

KEYNOTE SPEAKERS

GLOBAL CLIMATE CHANGE: NUTRITIONAL STRATEGIES FOR SUSTAINABLE LIVESTOCK PRODUCTION

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ABSTRACT

Environmental scientists have agreed that world is experiencing a slow but steady increase in global temperatures. Studies in the last few decades have shown that there was an increase in the global temperature by about 1.5o C over the last 50 years, and that if global warming is not mitigated we will be experiencing higher temperatures in the next fifty years to come. High temperatures can affect not only the productivity of animals but also can lead to increased breeding of certain animals and possibly extinction of animals. Crops and feed production will be affected due extreme weather conditions (such as drought, floods and heat wave) that arise from the high temperatures. It has been established that global warming is caused by greenhouse gases that are produced as a result of human activities although natural emission of GHGs are also contributing to the global warming. GHGs include carbon dioxide, methane ozone, CFCs and others. With the increase in world population it is inevitable that global temperature will be on the increase as a result of increased in human activities. The factors that contribute to the GHGs include methane gas emission from animals and livestock activities, factories, automobiles and also natural organic decomposition. Agricultural activities that increase GHGs include clearing of forests, livestock production, aquaculture and fodder and feed crops cultivation and undoubtedly the generation of energy from fossil fuel for machineries. It is known that ruminant's produces methane gas through rumen microbial fermentation and is expelled to the atmosphere through the process of eructation. Scientists have estimated that as much as 18% of GHGs from agriculture activities is attributed to ruminant production sector. For future sustainable livestock production, animals must be kept in efficient housing systems, fed available feedstuffs that are renewable, employing practical precision feeding, with low fossil energy inputs. Green technology will be used for farm activities to reduce waste and use of fossil energy. Animal nutritionists can play an important role in reducing the methane production by increasing the efficiency of livestock production through manipulation of rumen fermentation, precise feeding and the use of natural additives as growth promotants. From the holistic point of view, global warming issues in livestock production can be addressed in terms of mitigation of emission of GHGs (slow down global warming) and adaptation to the changing climate. For mitigating GHGs emission research need to be conducted in breeding animals for heat tolerance, improved efficiency in terms of reproduction and feed utilization, controlled environment and disease control. Biotechnology in crop science has produced crops that are heat, insect and fungus tolerant, and therefore have paved the way the future crops will look like. These GMO crops will play major roles in the future to providing food for people and animals. Production of GMO crops will need to be regulated so that they do not present threats to the human health. Efficiency in livestock production encompasses the production systems, feeding strategies and reproduction. As new varieties/breeds of animals are being introduced into developing countries, these animals need to be adapted to new environment. New housing systems need to be developed (with low energy input) to overcome high temperatures and heat stress. Feeding formulated diets designed to reduce heat stress is an important step towards precision feeding of animals. In fact, precision feeding (that is feeding exactly to what the animal needs) leads to high efficiency, low waste production and reduce

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feed costs. With the anticipated global warming animal productionists are faced with new challenges such as new emerging diseases, emerging predators and vectors, new animal husbandry regulations and management systems, and above all limiting feed resources. If animal scientists work closely and in collaboration with technologists the problems of housing, energy use at farm and new feed resources techniques in precision feeding can be resolved.

Keywords: *global warming, precision feeding, methane emission, nutrition under heat stress, GMO feedstuffs*

STRESS, NUTRITION AND IMMUNE REGULATION IN PIGS

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ABSTRACT

With respect to fast growing and global changes of international atmosphere, stresses have been concerned for decades in livestock industry. Major stresses including heat, nutrition and infection could alter not only the growth performance, but also systemic and local immune system. It is also well known that major stresses impact on gut health. Heat stress (HS) increased the permeability and the inflammatory responses in the gut. Nutritional stresses, such as fasting or fed with mycotoxin contaminated feed, induced the destruction of the tight junction proteins in the gut. Fasting suppressed pro-inflammatory cytokines, whereas deoxynivalenol (DON) up-regulated the recruitment of intestinal pro-inflammatory cytokines and the level of lymphocytes in gut. Pigs infected with pathogens such as Enterotoxigenic *E.coli* (ETEC) and porcine epidemic diarrhea virus (PEDV) lead to loosen up the intestinal epithelial barrier. On the other hand, supplementation of *Lactobacillus plantarum* or *Saccharomyces cerevisiae boulardii* reduced infectious stress by ETEC. It was noting that major stresses altered the permeability of the intestinal barriers and profiles of genes and proteins of pro-inflammatory cytokines and chemokines in porcine gut. However, it is not sufficient to fully explain the mechanism of gut immune system in pigs under stress condition. In near future, the interaction of gut and systemic immune system under major stresses should be defined precisely to overcome aforementioned obstacles.

**THE USE OF FEED BIOTECH PRODUCTS AS AN ALTERNATIVE TO SUPPORT
IMPROVEMENT OF POULTRY PRODUCTION**

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ABSTRACT

Development and use of biotechnology food products in support of improving the current livestock productions current and growing very fast. GM feed that has been circulating a lot in the poultry feed industry such as probiotics, prebiotics, phytobiotic, synbiotic, acidifiers and single cell proteins and genetic modified organism. The use of probiotics as feed additive has much to do as in broilers and laying hens. The use of probiotics in broiler feed did not affect production performance but can improve the quality of production performance of broiler meat as a decrease in cholesterol, abdominal fats content and yellow coloring of legs. The use of probiotics in the feed of laying hens lay eggs can extend, enhance and animal health, egg quality of laying hens. The use of other biotech products have helped increase chicken health, performances production quantity and quality of poultry production. The use of biotech feed products to day can even replace antibiotics in poultry production support.

Keywords: *Feed, Biotech Product Livestock production*

**MULTIFUNCTION ROLE OF TROPICAL FORAGE
IN AGROFORESTRY SYSTEMS**

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ABSTRACT

Cover crops are planted traditionally in three major plantation crops such as rubber, oil palm and coconut to suppress weeds, control soil erosion and to add nitrogen. Sown forages have a role in many systems to enhance production efficiency and contribute to other function such as erosion control, soil improvement, restoration of degraded land, and improving biodiversity. The major problem encountered with many sown forage species in plantation crops is lack of long-term persistence due to limitation of light transmission. Livestock are a crucial component of livelihoods and food security of nearly 1 billion people in the developing world, contributing 40% of the global value of agriculture output. Livestock contribute 15% of total food energy, 25% of dietary protein, and some micronutrient that are not available from plant. On the other hand agriculture, including meat and milk production, produces three main greenhouse gas GHG: carbon dioxide CO₂, methane CH₄ and nitrous oxide N₂O. Livestock systems are estimated to contribute about 50% of all agriculture sector GHG emissions, contributing up to 9% of all anthropogenic CO₂ emissions, 37-52% of CH₄, and 65-84% of N₂O. This article describes the environment in which the three major plantation types occur, discusses the adaptation and value of the most frequently encountered naturally occurring and sown forage species, and reviews the potential for making best use of existing forage resources in plantation crops. We also discussed the potential and the associated benefit soft tropical forage-based systems to contribute to reducing greenhouse gas emissions and to enhancing the eco-efficiency of farming in the tropics.

