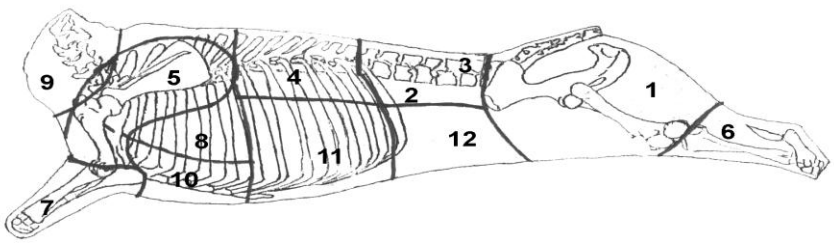

Figure 1. Scheme of cutting of carcass side into main parts
1.- Round/leg; 2.- Tender loin; 3.- Loin part; 4.- Back; 5.- Shoulder; 6.- Second thigh; 7.- Second fore thigh; 8.- Butt; 9.- Neck; 10.- Chest; 11.- Ribs; 12.- Belly

Conclusion

Possibilities for improvement of the quality of beef in population of Domestic spotted cattle breed by application of industrial crossing with French fattening breeds Charolais and Limousine were investigated. It was established that the highest gain was recorded for cattle of genotype G_1. The highest meat yield/dressing percentage of warm carcass sides was recorded in cattle of genotype G1, and at the same time in the same genotype lower percentage of bones in carcass sides and higher share of meat was determined. It was established that young cattle of genotypes 1 and 2, in regard to slaughter traits had advantages in relation to genotype 3. Highly significant effect ($P<0,01$) of genotype on meat yield/dressing percentage in carcass sides was established and lower absolute share of fat and bones in crosses with fattening breeds. Obtained results indicate justification of crossing for the purpose of obtaining of higher meat yield which is in the interest of the producer – farmer.

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The Effect of Pollution on Food Security of Floating Net Cage Aquaculture in The Lake

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Abstract

Food security in fishery is very important to protect public health and to enhance the economic growth. Food security in fishery started from medium for living fish (water quality), seed, fish feed, aquaculture practise until food processing. Floating net cage is the system of aquaculture which usually conducted in the lake/reservoir and river. Therefore the knownledge or characteristics of lake especially water quality lake must be known. There are more than 500 of natural lakes and 25 of reservoirs or man made/artificial lake in Indonesia. Both natural and artificial lake is very important for human being and environment, especially for energy (hydro electric power project) and drinking water. The other function of lake is for irrigation, flood control, tourism, aquatic sport, transportation as well as for floating net cage aquaculture. For example natural lake in Sumatera namely Lake Toba (North Sumatera) and Lake Maninjau (West Sumatera) used for hydro electric power project and fisheries. While artificial lake, actually the main function is for Hydro Electric Power Project, beside that the people used for aquaculture development, particularly floating net cage aquaculture for example Kuto Panjang (Riau), Saguling, Cirata, Jatiluhur in Citarum River Basin (West Java), Mrica (Central Java). However, pressure on the lake have been occurred namely sedimentation and water pollution and eutrophication from industry, agriculture and domestic activities. Along Citarum River Basin, there are more than thousand industry developed especially textile industry which dumped liquid waste into the river and finally enter the reservoir of Saguling, Cirata and Jatiluhur (Djuanda Reservoir), where the fishery activities developed. Several water quality parameters such as Biochemical Oxygen Demand (BOD), Chemical Oxygen Demand, H2S, dan Pb have increased while dissolved oxygen decreased. Several time "overturn" occurred in the reservoir, which caused massive killed of fish because of high concentration of chemical toxic substance like H2S and NH3. In addition, concentration of heavy metal such as Pb detected in fish meat of fish aquaculture and wild fish in Jatiluhur reservoir. In relation to food security this condition must be aware (precautionary principle/approach). In Cirata reservoir Koi Herpes Virus, because of bad sanitation and water quality. While in Maninjau, in the last two years the number of floating net cages reaches until 15,051 units, and the using of fish food increases. However in this period overturn occurred two times caused 14,000 ton and 500 ton of fish killed. The government of Republic of Indonesia already have law, regulation and guidelines concerning of Food, Food Security and Guideline of Conduct of Aquaculture Practices. While internationally FAO and WHO have published A Guide for national food safety authorities.

Keywords: ***floating net cages, food safety , heavy metal, natural and artificial lake, overturn***

Introduction

Food security in fishery is very important to protect the public health and to enhance the economic growth. Food security in fishery must be started from medium for living fish (water quality), seed, fish feed, aquaculture practise until food processing. Floating net cage is the system of aquaculture which usually conducted in the lake/reservoir and river. Therefore the knownledge or characteristics of lake especially water quality lake must be known.

Food security cosist of pathogen, toxic chemical in food like formaline, borax etc; and low sanitation and hygiene aspect of product and food processing. Pollutant or contaminant which often found in fishery product consist of biological pollutant and chemical pollutant. Biological pollutant namely vibrio parahaemolyticus, vibrio cholerae, salmonella and E. Coli. While chemical pollutant consist of heavy metal (Hg and Pb), histamine, marine biotoxin, hormon, antibiotics and pestiside as well as toxic material like formaline and rhodamine B (Anisya, 2011).

Fish processing technology is very important in food security to ensure the fish product safely and health. Therefore the method for fish processing refer to Good Manufacturing Practice (GMP), Standard Sanitation Operating Procedure (SSOP), as well as conduct Hazard Analysis Critical Control Point (HACCP) (Dirjen Fish Processing and Marketing of Fishery Product, 2007).

The Indonesian Government have paid attention on food security since published the Law of Food No. 7 Year 2004 and the Law of Fishery 2004, and other regulation and guidelines which publish by Ministry of Health and Ministry of Marine Affairs and Fishery. Regulation published by Ministry of Health or Board of Medicine and Food Supervision is Guidelines on Determine Good System of Food Production. While Ministry of Marine Affairs and Fishery published Guidelines on Good Aquaculture Practises. To supervise and prevent pollution on food, the government determined the maximum allowable pollutant in food, especially for metal and microbe.

As mentioned before that the media (water quality) and location of aquaculture conducted must be known. Therefore lake and reservoir in Indonesia where floating net cage aquaculture characteristic presented in this paper.

The lake consisted of two types, namely natural and artificial lake or manmade lake (reservoir). In Indonesia there are more than 500 of natural lake and 25 of artificial lake (about 5 artificial lake > 5,000 ha). In five ASEAN countries is estimated circa 1,540,000 ha of which one-third is reservoir area. Natural lakes are relatively important in Indonesia and the Philippines which have respectively 9 and 6 lakes of over 5,000 ha (Verdegem, 1999; Giessen, 1994). According to Verdegem (1999), in the five ASEAN countries, there were 55 reservoir of over 1000 ha, 27 of over 5,000 ha and only 14 of over 10,000 ha. The total surface area of reservoirs in Thailand and Malaysia increased almost linearly since the early sixties. The constructions of large reservoirs were built especially for power generation.

In Indonesia the biggest natural lake is Lake Toba, 1102,970 ha of surface area, while the reservoir is Koto Panjang,12,400 ha of surface area; both of the lakes are in Sumatera. The main function of natural lake is for conservation, however at present lake use for genetic resources, tourism, fisheries (aquaculture and fish capture), sport, transportation, sources of drinking water materials, and hydropower generation. While the main function of

reservoir is for hydropower generation, and also use for aquaculture, tourism, sport, transportation, flooding control, irrigation, sources of drinking water material, and industry.

The lake distributed in the Indonesia Archipelago mainly in Sumatera, Kalimantan, Java and Bali, Sulawesi, Maluku and Papua. The largest number lakes are located in Sumatera, over 170 lakes and surface water area circa 190,043 ha; including Lake Toba. Lake Toba is the biggest lake Indonesia, even the South Asian Countries. In Kalimantan there are 139 lakes and surface water area circa 84,231 ha. While in Sulawesi, there are 30 lakes, the second large surface area circa 141,871 ha. The numbers of lakes in Papua are 127 lakes and surface water area circa 59,830 ha.

The problem on lake ecosystem consisted of aquatic and terrestrial ecosystem in river basin and catchment area of the lake. The problem in terrestrial ecosystem is because of trees/forest cutting in upper area caused the erosion and sedimentation so that the lake becomes shallow. The other thing is settlement, industrial and agriculture waste as well as mining waste caused the aquatic pollution of the lake. The potential problem also occurred in the lake itself namely overfishing, massive aquaculture that potential to have over feeding. The waste of aquaculture and fish secretion potent the water pollution especially organic pollution caused eutrophication/nutrient enrichment and blooming of algae as well as massive of aquatic weeds growth especially water hyacinth. Algal bloom especialy of Microcystis which living in three reserviir along Citarum River produce biotoxin called microcystine. In last five year, in Cirata massive Koi Herpes Virus, because of bad water quality and sanitation as good media for growing KHV, caused massive killed of common carp. The most important is water pollution which caused by industrial activities particularly toxic chemical like heavy metal (Hg, Cd, Pb, Cr) and pesticide.

Natural lake

Lake Toba is located in Province of North Sumatera, about 176 km southern of City of Medan. The catchment area of this lake is 3,440 km^2, surface water area 1102,60 ha, and depth 435 m. The area of Lake Toba is national asset and has strategic value for North Sumatera Province. The lake has multifunction namely for drinking water material, tourism, agriculture, irrigation, fishery, transportation, and hydropower generation about 450 MW, and potent to develop more electric hydropower until > 1,000 MW. However, some potential problem of pollution will treat the Lake Toba such as domestic pollution of disposal, feces; pollution from hotel and restaurant, transportation waste like oil, animal dung, aquaculture particularly floating net cages (FNC) like feed fish waste, massive growth of water hyacinth. At present, there are about 1,347 units of FNC that belong to foreign company and 5,300 units FNC of local people. The other problem is agriculture waste like pesticide and fertilizer, and erosion as well as sedimentation.

The other lake in West Sumatera is Lake Maninjau. Lake Maninjau is volcanic lake, like Singkarak Lake. The area of surface lake is 9,950 ha, and depth maximum is 480 m. The function of this lake is for tourism, irrigation, genetic resources, source of drinking water, reservoir, hydropower generation (68 MW), fishery (aquaculture, there is about 16,000 of FNCs). At present the main issues of this lake is massive fish killed because of overturn. Last year (December 2009) about 14,000 ton fish killed namely common carp (*Cyprinus carpio*) and Nile Tilapia (*Tilapia nilotica*). It was the biggest disaster in the Lake Maninjau, especially for the farmer and about 150 billion rupiah lost. While this year (March 2010), about 500 ton fish killed and about 4.8 billion rupiah lost. Actually the floating net cages, year by year increase. In 1997, the number of FNC only 2,854, 2005 circa 8,251 units, 2008 circa 15,051

units and 2010 about 16,000 units. This activity has a large advantage to the farmers. However, the impact of FNC development is very big, caused by organic pollution, the increasing of NO_3, PO_4, H_2S and NH_3. The high concentration of NO_3 and PO_4, were caused by eutrophication of the lake, while high concentration of H_2S and NH_3 is poison for fish.

Reservoir (man made lake)

Reservoirs that will be described in this paper are three artificial lakes in along Citarum River in West Java. This river is the longest river in West Java (325 km long) and consisted of three large reservoirs, and the main function of two reservoirs is for hydropower generation, while one reservoir is multipurpose reservoir. The first two reservoirs, namely Saguling and Cirata, and the third is Jatiluhur which the largest reservoir in this area as well as the first large reservoir in Indonesia. The surface area of three reservoirs are 5,600 ha, 6,200 ha and 8,000 ha respectively. The capacity of hydropower generation is 700 megawatt, 1,500 megawatt, and 150 megawatt respectively. The electricity which resulted from those reservoirs is for Java and Bali requirement.

The three lakes already had an environmental pressured due to the high human population surrounding the lake cause high domestic pollution, high sedimentation of Citarum River, industrial pollution especially textile industries, more than thousand of industry in diferent type in this area. In the lakes it selves, aquaculture especially floating net cages developed excessively carrying capacity, eutrophication or nutrient enrichment already occurred, overturn which caused fish killed every year occurred.

Domestic pollution will caused organic pollution in the lake especially from household waste, bad sanitation system and bad habit of the people to dump debris to the river. This condition will be increased of Nitrogen and Phosphorous in aquatic ecosystem, especially NO_3, NO_2, NH_3, and PO_4, is good media for bacteria, virus to grow, for example KHV growing in Cirata Reservoir. While contain of industrial pollution usually are organic compound, and heavy metal especially Cr, Cd and Pb. Heavy metal pollution is very dangerous because of possibility of bioaccumulation through food chain and food web as well as decrease the food safety and food security.

Fisheries activities particularly floating net cages is intensive aquaculture need fish food with high protein content. Based on investigation about 30 % of food lost to the aquatic ecosystem, therefore NO_3, NH_3 and PO_4 increased. Besides that the content of BOD and COD also increased. In the table below presented the water quality characteristic in the three reservoirs. In the three table below presented the water quality characteristics of three reservoir along Citarum River.

Table 1 showed that generally the water quality characteristics of Saguling Reservoir was moderate for utilization of drinking water materials and fisheries, but for hydropower generation is still good. In each station the content of organic material is shown by COD, BOD higher compare with water quality standard particularly for drinking water material. The content of COD and BOD higher compare to water quality standard due to feed waste and domestic waste in the water was high; in contras concentration of DO (dissolved oxygen) was low, because oxygen was used by microorganism to degrade organic matter to be inorganic matter such as NO_3, NO_2, NH_3 and PO_4. The degradation of organic matter to H_2S is conducted by anaerobic bacteria.

Table 1. Water quality characteristic in the Saguling Reservoir for utilization of drinking water material (Class 2), fisheries (Class 3), and hydropower generation (Class 4), 2007.

Stat. No.	Class 2			Class 3			Class 4		
	Unmatched standard Parameter	Score Total	Standard Status	Unmatched standard Parameter	Score Total	Standard Status	Unmatched standard Parameter	Score Total	Standard Statue
1	DO, COD, BOD	-22	Moderate	NO_2, Cl_2, Zn	-20	Moderate	RSC	-10	Good
2	DO, COD, BOD, Cd	-32	Bad	H_2S, NH_3, NO_2, Cl_2, DO, Zn, Cd	-48	Bad	Cd	-2	Good
3	H_2S, DO, COD, BOD	-32	Bad	H_2S, NH_3, NO_2, DO, Zn	-36	Bad	RSC	-8	Good
4	DO, COD, BOD, Cd	-30	Moderate	NO_2, Cl_2, DO, Cd	-20	Moderate	Cd, RSC	-12	Moderate
5	H_2S, DO, COD, BOD	-36	Bad	H_2S, NH_3, NO_2, Cl_2, DO	-34	Bad	RSC	-8	Good
6	DO, COD, BOD, Cd	-30	Moderate	H_2S, NH_3, Cl_2, DO, Cd	-24	Moderate	Cd, RSC	-12	Moderate
7	H_2S, DO, COD, BOD	-30	Moderate	H_2S, NH_3, Cl_2, DO	-26	Moderate	RSC	-10	Good
8	H_2S, DO, COD, BOD	-30	Moderate	H_2S, NH_3, NO_2, Cl_2, DO, Zn	-36	Bad	RSC	-10	Good
9	DO, COD, BOD	-28	Moderate	H_2S, NH_3, NO_2, DO, Zn	-30	Moderate	RSC	-8	Good
	Score Average	-30	Moderate		-30	Moderate		-9	Good

In water sample was found some heavy metal such as cadmium (Cd). The sources of heavy metal is industrial waste especially from textile industry which in the Citarum River Basin or catchment area of Saguling Reservoir. The content of industrial waste was not only of heavy metal, but also organic compound.

The water qualities in Cirata Reservoir are bad especially for drinking water materials and fisheries, but are still very good for hydropower generation. Standard status of water quality in Cirata Reservoir particularly for drinking water material and fisheries is worst compare to Saguling Reservoir. The reason is the aquaculture activities more developed in this reservoir. There are circa 51,000 units of floating net cages in this reservoir whereas in Saguling only circa 25,000 unit. So, there are more fish feed required and more waste dump on the reservoir. Besides that the people who manage the floating net cages lived in here more than in Saguling. The daily activities of them conducted in the floating net. They dump domestic waste to the reservoir; E. coli and coli form are higher unmatched compare to water quality standard. This biological parameter increased due to the feces and dung. Actually, the water quality of Cirata Reservoir is bad for aquaculture utilization; however the cultivator still develop floating net cages for fish aquaculture. Therefore overturn often occurred in this reservoir, the thousand tones fish killed, billion rupiah lost. Another disadvantaged of water quality degradation is fish diseases. Invertebrate and bacteria which caused fish diseases (KHV) are growing in this condition very well.

As mentioned above that 30% of fish feed lost to the aquatic ecosystem, this condition caused sedimentation, the reservoir became shallow and very rich of organic material, decomposed occurred, the benthic animal can't live in. Although benthic animal is one of

natural fish feed besides plankton. This condition is organic pollution, the diversity of benthic animal and plankton decrease, water quality parameter such as BOD and COD increased but DO (dissolved oxygen) decreased, although oxygen is the most important for aquatic organism like fish. Table 2 is shown this condition.

Table 2. Water quality characteristic in the Cirata Reservoir for utilization of drinking water material (Class 2), fisheries (Class 3), and hydropower generation (Class 4), 2007.

Stat. No.	Class 2			Class 3			Class 4		
	Unmatched standard Parameter	Score Total	Standard Status	Unmatched standard Parameter	Score Total	Standard Status	Unmatched standard Parameter	Score Total	Standard Statue
1	H_2S, DO, COD, BOD, Hg, Se	-46	Bad	H_2S, NH_3, NO_2, Cl_2, DO, Hg	-42	Bad	-	0	Very Good
2	H_2S, DO, COD, BOD, Hg, Se, E. coli, Coli form	-80	Bad	H_2S, NO_2, Cl_2, DO, Hg	-40	Bad	-	0	Very Good
3	DO, COD, BOD, Hg, E. coli, Coli form	-68	Bad	H_2S, NO_2, Cl_2, DO, Hg	-38	Bad	-	0	Very Good
4	DO, COD, BOD, Hg, E. coli, Coli form	-70	Bad	H_2S, NO_2, Cl_2, DO, Hg	-30	Moderate	-	0	Very Good
5	DO, COD, BOD, Hg, E. coli	-47	Bad	H_2S, Cl_2, DO, Zn	-30	Moderate	-	0	Very Good
	Score Average	-62	Bad		-35	Bad	-	0	Very Good

The table above showed that water quality in Jatiluhur Reservoir relatively better compare to Saguling and Cirata Reservoir. It is because of the location of Jatilhur Reservoir in the lower part of Citarum River. Saguling and Cirata as filter for Jatiluhur, pollutant and mud from upper part were deposited in two reservoirs. However, organic material (BOD and COD) is high, unmatched compare to water quality standard for drinking water utilization, in contrast DO is low, unmatched also for drinking water utilization. The concentration of heavy metal Pb is higher for all utilization. The source of heavy metal is probably from industrial waste which a lot surrounding reservoir. The number of floating net cages in this reservoir is smaller compare to Saguling and Cirata. In Jatiluhur, there are about 15,000 units of FNC, and the FNC distribution only in the one site, because the local government gives permission to the farmer on this site. In general aquaculture activities in Jatiluhur reservoir is more organize than in Saguling and Cirata.

In the Table 4 presented the concentration of heavy metal in fish meat, fish feed, and sediment in Jatiluhur and Cirata Reservoir. This data compiled from our research in other research conducted by Institute of Ecology, which cooperation with Board of Cirata Reservoir Management.

Table 3. Water quality characteristic in the Jatiluhur Reservoir for utilization of drinking water material (Class 2), fisheries (Class 3), and hydropower generation (Class 4), 2007.

Stat. No.	Class 2			Class 3			Class 4		
	Unmatched standard Parameter	Score Total	Standard Status	Unmatched standard Parameter	Score Total	Standard Status	Unmatched standard Parameter	Score Total	Standard Statue
1	BOD, COD, DO, Pb	-32	Bad	H_2S, Pb	-20	Moderate	Pb	-2	Good
2	BOD, COD, DO	-30	Moderate	H_2S	-10	Good	-	0	Very good
3	-	0	Very good	H_2S	-10		-	0	Very good
4	BOD, COD, DO	-30	Moderate	H_2S	-10	Good	-	0	Very good
5	BOD, COD, DO	-30	Moderate	H_2S, Pb	-20	Moderate	Pb	-2	Good
6	BOD, COD	-10	Moderate	H_2S, Pb	-20	Moderate	Pb	-2	Good
	Score Average	-24	Moderate		-15	Moderate		-1	Good

Note: Water quality standard based on Regulation of Indonesian Government No. 82/2001, concerning Management of Water Quality and Water Pollution Control. Status Standard based on Storet Method (USEPA, Canter, 1997). Score 0: very good; (-1)-(-10): good; (-11)-(-30): moderate; > (-31)

Table 4 . Concentration of heavy metal in fish aquaculture and wild fish

Parameter	Concentration (mg/kg)				
	JTL-I01	JTL-I02	JTL-I03	JTL-I04	JTL-I05
Cd	-	-	-	-	-
Cr	-	-	-	-	-
Hg	0.016	0.010	0.023	0.014	0.019
Se	-	-	-	-	-
Pb	13.2	-	-	4.1	1.8

Note: (-) = undetected, value under detection limit, 0,002 mg/kg; JTL-I01= Pangasius (culture); JTL-I02= Common carp (culture); JTL-I03= Oscar (wild); JTL-I04=Tilapia (wild); JTL-I05= Tilapia (culture)

As mentioned before that Pb concentration in the water is high, and in Table 4 (the first analysis/measurement) showed that the concentration of Pb in fish meat also higher that other heavy metal (Hg). So, there is relation between Pb concentration in water and Pb accumulation in fish meat.

Table 5. Concentration of heavy metal in fish aquaculture and wild fish

Parameter	Concentration (mg/kg)					
	JTL-01	JTL-02	JTL-03	JTL-04	JTL-05	JTL-06
Se	-	0.005	0.005	-	-	-
Cd	-	-	-	-	-	-
Hg	-	-	0.005	0.015	0.055	0.005
As	-	0.020	-	-	-	-
Cu	-	-	-	-	-	-
Pb	1.610	0.385	1.925	1.460	0.365	2.335
Cr	-	-	-	-	-	-

Note: (-) = Undetected, value under detection limit 0,002 mg/L

Like in Table 4, Table 5 (the second analysis/measurement) showed that Pb was higher than others heavy metal of Se, Hg, and As. So, from this analysis of heavy metal can be concluded that Pb found in all species fish sample namely common carp, pangasius, tilapia and Oscar, in fish culture and wild fish. Compare to standard which published by FAO (1983) and Ministry of Health Republic of Indonesia (Board of Food and Medicine Supervision) (1989), concentration of Pb have been exceeded of standard 0.5 ppm.

Table 6. Concentration of heavy metal in fish meat of Nilem

Treatments	Heavy Metal		
	Pb (ppm)	Cd (ppm)	Hg (ppm)
No fish feed	0.100	0.011	0.0017
1% fish feed	0.100	0.252	0.001125
2% fish feed	0.120	0.067	0.000925
3% fish feed	0.100	0.128	0.00325
4% fish feed	0.100	0.118	0.0011375
5% fish feed	0.100	0.151	0.0007

Table 6 showed that in all treatments heavy metal of Pb, Cd and Hg was found in meat of fish, however all concentration still below the FAO Standard (1983) and BPOM R.I. (1989) that is 0.5 ppm. This experiment was conducted during 71 days and fish in seed phase (8-12 gram). While the condition fish presented in Table 4 and 5 was fish consumption, so the size was bigger (200 gram).

Research of heavy metal concentration in culture fish and wild fish also condusted by Institute of Ecology in cooperation with Lake Cirata Management Board (2009). The result showed that several heavy metal were found in the meat of fish (tilapia, common carp and bawal) namely Cd, Mn, Zn, Ni, Cu, Cr, As and Hg, however the concentration still low compare to standard of FAO (1985) and WHO (2003).

The concentration of heavy metal content in fish feed was varied, however heavy metal of Pb was found that was 0.155-0.25 mg/kg, while concentration of Zn and Cu was the highest, because both metal was trace element which required in the fish feed. In addition, research conducted by Institute of Ecology (2009) on concentration of heavy metal ini fish feed showed that nine heavy metal were found in the fish feed namely Cd, Mn, Zn, Ni, Cu, Cr, As and Hg.

Table 7. Concentration of heavy metal in fish feed sample

Parameter	Unit	Concentration		
		JTL-P01	JTL-P02	JTL-P03
Ash	%	18.50	15.17	21.40
N-total	%	1.66	3.74	5.09
P-total	%	0.007	0.009	0.014
Se	mg/kg	0.015	0.06	0.08
Cd	mg/kg	-	-	-
Hg	mg/kg	-	-	0,02
As	mg/kg	0.025	0.03	0.03
Zn	mg/kg	1.005	0.51	0.335
Cu	mg/kg	0.75	1.20	0.89
Pb	mg/kg	0.25	0.17	0.155
Cr	mg/kg	-	-	-

Note: (-) = undetected, value under detection limit, 0.002 mg/L

Table 8 showed that metal was found in the sediment, and Pb one of the highest concentration beside Zn. So from Table 4-8, concentration of Pb relatively higher than other heavy metal, however Pb is toxic metal. Based on research conducted by Institute of Ecology showed that nine heavy metal were found in the sediment namely Pb, Cd, Mn, Zn, Ni, Cu, Cr, As, and Hg. The heavy metal concentration in the sediment of Lake Cirata lower compare to EPA Regional V Standard (Ahmad *et al*, 2009).

Pb was found high in water, fish meat, fish feed and sediment. Based on standard of FAO (1985) and WHO (2006), the allowable concentration of both metal in fish and food was 0.5 mg/kg and 2 mg/kg respectively.

Table 8. Concentration of heavy metal in sediment sample

Parameter	Unit	Concentration	
		JTL-P01	JTL-P02
N-total	%	0.199	0.172
P-total	%	0.005	0.006
Se	mg/kg	0.095	0.065
Cd	mg/kg	-	-
Hg	mg/kg	0.065	0.095
As	mg/kg	0.075	0.055
Zn	mg/kg	39.58	44.39
Cu	mg/kg	4.81	5.90
Pb	mg/kg	10.03	10.81
Cr	mg/kg	0.21	0.36

Note: (-) = undetected, value under detection limit, 0,002 mg/L

Environmental management

The environmental management can be conducted outside the lake in the catchment area and in lake side it's self. The catchment area environmental management activities are tree planting to prevent erosion and sedimentation, by building small reservoirs or wet land area to trap sediment, disposal, and aquatic weeds before entering the reservoir. The local government of West Java Province plan to build several wet land area in the upstream of Citarum River to protect Saguling, Cirata and Jatiluhur Reservoir. The other thing is the industrial have to treat the waste before dumping to the river as well as prevent the people to dump disposal to the river. It is bad habit the people who live surrounding or in the flood plain area dump the disposal or domestic waste to the river. It is new paradigm that to prevent industrial pollution is not only to treatment the waste (*end of pipe*) but also to reuse, reduced and recycle the waste (*cleaner production*).

The other thing fish feed waste limited, aquatic pollution of organic material or eutrophication can be controlled. The most important thing is the frequency of overturn is seldom. The overturn is natural accident in the reservoir and lake that caused the massive killed of fish. To know earlier of overturn the early warning system has been utilized in Maninjau Lake. This system is to know the concentration of dissolved oxygen and temperature. One parameter or key parameter of aquaculture is dissolve oxygen. Fish require DO > 3 ppm for their living, especially for aquaculture > 5 ppm DO is required. The eco technology can be applied, to increased dissolved oxygen, input oxygen by using aerator in the water body.

The other environmental management of the lake that occurred fish killed is to identified the zone of fisheries, the FNC in a certain area, environmental friendly of aquaculture development, for example use double net. In this system the feed waste was trapped by second net and using by another species of fish namely tilapia, therefore the waste can be minimize. The time of overturn must be known by the farmer, so in the certain time the volume of fish can be reduced. The overturn usually occurred in the month of December-March. Utilization of fish killed for feed fish material, because the fish has high content of protein, and the price of fish mill is very expensive.

Conclusion

In relation to food security, the industrial pollution which content of heavy metal, so waste water treatment must be applied in industrial activities. Therefore, the pollutant, particularly toxic material like heavy metal will be reduced in the lake and reservoir. The most important thing is occurred the conflict of interest among department. For example the Ministry of Marine Affairs and Fisheries plan to enhance fish production through aquaculture in the sea and freshwater, also Department of Industry and Trade want to increase the production of textile to have more deviza, but the environment and organism living there in bad quality and disturbed, finally food security or food safety will decrease.

In addition, the aquaculture good practise must be applied in floating net cage aquaculture in the lake ecosystem. So that, the fish feed will be reduced, the quality of sanitation, hygiene, fish health and aquatic environment increase. In relation to law and regulation on food security must be applied, and law enforcement also must be enhanced.

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Characteristics of Indonesian Lakes and Fisheries Development

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Background

Natural watershed depression lakes originate and were generally formed during the ice ages by tectonic or volcanic activities. In contrast, river-bottom lakes like floodplains, flooded forests and the man-induced development activities create the ever-increasing number of reservoirs and artificial water impoundments. The relation of lakes to the other terrestrial and aquatic water systems is illustrated in Figure 1.

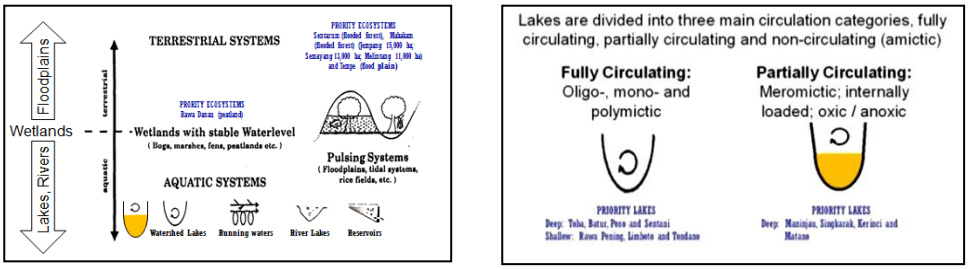


Figure 1. Terrestrial and aquatic water systems (left) and two types of deep lakes, fully circulating and partially circulating internally loaded meromictic lakes (right)

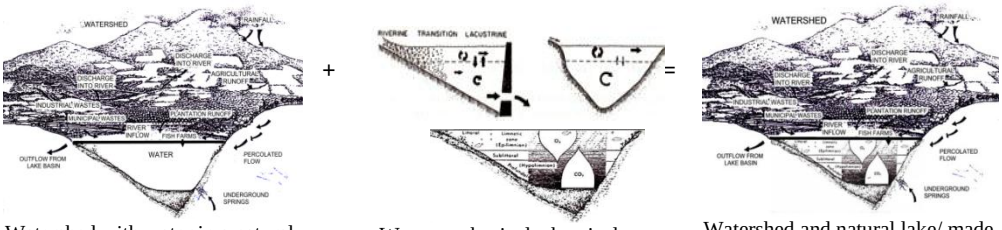
Forbes already in 1887 understood that “watershed” lakes are ancient isolated equilibriums and independent of the surrounding land and “river-bottom” lakes are appendages of rivers.

Some 100 years ago it was recognized that the temperature-dependent hydrodynamic and biogeochemical functions are driven by seasonal temperature changes and in the peculiarly behaving tropical lakes they are mostly irregularly wind-driven.

High temperature and the thermal barrier (metalimnion) between the upper water layer and deep water are important in structuring and distribution of substances and interrelationships and interaction of the ecosystem and bioactivity.

Watersheds of the river-dependent reservoirs are much larger in relation to the water surface area of the natural lakes. Because reservoirs are formed almost always in river valleys, the basins are usually narrow and elongated reservoir continuums from the riverine to transitional and to lake-like lacustrine conditions nearest the dam.

Whether naturally or artificially created, lakes share several common physical, chemical and biological characteristics, and the lake equilibriums alter, age and gradually dry out during the years. Lately land-use and agriculture related runoff, communal wastewaters and industrial effluents have taken a dominant polluter role with increasing consequences, such as siltation, eutrophication and bioaccumulation of harmful substances.



Watershed with water in a natural lake or made reservoir

Water as physical, chemical and biological intermediary

Watershed and natural lake/ made reservoir as an ecosystem

Figure 2. Building blocks, watershed and water (left) and the hydrodynamically and biogeochemically mediating water (center) and the entire holistic lake ecosystem (right)

For lakes nutrient-rich *external loading* from scattered and point sources is the main reason of eutrophication, causing algal blooms, reduced oxygen reserves and fish kills. Low oxygen levels or hypoxia affect fish production and oxygen controls release of phosphorus from the bottom sediment. In water, like in air, oxygen saturation must be at least 70-80% for the healthy higher life but continued anoxia in the near bottom water causes *internal loading.* The rapidly increasing *on-lake pressures* of the cage/ pen fish farms directly pollute the lakes. In some areas *airborne-precipitation* and *acid rain* may also need attention.

Objectives

Limnology, "oceanography" of inland waters, the principal fresh and brackish water science *is one of the three ecological divisions on earth*. The management and protection and fisheries-oriented applied limnology are multidisciplinary, interdisciplinary and sometimes transdisciplinary ventures that meld the often descriptive building blocks of the natural sciences (see Figure 3 and 4) with the technical, economic and social issues (Figure 3).

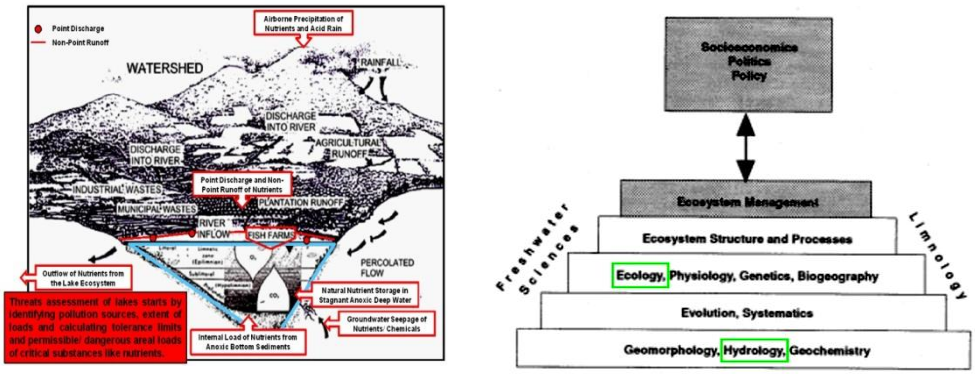
Limnology is the causal-synthetic analytical science which builds on the holistic and integrated management of the water bodies; i.e. interrelationships with watershed, water and the living resources alike, in which air above, landscape around, impoundment of water and water as the mediation ground for biogeochemical building blocks of the ecosystem are concurrently considered as management objects (see Figure 2). The self-purification capacity as a collective expression must be abolished because in nature such a capacity rarely exists. The lake ecosystems are naturally developed "equilibriums which steadily self-adjust to accomplish the greatest good which the circumstances will permit for all parties" (Forbes, 1887) and which must be properly conceptually understood (Figures 3, 4 and 6).

The *best available technology* mayfavor too efficient waste water treatment and the *best environmental practices* highlight the environmental issues but their goals may contradict when the controlling "excess" nitrogen load is removed. This may happen also in the "*heavily protected"* lakes, where nitrogen pollution is needed to keep the N/ P-ratio high.

Although in the world there is a large number of guidelines and frameworks to manage the temperate lakes sustainably, Indonesia needs local climate and lake type and characteristic specific guidelines since they range from lowland tropical ones to high altitude alpine lakes.

Methodology

In the systemic lake management, first a *water balance calculation* is made and *pressures* from the air, watershed, on-lake and deep water and bottom sediment, the long-term log book of the lake, are assessed and quantified (Figure 3, left), after which the *ecological situation* is judged by comparing the physical, chemical and biological characteristics of the water body to the reference conditions (see Figure 4). Based on the assessments, and conceptual frameworks of the *critical pathways* is made and the *areal loading* is calculated.



Courtesy of NOOA, Freshwater Imperative, USA, 1995

Figure 3. Assessment of pressures from the watershed (left) and collection of information from the various building blocks of the limnological assessment and preparing a management plan or, if necessary, restoration program for the lake ecosystem management (right)

If there are great changes in the characteristics of the ecosystem and the areal load exceeds the acceptable limits, a cause, consequence, correction, cost-benefit and concluding synthesis is made to improve or restore the ecosystem (see Figure 3, right).

Results

By studying the problem identification of the 15 various types of priority lakes selected by the Ministry of Environment for the national lake program to implement the Bali Declaration (2009), it became evident that problem identification needs improvements (Figure 1).

The Indonesian lakes hold some 500 km^3 of water (world 175,000 km^3) and reservoirs about 90 km^3 (world 7,000 km^3); in the rest of East/ Southeast Asia respectively 800 and 1,980 km^3. They hold 72% of fresh water (Zainuddin Amilia, 2010) and may foil the water crisis (Rachmat Witoelar, 2009) and their environmental and socioeconomic value is high.

However, most of the required basic information is limited and fragmented into various institutions or such data and information are not at all available and/ or are longitudinally and depth wise seldom adequate, making assessments and management planning fraught with dangers. Despite the excellent start of the tropical limnology in Indonesia some 90 years ago by the Sunda Expedition and continued 20 years ago by the Expedition

Indodanau, the Indonesian national limnology is largely at the descriptive stages. To advance the evidence-based management of lakes, is necessary to expand the limnological focus also longitudinally and depth wise on physical, chemical and productivity/ bioactivity studies (Figure 4).

To speed-up switch from the descriptive to analytical, causal-synthetic studies, first it is necessary to understand the basic latitudinal differences between temperate and tropical lakes. As examples the relative metabolic rate, capacity of water to hold oxygen and the rate at which zero oxygen (anoxia) is reached are used (Figure 5).

These alone stress that water as media should be the first study object but most often, however, it is the last, because the diversity of aquatic life and forms of life and species attract more. Water has also at high temperatures the highest heat absorption capacity and lakes can also modify the local microclimate.

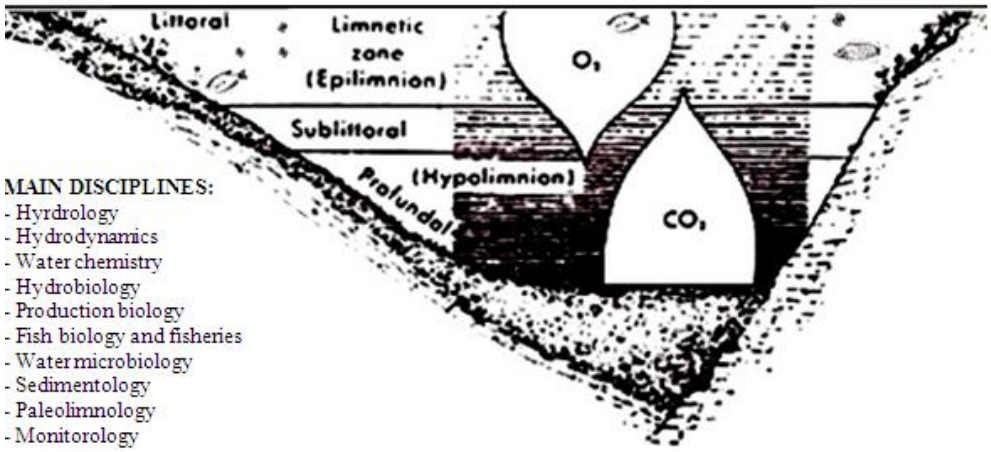


Figure 4. Lake, a water impoundment, and the net results of physical effects of light and wind (heat and thermal structure), biogeochemical metabolism (nutrients, oxygen, carbon dioxide, hydrogen sulfide, methane, etc.) and biological succession (algae, zooplankton, fish, etc.).

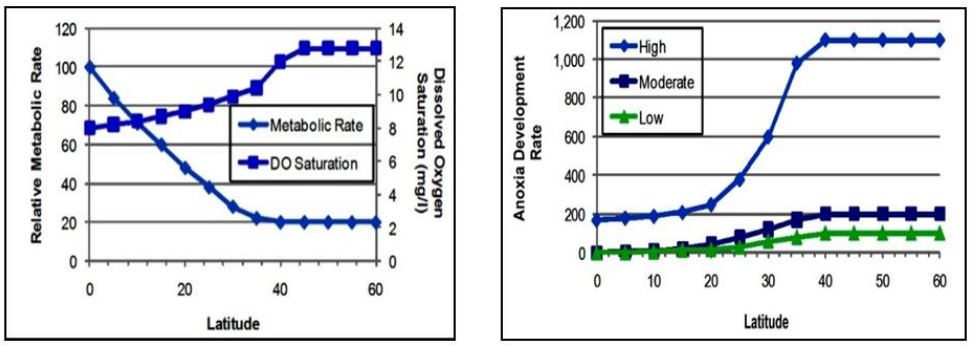
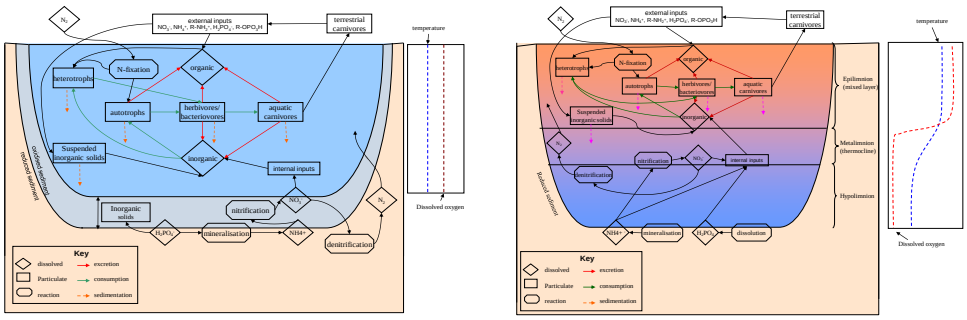


Figure 5. Latitudinal differences in metabolic rate, oxygen saturation (left) and anoxia development rate (right). In Indonesia, anoxia is not a direct indicator of eutrophication.

Biogeochemical functions and food webs

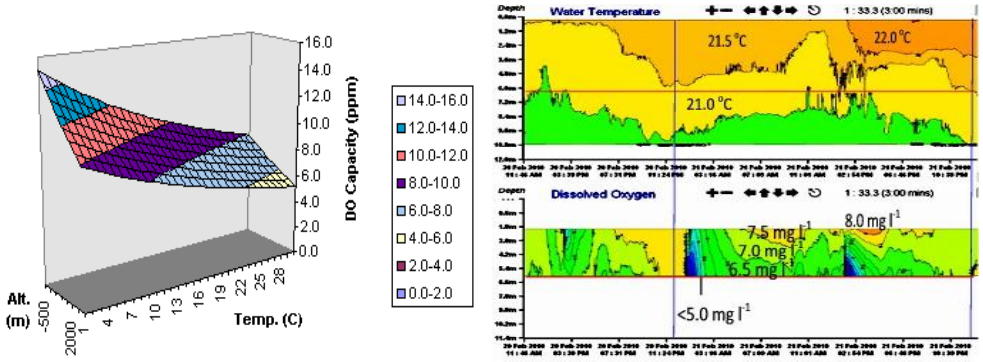
For basic biogeochemical functions, conceptual models have been developed to improve the understanding the key physical, chemical and biological processes occurring in lakes under non-stratified mixing conditions, where oxygen reserves are replenished (Figure 6, left) and in stratified conditions where oxygen reserves are consumed at various rates, (Figure 6, right). This happens in zero latitudes 4-10 times faster than in the temperate regions.



Courtesy of EU WFD, 2001

Figure 6. Light, temperature and wind form thermal stratification, drive turnover and mixing and biogeochemical metabolism govern oxygen conditions and the water ecosystem.

In temperate lakes mixing is cyclic while in tropical lakes turnovers/ mixing are irregular and often wind induced. Mixing of water cannot be overemphasized; it is vital to oxygen replenishment and for higher animal life like fish. Figure 7 (left) shows oxygen solubility and (right) how swift the changes in temperature and oxygen levels can be in 36 hours.



Courtesy of CWR-UWA, Australia, 2010

Figure 7. Oxygen solubility in various temperatures and altitudes (left) and the rapid diurnal variation of temperature and oxygen concentrations in water (right, vertical lines show midnight).

Internal waves

An internal wave (seiche) or sloping of the thermal barrier (thermocline), first recorded in 1519 but widely unknown in Indonesia, is generated by a heavy wind/ rainfall on one end of a lake/ reservoir and oscillations are damped by frictional resistance of water (Figure 8).

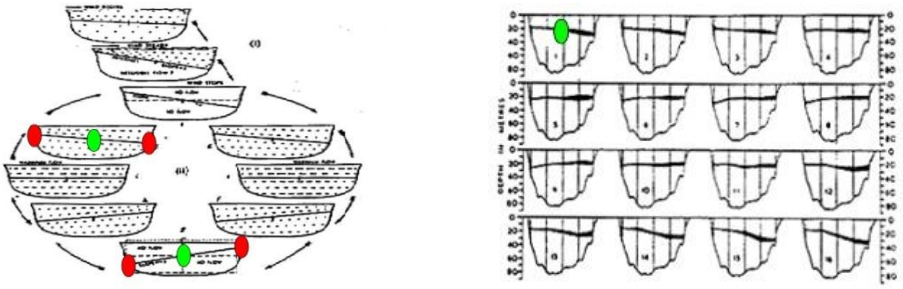
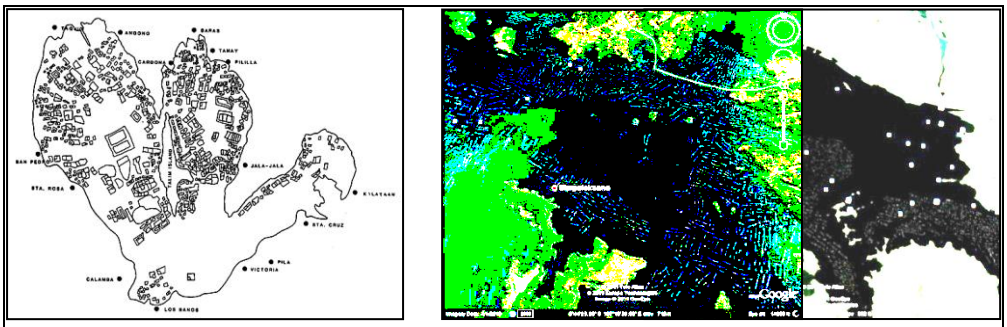


Figure 8. The schematic internal wave (left) and recorded wave in Lake Earn, Scotland (right). The black layer in the lake is the oscillating thermocline, the green dot the node of the wave and the red dot shows the shallow areas where fish farms are in greatest danger

In the uninodal seiche, the thermocline slope and vertical water displacement is greatest at windward and leeward ends, least at the node. These waves may also create turbulence in the near bottom water and enhance internal loading.The internal waves may also be a cause of the fish kills in Saguling, Cirata and Jatiluhur reservoirs, where the fish farms are located in the shallow bays and the anoxic water starts at the depth of 2-3 meters. In Figure 9, the excess number of farm units shown in three shallow areas may block oxygen replenishment routes; a part of the fish kills. Locating the units in the mainstream and covering only 1-5% of the surface area allows better oxygen exchange and is less risky for fish farmers (see Figure 8).



Laguna de Bay, The Philippines
Courtesy of Bulletin Today, May 2, 1982

Cirata

Jatiluhur
Courtesy of Google Earth, 2011

Figure 9. Excess fish farms in Laguna de Bay in 1982; Cirata and Jatiluhur in 2011

In fish farming, the relationship between the areal loading and phosphorus treatment rate shows highest retention in lakes which receive least pollution per lake surface area. The capacity sharply declines when the loading increases and exceeds the treatment capacity and the lake develops eutrophic. This is already happening in some Indonesian lakes.

Discussion and Conclusions

Lakes equilibriums have a limited tolerance/ capacity (erroneously called self-purification, resilience or carrying capacity) to adjust to and treat external, on-lake and in-lake influences to change. After a certain pollution stress, this threshold is exceeded and the internal loading becomes a significant factor to control. Consequently, the slowly progressing eutrophication may suddenly be a visible and sensed reality (algal blooms, fish kills, smell,

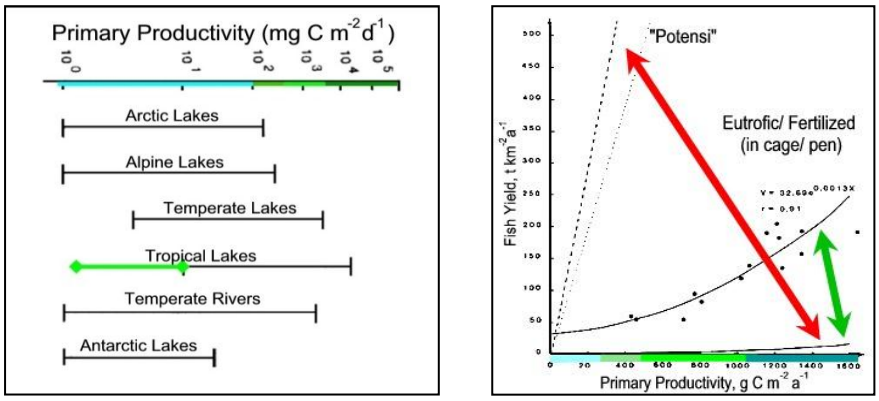
etc.) causing loss of e.g. environmental, economic, recreational, tourist and water and land value. When the point pollution is brought under control, on-lake and internal loads will become the main sources of nutrients and the significance of the agricultural run-off increases.

Primary productivity, fisheries and sustainability of fish resources

Eutrophication has both positive and negative consequences in lakes. Can artificial eutrophication be controlled in lakes, most probably not? The ranges of primary productivity of tropical and temperate freshwater bodies in Figure 10 are based on the data from Hill and Rai (1982) and Tundisi (1983).

In the graph (left), the range for tropical lakes is not necessarily correct for Indonesian large, deep lakes because in such lakes the range of species number is 5-40 and in temperate lakes 18-75 and primary productivity/ phytoplankton biomass is minimal; Batur 2.4 mg l^{-1}, Toba 0.2-0.4, Maninjau 0.2, Diatas 0.05, and Matano 0.002. If fish farms are introduced on such lakes, they do not have enough capacity to treat the nutrient load and dilution is insignificant due to the long water renewal time. In natural lakes, the first fish farms cause local, in the long-term lake-wide eutrophication, which may be difficult to restore.

In Figure 10 (right) primary productivity and fish yields (the dotted and dashed lines) represent the theoretically calculated yields, "potensi", the lowest curve shows the typical annual fish yields from tropical fresh waters and the dots/ curve represent tilapia yields of the fertilized ponds (Almazan & Boyd, 1978). This "potensi" and actual yields are quite apart from each other: Attempts to gain higher yields than the natural productivity are environmentally and economically harmful. The "potensi ikan KJA" for large lakes is 150-200 kg m^{-2} a^{-1} (DKP, 2005) goes beyond the vertical scale and *would need uniformly fertilized lakes* (Figure 11, left), which is not feasible for lake sustainability.



Courtesy of Almazan and Boyd, 1978

Figure 10. Overestimation of the biological productivity of tropical lakes (left) and fisheries yields (right) may create situations, where fishing efforts are not adjusted to the carrying capacity of the fish stocks. There are also wide gaps in the theoretical and actual fish yields (red arrow) and the actual and artificially nourished fish yields (green arrow). Blue color below the horizontal axis indicates oligotrophic and green/ bluegreen eutrophic water.

Figure 11 (left) suggests acceptable (dotted line) and ideal (solid line) total phosphorus concentrations for different use purposes of water. However, the ecosystem capacity to treat nutrients is low and meso- and eutrophic lakes (left, green vertical line) and lakes with total phosphorus concentrations above 50-100 µg l^{-1} (left, red vertical line) are beyond most restoration methods, requiring the best phosphorus load reduction.

The plans of DKP to improve fish yields, livelihoods and export revenue is “suicidal” in lakes like Maninjau, where the *oxygen rich* and immediately oxygen demanding *negative oxygen* layers meet at ~50 m depth. The direct organic oxygen consuming load from surface enhances “pathological” hypoxia, anoxia and fish kills, harming the multi-uses of lakes.

This means that the competent authorities should, *before any fish farming units are located on any lake in Indonesia,* evaluatethe combined areal load of total phosphorus and total nitrogen (g m^{-2} a^{-1}) from i) external diffuse runoff and point sources, ii*) on-lake fish farms* and iii) internally from the deep water and/ or bottom sediment.

Like Maninjau, *many other deep lakes are similar "sleeping bears*" with great amounts of dormant gases, nutrients and other substances blocked into the stagnant deep water below the oxygen rich surface water. These *internally loaded lakes should not be provoked* by fish farms, oxygenation or mixing the layers; instead the deep water could be siphoned out.

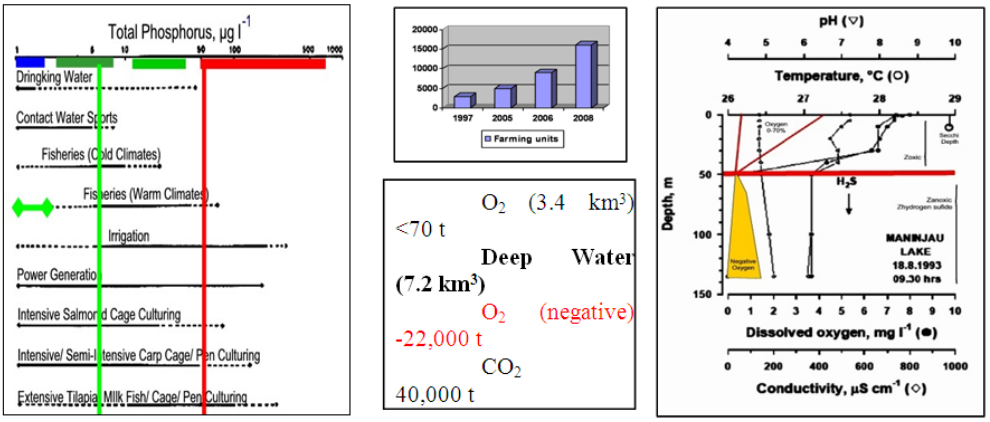


Figure 11. Fertility as total phosphorus concentration vs. fisheries expectations in various climates (left). In Indonesian lakes phosphorus levels are generally low, thus the line for fisheries in warm climates is misleading (green line). Characteristics of Lake Maninjau before fish farming (middle, down and right) & increase of on-lake fish farms (middle, up).

Also water level lowering by increased water withdrawal may reduce hydrostatic pressure and induce diffusive nutrient flux, i.e. “limnic eruption”, from the deep water into the middle/ upper water and earthquakes may aggregate gas bubbling and upward lift of nutrients.

Oxygen and fish

Basically, in lakes only capture fisheries should be supported, especially in lakes which are permanently stratified and have a naturally developed anoxic hypolimnion, which volume is larger than that of epilimnion. There is circumstantial evidence that since 1929 and 1993, the present oxygen conditions in Lake Maninjau are alarming, since the minimum oxygen

requirements for fish is in that temperature at night 7.0-7.5 mg l^{-1}, ~90% saturation, and 5-6 mg l^{-l} to survive. Specific attention should be given to lakes in small islands.

For optimal fish yields, diurnal oxygen saturation range is 80%-110% because at night metabolism and larger fish populations use-up the oxygen reserves. In unfavorable conditions pathological hypoxia, anoxia and fish kills cause great losses in farming (see Figures 5, 9 and 11, right). If the incremental areal nutrient load and biological oxygen demand exceed the treatment capacity of a lake ecosystem and Zoxic/ Zanoxic volume ratios are 0-40%/ 100-60%, permits should be considered and granted permits reassessed.

Conclusions

Freshwater is a finite resource and, in fact, both water demand and pollution are increasing. Thus much less freshwater is available than the potential shows (Wetzel, 2001). It makes it a great challenge to implement the objectives of the Bali Declaration. *Lake managers andthe competent authorities need a substantial knowledge-base on lakes, which requires research, studies and continued monitoring.Only if problems are correctly identified and quantified, the consequences and losses can be eased by the evidence-based management/ restoration programs.* This includes hydrological conditions (water security), keeping-up ecosystem equilibrium and fisheries capacity (sustainability) and especially the multipurpose use values of lakes and of the surrounding land. Lakes and reservoirs are basically for capture and leisure fisheries, which have to be based on the natural productivity. For sustainability of their values, it is better to locate fish farms in river systems or on land.

Regarding assessment of Lake Toba problems: i) the excellent study of the catchment ecology should have been connected to the lake and quantify the annual inflow, silt transport and nutrient loads from rivers (Zulkifli Nasution, 2006) and ii) the House probe team defined declining water level and sedimentation as damages (The Jakarta Post, 2010). However, they are inadequate evidences; the arbitrators should have also focused on the water ecosystem.

To improve evaluation of the ecological quality and changes in lakes, basic research and long-term monitoring are needed. In this respect, three educational programs are essential: i) basic limnology for fine-scale structures, functions and net results of the water ecosystem, ii) management-oriented limnology for understanding the causal-synthetic mechanisms of ecosystems and finding solutions to alleviate problems and iii) fisheries-oriented limnology for understanding trophic-dynamics, fish production and sustainability of fish populations.

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Opportunities for Sustainable Intensification of Agricultural Practices to Improve Crop Productivity of Small Holding Farmers in West Africa

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Abstract

The challenges to increase grain yields, improve ecosystem services, and increase farm profitability in West Africa largely revolve around environmental and economic constraints. Improving ecosystem services, with a focus on soil quality and water quantity will require the adoption of innovative crop rotations that employ both local and improved conservation agricultural practices (CAPs) such as reduced tillage, crop residue, legume cover crop, and integrated nutrient, water and pest management practices. Our goal is to provide food security by increasing economic returns to smallholder farming households dependent on rain-fed agriculture, through gender-sensitive farmer participatory development, and dissemination of sustainable CAPs. Surveys and meetings were conducted with farmers and treatments for testing were identified and implemented. To evaluate long term impacts of CAPS, mother trials with several components/treatment (<5) of CAPS with 3 to 5 replications and baby trials with few (<3) involving several farmers test-plots (78 in Ghana and 60 in Mali) were implemented. Treatments mainly included components of minimum tillage, crop residue management, crop rotations, water harvesting techniques and integrated fertilizer and weed management practices. One of the components of CAPs dealing with permanent groundcover, especially with cover crop is challenging to farmers because of conflicts with livestock management a key component of farming systems; and due to alternative uses of residues and problems with free animal grazing. Soil in most of the farmers fields were poor and had low soil organic carbon in upper west region of Ghana. Sole crops of maize or soybean produced greater grain yields and crop residue than intercropping in conventional tillage or minimum tillage. There was no significant difference in maize yield under conventional tillage or no-tillage in continuous maize cropping system. Soybean yields were increased with fertilizer application, particular phosphorous under both conventional and no-tillage conditions. Soybean growth and yield was not affected by tillage, residue management or the interaction of tillage and residue management. Although residue and grain yields tended to be higher under conventional tillage than under no-till. Maize planted on tied ridges, and tied ridges with grass strips or pigeon pea strips produced higher maize residues than maize planted on flat, and flat with grass strips or pigeon pea strips. Our results suggest that there are some opportunities to increase crop yields through use of intensive sustainable agricultural practices such as application of fertilizer, minimum tillage, crop rotations and water management practices.

Management of Water Saving and Organic Based Fertilizers Technology for Remediation and Maintaining The Health of Paddy Soils and to Increase The Sustainability of Rice Productvity in Indonesia

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Abstract

Rice is belong to the most important grain for Indonesia and act as the main food for majority of the people and it has a great impact on economic and political issue. The rice production in Indonesia is dominated by permanent flooding or inundation system. Intensification of permanent flooding of paddy soils not only reduces the soil biological power significantly, but also restricts the roots growth. The water requirement to produce one kilogram of rice in continuously irrigated fields is approximately 3,000 – 5000 L, while the theoretical minimum at the crop scale is as low as 600 L. Under anaerobic condition, soil organisms cannot growth optimally and estimated only about 25% rice roots can growth normally. Intensive use of inorganic fertilizers, particularly N fertilizers such as urea accelerates the mineralization of soil organic matter. Consequently, soil organic content was decreased rapidly to below minimum content of healthy soils. Various field studies indicated mostly of paddy soils in Indonesia has a low organic content (<< 2%). Under these conditions, the increasing of inorganic fertilizers dosage application may give a non-significant effect on rice production and paddy soils can be categorized as a sick paddy soils. Management of paddy soil health is urgently required to restore, improve and maintain the soils organic matter as heart of soil ecosystem. SOBARI (system of organic based aerobic rice intensification) as water saving technology combined with straw compost based fertilizers technology has two main goals: (1) to remediate or restore, improve and maintain the health and quality of paddy soils by integrating the management of soils soil biological power, water and soil organic matter and (2) to boost the growth and to enhance rice productivity in sustainable ways (efficient water and fertilizer use) by integrating the management rice biological power and integrated fertilizers management. The field results using several rice varieties in Indonesia revealed that the water saving technology combined with organic fertilizers (straw compost) can produce grain yield about 8 – 12 t/ha (average of an increasing about 50 – 150% compared to anaerobic rice cultivation) and the water irrigation was reduced by at least 30 - 50% and as well as inorganic fertilizers was reduced at least by 25% This high rice yield is highly correlated with the increasing of roots zone about 4 – 10 times, number of productive tillers about 60 – 80 tillers, number of panicles, length of panicles and number of grain/panicle, and as well as due to the increase of soil biodiversity. The reuse of straw or straw compost into soils within three years is expected to be able to remediate and improve the health of degraded paddy soils significantly.

Keywords: ***organic fertilizers, paddy soils, remediation, SOBARI, soil health, straw compost, water saving***

Introduction

Rice is one of the most important grains in Indonesia since this crop is the staple food for majority of the people. Increasing rice production to meet the population demand is one of the major issues in Indonesia's development program. Indonesia is the world's fourth largest populous country with 237 million people in the year of 2010, after China, India and United States, respectively. Despite the population growth has declined from 2.4% per annum during the late 1960s, by early 1970s to 1.3%, and in near future expected to decline to less than 0,9%, population size is increasing continuously and it is expected to be double size by 2050 (population closely is to 480 million). The rate of rice consumption is about 139 kg rice per capita per annum belong to the highest in the world compared with Japan, Malaysia and Thailand is only about 45 kg, 80 kg and 90 kg per capita per year, respectively. The currently rice consumption is about 33 million ton per annum and expected to increase to 38,5 million ton by year of 2025 (Suryana, 2008; Simarmata, 2008; Apryantono, 2008: McCulloh, 2008). Consequently, the rice production must be increased by at least 4 – 5 % per annum to ensure the sustainability of food security.

Rice cultivation covered a total of around 10 million hectares throughout the archipelago consisted of about 7,9 million ha paddy soils (lowland rice) and the rest is belong to upland rice. Paddy rice growing in Indonesia has changed dramatically during the last five decades. Either rice production or rice productivity had increased significantly from 9 million ton rice (productivity about 2,5 ton per hectare) in early 1970 to 29 million tons in 1989 (4.23 ton per hectare) to 60 million ton in 2009 (4,6 ton per hectare). The increasing of rice production was highly correlated with the adoption of new technologies, mechanization, chemical use (fertilizers, plant protection agents) and luxurious use of valuable water irrigation (Mutert and Fairhurst. 2002; ISB., 2004; ISB, 2009; Simarmata, 2009: FAO, 2004).

The challenge for agriculture is to increase the production to meet demand for food in a sustainable way. Declining soil health and soil quality, scarcity of water and mismanagement of plant nutrients have made this task more difficult. Can agriculture provide enough food for Indonesia and how is the sustainability of the rice productivity and food security?. The recent data revealed that the highly intensive of fertilizer use that has been introduced over the past five decades probably has reached a point of diminishing returns or leveling off (Simarmata and Joy, 2011; Abdullah *et al*., 2006: Anthofer, 2004: FSD, 2009)

The rapid growing of population in Indonesia not only led to an intensive agricultural practices, but also accelerate land conversion of agricultural land to nonagricultural use and to sped land degradation and environment hazard. The intensive uses of inorganic fertilizers and agrochemical products during the green revolution (at beginning year of 1960) have given a great impact on the decline of soil health and soil quality. Based on recently research, the soil organic content of agricultural soils have been decreased within 30 years sharply. It is estimated about 70% of paddy soils in Indonesia have a low organic content (<2 % of org-C) and low nutrients availability. This paddy soil is belong to degraded soil and can be categorized as unhealthy soils or sick soils and fatigue soils (Simarmata and Joy, 2011: Las *et al*., 2010). The extent of sick paddy soils is increasing continually due to inefficient use of inorganic fertilizers, especially nitrogen and mismanagement of soil organic matter. The excessive use of nitrogen has accelerated the decomposition of soil organic matter (Ingham, 2001: Simarmata, 2009: Abbott and Murphy, 2004: Gupta and Rog, 2004; Sullivan, 2004). A lot of attempt has been done to anticipate and to solve this

problems. The adoption of environmentally friendly agriculture (organic based agricultural and good agricultural practices) becomes more important and growing rapidly. An effort to remediate or restore the health and quality of degraded agricultural soils has been done since the last 10 years by using organic fertilizers and soil ameliorant

The success key of soil remediation (restoring) and maintain of paddy soils health is highly correlated with soil organic matter management and good agricultural practices in sustainable ways. Organic based fertilizers by using the rice straw or straw compost as a main source of organic fertilizers for paddy soils not only restore and improve the soils health, but also reduce inorganic fertilizers significantly. The use of straw compost not only remediate and increase the soils health, but also can reduce application inorganic fertilizers by at least 25% - 50%. Particularly, the supply of silica nutrient and potassium can rely on straw compost application. Every 5 ton straw contain about 70 – 100 kg urea, 50 – 60 kg Super phosphate and 100 – 150 kg KCl (Dobermann and Fairhurst. 2002, Husnain *et al*., 2008; Simarmata, 2009: Turmuktini *et al*, 2010).

Paddy rice cultivation (irrigated rice fields) largely depends on the water supply (irrigation) and it consume about 3000 – 5000 L of water to produce 1 kg of grain rice. The increasing scarcity of water threatens the sustainability of the irrigated rice production system and hence the food security and livelihood of rice producers and consumers. The task becomes more difficult due to global climate change. Therefore, a more efficient use of water is needed in rice production. Several strategies and management are being pursued to reduce rice water requirement, such as alternate wetting and drying, ground cover systems, system of rice intensification (SRI), aerobic rice and raised beds (Uphoff, 2004, Yuan *et al*., 2004, Yang *et al,* 2004; Wikipedi, 2009; Namara, *et al.,* 2004, Ho, 2004; Simarmata and Yuwariah, 2009)

Since 2006 we have developed system of organic based aerobic rice intensification system (SOBARI) to promote the soil biological activity (biodiversity) and to provide a favorable condition for rice roots growth and to increase the growth and yield of paddy rice (Simarmata, 2008). This technology is a holistic rice production system by using and integrating the soil biological power, plant, fertilizers and water management according to the plan and design. The main goals of SOBARI as a water saving and organic based fertilizers technology are (1) to remediate, improve and maintain the health and quality of paddy soils and (2) to increase the productivity of paddy rice in sustainable ways, (3) to promote efficient and eco-friendly paddy soil cultivation and (4) to reduce the inorganic fertilizers application by at least 25%.

The implementation of SOBARI now is beginning widely adopted in several provinces of Indonesia (West Java, Banten, East Java, Central Java, North Sumatera, South Sulawesi, and North Sulawesi, East Nusa Tenggara, Bali and etc.) and it shown a promising and higher rice yield about 50 – 100% compared with traditional methods of continuous flooding (permanent anaerobic) and able to save the water irrigation significantly. This high rice yield is highly correlated with the increasing of roots zone about 4 – 10 times, number of productive tillers about 60 – 80 tillers, number of panicles, length of panicles and number of grain/panicle, and as well as due to the increase of soil biodiversity (beneficial organism) under aerobic condition. Experience with SOBARI technology combined with the recycling of straw as organic fertilizers or straw compost was able to reduce inorganic fertilizers up to 25 - 50%, to reduce water irrigation to 30 – 50%, to enhance the soil biodiversity and nutrient availability and to increase the rice yield by at least 25 % compared to conventional methods or resulted about 6 – 11 ton grain yield of rice per hectare (Simarmata, 2008;

Simarmata, 2009). This paper is a review of SOBARI as a water saving and straw compost based technology for restoring or remediation, improving and maintaining of the health of paddy soils to increase rice productivity and ensure food security in sustainable ways.

System Of Organic Based Aerobic Rice Intensification

Definition

System of organic based aerobic rice intensification (SOBARI) is a holistic rice production system by integrating the soil biological power, plant, fertilizers (organic, biofertilizers and inorganic) and water management to achieve targeted main goal (by design). The main goals SOBAR are to achieve and to sustain high rice productivity, promotes and enhances agro-ecosystem health (soil quality & soil health), including biodiversity, biological cycles and soil biological activity. Therefore, the maximize of local input, especially organic fertilizers (straw compost, cow dung or others) and biofertilizers becomes the main priority and called as low external inputs for sustainable rice cultivation (LEISRC) (Simarmata, 2008).

The management of local inputs is designed to optimize the biological processes in achieving the desired rice productivity (outputs) and environmental quality. System of organic based aerobic rice intensification is working with the nature by optimize of organic- and biofertilizers application, water and culture of management to promote soil biological power and minimized the use of inorganic fertilizers.

Pillars of SOBARI

The key success of system of organic based aerobic rice intensification (SOBARI) in increasing rice growth and soil biological activity (soil biological power) in soils are highly depend on the four basic concepts or pillars of SOBARI, as follows: (1) the of paddy soil ecosystem, (2) living soil as natural fertilizers factory (biofertilizers plant) or bioreactor, and (3) biological power of rice and (4) integrated nutrients management.

Paddy ecosystem change

Traditionally paddy rice ecosystem is characterized by permanent flooding or inundation. Consequently, all aerobic organisms cannot life in soils due to oxygen depletion, which is required for aerobic respiration. Depletion oxygen leads to anaerobic conditions (anoxic conditions) and give a significant negative effect on the beneficial organisms activity and roots growth. By changing the rice ecosystem from anaerobic (permanent flooding) to aerobic conditions resulted a dramatically change in soils, especially on soil biological activity and the roots growth. The moist soils up to muddy condition provide oxygen for the respiration of soil organism. Meso- and micro fauna play an important role to create and to build a tunnels system, which is very important for air and oxygen supply under muddy condition. It seems, these tunnels support the biological activity in soil and it produced a lot of cast on soil surface. This cast is containing either macro- or micronutrients and as well as essential growth substances, like vitamins. In contrast under inundation or permanent flooding, there is no activity of aerobic and produced no cast on paddy soil surface (Uphoff, 2004; Hengsdijk, and Bindraban 2001: Simarmata, 2008; Bouman *et al*., 2002)). In addition, the roots growth and microbial activity was increased significantly under aerobic to muddy condition (Figure 2).

Figure 2. Roots system and tiller number of SOBARI rice under muddy condition/aerobic is about 3–5 greater than permanent flooding rice/anaerobic (Simarmata, 2007)

The population of beneficial microbe (nonsymbiotic nitrogen fixers such as *Azotobacter sp*&*Azospillum sp* and phosphate solubilizing bacteria) and roots growth were increased highly significantly compared with anaerobic conditions (Simarmata, 2008). The rice roots system under aerobic to muddy condition was about 5 – 10 times greater than flooding ecosystem. It has been revealed only about 30% of rice roots growing well. Consequently, the potential obtained yield of various rice varieties is the work of only 30% rice roots. If roots system growing optimally in step with the rice production, the potential yield of various rice variety may increase by at least 2 three times. Therefore to change permanent flooding rice ecosystem to aerobic conditions (field capacity to muddy) is absolutely necessary to increase the rice production and to save water irrigation significantly.

Living soil as natural biofertilizers factory

Paddy soil ecosystem under aerobic to muddy condition is living system and highly complex system characterized by various of biological, chemical and physical processes, which markedly influenced by environmental factors. Microorganisms inhabit the soil and together with exocellular enzymes and the soil meso fauna and macro fauna conduct all known metabolic reactions. A hectare of fertile healthy topsoil contains approximately 1200 kg of bacteria, 1200 kg of actinomycetes, 2400 kg of molds, 120 kg of algae, 240 kg of protozoa, 51 kg of nematodes, 120 kg of insects, 1200 kg of worms and 2400 kg of plant roots One per gram moist soil in rhizosphere contains approximately 1,200 x 10^6 of bacteria, 46 x 10^6 of actinomycetes, 12 x 10^5 of fungi and 5 x 10^3 of algae (Sullivan, 2001, Ingham, 2001). All these organisms from a tiny bacteria up to the large earthworms and insects will eat, grow and interact in the soil ecosystem to form food web that influence paddy soil ecosystem significantly. Food web is the community of organisms living all or part of their lives in the soil and it named as the living component of soil. Therefore under aerobic to muddy condition, paddy soil is life and act as natural fertilizers factory or biofertilizers plant. Contribution of soil beneficial organisms for nutrient availability was well known. The nonsymbiotic nitrogen fixers may contribute up to 50 – 100 kg N per hectare, phosphate solubilize microbes may increase the availability of P up to 50%. In addition, the mineralization rate of added straw was faster and provided more nutrient and

energy for biological activity to support the rice growth. Flooding permanently or under anaerobic condition will lead to close of natural valuable fertilizers factory, mean while the farmer spend a lot of money to buy in organic fertilizers (Uphoff, 2004, Simarmata, 2008).

Paddy soil as natural fertilizers factory (bioreactor) need a sufficient energy sources to allow the machine working properly. Energy source for biological machine is mainly depends on organic availability. Organic material is entry point of chemical energy to paddy soil ecosystem. Consequently, the activity of heterotrophic microbes, like bacteria,fungi and actimycetes are rely on organic supply. The present of microbe as first trophic level allow the energy flow to next trophic level in soil food web. As organisms decompose complex material, or consume other organisms, nutrients are converted from one form to another form, and are made available to plants and to other soil organisms. Soil-dwelling organisms release bound-up minerals, converting them into plant available forms that are taken up by the plants growing on the site. Organisms which are not directly involved in decomposing plants wastes may feed on each other or others wastes products or the other substances they release. Among the substances released by the various microbes are vitamin, amino acids, sugars, antibiotics, gums, and waxes, which are very important to sustain the soils remain alive and healthy. It is not surprisingly that aerobic to muddy condition of paddy soils are tilled and fertilized by soil organisms (Ingham, 2001: Sullivan, 2004).

The performance of Natural Fertilizers Factory (Biofertilires Plant) is highly depend on water management and organic supply (organic fertilizers). The water management or irrigation supply is aimed to maintain the paddy soil condition under field capacity to muddy conditions and to meet the water requirement of rice. Water conditions or water level is regulated through water canal. The distance between of water canal may range from 5 – 10 m depend on the level of paddy soils. The rice straw is used as a main source organic material to supply chemical energy the soil ecosystem and essential nutrient (macro- and microelements) for rice plant. The application of composted straw or other organic fertilizers play an important role to stimulate the growth of beneficial microbes (N-fixers, phosphate solubilizing and phytohormone producing bacteria) and to secure the of organic matter supply for soil fauna, which are act as soil engineers. The application of improved organic fertilizers (high content of humic substances, relative high of nutrition contents and other bioactive substances such amino acid, sugar and vitamins are highly recommended. The benefits of this products includes easily handling and application, relative lower in application rate (dosage), the standardized quality, relative free from contaminants (weed seeds and soil born diseases) and lasted longer in soils.

Biological power of rice

Since long time ago, people and scientist believe that rice is an aquatic plant and grows best in standing water. But recently was known that rice is not an aquatic plant. Although rice can survivewhen its roots are continuously submerged under water but does not thrive under hypoxic conditions. Rice does not grow as well in standing water as when its roots are able to get oxygen from direct contact with air. Under submerged conditions the roots grow is limited and rice plants spend lot of its energy and some of the roots' cortex disintegrates to form air pockets (*Aerenchyma tissue* pockets (aerenchyma) so that oxygen can reach root tissues. In addition, under flooded conditions up to 3/4 of roots may die by the time of flowering (panicle initiation) (Uphoff, 2004, Simarmata, 2008). Field results revealed that wider space combined with good water and nutrient management increased the tiller number and growth significantly (Figure 3).

Figure 3. The tiller number about 60 – 80 per clumps and growth performance of SOBARI rice (varierty of Ciherang), wihile the flooding rice produced only 20 -30 tillers (Doc. Simarmata, 2006 – 2007).

Water management is focusing to maintain an aerobic to muddy condition in paddy rice ecosystem. Young seedling 8 – 12 days old are planted twin (two of singly seedling are planted in line with 5 cm distance) combined with wider plan space (30 cm x 30 cm or 30 cm x 35 cm is highly recommended) to allow the roots of singly seedling grow independently at the beginning stage. Output oriented of organic based fertilizers is aimed to secure the supply of nutrient sufficiently and to maintain the optimum biological activity in soil ecosystem

Nutrients management

The intensive rice practices lead to the excessive nutrients removing from the paddy soils ecosystem. Fertilizers application of traditionally rice cultivation is focused only on primary nutrients such as N, P, and K, especially nitrogen (very cheap due to subsidized price). Consequently lead to nutrients depletion in soils. In addition, the excessive of nitrogen application will accelerate the decomposition of soil organic matter and harm the environment (soil quality, water and air pollution). At least sixteen and possibly nineteen elements are known to be essential for plant growth. All nutrients have to be present in adequate amounts according to plant development or stage of growth to ensure the high rice productivity. Therefore, the management nutrition or fertilization of SOBARI is based on outputs oriented (Figure 4).

The amount nutrients supplied into agro-ecosystem as inputs are calculated according to the plants requirement for supporting plant development and growth to achieve the targeted productivity (*outputsby design*). The plants needs of nutrients as raw materials based on the plant development. The nutrients availability in soils and agro-ecosystem (soil properties, landscape and climate) condition and as well as on the characteristic of applied fertilizers (solubility, salt index, reaction, etc.) must be integrated in designing of inputs management (raw materials management). It is necessary to keep the balance between inputs and outputs in order to avoid either nutrients depletion or excessive of nutrients supply, nutrients disorder in soils and to maintain the sustainability of natural resources. In the long run, it is possible to obtain high yield on sustained basis only when the nutrients balance is positive (when the input is larger than removal). Thus all removed nutrients from the soil by plants must be replaced in an ecologically responsible away.

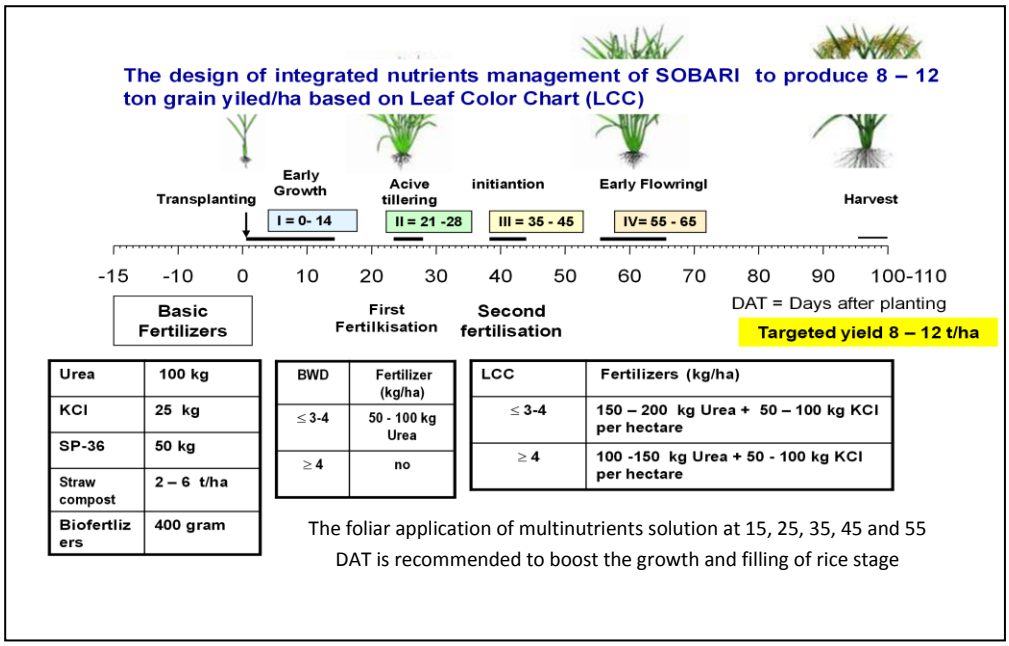

Figure 4. Design of integrated nutrients management output oriented of SOBARI based on leaf color chart (LCC)

In SOBARI is recommended the use of a balance application of organic and mineral fertilizers. The widely used of fertilization methods in SOBARI to produce about 8 – 12 ton rice grain yield (targeted production) are as follow; (1) the incorporation of rice straw or straw compost is done during the land preparation, (2) one day or before transplanting about 50 kg of urea, 25 kg KCL and 50 kg SP-36 (as alternative is 50 kg urea and 100 kg NPK) were applied into soil, (3) application of about 50 - 100 kg urea is done at 18 – 21 days after transplanting or after the first weeding (leaf color chart can be used as indicator to determine the nitrogen rate), (4) 50 - 100 kg urea and 25 - 50 kg KCl or 50 kg urea and 100 – 150 kg NPK (16:16:16) is applied 35 – 38 days after transplanting. In addition, enriched organic liquid fertilizers or multinutrition solution is sprayed to plant at 15, 15, 25, 35, 45 and 55 days after transplanting.

Summary of SOBARI field results

SOBARI is now widely adopted by incorporating the rice straw during land preparation. Young singly seedling is transplanted at 8 – 12 days old with wider spacing (30 cm x 30 cm or 30 cm x 35 cm) in square pattern. Seedling is planted at the marked intersection of planting space (25 x 30 cm, or 30 x 30 cm or 30 x 35 cm). A single seedling is planted in twin methods (two single seedling is planted in line about 5 cm distance from each others at point of planting cross section, called as Twin Seedling Method : “SOBARI-TS”) to allow the roots system of seedling growing properly at the beginning stage. The seedling is planted by slipping in sideways rather than plunging it into the soil vertically and it makes the shape of the transplanted seedling more like an L than like a J. With an L shape, it is easier for the tip of the root to resume its growth downward into the soil.

Water applications or irrigation is required to regulate the soil moisture under field capacity to muddy conditions until the beginning of grain ripening stage. The water regulation is

necessary to allow rice roots growing properly and to stimulate the growing of soil organisms and as well as its biodiversity. One or two days before weeding (manually or mechanically), the rice field is irrigated with 1- 2 cm depth or a thin layer to allow the removing of weeds the easily and to improve the soil aeration. Usually, weeding is done 3 times (2, 4 and 6 weeks after transplantation).

The field results of SOBARI from several Provinces or districts in Indonesia are summarized in Table 1. As shown in Table 1, adoption of SOBARI with various rice varieties under different planting season in several provinces of Indonesia are fairly easily to achieve 40 – 60 fertile tiller plant and contains about 150 – 250 grains per panicle. The average yield was ranged from 8 to 10 ton grain yield per hectare (about 50 – 100% higher than traditional flooded paddy rice). The highest yield about 12 ton per hectare in 2008 was obtained in SOBARI Experimental Research Station of SOBARI of Agriculture Faculty of Padjadajaran University,Bandung. SOBARI method raises not only the yield of paddy(kg of unmilled rice harvested per hectare) without relying on improved varieties, but also to increase the outturn of milled riceby 5 – 10%**.** This "bonus" on top of higher paddy yields is due to having fewer unfilled grains and fewer broken grains. In addition, based on field results and report of farmers groups and extension services have verified that SOBARI crops are more resistant to pests and diseases and as well as more resistance to abiotic or climatic stress (drought). The length of the crop cycle (time to maturity) is also reduced by 5 – 10 days. The earlier harvested time is highly correlated with no seedling stress during transplantation and after planting the roots can grow optimally without lag phase.

The key success of SOBARI depends on water irrigation and fertilizers management. The water supply by irrigation is focusing to maintain the soils under moist to muddy conditions to allow the roots system and soil biota growing optimally. Based on field experiences, the SOBARI method may reduce the water irrigation use by at least 30 % - 50 %. Output oriented integrated fertilizers application of SOBARI is aimed to provide a sufficient nutrient to meet the plant requirement based on targeted yield by design and to improve the soil quality and soil health. Therefore, the application of organic fertilizers and biofertilizers are need to increase nutrients availability and biological activity in soils. SOBARI is focusing on the application of rice straw as a main source of organic fertilizers. Actually, organic fertilizers in the form of rice straw is the main product of paddy rice cultivation. It well known that ratio between straw and grain yield ranged from 1.0 – 1.5 times. Straw is the only organic material available in significant quantities to most rice farmers. About 40 percent of the nitrogen (N), 30 to 35 percent of the phosphorus (P), 80 to 85 percent of the potassium (K), and 40 to 50 percent of the sulfur (S) taken up by rice remains in vegetative plant parts at crop maturity (Dobermann and Fairhurst, 2002).

Nutrient content of every 5 ton rice straw is equal to 50 kg N, 10 kg P_2O_5 and 120 kg K_20. Straw is also an important source of nutrients such as zinc (Zn) and silicon (Si). By incorporating the rice straw during land preparation combined with biofertilizers may reduce the application inorganic fertilizers significantly. Based on field report, the application of 2 – 6 ton/ha of compost straw combined with the application biofertilizers and enriched organic liquid fertilizers as multinutrition solution (Nazarudin *et al*., 2009; Simarmata, 2009) reduce the usage of inorganic fertilizers by 50%. SOBARI recommends the inoculation of piled straw with decomposer (sellulolitic and lignolitic combined with *Trichoderma, sp*) about 2 – 3 weeks before spreading in the rice field and incorporation during the land preparation to accelerate decomposition of straw and to suppress the pathogenic microbes in straw

Table 1. Unmilled Rice Yield Summary of the System of Organic Based Aerobic Rice Intensification (SOBARI) in Different Province/District in Indonesia (Planting Season 2007 – 2009)

Location	Yield (t/ha	Control (t/ha)	Increment (%)
SOBARI Experimental Fields and Demo Plot (Faculty of Agriculture of Padjadjaran Univ. Bandung), planted Rice (Ciherang, Dyahsuci, IR 64, Mekongga)	6 – 12	4 – 7	50 –150%
Demo Plot in Research Institute for Rice of Indonesia, harvested on July 2008 (Ciherang, Mekongga and Sintanur	7 – 10	-	-
West Java (Bandung, Garut, Sumedang, Bogor, Bekasih, Subang, etc.) with various rice variety	6 - 11	4 – 6	50 –100 %
Banten (Serang)	6 – 10	4 – 6	50 –100 %
East Java (Tulung Agung, Jombang, Madiun, Blitar, Mojokerto, Nganjuk, etc.)	6 – 10	4 - 6	50 –100 %
Central Java (Sragen, Sukoharjo, Wonogiri, Karang Anyar, Purworejo, Magelang, Semarang, etc.)	6 - 10	4 - 6	50 –80%
Nort Sumatera (Sergei, Tebing Tinggi, Tapanuli, etc)	5 – 10	3 - 7	50 –100 %
South Sulawesi (Gowa, Luwu, etc.) conducted by Famer Groups in 16 Subdistrict (Various Rice Variety)	6 – 10	3 – 6	50-200%
North Sulawesi (Minsel, Minut, etc.) conducted by Famer Groups with Various Rice Variety	6 – 10	3 – 7	50 –200 %
Nusa Tenggara (Kupang, Ende, Bajawa, Nagekeo, Rote Ndao) conducted by Famer Groups with Various Rice Variety	6 - 10	2 – 6	50 –300 %
West Kalimantan (2 ha)	8,9 ton	-	100%

Sources: Agricultural Services Agency of Bogor (2008), Agricultural Services Agency of South Sulawesi (2009), Agricultural Services Agency of Bajawa and Ende (2008), Agricultural Services Agency of South Minahasa (2008), Rice Institute for Rice of Indonesia, 2008, PT. SMS, 2008-2009

Management for Remediation The Health of Paddy Soils

Basic concept

The concept of restoring or remediation of soils health is based on basic function of healthy soils under natural ecosystems, such as (1) to provide an excellence growth media for roots system and soil organisms (2) regulating and partitioning water and solute flow; (3) filtering and buffering, degrading, immobilizing, and detoxifying organic and inorganic materials, including industrial and municipal by-products and atmospheric deposition; (4) storing and cycling nutrients and other elements within the earth's biosphere (5) to sustain and maintain a diverse community of soil organisms (soil biodiversity) which are important to control plant disease, insect and weed pests, to form beneficial symbiotic associations with plant roots, (6) to improve soil quality, water and nutrients holding capacity, (7) improve crop production (Seybold *et al*, 1998, Doran *et al*, 1996; 2000; 2004, Ingham, 2001).

Straw compost for restoring health of paddy soils

Briefly, the main function of organic fertilizers like compost straw in soils will act as: (1) a revolving nutrient bank account. Rice straw is derived from plant residues and it contain all

essential plant nutrients (macro and micronutrients) and chemical energy resulted from photosynthesis. Therefore, biomass of straw as organic matter is the storehouse for nutrients and chemical energy. Either nutrients or energy are released by microbial process (mineralization or decomposition and oxidation of organic substances). The released nutrients are highly to plant (plant-available form) and the stable organic fraction (humus) adsorbs and holds nutrients in a plant-available form, (2) entry point of energy flow (supply) into soils ecosystem, (3) an agent to activate and regulate the biological system in soils .and (4). an agent to improve soil healt and soil,quality (Simarmata, *et al*., 2011). Consequently, organic fertilizers in form straw compost plays an important and has a vital role in soils and it may categorized as heartof paddy soil ecosystem.

In addition, straw compost which is rich organic carbon (30 – 40 %) contain about 1.5% N, 0,3 – 0,5 % P_2O_5, 2 – 4% K_2O, 3 – 5 % SiO_2) and micro nutrients, such as Cu, Zn, Mn, Fe, Cl, Mo. Actually, the main product rice cultivation is not grain yield itself but the organic fertilizers in form rice straw. In General the straw production is about 1.5 x rice grain yield. If productivity of rice is about 6 – 8 ton grain yield/ha, than the production of straw is about 9 – 12 ton/ha. The nutrients contain of ice straw and it potential to substitute the inorganic fertilizers is presented in Table 2.

Table 2. Nutrients content of straw, economical value (IDR) and its potential to substitute the main inorganic fertilizers (Simarmata and Joy, 2011).

Nutrients	Contain (%)	kg/ton straw	Equivalent in 5 t straw kg/ha	
C-Org	40	400	2000,0	
N	0,65	6,5	72,2	Urea
P	0,1	1	13,9	SP-36
K	1,45	14,5	161,1	KCL
Ca	0,6	6	30,0	
Mg	0,2	2	10,0	
Si	5,5	55	275,0	
S	0,10	1	5,0	
Rice production in 2010 = about 60 million ton or 90 million ton of straw				

Equiavlent :			IDR
1.299,60	MT	Urea	4.678.560.000.000
250,20	MT	SP-36	900.720.000.000
2.174,40	MT	KCL	7.827.840.000.000

The price ***Urea Rp 3600 (Rp 1200, SP-36 Rp 4000 (Rp 1850) dan KCL Rp 6000***

As shown in Table 2, the reuse of straw as an organic fertilizers can substitute the inorganic fertilizers up to 50% of major nutrients, such N, P and K., while other nutrients (Ca, Mg, Si, S) and micro nutrients (Cu, B, Zn, Fe) could be fully substitute. The application of straw is also act refilling or recharging of the fuel or energy into soil ecosystem, every 5 ton straw will supply about 2000 kg organic carbon into soils. Thus, the activity of soil organisms are highly correlated with the supply of straw which act as energy source (fuel).

The composting of straw with appropriate technology will increase not only nutrient contents, beneficial organisms and but also important to control the pathogen microbes which has been existed in the straw. The increasing population of organisms in straw compost (bacteria, fungi, animal and others) will improve the quality of straw compost significantly (Figure 5). In general, the straw compost content about 30 – 40 % of total carbon (rich in humus and organic acids), 1 – 1,5 %, 0,5 % P_2O_5, N, 2 – 3 % K_2O, 3 – 5% of SiO_2 and other essential nutrients. In situ composting of straw has been developed (Simarmata and Joy, 2010) for the remediation of paddy soils in Indonesia (Figure 6).

Figure 5. In situ direct composting of straw (A = preparation of decomposer inoculant, B = compost box, pilling of straw and inoculation, C= Straw pile, D= covering straw pile G & H = mycelium of fungi is growing well on straw compost and I = mature straw compost (Simarmata & Joy, 2011).

Figure 6. Spreading and incorporation of straw compost and following with land cultivation or preparation (Simarmata and Joy, 2011)

Application Methods

Briefly, the application of composted straw can be applied or incorporated (1) before land preparation and (2) after transplanting for remediating or restoring the soil health of paddy soils.

(a). Before Land Preparation. Shortly before land cultivation with tractor, the composted straw (normally 2 – 3 weeks has a C/N ration to 30 - 40 after 2 – 3 weeks composting process) and it can be applied on the surface of paddy field homogenously. Subsequently, the land is cultivated using tractor or others equipment to incorporate the straw compost into the soils (Figure 2.3). In general one week after incorporation, the C/N ratio of soil will be decreased to below < 20 and the rice seedling or rice seed can be planted.

(b).After Transplanting. The application of mature straw compost (C/N ratio about 20 or less) is applied about 2 -3 weeks after transplanting. The incorporation or placement of compost straw within plant row is done after the first weeding (Figure 2.5.). During this activity, the weeds that still exist and growth closely to the rice clumps can be eliminated manually. Subsequently, it is recommend to applied of inorganicfertilizers to boost the plant growth.

Figure 2.5. Application of straw compost within plant row after the first weeding (about 2–3 weeks after transplanting). The straw is placed or incorporated into soils by stepping (Simarmata and Joy, 2011)

Field Results and Close Remarks

The application of compost straw or straw has shown a great impact on the improving the soil quality and soil health. The content of organic carbon, availability of nutrients and on biodiversity of organisms in soils were increased significantly. The application of 2 – 6 ton/ha of compost straw increase the rice yield and reduce the rate of inorganic fertilizers significantly (Table 3).

Table 2. Efect of straw compost the rice grain yield under different rate of potassium fertilizers (Nazarudin *et al*., 2010)

Straw Compost (ton ha^{-1})	Rate of KCl (kg ha^{-1})			
	0	50	100	150
0	2.90 a A	3.80 b A	6.20 c A	6.44 c A
2	4.20 a B	5.48 b B	8.49 c B	8.54 c B
4	7.21 a C	9.28 c C	9.49 c D	8.76 b C
6	8.84 a D	9.20 b C	9.20 b C	9.10 b D

Note: Value number within row or column followed by the same letter (capital for vertical in column) is not different significantly according to the LSD 0.95 test

It is estimated that application of 2 – 4 ton straw compost within 3 years (about 6 planting season) will be able to restore and improving the health of paddy soils. As indicator of the soil health recovery can be seen on the main indicator such as the biological, chemical and physical indicator. Briefly, the content of organic carbon must be much more than 2% and availability of major nutrients is at least belong to medium category. Subsequently, it is need the management of soil organic matter by using the straw as` main source of organic fertilizers. The habit to burn of straw must avoid or forbidden strictly. The straw burning not only polluted the environment but also led to the loss of major nutrients significantly. About 91% C, 91% N, 45 % P, 70 % K, 50 % Ca, 20 % Mg and others nutrients were loss due to the straw burning. (Husnain *et al*., 2008).

The success key of restoring and maintaining of soils health for sustainable rice cultivation to ensure the food security in Indonesia is highly depend on management of soil organic matter. The use of straw as the main source of organic fertilizers is absolutely correct. It has been ready available on site. Therefore it belong the low cost and effective agent that can be used as agent for re-healing and to maintain the health of the soils ecosystem. Thus, the SOBARI combined with management of rice straw can be implemented to speed the paddy soil remediation and to increase the rice productivity in Indonesia in sustainable ways.

Conclusions

The conclusions and suggestions can be summarized as follows:

1. System of organic based aerobic rice intensification (SOBARI) is a holistic of water saving and inorganic fertilizers reducing technology focusing on soil biological and rice power management and as well as integrated fertilizers management to increase rice production and to improve the soil quality significantly. It can reduce the water irrigation by at least 30 - 50%, seed rate about 75%, and inorganic fertilizers by at least 25%, and increase the rice yield about 50% - 100% compared with traditionally flooding rice cultivation.
2. The main goals of SOBARI as water saving and organic based fertilizers technology are (1) to remediate, improve and maintain the health and quality of paddy soils and (2) to increase the productivity of paddy rice in sustainable ways, (3) to

promote efficient and eco-friendly paddy soil cultivation and (4) to reduce the inorganic fertilizers application significantly.

3. Adopting the SOBARI as water saving and environmentally friendly cost reducing technology combined with straw and integrated nutrients managements will promote and speed the remediation or restoring and improving of paddy soils health and increase the sustainability of rice production
4. Furtherer research are need, especially on; (1) a comprehensive water requirements, (2) soil biological activity (macro- and microorganisms) and roots system under different water conditions, The application organic and Biofertlizers to substitute or reduce the inorganic fertilizers (5) the performance of roots system, growth and yield various rice variety under different planting season.

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New Fruit Technologies in Europe

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Abstract

Fruit production in Europe had a fast evolution in the last two decades. Important changes occurred in the variety and rootstock assortment in all species. By introducing medium and low vigor rootstocks it was possible to increase the planting density even in fast growing species as stone fruits. The new high density orchards imposed a radical change of the orchard management and a new range of growing technologies. The innovations always started in the fruit nursery and recently, new typologies of planting material are proposed. Soil preparation for the new orchards is sometime under the sign of the replanting problems due to the monoculture. New planting systems and tree canopies have been developed with the aim of better light use, ensuring high yield, high quality, but low training and maintenance pruning costs. In correlation, different hail and rain protection systems are in use. Grass cover soil management is generalized between rows, while on the row, herbicides or mechanical tillage is used. Fertilizers and irrigation are two important tools to control the tree growth and production. Generally, simple soluble fertilizers are applied with the irrigation water in function of the vegetation stage and plant needs. Depending of the production area, several systems of crop protection against wind, frost, hail, rain, birds are in use. Plant protection faces new challenges due to some specific requests of integrated and organic production systems, to the annual reduction of the approved pesticides number, to the rise of new pests and diseases and also due to the consumer concerns regarding fruit residues. Hand fruit picking is facilitated by self-propelled machines with fruit bins loading and downloading systems. For processing fruits mechanical harvest is largely used. Continuous develop and innovation of orchard technologies in European fruit industry is requested in order to maintain its sustainability and competitiveness, by producing top quality fruits, with nearly to zero residues and no environmental negative impact.

Keywords: ***crop protection, fertirrigation, fruit picking, fruit tree propagation, plant protection, planting systems, soil management, thinning***

Introduction

The evolution of the fruit production in Europe in the last century was marked by three important stages. The first one, a classic period, characterized by low planting densities of 300-400 trees/ha, high volume canopies (pyramid, leader, vase, etc.) ended in the 60' by adopting some medium vigor rootstocks, as MM106 in apple.

The intensive plantations were realized by reducing the distance between rows at 4-5 m and by increasing the planting density to over 1,000 trees/ha. Typical hedge canopies as Italian palmete, Simple palmete, Fan palmete, but also low volume canopies as Slender Busch, Slender Spindle, and Spanish open vase have been introduced. The intensive orchards were managed using a special adapted range of tractors and machineries.

In the 80' started the era of superintensive orchards with the high density apple orchards realized by the generalized use of low vigor rootstocks (M26, M9, M27). Planting systems as Super Spindle, Pillar System etc. brought the planting density over 8,000 tree/ha (Corelli

Grappadelli, 1997). There have been some tentative to further increase the planting density over 40,000 trees/ha in the biannual bearing “lawn system”.

After being promoted few years by nurseries, the very high density orchards trend diminished, especially because of the extremely high investment cost and of some fruit quality problems due to the bad light penetration. Some alternative canopy and planting systems as Drilling and Mikado, have been introduced (Widmer and Krebs, 1996).

In the last two decades, beside important changes in the variety and rootstock assortment in all species, the planting density stabilized between 2000 and 4000 trees/ha (DeJong *et a*l., 2006). Even in the fast growing species as stone fruits, it was possible to increase the planting density by using medium and low vigor rootstocks. Rootstocks as Gisela 5 in sweet cherry, Ishtara in plum, apricot and peach produced a real revolution in the orchard planting systems.

New Fruit Technologies

Nursery production

The fruit nurseries are generally the promoters of innovation and recently, new typologies of planting material are proposed. Stone fruits rootstocks are mostly *in vitro* micropropagated with all the benefits of this method: rapid propagation, uniformity, virus free, strong roots etc. Instead pomes fruits rootstocks are still propagated by traditional layering methods.

The first improvement of the planting material was the production of the feathered trees – knip. Even if initially, it was realized for apple trees, nowadays it is generalized for all the fruit species with multiple advantages on early fruit bearing and orchard establishment.

To reduce the number of planted trees/ha and so, the initial investment value, recently it was promoted by Mazzoni nursery in Italy, the Bi-baum® type of fruit trees. The trees are produced by double bud grafting with two vertical feathered axes, instead of a single one.

Micrografting is a new technique applied recently on large scale for the production of the pot grafted fruit trees. Using *in vitro* produced rootstocks, the micrografting is applied in January or early February under protected conditions. Scions start to grow and in late May or June, the potted trees are ready to be planted in the orchard. Until the winter time, the trees reach the high of 2.0-2.5 m. The method reduces the time of the production of grafted trees at 1.5 years, instead of 3-4 years and is applicable mainly to stone fruits.

In vitro micropropagation of varieties and cultivation on their own roots (without grafting) was successfully applied for some pear varieties (Stănică *et a*l., 2000), kiwi plants, olive trees, walnuts (Navacchi *et a*l. 2008) and is in the way to be extended for apricots, sweet cherries, peaches and other species.

Soil preparation

Sometime the soil for the new orchards has replanting problems due to the previous orchard monoculture. In that case, few supplementary measures are essential: removal of all roots rests of the previous crop; cultivation for 3-4 years with a legume crop, mainly

alfalfa or clover; one year cultivation with *Tagetes sp*. for soil restructuring and nematodes elimination (Ploeg, 1999); soil disinfection; fertilization etc.

In order to ensure good soil physical properties before planting is necessary a deep work with a ripper in two cross directions, at 60-80 cm depth.

Planting

Because of the high density and small planting distances between plants on the row, recently the trees are planted on open ditches (Figure 1). To increase the influence of the rootstock on the future tree growth and productivity, it is necessary to plant the fruit trees with the grafting point at least 20 cm above the ground.

Figure 1. Apple tree planting on open ditches

Planting distances are influenced by the species, the variety/rootstock vigor and by the canopy type (Table 1). Autumn is the best planting period for bare rooted trees. For potted trees planting can be executed during all the vegetation period, but is indicated to be finished before the end of June. After planting, fruit tree needs to be tide on the trellis system and usually, no pruning is applied. By exception, few vigorous lateral branches can be eliminated in order to balance the rapport with the tree leader.

Planting systems and tree canopies

Modern canopies and planting systems have been developed with the aim of better light use, ensuring high yield, high quality, and in the same time, low training and maintenance pruning costs (Sansavini and Corelli Grappadelli, 1997, Corelli Grappadelli and Marini, 2008.).

Introduction of knip-feathered trees have a major role on the orchard architecture and production:

- the single leader type canopies (Super Spindle, Vertical Axe, Solaxe etc.) have been generalized;
- fruits trees are full of lateral fruiting branches and fruit production starts in the first year after planting;
- under the fruit weight, the branches start to have a weeping position with a strong tendency for fruiting;
- pruning is limited to the elimination of the vigorous lateral branches;

Taking in consideration the fact that the new low vigor rootstocks have a superficial root system, usually a trellis system needs to be installed in order to sustain the fruit production. In the same time, the trellis can be used as a support for the hail protection nets and for the drip irrigation pipes. After many years of orchard experience with the single leader canopies, some new canopies with double axe - Parallel V, or even triple axe – Trident, have been introduced.

Table 1. Tree canopies and planting systems for apple and pear

Specie	Tree canopy	Distance between rows (m)	Distance on the row (m)	Trees/ha (no)
Apple	Spindle (Vertical axe)	3.5-4.0	1.2-1.6	1500-2000
			0.9-1.3	2000-2500
			0.8-1.1	2500-3000
			0.7-0.9	3000-3500
			0.6-0.8	3500-4000
	Mikado	3.8-4.0	1.6-1.8	1300-1500
	Drilling	3.8-4.0	1.2-1.4	1700-2000
	Solen	3.5-4.0	1.0-2.0	1200-2006
Pear	Spindle (Vertical axe)	3.5-4.0	1.5-2.0	1250-1850
	Palmete	3.0-3.5	2.0-2.5	1100-1450
	Mikado	3.8-4.2	1.5-2.0	1100-1750
	Drilling	3.8-4.2	1.2-1.7	1350-2550
	Gard Belgian	3.0 -3.5	1.0-1.5	1800-2650
	Drapeau Marchand	3.0 -3.5	1.7-2.5	1100-1600

The new type of double axe canopy (Parallel V) is recommended for the more vigorous varieties and species, because of the distribution of the growth energy on two directions. Bi-Baum® tree type is ideal to form a double axe canopy by providing a balanced structure ready to produce fruits (Musacchi, S. 2008). Bi-Baum® was first recommended for apple (Figure 2) and pears, but there are some new researches for extending the system to the more vigorous stone fruits species.

Figure 2. Bi-baum® System (Double axis) at apple

Similar studies have been developed the concept of the Trident canopy, the trees having three vertical parallel axes (Figure 3). Even the first yield is obtained in the second year after planting, by reducing the planting distances three times, the initial investment diminishes. Recently there are some approaches regarding the asymmetric orchard with interesting results regarding the light reception and tree photosynthetic efficiency (Losciale P. *et al*, 2010).

Figure 3. Trident (Triple axis) canopy at apricot

Soil management

After decades of research on different soil management systems, grass cover is now generalized between the tree rows. Grass is obtained by seeding or by natural covering.

The most important advantages of the grass cover system (Figure 4) are represented by the positive effect on the soil structure, soil content in organic matter, soil microbial activity, orchard biodiversity etc. In the same time, the soil erosion, water loss, energy consumption and the man work are reduced. In grass covered orchard it is possible to circulate and to do typical works after heavy rains or hail.

Figure 4. Soil grass covered management

To control the weeds on the row, herbicides or mechanical tillage are used. In the conventional and in some integrated orchards, herbicides (mainly glyphosate based) are sprayed on a 0.8-1.0 m wide strip along the tree row (Stefan and Bucur, 2000).

In order to reduce the chemicals use, alternative methods of weeds control are applied. One of the most efficient is the mechanical tillage on row made with different machines with trunk sensing device. The treatment is useful for soil aeration and for stimulation the roots growth in the depth.

Recently, some experiments have been done by covering the soil with reflective much. Besides the advantages of artificial mulching, the reflective mulch has the main task to improve fruit coloration (Costa G,*et al*., 2003).

Fertilization and irrigation

Tree growth and productivity are influenced by some technical operation as pruning, root pruning and fruit thinning, but irrigation and fertilization are the most two important tools. Based on the vegetation stage, plant needs and soil and plant agrochemical analyses, simple soluble fertilizers are applied with the irrigation water. The most important advantages of the method consist on direct root distribution and efficient use of the fertilizer, low energy request and low level of pollution.

Crop protection

Depending of the production area, several systems of crop protection against wind, frost, hail, rain, birds are in use.

Wind protection is necessary in windy areas, where the frequent winds can seriously affect the tree upright position and in the same time the fruit crop. Mixed species windbreaks are recommended to be planted around the orchard and even in the interior part of it. The windbreaks effectiveness on horizontal line is 10-15 times longer than the windbreak height. Within the windbreak different height species are planted from the smaller ones at the exterior to the taller ones in the central part.

Late frosts are serious challenges for early flowering species (stone fruits) but also, in some high altitude areas, for the normal flowering ones (apples and pears). One of the most diffused antifrost systems is the overhead irrigation but it needs a special knowledge regarding the timing, the water quantity etc. Other systems are using different air mixing devices as high tower wind machines or horizontal fans and recently, the mobile gas burners (“Frostbuster”) the work by the principle of energy transfer to the flower organs.

Hail protection is provided by the use of special plastic nets (Fig 5). If the initial hail nets were black, nowadays, the white ones (Crystal) are used. Hail net is installed normally on the same structure used as trees trellis, slightly modified and reinforced. The use of colored hail nets is still under research, different colors having different physiological effect (Vittone, 2006).

Figure 5. Apple orchard protected with anti hail net

Modified hail net by closing the lateral and the front side of the parcel is used frequently as a tool for orchard protection against birds especially in cherry. Many tests and research are in progress for replacing the hail net with an insect proof net in order to keep out and control specific orchard pests.

By using a special polyethylene transparent and reinforced fabric is possible to protect the orchard against rains. The technique is mainly used for sweet cherry and berries during the picking period in the rainy areas and it could provide also an earlier ripening (Figure 6). The trellis system in this case has to be stronger than the antihail one, by doubling the pole number and by a special anchorage.

Figure 6. Raspberry production under rain protection

Plant protection

Plant protection is a delicate subject and a difficult task for the fruit grower. New challenges regarding some specific requests of integrated and organic production systems, the annual reduction of the approved pesticides number, the rise of new pests and diseases are imposed because of the consumer concerns regarding fruit residues (Zavagli 2008, Zavagli, 2011).

Some important prevention principles are recommended to be followed:

- mixed planting of resistant and less sensitive varieties,
- orchard sanitizing and respect of hygiene rules,
- late and early treatments (dormancy period) to reduce the inoculums,
- summer pruning etc.

Alternative fighting methods are requested:

- insect mass capture and male disruption by the use of pheromones,
- installation of light capture systems and use of other physical methods of combat,
- biological control,
- use of repellent and insectproof bioproducts,
- installation of insectproof nets,

In the same time, when applying chemical treatments, farmers need to consider seriously some important elements:

- treat only when necessary (by monitoring and prediction models),
- use the best spraying technique (Loquiet *et al*. 2008),
- choose pesticides with shorter persistence or higher reporting limit,
- choose non toxic pesticides for birds, fish, honey bees, birds, warm blood animals etc.
- apply new chemicals more specific and less toxic,
- reduce doses (adapted to the risk, adapted to the canopy – EMR studies),
- increase intervals between treatment and harvest.

Fruit picking

In order to establish the right moment for the fruit picking, some traditional and modern methods are used. The number of days from fruit setting till the ripening moment is normally known for every cultivar. Apart of that, different changes of fruit characteristics as base color, covering color, seeds color, taste etc, could indicate the approach of picking maturity.

Some more elaborated methods are analyzing the changes in the fruit starch content, by coloring the fruit section with iodine in potassium iodinate. Reduction of coloration because of the starch hydrolisation into soluble sugars, shows the fruit maturation stage.

Fruit flesh firmness evolution is also used to appreciate the ripening status, by using appropriate device called penetrometer. New undisruptive methods are proposed to determine the optimal picking moment. The NIR (Near Infrared) technique creates the possibility of analyzing some fruit biochemical parameters on the tree (Peirs, 2003).

A similar method was developed by an Italian research group of the Bologna University by creating a system of light absorbance measurement with the aid of DA meter (Costa *et al*., 2010).

For fresh consumption hand fruit picking is still the most popular. To facilitate it, self-propelled machines with fruit bins loading and downloading systems are used (Figure 7). For processing fruits mechanical harvest is largely used. For processing in many fruit species, the mechanical harvest is applied (Corelli Grappadelli, 1988).

Figure 7. Self propelled machine for fruit picking

Conclusion

European fruit industry faces new challenges imposed by the necessity of maintaining its sustainability and competitiveness. Production of top quality fruits with nearly to zero residues and no environmental negative impact, requests the continuous develop and innovation of orchard technologies.

The future fruit production needs to be able to survive by reducing the inputs (specially the one based on conventional forms of energy), the man work and by adapting continuously to the climate changes and consumer needs (Bertschinger L., 2009).

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Empowering Business of "Garut Sheep" for Small Holder Farmers in West Java

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Abstract

There were approximately 6.2 million heads of sheep in West Java in 2010, consisting oflocaland Garut sheep as known as indigenoussheep. Localsheepdeveloped inWestJava is known as Priangan sheep, and historically as a result of crossing among local, Kaapstad andMerinosheep. The origin of the Garut sheep is believed from Garutregency, in which in Cikajang (Cibuluh, Cikandang, andCikeris villages)andWanaradja Districts. Most farmersinthetwodistrictsbelievedthat thesheepis a genetic resourceofGarutRegency. The ear of the sheep was mainly rudiment (rudimentaryear/rumpung on sundanese) or ngadaun hiris (sundanese), like aleaf of Cajanus cajan a smalltriangular. The tail was ngabuntut beuritor ngabuntut bagong (thintaillike a tail ofrat or tail of wild boar).Garut sheep is mostly kept as fighting ram or contest for pleasureorhobby. During ram fighting and contests, it is accompanied withtraditionalmusicandpencaksilat(traditional dance). Based on supporting carrying capacity, socialand economic development, WestJava has a high potential regionfor Garut sheep in Indonesia. In order toempower the business, the farmer'sorganizationsneed tobe developedintensivelyto be Breeder Associations, intensive researchon thegeneticimprovement, educating the consumers,and governments need todevelopcentral breedingfor the Garut sheep.

Keywords: empowering, farmers business, Garut sheep

Introduction

The population of sheep in West Jawa in 2010 was approximately 6.2 million heads, consisting oflocaland*Garut sheep* as known as indigenoussheep. Localsheepdeveloped inWestJava is known as *Priangan sheep*, and historically as a result of crossing among local, KaapstadandMerinosheep. The origin of the Garut sheep is believed from Garutregency, in which in Cikajang (Cibuluh, Cikandang, andCikeris villages)andWanaradja Districts. Most farmersinthetwodistrictsbelievedthat thesheepis a genetic resourceofGarutRegency. The ear of the sheep was mainly rudiment (*rudimentaryear/rumpung*) or *ngadaun hiris* (sundanese, *like aleaf of Cajanus cajan a smalltriangular*). The tail was *ngabuntut beuritorngabuntut bagong* (sundanese, *thintaillike a tail ofrat or tail of wild boar*) (Heriyadi andSurya, 2004;Heriyadi, 2011).

Garut sheep belongs to world genetic resources and is known as prolific sheep. Sheep plays an important role in farming systems in west Java and provides valuable commodities such as meat and organic fertilizer in the villages. Moreover, it has also has an important function for saving bank for smallholder farmers. Almost 90% of the sheep are bred in small holding. The management regime is also strongly associated with specific socio-cultural conditions and is variable from one village to other villages. Cut and carry system is a typical management, where the forage and other feeds are brought to the animals raised in confined housing. These conditions lead to the difficulties in evaluating the animal across the villages and in designing a breeding scheme.

West Java has a high potential in development of sheep, given with the potential for their own sheep, carrying capacity ofthe community sheep farmers, andotherphysicalenvironments. The governmentof WestJavahas outlined thelivestockdevelopmentprogramtoincreasesheep populationto be 10million heads.However,sheep farminginruralareainWest Javais generallyaside businessanddomestic meat sheep consumption is still low. This paper was addressed to review the possibility of empowering “Garut sheep" business in rural farmer’s communitiesinWestJava.

Development of "Garut Sheep" Farming

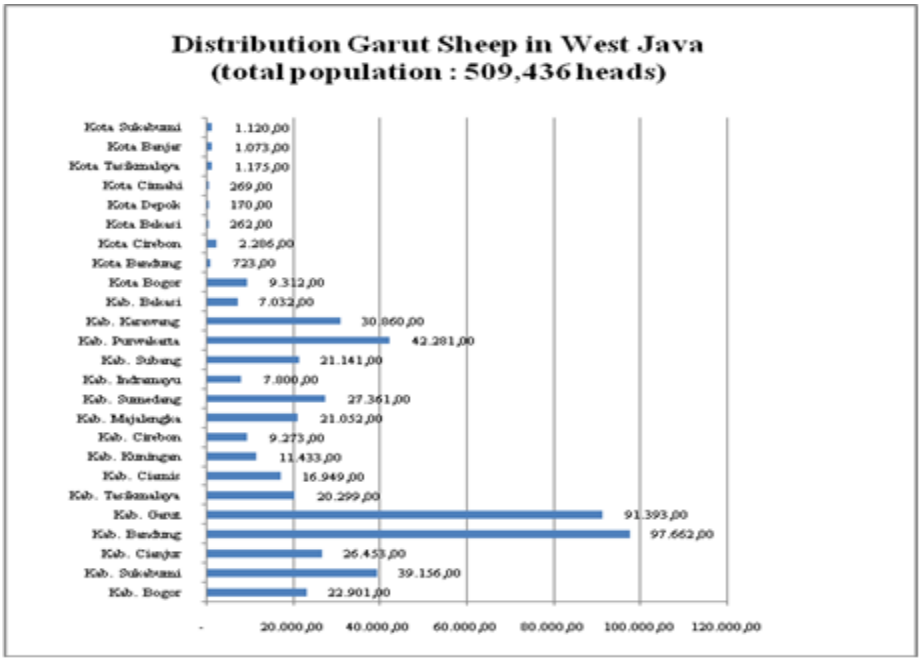
Population of goats and sheep in Indonesia in 2010 was only 8.86 percent of total livestock population, or approximately 26,747,946 heads, consisting of 16,110,709 heads goat 10,637,237 heads sheep, respectively. Low ratio population indicates that there is insecure population if the consumption increases drastically, thus although there is an increase in meat consumption, breeding population and productivity to produce lambs should be stable, in order to give the benefit to livestock farming as well as to increase in demand consumption or market.

Although the biggest increase in meat production came from sheep (15.3%), the consumption of lamb was still low at only 0.20 g / capita / year (Strategic livestock Planning, Directorate General of Livestock Services, 2010-2014), compared to France 13.89 g/capita/years, UK 16.94 g/capita/years, Australia 52.50 g/capita/years, and New Zealand 81.11 g/capita/years, respectively. The comparison with countries in Asia was also was still low, with only 10% of domestic consumption in India and about 3.9% of consumption in China (Heriyadi, 2009).

Sheep meat consumption needs to be promoted by informing to give the benefits for health, intelligence, and the brain performance, especially for brain development. Recently, consumer preferences for sheep meat consumption reached 2.0%, which was below the consumption of beef and chicken. The priority of meat production should be better directed to increase domestic consumption.

Based on the potentialconsumptionin the future, inWestJavawhichhascontributed57.98% of thenationalsheeppopulation(Department of Livestock Services West Java, 2010), has great potentialto contribute tothe production ofSheepmeatnationally.Contributionof livestockproductionof sheep, the sheepare stilldominatedbylocalsheep(*Priangan sheep*), while the *Garutsheep* witha bettergeneticpotentialthanany otherlocalsheep, should be developedin orderto increasethe productivity ofsheepinWestJava.

The Illustration below shows the distribution of Garut Sheep population in West Java across thedistricts. The sheep was well distributed in all areas, which directly had an impact to the carrying capacity of thesocial, economic and physicalsupport, particularly inthe central mountainregionsof WestJava. The population was closely related to theculturalcommunity, especiallycommunity of farmer groups (Garut sheep Farmers Association) inconducting an activityof*ram fighting contests.*



Source: Heriyadi, 2010

Performance Garut Sheep

Rahmat (2005) has studied on Garut Sheep, in three different locations, in which Margawati as government breeding centre, H. Osih and Lesan Putra as farmers. The results are summarized in Table 1.

Table 1. Body Weight, Body measurements, and Litter Size

Traits	Farm	Sex	Birth Type		
			Single	Twin	Triplet
Birth Weight (kg)	Margawati	Male	2.81	1.90	1.40
		Female	2.49	1.61	1.25
	H. Osih	Male	3.59	2.98	1.99
		Female	3.09	2.68	1.88
	Lesan Putra	Male	2.76	1.95	1.87
		Female	2.47	1.76	1.55
Weaning Weight (kg)	Margawati	Male	10.81	8.51	6.78
		Female	9.97	8.00	6.56
	H. Osih	Male	12.22	11.64	8.75
		Female	10.95	10.61	8.48
	Lesan Putra	Male	11.63	8.51	6.78
		Female	9.96	8.00	6.56
Liter Size (%)	Margawati		39.38	55.16	5.47
	H. Osih		40.16	50.82	9.02
	Lesan Putra		47.96	46.94	5.10

On Table 1; approximately 50% of Garut sheep gives twin birth, followed by single birth with approximately 45%, and triplet 5%. An increase in litter size leads to a decline in birth weight of lambs. Birth weight and weaning weight were also influenced by sex and birth type. Male is heavier than female for both birth and weaning weight. There is a trend that the famers are more favourable to breed single birth for fighting ram rather than twin and triplet birth. These are indicated by percentage of single birth in H. Osih and Lesan Putra was higher than the percentage of single birth in Margawati.

Mature body measurements of Garut sheep, including wither height, body length, heart girth, and chest width, and mature body weight are presented in Table 2.

The results above are essential for standardization of Garut sheep in a real origin population in West Java. The avarage body weights were 57.74 kg and 36.89 kg, for ram and ewe, respectively. These measurements could be used also for standardization in other areas for mature body weight.

Table 2. Body Measurements and Body Weight

Traits	Ram	Ewe
Body Weight, kg	57.74 ± 11.96	36.89 ± 9.35
Wither height, cm	63.41 ± 5.72	56.37 ± 4.58
Body length, cm	88.73 ± 7.56	77.41 ± 6.74
Heart girth, cm	74.34 ± 5.84	65.61 ± 4.85
Chest width, cm	22.08 ± 8.21	16.04 ± 2.05

Heriyadi, *et al*. 2002

In general, rations Garut Sheep consisted of forage and concentrates. Forage consisting of grass, legume, and the leaves are given singly or a mixture of two kinds of forage or mixed, while the concentrate is varied depending on the habits of each farmer. Most farmer gives concentrates consisting of mixtures of two or three kinds of feed ingredients, some give commercial concentrates. But in addition to forage and concentrates are given, almost all the farmer gives additional feed such as eggs, honey, herbs, supplements or drinks, especially just before and after a contest or at certain times as it deems appropriate (Heriyadi, 2005).

The Contest of Garut Sheep

AccordingtoSuleiman*et al*l (1999),Garut Sheep waskeptby thefarmersfor pleasure or hobby. The performance was shown in the ramfightingcontests, with usually being accompanied bytraditionalmusicandof *Pencak Silat* (traditional dance). The art of ram fightingis usually held inthe fields, wheretworams arecontestedbyclashingheadswith acertainagreement before the match. The decision of the winner is determinedbya referee. The history of art ram fighting began around the year 1937 in the village of Cibuluh, by the establishing a representative field as an initiative of *Mama Rubai* and *Ki Tasik*. The ram fighting was accompanied with well performed and strong fighter ram. The championship was part of the art with followed traditional drums.

Recently, the contest and ram fighting of *Garut sheep* have beeninstitutionalizedin Sundanese ethnicin WestJava, which is implemented regularly everySunday. The implementation has been locatedincertain locations, likewise contestof livestockinsome countries. Based on research conducted by Suleiman et. all. (1999), Garut

sheephusbandryhas contributedsignificantlytohousehold income, althoughthere is nostandardmarketprice ofthe benchmarkquality ongenetic quality.

Empowering Garut SheepBusiness

To empower business of Garut sheepfarming, several things thatneed to be concerned; Firstly, the development oflivestockshould be on the linewith culture. For example, inWestern countries, animal husbandry is on the line with*cowboyculture*, inIndiaandBali, the cattle is partoftheculturaland religionin all regions, thus thecattle farming businessgrows rapidly. In West Java, sheep farmingis moreculturallysanctionedandit is conservedby asocietyof Sheep and Goat Farmers Association as loversandfans ofGarut sheep. The contest is conducted everyweekin many regions. This condition should continue to preserve thebusiness as well astheir geneticquality. The historyGarutsheep need to be publishedby "open museum" likein severalcountries, such as Bali cattle is shown in Martin-Luther University museum in Germany. In addition, it is necessary tostrengthenthe presence ofGarutsheepin West Java.

Secondly, the researchersfromuniversities andresearch institutions, especially governmentof WestJavaprovince need to conduct in-depthandcomprehensiveresearch, particularlyin genetic potential of Garut sheep, in order togainworldwide recognition.

Thirdly, Demand forsheep, particularly forsheepmeatconsumption. It is necessary to inform to the consumerson thebenefits of sheep meat in particular. The perception of sheep meat containinghighcholesterol should be abolished, by the importance to consumesheep meat sinceearly age.

Fourthly, it is requiredthe government to setup the policiesregardingzoningfor the central development Garut sheep breeding farm, base on carrying capacity ofthe regionof WestJava

Conclusions and Recommendations

1. Based on the supporting carrying capacity and social and economic development, WestJava has a high potential region to develop and improve Garut sheep in Indonesia.
2. In order toempower abusiness in Garut sheep in West Java, farmer organizations need to be developed intensively to be a Breeder Association, intensive researchon the genetic quality of Garut sheep, conducting education to consumers and government need todevelopcentral breeding and suitable breeding design

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Biotechnology to Ensure Food Security

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Abstract

Food security consists of multiple dimensions of Food Availability, Physical Access to food, Economic Access and Food Utilization. Food availability is made up of production, imports and stockpiles, each of different importance to people from food excess or food deficit geographic areas. In the mid 20th Century, food production was a major agenda for international development aid and the subsequent "Green Revolution" of the 1960s helped avoid mass famine in many parts of the world. Today, in a globally connected food supply chain, export of food is becoming more important, and dependent on the excess over consumption at the source of production. Maintaining this excess in the face of growing demands for food and a declining resource base to grow food, as well as challenges from alternate uses for food biomass, all require significant increases in productivity and total yield production in the coming years. The Food and Agriculture Organization, U.N. has estimated that food production will have to increase by minimally 50% to keep pace with demand by 2050. Biotechnology is one set of technologies which offers potential to significantly increase production by increasing yield potential and reducing losses, and concurrently, ameliorating the effects of climate change on food availability, physical access and its utilization. Biotechnology applications ensure food security by increasing the supply of crop-based foodstuffs and include a) Improving conventional breeding using marker aided selection, b) Diagnostic and early detection tools for reducing losses caused by pests and diseases, c) Increasing the knowledge of genetics and ecology for managing yield and losses (Biodiversity management), and d) Genetic engineering for improved yield and pest resistance traits using transgenes (genetically-modified or "GM" crops). New traits to address changes in the biotic (insect, disease, weeds) and physical (soil, water, nutrients) environment have proven successful in countering extreme weather events arising from climate change. Many of these new traits, such as for drought and submergence tolerance, insect and disease resistance, have been derived through biotech tools such as genetic engineering or marker-aided selection. However, beyond R&D, farmer adoption of new technologies is an issue. A combination of supporting policies, technology and appropriate knowledge extension is required to address food insecurity using biotechnology.

Diversification of Food Products to Support Food Security: Development of Food Products Based on Sorghum Rice and Flour

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Abstract

In Indonesia grain sorghum was known since 1925 and cultivated in the semi-arid regions of Java and Nusa Tenggara islands. Production is still low, hardly any statistics are available. In 2008 central Java produced about 1.483 tons from 1.291 ha. Renewed interest in sorghum in West Java began in 1999 as cattle feed, mainly due to its resistance to drought. Demand for wheat flour in Indonesia is rising continuously as a consequence of population increase and changes in way of living particulary in urban areas. Dependencany on few crops such as rice and wheat flour may be solved by food diversification to ensure national food security. Various commodities suggested are root-crops (cassava, sweet potato and arrowroot) and minor cereal crops (sorghum, job's tears, millet). There are three types of grain sorghum based on tannin content, respectively type I (non-pigmented testa), type II (low-pigmented testa) and type III (pigmented testa and pericarp). Decortication produces sorghum-rice, which is more palatable and acceptable, because it is removes both pericarp and the testa to various degrees to yield sorghum-rice of yellowish white to greenish white color. Decortication may be done by traditional, mechanical and chemical methods. The overall quality of the decorticated product depends on inherent characteristics of the grains and the method of decortications employed, but good results for all three types of sorghum could only be obtained by abrasive decortication. Sorghum-rice could be used to manufacture sorghum flour. Sorghum-rice and flour are versatile food materials. Sorghum flour is free of gluten and can be added to a variety of food as they are almost taste and scent free and naturaly has a color ranging from creamy to pinkish or reddish brown depending on the sorghum cultivar and hulling efficiency of the sorghum-rice. Sorghum-rice and flour doesn't have good binding properties, which may be solved by applying specific prosessing or cooking methods such as cooking to induce gelatinition, pressure cooking or adding various food additives such as hydrocolloids, starches, food protein, sugar and mixing with other flours including wheat flour. Attempts have been done to manufacture food based on sorghum-rice respectively 'kerupuk', 'keripik', 'opak', 'rengginang' (rice-crispis), puff sorghum, snack based on puffed sorghum and food based on flour respectively bread, sweet rolls, cake, noodle and various cookies.

Introduction

Grain sorghum (*Sorghum bicolor* L. Moench) is native to tropical Africa. It is a leading cereal grain in both Africa and India, and now ranks fifth among the world's most important grains, respectively rice, wheat, corn and barley. The world's largest producers of grain sorghum are USA, India, Nigeria and Mexico, while the leading exporters are the USA, Australia and Argentina (US-Grain Council, 2008)

In Indonesia grain sorghum was known since 1925 and cultivated in the semi-arid regions of Java and Nusa Tenggara islands. Production is still low, hardly any statistics are available. In

2008 Central Java produced about 1.483 tons of sorghum grains from a total area of 1.291 ha (Anonymous, 2006).

Sorghum is inherently a very efficient crop, it emerges quickly and produces a rapid ground cover; it is efficient at using limited amounts of water and nutrients and has a high growth rate. It may be cultivated in various cropping systems repectively inter-cropping, sequential-cropping, relay-cropping, ratoon-cropping and rotational-cropping (Gruben and Partohardjono, 1996). Consequently cultivation of sorghum has great prospect especially for regions depending on rainfall for irrigation (Sihono, 2008; Sirappa, 2003).

Renewed interest in sorghum cultivation in West Java began in 1999, when some of Unpad's researchers got involved in an Agribisnis development project to use sorghum plants as cattle feed, mainly due to it resistance to drought. However, farmers involved in this project insisted besides feed the crop should also produce grains that can be sold for additional income. Since then research were carried out involving plant breeding, crop cultivation, postharvest handling and processing of sorghum grains. Recently plans have been made by the BPPT and The Indonesian National Reseach Institute (LIPI) to carry more advanced research on sorghum. Sorghum generally posseses seeds of bronze, brown to brownish red color due to high tannin contents. Pericarp and tannin must be removed to make the grains edible and acceptable. On the other hand a high tannin content makes the crop more birdproof and disease resistant. Nutritionally, however tannins react with proteins causing indigesbility of these proteins. Moreover tannins also cause sorghum products to be astringent and reddish of color (Rooney and Sullines, 1977 vide Sirappa, 2003).

Sorghum grains may serve as substitute for rice as well as wheat flour. Sorghum contains 70 – 90% carbohydrates, 7.4 – 14.2 % protein, 2.4 – 6.5 % fat, 1.2 – 3.4 % ash, containing iron, phosphor and calcium. Tannin content range beween 0.4 – 3.6 %. Tannins are present mainly in the pericarp and testa (Plessis, 1998).

Decortication of sorghum grains removes both the pericarp and testa from the endosperm (Sinuseng and Prabowo, 1999). The pericarp have a tendency to adhere firmly to the kernel depending on thickness. Both pericarp and testa contain pigments, while the sorghum endosperm is either white or yellowish white. Degree of decortication could be measured by hulling efficiency (FAO, 2004).

Sorghum-rice (decorticated sorghum) could be used as a substitute for rice and rice-based food products. It can also be milled to yield sorghum flour which may be used to substitute wheat flour either partially or totally. Demand for wheat flour in Indonesia is rising continuously as a consequence of population increase and change in way of living particulary in urban areas. Most urban citizens consume fast food and snacks which are mostly based on wheat flour.

Dependencany on few crops such as rice and wheat flour may be solved by food diversification. Various commodities suggested include employing root-crops (cassava, sweet potato and arrowroot) and minor cereal crops (sorghum, job's tears, millet). In this way Indonesian food security problems might be solved.

Types of Seed Sorghum and Decortications of Sorghum Grains

Types of Seed Sorghum

The sorghum seed consists of pericarp (bran) 6.0 – 9.3%, embryo (7.0 – 12.1%) and endosperm 80.0 – 84.6 % (Rooney and Miller, 1982; Wall and Ross, 1970). Grain sorghum maybe classified into 3 categories based on tannin content (Figure 1), respectively type I (non-pigmented testa), type II (low-pigmented testa) and type III (pigmented testa and pericarp) (Hagerman *et al*., 1988 vide Waniska, 2000).

Various breeding programs to obtain sorghum types adapted to Indonesian conditions have been done. The result are UPCA-S1, UPCA-S2, Katengu, Feterita (Mudjisihono and Suprapto, 1987), B-100, B-95, B-72 and B-68 (Soeranto, 2004) and Unpad 1.1 a white sorghum type (Anas, 2009).

tannin sorghum (Local cultivar) | white sorghum (Genotype 1.1.) | tannin sorghum (Genotype B-100)

Figure 1. Types of Grain Sorghum

Decortication of Sorghum Grains

Decortication of grain sorghum produces sorghum-rice, which is more palatable and acceptable because it is removes both pericarp and the testa to various degrees to yield sorghum-rice of yellowish white to greenish white color. Low tannin and tannin sorghum yield sorghum-rice of yellowish white color with reddish specks at the hyllum. Structure of sorghum grain is presented in Figure2.

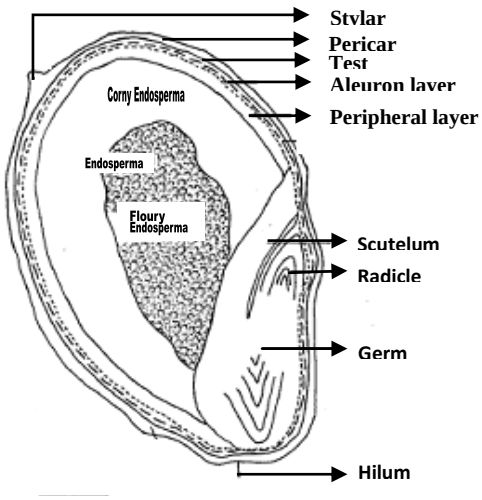


Figure 2. Sorghum Seed Structure (Rooney and Miller, 1981)

Decortication may be done by traditional, mechanical and chemical methods. The overall quality of the decorticated product depends on inherent characteristics of the grains and the method of decortication employed. White sorghum could be decorticated by the three pearling methods with satisfactory result. Low tannin and tannin sorghum grains could not be pearled with satisfactory result by the alkali debranning and traditional method (using mortar and pestle), but good results for all three types of sorghum could be obtained by abrasive decortication (Figure3).

White sorghum (Unpad 1.1) | Low tannin (genotype B-100) | Tannin sorghum (indigenous cultivar)

Figure 3. Sorghum-rice Decorticated by Mortar and Pestle Methods

Degree of abrasive decortication of sorghum could be measured by hulling efficiency (HE) (FAO, 1994). Sorghum-rice of the Unpad 1.1. and indigenous cultivar pearled to various hulling efficiency are presented in Figure4.

Indigenous Cultivar

(0') (1') (1,5') (2') (2,5')

White sorghum (Unpad 1.1)

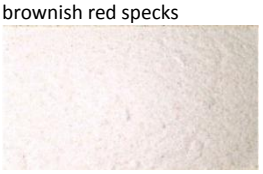
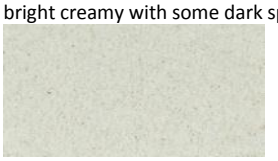
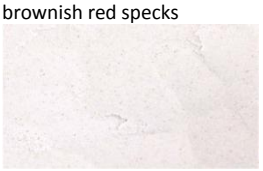
HE: 100% (0') HE: 39,36% (5') HE: 30,33% (7,5') HE: 24,25% (10')

Figure 4. Sorghum-rice with Various Hulling Efficiency (Mardawati, *et al*., 2009)

Sorghum Flour Milling

Sorghum grains can not be stored more than two months due to pest and disease infestation, but sorghum flour may be stored for more than six months (Dogget, 1988). Flour production involves grinding and refining cereal grains by a process called milling.

Table 1. Wholemeal and Sorghum Flour Fractions of to Two Sorghum Cultivar

Flour Fraction (Mesh)	Indigenous	Unpad 1.1
> 120	pinkish yellow with a small amount of brownish red specks	bright creamy
101 – 120	 pinkish yellow	 bright creamy with some dark specks
81 – 100	 pinkish yellow	 bright creamy
61 – 80	 pinkish yellow with a small amount of brownish red specks	 bright creamy with a large dark specks
41 – 60	 pinkish yellow with a small amount of brownish red specks	 bright creamy with some dark specks
21 – 40	 pinkish yellow with a small amount of brownish red specks	 bright creamy
> 80	 pinkish yellow with a small amount of brownish red specks	 bright creamy with some dark specks
Wholemeal	 pinkish yellow with a small amount of brownish red specks	 bright creamy

Sumber: Mardawati, *et al*., (2010)

Usually before milling the grains are subjected to a brief conditioning treatment (2–3 hours) to remove pericarp in large pieces and efficient separation of the desirable products (Gani, 2011). Small scale milling is done by hammer mill or disc mill and than fractionated by sieving. Generally cereal flour posseses a particle size more than 80 mesh. Yields of sorghum wholemeal was about 91-93% but sorghum flour more than 80 mesh was about 35%. The cultivar Unpad 1.1 yields bright creamy flour with some dark specks and the indigenous type yields pinkish yellow flour with a large amount of brownish red specks (Table 1). Sorghum flour has more advantages than sorghum-rice, it is more practical and can be enriched and mixed to yield composite flour suitable for producing various food products.

Food Products Based on Sorghum-rice and –Flour

Sorghum-rice Based Food Products

Sorghum-rice cooked by traditional method respectively boiling or steamming are has a sandy texture, due to the germ and embrionic disc. This can be removed by sieving or cooking in pressure cooker. The last method is more practical and has been used for manufacturing of food products such as crackers and other snacks ('kerupuk', 'opak', 'keripik').

Sorghum-rice cooked by traditional method and pressure cooking have a loose non- sticky texture. Therefore to produce 'opak' or 'keripik' a binder is needed such as tapioca. However in "kerupuk' manufacture the tapioca starch has to be pre-gelatinized and gelatinized. Type and concentration of binding agent for manufacturing various snack food based on cooked sorghum-rice is shown in Table 2. Moreover sorghum-rice still has reddish brown specks (except the white cultivar), that give specific coloring to the food product (Fig 5).

Table 2. Type and Concentration of Binding Agent for Manufacturing Various Snack Food Based on Cooked Sorghum-rice

Food Product	Type and Concentration of Binding Agent	Sorghum-rice Cultivar
Keripik	7.5% tapioca	Unpad 1.1
Keripik	7.5% tapioca	Indigenous
Kerupuk	Gelatinize 40% tapioca	Unpad 1.1
Kerupuk	Gelatinize 60% tapioca	Zh-30
Opak	40% tapioca	Unpad 1.1

indigenous Keripik | Unpad 1.1 keripik | Unpad 1.1 Opak | Zh-30 kerupuk stick

Figure 5. Snack Products Based on Sorghum-rice

Sorghum Flour Based Food Products

Sorghum flour is gluten free and contain a small amount of phenolic compounds. Gluten free flour may be used to produce food for people alergic to gluten, such as celiac deseases patients and autistic children. Tannin content of the sorghum flour causes a pinkish or brownish color, but also have antioxidant properties (US Grain Council, 2008). Sorghum flour color may be covered up by using food colors, cinnamon, cocoa powder, etc. Sorghum flour doesn't have good binding properties, which may be solved by applying specific prosessing or cooking methods such as cooking to induce gelatinization, pressure cooking or adding various food additives such as hydrocolloids, starches, food protein, sugar and mixing sorghum flour with other flours including wheat flour (composite flour). Substitution of sorghum flour in the production of various food products are shown in Table 3 and Figure6

Table 3. Percentage Sorghum Flour in Various Food Products

Food Product	Percentage Sorghum Flour	Binding Agent	Sorghum Flour Cultivar
Bread	20%	80% Wheat flour	B 100
Sweet rolls	30%	70% Wheat flour + sugar	Unpad 1.1
Cookies	100%	38% sugar	Unpad 1.1
Pound Cake 1	70%	30% Corn starch	Indigenous
Pound Cake 2	70%	30% Corn starch + 2% xanthan gom	Indigenous

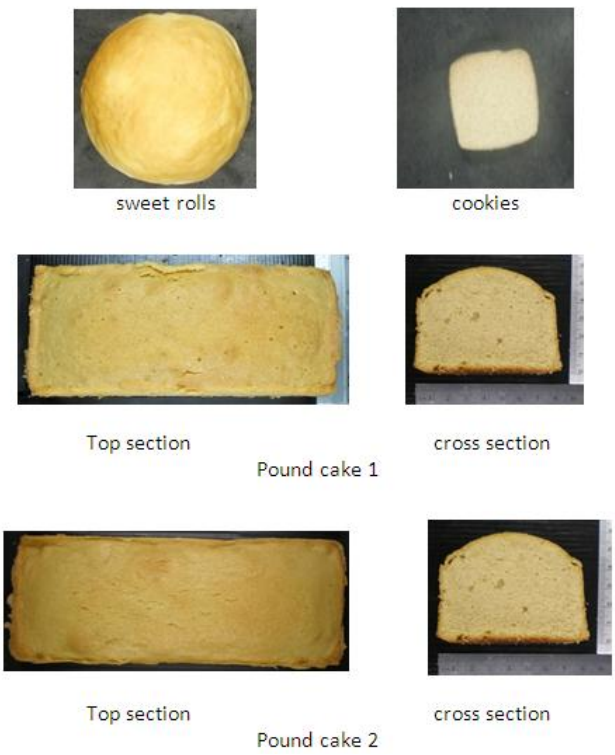


Figure 6. Product Based on Sorghum Flour

Conclusions

There are three types grain sorghum based on tannin content, respectively type I (non-pigmented testa), type II (low-pigmented testa) and type III (pigmented testa and pericarp).Decortication produces sorghum-rice, which is more palatable and acceptable. Sorghum-rice and flour are versatile food materials**.** Sorghum flour is free of gluten and almost taste and scent free with a color ranging from creamy to pinkish or reddish brown. Attempts have been done to manufacture food based on sorghum-rice respectively 'kerupuk', 'keripik', 'opak', 'rengginang' (rice-crispies), puffed sorghum, snacks based on puffed sorghum and flour respectively bread, sweet rolls, cake, noodles and various cookies.

Acknowledgment

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Application of Genomics Approaches to Unravel The Functional Biodiversity of Farm Animals

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Abstract

There are major challenges for animal production due to the global increase of population size and changes of social structure and wealth as well as climatic changes. There is a strong demand for resource efficiency of animal production and for farm animals with a high productive adaptability corresponding to the diversification of production and markets. In order make use of the existing genetic potential of a farm animal species their biodiversity must be determined. Consequently the identification of genetic resource is an important task and a prerequisite for conservation measures and/or utilization of the genetic variety. Since the emergence of DNA markers they are used as potential tools to estimate genetic distances and distinctness of breeds, lines, or local populations. The characterization of the genetic make-up together with records of phenotypic traits and breeding history facilitates the identification of genetic resources. Microsatellite markers are versatile tools to estimate genetic distance and are frequently used. However the application of DNA markers and the estimation of genetic distances do not provide a link to functionally relevant genetic variation. Currently SNP-markers, in particular the SNP-chip technology and new whole genome sequencing techniques allow a comprehensive analysis of the biodiversity. We aim to analyze the functional biodiversity of farm animals, i.e. the variation at different levels of the genotype-phenotype-map that is associated with functional consequences and reflects different physiological strategies that emerge during domestication and breeding of farm animals in order to cope with changing conditions. The manifestation of the genetic potential at the phenotypic level works via transcripts, proteins, metabolites, and other intermediate and molecular phenotypes. Therefore we integrate holistic approaches to characterize the genome, the transcriptome and in-depth phenotyping covering various structural and functional characteristics. The integration of analyses of several levels of the genotype-phenotype-map enables detecting causal relationships between genomic and phenotypic variation.This will improve our understanding of functional biodiversity and productive adaptability evolved in farm animal species. Finally this integrated approach of comprehensive determination of the genetic make-up and in-depth-phenotyping applying `omics´-techniques will allow obtaining DNA-markers and causal genes for selection in breeding routine and targeted conservation measures.

Ecological Rain-Fed Agriculture in Semiarid Loess Plateau of Northwest China

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Abstract

Rain-harvesting agriculture has been extensively expanded in north-west China particularly in the rain-fed agricultural areas of Loess Plateau since the end of 1980s. Currently, it is bringing about a revolutionary change to local social and economic and ecological developments, especially supplying enough food and clothing to afford 34.2 million people. This paper presents the developmental processes in traditional livelihood and current economy as a product of RHA impulse over last decades. The RHA plays a positive role to balance social-economical growth and natural ecosystem sustainability. Local GDP increased by 129%, average grain output per hectare increased by 63.1% while field soil erosion decreased by 32%, average soil moisture increased by 12.6% from 1989 to 2009. Importantly, the use efficiency of natural rainfall was improved from 4.5% to 25.8% and soil organic carbon from 0.23‰ to 0.31‰ simultaneously. The RHA is also found to change residents' lifestyle and livelihood pattern to meet the challenge of global climate change. We therefore propose an integrated model/concept of rain-harvesting ecological agriculture (RHEA), which incorporates the RHA theory into the coupled human & natural system (CHANS). The solution, evolution and popularization of RHEA in N-W China provide a novel understanding on the development and initiatives of dryland agriculture in semiarid rain-fed agricultural areas in the next 100 years.

Keywords: rain-harvesting agriculture, food security, field productivity, sustainability.

Biodiversity and Variety Improvement of Crop Plant

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Abstract

Crop diversity is the difference of cultivated plants used for producing food, fodder, fibre, fuel, pharmaceuticals, etc. Nearly 2,500 plant species have been cultivated worldwide and about 120-130 of them have been considered as the commercial crops. Among these, only 30 cropsprovide 95% of human food and just four of them, i.e. rice, wheat, maize and potatoes provide 60% food for the mankind. Crop plants evolved from the wild species through domestication bringing about genetic, physiological, morphological, and biochemical changes which render the plant differs from its wild form. Nowadays, a wide range of genetic and phenotypic diversity, called plant genetic resources (PGR), exists in the crop species. PGR provides the basic genetic material for improving crop varieties. Knowledge on the collection, documentation, characterization, evaluation and conservation of PGR are important to ensure that effort to save PGR from becoming extinct and make it always available to plant breeders are continued and enhanced. Variety improvement is a process of changing the genetics of plants using plant breeding technique in order to produce plants with characteristics desired by man. Techniques of plant breeding including conventional technique of selection and cross breeding, and the modern techniques of controlled pollination, hybridization, embryo rescue, recombinant DNA, doubling chromosome set/ number will be described in this paper.

Keywords:crop improvement, diversity, plant genetic resources

Introduction

Biodiversity or biological diversity refers to the variability among living organisms (plants, animals, fungi, microbes, lichens, etc) and the ecological systems in which the organisms live. Biodiversity involves ecosystem, species and genetic diversity.Ecosystem diversity refers to the differences of life forms in a given territory or area and the ecological processes that make them function. Species diversity encompasses the variety of different species within a geographical area. Genetic diversity denotes the variation of genes or the functional units of heredity within any species.

Variety improvement of a crop is an effort to improve the characteristics through reconstructing the genetic composition of the crop, so that it became more desirable agronomically and economically. Improvements can include things like better disease resistance, drought tolerance, fruit size and rapid growth comparing to the previous one. Plant genetic diversity is the only materials useful for variety improvement including the development of new cultivars.

The objective of this paper is to explain the origin, role and assessment of crop genetic diversity, and to share several points on the techniques of crop variety improvement for sustaining agricultural production.

Crop Diversity

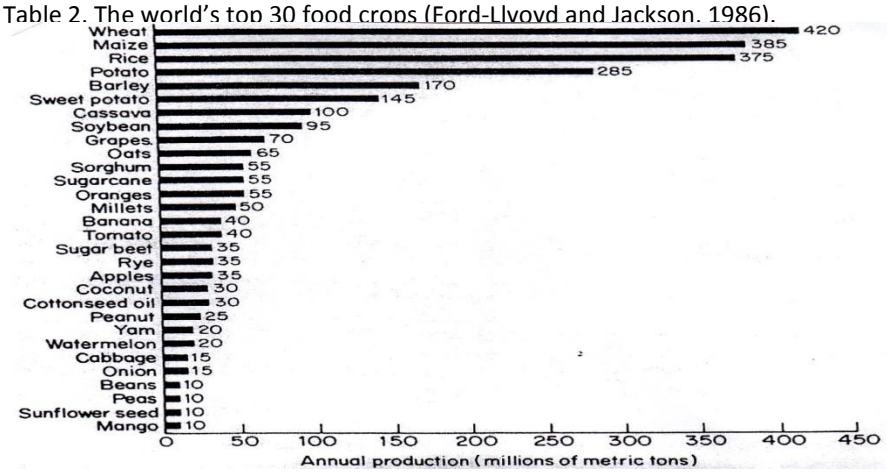
Crop diversity is the difference of cultivated plants used for producing food, fodder, fibre, fuel, pharmaceuticals, etc in any particular area or region. The example of crop diversity is an orchard garden where a large number (40 – 50) of different crop species could be present in one or two hectares of farmer's land (Table 1) (Hawkes, 1983).

Table 1: Crop diversity with different species of economic important in an orchard garden in the Southeast Asia.

Species	Common name
Root and tuber herbs	
Colocasia esculenta	Taro
Impomoea batatas	Sweet potato
Curcuma xanthorrhiza	Temu lawak
Zingiber officinale	Ginger
Amorphophallus campanulatus	Suweg
Maranta arundinacea	Arrowroot
Seed-sown herbs	
Solanum melongena	Eggplant
Cucurbita moschata	Squash
Amaranthus (tricolor ?)	Bayam
Mucuna pruriens	Cowage
Vigna sinensis	Cow pea
Hibiscus cannabinus	Java jute
Crotalaria anagyroides	Orok orok
Capsicum (annuum ?)	Chili pepper
Ocimum basilicum	Selasih
Climbers	
Dioscorea alata	Winged yam
Dioscorea bulbifera	Potato yam
Dioscorea pentaphylla	Ubi pasir
Dioscorea hispida	Asiatic bitter yan
Momordica charantia	Bitter gourd
Shrubs and small trees	
Musa x sapientium	Banana
Musa acuminata (seeded)	Banana
Ipomoea fistulosa	("For hedging")
Carica papaya	Pawpaw
Panax fruticosum	Kadongdongan
Jatropha curcas	Purging nut
Taxotrophis macrophylla	–
Gigantochloa atter, forma nigra	Black bamboo

Source: Hawkes, 1983

Table 2. The world's top 30 food crops (Ford-Llyoyd and Jackson, 1986).



Nearly 2,500 plant species have been cultivated worldwide for various reasons. This number includes about 850 food species which is covering 370 fruits, 280 vegetables, 60 edible flowers, 60 seeds/nuts/grains and 80 root and tuber crops (PROSEA, 1993-1999; Malik and Singh, 2006). Among these only 230 species are widely cultivated in which about 120-130 species are considered as the major or commercial crops. However, only 30 crops (Table 2) provide 95% of human food energy needs and just four of them, i.e. rice, wheat, maize and potatoes provide 60% food for the mankind (Ford-Llyord and Jackson, 1986).

Genetic Diversity

Although the number of major crops is significantly small but the genetic and phenotypic diversity within a crop are very great. For example, the number of distinct varieties of rice (*Oryzasativa*)worldwide is estimated more than 40,000 varieties. Also, the number of land races variety of potato (*Solanum tuberosum*) in Andes, South America was more than 175 varieties. Similarly, a great genetic diversity was also found in other major crops such as wheat, barley, maize, sorghum, millet, etc. (Harlan, 1975; Hawkes, 1983; Ford-Llyord and Jackson, 1986; Hasan, *et al,* 2008; Hasan & Abdullah, 1997).

Domestication Increases Crop Variability

The cultivated variety of a plant has evolved from the wild species through a process of domestication (Lester and Hasan, 1991; Simmonds, 1984; Hawkes, 1983). Since the beginning of agriculture about 10,000 years ago wild plants from natural population have been taken into cultivation by man (Harlan, 1975). During the course of cultivation, man consciously and unconsciously selects plants with useful characters and bringing about genetic, physiological, morphological and biochemical changes which render the plant differs from its wild form (Hawkes, 1983). Subsequent selection pressure occurred continually on the crops through the differentiation-hybridization cycle. In this cycle, cultivars were separated for a while by gene flow barrier due to geographical, ecological or morphological differences that allow crop establishes new sub-populations or cultivars. Soon or later, crossing between two cultivars results in a release of great variability within the crop. Selection pressure provided by different culturing practice, soil fertility, irrigation, etc accelerated the increase and distribution of variation in a crop (Ford-Llyord and Jackson, 1986).

Centre of Genetic Diversity

Genetic diversity of our major food crops mostly occurred in one of the eight mega-diversity of the world (Ford-Llyord and Jackson, 1986) (Table 3). It is the place or region where the crop shows the greatest variability ranging from new cultivars to landraces, weedy and wild relative forms. These regions were considered as the areas in which the domesticated plants have taken place and in some cases, were also considered the places of origin of the crop (Hawkes, 1983).

Plant Genetic Resources (PGR)

Now, a range of genetic and phenotypic diversity, called plant genetic resources (PGR) existed in the cultivated species ranging from the wild to fully cultivated state. Such diversity is an important plant genetic resources that provides the basic genetic material for improving crop varieties. PGR or germplasm is a living tissue from which the new plant can be grown. PGR is grouped into; i, primitive cultivars or landraces, ii, obsolete cultivars, iii,

modern or advanced or elite cultivars, iv, breeding lines, v, genetic stocks, vi, weedy types and vii, wild relatives (Hawkes, 1983).

Table 3. List of the world important crop in its centre of diversity and possible place of origin (Ford-Llyoyd and Jackson, 1986).

1. THE CHINESE CENTRE
Avena nuda, Naked oat (secondary centre of origin)
Glycine max, Soybean
Phaseolus angularis, Adzuki bean
Phaseolus vulgaris, Bean (recessive form; secondary centre)
Phyllostachys spp., Small bamboos
Brassica juncea, Leaf mustard (secondary centre of origin)
Prunus armeniaca, Apricot
Prunus persica, Peach
Citrus sinensis, Orange
Sesamum indicum, Sesame (endemic group of dwarf varieties; secondary centre)
Camellia (Thea) sinensis, China tea

2. THE INDIAN CENTRE
Oryza sativa, Rice
Eleusine coracana, Finger millet
Cicer arietinum, Chickpea
Phaseolus aconitifolius, Math bean
Phaseolus calcaratus, Rice bean
Dolichos biflorus, Horse gram
Vigna sinensis, Asparagus bean
Solanum melongena, Egg plant
Raphanus caudatus, Rat's tail radish
Colocasia antiquorum, Taro yam
Cucumis sativus, Cucumber
Gossypium arboreum, Tree cotton, 2x
Corchorus olitorius, Jute
Piper nigrum, Pepper
Indigofera tinctoria, Indigo

2a. THE INDO-MALAYAN CENTRE
Dioscorea spp., Yam
Citrus maxima, Pomelo
Musa spp., Banana
Cocos nucifera, Coconut

3. THE CENTRAL ASIATIC CENTRE
Triticum aestivum, Bread wheat
Triticum compactum, Club wheat
T. sphaerococcum, Shot wheat
Secale cereale, Rye (secondary centre)
Pisum sativum, Pea
Lens culinaris, Lentil
Cicer arietinum, Chickpea
Sesamum indicum, Sesame (one of the centres of origin)
Linum usitatissimum, Flax (one of the centres of origin)
Carthamus tinctorius, Safflower (one of the centres of origin)
Daucus carota, Carrot (basic centre of Asiatic varieties)
Raphanus sativus, Radish (one of the centres of origin)
Pyrus communis, Pear
Malus pumila, Apple
Juglans regia, Walnut

4. THE NEAR EASTERN CENTRE
Triticum monococcum, Einkorn wheat
Triticum durum, Durum wheat
Triticum turgidum, Poulard wheat
Triticum aestivum, Bread wheat (endemic awnless group; one of the centres of origin)
Hordeum vulgare, Endemic groups of cultivated two-rowed barleys
Secale cereale, Rye
Avena byzantina, Red oat
Cicer arietinum, Chickpea (secondary centre)
Lens culinaris, Lentil (a large endemic group of varieties)
Pisum sativum, Pea (a large endemic group; secondary centre)
Medicago sativa, Blue alfalfa
Sesamum indicum, Sesame (a separate geographic group)
Linum usitatissimum, Flax (many endemic varieties)
Cucumis melo, Melon
Prunus amygdalus, Almond
Ficus carica, Fig
Punica granatum, Pomegranate
Vitis vinifera, Grape

5. THE MEDITERRANEAN CENTRE
Triticum durum, Durum wheat
Avena strigosa, Hulled oats
Vicia faba, Broad bean
Brassica oleracea, Cabbage
Olea europea, Olive
Lactuca sativa, Lettuce

6. THE ABYSSINIAN CENTRE
Triticum durum, Durum wheat (an amazing wealth of forms)
Triticum turgidum, Poulard wheat (an exceptional wealth of forms)
Triticum dicoccum, Emmer
Hordeum vulgare, Barley (an exceptional diversity of forms)
Cicer arietinum, Chickpea (a centre)
Lens culinaris, Lentil (a centre)
Eragrostis tef, Teff
Eleusine coracana, Finger millet
Pisum sativum, Pea (one of the centres)
Linum usitatissimum, Flax (a centre)
Sesamum indicum, Sesame (basic centre)
Ricinus communis, Castor bean (a centre)
Coffea arabica, Coffee

7. THE SOUTH MEXICAN AND CENTRAL AMERICAN CENTRE
Zea mays, Corn
Phaseolus vulgaris, Common bean
Capsicum annuum, Pepper
Gossypium hirsutum, Upland cotton
Agave sisalana, Sisal hemp
Cucurbita spp. Squash, Pumpkin, Gourd

8. SOUTH AMERICAN (PERUVIAN-ECUADOREAN-BOLIVIAN) CENTRE
Ipomoea batatas, Sweet potato
Solanum tuberosum, Potato
Phaseolus lunatus, Lima bean
Lycopersicon esculentum, Tomato
Gossypium barbadense, Sea island cotton (4x)
Carica papaya, Papaya
Nicotiana tabacum, Tobacco

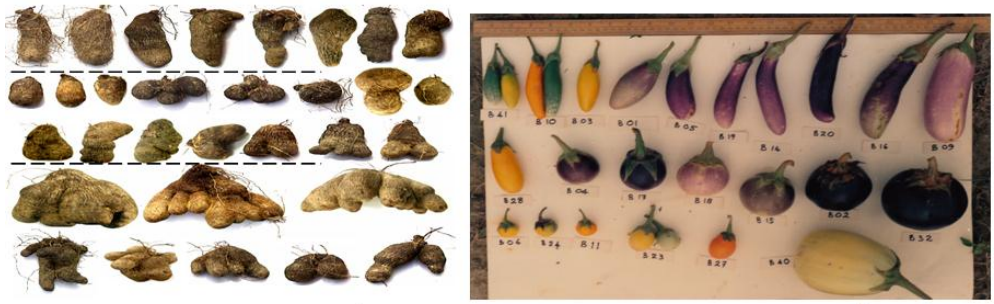
8a. THE CHILOE CENTRE
Solanum tuberosum, Potato

8b. BRAZILIAN-PARAGUAYAN CENTRE
Manihot esculenta, Manioc
Arachis hypogaea, Peanut
Theobroma cacao, Cacao (secondary centre)
Hevea brasiliensis, Rubber tree

Erosion of PGR

The existing of the present day plant genetic resources is seriously endangered. In nature, PGR has been reduced due to the industrial development, deforestation, climatic changes, changing in agricultural practices, disease and natural disasters. The diversity of local and well-adapted landraces has long been replaced by modern cultivars, particularly in

monoculture system where a single high-yield variety has been used, such as in rice and oil palm industries.



a b

Figure 1. Diversity of tuber morphology (size, shape and branching patterns) in traditional cultivars or land race of yam (*Dioscorea alata*) (a) and eggplant (*Solanum melongena*) (b) found in Malaysia (Hasan, *et al.,* 2008; Hasan & Abdullah, 1997)

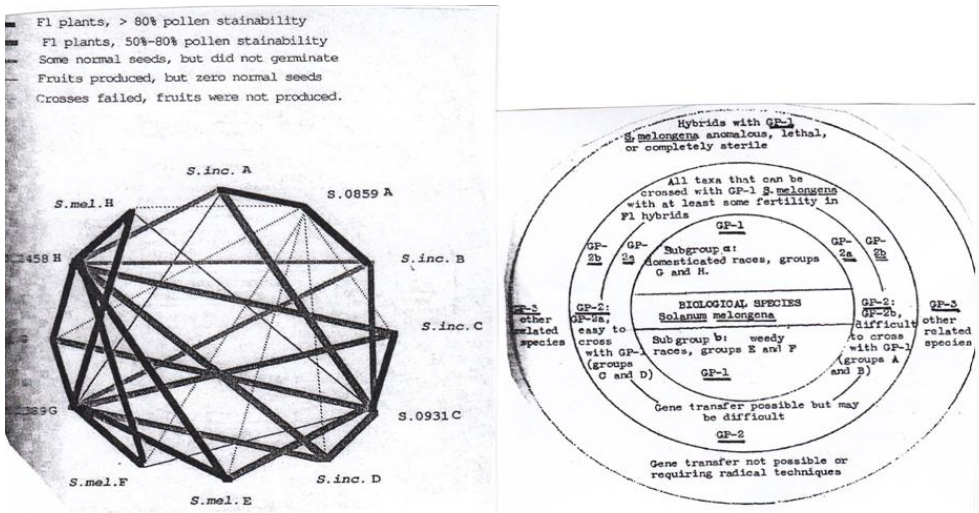


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Figure 2. **a.** Crossing polygon showing diversity in cross-ability relationship between cultivated *S. melongena* weedy form (E), landraces (F), obsolete cultivar (G) and elite cultivar/ black beauty (H), and its putative wild relatives *S. incanum*, groups A, B, C and D (Hasan, 1989).

b. Schematic diagram of primary gene pool (GP1), secondary gene pool (GP2) and tertiary gene pool (GP3) in eggplant genetic resources (Hasan and Lester, 1992).

Management of PGR

Proper management including collection, characterization, evaluation and conservation of PGR is needed to ensure that the genetic resources of a species are saved from becoming extinct and are always available to the breeders and other users. Collection involves

gathering samples of a species from populations in the field or naturalhabitats for conservation and subsequent use. Characterization refers to recording of characters which are highly heritable, easily identified (usually qualitative), and are expressed in all environments. Evaluation refers to documentation of additional characters (often quantitative traits relating to yield and symptom of diseases) which are thought desirable by the users or breeders of the crop (Hasan, *et al*., 2008; Hasan & Abdullah, 1997).

Conservation involves maintaining PGR whether in the natural habitats where they occur (*in-situ* conservation), or outside the native habitat (*ex-situ* conservation) such as seed storage in gene-bank which capable to maintain the integrity large number of the germplasms over prolonged periods of time (Figure 3).*In vitro* conservation through tissue culture techniques is also considered as an *ex situ* conservation (Figure 4a). *In vitro* conservation can maintain the materials in a pathogen-free environment and are not subjected to environmental disturbances. They fall under two categories: (i) slow growth procedures (Figure 4b) and (ii) cryopreservation (Figure 5).

Figure 3. Seed storage in National Genebank at Tsukuba, Japan (Visited by the author in 1994)

a b

Figure 4. a. *In-vitro s*torage of pineapple germplasm in standard tissue culture needed sub-culturing at every three months of culturing (Hasan and Nursuraya, 2007). b. Reduced growth storage of yam (*Dioscorea alata*) germplasm in medium culture supplemented with osmotic agent of mannitol (Hasan and Lasim, 1996)

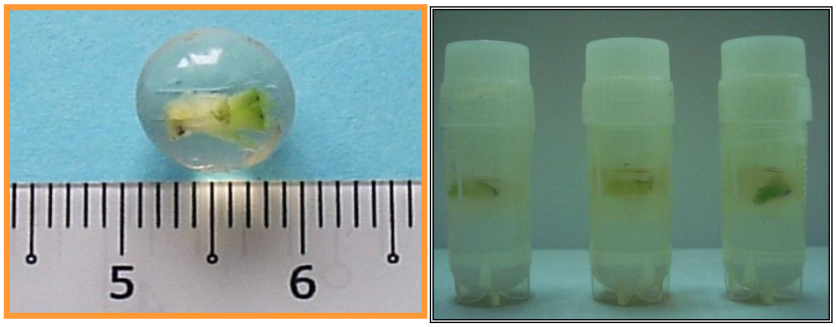

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Figure 5. Shoot-tip tissue culture encapsulated in alginate-bead (a) and kept in cryo-tube (b) for cryopreservation test in liquid nitrogen at -160° C (Hasan and Takagi, 1995)

Estimating Genetic Diversity

Estimating genetic diversity is essential for effective use of genetic resources in variety improvement. The level of genetic variability would determine the PGR either having a wide or narrow genetic base. Presently, beside agro-morphological traits, bio-chemical such as protein and molecular DNA (RAPD, RFLP, ISSR, etc) are also used as the markers to assess the extent of genetic variability present in the PGR collection (Figs. 6 and 7) (Hasan, *et al.* 2010, 2009, 2008, 2006; Hasan & Isa, 1998; Hasan & Abdullah, 1997).

