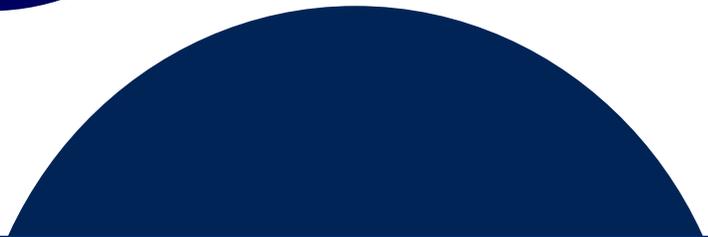
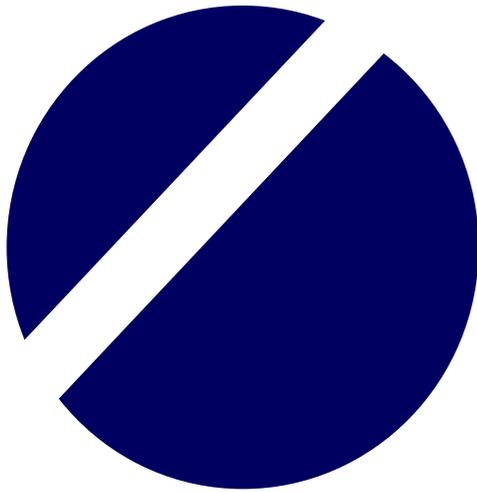


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F₂ SCREENING FOR RARE RESISTANCE ALLELES OF ASIAN CORN BORER, *Ostrinia furnacalis* Guenee (Lepidoptera: Crambidae) IN THE PHILIPPINES

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ABSTRACT

Ostrinia furnacalis Guenee commonly known as Asian Corn Borer (ACB) is the major pest of corn in the Philippines. This study attempted to determine the frequency of Cry1Ab resistance alleles in ACB that will serve as quantitative baseline data for monitoring ACB resistance in Bt corn in the Philippines. The initial frequency of Cry1Ab resistance alleles in ACB collected from corn farms in Lubao, Pampanga, Philippines was studied using the F₂ screen method from March 2006 to December 2008. A total of 2,606 females of ACB were used to establish 800 isofemale lines. From these isofemale lines, 37,702 larvae from the F₂ screen were susceptible to the toxin, Cry1Ab protein. The mortality rate for all the isofemale lines peaked during the first seven days of exposure to Cry1Ab. Two hundred fifty three (253) isofemale lines or 10% were observed to be partially positive to the toxin wherein four (4) isofemale lines (15, 19, 116, and 121) have survivors and successfully reached adult stage. However, due to the low survival of ACB larvae of each isolines, unsynchronized adult emergence and high percentage of male moth emergence, the re-testing of resistance to the isofemale lines were unsuccessful. Furthermore, an equal number of ACB larvae (37,702) were used for the control. A total of one thousand twenty seven (1,027) or 2.88% mortality was observed. This small percentage mortality from the control can be attributed to mechanical injury from handling. The calculated frequency of resistance alleles (E [pR]) was 3.12×10^{-4} . This shows that the resistance allele is rare.

Key words: Bt corn, Cry1Ab, isofemale, Lubao, refuge

INTRODUCTION

The advent of Bt corn and other genetically modified plants has revolutionized our approach to insect resistance management (IRM) for corn borers as well as in developing integrated pest management (IPM) strategies for corn borers. Resistance is expected to limit the usefulness of Bt hybrids. Hence, a serious effort to maximize and prolong the efficacy of the technology through better understanding of basic pest biology as it relates to Bt corn in the field is imperative.

The Philippines IRM strategy involves a high dose and unstructured refuge while other countries use the former also and structured refuge. The 80:20 structured refuge is an option based on three criteria such as: (1) Bt adoption has reached 80%, (2) contiguous area of 200 hectares planted to

Bt corn, and (3) corn – corn based cropping pattern with no temporal refuge being practiced (Barron *et al.*, 2010). This approach does not answer the quantitative basis of IRM for Bt corn in the country.

To date, 800,000 hectares of total corn production area is planted to Bt corn. The Philippines is now categorized as a biotech mega country (country with >50,000 hectares, or more of biotech crops) (James, 2007; James, 2012; James, 2014). Post-commercialization monitoring of the performance of Bt corn is required to detect the development of resistance to Asian corn borer (ACB), *Ostrinia furnacalis* Guenee, at the earliest possible time. At present, local resistance monitoring efforts are based on qualitative scouting for corn borer survivors in Bt corn. The Asian corn borer can become resistant to the Cry1Ab protein which is the toxic ingredient to the larvae. It is in the best interest of all stakeholders to preserve Bt proteins for the long term benefits they provide (MacIntosh, 2009). Although resistance to Bt corn has not yet been established among target pests, low levels of resistance might be present as in the European corn borer (Chaufaux *et al.*, 2001) and also with the ACB. Therefore, a proactive resistance monitoring program needs to be implemented for ACB to increase the chance of detecting resistance at the earliest possible time which would give stakeholders headway to implement appropriate mitigation measures.

Some of the methods used to monitor insect resistance to Bt corn are discriminating dose laboratory assays, surveys of Bt corn fields and screening against test stocks. All of these have limitations (Gould *et al.*, 1997; Roush and Miller, 1986 and Andow and Alstad, 1998). The two additional techniques include the monitoring of Bt sweet-corn sentinel plots and F₂ screen which allows the expression of recessive alleles. The high dose refuge strategy for IRM will work if we assume that the frequency of resistance to Bt corn is low such that resistant ACB survivors in Bt corn are able to mate with susceptible individuals (alleles) coming from the refuge. It is difficult to implement an adaptive IRM plan unless the frequency of resistance alleles in local ACB population is known (Andow and Alstad, 1998). The resistance allele frequency should be $<1 \times 10^{-3}$ for successful application of the refuge plus high – dose strategy (Roush, 1994).

This study attempted to determine the frequency of Cry1Ab resistance alleles in ACB that will serve as quantitative baseline data for monitoring ACB resistance in Bt corn in the Philippines. The allele frequency estimates involving the F₂ screen test will increase the level of familiarity for this species including more reliable prediction on both its susceptibility to Cry proteins and its response (resistance mechanism) to selection pressure (*i.e.*, Bt corn).

MATERIALS AND METHODS

Sampling

Asian corn borer females for mass rearing were collected through the combined use of light traps and sweep netting from corn farming areas in Lubao, Pampanga from March 2006 – December 2008 (Fig. 1). The sweet corn variety, Sugar 75 SG of Syngenta was preferentially used by the farmers. A total of 2,606 ACB females were used to establish isofemale lines. Collected females were individually held in 30 ml plastic cups immediately upon capture and provided with cotton containing 10% honey aqueous solution for food. Gravid females were reared at the Plant Quarantine Support Laboratory, National Crop Protection Center – Crop Protection Cluster (NCPC-CPC), College of Agriculture, University of the Philippines Los Baños (Fig. 2).



Fig. 1. Field collection of Asian corn borer, *Ostrinia furnacalis* Guenee, adults (from left to right: sampling area in Lubao, Pampanga, collection using insect net and light trapping).



Fig. 2. Rearing of Asian corn borer, *Ostrinia furnacalis* Guenee, larval cages (left) and adult cage (right).

F₂ Screen

The F₂ screening method is used for estimating the resistance allele frequency and identifying and recovering resistance alleles from natural populations. The key to this technique is to preserve genetic varieties among isofemale lines and concentrate all alleles into homozygous F₂ genotypes that can be detected using toxin bioassays. The method of Andow *et al.* (1998, 2000) was used to search for resistance alleles in ACB (Fig. 3). The F₂ screen involves a four-step process, namely: (1) sampling mated adult females from natural populations and establishing isofemale lines, (2) rearing and sib-mating F₁ progeny in each isofemale line, (3) screening F₂ neonates to evaluate susceptibility to Bt toxin, and (4) statistical analysis of the data. By sib-mating the F₁ generation, 1/16 of the F₂ larvae are expected to be homozygous for any resistance alleles that a field-collected female (or her mate) carried. Because each female carries at least four haplotypes (two of her own and two from her mate), each isofemale line enables the characterization of at least four alleles.

The progeny (F₁) from each field collected female were sib-mated within each family line and the larvae were reared on artificial corn borer diet using standard rearing techniques established for *O. nubilalis* (Guthrie *et al.* 1965; Lewis and Lynch, 1969) as described by Siqueira *et al.* (2004), except that a commercialized meridic diet (Southland Products Incorporated, Lake Village, AR) was used for larval rearing. Neonate larvae (F₂) resulting from sib-matings were assayed using 16x8 well bioassay trays with 1ml artificial ACB diet containing a diagnostic concentration (LC₉₉ = 120 ng/cm²) of Cry1Ab protein applied using surface contamination technique. Cry1Ab protein was prepared from recombinant *E. coli* (ECE 53) cells containing the Cry1Ab gene. The ECE 53 cells were obtained

from the Bacillus Genetic Stock Center, Ohio State University, Columbus Ohio. A total of 295 replicates were utilized for the bioassay trays.

The number of larvae screened in artificial diet was recorded. One potential pitfall of the F₂ protocol is the possibility of inbreeding depression which may limit the number of F₂ progeny or may cause some families to die out before the F₂ generation. However, by backcrossing the resistant families to a parental stock, the fitness estimates for the resistant genotypes will be unaffected (Andow and Alstad, 1998).

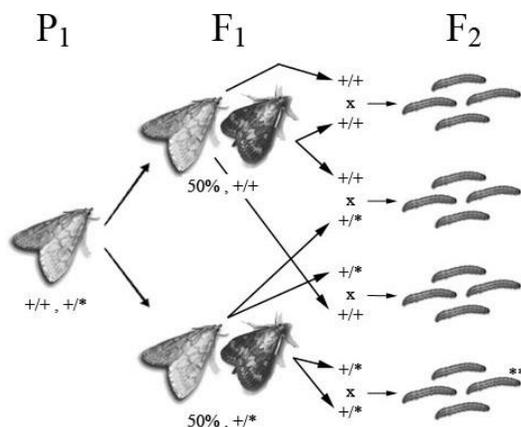


Fig. 3. F₂ screen for a rare resistance allele (*) in Asian corn borer (Adopted from Andow and Alstad, 1998).

Statistical Analysis

Results were analyzed using the modified method described by Andow *et al.* (2000) to derive the allele frequency as additional data becomes available. The expected frequency and confidence intervals of resistance alleles is calculated using generalized Bayesian statistics. Thus, similar to the above authors work, each isofemale line represents 1 independent Bernoulli trial, where a “success”, S , is a true positive from the F₂ screen, which is phenotypically determined as a line that feeds and develops on the diet with the Bt toxin. S is distributed as a binomial (N, P) where N is the number of F₂ lines evaluated and P is the phenotypic probability of success (*i.e.*, an isofemale line expressing resistance). In this case, because there is no prior data available, it was assumed that the prior distribution of P is Beta(u, v), wherein $u = v = 1$. $E[p_R]$ was the expected resistance allele frequency. A similar substitution enables the calculation of the variance.

RESULTS AND DISCUSSION

Resistance Allele Frequency

Eight hundred isofemale lines were produced (Table 1) from 2006-2008 of which 37,702 larvae, from the F₂ screen, were susceptible to Cry1Ab protein. The mortality rate for all of the isofemale lines is at its peak during the first seven days of exposure to the toxin (Fig. 4). Two hundred fifty three (253) isofemale lines or 31.63% were observed to be partially resistant to the toxin and four isofemale lines (15, 19, 116, and 121) had survivors (0.01%) that successfully reached the adult stage. However, due either to some low numbers of surviving larvae in each isolines, unsynchronized adult emergence, or high percentage of male moths that emerged, the re-testing of resistance to the these isofemale lines were unsuccessful. In addition, an equal number of ACB larvae (37,702) were used

for the control. A total of 1,027 or 2.88% mortality was observed. This small percentage mortality in the control can be attributed to mechanical injury due to handling.

Table 1. Calculated frequency of resistance allele using the Bayesian statistics (Andow *et al.*, 2000. *J. Econ. Entomol.* 93(1): 28-30.

Parent female lines (P ₁)	Isofemale lines screened (F ₂)	Resistant Alleles (S)	Frequency of Resistance Alleles E[p _R]	Variance of frequency of Resistance Alleles Var[P]
2, 606	800	4	3.12x10 ⁻⁴	3.88x10 ⁻⁷

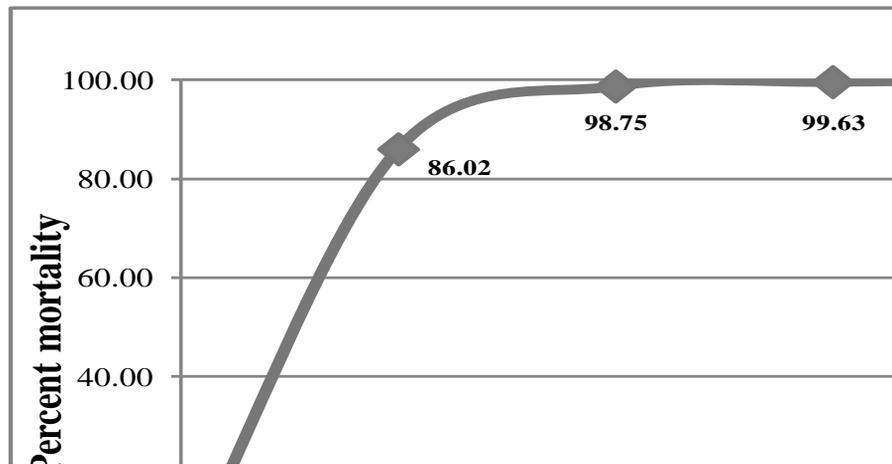


Fig. 4. Mortality rate of all the isofemale lines after exposure to Cry1Ab protein

With $S = 4$ and $N = 800$, the expected frequency of resistance alleles ($E[p_R]$) calculated was 3.12×10^{-4} and indicates that the frequency of Cry1Ab resistance alleles is rare (Fig. 5) and is in agreement with the findings of Andow *et al.* (2000) that this frequency estimate approaches the data needed to support one of the assumptions of the refuge plus high-dose strategy for resistance management in Bt corn.

Next to rice, corn is a major agricultural crop in the Philippines. There were 25 and 28 biotech crop countries in 2009 and 2014, respectively, all over the world including the Philippines (James, 2009; James, 2014). Clearly, the increased adoption of Bt corn in the country will significantly boost corn production. The data gathered from this study will serve as a valuable input in formulating the framework for regulatory decision making by the Bureau of Plant Industry (BPI). Specifically, the F₂ study provides the baseline data on the ACB resistance allele frequency which will serve as reference point for the continuous monitoring of the development of ACB resistance to Bt corn in the Philippines. Overall, the impact on BPI would be the capacity to carry out science-based decision making in IRM. BPI in the near term will be able to develop a proactive IRM and in the long term design an IRM strategy that will be adaptive to the level of ACB resistance in the field.

This is the first time that the F₂ screen test was conducted on the ACB. The results of this study can help in the capacity building of Filipino scientists in relation to the use of the said tests on IRM studies and strategies on Bt corn and other Bt crops that will be commercialized in the country.

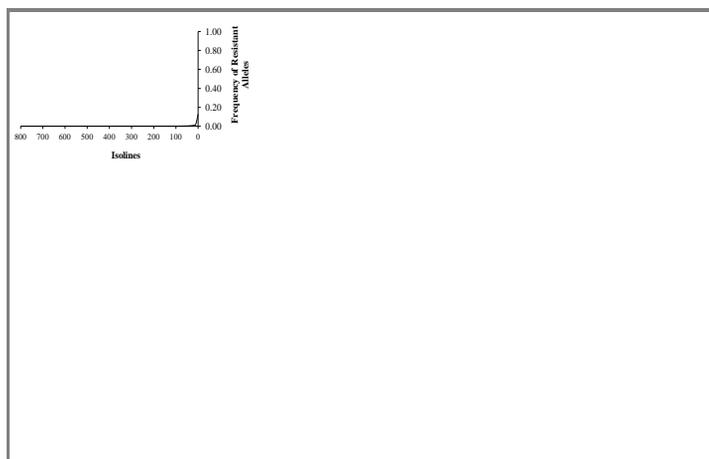


Fig. 5. Cumulative probability of detecting a resistant allele

CONCLUSION

The F₂ screen test was used to search for resistance alleles which will serve as reference point for continuous monitoring of the development of ACB resistance to Bt corn in the Philippines. The frequency of resistance alleles ($E[p_R]$) calculated was 3.12×10^{-4} indicating that the resistance allele is rare and this agrees with the findings of Andow *et al.* (2000).

The Bureau of Plant Industry, Department of Agriculture, Philippines and the technology developers can now use the above result as a baseline value in the quantitative monitoring of ACB resistance alleles where Bt corn is extensively grown in the Philippines during the last five years such as in Cagayan Valley and Central Luzon in Luzon and in South Cotabato and Sultan Kudarat in Mindanao.

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ARE RICE TENANTS PAYING MORE THAN THE TRUE ECONOMIC LAND RENT?

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ABSTRACT

Despite the advantages of owner-operated farms based on equity and efficiency grounds, land rental market plays a big role as a means of providing access of land to poor farmers. However, the existing policy on land rent is still anchored on the provision of a law that was codified in 1963. It is hypothesized in this paper that the basis for setting the land rent in 1963 may no longer be reflective of the true contribution of land in the production process given the advances in rice production technologies. The data used in this analysis was drawn from a survey conducted in the province of Laguna, Philippines for the 2014-2015 cropping cycle. Employing marginal productivity analysis using a Cobb-Douglas production function, it was established that majority of the leasehold tenants in the study sites paid more than the true economic rent of the land. The affordability concept which was the primary basis of codifying, by virtue of R.A. 3844, the rent pegged at 25 percent of the net income based on previous harvests is deemed obsolete. Back in the 1960s, the policy was relevant and appropriate because at the time, rice farming was very simple and the main factors of production were land and labor. In today's rice farming technology, the contribution of land as a factor of production has diminished because of the increasing capital intensity of rice production. The use of modern farm machineries like farm tractors, rice transplanters, reapers, threshers and even combine harvesters has increased the capital intensity of modern rice production resulting in the diminution of the contribution of land in the production process. The use of modern farm inputs like fertilizers and chemicals further pronounced the diminution of the contribution of land to the rice production process. Hence, in order to reflect the realities of modern rice farming technology and to adhere to the basic principle of land rent affordability, the land rental provisions as mandated in R.A. 3844 should be amended.

Key words: affordability, marginal productivity analysis, y, policy analysis, rice production technology, tenancy

INTRODUCTION

In land economic theory, land rent is an important theoretical concept that explains the value of land resources and the opportunity cost of owning them. It represents the economic return that one should pay or should charge for the use of land as a factor of production (Barlowe, 1963). Land rental market has been regarded as an effective mechanism to access land in both developed and developing countries. It incurs lower transaction costs than land sales and must only charge, accordingly, to the value of land in its contribution on the production process (Janvry and Sadoulet, 2001). Leasehold also reduces the chance of being cheated in terms of profit-output sharing (Harsianto, 1994).

Are rice tenants paying more than the true economic land rent?

In the Philippines, the Agricultural Land Reform Code (R.A. 3844, 1963) introduced the system of Agricultural Leasehold. As defined in Section 4 of its provisions, the act declared the abolition of share tenancy in the country and converted all agricultural lands to leasehold operations. As part of the compliance, “the lease must not exceed to the equivalent of 25% of the average value of normal harvest after deducting costs of production during the three agricultural years immediately preceding the date of the establishment of leasehold agreement (Sec. 34 R.A. 3844, 1963).). It is argued here that the 25 percent land rental ceiling was an operationalization of the affordability principle because the prevailing sharing arrangement at the time was 1/3:2/3 or roughly a rental fee of about 33 percent.

The implementation of agricultural leasehold shall cover the following: land areas part of the retention limit, tenanted agricultural lands not yet acquired by CARP for redistribution, and all other tenanted agricultural lands (Administrative Order No.2, 2006). This fundamental provision of the law was also adapted in the more recently enacted Comprehensive Agrarian Reform Law (R.A. 6657,1988). However, given the improvements of rice farming practices such as more inputs and introduction of technology, the contribution of land as a factor of production may have already diminished.

Data and Sources

Laguna Province, Philippines was selected as the area of study. Although some parts of Laguna have switched to industrial economy (Sta. Rosa, Canlubang and other parts in Western Laguna), majority of its citizens are still highly dependent on rice farming as their main source of income. Given its vast tracts of arable land devoted for rice farming, large portion of these lands are still operated by tenant farmers.

A farm survey was conducted with a use of a pretested questionnaire. Specifically data of profile of leaseholders, tenanted rice farm information, farm production information, rented equipment, and inventory of farm assets were gathered. On the other hand, secondary data were used for the list of the registered rice farmer-leaseholders in Laguna under the Comprehensive Agrarian Reform Program of the Department of Agrarian Reform.

The updated list of the registered rice farmer-leaseholders for 2014 under Comprehensive Agrarian Reform Program was provided by the Department of Agrarian Reform (DAR) office in Sta. Cruz, Laguna. From the list, top five tenanted municipalities were chosen namely: Victoria, Pila, Sta. Cruz, Lumban and Mabitac. However, the study disregarded the municipalities in the upland areas of Laguna to avoid differences in farming practices. The study considered 65 samples of varying farm sizes distributed on the chosen areas of study. Tenants were randomly selected per municipality.

Analytical Framework

Conceptually, economic land rent can be determined using two approaches: residual approach and marginal value product estimation. However, the paper only focused on the estimated economic land rent using the marginal productivity approach.

For the paper the *Cobb-Douglas Production Function* or C-D production function was used to determine the efficiency of land as a factor of production by estimating the Marginal Value Productivity of Land (MVP_L) for each tenanted rice farms of the sample.

Important assumptions considered for estimating the C-D production function:

The study used the annual normal production data of 2014 – 2015 cropping cycle. Also, by following Euler’s theorem, in case of constant returns to scale, each unit of factor of production should

be equivalent to its Marginal Value Productivity or MVP. MVP of a factor of production represents the return from the use of incremental unit of the factor (Anthony, 1996).

Mathematically the estimated C-D production function, is of the following form:

$$Y_A = \alpha X_1^{\beta_1} X_2^{\beta_2} X_3^{\beta_3} X_4^{\beta_4} X_5^{\beta_5} X_6^{\beta_6} X_7^{\beta_7} X_8^{\beta_8} X_9^{\beta_9} X_{10}^{\beta_{10}} X_{11}^{\beta_{11}} X_{12}^{\beta_{12}} X_{13}^{\beta_{13}} X_{14}^{\beta_{14}} X_{15}^{\beta_{15}} X_{16}^{\beta_{16}} e^{e_i}$$

Since Cobb-Douglas production function is not linear in parameters, the estimated C-D production function was transformed into linear in the parameters by taking the logarithms of both sides of the equation.

Taking the logarithm of both sides:

$$\text{Log}(Y_A) = \text{Log}(A X_1^{\beta_1} X_2^{\beta_2} X_3^{\beta_3} X_4^{\beta_4} X_5^{\beta_5} X_6^{\beta_6} \dots X_{16}^{\beta_{16}} e^{e_i})$$

Applying the logarithm rule: “The log of a product is the sum of the logs”.

$$\text{Log}(Y_A) = \text{Log}(A) + \text{Log}(X_1^{\beta_1}) + \text{Log}(X_2^{\beta_2}) + \dots + \text{Log}(X_{16}^{\beta_{16}}) + \text{Log}(e^{e_i})$$

Applying the logarithm rule: “the log of a variable raised to an exponent is the exponent times the log of the variable”.

$$\text{Log}(Y_A) = \text{Log}(A) + \beta_1 \text{Log}(X_1) + \beta_2 \text{Log}(X_2) + \dots + \beta_{16} \text{Log}(X_{16}) + e_i \text{Log}(e)$$

Simplifying our specified Double-Log (or Log-Log) model for C-D production function is given by the function:

$$\text{Log}(Y_A) = \text{Log}(\alpha) + \beta_1 \text{Log}(X_1) + \beta_2 \text{Log}(X_2) + \beta_3 \text{Log}(X_3) + \beta_4 \text{Log}(X_4) + \beta_5 \text{Log}(X_5) + \beta_6 \text{Log}(X_6) + \beta_7 \text{Dum}(X_7) + \beta_8 \text{Dum}(X_8) + \beta_9 \text{Log}(X_9) + \beta_{10} \text{Log}(X_{10}) + \beta_{11} \text{Log}(X_{11}) + \beta_{12} \text{Log}(X_{12}) + \beta_{13} \text{Log}(X_{13}) + \beta_{14} \text{Log}(X_{14}) + \beta_{15} \text{Dum}(X_{15}) + \beta_{16} \text{Dum}(X_{16}) + e_i$$

where,

Dependent Variable:

Y_A – Total output of palay, 2014 – 2015 cropping cycle (cavans; 50 kg/cavan)

α – Constant Term

Independent Variables:

X_1 – Land Area (ha)

X_2 – Total Labor: hired man-animal labor (mandays; 1 manday = 8 hours)

X_3 – Management or Entrepreneurship (mandays; 1 manday = 8 hours)

X_4 – Fertilizer (cavans; 50 kg/cavan)

X_5 – Plant Protection Chemicals: herbicides, pesticides, insecticides (liters)

X_6 – Seeds (cavans; 50 kg/cavan)

Dum X_7 – Dummy Variable for Variety of Palay (1 – Hybrid; 0 – Otherwise)

Dum X_8 – Dummy for Irrigation (1 – Irrigated; 0 – Otherwise)

X_9 – Machinery: Tractor (hrs)

X_{10} – Machinery: Thresher (hrs)

Are rice tenants paying more than the true economic land rent?

- X₁₁ – Fuel (L)
- X₁₂ – Oil (L)
- X₁₃ – Crude Oil (L)
- X₁₄ – Farming Experience (years)
- X₁₅ – Dummy Variable for Educational Attainment: High School
 - 1 – High School Graduate
 - 0 – Otherwise
- X₁₆ – Dummy Variable for Educational Attainment: College
 - 1 – College Graduate
 - 0 – Otherwise
- e_i – error term

Output Elasticities are represented by the parameters $\beta_1, \beta_2, \beta_3 \dots \beta_{16}$ representing the effects of land area, hired man-animal labor, management or entrepreneurship, use of fertilizer, use of plant protection chemicals, use of seeds, variety of palay (dummy), irrigation (dummy), use of tractor, thresher, fuel, oil, crude oil, years of farming experiences and educational attainment. However, the regression analysis provide the value of α in logarithmic form as the intercept. The logarithmic form should be removed first to get the value of α as an intercept.

Marginal Value Productivity of a factor of production represents the return from the use of the incremental unit of factor. For instance the marginal value product of Land (MVP_L) of each farm were calculated by taking the derivative of the estimated production function with respect to land multiplied to the price of the output.

Estimated Cobb-Douglas Production Function:

$$Y_A = \alpha X_1^{\beta_1} X_2^{\beta_2} X_3^{\beta_3} X_4^{\beta_4} X_5^{\beta_5} X_6^{\beta_6} X_7^{\beta_7} X_8^{\beta_8} X_9^{\beta_9} X_{10}^{\beta_{10}} X_{11}^{\beta_{11}} X_{12}^{\beta_{12}} X_{13}^{\beta_{13}} X_{14}^{\beta_{14}} X_{15}^{\beta_{15}} X_{16}^{\beta_{16}} e^{e_i}$$

Deriving the formula for Marginal Productivity of Land or MP_L based on the C-D production function:

$$\text{Magrinal Productivity of Land or } MP_L = \frac{\partial Y}{\partial X_1}$$

$$MP_L = \frac{\partial [Y(X_1, X_2, X_3, X_4, X_5, \dots, X_{10})]}{\partial X_1}$$

$$MP_L = \beta_1 \alpha X_1^{(\beta_1-1)} X_2^{\beta_2} X_3^{\beta_3} X_4^{\beta_4} X_5^{\beta_5} X_6^{\beta_6} X_7^{\beta_7} \dots X_{16}^{\beta_{16}}$$

$$MP_L = \frac{X_1 (\beta_1 \alpha X_1^{(\beta_1-1)} X_2^{\beta_2} X_3^{\beta_3} X_4^{\beta_4} \dots X_{16}^{\beta_{16}})}{X_1}$$

$$MP_L = \frac{\beta_1 (\alpha X_1^{\beta_1} X_2^{\beta_2} X_3^{\beta_3} X_4^{\beta_4} \dots X_{16}^{\beta_{16}})}{X_1}$$

$$MP_L = \beta_1 \frac{Y}{X_1}$$

Marginal Value Productivity of Land MVP_L = $\beta_1 \frac{Y}{X_1} P_y$ or MP_L (P_Y)
(price/hectare)

where,

Y – Total output of palay, 2014 – 2015 cropping cycle (cavans; 50kg/cavan)

X_1 – Land area (ha)

β_1 – Output elasticity of land

P_Y – Price of output (average prices of 2014 – 2015 cropping cycle; in pesos)

Hence, the Marginal Value Product of land (MVP_L) is argued here as the economically justifiable land rent for a rice farm based on its efficiency as a factor of production.

RESULTS AND DISCUSSION

Descriptive Analyses

There were 65 respondents representing the registered rice farmer-leaseholders in the province of Laguna. In the municipalities of Victoria, Pila, Mabitac, and Lumban more than half of the registered tenants are male. On the average, the tenants' age were 57 in Victoria, 55 in Pila and Mabitac, 56 in Lumban, and 49 in Sta. Cruz. However, in terms of the age of the total sample, it was observed that the average age of the respondents was 57 years. According to most of the tenants' interviewed, they remained as rice farmers since it is their only source of income. This shows, despite being close to retiring age, farmers are still into farming.

On average, the household is composed of 5 persons. Based on the interview, despite the availability of capturing the benefits of family labor, most of the leaseholders are highly dependent to hired labor since most of their family members are no longer into farming. Also, the mindset of their children were changed. Majority of them already seek for better opportunities outside the farm. Also, most of their children were already married and have families of their own. It was observed that the average household size of the respondents are: 5 in Victoria and Mabitac, 6 in Pila and Lumban, and 3 in Sta. Cruz.

Tenants interviewed are predominantly high school graduates. The distribution falsifies the usual connotation that farmers ended up being a farmer because they have no formal education. Also, 61 out of 65 households considered rice farming as their major source of income. Results show that rice farming plays an important role to most of the tenants' household in the province.

The annual gross farm income accounted includes both monetary and non-monetary income received by the rice farmer-leaseholder. The specific attributes considered were sales of palay, palay given away for harvesting and threshing, palay paid as rent, and palay used for home consumption. On average, a farmer earns an annual gross income of PhP 221,523.58 given the average farm size of 2 hectares.

The size of tenanted land of the total sample are mostly 1.5 hectares. On average, tenants leased two lands. Some of the tenants who intended to expand their farms do so by leasing another land since most of the available lands for tenancy were small. In terms of farming experience, the farmers predominantly have 13 years experience in the farm. However, in terms of years of tenancy, most of the farmers are only 10 years in the occupancy of the leased land. The tenanted rice farms were predominantly irrigated. About 70 percent of the farms have access to irrigation system and the remaining 30 percent uses water pump or *pogpog*. Notably, the respondents are all registered as tenants under the Comprehensive Agrarian Reform Program of the Department of Agrarian Reform.

Cobb-Douglas Production Function Analysis

In order to determine the efficiency of land as a factor of production, the Cobb-Douglas production function was used to estimate the marginal value productivity of land (mvpl) of each rice farm leaseholders. Results of the double-log regression analysis were presented in Tables 1 – 3.

Table 1 shows the model summary of the Pearson correlation result of the model. Based on the suggested qualitative interpretations of the correlation coefficient, 0.981 indicates that there is a very strong linear relationship between the independent variables used in the model. On the other hand, the adjusted r-square of the model is 0.950 which indicates the measure of adequacy or the goodness-of-fit of the model. It implies that 0.950 or 95 percent of the variation in the total output in rice production is explained by the independent variables in the model. The explanatory variables used were land area, hired man-animal labor, management or entrepreneurship, fertilizers, plant protection chemicals, seeds, variety of palay, irrigation, tractor, thresher, fuel, oil, crude oil, farming experience and educational attainment.

Table 1. The model summary of Pearson Correlation result, total output in cavans as the dependent variable, 65 rice farmer-leaseholders in selected municipalities of Laguna, 2014.

Model	R	R-Square	Adjusted R-Square	Std. Error of the Estimate
Pearson Correlation	0.981 ^a	0.963	0.950	0.37452

Predictors: (Constant), X₁ (ln land area), X₂ (ln hired man-animal labor), X₃ (ln management or entrepreneurship), X₄ (ln fertilizer), X₅ (ln plant protection chemicals), X₆ (ln seeds), X₇ (Dummy: variety of palay), X₈ (Dummy: irrigation), X₉ (ln tractor), X₁₀ (ln thresher), X₁₁ (ln fuel), X₁₂ (ln oil), X₁₃ (ln crude oil), X₁₄ (ln farming experience), X₁₅ (Dummy: educational attainment – High School), and X₁₆ (Dummy: educational attainment – College).

Dependent Variable: ln Total Ouput (cavans) – 50 kg/cavan

The analysis of variance is used to test the significance of the model. As shown in Table 2, the p-value of the model is 0.000 which means that the estimated C-D production function is significant at 1%.

Table 2. ANOVA of the regression results of the estimated C-D production function, 65 rice farmer-leaseholders in selected municipalities of Laguna, 2014.

Model	Sum of Squares	df	Mean Square	F	Sig.
Regression	173.409	16	10.838	77.270	.000 ^a
Residual	6.733	48	0.140		
Total	180.142	64			

Predictors: (Constant), X₁ (ln land area), X₂ (ln hired man-animal labor), X₃ (ln management or entrepreneurship), X₄ (ln fertilizer), X₅ (ln plant protection chemicals), X₆ (ln seeds), X₇ (Dummy: variety of palay), X₈ (Dummy: irrigation), X₉ (ln tractor), X₁₀ (ln thresher), X₁₁ (ln fuel), X₁₂ (ln oil), X₁₃ (ln crude oil), X₁₄ (ln farming experience), X₁₅ (Dummy: educational attainment – High School), and X₁₆ (Dummy: educational attainment – College).

Dependent Variable: In Total Ouput (cavans) – 50 kg/cavan

The double-log regression equation based on the results of the model is:

$$\ln Y (\text{Total Output}) = 4.756 + 0.283 \ln X_1 + 0.194 \ln X_2 + 0.209 \ln X_3 + 0.105 \ln X_4 + 0.002 \ln X_5 - 0.116 \ln X_6 - 0.013 \text{Dum } X_7 + 0.143 \text{Dum } X_8 - 0.043 \ln X_9 + 1.014 \ln X_{10} + 0.034 \ln X_{11} - 0.086 \ln X_{12} + 0.059 \ln X_{13} - 0.249 \ln X_{14} - 0.098 \text{Dum } X_{15} - 0.313 \text{Dum } X_{16}$$

where,

$\ln Y_A$ – Total output of palay, 2014 – 2015 cropping cycle (cavans); 50 kg/cavan

α – Constant Term

Independent Variables:

$\ln X_1$ – Land Area (ha)

$\ln X_2$ – Total Labor: hired man-animal labor (mandays; 1 manday = 8 hours)

$\ln X_3$ – Management or Entrepreneurship (mandays; 1 manday = 8 hours)

$\ln X_4$ – Fertilizer (cavans; 50 kg/cavan)

$\ln X_5$ – Plant Protection Chemicals: herbicides, pesticides, insecticides (liters)

$\ln X_6$ – Seeds (cavans; 50 kg/cavan)

$\text{Dum } X_7$ – Dummy Variable for Variety of Palay (1 – Hybrid; 0 – Otherwise)

$\text{Dum } X_8$ – Dummy for Irrigation (1-Irrigated; 0 – Otherwise)

$\ln X_9$ – Machinery: Tractor (hrs)

$\ln X_{10}$ – Machinery: Thresher (hrs)

$\ln X_{11}$ – Fuel (L)

$\ln X_{12}$ – Oil (L)

$\ln X_{13}$ –Gear Oil (L)

$\ln X_{14}$ – Farming Experience (years)

$\text{Dum } X_{15}$ – Dummy Variable for Educational Attainment: High School

(1 – High School Graduate, 0 – Otherwise)

$\text{Dum } X_{16}$ – Dummy Variable for Educational Attainment: College

(1 – College Graduate, 0 – Otherwise)

NOTE:Since $\alpha = e^{4.756}$, this means that $\log(\alpha) = 4.756$.Then, we remove the logarithm by taking the EXP (4.756) = 116.28

The multiplicative form of the estimated C-D production function based on the results of the model is:

$$Y = 116.28 (X_1^{0.283})(X_2^{0.194})(X_3^{0.209})(X_4^{0.105})(X_5^{0.002})(X_6^{-0.116})(X_7^{-0.013})(X_8^{0.143})(X_9^{-0.043})(X_{10}^{1.014})(X_{11}^{0.034})(X_{12}^{-0.086})(X_{13}^{0.059})(X_{14}^{-0.249})(X_{15}^{-0.098})(X_{16}^{-0.313})$$

Table 3 shows the regression results of the estimation of the C-D production function. The estimates showed that out of 16 explanatory variables 6 were considered significant. Assuming all variables in the model are held constant, the sample regression constant indicates that when the independent variables in the model is equal to zero the annual total output of the farm is equal to 116.28 or approximately 116 cavans. The output elasticities of the significant explanatory variables state that (1) For every 1 hectare increase in the farm size of the tenant, there is a corresponding 0.283 cavan increase in the annual total output of palay, *ceteris paribus*, (2) For every 1 manday increase in the employment of hired labor, there is a corresponding 0.194 cavan increase in the annual total output of palay, *ceteris paribus*, (3) For every 1 manday increase in the utilization of unpaid family labor or management, there is a corresponding 0.209 cavan increase in the annual total output of palay, *ceteris paribus*, (4) For every 1 cavan increase in the use of fertilizer, there is a corresponding 0.105 cavan

The summary of computed annual MVP_L (P/ha) and actual annual land rent paid (P/ha) are shown in Tables 4 and 5. The results showed that, on average, a rice farmer should pay only PhP 23,017.93/ha annually for the use of the land. However, comparing it to the actual land rent paid, a tenant is paying an excess of PhP 13,769.23/ha annually in terms of the efficiency contribution of the land as a factor of production on average. It is also shown that predominantly (18 out of 65) or about 27.69 percent of the sample have a MVP_L of PhP14,000.00 – PhP 24,000.00/ha. However, based on the actual land rent paid, the prevailing land rental market pegs the price mostly in the range of PhP 30,000.00 – PhP 41,000.00/ha. The gap between the values of actual land rent paid (P/ha) and computed MVP_L (P/ha) proves that tenants, in reality, are charged of land rents higher than the contribution of land as a factor of production.

Table 4. Summary of the estimated annual marginal value productivity of the each leased rice farm in selected municipalities in Laguna, 65 respondents, 2014.

Marginal value productivity of land (P/ha)	Respondent	
	No.	%
PhP 2000.00 and below	3	4.62
PhP 3,000.00 – PhP 13,000.00	16	24.62
PhP14,000.00 – PhP 24,000.00	18	27.69
PhP 25,000.00 – PhP 35,000.00	15	23.08
PhP 36,000.00 – PhP 46,000.00	9	13.85
PhP 47,000.00 – PhP 57,000.00	4	6.15
Total	65	100
Mean of the Computed MVP_L (P/ha)	PhP 23,017.93	
Mean of the Actual Land Rent Paid (P/ha)	PhP 36,787.16	

Source of data: Survey results, 2014 – 2015 cropping cycle.

Table 5. Distribution of the selected rice farmer-leaseholders by values of the actual annual land rent paid, Laguna province, 65 respondents, 2014.

Actual land rent paid ^a (P/ha)	Respondent	
	No.	%
PhP 6,000 – PhP 17,000	7	10.77
PhP 18,000 – PhP 29,000.00	13	20
PhP 30,000.00 – PhP 41,000.00	26	40
PhP 42,000.00 – PhP 53,000.00	13	20
PhP 54,000.00 – PhP 65,000.00	2	3.08
PhP 66,000.00 – PhP 77,000.00	1	1.54
PhP 78,000.00 – PhP 89,000.00	3	4.62
TOTAL	65	100
MEAN ACTUAL LAND RENT PAID (P/ha)	PhP 36,787.16	

^a Based on the actual annual land rent paid.

Source of data: Survey results, 2014 – 2015 cropping cycle.

Are rice tenants paying more than the true economic land rent?

As shown in Table 6, on average, results revealed that 50 out of 65 respondents (76.92 percent) were paying above the average computed marginal value productivity of land (P/ha). This is consistent to the information gathered from the tenants' assessment of the implementation of the agricultural leasehold. Majority of the tenants interviewed (63.08 percent) considered their pegged land rent as high based on their earnings from the rice farm.

Table 6. Comparison of the mean marginal value productivity of land (P/ha) and actual land rent paid (P/ha) of the rice farmer-leaseholders in selected municipalities in Laguna, 65 respondents, 2014.

Category	Respondent	
	No.	%
Actual Land Rent < MVP _L	15	23.08
Actual Land Rent > MVP _L	50	76.92
Total	65	100

Source of data: Survey results, 2014 – 2015 cropping cycle.

Actual Land Rent Paid (P/ha) above the Efficiency of the Leased Land (MVP_L)

As shown in Table 7, on average, the respondents paid 32.82 percent above the marginal value productivity of the land. However, in terms of the frequencies of the percentages paid, predominantly, tenants paid 25 – 34 percent above the average marginal value productivity of land. The results show, that in terms of the efficiency of land, majority of the pegged rent (50 out of 65) to the farmers overestimates the contribution of land as a factor of production.

Table 7. Summary of the frequency and percentage distribution of actual land rent paid above the marginal value productivity (P/ha) of the leased rice farm in selected municipalities in Laguna, 65 respondents, 2014.

Percentage paid above MVP _L	Respondent	
	No.	%
(%)		
3 – 8	7	14
9 – 16	10	20
17 – 26	6	12
25 – 34	12	24
35 – 42	5	10
43 – 50	3	6
51 – 58	3	6
59 and above	4	8
Total	50	100
Mean Percentage	32.82%	
Mode Percentage	25 – 34 %	
Total Sample	65	

Source of data: Survey results, 2014 – 2015 cropping cycle.

Marginal Value Productivity vs Actual Land Rent Paid

Based on the comparison showed in Table 8, on average, tenants are paying more than the and efficiency of land as a factor of production. On average a tenant is paying an excess of PhP 13,769.23/ha for MVP_L.

Table 8. Comparison of the mean marginal value productivity of land (P/ha) to the actual land rent paid of the 65 rice farmer-leaseholders in selected municipalities of Laguna, 2014.

Mean	Marginal Value Productivity (P/ha)	Actual Land Rent Paid (PhP)	Difference (PhP)
Value	23,017.93	36,787.16	-13,769.23

Source of data: Survey results, 2014.

SUMMARY AND POLICY IMPLICATIONS

Results showed that majority of the leasehold tenants in the study sites paid more than the true economic rent of the land. The affordability concept which was the primary basis of codifying, by virtue of R.A. 3844, the rent pegged at 25 percent of the net income based on previous harvests is deemed obsolete. Back in the 1960s, the policy was still relevant and appropriate because at the time, rice farming was very simple and main factors of production were land and labor. In today's rice farming technology, the contribution of land as a factor of production has diminished because of the increasing capital intensity of rice production. The use of modern farm machineries like farm tractors, rice transplanters, reapers, threshers and even combine harvesters has increased the capital intensity of modern rice production resulting in the diminution of the contribution of land in the production process. The use of modern farm inputs like fertilizers and chemical inputs further pronounced the diminution of the contribution of land to the rice production process. Hence, in order to reflect the realities of modern rice farming technology and to adhere to the basic principle of land rent affordability, the land rental provisions as mandated in R.A. 3844 should be amended.

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PHOSPHORUS FRACTIONS OF PADDY SOILS IN JAVA, INDONESIA

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ABSTRACT

Paddy fields in Java contributes 52% of Indonesia's rice production. The use of fertilizers has been increasing significantly since intensification programs were started in 1969 for paddy fields in Java. Reports concerning phosphorus (P) fractions of paddy soils in Java is not available at present. Therefore, soils P fractions were investigated in Java paddy soils. Top soils from 23 locations were collected from the surface layer (0-20 cm) in February 2012. They consisted of 7 locations from West Java, 11 locations from Central Java and 5 locations from East java. Selected physico-chemical properties and P fractions were analysed in Laboratory of Graduate School of Life and Environmental Sciences, Kyoto Prefectural University Japan and Laboratory of Soil Chemistry and Fertility, Department of Soil Science and Land Resource, Faculty of Agriculture, Bogor Agricultural University respectively. The results showed that physico-chemical properties in West Java, Central Java and East Java were different. The mean values of Resin P (readily available to plant) in East Java was higher than that of Central Java and West java. The mean values of $\text{NaHCO}_3\text{-P}_i$, -P_o and NaOH-P_o were comparable in West Java, Central Java and East Java. However soil P accumulation in West Java, Central Java and East Java were different. Soil P in West Java was accumulated mostly in NaOH-P_i and residual-P. Soil P in Central Java was accumulated mostly in NaOH-P_i , HCl-P_i and residual-P. Soil P in East Java were accumulated mostly in HCl-P_i and residual-P. Soil pH was negatively correlated with the NaOH-P_i . The values of Fe extracted by ammonium oxalate (Fe_o), Al and Fe extracted by dithionite-citrate-bicarbonate (Al_d and Fe_d), $\text{Al}_o+1/2 \text{Fe}_o$ can be used to predict the values of $\text{NaHCO}_3\text{-P}_i$, -P_o , NaOH-P_i , -P_o fractions in Java paddy soils. Resin- P_i was significantly correlated with $\text{NaHCO}_3\text{-P}_i$ and NaOH-P_i suggesting that $\text{NaHCO}_3\text{-P}_i$ and NaOH-P_i can be used to predict the value of resin- P_i . The different accumulation of P in West, Central and East Java needs attention in managing P fertilization.

Key words: accumulation, correlation, fertilization, management

INTRODUCTION

Java is a very important island as it serves as the rice bowl of Indonesia. It has about 2.12 million hectares of paddy fields, contributing 52% of national rice production. The use of fertilizers has been significantly increasing since intensification programs were started in 1969 (Adiningsih *et al.*, 1989). The main fertilizers are nitrogen (N), phosphorus (P) and potassium (K) fertilizer. The average production of rice in Java is 5.60 ton ha⁻¹ in one harvest. At present the production of rice in Java has

leveled, as there is no significant increase in production with the increase of fertilization. This may be due to the nutrient imbalance. The status of nutrients should be monitored to have an idea concerning the fertilization management in Java paddy fields. The balance of nutrient status in soil cannot be over emphasized.

The researches concerning nutrient balance in paddy soils have been conducted in some Asian countries (Nakamura and Matoh, 1996; Cho and Han, 2002; Setyorini *et al.*, 2004). These stressed on input and output of nutrients in the field. The studies of nutrient accumulation were conducted by Park *et al.* (2004) and Li *et al.* (2015). They studied about soil P status and accumulation in paddy soils in Korea and China respectively. Sofyan *et al.* (2004) reported that P status of paddy soils in Java were high, indicating that P was already accumulated in paddy soils. Reports concerning P fractions in Java paddy soils is not available at present. The accumulation of P fraction is presumed to be different in West Java, Central Java and East Java due to different climate type based on the rainfall which is different from west to east. According to the Köppen climate classification, climates of the western, central, and eastern part of Java are classified as tropical rain forest climate (Af), tropical monsoon climate (Am) and tropical savannah climate (Aw), respectively. This phenomenon results in a dryer eastern part compared to the western part. The climate type affects some soil properties and may influence P distribution of the soil P of paddy soil in Java. For sustainable and productive agriculture, different accumulation of P requires different management.

This research sought to identify the P fractions in paddy soils in Java and to elucidate the relationship between selected physico-chemical properties with the P fractions.

MATERIALS AND METHODS

Soil Sample

Top soils from 23 locations were collected from the surface layer (0-20 cm) in 7 locations from West Java, 11 locations from Central Java and 5 locations from East Java (Figure 1). Sampling sites ranged from 06°16'25.0" to 08°06'27.2" in the south latitude and from 107°17'08.7" to 112°32'15.0" in the east longitude.

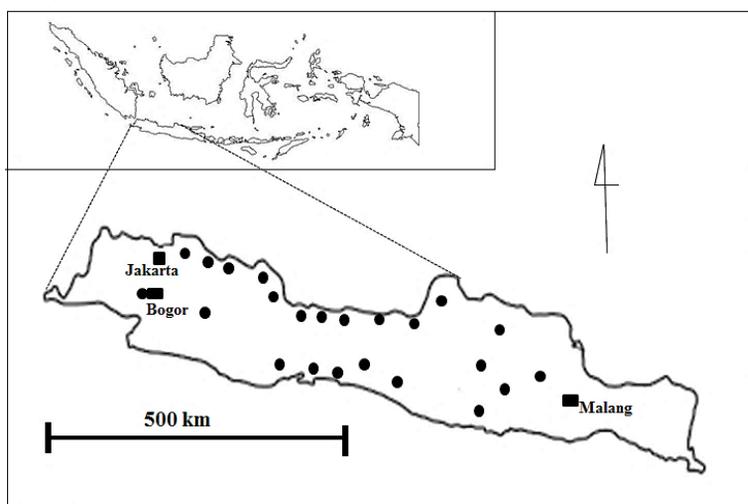


Fig. 1. Location of the sampling sites in Java paddy field

Analytical Methods

Evaluation of selected physico-chemical properties of the soils

To investigate the relationships with the soil P fractions, selected physico-chemical properties of the soils were analyzed with methods as described by Hartono *et al.* (2005). The analyses were conducted in Laboratory of Graduate School of Life and Environmental Sciences, Kyoto Prefectural University Japan. The content of total organic carbon (C) and total N in soil were measured with a NC analyzer (Sumigraph NC analyzer NC-800-13 N, Sumika Chem. Anal. Service). Cation exchange capacity (CEC) was obtained by extraction with 1 mol L⁻¹ NH₄OAc pH 7.0. The contents of exchangeable calcium (Ca) and magnesium (Mg) were determined by atomic absorption spectrophotometry (AA-640-12, Shimadzu Corporation, Japan) while those of exchangeable potassium (K) and sodium (Na) were determined by flame emission spectrophotometry (AA-640-12, Shimadzu Corporation, Japan). Base saturation was defined as the ratio of total exchangeable bases (Ca, Mg, K and Na) to CEC, expressed as a percentage.

The contents of dithionite-citrate-bicarbonate-extractable iron (Fe) and aluminum (Al) (Fe_d and Al_d) were determined according to the method of Mehra and Jackson (1960). The contents of oxalate extractable-Fe and Al (Fe_o and Al_o) were obtained by extraction with 0.3 mol L⁻¹ ammonium oxalate, at pH 3 for 4 hours in a dark room (McKeague and Day, 1966). Extracted Fe and Al of Fe_d and Al_d and of Al_o and Fe_o were filtered through a syringe filter with a 0.45 µm pore size (Minisart RC 15, Sartorius, Hannover, Germany). Contents of extracted Fe and Al were then determined by inductively coupled plasma-atomic emission spectroscopy (SPS1500, SEIKO). The clay content were determined by the pipette method after organic matter removal by oxidation with hydrogen peroxide and ultrasonic dispersion.

Evaluation of Soil P Fractions

The amounts of soil P in different fractions were determined by Tiessen and Moir (1993) method as follows: (i) resin strip in bicarbonate form (resin-P inorganic (P_i) that is readily available to plant), (ii) 0.5 mol L⁻¹ NaHCO₃ extracting P_i and P organic (P_o) (P which is strongly related to P uptake by plants and microbes, (iii) 0.1 mol L⁻¹ NaOH extracting P_i and P_o (P which is more strongly held by chemisorption to Fe and Al components of soil surface) (iv) 1 mol L⁻¹ HCl extracting P_i (Ca-P of low solubility).

Residual-P was determined by subtracting the sum of resin-P_i, NaHCO₃-P_i, -P_o, NaOH-P_i, -P_o, and HCl-P_i from the total P. Residual-P was interpreted as occluded P and recalcitrant organic forms (Tiessen and Moir, 1993; Dobermann *et al.*, 2002). The P fractionation analyses were conducted in Laboratory of Soil Chemistry and Fertility, Department of Soil Science and Land Resource, Faculty of Agriculture, Bogor Agricultural University.

The protocol of P fractionation is summarized in Fig. 2. P in solution was determined by the procedure of Murphy and Riley (1962).

Duplicate 0.5 g
soil samples

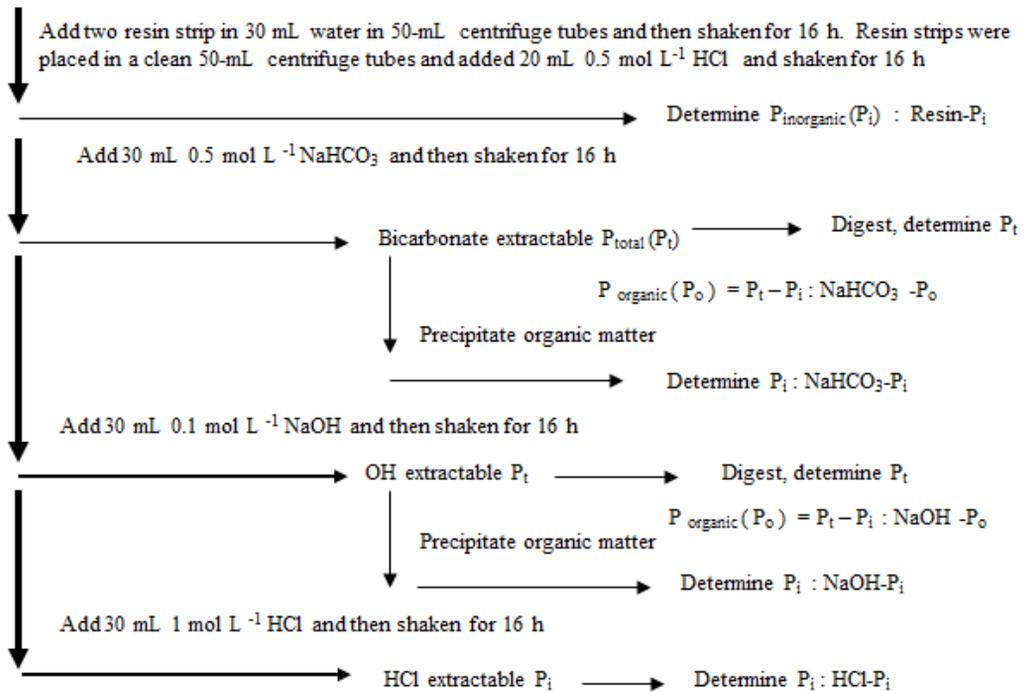


Fig. 2. Flow chart of the P fractionation into various inorganic- and organic-P fractions

RESULTS AND DISCUSSION

Selected physico-chemical properties

Selected physico-chemical properties are presented in Table 1. The soil pH in Java paddy soils were higher compared to those of Java upland soils (Hartono *et al.*, 2005). This is the effect of submergion for long time in paddy soils. The mean soil pH values in West Java, Central Java, and East Java were 6.21, 6.77 and 7.99 respectively. The mean values of pH in West Java, Central Java and East Java were higher than those of found by Darmawan *et al.* (2006b). The status of selected physico-chemical properties was judged according to the criteria published by Soepratohardjo *et al.* (1983). The mean values of soil pH in East Java was higher than that of West Java and and central Java. The mean values of total C in West Java was 19.1 g kg⁻¹ while in Central Java and East Java were 18.4 and 15.5 g kg⁻¹ respectively. All the mean values of total C were low. However the mean values of total C in West Java, Central Java and East Java were higher than those found by Darmawan *et al.* (2006a). The mean values of total N in West Java was 2.14 g kg⁻¹ while in Central Java and East Java were 1.84 and 1.39 g kg⁻¹ respectively. The mean values of total N in West Java was medium while in Central Java and East Java were low. The mean values of total N in West Java, Central Java and East Java were also higher than those found by Darmawan *et al.* (2006a). The mean value of CEC in West Java was 34.3 cmol_c kg⁻¹ while in Central Java and East Java were 32.5 and 46.0 cmol_c kg⁻¹ respectively. The mean values of CEC in West java and Central Java were high but in East Java was very high. The mean values of base saturation in West Java was 73.7% while in Central Java and East Java were 116% and 109% respectively. All the mean values of base saturation were very high.

For the mean values Fe_d and Al_d , the mean values in West Java were higher than those of Central Java and East Java while Fe_o and Al_o were comparable. For the clay content, the mean values of clay in West Java, Central Java and East Java were more than 40%.

The soil P fractions

The soil P fractions were presented in Table 2. The mean values of Resin- P_i (readily available to plant) in East Java was higher than those of Central Java and West Java. The mean values of $NaHCO_3-P_i$, P_o and $NaOH-P_o$ were comparable in West Java, Central Java and East Java. However soil P accumulation in West Java, Central Java and East Java were different. Soil P in West Java was accumulated mostly in $NaOH-P_i$ ($132 \pm 86.0 \text{ mg kg}^{-1}$) and residual-P ($326 \pm 285 \text{ mg kg}^{-1}$). Soil P in Central Java was accumulated mostly in $NaOH-P_i$ ($128 \pm 58.5 \text{ mg kg}^{-1}$), $HCl-P_i$ ($341 \pm 332 \text{ mg kg}^{-1}$) and residual-P ($357 \pm 260 \text{ mg kg}^{-1}$). Soil P in East Java were accumulated mostly in $HCl-P_i$ ($266 \pm 177 \text{ mg kg}^{-1}$) and residual-P ($179 \pm 117 \text{ mg kg}^{-1}$). The high standard deviation of the P fractions data in West Java, Central Java and East Java showed that the variation was high spatially. It indicated that the application of P fertilizer was not the same in the quantity even in the same region.