Keywords: *agroforestry system, forage, multifunction, tropical.*

NUTRIOPT SPLIT-FEEDING, OPTIMAL SUPPLY OF NUTRIENTS TO ENHANCE THE PERSISTENCY

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ABSTRACT

The protein, calcium and energy requirements of laying hens do not remain constant, but vary during the day depending on the hen's physiological requirements for the various stages of egg formation. The current feeding methods for laying hens based one diet with constant levels of nutrients may not result in optimal utilization of the nutrients (Chah, 1972, Leeson and Summers, 1997). When birds are offered diets that allow self-selection of nutrients, an increase in protein and energy intake in the morning has been observed, around the peak of egg production. Calcium intake is higher later in the day. In a study of Chah (1972) where hens were offered diets that allow self-selection of nutrients, the total daily protein, energy and calcium intake were respectively 11%, 8% and 26% lower, compared to hens fed a normal diet. This suggests that the hen is using the energy, protein calcium and phosphorous more efficiently by consuming these nutrients at periods of the day that the requirements are highest. Therefore the current practice of providing hens with only one diet with a uniform composition might not be the ideal approach for optimal nutrient utilization. Trouw Nutrition has developed the NutriOpt split-feeding system based on the concept of feeding hens two diets with different nutrients levels during the morning and the afternoon to meet the different requirements through the day. The success of the split-feeding system relates to the fact that current layer hen lines lay the majority of eggs during the morning (Etches, 1986, Leeson and Summers, 1978, Larbier and Leclercq, 1992). The interval between two successive ovipositions in a cycle is about 24 – 26 hours (Keshavarz, 1998). After each oviposition the subsequent ovulation occurs about 30 minutes later. Within the first four hours the egg white is formed. Thereafter the egg moves to the shell gland, where the shell is deposited around the albumen within approximately the next 20 hours (Larbier and Leclercq, 1992). Shell formation mainly takes place during the evening and night. As a result of this, hens will have higher protein and energy requirements during the morning and a higher calcium requirement during the evening and night. Phosphorous is also more required during the morning as it is mainly needed to reabsorb the calcium for the medullar bone used during the night to form the eggshell. Trouw Nutrition R&D has been working since 2005 to develop a novel feeding program for laying hens according to nutrient requirements for egg formation. Eight trials have been performed from 2005 to 2012. The main objective of the trials was to determine the nutrient requirements of energy, protein, calcium and phosphorous during the morning and the afternoon when two diets were fed. All trials contained a single feed control to corroborate the benefit of the split-feeding system against the current feeding system applied in the egg industry. As result of this project a split-feeding program has been designed using two dietsto meet the different requirements resulting from the egg formation phase, first, morning diet (focused on the requirements of energy,protein and phosphorous) and second, afternoon feed(focused on the requirements of calcium). Daily feed amounts are covered by 40% with the morning diet (from the start of the light period) and by 60% with the afternoon diet, which fits with the normal feed intake behaviour of the laying hens and has been experimentally confirmed (Keshavarz, 1998). The results obtained from the trials support that the split-feeding system: Reduces nutrients intake compared to a single feed, and as consequence (Reduces price of the equivalent diet and Reduces excretion of nutrients), Improves eggshell quality and laying persistency of older flocks, and All other performance parameters are at least comparable as those obtained with a single diet. The NutriOpt split-

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feeding is a new system for feeding laying hens. In addition to apply current knowledge in nutrition, it incorporates the advantage of adjusting the nutrient consumption to the different requirements throughout the day in order to meet the needs of hens in the egg formation process. With this system, the laying hen is closer to the voluntary and physiological feeding behaviour. This adaptation to the physiological requirements of the laying hen during the day allows the NutriOpt split-feeding to be a more efficient feeding programme, brings a more profitable and sustainable egg production with a lower production cost. NutriOpt split-feeding provides the following benefits: improve the quality of eggshell and increase the number of saleable eggs, more profitable egg production by reduction of production cost and improve the performance and a sustainable production.

**INFLUENCE OF SHRIMP WASTE, KATUK LEAF
(*Sauropus androgynus L. Merr.*), BROMELAIN AND GARLIC POWDER
ADDITION ON PERFORMANCE AND EGG QUALITY OF QUAIL**

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ABSTRACT

This experiment aimed to study effect of addition of shrimp waste, katuk leaf, garlic powder, and bromelain on performance, egg weight quality, and physical composition of the egg. This experiment used 160 quail pullet reared 10 weeks and divided into five treatments and four replicates. The experimental diets were P0 (control), P1 = P0+ 31.1 ppm bromelain, P2 = P0+ 0.45% waste shrimp powder, P3 = P0+10% katuk leaf powder, and P4 = P0+1% garlic powder. This study used a completely randomized design. The results showed that egg weight and yolk color score were significantly different ($P<0.05$) among the treatments. Egg production, feed consumption, feed conversion, egg white percentage, egg yolk percentage, egg shell percentage, index of eggs and Haugh Unit were not significantly different. All eggs were classified into AA quality. All treatments produced higher ($P<0.05$) egg weight compared to control. Egg yolk color score of katuk leaf powder group was significantly higher ($P<0.05$) than other groups. It is concluded that the addition of bromelain, shrimp waste powder, katuk leaf powder, and garlic powder did not affect performance. But those had significant different effect on egg quality.

Keywords: *Quail egg, Egg quality, Shrimp waste, Katuk leaf, Garlic, Bromelain*

INTRODUCTION

Quail have a high potential to produce eggs. Quail egg farms are able to produce proteins that help meet the needs of the people of Indonesia. Quail small body size gives the advantage because the land requirement is not too wide for maintaining them in large numbers. Another advantage is their very fast grow and mature, ie at the age of 35-42 days have started laying eggs. Their eggs production capable of reaching 200-300 eggs/year with a weight of 10 grams/egg. Based on data from the Directorate General of Livestock (2012) quail egg production in Indonesia in 2011 reached 16.926 million tons.

The most important factor in the maintenance of quail is feed. Quail nutritional needs should be met within the feed. Needs amount of feed for quail is usually more than 10% of their body weight. Shrimp waste is a waste of frozen shrimp processing industry that have potential and relatively high nutritional value. Garlic has a wide range of active substances in it. Katuk leaves contain high nutrients and bromelain is one of the sulfhydryl protease enzyme capable of hydrolyzing the bond polypeptides into amino acids.

Enzyme bromelain is a proteolytic enzyme such as renin (rennet), papain and fisin which has the properties of protein hydrolysis. Enzyme bromelain from pineapple weevil is one of the alternatives in order to utilize waste sehingga pineapple can provide added value for pineapple in addition to reducing the pollution problems of waste on the environment (Sebayang, 2006).

Shrimp heads flour can be used as animal feed ingredients. advantage of shrimp heads flour is a waste product of fisheries that have a fairly continuous availability, the price is quite stable and nutritional able to compete with conventional feed ingredients (Wanasuria, 1990). Berda- sarkan Syukron research (2006) the best Taraf administration shrimp heads in the ration of broiler chickens is as much as 6% .According Mawaddah study (2011), granting 10% katuk leaf meal in the diet of quail produce quality meat and eggs are better than the product quail. The treated extract katuk leaf meal at the same level. Garlic is thought to be able to optimize the metabolic functions of food ingredients so as to improve the efficiency of feed utilization. Each 2 kg of fresh ingredients, garlic produces 600 g of dry matter (Wiryanawan *et al.*, 2005).