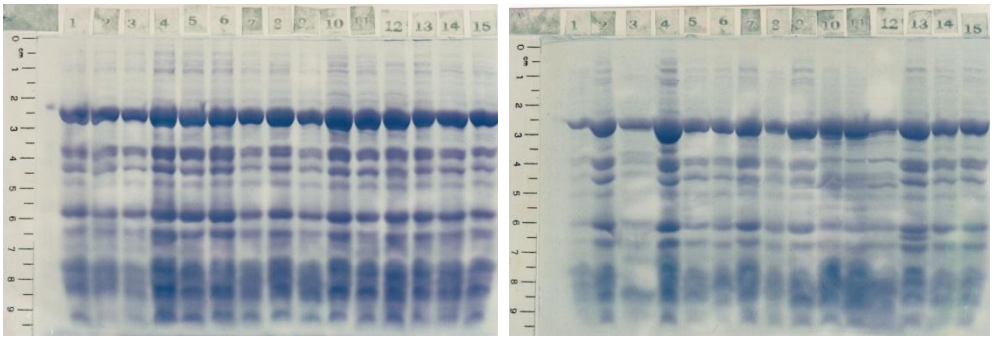

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Figure 6. Diversity of seed protein profile (banding pattern) in eggplant (*Solanum melongena*) genetic resources collected in Malaysia, where it was rather uniform in population of advanced cultivar (a) and quite diverse in old or obsolete cultivar (b) (Hasan and Isa, 1998)

Assessment of diversity is made by comparing the differences of the markers between individuals or populations using statistical analysis (Dunn and Everitt, 1982). Comparison of mean and variances among the populations are the simple way to estimate the diversity (Hasan and Abdullah, 1997). Calculating the similarity coefficient is another way to assess the PGR diversity (Table 4) (Hasan, *et al,* 2010 and 2009). Recently, multivariate analysis of principal component analysis (PCA) and cluster analysis (CA) using SPSS computer program is commonly used in diversity study. In this analysis, similarity or dissimilarity index of

individuals is used to separate the individual on the PCA and also to cluster the individuals in order to construct the hierarchical relationship among the individuals (Figs. 8a & 8b). The PCA is often used to find out the most variable characters in the population, while cluster analysis (CA) is used to elucidate the relationship among individual in the PGR collection (Hasan, *et al.* 2010, 2009, 2008 & 2006)

OPG-02 primer

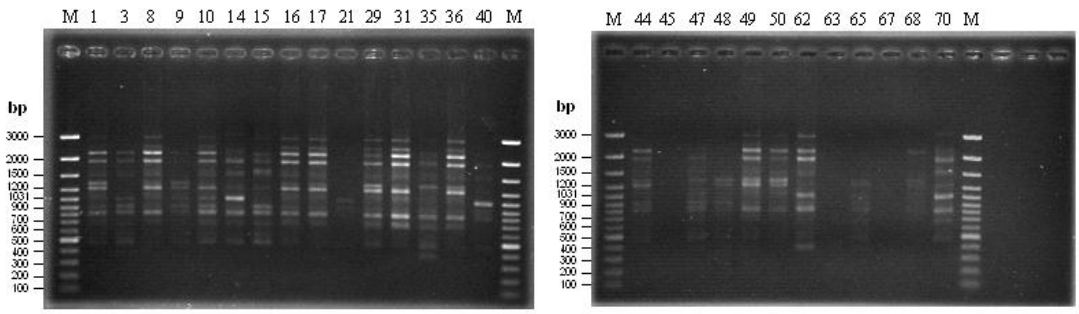


Figure 7. RAPD banding patterns of twenty seven (27) *D. alata* accessions based on a primer OPG-02. Each lane represents an accession marked above. Lane M is a marker of 100bp DNA ladder plus

Table 4. The similarity coefficient among twenty seven (27) accessions of yam (*Dioscorea* alata) genetic resources in Peninsular Malaysia after analyzed data from RAPD marker using Jaccard's similarity coefficient (Hasan *et al.,* 2006*)*

Acc. No.	1	3	8	9	10	14	15	16	17	21	29	31	35	36	40	44	45	47	48	49	50	62	63	65	66	68	70
70	0.49	0.63	0.52	0.51	0.49	0.75	0.61	0.47	0.53	0.61	0.56	0.51	0.67	0.51	0.75	0.49	0.49	0.67	0.53	0.51	0.53	0.53	0.54	0.58	0.51	0.54	1.00
1	1.00																										
3	0.61	1.00																									
8	0.89	0.61	1.00																								
9	0.87	0.63	0.84	1.00																							
10	0.79	0.72	0.86	0.81	1.00																						
14	0.56	0.63	0.56	0.61	0.60	1.00																					
15	0.56	0.74	0.53	0.54	0.60	0.58	1.00																				
16	0.87	0.63	0.91	0.82	0.88	0.58	0.54	1.00																			
17	0.86	0.61	0.89	0.81	0.82	0.53	0.56	0.95	1.00																		
21	0.60	0.63	0.60	0.68	0.59	0.75	0.61	0.58	0.53	1.00																	
29	0.79	0.61	0.89	0.74	0.82	0.60	0.49	0.91	0.86	0.56	1.00																
31	0.88	0.60	0.95	0.86	0.84	0.58	0.54	0.89	0.88	0.58	0.88	1.00															
35	0.68	0.61	0.68	0.67	0.65	0.70	0.67	0.70	0.65	0.63	0.75	0.70	1.00														
36	0.84	0.63	0.95	0.79	0.88	0.54	0.51	0.89	0.84	0.58	0.91	0.93	0.70	1.00													
40	0.56	0.67	0.60	0.68	0.56	0.79	0.61	0.51	0.52	0.82	0.52	0.58	0.67	0.54	1.00												
44	0.79	0.65	0.82	0.77	0.75	0.49	0.56	0.77	0.75	0.56	0.75	0.77	0.61	0.77	0.56	1.00											
45	0.79	0.64	0.86	0.77	0.86	0.49	0.56	0.84	0.82	0.56	0.82	0.88	0.61	0.84	0.53	0.86	1.00										
47	0.61	0.65	0.61	0.67	0.54	0.60	0.63	0.60	0.58	0.60	0.61	0.60	0.72	0.56	0.67	0.65	0.58	1.00									
48	0.82	0.54	0.79	0.84	0.68	0.60	0.49	0.81	0.80	0.60	0.79	0.77	0.72	0.74	0.60	0.79	0.72	0.72	1.00								
49	0.84	0.53	0.84	0.75	0.74	0.61	0.47	0.86	0.84	0.47	0.84	0.82	0.67	0.79	0.51	0.77	0.77	0.63	0.84	1.00							
50	0.82	0.61	0.79	0.74	0.68	0.53	0.53	0.74	0.72	0.56	0.75	0.74	0.65	0.74	0.60	0.75	0.68	0.72	0.75	0.77	1.00						
62	0.58	0.86	0.65	0.60	0.68	0.60	0.63	0.63	0.58	0.56	0.61	0.63	0.58	0.67	0.56	0.61	0.61	0.58	0.54	0.60	0.58	1.00					
63	0.56	0.60	0.60	0.51	0.60	0.61	0.61	0.58	0.56	0.54	0.63	0.61	0.67	0.61	0.54	0.63	0.67	0.60	0.56	0.61	0.53	0.60	1.00				
65	0.63	0.60	0.67	0.65	0.60	0.72	0.51	0.61	0.56	0.72	0.63	0.65	0.63	0.65	0.72	0.63	0.63	0.60	0.60	0.61	0.60	0.60	0.65	1.00			
67	0.74	0.67	0.77	0.82	0.74	0.58	0.51	0.75	0.70	0.65	0.74	0.75	0.67	0.75	0.65	0.81	0.77	0.74	0.84	0.75	0.74	0.67	0.54	0.72	1.00		
68	0.67	0.63	0.67	0.68	0.63	0.51	0.54	0.61	0.60	0.61	0.63	0.65	0.56	0.61	0.65	0.70	0.63	0.77	0.67	0.61	0.84	0.56	0.54	0.58	0.72	1.00	
70	0.49	0.63	0.52	0.51	0.49	0.75	0.61	0.47	0.53	0.61	0.56	0.51	0.67	0.51	0.75	0.49	0.49	0.67	0.53	0.51	0.53	0.53	0.54	0.58	0.51	0.54	1.00

Gene Pools Concept

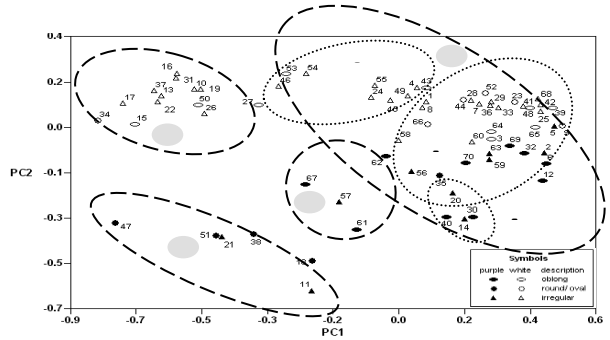
The ease of crossing among the groups of PGR also varies (Figure 2a). This variation is used to classify PGR according to the gene pools concept (Figure 2b) (Hasan & Lester, 1992) in order to assist breeders to select particular genes or variant for use in crop improvement. In this concept PGR is classified into four gene pools (GP) (Ford-Llyord and Jackson, 1986; Harlan, 1975).

GP1: The crop itself and wild relatives that are easily inter-crossed and produce fertile progeny.

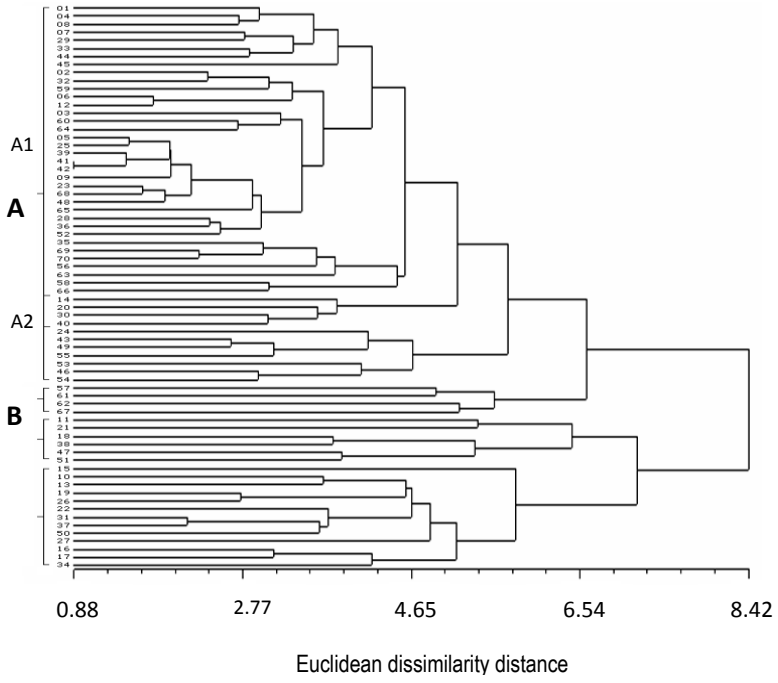
GP2: Species that cross to GP1 but with difficulty and produce progeny with reduced viability

GP3: Species that cross to GP3 only using advanced techniques (embryo rescue, tissue culture) and produce progeny with low viability

GP4: Other species that do not cross to GP1 at all. Molecular genetics and gene cloning techniques must be used to transfer these genes into species in GP1.



a



b

Figure 8. a. Two-dimensional plot of the first two principal components (PC-1 and PC-2). Accessions that are encircled by the indented lines and marked as groups A, B, C and D in the cluster analysis (Figure 9b) (Hasan, *et al.,* 2008).

b. The hierarchical dendogram showing the relationships among seventy accessions of *D. alata* collected in Malaysia (Hasan, *et al.,* 2008).

a b

Figure 9. Artificial cross-pollination in eggplant (a), and growing progeny of eggplant for selection (b). The work had been conducted by the author in 1990 – 1993 at UPM, Serdang, Malaysia (Hasan, 2005; Hasan & Abdullah, 1997).

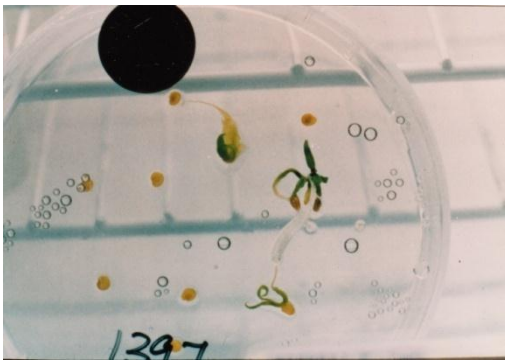


Figure 10. Germination of hybrid seed or embryo rescue in medium of tissue culture

Variety Improvement

Variety improvement is a process of plant breeding, that is a technique of changing the genetics of plants in order to produce plants with characteristics desired by man. Basically, the process started with the searching and identification of desirable trait from the gene pools and this greatly depends on the genebank collection. Then followed by creation of variation that normally done through the hybridization process (Figure 10a). The best plants are selected from the derived progeny and spread into a large population (Figure 10b). These plants are allowed to cross-pollinate, and seed is harvested from them. The process is repeated with many following generations, until the desired characteristics are exhibited on the general population. The new variety is then released and dispersed to the public.

Aspects of Variety Improvement

The main aspects of crop variety improvement are :

- *Improved yield:* To increase and stable crop yield production.
- *Improved quality:* To produce better size, colour, shape, taste and nutritive value in food.
- *Enhance biotic and abiotic resistance:*To increase resistance against pest and disease and environmental stress.
- *Shorter in maturity duration*: To reduce duration and synchronous maturity. Early maturity can make the crop double or triple cropping system and reduce the cost of production.
- *Modifying agronomic traits:*To change the agronomic characteristics of crops such as plant height, tillering, branching, determinate growth, etc.
- *Photo and Thermo insensitivity:* To develop varieties insensitive to light and temperature to permit crops grown in areas, which might not have been ideally suited for their growth (Example Strawberry in Cameron Highland)
- *Wider Adaptability:* To develop varieties with wider adaptability and thus help stabilize crop production under unfavourable environmental conditions (Hawkes, 1983).

Methods of Plant Breeding

Plant breeding can be accomplished through many different techniques ranging from simply selecting plants with desirable characteristics to more complex molecular techniques for improving crop variety (Allard, 1960). Any one technique adopted, however, is depending on the reproductive system of the crop and objectives of the improvement. Based on the reproductive system, the crops are divided into self-pollinated/ inbreeding and cross-pollinated/ outbreeding groups. The major different between the two groups is related to their influence on the genetic structure of the populations (Ford-Llyoyd and Jackson, 1986).

Conventional Plant Breeding

Conventional plant breeding is a breeding method practiced by the breeders since 1900 when the Mendel's laws of genes inheritance have been adopted as the scientific basis for plant breeding. As all traits of a plant are controlled by genes located on chromosomes, conventional plant breeding can be considered as the manipulation of the combination of chromosomes. In general, there are two main procedures in conventional breeding are used to manipulate chromosome combination for improving the crops.

1. Selection

Selection is the basic procedure in variety improvement. It generally involves first, selection large number (mass selection) of plants from the genetically variable original parent population. Second, the progeny from the individual selected plants are grown and the progenies are selected again and grown for over several generations. The final selected progeny are also tested for selection under different environmental condition. Thirdly, the new varieties are then compared to the existing commercial varieties in their yield performance and other aspects of agronomic importance. There are two selection methods are applied in self-pollinated crop, pure-line selection and mass selection. Also two

selection methods are applied in cross-pollinated crop, mass selection and recurrent selection (Allard, 1960).

2. Hybridization/ Cross-pollination

Hybridization is an effort to bring together the desired traits found in different plant lines into one plant line via controlled or artificial cross-pollination (Figure 9a) (Hasan, 2005). The first step is to generate homozygous inbred lines. This is normally done by controlled self-pollinating (Figure 9a) where pollen from male flowers is pollinated on the female flowers from the same plants. Once a pure line is generated, it is then out-crossed with another inbred line. The segregating F2 progeny is then selected for the desired traits (Figure 9b). If a trait from a wild relative of a crop species, such as resistance against a disease, is to be brought into the genome of the crop, the undesired traits (like low yield, bad taste, low nutritional value) are then removed by repeated back-crossing with the crop parent.

Modern Technique of Hybridization

In general, there are three types of hybridization: inter-varietal (narrow crosses), inter-specific and inter-generic hybrids (wide crosses). Beyond this biological boundary, hybridization cannot be accomplished due to sexual incompatibility (incongruity), which limits the possibilities of introducing desired traits into crop (Hasan, 1990; Hasan & Lester, 1990). Nowadays a number of techniques were developed to overcome these barriers and that allowed plant breeders to hybridize distantly related species.

Bridge Cross: When a direct cross between two species is not possible, an intermediate crossing with a third species, which is compatible with both species, is used to break down the crossing barriers.

Pollination using sub-optimal age of stigma: In order to overcome incompatibility barriers posed by the stigma or style, mature pollen is placed on a stigma that is immature to prevent presence of active factors that inhibit pollen tube growth.

Pollination with chemical application; Plant growth regulator (hormones) such as auxin, cytikinin and gibberellins are applied to flowers to promote pollination.

Pollen mixture: A mixture of incompatible pollen with temperature pre-treated compatible pollen is used in pollination. The compatible pollen is able to germinate and penetrate the style, thereby clearing the way for the incompatible pollen.

Style treatment: Heat treatment of stigma, style or the whole flower before pollination. Heat treatment can reduce the incompatible reaction of the stigma surface, the style or the whole flower before pollination.

In-vitro Methods: This technique can be considered as a high level of sophisticated tools applied in plant breeding. The technique is conducted under the aseptic condition. Some of the methods have been used as a routine tool and some others are still at the experimental stage in plant breeding.

a. Pollination after manipulation of style: Removal of the stigma or shortening of the style to remove the factors that inhibits pollen tube growth.

b. Graft style method: a style with pollen germinated on a compatible stigma is cut and put on an ovary of the other incompatible parent where the stigma has previously been removed.
c. *In vitro* pollination: Pollen is applied on the stigma under aseptic condition. The flower bud is removed from the plants and after dissection of flower parts, the bud is put on a growth medium. Fertilized ovules are then subsequently isolated and further cultured on fresh medium. Pollen and ovule can also be brought into direct contact for fertilization. Pollen can also be placed on the placenta of the ovary.
d. *In vitro* culture of excised ovaries: After pollination, the ovary is excised from the flower and cultured aseptically on a suitable medium.
e. Embryo rescue; If fertilization is possible between two species or genera, the hybrid embryo may abort before maturation due to incompatibility with the endosperm or surrounding maternal tissue of the ovary. If this does occur the embryo resulting from an interspecific or intergeneric cross can sometimes be rescued and cultured on media to produce a whole plant. Such a method is referred to as Embryo Rescue (Figure 10) (Hasan & Lester, 1990).
f. Induced mutation or mutation breeding: The mutation is induced by chemical mutagens like EMS and DMS, radiation (beta or gamma ray) and transposons to generate new variances with desirable traits and used to bred with other cultivars. The mutant hybrids are tested and further selected for desired traits. Because the great majority of mutants carry undesirable traits, this method has not been widely used in breeding programs.
g. Somaclonal variation: Plant breeders also generate genetic diversity within a species by exploiting a process of somaclonal variation, which occurs in plants produced from tissue culture, particularly plants derived from callus.
h. Somatic Hybrid Plants: Somatic hybrid plants are plants derived from the fusion of somatic cells. Cell fusion is done by making contact a number suspension cells after stripped of their cell walls, or referred as protoplasts. Methods such as particle bombardment, electroporation and polyethylenglycol permeabilisation are used in chloroplast fusion breeding. Since, protoplasts from distinctly unrelated species can be fused, the technique have been used to overcome sexual incompatibility of the cross species. The technique is uneconomic to practice, but contributes important knowledge in understanding somatic hybrid in plant.
i. Genetic Engineering: The incompatibility problem to transfer genes can be overcome by using the methods of genetic engineering or recombinant DNA technology, which in principle allow introducing valuable traits through carrier organism (other plants, bacteria, fungi, animals, viruses) into the genome of any plant. Recently, transgenic plants have been obtained using Agrobacterium-mediated DNA-transfer and direct DNA-transfer. When a desirable trait has been bred into a species, a number of crosses to the favored parent are made to arise the new plant similar to the favored parent as possible. The new plant is called transgenic plant or genetic modified organism (GMO).

3. Doubling the chromosomes set / Polyploidy/ Haploidization

Most plants contain two sets of chromosomes number and called diploid plant. Plants with three or more sets of chromosomes are also common and are referred to as polyploids. The increase of chromosomes sets can be artificially induced by applying a mitotic inhibitor, colchicine, which leads to a doubling of the chromosome number. The sterile hybrid can be retrieved and producing fertile progeny.

Haploid plants can be arisen in tissue culture from pollen (anther) culture or ovule (ovary) culture. The regenerated haploid plants are diploidized to obtain doubled haploids (homozygous) plants by using colchicine.

Conclusion

Regional centre of genebank should be established to conserve a vast amount of plant genetic resources present in this region. The PGR is extremely valuable genetic materials for the development of new variety and improving the existing one, particularly new variety with high resistant against biotic and abiotic stresses and high yield.

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SUSTAINABLE AGRICULTURE

Indicators of Agricultural Sustainability at the Regional Level a Case Study of Vidarbha

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Abstract

In this study, an attempt has been made to measure the Sustainability of Agriculture in Vidarbha region of Maharashtra state and to identify the factors responsible for sustainability. The sustainability is measured by developing Sustainability Index of Nagpur district with the help of various indicators of sustainability. The study was based on secondary data of area, production, productivity, population, agricultural population, weather, irrigation, area under high yielding varieties were collected from various Government publications of Maharashtra. The data for computation of indicators cost-benefit ratio for selected crops were adopted from the records of Agricultural Prices Cell (APC), Dr. Panjabrao Deshmukh Krishi Vidyapeeth, Akola. The data pertain to a period of 26 years i.e. from 1980-81 to 2005-06. The study was limited to the principal crops namely cotton, kharif jowar, soybean, tur, gram and sunflower cultivated in Nagpur district of Vidarbha.The result showed that the sustainability index developed through principal component analysis was seen declining over the years. The Sustainability index ranges from -0.308 to 0.350. Higher number of sustainability index represents higher sustainability whereas, lower number represents lower sustainability. This concludes that the Vidarbha agriculture is tending towards un-sustainability. The study also found that the productivities per hectare of cotton, tur, kharif jowar, sunflower and gram, gap in gross returns in cotton, kharif jowar and gram, cost-benefit ratio of cotton, kharif jowar and gram, parity index of kharif jowar and gram, availability of land per farmer, ratio of irrigated land to irrigable land, per capita production of foodgrains, ratio of agricultural population to total population, area under high yielding varieties and rainfall were the significant contributing variables for agricultural sustainability of the Vidarbha region.

Keywords: sustainability indicators, principal component analysis, sustainability index, Mann Whitney U Test

Introduction

Agriculture is a complex of processes taking place within biophysical, socio-economic and political constraints, which control the sustainability of the farming activities (Yunlong and Smit, 1994). The concept of sustainable agriculture combines characteristics such as long term maintenance of natural systems, optimal production with minimum input, adequate income per farming unit, fulfillment of basic food needs, and provision for the demands and necessities of rural families and communities (Brown *et al*. 1987). All definitions of sustainable agriculture promote environmental, economic and social harmony in an effort to attain the meaning of sustainability. The most relevant issue today is to design suitable technologies, as well as compatible strategies from the social, economic and ecological viewpoints that will bring about the necessary behavioral changes to achieve the objectives of sustainable agriculture.

Sustainability in relation to agriculture is becoming increasingly important as the linkages between the economy, society and the environment are more widely recognized (Vander Werf and Petit, 2002). Sustainability is a term with diverse interpretation; however, it is commonly applied to ecological or environmental, social and economic aspects of farming systems. Sustainable agriculture is essentially concerned with the ability of agro-ecosystems to remain productive in the longer term. In order to judge whether an agriculture system or farm is sustainable or not, an easy to use tool or method is required that can provide information understandable to practitioners such as farmers and to other stakeholders such as policy makers.

Sustainable agriculture is defined as a practice that meets current and long-term needs for food, fibre, and other related needs of society while maximizing net benefits through conservation of resources to maintain other ecosystem services and functions, and long-term human development. It emphasizes multidimensional (economic, environment and social) goals for addressing the problems of sustainable agricultural development.

Sustainability is a concept and cannot be measured directly. Appropriate indicators must be selected to determine levels and duration of sustainability (Zinck and Farshad, 1995). An indicator of sustainability is a variable that allows describing and monitoring the processed, states and tendencies of systems at the farm, regional, national or worldwide levels. It must be sensitive to temporal and spatial changes, predictable, measurable and interactive (Liverman *et al*. 1988). They possess multidimensional attributes–economic, environmental and social. Indicators can be meaningfully integrated into an aggregated index. They allow integrated assessments after taking into account all information provided by indicators.

Vidarbha was in the media spotlight for a spate of farmer suicides in recent years. Vidarbha accounts for 65 per cent of land under cotton cultivation in Maharshtra; and the state accounts for 36 per cent of the national area of cotton cultivation. But the productivity in the state is the lowest in the country: just above a quintal per hectare (Deshpande, 1997).

Repeated crop failures, inability to meet the rising cost of cultivation and indebtedness seem to create a situation that forces farmers to commit suicide. However, not all farmers facing these conditions commit suicide – it is only those who seem to have felt that they have exhausted all avenues of securing support have taken their lives (TISS, 2005).

The spate of suicides in the Vidarbha region, coupled with the declining share of agriculture in the Gross Domestic Product (GDP) and increasing burden on the agrarian system underlines the desperate plight of Vidarbha's farmer. These suicides point to a greater crisis in the agrarian system as a whole where, the suicide is a symptom of a greater malaise that threatens millions of farmers and the landless agricultural laborers in Vidarbha. This raises the issue of Sustainability in Vidarbha agriculture.

The objective of this study was aimed to investigate Sustainability of Agriculture in Vidarbha region with the help of various indicators and to identify the factors responsible for sustainability. The sustainability is measured by developing Sustainability Index of Vidarbha region with the help of various indicators of sustainability.

Materials and Methods

The study has been confined in the Vidarbha region of Maharashtra state. Indicators of sustainability of Vidarbha region have been constructed for the period 1980-81 to 2005-06

utilizing the data available through different sources. The study was limited to the principal crops namely cotton, kharif jowar, soybean, tur, gram and sunflower cultivated in Vidarbha region. Data pertaining to area, productivity, population, agricultural population, weather, irrigation, area under high yielding varieties were collected from various Government publications of Maharashtra state. The data for computation of indicators cost-benefit ratio for selected crops were adopted from the records of Agricultural Prices Cell (APC), Dr. Panjabrao Deshmukh Krishi Vidyapeeth, Akola.

The present investigation was aimed to study the Sustainability of Agriculture in Vidarbha region. Identification of meaningful indicators was an important task of the study. In this study several indicators pertaining to agro-diversity, agro-system efficiency, extent of the land resource base and food security were considered.

Several indicators of sustainability of Vidarbha region were classified as follows:

1) Agrodiversity
    i) Index of Surface Percentage of Crops
2) Agrosystem efficiency
    i) Crop yield
    ii) Gap in gross return
    iii) Cost-benefit ratio
    iv) Parity index
3) Land resource use base
    i) Availability of land per farmer
    ii) Irrigated land/ irrigable land
    iii) Cropping intensity
    iv) Area under High Yielding Varieties
4) Food Security
    i) Per capita production of foodgrains
    ii) Agriculture population / total population
    iii) Weather index

Analytical Tools

Development of Composite Sustainability Index

A set of data pertaining to 32 indicators was too large to comprehend and there were several approaches to represent them in a compact manner leading to development of composite index. Composite indices are aggregation of sets of variables for the purpose of meaningfully condensing large amounts of information. Various aggregation methods exist and the choice of an appropriate method depends on the purpose of the composite index as well as the nature of the subject being measured. The method used here was to perform a principle component analysis.

Principal Component Analysis

Principal component analysis (PCA), a popular multivariate technique, is mainly used to reduce the dimensionality of p multi-attributes to two or three dimensions. PCA summarizes the variation in a correlated multi-attribute to a set of a few uncorrelated components, each of which is a particular linear combination of the original variables. The extracted uncorrelated components are called principal components (PC) and are estimated

from the eigenvectors of the covariance or correlation matrix of the original variables. Therefore, the objective of PCA is to achieve parsimony and reduce dimensionality by extracting the smallest number components that account for most of the variation in the original multivariate data to summarize the data with little loss of information.

Principal component analysis can be performed either on the original variables, or their deviations from the means $x_j = X_j - X_j$ or the standardized variables (measured as the deviations of the X_j's from the means and subsequently divided by the standard deviation);

$$Z_j = x_j / S_{xj}$$

Principal components satisfy two conditions:

a. The principal components are uncorrelated (orthogonal) and

b. The first principal component P_1 absorbs and accounts for the maximum possible proportion of the total variation in the set of all X's, the second principal component absorbs the maximum of the remaining variation in the X's and so on.

In our study, a required number of components that explains at least 85 per cent of total variation were chosen for developing composite Index. SPSS, a standard software package was used for this purpose. A composite index was derived by taking sum of weighted PCA scores with eigen values as weights.

Classification of years on the basis of Sustainability Index

The degree of sustainability can serve as a basis for classifying years of low, moderate, sustainable and highly sustainable years which can help policy makers for developing suitable strategies to protect the farmers against preventable causalities.

For classification of years, the composite index scores developed through Principal Component Analysis was taken into consideration. The composite scores were correlated with the help of different indicators of sustainability studied above to classify the years and then final classification of years was done.

The composite indices were further used to classify years into different categories indicating varying levels of sustainability.

An year with composite score -

(1) falling below zero,
(2) falling in between 0 to 0.1,
(3) falling in between 0.1 to 0.3 and
(4) above 0.3

is classified as Not sustainable, Moderately sustainable, Sustainable and Highly sustainable respectively.

On the basis of the scores developed above, the classification of years was carried out. After classification of years, the means of Sustainable and Non-Sustainable years were compared with Non-Parametric Tests.

Mann Whitney U Test

The Mann Whitney U Test (also called the Mann-Whitney-Wilcoxon (MWW), Wilcoxon rank-sum test or Wilcoxon-Mann-Whitney test) is best known non-parametric significance test for assessing whether two samples of observations come from the same distribution. The null hypothesis is that the two samples are drawn from a single population, and therefore their probability distributions are equal. It requires the two samples to be independent, and the observations to be ordinal or continuous measurements, i.e. one can at least say, of any two observations, which is the greater. It was proposed initially by Wilcoxon (1945), for equal sample sizes, and extended to arbitrary sample sizes and in other ways by Mann Whitney (1947).

The basic steps in performing the Mann Whitney U test were as follows:

1. Arrange all the observations into a single ranked series.
2. Add up the ranks in sample 1. The sum of ranks in sample 2 follows by calculation, since the sum of all the ranks equals N(N+1)/2 where N is the total number of observations.
3. "U" is then given by:

$$U_1 = R_1 - \frac{n_1(n_1 + 1)}{2}$$

Where,

n_1 is the two sample size for sample 1
R_1 is the sum of the ranks in sample 1

$$U_2 = R_2 - \frac{n_2(n_2 + 1)}{2}$$

The smaller value of U_1 and U_2 is the one used when consulting significance tables. The sum of the two values is given by

$$U_{1+}U_2 = R_1 - \frac{n_1(n_1 + 1)}{2} + R_2 - \frac{n_2(n_2 + 1)}{2}$$

Knowing that $R_1 + R_2 = N(N=1)/2$ and $N = n_1 + n_2$, the sum is $U_1 + U_2 = n_1n_2$

The maximum value of U is the product of the sample sizes for the two samples. In such a case, the "other" U would be 0.

Results and Discussion

Triennium Average Performance of Sustainability Indicators

The performance of various Indicators of Sustainability in Vidarbha region during different segments of time periods from 1980-81 to 2005-06 is shown in Table 1. From the Table, it can be seen that the Index of Surface Percentage of Crops of Vidarbha region showed an

increase of 51.52 per cent from 1980-81 to 2005-06. It can be also seen from the table, that during the period 1980-81 to 2005-06, the yields of major crops in the Vidarbha region had observed a spectacular growth. The yield of sunflower has increased by 696.13 per cent. The yield of cotton has increased by 152.11 per cent from 1980-81 to 2005-06. The yield of gram, tur and kharif jowar had increased by 100.89 per cent, 59.46 per cent and 9.56 per cent respectively during the same period. Soybean was an introduced crop in the 1990s, the yield of which increased by 42.82 per cent from 1990-91 to 2005-06.

As far as gap in gross return of crops were concerned, the gap in gross return of cotton showed a highest increase of 653.55 per cent from 1980-81 to 2005-06. Similarly, the gap in gross return showed an increase of 368.06 per cent, 261.51 per cent and 102.60 per cent in gram, kharif jowar and tur respectively, during this period. Whereas, the gap in gross return showed an increase of 88.33 per cent from 1990-91 to 2005-06. The gap in gross return of sunflower declined by 51.64 per cent during the study period.

Regarding profitability of crops were concerned, the cost-benefit ratio of sunflower and soybean increased by 158.49 per cent and 15.65 per cent respectively, during the period 1980-81 to 2005-06. The cost-benefit ratio of kharif jowar, gram, cotton and tur decreased by 25.85 per cent, 18.75 per cent, 15.79 per cent and 10.29 per cent, respectively during the same period.

As far as input and output prices were concerned, the parity index of sunflower and soybean increased by 117.50 per cent and 17.56 per cent. The parity index of kharif jowar, gram, cotton and tur decreased by 20.93 per cent, 20.53 per cent, 8.97 per cent and 8.0 per cent, respectively during the same period.

The ratio of irrigated land to irrigable land, rainfall and cropping intensity increased by 96.30 per cent, 63.79 per cent and 7.44 per cent respectively, during the period 1980-81 to 2005-06. The availability of land per farmer, per capita production of foodgrains and ratio of agricultural population to total population decreased by 38.18 per cent, 39.46 per cent and 18.75 per cent respectively, during the same period.

Development of Sustainability Index through Principle Component Analysis

Table 2 presented the eigen values and cumulative per cent for a set of variables. It can be seen from the table that the component analysis produces components in descending order of their importance i.e., the first component explains the maximum amount of variation, and the last component the minimum. The first component only accounts for 38 per cent of variation. The second component explains 53 per cent of variation. It takes sixth components to produce 85 per cent of variation, and another two for 90 per cent variation. The first six components account for a sizeable amount of variation i.e., 85 per cent thus succeeded in effectively reducing the dimension of the problem. The subsequent components contribute very little.

Table 1. Triennium average of indicators of sustainability and related agricultural development in Vidarbha

Sr. No.	Indicators of Sustainability	Unit	1981-83	1990-92	2000-02	2004-06	Percentage increase (+)/ decrease (-)
1.	Index of Surface Percentage of Crops	-	0.33	0.38	0.50	0.50	51.52
2.	Crop yield in cotton	Kg/ha	576.33	823.67	1167.33	1453.00	152.11
3.	Crop yield in soybean	Kg/ha	0.00	6362.50	12225.67	9087.00	42.82
4.	Crop yield in tur	Kg/ha	5447.00	5600.67	9986.67	8686.00	59.46
5.	Crop yield in kharif jowar	Kg/ha	7542.33	9907	9963.00	8263.00	9.56
6.	Crop yield in sunflower	Kg/ha	1085.33	7897.67	12528.67	8640.67	696.13
7.	Crop yield in gram	Kg/ha	2725.00	4266	5998.33	5474.33	100.89
8.	Gap in gross returns in cotton	Rs.	956.24	2044.32	7350.18	7205.74	653.55
9.	Gap in gross returns in soybean	Rs	0.00	3344.78	9264.40	6299.27	88.33
10.	Gap in gross returns in tur	Rs	1601.93	4370.28	8580.46	3245.45	102.60
11.	Gap in gross returns in kh jowar	Rs	1776.10	2675.52	7239.85	6420.83	261.51
12.	Gap in gross returns in sunflower	Rs	2090.24	3790.72	2519.97	1010.87	-51.64
13.	Gap in gross returns in gram	Rs	1906.67	2627.09	4644.53	7623.29	368.06
14.	Cost-benefit ratio of cotton	-	1.33	0.99	0.97	1.12	-15.79
15.	Cost-benefit ratio of soybean	-	1.15	1.36	1.12	1.33	15.65
16.	Cost-benefit ratio of tur	-	1.36	1.37	1.21	1.22	-10.29
17.	Cost-benefit ratio of Kh.jowar	-	1.47	1.23	0.98	1.09	-25.85
18.	Cost-benefit ratio of sunflower	-	0.53	0.95	1.22	1.37	158.49
19.	Cost-benefit ratio of gram	-	1.28	1.35	1.16	1.04	-18.75
20.	Parity index of cotton	-	0.78	0.64	0.63	0.71	-8.97
21.	Parity index of soybean	-	0.56	0.77	0.56	0.66	17.86
22.	Parity index of tur	-	0.75	0.80	0.69	0.69	-8.0
23.	Parity index of kharif jowar	-	0.86	0.83	0.66	0.68	-20.93
24.	Parity index of sunflower	-	0.40	0.58	0.67	0.87	117.50

Sr. No.	Indicators of Sustainability	Unit	1981-83	1990-92	2000-02	2004-06	Percentage increase (+)/ decrease (-)
25.	Parity index of gram	-	0.78	0.85	0.74	0.62	-20.51
26.	Availability of land per farmer	Ha.	2.75	2.23	1.77	1.70	-38.18
27.	Irrigated land / Irrigable land	-	0.27	0.32	0.49	0.53	96.30
28.	Cropping Intensity	Per cent	110.34	117.66	119.93	118.55	7.44
29.	Area under High Yielding Varieties	-	24.94	30.56	29.39	31.81	27.55
30.	Per capita production of foodgrains	Kg/person/annum	200.41	187.75	139.52	121.32	-39.46
31.	Agricultural population / Total population	-	0.32	0.30	0.27	0.26	-18.75
32.	Rainfall	mm	760.23	1040.38	1025.36	1245.21	63.79

Table 2. Cummulative variations and eigen values accounted for by components

Set of Components	Eigen values	Cumulative Percentage	Set of Components	Eigen values	Cummulative Percentage
1	7.5838	0.379	11	0.2734	0.968
2	2.9809	0.528	12	0.2364	0.980
3	2.3891	0.648	13	0.1163	0.986
4	1.8738	0.741	14	0.1092	0.992
5	1.2658	0.805	15	0.0557	0.994
6	0.8515	0.847	16	0.0418	0.996
7	0.7459	0.885	17	0.0300	0.998
8	0.5616	0.913	18	0.0220	0.999
9	0.5108	0.938	19	0.0140	1.000
10	0.3323	0.955	20	0.0058	1.000

Sustainability Index

Table 3 presents the Sustainability Index of Vidarbha region derived from the various indicators of sustainability. It was observed that the results of the principle component analysis were astonishingly clear and appealing from an agricultural perspective. The numerical scores ranging from -0.308 to 0.350, which represented the Sustainability Index, obtained through principle component analysis. Further, it was observed that the index obtained through the principle component analysis was seen declining. Higher numbers represented higher sustainability whereas, lower number represented lower sustainability. The higher sustainability was observed in the year 1982-83 (0.350) followed by 1981-82 (0.326) and 1983-84 (0.275). The lower sustainability was observed in the year 2001-02 (-0.308). The higher a year's sustainability index, the better positioned it is to maintain favorable agricultural conditions into the future.

Table 3. Sustainability index of Vidarbha region

Years	Sustainability Index	Years	Sustainability Index
1980-81	0.233	1993-94	-0.081
1981-82	0.326	1994-95	0.075
1982-83	0.350	1995-96	0.028
1983-84	0.275	1996-97	-0.010
1984-85	0.217	1997-98	-0.258
1985-86	0.186	1998-99	-0.298
1986-87	0.157	1999-00	-0.216
1987-88	0.065	2000-01	-0.248
1988-89	-0.088	2001-02	-0.308
1989-90	0.093	2002-03	-0.225
1990-91	-0.008	2003-04	-0.227
1991-92	0.036	2004-05	-0.171
1992-93	0.097	2005-06	-0.198

The Sustainability Index was a measure of overall progress towards agricultural sustainability developed for 26 years. The main goal of this index was to take sustainability into account in the measurement of the agricultural development level. By facilitating, comparative analysis across indicators, the index provided a mechanism for making agricultural management more quantitative, empirically grounded and systematic.

The sustainability index permits comparisons of agricultural progress in a systematic and quantitative fashion. It represented a first step towards a more analytically driven approach to agricultural decision making. The sustainability index enables to identify the areas of success or failures. It helped to fill a long-existing gap in agricultural performance evaluation. It offered a small step toward a more vigorous and quantitative approach to agricultural decision making.

Classification of Years on the basis of Sustainability Index

After developing the Sustainability Index, it becomes useful to compare the years and their performance against each other that were similarly situated and the sustainability index makes such comparisons relatively easy to do. The composite indices scores were further used to classify years into different categories indicating varying levels of sustainability.

Table 4 showed the classification of years on the basis of Sustainability Index. It can be seen from the Table that classification of years were categorized into 4 different heads on the basis of composite index scores derived.

They were -

1) Not Sustainable (Below 0)
2) Moderately sustainable (0 to 0.1)
3) Sustainable (0.1 to 0.3)
4) Highly Sustainable (Above 0.3)

It can be seen from the table, the scores below zero or the negative scores represented Not Sustainable years. The years 1988-89, 1990-91 and years from 1996-97 onwards to 2005-06

were classified as Not Sustainable years. According to Zora (2006), 1997 was the first year in which farmer suicides began to be noticed in Vidarbha. In a report results obtained by Mishra (2006) in "Suicide of Farmers in Maharashtra", submitted by Indira Gandhi Institute of Development Research, Mumbai concluded that from 1995 to 2004, suicide deaths in Maharashtra increased from 11,866 to 14,729. Across divisions, Amravati and Nagpur were the ones with greater SMR (Suicide Mortality Rate) than the state average. The above results may be due to observed Not Sustainable years.

Table 4. Classification of years on the basis of sustainability index

Categories	Years	Percentage of Years
Not Sustainable (Below 0)	1988-89	46
	1990-91	
	1996-97	
	1997-98	
	1998-99	
	1999-00	
	2000-01	
	2001-02	
	2002-03	
	2003-04	
	2004-05	
	2005-06	
Moderately Sustainable (0 to 0.1)	1986-87	15
	1989-90	
	1992-93	
	1995-96	
Sustainable (0.1 to 0.3)	1980-81	27
	1983-84	
	1984-85	
	1987-88	
	1991-92	
	1993-94	
	1994-95	
Highly Sustainable (Above 0.3)	1981-82	12
	1982-83	
	1985-86	

Further, the scores ranging from 0 to 0.1 were classified as Moderately Sustainable years. The years 1986-87, 1989-90, 1992-93 and 1995-96 were classified as Moderately Sustainable years.

The scores ranging from 0.1 to 0.3 were classified as Sustainable years. The scores above 0.3 were classified as Highly Sustainable years. The years were 1981-82, 1982-83 and 1985-86.

In percentage terms, forty-six per cent of years from 1980-81 to 2005-06 were classified as Not Sustainable years. Fifteen per cent of years were classified as Moderately Sustainable, twenty seven per cent were classified as Sustainable and only twelve per cent were classified as Highly Sustainable years.

The classification of years formed on the basis on the sustainability index helped to identify appropriate years against which one can benchmark agricultural performance.

Identification of Factors Responsible for Sustainability

After classification of years, an attempt has been made to identify the factors responsible for sustainability. However, for the purpose of identification of factors responsible for sustainability, the years were classified into two main groups i.e., Sustainable years and Not Sustainable years. For this purpose, a non-parametric Mann Whitney U test has been carried out.

Table 5 presented the means of various indicators of sustainability. The means of Sustainable and Not Sustainable years were compared with the Mann Whitney U Test. The indicators of sustainability were tested with the help of hypotheses that there was no significant difference between the years of sustainability and years of not sustainability.

Productivities per hectare of major crop contributes to sustainability. From the Table, it can be seen that the productivities per hectare of cotton, tur, kharif jowar, sunflower and gram were the significant contributing variables for sustainability. Since soybean has come into existence in the 1990s, the data was not considered for sustainability. Therefore, all efforts should be made to improve the production per hectare for sustainable agriculture.

Gap in gross returns in cotton, kharif jowar and gram were the significant contributing variables for sustainability. Gap in gross returns in tur and sunflower was the non-significant variable for sustainability. Tur, being a drought resistant crop, suitable for dry-land farming in the Nagpur district and which is predominantly used as an intercrop with other crops have a dominating importance. There is a possibility of improving the productivity of tur by narrowing the gross returns or increase the productivity per hectare or offering proper remunerative prices to the crop. In Nagpur district, the area under cultivation of sunflower is less than one per cent. For sustainability of sunflower, efforts must be made to bring more land under sunflower crop. Also there is a need to decrease the gap in gross returns or increase the productivity per hectare.

As far as cost-benefit ratios of major crops were concerned, cotton, kharif jowar and gram were the significant contributing variables for sustainability. Cost-benefit ratio of soybean, tur and sunflower were the non significant variable for sustainability. Since soybean crop was an introduced crop in the Vidarbha region in the 1990s, it does not contribute to sustainability. For sustainability of tur and sunflower, effort must be made to cultivate these crops at low cost production technology to improve the gross returns. By improving the gross returns, sustainability of these crops will definitely increase.

Table 5. Non-parametric test applied for comparing means of indicators of sustainability

Sr. No.	Indicators of Sustainability	Unit	Not Sustainable years	Sustainable years	U_{Obt}
1.	ISPC	-	-	-	-
2.	Crop yield in cotton	Kg/ha	1139.30	1153.30	29.5*
3.	Crop yield in soybean	Kg/ha	-	-	-
4.	Crop yield in tur	Kg/ha	7913.10	8206.40	10.5*
5.	Crop yield in kharif jowar	Kg/ha	10064.00	12606.00	36**
6.	Crop yield in sunflower	Kg/ha	10524.00	11247.00	16.5*
7.	Crop yield in gram	Kg/ha	5363.50	5762.60	6.5*
8.	Gap in gross returns in cotton	Rs/ha	6496.70	1780.22	11.5*
9.	Gap in gross returns in soybean	Rs/ha	-	-	-
10.	Gap in gross returns in tur	Rs/ha	5483.1	3376.53	42.5
11.	Gap in gross returns in kharif jowar	Rs/ha	5613.60	2299.33	9.5*
12.	Gap in gross returns in sunflower	Rs/ha	4024.60	3103.33	74.5
13.	Gap in gross returns in gram	Rs/ha.	5310.30	2467.06	23.5*
14.	Cost-benefit ratio of cotton	-	1.03	1.19	35.2**
15.	Cost-benefit ratio of soybean	-	1.24	1.17	59
16.	Cost-benefit ratio of tur	-	1.22	1.25	73.5
17.	Cost-benefit ratio of kharif jowar	-	1.03	1.30	17.5*
18.	Cost-benefit ratio of sunflower	-	1.10	0.88	44
19.	Cost-benefit ratio of gram	-	1.11	1.27	41**
20.	Parity index of cotton	-	0.66	0.73	52.4
21.	Parity index of soybean	-	0.60	0.63	68.5
22.	Parity index of tur	-	0.71	0.70	73.5
23.	Parity index of kharif jowar	-	0.67	0.85	17*
24.	Parity index of sunflower	-	0.64	0.68	76
25.	Parity index of gram	-	0.67	0.79	37.5**

Sr. No.	Indicators of Sustainability	Unit	Not Sustainable years	Sustainable years	U_{Obt}
26.	Availability of land per farmer	Ha.	1.88	2.41	8.5*
27.	Irrigated land / Irrigable land	-	0.45	0.31	10*
28.	Cropping Intensity	-	113.48	118.71	52
29.	Area under High Yielding Varieties	Kg/ Person/ annum	30.62	28.65	40.5**
30.	Per capita production of foodgrains	Percent	156.57	190.65	32.5**
31.	Agricultural population / Total population		0.28	0.31	6*
32.	Weather Index	mm.	1063.40	924.87	19.5*

**, * Mann-Whitney U Test is statistically significant at 5 and 1 per cent.

The parity index of kharif jowar and gram were the significant contributing variables for sustainability. Parity index of cotton, soybean, tur and sunflower were the non significant variable for sustainability. For cotton, soybean, tur and sunflower, the existing gross returns were not adequate enough to cover the cost of cultivation. Efforts must be made to maintain parity between the input and output prices of these crops.

Availability of land per farmer, ratio of irrigated land to irrigable land, per capita of production of food grains, ratio of agricultural population to total population, area under High Yielding Varieties and Rainfall were the significant contributing variables for sustainability.

Conclusion

From this study, it is concluded that, the sustainability index of Vidarbha region developed through principal component analysis was seen declining over the years. The Sustainability index ranges from -0.308 to 0.350. Higher number of sustainability index represents higher sustainability whereas, lower number represents lower sustainability. This concludes that the Vidarbha agriculture is tending towards un-sustainability. The study also found that the productivities per hectare of cotton, tur, kharif jowar, sunflower and gram, gap in gross returns in cotton, kharif jowar and gram, cost-benefit ratio of cotton, kharif jowar and gram, parity index of kharif jowar and gram, availability of land per farmer, ratio of irrigated land to irrigable land, per capita production of foodgrains, ratio of agricultural population to total population, area under high yielding varieties and weather index were the significant contributing variables for agricultural sustainability of the Vidarbha region.

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Effect of Mulch, Clay and Organic Matter on Soil Chemical and Biological Properties of Sandy Soil and Growth of Physic Nut (*Jatropha curcas* L.)

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Abstract

Sandy soil for growth media of physic nut in Situbondo, East Jawa has low soil fertility. In this field study, the effect of addition of clay and organic matter with or without mulch on soil properties was conducted from May until November 2010. The treatments consisted of (1) Sandy soil (as control), (2) Incorporation of sandy soil with 10% clay and 1,6% organic matter, (3) Sandy soil covered with mulch and incorporated with 10% clay and 1,6% organic matter. Crotalaria juncea was used as a source of organic matter and mulch. The results showed that incorporation of sandy soil with 10% clay and 1,6% organic matter and mulch increased soil chemical properties of sandy soil which were expressed by increasing soil C-organic, N, P and K contents by 46%, 18%, 73% dan 48%, respectively. The treatmet also improved soil biological properties indicated by more population of soil fungi and bacteria growth on the media. As consequence the growth of physic nut was betterr than that grow on sandy soil.

Keywords: clay, Jatropha curcas L., mulch, organic matter, physic nut, sandy soil, soil properties

Introduction

Potential bio-energy crop *Jatropha curc*as L (physic nut) is best suitable planted on sandy soils because the plant needs well aerated soils and it is intolerant of waterlogged conditions (da Schio, 2010). However, this natural property of sandy soil can cause problems in their use for agricultural production. The sandy soil was inherently low in all aspects of soil fertility and has a capacity to retain water and applied nutrients (Farrington and Campbell 1970). On the other hand, in order to support a high biomass production physic nut indicates a high demand for nitrogen and phosphorus fertilization (Daey Ouwens *et al*., 2007). One strategy to support the growth of physic nut on sandy soil is by improving soil fertility.

Among soil properties, CEC, pH organic C and clay have significant effect on the availability of nutrients. Soils which have high amounts of organic matter and moderately weathered clays tend to have high CECs. As soils become highly weathered, the CEC of the soil decreases. Sandy soils, too, generally have lower CEC values. This is due to the lesser surface of sandy particles in comparison with clay minerals, which decreases the ability of sand particles to hold and retain nutrients (Brady and Weil, 2009). Mulching and incorporation of clay soil and organic matter to sandy soil might have positive effect on soil chemical properties of sandy soil which in turn might improve soil biological properties and growth of physic nut.

Some studies have reported that mulch applications significantly increased soil organic carbon, K, P, root and top biomass of cassava (Cadavid *et al*., 1998). Sinkeviciene *et al*. (2009) reported that mulch increased available P and K contents and crop yield. Application

of mulch on sandy soil with slope of 7% decreased soil and nutrient loss due to leaching (Wakindiki and Danga, 2011). However most of the studies were carried out focusing on the effect of mulching on soil properties and crop yield. There was a limited information regarding on the combined effect of mulch, clay and organic matter on soil chemical and biological properties. This study aimed to quantify the effect of mulch, clay and organic matter on soil chemical and biological properties and growth of physic nut.

Materials and Methods

Land Preparation

A field study was carried out at Situbondo, Each Java from April to November 2010. The soil texture is dominated by sand particles with proportion of 77% sand, 17 % silt and 6% clay. The site was chosen because it has been established as a seeding production area for jatropha (physic nut).

Stems of physic nut cultivar IP2 were grown at poly bags for 30 days before they were transplanted to the plot size of 9 m^2. Planting space of physic nut stem was 3 m x 3 m, so each plot consisted of 9 stems. Application of urea 50 g/plant and Ponska 100 g/plant were added twice, at 20 and 50 days after transplanting. The soil was iririgated periodically with interval of 10 days.

Experimental Design

The treatments were three kinds media growth of physic nut, namely (1) Sandy soil as a control, (2) Sandy soil + 10 % clay soil + 1.6% organic matter, and (3) Sandy soil + Mulch+ 10% clay soil + 1.6% organic matter. There were eight replicates of each treatment. The arrangement of treatments used a Randomized Block design.

Addition of Mulch, Clay Soil and Organic Matter

Two days after transplanting of physic nut stems, mulch of *C. juncea* straw was spread out manually in about 10 cm thick layer to the sandy soil with had been incorporated with 10% clay soil and 1.6% organic matter. Clay soil (67% clay) was collected from sub soil of the land located nearby experimental site. The clay soil was added with the rate 10% (equivalent to 248 kg/plot or 276 tones/ha). *Crotalaria juncea* was used as a source of organic matter which was planted on another plot 45 days before it was added to the sandy soil. The organic matter was added at rates of 1.6% (equivalent to 61.7 kg/plot or 69 tones/ha).

Soil Chemical Properties Measurements

Soil organic C content was measured by Walkey Black Method, total nitrogen was determined employing Kjehldahl procedure, soil available phosphorus was measured using Bray 1 methode, exchengable soil potassium was measured by Flame photometric. Soil samples were collected as bulk samples at 30 and 60 days after treatment (DAT).

Soil Biological Properties Measurements

Isolates extracted from soil samples were cultured on spesific media. To culture bacteria and actinomycetes, tryptic soy agar media was used. Fungi were cultured on Martin media agar. Soil was sampled twice, at 30 and 60 DAT.

Plant Growth Measurements

Variables measures were plant height and number of leaf of physic nuts. Measurements were done on three plants per plots which were choosen randomly. Plant height and number leaf were measured at 30, 60 and 90 DAT.

Results and Discussion

Soil Chemical Properties

All treatments applied on sandy soil significantly improved the soil chemical properties. The highest soil C organic content was in plot where sandy soil was covered with mulch and added with clay and organic matter (Table 1). Covering sandy soil with mulch which the soil had been added with clay and organic matter had C organic content 42% and 46% higher than untreated sandy soil at 30 and 60 days after treatment respectively. Addition of clay and organic matter did not significantly increase C organic content of sandy soil. Increasing of soil C organic content on the sandy soil covered with mulch and added with clay and organic matter might be due to decreasing loss of the materials by leaching during irrigation of the soil. Atreya *et al.* (2005) reported that mulching of acidic sandy loam reduced annual soil organic matter by 52%.

Table 1. Effect of mulch, clay and organic matter on C organic content of sandy soil at 30 and 60 days after treatment (DAT)

Treatments	C organic content (%)	
	30 DAT	60 DAT
SS (control)*)	0.74 a	0.76 a
SS + 10% CS+ 1,6% OM	0.84 a	0.87 a
SS + M + 10% CS + 1,6% OM	1.05 b	1.11 b
LSD 5%	0.16	0.22

*) SS = Sandy Soil, M = Mulch, CS = Clay Soil, OM = Organic Matter

Effect of treatment on sandy soil on soil total nitrogen content is presented in Table 2. Mulching of sandy soil which had been added with clay and organic matter signinificantly increased soil nitrogen content by 18% at 30 DAT.

The treatments also significantly increased available phosphorus and potassium content of the sandy soils. Mulching the sandy soil which had been added by clay and organic matter increased soil phosphorus content by 93% and 67% at 30 DAT and 60 DAT respectively (Table 3). The sandy soil covered by mulch and added with clay and organic matter had an exhangeable potasssium content 101 % and 48 % at 30 DAT and 60 DAT respectively than untreated sandy soil at the same date of soil sampling (Table 4).

Table 2. Effect of mulch, clay and organic matter on total nitrogen (N) content of sandy soil at 30 and 60 days after treatment (DAT)

Treatments	N content (%)	
	30 DAT	60 DAT
SS (control)*)	0.11 a	0.10
SS + 10% CS+ 1,6% OM	0.11 a	0.10
SS + M + 10% CS + 1,6% OM	0.13 b	0.12
LSD 5%	0.01	n.s

*) SS = Sandy Soil, M = Mulch, CS = Clay Soil, OM = Organic Matter, ns = not significant

Table 3. Effect of mulch, clay and organic matter on available phosphorus (P) content of sandy soil at 30 and 60 days after treatment (DAT)

Treatments	Available P_2O_5 content (%)	
	30 DAT	60 DAT
SS (control)*)	21.50 a	29.80 a
SS + 10% CS+ 1,6% OM	20.68 a	24.10 a
SS + M + 10% CS + 1,6% OM	41.49 b	49.80 b
LSD 5%	5.28	16.63

*) SS = Sandy Soil, M = Mulch, CS = Clay Soil, OM = Organic Matter

Table 4. Effect of mulch, clay and organic matter on exchangeable potassium (K_2O) content of sandy soil at 30 and 60 days after treatment (DAT)

Treatments	Exch K_2O content (%)	
	30 DAT	60 DAT
SS (control)*)	0.68 a	0.71 a
SS + 10% CS+ 1,6% OM	0.84 b	0.81 a
SS + M + 10% CS + 1,6% OM	1.37 c	1.05 b
LSD 5%	0.17	0.21

*) SS = Sandy Soil, M = Mulch, CS = Clay Soil, OM = Organic Matter

The positive influence of organic mulches on enriching of soil nutrients is well documented. Saroa and Lal (2004) found that mulching enriched total phosphorus concentration in the soil after 4 years of mulching from 601 up to 658 mg/kg and from 491 up to 694 mg/kg after 11 years of mulching. Wakindiki and Danga (2011) reported that application of straw mulch increased soil nitrogen, phosphorus and potassium concentrations. They attributed that increasing the nutrients in mulched soil due to minimization of soil and nutrients losses. In the present study, the soil nutrients increased in sandy soil covered with mulch and added with clay and organic matter might be due to minimization of soil loss and increasing the holding capacity of the sandy soil as a consequence of addition of clay and organic matter.

Soil Biological Properties

This study demonstrated that mulching of sandy soil which had been added with clay and organic matter enhanced soil biological properties which was expressed by increasing population of soil microorganisms. Table 5 indicated that sandy soil covered with mulch and incorporated with clay and organic matter had the highest population of soil fungi. Compared to the untreated sandy soil, mulching of sandy soil which had been added with clay and organic matter was occupied by fungi more than three folds and two folds at 30 DAT and 60 DAT respectively.

Table 5. Effect of mulch, clay and organic matter on fungi population of sandy soil at 30 and 60 days after treatment (DAT)

Treatments	Population of Fungi ($x10^3$ cfu/ml)	
	30 DAT	60 DAT
SS (control)*)	5.00 a	9.62 a
SS + 10% CS+ 1,6% OM	11.25 b	16.37 b
SS + M + 10% CS + 1,6% OM	16.87 c	20.62 c
LSD 5%	3.22	3.81

*) SS = Sandy Soil, M = Mulch, CS = Clay Soil, OM = Organic Matter

Treated sandy soil with mulch, clay and organic matter also had the highest population of soil bacteria (Table 6). The treatments increased population of soil bacteria by three folds and 1.5 folds at 30 DAT and 60 DAT respectively.

Table 6. Effect of mulch, clay and organic matter on bacteria population of sandy soil at 30 and 60 days after treatment (DAT)

Treatments	Population of Bacteria ($x10^3$ cfu/ml)	
	30 DAT	60 DAT
SS (control)*)	8.12 a	27.62 a
SS + 10% CS+ 1,6% OM	15.87 b	40.00 b
SS + M + 10% CS + 1,6% OM	24.37 c	46.12 b
LSD 5%	4.63	7.27

*) SS = Sandy Soil, M = Mulch, CS = Clay Soil, OM = Organic Matter

The effect of mulch to the sandy soil which had been added with clay and organic matter on population of actinomycetes is presented in Table 7. The treatments caused the sandy soil had the highest population of actinomycetes. Compared to the untreated sandy soil, addition of mulch, clay and organic matter to sandy soil increased population of actinomycetes by 21% and 46% at 30 DAT and 60 DAT.

Some studies have reported the positive effect of mulch on soil microorganisms population. Augmentation of population of soil fungi, bacteria and actinomycetes by mulching was reported by Gaur and Mukherjee (1980). Mundy and Agnew (2002) found that mulching increased fungi population of soil under vineyard. They attributed that increasing of soil microorganisms populations were due to increasing soil water content and decreasing soil temperature which were suitable conditions for soil microorganisms. Kumar *et al.* (2010)

reported that *C. juncea* used as bio-mulching increased soil water holding capacity by 56.9% and soil moisture by 68.5% at 30 cm depth.

Table 7. Effect of mulch, clay and organic matter on actinomycetes population of sandy soil at 30 and 60 days after treatment (DAT)

Treatments	Population of *Actinomycetes* ($x10^3$ cfu/ml)	
	30 DAT	60 DAT
SS (control)*)	8.87 a	9.12 a
SS + 10% CS+ 1,6% OM	10.50 b	11.37 b
SS + M + 10% CS + 1,6% OM	10.75 b	13.37 c
LSD 5%	1.58	1.68

*) SS = Sandy Soil, M = Mulch, CS = Clay Soil, OM = Organic Matter

In the present study, combined influence of mulch, clay and organic matter might occur on soil biological properties improvements. A previous study found that more soil microorganisms were present in soil fraction with a high content of clay and organic matter (Van Gestel *et al.* 1996).

Plant Growth

Mulching of sandy soil which had been added with clay and organic matter increased plant height and number of leaf of physic nut. Physic nuts grown on mulched sandy soil and added with clay and organic matter taller more than 42%, 30% and 28% than those growth on untreated sandy soil at 30, 60 and 90 DAT respectively (Table 8).

Table 8. Effect of mulch, clay and organic matter on plant height of physic nut growth on sandy soil at 30 and 60, and 90 days after treatment (DAT)

Treatments	Plant Height (cm)		
	30 DAT	**60 DAT**	**90 DAT**
SS (control)*)	46.25 a	73.50 a	97.66 a
SS + 10% CS+ 1,6% OM	55.37 b	81.83 b	107.00 b
SS + M + 10% CS + 1,6% OM	65.45 c	95.70 c	124.62 c
LSD 5%	4.75	4.89	9.28

*) SS = Sandy Soil, M = Mulch, CS = Clay Soil, OM = Organic Matter

Sandy soil treated with mulch, clay and organic matter also caused physic nut had the most number of leaf (Table 9). The treatments increased number of leaf by 97%, 70% and 47% at 30, 60 and 90 DAT.

Similar results of the influence of mulch on plant growth development was reported by Arin and Ankara (2001) and Salman *et al*. (1992). The pointed out that mulching increased soil temperature so that vegetative development of tomatoes was enhanced. In this present study, increasing of plant height and number of leaf of physic nuts grown on mulched sandy soil and added with clay and organic matter might because the treatments improved soil physical and biological of sandy soils which created favourable conditions for root growth. A

previous study reported that addition of clay and organic matter to sandy soil induced vegetative growth of subterranean clover which was indicated by increasing root length and fresh weight of shoots (Djajadi, 2007).

Table 9. Effect of mulch, clay and organic matter on number of leaf of physic nut growth on sandy soil at 30 and 60, and 90 days after treatment (DAT)

Treatments	Number of Leaf		
	30 DAT	60 DAT	90 DAT
SS (control)*)	53.75 a	125.16 a	197.33 a
SS + 10% CS+ 1,6% OM	81.37 b	151.83 b	238.08 b
SS + M + 10% CS + 1,6% OM	106.08 c	211.20 c	290.66 c
LSD 5%	8.90	15.60	15.93

*) SS = Sandy Soil, M = Mulch, CS = Clay Soil, OM = Organic Matter

Conclusion

Mulching of sandy soil which had been added with clay and organic matter improved soil chemical and biological properties of sandy soil which result in enhancing growth of physic nut. Nutrients contents (C organic, total N, availability P_2O_5 and exch. K_2O) of sandy soil were increased by addition of mulch, clay and organic matter. The treatments also developed population of soil fungi, bacteria and actinomycetes and also enhanced the plant height and number of leaf of physic nut.

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Sustainability of Food Resources by Eco-Farming Implementation: The Role of Farmer's Socio Economics

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Abstract

The agricultural sector in Indonesia is still the mainstay of the sector. It can be seen from the role of agriculture to GDP in 2010 increased from 14.5% to 15.3% (BPS, 2011), besides to meet the food needs of more than 235 million people and will continue to grow with growth rate is 1.49% per year (BPS, 2011). Agricultural development is an effort to support food security, now face various challenges that more serious and complex, especially environmental carrying capacity. This is because the agricultural sector produces waste and pollution are high enough, high in use of natural resources, especially for water and land as well as vulnerable to climatic shocks. With the limited carrying capacity of the natural environment and the climate change, the sustainable agricultural development is needed through implementation of the eco-farming concept. Eco-farming is a form of agriculture that seek wherever possible to achieve harmony with their environment by considering the social, economic and ecological. According to Leahy (2011), transfer of agricultural systems towards eco-farming is something that is urgent to end hunger and face the challenges of climate change. According to Amin (2010), other constraints in agricultural development efforts in rural on a households scale farmers is the problem of socio-economics of farmers, where farmers generally have a narrow land (subsistence), labor and fewer farmers generally have a fairly old age with the level education is low, it will limited to technology usage. Various studies have explained that the farmers' socio-economic variables affect the process of receiving an agricultural innovation (Mafimisebi et al, 2006; Rahman, 2007; Rezvanfar, 2007; Rezvanfar and Arabi 2009, Oladel and Rantseo, 2010) by using linear regression, the dependent variable is the socio-economic attributes of farmers (gender, age, experience, education, family size, income). Therefore needs to be done in-depth review of the factors of socio economics of the farmer. This paper is a review of the role of farmers' socio-economic towards applying eco farming.

Keywords: eco-farming, food resources, farmer socio economy, sustainability

Introduction

Lately the need for more attention to the environment of each construction sector continues to increase, because there has been climate change, decline in the quality of the environment and disruption of ecosystem balance. Environmental issues are increasingly complex issues is rooted in the growth of a rapidly growing population. Human interaction with the environment to meet their needs has led to the depletion of natural resources leading to massive environmental degradation, particularly in developing nations.

In the future population growth continues to increase, leading to increasing food demand. Learning from experience in the 20th century where even though food production exceeded population growth, but the condition can not be sustained because food production systems at the time did not consider environmental aspects. According to Salim (2010), conventional development has managed to increase economic growth, but failed in the social and environmental aspects. Therefore we need a proper system of food production in the agro-ecosystem to ensure its sustainability.

Development of agricultural production of food crops have been carried out with various programs, especially for rice intensification. These programs actually lead to saturation (leveling off) as a result of intensive use of chemical fertilizers, followed by the more limited availability of sources of nutrients for plants. These conditions provide opportunities for livestock development is through the use of animal manure to improve soil fertility, besides cattle can also use agricultural waste as feed. Thus the integration of livestock with crop production systems is expected to be one way out to realize food resources sustainability.

Crop livestock system (CLS) in Indonesia have long been known as paddy and livestock integration system (SIPT). This program has been implemented since 2002. SIPT program is to optimize the utilization of local resources such as the use of straw as livestock feed and cow manure can be processed into organic fertilizer which is very useful for improving the nutrients that plants need. With the integration of rice and livestock systems, is expected zero waste from both farming system (Director General of Livestock Production, 2002; Diwyanto *et al*, 2002). This program is very good concept if it can be implemented by farmers. But the results of the Muslim (2006) research, the implementation of the integration system of rice and beef (SIPT), concluded that the technology has not fully responded applied by farmers (in the case Majalengka) because the majority of the farmers did not use manure on the farm even when a packet recommendation in the SIPT activities. Implementation SIPT in West Lombok regency also experienced failures, because group members did not impound cattle ranchers. Priyanti (2007) states, the adoption of integrated crop livestock farming in Central Java, Yogyakarta and East Java also has not been implemented in a balanced way by most farmer-breeders. The results Elly (2008), states that livestock farmers who receive assistance on SIPT program in South Sulawesi have largely failed because the response of farmers in adopting the program have been inadequate. This indicates that the program was introduced to farmers has not been able to be implemented, so it needs to make a model sustainable farming system based on socio economic conditions of farmers and also consider the ecological dimension.

The Agroecosystem

An ecosystem is a natural system that is formed by dynamic interactions between biotic and non-biotic elements in a defined area. Biotic elements include plants, insects (pests, natural enemies, decomposers), microbes and other living organisms, and non-biotic elements comprise weather components such as temperature, relative humidity, wind, sunshine, rain and soil. Each element has its special characteristics and role in the system that, as a function of time and place, will influence the distribution and population of living organisms. The term ecosystem also involves nutrient and energy flows within the system. An agro-ecosystem is characterized by a much simpler composition with regard to the number of species residing in the system and the relative simplicity of energy flows than a natural, stable ecosystem. Therefore, the agro-ecosystem needs energy input to maintain its balance.

Agricultural activities are generally convert the natural ecosystem to artificial ecosystem that called agro-ecosystem. According to Conway (1997), agro-ecosystem is an ecological and socio-economic systems associated with the process of domestication of plants and / or animals by humans that aims to meet food needs. In the hierarchy can be seen in Figure1.

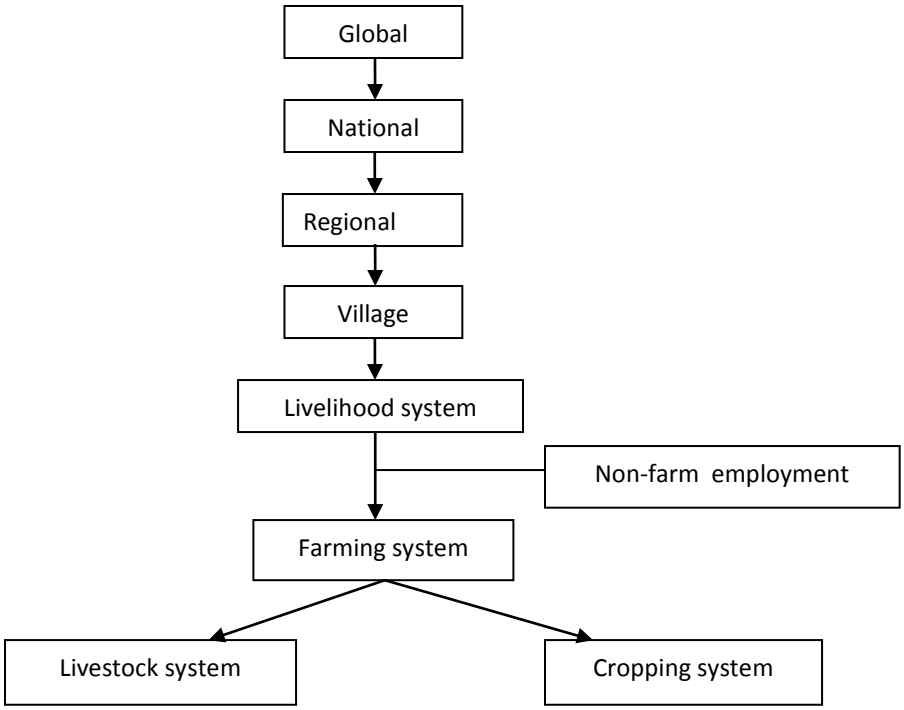

Figure1. Hierarchy of agro-ecosystems

Eco Farming and Food Resources Sustainability

Eco-farming is a form of agricultural cultivation that seek wherever possible to achieve harmony with their environment by considering the social, economic and ecological. Some experts say eco-farming as a reasonable organic farming, because the utilization of non-organic production inputs are still permitted in limited quantities by taking into account the carrying capacity and the ability to recover from environmental pollutants. In contrast to the absolute organic farming which does not allow the use of non-organic production inputs.

Eco-farming system characteristics include: (1) Utilization of local resources to the maximum but still pay attention to sustainability, (2) The use of external inputs are minimal, simply as a replacement if local resources are not available, (3) Emphasis on cultivation of food crops combined with other crops can be harvested before the main crop is produced, (4) Ensuring that the basic biological functions of soil, water, nutrients and humus can be maintained, (5) Maintain the diversity of plants and animals to balance ecological and economic stability by developing species and varieties local, and (6) Creating an attractive form of land management and capable of providing welfare for the local community (Egger,

1990). According to Leahy (2011), transfer of agricultural systems towards eco-farming is something that is urgent to end hunger and face the challenges of climate change.

The main obstacle in the utilization of waste in the system integration is one of the eco-farming system is the farmer have not use a forage crop waste and livestock waste optimally. This will cause high levels of pollution along with the increased productivity of livestock and rice farming partially. This condition is certainly influenced by the characteristics of farmers such as socio economics factors.

Farmer Socio Economy

National agricultural development in Indonesia is constrained, narrow land, labor becomes more scarce, aging, and less educated, and limited capital. Land the narrow limits of technology options, farmers are older and less educated slow to adopt technology, and limited capital hinder the application of technologies (Amin, 2010).

Various studies have explained that the variable characteristics of the farmer is the socio economic variables affect the process of receiving an agricultural innovation (Mafimisebi *et al*, 2006; Rahman, 2007; Rezvanfar, 2007; Rezvanfar and Arabi 2009) using linear regression, the dependent variable is a characteristic attribute of farmers (gender, age, experience, education, family size, income) and farm characteristics variables (type of farm, farm size and location). Meanwhile, according to Baidu-Forsan (1999), that farmer behavior adoption of technology can be measured from the attributes of farmers, farm attributes, the attributes of infrastructure and the perception of agricultural technologies. Oladel and Rantseo (2010), concluded from his research that the factors that still relevant to the application of farm technology by farmers is the level of education, number of livestock and income. So that the necessary effort to improve the level of education, access to farm and to training (Musaba, 2010).

Ghosh *et al*., (2005) states that the socio-economic factors include age, education, family income, land ownership, availability of resources, and socio-psychological factors are attitudes, motivation, risk, culture, and institutional real impact on farmers' technology adoption. Ismail and Dianpratiwi (2008), explains that the perception of the attitude of farmers, farmer knowledge and skills related to a technological innovation influence the adoption of technology. Responsiveness of farmers to new technologies is very dependent on the attitudes, behaviors and knowledge level of farmers. According to Lawa *et al* (2007), factors affecting technology adoption by farmers is the socio-economic characteristics of farmers, institutional factors, and characteristics of technology, use of production facilities, the cost of technology, the technical implementation of production technology, risk, communication networks, extension agents, and technical efficiency. The influence of each factor varied magnitude and direction determination is a decision of a technology adoption.

Based on a review of some results of previous studies, it is to create a model of sustainable farming systems to realize the sustainability of food resources it would require the implementation of the concept of eco-farming is based on the socio economics of farmers, especially small farmers with limited land ownership level. The following conceptual frameworks for the development of eco-farming model (Figure 2).

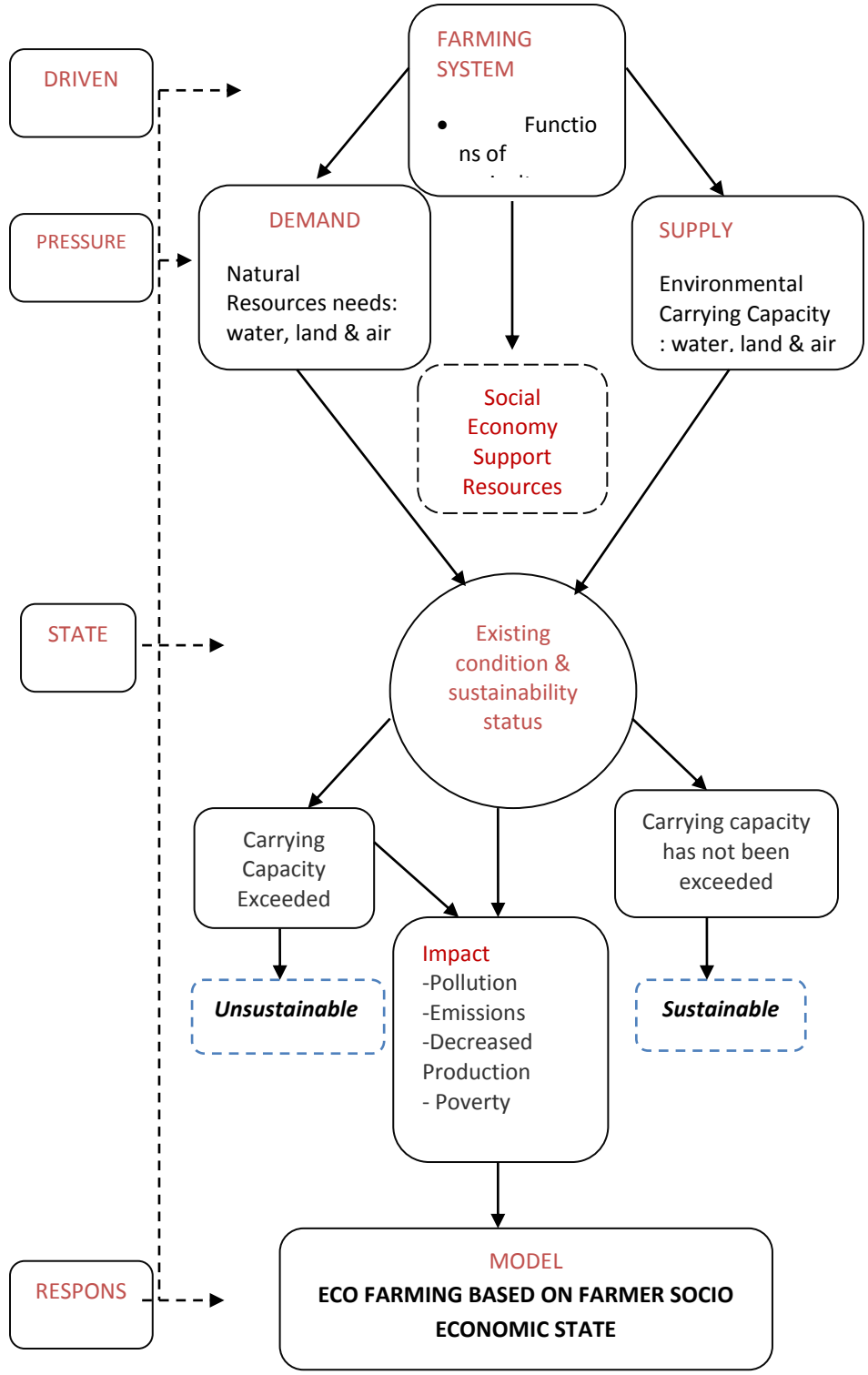

Figure1. Conceptual frameworks of Eco-farming model based on farmers' socio economics

Conclusion

Farmers' socio-economic factors play an important role in determining the characteristics of the farm, therefore it is necessary to study the existing conditions related to farming systems at farm household level, as well as the status of sustainability analysis to make the right eco-farming model for food resources sustainability.

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Utilization of Yard to Increase Household Income and Food Security

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Abstract

Food security is generally interpreted as a guarantee that every resident (anyone) in a country (anywhere, anytime) adequate food and nutritional needs as a key condition to achieving the health and welfare (Soekirman, 1996). The existence of dry land in the countryside in the form of cultivated land, gardens, fields and yards and empirically demonstrate the distribution of dry land tenure and ownership more evenly so. Utilization of the yard as a planting medium, aquaculture and breeding can increase the availability of food sources and increase household income sources, is expected to malnutrition and low income are a problem in the countryside will be overcome. Our study in collaboration with the National Food Security in West Java showed that both urban and rural areas are potential to be developed with the use of yard area by planting different crops or enterprises that have economic value. Results of the study showed that all four areas in West Java that used the yard received benefits for households to improve household consumption and household income. For urban areas, choice of plants are generally with the orientation to be more commercial. As for rural areas, the choice of plants or businesses in utilizing the yard, tend to be limited, although the thinking is economically good choice of plants or business and the orientation of the crop has begun to grow in areas other than rural food insecurity. Food insecurity in the village by utilizing their yards is still low of interest.

Keywords: resilience, food, yard, nutrition

Introduction

The existence of land in addition need to be maintained as a limited resource sustainability also requires efforts to improve land use to obtain or make alternative sources of household income of rural farmers. Development program in order to realize food security and nutrition, including through the use of yard area, diversification of food and nutrition and increased movement of food security of rural communities. Yard area is the land around the house that can be used for farming activities of economic value but do not interfere with the function of aesthetic grounds. Land use need to be integrated handling yard so as to keep the beauty and scenery, the source of family nutrition, pharmacy and life as a barn or living plants, including vegetables, fisheries and livestock. Utilization of the yard as a planting medium, aquaculture and breeding can increase the availability of food sources and sources of income generation tangga.Pemenuhan home food sufficiency is not only an obligation, whether moral, social and legal (including compliance with human rights), but also the establishment of investment human resources in the future. Besides fulfilling food security is also a prerequisite for the fulfillment of other basic rights such as education, employment and so on.

It is therefore necessary to review the implementation of the quality of consumption through the utilization of the yard with a focus on the study include: picture of the utilization of the yard, the picture of the culture or people's habits in using their yards for productive agricultural land use as well as an ideal model to the front yard to support the realization of food security in every home ladder.

The aim of this research is reviewing the implementation of the utilization of the yard as a source of income for food and income needs of households, and also recommend the use of a model compound in various base areas.

Materials and Methods

Assessment Methods

In general, the methods used in this study is descriptive method with survey techniques, methods and techniques used to gather primary and secondary data. Primary data obtained from the field with interviews of people in the village Rawan Food, Village Industry Base, Base Village Beach, Village Base Area Agriculture And Village Pantura in all four regions of West Java and officials from the offices, agencies and related institutions sampled, and observation. Secondary data obtained from the relevant agencies. Furthermore, primary data and secondary data obtained is processed, analyzed and described descriptively.

Place of Research

In the data collection techniques that will be sourced from the Food Rawan village data base is then sorted by region and then purposively sampled as follows see Table 1.

Table .1 Implementation Quality Study Area Consumption Through the use of compound

No	Region (Base)	Regency	Sub District	Village
1	Industrial	Purwakarta	Tegalwaru	Batunumpang
				Warung Jeruk
2	Agriculture	Majalengka	Dawuan	Pasirmalati
			Banjaran	Banjaran
3	Beach	Garut	Cikelet	Cigadog
			Cibatu	Sukalilah
4	Pantura	Subang	Pamanukan	Sukasari
5	Urban	Depok	Pancoran Mas	Kelurahan Cipayung Jaya

Data Analysis Phase

Assessing the Implementation of Quality Food Consumption Communities

To assess the quality of a society of consumption can be seen from the level of diversification of food and a good level of food sufficiency and food sources of carbohydrate source of protein.

$$\frac{\sum \text{kg food consumption}}{100 \text{ gram}} \times \text{nutritional content of food}$$

To determine whether sufficient levels of food consumption and food source of carbohydrate food source of protein in the household used the quality score calculation PPH (Hope Food Pattern) according to FAO - Rapa (1989) as follows:

$$SPPH = \sum_{i-1}^{n} E \times B$$

Identify utilization

To identify the use of the yard, the picture of the culture or people's habits in using their yards for productive agriculture can be analyzed descriptively by looking keragaan yard land use and calculate the income earned from the use of the yard.

Develop land use model ideal yard looking ahead to support food security in any realization of household.

In formulating the ideal model of land use to the front yard to support the realization of food security in every household, ie by evaluating the results of interviews with communities, stakeholders related to the utilization of the yard. Conclusion of the interview obtained by using the method of PRA (Participation Rural Appraisal).

Results and Discussion

Utilization yard in an urban area (Sample: Depok, Pancoran Mas District, Village Cipayung Jaya)

Utilization model yard

One of the groups who use the garden as a source of food and income sources for the fulfillment of nutrition and household food in West Java is a community in Pancoran Mas Village, District Pancoran Mas, Depok City incorporated in Kali Slippery Farmers group who use the yard to develop plants Plants annual leatherback form. Starfruit plants developed in the City area has been used as a Icon Depok Depok City as "Carambola Gods", a type of fruit is very distinctive so it is suitable to agro-climate conditions in the area. As the star fruit tree species are very suitable to be developed annually in the yard because the trees that shade memeiliki characteristics serves as a protection from the sun but can produce fruits that have high economic value. Currently Pancoran Mas Village community incorporated in this Kalilicin Farmers Group already has an extensive planting that if the total area of approximately 10 ha with a membership of approximately 25 people. From the information obtained through interviews with the Chairman of Farmers Group from a single tree that has productive starfruit can be produced fresh fruit for each harvest as much as 100 kg, with an average price of Rp. 5.000, - / kg of the obtained revenues of approximately Rp. 500.000, -. The average tenure as a star fruit tree crops pekarang each household is 2-3 trees per household to obtain a monthly income of about 1.5 million dollars to 15 million.

Adequacy level of food carbohydrate and protein sources

Table 2. Criteria for Adequacy of Food Consumption And Food Sources of Energy and Protein In Households

No.	Nutritional content	Factual Consumption	Consumption advice	Criteria
1.	Energy	3270,13 Cal/cap/day	2.200 Cal/cap/day	Enough
2.	Protein	182,94 gr/cap/day	50 gr/cap/day	Enough

Source: Results of Analysis, 2009

Diversification of Food Consumption

Table 3. Achievement Score Food situation Pattern Hope (PPH) Farmer Household Respondents

No	Food Group	Weight	Calorie	Range (%)	PPH Score	PPH Score Maximum
1	Grain	0,50	2010,21	61,47	30,74	25,00
2	Root	0,60	28,31	0,87	0,52	2,50
3	Animal Food	2,00	301,42	9,22	18,43	24,00
4	Oils and Fats	0,50	401,60	12,29	6,14	5,00
5	Fruits / seeds	0,50	80,56	2,46	1,23	1,00
6	Nuts	2,00	69,74	2,13	4,26	10,00
7	Sugar	0,50	187,33	5,73	2,86	2,50
8	Vegetables and fruit	5,00	190,95	5,84	29,20	30,00
	Number		**3270,13**	**100**	**93,39**	**100**

Source: Results of Analysis, 2009

Revenue from use yard

The average additional income per month in getting the yard in the village Cipayung Raya Depok Rp. 2770021 This is because the high sales in the town red Guava depok exports that reached even the average income derived by a resident of the town of Depok enough. In addition to the yard of a potential red Guava is high, other commodities commodities are planted in the yard depok also has considerable potential to be developed as potential leatherback is pursued by a group of farmers whose income ranges between 500 thousand to 15 million.

Use of Yard Areas Prone to Food (Sample: Majalengka, Dawuan District, Village Pasirmalati)

Utilization model yard

Utilization of the yard in the Village area is in dire need of food Prone creativity of every household, every household can not manage their yards as well. In the Village District

Pasirmlati this Dawuan yard utilization is still lacking. With the average area of the yard between 17 x 14 m2, in general, households do not optimally utilize the land garden. Most households use their yards only planted one or two types of plants, which is planted with mango and a kind of ornamental plants. Besides the existing courtyard used by poultry and sheep and ducks. The reason for the lack of utilization of the yard is because of the availability of water.

Level of food sufficiency carbohydrate and protein sources

Table 4. Criteria for adequacy of food consumption and food sources of energy and protein in households

No.	Nutritional content	Factual Consumption	Consumption advice	Criteria
1.	Energy	2281.59 Cal/cap/day	2.200 Cal/cap/day	Enough
2.	Protein	75.89 gr/cap/day	50 gr/cap/day	Enough

Source: Results of Analysis, Year 2009

Diversification of Food Consumption

Table 5. Achievement score food situation pala hope (pph) farmer household respondents

No.	Food Group	Weight	Calorie	Range (%)	PPH Score	PPH Score Maximum
1	Grain	0.50	1808.36	79.26	39.63	25.00
2	Root	0.60	82.08	3.60	2.16	2.50
3	Animal Food	2.00	75.40	3.30	6.61	24.00
4	Oils and Fats	0.50	151.94	6.66	3.33	5.00
5	Fruits / seeds	0.50	22.44	0.98	0.49	1.00
6	Nuts	2.00	32.71	1.43	2.87	10.00
7	Sugar	0.50	75.73	3.32	1.66	2.50
8	Vegetables and fruit	5.00	32.94	1.44	7.22	30.00
	Number		2281.59	100.00	63.96	100.00

Source: Results of Data Processing, Year 2009

Revenue from Use Yard

The average additional income per month in getting the yard in the village of Pasirmalati Rp. 77 thousand. For mango, coconut and jackfruit 30% self consumption and 70% sold. As for the goats and ducks to 100% sold and consumed 50% chicken and 50% for sale. The average results of the village yard Pasirmalati derived from annual crops such as mango, jackfruit, coconut and effort goats, ducks and chickens.

Utilization of Agricultural Base Yard Area (Sample Majalengka, District Banjaran, Village Banjaran)

Utilization model yard

Banjaran Banjaran Village District is the village that successfully utilize rumahtangganya yards. With this success has made the Village Banjaran election success successful villages in West Java level. People in the village population Banjaran including creative use of the yard. Spacious yard with an area average of 3 x 12 m2 planted with various crops such as cengek, tomato, scallion, sosin, spinach, kale etc. Additionally yard land used for sheep and cattle business as well with the chicken business carp, tilapia and carp.

Levels and adequacy of food sources carbohydrates protein

Table 6. Criteria for adequacy of food consumption and food sources of energy and protein in households

No.	Nutritional content	Factual Consumption	Consumption advice	Criteria
1.	Energy	2774,79 Cal/Cap/day	2.200 Cal/Cap/day	Enough
2.	Protein	115,01 gr/cap/day	50 gr/cap/day	Enough

Source: Results of Analysis, Year 2009

Diversification of food consumption

Table 7. Achievement score food situation pattern hope (pph) farmer household respondents

No.	Food Group	Weight	calorie	Range (%)	PPH Score	PPH Score Maximum
1	Grain	0.50	1948.93	66.54	33.27	25.00
2	Root	0.60	107.63	3.67	2.20	2.50
3	Animal Food	2.00	252.87	8.63	17.27	24.00
4	Oils and Fats	0.50	346.30	11.82	5.91	5.00
5	Fruits / seeds	0.50	45.94	1.57	0.78	1.00
6	Nuts	2.00	55.04	1.88	3.76	10.00
7	Sugar	0.50	115.34	3.94	1.97	2.50
8	Vegetables and fruit	5.00	56.74	1.94	9.69	30.00
	Number		2928.77	100,00	71.63	100.00

Source: Results of Data Processing, Year 2009

Revenue from use of yard

The average additional income per month in getting daripekarangan Rp. 431 500. Commodities grown in the garden of perennial crops such as mango, banana and cloves used for planting crops other than annual, as well as to plant different crops, including shops and pharmacies to live lives of 80% is consumed for the purposes rumahtangganya

and 20% sold, in addition to their yards as well in use for pond tilapia, carp gold and partly used for cattle sheep or rabbits.

Use of yard areas Pantura (Sample-Subang District, District Sukasari, Village Sukasari)

Utilization model yard

In the Village District Sukasari Sukasari average tenure of the population is 82.1 m2. Households in the Village District Sukasari already using land yard. Land management is a simple compound. Households used their yards by planting both seasonal and annual crops. In addition to the business of sheep. In detail as follows:Plant annuals planted the beans,eggplant, peppers and chili. Whereas annual crops are planted are mango, banana and tamarind. Watering in this village isirrigation**.**

Level of food sufficiency carbohydrate and protein sources

Table 8. Criteria for adequacy of food consumption and food sources of energy and protein in households

No.	Nutritional Content of Consumption	factual consumption	Suggested	Criteria
1.	Energy	2774,79 cal/kap/day	2.200 cal/kap/day	enough
2.	Protein	115,01 gr/kap/day	50 gr/kap/day	enough

Source: Results of Analysis, Year 2009

Diversification of food consumption

Table 9. Food Nutmeg State Achievement Scores Hope (PPH) Farmer Household Respondents

No.	Food Group	Weight	calorie	Range (%)	PPH Score	PPH Score Maximum
1	Grain	0.50	1957.50	70.55	35.27	25.00
2	Root	0.60	68.01	2.45	1.47	2.50
3	Animal Food	2.00	186.06	6.71	13.41	24.00
4	Oils and Fats	0.50	281.88	10.16	5.08	5.00
5	Fruits / seeds	0.50	85.48	3.08	1.54	1.00
6	Nuts	2.00	42.75	1.54	3.08	10.00
7	Sugar	0.50	97.52	3.51	1.76	2.50
8	Vegetables and fruit	5.00	55.60	2.00	1.76	30.00
	Number		2774.79	100,00	71.63	100.00

Source: Results of Data Processing, Year 2009

Revenue From use yard

The average income of the yard in the Village sukasari Rp. 295 500 plants with the commodity form of annual and seasonal crops combined with sheep. Crops are planted in the form chili, chillies, and eggplant, while the annual crop of mango and tamarind.

Use of Yard Areas Prone to Food Village(Sample-Purwakarta district, District Tegalwaru, Village Batu Tumpang)

Utilization model yard

Utilization of the yard in rural food insecure areas are in desperate need of creativity from every household, every household can not manage their yards as well. In the Village District Batutumpang this Tegalwaru yard utilization is still lacking. With an average of 30 m2 yard area, in general, households do not optimally utilize the land garden. Most households use their yards only planted with 1 or 2 types of plants, which is planted with mango and a kind of ornamental plants. Besides the existing yard used by livestock of sheep and ducks. The reason for the lack of utilization of the yard is because of the availability of water.

With the assistance of independent action program of food then there are additional efforts that attempted sheep in the yard, because there are several affinity groups that utilize the yard with cattle sheep business.

Level of food sufficiency carbohydrate and protein sources

Table 10. Criteria for adequacy of food consumption and food sources of energy and protein in households

No.	Nutritional content	Factual Consumption	Consumption advice	Criteria
1.	Energy	2002,56 Cal/Cap/day	2.200 Cal/Cap/day	Enough
2.	Protein	70,85 gr/cap/day	50 gr/cap/day	Enough

Source: Results of Analysis, Year 2009

Diversification of food consumption

Table 11. Circumstances achievement scores on food expectations (pph) farmer household respondents

No.	Food Group	Weight	Calorie	Range (%)	PPH Score	PPH Score Maximum
1	Grain	0.50	1203.43	60.09	30.05	25.00
2	Root	0.60	41.61	2.08	1.25	2.50
3	Animal Food	2.00	32.54	1.62	3.25	24.00
4	Oils and Fats	0.50	64.43	3.22	1.61	5.00
5	Fruits / seeds	0.50	7.69	0.38	0.19	1.00
6	Nuts	2.00	16.68	0.83	1.67	10.00
7	Sugar	0.50	617.06	30.81	15.41	2.50
8	Vegetables and fruit	5.00	19.11	0.95	4.77	30.00
	Number		2928.77	100,00	58.19	100.00

Source: Results of Analysis, Year 2009

Revenue from use of yard

Revenue earned per month in getting the yard by an average of Rp 62,200. Commodities grown in the garden of perennial crops such as mango, banana, coconut, and banana. While the crops are planted in the form of onion leaves, cassava leaves, peanuts and soybeans and partly used for cattle sheep.

Utilization yard in Industrial Area (Sample-Purwakarta district, District Tegalwaru, Warung Jeruk Village)

Utilization model yard

Communities in Warung Jeru Village, including the creative community in the use of yard area with land ownership on average 137.5 m2 planted with a variety of annual and seasonal crops. Annual crops are planted in the shop like orange mango, rambutan, papaya, and banana. While the seasonal crops such as chili, sosin, spinach, kale, cassava and various seasonings sometimes referred to tabulakar (herbs in the garden) like ginger. In addition land is also used for business yard goats and duck are also with the business goldfish. The reason for the lack of utilization of the yard is because of the availability of water.

Levels and adequacy of food sources carbohydrates protein

Table 12. Criteria for Adequacy of Food Consumption And Food Sources of Energy and Protein In Households

No.	Nutritional content	Factual Consumption	Consumption advice	Criteria
1.	Energy	2471,98 Cal/Cap/day	2.200 Cal/Cap/day	Enough
2.	Protein	82,62 gr/cap/day	50 gr/cap/day	Enough

Source: Results of Analysis, Year 2009

Diversification of Food Consumption

Table 13. Circumstances Achievement Scores On Food Expectations (PPH) Farmer Household Respondents

No.	Food Group	Weight	Calorie	Range (%)	PPH Score	PPH Score Maximum
1	Grain	0.50	1990.71	80.53	40.27	25.00
2	Root	0.60	37.97	1.54	0.92	2.50
3	Animal Food	2.00	86.43	3.50	6.99	24.00
4	Oils and Fats	0.50	171.81	6.95	3.48	5.00
5	Fruits / seeds	0.50	22.44	0.91	0.45	1.00
6	Nuts	2.00	36.35	1.47	2.94	10.00
7	Sugar	0.50	83.79	3.39	1.69	2.50
8	Vegetables and fruit	5.00	42.48	1.72	8.59	30.00
	Number		2471.98	100,00	65.34	100.00

Source: Results of Analysis, Year 2009

Revenue from use of yard

Revenue earned per month in getting the yard by an average of Rp 124,000. Commodities grown in the garden of perennial crops such as mango, banana, coconut, and banana. While the crops are planted in the form of onion leaves, cassava leaves, peanuts and soybeans and partly used for cattle sheep.

Use of Yard Areas Agriculture Industry (Sample-Garut District, District Cibatu, Village Sukalilah)

Utilization model yard

Cibatu Sukalilah Village District is the village that successfully utilize rumahtangganya yards, and managed to become the national champion of the successful utilization of the yard.

People in the village population Sukalilah including creative use of the yard. Spacious yard with an area average of 108.5 m2 planted with a variety of seasonal crops such as cengek, tomato, scallion, sosin, spinach, kale is also an annual crop of banana and papaya. Additionally yard land used for sheep and cattle enterprises chicken.

Levels and adequacy of food sources carbohydrates protein

Table 14. Criteria for Adequacy of Food Consumption And Food Sources of Energy and Protein In Households

No.	Nutritional content	Factual Consumption	Consumption advice	Criteria
1.	Energy	2325,52 Cal/Cap/day	2.200 Cal/Cap/day	Enough
2.	Protein	61,20 gr/cap/day	50 gr/cap/day	Enough

Source: Results of Analysis, Year 2009

Diversification of Food Consumption

Table 15. State Achievement Score On Food Expectations (PPH) Farmer Household Respondents

No.	Food Group	Weight	calorie	Range (%)	PPH Score	PPH Score Maximum
1	Grain	0.50	1239.43	53.30	26.65	25.00
2	Root	0.60	36.40	1.57	0.94	2.50
3	Animal Food	2.00	101.50	4.36	8.73	24.00
4	Oils and Fats	0.50	154.63	6.65	3.32	5.00
5	Fruits / seeds	0.50	30.77	1.32	0.66	1.00
6	Nuts	2.00	22.86	0.98	1.97	10.00
7	Sugar	0.50	711.85	30.61	15.31	2.50
8	Vegetables and fruit	5.00	298.08	1.21	6.04	30.00
	Number		2471.98	100,00	63.61	100.00

Source: Results of Analysis, Year 2009

Revenue from use of yard

Revenue earned per month in getting the yard by an average of Rp 199,750. Commodities grown in the garden of perennial crops such as mango, banana and papaya. While the crops are planted in the form sosin, kale, celery and garlic spinach leaves. The results of the annual crops of the yard 100% consumed by the household. While cattle are cultivated are goats.

Utilization in the Village Yard Prone Food (Sample-Garut District, District Cikelet, Village Cigadog)

Utilization model yard

Utilization of the yard in rural food-insecure rural areas Cigadog still lacking. It is in desperate need of creativity from every household, every household can not manage their yards as well. In the village is the utilization of the yard is still lacking. With the average area of the yard 2 mx 3 m, the average household does not optimally utilize the land garden.

With the assistance of independent action program of food then there is the addition of the business use of his yard with a cultivated sheep in the yard, because there are several affinity groups that utilize the yard with cattle sheep business. For More Clearly can be seen in Table 16 below:

Level of food sufficiency carbohydrate and protein sources

Table 16. Criteria for Adequacy of Food Consumption And Food Sources of Energy and Protein In Households.

No.	Nutritional content	Factual Consumption	Consumption advice	Criteria
1.	Energy	2173,19 Cal/Cap/day	2.200 Cal/Cap/day	Enough
2.	Protein	86,16 gr/cap/day	50 gr/cap/day	Enough

Source: Results of Analysis, Year 2009

Diversification of food consumption

Table 17. State achievement score on food expectations (pph) farmer household respondents

No.	Food Group	Weight	Calorie	Range (%)	PPH Score	PPH Score Maximum
1	Grain	0.50	1789.93	82.36	41.18	25.00
2	Root	0.60	60.91	2.80	1.68	2.50
3	Animal Food	2.00	63.76	2.93	5.87	24.00
4	Oils and Fats	0.50	98.51	4.53	2.27	5.00
5	Fruits / seeds	0.50	30.77	1.42	0.71	1.00
6	Nuts	2.00	28.77	1.32	2.65	10.00
7	Sugar	0.50	68.98	3.17	1.59	2.50
8	Vegetables and fruit	5.00	31.57	1.45	7.26	30.00
	Number		2173.19	100,00	63.20	100.00

Source: Results of Analysis, Year 2009

Revenue yard in the Village Cigadog

Revenue from use of the yard in the Village Cigadog around Rp. 58 750 thousand per month, 80% of his yard on his own consumption, 20% for sale.

Conclusions

1. Study results to-4 region in West Java, in the courtyard in urban land use and rural differences in the choice of plants and orientation of the crop. For urban areas, generally been carried out the choice of plants or businesses that are more economical or profitable and the orientation of the crop more commercially oriented or for sale. As for rural areas, the choice of plants or businesses in utilizing the yard, tend to be limited, although the thinking is economically good selection of plants or business genius and the orientation of the crop has begun to grow in areas other than rural food insecurity. Food insecurity in the village of interest in utilizing their yards is still minimal.
2. Sufficient levels of carbohydrate and protein food source which is calculated based on the criteria Widyakarya National Food and Nutrition (2004) showed that households in the four regions of West Java to meet the recommended intake for energy and protein and belongs to the criteria fairly.
3. Diversification of food consumption, especially food source of carbohydrates and proteins are still few in number. This is indicated by Hope Food Pattern Score (SPPH) achieved was still below the maximum score of PPH. Score PPH has been achieved is still dominated by the grains food group, this show is still dependent on food staples of the four regions of West Java, on rice as the main carbohydrate food sources.

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Harmonizing Agriculture, Forests and Fishery Management in the Design of REDD+ in Small Islands of Kepulauan Aru Regency, Maluku Province

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Abstract

Generally, ecology on big and small islands is different. In case of small islands ecology is more fragile, implementation of sustainable agriculture, forest and fishery should be handled carefully. The research regarding management of coastal area in small islands and its impact for sustainable development in Regency of Kepulauan Aru, Maluku Province was done. The results showed that the natural resources of Kepulauan Aru Regency is a very potential regency for fishery development and fishery sector has become as leading sector and prime mover for economical development in this region. Unfortunately, the coastal area, low land and high land have damaged by forest degradation, deforestation and others activities. In this case, forest quality can be improved through forest conservation action in Reducing Emissions from Deforestation and forest Degradation (REDD+) Project. The Special Design of REDD+ Project in Small Islands is needed and should be done by harmonizing Agriculture, Forests and Fishery management with involvement of all stakeholders in this region.

Keywords: deforestation, forest degradation, REDD+, sustainable agriculture.

Introduction

In general, ecology on small island is very specific and there is difference between small island and big island. Dahl (1998) states the ocean covers more than two thirds of our planet, and small island developing States are generally somewhere in the middle of it. Their environmental priorities concern the islands themselves and their immediate coastal areas, where the balance of population and resources is critical for the future. From the perspective of the island countries, the ocean is still an unlimited resource relative to the present scale of their human impacts. The effects on the ocean of the great centres of population and industry in continental countries are much more significant.

Natural resources management in small islands should be done fully attention and properly. Widespread environmental problems should be minimized i.e. the safe disposal of liquid domestic wastes (human wastes and urban sewage); the damage or destruction of productive coastal resources and fisheries; the steady reduction in forest cover (timber extraction, shifting cultivation and land clearing for agriculture); conflict of interest on land use and land tenure. In any case, we have to manage natural resource in small island wisely. In this region, efficient use must be made of all available land to meet the needs of the people for water, food, building materials and reasonable quality of life, and to maintain the functioning of natural systems on which all these depend. This requires comprehensive planning and careful allocation of land to the most appropriate use or combination of uses.

In the last two decades, the most threats of sustainable natural resource management in small islands is global warming. Global warming is a gradual warming of Earth's atmosphere reportedly caused by the burning of fossils fuels and industrial pollutant. It is caused by gaseous that trap suns heat in Earth's atmosphere. Primarily carbon dioxide and CFC

gaseous are to blame. In general, adverse effects of global warming on small islands is slight increase in sea levels, increases the temperature of sea and blamed for severe climatic change. Many efforts to handle global warming, one of them is REDD program. It is estimated that greenhouse gas emissions from deforestation and degradation contribute up to 20% of global emissions. REDD stands for 'reducing emissions from deforestation and degradation', and is an important part of global policies to address climate change. The term 'REDD-plus' or 'REDD+' is now also used frequently. REDD+ is similar to REDD, but instead of just covering deforestation and degradation, it includes other activities, such as the sustainable management of forests and the enhancement of forest carbon stocks (Graham, 2010). Therefore harmonizing agriculture, forest and fishery management in the design of REDD+ in small island is important.

Materials and Methods

Methods of the study are environmental policy study and literature study. Environmental policy is any [course of] action deliberately taken [or not taken] to manage human activities with a view to prevent, reduce, or mitigate harmful effects on nature and natural resources, and ensuring that man-made changes to the environment do not have harmful effects on humans (McCormick, 2001). Environmental policy study focuses on problems arising from human impact on the environment, which retroacts onto human society by having a (negative) impact on human values such as good health or the 'clean and green' environment. There are many environmental issues in the world. In general, environmental issues addressed by environmental policy include natural resource i.e. air and watershed management, water pollution, forest with its biodiversity protection, land conservation, coastal area protection, ecosystem management, etc. Material of the study are information regarding natural resources management which available on research report, annual report, text books and some articles from website. The site study is presented in Figure 1.

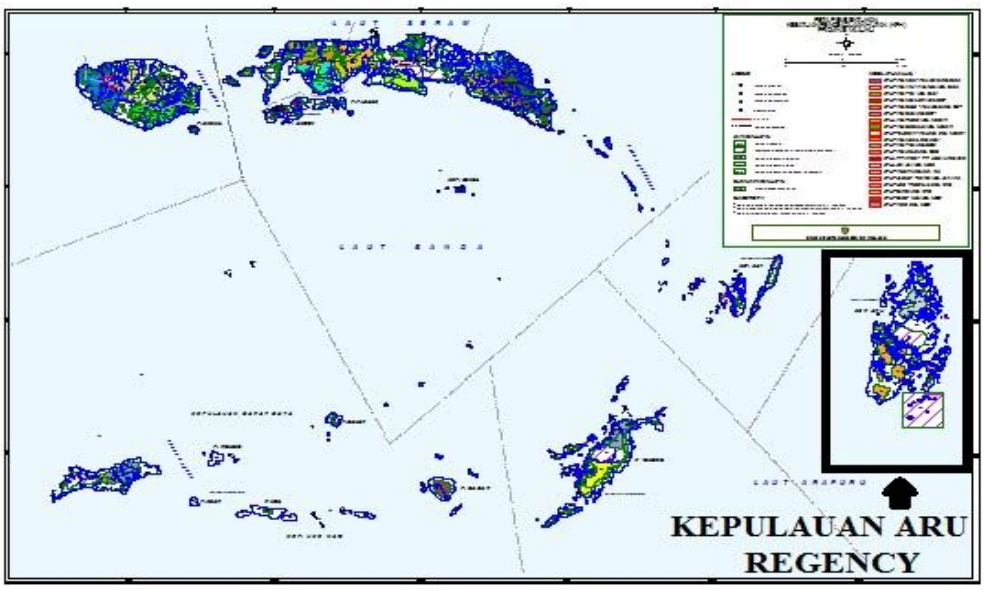

Figure 1. The site study
Source: Maluku Provincial Forestry, 2009

Results and Discussion

Forestry Management in Kepulauan Aru Regency

There are two Forest Management Units (FMU) in Kepulauan Aru Regency, namely Wokam and Kobror – Trangan. Wokan FMU area is 61,012 Ha (production forest 57,992 Ha and protected forest 3,020 Ha) with watershed area namely Wai Gaimar 69,160 ha. This watershed is rather and very critical 17,866 Ha (25.83 %). In general, forest resources in this FMU is good where primary and secondary forest in large area and open area or agriculture activities in forest area still limited. This forest can be functioned as catchment area and it is very important for local community to use water for their life and also for development of traditional agriculture and pastures.

Kobror-Trangan FMU area is 117,614 Ha (production forest 114,781 Ha and protected forest 2,833 Ha) with 4 watersheds i.e. Koba 3,910 Ha with rather and very critical area is 3,168 Ha (81.02 %), Waekoba 10,095 Ha with rather and very critical area is 1,358 Ha (13.45 %), Trangan 2,024 Ha and all of its watershed is very critical area 2,024 Ha (100 %) and Kobror 2,024 Ha with rather and very critical area is 15 Ha (0.74 %). Protected forest lies in all of small islands with dominance of forest mangrove and savana. Open area or critical land usually lies in bigger island in southern and western FMU. This critical land is caused by illegal cutting, shifting cultivation and forest fire, etc (Maluku Provincial Forestry, 2009).

In general, land cover of rather and very critical watershed is indicated with low density forest and dominated with shrubs, bush, grasses and open area. Based on sattelite imagery interpretation, mainly by using color composite of Landsat ETM+ band 542 year 2008, watershed area with rather and very critical in Aru archipelago can be recoqnized easily. Open area without vegetation is indicated with color of red to brown, shrubs, bush and grasses with soft green and dense forest with deep green. Furthermore, by using NDVI (normalized difference vegetation index) approach or ratio between NIR-RED with NIR+RED wavelengths, the land cover can be recoqnized well. The higher NDVI values indicated more abundant, greener vegetation like dense forest and lower NDVI values indicated grasses, shrubs, bush, bare soil, water, clouds due to lower reflectance values in the NIR wavelengths. NDVI values for dense forest, rare forest, grass-shrubs-bush, bare soil and water is 0.7-0.9; 0.5-0.6; 0.2-0.5; 0.1-0.2 and 0.0-0.1 respectively (Mardiatmoko, 2009). NDVI are indicators of intensity of biomass (JARS, 1993) and it is important for REL (reference of emission level) establishment mainly to support REDD+ project.

The main habitat at coastal area in Pulau-pulau Aru and Aru Utara Sub district is ecosystem of forest mangrove, coral reef, seagrass and algae. Composition of forest mangrove is Rhizophoraceae, Sonneratiaceae, Maliaceae, Myrsinaceae, Sterculiaceae, Acanthaceae and Arecaceae. Sea weeds distribution is separated randomly in surrounding of Aru archipelago.

Agriculture Management in Kepulauan Aru Regency

Potential areas for agriculture in Kepulauan Aru Regency is 1,200 ha in Kapron island. Based on ecological small island, sector development of agriculture in Kepulauan Aru Regency is not focused on wetland paddy. There is paddy production but from dryland which average area harvested dryland paddy is very limited 20 ha/year. Area harvested of food crop in year 2008 as follows maize 75 ha, cassava 243 ha, sweet potatoes 91 ha, peanuts 45 ha, mung beans 43 ha, soybeans 0 ha, taro 88 ha. Beside this crops, area harvested, production

and yield rate of low land vegetables can be described as follows*:* chili area harvested 7 ha, production 6 ton, yield rate 8.57 kw/ha; cucumber 2 ha, 7 ton, 35 kw/ha; egg pant 5 ha, 19 ton, 38 kw/ha; pulses 2 ha, 6 ton, 30 kw/ha; tomatoes 1ha, 3 ton, 30 kw/ha. The crops, area harvested, production and yield rate of low land fruits as follows: avocados area harvested 0.35 ha, production 15 ton, yield rate 15 kw/ha; mangoes 25.57 ha, 2,661 ton, 26.61 kw/ha; pepayas 1.05 ha, 364 ton, 36.40 kw/ha; bananas 5.28 ha, 649 ton, 64.9 kw/ha, pineapple 0.07 ha, 34 ton, 34 kw/ha and others (BPS-Statistics Indonesia, 2009).

Total area of agriculture activities in Kepulauan Aru Regency is not large and there is no problem regarding land use and land tenure. The most important thing is agriculture area expansion for the future should be planned well and wisely.

Fisheries Management in Kepulauan Aru Regency

Potential areas for fisheries and marine tourism in Kepulauan Aru Regency is high. Based on ecological small island, sector development of fisheries in Kepulauan Aru Regency is not focused on land area and production of inland aqua culture and open water fisheries but in coastal area and its surroundings area. According to Kakisina *et al* (2010) Kepulauan Aru Regency lies in Aru Sea and Arafura Sea and has high fisheries resources potency with its biodiversity. Therefore fishery sector has become as leading sector and prime mover for economical development in this region. Total marine water area which can be used as cultivation area is 3,311.73 Km^2 (seagrass 1,996 Km^2, fish 667 Km^2, sea cucumber 371 Km^2, cockle shells and bivalve 278 Km^2). In year 2009, only 506,6947 Km^2 (15.30%) of these area is used as cultivation activities i.e. Kobror, Koba, Maikor islands.

Number of fishery households is 969 households and non-powered fishing boats 231 units. Fish production and its value in year 2008 as follows: prawn 1,291.7 ton, IDR 17,682,380,000,-; skipjack 203.9 ton, IDR 1,223,572,000,-; indian mackarela 591.1 ton, IDR 1,182,180,000,- and others 8,179.5 ton, IDR 33,108,297,000,- (BPS-Statistics Indonesia, 2009). Kepulauan Aru Regency has many coastal area with beautifull beach and very potensial to develop marine tourism or ecotourism, diving clubs, hand line, trolling and sport fishing development.

Some problems regarding coastal area management is the damage or destruction of productive coastal resources and fisheries. Coastal area are destroyed by mining of c type excavation. Coral reefs are destroyed by construction or dredging, pollution, siltation and dynamiting or poisoning for fish. Mangroves are killed off by dredging or filling, or by changing essential patterns of water circulation and salinity. Location of coastal area and coral reefs damage can be found mainly in western and eastern Wokam island and eastern and southern Kobror island.

Community Perception on Natural Resources Management

In general, perception of local community on resources management ussually can be analyzed by using some questioner and interview. After that the survey result should be continued by Focus Group Discussion (FGD). This analysis is used to measure the objectivity of survey results which using questionare and interview. Arrangement of strategy for natural resources management by local government and other stakeholders can be done by SWOT analysis. SWOT analysis is a tool for auditing an organization and its environment. It is the first stage of planning and helps marketers to focus on key issues. SWOT stands for strengths, weaknesses, opportunities, and threats. Strengths and weaknesses are internal

factors. Opportunities and threats are external factors. Establishment of management concept priority for natural resource potency can be used by using AHP (analytical hierarchy process). AHP is an advanced technique that supports decision makers in structuring complex decisions, quantifying intangible factors, and evaluating choices in multi-objective decision situations. It is a comprehensive and rational decision making framework that provides a powerful methodology for determining relative worth among a set of elements (Jayaswal *et al*., 2007). It has particular application in group decision making and is used around the world in a wide variety of decision situations, in fields such as government, business, industry, healthcare, and education (Saaty & Kirti, 2008). Kakisina *et al* (2010) reported that policy priority for management of Kepulauan Aru Regency potency was community and government based management. In this case, the stakeholders agreed that management of natural resources potency in Aru coastal area should be done together by goverment and local community as first priority. Besides this report, strategy of management of coastal area potency on Kepulauan Aru was collaborative strategy which involved all of stakeholders. This collaboration must be implemented based on the role and competency from every stakeholder.

The management integration of forestry, agriculture and fisheries in small islands

Almost of all small islands in Kepulauan Aru Regency has damaged not only in high land but also low land and coral reefs. It must be handled through intensive conservation but financial support from goverment is very limited. In this case, the management integration of forestry, agriculture and fisheries should be done through REDD+ program because developed countries has commitment to support conservation fund. Actually, there is close relationship between agriculture and REDD+. According to Scherr *et al* (2011) the trajectory of agricultural development is a key determinant of the success of REDD+**.** The original model of REDD was to pay landowners the opportunity costs of forgoing forest development. Yet most agricultural production systems offer much higher financial benefits than natural forests. Given low relative carbon prices and the likelihood that they will remain low relative to agricultural options in the future, this payment model has limited scope. It is only relevant in the small set of circumstances where deforestation is principally due to conversion to very-low-value land uses like extensive grazing or small-scale subsistence food production and where governance systems are sufficiently developed to manage payments over the long-term. In most other cases, reducing deforestation requires more than shifting financial incentives at the margin—it requires transformation of the land rights and land use system to harmonize farm and forest policy. Thus successful REDD+ depends less on forestry strategies than on agricultural development strategies that retain and sustain forests. Therefore harmonizing agriculture, forest and fishery management in the design of REDD+ in Kepulauan Aru Regency should be tried. It is possible to be done because the stakeholders agreed that management of natural resources potency must be done together by goverment and local community as first priority.

Conclusions

Small islands ecology in Kepulauan Aru Regency is more fragile, implementation of sustainable agriculture, forest and fishery should be handled carefully and wisely. The stakeholders agreed that management of natural resources potency in Aru coastal area should be done together by goverment and local community as first priority.

The natural resources of Kepulauan Aru Regency is a very potential regency for fishery development and fishery sector has become as leading sector and prime mover for

economical development in this region. Unfortunately, the low land and high land have damaged by forest degradation, deforestation and others activities.
The damage area must be handled through intensive conservation but financial support from goverment is very limited. In this case, the management integration of forestry, agriculture and fisheries should be done through REDD+ program.

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Farmers are sacrificing their health for production of vegetables

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Abstract

Long term negative effect of pesticides on healthy of vegetable farmer has been studied in Bengkulu. Research was conducted to evaluate the effect of pesticides on the health status of farmer in Kabupaten Rejang Lebong, Province of Bengkulu. The result will be used for developing pesticides application and trade regulation by farmers in Bengkulu. One hundred of active farmers had been chosen to evaluate their behavior on handling pesticides on the field and their health status based on their feeling, urine and blood test. The results showed that most of the respondents understand how to handle pesticides at home. On the other hand, they did not apply a proper regulation on the field. The farmers usually contact directly with pesticides during application due to lack of protective cloths such as glasses, masker and gloves. As result, based on their feeling, more than 50% of the farmers felt tired, muscle cramps, dry throat, shortness of breath, headache, dizzy and other digestive system problems. The clinical test confirmed that health problems such as heart and kidney deterioration were also encountered among the farmers. These facts force local government to provide regulation on trade and application of pesticides.

Introduction

Pesticides carry health risk problems among farmers (Anonym, 2005; Forget, 1991). A group of farmers who deals with the pesticides on the field will be poisoned potentially. In developing countries including Indonesia, due to lack of appropriate resources, the spray operators are contact directly to pesticides due to lack standard procedures application such as cloths and its accessories, using leakage of sprayer, etc. Communities are also has potential health risk due to lack of proper handling mechanisms during application of pesticides.

Severe pesticide hazards occur mainly in developing countries. These caused by high application rate and even using the banned ones, all of these due to lack of information and knowledge. The standard procedure of pesticide application is usually disobeyed because of market unavailability or hard to be accessed, non-affordability, or even never think that it is important (Jeyaratnam *et al*., 1987; Forget, 1991). In contrast, farmers in developed countries have been protected by laws, and there is a good coordination between health and agriculture authorities to minimize the negative effects of pesticides on human healthy.

Organophosphates, carbamates, organochlorines and pyrethroids are pesticides commonly used by vegetable farmers in Bengkulu Province of Indonesia. The active ingredients have a potential to poison farmers and in the long term could create cancer (Maroni and Fait, 1993). There is lack of complete evaluation on the effect pesticides on farmer health in developing countries including Indonesia, such as a kind and degree of poisoning, and how many farmers that have been affected. Survey by Jeyaratnam (1990) in 4 (four) different Asian countries indicated that at least 3% of farmers have been affected by pesticides each year. Assuming population of Indonesian farmer of about 25%, it will be large number of farmers can be poisoned by pesticides. In the long term there will be large number of

occurrence of cancer, infertility, nerve damage etc. (Maroni and Fait, 1993). In coping with the negative effect of pesticides on human health requires knowledge on pesticides toxicology, poisoning symptoms, and developing strategy on preventing or controlling poisoning in the farmer's community (Wesseling et al., 1997; Wesseling et al., 2001).

The aims of the research was to evaluate the pattern of pesticides handling by farmers, to observe the awareness of farmers on the negative effects of pesticides on their healthy and to measure the health status of the spray operators on vegetables farming in Selupu Rejang District.

Materials and Methods

a. Research preparation on pesticides usage in Kabupaten Rejang lebong, Bengkulu Province of Indonesia

The district selection was based on data and map provided by Department of Agriculture Kabupaten Rejang Lebong. The district was the central of vegetable production in the area of research. The research was conducted in Selupu Rejang district, regency of Rejang Lebong, Bengkulu Province of Indonesia, from August to November, 2010. Selupu Rejang district was one of the agricultural area producing varieties of vegetables to be marketed to Bengkulu, Palembang, Jambi, Lampung, and Pekanbaru.

Survey method was conducted to collect the data, focusing on 4 (four) major issues which have been raised during the last decade in Bengkulu province, i.e. 1) driving force that farmers using pesticides intensively; 2) the understanding of spray operator on procedure of pesticide application and its effect on environment; 3) the knowledge of farmers on pests and its control, and 4) the test on health status of vegetable farmers (especially the spray operator) using blood and urine examination.

b. Research Implementation

Field survey

Primary data was collected based on open questioner distributed to the farmers (respondents) and on clinical test to selected farmers who are acting as spray operators. In order to be understood easily by respondents, the questioner was examined before distribution to the respondents. The researchers have selected 100 (one hundred) of farmers (equal to more than 10% of active farmers in Selupu Rejang district) to become respondents. About 50 (fifty) percents of them were selected at random for clinical test (blood and urine test). As the questioner was using an open question, the researchers were collecting data by direct meeting to the respondents. Meanwhile, clinical test was carried out by selecting 50 spray operators ranging from 20 to 80 years old. The clinical test was conducted by Health laboratory Ashifa at Bengkulu City supervised by one of academic staff of in Medical Study Program of University of Bengkulu.

Variables measured

1. The number of pesticides stored
2. Various kinds of pesticides stored
3. Pesticides application within a week
4. Volume of pesticides application in one day

5. Time spent for each pesticides application
6. Personal Protective Equipment during pesticides application
7. Skin contact during preparation of pesticides
8. Skin contact during pesticides application in the filed
9. Poisoning symptoms as experienced by farmers
10. Health status based on clinical test

Data Analysis

Collected data are not be analyzed statistically but presented in table and figures.

RESULT AND DISCUSSION

General condition

Farmers have intensively used pesticides, including using 2 to 3 different active ingredient in one single application, high dosage and more frequently during high pest population. Spray operators commonly without cloth protection during operation, smoking while spraying, and do not consider wind direction. In addition, the time period between pesticides application and harvest time commonly shorter than the recommendation this potentially increase the pesticide residues to the consumers.

Local government (Pemda Kabupaten Rejang Lebong) plays minimal control on the pesticides trading, therefore there are many kind of pesticides available for farmers. With minimum knowledge on pesticides usage, the farmers tend to apply the pesticides by trial. Meanwhile government officials who are in charge of educating the farmers (Penyuluh Pertanian Spesialist) have little effect on the farmers' behavior due to strong and persuasive sales promotion.

A. The way of farmers handling pesticides at home

Survey results showed that the farmers basically know the procedure of keeping pesticides at home. Figure 1 and 2 explains that about 70% respondent kept and use more than 1 kind of pesticides for preventing and eradicating pests (insecticides), fungus (fungicide), and weeds (herbicside). Fifty percent of respondent know how to keep pesticides in the save place, but about 30% still keep it in the kitchen.

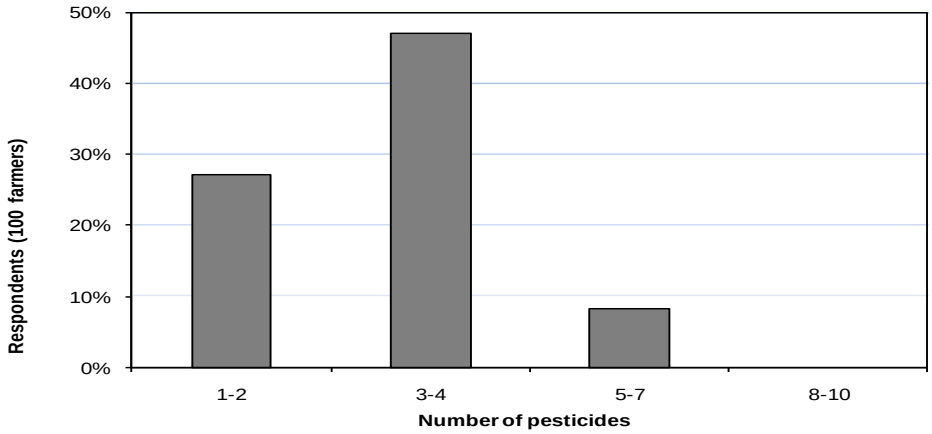

Figure 1 The number of pesticides stored at home

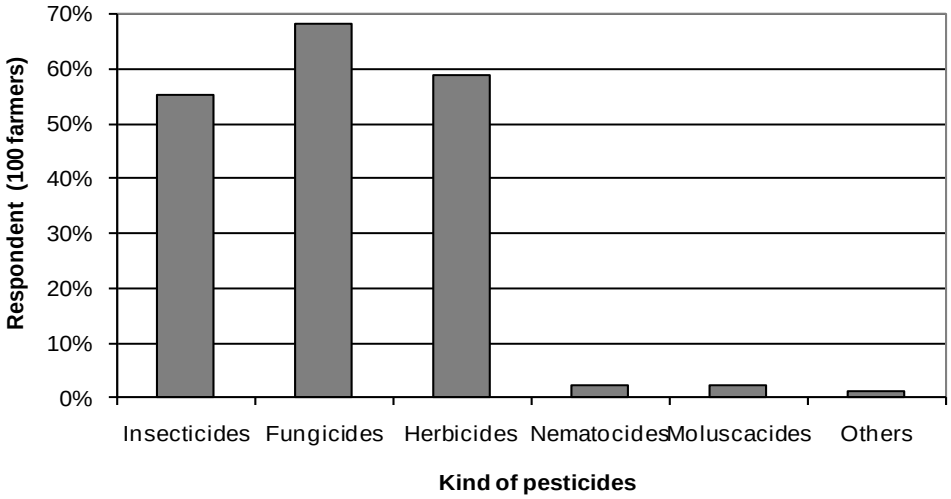

Figure 2. Various kinds of pesticides stored at home

B. Pesticides application pattern

The pattern of pesticides application by farmers can be seen in Figure 3 to 7. In general, about 60% farmers spray the plants less than 5 times a week (Figure 3), but other farmers (40%) spray between 6 to 15 times a week, this is possible as many spray operators also become employee of other farmers.

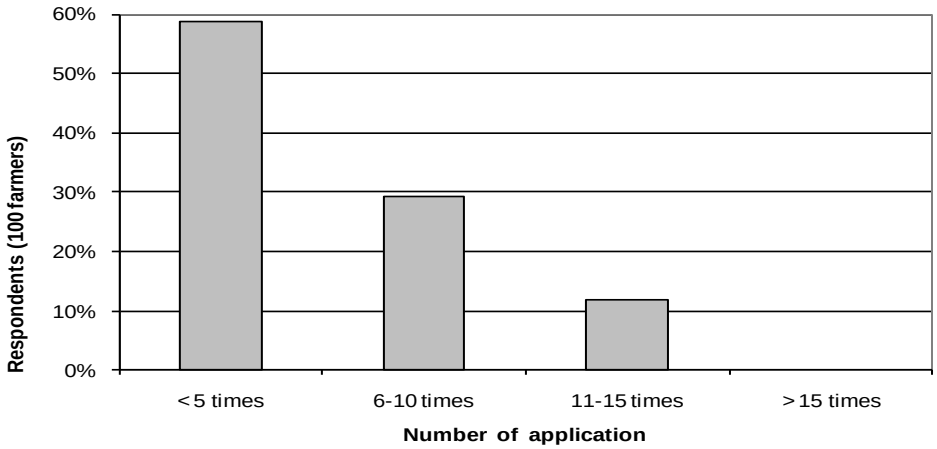

Figure 3. Pesticides application within a week

Within one day application, about 46% farmers spray 3 container (using back sprayer volume 14 l) (Figure 4), even though there was not available data on how large area that have been sprayed. The time spent for pesticides application less than 30 minutes (53%), but about 20% spent more than 30 minutes and even 18% respondent spent more than 1 hour (Figure 5).

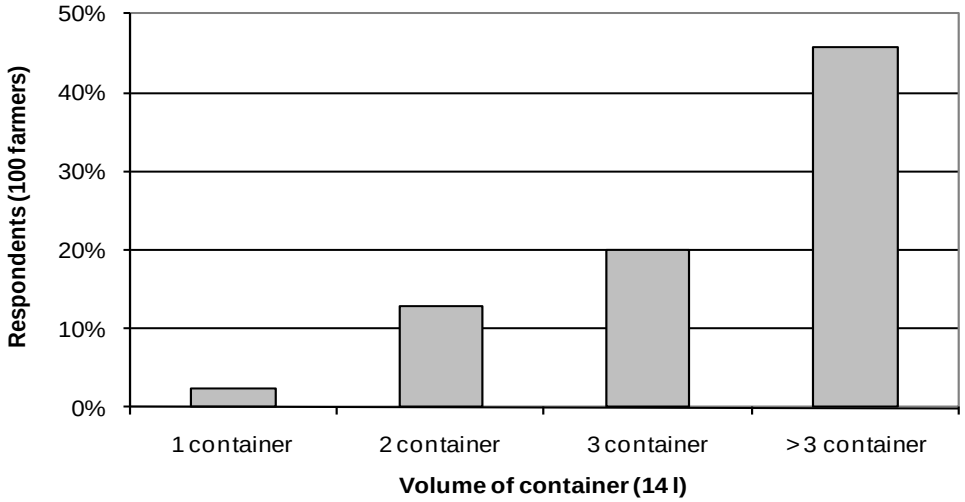

Figure 4. Volume of pesticides application in one day

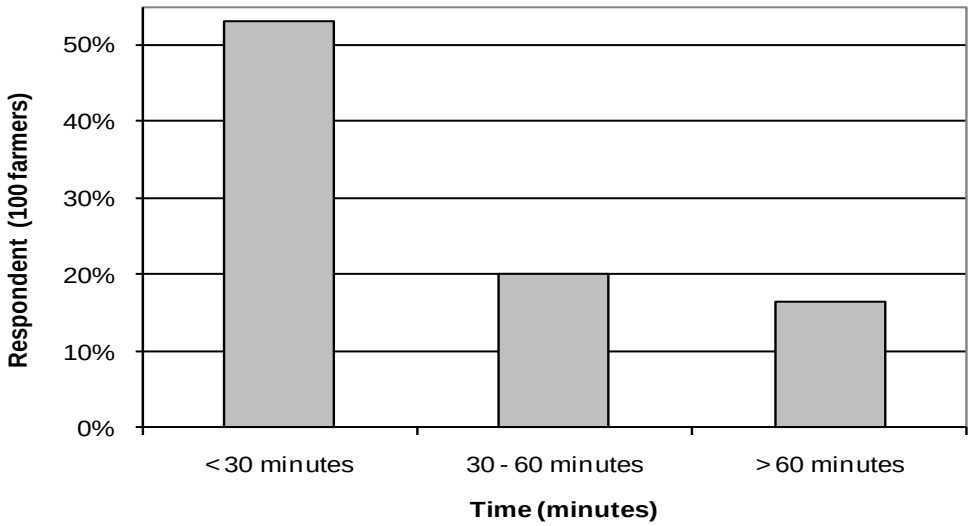

Figure 5. Time spent for each pesticides application

Information above portrays that most of the respondent used pesticides very intensive within one planting season. In addition, generally the farmers mix two or more pesticides to increase the toxicity to prevent and eradicate the pests. This kind of action did not use knowledge base, only based on information from experienced farmers. Therefore they do not know the effectiveness and accuracy of the mixture, and also the negative effect to their health and the environment. It is believed that the farmers were heavily exposed to pesticides for long time with high frequency application will increase the poisoning potency.

C. Pesticides exposure to the body

During spraying activity, the operator did not follow the standard cloth (Personal Protective Equipment) properly. Table 1 shows that the operators used minimum protection to their body, commonly they only use hat (80%), boots (65%) and double cloth (68%). Furthermore, only 11 % of the operators used gloves and masker, and even only 5% used glasses.

Table 1. Personal Protective Equipment during pesticides application

Personal Protective Equipment	Percent (100 farmers)
A. Head protection (hat)	80%
B. Glasses	5%
C. Masker	11%
D. Gloves	11%
E. Foot protection	
Boots	65%
Ordinary shoes	14%
Zandal	1%
F. Body protection	
Double shirts	68%
Single shirt	20%
Without shirt	1%

Meanwhile Figure 6 also explains that the operators admitted that during preparation of the pesticides, they were exposed to pesticides on their hands (76%), and to their legs/calf (9%) also foots (12%).

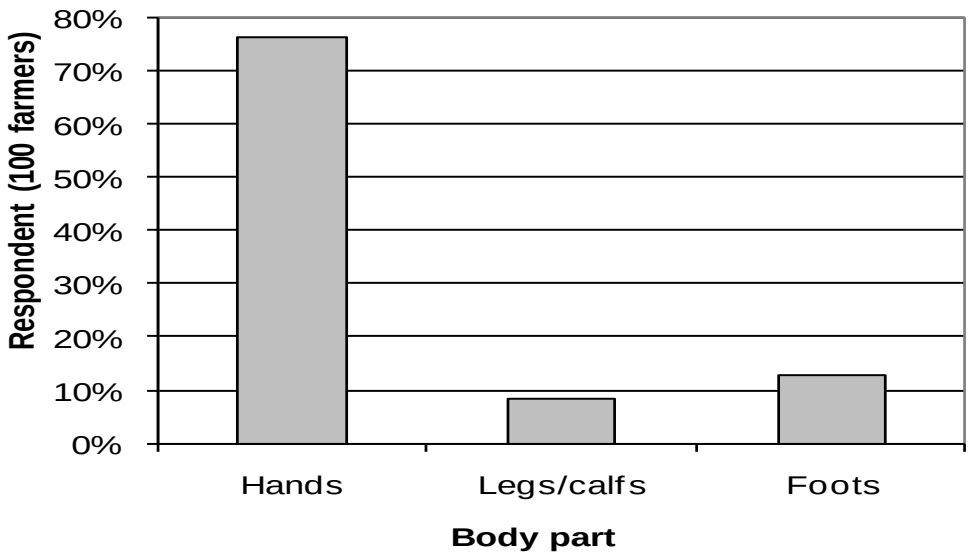

Figure 6. Skin contact during preparation of pesticides

In addition to inhaling the pesticides during application, the operators also admitted that most of their body subject to be contaminated by pesticides, mostly on their hands (81%), back and abdomen (72%), and others of about 40 to 55% (Figure 7). The understanding of using protection equipment was only on head, hands and foots/legs, but the other important body part were neglected such as eyes, nose and mouth.

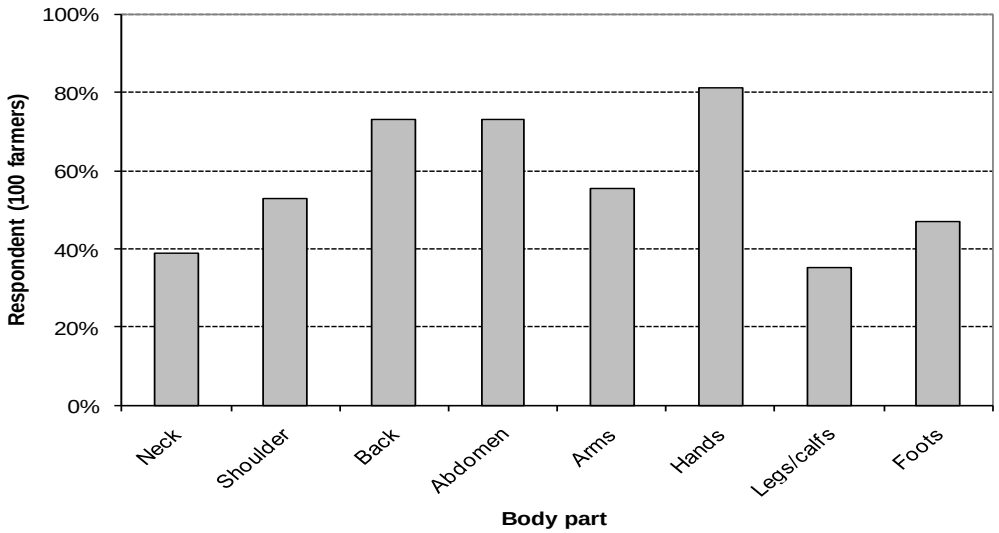

Figure 7. Skin contact during pesticides application in the filed

D. Health status

Long term exposure to pesticides experienced mostly by farmers affecting their health status. The respondents admitted that there were feeling of illness either their externally and internally of their bodies (Figure 8). But mostly felt tired (40%), muscle cramps (27%), dry throat and shortness of breath (21%), headache, dizzy, excessive salivation and other illness related to eyes and stomach.

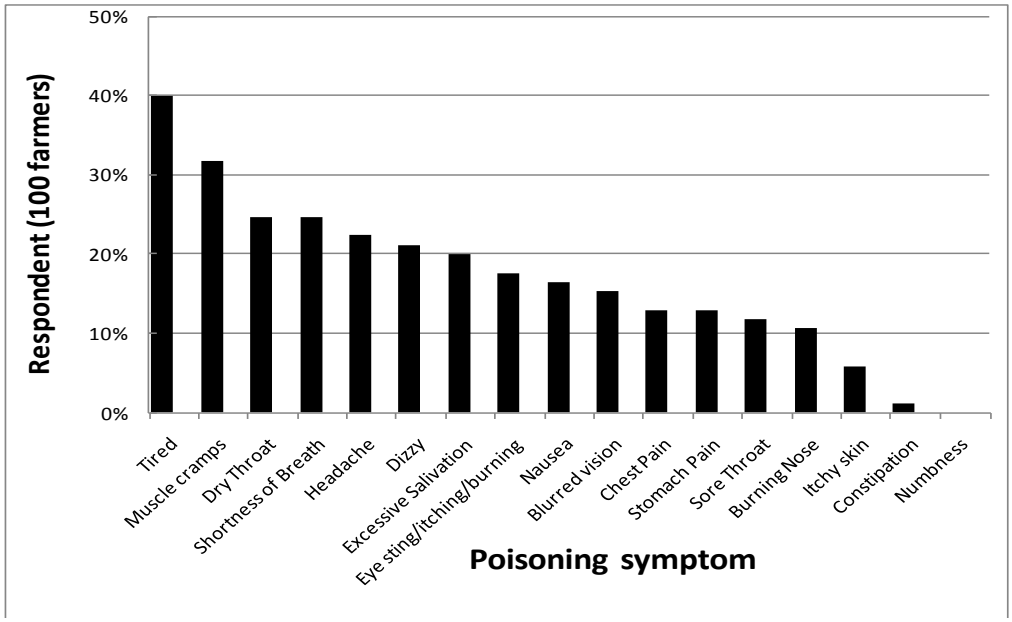

Figure 8. Poisoning symptoms as experienced by farmers

Clinical evaluation based on blood and urine test accentuate the respondents' admittance on their health status, in which 35% of them have problems on either deterioration in lever and kidney function or chronic infection (Figure 9). There was interesting evidence that the respondents that were found having clinical problem mainly in the age of over fifty years old. This is an indication that the effect of pesticides on health could be in the long term.

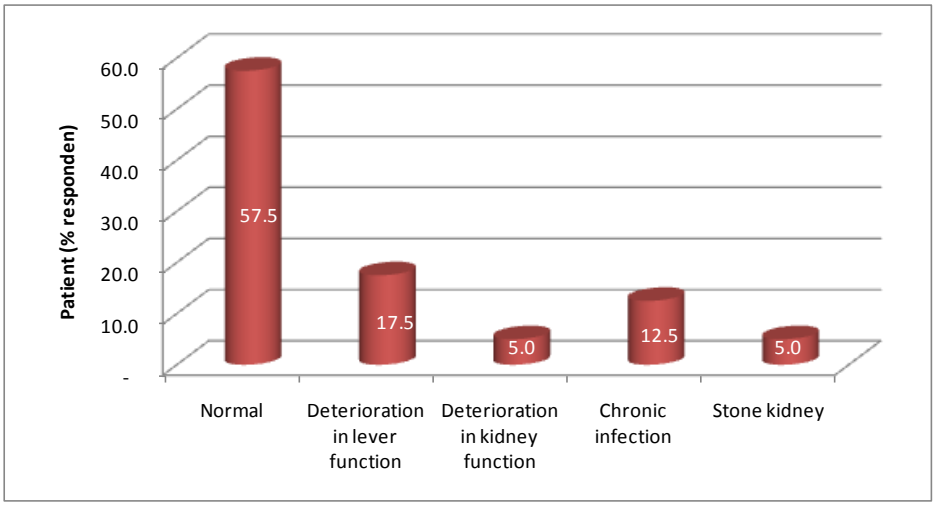

Figure 9. Health status based on clinical test

CONCLUSSION

The research concludes that the farmers basically understand on handling pesticide at home, but they tend to excessively use pesticides. In addition, the farmers were ignorant of having protective equipment during pesticides application, and ultimately about half of the spray operators had experienced on health problem either based on their feeling or clinical examination.

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Phosphorus Recovery from Agroindustrial Wastewater through Struvite Crystallisation: Principles and Applications

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Abstract

Intensified agriculture in response to the growing population has led to excessive nutrient discharges into waterbodies causing eutrophication, which affects biological diversity and water quality. One of such nutrients is phosphorus (P), an essential element for every living plant, and hence for food production. An attractive method to help minimise eutrophication is to remove P from wastewater by converting it from a soluble form into solid phase, i.e. a phosphate mineral commonly known as struvite ($MgNH_4PO_4.6H_2O$), through a controlled crystallisation process. The method has increasingly gained particular interest since struvite can be utilised as fertilizer which simultaneously provides Mg, N, and P in proportional amounts to crops. Since struvite has low solubility in water, it may also act as slow-release fertilizer and may not cause burning of roots. P removal and recovery as struvite is also an effective method to address the problem of operating difficulties in wastewater treatment facilities, since under favourable conditions struvite can spontaneously form in wastewater, and the resulting struvite crystals agglomerate and excessively cover the piping systems, pumps, screens etc. This paper discusses the principles of P recovery from agroindustrial wastewater through struvite crystallisation. The findings of a preliminary experiment examining the onset of struvite crystallisation from synthetic liquors are presented. Applicability of the struvite crystallisation method including its advantages and disadvantages, as well as typical operating conditions and the properties of the struvite crystals produced will be scrutinised.

Keywords: crystallisation, eutrophication, phosphorus, struvite, wastewater

Introduction

Intensified agriculture is an inevitable consequence of population growth. Such intensification demands extensive use of fertilizers, and over-application of phosphate fertilizers to soil has been frequently encountered [Harris *et al*., 2008; Nelson *et al*., 2003]. Accordingly, waste streams from agricultural activities contain considerable amount of soluble phosphate. Discharging such nutrient-rich streams to surface waters results in eutrophication which is unsightly, lethal to aquatic life, and negatively affects water quality [de-Bashan and Bashan 2004].

Phosphorus (P) is an essential element for living organisms, plants included, hence food production relies heavily on the availability of P. On the other hand, P is also regarded as the primary nutrient for eutrophication of surface waters. Efforts to prevent eutrophication by removing P from wastewater have been widely put forward.

A promising method to reduce eutrophication is to remove the excessive P from waterbodies by converting it into solid phase, i.e. a phosphate mineral known as struvite ($MgNH_4PO_4.6H_2O$), through a controlled crystallisation process. Struvite can be used as fertilizer since it has low solubility in water, hence would not quickly leach in waterbodies. Since the leaching potential is low, struvite can act as a slow release fertilizer, and can be applied at relatively high rates without damaging plant roots [Gaterrel *et al* 2000]. Struvite can be used individually or in combination with other agricultural fertilizers.

P crystallisation as struvite is also an effective method to address the problem frequently encountered in wastewater treatment facilities (WWTFs), since under favourable conditions struvite can spontaneously form in wastewater, and the resulting crystals agglomerate and excessively cover the piping systems, pumps, screens etc.

This paper discusses the principles of P removal and recovery from agroindustrial wastewater through struvite crystallisation. The findings of an experiment examining the effect of metal ions on the morphology of struvite are presented. Applicability of the struvite crystallisation method including its advantages and disadvantages, as well as typical operating conditions and the properties of the struvite crystals produced will be scrutinised.

Principles of P Recovery through Struvite Crystallisation

Agroindustrial waste is commonly land-applied or treated by anaerobic digestion. Land-application is not always desirable since it causes green house gas emissions, eutrophication, and groundwater pollution. Anaerobic digestion is preferred due to the possibility of alleviating enviromental concerns and generating biogas [Jin *et al* 2009]. However, anaerobic digestion of waste sludge gives rise to high concentrations of P and ammonia (N) in the sidestreams, and these high nutrient-rich loads have to be removed or preferably recovered. Moreover, anaerobic treatment increases the volume of the sludge. In this respect, many researchers have found that struvite crystallisation is an attractive option for P removal and recovery.

After sludge separation, subsequent liquors contain high concentrations of soluble P and N, and lower concentrations of soluble Ca and Mg. Significant proportions of this soluble P can be recovered by a crystallisation process unit placed after sludge digestion [Pastor *et al*., 2010].

In many places, the manure management for dairies consists of flushing the manure with water, followed by separating the coarse solids through screening or sedimentation. The liquid fraction is then sprayed to the fields as fertilizers. Such practice is favourable since the nutrients are recycled on-site. However, the liquid fraction generally contains high concentrations of both P and N in a ratio that is inappropriate for crop requirement [Harris *et al* 2008]. On the other hand, it has been reported [Suzuki *et al* 2007] that certain swine wastewater already contains Mg^{2+}, NH_4^+ and PO_4^{3-} in high concentrations. This condition is preferable since adjusting the pH of the wastewater to 8 – 9 would lead to spontaneous crystallisation of struvite. However, unless the crystallisation process is controlled, the struvite crystals can agglomerate and cause scaling of equipment, which complicates the

operation of WWTFs. Nutrient recovery from agroindustrial wastewater by struvite crystallisation is an advantageous method since the process prevents scaling problems [Doyle and Parsons 2002], removes soluble P [Battistoni *et al* 1997; Laridi *et al* 2005], as well as N [Çelen and Türker 2001; Altinbas *et al* 2002; Laridi *et al* 2005]. Integration of a struvite crystallisation unit into sidestream of WWTFs is becoming common practice [Caffaz *et al* 2008].

Availability of Struvite Ion Components

The three major components of struvite: magnesium (Mg^{2+}), ammonium (NH_4^+-N), and phosphorus (PO_4^{3-}-P) can come from a number of sources. As can be seen from its chemical formula: $MgNH_4PO_4.6H_2O$, once these three ions are present in equimolar ratios, it is theoretically possible for the struvite to crystallise.

In most cases, agroindustrial wastewater is deficient in magnesium [Pastor *et al* 2010]. The amount of magnesium ions in swine lagoon liquid is usually only half of that required stoichiometrically for struvite crystallisation [Nelson *et al* 2003]. As reported by Marti and co-workers [Marti *et al* 2010], effluents from sludge digestion systems usually have high concentrations of P, Mg^{2+}, NH_4^+ with Mg^{2+} being the least. Another group of researchers [Liu *et al* 2011] also stated that the magnesium content of agroindustrial wastewater is very low compared to the other two components: P and N. Hence, magnesium additions to wastewater is common practice to encourage struvite crystallisation. A number of different Mg-containing chemicals have been used as sources of magnesium ions. These sources are: $Mg(OH)_2$ [von Münch and Barr 2001], $MgCl_2$ [Pastor *et al* 2010; Forrest *et al* 2008], MgO [Quintana *et al* 2008], magnesite [Gunay *et al* 2008], and bittern [Lee *et al* 2003]. Location of WWTFs may also influence the availability of magnesium ions. The ions can be plentiful in hard water area, certain industrial areas, as well as in places where seeping of sea water may take place [Doyle and Parsons 2002]. Yet another source of magnesium ions is the support materials placed inside anaerobic digesters, such as bentonite, dedicated to provide surface area for biomass attachment [Doyle and Parsons 2002]. In such cases the release of magnesium ions into the surrounding water can be significant if these materials were left in the digester for a long time.

Agroindustrial wastewater typically contains high concentrations of N due to the degradation of organic matter and nitrogeneous material such as proteins. Manure of domestic animals and poultry are also nutrient-rich wastes as sources of N [Ren *et al* 2010]. In general, wastewater contains relatively excess N compared to other components [Doyle and Parsons 2002].

Anaerobic digestion process releases polyphosphates into the liquid phase, which simultaneously increases the concentration of P, Mg^{2+} and K^+ in the solution [Marti *et al* 2010]. Three soluble forms of P are usually present in wastewater: PO_4^{3-}, HPO_4^{2-}, and $H_2PO_4^-$ [Doyle and Parsons, 2002]. While sufficient P and Mg^{2+} contribute to the formation of struvite, the presence of K^+ might encourage the crystallisation of another phosphate mineral: $MgKPO_4$ [Huang *et al* 2011], reducing the yield of struvite crystals.

Effect of Component-Ion Molar Ratios

Although struvite crystallisation is theoretically possible when the ratio of Mg/N/P in a crystallising solution is 1:1:1, the crystallisation process is highly dependent on the pH of the solution (see chemical equation in the later section of this paper). It has been found

that depending on the stoichiometric condition alone, and neglecting the optimum pH level, the crystallisation of struvite might take a number of days [Battistoni *et al* 1997].

As guidance, for struvite to crystallise the ratio of Mg/N/P in the solution should be higher than equimolar. As an example, Mohan and co-workers [Mohan *et al* 2011] carried out experiments using three chemicals in a batch reactor (a 250-ml Erlenmeyer flask) to obtain a synthetic solution having an initial $NH^{+}/Mg^{2+}/PO_4^{3-}$ ratio of ~ 3.5/3/4. The pH of the solution was adjusted to 8.9 using 5.0 N NaOH and the crystallisation proceeded spontaneously. The experiment, which lasted about ten minutes, produced sufficient amount of struvite crystals.

As described earlier (see previous sub-section), the Mg content of the wastewater is usually in deficit, i.e. the Mg/P molar ratio is less than one, which theoretically would not encourage the formation of struvite. In such cases, addition of Mg-containing chemicals are needed and this addition may involve significant cost.

Song and co-workers [Song *et al* 2011] have used CO_2 stripping method and were successful in crystallising struvite without chemical additions. The main reason being the molar ratio of Mg/P was kept constant between 1.31 and 2.42. This condition is in agreement with similar experiments on struvite crystallisation from swine waste [Burns *et al* 2003; Jordaan *et al* 2010] where for an effective struvite crystallisation, the Mg/P ratio was maintained at 1.6.

Effect of pH

Struvite crystallises as white orthorhombic crystals according to the following reaction:

$$Mg^{2+} + NH^{+} + HnPO_4^{n-3} + 6\ H_2O \longrightarrow MgNH_4PO_4.6H_2O + nH^{+}$$

Depending on the pH of the solution, the values of **n** can be 0, 1, or 2. Obviously, the crystallisation process is highly dependent on pH. The range of pH suitable for struvite crystallisation can be sufficiently wide, namely from 7.00 to 11.00 [Ali 2005]. However, individual investigations have shown that the precise and optimum pH where struvite has the maximum potential to crystallise is narrower in range as can be seen in Table 1. It can be inferred from the table that in most cases, the optimum pH for struvite crystallisation is about 9.0. pH level plays an important role in struvite crystallisation process since increasing the pH from 7.0 to 9.0 would result in a 250 fold increase in the amounts of PO_4^{3-} ions, while NH_4^{+} reduces slightly [Stumm and Morgan 1970]. As described previously (see Availability of struvite ion components) pH adjustment is a crucial factor for the completion of struvite crystallisation.

Table 1. pH values for struvite crystallisation used by different researchers.

Crystallisation Time	mg:p	Agitation	pH	References
120 minutes	1:1 to 1.6:1	120 rpm	7.5-9.5	Nelson *et al.,* 2003
24 hours	1:1	200 rpm	7.0-11.0	Ali, 2005
25 minutes	1:2	None	9.0(fixed)	Le Corre, 2006
10.5 to 12.5 hours	0.8:1 to 1.7:1	unknown	8.7 (adjusted)	Pastor *et al.,* 2010
4 hours	1:1	air bubbling	8.09-8.43	Rahman *et al.,* 2011
4 hours	0.5:1 to 1.2:1	air bubbling	7.82 to 8.92	Liu *et al.,* 2011

Effect of Impurities

Although impurities are usually present in trace (or ppm) amounts, their effects on crystallisation can be significant [Hoang *et al* 2009], since these impurities may block the "active sites", thus hindering the growth of crystals. Depending on the type of wastewater, there is a wide range of impurities which can affect the crystallisation of struvite.

Wastewater originating from agroindustrial sector typically contains high concentrations of Ca^{2+} as impurities [Parsons *et al* 2001], which can interfere with struvite [$MgNH_4PO_4.6H_2O$] formation due to the tendency of Ca^{2+} to react with the phosphate ions forming calcium phosphate: $Ca_3(PO_4)_2$ [Pastor *et al* 2008; Song *et al* 2011]. According to Yi and co-workers [Yi *et al* 2005), however, such competition for phosphate ions could start taking place only when the Ca^{2+} concentrations are significantly high, i.e. above 300 ppm. A recent investigation by Pastor and his colleagues [Pastor *et al*., 2010] showed that the presence of Ca^{2+} could also give rise to the formation of both calcite: $CaCO_3$ and dolomite: $CaMg(CO_3)_2$, if the solution also contains sufficiently high concentrations of Mg^{2+}.

Swine wastewater usually contains high concentrations of Cu^{2+} and Zn^{2+} which originate from the food supplement given in the piggery. These two ions are regarded as not environmental friendly, so their presence in wastewater has been actively investigated [Lake *et al* 1985]. It has been found [Lisitsin *et al* 2005] that trace amounts of Zn^{2+} (about 2 ppm) was capable of retarding the growth of $CaCO_3$ crystals either in the bulk of the solution or as scale in the RO membranes of a desalination facility.

An Experiment on Struvite Crystallisation from Synthetic Liquors

We carried out a bench-scale experiment using synthetic liquors to investigate the effect of metal ions, namely Cu^{2+}, and Zn^{2+} on the morphological change of struvite.

Materials and Methods

All reagents used were of analytical grade (Merck®) or equivalent. $MgCl_2.6H_2O$ and $NH_4H_2PO_4$ were separately dissolved in doubly-distilled water, which was previously boiled to purge CO_2, to provide Mg^{2+}, NH_4^+-N, and PO_4^{3-}-P with varying concentrations. For pH adjustment, up to 9.00, NaOH solution was used. Effect of ppm amounts of metal ions (Cu^{2+}, and Zn^{2+}) on struvite crystallisation was investigated in this study, and for this purpose, crystals of $CuCl_2.2H_2O$, and $ZnCl_2$ were separately dissolved in $MgCl_2$ solution.

A series of batch crystallisation experiments were conducted using a one-litre clean ("scratchless") glass beaker. The beaker was agitated using a variable speed pitched blade impeller (in-house made), and was placed in a thermostatically controlled water bath. Stirring rate was fixed at 125 rpm as it was found that this speed was sufficient to make the solution visibly well stirred. The two solutions of $MgCl_2$ and $NH_4H_2PO_4$ were placed in the crystalliser, the timer was started (= which corresponded to t = 0 or zero time), and the impeller was activated vigorously for five seconds and then reduced to 125 rpm until the crystallisation completed. During the experimental run, pH values of the solution were measured (using combined pH meter: Martini-Mi 805-Romania) at 0, 1, 2, 3, 4, 5, 10, 15, 20, 25, 30, 35, and 40 minutes, respectively. As crystallisation proceeded, the pH of the solution decreased gradually and levelled off after about 30 minutes, and therefore extension to 40 minutes was considered to be the end of the crystallisation reaction. Next, the whole solution was filtered (0.20 μm Whatman filter paper) and the crystals were air dried overnight at room temperature. The morphology of these crystals were then

examined using a light microscope (Yenaco XS2-107 BN-Japan). After completion of one experiment, the beaker and its accessories were cleaned and prepared for the next run.

Results and Discussion

The typical morphology of the struvite crystals obtained is shown in Figure 1. Fig. 1.a depicts struvite crystallised in pure solution without impurities addition. As can be seen the elongated needle-like crystals confirm the typical struvite morphology [Stratful *et al* 2001; Kabdasli *et al* 2006]. On the other hand, Figs.1.b and 1.c. exhibit the crystals obtained when 20 ppm of Cu^{2+} and Zn^{2+} were added, respectively. The two images show that the crystals were no longer needle-like but agglomerated. This morphological change could be the result of Cu^{2+} and Zn^{2+} blocking the active sites of the crystal surface. Hence the growth of the crystals was stopped and the crystals failed to form the extended needle shape [Muryanto 2002]. Considering the agglomerated struvite crystals as shown in Figs.1b and 1.c., it is still difficult to distinguish the specific influence of either Cu^{2+} and Zn^{2+} on morphological change of crystals.

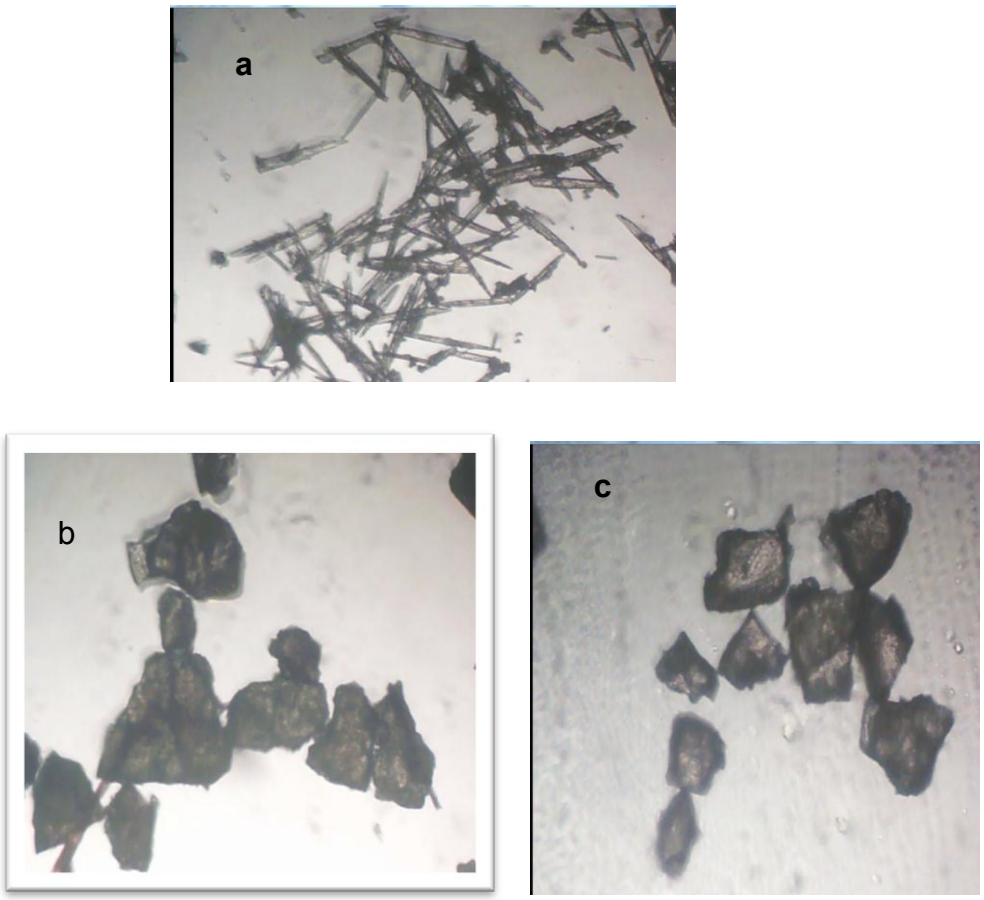

Figure 1. Struvite crystallised at 40 min. (a) pure solution; (b) 20 ppm Cu^{2+} added; (c) 20 ppm Zn^{2+} added. (solution concentration: 4.0 mM; image magnification: 40x).

Typical Full-Scale Struvite Crystallisation

The method of struvite crystallisation falls into three categories [Le Corre, 2006], namely, (1) selective ion exchange, (2) air-agitated reaction, and (3) stirred reaction. The third method comprises a fluidised bed reactor (FBR) and is by far the most promising. A number of industrial scale FBR for struvite crystallisation have been operational, such as the one shown in Fig. 2. As depicted in the figure, P- and N-rich filtrate from an anaerobic unit is pumped upward from the base of the inverted cone-shaped column (see Fig.3 for details of the FBR column). Near the base of the inverted cone (see Fig. 3), two separate inlets were made to supply NaOH and $MgCl_2$ solutions, for pH adjustment and Mg^{2+} additions, respectively. Once the pH level of the solution has been adjusted and reached the alkaline conditions of ≥ 8.0, struvite spontaneously crystallises, and subsequently carried to the upper section of the FBR by the continuous upflow stream. During this period, the struvite crystals collide among themselves and start agglomerating with each other to form pellets. The pellets fall to the base of the column due to their high density, while smaller struvite crystals are suspended inside the column and continue to grow or agglomerate. The struvite pellets with sizes from 1 to 2 mm in diameter, are periodically harvested from the bottom of the FBR. For easier handling, a straightforward additional process is usually required, such as air drying.

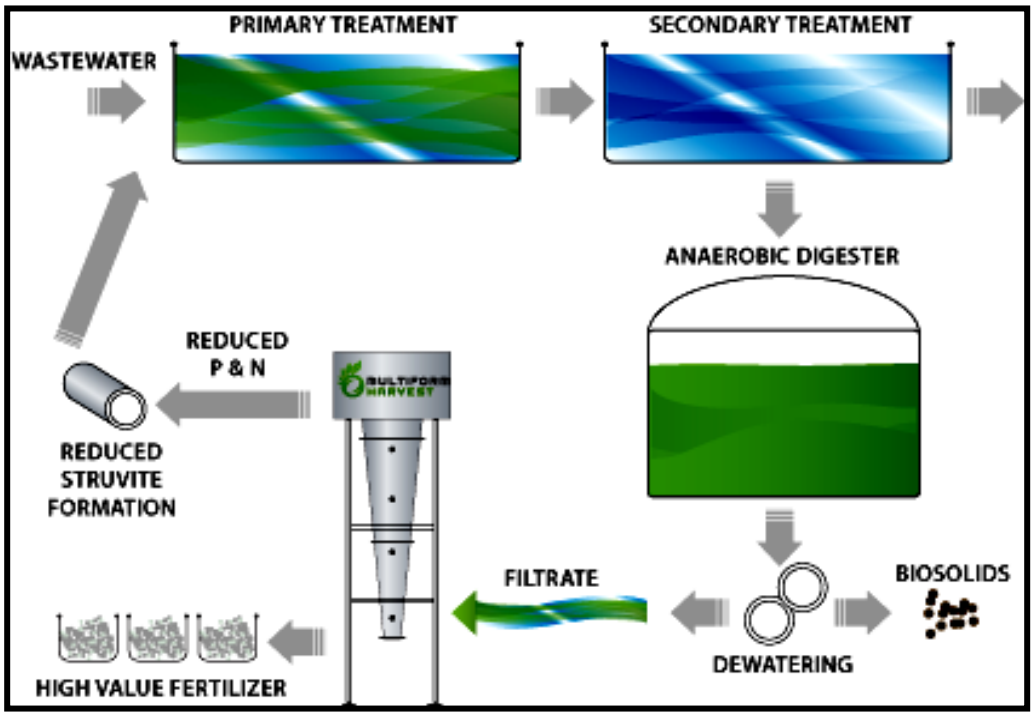

Figure 2. Schematic diagram of an enhanced biological phosphorus removal (EBPR) unit coupled with an FBR to recover P as struvite fertiliser (Courtesy of Multiform Harvest Inc. Seatle, Washington, USA, www.multiformharvest.com - personal communication with first author).

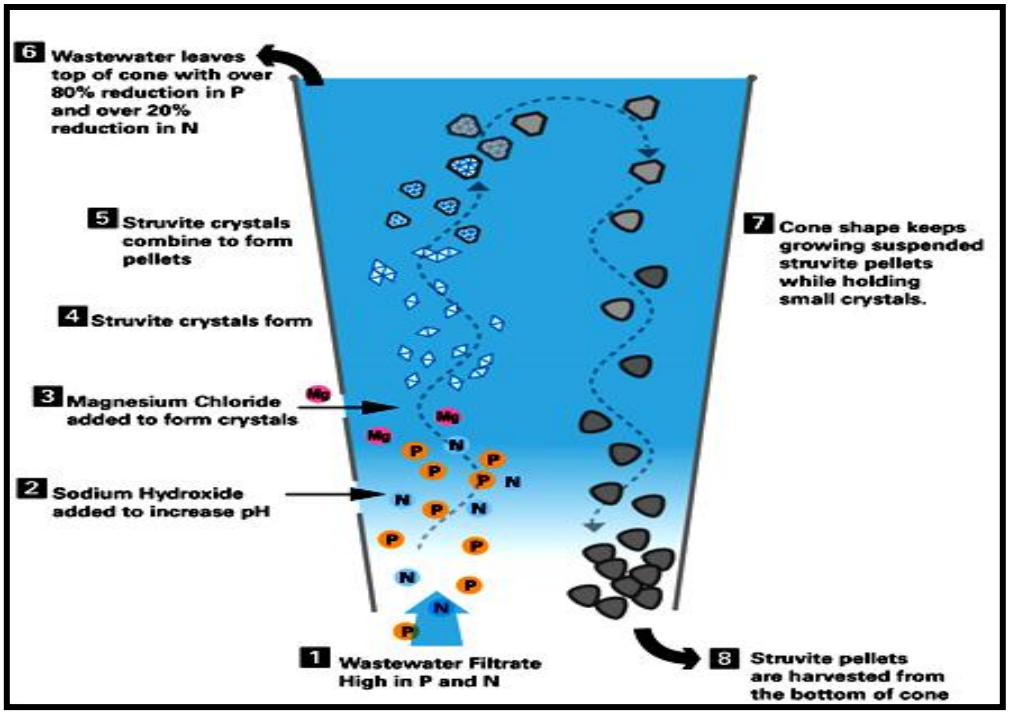

Figure 3. Detailed description of the FBR (Courtesy of Multiform Harvest Inc. Seatle, Washington, USA, www.multiformharvest.com - personal communication with first author).

Conclusions

Agroindustrial wastewater is a highly potential source for struvite crystals. Struvite crystallisation from agroindustrial wastewater needs addition of chemicals especially as sources of magnesium ions. Trace amounts of metal ions: Cu^{2+} and Zn^{2+} can change the morphology of struvite crystals from typically needle-like to become agglomerated crystals. A number of full-scale struvite crystallisation projects using agroindustrial wastewater are already in operation.

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Arbuscular Mycorrhizal Fungi Induced the Content of Isoflavonoid that Reduced Potato Cyst Nematode on Roots of Potato Plants

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Abstract

The activities of soil microorganisms have a significat influence on plant health. Plant-parasitic nematodes and arbuscular mycorrhizal fungi (AMF) often occur in the rhizosphere and roots of plants and therefore frequently encounter each other. Arbuscular mycorrhizal fungi may stimulate plant growth, whereas plant-parasitic nematodes usually suppress plant growth. Pot studies on the effects of AMF (0, 50, 100, and 150 spore per pot) on plant growth and nematode reproduction in potato-potato cyst nematode (PCN) pathosystem (inoculated at two densities, 0 and 4000 cysts per pot) was carried out. The experiment was also determined biochemical substantiation for the observed difference in resistance to nematode infection in roots colonized by mycorrhiza.The treatments were number of spores of AMF and number of cysts of PCN Results showed that AMF at the rate of 150 spores per pot significantly reduced the numbers of juvenils II, females and cysts. The mechanisms of AMF in reducing PCN activities was more likely because of the production of antifungi isoflavonoid which increased according to the rate of AMF. This experiment showed the potential of AMF to be used as biocontrolling agents of PCN on potato plants.

Keywords: antifungi, arbuscular mychorrizal fungi, isoflavonoid, potato cyst nematode

Introduction

Nematodes have the greatest impact on crop productivity such as potato. The most well known nematode disease are those caused by potato cyst nematodes such as *Globodera rostochiensis* (Woll.) dan *G. pallida* Stone (Turner and Evans, 1998; Bridge and Starr, 2007). *Globodera rostochiensis* (Woll.) has been reported in Indonesia in the area of potato plantatation at Malang, East Java (Daryanto, 2003b).

Management of *G*. *rostochiensis* is completed primarily through crop rotation and the planting of resistant or tolerant cultivars (Bridge and Starr, 2007). Current emphasis in low input-based agrotechnology for crop production systems has stimulated the study and management of microbial interactions in the rhizosphere. In the particular case of potato production in tropical countries, emerging technologies include the use of inoculants of arbuscular mycorrhizal fungi (AMF) during the production of crops, is of continuing interest (Vierheilig *et al.,* 2008). These fungi are generally present in all soils that establish an obligate mutualistic symbiosis with many plant species. *Glomus* species are the most diverse of the arbuscular mycorrhizas and are found in many soils all over the world (Abbott and Robson, 1991). Besides known as biofertiliser or bioameliorator, several studies have reported that AM fungi species increase host tolerance in some plant-nematode systems, such as *Meloidogyne*, *Heterodera, Rotylenchulus dan Pratylenchus* (Talavera *et al.*, 2001; Nagesh and Reddy, 2004; Castillo *et al.*, 2006). Improvement in the growth of mycorrhizal

tomato and carrot (Talavera *et al*., 2001), soybean (Morandi *et al.,* 1984), olive (Castilo *et al*., 2006) grown on soil inoculated with nematodes have been showed. However, there was no information available about the role of AMF in protecting potato plants against root-knot nematodes, especially in Indonesia. Furthermore, the mechanisms in which AMF reduced nematodes population have not been cleared. Physiological changes in AM roots could change resistance to nematodes by increased production of inhibitory substance. Plant that received nematode alone and mycorrhiza plus nematode recorded differential biochemical contents of proteins, total phenols and IAA (Nagesh and Reddy, 2004). Singh *et al*. (1990) showed that pre-occupation of the roots with AMF coupled with biochemical changes such as increases in lignins and phenols make Pusa Ruby resistant to root-knot nematodes. It seems that there is no single response in the interaction mycorrhiza-nematode and that the response depends on various factors, such as nematode, mycorrhiza species, plant species or environmental factors (Talavera *et al*., 2001).

The objectives of this study were: (i) to determine the effect of AMF on population of *G. rostochiensis* on potato plant-nematode system; and (ii) to determine whether the reduction of nematode populations was associated with the production of inhibitory substance in plants inoculated by AMF.

Materials and Methods

A pot experiment was undertaken in a glasshouse of Faculty of Agriculture Universitas Padjadjaran from August to October 2010. Certified Potato (*Solanum tuberosum* L.) cv. *Granola* seeds were planted in plastic pots (25 cm top diam.; 20 cm bottom diam.; 25 cm depth), filled with 5 kg of a sterile soil (soil analysis presented at Table 1).

Table 1. Analysis of Soil from Pangalengan West Java used in Experiment (Collected in July 2010)

Parameter	Methods	Value
pH H_2O	Potensiometri	6.5
pH KCl	Potensiometri	6.0
C-org (%)	Walkley & Black	5.52
N-org (%)	Kjeldahl	0.49
P_2O_5 (mg/100g)	HCl 25 %	11.86
K_2O (mg/100g)	HCl 25 %	17.91
CEC (cmol (+)/kg)	Perkolation/NH_4 Acetat pH 7	25.54

The treatment were number of spores of AMF per pot (0, 50, 100, 150) and juvenils of G. rostochiensis (0 or 4000 per pot). Inocula AMF (Glomus sp.) consisted of spores were obtained from AMF pot cultures using Sorghum as host plants. The spores of AMF were applied according to the treatments one week before planting to enable spores to germinate.

Inocula of PCN G. rostochiensis for experiments were obtained by extracting eggs from soil at the potato plantation at Pangalengan West Java using methods described by Bridge and Starr (2007). The eggs were hatched and submersed in water for three days to obtain second-stage juveniles (J2). The nematodes treatment were applied one week after planting. Each treatment was replicated three times and pots were arranged on a glasshouse bench in a randomized-block design. Plants were watered daily to field capacity

and fertilized with 25-5-20 liquid fertilizer (50ml of 1/1,000 diluted Hyponex) at the time of planting. The experiment ended seven weeks after inoculation with the nematodes.

Seven-week-old potato plants were harvested . Potatoes were pulled from the soil and weight of shoot and roots of were recorded. The soil from each pot was sieved through a 2.5 mm mesh to separate the roots from the soil. Roots were washed free of soil, finely chopped, mixed thoroughly again with the soils, and stored in plastic bags. The nematode population in soil and roots (number of female eggs and juvenils) were assessed from soil samples by methods described by Bridge and Starr (2007). The percentage of the root system colonized by the mycorrhiza was assessed in subsamples of 0.5 g of roots at harvest time by the methods of Brundrett *et al*. (1996).