The total P in Java paddy soils were much more higher than those of Java acid upland soils. The mean value of total P in Java acid upland soils was 487 ± 89.1 (Hartono *et al.* 2006). The amount of resin- P_i and $NaHCO_3-P_i$ fractions in Java paddy soils were much more higher than those of Java acid upland soils (Hartono *et al.* 2006). In Java acid upland soils, the mean values of resin- P_i and $NaHCO_3-P_i$ were $8.00 \pm 2.55 \text{ mg kg}^{-1}$ and $9.80 \pm 9.47 \text{ mg kg}^{-1}$ respectively. As for $NaOH-P_i$ (moderately resistant P), in Java paddy soils was also much more higher than that of Java acid upland soils. In Java acid upland soils the mean values of $NaOH-P_i$ was $63.4 \pm 2.77 \text{ mg kg}^{-1}$ (Hartono *et al.* 2006). The data suggested that P fertilization program in paddy soils was much more intensive compared to that of Java acid upland soils.

Correlation among soil P fractions and physico-chemical properties are presented in Table 3. Significant correlation values shown among Fe extracted by ammonium oxalate (Fe_o), Al and Fe extracted by dithionite-citrate-bicarbonate (Al_d and Fe_d), $Al_o+1/2 Fe_o$, $NaOH-P_i$, $NaHCO_3-P_i$, P_o , total P and residual-P (Table 4). Resin- P_i was not significantly correlated with all physico-chemical properties. $NaHCO_3-P_i$ was correlated significantly with Fe_o and $Al_o+1/2 Fe_o$ while $NaHCO_3-P_o$ was correlated significantly with Fe_d , Al_d , Fe_o and $Al_o+1/2 Fe_o$. $NaOH-P_i$ was negatively correlated with pH. It confirmed that high pH decreased the formation of $NaOH-P_i$ fraction. $NaOH-P_i$ was correlated significantly with Fe_d , Al_d , Fe_o and $Al_o+1/2 Fe_o$. $NaOH-P_o$ was correlated significantly with Fe_d , Al_d , Fe_o and $Al_o+1/2 Fe_o$. Significant negative correlation was shown between $HCl-P_i$ with clay. $HCl-P_i$ also had negative correlation with Fe_d , Al_d , Fe_o and $Al_o+1/2 Fe_o$. It confirmed that $HCl-P_i$ fraction was not influenced by the presence of Al and Fe oxides. The correlation analyses showed that $HCl-P_i$ positively correlated with soil pH although in these results it was not significant. Residual-P correlated significantly with Al_d .

Among soil P fractions, resin- P_i was correlated with $NaHCO_3-P_i$, $NaOH-P_i$, and total-P. It suggested that $NaHCO_3-P_i$ and $NaOH-P_i$ were released when resin- P_i depleted. $NaHCO_3-P_o$ and $NaOH-P_i$ were significantly correlated with residual-P. It suggested that some part of residual-P may transform to moderately resistant P when the latter fraction is depleted.

Phosphorus fractions of paddy soils.....

Table 1. Selected physico-chemical properties of the soil samples

Location	pH (H ₂ O)	Total-Cg kg ⁻¹	Total-N	CEC (cmolc kg ⁻¹)	Base	DCB		Oxalate		Al _o +1/2 Fe _o	Clay	
					saturation (%)	Fe _dg kg ⁻¹	Al _d	Fe _o	Al _og kg ⁻¹	(%)		
West Java												
Karawang	5.44	23.2	2.29	35.1	67.0	15.5	1.06	14.8	1.47	8.86	71.7	
Jatisari	5.49	21.6	2.19	39.9	74.7	15.7	1.01	12.7	1.49	7.86	69.7	
Pamanukan	6.86	27.0	2.52	42.3	86.7	12.1	1.27	9.32	1.63	6.29	80.7	
Indramayu	6.99	17.2	1.97	40.7	87.5	10.5	1.37	7.09	1.81	5.35	65.0	
Palimanan	7.31	8.10	0.78	34.6	87.4	6.25	0.70	4.61	1.11	3.42	52.4	
Cicalengka	5.40	13.1	2.90	24.5	50.9	40.7	5.11	28.5	6.60	20.9	48.6	
Cikarawang	5.99	23.6	2.35	22.8	61.8	54.1	4.83	27.5	8.86	22.6	47.8	
Mean	6.21	19.1	2.14	34.3	73.7	22.1	2.19	14.9	3.28	10.7	62.3	
SD	0.82	6.66	0.66	7.80	14.5	18.0	1.91	9.54	3.12	7.71	12.8	
Central Java												
Brebes	7.66	13.1	1.37	14.6	423	9.65	1.03	8.03	1.74	5.76	54.4	
Suradadi	7.42	16.0	1.73	42.7	102	11.5	1.22	8.12	1.73	5.79	41.1	
Batang	5.41	29.9	2.95	24.4	36.2	20.0	2.47	10.4	2.22	7.42	75.3	
Kendal	6.54	24.0	2.32	47.9	92.7	14.6	1.10	11.5	1.74	7.50	60.9	
Demak	8.28	15.9	1.59	42.5	131	9.33	1.26	6.39	1.75	4.94	56.4	
Jekulo	6.96	14.6	1.42	33.2	64.6	12.7	1.90	10.2	1.74	6.83	49.9	
Borobudur	6.15	14.7	1.51	14.3	76.1	5.49	1.90	11.5	6.17	11.9	13.7	
Kutoarjo	6.81	18.6	1.83	42.2	99.2	19.9	2.78	26.4	7.31	20.5	47.5	
Karanganyar	6.48	19.8	1.91	43.5	103	22.1	2.71	32.1	7.00	23.0	48.0	
Buntu	5.77	27.0	2.65	37.1	67.3	22.4	3.64	40.8	9.90	30.3	48.3	
Yogyakarta	6.99	9.00	1.00	14.8	81.9	5.56	1.54	14.0	3.94	10.9	16.4	
Mean	6.77	18.4	1.84	32.5	116	13.93	1.96	16.3	4.11	12.3	46.5	
SD	0.83	6.29	0.58	13.1	105	6.32	0.84	11.4	2.97	8.51	18.0	
East Java												
Bojonegoro	7.58	18.2	1.59	65.0	96.7	8.55	1.83	6.75	2.32	5.69	68.1	
Tambak Rejo	8.55	10.8	0.86	59.8	120	3.00	0.86	1.86	1.39	2.32	41.0	
Nganjuk	8.08	14.9	1.36	44.8	105	3.65	1.01	1.21	5.37	5.97	62.4	
Jombang	8.06	9.80	0.97	18.7	111	2.45	1.05	2.87	3.32	4.76	16.7	
Ponorogo	7.67	23.8	2.16	41.6	112	5.34	1.98	8.26	6.36	10.5	47.9	
Mean	7.99	15.5	1.39	46.0	109	4.60	1.34	4.19	3.75	5.85	47.2	
SD	0.39	5.72	0.52	18.1	8.79	2.46	0.52	3.13	2.07	2.97	18.1	

The subscripts d and o denote selective extractions with dithionite-citrate-bicarbonate and oxalate, respectively

Table 2. The soil P fractions and total P of the samples

Location	Resin-P ₁	NaHCO ₃ -P ₁	NaHCO ₃ -P ₀	NaOH-P ₁	NaOH-P ₀	HCl-P ₁	Residual-P	Total P
.....mg kg ⁻¹								
West Java								
Karawang	9.18	10.8	17.0	45.1	34.3	35.8	267	420
Jatisari	15.4	65.3	8.92	176	134	97.4	129	625
Pamanukan	11.8	26.8	14.3	89.1	67.8	46.0	169	425
Indramayu	14.1	31.7	9.18	92.1	70.1	58.4	141	416
Palimanan	12.6	20.8	17.1	43.9	33.4	43.5	111	282
Cicalengka	25.5	71.2	22.5	260	104	77.5	656	1217
Cikarawang	23.5	54.2	49.2	217	97.5	94.3	806	1342
Mean	16.0	40.1	19.8	132	77.3	64.7	326	675
SD	6.14	23.4	13.8	86.0	37.0	25.1	285	426
Central Java								
Brebes	24.0	45.4	13.6	106	81.1	180	357	807
Suradadi	13.2	23.0	20.8	61.2	46.6	75.8	260	501
Batang	10.3	20.6	17.7	172	96.4	26.0	614	957
Kendal	75.3	116	26.2	246	30.8	259	486	1239
Demak	8.95	28.7	12.5	51.4	38.0	287	319	745
Jekulo	31.4	55.0	22.2	161	52.8	566	1339	2228
Borobudur	16.3	49.2	11.9	92.1	74.7	1033	80.4	1358
Kutoarjo	23.5	53.9	14.9	96.6	66.2	134	111	500
Karanganyar	26.1	59.2	21.5	104	57.0	145	213	626
Buntu	28.1	85.3	17.5	188	70.4	188	292	869
Jogjakarta	31.6	71.9	10.1	127	43.1	855	156	1294
Mean	26.3	55.3	17.2	128	59.7	341	357	1011
SD	18.2	28.3	5.06	58.5	81.1	332	260	504
East Java								
Bojonegoro	27.3	35.5	6.12	67.5	85.6	138	142	502
Tambak Rejo	11.6	15.6	8.72	28.8	59.7	194	58.8	378
Nganjuk	28.1	30.5	8.44	49.3	74.5	157	142	490
Jombang	16.9	14.8	6.13	32.0	65.9	271	179	585
Ponorogo	52.1	59.3	7.99	66.5	49.7	570	372	1177
Mean	27.2	31.1	7.48	48.8	67.1	266	179	627
SD	15.6	18.2	1.26	18.3	13.8	177	117	317

Table 3. Correlation values among soil P fractions and selected physicochemical properties

	NaHCO ₃ -P _i	NaHCO ₃ -P _o	NaOH-P _i	NaOH-P _o	HCl-P _i	Residual-P	Total-P	pH	CEC	Base saturation	Clay	Fe _d	Al _d	Fe _o	Al _o	Al _o +1/2 Fe _o
Resin-P _i	0.79**	0.17	0.44*	-0.23	0.28	0.26	0.46*	0.01	0.13	0.03	-0.07	0.03	0.10	0.13	0.22	0.17
NaHCO ₃ -P _i		0.31	0.76**	0.07	0.29	0.26	0.54**	-0.37	-0.07	-0.06	-0.12	0.33	0.40	0.55**	0.43*	0.54**
NaHCO ₃ -P _o			0.61**	0.05	-0.20	0.60*	0.41	-0.44	-0.21	-0.19	0.06	0.83**	0.62**	0.51*	0.35	0.48*
NaOH-P _i				0.42*	-0.03	0.58*	0.59**	-0.69**	-0.26	-0.23	0.12	0.72**	0.66**	0.59**	0.34	0.53**
NaOH-P _o					-0.22	0.073	0.03	-0.42	-0.16	-0.02	0.20	0.43*	0.42*	0.21	0.23	0.23
HCl-P _i						0.05	0.68**	0.12	-0.39	-0.02	-0.69**	-0.34	-0.08	-0.10	0.18	-0.01
Residual-P							0.13	-0.28	-0.24	-0.13	0.10	0.49*	0.44*	0.20	0.07	0.16

* $P < 0.05$; ** $P < 0.01$

The results showed that the recommendation for P fertilization management in Java should be different due to the different accumulation of P in West Java, Central Java and East Java. It was reported that application of organic matter and silicate released P in NaOH-P_i and HCl-P_i fractions to resin-P_i (Iyamuremye *et al.*, 1996; Hartono and Bilhaq, 2014). Therefore organic matter and silicate fertilizer can be used to manage P fertilization in Java. However the amount of silicate fertilizer should be more applied in Central Java and East Java due to higher HCl-P_i in both locations.

CONCLUSIONS

The soil pH of paddy soils in East Java were higher than those of West Java and Central Java. The paddy soils in East Java were more alkaline. The CEC and base saturation of West Java, Central Java and East Java were very high with base saturations were close to 100%. However total C status were low and N status ranged from low to medium.

Soil P in paddy soils of Java accumulated in NaOH-P_i, HCl-P_i and residual-P_i. Soil P in West Java was accumulated mostly in NaOH-P_i and residual-P. Soil P in Central Java was accumulated mostly in NaOH-P_i, HCl-P_i and residual-P. Soil P in East Java were accumulated mostly in HCl-P_i and residual-P. The different in soil pH affected the soil P distribution. The values of Fe extracted by ammonium oxalate (Fe_o), Al and Fe extracted by dithionite-citrate-bicarbonate (Al_d and Fe_d), Al_o+1/2 Fe_o can be used to predict the values of NaHCO₃-P_i-P_o, NaOH-P_i-P_o fractions in Java paddy soils where their correlations were significant. Resin-P_i as very available fraction was significantly correlated with NaHCO₃-P_i and NaOH-P_i suggesting that NaHCO₃-P_i and NaOH-P_i can be used to predict the value of resin-P_i.

The very high P status in Java paddy soils and the different accumulation of P in West, Central and East Java needs attention to manage P fertilization. The organic matter and silicate fertilizer can be used to manage P fertilization in Java. More silicate fertilizer should applied in Central Java and East Java due to higher HCl-P_i in both locations.

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THE ROLE OF THE SUBAK SYSTEM AND TOURISM ON LAND USE CHANGES WITHIN THE SABA WATERSHED, NORTHERN BALI, INDONESIA

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ABSTRACT

Bali has recently become a main hub for tourism ensuring national food security as a consequence of the master plan for the acceleration and expansion of Indonesian economic development 2011–2025. Saba watershed, located in the north of Bali, has experienced drastic land use changes due to population growth and tourism development over the past two decades. A large proportion of the paddy fields managed through subak systems in the Saba watershed have been converted to other land uses. Here we explore the role of subak system under land use changes and tourism development. We conducted in-depth interviews using the RRA method to gather primary data from 20 subak areas up-stream, central, and downstream of the watershed. With the exception of Subak Pongjokukli, most subaks within Saba watershed cannot control for drastic land use changes occurring within subak areas. A subak-based agro-tourism development model is possibly the best way to promote sustainable development of both agriculture and tourism within Saba watershed due to its richness in natural resources and as evidenced by legal and financial support provided by the government.

Key words: converted paddy field, subak, sustainable agro-tourism

INTRODUCTION

Primary production (agriculture) and secondary (small-scale industries) and tertiary sectors (including tourism) have been the main reasons for economic growth in Bali, contributing to 18.08%, 15.57%, and 66.35% of total growth, respectively (Regional Development Planning Agency of Bali Province, 2012). Tourism represents an important sector in Bali's economy, which at the same time is used as the barometer to indicate the progress of the overall tourism industry in the whole of Indonesia. Bali is well-known as a popular international tourist destination, appearing often as "the best destination in the world" or "the best spa destination in the world" in international travel magazines, highlighting the awe-inspiring and admirable beauty and uniqueness of Bali to foreign nationals and its position as a tourism destination worldwide. Empirical data from the period 1969–2012 show that 25.23% of foreign tourists visiting Indonesia directly arrive in Bali (Bali Government Tourist Office, 2013). Based on the economic structure and other evidence, Bali has thus become a main tourist hub as well as a main economic factor for supporting food security which is written down in the Indonesia's Economic Master Plan 2011–2025 (The Coordinating Ministry for Economic Affairs, 2011).

Despite its economic benefits, tourism in Bali has been blamed for damage on the agricultural sector due to land use changes. As an environmental impacts, unfortunately, with the influx of tourism growth, rapid and unplanned development occurred in Bali, with then led to an increase in loss of important farmland due to encroaching development of more resorts for villas, hotels, restaurants, etc. (<http://share.nanjing-school.com>; Sutawa, 2012). Tourism has also heavily influenced and changed the traditional Balinese orientation towards land. Originally, before Balinese started leasing and selling land they considered land had no value except for its agricultural use. Parallel to the development of new villa and restaurant construction, is an increased demand and prices for land, and the use of farm land started to change (Parker, 2011). The sustainability of tourism is also dependent on an adequate water supply of sufficient quality and quantity but there is little research on the significance of water in tourism development. Whether in the developed or developing world, the per capita use of water by tourists far exceeds that of locals, and it is depended on the predominant tourist land use patterns and the hotel type (Cole, 2012). As Gossling (2001 *in* Cole, 2012) demonstrates water use grows exponentially with increasing hotel size.

Population growth and immigration Bali over the past decade are also likely contributors to the land use changes from the agricultural land to other land uses in Bali. Once one area has been converted to uses other than agriculture, the surrounding areas are also very likely to be progressively converted over the short term. These rapid changes likely occur as a consequence of the improvement in accessibility, housing, and/or industrialization of the area. In addition, increases in rice field prices in areas experiencing conversion encourage farm owners to sell their land. Further factors likely influencing land use change such as there is not allowing for building higher than 15 m based on regional regulation of Bali Province (2009-2029) and the agricultural land tax has progressively increased based on the warranty value of the taxed object.

Based on statistical data from 2012 (<http://bali.bps.go.id>), Bali Province (consisting of over 563,666 ha) comprises 81,744 ha (14.5%) of rice fields managed through 1,548 subak systems; 273,655 ha (48.55%) of other agricultural fields; and 208,267 ha (36.95%) of non-agricultural land. Over a period of 15 years (1997–2011), approximately 6,109 ha of rice fields were converted into non-agricultural land with an annual conversion rate of approximately 436.3 ha (0.5%). The top three out of the nine regencies experiencing land conversion in Bali were Denpasar, Klungkung, and Buleleng, with an annual conversion rate of 1.33% (35 Ha), 0.8% (31 ha), and 0.45% (50 ha), respectively. The rest area in 2013 was 81,165 ha, still dedicated to rice cultivation in Bali (Food Agriculture Agency of Bali Province, 2014). These data highlight the steep reduction in the extension of rice fields in Bali due to land use changes in the past, with no evidence that this trend will be reversed in the near future.

Saba watershed is one of the 391 watersheds in the Bali-Penida River Basin. It belongs to Saba-daya, one of the 12 watershed management units within this basin (Ministry of Forestry, 2011). It geographically lies in the regencies of Buleleng and Tabanan (Unda-Anyar Watershed Management Agency, 2008), expanding over an area of 14,019 ha. From these, approximately 4,156.82 ha represent rice fields supported from the eight rivers (Saba, Getas, Dati, Jehe, Bakah, Titab, Panas, and Ling) irrigating basin (Bali-Penida River Basin Agency, 2005; Unda-Anyar Watershed Management Agency, 2008), managed through 56 subak systems.

Subak is a traditional irrigation system used in Bali since the year 882 (Public Works Agency of Bali Province, 1997). According to Bali Province Regulation No. 9 published in 2012, subak is defined as a traditional organization for water and/or crops management at the farm level custom from socio-agricultural societies from Bali, characterized by religious and economical features, which have historical growth and continue to grow and develop in present times (<http://simkum.baliprov.go.id>). It

includes both *subak sawah* (wet land subak) and *subak abian* (dry land subak) systems, although *subak sawah* is the system known simply as subak. Thus, *subak sawah* (hereafter subak) was developed based on the hydrological features of the area, rather than on administrative boundaries, and whose partitions are clearly defined by membership and management structures. The subak system arose as a consequence of the *Tri Hita Karana* (three happiness causes) philosophy, and it was included as cultural landscape of Bali in the UNESCO World Heritage List on July 6, 2012 (Ministry of Education and Culture, 2013). However, the interaction of several factors are currently threatening the sustainability of irrigated rice culture, including (1) the declining interest of youth in rural areas to find a job in the farming sector, particularly as rice farmers; (2) the declining in rice field extension due to conversion into non-agricultural land; (3) the increasing conflicts in the use of water resources; and (4) the deforestation and pollution of freshwater systems for irrigation (Sutawan, 2004).

Downstream and upstream of Saba watershed are the areas experiencing faster land use change in the whole region. Based on a preliminary survey including the 53 head of subaks within Saba watershed attending the 1st Bali stakeholders meeting held on September 8, 2013, approximately 25.15% of 2,903.79 ha of rice fields have been converted to other land uses such as clove plantations, vineyards, housing, villas, hotels and restaurants, public services (police office and public yard), and private business (storages, trading companies, and gas stations), or have been lost through the Titab dam construction over the past two decades.

This research aims to explore the potential of subak system to control land use change, and the possibility to develop a sustainable development plan integrating agricultural production and tourism, especially within Saba watershed, northern Bali, Indonesia refers to some experiences in Asia or international levels. Lack of land use planning in Turkish Mediterranean in 1960's, for example, where 816 ha in agricultural lands and 457 ha in natural forest areas were converted into hotels, service buildings and urban settlements for the growing tourism industry, damages coastlines and coastal communities (<http://responsibletravel.org>). In addition, explosive tourism growth since the 1993 in the Angkor basin of Cambodia, for example, has driven increasing demand for water, wood, and biomass fuel, and rapid and extensive land-use and land-cover change (Gaughan, 2008). Furthermore, paddy landscape of the Dongshan River Basin in Taiwan has changed significantly from 472,759 ha (1991) to 406,064 ha (2011), showing a 14% decrease over two decades due to tourism and housing development (Lin and Huang, 2014). Rural tourism development is significantly as a way of adapting to the process of agricultural transformation in Taiwan when Taiwan's agriculture production and marketing became more difficult due to the process of agricultural globalization, particularly under the effect of joining the WTO (Tsai, 2007). Appropriate management planning with integrating the tourism carrying capacity assessment and land use policies supported by comprehensive data framework is necessary, particularly if new tourism zones or areas are to be developed in accordance with the protection of all the natural resources, including land for sustainable agricultural production (<http://responsibletravel.org>). In this case, community-based agro-tourism development including in Japan is very important to increase local people earnings and improve their lives as well as sustain their agricultural operations (Itagaki, 2013). Subak system as a manifestation of the *Tri Hita Karana* Philosophy with beautiful cultural landscape supported by great image for Bali as a popular tourist destination has great opportunity to implement agro-tourism concept in order to minimize land use change of rice field and to realize the sustainable subak system based rice culture in Bali.

METHODS

Study area

This study is under Research Institute for Humanity and Nature (RIHN), Japan, C-09-Init project on "Designing Local Frameworks for Integrated Water Resources Management" (2012-2016)

in collaboration with Bogor Agricultural University and Udayana University, Indonesia. One of its study locations is the Saba watershed, Northern Bali, Indonesia. It covers approximately 14,019 ha (Geospatial Information Agency, 2000 in Setiawan *et al.*, 2013), which is part of both Buleleng and Tabanan Regencies (Fig. 1) including approximately 4,156.82 ha irrigated rice fields. There have been more than eight sub-rivers (Saba, Getas, Dati, Jehe, Bakah, Titab, Panas, and Ling) for supplying irrigation water inside the area. It includes 56 subak systems (Table 1) that traditionally manage irrigation water on their rice fields and 34 *subak-abian*(s) (Table 2) that traditionally manage crops in dry land farming system (Bali Provincial Government, 2011). It also covers part of five districts and 35 village areas (Table 1). The population number inside the 35 villages was 124,935 persons (<http://bulelengkab.bps.go.id>). It seemly has great experiences in land use changes from rice fields to non-agricultural use in more than last two decades.

Table 1. Name, location and water source of each subak within Saba watershed.

Regency (District)	Village	Subak	Water source		
			River	Weir	
Buleleng (Banjar)	1. Munduk	1. Munduk	Pulopane, Panas, Ling	Pulopane, Lampah, Bolangan, Munduk	
	2. Gesing	2. Gesing	Soka, Keladi	Gesing Beten, Gesing Dur 1& 2	
	3. Kayu Putih	3. Sanda	Ling	Sanda	
		4. Bolangan	Ling	Bolangan	
		5. Bebau	Ling	Banyuatis	
4. Gobleg	6. Tagallinggah	Ling	Sanda		
5. Banyuatis	7. Banyuatis	Ling	Banyuatis		
Buleleng (Busungbiu)	1. Umejero	1. Umejero	Water spring	Umejero	
		2. Lebah	Water spring	Umejero	
		3. Tanah Pegat	Water spring	Umejero	
		4. Celagi	Water spring	Umejero	
		5. Pelapuan	Panas	Pelapuan	
		6. Bengkel	Jehe	Bengkel	
		7. Keckeran	Jehe	Bengkel	
		5. Kedis	8. Pinjinan	Jehe	Pinjinan
			9. Kedis	Jehe	Kedis
			10. Timbul	Jehe	Timbul, Sibetan
6. Tinggarsari	11. Tinggarsari	Saba-Getas	Tinggarsari		
7. Subuk	12. Gebang	Selat-Titab	Gebang		
	13. Celokah	Saba	Celokah		
	14. Bukit Pulu	Titab	Bukit Pulu		
	15. Batu Megaang	Titab	Batu Megaang		
8. Pucaksari	16. Titab	Titab	Titab		
9. Titab	17. Asah Uma	Saba-Titab	Asah Ume		
	18. Dukuh	Saba-Titab	Dukuh		
	19. Busungbiu	Panas-Ling	Busungbiu-Tunju		
Buleleng (Seririt)	1. Gunungsari	1. Tunju	Panas-Ling	Busungbiu-Tunju	
	2. Mayong	2. Mayong	Panas	Joanyar	
	3. Joanyar	3. Joanyar	Panas	Joanyar	

Regency (District)	Village	Subak	Water source	
			River	Weir
4. Ringdikit	4. Ringdikit	4. Mayong Pengulkulan	Panas	Joanyar
		5. Tebu Merta	Bakah	Tebu Merta
		6. Lebah Semawa	Saba	Lebah Semawa
		7. Rawa	Saba	Rawa
	5. Rangdu	-	-	-
	6. Patemon	8. Anyar Patemon	Panas	Patemon Kajanan
		9. Patemon Kajanan	Panas	Patemon Kajanan
		10. Karangsari	Saba-Panas	Puluran
	7. Seririt-Sulanyah	11. Belumbang	Saba-Panas	Puluran
		12. Kapal	Saba-Panas	Puluran
	8. Sulanyah	13. Babakan	Saba-Panas	Puluran
		14. Batan Bekul	Saba-Panas	Puluran
		15. Puluran	Saba-Panas	Puluran
	9. Tangguwisia	16. Uma Desa	Saba Hilir	Saba
		17. Tegal Intaran	Saba Hilir	Saba
		-	-	-
	10. Pengastulan	18. Pongjocukli	Saba Hilir	Saba
		19. Banyumati	Saba Hilir	Saba
		20. Yeh Anakan	Saba Hilir	Saba
		21. Pangkung Kunyit	Saba Hilir	Saba
		22. Banjar Munduk	Saba Hilir	Saba
	11. Lokapaksa	23. Tegallenga	Saba Hilir	Saba
		24. Kalisada	Saba Hilir	Saba
	12. Ularan	-	-	-
13. Uma Anyar	1. Tukad Sumaga	Saba Hilir	Saba	
	2. Berombong	Saba Hilir	Saba	
14. Banjar Asem	1. Pujungan	1. Tibu Dalem	Getas	Tibu Dalem
	2. Pupuan	2. Yeh Saba	Saba Hulu	Yeh Saba
		3. Anyar Pupuan	Saba Hulu	Yeh Saba
		4. Tengladan	Saba Hulu	Yeh Saba
15. Kalisada				
TOTAL	35	56		

Source: Field investigation in 2013–2014

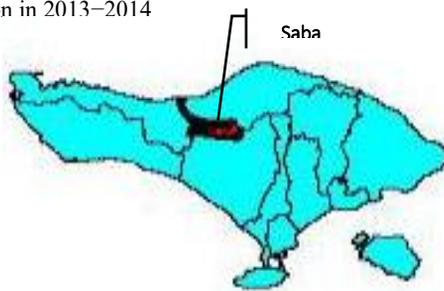


Fig. 1. Study area within Saba watershed

Table 2. Subak-abian list within Saba watershed

Regency (District)	Village	Subak Abian	Location Within Saba Watershed
Tabanan (Pupuan)	1. Bantiran	1. Teja Pala Amerta	Upstream
	2. Pupuan	2. Budi Tunggal	Upstream
	3. Pujungan	3. Batur Pendem	Upstream
Buleleng (Banjar)	1. Gobleg	1. Tunggal Sari	Upstream
		2. Guna Asri	Upstream
		3. Wija Sari	Upstream
		4. Galih Sari	Upstream
		5. Pala Sari	Upstream
		6. Taman Sari	Upstream
		7. Amerta Sari	Upstream
	3. Gesing	8. Wiratani I	Upstream
		9. Wiratani II	Upstream
		10. Wiratani III	Upstream
Buleleng (Seririt)	1. Gunung Sari	1. Widya Anantha Karya	Central
	2. Mayong	2. Anantha Sari	Central
	3. Rangdu	3. Taman Sari	Central
	4. Banjarasem	4. Mekar Sari	Downstream
	5. Umeanyar	5. Kertham Bhumi	Downstream
	6. Loka paksa	7. Wana Sari	Downstream
		8. Bila Sari	Downstream
		9. Gunungina	Downstream
		10. Kerta Bumi	Downstream
	7. Ularan	11. Eka Cita Wana	Central
	Buleleng (Busungbiu)	1. Busungbiu	1. Buana Sari
2. Kekeran		2. Padang Mesawen	Central
3. Bengkel		3. Widya Sari	Central
4. Umejero		4. Waa Amertha	Upstream
5. Kedis		5. Tri Dharma Laksana	Central
6. Tinggar Sari		6. Mandala Sari	Central
7. Pucak Sari		7. Mertha Sari	Upstream
	8. Pakang Aya	Upstream	
Buleleng (Gerokgak)	1. Tukad Semaga	9. Gunung Kutul	Upstream
		1. Giri Sari	Downstream
TOTAL		34	

The water quality within the Saba watershed (Table 3) is still good for irrigation and daily domestic use even downstream near the river mouth as well as upstream. River flows at the water sampling points are shown in Fig. 2. Based on the figure, with the exception of the Panas River, river flows in the wet season were approximately four times that in the dry season (Nakagiri *et al.*, 2013). During the period of 1995–2004, the annual rainfall tended to decrease with time. Before the great El-

Nino in 1997, rainfall reached 2,500 mm in 1995 but later on decreased significantly and reached below 1,500 mm (Setiawan *et al.*, 2013).

Table 3. Water quality inside Saba watershed.

Item	Value	Description
pH	Wet: 7.5–8.4	Alkaline (near normal)
	Dry: 7.9–8.2	Rich in mineral (Na, Mg, Ca)
Electric cond. (EC)	Wet: 23–49 mS/m	Slightly higher than normal (due to rich minerals)
	Dry: 18–41 mS/m	
Suspended Solid (SS)	Wet: 8–63 mg/L	Very clear in dry season
	Dry: <0.2 mg/L	Silty during the wet season
Total Nitrogen (TN)	Wet: <1 mg/L	Oligotrophic even downstream
	Dry: <1 mg/L	
Total Phosphor (TP)	Wet: <0.05 mg/L	Oligotrophic even downstream
	Dry: <0.05 mg/L	
Chemical Oxygen Demand (COD)	Wet: 3–17 mg/L	Slightly high although near normal
	Dry: 3–8 mg/L	

Source: Nakagiri *et al.*, 2013

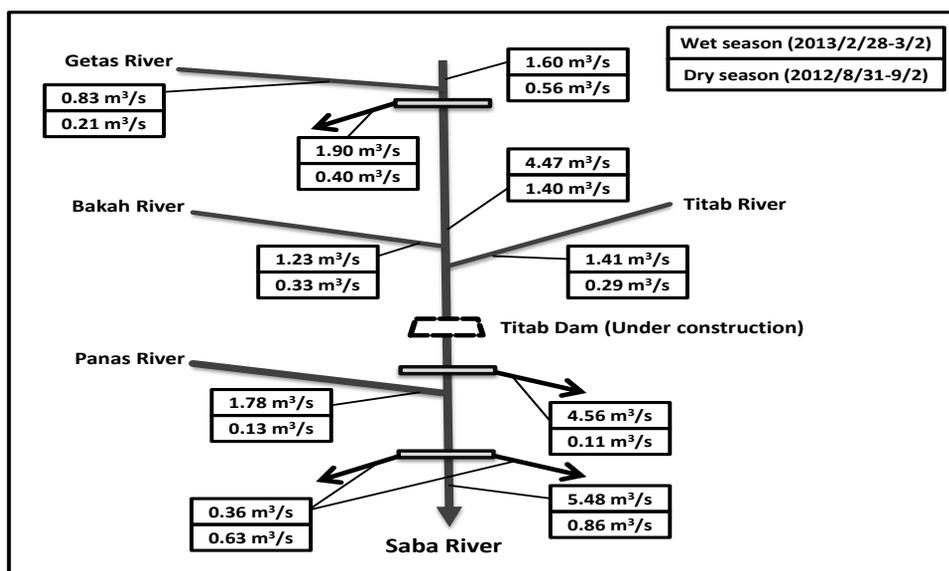


Fig. 2. River flow at water sampling points (Nakagiri *et al.*, 2013)

Data collection

This research focused on Saba watershed, one of the 391 watersheds in Bali Province. We selected 20 subak areas from a total of 56 subak systems, distributed in the upstream, central, and downstream areas of this watershed (Table 4) currently or recently experiencing land use changes. From the selected subaks, many have been identified as the potential destination for subak-based agro-tourism as a new strategy to sustain current levels of agricultural production in Bali.

Table 4. Subak areas studied within Saba watershed.

No	Subak	Location			Land Use Change to
		Village	District	Watershed	
1	Munduk	Munduk	Banjar	Upstream	Clove plantation and villa
2	Gesing	Gesing	Banjar	Upstream	Clove plantation and villa
3	Sanda	Kayuputih	Banjar	Upstream	Clove plantation
4	Bolongan	Kayuputih	Banjar	Upstream	Clove plantation and houses
5	Bebau	Kayuputih	Banjar	Upstream	Clove plantation and houses
6	Tegallinggah	Gobleg	Banjar	Upstream	Clove plantation
7	Bukit Pulu	Subuk	Busungbiu	Middle stream	Clove plantation
8	Titab	Titab	Busungbiu	Middle stream	Clove leave distillation
9	Asah Uma	Titab	Busungbiu	Middle stream	Titab Dam
10	Dukuh	Busungbiu	Busungbiu	Middle stream	Titab Dam
11	Belumbang	Sulanyah-Seririt	Seririt	Downstream	Vineyards, houses, hotel, public service, private business
12	Babakan	Tangguwisia	Seririt	Downstream	Vineyards
13	Batan Bekul	Tangguwisia	Seririt	Downstream	Vineyards, houses, hotel, villa, private business
14	Puluran	Pengastulan	Seririt	Downstream	Vineyards, houses, private business
15	Umadesa	Lokapaksa	Seririt	Downstream	Vineyards, houses, hotel, villa, private business
16	Ponjokcukli	Umeanyar	Seririt	Downstream	Vineyards
17	Banyumati	Umeanyar	Seririt	Downstream	Vineyards and villa
18	Yeh Anakan	Banjarasem	Seririt	Downstream	Fishpond, houses, villa, rice milling unit
19	Tegallenga	Kalisada	Seririt	Downstream	Vineyards, houses and villa
20	Kalisada	Kalisada	Seririt	Downstream	Houses and villa

Source: Preliminary field survey conducted on September 8, 2013

We interviewed the head of each subak area using the Rapid Rural Appraisal (RRA) method to gather primary data between August 27, 2014 and December 7, 2014. We also collected secondary data from appropriate sources such as the detail land use planning data of the Seririt District, the list of each subak members, written (*awig-awig*) and unwritten (*pararem*) regulations of each subak, and agreements between subak and investors regarding land use conversion inside the subak area and their contribution for sustaining the subak activities.

Data analysis

We identified the different subak areas for a potential target as agro-tourism destinations or for the development of tourism-related infrastructures using a transect method throughout the study period. We collected physical data in the form of observational and photographic documentation. All data were analyzed using descriptive qualitative analysis.

RESULTS AND DISCUSSION

Land use changes in subak areas

The operational foundation of the *Tri Hita Karana* defined three sub-systems within the subak: (1) The religious element (*parhyangan*) identified by the presence of *Ulun Danu*, *Ulun Empelan*, *Ulun Swi*, and *Bedugul* temples to honor God. The *Ulun Danu* temple which found near the lake, the *Ulun Empelan* temple is close to the diversion weir, the *Ulun Swi* temple is close to the division structure, the *Bedugul* temple is within the subak area, and the *Tugu Shrine* or *Ulun Carik* temple is within each farmer's land plot; (2) The social element (*pawongan*) includes members of subak and the subak board and its organization structure. Finally, (3) the environmental element (*palemahan*) includes the rice fields, irrigation network, water source, and all other sites used by the subak members to implement activities related to farming and religious ceremonies. These elements are strongly interconnected, and the destruction of one leads to the disappearance of the entire subak system.

The subak rule known as the *awig-awig* is established by the subak members through general consensus and approval by the subak assembly, and it is ultimately responsible for the implementation of the subak philosophy within the system. This rule includes both written and unwritten norms that are accepted by the subak members. In general, the rule comprises the name, address, and boundaries of each subak; ideological, constitutional, and operational philosophies, subak objectives; requirements, rights, and duties of the subak board; requirements, rights, and duties of the subak members; regulations regarding the subak assembly and board meetings; is absentia permits (*leluputan*) for subak members; the use of wooden bells (*kulkul*) in the subak; subak properties (temples, buildings, granary, symbolic/stamp, and the sources of income); ceremonies and the holiness of subak; rules regarding rice field boundaries, irrigation water, crops, pest and diseases control, prohibitions following transplantation; and penalties and punishments. Any additional unwritten rules (*pararem*) are also in support of *awig-awig* (PHDI Bali Province, 1984). An *awig-awig* is hand written on *lontar* (a palm tree) dry leaves using the old Balinese language and/or type-written. A ritual ceremony known as the *pasupati* is sometimes performed by all the subak members to pray for magic-religious strengths and the rightfulness of the *awig-awig*. A written *awig-awig* could be signed by the subak board, then registered at the local government by an officer. Table 5 shows the 16 subak areas (80%) that have written *awig-awig* while the remaining subak have only unwritten *awig-awig*.

Land use change is the main factor for the disappearance of subak systems and have been more profound in the upstream, central, and downstream areas of Saba watershed. In this region, many rice fields have gradually converted into other land use types over the past two decades. Table 5 shows that 36.97% of the rice fields have been converted into other land that uses the 1,223.42 ha of rice fields in 1991 to 771.13 ha in 2014. These rice fields (approx.. 165.91 ha) were converted into clove plantations, vineyards (129.72 ha), dragon fruit (1.5 ha), fishpond (20 ha), approximately 104.16 ha have been converted into houses, tourism-related infrastructure, offices, public yards, and other private business (e.g., shops, gas stations, and rice milling units), while the Titab dam construction (2011–2015) claimed a total of 31 ha.

The role of the subak system and tourism on land use changes.....

Table 5. Land use change experienced by the 20 subak areas

No	Subak	Rice field (ha)		Member (person)†	Land converted (ha)		Awig-awig	Water supply/ availability
		1991	2014		Plantation	Building		
1	Munduk	131.83	40	246	Clove (90.33)	Villa (1,5)	Written	Enough
2	Gesing	26	5	17	Clove (20.5)	Villa (0.5)	Unwritten	Deficit in Dry Season (DS)
3	Sanda	39	20	46	Clove (18.95)	Houses (0.05)	Written	Enough
4	Bolongan	35	15	44	Clove (18)	Houses (2)	Unwritten	Enough
5	Bebau	64	51	76	Clove (10)	Houses (3)	Unwritten	Enough
6	Tegallingah	16.20	9.09	49	Clove (7.01)	Houses (0.1)	Unwritten	Deficit in DS
7	Bukit Pulu	7.90	5.88	16	Clove (1.12)	Houses (0.9)	Written	Enough
8	Titab	12.93	12.83	36	0	Clove leave distillation industry (0.1 Ha)	Written	Enough
9	Asah Uma	25.00	7.00	44	0	Titab Dam (18)	Written	Enough
10	Dukuh	13.00	0.00	0	0	Titab Dam (13)	Written	Enough
11	Belumbang	115	78.1	160	Vineyards (10)	Houses, hotel, public service, private business (26.9)	Written	Enough
12	Babakan	12.3	11.3	28	Vineyards (0.77)	Houses (0.23)	Written	Enough
13	Batan Bekul	42.59	23.58	80	Vineyards (13.98)	Houses, hotel, villa, private business (5.03)	Written	Deficit in DS
14	Puluran	88	55	100	Vineyards (5)	Houses, private business (28)	Written	Deficit in DS
15	Umadesa	173.85	92.37	395	Vineyards (72.94)	Houses, hotel, private business (8.54)	Written	Enough
16	Ponjokcukli	17.91	14.05	34	Vineyards (3.86)	0	Written	Deficit in DS
17	Banyumati	84	71.56	156	Vineyards (10)	Villa (2.44)	Written	Deficit in DS
18	Yeh Anakan	95.78	71.68	132	Fish pond (20)	Houses, villa, RMU (4.1)	Written	Enough
19	Tegallenga	122.84	96.17	123	Vineyards (11.67)	Houses and villas (15)	Written	Deficit in DS
20	Kalisada	100.29	91.52	140	Dragon fruit (1.5), Vineyards (1.5)	Houses and villa (5.77)	Written	Deficit in DS
Total		1,223.42	771.13	1,922	317.13	135.16		

Notes: † Including converted land owners

Land conversion for clove, vineyards and/or dragon fruit plantations at the household level was evident in 16 subak systems. Approximately 20 ha of rice fields within Subak Yeh Anakan were also converted into fishpond during the 1990s. In addition, nine subak areas had succumbed to tourism development through the construction of tourism-related infrastructure in rice fields upstream and downstream of Saba watershed by developers not related to the subak system. Only one subak system, namely the Subak Ponjokcukli, has not experienced any construction-related land use changes because of its restrictive regulation, which does not allow constructions within the subak area. On the other hand, all other subak areas do not have any restrictive regulations in place to control land use change inside each area.

The reasons behind land use changes driven by rice fields owners fall within five categories: (1) economic reasons associated with variations in land prices; (2) extreme market prices due to a high demand for land to build houses or private business including tourism-related infrastructure such as villas, home stays, hotels, and restaurants; (3) limited irrigation water supply especially during dry seasons; (4) limited human resources available for the agriculture sector, especially for rice farming; (5) lack of fixed spatial and land use plans with the existing legal framework prohibiting the use of rice fields within the prescribed zones for non-farming activities; (6) lack of satisfactory compensating policies for traditional rice farmers. For economic reasons, some farmers have aimed to obtain higher benefits through agricultural practices by converting rice farms into other types of agricultural production, such as clove plantations in the uplands and vineyards in the downstream of the watershed. Annual gross margin yielded from a hectare of clove plantation is approximately IDR250,000,000 whereas rice farming generates only IDR32,000,000, approximately. Furthermore, the annual gross margin generated from a hectare of vineyards is approximately IDR60,000,000 compared to the IDR42,000,000 that can be obtained from rice farming. The exchange rate in 2014 is approximately IDR12,000 for USD1 (<http://bi.go.id>).

In the current economic climate, it is not unusual that the sale of rice field at the highest market price to build houses encourages the surrounding land owners to sell all or part of their land for similar purposes. As an example, the rice field price inside both Subak Dukuh and Subak Asah Uma reached IDR10 M for each 100 m² during the land acquisition process associated with the construction of the Titab dam between 2011 and 2015. Before the construction of the dam, rice field prices only reached IDR2.5 M per 100 m² of land, approximately. This project required approximately 138 ha of land, including a flooded area of 68.83 ha, a green belt of 44.67 ha, and 24.5 ha allocated for infrastructure and construction activities. Table 5 shows that approximately 31 ha of rice fields within the Subak Dukuh and Subak Asah Uma areas have been lost under the waters of the Titab reservoir. In this case, the entire extension dedicated to rice fields (13 ha) inside Subak Dukuh were flooded following the construction of the dam causing the subak to disappear altogether. This project was supported by the Indonesian government through the allocation of IDR481,893,341,000 including IDR468,716,771,000 allocated to the construction of the dam and IDR13,176,570,000 dedicated to project management. This dam was built to (1) regulate 1,794.82 ha of irrigated area including the Puluran irrigation command area (398.42 ha) and the Saba irrigation command area (1,396.40 ha), (2) to ensure a domestic water supply of 350 Ls⁻¹ for the Seririt and Banjar district areas; (3) to provide approximately 1.5 MW of electricity to the Busungbiu district area; and to promote (4) fishery industries and (5) tourism development (Bali-Penida River Basin Agency, 2012). In case of the land use change of rice field due to Titab Dam construction, some farmers who would like to continue their rice culture outside of Titab Dam location, feel worse-off due to the number of rice field can be paid by themselves are less than the number when it was sold. The losses is due to the price of rice field for each 100 m² is strongly different between IDR10 M (inside Titab Dam) and IDR40 M (nearest Titab Dam). Whereas, the rice field price near Seririt city highway reached IDR400 M per m². This land is not being mainly occupied by buildings such as business centers (shops or trading companies), hotels, and restaurants. For those areas with nice views, such as those

overlooking rice terraces, or those that have sunset, sunrise, beach, mountain, or waterfall views, have a great potential for tourism development and the land prices tend to be higher. Rice field prices for areas near Seririt city, such as those inside Subak Umadesa, readily available for the construction of houses or villas can reach up to IDR125 M per 100 m². In areas upstream of the Saba watershed, including areas within Subak Gesing, the price of rice farm lands can reach IDR50 M per m².

Other rice field owners have transformed their lands into houses for rent the rooms on a monthly or annual basis. In these cases, the monthly rate of a 12 m² single room ranges between IDR300,000 and IDR500,000. Some prestigious hotels such as Gran Surya have followed this conversion strategy inside Subak Belumbang, where currently a deluxe room can be offered for IDR750,000 a night. Lesong Hotel inside Subak Gesing in the uplands of the Saba watershed offers double rooms with mountain, sunrise, sunset, and rice terrace views at a rate of IDR800,000 a night. On the other hand, Villa Pelangi inside Subak Batan Bekul built on 600 m² of rice fields with the permission of local government and was offering in 2011 group rates (maximum 10 persons) of IDR15 M for one week. This evident highlights the current options that rice farm owners currently have regarding land prices and business opportunities besides rice farming. Unfortunately, when all rice fields in a subak have changed to non-agricultural uses, the subak itself disappears. To this end, several strategies could be proven efficient to mitigate land use change for irrigated land, including (1) careful spatial and land use planning considering water resources availability and (2) creation of a legal framework restricting the use of paddy land within prescribed zones, prohibiting non-farming activities through strict law enforcement and implementation (Sutawan, 2004).

Potential development of a subak-based agro-tourism industry

The Ministry of Tourism (<http://www.kemenpar.go.id>) encouraged local governments to decide on the use of local agricultural products, and stakeholders to synergize and integrate agriculture and tourist industries in an attempt to promote both agriculture and tourism sectors in Indonesia. Currently, two models of agro-tourism development are being considered in the region: capital-based and community-based agro-tourism (Budiasa and Ambarawati, 2014). Subak-based agro-tourism shows a great potential as a tool to develop community-based agro-tourism to achieve sustainable agriculture in Bali. This arose as a great opportunity for subaks to develop as business units, and received formal governmental support through the President instruction No. 3 in 1999 within the New Policies of Irrigation Management in Indonesia (Indonesia Government, 1999). Consequently, the tourism industry in Bali has developed on the basis of the Balinese culture as defined in the Bali Provincial Regulation No. 2 published in 2012, the improvement of Bali Provincial regulation No. 3 published in 1991, and the regulation oldest one No. 3 in 1974 (Bali Government Tourism Office, 2012). Furthermore, Balinese culture includes Balinese agriculture, and in particular, irrigated rice cultivation managed through the subak system.

Five of the subak studied here present a great potential for agro-tourism development: Subak Munduk and Subak Sanda upstream, Subak Titab and Subak Asah Uma in the central parts, and Subak Pongjokukli downstream of the Saba watershed. All these subaks are easily accessible by car. Subak Munduk is located at Munduk Village, well-known as an ecotourism destination for European tourists, especially from France, Italy, Germany, and the Netherlands. This area is located 25 Km, 42 Km, and 71 Km from the center of the Banjar district, and the urban centers of Singaraja and Denpasar, respectively.

Munduk Village is a tourist destination located between 500 and 1,500 m above sea level and presents a mean annual temperature of 26°C and annual rainfall of 22.3 mm. It is a popular destination due to the beautiful Melanting waterfall (100 m) and its proximity to the Tamblingan lake and natural forest. Subak Munduk comprises nine sub-subaks (*tempeks*) with 246 members managing 40 ha of

irrigated rice fields and 90.33 ha dedicated to clove plantation. Approximately 1.5 ha of rice fields inside Tempek Suradadi has been converted into Villa Puri Lumbung, representing the first tourist accommodation constructed in 1992 in this region; an additional extension of 1.5 ha were kept as rice fields by Nyoman Bagiarta, the villa owner, for its views from the villa. Local rice farmer usually produce local red rice of the variety *cendana* during the rainy season and hybrid white rice during the dry season. Red rice farming usually starts in January and continues through June, while white rice is cultivated between August and November. Annual cropping pattern that have been implemented within subak sample is shown in Fig 3. Currently, those land owners that have converted their rice fields into clove plantations continue to be subak members and obey *awig-awig* in Subak Munduk. In addition, the owners of the remaining 40 ha of rice fields under the subak have agreed to keep their land for beauty of rice terrace landscape to support rural tourism under the agreement of receiving compensation in the form of fertilizers and the land tax subsidies.

No	Subak	Annual Cropping Pattern in 2014												
		Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec	
1	Munduk	Local red rice						E	Hybrid rice				E	
2	Gesing	Local red rice						E	Hybrid rice				E	
3	Sanda	Hybrid rice			Hybrid rice			Empty (cut pest attack)						
4	Bolongan	Hybrid rice			Hybrid red rice			Empty (cut pest attack)						
5	Bebau	Hybrid rice				Hybrid rice				Hybrid rice				
6	Tegallinggah	E	Hybrid rice				Hybrid rice				Second crops			
7	Bukit Pulu	Hybrid rice			Hybrid rice			Hybrid rice						
8	Titab	Hybrid rice				Hybrid rice				Hybrid rice				
9	Asah Uma	Hybrid rice				Hybrid rice				Hybrid rice				
10	Dukuh	TITAB DAM UNDER CONSTRUCTION												
11	Belumbang		Hybrid rice				Hybrid rice / second crops				Hybrid rice			
12	Babakan	Hybrid rice				Hybrid rice				Hybrid rice				
13	Batan Bekul	Hybrid rice				Hybrid rice				Hybrid rice				
14	Puluran	Hybrid rice				Hybrid rice				Hybrid rice				
15	Umadesa	Hybrid rice				Hybrid rice				Hybrid rice				
16	Ponjokcukli	Hybrid rice				Hybrid rice				Hybrid rice				
17	Banyumati	Hybrid rice				Hybrid rice				Hybrid rice				
18	Yeh Anakan	Hybrid rice			Hybrid rice			Second crops			E			
19	Tegallenga	Hybrid rice				Hybrid rice				Second crops				
20	Kalisada	E	Hybrid rice				Hybrid rice / second crops				Second crops /Empty			

Notes:  = empty; Second crops involve soybean, corn, and/or ground nut.

Fig. 3. Cropping pattern inside 20 subak areas

Subak Sanda (20 ha) is located in Kayu Putih Village, close to Munduk and Gesing villages. Tourists staying in these villages often engage in walking and trekking activities in rice field within Subak Sanda. This region also includes a beautiful natural pool near the subak area formed by the Ling River that is suitable for bathing. The beautiful rice terraces inside the subak also represent a tourist attraction, together with the beautiful Lesong Hill and the sunrise and sunset views. This subak also has the potential to offer additional benefits to local farmers if the landscape is managed as an area of natural beauty able to offer agro-touristic commodities and attractions.

Subak Titab and Subak Asah Uma are located near the Titab dam site. The approximately seven hectares of rice fields remaining in the Subak Asah Uma, and presents the potential of becoming a gateway to access the Titab dam from upstream. However, this region has repeatedly suffered from heavy flooding from the Titab River, and the bridge across the river providing access to the dam has been washed out twice due to floods. These same events have also affected a number of rice fields, which were lost as a consequence. Nevertheless, the remaining rice fields in the Subak Titab, extending over approximately 12.83 ha, have potential to develop for community-based agro-tourism. Farming activities associated with agro-touristic commodities and ceremonies, the holiness of the subak, and its water sources could therefore be exploited as tourist attraction in both Subak Titab and Subak Asah Uma. In addition, the beautiful views of the rice terraces in Subak Titab are easily accessible from the public road; in fact, many tourists already stop in road view point nearby. Subak members can potentially benefit from agro-tourism activities in the future; however, they lose these opportunities when the land is sold to investors from outside the subak.

The last subak assessed in this study is the Subak Ponjokcukli, located inside the Village of Uma Anyar in the Seririt district, downstream of Saba watershed. It is a coastal area including 14.05 ha of rice fields and 3.86 ha of vineyards exploited by Hatten Wines. This area is easily accessible from the road up to the point where the subak area reaches the beach. This is a beautiful subak with a big water spring located behind the Segara Temple near the beach. It has great potential as a tourist attraction for religious tourism and one can bath in the holy water. Its beach is very popular among tourists for diving and watching the beautiful sunset. Putu Lidituari, a local employed at a small hotel in Buleleng regency, proposed a master plan for community-based tourism inside Uma Anyar Village including the Subak Ponjokcukli area. This plan was accepted and approved by the head of Uma Anyar Village, the head of customary village, and head of Subak Ponjokcukli. It requires an initial budget of approximately IDR2,000,000,000 as proposed through the head of Buleleng regency. This master plan is appropriate within current development plan, as Uma Anyar village is one of the eight villages included in the zoning detail plan for Seririt city resort 2014–2034 (Regional Development Plan Agency of Buleleng Regency, 2014). Based on this zoning plan, Uma Anyar Village has been allocated as both agriculture and tourism sub-zones. Tourism sub-zone include agricultural fields to be developed into tourism-related infrastructure, including villas, hotels, resorts, viewing points, and infrastructure to support other tourism activities. In this case, a community (subak)-based agro-tourism plan might be useful and helpful to maintain the subak system, to achieve sustainable agriculture, and to improve social welfare in Uma Anyar Village.

CONCLUSIONS AND IMPLICATIONS

Most subaks within the Saba watershed do not have enough power or control over large-scale land use changes inside the subaks area; to this end, only Subak Ponjokcukli downstream of the watershed have remained unchanged mainly because of the restrictive nature of its *awig-awig*, which prohibits subak members to build on their own rice fields. Land costs and increases in land prices are the main causes prompting farmers to sell their land. In fact, with the exception of Seririt City Resort, there are no specific spatial and land use plans or regulations prohibiting the conversion of rice fields into non-agricultural land types. In addition, the compensations and incentives to farmers to maintain traditional rice cultivation are limited in the region.

The development of subak-based agro-tourism models might be ideal to promote sustainable development of both agriculture and tourism sectors in Bali. Here, we identified several subaks inside the Saba watershed, such as Subak Munduk, Subak Sanda, Subak Titab, Subak Asah Uma, and Subak Ponjokcukli, as presenting great potential for the development of community-based agro-tourism industries based on their natural resources and governmental support.

The subak system in Bali as inscribed by UNESCO in 2012 is endangered by land use change. Thus, there is a pressing need to develop and improve strategies to control land use changes, especially inside the Saba watershed, including comprehensive spatial and land use planning for Seririt City Resort (2014–2034). Efforts to promote the interest of young people to preserve the subak system are also essential. Those subak systems presenting high potential for the development of integrated agro-tourism strategies should be empowered and financially supported by the central and local governments, and ultimately designed and managed by the local society, including young people.

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ANALYSIS OF THE TECHNICAL EFFICIENCY OF PINEAPPLE PRODUCTION IN SUBANG DISTRICT, WEST JAVA PROVINCE, INDONESIA

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ABSTRACT

Estimating technical efficiency of agriculture production is important for policy purposes. The study examined the technical efficiency of pineapple production of Subang district using cross section data from 142 pineapple growers. Primary data were collected from 4 villages in Subang districts in May-June 2013. Cobb-Dougllass production function model was used to estimate farm level technical efficiency using Maximum Likelihood Estimation (MLE) Method. The results showed that seed, hired labor, plant growth regulator, dung and chemical fertilizer have a positive and significant influence on pineapple yield. The average farm level of technical efficiency was 58.6 percent. The technical inefficiency was significantly affected by farmer's age, farming experience and off-farm income and land's ownership. The study revealed that there is possibility to further increase pineapple productivity.

Key word: Cobb-Douglas production function, Stochastic Frontier Analysis (SFA), Maximum Likelihood Estimation (MLE) Method, technical inefficiency

INTRODUCTION

As other tropical fruits, pineapple (*Ananas comosus*) spreads in many developing countries mainly in Asia, Africa and America continents. In Indonesia, pineapple cultivation spreads fairly in all provinces with the four biggest pineapple production centers are located at Lampung Province (33.47%), North Sumatra Province (14.98%), East Java Province (11.23%) and West Java Province (9.97%) (DG of Horticulture, 2013). In the last five years, there has been an increasing trend from other provinces mentioned above which grown pineapple commodity, from 19.42 percent in 2008 become 30.35 percent in 2012. This showed that pineapple commodity is very potential to be cultivated due to the increasing of fresh and processed pineapple's market demand in the last few years (Kleemann and Effenberger, 2010).

Fresh pineapple is the main raw material for pineapple processed products which become important in Indonesian economic sector as the highest volume and value in fruits export commodity in the last few years. It also became the third biggest fruit production in Indonesia after banana and mango commodities in 2012 (BPS, 2014).

Subang district is one of the main pineapple production centers in West Java Province where majority of the pineapple growers are the small scale's farmer which selling their outputs to the domestic (local) markets. The challenges faced by the pineapple growers with the limitation scale are lack of mechanical equipment, credit facility, land's ownership, low output price also lack of trust and commitment between the grower-buyer, lack of capital and plant diseases (Abbam, 2009 and Achaw, 2010). Main problems in pineapple cultivation in Subang district are decreasing of harvested land, production and productivity. As pineapple cultivated from generation to generation, farmers are using traditional technique with limited input and technology. Limited land area and scattered location of the land's ownership in Subang district caused diversity on the output been produced. One way to mitigate the problem is intensify the use of improved technologies and improves the efficiency of farmers in using inputs (Manganga, 2012).