Katuk leaves, waste shrimp, garlic, and bromelain contains good nutrition and still rarely used as animal feed. In addition, the availability can be obtained throughout the year. Some feed materials can be added to the feed of quail to supplement the nutritional needs of quail. The content of nutrients in each feedstuff is expected to give a good effect on the performance and quality of quail eggs. Therefore, it is necessary to study the effect of addition, bromelain, flour shrimp waste, katuk leaf flour and garlic powder to the quality of quail eggs. This study aimed to evaluate the effect of adding shrimp waste flour, flour leaves katuk (*Sauropus androgynus* L. Merr.), Garlic powder, and bromelain on the performance and quality of eggs (egg shell weight percentage, the percentage weight of egg yolk, egg white weight percentage , thick egg shell and yolk color) quail.

MATERIALS AND METHOD

This study used 160 quails grower phase that are ready for production and placed in 20 pieces of battery cages. Each cage contains eight quails. Each cage is equipped with a place to eat and drink. Each plot enclosure is equipped with double rations and the drinking water. The quail is divided into five treatments and four replications and maintained for 10 weeks.

Experimental diets consisted of basal ration supplemented with bromelain, shrimp waste meal, katuk leaf meal, and garlic powder in accordance with treatment. Drinking water was given every day during the study. Experimental diets were given after 2 weeks of maintenance. The composition of experimental diets is shown in Table 1. The nutrient content of the ration experiments are presented in Table 2.

The experimental design used in this research is completely randomized design (CRD) with 5 treatments and 4 replicates. The treatments provided are: P0: Rations control, P1: P0 + bromelain 0.625 mg/head /day, P2: P0 + 0.45% shrimp waste powder, P3: P0 + 10% katuk leaf meal, P4: P0 + garlic powder 1%.

Data were analyzed by analysis of variance (ANOVA). If significantly different between treatments was tested further by Duncan's multiple range test (Mattjik and Sumertajaya, 2006).

Parameters measured were as followed: production of eggs (%), feed conversion, feed intake, consumption of metabolizable energy (kcal/head/day), consumption of protein (g/head/day), egg weight (g/egg), proportion of yolk (%), egg white proportion (%) proportion eggshell (%), egg yolk color and Haugh Units (HU).

Table 1. Composition of experimental diets

Ingredients	P0	P 1	P2	P3	P4
	------(%)-----				
Yellow corn	46	46	46	46	46
Rice bran	9	9	9	9	9
Soybean meal	27	27	27	27	27
Fish meal	8	8	8	8	8
CPO	3	3	3	3	3
DCP	0,8	0,8	0,8	0,8	0,8
NaCl	0,2	0,2	0,2	0,2	0,2
CaCO ₃	5	5	5	5	5
Premix	0,4	0,4	0,4	0,4	0,4
DL-Methionine	0,6	0,6	0,6	0,6	0,6
Total	100	100	100	100	100
<i>Bromelain</i> (mg/head/day)	-	0,625	-	-	-
Waste shrimp meal (%)	-	-	0,45	-	-
Katuk leaf meal (%)	-	-	-	10	-
Garlic powder (%)	-	-	-	-	1

Table 2. Nutrients content of experimental diets based on calculation

Nutrient	P0	P1	P2	P3	P4
ME (kcal/kg)	2837,50	2825	2855,34	3017,5	2878,46
Crude protein (%)	22,44	22,60	22,58	25,74	22,61
Ether extract (%)	5,49	5,01	5,53	6,49	5,50
Crude fibre (%)	3,12	4,00	3,22	3,12	3,14
Ca (%)	2,65	2,78	2,72	2,68	2,65
P (%)	0,46	0,48	0,47	0,49	0,47

Prosedur

Shrimp waste powder. Shrimp waste powder wer obtained from the Institute of Fisheries and Freshwater Aquaculture, Bogor.

Katuk leaves meal. Leaves and stems were first separated. Katuk leaves then dried in the sun. Dried katuk leaves was then processed into katuk leaves meal.

Garlic powder. First, garlic was dried in the sun, then was ground into a powder.

Maintenance of Quail and Treatment Application

Quail used in the study were randomly placed into battery cages by the treatment given. Treatments of diet were given at the second week of maintenance. In the beginning quail were fed the basal ration for 2 weeks. Maintenance of quail lasted for 10 weeks and the first 2 weeks serve as a control.

One hundred sixty quails were placed in a cage. Each treatment consisted of 4 replicates with 8 quails for each experimental unit.

RESULTS AND DISCUSSION

Feed Consumption, Eggs Production and Ration Conversion

Feed intake did not show significantly different results. The value of feed intake in the control treatment, administration of bromelain, shrimp waste powder, flour katuk leaves, and garlic powder are respectively 22.76 ± 2.12 ; 22.22 ± 1.59 ; 22.84 ± 1.89 ; 24 ± 3.22 ; and 23.16 ± 6.07 g / head / day. Factors affecting feed intake is the large body of livestock, livestock activity, ambient temperature, quality and quantity of ration (NRC, 1994).

The production value of eggs in each treatment showed significantly different results. Egg production in the control treatment, administration of bromelain, flour shrimp waste, katuk leaf, and garlic respectively in the amount of $32.25 \pm 3.86\%$; $34.61 \pm 6.88\%$; $36.16 \pm 2.41\%$; $40.04 \pm 8.91\%$; and $39.50 \pm 5.90\%$. According Listyowati and Roospitasari (2004) Production of quail eggs is influenced by genetic and environmental factors such as diet, cage, temperature, environment, disease, and stress. Factors affecting feed intake is the large body of livestock, livestock activity, ambient temperature, quality and quantity of ration (NRC, 1994).

Feed conversion showed no significantly different results. Feed conversion in the control treatment, administration of bromelain, shrimp waste powder, flour katuk leaves, and garlic powder respectively is 9.13 ± 1.11 ; 7.43 ± 2.45 ; 7.57 ± 1.51 ; 7.37 ± 1.65 ; and 7.20 ± 3.30 . This suggests that the efficiency of feed utilization on all treatments are the same (average 7.74). Widjastuti and Kartasudjana (2006) states that the balance between feed consumed by the production of eggs produced in each treatment causes no different feed conversion.

Egg Production, Energy Consumsed, Protein Consumsed, and Percentage Weight Components of Quail Eggs

The average weight of quail eggs in each treatment showed significantly different results. Giving bromelain produces the greatest egg weight from other treatments that is equal to $9.09 \pm 0,31$ g. Bromelain has a high protein content. Quail egg weight is not only influenced by the quantity of feed consumed but also by the quality of feed, especially protein content (Mozin, 2006). Protein deficiency will result in a large decrease in the number of eggs and egg albumen (Amrullah, 2003).

Egg weight on the addition garlic powder and shrimp waste powder showed not significantly different results, and each has an eggs weight of $8.60 \pm 0,37$ g and $8.47 \pm 0,53$ g. On the addition of bromelain treatment showed a highest egg weight than the other treatment that was equal to 9.09 ± 0.31 g but the control treatment had the smallest egg weight of $7.84 \pm 0,82$ g. Results of the study had a lower weight value than that of Kul and Seker (2004) who obtained results of egg weight (g) of $11.28 \pm 0,06$ g.

Consumption of protein and metabolizable energy used to meet the maintenance, growth and egg production (Widjastuti and Kartasudjana, 2006). The magnitude of the weight of the eggs produced by the addition of bromelain treatment can be caused due to consumption of protein and metabolizable energy used to meet maintenance and growth are fulfilled, so that the remainder is used to produce large eggs. Requirement for maintenance and growth in the control treatment that has not been fulfilled resulting in the consumption of protein and metabolizable energy is not widely used for the production of eggs, so the weight of eggs produced is low.