The production of inhibitory substance including isoflavonoid, Indole Acetic Acid and Chitinase were assessed from root samples by High Performance Liquid Chromatography (HPLC) at Laboratory of Research Center for Biology. Data were analyzed by two-way ANOVA using SPSS 16 program. When F values were significant, means were compared by the LSD test (p_0.05).

Results

Population of Potato Cysts Nematodes *G. rostochiensis*

Numbers of cysts of *G. rostochiensis* 100 g of soil was significantly lower in mycorrhizal plants than non-mycorrhizal plants . The number of cysts decreased as the number of AMF spores increased from 50, 100, and 150 per pot. The average reductions were 42, 60 and 86 % respectively (Table 2). The number of female was lower compared to control only when the AMF spores applied at the highest rate, with the reduction of 70%.

Table 2. Population of cysts and female of *G. rostochiensis* on roots of potatoes inoculated by spores of AMF

AMF spores per pot	Cysts per 100 g of soil	Female per g of roots
0	16,7 c	6,7 b
50	9,7 b	5,3 b
100	6,7 ab	5,0 b
150	2,3 a	2,0 a

Within each column, values followed by the same letter are not different (p>0.05)

Mycorrhiza Colonisation and Content of Inhibitory Substance on Roots

The percentage of roots colonised by mycorrhiza increased according to the rate of number of AMF spores. However, no significant differences were observed in the percentage of roots colonized by the mycorrhiza in plants infested by PCN *G. rostochiensis* (Table 3).

Inoculation of *G. rostochiensis* significantly decreased the content of isoflavonoid in roots of potatoes up to 25% (Table 3). Increasing the spores number of AMF from 50, 100 and 150 per pot significantly increased the content of isoflavonoid 34%, 48%, and 64% respectively. No significant differences were observed in the content of chitinase and IAA as the effect of AMF and PCN inoculation.

Table 3. Root colonisation by AMF (%), Content of isoflavonoid, chitinase, and IAA on roots of potatoes inoculated by AMF and PCN *G. rostochiensis*

Treatments	AMF Root Colonisation (%)	Isoflavonoid (ppm)	Chitinase (µg/g/hour)	IAA (ppm)
AMF (spores/pot)				
0	0	9.8 a	0.13 a	13.6 ab
50	31 a	14.8 ab	0.13 a	10.3 a
100	37 a	18.8 b	0.13 a	16.3 b
150	52 b	26.9 c	0.14 a	13.5 ab
PCN (J2/pot)				
0	36 a	22.9 b	0.13 a	13.9 a
4000	43 a	17.4 a	0.13 a	13.0 a

Within each column, values followed by the same letter are not different (p>0.05)

Discussion

Establishment of AMF in plant root systems is thought to contribute to biological control of plant diseases primarily by menas of stress reduction. The mechanismss including improvement of P absorption by plants; competition with the pathogen for infection sites and nutrients; morphological changes in roots and root tissues; micorbial changes in the mycorrhizosphere; and changes in chemical constituents of plant tissues (Suresh *et al*., 1985; Linderman, 1996). The use of AMF to protect plants against PCN *G. rostochiensis* could be a useful for the integrated management of root-knot nematodes in potato production (de Ruijter and Haverkort, 1999).

In general, this experiment showed that inoculation of AMF on potato plant-PCN system, reduced the number of female and cysts of *G. rostochiensis.* This is concured Sikora (1981) who found that the population of a *Globodera rostochiensis* Wollenweber reduced when potato plants inoculated by *G. fasciculatum*. Moreover, Suresh *et al.* (1985) stated that mycorrhiza reduced the number of nematodes larvae up to 50% and female nematodes were significantly lower in mycorrhizal plants. Ingham (1988) was also found that population of juvenils 75% lower when cofee, wheat and tomato plants were inoculated by *Glomus mossea*. Furthermore, Smith *et al.* (2001) confirmed that population of nematodes *M. incognita* on cotton plants reduced if the the level of mycorrhiza colonisation was higher than 50%. In this experiment, addition of 150 spore per pot resulted 52% root colonisation and highest reduction in nematode populations. The development of nematodes would inhibited when mycorrhiza penetrated the roots (MacGuidwin *et al.,*1985; Suresh *et al*., 1985).

In this experiment, the mechanisms on the reduction of PCN by FMA were likely because of direct competition for infection sites and production of isoflavonoid as one of inhibitory substance to nematodes (Table 3). Morandi *et al.* (1984) found that the concentration of isoflavonoid (phytoalexin-like) increased in mycorrhizal soybean plants and considered that isoflavonoid have a function as nematicide. Suresh and Bagyaraj (1985) assumed that the occurence of nematicidal substance in mycorrhizal roots was because of improvement of plant vigor and nutritions.

The biochemical defense response to mycorrhizal colonization against root-knot nematodes has been documented (Smant *et al.,* 1997). Assay on the activities IAA oxidase enzymes in treated and healthy roots of tomato, indicated that IAA oxidase activities were maximum in *G. fasciculatum* colonized roots followed by the roots that received mycorrhiza plus nematode (Nagesh and Reddy, 2004). Demonstration of other change in chemical constituents due to AMF inoculation showed by Dehne *et al.* (1982) where concentration of anti-fungal chitinase were increased in AMF roots. However, this experiment did not show the difference in IAA nor Chitinase content as affected by AMF inoculation (Table 3). The effect of AMF on growth of plant infected by nematodes were influenced by some factors including plant cultivar, species of nematodes and fungi, status of soil nutrients, time of inoculation and harvest.

The growth of mycorrhizal plants infested by nematodes were generally had smaller leaves and unhealthy (data not shown). AM fungi compensate some parts of the plants that demolished by nematodes. Ingham (1988) showed that the growth of mycorrhizal plants received nematodes were bigger than non-mycorrhizal plants. AM fungi would not colonise the site infected by nematodes and vice versa, the nematodes do not infect the site colonised by AMF.

In general, plant-parasitic nematodes and AM fungi commonly occur together in the roots or rhizosphere of the same plant, each having a characteristic but opposite effect on plant vigor. The obligately symbiotic AM fungi may stimulate plant growth, whereas the obligate plant-parasitic nematodes usually suppress plant growth. This experiment indicates that AM fungi have potential as biocontrol agents when both groups of microorganisms occur simultaneously in the roots or rhizosphere of the same plant.

Conclusions

1. Inoculation of spores of AMF at the highest rate (150 spores per pot) reduced the population in term of number of female and cysts of *G. rostochiensis* on potato plant-nematode system.
2. The reduction of nematode populations was associated with the production of inhibitory substance isoflavonoid in plants inoculated by AMF.

Acknowledgement

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Analysis of Sustainability of Capture Fisheries Resources Management (A Case in Pangandaran the District of Ciamis West Java)

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Abstract

Research was aimed to assess the sustainability fisheries resources in the region of Pangandaran Ciamis West Java. The production of capture fisheries has been decreasing and was affected by the tsunami disaster. This research uses primary data and secondary data. Analysis of the sustainability of fisheries resource management was done using Rapfish analysis. Primary data were collected by using a stratified cluster random sampling as many as 186 fishermens and purposively acquired five employees Marine and Fisheries district of Ciamis West Java Province. Analysis of the sustainability of fisheries resource management was done using Rapfish analysis. Result of analysis showed that sustainability on capture fisheries was sufficient to support the resource management.

Keywords: fisheries, rapfish, sustainability

Introduction

The management of fisheries resources by integrating the various factors is interrelated and affect the viability of fishery resources, fishery commodity demand for the fulfillment of human needs and synergy fishermen in exploiting fisheries resources can not be separated from economic value. Economic activities in coastal areas generally involve more than one sector, including fisheries and marine tourism sector. Result the existing density of economic activity, many regions in Indonesia suffered severe environmental destruction that threatened its sustainability. Destruction such as sedimentation, destruction coral reefs, destruction of mangrove forests and coastal erosion are some common natural form destruction of occurred in coastal areas

Pangandaran is one of the areas included the Indian Ocean zone of WPP IX which encompasses the waters of the western tip of Sumatra island and the south coast of Java. This region is a key region for tourism and marine capture fisheries. Both of these sectors recorded a major contribution to the regional economy and society in the region. Bojongsalawe Coast Subdistrict Parigi Ciamis District who became one of the study site is one such example. Result uncontrolled exploitation of resources have been reason coastal erosion is a great surf in the South Coast of Java resulting in abrasion of approximately 600 meters long (Department of Marine and Fisheries District. Ciamis, 2007). The impact of the destruction is exacerbated by various forms of natural events. One example is the earthquake followed by tsunami that occurred in many coastal areas of Indonesia including the Pangandaran area which is also the main location of the study. The earthquake and tsunami that occurred on July 17, 2006 has destroyed much of the infrastructure and various facilities owned by fishermen, including boat and fishing equipment. Disasters that destroy economic assets in coastal areas like that, if not handled properly and thoroughly able to impact further on the attitudes and actions of economic actors to resources.

Materials and Methods

Fisheries resources is one form of natural resources that are renewable or having characteristics can be recovering or be able to renew themselves. The fishery is a typical example of a common property resource, and thus suffers from the common property problem. Individual fishermen have incentives to harvest more than what is socially optimal, because they do not take into account the stock externality of harvesting (Linda Nostbakken, 2008).

Fishery resources in addition to having ifat renewable, according to Widodo and Nurhakim (2002), fishery resources have a nature in general "open acces" and "common property" which means the use is open to anyone and ownership of a general nature. Resource properties such as this raises some consequences, among others: (1) Without the management will cause symptoms of over-exploitation, over-investmen) and over-employment: (2) There is need for property rights, for example by state property rights, by the community property rights or by private individuals property rights.

A fundamental property of all fisheries is that fishery resources are common property natural resources. Nikijuluw (2002) suggests that there are three special properties that is owned by the resources that are owned by the joint. These three specific traits are : 1) Ekskludabilitas, these properties related to control efforts and monitoring of access to resources: (2) Substraktabilitas, these properties is a situation where a person is capable and can withdraw part or all of the benefits and advantages possessed by others: (3) Indivisibilitas , these properties essentially cause the fact that common property resources is very difficult to for or separated, although the administrative division or separation can be performed by the management authority.

Sustainable management of fishery resources by Pitcher (1999) includes ecological, economic, ethical, social and technological, as follows: (1) The analysis reflects how the impact of ecological sustainability of fishery resources seen from the level of exploitation, diversity recruitment, changes in the size of catch, discard and by catch and primary productivity, the attributes are then measured in terms of scale: (2) economic analysis: (3) ethical analysis: (4) social analysis and (5) technology analysis.

The method used in this research is survey method. Source of data extracted from primary data and secondary data. Primary data obtained through interviews or interviews and giving a list of questions to the respondents that include: community leaders and owners of fishing as much as 186 people, 4 people Fisheries and Marine officers Ciamis District and an employee Ciamis District Regional Planning Agency. Secondary data obtained from annual reports written agency or agencies involved in this study. While the techniques of data collection through literature study and interviews, which ask respondents directly on matters related to this research.

Results and Discussion

The Level of Sustainable Fisheries Resource Management Approach Rapfish Analysis.

Methods of analysis used to determine the level of sustainable management of fishery resources using Rapfish (A Rapid Apprasial Tehnique For Fisheries) through the method of Multi Dimensional Scaling (MDS) to assess the level of sustainable management of fishery

resources. Technics Rapfish ordinated through a multi-dimensional scaling method is a statistical technique that tries perform a multidimensional transformation into is more simple dimensions (Fauzi and Anna, 2005).

Multi-dimensional approach to scaling in Rapfish give stable results (Pitcher, 1999) compared with dual variable method of analysis to another. All data of the attributes considered in this research then analyzed to determine the multidimensional point reflects the position of the sustainability of fisheries is based on two points of reference, namely the point of "good" and the point of "bad. The position of these points will be hard to imagine visually because a lot of dimensions, for simplicity used the method of analysis ordination with multidimensional scaling (MDS).

In the MDS two titk or the same object is mapped in a far-flung points. These points will be very useful in regression analysis to calculate the 'stress' that is part of the MDS method. Score on each attribute will form a matrix X (nxp), where n is the number of areas while the p number of attributes used. A good model is indicated by the S-Stress value smaller than 0.25 or S <0.25 and R^2 close to 1. Index scale that assessed the sustainability of the system has interval 0% - 100%. In this study there are four categories of status of sustainability, as seen in Table 1.

Table 1. Category Index and Status of Sustainability

Index value	Category
0.0 – 25.00	Bad: Not sustainable
25.01 – 50.00	Less: Less sustainable
50.01 – 75.00	Sufficient: Simply Sustainable
75.01 – 100.00	Good: Very sustainable

Geographically district located at 108 Ciamis 0 40 'east longitude and 7 0 20' south latitude with an area as a whole reached 2488 km2. Administratively Ciamis district borders the north Majalengka and Kuningan, west Tasikmalaya District, east of the Banjar and Central Java Province and south by the Indonesian Ocean.

The coastal areas of six subdistrict located in the district of Ciamis better known as the Pangandaran area. The six subdistricts each have the potential for coastal tourism and fisheries, which should be able to be processed as optimal as possible by not ignoring aspect of sustainability of capture fisheries. The six subdistricts are: Cimerak, Cijulang, Parigi, Sidamulih, Pangandaran, and Kalipucang.

Fisheries Resource Sustainability Approach Rapfish

Sustainability of multi-species fisheries resources in this research through Rapfish approach. Identification of fishing in the Region which includes Subdistrict Kalipucang Pangandaran, Pangandaran, Parigi, Cijulang and Cimerak based on the type of fishing gear used.

Ecological Dimension

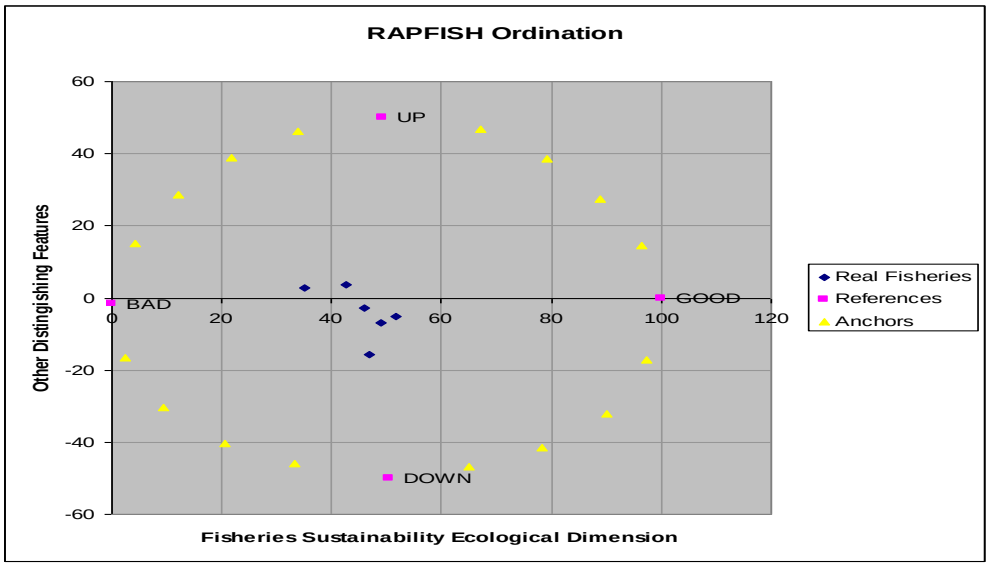


Figure 1. Rapfish ordination

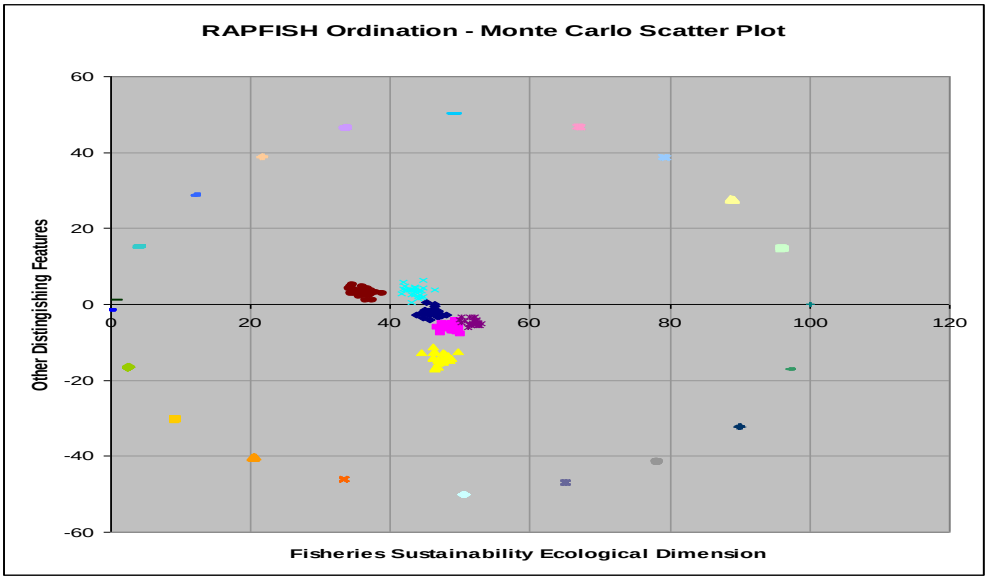


Figure 2. Rapfish ordination Monte Carlo scatter plot

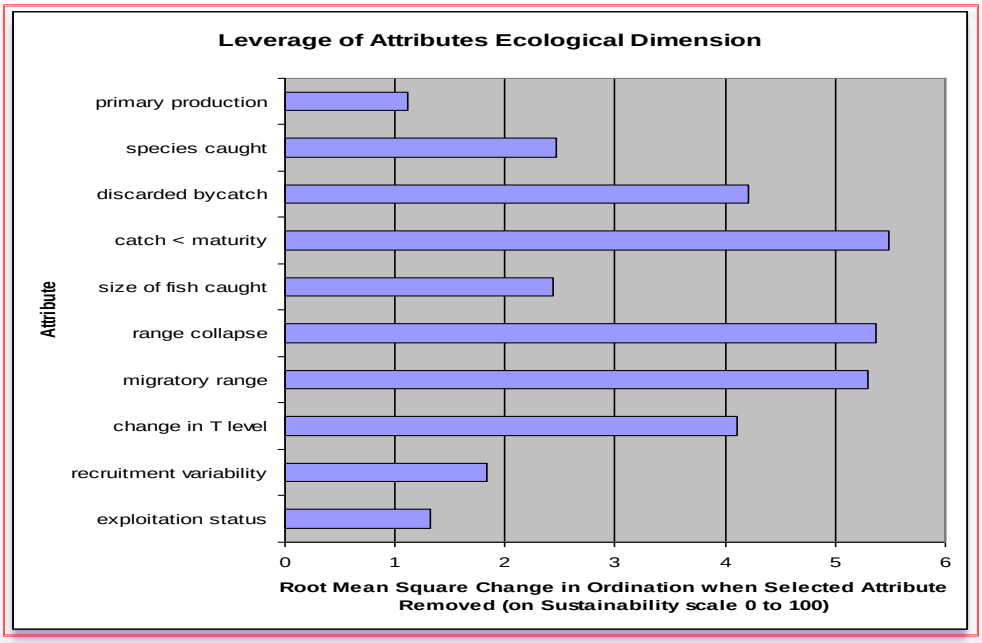


Figure 3. Leverage of attributes ecological dimension

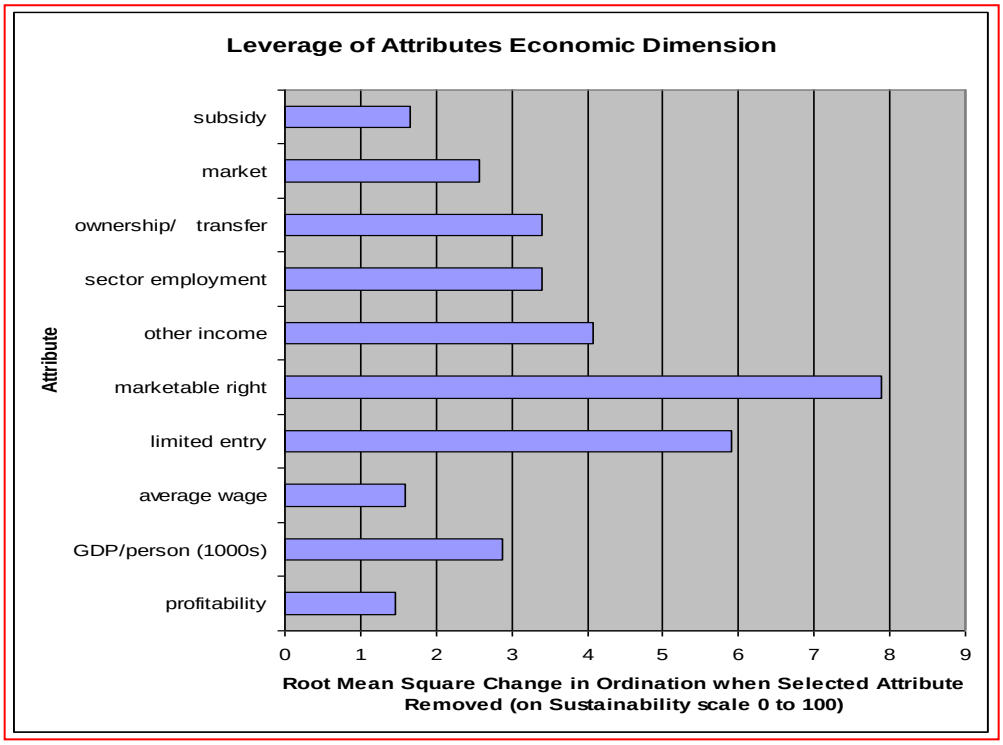


Figure 4. Leverage of attributes economic dimension

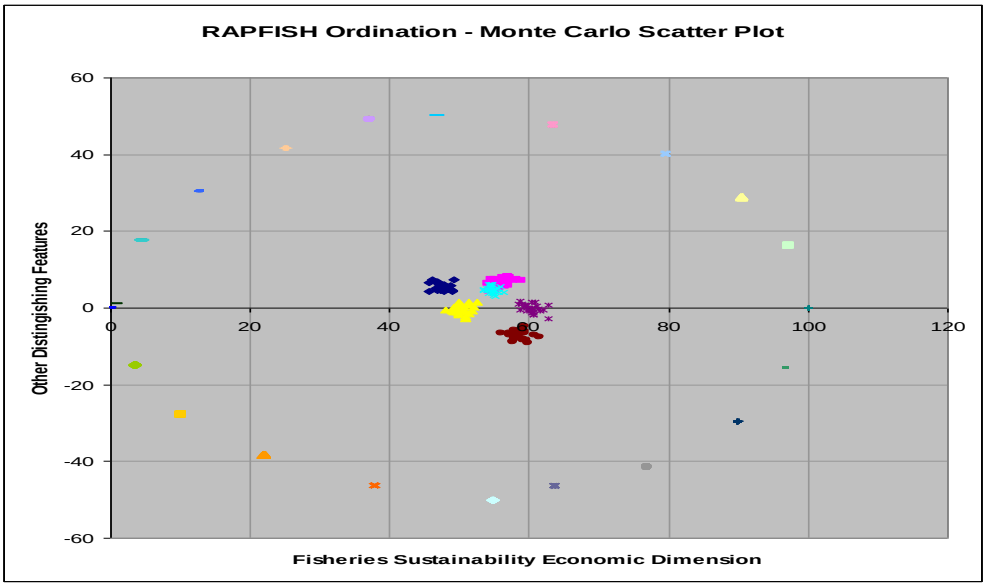


Figure 5. Radfish ordination-Monte Carlo scatter plot

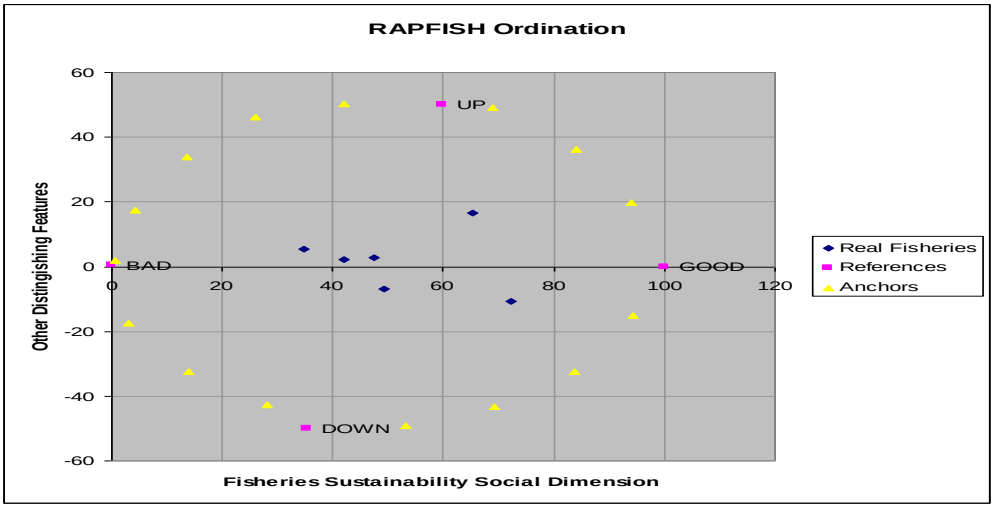


Figure 6. Radfish ordination

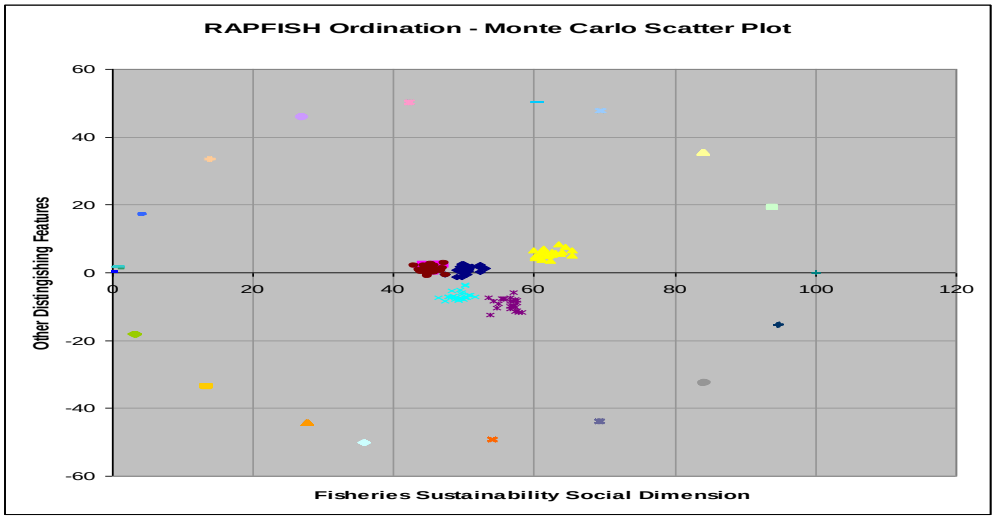


Figure 7. Radfish ordination-Monte Carlo scatter plot

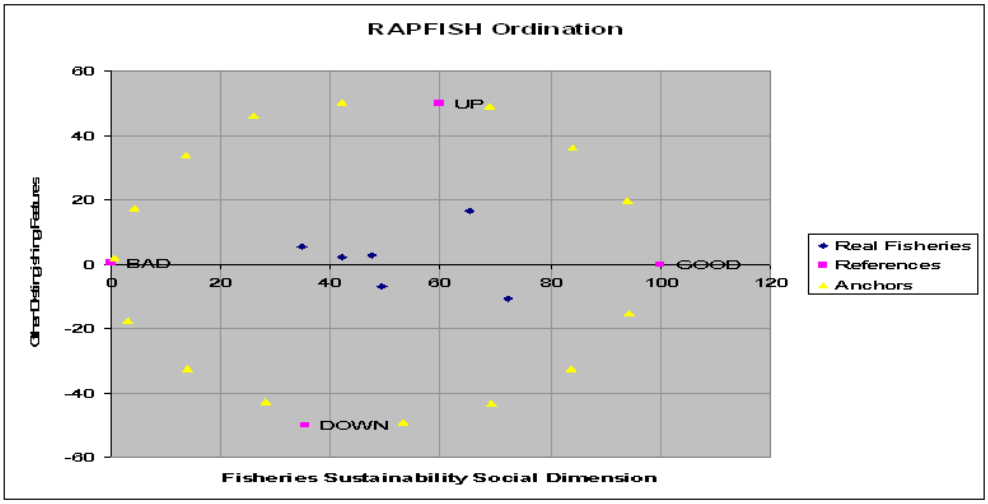


Figure 8. Radfish Ordination

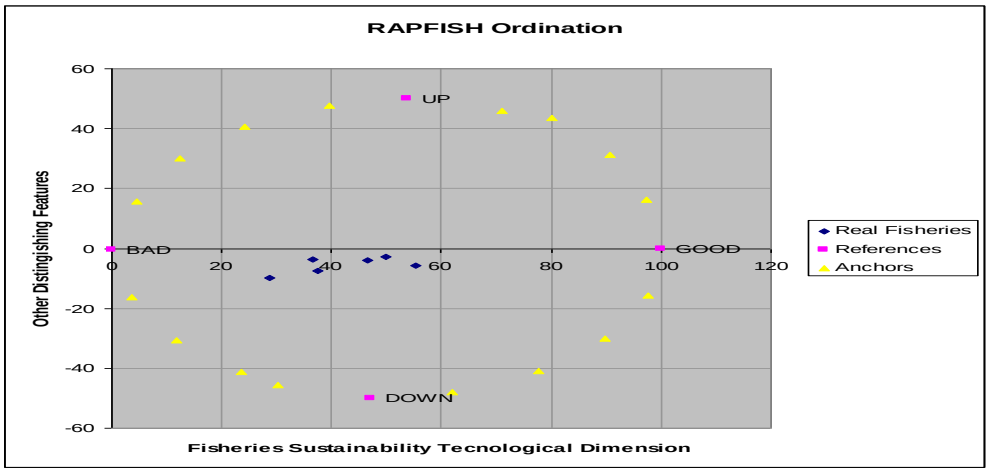


Figure 8. Radfish Ordination

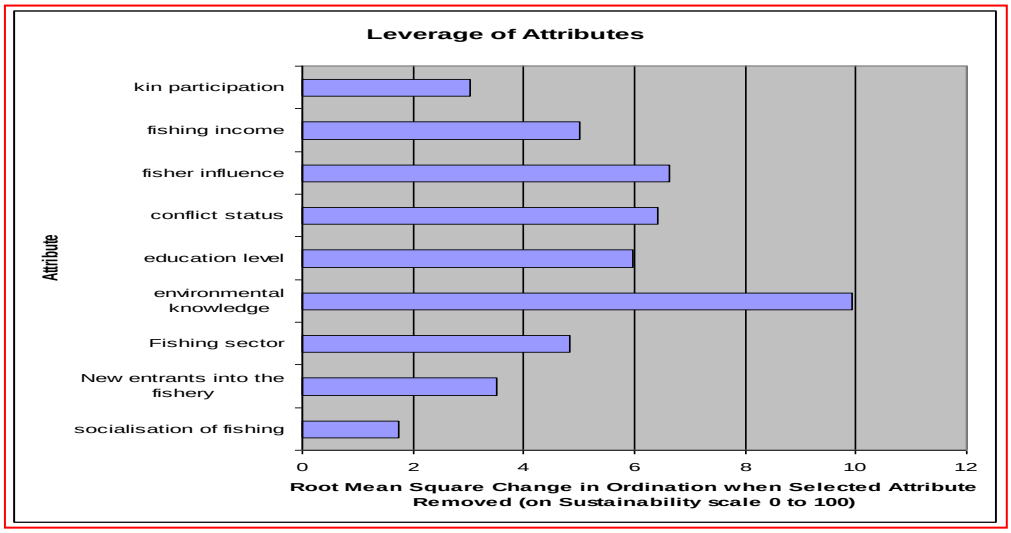


Figure 9. Leverage of attributes

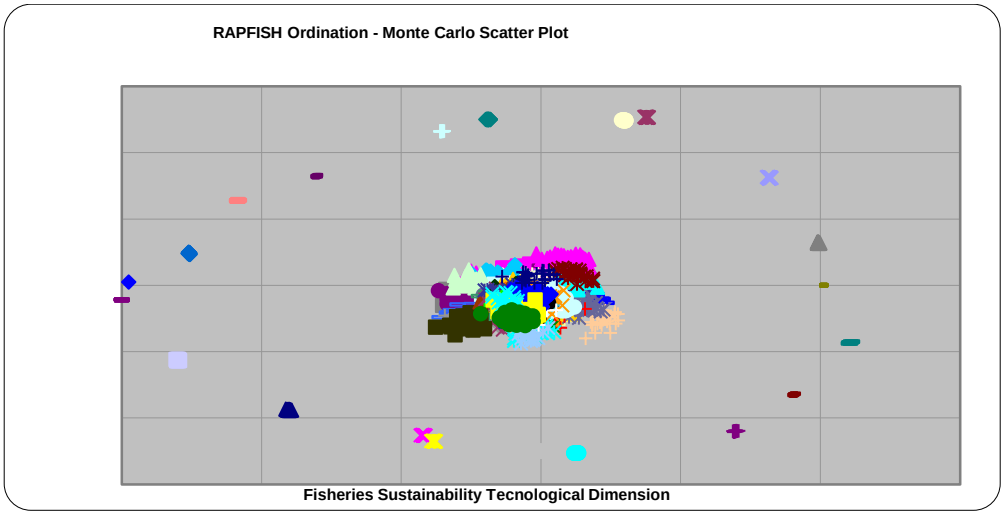
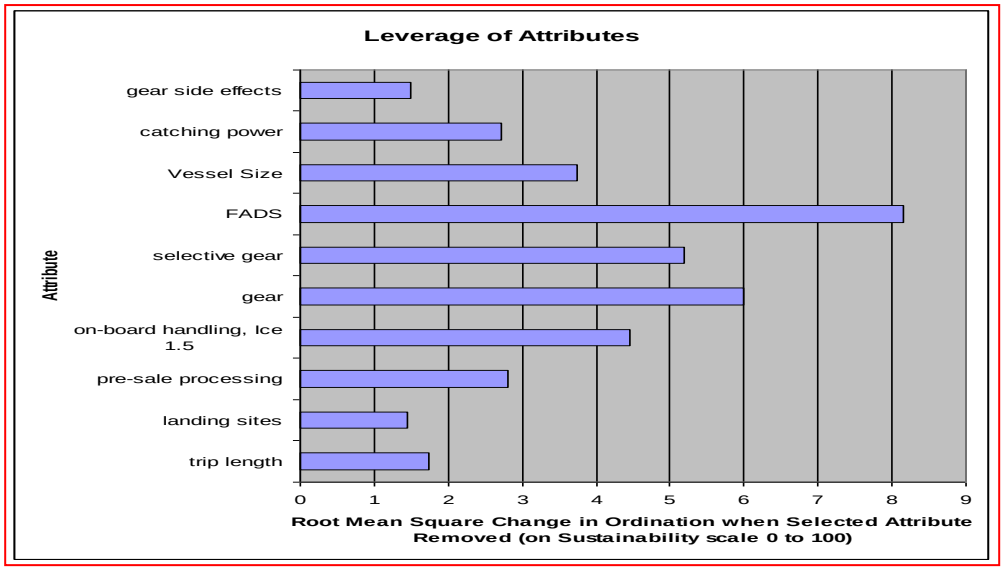


Figure 10. Radfish ordination-Monte Carlo scatter



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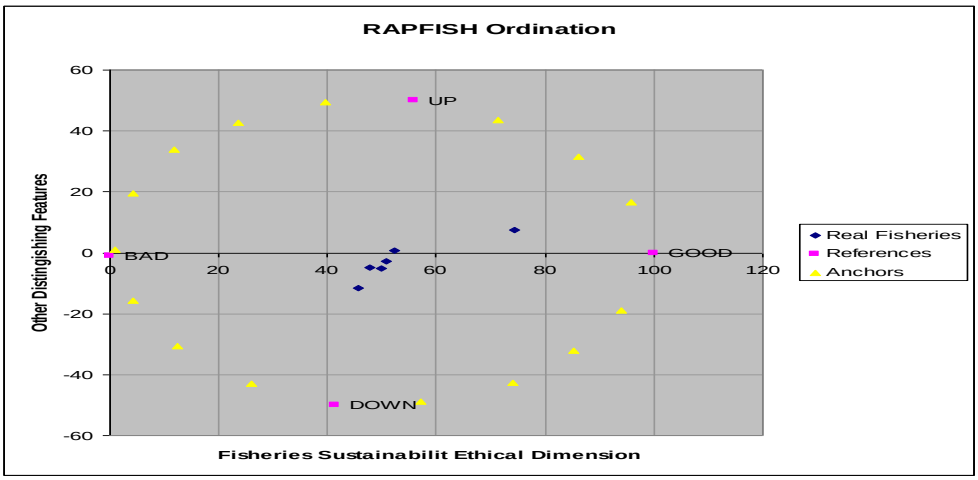

Figure 12. Radfish ordination

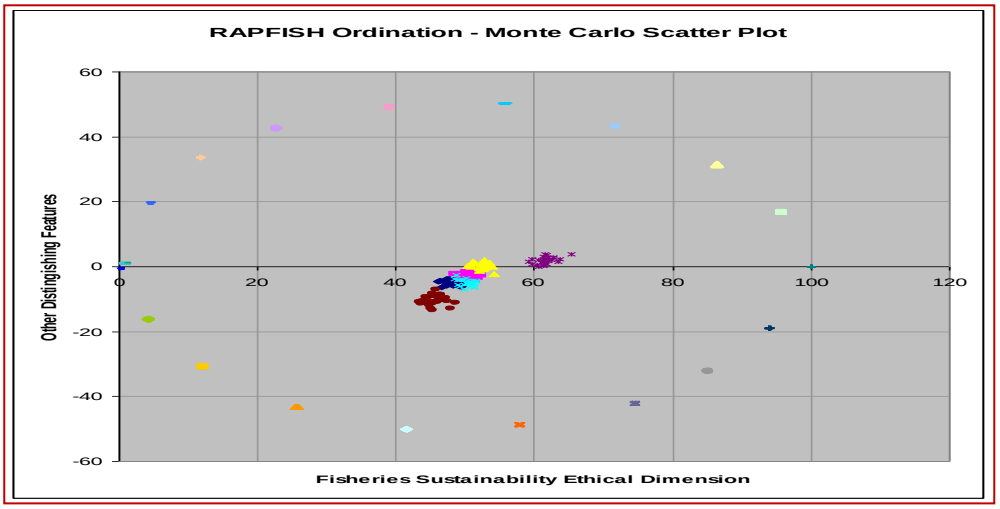

Figure 12. Radfish ordination-Monte Carlo scatter

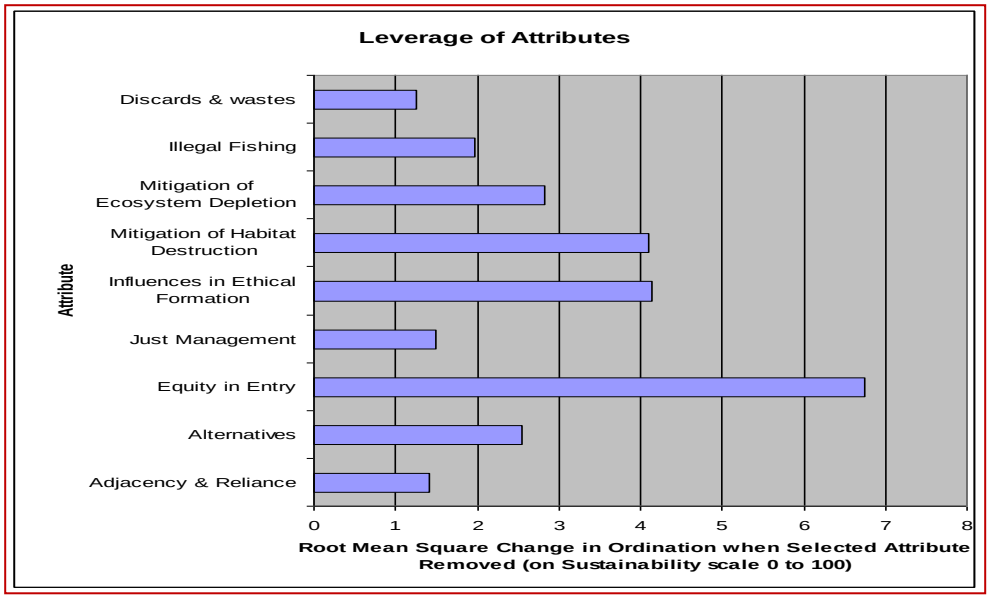

Figure 13. Leverage of attributes

Level of sustainability of management of fishery resources at Region of Pangandaran in West Java Ciamis District from the fifth dimension can be seen in Table 2. Ecological dimension by using nets stupid gear, nets Arad, giil net, trammel net and chart less supportive of the sustainability of fisheries resources while the level of longline fishing in relatively favorable conditions for sustainable management of fishery resources tingakat. The coefficient of determination (R2) of 93.29% means that the ecological dimension has a strong influence and relevance to the sustainability of fisheries resource management. Economic dimension by analyzing the types of fishing gear stupid nets, gill nets, trammel net in conditions unfavorable level of sustainability of fishery resources management while net Arad gear types, longline fishing line and chart the condition sufficient to support the sustainability of fisheries resource management. The coefficient of determination (R2) of 94.11% means that the economic dimensions of belonging and a strong link to the sustainability of fisheries resource management.

The social dimension with a type of fishing equipment nets stupid, Arad nets, trammel nets, and chart the condition of unfavorable levels of sustainability of fisheries resource management, while gill nets and longline fishing in the state sufficient to support the sustainability of fisheries resource management by (R2) of 90.55% means that the social dimension has a strong influence and relevance to the sustainability of fisheries resource managemen

Dimensions of technology with trammel net gear types and charts are in bad condition, while net stupid, Arad nets, gill nets and longline fishing in the state sufficient to support the sustainability of fisheries resource management by (R2) of 90.30% means that the dimensions of technological influence and relevance strong on the level of sustainable management of fishery resources. Ethical dimension to the type of fishing gear stupid nets, trammel net and chart in conditions less supportive while Arad nets, gill nets and longline fishing in the state sufficient to support the sustainability of fisheries resources

management, with (R2) of 92.68% means that ethical dimensions have an influence and a strong link to the sustainability of fisheries resource management.

Table 2. Matrix of Level Sustainability of Capture Fisheries Resources Management at Pangandaran Region Ciamis District of West Java Province.

Type of Trap	***Jaring***	***Jaring***	***Giil***	***Trammel***	***Pancing***	
Dimensi	**Dogol**	**Arad**	***Net***	***Net***	**Rawai**	**Bagan**
Ekological	45,89	48,82	46,94	42,53	51,61	35,14
The square Correlation Value (R^2)	93,29	93,29	93,29	93,29	93,29	93,29
Stressvalue /goodness of fit in MDS	15,90	15,90	15,90	15,90	15,90	15,90
Status of Level Sustainability	Less	Less	Less	Less	Sufficent	Less
Economic	46,94	62,62	54,19	59,22	67,64	65,37
Type of Trap	***Jaring***	***Jaring***	***Giil***	***Trammel***	***Pancing***	
Dimensi	**Dogol**	**Arad**	***Net***	***Net***	**Rawai**	**Bagan**
The square Correlation Value (R^2)	94,11	94,11	94,11	94,11	94,11	94,11
Stres Values/ goodness of fit in MDS	13,88	13,88	13,88	13,88	13,88	13,88
Status of Level Sustainability	Less	Sufficent	Less	Less	Sufficent	Sufficent
Social	47,55	41,87	65,40	49,32	72,34	34,69
The square Correlation Value (R^2)	90,55	90,55	90,55	90,55	90,55	90,55
Stress Value/goodness of fit in MDS	15,00	15,00	15,00	15,00	15,00	15,00
Status of Level Sustainability	Less	Less	Sufficent	Less	Sufficent	Less
Teknological	51,74	59,49	54,61	42,13	60,04	34,02
The square Correlation Value (R^2)	90,30	90,30	90,30	90,30	90,30	90,30
Stress Value/goodness of fit in MDS	17,15	17,15	17,15	17,15	17,15	17,15
Status of Level Sustainability	Sufficent	Sufficent	Sufficent	Bad	Sufficent	Bad

Ethical	47,81	50,84	52,33	49,85	74,46	45,55
The square Correlation Value (R^2)	92,68	92,68	92,68	92,68	92,68	92,68
Stress value goodness of fit in MDS	19,56	19,56	19,56	19,56	19,56	19,56
Status of Level Sustainability	Less	Sufficent	Sufficent	Less	Sufficent	Less

Conclusions

Level of sustainability of fishery resource management dimensions of ecological, social, technological and ethical in poor sustainability of fishery resources while supporting the economic dimension in condition enough to support the sustainability of fisheries resources. Conditions of fishing at Region of Pangandaran in West Java District Ciamis tends to decrease production, which must be anticipated by improving fish habitat, mangrove forest conservation, improved understanding and awareness of fishing for stabilizing the beach.

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Remediating The Degraded Land Due to Mining of Pumice Stone in The Northern Part of Lombok Island by Applying Silicate Rock-Organic Fertilizer

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Abstract

This research was aimed to identify the effects of the application of a remediating agent which was a mixture of ground basaltic-silicate rock powder with manure, termed as silicate rock-organic fertilizer (SROF), on soil quality of the degraded land due to mining of pumice stone in the northern part of Lombok Island. The degraded land was physiographically fixed, manually cultivated, and terraced accordingly to the land sloping. A completely randomized block design was applied with a treatment of five application rates of the remediating agent (0 – 10 t ha^{-1}). A week after application of the remediating agent, corn (var. BISI 2) was grown on the land. Soil organism-respiration rate was measured at 45 and 90 days after planting and several soil chemical properties were identified after crop harvesting. Results indicated that application of the remediating agent (SROF) significantly increased soil organism activity, soil cation exchange capacity, growth and yield of corn. It was concluded that the mixture of basaltic-silicate rock powder and manure may be used as an effective remediating agent to rebuild the physical, chemical as well as biological properties of degraded land due to mining.

Keywords: ameliorant, degraded land, pumice stone, remediation, silicate rock

Introduction

Mining of pumice stone in Northern Lombok – NTB, Indonesia, has destroyed hectares of farming land in the area. To rebuild the degraded land requires effective method and materials. Based on the local condition, it was proposed that basaltic rocks and organic matter (manure) – the environmentally sound and locally available materials may be used as the most appropriate remediating agent.

The finely ground basaltic rocks or silicate rock fertilizer (SRF) has been known to have multi functions, e.g., as a source of almost all nutrients essential for plants (except N) or multinutrient fertilizer and a general soil ameliorant (Priyono, 2005). Another well known natural material having multi functions similarly to SRFs is organic matter or manure.

A numbers of researchers showed that the application of SRFs increased pH of acidic soils (Holdren and Berner, 1979; Gillman, 1980; Gillman *et al*., 2001 and 2002; Leonardos *et al*., 1987 and 2000; Wang *et al*., 2000; Coventry *et al*., 2001; Harley, 2002; Priyono *et al*., 2002; Priyono *et al*., 2007), cation exchange capacity (Gillman *et al*., 2001 and 2002; Coventry *et al*., 2001), and reduced Al-toxicity effects and P adsorption by polyvalent cations (Mn, Fe, Al) in acidic soils (Coventry *et al*., 2001). The application of SRFs also increased organism

activity in acidic to neutral soils (Priyono *et al*., 2007) and reduced salt effects of saline-sodic soils on plant growth (Sumarlin, 2008). Another potentially beneficial effect of using SRF was associated to its high content of Si, i.e., 4 – 8 % Si extractable in 0.01M citric-oxalic acid (Priyono and Gilkes, 2008a). Results of a greenhouse experiment (Priyono and Gilkes, 2008b) indicated that the application of SRFs equivalent to 5 – 10 $t.ha^{-}$ increased 2 – 4 fold of the uptake of Si by ryegrass. The other proves were that applying Si-high containing fertilizers on several crops, mainly grasses increased the tolerance of the crops to pathogen attacks (Volk *et al*., 1958; Epstein, 1999) and reduced the toxicity effects of Al on corn (Corrales *et al*., 1997).

Based on above findings, it was presumed that the application of a mixture of SRF and manure, termed as silicate rock-organic fertilizer (SROF), had more effective effects in improving soil properties being suitable for plant growth than those materials were applied individually. Dissolution of plant nutrients from SRF was accelerated by the present of organic acid (Priyatna *et al*., 2007) or organic matter (Priyono *et al*., 2007), while the decomposition of organic matter that produced organic acids were speed up by adding SRF. Therefore, SROF was proposed to be the cheap, effective, and environmentally sound material to rebuild the physically, chemically, and biologically degraded land due to mining. The objectives of the research were to identify effects of application rate of SROF on soil quality as indicated by soil biological activity (respiration rate) and chemical properties, growth and yield of crop (corn).

Materials and Methods

Land Preparation

The research was carried out in Akar-Akar Village, in the northern part of Lombok Island, Indonesia from May to August 2009. The degraded land (0.5 ha) was prepared for the experiment by fixing its physiographical condition, i.e., by manual smoothening, cultivating (by using a hand tractor), and then terracing accordingly to land sloping. The terraces were functioned as border line of the experimental blocks. Each block was divided into 5 experimental plots of 25 m x 13 m each. Among the plots were separated by 50cm-width and 30cm-height border lines. The general condition of the degraded land and that after being smoothen and terraced are shown in Figure 1.

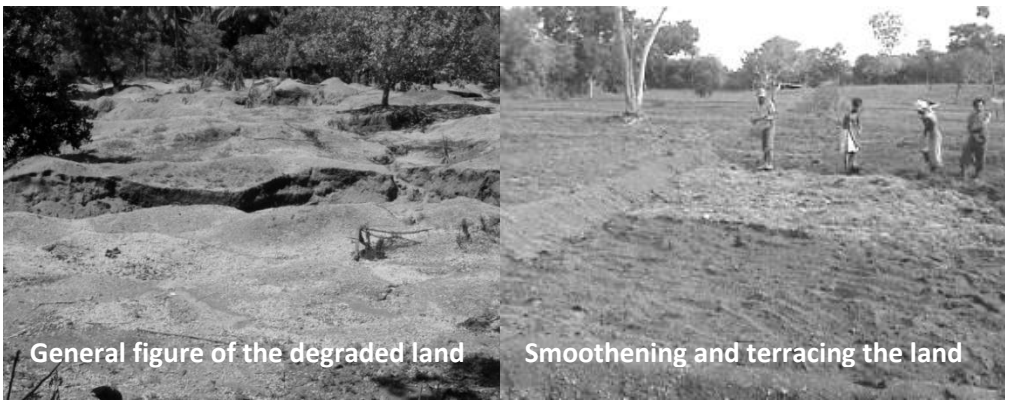

Figure 1. General condition of the degraded land in the northern part of Lombok Island (left) and the land after being smoothen and terraced (right)

A sprinkle irrigation system was prepared for watering in this experiment by utilizing a deep-ground water wheel of 80m depth. Water was pumped from the wheel at rates of 18 - 20 L s^{-1}. A sprinkle outlet was set in each experimental plot by connecting the PVC pipes to the main water line and then to the water pump.

Preparing Remediating Agent

Basaltic-silicate rocks – the quarry by product (Ø < 1 cm) was ground by using an attrition ball mill. The rock material (2 kg) and 5 kg of stainless steel balls (Ø = 22 mm) were put into a ball mill vial having capacity of 18.8 L. The mill was operated at speed of about 150 rpm for 30 minutes. This milling procedure was run several times to have enough amount of rock powder for this experiment. Chemical and mineral compositions of the rock powder are presented in Table 1. To provide a remediating agents (SROF), the rock powder was mixed with dry composted cattle wastes (manure) at a ratio of rock powder : manure = 1 : 4.

Experimental Design

A completely randomized block design consisting of 3 blocks was applied with a treatment of the application rates of SROF (0, 2.5, 5, 7.5, and 10 t ha^{-1}). SROF was evenly applied into about 5-cm depth of planting rows and were covered with soil, moisten to and maintained at field capacity for a week. Corn (var. BISI 2) was planted 1 seed per planting hole in the plant spacing of 20 cm x 80 cm. Basal fertilizers consisting of N and P in forms of urea (138 kg N ha^{-1}) and superphosphate-18 (27 kg P_2O_5 ha^{-1}) were applied. Crop watering by using sprinkle irrigation system was carried out in each 3 – 4 days, depending on soil moisture condition and growing stage of the crop. Weeding was carried out twice at 21 and 45 days after planting (d.a.p), and harvesting was in 97 d.a.p.

Table 1. Chemical and mineral compositions of rock powder used in this experiment

Main elements (% oxide) *	**SiO_2 52.3**	**Al_2O_3 24.8**	**CaO 4.8**	**MgO 1.8**	**K_2O 6.3**	**Na_2O 3.3**	**FeO 6.2**	**MnO 0.15**	**ZnO 0.01**	**CuO 0.33**	**others < 0.01**
Main minerals **	Gendrite; Fargasite; Albite; Anorthite; Muscovite										
Particle size (µm)			< 10	10–20	20-30	30-40	40–60	60–80	> 80		
Proportion (%) ***			28	5	5	6	8	8	40		

* Wet destruction by using HF (modified Jackson, 1956).
** Interpreted from XRD (X-ray diffraction) patterns.
*** Pipette method (modified Gee and Bauder, 1986).

The observed parameters in this experiment were (1) respiration rate of soil organism, (2) several chemical properties of residual soil (after harvesting), (3) weight of dry matter crop, and (4) yield of corn. The respiration rate of soil organism as an indicator of general soil quality was measured directly in field condition at 45 and 90 d.a.p. The measurement was carried out on 6 points in each plot. A day before measurement, soil was watered to field capacity. A 20cm-lenght of top-covered PVC pipe (Ø = 5 inches) was inserted into the ground up to 10cm-depth. The gas of CO_2 produced from soil organism respiration processes in 24 hours was collected by flowing the gas through a plastic line (Ø = 5 mm) into a plastic bottle containing 0.1N KOH, and then the quantity of CO_2 was measured with acid-base titration method (using 0.01N HCl).

Several chemical properties of soils in initial state (before planting corn) and those for residual soils (after harvesting) were identified. Soil acidity (pH_{H2O} 1 : 2,5) was measured by

using pH-meter, total C-organic by using dichromate oxidation (Walkey and Black, 1935), N total with Kjedahl, and extractable P with Bray II methods. Cation exchange capacity (CEC) of soil was measured by using a buffer solution of 1N ammonium acetate pH 7, and concentration of the exchangeable basic cations (Na, Ca, Mg, and K) in the filtrates of the extraction was measured with AAS.

Results and Discussion

Respiration Rates

Statistically, the application rate of SROF significantly increased soil organism activities (respiration rate). The relationships between respiration rate and application rate of SROF are presented in forms of linear regression in Figure 2.

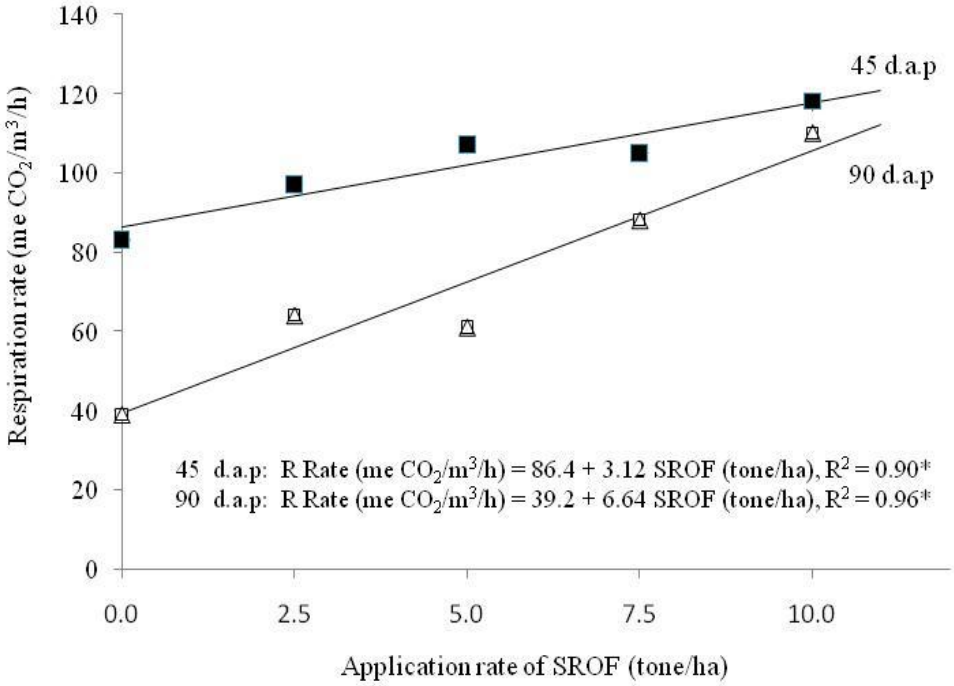

Figure 2. The Relationship between the application rate of silicate rock-organic fertilizer (SROF) and the average of respiration rate at 45 and 90 d.a.p.

As shown in Figure 2, the respiration rate at 45 d.a.p. is higher than for that of at 90 d.a.p. However, the intensity effect SROF application to the respiration rate at 45 d.a.p. was lower than for that at 90 d.a.p. Based on the regression equations, each adding 1 t ha^{-1} of SROF increased the respiration rate up to about 3.12 me $CO_2.m^{-3}h^{-1}$ for the measurement at 45 d.a.p. and about 6.64 me CO_2/m^3/hour for the measurement at 90 d.a.p. Clearly, remediation in relatively short period by using SROF up to 10 t ha^{-1} significantly improved soil quality of the degraded.

Changes of Soil Chemical Properties

Several chemical properties of soils at initial condition were compared to those after corn harvesting (Table 2). As shown in Table 2, clear changes of soil properties due to the application of SROF were the increases of soil C-organic content and CEC, while soil pH and exchangeable cations tend to decrease. The increase of C-organic content could be due to the direct effect of adding SROF as well as root exudates of the crop, which the increases of both constituents in soil may contribute to the increase of soil CEC.

Table 2. Several chemical properties of soils before remediation and after corn harvesting

No	Soil Properties	Unit	Initial Condition	Application Rate of SROF ($t.ha^{-}$)				
				0	2.5	5.0	7.5	10
1.	pH_{H2O} (1:2.5)	-	5.86	5.60	5.53	5.61	5.48	5.56
2.	C-organic	%	0.20	0.34	0.42	0.26	0.51	0.44
3.	N-total	%	0.02	0.01	0.02	0.02	0.01	0.01
4.	Extr.-P	$mg.kg^{-}$	2.80	3.20	4.10	6.40	4.80	2.80
5.	CEC	$cmol_c.kg^{-}$	4.50	4.80	5.20	5.26	5.55	6.06
6.	Exch. Ca	$cmol_c.kg^{-}$	3.80	4.20	3.74	3.34	2.93	2.23
7.	Exch. Mg	$cmol_c.kg^{-}$	0.70	0.66	0.52	0.31	0.43	0.62
8.	Exch. K	$cmol_c.kg^{-}$	0.56	0.41	0.48	0.62	0.22	0.32

If the direct effect of root exudates on CEC was ignored, each adding 1 t ha^{-1} of SROF will increase CEC up to about 0.6 $cmol_c.kg^{-}$. Practically, the increase of CEC has important meaning in association to the general soil properties in Northern Lombok, e.g., coarse-sandy textured and porous, which adding SROF may reduce plant nutrient loss from the soil due to leaching, mainly during the rainy season.

Corn Growth and Yield

The average weights of dry biomass, yield, and 1000 seeds are presented in Table 3. Statistically, the application rate of SROF significantly increased those three crop components.

Table 3. The average weights of dry biomass, yield, and of 1000 seeds of corn

Ratec of SROF	Dry Biomass	Yield	1000 Seeds
	…….($t.ha^{-}$) ……	……..(g) …….	
0.0	2.46 a	0.95 a	216.6 a
2.5	3.38 b	1.28 b	223.3 b
5.0	3.36 b	1.26 b	236.7 c
7.5	4.06 c	1.38 b	246.7 d
10.0	5.02 d	1.41 b	246.7 d

Note: values in the same column followed by the same letter were not significantly different based on the test of LSDα = 0.05.

As shown in Table 3, the average of dry biomass was nearly doubled due to adding SROF of 10 t h^{-1}, e.g., from 2.46 to 4.06 t h^{-1}. Total yield and weight of 1000 seeds each was increased about 40 %, e.g., from 0.95 to 1.41 t h^{-1} for yield and 216.6 to 246.7 g for weight

of 1000 seeds. Clearly, the application of SROF increased corn growth, quantity and quality of corn yield.

Although the total yield of corn (var. BISI-2) in this experiment was very low (< 2 tones/ha) relative to the potential yield of the corn variety (7 – 8 t h^{-1}), the application of SROF significantly improved the productivity of the land. To restore the degraded land into highly productive land may be need 3 – 4 times of SROF application into the soils.

Conclusion

The application of a mixture of basaltic rock powder and manure (SROF) up to 10 t ha^{-1} significantly increased soil quality and productivity of the degraded land due to mining of pumice stone (soil parent material) in Northern Lombok – NTB. The mixture (SROF) can be used as an effective and locally available remediating agent to rebuild the physically, chemically, and biologically degraded lands.

Acknowledgement

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Evaluation of Extension Worker's Attitude toward Integrated Farming System in Indonesia

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Abstract

This study aimed to determine the access of information channels and attitude of extension workers toward Integrated Farming System (IFS). The study used a survey design. Data were collected from 184 extension workers Sleman and Bantul districts, in Yogyakarta Province. F- test and Least Significant Difference (LSD) test, were employed to test hypothesis. Results revealed that extension workers accessed agricultural magazine rather than newspaper, journal of agricultural research and internet to gain information toward IFS. It was found that extension workers' attitude towards IFS was at high level. Hypotheses testing indicated that extension workers' attitude toward IFS by accessing journal of agricultural research was significantly different from which by accessing newspaper, agricultural magazine, and internet.

Keywords: access of information channels, attitude, extension workers, integrated farming system

Introduction

Indonesia has formulated national planning in terms of agricultural development based on existing problems such as restrictiveness of agricultural resources and infrastructures, weakness of capital accessibility and farmer institution, lack of transfer knowledge and technology, weakness of farmer participatory and movement and inefficiency of agricultural product marketing (Bappenas, 2007). Recently, the country is also encountering a threat of food crisis because of the climate change so that it will rapidly decrease the agricultural production by high level of crop failure and livestock mortalities (Subejo, 2009). Therefore, awareness from government is needed then the appropriate strategy should be applied to suffice the food demands in the future.

Currently, government has reintroduced a strategy which emphasizes the diversification in farming systems. The strategy, known as Integrated Farming System (IFS), is based on the indigenous knowledge which has been practiced long years ago by farmers. However, the farmers have ignored this indigenous pattern since the introduction of monoculture as new technology which is high yields oriented. Consequently, it causes ecological problems which can be alleviated by the adoption of IFS (Edwards, 1989). In Thailand, adoption of IFS has become important by combining rice farming with livestock and aquaculture enterprises to sustain agriculture development and to prevent resource degradation and food insufficiency in the future (Tipraqsa *et al*, 2007). In Europe, implementation of IFS has significantly improved the agro ecosystem and has provided significant evidence on the

worth of IFS to farmer, society, and the environment (Edwards, 1989). In Indonesia, an IFS approach has been developed for sustainable improvement and productivity based on crops and livestock in which food production for local consumption is needed (Adiningsih and Karama, 1992).

The implementation of IFS is highly dependent on extension workers' role as important facilitator and agent of change in the community (Steyn and Nunes, 2001; Rogers, 1983). It is because, in doing their job, extension workers provide extension services to the farmer by transferring new knowledge, integrating research and practice, connecting innovation and application, and also implementing the policy into farmer community (Hoque and Usami, 2007; Boonjing, 2008). The implementation of the strategy requires the active extension workers in their important role for the success.

The active extension workers can be illustrated by having positive behavior and high performance efficiency which are determined by attitude (Liaghati, 2008). In agricultural context, the attitude of extension workers is often based on quality and quantity of the information availability and the environment particularly in working area (Hyytia and Kola, 2006; Putra *et al*, 2011). It obviously indicates that the positive attitude of extension workers can be manipulated by policy mechanism and intervention by the government in decentralization system (Liaghati, 2008; Hyytia and Kola, 2006; Putra *et al*, 2011) particularly in Indonesia. The key role of extension workers should be noted because they are disseminator of strategy and policy which have been set up by the government.

Therefore, extension workers, as frontliners for applying IFS in farming area, have to improve their capabilities. One thing to improve the capability is by keeping frequently updated of new information because information is the essential ingredient providing the raw material in order to build knowledge structure (Potter, 2001). Information, as a message, will be delivered to extension workers through channel in which adaptable to the content of message with most impact to receivers (Ongkiko and Flor, 2003). By the new challenges such as new communication and information technology, and participation and decentralization, the better selection of information and communication channels is the more effectiveness of extension programs in agriculture (Allahyari, 2008; Saadi *et al*, 2008). Actually, extension workers usually access magazine, newspaper, journal of agricultural research, and internet to gain knowledge on IFS at different intensity (Putra *et al*, 2011). Magazine and newspaper become very popular to get information in conservation issues while journal of agricultural research is a very trusted channel (Yiridoe, 2010; Saadi, 2008). Meanwhile, internet as a popular new information channel in which contains unlimited knowledge is still ineligible to access in some particular area (Saadi, 2008). Nonetheless, it becomes very interesting to study the access of information access of extension workers toward IFS.

Sleman and Bantul districts in Yogyakarta Province are selected for the study. They have 70.43% or 224,398 hectares of agricultural area which are highly vulnerability to natural disaster. The local governments have applied IFS as a policy in agricultural development to retain the role as the highest rice producer in the province (BPS, 2009). So, the conducted study aims to determine the access of information channels and the attitude of extension workers toward IFS.

Materials and Methods

This research was designed as a survey research by using questionnaire to collect the data. Research was conducted at Sleman and Bantul, Yogyakarta Province from April to May 2010. The population of research was extension workers in those two districts. It consisted of 337 personnel; of whom 157 personnel were from Bantul and 180 personnel were from Sleman. By proportional stratified random sampling technique and computing by Yamane's formula (1967), 184 respondents were selected from the population. A questionnaire was used to collect data consisted of close and open ended questions. It was divided in to three parts; First was characteristics of respondents consisting of question about gender, age, educational background, working experience, monthly income, and working area; Second was access to information channels from which the respondents get their information about IFS. Four common information channels for extension workers such as newspaper, agricultural magazine, journal of agricultural research, and internet were offered to the respondents. Third was attitude of extension workers toward IFS consisting of 30 statements scored by Likert scale. The rating scale was measured in three intervals which were low level, medium level, and high level of attitude toward IFS. The reliability test of research instrument had been conducted resulting 0.86 as Cronbach Alpha Coefficient reliability. It meant that the questionnaire was reliable to gather information toward respondents. Furthermore, data were analyzed and presented by frequencies, percentages, mean, and standard deviation. ANOVA analysis was used for inferential statistical analysis to test the different attitude of extension workers toward IFS with different access of information channels.

Results and Discussion

Characteristics of respondents

The extension workers selected as respondent comprised of male (72.3%) and female (27.7%). The composition based on age was lower than 40 years old (38.1%) followed by 40 to 50 years old (32.6%), and more than 50 years old (29.3%) respectively. Meanwhile, the educational background of respondents was graduated from higher education (51.6%), diploma (26.6%) and high school certificate holder (21.7%). Most of respondents had 16 years to 23 years working experience (53.3%) and 1 year to 15 years (46.7%). The monthly income earned by extension workers was in the range from Rp. 1,000,000,- to Rp. 2,000,000,- (53.8%) and the rest were in range from Rp. 2,000,001,- to Rp. 4,000,000,- (46.2%). Of those respondents were randomly selected from both Bantul (46.7%) and Sleman (53.3%).

Access of information channels

Access of information channels of extension workers was the access of respondents to media which were newspaper, journal of agricultural research, agricultural magazine and internet. The results as shown on table 1 revealed that most of respondents accessed agricultural magazine (53.3%), followed by respondents who accessed internet (17.9%) and newspaper (17.4%). The least accessed was journal of agricultural research (11.4%).

Table 1. Extension workers classified by access of information channels to get information about IFS (n = 184)

Information Channels	Number	Percent
Newspaper	32	17.4
Journal of agricultural research	21	11.4
Agricultural magazine	98	53.3
Internet	33	17.9

Attitude of extension workers toward IFS

According to results shown on Table 2, respondents had overall attitude toward IFS at high level (overall mean = 4.07). It showed that 26 (twenty six) items of attitude toward IFS were at high level and 4 (four) items were at medium level. The three highest attitude's mean toward IFS were (item 28) the usage of livestock to support the organic matter on soil management (mean = 4.50), (item 17) the necessity of farmer participation in groups (mean = 4.47), and (item 8) the important role of extension workers toward IFS implementation (mean = 4.39). Moreover, 4 (four) items at medium level were (item 3) implementation of IFS by adding the awareness of natural disaster mitigation (mean = 3.67), (item 25) provision of subsidies and credit to farmers by government (mean = 3.50), (item 30) IFS has been adopted appropriately by farmer (mean = 2.97), and (item 1) high vulnerability of natural disaster in farming area (mean = 2.66). It, however, described that extension workers had high level attitude toward the implementation of IFS.

Table 2. Attitude of extension workers toward IFS

Items	Statements	Mean	S.D.	Attitude Level
1	High vulnerability of natural disaster in farming area	2.66	1.01	Medium
2	Land cultivation by IFS as an appropriate solution in Yogyakarta	4.34	0.51	High
3	Implementation of IFS by adding the awareness of disaster mitigation	3.67	0.79	Medium
4	IFS as issued policy by government in agricultural development	3.89	0.77	High
5	The positive impact of IFS for the future	4.06	0.74	High
6	The implementation of IFS effectively supported by government	4.29	0.54	High
7	The assistance of extension workers to IFS application by farmers	4.23	0.51	High
8	The important role of extension workers toward IFS implementation	4.39	0.59	High
9	Provision of technical assistance from extension workers to farmers	4.26	0.49	High
10	The support of IFS to sustainable agricultural development process.	4.22	0.76	High
11	The promotion of IFS on perennial crops and livestock based	4.13	0.56	High
12	Role of extension workers on synergizing stakeholders in agricultural	4.05	0.54	High

13	IFS gives more benefit to farmer's household	4.29	0.47	High
14	The need of continuous improvement on IFS	4.27	0.56	High
15	The role of extension workers on experience-based continuous improvement process.	4.08	0.87	High
16	The more advantages to the social capital of farmer's communities	4.22	0.44	High
17	The necessity of farmer participation in groups	4.47	0.55	High
18	Contribution of farmer groups to IFS success	4.19	0.47	High
19	Ecological balance as one of IFS purposes	4.29	0.51	High
20	The need of balance interaction between nature and human	4.28	0.82	High
21	IFS applied by interacting farmers and their land in balance	4.09	0.59	High
22	The need of economics growth by increase of productivity	4.11	0.60	High
23	Opportunity of farmers to increase the yields in cultivation process	4.18	0.49	High
24	The more income from agriculture sector when applying IFS	4.01	0.59	High
25	Provision of subsidies and credit to farmers by government	3.50	0.96	Medium
26	The need of intensive agriculture both upland and lowland area	4.18	0.48	High
27	The Combination crops and livestock on land cultivation	4.13	0.67	High
28	The usage of livestock to support the organic matter on soil management	4.50	0.53	High
29	Five times higher profit than conventional farming by IFS	4.23	0.54	High
30	IFS has been adopted appropriately by farmer	2.97	0.99	Medium
	Overall Mean	**4.07**	**0.29**	**High**

Extension worker's attitude on access of information channels

The comparison of overall attitude mean with different access of information channels using one-way ANOVA (table 3) illustrated that extension workers with different access of information channels had different attitude toward IFS ($p < 0.05$). Meanwhile, the continued LSD test for multiple comparisons of extension workers' attitude toward IFS by access of information channels (table 4) indicated that extension workers' attitude mean toward IFS by accessing journal of agricultural research was significantly higher than of which accessing newspaper ($p < 0.01$), agricultural magazine ($p < 0.05$), and internet ($p < 0.01$).

Table 3. Comparison of overall attitude mean toward IFS of extension workers classified by access of information channels using one way ANOVA (n = 184)

Attitude by information channels	n	Mean	S.D.	F	p-value	Interpretation
Newspaper	32	120.72	9.08	3.065*	0.029	Significant
Journal of agricultural research	21	127.05	9.90			
Agricultural magazine	98	122.39	8.49			
Internet	33	120.39	7.24			

*Significant at the 0.05 level

Table 4 Multiple comparisons of extension workers' attitude toward IFS by access of information channels using LSD test (n = 184)

Attitude by information channels		Information Channels			
		(1)	(2)	(3)	(4)
	Mean	120.72	127.05	122.39	120.39
(1). Newspaper	120.72	--			
(2). Journal of agricultural research	127.05	**	--		
(3). Agricultural magazine	122.39	Ns	*	--	
(4). Internet	120.39	Ns	**	ns	--

ns means not significant at 0.05
* means significant at 0.05
** means significant at 0.01

The characteristics could describe the background of respondents in this study. Result showed that 72.3% respondents were male which indicated the gender gap between male and female in this area. Nonetheless, since the extension workers were the field workers with high responsibility and mobilization, this profession was dominated by male more than female (Asadi *et al*, 2008; Allahyari, 2008). Unfortunately, more than 60% extension workers were 40 years old which implied they were also, currently, getting older (Hoque and Usami, 2008; Asadi *et al*, 2008). In educational background issue, it could be recognized that most of respondents had higher educational degree. It indicated that the human resources development for extension workers was in the appropriate level (Asadi *et al*, 2008).

Extension workers had been working long years in their profession (Leta *et al*, 2005) based on the evidence that most of respondents had been working as extension workers for more than 15 years by now. Although they had much experience, their monthly income was still at lower level. It will impact to their satisfaction in doing their job indeed because monthly income contributes in explaining the job satisfaction of extension workers (Asadi *et al*, 2008; Leta *et al*, 2005).

As shown on result, most of respondents (70.7%) accessed magazine and newspaper for gaining information on IFS. It might seem that agricultural magazine and newspaper could deliver the attractive message instead of journal agricultural research. Newspaper and agricultural magazine often published the popular knowledge in agricultural area so that it was preferred by respondents for enriching their knowledge. Meanwhile, the use of

newspaper and magazine was also very important for accessing the information about IFS which was closed to the conservation issues. Yiridoe (2010) argued that newsletters and agricultural magazines were the most important communication channels for accessing information on conservation practices and magazine usually contained popular information rather than unpopular. It implied that IFS issue was becoming popular through the extension workers at Bantul and Sleman.

The interesting finding on access of information channels illustrated that the access on internet was lower than access on agricultural magazine. Although the internet currently provided unlimited information toward IFS, the number of extension workers who accessed to internet was not quite different from those who accessed newspaper. It might indicate to the internet illiteracy of extension workers and ineligibility of access (Saadi *et al*, 2008). On the other hand, the most trusted information provider, journal of agricultural research, (Saadi *et al*, 2008) was only accessed by a few extension workers. Nevertheless, as long as the IFS became a policy of government in agricultural development, newspaper was an appropriate mass medium for accessing the dissemination of the policy because it could widely be accessed by common people (Sanya-Arj, 2002).

According to findings, extension workers had high level attitude toward IFS. The result might indicate that extension workers function as innovator in every innovation because of their role as change agents. Extension worker was one of the stakeholders of agricultural development that should have appropriate and progressive attitude toward technological innovation to be an integral part of sustainable agriculture (Aerni *et al*, 2009; Liaghati, 2008).

Furthermore, we could break down on which particular attitude contributed to the overall high attitude of extension workers toward IFS. It had been showed that the three highest attitude's mean toward IFS were on the usage of livestock to support the organic matter on soil management, the necessity of farmer participation in groups, and the important role of extension workers toward IFS implementation. It seemed that implementation of IFS required integration of social and technical aspects (Farmia, 2008). Technical side such as raising livestock, however, could supply the organic matter for soil management while non-technical sides such as extension workers and farming group played the important role in implementation to ease the adoption process at the grassroots level (Farmia, 2008; Steyn and Nunes, 2001; Onasanya, 2006)

The finding about statistical test showed that the attitudinal mean of extension workers toward IFS by accessing journal of agricultural research was significantly higher than by accessing agricultural magazine, newspaper and internet. The findings might indicate that the main responsibility of extension workers was to link from the research at research center to the implementation at field. Unfortunately, journal of agricultural research was at lowest percentage accessed by extension workers. Aside from unpopular information inside, it seemed that they didn't much understand their role as agent of change in farmers' community (Rogers, 1983). As extension workers, indeed, they should have aptitude on knowledge, leadership, emotion, skill, and smartness which cover the strategic planning, comprehensive knowledge, advance analytical thinking, intelligent, and knowledge management, because of those, extension workers would transform innovation by communicating the research findings to the implementation as facilitator (Boonjing, 2008; Steyn and Nunes, 2001). It emphasized extension workers to access the journal of agricultural research as the channel to publish research in agricultural area because, in fact,

the access of journal of agricultural research more likely implied to the higher level of attitude toward IFS.

Conclusions

Magazine and newspaper are the most access of information channels by extension workers for gaining the information toward integrated farming system. More than 70 % of respondents access magazine and newspaper rather than internet or journal of agricultural research. It is supported by the fact that, in rural working area, the ineligibility access of internet and computer exists. Aside from it, extension workers seem unwanted to access journal proven by the low percentage of respondents despite of high reliability of information provided. They seemed misunderstand with their role as agent of change, moreover, it is because the journal contains unpopular information despite of such kind of innovation instead. However, magazine and newspaper are expectedly opted because of the more popular information presented inside.

Meanwhile, the mass media such as newspaper and magazine become the appropriate channels to disseminate information on government's policy. IFS as local government's policy in Sleman and Bantul, is disseminated effectively by using newspaper and magazine. The large number of extension workers accessing mass printed media confirm the valuable transfer of knowledge toward integrated farming system. The contribution of newspaper and magazine is indispensable toward the adoption of innovation for extension workers.

The high level of attitude of extension workers toward integrated farming system denotes that they have positive response concerning the answer of agricultural problems encountered. Integrated farming system is the step forward of the national planning in revitalizing the agriculture, fisheries, and forestry which can assure the sustainability of agricultural development. The high level attitude of extension workers toward IFS in Sleman and Bantul becomes very important to encourage the farmers to implement the innovation so that the adoption in grassroots level will be effortless to be. Meanwhile, another result also confirms that highest attitude's mean is the raise of livestock is very useful to support the organic matter of soil management for land cultivation, farmer participation is necessary to accommodate in groups, and extension workers have an important role toward IFS' implementation. Those items represent the steadiness emotional response of extension workers for both in technical and social aspects. However, according to the results, raising livestock plays the important role in the technical aspect of farming system while farmer participation in a group is positively addressed in social aspect which underlines that the role of extension workers toward implementation of IFS at farmer level is significant. The findings turn out to be evidence that extension workers have certain attitude of both on social and technical aspects in integrated farming system.

The statistical analysis result demonstrated that the extension workers by accessing journal of agricultural research have significant highest attitude level toward integrated farming system rather than by accessing agricultural magazine, newspaper, and internet. It emphasizes the responsibility of extension workers for linking the research and implementation in the field. Although it is only accessed by a few extension workers, the result illustrates that journal of agricultural research is the important information channel to gain information toward IFS. However, by accessing the determinant information channel on attitude difference, the extension workers can play more strategic role as agent of change and facilitator through the development process in farming area.

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Numerous Factors Influencing Food Availability During Harvesting and Lean Seasons in West Timor

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Abstract

Problem of hunger in West Timor is very seasonal phenomenon that has to be faced by rural households routinely. Food shortage becomes more severe as the trend of food usage for social functions during harvesting season increase. The aims of this research were (1) to analyze the relationship between season, treatment and food security, and (2) to underpin numerous factors influencing food availability. The research was carried out during the harvesting and lean season in 2009 at 96 villages of four regencies in West Timor. Villages were purposively selected based on their potential agricultural product. Villages of each regency then were divided in four treatments, and thus there were six villages for every treatment. Respondents were 30 housewives of poor households randomly chosen. The result showed that food in most of households comes from their own farm, and hence it has short availability time. Therefore, intervention of the program using treatment of credit, pure storage and fixed storage (storage with contract) gave valuable lessons to the households in coping food shortage problems during lean season. Variables of area of possessing land, cash income and the number of productive labor have positive impact on food availability. In contrast, expenditure for festivals or parties negatively influences food availability in households.

Keywords: food availability, food security, lean season, West Timor

Introduction

Food availability in farmer's household is the availability of food physically in a household, which may come from farm, market, or gift, using for daily need of the household. In the village, food availability of households is very much depended on the harvesting of food crops from their own rice field, backyard or farm. The yield of the food crops is influenced by climate especially rainfall and its distribution.

Climactically, East Nusa Tenggara Province has classified as semi arid zone. The rain season is shorter than the dry season. The very short rain season indicates that farmers have very short time to work in their farm. Even though intensity of rainfall around Timor is usually high (in average, 1583 mm per annum during 1985 – 2004), however, its distribution is unbalance and affecting food production. Reports from Pos Kupang Newspaper (August 26-28, 2011) and our observation indicated that lean season of this year in Timor is earlier than before (starting in August) because many villages experienced not only fail to harvest, but also fail to plant.

The production of maize, the main staple food of majority of households in West Timor, in East Nusa Tenggara Province is lower (2,5 ton/ha) than that of National (3,7 ton/ha) (Statistic Indonesia, 2010). Phenomenon as above is routine for farmer's households in

West Timor villages. This condition becomes worse as consequence of over using of food during harvest season for social purposes such as religion, marriage, and cultural festivals. All these activities decrease food security of households that mainly harvest from their own farm. More over, the income contribution from off farm is low that is only 15-20 % (Suek, 2010), thus the accessibility of food becomes low. This condition certainly affects food security of household and then change consumption pattern of the household.

Considering two extreme food availability cycles, i.e. at harvest and lean seasons, this study was designed to stabilize food security in these two seasons as illustrated in Figure 1.

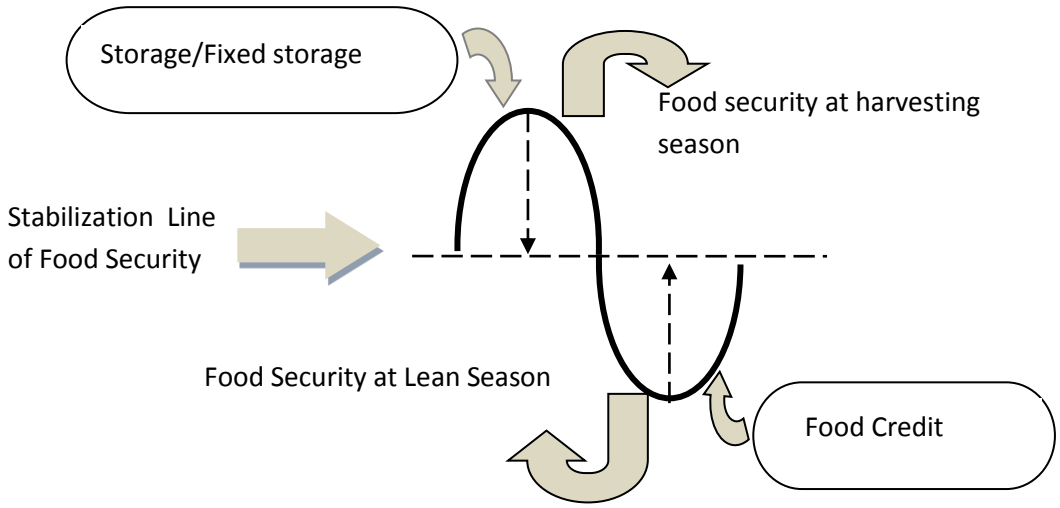

Figure 1. Illustration of food security stabilization achievement at harvesting and lean seasons

Illustration in Figure 1. explaining that intervention conducted during the lean season aims to ensure the minimal food availability in household. Some possibilities of intervention are giving food credit, storing extra food voluntary (pure storage) and storing extra food with contract.

Food credit, pure storage and storage with contract treatments are supposed to form a bridge between these two extreme seasons. During harvesting season, households are expected to use food wisely. At lean season, therefore, the households still have enough food. The study will find out how far the treatments give benefit to the households. Therefore, the objective of the research is to investigate food availability in household during harvesting and lean season, and also to analyze the variance factor influencing food security in households during harvesting and lean seasons.

Materials and Methods

The research was conducted at harvesting and lean season in 2009. The research covered 4 regencies in West Timor, i.e. Kupang Regency, Timor Tengah Selatan Regency, Timor Tengah Utara Regency, and Belu Regency. Twenty four villages were purposively chosen

from each regency; therefore, there are 96 villages in total. The villages were selected based on the potency of agricultural production, the distance from closet sub district (10 km), and accessibility. The 24 villages from every regency were then divided into 4 treatments: 6 villages for control, 6 villages for credit program, 6 villages for pure storage program, and 6 villages for fixed storage program. Except control villages, treatment villages were facilitated by 2 NGOs: Yayasan Alfa Omega (YAO) and Tanaoba Lais Manekat (TLM) by distributing food (rice and corn) for credit villages, and storage equipments (drum, jerry can, and plastic sacks) for storage villages. The facilitators also helped the group of participants to determine when the storage will be opened. There are 100 households participants in one village.

Respondents of the study were 30 housewife (or mature women responsible for preparing food for the family) randomly chosen per village. Similar respondents were interviewed at all seasons.

Collection of primary data was conducted using questionnaire with open and close questions. Secondary data were gathered from institutions related to the study issue. Observations to the groups during their activity were also carried out in addition to depth interview of key persons such as group leaders (head, secretary, and treasurer), head of village and NGO facilitators.

Data were divided into categorical data and numerical data. Categorical data were arranged for narrative explanation, while numerical data were tabulated and analyzed using statistical means (descriptive and inferential). Descriptive analyses were executed by calculating mean, percentage, and coefficient of variation, while inferential analyses were performed using multiple regressions with general formula: $Y = f(X_i)$; whereas Y is food availability, and X_i is variable that influencing food availability in household.

Results and Discussion

West Timor is administratively divided into 1 Kota Madya and 4 Regencies i.e. Kupang City, Kupang Regency, Timor Tengah Selatan (TTS) Regency, Timor Tengah Utara (TTU) Regency and Belu Regency. Total area of West Timor is 15,117.83 km square. Kupang Regency covers the widest area (39.01%), while Kupang City occupies the smallest area (0.17 %) (BPS NTT, 2010).

About 80% of households in West Timor are depended to agriculture sector. However, agriculture contribution to PDRB NTT within last 3 years tend to decrease down to 40.11% (BPS NTT, 2010). Although in general there was a shifting from traditional subsystem agriculture toward modern agriculture, the average income of a household is still low, ranged from Rp 3.5 million to Rp 4.5 million per year (Suek 2010, 2011), Compared to the income per capita written in Statistic of NTT (2010). This condition may cause households more vulnerable to famine.

Household Profile

Data from Table 1. showed the profile of household including land possessing, number of household members within working age, and number of household receiving BLT and Raskin. Although Area of owned land of stakeholder is big (ranged from 1.66 to 2.38),

however not all of these land are compact. Land use per household is around 60-75 % of owned land per year.

Table 1. Profile of respondent households

Profil RT	Treated Villages			
	Credit	Pure storage	Contract storage	Control
Area of possessing land, ha	1.32	1.18	1.42	0.78
Number of household member, mean of people	5	5	5	5
Number of working age household member, mean of people	3	3	3	3
Household receiving BLT, %	77,27	58,86	76,05	58,59
Household receiving Raskin, %	97,78	97,77	92,78	87,22

Source: Primary data
Note: BLT:= Bantuan Langsung Tunai (direct finance support); Raskin=Beras Miskin (subsidized rice for the poor)

Data of land ownership in Table 1 shows that 2,225 households (78.56 %) own dry land; 578 households (20.07%) own dry land and wet land, and 97 household (3.37 %) own wet land.

Production, Availability and Consumption of Food

FAO (1996) proposed 4 important components in the definition of food security: (1) enough availability of food, (2) stability of food availability without any fluctuation through the years, (3) food accessibility, and (4) food security. Moreover, Cohen (2002) mentioned 6 basic components that have correlation with food security in community: (1) social economy profile and demography character of community, (2) profile of food resource in community, (3) estimation of food security in household, (4) estimation of accessibility to the food resource, (5) estimation of affordability and availability of food, and (6) estimation of food production in the community.

Data in Table 2 describes the variation of food crops production per household. Except vegetables, the smallest of food crops production was found in villages having contract storage treatment. The highest production of maize and rice, on the other hand, was found in control villages.

Table 2. The mean of food crops production in households at harvesting season, 2009

Commodity	Treated villages			
	Control	Credit	Pure Storage	Contract Storage
Maize, kilogram/HH	200.66	166.90	71.57	140.47
Rice, kilogram/HH	297.01	212.89	59.98	149.51
Cassava, kilogram/HH	194.08	247.47	19.96	170.15
Pulse, kilogram/HH	113.69	79.89	6.15	45.14
Vegetables kilogram/HH	207.87	435.71	07.62	302.97

Source: Data Primer,
Note: Raw rice has been converted into rice with 60% out turn

Food availability in household at the time of interview (October-December 2009) were presented in Table 3.

Table 3. The mean of food availability in households in West Timor during lean season, 2009

Commodity	Treated villages			
	Control	Credit	Pure Storage	Contract Storage
Maize, kilogram/HH	41.48	46.77	56.15	36.69
Rice, kilogram/HH	53.61	76.86	27.43	112.32
Cassava, kilogram/HH	103.33	96.50	92.62	54.22
Pulse, kilogram/HH	15.39	14.19	7.04	7.63
Vegetables kilogram/HH	25.16	62.75	6.66	16.14

Source: Data Primer

The highest availability of maize was found in pure storage villages, while the lowest maize availability was found in contract storage villages. The highest availability of rice was found in contract storage villages followed by credit villages, control villages and pure storage villages. High availability of rice in contract storage villages as well as in credit villages mainly was caused by intervention of the programs. The Program intervention was aimed to anticipate food shortage in the villages.

The mean of weekly food consumption during lean season decreased by 13,54 % compared to it in harvesting season. In lean season, consumption of maize was 53,93 % of total consumption of it in harvesting season, while consumption of cassava, pulse and vegetables in lean season was respectively 73,14 %; 68,54% and 56,15% of it in harvesting season. Consumption of these commodities is quantitatively presented in Table 4.

Table 4. Weakly household consumption at harvest and lean season 2009, in kilogram

Commodity	Control		Credit		Pure Storage		Contract Storage	
	Harvest	Lean	Harvest	Lean	Harvest	Lean	Harvest	Lean
Rice	10.25	8.86	10.45	9.20	10.50	9.12	11.03	9.33
Maize	6.24	3.01	5.64	2.65	5.40	2.50	7.11	4.99
Cassava	8.79	7.54	9.59	7.96	7.88	5.00	11.42	7.06
Pulse	4.04	3.11	4.35	3.50	6.26	3.90	4.55	2.64
Vegetable	8.94	5.35	8.31	5.11	11.13	6.62	9.02	3.93

Source: Data Primer, processed

Non Food Expenditure

Non food expenditure is expenses of household not for food. Data in Table 5 indicates that in all treated villages, the highest non food expenditure of households during harvesting and lean seasons was for parties. While in contract storage villages during the lean season, the highest expenditure was for education.

Table 5. Yearly non food expenditure at harvest and lean season 2009, in percent

Non Food Expenditure	Control		Credit		Pure Storage		Contract Storage	
	Harvest	Lean	Harvest	Lean	Harvest	Lean	Harvest	Lean
Education	21.94	32.45	26.41	34.98	38.30	39.16	31.25	39.88
Clothing	16.03	11.47	7.88	1.77	10.09	8.44	9.45	10.69
D-Good	14.88	10.34	10.90	9.69	12.15	10.68	14.62	14.05
Tax	7.76	1.33	2.92	5.46	1.08	1.22	2.87	1.23
F-NF Party	39.38	44.42	51.90	38.10	38.37	40.50	41.82	34.15
				100.0		100.0		100.0
TOTAL	100.00	100.0	100.00	0	100.00	0	100.00	0

Source: Data Primer, processed

Note: DGoods=Durable Goods; F-NFParty=Family-Non Family Party

Household Income

Household income is the raw income received by household from different resources. It can be seen from Table 6 that agriculture is the most important income resource for households. Income from off farm is also important for the family.

Table 6. Household income from different resources in west timor during harvesting and lean seasons, 2009

Sources of Income	Control	Credit	Pure Storage	Contract Storage
Family Farm	1,018,270.79	1,171,547.58	875,188.92	1,163,268.91
Non Family Farm	684,422.67	1,102,344.58	1,712,346.00	884,891.43
Selling Livestock	459,578.42	376,210.04	370,393.37	561,838.39
Other Family Member Income	346,699.51	283,807.58	279,419.56	423,843.00
Off farm	550,839.91	541,455.18	771,577.64	856,179.52
TOTAL	3,059,811.30	3,475,364.96	4,008,925.49	3,890,021.25

Source: Data Primer, processed

Note: Other sources=the Income that is sent by household members who worked in other cities or foreign Countries

Relation between Season and Treatment on Food Availability

Purwanto (2009) proposed that in providing food, the food security aspects that should be considered are food availability, and the calorie and protein sufficiency in the households. The higher expenditure on food, the lower food security in the household.

The result of study showed that in harvesting season, the average of weekly cash expenditure for food was Rp. 53.472,35 and 53,18% of it was spent for purchased rice. On the other hand, in lean season the sum of Rp. 36.432,57 was disbursed for food and among it 59,87% was spent for rice. Rice was the biggest proportion of weekly consumption of household compared to maize and cassava. This is an indication of the changes of food habits in West Timor, which in the past the staple food of the rural households was maize and cassava. In fact, there are only 23,44% of household have rice field (wet land) or the majority (76,56%) of households depend on dry land farming. This means that most of households tends to purchase rice rather than using maize and cassava for their meal. Purwanto, *et al.,* (2009) reported that net producer of rice is only 24% of total households

in Indonesia. If there is an increase in the price of rice, it will reduce the purchasing ability of households, and therefore increase the risk of poverty.

From the perspective of harvest, credit villages and contract storage villages showed low production of maize and rice. However, these two treated villages performed high stock and consumption of rice. This is an indication that program intervention has created the possibility for household to credit rice during the lean season. Most of household preferred to buy or credit rice rather than maize. In pure storage villages, most of house hold stored maize in the storage container distributed by the intervention program.

From the perspective of community empowering, credit villages showed more dynamic than other treatments. About 70 % of participants said that food credit has been a big help for them during the lean season. They have no to be much worried about their food. Up to 30 % of participants in contract storage villages still do not want to open their food storage containers even though it has passed the contract time for the storage. They said that they will open the containers if they do not have anything else to eat.

Factors Affecting Food Availability

Food availability in household generally depends on cereal, tubers, pulse, vegetables and fruit both come from family farm or non family farm. Many indicators have been used to perform food security in household. The availability and consumption of food mainly are presented referring to the need of basic food that provides carbohydrate. Therefore, to analyses of food sufficiency, food from agriculture produce is usually converted according to Leung, Butrum and Chang, (1972). Then, by using *Ordinary Least Square* (OLS) method resulted from multi linear regression with the formula below, the coefficient of regression will be defined for each variables mentioned in Table 7.

$$\text{Log } Y = \log A + b_1 \log X_1 + b_2 \log X_2 + b_3 \log X_3 + b_4 \log X_4 + b_5 \log X_5 + U$$

Note: Y = Food availability (converted into kcal), according to Leung, Butrum and Chang, (1972); X_1 = Harvest in 2009 (converted into kcal); X_2 = Farm area (ha); X_3 = Household cash income (Rupiah/year); X_4 = number of productive age labor in the household (people); X_5 = expenditure for parties (Rp/year); U = Error/residual

Table 7. The Data of Factors Affecting Food Availability in west Timor

Variables	Coefisien	Std Error	t-Statistic	Probability
Intercept	2.352373566	0.145207368	16.2000978	1.67843E-50
X-1	0.372469394	0.016931948	21.99802418	2.77528E-82
X-2	0.681366502	0.185238554	3.67831904	0.000252422
X-3	0.089521118	0.012955075	6.910119555	1.07395E-11
X-4	0.319258914	0.177469888	1.798946956	0.07244924
X-5	-0.062227354	0.011978945	-5.194727335	2.67766E-07
R-Squared	0.523202608	F -Statistic		156.6982825
Adjusted R-Squared	0.519863691	Prob (F-statistic)		0.000000

Source; data primer processed using Microsoft Excel 2003

Harvest of agriculture commodity, area of possessing farm, household cash income, and the number of productive age labor in the household have positive effect on food availability in household. The larger of possessed land, owned cash and labor availability the more flexible for the farmers or household to use all these production factors to choose the best way to increase the productivity of farm. In contrary, Expenditure for party negatively affects the

food availability of household. The more spending on party the more risk of food shortage and famine for the family.

Conclusions

1. Most of food availability in West Timor rural households is much depended on the harvest from family farm that has short durability; therefore, intervention programs by crediting food, applying pure storage or contract storage create good lesson for household in coping food shortage during lean season.

2. The result from analysis showed that the harvest of agriculture commodity, area of possessing farm, household cash income, and the number of productive age labor in the household have positive effect on food availability in household. The higher productivity of farm it was meant the higher availability of food in the household. The more spending on party the more risk of food shortage and famine for the family.

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The Characterization and Evaluation of Local Upland Rice Cultivars to Blast Disease (*Pyricularia oryzae*) Resistance in Southeast Sulawesi

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Abstract

Blast disease, caused by Pyricularia oryzae is an important disease for upland rice. Blast disease causes a significant loss for farmers. One of the control methods is using resistant cultivar. The objective was to characterise and identify differences in the response of several Southeast Sulawesi upland rice cultivars to blast disease (Pyrricularia oryzae) in Southeast Sulawesi. This research was arranged in a randomized block design, with 6 local rice cultivars (pae Endokadia, pae Kori, pae Balaka, pae Enggalaru, pae Loiyo, pae Wulo), and one national variety Situpatenggang, with 4 (four) replications.The results showed that the highest plant height was pae Bakala cultivar, the highest number of plant per cluster was Situpatenggang variety, the fastest flowering and harvesting time was Situpatenggang variety, the highest average number of grains was found in pae Wulo, the highest number of pithy grains was obtained in pae Enggalaru, and the heaviest of 1000 grain was in pae Endokadia. Enggalaru and Endokadia cultivars had highest production (1.73 ton per hectare and 1.24 ton per hectare), compared to the others. The severity of blast disease on the local rice varieties was catagorized as moderate resistant (5.78% - 11.05%) at vegetative phase and as fairly resistant to moderately susceptible (14.47% - 27.38%) at generative phase. Situpatenggang variety was considered to be moderate susceptible (28.79%) at vegetative phase and susceptible (55.79%) at generative phase.

Keyword: blast disease, Pyricularia oryzae, resistant, up land rice

Introduction

Southeast Sulawesi has a high potency for dry-land areas. According to Central Beaureu of Statistics of Southeast Sulawesi (2009), the dry-land area in this province reaches 3,147,000 hectares. The dry-land area spreads out evenly in several districts, which is used to cultivate local and national up-land rice varieties. In general, up-land rice (*Oryza sativa* L.) cultivation is conducted at dry-land area, which water source is ony from rainfall. Because the water source is only from rainfall, there is no need to invest irrigation like for paddy rice. Sumarno and Hidayat (2007) pointed out several advantages of up-land rice cultivation, such as: 1) able to utilize soil nutrients efficiently and tolerant to low pH, making initial investment for soil amelioration can be minimized, 2) relatively low production costs and labor, 3) there are no need to build specific infrastructures such as irrigation, dams etc, that overall makes the investation is relatively low.

Not only Southeast Sulawesi has a high potency for dry-land areas but also it has a high number of promising local up-land rice germplasms that have traditionally been bred and cultivated by local farmers. Collection and evaluation of local varieties showed that several

varieties had diverse morphological characteristics and is also presumed has diverse resisitancy to plant diseases.

One of the important up-land rice diseases is the fungus *Pyricularia oryzae,* causing the blast disease for rice. This disease contributes significantly to the reduced production of up-land rice, experienced by farmers. Baker *et al.,* (1997) reported that the blast disease caused yield losses between 30% to 50% in Southeast Asia and South America. Research by Amir *et al.,* (1993) showed that the infection of pathogen *P. oryzae Cav.* on susceptible cultivars caused yield loss up to 50-90% or even failed to harvest. Therefore, it is necessary to conduct research on the characterization and evaluation of local upland rice cultivars to blast disease (Pyricularia oryzae) resistance in Southeast Sulawesi

Materials and Methods

Time and Place

The researh was conducted in farmer's field at Puriala, Konawe, Southeast Sulawesi, between November 2009 to November 2010. This area was an endemic area for blast disease.

Materials and Equipments

Materials used were six local up-land rice varieties (Pae Endokadia, Pae Kori, Pae Bakala, Pae Enggalaru, Pae Wulo, dan Pae Loiyo), and as the control was a national up-land rice variety. Other materials used were urea, SP36, and KCl fertilizers; glyphosat herbicide; deltametrin 25 g/l and Sipermetrin insecticide. Equipments used were hand tractor, knapsack sprayer, wiring net, scale, camera, etc.

Research Design

This research used Randomized Block Design (RBD) with seven treatment (varieties) (Pae Endokadia, Pae Kori, Pae Bakala, Pae Enggalaru, Pae Loiyo, Pae Wulo and Situ Patenggang). The treatments were repeated 4 times. Observed variables were plant height, number of cluster, flowering time,tinggi tanaman, jumlah anakan, umur berbunga, number of grain, and weight of 1000 grains. Data were analized ad means were separated using Duncan Multiple Range Test (DMRT) (Steel and Torrie, 1993).

Research Procedure

Land Preparation, Planting, and Plant Maintenance

Land was cleared of weeds, and followed by making plots according to treatments. Plot size for each treatment was 4 m x 5 m. Planting was conducted by making hole for seed, with planting distance of 80 cmx 40 cm. Each hole was planted wit 3-5 seeds. Fertilizers used were 90 kg N, 75 kg P_2O_5 and 75 kg K_2O. Half dossage of N, all dossage of P dan K were applied at planting, and half dossage of N was applied at 30 days after planting. Fertilizers were placed approximately 10 cm on the right and left of plants. Replacement of died plants was done at 1 week after planting.

Harvesting

Harvesting was conducted when 80-90% rice grains had shown signs of physiological mature, where rice grains had colored goldy yellow (for yellow cultivars) and black (for black cultivars), and tips of leaves had colored brown and drying out.

Observation and Data Collection

Observation was performed to determine the blast disease (*P. oryzae*) severity, at 30 days after planting. Determination of the infecton value used Standard Evaluasi System for Rice IRRI (1988) and Sijabat (2007), which is shown on Table 1. Observation data were tabulated and analised using analysis of varians (anova). Separating significant means used Duncan Multiple Range Test (DMRT) at 95%, using SAS program.

Table 1. Scoring for resistance to blast disease (*P. oryzae*)

Score	Category (symptoms)
0	Highly resistant (no infection symptoms)
1	Resistant (blast symptoms, with infected leaves 1 – 5%)
3	Moderately resistant (blast symptoms, with infected leaves >5 – 11%)
5	Fairly resistant (blast symptoms, with infected leaves >11 – ≤25%)
7	Moderately susceptible (blast symptoms, with infected leaves >25 – ≤50%)
9	Susceptible (blast symptoms, with infected leaves >50 – 100%)

$$\text{Formula : } Kp = \sum ((vi \times ni)/Z \times N) \times 100\%$$

Notes : Kp: Disease severity
vi: Score at infection category
ni: Number of leaves at infection category
Z: Score at the highest infection category
N: Observed leaf number

Results and Discussion

Plant Height (cm)

Averages of plant height at 1 to 11 weeks after planting (WAP) were shown on Figure 1. Based on anova of average plant height, it was found that cultivar treatments gave significant effect on plant height at 1 to 11 WAP. The highest average plant height at 1 to 9 WAP was on pae Endokadia, while at 10 and 11 WAP was on pae Bakala (126.40 cm). The lowest average plant height (104.10 cm) at 1 to 11 WAP was on variety Situpatenggang. The difference in plant height among cultivars was caused by genetic factor. This is in line with Mildaerizanti (2008) stating that diverse plant height was influenced by a genetic factor.

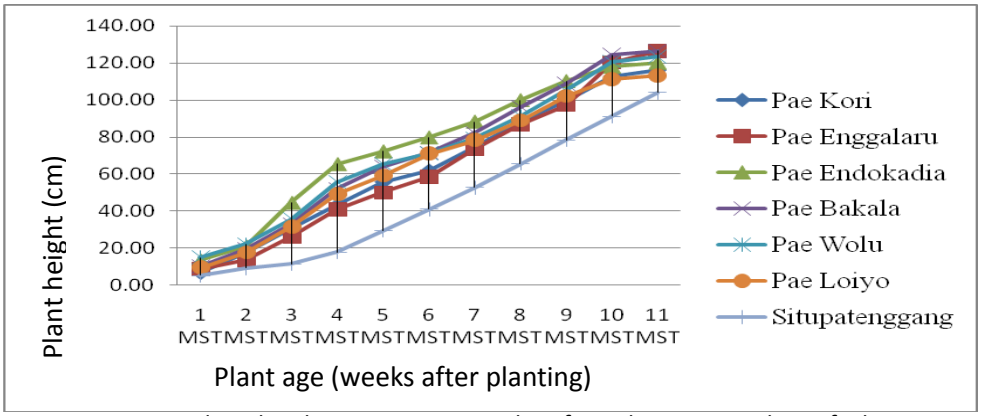

Figure 1. Average plant height at 1 to 11 weeks after planting Number of Plants per Cluster, Number of Productive Plants per Cluster and Precentage of Productive Plants per Custer

Research showed that Situpatenggang variety had the highest number of plants per cluster (43.65 plants), which was significantly different from other cultivars. Pae Wulo had the smallest number of plants per cluster (10.85 plants), which was different from treatments pae Enggalaru and Situpatenggang variety. Situpatenggang variety had the highest number of productive plants (27.45 plants), which was different from others varieties, while pae Wulo had the smallest number of productive plants (9.15 plants). Pae Loiyo had the highest number of average percentage of productive plants (97.28 plants), while variety Situpatenggang had the least (27.70 plants) (Table 2). Based on this reseach, Situpatenggang variety was the most severe of disease infection (28.79 – 55.79%). This was because the plant cluster was so dense, creating the micro climate was more suitable for pathogen growth and infection, and made plants more susceptible to the disease. Prayudi (2008) also observed that the more dense the rice population, the more severe disease incidence.

Table 2. The average number of plants per cluster, number of productive plants and precentage of productive plants on up-land rice cultivars

Treatment (Cultivars)	Number of plants per cluster (plants)	Number of productive plants (plants)	Precentage of productive plants (%)
Pae Kori	11.75bc	10.65bc	91.47ab
Pae Enggalaru	15.90b	14.25b	89.26ab
Pae Endokadia	12.55bc	10.00c	80.64b
Pae Bakala	13.70bc	11.05bc	80.80b
Pae Wulo	10.85c	9.15c	84.21b
Pae Loiyo	13.05bc	12.70bc	97.28a
Situpatenggang	43.65a	27.45a	27.70c

Note: Means in each olumn followed by a common letter are not different at the 5% probability level according to the DMRT

Flowering and Maturing Time

Situpatenggang variety had the fastest flowering (106.80 days) and maturing (127.20 days) time that were different than other cultivars, while pae Wulo had longest maturing time (157.60 days), which was different than other cultivars (Table 3). In tropical region, the optimum maturing time for up-land rice to reach maximum yield is generally around 120 days. The shorter cultivation time will generally contribute to the lower yield due to an inadequate time to utilize sun light and soil nutrients (Yoshida and Parao, 1976). Maturing time between 100 - 130 days is generally sufficient for best yield (Abdullah, 2004). In this research, pae Wulo cultivar had longer maturing time (157.60 days) than other cultivars, and gave the least weight of 1000 grains (23.10 g) and the highest number of empty-grains (77.33%).

Table 3. Average flowering and maturing time of up-land rice cultivars

Treatment (Cultivars)	Flowering Time (days)	MaturingTime (days)
Pae Kori	128.40a	149.60d
Pae Enggalaru	126.40bc	155.80b
Pae Endokadia	126.40bc	148.00e
Pae Bakala	125.20c	149.60d
Pae Wulo	127.40ab	157.60a
Pae Loiyo	125.80c	154.55c
Situpatenggang	106.80d	127.20f

Note: Means in each olumn followed by a common letter are not different at the 5% probability level according to the DMRT

Number of Grain, Number of Pithy Grain, Number of Empty Grain, and Precentage of Empty Grain

Pae Wulo had the highest average number of grain (229.30 grains), while Situpatenggang variety had the lowest (104.70 grains). Pae Enggalaru had the highest number of pithy grain (122.85 grain), while Situpatenggang variety had the lowest (37.25 grains). Pae Wulo had the highest average number of empty grain and percentage of empty grain (177.30 grains), while pae Endokadia had the lowest number of empty grain (59.45 grains) and pae Enggalaru had the lowest percentage of empty grain (34.20 %) (Table 4).

Weight of 1000 Grains

Pae Endokadia had the highest average weight of 1000 grains (30.89 g), while pae Wulo had the least weight (23.10 g), which was different than other cultivars (Table 5). Cultivars Enggalaru and Endokadia had a genetic potency to be further developed, because they had higher production (1.73 ton ha^{-1} and 1.24 ton ha^{-1}) than other cultivars. Cultivar Endokadia had an erect leaf habit, stronger stem, the highest weight of 1000 grains (30.89 g) and had a relatively low percentage of empty grain (43.38%). This finding is in line with Siregar (1981) who stated that a high rice production was generally correlated with an erect leaf habit (angle between stem and leaf > 30^{o}) and had compacted and strong stem.

Table 4. Average number of grain, number of pithy grain, number of empty grain, and precentage of empty grain of up-land rice cultivars

Treatment (Cultivars)	Number of Grain(grain)	Number of Pithy Grain (grain)	Number of Empty Grain (grain)	Precentage of Empty Grain (%)
Pae Kori	211.75c	95.85b	115.90c	54.73d
Pae Enggalaru	186.55d	122.85a	63.70f	34.20f
Pae Endokadia	137.05f	77.60c	59.45g	43.38e
Pae Bakala	156.65e	71.75d	84.90d	54.20d
Pae Wulo	229.30a	52.00e	177.30a	77.33a
Pae Loiyo	222.85b	74.15cd	148.70b	66.74b
Situpatenggang	104.70g	37.25f	67.45e	64.42c

Note: Means in each olumn followed by a common letter are not different at the 5% probability level according to the DMRT

Table 5. Average weight of 1000 grains of up-land rice cultivars

Treatment (Cultivars)	Weight of 1000 Grains (g)
Pae Kori	23.76e
Pae Enggalaru	27.73b
Pae Endokadia	30.89a
Pae Bakala	30.88a
Pae Wulo	23.10f
Pae Loiyo	25.63d
Situpatenggang	26.76c

Note: Means in each olumn followed by a common letter are not different at the 5% probability level according to the DMRT

Severity of Blast Disease *(P. oryzae)* at Vegetative and Generative Phase

The least severe of blas disease at vegetative and generative phases was on Pae Bakala, while the most severe was on Situpatenggang variety. Grouping of resistance levels based on IRRI (1988) and Sijabat (2007) showed that there were 5 (five) cultivars categorised as moderate resistant, 1 (one) cultivar as fairly resistant, and 1 (one) cultivar as moderately susceptible. Generative phase was generally categorised as fairly resistant (Table 6).

It can be assumed that in general all local rice cultivars were categorized as moderate resistant that presumably had some kind of a defense mechanism against blast pathogen infection. According to Agrios (2005) and Goodman (1986), plant resistance response can occur structurally and biochemically. Cultivars Enggalaru and Endokadia had a genetic potency to be further developed, because they had higher production (1.73 ton ha^{-1} and 1.24 ton ha^{-1}) than other cultivars, and moderately resistant to blast disease.

Table 6. Grouping of resistance category to blast disease (*P. oryzae*) at vegetative and generative phases on up-land rice cultivars

Treatment (Cultivars)	Disease severity at Vegetative phase (%)	Resistance Category/Type	Disease severity at Generative phase (%)	Resistance Category/Type
Pae Kori	8.53	Moderate resistant	22.02	Fairly resistant
Pae Enggalaru	6.55	Moderate resistant	16.74	Fairly resistant
Pae Endokadia	8.08	Moderate resistant	20.49	Fairly resistant
Pae Bakala	5.78	Moderate resistant	14.47	Fairly resistant
Pae Wolu	8.59	Moderate resistant	24.34	Fairly resistant
Pae Loiyo	11.05	Fairly resistant	27.38	Moderately susceptible
Situpatenggang	28.79	Moderately susceptible	55.79	Susceptible

Conclusions

Based on research results and discussion, it can be concluded that:

1. The highest average plant height was pae Bakala; the highest number of plant per cluster was variety Situ Patenggang; the fastest flowering and maturing time were Situ Patenggang variety; the highest number of grain was pae Wulo; the highest number of pithy grain was pae Enggalaru; and the heaviest of 1000 grain was pae Endokadia.
2. Cultivars Enggalaru and Endokadia had highest rice production (1.73 ton ha^{-1} and 1.24 ton ha^{-1}).
3. Severity to blast disease (*P. oryzae*) of local rice cultivars at vegetative phase was categorized as moderate resistant (5.78% - 11.05%), and at generative phase was categorised as fairly resistant to moderately susceptible (14.47% - 27.38%).
4. Severity to blast disease (*P. oryzae*) of Situpatenggang variety at vegetative and generative phases was categorized as moderately susceptible (28.79%) and susceptible (55.79%).

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Effects of Nitrification Inhibitors on Denitrification in Soils

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Abstract

Nitrous oxide is a biogenic greenhouse gas emitted to the atmosphere from soils and it is stimulated by agricultural management practices involves the use of nitrification inhibitors. Nitrous oxide (N_2O) is produced by biological processes in the soil such as denitrification and nitrification . This experiments was conducted under laboratory conditions to elucidate the effect of three nitrification inhibitors viz, 3,4 dimethylpyrazolephosphate (DMPP), 4-Chlormethylpyrazolephosphate (ClMPP) and dicyandiamide (DCD) on denitrification in soil incubated at 25^oC. The quantitative determination of potential denitrification capacity (PDC) was measured by gas chromatographically. DMPP, ClMPP and DCD were recommended used on rates of 90 kg N ha^{-1} corresponding to 0, 36 µg DMPP; 0, 25 µg ClMPP and 10 µg DCD g^{-1} dry soil. In all treatments, the influence of 1, 10, 50, 100, 250 and 500 times of the recommended-concentrations were examined. The results suggested that DMPP, ClMPP and DCD applied at rates generally recommended for agricultural use may not be effective to inhibit denitrification. The three NIs began to inhibit PDC at 10 to 50 times recommended concentration and were more effective in sandy than in loamy or clay soils. ClMPP influenced PDC at much lower concentration as DMPP or DCD.

Keywords: nitrification inhibitors, potential denitirification capacity

Introduction

Nitrification of applied nitrogen fertilizers leads to losses of nitrogen (N) as nitrate (NO_3^-) or as the greenhouse gas nitrous oxide (N_2O). These losses mean that plant use efficiency of nitrogen fertilizers is low. Reducing nitrification losses can be achieved with the use of nitrification inhibitors that suppress the activity of the autotrophic ammonia oxidizing bacteria (AOB). The application of these compounds that retard nitrification was used to improve N recovery and N use efficiency in agricultural soils, while the same time limiting the environmental impacts of N loss and thus improving sustainability (Fillery, 2007). It have beneficial effect on reducing nitrate leaching into ground water (Di et, al. 2004; 2005) and nitrous oxide emission to the atmosphere or denitrification (Weiske *et al*. 2001; Di et, al, 2006; 2007), affect N retention in the root zone and microbial biomass and activity in the rhizosphere and as a result increase plant growth (Wolt, 2004; Douma et.al, 2005; Malla *et al*, 2005, Moir, et.al, 2007). The use of nitrification inhibitors (NIs) show a reduction of about 60% in NO_3-leaching, 70% in N_2O emissions and an increase of more than 20% in crop and pasture yield can be achieved (Sahrawat, 2004; Douma *et al* 2005; Di. *et al*, 2007; Moir *et al*, 2007; Singh and Verma, 2007). Nitrification results in the formation of highly mobile nitrate, which is susceptible to loss from root zone by leaching and/or gaseous

emissions of di-nitrogen or nitrous oxide through denitrification. As the loss of soil N in solution or gaseous form can cause pollution as well as N deficiencies in crops and pastures, the prospect of actively regulating these soil processes has major implication for improving effiency of fertilizer nitrogen in agriculture and for plant productivity (Chen *et al*, 2008; Li *et al*, 2008: Stark and Richards, 2008). The reduced nitrification can have significant impacts on the soil carbon cycle and, for example, decrease organic decomposition. N species (i.e. ammonium vs. nitrate) may a more important driver of carbon cycling and ecosystem functioning than the quantity of N present in the system (Austin *et al*, 2006).

The using of Nitrification inhibitors in agriculture should be recommended in low concentration and capable to control nitrate supply to crop so that avoid the excess of nitrate supply in soils. The inhibitor has the specific influence that is only inhibit the nitritation and not for nitratation so that accumulation can be avoid. The Inhibitor should be bacteriostatic and not a bactericide which killing certain microorganism in soils like *Nitrosobacter* sp, *Nitrosococcus* sp. Recently, more than 300 type of nitrification inhibitors have been well recognized and used in agriculture. Some of these NIs consisted of N-heterocyclic compounds, acetylene derivates, sulphates and also various pesticides and herbicides (Regina *et al*, 1998; Mc Carty, 1999). Ammonium-recommendedd fertilizers are the most widely used source of N for crop production and keeping the applied fertilizer N in NH_4^+ form by using nitrification inhibitors (NIs) is a well documented strategy for reducing N loss and to minimize negative environmental impacts of the fertilizer-use. Three compounds have been commercialized as NIs for agricultural use including (i) nitrapyrin (2-chloro-6-trichloromethyl-pyridin, trade name N-Serve), (ii) dicyandiamide or DCD (trade name Didin, Alzon or and Ensan), and (iii) more recently DMPP (a pyrazole derivative, 3,4-dimethylpyrazole phosphate; trade name ENTEC) (Zerulla *et al*., 2001; Weiske *et al*., 2001; Barth *et al* 2006; 2008; Ali *et al*, 2008).

The objective of the present study was to evaluate under laboratory conditions the effect of three nitrification inhibitors viz., 3,4-dimethylpyrazole-phosphate (DMPP) 4-Chlormethylpyrazole phosphate (ClMPP) and dicyandiamide (DCD) on denitrification in three different type of soils incubated at 25^{o}C. Using this information, patterns and recommendations for the use of fertilizers containing nitrification inhibitors can be identified.

Materials and Methods

Soil samples and samples preparation

The soils samples in this study were clay, loam and sandy soils collected from Soil Experiment Station, Institute for Agronomy and Plant Protection in Giessen Germany and a sandy soil from Agrochemical Experimental Station, Bayerische Acetylen Soda Fabrik (BASF, Limburgerhof, Germany). The respective soils were classified as Typic Udorthent (clay soil) Typic Kandiudult (clay and loamy soil), and Typic Paleudult (sandy soil) according to Soil Taxonomy (Soil Survey Staff, 1999). These three experiment soils covered different types which are common in the field and varied in their chemo physical properties such as texture, pH, C-content, N (Table 1).

Nitrification inhibitors used in this study included 3,4dimethylpyrazole phosphate (DMPP, as pure 99,9 % active ingredient), 4-Chlor-methylpyrazole phosphate (ClMPP 99,7 %) both were produced by BASF AG, Ludwigshafen Germany) and dicyandiamide (DCD= Purity 96%

produced by SKW Trotsberg Ag. Trotsberg Germany). In experiments utilizing these three NIs, stock solution of the inhibitor was prepared in distilled water by mixing the inhibitor in soil, whereas for experiment with NIs as pure active ingredient as well as the control. Portions of this stock solution were used to achieve the desired level in soil. These three NIs were applied at recommendation rates 0, 36 µg, 0,25 µg ClMPP and 10µg DCD g^{-1} dry soil. These rates were equal to that incorporated in N-fertilizer for 90 kg N per Ha. The application rates used in the present study included 1, 5, 10, 25, 50, 100, 250, and 500 times of recommended concentrations.

Table 1. Chemical and physical properties of soils

Parameters	**Type of Soil**		
	Loamy clay	**Loam**	**Loamy sand**
C_{total} (%)	1,35	1,30	0,70
C_{H2O} (%)	0,40	0,55	0,27
N_{total} (%)	0,15	0,15	0,08
C/N	10	9	9
pH_{H2O}	6,30	7,00	7,00
pH_{KCl}	6,00	5,50	6,40
CEC (Ba in cmol/c/kg)	20,1	12,0	8,4
mWHC (%)	45,5	40	26,5
NH_4^+-N (µg g^{-1} dwt.s)	1,0	4,3	1,0
NO_3^--N (µg g^{-1} dwt.s)	10,6	7,9	17,4
NO_2^--N (µg g^{-1} dwt.s)	0,06	0,1	0,1
Fraction (%)			
Clay	51	24	6
Loam	41	46	19
Sand	8	30	75

Incubation trials

Incubations were carried out in Schott glass (capacity 250 ml) using 100 gram soil. The soil was moistened to 60% WHC with a solution containing $(NH_4)_2 SO_4$ to provide NH_4^+-N at 100 mg kg^{-1}. The bottle were covered with Parafilm that was perforated for gaseous exchange. Sufficient bottle were kept for each treatment to sacrifice four replicate bottles at each sampling interval for analysis of soil mineral N. To determine the NI-Inhibition on nitrification (ammonium decrease, nitrite and nitrate formation), the soil samples were treated with desired concentrations of DMPP, ClMPP or DCD according to Beck (1983) and incubated at 25^oC. Each sample (100 g moist soil) was placed in a 250 mL- flask of volume (for every 5 incubation times and 4 parallels), thoroughly mixed with fixed concentrations of each NI and added with 10 ml of a 1%-ammonium-sulfate solution. One extra soil sample from each treatment was used as a blank subsequently, the samples were kept into 60% of the $_M$WHC and the flask covered with perforated Parafilm (each 10 holes) (to ensure aerobic conditions and to reduce water evaporation) and in 0-7-14-28 days at 25 ° C incubated. As blank values (without NI each with or without ammonium addition) served three flasks, which were immediately deep frozen at -18 ° C incubated as control. One sample (20 g moist soil) was taken from each soil core and analyzed for inorganic N. The samples were mixed with 75 mL of 2 *M* KCl, shaken for 20 min, centrifuged, and the supernatants were stored at -20°C until analysis. At each sampling time, gravimetric moisture was determined after drying approximately 50 g of soil at 105°C for 48 h. Twenty 20 g of sample were used every incubation time and taken with a spatula and the concentrations of ammonium-N, nitrite-N and nitrate-N examined photo metrically

according to the below mentioned methods. Forty g of soil samples were weighed in 500 ml plastic bottles and added 200 ml of a mixture of 1 N NaCl and 0.1 N $CaCl_2$. Then the flasks shake for 30 minutes at 130 U-1 (VKS-shaker, Bühler, Tübingen). The filtrate of ammonium, nitrite and nitrate (filter, 595 ½ Schleicher &Schuell, Einbeck) were determined photo metrically (by U-3200 Hitachi, Japan). Ammonium-N was quantified according to Dev (1983) 2ml up to 25 ml of the above Filtrate were mixed with 2 ml of a salicylate-citrate solution and filled with distilled water up to 25 ml of volume, then it determined spectrophotometrically (by U-3200 Hitachi, Japan) at 655 nm. The ammonium concentrations were taken from a calibration curve of NH_4SO_4 and printed in µg NH_4^+-N g^{-1} dry soil. Nitrite-N was determined by using a photometer according to Dev (1981) with a-naphthylamine Sulfanilacid solution. For this purpose, 40 ml of filtrate in a flask was mixed with 2 ml of a-naphthylamine and Sulfanilacid solution. Nitrite reacts with sulfanilic acid in acidic solution of a red diazonium salt, the absorbance of the red color complex was measured at 535 nm spectrophotometrically. The nitrite concentration was taken from a calibration curve with $NaNO_2$ solution and expressed in µg NO_2-N g^{-1} dry soil.

The concentration of nitrate (NO_3-N) in the above Filtrate was determined by self-absorption directly (Navone, 1964). Five to 25 ml of the filtrate were filled up with distilled H_2O up to 25 ml of volume and mixed well homogenized with 1 ml H_2SO_4 (10%). The self-absorption of nitrate after 1 h was determined photo metrically at 210 nm. After reduction of nitrate by copper-plated zinc granule (about 3 days) the same sample again was measured and made a correction to the first reading to the absorption of the filtrate. The nitrate concentration was calculated net of a blank (salt solution) from a $NaNO_3$-calibration series and expressed in µg NO_3-N g^{-1} dry soil.

pH .Ten grams of air-dried soil in 50 ml mixed with 25 ml distilled water or a 0.01 M CaCl2 solution, and shaken for 1h (VKS shaker, Bühler, Tübingen). After equilibration (30 min) the pH was measured by potentiometric (Microprocessor pH-Meter 535, Multi-Cal, Weilheim).

Potential denitrification capacity

Denitrification as measured via acetylene-inhibition-method. This method can be used to determine either actual denitrification under field conditions or potential denitrification under optimized laboratory conditions (anaerobiosis, addition of substrates, optimum temperatures). Acetylene is used to block the conversion from nitrous oxide to dinitrogen, which means that all the denitrified nitrogen can be measured as nitrous oxide by gas chromatography. Nitrat can in O2 deficiency condition as an alternative to electron acceptor in term of a denitrification (N_2O, N_2 emission) by many soil bacteria and measured therefore as a potential denitrification capacity (PDC). To determine the potential denitrification (PDC,Pell *et al* 1998), 40 grams of the above prepared and NIs-treated test soil were weighed into gas-tight-sealable- paraffin 2000 mL glass bottles (Schott, Germany) The experimental flaks were fitted for gas sampling with screw caps, including Gummisepten (Verneret, France). Each sample was mixed with 20 ml of a KNO_3-glucose solution (50 g NO_3-N and 300 µg glucose-C g^{-1} dry soils). After gas-tight closure, the bottle purified and rinsed in 2 minutes with N_2 gas (Messer-Griesheim, Darmstadt) and 2% volume of the bottles-atmosphere replaced with acetylene (99.9%, Messer-Griesheim, Darmstadt, Germany). Acetylene blocks the N_2O reductase specifically and can be considered that the release of N_2O in the presence of C_2H_2 as a measure of the potential denitrification capacity (PDC). Later, the samples were incubated in the dark for 24 h and 48 h at 25 ° C. By using gas-tight Plastipak syringes (Becton Dickinson, Ireland), 50 ml of gas was injected into a gas

chromatograph with an electron capture detector (ECD). With an external standard, the PDC printed in µg N_2O-N g^{-1} dry soil h^{-1}. At the end of the experiment, the concentrations of NH_4+-N, NO_2-N NO_3-N in the soil samples were measure photometrically according to the above Methods.

Statistical analysis

Data were subjected to an analysis of variance followed by Duncan's multiple range tests (Gomez & Gomez, 1984). Results are reported as means of four replicates and are expressed on the soil dry weight basis.

Results and Discussions

The effect of increasing nitrification inhibitors on ammonium oxidation, nitrite and nitrate formation in clay soil

In Figure 1 is shown the effect of increasing NI-concentrations (DMPP, ClMPP and DCD) on the ammonium oxidation, nitrite or nitrate formation and on the pH gradient in the clayey soil. The ammonium oxidation is inhibited with increasing DMPP-concentrations, apparently at the base concentration continued with a rapid decrease of ammonium. A complete inhibition of ammonium oxidation in the clay soil reached within 1-2 weeks earlier at about 500 times the base concentration (0.36 µg DMPP g^{-1} dry soils, Figure. 1). The nitrite formation 250 to 500-fold increase in the DMPP concentration-based largely during the formation of nitrate is suppressed (= Nitratation) at these concentrations is not completely inhibited. After 28 days (end of the experiment) is about 30 -50% more nitrate was formed in the control (with ammonium fertilization; Fig. 1). The pH values on DMPP concentration in all areas in incubation times (especially after three weeks) were significantly dropped as a result of nitrification.

The influence of increasing ClMPP concentrations on the ammonium oxidation and of nitrite or nitrate formation, on the pH gradient in clay soil are shown in Figure 2. A complete inhibition of ammonia oxidation by ClMPP in over a period of 1 to 3 weeks was observed only at a 500-fold of base concentration. Compared to DMPP, ClMPP thus seems to inhibit the oxidation of ammonium was stronger. The pH values in the clay soil decreased less than that of DMPP. The initial pH values decreased as a result of nitrate formation after 4 weeks by about 0,2 of pH units.

Compared to DMPP and ClMPP, DCD resulted inhibition of ammonium oxidation in the first two weeks at the 50-fold of DCD-based concentration (equals to 500 µg DCD g^{-1} dry soil). Cause may be with the microbial hydrolysis of DCD (contains ~ 66% N) and release of ammonium in to the soil. The initial increase in ammonium formation (maximum after about seven days) is reflected in an increased nitrate formation again (especially in increase of the 250 - and 500-fold of base concentration). The nitritation was in the two controls (with and without ammonium addition) the highest (Fig. 1). The results showed that DCD in the high concentration range (for example in the 50 to 500 times the base concentration) rather than N-source for the heterotrophic microorganisms and less acts as a nitrification inhibitor for the nitrifiers. The additional NH_4^+ formation from the DCD-hydrolysis is also evident from the pH level.

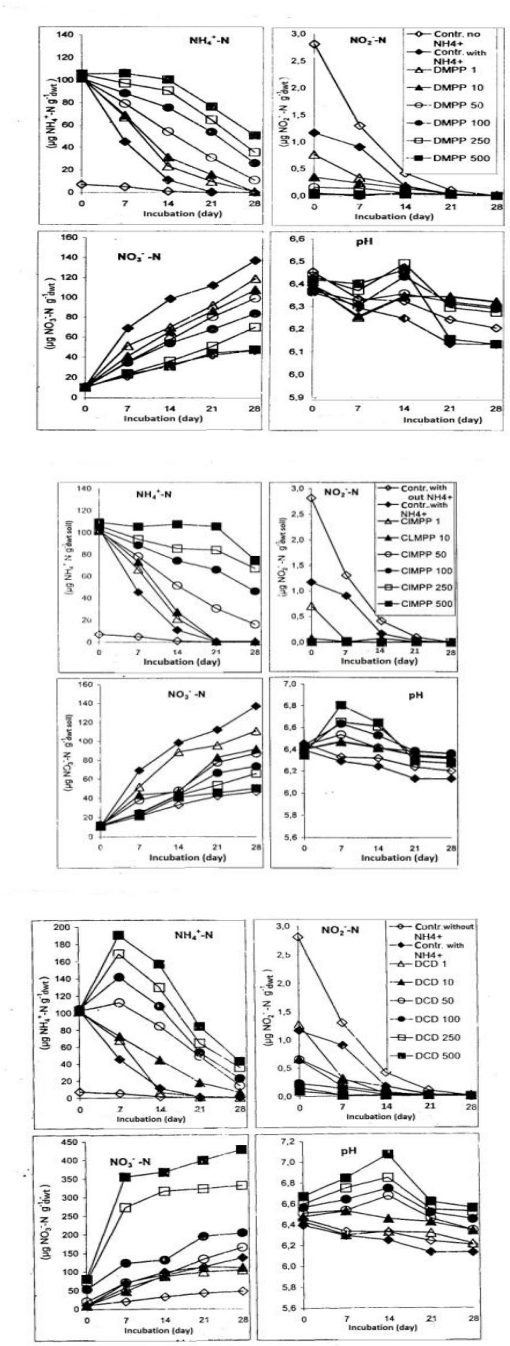

Figure 1: Effect of increasing DMPP, ClMPP and DCD concentrations on the ammonium oxidation, nitrite and nitrate formation and on the pH (H2O) values in clay soil during an incubation period of 28 days (60% MWC, adding 100 mg Ammonium-N g^{-1} dry soil, at 25 ° C). The recommended concentration equivalent to 0.36 µg DMPP, 0.25 µg ClMPP and 10 µg DCD g^{-1} dry soil.

Loamy soil

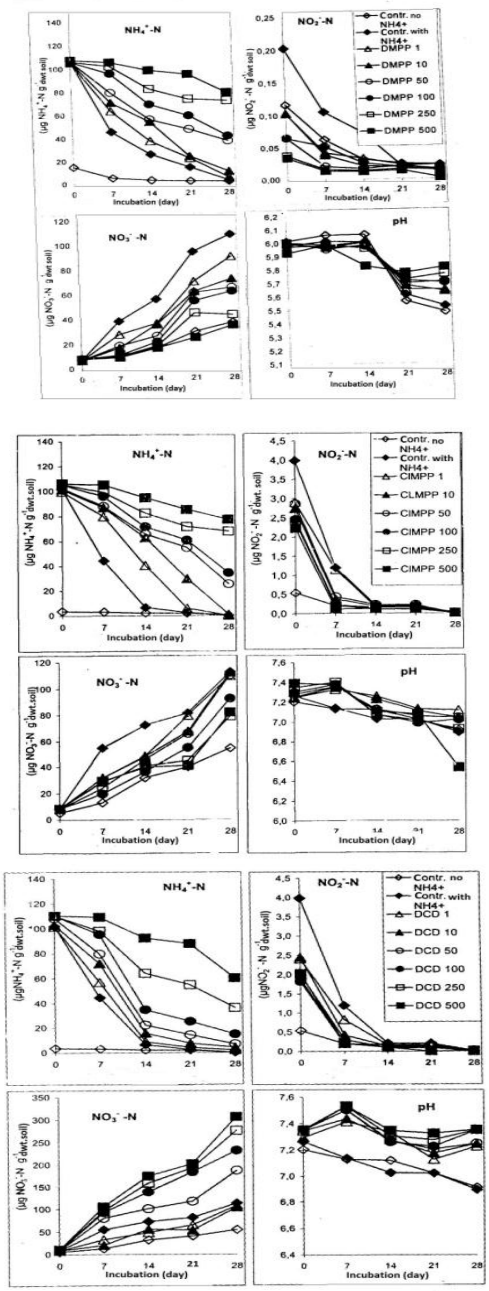

Figure 2: Effect of increasing DMPP, ClMPP and DCD concentrations on the ammonium oxidation, nitrite and nitrate formation and on the pH (H2O) values in the loamy soil during an incubation period of 28 days (60% MWC, adding 100 mg Ammonium-N g^{-1} dry soil, at 25 ° C). The recommended concentration equivalent to 0.36 µg DMPP, 0.25 µg ClMPP and 10 µg DCD g^{-1} dry soil.

Figure 2 showed the effect of increasing concentrations of DMPP on the decrease of ammonium, nitrite, nitrate formation and the pH change in the loamy soil. A comparison with Figure 1 clayey soil) that it showed as an almost identical to the measured parameters. Even in the loamy soil DMPP could only at about 500-fold of the base concentration effective to inhibit the ammonium oxidation in about 2 weeks. Thus, the minimum inhibitory concentration was temporarily to block the nitritation at 180 µg DMPP g^{-1} dry soil. A rapid decrease of ammonium still took place at the base concentration (0.36 µg DMPP g^{-1} dry soil). Compared to the control (with added ammonium), nitrite and nitrate formation was inhibited with increasing concentrations of DMPP. The inhibitory effect was already about 56% and 76% of control for nitrite and nitrate formation. The pH values were relatively constant in the first 2 weeks in all DMPP concentrations, indicating indirectly to delay of nitrate enrichment. The rapid pH drop after 14 days would indicate a decline in the inhibitory effect and on nitrate accumulation in a loamy soil.

Figure 2 is also shown the influence of rising ClMPP concentrations on the corresponding N reactions as well as on the pH values in loamy soil. As in the clay soil (Figure 1) found that a complete inhibition of ammonia oxidation in over a period of 1 to 3 weeks in first place of the 500 fold of base concentration (180µg ClMPP g^{-1} dry soil). The effect of ClMPP on nitrification was similar to how DMPP (Figure 1), as compared to DMPP; it seems that ClMPP affected the ammonium oxidation slightly more negative in this soil. And changes of the pH values were similar to those of the DMPP treatment (Figure 1).

In contrast to the clay soil (Figure 1) DCD could inhibit ammonium oxidation in loamy soil already over a 14 days period in the 250-500 fold of base elevated concentration. A portion of the added DCD was hydrolyzed but, then obviously strong and rash converted into nitrate. The pH values increased slowly in all DCD-concentration within 2 weeks significantly (DCD hydrolysis), but then decreased again. The decrease of pH (from pH 6.0 to about pH 5.5) was found in the two controls (with and without ammonium addition) was most intense on nitrification indirectly and indicated as an acidification point.

Sandy soil

The influence of increasing DMPP concentrations on the ammonium nitrite or nitrate formation and to the pH gradients in the sandy soil is presented in Figure 4. DMPP could not to inhibit the ammonium oxidation even at 500 times the base concentration completely. The nitrite and Nitrate concentrations of the untreated controls (with Ammonium) were at the highest. The differences between the concentrations were relatively low in terms of nitrite and nitrate formation. The simple base concentration had only a limited influence on the decrease in the ammonium concentration. The pH values were always significantly in the higher concentrations of DMPP over those of the control.

The influence of increasing ClMPP concentrations on the inorganic nitrogen transformations and on the pH values in the loamy sand is summarized in Figure 13 graphically. The effects of ClMPP were similar to those of DMPP (Figure 3). The pH values were increased at the end of the experiment, however, this indicate which indirectly to enhanced the nitrate formation.

The influence of DCD concentrations on the inorganic N transformations and on the pH gradients in the sandy soil is finally summarized in Figure 14. DCD inhibited ammonium oxidation in the 500 times of the base concentrations of in about a 7 days period to completely.

As in clay and loamy soil (Figure 1 and 2), DCD occurred to facilitate more intensive nitrate formation, confirming the incomplete inhibition of ammonium oxidation at the 100 -500 fold of base concentration. Apparently, part of the added DCD was rash degraded by microbial nitrification and speedily transformed into nitrate. The pH values were decreased significantly as a result of unbridled Nitratation. Overall, it came to conclude that DMPP and ClMPP began to inhibit the ammonium oxidation completely in over a period of 1 to 2 weeks at the 500-fold base concentration (minimum inhibitory concentration at 180µg DMPP g^{-1} and 125 µg ClMPP g^{-1} dry soils).

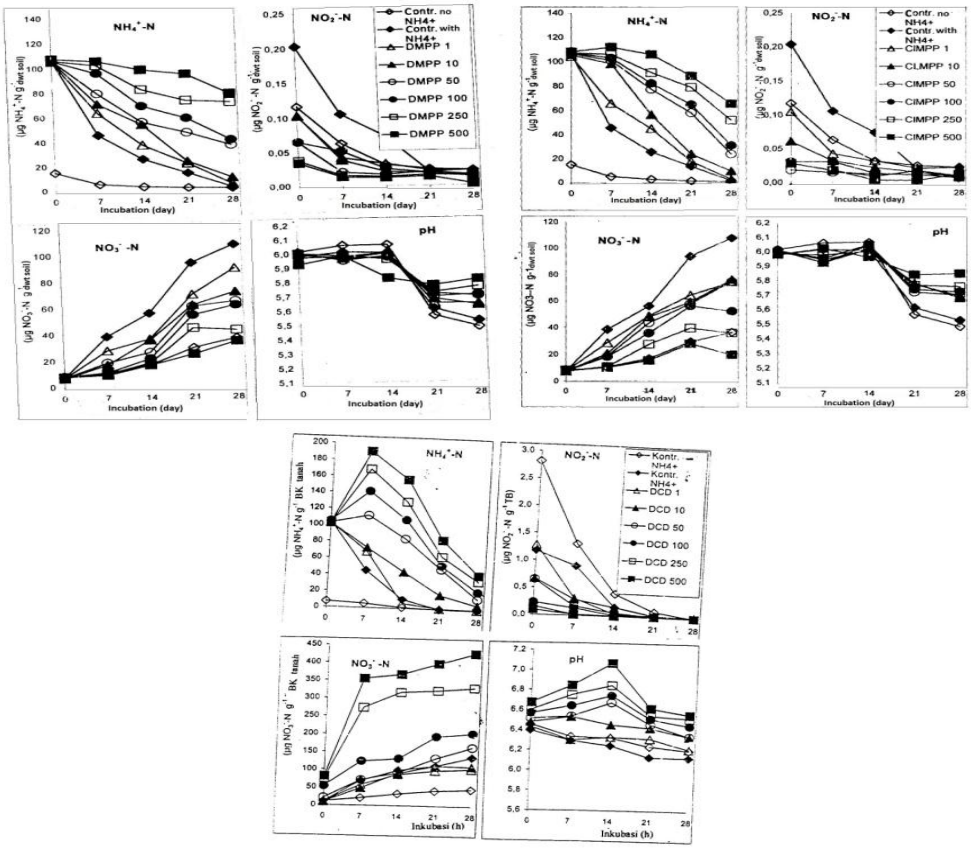

Figure 3: Effect of increasing DMPP, ClMPP and DCD concentrations on the ammonium oxidation, nitrite and nitrate formation and on the pH (H2O) values in sandy soil during an incubation period of 28 days (60% MWC, adding 100 mg Ammonium-N g^{-1} dry soil, at 25 ° C). The recommended concentration equivalent to 0.36 µg DMPP, 025 µg ClMPP and 10 µg DCD g^{-1} dry soil.

Influence of nitrification inhibitors on the potential denitrification capacity (PDC)

The influence of NIs- concentrations; DMPP, ClMPP or DCD on the potential denitrification capacity (PDC) and the NH_4^+, NO_2^- and NO_3^- concentrations in the three study soils were presented in Figure 4-6 and in Table 2. In clay soil occurred only an inhibitory effect on the PDC at about 10 to 50-fold of recommended concentration of DMPP, ClMPP or DCD (Figure 5). This No Observable Effect Level (NOEL- values) were for ClMPP already at 2.5 µg g^{-1} dry

soil , DMPP at 18 µg and DCD 250 µg g^{-1} dry soil. The Effective Dose of 50 % inhibition (ED_{50}- values) of ClMPP, DMPP and DCD were at about 187 µg, 270 µg and 2500 µg g^{-1} dry soil. ClMPP affected the denitrification at much lower concentration than as DMPP or DCD.

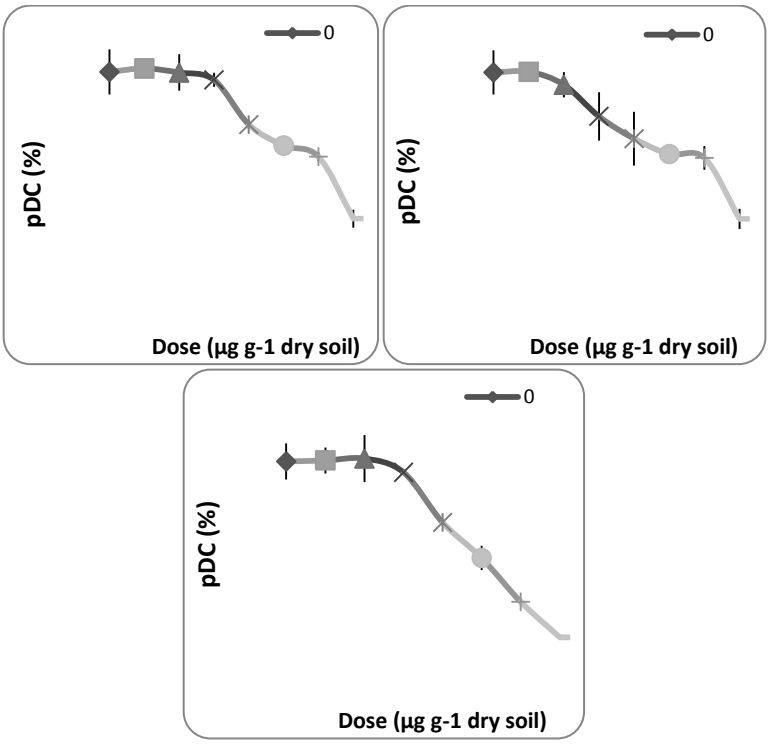

Figure 4.The effect of increasing the concentration of NIs; DMPP, ClMPP and DCD on potential denitrification capacity (% of control) in clayey soil. Recommendation dosage was 0.36 µg DMPP; 0.25 µg DCD; ClMPP and 10µg DCD g^{-1} dry soil

In the loamy soil (Figure 5), inhibitory effects of the three NIs on PDC occurred earlier as than in clay soil at 10 times of recommended concentration. The NOEL values for DMPP ClMPP and DCD were 3.6 µg and 2.5 µg, 100 µg DCD g^{-1} dry soil. At the concentrations of 105 µg ClMPP, 193 µg DMPP or 2049 µ DCD g g-1 dry soil, the ED50 values were achieved. Compared to the clay soil inhibitory effects of three NIs in loamy soil was reached at lower concentrations.

Similar to the loamy soil, the inhibition of PDC begun at about 10 times the recommended concentration of DMPP and ClMPP in the sandy soil (Figure 6, and Table 2.). The NOEL-values of PDC in sandy soil were 1.25µg ClMPP and at 3.6 µg DMPP g-1 dry soil, while by the DCD has already reached 100 µg g-1 dry soils, therefore at much lower concentrations than in clay and loamy soil. The ED50-values of ClMPP, DMPP and DCD are at about 98 µg, 150 µg or 1724 µg g^{-1} dry soil.

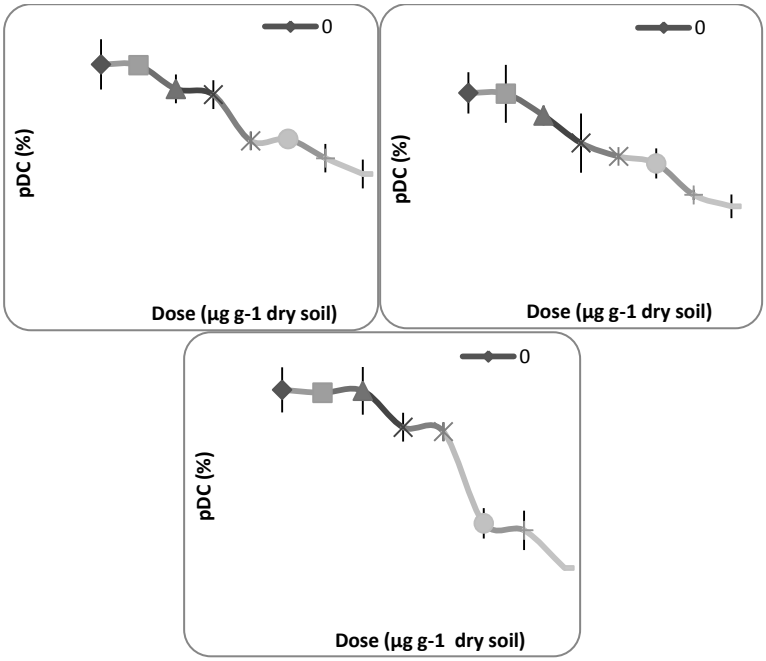

Figure 5. The effect of increasing the concentration of NIs; DMPP, ClMPP and DCD on potential denitrification capacity (% of control) in loamy soil. Recommendation dosage was 0.36 µg DMPP; 0.25 µg DCD;ClMPP and 10µg DCD g^{-1} dry soil

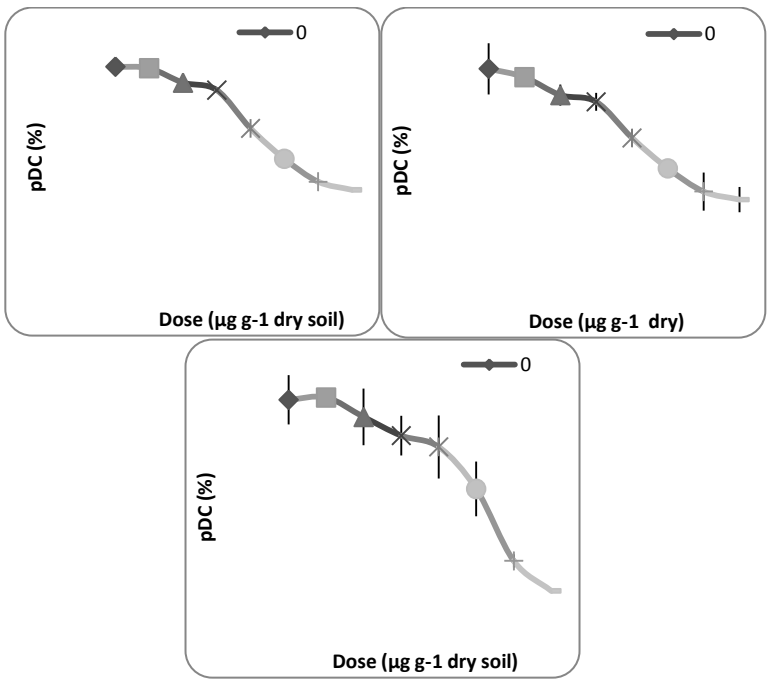

Figure 6. The effect of increasing the concentration of NIs; DMPP, ClMPP and DCD on potential denitrification capacity (% of control) in sandy soil. Recommendation dosage was 0.36 µg DMPP; 0.25 µg DCD;ClMPP and 10µg DCD g^{-1} dry soil

A comparison of these values with those in Figure 4 (clay) and Figure 5 (loam) showed that the inhibitory effects of ClMPP, DMPP and DCD decreased significantly in the three soil types in the ranking ClMPP> DMPP>> DCD. The influence of increasing concentrations of DMPP, ClMPP and DCD on the PDC and the concentrations of NH4 + -N, NO2-N and NO3- N in the clay, loamy and sandy soils were summarized in Table 2. It was clear that the nitrate formation decreased in the three experimental soils after the NI-treatment only in low mass, while the NH_4^+ levels increased and easily entered in the three soil types in comparison to the control. This confirms a slight inhibition of the PDC and ammonium oxidation. The NO2 - N-formation looks similar under NI application in the three test soils in comparison to the control (Table 2).

Table 2. The effects of nitrification inhibitors on potential dentrification capacity (PDC in µg N_2O g^{-1} dry soil 6 h^{-1}) in clayey, loamy and sandy soils.

x time of Recommended Concentration	Dose of Nitrification inhibitor (µg g^{-1} ds)	PDC (µg N_2O g^{-1} dwt.s 6h^{-1})		
		clay	loam	sand
	DMPP			
0 = Controll	0,0	3,87	1,886	3,96
1	0,4	3,93	1,871	3,94
10	3,6	3,86	1,730	3,48
50	18,0	3,73	1,657	3,37
100	36,0	2,96	1,281	2,48
250	90,0	2,60	0,985	2,52
500	180,0	2,41	0,760	2,15
	ClMPP			
0 = Controll	0,0	3,87	1,886	3,96
1	0,3	3,88	1,807	3,94
10	2,5	3,65	1,640	3,43
50	12,5	3,10	1,577	2,79
100	25,0	2,70	1,239	2,48
250	62,5	2,43	0,954	2,32
500	125,0	2,36	0,737	1,58
	DCD			
0 = Controll	0,0	3,87	1,886	3,96
1	10,0	3,89	1,909	3,89
10	100,0	3,93	1,719	3,94
50	500,0	3,64	1,537	3,15
100	1000,0	2,57	1,427	3,06
250	2500,0	1,81	1,016	1,09
500	5000,0	0,87	0,314	0,94

As the evidence available laboratory test data, the threshold of NIs-concentrations to provide a complete blockage of Nitratation over a period of 1-2 weeks for ClMPP was on average at 125 µg ClMPP and approximately 180µg DMPP g^{-1} dry soil and DCD was about 500µg up to 1000µg DCD g-1 TB (depending on soil types). The minimal inhibitory concentration with homogenous distribution in soils was around 15 times of recommended concentration for ClMPP or about 11 times for DMPP). These confirmed the relatively high specific effect of the two new nitrification inhibitors (DMPP and ClMPP). These thresholds inhibitory concentrations from the laboratory were resulted initially a contradiction to the

recommended dose for ENTEC in practice. The recent experiment reported that the effect of DMPP in practice was depending on the N-Fertilizing intensity. Further, DMPP in concentrations of 0.5 to 1.5 kg of active substance per ha seems was sufficient disincentive to inhibit nitrification in the field over a period 4 up to 10 weeks (Zerulla *et al*., 2001). By homogeneous distribution of this substance in a soil depth of 20 cm would the mean application rates DMPP-concentrations about 0.36 to 0.50 µg g^{-1} dry soil. The equivalent terms for ClMPP concentrations would around 0.25 µg up to 0.35 µ ClMPP g^{-1} dry soil. Such concentrations of DMPP or ClMPP would be lower in comparison to the above threshold inhibitory concentrations (250 to 500-fold recommended concentrations). With homogeneous incorporation into the topsoil DMPP should therefore in the recommended concentrations (0.5 to 1.5 kg DMPP ha^{-1}) inhibit the nitritation hardly efficient. DCD was applied in the field equal to about 10% of the fertilized N or at a dose of 90 kg N ha^{-1} in the topsoil or equals to 10 g DCD g^{-1} dry soil. This DCD concentration would not reach even with an intensive N fertilization up to 200-300 kg N ha^{-1}, in order to reach the above mentioned minimum inhibitory concentrations from the laboratory tests. Thus, there is a contradiction dose between the laboratory and in field applications.

In practice, however, DMPP as aggregates (with a grain diameter of about 4mm) formulated on ammonium-N applied, therefore it would not in a homogeneous distribution (Azam, *et al*., 2001; Zerulla, *et al*., 2001, Di *et al*, 2006). It can be assumed that the granules of ENTEC (N-fertilizer and DMPP) after rain hydrolyzed gradually with the result that ammonium and possibly DMPP (also CIMPP) to diffuse rapidly among the granules into the soil. It took consequently in the field, temporally and spatially different concentration gradients of DMPP (or CIMPP) and ammonium, which for DMPP between 0 and about 100 µg g^{-1} dry soil hat a granulate distance (in the 0-5 mm zone of the granule center) are expected (Azam *et al*., 2001). A comparison of the inhibitory concentrations for further inhibition of nitritation in the laboratory showed that a complete inhibition of ammonium oxidation was temporally and spatially under the pellets and around them is around given the least, especially since about 80% of DMPPs remains in a loamy soil over a 10-days period in around during 0-5 mm of the granules (AZAM *et al*, 2001). However, it remains very difficult to know the real, locally and time and the process of granules to inhibit nitritation in the field, because the concentrations of DMPP (or CIMPP) and ammonium because of the different diffusion rates should change continuously. It is likely that DMPP and CIMPP in soils with usual pH values from 5 to 6.5 predominantly active cation (diffuse through Protonitation) and due to their molecular sizes significantly slower than the ammonium. In Laboratory experiments, which were carried out under standard conditions and at different temperatures (4, 15, 25 o C) and soil moisture (18 or 20% of the $_{M}$WHC), let the example of a silty clay showed that only 5-15% of DMPPs after 10 days were in the 25 -40 mm zone around the granules, which confirmed the very low mobility DMPPs (Azam *et al*., 2001, Di, *et al*, 2007). In soils, the ratio of ammonium was changed to DMPP in the course of the time probably constantly. The inhibitions of Nitritation in the field were depending on such conditions, which in model experiment are hardly possible to create them. From the above considerations' can be concluded that the nitrification inhibition must take place substantially in close contact with the DMPP-granules. To clear up this point, some more field experiments in different soil types should be conducted.

Conclusions

Based on the obtained results above, the following conclusion can be drawn :

1. In all tested soil, the applied NIs; DMPP, ClMPP and DCD inhibited ammonium oxidation at recommended concentration only partially. On the average up to 125µg ClMPP, 180µg DMPP and 2500µg DCD g^{-1} dry soil were needed to inhibit the nitrification completely. The recommended applications rate of 0.5 kg - 1.5 kg DMPP ha^{-1} which corresponding to 0.36 µg DMPP - 0.50 DMPP µg g^{-1} dry soil were far below the concentration level, that inhibits ammonium oxidation completely in the three investigated soils. Thus even at highest tested inhibitor concentrations, nitrate and nitrite formation still occurred. A decrease in the soil pH was observed in all experiments but highest in control plots.
2. Effect of various concentrations of inhibitors nitrifications on potential denitrification capacity has been occurred clearly at 50 times of the recommended dose on clayey soil equals to 18 µg DMPP, 12.5 µg ClMPP and 500 µg DCD g^{-1} dry soil, and 10 times of recommended dose on loamy and sandy soils or equals to 3.6 µg DMPP, 2.5 µg ClMPP and 100 µg DCD g^{-1} dry soil.
3. Generally, ClMPP exhibited the strongest influence on soil mineral nitrogen dynalics in the three soils compared to DMPP and DCD. The NIs was generally the most effective in sandy soils than in clay or loamy soil.

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Yields Increasing of Sweet Potato (*Ipomea batatas l.*), Variety of Beauregard by Organic Cropping System at Desa Cilembu, Sumedang

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Abstract

Desa Cilembu, Sumedang is one village from several central production of sweet potato in West Java. One of the new sweet potato varieties was Beauregard from Solomon Island has been cultivated in this village and the yield of tuber currently decrease caused by increasing of soil bulk density and decreasing of organic contents. The application of organic cropping system is one way which can use to solve this problem. The aim of this research was to know the effect of a straw compost application as organic cropping system on soil compaction, soil moisture content, growth and yield of sweet potato. Field experiment was carried out during February until June 2011, at Desa Cilembu, Sumedang Regency. Experiments were arranged in Randomized Block Design using 5 doses of straw compost as treatments and replicated five times so that obtained 25 plots where each plots size is 1m x 0.75m. The treatments were without straw compost and applied artificial fertilizer (K1), 10 ton/ha straw compost and organic manure (K2), 15 ton/ha straw compost and organic manure (K3), 20 ton/ha straw compost and organic manure (K4), and 25 ton/ha straw compost and organic manure (K5). Result showed that 20 ton/ha dose of straw compost and applied 5 ton/ha organic manure on soil compaction was more decreasing, whereas soil moisture content, plant growth and tuber yield was more increasing. The average yields of treatment without straw compost and use artificial fertilizer was 9.78 ton/ha, grade B dominant, and fond, while organic cropping system by given 10 ton/ha; 15 ton/ha; 20 ton/ha; and 20 ton/ha straw compost and each treatment was applied 5 ton/ha manure as organic fertilizer respectively reached 15.12 ton/ha, grade A dominant, and very fond.

Keywords : organic cropping system, straw compost, sweet potato.

Introduction

Sweet potato is assumed it come from America. One botanist have estimated that regions as the origin of sweet potato were New Zeland, Polinesia, and midle America. Nicolay Pvanovich Vavilov, a Soviet botanist, fixed that the primary centrum as the origin of sweet potato is Middle America. Sweet potato began to spread out to entire the world in particular to the countries with tropical climate on the sixteen century. The Spains spread the sweet potato to Asian, in particular to Phillipine, Japan and Indonesia (Rukmana, 1997).

In Indonesia, many varieties of superrior potatoes have cultivated, among of them is Beauregard variety which was come from the Island of Solomon. But in developing this variety is not yet known even though it has high potential productivity is about, 22.6 ton/ha. The best texture soil for this variety is sabndy clay (Tjintokohadi and Ezette, 2010).

Desa Cilembu (Cilembu Village) is one central production of sweet potatoes in West Java. Many of sweet potatoes in different varieties those have been cultivated in Cilembu Village and Beauregard was one variety that is begun to cultivate. However, in it's development there was some problems, such the decreasing the quality of the existing soil in Cilembu Village. According to Tohidin (2006) there was occurence in the increasing of content

weight, hardening of soil in Cilembu Village. According to Arifin (2002) in the soil capability map expose at West Java reported that the kind of soil in the areal of Cilembu Village is Latosol (equivalent with Inceptisol, according to the Soil Survey Staff, 1999). Kind of soil like Latosol or in equivalent with Inceptisol have a value content weight as much as 0.93 – 1.11 g/cm^2 (Sarief, 1989).

In order to overcome those problems that can be conducted by the application of organic cropping system, mainly the organic fertilizer like manure and mulch (Solihin, 2007). The utilizing of fertilizer organic in those soil could incrase the content of organic materials (C) and also it could improved the physical soil nature, such as increasing the land water content and to decrease the content weight of those soil (Abiyanto, 2009).

Straw and agriculture yield waste from rice which is great enough in amount and it was not fully used. Rice straw production to day can reach 12-15 ton. The result from the research in Balai Penelitian Tanaman Pangan (Food Research Agency) showed that using of the straws have an influence to increase the sweet potatoes crop yield as much as 11,03%. However the using of straws have some weaks and could caused deficiency on sweet potatoes cultivation, in which, the growth of sweet potatoes, in the first month, have a few yellowish and they have a recovery and have a normal growth in the next month. It is caused by the occurance of nutrition competition elements between composting organism and plant. Therefore, it is better that the straws must be composted previously (Rukmana, 1997).

Compost are the organic materials that has peat process because there was an interaction among microorganism that they worked inside. Using compost as fertilizer is best because they can improve the nature in chemical, physical, and biology of soil, therefore they can increase soil moisture, to facilitate the growth of plant roots, to increase porosity, aeration, to improve drainage, to improve the compositition of soil microorganism, and to provide the macro and micro nutrition elements of soil (Murbandono, 1994).

With the cultivation pattern which have been applied by the community in Cilembu village, commonly rice-rice-sweet potatoes or rice-sweet potatoes-rice in a year, the availability of agriculture yields waste from the rice plantation, especially straws that can be used to converted into straw compost. The fertilization with organic materials such straw compost can become the proper method to overcome the above problems.

Thereby, it is felt a necessary the research that has conducted for “the effect of the using of the straws compost on the soil density and the growth and the yield of sweet potatoes (*Ipomea Batatas L*) cultivations with Beauregard variety in Cilembu Village, Sumedang Regency”. This research was designed, them to find out the effect of straws compost utilization on the soil density which is stated with the content weight, land water content and the growth then the yield of Sweet Potatoes Variety Beauregard Cultivation and the yield potential of Sweet Potatoes Variety Beauregard cultivation which was cultivated in the soil in the Cilembu Village.

Materials and Methods

The research was conducted in rice field at Cilembu village, Sumedang Regency, during January up to June 2011. The condition of the research soil, as following: Altitude of place is 900 – 1.000 m on over of the sea level, type of rainfall is C (rather wet), air humadity, 74,4 –

92,2%, air temperature 22,4 – 23,6^{o}C, Photoperiod was about 70,9 – 212,7 hour/month, and soil orde : Inceptisol and – granular.

The Tools were included: Hoe for cultivation of the sweet potatoes, to make a stack and to stir the compost and soil, label to remark a certain straws compost + manure and repetition of each unit of experiment as much as 25 units, *Emrat* designed to put water into plant, meter roll with minimal 10 m to measure a cultivation compartmen, pair of weight digitally, to consider the yield of sweet potatoes and data of the straws compost that will be given to each treatment and the soil sample, sample ring and stalk to press the sample ring in order to make a sampling of soil, wood hammer, thin and sharp knife, hoe, to take a soil and compost samples, bowl for placing the soil and compost samples, drying oven, to dried the soil sample and to measure the content weight and the land water content, clipper stalk, to take a sample of soil from oven, iron pipe (ø = 0,5 inch), to take a soil sample from the land, calculator, and software Excell 2007.

The Material were: sweet potatoes Seed of Beauregard variety which was yielded from the stem cutting or a tip cutting with lenght 25 cm that has two month old or more and they have stored during 4 days as much as 75, the straw compost as much as ± 15 kg that was obtained as a results of the composting, Soil from Cilembu Village, Sumedang Regency, and water for irrigation and composting.

The Method used in this experiment were, Randomized Block Design to try 5 doses treatment improviding the straws compost which replicated five times, thus there were 25 experimental compartments. Each of compartment is 1 m x 0,75 m in size. Usually, to cultivate the tuber plant used the compost and manure as much as 15 ton/ha (Wibowo, 1989). The following description for each treatment in providing of straw compost doses :

K_1 = 0 ton / Ha or 0 kg/compartment
K_2 = 10 ton / Ha or 0. 750 kg/compartment
K_3 = 15 ton / Ha or 1.125 kg/compartment
K_4 = 20 ton / Ha or 1.500 kg/compartment
K_5 = 25 ton / Ha or 1.875 kg/compartment

All of each treatments above added by 5 ton/ha or 0.375 kg/compartment manure as organic fertilizer. For the interest of analysis on Randomized Block Design, it was used a linear model in additive structure (Gaspersz, 1995).

$$Y_{ij} = \mu + \tau_i + \beta_j + \epsilon_{ij} \quad \text{..... (1) ; } i = 1, 2, ..., t \text{ ; } j = 1, 2, ..., r$$

In where Y_{ij} = Observation values on ith treatment in the recurring group up to j th
μ = Mean value of population
τ_i = Additive effect of recurring group for i th
β_j = Additive effect from recurring group for j th
ϵ_{ij}= Error effect of experiment from treatment ith on the recurring group j th

Results and Discussion

Sweet potatoes cultivation is categorized as annual plant that has a main body structure which consist of stem, tubers, leafs, flowers, fruits, and seeds. Cultivated stem has a circular form, no wood, no joint, type of its growth is upright and creeping. Cultivation with upright type has between 1-2 meter in length. The size of stem is classified into three group, that is,

big, moderate, and small. Usually the stem has an old green in color up to purplish (Rukmana, 1997).

Leaf form of the sweet potatoes are divided into three group that is, heart and round form, ellips, and spikes circle. A plant of sweet potatoes have many leaves in amount. A sweet potatoes variety with wide leaves have a higher productivity compared accompanying sweet potatoes with narrow leleaves, because the wide leaf have a better and effective photosyntezied than the sweet potatoes with narrow leaves. Sweet potatoes leaves have an old green in color and green-yellowish (Rukmana, 1998).

Floral wreath grow on the armpit of leaf. The sweet potatoes have a flowers with similar to trumpet in form, it is structured from five sheets of crown leaf, five sheet of flowers leaf, and a pistil stalk. Flower crown has a white and purple in color. Usually, the sweet potatoes flower with mature or reade to have a pollination in the morning which is begun on 04.00 – 11.00 hours. The pollination has occured, and flower being a fruit. Fruit of the sweet potatoes have a three boxes, has a hard peel and seeds (Rukmana, 1998 & Juanda, 2000).

Forming sweet potatoes in the sweet potatoes plant generally occurred in 20-25 days after cultivated. The sweet potatoes forms generally are from a round up to ellips with the surface in level or not level. Level form (round and ellips) and there is no many dents included into the sweet potatoes that have high quality (Rukmana, 1998 & Juanda, 2000).

Sweet potatoes peel structure has a variation between thin up to thick and generally it has a sap. Sweet potatoes have a white, yellow, purple, or purple reddish in color and they are depend on their varieties. Sweet potatoes has a flesh with the color related to the nutrition up to 7.100 IU. The flesh colors of sweet potatoes were white, yellow, or orange and a faw purple. The sweet potatoes that has a high beta caroten content only derived from orange and reddish sweet potatoes. Sweet rather that its power content is lower.

There are growth, yield, grading and organoleptic test of sweet potatoes variety Beauregard in Cilembu village, Sumedang Regency.

Total Sweet Potatoes Grown

The result from analysis on the total growth of sweet potatoes can be seen in Table 1. results showed that providing the straw compost have no significant effect to the growth amount of sweet potatoes, neverthelles, more providing compost, the total groeth of sweet potatoes more increase, with the increasing among 47-80%.

Table1. The effect of Providing the Straws Compost with 5 ton/ha Manure as Organic Fertilizer on Total Growth of Sweet Potatoes.

Treatment	Amount of Sweet Potatoes (pc)	Test Yield	Comparing (%)
No Compost	25	b	100
K2 (Provide compost 5 ton/ha)	31	ab	124
K3 (Provide Compost 10 ton/ha)	34	ab	136
K4 (provide Compost 15 ton/ha)	36	a	144
K5 (Provide compost 20 ton/ha)	36	a	144

Note : Treatment in average which was marked with the ssme alfabet have no significant differences according to Duncan test with confidence level 5%

From the Table 1, it can be seen that the providing the straws compost 10, 15, 20 and 25 ton/ha there was no significant more increase along with the increasing in amount of providing the compost that has been given, but the providing the straws compost 20 ton/ha have a significant differences compared with treatment without compost.

Total Sweet Potatoes Yield

The result showed that providing of the straws compost have a significant effect on the yield of sweet potatoes per compartment and per ha. More providing compost, more the sweet potatoes per compartment and per ha with the increasing among 114 – 230%.

From the Table 2, it can be seen that the providing of the straws compost 25 ton/ha have no significant differences with the providing 20 ton/ha however the amount of differences was enough large, that is, 1,03 ton/ha but both have a significant differences with other treatment.

The soil density and soil water content observed as supporting data. Providing straw compost 20 ton/ha and manure 5 ton/ha produce the lowest soil density (0,75gr/cm^2) and have a significant differences with the other, while for the soil water content (50 % weight). Table 1 and Table 2 showed that treatment providing straw compost 20 ton/ha and manure 5 ton/ha in generally have highest value for growth and yield of sweet potatoes.

Table 2. The Effect of the Straws Compost and with 5 ton/ha Manure as Organic Fertilizer on Sweet Potatoes Yield

Treatment	Sweet Potatoes Yield (ton/ha)	Test Yield	Comparing (%)
No compost	6.78	c	100
K2 (provide compost 10 ton/ha)	9.97	b	147
K3 (provide compost 15 ton/ha)	13.32	b	196
K4 (provide compost 20 ton/ha)	15.12	a	223
K5 (provide compost 25 ton/ha)	16.15	a	238

Note : Treatment in average which is marked with the same alfabet have no significant differences acording to Duncan test with confidence level 5%

Grading and Organoleptic Test

Table 3. The Effect of the Straws Compost and with 5 ton/ha Manure as Organic Fertilizer on Sweet Potatoes Grading and Fond Test.