Despite the importance of pineapple industry, there has been lack of studies that provides estimates of technical efficiency of pineapple farmers. Thus, the main objective of the present study is to examine technical efficiency in pineapple production in Subang district, by employing a Stochastic Frontier Analysis (SFA) and examine the factors affecting variations in these technical inefficiency scores. The results of this analysis offer information for policy makers and researchers to improve the existing condition.

METHODOLOGICAL FRAMEWORK

Farrell (1957) categorized economic efficiency into technical efficiency, allocative efficiency and scale efficiency. Technical efficiency is ability of a producer or farmer to use input and available technology with the most effective way. The approaches to measuring technical efficiency can be grouped into non parametric frontiers and parametric frontiers. Non parametric frontiers do not impose a functional form on the production frontiers, no assumptions about the error term and used linear programming approaches. The most popular non-parametric approach has been Data Envelopment Analysis (DEA). Parametric frontier approaches impose a functional form on the production function and based on the assumption that any deviation from the production frontier is attributed to the random component reflecting measurement error, statistical noise and a farm-specific inefficiency component (Ogundele and Okoruwa, 2006). The most common functional forms include the Cobb-Douglas, constant elasticity of substitution and translog production functions.

Since pineapple production in Subang district is an example of single output and multiple input production, this study focuses on the use of parametric approach namely SFA to assess the technical efficiency based on the production frontier model. SFA developed by Aigner *et al.* (1977) treats deviations from best-practice as comprising both random error and inefficiency. The software used to calculate technical efficiency score by SFA method is Frontier 4.1 (Coelli *et al.*, 1998). Under the parametric approach, this study used the Cobb-Douglas stochastic production frontier to estimate efficiency levels for the pineapple production in Subang district due to the methodology employed requires that the production function be selfdual. It is also worth stating that this functional form has been widely used in farm efficiency analyses for both developing and developed countries. The production function is generally specified as equation (1) (adopted from Battese and Coelli, 1995) :

$$\ln Y_i = \alpha_0 + \alpha_1 \ln X_{i1} + \alpha_2 \ln X_{i2} + \alpha_3 \ln X_{i3} + \alpha_4 \ln X_{i4} + \alpha_5 \ln X_{i5} + V_i - U_i \quad (1)$$

where, Y_i (output) is the yield of pineapple (kg/ha) of the i^{th} farms; X_{i1} denote quantity of seed (clump/ha) of the i^{th} farms, X_{i2} denote quantity of hired labors (man-days/ha) of the i^{th} farms, X_{i3} denote quantity of plant growth regulator (liter/ha) of the i^{th} farms and X_4, X_5 denote quantities (in kg/ha) of dung and chemical fertilizer of the i^{th} farms; α_s are parameters to be estimated; V_i is a random error which is associated with random factors outside the control of the firm and it is assumed to be symmetric independently distributed as $N(0, \sigma_v^2)$; and U_i is a non-negative random variable

associated with farm-specific factors (under the control of the firm), which lead to the i^{th} firm inefficiency of production and ranges between zero and one, identically and independently distributed as $N(0, \sigma_u^2)$. The model of the stochastic frontier is composed of two equations, one equation specifies the main factors of production and the other equation specifies the variables that are assumed to cause inefficiency.

The technical efficiency of production for the i^{th} firm at the t^{th} observation is defined by formula (2):

$$TE_i = \exp(-E[u_i | \epsilon_i]), \quad i = 1, 2, \dots, N \quad (2)$$

where TE_i is defined as pineapple grower's technical efficiency in i^{th} farm, $\exp(-E[u_i | \epsilon_i])$ is defined mean from u_i with condition ϵ_i . Technical efficiency score between $0 \leq TE \leq 1$. Technical efficiency score has negative relationship with technical inefficiency score and only use for function that has certain output and input (cross section data) (Coelli *et al*, 1998).

Adopted from Battese and Coelli (1995), the model for assessing technical inefficiency effects in this study were defined by equation (3):

$$U_i = \delta_0 + \delta_1 Z_1 + \delta_2 Z_2 + \delta_3 Z_3 + \delta_4 Z_4 + \delta_5 Z_5 + \delta_6 D_1 + \delta_7 D_2 + w_i \quad (3)$$

where, U_i is the technical inefficiency of the i^{th} farm, Z_1 is the farmer's age (in years), Z_2 is the formal education of a farmer (in years), Z_3 is the farming experience (in years), Z_4 is number of family member (in people), Z_5 is the share of off farm income (in percent), D_1 is the dummy for cropping pattern (1=intercropping, 0=monoculture) and D_2 is the dummy for land ownership (1=farmer's own, 0=rent); δ_i are vector of unknown parameters to be estimated and will have coefficient with negative signs; and w_i is unobservable random variables which are assumed to be identically distributed, obtained by truncation of the normal distribution with mean zero and unknown variance σ^2 , such that U_i is non-negative. The hypothesis of the study estimates that these variables also influence negatively the level of technical inefficiency of pineapple growers in the study area. Prior estimating SFA method, using Tobit regression had found out that there were no multicollinearity among factors affecting technical inefficiencies in pineapple production.

Efficiency measurement is the beginning step to exam individual production unit performance and to constitute a rational framework for the distribution of resource between and within production units

DATA INFORMATION

This study used primary data of 142 respondents from the total of 282 pineapple growers located in Subang district, West Java Province, which is one of the major pineapple growing districts in Indonesia. It accounted for 98.53 percent of the total West Java Province production in 2011. Based on the criterion of the highest area under pineapple, four villages were selected namely Tambak Mekar, Curug Rendeng, Bunihayu and Tambakan. From each selected village, simple random sampling technique was used to select pineapple growers proportionately to size. The number of respondents from Tambak Mekar, Curug Rendeng, Bunihayu and Tambakan villages were 21, 15, 28 and 78, respectively. Data were collected using a structured questionnaire through personal interviews and conducted in May-June 2013.

Data such as output, input use, socio-economic and institutional variables were collected. Descriptive statistic variables stated in Table 1.

Table 1. Variables of statistic descriptive used in the study

Variables (unit)		Means	Min	Max
Age	(the age of pineapple grower in years)	54	27	79
Education	(formal education of grower in years)	6.91	1	14
Farming Experience	(number of pineapple growing in years)	18.39	1	50
Family Member	(number of member per household in people)	4.48	1	9
Off-farm Income	(off-farm income of the pineapple grower in %)	1.57	0	98
<u>Dummy Variables</u>		Category	Frequency	Percent
Cropping Pattern	Monoculture (0)		37	26.06
	Intercropping (1)		105	73.94
Land Ownership	Rent (0)		29	20.42
	Farmer's own land (1)		113	79.58

RESULTS AND DISCUSSION

The estimated pineapple production function is furnished in Table 2. It is evident from Table 2 that all variables included in the model were positive and significant except for chemical fertilizer. In this study, chemical fertilizer variable had been used in terms of data limitation on single fertilizer used by the pineapple growers. Hence, increase in the use of inputs such as seed, hired labor, plant growth regulator and dung would increase the yield of pineapple by 0.13 percent, 0.16 percent, 0.48 percent and 0.24 percent, respectively.

Table 2. Maximum-likelihood estimates of the stochastic frontier production function for Pineapple in Subang, Indonesia

Variables	Regression coefficient	Standard error	t-ratio
Regression constant	4.125	1.035	3.985
Seed (clump/ha)	0.126 ^c	0.068	1.834
Hired labor (man-days/ha)	0.163 ^c	0.092	1.764
Plant growth regulator (liter/ha)	0.479 ^a	0.104	4.592
Dung (kg/ha)	0.241 ^b	0.102	2.359
Chemical Fertilizer (kg/ha)	0.115	0.089	1.289
Sigma-squared	0.408	0.111	3.649
Gamma	0.351	0.349	1.003

Note: a,b,c = Significant at $\alpha=0.01$, $\alpha=0.05$ and $\alpha=0.1$

The yield of pineapple varied between 928.07 and 100,000 kg/ha with an average of 14,243 kg/ha. The average seed, hired labor, plant growth regulator, dung and chemical fertilizer used by the pineapple growers were 35,651 clump/ha, 631 man-days/ha, 4.6 liter/ha, 14,086 kg/ha and 914 kg/ha, respectively. Table 2 indicates that the estimated variance parameter of the model (γ) was 0.35 which implies that about 35 per cent of the variation in pineapple output was due to farmer's practices rather than random variability. Sigma-squared is small (0.408), meaning that there were insignificant changes in the agricultural outputs of the pineapple growers over the past decade.

The percentage distribution of farms based on technical efficiency in pineapple production using decile class distribution is presented in Table 3. A perusal of Table 3 reveals that zero percent of the sample farmers were in the most efficient category (> 90 percent) and 13.3 percent were in 81-90 percent category. The mean level of technical efficiency was 58.6 percent, thus there is a possibility to increase the yield of pineapple production by 41.4 percent by using the resource at their disposal in an efficient manner without introducing any other improved inputs and practices and without any additional cost or to produce the current output, pineapple growers can reduce the use of inputs production by 41.4 percent with the existing technology. Based on Table 2, it can be concluded that pineapple growers were in the position of increasing returns to scale as according to the theory, increasing returns to scale suggests that the increase of output is higher than inputs. The five inputs variables using in this study are categorized as quasi-fixed inputs. These results were lower compare than pineapple technical efficiency research in Sri Lanka which was 85 percent with SFA method (Amarasuriya *et al.*, 2007). Idris *et al.* (2013) also found that pineapple was technically inefficient in Malaysia with non parametric approach (DEA method).

The minimum and maximum technical efficiency levels were 24 and 90 per cent, respectively. This shows that there is a wide disparity among pineapple growers in Subang district in their level of technical efficiency which indicating that there is possibility for improving the existing level of pineapple production through enhancing the level of farmer's technical efficiency. So, in the long run, it needs attention at policy level to introduce other best alternative farming practices and improved technologies to alleviate the overall Subang pineapple shortage.

Table 3. Frequency distribution of pineapple farms based on technical efficiency

Technical efficiency class (%)	No. of farms	Percentage to total farms
0 – 32	14	9.9
33-39	15	10.6
40-49	14	9.9
50-54	14	9.9
55-59	15	10.6
60 – 65	15	10.6
66 – 70	15	10.6
71 – 74	13	9.2
75 – 85	14	9.9
86 – 100	13	9.2
Total	142	100
Means		58.6 %
Min		24 %
Max		90 %

The determinant analysis of technical inefficiency is important as a basis for informing agricultural policy on what action to be taken to improve smallholder agricultural productivity, hence reduce resource wastage and improve farmer’s livelihoods (Tchale, 2009). Table 4 shows that technical inefficiencies are influenced negatively and significantly by share of off-farm income and land’s ownership.

In this study, a negative and statistically significant relationship between off-farm income and land’s ownership variables and technical inefficiencies were observed at 10 percent level of significance. A negative coefficient implies that any increase in the value of the variable lead to reduce the level of technical inefficiency (increase the efficiency).

Share of off-farm income contributes negatively to increase the technical inefficiency or this finding suggest that growers who are only involving in farming are less technical efficient than those who get the off-farm income. However, this result contradicts that of Amasuriya *et al.* (2007), Manganga (2012) and Mussa (2012) but consistent with results of Tchale (2009). The complementary income from other sources on and off the farm is likely to result in high on-farm productivity, as farmers can use income from other sources to invest in farm operations.

Land’s ownership is defined as farmer’s own land or land renting. The pineapple growers have to pay the land rent higher than if they only pay the land tax, so farmer who own the land will encourage increasing the technical efficiency.

Table 4. Estimates of the factors affecting technical inefficiency in pineapple production, Subang Indonesia

Variables	Regression coefficient	Standard error	t-ratio
Constant	-0.188	1.143	-0.164
Age (year)	0.012	-0.008	1.415
Education (year)	-0.039	0.044	-0.885
Farming experience (year)	-0.011	0.007	-1.399
Family member (people)	-0.022	0.069	-0.319
Off farm income (%)	-0.005 ^c	0.003	1.806
<i>Dummy</i> cropping pattern (1=intercropping, 0=monoculture)	0.982	0.914	1.074
<i>Dummy</i> land ownership (1=farmer’s own land, 0=rent)	-0.426 ^c	0.244	-1.743

Note: N = 142; a, b and c = Significant at $\alpha=0.01$, $\alpha=0.05$ and $\alpha=0.1$

CONCLUSIONS AND POLICY IMPLICATION

The pineapple production had been estimated by Cobb-Douglas production function and technical efficiency by Stochastic Frontier Analysis (SFA). The study revealed that inputs such as seed, hired labor, plant growth regulator, dung and chemical fertilizer have positive and significant influence on the yield of pineapple. Frontier production function analysis revealed that the average technical efficiency were 58.6 percent and hence output of pineapple can be increased by improving the technical efficiency of less efficient farms through suitable extension services delivery. The technical inefficiency is influenced by share of off-farm income and land’s ownership. In the long run, it needs attention at policy level to introduce other best alternative farming practices and improved technologies to alleviate the overall Subang pineapple shortage.

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MUTAGENIC ACTIVITY OF NUA POWDER EXTRACTS BY AMES TEST

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ABSTRACT

Most consumers believe that herbal products must be harmless. The herbs and spices were used as ingredient in seasoning powder to flavor food for a long time without concern for their toxicity and adverse effects. The objective of this study was to evaluate the mutation possibility of Thai savory powder, namely Nua powder. Nine vegetables are normally used in the seasoning powder recipe: mulberry (*Morus alba* Linn.), horse radish tree (*Moringa oleifera* Lam.), bamboo grass (*Tiliacora triandra* (Colebr.)Diels.), bustard cardamom (*Albizia myriophylla* Benth.), Chinese spinach (*Amaranthus lividus* Linn.), cowa (*Garcinia cowa* Roxb.ex DC.), chives (*Allium schoenoprasum* L.), Pak-wan tree (*Melientha suavis* Pierre.) and Kan tong (*Colubrina asiatica* L.ex Brongn.). The mutation evaluation was studied using the Ames test (reaction with and without nitrite salt). The experiments were conducted at the Institute of Food Research and Product Development (IFRPD) Kasetsart University from February 1 to 25, 2014. The samples were extracted using various solvents (dimethyl sulfoxide (DMSO) 80% methanol and hexane). After the reaction with /without nitrite salt, the extracts were further evaluated for mutation activity using *Salmonella typhimurium* strain TA98 and strain TA100 for detection of frameshift and base-pair substitution mutation, respectively. The extract was added at concentrations of 1.5, 7.5, 15 and 30 mg/plate into the plate culture to evaluate the correlation between the concentrations of the extracts and the number of bacterial colonies. The DMSO solvent and reaction without the nitrite salt system, did not exhibit any mutagenicity in the seasoning powder samples and all vegetables with either TA98 or TA100. In contrast, seasoning powder samples and four vegetables (horse radish tree, bamboo grass, chives and Kan tong) showed a weak positive mutation of *S. typhimurium* TA98. These results indicate that the extract from the four vegetables produced a direct mutagen after nitrosation. These four vegetables showed the same results, when 80% methanol was used as solvent. However, (bamboo grass and Kan tong hexane extract demonstrated weak mutagenic effect in the *S. typhimurium* colony after nitrosation, it can imply that the substance caused mutagenicity should be the polar compounds, because DMSO and methanol can extract polar substances but the hexane extract, did not show strong mutagenicity toward the bacterial colonies.

Key words: *S.typhimurium*, , nitrosation, Thai savory powder, frameshift mutation

INTRODUCTION

The utilization of herbs has been widely studied (in pharmaceutical products, food and beverage seasoning, cosmetics and supplementary applications). Seasoning powder has been widely used in Thai food recipes and is called Nua powder or Spicy powder. It is added into many types of food, such as mild soup, spicy lemon grass soup, spicy salad, fried food, curry, preserved food, and grilled food. This application is not only a part of conventional Thai philosophy, but it can also be used instead of conventional chemicals such as monosodium glutamate. This provides an alternative for customers who are allergic to monosodium glutamate, and who want to avoid monosodium glutamate in food recipes. Many people generally believed that herbal products are harmless. Unfortunately, there is no confirmed toxicology in food applications for this seasoning powder and the vegetables which are normally included in the seasoning powder. Some herbs or vegetables contain alkaloid compounds, for instance; *Ceylon cinnamon*. High consumption of these alkaloid compounds may result in high toxicology. According to Yu Yang and his colleagues (2014) reports, Chinese herbs containing aristolochic acid (AA) have been designated to be a strong carcinogen and associated with renal failure and urothelial carcinoma. Ungsurungsie *et al.*, (1984) reported the mutation of *Bacillus subtilis* after treating with herb extract (alkaloid compounds). In addition, this mutation could be explained by the nitrite or nitrate contained in the herbs or vegetables. In fact, nitrite and nitrate can come from many sources including green vegetables, tap water, fermented meat and human saliva.

Nitrite (and nitrite reduced from nitrate) can react with proteins (amine, amide, urea), to produce nitrosamine or nitroso compounds. Nitroso compounds are carcinogenic agents and can induce mutations 40 various species including humans (Bogovski,1981). Colorectal cancer is one of the cancers that could be affected by this nitroso compound (Loh *et al.*, 2011). Nitroso compounds comes not only from the reaction of nitrite and proteins, but also from the phenol compounds contained in vegetables. Jakszyn (2006) found that nitrosamine was contained in many kinds of food such as bacon, salty fish, dried shrimp, fermented fish, dried squid and some sauces. Therefore, nitrite and nitrate, nitrosamine or nitroso compound would have a strong effect on toxicology and mutation. Supatra *et al.* (2008) reported that extracts from some herbs (*Phyllanthus emblica* Linn. *Tinospora cordifolia* (Thunb.) Miers. *Acanthus ilicifolius* Linn. *Allium sativum* Linn. *Andrographis paniculata* (Burm.f.) Wall. Nees, *Murdania loriformis* (Hassk) Rao et Kammathy *Piper sarmentosum* Roxb. ex Hunter, *Zingiber officinale* Roscoe, *Senna siamea* (Lam.) Irwin & Barneby, *Azadirachta indica* A. Juss. var. *siamensis* Valetton, *Solanum trilobatum* L., *Solanum indicum* L., *Momordica charantia* L., *Alternanthera sessilis*(L.)DC., *Lonicera japonica* Thunb. *Cyperus rotundus* Linn.) could induce the reverse mutation in *Salmonella typhimurium* (TA 98). The nitrite or nitrate content in these samples may result in the mutation of *Salmonella typhimurium* (TA 98). For the same reason, the seasoning powder produced from local vegetables (as well as from the vegetables used as raw material in the seasoning powder) might contain nitrite or nitrate, which again may result in mutations in bacteria.

Therefore, the objective of this research work was to study the toxicology of this seasoning powder as well as of the vegetables which are normally used in it. The results obtained from this research can be used for choosing the type and amount of raw material to safely add to seasoning powder. The mutation evaluation was studied using the Ames test (reaction with and without nitrite salt). The experiments were conducted at Institute of Food Research and Product Development (IFRPD) Kasetsart University on October 1-30, 2014.

MATERIALS AND METHODS

Sampling, Seasoning powder preparation and extraction

Samples were collected from Ban Yang Lon Housekeeping Association in Sakon Nakhon on January 29, 2014. The experiments were conducted at the Institute of Food Research and Product Development (IFRPD) Kasetsart University, Bangkok from February 1 to 25, 2014.

According to the seasoning powder recipe of Ban Yang Lon Housekeeping Association in Sakon Nakhon, nine vegetable leaves mulberry (*Morus alba* Linn.), horse radish tree (*Moringa oleifera* Lam.), bamboo grass (*Tiliacora triandra* (Colebr.) Diels., bustard cardamom (*Albizia myriophylla* Benth.), Chinese spinach (*Amaranthus lividus* Linn.), cowa (*Garcinia cowa* Roxb.ex DC.), chives (*Allium schoenoprasum* L.), Pak-wan tree (*Melientha suavis* Pierre.) and Kan tong (*Colubrina asiatica* L.ex Brongn.) were prepared. The leaves were dried and blended to a fine powder.

A sample of 0.75 g was weighed and dissolved in 5 ml dimethyl sulfoxide (DMSO ,Sigma-Aldrich,USA, Analytical grade 99%). The final concentration of the solution was 0.15 g/ml. The solution was mixed for 15 min using a Vortex mixer before filtering through filter paper (Whatman No. 1) and the filtrate was kept at -20° C.

Extraction with methanol (Scharlau Chemicals,Bacelona,Spain, Analytical grade 99.8%) was done using a 30 g sample dissolved in 300 ml of 80% methanol (or 300 ml of hexane). The solution was mixed for 16 hrs in a shaker. The solution was filtered through filter paper (Whatman No.1). The solvent was evaporated using a rotary evaporator under low pressure at 45° C. The exact weight of the crude extract was recorded. The crude extract was then redissolved to a concentration of 0.1g/ml using DMSO and kept in a deep freezer at -20° C.

Microbial inoculation

Salmonella typhimurium (TA98 and TA100) was inoculated in nutrient broth according to the procedure developed by Kaew (1994). Each culture (10 µl) was pipetted into the flask containing the nutrient broth (No.2 , Oxoid Ltd,Basingtoke,UK, Analytical grade 99%). The flask was kept in an incubator at 37°C for 16 hrs.

Mutation evaluation of seasoning powder and each vegetable by Ames method (without nitrite salt addition)

The extracts (10, 50, 100, 200 µl) were added to 13x100 mm. Pyrex glass test tubes. Then, 500 µl of NaPO₄-KCl buffer and 100 µl of *Salmonella typhimurium* (TA98 or TA100) were also added. The test tubes were placed in a shaking water bath (Memmert Model SV 1422,Schwabach, Germany) at 37°C for 20 min. A sample of 2 ml of top agar (agar containing 10% 0.5M L-Histidine-HCl-0.5 mM biotin mixture) (Sigma-Aldrich Chemical Co,Poole,UK, Analytical grade 99%) was pipetted into each test tube and placed on a Vortex mixer for approximately 10 sec and afterwards was poured onto a culture plate. Each culture plate was incubated at 37° C for 48 hr after which, the number of mutation colonies was counted.

Mutation evaluation of the seasoning powder and vegetables (which are normally used in the seasoning powder) using Ames method (with nitrite salt addition)

Each extracts (10, 50, 100, 200 µl) was pipetted into a test tube and 750 µl of 0.2N HCl was added. The final volume was adjusted to 1,000 µl. The pH of the solution remained in the range 3.0-

3.5. Finally, 250 µl of 2M sodium nitrate (BDH Chemicals, Poole, England, Analytical grade 99%) was added to each test tube. Positive and negative controls were used. The positive control used 1-aminopyrine or 1-AP (the mutagen) at volumes of 10, 25, 50, 100 µl instead of the extracted samples and then followed the experiment as described above. The negative control used distilled water instead of the extracted samples. All test tubes were placed in a shaking water bath at 37°C for 3 hr. After that, the reaction was stopped by soaking in an ice box for 1 min and then adding 250 µl of 2M ammonium sulfamate. This was followed by further soaking of the test tubes for 10 min in the ice box. The nitrosation product was produced in this step, and then was readily used in the Ames method.

Ames test

S. typhimurium (TA 98) solution was incubated for 16 hrs before use. A sample of 100 µl of nitrosation product was mixed in test tubes with 500 µl of NaPO₄-KCl buffer (Scharlau Chemicals, Barcelona, Spain, Analytical grade 99.5%) and 100 µl of the incubated *S. typhimurium* (TA 98) was added. Finally, the mixture was placed in a shaking water bath for 20 min at 37° C. A sample of 100 ml of top agar was mixed with 10 ml of 0.5M L-Histidine/0.5 mM Biotin (Sigma-Aldrich Chemical Co, Poole, UK, Analytical grade 99%) and 2 ml of this solution was pipetted into each test tube. Each test tube was placed on a Vortex mixer for approximately 10 sec and then poured onto culture plates and incubated for 48 hr at 37° C. The number of mutation colonies was counted.

Data analysis

The concentrations of extracted samples were 1.5, 7.5, 15, and 30 mg/plate. The average number of mutant colonies obtained per plate was studied in four replications.

Principle of the experimentation

1) The extracted sample concentrations and the growth of colonies had to show a dose response relationship i.e., as the extracted sample concentration increased, the mutant colony increased.

2) The number of mutant colonies (affected by extracted sample) had to be higher than the number of mutant colonies (spontaneous mutation) in at least two extracted sample concentrations.

3) In addition, the number of mutations (affected by extracted sample) had to be twice as high as the spontaneous mutations in at least one extracted sample concentration. (Tejs, 2008)

RESULTS AND DISCUSSION

Tables 1 shows the mutation results of *S. typhimurium* (TA 98 and TA100). These results were obtained by treating the extracts (nine vegetables and seasoning powder samples extracted using DMSO) with *S. typhimurium* (TA98 and TA100). It was found that no extracted samples showed any mutation effect on *S. typhimurium* (TA 98 and TA 100).

However, after adding nitrite solution into the extracted solutions, Kan tong and Chives showed the mild mutation results. With a concentration value of 30 mg/plate, Horse radish tree and Bamboo grass showed stronger effects on mutagenicity (at 15 and 30 mg/ml) with only *S. typhimurium* (TA 98). It was noted that the mutation affected by these extracts was found to be the “frameshift mutation” type and was definitely not a “base-pair substitution mutation. This finding is similar to Thunyawan and Linna (2009)’s study, where it was discovered that the extract from some leaves of raw and blanched vegetables (Cha-Plu (*Piper sarmentosum* Roxb.), Yor (*Morinda citrifolia*

L.), Pak-Waan-Bann (*Sauropus androgynus* Merr.) and Pak-Khom (*Amaranthus lividus* Linn.) did not have mutagenicity without nitrite reaction, however after being reacted with nitrite solution in an acidic condition (pH 3-3.5) without activating system, they showed the mutagenicity toward both types of *S. typhimurium* (TA 98 and TA 100). This may be because fertilizers or insecticides residue can be accumulated in those vegetables. The nitrate is normally composed in the fertilizers or insecticides. When humans consume nitrate in those vegetables, nitrate is readily reduced to nitrite by oral bacteria. The nitrite then reacted with secondary amines (from some preserved foods such as sausages, bacon) and it generates a mutagenic nitrosamines. Some leaf vegetables were a rich source of polyphenols and flavonoids. Bamishaiye and colleague (2011) reported that they could detect the phytochemicals such as alkaloids, tannins, phenolics, saponins, flavonoids, and steroids in both aqueous and methanolic extracts of *Moringa oleifera* leaves maturation. It is possible that the mutagenic activity might be from polyphenolic or flavonoids compounds. Wakabayashi and colleague (1989) reported that some phenol and indole derivatives present in some vegetables are changed to mutagenic compounds by being reacted with nitrite (nitrosation). This is similar to the study of Kikugawa and Kato (1988), where it was discovered that the diazaquinone compounds, which show mutagenicity, were formed by the interaction between phenol and nitrite.

According to Flavia *et al.* (2012), some flavonoids have mutagenic activity; as is the case for quercetin, kaempferol and galangin. In the Ames test, those flavonoids were mutagenic toward the TA98 strain. This is similar to the experiment of Macgregor (1986), where it was discovered that quercetin was capable of causing base-pair substitutions and frameshift mutations in the Ames assay. According to Santos and colleague (2011)'s research, where it was discovered that the extracts obtained from *Qualea multiflora* and *Qualea grandiflora* exhibited mutagenicity, inducing frameshift mutations and base substitutions in DNA. The bacterial mutagenicity might be due to the presence of pentacyclic triterpenes and polyphenols, which are capable to form reactive oxygen species (ROS) and result in the liable to cause DNA damage.

In addition, Tables 1 shows that the seasoning powder did not produce any mutation effect on these bacteria. Four vegetables in the seasoning powder recipe—Kan Tong, chives, horse radish tree and bamboo grass—showed mutagenicity. The reason for the negative mutation result might have resulted from the very low concentration of these vegetables in the seasoning powder (compared to the other vegetables). Moreover, the chemical interaction of active agents and other chemical components of seasoning powder may affect this negative mutation results such as disappearance of amine, amide and urea and/or nitrite reaction to nitroso compound. According to Charoensin and Wongpoomchai (2010), who reported that *Moringa oleifera* (one of the vegetables which is normally a component in the seasoning powder recipe) showed non-mutagenic in both *S. typhimurium* TA98 and TA100 and was capable of antimutagenicity against the standard mutagen, acrylamide (AF-2). Some Chinese herbals remedies are generally prescribed as complex mixtures of several different medicinal plants so that the combination of these can enhance the main effect and reduce the toxicity of herbs (Yu Yang *et al.*, 2014).

Based on Prapasri's (1994) research, it was observed that some vegetables and fruits fiber have ability to absorb nitrite and also diminish the mutagenicity and mutagen formation of nitrite-treated sample extracts. The crosslinking of polysaccharides by lignin in those vegetables and fruits makes it possible for the plant to absorb the nitroso compounds, and result in the potential to diminish the bacterial mutagenicity (Tongyonk *et al.*, 2003).

Mutagenic activity of Nua powder extracts

Table 1. Mutation evaluation of *S. typhimurium* (TA98) and (TA100) using seasoning power and vegetable extracts as mutagen and using DMSO as the solvent

Sample	Concentration (mg/plate)	No. of mutation colony ^a /plate			
		TA98		TA100	
		Without NO ₂	with NO ₂	without NO ₂	with NO ₂
Kan Tong	1.5	13±4	19±6	82±8	102±19
	7.5	14±3	18±3	81±7	96±14
	15	17±3	35±2	81±3	133±7
	30	12±4	44±4	91±4	187±9
Pak -wan tree	1.5	15±3	18±6	86±3	123±23
	7.5	14±5	21±8	118±9	166±10
	15	13±6	28±2	129±6	175±10
	30	15±2	27±13	147±4	169±15
Chives	1.5	17±7	18±6	89±5	100±17
	7.5	13±3	31±5	92±20	96±14
	15	15±3	33±7	96±17	133±7
	30	16±4	45±5	85±4	187±9
Chinese spinach	1.5	15±5	13±2	95±18	83±5
	7.5	12±4	20±5	89±19	92±4
	15	14±7	16±4	81±14	96±11
	30	12±2	21±3	75±13	74±17
Cowa	1.5	18±4	18±7	91±30	91±10
	7.5	20±2	20±4	93±9	111±22
	15	13±3	20±5	93±9	126±12
	30	16±4	26±7	89±17	132±25
Bustard cardamom	1.5	21±4	18±2	73±11	85±20
	7.5	20±7	20±3	76±8	89±3
	15	20±6	17±3	82±5	84±4
	30	21±6	15±3	88±7	80±5
Horse radish tree	1.5	17±2	18±6	85±2	100±8
	7.5	14±2	20±6	95±3	93±14
	15	16±3	60±9	130±8	125±9
	30	15±3	100±21	152±10	130±18
Bamboo grass	1.5	21±5	18±3	56±33	76±9
	7.5	14±2	39±3	65±2	78±13
	15	16±3	91±11	73±3	94±8
	30	16±5	133±22	81±2	103±17

Sample	Concentration (mg/plate)	No. of mutation colony ^a /plate			
		TA98		TA100	
		Without NO ₂	with NO ₂	without NO ₂	with NO ₂
Mulberry	1.5	17±3	19±5	85±4	119±9
	7.5	14±5	25±5	103±2	163±8
	15	13±6	31±9	113±6	174±11
	30	14±2	34±7	120±3	181±25
Seasoning powder	1.5	16±5	12±3	75±8	78±6
	7.5	12±3	20±3	65±4	69±11
	15	15±4	26±5	85±5	102±11
	30	15±2	21±8	85±3	159±14
Negative control	-	22±2	18±3	120±23	121±14
Positive control ^b	-	21±4	582±54	86±8	1067±126

: a: mean(from 4 replicates) ± SD b: 1-Aminopyrene 7.5 µg/plate

The number of mutation colony in bold type: number of revertants doubles the spontaneous yields.

Table 2 shows the mutation evaluation using 80% methanol solvent and *S. typhimurium* (TA 98). These four samples posed mutation of *S. typhimurium* (TA 98) with the addition of nitrite. For the condition of without nitrite addition, mutation effects were not observed.

Table 2. Mutation evaluation of *S. typhimurium* (TA98) using vegetable methanol extracts as mutagen.

Sample	Concentration (mg/plate)	No. of mutation colony ^a /plate	
		without NO ₂	with NO ₂
Kan Tong	1.5	28 ± 1	32 ± 0
	7.5	37 ± 9	46 ± 2
	15	30 ± 8	49 ± 8
	30	22 ± 3	61 ± 4
Chives	1.5	32 ± 1	47 ± 1
	7.5	41 ± 1	56 ± 6
	15	30 ± 18	69 ± 9
	30	27 ± 0	60 ± 7
Horse radish tree	1.5	39 ± 4	31 ± 5
	7.5	33 ± 1	33 ± 4
	15	25 ± 5	33 ± 1
	30	20 ± 2	57 ± 5
Bamboo grass	1.5	20 ± 4	37 ± 2
	7.5	24 ± 1	48 ± 8
	15	18 ± 0	44 ± 2
	30	24 ± 1	71 ± 5
Negative control	-	25 ± 3	21 ± 2
Positive control ^b	-	19 ± 6	613 ± 25

a: mean (from 4 replicates) ± SD. b: 1-Aminopyrene 7.5 µg/plate

The number of mutation colony in bold type: number of revertants doubles the spontaneous yields.

Table 3 shows the mutation evaluation using hexane as solvent and *S. typhimurium* (TA98). Only two samples (Kan tong and Bamboo grass) produced mutation of *S. typhimurium* (TA98) with the addition of nitrite. Undoubtedly, mutation effects were not found in the system without nitrite addition. From the experimental results, it was found that the extracts derived by different solvents had different effects on mutagenicity of bacterial colonies. That is, the extracts of four vegetables obtained by DMSO and methanol have strong mutagenicity in *Salmonella* sp. but the extracts from hexane, only two vegetables showed mild mutagenicity. Solvents with different polarities may contribute to this finding as different substances were extracted. This is because, the extract from both solvent (DMSO and methanol) gave the polarity substances. These substances may have the effect on mutagenicity of the bacterial colonies. Consequently, the polar substances in the methanol extract may cause those four vegetables to have stronger mutagenicity than the hexane extract. It can imply that the polar substances caused mutagenicity, because DMSO and methanol can extract the polar substances but the hexane extract, which is non polar, did not show strong mutagenicity toward the bacterial colonies. This study is supported by the Pederson and Siak's experiment (1981), where it was observed that about a third of the bacterial mutagenicity in the diesel particle extract was recovered in fractions containing monosubstituted nitro-PAH compounds. The remaining activity was in the more polar fractions.

However, the mutation effect from the seasoning powder was found to be of low toxicity. The seasoning powder in commercial noodles has high mutation possibility, which is more than that of seasoning powder produced from vegetable. Oranuch, (2001) reported that seasoning powder of commercial noodle posed high mutation ability to *S. typhimurium* (TA98). Its mutation colony at concentration of seasoning powder of 2.4-9.8 mg/plate was found to be 42-243 colony/plate. Meanwhile the mutation colony of *S. typhimurium* (TA100) at seasoning powder concentration of 2.9-11.7 mg/plate was found to be 380-1,013 mg/plate. It means, the mutation possibilities found with TA 98 and TA 100 were higher than that of natural conditions (2-12 times and 4-11 times, respectively).

Table 3. Mutation evaluation of *S. typhimurium* (TA98) using vegetable hexane extracts as mutagen.

Sample	Concentration (mg/plate)	No. mutation colony ^a /plate	
		without NO ₂	with NO ₂
Kan Tong	1.5	18 ± 1	36 ± 0
	7.5	22 ± 5	21 ± 3
	15	25 ± 5	20 ± 4
	30	31 ± 3	52 ± 7
Chives	1.5	25 ± 6	17 ± 4
	7.5	30 ± 1	23 ± 5
	15	32 ± 4	33 ± 2
	30	24 ± 2	33 ± 1
Horse radish tree	1.5	23 ± 6	24 ± 1
	7.5	30 ± 2	29 ± 3
	15	32 ± 4	38 ± 1
	30	28 ± 1	33 ± 0
Bamboo grass	1.5	19 ± 5	27 ± 7
	7.5	30 ± 1	24 ± 5
	15	26 ± 1	50 ± 4
	30	26 ± 4	60 ± 6
Negative control	-	25 ± 3	21 ± 2
Positive control ^b	-	19 ± 6	613 ± 25

a: mean (from 4 replicates) ± SD b: 1-Aminopyrene 7.5 µg/plate

The number of mutation colony in bold type: number of revertants doubles the spontaneous yields.

Therefore, the seasoning powder produced from natural vegetable as well as from monosodium glutamate can both induce the mutation possibility, with the addition of nitrite. However, the one that is produced from vegetables has lower mutation possibility. Thus, this valuable data and information from this research work would be useful for the seasoning powder producers. They can choose the type and amount (recipe ratio) of vegetables for their production line, and then maintain the good quality and high safety of seasoning powder for consumers.

CONCLUSION

The Nua seasoning powder did not show any mutagenic activity. On the other hand, the extracts from vegetables (which are normally components of the seasoning powder) were not found to be “direct mutagens”. That is, in the condition of without nitrite salt reaction, *S. typhimurium* (TA98 and TA100) did not produce mutation colonies. Meanwhile, the Kan Tong, chives, horse radish tree and bamboo grass extracts with nitrite reaction were found to be mutagenic to *S. typhimurium* (TA98). Therefore, these extracts were only frameshift mutagens. They did not induce the base pair substitution mutation. We can conclude that some Nua powder ingredients might have mutagenic activities.

The methanol extracts obtained from Kan Tong, chives, horse radish tree and bamboo grass, produced mutation colonies of *S. typhimurium* (TA 98) after reaction with nitrite salt. Meanwhile, hexane extracts of Kan Tong and bamboo grass produced mutation colonies. Without nitrite addition, all four samples did not produce mutation colonies.

Our results suggest that some herbs and spice components of Nua powder exhibit mutagenic activities although the Nua powder by itself, did not exhibit mutagenic effects. Nua powder contained only trace amounts of these mutagenic herbs and some herbs, such as mulberry (Agabeyli, 2012) and horse radish tree (Charoensin and Wongpoomchai, 2010) were capable of antimutagenic activities. Thus, Nua powder can be considered genotoxically safe and should be used as flavor enhancers.

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APPLICATION OF *Trichoderma asperellum* FRESH CULTURE BIOPRODUCT AS POTENTIAL BIOLOGICAL CONTROL AGENT OF FUNGAL DISEASES TO INCREASE YIELD OF RICE (*Oryza sativa* L.)

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ABSTRACT

Dirty panicle, brown spot, narrow brown spot and sheath rot are important constraints for rice production in Thailand. This research sought to evaluate the efficacy of five isolates of *Trichoderma asperellum* to reduce these fungal diseases and to increase rice yield. Experiments were conducted at Kasetsart University, Kamphaeng Saen Campus, Nakhon Pathom, Thailand from June to September, 2014. Rice seeds var. Chai Nat 1 were soaked (Sk) for 24 hr in spore suspension derived from *Trichoderma* fresh culture bioproduct (semi-cooked broken milled rice colonized with fungus) at 100 g 20 L⁻¹. Soaked rice seeds were planted in cement circular wells. Rice plants were sprayed (Sp) with spore suspension (100 g-fresh culture 20 L⁻¹) three times during the growing period. The results revealed that *T. asperellum* isolate 01-52 and CB-Pin-01 increased plant height, number of panicles per square meter, 1,000-seed weight and yield. Dirty panicle infected seeds, brown spot and narrow brown spot were reduced when compared to untreated control. The highest percentage of rice roots colonized by *T. asperellum* was obtained from treatment 01-52 (Sk+Sp). Fresh culture bioproducts of *T. asperellum* 01-52 and CB-Pin-01 used for seed soaking and plant spraying have the potential to reduce several important rice fungal diseases and to enhance rice yield.

Key words: isolates, rice diseases, biofungicide, antagonistic fungus

INTRODUCTION

Rice (*Oryza sativa* L.) is very important cereal crop grown for consumption and export in Asia. Total rice yield from 17 countries of Asia during 2014-2015 was 424.12 million tons or 89.16% of world total yield. In 2014, 12 countries in Asia used 398.514 million tons of rice for consumption and utilization, which was 82.24% of the world usage (Original data from Foreign Agricultural Service and World Markets & Trade, Ministry of Agriculture, USA, May 2015, cited by Rice Department, 2015). Thailand was ranked as No.1 rice exporter of the world market in 2014. Rice exporting in Thailand provides the main income for our country. During the year 2014 (January - December), total milled rice exported from Thailand was 10.97 million tons with 5,439 million US\$ of exporting value (Thai Rice Exporters Association, 2015). The production of rice has several diseases such as dirty panicle or seed discoloration, brown spot, narrow brown spot and sheath rot which affect on rice yield. These diseases can cause great economic losses in grain and seed production which makes it unacceptable for consumption and seeding. Discoloration seeds with brown spot caused by fungi *Bipolaris oryzae* and *Curvularia oryzae* gave 41% abnormal seedlings

and 33% seed rot. The infected seeds may fail to germinate, transmit diseases from seeds to seedlings and from seedlings to growing plants (Ibrahim and Abo El-Dahab, 2014). Under favorable conditions of high temperature and humidity, high disease severity not only reduced 1,000-grain weight (16.61%) and the percentage of rice seed germination (34.31%) but also reduced the quality of brown rice and milled rice by decreasing head rice percentage of Jasmine 85 variety by 40.17% at discoloration disease level of 51-100% (Phat *et al.*, 2005). Disease severities of dirty panicle varied from 6.0-87.3% on different rice varieties. High disease severity caused as high as 72% yield loss in rice variety RD31 (Chimwai *et al.*, 2013)

In Thailand, dirty panicle disease is caused by at least six phytopathogenic fungi including *Curvularia lunata* (Wakk) Boedjin., *Bipolaris (Helminthosporium) oryzae* (Breda de Haan) Shoem., *Alternaria (Trichoconis) padwickii* (Ganguly) M.B. Ellis., *Cercospora oryzae* I. Miyake., *Fusarium incarnatum* (Roberge) Sacc. (*Fusarium semitectum* Berk & Rav.) and *Sarocladium oryzae* Sawada. Particularly, *B. oryzae* is highly pathogenic to seeds and seedlings, causing seed rot, root rot and reducing quality and quantity of rice seedlings (Bureau of Rice Research and Development, 2015; Chettanachit *et al.*, 2009; Jaisong, 2010). The favorable conditions for fungal infection are high temperature (28-33 °C) and humidity (>80% RH). Occurrence of such conditions during the panicle forming stage and milky stage usually causes severe damages on rice seeds. *B. oryzae* and *A. padwickii* were detected at high level, 19.2 and 14.4% seed infection, respectively (Jaisong, 2010). Some dirty panicle pathogens can cause diseases on rice leaves and seedlings including brown spot (*B. oryzae*), narrow brown spot (*C. oryzae*), sheath rot (*S. oryzae*) and seedling rot (*C. lunata* and *B. oryzae*) in nursery trays (Bureau of Rice Research and Development, 2015; Chettanachit *et al.*, 2009). These pathogens are widely disseminated by wind and are seed borne fungi.

At present, rice farmers have used several commercial available chemical fungicides to control various leaf and dirty panicle diseases because the commercial biofungicides are not readily available. However, many consumers are becoming increasingly concern about chemical contamination and residue accumulation in the environment and many other problems such as pesticide residues on food and pathogen resistance to chemical fungicides. Furthermore, the application of chemical fungicides to control rice diseases increases the cost of rice production. Thus, biological control (biocontrol) is an alternative method for the control of plant diseases. The most widely used biocontrol agent (BCA) throughout the world belongs to the genus *Trichoderma* (Harman, 2011a, 2011b). *Trichoderma* has been well-known BCA which provides high efficacy to control fungal pathogens directly by antibiosis and mycoparasitism. However, abilities to induce resistance to plant disease and increase plant growth are indirect benefits (Brunner *et al.*, 2003; Harman, 2011a, 2011b; Harman and Shores, 2007 and Saldajeno *et al.*, 2014). *T. asperellum* SKT-1 (SKT-1) is a valuable fungal BCA against various seed-borne diseases of rice including “Bakanae” disease (Watanabe *et al.*, 2007). Mixture of four isolates of *T. asperellum* was efficient in reducing severity of sheath blight and increasing rice yield and grain weight in Brazil (de França *et al.*, 2015).

The researches on *Trichoderma* spp. for plant disease control in Thailand have been started since 1985 (Inwang and Chamswarn, 1986). *T. asperellum* Samuels, Lieckf. & Nirenberg isolate CB-Pin-01 (formerly identified as *T. harzianum*) as shown in Fig. 1 was isolated from pineapple field soil in Chonburi Province at the Eastern part of a country. This isolate provided the most promising efficacy for the control of fungal plant diseases (Chamswarn and Tanangsnakool, 1996). During 1986 to 2010, there were reports which proving that *T. asperellum* CB-Pin-01 was the most effective antagonistic BCA for controlling various diseases caused by fungi and promoting plant growth of many kinds of crops, including cereal, fruit, vegetable, ornamental and hydroponic lettuces (Chamswarn and Intano, 2002; Chamswarn *et al.*, 1992; Intana *et al.*, 2003; Lamool, 2006; Thieankhao *et al.*, 2009; Thonkla *et al.*, 2007). Biocontrol mechanisms of *T. asperellum* CB-Pin-01 including competition, parasitism, antibiosis and induced systemic resistance in plants were determined (Intana *et al.*, 2003). Ability of *T. asperellum* CB-Pin-01 and 01-52 (formerly identified as

T. harzianum) to colonize rice roots has been found to induce resistance in rice plant against dirty panicle and brown spot (Charoenrak *et al.*, 2012). Promotion of plant growth was attributed to phosphate solubilization and the production of auxins as well as other plant growth promoting-like substances such as pentyl pyrone, harzianic acid and harzianic acid isomer (Intana *et al.*, 2003; Promwee *et al.*, 2014). Since 2002, *T. asperellum* CB-Pin-01 has been distributed as pure stock culture in powder bioformulation used for producing fresh culture bioproduct (semi-cooked broken milled rice colonized with fungus) by simple procedure developed by Chamswarnng and Intanoo (2002). Spore suspension prepared from fresh culture bioproduct was widely used for seed soaking and plant spraying against fungal diseases of various plants except rice during 2002 to 2005. Among the other *T. asperellum* isolates, 01-52, T50, M23 and T35-Co4 (formerly identified as *T. harzianum*) used in this study the latter three isolates have never been tested for the control of rice diseases. While isolate 01-52 prepared as powder and pellet formulations were evaluated for the efficacy to control rice dirty panicle, brown spot, narrow brown spot, sheath rot and sheath blight (Chamswarnng *et al.*, 2012a, 2012b, 2014).

Most of rice production in Thailand is in lowland or water flooding areas. The control of rice diseases by coating rice seeds with *Trichoderma* spp. or applying spore suspension through irrigation system was early thought to be impossible since *Trichoderma* isolates are originally isolated from soil which may not survive in water. In addition, the use of *T. asperellum* as a BCA for the control of dirty panicle and several diseases of rice has not widely been reported. Nevertheless, Lamool (2006) demonstrated that *T. asperellum* CB-Pin-01 could colonize and survive on root surface of lettuce (Cos) grown in the Nutrient Film Technique (NFT) hydroponic system, effectively reduced *Pythium* root rot incidence and promoted lettuce growth. Therefore, the research on the application of *T. asperellum* CB-Pin-01 against rice diseases and enhancement of rice growth and yield was initiated (Chamswarnng and Intanoo, 2007; Chamswarnng *et al.*, 2005). The initial results revealed that *T. asperellum* CB-Pin-01 and 01-52 had promising potential to reduce *Rhizoctonia* sheath blight, brown spot, narrow brown sot, sheath rot and dirty panicle diseases, enhance seedling and plant growth, increase yield and was able to colonize the roots of rice (Chamswarnng *et al.*, 2012a, 2012b, 2012c, 2014). Other three isolates of *T. asperellum* have never been evaluated for the antagonistic effects against rice diseases as well as the ability to promote plant growth and increase rice yield.

Therefore, this research was conducted to; (1) evaluate the efficacy of five antagonistic *T. asperellum* isolates prepared as fresh culture bioproduct as potential BCA of rice diseases including dirty panicle, brown spot, narrow brown spot, sheath rot and (2) assess of the growth promotion and yield enhancement effect of *T. asperellum* on rice.

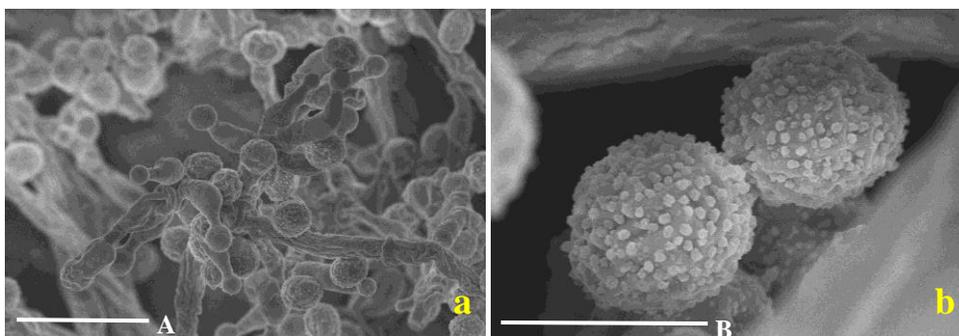


Fig. 1. Morphological characteristics of *Trichoderma asperellum* (CB-Pin-01) (a) conidiophores and young conidia, (b) conidial surface with pyramidal warts. Bars: A 10 μ m, B 3 μ m (courtesy J. Unartngam)

MATERIALS AND METHODS

Trichoderma asperellum isolates

All *T. asperellum* isolates (Molecular taxonomic based; Unartngam, personal communication) used in this study were received from Assoc. Prof. Dr. Chiradej Chamswarnng of Plant Disease Biocontrol Laboratory, Department of Agriculture at Kamphaeng Saen, Kasetsart University, Kamphaeng Saen Campus, Nakhon Pathom Province, Thailand. *Trichoderma* isolates M23 (Gesnara, 1994), 01-52 (Chamswarnng and Intanoo, 2007), T50 and T35-Co4 (Intana *et al.*, 2003), and CB-Pin-01 (Inwang and Chamswarnng, 1986) were formerly identified as *T. harzianum* based on fungal morphology and culture characteristics. The four *Trichoderma* isolates 01-52, T50, M23 and T35-Co4 effectively inhibited dirty panicle pathogens (*B. oryzae*, *C. lunata* and *A. padwickii*) *in vitro*, promoted rice seedling growth, and colonized rice seedling roots (Charoenrak *et al.*, 2009). *T. asperellum* isolate CB-Pin-01 (a parent strain) was used as a comparative isolate. Details of origin and efficacy of these *T. asperellum* isolates were described in Table 1.

Table 1. Original sources and efficacy of *Trichoderma* isolates in Thailand.

<i>Trichoderma asperellum</i> Isolate	Source	Efficacy
CB-Pin-01 (Fig. 1)	Isolated from pineapple field soil, Chonburi Province (Inwang and Chamswarnng, 1986)	An indigenous parent strain which control root and stem rot of durian and tangerine; stem rot of tomato and chili; damping-off of cotton, vegetable soybean, yard long bean and cucumber; seedling blight of barley; anthracnose of chili, asparagus, mango, grape, root rot of hydroponic lettuce; dirty panicle, brown spot, sheath blight and seedling rot of rice <i>etc.</i>
T50	Isolated from durian orchard soil, Songkla Province (Intana <i>et al.</i> , 2003)	Promote growth and induce resistance in cucumber and tomato against root rot caused by <i>Pythium irregulare</i> , <i>P. aphanidermatum</i> and <i>Pseudocercospora fuligena</i>
T35-Co4	Benomyl resistant mutant strain derived from isolate T35, isolated from durian orchard soil, Surat Thani Province (Intana <i>et al.</i> , 2003)	Produce pentyl pyrone and oxazole which promote growth and colonize roots of cucumber
01-52	Benomyl resistant mutant strain derived from isolate CB-Pin-01 (Chamswarnng and Intanoo, 2007)	Reduce dirty panicle, brown spot, sheath rot, sheath blight, seedling rot and enhance yield of rice
M23	Benomyl resistant mutant strain derived from isolate CB-Pin-01(Gesnara, 1994)	Reduce stem rot of tomato; seedling blight of barley caused by <i>Sclerotium rolfsii</i> ; reduce root rot of durian caused by <i>Phytophthora palmivora</i> and root rot of tangerine caused by <i>Phytophthora parasitica</i>

Preparation of *Trichoderma* spore suspension

Isolates of *T. asperellum* were cultured on semi-cooked broken milled rice as fresh culture bioproduct following the procedure of Chamswang and Intanoo (2002). Briefly, four parts of broken milled rice or milled rice (var. Khao Dawk Mali 105) and two parts of tap water (v/v) were added into automatic rice cooker. After 10-15 min, switch of rice cooker turned off, 250 g of semi-cooked broken milled rice was taken and put into heat tolerant plastic bag (8x12 inch), then rice seeds were flatten and the open-end of the bag was folded during cooling down period. Aliquot (0.25-0.5 g-powder) of pure stock culture of each *Trichoderma* isolate prepared as powder bioformulation (unpublished data) was added into the nearly cold semi-cooked broken milled rice, then sealed with plastic sealer. Each bag was punched in the area under sealed line with fine needle for 20 to 30 times. Rice seeds in each inoculated bag was flatten, then upper part of bag was lifted to bring air into the bag. All inoculated bags were incubated at 25-30 °C for 6-7 days (d). Spore suspension was prepared by mixing 100 g fresh culture with 1 L of cleaned water. Dark greenish *Trichoderma* spores covered on whole surface of semi-cooked broken milled rice (Fig. 2) were washed from rice seeds by stirring fresh culture-water mix for 2-3 minutes (min). The resulting spore suspension was filtered through cheesecloth or 50-100 mesh screen before added with cleaned water to obtain 20 L spore suspension. The final concentration of spore suspension was 10^6 CFU ml⁻¹.



Fig. 2. Fresh culture bioproduct of *Trichoderma asperellum* CB-Pin-01 (a,b) showing white mycelial growth on semi-cooked broken milled rice after 2 d incubation period, (c,d) whole surface of semi-cooked broken milled rice after 7 d incubation.

Rice seed soaking and plant spraying with *Trichoderma* isolates

Ten grams of rice seeds (paddy) var. Chai Nat 1 were placed in double layers of cheesecloth and soaked in spore suspension of *T. asperellum* (100 g 20 L⁻¹ of water) for 24 hr. After spore suspension was drained off, seeds were kept moist and incubated for 15-18 hr. Germinated rice seeds were planted in cement circular well (80 cm in diameter and 40 cm in height) contained with clay soil, five

seeds per sowing site and five sites for each well. At 14 days after sowing (DAS), only one rice seedling was maintained at each site. The whole rice plants were sprayed with each *Trichoderma* isolate for three times at the booting stage (50 DAS), 5% of panicle formation stage (70 DAS) and complete panicle formation stage (90 DAS).

Experimental Design

The Randomized Complete Block Design (RCBD) was performed with nine treatments, four replications (one cement circular well per replication) for each treatment and five plants per replication. Five treatments comprised of rice seed soaking (Sk) and whole plant spraying (Sp) with each isolate of *T. asperellum*, i.e. Treatment 1 (T1)-SK and Sp with *T. asperellum* CB-Pin-01; T2-Sk and Sp with *T. asperellum* 01-52; T3-Sk and Sp with *T. asperellum* T50; T4-Sk and Sp with *T. asperellum* M23 and T5-Sk and Sp with *T. asperellum* T35-Co4. Two treatments were rice seeds soaked only with each of two *T. asperellum* isolates, i.e. T6-Sk with *T. asperellum* CB-Pin-01 and T7-Sk with *T. asperellum* 01-52. The other two treatments were rice seeds soaked with cleaned water served as a positive control; i.e. T8-Sk with water and the whole plants sprayed with chemical fungicide was the negative control i.e. T9-Sp with propiconazole + difenoconazole 30% W/V EC. For the management of rice plants, each well was applied with chemical fertilizers as recommended by the Rice Department. Two chemical fertilizer formulations were applied including 16-20-0 (156.25 kg ha⁻¹), 46-0-0 (125.00 kg ha⁻¹), and 46-0-0 (62.50 kg ha⁻¹) at 15, 45 and 55 DAS, respectively.

Dirty panicle pathogens inoculation

The dirty panicle infected rice seeds were surface sterilized with 0.525% sodium hypochlorite for 2-3 min, blotted dry and placed on potato dextrose agar (PDA) with five seeds per plate. All plates were incubated at room temperature (25-30 °C) until fungal mycelia grew out from the rice seeds. Several pieces of agar at the margin of fungal colonies were cut and transferred to potato dextrose agar (PDA). All pure cultures of *B. oryzae*, *C. lunata* and *A. padwickii* were stored at 10 °C until used. Each pathogen was grown on autoclaved rice seeds contained in plastic bag and incubated under near ultraviolet (NUV) light for 10 d or until the spores were observed (modified from Chamswarnng and Intanoo, 2002). Three dirty panicle pathogens were prepared separately as spore suspension and numbers of total spores in the mixture of three pathogens (1:1:1 by volume) were standardized at 10⁴ spore ml⁻¹ by using haemocytometer. The test plants were inoculated by spraying spore suspension on the whole rice plants at the early stage of panicle formation.