The percentage of egg whites on all treatments showed no significantly difference, i.e. ranging 54.06 – 55.59%. Hazim et al. (2011) measures the egg whites percentage of

53.10%. Kul and Seker (2004) obtained the higher egg whites percentage of 59.83. Likewise, the percentage of egg yolk and eggshell were not significantly different. Kul and Seker (2004) reported that the percentage of yolk $32.71 \pm 0.12\%$ and the percentage of eggshell $7.47 \pm 0.04\%$. According to Song et al (2000) quail egg yolk has a percentage of 29.42 to 33.38%, from 58.88 to 63.52% egg white, and eggshell 6.61 to 7.99%.

Table 3. Feed consumption, egg production, feed conversion, nutrient intake, egg weight and percentage weight components of quail eggs given experimental diets

Parameters	Treatments				
	Control	<i>Bromelain</i>	Shrimp waste	Katuk leave	Garlic
Feed intake (g/head/day)	22,76±2,12	22,22±1,59	22,84±1,89	24±3,22	23,16±6,07
Egg production (%)	32,25±3,86	34,61±6,88	36,16±2,41	40,04±8,91	39,50±5,90
Feed conversion	9,13±1,11	7,43±2,45	7,57±1,51	7,37±1,65	7,20±3,30
Energi intake (kcal/head/day)	64,86±4,94	62,77±4,48	65,23±5,39	72,42±9,72	66,65±17,49
Protein intake (g/head/day)	5,13±0,37	5,02±0,36	5,16±0,43	6,18±0,83	5,23±1,37
Egg weight (g)	7,84 ^c ±0,82	9,09 ^a ±0,31	8,47 ^{ab} ±0,53	8,15 ^b ±0,84	8,60 ^{ab} ±0,37
Albumin (%)	54,06±0,44	55,59±0,84	55,59±0,88	55,19±0,85	54,88±0,63
Yolk (%)	30,02±0,62	29,56±0,76	30,45±0,70	30,63±0,54	30,5±0,97
Shell (%)	9,79±0,10	9,81±0,39	9,47±0,21	9,75±0,21	9,78±0,08

Remark: different superscript within the same row indicate significantly different (P <0.05)

Table 4. Quail egg quality given experimental diets

Parameters	Treatments				
	Control	<i>Bromelain</i>	Shrimp waste	Katuk leave	Garlic
Egg index	78.81±4.36	0,80±0,71	0,81±1,19	0,80±1,11	0,81±1,11
Yolk colour score	4,13 ^b ±0,91	4,10 ^b ±0,20	4,17 ^b ±0,21	6,10 ^a ±0,43	4,24 ^b ±0,27
Shell thickness (mm)	0,168 ^a ±0,01	0,167 ^a ±0,01	0,155 ^b ±0,01	0,158 ^{ab} ±0,01	0,165 ^{ab} ±0,01
Haugh Unit	92.64±1.01	91,67±0,49	91,95±1,58	91,24±1,02	91,67±1,04

Remark: different superscript within the same row indicate significantly different (P <0.05).

Egg Quality

The quality of the eggs is a collection of factors that affect the valuation and tastes of consumers on the quality of the eggs. Consumers are always looking for fresh eggs, with standard weight, good eggshell quality, yolk color attractive (yellow) and a relatively thick egg white (Yuwanta, 2010). In this study, the egg index was not significantly different between treatments. This shows the shape of eggs in each treatment is more rounded than the results Kul and Seker (2004), ie with an index of 0.75 ± 0.22 eggs.

Scores yolk color in this study was significantly different. In the treatment of leaf powder katuk have egg yolk color score highest than the others, namely 6.10 ± 0.43 . In the administration of bromelain treatment, waste flour shrimp, garlic, and controls were not significantly different. Hulshoff *et al.* (1997) reported that among the vegetables and fruits were studied in Indonesia, the highest katuk leaf contains carotene. This shows that the carotene pigment found in leaves katuk have a role in improving the yolk color scores.

Eggshell thickness was significantly different in all treatments. Control treatment and administration of bromelain has the thickest shell, each of which is 0.168 mm and 0.167 mm. Treatment by administering powdered shrimp waste has the most thin shell that is 0.155 ± 0.01 mm. Eggshell thickness in treatment provision garlic powder and leaves katuk respectively 0.165 mm and 0.158 mm. Kul and Seker (2004) obtaining eggshell thickness values higher than this study is 0.231 mm.

Haugh unit in this study showed no significantly different results for all treatments and eggs belong to the quality of the AA indicated by HU value >91.24 , i.e. above 72 (USDA, 2000). Haugh unit quail eggs on research Kul and Seker (2004) was 85.73 ± 0.15 was lower than HU in this study.

CONCLUSION

All treatments can provide a high quail egg weight compared with the control. The percentage weight of the composition of quail eggs are not affected by the provision of treatment. Giving katuk leaves can increase the value of yolk color scores and maintain a quail egg production. The addition of bromelain, flour shrimp waste, katuk leaf, and garlic does not affect performance and can maintain the quality of quail eggs.

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THE EFFECT OF HERB MEDICINE SUPPLEMENTATION ON BLOOD PROFILES OF LAYING QUAIL

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ABSTRACT

Herb medicine was called “Jamu Ternak” has been popular in the people to stimulate immunity and treat diseases of poultry. The study was aimed to evaluated the effect of herb medicine supplementation through drinking water on blood profile of laying quail. This experiment used 400 heads of quail females, aged 36 weeks. Quails were reared until 8 weeks. Quail ration containing 18% crude protein and 2950 kkal/kg metabolizable energy. Herb medicine used as much as 9 kinds herb (*Alpinia galanga*, *Curcuma domestica*, *Kaemferia galanga*, *Curcuma xanthorrhiza*, *Zingiber officinale* Rosc, *Allum sativum* Linn, *Cinnamomun zeylanicum*, molasses and EM4). All herbs mashed, filtered and fermented for 5 days. Feed and water were given *ad libitum*. The experiment using a completely randomized design 5 treatments and 2 replications. Data were analyzed descriptively. The treatments consisted of herb medicine by drinking water, i.e drinking water with 0% herb medicine supplementation (control), drinking water was given herbs medicine 30 ml / liter of water (P1), drinking water was given herb medicine 60 ml / liter of water (P2), drinking water was given herb medicine 90 ml / liter of water (P3), and drinking water was given herb medicine 90 ml / liter of water (P4). The Treatment of P1, P2, and P3 was given herb medicine twice a week but P4 treatment only one times a week. The variables measured were hematological blood of quali ie haemoglobin, PCV (hematocrite), erythrocyte, leucocyte, heterophyl and Lymphocyte. The results showed that haemoglobin and hematocrit (PCV) in 60-90 ml/l of water and the leukosyte in 90 ml/l of water was given twice times a week of herb medicine supplementation higher than other treatments. The value of erythrocyte no difference among the treatments. Beside that the doses of herb medicine supplementation at 90 ml/l of water of with was given one times a week had H/L lowest than other treatments.