Treatment	Dominant Grade	Fond Test
No compost	C	Fond
K2 (provide compost 10 ton/ha)	B	Fond
K3 (provide compost 15 ton/ha)	A	Fond
K4 (provide compost 20 ton/ha)	A	Very fond
K5 (provide compost 25 ton/ha)	A	fond

Note : Treatment in average which is marked with the same alfabet have no significant differences according to Duncan test with confidence level 5%

Sweet potatoes grading was used grade standard from Food Research Agency. Grade A of sweet potatoes weight is more than 400 grams/pc, grade B 1s 200-400 grams/pc, and grade C is less than 200 grams/pc. Organoleptic test conducted in August 2011 by 25 students as tester. Result of grading and organoleptic test showed in Table 3.

Conclusion

Organic system cropping for sweet potatoes, variety Beauregard, in Cilembu village, Sumedang Regency was caused the density of soil more decreasing, whereas the water content, growth and cultivation yield more increase. Providing the straws compost 20 ton/ha and manure 5 ton/ha yielded the lowest soil density (0,75gr/cm^2) and have a significant differences with the other, while for the soil water content (50 % weight), the growth and yields (15.12 ton/ha), dominant grade A, and very fond, in generally have highest value, eventhough have no a significant differences with providing 25 ton/ha

Suggested tried to apply on the soil in wider areal, and conducted the research in similar with providing the other organic ferilizer which was much available in this area, such as manure from the bulls and cock which was conducted usually by the local farmers.

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AGROSOCIO-ECONOMY

The Potentials and Pitfalls of Ecotourism Development on Natural Resources Conservation Area in Indonesia

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Abstract

Any expert on ecotourism will always objectively say that Indonesia has more than abundant resources for ecotourism, not only in number and variety but also in distribution and demand. However, after more than 20 years the ecotourism paradigm has been adopted in Indonesia –as well as more than 30 years nature conservation area also was developed in Indonesia – unfortunately the development processes are not giving a significant result. A good understanding on the characteristics of resources and demand is a keyword to grab the potentials, meanwhile being honest to the dynamic of management is demanded to solve the pitfalls. Knowing well about ecotourism-resources characteristics is very important for finding and promoting the uniqueness of ecotourism objects and activities. Having detail in demand characteristics is very useful to mobilize and distribute the number of visit and the number of visitor as well. Further, frankly assessment in man-power is needed to bridge the obstacles.

Keywords: ecotourism, nature conservation area, Indonesia

Introduction

For many years since the early of 70s, Indonesia has been putting tourism sectors as a primary sector (after gas and oil) to support the national income, but up to now the result is still hard to be classified as advantageous. Even in the early year, Bali, Bunaken, Borobudur, Toba Lake and Tana Toraja have been appointed by the government to became major destination at national level, but to date only Bali which has its fruitful-ways to gain the tourism benefit at a certain level and aspect. For many decades Bali has played important role contributing the number of international tourist to Indonesia which is for the year of 2010 totally Indonesia only grab 7 millions foreign tourists (that speechless comparing to 24.58 millions foreign tourist in Malaysia, 9.16 millions foreign tourist in Singapore, 15.84 millions foreign tourists in Thailand, and 55.67 millions foreign tourist in China on the same year).

The same story of "unclear" development dynamics is also facing the ecotourism development in Indonesia. At least 10 years before ecotourism-paradigm boosted by scholars and practitioners world-widely on early 90s, actually since 1980 Indonesia already has started to develop its conservation area nationally with "high-speed" motivation on conservation and outdoor recreation programs. Nowadays, Indonesia might be the single country which has a such big number of conservation area in the world; not only about the wide of area but also about the categories. Nationally, to date due to the national official-data there are 50 National Parks (12.298.216 Ha in total), 248 Restricted Nature Areas (4.586.665 Ha in total), 75 Wild-life Restricted Areas (5.099.849 Ha in total), 14 Game Reserves (224.816 Ha in total), 22 Provincial Parks (344.174 Ha in total) and 105 Nature

Recreation Parks (257.348 Ha in total) located at almost any province in Indonesia. In number and area, it may call enormous, unfortunately but not yet for the number of visitors that only 670.084 visitor of National Parks and 1.160.296 visitors of Nature Recreation Parks in the average of the last five years.

Any tourism expert who pay attention on Indonesia, objectively, will always say that Indonesia has spectacular potential of tourism, both in term of resources and demands. However, up to now no body who are willing to tell all the truth of weaknesses bringing the "unsatisfied-condition" of tourism development in Indonesia. After more than 40 years Indonesia did all the best to develop and gain benefit from tourism sectors, probably it is a time to find the answers why the results are still far away from hope. Therefore, this paper will try to share a twenty-years experiences in ecotourism fields which describes the dynamics of ecotourism sectors in Indonesia; especially focused on the potentials and pitfalls of ecotourism development on nature conservation programs as a significant element of the whole ecotourism sectors.

Material And Methods

This paper was written based on professional experiences (as an academician and practitioner) of more than 20 years experiences involved on many ecotourism studies and projects in Indonesia. The primary data for this paper was based on travelling-log-books; in which personally the author always wrote down all observation result during any project. To write this paper, the travelling-log-books have been re-read to extract and figure-out the potentials and pitfalls of ecotourism development on nature conservation programs in Indonesia. Further, to date secondary data have been used to articulate and to complete and up-date the recent situation and information of any related object cited for the primary data.

The evaluation method of Avenzora (2003 and 2008) has been applied to approach and evaluate any information from travelling-log-books. The uniqueness, beautifulness, rareness, accessibilities, seasonality, social-functions and sensitivities of each destination area in the log-books are becoming major criteria to evaluate the potentials of resources. Further, education system, human resources, bureaucratic-system, and ecotourism net-working are turning into the main of external ecotourism-element to appraise and describe the pitfalls. Further, the Likert-scale has been enlarged to become 1 to 7 (instead of score 1 to 5; where score 1 is for very bad gradually up to score 7 is for a very good) and used to score the condition of any potential and pitfall. Further more, any criteria of potentials and pitfalls has been broken-down into 7 indicators in order to build a systematic scoring system. Finally, an expertise-judgment of 3 experts on ecotourism has been applied to point out final score of any criteria.

Results And Discussions

Almost all provinces in Indonesia has one national park, which are only West Papua Province, West Sulawesi Province, South Kalimantan Province, Bangka Belitung Province and Riau Islands Province those are having no national park in the area. While, Jambi Province, Papua Province, East Kalimantan Province, Central Kalimantan Province and West Java Province have two national park in their area. Further, West Kalimantan Province, East Nusa Tenggara Province, and East Java Province are having 3 and 4 national parks in their area. The detail number of nature conservation areas was shown at Table 1 and the

distribution of nature Conservation Area at any province in Indonesia can be seen on Table 2.

Table 1. The Number of Nature Conservation Area in Indonesia

No.	Nature Conservation Area	Amount	Area (Ha)
1.	Strict Nature Conservation Area		
	a. Terrestrial	243	4.333.630
	b. Ocean	5	152.610
2.	Wildlife Conservation Area		
	a. Terrestrial	73	5.052.973
	b. Ocean	2	5.220
	National Park		
3.	a. Terrestrial	43	12.284.031
	b. Ocean	7	4.043.541
4.	Recreational Nature Area		
	a. Terrestrial	104	258.469
	b. Ocean	17	491.284
5.	Provincial Park	21	344.174
6.	Game & Hunting Park	14	225.103
	Total Amount of Area		
	a. Terrestrial	452	22.498.383
	b. Ocean	39	4.692.655

Source: calculated from www.dephut.go.id

Due to the distribution pattern of conservation area in Indonesia, can be concluded that those conservation areas are actually located in golden-distance from the central of population. While, the Table 3 shows that the ecotourism resources elements at the national parks are averagely scored by the experts as 6 for the uniqueness, 6.5 for the beautifulness, 6 for the rareness, 6 for seasonality, 5.5 for sensitivity, 3.5 for accessibility and 5.5 for the social-functions. Furthermore, the total number of visitor of main categories of nature conservation area in each province is shown on Table 4; wherein indicates that the ratio of the total number of visitor to the conservation area and the total number of population at the province is not yet showing a better rate of the number of visit.

Regarding the result of scoring, it may conclude that the deficient of actual demand at any conservation area in each province is not attributable to the quality of on-site ecotourism experience (as the internal ecotourism elements) at the focal-point of each ecotourism destination on nature conservation area, but it is strongly predicted that the lack of actual demand mostly down to the external ecotourism-elements and experiences at the surrounding area of the main focal point. Many literatures (e.g. Clawson, 1966) stated that the quality of total ecotourism experiences is divided into 5 phases of ecotourism experiences, i.e.: planning phase, departure phase, on-site experience phase, on the way back phase and recollection phase. While, Avenzora (1997) found that the quality of the in-situ ecotourism experiences in Gede-Pangrango National Park amongst the respondent was only support 19.3% of the total quality of their ecotourism experiences in BOPUNJUR (Bogor-Puncak-Cianjur) Destination Area.

Table 2. The distribution of nature conservation area in Indonesia

No	Province	Recreational Nature	National Parks	Provincial Parks

		Parks					
		Amount	**Area**	**Amount**	**Area**	**Amount**	**Area**
1.	NAD	2	231,400	1	1,094,692	1	6,300
2.	North Sumatra	5	3,071	1	108,000	1	51,600
3.	West Sumatra	4	40,509	1	190,500	1	12,100
4.	Riau	2	6,778	1	144,223	1	6,172
5.	Kep. Riau	NA	NA	NA	NA	NA	NA
6.	Jambi	1	425	4	1,651,286	1	15,830
7.	Bengkulu	3	15,877	NA	NA	1	1,122
8.	South Sumatra	1	50	1	202,896	NA	NA
9.	Bangka Belitung	NA	NA	NA	NA	NA	NA
10.	Lampung	NA	NA	2	490,621	1	22,245
11.	DKI Jakarta	1	99	1	107,489	NA	NA
12.	Banten	NA	NA	1	123,156	NA	NA
13.	West Jawa	18	4,971	3	143.553	3	632
14.	Central Jawa	5	270	3	123,760	1	231
15.	DI. Yogyakarta	2	4	NA	NA	1	617
16.	East Jawa	3	20	4	176,696	1	27,828
17.	Bali	3	15,566	1	9,003	1	1,392
18.	West Nusa Tenggara	9	13,428	1	41,330	1	3,155
19.	East Nusa Tenggara	8	102,647	4	314,054	1	1,900
20.	West Kalimantan	2	835	4	1,203,090	NA	NA
21.	Central Kalimantan	3	2,064	2	983,740	NA	NA
22.	South Kalimantan	2	2,135	NA	NA	1	112,000
23.	East Kalimantan	2	61,930	2	1,559,129	1	61,850
24.	North Sulawesi	3	3,196	2	367,720	NA	NA
25.	Gorontalo	NA	NA	NA	NA	NA	NA
26.	Central Sulawesi	5	92,738	2	580,596	1	7,128
27.	South Sulawesi	11	152,870	2	574,515	1	3,475
28.	Southeast Sulawesi	4	163,500	2	1,495,194	1	7,877
29.	West Sulawesi	NA	NA	NA	NA	NA	NA
30.	Maluku	5	5,085	2	356,300	NA	NA
31.	Papua	7	197,323	3	4,317,310	NA	NA
	Total	**111**	**1,016,936**	**50**	**16,215,444**	**21**	**343,454**

Source: Calculated from The Ministry of Forestry 2010.

Table 3. score of the ecotourism potential at nature conservation area in Indonesia

No	**Nature Conservation Area**	**Evaluation Criteria**						
		A	**B**	**C**	**D**	**E**	**F**	**G**
1.	Strict Nature Conservation Area	4	4	4.5	4.5	4	6	4
2.	Wildlife Conservation Area	4	4	5	4	4	6	4
3.	National Park	6.5	6	6	6	5.5	5.5	3.5
4.	Recreational Forest Park	5.5	5.5	4	6	4.5	4.5	6
5.	Provincial Forest Park	5.5	5.5	4.5	6.5	4.5	5	6.5
6.	Game and Hunting Area	5	4.5	4.5	4.5	5	6	4

Note: A. Beautifulness; B. Uniqueness; C. Rareness; D. Seasonality; E. Social Function; F. Sensitivity; G. Accessibility

Table 4. The Number of Ecotourism Visitor at Nature Conservation Area on 2010 in Indonesia

No	Province	Number of visitor	Number of Population	Ratio (%)
1.	NangroeAceh Darussalam	923	4,494,410	0.021
2.	North Sumatera	477	12,982,204	0.004
3.	West Sumatera	8	4,846,909	0.001
4.	Riau	136	5,538,367	0.003
5.	Riau Island	NA	3,092,265	NA
6.	Jambi	4799	7,450,394	0.064
7.	South Sumatera	113391	1,715,518	6.609
8.	Bangka Belitung	NA	7,608,405	NA
9.	Bengkulu	699	1,223,296	0.057
10.	Lampung	37725	1,679,163	2.247
11.	Banten	4356	9,607,787	0.045
12.	DKI Jakarta	981	43,053,732	0.003
13.	West Java	1059453	32,382,657	3.272
14.	Central Java	73452	3,457,491	2.124
15.	Yogyakarta	64490	37,476,757	0.172
16.	East Jawa	162476	10,632,166	1.528
17.	Bali	35729	3,890,757	0.918
18.	NTB	12023	4,500,212	0.267
19.	NTT	44982	4,683,827	0.960
20.	West Kalimantan	432	4,395,983	0.009
21.	Central Kalimantan	10194	2,212,089	0.461
22.	South Kalimantan	1384	3,626,616	0.038
23.	East Kalimantan	574	3,553,143	0.016
24.	North Sulawesi	10288	2,270,596	0.453
25.	Gorontalo	NA	2,635,009	NA
26.	Central Sulawesi	4524	8,034,776	0.056
27.	West Sulawesi	NA	2,232,586	NA
28.	South Sulawesi	714485	1,040,164	68.689
29.	Southeast Sulawesi	1761	1,158,651	0.152
30.	Maluku	226	1,533,506	0.015
31.	North Maluku	NA	1,038,087	0
32.	West Papua	77	760,422	0.010
33.	Papua	6961	2,833,381	0.247

Note: 1. The number of visitor is an aggregate number of domestic and foreign tourist.
2. Calculated from The Ministry of Forestry 2010.

More, the insufficient actual demand is not only an important indicator for saying that the national park and other conservation areas are not yet becoming a popular site to be visited by the inhabitants in each province, but also describing there are many lacks of prerequisite to drive the potential demand into an actual demand. Ideally, in easy speaking the potential demand of conservation areas in each province should be equal to the total population in the province; that might be equal to at least a single visit by any inhabitant to any chosen area in the province per year.

There are two reciprocal conditions which are robustly envisaged be the occasion of low actual demand at any nature conservation area (as a recreation or ecotourism destination) in each province, i.e.: the population annual income and the condition of infrastructure.

Almost all provinces in Indonesia (except Jakarta) still in a condition of low average income of population; which is leading to a low expenditure to consume. In the last 10 years, the average of population per capita income in Indonesia is only around 1350 USD (650 USD in 2000 and 2900 USD in 2010; Biro Pusat Statistik, 2011). Avenzora (2004) reported that by this income, the population can not put a recreation activity as a primary need; which is even for the last 10 years there were more population in the city area already put the recreation activity as the secondary need (move from tertiary need on the 90s) but most of them still decided to use their income for a culinary related recreation activities in the city they live.

Further, recreation activities actually already increase and take better position on their living-need, but their recreation pattern is not yet belonging to ecotourism-travelling-statistic (as their activities do not yet meet the tourism-statistic criteria). Further, the condition of low income also create a domino effect to the conservation area management; as the conservation area institution hard to put a ticket price (for any service of recreation and ecotourism activities) on an ideal rate. In 1998 the ticket price for a visitor to national park was only IDR 1500, and this price remain stable (if can not said as "has no chance to be increased") until the year of 2010 where the ticket price offered at IDR 2500. The result was the income for conservation area institution also became low, therefore it is understandable that the conservation area institution become incapable to set their "investment" schemes on an ideal schedule and being infertile to manage their area in better way. These condition bring many loops of reciprocal negative impacts; both on resources-management aspects and on demand-behavior.

Telling the condition of infrastructures, Avenzora (1997) found that 78 % of respondent put the regular transportation schedule to a natural conservation area becoming a major obstacle for the population going to the destination. Actually the accessibility to destination already provided by a better route of national-road up to regency-road, but the transportation modes are not yet fulfill better console for the recreation-taker during their travelling to the nature conservation area as a destination of their recreation or ecotourism activities. Further, Avenzora (2004) found that 81% of respondents asked for a direct shuttle-bus (instead of connecting transport) to travel them from their city to the recreation focal point at the nature conservation area; which was not only to decrease the cost for transportation but also in order to make sure that they will have more time to do recreation activities in the destination (with 4-5 hours on travelling) without any worry to missed "the last train" to back home.

Beside all those dynamics of ecotourism resources on nature conservation area in Indonesia (as described above), at least there are two important issues that have to be portrayed for discovering the "unclear-phenomena" of ecotourism development on nature conservation area in Indonesia, i.e : the human resources capacity and the bureaucratic-system. Outspokenly, the capacity of human resources in the system of ecotourism development on nature conservation area in Indonesia is still in a very serious problems; not only in term of final professionalism appearances but also in terms of education back-ground, experiences and working-motives that all needed to build a better professionalism appearances.

The root of problem in human capacity on the ecotourism development on nature conservation area in Indonesia is the education back-ground. Even there is no official data that can be cited to describe the condition, but one may claim that more than 95 % human resources in the system of the ecotourism development on nature conservation area in

Indonesia are not having sufficient knowledge on ecotourism-field. Other people may say that the claim is just a very subjective and unfair, but probably they will change their comment if they try to trace the root of problems starting from the tourism-education history in Indonesia. Since 1956 (the year of NHI in Bandung was found) most of the tourism education in Indonesia until now is still only focused on hotel management, travel management and tourism economic; in which until the decade of 80s the curricula of study is still only at of a 3-years Diploma Level. Even on the decade 80s there were some university/high education institution started to offer Sarjana Degree (a 4-years Diploma) but most of them just concern and offer the perspective of tourism economic; meanwhile actually there are at least 18 fields of knowledge and sciences should be involved in a study of ecotourism/tourism (e.g. please see Cooper *et al*, 1999).

The result of those situations are many of tourism "expert" and practitioners in Indonesia are very ready for any tourism-skill-activities, but sorry to say that most of them are still incompetent to deliberate a comprehensive and genuine development concepts. At the end, as one can see in Indonesia nowadays there are more and more of a "copying success story" ecotourism development projects competing negatively from one place to another. Further, experiences and working motives among ecotourism practitioners in Indonesia become a serious issue since most of the ecotourism project in Indonesia – that lead by NGOs-institution in the early or the middle of 90s – had to face a limited budget and discontinued-funding. This condition trapped many ecotourism practitioners to incomplete process for gaining better experiences. Even most of them eager to write a long paper of CV on ecotourism experience, but if they want to be honest, however, the reality will tell us that actually most of them are having deficient experiences; as most of their projects were only fit in to an initial-phase of ecotourism development programs.

Tour on duty, structural-functional officer-system and discontinued budget are becoming important dynamics to be discussed on talking about bureaucracy on ecotourism development on nature conservation area. For many years until now, most of decision making position on a ecotourism institution of a nature conservation area are belong to group of structural officer; who will in average be having tour on duty for every 2-3 years from one working-station to another. This situation is not only creating a short supply of command (while most of ecotourism projects belong to a mid up to a long development program), but also generate an unproductive transition periods of command. This situation become worth since most of the sub-ordinate officers on any nature conservation area are also not having enough knowledge on ecotourism field. Regarding their formal education back-ground and their yearly activities, it is strongly predicted that less than 0.1 percent of them who ever had formal study on ecotourism (even for partial subject only) or ever had comprehensive extension/training on ecotourism.

Even since the middle of 2000 the Ministry of Forestry provided such kind of regular extension programs (such as SECEM-Program; a collaboration program between the Ministry of Forestry of Republic Indonesia and KOIKA of management course on ecotourism and conservation field), however the extension program can not yet supply enough place for thousands officer. More, on the one hand, mostly extension or training program on ecotourism will be "clutched" by the upper position first – as a part of modus operandi of seniority – while on another hand the senior officer will be transferred soon to other working station due to the tour on duty system.

The scoring result on human resources capacity of nature conservation destination showed on Table 5. Even from the table one can see that many bad scores for most of the criteria,

but not for the criteria of management spirit. The high value of management-spirit was mostly supported by the field officers at each ecotourism destination on nature conservation area. After many years of working period, many of them still loyal to be a volunteer or to be a yearly contract base worker just due to their interest on nature conservation and ecotourism activities. Actually it is really a good point of human resources and should not be longer annoyed by the upper system.

Table 5. The Score of Human Resources Capacity on Ecotourism Field in Nature Conservation Area

Criteria of Human Resources Capacity	Score of Criteria
1. Formal Education	3
2. Formal Extension	3
3. Cognitive Ability	3
4. Skill Ability	4
5. Formal Experience	3.5
6. Working Capacity	4
7. Working Spirit	5.5

The situation of ecotourism management in nature conservation area also become worth since the limited budg*et al*so followed by complicated budgeting system and the administration system of governmental-finance for ecotourism development on nature conservation area. The system is also not giving a better atmosphere for running an ideal "business" of ecotourism; for example the basic rule of to pay-out a budget is really a time consuming and long procedure of administration-prerequisite. Data at the Ministry of Forestry (www.dephut.go.id) showed that the total budget on 2010 for ecotourism and environmental services for all national parks in Indonesia was only around 90 millions USD; it means only around 170-grand USD for each national park. This number of budget of course merely far from enough to fulfill and run a good development and management of ecotourism program. Further more, after many decades of "complicated" bureaucratic budgeting system applied in Indonesia, the system of multi-years project of budgeting system was just applied since few years ago in the middle of 2000s. However, this new system does not yet answer the problem and also does not running well since the political-election period is not fit to development phase.

Conclusion

Regarding all situation described above probably now one may sure to conclude that it is true Indonesia has more than abundant potential on ecotourism, not only in supply but also in demand. The most crucial problem on the development of ecotourism on nature conservation area in Indonesia is the human resources capacity; which also unsupported by a better bureaucracy system. A low capacity of human resources brings domino effect of unsatisfactory planning and its reciprocal impacts, while a complicated bureaucracy system brings trickle down impact on an awful management system.

To increase the benefit of ecotourism development on nature conservation area in Indonesia, some important starting-effort to do is not only to solve the pitfall on the human resources capacity and management system, but also to really practice the multi-discipline nature of ecotourism. It is very important to be realized that an ecotourism development is

not a kind of development on a focal point at any destination only, but actually it has to be a total development collaboration of every living-element in the region.

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Instability in Selected Malaysian Crop Production in Reference to the National Agricultural Policies

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Abstract

Instability in crop production has raised a concern in the Malaysian agriculture sector on which 15% of the population depends as their income for livelihood. Instability mainly influences farm income and production of crops. To understand the instability this study examined four major crops of Malaysian agriculture from 1984 -1997 and 1998 – 2008. This period includes the National Agricultural policies of Malaysia. The results show growth of production in oil palm, and rice was from area increase and yield increase respectively and drastic decrease in rubber and cocoa production due to less area acreage. The study identified the major factors for instability in crop production, and recommended that policy makers address instability by looking into the proper area distribution among crops and to improve the yield of crops by introducing new varieties and technologies.

Keywords: Instability, agricultural policy, Malaysia

Introduction

Agriculture in Malaysia has been improving with the growing economy. However, internally the sector has been facing structural imperfection such as land fragmentation, labor shortage and yield stagnations. As a result of which productivity and profitability are affected. Malaysian agriculture is in the process of drafting new policy for the sector for the period of 2011 and 2020 as the third national policy ended in 2010. However, the instability has become a matter of concern, as instability in agricultural and food production is important for food management and macro-economic stability (Chand and Raju, 2009). Volatility affects the farmers in their farming decisions and which in return increases the risk in farm production and effects' farmer's income and also impacts the low-income families as the instability in production will lead to instability in price of the agricultural products.

The agriculture sector of Malaysia covers a crop establishment of 9,175 hectares in 2008 with coverage of oil palm with 76.9%, rubber 5%, vegetables 3.4%, fruits 3.3 % and others 11.4% (Department of Statistic, 2010). It provides employments and livelihood for 15% of the total population. Even though there has been an improvement in the agricultural sector with the use of new technologies and policies, it was seen that there was large oscillation in farm production and income, which is affecting the farmers to stick to farming and farming decisions.

Accordingly, to improve the agricultural sector, the National Agricultural Policy (NAP) was formulated. The first NAP formulated for 1984 -1991 has made it possible for the agricultural sector of Malaysia to reach an average growth rate of 3.2 per cent per annum for the time period (Government of Malaysia, 1985). Focusing on expansionary policy on export crops like oil palm, rubber and cocoa as there were abundant land and manpower available. In absolute terms, the total value-added of the sector increased from RM11.9

billion in 1985 to RM16.2 billion in 1995(MOA, 1998a). The Second NAP (1992-1997) emphasizes on increasing productivity, efficiency and competitiveness, with further expansion of area under oil palm. The NAP3 (1998-2010) emphasized more in maximizing income through optimum utilization of resources and increasing agriculture's contribution to the GDP and with a specific objective to improve food security.

The structural changes in the economy have brought forward new issues and challenges in the agricultural sector, in particular, acute labor shortage, limited availability of suitable land and variation in yield area and production which in return negatively affected the stability and security of the country's agricultural supply. This instability and insecurity, if left unchecked, can have serious economic, social and political implications (Ministry of Agriculture, 1998b). The study identifies the changes in crop production from 1984 – 2008 and studies the instability and the reason for the crop instability.

Materials and Methods

Table 1. National agriculture policy of Malaysia

Year	**1984-1997**	**1998-2010**
Policy	NAP 1 and NAP 2	NAP3
Study Phase	I	II
Emphasis of NAP 1(1984-1991)		
• To develop the export oriented sector • Government invested heavily on: • Infrastructure • Institutional building • New land developments for oil palm and cocoa • In situ development to resolve uneconomic farm size and low productivity among smallholders		
Emphasis of NAP 2 (1992-1997)		
• Increasing productivity, efficiency and competitiveness • Increasing land areas for palm oil (plantation crop) • Development of agro-based industry		
Emphasis of NAP3 (1998-2010)		
• Increasing the competitiveness of the agricultural sector • Maximizing income through: • Optimum utilization of resources • Increasing agriculture contribution to national GDP • Increasing incomes of producers		

Source: Ministry of Agriculture (various issues)

The paper used time-series data from the Department Of Statistics on area, yield and production of selected major crops viz, rice, rubber, oil palm and cocoa. The data on area, yield and production were further split into two-phases, i.e. Phase I (1984-1997) which includes NAP1 and NAP2, and Phase II (1998-2008) that includes NAP3. The years are separated according to the implementation of the NAP's. The reason behind the selection of the NAP as a base for division is to test instability of production of crop after the implementation of the NAPs in Malaysia. NAP1 and 2 focuses on the cultivation of crops which depended mostly on traditional methods. So production growth was mostly due to

increase of the cultivated area. Whereas NAP3 emphasizes growth in crop production were mainly due to the adoption of HYVs, improved technology, improved agricultural and management practices. Table 1 shows the framework of the study with respect to the agriculture policies.

The methodology used was previously applied to study the instability of Indian agriculture by Hazell (1982). The method is an original work on the "exact covariance of products of random variables" by Bohrnstedt and Goldberger (1969). This method was also used by Ray (1983a), Ray (1983b), Mahendradev (1987) and Rao *et al*., (1988) to estimate instability in agricultural production.

The study uses coefficient of variance (CV) as a standardized measure of instability, accordingly the change in CV in period II over Period I was calculated.

$$CV = \left(\frac{6}{\mu}\right) * 100$$

CV is calculated by dividing standard deviation (6) by mean (μ) and multiplied by 100 to express it in percentage. The average production was calculated by

For entire production : $E(P) = \mu a \mu y + CV\ (a, y)$

For each period : $E(P_I) = \mu a_I \mu y_I + CV(a_I, y_I)$

$E(P_{II}) = \mu a_{II} \mu y_{II} + CV(a_{II}, y_{II})$

The Change in average production : $\Delta E(P) = E(P_{II}) - E(P_I)$

The instability was computed by disintegrating the variance of production into area, yield variance and area- yield covariance. The production variance V(P) was determine by using the formula from (Mood *et al*. 1974):

$V(P) = \mu_a^2\ V(a) + \mu_y^2\ V(y) + 2\mu_a\mu_y\ CV(a,y) - \{\ CV(a,y)\}^2$ + E $\{(a - \mu_a)^2\ (y - \mu_y)^2\} + 2\mu_y E[(a - \mu_a)^2(y\mu - \mu_y)] + 2\mu_a E[(a - \mu_a)(y - \mu_y)^2]$

Where V(a), V(y) and CV(a,y) are the variance of area and yield and μa and μy are means of area and yield respectively. Covariance of area-yield is represented by CV(a,y).

Results and Discussion

Variability in agricultural production consists of variability in area and yield and their interactions (Chand *et al*., 2008). However, variation in cultivated area and yield depends largely on climate factors, distribution and availability of inputs. The change for production of the crops of Malaysia during the entire period (1984-2008), as shown in Table 2, illustrate an increase of production for oil palm (633%) which was due to a large increase in crop area (9%) and while yield due to the introduction of improved varieties was rather constant (0.7%) as expansion of oil palm area was emphasized during the NAP2 and NAP3. Rice too showed an increase in production of 49% with an increase in area (0.19%) and yield (1.7%). There is a decrease in production for rubber (-88%) and cocoa (-97%), with a decrease in area for rubber (-8%) and cocoa (-11%), even though rubber showed an increase in yield by

0.23%. The result indicates that increase in crop area and yield increased crop production. The result indicates that as the land use is not well distributed among crops so policies for better land utilization are highly recommended. As land becomes limited, more importance should be given to increase yield through improved technology.

Table 2. Total change in area, yield and production of crops in Malaysia (1984 – 2008)

Crop	Change in mean Area (%)	Change in mean Yield (%)	Change in area – yield covariance (%)	Total change in Mean production (%)
Rice	0.19	1.70	0.03%	49.76%
Rubber	-8.47	0.23	-0.07%	-88.51%
Oil palm	9.03	0.70	-0.16%	633.09%
Cocoa	-11.78	-0.86	0.76%	-97.99%

Instability in Malaysian Crop Production

In table 3 the instability of crops between the two phases i.e. Phase I and Phase II is shown which provide a clear picture of variation between the period and among the major crops of Malaysia. As coefficient of variation (CV) is used to measure instability in this study therefore the CV of crops for each period are shown in the Table 3. Among all the crops highest instability in area, yield and production was seen in cocoa of 126%, 112% and 143% respectively followed by increased in rubber instability by 94 % for area and 64% for production where as yield instability decreased by 4%. In case of oil palm, area and production instability decreased in the II period compared to period I by 55% and 45% respectively where as yield instability increased by 55%. Out of the four major crops rice showed decrease in instability in area, yield and production by 39%, 21% and 43% respectively. Therefore it is seen that change in crop area play's the major role in increasing instability in production i.e. expansion of area for oil palm during NAP1 and NAP2 contribute to instability among the crop production.

Variance in area and yield is shown in Table 4 where it indicates that the area variance for rice is constant in both the phase while yield variance was higher in phase I (0.54) compared to phase II (0.20). The main source of instability between the phases was due to yield variance. But in case of cocoa both area variance and yield variance played major role in production variance. Area-yield covariance has significant role in production variance in case of oil palm and rubber.

Therefore the major reason of production instability is area variance. These findings indicate pressing need for instituting policies to stabilize acreage among the major crops in Malaysia. The minor yield instability can be stabilized by the introduction of better varieties and improved management practices.

Table 3. Coefficient of variation of area, yield and production for phase I and phase II

Crop	Phase I CV (%)	Phase II CV (%)	% change in CV (phase I and II)
Rice			

Area	2.77	1.67	-39.71
Yield	9.24	7.26	-21.43
Production	11.20	6.30	-43.75
Rubber			
Area	25.82	50.17	94.31
Yield	5.65	5.41	-4.25
Production	29.54	48.53	64.29
Oil palm			
Area	36.56	16.45	-55.01
Yield	3.74	5.80	55.08
Production	37.66	20.54	-45.46
Cocoa			
Area	30.64	69.32	126.24
Yield	15.21	32.33	112.56
Production	36.07	87.84	143.53

Table 4. Changes in production variance from NAP1 / NAP2 to NAP3 (1998-2008)

Crop	Phase	Area Variance (%)	Yield Variance (%)	Area –yield covariance (%)
Rice	I	0.04	0.54	0.05
	II	0.04	0.20	0.01
Rubber	I	0.18	0.23	-0.11
	II	0.53	0.40	0.03
Oil Palm	I	4.68	0.35	-0.34
	II	0.19	0.86	0.06
Cocoa	I	3.07	1.56	0.20
	II	0.80	2.27	0.21

Conclusions

This paper examined the instabilities of the major crops of Malaysian agriculture and identified the components of change in crop production. The National Agricultural Policy was adopted in 1984 as an attempt to provide guidelines to help unify and direct efforts of all involved in agricultural development (Courtenay, 1987). These efforts and policies no doubt increased crop production but also created instability as well. It was found that among the four major crops, rice and oil palm increased production while rubber and cocoa suffered production decline mainly due to variance in crop acreage. Coefficient of Variation was higher in rubber and cocoa due to instability in crop area and yield. Crop area plays a significant role in the instability of crop production of the major crops in Malaysian agriculture. As the NAP1 focused more on area expansion, oil palm production increased at the expense of cocoa and rubber in terms of area and due to better economic returns. Rice production increased marginally mainly through yield improvements compared to the other three crops.

As future land expansion is limited, more efforts should applied towards using HYV's, improved farm management practices and adoption of new technologies. Agricultural

policies should be geared towards better distribution and fixed zoning of crop acreage while increasing crop intensity so as to reduce instabilities in crop production.

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Public Perception of Food Alternatives for Rice in Bandung

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Abstract

The increasing of population geometrically and food production arithmetically become interesting issue at present and in the future. Furthermore, it will lead the world into a food crisis approximately in 2020. These conditions create demand for alternative food (non-rice food) which needs to be organized continuously. To Respond the issue, the government of West Java creates public movement such a day without rice by issuing government decree number 60 year 2010 concerning the Acceleration of Local Food Resources with directives from presidential decree number 22 year 2009 concerning Food Diversification. The purpose of this study was to determine the public perception of alternatives substitution of rice, most appreciated alternative foods and community expectations on food consumption of alternatives substitution of rice. Methods in this study uses primary data, the respondents were civil servants of government province of West Java. They were treated by consuming non rice food of their lunch in West Java Cooperative Fair 2011. The results showed that substitution alternative of rice such as cassava rice, corn rice, sorghum and rice from hanjeli seed have no different response with regular rice. The most preferred alternative food to substitute rice was cassava rice because the taste was more familiar and the shape resembled with rice. People expected to have a proper access to alternative substitution food with cheap price.

Keywords: rice, food alternatives, Presidential decree No.22 /2009, public perception

Introduction

Increasing number of population in geometrically, while food production grows in arithmetically will be an important issue regarding agro-food business nowadays and in the future. Moreover, it will bring the world into a food crisis approximately in 2020 These conditions create demand for alternative food (non-rice food) which needs to be organized continuously.

As world population increase, the food demand raise by 70%. In 2010, the world's rice requires approximately 380 million tons and will increase approximately 465 million tons in 2030 due to population growth from 6 to 8 billion people. The population reaches 9 billion by 20 years and it will take 2.1 to 3 billion tons/day of rice production. Not only by limited land resources, but also water supply is greatly reduced causing drop crop production. Currently, rice exporter country becomes net importer from neighboring countries. It is not only happen to ASEAN countries, but as well as China, Japan and Korea.

Indonesia imports rice from Thailand and Vietnam of 1.848 million tons in April 2011, equivalent to 92.51% of the government import permit which allocates about 2 million tons in the period October 2010 to March 2011. This import was due to the shifting productivity from productive land become less productive land, hence it declines the production. The dependence of rice import continues to fluctuate and tend to increase (Council of Food Security, 2006, in Sektyanu 2011). Thus saving food by way of diversity is necessary to

strengthen food security. This preliminary study will access the public perception of alternatives to rice such as rice made by cassava, corn, sorghum and hanjeli.

Materials and Methods

Objective

Research uses a case study. The objective was to analyze the perception of consumers toward good alternative to substitute rice and to identify which is the most preferred alternative by consumers. This study was conducted in the provincial government of West Java coincidence with campaign for the Movement Day without Rice (without rice on a day) which was usually held every Wednesday in the West Java provincial government. Governor of West Java has suggested to all regions in West Java to support the program. As part of the implementation, the provincial government serves food substitute to rice in West Java Expo Cooperative Fair 2011 on July 2011.

How to determine respondents

The number of respondents for this study was about 64 respondents using purposive sampling. The respondents were local government employees who attended the lunch on West Java Expo Cooperative Fair 2011 session. Data collection techniques used in the study were primary observation, interview, questioner, and literature study. Data was analyzed using descriptive analysis.

Results and Discussion

Characteristic of respondent

Most of the respondents are women (52.5%), while men respondents only 43.75%; because women was interested to know about food innovation. Age of respondents <25 years (12.5%), 26 - 35 years (25%) and >35 years (62.5%) and average-married (75%). Generally, respondents have a long working period such as >10 years (37.5%), 10-20 years (31.25%) and >20 years (31.25%). Education varies from junior high (6.25%), high school (12%), Strata 1 (75%) and Strata 2 (6.25%) and so does the income. Income <3 million (18.75%), between 3-5 million (43.75%) and >5 million (37.5%). Their income seemed enough to support his family, which the average number of family members between 3-5 people (75%) and other more than 5 people (25%).

Respondents are familiar with rice alternative foods, because the local government held a program Jabar "One Day No Rice", which its implementation every Wednesday. Therefore the local cafetaria provided cassava rice, maize rice, sorghum rice and hanjeli rice every Wednesday.

The research showed that not all respondents consumed all kinds of alternative food; only few consume 2 or 3 times. Respondent refers to someone who consumes cassava rice, maize rice, sorghum rice or hanjeli rice at least one time. Distribution of respondents is consuming cassava rice (31.35%), corn rice (28.12%), sorghum rice (18.75%) and hanjeli rice (21.87%).

Consumer perceptions to alternative food of rice

One way to analyze consumer behavior is through the perception to the products. By understanding consumer perception, industries will be able to identify the strength, weakness, opportunity and threats in the market. It rely on consumer perception as the internal factors that influence consumer in decision making. Definition of perception according to Schiffman and Kanuk (2004) is "the process by which an individual selects, organizes, and interprets stimulus into a meaningful and complete picture of their world. From this definition, perception is a process that makes a person choose, organize, and interpret stimulus that he received into a meaningful picture of their world.

The same way what Robin (2006) argued, the factors that influence a perception are people, target and situation. So perception is very subjective because it influenced by experience, expectation, motive, personality, and individual physical condition. Leavitt (in Rosyadi, 2001) distinguishes between perception into two points of view; the narrow and broad views. A narrow view perception as how a person sees something. While a broad view perception as how a person interprets something. Differ from narrow view, broad view not only just sees something but also the sense of it.

From Table 1, it can be concluded that cassava rice is the favorite food than other food and corn rice became the second option., Sorghum and hanjeli rice tendency to shift corn rice because sorghum and hanjeli rice are the unique product and sensational. Respondents prefer cassava rice because cassava is more familiar than other product such as snack or junk food.

Table 1. Frequency respondents in consuming rice alternatife food

Product	Consume		Number (%)
	1 time (%)	> 1 time (%)	
Cassava Rice	10.93	20,32	31.35
Corn Rice	15.62	12.50	28.12
Sorghum Rice	12.50	6.25	18.75
Hanjeli Rice	7.81	14.06	21.87
Total	46.87	53.13	100

The main reason consuming rice alternative food are unique (32.82%), benefits (31.25%), attractive appearance (28.12%) and taste (7.81%). According to respondents, rice alternative food (cassava rice, corn rice, sorghum rice and hanjeli rice) content fiber higher than rice. Although rice alternative food good appearance, but the color (was not as bright as rice). Hopefully, Bleching that food, do not use bleach and cultivated without the flavoring.

The research showed that all kinds of rice alternative food were delicious (73.43%), less delicious (18.75%) and not delicious (7.82%). Although the food was good, some respondents were feel it unfamiliar (25%). If they consume rice alternative food, they need more variety of food combine with fish, beef, and chicken. This is to compensate the inconvenience of consuming these foods. Actually, the diversification of these foods will increase the expenditure. The data showed that not all respondent willingness to consume

alternative food, because many respondents got health issue such stomach ache after consuming the alternative food. Reasoning willingness to consume alternative food can be seen in Table 2. .

Table 2. Reasoning willingness to consume rice alternative food

Reason / Product	Goverment Program (%)	Saving Rice (%)	Healthy Living (%)	Common (%)	Number (%)
Cassava Rice	15.62	4.68	9.37	1.56	31.35
Corn Rice	20.31	1.56	4.68	1.56	28.12
Sorghum Rice	18.75	0	0	0	18.75
Hanjeli Rice	21.87	0	0	0	21.87
Total	76.56	6.25	14.07	3.12	100

According to respondents, willingness to substitute rice with rice alternative food (cassava rice, corn rice, sorghum rice and hanjeli rice) was to support government programs (76.50%) such The Diversification of Food Consumption (PKP). Respondents were the West Java local government employees, so the answer was subjective. Hence, it is necessary to conduct the other research, so that the answer more objective

World Bank (2006) stated that food security that is intended to improve nutrition is a very profitable investment. There are at least three main reasons a country needs to implement. First of all, it has a high economy return; secondly, it supports economic growth; and thirdly, it reduces poverty through improving productivity, reduce sick days and health costs.

Indonesia's success in the food self-sufficiency in rice commodity is not always followed by a reduction in food-insecure population. Therefore, food security is a priority in Indonesia since it is not merely from the aspect of food supply through food self-sufficiency efforts, but more important is to realize food security of households to help reduction in food-insecure population (Nuhfil Hanani, 2011).

Definition of food security varies widely, but it generally refers to the definition of the World Bank (1986) and Maxwell and Smith (1992) that is "the access to all the people all the time at which enough food for healthy living (secure access at all times to sufficient food for a healthy life). Indonesia applies it in the Law of Food No. 7 1996 : food security is the fulfillment of conditions for household food security reflected in the availability of sufficient food, both in quantity and quality, safe, equitable and affordable (Yuli Hariyati and Sugeng Raharto, 2011).

According to Steven *et al* (2000), country that conducts food self-sufficiency program at the national level, still found food-insecure population due to the barriers on access and distribution of food. One way that the household families do in dealing with problem of food access is they change their quality of food they consume; reduce food portions from adults to children (Maxwell and Smith, 1992). Furthermore, the efforts on consumption of food need to focus on (1) the increase and diversification of food consumption through the provision of alternative food rice, (2) the campaign to strengthen local non food consumption of rice as a source of carbohydrates that pioneered by the local government in every meeting routine and on a larger event, (3) campaign material consumption of organic

food, (4) increase awareness and early warning of food insecurity and malnutrition, (5) mapping of food insecurity at the village level and (6) campaign consumer awareness in security food (Andy Mulyana, 2011).

"One Day No Rice" was the local government program, that expected to reduce the dependency on rice, in accordance with the mandate of this program from Presidential Decree No. 22 of 2009 on Food diversified. Indonesia actually has a diverse potential in the development of non-rice staple foods like corn, hanjeli, buckwheat, barley and various other types of Cereal. However, because so far only focused government policy on rice, then the existence of other types of other carbohydrate food sources neglected (Tati Nurmala and AEP Wawan Irwan, 2007). Hence, there is a need for alternative food instead of rice. If we consider all the citizens of West Java consume rice, one day is equivalent to the yield requirement of 3,000 hectares of rice.

The Constrain and Responden Expectation on "a day without rice" Program

The limitation for running this program were: more expensive than rice (32.81%); the material was not available (31.25%); family member dislike the product (32%); and distribution is not running well (15.62%). The response of respondents could be understood, because the raw materials of food alternatives is rarely found and sold in traditional markets and supermarkets

Alternative food has not been marketed well, because farmers rarely seek sorghum and hanjeli commodities. The farmer changed to commodity more marketable. Based on Nurmala Tati *et al*. (1998) research, there are several traditional markets that sell hanjeli in Sumedang Regency, Bandung, Garut and Purwakarta. This research emphasizes the aspects of cultivation. It was reported that West Java prospective for rice alternative food such as hanjeli, sorghum, wheat, barley and buckwhea.

If an alternative food is spread in traditional markets or supermarkets, hopefully it will be alternative form for alternative foods like grains of rice (59.37%). Respondent's expectation about the form of rice alternative food form can be found in Table 3.

Table 3. Respondent Expectation about The Form of Alternative Food to Rice

Expectation / Product	Grain of Rice (%)	Ready Half-Cooked (%)	Mix Product (%)	Others (%)	Number (%)
Cassava Rice	23.43	.12	3.12	1.56	31.35
Corn Rice	10.93	6.25	7.81	3.12	28.12
Sorghum Rice	10.93	4.68	1.56	1.56	18.75
Hanjeli Rice	14.06	3.12	1.56	3.12	21.87
Total	59.37	17.18	14.07	9.38	100

Producing ready half-cooked product, mix product or others product should involve food technology expert. The expected product, such as "Soy Joy" cracker or biscuits Melania from 521 MLD was produced from soybean powder. For introducing new product, it should be attractive appearance, tasty, practical and well distributed.

The process adoption of alternative food on rice is through five steps that is awareness, interest, evaluation, trial and adoption (Kotler, 2003). Awareness is the public aware of new foods, interest is the people interested in taking, evaluation is the people consider whether to consume, trial is the people try to consume, and adoption is the people decided to use these foods in daily.

Respondent's willingness to pay varies. If the price of rice between 6500,- – Rp. 7000,- IDR, the availability of the respondent to pay the alternative food between 5500,- – 6000,- IDR/kilogram. The ranking of expensive price, from the highest to lowest are hanjeli rice, corn rice, cassava rice and sorghum. Thus hanjeli rice sorghum and rice is no longer inferior, because of the uniqueness. Availability willingness to pay for rice alternative food can be found in Table 4

Table 4. The respondents willingness to pay for rice alternative food

Willingness to Pay / **Product**	**Rp.5.000 /Kg (%)**	**Rp. 5.500 /Kg (%)**	**Rp. 6.000 /Kg (%)**	**Number (%)**
Cassava Rice	12.50	4.68	4.68	21.87
Corn Rice	4.68	4.68	7.82	28.12
Sorghum Rice	14.06	3.14	1.57	18.75
Hanjeli Rice	3.14	15.62	23.43	31.35
Total	34.38	28.12	37.50	100

Consumer willingness to pay a premium personal or subjective, means that the characteristics of influences, hence the social factors (income, education, and employment) as well as demographic thought to affect consumer willingness to pay a premium (Grannis, *et al.* 2000; Akungor, *et.a.* 1999). Respondent Willingness to pay depend on income and purchasing power. There is a tendency that respondents dedicate only for trying, hence although purchasing power is decreased, they eager to buy. Repetition of purchase depends on the evaluation of the first purchase. As expectation, if the food is consistent, the loyal consumers will buy (Schiffman, 2004).

Conclusions

Rice Alternative food such as cassava rice, corn sorghum rice and hanjeli rice are no different than regular rice. Cassava rice is the most preferred, corn rice become the second choice. Hanjeli rice and sorghum are tend to shift the others because of the uniqueness. The main reason to consume alternative foods is to support the government program "Without Rice on a Day ".

Limitation to consume rice alternative food are the expensive price, product are not available in traditional markets and supermarkets, the distribution is not running well and the family members did not like that foods. The expectation of food alternative to rice is grain rice because they are more familiar. The ranking of expensive price, from the highest to lowest are hanjeli rice, corn rice, cassava rice and sorghum. Thus hanjeli rice sorghum and rice is no longer to be inferior, because of the uniqueness.

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Physiological Response of Tomato (*Lycopersicon esculentum L.*) to Boron Fertilizers Under the Varied Soil Lime Content

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Abstract

Boron (B) is essential for yield and quality, relatively small amounts of B are required to support the process of plant growth. Thus, varied solubility of different B sources has a great importance in order to avoid potential toxicity or deficiency of B to agricultural crops. In the present study, different B sources as boron fertilizers were tested for tomato plants to correct potential B deficiency or minimize the risk of B toxicity under the varied soil lime content which consist of most of the agricultural area in Turkey. A pot experiment was conducted based on completely randomized design using the seven different soils containing varied levels of calcium carbonate. Tomato variety of Sedir (Lycopersicon esculentum L.) was used for the present study. Five B sources, ground colemanite (2CaO.3B2O3.5H2O), borax decahidrate (Na2B4O7.10H2O), borax pentahidrate (Na2B4O7.5H2O), sodium metaborate tetrahidrate (NaBO2.4H2O), boric acid (H3BO3), were applied at the rate of 1.5 kg B ha-1 to the experimental soils. In addition, a basal dressing of some macro and micro nutrients was applied to all pots for normal plant growth. The plants were harvested after eight weeks. Dry matter yield was recorded, and total B and nitrogen concentrations in the top of plants were determined. According to the results of experiment, dry weights were significantly affected by application of B fertilizer as an average of B sources. The results clearly showed that tomato plants differently responded to the B sources having different solubility under the varied soil conditions. Boron and nitrogen nutrition of tomato plant was also significantly influenced by B sources under the varied soil conditions.

Keywords : Boron fertilizers, soil lime, tomato

Introduction

Tomato (*Lycopericon esculentum L.*) is one of the most popular vegetables in Turkey, and it has a great importance in the processing industry. Therefore, fruit quality of tomato has been a major concern for modern agriculture and crop marketing. Among the many factors limiting the yield and fruit quality of tomato, balanced nutrition is highly important for optimal yield and quality. Many studies have clearly revealed that plant nutrients such as calcium, boron had important effect on fruit quality like fruit cracking, fruit color, brix and other standard properties of tomato (Chude and Oyinlola, 2001; Davis *et al*., 2003; Heckman, 2009; Huang and Snapp, 2009). Hence, B has many physiological function such as water relations, sugar translocation, cation and anion absorption, pollen viability and the metabolism of N, P, carbohydrates and fats in the plants (Oyinlola, 2005). Boron also encourages proper nutrient balance, cell wall stability and assimilation as well as synthesizes transport (Allen and Pilbeam, 2007; Helal *et al*., 2009).

While boron (B) is essential for yield and quality, relatively small amounts of B are required to support the process of growth. Crop sensitivity to B deficiency or toxicity vary widely

depending on plant species together with soil and other environmental interactions affecting B availability and optimal plant growth (Gupta, 1993). Soils are normally considered to have sufficient levels of B when the soil analysis level is greater than 1.0 ppm. Whereas, soil extractable B levels above 1.5 ppm have the potential to cause B toxicity in sensitive crops. It has been reported that symptoms of B excess in sandy, loamy sand, loamy, and clayey soils occur when B content extracted with boiling water exceeds 0.80 mg kg^{-1}, 1.00 mg kg^{-1}, 1.20 mg kg^{-1} and 2.00 mg kg^{-1}, respectively (Robertson *et al*., 1975). Thus, considering the narrow range between optimum and toxic B concentrations, it is necessary to be careful when applying B fertilizers to the soils (Rudolf *et al*., 2008). On the other hand, B availability to plants depends on a large number of other factors (Kastori, 1990). For example, the interactive relationship and balance ratios among the essential plant nutrients is one of the important criteria for plant B nutrition. Soil pH and lime content are also the most important soil factors that affect B use eficiency. Many studies revealed that there was a significant interaction between soil pH and B availability to plants especially above pH 6.5 (Peterson and Newman, 1976; Gupta and Macleod, 1981). It has been reported that B could be adsorbed by calcium carbonate in the soils (Goldberg and Forster, 1991). Thus, the relationship between soil B level and plant B uptake can be varied depending on soil lime contents (Kızılgöz, 2009).

Hence, varied solubility of different B sources depending on varied soil conditions has a great importance in order to avoid potential toxicity or deficiency of B to agricultural crops. For example, colemanite as B source is only used for soil application, and it dissolves slowly. Borax and boric acid easily dissolve in soils, whereas pentahydrate borax dissolves more slowly than decahydrate borax and faster than anhydrous borax. Solubor dissolves more quickly than borax and boric acid. In the present study, different B sources as B fertilizer having varied solubility were tested for tomato plants to correct potential B deficiency or minimize the risk of B toxicity under the varied soil lime content, which consist of most of the agricultural area in Turkey.

Materials and Methods

A pot experiment, based on completely randomized design, was conducted using the seven different soils containing varied levels of calcium carbonate. Tomato variety of Sedir (*Lycopersicum esculentum L.*) was used for the present study. Five B sources, ground colemanite ($2CaO.3B_2O_3.5H_2O$), borax decahidrate ($Na_2B_4O7.10H_2O$), borax pentahidrate (Etibor 48 - $Na_2B_4O7.5H_2O$), sodium metaborate tetrahidrate ($NaBO_2.4H_2O$), boric acid (H_3BO_3), were applied at the rate of 1.5 kg B ha^{-1} to the experimental soils having varied lime contents. In addition, a basal dressing of some macro and micro nutrients were applied to all pots for normal plant growth. The experiment was conducted about eight weeks, and the plants were harvested after 56 days. Plants were then washed thoroughly in distilled water and dried in the oven set at 65°C., and than dry matter yield was recorded. Boron concentrations in the tops of tomato plants (shoots + leaves) were determined by the method of Azometin-H (Wolf, 1972). The composite soil samples used for the experiment were air-dried and ground to pass through a 2 mm sieve for further analysis. The extractable soil B contents were determined by ICP according to the method of Cartwright *et al*. (1983). Determinations were also made for available soil phosphorus (Olsen *et al*., 1954), soil organic matter (Walkley, 1947), saturation percent (Richards, 1954), $CaCO_3$ (Allison and Moodie, 1965), pH (Jackson, 1958) and electrical conductivity (E.C.) (Richards, 1954). Experimental data were also subjected to the definitive statistical analysis (StatMost,

1995) and MSTAT statistical program. Selected soil properties for the experimental soils were presented Table 1.

Table 1. Selected physical and chemical properties of the experimental soils

Soil properties	Soil type						
	1	2	3	4	5	6	7
Texture	SCL	SL	L	CL	CL	C	L
$CaCO_3$, %	7.8	3.6	12.8	11.8	13.6	24.0	11.7
pH, 1:2.5	7.86	7.75	7.84	7.99	7.82	7.79	7.77
EC, mmhos cm^{-1}	0.96	0.79	0.98	0.83	1.04	1.59	1.74
Organic matter, %	1.82	1.89	1.75	1.83	1.55	1.65	1.34
Available P_2O_5, kg da^{-1}	5.54	4.75	7.76	7.24	7.93	9.51	6.78
Available K_2O, kg da^{-1}	25.0	34.4	28.3	40.8	26.2	35.6	26.5
Extractable B, mg kg^{-1}	0.37	0.53	0.79	0.42	0.82	1.14	0.93

Results and Discussion

Dry Matter Yield

According to the results of experiment, dry weights of tomato plants were significantly ($P < 0.01$) affected by application of B fertilizers depending on varied soil conditions as average of B sources (Table 2). Positive effects of B fertilizers on plant growth were also found in other studies conducted with tomato plants (Oyewole and Aduayi, 1992; Chude and Oyinlola, 2001; Davis *et al*., 2003). The results clearly showed that tomato plants differently responded to the B sources having different solubility under the varied soil conditions. However, the relationship between soil B levels and dry matter yield of tomato was nonlinear under the application of different B sources (Fig. 1.). The highest dry matter yield of 8.08 gr pot^{-1} was obtained for experimental soil no. 3, having extractable B of 0.79 mg kg^{-1}, whereas the lowest dry matter yield of 6.53 gr pot^{-1} was obtained for experimental soil no. 2 having lower extractable B of 0.53 mg kg^{-1} as average of different B sources.

Table 2. Dry matter yields of tomato plants as affected by different boron fertilizers under the varied soil conditions (gr pot^{-1})

Soil type	Boron fertilizers					
	Ground colemanite	Borax decahidrate	Borax pentahidrate	Sodium m. tetrahidrate	Boric acid	Av.**
1	10.6	6.49	6.95	5.43	6.57	7.10 b
2	6.94	6.75	6.59	5.84	6.52	6.53 d
3	6.77	7.67	8.93	7.79	9.25	8.08 a
4	6.52	7.21	7.75	5.94	6.72	6.83 c
5	6.48	7.09	7.65	6.93	6.37	6.90 ab
6	6.57	9.05	7.44	5.84	6.00	6.98 ab
7	7.57	7.41	8.09	8.43	8.47	7.99 a
Av.**	7.23 b	7.38 b	7.62 a	6.60 c	7.13 c	

** $P < 0.01$

The findings also indicated that dry matter yields of tomato plants under the varied soil conditions was significantly ($P < 0.01$) varied depending on B sources. Differences in the dry matter yield based on applicated B forms were found agronomically significant as average of experimental soils. Whereas, in other studies, it has been raported that these differences based on B sources were agronomically significant (Shorrocks, 1997). The highest dry matter yield of 7.62 gr pot^{-1} was obtained by the application of borax pentahidrate, whereas the lowest dry matter yield of 6.60 gr pot^{-1} was obtained by the application of sodium metaborate tetrahidrate.

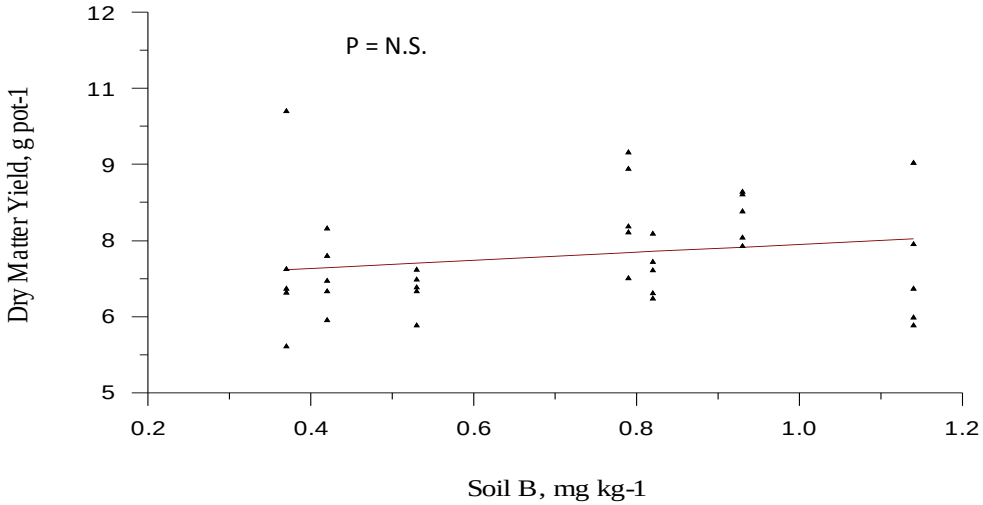

Fig. 1. Nonlinear relationship between dry matter yield of tomato plant and soil B levels under the different B sources treated to the experimental soils

Boron and Nitrogen Levels in the Tomato Plants

Boron nutiriton of tomato plant was significantly ($P < 0.01$) influenced by B sources and varied soil conditions (Table 3). The concentration of B in tomato plants was ranged from 25.85 mg kg^{-1} to 48.04 mg kg^{-1} depending on soil types, whereas it was ranged from 27.07 mg kg^{-1} to 37.00 mg kg^{-1} as to different B sources. On the other hand, the highest B concentration of 53.31 mg kg^{-1} was obtained for experimental soil no. 6 by the application of sodium metaborate tedrahidrate, whereas the lowest B concentration of 17.45 mg kg^{-1} was obtained for experimental soil no. 4 by the application of ground colemanite. It has been reported that B sufficiency levels in the dried leaf tissue of tomato plant were in the ranges of 25 to 75 mg kg^{-1} (Jones *et al.*, 1991). Soil B application also significantly ($P < 0.05$) affected total B uptake of the tomato plants (Table 4). Total B uptake of the plants was ranged from 176 µg pot^{-1} to 337 µg pot^{-1} depending on soil types, whereas it was ranged from 196 µg pot^{-1} to 266 µg pot^{-1} as to different B sources. The highest B uptake of 464 µg pot^{-1} was obtained for experimental soil no. 6 by the application of borax decahidrate, whereas the lowest B uptake of 114 µg pot^{-1} was obtained for experimental soil no. 4 by the application of ground colemanite. Positive effect of B fertilizers on the B levels of the plants was also determined in other studies (Diab, 1992; Chude and Oyinlola, 2001; Hellal, 2009).

Significant differences ($P < 0.01$) were also detected among the tomato plants as to their effectiveness in the physiological N use (Table 5). The N concentration of plants was ranged

from 2.61% to 3.19% depending on soil types, whereas it was ranged from 2.62% to 3.12% as to different B sources. Total N uptake of tomato plants was also significantly ($P < 0.05$) influenced depending on varied soil conditions (Table 6). Total N uptake of the plants was ranged from 185 mg pot^{-1} to 249 mg pot^{-1} depending on soil lime contents. Whereas, effect of different B sources on the total N uptake of plants was found non significant. Total N uptake of the plants was ranged from 191 mg pot^{-1} to 228 mg pot^{-1} depending on different B sources. The highest N uptake was obtained by the application of borax pentahidrate.

Table 3. Boron concentrations in tomato plants as affected by different boron fertilizers under the varied soil conditions (mg kg^{-1})

Soil type	Boron fertilizers					
	Ground colemanite	Borax decahidrate	Borax pentahidrate	Sodium m. tetrahidrate	Boric acid	Av.**
1	23.89 de	26.86 d	25.14 d	33.55 bc	32.25 bc	28.33 b
2	26.20 d	37.38 bc	28.32 c	32.13 c	26.15 d	30.04 b
3	23.45de	29.42 c	30.69 c	36.63 bc	41.43 ab	32.32 ab
4	17.45 f	26.42 d	26.98 d	31.53 c	26.88 d	25.85 c
5	24.09 de	33.66 bc	34.44 bc	37.43 bc	30.38 c	32.00 ab
6	41.97 ab	51.33 a	49.83 a	53.31 a	43.79 ab	48.04 a
7	32.50 bc	43.48 ab	41.29 ab	34.46 bc	33.35 bc	37.01 b
Av.**	27.07 c	35.51 b	33.81 b	37.00 a	33.46 b	

** $P < 0.01$; * $P < 0.05$

Table 4. Total B uptake of tomato plants as affected by different boron fertilizers under the varied soil conditions (µg pot^{-1})

Soil type	Boron fertilizers					
	Ground colemanite	Borax decahidrate	Borax pentahidrate	Sodium m. tetrahidrate	Boric acid	Av.*
1	240 d	174 de	175 de	182 de	212 de	196 d
2	181 de	252 d	187 de	188 e	171 de	195 d
3	159 f	225 de	274 c	287 c	383 ab	265 bc
4	114 fg	192 de	209 de	186 de	180 de	176 de
5	156 de	239 d	263 c	259 d	193 de	221 c
6	276 c	464 a	371 ab	311 bc	263 c	337 a
7	246 d	321 bc	333 bc	291 bc	282c	295 b
Av.*	196 c	266 a	259 a	243 ab	240 b	

* $P < 0.05$

The results also indicated highly significant positive correlation ($r = 0.774^{**}$, $P < 0.01$) between the B and N concentrations in tomato plant (Fig. 2.). The interactive relationship between B and N levels can be explained by the integration in nutrient uptake and balance in the presence of sufficient concentrations of these nutrients (Shaaban and El-fouly, 2001). Boron encourages proper nutrient balance and cell wall stability together with many assimilation and synthesis progress (Allen and Pilbeam, 2007; Helal *et al*., 2009). The positive relationship between B and the concentrations of B, N, P and K in wheat plants was also found by Diab (1992). The relationships between B and N levels were also determined in other studies conducted with varied plants (Carnacho and Gonzales, 1999; Hellal *et al*., 2009).

Table 5. Nitrogen concentrations in tomato plants as affected by different boron fertilizers under the varied soil conditions (%)

Soil type	Boron fertilizers					
	Ground colemanite	Borax decahidrate	Borax pentahidrate	Sodium m. tetrahidrate	Boric acid	Av.**
1	2.58	2.54	2.40	2.86	2.70	2.61 c
2	2.41	3.17	3.05	3.06	2.86	2.91 ab
3	2.84	2.94	3.08	3.17	3.28	3.06 a
4	2.25	2.57	2.65	3.09	2.70	2.65 c
5	2.41	2.97	3.03	3.10	2.72	2.84 ab
6	2.96	3.16	3.42	3.39	3.04	3.19 a
7	2.89	3.17	3.22	3.18	2.86	3.06 a
Av.**	2.62 c	2.93 b	2.98 b	3.12 a	2.88 c	

** P < 0.01

Table 6. Total N uptake of tomato plants as affected by different boron fertilizers under the varied soil conditions (mg pot^{-1})

Soil type	Boron fertilizers					
	Ground colemanite	Borax decahidrate	Borax pentahidrate	Sodium m. tetrahidrate	Boric acid	Av.*
1	260	163	167	157	180	185 c
2	167	213	203	180	187	190 c
3	193	227	277	247	303	249 a
4	147	183	207	180	183	180 c
5	160	210	230	217	170	197 c
6	193	287	253	200	183	223 b
7	220	233	260	273	243	246 a
Av.	191	217	228	208	207	

* P < 0.05

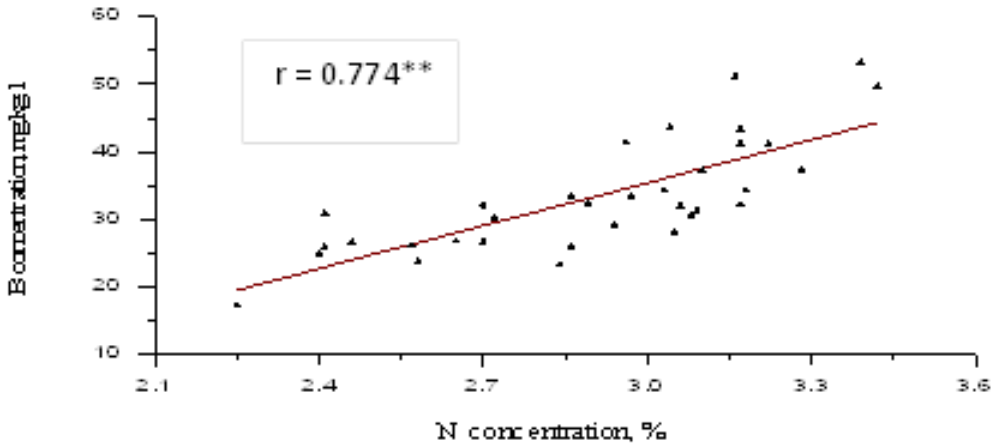

Fig. 2. Interactive relationship between B and N concentrations in tomato plant under the different B sources treated to the experimental soils

Conclusion

Tomato plants exhibited a wide range of beneficial responses to different B sources under the varied soil lime contents from low to high levels. The relationship between soil B levels and dry matter yields of the tomato plant was nonlinear under the experimental conditions. The variability of plant respons to the B sources was possibility associated with the different solubility of B sources especially depending on soil lime contents. On the other hand, the findings clearly revealed that there was a significant interactive relationship between B and N concentrations in tomato plants. This positive relationship can be explained by the integration in nutrient uptake and balance in the presence of sufficient concentrations of these nutrients. Thus, positive relationship among these nutrients will have also a positive effect on the plant growth. As a result, the experimental findings showed that different B sources should be tested and calibrated, especially for varied soil pH and lime contents. Well-understanding of solubility of B sources and interactive relationships among the plant nutrients under the varied soil conditions will also significantly contribute to correct potential B deficiency or minimize the risk of B toxicity to the agricultural plants.

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Evaluation of Food Safety Concept in Indonesian Food Security Policies

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Bandung, Indonesia

Abstract

Indonesia as an agriculture-based country, rich with natural resources, and has a good potential to manage her resources and should be able to provide sufficient food to consume. Moreover, since food is one of human basic needs, everyone deserved to obtain safety foods. Food security is an ability to provide sufficient food with respect to its quantity and quality, safety, availability and distribution. Hitherto, food security program is focusing more on quantity aspect. In other words, if food consumption needs have already fulfilled, then it considered that food security has already achieved. Food security related to the eligibility of food consumption not only sufficient in quantity, but also in its quality. The objectives of this paper are (1) to identify how food safety concept integrated into food security programs in Indonesia (2) the sources that caused food borne diseases and (3) efforts carried out in order to achieve sustainable food safety, and (4) to identify involved institutions in supporting and ensuring food safety in Indonesia. According to this study, even though Indonesia has achieved self-sufficiency on food in 1984, many poor people have low nutrient status. Moreover, the number of intoxication of food cases has been increasing. Based on the socio economic aspects, as people cannot afford to consume healthy food will lead to increase the prevalence of food borne diseases, such as diarrhea. Number of death caused by diarrhea was increased. Based on food case field studies, food borne diseases were caused by catering and household foods, processing, and junk foods. Finally, food safety concept has not good integrated into food security program. In addition, several life styles, such as consumption of junk foods resulted in food borne diseases. Therefore, the use of HACCP safety standards and organic farming methods are some of the efforts that need to be encouraged to achieve sustainable food safety. Similarly, several institutions have involved in ensuring food safety in Indonesia, but their preferences need to be improved.

Keywords: food safety, food security

Introduction

Food is one of the basic human needs to survive, and then food problems are very important issues. Providing enough food can make a country get a positive appreciation but instead can also be a serious problem if food needs are not fulfilled. Food security program in Indonesia has been reinforced in Food Act 7 of 1996 about food and Government Regulation number 68 of 2002 about Food Security.

Food security is an ability to provide sufficient food in quantity and quality, safe, available and well distributed. Moreover, due to food is one of the human basic needs, everyone deserve to obtain safe foods. From the definition, there are two important parts, namely adequate in terms of quantity and adequate in terms of quality. According to the National *Widyakarya* of Food and Nutrition in 2000, the normal human food needs are 2500 kcal/capita/day and 55 grams of protein/capita/day. From the food quality side, there are

two aspects supporting food safety, namely nutrient content quality standard and food quality standards from contaminant materials that can bring about diseases *(food borne diseases).*

Discussion

Although definition and paradigm of food security in the world has been discussed extensively, especially since 1943 in the Conference of Food and Agriculture issuing "secure, adequate and suitable supply of food for everyone" concept, in the national level, definition of food security was in debate particularly during 1970s and 1980s period. The debate on food autonomy (which means food self-sufficiency) and comparative advantage production view, which deal with the choice of, produce or import food, were main concerns in Indonesian food policies development. However, the government in the new order era has decided to reach food self-sufficiency in order to reduce food dependency from other food producing countries. As a result, Indonesia obtained food self-sufficiency officially in 1984.

Briefly, the development of food security policy, with its accompanying attributes can be seen from the following figure.

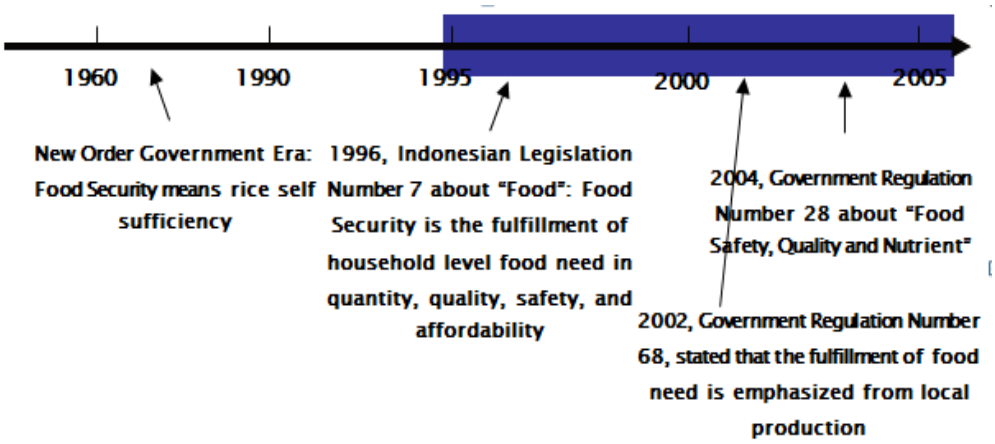

Figure 1. Development of Food Safety Concept in Indonesia's Food Security PolicyThe striking milestone in food security policy is a release of Indonesian Legislation Number 7/1996 about Food. This legislation stated, "Food security is the fulfillment of household level food need in quantity, quality, safety, and affordability". Since the time, food security has been developed based on household level that emphasizes human development, instead of production aspects. Furthermore, the legislation mentioned that food security is orchestrated responsible of government and society.

In 2002, the government issued Indonesian Regulation Number 68/2002 about Food Security. The regulation stated that the fulfillment of food need gives stress on local production. The legalization of this regulation indicated that food reserve system development was determined conforming national concern (as food producer country) than relies on other countries' interest. According to this legislation, society has a large opportunity to generate food security in several activities, such as production, trading, distribution and consumption. In order to control and evaluate national food security, the

government established Food Security Board (*Dewan Ketahanan Pangan*) over Indonesia region. Hitherto, 30 Food Security Boards have been formed at province level and 336 at district level. The national board is responsible for several activities, namely: (a) develop national food security policy, such as national food security roadmap, (b) coordinate strategic policy development (import policy, subsidy, price, food reserve and "food for the poor" program), (c) coordinate food and nutrient vulnerability treatment, (d) consolidate data and information (production, consumption, export, import, food stock, price), and (e) develop commitment among provincial and district food security boards.

Since 2004, policy on food security gave more emphasis on food quality, safety and nutrient considerations, which was signified by the issue of Government Regulation Number 28/2004 about Food Safety, Quality and Nutrient. This regulation explicitly contains food safety principles, such as standards and other stipulations to prevent food from danger, physically, biologically, and chemically. Another important substance of this regulation is an obligatory for food industry (particularly big scale industries) to have food certification in order to ensure food from dangerous contents. The important of food nutrient is also underlined in this regulation including food fortification and national nutrient sufficiency standard. In relation to food safety issue, this regulation can be mentioned as an important milestone.

The government has made various efforts to increase productivity of food crops. Program on food consumption patterns is also recommended for people with non-rice consumption patterns, which is diversification of food products to reduce consumption of rice. Unfortunately, food security programs that have been promoted both by the government and non-government agencies were identified with programs to minimize consumption of rice as a staple food for most of the Indonesian population. Food security program still focused on quantity aspect, while food security program should be related to eligible food consumption that is not only in sufficient in quantity, but also in quality.

Food security conditions and efforts are needed to prevent food from possible contamination of biological, chemical and physical objects that can danger human life. Basically food safety program will be more easily applied in the community if the community understand and realize the nature of food security and its impact on public health daily. This condition can be fulfilled if the motivation of people to get better lives and support from the government in the form of socialization programs that refers to food safety programs.

Food Availability (Food Quantity and Quality)

From the availability of food quantity, aspect that need to be considered is the level of availability of food crops for the community, both from the aspect of production and productivity of food crops. Food production level that describes the condition of food availability in Indonesia is presented in Figure 2.

Figure 2 shows the production increase of some food commodities, namely paddy, corn, cassava, soybeans, sugar, palm oil and meat. Seven commodities, which are main foods, were more available to the public.

Statistical Centre Agency of Indonesia (2009) informed that in the last ten years the productivity of food crops such as paddy, corn and soybean had steadily increased. While other food crops such as peanut, green bean and sweet potatoes were not significantly

increase. However, the opposite has occurred in the harvested area of crops. Food security conditions in Indonesia in quantity aspect can be examined by the comparison of food production (especially paddy and rice) with the need for food. The needs for rice in the community from year to year is increase based on the increase in population. However, in Indonesia this was followed by the increase in the quantity of rice, therefore the availability of rice can fulfill the human needs. Suryana (2009) informed that in 2008 Indonesia had a surplus of rice which mean that Indonesia did not necessary to import rice anymore. This condition shows that in quantity aspect, Indonesia has achieved food self-sufficiency.

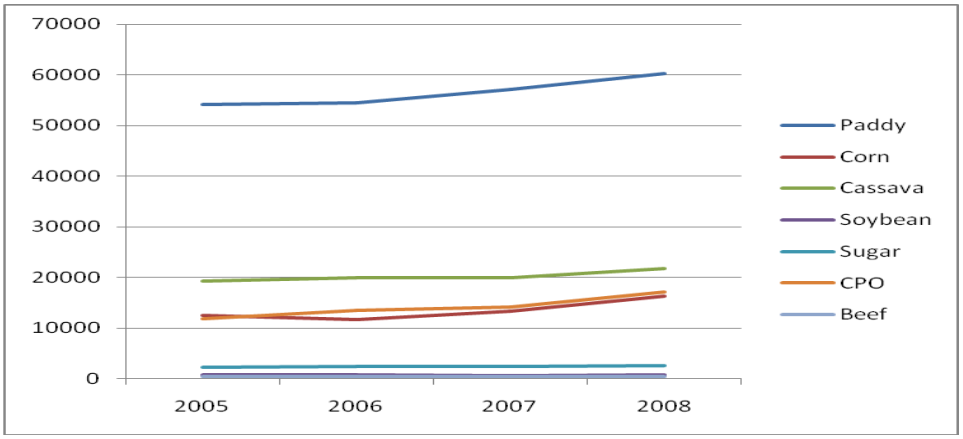

Figure 2. Food production in Indonesia (2005 - 2008), source : BPS RI, 2009 The increasing of population resulted in the increase in human food needs. Increasing food consumption needs force the government and society to prioritize the fulfillment of the food needs. Nowadays, issues of food quality are still regarded as being secondary, whereas in the previous years there was concern from the government through the government's recommendations regarding "4 *sehat* (healthy) - 5 *sempurna* (perfect)". However in the recent years, it is not a major concern anymore.

Furthermore, even for kinds of food commodities that looked healthy, some hazardous cases could happen whenever these commodities consumed. The results of laboratory test conducted and informed by Indonesian Food Security Agency (Badan Ketahanan Pangan), 2011, had specifically mentioned that from the evaluation among 11 provinces, it was found hazardous pesticide residu in some commodities.

Table 1. Residual Pesticide on Local Vegetables and Fruits at the Provincial Level in Indonesia

No	Province	Commodity	Types of Residues that are Prohibited	Residue Levels (ppm)
1	Nangroe Aceh Darussalam (NAD)	Cauliflower	Lindan	0,007
2	North Sumatera	Banana	Heptachlor	0,0090
3	Jambi	String bean	Dieldrin	0,0781
4	South Sumatera	String bean	DDT	0,054

5	Middle Java	Tomato	Dieldrin	0,0090

Kimia Agro (Chemical Agro), as the institution under West Java – Governmental Department of Food Agriculture for the year of 2010 reported that residual pesticide found on some vegetables in this province is presented on the following Table 2.

Table 2. Residual Pesticide on Local Vegetables and Fruits at the District level in West Java

No	Commodity	Sample of Origin	Analysis Results (mg/kg)
1	Green Petial Petsai	Cicendo, Bandung	0,28802
2	Red Pepper	Pacet, Cianjur	1,59125
3	Hot Red Pepper	Pacet, Cianjur	0,67074
4	Beans	Pacet, Cianjur	0,56279
5	Petsai (white cabbage)	Pacet, Cianjur	0,08738
6	Hot pepper	Pacet, Cianjur	0,58066
7	Tomato	Pacet, Cianjur	0,53708
8	Spinach	Pacet, Cianjur	0,00166
9	Shallot	Jati Tujuh, Majalengka	0,00100
10	Salad	Lembang	0,00216
11	Brocoli	Cibodas, Lembang	0,13861
12	Kaelan	Pacet, Cianjur	0,26007
13	Pea	Pacet, Cianjur	0,00104
14	Carrot	Cibodas, Lembang	0,01889
15	Raddish	Cibodas, Lembang	0,02791

Source : Chemical Agro, 2010

Other information had also been reported by Setyabudi (2008) related to residual pesticide that was found on Orange fruit in Bogor region. From kinds of orange what so called Lokan and Mandarin Markot that were sold in fruit shops, it was found that residual pesticide of Klorpirifos was 0.016 and 0.009 respectively.

Food security becomes a subject that needs serious and sustained attention because besides being a source of energy and nutrients, food also can be a source of contamination and disease. Unhealthy food can lead to various diseases such as food intoxication and diarrhea.

Based on food intoxication outbreaks reported in 2005, commonly patient has the same complaint, namely diarrhea, abdominal pain, dizziness, and vomiting with no bleeding and only some patients accompanied with fever. According to the investigations, it was found that some cases of food intoxication caused by positive S*taphylococcus aureus, Vibrio cholera, E. Coli*, histamine, fungus, and *Salmonella* (Ministry of Health, 2005). The development of food intoxication in Indonesia from 2000 to 2007 is presented in Table 3.

Table 3 shows that the level of food intoxication in Indonesia is fluctuate. This indicates that food consumed is still in unsafe conditions. Even in the year 2006, the number of food intoxication cases reached 10980 cases and 277 people died. Therefore, efforts to socialize food safety must be carried out more intensively.

Table 3. The development of food intoxication in Indonesia from 2000 to 2007

Year	Number of extraordinary hazard	Number cases	Death
2000	16	5680	109
2001	12	4428	100
2002	15	5789	94
2003	22	4622	128
2004	16	3314	53
2005	12	5051	127
2006	16	10980	277
2007	8	3661	46

Source : Indonesian health profile 2007, 2008

Food intoxication can be caused by various factors, both from households and from the environment. The study of food cases (Ministry of Trade Republic of Indonesia, 2007), found that four major issues related to consumer safety of foods consumed are:

1. Food intoxication can occur because of damaged and contaminated foods, or mix with hazardous materials
2. The use of illicit substances including preservative materials, coloring agents, sweeteners and other additional materials.
3. Conditions of labels for industrial products, food and beverages that are not in accordance with the provisions of food labels and advertisements (PP 69 of 1999), and are not follow the law from the ministry of health.
4. Products of food and beverage industry are expired.

From the data of the percentage of food intoxication sources in Indonesia (Source : PPM & PL, 2002) , foods from catering and households are the largest sources causing food intoxication. Foods from catering become the main sources of food intoxication caused by a plenty amount of food cooked through heating process to be served for many people. Processing food, heating in high temperatures for a long time will bring about heat resistant bacteria during the cooking process, which will form spores during the cooling process. The bacteria will germinate spores when food is cooling slowly to reach a safe temperature (4 ^{o} C or lower). The slow cooling process plays an important part in the formation of spores during the cooling process. From catering and household foods, processed food also gives considerable impact in the cause of food intoxication. This caused many processed foods contain chemicals such as preservatives, colorings, sweeteners and additional materials.

Diarrhea can cause malnutrition through the mechanism of fluid loss (dehydration) and electrolyte imbalance in body fluids during diarrhea. Diarrhea also affects the absorption of nutrients/mal-absorption, which can cause the body's nutritional deficiencies and growth disorders (Anonimous, 2005). Indonesian nutrition status, aged 0 to 59 months are presented in Table 4.

During the 4-years period from 1998 to 2001, the number of infants' malnutrition was decrease and the number of infants' good nutrition was increase. However, since 2002, this situation is turn around, infants malnutrition rates increased and infants' good nutrition rates decreased. This shows the nutrition status in Indonesia was decline, so that

malnutrition cases, especially for infants should get serious attention. The causes of infants and toddlers death in Indonesia are presented in Table 5.

Table 4. Toddlers (0-59 Months) the nutritional status, Year 1998-2005 (%)

Nutrition Status	1998	1999	2000	2001	2002	2003	2005
Malnutrition	10.51	8.11	7.53	6.30	7.47	8.55	8.80
Poor nutrition	19.00	18.25	17.13	19.80	18.35	19.62	19.24
Good Nutrition	67.33	69.06	72.09	71.10	71.88	69.59	68.48
More Nutrition	3.15	4.58	3.25	2.70	2.30	2.24	3.48

Source : BPS RI, 2009

Table 5. Causes of Infant and toddler death in Indonesia (2005)

No	Cause Death	Infants (%)	Toddler (%)
1	Pneumonia	22.3	23.6
2	Diarrhea	9.1	15.3
3	Severe infections	10.6	15.1
4	Other problems (including accidents)	5.5	14.7
5	Neonatal	44.5	11.2
6	Typhoid	1.2	3.8
7	Malnutrition and BGM	1.7	3.6
8	Malaria	0.8	2.9
9	Measles-complications	0.8	2.9
10	Vomiting, dehydration	1.3	1.6
11	Pertussis	0.3	0.2
12	Without cause	0.6	0.05

Source: Ditjen PP-PL, Depkes RI

From the percentage of infant and toddler mortality in Indonesia, diarrhea was the fourth main cause after neonatal, pneumonia and severe infection. However, at the toddler ages, diarrhea became the second cause of death after pneumonia. This shows diarrhea can be potential causes of death in infant and toddler ages. The information from Indonesia health profile 2007 indicated that explotion of death by diarrhea was occured on 2006.

The Sources of Main Hazards of Food Insecurity

According to Zaenab (2005) the factors that cause the occurrence of food intoxication cases are related to:

1. Six hygiene principles of sanitation and food processing which includes food selection (raw food ingredients (fresh), processed foods (plant) and ready to consume foods, food storage, food processing, storage of cooked food (container, temperature and waiting time), transporting food and serving food

2. The existence of microorganisms causes food intoxication such as the type of vibrio infection, Parahaemoliticus, Salmonella spp, E. Parahaemoliticus, Salmonella spp, E. Coli pathogen, and others, and the type caused by aereus stphylococcus intoxication, Clostridium botulinum, Aereus bacillus and Clostridium perfringes.

According to POM RI (2006), diseases can be caused by unsafe foods sourced from various hazards, such as biological, chemical and physical hazards. Biological hazards originate from microbes (viruses, parasites, molds and bacteria), farm animals, pets and rodents (mouse, insects, flies and cockroaches). Chemical hazards derived from natural toxins (palotoksin, amatoksin, poison mushrooms, toxic HCN in cassava, jengkolat acid in jengkol, toxins in fish tetrodotoksin wrapping); chemical contamination (paint solvents, pesticides, paint, chemical components of the equipment or packaging of food); and heavy metals (mercury, lead, cadmium, arsenic, copper, zinc and tin, from contaminated water, from cooking utensils/packaging containing hazardous metals and experienced surface erosion, from textile dyes, from the air contaminated by gas and dust, vehicle exhaust). While the physical hazards comes from the piece cans, glass, twigs or wood and plastic, hair, nails, jewelry and pieces of stone or gravel.

Apriyantono (2002) revealed many protein of food denatured when heated in a moderate temperature (60-90 ^{o} C) for one hour or less. Denaturation of proteins can lead to excessive insolubilization that may affect the functional properties of proteins depend on its solubility which can result in off-flavor, rancid, changes in texture, and color changes during storage of foodstuffs. Similarly, baby food processing that is usually based milk is rich in lysine and reducing sugar lactose must be observed. If the process is not well controlled, the number of lysine residues will become unavailable for reaction with lactose. As a result Protein Efficiency Ratio (PER) of baby food will be decline.

Hariyadi (2009) explained that generally Indonesian people prefer most processed foods cooked with high temperature for a long time. It could be the highest possibility of food intoxication caused by bacteria that form heat resistant spores during cooking. These causes mostly do not occur in households with small number of cooking. A small number of food cooking can faster to be cooled. Indonesian people habit that storing food at room temperature and lack of cooling facilities growing rapidly causing the return of these spore-forming bacteria. From the various cases above obtained information that the food processing must be get serious attention because the treatment should provide a nutritious food but if the food processing, especially with high heating and cooling is not balanced will be able to produce toxic compounds, which will contribute to the food becomes unsafe.

Borgdorff *et al* (2005) explained transmitted diseases caused by food derived from biological hazards of microbiological agents such as *Bacillus cereus, Cryptosporidium, Eschericia coli* O157: H7, *Salmonella enterica* serotype *paratyphi B, Salmonella typhimurium, Shigella sonnei, and Vibrio cholera.* Research results of Harlia *et al* (2001) conducted in Kosambi market showed that chicken liver proved contain heavy metals cadmium, therefore it should not be consumed by infants and pregnant women. Considering long-term effects, people can consume liver, but not for every day consumption.

Research from Balia *et al* (2004) concluded that food security status of livestock, based on content of heavy metals, has been contaminating food or organs of livestock. This was because in many processed poultry products in Bandung area, such as fresh milk, beef meatballs, lamb kidney, chicken liver and pork liver have heavy metal content of lead (Pb). Similarly, the content of heavy metals cadmium (Cd) in fresh milk and milk pasteurization are also founded. Even though the content of Pb and Cd found in livestock products was still within the allowable limit (smaller than the Maximum Residue Limit (BMR) determined by the Director General of POM in 1998, which refers to the provisions of the FAO (2.000 ppm for Pb) and Cd limits by *The Environmental Protection Agencies (EPA)* is 0.0100 ppm in

1985), it is need to remember to maintain equipment that also contribute to Cd pollution. Although the content of Cd was very small but it extremely dangerous in the long term. Cd will be accumulated in the human body (Darmono, 1995). In this study it was also revealed that the content of Cd on beef meatballs have exceeded the BMR.

Research conducted by Rahman (2006) shows that results of analysis of heavy metal content of Pb and Cd in crustaceans (shrimp and small crab) on two shores (Takisung and Batakan) has exceeded the normal threshold that is defined by FAO. Therefore, contamination of Pb and Cd in the two shores was reasonably high in the of crustaceans (shrimp and crab) organism's body.

The research by Panggabean *et al* (2008) showed that Pb contaminated local beef innards more than imported beefs did. This research also found that 32 of 146 active samples (sampling from the traditional markets, supermarkets, distributors and importers) and 10 samples of passive activities (from direct customer in Jakarta) contain Pb (lead) content that exceeds BMR consisting of 11 samples (34.4%) of imported innards and 21 samples (65.6%) of local innards. Another research from Research Winiati and Rosaria (2008) showed that red chili paste samples stored in plastic containers and food additives use (Rhodamin B colorant and sodium benzoate) positive for E. Coli.

Schumann (2001) revealed the most recent and best controlled studies, however, support the hypothesis that iron stores were related to cardiovascular risk. Amplified iron oxidative stress may also increase DNA damage, oxidative activation of precancerogens and support tumor cell growth. This was supported by experimental, clinical and epidemiological observations. Due to these mechanisms high iron stores may present a health hazard.

All preceding cases describe food security status in Indonesia. Many foods are not suitable for consumption that could be originated from raw materials, processed foods and ready meals. Moreover, many cases in several regions in Indonesia showed that particular foods derived from cattle were classified as unsafe for consumption, and then efforts to improve food safety should be improved. The above presentation also shows the factors that engender food insecurity is derived from environmental factors (food treatment), which causes the emergence of microorganisms create food intoxication. Microorganisms can arise from raw materials, storage to processing activities.

Conclusions

- In short, food safety concept was not fully integrated yet into food security practices. Secondly, several life styles, such as consumption of snacks/junk food has resulted in food borne diseases.
- In order to ensure food safety, sanitary standards that provide an important means of protecting human health from unsafe food must be implemented.
- The development of organic farming and HACCP safety standards that was carried out by Chemical Agro and Food Safety Division under the Food Security Agency at the Provincial level can be other alternatives in ensuring healthy and safe foods.

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Socio-economic Interfaces of African Indigenous Vegetables in a Subsistence Economy and the Implication for Food Security in Western Kenya

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Abstract

African indigenous vegetables (AIVs) play a crucial role in food nutrition and general livelihoods for both rural and urban populations in Africa. The vegetable production contributes substantially to household income and general household subsistence. Erratic weather conditions coupled with land pressure continues to inhibit supply of the vegetables. Increasing awareness of the nutritional and medicinal value of the indigenous vegetables has also triggered unequalled demand. Socio economic factors influencing AIV production among smallholder farmers were investigated. Employing a systematic random sampling, a total of 240 households were interviewed by a questionnaire. Cross tabulations, multiple regressions and correlation analyses were used to analyse the data. Majority of farmers use non-certified local market seeds and organic fertilizers. Agro ecological zone of the farmer, gender, and scale of production significantly influenced the size of land allocated to AIVs. Other significant factors included; education level of the head of household, access to water for irrigation and extension services. Production of maize (staple food) and livestock compete with AIVs for limited land resource. Remarkably, the choice of seeds and the incidences of pests and diseases were positively correlated. The age, education level, scale of production and off farm income also interrelate to influence allocation of resources and choice of technology in production of AIVs. Farmers in Western Kenya are smallholders relying mainly on farming as a source of livelihood where socio economic factors dramatically interact to influence significantly the decision they make in the production of the AIVs. It is recommended that to encourage increased production and productivity of African indigenous vegetables, farmers should be supported to adopt improved seeds and irrigation technologies. In addition the Government and NGOs should provide tailored agricultural information services to farmers in Western Kenya.

Keywords: African indigenous vegetables, subsistence economy, Western Kenya

Introduction

The debate as to whether African Indigenous Vegetables could break the jinx of food insecurity in Africa lingers. African indigenous vegetables have a potential to reduce the food shortage in Sub-Saharan Africa (Alistair, 2006 & NRCNA, 2006). Africa as a region is home to hundreds of indigenous vegetables that have fed Africans for tens of thousands of years. These plants are resilient enough and can thrive even in poor soil and well-suited to the small plots and resource constrained rural population. Over the years these species have received little or no attention from the research community. However, despite increasing research interests in the recent past, the vegetables are still overlooked by many scientists, donor community and policy makers in and outside the region. Efforts to explore

the potential of such vegetables could lead to enhanced agricultural productivity, more-stable food supplies, and higher incomes in rural areas across the continent.

It is documented that, historically Africans depended mainly on traditional food plants until about five centuries ago, when adventurers and slavers sailing the western seaboard introduced a collection of American crops. These crops included maize, cassava, groundnut, sweet potato, tomato, common bean, chili peppers, and pumpkin. During the subsequent centuries the crops spread across Africa as more farmers integrated them into their traditional livelihood strategies. The last two centuries, during the colonial era, indigenous crops further suffered neglect with the colonial policies advocating for a shift to familiar crops of mercantile interest, such as tea, cane, cocoa, coffee, cotton, among other crops. These crops were considered valuable in the target markets. Consequently, African indigenous crop were neglected because they were considered "poor people's plants." These subsistence crops were almost entirely ignored in organized agriculture. The valuable exportable cash crops were cultured, harvested, graded, and protected against rodents, insects, and decay with exceptional effectiveness and dispatch. Like grains and fruits, the historical discrimination did not spare Africa's ancient vegetables. It is noted that long ago, hundreds of leaves, roots, tubers, corms, rhizomes, bulbs, seeds, buds, shoots, stems, pods, or flowers were eaten. However across Africa today the main vegetables are crops such as kales, sweet potato, cooking banana (plantain), cassava, peanut, common bean, peppers, eggplant, and cucumber where majority come from a mere 20 or so species with almost all of foreign extraction. Despite the focus on exotic vegetables, peasant crops, grown by poor people have been found to be robust, productive, self-reliant, and useful with great potential to food security and nutrition (NRCNA, 2006).

Climatic variations in the recent past have posed serious challenges in growing of traditional conventional food crops such as, maize beans and wheat (GoK 2010). Decreasing productivity of traditional crops, coupled with increasing population pressure impedes efforts to overcome food insecurity in Kenya. There is an urgent need to establish and develop alternative and new sources of food that could cope with the challenge. Research and policy has now shifted focus to the cultivation of Indigenous crops. These crops have been dubbed "emerging crops" or "neglected crops" or "orphaned crops". The Government of Kenya, in the bid to tap the potential of these crops, has since developed a policy on emerging crops.

The objective of this study was to investigate socio economic factors influencing African Indigenous Vegetables among smallholder farmers in Western Kenya. The paper is divided into four sections; section one presents the background information on African Indigenous Vegetables, section two shows the methodology that was followed in conducting the study. While section three presents the results and discussions, section four give the conclusions and recommendations.

Materials and Methods

Theoretical Model

Microeconomic theories of the households attempt to capture the intricate structure of household and its distinctive behaviour. According to Mattila-Wiro (1999) information on demographic structure, decision making process, resource allocation, income earning mechanisms and gender division of lobour is prerequisite for understanding the effects of

public and private sector interventions at the micro level as well as their macro level consequences. Classical economic models assume that household is a rationally behaving unit and that the households attach value of consumption and production of goods as determined by market mechanisms. The view of households in traditional consumer theory is twofold; as a consumer and producer. Akin to a competitive firm in a market economy; households also face production possibility frontier, limited resources with alternative uses and fixed technologies. In a semi-subsistence economy household land is a major constraint and household decision in allocation of the resource is influenced by several demographic and external factors.

Specification of Empirical model

Land ratio allocated to African indigenous vegetables is used as a proxy for the decision to produce the vegetable. The ratio of land to various enterprises is also entered as independent variables to capture how the decision of household in allocating resource to other enterprises influences the decision of AIVs.

Focusing on factors influencing allocation of land resource to alternative farm enterprises, multiple regression analysis is specified as follows:

$$Y = \alpha + \beta_i X_i$$

Where:
Y = Percentage of land size in acres allocated to African indigenous vegetables

X_i = Socioeconomic variables influencing the size of land

α = is the constant and β_i are the coefficients where the sing could be either negative or positive for all variables.

Specification of the Variables used in the Regression Model

X_1 =Agro Ecological Zone

X_2 =Age of the Head of Household

X_3 =Education Level of the Head of Household

X_4 =Off Farm Income

X_5 =Distance to the Nearest Market

X_6 =Gender of the Head of Household

X_7 =Employment of the Head of Household

X_8 = Overall Land Size

X_9 =Fraction of Land Size allocated to Exotic Vegetables

X_{10} =Fraction of Land Size Allocated to Maize

X_{11} = Fraction of Land Size Allocated to Livestock

X_{12} = Extension Services Received

X_{13} = Access to Water for Irrigation

Data Collection and Analysis

The data was collected from a population of farmers in western Kenya. A household survey of small holder farmers in Keiyo North, Lugari and Central Bungoma districts of western part of Kenya was conducted during the month of July, 2010. Questionnaires were used to collect the primary data. The choice of the district was based on three major agro ecological zones of High, Medium and Low altitudes in Keiyo, Lugari and Bungoma districts respectively. The actual households were sampled out from these areas for survey.

Sampling process started by identifying and naming all the provincial boundaries (divisions and locations) within each district as units in the population from which to obtain the sample. From the sampling frame, the actual units of sampling were picked for the final sample using systematic random sampling where every fourth household was picked. Since in statistics a sample size>30 approximates normal distribution where inference can be statistically drawn, a sample size of 30 from each of the locations in the three districts was picked. A total of 240 farm households were interviewed.

Data was analysed using SPSS V17.0. Descriptive statistics was used mainly to present exiting relationships between variables. Cross tabulations, charts and tables were used to summarize the results. Regression and correlation analyses were used to obtain specific relationships.

Results and Discussions

General Farmer Characteristics

Majority of the farmers in the three districts are mainly subsistence smallholders with some few farmers renting out their farms to other farmers while others rent from other farmers. Most of the respondents interviewed in the three districts were heads of households (66 percent) implying the reliability of the household information obtained. Table 1 below presents descriptive statistics of mean farm size, market distance and the size of land rented.

Table 1. Descriptive statistics

Variable	N	Minimum	Maximum	Mean	Std. Deviation
Market Distance(Km)	239	0	65	12.30	13.537
Farm Size(hectares)	239	0	14.8	1.52	1.8588
Size Of Land Rented (Hectares)	240	0	2	0.06	0.2284

Source: Authors survey data

Members of the households interviewed were between 5 to 10 members. However the number of adults aged between 18 and 60 years were between 2 and 5 members per household meaning that majority of the household members were aged below 18 and/or above 60 years implying a higher dependence ratio among the households in the district. This has implication for the household labour and. Gender, as expected had more male than female (table 2), but more households in Bungoma district were dominated by female.

Table 2: Gender of Head of Household

District	Female	Male
KEIYO	17	26
LUGARI	24	27
BUNGOMA	59	31
TOTAL	10	90

Source: Authors' survey data, 2010

Given the land size and the reliance on farming highlights the significance of small scale farming in general household livelihood of the household in these economies. This is compounded further by the level of education, land tenure and the role of gender. With majority of heads having limited professional training imply inaccessibility to formal employment, hence low off farm income. Communal land ownership is frequently considered a constraint on farm productivity; farmer's effort may be side-tracked in the bid to balance socio-cultural obligations with the demands of commercial agriculture (Kingi and Kompas, 2005). Lack of security may also impede access to credit services that accrue due to other tenure systems.

There were variations in the percentages of land allocated to various enterprises of the overall land size against individual enterprises reflecting possible intercropping among some enterprises in the farm and renting of land outside the farm as noted earlier. Input use in AIV production varied as indicated in figure 2. African indigenous vegetables occupy about 9.6 percent of the total land size on average in the three districts. Moving from up lands to lowlands, Keiyo district allocated the highest percentage of land size to the AIVs. Comparatively farmers grow less exotic vegetables (Kales e.g. Cabbage and *Sukumawiki*) with an average of 6.61 percent of the total land cultivated. Keiyo district still had the highest percentage allocation of average land size of 15.82 followed by Bungoma and Lugari at 8.2 percent and 7.6 percent respectively.

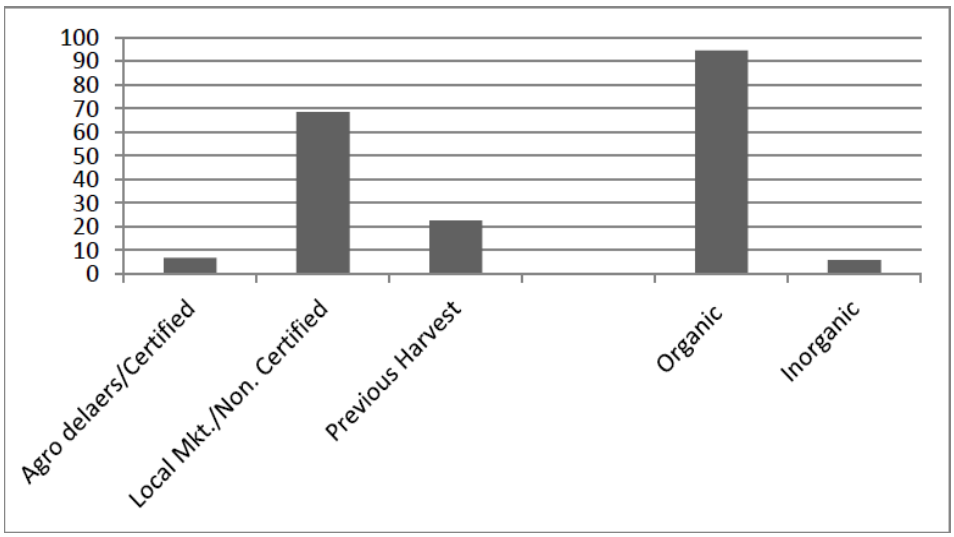

Figure 1. Seeds and Fertilizer use in AIV production.Keiyo district with the least average land had a higher percentage of land allocation to both AIVs and Banana productions. Unpredictably, Sorghum and Millet production exceeded that of traditional cereals e.g. Maize and Common beans.

Factors Influencing AIV Production

Multiple regression analysis was used to determine socioeconomic factors influencing household decision to allocated land resource to African indigenous vegetables. Table 2 indicates the variables influencing the farmer decision.

Table 3. Factors influencing land allocation to AIVs

Estimated parameters of Factors influencing AIV Land Allocation		
Variables	**Coefficients**	**Std. Error**
(Constant)	38.111	(4.316)***
Agro Ecological Zone	-5.253	(0.991)***
Age of the Head of Household	-0.016	(0.212)
Education Level of the Head of Household	2.420	(1.098)**
Off Farm Income	0.995	(1.779)
Distance to the Nearest Market	0.039	(0.059)
Gender of the Head of Household	-6.299	(2.397)***
Employment of the Head of Household	-1.450	(2.249)
Overall Land Size	-0.534	(0.157)***
Fraction of Land Size allocated to Exotic Vegetables	0.063	(0.075)
Fraction of Land Size Allocated to Maize	-0.293	(0.041)***
Fraction of Land Size Allocated to Livestock	-0.313	(0.053)***
Extension Services Received	3.528	(2.025)*
Access to Water for Irrigation	-4.495	(1.804)**
R2= 0.471		
Adjusted R Square=0.428		
Model F-Value 11.654***		
Level of significance denoted as *, **, *** representing 10%, 5% and 1% respectively		

Source: Authors Survey Data, 2010

Agro ecological zone significantly influenced the ratio of the size of land allocated to indigenous vegetable. Moving down the zones, a farmer in Keiyo is likely to allocate more land compared to a farmer in Lugari and Bungoma districts respectively. Indigenous production is influenced by the gender of the head of household. Female headed households are likely to allocated more land and by extension more resources to indigenous vegetables.

Indigenous vegetables compete for land resource with maize (staple food) and livestock production. Remarkably, as the scale of production increases, the ratio of land allocated to indigenous vegetables diminishes. While education level of head of household favoured the production of indigenous vegetables, poor access to water for irrigation discouraged its production. Farmer's access to extension services also evidently influenced increase production of the vegetables.

When the ratio of land size allocated to AIVs is controlled for, partial correlation (table 4) shows that agro ecological zone of the farmer is positively correlated with the choice of seeds and the method of water abstraction for irrigation in AIV production. There was also a positive relationship between the choice of seeds and the incidences of pests and diseases. The method of water abstraction was also positively correlated with the education level of the head of household and scale of production. However, access to water for irrigation negatively correlates with the abstraction method. Farm size also negatively correlated with off farm income with a likely scenario that as the size of land decreases households become more dependent on off farm sources for their livelihoods.

Conclusions

Majority of farmers rely on farming as a source of livelihood and that socio economic factors dramatically interact to influence significantly the decision of farmers in the production of the African indigenous vegetables. Land is a common and key constraint among farmers with an average land holding of 1.52 hectares. AIV competes for limited farm resources receiving less attention compared to other crops in terms of land allocation and general crop husbandry. *Sucha (black night shade)* was found to be a widely grown vegetable in the region among all the AIV farmers. Organic fertilizers were also used by most farmers as oppose to inorganic fertilizers. Majority of farmers use non-certified local market seeds and organic fertilizers.

Production of the AIVs in terms of decision of land size allocation is influenced by several socio economic factors within the household. Agro ecological zone of the farmer, gender, scale of production, and production of maize (staple food) and livestock significantly influenced the amount of land the farmer allocated to AIVs. Other significant factors included; education level of the head of household, access to water for irrigation and extension services. Remarkably, the choice of seeds and the incidences of pests and diseases were positively correlated. The age, education level, scale of production and off farm income also interrelate to influence allocation of resources and choice of technology in production of African indigenous vegetables.

It is recommended that AIV value chain should be developed and farmers supported to integrate in the for access to both input and output markets. Specifically, the Government of Kenya and NGOs should intervene by supporting farmers to access and use certified seeds and water for irrigation. Varieties and production of certified seeds should factor in the indigenous knowledge, and tests and preferences of the local farmers to ensure adoption. Affordable irrigation technologies should be provided also to reduce reliance on rain-fed AIV production. Ministry of Agriculture should also intensify extension services with the information tailored and packaged to target AIV husbandry through the socioeconomic agents as stated. Further stud could be done on the specific role of AIVs in the household dietary and food security.

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Promoting Forest and Non Timber Forest Cultivation to Increase Farmer's Income on Small Scale Private Forest (A case study at Tanjung Raya Village, Samarang Sub District, Garut, West Java)

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Abstract

More than 60% of Javanese population with relatively small land ownership depend on agricultural sector (0.3 ha/household). They live in dense populated villages surrounded by forest; resulting high pressure on forest existence. Consequently, frequent incidents of social conflict occur and deteriorate forest condition and eventually prone to flood, erosion and landslide. An alternative solution to accommodate rehabilitation of forest function and fulfilling community needs is forest and non timber forest cultivation simultaneously. This new cultivation technique was introduced to farmers through experimental plot. The research objective was to study Eucalyptus urophylla cultivation together with Andropogon zizanioides (vetiver oil plants) on small scale private forest in Tanjung Karya Village, Samarang sub district, Garut, West Java. Participatory approach was used in developing the model; hence farmers participate actively in all stages of model establishment. Series of discussions (individual interviews and continued with Focus Group Discussion/FGD) were carried out in advance to gain better mutual understanding about the purpose of the research. Further information about people's preferences towards tree crops combination was also previously collected. The results showed that although a light demanding species, vetiver oil plants could grow well under E. urophylla until it has harvested (13 months old) for three rotation periods. By integrating tree and non timber forest plants, farmer could improve not only soil condition but also their income significantly.

Keywords: ***A. zizanioides (vetiver oil plants), Eucalyptus urophylla, farmer's income, rehabilitation, small scale private forest***

Introduction

The disturbance of natural forest resources which has multi functions would give negative impacts not only on the existence of flora and fauna, but also to the community around and even far from the forest. Forest degradation causes declining of forest function in providing life resource for the people and its role as source of foreign exchange of the country. Forest deterioration even might cause damage in ecological function especially as supporting factor of community life, which in turn could generate serious disaster for human being.

In dense populated area surrounded by forest like in Java island, it will increase pressure towards forest area. Average land ownership in Java is only 0.3 ha/household (60% of Javanese population) (Raswa, 2006; Getteng, 2011; Jemabut, 2011). With this small ownership, it is difficult for the farmers to fulfill their daily necessities. In order to fulfill basic necessities each household should have/cultivate at least 2 ha/land (Sumarno and Kartasasmita, 2010; Getteng, 2011). The figure is much less if compared with farmers in

Thailand, Malaysia and Australia which in average cultivate 5 ha, 4 ha and 100 ha respectively (Sumarno and Kartasasmita, 2010). With minimum land ownership, it is difficult also for the farmer to cultivate their land efficiently. In Tanjung Karya village, Garut, most people used to cultivate annual crops (vegetable and fruit crops) and other cash crops like vetiver oil plants (*A. zizanioides*) with minimum input application. Consequently, after several years, soil condition has become deteriorated & production decreases significantly. In addition, topography in Tanjung Karya Village is mostly steep and undulating (Fig.1). Intensive farming cultivation together with extreme landscape has caused the area is prone to land slide, flood and soil erosion like what happened in the area in January 2010 (Fig.2).

Figure 1. Land condition in research site

Figure 2. Landslides in Tanjung Karya , with steep and undulating topography, Jan 2010

Since last three decades, Government of Indonesia has initiated several programs aiming to overcome the problem of deforestation and forest degradation. However, the result was not success and satisfy. Conflict of interest between ecological and economic consideration often collide each other, hence that hindered the success of the programs. Therefore, more comprehensive and reasonable efforts are required to solve the problem. The emerge question is, how to adapt those two aspect properly. Alternative solution to solve the problem is the effort to accommodate and link between ecological consideration in one hand and urgent social economic needs on the other hand. This could be done by introducing a new cultivation technique which is adaptive and applicable for the farmers through combining long life tree with non timber forest plant (cash crop) simultaneously.

In order to show farmers this new farming technique, a participatory demonstration plot is needed to be established. Previously, discussions with local people were carried out to gain better mutual understanding about the purpose of the research and what they could learn and what benefits they might get from it. Further information about people's preferences towards tree crops combination was also collected previously. It is expected that by accommodating people's expectations and desires, it would increase their awareness about the importance of integrating forest and non timber forest cultivation to rehabilitate soil condition, preventing natural disaster and increasing farmer's income simultaneously. The objective of the research was to promote intercropping cultivation of forest and non forest product to increase farmer's income on small scale private forest while rehabilitating degraded forest land.

Materials and Methods

Location and time

The research was conducted in Tanjung Karya Village, Samarang Sub District, Garut Regency at approximately 6º56'49'' - 7 º45'00'' South latitude and 107º25'8'' - 108º7'30'' East longitude with elevation 1300m above sea level. Average annual rainfall in the village was 2589 mm with 9 wet months and 3 dry months. Average monthly temperature varies from $24^{o}C - 27^{o}C$. Topography of research site varies from flat, hilly and mountainous. In general, soil condition in the village is relatively fertile and most is private owned and cultivated with annual food crops. Total area of the village was 477.869 ha (Pemerintah Kabupaten Garut, 2011). The research has been conducting since 2006.

Approach

The research was an action research through establishment of participatory demonstration plot on small scale private owned forest land. It was implemented in two main phases i.e. survey and demonstration plot establishment. First stage (interviews and discussion with candidates of participants) was intended to study about biophysical condition of the research site, social economic and cultural condition of local community, people's dependency upon land (both agricultural and forest land) and to investigate prospect of community participation. From the initial research phase, description about existing farming system, land productivity, constraint and problem people commonly face could be anticipated before. The next step was formulation of plot design including species combination which was going to be planted in the research plot. By owning this information, it was expected that candidates of participants would really understand about the purpose of the research and eventually could increase their active participation in demonstration plot establishment.

Method of data collection and analysis

Data presented in this paper were collected from field survey (primary data) and literature study (secondary data). Primary data was collected through direct interviews with selected respondents (local farmers, key persons, and intermediate trader) including social economic condition of the people, farming system commonly applied, productivity of tree crop planted (tree age, price, height and diameter) and marketing prospect. Whereas secondary data was collected from several publications. All data collected then would be analyzed descriptively.

Results and Discussion

Social economic condition of the people in the research site

In 2010, total population of Tanjung Karya village was 8335 consisting of 4174 man and 4161 woman. Average population density in the village was 46.25 persons/km^2. Level of education of the people in the village is relatively low. Most of them do not finish their basic education/elementary school (59.7%) and there is only 14,2% who could reach Senior High School. Main occupations of the people are farmers or on-farm labours. Most of the land is intensively cultivated for paddy, maize, cassava, sweet potato, vetiver oil pant and other fruit and vegetable crops like tomato, eggplant, chilly, long bean and peanuts. For

fulfilling their daily needs, people totally depend on their on-farm job (Desa Tanjungkarya, 2010).

Size of landholding is relatively not so wide ranging from 210 – 850 tumbak/bata (1 tumbak/bata ≈ 14 m²) or around 2940 m² - 11.900 m² but most of them has less than 0,5 ha/household, even some of the people in the village are landless. In order to meet daily necessities, landless people used to rent land and develop benefit sharing cultivation. After cultivated for several periods (usually 4 – 5 years) with limited input application, usually soil fertility declines very rapidly and land tenant then just abandon those degraded land.

Agreement Achieved

Based on the baseline survey and initial information gathered, 2.5 ha demonstration plot of *E. urophylla* and *A. zizanioides* (vetiver oil plant) combination was established. *E. urophylla* and *A. zizanioides* were planted together with 3 x 3 m and 0.5 x 0.5 m spacing respectively. There are 8 farmers involved in research plot establishment. *Eucalyptus* trees were planted first and vetiver oil plant was planted three months afterwards (Fig. 3&4).

Figure 3. Vetiver oil plant (*Andropogus zizanioides*)

Figure 4. *E. urophylla* tree stands intercropped with vetiver oil plant

Location of the plot is on private owned forest. Hence, success key of plot establishment is willingness of the people to maintain tree-crop in the research plot. A mutual agreement was designed in advance to guarantee that *Eucalyptus* trees will not be cut before research is finished (6 years). It was not easy to achieve mutual agreement since average land holding of participating farmers are in general not so wide (less than 0,5 ha) and vetiver oil pant is light demanding species which they prefer more than *Eucalyptus* trees.

General description about tree-crop planted in the research plot

Selection of *E. urophylla* and *A. zizanioides* (vetiver oil plant) are based on several considerations either by social economic and ecological aspects. Description about those species as follow:

E. urophylla

E.urophylla is an evergreen tree up to 45 m tall, or, in unfavorable conditions, a shrub; bole straight, branchless for up to 30 m. *E. urophylla* frequently occurs as dominant species in open, often secondary mountain forests. It grows on mountain slopes and in valleys and is commonly found on basalt, schists and slates, but rarely on limestone.

Spacing varies with purpose of the plantation. For pulpwood, 3 x 2 m is commonly used, and for fuel wood or poles spacing may be closer. It is essential to keep the field free of weeds until the trees are 6 months old. Thinning is done every 2 years from the age of 3 years onwards. *E. urophylla* has good coppicing ability and can be expected to produce at least 3 coppice rotations after the initial seedling rotation. *E. urophylla* actually is a multipurpose tree species (MPTS). It makes satisfactory fuel wood and charcoal. It is also suitable as a source of mid-density to low-density eucalypt fiber for pulp and paper production. In Timor, the wood is used in heavy construction, bridging, flooring and framing. The round wood is used for building poles and fence posts. The bark has a tannin content of over 10%, but it is not used commercially. The leaves yield consist of pale yellow oil. The essential oil is a good source of paracymene, possesses disinfectant properties and utilized in soap making and perfumery industry (World Agroforestry Centre, 2004).

Specific reasons were mentioned by farmers to select *E. urophylla*:

- A great demand of forest products, which may be manufactured from *Eucalyptus* sp. Wood (67% of respondents)
- *E. urophylla* is very potential and preferred as pulp and paper raw material since it contains full filament (23% of respondents)
- Price of timber product including *E. urophylla* tends to increase from time to time (93% of respondents
- Fast growing, good coppicing and drought resistant tree species (70% of respondents)
- Planting of Eucalyptus is can be used to reclaim the degraded lands (17% of respondents)

Several studies also go along with those reasons and indicate that *E.urophylla* in the future would become one promising species and the popularity of this species has increased markedly for plantations in humid and sub-humid tropical climates that endure several months of drought annually (the wet/dry tropics) such as parts of Indonesia, Brazil and southern China (Eldridge *et al*. 1993 in Anonym, 2004). Yields of 20-30 m^3 ha^{-1} yr^{-1} of *Eucalyptus* have been reported under favorable growing conditions (Anonym, 2004). It can improve soil fertility, especially when planted on marginal agricultural lands or other areas with degraded soils. The usage of nutrient by *Eucalyptus* is efficient and lower than other planted tree species and agricultural crops (Anonym, 2009). This species is also reported use water more efficiently than other vegetations (FAO Regional Office For Asia and The Pacific Bangkok, 1993).

***A. zizanioides* (vetiver oil plant)**

A. zizanioides grows naturally in swamp areas of northern India, Bangladesh, Burma (Myanmar) and occurs probably naturalized in many parts of South-East Asia. Under favorable field conditions vetiver clump cuttings (splits) start sprouting a week after transplanting, but growth is generally slow during the first 3 months. In one year the root system becomes well developed. Some roots may reach a depth of up to 4 m. *A. zizanoides* is grown for its oil mainly in Haiti, West Java, India, Réunion, China and Brazil. From the rhizome and roots, vetiver oil is steam-distilled, which is used in perfumes, deodorants, soaps and other toilet articles. Its scent is heavy and woody (Sulistyawan, 2010).

Traditionally, *A. zizanioides* is planted in southern India in strips as permanent field boundaries and occasionally in contour strips to control erosion, while in Java it is planted to protect sloping drains. The use of vetiver in erosion control spread first from India to the Caribbean and Fiji and later to many tropical areas, including all countries of South-East Asia. Since the late 1980s, its planting for erosion control has been promoted strongly, not only around fields, but also to protect terraces and road shoulders. Strips of densely packed, stiff and tough grass stems break the speed of run-off water and divide it evenly, reducing the risk of formation of run-off streams and gully erosion. Very dense root system has a strong tendency to grow downwards and effectively anchors strips of plants and soil behind it (Damanik, 2005; Emmyzar, et.al, 2006).

Indonesia and Haiti export the largest quantities of vetiver oil, about 50—100 t/year each, while China exports about 20 t/year. The largest area of production in Indonesia is in Garut Regency in West Java where it is grown on about 20,000 ha land. The main importing countries are the United States and Western Europe (each with 100 t/year), and Japan (10 t/year). The price of vetiver oil varies between years and sources. In the 1990s, it was valued at about US$ 135—155/kg, oil from Haiti at US$ 90—100/kg and oil from Indonesia at US$ 54—62/kg; vetivery acetate at US$ 160/kg (Anonym, 2004).

The average maximum temperature required for good growth is 25°—35°C; absolute maxima may be about 45°C. It should not be shaded permanently, although healthy hedges of vetiver can be maintained in sugar-cane plantations, as the plants recover quickly after the harvest of the cane. The species is tolerant of very poor and adverse soil conditions. It can survive fire, rough trampling and grazing. For the production of vetiver oil, light sandy soils are required to facilitate harvesting of the smaller roots, which contain most oil.

To establish vetiver, early weeding is important. Weeding is done 3—4 times in the first year and a few times in the second. The final weeding is done just before harvesting to avoid roots of weeds in the harvested vetiver roots. Intercropping with short-duration pulses can be done during the early stages of growth. Intercropping of coconut and areca palm plantations with relatively open canopy is sometimes practised. Irrigation is sometimes economic. For erosion-control hedges it is essential to fill gaps between plants. Roots and rhizomes of vetiver are harvested 15—18 months after planting when their essential oil content is highest. In Java harvesting is sometimes done already after 12 months, elsewhere it is sometimes postponed until after 24 months, which results in lower yields, but higher quality oil, being heavier and darker coloured. The use of a single disk plough digging up to 40 cm depth was efficient and effective. However, on steep area, harvesting lead to serious erosion. The average yield of air-dried roots of vetiver varies from 1—2.4 t/ha, commonly yielding 12—17 kg oil. (Sulistyawan, 2010)

The growth of tree-crop in demonstration plot

A. zizanioides (vetiver oil plant) is mostly preferred by the community in the village. This species could provide cash income fast. Harvesting period varies from 8 – 13 month after cultivation. Beside the handily of cultivation, price of vetiver oil plant tends to increase intermittently. Marketing of this product is not difficult as demand for vetiver oil plant also tends to increase from time to time. Even, consumers (intermediate trader, local industry) are willing to buy it long before harvesting period. Unfortunately the species has disadvantage root character. When it is planted on steep area, monoculture and incautiously harvested, it will take away/remove soil surround the root area and leave unsolid soil. When rain comes, it will lead to a severe surface erosion. Therefore intercropping planting with tree stands having long and deep root is needed in such area to maintain stability of the soil after harvesting.

Fortunately, despite those disadvantages, vetiver oil grow well under *E.urophylla* until it has harvested even until three rotation periods, although its production tends to decrease gradually. Description of growth of vetiver oil plant *E. urophylla* is presented in the picture below.

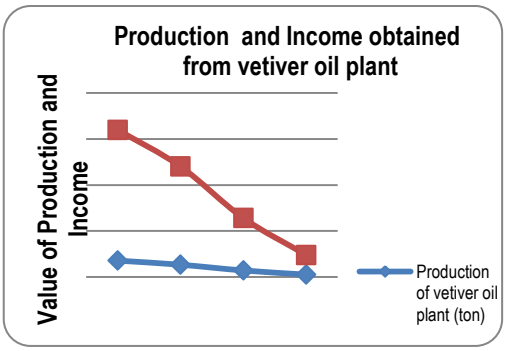

Figure 5. Production and income gained from vetiveroil plant

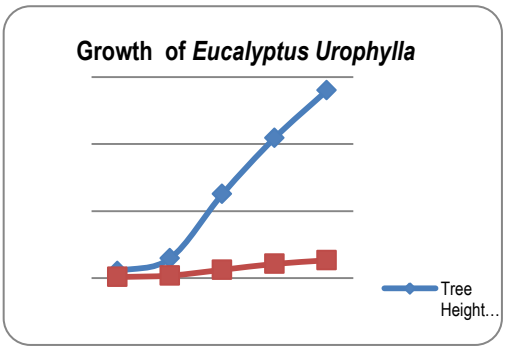

Figure 6. Growth of *Eucalyptus urophylla*

As a light demanding species, cultivating vetiver oil plant under *E. urophylla* decrease its production gradually from 100%; 75% and 40% respectively during three rotation periods (Fig.5). However the decreasing is compensated by the significantly growth of *Eucalyptus* (Fig.6).

Income Gained from Integrating Tree-Crop Cultivation

Table 1. Income obtained from monoculture cultivation of *A. zizanioides* (vetiver oil plant)/ha

No	Monoculture cultivation of *A. zizanioides* (0.5x0.5 m)	Total Income
1	Average production of vetiver oil plant	2.8 ton/ha*)
2	Current price of dried roots of vetiver oil plant	Rp 4000*)
3	Total income gained for one rotation period	Rp 11,200,000
4	Cost of production until harvesting 20%	Rp 2,240,000
5	Total net income	Rp 8,960,000
6	Total income gained during 6 years monoculture cultivation/ha (5 rotation)	Rp 44,800,000

Source: Primary data analysis Note: Assumed, price and level of production is stable during 6 years rotation period

Table 2. Income obtained from intercropping of E. urophylla and A. zizanioides during 6 years cultivation

No	Intercropping cultivation of *E. urophylla* (3x3) and *A. zizanoides* (0.5x0.5 m)	Description
1	Survival rate of tree stands (± 70%)	770 trees/ha*)
2	Average diameter of tree stands	20 -25 cm
3	Average height	15 – 20 m
4	Estimation of volume/tree stand	0.1 m3*)
5	Total of volume/ha	77 m3
6	Current price of *E. urophylla* (Rp/m3)	Rp 1,000,000,-*)
7	Estimation of Gross income/ha	Rp 77,000,000
8	Estimation of cost of production until harvesting 20%	Rp 15,400,000
9	Estimation of net income from *E. urophylla* (Rp)	Rp 62,600,000
10	Average production of dried roots of vetiver oil plant	2.8 ton/ha*)
11	Current price of dried roots of vetiver oil plant (Rp/kg)	Rp 4,000
12	Total net income gained for first rotation period (Rp/ha/year)	Rp 8,960,000
13	Average production of dried roots of vetiver oil plant at second period (75 % of initial production)	2.1 ton/ha
14	Total income gained for second rotation period (Rp/ha/year)	Rp 6,720,000
15	Average production of dried roots of vetiver oil plant at third period (40% of initial production)	1.12 ton/ha
16	Total income gained for third rotation period (Rp/ha/year)	Rp 3,584,000
17	Additional income obtained from intercropping cultivation (9 +12+14+16)	Rp 81,864,000
18	Additional income obtained if compared with Monoculture cultivation of *A. zizanioides* (during 6 years cultivation)	Rp 37,064,000 (82.7% increase of income)

Source: Primary data analysis; Note: *) Assumed production and price

As already mentioned before, although planted under tree stands declined production of vetiver oil plant gradually, this loss is substituted by tree production. Preliminary rough calculation of income obtained within 6 year rotation period (without calculating present value of money/interest rate), presented in table. 1 & 2.

Table 1 and 2 shows, intercropping cultivation of *E. urophylla* and *A. zizanioides* increased total income of the farmers significantly (82.7%) compared with monoculture vetiver cultivation. It also guaranteed sustainable income for the farmers although main portion of income will be attained at the end of *E. urophylla* rotation period (6 years).

Conclusions

1. Although a light demanding species, *A. zizanoides* still could grow well under tree stands.
2. By integrating tree stands and non timber forest plants, farmer was able to increase their income significantly although major part of income would be obtained at the end of tree rotation period (6 years).
3. Intercropping tree-crop cultivation between *E. urophylla* and *A. zizanoides* could maintain stability of the soil in the research plot and prevent from erosion and land slide incidents
4. Further study to compare soil condition between monoculture cultivation of *A. zizanoides* and intercropping cultivation with *E. urophylla* need to be done to investigate how far the decrease in erosion level.
5. More demonstration plot with different combination of timber and non timber species cultivation need to be introduced on areas prone to natural disaster such in Tanjung Karya.

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Environmental Friendly Attitudes of Women in Forest Management Based on Communities Empowerment (PHBM)

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Abstract

The aim of this research was to study the womens' environmental attitude and their contribution to forest management. The case study with qualitative approach has been applied. The stages in data analysis applied were categorization, organization, interpretation and understanding process. It could be concluded that the farmer women society showed their attitude that are harmony with environment including taking good care of vanilli plant in preparating, planning, implementing, controlling and evaluating the program. For continuation of the program it is suggested to provide women farmers with sustainable market for their products.

Keywords: environmental friendly attitude, women participation.

Introduction

Several studies have shown that program in which women included it shows a success, it could be that women are very strategic role in "Forest Managemnent with Society Program" (PHBM) activities. PHBM Programe that includes women could possibly stop logging. Similarly, PHBM will remove the negative image of community and rural women who have often been accused as forest encroachers for the purpose of meeting the needs of domestic energy (fuelwood). Based on the description, it can be argued that the achievement of gender equality conditions in forest management will significantly affect the conservation of forest environments, as well as significant that the PHBM will be more meaningful if the vision of gender. Research problem is : a. The extent of environmentally friendly attitudes of women in managing the PHBM; b. How environmentally friendly attitude contributions of women in supporting the success of PHBM

Attitudes of women who prefer the softness, cohesiveness and emotional relationships maintained and manicured natural causes no occupied or destroyed for the sake of power that tend to result in death (Mary Daly, in Putnam Tong, 1998). Knowledge of women who specifically due to their proximity to natural causes only women can preserve their love of nature for survival and not on mortality (Griffin, in Putnam Tong, 1998). Such views indicate that women have a character that is environmentally friendly. This can be explained by the paradigm of mutual relations between social systems and ecosystems of Terri Rambo (1983). This view explains that there is interplay between the social systems with ecosystems through the flow of energy, materials, and information. The various inputs from the social system into the same ecosystem as the ecosystem in the place of the social system, only the inputs that go into the ecosystem of the social system derived from human activities. In case this is meant by human activity is the act of female-friendly environment.

Such views reinforce the assumption that the advantages possessed by these women PHBM Programe will be managed by way of empowerment of women through programes that are gender perspective. It has been demonstrated from various research results. Search a variety of research related to women's empowerment model described in environmentally

preservation of the FAO results of research conducted in 1970 on the island of Java, about women's interaction with the forest environmentally, suggesting that there is an increasing amount of biomass consumption in 1981 reached 0.82 m3/men/year up to 0.85 m3 / men / year in 1986. In these studies proved that there is a physical impact on the environment that hinder sustainable development programs. This study focuses on the study of biomass consumption and the empowerment of women in environmental management.

Another study conducted by the UNDP-World Bank (1997) on women's participation in managing water, indicating that the inclusion of women in the provision of clean water is more effective. This study shows that the significant role of women in environmentally conservation related to water supply. However, reality showed the extension and technical training more clean water provision is aimed at men.

Women's Research Center (P3W) Universitas Padjadjaranin collaboration with the Department of Infrastructure Project SIWU (2000) conducted action research (action research) to target women who are unemployed bakulan traders because of the relocation market. Involving them in the waste management market (though worthy of waste into a product worth selling and improper waste into compost, though) besides contributing in the hygiene market environment, it also seeks to change attitudes towards household waste and provide for women's economic contribution.

Materials and Methods

Research carried out by the method of "Case Studies", the research that seeks to uncover a holistic (whole) subjects studied, in PHBM Program activities. Approach to research conducted with qualitative and quantitative manner. Details of the research activities include: Subjects were residents PHBM project participants, both men and women who are in the Village District Padasari Cimalaka, Sumedang District. Research paradigm is a sociological perspective associated with the paradigm of social facts, social definition and social behavior of feminist ecology. This means that socio-cultural background will be reviewed in the mechanism and process of PHBM Programe activities, in order to do activity of environmental conservation or environmentally friendly in a gender perspective.

Variable study consisted of environmentally friendly attitudes of women; The role of women in the management of PHBM Program and Implementation PHBM ; Contributions environmentally friendly attitudes of women in the success of PHBM. Sampel purposively determined location (aim), namely Sub Cimalaka, Sumedang District which is a PHBM Project activities of the Forest Service. Determined purposively sampled respondents (aims) of citizens, both men and women who participated in the activities of PHBM. At the time of the research has netted a total of 14 people consisting of 6 men and 8 women, consisting of members of the group of 11 people and village officials 3 people. Sources of primary data obtained from the individual as the unit of analysis through in-depth interview technique and through group discussions directional (focused group discussions / FGD). While secondary data from documents, decrees and regulations, as well as the research results obtained from different institutions. Depth interviews were conducted in the homes of informants and focus group discussions conducted in the home group leader. Qualitative data analysis done by organizing the data according to the pattern units, such as: environmentally friendly character of women, the pattern of eco-friendly women's role in the management of PHBM, the pattern of women's contribution to eco-friendly in the success of PHBM , and patterns of gender oriented forest management. Interpretation of

data is done through in-depth understading (Verstehen), namely to understand and uncover the meanings, feelings and thoughts of the study subjects.

Results and Discussion

Women's Eco-Friendly Attitude

This is shown in terms of, Sustainable character : Sustainable character in system means that the activity of the potential and show the progress in a variety of things that support productivity. Vanilla planting programs around the forest potential when seen from the friendly environment, because the cultivation of vanilla in addition to any economic value for farmers, also sustained in conserving the forest, so there is no erosion or loss of water sources. Experience after the plant vanilla, PHBM program was a source of water is always available, both for everyday life as well as for paddy rice farming. Plants "cebreng" as a pillar vanilla plants, turned out to care for water conservation. “Cebreng” plant traits, easy to grow and the leaves can be given as fodder. Such conditions as a mechanism to preserve the ecological cycle in plants, animals (natural resources) and economic productivity (economic resources). Such a condition has been running long enough.

The value of sustainability of the CBFM program can be studied from the development activities. As was revealed earlier that this program continues to grow. Beginning in 2001, Mr. Entis with 24 group (Farmer Forest / KTH) try first area of approximately 6 ha located in the region Tanjungkerta RPH, BKPH Tampomas. The following year, expanded to 8.5 ha. Good productivity, encourage an increase in the interest of farmers. In 2003 opened more land for the vanilla area of approximately 20 ha in RPH Naluk, still in the subordinate BKPH Tampomas. Farmers involved are members of KTH in the Village and Village Narimbang Conggeang Naluk District, District Cimalaka, Sumedang, as well as in the Village Padasuka, District Cimalaka. The concept of cooperation was made in the context of business continuity and preservation of forests, namely through the concept of mutually beneficial cooperation between communities and Perhutani.

Studied from the pattern of partnerships happen mutually beneficial cooperation mechanism that is in terms of planting and maintenance, the community acts as a contributor of energy and expertise. “Perhutani” while acting as land and capital, seedlings, planting wages and organic fertilizers. Thus the pattern will clearly indicate the existence of business continuity because it contributes to the economic needs of farmers who support the survival of farmers. Similarly, there was an agreement for the environmental compatibility of menunjuang harmony and community agencies namely: the existence of an agreement with the provisions of “Perhutani” each share 42.5% of net income. The rest, to the village by 5%, and 10% for management fee (administration fees).

The market opportunity is still open for vanilla products, including applicants from overseas (Japan) as a buyer want to stick with the contract system. But the farmer refused, because there is already tretap from domestic customers, as of Klaten and Bali Province.

Attitudes to maintain an environment : Attitudes to maintain an environment other than that have been nurtured since consciousness there PHBM Programe, as well as residents awakened to the existence and life, which depends on the potential of forests, ranging from agricultural crop production, orchards, vegetables, firewood and water resources. A resident stated:

"Forest livelihoods and sources of business"

One other resident reinforce the need to preserve the forest environmentally because:

"Forest is the mother of human life"

Both statements are in addition to awareness, also will automatically maintain both an Environmental followed with an attitude. Attitudes related to maintaining enviroment are 3 things:

a. Maintaining environmentally associated with agricultural programs source of life, namely planting vanilla, which formally gets referrals and as a partner with Perhutani
b. Maintaining an environment with concrete efforts directly participate in maintaining environmentally. What does the pack entis and his friends deliberately localize "suren"
c. Tree planting
d. Managing water resources for life

The second effort at first been challenged Perhutani, because she thought penetrated the forest. According to informan, separated plants then the citizens will not be penetrated planting every where, controlled and can be nurtured. Such an attitude shows how dependence on the forest residents. The next chairman of the group stated:

"Women creative here, what has not occurred to the men by the women thought to be making money; in the woods a lot of fodder, so if there is damage to the forest will be angry".

Sustainability efforts are also shown in the managers of the cycle of mutual benefit, as stated by one informant:

"Livestock sheep with mutually beneficial forest cycles, always using organic fertilizers".

Forests for sustainable business evidenced by the role of forests as a source of water. PHBM project from the forest into a source of water for several districts in West Java. As explained by one informant:

"People here keep the water source, due to be aware of and feel the benefits".

Society has tried to maintain water resources for the sustainability of business and life. Communities tried to maintain water should not be dominated by business interests or individuals. Society never retain water sources, informants described

"People tried to keep the source water is not used commercially, because it will damage the environment"

The statements indicate an awareness has been part of environmentally system, because the forest has provided a source of life (energy, matter and information). Similarly, the society's commitment to maintain natural resources and manage forests with two objectives, namely to the source of life and environment, means that society has an

associative interaction with the environment and participate in the program environmentally

How the role of women in PHBM project management is illustrated through their participation in the planting of vanilla. The concept of participation in project implementation through the cultivation of vanilla CBFM include the following stages: a. Preparation, b. Plan. c. Implementation. d. Monitoring and evaluation. e. enjoying the results

Women's participation in the CBFM program participation if the following steps indicate the following:

(1) The preparation phase; this phase of activity begins:

a. Socialization in the group that carried out by “Perhutani” and village officials
b. Consultation group to determine which plants will be planted, the determination of arable land area for members of the group and the formation of group organizations (chairman, secretary and treasurer)

(2) The next stage is the planning; activities include the distribution of arable land, of the agreement determined the average arable land from 0.25 to 0.28 ha per member depending on the ability of members of the land is mainly concerned with the willingness of labor and capital. This stage is also planned in other types of plants will be planted in addition to vanilla as intercropping, marketing, distribution of tasks to siskamling (ronda) especially before the harvest, distribution of tasks for daily pickets to keep the plants from pests of plants (especially monkeys). At this stage of preparation and planning role of women is not so prominent these activities mostly done by men.

(3) Implementation phase; activities include land clearing that is clean in preparation for planting crops host plant, host plant, vanilla seeds to plant, fertilizing, maintenance of wild plants (ngababad), pollination, harvesting, drying and marketing. At this stage the role of women seemed to stand on certain activities. Work to clear land is generally only done by men, because this work is considered to be quite heavy., While the women began working on the activities of the host plant to plant and sow seeds for vanilla, which was conducted jointly with men. Specialized in pollination activity is performed only by women, because this work requires precision and women considered to have the character to do the job. Pollination is quite time-consuming job, because it is done for two to three months for a full day from morning until evening. Plant maintenance jobs done by women with men that keep crops from wild plants disorder . Two months before the harvest full well guarded during the day or night to avoid losses due to theft is rampant. Job keeping the fruit vanilla done by women along with men. Recent work of harvesting fruit vanilla performed by women and men, while drying the fruit vanilla is generally done by women. Marketing of the work activities are usually done by men from the beginning to find a buyer, determine the price to sell.

Other activities are usually done by women is growing between the crop (intercropping) in the PHBM area. In intercropping plant activity is the role of women is very prominent,

especially in planting various vegetables such as tomatoes, scallion, chili, cayenne, mustard greens, medicinal plants, medicinal (kunjit, ginger, laos). The whole plant is the domain of women ranging from growers, harvesting up to sell (or sold themselves on middlemen to market). Income from crops by women between the garden provides a substantial contribution to supplement the family income. Between these reliable plants for additional pengahasilan because harvest time is too long and provide a regular income. Between other plants in the form of annual cash crops such as mahogany, "suren", jackfruit, avocado, coffee, bananas are also sown by women.

(4) Monitoring and evaluation stage; include vanilla crop monitoring and supervision organizational KTH.

a. Supervision of vanilla plants, ie plants that performed maintenance on a regular basis (cleaning plant / ngababad) and intensive supervision of the plant two months before the harvest by ronda. This activity is conducted jointly by men and women.
b. Supervision in the organizational KTH, conducted by the chairman of KTH and devices (secretary and treasurer) of the members. Surveillance instruments made by Perhutani a monthly list of fields to be reported every six months by KTH to Perhutani. Checklists in the form of notes on the extent of arable land, crop, productivity and sales results.
c. Surveillance in environmental sustainability, namely the supervision of forest encroachers who often perform well logging for firewood and building or supervision on water resources. Supervision of environmental conservation activities carried out by men and women. From some of the statements of respondents stated that women play an important role in monitoring environmental sustainability, as stated in the following statement :
   "Women's would be angry if there is any disturbance of water sources".

d. Evaluation is usually conducted on the harvest and sale of vanilla. Evaluation results are recorded by a chairman for the unknown factors become obstacles and how to overcome obstacles. Constraints are most felt by farmers KTH is the falling price of vanilla. At the first harvest of vanilla price of Rp 300,000 per kilo, the second crop of Rp 25,000 per kilo and the third harvest of Rp 8,000 per kilo for organic and Rp 5,000 per kilo for the non-organic.In this stage women also have contributed significantly, especially in the supervisory activities of plant and maintain plants before harvest, while the control activities in organization and evaluation activities generally do not involve women.
e. Phase enjoy the results; PHBM project provides an opportunity to rural communities living around forests to improve their lives through land management Perhutani produced by them through the vanilla crop. From the land tilled by members of the KTH while providing productivity vanilla plant that have good market opportunities, they also grow crops that provide income between significant for the survival of their families. This work was done by using family labor, both men and women. Mutually beneficial cooperation between Perhutani and rural communities around the forests provide considerable benefits perceived by members of the family farm as well as by Perhutani KTH.

Insights on gender in environmentally community PHBM

The role of women in the management of PHBM to receive recognition from men. This is apparent from the following statement:

"Men recognize women's skills in the cultivation and marketing of agricultural products".

From the above statement it appears that recognition of the role of women not only in terms of labor productivity but also its role in preserving environmentally. That men and women can work together as complementary to the welfare of families very well understood by society, not only in productivity and employment but also in decision-making.

Financial resources more families obtained from the work of the women's

Decision making is an important concept in analyzing the role of women in gender insight, because it shows the allocation of power between men and women in the household. From the observation in the field of women in decision-making than dominant in the domestic sphere, it also has the power to take decisions in the productive sphere. In the realm of men earning more informed decisions in marketing activities, while making use of the proceeds from the sale of vanilla and other crops is determined entirely by women.

Men more involved in forestry organizations

Contributions environmentally friendly attitudes of women in the success of PHBM Programe in productive activities : Women in this area including the communities productive. Women's labor productivity is shown by the enormous outpouring of labor in subsistence activities and the amount of income. The extent of labor shedding of women described by the informant, that the average in one day make a living from their work from 08.00 until 14.00 the job done in time serve targeted is the maintenance of plants, namely ngababad (clean vanilla plants from weeds), ngored (menggemburkan plants) and fix the fence and collect firewood as well as preserve the forest environment of the browser. At 14.00 after they went home to rest and do the housework until near sunset. Contribution of women's role in household income is significant, as shown by the fact the falling price of vanilla on the lowest price level (when the study was conducted Rp 8000 / kg wet), family finances were helped thanks to the sale still crop interrupted (vegetables, fruits , tubers, medicinal, medicinal) are grown and marketed by women. Revenues from the plant sale is varied depending on the sidelines of the kind of results that are marketed and the number of results that are marketed. However, from about 0.25 to 0.28 hectares of land under cultivation in the CBFM project, revenues from the sale of crops between the average of Rp 150,000 - Rp 250,000 per week. Revenue is significant in meeting the needs of the household. Income from the vanilla growers are also very varied depending on the area of land under cultivation, the amount of vanilla plants and plant fertility. Although the level of crop productivity in the vanilla harvest third pretty good (an average of 100 kg wet) compared with the first and second harvest, but the low market price of vanilla (Rp 8,000 per kg wet) lead farmers do not gain anything compared to the costs must be removed (to 0.25 ha required cost of Rp1000.000). In this situation the contribution of women in both the outpouring of employment, income and decision-making is in the CBFM project means enough. Environmental sustainability : Women's role in supporting environmental sustainability is evidenced by the prominent activities on women in the use of plants "cebreng" (tuturus). Other than as barriers to erosion as well as feed crops cebreng

delicious lamb that many managed by women. Thus women have to help preserve the environment as well as productive. As one farmer stated: *Women'is more diligent and skillful in raising livestock sheep*

Conclusion

The farmer society (women) had showed the environment harmony attitude with environment. The contribution of the women environment harmony attitude to the PHBM Programe showed in raising vanilli plant, started with preparing, planning, action, controlling, evaluation, and benefit form. For everlasting of the environment harmony attitude with environment of women need insurance with product market and to give many action programe spesial for women.

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AGRITECHNOLOGY PLANT SCIENCES

Antagonistic Activity of *Rhodotorula* spp. Against Spoilage-Causing Moulds on Tomatoes

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Abstract

Six strains of Rhodotorula spp. (Rhodotorula sp. UICC Y-318, Rhodotorula sp. UICC Y-325, Rhodotorula sp. UICC Y-332, Rhodotorula sp. UICC Y-381, Rhodotorula sp. UICC Y-384, and Rhodotorula sp. UICC Y-386) were investigated as antagonistic yeasts against Aspergillus ochraceus D1.2.2.SSM3, A. terreus D2.2.MC, and Drechslera sp. D1.3.MC. The yeast strains were isolated from plants at Cibodas Botanical Garden, and the mould strains were obtained from University of Indonesia Culture Collection (UICC). Antagonistic test by using strip method showed that higher percentage of inhibition of mould colonies was shown by Rhodotorula sp. UICC Y- 325 against Drechslera sp. (28.12%—72.14%), Rhodotorula sp. UICC Y-381 against A. ochraceus (54. 28%—72.46%), and Rhodotorula sp. Y-318 against A. terreus (21.76%—58.10%).

Keywords: Aspergillus ochraceus, A. terreus, biological control, Drechslera sp., Rhodotorula sp., tomato, yeast.

Introduction

Spoilage-causing moulds are one of destructive postharvest pathogen of tomatoes fruit during storage. *Aspergillus ochraceus* Wilhelm, *Aspergillus terreus* Thom, and *Drechslera* sp. are several spoilage-causing moulds that can infect tomatoes fruit (Oetari *et al*., 2007). The spore of spoilage-causing moulds is very common in atmosphere and this infection of fruit occurs mainly at wound site during harvest and packing (Qing & Tian, 2000).

Fungicides have been the most effective means of controlling tomatoes (Conway *et al*., 2007), but public concern about food safety and the development of fungicides resistance by pathogen has increased the search for alternative methods to controlling pathogen (Dorby, 2006; Lima *et al*., 1999; Korsten *et al*., 1998; Vero *et al*., 2002). Biological control of spoilage-causing moulds of tomatoes during postharvest storage by antagonistic microorganism seems increasingly promising to replace the fungicides (Chalutz & Wilson, 1999; Lima *et al*., 1999).

Some yeast have been reported to effectively reduce various postharvest diseases of fruit (Jones & Dov, 2002; Leibinger *et al.,* 1997; Lima *et al*., 1999; Zhang *et al*., 2007). Yeast is potential to be used as biological control because of their high inhibitory capacity, rapid colonization, and modes of action mainly based on competitive of nutrients, direct physical interaction with fungal hype and production of cell-wall lyctic enzyme (Lima *et al*., 1999). However, it is important to point out that most of selected yeast has frequently been tested for antagonistic activity on only few postharvest pathogens, while it is essential for the

commercial development of biocontrol agent to have a wide range activity (Janisiewicz & Lise, 2002).

In this work we observe about antagonistic activity of six strains of *Rhodotorula* spp. against three pathogens with *strip method* on PDA plate, with some modification experiment by Azizmohseni (2007).

Materials and Methods

Antagonists

Rhodotorula spp. strain UICC Y-318, UICC Y-325, UICC Y-332, UICC Y-381, UICC Y-384, UICC Y-386 were isolated from tomato plants at Cibodas Botanical Garden, West Java. The yeast was grown in yeast malt agar (YMA) in tubes at room temperature for 2 days. The yeast was suspended with 5 ml of sterile water and then homogenized with vortex. Suspension of *Rhodotorula* spp. was adjusted about 1.0 to 6 x 10^8 CFU per ml using TPC (Total Plate Count) method.

Pathogens

The *Aspergillus ochraceus*, *A. terreus*, *Drechslera* sp. are the pathogens used in this study were obtained from tomatoes plants at University of Indonesia Culture Collection (UICC) and Cibodas Botanical Garden, West Java. The pathogens were grown on potato-dextrose agar (PDA) in the tubes at room temperature for 2-4 days. The culture was added with 5 ml of sterile water and then homogenized with vortex.

Evaluation of Antagonistic Activity of *Rhodotorula* spp. Against Spoilage-Causing Moulds by Strip Method in Potato-Dextrose Agar (PDA)

The suspension of 48 h suspension of 20µl of antagonistic yeast was applied to each side of PDA plate (1cm from the center) to generate two steak of yeast inoculates. Four hours later, 20µl of spore suspension of pathogens were inoculated along the center line of the same PDA plate. The plates were sealed with parafilm and incubated at room temperature for 6 days. There were tree replications in each treatment. The antagonistic ability of *Rhodotorula* spp. were determined by examining their abilities to reduce the colony growth of spoilage-causing moulds by comparing the width of colony growth of pathogens on treatment with control of pathogens from day 2 until day 6. The formula to account the percentage of colony reduction of pathogens is determined by:

Percentage of colony reduction of pathogens = $\frac{a-b}{a} \times 100\%$

Explanation
a = the width of colony of control pathogens
b = the width of colony of pathogens in treatment

Results and Discussion

The use of biocontrol agent to manage spoilage-causing moulds on tomatoes fruit has been explored as an alternative to the use of synthetic fungicides. Several commercial products are now available (Dorby, 2006). Further observation about new antagonist is desirable because antagonist from specific area may be more effective against the pathogens strain present in that area (Vero *et al*., 2002). The experimental data in this paper demonstrate the occurrence of *Rhodotorula* spp. strains to control the growth of spoilage-causing moulds on tomatoes by strip method.

After 6 days incubation it was showed that *A. ochraceus* could grow normally in control PDA without antagonistic yeast (Table 1). At day 2 *A.ochraceus* has grown sporulated in all part of colony. The color of spores is ochre and the shape is granular. *Rhodotorula* spp. could reduce the colony growth of *A. ochraceus* (Table 2). Strain UICC Y-381 showed the highest ability (54.28-72.46%) to reduce colony growth of *A. ochraceus* during 6 day incubation.

After 6 days incubation, *A. terreus* could grow normally in control PDA without antagonistic yeast (Table 1) and not all strain could reduce the colony growth of *A. terreus* (Table 3). Strain UICC Y-325 at day 2-4 reduced the growth of *A.terreus but* in day 5 until day 6 incubation the growth of *A.terreus* was better than control. Strain UICC Y-318 showed the highest ability (21.76—52.27%) to reduce colony growth of *A. terreus* during 6 days incubation.

After 6 days incubation showed that *Drechslera* sp. could grow normally in control PDA without antagonistic yeast and all strain of *Rhodotorula* spp. could reduce the colony growth of *Drechslera* sp. (Table 4). *Rhodotorula* sp. UICC Y-325 showed the highest ability (28.12-72.14%) to reduce colony growth of *Drechslera* sp. During 6 day of incubation.

Table 1. Average of width three spoilage-causing moulds on PDA during 6 days incubation on 25-27 ^{0}C

No	Code UICC	Species Name	Width (mm) \ Day of observation				
			d-2	d-3	d-4	d-5	d-6
1	D.1.2.2.SS.M3	*Aspergillus ochraceus*	8.68	16.46	22.95	33.69	41.06
2	D.1.3.MC	*Aspergillus terreus*	7.22	12.89	20.05	25.94	29.50
3	D.2.2.MC	*Drechslera* sp.	10.47	13.26	17.89	33.48	37.96

Table 2. Percentage of colony reduction of Aspergillus ochraceus on PDA during 6 days incubation on 25-27 0C

Code of Yeast strain	Day of observation and colony reduction (%)				
	d-2	d-3	d-4	d-5	d-6
UICC Y-318	41.15	22.57	0.57	-0.35	13.72
UICC Y-325	51.28	66.06	63.92	46.99	42.71
UICC Y-332	48	67.25	36.66	27.58	25.56
U ICC Y-381	54.28	70.16	66.77	72.46	60.13
UICC Y-384	55.76	66.45	53.02	41.53	35.74
UICC Y-386	41.48	43.30	33.80	24.07	21.96

Table 3. Percentage of colony reduction of Aspergillus terreus on PDA during 6 days incubation on 25-27 0C

Code of Yeast strain	Day of observation and colony reduction (%)				
	d-2	d-3	d-4	d-5	d-6
UICC Y-318	52.27	58.10	50.55	37.55	21.76
UICC Y-325	34.91	18.58	0.68	-2.67	-9.84
UICC Y-332	48.51	61.62	62.41	20.09	4.71
UICC Y-381	68.77	21.21	26.04	33.93	15.68
UICC Y-384	31.1	57.38	18.53	11.38	0.45
UICC Y-386	40.56	58.79	28.20	1.28	-2.05

Table 4. Percentage of colony reduction of Drechslera sp. on PDA during 6 days incubation on 25-27 0C

Code of Yeast strain	Day of observation and colony reduction (%)				
	d-2	d-3	d-4	d-5	d-6
UICC Y-318	42.81	61.72	63.49	63.46	28.11
UICC Y-325	72.14	64.86	70.82	69.80	28.12
UICC Y-332	59.89	65.46	63.22	53.11	44.76
UICC Y-381	41.83	59.28	28.79	35.72	24.32
UICC Y-384	67.24	65.46	28.79	35.72	24.32
UICC Y-386	67.24	51	8.44	20.28	1.87

From the Table 2,3,4 we can suggest that *Rhodotorula* spp. reduce the colony growth of spoilage-causing mould at room temperature for 6 days. However the antagonistic ability of yeast strain against pathogens were different. These also suggested that *Rhodotorula* spp. could be applied as biocontrol agent to reduce spoilage-causing mould on fruit in tropis climate like Indonesia. All strain of *Rhodotorula* spp. were isolated from tomatoes plant (fruit, leaves and stem), which suggested that they get the nutrition from plant, but in this antagonist test, they were grown on PDA, which is not they natural nutrition. However, the *Rhodotorula* spp. can grow well in that medium. This was shows that the yeast have high ability to adapt on different nutrition.

The mechanisms of suppression by the introduced microorganisms are not yet understood. Previous studies established that nutrient competition is a mechanism of biocontrol that is likely occured in many yeast isolates. *Sporobolomyces roseus* and *C. Laurentii* were known to utilize the same nutrients like sugar (glucose, fructose and sucrose) that is required for germination and growth by *Botrytis cinerea.* (Filonow, 1998). Other mechanism is production of cell-wall lyctic enzyme, e.g. β-1.3-glucanase, chitinase that can damage the hypha wall (Lima *et al*., 1999; Zhang *et al*., 2007). Hypha can be degraded by yeast to simple compounds that can be used as sourced of nutrient for their growth and reproduction. However, the mechanism of *Rhodotorula* spp. to reduce colony growth of spoilage-causing moulds was not examined during the study. And therefore should be investigated further.

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Indonesian Rainfall Patterns: A Dramatic Shift

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Abstract

Indonesia's climate is influenced by various Earth-system processes including the El-Niño Southern Oscillation (ENSO), the Indian-Ocean Dipole, monsoon variability and the Julian-Madden Oscillation[8]. Previous analyses of rainfall data from 1951-1997 indicated a tight coupling between Indonesian rainfall anomalies during the dry season, peaking in August. Nevertheless, rainfall anomalies tended not to persist from the dry season into the wet season and rainfall in the heart of the wet season tended to be spatially incoherent and inherently unpredictable[9,10]. Whether this pattern extended into recent years, being subject to accelerated global warming, was unknown. Based on rainfall data covering period 1900-2008, shown that the Indonesian rainfall variability has dramatically changed from the early 1970s onwards. Instead of being seasonally confined, ENSO events have switched into a mode in which they also control Indonesian rainfall anomalies outside the dry season. This prolongation of ENSO influences and extended El-Niño episodes from the early 1990s onwards explains the observed decline in annual-mean rainfall by ~20% in the entire Indonesian region and why this decline occurred spatially coherent and during all seasons of the year. Given the widespread impacts of ENSO events, we anticipate these findings to be a clue in the understanding of climate-change impacts on changes in climate variability in the Indo-Pacific region. For instance, the sudden onset and persistence of intensified ENSO influences over the last 40 years, revealed here via an analysis of Indonesian rainfall anomalies, could point to a dramatic and shift in ENSO-related climate variability associated with changes in the Walker circulation influencing most areas across the tropical Pacific Ocean.

Keywords: climate variability, ENSO, Indonesian rainfall

Introduction

Indonesia is located in a climatic zone influenced by key drivers of climate variability; that is, the El-Niño Southern Oscillation in the tropical Pacific Ocean (Adrian and Djamil, 2008; D'Arrigo and Smerdon, 2008; Giannini *et al*., 2007; Haylock and McBride, 2001; Hendon, 2003) and the Indian Ocean Dipole associated with climate feedbacks in the tropical Indian Ocean (Saji *et al*., 1999; Ashok *et al*, 2001). Indonesia's weather is strongly influenced by monsoons on a seasonal basis being modified by these key drivers of climate variability (Adrian and Djamil, 2008; D'Arrigo and Smerdon, 2008; Giannini *et al*., 2007; Saji *et al*., 1999; Ashok *et al*, 2001; Qian *et al*., 1999) and, in austral summer, by atmospheric disturbances associated with the Julian-Madden Oscillation (Hidayat and Kizu, 2010).

Evidence of changes in Indonesia's climate in recent decades can therefore give important clues on the effect of global warming on climate variability in the Indo-Pacific region and changes in the frequency and duration of "extreme events" such as floods and droughts. Many countries of tropical influences including Indonesia have experienced pronounced drought periods in the last two decades. The particularly intense 1997 -1998 El Niño brought the worst drought in over 50 years to Indonesia (Kirono *et al*., 1999) (and many

other tropical countries in the world) and helped to trigger and feed forests fires across Borneo that affected more than a million acres (Murty *et al*., 2000).

Previous studies (Haylock and McBride, 2001; Hendon, 2003) based on rainfall data from 1951-1997 found that ENSO-events had a pronounced influence on Indonesian rainfall exclusively during the dry season (peaking in August), but could not be used to predict rainfall in the wet season (December-February). However, these studies (Haylock and McBride, 2001; Hendon, 2003) did not include the most recent data revealing a dramatic annual-mean rainfall decline commencing in the late 1980s (Figure 1). For instance, annual-mean rainfall anomalies in the Indonesian region attained record minimum values of -5.4, -7.1 and -4.9 cm/month in the years 1993, 1997, and 2004, respectively. This corresponds to relative rainfall declines in a range of 22-32% when compared with the long-term annual-mean of 22.5 cm/month for the 1900-1979 period. The driest year on record occurred during the 1997/98 El-Niño. Interestingly, the rainfall data show no evidence of increases in annual-mean precipitation in Indonesia during the wetter years of the last 20 years.

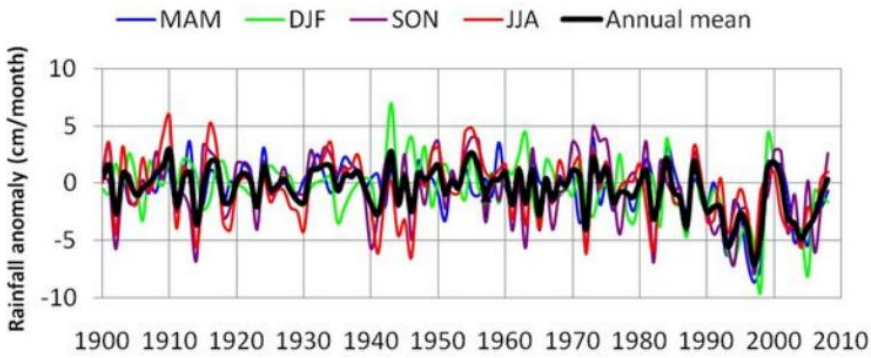

Figure 1. Indonesian rainfall anomalies (cm/month) during 1900-2008. Shown are averages for DJF, MAM, JJA, SON, and the annual mean. Arrows indicate events in which local maxima/minima of the annual-mean value are predominantly triggered by local maxima/minima of the JJA and/or SON value. Anomalies refer to the long-term mean value for the 1900-1979 period of 22.5 cm/month.

Materials and Methods

Rainfall data were taken from monthly terrestrial precipitation data set for the period 1900-2008 developed at the University of Delaware provided at ½ degree spatial resolution. Indonesian rainfall data are primarily based on station observations from the Global Historical Climate Network, version 2 (GHCN), and station observations from the Global Surface Summary of the Day (GSOD).

The time series of NINO 3.4 was computed from monthly sea surface temperature (SST) anomalies in the region 5^{o}N–5^{o}S, 120^{o}–170^{o}W from the HadISST1 data set (version 1.1) produced by Meteorology Office Hadley Centre for Climate Change. Monthly values of the Dipole Mode Index (DMI) were computed from the difference of SST anomalies between the western equatorial Indian Ocean (50^{o}–70^{o}E, 10^{o}S–10^{o}N) and the south-eastern equatorial Indian Ocean (90^{o}–110^{o}E , 10^{o}S–0^{o}N), again using HadISST1 data. Correlation values are calculated from the commonly used Pearson method.

Results and Discussion

In order to get a clearer picture of the observed rainfall decline in Indonesia, we separated the Indonesian region into four climatic sub-regions, similar to the approach taken in previous studies (Aldrian and Susanto, 2007; Aldrian, 2003). Region A comprises southern parts of Sumatra, the whole of Java, adjacent islands (Timor), and southern parts of Kalimantan and Sulawesi (Celebes). Region B contains the northern parts of Sumatra and Kalimantan including Borneo. Region C includes northern parts of Sulawesi and islands located in the Banda Sea. Region D includes the whole of Papua. Region A, containing most popular tourist locations, displays a monsoonal rainfall pattern (Aldrian and Susanto, 2007; Aldrian, 2003) with a dry season during June-October and a wet season during December – February. Region B displays an "equatorial" rainfall pattern (Aldrian and Susanto, 2007; Aldrian, 2003) that, in addition to a rainfall maximum around November, displays increased rainfall around April. Region C displays a "local" rainfall pattern (Aldrian and Susanto, 2007; Aldrian, 2003) displaying a wet period peaking in June and July. We introduce a fourth region D representing rainfall anomalies over Papua that previous studies (Aldrian and Susanto, 2007; Aldrian, 2003) included in region A. Rainfall over Papua, however, is usually much steadier during the year, ranging between 20 and 30 cm/month (results not shown), than in our region A where rainfall tends to range between 5 and 30 cm/month (results not shown). Nevertheless, while intra-annual rainfall patterns are vastly different among these regions, all regions show a dramatic decline in rainfall during 1990-2008 in all months of the year by 4.5±3.3 cm/month (or 20.7±14.7%) at 95% confidence level in comparison with the 1900-1989 average (Figure not shown). This decline had the most dramatic effect during the dry season in region A (Sumatra, Java and southern Kalimantan and Sulawesi) where, on average, the number of months per year of rainfall <5 cm/month has increased from 1 to 5 during the last 20 years (results not shown). Given the reported (Haylock and McBride, 2001; Hendon, 2003) seasonal restriction of atmospheric and climatic influences on Indonesian rainfall, the observed rainfall decline across the entire Indonesian region and in all seasons is an intriguing scientific puzzle.

In agreement with previous studies (Haylock and McBride, 2001; Hendon, 2003), our correlation analysis indicates an overall pronounced ENSO influence on seasonal rainfall anomalies during the periods June-August (JJA) and September-November (SON). Also, these seasonal rainfall anomalies appear to control the annual-mean rainfall anomaly (Haylock and McBride, 2001) (see Figure 1). Hence, we can attempt to reconstruct annual-mean rainfall anomalies from seasonal averages of climatic indices. To this end, we used NINO 3.4 (Bunge and Clarke, 2006), reflecting ENSO anomalies, and, in addition to this, the Dipole Mode Index (DMI) (Saji *et al*., 1999), reflecting anomalies associated with the Indian Ocean Dipole, for definition of a combined index κ, given by:

$$\kappa(\mathrm{NINO3.4, DPI}) = (1-\varepsilon)\frac{\mathrm{NINO3.4}}{std(\mathrm{NINO3.4})} + \varepsilon\frac{\mathrm{DPI}}{std(\mathrm{DPI})} \quad \ldots\ldots(1)$$

where ε is a weighting factor and *std.()* denotes the standard deviation. The calibration of ε follows from correlations of (1) with annual-mean rainfall anomalies whereby the latter are normalised by their standard deviation. In order to reveal changes in rainfall dynamics in the past decades, this calibration and standard deviations are exclusively calculated from values for the period 1900-1979. Once the best value of ε is found, annual-mean rainfall anomalies can be reconstructed via:

$$\text{Reconstructed rainfall anomalies} = -std(\text{rainfall anomalies}) \cdot \kappa(\text{NINO3.4, DPI}) \quad \ldots.(2)$$

whereby the standard deviation of rainfall anomalies is a constant being derived from 1900-1979 values. SON anomalies of the climate indices used together with ε = 0.3 gave the best correlation of a value of -0.80. At 95% confidence level, the correlation value lies in a range between -0.87 to -0.70 when using Fisher's transformation (Plata, 2006). Note that the Indian Ocean Dipole has a small but not dominant influence and that JJA values of climate indices gave a slightly reduced correlation value of 0.73 (results not shown). Obviously, the reconstruction of annual-mean rainfall anomalies in the Indonesian region works well for the 1900-1979 period, but significant differences become apparent from the late 1980s onwards (Figure 2). What has changed?

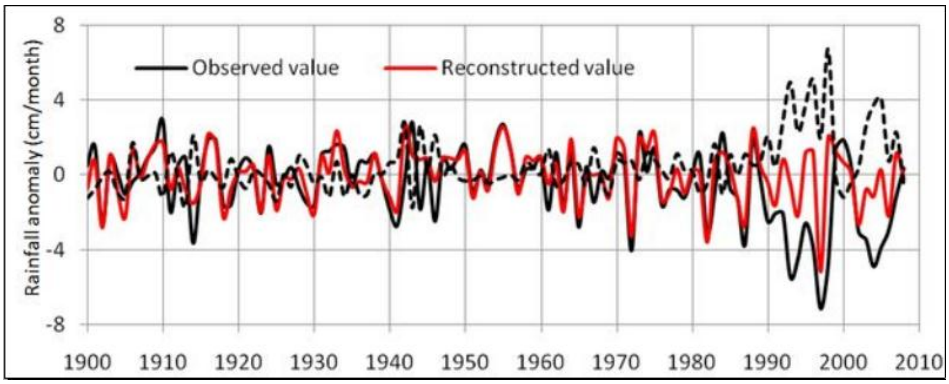

Figure 2. Observed and reconstructed annual-mean Indonesian rainfall anomalies for 1900-2008. Reconstructed values are based on equation (1) with ε = 0.3. The dashed curve shows the difference between reconstructed and observed values.

In order to identify trends and shifts in Indonesian rainfall patterns, we calculated cross-correlations of the entire time series based on a sliding window of 11 years in width. Although this method involves relatively low confidence levels, which is unavoidable, it can give first indications of intra-decadal and inter-decadal changes. For instance, the sliding-window correlation between JJA and SON values of NINO 3.4 with DJF rainfall anomalies indicates that there was no apparent correlation prior to the 1970s whereas the situation suddenly and drastically changed to a strong negative correlation over the following 4 decades (Figure 3a & b). A direct correlation using JJA values of NINO 3.4 for the period 1971-1990 gives a correlation value of -0.89 and Fisher's transformation gives a value in the range of -0.96 and -0.74 at 95% confidence level. Such unprecedented high correlations imply that ENSO events started to control rainfall anomalies outside the dry season from the early 1970s onwards. This effect is seen as a continuous pattern in DJF rainfall anomalies and as episodes in MAM rainfall anomalies (see Figure 3a&b). Note that DMI influences are void of such clear changes (results not shown). As a result of these prolonged ENSO influences, rainfall anomalies between the dry (JJA) and the wet (DJF) started to become highly correlated with each other from the early 1970s onwards (Figure 3c). Here the direct correlation for the 1970-2008 period gives a high correlation value in a range between 0.70 and 0.91 at 95% confidence level. Hence, the unpredictability of Indonesian's rainfall during the wet season indicated in past investigations[9,10] has changed to a fairly predictable state over the last 40 years. In addition to this are episodes in the 1980s and

from the 1990s onwards during which strong positive correlations even extend into the MAM period of the following year.

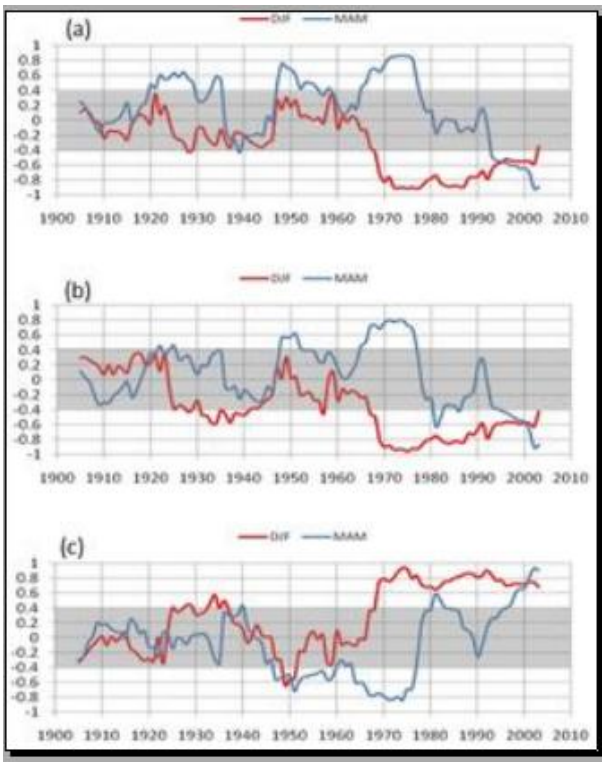

Figure 3. Sliding-window correlation (window size 11 yrs) between : a) JJA anomalies of NINO 3.4, b) SON anomalies of NINO 3.4, and c) JJA rainfall anomalies with average rainfall anomalies in the following DJF and MAM periods.