Data gathering

Rice growth, disease incidences and yield components of each treatment were recorded from 108 d-old rice plants in four replications (wells). Rice growth was determined by measuring plant height from base of tiller to terminal of flag leaf. Number of panicle per square meter was counted from five hills of each replication, four replications per treatment. Severity of brown spot and narrow brown spot diseases was assessed from five flag leaves of each replication and four replications per treatment (modified form Chettanachit *et al.*, 2009). While severity of sheath rot was enumerated by counting number of rotted sheath or sheath rot infected panicles from total plants in five hills collected from each replication. The severity of dirty panicle disease was determined as the percentage of healthy seeds, dirty panicle infected seeds and empty seeds (un-fertile seeds, undeveloped seeds) derived from 5 g of seeds in each replication and four replications per treatment. For the yield assimilation, rice panicles were harvested from all plants in each of four wells (replications). The moisture of rice seeds was reduced to 15% before all seeds were detached from panicles for the evaluation of total rice yield and weight of 1,000-randomly collected seeds. The percentage of rice root colonized with *T. asperellum* was detected by collecting the rice root systems from harvested plants of each replication. The roots were washed three times under running tap water, cut to a single piece, blotted dry in

sterilized tissue paper and placed on Martin's medium (Johnson and Curl, 1972) supplemented with 100 ppm Rifampicin (Pond's Chemical Thailand R.O.P). After incubation under light at room temperature (25-30 °C) for 3-5 d, the numbers of rice roots colonized with *T. asperellum* were recorded by observing mycelial growth and sporulation of *T. asperellum* from the root samples.

Statistical analyses

All data were statistical analysis using analysis of variance (ANOVA). The significance of difference between the treatment means was determined by Least Significant Difference (LSD) using the statistical programs R (R Core Team, 2014). The significance level was set at $P \leq 0.05$.

RESULTS AND DISCUSSION

Rice plant growth and yield

All *Trichoderma* treatments either rice seed soaking (Sk) or seed soaking combined with plant spraying (Sp) with *T. asperellum* except T50 (Sk+Sp) enhanced the height of rice plants by 1.23-4.77% when compared with untreated control. Treatments with seed soaking and spraying (Sk+Sp) with isolate CB-Pin-01 and only Sk with isolate 01-52 provided significant plant heights with 135.30 cm and 136.20 cm, respectively as compared to the control (130.00 cm). There were no significant differences in the panicle numbers per square meters among all *Trichoderma* treatments except treatments M23 (Sk+Sp) and CB-Pin-01 (Sk) when compared with untreated control. However, seed soaking and plant spraying (Sk+Sp) with isolates 01-52 and T50 increased panicle numbers by 7.38 and 7.78%, respectively when compared to untreated control. While other isolates failed to increase the panicle number relative to the control (Table 2). Results obtained from Table 2 indicated certain effects of either soaking or seed soaking combined with plant spraying on rice height, but less effects on panicle number per square meter. This was probably due to the differences in efficacies among the isolates of *T. asperellum*.

Table 2. Plant height and number of panicles per square meter (m^2) of rice plant (var. Chai Nat 1) grown with seeds soaked only (Sk) or seeds soaked and plants sprayed (Sk+Sp) with fresh culture bioproducts of various *Trichoderma asperellum* isolates.

Treatment	Height (cm)	Panicle number per m^2
CB-Pin-01 (Sk ¹ +Sp ²)	135.30 ab ^{3/} +4.08%) ^{4/}	78.62 ab (-0.35%) ^{4/}
01-52 (Sk+Sp)	131.80 abc (+1.38%)	84.72 a (+7.38%)
T50 (Sk+Sp)	130.00 c (+0%)	85.04 a (+7.78%)
M23 (Sk+Sp)	132.10 abc (+1.61%)	67.20 c (-14.83%)
T35-Co4 (Sk+Sp)	131.60 bc (+1.23%)	76.20 b (-3.42%)
CB-Pin-01 (Sk)	133.70 abc (+2.85%)	64.80 c (-17.87%)
01-52 (Sk)	136.20 a (+4.77%)	77.92 ab (-1.24%)
propiconazole + difenoconazole (Sp)	131.80 abc (+1.38%)	78.40 ab (-0.63%)
Control untreated	130.00 c -	78.90 ab -

^{1/} Sk = Soaking rice seeds in *Trichoderma* spore suspension prepared from *Trichoderma* fresh culture bioproduct (100 g 20 L⁻¹ of water).

^{2/} Sp = Spraying on the whole rice plants with *Trichoderma* spore suspension prepared from *Trichoderma* fresh culture bioproduct (100 g 20 L⁻¹ of water).

^{3/} Means in each column followed by the same letter (s) are not significantly different according to Least Significant Difference (LSD) ($P \leq 0.05$).

^{4/} Percentage of increment (+) or decrement (-) of each treatment mean when compared with untreated control.

Results from Table 3 showed that soaking rice seeds and spraying plants (Sk+Sp) with isolates T35-Co4 and CB-Pin-01 and soaking seeds with isolates CB-Pin-01 and 01-52 significantly increased seed weight per panicle by 34.76, 17.60 and 21.46, 17.17%, respectively when compared with untreated control. However, treatments T50 (Sk+Sp) and M23 (Sk+Sp) also increased seed weight but not significantly different with untreated control. For rice yield, the treatment 01-52 (Sk+Sp) provided the highest rice yield with 2,398.0 kg ha⁻¹ which was 17.66% of yield increment when compared with untreated control (2,038.0 kg ha⁻¹). The yield of 01-52 (Sk+Sp) was significantly higher than those from chemical fungicide treatment (2,036.0 kg ha⁻¹). However, there were unexpected reduction of rice yields but nonsignificant relative to untreated control in other treatments of *T. asperellum*, including T50 (Sk+Sp), M23 (Sk+Sp), T35-Co4 (Sk+Sp), CB-Pin-01 (Sk) and 01-52 (Sk), even though the increments of seed weight per panicle were obtained. The treatments T50 (Sk+Sp) and CB-Pin-01 (Sk+Sp) significantly increased the 1,000-seed weight by 12.03 and 9.99% when compared with untreated control. The 1,000-seed weight of T50 (Sk+Sp) and CB-Pin-01 (Sk+Sp) (26.35 g and 25.87 g, respectively) were comparable to those all *Trichoderma* treatments and of chemical fungicide treatment (24.92 g) (Table 3).

Table 3. Seed weight per panicle, weight of 1,000-seed and yield of rice plant (var. Chai Nat 1) grown with seeds soaked only (Sk) or seeds soaked and plants sprayed (Sk+Sp) with fresh culture bioproducts of various *Trichoderma asperellum* isolates.

Treatment	Seed weight (g/panicle) ^{1/}	Weight of 1,000-seed (g) ^{1/}	Rice yield (kg ha ⁻¹) ^{1/}
CB-Pin-01 (Sk ^{2/} +Sp ^{3/})	2.74 b ^{4/} (+17.60%) ^{5/}	25.87 ab (+9.99%) ^{5/}	2,204.00 ab (+8.15%) ^{5/}
01-52 (Sk+Sp)	2.33 c (0%)	24.52 abc (+4.25%)	2,398.00 a (+17.66%)
T50 (Sk+Sp)	2.69 b (+15.45%)	26.35 a (+12.03%)	1,829.80 cd (-10.22%)
M23 (Sk+Sp)	2.70 b (+15.88%)	25.11 abc (+6.76%)	1,756.20 d (-13.83%)
T35-Co4 (Sk+Sp)	3.14 a (+34.76%)	24.35 bc (+3.53%)	1,889.00 cd (-7.31%)
CB-Pin-01 (Sk)	2.83 ab (+21.46%)	25.33 abc (+7.70%)	1,957.60 bcd (-3.95%)
01-52 (Sk)	2.73 b (+17.17%)	24.26 bc (+3.15%)	2,076.00 bc (+1.86%)
propiconazole + difenoconazole (Sp)	2.35 c (+0.86%)	24.92 abc (+5.95%)	2,036.00 bc (-0.10%)
Control untreated	2.33 c -	23.52 c -	2,038.00 bc -

^{1/} Rice seed weight per panicle, weights of 1,000-seed and rice yield (kg ha⁻¹) derived from paddy seeds with 15% moisture content.

^{2/} Sk = Soaking rice seeds in *Trichoderma* spore suspension prepared from *Trichoderma* fresh culture bioproduct (100 g 20 L⁻¹ of water).

^{3/} Sp = Spraying on the whole rice plants with *Trichoderma* spore suspension prepared from *Trichoderma* fresh culture bioproduct (100 g 20 L⁻¹ of water).

^{4/} Means in each column followed by the same letter (s) are not significantly different according to Least Significant Difference (LSD) (P≤0.05).

^{5/} Percentage of increment (+) or decrement (-) of each treatment mean when compared with untreated control.

Comparison among treatments of seed soaking only or the combination of seed soaking and plant spraying with isolates CB-Pin-01 or 01-52 revealed the obvious effect of plant spraying on the increment of rice yield, but less effect on the increment of seed weight per panicle and 1,000-seed weight relative to untreated control. Among *Trichoderma* treatments, CB-Pin-01 (Sk+Sp) and 01-52 (Sk+Sp) provided most positive effects on the increment of both 1,000-seed weight and rice yield shown in Table 3 and rice growth (height and panicle per square meter) presented in Table 2. These

results are in line with those reported by Chamswarnng *et al.* (2012a), as the powder formulation of *T. asperellum* isolate 01-52 increased 1,000-seed weight, total yield and seed weight per panicle of rice variety Chai Nat 1. Whereas, pellet formulation of isolate 01-52 provided the yield increment of rice variety Pathum Thani 80 (Chamswarnng and Kumchang, 2012). Our results may be further explained and supported by recent findings of Doni *et al.* (2014a) who reported that all seven *Trichoderma* spp. tested were able to increase several rice physiological processes which included net photosynthetic rate, stomatal conductance, transpiration, internal CO₂ concentration and water use efficiency. These *Trichoderma* spp. isolates were also able to enhance rice growth components including plant height, leaf number, tiller number, root length and root fresh weight. The findings of Li *et al.* (2012) and Makino (2011) which concluded that improving rice physiological characteristics could contribute the achievement of high rice yield. Our results showed that the positive effects of most *T. asperellum* isolates on rice growth and yield were comparable to or even better than the use of chemical fungicide (propiconazole + difeniconazole) spraying. These finding seems to indicate the potential of *T. asperellum* isolates to provide better promotion of rice growth and yield, whereas using chemical fungicide lacks of capability to promote rice growth and yield.

Enhancement of plant growth induced by *Trichoderma* species has been reported across a large number of different groups of plants including vegetable, cereal, ornamental and forestry crops. For cereal crops such as maize (*Zea mays*) and wheat (*Triticum aestivum*) treated with *Trichoderma* spp. were resulted in enhanced stem, shoot length and shoot dry biomass, longer ear heads and more seeds. Similarly, *Trichoderma* isolate T52 enhanced shoot and root dry weights in rice by up to 38% (da Silva *et al.*, 2012). Various mechanisms have been proposed to explain plant growth promotion associated with *Trichoderma* species. These include synthesis of phytohormones, either by microbe or plants; production of vitamins; enhanced solubilization of soil nutrient; increased uptake and translocation of nutrient; enhanced root development and increased in the rate of carbohydrate metabolism; photosynthesis and plant defense mechanisms (Harman, 2006; Harman and Shores, 2007). Recently, several reports supported the efficacy of *Trichoderma* spp. for enhancing growth and yield of other monocots, maize and wheat (Akladios and Abbas, 2014; Harman, 2011a, 2011b; Kucuk, 2014) and ameliorated stress tolerance (Vinale *et al.*, 2008). Benitez *et al.* (2004) reported that *Trichoderma* strains were able to produce plant growth hormones like cytokinin-like molecules, e.g. zeatin and gibberellin GA3 or GA3-related which directly affected on plant growth.

Rice disease incidence and seed quality

The treatments Sk+Sp with *T. asperellum* isolates CB-Pin-01 and T50 significantly increased percentage of healthy seeds by 12.20 and 13.01%, respectively when compared with untreated control. Determination of dirty panicle infected seeds or discolored seeds revealed that the treatments Sk+Sp with isolates CB-Pin-01 and T50 and Sk with 01-52 provided significant reduction of discolored seeds by 43.73, 33.75 and 36.76%, respectively when compared with untreated control. All *Trichoderma* treatments except the treatment 01-52 (Sk) and 01-52 (Sk+Sp) significantly reduced the empty seeds by 17.00-23.98% when compared with untreated control. The other two *Trichoderma* isolates, M23 and T35-Co4 with Sk+Sp provided insignificant increase in healthy seeds and reduced dirty panicle infected seeds but significantly reduced empty seeds when compared with untreated control treatment (Table 4 and Fig. 3). Results from Table 4 also showed that there were non-significant differences in healthy seed percentages, dirty panicle infected seeds and empty seeds between treatments CB-Pin-01 (Sk+Sp) and CB-Pin-01 (Sk) as well as 01-52 (Sk+Sp) and 01-52 (Sk). Form the practical point of view, only soaking rice seeds with effective *T. asperellum* isolate may provide significantly increase healthy seeds, reduce dirty panicle infected seeds and empty seeds. The obtained results were also comparable to the use of chemical fungicide. The T50 (Sk+Sp) was another treatment which gave results similar to those four previous mentioned treatments.

Table 4. Percentages of healthy seeds, dirty panicle infected seeds and empty seeds of randomly harvested seed samples from rice plants (var. Chai Nat 1) grown with seeds soaked only (Sk) or seeds soaked and plants sprayed (Sk+Sp) with fresh culture bioproducts of various *Trichoderma asperellum* isolates.

Treatment	Healthy seeds (%) ^{1/}		Dirty panicle infected seeds (%) ^{1/}		Empty seeds (%) ^{1/}	
CB-Pin-01 (Sk ^{2/} +Sp ^{3/})	75.40 a ^{4/}	(+12.20%) ^{5/} (+10.96%) ^{6/}	8.97 d	(-43.73%) ^{5/}	14.40 cd	(-17.00%) ^{5/}
01-52 (Sk+Sp)	67.94 b	(+1.10%)	12.60 bcd	(-20.95%)	18.43 a	(+6.22%)
T50 (Sk+Sp)	75.94 a	(+13.01%) (+11.74%)	10.57 cd	(-33.75%)	13.19 d	(-23.98%)
M23 (Sk+Sp)	67.00 b	(-0.30%)	18.17 a	(-13.99%)	13.25 d	(-23.63%)
T35-Co4 (Sk+Sp)	71.04 ab	(+5.71%)	18.52 a	(-16.19%)	13.84 d	(-20.23%)
CB-Pin-01 (Sk)	71.98 ab	(+7.11%)	11.58 bcd	(-27.35%)	14.04 cd	(-19.08%)
01-52 (Sk)	69.74 ab	(+3.78%)	10.08 cd	(-36.76%)	16.61 abc	(-4.27%)
Propiconazole + difenoconazole (Sp)	67.96 b	(+1.13%)	14.13 abc	(-11.36%)	15.72 bcd	(-9.39%)
Control untreated	67.20 b	-	15.94 ab	-	17.35 ab	-

^{1/} Percent healthy seeds, dirty panicle infected seeds and empty seeds from three replications/treatment (5 g per replication).

^{2/} Sk = Soaking rice seeds in *Trichoderma* spore suspension prepared from *Trichoderma* fresh culture bioproduct (100 g 20 L⁻¹ of water).

^{3/} Sp = Spraying on the whole rice plants with *Trichoderma* spore suspension prepared from *Trichoderma* fresh culture bioproduct (100 g 20 L⁻¹ of water).

^{4/} Means in each column followed by the same letter (s) are not significantly different according to Least Significant Difference (LSD) (P≤0.05).

^{5/} Percentage of increment (+) or decrement (-) of each treatment mean when compared with untreated control.

^{6/} Percentage of increment (+) of treatment mean when compared with chemical fungicide treatment.



Fig. 3. Various characteristics of rice seeds (var. Chai Nat 1) (a) healthy seeds, (b) dirty panicle infected seeds and (c) empty seeds

The results obtained in Table 5 revealed that all *Trichoderma* treatments and chemical fungicide treatment significantly reduced leaf brown spot on rice plants by 16.04-35.15% and 14.92%, respectively when compared with untreated control. Among these *Trichoderma* treatments, isolates

T35-Co4 Sk+Sp and 01-52 with Sk provided significantly lower brown spot incidence (4.65% and 4.85%, respectively) compared to chemical fungicide treatment (6.10%). Whilst the brown spot incidences of treatments CB-Pin-01 (Sk+Sp), 01-52 (Sk+Sp), T50 (Sk+Sp), M23 (Sk+Sp) and CB-Pin-01 (Sk) were lower than those of chemical fungicide treatment but with non-significant different. For narrow brown spot, all *Trichoderma* treatments reduced disease incidences by 0.19-19.78% while 40.95% of significant disease reduction was found in chemical fungicide treatment. Narrow brown spot incidences of treatments M23 (Sk+Sp) (10.70%) and T50 (Sk+Sp) (9.85%) were not significant different from the control (10.72%). The lowest narrow brown spot incidence was found in a treatment 01-52 (Sk) (8.60%), but this was not significant different with treatments CB-Pin-01 (Sk+Sp) (9.00%), CB-Pin-01 (Sk) (9.27%), 01-52 (Sk+Sp) (8.70%) and T35-Co4 (Sk+Sp) (8.97%). All *Trichoderma* treatments except treatment 01-52 (Sk+Sp) reduced rice sheath rot (8.56 - 40.21%), while treatments CB-Pin-01 (Sk), M23 (Sk+Sp) and 01-52 (Sk) significantly reduced sheath rot (40.21, 34.02 and 32.47%, respectively) as compared to untreated control. Chemical fungicide reduced this disease by 23.30% as compared to untreated control. The sheath rot incidences resulted from using all *T. harzianum* isolates (5.80-8.87%) except 01-52 (Sk+Sp) (10.22%) were comparable to the use of chemical fungicide (7.44%) (Table 5 and Fig. 4).

Table 5. Percentages of brown spot, narrow brown spot and sheath rot disease incidences on rice plants (var. Chai Nat 1) grown with seeds soaked only (Sk) or seeds soaked and plants sprayed (Sk+Sp) with fresh culture bioproducts of various *Trichoderma asperellum* isolates.

Treatment	Brown spot (%) ^{1/}		Narrow brown spot (%) ^{1/}		Sheath rot (%) ^{2/}	
CB-Pin-01 (Sk ^{3/} +Sp ^{4/})	5.58 bc ^{5/}	(-22.16%) ^{6/}	9.00 bc	(-16.04%) ^{6/}	7.45 bcd	(-23.20%) ^{6/}
01-52 (Sk+Sp)	6.02 b	(-16.04%)	8.70 c	(-18.84%)	10.22 a	(+5.36%)
T50 (Sk+Sp)	5.35 bcd	(-25.38%)	9.85 ab	(-8.12%)	8.87 abc	(-8.56%)
M23 (Sk+Sp)	5.72 b	(-20.22%)	10.70 a	(-0.19%)	6.40 cd	(-34.02%)
T35-Co4 (Sk+Sp)	4.65 d	(-35.15%)	8.97 bc	(-16.32%)	8.53 abc	(-12.06%)
CB-Pin-01 (Sk)	5.50 bc	(-23.29%)	9.27 bc	(-13.53%)	5.80 d	(-40.21%)
01-52 (Sk)	4.85 cd	(-32.36%)	8.60 c	(-19.78%)	6.55 cd	(-32.47%)
Propiconazole+ difenoconazole (Sp)	6.10 b	(-14.92%)	6.33 d	(-40.95%)	7.44 bcd	(-23.30%)
Control untreated	7.17 a	-	10.72 a	-	9.70 ab	-

^{1/} Percentages of brown spot and narrow brown spot disease incidences were calculated from five flag leaves of each replication (four replications/treatment).

^{2/} Percentage of sheath rot disease = (number of sheath rot infected panicles x 100)/Total panicles

^{3/} Sk = Soaking rice seeds in *Trichoderma* spore suspension prepared from *Trichoderma* fresh culture bioproduct (100 g 20 L⁻¹ of water).

^{4/} Sp = Spraying on the whole rice plants with *Trichoderma* spore suspension prepared from *Trichoderma* fresh culture bioproduct (100 g 20 L⁻¹ of water).

^{5/} Means in each column followed by the same letter (s) are not significantly different according to Least Significant Difference (LSD) (P≤0.05).

^{6/} Percentage of brown spot, narrow brown spot and sheath rot disease increment (+) or decrement (-) of each treatment mean when compared with untreated control.



Fig. 4. Symptoms of rice diseases caused by fungi (a) dirty panicle or seed discoloration, (b) brown spot, (c) narrow brown spot and (d) sheath rot.

From overall consideration, the use of *T. asperellum* isolate 01-52 for rice seed soaking was the most effective treatment that provided high reduction of all three diseases. Brown spot, narrow brown spot and sheath rot were respectively reduced by 32.26, 19.78 and 32.47% relative to the control. The treatment CB-Pin-01 (Sk+Sp) provided similar with non-significantly lower efficacy when compared with a treatment 01-52 (Sk). Another three treatments, CB-Pin-01 (Sk), 01-52 (Sk+Sp) and T50 (Sk+Sp) also provided similar efficacy for reducing all three diseases when compared with a treatment CB-Pin-01 (Sk+Sp). Among *Trichoderma* treatments, the highest reduction of brown spot, narrow brown spot and sheath rot were resulted from treatments T35-Co4 (Sk+Sp) (35.15%), 01-52 (Sk) (19.78%) and CB-Pin-01 (Sk) (40.21%), respectively. Among three diseases, only soaking rice seeds with *T. asperellum* isolates CB-Pin-01 and 01-52 provided higher or non-significant reduction of those diseases when compared with the treatments CB-Pin-01 (Sk+Sp) and 01-52 (Sk+Sp). Most *Trichoderma* treatments, with several exceptional cases, reduced brown spot and sheath rot comparable to the use of chemical fungicide (Table 5). The results seems to indicate that soaking rice seeds without plant spraying with effective *T. asperellum* isolate may be the promising practice for reducing rice diseases. This finding is also an alternative practice for reducing the use of chemical fungicide.

A reason for the reduction of brown spot, narrow brown spot and sheath rot of rice, was probably due to the antagonistic activities of *T. asperellum* through seed soaking singly (Sk) or seed soaking and plant spraying (Sk+Sp). Efficacy of *T. asperellum* for controlling fungal pathogens through competition, antibiosis, mycoparasitism and induced resistance has been well known and accepted. Our previous results indicated the efficacy of *T. asperellum* isolates to suppress rice fungal pathogens *in vitro*, such as, *A. padwickii*, *B. oryzae*, *C. lunata* and *S. oryzae* (Charoenrak *et al.*, 2009 and unpublished data). Moreover, recently study revealed that soaking rice seeds with *T. asperellum* as dry powder, pellet or fresh culture formulation increased chitinase activity in 45-d-old rice leaves which indicated the possibility of induced systemic resistance occurred in rice plants against both dirty panicle and leaf brown spot diseases (Charoenrak *et al.*, 2012).

Roles of seed soaking and plant spraying applications on disease reduction were supported by the research results of Perelló *et al.* (2009) which indicated that the antagonism of *Trichoderma* spp. was effective at an early stage of the *Septoria tritici* blotch disease on wheat under field conditions only. In addition, the comparison of application techniques, spraying spore suspension onto leaves and the coating seeds of wheat revealed that both were effective in decreasing the disease. *Trichoderma* spp. could restore vigor and improve germination, even in the presence of any pathogenic organisms (Björkman *et al.*, 1998). Early works of Chamswarnng *et al.* (2009) and Chamswarnng *et al.* (2012c), revealed that dirty panicle infected rice seeds soaked overnight in spore suspension of *T. asperellum* had the highest percentage of germination and also promoted seedling growth with the longer shoots of 7-d-old rice seedlings when compared to untreated control.

Moreover, detection of dirty panicle pathogens by the blotter method indicated the lowest detected *Fusarium* spp. and *Curvularia* spp. counts from seeds treated with *T. asperellum* compared to untreated control. The results showed reduction of rice pathogens and improvement of seed health and vigor of dirty panicle infected rice seeds derived from all *Trichoderma* treatments. Therefore, the reduction of foliar diseases on rice plants prior to harvest in this study may resulted from the reduction of rice pathogens by seed soaking with *T. asperellum*. On the other hands, the use of *T. harzianum* application through seed biopriming in rice and wheat plants enhanced drought tolerance (Shukla *et al.*, 2012, 2015).

Rice root colonization

Rice roots of 28-d-old seedlings were found highly colonized by *T. asperellum*, from 17.77 to 100%. However, the root colonization percentages of all *Trichoderma* treatments were markedly reduced to 12.50-56.25% after harvesting. Detection of root colonization by *T. asperellum* from harvested rice plants indicated that isolates 01-52 and T50 with Sk+Sp provided the highest root colonization, both with 56.25%, followed by isolate 01-52 with Sk (43.75 %), isolate CB-Pin-01 and isolate T35-Co4 with Sk+Sp, both with 31.25%. The lowest root colonization percentage was found in *Trichoderma* treatment M23 with Sk+Sp (12.50%). Interestingly, soaking rice seeds singly with isolates 01-52 and CB-Pin-01 (Sk) provided non-significant but lower percentages of root colonization (43.75% and 26.25%) than those the same isolates with Sk+Sp (56.25% and 31.25%) (Table 6 and Fig. 5).

Table 6. Colonization percentages of *Trichoderma asperellum* from fresh culture bioproduct on roots of harvested rice plants at 28 and 108 d after sowing (DAS).

Treatment	Root colonization (%) ^{1/}	
	28 DAS	108 DAS
CB-Pin-01 (Sk ^{2/} +Sp ^{3/})	100.00 a ^{4/}	31.25 bc
01-52 (Sk+Sp)	88.89 b	56.25 a
T50 (Sk+Sp)	77.77 c	56.25 a
M23 (Sk+Sp)	100.00 a	12.50 d
T35-Co4 (Sk+Sp)	17.77 d	31.25 bc
CB-Pin-01 (Sk)	100.00 a	26.25 cd
01-52 (Sk)	88.89 b	43.75 ab
propiconazole +difenoconazole (Sp)	ND ^{5/}	ND
Control (H ₂ O)	ND	ND

^{1/} Root colonization percentage was calculated from $[(C_1/C_2) \times 100]$, where C_1 is the number of root segments which were colonized by *T. asperellum* and C_2 is the total number of root segments from each treatment which were placed on Martin's medium.

^{2/} Sk = Soaking rice seeds in the *Trichoderma* spore suspension prepared from *Trichoderma* fresh culture bioproduct (100 g 20 L⁻¹ of water).

^{3/} Sp = Spraying on the whole rice plants with *Trichoderma* spore suspension prepared from *Trichoderma* fresh culture bioproduct (100 g 20 L⁻¹ of water).

^{4/} Means in each column followed by the same letter (s) are not significantly different according to Least Significant Difference (LSD) ($P \leq 0.05$).

^{5/} ND = Not determined

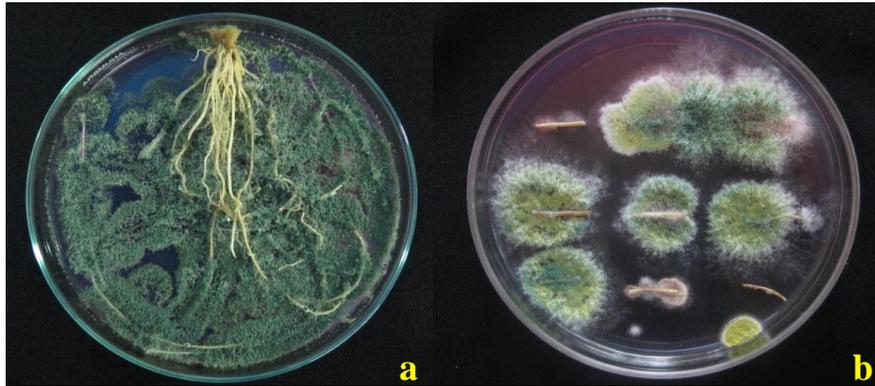


Fig. 5. Mycelia and spores of *Trichoderma asperellum* isolate 01-52 produced from roots of (a) 28-d-old rice seedling and (b) 108-d-old rice plants. All roots were placed on Martin's medium.

Based on our results, obvious evidences of rice root colonization by all tested isolates of *T. asperellum* from seedling to harvesting stage of rice plants might play important roles on the reduction of brown spot, narrow brown spot, sheath rot (Table 5), dirty panicle infected seeds (Table 4) and the increment of yield (Table 3). High percentages of root colonization by *T. asperellum* isolates 01-52 (56.25 %) and CB-Pin-01 (31.25 %) recovered from roots of harvested rice plants indicated the ability of these isolates to penetrate and live in rice roots in a manner similar to mycorrhizal fungi. In addition, there was a possibility that these isolates restricted to the epidermis and outer cortex of roots similar to previously reported in cucumber plants treated by *T. asperellum* T203 in hydroponic system (Yedidia *et al.*, 2003) and in hydroponic lettuce roots treated by *T. asperellum* CB-Pin-01 (Lamool, 2006). However, the lowest percentage of root colonization by *T. asperellum* isolate M23 at 108 d after sowing (12.50%) also indicated the lack of survival ability of fungal mycelia on rice roots of this isolate. Previous reports indicated that root colonization by *T. harzianum* resulted in increased level of plant enzymes, including various peroxidases, chitinases, β -1,3-glucanases, lipoxygenase-pathway hydroperoxide lyase and compounds like phytoalexins and phenols to provide durable resistance against diseases and stress. (Harman, 2006; Hoitink *et al.*, 2006). Vinale *et al.* (2008) reported that *Trichoderma* species were capable to colonize plant root and produced compound that changed plant metabolism and stimulate plant defense. After penetration into the plant, metabolites such as peroxidase, xylanase, glucanase, swollenin, cellulase and endochitinase would be produced and acted as elicitors which activated defense-gene, known as mycoparasitic gene and induced resistance against pathogens (Benítez *et al.*, 2004; Shores *et al.*, 2010; Thakur and Sohal, 2013). Above mentioned reports could explain our results which showed the relationship between high root colonization percentages by *T. asperellum* 01-52, CB-Pin-01 and reduction of rice diseases as well as enhancement of plant growth and yield.

There were many researchers reported the abilities of root colonization by *T. harzianum* to increase nutrient uptake and fertilizer utilization efficiency which resulted in plant growth promotion. *T. harzianum* T22 treated maize seed showed the high efficacy to increase the nitrogen use efficacy (Akladios and Abbas, 2014; Harman, 2011a, 2011b). Similar to Lamool (2006) who reported that the application of spore suspension of *T. asperellum* CB-Pin-01 (1 kg-fresh culture 2,000 L⁻¹) into nutrient solution of lettuce grown in the NFT hydroponic system effectively provided not only the reduction of Pythium root rot but also the promotion of lettuce growth. Root colonization by *T. asperellum* CB-Pin-01 also resulted in the enhancement of phosphorus, potassium and nitrogen in lettuce leaves by 70, 18 and 6% as compared to untreated control, respectively. There was previous suggestion that elicitors produced by the *Trichoderma* activated specific gene expression pathways involved in plant defense response which resulted in the promotion of root growth leading to enhance

nutrient uptake and subsequent plant growth promotion (Yedidia *et al.*, 2003). Moreover, phosphorus (P) is an important element in a complete and balanced fertility program that can improve crop health and reduce the incidence and severity of many crop diseases. Rice plants treated with *Trichoderma* spp. also had better nutrient uptake. This occurrence enhanced the physiological process within the rice plants, leading to good growth performance (Doni *et al.*, 2014a).

From our experiment, all five isolates of *T. asperellum* used for soaking rice seeds and plant spraying effectively reduced all three leaf disease incidences (Table 5) and dirty panicle infected seeds (Table 4). While only two isolates (01-52, CB-Pin-01) increased yield, especially isolate 01-52 which provided the highest increment of total yield (Table 3). These results were similar to the report of Chamswarnng and Intanoo (2007) which obtained dirty panicle reduction and yield increment by using spore suspension prepared from fresh culture bioproducts of *T. asperellum* CB-Pin-01 and 03-7/I34 for seed soaking and plant spraying. Doni *et al.* (2014b) reported that soaked rice seeds in 10^7 ml⁻¹ spores of seven *Trichoderma* spp. isolates for 30 min significantly increased rice seedling growth, germination rate, vigor index and speed of germination. They suggested that the healthy and invigoration rice seedlings would affect the enhancement of rice quality and yield.

CONCLUSION

Application of five *T. asperellum* isolates as fresh culture bioproduct by soaking rice seeds (var. Chai Nat 1) singly or in combination with rice plants spraying for three times for reducing diseases and increasing yield of rice was conducted. *T. asperellum* 01-52 gave the highest rice yield, enhanced 1,000-seed weight, increased healthy seeds, whilst reduced dirty panicle discolored seeds, brown spot, narrow brown spot and sheath rot incidences when compared with untreated control. In addition, *T. asperellum* isolate 01-52 provided comparable efficacy with the isolate CB-Pin-01. Most *Trichoderma* treatments provided better overall efficacies than the use of chemical fungicide. All five isolates of *T. asperellum* were able to colonize rice roots from seedling (28-d-old) to harvesting stage (108-d-old) of rice plants. Seed soaking and plant spraying with *T. asperellum* 01-52 and CB-Pin-01 can be a promising, low cost method and environmentally friendly way to be easily implemented using in normal agricultural practices and sustainable organic rice production in the future. However, another field trials using *T. asperellum* isolates 01-52 and CB-Pin-01 in comparison with chemical fungicide should be investigated in fields at various locations or even of different seasons and rice varieties to validate and strengthen the results obtained from this study. The costs and benefits derived from using these biocontrol agents should be considered and compared with the use of chemical fungicide.

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CO₂ AND CH₄ EMISSIONS ON DIFFERENT WATER MANAGEMENT AND PESTICIDE TREATMENTS IN RICE FIELDS OF TIDAL PEAT SWAMP

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ABSTRACT

Utilization of peatlands for agriculture will be associated with water management and pesticides. Rice cultivation in peatlands become potential contributor to global warming through carbon dioxide (CO₂) and methane (CH₄) emission. The study was conducted in tidal peat swamp at Central Kalimantan Province at October 2012 to March 2013. The study aims to evaluate the effect of water management and pesticides on CO₂ and CH₄ emissions. The study was designed in a split plot design with three water management treatments as the main plot and six pesticide treatments as subplots. The water management treatments: control (A0), saturated water (A1), and intermittent irrigation (A2). The pesticide treatments: control (P0), paraquat during tillage (P1), fenobucarb for every week application (P2), fenobucarb for every two weeks application (P3), difenoconazole for every week application (P4), and difenoconazole for every two weeks application (P5). Fenobucarb which is applied every week (P2) was able to suppress CO₂ flux as much as 40%. Intermittent irrigation (A2) could mitigate CO₂ flux measured from soil as much as 36%. Suppression mechanism of GHG emissions appear to be associated with the binding mechanism between pesticide and phenolic acids.

Key words: butylphenyl methylcarbamate, difenoconazole, saturated, intermittent irrigation, paraquat dichloride,

INTRODUCTION

Peatlands are wetland ecosystems that are characterized by the accumulation of organic matter called "peat" which derives from dead and decaying plant material under high water saturation conditions (Parish *et.al.* 2008). Peatlands are known as a very important carbon stock (Andriesse, 1988; Page and Rieley, 1998), and influence the stability of carbon dioxide (CO₂), methane (CH₄) and nitrous oxide (N₂O) (Parish *et.al.* 2008). The peat composition of Central Kalimantan is dominated by lignin (Sabiham, 2010). Lignin disintegrates under aerobic and anaerobic conditions and result in several derivative phenolic acids (Stevenson, 1994; Orlov, 1995). *Ferulic*, *synapic*, *p-coumaric*, *vanilic*, *syringic*, and *p-hydroxybenzoic* acids are the most important derivative phenolic acids found in the peatlands of Central Kalimantan (Sabiham, 1997; Mario and Sabiham, 2002) (Fig. 1). These acids were categorized as the main sources of carbon-release due to the high content of carboxyl (–

COOH) and methoxy (–OCH₃) groups. COOH could be broken down completely into CO₂ and H₂O through the processes of oxidation-reduction. CO₂ could also be released when the methoxy groups (–OCH₃) changed into –OH groups during the phenol-OH formation through the processes of demethylation, hydroxylation and oxidation (Sabiham, 2010). The other important gas, methane (CH₄), is produced by strict anaerobic bacteria (methanogens) (van der Gon and Neue, 1995).

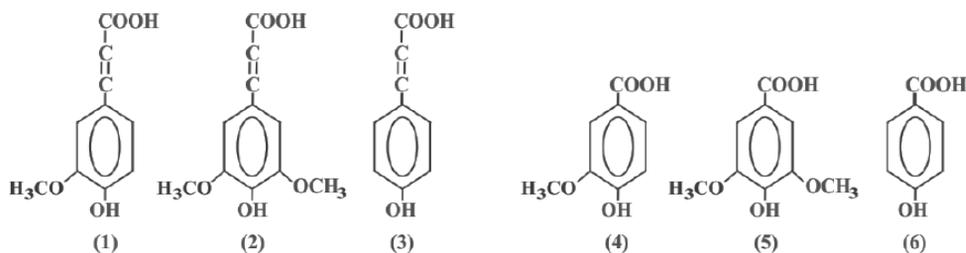


Fig. 1. The most important derivative phenolic acids in the peats of Jambi and Central Kalimantan: (1) Ferulic acid; (2) Synapic acid; (3) *p*-Coumaric acid; (4) Vanilic acid; (5) Syringic acid; and (6) *p*-Hydroxybenzoic acid.

When peatlands are disturbed, they can become significant sources of CO₂ and CH₄ (Parish *et.al.* 2008). Water management is one of strategies in mitigating emissions in rice field. Previous studies reported that intermittent irrigation was able to reduce CH₄ emission on paddy field about 36% to 79% compared to the flooding method (Shin *et.al.*, 1996; Setyanto, 2006).

The development of peatlands for agriculture is also associated with pesticides. Pesticides have become a mainstay of farmers to overcome the problem of pests and diseases of rice crop. Although pesticides are used in agriculture, little is known about their effect on greenhouse gas emission. According to Stevenson (1994) pesticide or its decomposition product is able to form a chemically stable relationship with organic materials. Two mechanisms may occur for the formation of chemical bonds between pesticides and organic materials, they are (1) pesticide can be directly attached to the reactive part of the surface of colloidal organic by chemical bonds, and (2) during the process of humification, pesticide may insert into the structure of newly formed humic and fulvic acid. Through this binding mechanism, then the decomposition of organic acids into CO₂ and CH₄ can be inhibited.

Paraquat dichloride (1,1'-dimethyl-4,4'-bipyridinium) is one of a group of bipyridylum herbicides (Fig. 2). Paraquat dichloride is a non-selective contact herbicide used widely throughout Indonesian farmers for control of broadleaf weeds and grasses. Difenconazole (1-[2-[2-chloro-4-(4-chloro-phenoxy)-phenyl]-4-methyl[1,3]dioxolan-2-ylmethyl]-1H-1,2,4-triazole) is a broad-spectrum fungicide used for disease control in many fruits, vegetables, cereals and other field crops (Fig. 2). This fungicide belongs to azole fungicides group. Fenobucarb (2-(1-methylpropyl) phenyl methyl carbamate) is one of a group of carbamate insecticides (Fig. 2). It is non-systemic insecticides with contact action to control leafhoppers, plant hoppers, trips, and weevils on rice, and several field crops. Those pesticides are commonly used by farmers to overcome pest and disease problems (The British Crop Protection Council, 1997).

This study therefore sought to evaluate the effects of water management and pesticides on CO₂ and CH₄ emissions from rice field in tidal peat swamp.

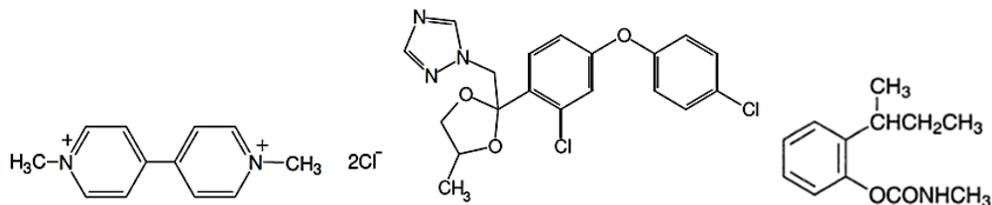


Fig. 2. Herbicide paraquat dichloride (left), fungicide difenoconazole (middle) and insecticide fenobucarb (right) (The British Crop Protection Council. 1997)

MATERIALS AND METHODS

Research Site

The study was carried out in the agricultural field located in Kanamit Jaya Village, Pulang Pisau District, Central Kalimantan Province (-2° 55'46"N and 114° 10'16" E). The study was conducted during the rainy season from October 2012 to March 2013 in a tidal peat swamp. Peatland used in this study was shallow peat with 50 -100 cm in depth, with hemic level of peat decomposition rate (rate of peat's decomposition around 33-66%). Land research included into zone of tidal freshwater swamp, which is land still directly influenced by tidal energy in the form of rising and declining water stream following cyclical movement of the tide (Subagyo, 2006).

Research Design

The experiment was conducted in plots set-up as split plot design with 3 replications. The main plot was water management treatments (A) and the subplot was pesticides treatments (P) and each plot measured 5m x 4m. Water management was carried out with three treatments: control without water management (A0), saturated water (A1), and intermittent irrigation (A2) (Fig. 3).

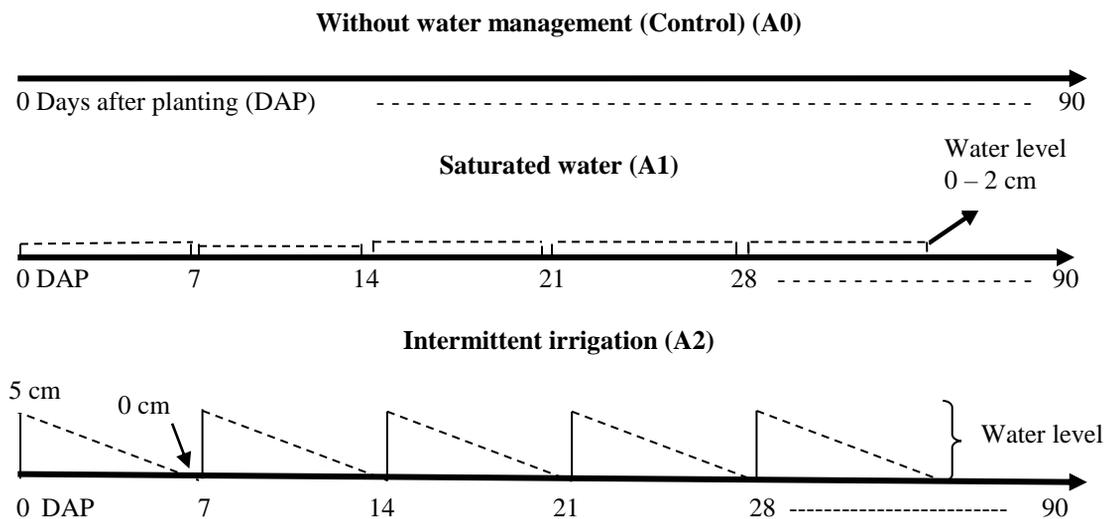


Fig. 3. Water management treatment scheme.

Saturated water treatment was maintained in water level as high as 2 - 5 cm from soil surface. While intermittent irrigation treatment was maintained through filled up the plot with the water as high as 5 cm from soil surface and gradually lowered the water surface until it reaches 0 cm in 7 days. Then on day 8 onwards was done the same way until the maturity stage. The water level was monitored and maintained continuously according to treatments. PVC pipes (5-cm diameter, 70-cm long) were installed horizontally on the soil surface in each plot to remove excess water during heavy rains. Piezometers were installed on each plot at a depth of 80 cm from the soil surface to monitor water level. The experimental plot was bound by plastic with 60 cm depth and 20 cm above to avoid lateral movement of water.

The pesticide application was laid out in six different treatments:

1. control without pesticides (P0),
2. herbicide paraquat during tillage (P1),
3. insecticide fenobucarb for every week application (P2),
4. insecticide fenobucarb for every two weeks application (P3),
5. fungicide difenoconazole for every week application (P4),
6. fungicide difenoconazole for every two weeks application (P5).

The pesticides used in the experiment were commonly used in the agricultural fields, which were obtained from a local agricultural store. The pesticides used were: herbicide paraquat dichloride, insecticide fenobucarb, and fungicide difenoconazole. The plots were sprayed with three different pesticides individually (except control) at company recommended rates of 3 liters ha⁻¹ for paraquat, 1 liter ha⁻¹ for fenobucarb, and 0.5 liters ha⁻¹ for difenoconazole. The herbicides paraquat was applied only once at three weeks prior to planting. Application of insecticide fenobucarb and fungicide difenoconazole was started at 7 days after planting according to the treatment. During flowering period (70 days after planting), pesticide application was stopped temporarily to minimize disruption of the flowering process, then continued after flowering period elapsed up to plants approached grain maturity.

Inpara 5, a variety of rice that adapts to swampland, was planted at 20 days after germination with a distance of 20 cm (between row) x 10 cm (on a row) and 40 cm (hallway) with 2 plants per hill. Rice crops were fertilized with 250 kg urea.ha⁻¹, 135 kg SP36 (superphosphate, 36 percent P₂O₅).ha⁻¹, and 100 kg KCl (potassium chloride).ha⁻¹. These application rates are the recommended amount for application to a rice crop in peatland (Noor, 2001).

Flux Measurement

Gas flux measurements were made using closed chamber technique adopted from the International Atomic Energy Agency (IAEA) (1992). Closed chamber were made of Plexiglass with 50 cm wide x 50 cm long x 100 cm high, equipped with a fan (12 V DC) and a thermometer. The flux chambers were placed covered four hills of rice plants at 30, 60 and 90 days after planting to measure of CO₂ and CH₄ emission from soil and plants. Another chamber with 17 cm wide x 50 cm long x 35 cm high size was placed in the ground in the hallway between the rice plants to measure of CO₂ and CH₄ emission from soil. One set of closed chambers were placed at each plot for all treatments with three replications.

Gas samples were collected in gas-tight syringes (Terumo, 10 ml) for six time points at 3 minute intervals to determine the change in trace gas concentration over time. Fluxes were measured at 0 day after planting (DAP), 30, 60 and 90 DAP during rice plant grow stages and done in the morning between 6:00 a.m. to 8:00 a.m. Gas samples have been placed in a cooling box and then taken to the laboratory of Indonesian Swampland Agriculture Research Institute at Jalan Kebun Karet,

Loktabat Utara, Banjarbaru South Kalimantan to be analyzed on the same day every time the gas samples taken. Gas samples were analyzed using a CP-4900 Micro-GC gas chromatography (GC) with a flame ionization detector (FID) to analyze CH₄, and thermal conductivity detector (TCD) to analyze the CO₂ gas. Before the gas analysis is done, the GC was calibrated using the standard gas at concentrations of 10 ppm for CH₄ and 600 ppm for CO₂. Gas (carrier gas) used was helium UHP (ultra high purity) 150 kPa with a gas purity of 99.99%. Column temperature was 100 °C, Injector time was 255 ms, and flow mode was Continuous. Results of the analysis indicate gas concentration (ppm), which is used to determine the rate of change of the concentration of gas per unit of time used in the calculation of GHG fluxes.

Calculation of flux at each treatment using the equation adopted from IAEA (1992) as follows:

$$E = \frac{Bm}{Vm} \times \frac{\delta Csp}{\delta t} \times \frac{V}{A} \times \frac{273.2}{T+273.2}$$

Where E is CO₂ or CH₄ emission (mg.m⁻².min⁻¹), V is volume of the closed chamber (m³), A is area of the base closed chamber (m²), T is average air temperature inside the close chamber (°C), $\frac{\delta Csp}{\delta t}$ is concentration changing rate of CH₄ or CO₂ (ppm.min⁻¹), Bm is molecular weight of CH₄ or CO₂ under standard conditions, Vm is volume of gas at standard temperature and pressure conditions is 22.4q litre at 23°K.

Phenolic acids measurement

Soil sample for organic acids were taken at the time of CO₂ and CH₄ sample were also taken. Soil samples were taken at five points diagonally to the plots and then mix into one soil sample. Phenolic acids were calculated using method by Angeles *et al.* (2006). 5 grams of fresh peat samples were put into a bottle, and then 25 ml of distilled water was added and shaken for 30 minutes. Then sample centrifuged with 3000 rpm for 60 minutes, and about 20 ml of the supernatant filtered using 0.45µm Millipore filter. The analysis focused on the measurement of the concentration of *ferulic acid*, *synapic*, *p-coumaric*, *vanilic*, *siringic*, and *p-hydroxybenzoic* using partition separation method with a reversed-phase C18 column (2.9x300 mm µBondapakTM) and UV detector with light-D2 at wavelength of 280 nm. High Performance Liquid Chromatography (HPLC) Shimadzu 20A used for quantitative analysis of derivative phenolic acids.

Statistical analysis

The data were statistically analyzed using ANOVA with SAS Statistics 9.1 software. Multiple mean comparisons were made by Duncan's Multiple Range Test (DMRT).

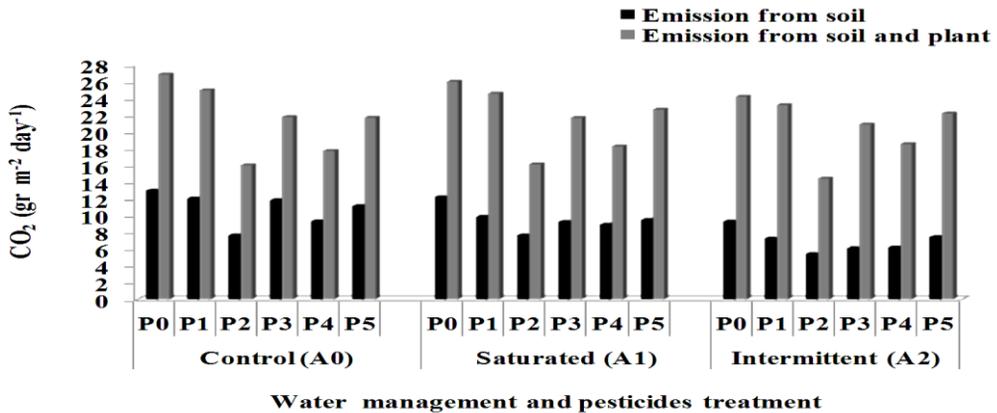
RESULTS AND DISCUSSION

Trace gas flux

Production and consumption of CO₂ and CH₄ take place in soil environments. The study found that during the period of flux measurements, CO₂ emission measured from soil and plant was higher than CO₂ emission measured from soil for all treatments (Fig. 4). On the contrary CH₄ emission recorded higher at measured from soil than CH₄ emission measured from soil and plant (Fig. 5).

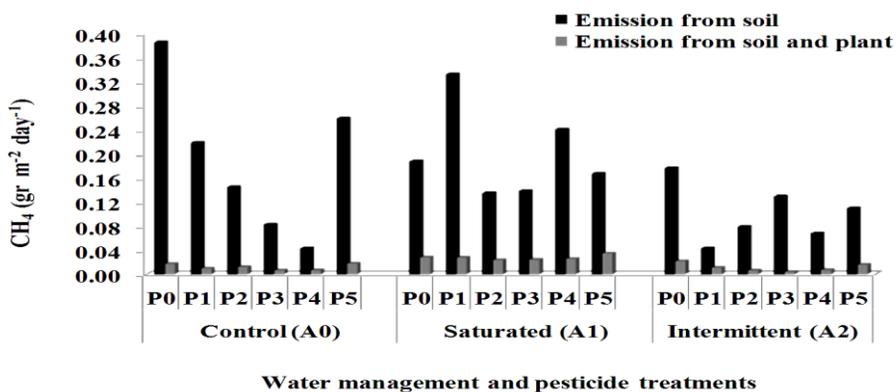
Heterotrophic and microbial decomposition of soil organic matter and root respiration from plants are responsible for CO₂ and CH₄ production in soil (Satomayor and Rice, 1999). Microbial

communities and decomposition processes in the rhizosphere were higher than the soil that was not influenced by the presence of plant roots (bulk soil). Faster decomposition process is causing CO₂ emissions in the rhizosphere higher than soil. Meanwhile, CH₄ was produced in anaerobic conditions by methanogenic *Archaea* bacteria. These bacteria grow at a pH between 6 and 8, some are known to occur under more acidic conditions (Whalen, 2005; Lay 2009). Only a fraction of the methane produced emitted into the atmosphere, a large number of other consumed by methanotrophic bacteria. CH₄ consumption occurs in the zone around the plant roots oxygenated. Potential of methane oxidation by methanotrophic is usually larger than potential of methane production by methanogens (Segers, 1998). Consequently, methanotrophic bacteria can reduce the amount of methane released into the atmosphere substantially. In line with the statement above, the research found that CH₄ recorded in very small quantities and CH₄ emissions measured from soil was higher than CH₄ emissions measured from soil and plants.



Without any pesticide/control (P0), Paraquat at tillage (P1), Fenobucarb every week application (P2), Fenobucarb every two week application (P3), Difenconazole every week application (P4), Difenconazole every two week application (P5).

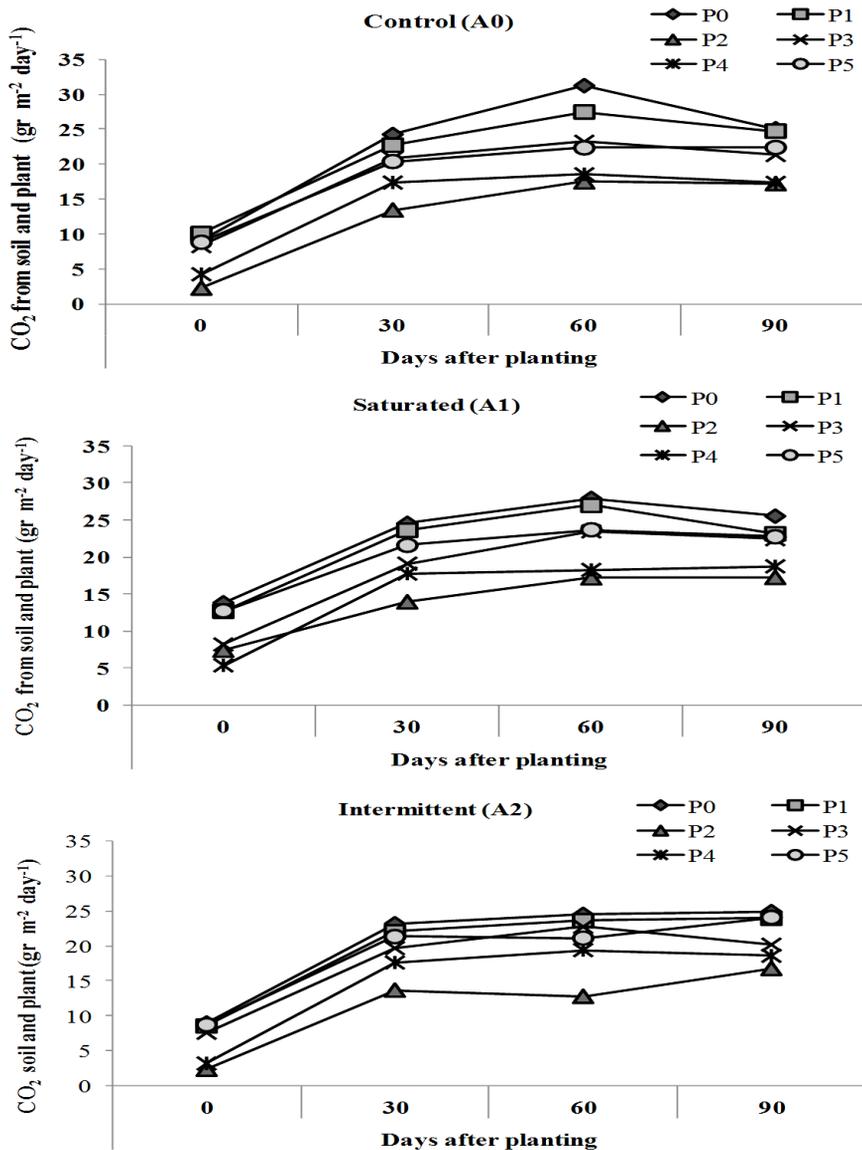
Fig. 4. Flux of CO₂ measured from soil and plant and from soil (without plant)



Without any pesticide/control (P0), Paraquat at tillage (P1), Fenobucarb every week application (P2), Fenobucarb every two week application (P3), Difenconazole every week application (P4), Difenconazole every two week application (P5).