Keywords: *Herb medicine (jamu ternak), Blood profile, Quail, Hematological*

INTRODUCTION

Herbs medicineis a lot of usedasa substitute for antibioticsforrearing poultrysuch aschicken, duckandquail. Herbs supplementationformulascreated by the farmer withvariety of herb dependingon thepurpose of supplementationandexperienceby farmer. In general, herbal plants (rhizomes, leaves, stems, roots, flowers and fruits) have active compounds such as alkaloids, flavonoids, tripenoid, glycosides and essential oils. The kind and amount of herbs were used by the researcher generallyeasy to get on the field such as *Morinda citrifolia* in quail (Wardiny *et al.*, 2013), *Aloe vera* in laying chicken (Bintang *et al.* 2005), *Allium sativum* as antioxidant and it used in broiler chicken (Retnani *et al.*, 2010 and Mide, 2008), *Andrographis paniculata* (Suci *et al.*, 2012), *Curcuma domestica* in broiler chicken (Bintang dan Nataamijaya, 2005), papaya leaf meal in chicken (Wijastuti, 2009, Suci *et al.*, 2013). The resultsof herbalsupplementationin

poultry varies greatly. Many factors influence the results such as the amount of active compounds in herbs, infection of *Ascaridia galli* (Deka dan Borah ,2008) or doses used in this experiment.

Herbs medicine can be given by feed or drinking water with various kinds of processing techniques such as meal and extract of herbs. The extract of herbs can do with boiled in the water or extract with ethanol.

MATERIAL AND METHODS

Four hundred quails reared on the farm were given diet containing 18% crude protein and 2600 kcal/kg metabolizable energy (Table 1). The diet did not contain a feed additive such as antibiotic. Diet consumption was \pm 19 g per day per head. Quails placed on the colony battery cages as many as 10 pieces, each colony battery contains 40 quails.

Table 1. Ingredient composition of laying quail diet used this experiment

	Total (%)
Yellow corn	53.00
Rice bran	4.00
Soybean meal	25.00
Fish meal	7.50
Crude Palm Oil	3.00
CaCO ₃	6.00
Dicalcium Phosphat	0.60
Salt	0.30
Premix	0.50
DL-Methionine	0.10
Total	100
Nutrient content :	
Dry matter (%)	88.57
Crude protein (%)	18.90
Crude fiber (%)	3.12
Ether extract (%)	5.24
NFE (%)	50.68
Ca (%)	3.2
P (%)	0.68
Metabolizable energy (kcal/kg)	2950

Laboratory Analysis Results Nutrition and Feed Technology, Faculty of Animal Husbandry, Bogor Agricultural University (2012).

Each cage was given a feed and drinking water *ad libitum*. The quails were reared 8 weeks. Herb supplementation was given through drinking water is done for 2 times a week. The quails were given herbs medicine supplementation as much as 30, 60 and 90 ml/l and had two a control such as without herbs medicine and it was given 90 ml/l for once a week. All herbs cleaned, smoothed, filtered and taken to the juice. Furthermore, the juice was added 300 ml molasses and 300 ml EM4. After mixture very well added the water become 30 l. Herb medicine put into a sealed

container and stirred and sealed, fermented for 5 days. Every day the herbs stirred 5 minutes and container closed again.

Table 2. Composition of herbs

No	Herbs	Composition
1.	<i>Kaempferia galanga</i> L	750 gram
2.	<i>Ginger officinale</i> rosc	375 gram
3.	<i>Curcuma domestica</i>	375 gram
4.	<i>Langkuas galanga stunz</i>	375 gram
5.	<i>Allium sativum</i> .l	750 gram
6.	<i>Curcuma xanthorrhiza</i> roxb	375 gram
8.	<i>Cinnamomun burmanii</i> b	187.5 gram
9.	Molases	300 ml
10	Probiotik/EM4	300 ml
11	Water	30 l

Haematological

Blood was collected from individual quail. Each repetition was taken 3 quails. Blood was taken by artery in the wing. Sterile vials with EDTA were used as anticoagulant for collection of blood. Plasma serum analyzed haematology profile.

Design Experiment

The experiment used the completed design with two repetitions. Analyzed of the data used descriptive analysis.

RESULT AND DISCUSSION

Active Substantive of Herb Medicine Supplementation

Phyto chemical analysis results of herb medicine supplementation in this experiment in Table 3. The result showed that herbs medicine contains saponins, flavonoids and tannins. Tannins in the body can reduce cholesterol because working tannins bind fatty acids in the digestive tract, while saponins inhibit cholesterol by saponification process. Tannin, saponin and flavonoid can reduce absorption of nutrient in the diet if it were not optimal in the body.

The results of the haematology of the quail showed that all treatments did not influence of all variables. Hemoglobine, erythrocyte (PCV), leucocyte and differentiation from leucocyte had same values. The blood profile of quail aged 28-42 days such as erythrocytes, hemoglobin and hematocrite (PCV) did not significantly affected by administration of turmeric powder in diet doses 0.1-1% (Napirah *et al.* 2013). Noni leaf extract in drinking water at a quail starter period did not affect blood profile but tend to increased erythrocytes and leucocyte in the provision of 15% (Wardiny *et al.* 2012).

Result this experiment when compared with the results Napirah *et al.* (2013) showed that hemoglobin content of this experiment range from 6-8.8 g/dl was lower. Lowered content of hemoglobin associated with digestibility and absorption of nutrients, especially protein, amino acids and minerals Fe. The quail were given the herbs medicine did not increased hemoglobine levels so that the herbs medicine was given did not increased the absorption of nutrients. Tuleun *et al.* (2013) hemoglobin content of blood is also not affected by dietary protein of 17-21%.

Table 3 Results of phytochemical analysis of herbs medicine

Active substances	Result ¹
Alkaloid	-
Flavonoid	+++
Phenol Hidrokuinon	+
Steroid	-
Triterpenoid	++
Tannin	+
Saponin	+

Laboratory of Biochemistry, Faculty of Mathematics and Natural Science, Bogor Agricultural University (2012).

The content of erythrocyte this research in egg-laying period range 7.6-7.7 million/mm³) was higher than erythrocytes in quail starter period (1.79 million/mm³) (Wardiny *et al.*, 2012). Moreover there was not effect of herbs on the content of quail erythrocytes. This is similar to results obtained by Wardiny *et al.* (2012) and Napirah *et al.* (2013), the content of quail erythrocytes were not influenced by administration of noni leaf extract in drinking water(5-15%) and turmeric powder in feed (0.1-1%).

The blood PCV (hematocrite) content of laying quail showed result range 35.5% -37.7% was not different if compared with quail given turmeric as much as 1% ie 39.50% (Napira *et al.*, 2013). According Tuleun *et al.* (2013) the content of blood PCV laying quail period did not significantly affected by the protein content of the ration, but there were a tendency that dietary protein 20-21% higher than the protein content of the ration 17%, 18% and 19%. The diet protein of quail this research by 18%. According Tuleun *et al.* (2013) blood PVC around 5.52±31.5%

Blood leucocytes of results this research range 8.2-10.9 thousand % / mm³. Herbs medicine supplementation doses 30 - 90 ml via drinking water did not increased leucocytes. Leucocytes blood increases when there is an indication of infection in the body. Levels of blood leucocytes from quail were given the herbs medicine did not different if was compared with control (without herbs medicine) so that it can be stated there was no noticeable increase of antibodies in the body. Napirah *et al.* (2013) states granting turmeric powder in the diet at the age of 42 days quail was not affected by doses of turmeric powder 0.1% -1%.