Conclusions

The rainfall decline in the Indonesian region in the last two decades has been preliminarily caused by two factors: 1) prolongation of ENSO influences controlling now rainfall anomalies in most seasons of the year and 2) extensive El-Niño-dominated episodes from the early 1990s onwards except for a strong and transient La-Nina intermission in 1999/2000. Whether this shift of rainfall dynamics in the Indonesian region will continue to exist into the future and whether it has been caused by global warming trends cannot be concluded with any statistical confidence. Nevertheless, findings of this research indicate a sudden and still ongoing shift in ENSO-related dynamics in the Indio-Pacific region that deserves further investigation.

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Agronomic Trait Evaluation of Transgenic Rice Line With *Db1* Transgene

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Abstract

Genetic improvement of rice crop resistance to brown planthopper is one of the main goals in rice breeding programs in Indonesia. To achieve this goal, Db1 transgene (Mannose-Binding Lectin Family Gene) driven by sucrose synthase-I promoter, which was isolated from Dioscorea batatas and proven to be effective against sup-sucking insect, had been succesfully transformed into rice genome cv.Taichung-65 by using Agrobacterium-mediated transformation and one homozygous line has been selected. However, plants derived from genetic transformation frequently show phenotypic abnormality, hence agronomic trait evaluation is required to clarify the occurrence of this phenomenon. The objective of this experiment was to obtain transgenic rice plants with normal phenotypes (cv. Taichung 65). Forty rice plants were used in this experiment, consist of 20 T_3 generation of transgenic rice plants cv. Taichung-65 and 20 non-transgenic rice lines of cv. Taichung-65 as check. Data were analyzed by student's t-test. Significant differences were found in heading date, days to panicle exertion, plant height, wet weight of panicle, dry weight of panicle, grain number per panicle, grain weight per panicle, and grain filled per hill. These results suggest that agronomic trait differences in transgenic plants caused by somaclonal variation during in-vitro culture and transformation events.

Keywords: agronomic trait, Db1 transgene, somaclonal variation, transgenic rice.

Introduction

Brown planthopper (*Nilaparvata lugens;* BPH) is one of the most dangerous and harmful pest for rice cultivation, especially in Southeast Asia and East Asia. This pest has become an important issue for farmers and decision makers. The pest attacks rice cultivation on large areas within a short time and may cause huge losses of rice yield even no yield (Baehaki, 2009). Due to this condition the effective way to control BPH is highly needed. Developing new rice lines resistant to BPH through transfer gene technology is one of promising effort to overcome the problem. The application of gene transfer technology has been recognized as the economical and environmentally sustainable approach because of reducing insecticide application (Maqbool *et al*., 2001).

Currently some genes have been utilized to developing resistant plants against insects (horizontal resistance) is the *Cry* gene, which isolated from *Bacillus thuringiensis* (*Bt*) expressing the *Bt* toxin (protein crystals that toxic to insects). Various crops such as rice, maize, sugarcane, tobacco, etc., have been transformed with a various types of the *Bt* genes (*cry1Aa*, *cry1Ab*, *cry1Ac*, *cry2A*, etc.); Rao *et al*., 1998; Maqbool *et al*., 2001; Rahman *et al*. , 2007).

The availability of cloned genes regulating resistant traits against some pests has become an opportunity to developing new cultivars. One of the potential genes for this purpose is *Db1* gene. *Db1* is a gene which was isolated from *yam bean*, known as *mannose-binding lectin* consist of some identic 12-kDa sub-units. The mannose-binding lectin is a protein that contributes to the immune response, which can bind to carbohydrates on a various of pathogens. Around 58% of *Db1* nucleotide have the same motif (homology) with *Snowdrop lectin Gna*, so it classified to *Gna-related lectin family* and this gene has been successfully utilized to increase resistance ability against insects in tobacco, maize, and rice (Gaidamashvili *et al*., 2004).

Based on these facts, it is worth if *Db1* gene can be applied to develop the transgenic rice plants with resistant ability against pests, especially brown planthopper. Transgenic rice cv. Taichung-65 expressing *Db1* gene and driven by *sucrose-synthase-1 (Rss1)* has been successful developed by Prof. Kinya Toriyama, Lab. of Environmental Plant Biotechnology, Tohoku University. The selected homozygous *Db1* line, especially driven by *Rss1* promoter, will be tested for BPH resistance in Indonesia (Toriyama, 2010).

However, before integrated into conventional rice breeding program in the future, agronomic trait evaluation of transgenic *Db1* must be performed in order to assess the occurrence of somaclonal variants by comparing it with its original cultivar (Taichung-65). The objective of this current experiment was to obtain transgenic rice line with agronomic with normal phenotypes. This is due to transgenic plants sometimes shown phenotypic variation on agronomic characters. Jiang *et al*. (2000) reported that some transgenic rice showed abnormal phenotypes as observed on necrotic leaves, dwarfism, reduced fertility, reduced panicle and grain fertility. This is because of the instability of the genome of transgenic plants which had been cultured by *in vitro* and the effect of gene transfer (Cassels and Curry, 2001).

Materials and Methods

The experiments were conducted at Transgenic containment, ICABIOGRAD, Bogor from July 2010 to January 2011. Forty rice plants consisted of 20 transgenic plants of cv. Taichung-65 with Db1 (T_3 generation) and 20 plants of non-transgenic of cv. Taichung-65 as the check. Transgenic plant was derived one T_2 plant that supposed to be homozygous based on detection of a single copy number of *Db1* transgene (Toriyama, 2010). Observations of agronomic characters were made at vegetative phase (4 and 8 weeks after planting) for plant height (cm) and number of tillers. At the generative phase (12 weeks after planting) observations were made for plant height, number of tillers, number of productive tillers, number of panicles per hill, chlorophyll content index. At harvesting date, the observations were made for panicle length, number of grains per panicle, number of filled grain per panicle, number of unfilled grain per panicle, weight of 1000 grains, dry weight of panicle, and grain weight per panicle. Observed data were then analyzed with student *t-test* using SPSS v.17.0 software.

Results and Discussion

Data is presented in Table 1 showed the significant differences of phenotypic appearance of heading date, days to panicle exertion, plant height, wet weight of panicle, dry weight of panicle, grain number per panicle, grain weight per panicle, and grain filled per hill showed when its compared with controls plants (cv. Taichung 65).

Tabel 1. Comparative performance of the transgenic plants and non-transgenic original plant

Genotype	Days to panicle exertion (dap)	Heading date (dap)	Plant height (cm)	Number of tillers	Number of productive tillers	Chlorophyll (CCI)	Number of panicles per hill	Panicle length (cm)	Wet weight of panicle (g)	Dry weight of panicle (g)	Grain number per panicle	Grain weight per panicle	Filled grain per hill	Unfilled grain per hill	Weight of 1000 grains (g)
GMO	106.4 ±0.32	109.4 ± 0.24	123.01 ± 0.95	11.32 ± 0.75	19.10 ± 1.40	20.20 ± 0.61	19.10 ± 1.40	21.22 ± 0.38	3.16 ± 0.18	2.86 ± 0.15	135.06 ± 5.26	2.75 ± 0.15	110.19 ± 5.96	25.20 ± 3.91	23.89 ± 0.32
Non-GMO	103 ± 0.31	106.35 ± 0.27	129.81 ± 0.74	12.58 ± 0.88	18.50 ± 1.07	18.95 ± 0.71	18.50 ± 1.07	21.78 ± 0.26	4.03 ± 0.16	3.64 ± 0.14	169.57 ± 4.95	3.62 ± 0.13	158.02 ± 11.83	23.89 ± 3.15	23.43 ± 0.342
Significancy	*	*	*	ns	ns	ns	ns	ns	*	*	*	*	*	ns	ns

Note: Data show mean values with standard error of means. Differences between mean values of transgenic and non-transgenic plants are non-significant (ns) or significant (*) by *Student's t-test* at 0.05 significance level.

Transgenic rice plants have the possibility of phenotypic changes in appearance due to the influence of environment and also due to somaclonal variation derived from *in vitro* culture as well as due to transgene insertion. Genetic and environmental factors could cause differences in agronomic characteristics such as the productive tillers number (Grist, 1965), loss of tillers number (Arraudeau and Vergara, 1992), plant height (Patnaik *et al*., 1999), and harvest age (Jiang *et al*., 2000). Somaclonal variation is estimated to be one that cause of this difference. Somaclonal variation is a genetic alteration that generated through tissue culture (Larkin and Scowcroft 1981; Scowcroft *et al*., 1985). Somaclonal variation can occur as a result of injuries on the target gene (wounding) by the transformation, tissue culture media, and tissue culture environment (Cassels and Curry, 2001).

According to Wattimena (1992) somaclonal variation derived from genetic diversity of explants and genetic alteration that occurs in tissue culture. Genetic diversity of explants is caused by mutated cells and the polisomic of specific tissues. Genetic alteration that occurs in tissue culture is caused by doubling the number of chromosomes (fusion endomitosis), changes in the structure of chromosomes (crossover), changes in gene and cytoplasm (Evans and Sharp 1986; Ahlowalia 1986). The alteration that occurs at genetic level has led to the phenotypic appearance changes in transgenic plants.

The significant differences can also be caused by environmental influences that explained by Zainal and Amirhusin (2005) who mentioned that the quantitative agronomic characters were observed visually influenced by the environment, and each genotype has a different response to the environmental carrying capacity. Environmental factors such as pests, diseases and temperature can also affect the trait of filled of grain. Temperatures above 35℃ during the period of anthesis until flowering can lead to sterility pollen (Satake and Yoshida, 1978 in Jagadish *et al*., 2007). In addition, the panicle is formed at the later tillers are usually small and too late to rippened, so that the grain only half-full or empty at harvest time (Arraudeau and Vergara 1992).

The non significant differences on traits between transgenic and non-transgenic plant showed by the number of tillers, number of productive tillers, chlorophyll content, number of panicles per hill, panicle length, unfilled grain per hill, and weight of 1000 grains. Grist (1965) states that the number of productive tillers in rice plants affected by genetic factors and environment. The lost number of productive tillers was affected by environmental factors caused by competition of tillers and nitrogen deficiency (Arraudeau and Vergara, 1992).

Conclusion

There were significant differences on some agronomic characters i.e. days to panicle exertion, heading date, plant height, wet weight of panicle, dry weight of panicle, grain number per panicle, grain weight per panicle, grain filled per hill. These results suggest that agronomic trait differences in transgenic plants caused by somaclonal variation during in-vitro culture and transgene insertion.

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Residual Effects of Vesicular Arbuscular Mycorrhiza and Bokashi on Growth and Yield of Cilembu Sweet Potato (*Ipomoea batatas* (L.) Lamb.)

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Abstract

An experiment studying the residual effects of Vesicular Arbuscular Myccorrhiza (VAM) on growth and yield of Cilembu sweet potato Eno cultivar was conducted at the Agricultural Experimental Site of Faculty of Agriculture Universitas Padjadjaran at Jatinangor from November 2003 to April 2004. The altitude of the location was 754 meters above sea levels. Randomnized Block Design was used with factorial pattern of two factors and three replications. The first factor was residual of VAM (m), which were consisted of four levels, m0 = 0 g/plant, m1 = 5 g/plant, m2 = 10 g/plant and m3 = 15 g/plant. The second factor was residual of chicken dung bokashi (b), which were consisted of three levels, b1 = 0 ton/ha, b2 = 5 ton/ha and b3 = 10 ton/ha. The result of the trial showed that in the young stages of sweet potato, bokashi was more influential than VAM. There were interaction effects between higher dosages of VAM residue with higher dosages of chicken dung bokashi residue on the tuber weight per plant. VAM residue at 10 g/ plant and bokashi residue at 10 ton/ha produced highest yield of sweet potato and was significant as compared to control (no applications).

Keywords: Mycorrhiza Vesicular Arbuscular (VAM), bokashi, Cilembu sweet potato, Inceptisol soil

Introduction

According to Indonesian statitical figures sweet potatoes are planted in about 0.4 million hectares or 3% of lands planted with food crops. National yield is only 7.5 ton/ha which is low. Soenarjo, 1984 stated that between 1968 to 1984 the hectarage planted with sweet potatoes were decreasing. Usually the dwindling of the hectarages were because of irrigation canal betterments and competition with more profitable crops. Until now there are no significant advancement of size as well as technologies on the growing of this crop. In 2006, 176,510 hectares of sweet potatoes yielded 10.51 ton/ha, and produced 1,854,238 ton. In 2008, 174,026 hectares yielded 10.64 ton/ha and produced 1,850,828 tons (Badan Pusat Statistik, 2008).

Farmer's average yield is between 7 to 10 ton/ha while from the experimental plots with ideal inputs of agriculture the yield could reach up to 20 or up to 30 ton/ha. Therefore increasing farmers' yield to 20 ton/ha is possible (Hendroatmodjo, 1984). Compared with Philippine farmers who yielding only 5.0 ton/ha, Indonesian farmer's yields are higher. The highest yield 25 to 30 ton/ha is obtained from Taiwan's farmers. Indonesian sweet potatoes were planted in less fertile soils and generally sweet potatoes is not considered as very important crop except in certain small areas where the very good quality varieties are grown.

Technically effort to increase yields could be obtained by using high quality cuttings, fertilizers and manures applications, good rearing and control of pests and diseases

(Soenarjo, 1984). Sweet potatoes is planted in 33 provinces in Indonesia, West Java produced the highest which was 375,708 tons in 2008 or 20.3% of national production and followed by Papua which produced 350, 964 tons (18.9%), East Java 136,556 tons (7.38%), Central Java 117,159 tons (6.33%) and North Sumatra 114,186 tons (6.17%) (Badan Pusat Statistik 2008).

In tropical countries in Africa, in India, and Indonesia, sweet potato is grown by poor peasants who grow this crop to fulfill their own additional carbohydrate needs. Hence it is looked upon as low prestige crop. But when it is grown as a modern industrial purposes as in the US, Japan and Taiwan, it becomes high value crop. Sweet potato is a source for producing substances for food and feed industries and raw materials for many industries. World sweet potato production was 124 million tons with 9.2 millions hectares (Huaman and Zhang, 1997).

Mycorrhiza is a symbiotic relationship between a fungus (*mykes*) and roots (*rhiza*) of a vascular plant (Setiadi, 1989). Bokashi is a fermented organic fertilizer using non pathogen microbes innovated by Teruo Higa from Ryukyus University of Japan. In this trial chicken dung bokashi was used.

Materials And Methods

Purpose of trial was to find out the residual effects of Vesicular Arbuscular Mycorrhiza (VAM) and Bokashi on growth and yield of Cilembu sweet potato (*Ipomoea batatas* (L.) Lamb.). Trial location was at the Agricultural Experimental Station, Faculty of Agriculture, Padjadjaran University, Jatinangor, Sumedang at the altitude of 754 meters above sea level. According to Schmidt and Ferguson (1951) the climate was C type with average precipitations of 2179 mm per year. Trial was commenced from November 2003 to April 2004. Materials used were sweet potato seedlings Eno variety originated from Cilembu village. Urea (45% N), SP-36 (36% P_2O_5), and KCl (60% K_2O). Insecticide used was Decis 2.5 EC. Agricultural tools used were hoes, small hoes with small handle (Kored, Sd), dibbling sticks, pegs, a metering tape, balances, paper envelopes, plastic bags, sags, knives, scissors, paper labels and rafia strings.

Randomnized Block design with two factors and three replications was used in this trial. The first factor was VAM residue dosages (m), the second factor was chicken dung bokashi residue dosages (b). VAM residue dosages were of four levels: m_0 = 0 g VAM/plant, m_1 = 5 g VAM/plant, m_2 = 10 g VAM/plant and m_3 = 15 g VAM/plant. Chicken dung Bokashi residue dosages were of three levels b_0 = 0 ton/ha Bokashi, b_1 = 5 ton/ha Bokashi, b_2 = 10 ton/ha Bokashi. There were 12 treatment combinations replicated three times to obtain 36 treatment plots. Plot size was 3.5 m x 2.8 m, there were four bunds in each plot, there were 14 plants on every bund. Plant population was 56 in each plot. Treatment combination is shown at Table 1.

Soil analysis conducted before trial showed that organic-C content was low (1.6%), N-organic medium (0.4%), C/N ratio low (4). Available P content was medium (16.1 mg/100 g), and K content was low (20 mg/ 100 g). Soil pH was acidic (pH 5.2). Sweet potatoes were tolerant to acidic soils since it pH live range is pH 4.5 to 7.5 (Onwueme, 1978). High cation exchange capacity (26.4 me/100 g) was able to absorb, retain and prepare the plant nutrient for sweet potatoes. VAM infections on plant roots was measured by counting the vesicles and arbuscles found in the plant roots. Analysis was conducted at the Soil Microbiology Laboratory Universitas Padjadjaran at Jatinangor is shown at Table 2.

Table 1. Treatment combination

VAM residue dosages	Chicken Bokashi residue dosages		
	b0 = 0 ton/ha Bokashi	b1 = 5 ton/ha Bokashi	b2 = 10 ton/ha Bokashi
m0 = 0 g VAM/plant	m0 b0	m0 b1	m0 b2
m1 = 5 g VAM/plant	m1 b0	m1 b1	m1 b2
m2 = 10 g VAM/plant	m2 b0	m2 b1	m2 b2
m3 = 15 g VAM/plant	m3 b0	m3 b1	m3 b2

Table 2. VAM infection grade (%)

Treatments	Chicken dung Bokashi residue dosages		
	b0 = 0 ton/ha Bokashi	b1 = 5 ton/ha Bokashi	b2 = 10 ton/ha Bokashi
VAM residue dosages	Infection (%)		
m0 = 0 g VAM/plant	0.0	1.0	1.3
m1 = 5 g VAM/plant	13.3	33.3	50.0
m2 = 10 g VAM/plant	20.0	36.7	56.7
m3 = 15 g VAM/plant	36.7	53.3	56.7

The same letters following figures within one column mean no significancy among treatments (Duncan's Multiple Range Test at 5% level)

Results and Discussions

VAM Infection on Roots

After VAM and bokashi residues were applied, there were eight combination tratments with less than 50% root infections and four with more than 50%. There were infected roots on the treatments without VAM residue. These were indications that the roots were infected by indigenous VAM. Bokashi residues improved conditions which enable VAM to infect root cells after which mutualistic symbiosis would happen between VAM and roots cells. According to Gunawan (1993) in the wet season water would stimulate spores to grow and infect root cells. Percentage of infections are shown at Table 2.

Vine Length

Analysis results showed that there were no interaction between VAM residue and Bokashi residue on the vine length. Independent effects of VAM residue at 10 g/plant influenced vine length at 9 WAP, while Bokashi residue influenced vine length at 6, 9, 12, 15 and 18 WAP were shown at Table 3.

Leaf Area Index

Analysis results as shown at Table 4 stated that there were no interaction between VAM residue and bokashi residue on the leaf area index. Effects of VAM residue were non sigficicant influence on the leaf area index, while bokashi residue influenced significantly on the leaf area index at 15 WAP only as compared to control (no application).

Table 3. Vine lenght (cm). WAP = weeks after planting

Treatments	Vine Length Averages				
	6 WAP	9 WAP	12 WAP	15 WAP	18 WAP
VAM residue dosages					
m_0 = 0 g VAM/plant	110.48 a	172.30 a	214.33 a	239.11 a	256.30 a
m_1 = 5 g VAM/plant	98.00 a	150.96 a	200.52 a	235.81 a	253.89 a
m_2 = 10 g VAM/plant	132.07 a	209.78 b	246.41 a	280.52 a	302.74 a
m_3 = 15 g VAM/plant	124.89 a	182.19 ab	245.04 a	268.96 a	313.93 a
Bokashi residue dosages					
b_0 = 0 ton/ha Bokashi	97.78 a	148.14 a	201.11 a	228.36 a	258.78 a
b_1 = 5 ton/ha Bokashi	116.78 ab	181.03 b	224.64 ab	254.36 b	270.08 ab
b_2 = 10 ton/ha Bokashi	134.53 b	207.25 b	253.97 b	285.58 b	316.28 b

The same letters following figures within one column mean no significancy among treatments (Duncan's Multiple Range Test at 5% level)

Table 4. Leaf Area Index

Treatments	Leaf Area Index Averages				
	6 WAP	9 WAP	12 WAP	15 WAP	18 WAP
VAM residue dosages					
m_0 = 0 g VAM/plant	1.55 a	2.60 a	1.70 a	2.26 a	3.12 a
m_1 = 5 g VAM/plant	2.10 a	2.24 a	2.27 a	2.48 a	3.25 a
m_2 = 10 g VAM/plant	2.15 a	1.79 a	2.24 a	2.51 a	2.75 a
m_3 = 15 g VAM/plant	2.02 a	2.22 a	1.64 a	2.15 a	3.46 a
Bokashi residue dosages					
b_0 = 0 ton/ha Bokashi	2.04 a	1.88 a	2.04 a	1.67 a	2.56 a
b_1 = 5 ton/ha Bokashi	1.80 a	2.40 a	1.99 a	1.80 a	3.40 a
b_2 = 10 ton/ha Bokashi	2.02 a	2.36 a	1.87 a	3.59 b	3.47 a

The same letters following figures within one column mean no significancy among treatments (Duncan's Multiple Range Test at 5% level)

Tuber Numbers / Plant

Analysis on the tuber number per plant at Table 5 showed that there were no interaction between effects of VAM residue and bokashi residue. VAM residue and bokashi residue were independent and were not significant on the tuber number / plant as compared to control.

Tuber Weight / Plant

Table 6 showed that without bokashi residue, addition of VAM up to 15 g/plant didn't give any significancy on the tuber weight/plant and vice versa, without VAM residue addition of bokashi up to 10 ton/ha were not significant as compared to control (no bokashi nor VAM applications). In the treatments where bokashi and VAM were increased, they were showing gradual increase of significancy as well. Application bokashi 10 ton/ha and VAM at 10 g/plant resulted highest tuber weight/plant ($m_2 b_2$).

Table 7 showed that sweet potato yields can be obtained by applications of more inputs for instance VAM and bokashi. In this case application of 10 g VAM residue/plant plus bokashi residue at 10 ton/ha were the highest yield 25.41 ton/ha.

Table 5. Tuber Numbers / Plant

Treatments	Tuber Numbers/Plant Averages
VAM residue dosages	
m_0 = 0 g VAM/plant	1.56 a
m_1 = 5 g VAM/plant	1.40 a
m_2 = 10 g VAM/plant	1.59 a
m_3 = 15 g VAM/plant	1.56 a
Bokashi residue dosages	
b_0 = 0 ton/ha Bokashi	1.50 a
b_1 = 5 ton/ha Bokashi	1.53 a
b_2 = 10 ton/ha Bokashi	1.56 a

The same letters following figures within one column mean no significancy among treatments (Duncan's Multiple Range Test at 5% level)

Table 6. Interaction effects VAM residue and bokashi residue on the tuber weight/plant (g)

VAM residue dosages	Chicken bokashi residue dosages		
	b_0 = 0 ton/ha Bokashi	**b_1 = 5 ton/ha Bokashi**	**b_2 = 10 ton/ha Bokashi**
m_0 = 0 g VAM/plant	177.22 a A	191.11 a A	210.33 a A
m_1 = 5 g VAM/plant	165.22 a A	220.00 a A	383.89 bc B
m_2 = 10 g VAM/plant	184.89 a A	343.89 b B	444.78 c C
m_3 = 15 g VAM/plant	163.89 a A	338.33 b B	326.67 b B

Average values coded with the same small letters in the same columns were not significant according to Duncan's Multiple Range Test 5% level.
Average values coded with the same capital letters in the same rows were not significant according to Duncan's Multiple Range Test 5% level.

Table 7. Tuber yield (ton/hectare)

VAM residue dosages	Chicken Bokashi residue dosages		
	b0 = 0 ton/ha Bokashi	**b1 = 5 ton/ha Bokashi**	**b2 = 10 ton/ha Bokashi**
m0 = 0 g VAM/plant	10.12	10.92	12.01
m1 = 5 g VAM/plant	9.44	12.57	21.93
m2 = 10 g VAM/plant	10.56	19.65	25.41
m3 = 15 g VAM/plant	9.36	19.33	18.66

Average values coded with the same small letters in the same columns were not significant according to Duncan's Multiple Range Test at 5% level.
Average values coded with the same capital letters in the same rows were not significant according to Duncan's Multiple Range Test at 5% level.

Conclusions

Bokashi residue influenced vine length at 6, 9, 12, 15 and 18 WAP. Bokashi residue influenced significantly on the leaf area index at 15 WAP as compared to control, no application. Bokashi residue was more influential on the vine growth and leaf area index than VAM residue.

In the treatments where dosages of bokashi and VAM were increased, they were showing gradual increase of significancy as compared to control. Giving bokashi to 10 ton/ha and VAM at 10 g/plant resulted highest tuber weight/plant ($m_2 b_2$) or equal with 25.41 ton/ha. Control treatments only yield 10.12 ton/ha.

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Reformation of Shifting Cultivation Farming System Towards Permanent and Sustainable Cultivation

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Abstract

Cultivation of estate crops especially rubber and oil palm is believed to be able to increase farmer's income and reduce shifting cultivation significantly. Cultivating estate crops, enables peoples to gain sufficient income to fulfill their basic needs. Establishment of rubber and oil palm require investment cost Rp 34.39 million/ha and Rp 33.71 million/ha respectively. Estimation of net present value (NPV) which is going to be obtained at 12.5% interest rate/year is Rp 44.86 million/ha and Rp 45.02 million/ha, whereas expected internal rate of return (IRR) is 22,92% and 24,46% for rubber and oil palm respectively. Hence, farmers would gain monthly net income Rp 1.36 million/ha and Rp 1.14 million/ha for rubber and oil palm respectively. This confirms that rubber and oil palm plantation are feasible to establish. In order to assure the reformation process from shifting cultivation towards sustainable and permanent cultivation goes effectively, thorough planning of target, identification about existing condition, anticipation about constraints that might hinder the process and formulation of operational pace need to be determined clearly beforehand. This could be done through multi stakeholder's participation.

Keywords: estate crops, participation, permanent cultivation sustainable, reformation, shifting cultivatio,.

Introduction

Shifting cultivation (SC) is deemed to be the earliest subsistent farming system ever known and inherited since ancient times. Mering (1990) in Arkanudin (2009) stated that such cultivation system has been practiced by people in Kalimantan since 6000 years BC. Main characteristics of the system are slash, cut and burn in land preparation. After cultivated for several years, it will be followed by relatively longer fallow period.

In the past, when land availability was still abundant, this cultivation technique was relatively save and sustainable. In the system, short period of cultivation (usually 1 – 3 years) would be followed by fallow period which was relatively much longer (7– 10 years). Fallow period enabled the land to recover its soil fertility naturally. However, due to rapid population growth together with limited land availability and increase of basic economic needs have induced drastic change of the system in which short period of cultivation is followed by short fallow period. In addition, in many cases, the system has been manipulated by some imprudent people to camouflage their illegal logging activities. As a result, the situation has generated forest fire events more frequently and instigates wider critical land. Therefore, it is urged to ameliorate the system towards more sustainable and permanent cultivation. Cultivation of estate crops especially rubber and oil palm is believed to be able to increase farmer's income and reduce shifting cultivation significantly. The purpose of the paper was to give a description about investment cost and feasibility of rubber and oil palm plantation establishment in order to develop sustainable and

permanent farming system. Data and information presented in the paper were taken from literature study.

Materials and Methods

Data and Assumptions Persist

This is a literature study through collection of data and information from Directorate General of Estate Crop, Statistic Central Bureu, Oil World dan other research institutes and publications. Collection of data and information was done from January – July 2011. Considering variations of collected data, several data adjustments based on inflation and exchange rate were made in data analysis and processing. Several limitations and assumptions used in this analysis:

a. Calculation is based on one hectare rubber or oil palm plantation investment.
b. Productivity of oil palm plantation uses production pattern of third classification area i.e. 20.33 ton FFB (fresh fruit bunches)/ha/year. Whereas productivity of rubber/ha is assumed 1,300 kg dried rubber/year.
c. Price of FFB fluctuate from Rp 600/kg – Rp 2000/kg, its price used is Rp 1300/kg
d. Price of raw rubber at farmer level in the last few years fluctuate from Rp 5000 – Rp 17 000 hence, its price used is Rp 11000/kg
e. Labour cost is Rp 50,000/man-day
f. Financial analysis is based on one rotation period (30 years for rubber and 25 years for oil palm) starts from 2011 with 12.5% interest rate/year.

Data Analysis

All collected data and information was processed and analyzed by using financial analysis based on several feasibility criteria i.e. Net Present Value (NPV), Internal Rate of Return (IRR) and Benefit Cost Ratio (B/C ratio) which is formulated as follow (Gittinger,.1986):

a. $$NPV = \sum_{t=0}^{n} \frac{Bt - Ct}{(1+i)^t} \quad \text{..... (1)}$$

where:
NPV = Net Present Value,
Bt = benefit or revenue at year t,
Ct = cost at year t,
i = interest rate of Bank,
n = period of project.
One project is considered to be financially feasible if NPV is > 0.

b. $$IRR = i1 + \frac{NPV1}{NPV1 - NPV2}(i2 - i1) \quad \text{..... (2)}$$

where:
IRR = Internal Rate of Raturn,
i1 = interest rate to produce NPV1 positive close to zero,
NPV1 = value of NPV close to zero positive,
i2 = interest rate to produce NPV2 negative close to zero,
NPV2 = value of NPV close to zero positive.

One project is considered to be financially feasible if IRR value is higher that interest rate from Bank.

c. $$B/C = \frac{\sum_{t=0}^{n} \frac{Bt}{(1+i)^t}}{\sum_{t=0}^{n} \frac{Ct}{(1+i)^t}}$$ ..... (3)

where:

B/C = Benefit Cost Ratio,
Bt = benefit or revenue at year t,
Ct = cost at year t,
i = interest rate,
n = period of project

One Project is considered to be financially feasible if B/C ratio > 1.

Result and Discussion

General Condition of Shifting Cultivation

Until now there was no accurate data about how many households actually practice shifting cultivation and how large area has been used for such farming system. Food and Agricultural Organization (1955) in Dove (1988) estimated there was about 10 % of world population over one third of world agricultural land applied SC. Subsequently, according to Spencer in Dove (1988), in 1966 in Indonesia there were almost 4 million people practicing SC over an area of 85 million ha. Next, Food and Agricultural Organization (1990) in Barchia (2009) estimated that number of population involved SC in 1990 was 6.39 million covering an area of 12.24 million ha and in 2010 it increased to become 8.65 million people covering 16.44 million ha. Although data and information about number of people who practice and area of shifting cultivation are very limited, the system continuous developing in many parts of Indonesia. Consequently, its impact as a result of slash and burn an activity tends to increase also, especially in dry season. Slash and burn are two main activities applied in the system.

As cultivation system inherited since long time ago, SC has become a culture/tradition for the people living outside Java Island like Sumatera, Kalimantan, Sulawesi and Papua. Practice of shifting cultivation in those islands is almost similar, started with slash, cut and burn. Planting usually is done for 1 – 3 year period (depend on soil fertility) and then followed by fallow period which is relatively longer than planting period i.e. between 7 – 10 years before it is recultivated again.

According to Banyamine (2009), shifting cultivation is an integrated and continues agricultural system in time and space. It is done by shifting/moving from one area to another as a main characteristic of local knowledge/wisdom. Fallowing period is meant to give the land an opportunity to take a break for certain period (10 – 15 years) before recultivate in the next rotation period.

In West Kalimantan, SC has been practicing by Dayak tribe since long time ago using simple equipments like adze or chopping knife. It has been done very restrictly and followed common knowledge i.e. forest is not an asset or treasury but is “common home” for all the people living in and around it. Therefore several accusations directed to them which

blamed those people for the vanish/extinct of natural forest in the area is really unreasonable. In utilizing natural resources including surrounding forest, Dayak people always base on their religious system that lead them to be always in harmony with dynamics of universe (Arkanudin, 2009).

Subsequently, Arkanudin (2009) also notified that for Dayak people (who practice SC) mostly was subsistence community. For them, productivity in SC activities is not their first priority. Crop diversity and sustainability are much more important than that. As symbol of land ownership, most of them used to plant fruit or rubber trees on their land. The symbol declares that the land will be recultivated in the next rotation period.

In 1990, World Bank and FAO stated that main cause of deforestation is slash and burn farming system. However, further analysis showed several evidences and proved that judgment which mentioned that shifting cultivation is the main cause of deforestation is misleading. Since then, many began to realize that development of wood industrial sector in many parts of Indonesia, especially after most of forest area in the country has been allocated to concessionaires actually the main cause of deforestation in Indonesia. Many remote areas which has never been able to be accessed before became accessible as lot of new infrastructure (road, bridge, drainage facilities and others) was built to enable concessionaires operate their work. Indirect impact of this situation is the remote area becomes accessible also for local community hence their area for SC also becomes wider and farther than before. Other indirect impact also persists i.e. lots of new comers (job seekers) came to the area and some of them followed local community to practise slash and burn cultivation. As a result, forest fire incidences and natural devastation increased due to hasty implementation of slash and burn activities done by those new comers. (Sunderlin and Resosudarmo, 1997).

Many efforts have been developed by the Government especially Ministry of Forestry, Ministry of Transmigration, Ministry of Agriculture, Ministry of Home Affairs and Ministry of Social Affairs to control the expansion of SC. Several guidance and counseling for community practicing SC through different programs such as Rehabilitation program, Timber Estate Program, Rice field construction, Transmigration Program, Nucleus Estate Smallholder Program and Resettlement Program have been widely implemented in many parts of Indonesia (Sulistyo, 2011). Some of those efforts show satisfying results but most failed. On example of success program to control shifting cultivation is development of Smallholder Rubber Development Project (SRDP) and oil palm Nucleus Estate Smallholder (NES) in Sanggau Regency, West Kalimantan. Rubber and oil palm plantation managed by shifting cultivation household could contribute significant income for the farmers. Total income gained from rubber and oil palm cultivation is Rp 24.82 million (72% of total family income) and Rp 27.98 million (63% of total family income) respectively (Agus, et.al, 2010). Success story from Sanggau through development of estate plantation by involving SC household should be a pilot model for controlling SC in other parts of Indonesia.

Prospect of Estate Plantation

Estate commodity, especially rubber and oil palm are very potential to be developed as world demand for these products tends to increase intermittently. Consequently, price of these commodities also increase considerably in the last few years, and it is predicted that its price would be stable in the next few years.

Study on Task Force Rubber Eco Project (REP) by International Rubber Study Group (IRSG) in 2005 projected that demand for natural and synthetic rubber in 2035 would be 31.3 million ton and 15 million ton of it is natural rubber. Production of natural rubber in 2005 was 8.5 million; hence there is a need to increase production 6.5 million ton in 2035. Therefore it is urged to have an effort for large rejuvenation and expansion of plantation. Subsequently, price of natural rubber for standard Indonesia rubber-20 (SIR 20) has significantly increased from US $ 1/kg in 2002 to become US $ 1.90 in 2005. In 2007, it is predicted it will reach US $ 2.00/kg and it will always be constant in the long run until 2020 (Anwar, 2006).

Average yearly increase of demand for world palm oil during 1993 – 2006 was 8.1 %, whereas its yearly increase of production was only 7.9 %. As a result, crude palm oil (CPO) price increased also significantly from US $ 400/ton in 2005 to become US $ 1,000/ton at the beginning 2008 (Directorate General of Estate Crop, 2008). Subsequently, Food and Agricultural Policy Research Institute (2011) declared that demand for world palm oil would increase from 47.1 million ton/ha in year 2010/2011 to become 70.5 million ton in 2025/26. While world CPO price will continued increasing from US $ 946/ton 2010/2011 to become US $ 1,163/ton in 2025/2026.

Feasibility Analysis of Estate Crop Business

Investment needed to establish rubber plantation or oil palm plantation varies, depend on land condition, use of input (fertilizer), seedling, other production inputs and estate management model. Based on technical standard, for establishing 1 ha of estate crop plantation of rubber and oil palm is Rp 34.39 million/ha and Rp 33.71 million/ha respectively.

Table 1. Cost and Financial Analysis of Rubber Plantation

Investment Cost			
Description	**(Rp/ha)**	**Description**	**(Rp/ha)**
Pr H-0	12,527,500	Pr H-4	4,126,500
Pr H-1	5,081,000	Pr H-5	4,376,500
Pr H-2	4,114,000		
Pr H-3	4,164,000	Total Investment	34,389,500
Operational Cost			
Description	**(Rp/ha)**	**Description**	**(Rp/ha)**
Po H min	7,015,000	Po H max	10,540,000
Po H average	9,138,575		
Result of Financial Analysis			
NPV (Rp)	44,859,490	B/C	1.7657
IRR (%)	22.92	Income (Rp/ha/month)	1,364,446

Note: Pr H: Pre harvesting; Po H: Post harvesting

Table 2. Cost and Financial Analysis of Oil Palm Plantation

Investment Cost			
Description	**(Rp/ha)**	**Description**	**(Rp/ha)**
Pr H-0	15,085,000	Pr H	5,492,500
Pr H-1	7,343,250		
Pr H-2	5,791,250	Total Investment	33,712,000
Operational Cost			
Description	**(Rp/ha)**	**Description**	**(Rp/ha)**
Po H	5,492,500	Po H max	15,085,000
Po H average	8,959,356		
Result Financial Analysis			
NPV (Rp)	45,019,940	B/C	1.2437
IRR (%)	24.46	Income (Rp/ha/month)	1,135,872

Note: Pr H: Pre harvesting; Po H: Post harvesting

With total investment cost Rp 34.39 million/ha, added by operational cost for rubber plantation which varies between Rp 7.01 – Rp 10.54 million/ha, within 25 years by using 12.5% interest rate, it would earn net present value (NPV, df=12.5%) Rp 44.86 million/ha, internal rate of return (IRR) 22.92 and Benefit Cost Ratio (B/C) 1.77. This figure indicates that this investment is feasible to be developed. From this revenue, rubber farmer would get average monthly income Rp 1.36 million/ha (Table 1).

For establishment of oil palm estate plantation, it would require Rp 33.71 million/ha for investment and operational cost which vary between Rp 5.49 – Rp 15.09 million/ha. Within 22 years, it will earn net present value (NPV df=12.5%) Rp 45.02 million/ha, IRR = 24.46 and B/C = 1.24 which means feasible to be developed. Average monthly income which would be gained by farmer is Rp 1.14 million/ha (Table 2).

Reformation of Shifting Cultivation

Considering existing condition and land use tendency in the last 20 years, it is estimated that deforestation and forest degradation will keep continuing. Shifting cultivation practice and total critical land will remain increasing as well. If accurate solution for the problem is not prepared soon, total forest area in Indonesia will reduce drastically and eventually it will vanish someday. On the other side, prospect of estate plantation business is very potential and feasible to be established as alternative solution for shifting cultivation management. It seems that estate plantation by involving shifting cultivation household would be one possible answer for the problem. Therefore, if government is really intending to manage SC practice and reduce critical land while improving social economic condition of the people, a thorough and comprehensive planning of estate crop establishment is urged to be well formulated.

In planning design, there are several process need to be accomplished i.e. process of target determination, formulating existing condition, identification of influencing factors and organizing actions needed to achieve target determined before. Planning process is

initiated by target plotting. In this phase, related stakeholder especially local/regional government decide target of SC area which is going to be managed within next 5 – 10 years.

Next, local government as decision maker should identify general condition of land use in the target area and then identify supporting factors and anticipate difficulties that might hinder success of target achievement.

Last but not least, rapid action needed is defining several possible alternatives to accomplish target which are going to be achieved and select best alternative (the most adaptive, beneficial and sustainable) which could provide optimal result. To accomplish those action, all stakeholders concerning shifting cultivation management (investor, central and local government, industry, etc) should coordinate and work together. Special workshop to formulate planning of land use in the next 5 – 10 years need to be carried out to gain mutual understanding among stakeholders about the problem and strategic plan to solve it. It is also intended to accommodate expectations from each of them while identifying what contribution each of them might allocate for the success of the program.

Conclusion

Based on explanation described before, it could be concluded that SC either done by local people traditionally or by new comer which can be categorized as forest squatter has emerged several negative impacts both direct and indirectly for natural resources. Therefore it is very urged to manage and direct this farming system towards permanent and sustainable cultivation. Establishment of rubber and oil palm plantation is one promising alternative of permanent cultivation which is feasible to be developed. In order to guarantee reformation process from shifting cultivation towards permanent and intensive cultivation runs smoothly, it is essential to formulate comprehensive planning including identification of target and existing land use condition, complete information about supporting factors and obstacles that might hider target achievement and formulating operational activities. Planning preparation should be done by involving related stakeholder and carried out in particular workshop.

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The Abilities of Endophytic Fungi from Tomato Roots in Suppressing Root Knot Nematodes (*Meloidogyne* spp.) in Tomato

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Abstract

Endophytic fungi are microorganisms that inhabit the internal plant tissues without causing any apparent disease. The endophytic fungi may have beneficial effects on the host through their abilities in suppressing plant diseases. This paper discusses the abilities of endophytic fungi isolated from tomato roots in suppressing root knot disease intensity and population of Meloydogyne spp.. The endophytes were isolated from roots of tomato obtained from several areas in West Java (Jatinangor, Sumedang and Lembang, West Bandung). Concerning that some latent pathogens might be isolated as endophytes, the isolates were examined for their pathogenecity to tomato seedlings. The non pathogenic isolates were examined for their suppressive effects on Meloidogyne spp. in tomato. The results showed that from 7 isolates tested, 6 isolates reduced the numbers of galls by 50.1 - 69.2 %. All endophytic fungal isolates suppressed the population of second-stage juvenile of Meloidogyne spp. in the soils by 55.3 – 76.9 %. Three isolates which were CL1 (Chaetomium sp.), FL3 (Fusarium sp.) and FL4 (Fusarium sp.) increased the growth of treated tomato.

Keywords: endophytic fungi, Meloidogyne, root knot disease, tomato

Introduction

Root knot nematode (*Meloidogyne* spp.) is one of limiting factors in tomato production. The nematode infection resulted in formation of galls/swelling of the infected roots. The galls formation leads to inhibition of water and nutrients absorption. Aboveground symptoms include growth inhibition and fewer, small, pale green, or yellowish leaves that tend to wilt in warm weather. Severe infection particularly in young plants may cause stunting (Agrios, 2005; Luc, 1995).

Based on their pathogenecity *Meloidogyne* spp. are considered as sedentary endoparasites. The nematode inocula include eggs and second-stage juvenile (juvenile II). Each female lays approximately 500 eggs in gelatinous substances. The first-stage juvenile (inside the eggs) will emerge from the egg, that are considered as second-stage juvenile, which will penetrate the root tip of their host plants and become sedentary and feeds on the surrounded cells by means of its stylet. The nematode secretes saliva into the cells that stimulates cell enlargement. The nematode then undergoes a second molt and become third-stage, then fourth-stage juvenile. After the final molt the worm-like adult male emerges from the root and becomes free-living in the soil, while the female (like pear shaped) continues to parasite the roots (Agrios, 2005; Luc, 1995).

The nematode can be controlled by pesticide application into the soil. However, the increase in awareness on environmental issues has encouraged the development of environmentally-friendly control measures, such as biological control. Microorganisms that

are usually used for biological control agents of plant parasitic nematodes are rhizosphere microorganisms. Microorganisms that are also potential as biological control agents include endophytes which are microorganisms that reside the internal plant tissues without inciting any disease symptom such as endophytic fungi (Backman & Sikora, 2008).

The endophytic fungi have been found in many plants investigated (Stone *et al*., 2004; Istifadah & Suganda, 2010). Colonisation of endophytic fungi in the roots was more prominent rather than within the leaves (Boyle *et al*., 2001). Even though some endophytes may have no influence on the host plants or even as latent pathogens, many of them benefit the host plant through their antagonistic effects on plant pathogens (Backman & Sikora, 2008; Istifadah & Suganda, 2010).

The abilities of endophytic fungi in suppressing plant pathogens, including plant parasitic nematodes have been reported (Sikora *et al*., 2008) . The endophytic fungi have been reported to have antagonistic effects on burrowing nematode *Radophulus similis in vitro* (Athmann *et al*., 2006) and reduced its population in the soil (Pocasangre *et al*., 2000). The endophytic fungi have been reported to produce secondary metabolites toxic to plant parasitic nematodes such as *Meloidogyne* spp. (Hallmann & Sikora, 1996; Lingga, 2010) and *Globodera rostochiensis* (Istifadah *et al*., 2010) in vitro. This paper discusses the abilities of endophytic fungi from tomato roots in suppressing the root knot nematode (*Meloidogyne* spp.) in tomato.

Materials and Methods

The endophytic fungi were isolated from roots of wilds tomato from Jatinangor, Sumedang and organic tomato from Cibodas, Lembang, West Bandung. The root sample were washed under running tap water and then cut into 1.5-2 cm segments. The root segments were surface sterilized by submersion in 96 % ethanol for 1 minute, followed by submersion in solution containing 2 % chlorine for 3-5 minutes and submersion in 96 % ethanol for 30 seconds. The segments were then plated out on *Potato Dextrose Agar* (PDA) supplemented with chloramphenicol (50 mg l^{-1}) and incubated in room temperature (20 ^{o}C). Contamination by saprophytic fungi was checked by imprinting some segments on PDA before culturing. Mycelia that emerged from the root segments were sub-cultured to obtain pure cultures.

Endophytic fungi were inoculated into tomato seeds by placing the surface sterilized seeds on the endophyte isolate culture (Narisawa *et al*., 1998). The germinated seeds were planted into plastic cups containing mixture of sterilized soil mixed with rice husk charcoal (1:1, v/v). To ensure the colonization, the endophyte isolates were inoculated again by drenching 3 ml of the fungi spore suspension (spore density 10^{7}/ml) per planting site. The experiment was arranged in Completely Randomized Design with treatments of the endophytic fungal isolates, positive control (tomato inoculated with the nematode) and negative control (uninoculated tomato). Each treatment was repeated three times.

Inoculum of *Meloidogyne* spp. was prepared by extracting the nematode eggs from root galls. The root galls were cleaned with running tap water, cut into 0.5-1 cm in length and then soaked in NaOCl 0.1% for 5 minutes. The suspension was screened with series of screens with different pore size (750 µm, 50 µm and 35 µm pore screens). The debris left on the 50 µm and 35 µm screens were collected in beaker glass. The nematode inocula (eggs and juvenile II) per ml were counted using counting chamber under binocular microscope.

The tomato seedlings (treated and untreated) were transplanted into polybag and inoculated by *Meloidogyne* spp. inocula (2000 eggs per plant) at 2 weeks after transplanting. For nematicide treatment, the nematicide with active ingredient carbofuran was applied at dosage, 2 g/plant.

The numbers of galls in tomato roots were observed 5 weeks after nematode inoculation. The number of second-stage juvenile per 100 g soil was observed. The juvenile was isolated/extracted from soil sample by Baermann methods (Luc *et al*., 1995). The other parameters observed were fresh and dry weight of tomato shoots.

To confirm the antagonistic effects of the endophytic isolates on *Meloidogyne* spp., in vitro experiment was also conducted. The endophytic fungi were cultured in potato dextrose liquid for 2 weeks. The filtrate was separated by centrifugation at 8000 rpm for 3 minutes. The nematode suspension was added to the filtrate (1:1, v/v) and incubated for 24 hours. For check, the nematode was soaked in sterile water for 24 hours.

Results and Discussion

Isolation of endophytic fungi from roots of tomato resulted in 7 non pathogenic isolates. Most of endophytic fungal isolates examined reduced the numbers of galls in tomato roots by 50.1- 69.2 % (Table 1). The abilities of endophtic fungi in inhibiting the development of galls, maybe related to the colonization of the endophyte in the root tissues and their abilities to produce toxic metabolites. In the in vitro experiment, culture filtrate of all endophytic fungal isolates examined killed the second-stage juvenile of *Meloidogyne* spp. soaked in the filtrate for 24 hours. *Chaetomium* spp. are known to produce a range of antimicrobial secondary metabolites (Cole, 2003; Istifadah *et al*., 2006). Non pathogenic endophytic *Fusarium* was also reported to produce secondary metabolites that were toxic to *Meloidogyne* spp. (Halmann & Sikora, 1996; Lingga, 2010) or *G. rostochiensis* (Istifadah *et al*., 2010). Sedentary endoparasitic nematode such as *Meloidogyne* spp. were more sensitive to the secondary metabolite produced by the endophytes rather than migratory nematode (Halmann & Sikora, 1996; Kimmons *et al*., 1990).

Table 1. Numbers of galls in tomato roots inoculated by *Meloidogyne* spp. in various treatments

Treatments	Number of gall	Level of suppression (%)
Isolate CL 1 (*Chaetomium* sp.*)*	5.3 ab	69.2
Isolate CL 2 (*Chaetomium* sp.)	6.0 abc	65.4
Isolate FL 1 (*Fusarium* sp.)	8.7 bc	50.1
Isolate FL 2 (*Fusarium* sp.)	5.3 ab	69.2
Isolate FL 3 (*Fusarium* sp.)	6.0 abc	65.4
Isolate FL 4 (*Fusarium* sp.)	10.7 cd	38.5
Isolate FJ (*Fusarium* sp.)	5.3 ab	69.2
Nematicide (Carbofuran)	3.3 a	80.7
Check	17.3 d	-

Note : Data in each column followed by different letters were significantly different ($P < 0.05$) based on Duncan's Multiple Range Test (DMRT)

The endophytic fungi also reduced the numbers of second-stage juvenile in 100 g soil by 55.3 – 77.7% (Table 2). The reduction of the juvenile numbers in the endophyte treatments, possibly related to the reduction of the numbers of galls and also the inhibition of egg mass

production by the female nematode. Kimmons *et al*. (1990) reported that endophytic fungus, *Acremonium coenophialum* inhibited the production of egg mass of *Meloidogyne* spp. by 90%.

Table 2. Numbers of second-stage juvenile of *Meloidogyne* spp. in 100 ml soil in various treatments

Treatments	Numbers of juvenile II	Level of suppression (%)
Isolate CL 1 (Chaetomium sp.)	273.3 a	62.6
Isolate CL 2 (Chaetomium sp.)	293.3 a	59.9
Isolate FL 1 (Fusarium sp.)	168.9 a	76.9
Isolate FL 2 (Fusarium sp.)	288.9 a	60.5
Isolate FL 3 (Fusarium sp.)	326.7 a	55.3
Isolate FL 4 (Fusarium sp.)	264.4 a	63.8
Isolate FJ (Fusarium sp.)	206.7 a	71.7
Nematicide (Carbofuran)	162.2 a	77.8
Check	731.1 b	-

Note : Data in each column followed by different letters were significantly different ($P < 0.05$) based on Duncan's Multiple Range Test (DMRT)

In this study, some endophytic fungal isolates increased the growth of tomato plants (Table 3). Among the isolates examined, isolate CL1 (*Chaetomium*), FL 3 (*Fusarium* sp.) and FL 4 (*Fusarium* sp.) increased the shoot fresh and dry weight of treated tomato plants, 2.0 - 2.6 times compared to the check. Some endophyte isolates have been reported to promote the growth of their host plants (Istifadah *et al*., 2008; Istifadah, 2010; Istifadah & Suganda, 2010; Varma *et al*., 1999) as they can produce phytohormone (Hyakumachi & Kubota, 2003; Varma *et al*., 1999).

Table 3. Growth of *Meloidogyne* spp.-inoculated tomato plants in various treatments (35 days after inoculation)

Treatments	Shoot fresh weight (g)	Shoot dry weight (g)
Isolate CL 1 (*Chaetomium* sp.*)*	14.0 d	0.89 d
Isolate CL 2 (*Chaetomium* sp.)	13.9 d	0.45 abc
Isolate FL 1 (*Fusarium* sp.)	6.6 abc	0.39 abc
Isolate FL 2 (*Fusarium* sp.)	3.9 a	0.28 ab
Isolate FL 3 (*Fusarium* sp.)	14.3 d	0.71 cd
Isolate FL 4 (*Fusarium* sp.)	12.3 cd	0.74 cd
Isolate FJ (*Fusarium* sp.)	8.7 bcd	0.59 bcd
Nematicide (Carbofuran)	6.3 abc	0.25 a
Check	5.3 ab	0.36 ab

Note : Data in each column followed by different letters were significantly different($P < 0.05$) based on DMRT.

In this study, the endophytic fungi showed their abilities to suppress the nematode infection and population in the soil. The fungi are potential to be developed as biological control agents. However, formulation and delivery methods of the best isolates are still required to be further studied.

The use of endophytes in biological control has several advantages. The colonization of the endophyte in the host tissues in advance to the pathogen allows them to prevent the host

tissue from the pathogen infection. In addition, as resident of plant tissues, the endophytes are also protected from the fluctuating environment and lack of nutrition.

Conclusion

Among seven non pathogenic isolates of endophytic fungi from tomato roots, there were six isolates reduced the numbers of galls by 50.1 - 69.2 %. All endophytic fungal isolates suppressed the population of second-stage juvenile of *Meloidogyne* spp. in the soils by 55.3 – 76.9 %. Three isolates which were CL1 (*Chaetomium* sp.) FL3 (*Fusarium* sp.) and FL4 (*Fusarium* sp.) increased the growth of treated tomato.

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Non-Irrigated Upland Cultivation - Utilizing The Concept of Transpiration Coeffisient

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Abstract

Upland irrigation as an effort to fulfill the balance relationship of water amount distributed and crop effective water required has been oftenly ineffective due to water waste which linked with actual soil physical condition and the surrounding environment. Such a condition become worsening at an extrem period of drought, forcing to alternatively stop water distribution to crop areas of less unpotential benefit. The concept of transpiration coefficient here to might redeem the problem where water need as a minimum factor can be reduced to the least water need as effective as required by the respective crops. It termed: "a certain amount of water required to produce a certain weight of crop dry substances through transpiration process" The application of the concept as a system of efficient water management had been been demonstrated in a case study at the time of widely influence of El Nino in 1997 throughout Asia-East Asia in a remote upland area of Jelegong-Soreang, Bandung Sub District, West Java. The result had proven a promising highlight of upland potency to the additional income of the farmers who used to leave their land bare during dry seasons. Hence, evaluation to validate the concept needs to be actualize in other non-irrigated upland areas.

Keywords: drought period, non-irrigation, transpiration coefficient, upland cultivation

Introduction

In an irrigation system sufficient water source to be distributed is a must. Sufficient means that there is a part of water amount in the root zone effectively used by the crops and another part becomes lost due to natural seepage, percolation, evaporation, transpiration and micro organism metabolism as well as due to culture technique mismanagement. (*e.g.* method of irrigation - Baumann *et al.*, 1974 and method of effective-selective fertilization - Finck, 1976, to name a few).

For water is available during planting season the lost is tolerated. At limited water period, however, water distribution should be carried out in such a way that available water can be utilized as effective and efficient as possible. In that case, efforts in optimizing water distribution system might be one of the good solution but not the best, since there was still an amount of water that has to be considered 'lost' in system optimization due to system as well as technical errors.

Hence, the concept discussed in this paper might be an opportunity to develop a water less agricultural production system, so called as transpiration coefficient.

Materials and Methods

Conceptual Terms

Three dominant aspects involved in the concept of water efficient cultures (Kramadibrata, 1997). They are crop physiology (CP), field technique (FT) and water log accummulation (WA). Hence, an effective irrigation could be defined in a functional term:

Y = f (CP, FT, WA) ……(1)

CP is related to specific property of the any crops on water need in its productive cycles. The property correlates positively to certain extent with nutrients availability in soil solvatation where water for a crop life cycles will be lessening if avalable nutrients are at its optimal level. The fact could be manipulated by application of appropriate and effective fertilizer (Finck, 1976: Kramadibrata, 1978). Whereas FT is linked with water availability that filled soil matrix at its field capacity and WA is an actual amount of available water in the root zone. The existance of the water is naturally occured in a kind of impervious soil holes due to micro organism activities (larves, worms) and organic remains and/or artificially made by a special soil manipulation (Abdul-Rozak, 1993).

Practically Y would be an amount of water [mm] or [L] that effectively used by a crop in its productive cycle to produce a certain yield, termed JE. Hence, CP, FT and WA should have its conversion terms in water amount. In the case, CP is represented by amount of specific water need used by crop (Js), which is based on transpiration coefficient, *i.e.*, *"a certain amount of water required to produce a certain weight of crop dry substances through transpiration in photosynthesis process".*

The FT would be amount of available water in soil matrix (**Jm**), obtained from actual field capacity. And WA would be amount of water that flowed or spread out from water log (**Ja**) spread to the surrounding. To be noted here that water log is the amount of water accumulated in the respective impervious hole which is formed naturally or made artificially (Abdul-Rozaq, *ibid*). The later is a kind of water storage placed at a soil depth of 5-15 cm which will be used as spare water by the crops at drought season.

Therefore, the relation of those terms applies:

JE = f (Js, Jm, Ja)……(2)

In case that carbohydrate (CH) is the weight of crop dry substances desired, JE is the certain amount of water transpired in photosynthesis to produce - say, one kilogram CH of a certain crop in its production cycle. In form of rice, for instance, it is around 35 percents of the total biomass product. Analogically, Y is the amount of water transpired through photosynthesis to yield one kg dry subtances (CH, fat, protein, oil, etc) of a certain crop related to percentage of any substrate obtained from a biomass product.

Based on rice, the equation applies:

J E = f (Js, Jm, Ja) x 0, 35 ……(3)

For calculating variables mentioned, they have to be converted in a similar unit, e.g., in cm height of water column pressure (cmWC) on a certain area [cm^2], pF value, air pressure, or static load [kN/m^2 = kPa]. The relation of those variables is presented in Table 1. that shows

available water in unit of pressures at intervals of pF values from permanent welke point (PWP, pF = ~4.2) to field capacity (FC_{min}-FC_{max}, pF = 2.0-2.2). Hence, the amount of available water for crop (FCc) in various soil textures at a certain field capacity, say, FC at pF = 2.0, can be calculated as shown in Table 2.

Referring to Table 1 shown previously, the amount of available water in soil matrix that can be used by the crops (Jm) lies between the interval of WP (pF < 3,5) and FC (pF ~ 2,4) whereas the amount of water specifically required by a certain crop and/or a certain group of crops (**Js**) is calculated based on from the pressure difference between PWP (pF~ 4.2) and actual FC (pF = say, 2.0), so there is a ΔpF = ~ 2.2, that is equal to a water amount of 11-30 mm or around 11,000-30,000 liters water available in the root zone (30 up to 40 cm soil depth) a hectare.

Hence, there will flow a certain amount of water from soil matrix to the root zone where it is immediately sought by crop roots until Js and Jm reach their equillibrium. So, **Js ≈ Jm.** In the contrary, at **Js < Jm**, there will flow no water to the root zone. The former is obvious that distribution of water occurs only if it is needed, and the later, the crop - at its large can utilize the available water as maximum as it is required.

Table 1. Pressure conversions in Js, Jm and Ja

Pressure				Description
cmWC	**pF**	**Atm**	**kPa**	
15848,9	4,2	15,33	15,548	----------Permanent Welke Point (PWP)---------- ⇑
3162,3	3,5	3,06	3,102	Permanent Welke (WP) Jm
1000,0	3,0	0,97	0,098	Js ⇑ ⇓
251,2	2,4	0,24	0,025	---------Minimum Field Capacity (FCmin)--------- ⇑
100,0	2,0	0,10	0,001	⇐ ◊ ⇒
63,1	1,8	0.06	0,006	⇓ ---------Maximum Field Capacity (FCmax)--------
1,0	1,0	0,01	0,0001	Ja (percolation) ⇓

PWP = permanent welke point; WP = welke point; FC = field capacity; FCmin = FC minimum (pF ~2.4); PCmax = FC maksimum (pF ~ 1,8-2.5); Js = specific water amount requird; Jm = amount of available water in soil matrix; Ja = amount of available water in water log..

Source:: Schroeder, 1972 - Modified, Kramadibrata, 1978

Table 2. Available water for crop (FCc) between FC and PWP

Soil texture	PWP (pF = 4,2) [mm]	FC (pF = 2,0) [mm]	FCc (ΔpF = 2,2) [mm]
Sand (S)	2,5	13,5	11,0
loamy sand (lS)	3,0	21,0	18,0
sandy loam (sL)	4,5	25,5	21,0
Loam (L)	10,5	36,0	25,5
Clay (C)	18,0	40,0	22,0
Organic soil (O)	30,0	74,0	44,0

Source: Czeratzki (in Withers/Vipond/Lecher,1978) - modified, Kramadibrata, (1978)

While water log (**Ja**) is actually the free water, the so called percolation that trapped in the respective water holes. Later it becomes water pockets in the soil as spare water in time of draught which flows based on hydraulic gradient and/or osmotic pressure. So, **Ja** is available there as unused spare water as long as **Js > Jm** and would be supplied just a slightly part in JE calculation. Therefore, it could be discarded (Kramadibrata, 1978). The equation applies:

JE = f (Js, Jm)……..(4)

The functional relation between the variables should be observed in details in forms of action research both under controlled and uncontrolled condition to obtain a relevant relation at a high determinant value (linear, exponential, geometric, logarithmic and/or polinomial equation).

Aspect of Crop Physiology

General term represented by Js includs a theoretical concept which states that each crop has a specific property to transform the amount of water absorbed from the root zone and excerted through photosynthesis process to the air in forms of transpiration through which process various kinds of substrates are produced in the crop.

Generated from photosynthesis equation, the concept is defined as the amount of water required [L] by a certain crop along its production cycle (from seed to yield) to yield a certain amount of dry substrate [kg] at time of harvesting.

$$nCO_2 + nH_2O \xrightarrow[\text{Chlorophyl + energy (photon)}]{} (C_6H_{12}O_6)_n + n\,O_2$$

(4)

(carbohydrate)

Compilation of researchs reported by Baumann, *et al.*, 1974; Finck, 1976; Geisler, 1974; Withers, *et al.*, 1978) linked with the relation of various crops and its water requirement is presented in Table 3.

Table 3. Production of crop dry substrates (DS) based on transpiration coefficient (TC)

Crop	JE [L/kg DS]	Production Cycle [days]	Dry Substrate [kg/ha]
Graminae	471-699	40-60	768-1139
Wheats	411-520	50-120	1031-1305
Wetkand Paddy	550-625	100-110	858-976
Upland Paddy	422-511	105-115	1050-1271
Legumes	403-514	60-90	1044-1331
Earthfruits	298-314	90-150	1709-1800
Maizes	142-315	90-115	1703-3778

JE = effective water need of the crop based on transpiration Coefisient

Sou*rce*: Baumann, *et al*., 1974; Finck,1976; Geisler, 1974 – modified, Kramadibrata,1978.

It is seen from the table that transpiration coefficient (TC) of a crop is determined by a certain amount of water effectively required to produce one kilogram dry substrate (DS) along its production cycle. For instance, graminae needs around 471 up to 699 liter of water

to produce one kilogram dry substrate along its production cycle of 40 up to 60 days. This summed up to 768 up to1139 kilograms a hectare .

Table 4. Production of Dry Substrate (DS) at 50% TC 50% and at ΔpF ≈ 0,7

Crop Types	TC 50% [L/kg DS]	DS [kg /ha]	DS [kg /ha]
Graminae	585.0	367.13	420 *
Wheats	465.5	461.38	720 *
Wetland Paddy	587.5	365.57	560 *
Upland Paddy	466.5	471.51	512 *
Legumes	458.5	468.43	587 **
Earthfruits	306.0	701.87	1690 ***
Maize	228.5	939.93	990 **

* water content 14-16%; ** 18-20%; *** 16% of fresh products
Source : Finck (1976 – modified, Kramadibrata, 1978; 1996)

In terms of water need and fertilizer dosage relationship there has not been many reported yet since field climate hazzards are difficult to control. However, the result obtained by Finck (*ibid*) highlighted a possibility that water less cultivation could also carried out either based on that fact or on Transpiration Coefficient, or both of them (Fischbeck *et al*., 1975). The concept discussed herewith sofar, is the combination of both methods where there is an opportunity to use water efficient as well effective, particularly in non irrigated upland by diversified crop cultures (Kramadibrata, 1978).

Aspect of Field Techniques

General situation in an irrigation system is the occurence of wetting process of the upstream channels up to its saturation level before flowing down to the downstream channels and so on to the desired plots of agricultural sites, and going so on to other plots of other sites (Withers *et al*., 1978).

It is obvious that water lost has been taken place many times during wetting process where some parts of water amount is being used to fill up pores in the soil matrix, both in the channels and in the desired sites beyond, up to their field capacities. The stream of water infiltration to the channel surface is gradually lessening, so irrigation water flow further along the channels. Along with the process water lost happened all the time in forms of seepage through channel/ditch walls, percolation, direct evaporation (soil and water surfaces) and evapo-transpiration (soil surface and vegetation surrounding the channels such as grass, weeds, bushes and trees).

Aspect of Accumulated Water Log

The aspect includs some concepts in utilizing various phenomena, i.e.; osmotic pressure that leads to pitcher irrigation (Siyal, *et al*., 2009), mulching to catch condensation water, water log as storage water in draught period, impervious surface channels resulted from wheel traffic of agricultural vehicles that hold water on the soil surface and water detention structures enable water to perforate into the soil.

Field Practice

Table 1 above could be interpreted in the calculation which based on the limit between welke point (WP) and minimum field capacity (FC_{min}) and the limit between WP and maksimum field capacity (FC_{max}). Hence, the following calculation applies:

The Limit Between WP and FC_{min}

In a pessimistic assumption, if water availabilty in a land is the amount of water contained in the interval between WP (say, pF ~3.5) and FC_{min} (say, pF ~2.4), so there is a different pF values of 1.1 which is adequate to water pressure on an area of $1cm^2$ at a height of 12.59 mmWC, or 0.01259 mWC. In a hectare land this is equal to 0.01259 m x 10,000 m^2 = 1259 m^3 water a hectare. Assuming that only 50 percent of that amount could be utilized, so there would be around 679.5 m^3 water available a hetare.

The Limit Between WP and FC maks

Analog as above, if water availabilty in a land is the amount of water contained in the interval between WP (say, pF ~3.5) and FC_{min} (say, pF ~1.8), pF different is 1.7 that is equal to 0.0502 mWC. Also by assuming of 50 percents of save usage, there would be around 2506 m^3 a hectare.

Result obtained from conversion, the amount of water in forms of dry substrates of the crops - calculated based on transpiration coefficient as presented in Table 2, is shown in Table 3.

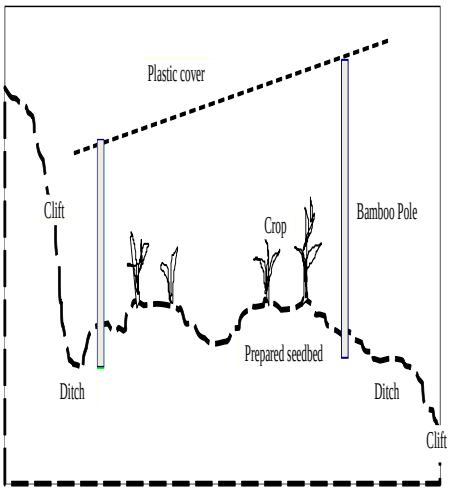
Obviously without any drops of water a land having such a condition of field capacity as shown in Tables 1 and 2, sufficiently potential to yield a certain amount of dry substrates. Consequently, it is considerably fund saving since costs for irrigation water, maintenance of channels and other water constructions would be eliminated, or expended for other costs only if it is necessary.

In addition, there is an alternative to select and cultivate certain crops which are suitable to the actual water availability of the field. They require a lower amount of water than paddy along their life cycles as well as have a higher market values. Some trash crops such as legumes, earthfruits and maize, as shown in Table 3, have both advantages.

The actualization of the concept was observed in a demonstration plot (Demplot) under which various trash crops were cultivated. It was located on a hang site of an upland area where no any irrigation system constructed. The observation was conducted at time of long period of drought in 1997 which was influenced by *El Nino.* The location is in Jelegong village, Soreang sub district, Bandung Distrct, West Java. The preparation had been carried out by the site farmers.

All over the demplot was covered by a transparant plastic roof construction supported by bamboos poles and no wall, so sunlight could go through the roof to the soil surface and fresh air could circulate and pass freely through the construction and the cultivated crops. The construction was designed in a way, as shown in Figure 1(a), that rainfall (if any) would be detented and flowed away from the demplot through ditch constructions. The aim of the construction was obviously to let sunlight and fresh air in but prevent from water inflow into the demplot.

Field capacity of the demplot was periodically measured using potentiometers installed during the whole period of trash crop life cycle (around 120 days).



Blind Test		
amp1	bmq2	ctp1
cmq2	dmq1	ctq3
btp1	atp3	amq1
amq3	btp2	cmp2
btq3	cmp1	bmp1
btp3	atq2	bmq3
bmp2	dtq1	dtp3
atp1	cmq1	ctq2
dtp2	btq2	btq1
atq1	atq3	ctq1
dtq2	ctp3	amq2
dmp3	cmq3	atp2
ctp2	amp2	amp3
dtq3	dmq2	bmq1
bmp3	dmp1	cmp3
dmq3	bmp2	atp1

(a) (b)

Figure 1. a. Cross-Section of the demonstration plot; b. A randomized block design

The experiment method employed a randomized block design (RBD) at four replications, as shown in Figure 1(b) Two treatments applied were: planting four local crop varieties , *i.e.*, a = tomato; b = terong; c = maize; d = soybean, and applying four technical seedbeds, *i.e.*, m = covered by dark plastic layer; t = uncovered; p = tilled; q = untilled. Whereas NPK fertilizer and stall mist were given at a rated dosage to all plots, includings the control plots. The covered demplot was devide into 48 tested plots of 1x1 m^2 each. A control plot was placed outside the demplot, *i.e.*, a bar plot, no treatments at all.

Results and Discussion

The result obtained from field capacity measurement using potentiometer is presented in Table 5 through which the amount of water transpired in photosynthesis (JE) could be actually determined based on the average different of pF values between pF at PWP and actual pF (ΔpF).

Table 5. Actual pH dan pF of the soil and its conversion to water availability (Je, L/Ha)

Date	3/5/97	10/5/97	17/6/97	26/6/97	3/7/97	16/7/97	23/7/97
pH	6.0	6.0	6.0	6.1	6.1	6.1	6.2
Time							
10 am	2.35	2.35	2.36	2.36	2.37	2.38	2.39
12 pm	2.42	2.44	2.45	2.45	2.47	2.48	2.48
2 pm	2.45	2.46	2.47	2.48	2.49	2.50	2.51
pF	2.41	2.42	2.43	2.43	2.44	2.45	2.46
pF at PWP	4.20	4.20	4.20	4.20	4.20	4.20	4.20
Average ΔpF	1.79	1.78	1.77	1.77	1.76	1.75	1.75
JE(x 1000)	549.205	546.136	543.068	543.068	540.000	536.932	536.932

Source: Field Data (Kramadibrata, Mei-September 1997)

Qualitative results are shown in Figure 2 to 5, indicated that all trash crops cultivated were well grown after 1 week planting.

Figure 2. Demonstration plot before and after crop planting (10 days seeds)

Figure 3**.** Crops growth under plastic roof construction at weeks 1 to 3

Figure 4. Crop growth at weeks 3 to 7 (tomato, eggplant, maize and soybean)

Figure 5. Crops and its yields ready to harvest at the last day of week 7 (tomato and maize)

At the periode of approaching week 3 after planting there was a sign of watter deficiency on eggplant, whereas tomato, maize and soybean were at their generative growth phases. Then at week 4 after planting, tomato and maize had already shown their young fruits. Soybeans in several plots indicated symptoms of water deficiency though, but most of them had passed their generative phases and starting to fruit. At the end of week sixth after planting tomato was ready to be harvested, while maize and soybean were ready at the end of week seventh.

Table 6**.** Yields of trash crops in various tested plots

Crop	Tested plots	Biomass [g]	Yield [g]	Total [g]	Yield [%]	Description
a	amp1	20.6	90.6	111.2	81.47	11 out of 12 tested plots (91,7%) grown and yielded*): 1) 3 plots of mp (25,0%) = A 2) 2 plots of mq (16,7%) = B 3) 3 plots of tp (25,0%) = C 4) 3 plots of tq (25,0%). = C
	amp2	16.7	52.2	68.9	75.76	
	amp3	20.2	55.8	76.0	73.42	
	amq1	35.7	44.9	80.6	55.71	
	amq2	23.2	44.8	68.0	65.88	
	atp1	23.1	41.4	64.5	64.19	
	atp2	20.1	45.3	65.4	69.27	
	atp3	25.6	51.9	77.5	66.97	
	atq1	25.7	74.3	100.0	74.30	
	atq2	34.3	38.4	72.7	52.82	
	atq3	20.3	65.0	85.3	76.20	
Total	11	265.5	604.6	870.1	69.49	
b	bmp2	15.9	14.2	30.1	47.18	3 out of 12 tested plots (25,0%) grown and yielded*): any plots of mp, mq, and tp (8,3%). = D
	bmp3	10.9	-	10.9	0.0	
	bmq1	19.2	35.0	54.0	64.81	
	bmq3	12.9	-	12.9	0.0	
	btp1	14.7	-	14.7	0.0	
	btp2	14.2	15.7	29.9	52.51	
	btp3	16.3	-	16.3	0.0	
	btq1	21.8	-	21.8	0.0	
	btq2	13.6	-	13.6	0.0	
	btq3	16.7	-	16.7	0.0	
Total	10	156.2	64.9	229.9	28.22	
c	cmp1	436.7	142.3	579.0	24.58	7 out of 12 test plots (58,3%) grown and yielded*): 1) 2 plots of mp (16,7%) = E 2) 2 plots of mq (16,7%) = E 3) 1 plot of tp (8,3%) = F 4) 2 plots of tq (16,7%) = FG
	cmp2	429.8	136.9	566.7	24.16	
	cmq1	202.8	90.0	292.8	30.74	
	cmq2	496.3	161.7	658.0	24.57	
	ctp1	448.8	210.3	659.1	31.91	
	ctq2	242.7	135.0	377.0	35.81	
	ctq3	488.0	130.7	618.7	21.12	
Total	7	2745.1	1005.9	1654.8	60.79	
d	dmq1	2.02	1.27	3.29	38.60	6 out of 12 tested plots (50,0%) grown and yielded*): 1) 1 plot of mq (8,3%) = H 2) 2 plots of tp (16,7%) = H 3) 3 plots of tq (25,0%). = H
	dtp2	3.25	2.15	5.40	39.81	
	dtp3	3.70	1.93	4.63	41.68	
	dtq1	4.25	2.46	6.71	36.66	
	dtq2	2.13	0.90	2.21	40.72	
	dtq3	5.21	3.65	8.86	41.20	
Total	6	205.6	123.6	311.0	39.74	
Total	34	3372.4	1799.0	3065.8	58.69	

a = tomato; b = eggplant; c = maize; d = soybean; plot area = 1 m^2; m = darky plastic covered; t = uncovered; p = untilled; q = tilled; 1,2,3 = replications

* same capital letter = not significantly different atα= 0.05

Quantitative results are shown in Table 6 - based on statistical analysis at 5 percent level of confidence, it was indicated that 91.7 percents of tomato were able to grow and yield its fruits, while eggplant, maize and soybean reached a total yield of 25.0; 58.3 and 50.0 percents, respectively.

On the other perspective – indicated in Table 7, the total production of dry substrates of all crops was 1903.9 kg a hectar, in which 69.48 percents were obtained from tomato, 29.35 percents from eggplant, 60.77 percents from maize and 39.75 percents from soybean.

To note from statistical analysis, it showed that different treatments of technical seedbeds had been caused a variation of significant differences to the obtained yields. But the results were considered unvalid to avoid any misinterpretation.

Table 7. Production of dry substrates a hectare

Crop	Biomass [kg]	Yield [kg]	Total Weight [kg]	Percentage of Yield
Tomato	389.09	894.89	1287.98	69.48
Eggplant	1551.85	690.38	2352.23	29.35
Maize	430.94	667.56	1098.50	60.77
Soybean	666.68	439.84	1106.52	39.75
Total Yield	1903.90	2692.67	4596.57	58.58

*Source:*Kramadibrata, Field Data, Mei-September 1997

The entire yield was actually represented 65 percents of the total production at normal water availability. This showed that there was a possibility to apply the concept of TC in a greater scale, particularly in efforts to increase the productivity of both upland and dryland in the cultivation of fallow system, neither it has nor it has not an irrigation system.

Conclusions

The concept of utilizing transpiration coefficient is theoretically and practically possible as had been proven in a partial upland cultivation. Nevertheless, further information from research and observation in a broader area should be still supplied to actualize in the field practice.

Practical problems might come from the fact that soil field capacity varies from one site to another, so its actual water availability should be determined first. Hence, undisturbed soil sampling from root zone to determine the actual negative pressure in soil laboratorium would give more representative data of actual soil water availability and so the suitable crops selected.

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Prospect and Challenge of The Usage of Portable Near-Infrared Spectrometer to Assess Fruit and Vegetable Quality in Indonesia

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Abstract

Measuring quality attributes, particularly those related to internal quality has become one of the future researches in the field of non-destructive quality evaluation. The usage of portable near-infrared (NIR) spectrometer to assess the quality of fruit and vegetable is quite common in several countries. Our previous researches in Japan utilized fruit vegetables. The data results of NIR absorption spectra were transformed with second derivative spectra and analyzed by the Un-scrambler multivariate calibration software. The usage of NIR showed better results of coefficient correlation for detecting internal quality of fruit and vegetables. Prospect of the usage of portable NIR for rapid and accurate measurement on fruit and vegetables quality in Indonesia were explored. There were so many exotic products among Indonesian local cultivars which have not been researched for its quality using non-destructively NIR method. The non-destructive method to assess quality was not so common and popular in Indonesia, because the method and apparatus was not familiar enough. However it is needed to guarantee the quality of the fruit and vegetable to be consumed or exported, if possible in a very simple and efficient way such as portable NIR. The usage of portable NIR to assess the quality of the fruit and vegetable still has challenges in Indonesia. In developed countries, the non-destructive quality assessment on fruit and vegetable is the guarantee mark that could increase the price of the product. The internal quality assessment that could be detected by NIR is various depend on the fruit and vegetable. Several researches have successfully determined the quality attributes of fruit and vegetable such as sugar content, total soluble solid, moisture content, firmness, and color. Recently, NIR is also used by researchers to detect insects and diseases contained in the fruit and vegetable. It is very important and useful tool especially for high-value fruit and vegetable products to be exported overseas.

Keywords: fruit, portable NIR, quality, vegetable,

Introduction

Farmers usually use ripeness stage as the basis to determine when harvest fruits and vegetables. In this case, fruit ripeness is subjectively determined based on skin/peel color or on the time elapsed after flowering or planting. This method is simple and easy especially for skilled pickers. However, it may become a problem when under-experienced pickers may not be able to determine the ripeness objectively, time consuming, and bruising occured by touching frequently. Moreover, the skin/peel color of some fruit and vegetable cultivars are almost the same before and after ripening such as mango *cv harumanis*, apple *cv Malang*, snake fruit, jack fruit, durian, mangosteen, sawo (*Achras sapota* L.), avocado, rambutans *cv rapiah*, and bitter gourd. Consequently, serious losses in terms of fruit rejects from sorting machine have been experienced due to variations in internal quality of the fruits. For most fruits and vegetables, quality firmness is an important indicator of physical

quality, shelf life and consumer's acceptance (Kader, 2002). On the other hand, internal quality attributes associated with chemical content such as total soluble solid, moisture content, sugar content and its composition, vitamin-C are very difficult to be detected by non-destructive methods. These internal quality contents are significant indicators of healthy food (Saltveit, 2005).

Nowadays, not only fruit color, measuring other quality attributes, particularly those related to internal quality, can be predicted and sorted more correctly, objectively, and non-destructively using spectroscopic measurement (Saltveit, 2005).

Non-destructive quality evaluation has been generally limited to laboratory conditions using a set of near-infrared (NIR) instrument on harvested fruit (after harvesting). More interest is given for on-tree/plant measurement whereas quality evaluation was conducted using portable NIR spectroscopy while the fruit is still attached to the tree/plant before harvesting. The portable NIR spectroscopy can be used not only in laboratory, but also convenient to be used in the field because it is very compact and relatively light weight. The on-tree quality evaluation has been successfully done for mango (Saranwong *et al*., 2004) and apple fruit (Zude *et al*., 2006). For tomato, portable NIR technique has been tested to determine soluble solids content, moisture content, hue color value, and firmness of fruit on-tree and after-harvesting (Kusumiyati *et al*., 2007).

Measuring on-tree spectral data in the field can be done using a portable NIR instrument. NIR data on fruit quality parameters can be integrated in calibration and prediction models. If strong correlation among quality parameters could be established, the models can be used to determine the harvesting time and eliminate fruits rejected from sorting machine (Peirs *et al*., 2003).

Therefore, the importance of portable NIR technique as a useful apparatus to determine quality attributes for some fruits and vegetables has been widely recognized. However, the research on the usage of portable NIR technique to determine quality attributes for local fruit and vegetable cultivars in Indonesia, especially while still attached to the tree/ plant has been rarely conducted.

The aim of this research was focused on analyzing the prospect and challenges of using portable NIR spectrometer to determine the internal quality parameters for local fruits and vegetables in Indonesia based on previous researches.

Materials and Methods

The method of this research is descriptive analysis. As comparison, data was collected from previous experiments. One of the experiments was conducted from May to August 2005 on bitter gourd. The bitter gourd fruits were all in the 150-250 g size band and were harvested from the hydroponics area at the University of the Ryukyus, Okinawa, Japan. The harvest times for all the fruits were determined 15 days after pollination. The fruits were analyzed in the laboratory at 25oC and all of the measurements were made on the harvest day. The total number of samples, which were used for the soluble solid content and moisture content analysis was 53. Other experiment data were collected from the same place on August to May 2007 on tomato fruit at different maturity stage. Briefly, in this experiment, the tomatoes were harvested in winter (from January to March 2005) and summer (from June to August 2006. All of the samples were harvested during the commercial harvesting time (about 2 weeks after spraying the oxicyne tomato hormone) from the university

experimental station, based on the visual appearance in the range from a red yellow color to the fully ripened tomato (fully red color). The total data used in this experiment included 247 tomato fruits, consisted of 182 tomato fruits from the winter season, and 65 tomato fruits from the summer season.

Spectra Acquisition Method by Portable NIR

Figure 1 shows a portable type of NIR (FANTEC FQA-NIRGUN, Japan) used in the researches. The wave length range is 600-1100nm with 2nm intervals. It looks like a gun, and it's very compact and convenient to be used in the field because of its size (250 mm length) and light weight (800 g). Spectrum collection and data storage took no longer than 30 miliseconds per fruit. The raw intensity spectrum for a fruit was the average of 50 individuals intensity scans. The light from a tungsten halogen lamp was directed by fiber optics to the fruit placed in a black holder. The fruits were placed under a black cloth cover to avoid any scattered sunlight during the measurement.

Figure 1. NIRGUN portable equipment (Constituent Measurements (soluble solid content, moisture content, and fruit Color)

The fruit soluble solid content was estimated from a small sample of fruit juice (without seeds for the bitter guard) from the middle part of the fruit. The soluble solid content level was measured using a digital refractometer (Atago-PR1, Japan) after extraction of juice and the readings given in ^{o}brix.

The fruit moisture content was measured by cutting the fruit into slices. The moisture content was measured by a drying method using an oven at the constant temperature of 105^{o}C for 24 hours. After 24 hours, the fruit is removed from the oven, and pre-cooled in a dessicater before weighing. The fruit moisture content was calculated as the percent moisture on a wet basis.

The measurement of tomato fruit composition for hue color was determined from the freshly harvested tomatoes. The hue color value was obtained in terms of the L, a, and b values using a digital colorimetric instrument (CR-200 Minolta, Japan).

Data Analysis

The Un-scrambler software version 7.51 (Camo, Oslo, Norway) was used for the data analysis. The NIR original spectral data were obtained using the portable NIR and stored as absorbance spectra and pretreated by second derivative processing of the spectra. The

calibration model was used to estimate the constituent of the prediction set. The prediction values were compared to the actual measured values using a regression analysis. The statistical results of the coefficient regression (R) calibration and validation, standard error prediction (SEP), and standard error calibration (SEC) were used to check the success predictability of the model.

Results and Discussion

The best correlation coefficient of the soluble solid content and moisture content showed promising results for predicting each constituent (Table 1), even though they were not very high, (R~0.65-0.81). It is suspected that the light cannot be completely absorbed due to the jagged edge/uneven shape on the fruit surface and the homogenous sample data (Table 2).

Table 1. Result of calibration and validation by PLSR in bitter gourd

Constituent	Validation			Calibration		
	R	SEC	Bias	R	SEP	Bias
SSC (Brix)	0.64	0.25	2E-08	0.6	0.26	0.0003
Moisture (%)	0.64	1.39	-3E-06	0.55	1.52	0.0031

Table 2. Statistical characteristics of samples used in data analysis of bitter guard

Sample sets	SSC (^{o}Brix)	Moisture Content (%)
Range	2.10 - 3.63	86.69 - 93.18
Average	2.93	89.99
Standard deviation	0.33	1.83

Table 3. Result of calibration and prediction models by PLSR in the tomato

Constituent	Calibration		Prediction			PLS component
	R	SEC	R	SEP	RPD*	
Soluble solids content (^{o} brix)						
After harvesting	0.95	0.26	0.92	0.33	2.47	5
On tree	0.84	0.45	0.77	0.50	1.57	5
Moisture content (%)						
After harvesting	0.81	1.85	0.80	2.04	1.66	5
On tree	0.84	1.78	0.82	1.91	1.71	4
Hue color value						
After harvesting	0.94	3.32	0.87	5.57	2.02	5
On tree	0.91	3.92	0.84	6.09	1.81	5

* RPD is the ratio of standard deviation of reference data in validation set to SEP

In general, portable NIR spectrometer was able to estimate the soluble solids content, moisture content, and hue color values by the developed PLS model for the tomato (calibration correlation and SEC were 0.95 (0.26), 0.81 (1.85), and 0.94 (3.32) respectively) from samples in the winter season and summer season. The correlation coefficients and SEP of the prediction set for the tomatoes were 0.92 (0.33), 0.80 (2.04), and 0.87 (5.56) respectively (Table 3).

The positive and similar trend linear correlations of the soluble solids content, moisture content, and hue color value during the winter season and summer season are shown for

the tomatoes. Variance in the quality attributes of the tomato from winter season and summer season was high in range and could make the models more robust.

There are so many exotic products in Indonesian local cultivars, such as mango, papaya, apple, snake fruit, jack fruit, durian, mangosteen, sawo (*Achras sapota L.*), tomato, bitter gourd, which have not been researched for its quality using non-destructively NIR method. It is promisingly feasible for fruit and vegetable products to be exported, because quality guarantee mark may increase their price. The quality guarantee may also include free from insects and diseases. This method is also useful to reduce bruises occured from sorting by physical touches.

Challenges of this non-destructive method is mostly to introduce this method to the farmers, particularly since the quality guarantee mark detected by NIR is not familiar. In the beginning, it may be costly for the investment to buy the apparatus and for training the farmers to use this method appropriately, which in turn would increase the price of the product for consumer. In the early stages, this method could only be used for some certain level of farmers, since there are still so many illiterate or unskilled conventional farmers in Indonesia, as well as many other developing countries. However it is predicted that after some successful researches and field practices, the implementation of portable NIR spectroscopy will be widely accepted, particularly for high-value fruit and vegetable products in which the consumers demand quality as their first priority. In turn, the investment of using this method will be paid off by more profit for the farmers. It is also predicted that after wide implementation of NIR spectroscopy, the price of portable NIR would be significantly reduced.

As mentioned previously, the usage of portable NIR spectroscopy could be very useful for direct and non-destructive measurement, in which the main factor analysis-derived quality components could be predicted with sufficient accuracy. The other advantage of spectroscopic readings is able to determine the best harvest time and sorting based on the fruit quality. If the calibration model could be formulated precisely, the NIR measurement can be applied directly and non-destructively by the farmers themselves. So that after measurement, the product is still marketable and profitable for farmers. For example, in Japanese market, the fruit can be sold for higher prices because of its guaranteed sweetness taste and other quality level if growers noted the tastes of the fruit. The sensor from NIR will assure consumers the quality level of fruit. Moreover, the method is friendly to the environment because of the elimination of chemical material usage during fruit quality analysis. The usage of portable NIR to predict quality assessment non-destructively is not only scientifically important but also applicable for farmer, useful for daily practice, and may promisingly will lead to contribute improvement on agricultural productivity in the world.

Conclusions

The application of NIR measurement could be used to predict the optimal harvesting time of a fruit or vegetable on tree or after harvesting depending on the farmer's purpose. Based on the on-tree NIR measurement technique, the farmers is able to determine the harvesting time as for whether the fruit or vegetable is ready to be harvested or will be postponed until the desired levels of the soluble solids content, moisture content or hue color value has been obtained. This NIR quality assessment has a promised prospect as one of the future researches in the field of non-destructive quality evaluation.

There were several challenges to use portable NIR in Indonesia for some local cultivar of fruits and vegetables, particularly in terms of introducing this method for the regular farmers around the country. In this context, the research and field implementation of NIR on local Indonesian fruit and vegetable cultivars should be encouraged further. Also, this promising on-tree quality assessment should be viewed as one of the feasible effort to agriculture development in Indonesia as a significant agrarian country in the world.

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Enzymatic Production of Monoglyceride Through Esterification System

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Abstract

Coconut and palm kernel oils were recognized as health oils in Ayurvedic Medicine. Modern research has found a common link between these two natural health products their fat or lipid content. Medium chain fatty acids and monoglycerides found primarily in these two tropical oils and mothers milk had functional effects. The aim of this research was to develop esterification system for the enzymatic production of monoglyceride (MG). Novozyme® 435 was employed as catalyst in esterification system. This research was using Central Composite Design from Response Surface Methods (RSM) as experimental design. Temperature and time of reaction were as variables. Semi continuous circulated packed bed reactor had residence time of 23.57 minute, glycerol/ oil molar ratio of 5:1, solvent/substrat ratio of 8.8:1 (wt/wt), and the process produced MG up to 80%. Optimization was obtained quadratic equation which was $Y= - 61.700 + 6.088\ x_1 + 3.259\ x_2 - 0.065\ x_1^2 + 0.017\ x_1x_2 - 1.792\ x_2^2$ *with* $R^2 = 0.5408$*. The optimum temperature and time reaction of 46.92°C and 1.1 hour, respectively and predictive value for MG content was 82,96%.*

Keywords: ***candida antartica lipase, esterification system, monoglyceride, optimization, organic solvent***

Introduction

Coconut and palm kernel oils were recognized as health oils in Ayurvedic Medicine. Modern research has found a common link between these two natural health products their fat or lipid content. Medium chain fatty acids and monoglycerides found primarily in these two tropical oils and mothers milk had functional effects. Monoglycerides (MGs) are emulsifier that widely used as food additives. MGs are used in bakery products, margarine, dairy products, and confectionary because of their emulsification, and conditioning properties (Damstrup *et al.,* 2005). Monoglyceride of lauric acid, is one of the products derived from oil, which has privileges. Privileges this monogliceride beside as food preservative and sanitizer, it also has an ability to destroy the herpes and HIV-1 virus. Monolaurin can reduce the risk of transmission HIV virus to infants from HIV-infected pregnant women, and effectively inhibit vegetative cells of *B. Cereus* by damaging the membrane, causing leakage of intracellular proteins (Cotton and Marshall, 1997; Kabara, 1983). Nowdays, monoglyceride of lauric acid is already used in the cosmetics, pharmacy and medicine industry.

Production of MG can performed chemically and enzymatically. Today, commercial MGs are manufactured by chemically glycerolisis of fat/oil. This chemical reaction take a long time, not selective products, and use large amounts of energy. In addition, this method use high temperature (220-260 °C) and will produce undesirable products such as dark color, burnt taste, and off flavor. Enzymatic production of MG was choosen by researchers in recent years, because the catalytic activity of enzymes that are very high and the ability to work at

relatively low temperature (McNeill *et al.,* 1992). The development of enzyme technology immobile increased enzyme stability (Haryadi, 1996).

In this study, MG was produced by esterification reaction of lauric acid and glycerol using lipase immobile from *Candida antartica*. Factors that influence enzymatic synthesis of MG by esterification includes: temperature, reaction time, enzyme dosage, and amount of solvents. The present study was aimed at optimizing enzymatic esterification in organic solvents for monoglyceride production.

Materials and Methods

Materials and Tools

Commercially lauric fatty acid (technical grade) with 98,9% lauric acid and water content 0,6%, glycerol (technical grade) was purchased from Sigma, Novozym® 435 purchased from Novozymes A / S (Bagsvaerd, Denmark). The enzyme was a *Candida antartica* lipase that was produced by submerged fermentation of a genetically modified *Aspegillus oryzae* microorganism and that was subsequently adsorbed on a macropirous resin. Standard of monolaurin purchased from Sigma, hexane (techinal grade), tert-butanol p.a (purity:99%) purchased from Sigma. The chemicals used for analysis (NA2S2O ¬ 3) 0.1N, Wijs solution, Alcohol 95%, PP and starch indicator, 0.01N NaOH, chloroform, dimethyl formamide (DMF), benzene, and aquades were provided from Food chemistry Lab-SEAFAST Center. The tools that was used in this research: packed bed reactor which is equipped with a stock tank, peristaltic pump (BT 100-1 F), waterbath (Stephen Haake, Germany) as well as container container products, Gas Chromatography with a detector FID (Flame Ionization Detector), analytical balance, Aw-meter (Shibaura WA-360), pH meter, filter paper and glass equipment.

Esterification Procedure

Glycerol/fatty-acid esterification was carried out in packed bed reactor. The substrates amounts in the molar ratio lauric acid: glycerol (1:5). The amount of enzyme added after stabilization of the temperature was 5% (w/w) of the lauric acid. The reactions were performed in pure *n*-hexane/tert-butanol (3:7, v/v).

Methods

Analysis of raw materials

Fatty acid and enzyme used in the esterification system is analyzed before hand to determine the quality of raw materials. The analysis was conducted on the water content (AOAC, 1995), free fatty acid (FFA) (AOCS Official Method Ca 5a-40, 1998), and peroxide value (AOAC, 1995), GC-FID analysis (Modification of AOAC Official Method 993.18, 1995). Enzyme characterization was carried out first with measurement of pH and Aw enzyme (Haryadi, 1995).

Stages of monoglyceride production

Optimization production of monoglyceride

In this stage, the aimed was to find the optimum conditions of esterification process which produce products with maximum MG composition. The experimental design used in this study follow design of the Central Composite Design (CCD) of Response Surface Methodology (RSM) with two variables, time and temperature esterification reaction. Response Surface Model was used to see the effect of treatment time and reaction temperature on yield of products, the composition of MAG. Midpoint was taken from the temperature and reaction time on previous research results. The data obtained is analyzed with SAS program 9.1.3

Experimental design and statistical analysis

Response surface methodology (RSM) enables evaluation of effects of multiple parameters, alone or in combination, on response variables. Software SAS 9.1.3 was used to assist the design, statistical analysis, and reaction optimization. A two factor fractional factorial design with five central points was adopted to optimize the semicontinuous esterification of lauric acid. Two factors chosen for the optimization were reaction temperature (T) and time reaction (t). The variables and their levels were selected from single factors studies and are presented in Table 1.

Table 1. Set Factor Levels and Observed Responses in Response Surface Methodology Experiments for Enzymatic Esterification in the Packed Bed Reactor

Code for Temperature (T)	Code for Time (t)	MG content (%)
-1	-1	86,75
-1	1	79,16
1	-1	80,82
1	1	73,4
-1.414	0	73,94
1.414	0	79,04
0	-1.414	79,38
0	1.414	77,84
0	0	81,72
0	0	79,63
0	0	79,82
0	0	82,9
0	0	81,12

$$Y = \beta_o + \sum_{i=1}^{2} \beta_i X_i + \sum_{i=1}^{2} \beta_{ii} X_i^2 + \sum_{i=1,j=2}^{2\text{-}1,2} \beta_{i,j} X_i X_j + \varepsilon$$

Y is the response (the content of MGs, wt %), β_o) intercept, β_i first-order model coefficients, β_{ii}) quadratic coefficients for the *i*th variable, β_{ij}) interaction coefficients for the interaction of variables *i* and *j*, and *Xi*) independent variables. Second-order coefficients were generated by regression analysis with backward elimination. Responses were first fitted for the factors by partial least-squares regressions. The fit of the model was evaluated by the coefficients of determination (*R*2) and analysis of variance. The insignificant factors were eliminated from evaluation and the model was finally refined.

Result and Discussion

Chemical characteristics of raw materials

Lauric acid used in this study has a water content of 0.34 ± 0.02% (db). Fatty acid content 98.91 ± 0.34%, peroxide value 1.23 meq O2/kg. According to De Greyt *et al*. (1997), raw material has requirement for peroxide value is lower than 10 meq O2/kg. Enzyme Novozyme® 435 has pH and Aw are 4.38 ± 0.02 and 0.62 ± 0.004 at a temperature of 29.5°C, respectively. Purity content of lauric acid in raw materials is shown by gas cromatography (GC) only one peak with retention time 7.312 minutes (Fig. 1). Its means that the content of lauric acid as raw material is pure.

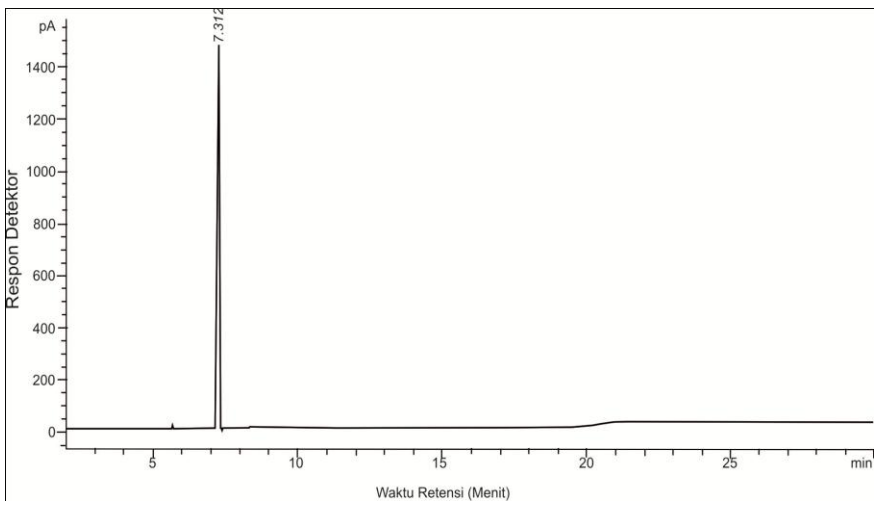

Figure 1. Chromatogram of lauric acid esterification of fatty acids with glycerol

The discovery of the high stability of lipases in organic solvents offered the possibility of a reverse reaction of hydrolysis, esterification using free fatty acids (FFA), or transesterification using esters. For that purpose conditions are needed, in which the enzyme will catalyze the synthesis reaction rather than the hydrolysis. Most important, low water content and low water activity are necessary. The reaction of fatty acids and glycerol can be

seen in Figure 2. Triglycerides are formed when three fatty acid with one molecule of glycerol esterified (Winarno, 2002). Glycerol will cause the reaction equilibrium towards to the right direction so that esterification reaction produce high MG (Fischer, 1998).

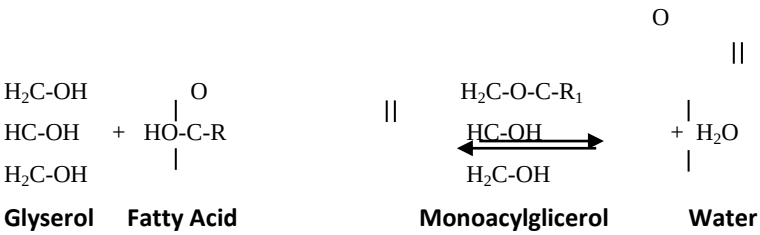

Figure 2. Esterification reaction of one molecule of glycerol with one fatty acid (Winarno, 2002)

Esterification system used in this study was semicontinous reaction and took place in circulated packed bed reactor. Residence time was determined before. Continuous esterification, where substrate in reactor flow to the enzyme reactor and then returned to the substrate reactor until the time and reaction temperature was reached. Scheme of flow reaction is shown in Figure 3. Time when molecules through enzyme reactor with a specific flow rate called the residence time (Yang *et al*., 2005). Residence/space-time is analogous to reaction time on a batch system and is defined as time needed to process reactants for one volume of reactor in certain conditions (Levenspiel, 1972).

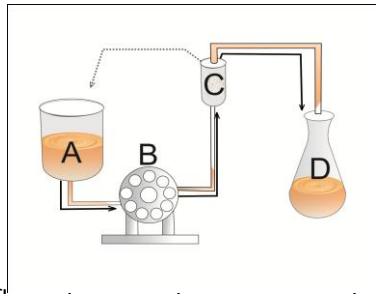

Figure 3. Scheme flow of semicontinuous enzymatic esterification reaction in a packed bed reactor

In present study, space-time is 23.57 minutes, where the substrate flow rate used is 0.7 ml / min. Working volume of enzyme reactor is 15 milliliters and then filled with 4.6 grams Novozyme® 435. The process of semicontinuous enzymatic esterification reaction then performed with the best composition and the glycerol-soluble solvent from previous trial study, substrate ratio (1:5 mol/ mol) and the ratio of the amount of substrate with solvent 1:8,8 (w/v). Selection of solvents is an important factor in enzymatic esterification reaction, another factors are solubility of substrates and products in solvent, solvent hydrophobicity, reactivity solvents, density, viscosity, surface tension, toxicity, easy/absence of fire, how to waste disposal to environment, and of course, cost issues (Dordick, 1989). From these various factors, main attention is the problem of solvent hydrophobicity (Hariyadi, 1994). Thus, a high catalytic activity is generally observed in a hydrophobic solvent, while hydrophilic solvent which does not support the emergence of catalysis activity. hydrophobicity parameters used were log P values of solvents used. Solvents used in this study were hexane with its log P value is 3.5, while log P tert butanol is 0.4 (Damstrup *et al*.,

2006). Log P values of solvents used in this study was 1.33. it means that the solvent in this study tend to be hydrophilic.

Optimization Monoglyceride Production

The whole treatment consists of 13 units of an experiment where each treatment following to the Central Composite Design (CCD) with two factors of Response Surface Method (RSM). Results of optimization with 13 treatment on Packed bed reactor circulation and then visualized with response surface methods so as to obtain a quadratic equation as shown in Table 2.

Table 2. Characteristics of Mathematical Equations Based on Central Composite Design of Response Surface Method (RSM) and Value Prediction for Optimum Product Quality

Responses	Persamaan kuadratik	R^2	Karakteristik Model	Prediksi Variabel Optimum	Nilai Prediksi Parameter Maksimum
Kadar MAG (%)	$Y= - 61,700 + 6,088\ x_1 + 3,259\ x_2 - 0,065\ x_1^2 + 0,017\ x_1x_2 - 1,792\ x_2^2$	00,5408	Maksimum	Suhu 46,92 ^oC & waktu 1,1 jam.	82,96%

Figure 4 shown the effects of temperature and reaction time on monoglyceride content. Based on the figure seem a curve on the Y axis (reaction time) is more convex than the curve on the X axis (reaction temperature). Based on two-dimensional contour drawing MG (Fig. 3) appears inflection point the curve on the Y axis (reaction time) more sharply than the curve on the X axis (reaction temperature). This suggests that the influence of reaction time is greater to improving product MG compared than reaction temperature. On the Y axis seen MG will reach a maximum at reaction time 1.1 hours, but the MG will decrease with increasing time of more than 1.1 hours. On the X axis appears that MG will reach a maximum at reaction temperature of 46.92 °C (on actual 47±5 °C), but will decline rapidly with increasing reaction temperature is more than 46.92 ^oC (on actual 47±5 °C). Canonical analysis shown that MG at a stationary point shows the maximum value equal to 82.96% (Fig. 3).

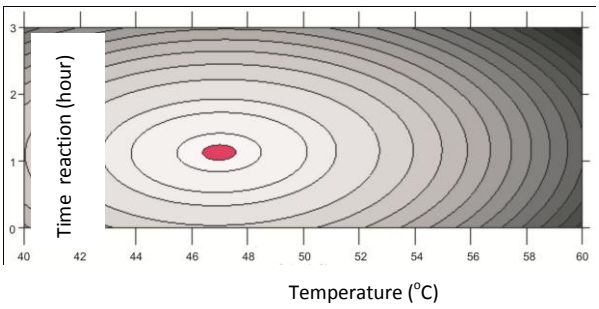

Figure 4. Contour Response for MG content

Mathematical models (content of MG as response) were established and used for interpretation, predictions, and optimization. The model can be used for the evaluation of

parameters. The effect and significance of each parameter can be seen from the plot of main effects (Fig. 4). The time reaction was the most significant factor for MG content. Verification was done to saw consistency MG products by looking at value Coefficient of Variation (CV). Verification based on optimum conditions performed five replications of the optimum MG content parameters (temperature 46.92 ° C&time 1.1 hours). Consistency is evaluated based on the CV. Verification result shown that MG content was 83.19 %±3.51 and CV value was 4.25%. Verification result is higher that predictive value from response surface analysis (82.96%).

Conclusion

Semi continuous esterification enzymatic in circulated packed bed reactor had residence time of 23.57 minute, glycerol/ oil molar ratio of 5:1, solvent/substrat ratio of 8.8:1 (wt/wt), and the process produced MG up to 80%. Optimization was obtained quadratic equation which was $Y= - 61.700 + 6.088\ x_1+3.259\ x_2 – 0.065\ x_1^2 + 0.017\ x_1x_2 – 1.792\ x_2^2$ with $R^2 = 0.5408$. The optimum temperature and time reaction of 46.92^oC and 1.1 hour, respectively and predictive value for MG content was 82,96%.

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Evaluation Drought Tolerance Level of Sweet Potato (*Ipomoea batatas* L.) Germplasm from NTT Province

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Abstract

Sweet potato is one of the most promising staple crops in NTT; however, the crop yield and quality in this region is relatively low due to erratic climatic condition and low genetic potency of the genotypes cultivated. NTT province is rich of sweet potato germplasm that can serve as basis for selection of high yielding varieties which are able to cope with erratic climatic condition of the region. Twenty five sweet potato genotypes (21 local clones and 4 check varieties) were elucidated to identify high yielding genotypes with tolerance to drought stress. The study was carried out in a split-plot design with three replications in field experimental station at Faculty of Agriculture, Nusa Cendana University, during June to November 2008. Two irrigation regimes (optimum and stress conditions) were assigned as main plot and 25 sweet potato genotypes as sub-plot. Results of the study revealed significant interaction effect between irrigation regimes and sweet potato genotypes on variable observed. Root yields of most genotypes tested were quite low indicating high stress condition of the trial location. Two check varieties, Sari and Cangkuang, produced only about one half and one quarter of their yield potential on optimum and stress conditions, respectively. Fourteen of 25 genotypes tested produced tuber yield above 10 t ha^{-1} in optimum condition but only five genotypes did similarly in stress condition. The highest root yields were produced by local genotype LB-01, 32.4 and 17.5 t ha^{-1} in normal and stress conditions, respectively. LB-01, NBN-01 and the check variety Kidal performed the best in optimum condition and LB-01, ON-06, and Kidal performed the best in stress condition. Only three local genotypes (LB-01, NBN-01, ON-06) and the check variety Kidal were considered tolerant and high yielding based on STI, GMP, SSI and YL selection indices.

Keywords:drought tolerance, genotype, germplasm, sweet potato, yield potential.

Introduction

In Indonesia, sweet potato [*Ipomoea batatas* L.) is the second most important root crop after cassava, in terms of harvest area, tuber production, and consumption. Sweet potato was used as food, feed, and raw materials for industries since it has potential carbohydrate, vitamin, and mineral sources (Juanda & Cahyono, 2000; Rabah *et al*., 2004). Due to its high nutritional content, sweet potato is the major staple food in several provinces in Indonesia such as Papua, Papua Barat, and some parts of other provinces like NTT, NTB, etc.

In NTT province, sweet potato has been grown for generations by local farmers and has been used as substitute for maize during famine period. However, sweet potato can not be relied on due to low root yield and root quality obtained by the farmers. This can be attributed to several factors such as erratic climatic condition, traditional farming practice, and low yield potential of the cultivars grown. Among these, erratic climatic condition is the most important and permanent factor in this region where long dry season and drought

stress is the main constraint for crop production. Development of sweet potato cultivars with high yield potential and tolerance to drought stress is the best option to solve this problem. One of the main goals in breeding programs is selection of the best genotypes under drought stress conditions (Richards *et al*., 2002). Availability of local sweet potato germplasm with wide genetic diversity (Ndiwa *et al*., 2007) is genetic resources that can be selected and utilized to develop high yielding and drought tolerant sweet potato varieties adapted to NTT agro-climatic conditions.

Water deficit, commonly known as drought, can be defined as the absence of adequate moisture necessary for normal plant growth and to complete the life cycle (Zhu, 2002). A number of selection indices have been proposed to determine drought tolerance in many crop species, including stress tolerance index (STI; Fernandez, 1992), stress susceptibility index (SSI; Fischer and Maurer, 1978), percent yield loss (YL; Blum, 1980), and geometric mean productivity (GMP; Golbashy *et al*., 2010). Some researchers found that the cultivars which had the lowest SSI values were more drought resistant than cultivars with highest SSI values (Bahar and Yaldirim, 2010; Sio-Se *et al*., 2006). On the other hand, cultivars which had high STI values were drought tolerant and those with low STI values were drought susceptible. Golbashy *et al*., (2010) found that STI and GMP indices have similar ability to separate drought sensitive and tolerant genotypes, which are in line with observation by Sio-Se *et al*., (2006). These indices, therefore, can also be employed to select high yielding and drought tolerant sweet potato genotypes, specifically adapted to semi arid climatic condition of NTT province. Therefore, it is necessary to identify selection indices able to distinguish high yielding and drought tolerant sweet potato cultivars in drought stress and in non stress (optimum) conditions. The present study was carried out to determine yield potential of local sweet potato germplasm from NTT and 2) to determine sweet potato genotypes with good drought tolerance level based on the selection indices employed.

Materials and Methods

Site Description and Treatments

This experiment was carried out in Research Station of Agriculture Faculty, University of Nusa Cendana, during dry season 2008 (June to November 2008). Research site location was located at 100 m above sea level, and the soil was an Alfisol (USDA) or Luvisol (FAO). A split plot design was employed consisted of irrigation regime as the main plot and sweet potato genotype as sub-plot treatments. Irrigation regime was of two levels, i.e. minimum/stress condition and normal (optimum) conditions. In both stress and normal condition, the plants were irrigated once a day during the first three days after planting, followed by three days interval irrigation until one month after planting. Irrigation treatments were applied at one month after planting until harvest (5 months after planting). The plants were irrigated once in two weeks in the stress condition and once a week in normal condition. In the sub-plot treatment, 25 sweet potato genotypes were included, consisted of 4 check varieties (obtained from Balitkabi Malang) and 21 landraces (local genotypes) obtained from various parts of NTT province (Ndiwa *et al*. 2007). All treatments consisted of three replicates, and in total, 75 experiment units were observed.

Planting of Sweet Potato

After land preparation**,** the sweet potato genotypes were grown in a single row plot of 3 m length and 1 m width, and the space between the plots was 1 m. Shoot cuttings of 25 – 30

cm in length with 4-5 nodes were planted at a spacing of 50 cm between plant and 1 m between rows. A total of 5 cuttings were planted in each row. Field plots were subjected to normal agronomic practice except irrigation treatment that was applied one month after planting. Fertilizer was applied at sowing time with composite fertilizer (36% P_2O_5, 36% N and 36% KCL) at a rate of 150 kg ha^{-1}.

Observation

The main variable observed in this experiment was root yield per plot that was later converted to yield per ha. Root yield parameter was used to assess drought tolerance level based on the following indices (Fischer and Maurer, 1978; Blum, 1980; Fernandez, 1992; Sio-Se *et al.,* 2006).

Stress Tolerance Index (**STI**) = $\frac{\boldsymbol{Yp\ x\ Ys}}{\boldsymbol{(Y.p)^2}}$

Stress Susceptibility Index (**SSI**) = $\frac{\boldsymbol{1-(Ys/Yp)}}{\boldsymbol{1-\ (Y.s/Y.p)}}$

Yield Loss (***YL***) = $\left(\frac{\boldsymbol{(Yp-Ys)}}{\boldsymbol{Yp}}\right)\mathbf{x\ 100}\%$

Geometric Mean Productivity (**GMP**) = $\sqrt{(Yp)(Ys)}$

Where **Ys** and **Yp** are stress and non-stress (potential) yield of a given genotype, respectively. **Y.s** and **Y.p** are average yields of all genotypes under stress and non-stress conditions, respectively. Data were recorded from 5 plants in each plot for root yield and were subjected to variance analysis appropriate for split plot design using Gentstat ver. 12 software's. Means were compared using Duncan's Multiple Range Test at 0.05 level of probability when F test is significant. Correlation analysis was also carried out among drought stress indices employed.

Results and Discussion

Root Yield Distribution

Variance analysis revealed significant interaction effect between irrigation levels and genotypes on root yields of sweet potato genotypes (data not shown). As with irrigation by genotype interaction, each of the main plot treatment (irrigation) and sub-plot treatment (genotype) also exhibited significant effect on root yields. Mean root yields of the genotypes under normal (Yp) and stress (Ys) conditions are presented in Table 1. Average root yields of the 25 genotypes tested was 12.6 t ha^{-1} in normal condition and 6.0 t ha^{-1} in stress condition. On average, root yields decline was about 50% in stress condition compared to optimum condition. This decline was much higher than that observed by Ekanayeke *et al.*, (1990) indicating high drought stress index (0.6) of the trial condition. Yields of the four check varieties were far below their potential yield. Only two of the four check varieties produced root yield above 10 t ha^{-1}, i.e. Kidal (23.7 t ha^{-1}) and P. Solossa (20.1 t ha^{-1}).

In normal condition, root yields of all local genotypes, except LB-01 and NBN-01, were lower than two check varieties, Kidal and P.Solossa; however, six local genotypes (28% of all

local genotypes evaluated) produced root yield above 10 ton.ha^{-1} and were higher than the other two check varieties, Sari and Cangkuang. The latter two varieties have potential yield between 25-35 t ha^{-1} implying that the six local genotypes mentioned previously will potentially have higher yield potential than the two check varieties. The research results showed only two local genotypes, i.e. LB-01 (32.4 t ha^{-1}) and NBN-01 (23.7 t ha^{-1}) that produced root yield above 20 t ha^{-1}. Thus, in total only 16% or 4 out of 25 genotypes evaluated (two local genotypes and two check varieties) produced root yield > 20 t ha^{-1} in normal condition.

In stress condition, only five genotypes, one check varieties (Kidal) and 4 local genotypes (LB-01, NBN-01, ORM-02, ON-06) yielded more than 10 t ha^{-1} fresh tuber. The rests of local genotypes and check varieties produced lower tuber yield, mostly far below 10 t ha^{-1} in stress condition. Even, the local genotype NMT-01 produced no tuber at all as it suffered significant reduced growth and development or died before harvest due to the stress condition imposed.

Table 1. Average root yields of sweet potato genotypes under optimal (Yp) and stress (Ys) conditions, and calculated drought tolerance indices.

Genotype	Tuber yield (t.ha^{-1})			YL	SSI	STI	GMP
	Yp	Ys	MP				
Cangkuang*	11.7 ^{e}	4.1 ^{cdefg}	7.9	65.1	1.3	0.3	6.9
EBS-01	14.0 ^{f}	3.4 ^{bcde}	8.7	76.0	1.5	0.3	6.9
EBS-02	4.6 ^{a}	2.1 ^{bc}	3.3	55.4	1.1	0.1	3.1
Kidal*	23.7 ^{j}	14.8 ^{l}	19.2	37.7	0.7	2.2	18.7
LB-01	32.4 ^{k}	17.5 ^{m}	25.0	45.8	0.9	3.6	23.8
LBK-01	9.4 ^{cd}	6.5 ^{h}	8.0	30.3	0.6	0.4	7.8
NBN-01	23.6 ^{j}	12.2 ^{k}	17.9	48.6	0.9	1.8	16.9
NFB-01	4.7 ^{ab}	2.9 ^{bcd}	3.8	38.2	0.7	0.1	3.7
NLK-01	9.3 ^{cd}	5.4 ^{fgh}	7.3	41.4	0.8	0.3	7.1
NMT-01	6.3 ^{b}	0.0 ^{a}	3.2	100	1.9	0.0	0.0
NPL-01	6.4 ^{b}	2.6 ^{bcd}	4.5	60.0	1.2	0.1	4
ON-02	16.9 ^{h}	5.6 ^{gh}	11.3	66.8	1.3	0.6	9.8
ON-03	6.4 ^{b}	3.0 ^{bcd}	4.7	53.0	1.0	0.1	4.4
ON-05	8.5 ^{c}	6.3 ^{h}	7.4	25.8	0.5	0.3	7.3
ON-06	17.0 ^{h}	14.1 ^{l}	15.6	17.4	0.3	1.5	15.5
ON-07	15.2 ^{fg}	5.7 ^{gh}	10.4	62.3	1.2	0.5	9.3
ORM-02	16.0 ^{gh}	10.2 ^{j}	13.1	36.2	0.7	1.0	12.8
ORM-03	5.8 ^{ab}	3.5 ^{bcdef}	4.6	40.6	0.8	0.1	4.5
P. Solossa*	20.1 ^{i}	8.4 ^{i}	14.3	58.2	1.1	1.1	13
PSU-02	10.7 ^{de}	1.6 ^{ab}	6.2	84.6	1.6	0.1	4.2
PSU-03	11.6 ^{e}	4.2 ^{defg}	7.9	63.6	1.2	0.3	7.0
PSU-04	9.6 ^{cd}	3.3 ^{bcde}	6.4	65.8	1.3	0.2	5.6
Sari*	12.2 ^{e}	5.2 ^{efgh}	8.7	57.1	1.1	0.4	8.0
SEO-01	12.2 ^{e}	5.2 ^{efgh}	8.7	57.4	1.1	0.4	7.9
TBU-02	6.1 ^{ab}	3.2 ^{bcde}	4.7	48.0	0.9	0.1	4.4
Mean	12.6	6.0	9.3	53.4	1.0	0.6	8.5

*National Check variety, Yp (normal/optimal condition), Ys (stress condition), MP (mean productivity), YL (percent yield loss), STI (stress tolerance index), SSI (stress susceptibility index), GMP (geometric mean productivity).

Level of Tolerance to Drought

Genotypes evaluated were selected based on calculated drought tolerance indices employed (Table 2). Similar number of genotypes was selected based on high yielding performance, respectively, LB-01, Kidal, NBN-01, P.Sollosa in normal, and LB-01, Kidal, NBN-01, ON-06 in stress conditions. This implies that the genotypes that yielded better in normal condition tended to do so in stress condition.

Drought tolerance levels of the genotypes tested were assessed based on four selection indices, i.e. stress tolerance index (STI), stress susceptibility index (SSI), percent yield loss (YL), and geometric mean productivity (GMP). Research results reveal that STI values of the genotypes ranged from 0.0 to 3.6 (Table 1). Only five genotypes were considered tolerant and were selected, based on STI values (≥1.0), i.e. LB-01, NBN-01, Kidal, P. Solossa, and ON-06 (Table 1 and 2). Sweet potato genotypes with high STI values are considered drought tolerant while those with lower STI values were considered drought susceptible (Fischer and Maurer, 1978). The five genotypes considered tolerant based on STI were also considered tolerant based on geometric mean productivity, GMP (Table 2). This finding was supported by highly significant correlations observed between the two selection indices (Table 3). It is interesting to note that four of the five genotypes selected based on STI and GMP were also the best yielding genotypes in both normal and stress conditions (Table 1 and 2). This implies that STI and GMP are reliable selection indices to select for drought tolerant and high yielding genotypes in both normal and stress condition. Results of this study were closely in line with previous works (Sio-Se *et al*., 2006; Golbashy *et al*., 2010).

Stress susceptibility index (SSI) of the genotypes tested (Table 1) ranged from 0.3 (ON-06) to 2.3 (NMT-01). Selection based on SSI favored genotypes with low SSI values (<0.75), i.e. ON-03, ON-05, Kidal, ORM-02, and LBK-01. A high SSI value indicates high susceptibility to drought while a low SSI value indicates tolerance to drought (Fischer and Maurer, 1978). The five most tolerant genotypes selected on the basis of SSI were also the most tolerant on the basis of percent yield loss (YL) (Table 2). Genotypes with low SSI values and low proportion of yield reduction (YL) in stress condition were identified as tolerant genotypes; however, these genotypes were in general low yielding in both normal and stress condition (Table 1 and 2). Out of five tolerant genotypes selected based on SSI and YL, only Kidal and ON-06 produced high yield (above 10 $t.ha^{-1}$) in both normal and stress conditions. This implies that SSI and YL were reliable indices to select for drought tolerant genotypes but partly determinative in selecting the best yielding genotypes in both normal and stress condition. Similar results were reported by other workers (Bahar and Yaldirim, 2010; Sanjari, *et al*., 2010; Golbashy *et al*., 2010).

Positive and significant correlation between Yp and Ys with STI and GMP (Table 3) indicates that STI and GMP can be used to select for drought tolerant and high yielding genotypes in normal and stress conditions. On the contrary, negative and significant correlation between Ys and either SSI or YL implies that SSI and YL were able to detect drought tolerant genotypes but partly effective in selecting high yielding genotypes in stress condition.

Overall, results of the present study revealed eight drought tolerant sweet potato genotypes on the basis of four selection indices, i.e. STI, SSI, YL and GMP. However, only four of the eight genotypes were drought tolerant and high yielding in both normal and stress condition.

Table 2. Selected genotypes based on drought tolerance indices

Selected genotypes	Drought tolerance indices
LB-01, Kidal, NBN-01, P.Solossa	Yp
LB-01, Kidal, NBN-01, ON-06	Ys
LB-01, Kidal, ON-06, NBN-01, P.Solossa	STI
LB-01, Kidal, NBN-01, ON-06, P.Solossa	GMP
ON-06, ON-05, Kidal, LBK-01, ORM-02	SSI
ON-06, ON-05, Kidal, ORM-02, LBK-01	YL

Yp (normal/optimal condition), Ys (stress condition), MP (mean productivity), YL (percent yield loss), STI (stress tolerance index), SSI (stress susceptibility index), GMP (geometric mean productivity).

Table 3. The correlations between selection indices and mean yiled of sweet potato genotypes under normal and stress conditions.

	Ys	YL	STI	SSI	GMP
Yp	0.92**	-0.19^{ns}	0.93**	-0.18^{ns}	0.84**
Ys		-0.50*	0.92**	-0.48*	0.57**
YL			-0.26^{ns}	0.99**	0.29^{ns}
STI				-0.24^{ns}	0.69**
SSI					0.70**

**: Significant at P≤0.01 level, *: Significant at P≤0.05 level, ns: Not significant

Conclusions

The present study demonstrated significant effect of drought stress on sweet potato genotype's root yields. Local genotypes LB-01 (32.4 t ha^{-1}), NBN-01 (23.6 t ha^{-1}), and check variety Kidal (23.7 t ha^{-1}) were the best genotypes under normal condition and local genotypes LB-01 (17.5 t ha^{-1}), ON-06 (14.1 t ha^{-1}), and check variety Kidal (14.8 t ha^{-1}) were the best genotypes under stress condition. Three local genotypes (LB-01, NBN-01, ON-06) and the check variety Kidal were selected as drought tolerant and high yielding varieties in both normal and stress condition based on STI and GMP. These two selection indices were reliable to be employed in selection of drought tolerant and high yielding sweet potato genotypes in future works.

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Laboratory Bioassay of Entomophatogenic Fungi *Beauveria Bassiana* and *Metarrhizium Anisopliae* for Control of Sweet Potato Weevil (*Cylas formicarius* Fab.)

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Abstract

Sweet potato weevil (Cylas formicarius Fab.) is an important pest of sweet potato in the tropics, including Indonesia. The weevil is not easily controlled using conventional method of insecticide spray since the weevil stays and feeds inside the sweet potato tubers and the stems. Alternative control method that could be more effective includes the use of entomopathogenic fungi such as Beauveria bassiana and Metarrhizium anisopliae. This study was carried out to investigate the potential of these two entomopathogenic fungal species to control C. formicarius. Several conidial concentrations of B. bassiana and M. anisoplia were sprayed on C. formicarius adults and the insect mortality was observed daily until the last insect died. Research results showed that percentage of insect mortality at 7 day after inoculation (DAI) ranged from 10 to 80% for B. bassiana and 15 to 90% for M. anisopliae inoculations, depending on the conidial concentrations tested. At the lowest conidial concentration (10^1 condia.mL^{-1}), the test insects inoculated with B. bassiana were all dead at 22 DAI while those inoculated with M. anisoplia were all dead at 19 DAI. The median lethal concentration (LC_{50}) of M. anisopliae to kill 50% of the test insects (9.66×10^4 conidia.mL^{-1}) was lower than that of B. bassiana (1.28×10^6 conidia.mL^{-1}). Similarly, median lethal time (LT_{50}) of M. anisopliae (3.99 days) was shorter than that of B. bassiana (4.77 days). The results demonstrated that the two fungal species were pathogenic on C. formicarius. Although M. anisopliae had lower LC_{50} and LT_{50} than B. bassiana, effectiveness of the two fungal species against C. formicarius was not significantly different due to overlapping fiducial limits of the two parameters.

***Keywords: Beauveria bassiana, bioassay, Cylas formicarius, Metarrhizium anisopliae*.**

Introduction

Sweet potato (*Ipomoea batatas* L.) is the second most important tuber crop in Indonesia. In many parts of Indonesia, including NTT province, sweet potato is used as the main staple food due to its high carbohydrate content and sources of vitamins (A, B_1, B_2 dan C) and essential elements (Juanda & Cahyono, 2000). Sweet potato production in NTT province fluctuated year by year, and total tuber production in the region in 2009 was 99,233 t (BPS NTT, 2009). This fluctuation was caused by many factors such as fluctuation in total harvest area, climatic condition, and yield loss due to major pests and diseases. One of the most important pests of this crop is sweet potato weevil (SPW) (*Cylas formicarius*) that can cause severe yield loss (up to 80%) and significant decreased tuber quality (Mao *et al*., 2004; Zuraida *et al*., 2005; Juanda & Cahyono, 2000).

C. formicarius is difficult to control using conventional control method such as pesticide spray since the weevil feeds and lives within the stems and tubers of sweet potato. A more effective control measure would be through biological control method such as application

of entomopathogenic fungi *Beauveria bassiana* and *Metarrhizium anisopliae*. These two fungal species were found to be effective against various insect pest species such as *Anisopliae australiae* and *Oryctes rhinoceros* (Lolong & Soekarjoto, 1990), *Heliothis armigera* (Tjahjadi, 1989), *Callosobruchus maculates* (Murad *et al*., 2006), fruit fly (Toledo *et al*., 2006), *Polyphagotarsonemus latus* (Nugroho & Ibrahim, 2004), *Delia radicum* (Bruck *et al*., 2005), olive fuit fly, *Bactrocera oleae* (Mahmoud, 2009), and *Tetranychus cinnabarinus* (Shi & Feng, 2004).

Previous studies revealed that lethal effect of *B. bassiana* and *M. anisopliae* varied among various insect pest species. Conidial concentration of *M. anisopliea* to kill 50% population of test insects, for instance, was about 10^4 conidia mL^{-1} for *C. maculates* (Murad *et al*., 2006), and $2.77x10^7$ conidia mL^{-1} for *P. latus* (Nugroho & Ibrahim, 2004). As a compariosan, Nugroho & Ibrahim (2004) also reported an LC_{50} of $2.74x10^6$ conidia mL^{-1} *B. bassiana* against broad mite (*P. latus*). In addition, Toledo *et al.*, (2006) reported different LC_{50} and LT_{50} values of either *M. anisopliae* or *B. bassiana* isolates against different species of fruit fly (*Anastrepha ludens* and *Ceratitis capitata*). These results indicate wide pathogenicity range of the two entomopathogenic fungal species.

The existence of wide pathogenicity range of these fungal species implies that effectiveness of these fungi as biocontrol agents depends on their interaction with the host species. Therefore, it is important to carry out a bioassay study prior to employing these fungi as biocontrol agents for an insect pest species. Considering the wide host range of *B. bassiana* and *M. anisopliae*, it is plausible to assume that these two entompathogenic fungal species are also potential biocontrol agents for sweet potato weevil, *C. formicarius*. However, published results of effective concentrations of these two fungal species against *C. formicarius* are very limited. This study was, therefore, carried out to determine effective conidial concentration of *B. bassiana* and *M. anisopliae* against *C. formicarius*.

Materials and Methods

This study was carried out in Microbiology Laboratory, Faculty of Agriculture, University of Nusa Cendana, from July to September 2010. A laboratory experiment method was empoyed consisted of several conidial concentations of *B. bassiana* and *M. anisopliae*. The concentrations tested were 10^1, 10^3, 10^5, 10^7 and 10^9 conidia mL^{-1} for each fungal species. A blank control treatment using distilled water spray was also included in each bioassay. The test insects were *C. formicarius* adults that were mass propagated following the method of Zuraida *et al.,* (2005) to obtain homogeneous and same age test insects. As many as 20 insects were exposed in every concentration treatment, each with three replicates.

B. bassiana and *M. anisopliae* isolates were obtained from Balai Proteksi Tanaman, Dinas Pertanian dan Perkebunan Provinsi NTT. Before inoculation, the fungal isolates were grown in the laboratory to obtain pure culture followed by fungal indentification based on the procedures proposed by Watanabe (2002). Conidial density of either *B. bassiana* or *M. anisopliae* in the stock medium was calculated by examining the stock solution (100 µl condial suspension) using a haemocytometer, and serial dilutions were made to obtain the working concentrations for the bioassay. Inoculation was carried out by applying 10 µl conidial suspension using micro pippet on the surface of adult C*. formicarius* prepared in a petridish (20 insects per petridish) and maintained at 25°C. The base of the petridish was covered with wet towel paper and wet cotton to maintain high moist condition (80% RH).

Food for the test insects was freshly provided daily, in forms of pieces of sweet potato tuber.

The test insects were observed daily for mortality from one day after inculation (DAI) until the last insect died. Percent mortality was calculated based on the following formula:

$$\%\ mortality = \frac{Number\ of\ dead\ insects}{Number\ of\ dead + alive\ insects}\ x\ 100\%$$

Percent mortality data at 7 DAI were subjected to probit analysis to generate a concentration-mortality relationship for the estimates of LC_{50} and LC_{99}, and their Fiducial Limits (FL) for each fungal species. In addition, LT_{50} and LT_{99} values were also estimated based on daily (seven days) mortality data of each fungal isolate at comparable concentration 10^9 conidia mL^{-1}. The difference in effectiveness between two fungal species was considered significant when 95% Confidence Interval (Fiducial Limit) did not overlap. All data analysis was performed using a Genstat release 12 software package.

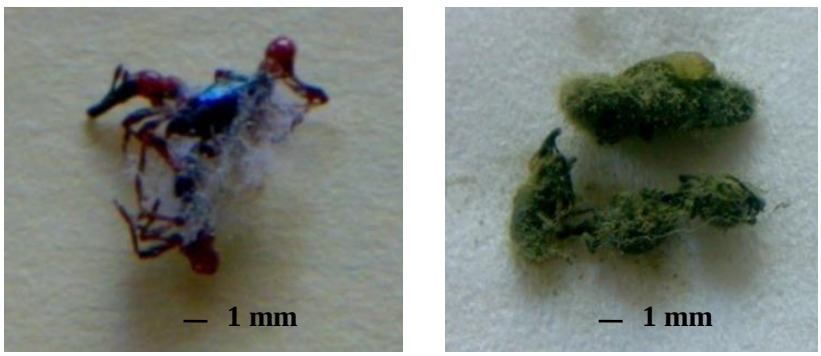
Results and Discussion

Characteristics and Viability of *B. bassiana* and *M. anisopliae*

B. bassiana dan *M. anisopliae* condia started to grow on a PDA medium at 3 DAI and the colonies of the two fungi fully covered the petridish (Ø: 9 cm) at 6 DAI and 7 DAI for *B.bassiana* dan *M. anisopliae*, respectively. *M. anisopliae* colony was first whitish in color but later became dark green, and the shape of the condia were round, consistent with previous observation by Prayogo *et al*. (2005). The colony of *B. bassiana* appeared white in color during the growing stage in the petridish and the conidia were round. The conidia shape observed in this study was inline with the previous results by Sudarmadji (1997).

Conidia of either *B. bassiana* or *M. anisopliae* released from condiophore will immediately sporulated in a moist condition. Results of the present study showed that conidial viability of *B. bassiana* and *M. anisopliae*, were, respectively, 85.13% and 86.45%, which were considered high and were eligible to be employed in the bioassay study.

Symptoms and Signs of The Infected Insect



(a) (b)

Figure 1. *C. formicarius* adults infected with (a) *B. bassiana* and (b) *M. anisopliae*.

C. formicarius adults infected with either *B. bassiana* or *M. anisopliae* showed physical symptoms such as very weak and slow movement, and later became paralysis. The test insect infected with *B. bassiana* started to die one day after inoculation using 10^9 conidia mL^{-1}, and *B.bassiana* sporulation over the weevil's surface took a whitish color (Figure 1a). Nugroho and Ibrahim (2004) reported similar whitish color over surface of broad mite's cadaver 4-5 days after treatment with *B. bassiana* conidia. Unlike *B. bassiana*, *M. anisopliae* appeared greenish when sporulation occurred on the weevil's surface (Figure 1b). Similar results were reported by Nugroho and Ibrahim (2004).

Effect of *B. bassiana and M. anisopliae* Inoculation on Insect Mortality

The test insects started to die at one DAI in inoculation with 10^9 conidia mL^{-1} of each fungal species. Percent mortality on the first DAI was 10% and 15% for *B. bassiana* and *M. anisopliae*, respectively. At lower conidial concentrations between 10^7 – 10^3 conidia mL^{-1}, lethal effect of both fungal species also started to appear on the first DAI but in lower percent mortality or even appeared in the second and the third DAI. Lethal effect of *B. bassiana* at the lowest condial concentration (10^1 conidia mL^{-1}) started to occur at 6 DAI while that of *M. anisopliae* occoured at 5 DAI.

Inoculation using *B.bassiana* at various conidial concentrations killed all the test insects at 9, 12, 15, 19, and 22 DAI for 10^9, 10^7, 10^5, 10^3, and 10^1 conidia mL^{-1}, respectively, while that of *M. anisopliae* in the same range of conidial concentrations occurred at 8, 11, 14, 12, and 19 DAI. These reuslts indicate that for each of the fungal species bioassay, higher concentration killed all test insects faster than lower concentrations. Plausible explanation for this finding is that when higher conidial concentration was applied, there was a higher chance and higher number of fungal conidia that get contact with the insect surface, and hence more chance for infection to take place that may render the test insects to die sooner. Opposite situation may occur for lower conidial concentrations.

Concentration and Mortality Relationship

Mortality data of C*. formicarius* adults recorded at 7 DAI were subjected to probit analysis to obtain regression relationship between concentration and mortality. A linear relationship between conidial concentration of each fungal species and weevil's mortality was attained using Probit analysis (Table 1). The test for goodness of fit indicates no significant heterogeneity in the linear relationships for all fungal isolates tested (P>0.05) (Table 1).

The linear relationship between conidial concentration and weevil mortality as presented in Table 1 implies that the higher the conidial concentration the higher the weevil mortality will be observed. This relationship applies for both *B. bassiana* and *M. anisopliae* inoculations. Lethal concentration of *M. anisopliae* to kill 50% population of the test insects (LC_{50}) was 9.66 x 10^4 conidia mL^{-1}, which was lower than that of *B. bassiana* (1.28 x 10^6 conidia mL^{-1}). Considering to this data point alone may lead to conclusion that *M. anisopliae* was more effective than *B. bassiana* against *C.formicarius*. However, based on the 95% Fiducial Limit data (Table 1), it is clear that an overlap did occur between LC_{50} fiducial limits of the the two fungal species, which suggests that, statistically, effectiveness of the two fungal species against *C. formicarius* did not differ significantly. Similar with LC_{50}, LC_{99} of *M. anisopliae* (9.81 x 10^{12} conidia mL^{-1}) was much lower than that of *B. bassiana* (1.06 x 10^{15}conidia mL^{-1}) but overlap fiducial limits of the two species (Table 1) renders them not to differ statistically in their effectiveness against *C. formicarius*.

When log conidial concentration of each fungal species was plotted against probits of mortality, a linear response curve was observed (Figure 2). The linear response curve in Figure 2 demonstrates a good fit between observed and expected mortality data, either for *B. bassiana* or *M. anisopliae* bioassay. This good fit was supported by non significant heterogeneity in the linear relationship (P=0.85 and P=0.78 for *B.bassiana* and *M.anisopliae*, respectively). Figure 2 also demonstrates that the two response curves were very close at the lower conidial concentrations but then started to appart from each other in higher concentrations. This was an indicative of different slope of the two response curve, where response curve of *M. anisopliae* had higher slope (0.29) than *B. bassiana* (0.26) (Table 1).

Table 1. Summary of probit parameters of *C. formicarius* adults inoculated with several conidial concentrations of *B. bassiana* and *M. anisopliae*

Fungal species[a]	Slope (± SE)	Intercept	LC_{50}[b]	95% FL[c]	LC_{99}	95% FL	$\chi^2_{(P)}$[d]
Bb	0.26 (0.03)	-1.59	$1.28x10^6$	$3.19x10^5$-$6.20x10^6$	$1.06x10^{15}$	$1.30x10^{13}$-$1.03x10^{18}$	0.85
Ma	0.29 (0.03)	-1.45	$9.66x10^4$	$9.74x10^4$-$3.5x10^5$	$9.81x10^{12}$	$3.10x10^{11}$-$1.71x10^{15}$	0.78

[a]Bb (*B. bassiana*) and Ma (*M. anisopliae*), [b]LC (Lethal Concentration, in conidia mL-1), [c]FL (Fiducial limit). [d]Chi square probability of heterogeneity in linear relationship between concentration and mortality.

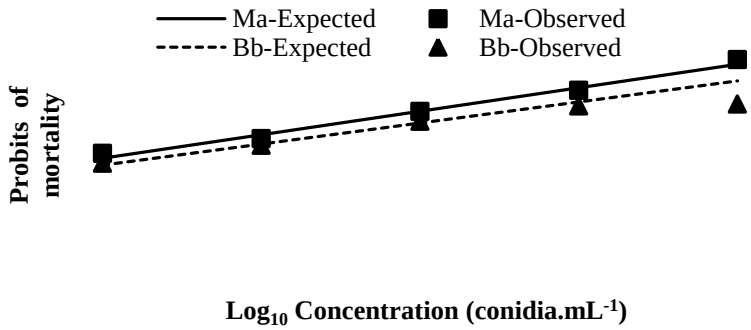


Figure 2. Response curve of log concentration of either *B. bassiana* (Bb) or *M. anisopliae* (Ma) and probits of mortality of *C. formicarius*.

Overall, the present study revealed that either *B. bassiana* or *M. anisopliae* was lethal to the sweet potato weevil, *C. formicarius*. Probit parameters was obtained in this study demonstrated that *M. anisopliae* had lower LC_{50} and LC_{99} values, and a steeper slope than *B. bassiana* indicating that the former was more effective than the latter but, statistically the two fungal isolates were not significantly different in their effectiveness against *C. formicarius* due to overlap fiducial limit. Conidial concentration of *M. anisopliae* required to kill 50% population of *C. formicarius* (9.66×10^4 conidia mL^{-1}) was very close to that of previous tests of this fungus against *C. maculates* (1.4×10^4 conidia mL^{-1})(Murad *et al*., 2006). However, it was much lower than LC_{50} of this fungus against broad mite, *P. latus* ($2,77 \times 10^7$ conidia mL^{-1}) (Nugroho & Ibrahim, 2004). LC_{50} of *B.bassiana* against *C. formicarius* (1.28×10^6 conidia mL^{-1}) was close to that of previous test of this fungus against

broad mite (2.74 x 10^6 conidia mL^{-1}) (Nugroho & Ibrahim, 2004), which indicates almost similar lethal effect of the fungal species agains the two insect species. Unlike *C. formicarius* bioassay, Nugroho & Ibrahim (2004) reported that LC_{50} of *B. bassiana* against broad mite was higher than that of *M. anisopliae*, which supports the notion that effectiveness of different entomopathogenic fungal species is determined by their interaction with the host insect species.

Time and Mortality Relationship

As with concentration and mortality relationship, a linear relationship between time (after inoculation) and weevil's mortality was also observed at concentration 10^9 conidia mL^{-1} of each fungal species (Table 2). The test for goodness of fit also indicates no significant heterogeneity in the linear relationships for the two fungal isolates (P>0.05) (Table 2).

Table 2. Time-mortality relationship of *C. formicarius* inoculation at comparable concentration 109 conidia mL-1 of *B. bassiana* and *M. anisopliae*

Fungal species[a]	Slope (± SE)	Intercept	LT_{50}[b]	95% FL[c]	LT_{99}	95% FL	$\chi^2_{(P)}$[d]
Bb	0.37(0.03)	-1.75	4.78	4.43-5.13	11.14	10.15-12.51	0.99
Ma	0.36(0.04)	-1.44	3.99	3.62-4.36	10.43	9.32-12.05	0.92

[a]Bb (B. bassiana) and Ma (M. anisopliae), [b]LC (Lethal Time, in days), [c]FL (Fiducial limit). [d]Chi square probability of heterogeneity in linear relationship between concentration and mortality

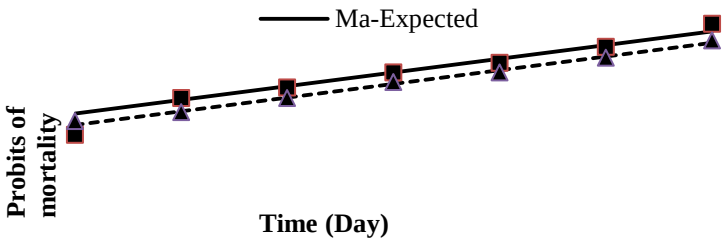


Figure 3. Response curve between time (day) after inoculation and probits of mortality at comparable concentration 109 conidia mL-1.

The linear relationship between time after inoculation and weevil mortality (Table 2) indicates that weevil's mortality increased with the increase of time after inoculation. Time (in days) of *M. anisopliae* at 10^9 conidia.mL^{-1} to kill 50% *C. formicarius* population was 3.99 days, which was lower than that of *B. bassiana* (4.78 days). As with LC_{50}, there was an overlap between LT_{50} fiducial limits of the two fungal isolates (Table 2), which suggests that the two fungal species did not differ statistically in terms of time required to kill 50% *C. formicarius* population. The same situation holds for the other probit parameter, the LT_{99}. Probit parameters in Table 2; therefore, suggest that effectiveness of *B. bassiana* and *M. anisopliae* against *C. formicarius* was not significantly different.

Similar with concentration-mortality relationship, a linear response curve was observed when time after inoculation (in days) was plotted against probits of mortality of each fungal species (Figure 3). There was a very good fit between observed and expected probits of mortality data over time, and the two response curves were apparently very closely parallel. The high parallelism between the two curves was due to the overlap, and hence non significant, slopes of the two curves (Table 2).

Results of the present study revealed that LT_{50} of either *B. bassiana* (4.78 days) or *M. anisopliae* (3.99 days) was not substantially different from that of previous bioassay of these two fungal species on *P. latus* at 10^8 conidia mL^{-1}, where time required to kill 50% *P. latus* population for *B. bassiana* was 3.35 days and that for *M. anisopliae* was 4.28 days (Nugroho & Ibrahim, 2004). Overall, the present study results demonstrate that both *B. bassiana* and *M. anisopliae* were effective against *C. formicarius* in this laboratory bioassay. These two fungal isolates are, therefore, potential biocontrol agents for sweet potato weevil, *C. formicarius*, and further studies for their application in the field are required.

Conclusions

1. Entomopathogenic fungi *B. bassiana* and *M. anisopliae* proved effective against *C. formicarius* with LC_{50} values of 1.28×10^6 and 9.66×10^4 conidia mL^{-1}, respectively.
2. The LT_{50} of *B. bassiana* against the weevil was 4.78 days while that of *M. anisopliae* was 3.9 days.
3. *M. anisopliae* exhibited lower LC_{50} and LT_{50} values than *B. bassiana* but the two fungal species were considered not significantly different in their effectiveness as biocontrol agents for *C. formicarius*.

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Simulation Model For Corn (*Zea mays*, L.) Planting Time Determination In Dryland Of Timor, East Nusa Tenggara Province

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Abstract

In the last decade, corn farmers in West Timor have frequently experienced planting failure due to erratic rainfall and dry spells. A reliable planting time determination will be highly appreciated not just by farmers but by politicians alike. This study was meant to build a model of planting time for corn based on crop water requirement, rainy season forecast, water balance, and dry spell occurrences. Crop water requirement is specifically assessed for seed germination and early growth of seedlings on representative soils (an Entisols and a Vertisols). Water balance was based on Cocheme-Franquin model in determining water surpluses and deficits during the year. Rainfall forecasting was based on a combination of ARIMA model and SOI values. Dry spells were calculated based on Oldeman and Frère method. Results showed that there were three determined factors in planting time of corn namely: water deficit (rainfall minus ETo), SOI values, and dry spell (rainfall < 3 mm) probability during two consecutive days in a decade. The model is: (1). IF SOI 3 preceding months >+7 THEN Planting = 1 ELSE Planting = 0; (2). IF PDD < 50% THEN Planting time = 1 ELSE Planting time = 0; (3) IF (Rainfall-ETo)>0>soil water deficit THEN Planting time = 1 ELSE Planting time = 0.

Keywords: ***corn (Zea mays), dry spell, planting time, Southern Oscillation Index (SOI), Timor***

Introduction

In the last 10 to 20 years, dryland corn farmers in dry areas (i.e. areas of class D, E, and F according to Schmidt and Ferguson classification system) of Timor have been experienced planting failure due to erratic rainfall's frequent occurrence. According to Badan Meteorologi, Klimatologi, dan Geofisika (BMKG) (2009), these areas comprise more than 90% of West Timor. Included in this erratic nature of rainfall are dry spells. Even though, farmers in these areas already had their own planting time determination and dry spell avoidance methods based on long time experiences, these methods have been upset by the latest changes in the rain pattern.

Farmers may eventually adjust to this situation and develop new methods to cope with the new climate pattern. However, it will probably take generations for them to make such adjustments themselves and this will also mean prolonged planting and crop failures for the time. Therefore, an intervention in the form of planting time determination methods from out side the farmers should be fabricated. The intervention has to be in the form of appropriate methodologies in planting time determination by taking advantage of present

scientifically and empirically proven methodologies on rainy season onset and crop water requirement determinations.

Various climatological and meteorological tools such as water balance (Kasei and Afuakwa, 1991; de la Casa, 2009; Geru *et al*., 2010) and Southern Oscillation Index (SOI) (Punyawardena and Cherry. 1999; Haylock, M and J. McBride. 2001; Kirono and Partridge, 2002; Aldrian and Susanto, 2003; Kirono and Sulaswono, 2003; Korecha and Barnston, 2007) have been utilized to determine the onset of a rainy season and the length of growing period in tropical areas. Geru *et al*. (2010) applied water balance method to determine the growing season in Timor. Aldrian and Susanto (2003) agreed that there is a significant response of the rainfall pattern to ENSO (El Nino Southern Oscillation) in Timor for five to six months (from June to November) prior to the onset of the rainy season. An accumulation of rainfall of 50 mm in a decade is also frequently used by BMKG to establish the onset of a rainy season (personal communication with A. Geru).

When applying the water balance method, all the above authors used the start of the rain water surplus as the beginning of the growing period and plant water requirements to determine the length of the growing season (Kasei and Afuakwa, 1991). However, the planting time of corn does not always coincide with the onset of the rainy season. The planting time of corn has to consider the amount of water to initiate seed germination and to ensure a vital growth of the seedlings. Therefore, corn planting may not start soon after the onset of the rainy season. Corn planting will start when the water surplus exceeds or at least the same as the amount of soil moisture needed for germination (during the establishment stage of corn). This will be reached when the soil profile around (a little bit deeper than) the planting depth is filled with water to the field capacity of the soil (Doorenbos *et al*., 1986; Brady and Weil, 2002).

Beside on the above mentioned considerations, the determination of the planting time of field crops in drylands has to consider also the possibility of dry spell occurrences (Oldeman, and Frère, 1982). Normally, following the onset of the rainy season, there always be dry spells which, most of the time, result in the wilting or even the death of the newly germinating corn plants.

This study is meant to combine climatological and meteorological tools in determining the onset of the rainy season with soil moisture requirement in an integrated model to determine the planting time of corn in dryland areas of Timor, East Nusa Tenggara Province.

Materials Methods

This study was based on analyses of climatological data for Kupang area and on a field experiment. Climatological data were compiled at Lasiana BMKG Station of Kupang, Timor, East Nusa Tenggara Province. Climatological data were daily rainfall and potential evapotranspiration, and monthly southern Oscillation Index and were utilized to predict the onset of rainy season and the decadal (a ten day period) water balance. The field experiment was conducted during the dry season of 2009 (late August to early November) at BPTP experiment station in Naibonat Kupang, Timor, East Nusa Tenggara Province (Figure 1) to study the used of rice straw mulching on corn field as an appropriate technology to overcome soil moisture shortages during a dry spell occurrence.

Daily rainfall records for 21 years from 1985 to 2005 were analyzed for average decadal rainfall, frequency and probability of having a decade with one and two consecutive dry-

spell days. Dry spell analyses (definition, decadal frequency and its probability) were based on the procedure suggested by Oldeman and Frère (1982). Dry spell was defined as a day with less than 3 mm rainfall. Decadal frequencies and probabilities were calculated for a decade which have one day and for a decade with two consecutive days of dry spell. Decadal rainfall was also used for a random walk autoregressive integrated moving average (ARIMA) model in forecasting the onset of the rainy season in Kupang. The onset of the rainy season starts when an accumulation of 50 mm in a decade had been reached.

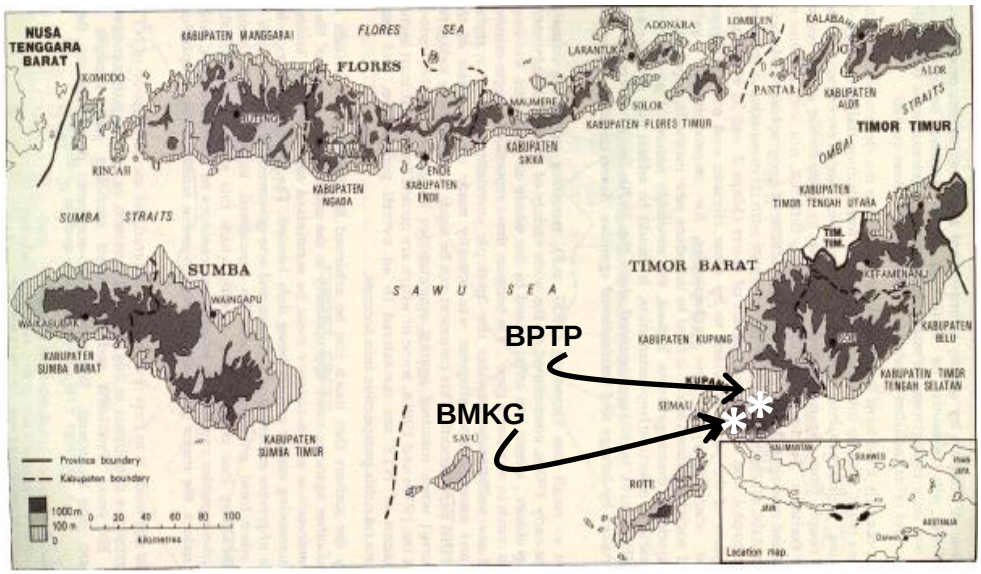

Figure 1. Field experiment location at BPTP Naibonat and location of BMKG Lasiana, Kupang, Timor, East Nusa Tenggara Province

Water balance was constructed by using the decadal rainfall and potential evapotranspiration data based on Cochemé and Franquin (1967). Monthly rainfall for 21 year from 1985 to 2005 were also utilized in building the rain regionalization of the province of East Nusa Tenggara. The regionalization produced by BMKG (Badan Meteorologi, Klimatologi, dan Geofisika, 2009) which was based on Schmidt and Ferguzon classification system.

Monthly Southern Oscillation Indices (SOI) were averaged for every three months during the period of June to October of 2010 i.e June to August, July to September, and August to October. The average SOI for the three-month periods above +7 became the threshold of the onset of rainy seasons.

Soil moisure deficit was calculated based on the difference between the present soil moisture during the dry season (prior to rainy season) and the soil moisture at field capacity. For this purpose, three soil samples extracted from the field of Badan Pengkajian Teknologi Pertanian (BPTP) Naibonat, Kupang and from the field in the BMKG station of Lasiana Kupang (Figure). Soil samples from these areas were to represent deep (solum > 120 cm) and shallow (solum < 20 cm) soils of Timor, respectively. The soil samples were used to determine their present gravimetric soil moisture contents and the field capacity values.

The field experiment at BPTP Naibonat was a one factor experiment and arranged in a Completely Randomized Block Design (RCBD) with two treatments i.e. with mulch and without mulch. Every treatment was laid down in three replications. The dimension of an experimental plot was 14 m x 3 m. Twenty one kilograms of dry rice straw was evenly distributed on each mulch treated plot. Two seeds of BISI–2 corn variety was planted per dible hole at 50 cm by 70 cm spacings and watered based on crop water requirement calculations suggested by Doorenbos *et al*. (1986) i.e. Etcrop = Eto x kc for each corn growth stage. Time of watering was determined by only inspecting the corn plant indicator i.e. when the corn leaves start to curl (a deficit irrigation). This was done according to the farmers practice in time of watering.

Rainy Season Forecasting Sub-Model

The presentation of results and discussions of this study will be in a logical order to build the algorithm of the proposed planting time determination model. It starts with the presentations of rainy season forecasting sub-model followed by crop water requirement sub-model and ended with the proposed planting time model.

Southern Oscillation Index

Based on Southern Oscillation Index for 2010, the average index for three months prior to the suspected onset of the rainy season for the area i.e. October (early onset), November (normal nset), or December (late onset) were 21, 21, and 20, respectively (Table 1). An average of index >+7 has been recognized by Kirono and Partridge (2002) as the threshold SOI for the onset of rainy season especially for Indonesia. Therefore, it can be concluded that the onset of rainy season for Kupang area in 2010 can be as early as October or as late as December.

Since this tool of determining the onset of the rainy season is still ambiguous, some other tools are needed as will be explained in the next sections. However, SOI can still be included in the proposed model for determining the planting time. It is viewed as the first screen in the process of planting time model building.

Table 1. Southern Oscillation Index (SOI) and the average of three month indices prior to suspected onset of rainy season (early: October; normal: November; or late: December).

SOI	Month											
	Jan	**Feb**	**Mar**	**Apr**	**May**	**Jun**	**Jul**	**Aug**	**Sep**	**Oct**	**Nov**	**Dec**
Monthly	-10.1	-14.5	-10.6	15.2	10.0	1.8	20.5	18.8	25.0	18.3	16.4	27.1
July- Sept. average									21			
Aug.- Oct. average										21		
Sept.- Nov. average											20	

Autoregressive Integrated Moving Average (ARIMA)

Autoregressive Integrated Moving average (ARIMA) procedure for 21 years of daily rainfall records for Kupang area showed that typically the onset of rainy season for this locality is at the end of the third decade of November where the accumulated rainfall at this point of the

year exceeds 50 mm (Figure 2). This number is set by BMKG as the accumulated rainfall threshold to start a rainy season.

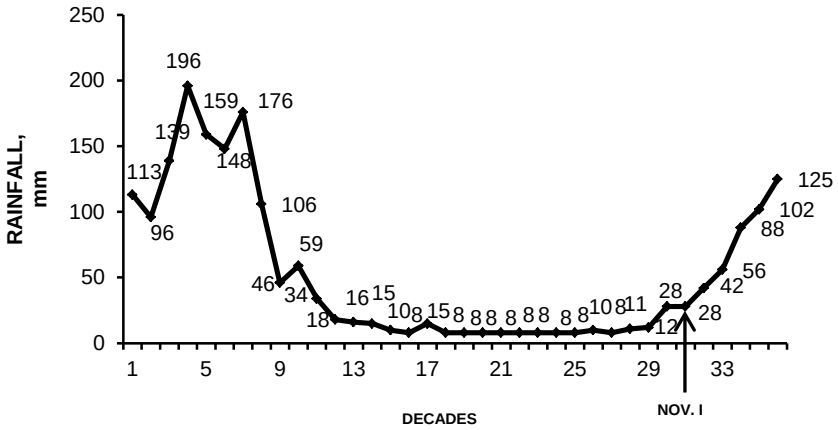

Figure 2. Estimation of rainy season onset for Kupang area based on autoregressive integrated moving average (ARIMA) which is at the end of the first decade of November (see arrow) where the accumulated rainfall up to this point is higher than 50 mm.

However, for the purpose of planting time determination, the accumulated rainfall has to surpass the potential evapotranspiration (ETo). As a consequence, the following water balance presentation should also be considered.

Water Balance

The explanation in ARIMA model has set the end of the first decade of November as the onset of the rainy season in Kupang area (see previous section). However, water balance presentation for the area (Figure 3) shows that during the first and second decades of November the rainfall is still less than the potential evapotranspiration (rainfall – Eto < 0). In other words, there is still a deficit in water availability to start a growing period. The surplus of water is reached at the third decade of November.

Nevertheless, this approach still needs to be coupled with information on the soil moisture requirement for seed germination. This can be obtained by considering the soil moisture content prior to the fall of rain to the area (when the soil is still dry) and the field capacity and the permanent wilting point of the soil.

Soil Moisture Requirement for Seed Germination Sub-model

Field capacity (FC) of soil at BMKG field was 30% (w/w) and soil moisture at dry condition prior to irrigation or rain (CW) was 22% (w/w) (Table 2). This means that before the onset of the rain, the soil was under a deficit of water as much as 8% (FC-CW). Therefore, in order to plant corn, an amount of water equal to the deficit water has to be present in the soil profile to the depth of planting. By doing this, the soil moisture content is brought to the field capacity level.

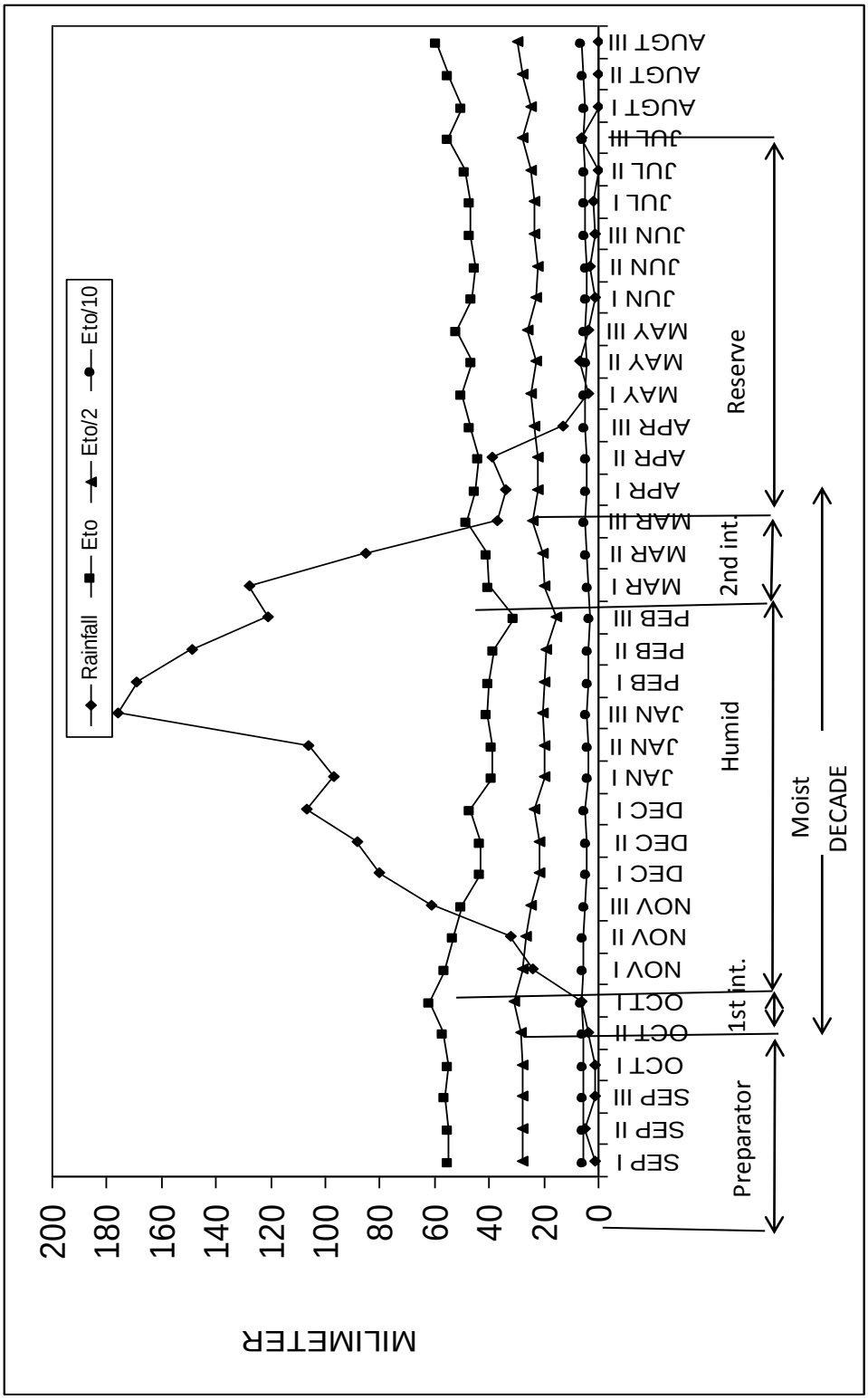

Figure 3. Water balance for Kupang area based on Cocheme-Franquin (1967) Model (1st int. = first intermediate; 2nd int. = second intermediate).

Table 2. Hydrological measurements of BPTP (heavy clay) and BMKG (clay loam) soils.

Soil	Bulk Density	Field Capacity (w,v)	Permanent Wilting Point	Available water	Current water content	Deficit water †	mm of water per 10 cm
	Mg m^{-3}	---------- % w (v)‡ ----------					James, 1988
BPTP (clay)	1.3	35 (43)	18 (23)	17 (21)	22 (28)	13 (16)	18
BMKG (clay loam)	1.3	30 (37)	16 (20)	14 (18)	22 (28)	8 (10)	10

† Deficit water = Field capacity – current water content; current water content = water content at dry condition prior to irrigation or rain;

‡ Numbers outside and inside parenthesis are percent

James (1988) suggested that at this deficit water (8%) this kind of soil (clay loam soil) has to receive irrigation or rain water as musch as 10 mm per 10 cm of soil depth. If the planting depth around six to seven centimetres, the depth of wetting to a field capacity has to be at least 10 cm. The amount of water needed to reach this depth has to be at least 10 mm. Therefore, the onset of planting time for corn coincides with the third decade of November where there is a surplus of water as much as 11 mm (Figure 3).

Similarly for the soil at BPTP field, the field capacity of the soil (clay soil) was 35% and the soil moisture at dry conditions prior to the rain was 22%. This brings about a water deficit of 13% by weight bases. James (1988) suggested 18 mm per 10 cm soil depth of water is needed to bring back the soil moisture to its field capacity before planting. Since the depth of the dibble hole was normally six to seven centimetres, the wetting front at field capacity has to reach at least 10 cm depth in the soil profile. Therefore, the onset of the planting of corn should be when the water surplus is ≥18 mm. Figure 4 dictates that this can happen at the beginning of the first decade of December where the water surplus is as much as 37 mm.

Dry Spell Sub-Model

Despite the more exact tool as explained in the previous section, another factor to consider in the corn planting time determination is the probability of having a decade with two consecutive dry spell days (days with ≤ 3 mm of rain per day). As shown in Table 3, a 21-year daily rainfall records show that starting from the third decade of November to the second decade of December, the probability of having a decade with two consecutive dry spell days (PDD) is < 50%. Oldeman and Frère (1982) suggested < 50% PDD as a safe threshold. This implies that choosing the third decade of November and the first decade of December as the planting date of corn, respectively, for BMKG and BPTP field is a wise decision.

Mulch Experiment

Even though the chosen planting time of corn is a safe decision by considering PDD < 50% can still pose a threat to the survival of the seedling during the establishment stage of corn. Mulch application can be an anticipative appropriate technology to overcome a dry spell menace.

The mulch experiment conducted in the BPPT field revealed that mulching can delay irrigation for four days, save one irrigation (reduce the irrigation frequency from five times for no mulch to four time for mulching), and saved 12 mm of irrigation water (638 mm for no mulching and 626 mm for mulching) for corn plant without reducing corn yields (Table 4). It can then be implied that mulching can overcome the dry spell problem when planting time is followed by a high (≥ 50%) probability of having two consecutive dry spell days.

Table 3. Frequency of having one dry spell in a decade F(D), frequency of having two consecutive dry spells in a decade F(DD), probability of having one dry spell in a decade P(D), and probability of having two consecutive dry spells in a decade P(DD) for Kupang area, East Nusa Tenggara Province. A dry spell is a day with less than 3 mm f rainfall. Data from 21 year daily rainfall records.

Month	Decade	F(D)	F (DD)	P (D)	P (DD)
				---------- % ----------	
November	III	8	2	38	25
December	I	5	2	24	40
	II	6	1	29	17
	III	2	1	10	50
January	I	2	0	10	0
	II	3	0	14	0
	III	2	1	10	50
February	I	1	0	5	0
	II	2	0	10	0
	III	5	2	24	40

Table 4. Comparison of total water, corn yield, water use efficiency, and irrigation frequency under mulching and no mulching treatments.

Items of Observation	No Mulching	Mulching	Remarks
Total water, mm	638	626	12 save
Corn yield, kg ha^{-1}	3412.7	5039.7	1627 save
WUE, kg yield m^{-3} water	0.5	0.8	
WUE, m^3 water kg^{-1} corn yield	1.9	1.2	
Irrigation frequency	5 times	4 times	1 time save

Planting Time Proposed Model

Planting time determination for corn has to start with determining the onset of the rainy season based on SOI and water balance (Figure 4). Second is to have the surplus of water (based on water balance model: (rain water-ETo)>0) higher than the deficit soil moisture (field capacity minus current water) to initiate seed germination. The third step is threshold of the PDD < 50% after the onset of the rainy season. Therefore, the proposed model for corn planting time is as follows: 1) IF (average SOI of three preceding months >+7 and (rain water-ETo)>0) THEN planting time = 1 ELSE planting time = 0; 2) IF (rain water-ETo)>0>soil water deficit THEN Planting Time = 1 ELSE planting time = 0; 3)IF PDD < 50% THEN planting time = 1 ELSE planting time = 0

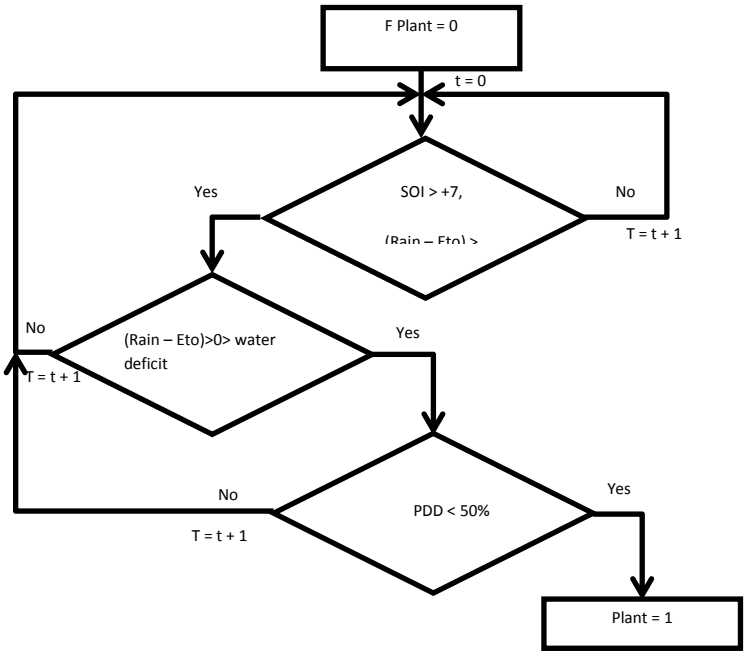

Figure 4. Proposed corn planting time model for dryland of Timor, East Nusa Tenggara Province. T = time; t = decade in the algorithm reiteration.

Conclusions

1. The proposed simulation model consists of three sub models i.e. rainy season forecast based on water balance and SOI, soil moisture requirement for germination, and probability of a decade with two consecutive dry spells.
2. Mulching can be utilized to overcome a planting failure dry spells.

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4. Head and Staffs of BPTP NTT Office, Naibonat Kupang.

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Efficacy of New Formulation of 1-Methylcyclopropene for Improving Postharvest Quality of Pelargonium Flower

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Abstract

Pelargonium is the most flowering plants that have a variation of flower color. It is also popular as bedding plants and hanging basket plant. Reduction of postharvest quality, flower senescence flower and bud drop, are affected by several factors such as ethylene. 1-MCP is a chemical compound that has a function as ethylene inhibitor. Commonly, 1-MCP is a volatile compound. Therefore its application is limited in enclosed area. New formulation of 1-MCP, sprayable formulation, was found, that can be used in opened area and in the field. This study was conducted to find out the optimum concentration of sprayable 1-MCP to improving postharvest quality of pelargonium flower. Pelargonium zonale hybrids 'Katinka' was used in this investigation. This study was conducted in Randomized Block Design with two replications. Six concentrations of sprayable 1-MCP (0.5, 1, 2, 5, 10 and 20 µL L-1), one concentration of 0.2 µL L-1 of volatile 1-MCP and one control (without 1-MCP) were used in this experiment. The result showed that sprayable 1-MCP at 5 µL L-1 was effective in reducing ethylene reducing ethylene-induced petal abscission and extending floret longevity of flower.