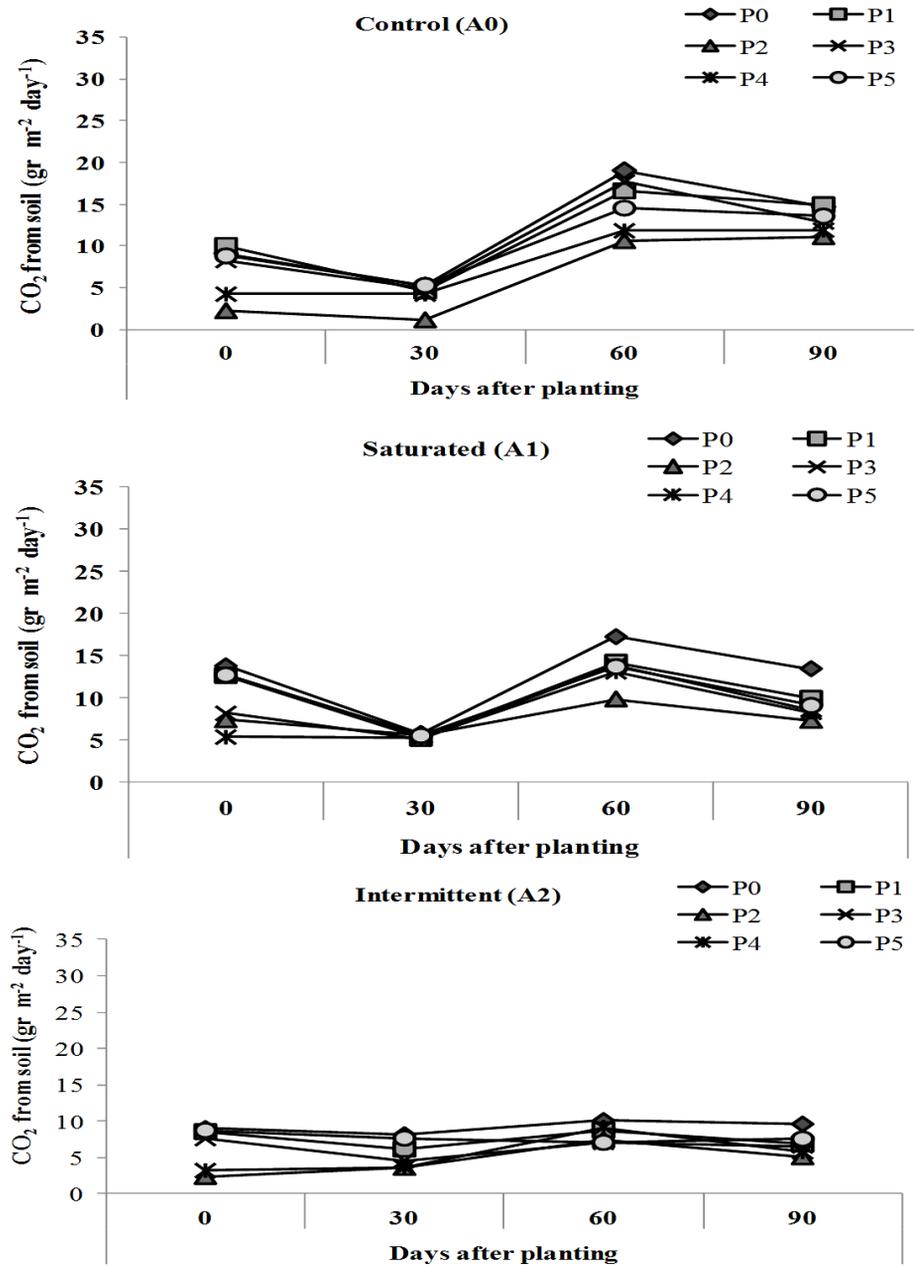
Fig. 5. Flux of CH₄ measured from soil and plant and from soil (without plant)

In line with the statements above, different trend CO₂ emissions were also observed between emission measured from soil and emissions measured from soil and plant. Trend CO₂ emission measured from soil and plant showed increasing along with the growth of plants and decreased at 90 day after planting (Fig. 6) while CO₂ emissions measured from soil fluctuated during the observation period (Fig. 7).



Without any pesticide/control (P0), Paraquat at tillage (P1), Fenobucarb every week application (P2), Fenobucarb every two week application (P3), Difenoconazole every week application (P4), Difenoconazole every two week application (P5).

Fig. 6. Carbon dioxide flux from soil and plant at different water management and pesticides treatment.



Without any pesticide/control (P0), Paraquat at tillage (P1), Fenobucarb every week application (P2), Fenobucarb every two week application (P3), Difenoconazole every week application (P4), Difenoconazole every two week application (P5).

Fig. 7. Carbon dioxide flux from soil and plant at different water management and pesticides treatment.

Carbon dioxide emission measured from soil mainly affected by ground water level that create aerobic and anaerobic condition. Aerobic conditions related to oxygen penetration in peat

when the water table declines, favors CO₂ and inhibits CH₄ production, whereas anaerobic conditions associated with high water content, favor lower CO₂ and greater CH₄ production rates (Moore and Dalva, 1997; Turetsky and Ripley, 2005). Meanwhile CO₂ emission measured from soil and plant was affected by water level and plant rhizosphere. Ekberg *et al.* (2007) reported that; (1) increasing the availability of substrates such as dead roots in the rhizosphere, thus increasing the quality of decomposer community, (2) increasing the availability of root exudates and will increase the decomposition of lignin derivatives, (3) the composition of the structure of dissolved C organic. Decomposition process in rhizosphere is causing CO₂ emissions measured from soil and plant was linear with rice growing.

Methane flux

Methane emission was observed very small concentration hence it was no clear effect in inhibition of pesticides and water management to CH₄ emission. Although plots with pesticides treatment (P1, P2, P3, P4, and P5) had lower CH₄ emissions relative to the control (P0), and intermittent irrigation (A2) had lower CH₄ emissions compared to saturated (A1) and control (A0). However, statistical analysis showed water management and pesticides and interaction between those treatments had not significant effect on CH₄ emissions both from soil and from soil and plant (*p* > 0.05) (Table 1 and 2). Hence further discussion will focus on CO₂ emission.

Table 1. Methane flux for different pesticide treatments

Pesticide treatments	CH ₄ from soil				CH ₄ from soil and plant			
	30 DAP	60 DAP	90 DAP	MEAN	30 DAP	60 DAP	90 DAP	Mean
----- gr m ⁻² day ⁻¹ -----								
P0	0.003a	0.004a	0.002a	0.003a	0	0	0	0
P1	0.002a	0.003a	0.001a	0.002a	0	0	0	0
P2	0.002a	0.001a	0.000a	0.001a	0	0	0	0
P3	0.002a	0.001a	0.001a	0.001a	0	0	0	0
P4	0.001a	0.002a	0.001a	0.001a	0	0	0	0
P5	0.002a	0.003a	0.001a	0.002a	0	0	0	0

DAP = days after planting; Without any pesticide/control (P0); Paraquat at tillage (P1); Fenobucarb every week application (P2); Fenobucarb every two week application (P3); Difenconazole every week application (P4); Difenconazole every two week application (P5).

The number followed by the same letters in the same column indicate no significant difference between treatments at 0.05 level DMRT.

Table 2. CO₂ flux from different water management treatments.

Water management treatments	CH ₄ from soil				CH ₄ from soil and plant			
	30 DAP	60 DAP	90 DAP	Mean	30 DAP	60 DAP	90 DAP	Mean
----- gr m ⁻² day ⁻¹ -----								
Control (A0)	0.002a	0.003a	0.000a	0.002a	0.000	0.000	0.000	0.000
Saturated (A1)	0.001a	0.003a	0.002a	0.002a	0.000	0.000	0.000	0.000
Intermittent (A2)	0.002a	0.001a	0.000a	0.001a	0.000	0.000	0.000	0.000

DAP = days after planting; Without any pesticide/control (P0); Paraquat at tillage (P1); Fenobucarb every week application (P2); Fenobucarb every two week application (P3); Difenconazole every week application (P4); Difenconazole every two week application (P5).

The number followed by the same letters in the same column indicate no significant difference between treatments at 0.05 level DMRT

Carbon Dioxide flux

Pesticides from the group of *bypirilidium*, *carbamate* and *azole* were used in this study had lower emissions of CO₂. Plots treated with herbicide (P1), insecticide (P2 and P3) and fungicide (P4 and P5) had lower CO₂ emissions relative to the control (P0) plot (Table 3). Statistical analysis showed pesticides treatment had significant effect on CO₂ emissions both from soil and from soil and plant ($p < 0.05$).

Table 3. Carbon dioxide flux from different pesticide treatments

Pesticide treatments	CO ₂ from soil				CO ₂ from soil and plant			
	30 DAP	60 DAP	90 DAP	Mean	30 DAP	60 DAP	90 DAP	Mean
	----- gr m ⁻² day ⁻¹ -----							
P0	6.39d	15.46c	12.58e	11.48d	23.98e	27.88d	25.15c	25.67e
P1	5.32bc	13.20b	10.57d	9.70c	23.07e	26.47d	24.23c	24.59d
P2	3.47a	9.33a	7.84a	6.88a	13.68a	15.81a	17.07a	15.52a
P3	4.90b	12.88b	9.39bc	9.04c	19.86c	23.12c	21.28b	21.42c
P4	4.37ab	11.35b	8.66ab	8.13b	17.57b	18.73b	18.21a	18.17b
P5	6.16bc	11.77b	10.08cd	9.34Cc	21.09cd	22.37c	23.05bc	22.17c

DAP = days after planting; Without any pesticide/control (P0); Paraquat at tillage (P1); Fenobucarb every week application (P2); Fenobucarb every two week application (P3); Difenconazole every week application (P4); Difenconazole every two week application (P5).

The number followed by the same letters in the same column indicate no significant difference between treatments at 0.05 level DMRT.

The insecticide fenobucarb, applied every week (P2), significantly suppressed CO₂ flux both measured from soil and from soil and plant, and was able to suppress CO₂ emission from soil and from soil and plant by around 40% compared to control (P0). While other treatments were able to suppress CO₂ emissions from soil as much as 28%, 21%, 19%, 15% and from soil and plant as much as 29%, 17%, 14%, and 6% for P4, P3, P5, and P1, respectively.

The study also demonstrates that the use of the same active ingredient but different application frequencies (P2 and P3, P4 and P5) produce different flux levels. As indicated in the treatment of P3 and P5 (fenobucarb and difenoconazole applied every two weeks) significantly had higher CO₂ and CH₄ flux than P2 and P4 (fenobucarb and difenoconazole applied every week).

Observations also were carried out on CO₂ flux resulting from the three water management treatments. During the rice growing period, the rate of CO₂ emissions in the intermittent irrigation (A2) was decreased as it is compared to saturated condition (A1) and control (A0) (Table 4). Intermittent irrigation (A2) could mitigate CO₂ flux from soil as much as 36% compared to saturated condition (A1) 12% and control (A0). Statistical analysis showed water management had significant effect on CO₂ from the soil ($p < 0.05$), but had no significant effect on CO₂ emissions from soil and plant ($p > 0.05$).

Treatment without water management such as environmental design A0 tend to experience fluctuations in water level changes very quickly, these conditions favor the formation of CO₂ flux. Agus *et al.* (2010) in his research on peatland in Central Kalimantan reported that the ground water level is the dominant factor affecting CO₂ emissions. Up to 50 cm of ground water table is positively correlated to CO₂ emissions. Meanwhile, land flooded continuously at saturated conditions such as treatment A1 produced an environment that was very conducive to the formation of CO₂ flux. This is due to peat decomposition of organic matter under anaerobic conditions can produce organic acids,

CO₂ and CH₄, whereas peat material in aerobic conditions can produce CO₂ (Morril *et al.* 1982). Peat can also produce CO₂ under anaerobic conditions in the present of acetic acid, propionic acid, and butyric acid as an electron donor (Rinnan *et al.* 2003). Intermittent irrigation treatment (A2), appears to provide better results for the suppression of CO₂ flux. Period of land were flooded and not flooded produce aerobic and anaerobic conditions that can inhibit the formation of CO₂ flux.

Table 4. CO₂ flux from different water management treatments.

Water management treatments	CO ₂ from soil			Mean	CO ₂ from soil and plant			Mean
	30 DAP	60 DAP	90 DAP		30 DAP	60 DAP	90 DAP	
Control (A0)	4.23a	15.04c	13.16c	10.81c	19.81a	23.36b	21.32a	21.49a
Saturated (A1)	5.44b	13.69b	9.47b	9.53b	20.08a	22.90b	21.62a	21.53a
Intermittent (A2)	5.64b	8.27a	6.91a	6.94a	19.58a	20.69a	21.41a	20.56a

DAP = days after planting

The number followed by the same letters in the same column indicate no significant difference between treatments at 0.05 level DMRT

During the rice growing period, variation of groundwater level for each water treatment can be seen in Fig. 8. Tidal peat swamp that used in this study directly influenced by the tide and receding of water, the land will be inundated with water and dry quickly. Moore and Dalva (1993) reported a close relation exists between water table and GHG emissions. Intermittent irrigation was designed to create soil environment not only to ensure the availability of sufficient water to support plant growth but also to depress emissions of CO₂. Intermittent irrigation systems are closely related to groundwater level that creates aerobic and anaerobic condition. A temporal shift between aerobic and anaerobic conditions is a major control for peat accumulation and CO₂ and CH₄ production rates (Aragonés and Blodau, 2012). Hooijer *et al.* (2010) on his research in drained peatlands in Southeast Asia reported that each 10 cm reducing water level will increase 9.1 t CO₂ ha⁻¹y⁻¹ of CO₂ emission rate. Quick changing in water level was identified as the factor which supported CO₂ and CH₄ formation.

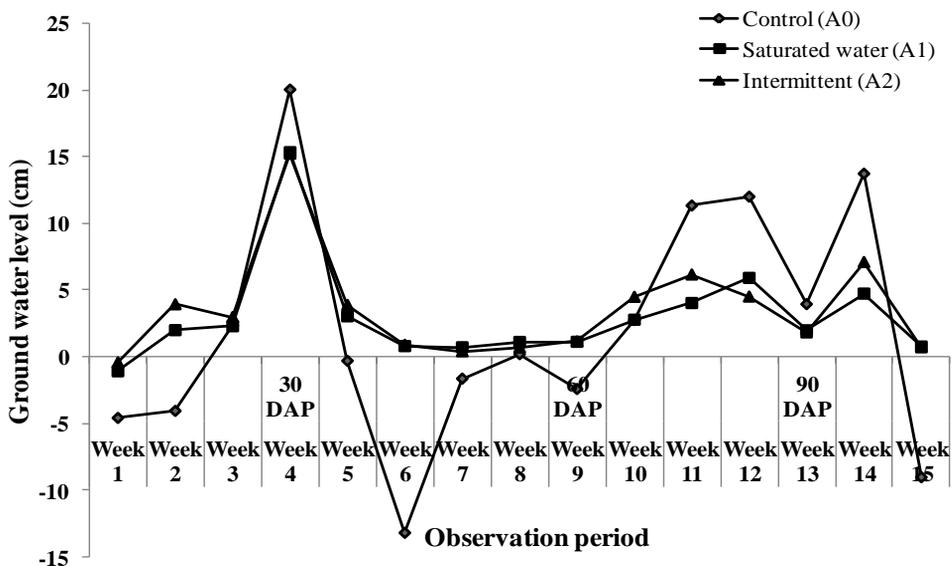


Fig. 8. Groundwater level from rice field under water management treatments at tidal peat swamp

The data obtained from interaction between water management and pesticide treatments known that P2 and A2 had the lowest CO₂ flux (Table 5), but analysis of variance showed that there was no interaction between those treatments on CO₂ flux.

Table 5. Flux of CO₂ on the interaction between water management and pesticide treatments

Pesticide & water treatment	CO ₂ from soil				CO ₂ from soil and plant			
	A0	A1	A2	Mean	A0	A1	A2	Mean
	----- gr m ⁻² day ⁻¹ -----							
P0	12.97	12.19	9.27	11.48	26.83	25.98	24.19	25.67
P1	12.03	9.81	7.25	9.70	24.96	24.56	23.18	24.23
P2	7.62	7.62	5.41	6.88	16.01	16.12	14.42	15.52
P3	11.81	9.22	6.10	9.04	21.75	21.65	20.86	21.42
P4	9.30	8.90	6.18	8.13	17.72	18.25	18.54	18.17
P5	11.12	9.46	7.43	9.34	21.68	22.65	22.17	22.17

Without water treatment /control (A0); Saturated condition (A1); Intermittent irrigation (A2); Without any pesticide/control (P0); Paraquat at tillage (P1); Fenobucarb every week application (P2); Fenobucarb every two week application (P3); Difenconazole every week application (P4); Difenconazole every two week application (P5).

There was no significant interaction between water management and pesticides treatments ($p > 0.05$).

The results of this study reinforce the importance of water conservation in controlled conditions in peatlands. Water management is important to be done in GHG mitigation and to prevent peat subsidence rate due to acceleration of the process of organic matter decomposition.

Interaction between Pesticides, Phenolic Acids and CO₂ Flux

The inhibition of CO₂ flux on pesticide treatment plots suggested that interaction between phenolic acids and pesticides leads to low flux. Most of pesticides will fall to the ground at the time applied (Sa'id, 1994) and may form stable chemical bonds with organic acids (Stevenson, 1994). The results showed that the amount of phenolic acids decreased during the observation period except for synapic acid and *p*-hydroxybenzoic acid (Fig. 9).

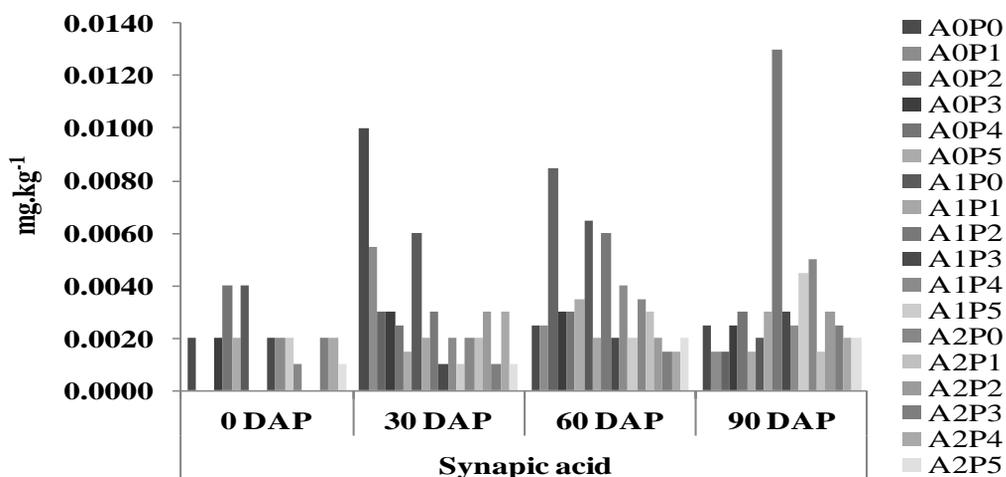


Fig. 9. Phenolic acids on different water management and pesticides treatment

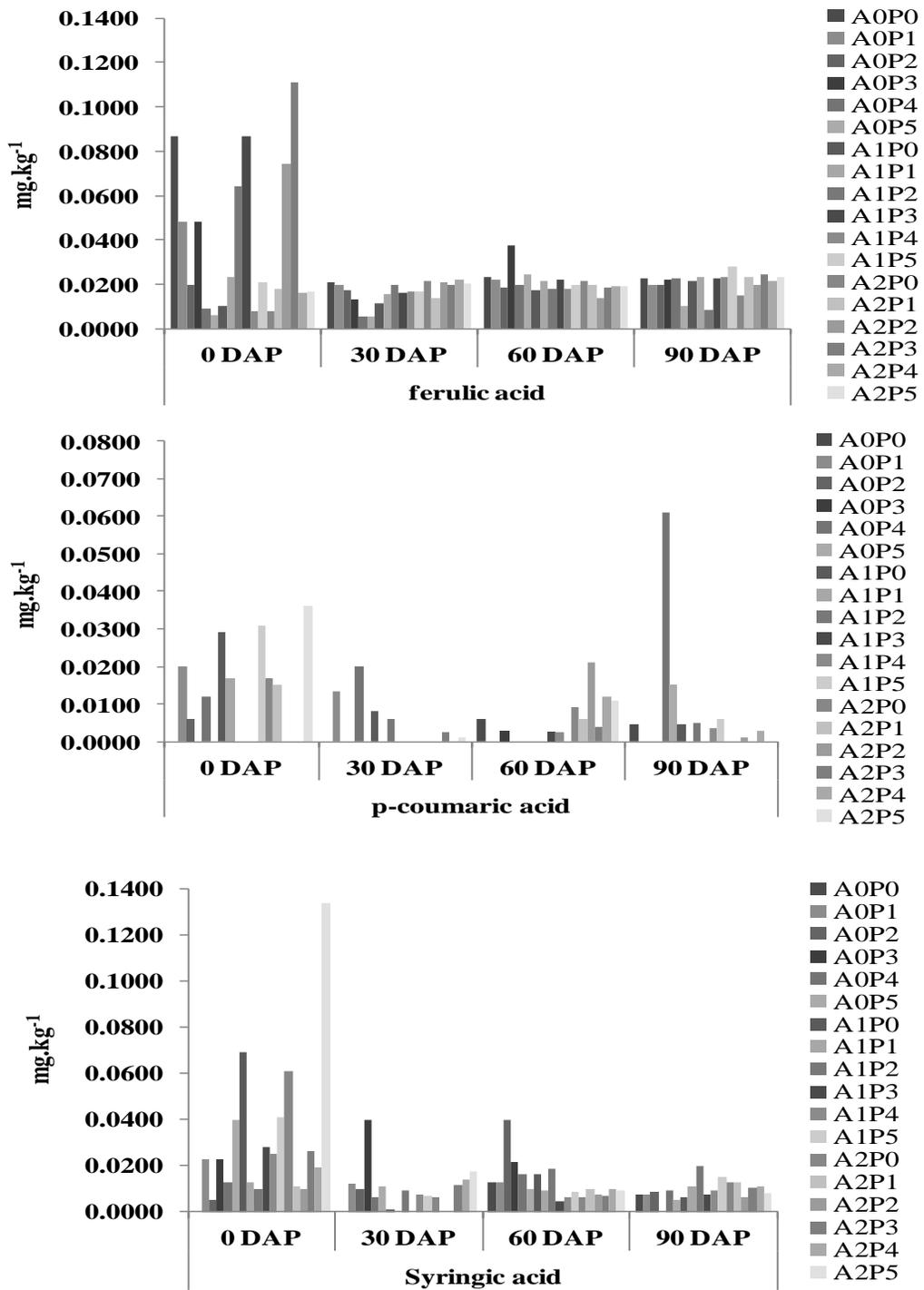
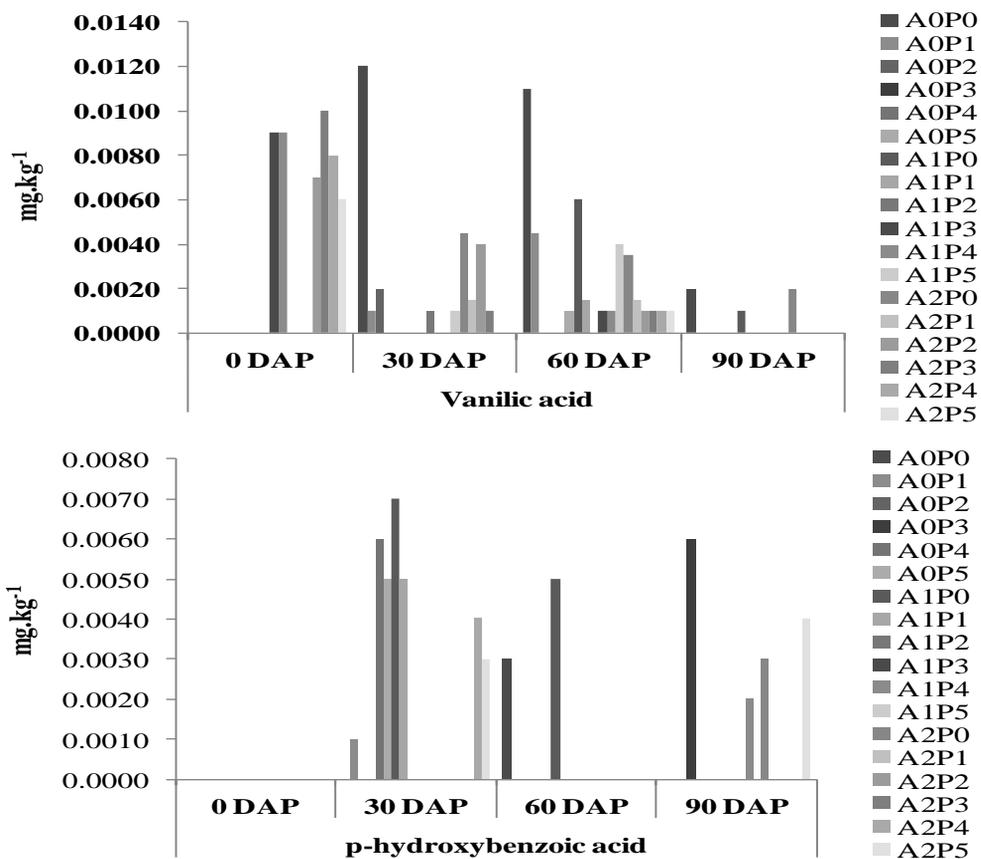


Fig. 9. Phenolic acids on different water management and pesticides treatment (continued)



Without any management water / control (A0), Saturated water (A1), Intermittent irrigation (A2), Without any pesticide/control (P0); Paraquat at tillage (P1); Fenobucarb every week application (P2); Fenobucarb every two week application (P3); Difenconazole every week application (P4); Difenconazole every two week application (P5).

Fig. 9. Phenolic acids on different water management and pesticides treatment

An interaction between chemicals substances in pesticides with soil organic matter is a very important factor that influences the fate of pesticides in soil as well as decomposition of organic matter into CO₂ and CH₄. Peat contains many aliphatic and aromatic acids. In acidic conditions, the charge on peat organic matter is determined by the ionization of carboxylic groups that will support the adsorption of the cationic pesticide. Paraquat is pesticides that can be adsorbed either on the soil surface by replacing inorganic cations or by ionic interaction mechanism with the negative charge on the surface of the ground, where the electrostatic effect be the deciding factor (Arce et al. 2011). Stevenson (1994) reported that paraquat has binding affinity through various ways, such as physical bonding, hydrogen bonding, ion exchange and protonation. The divalent paraquat, have the potential to react with more than one side of the negative charge on the colloidal humus soil, for example through two ion COO⁻. Khan (1989) also reported that paraquat is absorbed in a larger amount by humus, fulvic acid and by organo-clay complexes than Diquat. Binding mechanism between Diquat and paraquat with phenolic acids may illustrate as follows (Fig. 10).

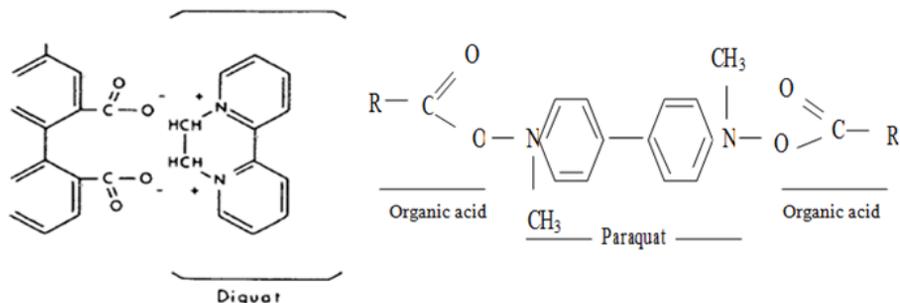


Fig. 10. Illustration of binding mechanism between diquat and paraquat with phenolic acids (Stevenson, 1994).

Furthermore Stevenson (1994) also reported that pesticides can also form a stable bond with the organic material through hydrogen bond (H-bond) (Fig. 11).

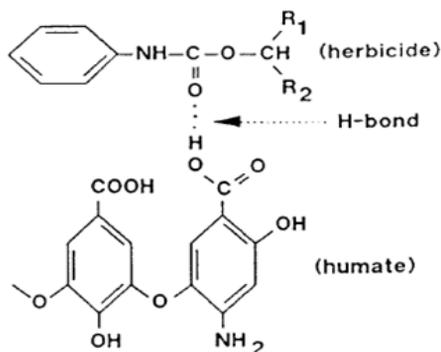


Fig. 11. Illustration of binding mechanism between herbicide with phenolic acids through hydrogen bonding (Stevenson 1994)

Pesticide can be absorbed by soil colloids, and absorption can be very strong, strong, moderate and weak depending on the type of soil (Zimdahl 1993). The ability to absorb mineral soil monmorillonit higher than allopan and kaolinite. Uptake will increase with increasing organic matter content, clay, kation exchange, and decreases with increasing pH and temperature. Further Zimdahl (1993) and Waldron (1992) reported that soil with high organic matter adsorb pesticides greater than sandy soils. Through this binding mechanism, the pesticide becomes inactive and prevents decomposition of phenolic acid into CO₂. Further studies in more controlled conditions are needed to determine the binding mechanism of pesticides and phenolic acid related to CO₂ and CH₄ emission in tidal peat swamp.

CONCLUSION

Pesticides paraquat, fenobucarb and difenoconazole used in this study had not led to an increase in CO₂ flux in peatland's rice field. Fenoburarb which was applied every week (P2) had the lowest CO₂ flux and was able to suppress CO₂ flux as much as 40%. Meanwhile, the result of water management treatment shown that intermittent irrigation (A2) significantly reduced CO₂ flux measured from soil as much as 36%. There was no significant interaction between pesticide and water management treatments, but the data obtained showed that interaction between P2 and A2 had the lowest CO₂ flux. Binding mechanisme between pesticides and phenolic acid may reduce the rate of decomposition of phenolic acids into gases (CO₂ and CH₄) and lead to lower CO₂ flux.

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INFLUENCE OF LIVING MULCH ON WEED GROWTH AND RICE PRODUCTIVITY IN PADDY RICE FIELDS

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ABSTRACT

Polyculture systems have the potential to improve crop performance. A new polyculture system was tested in rice (*Oryza sativa* L.) paddy fields. *Spirodela polyrhiza* was used as living mulch, as it does not compete with rice for light or nutrients. The rice cultivars, Akamai and Kichinkokumai, were used to compare living mulch with rice bran as a non-living mulch and the control treatment without mulch. Treatments were applied 7 days after transplanting in an experimental field of the Agricultural and Forestry Research Center, University of Tsukuba, Japan, from June to November in 2009 and 2010. The excessive biomass of *S. polyrhiza* decreased rice growth. Beneficial effects of *S. polyrhiza* on rice yield were only observed for Akamai. The Akamai yield was increased by 13% compared to the control due to an increase in panicle numbers. Rice bran application did not affect Akamai yield. The cumulative solar radiation and water temperature were low in the *S. polyrhiza* treatment compared to the control, but the same as the control for the rice bran treatment. Reduction in radiation and temperature by a floating mat of living mulch directly affected weed growth. Therefore, *S. polyrhiza* has the potential for being a living mulch in rice paddy fields, preventing weed growth and increasing rice yield for specific rice cultivars.

Key words: duckweed, polyculture system, rice bran, *Spirodela polyrhiza*, weed suppression

INTRODUCTION

In terms of productivity, modern agriculture has developed successfully with the use of chemicals and mechanization (Piper, 1999). However, these developments have also brought environmental problems, such as soil erosion, water pollution of lakes, and disturbance of ecosystems (Oyanagi, 2006). Reduced use of agricultural chemicals in crop production systems has affected weed production and crop productivity, as weeds are recognized as the major contributors to reductions in crop yield (Uchino *et al.*, 2011). Grain yield losses from weed infestation are estimated to be 44-96% compared with weed control plot, depending on the rice production system (Ampong-Nyarko and De Datta, 1991) and farmers' income (Bärberi, 2003). Growing cover crops is suggested as one way to achieve a sustainable agricultural system (Sakai and Hara, 2004; Sustainable Agriculture Network, 1998).

The main benefits of cover crops are weed control, reduced soil erosion, prevention of soil nutrient loss, enhanced N fixation, and increased biological diversity (Hartwig and Ammon, 2002; Sustainable Agriculture Network, 1998). A cover crop is defined as (1) any living ground cover plant that is grown during the season or off-season and is killed before planting the main crop, or (2) a plant grown at the same time as the main crop that serves as living mulch during the growing season (Hartwig and Ammon, 2002; Teasdale, 2003). For example, legumes (Hiltbrunner *et al.*, 2007), Chinese milk

vetch (*Astragalus sinicus* L.; Samarajeewa *et al.*, 2005), clover (*Trifolium* spp.; den Hollander *et al.*, 2007), rye (*Secale cereale* L.), Italian ryegrass (*Lolium multiflorum*), and hairy vetch (*Vicia villosa* Roth) are widely used as cover crops in upland fields (Komatsuzaki, 2004) and during the fallow season in rice paddy fields (Ueno, 2004). Living mulch cover crops, such as Chinese milk vetch (Mineta *et al.*, 1997) and legume (Asagi and Ueno, 2009), are used only in upland fields converted from paddy fields to improve soil fertility of the fields. Cover crops may reduce weed germination through allelopathic and mechanical effects (Teasdale *et al.*, 2007), as well as competition between the cover crop and weeds for limited resources such as light, nutrients, water, and space (Piper, 1999). Competition is also affected by cultural practices, such as plant density and the planting date and method (Ampong-Nyarko and De Datta, 1991).

A cropping system that cultivates two or more crops by mixing species at the same time in the same field is called a polyculture system. This type of system has the potential to improve cropping system performance in terms of yield, nutrient cycling efficiency, weed suppression, and enhanced efficiency in utilizing land, labor, natural resources, pest control, and carbon fixation (Piper, 1999). The potential benefits from using the polyculture system in rice paddy fields, as proposed by Sakai *et al.* (2003), are (1) increased carbon fixation by vertical habitat segregation, (2) reduced use of fertilizer and agrochemicals, (3) improved fertilizer use efficiency through plural plants, and (4) improved landscapes in paddy fields.

The potential for polyculture involving rice and water hyacinth (*Eichhornia crassipes*) was cited by Sakai *et al.* (2008). They found that rice yield was not affected significantly after inoculation with water hyacinth at a rate of up to half the rice planting density, but yield was compromised as the water hyacinth planting density became greater than half the rice planting density, as paddy rice and water hyacinth competed for light and nutrients. Nevertheless, the total biomass of this system increased as the planting density of water hyacinth increased.

Azolla spp., a floating aquatic plant, is capable of suppressing weed emergence in rice paddy fields (Biswas *et al.*, 2005; Satapathy and Sing, 1985; Shougomori and Yoshioka, 1999), but the impact on rice yield has not been reported. Most *Azolla* spp. are used as a source of N bio-fertilizer in rice fields (Amano and Ahmed, 2004). Although *Azolla* spp. and water hyacinth (*E. crassipes*) have some advantages, both are difficult to grow in rice paddy fields in Japan. *Azolla japonica* is very difficult to distinguish from *A. cristata*, which is defined as an invasive alien species by law in Japan (Ministry of the Environment, 2012a). Water hyacinth is quite common as an ornamental aquatic plant, yet it is one of the 100 most aggressive plant species in the world (Ministry of the Environment, 2012b).

Lemna spp. and *Spirodela* spp., also known as duckweed, are floating aquatic plant species that are widely distributed in tropical and sub-tropical regions (FAO, 2013). These two species affect N fixation by cyanobacteria (Duong and Tiedje, 1985), which are used in water purification because these accumulate heavy metals (FAO, 2013; Zimmo *et al.*, 2004) and as animal feed (FAO, 2013). Cyanobacteria also improve N use efficiency by reducing the amount of ammonia volatilized from urea, resulting in increased rice yields (Li *et al.*, 2009). Both of these duckweed species are domestic aquatic plants in Japan (Kadono, 1994). A domestic floating aquatic plant grown as a living mulch might be the best choice to construct a polyculture system in rice paddy fields and to reduce light competition. Therefore, this study sought to determine the potential for using *S. polyrrhiza* as a living mulch in a rice paddy field to improve rice productivity and weed control.

MATERIALS AND METHODS

The experiment was conducted in an experimental field of the Agricultural and Forestry Research Center (AFRC), University of Tsukuba, Japan, from June to November in 2009 and 2010. The experimental field (29.1 × 1.8 m) was divided into two areas to accommodate the rice (*Oryza sativa*

L.) cultivars Akamai and Kichinkokumai. Compound fertilizer (N:P₂O₅:K₂O = 6:8:4) was applied as a basal dressing at a rate of 6.3 g N m⁻², 8.4 g P₂O₅ m⁻², and 4.2 g K₂O m⁻². Eighteen-day-old rice seedlings were transplanted on June 16, 2009, and June 15, 2010, with spacing of 30 × 15 cm (22 hills m⁻²), with four seedlings per hill.

S. polyrhiza collected from a lotus field near Lake Kasumigaura, Tsuchiura, Japan, was used as living mulch in a rice paddy field. *S. polyrhiza* was washed with tap water and weighed before being applied to experimental plots 7 days after transplanting (DAT) the rice. In 2009, *S. polyrhiza* was applied at a rate of 0.64 kg FW m⁻² as 100% of the flood water surface area in a plot size of 0.9 × 1.05 m (0.945 m²) and compared with no mulch as the control. In 2010, the treatments were *S. polyrhiza*, applied at 1.57 kg FW m⁻², rice bran applied at 0.1 kg m⁻², and no mulch as a control, in 1 × 1-m plots. The growth of *S. polyrhiza* varied yearly, as affected by the different environmental conditions in the lotus fields. A piping system was used to maintain the water level at a depth of 8 cm throughout the season. Plots were separated by 20-cm-high plastic panels to prevent aquatic plants from drifting to neighboring plots. Akamai and Kichinkokumai were harvested in October and November, respectively.

Plant growth in terms of plant length, stem number, and SPAD value was measured on five hills per plot every 3 weeks in 2009 and 10 hills per plot every 2 weeks in 2010. The leaf area index (LAI) of the rice plant canopy was measured once a week using LAI-2000 (LI-COR, USA). Optleaf (Y-1W, Taisei E&L, Japan) was set at ground level in the floodwater to measure the cumulative amount of solar radiation every week. The biomass of *S. polyrhiza* in each 25 × 25-cm quadrat was measured every 2 weeks after inoculation and returned to the field immediately after the fresh weights were recorded. The weeds in a 30 × 30-cm quadrat were counted every 2 weeks during the growing season. After the rice was harvested, weeds were sampled and counted, and their dry weight was recorded. A thermorecorder (TR-52, T&D Corporation, Japan) was used to record the water temperature at 60-minute intervals. At maturity, five hills of rice plants per plot were harvested at ground level to measure yield components and brown rice yield.

The experiment was conducted in a randomized block design with three replicates for each rice cultivar. Data were analyzed using a t-test in 2009 and Tukey's HSD test in 2010, using JMP software (Ver. 8.01, SAS, Japan).

RESULTS AND DISCUSSION

Growth of *S. polyrhiza*

Duckweed reproduces asexually. Branching and fragmentation of the shoots result in development of a new frond (Lemon and Posluszny, 2000). Generally, the growth of aquatic plants is affected by plant photosynthesis, respiration, and plant tissue decay, which are influenced by light availability, water temperature, water velocity, nutrients, dissolved inorganic carbon concentration, plant density, and self-shading (Carr *et al.*, 1997; Lemon *et al.*, 2001).

At the initiation of this experiment, the entire areas of *S. polyrhiza* plots were covered. However, factors affecting plant density and self-shading resulted in a decline in *S. polyrhiza* fresh weight biomass (Fig. 1), which decreased with inoculation time (approximately 60% after 35 DAT) for both Akamai and Kichinkokumai. Lemon *et al.* (2001) mentioned the variation in population growth rates that affected frond development and retention times. The short lifespan of *S. polyrhiza* affected frond production rate, and ultimately, plant survival.

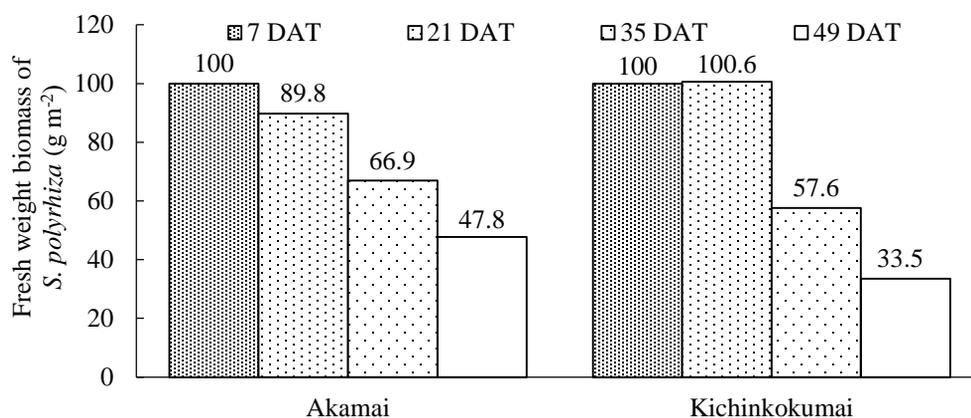


Fig. 1. Changes in fresh weight biomass of *S. polyrhiza* during the growing period for Akamai and Kichinkokumai in 2010. (Numbers represent the percentage of fresh weight biomass compared to 7 DAT)

Effects of Living Mulch on Growth and Yield of Rice

Basic information on the living mulch growth characteristics for a candidate species is essential. Generally, cover crops compete with the main crop for nutrients and light (Piper, 1999; Sakai *et al.*, 2008). *S. polyrhiza* is a floating plant mat. Inoculation with 100% *S. polyrhiza* did not result in competition for light on rice plant as showed the same results of the *Azolla* spp. (Biswas *et al.*, 2005; Lumpkin and Plucknett, 1980) and also did not compete for nutrients. Inoculation of *S. polyrhiza* significantly increased the growth of Akamai (Fig. 2a) and Kichinkokumai (Fig. 2b) compared with the control in both experiments.

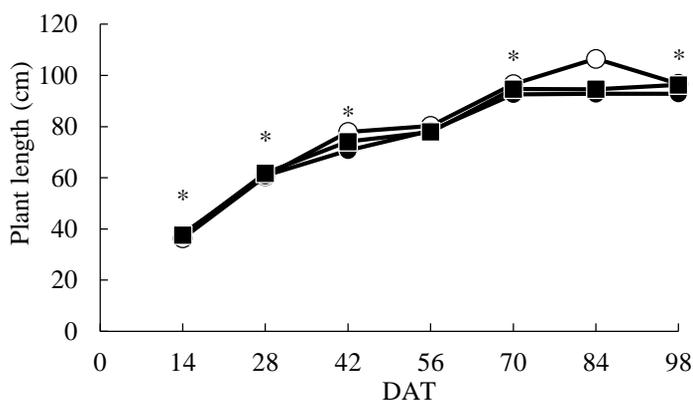


Fig. 2a. The growth of Akamai under *S. polyrhiza* (-○-), rice bran (-●-) and no treatment (-■-) in 2010. (*significantly different at $p = 0.05$. DAT, days after transplanting.)

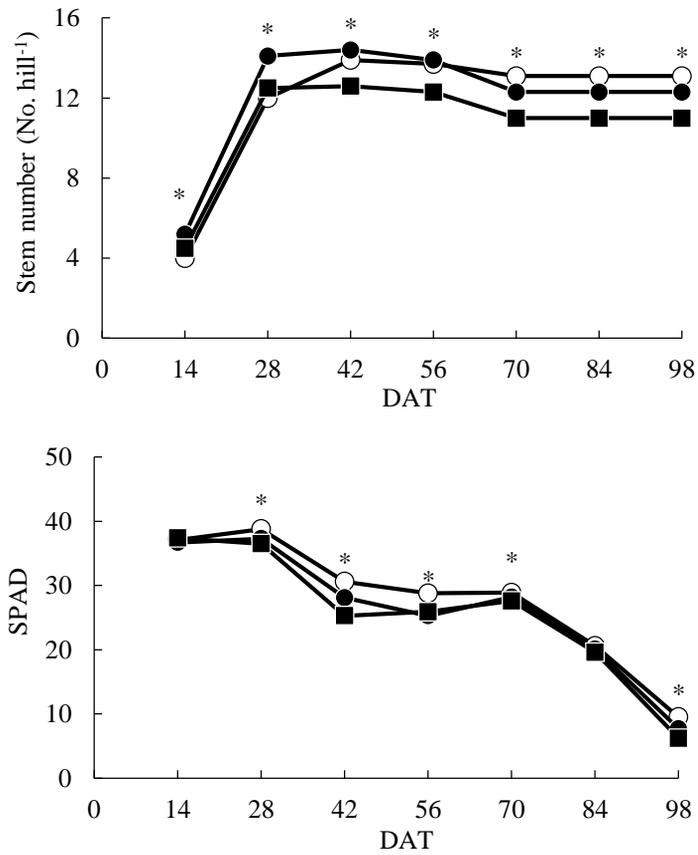


Fig. 2a. The growth of Akamai under *S. polyrhiza* (-○-), rice bran (-●-) and no treatment (-■-) in 2010. (*significantly different at p = 0.05. DAT, days after transplanting.)

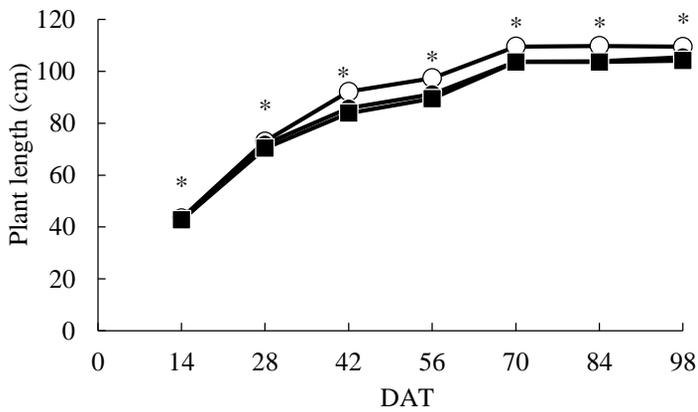


Fig. 2b. Growth of Kichinkokumai under *S. polyrhiza* (-○-), rice bran (-●-) and no treatment (-■-) in 2010. (*significantly different at p = 0.05. DAT, days after transplanting.)

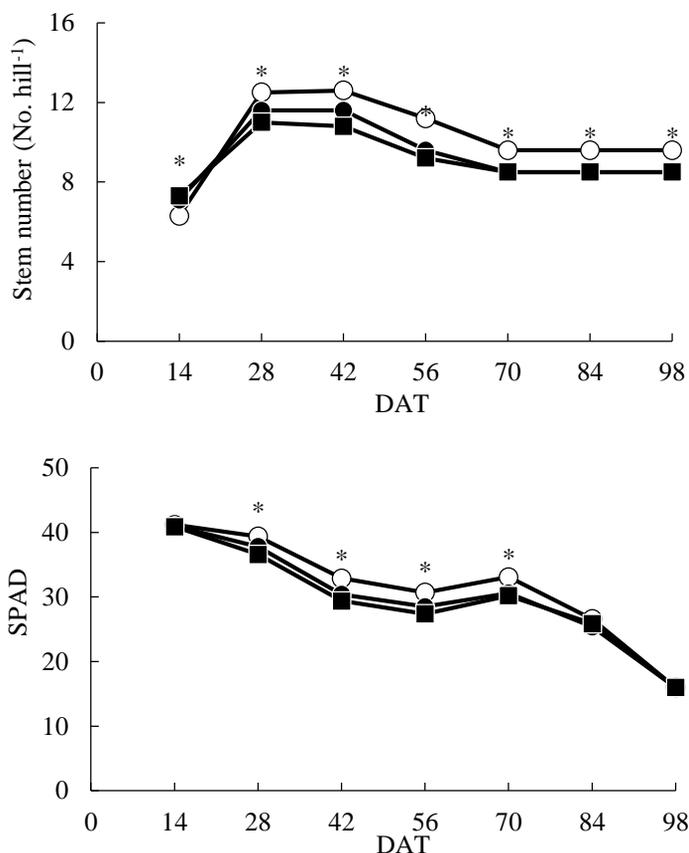


Fig. 2b. Growth of Kichinkokumai under *S. polyrhiza* (-○-), rice bran (-●-) and no treatment (-■-) in 2010. (*significantly different at $p = 0.05$. DAT, days after transplanting.)

Incremental rice growth attributed to *S. polyrhiza* might have resulted from the release of nutrients through *S. polyrhiza* decomposition. Li *et al.* (2009) showed that N released from duckweed through decomposition was returned to the floodwater or soil. Moreover, Duong and Tiedje (1985) mentioned that *S. polyrhiza* fixed N in their leaves via cyanobacteria attached to the lower epidermis or the reproductive pockets of the leaves. The application of the non-living mulch, rice bran, increased rice growth because it contains oil, proteins, vitamins, enzymes, essential minerals, microorganisms, and natural toxicant constituents (Barber and de Barber, 1980), which are released to the floodwater and soil. Furthermore, the quantity of *S. polyrhiza* affected rice growth by 68% in Akamai (Fig. 3a) and by 65% in Kichinkokumai (Fig. 3b), with the biomass of *S. polyrhiza* being inversely proportional to the plant length. The biomass of *S. polyrhiza* significantly affected the number of stems of Akamai (Fig. 3a), but of Kichinkokumai (Fig. 3b). The LAI of the rice leaf canopy was not significantly different among treatments from 14–56 DAT for both rice cultivars. However, the LAI trend after 14 DAT was slightly higher under the rice bran treatment; thereafter, the trend was higher under *S. polyrhiza* treatment (Fig. 4).

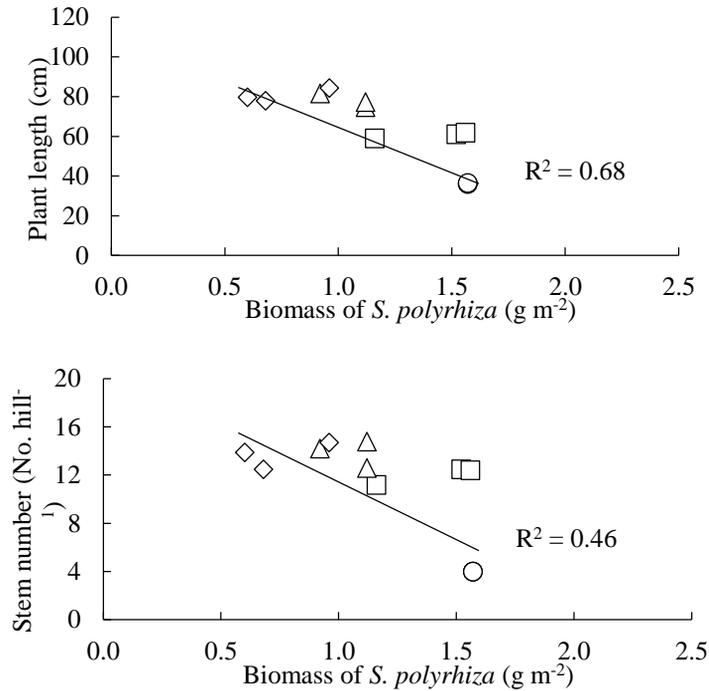


Fig. 3a. Correlation of the fresh weight biomass of *S. polyrhiza* with plant length and stem number in Akamai at 7 DAT (○), 21 DAT (□), 35 DAT (△), and 49 DAT (◇).

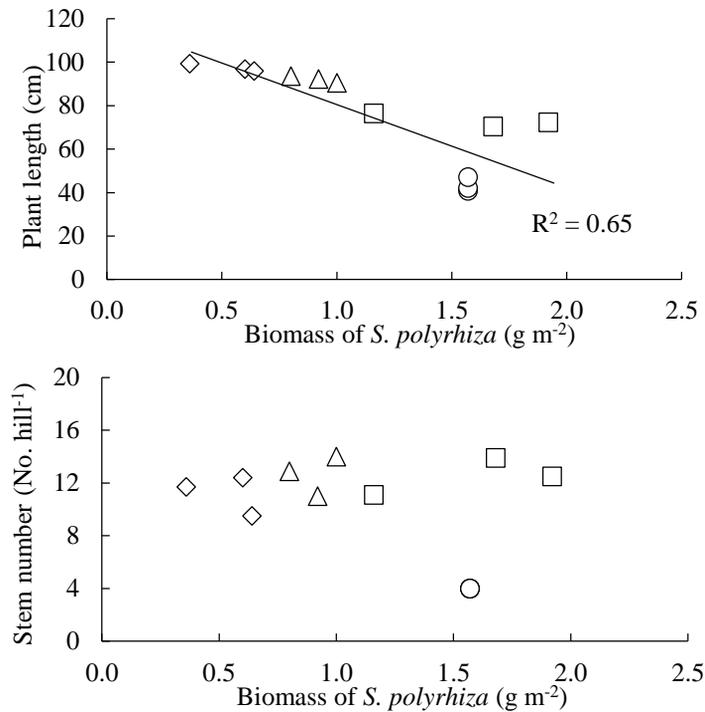


Fig. 3b. Correlation of the fresh weight biomass of *S. polyrhiza* with plant length and stem number in Kichinkokumai at 7 DAT (○), 21 DAT (□), 35 DAT (△), and 49 DAT (◇).

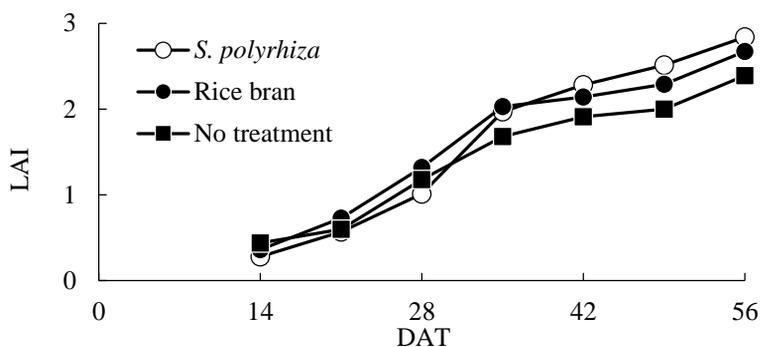


Fig. 4. Leaf area index (LAI) of the Akamai rice leaf canopy in 2010.

The effects of *S. polyrhiza* and rice bran on rice yield and yield components are shown in Table 1. The two cultivars differed in their response to the *S. polyrhiza* inoculation. We used two different cultivars, Akamai and Kichinkokumai, in this experiment because the polyculture system might be suitable in the local area where environmentally friendly agricultural practices are conducted. Both rice cultivars were glutinous rice. An old local landrace, Akamai is a redkerneled rice which is producing in Japanese local area. Kichinkokumai is a blackkerneled rice which was introduced from China. Both cultivars had premium prices based on their unique color and taste. Therefore, the historical trails of these cultivars are not clear, but the productivity of Kichinkokumai was less than that of Akamai due to the low number of panicles per hill in the former. Kichinkokumai is susceptible to the cold temperature (Yonekawa *et al.* 1999).

Table 1. Brown rice yield and yield components of Akamai and Kichinkokumai in 2009 and 2010.

Treatment	Panicle number (No. m ⁻²)	Grain number (No. panicle ⁻¹)	Percentage of ripened grains (%)	1000-grain weight (g)	Brown rice yield (g m ⁻²)
Akamai, 2009					
<i>S. polyrhiza</i>	321 a	119 a	89.0 a	18.6 a	479 a (92)
No treatment	342 a	108 a	90.1 a	19.0 a	523 a (100)
Kichinkokumai, 2009					
<i>S. polyrhiza</i>	227 a	111 a	46.9 a	23.2 b	276 a (89)
No treatment	215 a	108 a	57.6 a	23.4 a	312 a (100)
Akamai, 2010					
<i>S. polyrhiza</i>	272 a	106 a	92.2 a	17.4 a	460 a (113)
Rice bran	261 a	97 a	90.1 a	17.6 a	404 a (100)
No treatment	240 a	91 a	91.3 a	20.6 a	406 a (100)
Kichinkokumai, 2010					
<i>S. polyrhiza</i>	206 a	119 a	17.6 b	22.6 a	95 b (44)
Rice bran	187 a	107 a	32.8 ab	23.1 a	151 ab (70)
No treatment	184 a	101 a	51.4 a	23.2 a	217 a (100)

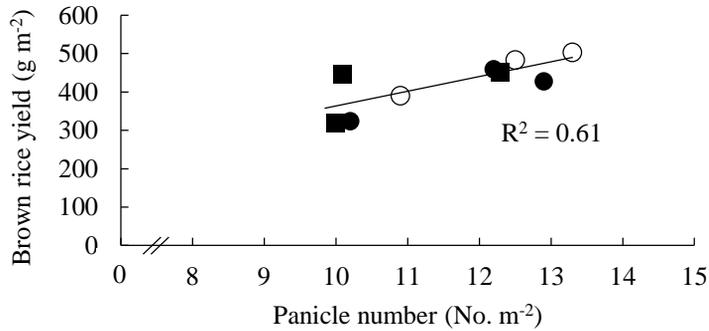
Notes: 1) The values with the same letter within each column are not significantly different at $p = 0.05$ by t-test (2009) and Tukey's HSD Test (2010).

2) The number in the parentheses means the percentage to no treatment.

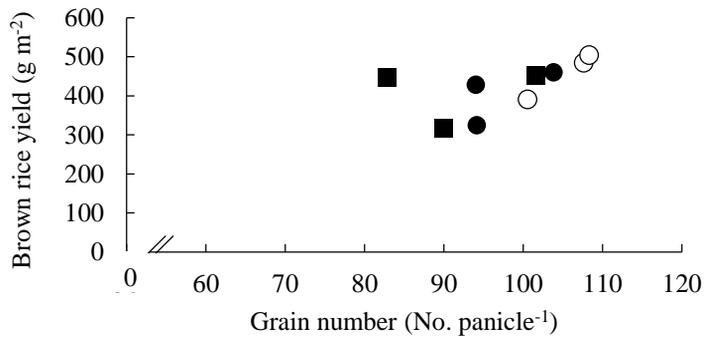
In 2009, inoculation by *S. polyrhiza* did not have an effect on the yield and yield components of the two rice cultivars, with the exception of the 1000-grain weight of Kichinkokumai. In 2010,

inoculation by *S. polyrhiza* affected all yield components. Some of differences were not statistically significant, but the brown rice yield of Akamai increased by 13% with *S. polyrhiza* inoculation. The yield of Kichinkokumai decreased by 56% due to a significantly low percentage of ripened grain. The Akamai brown rice yield was correlated with panicle number ($R^2 = 0.61$; Fig. 5). Rice bran application did not affect the brown rice yield, although some yield component parameters increased, according to Xuan *et al.* (2003).

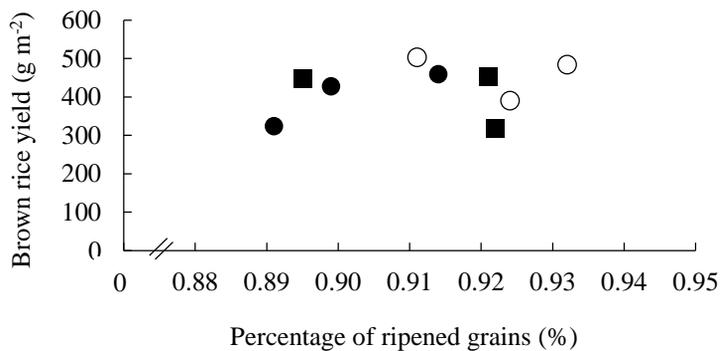
a) Panicle number



b) Grain number



c) Percentage of ripened grains



d) 1000-grain weight

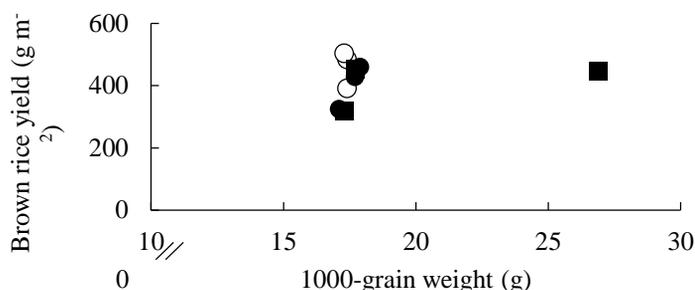


Fig. 5. Correlation of brown rice yield with panicle number (a), grain number (b), percentage of ripened grains (c), and 1000-grain weight (d) in Akamai under *S. polyrhiza* (○), rice bran (●), and no treatment (■).