Blood leucocyte is play a role in keeping the immune system from fighting infections. When the infection in quail was reduced so the neutrophil was reduce also but monocyte was increase. Supplementation of herb medicine was not affect the content of lymphocytes, monocytes, basophils eosiofil and heterophile on laying quail. Results of the research administering drinking water showed lymphocyte content was far below the results of Napirah *et al.* (2013). Leucocytes, neutrophils, lymphocytes and monocytes with the addition of 0.1-1.0% of turmeric powder in ration were affected by age and doses (Napirah *et al.*, 2013). Quail age of 42 days content of lymphocytes 62.50% and when quail was given 1% turmeric showed result 74.25% (Napirah *et al.* 2013). Monocyte levels of result this experiment was not difference from the research of Napirah *et al.*(2013).

Table 3. Haematology analysis of quail blood profile were given supplementation of herbs medicine

Variables	Control 1 (Without herbs medicine)	Control 2 (given herbs medivine, once a week	Given herbs medicine, twice a week		
			30 ml/l	60 ml/l	90 ml/l
Haemoglobine (g/%)	6.7 ±1.8	7.4±1.8	6.0±0.7	7.4±0.8	8.8±1.0
Packed Cell Volume (%)	35.5±4.7	37.3±4.5	35.5±4.8	37.7±3.6	36.2±4.6
Erythrosit (x10 ⁶)/mm ³	7.8±0.7	7.5±0.6	7.7±0.7	7.6±0.8	7.6±0.8
Leucocyte (x10 ³) %/mm ³	9.0±2.1	11.6±2.8	8.6±2.6	8.2±1.1	10.9±2.7
Differentiation of Leucocyte					
Lymfosit (%)	34.3±6.0	40.7±8.0	36.7±9.0	44.7±9.9	32.7±6.0
Monosit (%)	6.0± 2.6	3.0±2.0	3.2±1.8	4.2±1.8	4.0±2.0
Basofil (%)	3.0±1.0	4.3±1.5	4.0±1.3	3.2±2.4	2.8±1.8
Eosinofil (%)	5.3±0.6	3.3±2.5	6.2±2.5	4.5±1.4	4.3±2.3
Heterofil (%)	51.7±7.6	48.7±6.8	46.7±10.0	44.3±10.4	56.2±5.6
Heterofil/lymfosit	1.6±0.5	1.2±0.4	1.4±0.6	1.1±0.4	1.8±0.4

Heterophil limfocyte ratio indicated higher than normal range that is 0.34 to 0.43. H/L ratio this experiment indicated the quail was stress in the all treatment. Quails used in the Research were stress causes a change of commercial feed into rations that do not use antibiotics. Doses of herbs used have not been able to prevent the occurrence of stress.

CONCLUSION

Herbs medicine supplementation in laying quail through drinking water (30 to 90 ml L-1) was not effect in haematological profile.

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SUPPLEMENTATION OF *Azolla pinnata* TO SORGHUM BASE DIET ON EGG QUALITY OF JAPANESE QUAILS

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ABSTRACT

Azolla pinnata is a water plant potential for animal feed. *Azolla pinnata* contain beta carotene that useful to enhance the color of egg yolk and as an antioxidant. This experiment was conducted to study the effects of *Azolla pinnata* supplementation in sorghum base diet on egg quality of Japanese quails. The experiment used 30 Japanese quails (44 day of age) which were reared for 2 weeks. This experiment used 5 dietary treatments and 6 replications. The dietary treatments were: T0 = corn basal diet as control diet, T1 = sorghum basal diet without *Azolla pinnata* supplementation, T2= T1 + 1% *Azolla pinnata*, T3 = T1 + 2% *Azolla pinnata* and T4 = T1 + 3% *Azolla pinnata*. Water and feed were provided *ad libitum*. At the end of feeding trial, all egg from each treatment was collected for egg quality measurement. There were no difference on egg weight, shell weight, shell thickness, Haugh Unit due to *Azolla pinnata* supplementation. However, egg yolk color of *Azolla pinnata* treatment increased as compared to control diet (T1) and corn base diet (T0). The MDA (malondialdehyde) level of birds fed *Azolla pinnata* decreased as compared to T0 and T1. In conclusion, *Azolla pinnata* supplementation improved egg yolk color and antioxidant status of Japanese quails.

Keywords: *Azolla pinnata*, egg quality, Japanese quails, MDA

INTRODUCTION

Corn is a mayor energy sources for poultry feed, it is contain 9% protein and 3350 kkal/kg of metabolizable energy (NRC, 1994). Yellow corn especially is used for laying hen diet not only for energy source but also for carotenoid source. Today, corn utilization not only for food and feed but also for fuel, so the price of corn increase every year. There fore we need look for another energy resources for poultry feed. Sorghum is promissing energy alternative for replace corn in poultry feed. Sorghum contain 11% protein and 3288 kkal/kg (NRC, 1994). The energy contain of sorghum near to energy of corn, but there is a litle problem if we use sorghum for laying hen diet. Sorghum has no carotenoid as much as corn, so we have to supplement with other carotenoid source. One of carotenoid source is *Azolla pinnata*.

Azolla pinnata is an aquatic plant abudantly in paddy fields and ponds in Indonesia. It contains 140 - 300 g/kg crude protein (Subudhi and Singh, 1978; Sreemannaryana, *et al.* 1993). Khatun *et al.* (1999) reported that *Azolla pinnata* can be used as feed ingredient for laying hen. *Azolla pinnata* contain 1188 mg/kg beta carotene that useful to enhance the color of egg yolk and as an antioxidant. Mithraja *et al.* (2011) reported that *Azollapinnata* contains various antioxidant such as tannin, phenolic content and flavonoids. *Azolla pinnata* can absorbed heavy metal such as lead and zinc from polluted water (Jain, et al. 1990, 1992).

The purpose of this experiment was to study the effect of *Azollapinnata* supplementation to sorghum base diet on egg quality of Japanese quails.

MATERIALS AND METHODS

Bird and Housing

This experimental was conducted at Laboratory of Poultry Nutrition, Faculty of Animal Science, Bogor Agricultural University. The experiment used 30 Japanese quails (44 day of age) which were reared for 2 weeks. This experiment used 5 dietary treatments (6 birrds/treatment). The quails were reared on coloni cage in open side house with standard management conditions throughout the experiment period. Feed and water were provided *ad libitum*.

Experimental diet and chemical analysis

Experimental diet was formulated to met Japanese quails requirement according to Leeson and Summer (2005) recommendation. Composition and nutrients of experimental diet was shown in Table.1and Table2.

Table.1. Composition of experimental diet (% as fed)

Ingredient	T0	T1	T2	T3	T4
Yellow corn	40	-	-	-	-
Sorghum	-	40	40	40	40
Rice bran	4	4	4	4	4
Soybean meal	35	35	35	35	35
Fish meal	10	10	10	10	10
CPO	5.5	5.5	5.5	5.5	5.5
NaCl	0.5	0.5	0.5	0.5	0.5
DCP	0.1	0.1	0.1	0.1	0.1
CaCO ₃	3.4	3.4	3.4	3.4	3.4
Premix	0.5	0.5	0.5	0.5	0.5
Total	100	100	100	100	100
<i>Azolla pinnata</i>	-	-	1	2	3

T0= corn base diet ; T1=sorghum base diet; T2,T3,T4 = sorghum base diet supplemented with 1%,2% or 3% *Azolla pinnata*.