Keywords: Ethylene, flower, senescence, 1-MCP

Introduction

Pelargonium zonale hybrid. is an ornamental plant. It is one of the most important species in the bedding plant business in Europe, with approximately 38 million units sold at Flora Holland in 2008 (Flora Holland, 2009). In addition, it can be used not only as bedding, a potted or a hanging basket plant, but also as a medicinal, an aromatherapy, and a mosquito repellent plant, such as *Pelargonium graveolens*.

Giving the high demand of *Pelargonium* in Europe, some strategies are necessary to produce the high quality of *Pelargonium* crops. One strategy is by improving postharvest quality, because quality loses on *Pelargonium* commonly occurs during postharvest, such as during shipping and marketing. The parameter of postharvest quality of ornamental plants depends on postharvest vase life, such as flower and cutting longevity (Seglie *et al.*, 2010). Unfavorable condition during shipment and marketing, such as temperature extremes, long period of darkness, and exposure to ethylene causes a reduced quality of flowers and also cuttings (Rapaka, *et al.,* 2008).

In the case of *Pelargonium*, exogenous (from environment) and endogenous ethylene can be a main problem to its postharvest vase life, since it is one of ethylene sensitive plants. It enhances senescence and abscission of vegetative and generative organs. Ethylene induces cutting senescence showing leaf yellowing due to a loss of chlorophyll, wilting, premature rooting and decay during rooting period (Kadner and Druege, 2004). It also induces flower senescence or abscission due to the death process of flower caused by ethylene. Some reports showed that the continuous exposure 1 $\mu l.L^{-1}$ exogenous ethylene completely

causes petal abscission within two hours from flower of *Pelargonium hortonum, Pelargonium domesticum* and *Pelargonium peltatum* (Jones *et al*., 2001; Evensen, 1991; Cameron and Reid, 2001).

1-Methylcyclopropene (1-MCP) is effective as ethylene blocker by suppressing ethylene action. The advantages of 1-MCP are that it is not toxic and environmentally friendly. Because of that, 1-MCP is very popular, used for minimizing the postharvest loses in horticultural crops, such as ornamental plants. The exposure 1.0 $\mu l.L^{-1}$ of 1-MCP for 4 to 6 h was effective in reducing petal abscission of *Pelargonium x hortonum,* (Kim *et al*., 2007; Jones *et al*., 2001).

The commercial product of 1-MCP is a powder formulation. It releases 1-MCP gas after the addition of a few drops of water or buffer solution. The application of gas 1-MCP needs an enclosed area to prevent releasing or loosing of 1-MCP to another area, otherwise the effect of 1-MCP is not effective in improving postharvest quality and it is quite complicated to apply. Recently a new formulation of 1-MCP, sprayable formulation, was founded. This new formulation makes it quite easier than previously formulation because it does not need an enclosed area to apply it. It can be applied in outdoor like in the fields as pre-harvest treatment. It has already been investigated on some fruits. It is effective in increasing the postharvest quality of apples, pears, and plums caused by ethylene (Elfving, *et al*., 2007; Manganaris, *et al.,* 2008; Mogia and Pereira, 2008; Yuan and Li, 2008). In this report we are reported the effectiveness of this new formulation on postharvest quality of *Pelargonium* cultivars 'Katinka'.

Materials And Methods

Flower Preparation

Developed blooming floret (individual flower) were harvested for the investigated flower. The florets were harvested at half-opened floret stage (Figure 1). After harvesting, florets were immediately transferred to interior environment room (IE) under the conditions: 20 ± 0.5 °C day and night temperature and exposed to 12 h of light from 20 µmol m^{-2} s^{-1} cool white fluorescence tubular for 1-MCP and ethylene treatments.

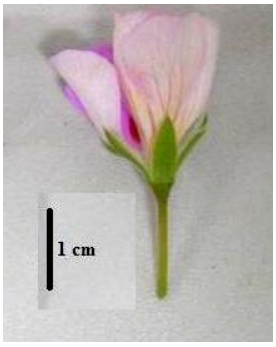

Figure 1. Development stage of floret of 'Katinka' for experiment at the beginning of the experiment, the investigated florets were prepared and placed in a 1.5 ml reaction tube containing water. After that they were placed into 54 L glass chambers for 1-MCP treatments.

1-MCP and Ethylene Application

Six concentrations of sprayable 1-MCP (0.5, 1, 2, 5, 10 and 20 µL L^{-1}) one concentration of 0.2 µL L^{-1} of volatile 1-MCP and one control (without 1-MCP) were used in this experiment. 1-MCP were prepared in two formulations, volatile 1-MCP (SmartFresh) and sprayable 1-MCP (AFxRD-038). For sprayable 1-MCP, the amount of powder formulation was weight, inserted into 1 L spray tank, dissolved in tap water and swirled for 2 – 5 minutes. 1 % Tween-20 was added to the 1-MCP spray solution as a wetting agent. 1-MCP solutions were sprayed to plant materials in enclosed space after 5 minutes mixing. The application of volatile 1-MCP was applied by weight and placed a powder formulation in a scale pan inside the sealed glass chambers. To release 1-MCP gas, a few drops of tap water were added to the powder. After 6 hours incubation, the chambers were ventilated. For controls, there was sprayed with 0.1% Tween-20 diluted in tap water. Six hours after 1-MCP treatment, the 1-MCP treated flowers and also control were transferred and randomly placed into two different 200 L sealed glass chambers. During the experiment periods, one glass chamber was ventilated with air containing ethylene 1 µL L^{-1} and a second glass chamber, as a control, was ventilated with ethylene free-air. A simple diffusion system was used for the method of ethylene treatment.

Determination of Display Quality of Florets

To determine the display quality of florets, the percentage of petal abscission and floret longevity was recorded. The percentage of petal abscission was evaluated by counting the number of petals dropped in response to a gentle shake. The evaluation was done at 2, 4, 15, 24, 48, 72 and 96 hours after 1-MCP treatment. The percentage of petal abscission was calculated using the equation:

$$Petal\ \ abscission\ (\%) = \frac{Number\, of\ \ petal\ \ drop}{Total\ \ petal\ \ per\ \ floret} x100\%$$

The floret longevity was determined as the number of days from the beginning of the experiment until 60% of all petals are dropped.

Results and Discussion

Petal Abscission

The application of 1-MCP delayed 1 day of petal abscission. Treating 'Katinka' florets with volatile 1-MCP 0.2 µL L^{-1} resulted 13.89 % petal abscission at 48 hours of experiment period. It was significantly different to all treatment. The application of sprayabale 1-MCP was also effective in preventing ethylene-induced petal abscission and was significantly different compared to control and volatile 1-MCP 0.2 µL L^{-1}. After 48 hour of exogenous ethylene, the range of petal abscission of sprayable 1-MCP was from 31.67 % (sprayable 1-MCP 10 µL L^{-1}) to 48.33 % (sprayable 1-MCP 0.5 µL L^{-1}). Petal abscission in all 1-MCP treated floret dramatically increased after 72 hour and nearly 85 % petal abscisiion after 96 hours. Sprayabl 1-MCP 5 µL L^{-1} was sufficient and effective in reducing 10 % ethylene induced petal abscission compare to control at 96 hour of exogenous ethylene exposure. It was not significantly different compared to volatile 1-MCP 0.2 µL L^{-1}, sprayable 1-MCP 10 and 20 µL L^{-1} (Figure 2).

Floret Longevity

Floret longevity was measured until ≥ 60% of the petal abscission in response to a gentle shake. The statistic analysis showed that the application of both of 1-MCP formulations, sprayable and volatile 1-MCP, given a significance effect on floret longevity of *Pelargonium zonale* 'Katinka' (p-value > 0.01) (Table 1). The mean comparison among treatment by using Tukey's test showed that volatile 1-MCP treated florets at 0.2 µL L^{-1} resulted the longest of floret longevity of 3.08 days and were not significantly different compared to sprayable 1-MCP treated florets at 5, 10 and 20 µL L^{-1}, whereas control florets resulted the shortest of floret longevity reached 1.53 days, and was significantly different compared to other treatments.

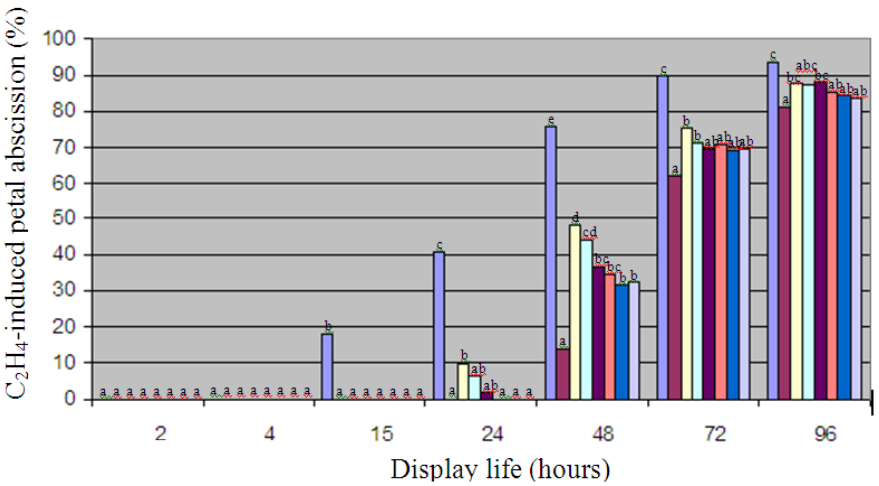

Figure 2. Effect of 1-MCP treatments on C_2H_4-induced petal abscission of *Pelargonium* 'Katinka'continued with continuous 1 µL L^{-1} ethylene throughout the experiment. The mean value followed by the same letters in each cultivar are not statistically different at p=0.05 by Tukey's HSD test (performed separately within each hour).

Table 1. The effect of various concentrations of sprayable 1-MCP (0.5, 1, 2, 5, 10 and 20 µL L-1) and 0.2 µL L-1 volatile 1-MCP on floret longevity (days) of Pelargonium zonale hybrid 'Katinka'. After 1-MCP treatment they were treated with 1 µL L-1 ethylene during 96 hours of experiment period

Treatment	Floret Longevity (days)
Control	1.53 c
Volatile 1-MCP (0.2 µL L-1)	3.08 a
Sprayable 1-MCP (0.5 µL L-1)	2.47 b
Sprayable 1-MCP (1 µL L-1)	2.61 b
Sprayable 1-MCP (2 µL L-1)	2.67 b
Sprayable 1-MCP (5 µL L-1)	2.75 ab
Sprayable 1-MCP (10 µL L-1)	2.81 ab
Sprayable 1-MCP (20 µL L-1)	2.75 ab

The mean value in the same column followed by the same lowercase are not statistically different at p=0.05 by Tukey's HSD test.

The quality of *Pelargonium* florets were highly dependent on display life of florets which is affected by many factors such as ethylene. A new application technique of 1-MCP, sprayable 1-MCP, was found to prevent the deteriorative effect of ethylene. This study reported the efficacy of the new formulation as sprayable 1-MCP, in comparison with the volatile 1-MCP in preventing the ethylene-induced florets longevity. Ethylene can be a main problem in postharvest vase life of *Pelargonium* by reducing its postharvest quality. Mainly ethylene induces flower senescence or abscission in flowers (Jones *et al*., 2001; Evensen, 1991; Cameron and Reid, 2001). This study reported that the application of volatile 1-MCP was more effective than sprayable 1-MCP in improving postharvest quality of *Pelargonium* florets. Probably the different velocity of 1-MCP from volatile 1-MCP and sprayable 1-MCP entering the cell and binding the ethylene receptor which is affected by form or phase of the active substance causes the difference efficacy. Volatile 1-MCP diffused rapidly into the plant tissue, whereas sprayable 1-MCP required a much longer time than volatile 1-MCP to diffuse into the tissue. Sisler *et al* (2009) showed that N-N-Diprophyl-(1cyclopropenylmethyl)amine as a gas was more active than the carbonate salt in preventing the degradation of chlorophyll.

According the data, the effectiveness of 1-MCP was affected by the application methods and concentration. This is similar with Blankenship and Dole (2003), Dole and Wilkins (2005) who reported the effectiveness of 1-MCP was highly dependent on cultivars, concentration, duration of exposure, temperature, stage of development and plant maturity. Sprayable 1-MCP was effective in preventing the deteriorative of ethylene effect and improving the display life of *Pelargonium* florets in comparison to 1-MCP untreated florets. In opened space application, a high concentration of sprayable 1-MCP is required to improve display quality of *Pelargonium* florets. 5 µL L^{-1} of sprayable 1-MCP was sufficient and effective in preventing petal abscission up to 96 hours of exogenous ethylene exposure (Figure 2). These concentrations had similar effect with application of 0.2 µL L^{-1} volatile 1-MCP. Sprayable 1-MCP was also effective in reducing ethylene-induced petal abscission (Figure 2) and extending floret longevity (Table 1). However in the terms of concentration, sprayable 1-MCP is required in the high concentration for the application in an opened space application. Its efficacy depends on the ethylene sensitivity of each cultivars. Jones *et al*. (2001) and Kim *et al.* (2007) showed that 1-MCP did not completely inhibited petal abscission in the most ethylene sensitive cultivar of zonal *Pelargonium* (*Pelargonium* x *hortonum* Bailey) and regal *Pelargonium* (*Pelargonium* x *domesticum* Bailey).

Conclusion

Based on this study, it can be concluded that Sprayable 1-MCP was effective in preventing the deteriorative of ethylene effects in floret. However, volatile 1-MCP gave a better result than sprayable 1-MCP. Sprayable 1-MCP can be used in enclose or open space applications. Thus, it makes easier and faster application in open-space conditions than volatile 1-MCP. Five µL L^{-1} of sprayable 1-MCP was sufficient and was as effective as volatile 1-MCP 0.2 µL L^{-1} in reducing ethylene-induced petal abscission in all cultivars up to 96 hour of exogenous ethylene exposure and extending floret longevity 1 day compared to untreated floret.

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Genetic Diversity of Morphological Responses ond The Relationships Among Javanese Winged Bean (*Psophocarpus tetragonolobus* L. DC.) Accessions

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Abstract

This research was conducted to determine the extent of diversity and relationships among 27 winged bean accessions collected from several region in Java Island. Characterization of 29 traits was conducted in a field experiment started from May 2009 untill september 2010 at experimental station of Faculty of Agriculture, Universitas Padjadjaran, Jatinangor Sumedang. Experiment were arranged in a Randomized Block Design and replicated twice. Treatment was 27 winged bean accessions collected from various region of Java Island. 29 characters observed were morpho agronomic characters which were measured by referring to plant descriptors issued by IBPGR (International Board of Plant Genetic Resources). The experiment was conducted in two growing seasons. Correlation analysis was done on several quantitatively measured characters. To estimate the extent of diversity, data were analyzed multivariately using principle component analysis and cluster analysis. Results showed that there were extremely large diversity among 27 javanese wingedbean accessions. Results of PCA analysis indicated that accessions from west Java were distributed in four quadrant of biplot. This means that wide variability existed among western accessions. Meanwhile, rest accessions from central and eastern Java seemed to be affiliated in the same cluster with accessions from west Java. Fact that three accession from west Java was separated different cluster also indicated that wingedbean seed trafficking might start from west java, central and eventually east Java. This prediction was in accordance with previous assumption that west Java had been center of diversity of winged bean. The results of the characters distribution and phenotipic diversity analysis permitted broad generalization about collection, coservation (core collection), and uses (breeding purposes).

Keywords : accession, diversity, genetic, seed-trafficking, winged bean

Introduction

Winged bean *(Psophocarpus tetragonolobus*) is one of the legumes crop which has great potential in meeting the protein needs of people in tropical region. Nutrient contents of Winged bean seeds reach the range of 29.8 to 39% for protein, 15 - 18% for fat, and 23.9 to 42% for carbohydrate (Tadera *et al*., 1984; Amoo *et al*., 2006), a highly competitive value with of soybean or peanut. As with soybeans, Winged bean can be processed to be tempe and so called winged bean milk. Apart from that, Winged bean young pods can be used as a vitamin A riched vegetable. Winged bean also produces edible fresh tubers which can also be processed into flour, made chips, and so on. Winged bean tubers as the potential source of food is very prospective. Winged bean tuber contained 4 to 6% protein, 9.5 to 19.6% starch, and 68 -75% of dry material (National Academic of Science, 1981).

Based on these potentials, it is reasonable to pay more attention to Winged bean through a more focused development efforts. One form of Winged bean plant development is through the breeding program. In breeding a crop commodity, genetic diversity or variability was a major prerequisite for breeding goals to be achieved (Fehr, 1987). Futhermore, a wide genetic diversity would be determinnant factor for the success of selection process and technically will help increasing the value of genetic gain (Baihaki, 2000).

Center of winged bean's origin remains in dispute and debatable. Winged bean center of origin are suggested to cover areas of Southeast Asia, India and Madagascar (National Academy of Science, 1981). Winged bean is spreaded from East Africa, India to Papua New Guinea, while the center of origin are assumed to be the Indochina-Indonesia and East Africa. Center of greatest diversity is located in Indonesia and Papua New Guinea Islands. Before its potential was understood, Winged had been grown as a horticultural crop in East Africa, parts of India, Sri Lanka, Thailand, Indo-China, Malaysia, Indonesia, the Philippines and some islands in the Pacific. Now, winged bean has been introduced to tropical regions, and some areas subtropika (www.prosea.org).

The use of vernacular names to trace the origin of the *P. tetragonolobus* is at best precarious. Rumphius, who lived on the Island of Amboina from 1653 to 1702, first wrote of *P. tetragonolobus* (Merril, 1917 cit. Hymowitz and Boyd, 1977). Rumphius believed that the cultigen had been brought to the Moluccas from elsewhere, probably from Bali or Java and that the vernacular name given to the plant, botor, was an Arabic word (Burkhill, 1935 cit. Hymowitz and Boyd, 1977). According to dictionary of Arabic, the word botor does not exist in the Arabic language. Heyne, as reported by Burkhill (1935) cit. Hymowitz and Boyd, (1977), suggested that botor is a Sundanese name for the pod. According to the Indonesian language dictionary edited by Poerwadarminta (1976), botor is a plant which fruits like a bean, *P. tetragonolobus* (tumbuhan yang buahnya kacang). If Heyne is correct, perhaps the winged bean was originally from western Java or first brought to western Java from elsewhere. Many of the indigenous, as well as introduced, cultivated, and naturalized species have vernacular names that are specific to a particular geographical region, while others refer to a general class of plants. For example, kacang or kachang are general terms for beans. Specifically, kacang botor refers to *P. tetragonolobus*. However, very often this system of vernacular nomenclature can be misleading. For example, the peanut in Indonesia has been called kacang manila. In this case, reference is given to the place of origin of an early introduction into the country. Similarly, the vernacular names Goa bean, Manila bean, or Mauritius bean for *P. tetragonolobus* probably refer to the place of introduction of the species as it moved about from one country to another (Hymowitz and Boyd, 1976). Attempts to relate the origin of a domesticate to the number of vernacular names in a country or region can be misleading. Very often the number of vernacular names for a domesticate is a function of the number and special interests of botanists in the region.

Ignoring the vernacular names point of view, evaluating the genetic diversity in Java island especially in west java, deserve to be conducted due to its widespread planting. From data of etnobotanic and traditional uses, it is generally assumed that this spesies had been introduced and cultivated from long time ago to this island for various purposes and uses. To proove wether genetic variability really exist, this research to evaluate morphological response of several javanese winged bean accessions and to asses the relationship among them, was conducted.

Materials and Methods

Experiments to assess the genetic diversity and relationship of 27 Javanese winged bean accessions was conducted in two growing seasons started from May 2009 until October 2010 in Experimental station of Universitas PadjadjaranFaculty of Agriculture Jatinangor. Each experiment was arranged in Randomized Block Design (RBD), the treatment were Winged bean accessions consisted of 27 level which was replicated twice. Plot of experimental unit was one row plot (plot of one line) with distance between row 1 m and plant 0.5 m. Each plot consisted of 10 holes and each hole was planted with 3 seeds. Twenty-seventh accessions evaluated were presented in Table 1.

Table 1. Name, origin and region of accessions evaluated

No	Accessions	Origin	Region
1	8.12	Ds Cibitung Kidul, Buah Dua Sumedang	Western Java
2	8.29	Sleman, Yogyakarta	Central Java
3	8.11	Ds. Pangadegan, Ranca Kalong Sumedang	Western Java
4	8.1O	Cileunyi, Bandung	Western Java
5	8.60	Ds, Sukatali, Situraja Sumedang	Western Java
6	8.90	Ranca Batok, Bandung	Western Java
7	set 1	Purwokerto	Central Java
8	8.17	Ds. Jati Selatan, Sidoarjo Jatim	Eastern Java
10	Dj3	Yogyakarta	Central Java
11	Kr1	Karawang	Western Java
12	8.13	Ranca Kalong Sumedang	Western Java
13	8.70	Cileunyi, Bandung	Western Java
14	8.3O	Purwokerto	Central Java
15	8.16	Yogyakarta	Central Java
16	8.50	Ciwidey, Bandung	Western Java
17	8.15	Sambung Pare, Tasikmalaya	Western Java
18	8.30	Subang	Western Java
19	8.40	Ds. Sawangan, Subang	Western Java
20	8.20	Ds. Cembul, Sagala Herang, Subang	Western Java
21	8.14	Karang Pawitan, Garut	Western Java
22	D.T.	Cipeundey, Sukabumi	Western Java
23	8.1	Cianjur	Western Java
24	Dj1	Yogyakarta	Central Java
25	Nth1	Cileunyi, Bandung	Western Java
26	Tto1	Subang	Western Java
27	Nth 2	Cileunyi, Bandung	Western Java

Observed variables were several morphological and agronomic characters. Correlation analysis was done on several quantitative characters. To assess diversiy, data were analyzed using multivariate analysis. Entire data characters were firstly developed into standardized data sets for multivariate analysis with principal component analysis (PCA). Interpretation of the results of PCA was done thorugh biplot of scores of the first and second components to determine the relationship between genotype. Euclidean distance matrix results through

the cluster analysis was used to create a UPGMA cluster dendrogram (Unweighted Pair Group Method Arithmetic).

Results and Discussions

Several quantitatively measured characters showed different degree of variation among 27 Javanese winged bean accessions (Table 2). Most of characters observed were shown to be varied among characters with averaged coefficient of variation 17,1 %. The lowest coefficient of variation were shown in stamen length (2,08%), as well as stylus length (2,98%), sepal length (5,01%) and pod width (8,87%) which were also categorized low (below 10%). The, most varied character based on coefficient of variation was number of pod per plant followed by other yield components characters such as single tuber weight, tuber weight per plant and so on. It is well known that performance of quantitative characters such as yield and yield component are much influenced by environmental change. On the other hand, this high degree of variation also indicated that there were difference of adaptability level of accessions in local environment of Jatinangor.

Table 2. Statistical parameters of quantitatively measured characters

Characters	Mean	Min	Max	Stdev	CV
Leaf length (cm)	7.207	5.400	8.965	0.747	10.36%
Leaf width (cm)	5.225	4.040	6.445	0.577	11.05%
Stem diameter (cm)	0.764	0.555	0.997	0.108	14.18%
Petiolus length (cm)	6.098	4.815	7.345	0.674	11.05%
Number of trifoliate	34.185	22.700	46.750	5.636	16.49%
Sepal length (cm)	1.411	1.250	1.575	0.071	5.01%
sepal width (cm)	1.040	0.817	1.383	0.113	10.84%
Petal length (cm)	3.120	2.023	4.303	0.583	18,68%
Pedunculus length (cm)	1.082	0.767	1.467	0.176	16.30%
stamen length (mm)	3.164	3.058	3.296	0.066	2.08%
stylus length (mm)	3.655	3.450	3.900	0.109	2.98%
Number of pod per plant	15.852	5.500	50.250	10.035	63.31%
Pod length (cm)	21.713	14.865	39.825	4.966	22.87%
Pod width(cm)	1.939	1.535	2.290	0.172	8.87%
Number of seed per pod	11.840	9.100	16.650	1.864	15.74%
100 seed weight (g)	42.932	29.950	60.825	7.545	17.57%
tuber diameter (cm)	1.849	0.888	2.475	0.329	17.79%
Tuber weight per plant (g)	31.118	17.825	54.963	9.973	32.05%
Single tuber weight (g)	16.843	7.125	25.815	4.362	25.90%

Several characters showed significant correlation one another. Based on correlation analysis, it was known that leaf length was positively correlated with stem diameter,

petiolus length, number of trifoliate, pod number per plant, and pod length, It means that the longer leaf length, the longer stem diameter, petiolus length, pod number per plant, and pod length. In contrast, stamen length was negatively correlated with pod length, seed number per pod, and 100 seed weight. This meant that the longer stamen, the shorter pod length, the smaller number of seed per pod, and the smaller seed size. Table 3 showed complete matrix of correlation coefficient among quantitatively measured characters.

Results of principal components analysis showed that the first significant ten principal components could explain 82,6% of the total diversity (Table 4). The first principal component accounted only for 16.0% of the total diversity, the second component 12,5%, and the next major component of the three, four, up to ten, respectively for 12,1%, 10%, 7,8%, 6,1%, 5,4%, 4,5%, 4,2% and 3,8%. This means that such ten principal component could be used, instead of 29 characters, to described variation among 27 winged bean accessions.

Based on eigen vector analysis of data matrix (Table 4), characters with eigen vector above 0,5 were assumed to have correlation with its real averaged value. The correlation between the main component score with the average real character indicated that these characters showed a wide variation. Table 4 showed that characters of leaf length (PD), stem diameter (DB), number of trifoliate (JT), pedunculus length(PTB), pod colour (WPlg), wing colour (WSPlg) were determinant in first principal component. This meant that such variation of such characters determined the variation from which PC1 could explained (16%). Characters of pod length (PPlg), seed number per pod (JBPlg), calyx colour(WK) seemed to be determinant in 12,5% variation explained by second principal component. Characters of petiolus length (PPtl), tuber diameter (DU), and single tuber weight (BUT) seemed to be varied based on third principal component which accounted for 12,1% of total variation. Based on fourth principal component which explained 10% of total variation, characters of stem diameter (DB) and pod width (LPlg) seemed to be determinant. To explain the distribution pattern of accessions based on its relationship, the score of first and second principal component of accessions (data non presented) could be used to construct a biplot. Biplot constructed from the principal component score one and two is presented in Figure 1.

Figure 1 showed that accessions from Java, especially West Java had a fairly wide variation. This was shown from the position of accession points scattered in the four quadrants in the biplot. If related with characters observed, this divergence pattern in biplot was formed based primarily on those characters that were correlated with first and second component scores. Accessions set 1 were far located from other accessions in one quadrant. That accession was especially different from other based on characters of pod length, seed number per plot, and calyx colour. This significant different performance was probably due to such accession was selected from radiated accessions. Most of middle Java accession occupide quadrant I and IV (negative score of PC 1) meaning that they had shorter leaf length, stem diameter, pedunculus length, and small number of trifoliate. Accession from Jogja were seperated each other but still each of them had associated with certain accession from west Java. To further examine the genetic relationships among accessions, accessions were grouped based on the level of genetic relationship. Such Relationship was presented in the form of dendrogram in Figure 2.

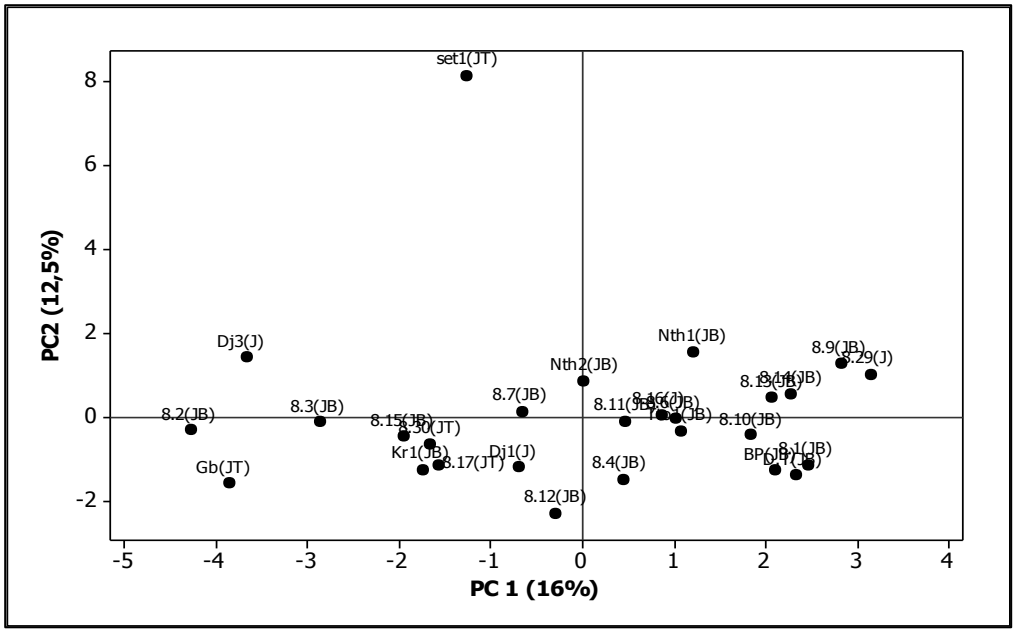

Figure 1. Pattern of divergence between 27 Javanese accession

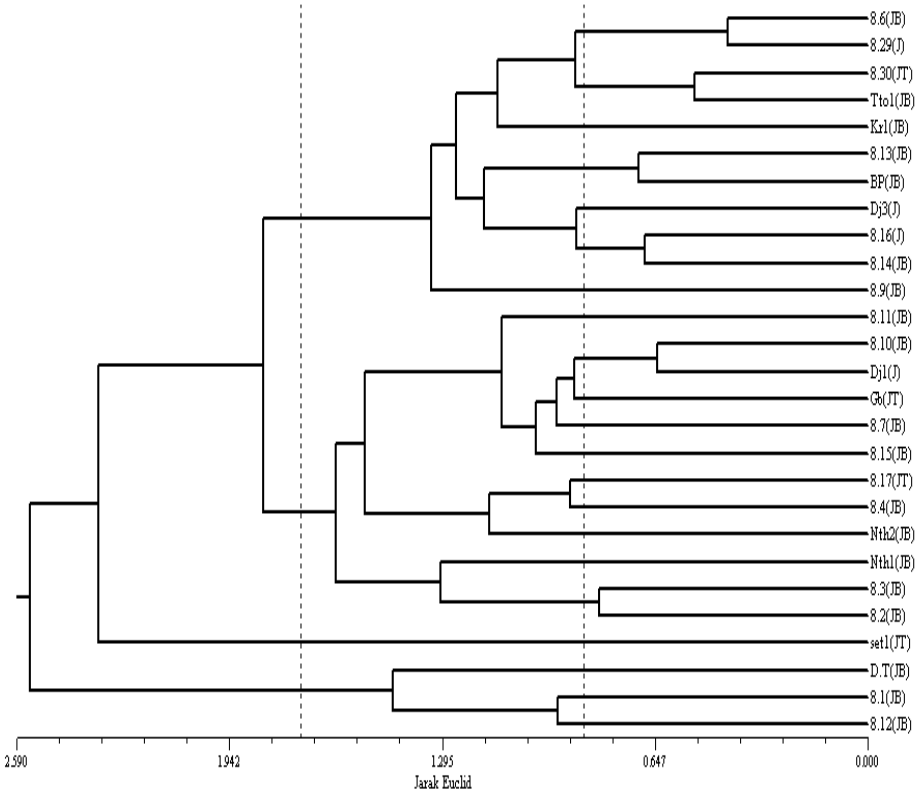

Figure 2. Grouping of 27 Javanese winged bean accession based on 29 morphological and characters

Table 3. Matrix of correlation coefficient among several quantitatively measured characters

Characters	PD	LD	DB	PPtl	JT	PK	LK	PM	PTB	PBS	PPtk	JPT	PPlg	LPlg	JBPlg	B100	DU	BUT
LD	0.149																	
DB	0.57**	0.147																
PPtl	0.595**	0.476*	0.54**															
JT	0.4*	0.209	0.689**	0.392*														
PK	0.04	-0.222	-0.102	-0.03	-0.24													
LK	0.251	-0.285	0.058	0.054	-0.137	0.41*												
PM	0.034	-0.029	-0.09	0.069	-0.23	0.257	0.102											
PTB	0.182	0.235	0.165	0.215	0.171	-0.305	-0.098	0.074										
PBS	0.095	-0.202	0.263	0.2	0.043	0.146	0.025	0.295	0.1									
PPtk	0.31	0.201	0.426*	0.129	0.389*	-0.024	-0.186	-0.023	0.051	0.125								
JPT	0.442*	-0.15	0.407*	0.073	0.44*	0.055	0.08	0.029	0.276	0.409*	0.35							
PPlg	0.403*	0.337	0.012	0.323	0.057	0.045	0.098	-0.128	0.078	-0.426*	-0.098	-0.127						
LPlg	0.118	0.217	0.055	0.112	0.097	0.118	-0.118	0.245	0.057	-0.125	0.205	-0.103	-0.05					
JBPlg	0.217	0.348	-0.182	0.229	-0.133	0.277	0.189	-0.055	0.04	-0.394*	-0.229	-0.111	0.815**	0.046				
B100	-0.12	-0.027	0.073	-0.361	0.125	-0.13	-0.101	0.05	0.197	-0.414*	0.193	0.159	-146	0.263	-0.188			
DU	-0.067	-0.039	0.063	-0.304	0.042	-0.347	-0.114	-0.209	0.273	-0.167	-0.23	-0.03	-0.164	-0.124	-0.158	0.144		
BUT	0.014	0.155	0.273	-0.021	0.19	-0.483*	-0.022	0.021	0.491*	-0.219	-0.071	-0.073	-0.155	-0.047	-0.216	0.164	0.627**	
BUTgl	0.061	0.116	0.282	-0.079	0.216	-0.393*	0.052	0.079	0.353	-0.064	0.013	-0.074	-0.183	-0.077	-0.274	0.067	0.713**	0.859**

* Significant at 5% level of probability

** Significant at 1% level of probability

Table 4. Results of principal component analysis

Characters	Principal Component									
	PC 1	**PC 2**	**PC 3**	**PC 4**	**PC 5**	**PC 6**	**PC 7**	**PC 8**	**PC 9**	**PC 10**
PD	0.538*	0.259	0.494	0.259	0.321	0.012	0.032	0.094	-0.054	-0.045
LD	0.394	0.474	0.119	-0.383	-0.217	0.109	0.075	-0.024	0.333	-0.223
DB	0.558*	0.125	0.218	0.609*	-0.093	0.019	0.285	0.026	0.184	-0.051
PPtl	0.371	0.373	0.56*	0.188	-0.018	0.33	0.16	-0.058	0.235	0.003
JT	0.577*	0.103	0.168	0.42	-0.183	-0.198	0.233	0.169	0.272	0.149
PK	-0.322	-0.346	0.495	-0.063	0.419	0.077	0.138	0.152	-0.112	-0.298
LK	-0.137	-0.113	0.231	0.24	0.736*	-0.099	-0.152	-0.216	0.168	0.024
PM	0.12	-0.314	0.064	-0.189	0.383	0.556*	-0.209	-0.045	0.079	0.017
PTB	0.568*	0.211	-0.207	-0.042	0.067	0.093	-0.329	0.333	-0.129	0.12
PBS	0.022	-0.297	0.203	0.491	-0.22	0.624*	-0.293	0.064	-0.146	-0.124
PPtk	0.588*	-0.223	0.242	0.009	-0.293	-0.035	0.08	0.11	-0.027	-0.162
JPT	0.44	-0.275	0.293	0.444	-0.032	-0.253	-0.405	0.253	-0.208	-0.09
PPlg	-0.009	0.729*	0.459	-0.281	0.207	-0.231	0.017	0.02	-0.073	0.05
LPlg	0.439	-0.33	0.108	-0.536*	0.034	0.163	0.328	-0.156	0.115	0.025
JBPlg	-0.124	0.507*	0.451	-0.422	0.302	-0.183	0.059	0.082	-0.148	-0.046
B100	0.341	-0.365	-0.264	-0.227	0.012	-0.534*	0.119	0.074	0.024	0.174
DU	0.146	0.221	-0.768*	0.198	0.189	-0.094	0.179	0.163	-0.202	-0.202
BUT	0.431	0.276	-0.688*	0.185	0.216	-0.008	-0.035	-0.057	0.204	0
BUTgl	0.423	0.263	-0.7*	0.247	0.275	0.151	0.011	-0.09	0.088	-0.078
WB	-0.228	0.159	-0.029	0.391	-0.123	-0.161	-0.167	-0.779*	0.1	-0.112
WK	-0.328	0.72*	-0.185	0.191	-0.133	0.152	-0.188	0.025	-0.291	0.103
BD	-0.207	-0.228	-0.037	0.489	0.638*	-0.177	0.244	0.143	-0.066	0.085
WBtg	-0.17	-0.264	0.362	0.332	-0.324	-0.211	-0.075	-0.077	0	0.57*
WPlg	-0.791*	0.172	-0.112	0.183	-0.014	0.11	0.002	0.318	0.386	0.063
WSPlg	-0.758*	0.081	-0.024	0.147	0.015	0.054	0.037	0.34	0.51*	0.036
TSPlg	-0.429	-0.002	-0.135	0.14	-0.421	0.111	0.549*	0.127	-0.261	-0.194
BPlg	0.114	-0.173	-0.119	-0.363	-0.161	-0.221	-0.567*	0.21	0.315	-0.174
WBj	0.253	0.11	-0.18	-0.286	0.141	0.443	0.111	0.039	-0.095	0.61
PBj	-0.116	0.85*	0.214	0.088	-0.092	-0.081	-0.174	0.043	-0.095	0.017
Eigen Value	4,64	3,64	3,52	2,89	2,27	1,78	1,57	1,29	1,22	1,12
Percent explained (%)	16,0	12,5	12,1	10,0	7,8	6,1	5,4	4,5	4,2	3,8
Cumulative(%)	16.00	28,5	40,7	50,7	58,5	64,6	70,0	74,5	78,7	82,6

* significant based on 0,5 loading factor.

Based on figure 2, it was known that 27 Javanese accession were grouped into four main cluster. First cluster consisted of 8.12, 8.1, and D.T (west java). Second cluster consisted only of set 1 (Central Java), third cluster consisted of 8.2, 8.3, Nth 1, Nth 2, 8.4, 8.17, 8.15, 8.7, Gb, Dj1, 8.10, and 8.11 (mostly west java). The fourth cluster consisted of 8.9, 8.14, 8.16, Dj3, BP,8.13, Kr1, Tto1, 8.30, 8.29, and 8.6. In accordance with biplot, dendrogram also showed that Set1 was not associated with other accessions. Dendrogram also indicated that each of other accessions apart from west java accession, were always associated with certain west java accessions. Whereas each cluster except cluster II always contained accession from west Java. This facts indicated that accessions from west java showed wide variation and probably indicated that former hypothesis that Java especially west java could be center of diversity, was true (Hymowitz and Boyd, 1977). Fact that three accession from west java was seperated different cluster also indicated that wingedbean seed trafficking might start from west java, central and eventually east Java. This prediction was in accordance with previous assumption that west java had been center of diversity of winged bean. The results of the characters distribution and phenotipic diversity analysis permitted some broad generalization about collection, coservation (core collection), and uses (breeding purposes).

Conclusions

There was a wide diversity of 27 Winged bean accessions with four main clusters spaced far enough. Several morphological and agronomic characters showed high degree of variation among those accession. Three west java accession, 8.12, 8.1, and D.T were grouped in different cluster due to their higher pod number per plant. Set 1accession was occupied alone in different cluster probably due to its longest pod length. The rest accession were grouped in other two different cluster. The wide distribution of west Java accession in three cluster probably indicated that west Java could be center of winged bean diversity and winged bean seed might be trafficked from west Java, central java and ended in east Java.

Acknowledgement

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Influence of Fermentation by Using *Bacillus licheniformis* and *Bacillus megaterium* on Crude Fiber, Fat, Tannin, and Protein Content of Saba Banana (*Musa balbisiana* colla) Peel

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Abstract

An experiment to examine the content of crude fiber, fat, tannin, protein of saba banana (Musa balbisiana Colla) peel fermented by both Bacillus licheniformis and Bacillus megaterium for fish feed raw materials had been done. The purpose of this study was to determine the ability of both bacteria in lowering crude fiber, fat, and tannin as well as in increasing protein of banana peel so it can be used as raw material for fish feed. Fermentation was arranged in completely randomized design (CRD) with 3 replications. The results showed that fermentation by using B. licheniformis with a dose of 5% gave the most high compared with other dose, fermentation could decrease as much as 23.42% of crude fiber, 50% of fat, 82.93% of tannin and could increase the protein content achieve to 47.23%.

Keywords : Bacillus licheniformis, Bacillus megaterium, cellulose, saba banana peel, vegetablefeed substance

Introduction

Feed for fish must contain the required nutrients including protein, fat, carbohydrates, vitamins and minerals. Among all the major of fish feed nutrients, protein is the most important, so that in preparing the formulation of feed required is a minimum requirement of protein to support growth and optimal feed conversion. Feed protein sources from plants are generally cheaper than animal protein. Commercial fish feed typically contains high levels of 25-45% protein, so only plants with high protein levels that can be used as a protein source in fish feed. But there are obstacles in its utilization in feed restriction and limitation of fish to digest plant fibers.

One alternative source of vegetable protein that potential to be utilized in fish feed is the saba banana peel. Saba banana peel is a potential of vegetable protein source of fish feed because it contains a fairly complete nutrition. Based on proximate analysis at the Laboratory of Nutrition and Food Chemistry Ruminant Livestock Animal Husbandry Faculty of UNPAD, saba banana peel contained 17.12% crude fiber, fat at 4.94%, and protein at 9.55%. Saba banana peel also contains tannins of 0.967% which provided a sense of brown in banana, this substance can not be digested by animals so it is needed a Tanase or gallic acid to break down into glucose (Anwar *et al.,* 2007). The main problem in the utilization of banana peel as a vegetable feed substance is content a high crude fiber. Feed with high-fiber ratio can decrease digestibility, efficiency and growth ratio (Ramli *et al.,* 2005). Improving feed nutrition is essential in the terms of the quality of protein so that it can also enhance the growth and survival of fish. Efforts to improve the benefits of saba banana peel can be done in various ways, including through biodegradation by microbes with the fermentation process.

The purpose of this study was to determine the ability of *Bacillus licheniformis* and *Bacillus megaterium* in lowering crude fiber, fat, and tannin as well as in increasing protein in the saba banana peel fermentation. The purpose of this study was to improve the quality of saba banana peel so it can be used as vegetable feed substance for fish.

Materials and Methods

The experiment was conducted in February-May 2011 in the Laboratory of Microbiology Department of Biology Faculty of Mathematics and Natural Sciences, Universitas PadjadjaranJatinangor. A 3 x 3 factorial completely randomized design (CRD) with three replications was applied. Variables measured were crude fiber, fat and protein and tannins. Data were then statistically analyzed using Analysis of Variance with probability level of 0.05 ($\alpha = 0.05$), followed by Duncan's multiple range test (Gomez and Gomez, 1984). SPSS version 13.0 was employed to calculate data.

Results and Discussion

Decrease Crude Fibers, Fat and Tannins

Before fermented, saba banana peel contains crude fiber 17.12%, fat 4.94% and tannin 0.967%, however after treatment and fermentation for 5 days these components decreased as shown in Table 1.

Table 1. Crude fiber, fat and tannins of saba banana peel after fermentation process

Treatment and Dose	Crude Fiber (%)		Fat (%)		Tannin (%)	
	Initial Fermentation	End fermentation	Initial Fermentation	End fermentation	Initial Fermentation	End fermentation
BL 1%	17,12	13,77	4,94	2,75	0,967	0,12
BM 1%	17,12	14,21	4,94	2,60	0,967	0,32
BL&BM 1%	17,12	14,52	4,94	2,51	0,967	0,18
BL 3%	17,12	13,29	4,94	2,63	0,967	0,20
BM 3%	17,12	13,59	4,94	2,42	0,967	0,37
BL&BM 3%	17,12	14,21	4,94	2,41	0,967	0,21
BL 5%	17,12	13,11	4,94	2,47	0,967	0,17
BM 5%	17,12	13,60	4,94	2,36	0,967	0,28
BL&BM 5%	17,12	13,97	4,94	2,45	0,967	0,31

Description: BL = *Bacillus licheniformis*, BM = *Bacillus megaterium*

The data show that each type of bacteria with different doses gives different effects to the decline in crude fiber and the highest result by *B. licheniformis* with a dose of 5% from 17.12% to 13.11%. And *B. megaterium* with a dose of 5% can reduce fat from 4.94% to 2.36%. The highest tannin decreased by *B. licheniformis* with a dose of 1%, from 0.967% to 0.12%. *B. licheniformis* with a dose of 5% to give the most high, fermentation can decrease as much as 23.42% crude fiber, fat by 50%, as much as 82.93% tannin.

Decrease in crude fiber may occur due to *B. licheniformis* dan *B. megaterium* can produce cellulase which can hydrolyze cellulose (Kim and Kim, 1993). It is also supported by the opinion Supriyati *et al.,* (1998), who states that the process of fermentation caused by the breakdown of certain enzymes on the substances that can not be digested, such as cellulose and hemicellulose into simple sugars.

Fat loss occurs due to *B. licheniformis* can produce lipase (Pikoli *et al.,* 2000), as well as *B. megaterium* (Sekhon *et al.,* 2005). Fat can be simplified into fatty acids after fermented by microbes that aid is more easily digested by fish (Afrianto and Liviawaty, 2005) and lipase is an enzyme that can hydrolyze fats into free fatty acids (Macrae, 1983).

According to Mondal and Pati (2000), *B. licheniformis* is a bacterium which can produce Tanase, therefore decrease the highest tannin by this bacteria. Tannins can not be digested by animals so Tanase needed to break down into glucose or gallic acid (Anwar *et al*., 2007). The compounds are broken down in simple carbohydrates when degraded by enzymes have the ability to bind tannins, thus decreasing the amount of tannins bound by the other simple compounds (Aw and Swanson 1985).

Increase Protein

Before fermented, early protein of saba banana peel was 9.55% and after treated and fermented, is expressed increased the protein of saba banana peel. Data elevated protein in saba banana peel under the influence of fermentation by bacteria type and dose of inoculum can be seen in Table 3.

Table 3. Increase of Protein of Saba Banana Peel after Fermentation process

Treatment and dose	Protein (%)	
	Initial Fermentation	End fermentation
BL 1%	9,55	12,62
BM 1%	9,55	12,87
BL&BM 1%	9,55	11,78
BL 3%	9,55	12,80
BM 3%	9,55	13,10
BL&BM 3%	9,55	12,07
BL 5%	9,55	14,07
BM 5%	9,55	13,71
BL&BM 5%	9,55	13,21

Based on the results obtained, each type of bacteria with different doses gives different effects on increasing protein. *B. licheniformis* with a dose of 5% could increase the protein more higher, from 9.55% to 14.07%. Fermentation by *B. licheniformis* with a dose of 5% to give the most high, could increase the protein content of 47.23%.

The opinion Nurhayani, *et al.,* (2000) that an increasing number of mass of microbes will increase protein content on fermentation products. Therefore, increased protein content of fermented saba banana peel is a reflection of the number of bacterial cell mass. In addition, microbial fermentation process is also synthesize an enzyme which is a protein that can

degrade the compound - complex into simpler compounds. This is called a process of enrichment of protein material (Puspitasari and Sidik, 2009).

Conclusion

From the results of this research could be concluded that the fermentation by *Bacillus licheniformis* and *Bacillus megaterium* singly and mixed can to decrease of crude fiber, fat, and tannin as well as increase protein of saba banana peel. *Bacillus licheniformis* with a dose of bacterial inoculum of 5% is the most effective dose and to improve the nutritional quality of saba banana peel by lowering levels of 23.42% crude fiber, fat by 50%, tannins by 82.42%, and increasing the protein content by 47, 33%.

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The Effect of *Cymbopogon nardus* Linne Rendle on Rice Storage Pest *Sitophilus oryzae* Linn (Coleoptera: Curculionidae)

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Abstract

Sitophilus oryzae Linn was known as a store of rice pest that can be reduce rice's quality. The aim of this research was to examine Cymbopogon nardus Linne Randle oil as botanical pesticide to S. oryzae. The method used was Complete Randomized Design (CRD) with five times replication for each treatment. Concentration of Cymbopogon nardus oil treatment used were: 0%, 25%, 50%, 75%, and 100% which observed on 24 hours. The results showed that Cymbopogon nardus oil caused mortality of S. oryzae at concentration of 20-95%, and Cymbopogon nardus oil 75% was the most effective to control S. oryzae.

Keywords: ***botanical pesticide, concentration, cymbopogon nardus, rice, sitophilus oryzae Linn***

Introduction

Rice is a staple food for Indonesian people. One of the problem for stored rice is infestation by *Sitophilus oryzae*. According to WHO 80% of rice in around 38 countries rice had been attacked by *S. oryzae* (Hanny, 2002).

One of potential botanical pesticide is *Cymbopogon nardus* Linne Randle especially its leaves and stem. *C. nardus* contain dipenten, terpen, metil heptenon, D- sitronelal, L-borneol. It also contains volatile compounds such as linalool, alkohol, nerol, geraniol, geraniol asetal, sitronelal asetat dan sitronelil n-butirat, metil eugenol, ses quisitronelene, seskiuterpen, farnesol (Guenther, 1990). Based on above explanation, was conducted test about the effect of Cymbopogon nardus to mortality of Sitophillus oryzae. Those compounds have insectisided effects.

Materials and Methods

The method used was dipping method, with 5 concentrations 0 %, 25%, 50%, 75%, dan 100% with 5 replications.

Figure 1. *S. oryzae*

The second experiment which conducted was choice test with olfcactometer. The Khairomone were coconut oil, soybean soil and corn oil as source of khiromone.

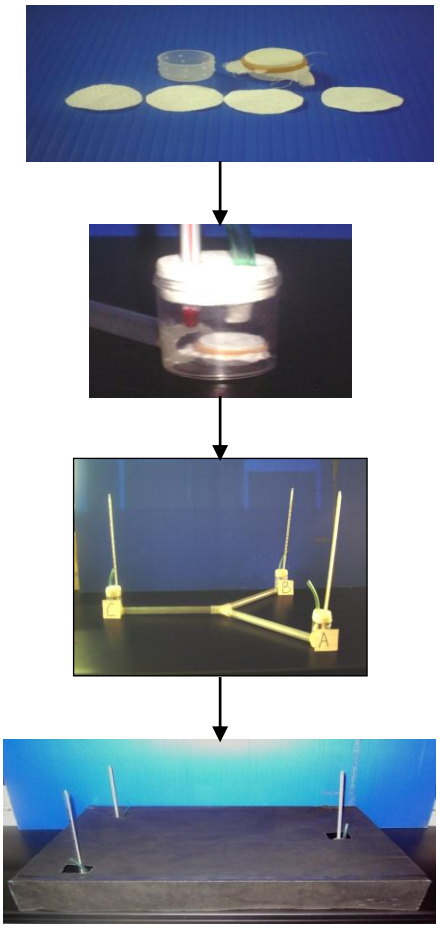

Figure 2. Olfactometer

Results and Discussions

The Concentrations of *Cymbopogon nardus* to the mortality of *S. oryzae*

The result can be shown on Table 1. *Cymbopogon nardus* can cause death of *S. oryzae* with dessication by losing the fluidity in its body by cuticle and respiration system as fumigant as strong fragrant of bioactive. According to Budiarto (2000) the insecticide can inhibit on respiration system by the inhibition of transport electron and oxidative phosporilization.

The insect attractiveness to khairomone be cause of its detected by sensory organ on its antennae. The khairomone which is metabolite plan secunder product can be act as an attractant. Corn oil compose of 45% oleac acid, 40% linoleat acid, 7% palmitat acid, 3%

stearat acid. Soybean contain of 30% oleac acid, 55% linoleat acid, 9% palmitat acid, 4% stearat acid (Patton and Saunders, 1963).

Table 1. Observations of Cymbopogon nardus Effect to the Mortality of S. oryzae

Concentration (%)	Mortality 24 Jam					Average
	1	2	3	4	5	
0 (Ethanol)	0	0	0	0	1	0,2
25	3	1	3	3	1	2,2
50	1	12	17	7	10	9,4
75	19	13	15	17	16	16
100	18	20	20	20	18	19,2

Choice Test of *S. oryzae* to Oil

Table 2. Choice Test of S. oryzae to Oil

No.	Oil	Amount		
		Start Position (C)	Oil (A)	(B)
1.	Corn	43 %	35 %	22 %
2.	Coconut	39 %	30 %	11 %
3.	Soybean	37 %	34%	29 %

Conclusion

Cymbopogon nardus Linne Randle could influence mortality of S. oryzae with 75% and *S. oryzae* was attracted to corn and soybean oil.

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Feasibility Test of The Biopore Absorption Hole to Improve Infiltration Rate

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Abstract

The objective of the research of the Biopore Absorption Hole (BAH) was to know the feasibility of the hole, which was used difference of microbes as an activator in the process of decomposition of organic material, in an effort to improve water infiltration rate into the soil. The research was conducted on experimental field of Industrial Technology Faculty of Agriculture, Padjadjaran University, Jatinangor, at ± 753 m altitude above sea level. Research carried out for 45 days starting in October-November 2010. The methodology of the research uses experimental design of randomized block design, which consists of 5 treatments with 6 replicates. The results of this study indicated that arrangement different organisms in each treatment into "Lubang Resapan Biopori (LRB)" gives a significant value to the rate of infiltration in the BAH. The hole biopori with the addition of organisms earthworm treatment gives the best results with the infiltration rate of 0.038 liters / sec but that the recommendation of the organism an effective and feasible in making biopori, was dried microbes (bio-active Semanggi) because according to the statistical test does not give different results significant with earthworms. Constant infiltration rate (fc) on the surface that was equal to 0.3 cm/min shows that only a slight change soil physical properties on the soil surface.

Keywords : hole absorption of biopore, infiltration, organism

Introduction

According Soemarto (1995) infiltration is a transfer of water from the upper surface of the soil into the ground. Power infiltration is the maximum possible rate of infiltration, which is determined by the surface conditions including soil layers. Magnitude of the infiltration is expressed in cm/hour or mm/day.

Infiltration is the pervasiveness of water into the soil through the soil surface, and a subsurface flow (Soewarno, 2000). Water infiltration into the soil may reduce the proportion of water flowing on the soil surface (runoff), so the water can be stored as a reserve of ground water and underground water (Murtilaksono, Siregar, Darmosarkono, 2007). In addition, infiltration plays an important role in controlling runoff and erosion (Murtilaksono.dkk, 2007)

Infiltration capacity is the ability of the soil in to speeded water (infiltrate) contained in surface water or surface water flow into the interior of the land, with the permeation of the surface water runoff will be very influential. Clearly, the greater the flow capacity of the flow of water infiltration on the surface of the soil decreases (slightly). Conversely, the smaller the infiltration capacity due to the number of soil pores are clogged, the flow of surface water increases or increases (Kartasapoetra, 1989).

Lee (1990) says that the infiltration capacity is a dynamic nature, the capacity is greatest when the rainfall started, and decreased progressively when kolid-colloidal soil expands and reduces pore size, fine materials leached from the surface into the pores pores and inhibit the movement of water, soil close to saturation, and reduced hydraulic gradient.

Surface run-off is part of the rainfall that flows above ground into the rivers, lakes, and oceans. Precipitation that falls to the ground level there is a direct entry into the soil or water is called infiltration. Others did not get into the soil and therefore flows above ground level to a lower place. There is also a part of the rainwater that had entered into the soil, especially on land that nearly or already saturated, the water out onto the ground again and then flows into the lower

One way that can be taken to prevent the flow of runoff water which is then wasted by increasing infiltration is through the creation of Biopore Absorption Hole. Meaning of the definition and understanding biopore absorption hole is the hole diameter of 10 to 30 cm with a depth of 30 to 100 cm filled with organic material (organisasi.org 2010). This organic material further into the feed material (energy source) for various soil fauna to do activities, so that their activity will increase, by increasing their activity the more biopori formed (Biopori.com, 2010). So, biopori are holes that form pores in the soil due to activities of organisms in it, such as worms, plant roots and other soil fauna (Biopori.com, 2010).

Other organisms such as microbes and bacteria in the soil can participate in making a hole in the soil pores, but there is no reference to the time required microbes and bacteria to make holes in different types of soil pores. Each soil type has a different structure, then the speed of the activity of soil organisms were different in each type of soil.

With the application of biopore absorption hole is expected to increase in pore holes in the ground as a new space to store water so as to increase the infiltration rate and obtained the organism with the fastest level of activity for each pore of making holes in the ground. To determine the rate of decline in the infiltration of water into the ground, we need to hold the pace of research on the relationship of infiltration and recharge biopori hole.

Soil organic matter is one of the material forming the soil aggregates, which have a role as an adhesive material between the soil particles together into aggregates to the soil, so that the organic material is important in the formation of soil structure. Influence of organic matter on soil structure is related to the texture of the treated soil (Stevenson, 1982).

The number and types of microorganisms to determine the success of the process of decomposition, or composting. The process of decomposition of organic material in nature is not done by a monoculture of microorganisms but by consortia of microorganisms. Several types of microorganisms commonly found in garbage piles are listed in Table 1.

Biopori are holes in the ground formed by a variety of activity, the organisms in it, such as worms, plant roots, termites and other soil fauna. The holes are formed will be filled with air, and it will be a place of passage of water in the soil (www.biopori.com).

An increasing number of biopori can be done by making a vertical hole into the ground. The holes are then filled with organic materials, such as organic waste, household garbage, grass clippings or other vegetation, and the like. This organic material will eventually be used as an energy source for organisms in the soil so that their activity will increase. With the increase in their activity, the more biopori formed (www.biopori.com).

Table 1. Types of microorganisms

	Bacteria	Fungus
Mesosfil	Pseudomonas spp. Achromobacter spp. Bacillus spp. Flavobacterium spp. Clostridium spp. Streptomyces spp.	Alternaria spp. Cladosporium spp. Aspergillus spp. Mucor spp. Humicola spp. Penicillium spp.
Termofil	Bacillus spp. Streptomyces spp. Thermoactinomyces spp. Thermus spp. Thermonospora spp. Microplyspora spp	Aspergillus dpp. Mucor pusillus Chaetomiumthermophile Humicola lanuginosa Absidia ramosa Sprotricbum thermofphile Torula thermophile (yeast) Thermoascus aureanticus

Source: balittanah.litbang.deptan.go.id, 2010

The sinergism between the vertical hole is made with biopori formed will allow these holes use as artificial water holes infiltration is relatively inexpensive and environmentally friendly Hole absorption is then given a nickname Biopore Absorption Hole.

But in reality on the ground soil organisms are not all able to grow optimally in all soil types and are able to make the soil porosity. With the above statement what is necessary to find organisms that can make holes in the soil pores as well as how long it takes to get the maximum results biopori application. With the principles set out above may be conducted feasibility testing whether biopori the organism can actually make porosity of soil and how long it takes.

The objective of the research of the Biopori Absorption Hole is to know the feasibility of the hole, which is use difference of microbes as an activator in the process of decomposition of organic material, in an effort to improve water infiltration rate into the soil.

Materials and Methods

The experiment was conducted in field trials of Faculty of Agro-Industrial Technology, Universitas Padjadjaran, Jatinangor. Research sites located at an altitude of ± 753 m above sea level. Implementation of the study began in October-November 2010.

The instrument used for this study are:

1. Soil hand-auger, to make the biopore absorption hole in the soil.
2. Measure the container, to measure how much water is given.
3. A plastic sheet to protect the biopore absorption hole due to the rain.
4. Nails, hammer and wood as a fence of the field trials.
5. Penetrometer and Stop watch
6. A set of Double Ring Infiltrometer and Munssel Color Chart.
7. A gallons of water plastic and a standing bench plastic for place the gallons.

Materials used for this study are: (1) Organic Materials. (2) Earthworm.(3) EM-4 (microorganisms mixed with liquid chemicals). (4) Microbes powder (example: Semanggi Bio-active).

Experimental design used was Randomized Block Design, which consists of 5 treatments with 5 replications. The five treatments were:

A = the hole without any treatment.
B = the hole filled organic material.
C = the hole filled organic material plus Earthworms.
D = the hole filled organic material plus the EM-4.
E = the hole filled organic material plus the Semanggi Bio-Active.

Each treatment consisted of six repetitions, so that there are 30 combinations of treatments.

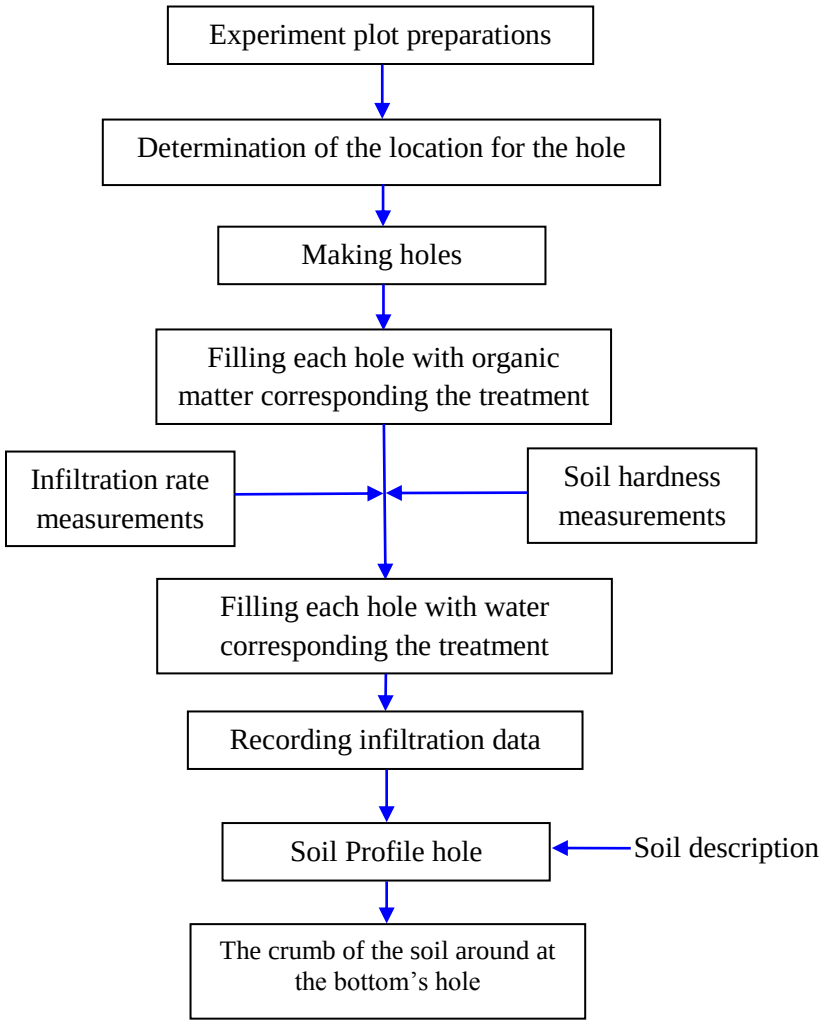

Figure 1. The schematic diagram of research implementation

Results and Discussion

Undisturbed soil sampling to determine the physical condition of the soil carried out before the experiment by using a ring sample at a depth of 0-20 cm and 20-40 cm and then examined in the laboratory to determine the physical condition of the soil in field trials. The results of laboratory analysis showed that the soil has a clay content of dusty, with a total pore space of about 50% only, with the same permeability rate of 0.00127 cm/min. This shows that the physical condition of the soil is lacking in the ability to absorb water into the ground or have very low infiltration capacity.

In the implementation of the field trail 30 holes made using hand-auger, the number of the holes were comes from 5 treatments with 6 replications different. For each treatment begins with the collection and processing of organic material (leaves and grass) which is then inserted into the biopore absorption hole.

Each hole is filled in accordance with the treatment of organic materials, ie for the holes B, C, D, and E are each comprised of 6 holes. The fill's hole leave a height of 10 cm from soil surface. The holes B after the organic material is inserted into the hole, without any additional ingredients. The process of decomposition of organic material occurs naturally. The holes C after incorporated by organic material then added 400 grams of earthworms (Lumbricus) each hole. The holes D, before the organic material inserted into the hole, first mixed with the EM-4 as one treatment. A total of 20 ml of EM-4 mixed with 2 liters of water mixed with organic material in each hole. The holes E, before input into the hole first mixed with clover as much as 5 grams of microbial dry powder can be mixed with organic materials and water for all holes E.

After left for 10 days, the measurements were taken in the first time, a measuring instrument that gallon plastic as water supply is connected with a hose into the hole. Decrease in water level in the gallon of water infiltration into the hole. Measurement of water drop on each hole is done every week for each treatment trial holes.

The results from the first measurement can be seen that the hole E has the fastest time in as many as 12 liters of water absorb into the ground with a time of 438 seconds, while the holes A and D have the longest time to absorb as much as 12 liters of water is 1222 seconds and 1202 seconds. It can be expected while the organic material is mixed with organisms that have not been mixed properly, so that the decomposition time is still very short, the process of decomposition of organic material has not occurred to the fullest.

Besides mixing the organic material in the hole D has the effect of soil organisms that can cause death or undeveloped. Chemicals contained in the EM-4 could be one cause. With no growth of organisms in the hole D, so that will inhibit the manufacture of small holes in the ground because the area of the experiment no plant roots that help making the holes in the ground, so progress and movement of organisms at the first measurement to be the only way for the occurrence holes in the ground (biopore).

While on the hole B, C, and E development already occurring soil organisms, resulting in a time difference of only about 1 minute between holes. Over the past 10 days has not been decomposed organic material all by soil microbes, and the development and activity of soil organisms have not been too much so just some produce small holes in the ground.

Table 2. The results of the first measurement for each treatment

Hole	Volume (l)	Infiltration Time (hour.minute.seconds)
A	12	00.20.22
B	12	00.09.57
C	12	00.08.55
D	12	00.20.02
E	12	00.07.18

And when converted into graphic form, then the result will be as below:

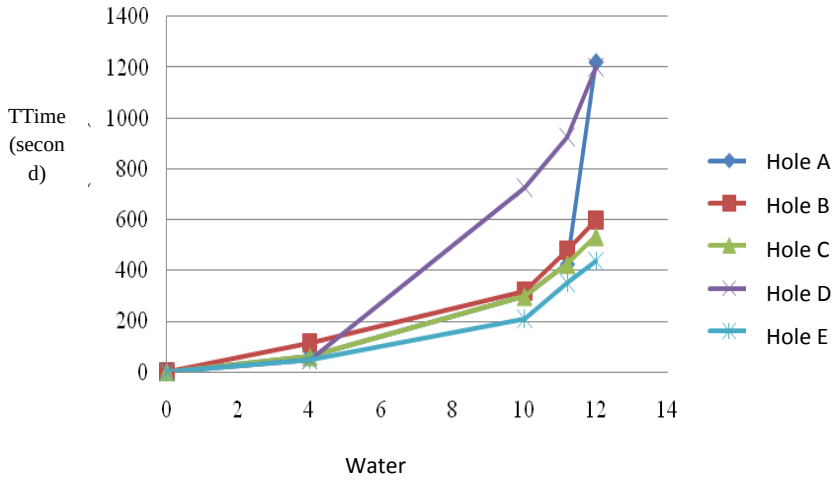

Figure 2. Graph rate of water infiltration in the biopore absorption hole

At the hole A measurements, which the hole without any treatment performed by the addition of water level measurements in the hole. A measurement at the hole is done by measuring the height of standing water in the hole. This measurement is done using an earring attached to a yardstick.

After the first measurement is completed then the measurements were taken to two performed 5 days later. And after measurement to measurement 2 to 3, 4, 5, and 6 performed in an interval of 7 days per measurement. The volume of water that entered each time the measurement is 12 liters, the results obtained are as follows in Table 3.

Table 3. Results total measurement time infiltration in biopore absorption holes (seconds)

Treatment	Replication						Total	Average
	1	2	3	4	5	6		
A	1222	1845	1725	1625	1514	1703	9634	1605,67
B	597	305	1202	638	150	337	3229	538,17
C	535	224	259	331	272	265	1886	314,34
D	1202	638	1330	745	271	331	4517	752,83
E	438	643	210	315	238	211	2055	342,50
Total	3994	3655	4726	3654	2445	2847	21321	

A measurement at the hole A, the results obtained from measurements of 6 times the average was 1605.67 seconds, or approximately 26 minutes 46 seconds. In the hole A the infiltration is not running smoothly. This is due biopori making a hole in the ground in a

completely done by nature. So there is no organic materials that provoke the organism to move and evolve. However, for a hole with no treatment, median time 12 liters of water to absorb into the soil that is 1605.67 seconds or 0.0075 liters / sec is something very good. The average time of infiltration in the hole A was 26 minutes 46 seconds.

Measurements on other holes that were given the addition of micro-organisms and worms showed activity that occurs biopori in the soil so that water can be absorbed quickly.

- Measurement of the hole B will produce an average infiltration time of 538.17 seconds. But hole B have continued to rise until it reaches its peak at the time of fifth measurement of 150 seconds to absorb 12 liters of water into the ground. Infiltration time average of hole B is 8 minutes 58 seconds.
- Measurement of the hole C, a hole filled with organic ingredients combined with earthworms. The infiltration time in the hole C having the fastest average of 314.34 seconds or 0.038 liters/sec. The average time of infiltration obtained hole C is 5 minutes 14 seconds to absorb 12 liters of water into the ground.
- Measurement of the hole D, pit filled organic matter with EM-4. The field fact found, even the hole D is included into the hole to get treatment but the results give very slow infiltration compare to three other holes. Apparently chemical content in the EM-4 can be a barrier to active organisms in the soil. The average time of infiltration produced by the hole D is 752.83 seconds or 12 minutes 33 seconds.
- Measurement of hole E, the holes are filled with organic materials and bio-active Semanggi. The fact the powder is able to work actively and make a quick organic material decomposes in the soil and olso preferably to oragnisme. Proven results obtained by the hole E can compensate for holes C which filled by earthworms. The average time of infiltration resulting hole E is 342.5 seconds, or about 5 minutes 43 seconds.

Statistical test followed by Duncan's range test at 5% level, resulting in the results table as follows in Table 4.

Table 4. Effect of composition of the treatment of infiltration rate increasein absorption biopore hole (ABH)

Treatments	Infiltration Average in ABH (seconds)	Result test
A	1605,67	a
B	538,17	c
C	314,34	d
D	752,83	b
E	342,50	d

And can be seen from the table above that the treatments C and E have the same letter, it shows that the treatments C and E not give a non significant result. Thus the organism at treatments C and E have the same speed in making biopori in the ground. The results of test can point out the organism that works in an efficient and effective to increase infiltration.

There is a fear that the creation of hole absorption biopori will crumble the soil physical structure changes in the form of a process of soil aggregate breakdown, when the these changes occur at the soil surface will cause the soil susceptible to water erosion. To answer these concerns do scientific effort that is the measurement of infiltration, after absorption

hole biopori is made and in it decomposes organic materials by using gauge double ring infiltrometer.

Table 5. Data Measurement Results Infiltrometer (doule ring) per-minute

t (minutes)	The rate of decline in water level (cm/min)	t (minutes)	The rate of decline in water level (cm/min)
1	1,5	9	0,3
2	1	10	0,3
3	0,7	11	0,3
4	0,5	12	0,3
5	0,4	13	0,3
6	0,35	14	0,3
7	0,3	15	0,3
8	0,3	16	0,3

Measurement data in the table above shows that the infiltration around the hole absorption biopori occurred at the soil surface as the infiltration rate curve of undisturbed soil. The results of the measurement followed by a linear regression analysis, it turns out the calculations show that at the soil surface up to a certain depth, soil physical condition has not changed.

Conclusions

- In compare with observations supporting, then a slight decrease in time in treatment A, which is 6 minutes longer than the supporting observations. Time that occurs in treatment B, C, D, and E increased with reference siginficant enough time has been measured at supporting observations. The best enhancement produced by treatment of C was the hole by the addition of earthworms organism which was 12 minutes 48 seconds. This could be caused by the worm have the physical size of the biggest and most capable of rapidly decomposing organic waste, so there are many activities that lead to the formation of holes in the soil pore
- When compared with the observations support, in the undisturbed soil sampling, then there was little difference in the level of permeability of the soil that has the value 0.00127 cm/min. Because the permeability of the soil conditions greatly affect the soil infiltration rate, only the value of the rate of infiltration and permeability rates are still at a fairly small value, so it could be concluded that changes in soil physical properties that occur in the leach pit biopori little impact on soil physical properties on the soil surface.

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Relationship between Slope and Soil Physical Properties A Case Study at Pasirwangi, Garut, Indonesia

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Abstract

The aim of the research was to investigate the effect of slope steepness and slope position on porosity and hydraulic conductivity of soil.The research was conducted from July to September 2007 at Pasirwangi, Garut, Indonesia. It was carried out using physiographic free survey method, which survey was based on land physiographic appearance. Soil sampling was carried out into transect on the same slope characteristics without calculating the range between points of observation.Soil samples were carried out onto three classes of slope as follow: 8 – 15 %; 15 –25 %; and 25 – 40 %. Each class was divided into three slope positions, i.e upper slope, middle slope and lower slope. Four soil sampleswere taken from each slope steepness and position, resulting 36 points of observation.The laboratory data then analyzed by statistical test.The result of the research showed that the higher the slope class, the lower the porosity, while permeability is getting higher. The lower the position of slope, the higher the permeability and porosity.Steep slope (25 – 40%) and upper slope possess the highest porosity, while the highest permeabilityexists on the slopes steepness of 8 – 15%. The lower slope possess the highest permeability and porosity.

Keywords : permeability, porosity, slope

Introduction

A watershed isan area that receives, collects and transports rain through the river to the sea or lake which is physically limited by the form of topography such as ridges and mountains. Watershed consists of several component such as : vegetation, soil, topography, water or river, and human. Fulfillment of human needs results the increasingofnature'sdamage and finally lead to the instability of watershed. Erosion is an indicator to determine whether the watershed has been disrupted or not. A major cause of erosion in Indonesia is the tropical climate and topography condition which is characterized by a high average rainfall intensity and hilly topography. Indication of soil degradation can be identified by the decline of physical and chemical properties of soil showed by a change in the texture, loss of nutrients and organic matter, lower infiltration capacity, lower water holding capacity, lower structural stabilityand increasing density of the soil.

The detachment of soil occur when rain hits the ground. Most of the rain water will flow on the soil surface andit's destructive power will create greater detachment along the steep and long slopes. The steeper the slope, the quicker the flow of the water.It results higher detachment and transportability of water on soil and creates a faster run off. In the end stage off runoff, water is collected on the bottom slope. It makes the deposition of dissolved materials is higher in the bottom slope position, hence bottom slope usually have a better soil quality. (Kinnel, 1983).

Soil properties that affect the ability of the soil erosion is determined by the infiltration capacity, permeability, water holding capacity, properties that determine the resistance of

soil dispersion by water (Judge *et al*, 1986) and resistance from transportability by runoff (Wischmeier and Mannering, 1969). Those properties consist of bulk density, porosity and water content.Infiltration capacity will change according to season, soil management and the most importantis soil permeability (Quansah, 1983).

Pasirwangi wasconsidered as critical zone in West Java. Erosion should be decreased in that area by doing some efforts of conservation techniques. In order to overcome the erosion which is occurs in Pasirwangi, a research should be conducted to investigate the factors that affect erosion. The sensitivity of the soil against erosion influenced by several physical properties of soil. Besides that Pasirwangi is well known as center for agricultural activity which needs to be protected to produce land sustainability.

Materials and Methods

The study area islocated at Pasirwangi village, district of Pasirwangi, regency of Garut, West Java, Indonesia(figure 1). It is located about 80 km from the capital city of West Java province (Bandung) within the coordinates7°10’00”- 7°20’00” South and 107°40’00”-107°50’00” East

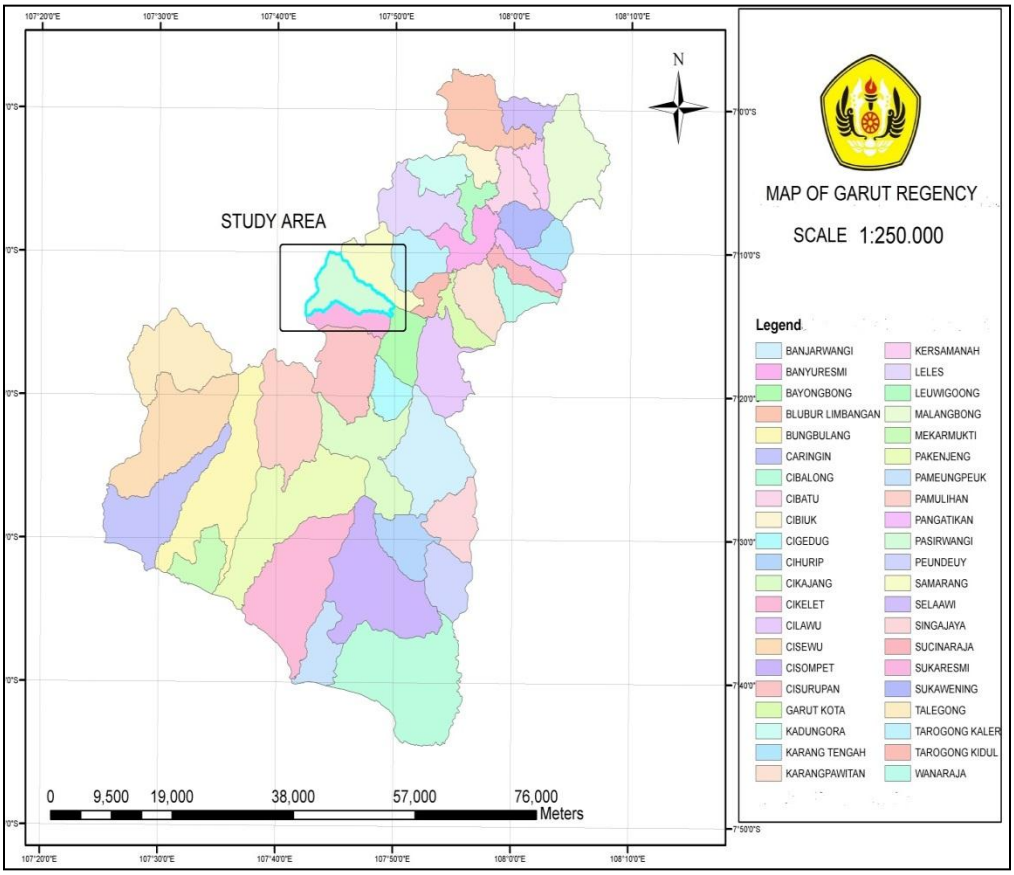

Figure 1. Map of the study area

The research used independentphysiographicsurvey, based on physiographical appearance. Soil sampleare collected randomly without taking into account the distance between

observation points. The observation consists of slope and soil observations on each land unit. It was done based one different land units, i.e.class of slopeand the same land units, climate, practice of conservation and soil type. The survey was conducted to obtain slope classdata (%). Observations performed on the various classes of slope and slope position.

The data and samples from field are collected, then analyzed in laboratorium to determine soil properties such as porosity and permeability. The laboratory data then analyzed by SPSS to determine the effect of factorson the parameters.Minipit was used for determining soil structure. Undisturbed soil sampling (using core sampler) are collected to determine permeability.

Results and Discussion

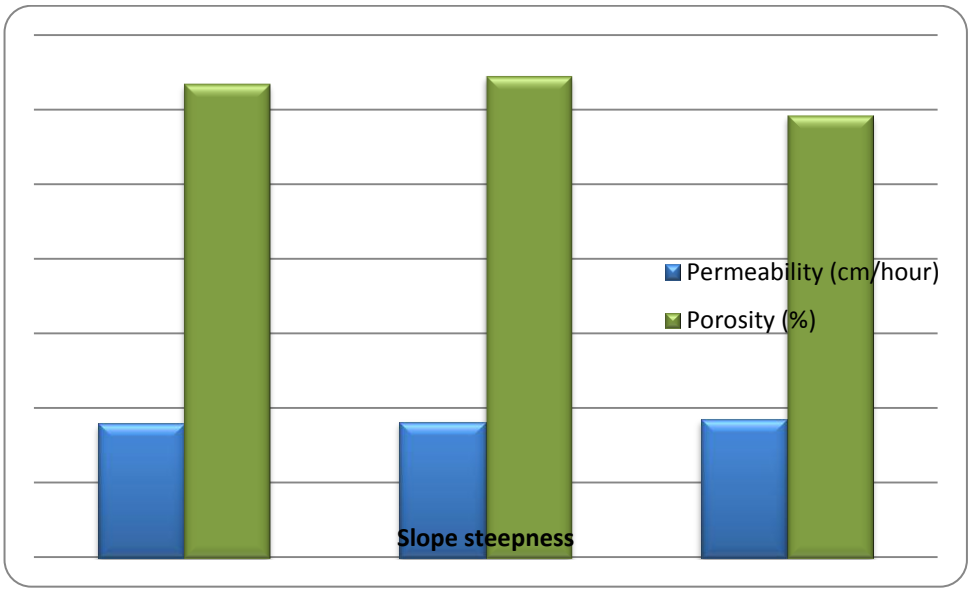

Figure 2.The relationshipbetween slope steepness with soil porosity and permeability

The Relationship between Slope Steepness and Soil Permeability

Faster soil permeability on the steeper slopes is indicated in figure of relation between slope steepness with soil porosity and permeability (figure 2). The soil permeability on the flat slope reached17.96 cm/h, onthe slightly slope 18.15 cm/h and the steep slopes 18.50 cm/hr.Slope steepness gave no significant effect on the soil permeability (significancylevel : 0.997). Faster rate of permeabilitypresumably caused by alteration on soil texture (coarser texture). Higher sand content, lower clay and silt content on the steeper slope lead to the decreasing of permeability. Those three causal factor lead to higher infiltration because soil contain more pores.

The Relationship between Slope Steepness and Soil Porosity

Statistical test showed that there was no significant effect to the soil porosity by slope steepness treatment (significancylevel : 0.75). Figure 2 shows that the steeper the slope the lower the porosity of the soil. This is presumably due to the clogging of pores by particles

caused by aggregates destruction in the process of erosion.Uehara in Saribun(1997) stated that the porosity on the steeper slopes was decreased. This is presumably because the organic material content on that site was low. Organic material affects the condition of the soil. Organic material will initially create a comfort condition for microorganism to live which eventually will create a better soil porosity by their activity.

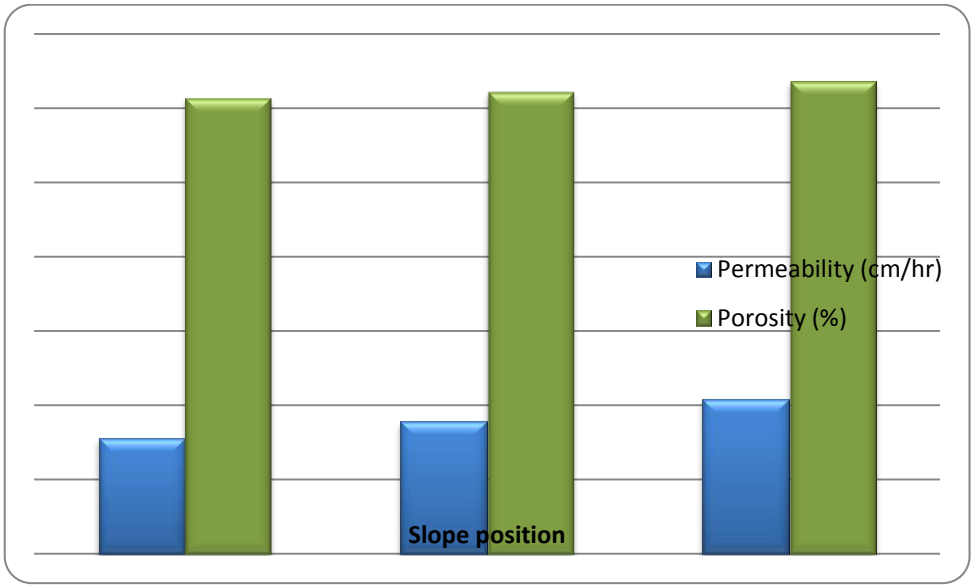

Figure 3.The relationshipbetween slope position with soil porosity and permeability

The Relationship between Slope Position and Soil Permeability

The highest permeability was found atlower slope position (20.780 cm/h) compared with other slope position. Permeability rate at the upper slope was 13.125 cm/h and 12. 339 cm/h on middle slope (Figure 3). Those address us to one conclusion that lower the slope position, higher the permeability. The main reason for that condition was a higher porosity in the lower slope. High pore content will create space for water to go through. On the upper slope, the permeability was not occur with higher rate. It was because the total porosity was lower in that area. Based on statistical analysis, treatment gave no significant effect to the soil permeability (significancylevel : 0.174).

The Relationship between Slope Position and Soil Porosity

Based on statistical analysis, slope position gave no significant effect on the soil porosity (significancylevel : 0.619). From figure 2, we can observe the porosity condition on each slope position. The lower the slope, the greater the porosity of the soil. Upper slope possess 61.331%, 62.146% and 63.591% in the middle and lower slopes. This is presumably due to a rough texture in the lower position. It is known that the sand and dust fraction is transported from the upper slope slopes to the lower slope and accumulated on the lower slope. In the process of erosion, silt fraction will be first detached and transported, thensand fraction will follow. While the clay fraction is more difficult to be detached, but when it is already detached from its aggregate , it will be transported very far (Wischmeir and Smith, 1978). Besides that, the higher permeability in the lower slope might be caused

by the process of run off that occur with high velocity, means there were more sand fraction delivered to the lower position. Another reason was the organic material which collected in the lower position, hence the soil pore enhanced. Due to the enhancement of soil pore, the soil porosity increased.

Conclusions

1. The steeper the slopes, the lower porosity and the higher soil permeability. The higher the position of slope, the higher porosity and the lower the permeability.
2. Steep slope (25-40%) and upper slope possess the highest porosity, while the highest permeabilityexistson the slopes steepness of 8-15%. The lower slope possesses the highest permeability and porosity.

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The Potential of *Trichoderma* Isolated from Cocoa to Control Black Pod Diseases on Cocoa Pod

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Abstract

Black pod disease cause by Phytophthora palmivora is one of the important diseases limiting cacao yields in Aceh. Research on biological control of black pod disease has been conducted on cacao pods in Aceh. Research on molecular systematics of Trichoderma isolates from Aceh was done at Systematic Mycology and Microbiology Lab, USDA-ARS Beltsville, MD, under the WCF-Aceh Cacao Fellowship program. Molecular characterization of Trichoderma isolates from Aceh identified three species of Trichoderma which were : T. virens, T. asperellum, and T. longibrachiatum. The potential effect of T. virens in controlling black pod disease compared to T. harzianum have been evaluated in cocoa pods. The results of this study indicated that T. virens affected the incubation period and the diameter of lesion caused by P. palmivora in cocoa pods. Althought T. harzianum was able to colonize the fruit tissue but T. virens more effective in controlling pathogens than T. harzianum.

Keywords: black pod diseases, cocoa, Phythopthora palmivora,Trichoderma

Introduction

Commodities that are being developed on a large scale in the Aceh region are the Cocoa plant. The development is conducted from the nursery to post-harvest, however, this widespread development still faces obstacles, including the existence of pests and diseases.

All parts of cacao plants can be infected by pathogens, ranging from roots, stems, flowers, fruit and leaves. The losses are very high when the diseases was infected the cocoa pod. Cocoa black pod disease caused by *Phytophthora palmivora* was the most important disease of cocoa cultivation. One of controls that can be developed is the use of antagonistic fungi to suppress fungal inoculums in the soil.

One of the biological control agents that has been discovered and widely used is fungal antagonist Trichoderma (Chet *et al*., 1979; Elad *et al*., 1982). The fungi can also increasing plant growth. Responses to application of *Trichoderma* spp. are characterized by increased germination percentage, plant height and dry weight and a shorter germination time in vegetables (Baker *et al*., 1984; Chang *et al*. 1986, Paulitz *et al*., 1986) and earlier flowering and an increase of number of blooms in Periwinkles *(Vinca minor* L.) and Petunia *(Petunia hybrid* Vilm) (Chang *et al*., 1986).

Endophytic fungi are the non-pathogenic fungi associated with plant tissues without causing disease in plants. The fungi have great opportunity as biocontrol agents to control plant pathogens. Therefore it was necessary to conduct a series of study from isolating and identifying fungi associated endofit Trichoderma in plants and tested their ability to control black pod disease on cocoa pods.

Materials and Methods

Isolation and identification endofit fungi

For all surveys of endophyte infection in the present study, healthy leaves, root and steam were collected, washed in running tap water and processed within 4 h of collection (Arnold *et al* 2000). From each leaf, root and stem were cut into 2 cm than were surface sterilized by sequential washes in 90% ethanol (5 min) and 2.5% clorox (2 min), rinsed with sterile water, and allowed to surface-dry under sterile conditions. This method of surface-sterilization eliminates epiphyllous microorganisms from endophyte cultures (Arnold *et al* 2000).

Endofitik fungi isolates from Cocoa ware purified on PDA (Potato Dextrose Agar). DNA was amplified by PCR using internal primers Ef-700F (5'- TCT ACC AGT GCG GTG GTA) 18 b - Reverse primer: ef2 (GGA (G/A)GT ACC AGT (G/C)AT CAT GTT) 21 b. DNA sequences that have been found was identified in the GenBank database. Level, genus level identification databases based on the statement above 90%. Molecular identification process carried out in the Systematic Mycology and Microbiology Lab, USDA-ARS Beltsville, MD, under the WCF-Aceh Cacao Fellowship program .

Preparing *Trichoderma* Spore and *Phythopthora* isolate

Spore suspension

Trichoderma spp that have been grown on rice media for two weeks, was taken about 1 g and then put into measuring cup and mixed with 1 ml of distilled water. The mixture was stirred for 15 minutes until evenly distributed,mixed with 1 ml suspension into 9 ml distilled water until the next 10^{-4} dilution. Taken 1 ml of dilution and above board haemacytometer drops, then we observe under the microscope and then repeated 3 times to obtain representative results.

P. palmivora was isolated colected from cocoa plants in the cacao plant research center Jember, then cultured and propaga ted in PDA. *P. palmivora* then put on to PDA using a needle loop for cultivation than incubated in the incubator for 2 weeks.

Preparation of cocoa Pod

Cocoa pods used in the study were cocoa lindak, ± 3 months old and that were taken directly from the cocoa plantations in Geumpang, Aceh Pidie, the healthy cocoa pods were sterilized using 70% alcohol and then air dried.

Aplication of Trichoderma spore

Trichoderma spp conidia suspensions were prepared, and then sprayed on any surface of cocoa pods were 35 x 10^6 spores in 100 ml distilled water under treatment and then left for 1 day.

Inoculation Phytophthora Palmivora

Inoculation were done by injuring certain part of the epidermis layer of cocoa pods by using the needle on each fruit, and then the pods were inoculated by plug of *P. palmivora* culture (1cm x 1cm), then incubated for 24 hours.

Measurement: 1. Incubation period of the first appearance of the symtomp; 2. Diameter spot: Observation of the diameter of the spot on each fruit was observed on day 6 and 7 after application of *Trichoderma* spp, using a ruller; 3. Reisolation of *T. virens* and *T. harzianum* from inoculated Cacao pods: The growth of fungal mycelium antagonists was checked by cutting the inoculated part and sterilized by using alcohol 2.5 % chloroks and then air dried. The samples were placed into petridish containing PDA and incubated for one weeks.

Results and Discussion

Trichoderma isolated from Aceh cocoa plantation were identified under molecular work, Three species of *Trichoderma* were shown on Figure 1.

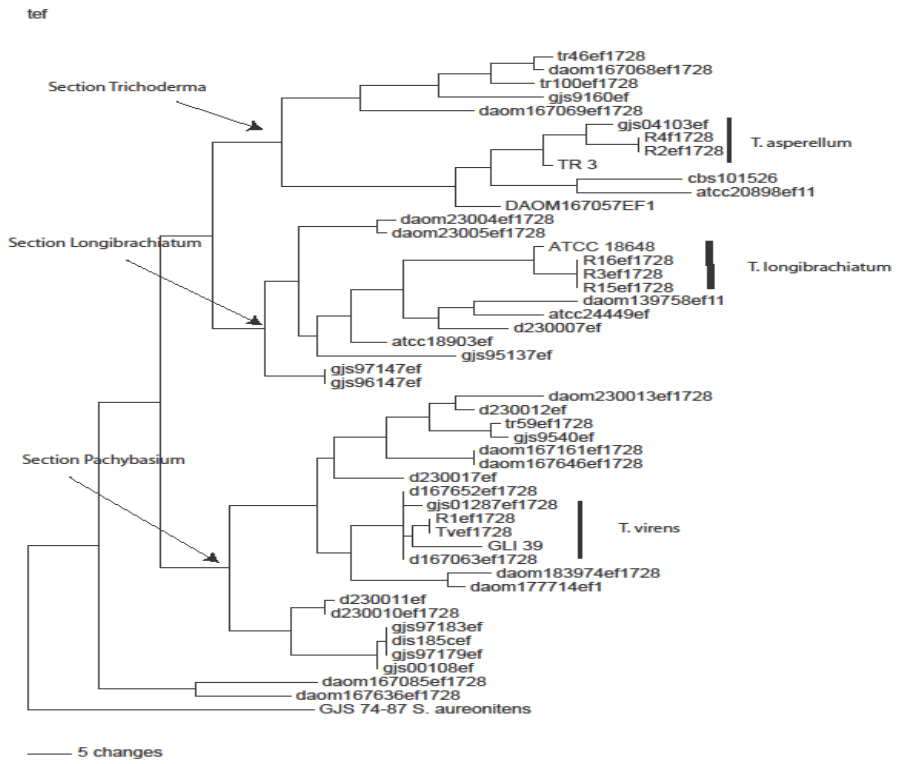

Figure 1. Phylogenetic tree of representative isolates of *Trichoderma* belonging to Sections *Trichoderma*, inferred by parsimony analysis Primer Ef-700F and ef2. DNA sequences that have been found later than the species in the GenBank database. Level, genus level identification databases based on the statement above 90%.

Incubation Periode

The incubation period of *P.* palmivora in various treatment can be seen in Table 1. *T. virens,* and *T. harzianum* affect the incubation period *Phytopthora palmivora*.

Table 1. The incubation period of *P. palmivora* in various treatment

Treatment	Repetition																		Average
	1			2			3			4			5			6			
P1	4	4	4	3	3	3	4	4	4	3	3	3	3	3	3	4	4	4	3,5
P2	-	-	-	-	-	5	6	-	-	-	-	-	4	-	4	4	4	-	4,5
P3	6	4	4	-	5	5	3	3	-	4	4	4	3	3	3	3	3	3	3,75
P4	5	5	5	4	4	4	6	4	4	3	3	3	3	3	3	4	4	4	3,94

Description: – = non-occurrence of the incubation period

Table 1 showed that aplication of antagonistic agents T. virens (P2) showed longest incubation period of the fungus P. Palmivora (4.5 days). It indicates that *T. virens* was more effective in inhibiting the development of pathogens.

Diameter of Cocoa Spot at 6 and 7 Day after Inoculation

Observations diameter of cocoa spot at 6 and 7 day after inoculation were shown on the Figure 2. *Trichoderma* as agent antagonist have been shown significantly affect of the the diameter of spotting a pod at 6 and 7 HSI. The average diameter spots on as effect of an antagonist agent can be seen in Figure 2.

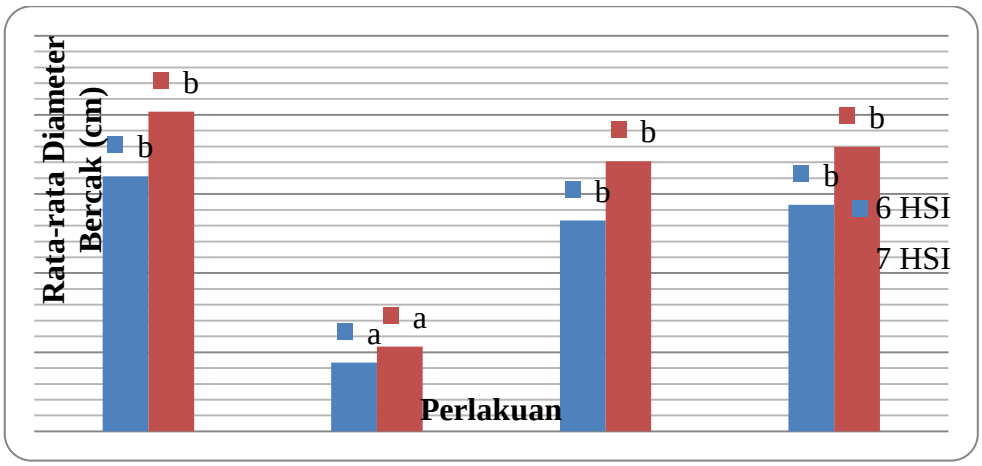

Figure 2. The average diameter of the spots with the different treatment of *Trichoderma* spp species (P1 = control, P2 = *T.virens*, *T.harzianum P3* =, *P4* = *T.virens* + *T. harzianum);* numbers followed by the same letter indicate not significantly different based on LSD at the 0.05 level. Data Transformation √ x + 0.5)

Reisolation of *T.virens* and *T harzianum* from Inoculate Pod

The results of observation after reisolate of fruit tissue which had been grown on PDA medium showed that at day 8 was found fungus *T. harzianum* and *P. palmivora*, whereas *T. virens* many secretes enzymes and toxins that produced, this proves that the *T. virens and T. harzianum* were able to get into the fruit tissue and effective in controlling pathogens.

Related to this, Blakeman & Fokkeman (1982) stated that *T. virens* will inhibit pathogens in the infection process, because at the beginning and its development were needed nutrients and enough space to infect plants. While on combination treatment (P4) shows an incubation period of the fungus *P. palmivora* (3.94 days) were not significantly different with the treatment of *T. harzianum* (P3) with an average incubation period of 3.75 days. *T. harzianum* (P3) show the average incubation period were not much different from the treatment of the pathogen *P. palmivora* (P1). This perhaps because *T. harzianum* was not able to develop properly on the fruit. The original habitat of *T. Harzianum* were in the soil so its growth progress was slower compared to *T. virens*. In the combination treatment (P4), there might was be competition between *T.virens with T. harzianum* in the struggle for nutrients and ability of each species of *Trichoderma spp* in controlling different fungal pathogens, this was because the morphology and physiology were different. For example, *Trichoderma harzianum* and *Trichodrma hamatum* produce the enzyme b-1 ,3-glucanase and chitinase that can cause exolisis host hyphae (Chet, 1987). Mahr (2005) states that *T.virens* produce toxins, antibiotics and various enzymes such as endo, ekso and glikonase, selobiase and chitinase.

Figure 2 shows that the smalest spots on the inoculated cocoa pod was found in the treatment of *T. virens* (P2). This may because the spores *T. virens* growing faster than the growth of spores of *P. Palmivora*. According to Mahr (2005) , T.virens produce toxins, antibiotics and various enzymes such as endo ekso and glikonase, selobiase and chitinase. The research was supported by Muttaqin (2011) which stated that the effectiveness of local isolates of the fungus T.virens was proven potential in controlling Phythopthora palmivora to be able to get it on the plants and great potential as a biocontrol. The other reason was due to *T. virens* belongs to class of free-living fungi benefit to the plant and also the role of auxin in increasing the growth and development of the germination response Arabdiopsis (Bais *et al*., 2006). While the diameter of the longest found in the control treatment (P1), it because of *P. palmivora* more freely infect host (cacao fruit) due to the absence of antagonist agents that suppress pathogens. In order to *T. harzianum* (P3) and *T. virens + harzianum* (P4), the diameter spots did not differ significantly with treatment without giving the agent an antagonist. Each *Trichoderma* have a different mechanism of attack patogent , *T.virens* works by producing a toxin, while *T. harzianum* to produce mycoparasitic, maybe in this study the mechanism of toxin is effective to control *Phytopthora*, while on combination treatment (P4).

Conclusion

Aplication of *T.virens* (P2) was more effective in suppressing the development *P.palmivora* compared to other treatments. Aplication *T. virens* (P2) on cocoa pod contribute to the shorter incubation period, lowest of diameter cocoa spot. This result can be concluded that the treatment with *T. virens* was very effective in controlling black pod disease.

Acknowledgements

A part of this work was supported by Swiss contact under PEKA Project through WCF-Aceh Cocoa Fellowship. We thank to Garry Samuel and Ed Ismael at laboratory Systematic Mycology and Microbiology Lab, USDA-ARS Beltsville, MD, for fungi identification work and team work of Phytopathology Laboratory of Agrotechnology department, Faculty of Agriculture Syiah Kuala University.

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The Effect of Phosphates Solubilizing Bacteria to The Growth and Crop Production of Corn Plant (*Zea mays* L.)

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Abstract

The aim of this research was to know the effect of phosphates solubilizing bacteria to the growth and crop production of corn plant (Zea mays L.). This research was arranged in Completely Randomize Design (CRD). The phosphates solubilizing bacteria (PSB) consist of Bacillus megaterium and Pseudomonas sp. The treatment in this experiment was given the PSB dosage 0, 10, 20, and 30 mL/plant and control positive (NPK fertilizer 5 g/plant) and replicated five times for each treatment, and each replication consisted of five plants. The PSB was inoculated on planting hole of corn plant. The corn plant growth was observed every 2 weeks, and plants were harvested at 15 weeks. The parameters that observed were plant height, root length, plant-root biomass, and plant production was dry seeds weight. Data were analyzed by one-way ANOVA, with Brown-Forsythe test at level 5%. The results of this research showed that the treatment of the PSB dosage and NPK fertilizers gave no significant difference (α>0.05) for growth and crop production of corn plant. The best growth of corn plant was given by NPK treatment but no significantly different with PSB treatment. The best dry seed weight (173.19 ± 43.60 g/plant or 17.31 ± 4.36 ton/ha) gave by PSB dosage 20 mL/plant, but the best RAE (Relative Argonomic Effectiveness) gave by PSB dosage 10 mL/plant.

Keywords: ***corn plant (Zea mays L.), , growth, phosphates solubilizing bacteria, production.***

Introduction

Potential agricultural land in Indonesia ranges from about 41 million ha, but 20 million ha of land of which is critical. Critical soil is poor soil nutrient (nutrients) that are not or less productive for crops. The most important nutrients that plants need are nitrogen (N), phosphorus (P) and potassium (K). P element is an element that is needed by corn plants (Rubatzky and Yamaguchi, 1998). Corn crop (Zea mays L.) is one type of food crop seeds, corn became the second most important commodity crops after rice. Based on the order of the world's staple food, corn ranks third after wheat and rice (Anonymous, 2000). Corn is an important and popular food, the popularity of corn increased rapidly in Europe and Asian countries which is characterized by increasing corn production (Rubatzky and Yamaguchi, 1998). The growth of corn crop is one of that many plants which require nutrients, especially phosphate elements.

Khan *et al*., (2009) suggested that the elements contained in the soil phosphate have an important role as well as a macro element required by plants. The role of phosphate, among others, is for growth of cells in plant tissues, the formation of fine roots and hair roots, strengthens stem cell walls, so that plant roots do not easily fall down, improve crop quality, the formation of flowers, fruits and seeds as well as strengthen the resistance of plants to disease or attack pests. When a ground element of phosphate is deficient, the

plant will experience a variety of metabolism obstacles, for example, in the process of protein synthesis. Elements of phosphate deficiency in plants can be observed visually, the emergence of a purplish or reddish color on older leaves is caused by the formation of pigment antosianin. These pigments are formed because they accumulated glucose in the leaves as a result of inhibition of protein synthesis. Other symptoms in plants due to lack of phosphate element is necrosis (tissue death) on the blade and petiole which is followed by the weakening of stems and roots of plants (Salisbury and Ross, 1995).

Phosphate content of elements in soil varies widely, but generally low. Elements of organic phosphate are about 30-50% of total phosphate in soil and humus. As a result, lower phosphate content of the element than nitrogen (N), potassium (K), and calcium (Ca), one way to meet the needs of phosphate in the soil element is the provision of chemical fertilizer phosphate (Elfiati, 2005). Provision of phosphate fertilizer plants were only able to be utilized by 10% to 20% (Hasanuddin and Gonggo, 2004). Thus, effort to address the low phosphate provided by adding fertilizer is inefficient, even too expensive. In addition, the use of phosphate fertilizers such as TSP is continuously in the future will damage the original texture of the soil loose and fertile become hard and barren (Elfiati,2005).

To improve production yield and quality of corn, then an alternative is needed to address the low elements are available phosphate in soil that is by making use of free-living soil microbes and has the ability to solubilize phosphate in the soil. One of the microbes that have the ability to solubilize phosphate in the soil is bacteria. In the presence of the bacteria, the ability of plants to absorb elements of phosphate in the soil to its needs will increase (Regis, 2001). In addition, the use of phosphate solubilizers as a bacterial biofertilizer have the advantage, that is cost effective, does not pollute the environment and be able to help improve the solubility of phosphate is trapped in acid soils. The use of phosphate solubilizing bacteria can produce growth regulating substances and resist penetration of root pathogens by producing antibiotic compounds (Hasanuddin and Gonggo 2004).

Khan *et al*., (2009) reported that among the soil bacterial genera Pseudomonas, Bacillus, and Enterobacter are the most powerful P solubilizers. Bacillus megaterium, B. circulans, B. substilis, B. polymyxa, B. sircalmous, Pseudomonas striata could be referred as the most important strains to solubilize the phosphate rocks. Soil microbes such as bacteria Pseudomonas sp. and Bacillus sp. are a type of phosphate solubilizing bacteria that can produce organic acids such as citric acid, glutamate, succinate, lactate, oxalate, glioksalat, malic, fumaric, tartaric, and α-ketobutarat, with increased organic acids are usually followed by a decrease in pH, so resulted in the dissolution of phosphate is bound by Ca (Elfiati,2005).

Soil P is precipitated as orthophosphate and adsorbed by Fe and Al oxides is likely to become bio-available by bacteria trough their organic acid production and acid phosphatase secretion. Although, high buffering capacity of soil reduces the effectivenes of PSB in releasing P from bound phosphates, however, enhancing microbial activity through P solubilizing inoculants may contribute considerably in plant uptake. Phosphate solubilizing bacteria are very effective for increasing the plant available P in soil as well as the growth and yield of crops (Khan *et al*., 2009). Based on above the background, it is necessary to do research on phosphates solubilizing bacteria on the growth and production of corn (*Zea mays* L.).

Material and Methods

The experiment was conducted at the Agriculture field in Kecamatan Dlanggu, Mojokerto, East Java. The isolate of phosphates solubilizing bacteria (PSB) consist of *Bacillus megaterium* and *Pseudomonas* sp. from the collection of the microbiology laboratory, Department of Biology Faculty of Science and Technology, Airlangga University, Seed Corn (BISI-2). The design of this study using Complete Randomized Design, treatment in this study were PSB bacteria dose: 0, 10, 20, and 30 mL/plant, and the positive control consisted of chemical fertilizer NPK 5 g/plant, with 5 replications for each treatment, and each replication consisted of 5 plants.

Liquid Fertilizer of Phosphates Solubilizing Bacteria (PSB)

Preparation of starter bacteria was done by pure cultures of each *Bacillus megaterium* and *Pseudomonas* sp. as Phosphates Solubilizing Bacteria **(**PSB) which were grown in NA medium+1% glucose and in NB medium+1% glucose and then incubated at room temperature for 2 days. PSB made by taking 10 ml of pure cultures of each the *Bacillus megaterium* and *Pseudomonas* sp into 280 ml NB+1% glucose medium aseptically, after incubated at room temperature for 2 days. At the end of incubation the number of bacterial cells counted (TPC 10^8 sel/ml media), and liquid fertilizer PSB applied in the field.

Implementation of the Experiment

Soil preparation was done by fertilizing and cleaning soil from weeds and the reminants of roots, the experiment land area was 15 x 6 m^2, the plot was made with size 2,5 x 0,8 m^2, each plot separated by 2 m wide beds, 100 g/plant given manure as a fertilizer base. Plant's holes were separated with a distance of 80 x 20 cm^2, each hole was filled with 2 seeds, furthermore, PSB bacteria in liquid medium was poured on a hole of plants with doses of treatment, it was also performed a positive control treatment of chemical fertilizer NPK 5 g/plant. Addition of PSB and chemical fertilizer NPK was repeated at age 25 days and 45 days. Replanting seeds, weeding and watering were also done. The Corn plant growth was observed every 2 weeks, the plants were allowed to grow for 15 weeks, and then at the end of harvest they were observed (plant height, root length, plant-root biomass, and production dry seed weight). Data growth and crop production are analysed with one-way ANOVA, if there is a real difference followed by Brown-Forsythe test at 5% level.

Results and Discussion

Parameters observed in this study were, growth (plant height, root length, plant biomass, root biomass) and crops production (weight of dried seed) of corn plants (*Zea mays* L.).

Figure 1. showed the growth of corn plants (*Z. mays* L.) at the age of 2, 4, 6, and 15 weeks of AP (after planting). Figure 1. also showed the presence of different plant height between treatments, at the age of 2 weeks, treatment of PSB 10 mL/plant gave the highest for plant height (36.97 ± 2.72 cm), the highest for plant height in 4 weeks AP also gave by treatment of PSB 20 mL/plant (120.97 ± 5.55cm) in 6 weeks AP gave by PSB 30 mL/plant (182.96 ± 4.74) and at harvest 15 weeks of AP gave by the treatment of NPK (216.96 ± 19.69 cm).

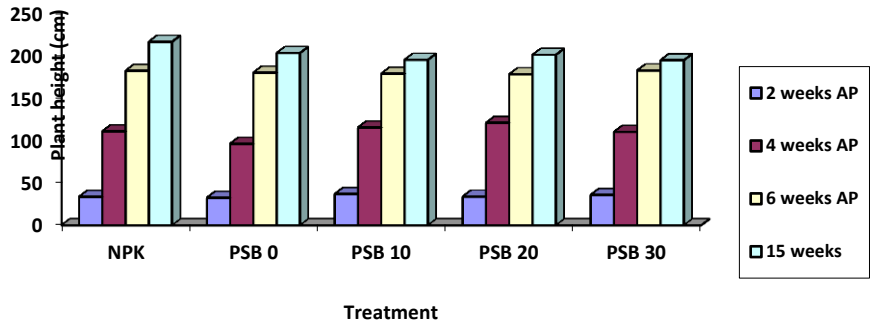

Figure 1. Effect of *PSB* on plant height of corn ages 2, 4, 6, and 15 weeks of AP (after planting)

Description:
NPK : chemically fertilized control plants (NPK 5g/plant)
PSB 0 : non inoculated with Phosphates Solubilizing Bacteria
PSB 10 : inoculated with *Phosphates Solubilizing Bacteria* 10 mL/plant
PSB 20 : inoculated with *Phosphatses Solubilizing Bacteria* 20 mL/plant
PSB 30 : inoculated with *Phosphates Solubilizing Bacteria* 30 mL/plant

Table 1. The Effect of Phosphates solubilizing bacteria (PSB) on plant growth of corn plant *(Z. mays* L.*)* at harvest (15weeks of AP)

Treatment	Plant height (cm/plant)	Root length (cm/plant)	Plant biomass (g/plant)	Root biomass (g/plant)
NPK	216.96 ± 19.69	18.25 ± 1.57	383,20 ± 72.86	34,80 ± 18.51
PSB0	201.40 ± 6.81	17,64 ± 2.19	338,80 ± 71.81	34,04 ± 12.62
PSB10	195.52 ± 18.09	17,16 ± 3.08	339,68 ± 23.60	27,16 ± 7.44
PSB20	203.56 ± 14.73	17,96 ± 1.98	315,60 ± 62.57	37,24 ± 8.25
PSB30	195.04 ± 6.54	17,73 ± 1.78	357,20 ± 49.93	40,60 ± 9.52

Description:
NPK : chemically fertilized control plants (NPK 5g/plant)
PSB 0 : non inoculated with Phosphates Solubilizing Bacteria
PSB 10 : inoculated with Phosphates Solubilizing Bacteria 10 mL/plant
PSB 20 : inoculated with Phosphatses Solubilizing Bacteria 20 mL/plant
PSB 30 : inoculated with Phosphates Solubilizing Bacteria 30 mL/plant

Table 1. showed the effect of phosphates solubilizing bacteria (PSB) with different doses of the average plant growth corn plant *(Z. mays* L.*)* at harvest 15 weeks of AP. The test results showed statistically no significant difference ($\alpha > 0.05$) between treatment of PSB and control positive (chemical fertilizer NPK) on plant growth Corn plants. The highest for growth of corn plant gave by NPK fertilizer, but NPK treatment gave results not significantly different ($\alpha > 0.05$) with treatment of PSB. Between PSB treatment, the PSB of 20 mL/plant provides the highest yield for plant height (203.56 ± 14.73 cm/plant), and for root length (17,96 ± 1.98 cm/plant) while PSB of 30 mL/plant gave the highest yield for plant biomass (357,20 ± 49.93 g) and root biomass (40,60 ± 9.52 g/plant).

Table 2. Effect of Phosphates Solubilizing Bacteria (PSB) on crop plant of Corn plant *(Z. mays* L.*)* at harvest (15weeks of AP)

Treatment	dry seed weight (g/plant)	dry seed weight (tonnes/ha)	RAE (%)
NPK	158.16 ± 18.81	15.81 ± 8.81	-
PSB0	161.40 ± 60.97	16.14 ± 6.09	-
PSB10	164.68 ± 39.25	16.46 ±3.92	203
PSB20	173.19 ± 43.60	17.31 ± 4.36	128
PSB30	131.92 ± 23.81	13.19 ±23.81	0

Description:
NPK : chemically fertilized control plants (NPK 5g/plant)
PSB 0 : non inoculated with Phosphates Solubilizing Bacteria
PSB 10 : inoculated with Phosphates Solubilizing Bacteria 10 mL/plant
PSB 20 : inoculated with Phosphatses Solubilizing Bacteria 20 mL/plant
PSB 30 : inoculated with Phosphates Solubilizing Bacteria 30 mL/plant

$$\text{RAE (Relative Agronomic Effectiveness)} = \frac{\text{Dry seed PSB- Dry seed NPK}}{\text{Dry seed PSB- Dry seed PSB 0}} \times 100\ \%$$

Table 2. showed the effect of Phosphates Solubilizing Bacteria (PSB) with different doses of the average yield of crop production of corn plant (*Z. mays* L.) at harvest 15 weeks of AP. The statistically test results showed no significant difference (α>0.05) between treatment of PSB and control positive (chemical fertilizer NPK) on plant crop production of corn plants, but PSB treatment 20 mL/plant gives good results for the highest dry seed weight (173.19 ± 43.60 g/plant, equivalent to 17.31 ±43.60 tones/ha). But the best RAE (Relative Argonomic Effectiveness) gave by PSB treatment 10 mL/plant, for corn plants.

Based on the results of statistical tests, the results of this study showed no significantly difference (α>0.05) between treatment of PSB and control positif (chemically fertilizer NPK) on plant growth and crop production of corn plants. This can be caused by phosphates solubilizing bacteria (PSB) as soil microbes such as bacteria *Bacillus megaterium* and *Pseudomonas* sp**.** are a type of PSB that can produce organic acids such as citric acid**,** glutamate**,** succinate**,** lactate**,** oxalate**,** glioksalat**,** malic**,** fumaric**,** tartaric, and α-ketobutarat**,** with increased organic acids are usually followed by a decrease in pH**,** so resulted in the dissolution of phosphate is bound by Ca (Khan *et al*., 2009)**.** Elements of phosphate (P) is an essential element that plays an important role in the process of photosynthesis and stimulates root development, plant biomass, seed formation, and accelerate the maturity of the fruit and enhance the disease resistance of plants against attack (Gonggo *et al*., 2006).

The optimum growth of PSB is at neutral pH and increases with increasing soil pH. Generally, the dominant PSB live in a pH range from 4 to 10.6. This corresponds to the soil in this study which has acidic pH 6.4. In acid soil, phosphate ions bound by Al and Fe to form compound P is less available to plants. By giving the PSB into the soil, pH decrease and P will become available again. In this study, the initial soil pH has a value of 6.4 and after addition of PSB (post harvest) soil pH decrease to 6.0, meaning that phosphate solubilizing bacteria are very effective to solubilize for dissolving the phosphate-bound P into a soluble P by deacreasing soil pH. Hasanudin and Gonggo (2004) stated that organic acids released by

bacterial phosphate solvent can khelat Fe, Al, Ca, and Mg so that the phosphate is bound strongly to soluble and available.

According to Khan *et al*., (2009), *Bacillus* sp. can dissolve P from the layer of corn plant roots and *Pseudomonas* sp. able to increase soil P extracted in acid to 50%, also reported that addition of PSB (*Bacillus* sp.) On the acid-tolerant crops such as corn can increase grain yield of these plants. Some researchers argue that effective PSB is not only due to its ability to increase the availability of P but also due to its ability to produce plant growth regulators, mainly by bacteria that live on the root surface such as *Pseudomonas* sp. These bacteria can produce growth regulators such as indole acetic acid (IAA) and gibberellins acid (GA3). In addition, some PSB can also serve as a biocontrol that can improve the health of roots and plant growth through the protection against disease.

Widawati and Suliasih (2006), reported that inoculant of potensial PSB increased the fresh plant production of green mustard (*Brasica caventis* Oed.), the mix isolate of potential PSB (*Klebsiella aerogenes, Chromobacterium lividum,* and *B. megaterium* injected in peat fine) was the best to increase the fresh plant production of green mustard until 32,87% then control. Elkoca *et al*., (2008) reported that seed inoculation with *Rhizobium* and phosphate solubilizing bacteria (PSB) such as *Bacillus subtilis* and *B. megaterium* significant increases of the seed yield of chickpea (*Cicer arietinum* L.) under different inoculation treatments ranged between by 18% *Rhizobium* and 30.5% (*Rhizobium* + PSB *Bacillus subtilis* and *B. megaterium*) over the control N, P and NP. In inoculation, seed inoculation with *Rhizobium* and PSB can substitute costly NP fertilizers in chickpea production. In this study PSB of 20 mL/plant gives the best result for dy seed weight 173.19 ± 43.60 g/plant equivalent of 17.31 ± 4.36 tones/ha, compared with the national standard value, because according to the Directorate General of Food Crops in 2010, the dry weight of corn seeds nation wide in 2009 of 16.47 tones / ha whereas in East Java at 5.19 tones/ha.

Development of environmentally sound agricultural production approach and income of farmers should be based on improving the effectiveness of fertilization on soil productivity and agricultural production systems. How to cultivate using chemical fertilizers and excessive constantly need to be revisited, especially to overcome the saturation of P fertilizer, in contrast to the effective use of chemical fertilizer P, but still cause the resulting negative impact on the environment (Saraswati and Sumarno, 2008). The RAE formula (Relative Effectiveness Argonomic) can determine how effective the use of biofertilizer such as PSB on crop production (Saraswati *et al*., 2008) it caused by the RAE outcome could determine the use of biofertilizer for the replacement of chemical phosphate fertilizers in the future.

From the results of this study indicate that the PSB 10 mL / plant gave the highest RAE value (203%), meaning that the use of PSB 10 mL/plant was quite effective compared to the PSB 20 and 30 mL/plant. This is because the number of bacteria at doses of PSB 10 mL/plant only slightly, so that the nutrients in the soil necessary for the development of bacteria sufficient. Bacteria need nutrients and optimal temperature for growth, the amount of nutrients that were lacking and the temperature was not optimal, it can lead to reduced bacterial growth (Martinko and Madigan, 2005). Bacterial populations in soil is influenced by several factors, including nutrients, pH, and temperature (Muslim, 1995). The use of small doses of PSB, will also be economically viable for farmers.

Conclusion

From the results of this study, can concluded that addition of 20 mL/plant of phosphates solubilizing bacteria (PSB) could improve crops production of corn plants (*Z. mays* L), the highest dry seed weight of 173.19 ± 43.60 g/plant, equivalent to 17.31 ±43.60 tones/ha was achieved by treatment of PSB 20 mL/plant. But the best RAE (Relative Argonomic Effectiveness) gave by PSB treatment 10 mL/plant, for corn plants.

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Inducing Somatic Embryos of Soybean *Glycine max* and *Glycine soja* on Sucrose Concentrations Variation

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Abstract

Production and productivity of Soybean in Indonesia have not met a national demand. The government program is to improve Soybean productivity and production to achieve self sufficiency on 2015. Genotype and sucrose in the medium tissue culture is important inducing somatic embryos. The objectives of this experiment was to inducing somatic embryos of Glycine max varieties Anjasmoro and Glycine soja varieties Mallika at the variation of sucrose concentration for MS medium by in vitro. The research was used immature cotyledons explant which was conducted at Greenhouse and Biotechnology Laboratory, Agrotechnology of UPN "Veteran" Yogyakarta, Indonesia from Mei to October 2010. The experiments were arranged in factorial Completely Randomized Design with two factors and replicated ten times. The first factor was explants soybeans were: Mallika (Glycine soja) and Anjasmoro (Glicyne max). The second factor was concentration sucrose: 20 g/l. 30 g/l, and 40 g/l. Data were subjected to an analysis of variance followed by Dunnet's Significance Test (DST) at 5% significance level. The results showed that material explant Glycine max and Glicyne soja not induced somatic embryos. The best sucrose concentrations 20 g/l for MS medium increased to time of embryos (days), growth percentage of embryos and fresh weight callus. The combination treatment Glicyne max and sucrose 20 g/l to increased dry weight callus.

Keyword: Embryos somatic, soybean, sucrose

Introduction

The Soybean is a crop that has rich nutritional and includes 10 crop commodities besides rice and corn commodities. In recent years, soybean production is still the range of 600-700 thousand tons per year, while the demand has reached 2.0 million tons. The low national production of soybean, as well as the total area of plantations is still limited or declining, as well as productivity per unit area remains low. This is caused by the use of low-quality seeds and by the onset of the disease (Anonymous, 2010).

Propagation in the conventional soybean plants generally requires a long time as well as a vast place so its needs to be done in biotechnology and by vegetative propagation of plants through tissue culture techniques *(in vitro).* Technology is an *in vitro* culture technique in plant breeding pieces of tissue in a sterile artificial media. The technology is based on the properties of the cell that each individual is able to form a new whole that has properties identical to the parent cell, especially the young (Wahyurini, 2008). The medium used for cultivating the tissue sections containing foods such as macro elements and micro nutrients. In addition, in the medium was also added source of carbon derived from sucrose, vitamins and growth regulators that serve to spur growth and improve the ability of cells to multiply and develop into a candidate plant (Gamborg and Shyluk, 1981 and George and Sherington, 1984).

Regeneration of plant tissue culture can be done through somatic embryogenesis and organogenesis. Somatic embryogenesis is widely used because it can accelerate the discovery of the success of transgenic crops with a high opportunity for transformation of somatic embryos which can be derived from a somatic cell. Somatic embryos can be induced directly from tissue explants or indirectly through a callus phase. Plant regeneration from callus cultures often show genetic diversity that somatic embryogenesis is more efficiently used directly in the application of biotechnology for plant breeders.

Currently somatic embryogenesis is well known as regeneration induction to way of tissue culture explants, or indirectly through a callus phase. Its success is largely determined by media formulations optimized for each stage of culture (Yusnita, 2003). The successful regeneration of soybean plants is also highly dependent on the genotype used. From previous studies induction of somatic embryogenesis in peanut, which mostly done by using several concentrations of sucrose, still produce a diverse number of embryos. So, on that ground, conducted research on somatic embryos soybean of white and black, is still the same family with peanuts as *Leguminoceae*. The research to know about the concentration of sucrose is right for the formation of soybean embryos. A problem in this study is on how the provision concentration sucrose effect on increasing the amount of soybean embryos and in a short time of planting material a bit.

Materials and Methods

This research has been conducted in the greenhouse and laboratory Agrotechnology Department of Biotechnology in May until October 2010. The materials used were: soybean seed varieties *Anjasmoro* and *Mallika*, poly bags, sand, manure, MS medium (Murashige and Skoog), jelly, sucrose, 2.4 D, disinfectants (Furadan, agrimycin, Benlate, 96% alcohol, bayclin 50%, sublimate 0.1%), sterile distilled water, aluminum foil, filter paper, gloves, and detergents. The tools used were: the culture bottles, beakers, Petridis, pH sticks, Laminar Air Flow (LAF), disinfect sets, lighting Bunsen and autoclave. The experiment was conducted using a Factorial Completely Randomized Design with 2 factors, with ten replications.

The First factor was explants material (genotype soybean) comprising 2 levels: white soybean Anjasmoro (K1) and black soybean Mallika (K2). Whereas the second factor was concentration of sucrose, which comprises three levels: 20 g / l (S1), 30 g / l (S2) and 40 g / l (S3). The data were analyzed by Variance Analysis at the level 5%. Therefore, to know there were a significant differences between the treatments then the test by *Dunnet's Significance Test (DST)* at 5% significance level.

Results and Discussion

The results of the present analysis shows that the treatment appears embryo explants material significantly affect the concentration of sucrose but the treatment did not significantly affect time of embryos. Average value the time of embryos can be seen in Table 1.

Table 1 shows that treatment of explants s K1 faster as the time of embryo than K2 treatment. At treatment concentrations of sucrose showed a significant S1 faster when compared treatments S2 and S3. In the early growth response of black soybean callus showed a faster growth than white soybean. In morphology the size and shape of white

soybeans *Anjasmoro* greater than *Mallika* black soybeans, so cotyledon as food reserves could supply the cells forming the meristem cells.

Table 1. The mean time of embryos (days)

Treatment	S1 (20 g/l)	S2 (30 g/l)	S3 (40 g/l)	Mean
K1 (white soybean)	22,25	22,25	24,75	23,167 a
K2 (black soybean)	25,50	26,25	26,25	26,00 b
Mean	23,88 p	24,38 q	25,25 q	(-)

Note : Mean followed by same small letter indicates no significant difference in the test DST 5%. Sign (-) showed no interaction

The embryo appears at day 22 after planting some sucrose treatment. Provision of sucrose with a concentration of 20 g / l markedly more rapid time of the embryo, this is due to sucrose with 20 g / l which was the best carbon source that acts as a raw material producing energy in the process of respiration (Katuuk, 1984). This energy which is used in cell-cell division to form embryos. The results of the analysis of the growth percentage of embryos showed that the treatment material did not significantly, but the concentration of sucrose significantly. Average value of the growth percentage of embryos can be seen in Table 2.

Table 2. The mean of the growth percentage of embryos

Treatment	S1 (20 g/l)	S2 (30 g/l)	S3 (40 g/l)	Mean
K1 (white soybean)	81,25	75,00	81,25	79,17 a
K2 (black soybean)	87,50	57,50	68,75	71,25 a
Mean	84,38 p	66,25 r	75,00 q	(-)

Note book: Mean followed by same small letter indicates no significant difference in the test DST 5%. Sign (-) showed no interaction

Table 2 shows that the treatment was not significantly different explants K1 with K2 treatment. At treatment concentrations of sucrose showed a greater percentage of significant S1 explants which are capable of forming embryos than treatment S2, and S2 greater than S3 significant. There was no interaction between sucrose concentration and explants material percentage of explants capable of forming embryos.

Treatment of different materials the genotype explants showed no significant difference in the percentage of embryos. Soybean embryo explants of black and white soybeans has the ability and equal opportunity for the proliferation or growth of cells so that the percentage of growing embryo no significant difference. The key to success in the formation of callus tissue of life is the existence of a sterile nutrient medium that has an optimum and suitable environment and culture (Ursila, 2004).

Provision of sucrose with a concentration of 20 g / l was the significant percentage of the growth of embryos than other treatments. The success of plant tissue culture is highly dependent on the media used. Tissue culture media does not only provide macro and micro nutrients but also the carbohydrates that in general the form of sugar. Sugar is a source of carbon instead of carbon usually obtained plants from the atmosphere in the form of CO2 into a component for photosynthesis (Gunawan, 1988).

The results of the analysis of the number of embryos per explants showed that the treatment material and the concentration of sucrose does not significantly affect the number of embryos per explants The average number of embryos per explants can be seen in Table 3.

Table 3 shows that treatment of explants K1 and K2 were not significantly different. At the concentration of sucrose treatment S1, S2 and S3 were not significantly different between treatments. There was no interaction between sucrose concentration and explants material to the average number of embryos per explants.

Table 3. Average number of embryos per explants

Treatment	S1 (20 g/l)	S2 (30 g/l)	S3 (40 g/l)	mean
K1 (white soybean)	31,39	29,67	32,21	31,09 a
K2 (black soybean)	27,29	27,75	37,34	30,79 a
mean	29,34 p	28,71 p	34,78 p	(-)

Note book: Mean followed by same small letter indicates no significant difference in the test DST 5%. Sign (-) showed no interaction

Treatment of different genotype materials explants showed no significant difference in the number of embryos per explants. In forming callus growth, white soy faster growth, but the subsequent development of both materials explants showed the same ability to form embryos. This is due to the development of cells forming embryonic cells is influenced by the nutrients contained in the media.

Provision of sucrose with a concentration of 20 g / l apparent greater number of embryos per explants compared to other treatments. This is because sucrose was an important carbon source used as a constituent of cells, cell division, cell enlargement and differentiation of cells that can form the plant shoots, and embryos as well (George and Sherrrington, 1984). The results of the analysis of wet weight of explants material shows that the treatment effect was not significant, but the treatment concentration of sucrose significantly affects callus wet weight. Average value of wet weight of callus can be seen in Table 4.

Table 4. shows that treatment of explants K1 and K2 were not significantly different in the concentration of sucrose treatment showed S1, the callus markedly more severe than the S2 and S3. There was no interaction/relation between sucrose concentration and explants material to wet weight of callus.

Treatment of different genotype materials explants showed no significant difference in wet weight of callus. In forming callus growth, white soybean faster growth, but subsequent

developments showed the same ability proliferating. This is due to the development of callus formation and embryos are fixed, so the plant will produce the same wet weight (Andarwening, 2009).

Table 4. The mean wet weight of callus (g)

Treatment	S1 (20 g/l)	S2 (30 g/l)	S3 (40 g/l)	Mean
K1 (white soybean)	1,365	1,142	1,212	1,240 a
K2 (black soybean)	1,281	1,098	1,177	1,186 a
Mean	1,323 p	1,120 q	1,195 q	(-)

Note book: Mean followed by same small letter indicates no significant difference in the test DST 5%. Sign (-) showed no interaction

On a wet weight parameters of callus that the granting of the concentration of sucrose 20 g/l showed the greatest callus wet weight compared with other treatments. The state thus induced cells in tissue explants grown on media with the addition of sucrose 20 g/l more rapidly receive the nutrients necessary for its development, while also influenced by the ability of the plant itself in receiving nutrients. Growth in the general sense is the formation, among others, the volume size, weight and number of cells (Salisbury and Ross, 1992). Dry weight analysis results showed that treatment of explants material and the concentration of sucrose does not significantly affect the dry weight of callus. There was interaction between the materials treated explants with sucrose concentration on dry weight of callus. Average value of dry weight of callus can be seen in Table 5.

Table 5. The mean dry weight of callus (g)

Treatment	S1 (20 g/l)	S2 (30 g/l)	S3 (40 g/l)	mean
K1 (white soybean)	1,734 ab	1,722 b	1,769 a	1,742
K2 (black soybean)	1,699 b	1,530 c	1,701 b	1,643
mean	1,717	1,626	1,735	(+)

Note book: Mean followed by same small letter indicates no significant difference in the test DST 5%. Sign (-) showed no interaction

Table 5 shows that the combination treatment K1S3 (white soybean and sucrose 40 g/l) did not differ significantly with treatment K1S1 (white soybean and sucrose 20 g/l) but significantly different from the other treatment combinations on the dry weight of callus. Combination treatment K2S2 (black soybeans and sucrose 30 g /l) were significantly lighter weight than other treatments. According to Septiana (2010) the process of explants growth and development can be realized by the assimilated accumulation which would be translocation into various cell explants required. If explants are not able to form assimilate sufficiently, the explants will growth vary.

K1S3 combined treatment (white soybean and sucrose 40 g / l) and K1S1 (white soybean and sucrose 20 g / l) produced the greatest dry weight of callus compared to other treatments. Dry weight of callus showed progression and cell growth. The white soybeans in sucrose concentration of 20 or 40 g / l produced the greatest dry weight of callus may be due to the density concentration and induces the cells to maintain the acidity of H + so that the water potential in the cell down and eventually enters the cell and places cell development. Provision of sucrose as a substitute for carbon is very involved in the process of photosynthesis, so the number of assimilate formed in the formation of many organs of plants (George and Sherrrington, 1984).

A B C

Figure 1. Embryo somatic with treatment K1S1 (A) after three weeks ; K2S1 (B) after four weeks ; K1S1 (C) after ten weeks

Conclusion

1. Treatment of white soybean (K1) and black soybeans (K2) does not affect the induction of somatic embryos *in vitro*.
2. The treatment concentration of sucrose 20 g / l may increase to time of embryos (days), growth percentage of embryos and fresh weight callus.
3. The combination treatment *Glicyne max* and sucrose 20 g/l to increased dry weight callus.

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ANIMAL PRODUCTION

Isolation and Characteristic of *Lactobacillus sp.* Isolated from Milks of Cattle, Goat and Homemade Yogurt's for Potential as Probiotic

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Abstract

The term probiotic has gained augmented popularity recently due to its perceived health benefits. Probiotic microorganisms consist primarily of lactic acid bacteria (LAB) and received great attention. Lactobacillus one of the probiotic classified as a Gram positive bacteria, catalyze and oxidase negative reaction and non-sporing rods or cocci. The aim of this study was to isolate and identify the Lactobacilli from dairy milks such as cattle's milk (Brahman, Jersey and Australian Friesian Sahiwal), goat's milk (Capra hircus) and homemade yogurts at Bestari Jaya's farm, Selangor. A total of 101 LAB bacteria had been isolated during January until April 2011. Classical identification on morphological, biochemical and molecular characteristic were studied. Those isolates were enriched in MRS broth and isolated using MRS agar plate in anaerobic condition at 37^{o}C. The phenotypic characteristic was determined through macroscopic examination on agar plate and Gram stained. Subsequently, biochemical test were conducted to determine the catalyze and oxidase reaction of the isolates. Out of 101 isolates, only 55 isolates were successful examined as Gram positive and negative for oxidase and catalyze test belong to Lactobacillus genus. However, for reliable identification, molecular technique application of 16S gene sequencing will be applied in order for the species identification of Lactobacillus. Probiotics characteristic on antibiotic resistance, pH and temperature tolerance, and plasmid profiling will be also carry out through in this study due to the capability of the important probiotic strain which has beneficial health care.

Keywords : cattle's milk, characteristics of Lactobacillus sp., goat's milk, PCR, yoghurt

Introduction

Probiotic is defined as live microorganisms which when administered in adequate amounts confer a health benefit on the host (Guarner *et al.,* 2005). Microbes from many different genera are used as probiotic but the most commonly utilized strains are lactobacilli. Lactobacilli are known to be the foremost bacteria in the lactic acid bacteria (LAB) group and are broadly used as food-associated microorganism as they are generally recognized as safe (GRAS) and the lactic acid and other organic acids produced by these bacteria act as natural preservatives as well as flavor enhancers. Probiotic are naturally available in foods such as yogurt, fermented and unfermented milk and cheese but the dominant food vehicles remain to be yogurts and fermented milks. Yogurt is a fermented milk product which is believed to be one of the best-known foods that contain probiotic. There are several selective media to isolate LAB and numerous tools to characterize them at genus, species or strain level. Modern molecular techniques have become increasingly important

and are extremely valuable for species identification or differentiation of probiotic strains, specific characterization and detection of strains selected for application as probiotic. Most commonly used methods are polymerase chain reaction (PCR) and pulsed-field gel electrophoresis (PFGE). There is a rising requirement for accurate quantity and quality control of the probiotic products and hence methods for specific identification of probiotic strains. Consequently, the aim of this effort was to isolate and identify *Lactobacillus* genus from locally made fresh yogurt through biochemical and molecular method using 16S rRNA gene sequence analysis and to study the phenotypic characteristics of the isolated strains.

Material and Method

Isolation of Lactobacillus

Lactobacillus was isolated from dairy samples which from cow's milk (Brahman, Jersey and Australian Friesian Sahiwal), goat's milk (*Capra hircus*) and homemade yogurts at Bestari Jaya Farms, Selangor. Serial dilutions of MRSB from 10^{-1} to 10^{-10} were prepared and incubated anaerobically in anaerobic jar contained Anaerocult @ A (Merck) for 24 hours at 37^{o}C. A 0.1ml of inoculum was spread plate onto MRS agar and incubated in anaerobic condition. After 72 hours incubation, 101 of single colonies of each sample were randomly selected and examine its morphology. Colonies with gram positive were only further tested to catalase and oxidase reaction.

Genomic DNA Isolation and Quantification

In this study, *L. casei* ATCC 393 culture was used as a positive control. The extraction of genomic DNA was performed using Phenol-Chloroform protocol according to Parenrengi *et al*., (2000) with some minor modifications. The concentration and purity of the DNA extracted were determined by a Nano-spectrophotometer (Pearl, Implen, Germany).

Polymerase Chain Reaction (PCR)

PCR reactions for genus identification were carried out in a total volume of 25 µl per reaction using 16S rRNA primers according to Abu Bakar *et al*., 2010. A PCR master Mix was freshly prepared and amplified in a thermocycler (SensQuest) using the following conditions: the initial denaturation step was performed at 95°C for 3 minutes and the target DNA was amplified in 40 cycles where each cycle consisted of denaturation at 95°C for 30 seconds, annealing at 61°C for 30 seconds and extension at 73°C for one minute. The final extension step was performed at 73°C for 5 minutes and the holding temperature was at 10°C (An *et al*., 2006).

Results and Discussion

Morphological and Biochemical Test

After morphological examined, only a pure culture was chosen which 7 colonies isolated from cow's milk, 28 colonies isolated from goat's milk and 20 colonies isolates form homemade yogurts were examined as rod shape, gram positive, negative catalase and negative oxidase results. A total number of 55 isolates were proceeded to molecular analysis.

Genomic DNA Extraction and Quantification

The quality of genomic extracted was determined by 1.0% of agarose gel electrophoresis and quantify using nano-spectrophotometer (Pearls, Implen, Germany). All the genomic isolates were successful extracted as shown in Figure 1 and the A_{260}/A_{280} ratio value resulted between 1.7 to 1.9.

Amplification of *Lactobacillus* Genus

PCR amplification was performed using genus-specific primers, Lacto-16S-Forward and Lacto-16S-Reverse to confirm the isolates belong to the genus *Lactobacillus*. Hence, all the 55 isolates managed to amplify the *Lactobacillus* genus with approximately 216 bp band.

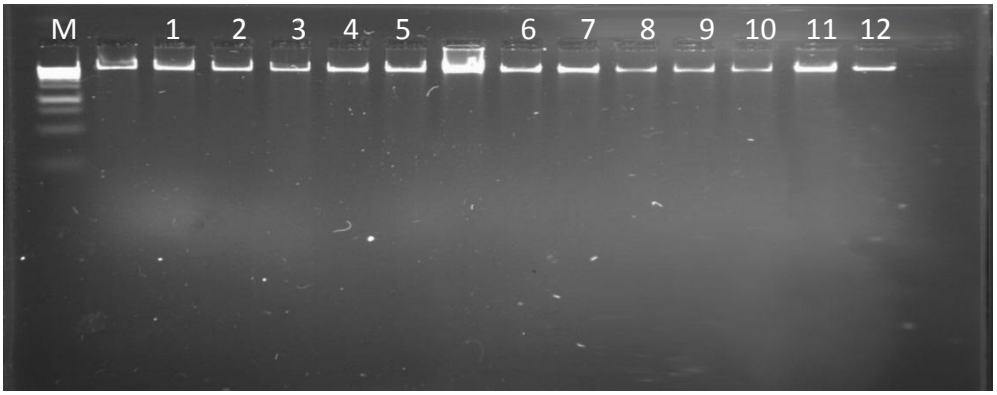

Figure 1. A representative of 1.0% of agarose gel electrophoresis for genomic DNA extraction of *Lactobacillus* cultures. Lane M=1 kb DNA ladder marker; Lane 1: *L. casei* ATCC 393; Lane 2: LG01R-LG12

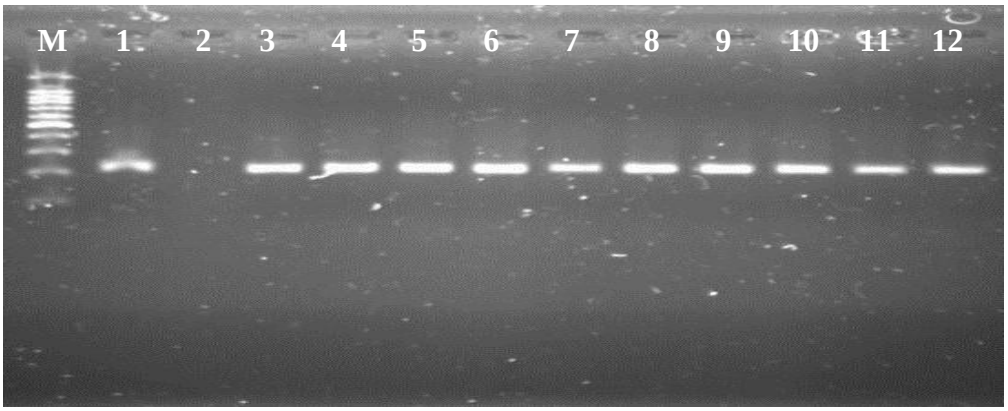

Figure 2. 2.0% of agarose gel electrophoresis of amplified 16S rRNA gene for identification of *Lactobacillus* genus using primer Lacto-16S (forward and reverse) with molecular weight of PCR product approximately at 216 bp. Lane M: 100 bp DNA Ladder, Lane 1: Positive control (*L. casei* ATCC 393), Lane 2: Negative control (without DNA), Lane 3-12: LG01-LG10.

In this study, *Lactobacillus* as a probiotics microorganisms are the most source among the other probiotics from animal or human origin. However, some studies show that it's also found in non-dairy fermented substrates such as fruits and vegetables, meats, cereals and soya. Recommended by De Man *et al*., (1960); De Man, Rogosa, Sharpe agar and broth (MRSA and MRSB) as medium for use in the isolation, enumeration and cultivation of *Lactobacillus* species. A few studied by Martin *et al*. (2010); Franciosi *et al*. (2009); Ortu *et al*. (2007); and Erdogrul and Erbilir, (2006) said that these media to support luxuriant growth of all lactobacilli from oral, fecal, dairy, and other sources contained peptone and dextrose which supply nitrogen, carbon, and other elements necessary for growth. Two biochemical tests which catalase and oxidase was tested to these isolates. Catalase reaction was test to detect the presence of gas or hydrogen in culture which its break down the hydrogen peroxide to hydrogen and oxygen. Probiotics are facultative anaerobic bacteria that catalase negative, no oxygen or bubbles produce. Then, oxidase test utilized to look out for the presence of the enzyme indophenol oxidase. N,N,N,N,-tetrammethyl-para-phenylenediamine oxidized in the presence of atmospheric oxygen by formation causing of indophenol oxidase. However, the physiological and biochemical criteria are sometimes ambiguous in terms of precise identification of the interest microorganism (Martin *et al*., 2003). Thus, there has been an explosion in the development and application of molecular tools for identifying microbes and analyzing their activity in the recent years. These tools are increasingly applied to strains of probiotics for identification and analysis of their activity (Amor *et al*., 2007).

Since genotypic approaches hold the most promise for the rapid and accurate identification of lactobacilli (Tannock, 1999), many of these tools are based on 16S rDNA sequences and exploit PCR techniques. These 16S rDNA based methodologies are robust and superior to traditional methods based on phenotypic approaches which are often unreliable and lack the resolving power to analyze the microbial composition and activity of bacterial populations (Amor *et al*., 2007). According to Singh *et al*. (2009), identification of lactobacilli using biochemical method have problems identification due to large numbers of species groups and plasmids transfer among them and this approach need plenty of cumbersome biochemical tests. Therefore, they are not sufficient for differentiation and need to proceed with sensitive molecular methods to obtain more reliable identification. In 2010, Markiewicz *et al*. said there are alternative procedures of rapid identification and typing of *Lactobacillus* isolates to be more equally effective and shortened from 1 week to 2–3 days in comparison to biochemical methods. So, in this study, a set of 16S primer for *Lactobacillus* genus designed by Abu Bakar *et al*. (2010) was used and gave successfully PCR results with size of product in 216 base pair. However, this study is still needed to identify until the species level using molecular methods such as sequencing or others DNA fingerprinting method. From these findings, all isolates will then proceed to analysis the species of *Lactobacillus*. These cultures were maintained as Unisel's probiotics culture collection at *International Islamic Academy for Life Sciences and Biotechnology* (IAB) Laboratory, Shah Alam, Selangor.

Conclusion

Based on the research conducted, it was apparent that *Lactobacillus* genus dominated the probiotics microorganisms present in the dairy milk. All the isolates obtained at random from the sample exhibited characteristics favorable to that of *Lactobacillus* genus and were further continue species level through molecular analysis to authenticate the presence of the desired strain of *Lactobacillus*. Thus, subsequent to this discovery, an advance analysis

should be conducted at species level with more rapid and precise to provide a better understanding that could discriminate among these strains of Lactobacillus.

Acknowledgement

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Live Weight Changes of Bali Cattle as Draft Animal Under The Integration of Oil Palm-Cattle System in Bengkulu

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Abstract

The importance of Bali cattle rearing under the oil palm plantation has multi-purpose functions. These include as for weeding control, saving labor costs, providing manure compost, cutting down fertilizer cost, calf life saving, and as working animal for transporting Fresh Fruit Bunch (FFB) of the oil palm. When the Bali cattle was used as working animal, the severity of draft load could severely affect the live weight through decreasing feeding time and rumination. The aim of this research was to evaluate the effect of work on live weight of Bali cattle under the integration of oil palm-cattle system in Bengkulu. Mature (2.4 years old) Bali cattle (20 cows and 20 bulls) with the body condition score of 7 (range 0-10) were randomly assigned to two groups of working and control group. Working group start working by pulling cart from home to the plantation, pulling cart with FFB from plantation to collection site, and from plantation back to home. Animal graze native pasture under oil palm plantation during working days and housed after work. Parameters measured were body weight, hearth girth, withers height and body length. All data were analyzed using analysis of variance using one way design and the Duncan Multiple Range Test (DMRT) were used to examine the significant differences between mean values. The result of the research showed that there is no significant effect (P>0.05) of work on live weight and there is no significantly differences of live weight (P>0.05) was found between female and male Bali cattle. Our findings also showed that working cattle had no significant effect (P>0.05) on hearth girth, withers height and body length.

Keywords: Bali cattle, integration, liveweight, oil palm, working animal

Introduction

To fulfill the domestic meat consumption Indonesia has largely depends on imported beef meat, mainly from Australia. This was due stagnant of cattle population growth which is not macthed to the beef demand. One of the reason for this was decreasing available land for pasture and cattle rearing (Sutardi, 2001). In Sumatera, the decreasing land available for cattle rearing was mainly due to vast growing of housing and oil palm industry. Although the integration of oil palm-cattle system has not been popular in Indonesia, the use of un-used land in the oil palm area for rearing cattle has been studied in Malaysia (Wan Mohamad, 1978; Chee dan Faiz, 1990; Tajuddin *et al*., 1990).

Oil palm farmers has found difficulties in transporting their Fresh Fuit Bunch (FFB) from harvesting site to collecting site or Crude Palm Oil factory. There are several benefits of introducing Bali cattle into the oil palm plantation, as draft animal for carrying and transporting FFB from the field to the collecting site or factory, as for weeding control, saving labor costs, providing manure compost, cutting down fertilizer cost, and calf life saving. Bali cattle has superiorities in some aspects such as high fertility rate, high

adaptation ability, good ability to recover condition after poor usage, and good working ability (Martojo, 2003; Chamdi, 2005)

The use of cattle for draft animal in certain circumstances has reduced the growth as well as the reproduction performances, depending on the severity of draft load. The reduction of productive and reproductivity was directly caused by reducing the time avaliable for the cattle to feed and ruminate, increasing body heat, and increasing energy needs (Teleni dan Murray, 1991).

Materials and Methods

Research was conducted in oil palm plantation of PT. Agricinal, Bengkulu Utara Regency, Bengkulu Province, Sumatera, Indonesia. Forty (40) Bali cattle having 3 years old and *Body Condition Score* of 7 were divided into four treatments having 10 cattle each, grouped as Bull *No-Working,* Bull *Working,* Cow *No-Working, and* Cow *Working.* The *Working* Group were subjected to work, as:

1. Work-1 = Pulling carriage from farmer's home to harvesting site.
2. Work-2 = Pulling carriage from Fresh Fruit Bunch harvesting site to collection site,
3. Work-3 = Pulling carriage from harvesting site to farmer's home

During working time, physiological parameters (rectal temperature (^{o}C), pulse rate (x/minute), and respirastion rate (x/minute) were regularly measured to checked all animals used were not fatique (Uphadyay dan Madan,1985;). Draft parameters such as distance (km), duration (hour), load (kg), and speed (km/hour) were also measured for the *Working Group.*

Live weight measurements were conducted every 2 weeks for 11 weeks, while data obtained were analysis using ANOVA in which the difference between mean were tested using DMRT (Daniel,1991).

Results and Discussion

Data on live weight of *Working* and *No-Working* bull and cow measured during the experiment are presented in Table 1. It is shown that there is no effect (P>0.05) of work on live weight during 11 weeks measurement period.

Table 1. Mean ± standard deviation of live weight (kg) measured during 11 weeks of *No-Working* and *Working* bull and cow.

Weeks	Bull		Cow		P
	No-Working	***Working***	***No-Working***	***Working***	
1	186.30 ± 13.31 [a]	202.45 ± 12.27 [a]	176.25 ± 6.94 [a]	181.95 ±12.60 [a]	0.05
3	187.00 ± 12.81 [a]	203.75 ± 12.22 [a]	176.80 ± 7.53 [a]	182.55 ±14.93 [a]	0.05
5	187.68 ± 12.86 [a]	204.05 ± 11.13 [a]	176.40 ± 7.26 [a]	183.85 ±13.62 [a]	0.05
7	188.25 ± 12.69 [a]	203.55 ± 11.62 [a]	176.70 ± 7.32 [a]	184.90 ±13.76 [a]	0.05
9	188.10 ± 12.23 [a]	203.90 ± 11.56 [a]	177.60 ± 7.76 [a]	184.70 ±13.79 [a]	0.05
11	187.50 ± 12.85 [a]	203.22 ± 12.82 [a]	177.21 ± 10.77 [a]	184.75 ±17.50 [a]	0.05
P	0.05	0.05	0.05	0.05	

Note : Means with different superscripts, in the same measurements within the same column or the same row, differ significantly

Draft animals in good condition and suitable live weight are essential requirements to ensure timeliness in carrying FFB. This is so because work output is a function of body size and working animals use fat reserves to fuel muscular activity during work. Under practical conditions, in Indonesia for example, most working animals lose weight (*e.g.,* Ffoulkes, 1986; Winugroho *et al.,* 1989). In this experiment, Table 1 indicated there is no indication of Bali bulls and cows used for pulling FFB losed weight. This would due to the the duration of and the workload to which the bulls and cows were subjected could be categorized as light work (Dwatmadji, 2007). The longest working distance travelled in this experiment was 3.1 km, with maximum load of 269 kg, speed of 0.6-1.8 km/hour, and duration of 20-58 minute. As there was no live weight losses of all animals also indicated that there was no energy deficits and body condition losses in the working animals. Such losses would probably adversely affect production and reproduction efficiency of cows in the future (Teleni and Hogan, 1989; Teleni *et al.,* 1989; Zerbini *et al.,* 1993).

Similar to the the data of live weight, there is no significantly difference between the *Working* and *No-Workng* Group of all measurements of hearth girth, withers height and body length. Measurements of physiological parameters has also indicated that there was no fatique condition observed of the all *Working* bulls and cows (Uphadhay and Madan, 1985; Pearson, 1985). The result of the present experiment has higlighted that the use of Bali bulls and cows as draft animal in oil palm plantation would not be detrimental to the animals.

Conclusion

It can be concluded that there were no live weight losses of the bulls and cows used for draft animal under oil palm plantation, and both bulls and cows used for draft animal was not in fatique condition.

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Characterization of Cellulose Enzyme from Milkfish *(Chanos chanos)* Gastrointestinal As Potential Agent to Degrade Cellulose

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Abstract

Milkfish (Chanos chanos) is a herbivore fish that consumes phytoplankton. There are so many floral bacteria that help milkfish in digestion process. Cellulolytic bacteria that produce enzyme from the milkfish digestive track have potential to be used by human. Cellulose enzyme can be used for composting agriculture waste, biorefining for producing eco health paper, increasing digestibility of forage and biofuel production. The purpose of this study was to isolate cellulolytic bacteria and to characterize the cellulose enzyme from bacteria isolated from milkfish's gastrointestinal. Milkfish was dissected aseptically. One gram of sample of stomach and intestine that have been mashed suspended into 9 ml of physiological saline solution then inoculated on Carboxyl Methyl Cellulose (CMC). Calculation and testing of cellulose activity was performed by Miller's Method. Characterization of enzyme and testing the enzyme activity in degrading organic waste were also conducted. Eight cellulolytic bacteria were successfully isolated from Milkfish digestive tract. Two isolates with the largest cellulolytic index and the biggest sugar reduction were BSA B1 and BSA E2. Both isolates had an optimum enzyme activity at pH 9 and temperature at 50 ° C. BSA B1 had Avicell specific substrate, meanwhile BSA E2 had specific substrate on CMC. The potential of the two isolates in degrading organic waste in the form of straw and banana peels was good. Activities of BSA B1 and BSA E2 enzyme on substrate straw waste were 0.084 and 0.052 units per ml, respectively. Meanwhile activity of BSA B1 and BSA E2 enzyme on a banana peel substrates were 0.35 and 0.03 units per ml.

Keywords: Cellulolytic bacteria, cellulose enzyme, milkfish,

Introduction

Milkfish (*Chanos chanos*) is a herbivore fish that consumes phytoplankton and has a long digestive track (Lathifah, 2009). There are so many floral normal bacteria that help milkfish in digestion process. Flora normal bacteria have potential in breaking phytoplankton cell wall that contains cellulose. Cellulolytic bacteria that produce enzyme from the milkfish digestive track have potential to be used by human. Cellulose enzyme can be used for composting agriculture waste, bio refining for producing eco health paper, increasing digestibility of forage and bio-fuel production (Alexander, 1965 ; Dina *et al*, 2007; Maryandini *et al*, 2009).

The potency of cellulose enzyme as cellulose degrading agent should be developed, to reuse the organic waste become something useful, like organic fertilizer (compost) and to decrease a number of cellulose or organic waste that may cause negative effect to environment. For this reason, it is valuable to know the characteristic of enzyme in order to be utilized optimally. The purposes of this experiment were therefore to obtain the

cellulolytic bacteria from milkfish (*Chanos chanos*) stomach, and to characterize of the enzyme.

Materials and Methods

Acclimatization milkfish

Milkfish was obtained from Center of Aquaculture Development Briny / BBPBAP (*Balai Besar Pengembangan Budidaya Air*), Jepara. Sample was acclimatized for two days in Animal Taxonomy Laboratory, Biology Faculty, Universitas Gadjah Mada, by giving phytoplankton as the feed.

Isolation of bacteria

Milkfish dissected aseptically to take the digestive organ (stomach). One gram of sample of stomach and intestine that have been mashed, suspended into 9 ml of physiological saline solution then inoculated on enrichment media Trypotone Soya Broth (*TSB*) for 24 hours. Inoculation was conducted on selective media Carboxyl Methyl Cellulose (*CMC*) agar. The colonies that have been grown, washed by congo red. Colonies that showed clear zone after spilled by congo red can be categorized as cellulolytic bacteria.

Growth curve measurement

Bacteria growth measurement and enzyme activity testing were done by interval within 24 hours. Growth of bacteria is measured by using spectrophotometer by wave length 660 nm.

Cellulose activity testing

Five ml of samples were centrifuged by using speed 10.000 rpm for 10 minutes. Supernatant is the crude enzyme that will be tested the activity. The cellulose enzyme activity was measured by Miller's method (1959) by adding 1 ml of crude enzyme into 1% CMC substrate in phosphate buffer of pH 7 for 15 minutes. The reaction was stopped by adding of 2 ml of Dinitro Salicylate Acid (DNS) and boiled for 15 minutes, and then the sample awaited until the temperature down become room temperature and measured by wave length 540 nm. One unit of cellulose activity is defined as number of enzyme that produces 1 µmol of glucose per minute.

Enzyme characterization

Cellulose enzyme characterization included determination of pH, optimum temperature, and the suitable substrate. Characterization of cellulose activity was obtained by mixing 1 ml of crude enzyme with 1 ml of substrate, then incubated for 10 minutes and the reaction is stopped by heating, level of reduction sugar production stopped by heating, and then the reduction sugar product that has been created, measured by spectrophotometer.

Determination of optimum pH

Determination of optimum pH was done by dissolving of the crude enzyme into 1% of CMC substrate in the different pH condition, (acid condition in pH 2 and 4, neutral condition in

pH 7, and alkali condition in pH 8 and 10) by using citrate acid buffer (pH 2 and 4), phosphate buffer (pH 7), and NaOH Glycine buffer (pH 8 and 10).

Determination of Optimum temperature

Determination of optimum temperature was done by testing cellulase activity on different temperature (20°C until 50°C within interval 10°C) in 1% of CMC substrate in the optimum pH buffer and incubated for 30 minutes

Determination of substrate

Determination of optimum substrate was done by testing cellulase activity in different substrate (CMC, AVICEL, Whattman Paper) in the suitable buffer and optimum pH, incubated in optimum temperature.

Enzyme activity testing for degrade cellulose

Enzyme activity testing for degrade cellulose was done by mashed straw and banana peel then straw and banana peel powder created as a substrate for enzyme activity test in the suitable buffer and optimum pH, incubated in optimum temperature.

Results and Discussion

BSA B1 showed the ability in producing reduction sugar for 0,281% and cellulolytic index for 1.036. As for BSA E2 isolates showed the second biggest for reduction sugar and cellulolytic index, were obtained level of reduction sugar 0.231% and cellulolytic index 0.756. Determination of reduction sugar level was done to decide how the ability of isolates in breaking cellulose that has been contained in CMC substrate into monomer-monomer of simple sugar. Cellulose is the polysaccharide that consist of glucose monomer that has been bound by -1,4 glycoside bond. By cellulase enzyme, that bond will be degraded until cellulose become monomers. Cellulolytic index is decided by calculation of clear zone diameter that has been created by colonies of bacteria isolates after the isolates were spilled by Congored reagent.

Table 1. Cellulolytic Index and sugar reduction BSA B1, BSA E2, LBA, BSA B3

No	Isolat Code	Sugar Reduction (%)	Cellulolytic Index
1.	BSA B3	0.213	0,667
2.	LBA	0.21	0,463
3.	BSA E2	0.231	0,705
4.	BSA B1	0.281	1,036

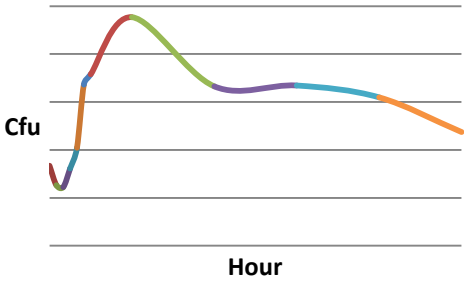

Figure 1. Bacteria BSA B1 growth curve

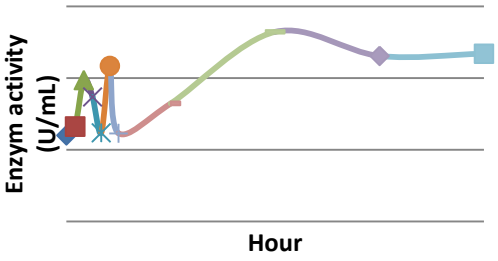

Figure 2. BSA B1 enzyme activity

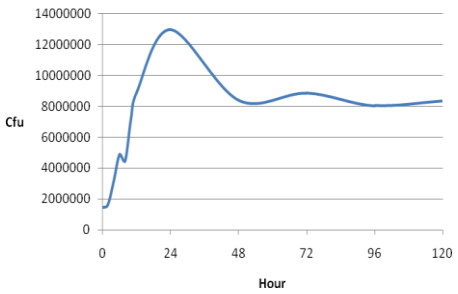

Figure 3. Bacteria BSA E2 growth curve

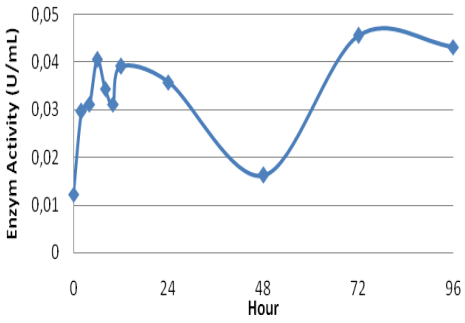

Figure 4 . BSA E2 enzyme activity

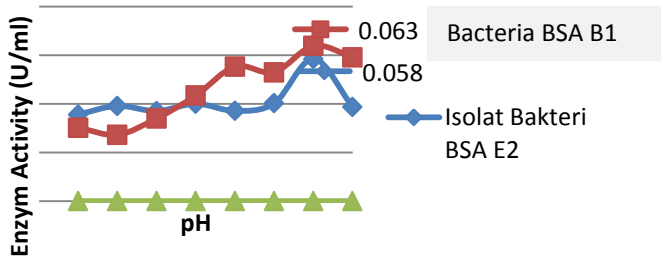

Figure 5. Enzym activity by BSA B1 dan BSA E2 in different pH

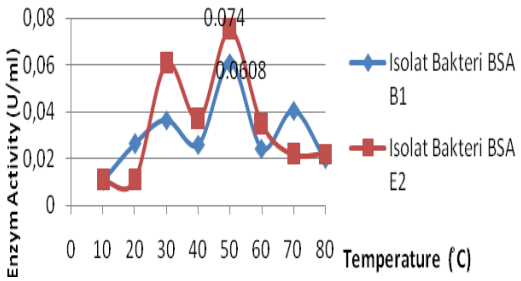

Figure 6. Enzym activity by BSA B1 dan BSA E2 at different temperature

These two isolates have the enzyme optimum activity at pH 9 and temperature is 50°C.

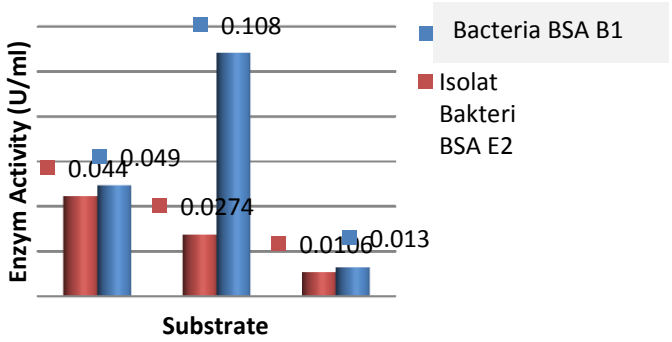

Figure 7. Enzym activity by BSA B1 dan BSA E2 at different substrat

Isolates of BSA B1 matched at AVICEL substrate, while BSA E2 matched at CMC substrate.

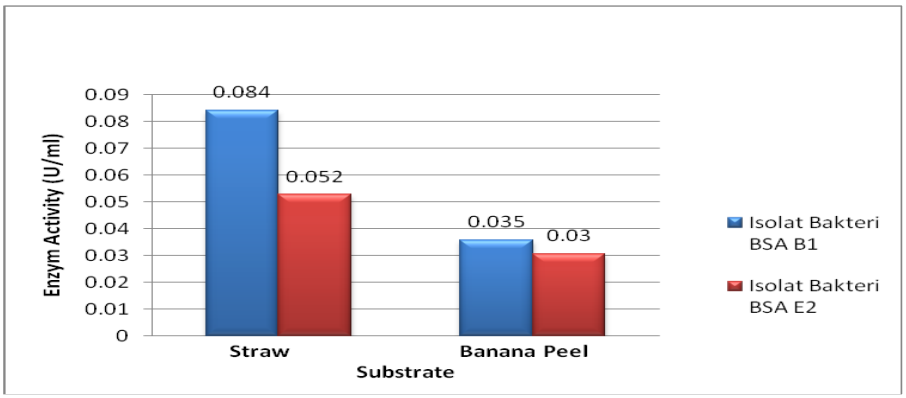

Figure 8. Ability cellulase enzyme in degrading cellulose

The potency of two isolates in degrading of organic waste likes straw and banana peel can be considered good enough. Enzyme Activity of BSA B1 at straw substrate as big as 0.084 unit/ml, while BSA E2 enzyme isolates 0.052 unit/ml. Enzyme activity of BSA B1 at banana peel substrate as big as 0.35 unit/ml, while BSA E2 enzyme isolates as big as 0.03 unit/ml.

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Layer Productivity as Affected by Different Feeding Portion

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Abstract

The research was aimed to determine feeding portion that support optimal productivity. The materials used on experiment were 11 weeks of 252 heads of Lohmann Brown. Feeding portion treatments were as follows: T1 = 100% (once at morning); T2 = 30:70%; T3 = 40:60%; T4 = 50:50%; T5 = 60:40%; T6 = 70:30%; T7 = 100% (once at noon). Each treatment was replicated 4 times with 9 hens per unit experiment. Completely randomized design was used for this laboratory experiment which applied when the hen reached 22 weeks old. Split in time design was used for the following variables : feed intake, Hen Day Production (HDP), egg mass, egg weight, feed conversion ratio (FCR). The Feeding portion treatment as main plot and age as subplot. The observation divided into 2 periods of age, 22-26 weeks and 27-32 weeks. The result showed that was not an interaction between treatment and age that influenced feed intake, HDP, egg weight, egg mass, and FCR. There was interaction between treatment and age for FCR at 22-26 weeks. The treatments influenced intake and egg weight at 22-26 weeks and at 27-32 weeks. Hen Day Production at 27-32 weeks, shell thickness at 22-26 weeks were affected by the treatments. Egg mass and FCR were not influenced by treatment. This research concluded that giving feeding portion of 70:30% at 22-26 weeks had egg weight higher than other treatments.

Keywords: feeding portion, layer, productivity

Introduction

One solution to increase business efficiency is by optimizing the performance or productivity of laying hens. Indicator of success lies in maintaining the productivity of laying hens in this case the production of eggs. Improved feed quality and better management will determine the efficiency of business. Increased business efficiency through improved quality of rations is relatively inexpensive and therefore management plays an important role in improving business efficiency.

Management of rationing applied by the farmer is to regulate the amount of diet in a day. Determination of feeding portion in addition to considering the environmental conditions should also refer to the cycle of egg formation. The process of egg formation takes approximately 24 hours. Eggs produced on a particular day is the result of metabolic processes on the previous day. Oviposition processes perform mostly in the morning until noon (Ezieshi *et al.*, 2003). Therefore in the morning, nutrient intake needed sufficient for the formation of eggs. Nutrients, especially energy and more protein, are needed in the morning but the calcium is needed in the afternoon, because the process of eggshell formation occurs at night (Rao *et al.*, 2002).

Variations in temperature and high humidity in the morning and afternoon led to inefficient use of nutrients if not offset by setting the appropriate portion of rationing. Excessive provision of diet in the morning is not effective because the body temperature will rise as a result of the thermogenic process of digestion and metabolism. The thermogenic effect of

rationing in the morning will coincide with an increase in environmental temperature would exacerbate heat stress. According to Rao *et al.* (2002) thermogenic effect ended after 8-10 hours at a temperature of 35 ^{o}C, compared with only 2 hours at 20 ^{o}C. This requires farmers to keep poultry from the diet during hot weather or during the day where the temperature increases and reaches its peak.

Based on the description above would need a study to provide information regarding the effect of feeding portion on productivity of laying hens. The purpose of this study was to examine the feeding portion to productivity of laying hen. The results of this study is expected to be a reference for farmers in relation to the management of feeding regimes.

Materials and Methods

Research conducted in the experiment laboratory, in Bandungrejo village district, Mranggen, Demak regency. The materials used were 11 weeks of 252 heads of Lohmann Brown laying hens placed in battery cages with size 30 x 35 cm/head. Battery cages equipped with a sectional diet and drinking places.

Layout Experiment

Chicks were equally and randomly divided and distributed into seven groups of treatments (T1, T2, T3, T4, T5, T6 and T7) having four replications in each. Each dietary experiment groups consist of 36 chicks distributed in four replicated pens (R1, R2, R3 and R4) with nine chicks in each. Feeding portion treatments were as follows: T1 = 100% (once at morning); T2 = 30:70%; T3 = 40:60%; T4 = 50:50%; T5 = 60:40%; T6 = 70:30%; T7 = 100% (once at noon).

Management

Cage cleaning and disinfection carried out before the chickens come. Chicken of the new arrivals were given electrolytes and vitamins through drinking water to supply the energy lost during the trip. Grower diet were given after 3 hours of drinking. Vaccination carried out periodically according to age and needs (Layer Management Guide). Coryza vaccine (Vac IC) given at the age of 13 and 18 weeks via intramuscular. Route ND, IB, and EDS vaccine were given at 15 weeks via intramuscular. ND vaccine via drinking water given at age 19 weeks. Revaccination ND performed at 1 month through drinking water. During the adaptation period were given diet with the same amount in accordance with the needs (Layer Management Guide). Crumble diet during the grower (B22) produced by PT CPP Semarang. Chickens were fed twice a day (50:50%). Weighing the remain of the diets carried out every week. Body weights were weighed once every 2 weeks by taking 30% population of the chicken to monitor the uniformity of body weight. Grower diet with the mixing layer diet started at age 19 weeks. Turnover diet was done in stages over 12 days with details of the first 4 days of 75% grower diet: 25% layer diet, the second 4 day 50% grower diet: 50% layer diet, and the third 4 day 25% grower diet: 75% layer diet. Furthermore, given 100% layer diet. Layer diet is a complete feed-shaped kibble (T24K) produced by PT CPP Semarang. The composition of layer diet were presented in Table 1. Weighing the overall body weight performed at the age of 21 weeks. Average feed intake and body weight at age 21 weeks, 78,86 g ± 6,68 g (cv = 8,47%) and 1596,34 g ± 89,75 g (cv = 5,62%) respectively.