Effect of Living Mulch on Weed Growth

The number of weeds was recorded during the growing season (Table 2). The day before applying treatments, the number of weeds did not differ significantly among treatment plots. Two weeks after inoculation, the number of weeds rapidly decreased in both rice fields by 85% and 96% for Akamai and Kichinkokumai, respectively. The suppression of weed number by *S. polyrhiza* continued up to 6 weeks after inoculation (WAI).

Table 2. Effect of *S. polyrhiza* and rice bran on the number of weeds (No. m⁻²) in Akamai and Kichinkokumai in 2010.

Treatment	0 WAI	2 WAI	4 WAI	6 WAI
Akamai				
<i>S. polyrhiza</i>	5603 a (97)	889 b (15)	1256 b (22)	1011 b (18)
Rice bran	5570 a (96)	5303 a (92)	5392 a (95)	5211 a (93)
No treatment	5800 a (100)	5788 a (100)	5656 a (100)	5586 a (100)
Kichinkokumai				
<i>S. polyrhiza</i>	6308 a (108)	203 b (4)	303 b (5)	311 b (6)
Rice bran	6667 a (114)	5319 a (95)	5126 a (90)	5367 a (95)
No treatment	5853 a (100)	5614 a (100)	5718 a (100)	5641 a (100)

Notes: 1) The values with the same letter within each column are not significantly different at $p = 0.05$ by Tukey's HSD Test.

2) The number in the parentheses means the percentage to no treatment.

3) WAI means week after inoculation.

The mechanism of weed suppression by *S. polyrhiza* was based on the sunlight-blocking action of the living mulch mat (Lumpkin and Plucknett, 1980). The number of weeds in rice bran was decreased by 5–8% at 2 WAI, but it was not significantly different from the control. The decline in the number of weeds might have depended on the distribution of rice bran, allelochemicals (Nakai and Toritsuka, 2009; Xuan *et al.*, 2003), and the amount of rice bran (Nakai and Toritsuka, 2009) on the soil. Therefore, the amount of rice bran applied should be increased when it is scattered on the water surface.

The reduction in weed growth at rice maturity is shown in Table 3. *S. polyrhiza* caused a reduction in the number of weeds and the dry weight of weeds in both rice fields and during both years, especially in 2010. The number and dry weight of weeds were reduced by 98% and 96%, respectively, in Kichinkokumai, although weeds in Akamai were reduced by only 3% and weed biomass by 50% in the *S. polyrhiza* plot. Rice bran application showed low ability to control weed growth as compared with *S. polyrhiza* in both fields.

Table 3. Effect of *S. polyrhiza* and rice bran on weed growth at the maturity stage of Akamai and Kichinkokumai in 2009 and 2010.

Treatment	Akamai		Kichinkokumai	
	Number (No. m ⁻²)	Dry weight (g m ⁻²)	Number (No. m ⁻²)	Dry weight (g m ⁻²)
2009				
<i>S. polyrhiza</i>	55 a (152)	10 a (89)	58 a (21)	8 b (63)
No treatment	36 a (100)	12 a (100)	281 a (100)	12 a (100)
2010				
<i>S. polyrhiza</i>	1207 a (97)	68 a (50)	78 a (2)	3a a (4)
Rice bran	1507 a (121)	98 a (72)	2163 a (59)	71 a (99)
No treatment	1241 a (100)	136 a (100)	3700 a (100)	72 a (100)

Notes: 1) The values with the same letter within each column are not significantly different at p = 0.05 by t-test (2009) or Tukey’s HSD Test (2010).

2) The value in the parentheses means the percentage to no treatment.

Environmental Conditions

Inoculation with *S. polyrhiza* and rice bran affected the environmental conditions of the rice paddy. Roger (1996) reported that the highest temperatures in the rice field ecosystem usually occur in the water. Therefore, inoculation with living mulch on the water surface reduced the water temperature by 0.9–2.1°C and reduced heat during the daytime (Li *et al.*, 2009). Inoculating with *S. polyrhiza* significantly reduced the cumulative water temperature, by 3–15%, compared to the control (Table 4), whereas rice bran had no effect.

Table 4. Cumulative water temperature (°C) from June 22 to August 9, 2010.

Treatment	Akamai	Kichinkokumai
<i>S. polyrhiza</i>	1333 c (85)	1310 c (97)
Rice bran	1388 a (101)	1340 b (100)
No treatment	1379 b (100)	1346 a (100)

Notes: 1) The values with the same letter within each column are not significantly different at p = 0.05 by Tukey’s HSD Test.

2) The value in the parentheses means the percentage to no treatment.

During the first 21 days, the maximum temperature was recorded on June 28 (13 DAT). Temperature changes during the day started at 8:00, the temperature rapidly increased until 15:00, and then it slightly decreased in the evening (Fig. 6). The highest *S. polyrhiza* treatment temperature was 6°C at 15:00. Solar radiation was reduced by 15% in Akamai and by 12% in Kichinkokumai with *S. polyrhiza* inoculation (Table 5). Solar radiation is defined as the radiation that reaches the soil surface and penetrates the canopy of rice plants, water layer, and living mulch. Therefore, the mechanism by which floating plants reduce solar radiation is that they act as a floating mat, absorbing the radiation and reducing light intensity (Li *et al.*, 2009; Lumpkin and Plucknett, 1980).

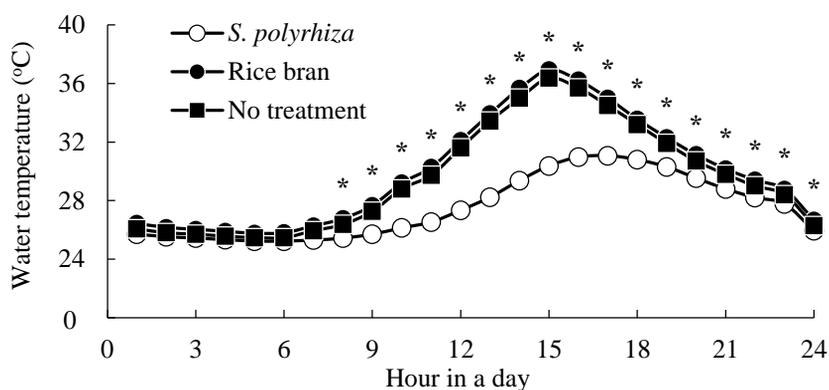


Fig. 6. The effect of *S. polyrhiza* and rice bran on water temperature on the day of maximum temperature (June 28, 2010 [13 DAT]) for Akamai.

Table 5. Cumulative solar radiation (MJ m⁻²) from June 29 to August 31, 2010.

Treatment	Akamai		Kichinkokumai	
<i>S. polyrhiza</i>	1096	c (85)	1071	c (88)
Rice bran	1197	b (98)	1168	b (95)
No treatment	1222	a (100)	1225	a (100)

Notes: 1) The values with the same letter within each column are not significantly different at p = 0.05 by Tukey's HSD Test.

2) The value in the parentheses means the percentage to no treatment.

CONCLUSION

The inoculation of living mulch (*S. polyrhiza*) in this study, promoted rice growth and yield, especially for Akamai, by preventing weed growth. The reduction in weeds was related to decreased solar radiation, which was blocked by the *S. polyrhiza* mat, and the consequent decrease in water temperature. The results from this study support growing floating aquatic plants as living mulch. Therefore, the adoption of a suitable polyculture system of rice production and weed control in rice paddy fields should be pursued carefully, taking into account the characteristics of the rice and the living mulch, as well as cultural practices.

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**REPELLENCY OF MARIGOLD, *Tagetes erecta* L. (Asteraceae)
VOLATILE ORGANIC CHEMICALS TO EGGPLANT FRUIT AND SHOOT
BORER, *Leucinodes orbonalis* Guenee (Lepidoptera: Crambidae)**

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ABSTRACT

Insecticide use in eggplant is frequent due to the eggplant fruit and shoot borer (EFSB), *Leucinodes orbonalis* Guenee (Lepidoptera: Crambidae). Food safety, environmental and human health issues for applicators have been raised. There is a need to explore ecological approaches to reduce pesticide use. We tested the hypothesis that volatile organic chemicals from marigold flowers, *Tagetes erecta* L. (Asteraceae) disrupt host-finding behavior of EFSB. The methods include host-finding behavioral assays in cages and Y-tube olfactometric bioassays to identify volatile chemicals in marigold flowers that repel EFSB. Behavioral bioassays were conducted at the National Crop Protection Center, College of Agriculture, University of the Philippines Los Baños from 2010 to 2011. Olfactometric bioassays using volatile organic chemicals of marigold flowers were conducted in 2013-2014. Fewer female adult landings were observed on the host plant, eggplant when marigold was present than eggplant alone, in laboratory cage bioassays over a 24 period. The percentage of damaged eggplants with marigold was significantly lower than sole eggplant. The average percent of shoot damage per eggplant with marigold was likewise significantly lower than sole eggplant. The repellency of volatile organic chemicals emitted by marigold flowers was likewise demonstrated with citral and 1-dodecene showing 64-68% repellency to female EFSB. A 10- flower equivalent displayed 72% repellency. Field trials need to be conducted to evaluate the appropriate cropping system that can effectively reduce EFSB populations.

Key words: host-habitat finding behavior, olfactory response, volatile organic compounds

INTRODUCTION

Eggplant, *Solanum melongena* L. (Solanaceae) ranked first in area, volume and value of production among the vegetables commercially grown in the Philippines (BAS, 2012). It is host to several major insect pests including whitefly, leafhopper, thrips and eggplant fruit and shoot borer (EFSB), *Leucinodes orbonalis* Guenee (Lepidoptera: Crambidae) the most destructive, bores into flowers, leaf petioles, shoots and fruits thereby adversely affecting plant vigor, yield and fruit quality resulting in yield reduction of as high as 70% (Navasero and Calilung, 1990; Islam and Karim, 1991; Dhandapani *et al.*, 2003). Pest management recommendation for EFSB accepted in South and Southeast Asia is the integration of the use of resistant variety, sex pheromone, cultural control and chemical insecticides (Srinivasan, 2008). In the absence of commercially viable resistant varieties, the use of sex pheromone and only a few available alternative cultural management tactics gave a level of control unacceptable to producers which make them more and largely dependent on the use of insecticides. In the Philippines, insecticide application was about 56 times during a cropping season equivalent to about 41 L in volume about 20 years ago (Gapud and Canapi, 1994; Orden *et al.*, 1994).

Other cultural management practices should be tested to make available alternatives to chemical pesticides, such as intercropping and companion planting.

Host-plant location activity is crucial for phytophagous insects to fulfill their nutritional requirements and to find a suitable oviposition site (Bruce *et al.*, 2006). Insects use plant chemical cues for host-finding activity. However, plant volatiles have also been demonstrated to protect plants by attracting herbivore enemies, including parasitic wasps, predatory arthropods and possibly even insectivorous birds (Unsicker *et al.*, 2009). Beneficial organisms and herbivores are attracted by chemical cues. Benzaldehyde and phenylacetaldehyde attract both pollinators and florivores while other compounds attract either pollinators or florivores (Theis, 2006). Syrphid flies use olfactory cues but not visual cues to find a pollen/nectar host-plant. Among the compounds eliciting an antennal response are methyl salicylate and 2-phenylethanol (Primante and Dötterl, 2010).

Floral volatiles serve as attractants for species-specific pollinators whereas the volatiles emitted from vegetative parts, especially those released after herbivory, appear to protect plants by deterring herbivores and by attracting the enemies of herbivores (Pichersky and Gershenzon, 2002). *Mythimna separata* Walker (Lepidoptera: Noctuidae) caterpillars utilize plant volatile information to sense their environment and modulate their daily activity patterns, thereby potentially avoiding the threat of parasitism (Shiojiri *et al.*, 2006). Intercropping of crop plants is common in the tropics with effects on pest population dynamics that minimise crop damage (Perrin and Phillips, 1978). Intercropping also ensures food security and additional income to farmers. Surrounding plants of field crops that are attractive to natural enemies, may be a good option to farmers, since this practice could lead to increased diversity of arthropod species and, consequently, the natural control of pest populations.

Marigold, *Tagetes erecta* L.(Asteraceae) is a versatile plant with potential for pest management. Marigold rows next to onion fields resulted in higher number of entomophagous species, potentially enhancing the natural control of onion pests and providing an alternative to crop sprays for organic control of onion pests (Silveira *et al.*, 2009). Marigold also possesses nematode suppressive potential that should be explored further as a green cover crop. Marigold is well known for its ability to produce compounds such as α -terthienyl that are allelopathic to many species of plant-parasitic nematodes (Hooks *et al.*, 2010). Intercropping with marigold induced a significant reduction in tomato early blight caused by *Alternaria solani*, by means of allelopathy, altering the microclimatic conditions around the canopy and acting as a physical barrier against conidia spreading (Go´mez-Rodr´ıguez *et al.*, 2003). Marigold oil also possesses a significant but limited and dose-related antifungal and insecticidal activity to white termite, *Odontotermes obesus* Rhamb. (Isoptera: Termitidae) (Singh *et al.*, 2003).

Intercropping eggplant with radish or basil has the potential for insect pest management as preliminary studies conducted in the field showed reduced fruit damage (Navasero and Calumpang, 2013; Gonzales *et al.*, 2004). Intercropping of lemon grass in eggplant resulted in reduction in leafhopper, whitefly and aphid counts up to 10 weeks after transplanting. This resulted in significant differences in average number of damaged shoots for eggplant intercropped with lemon grass (4.75) and without lemon grass (6.25) (Calumpang *et al.*, 2013).

It is important to explore other strategies that can contribute to reducing insect pest pressure and reliance on insecticides. The impact of marigold on insect pest behavior when grown together with eggplant has not been evaluated adequately; there remains a need to conduct more basic studies to provide the basis for the possible utilization of marigold as an intercrop. This strategy would increase the versatility and importance of marigold in vegetable production because it holds the potential for addressing both disease and insect pest problems.

In the current study, we examined the influence of marigold on the host-finding and oviposition behavior of eggplant fruit shoot borer, *L. orbonalis* in laboratory cage experiments. Furthermore, we determined the volatile organic chemical (VOC) profile of marigold floral headspace and related these chemicals to the olfactory response of *L. orbonalis*.

MATERIALS AND METHODS

Insects

Eggplant fruit and shoot borer, *L. orbonalis* larvae were collected from infested eggplant fruits in Calamba, Laguna, Philippines. These insects were reared in the laboratory in plastic pans lined with paper towel and fed daily with chips of eggplant fruits under controlled temperature (28°C), L16:D8 photoperiod and relative humidity (70-80%). Pupae were subsequently collected and placed in Petri plates for holding. When ready to emerge, pupae were placed in a cage and transferred to egg-laying chambers for mating and oviposition. The egg-laying chamber was a glass ball jar with mesh cloth strips for oviposition. A cotton ball soaked in 10 percent sucrose, was hung on a wire attached to the mesh cloth cover, and offered as adult food. After 3-4 days, adults were removed and the mesh cloth strips with egg masses were transferred to eggplant chips for further rearing according to the method described by Gonzales (1999).

Host finding behavior and damage potential bioassays

The response of eggplant fruit shoot borer (EFSB) adults was observed in screen cages using potted eggplant (flowering stage) and marigold in choice and no-choice tests in laboratory from 2010 to 2011. One potted eggplant and one potted marigold plant were placed inside wire screen cages (75x75x150 cm) after which, 2 males and 2 females of the laboratory reared adult moths were released into the cage. The behavior of these moths was monitored by recording whether they alighted on the plant or the cage at 0, 1, 2, 3, 4, 5, 6, 7 and 24 h after introduction under dark room conditions, with 3 h artificial light prior to 24 h readings. Controls were exposed to 1 potted eggplant per cage in a separate room from the choice test setups. The assays were conducted under ambient room conditions (32 °C; 70–80% RH). The adults were used only once. Each cage setup represented one replicate and the experiment was replicated 25 times with the adults used only once.

Shoot damage was monitored and assessed two weeks after introduction. The number of damaged shoots was counted and percent damage computed based on total number of shoots per plant.

Collection of Volatile Organic Chemicals (VOCs)

Marigold plants were grown and maintained at the National Crop Protection Center (NCPC), College of Agriculture, University of the Philippines Los Baños. Marigold flowers, 100 pieces in full bloom, were collected early in the morning and the VOCs were collected by trapping using 2 g Tenax TA (Supelco, USA) under vacuum using a mini-pump (Sibata MP 1, Japan) at a flow rate of 2.5 L/min for 4 h at room temperature. This was followed by Soxhlet extraction in *n*-pentane AR (JT Baker, USA) and concentrated using a micro Kuderna Danish set up.

GC-MS Analysis

GC-MS analysis was conducted at the Natural Science Research Institute, University of the Philippines Diliman, Quezon City. Separation of the collected VOC mixture was achieved using a 5% phenyl methyl polysiloxane capillary column (30m x 0.25mm i.d x 0.5 um film thickness) installed in a Shimadzu 2010 gas chromatograph- mass spectrometer (GC-MS). Extracts were analyzed with the following method: 1 uL injections; injector temperature, 250 °C; splitless mode; oven temperature, 60

°C for 3 mins; programmed at 5 °C/min to 250 °C and held for 5 mins; electron impact mode (70 eV; carrier gas, helium. Tentative identifications were made by comparison of the spectra with authentic standard materials and mass spectral database (NIST107.Lib).

Y-tube olfactometric bioassay

The responses of female EFSB to VOCs found in marigold flowers were assayed in Y-tube olfactometer as described in Calumpang and Navasero (2013) in an air-conditioned room (27-28 °C, 52-59 % relative humidity). Charcoal filtered airstream was supplied to each arm of the olfactometer using an electric mini-pump (Shibata MP-2N, Japan), passing through the odor chamber with a filter paper dosed with the test chemical and the other arm had a filter paper dosed with *n*-hexane, serving as the control. The bioassay was conducted using 1 day-old mated adult females prior to the experiment. Within 10 mins, the first choice of the EFSB between the two arms of the Y-tube olfactometer was noted and considered repelled if it entered the control arm. Unresponsive adults were not considered. The set-up was washed with mild detergent, rinsed twice with technical acetone and allowed to air-dry. The adults were used only once; one adult was used per replicate with 25 replicates per treatment. Percent repellency was determined based on the number of individuals responding negatively, divided by the total number of individuals tested and multiplied by 100.

Statistical Analysis

The data obtained in the host-finding bioassay was analysed statistically by using the *t*-test: Two-sample unequal variances (t critical 2-tail) at 5% significance level.

RESULTS AND DISCUSSION

Host-finding behavior laboratory bioassays

Basic studies on EFSB behavior were conducted to elucidate the mechanism for the reduced counts in eggplant-marigold intercrop in preliminary small plot field trials at NCPC, UPLB as well as crop diversification trials conducted by other researchers (Sujayanand *et al.*, 2015). Oviposition preference bioassays conducted under laboratory conditions revealed that EFSB adult females constantly landed on the host plant, eggplant leaves and shoots immediately after introduction, with 1 adult spending about 1-2 mins on the leaves within the first 7 h. The number increased to 1.8 adults per plant after 24 and 48 h after introduction (Fig. 1) but this was reduced to 0.4 to 0.5 adult per eggplant when marigold was present (Fig. 2). The cage experiments were a valuable tool for evaluating the potential of repellents to control eggplant fruit and shoot borer.

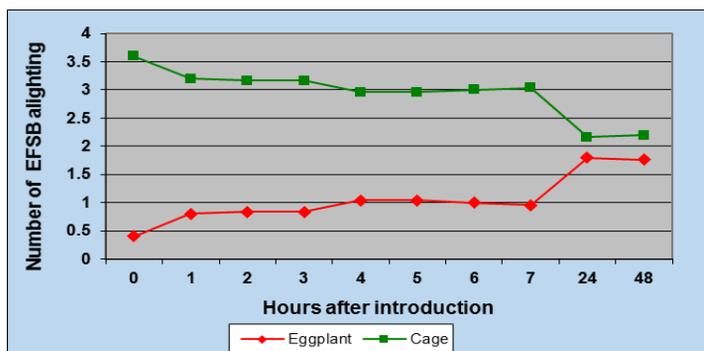


Fig.1. Number of eggplant fruit and shoot borer female adults alighting on eggplant in cage experiments

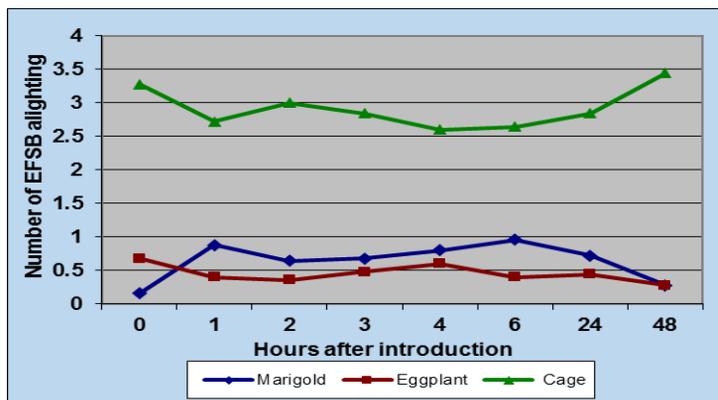


Fig. 2. Number of eggplant fruit and shoot borer female adults alighting on eggplant with marigold (1:1) in cage experiments

Damage potential laboratory bioassays

The percentage of damaged eggplants with marigold (36%) was significantly lower than sole eggplant (84%). Interestingly, we noted that the average percent shoot damage per eggplant with marigold was also significantly lower (12%) than sole eggplant (38%) (Table 1).

Table 1. Mean percent shoot damage per plant from behavioral assays in cages.

Treatment	Percent of Replicates with damage *	% Shoot Damage per plant*
1. Eggplant alone	84 a	38 a
2. Eggplant with marigold	36 b	12 b

*Means of 25 replicates (1 plant per replicate); Means in a column followed by a different letter are significantly different at 0.05 probability level using *t*-test : two sample assuming equal variances (*t* critical 2-tail)

Our behavioral and olfactory bioassay results are consistent with crop diversification field trials of eggplant with marigold intercrop and maize as border crop showing significantly lower percentage fruit infestation by *L. orbonalis* than eggplant alone. In addition, leafhopper and white fly populations were reduced (Sujayanand *et al.*, 2015).

Similar results using other herbs were reported recently. Intercropping eggplant with lemon grass or radish likewise reduced hoppers, aphids and whiteflies counts, shoot damage and increased the yield of eggplant in small plot field trials (Calumpang *et al.*, 2013; Navasero and Calumpang 2013). Coriander and fennel were equally effective in reducing *L. orbonalis* infestation when intercropped with eggplant. Intercropping reduced the fruit damage to 12–15% compared with the plots with eggplant monoculture (Satpathy and Mishra, 2011). This strategy has the potential to reduce farmer expense for insecticides to manage these insect pests

GC-MS analysis of marigold floral volatile organic chemicals

The presence of α -pinene, citral, 4-terpineol, limonene, 1-dodecene, 1-tetradecene and 1,8 cineole in marigold floral volatile extracts was demonstrated in the headspace of marigold flowers (Fig. 3). These are volatile organic chemicals, some of which are bioactive against insect pests (Bruce *et al.*, 2002; Ukeh *et al.*, 2009; Nerio *et al.*, 2010).

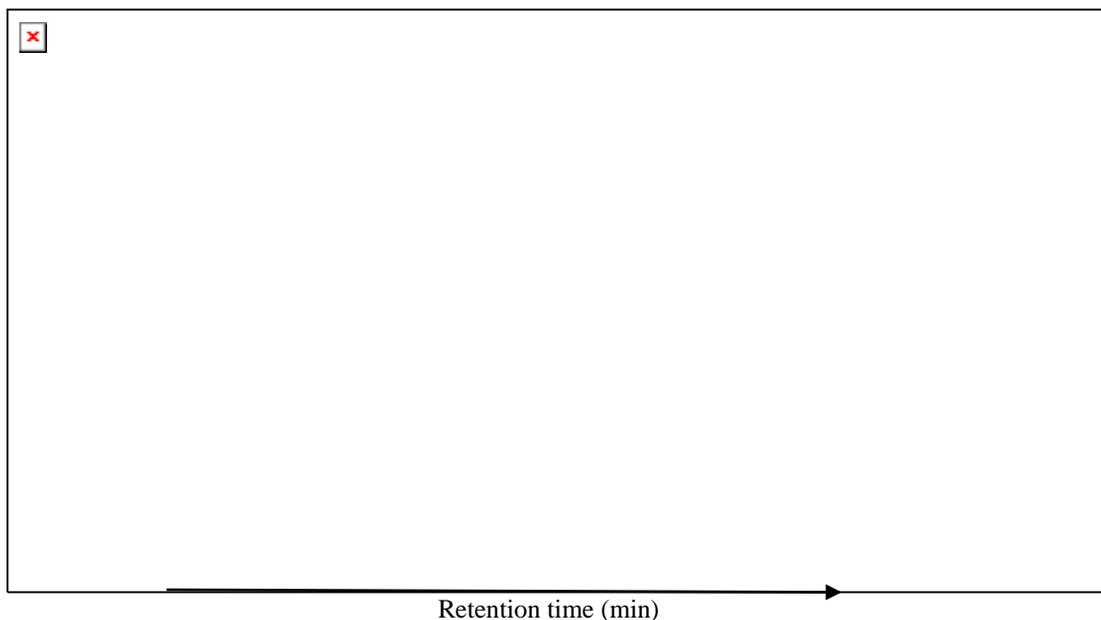


Fig. 3. Total ion chromatogram (TIC) obtained by GC-MS analysis of volatile organic chemicals found in marigold flower headspace.

African marigold, *T. erecta* floral odors, such as benzaldehyde (\pm)-linalool, phenylacetaldehyde and (S)-(-)-limonene were found attractive to cotton bollworm, *Helicoverpa armigera*. Alternatively, marigold flower oil has been characterized by the presence of 1, 8-cineole, α -pinene, limonene, α -terpineol, piperitone, piperitenone, piperitenone oxide and sabinene as the major compounds (Ogunwande and Olawore, 2006; Krishna *et al.*, 2004).

Olfactory response of EFSB to marigold floral volatile chemicals

The repellency of some volatile organic chemicals found in marigold flowers was demonstrated for eggplant fruit and shoot borer females (Fig. 4). 1-Dodecene and citral demonstrated 68% and 64% repellency, respectively. These chemicals could be part of the chemical cues blend received by eggplant fruit and shoot borer causing it to spend less time on eggplant, based on our behavioral assays. A 10- flower equivalent displayed 72% repellency.

The other volatile organic chemicals did not display activity when tested singly. However, headspace volatile organic chemicals emitted by plants (which act as chemical cues) are usually a combination that are found in varying ratios in flowering plants. Insects usually rely on ubiquitous plant volatiles and not those taxonomically characteristic of host plants as host odor cues. However, such an odor coding system has the advantage of providing flexibility; it allows for adaptation to changing environments by alterations in signal processing while maintaining the same peripheral olfactory receptors (Bruce and Pickett, 2011).



Fig. 4. Percent repellency of some marigold floral volatile organic constituents to eggplant fruit and shoot borer.

1- Dodecene is found naturally in plants and is known to be effective as a deterrent or controlling mechanism for insect pests of different orders such as: Lepidoptera, Coleoptera and Orthoptera. 1- dodecene and 3-tetradecene emitted by soybean leaves (PI 227687) were found repellent to cabbage looper and Mexican bean beetle (Liu et al, 1988 and 1989). Meanwhile, citral from natural sources is a mixture of two geometric isomers geranial and neral. As a component of ginger, *Zingiber officinale* (Zingiberaceae), citral showed significant repellency against the maize weevil, *Sitophilus zeamais* together with 1,8-cineole (Ukeh et al 2009). Limonene, α -pinene and 1,8 cineole are isoprenoids which produces electrophysiological responses in several Lepidopteran, Hemipteran and Coleopteran insect pests (Burguiere et al 2001; Ibrahim et al 2001).

Chemical investigations on the leaf essential oil of *Tagetes erecta*, performed by HPLC, GC and GC-MS techniques, showed the presence of 26 components, accounting for 89% of the total oil. The major constituents were (Z)- β -ocimene (42.2%), dihydrotagetone (14.3%), (Z)-tagetone (8.3%), limonene (7.3%), (E)-ocimenone (6.1%) and (Z)-ocimenone (5.3%). The leaf oil of marigold, which is rich in (Z)- β -ocimene, has a statistically significant antifungal and insecticidal activity (Singh *et al.*, 2003).

CONCLUSION

Experimental evidence for the role of olfaction in host-plant selection behavior and repellency of marigold to *L. orbonalis* has been obtained in the current study. Non-host volatiles may be exploited for control of EFSB by disrupting both mate- and host-finding behavior. This information is useful in developing intercropping or diversified cropping systems for eggplant pest management.

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YIELD AND QUALITY OF TWO KENAF VARIETIES AS AFFECTED BY HARVESTING AGE

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ABSTRACT

Kenaf (*Hibiscus cannabinus* L.) is grown either as an animal feed or as a source of fibre. In Malaysia, the variety V 36 has been planted since 2001 and currently a new variety MHC 123 is being evaluated. This experiment was conducted at the Malaysian Agricultural Research and Development Institute (MARDI) Serdang, (latitude N 3° 17', longitude E 101° 46') from August to December 2012 to determine the optimum harvest age for the two varieties of kenaf for animal feed and for fibre production. The two varieties were harvested at 8, 12, 16 and 20 weeks after planting (WAP). Mean biomass yield of MHC 123 (11.7 t ha⁻¹) was significantly higher (p<0.05) compared to V 36 (8.7 t ha⁻¹). The crude protein (CP) of MHC 123 decreased slowly from 18.06 to 17.22% while CP of V 36 declined rapidly from 21.72 to 11.32% between 8 and 12 WAP. Acid detergent fibre content in MHC 123 increased slowly (31.72 to 36.86%) compared to V 36 (39.57 to 55.59%) from 8 to 12 WAP. From these findings, MHC should be harvested at 12 WAP while V36 should be harvested at 8 WAP for use as forage. Tests on the fibre showed that the tensile strength for both varieties was highest at 16 WAP and both varieties should be harvested at this age for fibre production.

Key words: kenaf, animal feed, fibre, harvest dates

INTRODUCTION

Kenaf (*Hibiscus cannabinus*) has become an important industrial crop in Malaysia as a source of natural fiber for industrial purposes such as for making ropes and sacks and recently as composite building materials. It has been viewed as potential alternative to replace tobacco as a cash crop (Wong *et al.*, 2008). Kenaf was introduced in Malaysia in early 2000 mainly for animal feed. It was usually harvested at an early stage (6-10 weeks) before the start of the reproductive stage. The crude protein (CP) in kenaf is between 21-34% in leaves, 10-12% in stalk and 16-23% in the whole plant (Mat Daham *et al.*, 2006). The CP content of about 20.9% is comparable to white lead tree (*Leucaena leucocephala*) and asystasia (*Asystasia intrusa*). At the young stage of growth (4 weeks), kenaf CP content can reach 28-30% and crude fibre (CF) is less than 20%. However, at 8 weeks cutting, the CP decreased to 16% and CF increased to 36.6% (Wong *et al.*, 2008). Liang *et al.*, (2003) suggested that for the production of high protein feed, suitable age of cutting was between 6-8 weeks. The profile of amino acid of kenaf is similar to alfalfa hay but lower than that of soybean meal. The content of hemicelluloses is about 15%. Tannin content in kenaf is between 1.69-1.97 mg g⁻¹ and these values are lower than those found in oil palm frond or white lead tree (Wan Zahari *et al.*, 2003). Total digestible nutrients (TDN) and metabolisable energy (ME) in kenaf were sufficient to meet daily requirement of ruminant livestock. The average daily weight gain of growing Charoke beef cattle was about 0.66 to 0.71 kg when 30 to 20% kenaf was incorporated in a mixed ration, respectively.

Kenaf has a high growth rate, rising to a height of 4-6 m in about 4-5 months. The dry matter yield averages 6-10 t ha⁻¹ year⁻¹ but new varieties may reach up to 30 t ha⁻¹ year⁻¹. It is similar to jute in many of its properties and may be used either as an alternative, or in admixture with jute. Kenaf has several uses; traditionally, the main interest is its bast fibres, derived from the phloem, but recently the core fibres (xylem and pith) have also been proven useful for industrial application. Bast fibre can be used to make various high quality products such as composite product for building and automotive material. Kenaf fibre is also a replacement for glass fibre and other synthetic fibre. The core fibre can be used directly as a bioremediation agent, animal bedding, low density particle board, soil amendment, oil absorbent in chemical industry and in ethanol manufacture (Sellers *et al.*, 1999; Agbajeet *et al.*, 2008). In the United States kenaf has been planted for paper products (newsprint, bond paper and corrugated liner board) and also for building material (Charles *et al.*, 2002) while in China kenaf is used in the textile industry. Bangladesh, Thailand and Vietnam also produce kenaf but the usage of kenaf is for low value products like rope and sack. In Malaysia, kenaf is grown commercially in many states including Kelantan, Terengganu, Pahang, Kedah and Perlis. From its initial use as an animal feed kenaf is currently grown for its biomass as a primary source for biocomposite products that are regarded as high value building materials.

Harvesting time is an important factor affecting kenaf properties (Zhou *et al.*, 1998). The suitable harvesting age for kenaf depends on the usage and kenaf variety. Various industries harvest the kenaf plants for products at different harvesting times during the plant life cycle. Normally for the purpose of producing fibre kenaf is harvested after the fibre matures (4-5 month after planting or after flower blooming) but for other uses of kenaf especially for forage, kenaf is harvested in 7-8 weeks after planting. This is due to high protein content in the younger plant (Wong *et al.*, 2003). Besides that, fibre from juvenile plants of jute and kenaf was reported to be 'silk-like', fine in texture, very flexible and thin. The yield could be lower but for an annual plant, possibly two crops can be harvested in one season to give the same yield of fibre but with much less lignin. The fibre which can be used for textile may command a higher price resulting from such early harvesting (Roger and James, 1999). For kenaf, lignin occurred in bast fibre at 30 days after planting (Sellers *et al.*, 1999). Immature plant may be low in lignin thus making it more suited for paper manufacture since there would be little chemical pulping required to remove the lignin. The age of kenaf at harvest can influence plant composition and protein content.

Kenaf has a good quality as a source of animal feed and fibre but the limiting factor is the low dry matter yield from the currently grown variety V 36. New varieties have to be introduced and one of the potential variety that is being studied is MHC 123. The growth period for each kenaf variety may be different, so information on harvesting age and yield component on MHC 123 are needed to clarify the ability of MHC 123 to replace V 36. Thus, the objective of this experiment was to determine the optimum harvesting age for MHC 123 and V 36 and to determine the effect of harvesting age on quality and yield of kenaf biomass for forage and fibre.

MATERIALS AND METHODS

The experimental design was a two-factor experiment arranged in a Randomized Complete Block Design (RCBD) with four replications. Treatments comprised of four harvest ages (8, 12, 16 and 20 weeks after planting (WAP)) and two varieties (V 36 and MHC 123). For the production of kenaf as animal feed, the data used were confined to two harvest ages: 8 and 12 WAP as the crop would be too mature as a forage beyond 12 weeks. On the other hand for fibre production the data used were restricted to the 16 and 20 weeks after planting as less mature plants are unsuitable for fibre. Planting material was obtained from Malaysian Agricultural Research Development Institute (MARDI) Serdang. Seeds of two varieties were directly sown into a well prepared site in rows. The row spacing was 35 cm x 5 cm as recommended by MARDI for kenaf V 36. The experiment was conducted at the Malaysian Agricultural Research and Development Institute (MARDI) Serdang, Research Station (latitude N

3°17', longitude E 101° 46'). The soil was ploughed on 1 August 2012 and harrowed a day after. The seeds were sown 2 weeks after ploughing. Planting depth was 2 cm-2.5 cm and sowing was done manually and the plants were thinned to the defined population density at 2 WAP. A basal compound fertilizer, NPK nitrophospka green (15:15:15) was band applied after sowing at the rate of 300 kg ha⁻¹. A subsequent application of 300 kg ha⁻¹ NPK nitrophospka blue special (12:12:17:2 + Te) was applied at 6 WAP. Weeds were manually controlled.

Measurements

Plant height

Plant height was measured with steel ruler from ground level to the highest shoot. The measurements were taken on 10 randomly marked plants from each plot every 2 weeks until harvest.

Stem diameter

Stem diameter was measured 2 cm above the soil using a digital calliper Mitutoyo every 2 weeks until harvest. Measurements were taken on 10 randomly marked plants from each plot.

Dry matter yield

Above ground dry matter yield was measured from four random samples taken from each plot using a 2 m x 1 m quadrat. Samples were harvested manually from each plot and were weighed and dried in the forced-air dryer at 60 °C until constant weight.

Leaf, stem yield and leaf to stem ratio

Kenaf was sampled by using a 0.3 m x 0.3 m quadrat (four replicates for each sampling) at 8 and 12 WAP. The harvested plants were cut at ground level and fresh weight was recorded. Leaves were removed from the stalks and were weighed separately, before and after the samples were oven dried at 50 °C until constant weight. Leaf to stem ratio of the plant is the ratio of the leaf biomass to the stem biomass.

Acid detergent fibre (ADF) and crude protein (CP)

Kenaf was sampled by using a 0.3 m x 0.3 m quadrat (four replicates for each sampling) at 8 and 12 WAP. The plants were harvested at ground level. The plants were oven dried at 50°C until constant weight. Dried samples were ground through 1 mm screen and analysed for CP and ADF content by using Near Infrared Reflectance Spectrophotometer (NIRS) (Brand FOSS, model no. N6500) using prior calibration specifically for kenaf. Acid detergent fibre measures the most indigestible component of the cell wall (mainly cellulose and lignin) and is negatively correlated with digestibility of animal feeds (Van Soest, 1967).

Number of days to flowering

The number of days to flowering was quantified from planting date to the day when the first 50% flower and 100% flower of the marked plants bloomed with white yellowish petals.

Fibre quality

Bast yield, core yield and bast and core ratio

Kenaf was sampled using a 0.3 m x 0.3 m quadrat (four samples for each plot) at 16 and 20 WAP. The bark (containing bastfibre) was peeled from each stalk section manually and both components (bast and core) were oven dried at 60°C until no further weight loss was observed. Weights were recorded before and after the drying process. Bast and core ratio represent the percentage by weight of bast compared with the total stalk weight.

Tensile strength test

A random sample of five plants were harvested from each plot and the bark was peeled manually by hand from each stalk to separate the bast and core. The bast section was soaked in water in a fibre tank until the fibres were separated from each other. The process is called the retting process. The retting process took about 30 days to be completed. The fibres were then oven dried at 60°C until constant weight. These fibres were used for the tensile and water absorption test.

Fibre bundle test was performed using an Instron machine 3366 with a crosshead speed of 5 mm/minute. The kenaf fibre samples were glued at rectangular cardboard with a dimension of 30 mm x 70 mm. The same weights of fibre were used for each sample. In this trial, weight of each sample was 0.0120g. The cardboard contained a circular hole right at the centre with a diameter of 20 mm. A detailed illustration of the sample is illustrated in Figure 1.

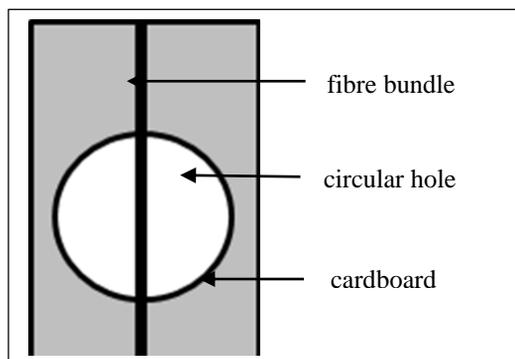


Fig. 1: Mounting card of fibre test piece

Water absorption

Bundles of kenaf fibre were immersed into distilled water which was kept in a covered bottle at room temperature. The moisture content was calculated using Equation 1. A fixed amount of fibre was weighed before and after immersing into water. The weight was taken using an electronic balance after removing the kenaf fibre from the water and wiping them dry. Weighing was done every day until a constant weight was achieved.

$$\text{Moisture (\%)} = \frac{W_t - W_0}{W_0} \times 100 \quad \text{Equation 1}$$

W_t = Weight after immersion

W₀ = Weight before immersion

Data analysis

Analysis of variance (ANOVA) was performed to examine the effect of harvest age and variety on the measured variables tested using SAS® version 9.2. When the effects were significant ($p < 0.05$), means were separated using least significant difference (LSD)

RESULTS AND DISCUSSION

Plant height and stem diameter

Plant height and stem diameter was monitored at 2-weekly intervals. Plant height and stem diameter increased ($P < 0.05$) as maturity advanced for both varieties (Table 1). MHC 123 showed a higher plant height and bigger stem diameter compared with V 36 and this was also reflected in the dry matter yield. The results of plant height and stem diameter from this experiment were considered normal for kenaf planting in the tropical area like Malaysia.

Table 1: Plant height and stem diameter

Treatment	Plant height (cm)	Stem diameter (mm)
Variety		
MHC 123	177.6a	14.36a
V 36	163.0b	12.68b
Week (W)		
W 2	26.2h	2.99h
W4	88.5g	8.33g
W6	141.2f	11.6f
W8	184.2e	14.2e
W10	219.3d	16.4d
W12	243.6c	18.5c
W14	252.8c	19.7bc
W16	270b	20.29b
W18	278b	22.57a
W 20	296a	23.1a
Significance level		
V	**	**
W	**	**
V x W	**	ns
Mean	170.3	13.5
CV	11.16	15.87

Means with the same letter were not significantly different ($P > 0.05$) using LSD.

Above ground biomass

The dry matter yield of kenaf is shown in Table 2. There were significant differences in dry matter yield (DMY) of kenaf between varieties and harvesting age. There was no interaction between harvesting age and variety, thus the discussion is focused on the main effects of varieties and harvesting age. The results showed that the DMY increased significantly ($P < 0.05$) from 1.71 kg plot⁻¹ at 8 weeks to 2.24 kg plot⁻¹ at 12 weeks. However no significant increase in dry matter yield was shown thereafter

at 16 and 20 weeks. Based on these values the extrapolated yield were 8.5, 11.2, 10.9 and 10.2 t ha⁻¹ at 8, 12, 16 and 20 weeks, respectively. The earliest harvest age at 8 weeks gave the lowest biomass because the plants were still actively growing. A similar finding was reported by Charles (1992) who found lower dry matter yield at 8 WAP (4.7 t ha⁻¹) compared with 12 WAP (7.5 t ha⁻¹) for varieties Everglades 41, Cuba 2032, Guatemala 4, Guatemala 45, Guatemala 48 and Guatemala 5. Similarly, Aminah *et al.* (2006) reported that dry matter yield of kenaf variety HC 13, Thai Kenaf, Tainung V 28, Tainung 2, Khonkaen 60 at 8 WAP ranged between 1 to 6 t ha⁻¹. Dry matter yield for forage in our study was considered high when compared to other varieties while for fibre it was similar with finding from Noor Syahira (2010) where dry matter yield for MHC 123 was 11 t ha⁻¹ at 16 WAP.

Table 2: Dry matter yield based on harvest age and variety

Treatment	Dry matter (kg plot⁻¹)	Yield (t ha⁻¹)
<u>Harvest age (H)</u>		
8 WAP	1.71b	8.5b
12 WAP	2.24a	11.2a
16 WAP	2.18a	10.9a
20 WAP	2.03a	10.2a
<u>Variety (V)</u>		
MHC 123	2.34a	11.7a
V 36	1.74b	8.7b
Significance level		
Harvest age	**	**
Variety	**	**
H x V	ns	ns
Mean	2.43	12.15
CV	13.66	13.66

Means with the same letter were not significantly different among harvest age and variety (p>0.05) using LSD** p<0.01, ns: not significant.

Dry matter yield of MHC 123 at 11.7 t ha⁻¹ was significantly higher than V 36 (8.7 t ha⁻¹). In Malaysia, DMY at maturity stage (16-20 WAP) ranged from 10-15 t ha⁻¹ (Nik Ablah, 2012). In well established BRIS fertile soil, yield can reach up to 20 t ha⁻¹ at maturity stage.

Number of days to flowering

MHC 123 was later flowering than V 36 (Table 3) which started flowering after 51 DAP and achieved 50 % flowering at 78 DAP followed by 100 % flowering at 103 DAP. V36 started to flower 10 days earlier than MHC 123 (41 DAP) and 50 % of total flowering at 67 DAP followed by 100 % flowering at 88 DAP. For kenaf, the crop is considered mature when 50% of flowers have bloomed. At maturity stage the kenaf growth rate started to slow down and the fibre becomes harder. Noor Syahira (2010) explained that the period of flowering determines the growth of kenaf. Full bloom at 0-30 DAP is considered as very early, 31-60 DAP as early, 61-90 DAP as intermediate and 91-120 DAP as late flowering variety. Thus, MHC 123 was classified as late flowering while V 36 was intermediate. Lateness in flowering gave an advantage to the MHC 123 by extending the period of accumulating biomass as kenaf growth rate was higher at the vegetative phase than the reproductive phase. Lateness in flowering was one of the reasons MHC 123 was greater in DMY compared to V 36. This finding concurred with Noor Syahira (2010) who reported that MHC 123 was later in flowering compared with V 36 at every flowering stage with 57.7, 78.8 and 101.5 DAP for MHC 123 at first flowering, 50%

flowering and 100% flowering respectively and 45, 65.2 and 92.7 DAP for V 36 at first flowering, 50% flowering and 100% flowering respectively.

Table 3: Mean number of days to flowering

Treatment	First flowering	50% flowering	100% flowering
Variety (V)			
MHC 123	51.8a	78.8a	103.3a
V 36	41.5b	67.5b	88.8b

Means within each column with different letters were significantly different at $P < 0.01$ using LSD

Forage quality

Crude protein content

Harvest age showed significant effect ($P < 0.01$) on crude protein (CP) content but no significant difference was shown between varieties. There was significant interaction ($P < 0.05$) between harvest age and variety on CP as shown in Table 4. Thus, the result will focus on interaction between harvest age and variety. The crude protein content from 8 WAP (18.05 %) to 12WAP (17.22 %) in MHC 123 decreased slowly but in V 36 (21.72 to 11.31 % of CP) showed a rapid decline from 8 to 12 WAP (Figure 2). The decrease in CP content at 12 WAP was mainly due to increase in plant maturity and rapid accumulation of fibrous component (Anutet *et al.*, 2009). The CP content in the study by Wan Zahari (2004) was similar with the current work which showed that at 8 WAP the CP content was 21% for V 36. Philips *et al.*, (2002) recorded lower CP (15%) as compared to the MHC 123 and V 36. This shows that the CP content varies between varieties. Even with a lower value of CP content at 12 WAP compared to 8 WAP for MHC 123 the value of 17% CP is acceptable even for dairy cattle (Nocek, 1987). The CP content for MHC 123 was similar with the finding of Abdullah *et al.*, (2006) where CP content ranged between 18 to 15%.

Table 4: Mean crude protein content

Treatment	CP (%)
Harvest age (H)	
8 WAP	20.3a
12 WAP	14.3b
Variety (V)	
MHC 123	18.1a
V 36	16.5a
Significance level	
Harvest age	**
Variety	ns
H x V	**
Mean	17.29
CV	9.62

Means with the same letter were not significantly different among harvest age and variety $p > 0.05$ using LSD.

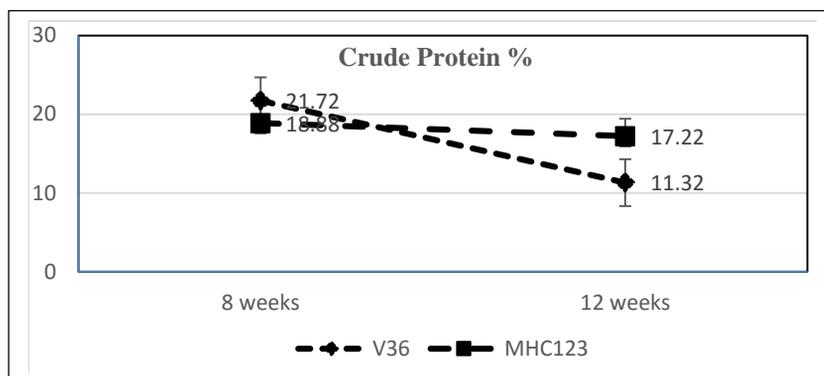


Fig. 2. Crude protein content of two varieties of kenaf harvested at 8 and 12 weeks. (Bars denote standard deviation $n=4$)

Acid detergent fibre content

Acid detergent fibre (ADF) content was significantly affected by harvest age and variety (Table 5). There was also significant interaction between harvest age and variety (Figure 3). Thus the result will focus on interaction between harvest age and variety. At both harvest age, V 36 showed significantly higher ($P<0.05$) ADF content compared with MHC 123. ADF content in MHC 123 increased slowly from 8 to 12 WAP (31.72 to 36.86 %) but in V 36 it increased at a much faster rate (39.57 to 55.59 %). For kenaf, after 50% flowering, the growth rate slowed down and the young stalk became fibrous. MHC 123 was late in flowering compared with V 36. MHC 123 takes about 11 WAP to 50% flowering while V 36 takes about 9 WAP to 50% flowering. ADF content in MHC 123 increased slowly from 8 to 12 WAP because in that period MHC 123 was still actively growing and the fibre in the stalk was still immature. In contrast, the ADF content in V 36 increased rapidly from 8 to 12 WAP because V 36 reached the 50% flowering very early at 9 WAP and the stalk has reached full maturity.

Table 5: Mean acid detergent fibre content

Treatment	ADF (%)
Harvest age (H)	
8 WAP	35.6b
12 WAP	46.2a
Variety (V)	
MHC 123	34.3b
V 36	47.6a
Significance level	
Harvest age	**
Variety	**
H x V	**
Mean	40.93
CV	7.61

Means followed by the different letters were significantly different among harvest age and variety using the LSD ($P<0.05$) ** $P<0.01$.

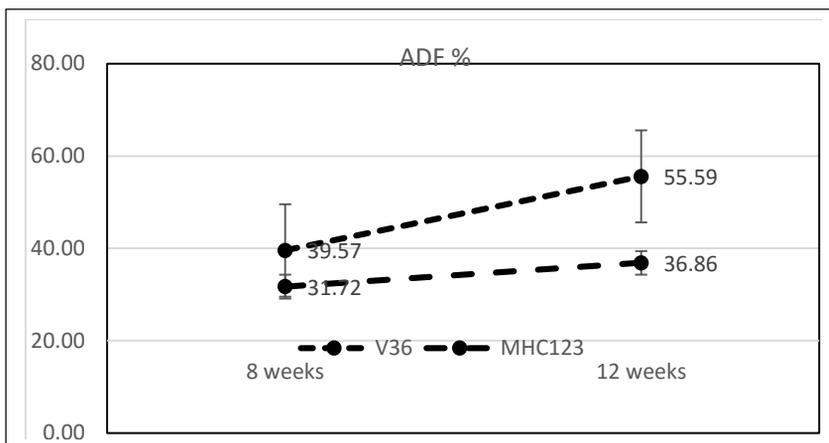


Fig. 3. Interaction between harvest age and variety on acid detergent fiber content. (Bars denote standard deviation $n=4$)

Correlation coefficients between crude protein and acid detergent fiber content

In order to determine the relationship between crude protein and acid detergent fiber content, the correlation between crude protein and acid detergent fiber content were analyzed. There was a significant negative correlation ($r=-0.776$ $P<0.01$) between crude protein and acid detergent fiber content

Leaf yield, stem yield and leaf to stem ratio

Table 6 shows the leaf yield, stem yield and leaf to stem ratio of kenaf. Both leaf and stem yield were significantly higher ($P<0.01$) at 12 weeks harvest compared to 8 weeks. However leaf to stem ratio between the two harvest dates was not significantly different. Generally as maturity advanced, leaves at lower plant part senesced and yield of the stems compared to leaves increased (Anutet *et al.*, 2009). This could be the reason stem yield at 12WAP was higher compared with 8 WAP. Leaf yield of MHC 123 and V 36 ranged from 1.6 to 1.7 $t\ ha^{-1}$ while stem yield of MHC 123 and V 36 ranged from 8.0 to 9.9 $t\ ha^{-1}$. There were no significant difference in leaf and stem yield between the two varieties, however leaf to stem ratio of MHC 123 (0.37) was significantly higher ($P<0.05$) than V 36 (0.16).

Table 6. Mean leaf yield, stem yield and leaf to stem ratio.

Treatment	Leaf ($t\ ha^{-1}$)	Stem ($t\ ha^{-1}$)	Leaf to stem ratio
Harvest age (H)			
8 WAP	1.3b	6.4b	0.20a
12 WAP	2.0a	11.5a	0.17a
Variety (V)			
MHC 123	1.6a	8.0a	0.37a
V 36	1.7a	9.9a	0.16b
Significance level			
Harvest age	**	**	ns
Variety	ns	ns	*
H x V	ns	ns	ns
CV	29.6	18.38	23.23

Leaf yield, stem yield and leaf to stem followed by the same letter are not significantly different among harvest age and variety using LSD ($P>0.05$). ns: not significant * $P<0.05$ ** $P<0.01$

Fiber quality

Bast yield, core yield and bast to core ratio

The components of kenaf as a fibre material include the bast and core fibre. Table 7 shows the yield of bast and core fibre at two harvest ages of the two tested varieties. There were significant differences in bast yield and core yield between the two harvest ages. Harvesting at 20 WAP gave a significantly higher ($P<0.01$) bast yield (3.5 t ha^{-1}) compared with harvesting at 16 WAP (3.2 t ha^{-1}). Similarly, core yield was also significantly higher ($P<0.01$) at harvest age of 20 WAP (5.4 t ha^{-1}) compared with harvest age of 16 WAP (5.0 t ha^{-1}). Bast to core ratios at 16 and 20 WAP (0.65-0.66) were not significantly different between varieties and between harvest age. MHC 123 was significantly ($P<0.05$) higher in bast yield which was 3.4 t ha^{-1} compared to V36 with 3.3 t ha^{-1} . There were no varietal differences in core yield which were 5.1 and 5.2 t ha^{-1} for V36 and MHC 123 respectively. In addition, there was a significant positive correlation ($r=0.767$ $P<0.01$) between bast and core yield, indicating that when bast yield increased, core yield also increased.

Table 7. Mean bast yield, core yield and bast to core ratio of two varieties of kenaf at 16 and 20 weeks.

Treatment	Bast yield (t ha^{-1})	Core (t ha^{-1})	Bast to core ratio
Harvest age (H)			
16 WAP	3.2b	5.0b	0.66a
20 WAP	3.5a	5.4a	0.65a
Variety (V)			
MHC 123	3.4a	5.2a	0.66a
V 36	3.3a	5.1a	0.65a
Significance level			
Harvest age	**	**	ns
Variety	*	ns	ns
H x V	ns	ns	ns
Mean	3.35	5.16	0.65
CV	8.65	11.99	9.37

Bast yield, core yield and bast to core ratio followed by the same letter are not significantly different among harvest age and variety, ** $P<0.01$, * $P<0.05$. ns : not significant.

Tensile test

There was significant interaction between varieties and harvest age on maximum tensile strength ($P<0.01$) (Table 8). Thus, the result will focus on the interaction between harvest age and variety. Figure 4 shows that the tensile strength of MHC 123 was significantly higher compared with V 36 at both harvest age, 16 and 20 WAP. MHC 123 showed a rapid decline in tensile strength from 16 WAP (149.8 MPa) to 20 WAP (53.6 MPa). V 36 also showed a decline in tensile strength but not as drastically as MHC 123 (80 to 39.6 MPa). From the result it could be concluded that maximum tensile strength was at 16 WAP for both varieties but there was a greater decline in tensile strength with later harvest in MHC 123 than V36. Norlin *et al.* (2011) also reported a similar result where maximum tensile strength was found at harvest age of 16 WAP with 118.30 MPa for kenaf fibre. Therefore based on this report the suitable harvesting age for producing fibre for MHC 123 and V36 was at 16 WAP.

Table 8. Maximum tensile strength of two kenaf varieties at 16 and 20 weeks harvest.

Treatment	Maximum tensile strength (MPa)
Harvest age (H)	
16 WAP	114.9a
20 WAP	46.6b
Variety (V)	
MHC 123	101.7a
V 36	59.8b
Significance level	
Harvest age	**
Variety	**
H x V	**
Mean	80.67
CV	16.09

Means with same letter are significant different, ** P<0.001.