Table.2. Nutrients content of experimental diet (% as fed)

Nutrients	T0	T1	T2	T3	T4
Dry matter	88.78	88.18	89.07	89.95	90.84
Ash	13.91	14.08	14.15	14.22	14.29
Crude protein	22.71	23.42	23.63	23.84	24.04
Crude fiber	5.07	4.57	4.60	4.63	4.66
Crude fat	1.87	2.16	2.20	2.24	2.27
Nitrogen free extract (NFE)	44.26	43.77	43.83	43.94	44.11
ME (kcal/kg)	3744	3606	3606	3606	3606

Fresh *Azolla* was harvested from pail in our laboratory and then sun-dried. It was ground (4 mm screen) and store in air-tight plastic bag until used in the diet. The dietary treatments were : T0 = corn basal diet as control diet, T1 = sorghum basal

diet without *Azolla pinnata* supplementation T2= T1 + 1% *Azolla pinnata*, T3 = T1 + 2% *Azolla pinnata* dan T4 = T1 + 3% *Azolla pinnata*. MDA from egg was measured according to Capeyron (2002).

Data collection

After 2 weeks feeding trial, all egg from each treatment were collected for measuring egg quality. Feed intake was measured weekly, egg was collected everyday for calculated the hen day production and egg mass.

Data analysis

This experiment actualy was preliminary study, due to limited animal unit we do not used statistical analysis but we used discriptived analysis.

RESULTS AND DISCUSSION

Azolla pinnata in our experiment contain 20.81% crude protein, 3.11% crude fiber and 1188 mg/kg of beta carotene. This results indicated that *Azolla pinnata* can be used as protein and carotenoid source in laying quails diet. The results of supplementation *Azolla pinnata* on laying performance are showed in Table 3.

Table 3. Supplementation of *Azolla pinnata* on laying quail performance

Parameter	T0	T1	T2	T3	T4
Feed intake(g/day)	22.09± 1.01	23.35±1.27	21.32±1.71	22.12±2.98	19.87±2.09
Hen day (%)	53.33±13.1	71.67±20.9	63.33±13.1	51.67±20.0	50.00±13.6
Egg mass(g/day)	5.03±1.24	6.48±1.89	5.92±1.23	4.79±1.85	4.23±1.15
Feed conversion	4.39±1.11	3.60±1.22	3.60±0.77	4.62±1.91	4.70±1.91

T0= corn base diet ; T1=sorghum base diet; T2,T3,T4 = sorghum base diet supplemented with 1%,2% or 3% *Azolla pinnata*.

As showed in Table 3, feed intake of quails fed sorghum base diet (T1) was higher than control group (corn base diet). This data indicated that sorghum was palatable for quails. Another reason why the feed intake increased in T1 group, because energy level of T1 diet lower than control (T0). The quails will consume more diet to meet energy requirement. If we see the data of hen day production, quail fed sorghum diet and sorghum supplemented with 1% *Azolla pinnata* give improved egg production. This data indicated that sorghum could replace corn in laying quail diet. Feed conversion ratio also improved in T1 and T2 treatment. This positive effect of feeding sorghum and *Azolla pinnata* should be confirm in larger scale animal unit in the next experiment.

The effect of *Azolla pinnata* supplementation on egg quality was shown in Table 4. The egg weight of quails fed sorghum and *Azolla pinnata* supplementation was a little bit lower than egg from quail fed corn base diet (control). On the other hand, yolk percentage of quails fed T1 and T3 diet was higher than control group. But only quails fed T2 diet had lower albumen percentage as compared to other feeding treatment. Increasing yolk weight/percentage in quails fed T1 and T3 diet might be due to increasing crude protein and fat in those diet (Table 2).

The egg shell percentage and shell thickness of quails fed 2% and 3% *Azolla pinnata* were higher than other treatment. This data indicated that *Azolla pinnata* might be involved in shell calcification or calcium metabolism. All qualis fed *Azolla*

pinnata had higher haugh unit value than quails fed corn diet or sorghum diet (T1). It is mean *Azolla pinnata* might be increased the thickness of albumen.

Table 4. Supplementation of *Azolla pinnata* on egg quality of Japanese quail

Parameter	T0	T1	T2	T3	T4
Egg weight (g)	9.4±0.21	9.0±0.71	9.3±1.07	9.3±1.05	8.5±0.78
Yolk weight (g) (%)	2.6±0.10 27.57	2.7±0.13 30.71	2.4±0.22 25.99	2.8±0.62 29.70	2.4±0.35 27.74
Albumen (g) (%)	6.0±0.29 63.24	5.6±0.62 61.60	6.2±0.92 65.98	5.6±0.42 60.61	5.3±0.42 62.76
Egg shell (g) (%)	0.9±0.12 9.19	0.7±0.05 8.22	0.7±0.09 8.03	0.9±0.14 9.69	0.8±0.01 9.51
Shell thickness (mm)	0.15±0.01	0.15±0.02	0.15±0.02	0.18±0.01	0.16±0.01
Yolk color	2.67±0.58	1.40±0.55	4.00±1.00	4.66±0.29	7.00±0.00
Haugh Unit	91.90±8.92	89.47±3.42	92.98±2.89	92.20±1.81	94.78±0.21
MDA (mg/100g)	0.78	1.23	0.46	0.42	0.55

T0= corn base diet ; T1=sorghum base diet; T2,T3,T4 = sorghum base diet supplemented with 1%,2% or 3% *Azolla pinnata*.

Yolk color score of egg from quail fed *Azolla pinnata* was higher than corn diet or sorghum diet (T1). Increasing yolk color score due to increasing level of *Azollapinnata* supplementation. In this experiment, *Azolla pinnata* contain 1188 mg/kg beta carotene. Beta carotene is one of carotenoid that could made yolk color value increased. Another function of beta carotene was antioxidant agent, as indicated by the MDA value in Table 4. All quails fed *Azolla pinnata* diet had lower MDA level than quails fed corn diet or sorghum diet (T1). As reported by Mithraja *et.al.* (2011), *Azolla pinnata* contain tannin, phenolic and flavonoid compound as anioxidant agent for animal feed. Another report by Radhakrisnan *et.al.* (2014) showed that *Azolla pinnata* improved non-enzymatic antioxidant (Vit.E and Vit.C) in *Macrobrachium rosenbergii*. In our experiment, there are two positive effect from *Azolla pinnata* supplementation, first, *Azolla pinnata* as carotenoid source that improve yolk color score and the second, *Azolla pinnata* as antioxidant agent that decrease the MDA level in quail egg.

CONCLUSION

Azolla pinnata increased yolk color score and antioxidant status in Japanese quail egg.

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PERFORMANCE AND SELENIUM (Se) CONTENT OF MEAT OF KAMPONG CHICKEN FED Se AND VITAMIN E FORTIFIED DIET

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ABSTRACT

Mineral Selenium (Se) and vitamin E are essential micronutrient and as antioxidant in poultry for normal health. The fortification of them could prevent the damage of phospholipid membrane and enzymes and other important molecules due to free radicals. This study was aimed to evaluate the performance and Se content of meat of kampong chickens fed diet fortified by Se and vitamin E. One hundred and sixty Kampong chickens aged 8-13 weeks and Se content of the meat. The birds were fed control diet (P0); P0+ Se organic 0.2 ppm (P1); P0 + vitamin E 200 ppm (P2); P0 + combination of Se organic 0.2 ppm and vitamin E 200 ppm (P3). This experiment used a Completely Randomized Design (CRD), with 4 treatment, 4 replication, and 8 chickens. The data were analyzed using Analysis of Variances (ANOVA) for performance's parameters. The performance's parameters observed were feed consumption, body weight gain, feed conversion, final body weight. For meat analysis, one composite sample per treatment was used to analyze for Se content, and analyzed descriptively for Se content. The results showed that Se and vitamin E fortification, or its combination did not affect the performances (feed consumption, body weight gain, feed conversion, and final body weight) of kampong chickens. The treatments increased Se content of the meat 58.62% (P2), 234.48% (P3) and 717.24% (P1) than the control (P0). Fortification diet with Se organic 0.2 ppm, vitamin E 200 ppm, and combination of vitamin E 200 ppm and Se organic 0.2 ppm increased Se content of meat.