Diet in the morning were given every day at 07.00 a.m while at noon the diet given at 01.30 pm. Amount of diet is adjusted to the standard requirements Lohmann strain. The remaining diet weighed every week to determine feed intake. Recording the number and weight of eggs weighing were done every day. Daily data accumulated during the week for an average taken weekly. Measurement of egg shell was done with 1 hour soaking in water for easy take off membrane. Then dried in oven for 1 hour (Hasnath, 2002) and shell was ready to measured the thickness.

Table 1. The composition of layer diet (T24K)

Nutrient	Content
Gross Energy (kal/g)	3624,62
Metabolizable Energy (kal/g)*	2758,26
Dry matter (%)	90,34
Ash (%)	11,63
Crude protein (%)	20,22
Ether extract (%)	7,13
Crude fiber (%)	6,00
Nitrogen free extract (%)	45,36

* (Indreswari, 2007)

Statistical analysis

All the recorded and calculated data were analyzed for ANOVA (Steel and Torrie, 1980) using a Completely Randomized Design (CRD) with the help of computer packaged SAS program (SAS Institute, 1996). Completely randomized design was used for this laboratory experiment which applied when the hen reached 22 weeks old. Split in time design was used for the following variables : feed intake, Hen Day Production (HDP), egg mass, egg weight, FCR. The feeding portion treatment as main plot and age as subplot. The observation divided into 2 periods of age, 22-26 weeks and 27-32 weeks. Duncan Multiple Range Test (DMRT) was calculated to compare the variations between the treatments were ANOVA showed significant differences.

Results and Discussion

Feed Intake

There was no interaction between treatment and age to feed intake (Table 2). This means that the treatment effect was relatively similar at all ages. The treatment feeding portion highly significant (P <0,01) to all ages. It can be shown that the chicken that got more share of diet in the morning will consume the diet more responses. This is understandable because the chickens try to meet the need of nutrients for egg formation. Duncan and Hughes (1975) stated that the feed intake patterns are influenced by egg formation cycle.

Laying hens to eat more on days in which the egg is formed than on the day which does not occur the formation of eggs (Roland *et al.*, 1972). During this study as much as 88,09% of the process oviposition occurs between 06.00 to 12.00 a.m. (Indreswari, 2007). The results Ezieshi *et al.* (2003) showed the process of oviposition a lot more done at 09:00 to 10:00 a.m. The smallest HDP value in the study Ezieshi *et al.* (2003) generated at 02:00 to 03:00 p.m. and 03:00 to 04:00 p.m.

Table 2. Layer Productivity

Variable	Feeding Portion (%)							Pr>F		
	100 in the morning	30:70	40:60	50:50	60:40	70:30	100 in the afternoon			
	T1	T2	T3	T4	T5	T6	T7	T	age	T x age
Feed intake (g)										
22-26 weeks	93,28^{A}	91,00^{B}	91,30^{B}	91,10^{B}	90,35^{B}	91,05^{B}	88,31^{C}	0,0001	0,0001	0,8467
27-32 weeks	103,19^{A}	101,99^{BC}	101,78^{C}	101,75^{C}	102,01^{BC}	102,78^{AB}	102,41^{ABC}	0,0046	0,0001	0,2612
Egg Production/HDP (%)										
22-26 weeks	21,22^{ab}	19,76^{ab}	24,13^{a}	21,11^{ab}	16,83^{c}	23,25^{a}	18,57^{ab}	0,1069	0,0001	0,9285
27-32 weeks	88,11^{abc}	89,90^{a}	84,78^{c}	88,66^{ab}	87,42^{abc}	86,48^{bc}	86,29^{bc}	0,0317	0,0001	0,1348
Egg weight (g)										
22-26 weeks	40,87^{A}	36,85^{ABC}	38,09^{AB}	34,59^{ABC}	29,40^{C}	41,24^{A}	31,98^{BC}	0,0086	0,0001	0,2025
27-32 weeks	58,98^{B}	58,68^{B}	59,45^{A}	59,79^{A}	58,99^{B}	59,81^{A}	58,73^{B}	0,0001	0,0001	0,8628
Egg mass (g/heads)										
22-26 weeks	10,62^{a}	10,61^{a}	12,80^{a}	11,25^{a}	11,13^{a}	12,39^{a}	9,69^{a}	0,6296	0,0001	0,9107
27-32 weeks	52,09^{ab}	52,92^{ab}	50,52^{b}	53,11^{a}	51,65^{ab}	51,79^{ab}	50,70^{b}	0,0514	0,0001	0,1547
Feed conversion ratio										
22-26 weeks	11,77^{a}	17,61^{a}	18,38^{a}	23,56^{a}	22,88^{a}	10,27^{a}	21,56^{a}	0,3397	0,0001	0,0134
27-32 weeks	2,01^{a}	1,98^{a}	2,08^{a}	1,95^{a}	2,02^{a}	2,02^{a}	2,08^{a}	0,2894	0,0001	0,8860

Test results (Table 2) showed that at 22-26 weeks of age there were differences between the feed intake of T1 with other treatments. This is because the chickens are treated T1 (100%) in the morning not having a high heat load, so the chicken will eat plenty to meet nutrient needs. Chicken with T7 treatment had the lowest levels of intake. This is due to the chicken have not been able to adapt to changes in the feeding portion. While at the age of 27-32 weeks, feed intake T1 was not significantly different with the feed intake of T6 and T7. Chickens did not get diet in the morning (T7) apparently had the same level of intake of chicken fed with rations 100% in the morning. This is because the chickens (T7) was able to adapt to try to meet the need of nutrients through a lot of eating in the afternoon. This is supported by environmental temperature begins to decline so that the chicken did not have an excessive heat load that blocks consumed diets. Age was highly significant (P <0.01) effect on feed intake at the age of 22-26 weeks and at the age of 27-32 weeks. Age would be accompanied by an increase in the feed intake because the rate of egg production also increased.

Hen Day Production

There was no interaction between feeding portion and the age of HDP (Table 2). This showed the influence of relatively equal treatment to all ages. Feeding portion did not affect significantly (P> 0,05) to HDP at the age of 22-26 weeks but otherwise at the age of 27-32 weeks (P <0,05). No treatment effect on HDP at the age of 22-26 weeks due to laying hen had not entered the peak phase of production so that the treatment had no effect on egg production. While the chicken at the age of 27-32 weeks of entering the peak of production in which the number of eggs produced more than the age of 22-26 weeks so that the treatment effect on egg production.

Some studies showed no effect on feeding time to HDP (Bootwalla *et al.*, 1983, Samara *et al.*, 1996 and Backhouse and Gous, 2005) while another study showed a decrease in chickens given a ration HDP if only in the afternoon (Avilla *et al.*, 2003). Avilla *et al.* (2003) showed that egg production of chickens given a diet of 100% at 11.00 a.m. was lower when compared with chickens given a diet of 100% at 06.30 a.m. and chickens treated 50% at 06.30 and 50% in at 03.30 p.m. This is because the heat load in chickens fed diet 100% at 11.00 was higher than other treatments due to heat stress.

Egg Weight

There was no interaction between treatment and age in influencing the weight of the egg. This indicated the feeding portion has the same effect on all ages. Feeding portion influenced egg weight (P> 0.01) both at the age of 22-26 weeks and 27-32 weeks. Feeding portion influenced egg weight at the age of 22-26 weeks, although HDP analysis showed a nonsignificant result. This is because the weight of the egg is the first response will be accepted if there was a decrease in feed intake (Sloan and Harms, 1984, cited by Bird *et al.*, 1996). Egg weight at the age of 22-26 weeks was much influenced by the level of consumption. The higher feed intake will produce the higher the egg weight (Amrullah, 2003). Backhouse and Gous (2006) states there was a trend of increasing egg weight in chickens given a diet more in the afternoon or with a portion more than once per day. The chickens fed diet once a day in the morning or afternoon course will result in lower egg weights.

Egg mass

There was no interaction between treatment and age in influencing the egg mass. This showed the influence of relatively equal treatment to all ages. No significant effect (P> 0.05) on egg mass both at the age of 22-26 weeks and at the age of 27-32 weeks. This is because the rate of increase between the HDP is not equal to the rate of increase in the egg weight. Egg mass is a multiplication of HDP with mean weight of eggs produced on a particular day (Amrullah, 2003) so that the egg mass is a combination effect of HDP and weight of eggs.

Shell thickness

Feeding portion effect on eggshell thickness (P <0.05) at 22-26 weeks but not at 27-32 weeks (Table 2). Shell thickness at the age of 26 weeks of which are influenced by the size of the egg and feeding portion. The smaller eggs tend to have a thicker shell (T4, T5 and T7) and the larger eggs will reduce the size of its thickness due to the same amount of calcium used to form a shell with a larger area (T1 and T6). Jejunum which serves as the absorption of calcium has a constant permeability of calcium during egg formation cycle. According to Waldroup and Hellwig (2000) increased absorption of calcium during eggshell formation is not due to increased mucosal capacity. Chickens treated with more servings during in the afternoon will likely have a thicker shell (T2, T4 and T7). It is associated with a time of egg shell formation that occurs in the afternoon. Use of calcium derived from the ration in the morning is very low due to the inability of laying hen to take calcium from the crop into digestive tract. Feeding in the afternoon or evening approached the period of shell deposition will improve the use of calcium which would then be manifested in shell eggs (Farmer *et al.*, 1983; Moreki, 2005). This is because the process of eggshell formation occurs at night.

Feeding portion have no effect on eggshell thickness at 27-32 weeks of age although egg weight parameters at the same age show an effect. This is because the difference in length of egg formation. The length of stay in the oviduct will affect the thickness of the shell. The chickens that produce higher eggs weight (T3, T4, and T6) tended to last longer in the oviduct compared to the chickens that produce eggs lower weight (T1, T2, T5 and T7) (Indreswari, 2007). This event will increase the amount of secretion of calcium for the eggshell in larger eggs and therefore at the age of 32 weeks there was no treatment effect on eggshell thickness.

Egg shell is thicker at 32 weeks than 26 weeks. The ability of calcium absorption was higher in the age of 32 weeks. The opposite was reported Al Batshan *et al.* (2004) that increasing age, eggshell thickness will decrease. Old chickens have the same ability or better in producing shell than young chickens (Rolland *et al.*, 1978).

Feed conversion ratio

The results showed the interaction between feeding portion and age in influencing FCR at age 22-26 weeks (Table 2). This shows feeding portion has different effects at each age. In contrast to the age of 27-32 weeks there was no interaction effect. There was no interaction between feeding portion and age. Feeding portion have no significant effect (P> 0.05) on the FCR. This is due to the lack of effect of treatment of egg mass in the second period. Age has very significant (P <0.01) on FCR. Increasing age will further lower the value of FCR. This is due to increasing with age, the ability of nutrient use will increase.

Conclusion

Chickens were fed with feeding portion 70:30% at 22-26 weeks have the highest egg weight compared to other treatments while the HDP does not seem to be influenced by the feeding portion because at that period still growing chickens in order to achieve optimal weight. Chickens get feeding portion 50:50% at the peak of production, at the age of 27-32 weeks is the best, especially in sufficient calcium for eggshell formation.

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The Effect of Kombucha Supplementation in The Ration on Quails Body Weight and Dressed Carcass Weight

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Abstract

Hundred quails were used in this experiment to study the effects of Kombucha supplementation in the ration on quails body weight and dressed carcass weight, were studied for five weeks. This research used a Completely Randomized Design (CRD) with six treatments of Kombucha tea (0, 10, 15, 20, 25 and 30%), repeated four times. The ration treatments were: P-0 basal diet as control, P-1 basal diet + 10% kombucha; P-2 basal diet + 15% kombucha; P-3 basal diet + 20% kombucha; P-4 basal diet + 25% kombucha; and P-5 basal diet + 30% kombucha. Results indicated that the highest body weight (163.1 g) was get from the quails that fed basal diet + 25% kombucha (P-4) and the quails that fed only basal diet (P-0) has the lowest body weight (146.475 g). For dressed carcass, quails that fed basal diet + 30% kombucha (P-5) has the lowest carcass weight (116.85 g), and the highest carcass weight (141.125 g) are the quails that fed basal diet + 10% kombucha.

Keywords: body weight, dressed carcass, kombucha, quails, supplementation

Introduction

Quail is a collective name for several genera of mid-sized birds in the pheasant family Phasianidae, or in the family Odontophoridae. The quails are small, plump terrestrial birds. They are seed eaters, but will also take insects and similar small prey. They nest on the ground. They are capable of short, rapid bursts of flight. Some species, including the Japanese and Common Quail, are migratory and fly for long distances. Some quail are farmed in large numbers. These include Japanese Quail, also commonly known as coturnix quail, which are mostly kept to produce eggs that are sold worldwide. Japanese Quail (*Coturnix coturnix japonica)* endemic to East Asia, abundant across most of its range. They migrate to Manchuria, southeastern Siberia, and northern Japan, and winter in southern Japan, the Korean Peninsula, and southern China. Their preferred habitats are grasslands and cultivated fields. The plumage is a speckled yellow-brown, with a creamy white strip above the eye. Adults are approximately 20 centimeters in length. The quail is a small bird that inhabits woodland and forest areas around the world. There are thought to be more than 15 different species of quail, with each species of quail being found in different parts of the world and all have slightly different appearances depending on how they have adapted to their environment. Although the quail is very small sized bird, the quail belongs to the same bird family as pheasants. Quails range in size depending on the species from the Japanese quail which is around 10cm tall to the larger mountain quail that can grow up to 25 cm tall. Quails are generally solitary birds and spend most of their time either on their own or in a pair with just one other quail. During the mating season it is common to see large flocks of quails as family groups convoy together in groups of up to 100 quail individuals. Quails do not tend to migrate and therefore spend their lives within the same area. In some parts of the world, quails are kept as poultry birds both for the small amount of meat that they contain and for the quail's brightly coloured eggs. These tiny coloured

eggs are seen as a delicacy in some parts of the world and can often be found on menus in posh restaurants. When quails reach 2 months old, they are then able to mate. Quails tend to breed in more open areas such as farmland and lay their eggs in nests. Quail clutch sizes can vary between one and 12 eggs depending on the species of quail and the baby quail chicks hatch out of their eggs in less than a month.

Feeding Quails

In recent years quail have also been successfully reared by methods similar to those applied to chicks. Feed ingredients are similar, but the amounts are adjusted to meet the nutritive requirements of the species (Heuser, 2003). Feeding to add flesh at a rapid rate, must consider the amount of protein that is needed because without that the birds will not fatten up quickly (Batty, 2005). However, high protein food, usually special pellets or turkey crumbs; must be fed all the time on an ad lib basis. The adults eat layers pellets finch food, chick grit, mealworms and fresh greens. Rear them on either chick crumbs, which are 20- 22 percent protein, or turkey starter crumbs, which are 26-29 per cent protein. Nutrients requirements of Japanese quail (*Coturnix coturnix japonica)* for breeding, energy base 1.361 kcal ME/lb. The metabolic rate of birds will be affected by the type of diet (Ensminger, 1990). Put pebbles in the water dish so that the young quail, which resemble bumblebees. If the chicks are adult-reared the parents will brood them if the aviary is large it will make sense to confine the birds in part of the floor area until they fly at about three weeks of age, so that they don't become separated from their parents. At this age you can begin diluting their crumbs with cheaper and less nutritious food. If the chicks are hatched in an incubator you will need to offer them heat, such as a brooder or infrared light, for the first couple of weeks. Japanese quail mature in about 6 weeks and are usually in full egg production by 50 days of age. With proper care, hens should lay 200 eggs in their first year of lay. Life expectancy is only 2 to 2½ years.

Body Weight of Japanese Quails

If the birds have not been subjected to genetic selection for bodyweight, the adult male quail will weigh about 100–140 g, while the females are slightly heavier, from 120–160 g. They fatten readily with a high conversion rate for the food eaten (Batty, 2005). The Japanese quail is a fast growing hardy bird. They are mature at around 6 weeks of age and are laying eggs by around 7-8 weeks of age. Males are characterized by a rusty brown throat and breast feathers while the hens have a lighter cream colored feathering on the neck with black stripes and dotting on the breast. The primary reasons for keeping quails, are for meat, eggs and as pet (hobby). The meat taste very delicious. Quail is small birds with plump breasts and a mild flavour. The hen is plump and tender, and the cock is almost as good, with a spectacular green neck and long speckled tail feathers. While most other types of fowl require a farmyard setting with plenty of room, even city dwellers are able to raise these small birds if their zoning requirements permit.

Kombucha Supplementation

Kombucha is fermented tea that is often drunk for medical purposes. There are scientific studies that support the health benefits of Kombucha as antimicrobial (Sreeramulu, *et al*., 2000; Cetojevic-Simin *et al*, 2008). Kombucha contains multiple species of yeast and bacteria, as well as the organic acids, active enzymes, amino acids, and polyphenols produced by those microbes. Yeast ferments contained in kombucha tea are *Candida albicans, Saccharomyces sp.*, and *Pichia fermentans* while the bacteria are *Acetobacter*

xylinium, Gluconicum bacteria, and *Acetobacter ketogenum* (Williams, 2001). According to Akhadianto (2009), Kombucha fermented tea, has no negative effect, when supplemented in broiler ration. To carcass weight, it more 2% compared to the broiler with no kombucha fermented tea in the ration. Even there is no significancy on the liver, gizard and hearth, and the abdominal fat. Various compounds have been added to livestock and poultry diets to increase the efficiency of food utilization. Most of these additives do not supply nutrients although they effect food utilization in some species. Besides some organic acids, Kombucha tea fermentation, has vitamin B1, B2, B3, B6 and B12 which effective in metabolism process of some food ingredients (Frank, 1995).

Results And Discussions

1. The Effect of Kombucha Supplementation in Ration on Body Weight

In Table 1, presents the body weight of quails that fed with Kombucha supplemented ration.

Table 1. The body weight of quails that fed Kombucha supplementation in ration (g)

Replication	R-0	R-1	R-2	R-3	R-4	R-5
I	155.5	164.4	167.4	164.7	156.2	155.6
II	153.6	173.5	164.2	166.5	161.2	154.1
III	130.3	156.0	161.9	152.7	154.5	162.1
IV	146.5	158.5	148.0	159.5	162.9	163.1
Average	146.475	163.1	160.375	160.85	158.7	158.725

From Table 1, the average body weight ranged between 163.7 g – 146.475 g; and the quail that fed normal ration (R-0) that has not supplemented with Kombucha are the lightest (146.475 g). These results showed that the Kombucha supplemented groups improved the body weight than the control groups, that feed no Kombucha tea fermentation supplementation in the diet. It means that the beneficial effect of kombucha tea on ration. The improvement in the body weight in this study may be due to the increased efficiency of digestion and nutrient absorbtion processed due to precence of the kombucha tea. As a consequence, there is an improvement in the intestinal environment, increasing the efficiency of digestion and nutrient absorption processes. The adult male quail usually will weigh about 100–140 g, while the females are slightly heavier, weighing from 120–160 g. Combucha tea fermentation has useful medical effect to intestine and also anti-bacterial effect as asetic acid, glucoronic acid, gluconic acid, citric acid, oxalic acid, lactic acid and butiric acid (Williams, 2001). According to Akhadianto (2009), Kombucha fermented tea, has no negative effect, when supplemented in broiler ration. To carcass weight, it give more 2%; compared to the broiler with no kombucha fermented tea in the ration.

2. The Effect of Kombucha Supplementation in Ration on Dressed Carcass Weight

In Table 2, there is data of Dressed Carcass Weight of quails that fed Kombucha Supplementation in ration. From Table 2, the average dressed carcass of quails are between 116.85 g – 141.125 g, and the highest dressed carcass weight is the quail that fed with basal diet +10% kombucha (R-1); and the lightest dressed carcass weight is the quail that fed with basal diet +30% kombucha (R-5). The dressed carcass, are light because many fat in the abdominal and the fat in quail meat are smallest, and then they have smallest carcass

weight. Organ percentage in the quails that fed Kombucha supplementation, even there is no significancy, but has more abdominal fat (Akhadiarto, 2009). So, the dressed carcass weight more lighter, because some fat was thrown when dressing carcass.

Table 2. The Dressed Carcass weight of quails that fed Kombucha supplementation in ration (g)

Replication	R-0	R-1	R-2	R-3	R-4	R-5
I	144.1	146.4	155.9	133.6	135.6	128.0
II	134.7	144.1	135.3	123.3	110.5	118.3
III	125.4	149.8	131.9	129.1	109.5	105.9
IV	134.8	124.2	139.5	133.1	134.9	115.2
Average	134.75	141.125	140.65	129.775	122.625	116.85

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Occurrence of Pork Derivative in Confectionery Product Upon Malaysia Market

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Abstract

Adding porcine ingredients in food manufacturing without labeling can considered as food adulteration. Thus, molecular technique especially PCR based method has been proposed as useful means for identifying species of origin in foods. The present study was carried out for detection of porcine ingredients in different food samples namely from raw to highly processed food using species specific PCR detection of conserved region in the mitochondrial (mt) 12S ribosomal RNA (rRNA) gene.

Keywords: 12S rRNA, Confectionery product, Malaysia, , PCR, Pork

Introduction

Every ounce of pig can be use to make an astonishing 185 products. A frequent adulteration of meat products is the addition of pork (*Sus scrofa*) to beef (*Bos taurus*) products for economic gain (Mafra *et al*., 2008). In addition, fail to label can remarkably give negative consequences. A number of approaches had been used to identify porcine ingredients in processed food. DNA molecules have been the target compound for species identification due to the high stability compared to proteins and it presence in most biological tissues regardless of tissue origin. Some of the PCR method that had been used to detect the presence of pork DNA in food mixtures are quantitative competitive PCR (QC-PCR), species specific PCR using 12S rRNA and cytochrome b, PCR-RFLP, multiplex PCR and real-time PCR with TaqMan probes (Che Man *et al*., 2007).The present study is aimed to identify pork ingredients at molecular level using species specific PCR based on mitochondria 12S rRNA gene with the following objectives:

(a) To evaluate the effect of different food matrices in terms of quality of the extracted DNA.
(b) Screening commercial food samples using selected DNA extraction method.

Materials and Methods

DNA extraction CTAB 2 method (Mafra *et al*., 2008)

0.1 g samples were mixed with 1 ml CTAB buffer [20g CTAB/l, 1.4 M NaCl, 0.1 M Tris-HCl, 20mM EDTA] and 70 µl proteinase K and incubated at 65^{o}C for one hour. The samples were then centrifuged for 15 minutes at 14 000 rpm. The supernatant was transferred to a new 1.5 ml tube extracted with 400 µl chloroform and centrifuged for 10 minutes at 12 000 rpm. The upper phase was transferred into a new 1.5 ml tube and double volume of CTAB was

added. Then, the mixture was incubated for one hour and later centrifuged for 10 minutes at 12 000 rpm.

Precipitation was added with 350 µl 1.4 M NaCl and 350 µl chloroform. Again, centrifuged for 10 minutes at 12 000 rpm. The upper phase was transferred into a new 1.5 ml tube and added 600 µl isopropanol. Extracts were then mixed by inverting tubes 50 times and placed in a freezer (-20°C) for 30 minutes. The supernatant was discarded and the pellet was washed once with 500 µl of 70% ethanol and air dried for approximately 30 minutes. The pellet was dissolved in 70 µl of TNE buffer and stored at -20 ^{o}C until use.

DNA extraction CTAB 1 method (Tung N. *et al.,* 2009)

Approximately 0.1g of minced samples was transferred into a plastic test tube containing 5ml of CTAB buffer (20g CTAB/L, 1.4 NaCl, 0.1 M Tris HCl, 20 mM EDTA). The samples were then incubated in the water bath at 65°C for overnight. An aliquot (1 ml) was transferred to 1.5 ml eppendorf tube, centrifuged it at 13000 rpm for 10 minutes. Then, 400 µl of supernatant was taken to a new appendorf tube and added with 300 µl of chloroform. The mixture was mixed gently to prevent DNA from destroyed.

Then, the mixture was centrifuged for 10 minutes at 13000 rpm. After that, 300µl of upper layer was removed to a new eppendorf tube and added with 600 µl of absolute isopropanol. The sample need to be cooled at 4°C for 10 minutes followed with centrifuged for 10 minutes at 12000 rpm. The supernatant was discarded and 600 µl of ethanol was added and centrifuged again at 10000 rpm for 5 minutes. Discarded the supernatant and the pellet was dried at room temperature. After that, added 70µl of distilled water or TE buffer to dilute the DNA.

DNA extraction triomic kit (Ecocillus (M) Sdn. Bhd., 2009)

Isolation of DNA was performed based on the manufacturer's instruction. Approximately 0.1 g of samples was weighted into sterile 1.5 ml appendorf tube containing 400 µl of R1 solution and vortex for 30 seconds. The tubes were incubated at 65 ^{o}C in the water bath for 15 minutes. The tubes were mixed by gently flicking every 15 minutes. Then, 800 µl of R2 solution was added to the tubes and inverted it gently. Afterwards, centrifuged at 9000 rpm for about 10 minutes. 600 µl of the supernatant containing DNA was transferred into a new appendorf tube. A volume of 800 µl of R3 solution was added and inverted slowly for 30 seconds and centrifuged the supernatant at 9000 rpm for 10 minutes. The DNA pellet was dried at 37 ^{o}C for 20 minutes and resuspend with 70 µl of sterile TE buffer.

Gel electrophoresis of genomic products

The DNA integrity including degraded and contaminants can also be evaluated by electrophoresis of genomics. The isolates DNA were checked by Agarose gel. 1 % Agarose was prepared in 1x TAE buffer.

Polymerase chain reaction (PCR)

Table 1. Sequence of primer used to amplify pork specific gene

Primer	Sequences	Genes Amplified	PCR Product
HAL Primer (+)	5'- CCA CCT AGA GGA GCC TGT TCT ATA AT – 3'	Pork	387 bp
HAL Primer (-)	3'- GTT ACG ACT TGT CTC TTC GTG CA – 5'	Specific	

(Source: Rodriguez *et al.*, 2004)

Gel Electrophoresis of Amplified PCR Products

The PCR amplification products were observed using agarose gel electrophoresis. A 2 % Agarose -TBE gel was made.

Results and Discussion

Genomic Electrophoresis

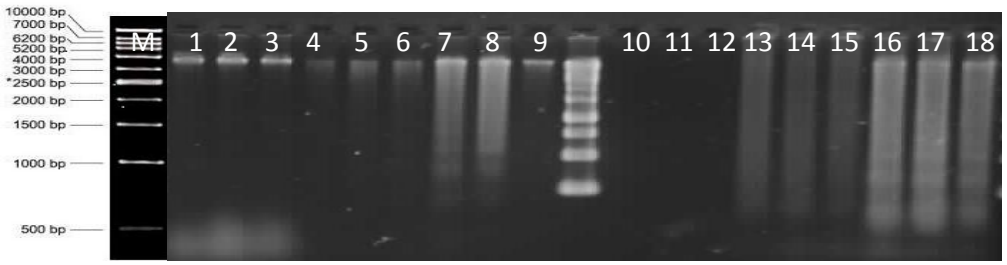

Figure 1. Agarose gel electrophoresis (1%) of total DNA from six categories of positive pork samples samples from the CTAB 2 method.

Note:
Lane M : 1 kb DNA marker (Vivantis Co.);
Lane 1-3 : Pork meat;
Lane 4-6 : Pork sausage;
Lane 7-9 : Lard;
Lane 10-12 : Pork oil;
Lane 13-15 : Biscuit contain oil;
Lane 16-18 : Biscuit contain lard

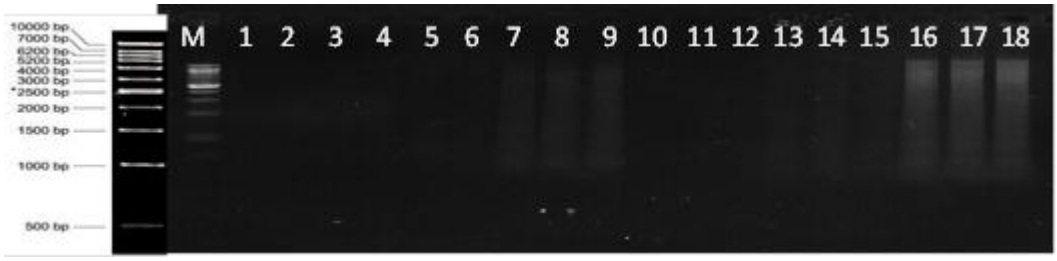

Figure 2. Agarose gel electrophoresis (1%) of total DNA from six categories of samples from the CTAB 1 method.

Note:
Lane M : 1 kb DNA marker (Vivantis Co.);
Lane 1-3 : Pork meat;
Lane 4-6 : Pork sausage;
Lane 7-9 : Lard;
Lane 10-12 : Pork oil;
Lane 13-15 : Biscuit contain oil;
Lane 16-18 : Biscuit contain lard

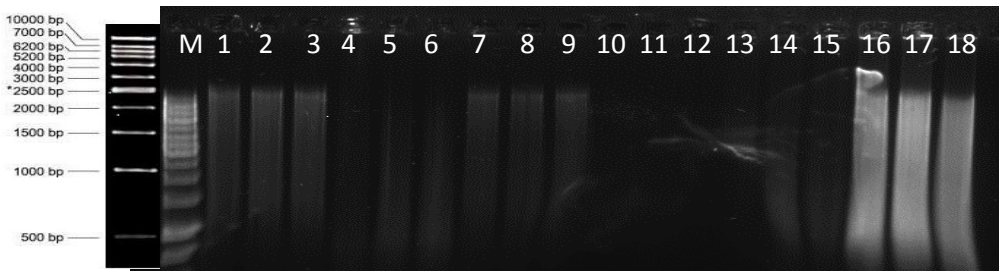

Figure 3. Agarose gel electrophoresis (1%) of total DNA from six categories of samples from the Triomic method.

Note:
Lane M : 1 kb DNA marker (Vivantis Co.);
Lane 1-3 : Pork meat;
Lane 4-6 : Pork sausage;
Lane 7-9 : Lard;
Lane 10-12 : Pork oil;
Lane 13-15 : Biscuit contain oil;
Lane 16-18 : Biscuit contain lard

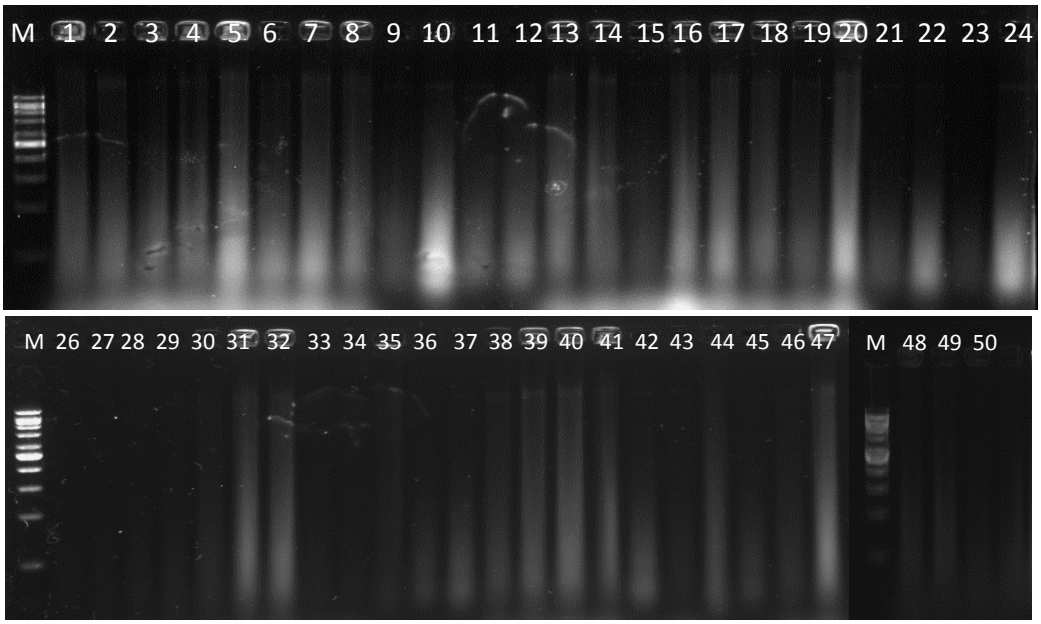

Figure 4. Agarose gel electrophoresis (1%) of DNA extraction from different commercial brand biscuits using CTAB 2 method.

Note :
Lane M : 1 kb DNA marker (Vivantis Co.);
Lane 1-50 : Commercial biscuits from different brands.

Polymerase Chain Reaction (PCR) Amplification

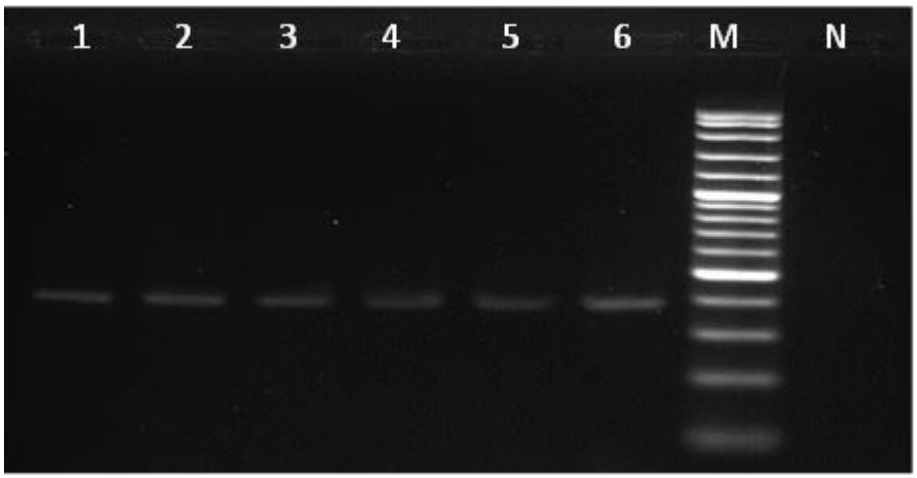

Figure 5. Agarose gel electrophoresis (1.8%) of porcine PCR product using CTAB 2 method.

Note :
Lane M : 100bp DNA marker (Vivantis Co.);
Lane N : Negative control ;
Lane 1 : Pork meat
Lane 2 : Pork sausage
Lane 3 : Pork lard
Lane 4 : Pork oil ;
Lane 5 : Biscuit contain lard
Lane 6 : Biscuit contain oil

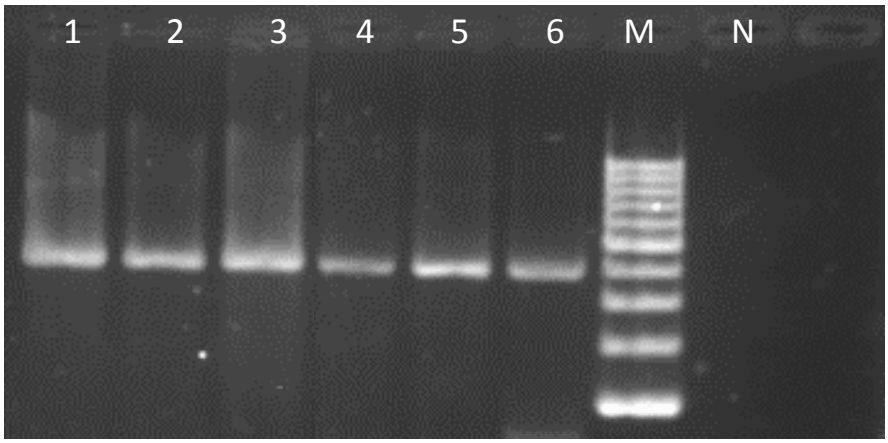

Figure 6. Agarose gel electrophoresis (1.8%) of porcine PCR product using CTAB 1 method.

Note :
Lane M : 100bp DNA marker (Vivantis Co.);
Lane N : Negative control ;
Lane 1 : Pork meat
Lane 2 : Pork sausage
Lane 3 : Pork lard
Lane 4 : Pork oil ;
Lane 5 : Biscuit contain lard
Lane 6 : Biscuit contain oil

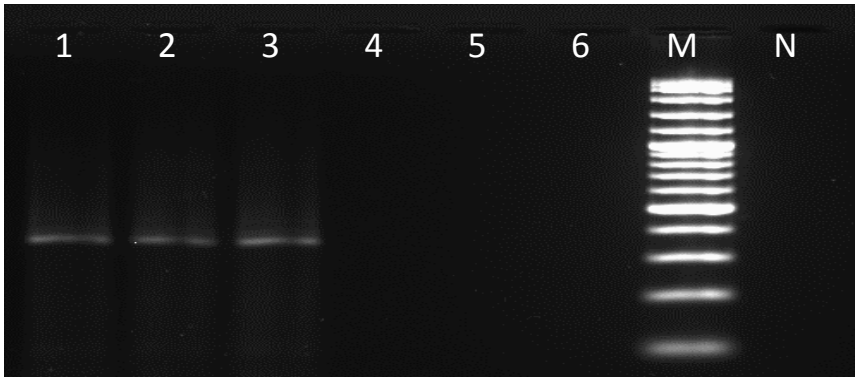

Figure 7. Agarose gel electrophoresis (1.8%) of porcine PCR product using Triomic method.

Note :
Lane M : 100bp DNA marker (Vivantis Co.);
Lane N : Negative control ;
Lane 1 : Pork meat
Lane 2 : Pork sausage
Lane 3 : Pork lard
Lane 4 : Pork oil ;
Lane 5 : Biscuit contain lard
Lane 6 : Biscuit contain oil

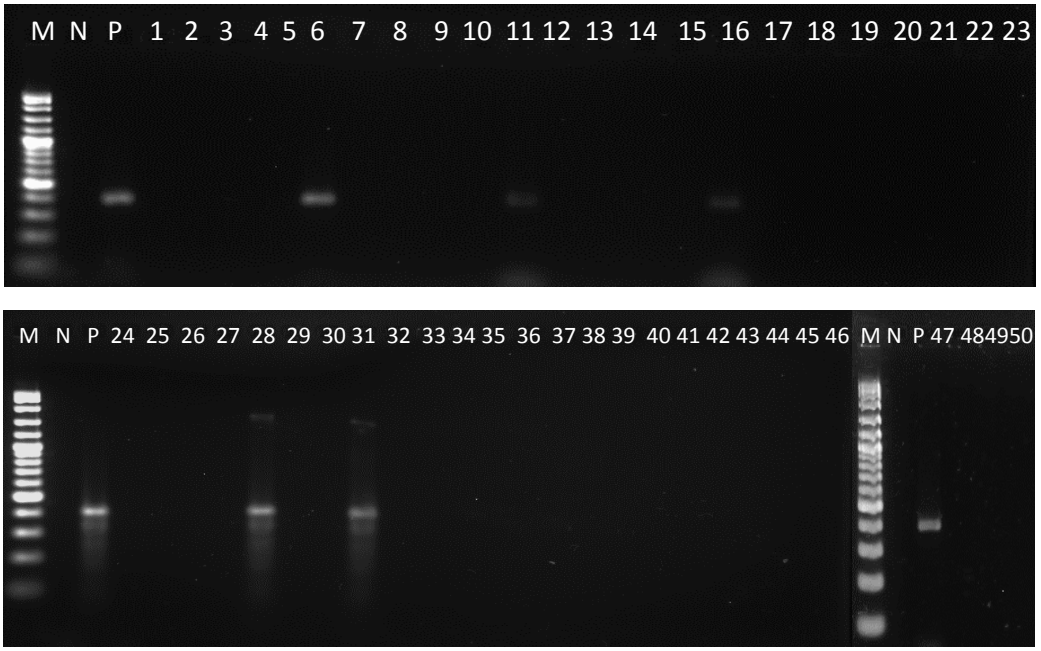

Figure 9. Agarose gel electrophoresis (1.8%) of 12S rRNA PCR products amplified from different commercial brand biscuits.

Note :
Lane M : 100bp DNA marker (Vivantis Co.);
Lane P : Different brand of sausages;
Lane M : Pork meat;
Lane S: Pork sausage

Conclusion

Since PCR entail high quality DNA template, the method used for the extraction of DNA is very critically importance. DNA extraction method CTAB 2 extraction DNA had shown more promising result in PCR and genomic amplification. All the bands in PCR were very clear as well as genomic electrophoresis. Out of 100 samples, 5 biscuits were detected with positive porcine ingredient.

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Nutritional Contents of Gecko's Flesh (*Gekko spp*)

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Abstract

Recently, gecko's flesh used as a raw material in alternative medicine. Consumer believes that gecko's flesh has high potency in treatment. The composition of gecko's flesh has never been published, therefore this study was conducted to identify the nutritional content of gecko's flesh, which includes the content of carbohydrates, protein, fat, crude fiber, water and ash. One hundred geckos without differentiated sex were slaughtered and skinned. Gecko's flesh samples were obtained by flesh separation technique from the bone and then put into plastic and analyzed. Analysis of nutrient content was conducted at the Laboratory of Biotechnology, Faculty of Agricultural Technology, Universitas Gadjah Mada, Yogyakarta. Proximate analysis for nutrient contents were conducted by applying micro-Kjeldahl method (protein), Soxhlet (fat), by difference (carbohydrates), acid-base (crude fiber), thermogravimetri (water), and muffle furnace (ash). The results showed that gecko's flesh carbohydrate (3.21±2.10%) and protein (21,42±2,20%) were higher than meat of cattle, buffalo, goat, chicken, and duck; and its fat content was 1.28± 0.28%, which is lower than those livestock's meat. This study also revealed that crude fiber (0.45±0.13%) and ash (1.46±1.37%) in gecko flesh were similar with those of livestock's meat.

Keywords : Gecko's flesh, nutrient content.

Introduction

Meat is a product with high nutritional content that has a potential for growing and developing the body. Meat is a source of energy, protein, vitamins and minerals. Meat consumption in Indonesia mostly derived from poultry because of its lower price than other livestock meat. There are other animals that produce meat, even though unusual, those are gecko, dog, snake, and bat flesh. Those meats are grouped as exotic meat. Survey indicated that people consumed those exotic meats for therapeutic purposes. The aim of this study was to find out the nutritional content of gecko's flesh and compared with other livestock's meat. Nutritional assessment of gecko's flesh consists of carbohydrates, ash, protein, fat, minerals and water.

Proximate analysis is a method of chemical analysis to identify nutrient content of a food or food ingredient (AOAC, 1970). This method was developed by Henneberg and Stokman in Weende Experiment Station in Germany at 1865. Proximate analysis comprises of tests for level of carbohydrates, protein, fat, moisture, ash and crude fiber.

Materials and Methods

Materials

The tools used for sampling meat geckos were knives, masks, gloves, weights, ice box, plastic trays, plastic bags, etiquette. The tools used in the analysis of water content were analytic scales, weighing bottle, spoon, oven, desiccators, and clamp. The tools used in the analysis of fat content were Soxhlet extraction tube, thimble, Soxhlet distillation equipment, electric stove, measuring cups, oven, desiccators, and analytical scales. The tools used in the analysis of total protein were analytic scales, filter paper, Kjeldahl flask, one and 10 ml measuring pipette, electric stove, hood, destilator unit (kjeltec), Erlenmeyer tubes, burette, stative, funnel, measuring cup, glass beaker, and the spoon / spatula. The tools used in the analysis of crude fiber content were analytic scales, electric stove, oven, measuring cups, Erlenmeyer tubes, filter paper, spatula/spoon, and desiccators. The tools used in the analysis of ash content were a porcelain crucible, electric stove, eksicator, oven, clamp, analytical balance, Muffle Furnace, ladle, and the smoke chamber.

One hundred geckos obtained from the Pasty Market, Dongkelan, Bantul, Yogyakarta without differentiated sex was used in this study. Materials used in the analysis of water content and the ash are gecko's flesh samples. Materials used in the analysis of meat fat content were gecko's flesh, and petroleum ether. Materials used in the analysis of total protein were gecko's flesh, a catalyst (HgO and K_2SO_4), a solution of concentrated H_2SO_4, distilled water, NaOH-$Na_2S_2O_3$, 4% boric acid solution, BCG-MR, and a solution of HCl 0.02 N. Materials used after in the analysis of crude fiber content were the meat samples gecko, 1.25% H_2SO_4 solution, 1.25% NaOH solution , distilled water, and 95% alcohol.

Methods

Research methods included three phases: preparation, research, data collection and processing. Preparation phase of research include the purchase of geckos, slaughtering and meat sample selection. Phase of research included the analysis of nutritional content of gecko meat samples consisted of 10 tails gecko's flesh. Given the weight of meat on each tail geckos were too small, each samples consisted of 10 tails gecko's flesh. The last phase was data collection and processing.

Sample selection was done by selecting a gecko which has a great body. Each samples consisted of 10 geckos. Samples that have been selected will then be slaughtered using a sharp knife. The sampling of meat by separating the flesh from its bones and put into plastics bag and weighed. Analysis on the nutritional content of meat gecko was performed at the Laboratory of Biotechnology, Faculty of Agricultural Technology, UGM, Yogyakarta.

Water content analysis (AOAC, 1970)

Determination of water content of samples based on the weight difference of samples before and after drying. Porcelain saucer to be used, was dried about an hour at a temperature of 105^{o}C, and then was cooled in a desiccator for 30 minutes and weighed until the weight remains (A). The sample was weighed about two grams (B) in the pot, dried in an oven at 100-105^{o}C for five hours or fixed weight. The cup containing the sample was cooled in a desiccator for 30 minutes and then weighed until the weight remains (C). Water content was calculated by the formula:

$$\text{Water content (\%)} = \frac{(A+B) - C}{B} \times 100\ \%$$

Ash content analysis

Determination of ash content based weighing the residual minerals as a result of burning organic material at a temperature of about 550^{o}C. Porcelain saucer was dried in an oven dive one hour at 105^{o}C temperature, then was cooled 30 minutes immersion in a desiccator and was weighed to get heavy equipment (A).Samples were weighed as much as two grams (B), then were inserted into a porcelain cup and were crossed over the flame until no longer smoky. Then it was inserted inserted into an electric arc furnace (furnace) with a temperature of 650^{o}C for ± 12 hours. Furthermore, the cup was cooled for 30 minutes in the desiccator, and then was weighed to get heavy equipment (C). The ash content was calculated using the formula:

$$\text{Ash content (\%)} = \frac{(A+B) - A}{B} \times 100\ \%$$

Fat content analysis by Soxhlet method (Woodman, 1941)

The flask that has been dried in an oven (105^{o}C) were weighed to get heavy equipment (A). A total of two grams of sample (C) was wrapped in fat-free filter paper and then inserted into the fatty sheath. Sleeve was inserted into the Soxhlet tube. A total of 150 ml of chloroform was introduced into the flask fat. Samples were refluxed for eight hours; after the solvent has been shown clearly indicate all the fat has been extracted. Furthermore, the existing solvent in the flask was evaporated to separate fat and fat solvents, and then the flask was dried in an oven 105^{o}C for 30 minutes.

Furthermore it was weighed to get heavy equipment (B). Fat content calculated by the formula:

$$\text{Fat content (\%)} = \frac{(B - A)}{C} \times 100\ \%$$

Analysis of total protein levels with Mikrokjeldahl method (AOAC, 1970)

Determination of protein content was done by micro-Kjeldahl method. A sample of 0.75 g was inserted into the Kjeldahl flask, and then 6.25 gram of K_2SO_4 and 0.6225 grams of $CuSO_4$ as a catalyst were added. A total of 15 ml and 3 ml of Concentrated H_2SO_4.H_2O_2 solution were added slowly into the flask and allowed to stand for 10 minutes in the hood. The next stage was the destruction process at a temperature of 410^{o}C for two hours or until a clear solution was obtained, Furthermore it was allowed to stand until it reaches room temperature and and then 50-75 ml of distilled water were added into the flask.

An Erlenmeyer was prepared by added 25 ml of 4% H3BO3 solution. This solution containing an indicator that composed from green bromochresol 0.1% and 0.1% methyl red (2:1) and were used as a reservoir distillate. Flasks Kjeldahl distillation equipment was mounted on a series of steam. After that 50 ml of Na2 (SO4)3 (alkaline) added to the Erlenmeyer, then were distilled and the distillated products were collected in Erlenmeyer until the volume reached 150 ml and showed a green-colored distillate. Distillate titrated with 0.2 N HCl, made up the color changes to gray naturally. The distillated products then were used as sample of blank stage. Protein tests were carried out duplicate samples. Protein content was determined by the formula:

$$\text{Protein content (\%)} = \frac{(AB) \times \text{Normality of HCl} \times 14.007 \times 6.2 \times 100\%}{W\ (g)}$$

Description: A = ml titration of HCl sample
B = ml HCl blank titration

Crude fiber analysis

A total of two grams of dry material that has been extracted fat were inserted into into the tube Erlenmeyer. The material was added into the Erlenmeyer flask containing 1.25% H2SO4 solution and then was boiled for 30 minutes. The suspension then was filtered using filtered paper, the residue left in the Erlenmeyer was washed using boiled aquadest. Residues contained in the filter paper then were washed until the washing water no longer acidic. Residues of litmus paper and then were transferred into an erlenmeyer and furthermore 200 ml of 1.25% NaOH were added. After that it were boiled again for 30 minutes and were filtered with filter paper that has been constant and the residue was washed using 95% alcohol or less as much as 15 ml. Filter paper and the residue then was dried and weighed until reached constant weight.. Calculation of residual weight is the weight of residual crude fiber.

Carbohydrate content analysis

It was done by calculating the residual (by difference):

The level of carbohydrates (%) = 100% - (moisture content+protein+fat+ash+crudefiber).

Data Collection and Analyzed

The test results of the nutrient content of meat involved protein, fat, carbohydrates, water, ash, and crude fiber were recorded and analyzed in statistic descriptively.

Results and Discussion

Table 1. Average Nutritional Value of Gecko's Flesh in 100 Gram (%)

Component	Nutrient Content (%)
Carbohydrates	3.21±2.10
Protein	21.42±2.20
Fats	1.28±0.28
Crude Fiber	0.45±0.13
Water	72.63±1.78
Ash	1.46±1.37

The results obtained is presented in Table 1. The carbohydrate level of meat gecko was higher than other livestock's meat. Carbohydrate content of gecko's flesh was 3.21±2.1%. It was higher than other livestock's meat content, which approximately 0.5-1.0% (Syamsir, 2010). There is no carbohydrate content (0%) in beef and meat of buffalo, sheep, chicken, or duck (Muchtadi, 1989). High muscle activity of gecko could be a reason of this condition. Glycogen is the carbohydrates in meat; it would be produced if the animal activity increases. As consequences, the glycogen levels increased, due to lower fat content as the demolition of energy.

Table 2. Nutrient comparison of meats in 100 grams (%)

Component	Cattle	Buffalo	Goat	Chicken	Duck
Protein	18.8	18.7	16.6	18.2	16.0
Fat	14.0	0.5	9.2	25.0	28.6
Carbohydrates	0	0	0	0	0

Source : (Muchtadi, 1989).

Table 3. Nutrient compotition in 100 gram meat

Nutrient	Nutrient Composition 100 Gram Meat (%)
Protein	22.0
Fat	24.0
Non Protein :	
Carbohydrates, organic salt, soluble nitrogen substances, mineral, and vitamin	3.5

(Syamsir, 2010).

Gecko average protein content of meat was 21.42±2.20%. Gecko meat protein content is almost the same as the protein content of meat in general. According to Levie (1967), meat contains 15-20 percent protein and according to Syamsir (2010) meat contains 22 percent protein. Crude fiber content of meat gecko is 0.45±0.13%. Gecko meat fiber content is almost the same as the fiber content of fat meat in general (a common meat consumed).

The water content of gecko's flesh average of 72.63±1.78%. Moisture content in meat gecko is almost the same as the water content in meat in general is 56% (Syamsir, 2010) while according Soeparno (1992), muscle contains about 75% water with a range of 68-80%. Water content in foodstuffs in determining the acceptability, freshness, and durability of the material (Winarno, 1997).

The ash content of gecko's flesh 1.46±1.37%. The ash content of both the animal's meat is almost the same as the ash content of meat in general, according to Sudarmadji *et al.* (1996) and Gracey (1999), ash content in fresh meat is 1%. According Soeparno (1992), the proportion of ash content in meat is affected by several factors, including genetic factors, gender, physiological animal, age, body weight, and nutrition. In proportional also be changed, when the proportion of one variable changes.

Conclusions

Gecko's flesh contained carbohydrate (3.21±2.10)% and protein (21.42±2.20)% that were higher than other livestock meat (cattle, buffalo, goat, chicken, and duck) but gecko's fat content, that was (1.28± 0.28)%, was lower than the others meat. Its crude fiber content was (0.45±0.13)% and ash was(1.46±1.37)%, which were similar with other meats.

Acknowledgement

Thanks for Faculty of Veterinary Medicine, Universitas Gadjah Mada which founded this study by Hibah Kompetisi Fakultas Kedokteran Hewan UGM, 2010.

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Effect of PUFA Supplementation on Cholesterol, Fat Content, Water Content, and Protein Content of The Simental-Ongole Cross Bred Meat

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Abstract

This study aims to determine the effect of Poly Unsaturated Fatty Acids (PUFA) supplementation of Lemuru Fish Oil (LFO), Palm Oil (PO), Soybean Meal (SM) and Fish Meal (FM) in cattle feedlot rations Simental-Ongole Cross Bred on the cholesterol, fat content, water content, and protein meat content. The treatments were as many as 5 kinds: Basal Feed (BF) (30% rice straw fermented (RSF) + 70% Basal Concentrate (BC), 30% RSF + 65 % BC + 5 % PO, 30 % RSF + 65 % BC + 5 % LFO, 30 % RSF + 60 % BC + 10 % FM, and 30 % RSF + 60% BC + 10 % SM. LFO and PO protection previously performed by saponification, while the SM and FM protected by 5% formaldehyde. Each treatment was given to the Simental-Ongole Cross Bred for 4 months of fattening feedlot model. All cattle treated is slaughtered and then meat was taken on the Longissimus dorsi (Ld) for determination of cholesterol, fat, water, and protein meat content. The result showed that PUFA supplementation was significantly affected ($P \leq 0.05$) cholesterol content and was not significantly affected ($P \geq 0.05$) fat, water, and protein meat content. Cholesterol content of meat from FM, SM, PO, LFO, and BF were 22.5 mg $100g^{-1}$, 24.16 mg $100g^{-1}$, 29.3mg $100g^{-1}$, 31.4 mg $100g^{-1}$, and 39.8 mg $100g^{-1}$, respectively, fat content were 3.41%, 3.26%, 3.62%, 3.38% and 3.45%, subsequently, water content were 75.05%, 74.57 %, 75.24%, 73.90%, 71.97% accordingly, and protein content were 21.75%, 22.60 %, 22.29%, 18.56%, and 21.99% respectively. Conclusion beef cattle that received PUFA supplementation obtained meat of lower cholesterol content than those fed without PUFA supplementation. PUFA supplementation either derived from waste oil (LFO and PO) as well as from not waste oil (SM and FM) are able to produce low cholesterol and fat meat content and high protein meat content compared to those without PUFA supplementation in the ration of fattening cattle Feedlot Simental-Ongole Cross Bred. Supplementation of Soybean Meal produced meat low in cholesterol, low fat and high protein

Keywords: ***cholesterol, cross bred, meat fat, meat protein, meat water, PUFA, simental-ongole***

Introduction

Beef cattle has been established as a priority commodity meat producer in Indonesia. Beef Livestock in Indonesia contributes 80-90 percent of the national meat production. Simental cattle which include exotic breed of beef cattle was one of cattle that crossed with Peranakan Ongole (PO) or Ongole Cross Bred which fattened by the farmer in feedlot system. Feedlot fattening carried out by providing forage feed (rice straw): concentrates in the ratio 30:70%. The proportion of the high concentrate in the diet besides to increase the rate of daily weight gain also will increase the deposits of fat and cholesterol content in meat. On the other hand, consumers want beef low in fat and cholesterol but high content of nutrition sources of food, also wants a wholesome, safety, sound, and consumed halal (Muslem Food) so that it can provide enjoyment during eating meat and nutritional needs

are met without suffering side effects that may be harmful to health. It is one of them determined by the performance of nutrients quality

Based on those background a formula of feed are made to produce meat in accordance with the needs of consumers. Implementation of productivity improvement methods in crossing cattle feedlot system with the results of low cholesterol meat, low fat and high protein levels at this time becomes very important to consider. Poly Unsaturated Fatty Acid (PUFA) in the feed materials of vegetable or animal both are fit for use as a source of nutrients to improve production performance of fattening beef cattle feedlot system (Orellana, *et al*., 2009) and quality of meat (Scollan *et al*., 2001dan Riyanto, *et al*., 2011). Altering the PUFA concentrations of meat is rarely directly related to the fatty acids composition of the diet (Hristov, *et al*., 2005). PUFA local sources of feed ingredients can be obtained from plants and animals. Flour and fishoil are a source of PUFA of the animal whereas the palm oil and soybean meal as a feed source of PUFA vegetable oils for beef cattle fattening (Riyanto, *et al*., 2010). PUFA supplementation either derived from waste oil (LFO and PO) as well as from not waste oil (SM and FM) are able to produce low-cholesterol meat compared to those without PUFA supplementation in the ration of fattened Simental-Ongole Cross Bred (Riyanto, dkk., 2011). The nutrition of content PO are cholesterol 59.35 mg 100 g^{-1}, water 74.70%, protein 23.69%, and fat 4.63% (Riyanto, 2010). This study aims to determined the formulations of feed for cross bred cattle feedlot that meets balance nutrient and obtained a low fat and cholesterol content but high in protein level.

Materials and Methods

Materials

This research used 30 heads of Simental Ongole Cross Bred weight approximately range of initial body weight of 350 kg of 30 head. Provided Basal Feed (BF) consisted of rice straw fermentated (RSF) and Basal Concentrate (BC) with a ratio = 30%: 70% (based on Dry Matter). The composition of Basal Concentrate (BC) ingredients preparation of the concentrate were soybean meal 8%, palm meal 5%, coconut meal 20%, corn milled 6%, rice bran 30%, pollard 14%, cassava by product 14%, minerals 2%, molasses 1%, and salt 1%.

The five treatments were :

1. BF = Basal Feed (BF) (30% rice straw fermented (RSF)+70% Basal Concentrate (BC),
2. PO=30% RSF+65%BC+5%PO,
3. LFO=30%RSF+65%BC+5%LFO,
4. FM = 30%RSF+60%BC +10%FM, and
5. SM = 30%RSF+60% BC +10%SM.

LFO and PO protection previously performed by saponification refers to a manner ingredient KOH and transform into salt Ca used CaCl, while SM and FM feedstuffs are protected by the addition of formaldehyde 37% (v / v) as much as 2 percent of dry matter of feed ingredient (Riyanto , *et al*., 2011). Nutrient content of each treatment is presented in Table 1.

Table 1. Nutrient content of treatment rations

Nutrisi	BF	PO	LFO	FM	SM
Nutrien (% DM) :					
Crude Protein	11.97	11.65	11.58	13,73	13,50
Exstract Eter	7.10	6.11	5.84	6,32	4,43
Crude Fiber	11.25	11.06	11.04	13,45	10,95
Ash	17.25	17.24	17.28	19,32	19,25
Extract Non-nitrogen	34.03	54.21	54.52	54,36	51,87
Fatty acids (mg 100 g^{-1}) :					
Oleic acids (C18:1)	864.88	1079.86	1541,13	54,36	826,09
Linoleic acids (C18:2)	431.69	427.40	634,62	470,80	395,79
Linolenic acids (C18:3)	27.66	33.49	37,67	39,19	26,32

BF (Basal Feed), PO (Palm Oil), LFO (Lemuru Fish Oil), FM (Fish Meal), SM (Soybean Meal)

Methods

Simental Ongole Cross Bred individually were caged in pens randomly, the position of stall tail to tail. Water for drinking was given ad libitum, Each treatment was given to the Simental-Ongole Cross Bred for 4 months of fattening model Feedlot beef cattle housed in Animal Husbandry Department of Agriculture Faculty of Sebelas Maret University at the Surakarta, Central Java Province, Indonesia. All cattle are slaughtered in the Surakarta Slaughter House. Cholesterol content of meat was determined by spectrophotometer method Liebermann-Burchad solution according to Plummer (1971). Levels of polyunsaturated fatty acids include oleic acids (C18: 1), Linoleic Acids (C18: 2), and Linolenic Acids (C18: 3) conducted using a gas chromatograph (GC). Proximate test and determination of cholesterol content of meat in Meat Processing Technology Laboratory and Department of Animal Husbandry Sebelas Maret University in Surakarta. Determination of levels of polyunsaturated fatty acids (PUFA = Poly-unsaturated Fatty Acids) of meat at the Laboratory of Chemistry and Biochemistry Center for Inter-University (Pusat Antar Universitas), Gadjah Mada University, Yogyakarta. The process of cutting carcasses in Surakarta Slaughterhouse method AMLC (1993). Meat samples taken from the Longissimus dorsi muscle (LD; Shortloin meat). Proximate feedstuffs and nutrien meat content were tested include the water content of meat by AOAC method (1984), protein content based on Kjeldahl method according to AOAC (1984). and fat content were determined by extracting samples of meat soxhlet method according to AOAC method (1984). Experiment was use the Completely Randomized Design (CRD) in the 5 kinds of treatment with 6 replications in each treatment. Data obtained from the study after tabulated and analyzed used various analysis CRD pattern. When significant results were tested more by Duncan Mean Range Test (DMRT) test.

Results and Discussion

Performance Simental Ongole Cross Bred nutrition quality meat results of research has been measured cholesterol levels, water content, fat content, and protein content in Table 2.

From the variable analysis of the Simental Ongole Cross Bred meat nutritional quality performance of that provides the effect of PUFA supplementation significantly ($P \leq 0.05$)

only on levels of cholesterol meat, but the effect is not significant (P≥0.05) on levels of fat, water content and levels of meat protein.

Table 2. Effect of PUFA supplementation on the cholesterol, fat, water, and protein the Simental-Ongole Cross Bred meat content

Parameter [1]	BF	LFO	PO	SM	FM	P-value[2]
Cholesterol (mg 100 g^{-1})	39.8^{a}	29.3^{b}	31.4^{b}	22.5^{c}	24.16^{c}	≤0.05
Fat content (%)	30,45	3,38	3,62	3,26	3,41	≥0.05
Water content (%)	71,97	73,90	75,24	74,57	75,05	≥0.05
Protein content (%)	21,99	18,56	22,29	22,60	21,75	≥0.05

[1] Parameter value are means of BF (Basal Feed), LFO (Lemuru Fish Oil), Palm Oil (PO), Soybean Meal (SM) and Fish Meal (FM) treatments

[2] P-value for main effects of parameter; means a common superscript at rows are differ significantly affect (P≤0.05)

Cholesterol meat content

Simental Ongole Cross Bred that have been given a basal concentrate feed without PUFA supplementation produced meat with the highest cholesterol levels 39.8 mg $100g^{-1}$ (P ≤ 0.05) compared with the supplemented PUFA. Cholesterol content were made from PUFA supplementation base-on oil (Lemuru Fish Oil 29.3 mg $100g^{-1}$ and Palm Oil 31.4 mg $100g^{-1}$) both have the levels did not differ (P≥0.05), as well as made from base on non- oil (Fish Meal 24.16 mg $100g^{-1}$and Soybean Meal1 22.5 mg $100g^{-1}$). Palm Oil Supplementation has resulted in meat with cholesterol levels 21.1% lower than without PUFA supplementation (31.40 mg $100g^{-1}$ vs. 39.80 mg $100g^{-1}$), Lemuru Fish Oil 26.4% (29.30 mg $100g^{-1}$ vs 39.80 mg $100g^{-1}$), Fish Meal 39.30% (24.16 mg $100g^{-1}$ vs. 39.80 mg $100g^{-1}$), and Soybean Meal 43.5% (22.50 mg $100g^{-1}$ vs. 39.80 mg $100g^{-1}$). Orellana, et.al.,, (2009) stated that the cholesterol concentration in the Longissimus muscle is total cholesterol obtained for both the breeds (0.80 and 0.73 mg g^{-1}) were similar to those recorded by USDA (2000), and were high compared to those obtained in other studies (Holbert *et al*., 1996). Guyton (1971) mentioned that cholesterol is fat soluble and slightly soluble in water so as to form an ester with fatty acids. Griswold *et al*. (2003) demonstrated that the response to added soybean oil depended on the forageto-concentrate ratio in the diet, with increased linoleic acid content of muscle tissues when greater forage was present in the diet. Rhee, *et al*., (1982) said that in muscles that contain relatively few intra-muscular fat content tend to contain lower levels of cholesterol in the muscles of many intra-muscular fat. Rusman (1997) stated that meat of ACC (Australian Commercial Cross) cattle are fattened in feedlot system in Indonesia contain cholesterol 34.28 mg 100 g^{-1}. Cholesterol content in this research was included safety to be consumed as food . The cholesterol content of male Ongole Cross observed in this study was still categorized as acceptable, as it was still in the ranges of 70 mg 100 g^{-1} (Simonsen and Rogovski, 1989), 70 mg 100 g^{-1} (Soeparno, 1997), 75 mg 100 g^{-1} (Judge, at.al., 1989) and 62-82 mg 100 g^{-1} (AMLC, 1993). Riyanto (2003 and 2010) cholesterol levels of beef cattle PO Shortloin, Silverside, Chuck, and the brisket point end were 55.77 mg $100g^{-1}$, 59.49 mg $100g^{-1}$, 62.05 mg $100g^{-1}$ and 60.12 mg $100g^{-1}$ respectively.

Fat meat content

Although different sources of PUFA did not affect the fat content of meat, but from the fat content of profile data appeared that supplementation of Oil Palm produced meat with higher fat content compared with other treatments and the lowest of supplementation was Soybean Meal. Simental Ongole Cross Bred meat was contained fat with decreased levels of 5.5% through supplementation compared with Soybean Meal without PUFA supplementation (3.26% vs. 3.45%). Factors that may affect the fat content of meat is a breed, species, feed, and individual animals (Judge, *et al*., 1989). Shackelford (1995) said that meat with a high water content tend to contain low levels of fat. Riyanto (2003 and 2010) mentioned that the fat content of beef PO from meat Shortloin, Silverside, Chuck, and the Brisket point end were 4.92%, 4.42%, 4.48% and 4.69% respectively. Lawrie (1995) stated that intramuscular fat content of 3-7% rate to remain high consumer acceptance based on aspects of beef palatability. The mean values obtained from intramuscular fat of Longissimus muscle fell within the recommended limits. Taken together, the meat of Criollo Argentino steers may have better palatability and more healthy, although the risk to health is difficult to evaluate (Orellana, et.al., 2009). Enhancing the linoleic acid content of the diet for finishing cattle through either oils or oilseeds failed to enrich muscle or adipose tissue with linoleic acid due to the extensive biohydrogenation that occurs in the rumen (Mir *et al*., 2003), but it increased linoleic acid concentrations in other studies (Madron *et al*., 2002).

Water meat content

The water content of beef have different levels are not significant at each treatment ($P \geq 0.05$). Table 2 showed that the data content of the water beef supplemented PUFA produce meat with higher water content compared to those were given only the basal concentrate without PUFA supplementation. This indicated that the meat of Simental Ongole Cross Bred in PUFA supplementation containing a lower dry matter without PUFA supplementation. The water content of meat can be affected by fat content, high accumulation of intra-muscular fat content of meat can loosen the bond tissue structure of the meat so much water that released then the high fat content in meat water content tended to decline. When the water content of animal meat is high then the fat content will drop (Browning, *et al*., 1990). Lawrie (1995) mentioned that increased protein bound with water then the water content of meat increases. Riyanto (2003 and 2010) noted that the water content of beef PO from meat Shortloin, Silverside, Chuck, and the brisket point end were 74.76%, 75.14%, 74.83% and 74.06%. respectively

Protein meat content

Protein content of beef have no significant levels in each treatment ($P \geq 0.05$). Data meat protein sources supplemented PUFA (Table 1) contain higher protein compared to those given only the basal concentrate without PUFA supplementation. PUFA supplementation based on Soybean Meal and Oil Palm produce meat with higher protein levels of 2.8% (22.6% vs. 21.29%) and 3.9% (22.29% vs. 21.29%) than without PUFA supplementation respectively. High and low protein content of beef were associated with the water content and levels of intra-muscular fat. Kramlich, *et al*., (1973) said that contain high levels of muscle proteins when low intramuscular fat content and high water content. Forrerst, *et al*., (1975) ask that with differences water content between the muscles may resulted in an differences protein content, it is caused by the hydrophilic properties of muscle proteins,

resulting in a lot of meat water molecules are bonded by PO beef protein content of 24.81% (Soeparno, 1994). Riyanto (2003 and 2010) said that the protein content of beef PO catlle from meat Shortloin, Silverside, Chuck, and the brisket point end were 22.77%, 26.08%, 24.29% and 21.61% respectively.

Conclusions

1. PUFA supplementation based on waste oil (Palm Oil and Lemuru Fish Oil) and non-waste oils (Fish Meal and Soybean Meal) on the basal feed (rice straw fermented 30%: 70% concentrate basal) Simental Ongole Cross Bred cattle fattening feedlot, both are produce meat with higher levels of lower cholesterol as compared with no supplementation of PUFAs.
2. PUFA supplementation based on Palm Oil, lemuru Fish Oil, Fish Meal, and Soybean Meal has resulted in meat with the lower cholesterol levels 21.1%, 26.4%, 39.30% and 43.5% than without PUFA supplementation on basal feed.
3. Fat content, water and meat protein of Simental Ongole Cross Bred cattle fattening feedlot have levels within the range that does not differently both supplemented PUFAs and without PUFA supplementation.
4. Based on the research data showed that PUFA supplementation based on Soybean Meal is more effectively used as a source of PUFAs to improve the nutritional quality meat of Simental Ongole Cross Bred are able to produce meat with an indication of low-fat, low cholesterol, and high protein content.

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Anticancer Activity of Chitosan from Local Chitin Waste of Fishery Products In Vitro

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Abstract

The aim of the experiment was to produce bioactive compound of chitosan enzymatically from local chitin waste to be applied in functional instant drinks. The activity of anticancer in this product was determined using in vitro assay. The chitin waste was obtained from crab shells as by- product of canning crabs meat industry in Cirebon West Java Indonesia. Production of chitosan enzimatically was using chitin deacetylase enzyme produced by Bacillus papandayan isolated from Kamojang Creater, West Java. This experiment resulted the technology of process and production of chitosan which degraded enzimatically. The product was chitosan-tea drink which is ready to be dissolved in water. The physical characteristics of Chitosan-tea drink instant which is mixture between flour and dry chitosan gel (1.5 cm x 1.5 cm) were having soft surface, brown-clear color, and smooth. Formulation of chitosan-tea drink (23.7 g per pack/one serve) consisted of 22.5 g sorbitol, 0.375 g green tea extract, and 0.8 g chitosan). This chitosan-tea drink instant contained 0.22% w/w water, 0.11% w/w ash , 0.03% w/w protein, 0.002% w/w lipid, and has 58.5 kkal calory which is suited SNI 01-3722-1995 national standard. The product was then tested for toxicity by in vitro using AH 109 cancer cells. The test showed that chitosan was cytotoxic to cancer cells AH109 with a value of KI_{50} (tg/mL) that was equal to 189.00 for exposure for 8 hours and the value of 1.20 for 24 hours.

Keywords: chitosan, anticancer, in vitro

Introduction

Chitin is an insoluble polysaccharide consisting of ß(1-4) linked N-acetyl-D-glukosamine (GlcNAc) units that most abundant polysaccharide in nature after cellulose. It is widely distributed as structural component of crustaceans, insects, and other arthropods, as well as component of cell walls of most fungi and some algae. About 10^{11} tons of chitin is produced annually in the aquatic biosphere alone, however, only 0.1% of this material is currently being converted to valuable product. Indonesia chitin waste which has not been exploited 56.200 metric of ton per year (Department of Marine and Fishery, 2003). Chitosan is chitin which has been eliminated its acetyl group leaving free amine residue that making it as polycationic character.

Conversion of chitin to chitosan in industry is generally done using termochemical technique, that use strong alkali at high temperature. This process requires high energy, maintains high temperature and produces waste and basic product with high concentration so is potencial to become toxic in environment. Alternatively, deacetylation conversion of chitin to chitosan can be done enzimatically by *chitin deacetylase*=CDA. This process makes chitosan easier to be controlled, more efficient, specific and safe to environment.. *Chitin deacetylase* synthesized by various crops, bacterium, mushroom, and sea organism (Kupiec and llan, 1998). Mushroom *Colletotrichum lindemuthianum* (Tsigos and Bouriotis 1995,

Tokuyasu *et al*., l996, Tsigos *et al*. 2000), *Mucor rouxii* (Kafetzopoulos *et al*., 1993; Kolodziejska *et al*, 1998), *Absidia coerulea* (Gao *et al*., 1995) and *Aspergillus nidulans* (Alfonso *et al.*, 1996) proven produced CDA. Local isolate of producer CDA, *Bacillus papandayan* K29-14, has been reported by Rahayu *et al.* (2004). Purification with column chromatography reported by Rochima (2004).

Waste treatment of crab chitin to improve added value need to be done. Waste treatment technology input of chitin is expected will increase its market price. On the other hand, formulation of food product bases on chitosan is needed to applied as coroner heart sickness inhibitor. This thing is constituted by till now supplement of food (*neutraceutical*) is containing chitosan in international market which high, that is Rp 250.000-300.000/100 item capsule for a few certain merk. Unfortunately, neutraceutical unable to be enthused public because its form looking like drug.

Chitosan is composed primarily of GlcNAc and GlcN (2-amino-2-deoxy-ß-D-glucopyranose) residu. Unlike most polysaccharide, chitosan has three types of reactional functional groups, an amino group as well as both primary and secondary hydroxyl groups at C-2, C-3, and C-6 position respectively. Amino group (NH_2) what causes chitosan to have the character of dissolving water so that easy to be application (Bastaman, 1989). This positive charge makes chitosan can tie compound around which haves negative charge, like cholesterol, fat, bile acid, and some other fat generations at the time of passing alimentary canal, and releases it through faeces (Furda, 1980). Chitosan can absorb 97% body fat, binding ability of fat by chitosan had been proved by Japan researcher long time ago (Sugano *et al*, 1980).

Cancer is one disease that can cause death and the existing treatment methods still have several weaknesses, among others because it has a selective toxicity is low, so these drugs also attack normal body cells resulting in side effects serious enough. Search of new drugs that effectively and safely continue through the synthesis or the use of natural resources. The search for anticancer drugs can be done using various methods of testing as a tool to detect the presence of the anticancer activity of the material under study. The balance of apoptosis and cell proliferation is a key determinant of growth in all normal tissues. Apoptosis is also an important phenomenon in the destruction of tumor cells by a chemotherapeutic, γirradiation and immunotherapy that works by stimulating the onset of apoptosis in target cells while the previously known that a direct cytotoxic effect of chemotherapy on tumor cells (Kaufmann and Earnshaw, 2000; Herr and Debatin, 2001; Hu and Kavanagh, 2003).

The research objective was to produce chitosan from chitin waste enzymatically small crab is formulated in the instant drink product, characterize the physio-chemical properties, and then tested its activity against cancer cells.

Materials and Methods

This work was conducted from January 2010 to October 2010 in Laboratory of Technology of Industry of Postharvest Fishery, Fishery and Marine Science Faculty of Padjadjaran University and Laboratory of Microbiology and Biochemistry Biotechnology Research Center Bogor Agricultural University.

Chitin preparation

Bacillus papandayan isolate was collection of Laboratory of Microbiology and Biochemistry Biotechnology Research Center Bogor Agricultural University. Growth isolate media (Sakai *et al* 1998) i.e: Bacto Agar, Ammonium sulphate, K_2HPO4, NaCl, $MgSO_4$. $7H_2O$, Yeast Extract, Bacto trypton, Coloidal chitin. Coloidal chitin made of chitin powder Sigma based on Arnold and Solomon method (1986). Chitin glycol made of chitosan glycol based on Trudel and Asselin method (1989).

Chitin deacetylase production

Culture of *Bacillus papandayan* has been fermented in Sakai media at pH 8.0 and 55 ^{o}C for 2 day. After completed, enzyme is harvested by sentrifugation 8000 rpm for 15 min. Supernatant dissociated and tested activity of CDA according to Tokuyasu *et al*., 1996.

Chitin deacetylase assay

Mixture consists of 50 µL chitin glycol 1%, 100 µL 0.2 M borate buffer pH 8.0 and 150 µL enzyme. Incubation it for 30 min. at 55 ^{o}C. Inactivation enzyme in 100 ^{o}C for 15 min. 200 µL of mixture was added 200 µL $NaNO_2$ 5%, 200 µL acetic acid 33%, then vortex and let 10 min. After that, is added 200 µL ammonium sulphamate 12.5% then shaker 30 min at room temp. Then is added 800 µL HCl 5% and 80 µL 0.1% indol in absolute ethanol (prepared when will be applied). Boiling it for 5 min. then cooling, It is added 800 µL absolute ethanol before measurement of absorbance at wavelength 492 nm (Tokuyasu *et al*., 1996).

Chemicall technique and deacetylation enzymatic of chitosan

Crabshell chitin waste was obtained from Bondet Cirebon Indonesia. It is washed and sun-drying for two-day, then flouried until 177 mm to 325 mm particles. Demineralization by addition of HCl 1 N 1:7 ratio, heated 90 ^{o}C 1 h, decantation, then cleaned again until pH 7 and dried. Deproteination by added of NaOH 3,5% ratio 1:10, then heated 90 ^{o}C 1 h, more decantation, washed until pH 7, then dried. Bleaching by addition of H_2O_2 2% ratio 1:10 till get is white chitin flour (Suptijah, 1992)

Deacetylation of chitin flour chemically by soaking in NaOH 50% (1:10) at 80 ^{o}C for 1 h. Chitosan formed then rinsed with water until neutral, then is measured degree of deacetylation. Enzymatic deacetylation by CDA which precipitated ammonium sulfate. 1 ml soluble chitosan 1% incubated with CDA 0.04 U/mg chitosan at 55 ^{o}C for 24 hours (Rochima, 2005)

Formulation of Chitosan-Tea instant drinks

Formulation of chitosan-tea drink refers to Palupi , 2006. Chitosan 1 g dissolved in 100 mL acetic acid 1% (b/v), added sorbitol. 0,25 % (b/v), heated at 85 ^{o}C, then put into petridisk (D=10 cm). After printed, chitosan layer dried in oven. Formulation of green tea ehich acceptable organoleptically is consisted of green tea extract 0.25% b/v and sorbitol 1.5% b/v, and chitosan instant (chitosan 1% b/v, acetic acid 1% v/v, and sorbitol 0.25% b/v).

Preparation of cultured cancer cells

The prepartion of cultured cancer cells was done according to Ananta (2000). Cell suspension: AH109 cancer cells in a frozen state stored in a tank containing liquid nitrogen after being expelled, experience the process of thawing, in advance, ie ampoules containing the cells were incubated at 37 ° C pads or held by hand until the content of it melts. Centrifuged at 228 XG for 10 minutes, the supernatant discarded and the pellet plus basal medium, then centrifuged again at 228 XG for 5 minutes, so preservative material of cells and cells that have died can be removed from the cell culture. Pellet cell growth media was then added and homogenized, then the cell suspension was transferred into a flask with 5 ml of growth medium, then incubated in an incubator with 5% CO_2 at 37 ° C. Maintenance of cell turnover or laundering is done by the media every 3 days or if the suspension has changed color from red to yellow to indicate there has been a decrease in pH due to metabolic activity of cells, used for culturing cells that are in the logarithmic phase of growth curve .

Monolayer: Maintenance of a layer of cells (AH109 cells) with cell suspension, washing or replacement only in the media in the flasks added by enzyme solution 0.02% trypsin in 0.5% EDTA-PBS, for the removal of the cells attached to the flask wall. Media that will be replaced at first discarded all so that only the remaining cells attached to the wall flask, then added to 500 mL of trypsin solution (for 5 ml culture volume) and incubated in the incubator for 8 (eight) minutes. Cells that are attached to will be detached, then add just enough PBS before the cell suspension was transferred into centrifuge tubes and performed the cell washing procedure as has been done on cell suspensions.

Cytotoxicity activity assay

Chitosan at various concentrations to test the activity of cytotoxicity against AH109 cancer cells by MTT assay method. The principle of measurement is the ability of living cells to change compounds the pale yellow MTT into blue formazan compound. Cancer cells (2 x 10^4) grown in 96 well plates, as many as 100 mL per well. After 24 hours, against cancer cells were given various concentrations of test compound and incubated again for 24 hours. 10 mL MTT was added into the test plate and to control, and then incubated in an environment with 5% CO_2 at 37 º C for three hours. In each well, which is formed formazan was dissolved with 100 mL 1 N hydrochloric acid. Absorbance of the dissolved formazan was measured at 450 nm (Yang *et al*., 2005).

Results and Discussion

The condition of industrial crab waste

The main source of chitin for this research came from a small crab waste is a byproduct of small crab meat canning industry as shown in Figure 1 below. Waste is processed by a small crab is an industry miniplan households assisted small crab meat canning industry is located in the area Bondet, Cirebon regency. Miniplan scattered throughout the Cirebon area totaling about 20 pieces, and the total production of small crab shell waste waste about 10 tons perday.

Small crab waste treatment process begins with washing, boiling, peeling and sorting. Small crab body parts were separated into the main part of the meat, and the waste includes

shells, claws large and small claws. Small crab meat from miniplan will immediately be packed to be sent to a small crab meat canning industry Philips Pemalang PT Central Java to be exported to various countries, while the waste is dried and packaged for direct sale or processed into flour chitin. Waste of small crab claw shells and dried sold in local market at a price of Rp 1800.00 per kilo, while the waste of small pincers USD 1500.00

1.Crabs miniplan Cirebon

2. Crabs

3.Washing

4.Boiling

5.Sorting

5a.Crab shell

5b.Big tweezers

5c. Small tweezers

6. Packing

7.Chitin flour

Chitin Deacetylase production

Morphology and characterization of *Bacillus papandayan* producer CDA, according to Rahayu *et al*. 2004 as follows: Gram-positive bacteria bar shape, having spore, and motile. Optimum pH=7 at 55 ^{o}C. Free filtrate cell harvested at phase stationer on 28-32 incubation periods. Optimum CDA activity at pH=8, 55 ^{o}C, and activated by $MgCl_2$ 1 mM.

Figure 1. *Bacillus papandayan* K29-14 in chitinolityc media

Free filtrate activity of cell CDA at this research is 0.005 U/ml, smaller than CDA activity from *M*. *rouxii* 0.0305 unit/ml (Kafetzopoulos *et al*. 1993) and CDA from *C. lindemuthianum* 0.0195 unit/ml (Tokuyasu *et al*. 1996). Tsigos and Bouriotis (1995) tested activity CDA from different strain of *C. Lindemuthianum*, it's activity 0.002 unit/ml. From third of above researcher, only Tokuyasu *et al* (1996) who applied the same method like in this research, while Kafetzopoulos *et al*. and Tsigos and Bouriotis applies method is having Bergmeyer (method reaction of three enzymes). This research applies method Tokuyasu *et al*., because easy practically. It also applies a few of raw material, and low cost.

Formulation of Green Chitosan-tea drink instant

The result of quality product analysis in Tables 1 has suitable to standard SNI 01-3722-1995 (orange taste drink powder). SNI orange taste drink selected as comparator because until now hasn't standard for chitosan. SNI 01-3722-1995 assumed to be nearest of instant drink product of chitosan-green tea. With the same analytical method, functional drink of tomato-cinnamon chosen beloved has antioxidant activity 5.44 mM Trolox ® (Radianti, 2005). Thereby, instant drink of chitosan-green tea showed high antioxidant activity (8.41 mM Trolox ®) if compared to instant drink of tomat-kayu manis one, tomato is rice of licopene have a good antioxidant activity and cinnamon that is also rich phenol.

Characteristics of chitosan-green tea powder such as white-brown colour of gel (1,5 cm x 1.5 cm) which has smooth surface and shiny and soft. Its weight is ± 23.7 grams per pack/once consumption (sorbitol 22.5 grams, green tea extract 0.375 grams, chitosan 0,8 grams).

MTT assay for in vitro

In vitro testing that can either guess the response of tumors to the drug and the results of these estimates will be invaluable. Test whether a chemical component has anti-tumor activity can be done through two ways, namely in vivo and in vitro, because the test in vivo is very costly and time it was developed in vitro assays using cultured cancer cell lines such as cultured strains KR-4 (lymphoblastoid B humans). Testing the activity of proliferation of cancer cells and normal cells using the Alamar blue method or methods of MTT (3 - [4,5-dimethylthiazol-2-yl] -2.5 diphenyl-tetrazolium bromide) in 96 wells flat plate.

This observation is based on MTT reduction by mitochondrial succinate dehydrogenase of living cells provide formazam blue color that can be measured with a spectrophotometer.

Based on the amount of chitosan is given, ie at concentrations of 0, 10.20, 50, 100 and 200, and then exposed for 24 hours with MTT method, the obtained data as follows:

Table 1. Result of achitosan-green tea drink instant product analysis (150 mL water per serving)

Parameter	Unit (b.b.)	Chitosan-green tea Drink instant		Comparation[b]
		Dry[a]	After boiling	
Water content	% b/b	0.22	85.30	Max.0.5
Ash content	% b/b	0.11	0.04	Max. 0.1
Protein content	% b/b	0.03	0.94	-
Fat content	% b/b	0.002	0.08	-
pH	-	-	5	-
TAT	%	-	1.73	-
Tot. Carbohyd.	% b/b	0.37	13.64	-
Tot. Susp.. Solid	^{o}Brix	-	1.354	-
Vitamin C	mg/100g	-	-	Min 300
Antioxydan act.	mM Trolox® % b/b	-	8.41	-
Tot. Glucose (as sucrose)	Kcal	-	-	Max.78
Calory	-	58.5	58.5	Max. 312
Shynthetic sweetener		-	-	no saccharine and syclamate

a Chitosan-green tea instant product which has not been poured boiling water into
b SNI 01-3722-1995 (SNI orange taste drink powder which has not been poured boiling water)

Table 2. Cytotoxic activity of chitosan after 24 hours of contact with cancer cells AH109

Observation	Absorbance of control cell	Absorbance of control medium	Formazan absorbance of AH109 cells after contact with chitosan for 24 hours at certain concentration (µg/mL)				
	(+)	(-)	12.5	25	50	100	200
1	0.628	0.124	0.536	0.531	0.518	0.149	0.134
2	0.621	0.117	0.529	0.550	0.545	0.120	0.137
3	0.641	0.114	0.508	0.510	0.573	0.133	0.140
Means	0.630	0.118	0.524	0.530	0.545	0.134	0.137
IC_{50} (%)			16.772	15.820	13.439	78.730	78.254

Cell culture is a method of studying the behavior of animal cells free of systemic diversity that usually appears in animals during normal homeostasis and under the pressure of the experiment. Cells used may be a flow cell, namely cell population derived from a particular network resource that has cultured further, until it reaches the sub culture. There are two types of cultured cancer cell line that is attached to form a layer of culture (monolayer) on a solid substrate, or a suspension in culture media. Both of these cell types have different properties, where the cell suspension does not require support or supporting material to stick, otherwise the cell layer requires support. The suspension usually from hemopoetik, blood cells or cells from malignant tumors, whereas monolayer cells normally to cells derived from tissue (Freshney, 1994).

The observations in this study is the proliferation of cells. Cancer cells are not normal cells, therefore it does not follow the normal rules of normal cell division. Normal cell cycle is controlled by a group of cyclin proteins takes place through a phase of mitosis (M), gap-1 (G1), DNA synthesis (S phase), gap-2 (G2), mitosis (M) and so on. Daughter cells of mitosis results regularly into the cycle in the G1 phase, some daughter cells enter the resting phase (G0). Cells at G0 phase can be actively re-entered the G1 phase of the cell cycle (Slinerland and Tannock, 1998). The speed of proliferation of tumor cells is different. Proliferating cells are not often encountered because of cell death at high speed.

Cell response to a compound depends on the type of cell, the weight of pressure (compound concentration) and duration of contact with the compound. To see the effect of length of exposure to levels of cytotoxicity, chitosan exposure performed at different times, ie 8 and 24 hours of exposure and the value of KI50 obtained results that exposure for 8 hours obtained KI50 (µg / mL) at 189.00, while if exposed for 24 hours to reach 1.20. This suggests that the longer the contact with the chitosan compound trials of cancer cells, the greater the effect sitotoksiknya, as indicated by the declining value of KI50 in a longer contact time.

Conclusion

Product yielded in the form of instant tea-chitosan drink readily dissolved in water to be consumed. Fisically, chitosan-tea drink instant performance were white-brown flour mixed with dry chitosan gel (1.5 cm x 1.5 cm), soft surface, brown clear color, and smooth. Formulation of chitosan-tea drink was weight 23.7 gram perpack/once consumption (sorbitol 22.5 gram, green tea extract 0.375 gram, chitosan 0.8 gram). Chemically, chitosan-tea drink instant have water content 0.22% w/w, ash content 0.11% w/w, protein content 0.03% w/w, lipid content 0.002% w/w, calory 58.5 kcal which suitable with SNI 01-3722-1995 standard. Chitosan are cytotoxic to cancer cells AH109 with the KI50 (µg / mL) of 189.00 for exposure for 8 hours and the value of 1.20 for 24 hours.

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Relationship Between Body Part Measurement, Body Weight and Flying Speed of Racing Pigeon (*Columba linia*) at Local Tournament of Sprint Racing (Case at Local Tournament of the Sprint Pigeon in the District in of Bandung)

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Abstract

A research on relationship between body part measurement, body weight and flying speed of racing sprint pigeon was conducted at the local championship of racing sprint pigeon in different locations (Adipura Gede Bage, Batu Jajar, Dago Resort and Lanud Sulaeman. Bandung). The objective of research was to find out the coefficient of correlation between body part measurement and flying speed, and their best equation of regression. The method of research was descriptive and data were collected using census. Total 200 heads of racing sprint pigeons were available to observe of body part measurement (wings length, wings stretch, primer feather length of wings, wings dense, and length of tail feather, width of breast circumstance of breast, length of back, and length of os. Pubis) body weight and flying speed (meter/sec.). The Result showed that coefficient of correlation between body part measurement and flying speed were low category and the best equation was multivariate linear regression. It was concluded that body part measurement and body weight were not influence for flying speed.

Keywords: body part measurement, body weight, championship of racing sprint pigeon, flying speed

Introduction

Racing pigeon in Indonesia is the local pigeons which originated from wild pigeons that had long cultivated the deployment of European origin (Antawijaya, 1988). Male racing pigeon body shape performance is bigger than the females. there is a knob above the beak, sturdy and big neck, burly body, and swelled chest, having a loud snoring, but rather narrow pubic bone.

Pigeons belong to a class: Aves, Sub Class: Neornithes, Super Order: Neognathae, Order of the Columbiformes, suborder Columbae, Familie: Columbidae, Genus: Columba Species: Columba livia (Levi, 1945).

In Indonesia, pigeon besides as meat source, also as pet especially as racing pigeon. Racing pigeons is a descendant of the rock-pigeon from *Columba livia* species. Sprint racing pigeon was used in the basic flying speed race. The racing pigeons fly and traveled around 100 – 1000 kilometers at speeds of 50 miles /hour (90 km /h), while the sprint racing pigeons have been trained only for around 1000 meters, flying only 2 meters above to meet the jockey who holds the female dove mate. The average racing pigeon's body weight are 2.5 kg body weight (ranged between 2 – 4 kg), length from tip of beak to tip of tail is 13 inches.

Since 1970 the sprint racing pigeons more popular and called as *andokan pigeon*. The first sprint racing pigeons was held in East Java, especially on the island of Madura, as

traditional performance for welcoming the tourists, but when they were popular, they grouped in PPMBSI (Persatuan Pencinta Merpati Balap Spint Indonesia- Association of Indonesian Sprint Racing Pigeon Lovers). The Sprint Racing pigeons which are flying as high as two meters above the ground, will prouped as good catepory. Until now the association have no information regarding the standardization of ideal body size that can support the speeds of sprint racing pigeon , so research on the relationship between the body sizes with flying speeds need to be investigated.

Although the racing pigeons was released from places hundreds of miles away, they can come home, so scientists think that the pigeons have a magnetic field detection power of the earth. When they are flying, there are synergistic movement between the primary feather of wings and secondary feathers that cover the wing, feather that accelerating the speed of flying and tail feathers to determine the flight direction.

There is retionship between the flying speed and body movement between the working muscles of the body and force the body length, but there is no scientific data about it. Flying is a simultaneous movement of body muscles and body part on body lifting. The aims of this study was to find the effect of pigeons body part on flying speed in racing.

Materials and Methods

200 male sprint racing pigeons, which were participated in several races in Bandung area (Sulaeman airport, Field of Adipura Gede Bage, Batu Jajar and Dago Resort).

Research method was survey, using purposive sampling for data collection, which was taken from 1^{st} to 5^{th} rank pigeon champion of every local championship. Data were analyzed using correlational and multiple linear regression Step Weis flying speed as the dependent variable and independent variables consisted of 11 variables including: (*a*). length of wings (LW) is the length of the base of the wing (os. Phalangs) to the tip of *os metatarsus*, (*b*). Length of stretch wing (LoSW) was the length of the os. humerus to the tip of the wing feathers, (*c*). Length of primary feathers of wings (LoPFoW) was the distance between the base of the primary feathers of wings with the tip of the primary feathers of wings, (*d*). Dence of the base of the wing (DoBW) is the arm wing thickness, (*e*). Length of the feather of tail (LToT) was the distance between the base of the feather of tail to tip of tail feathers, (*f*). Width of the chest (WC), a distance of protrusion of the shoulder joint (*os.scapula*) left and right, (*g*). Chest Circumference (CC) was the circumference circle the body. (*h*). Length of the neck (LN) was the distance between 7^{th} *os. vertebrae* to limit head bone, (*i*). Length of the back (LB) was the distance between the bone joints of the neck with the backbone to the border of the os. vertebrae caudal, (*j*). Distance between the os. Pubic (DoP), (*k*). Body weight (BW) was a weight of life adult male racing pigeon, (*l*). Flying speed (FS) was a quotient between the distance with time.

After tested correlationship using a scatter diagram, where the independent variable X was the horizontal axis and the dependent variable Y is the vertical axis, then the point was determined by the ordinate (Y) and absis is (X), found the grafic forms of regression. Based on the description of the scatter diagram, a linear regression analysis was defined using a simple general linier model:

$\mathbf{Y = \beta_o + \beta_1 x_1 + \mathcal{E}}$, whereas estimation Model: $\mathbf{\hat{Y} = b_o + b_1 x_1}$

where: $\hat{Y}$ = Estimation flying speed , b_o = mean, b_1 = coefficient of regression , x_1 = independent variable, $\mathcal{E}$ = Error.

In this study the independent variables are 11 variables, then the multiple general linear regression model with form is:

$\mathbf{Y = = \beta_o + \beta_1 x_1 + \ldots\ldots + \beta_n x_n + \mathcal{E}}$, then estimation form is: $\mathbf{\hat{Y} = b_o + b_1 x_1 + b_2 x_2 + + \ldots \ldots + b_n x_n}$

Compatibility equations in suspect flying speed is shown by the coefficient of determination by the formula: $\mathbf{R^2 = (TSreg) / (\Sigma yi^2)}$ (Sujana, 2002). Where: TSreg = Total Squares of Regression ; Σyi^2= Total Square of data.

Five grouping of coefficient of correlation are: very high from 0.91 to 1.00; high 0.71 to 0.90; moderate 0.41 to 0.70; low 0.21 to 0.41 and very low 0, 00 to 0.20 (Surya, 2006).

Tools used: (*a*). Measuring band, (*b*). Digital Scales, (*c*). The pair of compasses, (*d*). Stop Watch.

Result and Discusion

The results of data analysis through statistical analysis showed in Table 1.

Table 2. Data Structure of sprint racing pigeons as 1st to 5th champion at race of local championship

Body Part Measurement	N	Mean of 1st Champion	Mean of 2nd Champion	Mean of 3rd Champion	Mean of 4th Champion	Mean of 5th Champion	CV (%)
FS (m/sec.)	40	56.06±3.26	53.97±2.58	53.07±2.71	52.96±2.97	50.68±2.02	3.98-5.82
LW (cm)	40	15.50±1.18	15.82±1.05	15.54±1.06	15.42±1.09	15.24±1.17	6.62 – 7.71
LoSW (cm)	40	31.39±1.13	31.71±0.02	31.26±0.96	31.49±1.06	31.24±1.14	3.06 – 3.60
LoPFoW (cm)	40	17.01±0.62	17.16±0.54	16.00±0.53	17.08±0.47	17.01±0.53	2.70 – 3.60
DoBW (cm)	40	1.05±0.14	0.99±0.20	1.01±0.12	0.99±0.16	1.01±0.145	11.8–20.16
LoFoT (cm)	40	12.62±0.79	12.81±0.82	12.70±0.79	12.81±0.88	12.44±0.90	6.20 – 7.20
WoB (cm)	40	7.87±0.24	7.86±0.22	7.84±0.241	7.88±0.22	7.76±0.24	2.50 - 3.14
CC(cm)	40	27.85±0.53	27.76±0.46	27.86±0.60	27.89±0.52	27.59±0.57	0.45 – 0.60
LN (cm)	40	8,87±0.71	8.66±0.78	8.87±0.81	8.84±0.78	8.46±0.85	0.77 – 0.84
LB (cm)	40	14.61±0.65	14.50±0.38	14.59±0.49	14.59±0.46	14.43±0.49	0.38 – 0.54
DoP (cm)	40	0.55±0.04	0.54±0.03	0.55±0.04	0.55±0.04	14.43±0.35	0.03 – 0.31
BW (gram))	40	388.92±8.43	387.56±7.78	388.39±9.22	389.08±7.82	384.64±9.88	7.78 – 9.88

From Table 1, it shows that the flying speed (FS) on the 1st to 5th champion was ranged from 56.06 m/sec. to 50.68 m /sec. which is one of the criteria considered. But this difference between 1st champion and 5th champion is not significant. Statistically anaylisis of coefesien variation is still between 3.08 to 5.82% , it means that data are uniform. Flying speed is a combination of the potential size of the body part, the working muscles that attach to the bones of the body, the mecanical force that make the pigeons fly, the motivation to fly and environmental conditions (wind, temperature) at which pigeons in race (Cartmill, 1991 ; Djannah and Slistyani, 1986). Length of wings (LW) ranges from 15.50 to 15.24 centimeter, Length of wing consists of the os. Humerus, ulna and metatarsus place attachment of the muscles around the wing and Length of Premary feather of wing (LoPFoW) embedded surface which consists of the primary feathers of wing (10 sheets) which length ranges from 17.16 to 16.00 centimeter and all data were homogen. Secondary feathers of wings (8 sheets) serves to give a boost to the body to increase the flying speed . Length of stretched

wing (loSW) ranged from 31.71 to 31.24 centimeter which is almost double the length of the length of wing.

Data from Table 1 also showed that Dence of base of the wing(DoBW) of 1st champion to 5th champion ranges between 0.99 to 1.05 centimeter, for all pigeons showed high heterogeneous levels (11.8 to 20.16 %) when compared with other body parts. Between the base of the wing with the body formed a palpable glenoid, glenoid joint strengths can support the wings with a strong movement, so will the maximum flying speed. The formation of a strong joint glenoid must go through a routine practice (Sutejo, 2002). Feather of tail is very special and only owned by poultry, parallel to the body is not higher and not lower (Levi, 1945). The number of tail feathers and there are 12 sheets, when they are still, the tail just like covered corn (Darwati, 2003). When the gigeon are flying and turn, going down and rest, the tail feather serves to control (Tyne *et al*, 1976). The tail feathers consist of main tail feathers which covered between upper and under tail feathers (Levi, 1945).

Width of breast of 1st to the 5th champion of about 7.76 to 7.88 centimeter, to be uniform; with coeficien of variation approximately 2.50 to 3.14%. Wide Width of breast means that the pigeon respiratory organ are good, that the O_2 availability during breathing, as well as they training or race which requires high quantities of Oxygen input. Oxigen needs can be held by by the lungs because there are some air sac system (interclavicular sac, antherior thoracic sac and posterior thoracic sac) which ensures the supply of air into the parabrochus to the lungs. It works paralel; the air enter from one end through the other end . Chest Circumtance ranged between 27.59 to 27.89 centimeter, also uniform, the diversity of the data can be said to be uniform with Co-eficient of variation between 0.45 to 0.60% . Large chest Sircumtances is guaranteed the respiratory and cardiac conditions are well developed, will support the vitality of sprint racing pigeon.

Length of neck (LN) ranged from 8.87 – 8.46 cm with a CV range between 0.77 – 0.84%. length of neck will affected the aerodynamics of air during flight. The length of the neck formed of 13 segment of os. Vertebrae. The elasticity and flexibility of the neck will affected the time preparing flying and landing when they find a jockey who holds the female partner.

Length of the back (LB) of the sprint racing pigeon 1st to 5th championracing pigeon sprintranged between 14.61 – 14.43 centimeter, the 1st champion has the longest LB, and data has no significant, with a CV range between 0.38 – 0.54%. The length of the back consists of the caudal vertebrae, pygostyle, os synsacrum, scapula and lumbar vertebrae. This section is an essential part of the whole movement,look like a car chassis. In the framework of the backbone will stick to the inside of the air sac as well as kidneys from pigeons, so it will affect air circulation and filtering waste from the metabolism, so that the uric acid would be thrown away.

Distance of the os pubis (DoP) ranged from 1st to 5th champions, are ranged from 0.55 to 0.55 centimeter also has no significant with diversity level data uniformity, which is between 0.03 to 0.31%. DoP is useful supporting the flight, however, the pubic bone is very influential when pigeons is landing.

Body weight (BW) ranged from 384.64 to 388.92 grams, are non significant with a range between 7.78 to 9.88%. The 1st champion does not have the smallest BW, according to logically reason that the heavier body weight will be the stronger pull of gravity, so it

will have an influence on the flying speed of a sprint racing pigeon. Part of body size measurement and body weight is estimated to have a relationship with a flying speed.

Coeffisien correlasion (CC) between the flying speed with the body part are listed in Table 2 below:

Table 3. Coefisien of Relationship beetwen body part and flying speed of 1^{st}-5^{th} champion at race of local championship

Body Part	FS of 1^{st} Champion	FS of 2^{nd} Champion	FS of 3^{rd} Champion	FS of 4^{st} Champion	FS of 5^{st} Champion
LW (cm)	-0.091*	-0.041*	0.094*	0.177*	-0.102*
LoSW (cm)	0.035	-0.078	-0.047	0.045	-0.076
LoPFoW (cm)	0.269	0.005	0.032	0.049	0.054
DoBW (cm)	0.023	0.168	0.030	0.002	0.091
LoFoT (cm)	0.092	0.168	0.006	0.158	0.054
WoB (cm)	0.062	0.319	-0.096	-0.049	0.037
CC(cm)	0.014	0.000	-0.141	0.197	0.037
LN (cm)	0.029	0.099	-0.122	0.177	0.248
LB (cm)	0.010	0.099	0.037	0.028	-0.097
DoP (cm)	0.232	0.056	0.037	0.167	-0.074
BW (gram))	0.173	0.201	-0.048	0.010	-0.074

** Correlation is significant at the 0.01 level (2-tailed); * Correlation is significant at the 0.05 level (2-tailed)

In Table 2, it shows that coefisient correlasion (CC) between the body part with the flying speed in 1^{st} to 5^{th} champions are very low, although there is a negative and positive CC. Negative CC is the one variable increases the other variable that is correlated decline, while the CC value is positive when one variable increases followed by an increase in variable any others. LW has a negative CC with the FS on the 1^{st}, 2^{nd}, and 5^{th} champions. LoSW have a negative CC with FS in the 2^{nd} , 3^{rd} , and 5^{th} champions. WB has a negative CC with the FS on the 3^{rd} and 4^{st} champions. SB had a negative CC with FS in 3^{rd} champion. LB has a negative CC with the FS at 5^{th} champion. DoP have a negative CC with the FS on the 5^{th} champion. BW has a negative CC with the FS on the 3^{rd} and 5^{th} champion. So far that has CC a significant (significant at the level of 0.01 and 0.05) only between the LW with the FS on the 1st to 5^{th} champion.

LoPFoW and DooP have a high CC value (0.269 and 0.232) on the 1^{st} champion. The highest CC is the 2^{nd} champion between WoB, BW with FS (0.319 and 0.201). On the 5^{th} champion has a CC very high value between LN to FS (0.246).

The data in this study data were analysed using multiple linear regression backward method. The varable will analyzed one until reached significant levels, to determined the existence of linearity in the variable flying speed with the size of body parts as the independent variable. Multiple Linier Regression, R^2 and SE of 1^{st} – 5^{th} Champion at Race of Local showed in Table 3.

From Table 3, it shows that the Co-efficient of Determination (R^2) are <0.5 (between 0.18 to 0.34)means that there is low relationship between the part of the body size with a flying speed (Santosa, 2002). However, part of body size in sprint racing pigeon has no-significant among other, it means that the sprint racing pigeon has a proportional body shape.

Table 4. Multiple linier regression, R^2 and SE of 1^{st}- 5^{th} champion at race of local championship

Champion of Local Championship	Multiple Linier Regression	R^2	SE
1^{st} Champion	$\hat{Y}=143.82+0.22x_1-0.34x_2+0.39x_3+1.90x_4+0.49x_5+5.02x_6-1.99x_7+0.07x_8-0.07x_9+1.75x_{10}-0.25x_{11}$	0.26	3.30
2^{nd} Champion	$\hat{Y}=37.55-1.57x_1-0.15x_2+2.18x_3+3.97x_4+1.40x_5+5.72x_6-3.42x_7+0.33x_8-0.86\ x_9+3.18x_{10}+0.1x_{11}$	0.34	2.47
3^{rd} Champion	$\hat{Y}=130.07-0.1x_1-0.5x_2+1.5x_3+3.63x_4+0.45x_5-2.37x_6-1.92x_7-0.69x_8+6,51\ x_9-2.67x_{10}-0.16x_{11}$	0.18	2.89
4^{th} Champion	$\hat{Y}=11.29-0.52x_1+0.57x_2+0.01x_3-0.72x_4-0.21x_5-0.13x_6+2.37x_7+1.04x_8-1.33\ x_9+9.56x_{10}-0.11x_{11}$	0.18	3.18
5^{th} Champion	$\hat{Y}=29.38-0.1x_1-0.1x_2+1.42x_3+0.29x_4+0.45x_5+0.37x_6+0.42x_7-0.21x_8-0.14x_9+20.14x_{10}-0.04x_{11}$	0.21	2.12

Where: x_1 = LW , x_2= LoSW , x_3= LoPFoW, x_4= DW, x_5= LoFoT, x_6= WB, x_7= SB, x_8=LN, x_9= LB, x_{10}= LooP, X_{11}= BW and $\hat{Y}$ = estimation of Flying Speed.

Because there is no-significant correlation among others, so there are some factors that influence the flying speed, such as feeding, doping, training, disease control, environmental (wind), inteligent, and fly motivation , fly pattern and the jockey behavior.

Conclusions

Flying speed of the sprint race pigeon was not influenced by body part and body weight. Other factors, such as feeding, doping, drying, training, disease control, environmental (wind) and the behavior of the jockey, determined the champion achievement. Flying speed, body part and body weight and other factors were one unity.

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Performance of Sheep and Goat with Rotational Grazing Under Oil Palm Plantation Based on Animal Unit Equivalent (AUE)

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Abstract

The integration between livestock and oil palm plantation would play an important role for national meat self sufficiency and strengthening food security through optimization of decreasing land availability for livestock. It was estimated that more than 50% of oil palm plantation was owned by small farmer which was mostly prefer to keep sheep or goats rather than cattle. The objective of this research was to determine the effect of AUE (Animal Unit Equivalent) on performance of sheep or goats reared on rotational grazing under oil palm plantation. Total of 16 animals (8 sheeps and 8 goats) were used in this experiment. Eight sheeps were kept for two treatments of 1 AUE and 0.75 AUE, while the other eight goats were also kept for two similar treatments of 1 AUE and 0.75 AUE. Based on initial liveweight of the animals, two size of paddock (1 AUE and 0.75 AUE) were established to accomodate treatment of rotational grazing comprising 4 sub-paddocks grazed over 4 week-cycle from 08.00-16.00. The animals were housed at night; all animals were routinely feed with concentrate (1% of body weight) after being pastured under oil palm plantation. Parameters observed were body weight, ADG, body size (hearth girth, withers height and body length), and were all measured weekly for 8 weeks. Result indicated that AUE had significant effect (P<0.05) on average daily gain (ADG) in which ADG of goat was 26 gram day $^{-1}$ (0.75 AUE) and 2.3 gram day $^{-1}$ (1 AUE). Similar result was also found in sheep, reaching 39.6 gram day^{-1} (0.75 AUE) and 26.2 gram day^{-1} (1 AUE). It was found that there was no significant differences (P>0.05) on body size except height of withers within sheep. This study point out that under oil palm integration the performance of sheep showed better than those of goats.

Keywords: sheep goat performance oil-palm rotational grazing

Introduction

Sheep and goat (small ruminants) are highly valued as sources of income generating and asset of farmer in rural areas of developing countries. Therefore sheep and goats were documented have play an important role in food security, household stable, and economic transformation of farmer in developing countries such as in Indonesia, Thailand, Vietnam, Malaysia, Philiphina, and Kenya (Peischel, 2005; Ali, 2006; Gray and Wagner, 2006; Mui *et al.,* 2006; Sivasupramaniam, 2006; Villar, 2006; Waruiru, 2002, and Devendra 2007) and in Bangladest, goat known as a "*national treasure*" (Machmud, 2006)

Providing feed sufficient in quantity and quality throughout the year is the major constraint for ruminant developement in Indonesia.Decreasing land for livestock production mainly due to increasing land used for housing and plantation especially oil palm plantation. According to Chee and Faiz (1990) integration between animals and oil palm plantation have been associated with increased production of meat without the opening up of large

new areas of land because natural pasture under oil palm plantation is source of livestock feed. Moreover, the advantages integration of livestock-oil palm plantation were reduced cost of weeding control, reducing chemical fertilizer, speeded rate of nutrient cycle through urine and manure, minimize deterioration in soil fertility, increased crop yields, increased productivity per hectare as well as for draft animal power to carry fresh fruit brunch (Dwatmadji *et al*., 2004). System integration beef cattle with oil palm plantation is being started in Bengkulu since 2004, this system is aimed to increase the national meat self sufficiency through optimization of land use. It was estimated that more than 50% of oil palm plantation was owned by small farmer which was not suitable for rearing cattle, therefore goat or sheep could be more appropriate to this integration. The integration of goats and sheep with coconuts over three years increased the income of farmers in Philippines Integration between sheep and oil palm plantation has been practiced in Malaysia

According to Muir (2006) one of the most important factors influencing the productivity of animals grazed on pastured was stocking rate. Stocking rate can be calculated on the basis of animal number per hectare or on the basis of Animal Unit/hectare. The animal unit equivalent (AUE) concept is the most widely used to determine the carrying capacity of grazing animals on pastures, because AUE can be used to adjust stocking rates based on the initial live weight of livestock before entering to the paddock. The objective of this research was to determine the effect of AUE (Animal Unit Equivalent) on performance of sheep or goats reared rotational grazing under oil palm plantation.

Materials and Methods

This experiment was conducted in oil palm small holder enterprise at Pasar Pedati, Central Bengkulu. The ages of oil palms was 9 years and size of plantation was 5 ha. The natural pasture under oil palm plantation was a complex mixture of grasses pre dominantly *Axonopus compressus* , broad leaves, legume and fern. Two size of paddock (1 AUE and 0.75 AUE) was established to carry four animals based on initial live weight of 4 experimental animals, these paddocks then divided into 4 sub-paddock of equal size. All experimental animals were purchased from local seller and was dewormed before starting the experiment to be sure that all experimental animals were free from gastro intestinal parasites. The averaged of initial live weight of goat was about 7.2 kg and sheep was about 12 kg, the age goats and sheep was 5-7 months when grazing began. Eight sheep were divided two treatments of 1 AUE and 0.75 AUE, while the other eight goats were also kept for two similar treatments of 1 AUE and 0.75 AUE. All animals grazed rotationally over 4 week-cycle from 08.00-16.00. After being pastured under oil palm plantation the animals were housed at night and were routinely feed with concentrate (1% of body weight), and water were provided *at libitum*.

Parameters observed were body weight, ADG, body size (hearth girth, withers height and body length), all parameters measured in the morning before all animals entering the paddock. Data were collected weekly for 8 weeks. Heart girth, withers height, and body length were taken by a tape measure and body weight was taken using a digital scale. Withers height (WH) was measured as the distance from the floor to between the shoulders, body length (BL) was measured as distance from the occipital joint to the first caudal vertebra, heart girth (HG) was measured just behind the scapula. Two experienced persons carried out the body measurement, the average scores taken to avoid biasness since the exercise is subjective. Data were tabulated and expressed as mean and standard

deviation,. SPSS programmed for window version 15 was used for the statistical analyses especially for the data of ADG

Results and Discussion

The average of initial body or live weight of goats and sheep were 7.72 + 1.24 kg (1AUE) and 7.72 + 1.12 kg (0.75 AUE) and 12.09 + 3.09 kg (1 AUE) and 12.29 + 2.09 kg respectively. While the average of final live weight of goats and sheep were 7.68 + 0.98 kg vs 8.26 + 1.12 kg and 12.69 + 3.47 vs 13.12 + 2.78 kg. It was found that the live weight of goats tend to decrease until week 5 and then slightly increase from week 6 up to the end of the experiment. While, live weight of sheep increase gradually either in 1 UAE or 0.75 AUE.

Table 1 showed that AUE had significant effect ($P<0.05$) on average daily gain. It was calculated that average daily gain (ADG) of goat was 26 gram day -1 (0.75 AUE) and 2.3 gram day -1 (1 AUE). Babayemi and Bamikole (2006) found that weight gain of grazing goat was 7.74-27.38g/day depend on the quality of pasture. Result also indicated that ADG of sheep was higher than that of ADG of goat, reaching 39.6 gram day^{-1} (0.75 AUE) and 26.2 gram day^{-1} (1 AUE). Pasture production seems influenced the ADG both in goats and sheep. de Araújo Filho and Araújo Crispim (2002) and Haji Baba *et al* (1998) also reported that sheep had better daily weight gain than those recorded in goats.

Table 1. Average of live weight gain (gram) per week of goats and sheep with different AUE

Week	Goat		Sheep	
	0.75 AUE	1 AUE	0.75 AUE	1 AUE
1	86.7 ± 1.17	-37.7 ± 1.24	187.3 ± 3.09	108.7 ± 0.97
2	115.0 ± 1.12	-72.3 ± 1.46	150.3 ± 3.22	160.7 ± 0.99
3	220.3 ± 1.00	-51.0 ± 1.20	237.3 ±3.24	143.0 ± 0.76
4	176.3 ± 1.25	40.0 ± 2.00	180.3 ± 3.29	242.7 ± 1.89
5	165.7 ± 0.25	57.7 ± 2.18	346.7 ± 3.32	260.7 ± 1.59
6	266.3 ± 1.96	94.6 ± 1.24	391.0 ± 3.25	201.3 ± 1.72
7	246.0 ± 2.25	63.3 ± 2.89	413.0 ± 3.36	228.0 ± 1.53
Average	182.3 ± 1.35	16 ± 1.14	277.5 ± 1.52	192.1 ± 1.29
ADG (g/d)	26.00^{b}	2.30^{c}	39.60^{a}	26.20^{b}

^{abc} mean in same colum differ significantly ($P<0.05$)

Moreover, Haji Baba *et al.* (1998) also reported that the ADG of sheep grazed under oil palm plantation in Malaysia were 23.14 g/day (non supplemented) 31.71 g/day (supplemented 3% concentrate) and ADG of goats were 21.96 g/day (non supplemented) 26.18g/day (supplemented). According to Budisatria *et al.* (2010), the performance of sheep and goats depend on the agro-ecological condition (the lowlands, the middle zone, and the uplands). In middle and uplands which high quality of leaves was abundant, goats grew significantly faster than sheep (69 g d−1 vs. 65 g d−1). The differences of ADG between sheep and goats in our research could be due to the differences of feeding behavior between two animals and the quality of natural pasture under oil palm plantation. Dwatmadji *et al.* (2009) found that crude protein content of broad leaves a quite high; however the presence of secondary metabolite compound could affect the palatability and digestibility. Fajemisin *et al.* (1996) said that goat will only browse the most nutritious feed available. According to Aregheore (1998) unlike other ruminant animals, goats are selective, and the selectivity depend on the quality and quantity of the pasture (Quick and. Dehority,

1986). Moreover, Quick and Dehority (1986) said that sheep and goats have differences in intrinsic digestive function and capacity.

The average of the body size (hearth girth, withers height and body length) can be seen in Table 2-4 below. According to Olatunji-Akioye, and Adeyemo. (2009) and Pesmen and Yardimci (2008) chest or hearth girth is a useful tool that can be used to predict weight of commercial sheep and goat goats. However in our research, due to limiting experimental animal used the correlation between hearth girth and live weight were not calculated.

Average of wither height (cm) per week of goats and sheep with different AUE can be seen in Table 3. It was found that height of withers of sheep (1 AUE vs. 0.75 AUE) was significantly ($P < 0.05$) different. Pesmen and Yardimci (2008) found that there was no correlation between wither height and live weight ($r=0.225$).

Table 2. Average of hearth girth (cm) per week of goats and sheep with different AUE

Week	Goat		Sheep	
	0.75 AUE	1 AUE	0.75 AUE	1 AUE
1	43.67 ± 2.66	43.67 ± 1.53	49.33 ± 3.09	52.33 ± 4.50
2	42.17 ± 3.59	41.67 ± 3.21	48.67 ± 2.51	52.00 ± 3.54
3	42.33 ± 2.66	42.00 ± 1.00	49.00 ±2.57	51.67 ± 5.02
4	41.67 ± 2.81	42.33 ± 2.00	50.33 ± 2.77	52.00 ± 4.00
5	44.00 ± 4.72	43.67 ± 2.08	54.67 ± 2.11	52.67 ± 5.02
6	45.33 ± 3.65	44.00 ± 1.00	56.00 ± 2.29	52.83 ± 5.00
7	45.34 ± 3.81	45.00 ± 1.00	56.00 ± 2.24	53.50 ± 1.53
8	46.00 ± 1.53	46.00 ± 1.53	57.00 ± 1.53	53.83 ± 4.78
Average	43.81 ± 2.48	43.33± 2.12	52.63 ± 2.16	52.60 ± 3.06

Table 3. Average of wither height (cm) per week of goats and sheep with different AUE

Week	Goat		Sheep	
	0.75 AUE	1 AUE	0.75 AUE	1 AUE
1	38.67 ± 3.93	40.17 ± 2.08	45.00 ± 3.20	45.53 ± 2.51
2	39.17 ± 3.62	40.33 ± 1.89	46.00 ± 2.61	45.67 ± 3.54
3	39.30 ± 3.79	40.33 ± 1.83	47.83 ±2.25	46.00 ± 3.61
4	39.36 ± 3.69	40.83 ± 2.93	48.33 ± 2.03	46.67 ± 3.16
5	39.37 ± 3.06	41.00 ± 2.65	49.00 ± 2.00	47.33 ± 3.02
6	40.00 ± 3.46	41.00 ± 2.65	4900 ± 2.00	47.50 ± 3.27
7	41.00 ± 2.65	41.17 ± 2.36	49.33 ± 2.51	47.66 ± 3.51
8	41.33 ± 2.89	41.83 ± 2.47	49.50 ± 2.77	48.00 ± 3.27
Average	39.73 ± 3.17	40.83± 2.08	48.01 ± 2.15	46.40 ± 3.08

The wither height of goats and sheep in this research were about 38.67-41.83 cm and 45-49.5 cm respectively (Table 3). Utami (2008) reported that wither height of local sheep is about 46.8-49.9 cm. While Pesmen and Yardimci (2008) found that coefficient correlation between wither height and live weight of Saanen goat was 0.549.

Table 4. Average of body length (cm) per week of goats and sheep with different AUE

Week	Goat		Sheep	
	0.75 AUE	1 AUE	0.75 AUE	1 AUE
1	26.33 ± 3.06	28.67 ± 0.58	45.00 ± 3.20	52.33 ± 4.50
2	27.00 ± 3.46	28.33 ± 1.15	46.00 ± 2.61	52.00 ± 3.54
3	28.00 ± 2.65	28.00 ± 1.73	47.83 ±2.25	51.67 ± 5.02
4	28.00 ± 2.65	28.67 ± 0.58	48.33 ± 2.03	52.00 ± 4.00
5	28.33 ± 3.06	29.00 ± 1.00	49.00 ± 2.00	52.67 ± 5.02
6	28.67 ± 2.52	29.33 ± 2.08	4900 ± 2.00	52.83 ± 5.00
7	29.66 ± 3.06	30.32 ± 2.07	49.33 ± 2.51	53.50 ± 1.53
8	30.00 ± 2.65	31.00 ± 1.73	49.50 ± 2.77	53.83 ± 4.78
Average	28.25 ± 2.69	29.17± 1.58	48.01 ± 2.15	52.60 ± 3.06

Average of body length (cm) per week of goats and sheep can be seen in Table 4. According to Pesmen and Yardimci (2008) body measurements including body length differ according to the factors such as breed, gender, yield type and age.

Conclusion

Natural pasture under oil palm plantation can support the performance of sheep and goats depending on the animal unit equivalent. It was also found that sheep had better performance than those recorded in goats.

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FOOD TECHNOLOGY

Optimization of Fish Gelatin Extraction from Starry Triggerfish (*Abalistes stellaris*) Skin

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Abstract

Response Surface Methodology (RSM) was adopted to establish the optimum condition in gelatin extraction process from starry triggerfish skin. In this study, 30 experiments were performed using a four factor, 3-levels Central Composite Design (CCD). In this study, a combination of both alkali and acid pre-treatment were applied. Gelatin extraction was carried out using hot-water extraction. The effects of four variables which were NaOH concentration (X_1, M) and acetic acid concentration (X_2, M) during pre-treatment, and extraction temperature (X_3,˚C) and time during (X_4, min) extraction with three levels for each were studied in order to obtain the maximum gelatin yield and gel strength. RSM analysis suggested that the maximum gelatin yield and gel strength could be obtained at a 0.1 M NaOH and 0.05 M acetic acid, temperature of 67.82˚C and extraction time of 120 minutes. Gelatin yield could be predicted using quadratic model, while gel strength could be predicted using linear model. The experimental value for gelatin was 8.09 % and 252.73g for gel strength, while the predicted value was 7.48 % and 252.77 g for gelatin yield and gel strength respectively. This study shows that RSM could be used to predict gelatin extraction from starry triggerfish.

Keywords : fish skin, gelatin, starry triggerfish, response surface methodology

Introduction

Gelatin is a denatured protein derived from collagen by thermo-hydrolysis and has a thermo-reversible transformation between sol and gel (Cho *et al*., 2004). Gelatin regarded to an important, functional and good source of biopolymer (Kasankala *et al*., 2007; Zhou and Regenstein, 2005; Cho *et al*., 2004) with special and unique hydrocolloid for wide range of applications in various industries (Karim and Bhat, 2009) such as food, pharmaceutical, photographic and cosmetics industries.

Gel strength, viscosity, and melting point are the most important properties characterizing gelatin for food applications and factors affected these properties are concentration of the gelatin solution, gel maturation temperature, pH and salt content (Choi and Regenstein, 2000). In the extraction process, the factors influencing the length of the polypeptide chains and the gelatin functional properties are processing parameters which are temperature, time and pH, the pre-treatment, and the properties and preservation method of the stating raw material (Karim and Bhat, 2009).

The main origins of commercial gelatin are derived from mammalian sources mainly porcine or bovine (Gudmundsson, 2002). The increasing research to find alternative sources of gelatin is motivated by several issues such as religious restrictions of Judaism and Islam to consume porcine and bovine gelatin (Choi and Regenstein, 2000), religious restriction of Hindus to consume bovine gelatin and concerns over bovine spongiform encephalopathy (BSE) crisis (Zhou and Regenstein, 2005). Furthermore, the by-products of fishery industries

such as fish skins, bones, scales and swim bladders can yield additional income both economic and waste management benefits for fish industry because of the large quantities of these materials generated (Choi and Regenstein, 2000).

An optimization of the extraction procedures and a better knowledge of the properties of fish-gelatin could be helpful in rationalizing the use of fish residues (Gomez-Guillen *et al*., 2002). Starry triggerfish skin is always removed prior to filleting or cooking. Thus, it is a potential source of fish gelatin. Besides that, the use of fish skin to produce gelatin may reduce the disposal and environmental problem from fisheries waste. The aim of this study is to determine the optimum conditions to get the maximum yield and gel strength of starry triggerfish gelatin by using Response Surface Methodology (RSM).

Materials and Methods

Materials

Starry triggerfish (*Abalistes stellaris*) were purchased from a local supplier in Kuala Terengganu. All chemicals and reagents used were of analytical grades.

Methods

Raw materials preparation

Whole starry triggerfish was beheaded and eviscerated prior to filleting. Then the skin was removed from the fillet. The skin was washed to remove any contaminants. The skin was cut into small size about 1 cm^2 and washed with tap water to remove any contaminants. Then the skin was packed in plastic bags (30 g per plastic bag) before they were stored and frozen at -40˚C until further use.

Experimental design

In order to establish the optimum condition for gelatin extraction from Starry triggerfish skin, Response Surface Methodology (RSM) was used in this study. Four independent variables which were concentration of NaOH (M), concentration of acetic acid (M), extraction temperature (˚C) and extraction time (hours); with three levels (-1, 0 and 1) was examined in this experiment. Two response variables were chosen i.e. gelatin yield and gel strength. The experimental design used was Central Composite Design (face centered). Table 1 shows the experimental ranges and values of the independent variables used in this study.

Table 1. Experimental range and values of the independent variables for gelatin extraction

Independent variables	Symbol	Range and levels		
		-1	0	1
Concentration of NaOH (M)	X1	0.1	0.2	0.3
Concentration of acetic acid (M)	X2	0.05	0.13	0.2
Extraction temperature (˚C)	X3	40	55	70
Extraction time (minutes)	X4	120	180	240

Pre-treatment

The pre-treatment and gelatin extraction was carried out as described by Yang *et al*. (2007) with slightly modification. A cleaned skin (30 g) was added to a flask and was treated with NaOH (1:6 w/v) with varying concentration (factor X_1, mol/L) for 60 minutes. Then, the sample was drained and rinsed with tap water using muslin cloth. The above procedures were repeated 2 times. Afterwards the samples were treated with varying concentration of acetic acid (1:6 w/v, factor X_2, mol/L) for 45 minutes. The sample was then drained using muslin cloth and rinsed with tap water (1:6 w/v) for 3 times. All the solutions used in the above steps were kept at 4˚C.

Gelatin extraction

After the above pre-treatment procedures, distilled water (the ratio of sample to water was 1:4, w/w) was added to the flasks. Aluminum foil was used to cover the flasks and sample was extracted in a water bath at different extraction temperatures (factor X_3, ˚C) and extraction time (factor X_4, minutes), depending on the design. Then, the gelatin solution was centrifuged and filtered by muslin cloth. Finally, the gelatin solution was freeze dried prior to determination of gelatin yield and gel strength.

Determination of gelatin yield

Calculation of gelatin yield was employed as described by Liu *et al*. (2009). The gelatin yield was calculated as follows:

Gelatin Yield (%) = Weight of dried gelatine (g) / Weight of skin used (g) x 100

Determination of gel strength

Determination of gel strength was carried out using the procedure described by Norziah *et al*. (2009) with some modification. The gelatin gel was prepared by dissolving 6.67% (w/v) dried gelatin in distilled water and then heated at 60˚C in a shaker water bath for 30 minutes until gelatin was completely dissolved. The gelatin solution then cooled to 25˚C and poured into Bloom jar. Then it was left in refrigerator at 5-7˚C for 16-18 hours prior to analysis.

Texture analyzer TA XT2 (Stable Microsystems, Surrey, UK) was used to determine the gel strength with load cell of 5 kg and cross-head speed of 1 mm/s. A flat bottom plunger of 0.5 in diameter was used. The bloom value (g) was obtained after the plunger penetrates into the gel to a depth of 4 mm. At this depth, the maximum force reading (the resistance of penetration) was obtained and translated as the bloom strength (g) of the gel.

Statistical analysis

Experimental data obtained in the optimization of gelatin extraction from starry triggerfish using Response Surface Methodology were analyzed using Design Expert Version 8.0.4 (Stat Ease Inc.). The software generated the experimental runs and automatically analyzed the experimental data and gave the important analysis including model adequacy checking, sequential model sum of squares, ANOVA table, final equation, diagnostic case statistics, response surface plots, suggested solutions, and point prediction table. Data on response variables i.e. gelatin yield and gel strength were stated as mean±standard deviation.

Results and Discussion

Table 2 shows the experimental data obtained in this study. The range of gelatin yield and gel strength obtained was 0.4% - 10.1% and 0 - 338.3 g, respectively. The highest gelatin yield obtained from starry triggerfish skin was 10.1% while the highest gel strength was 338.3 g.

In order to analyze the experimental data, Design-Expert version 8.0.4 software was used in this study. Firstly, model adequacy checking was carried out to examine whether the fitted model provides an adequate approximation to the true system. Then, the data for each response was analyzed separately for analysis of variance (ANOVA).

After each response had been analyzed, the optimization was carried out by combining both responses. The purpose of optimization module in Design-Expert software is to search for a combination of factors that simultaneously satisfy the requirement placed on each of the responses and factors.

Model adequacy checking

The experimental data obtained were analyzed for model adequacy checking to the responses, i.e. gelatin yield (GY) and gel strength (GS). The software has suggested a quadratic model for gelatin yield and a linear model for gel strength.

The suggested quadratic model for gelatin yield was $GY = +6.42 – 0.30*A + 3.11*C + 1.70*A^2 – 4.10*C^2$, while for gel strength was $GS = +148.78 + 121.63*C$.

Analysis of variance (ANOVA)

Table 3 shows the ANOVA table for gelatin yield and gel strength, respectively (which was obtained after model reduction had performed). It is necessary to examine the fitted model to ensure adequate approximation is provided to the true system (Myers and Montgomery, 2002).

The “F-value” for both models was 87.67 and 59.34 respectively, indicating that the model is significant and there is only 0.01% chance that the model could occur due to noise. Meanwhile the “p-value” less than 0.05 implied that the models terms were significant. However, for gelatin yield response; model term A with “p-value” more than 0.05 was not reduced because it was required to support hierarchy. Since model term A^2 is significant, A must be included in the model. So, the significant model terms in this study for gelatin yield response are model terms A, C, A^2, and C^2. Meanwhile, the significant model term for gel strength is only model term C.

Table 2. The experimental data obtained for gelatin extraction from starry triggerfish skin using Central Composite Design (CCD) (face-centered)

Std	Run	Factor 1	Factor 2	Factor 3	Factor 4	Response	
		NaOH conc.(M)	CH_3COOH conc. (M)	Extraction temp. (˚C)	Extraction time (min)	Gelatin yield (%)	Gel strength (g)
15	1	0.10	0.20	70	240	6.9	198.96
20	2	0.20	0.20	55	180	6.6	100.00
16	3	0.30	0.20	70	240	7.7	243.28
24	4	0.20	0.13	55	240	5.9	171.65
1	5	0.10	0.05	40	120	0.4	0.00
2	6	0.30	0.05	40	120	0.4	0.00
22	7	0.20	0.13	70	180	4.9	285.74
3	8	0.10	0.20	40	120	1.1	11.85
30	9	0.20	0.13	55	180	7.1	20.06
7	10	0.10	0.20	70	120	7.7	334.23
27	11	0.20	0.13	55	180	6.5	153.47
26	12	0.20	0.13	55	180	7.0	178.88
28	13	0.20	0.13	55	180	6.4	224.8
9	14	0.10	0.05	40	240	0.4	0.00
29	15	0.20	0.13	55	180	6.8	133.73
18	16	0.30	0.13	55	180	7.0	242.07
10	17	0.30	0.05	40	240	0.5	0.00
8	18	0.30	0.20	70	120	5.4	287.98
4	19	0.30	0.20	40	120	0.9	45.91
21	20	0.20	0.13	40	180	0.6	17.48
14	21	0.30	0.05	70	240	7.6	185.35
19	22	0.20	0.05	55	180	4.2	128.52
5	23	0.10	0.05	70	120	7.6	246.95
11	24	0.10	0.20	40	240	1.2	25.35
17	25	0.10	0.13	55	180	10.1	40.45
25	26	0.20	0.13	55	180	6.2	284.03
23	27	0.20	0.13	55	120	6.6	338.30
12	28	0.30	0.20	40	240	1.0	28.49
13	29	0.10	0.05	70	240	7.6	234.17
6	30	0.30	0.05	70	120	7.1	301.68

Table 3. ANOVA table for response of gelatin yield and gel strength

Source	Sum of Squares	df	Mean Square	F Value	p-value Prob > F	
Gelatin yield (%)						
Model	245.00	4	61.25	87.67	< 0.0001	significant
A- NaOH conc.	1.62	1	1.62	2.32	0.1404	
C- extraction temp.	174.22	1	174.22	249.38	< 0.0001	
A^2	9.99	1	9.99	14.30	0.0009	
C^2	57.81	1	57.81	82.75	< 0.0001	
Residual	17.47	25	0.70			
Lack of Fit	16.83	20	0.84	6.64	0.0225	significant
Pure Error	0.63	5	0.13			
Cor Total	262.47	29				
Gel strength (g)						
Model	2.663E+005	1	2.663E+005	59.34	< 0.0001	significant
C- Extraction temp.	2.663E+005	1	2.663E+005	59.34	< 0.0001	
Residual	1.256E+005	28	4486.85			
Lack of Fit	85580.74	23	3720.90	0.46	0.9049	Not significant
Pure Error	40051.08	5	8010.22			
Cor Total	3.919E+005	29				

Optimization

For optimization, the goal for independent variables for NaOH concentration (X_1) and extraction temperature (X_3) had been set "to be in range". Since the other two independent variables, i.e. CH_3COOH concentration (X_2) and extraction time (X_4) did not affect both responses, it had been set to the lower limit. The lower limit for CH_3COOH concentration and extraction time was chosen to reduce chemical cost and extraction time. Meanwhile, both responses were set at "maximize". The optimum conditions obtained were 0.1M of NaOH and 0.05M of CH_3COOH during pre-treatment, 67.82°C of extraction temperature and 120 minutes of extraction time with the predicted value of 7.48% for gelatin yield and 252.77 g for gel strength.

The experimental values for gelatin yield and gel strength were 8.09% and 252.7g, respectively under the optimal operational conditions. This result is in close agreement with the predicted value at optimum conditions as mentioned above.

In this research, a combination NaOH and CH_3COOH were used in the pre-treatment step before hot-water extraction. According to Zhou and Regenstein (2005), alkaline pre-treatment followed by an acid treatment was found to remove unwanted material such as non collagenous protein with minimum collagen loss and securing a high gelatin yield and good gel strength.

The resulted quadratic model suggested for gelatin yield from starry triggerfish skin was in agreement with previous study on optimization of gelatin from yellowfin tuna skin (Cho *et al*., 2005) and shark cartilage (Cho *et al*., 2004). However, this finding was in contrast with previous study on optimization of gelatin from cobia skin (Amiza *et al*., 2009), lizardfish scale (Wangtueai and Noomhorn, 2008), channel catfish skin (Yang *et al*., 2007) and pollock

skin (Zhou and Regenstein, 2004), whereby for these studies, both linear and quadratic model could predict the gelatin yield response.

Meanwhile, the linear model for gel strength was not in agreement with other previous study on optimization of gel strength from fish gelatin. In the optimization of fish gelatin extraction from lizardfish scales and pollock skin, both linear and quadratic models could be used to predicted gel strength model (Wangtueai and Noomhorm, 2008; Zhou and Regenstein, 2005). Meanwhile, optimization of fish gelatin from grass carp fish skin and yellowfin tuna skin could be predicted by quadratic model only (Kasankala *et al*., 2007; Cho *et al*., 2005). While Yang *et al*., 2007 reported that no model could predict the gel strength of gelatin from channel catfish skin. This difference in prediction model for gel strength could be due to differences in raw materials, pre-treatments and extraction condition used.

The gelatin yield obtained from starry triggerfish skin is in similar range with that of tilapia (5% to 8%) (Jamilah and Harvinder, 2002). However, the gelatin yield obtained were lower compared to those from cod (14%) (Gudmundsson and Hafsteinsson, 1997), pollock skin (18%) (Zhou and Regenstein, 2004), lizardfish scales (10.97%) (Wangtueai and Noormhorm, 2008), grasscarp fish skin (19.83%) (Kasankala *et al*., 2007), yellowfin tuna skin (89.7%) (Cho *et al*., 2005), shark (79%) (Cho *et al*., 2004) and cobia skin (19.90%) (Amiza *et al*., 2010). The gelatin yield from starry triggerfish skin was higher compared to hake skin (6.5%) (Gomez-Guillen *et al*., 2002). The lower extraction yield of fish gelatin could be due to the loss of extracted collagen by leaching during washing step and incomplete hydrolysis of the collagen (Jamilah and Harvinder, 2002) and protein degradation (at higher temperature) (Cho *et al*., 2005).

For gel strength, the maximum value for starry triggerfish gelatin was 252.73g. Catfish gelatin also gave similar gel strength of 252 g (Yang *et al*., 2007). However, the gel strength of starry triggerfish gelatin was higher than those reported from red tilapia gelatin (128.1 g) and black tilapia gelatin (180.7%) (Jamilah and Harvinder, 2002) and Alaskan pollock gelatin (98 g). Lower gel strength of fish gelatin has been reported for grass carp (267 g) (Kasankala *et al*., 2007) and yellowfin tuna (426 g) (Cho *et al*., 2005). According to Karim and Bhat (2009), the variation in Bloom values found for various gelatin arises from the differences in proline and hydroxyproline contents for different species and associated with the temperature of the habitat of the animal. Yang *et al*. (2007) reported in optimization of the extraction channel catfish skin gelatin, the gel strength decreased when the temperature increased. They stated the high extraction temperature caused protein degradation and denaturation which in turn, produced small protein fragments and a lower gelling ability. Badii and Howell (2006) also reported similar finding.

Conclusion

The optimum conditions for gelatin extraction from starry triggerfish skin were 0.1 M NaOH, 0.05 M acetic acid (CH_3COOH) during pre-treatment, extraction temperature of 67.82˚C and extraction time of 120 minutes. The predicted values for both responses were 7.48% for gelatin yield and 252.77 g for gel strength. Under these optimum conditions, the experimental values for gelatin yield and gel strength was 8.09% and 252.7 g, respectively. This study shows that RSM could be used to predict gelatin extraction from starry triggerfish.

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Optimization of Gelatin Extraction Parameters from Cobia *(Rachycentron canadum)* Skin

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Abstract

This study reported the optimization of gelatin extraction from cobia (Rachycentron canadum) skin. The effect of four independent variables which were acetic acid concentration, skin to water ratio, extraction temperature and extraction time on gelatin yield and gel strength were determined using Response Surface Methodology (RSM). The generated models gave a linear fit for gelatin yield and a quadratic fit for gel strength. The optimum conditions for gelatin extraction from cobia skin were 0.15mol/L for acetic acid concentration, 82.4^{o}C of extraction temperature, 6 h of extraction time and 1:6 of skin to water ratio. The predicted responses (20.59% yield and 226.86g gel strength) closely matched the experimental yield of 20.10% and gel strength of 205.6g. RSM could be used to predict cobia gelatin extraction.

Keywords : Cobia, fish skin, gelatin, response surface methodology

Introduction

Gelatin was denatured form of collagen derived from the fibrous protein (Karim & Bhat, 2009) which is produced by partial hydrolysis of nature collagen (Norziah *et al*., 2009). Gelatin source is commonly from bovine, porcine and fish. The interest in fish gelatine production has icreased significantly growth compared to the other sources for religious and social reason. So far, several studies of gelatine from various species of fish have been published either from cold water fish e.g. cod (Gudmundsson, 2002; Arnesen & Gildberg, 2006), pollock (Zhou & Regenstein, 2004), salmon (Arnesen & Gildberg, 2007) or warm water fish e.g Nile perch (Muyonga *et al*., 2004), giant grouper and golden snapper, (Irwandi *et al*., 2009).

However, the utilization of fish gelatin is apparently limited due to its low quality such as dark colour, strong odor (Montero & Gomez-Guillen, 2000) low gel strength and melting point. Previous researchers have reported that certain parameters involved in the processing of gelatine such as temperature, time, pH, concentration of solution during pre-treatment and extraction will affect the quality and quantity of the final product (Gomez-guillen & Montero, 2001; Karim & Bhat, 2009). It is therefore very important to thoroughly control all the processing parameters in order to obtain optimum extraction conditions thus improve the rheological properties as well as maximize the yield of gelatine.

The aim of this study is to optimize extraction conditions for cobia skin gelatin with Response Surface Methodology (RSM). In food industry, RSM has important application in the design, development and formulation of new products, as well as in the improvement of existing product design (Granato *et al*., 2011). In this study, cobia *(Rachycentron canadum)* skin was chosen as raw material to produce gelatin. Impressive growth performance coupled with great fresh quality that is suitable for sashimi, tempura, broiled,

fried or steamed dishes makes the market demand for this species increases especially in Taiwan, China and Japan (Kaiser & Holt, 2005).

Materials and Methods

Materials

Marine cultured cobia was purchased from Langkawi Island, Kedah, Malaysia. All the chemicals and reagents used in this study were of analytical grade.

Raw materials preparation

Fresh cobia were eviscerated and filleted, and its skin were separated from the fillet. The fresh cobia skin were packed in polyethylene and kept frozen (-40^{o}C) until required for extraction. Bovine gelatine (230 blooms) and porcine gelatine (300 blooms) were purchased from Sigma Chemical Co. (St. Louis, MO, USA).

Gelatin extraction during optimization

All the method used during gelatine extraction was based on Zhou & Regenstein (2004). Thawed skin (40g) was treated with acetic acid at different concentration (factor A) for 1 hour at 4^{o}C. The treated skin were drained and rinsed for two times. Then, the sample were mixed with distilled water at total ratio of skin/ water (factor D) followed by extraction at temperature (factor D) for a period of time (factor C).

Experimental design

Response surface methodology (RSM) was applied to optimize the extraction parameters by using a commercial statistic package Design-Expert version 8.0.4 (Stat Ease Inc., Minneapolis, U.S.A). A Central Composite Design (face-centered) was employed with four independent variables (A, concentration of acetic acid; B, extraction temperature; C, extraction time and D, skin/water ratio) that were coded to three levels -1, 0 and +1. Yield of gelatin and gel strength were the dependent variables. The experimental design consists of 30 points with 16 factorial points, 8 axial points and six replicates of the centre point. The experimental design used is shown in the Table 1.

Table 1. Factors and levels fot two-level fractional factorial design.

Independent variable	symbol	Level		
		-1	0	+1
Concentration of acid (mol/L)	A	0.05	0.10	0.15
Extraction temperature (oC)	B	50	75	100
Extraction time (min)	C	1	4.5	8
Skin / water ratio (w/v)	D	½	¼	1/6

Gelatin yield

Calculation of gelatin yield was applied as described by Liu *et al*. (2009). The gelatin yield calculated as followed: Yield (%) = Weight of dried gelatine (g) x 100/ Weight of skin used (g)

Gel strength

Determination of gel strength was carried out using the procedure described by Norziah *et al*. (2009) with slight modification. The gelatin gel was prepared by dissolving 6.67% (w/v) dried gelatin in distilled water at room temperature for 30 min and then heated at 60˚C in a water bath shaker for 30 minutes until it was completely dissolved. The gelatin solution was then cooled to 25˚C and poured into Bloom jar. Then it was left in refrigerator at 5-7˚C for 16-18 hours prior to analysis. Gel strength was determined on Texture analyzer TA XT2 (Stable Microsystems, Surrey, UK) according to British Standard BS 757 (BSI, 1975).

Results and Discussion

Table 2. Experimental design and results for gelatin extraction

Run	Factors (actual value)				Response	
	A:Concentration of acid (mol/L)	B:Extraction temperature (°C)	C:Extraction time (hr)	D: Skin/ water ratio	GY (%)	GS (g)
1	0.10	75.0	4.5	2.0	15.79	203.00
2	0.05	75.0	4.5	4.0	15.99	119.30
3	0.10	75.0	1.0	4.0	9.40	300.20
4	0.05	50.0	1.0	6.0	6.20	117.10
5	0.05	50.0	8.0	6.0	11.40	28.20
6	0.10	75.0	4.5	4.0	12.00	237.84
7	0.05	100.0	8.0	2.0	18.40	44.50
8	0.10	75.0	4.5	4.0	12.36	243.87
9	0.15	100.0	8.0	2.0	21.00	86.80
10	0.15	100.0	1.0	6.0	12.40	228.10
11	0.15	50.0	8.0	6.0	15.40	111.10
12	0.05	100.0	8.0	6.0	29.06	73.50
13	0.15	75.0	4.5	4.0	17.79	199.20
14	0.15	100.0	1.0	2.0	21.59	284.60
15	0.10	75.0	4.5	6.0	18.60	241.80
16	0.05	100.0	1.0	2.0	11.42	270.40
17	0.05	100.0	1.0	6.0	24.05	67.60
18	0.15	50.0	8.0	2.0	14.99	41.30
19	0.15	50.0	1.0	6.0	7.00	110.10
20	0.10	75.0	4.5	4.0	12.66	253.33
21	0.15	100.0	8.0	6.0	33.99	45.50
22	0.10	75.0	8.0	4.0	20.40	124.80
23	0.05	50.0	8.0	2.0	12.10	2.50
24	0.10	75.0	4.5	4.0	12.33	316.58

25	0.05	50.0	1.0	2.0	4.40	16.50
26	0.10	75.0	4.5	4.0	12.66	219.92
27	0.10	50.0	4.5	4.0	10.62	49.90
28	0.10	75.0	4.5	4.0	12.33	292.32
29	0.10	100.0	4.5	4.0	23.59	110.70
30	0.15	50.0	1.0	2.0	4.80	149.90

Based on the result of the screening process that had been performed before optimization, four independent factors were chosen for optimization i.e. concentration of acetic acid (A: 0.05-0.15 mol/L), extraction temperature (B: 50-100^{o}C), extraction time (C: 1 – 8 hrs), skin to water ratio (D: 1:2 – 1:6) and 2 responses i.e. gelatin yield (GY) and gel strength (GS). Acetic acid was used for pre-treatment as preliminary studies showed that acetic acid could minimize the loss of collagen in fish skin. According to Gomez-Guillen and Montero (2001), small molecular size and low ionization of the acetic acid molecule have been proposed as a key factor for a suitable collagen swelling prior to conversion into gelatin.

The relationships between these four factors were studied in 30 runs of experiment using Central Composite Design (face-centered). The experimental conditions and its responses were shown in Table 2. The experimental sequence was randomized in order to minimize error.

Based on the result obtained, it was found that GY and GS ranged from 4.4% to 33.9% and 0.0g to 316.58g respectively. The results obtained were then further analyzed by following these steps; model fitting, analysis of variance (ANOVA), model diagnostic plot, response surface plot, optimization and verification.

Model adequacy checking

The software automatically analyzed the experimental data obtained and suggested the suitable models for both responses, i.e. gelatin yield (GY) and gel strength (GS). In this study, transformation of data was applied. This transformation applied a mathematical function to all the response data in order to meet the assumptions that made the ANOVA valid. Hence a Base 10 log and Square Root was applied to these responses. Based on the result, the linear model was suggested for GY whereas quadratic model was suggested for GS.

Analysis of variance (ANOVA)

It was necessary to examine the fitted model to ensure adequate approximation is provided to the true system (Myers and Montgomery, 2002). The significance of terms for each model was judged by using p-value, R-squared value and “lack of fit” value. The p-value for a model should be less than 0.05 to be strongly significant and between 0.05 and 0.10 is marginally significant. Table 3 shows the ANOVA table obtained after model reduction had performed (i.e. after insignificant terms with p-values > 0.05 were removed).

For GY, the model F-value is 38.44 and the p-values is <0.0001 indicating that the model is significant. The lack-of-fit ($p \leq 0.05$) of the model indicated that the quadratic model may not be suitable to predict gelatin yield. However, an analysis which involved a large amount of data, a model with a significant lack of fit could still be used (Pua *et al*., 2007). Furthermore

the goodness of the model is and then relied on confirmation runs to validate the experimental results, not the value of the lack of fit. The F-value of the model for GS is 24.39 and the p-value is <0.0001, respectively which implies that the model is significant. Linear terms , acetic acid concentration (A), extraction temperature (B) and extraction time (C), quadratic terms of B^2 and interaction terms BD showed significant effect on gel strength. "Lack of Fit value" of 0.0671 implies that the model fits well with the experimental data.

Table 3. ANOVA for the regression model and the respective model terms

Source	Sum of Squares	df	Mean Square	F Value	Prob > F
Gelatin Yield (GY) (%)					
Model	1.03	3	0.34	38.38	<0.0001
B	0.62	1	0.62	68.82	<0.0001
C	0.37	1	0.37	41.72	<0.0001
D	0.041	1	0.041	4.59	0.0417
Residual	0.23	26	8.950E-003		
Lack of Fit	0.23	21	0.011	146.25	<0.0001
Pure error	3.782E-004	5	7.564E-005		
Total	1.26	29			
Gel Strength(GS) (g)					
Model	481.99	6	80.33	19.55	< 0.0001
A	47.51	1	47.51	11.56	0.0025
B(extraction time)	49.75	1	49.75	12.11	0.0020
C	113.75	1	113.75	27.68	< 0.0001
BD	34.54	1	34.54	8.41	0.0081
B^2	235.76	1	235.76	57.38	< 0.0001
Residual	94.50	23	4.11		
Lack of Fit	88.30	18	4.91	3.96	0.0671
Pure error	6.20	5	1.24		
Total	576.49	29			

df: degree of freedom, A: concentration of acetic acid, B: extraction time, C: extraction temperature, D: skin to water ratio

Response surface plots

The relationship between responses and significant variables is shown in Figure 1a-1d. Extraction temperature, extraction time and skin to water ratio gave linear effects on GY as shown in Figure 1a and 1b, while acetic acid concentration does not show any significant effect. The same result was obtained by Al-Saidi *et al*. (2011) whom reported that the higher the temperature used the higher the gelatine yield, for all acetic acid concentrations. According to Samart *et al*. (2010) the extraction process with high temperature could influence the release of the polypeptide chains from the fish skin. Other researchers also

reported that extraction yields increased gradually with increasing extraction time (Cho *et al*., 2006; Arnesen & Gildberg, 2007).

Concentration of acid (A) show a linear effect on gel strength as shown in Figure 1c, which was consistent with Cho *et al*., (2006) and Mohtar *et al*. (2010) studies. GS increased with increase in extraction temperature and slightly decreased as the temperature increased above 80ºC (Figure 1c). Another factor that significantly affects gel strength is extraction time. The lower the extraction time, the higher the GS obtained (Figure 1d). GS declined at higher extraction time and temperature due to the breakage of hydrogen bonds and free amino acid hydroxyl groups (Cho *et al*., 2006). Besides, high extraction temperature also caused protein degradation and denaturation that produced small protein which have a lower gel strength value (Yang *et al*., 2007).

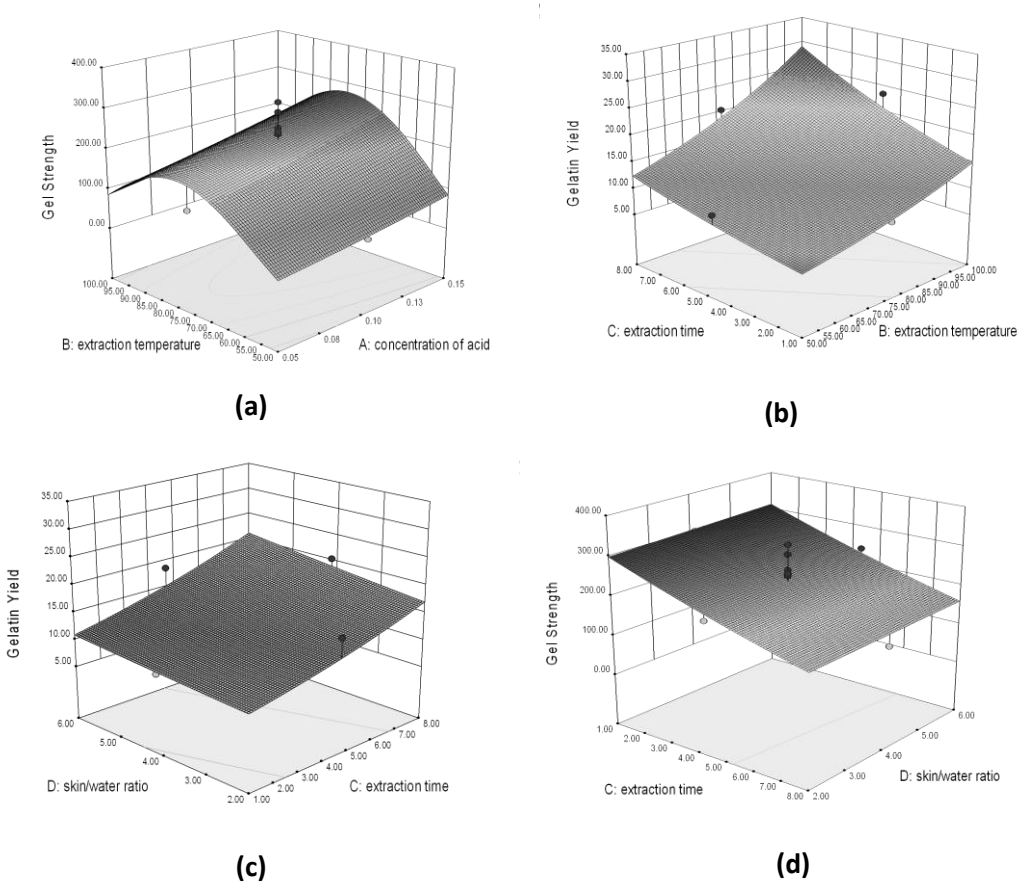

Figure 1 : a. Effect of extraction time and temperature on GY; F b. Effect of skin to water ratio and extraction time on GY; c. Effect of concentration of acid and extraction temperature on GS; d. Effect of extraction time and skin to water ratio and extraction time on GY.

Optimization and verification

Optimization process was carried out to determine the optimum condition for gelatin extraction. The desired goal for independent parameters which consist of extraction

temperature, extraction time and reaction time were set within the range while the responses; GY and GS were set to maximum value to achieve the highest performance. Table 4 summarized the limitation used for optimization study. Under the optimum conditions (concentration of acetic acid 0.15 mol/L, extraction temperature 82.4^{o}C, extraction time 6 hours and skin to water ratio 1:6), the predicted values are GY of 20.59% and GS of 226.86 g. Using the optimum conditions, verification experiments were conducted and the experimental values obtained were GY of 20.10% and GS of 205.6 g, which agrees well with the predicted response value.

Table 4. Optimization constraints used for optimization of gelatine extraction

Criteria	Goal	Lower Limit	Upper Limit
Concentration of acetic acid (mol/L)	In range	0.05	0.15
Extraction temperature (^{o}C)	In range	50.0	100.0
Extraction time (hour)	In range	1	8
Skin to water ratio (w/v)	In range	1:2	1:6
Gelatin yield (%)	Maximum	4.4	33.9
Gel strength (g)	Maximum	0.0	316.58

Gelatin yield

The value of yield obtained in this study were slightly higher compared to those reported for other species such as hoki 17.4% (Mohtar *et al*., 2010), channel catfish 19.2% (Yang *et al*., 2007) and grass carp 19.83% (Kasankala *et al*., 2007). This difference could be due to different collagen content in raw material (Kolodziejska *et al*., 2008) and different processing parameters (Montero & Gomez-guillen, 2000; Kolodziejska *et al*., 2004).

Gel strength

Many studies has reported similar gel strength of fish gelatin which were 197g for hoki (Mohtar *et al*., 2010), 340g for megrim (Gomez-Guillen *et al*., 2001), 267g for grass carp (Kasankala *et al*., 2007) and 252g for channel catfish (Yang *et al*., 2007). It is impossible to obtain gel with comparable gel strength due to the differences in the preparations of the samples, the experimental gel strength settings, and the equipments used (Boran and Regenstein, 2009). According to Holzer (1996), commercial gelatin commonly has gel strength value vary from 100 to 300g.

Conclusion

This study showed that RSM has been successfully used to optimize the extraction parameters of cobia skin gelatin. The optimum conditions obtained were as follows: acetic acid concentration of 0.15mol/L; extraction temperature of 82.4^{o}C; extraction time of 6 hrs and skin to water ratio of 1:6. This study shows that RSM can be used to predict gelatin extraction from cobia skin.

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Identification of Pork Contamination in Meatballs of Local Market Using PCR-RFLP Analysis

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Abstract

This research applied and evaluated a PCR-RFLP procedure to detect pork contamination in meatballs from local market in Yogyakarta using cytochrome b gen. To confirm the effectiveness and specificity of this fragment, sixteen meatballs DNA samples from different "warung bakso" were isolated and amplification, then PCR amplicon was digested by BseDI restriction enzyme to distunguished pork existence in meatballs. BseDI restriction enzyme was able to cleave porcine cytochrome b gene into two fragment (131 bp and 228 bp). The meatballs of local market showed that seven of sixteen "warung bakso" were detected pork contamination. In conclusion, specific PCR amplification of cyt b gen and cleaved by BseDI restriction enzymes seems to be a powerful technique for the identification of pork existence in meatball because of its simplicity, specificity and sensitivity. Furthermore, pork contamination intended for sausage, nugget, "abon" and "dendeng" can be checked. The procedure is also much cheaper than other methods based on PCR, immunodiffusion, and other techniques that need expensive equipment.

Keywords: detection, meatballs, polymerase chain reaction, pork

Introduction

The identification of the meat species is the sensitive and important issues of the food product especially in moslem country. The food safety and assurance consist some aspects including healthy, hygienist and *halal* authentication. An effective method for detecting pork in food from meat or non meat products is essential to avoid falsification. The falsification of food products including the meat product and non meat products is becoming common due to get higher profit by mixing *halal* meat with pork.

The reliable, sensitive and exact procedure for the authentication is crucial for law enforcement, quality assurance and consumer protection. Authenticity testing and analytical techniques have improved immeasurably since then and must now draw on a wide variety of techniques and methods, each appropriate and specific to deal with a particular problem (Hargin, 1996). Several different techniques are currently used for determination of species, primarily relying on detecting differences in proteins and are either electrophoretic or immunological methods. One of the major drawbacks with such methods is that they do not perform well with heat treated samples. More recently DNA techniques have been applied to meat specification with advantage that the DNA is much more stable to cooking temperatures.

Polymerase chain reaction (PCR)-based assays are currently the method of choice for species identification. PCR analysis of species-specific mitochondrial DNA sequences is the most common method currently used for identification of meat species in food and animal

feedstuffs (Bellagamba *et al*., 2001). The advantage of mitochondrial-based DNA analyses derives from the fact that there are many mitochondria per cell and many mitochondrial DNA molecules within each mitochondrion, making mitochondrial DNA a naturally amplified source of genetic variation (Partis *et al*., 2000; Aida *et al*., 2005, Girish *et al*., 2005). Additionally, procedures like polymerase chain reaction mean that the sensitivity of these test are greatly enhanced and positive reaction are possible from the merest trace of the species under investigation. Erwanto *et al*., (2011) has been reported that digestion of PCR products of mitochondrial cytochrome b gene using BseDI restriction enzyme was able to distinguish the existence of pork existence in processed food which contaminated with 0.1% level of pork.

However the traceability and identification of pork and other derivate porcine contamination in most popular food in Indonesia was not previously done. Therefore this paper was aimed to detection of pork contamination in meatballs from local market using PCR-RFLP methods through *cytochrome b* gene.

Materials and Methods

Sample Preparation and DNA Extraction

Meatballs from "warung bakso" were obtained from local market. From Table 1, the list of "warung bakso" location where samples have been obtained.

Table 1. List of meatballs sample from various "Warung Bakso"

Samples code	Addresses
A	Jalan Gejayan, Yogyakarta
B	Jalan Malioboro, Yogyakarta
C	Jalan Mataram, Yogyakarta
D	Jalan Bhayangkara, Yogyakarta
E	Jalan Pasar Kembang, Yogyakarta
F	Jalan Bhayangkara, Yogyakarta
G	Jalan Solo, Yogyakarta
H	Jalan Solo, Yogyakarta
I	Jalan Panjaitan, Yogyakarta
J	Jalan Bantul, Yogyakarta
K	Jalan Bugisan, Yogyakarta
L	Jalan Letjend Suprapto, Yogyakarta
M	Jalan Aipda K.S. Tubun, Yogyakarta
N	Jalan Pemuda, Klaten
O	Jalan Dewi Sartika, Klaten
P	Jalan Solo, Yogyakarta

DNA was extracted from meatball samples using the *Nucleospin® Food* DNA Kit protocol provided by *Macherey-Nagel*. Approximately 200 mg of meatballs was blended using a commercial blender and placed in a 1,5 ml microcentrifuge tube. A-550 µl of *lysis buffer CF* and *Proteinase* K were added and mixed by vortexing. The mixture was incubated at 65°C in a water bath overnight to disperse the sample until the tissue was completely lysed. Sample was centrifuged at 10,000 g for 10 minutes. An 300 µl samples were then added with 300 µl binding buffer C4 and 300 µl ethanol. The mixture was mixed by vortexing for 30 seconds, added with 300 µl ethanol, mixed vigorously and placed on *NucleoSpin® Food Column*. 750 µl samples was subsequently poured in the collection tube, placed in table top centrifuge,

and spun at 11,000 g for 1 min. The flow-through and *collection tube* were discarded and the *NucleoSpin® Food Column* placed in a new 2 ml *collection tube*. A-400 µl of *wash buffer* was added and spun at 11,000 g for 1 min. The flow-through and collection tube were discarded and the *NucleoSpin® Food Column* was placed in another 2 ml *collection tube*. An 700 µl buffer C5 was added The high filter tube was dried by centrifugation at 11,000 g for 1 min, and the supernatant flow-through was discarded. The *NucleoSpin® Food Column* was placed in a clean 1.5 ml micro centrifuge tube. A-200 µl of *pre-warmed elution buffer* was added and incubated for 5 min at room temperature and then spun at 11,000 g for 1 min to elute. The DNA solution was stored at -20 °C for further analysis.

PCR Amplification of Cytochrome 2b of Mitochondrial Gene

The set of primers used for amplification consisted of Cyt b-FW and Cyt b-REV oligonucleotides as follows:

CYT b FW 5'-CCA TCC AAC ATC TCA GCA TGA TGA AA-3'
CYTb REV 5'-GCC CCT CAG AAT GAT ATT TGT CCT CA-3'

Amplification of the mt cyt b gene was performed in a final volume of 25 µl containing 250 ng of extracted DNA, mega-mix royal (optimized mixture of *Taq* polymerase, anti-*Taq* polymerase monoclonal antibodies in 2 X reaction buffer (6 mM $MgCl_2$ with 400 µM dNTPs, stabilizer and blue loading dye) (Microzone Ltd, West Sussex, UK), and 20 pmol of each primer. Amplification was performed with a thermal cycler according to the following PCR step-cycle program: pre-denaturation of 94 °C for 2 min to completely denature the DNA template, followed by 35 cycles of denaturation at 95 °C for 36 s, annealing at 51 °C for 73 s, and extension at 72 °C for 84 s. Final extension at 72 °C for 3 min followed the final cycle for complete synthesis of elongated DNA molecules. Two microlitres of PCR products were electrophoresed at constant voltage (50 V) on 2% agarose gel (Promega, Madison, USA) for about an hour in 1x TBE buffer, pH 8.0 and stained by ethidium bromide. A-100 bp DNA ladder (Promega, Madison, USA) was used as size reference. The gel photo was taken using the Syngene gel documentation system.

Restriction Fragment Length Polymorphism

Two units/µl of RE *BseDI* (Fermentas) were applied to 10 µl of amplified DNA in a final volume of 20 µl digestion mixture [containing 1x reaction buffer (10 mM Tris-HCl, 100 mM KCl, 1 mM EDTA, 0,2 mg/ml BSA, 1 mM DTT and 50% glycerol)] and were incubated at 55 °C for 3 h for optimal result. A-5 µl of the digested samples were electrophoresed at constant voltage (50 V) on 2% agarose gel (Promega, Madison, USA) for about an hour in 1x TBE buffer, pH 8.0 and stained by ethidium bromide. A-100 bp (Promega, Madison, USA) was used as size reference. The gel photo was taken using the Syngene gel documentation system.

Results and Discussion

DNA was extracted from meatball products using Nucleo Spin Food DNA Kit. Genomic DNA was used as template for the amplification of PCR with the universal primer. The first step of the protocol for PCR analysis is the extraction of DNA genome. The extraction genomic DNA from the meat sample both of meatball cooked sample are reliable and there are no difficulties. Figure 1 showed the electrophoretic patterns of genomic DNA from local market meatballs.

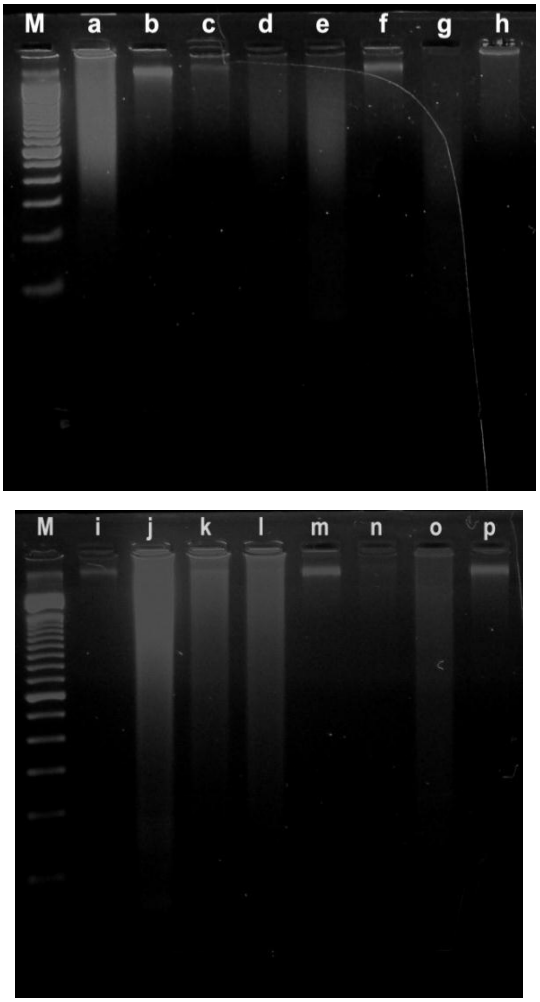

Figure 1. Total genomic DNA extracted from local market meatballs. M: marker 100 bp DNA ladder (Invitrogen), a-p : genomic DNA of meatballs from different "warung bakso"

PCR has a potential sensitivity and specificity required to achieve detection of a target sequence from template DNA. The amplicons of the cytochrome b gene of the meatball from sixteen location of "warung bakso" are shown in Figure 2. Gene of cytochrome *b* was used for the amplification of PCR and resulted DNA fragment of approximately 360 bp for beef sausage before or after frying and chicken nugget. These small amplicons are ideal for use with processed foods where DNA commonly was degraded. This result indicated that pig, beef, and chicken DNA in processed meat was successfully amplified in PCR reaction.

PCR-RFLP allows the amplification of a conserved region of DNA sequence using PCR and the detection of genetic variation among species by digestion of the amplified fragment with restriction enzymes.

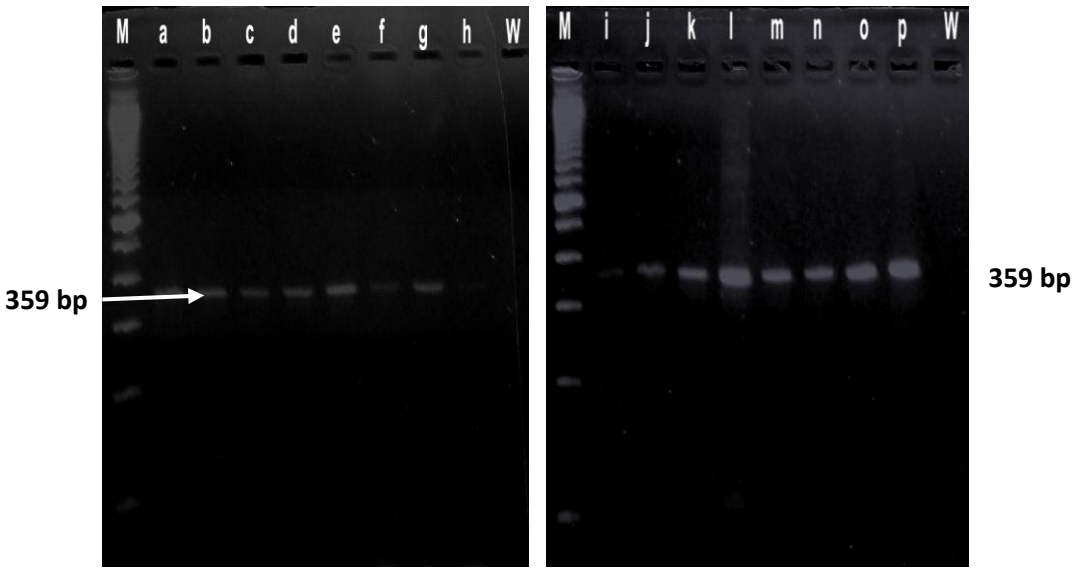

Figure 2. PCR products of cytochrome b gene fragments 359 bp long of samples from different meatballs product separated by 2% high-resolution agarose gel electrophoresis. PCR amplification using cyt b universal primer. M: marker 100 bp DNA ladder (Invitrogen), a-p: PCR products of cyt b gene from sixteen source of meatballs sample, W: water (negative control).