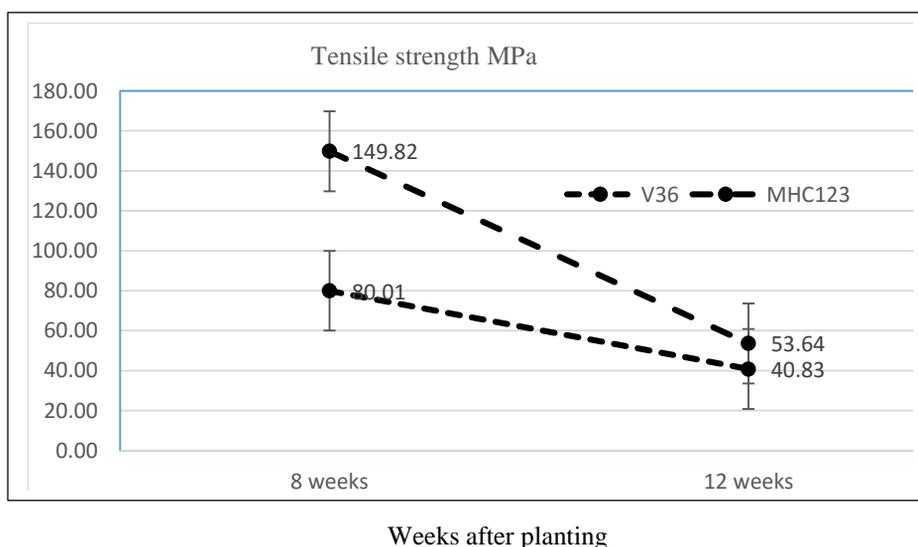


Fig. 4. Interaction between variety and harvest age on tensile strength. (Bars denote standard deviation, n=4)

Water absorption

Significant differences in water absorption was observed among day after immersion (DAI), harvest age (H) and variety (V) as shown in Table 9. However, there was no significant interaction between the factors on water absorption of the fibre. Thus, the result will focus on the main effects on water absorption. From day 1 to day 4 there was slow absorption while from day 4 to day 6 there was a rapid increase in water absorption which became stable from day 6 to day 8. Kenaf fibre has a hydrophilic characteristic, the penetration of water to the fibre makes the fibre swell until a maximum water absorption is achieved. In this study, the maximum water absorption started at 6 DAI.

Fibres from plants harvested at 20 WAP (122.9%) had significantly higher ($P<0.05$) water absorption compared to those harvested at 16 WAP (117.4%). V 36 had a significantly higher water absorption compared to MHC 123 with 124.3 and 116 % respectively. The desirable quality of kenaf fibre is one with lower water absorption. Therefore, the best quality of fibre was obtained from MHC 123 harvested at 16 WAP. For V 36, a better quality fibre was also found at harvest age of 16 WAP. The higher quality of fibre at 16 WAP was possibly due to the higher maximum tensile strength as compared with the fibre harvested at 20 WAP. From the observation, the fibre from harvest at 20 WAP was easy to break by hand and this could be the reason for the higher water penetration, because once the micro-cracking occurs the hydrophilic characteristic of the fibre will contribute to more water penetrating, creating swelling stress and lowering the fibre strength (Norlin *et al.* 2011).

Table 9: Mean water absorption

Treatment	Water Absorption (%)
1	69.5e
2	77.7e
3	87d
4	99.7c
5	133.6b
6	160.7a
7	166.3a
8	166.6a
Harvest age (H)	
16 WAP	117.4b
20 WAP	122.9a
Variety	
MHC 123	116.0b
V 36	124.3a
Significance level	
Harvest age	*
Variety	**
Day after immersion	**
H x V	ns
H x DAI	ns
DAI x V	ns
H x V x DAI	Ns
Mean	120.3
CV	10.80

Means within day after immersion, harvest age and variety followed by the same letter are not significantly different using the LSD, ** $P<0.01$, * $P<0.05$. ns : not significant.

CONCLUSION

The results of this study indicate that varietal differences occur in kenaf and the harvest management should be based on these varietal differences. The new variety MHC 123 showed better yield and forage quality than the current variety V 36. For the purpose of animal feed MHC 123 can be harvested at 12 weeks after planting while V 36 should be harvested earlier. For fibre production both varieties should be harvested at 16 weeks after planting as delayed harvest will cause a reduction in fibre quality in terms of tensile strength and water absorption.

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**IMPACT OF FARM-BASED LEARNING PRACTICES ON YOUNG FARMERS:
CASE FROM AN ORGANIC FARM IN OGAWA TOWN,
SAITAMA PREFECTURE, JAPAN**

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ABSTRACT

The Japanese agricultural sector has been facing issues such as lack of successors and increase in elderly farmers in recent years. Finding ways to attract and offer learning opportunities for the young generation to become new farmers has become a serious concern. This study aims to clarify the role of the farmer as “educator or teacher”, the methods these farmers utilize to foster the motivation of the youth, and the significance of their impact. A series of farm visits at Shimosato Farm was conducted in 2012 and 2013. Moreover, life history interviews of selected learners were conducted in December 2013. This study focuses on Yoshinori Kaneko as a pioneer of organic farming (OF) since 1970 and farm stay since 1979, and his learners who did one-year farm stay in his Shimosato Farm in Ogawa Town, Saitama Prefecture. In response to lack of successors and increase in elderly farmers, he has been playing three (3) important roles. First, differing from institutions and schools, he has been able to spontaneously motivate and attract youths from non-farm households to do farm stay and has been accepting learners since 1979. Second, he has broadened the concept of OF from focusing solely on monetary gain to include its intrinsic value related to environmental conservation and food safety and security. Third, in recent years, he has been actively involved in institutions and colleges establishing or incorporating farm stays, organic agriculture curriculum and other active learning methods in their activities. In fact, he was one of the founders, and currently serves as a board member and lecturer at the Japan Institute of Agriculture Management. This study found that learners have engaged in ‘alternative’ farm stay for different reasons and duration. Among the 90 learners, more than 50% reported that they are currently engaged in farming. This conveys that farm stay seems to play a role in addressing the issue of lack of successors. In addition, the three selected learners showed distinctive and spontaneous action towards farm stay and OF, and in fostering the motivation of the next generation. Text-mining analysis and life history interviews revealed that learners considered the importance of connectedness (*tsunagari*), and mentor-companion relationship (*shitei dogyo*) remained strong, even after the farm stay since their ages happened to be near their mentor’s.

Key words: ‘alternative’ farm stay, learners, connectedness, farm-based learning practice, organic farm, Yoshinori Kaneko, young farmers

INTRODUCTION

The Japanese agricultural sector has been facing issues such as lack of successors and increase in elderly farmers in recent years. Finding ways to attract and train the young generation to become new farmers has become a serious concern. If not properly and immediately addressed, this issue may cause further absence of farm successors that will lead to more abandoned farmlands, excessive depopulation of rural areas and the worst scenario, loss of the country’s distinctive culture and agriculture. Although the Japanese government has been addressing these issues, new farmers have decreased by about 30% from 81,000 in 2006 to 58,100 in 2011. Moreover, the number of new farmers aged 39 years old or less has not made an annual increase of more than 15,000 persons in recent years (MAFF, 2012).

With regards to learning methods for young farmers, past research shows that courses of study are offered by academic institutions under the Ministry of Education, Culture, Sports, Science and Technology (MEXT) and varying institutions under the Ministry of Agriculture, Forestry and Fisheries (MAFF). The former includes agricultural high schools (Sasaki, 2014) and Colleges of Agriculture in universities (Inaizumi *et. al.*, 2014a), while the latter refers to agricultural extension (Fujita 1997 and 2011, Shiomi 2000), experimental stations and agricultural colleges (Fujita, 1997 and 2011; Inaizumi, 2006; Shiomi, 2000). Agricultural colleges are farmers’ training centers (originally non-formal school type), which are under the supervision of the MAFF instead of MEXT. The implementation level and content varies as well. However, in order to achieve steady progress of learning, these institutions commonly combine lecture and learning methods in accordance with a pre-set curriculum. Nowadays other places of learning, such as school education (*gakko kyoiku*), non-formal school (*juku* style) and farm practice, have become available. Although these institutions consider themselves as “place for learning”, most of them are more and more transforming into formal schools. Of all the learning methods available, such as lectures, on-farm trials and seminars, many institutions give special focus to practicum within and/or outside their vicinity (Chiba Prefecture Agricultural College, 2015; Tokyo Metropolitan Agricultural High School, 2015). According to Inaizumi (2003), practicum is the most effective educational method. The Japan Institute of Agricultural Management (2014) and Nagano Prefecture Agricultural College (2015) have used long-term practicum as their pillar of education. In order words, learning methods offered by most academic and instructional institutions seem to significantly involve farmers. However, farmers as “educators or teachers” in the context of fostering motivation (or lighting the fire) of young farmers have not been fully examined.

This study sought to determine the roles of farmers as “educator or teacher”, the methods utilized to foster the youth and the significance of their impact. Differing from the “educators or teachers” in high schools, agricultural colleges and universities, and extension workers, this study hypothesizes that farmers themselves may provide essential and valuable knowledge or insights while fostering the motivation of young farmers.

As a pioneer of organic farming (OF) and farm stay, this study focuses on Yoshinori Kaneko (Farmer YK) and his learners who did one-year farm stay in his Shimosato Farm in Ogawa Town, Saitama Prefecture. In response to issues related to lack of successors and increase in elderly farmers, Farmer YK has been playing three important roles. First, differing from institutions and schools, he has been able to spontaneously motivate and attract youths from non-farm households to do farm stay and has been accepting learners since 1979. Second, he has broadened the concept of OF from focusing solely on monetary gain to include its intrinsic values related to environmental conservation

and food safety/security. Third, in recent years, he has been actively involved in institutions and colleges have establishing or incorporating farm stays, organic agriculture curriculum and other learning methods in their activities. In fact, he was one of the founders and currently serves as a board members and lecturer at the Japan Institute of Agriculture Management.

As shown in Table 1, various aspects of OF development and initiatives by Farmer YK, his learners and Ogawa town have been heavily researched. These concrete cases with public and social significance seem to have spontaneously attracted or motivated youth, even those from non-farm households through the years.

METHOD OF THE STUDY

Selection of the Study Area and Case Respondents

Ogawa Town. Ogawa town is located in the middle of Saitama, approximately 60 km from Tokyo (Fig. 1). The town can be characterized as having limited flat areas conducive for agricultural production, a limited number of farmers mostly operating on a small-scale, and a lack of farm successors (MAFF, 2010). This study area was selected as the hometown of Farmer YK, who addressed the above-mentioned issues by promoting OF through various public-private partnerships focusing on the development of the whole town, which was duly recognized as one of the country's

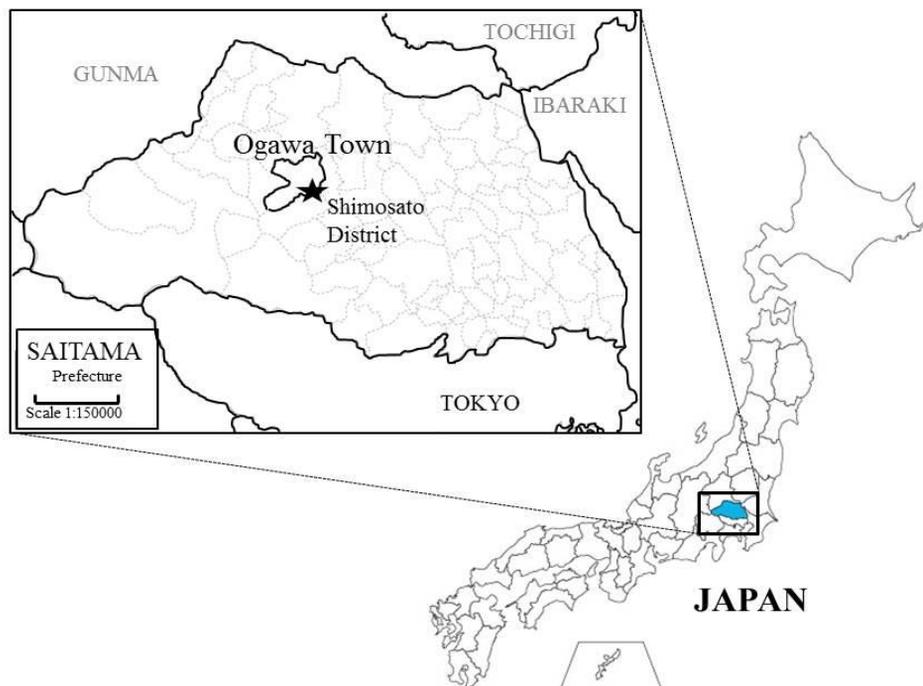


Fig. 1. Location of the Study Area

Note: Prepared by authors.

Organic Model Town in 2008. It is noteworthy that among the town's 41 new farmers from 1983 to 2011, as many as 29 farmers have engaged in OF, conveying its significant contribution in increasing the number of farmers (Promotion Council of Organic Agriculture Ogawa Town, 2013 as cited by Shimoguchi *et. al.*, 2015 in print).

Farmer YK. One of the first graduates of the National Farmer Academy, he pioneered the practice of OF in Ogawa town, throughout four decades of various OF initiatives and partnerships, not limited to his town, which eventually made him well-known not just in Japan, but also around the world. Farmer YK was selected based on his pioneering work on OF and farm stay, fostering motivation of youth including non-farm household individuals through a one-year farm stay, fostering good connections with others, and establishing key organizations. These spontaneously transformed his district to an organic community, an initiative duly recognized by MAFF through the awarding of the Emperor's Cup in 2010. It should be noted that farm stay, a kind of apprenticeship, is the most common and traditional farmer learning practice in Japan.

Learners. Individuals of all ages and from various walks of life have been accepted in Shimosato Farm since 1979 for different reasons and duration of farm stay. For this study, learners who did one-year farm stay are selected, since the longer exposure to farm life, and closer interaction with Farmer YK and other people involved may reflect significant impact. Since these learners learned directly from Farmer YK (refers to as the first generation), this study refers to these learners as the second generation. In addition, three learners were selected for further analysis based on the results of text-mining analysis and the following five reasons: no farming background, one of the first Shimosato Farm learners (1980s), decision to engage in OF after farm stay, change of residency to Ogawa town, and currently own/manage an organic farm within the town. It is noteworthy that the selected learners for the study matched with Farmer YK's advice and choice.

Data Collection

Both primary and secondary data were utilized in this study. Primary data were collected in two ways. First, a series of farm visits were made to Shimosato Farm in September 2012, and July, September and November 2013. Actual interactions between Farmer YK and his learners were also observed. Second, life history interviews of three selected learners were conducted in December 2013, to further determine and capture the details of the learners' relationship with Farmer YK, their farm stay experience and its impact. Questions included technical knowledge on OF from Farmer YK and views on agriculture and farm village. All interviews were recorded using an IC recorder and transcribed immediately thereafter.

Secondary data were mainly taken from a book entitled *Ichi Ryu Man Bai* (Orito and Morimoto, 2009), which means "A single seed can eventually produce a great harvest" in English. Published for the 30th Wedding Anniversary of Farmer YK and his wife, this document contains: (1) personal profiles (e.g. age, educational attainment, prior occupation, year of farm stay); (2) reason(s) for doing farm stay; (3) if farmer, current farm profile (e.g. farm size, produce) and difficulties as a new farmer; (4) farm stay experience (e.g. most memorable experience, struggles during farm stay, lessons after farm stay); and (5) congratulatory messages, submitted by a total of 90 learners. Since all farm records were destroyed by fire at Farmer YK's ancestral home, this document is considered the only comprehensive document available regarding his learners.

In this study, the learners have been categorized into two types, namely Type A learner (identified oneself as a farmer as of 2009) and Type B learner (identified oneself as a non-farmer as of 2009).

Data Analysis

This study utilized descriptive analysis, text-mining analysis and life history approach. Text-mining enables the extraction of interesting or valuable information from unstructured text (Ishida and Mingzhe, 2012). In this study, the original texts from *Ichi Ryu Man Bai* (Orito and Morimoto, 2009) were used in morphological analysis, wherein morphemes were identified and parsed. Morpheme refers to the smallest unit with a meaning in a language. The texts were listed from the highest number of appearances for both Type A learner and Type B learner by each question.

Table 1. List of Some Publications on Farmer YK and Ogawa Town's OF Initiatives and Impacts¹

Author(s) and Year Published	Contents
Iizuka, 2009	Motivation and views on life of organic farming women compared to city women
Inaizumi <i>et. al.</i> , 2014b	Learning practices in Shimosato Farm and its impact to young farmers
Kaneko, 1992, 1998	Farmer YK's philosophies and motivation on OF
Kaneko, 2003	Farmer YK's organic vegetable farming
Kaneko, 2008	Initiatives of farmer YK through the years; OF development in Ogawa town; Establishment of the Organic Model Town
Kaneko, 2012	Sustainable recycling model of Shimosato Farm
Kaneko, 2015	Partnerships with local industries towards community development
Kaneko and NPO Tsubasa Yu, 2015, and Takahashi, 2013	TEIKEI system (voluntary co-partnership between consumers and farmer; Partnerships and projects with local industries such as <i>tofu</i> company, sake company and reform company); <i>KOMEMAME</i> Project since 2009 (With the assistance of NPO Tsubasa Yu, the reform company will purchase all the harvested rice from the farmer group, and pay outright at the set price, which can sustain the next harvest)

Author(s) and Year Published	Contents
Oguchi, 2012	Three (3) stages of organic agriculture development process in Ogawa town: Foundation stage (1972-early 1980s), Enrichment stage (mid-1980s-2000) and Expansion stage (2000 onwards)
Oguchi, 2013	Important role of new young farmers involved in the development of OF in Ogawa town
Orito, 2014	Concept of Teikei system; Farmer YK's shift to Orei-sei system (or Gratitude system); Loyal partner-consumers for more than 20 to 38 years
Shimoguchi <i>et. al.</i> , 2015 in print ²	Institutional support of Ogawa town office and an active NPO
Tachikawa, 2011	Background and initiatives of Improvement Measures Council of Land, Water and Environment Conservation in Shimosato District established in 2008
Takahashi, 2007	Initiatives of Ogawa Town Organic Producers Group (OTOGP) established by Farmer YK and his trainees in 1995; Importance of collaboration with environment conservation initiatives of the community, partnerships with local industries, and building relationships with people in the community other than farmers
Takahashi, 2013	Characteristics of trainees who became members of OTOGP
Watanabe, 2013	Partnerships with <i>tofu</i> company

Notes: 1) All publications are in Japanese.

2) Although Nakajima (2010a, 2010b, 2011) made comprehensive reviews of the policies related to organic agriculture in Japan, some studies focused on the role of public and private institutions in the development of organic agriculture.

Life history is a popular qualitative approach used by researchers from various fields of science to reveal further details or realities of a given study or research topic (Ojermark, 2007). In this study, the transcribed information from the interviews was arranged into categories (*e.g.* farm stay route, farm stay course and details, and future vision) for easier comparison and discussion. Each category was further narrowed down into perception, judgment and action.

RESULTS AND DISCUSSION

Organic-Related Learning Practices in Japan

In Japan, there are various organic-related learning practices available such as books, internet sites, farmers’ fairs, seminars and lectures, and farm experience through farm visit and farm stay. Table 2 conveys that since all practices have merits and demerits, it is possible to choose one practice or combine various practices that will answer individual needs and contribute towards a particular goal.

Table 2. Merits and Demerits of Various Organic-Related Learning Practices in Japan

Learning Practices	Advantages	Disadvantages
Books	One can engage in systematic learning by himself/herself at any given time and place.	Once the book is published, it is quite difficult and costly to revise and republish. Learning by experience using own senses such as touch and sight is quite difficult.
Internet Sites	One may easily obtain the latest information (including videos, pictures and audios), which is commonly protected by internet environment restrictions.	One may find it difficult to engage in systematic learning due to various distractions such as numerous internet games, bad internet connection. Obtained data may not also be commonly acceptable.
Farmers' Fair	One can have the chance to learn by actual observation, contact and interaction.	Since this is supposed to be a festive event, one may find it difficult to engage in systematic and advance learning.
Seminars and/or Lectures	One can achieve systematic and highly specialized learning, and have the opportunity to increase knowledge and deepen understanding by raising questions and having discussions on the spot	There are limitations in space and time. Moreover, specialization or focus on a given topic is unavoidable. Thus, one may not achieve systematic learning.
Farm Experience (Farm Visit & Farm Stay)	One can learn through hands-on farm activities.	It is impossible to cover all the activities in one program. Thus, systematic learning is difficult to achieve.

Note: Prepared by authors.

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Table 3 shows that there are a total of 145 enterprises, mostly family farms, and 47 educational institutions offering organic-related farm stay in Japan. Depending on the enterprise or institution, the content, style and length of learning method vary. It should be noted that Shimane Prefectural College for Agriculture and Forestry (Shimane Prefectural College for Agriculture and Forestry, 2015; categorized under Farmers Training Center) is one of the few educational institutions offering an official organic agriculture curriculum to-date.

Table 3. Types of Places that Offer Organic Related-Farm Stay

Type	Number
Enterprise	
Family Farm	118
Agricultural Company	27
Total	145
Educational Institution	
Public Institution	42
Private Institution	5
Total	47

Sources: Organic Farming Promotion Council (yuki-hajimaru.net); MAFF, 2013; New Farmer Consultation Center (<http://www.nca.or.jp/Be-farmer/>)

Note: Shimane Prefectural College for Agriculture and Forestry (categorized under Farmers Training Center) is one of the education institutions, which offer an official organic agriculture curriculum.

So, what can one expect from the farm stay experience? Farm stay at a family farm is very appealing for aspiring farmers since one can fully experience a farmer's life while enjoying nature and rural life, especially for those who have the opportunity to stay with the host family. Moreover, there is also step-by-step exposure to farm activities from land preparation to harvesting, then marketing and delivery. Through this experience, one can assess whether he/she is inclined or not to engage in farming or related undertaking for small-scale business or subsistence.

On the other hand, farm stay at an agricultural company is commonly recommended to aspiring farmer entrepreneurs or farm company employees. One can experience working with different people and learn how an agricultural company operates. Since most companies have various divisions such as production, marketing and manufacturing, detailed experience and acquired knowledge may be limited to the particular division where he/she had the opportunity to work at.

Based on the above-mentioned, interest, purpose and available time seemed to be key factors in choosing the type of farm stay.

Learning Practices in Shimosato Farm

Shimosato Farm offers both farm visit and farm stay. Held once every two months, the farm visit is a half-day activity offered at 2,500 yen per person, which includes two lectures, followed by a farm tour. Figure 2 shows the content of farm visit as of May 2015. The lecture of Farmer YK includes his motivation and philosophy of organic agriculture, the development process and practices of Shimosato Farm, and how organic agriculture has transformed the community through the years

with its partners and promotion initiatives. Varying every session, the guest speaker comes from the group of community partners. For example, the *Tofu* company president was at the farm in March 2015 to share how the partnership between the farm and his company was established and evolved into having a significant impact on the community.

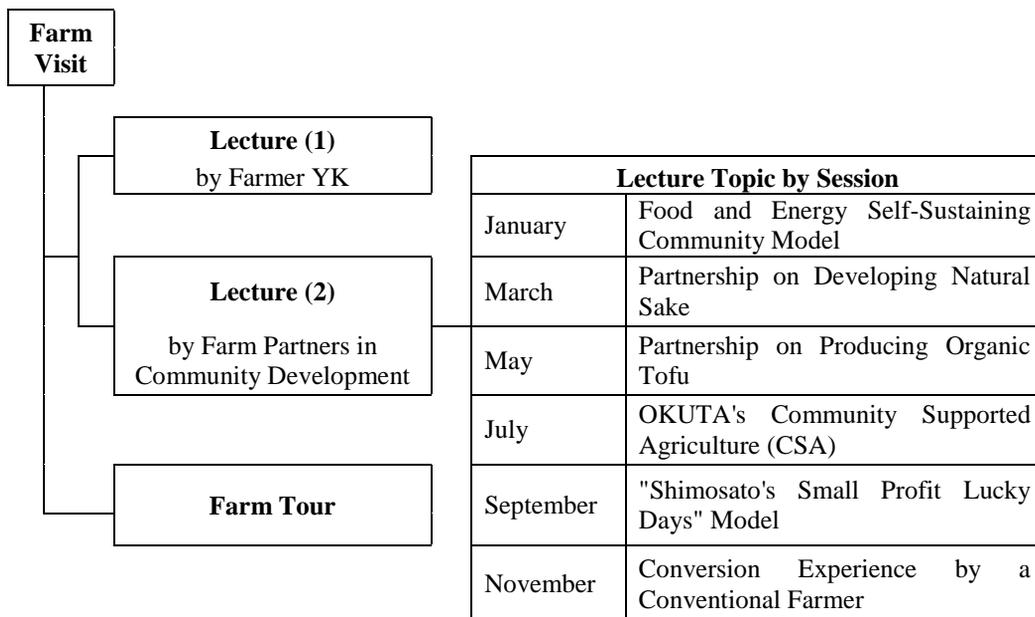


Fig. 2. Contents of the Farm Visit in Shimosato Farm as of May 2015

Source: Shimosato Farm Webpage (<http://www.shimosato-farm.com/watching2.html>)

Note: Prepared by author.

After two lectures in every session, Farmer YK personally leads the farm tour by showing his farm to the participants. If his schedule does not permit, one of his learners usually conducts the tour on his behalf. During the tour, participants will see first hand and learn about his farm including vegetable and fruit production in plots and greenhouses, various animals, fertilizer production, energy recycling initiatives (*e.g.* agricultural machines and car running on straight vegetable oil (SVO), glass green house, bio-gas, wood boiler) and learners at work. Although the contents of Farmer YK's lecture and farm tour seem to be the same every session, additional or changed information and farm landscape (*e.g.* use of new farming method or technology, new learners, updates on various initiatives, seasonal vegetables and fruits) can also be expected as the season changes. Thus, it is very common to find people who have participated more than once. On the average, there are about 70 participants per session.

The farm stay has been offered since 1979 when Farmer YK and his wife decided to accept learners interested in OF and sustainable living, and environmental conservation enthusiasts. The idea came from his wife, who first learned about organic farm school when she visited several European organic farms in 1978 (Kaneko, 2008). Interview with Farmer YK revealed that the content, length and schedule of the farm stay are flexible. Length of stay may be a day, a few days, a year or more. Working individuals may come once or twice a week, during weekends or even on an irregular basis. Considering only his long-term learners, he estimated that he already had more than 100 learners who engaged in one-year farm stay. Fukuda (2013) reported that Shimosato Farm had hosted more than 300 learners in the last 30 years. This figure definitely includes short-term learners.

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This study has also clarified that the farm stay does not follow a special training program with specific schedule, handouts, topics and activities. Learners are exposed to various activities or farm work at a given time, day or season. Since the learners live with Farmer YK and the rest of his family, household chores are also part of their responsibility.

It should be noted that the farm stay being offered by Shimosato Farm does not refer to a commercial farm stay or farm tourism. According to Kidd *et al.*, (2004), one has to pay for farm accommodation and various farm activities in the case of commercial farm stay or farm tourism. In this study, farm stay here refers to 'alternative' farm stay, a little similar to Willing Workers on Organic Farms (WWOOF). McIntosh and Bonnemann (2006) explained that in WWOOF, 'alternative' farm stay experience involves interested individuals who work voluntarily, while enjoying farm life offered by the host organic farm in lieu of payment. Moreover, the experience at a WWOOF farm is significantly different from commercial farm stay, "in terms of the rurality of the experience, the opportunity to learn organics, the personal meaning of the experience, and the element of sincerity in the experience". However, unlike in WWOOF, there is no prior requirement of membership or standard procedure to follow to avail the benefits of WWOOF members (commonly known as WWOOFers). In addition, learning experience from Farmer YK seemed to be the priority of the learners rather than just offering voluntary work.

Those who wish to engage in farm stay in Shimosato Farm are recommended to attend the farm visit or try the short-term farm practice to get acquainted with the farm, its initiatives and staffs. After which, he/she will undergo a personal interview initiated by Farmer YK and his wife. Initially, the couple preferred young individuals aged around 30 years old since their adaptability and learning ability are more flexible. However, this changed from the late 90's, when interests in OF were expressed by people in their 40s and above. Among them were farmers who wished to convert to OF.

The day of the learner(s) usually begins with feeding the animals and preparing or helping prepare breakfast. During breakfast, informal conversation and interactions focus on the activities for the day. The farm produces various commodities for its own consumption and its partner households. Some of the commodities are rice, wheat and 10 to 15 kinds of vegetables. Farm activities include land preparation, sowing, transplanting, maintaining (e.g. watering, weeding) and harvesting of various produce. The farm does not have manuals or formal lectures for the learners, so the learning process is mainly based on observation and imitation (*miyo mimane*) and learning by doing (*nasu koto ni yotte manabu*). In other words, he/she learns by observing, imitating and/or doing how Farmer YK does various farm activities. With a mixture of household chores, farm work and community interaction, the learner also has the opportunity to experience a farmer's life and assess if he/she is inclined to independently engage in such an undertaking in the (near) future.

It should be noted that learner(s) usually stayed at Farmer YK's house and experienced various farm activities with close interaction with Farmer YK during the early years of farm stay. However, with the increasing number of learners in recent years, some learners need to stay at a nearby apartment because the ancestral home could not accommodate everyone. Moreover, sharing of learning experience and knowledge among learners is common since the busy schedule of Farmer YK does not permit him to be with them all the time.

Recent report (MAFF, 2015) conveys that there is an apparent decreasing trend in enrollees from farm households in the country's agriculture-related schools. In 2011, the share of these enrollees became less than 50%. In addition, the number of enrolled sons/daughters from farm households and who are willing to do learn how to farm appeared to be declining. The same report noted that only 63% and 33% of the graduates hailing from farm households and non-farm households, respectively, were engaged in farming after graduation. Although MAFF (2009) recognizes the need to increase new farmers from non-farm households, above-mentioned figures show that individuals from non-

farm households seemed to have difficulty in becoming new farmers. Factors in consideration are limitations under the farm successor system (Uchiyama, 2014), no access to rights to farmland ownership, high capital requirement for purchasing machineries and facilities (MAFF, 2009).

Looking at the learners in Shimosato Farm, Table 4 shows that among the 90 learners, 46 learners were currently engaged in farming (referred to as Type A learner), of whom 37 (80%) were males. Although significantly dominated by males, women’s role and active engagement in farming cannot be disregarded (Iizuka, 2009). The average age of learners during farm stay was 30 years. Comparing age by current occupation, Type A learners were older compared to Type B learners for both males and females.

In general, this study found that Shimosato Farm has been steadily contributing to the increase in new farmers. It is important to stress that the "impact" is not always in terms of figures, since there are also other factors involved especially to new farmers with non-farming family background.

Table 4. Profile of Learners in Shimosato Farm, 2009

Current Occupation	n	Age			
		2009		During Farm Stay	
		Average	S.D.	Average	S.D.
Type A Learner					
Male	37	45.2	9.7	33.0	11.6
Female	9	37.9	13.1	29.8	7.2
Total	46	43.8	10.8	32.3	10.9
Type B Learner					
Male	25	39.0	11.7	28.3	10.3
Female	19	34.5	6.3	26.0	4.9
Total	44	37.4	10.3	27.4	8.7
All Learners					
Male	62	42.9	10.9	31.3	11.3
Female	28	36.1	9.8	27.8	6.2
Total	90	41.1	11.0	30.5	10.3

Source: Orito and Morimoto, 2009

Note: 2009 refers to publication year.

Reasons for Doing Farm Stay and Lessons during Farm Stay

Using text-mining analysis, text data on reasons for doing farm stay and lessons during farm stay for both Type A learners and Type B learners were analyzed as shown in Tables 5 and 6, respectively. It was found that there were many keywords related to reasons of engagement.

However, referral of a friend and book or magazine articles seemed to be more important as shown in the following statements:

I read a book entitled “Warera Hyakusho no Sekai” (The World of Farmers in Japanese published by the Japanese Society of Organic Agriculture Science in 1983) and came across Orei-sei, which I became very much interested in. With the strong will to experience genuine organic agriculture, I went to Shimosato Farm. (Type B learner, #10)

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Since I believed that organic agriculture is the only way not to further pollute the earth, I started studying and researching about organic agriculture. One day, a friend of mine told me about Mr. Kaneko. Soon after, I participated in the farm visit, wherein I came across a document stating that the farm practices advocate recycling. This immediately made me ask to be a learner at the farm. (Type B learner, #12)

Table 5. Text-Mining Analysis of Reasons for Doing Farm Stay

Keywords	No. of Answers	Choice Probability (%)	Type A Learner	Type B Learner	
Farm stay	38	42.2	22	16	
Shimosato farm	33	36.7	19	14	
Organic agriculture	22	24.4	6	16	**
Agriculture	20	22.2	13	7	
Kaneko	17	18.9	9	8	
Referral	17	18.9	9	8	
Reason	14	15.6	5	9	
Work	12	13.3	6	6	
Oneself	12	13.3	6	6	
Indebtedness	11	12.2	7	4	
Time	11	12.2	3	8	*
None	11	12.2	5	6	
Trainee	9	10.0	4	5	
Moment	9	10.0	4	5	
Way	8	8.9	5	3	
Farm	8	8.9	4	4	
Ancestral home	7	7.8	3	4	
I	7	7.8	1	6	**
Doubt	7	7.8	3	4	
Tomoko	6	6.7	1	5	*
Yoshinori	6	6.7	3	3	
Farm household	6	6.7	2	4	
University	6	6.7	4	2	
Book	6	6.7	3	3	

Source: Orito and Morimoto, 2009

Notes: 1) Only keywords with a frequency of 6 and more are shown.

2) ** Denotes significant at 5%; * denotes significant at 10% using Chi-Squared Test

3) Original text in Japanese was carefully translated into English by the authors.

Statistically, high interest for “organic agriculture” among Type B learners showed significance at 5%. Moreover, Type B learners showed the use of “I” as subject, conveying that their decision to do farm stay at Shimosato Farm seemed to be a voluntary self-action. Some even went directly to Shimosato Farm or phoned Farmer YK without any referrals. Here are some comments from the learners:

Since I had an interest in organic agriculture, I decided to engage in farm stay soon after graduation. (Type B learner, #22)

Before this farm stay, I was interested in permaculture, which brought me to New Zealand, where I did farm stay for 1 year. I first heard about Mr. Kaneko from my host farmer who had been to Shimosato Farm. My host farmer even recommended some books. Since I am also from Saitama and did not know about organic agriculture in Japan, I did farm stay at Shimosato Farm immediately after I came back. (Type B learner, #61)

I wanted to be a farmer so I participated in the farm visit. I was able to capture the whole concept of the farm, which involves various aspects including organic agriculture, bio-gas and solar energy generation. Soon after, I asked Mr. Kaneko to accept me as one of his learners. (Type B learner, #85)

Table 6. Morphological analysis of lessons during farm stay

Keywords	No. of Answers	Choice Probability (%)	Type A Learner	Type B Learner	
Learn	24	26.7	14	10	
Self	16	17.8	11	5	
Matter	16	17.8	12	4	**
Person	15	16.7	8	7	
Important	15	16.7	7	8	
Agriculture	14	15.6	7	7	
Live	13	14.4	8	5	
Yoshinori	12	13.3	7	5	
Shimosato Farm	11	12.2	8	3	
Connectedness	10	11.1	6	4	
Organic agriculture	10	11.1	8	2	*
Lifestyle	10	11.1	5	5	

Source: Orito and Morimoto, 2009

Notes: 1) Only keywords with a frequency of 10 and more are shown.

2) ** denotes significant at 5%; * denotes significant at 10% using Chi-Squared Test

3) Original text in Japanese was carefully translated into English by the authors

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With regard to lessons during farm stay, connectedness seemed to be a relevant key word although it is not statistically significant (Table 6). There are several past studies on connectedness. For example, Gosling and Williams (2010) conducted a study on the connectedness to nature, place attachment and conversion behavior among farmers. The essence of connectedness, and the profile and original text of the learners who mentioned connectedness (and similar terms such as connect) are shown in Table 7.

The responses of some learners are as follows:

Importance of vegetables. Connectedness to people. Importance of soil. Willing to do. Heart of compassion. Joy of farming. I thought that one has to use all his five senses in farming. Perception of agriculture is molded by meeting people.
(Type A learner, #2)

Table 7. Profile and Full Text of Trainees who Mentioned "Connectedness" in Lessons During Farm Stay

No.	Learner Type	Sex	Age in 2009 ¹	Year of Farm Stay	Age when Farm Stay was Done	Original Text*
1	A	Male	49	1993	33	I was able to <u>connect</u> with people and meet people from various specializations, and learn the importance of food.
2	A	Female	23	2009	23	Importance of vegetables. Connectedness to people. Importance of soil. Willing to do. Heart of compassion. Joy of farming. I thought that one has to use all his five senses in farming. Perception of agriculture is molded by meeting people.
3	A	Female	25	2005	21	I was able to meet people who could <u>connect</u> through each other's heart and mind. I enjoyed most the daily meeting and talking with co-learners and people of Ogawa Town. Although I seldom meet everyone after the farm stay, just the thought that I have friends/colleagues who are working hard and share the same philosophy on organic farming, child-rearing and food, gives me reassurance and power to move forward.
4	A	Female	31	2004	26	Live (stay alive) through <u>connectedness</u> . During my one-year farm stay, I was able to seriously think what learners should do to live the life that they desire.

No.	Learner Type	Sex	Age in 2009 ¹	Year of Farm Stay	Age when Farm Stay was Done	Original Text*
5		Female	42	2006	39	I still think that one should value " <u>connectedness</u> ". Whatever one does and wherever one lives, connectedness with people is the most important.
6	B	Male	44	1994	29	Importance of <u>connectedness</u> (person to person)
7		Female	29	2007	27	The most important basics for a person to become a farmer are having the great ability to <u>connect</u> with people and knowing that agriculture is an interaction of life.
8		Female	28	2007	26	Importance of <u>connecting</u> with people. Mr. Yoshinori has always mentioned this. I also learned that one could live a prosperous and happy life by just having an "organic network" and food to eat.
9		Female	36	2008	35	Importance of communicating with people. More than learning about farming technologies, <u>connectedness</u> to people is important. In order to move on, I think that it is also <u>connected</u> with the future.
10		Male	34	2008	33	Communication with living things. Personal relationship. Organic-related appreciation and steady accumulation of good things. Environment conservation. Way of life according to nature. I was able to experience living a farmer's life. I was in-charge of cooking and handling the wood boiler during farm stay. Although I thought that Shimosato Farm and Ogawa Town are not places of comfort, I very much value being able to do organic farming, live, experience, learn, and <u>connect</u> with people, while being surrounded by people and animals with unique characteristics.

Source: Orito and Morimoto, 2009

Notes: 1) Publication year

2) Original text in Japanese was carefully translated into English by the authors.

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Communication with living things. Personal relationship. Organic-related appreciation and steady accumulation of good things. Environment conservation. Way of life according to nature. I was able to experience living a farmer's life. I was in-charge of cooking and handling the wood boiler during farm stay. Although I thought that Shimosato Farm and Ogawa Town are not places of comfort, I very much value being able to do organic farming, live, experience, learn, and connect with people, while being surrounded by people and animals with unique characteristics. (Type B learner, #10)

These contexts clarified that being able to feel the connectedness to a different environment and lifestyle seemed to be important. Moreover, some farmer learners seemed to have been strongly influenced by Farmer YK's philosophies and way of life. One example can be drawn from the following statement:

Importance of connecting with people. Mr. Yoshinori has always mentioned this. I also learned that one could live a prosperous and happy life by just having an "organic network" and food to eat. (Type B learner, #8)

It became clear that Farmer YK and his learners valued connectedness (*i.e.* person to person, person to environment, person to plants and animals). Moreover, there is a sense of mutual trust among the people involved. In other words, Farmer YK has clearly conveyed to them that OF itself is connectedness.

Life History of Selected Learners

In order to further clarify the impact of Farmer YK's learning method, life history interviews were conducted with three selected learners, named Farmers A, B and C. Interview results were categorized into farm stay route, course and details of farm stay and future vision, as shown in Table 8. Each category was further narrowed down into perception, judgment and action. Perception refers to what made him/her aware and how. Judgment refers to what the learner selected among the choices based on his/her perception. Action refers to actual execution based on a particular judgment.

All the respondents engaged in farm stay during the 1980s. With regard to farm stay route, the respondents seemed to strongly prefer rural life (*e.g.* living in Hokkaido) and to have great awareness and concern about pollution and other environmental issues, which were common concerns at that time as shown in the following statements:

I wanted to live in a place surrounded by nature, unlike the cities, like Tokyo where I come from. I wish to enjoy producing vegetables on my own. (Farmer A)

Since I was in elementary school, I wished to stay in the countryside because I have perceived agriculture as an escape from the city. I got interested in organic farming after being dismayed with combined pollution.

Written by a famous Japanese writer named Ms. Ariyoshi, the book, "Fukugo Osen" (combined pollution in English) discusses the agricultural chemical contamination, environmental damage and other issues related to combined pollution in Japan. (Farmer B)

I have been interested in pollution issues and conservation of nature. Working at the sewage treatment plant made me question the treatment itself, since it requires another kind of energy to process. (Farmer C)

Table 8. Interview Summary of Selected Learners

		Farmer A	Farmer B	Farmer C
Age During Farm Stay		24	24	31
Sex		Male	Male	Female
Year Started Farm Stay		1983	1985	1987
Farm Type		Farm Corporation	Commercial Farm	Subsistence Farm
Farm Stay Route	Perception	I wanted to live in a place surrounded by nature, unlike in the cities, like Tokyo where I come from. I wish to enjoy producing vegetables on my own.	Since I was in elementary school, I wished to stay in the countryside because I have perceived agriculture as an escape from the city. I got interested in organic farming after being dismayed with combined pollution. Written by a Japanese famous writer named Ms. Ariyoshi, the book <i>Fukugo Osen</i> (means combined pollution in English) discussed the apparent agricultural chemical contamination, environmental damages and other issues related to combined pollution in Japan.	I have been interested in pollution issues and conservation of nature. Working at the sewage treatment plant made me question the treatment itself, since it requires another kind of energy to process.
	Judgment	I realized the difficulty of becoming a new dairy farming. I was interested in circulation agriculture, which produces many commodities in small amounts.	I realized the difficulty of venturing into dairy farming because it requires huge initial investment. I felt better when Farmer YK told me that I can be a farmer after one year.	I discovered that Shimosato Farm has a recycle simple toilet, which is similar to Nepal's fallen leaves toilet. It was love at first sight!

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	Farmer A	Farmer B	Farmer C
Action	After the training at a dairy farm, I became a salaryman. But not for long, I quitted my job and joined Shimosato Farm's farm stay. Right after I decided to become independent and establish my own organic farm in Ogawa Town.	After the training at a dairy farm, I went directly to Shimosato Farm. Right after the farm stay, I became an independent farmer in Ogawa Town.	After resigning from my work at the sewage treatment plant, I joined Shimosato Farm's farm stay. After the farm stay, I became an independent farmer in Ogawa Town.
Course and Details of Farm Stay	Perception	I personally felt and experienced the connectedness to the general farm activities, life at Shimosato Farm, moment with Ms. Sawako Ariyoshi, and circle of friends in organic farming. Ms. Ariyoshi is one of the partner consumers of Farmer YK when he started TEIKEI.	I learned the plan and action of each farm activity by observing and doing. Since I did not have any knowledge about farming and farming village. I did not understand the implication of each activity.
	Judgment	I realized my lack of background on farming and farm life. However, there was a need for new farmers in the village.	At any rate, I always just followed and accompanied Farmer YK.
	Action	I practiced based on <i>miyo mimane</i> (by observation and imitation) and/or <i>nasu koto ni yotte manabu</i> (learning by doing). I always followed him. Through connectedness,	I made sure that I worked with Farmer YK at all times. Mentor-companion relationship. In general, I did everything.
			My case was very different from the current situation. I was the only one who stayed full time in the house of Farmer YK. Number of learners living-outside or sometimes staying in the house had no limitations. I had freedom to do as I wish. So, I was able to learn at my own pace.
			Since I am not a person who usually asks question, I learned by observing and imitating while thinking, and doing by trial-and-error.

		Farmer A	Farmer B	Farmer C
		I was able to take the opportunity to become independent.		
Future Vision	Perception	I perceive that the change of generation in the village and Ogawa town will be at a fast pace. Lack of farm successor and difficulty for new farmers to acquire farmlands are prevalent issues.	Enjoy. I enjoy doing farming in these climate and landscapes. I feel reassurance with the food and energy I have, most especially after the Great East Japan Earthquake, Tsunami and Nuclear Meltdown in March 2011.	My interest in organic farming was not related to consumption but more on the process wherein living things and life itself circulates.
	Judgment	I need to serve as a consolidator of farmland for a certain period and to share my organic business in order for interested learners to operate independently. Fortunately, I have already achieved a social status and good track record with trust and confidence from the community. I have been consolidating limited available farmlands and sharing these with my learners who wish to become an independent farmer.	If no one will do it, I am eager to do it.	I wish to continue and promote farming by hand, which evolves Japanese traditional farming, small-scale farmland and non-utilization of agricultural machineries.
	Action	I will continue supporting new farmers by consolidating limited available farmlands.	Creation of landscapes can attract people and increase number of supporters/friends. To do so, direct and personal connectedness (connections) rather than virtual one is important.	Although it is difficult to have it as a main job, farming as a lifestyle and for subsistence purposes is very much enjoyable. In order to gain more friends, I wish to share my joy and knowledge to young wives.

Source : Life history interview, December 2013

Notes: 1) Original text in Japanese was translated into English by the authors. .

2) When the parents of Farmer YK were still alive, he/she worked with them at times

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In addition to these perceptions, their initial impressions of the Shimosato Farm and Farmer YK led them to even resign from their respective jobs in order to start farm stay. In fact, they even transferred their residence and established their own farms in Ogawa town. These convey their dedication, and love for farming and the village, as evidenced by the following statements:

After training at a dairy farm, I became a salaryman. But not for long, I quit the job and joined Shimosato Farm's farm stay. Right after, I decided to become independent and establish my own organic farm in Ogawa Town. (Farmer A)

After the training at a dairy farm, I went directly to Shimosato Farm. Right after the farm stay, I became an independent farmer in Ogawa Town. (Farmer B)

After resigning from my work at the sewage treatment plant, I joined Shimosato Farm's farm stay. After the farm stay, I became an independent farmer in Ogawa Town. (Farmer C)

With regard to the nature of farm stay, since respondents did not have prior knowledge about farming and farm life, and did not receive 'formal' instructions, practicing was based on *miyo mimane* (observation and imitation) and *nasu koto ni yotte manabu* (learning by doing), as shown by the following statements.

I practiced based on miyo mimane (by observing and imitating) and nasu koto ni yotte manabu (learning by doing). I always followed him. Through connectedness, I was able to take the opportunity to become independent. (Farmer A)

I made sure that I worked with Farmer YK at all times. Mentor-companion relationship. In general, I did everything. (Farmer B)

Since I am not a person who usually asks questions, I learned by observing and imitating while thinking, and doing by trial-and-error. (Farmer C)

The above statements also convey that Farmer YK was in the stage of figuring out how to operate his farm and conduct the farm stay. In other words, it seemed that Farmer YK himself was in a process of trial-and-error for the first few years after his farm started accepting learners. In addition, the selected learners seemed to have experienced *shitei dogyo* (mentor-companion relationship) with Farmer YK since they were of the same age. Through this relationship, they experienced work in fellowship, mutual learning and always looking towards a common purpose.

Speaking of their future visions, the selected learners reveal their greater purposes beyond their own welfare and needs. They clearly address prevalent issues on the lack of farm successors, struggles of new farmers, and detachment from farming and environment, as shown in the following statements:

I need to serve as a consolidator of farmland for a certain period and to share my organic business in order for interested learners to operate independently.

Fortunately, I have already achieved a social status and good track record with trust and confidence from the community. I have been consolidating limited available farmlands and sharing these with my learners who wish to become an independent farmer. I will continue supporting new farmers by consolidating limited available farmlands. (Farmer A)

Creation of landscapes can attract people and increase the number of supporters and friends. To do so, direct and personal connectedness (connections) rather than virtual one is important. (Farmer B)

I wish to continue and promote farming by hand, which evolves Japanese traditional farming, small-scale farmland and non-utilization of agricultural machines.

Although it is difficult to have it as a main job, farming as a lifestyle and for subsistence purposes is very enjoyable. In order to gain more friends, I wish to share my joy and knowledge to young wives. (Farmer C)

With regards to existence and characteristics of learning practices by the second-generation, this study was able to identify that the selected Farmers A, B and C followed their mentor, Farmer YK in fostering motivation and offering learning practices to the youth and other individuals (potential third generation) at their respective farms.

CONCLUSIONS

Focusing on Farmer YK as “teacher and educator” based on his learning initiatives for young farmers through ‘alternative’ farm stay at Shimosato Farm, this study found that the farm stay seemed to be a life-changing experience for the learners, even though their reasons for engaging in farm stay and the duration of their farm stay varied. The learning practices also showed positive impacts, conveying that anyone can benefit from ‘learning without a curriculum’.

Some impacts are as follows: First, among the 90 learners, more than 50% (46 learners) reported that they are currently engaged in farming. This conveys that farm stay seems to play a role in addressing the issue of lack of successors. Second, the three selected learners showed distinctive and spontaneous action towards farm stay and OF, and in fostering the motivation of the next generation. Text-mining analysis and life history interviews revealed that learners considered the importance of connectedness (*tsunagari*), and mentor-companion relationship (*shitei dogyo*) remained strong, even after the farm stay since their ages happened to be near their mentors.

There are two aspects for further study. First, although awareness on pollution and environment has been strongly emphasized by the three selected learners, these did not emerge as keywords based on text mining analysis. Thus, it is important to reexamine the views and activities of the 90 learners regarding this aspect. It is noteworthy that there was high concern for pollution and food contamination in the 1970s and 1980s.

Second, there is a need to research the learning practices of modern exemplary farmers group by clarifying and understanding Farmer YK’s learners (also known as second generation) who offer learning practices (including farm stay) to the youth and other individuals (potential third generation) at their respective farms. There may be some evolution or changes in their practices (with respective impacts) compared to what they have experienced and learned from the Farmer YK (first generation).

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BLUNT-HEADED BURROWING FROG (*Glyphoglossus molossus*) RAISING IN A SUSTAINABLE AGRICULTURAL SYSTEM

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ABSTRACT

Blunt-headed burrowing frog (BhBF) (*Glyphoglossus molossus*) is currently a species at risk of extinction because people catch them for food and also sell them in the marketplace for profit. The objectives of this study were to investigate BhBF nature and to develop the raising method of BhBF in sustainable agricultural system. The experiments were conducted in 2 areas of naturally ecological system; Area 1) Nong Thaag Laan village no. 5, Huay Mon Thong sub-district and Area 2) Kasetsart University, Kamphaeng Saen Campus (KU KPS). Both areas are located in Kamphaeng Saen district, Nakhon Pathom province, Thailand and the experiments were conducted from May 2012 to June 2014. The BhBF breeding trial started with the tadpoles aged 5 days and continued to the age of 20 days which was carried out in 80-cm diameter cement ponds. The density rate of 1,200 tadpoles/pond gave the highest growth with the average length of 2.79 cm. It is not significantly different from the density rate of 1,500 tadpoles/pond (average length of 2.63 cm) but significantly different from the density rates of 1,800 (average length of 2.34 cm) and 2,100 tadpoles/pond (average length of 2.38 cm). No difference in the tadpole length was found between the density rates of 1,800 and 2,100 tadpoles/pond. The tadpole in the density rate of 1,200 tadpoles/pond condition weighed the heaviest (7.61 g) and was significantly different from the tadpoles in other conditions, the density rates of 1,500 (6.25 g), 1,800 (4.71 g) and 2,100 tadpoles/pond (5.60 g), at $p < 0.0001$. No difference in tadpole weight was found between the groups with the density rate of 1,800 (4.71 g) and 2,100 tadpoles/pond (5.60 g). The highest survival rate of BhBF tadpoles was observed in the density rate of 1,800 tadpoles/pond condition and the survival rate is significantly higher than other conditions at $p < 0.0012$. The length of BhBFs from the density rates of 1,200 and 1,500 tadpoles/pond were different from the length of BhBF at the density rates of 1,800 and 2,100 tadpoles/pond. Even though the breeding of BhBFs with the lowest density rate gave the highest lengths and weights, it had the lowest survival rate, hence, it was not recommended. In the Area 1, farmers could catch BhBFs up to 1.5 kg (about 20-30 BhBFs) after one year. In the Area 2, the researcher found that about 20-30 BhBFs had mated and spawned their eggs in a pond 1 year later.

Key words: blunt-headed burrowing frog, feeding, sustainable agricultural system

INTRODUCTION

Thailand is a warm and humid country where the climate is very suitable for amphibians and makes an ideal home for a large number of amphibians. Thailand has ample water resources that are necessary to support the life cycle of amphibians. Rainy season (June - September) is a period of abundant rain and amphibians are very active. Most amphibians lay eggs in water and the tadpoles, the aquatic larvae, live their stage entirely in water before metamorphosing into adults when they then

reside on land. There were 137 species of amphibians reported by The Royal Forest Department (Nabhitabhata and Chan-ard, 2005). Unfortunately, the status of many species has been classified as at risk under the IUCN Red List of Threatened Species because of the influence of climate change and habitat fragmentation, as well as a lack of knowledge about ecological, behavioral, and taxonomic data. This threatened species include a blunt-headed burrowing frog (BhBF) or *Glyphoglossus molossus*, a species of frog in the Microhylidae family, which is a native amphibian in Cambodia, Lao People's Democratic Republic, Myanmar, Thailand, and Vietnam (Nabhitabhata and Chan-ard, 2005; Altig and Rowley, 2014). It is monotypic within the genus *Glyphoglossus*. The world's most avid frog eaters are almost certainly reported in Asia, in countries such as Indonesia, China, Thailand and Vietnam. South America is also a big market (Henley, 2009). Recently, this animal was close to being classified as a threatened species under IUCN because of the effects of forest degradation and over harvesting for food during the breeding seasons by local people. Dijk and Chan-ard (2010) also reported that BhBF is currently a species at risk of extinction because people catch them for food, and also sell them in the marketplace for profit and there are no frog raising for substitution. This situation has affected some certain wildlife animals to the risk of extinction as well as has a negative impact to the economical and social safety of the country (Kaenchan, 2001). If raising BhBF in a sustainable farming system as a new source of income for farmers is developed, it will provide helpful guidelines for the sustainable BhBF production technology. In order to encourage farmers to preserve BhBF through sustainable agriculture, this study sought to investigate the BhBF biology and to develop the raising method of BhBF in a sustainable agroecosystem.

MATERIALS AND METHODS

Nurturing Young BhBFs

Five-day-old BhBF tadpoles were raised from the breeding pond and put into four cement ponds of 80-cm in diameter, filled with water to a depth of 10 cm. Each pond had different BhBF density rates as followings: 1,200; 1,500; 1,800 and 2,100 tadpoles/pond, respectively. Three replications for each treatment were carried out. All of them were fed with protein powder food (including not less than 40% protein) at 5% of the total weights of the tadpoles, once a day, between 7.00-7.30 a.m. After 10 days, the water level was raised to 20 cm. The measurements of their weights and lengths were performed at 5-day intervals until Day 20. Then, the tadpoles were moved to a prepared holding pond prior to release. Nurturing the young BhBFs has been conducted from May16 to June 5, 2012, at KU KPS (Fig 1).

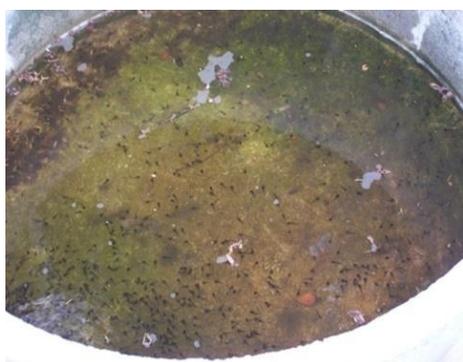


Fig 1. Young BhBFs in 80-cm cement ponds.

Raising BhBF in Areas Practicing Sustainable Agricultural Systems

Young BhBFs were nurtured from June 2012 to June 2013 and then released into 2 areas of sustainable agriculture systems where both areas were selected based on the following criteria; 1) conservation of natural resources, 2) use of local produces instead of using external factors, 3) use of local knowledge base for further development, 4) no pollution, and 5) principles of self-reliance among farmers (Chanthalakkhana and Sakulmun, 2003). The two different areas were Nong Thaag Laan village no. 5, Huay Mon Thong sub-district, Kamphaeng Saen district, Nakhon Pathom province (Area 1) and Kasetsart University (KU KPS) in Nakhon Pathom province (Area 2). Nong Thaag Laan village no. 5 is where a lot of BhBFs were found in the past and the BhBFs were one of their household food in the village. However before this study, there was none of them according to the villagers who wished to have BhBFs again as in the past. Similarly, Kasetsart University (KU KPS) in Nakhon Pathom province (Area 2), a higher education institution, where we previously surveyed and also found none of them. Therefore, both areas of sustainable agriculture system were selected to conduct the experiments to find out whether BhBFs could adapt and survive in these two areas or not. The followings were experimental design of releasing BhBFs in Area 1 and Area 2.

Area 1: Six hundred of 21-day-old BhBFs were released into the natural ecological system, in a collaborative experiment with a farmer in Nong Thaag Laan village. This was done following the procedure of participative research funded by the Thailand Research Fund (TRF) regional research division. This experiment was conducted from June 2012 to June 2013 (Fig 2 and 3).



Fig 2. Farmers were releasing BhBFs into a natural ecological system.



Fig 3. Natural ecological system of Area 1.

Area 2: Six hundred of 21-day-old BhBFs were released into natural ecological system at KU KPS in the same period as Area 1 (Fig. 4).

RESULTS AND DISCUSSION

Nurturing Young BhBFs

The BhBF breeding trial started with the tadpoles aged 5 days and continued to the age of 20 days which was carried out in 80-cm diameter cement ponds. Result showed that the density rate of 1,200 tadpoles/pond gave the highest growth with the average length of 2.79 cm but not significantly different from the density rate of 1,500 tadpoles/pond (average length of 2.63 cm). However, tadpole length from the density rate of 1,200 tadpoles/pond was significantly different from the density rates of 1,800 (average length of 2.34 cm) and 2,100 tadpoles/pond (average length at 2.38 cm) (Table 1). No difference of the tadpole length was found between the density rates of 1,800 and 2,100 tadpoles/pond (Fig 4 and 5).

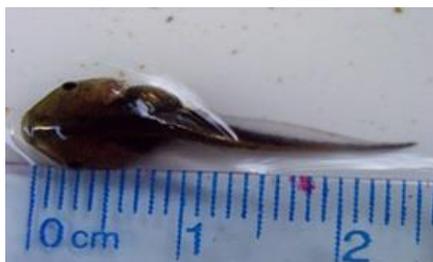


Fig 4. A tadpole of BhBF at the age of 15 days.