The present study designed to extend PCR-RFLP application for the specific identification of pig, targeting sequences of the cyt b mitochondrial DNA gene. PCR product was digested using *BseDI* restriction enzyme at 55°C for 3 h. The result of RFLP analysis of meatballs sample from various places (Fig. 3.) indicated some positive pork contamination. Erwanto *et al*. (2011) reported that cyt b gene from pig species could be digested by *BseDI* restriction enzyme and 0.1% pork contamination level was able to determine. Data of pork contamination were detected from seven "Warung Bakso". *BseD*I cleavage bands visualized in the gel were enough and suitable for the discrimination of pork in processed food. *BseD*I endonuclease cleaved the cytochrome b gene products of pig species into two DNA fragments of 228 and 121 bp (Figure 3 lane a, b, e, f, g, h and l),

Result of PCR amplification was similar to Aida *et al.* (2005) which reported the presence of 360 bp fragment. This result also indicated that meatball which commonly cooked for more than 30 min did not produced degraded DNA into small size (< 200 bp). Tanabe *et al.* (2007) investigate the feasibility of using mitochondrial cyt b gene to detect pork in commercial food products from a market and evaluated them for the presence of porcine DNA. Electrophoresis of PCR product clearly detected porcine DNA, while no amplification occured in others meat sources: cattle, chicken, sheep and horse.

The optimum PCR conditions are affected by some factors including the temperature, design primer, polymerase enzyme and also buffer quality. Severals analytical methods using PCR have been developed to qualitatively detect the presence of non halal ingredients of meat species. Tajima *et al*. (2002) using PCR with species specific primer, Tanabe *et al.* (2007) PCR with cloning of gene target, Aida *et al.* (2005) using PCR Restriction Fragment Lenght Polymorphism (PCR-RFLP) and Walker *et al.* (2003) using PCR amplification of short interspersed elements

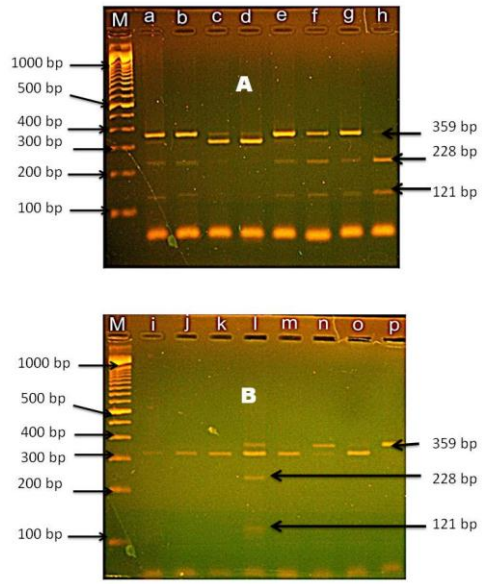

Figure 3. Electrophoretic patterns of cytochrome b gene after digested by BseDI restriction enzymes. A) M: marker 100 bp DNA ladder, a-h: from various places a. jl. Gejayan, b: jl. Malioboro, c: Jl. Mataram, d: Jl. Bayangkara 1, e: jl. Pasar kembang, f: Jl. Bayangkara 2, g: Jl. Solo 1, h: Jl. Solo 2, B). M: Marker, i-p: meatball samples from various places 9, i: Jl. Panjaitan, j: Jl. Bantul, k: Jl. Bugisan, l: Jl. Letjen Suprapto, m: Jl. Aipda K.S. Tubun, n: Jl. Pemuda Klaten, o: Jl. Dewi Sartika Klaten, and p: Jl. Solo 3.

*BseD*I cleavage bands visualized in the gel were enough and suitable for the discrimination of pork in processed food. *BseD*I enzyme cleaved the cytochrome b gene products of pig species into two DNA fragments of 228 and 131 bp. This research finding showed that some places of warung bakso have been contaminated with pork. The founding of pork contamination in meatballs of local mark*et al*lows us to give some predictions. The first prediction is some meatballs producer may be chopped his raw meat in the same places with pork chopping machine. The second interpretation, meatballs producer substituted beef with pork for economic reason. Pork was cheaper than beef, consequently replacement beef with pork increased their advantage.

Ong *et al*. (2007) used various restriction enzymes like *Alu*I, *BsaJI* and *Rsa*I to identify pork from different mixture of meat chicken, pig and beef in various modifications. Other study by Fajardo *et al*. (2006) reported PCR-RFLP based on the mitochondrial 12S rRNA fragment targeted cannot be applied for species identification in thermally treated meats. In the analysis of food matrices in which thermal action or other processing effects may degrade the DNA present in the food tissues. However this result allow us to reported that PCR-RFLP of the mitochondrial cytochrome b gene is a suitable alternative that can be applied to the detection of pork in processed meat in commercialized products such as meatball and sausage.

BseDI represent one of the restriction enzyme of type II which is able to recognize the nucleotide sequence and cut the DNA molecule specifically. *BseDI* is isoschizomer of *Bsa*JI,

and *Sec*I. This enzyme cut the sequence of DNA molecule at sequence of C CNNGG. *BseDI* enzyme was obtained from bacterium of *Geobacillus strearothermophilus* (Fermentas AB) and *Bacillus stearothermophilus* RFL1434 (US, Biological), while *Bsa*JI enzyme from bacterium of *Geobacillus strearothermophilus* J695 (New England Biolabs).

Conclusions

PCR-RFLP represents the appropriate method to identify a certain species in cooked meat. There were seven positive pork contaminations from sixteen "warung bakso" around Yogyakarta. Identification the species through analysis of restriction patterns is quite and practical application.

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Carotenoid, Total Phenolic Content, and Antioxidant Activities of "Jintan Leaves" (*Plectranthus amboinicus* L. Spreng.)

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Abstract

Studies on antioxidant activity, total phenolic content and carotenoid content in the leaves of the cumin (Plectranthus amboinicus L. Spreng.) have been carried out. The purpose of this study was to determine the antioxidant activity, total phenolic content and carotenoids content at leaves extract of cumin (Plectranthus amboinicus L. Spreng.) Antioxidant activity in study was measured by DPPH method, and reducing power method, where as phenolic content was measured by staining with Folin Ciocalteau method, and the measurement of carotenoids was carried out with lichtenthaler method. Data obtained from this research were tested with RAK (Randomized completely block design) test with 5 treatments (hexane, ethyl acetate, acetone, ethanol, and water as solvents). The results showed that the highest of antioxidant activity by DPPH method and reducing power method; total phenolic content and the content of carotenoids were found in ethyl acetate extract. Antioxidant activity by DPPH method was 83.31 ± 2.00%, while the reducing power method was 4.777 ± 0.1131 mek $K_4Fe(CN)_6$ / g extract, and total phenolic content was 32.09 ± 3.87 mg / g extract and the content of carotenoids was 5,2394 ± 0,1715 mg / g extract.

Keywords: antioxidant activity, carotenoid, free redical scavenger, phenolic, reducing power

Introduction

'Jintan' (*Plectranthus amboinicus* L. Spreng.) grow wild around the Himalayan mountain and other places at 1100 m above sea level. It could be plant in the garden or in pots as spices. Had the leaves yellowed green, oval, serrated and coarse, thick and has a rather unpleasant odor. The leaves can be used as a medicine and can treat various diseases. Commonly used for heartburn pain, flatulence, mouth ulcer, fever, inflammation of the nose, constipation and bitten by poisonous insects (Anonim, 2008).

The various functions in health might be caused by antioxidant compounds in this plant. Besides beneficial to health, antioxidant also plays important role to maintain the quality of food products. Various damages such as rancidity, changes in nutritional value, changes in color and aroma, as well as other physical damage to the food product due to oxidation can be inhibited by antioxidants. There are two kinds of mechanism of action of antioxidants : primary and secondary antioxidants. Primary antioxidants functions as donor of hydrogen atom. Secondary antioxidant functions as decreasing the rate of autooxidation with various mechanisms out of chain termination mechanism by conversion autooxidation with lipid radicals to form more stable (Gordon, 1990).

Some vitamins such as carotenoid, ascorbic acid, and tocopherol are believed to have functions as natural antioxidant, besides phenolic compounds. Phenolic compounds spread

in many kinds of plant. It has function as antibiotic, natural pesticide, pigmentation in plant, facilitate polination in plant, and as barirer to UV-light (Shahidi & Nazck, 2004). Carotenoid is a natural yellow pigment, has '*isoprenoid structure'* which consisted of 8 isoprenes unit. One example of it is beta carotene, which could functions as antioxidant by inhibit formation of hydroperoxide (Gross, 1991).

The aims of this research were: 1) determination of antioxidant activity of jintan leaves extracted with several kinds of solvent by free radical scavenger method and by reducing power method; 2) determination of phenolic content of jintan leaves extracted with several kinds of solvent and their carotenoid content.

Materials and Methods

Materials

Jintan leaves (*Plectranthus amboinicus* L. Spreng.) were obtain from Kopeng, Central Java (altitude 1100 m above sea level). Reagents : n-hexane, ethyl acetate, ethanol, DMSO (*Dimethyl Sulfoxide*), DPPH (*1,1-diphenyl-2picrylhydrazil*), Folin-Ciocalteu, Na_2CO_3, galic acid, NaH_2PO_4, $Na_2HPO_4.{}_7H_2O$, $K_3Fe(CN)_6$, $FeCl_3$, $K_4Fe(CN)_6$, and TCA (Merck, Jerman).

Apparatus

Mortar and pestle, waterbath, 4 decimal analytical balance (Mettler, H80), oven (WTB, Binder), dessicator (Glaswerk Wertheim, GL32), shaker (IKA Labortechnik, KS 501), rotary evaporator (Heidolph, Laborota 4000), spectrophotometer (UV-VIS mini-1240, Shimadzu), centrifuge (Hettich, EBA 21).

Methods

Sample extraction (Yen *et al*.; 1996; Yen & Hsieh, 1998; Zhu *et al*., 2007 modified)

Jintan leaves was crushed with mortar and pestle, then extracted by several kinds of solvent (hexane, ethyl acetate, acetone, ethanol, and water), with ratio of sample: solvent = 1: 5 (w / v), by macerated and shook for overnight. After filtered, the residue were macerated again with the same solvent. The filtrate were collected together and concentrated with a rotary evaporator, and dried with N_2 vapor and weighed; these were data for yield.

For extraction with water, after macerated, the extract was not concentrated with rotary evaporator, instead the filtrate was accomplished to 100 ml in volumetric flask. 20 ml of it was dried in a petri dish, and weighed. The rest of the filtate was used for determination of the antioxidant activity, phenolic and carotenoid content.

Determination of antioxidant activity by free radical Scaveging method (Banerjee *et al*., 2005)

1 ml sample was added with 3 ml DPPH 0.2mM in ethanol 95 %, then incubated in room temperature for 30 minutes, avoid from light. The absorbance was measured with UV-VIS

spectrophotometer on 517 nm. Ethanol was used as blank for spectrophotometer. The inhibition was calculated with formula :

$$\% \text{ Inhibition} = \frac{\text{Absorbance of control} - \text{Absorbance of sample}}{\text{Absorbance of sample}} \times 100\ \%$$

Determination of antioxidant activity by reducing power method (Yildirim *et al*., 2001 modified).

0,5 ml sample was added with 1,25 ml buffer phosphat 0,2 M pH 6,6 and 1,25 ml $K_3Fe(CN)_6$ 1 %, and incubated at 50°C for 30 minutes. Then 1,25 ml TCA 10% was added and sentrifuge on 3000 rpm for 10 minutes. 1,25 ml of upper layer was then added with 1,25 ml sample solvent (aquadest / DMSO) and 0,25 ml $FeCl_3$ 0,1 %. This mixture was incubated again on 37 °C for 10 minutes, then the absorbance was measured on 765 nm. Sample solvent was used as a blank (distilled water / DMSO). K4Fe (CN) 6 were used as standards, and the antioxidant activity was expressed as mek / 100 g sample.

Determination of phenolic content (Jayaprakasha & Jaganmohan, 2000).

0.2 ml sample in DMSO (in water for water extract) was added with 1 ml 10% Folin-Ciocalteu and 0.8 ml 7.5%. Na2CO3. After incubation in room temperature for 30 minutes, the absorbance was measured at 765 nm. Total phenolic content expressed in mg gallic acid /100gram sample.

Determination of carotenoid (Lichtenthaler, 1987)

Crude extract was dissolved with acetone 80%, and the absorbance was measured at 663 nm, 646 nm, 470 nm. Carotenoid content was determined by formula :

$$Ca = 12{,}21\ A_{663} - 2{,}81\ A_{646}$$

$$Cb = 20{,}13\ A_{646} - 5{,}03\ A_{663}$$

$$Cx+c = \frac{1000\ A_{470} - 3{,}27\ Ca - 104\ Cb}{229}$$

Ca : Chlorophyl a

Cb : Chlorophyl b

Cx+c : Carotene

Data analysis

Data of antioxidant activity, total phenolic and carotenoid content by several kinds of solvent (hexane, ethyl acetate, acetone, ethanol and water) were analyzed by Randomized Block Design, with kinds of solvent (hexane, ethyl acetate, acetone, ethanol and water) as treatments; each were repeated 5 times. HSD Test (Honestly Significant

Difference) 5% was used to compare significantly difference among the antioxidant activities, total phenolic and carotenoid content (Steel and Torie, 1980).

Results and Discussion

Yield of Jintan leaves extract

Yield of jintan leaves extract on Table 1 showed that the most efective solvent for extracting jintan leaves was ethanol, followed by acetone, water, ethyl acetate and hexane. This result was influenced by polarity of each solvents : ethanol was the most polar compare to other solvents that were semi polar or non polar.

Table 1. Yield of Jintan leaves extracts

Extract	Yield ± SE (%)
Hexane	0,21 ± 0,13
Ethyl acetate	0,68 ± 0,37
Acetone	1,32 ± 0,32
Ethanol	1,41 ± 0,23
Water	1,29 ± 0,07

Antioxidant Activity by Free Radical Scavenger Method with DPPH

Data of antioxidant activity by free radical scaveging method on Table 2, showed that the highest free radical scaveging activity obtained in ethyl acetate extract (83.31%) that were significantly higher than other extracts (ethanol extract, 52.15%; hexane extract, 51.43%; acetone extract, 43.00%; and water extract, 14.19%).

Table 2. Antioxidant activity of Jintan leaves extracts by free radical Scavenger method

Extract	% Inhibition ($\bar{X}$ ± SE)
Hexane	51,43 ± 0,85 (c)
Ethyl acetate	83,31 ± 2,00 (d)
Acetone	43,00 ± 0,74 (b)
Ethanol	52,15 ± 0,58 (c)
Water	14,19 ± 0,84 (a)
BHT	90,66 ± 5,94

W = 1,8594

Note : value followed by different alphabet means significally different in treatments

This results showed that ethyl acetate was the best solvent for extracting antioxidant compounds in jintan leaves, especially which had free radical characteristics. A synthetic antioxidant - BHT - was used as comparison, because it is often used as a food preservative. The antioxidant activity of ethyl acetate (83,31%) was high that almost the same compared to antioxidant activity of BHT (90.66%).

IC_{50} value of jintan leaves by free radical scavenger method extracted with several kinds of solvent were shown on Table 3. IC_{50} value means concentration of sample that could inhibit 50% of DPPH, as free radical. Thus low IC_{50} values implicated high antioxidant activity.

Table 3 showed that the lowest IC_{50} value found in ethyl acetate extract (267.36 µg/ml) followed by ethanol extract (407.65 µg/ml), hexane extract (495.49 µg/ml), acetone extract (1271.47 ug / ml), and water extract (1665.79 g / ml); whereas the IC_{50} value of BHT was 81.71 ug / ml. Therefore the highest antioxidant activity was found in ethyl acetate extract with IC_{50} value of 267.36 µg / ml.

Table 3. IC_{50} Value (µg/ml) of Jintan Leaves extracts by free radical Scavenger method

Extract	µg/ml ($\overline{X}$ ± SE)
Hexane	495,49 ± 42,96 (b)
Ethyl acetate	267,36 ± 58,98 (a)
Acetone	1271,47 ± 87,92 (c)
Ethanol	407,65 ± 38,55 (b)
Water	1665,79 ± 123,98 (d)
BHT	81,71 ± 10,29

W =134,25

Note : value followed by different alphabet means significally different between treatments

Antioxidant Activity of Jintan Leaves Extracts by Reducing Power Method

Antioxidant activity of jintan leaves extracts by reducing power method on Table 4 showed that the highest antioxidant activity of jintan leaves extracts was found in the ethyl acetate extract (4.77 mek K4Fe (CN) 6/ g extract), followed by water extract (3.7152 mek K4Fe (CN) 6 / g extract), hexane extract (1.5387 mek K4Fe (CN) 6 / g extract), ethanol extract (0.3022 mek K4Fe (CN) 6 / g extract), and acetone extract (0.2877 mek K4Fe (CN) 6 / g extract). This results indicated that ethyl acetate extract contained more reducing compounds compared to other extracts.

The antioxidant activity of BHT was 4.53 mek K4Fe (CN) 6 / g extract, lower than the antioxidant activity of ethyl acetate extract. It could be concluded that jintan leaves had huge potential as natural antioxidants. However, when calculated in mg/g leaves, the highest reducing power was found in water extract followed by ethyl acetate, ethanol, hexane, and acetone extracts.

Table 4. Antioxidant activity of Jintan Leaves extracts by reducing power method (mek $K_4Fe(CN)_6$/g Extract)

Extract	mek $K_4Fe(CN)_6$/g Extract ($\overline{X}$ ± SE)	mek $K_4Fe(CN)_6$/g leaves ($\overline{X}$ ± SE)
Hexane	1,5387 ± 0,0162 (b)	0,0034 ± 0,0055 (b)
Ethyl acetate	4,7700 ± 0,1131 (d)	0,0355 ± 0,0196 (d)
Acetone	0,2877 ± 0,0304 (a)	0,0040 ± 0,0006 (a)
Ethanol	0,3022 ± 0,1020 (a)	0,0077 ± 0,0052 (c)
Water	3,7152 ± 0,0998 (c)	0,0538 ± 0,0063 (e)
BHT	4,53 ± 0,5314	4,53 ± 0,5314

W= 0,1175 (BNJ 5%); 0.0170(BNJ 5%)

Note : value followed by different alphabet means significally different between treatments

Phenolic Content of Jintan Leaves Extracts

Phenolic content of jintan leaves extracts in Table 5 showed that the highest total phenolic content of jintan leaves extracts was found in ethyl acetate extract (32.09 mg/g extract) and hexane extract (24.90 mg/g extract) that were significantly higher than water extract (15.52 mg/g extract), ethanol extract (3.89 mg/g extract) and acetone (3.77 mg/g extract).

Table 5. Total Phenolic Content (mg/g extract) of Jintan leaves extracts

Extract	mg/g extract ($\bar{X}$ ± SE)	mg/g leaves ($\bar{X}$ ± SE)
Hexane	24,90 ± 0,93 (c)	0.0538 ± 0.0318 (a)
Ethyl acetate	32,09 ± 3,87 (c)	0.2181 ± 0.1273 (c)
Acetone	3,77 ± 0,29 (a)	0.0532 ± 0.0094 (a)
Ethanol	3,89 ± 0,51 (a)	0.0520 ± 0.0174 (a)
Water	15,52 ± 9,96 (b)	0.1999 ± 0.1204 (b)

W = 7,6539 (BNJ 5%); 0,0817 (BNJ 5%)
Note : value followed by different alphabet means significally different between treatments

It influenced by the polarity of ethyl acetate and hexane that were more suitable to extract phenolic compounds in jintan leaves. Solubility of phenolic compounds in ethyl acetate showed that phenolic compounds in jintan leaves were semi-polar, whereas the high phenolic content in hexane extract might be caused by the presence of other compounds that could be extracted by hexane, possibly this compound was carotenoid.

Correlation between antioxidant activity by free radicals scavenging method and phenolic content in Figure 1 showed r = 0.500. It meant that only some part of antioxidant activity in jintan leaves were caused by phenolic compounds. Phenolic compounds were not major source of antioxidant in jintan leaves. Jintan leaves were also contained carotenoids that were had a role as antioxidant because they could react with a reducing agent to form stable products (Gordon, 1990). Therefore, relationship between carotenoid content with antioxidant activity by reducing power method also be reviewed later.

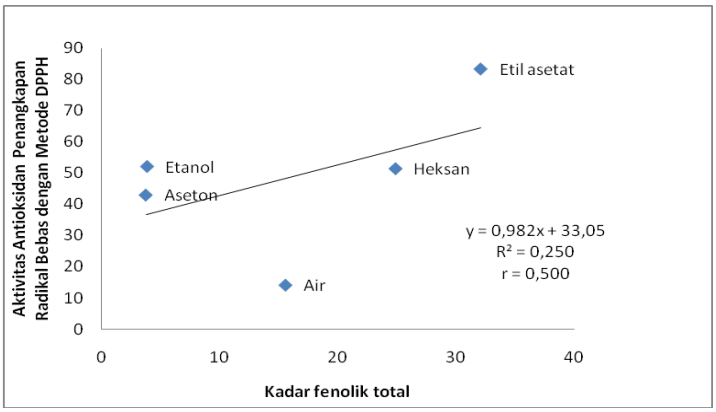

Figure 1. Correlation between antioxidant activity by free radical scavenger method (% inhibition) and phenolic content (mg/g Extract) of Jintan leaves extracts.

Correlation between antioxidant activity by reducing power method and total phenolic content in Figure 2 showed r = 0.7918, meant the phenolic compounds in jintan leaves extract had a major role as the reducing agent. They play more as reducing agent than as free radical scavenger. According to Hart (1983), phenolic compounds were easily to be oxidized, therefore they had characteristics as reductor.

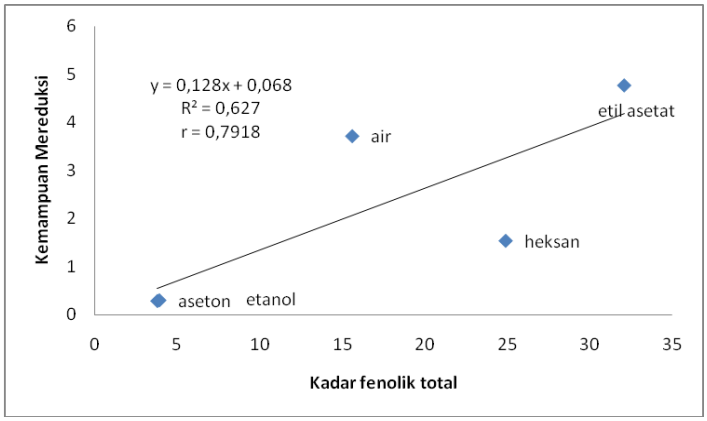

Figure 2. Correlation between antioxidant activity by reducing power method and phenolic content of jintan leaves extracts

Carotenoid content in Jintan leaves extract

Table 6. Carotenoid (mg/g extract) content of Jintan leaves extract

Extract	mg/g ($\bar{X}$ ± SE)
Hexane	0,7048 ± 0,1041 (a)
Ethyl acetate	5,2394 ± 0,1715 (e)
Acetone	1,2505 ± 0,2149 (b)
Ethanol	2,1815 ± 0,1571 (d)
Water	1,7891 ± 0,1684 (c)

W = 0,2888

Note : value followed by different alphabet means significally different between treatments

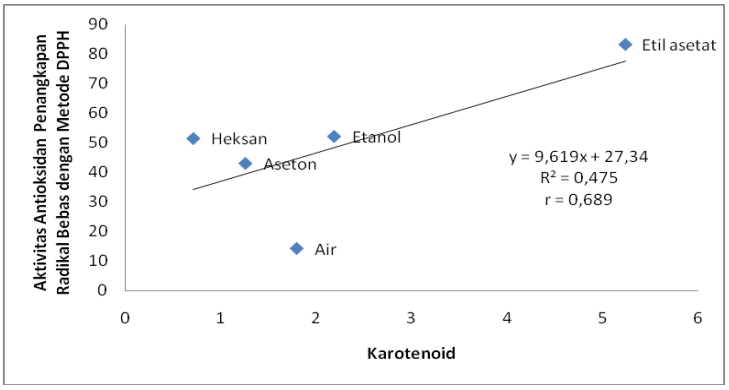

Figure 3. Correlation between antioxidant activity by free radical scavenger method and carotenoid content (mg/g Extract)

Carotenoid content in jintan leaves extracts on Table 6 and Figure 3 showed that the highest carotenoid content in jintan leaves extracts was found in ethyl acetate extract (5.2394 mg/g extract), followed by ethanol extract (2.1815 mg/g extract), water extract (1.7891 mg/g extract), acetone extract (1,2505 mg/g extract), and hexane extract (0.7048 mg/g extract). The polarity of the ethyl acetate was more suitable for extracting carotenoids in jintan leaves.

Correlation between antioxidant activity by free radical scaveging method and carotenoid content showed r = 0.689, meant that the carotenoid compounds in jintan leaves extract had antioxidant activity toward free radicals. Compared with the correlation between phenolic compounds and antioxidant activity by free radicals scavenger method that had r = 0.500; indicated that the antioxidant compounds in jintan leaves were more influenced by carotenoids than influenced by phenolic.

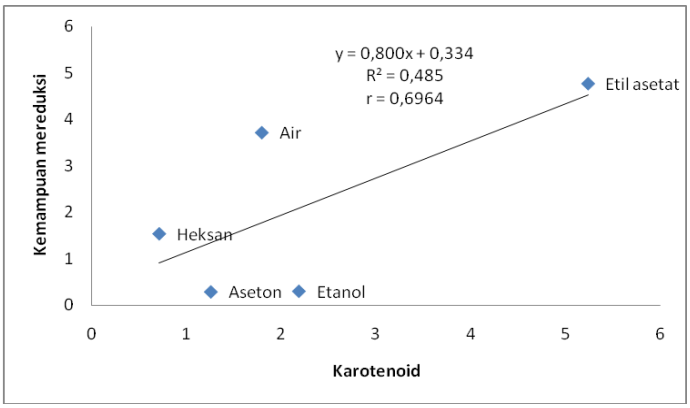

Figure 4. Correlation between carotenoid content (mg/g extract) and antioxidant activity by reducing power method

Correlation between carotenoid content and antioxidant activity by reducing power method showed r = 0.6964 which meant that some of carotenoid compounds in jintan leaves extract could reduced $K_4Fe(CN)_6$ well. When compared to correlation between phenolic content and antioxidant activity by reducing power that had r = 0.7918; it could be concluded that phenolic compounds in jintan leaves were more effective as antioxidant than the carotenoids.

Conclusions

From the research, it can be concluded that :

1. The highest antioxidant activity of jintan leaves both by free radicals scavenger method and by reducing power method were found in the ethyl acetate extract : 83.31%; and 4.77 mek $K_4Fe(CN)_6$/g extract;
2. The highest of phenolic content and carotenoid content in jintan leaves were found in ethyl acetate extract : 32.09 mg/g extract; and 5.2394 mg / g extract.

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Optimization Formulation of Functional Beverages Based on Medium Chain Triglyceride (MCT) and Virgin Coconut Oil (VCO)

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Abstract

Virgin Coconut Oil (VCO) were known as functional food oil which was rich of Medium Chain Fatty Acids (MCFA). VCO contains medium chain triglycerides (MCT/Medium Chain Triglycerides), and contain about 50% lauric acid which is an essential fatty acid in building and maintaining immune system. The aim of this study was to formulate new functional beverage based on Medium Chain Triglyceride (MCT) and Virgin Coconut Oil (VCO). Microemulsion in this study was processed based on Florin Vlad method. The best formula functional beverage was microemulsion from MCT oil, with inggredients water 33%, solvent 15%, surfactant 11.6%, MCT 40%, and CMC 0.4%. MCT functional beverage had characteristics stability tend to decreased at high temperature and viscosity tend to increase during storage. Acid value and peroxide value on products tend to increase especially at storage temperatures of 30 and 37.5 °C. Homogenization using speed of 8000 rpm for 10 minutes droplet size ranged from 4.0 to 12.1 µm.

Keywords:, medium chain triglyceride, physical characteristics, virgin coconut oil

Introduction

Coconut (*Cocos nucifera*) has a strategic role for the Indonesian citizen due to Indonesia palm plantation reached 3.882 million hectares which is the world's largest coconut plantation. Indonesia oil production ranks the second in the world, which amounted 16.146 billion grains (24.4% of world production). But until now the beneficial coconut oil as food is still limited. Coconut mostly used for vegetable oil and edible oil. Variety of products processed from coconuts by products such as coconut milk, desiccated coconut, nata de coco, coir fiber, coconut sugar and activated charcoal can increased value-added of coconut. Development of coconut products as medical products such as virgin coconut oil economically much higher than conventional product of coconut, it means value added. Virgin Coconut Oil (VCO) is pure coconut oil made from fresh coconut meat which is processed at low temperatures or without heating and without addition of chemicals, so that the valuable content in the oil can still be maintained. Virgin Coconut Oil (VCO) were known as functional food oil which was rich of Medium Chain Fatty Acids (MCFA). MCT oil has a content of 52-68%, with the highest amount of MCT in the form of lauric acid ranges from 45-55%. Fatty acids are easily absorbed by the body because the molecular size is not too big as in long-chain fatty acids (Marina *et al.*, 2009; Marten *et al.*, 2006; Nevin and Rajamohan, 2008). According to Marina *et al.*, 2009; Marten *et al.*, 2006; Nevin and Rajamohan, 2008; VCO is able to prevent and treat various diseases such as viruses, bacteria, increase endurance, softens the skin and so on. Among them the ability due to the VCO contains medium chain triglycerides (MCT/Medium Chain Triglycerides), and contain about 50% lauric acid which is an essential fatty acid in building and maintaining immune system. However, oily taste of the VCO is one problem in consuming VCO, moreover

generally the use of VCO is a peroral. So although efficacious, people are still reluctant to consume the VCO directly. To overcome this problem, VCO is made into a beverage emulsion for functional beverage. The basic emulsion was developed into the beverage product by added water and other additives. Unpleasant Ingredients taste can be made more delicious on oral when formulated into an emulsion. In addition, oral emulsions has advantages compared with other oral dosage in terms of absorption and bioavailability (Ansel *et al.*, 1999). Oral emulsion can be a single type: oil in water (o/w) or water in oil (w/o) and double emulsion: water in oil in water (w/o/w) or oil in water in oil (o/w/o).

Emulsion has been widely used in technology cosmetic and pharmaceutical formulation for years (Tenjarla, 1999; Kanebo, 1991). However, it has limited stability of thermodinamic. There is a separation into two liquid phase components. Limited stability of thermodinamic occur because emulsion products release its oil, and eventually phase separation occurs. Unlike emulsions, the microemulsions (including miselar solution) usually a clear dispersion has formed spontaneously without energy input at the time of its constituent components mixed well with each other. Due to the size of oil droplet dispersion of microemulsion is very small 10-200 nm, in many instances the size of the diameter of oil droplet dispersion is less than 140 nm, visible light can not be described and thus appears as a solution isopropik microemulsions are clear/translucent. Microemulsion is dispersions of oil and water and thermodynamically stable, it is stabilized by a surfactant and also a co-surfactant.

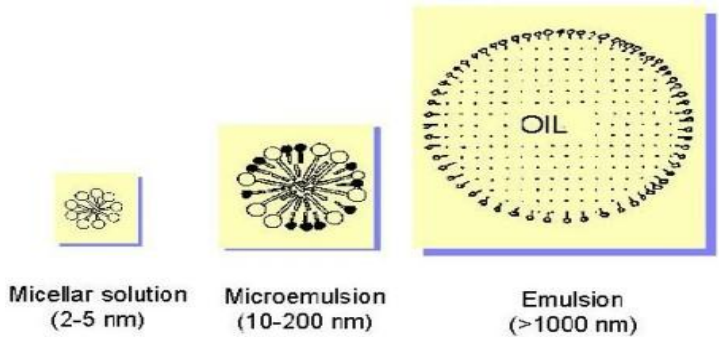

Figure 1. Structures of Micellar solution, Microemulsion and Emulsion

The aim of the present study is to formulate microemulsion of new functional beverage based on Medium Chain Triglyceride (MCT) or Virgin Coconut Oil (VCO) which has stable emulsion with a good physical properties and nutritional value.

Materials and Methods

MCT, VCO, bottled water, emulsifiers (CMC / Carboxymethyl Cellulose), co-solvent (alcohol group and the sugar alcohol), surfactant (Tween 80, and sucrose fatty acid ester / Ryoto Sugar Esters: type 1570 (HLB 15), S-1670 (HLB 16), type P-1570 (HLB 15), food preservatives, chelating, antioxidant, sweetener HFS-55, citrus flavor type R-9904895. Tools that used are homogenizer, magnetic stirrer, viscometer, stopwatch, and material support of packaging bottles, plastic buckets, funnels, plastic jerry cans, and plastic measuring cups. Microemulsions are mixture of water, solvent or co-solvent, oil, and emulsifiers or surfactant/co-surfactant.

Method/technique of processing microemulsions

Based on patent No.U.S. 6902756 B2, June 2005 by Florin Vlad, oil in water microemulsion (O/W) contains about 30% oil, 1-30% surfactant with a hydrophilic-Lipophilic Balance value / HLB 9-18 (the best range 12 - 15), co-solvent is less than 20%, and about 30% is water. The oil as MCT was used about 25-30%. For comparison, VCO used also to create a microemulsion. The solvent was using distilled water or purified water. Co-solvent was using food-grade sorbitol. Surfactant which is used was HLB values between 9-18 (Best 12-15) or 10-18 (Best 13-16). In making microemulsion, surfactants with HLB values outside these ranges must be mixed with other surfactants which have HLB values in the range or other HLB value (not in range), all mixed surfactant system produces the desired HLB value. HLB values of mixed surfactant system are calculated or determined in the same way with a single surfactant HLB value. Surfactants are using ratio 85% Tween-80 (HLB 15) and 15% GMS cithrol (HLB 4,4), so HLB surfactant system is 13,41.

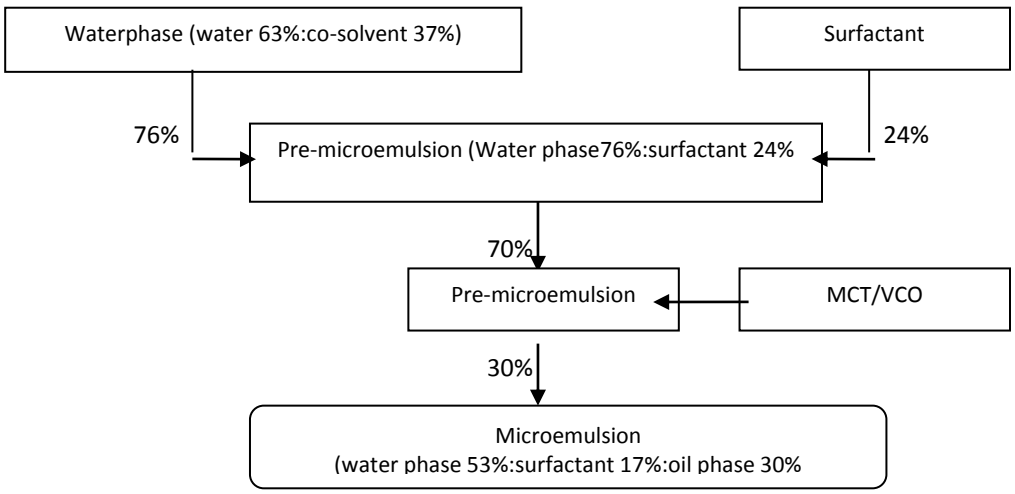

Figure 2. Flow chart of microemulsion processing modification method Vlad

Ingredients for making functional beverage

Functional beverage making started from a basic formulation (BF) given in Table 1. All the experiments were performed in duplo.

Evaluation of physical and chemical properties

Homogeinity and tranlucently (visual observation); stability and emulsion activity observed by centrifugation; viscosity observed used rheometer; diameter/droplet size by microscope electron;acid value and peroxide value based on AOAC (2006) methods

Statistical analysis

Optimization process of beverages microemulsion was using Response Surface Methods (RSM), with parameter that observed was separation percentage and analyzed using Design expert v7.1.6.

Table 1. Amount of inggredients used in formulation of functional beverage

Run	Air (%)	Solvent (Sorbitol) (%)	Surfactant system		MCT (%)	CMC (%)
			Surfactant (Tween-80) (%)	Co- Surfactant (%)		
1	36	17.50	13.85	2.45	30	0.2
2	33.58	17.98	10.42	1.84	35.98	0.2
3	32.80	20	5.78	1.02	40	0.4
4	30	17.50	10.45	1.85	40	0.2
5	30	15	16.83	2.97	35	0.2
6	36	19.80	3.4	0.6	40	0.2
7	30	20	16.83	2.97	30	0.2
8	36	20	11.56	2.04	30	0.4
9	36	17.30	5.35	0.95	40	0.4
10	36	17.50	13.85	2.45	30	0.2
11	30	15	12.41	2.19	40	0.4
12	30	17.50	18.78	3.32	30	0.4
13	36	19.80	3.40	0.6	40	0.2
14	30	20	8.24	1.46	40	0.3
15	36	15	7.48	1.32	40	0.2
16	36	15	15.81	2.79	30	0.4
17	36	15	7.48	1.32	40	0.2
18	30	17.50	18.78	3.32	30	0.4
19	30	15	21.08	3.72	30	0.2
20	33	15	14.19	2.51	35	0.3
21	36	20	7.48	1.32	34.80	0.4
22	36	15	11.56	2.04	35	0.4
23	30	20	16.83	2.97	30	0.2
24	33	15	9.86	1.74	40	0.4
25	30	20	12.41	2.19	35	0.4

Result and Discussion

Formulation of functional beverage

Formulation of functional beverage as microemulsion were based on composition Vlad (2005), which are consists of water 30-36%; co-solvent 15-20%, surfactant 4-24,8%; oil 30-40%; and stabilizer 0,2-0,4% with low agitation technic using inverter (1000 rpm for 60 minute). Surfactant system is using ratio 85% Tween-80 and 15% GMS, so Hidrophilic Lipophilic Balance (HLB) value of surfactant system is 13,41. Inverter is used in surfactant system because the mixing technic is low agitation to minimize foaming. Formulation of functional beverage based on design of experiment using mixture desing from response surface methods (RSM). There are 25 treatment (running) as seen in Table 1.

Homogeneity of microemulsion

All Formulas observed the homogeneity for seven days. The way to prevent droplet back in unity is using high concentrated emulsifier. While homogenize, emulsifier absorb the droplet surface and formed layer that prevent the droplets back in unity. According to O'Brien (2004), food industry must selected suitable emulsifier for each food products. Factors that should be consider when using emulsifier are the number, food characteristics when homogenize, solution property, price, avaibility, legality, stability property, and physicochemical of products which organoleptic accepted by consumer. Based on those factors, the selected formula was formula which has lower concentration of surfactant.

Based on response surface analaysis, the optimum formulas is formulas 14 with percentage inggredient water 33%, solvent 15%, surfactant 11.6%, MCT 40%, and CMC 0.4%. The response equation was Y = 9,52 (Mean Model) and significant ($p<0,05$) (Figure 3). Desirability of the response was 1. Separation percentage of Formula-14 is 0, it means that there was no separation in seven day storage, different with the predicted value of the response 9.52%.

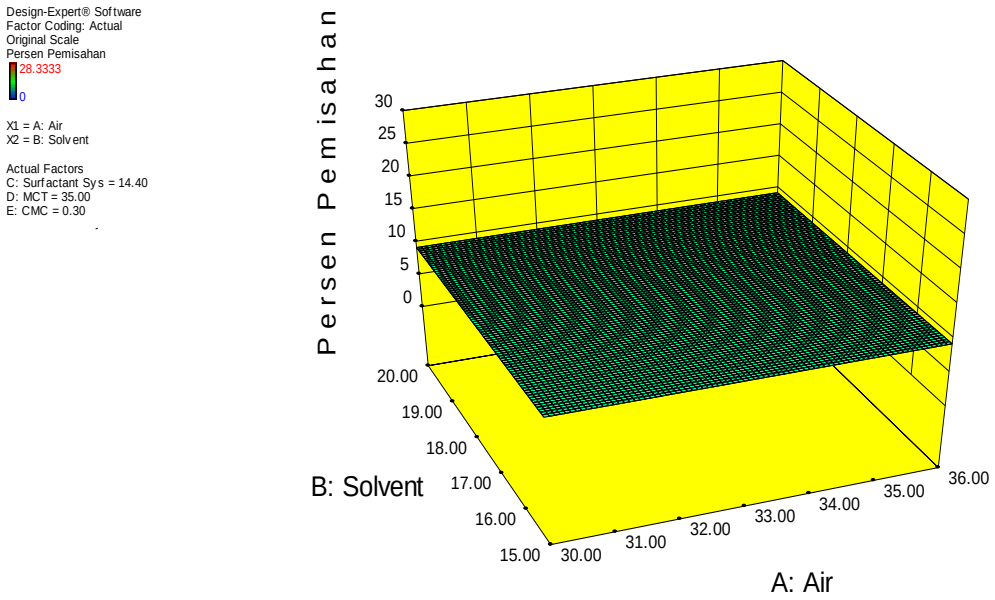

Figure 3**.** Response percent separation from functional beverages formulas

The best formulas were used in physical properties observation including stability, activity, and viscosity of microemulsion for seven days with oil based on MCT and VCO. The observation result can be seen at Table 2.

Table 2. Physical properties of the chosen formulas with MCT and VCO as the oil ingredient

Treatments	Physical Properties Day-1			Physical properties Day-7		
	Viscosity (cP)	Activity	Stability	Viscosity (cP)	Activity	Stability
			MCT as oil			
FM-14	1043.3	100	88.42	1040.0	100	89.93
			VCO as oil			
FV-14	1033.3	100	94.00	1000.0	100	92.00

Based on observation result emulsion stability at Day-7 with MCT as the oil inggredient is lower than VCO. But stability of the MCT formula in Day-7 higher than the first day (D-1) and stability of VCO formulas in Day-7 is decreased from first day (Day-1). The selected formula is FM-14 (using MCT as oil) because the product has better stability for seven day storage (Fig. 4).

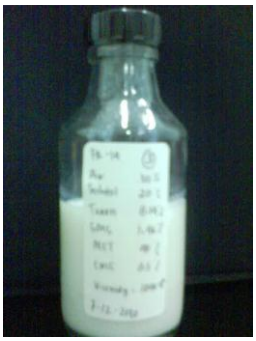

Figure 4. The selected formula (F-14) using MCT as oil

Functional beverage characteristics

Emulsion stability

Based on study shown that emulsion stability was effected by temperature storage. Products stored at ambient temperature (30 ^{o}C) was better than stored at 37.5 and 45 ^{o}C. The emulsion stability rate decreased faster with higher storage temperatures (Fig. 5A). Storage for 14 days, emulsion stability at temperature 30^{o}C was 84.16%, while at temperature of 37.5 and 40 °C were 82.17 and 80.56% respectively, lower than microemulsion on day 0.

Viscosity

Emulsion viscosity tend to increase during storage. Emulsion viscosity at the beginning was 1326.7 cP then rise on seventh day of storage to around 1853.3-1943.3 cP, while the viscosity increase from day 7 to day 14 was not too high (Fig. 5B). High viscosity of an emulsion system will create a durable stability of the emulsion, because of the dispersed phase (droplet) will be prevented from joining with each other (Tantayotai & Pongsawatmanit, 2004).

Acid Value

Acid value on products tend to increase especially at storage temperatures of 30 and 37.5 °C. At the beginning of storage, the acid value of emulsion beverage was 6.47 ml KOH/g. After storage for 14 days at a temperature of 30 and 37.5 ° C acid value slightly increased to 6.64-7.19 ml KOH/g (Fig. 5C). Acid value increased because of hydrolysis of triglycerides which then breaks down into fatty acids and glycerol. According to Buckle (1987) hydrolysis of triglycerides can be caused by the activity of enzymes and microbes.

Peroxide value (PV)

Peroxide value of emulsion beverage increased during storage (Fig. 5D), except in storage with a temperature 37.5 ° C which tends to decrease. The highest peroxide value occurs at a temperature storage of 45 °C increased from 2.54% at day-0 to 3.89% on the 14th day of storage.

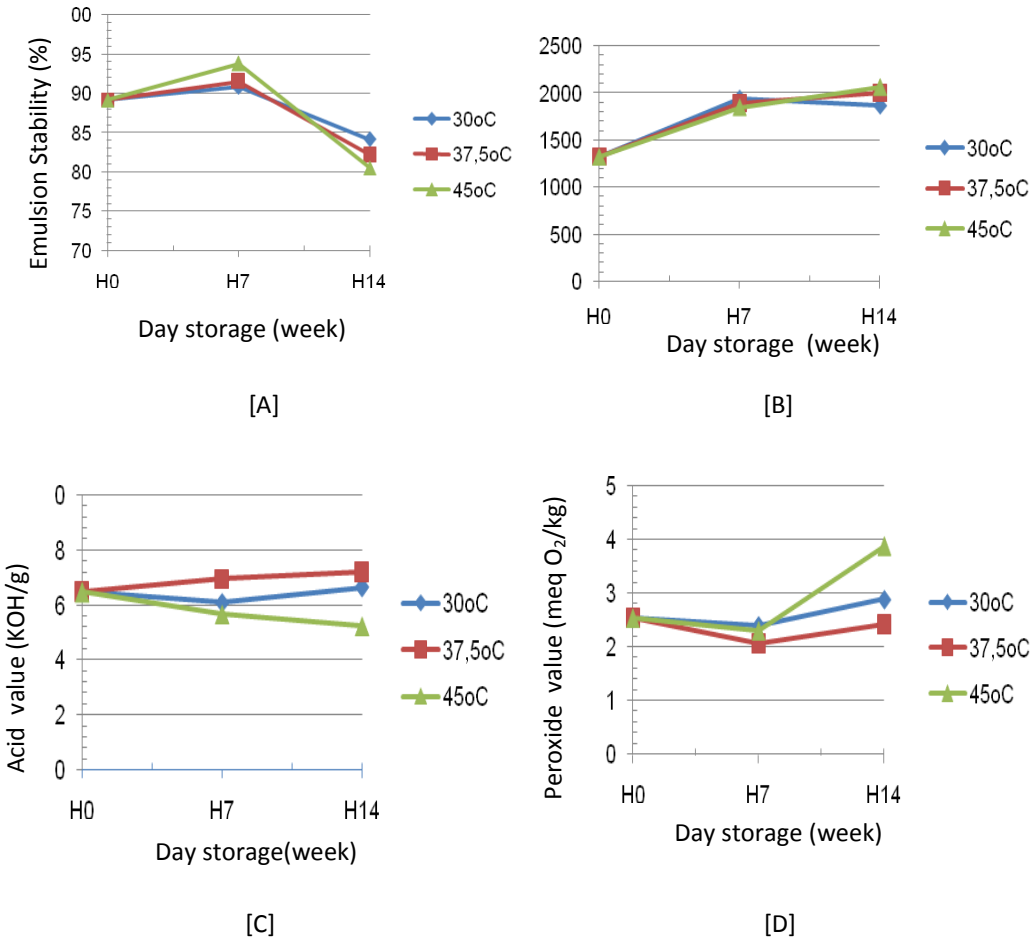

Figure 5. Changes in emulsion of functional beverage at various storage temperatures: [A] stability; [B] viscosity; [C] acid value; [D] peroxide value

Free Fatty Acid (FFA) number (%)

Product emulsion Formula FM-14 stored for 30 days at ambient temperature observed free fatty acids (FFA) content. Average FFA'c content at Day-0 (H-0) was 1.30% and at Day-30 (H-30) was 1.94%. From these results it appears that FFA levels increased along storage.

Fatty acid composition

Beverage products emulsion MCT was an emulsion product designed to contain a minimum of 45% MCT. Largest fatty acid component in the MCT of Cooconut Virgin Oil (VCO) were lauric acid (C12: 0), followed by myristic acid (C14: 0) and palmitic (C16: 0). Before storage, beverage emulsions containing lauric acid 42.65%, whereas myristic and palmitic acids respectively 0.16 and 0.25% (Fig. 6). During storage a slight increase in lauric acid, especially at storage temperature of 37.5 and 45 °C, but at temperature 30 °C decreased slightly because of hydrolysis of triglycerides into free fatty acids and glycerol.

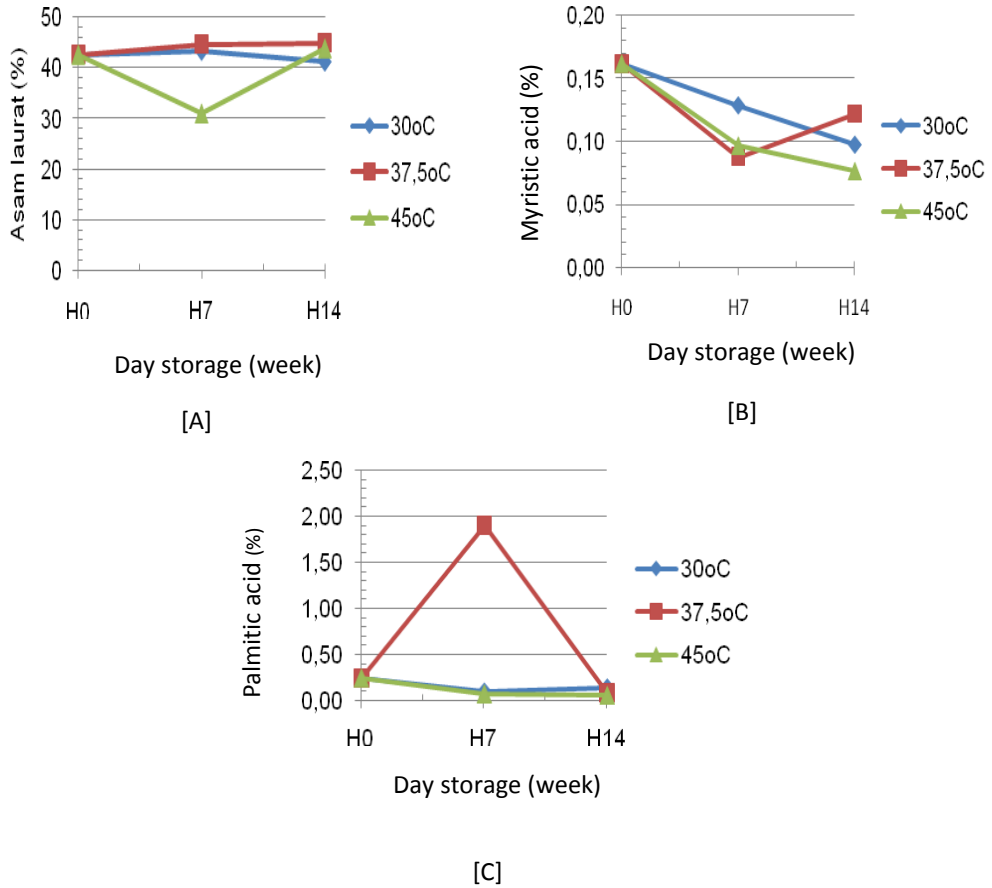

Figure 6. Changes in fatty acids composition of emulsion functional beverage at various temperature [A] lauric acid; [B] myristic acid; [C] palmitic acid

Droplet size

Emulsion formation involves a number of stages depending on the materials and methods used. The process to change two different liquids into an emulsion known as the homogenization process, including using a homogenizer (Loncin&Merson, 1979 in O'Brien, 2004). The main purpose of homogenization is to create an emulsion, which is largely droplet scattered in the optimum range that has previously been set by food processing (O'Brien, 2004). It is thus important to consider factors that determine the size globula in the process of homogenization.

Emulsion formation of two separate fluid obtained from the homogenization of the primary, while the shrinkage droplet in emulsions produced by homogenization of the secondary (O'Brien, 2004). Based on measurements globula conducted sampling in microemulsion-based beverage products of MCT in this study (3-5 observation points). Homogenization using speed of 8000 rpm for 10 minutes droplet size ranged from 4.0 to 12.1 µm fat (Fig. 7).

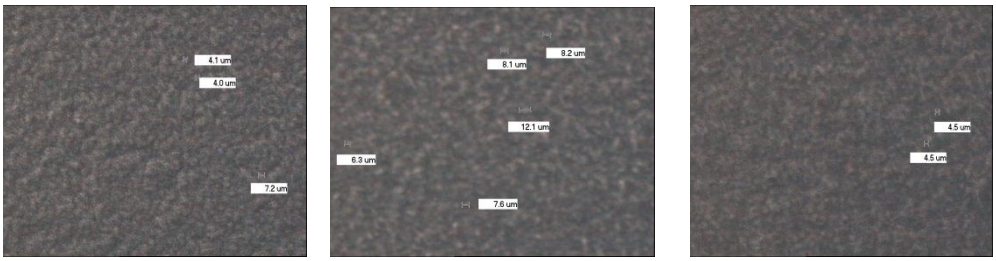

Figure 7. The size of microemulsion droplet homogenizer speed 8000 rpm, 10 minutes

Conclusion

Microemulsion in this study based on Florin Vlad method. The best formula functional beverage was microemulsion from MCT oil, with inggredients water 33%, solvent 15%, surfactant 11.6%, MCT 40%, and CMC 0.4%. Functional beverage characteristics had stability tend to decreased at high temperature, Emulsion viscosity tend to increase during storage. Acid value and peroxide value on products tend to increase especially at storage temperatures of 30 and 37.5 °C. Homogenization using speed of 8000 rpm for 10 minutes droplet size ranged from 4.0 to 12.1 µm fat

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Chemicals and Appearance Characteristics of Noodles Producing from Composite Flour based on Yam (*Dioscorea alata* L.) and The Beans

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Abstract

This research has done at Laboratorium of Crop Production and The Seed Technology Faculty of Agriculture University of Nusa Cendana, from June to August 2010. This research was aimed to find out the Chemical and Appearance Characteriatic of noodles from different composition of composite flour of yam (Dioscorea alata L.) and beans. Its begin with the preparation of collected yam (Dioscorea alata L.), cowpea (Vigna unguiculata) and dolichos bean (Lablab purpureus L.) from local market Oemofa village and stored in refrigerator at the temperature of 14 ^{o}C to 18^{o}C and mung bean (Phaseolus radiatus L.) bought from the trader at the local market. The procedure of work include analysis of nutrients (carbohydrate, protein, fat) and callori of noodle were based on AOAC procedures. The results show that noodles from composite flour based on yam (Dioscorea alata L.) and the beans have height level of nutrition. The noodles with highest carbohydrate level found in flour purple flesh yam - dolichos bean about 75,62%; 16,67% protein content found in noodles from flour based on purple flesh yam - mung bean, the highest fat content was in noodles with flour of purple flesh yam - mung bean which about 11,86%, and the highest energy produced from noodles with flour of white flesh yam - mung bean. Comparing to wheat noodles and noodles from composite flour, nutrition content of noodles produced was better than wheat noodles. The percentage texture of noodles with flour white flesh yam - cowpea was 65%, the percentage taste of its with flour purple flesh yam - cowpea was 55%, the percentage smell of with method (purple flesh yam- mung bean and color of noodles with flour (purple flesh yam dolichos bean is 55%.

Keywords: cowpeas , dolichos bean, noodles, organoleptic propertie , yam

Introduction

The noodle is a food made from any of a variety of doughs such rice, wheat, buckwheat, potato, maize, legume, nut and others, Most raw materials of noodle originated from rice and wheat. Nowadays, the needs of rice and wheats are increases too and the diversity of food are become major needs. There are many biodiverses are explored. One of them is *dioscoreae* (yam) which contains high carbohydrates amount but low of energy and protein (Markus and Oematan 2007) preliminary experiment. In order to increases protein daily intake, legume or beans are consumsed. Moreover, in relation to explore the biodiversity, indigenous bean such as cow peas, dolichos bean are used as a source of raw material of processed food

Basicly noodle is not an original food in Indonesia, but noodles is originated form Asian contries. Now the consumption of noodle increases because people trend to eat a number of energy sources and variuos nutrition sources. To produce noodle, flour is needed as a main material, on the other hand materials for making flour are limited, specially in Indonesia because in fact Indonesia has to import wheat as raw material of flour. One

reason for reduce the relying on wheat is by explored the local biodiversity and local potential crops. As tropical country there are a number of plants can produce flour. One of the plants is yam (*Dioscoreae alata*). Tuber of this crop contains high level of carbohydrate. Based on this fact yam flour may use to produce noodle.

Actually making noodle needs hard flour with protein content. Therefore, supplementation of protein is required. Producing noodle from beans and yam flour only may have unsuitable preferences. Hence, the proportion of flour or the compositions are important. This research was done to get the best proportion of composite based on yam and beans. Related to the quality of noodle made (oh, 1986) this research was done to analyzed the chemical properties of noodles from plam flour added with dolichos flour, to find out the organoleptic characters of noodles making from yam four added with bean flour such mungbean flour, cowpea flour and dolichos bean flour .

Materials and Methods

Raw Materials

Two cultivars of yam were used for making yam flour i.e. purple flesh yam and white flesh yam originated from West Timor Indonesia. Three kinds of dehulled beans were used for producing flour: mungbean (*Phaeseolus radiatus*), cow peas (*Vigna unguiculate*) and dolichos bean (*Lab Lab dolichos*)

Flour Preparation

Dehulled beans were prepared by soaking into the warm water for six hours the manually dehulled. Yam flour were prepared by peeling the tuber, slided, dried it to become crackers then milling it, dehulled beans and Yam were milled using waring blender. After milling flour was sieved by 150 mesh sieve.

Methods

Flour and Noodle Analysis

Moisture content was determined by gravimetric method. Protein was measured by macro Kjedahl AACC method 46-11using a factor of 6,25 to calculate protein crude, and moisture content by method 44-15A (AACC 1983). Carbohydrate amount was calculated by using carbohydrates by different. Fat content was measusred by using soxlet analysis (Apryantono, 1989)

Noodle making

Noodle was made by hand dough with the composition of yam and bean flour 50%. There were 6 treatments which the proportion of yam and bean flour to produce composite flour based on yam and bean. The treatments were:

A= 50% purple flesh yam flour and 50% mungbean flour
B= 50% white flesh yam flour and 50% mungbean flour
C = 50% purple flesh yam flour and 50% cowpea flour
D= 50% white flesh yam flour and 50% cowpea flour
E = 50% purple flesh yam flour and 50% dolichos bean flour

F = 50% white flesh yam flour and 50% dolichosbean flour

Wet noodle were prepared by using 100g of flour (50 g yam flour and 50g of bean flour). One egg and 40mL of water were added to produce dough, then mix it before mold with home industries pasta machine.

Sensory Appearance

The color, tastes, and aroma were evaluated by 40 untrained panelist. The panelists were the students of Agriculture Faculty who always eat noodle. The color, tastes, and aroma of samples were scored by placing mark on a five line scales ranging from 1 to 5 with 1 is extremely dislike and 5 is extremely like (Branger,*et al*, 1999) and (Lawless. & Heymann, 1998).

Results and Discussion

Six types of noodles were produced at the Laboratory of Crop Production and Seed Technology. Each noodle was made with the compositon of 50% of yam flour and 50% of beans flour. After preparing all, the noodles were separated to measured the chemical characteristics and some were boiled for panelist test .

Chemical Properties

There was no differences in protein content of noodle. Protein content ranges from 15.42 – 16,67 %. This fact shows that even noodles produce from different color of yam and different types of beans, but the protein content is same. This is because protein is supply from mungbean flour, dolichos bean flour and cowpeas flour. It is supported by Purwanto (2005), Subagio (2006) and Fachruddin (2007) that mungbean contain 22 % of protein, dolichos bean 17.1 % and cowpea contain 25% respectively. it also presented that with 50 % of substitute bean flour has not given any different to protein content of noodle.

The measuring showed a significant difference of fat content. According to measurement, the highest fat content of noodles was shown by noodle making from purple flesh yam with mungbean flour (11.86 %). On the other hand, noodle from purple flesh yam with dolichos flour showed the lowest fat content (5.34 %). For the remaining noodles, fat contents were different among noodle from purple flesh yam with cowpea flour, white flesh yam with cowpea, dolichos bean and mungbean respectively (Table 1)

After calculated by different carbohydrate. There were significant differences of carbohydrate amounts of the six noodles. Noodle from white flesh yam with cowpea has the highest amount of carbohydrate (Table 1) whilst noodle from purple flesh yam and mungbean flour has the lowest content (67.89 %).

Based on fact, noodles producing from yam flour composite to beans flour had high calories (Table 1). There were no significant differences in energy content of noodles neither noodle made from purple flesh yam or white flesh yam with mungbean, dolichos bean and cowpea. The high content of energy may be supported by measuring the energy of yam either purple flesh or white flesh 363.55 kcal, 360.8kcal respectively. Comparing to rice and instant noodle, these six noodles had double amount of calories, which supported by Merdiyanti (2008) that instant noodle had 350 kcal.

Table 1. Nutritions composition of noodles from composite flour based yam and bean flour.

Treaments	Carbohydrate Content (%)	Protein Content (%)	Fat Content (%)	Energy (Kkal)
Purple flesh yam and Mungbean	67,89a	16,67a	11,86e	801,03a
White flesh yam and Mungbean	75,32d	15,81a	5,89b	809,90a
Purple fleh yam and dolichos bean	75,62d	16,11a	5,34a	809,08a
White flesh yam and dolichos bean	71,37bc	16,55a	8,41c	800,70a
purple flesh yam and cowpea	69,46ab	15,71a	10,71d	800,04a
White flesh yam and cowpea	72,68cd	15,41a	8,60c	808,53a

Note: Number in the same column followed by the same letters was no significant differences at DMRT 5%.

Comparing to wheat noodles and noodles from composite flour, chemical composition of the six noodles produced were better than wheat noodles and noodle from sweet potato such calories, protein content, fat carbohydrate (Nutrition fact of mie telo). The percentage texture of noodles with flour white flesh yam - cowpea is 65%, the percentage taste of its with flour purple flesh yam - cowpea is 55%, the percentage smell of with method (purple flesh yam- mung bean and color of noodles with flour (purple flesh yam dolichos bean is 55%.

Appearance characteristics

Level of acceptance on texture ranging from like to extremely hard to extremely soft (Figure 1). However most of panelists preferred hard to fair hard for all noodles. In fact noodle from purple flesh yam and cowpeas has 55%, white flesh yam and cowpea 50%, purple flesh yam and mungbean flour 45%, white flesh yam and mungbean flour 35%, white flesh yam and cowpea 30% These variation may be caused by the various of these three of bean: mungbean, cowpeas and dolichos bean where the proportion of flour were not affected because the proportion yam and bean flour is 50%

Preference test indicated that perception of panelists for color is ranging from extremely dislike to extremely like (Figure. 2). It shows that perception of panelists like the color of noodles making from purple flesh yam and dolichos bean flour with the highest precentage (55%). This means that panelists really like the light purple color of noodle. Only 10 to 15 % of panelists argued that noodles had bad color.

After smelling the noodles , the preference data showed that 75 % of panelist like a beany flavour of mungbean either combined with purple or white flesh yam flour. (Figure 3) Compare to the two other beans (dolichos bean and cowpea). The testers commented most on noodle from yam flour with cowpea than dolichos bean. This fact may be caused by unfamilar flavour with dolichos bean. This is also supported by Yaw and Powers (1986) . The preference percentage of 75% by panelist mean that they like noodle moderately.

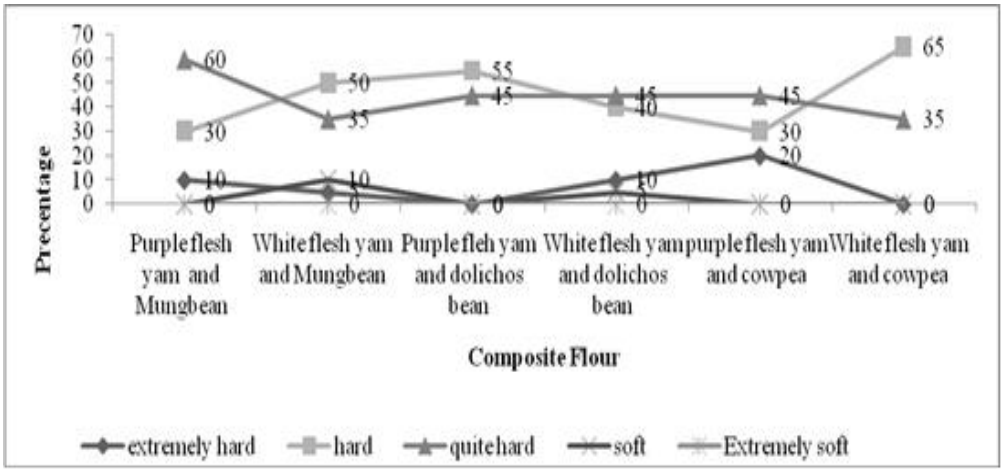

Figure 1. Texture of noodles from 6 types of composite flour

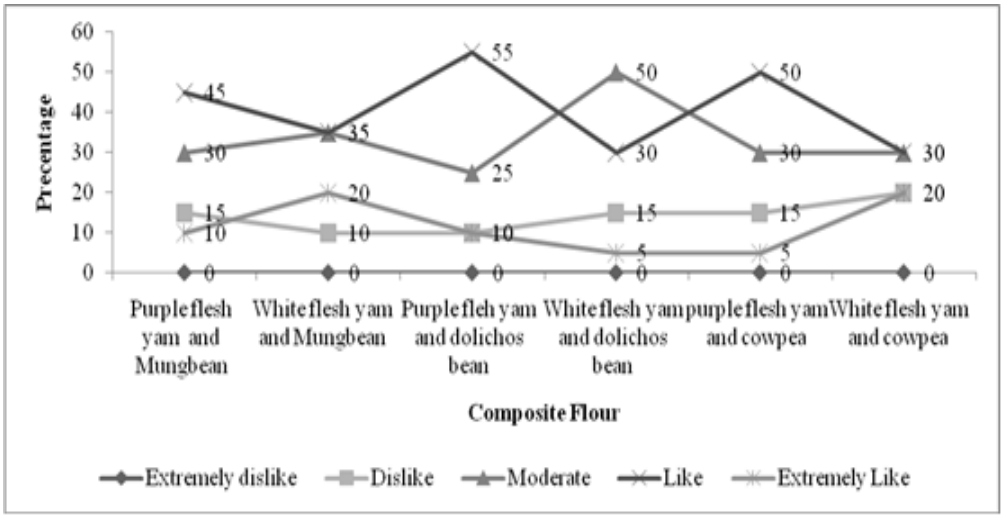

Figure 2. Color of noodles from 6 types of composite flour

Based on taste panel data for the noodle (Figure 4), this indicated that an average 40 –50 % of panelist like to moderately like all noodles. They also gave high mark on noodle from purple flesh yam composite to the three beans flour except noodle from white flesh yam composite with dolichos bean flour. Only few panelist commented that bad taste, However, if comparing to extremely like and bad taste, extremely like taste had more panelists perception than it.

Overall perception on appearance characteristics, noodle made from purple flesh yam combine with mungbean flour and cowpea were more preferable than dolichos. Noodle from white flesh bean had less preferable compared to purple flesh yam.

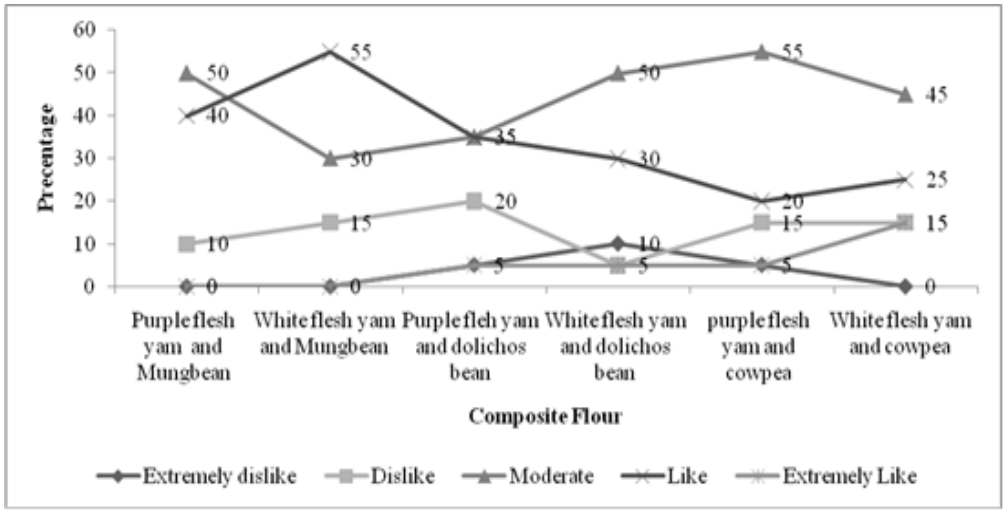

Figure 3. Taste of noodles from 6 types of composite flour

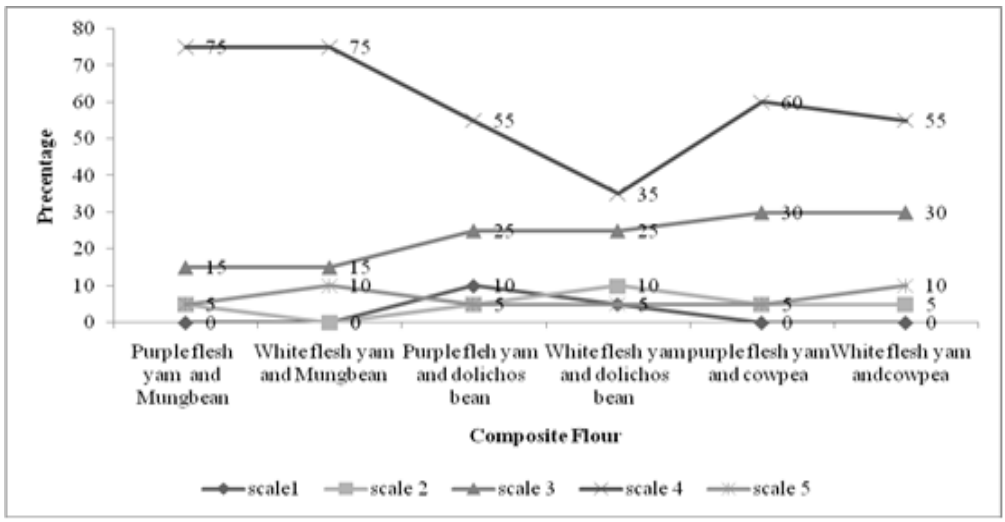

Figure 4. Flavour of noodles from 6 types of composite flour

Conclusion

Noodle produced from composite flour of yam and beans had a high content of chemical such protein, fat and carbohydrate calories. For both color of flesh tuber purple as well white flesh. Beside that, the appearance characters such texture, color, flavour and taste of noodles were most dominant by noodle which made from purple flesh yam composite with mungbean, dolichos bean and cowpea. The highest of appearance characteristics were: percentage texture of noodles with flour white flesh yam - cowpea is 65%, the percentage taste of its with flour purple flesh yam - cowpea is 55%, the percentage smell of with method (purple flesh yam- mung bean and color of noodles with flour (purple flesh yam dolichos bean is 55%.

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The Effect of Type of Packaging and Storage Time on The Quality of Patchouly Oil

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Abstract

Patchouly oil is one of the foreign exchange source from the group of essential oils. Patchouly oil obtained by distillation process dry leaves. One of the factors that cause decline in the quality of patchouli oil is in appropriate storage condition. This study aimed to investigate the effect of packaging types and storage time on the quality of patchouli oil. The method used in this research was descriptive analysis. Type of packaging used for storage were dark glass bottled, clear glass bottles, dark-colored plastic bottles, clear plastic bottles, and cans. Each treatment was repeated 3 times. The parameters measured were levels of Patchouly Alcohol (PA), color changes, specific gravity, acid number, ester number, refractive index, and solubility in 90% alcohol. All treatments during the storage period showed the changes of both in terms of appearance (physical) as well as in terms of chemistry. Research data showed that, the quality of patchouli oil still fulfill the SNI 06-2385-2006 standard. In conclusion, storage treatment using dark glass bottles (A) was the best storage because the patchouli oil showed a fairly stable treatment in maintaining its quality and the packaging using can was not recommended because it caused of corrosion.

Keywords: essential oils, packaging, patchouly oil, storage

Introduction

Patchouly oil is one of the high potential export commodities in Indonesia. With the production of 908.460 kg at 2009 (Dewan Atsiri Indonesia, 2011), this oil contributed to 60% of essential oils export. This cultivation and distillation primary in the western of Aceh Nangroe Darussalam, North Sumatra Province, Tapanuli and Simalungun. However, during the last few years West Sumatra, North Sumatra's eastcoast and West Java cultivated this commodity.

This oil was produced from pogostemon plant (Pogostemon Cablin Benth) using distiller. Distillation method that generally used was water and steam. The biggest oil content was in the leaf, while the straw contained only a few amount of oil.

Patchouly oil widely use as a fixative agent in fragrances industry. A Fixative agent was used to tide the aroma component in the perfume, therefore this component stay longer. Moreover, due to the antibacterial characteristic, the oil can be used as a raw material in pharmacy industry.

The oil aroma was contributed from some component ie terpene, alcohol, aldehid and ester. Patchouli alcohol as an ester is its main constituent that defined the aroma of the oil (Ketaren, 1985). The other component contained in the oil are camphor, eugenol, benzaldehyde, cinnamic aldehyde, cariofilen, α-patchaolena and bulnessen (Trifilieff, 1980). Characteristic for many of these oil constituents is their instability and the ease with which intramolecular rearrangements occur. These properties been a great hindrance to the study of the effect of storage condition to the oil quality.

Therefore, this study aimed to find out the effect of packaging type and storage time on the quality of patchouli oil. Hopefully, the result can be used to predict the best packaging type to maintain its quality standard.

Materials and Methods

Materials

Sample of patchouly oil was obtained from Cooperation of Pelopor Mandiri, Desa Pamulihan, Kecamatan Rancakalong, Kabupaten Sumedang.

Methods

The method used in this research was descriptive analysis. Type of packaging used for storage were dark glass bottled, clear glass bottles, dark-colored plastic bottles, clear plastic bottles, and cans. Each treatment was repeated 3 times with 9 weeks of storage time. The parameters measured were levels of Patchouly Alcohol (PA), color changes, specific gravity, acid number, ester number, refractive index, and solubility in 90% alcohol. Standard measurement for Patchouly oil was used SNI 06-2385-2006 standard.

Patchouly alcohol

Patchouly alcohol content was measured every week along storage time using alcoholmeter. The pathcouly oil was poured in the measurement glass of 100 ml. Alcohol meter was soaked to the glass. Result from alcohol meter was calibrated using Gas Chromatography method.

Specific gravity (SNI 06-2385-2006)

The pycnometer was cleaned by filling it with ethanol and diethyl ether, emptied and dried carefully with dry air, permitted to stand for 30 minutes and weighed accuracy. Then the pycnometer was filled with distilled water, placed it in a water bath at 25^{o}C for 30 minutes. It was dried and weighed. The pycnometer was cleaned by filling it with ethanol and diethyl ether again, emptied and dried carefully with dry air, permitted to stand for 30 minutes and weighed accuracy. Then the pycnometer was filled with the sample, placed it in a water bath at 25^{o}C for 30 minutes, dried and weighed. Specific gravity was difference between mass of pycnometer fill with sample and empty pycnometer divided by difference between mass of pycnometer fill with water and empty pycnometer

Acid number(SNI 06-2385-2006)

The patchouly oil of 2 gram was weighed accuracy in the saponification flask (Erlenmeyer). 10 ml alcohol and 3 drops of phenolphthalein solution were added in the flask. The sample was titrated with KOH 0,1 N. The first appearance of a red coloration was considered the end point.

Ester number(SNI 06-2385-2006)

The patchouly oil of 2 gram was weighed accuracy in the saponification flask (Erlenmeyer). 10 ml alcohol and 25 ml of KOH 0,5 N solution were added in the flask. The sample was refluxed about 1 hour, then cooled. The sample was added with 3-5 drops of phenolphthalein solutionand titrated with HCL 0,5 N. In order to determine the amount of alkali consumed, a blank determination was carried out, observing the same condition but omitting the oil.

Refractive index (SNI 06-2385-2006)

The prisms of the instrument were cleaned using alcohol and ether. Sample was dropped on the prism, closed firmly and allowed for awhile until the instrument and the sample have the same temperature. The result was red directly from the instrument.

Solubility in 90% alcohol (SNI 06-2385-2006)

1 ml oil was introduced into a 10 ml glass-stoppered cylinder, and slowly added 90% alcohol. Addition of alcohol was continued until the solution was clear in it's appearance.

Color changes

Color changes was measured using digital camera with digital color meter software. The sample was recorded it's color in the certain box in order to manage the light.

Results and Discussion

Initial quality of Patchouly oil

This assessment aimed to gain information about the charachteristic of the oil before treathed with type of sorage bottle. Result of the study showed in Table 1. All of the parameters were met the SNI 2006 standard for parchouli oil.

Table 1. The Initial quality of patchouly oil

Quality Parameter	Result
Patchouly Alcohol	29%
Specific gravity	0.9499
Refractive index	1.508
Ester number	8.4150
Acid number	2.4499

Effect of packaging type on Patchouly alcohol content

Data on the patchouly alcohol content of the oil in the different packaging type at 9 weeks storage time reseach were reported on Table 2.

Table 2. Patchouly alcohol content at 9 weeks storage time

Quality Parameter	**Patchouly alcohol content (%)**

	1	2	3	4	5	6	7	8	9
Dark glass bottle	29	29	29	29	29	29	29	29	29
Clear glass bottle	29	29	29	29	29	29	29	29	29
Dark-colored plastic bottle	29	29	29	29	29	29	29	29	29
Clear plastic bottle	29	29	29	29	29	29	29	29	29
Can	29	29	29	29	29	29	29	29	29

The study showed that there were no differences on patchouli alcohol content between all treatment. It may be caused by the short time storage, therefore the change on patchouli alcohol content have not been detected. According to Ketaren (1985), patchouli alcohol is a terpene –O which have a high number of its solubility in alcohol and high stability on oxidation and resinification. This lead to the level off of its patchouli alcohol content.

Effect of Packaging Type on Specific Gravity

The average values of specific gravity of the oil in the different packaging type at 9 weeks storage time reseach are reported in Table 3. It can be founded that specific gravity of the oil in the different type package tended to increase along the storage time. However, these values were nearly same as the value fo specific gravity stated in the SNI 2006 (0,950 – 0,975).

Table 3. Specific gravity at 9 weeks storage time

Type of packaging	Specific gravity (g/ml)			
	0	3	6	9
Dark glass bottle	0.950	0.950	0.955	0.957
Clear glass bottle	0.950	0.951	0.956	0.960
Dark-colored plastic bottle	0.950	0.953	0.954	0.957
Clear plastic bottle	0.950	0.949	0.954	0.957
Can	0.950	0.951	0.957	0.951

Increasing of the specific gravity may due to the oxidation prosess. Some components were oxidated and the product of the process would increase the molecule weight. The component of patchouli oil that proned to oxidate were benzaldehide, patchouly alcohol, patchoulena, and eugenol benzoate ((Sastrohamidjojo, 2002; Guenther, 1974).

Effect of packaging type on refractive index

Refraxtive index is a dimentionless number that describes how the light propagates through the oil. The value varies among the essensial oils depends on the component ot the essensial oils. The longer carbon chain in the oil, the higher number of refraxtive index. The variation of refractive index of the oil in the different packaging type at 9 weeks storage time reseach are stated in Table 4.

Table 4. Refractive index at 9 weeks storage time

Type of packaging	Refractive index			
	0	3	6	9
Dark glass bottle	1.508	1.510	1.508	1.510
Clear glass bottle	1.508	1.511	1.507	1.508
Dark-colored plastic bottle	1.508	1.509	1.507	1.507
Clear plastic bottle	1.508	1.508	1.508	1.507
Can	1.508	1.507	1.508	1.508

The mean value of refraxtive index of the sample using ABBE refractometer between 2.507 – 1.510. Therefore, although the oil was storage for 9 weeks, the refractive index values met the SNI 0602385-2006 standard (1.507 – 1.515).

The research showed that during the storage time the refarxtive index of the oil nearly constant. It might due to the not enough time to change on the composition of the essensial oils component. Among the different type of packaging there was not a different number of refraxtive index too. The refraxtive index that is influenced by the long of carbon chain may change along the long storage time caused by oxidation of some component, however in the range of research storage time, the refraxtive index remained constant.

Effect of Packaging Type on Acid Number

The acid number reflected the free acid in the oil. It is defined as the milligram KOH needed to neutralize the acid in one gram of the oil. The free acid in the oil caused by the oxidation process and ester hydrolysis in the oil. The study resulted that the acid number of the oil between 0.8828 – 2.45 mgKOH/g. According to the SNI 06-2385-2006 standard, the maximum acid number was 5 mg KOH/mg. Therefore, the acid number of sample met this standard. The acid number of the samples were shown in Table 5.

Table 5. Acid number at 9 weeks storage time

Type of packaging	Acid number mg KOH/g			
	0	3	6	9
Dark glass bottle	2.5000	1.2805	0.8864	0.9209
Clear glass bottle	2.5000	1.0018	0.8817	0.9238
Dark-colored plastic bottle	2.5000	0.9648	0.8853	0.8828
Clear plastic bottle	2.5000	0.9648	0.8848	0.9233
Can	2.5000	1.0019	1.0673	1.0687

The study showed that the acid number tended to decreased along the storage time. This might due to natural oxidation reaction whereas the some components including acid in the oil were oxidated and performed ester component. The acid component were sensitive to temperature and oxygen, the present of oxygen in the packaging tended to process of oxydation (Sutresna, 2000).

The acid number of the oil that stored in the can tended to increase during storage time. This might caused by a reaction between can with the patchouli component. Can that made from tin can be a catalisator for oxidation reaction of patchouli constituent (alcohol and aldehyde) became organic acid that lead to increase the acid number.

Effect of Packaging Type on Ester Number

The ester number predicted the organic ester in the oil. It is defined as the milligram of natrium hydoxyde to saponify the ester in a gram of oil. Ester number of the samples oils in the range of 8.415 – 15.479 mg NaOH/g. Since the SNI standard for ester number were 5 – 25, therefore the ester number of the samples met the SNI standard. The ester number of patchouli oil during 9 weeks of storage time were shown in Table 6.

Table 6. Ester number at 9 weeks storage time

Type of packaging	Ester number mg NaOH/g			
	0	3	6	9
Dark glass bottle	8.4150	7.2460	9.8320	13.1120
Clear glass bottle	8.4150	9.3470	9.7950	13.6330
Dark-colored plastic bottle	8.4150	9.3500	9.5780	12.6360
Clear plastic bottle	8.4150	7.7140	9.8240	14.0910
Can	8.4150	12.3790	11.0150	15.4790

The study explained that ester number increased along 9 weeks of storage time. The increasing of ester number might indicate esterification reaction in the oil. Organic acid and alcohol contained in the oil could react each other to perform the ester compound. Since the aroma component majority were the ester compound therefore the higher ester number, the higher aroma constituent in the oil.

Effect of Packaging Type on Solubility in Alcohol

Solubility in alcohol describe the number of volumes of dilute alcohol required for the complete solubility of one volume of oil (Guenther, 1948). The value was determined by component in the oil. The oil that high number of oxygenated component will more soluble in the alcohol, whereas the oil that have high content of terpenes less soluble in the alcohol.

The research found that solubility in alcohol off all samples have the same value (1:1) along the storage time and in the different packaging type. This indicated that the patchouli oil was easily soluble in the 90% alcohol and did not influenced by the storage time. Solubility in alcohol in the oil caused by oxygened component such as patchouli alcohol, norpatchoulenol, and pogostol (Ketaren, 1985). These constituent have polarity almost the same witk the polarity of alcohol (etanol) therefore the oil was easily soluble in the alcohol.

Effect of Packaging Type on Color Change

Color is a quality parameter that common used for consumer to decide the value of the patchouli oil beside it's patchouli alcohol content. The consumer tended to choose the light yellow compared to dark yellow color. In this study, color assessment was used digital camera and was analyzed by digital color meter software with a number of R (red), G (green) and B (blue) value.

Using the visual assessment there was not identified a change in color during storage time. However, by using the digital color meter it was recognized a color change during these

time. The study indicated that the red color increased along the storage time, whereas the green and blue color decreased during that time. Data of the color changes during the storage time were shown in Table 7.

The red color of patchouli oil tended to increase during storage time in all treatment except in patchouli oil that storage in the dark bottle. This may due to high stability of the oil when stored in the dark glass bottle, whereas light can be minimized to reach the oil that could pressdown the oxidation reaction. The clear glass bottle, plastic and can caused the higher red color becaused that materials allow the light came through to the oil, or the component in the packanging itself react with the oil that would produce product with the redish/darken color.

Table 7. Color changes during 9 weeks storage time

Treatment															
Weeks	**A**			**B**			**C**			**D**			**E**		
	R	**G**	**B**	**R**	**G**	**B**	**R**	**G**	**B**	**R**	**G**	**B**	**R**	**G**	**B**
1	77	0	3	69	0	3	72	0	6	75	0	6	69	0	10
2	72	0	7	68	0	9	70	0	7	67	0	6	60	0	8
3	76	0	11	75	0	10	70	0	8	74	0	6	66	0	8
4	76	0	9	76	0	10	65	0	7	74	0	7	65	0	9
5	78	0	9	72	0	6	75	0	7	77	0	5	62	0	8
6	84	0	9	78	0	10	76	0	8	72	0	8	77	0	12
7	80	0	10	78	0	10	84	0	11	79	0	11	66	0	11
8	72	0	9	77	0	7	82	0	8	77	0	9	73	0	11
9	77	0	10	84	0	4	81	0	7	81	0	12	79	0	12

Notes :
R: *Red* (bit)
G : *Green* (bit)
B : *Blue* (bit)
A : dark glass bottle
B : clear glass bottle
C : dark plastic bottle
D : clear plastic bottle
E : can

Conclusions

- During storage time spesific gravity, refractive index, and ester number tended to increase.
- Oil storage in can leaded to significant changes in its chemistry quality parameter and in visual quality (color).
- Dark glass bottle gave was the best package to store the patchouli oil

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The Potency of Banana Tissue Culture Development to Answer the Malnutrition Problems

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Abstract

Bananas contain high amounts of carbohydrates. These fruits are also rich with vitamin A and B6. Furthermore, bananas can be used to produce fibres and as ornamental plants. They can be grown easily and are relatively cheap. These features have made banana as one of the promising candidates to solve the malnutrition problems, especially in vitamin A, pottasium and iron. The objectives of our study were to analyze the potency of bananas to answer the malnutrition problems and identify some steps that need to be done especially in tissue culture technology to produce superior banana seeds. The analysis was performed by SWOT (Strengths, Weaknesses, Opportunities, Treaths) analysis. From this SWOT analysis, the strategies were designed. SWOT analysis results showed that it is possible to solve some malnutrition problems in Indonesia with bananas. The strategies to do that included development of banana varieties that were enriched in vitamin A and iron using tissue culture techniques and establishment of cooperations among farmers, research institutions, industries, and government.

Keywords: banana, malnutrition, SWOT, tissue culture

Introduction

Bananas are monocot plants that contain high amount of fiber. These plants originally come from South East Asia (Robinson & Sauco, 2010). There are some varieties of banana like Kapok, Big Mike, Gros Michel, and the most popular one “Cavendish” (NEP, 2008). Bananas production is increased yearly; this fact is caused by the high bananas demand in the market. Figure 1 shows the increase of bananas production from 1985 until 2002. Economically, bananas (*Musa x paradisiaca L.*) are the most important type of tropical fruit (Naturland, 2001).

Bananas are very rich in carbohydrates, due to that bananas become an alternative for energy source. In an article Vecchi (2010) described, research has proved that just two bananas provide enough energy for a strenuous 90-minute workout. Additionally, 0.68 mg vitamin B6 is available in 118gr banana (The George Mateljan Foundation, 2010). Englberger *et al*. (2003) described that banana contains provitamin A and other carotenoids.

Bananas reproduce vegetatively. To accelerate the vegetative reproduction, tissue culture can be carried out. This technique has been introduced since 1902 by Gottlieb Haberlandt

(Bhojwani & Razdan, 1996). Tissue culture techniques enable researchers to manipulate the reproduction process of plants to get superior seedlings.

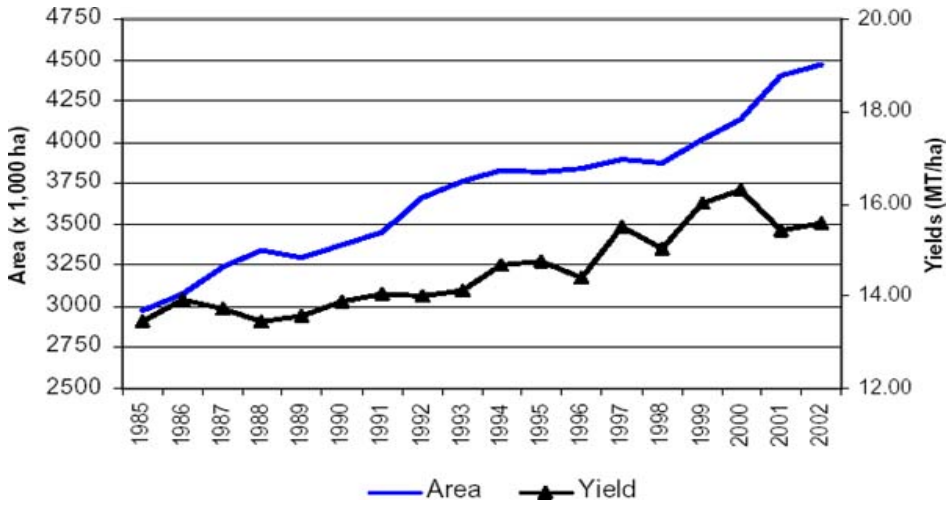


Figure 1. Dessert bananas: world areas and yields 1985-2002(FAO Corporate Document Repository)

Research about banana tissue culture has been established in Indonesia. Hoesen & Gandawidjaja (1993) collected shoots of *Musa bornensis* and propagated the shoots using MS basis media combined with Cytokinine BA and sulphate. Meldia Y from 1995 until 1996 studied the effects of carbon sources and macro nutrient content to the banana germplasm storage. The research result indicated that the buds which was grown on glucose and sucrose media without MS (Murashige and Skoog) ingredients do not form roots, but still produce shoots with small number compared with media that contains MS ingredients (Indonesian Ministry of Agriculture, 2009). Nisa & Rodinah (2003) conducted a research on the interaction between a mixture of Naphthalene Acetic Acid (NAA) and kinetin on the growth of some bananas strain (kapok, raja, and mauli). Treatment with NAA 0.4 mg/l and kinetin 6 mg/l in mauli cultivars gave the highest results on the percentage of live explants which is 87.5% and the lowest percentage of contamination that is less than 5% while the provision of NAA 0.8 mg/l and kinetin 9 mg/l in kepok cultivars showed the fastest time of callus growth that is 11 days. In their research Nisa and Rodinah used banana heart as explants.

This background has been drawing us to further investigate the potency of banana tissue culture to answer the malnutrition problems in Indonesia. Is it possible to make banana as an agent to solve some malnutrition problems? And what are the strategies that can be implemented?

Materials and Methods

The study was conducted by data collection to be analyzed based on SWOT (Strength Weakness Opportunity Threat) and TOWS (Threat Opportunity Weakness Strength) analysis. Data collection was aimed to obtain primary and secondary data. The primary data was obtained from the banana's seed multiplication activities in the laboratory and discussion with managers and staff from UPT Balai Benih Induk Pertanian dan Kehutanan

Dinas Pertanian dan Kehutanan DKI Jakarta about the banana tissue culture prospects. Secondary data was obtained from journals and articles about bananas and tissue culture.

To build the real analysis of each aspect assessed in SWOT analysis, some points need to be reviewed. There are four aspects considered in the SWOT analysis, the strengths and weaknesses which were the measures of internal potency, and opportunities and threats which were the tools to measure the external potency. The SWOT analysis results are used in TOWS analysis. In TOWS the combination of internal and external factors were used to design the strategies for the development of banana tissue culture in SC Tissue Culture Laboratory (Arafat, 2005).

Results and Discussion

SWOT Analysis of Banana Tissue Culture Development to Answer the Malnutrition Problems

Strengths

Researchers as human resources play an important role in the development of tissue culture in Indonesia. Many Indonesian researchers have successfully performed the tissue culture of banana, orchid, taro, and other crops. In addition, the researchers are able to manipulate and produce a superior seeds of plants by tissue culture.

Various studies conducted by national and international research agencies can be adapted to develop banana tissue culture. These studies can be accessed online or through books and journals. With holistic and actual knowledge of banana tissue culture, banana tissue culture can be optimized to produce a superior banana seed.

Tissue culture techniques enable the production of similar crop seeds, with significant amounts and in a relatively short time. This is the potential power of banana tissue culture development. The other advantages of banana tissue culture are: pest and disease free seedlings; uniform growth, increases yield; allow the maximum land use in low land holding country like India; no staggered harvesting; and 95% until 98% plants bear bunches (Jain Irrigation System Ltd, 2008).

Nutritional content of banana is high and can be assumed as strength in the development of banana tissue culture. Mukhtasar (2003) mentioned that banana has economic value mainly in its fruit. Data showed 118 g banana has 2.83 g fibre, 95.58 IU vitamin A, 0.37 mg iron, 22.54 mcg folate and 0.68 mg vitamin B6 (The George Mateljan Foundation, 2010). Folate plays important rule in the formation of DNA, and it is suggested for pregnant woman to consume banana in order to support the foetus development (Brigham and Women's Hospital, 2006). Besides that, bananas also contain potassium and pectin which are protective against heart disease, and the pectin alone can help maintain the gastrointestinal tract (Mitrea, 2007).

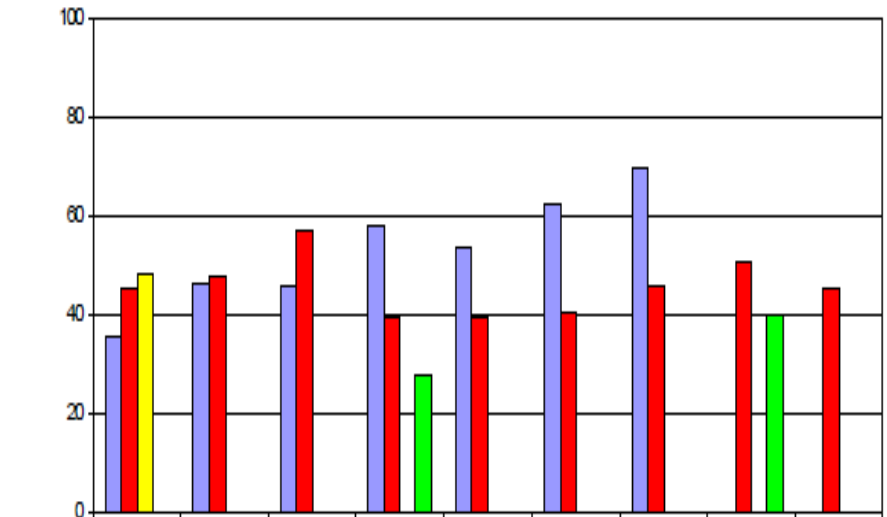
Weaknesses

The researcher's result in tissue culture still not produced in big scale. This is an impact of the long and multilevel examinations to observe the plant's quality and the safety. These examinations need a strong financial support. However the laboratories in Indonesia have a relatively limited budget.

According to Indonesian Ministry of Agriculture (2009) there are a numerous researches about tissue culture in the same objects which were conducted in different laboratories. Even though, there is no cooperation between researchers from different research agencies.

Opportunities

World Health Report in 2002 identified a lot of people suffer from iron, vitamin A, zinc, and iodine deficiencies. These problems are found especially in developing countries (WHO, 2002). VAD (Vitamin A Deficiency) and IDA (Iron Deficiency Anaemia) also occur in Indonesia (Atmarita, 2006), Figure 2 shows the prevalence of IDA in Indonesia based on the data from National Health and Household Survey (NHHS) until 2001. IDA adversely affects cognitive performance, physical growth of infant, immune status, morbidity from infection, and the physical capacity and work performance of adolescents and adults (WHO 2001). VAD lead to night blindness, xerophthalmia, keratomalcia, blindness and immune depression problems (Mayer, *et al*. 2008). (The DPD's chairman Adrianto Persagi said, the invisible malnutrition phenomena is still high. Persagi explains "At the national level, more than thirty percent of children under five years old are stunted (height growth was based on age), due to low levels intake of macro and micronutrients (Candra, 2009). Malnutrition case is one opportunity for the banana tissue culture, because banana contains high amounts of nutrient and is relatively cheap.



	0-4 years	5-9 years	10-14 years	15-44 years	45-49 years	55-64 years	65+ years	Pregnant women	Lactating mother
M-1995	35.7	46.4	45.8	58.3	53.7	62.5	70.0		
F-1995	45.2	48.0	57.1	39.5	39.5	40.5	45.8	50.9	45.1
M+F 2001	48.1								
M-2001				27.9				40.1	

Figure 2. Prevalence of Iron deficiency anaemia (NHHS 1995 and 2001) by Atmarita 2009

Table 1. Estimated prevalence (percent) and number (million) of stunted children from 1980 to 2005

UN Regions and Subregions	1980		1985		1990		1995		2000		2005	
	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.
Africa	**40.5**	**34.78**	**39.2**	—	**37.8**	**41.68**	**36.5**	**44.51**	**35.2**	**47.30**	**33.8**	**49.40**
Eastern	46.5	12.88	46.9	14.83	47.3	17.13	47.7	19.28	48.1	22.03	48.5	24.41
Northern	32.7	6.01	29.6	6.01	26.5	5.55	23.3	4.90	20.2	4.44	17.0	3.86
Western	36.2	9.034	35.8	10.51	35.5	11.99	35.2	13.47	34.9	14.74	34.6	16.03
Asia	**52.2**	**173.37**	**47.7**	**169.72**	**43.3**	**167.66**	**38.8**	**143.49**	**34.4**	**127.80**	**29.9**	**110.19**
South Central	60.8	89.36	56.5	93.45	52.2	93.36	48.0	83.62	43.7	78.53	39.4	72.28
South-Eastern	52.4	27.71	47.5	26.47	42.6	24.24	37.7	21.51	32.8	18.94	27.9	15.78
Latin America and the Caribbean	**25.6**	**13.19**	**22.3**	**11.87**	**19.1**	**10.38**	**15.8**	**8.59**	**12.6**	**6.82**	**9.3**	**5.11**
Caribbean	27.1	0.92	24.4	0.86	21.7	0.81	19.0	0.71	16.3	0.61	13.7	0.51
Central America	26.1	3.87	25.6	3.81	25.0	3.87	24.5	3.94	24.0	3.92	23.5	3.82
South America	25.1	8.38	21.1	7.35	17.2	6.05	13.2	4.55	9.3	3.16	5.3	1.84
Oceania	Data not available											
All Developing Countries	**47.1**	**221.35**	**43.4**	**220.10**	**39.8**	**219.73**	**36.0**	**196.59**	**32.5**	**181.92**	**29.0**	**164.70**

Source: WHO (1999a).

Table source: Gillespie, Stuart and Lawrence Haddad Attacking the Double Burden of Malnutrition in Asia and the Pacific, 2001.

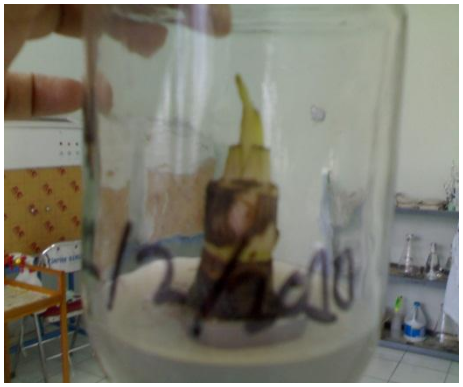
National research agencies have conducted research on banana tissue culture and other crops. Those agencies include Tissue Culture Laboratory Coffee and Cocoa Research Centre in Jember, Horticulture Research and Development Centre in Jakarta, BPPT Tissue Culture Laboratory in Serpong, and national universities. The information obtained from these studies open up opportunities for researchers to develop banana tissue culture.

Indonesia is a tropical country, and ideal for growing banana plants. The opportunities provided by natural condition support the development of banana tissue culture in Indonesia.

Indonesian biodiversity, as an asset, is a good opportunity for banana tissue culture. Biological diversity in bananas which is consisted of 102 cultivars (Suyanti & Supriyadi, 2008) proves the richness of biodiversity in Indonesia. This biodiversity has to be utilized for the nation welfare. Diverse collection of banana cultivars enables the exploration of bananas to find the appropriate seeds with high nutritional value.

Threats

The threat of banana tissue culture related to the problems in the tissue culture process are; contamination that caused by fungi and bacteria, browning due to phenolic activity in plants, a dwarf plants that lose its palisade tissue in the leaf produced by the explants, and genetic variability (BPPT Tissue Culture Laboratory, 2010). This threat will inhibit the tissue culture process.



A

B

Figure 3. Threats during tissue culture process in Seed Centre Tissue Culture Laboratory: A. Browning process in the bud of banana, B. A banana bud contaminated with fungi.

Bananas which were imported from the United States, Cuba and other countries have superior quality. In addition to the sweet taste and the size, imported bananas are better than the local counterpart. The quality of these imported bananas is a threat for the commercialization of banana fruits produced by national farmers.

There are several obstacles that need to be watched carefully during banana growth according to the Executive Summary of National Discussion in The Development of Leading Indonesian Fruits (2003) mainly; bananas are generally vulnerable to be attacked by soil pets, low banana productivity with varying quality of its short shelf, banana needs a long time for breeding process, the availability of genetic material for breeding purposes is still low, genotype diversity of bananas is very high causing difficulty in identifying the inheritance patterns that controls each idiotipe, lack of qualified banana seeds availability, cultivation technique not yet mastered, and soil borne disease attacks like *Fusarium spp.* and *Ralstonia solanacearum* has not been encountered in large scale plantation.

TOWS analysis of banana tissue culture development to answer the malnutrition problems

There were some strategies designed based on SO (Strengths and Opportunities): Develop banana cultivars with high provitamin A, pectin and iron content in order to solve the national malnutrition case is a strategy that can be implemented by the laboratory. Banana was a candidate to solve the malnutrition problem for the Iron Deficiency Anaemia and Vitamin A Deficiency, because it contains high amounts of vitamin A and iron; Researchers from a research agency can visit other national research agencies to look for superior banana seeds or optimize the conditions in banana tissue culture; Biodiversity of banana in Indonesia is high and this power can be used for the production of superior bananas. National researchers can perform the most qualified banana tissue culture from these bananas to handle the national food needs and supply seeds for commercial banana plantations.

Conduct cooperative researches with national research agencies is a strategy that can be implemented by researchers according to WO (Weaknesses and Opportunities) analysis. This step will enable a knowledge transfer process, to eliminate problems which have been faced in the laboratory during plants processing and avoid the same research to be done.

Beside that to solve the financial problems ther must be a working team that include ABG (Goverenment, Busniess and Academic) that put a focus on banana fruits production.

There were several strategies that could be applied for the development of banana tissue culture based on ST (Strengths and Threats). Firstly, ensure the transfer space was sterile to reduce the risk of contamination. Secondly, initiate the tissue culture process with banana heart as explants to minimize the contamination (Nisa & Rodinah, 2003). The banana heart is less contaminated with endogenous pathogens compared with the shoot, that fact lead to a lower contamination number in initiation process with banana heart as explants. Thirdly, provide a suitable condition for bananas plantation, to produce superior fruits.

Limit the banana collection was a strategy that could be implemented according to WT (Weaknesses and Threats) analysis. Restrict the banana collection aims to let the laboratory be focused on the development of banana quality. Laboratory should emphasize the orientation on the quality to eliminate various threats and vulnerabilities.

Conclusions

Based on the TOWS analysis there were some efforts that need to be done for the development of banana tissue culture to answer malnutrition problem. Such effort can include develop bananas with high vitamin A and iron content; national research agencies was supposed to conduct cooperative research and collaborate with people from business sector; ensure the transfer room was sterile to reduce contamination during tissue culture process; perform banana tissue culture with banana heart as explants; limit the collection of cultured banana and emphasize the orientation on the quality of the produced bananas.

Banana tissue culture, for large scale bananas production is urgently needed. This is the potential way to solve the Vitamin A Deficiency and Iron Deficiency not only in Indonesia.

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Quality Degradation of Mashed Red Chilli Based on Capsaicin During Processing

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Abstract

The objective of this reseearch was to analyze and identify the relation between processing temperature and heating time during processing of mashed red chilli to minimize degradation of capsaicin. Factorial Complete Randomized Design with three replications was used. The treatments were the processing temperature (70^0C, 80^0C, 90^0C) and the heating time (10, 15, 20 and 25 minutes). Parameters analyzed were capsaicin content, pH and particle size. Data were analyzed using anova, Duncan Test and surface response graph. Results showed that processing temperature of 70^0C and 80^0C indicated not significant difference in capsaicin content of 0.05%, while processing temperature of 90^0C caused capsaicin content dropped sharply, its about 0.03%. Result also showed that the longer heating time, the larger capsaicin content lost, however heating time of 20 minutes at 80^0C caused capasaicin lost lower than processing time of 25 minutes. During processing, pH content and particle size were stable. Research concluded that in order to prevent the quality degradation of mashed red chilli during processing, temperature of 80^0C for 20 minutes is the best processing temperature and heating time to process mashed red chilli to minimize capsaicin content of 0.072 %, pH of 5.54 and particle size of 0.0028 mm.

Keywords: capsaicin, mashed red chilli, , particle size, quality degradation

Introduction

Red chilli (*Capsicum annuum* L.) is a vegetable that can be marketed in the form of fresh and processed. In general, red chilli used as a flavoring food, mixed dishes and industrial raw materials. According to the Central Bureau of Statistics (2007), consumption levels of Indonesian society for red chili is 4.16 kg / capita / year. Chili needs will increase in line with the population. As a horticultural commodity, a red chili is categorized as a perishable product with its shelf life just four days after harvesting. In the sort shelf life of chili affect the supply of fresh chili in the market. These fluctuations also affect the selling price of chili. In the harvest season, chili price is relatively low ranging from Rp 12.000,- up to Rp 16,000,- / kg, but after the harvest of red chilli prices reach Rp 70,000,- to Rp 80,000,-/kg. Even in January 2011, the price of chili at Rp 110,000, -/kg. To overcome this problem needs to be done pickling chilli that are simple, easy, cheap and durable.

Processing of dried red chili such as chili, chili powder or kocukaro, chili oil or oleoresin, chili and pasta sauces have been made, and costs required for processing is relatively high. For consumers daily consumption, especially cathering, restaurants, mom-housewife, need chili peppers in the form of semi-solid product, mashed chilli is more preferred because of practical use, it's cheap, its widespread use and feasible. Research results of Septiarida

(2003) showed that by processing of minced chili was more profitable than processing a dried chili pepper.

As an industrial raw material, mashed red chilli can be processed into refined products such as bottles of hot sauce, spicy sweet, hot sauce bottles beauty, sweet chilli sauce, extra spicy, seafood sauce, sambal fried chicken, sambal plows, balado sauce, chili sauce, pasta chilli, and seasoning ingredients for canned food products (beef rendang, fish, etc.). In the Middle East countries and India, they generally use mashed chili for making chili curry, sauces, meat and pasta mixture, and processing of oleoresin-curry (Ahmed *et al*., 2000).

The need of mashed red chili as industrial raw materials will continue to rise because of increased demand and consumer tastes for processed chilli products that range. In addition, the red pepper processing industry continues to create diversified products according to consumer tastes. It require quality characteristics of raw materials, especially color and pungency. For example, spicy chilli pungency require different indices with sweet or spicy sauce is very spicy (hot spicy). Likewise, the color needs and level of spiciness to plow different condiment for raw materials sardines and rendang. During this chilli suppliers have not been clear standards for the quality criteria required for industrial raw material needs of chili so that the quality products varies at each stage of distribution; so it will affect the consistency of product quality. Though the quality indicator is determined by the color of red chilli and spiciness level reflected by the content of capsaicin.

According to Hasbullah (2007), mashed red chilli processing was very easy. Processing is usually performed conventionally without the heating process, but the mashed chilli have relatively short shelf life ± 7 days with 40-50% moisture content. After a week of storage of mashed chilli begin to occur the damage is characterized by slightly sour taste, distinctive aroma not deviate chilli, red color changed to brown, less attractive appearance and the surface covered chilli mushrooms (Kumara, 1986).

Food processing technology with blanching involves heating, exhausting, and pasteurized to extend shelf life. The results of Renate (2004), the heating process in the manufacture of red chilli puree made exhausting at a temperature of 82^0C for 10 minutes to extend the shelf life of 2 to 3 months. The heating process can affect the value of nutrients such as vitamins, colors and content of capsaicin (the level of spiciness) especially when heat is used too high or too long. During the heating process it changes color from bright green to olive brown on green chilli puree form changes in chlorophyll a pheophitine. Degradation of color and capsaicin increases with increasing temperature (Roche *et al*., 1993, Ahmed *et al*., 2000 and Ahmed *et al*., 2002). Blanching time and temperature can affect the color degradation chili puree, pea puree and mango puree (Ichier *et al*., 2006, Ismail *et al*., 2006, and Vasquez *et al*., 2007). Changes in red to brownish red color occurs in a modified red chilli puree with carotenoids from melinjo skin after two months of storage (Renate, 2008). In addition to color and capsaicin degradation by heating, there is also sugar-reducing sugar reaction with the amino acid that produces a brown color (browning). Therefore, the use of heat need to be tailored to the characteristics of food ingredients in this chili grind.

The objective of this research was to analyze and identify the relation between processing temperature and heating time during processing of mashed red chilli to minimize degradation of capsaicin.

Materials and Methods

Research was conducted at the Laboratory of Process Engineering of Agricultural Products Processing, Science Laboratory University of Jambi, from Maret to June 2011. Raw materials used in the processing of mashed red chili was fresh curly red chili comes from the highlands of Kerinci regency in Jambi and supporting material such as pure capsaicin powder, sodium chloride, citric acid, sodium benzoate, as well as materials for analysis such as methanol, acid chloride, etc. .

The equipment used to processed was chopper of red chili, stove, waterbath, and bottles. The equipment used for analysis was the pH meter Hanna Instruments brand type 711-8519, spectrophotometer Genesys 10uv brand, brand strirrer Biby Strrer magnetic type B 212, Analytical balance Precisa brand At 220 A type, a digital brand hand refractometer Atago S / Mill-E.

Research used Factorial Complete Randomized Design with three replications. The treatments consisted of 2 factors. The first factor was the processing temperature (70^0C, 80^0C, 90^0C) and the second factor was heating time (10, 15, 20 and 25 minutes). All experiments will be repeated 3 times.

Implementation research

The first criteria chosen for red chili are those not defective, full red colour and uniform size (in 1 kg there are 40 to 45 fresh red chillies). Red chilies that have been sorted and separated from the stems and then weighed 150 g to each set of experiments, then washed with running water and drained. Furthermore, red chili was blanced in water at temperature of 80^0C for 3 minutes. Red chili then was ground by using blender or chopper. During grinding, 0.5% citric acid, benzoic acid 0.1% and 5% natrium chloride were added. The percentage is calculated from the total weight of chilli used. The result mashed chili was then analyzed for capsaicin content, pH and particle size. Data were processed with anova, Duncan Test and surface response graph.

Results and Discussion

Capsaicin content

Heat process of mashed red chilli can cause degradation of capsaicin content. Heating process temperature of 70^0C for heating time 20 minutes, produced mashed red chili with capsaicin content higher than the heating time of 10, 15 and 25 minutes. Processing temperature of 80^0C for 20 minutes caused capsaicin content was relatif stable compare to its fresh chilli of 0.199 %. However, processing temperature of 90^0C caused capsaicin content dropped sharply. The heating process can affect the value of nutrients such as vitamins, colors and content of capsaicin (the level of spiciness) especially when heat is used too high or too long. Degragation of capsaicin of mashed red chilli during heat process illustrated on figure 1.

Degradation of capsaicin increases with increasing temperature (Roche *et al*., 1993, Ahmed *et al*., 2000 and Ahmed *et al*., 2002). Blanching time and temperature could affect the color degradation of chili puree, pea puree and mango puree (Ichier *et al*., 2006, Ismail *et al*., 2006, and Vasquez *et al*., 2007).. In addition to capsaicin degradation by heating, there is also sugar-reducing sugar reaction with the amino acid that produces a brown color

(browning). Renate (2008) stated that red chilli puree made by exhausting at a temperature of 82^{0}C for 10 minutes could extend the shelf life 2 to 3 months.

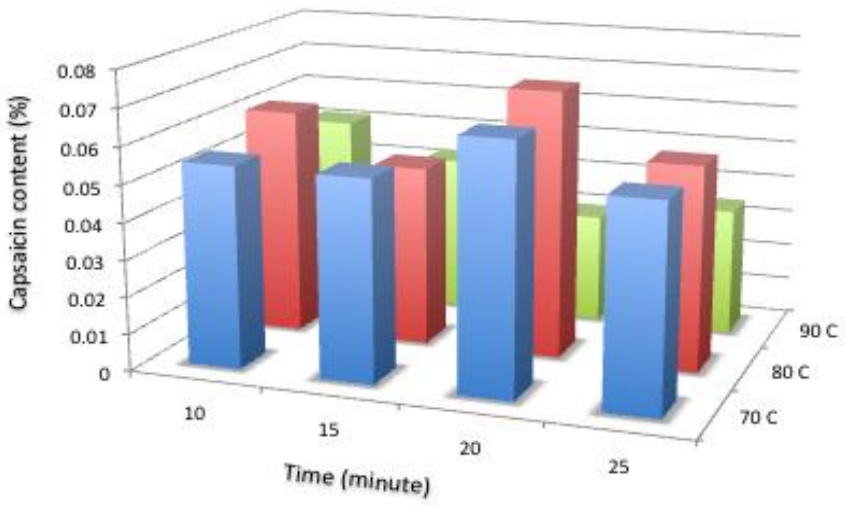

Figure 1. Capsaicin degradation of mashed red chilli during heat process

pH

The processing temperature and heating time was not significantly different on pH of mashed red chili. The highest pH is 5.33 and the lowest pH is 5.75. Most mashed red chilli had the same pH. It means that at the beginning of the process, temperature and heating time can't affect pH. It is assumed that pH may change when mashed red chilli store for several weeks.

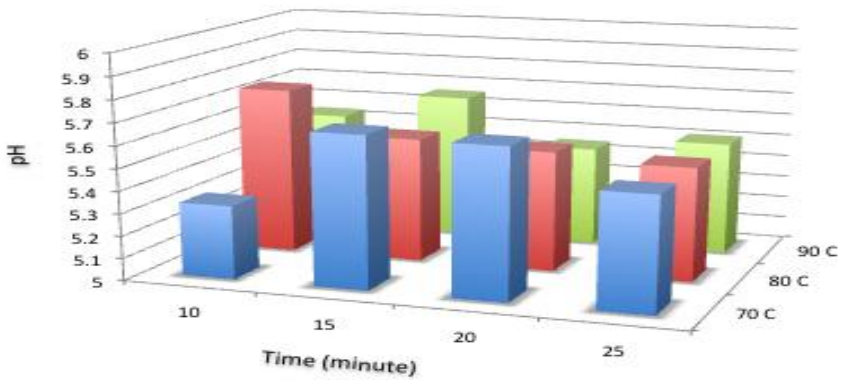

Figure 2. pH of mashed red chilli during heat process

Particle size

Particle size of mashed red chilli affects heat penetration on degradation of capsaicin.. When processing temperature was high, it was assumed that the particle size of mashed red chilli increased and it caused degradation of capsaicin fastly. Figure 3 illustrated the changing particle size of mashed red chilli during heat process.

Heating process temperature of 70^0C, particle size was stable, as a result degradation of capsaicin was stable. Processing temperature of 80^0C and heating time of 10 and 15 minutes, caused particle size was larger than heating time of 20 and 25 minutes, as a result degradation of capsaicin was low. On the other hand, processing temperature of 90^0C caused degradation of capsaicin declined due to larger particle size.

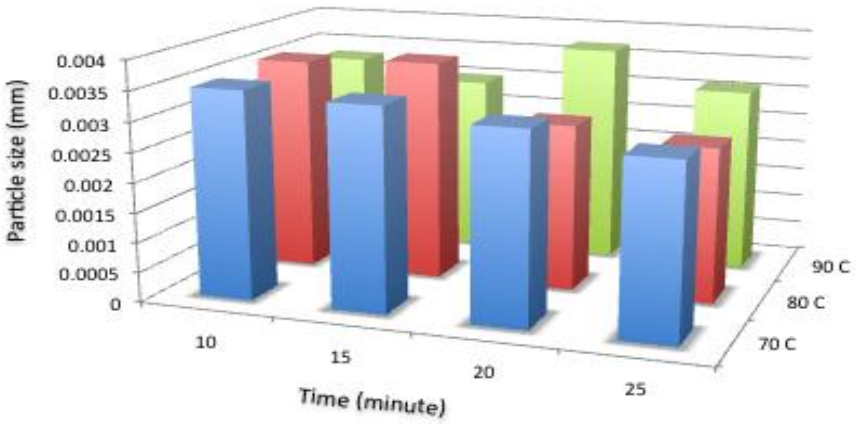

Figure 3. Particle size of mashed red chilli during heat process

Conclusion

Research conclude that temperature of 80^0C and heating time of 20 minutes was the best treatment to process mashed red chilli in order to minimize its capsaicin content of 0.072 %, pH of 5.54 and particle size of 0.0028 mm. It is recommended to used temperature process and heating time above to process mashed red chilli in order to prevent the quality degradation of mashed red chilli during processing,

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Improving the Quality of Meat from Old Cattle through Extended Cooking

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Abstract

The experiment investigated the effect of extended cooking on beef quality. The effects of position of the raw meat sample within the loin, cooking time, and sex on the WB shear value and cooking loss were studied. Loin samples were collected from nine randomly selected steers and cows. Loins were randomly allocated to cooking times of 2.5, 5, 10, and 15 hours. Data were analyzed with the mixed model procedure of the SAS system. The model contained fixed effects of sex, cooking time and position of loin and all first order interactions. Loins were used as a random effect, within which sex was nested. The result indicated that cooking time significantly affected the WB-shear value (P<0.01), whereas loin position and sex had no such effect (P<0.1). WB-shear value declined from 5.27 ± 0.19 kg to 3.53 ± 0.19 kg as cooking time increase. Posterior end of loin had higher values than those from anterior end (4.70 ± 0.19 kg vs. 4.20± 0.19 kg; P<0.1), and steer had lower value than cows (4.10 ± 0.21 kg vs. 4.71± 0.23 kg; P<0.1). There were significant effects of cooking time, loin position, and sex on cooking loss. Cooking losses increased from 32.57 ± 0.64% to 40.80 ± 0.64% as cooking time rose. Loins from anterior end had lower cooking losses (36.66 ± 0.64%) than those from posterior end (38.72 ± 0.64%). It is concluded that extended cooking could increase the meat quality of old cattle. Therefore, extended cooking is recommended to apply in producing meal made from beef of old cattle.

Keywords: extended cooking, meat quality, old cattle

Introduction

The beef available at public markets in Indonesia comes from both young and 'old' animals. The latest one is believed to be relatively tougher than the first one. Research indicates that as beef animals age, their meat contains more heat stable collagen cross-linkages and has lower solubility upon heating than that from young ones (Berry *et al*., 1974), and as a consequence is tougher. It has been shown in a number of countries that, for beef consumers, tenderness is the most important of the quality characteristics (Huffmann *et al*., 1996). At the consumer level, cooking methods have a great effect on the tenderness of cooked beef. Consumers in Indonesia traditionally cook meat at a low temperature for a long time in order to attain a tender product. This is done on the assumption that the meat available to them comes from old animals, and is assumed to be tough. Traditional practice is for slow cooking, which consumers believe will cause the meat to become more tender.

Laakkonen *et al*. (1970) have confirmed that low temperature cooking for a long time can be beneficial to meat tenderness. In that method, collagen is degraded into the soft gelatine form, and muscle fibre hardening is prevented, resulting in more tender meat (Cross *et al*., 1986). In this paper, the term of 'slow cooking' and/or 'extended cooking' will be used interchangeably to refer to a low temperature cooking for a long period.

Materials and Methods

Nine striploins from steer carcasses (six dentition) and nine striploins from cow (full mouth) were used in this experiment. All cattle were finished on grass. Striploins were vacuum packed and aged at 1°C for 14days before subsequently frozen. Each frozen striploin was thawed by holding it for 48h at 4°C and all connective tissue and subcutaneous fat was removed. Samples for cooking were cut to approximately equal size (7.5±0.5cm by 8±0.5cm with 3.25±0.25cm height) and equal weight (230±1g). The four cooking samples were randomised into four different cooking times (2.5, 5, 10 and 15h of cooking), which were referred to as treatments 1, 2, 3 and 4 respectively.

Each cooking sample was weighed, and then vacuum-packed in a Cryovac bag. A K-type thermocouple was then inserted into the geometric centre of meat. An oven (Turbofan oven, type E31W) was set to 80°C (to reach an internal meat temperature of 70±1°C) and allowed to preheat for 30 minutes. Each sample was placed in the centre of an aluminium-baking tray, then put in the centre of the oven and cooked for the selected cooking time. When a sample being cooked, the other three cooking samples from each loin were stored at 1°C in a chiller until required.

After cooking, each sample was cooled under cold running water for 15 minutes. Each sample was then washed with tap water to remove all exterior fragments and was dried with paper towel before being weighed. Each cooked sample was weighed and then placed in a polyethylene bag and stored overnight in a 1°C chiller. The cooking loss was determined by comparing pre-cook weight with post-cook weight.

A flat-vertical surface of sub-samples for WB shear force was taken from cooked meat samples which have dimensions of approximately (6.6 x 15 x 60) mm. The shear force values were determined using a Lloyd LRX instrument (Lloyd Instrument Ltd., Hampshire, England). Six replicate WB shear force values were recorded for each samples.

The effects of cooking time, loin position and sex on the WB shear force values and cooking loss were tested using the mixed procedure of SAS (SAS, 1996) which fits a mixed linear model. Cooking time, loin position, and sex were treated as fixed effects, whilst striploins nested within sex was classed as random effects.

Results and Discussion

The effect of cooking time on wb shear force values (tenderness)

The WB shear force of the meat samples decreased highly significant ($P<0.001$) from 5.27 ± 0.19 to 3.53 ± 0.19kg as there was an increased in cooking time (Table 1 and Table 2), indicating increasing tenderness.

Those findings support the studies of Bouton *et al*. (1981) and Cross *et al*. (1986), who found that slow cooking avoided muscle fibre hardening, and hence enhanced meat tenderness. The WB shear force value is influenced by the number of myofibrils per cross-sectional area (Bouton and Harris, 1972). Therefore, it is possibly that the low values of WB shear force at 15h cooking period might be due to the different properties of myofibrils present, as caused by slow cooking procedure.

Table 1. Least squares means (± standard errors) of WB shear force and cooking loss as influenced by cooking time, loin position and sex in roasted beef striploins from steers and cows

	WB shear force (kg)	Cooking loss (%)
Cooking time		
2.5 hours	5.27±0.19	32.6±0.64
5 hours	4.73±0.19	37.2±0.64
10 hours	4.08±0.19	39.6±0.64
15 hours	3.53±0.19	40.8±0.64
Loin position*		
1	4.20±0.19	36.7±0.64
2	4.36±0.19	37.3±0.64
3	4.35±0.19	37.5±0.64
4	4.70±0.19	38.7±0.64
Sex		
Steer	4.10±0.2	36.1±0.78
Cows	4.71±0.23	38.9±0.86

*1 = anterior to 4 = posterior end of the loin

Table 2. F-values of the effects of cooking time, loin position, sex and interaction between position and cooking time on the WB shear force and cooking losses of steer and cow beef

	Cooking time	Loin position	Sex	Cooking time x loin position
WB shear force	32.55***	2.60	3.83	n.i
Cooking loss	27.73***	2.27	3.2	n.i

*** = P<0.001 * = P<0.05
** = P<0.01 n.i = no interaction found

Extended cooking at a low temperature reduced the variability in the cooked product as represented by a declining value of the SD of the WB shear (Figure 1) from 0.92 ± 0.08 to 0.48 ± 0.07, as the cooking time increased.

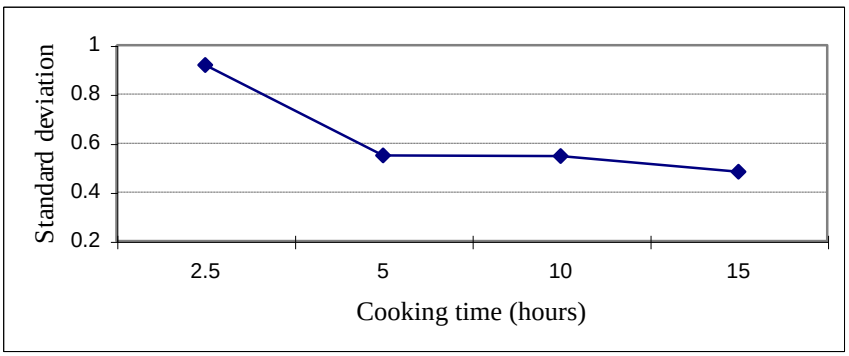

Figure 1. The cooking time effect on the SD of WB shear force value

The most dramatic reduction in the variability of cooked meat occurred between the 2.5h and 5h cooking times, and then variability remained relatively constant up until 15h of

cooking. It seems that in the first period (2.5h of cooking) not all myofibrills had completely undergone protein denaturation, whereas at 5h, it is likely that muscle protein denaturation was complete, and hence no further reduction in variability was found in the cooked product up until 15h.

The effect of loin position on the WB shear force value

Loin position did not significantly affect the WB shear force value (P>0.05). The shear value at anterior end was 4.20± 0 . 19kg, whilst that at posterior end was 4.70 ± 0.19kg (Table 1). However, there was a tendency for the WB shear force value to increase, and hence for the meat to be tougher, as the sample was progressively taken closer to the posterior end (Figure 2). The reason for this is unclear. This finding possibly could be due to a lower myofibrillar density and a low degree of myofibrillar strength at the anterior end rather than at the posterior end as the WB shear force value corresponds to the number of myofibrils per cross-sectional area (Bouton and Harris, 1972a) and its strength (Bouton and Harris, 1972b).

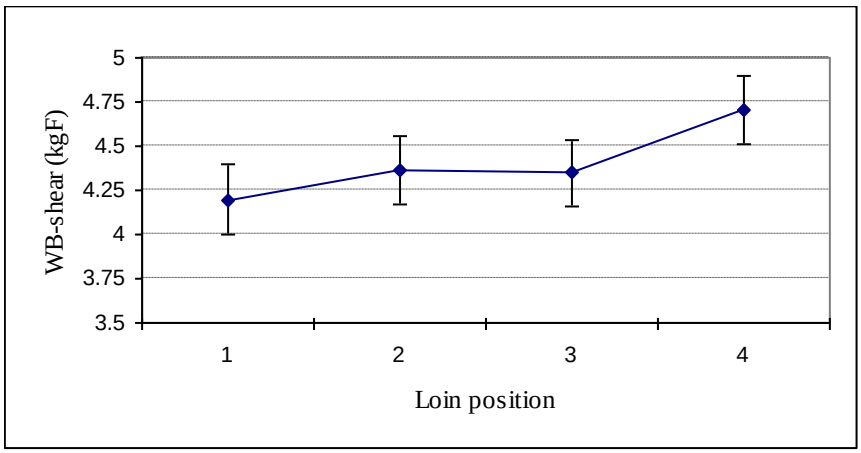

Figure 2. The WB shear value from different loin position (1=anterior to 4=posterior)

The effect of different sex on the WB shear force

The WB shear force value of meat samples from steer (4.10 ± 0 . 21kg) was lower than that from cow (4.71 ± 0 . 23kg); however those differences was not significant (P>0.05, Table 1.). The difference of WB shear force between steers and cows at different cooking time is presented in Table 3. Values at 5 and 10h cooking time from the steer samples were lower than those from cows. However, those at 2.5 and 15h were not different. Because cows are generally much old than steers when processed, this makes a clear understanding difficult. The effect could be intrinsic or result from fat cover or age. Crouse *et al*. (1983) suggested that sex influences on tenderness might be due to differences in the connective tissue component rather than in myofibrils. Boccard *et al*. (1979) indicated that differences in WB shear force value amongst the sexes could be due to the differences in the chilling rate within their carcasses as they have different fat thickness.

Table 3. The mean values ± standard error of WB shear force values of meat samples from steers and cows at different cooking times

Cooking times (hours)	WB shear force value (kg)	
	Steer	Cow
2.5	5.39±0.23	5.53±0.10
5	4.55±0.14	5.23±0.15
10	3.91±0.10	4.43±0.06
15	3.52±0.15	3.63±0.08

Meat samples from steers had a lower WB value than that from cows at cooking times of 5 and 10h, indicated a faster declining rate in the WB shear force of meat from steers than that from cows.

Study of Husband and Johnson (1985) indicated that leaner carcasses would have a more rapid chilling rate and hence are more prone to cold shortening. The current experiment involved steers with a mean hot carcass weight of 340.8± 3 2 .3kg and a mean fat depth of 13.2 ± 9 .87mm, whilst those of cows weighed 184.2 ± 3 1.1kg and had 4.5 ± 5 .36mm of fat cover. Therefore, the different in tenderness between steers and cows in this current study was possibly due to the different in their cooling rate as they are different in their fat cover. That reason is supported by the investigation of Lochner *et al*. (1980) who indicated that the more tender meat from fatter animal was not related to the prevention of cold shortening, but by unknown mechanism that supported by the sustaining physiological temperature during the first 2 to 4h post-slaughter.

The effect of cooking time, loin positions and sex on cooking loss

This experiment also revealed that the longer the cooking time, the higher the cooking loss (Table 1). Moreover, the increased in cooking loss with longer cooking time was also highly significant (Table 2). Cooking time and loin position had a highly significant effect on cooking losses (P<0.001), whilst sex only had a significant effect (P<0.05). Loin position had a pronounced effect on the cooking loss (P<0.001, Table 2). Meat samples from anterior end had significantly lower cooking loss (36.7 ± 0 .6%) than that from posterior end (38.7 ± 0 .6%, Table 1, Figure 3.). Cooking loss of meat from steer was significantly (P<0.05) lower than that from cows (36.1 ± 0 .8% and 38.9 ± 0 .9% respectively, Table 1).

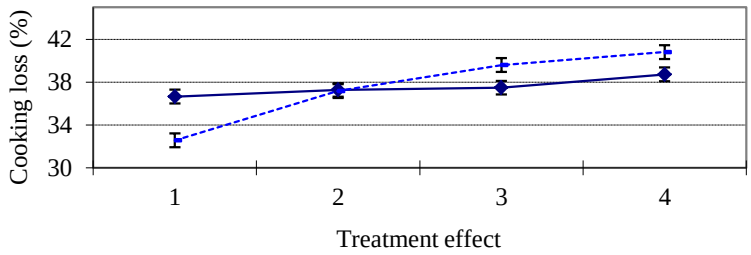

Figure 3. The effect of loin position (___) and cooking time (- -) on the cooking loss (%)

The cooking loss was observed to rise as the cooking time increase (Figure 3). This could be caused by the shrinkage of the myofibrillar and connective tissue structure (Bendall and Restall (1983). Collagen shrinkage played a major role in increasing the cooking loss. Steer

beef had lower cooking losses than that from cows ($P<0.05$). The sex difference in cooking loss is most likely to have resulted from differences in collagen properties due to the different ages of the steers and cows used. Sex differences do not have a significant relationship to the collagen content of the meat (Bailey and Light, 1989). In contrast, age has a great influence on the amount and structure of collagen (Kurth, 1993).

Conclusions

Slow cooking resulted in a large decreased in WB shear force old animals, both steers and cows. The tenderness of meat from steers was lower than that from cows, which could be due to different properties of collagen within those samples. There was a substantial decreased in the variation in WB shear force, suggesting a much more consistent product from slow cooking procedure.

Cooking the loin for 15h resulted in more tender beef; however, the cooking loss at that period was consequently higher.

The differences in tenderness between steers and cows in this experiment was remain unclear, whether this is due to the differences in sex *per se* or differences in their age. Further studies are needed to investigate the effect of slow cooking on the tenderness of beef from different sex of similar age and from different age of the same sexes.

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Increasing of Conjugated Linoleic Acid of Dairy Milk with Additional Rice Meal Fermented and Soybean Oil

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Abstract

An experiment has been carried out to produce milk containing conjugated linoleic acid in high concentrations. The study was carried out experiments in vivo, using a Latin Square Design 6x6x6, with factorial 2x3 treatment. The first factor (A) was a type of diet, consisting of feed used for cows in group of dairy farmers Margo Mulyo, Kemutug village, district Baturaden, Banyumas (a1), and concentrate containing 30% rice bran + 20% tapioca by product which was fermented using Saccharomyces cerevisiae, +3% soybean oil (a2). As a second factor (B) were the time of milking, that are before grass feeding (b1), 2 hours after grass feeding (b2), and 4 hours after grass feeding (b3), thus there were 6 treatments. As the column were 6 sampling periods and as the row were 6 cows. The variables measured were fat, and conjugated linoleic acid of milk. The results indicated that concentrate containing 30% rice bran + 20% tapioca by product which was fermented using Saccharomyces cerevisiae + 3% soybean oil increasing 88.89% of conjugated linoleic acid and time of milking 4 hours after grass feeding increasing 30.56% of conjugated linoleic acid compare time of milking that are before grass feeding and increasing 34.29% if compare time of milking 2 hours after gras feeding. Based on the results of this study concluded that the concentrate containing 30% rice bran + 20% tapioca by product which is fermented using Saccharomyces cerevisiae + 3% soybean oil was the best feed formula, and cows were milked four hours after grass feeding was the best milking time.

Keywords: conjugated lenoleic acid, milking interval, soybean oil.

Introduction

The high content of fat and cholesterol in the body, the occurrence of atherosclerosis and cancer are a problem for many people, as a result of increasing incomes and change of diets. These problems can be overcome by consuming foods that contain conjugated linoleic acid (CLA).

CLA is an essential nutrient, present in ruminant products, including milk, as anticarcinogenic (Donovan *et al*., 2000), to prevent mammary gland tumors (Garcia *et al*., 2000) antiatherogenic (Scollan *et al*., 2006), lean body mass promotans and anti-diabetic (Gulati *et al*., 2000), anti-oxidative, cholesterol-depressing and promoting growth, reducing atherosclerosis, and lowering adipose fat (Gillis, 2004), increasing High Density Lipoprotein (HDL) blood plasma (Choi *et al*., 2006). CLA is synthesized either in the rumen as a result of incomplete bio-hydrogenation of linoleic acid (Noci *et al*., 2005), as well as on tissues through the Δ9-desaturation vaccenic acid (VA; 18:1 trans-11) (Corl *et al*., 2001). Based on the benefits that have been described, it's indicating that the CLA is required for human health.

Nutritional strategies to enrich CLA production in ruminant products can be achieved by increasing the supply of C18: 2 (linoleic acid) in the reticulo-rumen. The acid can be obtained from the molds, which are bioactive and contain linoleic acid. Results of the experiment made by Suhartati *et al*., (2005) showed that the pollard fermentation using Saccharomyces cerevisiae was able to increase linoleic acid, but the results have not been significant. Pollard is an energy source that can be replaced by other feedstuffs that is easy to be obtained, i.e. rice bran and tapioca byproduct. To get the linoleic acid in sufficient quantity, we still need to add a compound that is able to supply a source of linoleic acid, i.e. vegetable oils (Flowers *et al*., 2007) or grain oils (Pavan and Duckett, 2007). Soybean oil as a grain oil, is a good source of unsaturated fatty acids (Kahrizi *et al*., 2007), which is able to increase the content of linoleic acid in the substrate, which will be hydrogenated by rumen bacteria and will produce intermediate compounds, i.e. CLA which is needed by humans.

Materials and Methods

The materials for research were six cows, grass, concentrates and soybean oil. This study was conducted with experimental methods in vivo, using a Latin Square 6 x 6 x 6. The treatment is arranged in factorial 2 x 3. The first factor (A) was type of diet, which consisted of the diet used for cows in the Margo Mulyo Group (a1) and concentrates containing 30% rice bran + 20% tapioca byproduct which were fermented using Saccharomyces cerevisiae, plus 3% soybean oil (a2). Nutrient content of grass and concentrates containing 30% rice bran + 20% tapioca byproduct fermented using Saccharomyces cerevisiae are listed in Table 1. As a second factor (B) was milking time after grass feeding, before grass feeding (b1), 2 hours after grass feeding (b2) and 4 hours (b3) after grass feeding. As the columns were 6 periods of sampling and the line included 6 cows. The variables measured were fat content, which was measured using Lacto Scan and CLA of milk, which was measured using Chromatography Gas. Data obtained was analyzed using analysis of variance, Directly Significant Different test and to find the response an Orthogonal polynomials was performed (Steel and Torrie, 1993)

Table 1. The nutrient content of grass and concentrates

Feedstuff	DM (%)	Based on Dry Matter (DM)				
		Crude Protein	Extract Ether	Crude Fibre	Ash	Nitrogen Free Extract
Grass	12.80	9.27	2.84	33.58	11.96	42.35
Concentrates containing fermented rice bran and tapioca byproduct	70.37	13.88	6.26	12.29	4.37	59.06
Control diet	86.27	10.04	15.83	19.69	8.98	45.46

Description: Analysis at Nutrition and Feed Science Laboratory, Animal Science Faculty of Jenderal Soedirman University (2010)

Results and Discussion

Milk fat

Milk fat content ranged from 2.85% to 4.45% (Table 2). Based on the analysis of variance, diets and the interaction between diet and milking time had no significant effect (P> 0.05)

on milk fat content, while the milking time had significant effect (P <0.01) on milk fat content.

Based on HSD test, the milk fat content before grass feeding was lower than 2 and 4 h after grass feeding. The orthogonal polynomial test showed that milking time had quadrater responses (P <0.01) on the fat content of milk by the equation: Y = 3.04 + 1.04 x - 0.18 x2; coefficient of determination (R2) = 0.57; P (2.89; 4.54) (Figure 1)

Table 2. milk fat and conjugated Linoleic Acids content of dairy milk (%)

Treatment	Milk Fat (%)*	CLA (% Fat)**
a_1b_1	3.23 ± 0.514	0.24 ± 0.02
a_1b_2	4.45 ± 0.504	0.27 ± 0.02
a_1b_3	4.14 ± 0.365	0.32 ± 0.10
a_2b_1	2.85 ± 0.507	0.48 ± 0.05
a_2b_2	4.30 ± 0.711	0.44 ± 0.02
a_2b_3	4.33 ± 0.615	0.62 ± 0.13

Description:* Analysis at Koperasi Pesat Karang Kemiri, Purwokerto (2010)
** Analysis at Kimia Terpadu Laboratory, IPB Bogor (2010)

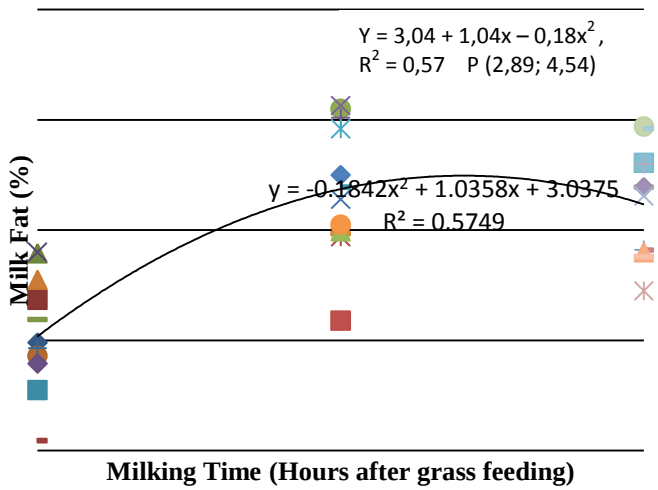

Figure 1. Effect of milking time on milk fat content of dairy milk

Conjugated Linoleic Acids (CLA)

Data from Table 2 showed that Conjugated Linoleic Acids (CLA) ranged from 0.24% to 0.62%. Based on the analysis of variance there was no interaction effect between types of feed and milking time (P> 0.05) but both (the type of feed or milking time) had highly significant effect (P <0.01) on milk CLA content of milk. CLA milk feed treatment (0.51 ± 0.11%) increased by 88.89% when compared with the control feed (0.27 ± 0.07%) (P <0.01). Orthogonal polynomial test showed that milking time gives a linear response (P <0.01) on

the CLA content of dairy milk by the equation Y = 0.32 + 0.026 x; coefficient of determination (r2) = 0.10 (Figure 2).

The highest fat content was achieved about three hours after grass feeding. These results indicated that for milk fat synthesis was required within 3 hours after grass feeding, although between 3 and 4 hours after grass feeding had no significant differences of effect (P>0.05). Fat content of dairy products was influenced by fermentative digestion in the rumen. According to Putra (2006) digestion of feed in fermentation, both dry matter (DM) and organic matter (OM), degraded the higher when the fermentation process took place. Based on the research results of Putra (2006) the highest Volatile Fatty Acid (VFA) production at 3 to 4.5 hours of incubation was achieved. VFA at 3 hours of incubation increased as a result of microbial growth that had begun to increase with a more stable ecological condition, thus feed degrading activity also increased. During the digestion process in the rumen, fermented grass produced volatile fatty acids consisting of acetic acid, propionate and butyrate. Based on research done by Vlaeminck *et al*., (2006), acetic acid is the highest VFA (66%), followed by propionate (21%) and the least is butyrate (13%). The proportion of VFA partially was influenced by the type of feed. Feedstuffs containing high crude fiber will produce acetic acid with a percentage more. From table 1, grass is the feed material with a high content of crude fiber, which is 33.58% (Table 1). Acetic acid is a precursor of milk fat, thus after the cows are fed grass, milk fat content will increase, and the more time available for the synthesis of milk fat content is also growing.

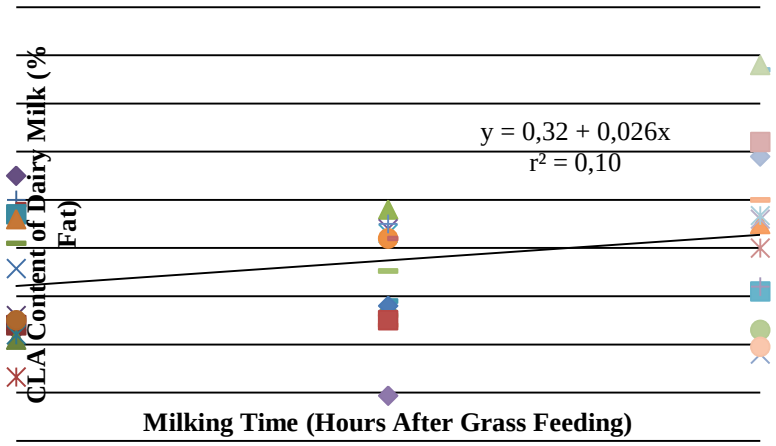

Figure 2. Effect of milking time on CLA content of dairy milk

Dietary treatment that contain fermented rice meal and tapioca byproduct added with 3% soybean oil could increase 88,89% CLA. Soybean oil has a large proportion of C18:2 (51% wt/wt), the precursor of CLA, and has been an effective dietary supplement for increasing the CLA content in Milk (Dhiman *et al*., 2000), (this outlook was supported by results of the analysis made at the Laboratorium of Kimia Terpadu IPB, namely that soybean oil contained 48.16% linoleic acid). Kelly *et al*., (1998) showed that CLA concentration in milk fat could be increased by dietary PUFA supplementation, especially oils rich in linoleic acid. The cis -9, trans-11 CLA isomer is derived directly from C18:2 isomerization during ruminal biohydrogenation, and is the major CLA isomer found in animal derived foods (Huang *et al*., 2008). Besides that, the oil also serves as a defaunating agent, so the population of protozoa in the rumen could be decreased. Protozoa are predators of bacteria so that the

declining population of protozoa may enhance the growth of bacteria (Muhtarudin and Liman, 2006), including group A bacteria that can hydrogenate linoleic acid and α-linolenic acid in the rumen, as the end product is trans-11 C18: 1. Group A bacteria are part of the Butyrivibrio fibrisolvens group, an ill-defined taxon that includes the genera Butyrivibrio and Pseudobutyrivibrio (Kim *et al*., 2008). In the rumen, linoleic acid that had undergone an isomeration become Cis-9, trans-11 CLA, and was then hydrogenated by bacteria group A into the trans-11 C18: 1. The hydrogenation of Cis-9, trans-11 CLA to trans-11 C18: 1 is faster than the hydrogenation of trans-11 C18: 1 to stearic acid (made by the group B bacteria), thus the accumulation of trans-11 C18: 1 in the rumen made it more available to be absorbed into tissues, including the mammary tissue. Lactation cows have the ability to synthesize the endogenous CLA in the mammary tissue, namely de-saturated trans-11 C18: 1 experienced by the Δ9-desaturase to cis-9, trans-11 C18: 2 (CLA) (Bauman *et al*., 1999) which is the main CLA isomer in ruminant fat (75-90% of total CLA) and mainly derived from endogenous synthesis via Δ9-desaturase in the mammary gland (Lock and Bauman, 2004). Increased rumen production of trans-11 C18:1 resulted in increased synthesis of cis-9, trans- 11C18:2 catalyzed by Δ9- de-saturated in the tissues of ruminants (Whitlock *et al*., 2006).

Conclusion

The concentrate containing 30% rice bran + 20% tapioca byproduct which is fermented with *Saccharomyces cerevisiae* + 3% soybean oil was the best feed formula, and milking four hours after grass feeding was the best milking time.

Acknowledgements

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Chitin Oligomer Production with Unique Chitinase *Bacillus* sp Strain SW71 Enzyme from Dams Water Shrimp

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Abstract

A chitinase-producing bacterium, Bacillus sp strain SW71 was isolated from Pinrang shrimp dams water, South Sulawesi, Indonesia. The bacteria was grown in the liquid medium supplemented with colloidal chitin (0.5%) as a carbon source for 46 hrs. at 37oC to produce chitinase. The optimal temperature and pH for the chitinase activity were 60-70oC and 7-8, respectively. The chitinase was very stable at 60-70oC up to 4 hrs. The enzyme of 0.0085 IU/mg chitin on 1 and 0,5 % colloidal chitin substrate were used to produce chitin oligomers through incubation at one until twelve hours. The reaction products were analyzed and fractionated using HPLC. Free cell supernatant and purified enzyme were shown tetramer and pentamer composition after incubation during twelve hours.

Keywords: chitinase, chitine oligomer, olisaccharide

Introduction

Chitin is among the most abundant biomass present on Earth. Chitin, a polymer of β1,4-N-acetylglucosamine (GlcNAc) units is the second most abundant polymer in nature, after cellulose. Chitin is present in insect, crustaceans and in most fungi. Commercial chitin can be produced from crabs and shrimp shells. Chitin and its derivatives are of interest because they have various biological activities, such as those of an immunoadjuvant and a flocculant of waste water sludge, and agrochemical uses (Flach *et al*., 1992). Chitinase plays an important role in the decomposition of chitin and potentially in the utilization of chitin as a renewable resource. Chitin and chitinases (EC 3.2.1.14) have an immense potential. Chitinolytic enzymes have wide-ranging applications such as preparation of pharmaceutically important chitooligosaccharides and N-acetyl D:-glucosamine, preparation of single-cell protein, isolation of protoplasts from fungi and yeast, control of pathogenic fungi, treatment of chitinous waste, and control of malaria transmission (Dahiya *et al*. 2005). Chitin oligosaccharides are prepared by partial hydrolysis of chitin with hydrochloric acid or enzymatically by degradation and transglycosylation. This preteated chitin is then mixed with a chitinolytic enzyme to hydrolyze it to monomer or oligomer of N-acetylglucosamine. Chitin oligosaccharides or chitin oligomers produced by enzymatic hydrolysis of chitin can be used in human health care. Muzarelli (2010) reported that citin and chitosan as immunoadjuvants and non allergenic drug carriers.Chito-oligomers produced by enzymatic hydrolysis of chitin have been of interest in recent years due to their broad applications in medical, agricultural, and industrial applications, including antibacterial, antifungal, hypocholesterolemic, and antihypertensive activity, and as a food quality enhancer. Chitin and chitosan-derivatives have gained wide attention as effective biosorbents due to low cost and high contents of amino and hydroxyl functional groups which show significant adsorption potential for the removal of various aquatic pollutants (Bhatnagar and Sillanpaa (2009).

The commercial interest in the utilization of chitin and it's derivatives has led to the need for inexpensive, reliable sources of active and stable chitinase preparation. Production of inexpensive chitinolytic enzyme is also important in the utilization of shelfish waste to solve environmental problems as well as to promote the economic value of marine product (Wang *et al*., 1985).

Chitinases, a group of enzymes capable of degrading chitin directly to low-molecular-weight product, have been shown to be produced by a number of microorganism. Almost all of the reported chitinase-producing strains will use chitin or colloidal chitin as a carbon source (Wang *et al*., 1995). Some of the chitinases produced by bacterial systems that have gained worldwide research interest for their diverse properties and potential industrial uses.

Material And Methods

Chemicals

Colloidal chitin was prepared from commercial chitin by the method of Arnold and Solomon, (1986). All chemicals were purchased from commercial suppliers.

Microorganism

Bacillus sp strain SW71 was isolated from Pinrang shrimp dams water, South Sulawesi, Indonesia and maintained at 37^{o}C on Sakai medium agar plate containing: 0.7% $(NH_4)_2SO_4$, 0.1% K_2HPO_4, 0.1% NaCl, 0.01% $MgSO_4.7H_2O$, 0.05% yeast extract, 0.1% bacto tryptone, 2% colloidal chitin and 1.5% bacto agar).

Enzyme production

The activities of chitinase and protein concentration in the culture supernatant at different stages of growth of *Bacillus* sp. SW71 were observed to obtain maximal production of the enzyme. *Bacillus sp* SW71 was cultivated in 10 ml of liquid medium in 50 ml erlenmeyer flask containing: 0.7% $(NH_4)_2SO_4$, 0.1% K_2HPO_4, 0.1% NaCl, 0.01% $MgSO_4.7H_2O$, 0.05% yeast extract, 0.1% bacto trypton, 0.5% colloidal chitin, pH 7 for 12-20 hours. Ten milliliters of the seed culture was transferred into 90 ml of the same medium and grown for 72 hours at 55^{o}C at water bath shaker (120 rpm). The culture broth was centrifuged for 15 minutes at 8000 g (4^{o}C) and the supernatant was used for enzyme purification.

Purification of Chitinase

Ammonium sulphate was added to the supernatant at 50% saturation. The solution was left overnight at 4^{o}C. After centrifugation for 20 minute at 9000 g (4^{o}C), the precipitate was collected to obtain crude chitinase. The crude chitinase was then partially purified by precipitation with ammonium sulphate, Partially purified chitinase was loaded into chromatography on Butyl Sepharose column (1 x 14 cm) continued by Sephadeks G 100 column (1 x 14 cm) equilibrated with the 50 mM Tris-Cl buffer. The contaminant and unabsorbed protein was washed from the column with the same buffer, and the enzyme was eluted with a linear gradient of 0-30% Tris-Cl buffer pH 8. The active fractions were pooled and concentrated through freeze dry.

Enzyme assays

Chitinase activities were measured with colloidal chitin as a substrate. The reaction mixture consisting of 150µl of enzyme solution, 150µl of buffer and 300µl of 0.5% (db) colloidal chitin was incubated at 60°C, except its stated. The mixture was then centrifuged immediately at 10000 rpm for 4 minutes. The control was prepared by addition of the enzyme to the supernatant obtained after centrifugation of the mixture of substrate and buffer. The amount of reducing sugar produced was measured by the modified Schales method (Imoto and Yagashita, 1971), with GlcNAc as the standard. One unit of activity was defined as the amount of enzyme which liberated 1µmol of GlcNAc per minute.

Hydrolysis with a crude, Amonium sulphate precipitate dan pure enzyme complex. The samples of low molecular weight chitin were prepared by hydrolysis of 1% w/v solution of colloidal chitin in 0,2 M sodium acetate at 60°C for 1 until 12 hours by adding culture supernatant (FC and FCSH), ammonium sulphate precipitate (AS) or pure enzyme (PE) as 0,0085 IU/mg chitin. After that the hidrolysate was heated for 15 min a boiling water bath and then centrifuged.

Results And Discussion

Characteristics of Chitinase-producing strain

A DNA fragment amplified from the genomic DNA of strain SW71 with a set of primers of 16S rRNA was directly sequenced. The 1063 bp were sequenced. The sequence homology was analyzed by BLAST programs. The phylogenetic tree was performed with TreeCon program. The similarity rank analysis showed that the isolat SW71 was closely related to *Bacillus* species. *Bacillus* sp. B-3 reported by Karita *et al*., (2001) is the nearest neighbor (98 homology).

Crude Chitinase Characteristics

Effect of temperature and pH on activity of purified Chitinase

The highest activity of purified chitinase was detected at 60-65°C (Figure 1a) and at pH of7 of Tris-Cl buffer (Figure 1b).

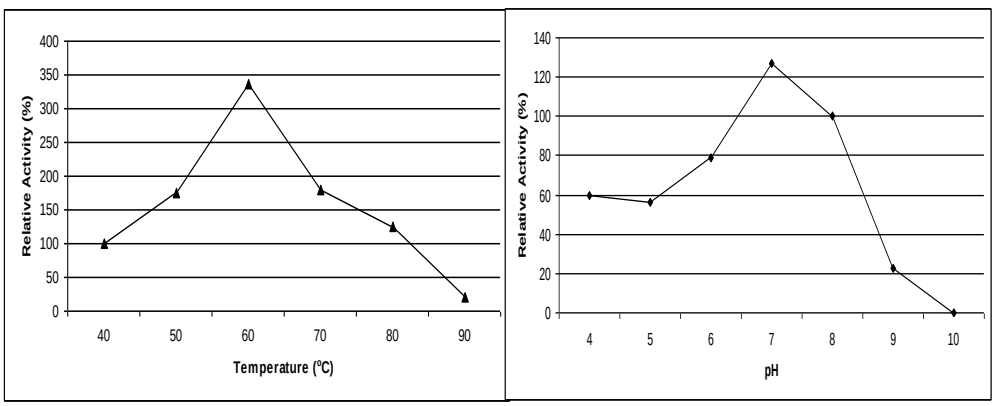

Figure 1. Effect of temperature (a) and pH (b) on activity of purified Chitinase

Effect of temperature on enzyme activity was analyzed by assay the enzyme at various temperatures. The chitinase activities were measured at various pH adjusted by the following buffer: citrate buffer (pH 4-6), phosphate buffer (6-8) and Tris-Cl buffer (8-9), boric acid-borax buffer (pH 9) and glycine-NaOH buffer (pH 10). The optimum temperature of the crude chitinase was 60^{o}C and the enzyme work better alkaline condition. Thermostability of the enzyme was measured by incubation at 60^{o}C, 70^{o}C and 80^{o}C for several hours. The enzyme from *Bacillus* sp. strain SW71 was highly stable at both temperature of 60^{o}C and 70^{o}C.

The effect of various cations addition the crude chitinase activity was also studied (Figure 2). It was carried out by preincubating the enzyme with each addition in 50 mM of phosphate buffer (pH 8) for 5 minute at 55^{o}C and then their activities were measured at 60^{o}C. The crude chitinase activity was not inhibited by addition some of metals cation Ca^{+2},Ni^{+2}, Mg^{+2}, Co^{+2} and Li^{+2}, except Mn^{+2} and Fe^{+2} appearance have a litte effect to decrease enzyme activity. In case of addition some metal ion had no effect to their activity, this is charasteristics for non metaloenzyme.

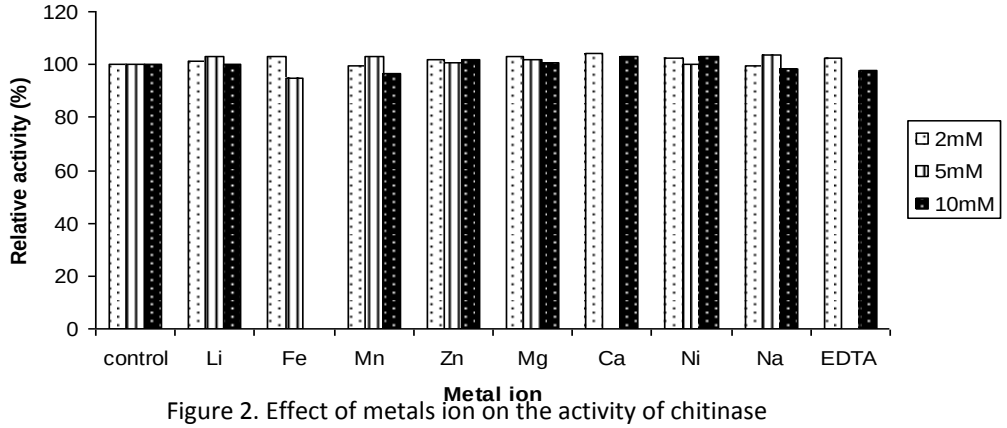

Figure 2. Effect of metals ion on the activity of chitinase

Thermostability of purified enzyme

Thermostability of the enzyme was determined by incubation at 60, 70, and 80^{o}C for several hours and then measured the residual activity (using 50 mM Tris-Cl buffer, pH 8) at the different temperature. The purified chitinase was highly stable at 60 and 70^{o}C (Figure 3).

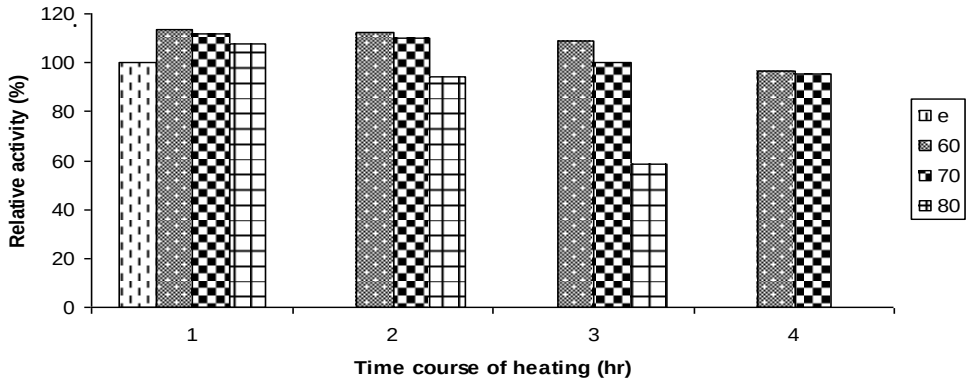

Figure 3. Thermo stability of purified chitinase at various temperature

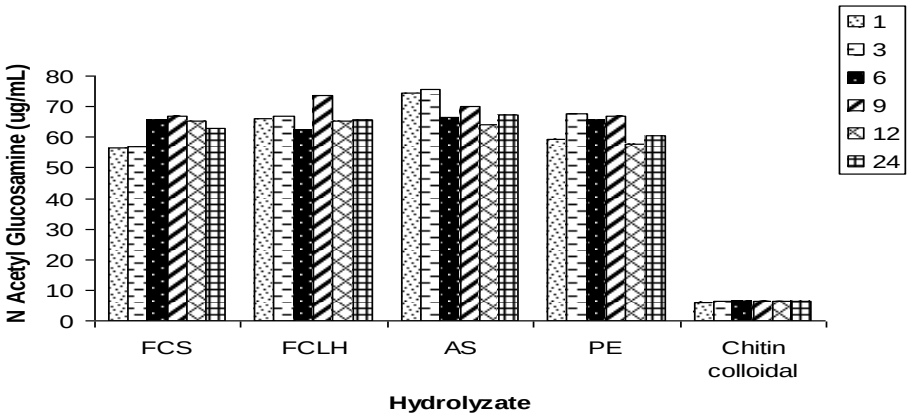

Figure 4. N-Acetyl clucosamine production from chitin oligomer hidrolysate for 1(one) until 24 hours ezymatic reaction (FCH= free cells supernatant, FCLH= heated free cells supernatant on 60^{o}C,AS=Ammonium sulphate precipitate, PE= purified enzyme)

A chitinase purified from culture filtrates of *Bacillus* sp SW71 efficiently catalyzed a hidrolisys reaction on 1% colloidal chitin substrate in a buffer medium containing ammonium phosphate, converting the with 0,0085 IU/mg chitin had yielded pentamer unit for reaction during 12 hours polymer into N-acetyl glucosamine monomer showed AS and FCLH hydrolysate were most efficient to Hydrolysis chitin colloidal substrate than the others enzyme hydrolysate for 1-9 hours. It is appropriate to use a 1% chitin solutions for hydrolysis and this produces maximal yied N-acetyl glucosamine monomer. HPLC analysis used to detection mono-hexamer chitin olygomer composition on 12 hours reaction time (Table 1). That results showed AS and PE hydrolysates has higher pentamer than dimer and monomer N–acetyl glucosamine concentrations. In this case, the addition of time course to the reaction system resulted in a remarkable increase of the pentamer productions, which are desirable as biologically active oligosaccharides.

Table 1. Composition and oligomer Chitin concentration from HPLC analysis

Sample	Reaction time (hr)	Monomer (ppm)	Dimer (ppm)	Trimer (ppm)	Tetramer (ppm)	Pentamer (ppm)	Hexamer (ppm)
AS	12	-	59,2	-	-	197,7	-
FCSH	12	73,8	-	60,5	-	-	-
PE	12	88,4	61,4	-	-	144,7	-

The optimum temperature of crude enzyme was about 60-65^{o}C. It was similar to that reported by other researchers (Ueda and Arai, 1992, Ueda *et al*., 1995). The thermo stability of chitinase from *Bacillus* sp. SW71 was quite remarkable, it is stable at 60-70^{o}C and maintains of its stability even after 5 hours at 60^{o}C in case of crude enzyme. A similar phenomenon was found in case of purified enzyme. This is the first report on bacterial chitinase from shrimp dams that was stable up to for 5 hrs at 70^{o}C.

Nam-soo and Kim (1991) stated that the increased of enzyme thermostability can be explained in two mechanisms, namely intrinsic and extrinsic stabilizing mechanism. In case

of intrinsic stabilizing mechanism, the molecular structure of enzyme itself is of central important for improved thermal stability. Factors contributing to intrinsic mechanism are electrostatics interaction (e.g., formation of salt bridge), hydrogen bond, hydrophobic interaction, content of aliphatic amino acids, intra molecular disulfide linkage, and compact packing. In case of extrinsic stabilizing mechanism, multipoint interaction of enzyme with other cellular component and the presence of specific thermo-stabilizing factors are important for increased thermostability. Factors contributing to extrinsic stabilizing mechanism are protein-protein contact, bindings of substrates and other cellular component of low molecular weight, prosthetic groups, metal cations etc.

The optimum pH for the chitinase produced by strain *Bacillus* sp SW71 was nearly neutral or slightly alkaline. These were similar to these of chitinase from *Pseudomonas aeruginosa* K-187 (Wang and Chang *et al*., 1997). Other chitinase produced by bacteria belonging to the genus of *Bacillus* have been reported by several researchers. The chitinase from *Bacillus circulans* WL-12 and B*acillus* strain MH-1 work better at acidic conditon (Watanabe *et al*., 1992, Sakai *et al*., 1998). The chitinase from *Aeromonas* Sp No. 10S-24 was also optimum at an acidic condition (Ueda and Arai, 1992). Chitinase from *Beauveria bassiana* was reported to be active at pH 9.2, however, it was not thermostable (Suresh and Chandrasekaran, 1999).

The inhibition pattern of metal ions and organic compound on chitinase from *Bacillus* sp SW71 differ from that of the thermostable chitinase reported by other researchers. Addition of $CaCl_2$ did not have significant effect on the chitinase activity from *Aeromonas* so No. 10S-24 (Ueda and Arai, 1992) or *Pseudomonas aeruginosa* K-187 (Wang and Chang, 1997). However, chitinase from *Bacillus* sp SW71. was not inactivated by Ca^{+2}. Mostly, thermostable enzymes required metal cation such as Ca^{+2} as specific stabilizing factors. For examples, a thermostable protease (thermolysin) is stable when Ca^{+2} ion is present (Nam-Soo and Kim, 1991).

To choose which one enzyme hydrolysate can produce high oligomer, We decide to choosed hydrolysate could produced lower N- Acetyl glucosamine more than 9 hours. So to obtain the chitin olygomer composition, HPLC analysis used to detection mono-hexamer chitin olygomer composition on 12 hours reaction time (Table 1). That results showed AS and PE hydrolysates has higher pentamer than dimer and monomer N–acetyl glucosamine concentrations. In this case, the addition of time course to the reaction system resulted in a remarkable increase of the pentamer productions, which are desirable as biologically active oligosaccharides.

Molecular weight of chitin hidrolysis product very important, because of molecular weight product is dependent to functional characterystic and the best bioactive is oligomer composition (Curroto and Aros 1993, Suzuki 1996, Kolodziejska *et al.* 2000). Ilyina *et al*. (2000) reported chitinase from *Streptomyces cursanovii* which yielded oligosaccharides with degrees of polymerization (DP) ranging from 2 to 9. Due to hydrolitic N-deacetylation, conventional partial acid hydrolysis of chitin gives only low yields of the desired pentamers, hexamers and heptamers (Roby *et al*.(1987)). Murao *et al.* (1992) reported a novel chitinase from *Vibrio alginolyticus* which yielded chitotriose and chitopentaose from colloidal chitin substrate.Yoon (2005) reported that diacetyl-chitobiose (GlcNAc)2 was produced exclusively using a *Vibrio furnissii* chitinase (chi E, 89 kDa) hydrolysis of chitin treated with dimethylsulfoxide containing lithium chloride. This method represents a novel way for the preparation of colloidal chitin. The preparation of a homogeneous series of chitin/chitosan oligomers with the same distribution of degrees of polymerization (DP) ranging from 2 to

12, but with various average degrees of N-acetylation (DA) from 0 to 90% is described by Trombotto *et al*.(2008). In this case, the relative proportion of each chito-oligomer present in the mixture can be estimated precisely as a function of DA considering oligomers of same DP. A chitinase from *Tricoderma reesei* also showed efficient transglycosylation reaction with the tetramer, giving the hexamer and dimer as the major products (Usui *et al*. (1990). In our research, chitinase from *Bacillus* sp. SW71 had enzyme activity 0,121 IU/mL or 0,0085 IU/mg chitin could react with 1% chitin colloidal for 12 hours, was produced chitin oligosaccharides with degrees of polymerization (DP) ranging from 2 to 5.

Degrees of polymerization (DP) chitooligosaccarides very important to biological activity, for examples chitohexaose and chitoheptaose show anti-tumor activity and are efficient elicitors of chitinase activity in melon plants (Roby *et al*.(1987). Chitooligosaccharides had activity as antibacterial, antitumor, and immunostimulant (Meidina (2005), Semenuk *et al*. (2001), Suzuki (1996)). Yeon (2004) reported that N-acetyl chitohexaose and chitohexaose had growth inhabited to Meth A-solid tumor cells. Hertriyani (2005) report*et al*so that chitin oligomers produced from chitinase enzyme from *Bacillus licheniformis* MB2 could activated *in vitro* lymphocite proliferation. Tanigawa *et al*., (1991) reported that no tocixity from chitin oligomers concentration at 400 µg/mL to L929 macrophage cells line, if compared with lipopolysccaride (LPS) tocixity at 100 µg/mL. Koide (1998) reported that Hexa-N-acetylchitohexaose dan chitohexaose as antiallogenic mouse tumor sarcoma. Wahyuni (2006) reported that chitosan oligomers produced from chitosanase enzyme from *Bacillus licheniformis* MB2 could activated *in vitro* lymphocite cells proliferation and inhibited some cancer cells proliferation better than chitosan polymer.

Bacause of the fact that chitin oligosaccharides with degrees of polymerization (DP) ranging from 2 to 5 could production from chitinase from *Bacillus* sp. SW71 with 0,0085 IU/mg chitin had yielded pentamer unit for reaction during 12 hours, the possibility of the preparation by varying condition of hydrolysis is needed of this approach. The result obtained show a clear ooportunity of using the chitinolytic enzyme from *Bacillus* sp. SW71 for the preparation of chitin oligosaccharides.

Conclusions

Bacillus sp. SW71 isolated from Pinrang shrimp dams water, South Sulawesi, Indonesia, produced thermostable chitinase.The optimum temperature and pH for the enzyme were 60 -79^{o}C and 7 -8, respectively. The enzyme was also stable at 60-70^{o}C up to 4 hrs. The crude chitinase activity was not inhibited by addition of 0,5-10 mM some of metals cation Ca^{+2},Ni^{+2}, Mg^{+2}, Co^{+2} and Li^{+2}, except Mn^{+2} and Fe^{+2} appearance have a litte effect to decrease enzyme activity.Chitin oligosaccharides with degrees of polymerization (DP) ranging from 2 to 5 was production by chitinase from *Bacillus* sp. SW71 with 0,0085 IU/mg chitin had yielded pentamer unit for reaction during 12 hours.

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Amiza Mat Amin	Department of Food Sciences, Faculty of Agrotechnology and Food Sciences, University Malaysia Terengganu, 21030 Kuala Terengganu, Terengganu	Optimization of Fish Gelatin Extraction from Cobia (*Rachycentron Canadum*) Skin

THE BEST OF POSTER PRESENTERS

Author	Institution	Title
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Sumadi	Laboratory of Seed Technology, Department of Agronomy , Faculty of Agriculture. Universitas Padjadjaran – Bandung, Indonesia	Growth and Yield of Two Soybean Cultivar Seed After three months Storage Periods
Roshita Ibrahim	Department of Food Science, Faculty of Agrotechnology and Food Science, Universiti Malaysia Terengganu	Microbiological Quality of Food Contact Surfaces at Selected 'Satar' Premises in Kuala Terengganu, Malaysia

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