Fig 5. A tadpole of BhBF at the age of 20 days.

The tadpole weight at the density rate of 1,200 tadpoles/pond gave the heaviest value (7.61 g) and was significantly different from the weight of the tadpoles at the density rates of 1,500 (6.25 g), 1,800 (4.71 g) and 2,100 (5.60 g) tadpoles/pond at $p < 0.0001$. There was no difference in the tadpole weight, between the groups with the density rate of 1,800 and 2,100 tadpoles/pond. However the results showed that the highest survival rate of BhBF tadpoles was found at the density rate of 1,800 tadpoles/pond and this was significantly different from the remaining treatments at $p < 0.0012$.

Although the breeding of BhBFs with the lowest density rate gave the highest length and weight, it had the lowest survival rate. Hence, it was not recommended. The greater the number of BhBFs was released into the natural ecological system, the greater output was obtained for farmer consumption and selling into the market. From Table 1, the release of BhBF tadpoles at the density

rate of 1,800 tadpoles/pond in the cement pond showed the highest survival rate of up to 96.19%, followed by the density rate of 2,100 tadpoles/pond with a survival rate of 68.92% and 1,200 tadpoles/pond with the lowest survival rate of 47.69%. Therefore farmers were recommended to raise the BhBF tadpoles at the density rate of 1,800 tadpoles/pond (Table 1).

Table 1. Length, weight, and survival rates of BhBFs raised in 80-cm diameter cement ponds at different density rates of tadpoles/pond.

	Treatments				p-Value
	1,200	1,500	1,800	2,100	
	T1	T2	T3	T4	
Length (cm)	2.79 ^a	2.63 ^a	2.34 ^b	2.38 ^b	0.004
Weight (g)	7.61 ^a	6.25 ^b	4.71 ^c	5.16 ^c	0.0001
Survival Rate (%)	47.69 ^c	55.82 ^{bc}	96.19 ^a	68.92 ^b	0.0012

The growth of BhBF tadpoles on the second and third week were shown by their increasing length and weight and the rapid development of front and back legs. The growth of BhBFs at the fourth week showed that their length and weight tended to decrease due to their transformation in shapes and sizes, with shortened tails. Different densities did not affect the growth or give feed conversion but affected its survival rates which were proportional to the density. Although the higher density gave higher survival rate, this is not consistent with those of other aquatic animals. Govindarajulu and Anholt (2006) found that there was interaction of temperature and tadpole densities in which the survival of the tadpoles increased with temperature if the tadpoles were released at low densities, but at high densities, the survival decreased with increasing temperature. In this research, the experiments were conducted during rainy season which the temperature was ranged at 27-30°C but the result demonstrated that at the density rate of 1,800 tadpoles/pond gave the highest survival rate and higher than at the density rate of 1,200 tadpoles/pond.

In addition, Lane *et al* (2011) found that tadpoles raised at a density of 5 per 3 liters (1.6 per liter) began to reach metamorphosis faster than those raised at a density of 15 per 3 liters (5/liter). In our research, the lowest density (1,200 tadpoles/pond) or 12 tadpole/liter gave the highest weight and we also observed their fastest metamorphosis.

According to Soonthornvipat and Soonthornvipat (2011), the survival rate is directly proportional to the density rate. This was assumed for three main reasons: 1) During the first two days, the food given (5% of total weights) to the density rates of 1,200 and 1,500 tadpoles/pond conditions was not totally consumed due to its being an unfamiliar food. There were some food scraps and rotting leftovers, causing death of the tadpoles. 2) The BhBF tadpoles at the density rate of 2,100 tadpoles/pond had a survival rate of 68.92% which was less than the survival rate of 1,800 tadpoles/pond. This could be explained by the better proportion of food with the tadpole numbers, therefore no rotting food was left in the pond to cause the death of tadpoles. 3) The trial breeding of BhBF tadpoles at a density rate of 1,800 tadpoles/pond had the highest survival rate of 96.19%. This number of tadpoles was considered to be suitable to the pond size and corresponded to the previous report of Soonthornvipat and Soonthornvipat (2011) which showed that breeding of BhBF tadpoles should be at the density rate of 2,000 tadpole/m² (p<0.05). No rotting food left in the pond and no problem of insufficient food. At this density, the survival rate was higher than the density rates of 500, 1,000 and 1,500 tadpole/m².

Raising BhBFs in Sustainable Agricultural Systems

In Area 1, the farmers could catch BhBFs up to 1.5 kg (about 20-30 BhBFs) per year after the release of the BhBF (Fig 6). Before the trial breeding, BhBF were not found in Nong Thaag Laan village for the last 10 years. After participating in the project and releasing 600 BhBFs into their natural habitats, the farmers perceived that some BhBFs could be found within one year. It showed that to increase the BhBF numbers, the breeding areas and the appropriate natural habitats must be prepared by introducing a sustainable agricultural system. This enables the BhBF to increase in numbers in the natural ecological system. When the number of BhBFs increase, the farmers can catch them for sale or consumption in order to reduce their family expenses. Therefore, if there is a study on how to raise BhBF in sustainable farming system under principles of benefits sharing among one another, it will help increase the farmer's income.



Fig 6. Farmers could catch BhBFs up to 1.5 kg (about 20-30 frogs) after 1-year of raising.

In Area 2, the researchers found that about 20-30 BhBFs had mated and spawned their eggs in a pond at one year after release. Within the area of KU KPS, where no BhBF could be found for the last 20 years, the researchers released such frogs and subsequently observed BhBFs to have mated and laid a lot of eggs. After releasing about 600 tadpoles to the experimental areas, we found that about 20-30 frogs could be caught in the first year and sold for approximately 450 baht. It showed that proper management of natural ecological system for BhBFs would be really useful to all farmers and the community. A sustainable farming system includes the reduced use of chemical substances and planting of trees. This allows the BhBFs and people to live in harmony. All these elements have positive effect on the environment and encourage the increase of natural species such as the BhBFs (Fig 7 - 9).



Fig 7. BhBF eggs in a pond.



Fig 8. Tadpoles of BhBFs in a pond of Area 2



Fig 9. The natural ecological system in Area 2

CONCLUSIONS

Breeding BhBFs and raising them in their natural habitats or in sustainable agricultural systems can reduce the risk of their extinction and at the same time provide farmers a source of food and additional income. The process described in this research can serve as a guideline to both researchers and farmers for further studies on BhBF or in making BhBF a viable component of the natural ecological system.

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DECOMPOSITION OF THE EFFECTS OF SMALL-SCALE IRRIGATION SYSTEMS ON OUTPUTS OF SELECTED LOWLAND AND UPLAND RICE IN THE PHILIPPINES

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ABSTRACT

In order to better harness the potentials of small-scale irrigation systems, this paper identifies the sources of improvements in rice outputs of lowland and upland ecosystems. Analysis of cross-section data sets for dry season production from 60 lowland rice farms in Laguna in 2010, and 60 upland rice farms in Ilocos Norte in 2012 with the use of the stochastic frontier production function approach showed that using small-scale irrigation systems such as shallow tubewells and small water impoundment projects improves yields of rice farms. The use of small-scale irrigation systems can increase rice productivity through technical change and improvement in technical efficiency. Shallow tubewells were found to be effective in increasing lowland rice productivity because it can shift the production function upward. Meanwhile, productivity improvements in upland farms using small water impoundment projects were largely due to higher technical efficiency attained by these farms.

Key words: climate change, stochastic frontier production function, technical change, technical efficiency

INTRODUCTION

The effects of climate change on the agriculture sector have become very evident, and these are expected to become more serious in the coming years. Climate projections for the Philippines under the Hadley Centre Coupled Model, version 3 (HadCM3) A1B¹ scenario described large seasonal variations in rainfall in 2020 and 2050 (Hilario *et al.*, 2009). It was also projected that the largest seasonal rainfall variation (from -35% to 45%) would be in the months of June to August and March to May.

A major problem in the agriculture sector that can be linked to climate change is the increasing scarcity of irrigation water, which can translate to socioeconomic impacts such as food insecurity and insufficiency. Rapera and Quilloy (2014) noted that there had been massive agricultural losses that resulted from prolonged droughts in key food producing areas in the Philippines particularly in rice producing regions.

¹ A climate change scenario where energy use does not rely too heavily on one particular energy source, on the assumption that similar improvement rates apply to all energy supply and end-use technologies (Hilario *et al.*, 2009).

Irrigation development is a potential means of increasing agricultural productivity in the Philippines. However, irrigation development faces three major constraints (Aragon *et al.*, 2011). First, the bulk of public funds for irrigation is channeled to the construction of new, and improvement of existing, large-scale gravity irrigation systems, i.e., national and communal irrigation systems (NIS and CIS). Second, since the current irrigation development programs are focused on gravity systems, the benefits from irrigation water are offset by the high cost of construction, operation, and maintenance of irrigation infrastructures. Third, the weak institutional arrangements in cost sharing and irrigation management among the different stakeholders, including the users, make these types of irrigation systems cost-ineffective. Roughly 50% of the total irrigable area remains undeveloped. Aragon *et al.* (2011) attribute the sluggish irrigation development in the country to these constraints.

Section 32 of the Philippine Agriculture and Fisheries Modernization Act (AFMA) provides for serious efforts in formulating plans for the promotion of private sector-led development of small-scale irrigation systems (SIS). Ella and David (2001) noted that SISs have short gestation period, cost effective, amenable to privatized ownership and management, and can promote crop diversification. Because of these characteristics, SISs are seen can potentially contribute towards making small-scale farms economically viable enterprises.

Shallow tube wells (STWs) and small water impoundment projects (SWIPs) are among the SISs that have been well studied in the Philippines since early 2000 (David, 2003). A shallow tubewell is a tube or shaft which is set vertically into the ground for bringing ground water to the soil surface through engine suction. Commonly, it is set at a depth of less than 20 m, and it extracts water by means of suction lifting. The limit of suction lift of STWs ranges from 6.7 m to 7.6 m. A single STW unit has the capacity to irrigate four to five hectares of lowland rice areas at an appropriate flooding depth of 5-10 cm during the early stages of growing period. On the other hand, a SWIP is a small reservoir-type irrigation system, which is typically an earthen embankment constructed with spillway facilities (David 2003). A SWIP with a height of 15 m can irrigate a maximum production area of 50 ha. SWIP development in the Philippines roughly covers 12% (22,348 ha) of upland rice areas (Macabasco, 2010).

Irrigation, in general, can increase land productivity or yield of rice farm by about 4 to 7 mt/ha/year (David, 2003) as it enables farmers to plant rice even in dry seasons, thereby doubling the number of cropping per year. David (2003) further reported that STWs have shown the most promise among the SISs that they have studied, but this system is not appropriate in upland areas where drilling towards the shallow aquifer becomes very costly. SWIPs, according to them, is more appropriate for upland agricultural production. STWs showed the highest land productivity improvement in rice production at about 7 mt/ha/year, while SWIPs were reported to perform similarly with national and communal irrigation systems, providing land productivity improvement of roughly 4 mt/ha/year.

The cost-effectiveness of SISs was demonstrated in a simulation modeling study by Ella and David (2001). Using the best estimates of technical parameters such as irrigation intensity, effective cropped area, and crop yield, simulation results showed that STW and SWIP provided higher benefit-cost ratio than either NIS or CIS.

While studies show that SISs promote higher yield in both lowland and upland rice ecosystems, the specific sources of yield or output improvements from these systems have not yet been identified. Information on the specific sources of output improvements is important in order to better harness the potentials of SISs in improving rice output in the country.

THEORETICAL FRAMEWORK

There are two main ways of increasing total farm production. One way is by increasing the levels of input use given the current state of the art or technology. Based on economic theory, this is warranted provided the current level of input use is still less than the economically optimal level. Production can likewise rise through increase in total factor productivity (TFP)—the output increase that cannot be attributed to increased use of inputs. TFP has four component sources, namely: (1) technical change, (2) scale efficiency change, (3) technical efficiency change, and (4) allocative efficiency change (Kumbhakar, Denny, and Fuss, 2001; Balk, 2001 as cited in Coelli, *et al.*, 2005). The first results in a shift in the production frontier or technology. It enables farm to produce more output from a given level of input use. The second results in the farm's improved ability to use inputs and available technology. It enables the farm to produce the maximum possible output, defined by the existing technology, using given levels of inputs. The third refers to the improvement in the size of operations of the farm and its move towards technically optimum scale of operations, e.g. it enables farm to take advantage of economies of scale if there is any. The fourth measures the effects of change in the composition of input vector over time (Coelli *et al.*, 2005). It enables farm to use optimal proportion of inputs.

This study focuses on the first two sources of TFP growth, which can directly result from the use of SISs, as well as the change in the input levels. These sources of TFP growth or output growth are illustrated in Figure 1. Suppose we let Equation 1 be a Cobb-Douglas production function for rice where q is rice output, \mathbf{x} is a vector of inputs, and t denotes time trend:

$$q = f(\mathbf{x}; t) \tag{1}$$

Logarithmic differentiation of Equation 1 with respect to t gives:

$$\dot{q}_t = \beta_t + \sum_i \beta_{it} \ln x_{it} \tag{2}$$

where \dot{q}_t is the percentage growth rate in q over time. β_t is exogenous or neutral technological change because it is the rate of growth in output if growth rates in the levels of all x are zero, that is, levels of input use did not rise. β_{it} is a vector of endogenous or biased technological change parameters which measure changes over time in relative productivity of all inputs, \mathbf{x} , due to the change in the technology.

If a farm does not use SIS in period t , its frontier and average production functions are given by $q_t^{max} = f(x)$ and $q_t^{ave} = f(x)$, respectively. The frontier production function represents the maximum attainable output levels at specific levels of input x and at a given production technology without SIS. A farmer who operates at the frontier is technically efficient. Meanwhile, the average production function gives the input-output combinations when the farm is technically inefficient relative to the production frontier.

Suppose the rice farm adopts SIS at period $t+n$, where $t+n$ denotes some future time period. Suppose further that output increased from q_1 to q_5 between time period t and $t+n$. The difference in output corresponding to the vertical distance from q_1 to q_5 may be decomposed into the respective contributions of higher input use, technical efficiency improvement, and technological change (Quilloy, 2015).

Let the farm utilize \mathbf{x} at the level \mathbf{x}_1 which is below the optimal level, say \mathbf{x}^* , and which gives the technically inefficient output level q_1 . Without an improvement in technical efficiency, increasing input use from \mathbf{x}_1 to \mathbf{x}^* results in an increase in output from q_1 to q_2 provided the marginal productivity of \mathbf{x} is positive. The difference between q_2 and q_1 (measured along the q axis) is the

difference in output due to higher levels of input use.

At x^* , achieving technical efficiency, through improvement in production practices, with respect to the frontier production function of the old technology raises output from q_2 to q_3 . This difference in output corresponds to the contribution of technical efficiency improvement which can be accomplished by improving the ways farm operations are executed at the given technology.

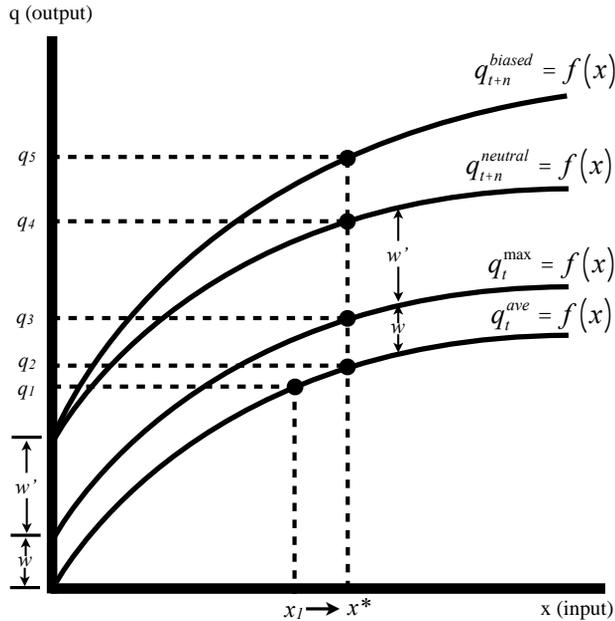


Figure 1. Decomposition of farm productivity gains from adoption of small scale irrigation

Suppose SIS is effectively introduced to the farmer at period $t+n$. When the technology is perfectly adopted such that the farmer becomes technically efficient at this new technology, output will increase from q_3 to q_4 or from q_3 to q_5 , depending on the neutrality of the technology. At input level x^* , the neutral technological effect of SIS is the difference between q_3 and q_4 which is of magnitude w , equal to the shift in the q -intercept. Meanwhile, at the same level of input use, the biased technological effect is the difference between q_4 and q_5 which arises from the difference in the slopes of the two curves (i.e. the difference in productivity after SIS is adopted) at the same level of input use, x^* .

The difference in output at any given level of input use, say x^* , shown by the vertical distance from q_2 to q_5 is the TFP or the increase in output that is not due to the increase in levels of input use. Thus, output can increase by improving TFP and/or increasing input use.

The magnitude of yield effects arising from improvement in TFP and increases in input use described above can be estimated by performing decomposition analysis of the underlying production functions. Recent empirical agricultural productivity studies (Rao 2012, Kumar and Singh 2013, and Hugar and Patil 2007) have employed the decomposition analysis technique in disaggregating the effects of technical change on agricultural production.

Secondary cross-section data obtained from Agrupis (2013) show that mean yields between STW and non-STW farms and those between SWIP and non-SWIP farms have huge disparities. In the

lowland rice ecosystem, the difference in mean yield was roughly 23 cavans² per hectare which is about 26% improvement in rice yield of STW farms over that of non-STW farms. Meanwhile, the yield gap between SWIP and non-SWIP farms was approximately 56.19 cavans per hectare which amounts to about 44% of yield improvement for SWIP farms in comparison with non-SWIP farms.

This paper aims to decompose the output effects of SISs under lowland and upland rice production conditions into the principal components arising from changes in total factor productivity and differences in input levels. In addition, the paper identifies the mechanisms by which small scale irrigation technologies (STW and SWIP) can produce desirable and sustainable effects on rice productivity.

RESEARCH METHODS

In order to decompose the output effects of SIS under lowland and upland rice production conditions, four production functions for rice were estimated. These functions apply to: (a) STW farms or lowland rice farms with STW, (b) non-STW farms or lowland rice farms without STW, (c) SWIP farms or upland rice farms with SWIP, and (d) non-SWIP farms or upland rice farms without SWIP.

Estimation of the Stochastic Frontier Production Function

The estimated production functions of the Cobb-Douglas form followed the stochastic frontier production function specifications of Kumbhakar and Lovell (2000) and Coelli *et al.* (2005). A stochastic frontier production function is a statistical formulation of a production function where output values are bounded above by the stochastic quantity $f(\mathbf{x}_i; \boldsymbol{\beta})e^{v_i}$ (Aigner *et al.*, 1977, and Meeusen and van den Broeck, 1977). The consideration of the potential contribution of technical efficiency on the productivity gains from using SIS was made possible by the use of stochastic frontier production functions in this study.

The general logarithmic form of the stochastic frontier production functions used in this study is expressed as:

$$\ln q_{nj}^s = \ln r^s + \sum_{i=1}^4 \hat{\alpha}_i b_i^s \ln x_{nij}^s + \ln v_{nj}^s - \ln z_{nj}^s \quad (3)$$

where q_{nj}^s = rice output of farmer j ($j = 1, 2, \dots, 30$) in farm site s ($s = L$ for lowland and U for upland) with a given technology n ($n = 0$ for farms without SIS; $n = 1$ for farms with SIS); x_{nij}^s = production input i ($i = 1$ for land area in hectare, 2 for labor in manday, 3 for fertilizer in kg of nitrogen, and 4 for seeds in kg) of farmer j in site s with technology n ; ρ^s = technology parameter ($\rho = \delta$ for farms with $n = 0$; $\rho = \theta$ for farms with $n = 1$). The technology parameters have the following restrictions: $\delta, \theta > 0$.

Equations (4) to (7) respectively describe the production process for non-STW, STW, non-SWIP, and SWIP farms:

$$\ln q_{0j}^L = \ln \sigma^L + \sum_{i=1}^4 \hat{\alpha}_i b_i^L \ln x_{0ij}^L + \ln v_{0j}^L - \ln z_{0j}^L \quad (4)$$

$$\ln q_{1j}^L = \ln \sigma^L + \sum_{i=1}^4 \hat{\alpha}_i g_i^L \ln x_{1ij}^L + \ln v_{1j}^L - \ln z_{1j}^L \quad (5)$$

² Cavan is a local unit of measure that is equivalent to 50 kg.

$$\ln q_{1j} - \ln q_{0j} = (\ln q - \ln d) + \sum_{i=1}^4 \hat{\alpha}_i \ln x_{0ij} (g_i - b_i) \hat{u} + \sum_{i=1}^4 \hat{\alpha}_i g_i (\ln x_{1ij} - \ln x_{0ij}) \hat{u} + (\ln z_{0j} - \ln z_{1j}) + (\ln v_{1j} - \ln v_{0j}) \quad (9)$$

If the terms in Equation (9) are expressed in antilogs, the left-hand side term is the difference in output between farms with and without SIS, respectively. The first term on the right-hand side refers to the change in output due to neutral technological change, the second term pertains to the change in output due to the changes in production elasticities or biased technological change, the third term corresponds to the change in output due to changes in the quantity of in input usage, the fourth term refers to the change in output arising from technical efficiency improvement, and finally, the fifth term captures the difference in output which can be attributed to random error.

Data Requirements and Sources

Two sets of secondary data were used in this study. The first data set comprised of the input-output data for rice production during the dry season (November 2009 to February 2010) from 60 random samples of lowland rice farms in the municipalities of Calauan, Victoria, Pila, and Sta. Cruz in Laguna Province. This data set was composed of production information for 30 STW-irrigated farms and 30 non-STW-irrigated farms. Both farm groups availed of water from the NIS. Non-STW farms relied solely on the NIS for irrigation water during the dry season. STW farms used their STWs during the dry season to augment the irrigation water obtained from the NIS.

The second data set covered the dry season production cycle in 2012 of rainfed rice farms in Pimentel, Batac, Ilocos Norte Province. It was obtained from a survey of thirty randomly selected SWIP farms and 30 randomly selected non-SWIP farms. To avoid selection bias, the chosen SWIP farms did not have sources of irrigation water other than rainfall and SWIP. Meanwhile, non-SWIP farms were solely dependent on rainfall.

RESULTS AND DISCUSSION

Input Use

Input use on a per hectare basis significantly varied between farms which used and did not use SIS for both types of rice production ecosystems (Tables 1 and 2). In lowland ecosystems, STW users had lower per hectare utilizations of labor, fertilizer, and seeds. Increased water availability through the use of STWs could have possibly resulted to better seed germination and better weed control, thereby reducing seed and labor requirements. Nonetheless, the lower use of fertilizer with enhanced water availability was quite contrary to the general expectation that the increased availability of water would induce greater fertilizer use as there would be improved fertilizer productivity through better fertilizer absorption.

Upland farmers have much less control over water availability because there is uncertainty in water supply. If farmers do not harness the potential gains from higher input productivities, they might end up at a loss in the current and succeeding cropping seasons. Hence, everything else being the same, it is strategic for upland farmers to increase input use when SWIP is available.

Table 1. Difference in mean levels of input use of lowland farms by farm group, dry season production cycle, 2010, Laguna, Philippines

Decomposition of the effects of small-scale irrigation systems.....

Variable	Farm Group		Percentage Difference (%)	t-value	P-value
	STW	Non-STW			
Land area, ha	1.80	0.93	93.55	3.9199	0
Labor, md/ha	91.32	124.65	-26.74	-1.8555	0.07
Fertilizer, kgN/ha	92.55	126.80	-27.01	-2.2259	0.03
Seeds, kg/ha	70.00	130.00	-46.15	-3.7435	0

Table 2. Difference in mean levels of input use of upland farms by farm group, dry season production cycle, 2012, Barangay Pimentel, Batac City, Ilocos Norte, Philippines

Variable	Farm Group		Percentage Difference (%)	t-value	P-value
	SWIP	Non-SWIP			
Land area, ha	0.59	0.59	0.05	0.003	1.00
Labor, md/ha	53.04	51.98	2.04	0.1289	0.90
Fertilizer, kg N/ha	152.45	190.89	-20.14	-2.0872	0.04
Seeds, kg/ha	271.84	211.73	28.41	1.9689	0.05

Upland farms who are not SWIP beneficiaries might be cost minimizers as they might tend to minimize their operating capital at risk because they do not have sources of irrigation water other than rainfall during the dry cropping season. On the other hand, farms with SWIP might be output maximizers as they take advantage of the available water from SWIP to effectively increase their yield.

Technical Change

Maximum likelihood estimation of the stochastic frontier production functions resulted in the data presented in Table 3 for the lowland rice ecosystem and Table 4 for the upland rice ecosystem. The regression coefficients measure the percentage contribution of specific production factors to rice output.

For the lowland rice production frontier model with STW, the coefficients for labor and seeds, and the intercept are statistically significant at least at 10% probability level. On the other hand, the coefficient for fertilizer is the only one that is statistically significant at 10% in the rice production frontier model without STW. Meanwhile, the intercept and coefficients for land, fertilizer, and seeds are statistically significant at least at 5% level in the upland rice production frontier model with SWIP. For the upland rice production frontier model without SWIP, the intercept and the coefficients for all considered production factors are statistically significant at least at 5%.

The higher intercept terms for STW and SWIP farms imply that, among other things, SISs have remarkable input-neutral technological effect on dry season rice productivity—the existence of SISs have made production function frontiers higher. These suggest that rice production systems, whether in the lowland or upland, become more productive with the use of SIS.

When water is made available, positive productivity gains can also be observed for each input. The productivity gains manifest as increases in the production elasticities (slope coefficients) of specific inputs. In both ecosystems, labor and seeds appear to be more productive with STW or SWIP. Seed germination can be enhanced with sufficient water during planting thereby encouraging STW farmers to reduce their seeding or planting rates. On the other hand, higher labor productivity is observed for SIS farms because sufficient water during important phases of rice production (especially after planting) aids in weed control, thereby effectively reducing labor use while still achieving higher levels of output.

In the upland rice ecosystem, SWIP farms were observed with higher fertilizer productivity as compared with non-SWIP farms. Because of limited and intermittent water from the impoundment, farmers were better off using more fertilizer and this resulted to higher output. In the lowland, fertilizer was less productive for STW farms as compared with non-STW farms probably because the water requirement of fertilizer might have already been supplied by the NIS.

Table 3. Maximum likelihood estimates of the parameters of the lowland rice frontier production function by farm group, dry season production cycle, 2010, Laguna, Philippines

Item	Farm Group					
	STW			Non-STW		
	Coefficient	t-ratio	P-value	Coefficient	t-ratio	P-value
Intercept	24.15***	53.67	0.0000	-2.32*	-1	0.3269
Land area, ha	3.92**	0.56	0.5805	-0.01*	-0.03	0.9763
Labor, md	0.24**	2	0.0565	-0.29*	-1.12	0.2734
Fertilizer, kg N	0.17**	1.31	0.2021	0.36*	1.89	0.0704
Seeds, kg	0.29**	2.64	0.0141	0.07*	1.4	0.1738
Returns to Scale	4.62**			0.13*		
Log-likelihood	19.39**			6.32*		
No. of observations	30			30		

***significant at 1%

**significant at 5%

*significant at 10%

Table 4. Maximum likelihood estimates of the parameters of the upland rice frontier production function by farm group, dry season production cycle, 2012, Barangay Pimentel, Batac City, Ilocos Norte, Philippines

Item	Farm Group					
	SWIP			Non-SWIP		
	Coefficient	t-ratio	P-value	Coefficient	t-ratio	P-value
Intercept	5.06***	4.93	0.0000	3.14***	6.66	0.0000
Land area, ha	0.91***	3.4	0.0023	0.42***	3.52	0.0017
Labor, md	0.27***	1.44	0.1623	0.21***	2.38	0.0253
Fertilizer, kg N	0.55***	5.48	0.0000	0.28***	6	0.0000
Seeds, kg	0.24***	2.31	0.0294	0.22***	4.23	0.0003
Returns to Scale	1.97***			1.13***		
Log-likelihood	35.58***			75.22		
No. of observations	30			30		

***significant at 1%

**significant at 5%

Technical Efficiency

The mean technical efficiency of each farm group, as well as the distribution of rice farmers according to specific ranges of technical efficiency are shown in Table 5. In both lowland and upland ecosystems, mean technical efficiencies during the dry season were higher and more pronounced for farms which used STW and SWIP, respectively. Furthermore, the difference in mean technical efficiency was higher in upland farms than in lowland farms.

STW irrigation did not cause much improvement in technical efficiency among lowland rice farm given the statistically insignificant difference in the mean technical efficiencies of STW and non-STW farms. Since all farms in the sample are connected to the NIS, the farms' basic water requirement was still met even if STW was not used. STW was used only to augment the water supply provided by the NIS.

Table 5. Frequency distribution of farm technical efficiencies by ecosystem and by and by farm group

Technical Efficiency (%)	Lowland Ecosystem		Upland Ecosystem	
	STW	Non-STW	SWIP	Non-SWIP
	(%)			
96 - 100	23	20	83	17
91 - 95	17	13	17	23
86 - 90	17	10	0	3
81 - 85	10	13	0	37
80 and below	33	43	0	20
Total percentage of farmers	100	100	100	100
Mean technical efficiency	86	80	96	83
Difference in mean technical efficiency	6		13**	
t-value	0.9295		2.0140	
P-value	0.3565		0.0487	

**significant at 5%

SWIP farms were more technically efficient than non-SWIP farms given the statistically significant mean difference of about 13 percentage points. Unlike in the lowland, in the upland, where water is more limited, if farmers do not source their irrigation water from SWIP, farmers are left to depend on rainfall for irrigation. Furthermore, farmers need to optimize the use of the available water stored in the small water impoundment as water losses are inevitable due to natural hydrologic processes, specifically evaporation and seepage.

This result implies that using SWIP might have enabled farmers to utilize their inputs more appropriately and to operate more closely to their production frontier as compared to farmers with non-SWIP farms that depended entirely on rainfall for irrigation. Because of limited water in the upland areas, SWIP farmers might have taken advantage of water availability from SWIP during the dry season. The SWIP might have enabled the farmers to become more technically efficient in production thereby resulting in higher yield.

Components of the Output Gap

Table 6 shows the estimated differences in mean dry season yields (MDSY) in lowland and upland rice production systems. In both ecosystems, MDSY of farms with SIS was statistically higher than that of farms without SIS. Furthermore, the effect of SIS on rice production was more pronounced with upland farms than with lowland rice farms. The MDSY of STW farms was 101.94 cavans per hectare which was roughly a 26% improvement over that of non-STW farms. For SWIP farms, the MDSY was about 157 cavans per hectare, representing a 44% improvement over that of non-SWIP farms.

Table 6. Differences in mean yields of rice by production ecosystem and by farm group

Item	Lowland Ecosystem		Upland Ecosystem	
	STW	Non-STW	SWIP	Non-SWIP
Mean yield (cavans per hectare)	101.94	78.68	156.99	100.8
Difference in mean yield (cavans per hectare)	23.26		56.19	
Percentage difference in mean yield (%)	25.90		44.30	
t-value	1.924		2.842	
P-value	0.053		0.006	

Output differences were disaggregated into the separate contributions of neutral technical change, biased technical change, change in technical efficiency, and changes in the levels of input use. As can be gleaned from Table 7, the major source of output change in lowland rice farms was neutral technical change. On the other hand, the output improvement in upland rice farms was primarily due to improvement in technical efficiency.

In lowland rice farms, roughly 26.47% of the output improvement came from neutral technical change arising from the positive difference between the production frontier intercepts of STW and non-STW farms.

Biased technical change, measured as the sum of the product of differences in production elasticities between STW/SWIP and non-STW/non-SWIP rice production and the quantity of input use in the non-STW/non-SWIP rice production, accounted for about 2.42% difference in output in the lowland ecosystem. It can be observed from Tables 3 and 4 that technical change involved in shifting from non-STW/non-SWIP to STW/SWIP is biased towards all inputs (except for fertilizer in the STW rice production) as evidenced by higher partial elasticity coefficients for all inputs in the STW and SWIP rice production frontier model. This means SIS technology is land-, labor-, fertilizer-, and seed-using technology.

The output effect of the differences in the levels of input use was measured as the sum of the product of the differences in the quantity of input usage between STW/SWIP and non-STW/non-SWIP farms and the respective production elasticities of the STW/SWIP rice production. This component also had modest contribution to the difference in output between the two lowland farm groups. Higher land area planted brought about around 2.59% improvement in output. The reductions in the use of labor, fertilizer, and seeds had negative but negligible effects on lowland rice output. The differences in the rates of input use between STW and non-STW farms brought about a net output improvement of approximately 2.28%.

Table 7. Percentage contribution to rice output due to STW/SWIP

Source of Difference in Output	Type of Irrigation Technology	
	STW	SWIP
	(%)	
A. Neutral technical change	26.47	1.92
B. Biased technical change	2.42	2.79
Land	-0.29	-0.26
Labor	2.56	0.24
Fertilizer	-0.92	2.66
Seeds	1.07	0.15
C. Difference in input use	2.28	-0.06
Land	2.59	0.00
Labor	-0.07	0.01
Fertilizer	-0.05	-0.12
Seeds	-0.18	0.06
D. Difference in technical efficiency	6.00	13.00
E. Predicted percentage difference in output (A+B+C+D)^a	37.17	17.65

^a Calculated based on equation 9 and represents the sum of the principal components of the yield effects of STW/SWIP based on the respective production frontier functions

The final component of the output effect of STW irrigation was the difference in technical

efficiency between STW and non-STW farms. The difference in technical efficiency between these two farm groups brought about approximately 6% increase in output. The predicted total output effect coming from all the principal output gap components in lowland rice farms amounted to about 37%.

In contrast with that in the lowland ecosystem, the percentage difference in dry season output between SWIP and non-SWIP farms was largely a result of higher technical efficiency of SWIP farms. The higher technical efficiency of SWIP farms caused around 13% difference in output among upland rice farmers. Technical change accounts for roughly 4.71% of the improvement in output between SWIP and non-SWIP farms, which in comparison with that in the lowland condition is significantly lower. Of the total technical change effect, neutral technical change caused a 1.92% increase in output. Meanwhile, biased technical change brought about 2.79% increase in output.

Improvements in the productivities or production elasticities of land, labor, fertilizer, and seeds gave rise to the biased technical change. The productivities of labor, fertilizer, and seeds positively contributed to higher output among SWIP farms. Conversely, higher land productivity had a negative effect on the output difference.

The final contributor to the output gap is the difference in the levels of input use. The increase in the quantities of land, labor, and seeds led to higher output among SWIP farms. Meanwhile, the reduction in the fertilizer use had contributed negatively to the output level. As compared with non-SWIP farms, SWIP farms utilized significantly lower level of fertilizer during their dry season production cycle.

CONCLUSIONS AND POLICY IMPLICATION

As the environmental conditions resulting from climate change become more and more unpredictable, rainfall now and in the coming years might not coincide with its patterns and intensities as observed during the past decades. This has serious implications as to when farmers should plant and harvest, how much they should produce, and how much input they should use. Failing to adapt to these new environmental conditions may render tremendous agricultural losses in the agriculture sector, particularly in rice production.

One of the effective and sustainable options for improving rice yield despite the challenges posed by climate change is the use of small-scale irrigation systems. Through decomposition analysis, this paper analyzed the output effects of using shallow tubewell irrigation in lowland rice production ecosystems and small water impoundment project in upland rice production ecosystems.

Using small-scale irrigation systems, such as STW and SWIP as means to bridge the gap in water supply in rice production during dry cropping periods, improves yield of rice farming by triggering structural changes in the rice production function. These structural changes come in the form of higher intercepts, production elasticities, and technical efficiency. However, the magnitudes of the source structural changes vary between lowland and upland rice ecosystems.

Using STW irrigation in lowland rice production increases farm output primarily because of neutral technical change. This suggests that at the very least, STW irrigation can shift the farmer's production function upward. Thus, without even improving individual input productivities, STW irrigation enables rice farms to produce relatively higher levels of output at low input levels.

The SWIP aside from shifting the production function of upland rice farms upward, primarily allows farmers to operate closely to their production frontier as evidenced by the positive difference in technical efficiency between SWIP and non-SWIP farms. Hence, the positive output effect of using SWIP is based on its ability to enhance technical efficiency of upland rice farming.

The above-mentioned advantages of using STW in lowland and SWIP in upland rice production ecosystems emphasize the relevant role of SIS in increasing rice production. However, the expected improvement in yields due to SIS might be realized in other rice production areas if the choice of irrigation system is anchored on the type of ecosystem within which it is situated.

Based on the findings of this paper, policy strategies should be formulated to promote the use of small-scale irrigation systems in lowland and upland rice production areas in support of the country's irrigation development thrust, whose long-term goal is the sustainable improvement in national rice farm productivity. The policy strategies should focus on identifying effective means to promote widespread and sustainable use of appropriate small-scale irrigation systems in varied rice production ecosystems.

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**CHALLENGES FACED BY PHILIPPINE AGRICULTURE AND UPLB's
STRATEGIC RESPONSE TOWARDS SUSTAINABLE DEVELOPMENT AND
INTERNATIONALIZATION**

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ABSTRACT

The Philippines' being an agricultural country need to invest in promoting inclusive growth, and build more sustainable agriculture and food systems, that are resilient to calamities and respond effectively to climate change impact.. The main goal is to develop the agriculture sector to be able to attain food self sufficiency, uplift the rural community and increase the income of the farmers. Agriculture's contribution to the economy is estimated to be about 40 per cent of gross domestic product (GDP) and two-thirds of jobs arise from agriculture. However, agricultural sector has been beset with persistent challenges resulting in low farm incomes, low rural employment, lack of food security, and meager agricultural competitiveness. Among the 17 targeted areas of the Global Sustainable Development Goals, the Philippines should specifically pursue the goals to end/minimize hunger, achieve food security, improved nutrition, and promote sustainable agriculture.

The status of Philippine agriculture as well as the challenges and strategies for sustainable development were discussed. Challenges identified were lack of important provisions that will guarantee food security and reduce poverty in the countryside, lack of programs that will link agriculture with the industry, and insufficient activities aimed to vigorously transfer advanced production, post-harvest, and processing technologies to the stakeholders, among others. The University of the Philippines (UPLB), as the premier university offering agricultural degrees in the country serves as the leader in developing network of an integrated system in agriculture education that effectively responds to the challenges faced by the agriculture sector. For instance, UPLB's programs in the attainment of food and nutrition security as well as its visions in internationalization were discussed. Programs and plans on responsive and innovative teaching methods and strengthen RDE integration were adopted, geared towards the achievement of the four dimensions of food security.

INTRODUCTION

Significant achievements in the areas targeted by the Millennium Development Goals (MDGs) established in 2001 and ending in 2015 were achieved. However, there were still many unfinished business that need to be tackled globally. Hence, major challenges were identified to address a more ambitious new agenda for sustainable development. During the United Nations Sustainable Development Summit in New York, USA last September 27, 2015, more than 150 world leaders agreed to adopt the global Sustainable Development Goals or SDGs (UNDP, 2015). The SDGs followed and expanded the MDG by defining a common agenda to achieve inclusive growth, end poverty, and protect the environment by 2030. In order to realize these goals there should be strong leadership at all levels from multilateral system and broad coalition of world leaders in order to build more environmentally sustainable agriculture and food systems and well designed social protection systems (Da Silva, 2015)

The SDG's consisted of 17 targeted areas envisioned to be accomplished by 2030 (Denning, 2015). These were to sustainably end poverty, zero hunger, attain good health and well-being, quality education, gender equality, clean water and sanitation, affordable and clean energy, decent work and economic growth, industry, innovation and infrastructure, reduce inequality, sustainable cities and communities, responsible consumption and production, climate action, life below water, life on land, peace, justice and strong institution and partnerships for the goals. Specifically, the goals to end hunger, achieve food security, improved nutrition and promote sustainable agriculture, should be urgently pursued as rapid progress in these areas is the key to the other goals (da Silva, 2015). In addition, harnessing the power of technology, mobilizing new and open data sources, and partnering across sectors are key elements in achieving these goals faster and make their progress more transparent to the stakeholders in the local, regional, national and global (Anonymous, 2015).

The key to achieving the objectives of SDG by 2030 are food security, nutrition and sustainable development in agriculture. The Philippines being an agricultural country needs to invest in promoting inclusive growth, and build more sustainable agriculture and food systems, that are resilient to calamities and respond effectively to climate change impact.

STATUS AND CHALLENGES OF PHILIPPINE SUSTAINABLE AGRICULTURAL DEVELOPMENT

Agriculture is still a key component of the Philippine economy and agricultural development remains having a significant role in the advancement of rural communities. Although agriculture, fishery and forestry represent just about 20 percent of the economy's domestic output (GDP), about 37 percent of jobs still depend on agriculture. If outputs related to agro-processing, agricultural inputs, manufacturing and trading (i.e. agribusiness sectors) as well as basic agricultural production are considered agriculture contributions to the economy, about 40 percent of GDP and two-thirds of jobs in the economy arise from agriculture (Pangilinan, 2015). Despite the successes gained in agricultural development by the Philippines in the late 1960's, the agricultural sector has been beset with persistent challenges resulting in low farm incomes, low rural employment, lack of food security, and meager agricultural competitiveness.

Although the ultimate goal of attaining agricultural development has not yet been achieved, attempts of several administrations in the country have been geared towards its attainment. These included projects of the Department of Agriculture such as reforms made to enable farmers to enjoy higher farm gate prices and to establish a rural credit system, projects to speed up farmers' organizations access to financing management expertise and marketing rice and corn production enhancement programs, assistance for farmer to form functional groups and cooperatives, food production and food security programs, Agriculture and Fisheries Modernization Act of 1998 or AFMA (with visions of transforming and modernizing the country's agriculture and fisheries sector), countryside assistance for rural employment and services, transport programs and interventions for agricultural products, and two million hectares of new lands developed for agribusiness.

The most recent report of the current administration regarding agricultural development was embodied in the State of the Nation Address of President Benigno S. Aquino. The report has mentioned that challenges to agricultural development have been further intensified by the current population of about 100 million and yearly natural calamities (like typhoons and El Nino phenomenon) that affect agricultural productivity. The current administration reported that two sectors made breaking records in rice production and in introducing stricter laws against illegal fishing. More than half of irrigable land in the country is now covered by government irrigation facilities exceeding the target. With financial assistance from the World Bank, the DA is constructing 213 kilometers of farm-to-market roads with 11% more than half-way completion. Establishment of agricultural trading centers, a depot where farmers can bring their crops and sell directly to customers, was also reported. Rice production rose by

4.98% from 2010 to 2014 although intended 100% rice self-sufficiency was not achieved in 2013 as predicted. Corn production volume and values grew and economic gains from high-value crops like coconut, sugarcane, banana, mango, livestock, poultry, and fish have also increased. The current budget requested for agriculture will be predominantly focused on meeting rice production and consumption targets, completion of irrigation projects for more agricultural lands, ensuring food buffer stocks to prepare for any shortage providing credit for farmers and fisher folk and research and development for agriculture.

Challenges faced by agricultural sector were identified. Among the apprehensions about the current national agricultural development agenda is that projects lack important provisions that will guarantee food security and reduced poverty in the countryside, lack of provisions that agriculture could easily be linked to industry, no provisions to enable farmers to semi-process their own products and improve the marketing and distribution of local agricultural products, farmers are not provided with better opportunities to be organized in order to link them to markets (like restaurant chains, supermarkets, food processing companies), and lack of plans for improving land tenure for farmers. Efforts to eliminate or minimize corruption in the government as well as to address smuggling and other irregularities have been pointed out.

STRATEGIES FOR SUSTAINABLE AGRICULTURAL DEVELOPMENT

An important strategy for sustainable agricultural development in the country is to re-orient focus programs toward farmer-centered development. The program should be income-based to enable farmers to access of markets, packaging, value-added processes and post-harvest facilities (Pangilinan, 2015). More so, to include practices that are economically viable, socially responsible and ecologically sound, where sustainable agricultural development is measured by income derived by the farmers.

Strategic approaches towards strengthening agricultural education requires revisiting and formulating plans for offering quality programs that are designed to develop students' skills and attitudes that respond to the need of the industry. The role of agricultural education institutions and its contributions to academic, research and development, technology transfer and entrepreneurial programs are expected to bring about innovations in agriculture and related fields. At present, there are about 124 universities and colleges, some of them have several campuses offering agricultural sciences degree. These schools were strategically located all over the country, an advantage in bringing the opportunity for the students in rural areas to take up agriculture as their profession if given incentives and financial assistance to deserving students. The RDE activities of these schools generate technologies that were responsive to the need in their respective locations, provide training to farmers and agricultural technicians, serve in facilitating information disseminations and effective communication and assist the LGU's in the preparation of their respective agriculture development (Rola, et al., 2012)

The University of the Philippines Los Baños (UPLB), the premier university offering agricultural degrees in the country serves as the leader in developing network of an integrated system of agriculture education that effectively responds to agriculture and rural development needs of the country. In partnership with the Commission on Higher Education (CHED), UPLB, endeavours to address the growing concerns or issues of the drastic decline of student enrollees in agriculture and agriculture-related courses, including the offering of programs that are responsive to the needs of the domestic and international agencies and most importantly the production of highly competent and competitive graduates, equipped in meeting the demands of the industry and the labor market (Anguilo-de Asis, 2013). In line with this, CHED initiated the passage of Agriculture Education Act of 2010 or an act rationalizing agriculture education in the Philippines by establishing a national system of agriculture education institutions, providing for mechanisms of implementation, and for other purposes urges the state to establish, maintain, and support a complete an integrated system of agriculture education relevant to the needs of the economy, community and country.

UPLB identified issues and strategic/innovative approaches on academic, R & D and mode of delivery of technical outputs in strengthening agricultural sciences. Specifically, it includes initiatives on UPLB's innovative teaching methods of faculty in degree, non-degree and graduate programs geared towards preparing graduates for global opportunities and competitiveness. The College of Agriculture (CA) developed responsive programs such as institution of new programs and courses. These are the B.S. Agricultural Biotechnology (with 13 new courses), BSA major in Landscape Agroforestry and new courses in BS Food Technology (Angeles, 2014). In the pipeline are BS Landscape Agriculture, BSA major in Organic Agriculture, MS in Food Control system and MS Food Security, among others..

UPLB College of Economics and Management (CEM) included agricultural entrepreneurship in their program to invoke that agriculture is a prestigious and profitable profession and not just cultivation of soils or cutting grasses but making money and business. Action researches were encouraged to promote an entrepreneurial mindset among UPLB students, faculty and researchers (Pabuayon and Depositario, 2015). The program includes promotion of entrepreneurship as an option to student's specialization and acceleration of commercialization of UPLB technologies. A multi-disciplinary approach was adopted with community-based involvement in holistic research and development efforts towards attaining the goals and objectives set for uplifting the lives of rural farmers as well as the general population.

CEM-UPLB is currently managing the UP Center for Agribusiness for Entrepreneurship, a multidisciplinary program envisioned to integrate small farmers into the market-driven economy through entrepreneurship. Specifically, the center aims to 1) develop a new generation of farmers and entrepreneurs through research, training and public service; 2) strengthen UPLB's partnership with agribusiness industry, government, and farmer groups; and 3) promote transdisciplinarity and sustainability in key agribusiness programs. These activities will improve the performance and sustainability of local entrepreneurs and SME's, which accounts for majority of businesses and employment in the Philippines (Pabuayon and Depositario, 2015).

UPLB's PROGRAMS FOR THE ATTAINMENT OF FOOD and NUTRITION SECURITY AGENDA

Food security is defined by the World Health Organization (WHO) as existing when all people at all times have access to sufficient, safe, nutritious food to maintain a healthy and active life. There were four dimensions of food security, namely, food availability, physical food access, economic food access, and food utilization ((Teng, 2015). Food availability concerns the agricultural and fisheries production poor performance, effect of environmental degradation and climate change, increased demand for food due to population growth, urbanization and higher income and changes in food preference. Food access is affected by modern supply chain in food distribution and trade and higher food prices. Food utilization issues include malnutrition, access to better food quality, food safety and food loss and waste.

UPLB's contribution to the SDGs is the creation of the **Interdisciplinary Studies Center on Food and Nutrition Security and Safety (ISCFNS)** in 2014 to promote food security through research, instruction and extension. Its main mission is to promote food security through research and education taking into consideration the many intricacies of providing sufficient and nutritious food to the country's population. The UPLB-ISCFNS is envisioned to be a national center of excellence in interdisciplinary research, teaching and training on food and nutrition security. The center will be identified as a national leader in manpower development, research, policy advocacy, capacity building and knowledge management in country's national food and nutrition security. It is a center "without walls" under the Office of the Chancellor whose function is carried by an interdisciplinary group headed by a chair with members from various disciplines across nine colleges and one school.

The research thrusts of the Center revolves around the four elements of food security.

A. Production and Postproduction System

1. Sustainable Food Production System: Seed production and processing system, breeding for high yield and quality and tolerance to environmental stresses; land use and suitability mappings, water and fertilizer use efficiency; higher cropping intensity; farming system; pest surveillance and management; site-specific technologies; farm mechanization; postharvest handling system; diversification of food sources; food processing of alternative staples; functional nutrient analysis of other staples; organic agriculture; smart agriculture; precision agriculture; urban agriculture and production in changing climate.
2. Agricultural Innovation: Creative research for efficient production and distribution; novel and basic research on varietal development, seed systems, production and postproduction; food processing; food quality; marketing and trade; and nutrition and consumption.

B. Food Safety, Nutrition and Consumption

1. Food Product Quality and Safety: Safety standards like GAP, HACCP, GMP; pesticide residue; microbial analysis; heavy metals analysis; diet diversification; and dietary intake analysis.

C. Trade and Distribution System

1. Socio-economics and policy research: Market price and income dimensions of the food system; societal strata as element of food access; economic analysis of food safety regulations; buffer stock management and trade policies; cost and benefits of technology adoption in the supply chain; supply chain efficiency; impact of AEC 2015; supermarket revolution; connectivity and efficient logistical services.

D. Knowledge Management

1. Capacity building: Structural administrative processes on food security; training and skill development, fora, symposia, scholarship, information retrieval and storage, R & D institutions directory and capacity on food security.
2. Food Security Communication and Early Warning System: Information retrieval, storage and communication, website management; production modelling, pest epidemiology; flooding and drought forecasting; quality forecasting; IEC, policy papers, occasional papers.
3. Food Security Education: Graduate and undergraduate curricular programs on food security and safety eg. MS in Food Security, MS in Flood Control Systems; PhD in Development Studies major in Food Security

The first major activity of the UPLB-ISCFNS was the recently concluded National Conference on Food and Nutrition Security held on October 12-13, 2015 at Crimson Hotel, Philippines. The objective of the conference is to set the strategic directions and actions on the Philippine Food and Nutrition Agenda. Specifically, the conference aims to 1) discuss the pressing issues, imperatives and challenges surrounding the dimensions of food and nutrition security, 2) present the current status of

Challenges faced by Philippine agriculture and UPLB's strategic response.....

the food security in the country, 3) discuss the role of the environment for sustainable food production, and 4) formulate strategies and directions adding to food security research, development and policy.

There were 3 plenary lectures, namely, Global Food Systems and Sustainable Development Goals; From Farm to Table to Farm: Issues on Food and Nutrition Security and Partnerships for Food Security. Twenty (20) invited papers dealing on food availability, accessibility, utilization and stability were presented. Workshops were held with the participation of 14 government agencies, state and private schools, colleges and universities, industries, individuals, among others.

The workshop outputs identified the constraints and researchable areas in the four elements of food security, such as:

- 1) Availability: Inadequate information on the supply chain of different commodities; insufficient water irrigation facilities in the production areas; failure of micro-scale food businesses to be accredited by the national regulatory agency; and lack of more advanced food preservation and processing technologies.
- 2) Accessibility: Constraints identified were the high cost of conventional production system; extreme weather variability that affected the crops, livestock and poultry; and the sites of the National Greening Program were located in upland and remote areas, making it inaccessible to the clientele;
- 3) Stability: The stability of agricultural production were due to climate change; environmental degradation; and effect of natural calamities as well as the perishability of agricultural products; and
- 4) Utilization: Problems identified were food safety especially on street foods; lack of awareness on nutrient composition of stable food and acceptability of type of food preparation especially to children. The conference culminated with the presentation of the draft Food and Nutrition Security RDE agenda to stakeholders.

UPLB RESPONSE TO CHALLENGES IN INTERNATIONALIZATION

In December 2015, the ASEAN Economic Community (AEC) will commence the transformation of ASEAN into a single market and production based in the region. One of the important components of integration is the role of education in providing skilled labor and services. My paper deals on how the University of the Philippines (UP) as a national university, contributes to the internationalization efforts of higher education sector and how UPLB as a constituent unit, respond to both and efforts of the Commission on Higher Education and the UP System. Specifically, it will focus on the important role of UPLB in manpower development equipped with agricultural knowledge in science and technology (Sanchez, 2015).

As lead agency responsible for formulating plans, policies and programs for the development and improvement of Philippine higher education system, the CHED has laid down roadmap for Higher Education Reform Agenda with the main goal of providing relevant and quality public higher education accessible to all who seek and deserve it. Apart from the roadmap, CHED also plans to carry out: 1) A policy framework for the internationalization of higher education; 2) The inclusion of more Philippine universities in the AUN; 3) The participation in the ASEAN International Mobility of Students (AIMS) Programs and 4) The development of a better credit transfer system and diploma supplement which will enable the comparability of Philippine higher education with those of other countries.

Under its Charter of 2008 (Republic Act 9500), UP is mandated to perform its unique and distinctive leadership in higher education and development. President Alfredo E. Pascual presented a blue print for a great ideal that is “One University, One UP” The plan includes strategic initiatives such

as: 1) Recruiting the best and the brightest students as we continue to address issues of admission and democratic access and ensure that those who are qualified are able to enrol; 2) Strengthening the staff with enough incentives to retain them so that they improve their capacity particularly in research; and 3) Intensifying internationalization and formalizing quality assurance.

With ASEAN integration, it would be expected that the advantage of our graduates will somehow be challenged by graduates of other ASEAN universities. Hence there is a need to develop graduates with core competencies and ensure that the curriculum is consistent with the ASEAN 2015 qualification framework requirements.

UPLB Strategic Vision

Adhering to the CHED Action Plan (2011-2016) and UP Strategic Plan (2011-2017), UPLB has laid down its own Strategic Plans to achieve its vision of a globally competitive graduate and research university contributing to national development. We will strive to meet three core objectives: 1) To sustain academic excellence and leadership; 2) To utilize UPLB's knowledge and technologies toward inclusive growth; and 3) To create an enabling environment for creativity and innovation

Objective 1: To sustain academic excellence and leadership:

1) Academic Programs: Build on existing academic programs by expanding and revitalizing degree offerings such as Satellite and off campus degree programs, dual degree and honors programs, AUN accreditation and straight PhD programs; and

2) Knowledge and Capacity Exchange Program: Foster knowledge sharing and collaboration among UPLB constituents such as formal/informal learning sessions, mentoring sessions, academic leadership conference, webinars with foreign universities and visiting professorship

Objective 2: To utilize UPLB's knowledge and technologies toward inclusive growth:

1) Strategic Inter-disciplinary Action Program: Enhance UPLB's inter-disciplinary niche on food security and safety. This includes: Strong food and security and safety centers and innovation hub; and

2) Knowledge and Capacity Exchange Program (External): To ensure the testing, application and diffusion of UPLB's knowledge and technologies thru projects on continuing education program and public service.

Objective 3: To create enabling environment for creativity and innovation:

1) Resource Mobilization: Raise and mobilize necessary resources for sustainable programs which include external support portfolio for donors;

2) Policy and Implementation Improvement Program: Address inefficiencies that hinder innovation and academic excellence such as projects on campus safety and security; procurement procedure reforms, and streamlining of processes; and

3) Communication: Improve the projection of UPLB to the broader community, attract investments and highly qualified students and personnel such as UPLB brand and alumni engagement

UPLB Interdisciplinary Studies Centers

As an effort to put in place formal structures to encourage faculty and researchers from different disciplines to work together, UPLB launched 18 interdisciplinary studies centers, envisioned to boost scientific and technical know-how in four clusters: agriculture, technology, environment and development. The most active Centers currently pursuing international linkages for research collaboration include: 1) Interdisciplinary Studies Center on Food Security and Safety; 2) Interdisciplinary Biofuels Studies Center; 3) Climate Risk Studies Center; 4) Center for Integrated Natural Resource and Environment Management; 5) Nanotechnology Program; and 6) Natural Products Interdisciplinary Studies Center

CONCLUSION

In conclusion, the challenge of achieving the Sustainable Development Goals is realized if there will be collective efforts of the global community (UNDP, 2015). Every country is enjoined to participate in SDG agenda to be able to contribute in nourishing 9.5 to 13.3 billion people by 2100. Denning (2015) stated that the targets toward Sustainable Global Food System can only be realized when all four dimensions of food and nutrition security are addressed.

1. **Availability:** Improved productivity through increase agricultural productivity using new technologies; reduce losses; encourage sustainable international trade; and supportive agricultural policies.
2. **Accessibility:** Improved distribution and access through modern supply chains (inclusive), capacity for distribution food, capacity for food trade, affordability of food spikes and safety nets.
3. **Utilization:** Issues on food utilization include: nutrition security, linkage to poverty, food quality and safety and malnutrition. Solutions on these issues are reduction food losses and wastes by reducing post-harvest and processing losses through technology, infrastructure, information, and policies and reduction of retail and consumer waste through education, advocacy and policies
4. **Stability:** Involved nutrition security (shift to healthier diet), food safety (improve infrastructure and hygiene). Producers should promote diversification and nutrition-sensitive agriculture while consumers should promote dietary diversification and balanced nutrition; discourage overconsumption; improve food safety through education, advocacy, policies and research.

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