Keywords: *Kampong chicken, Se, Vitamin E, Performance, Se content of meat*

INTRODUCTION

Mineral Selenium (Se) and vitamin E are essential micronutrient and as antioxidant in poultry for normal health. The fortification of them could prevent the damage of phospholipid membrane and enzymes and other important molecules due to free radicals. Free radicals are compound that contain one or more single electron, that make them very reactive. Naturally, free radical has formed in the body through a complex chemicals process. This condition was side product from oxidation process in the cell. The function of antioxidant is to change the harmful compound to harmless compound (Surai 2003). Vitamin E has a role as antioxidant in the body that can act as scavenger to free radicals that come into the body or that formed by normal metabolism process. Vitamin E fortification 250 ppm significantly increased antioxidant status in quail that in heat stress condition (Sahin and Kucuk, 2001). Selenium is trace mineral that can effectively act as important antioxidant for animals (MacPherson, 1994).

Vitamin E and selenium working together as antioxidant that stabilized free radicals through chemical reaction, that superoxidation (O_2) which the results of over oxidation due to heat stress, and then stabilized by Glutathion peroxidase to

more stabil form with selenium. Antioxidant consumption can kept natural antioxidant status in animal (Surai 2003).

This study was aimed to evaluate the performance and Se content of meat of kampong chickens fed diet fortified by Se and vitamin E.

MATERIAL AND METHODS

One hundred and sixty unsexed, 8 weeks of age, kampong chickens (*Gallus gallus domesticus*) with average initial body weight of 602.13 ± 80.32 g head⁻¹ used in this study. The chickens were allocated in 4 treatments, 4 replications with 10 chickens in each replication. The diet used in this study was broiler commercial diet (BR21E). The supplements used were Vitamin E 50 as source of vitamin E and Se optimin 15 as source of mineral Se. Nutrient content of the diet was showed in Table 1. Fortification of vitamin E and Se in treatment diet was showed in Table 2. Parameter observed were feed consumption, final body weight, body weight gain, feed conversion and Se content in the meat.

Table 1. Diet nutrient content

Nutrient	Content
Dry matter (%) ¹⁾	87.85
Ash (%) ¹⁾	4.96
Crude protein (%) ¹⁾	21.78
Crude fiber (%) ¹⁾	5.89
Ether Extract (%) ¹⁾	5.15
Nitrogen Free Extract /NFE (%) ¹⁾	50.07
Ca (%) ¹⁾	0.78
P (%) ¹⁾	0.74
Se ²⁾ (ppm)	0.00284
Vitamin E ³⁾ (ppm)	624.9

¹⁾Analyzed at Laboratory of Feed Science and Technology, Departmen of Nutrition and Feed Technology, Faculty of Animal Science, Bogor Agricultural University (2013); ²⁾Analyzed at Laboratory of Pengujian Balai Besar Penelitian dan Pengembangan Pascapanen Pertanian (2013); ³⁾Analyzed at Laboratory of Dairy Nutrition, Departmen of Nutrition and Feed Technology, Faculty of Animal Science, Bogor Agricultural University (2013)

Table 2. Vitamin E and Se fortification in diet

Treatment	Vitamin E (ppm)		Se (ppm)	
	Fortification	Total in diet	Fortificatin	Total in diet
P0	-	624.9	-	0.00284
P1	-	624.9	0.2	0.20284
P2	200	824.9	-	0.00284
P3	200	824.9	0.2	0.20284

P0=control diet; P1=P0+ Se organic 0.2 ppm; P2=P0 + vitamin E 200 ppm; P3=P0 + combination of Se organic 0.2 ppm and vitamin E 200 ppm

RESULTS AND DISCUSION

The results showed that Se and vitamin E fortification, or its combination did not affect the performances, i.e feed consumption, body weight gain, feed conversion, and final body weight of kampong chickens (Table 3).

Table 3. Performance of kampong chickens during 5 weeks (8-13 weeks of age)

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Parameters	P0	P1	P2	P3
Cumulative feed consumption (g head ⁻¹)	2448.50 ± 171.13	2327.65 ± 111.08	2473.33 ± 50.38	2466.03 ± 161.10
Average feed consumption (g head ⁻¹ day ⁻¹)	69.96 ± 4.89	66.50 ± 3.17	70.67 ± 1.44	70.46 ± 4.60
Initial body weight (g head ⁻¹)	602.13 ± 16.52	606.08 ± 12.61	599.50 ± 10.09	600.83 ± 6.70
Final body weight (g head ⁻¹)	1308.65 ± 51.97	1306.30 ± 78.47	1303.65 ± 41.05	1305.61 ± 27.23
Body weight gain (g head ⁻¹)	706.53 ± 38.22	700.23 ± 67.20	704.15 ± 40.09	704.78 ± 27.50
Feed conversion ratio (FCR)	3.49 ± 0.10	3.42 ± 0.38	3.61 ± 0.31	3.60 ± 0.12

P0=control diet; P1=P0+ Se organic 0.2 ppm; P2=P0 + vitamin E 200 ppm; P3=P0 + combination of Se organic 0.2 ppm and vitamin E 200 ppm

The treatments increased Se content of the meat 58.62% (P2), 234.48% (P3) and 717.24% (P1) than the control (P0). This study used Se organic (selenomethionine) that absorbed by active transport mechanism with methionine then deposit into muscle.

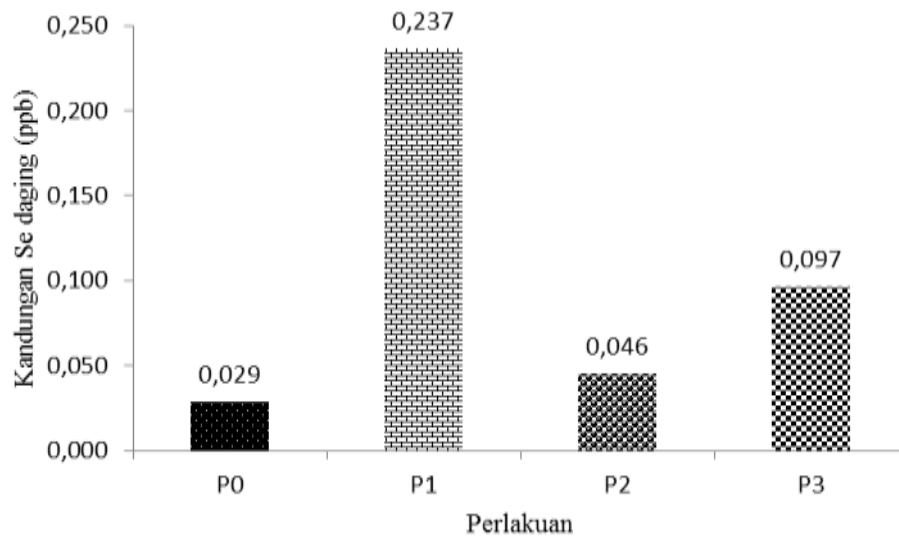


Figure 1. Selenium (Se) content of the kampong chicken meat.
 P0=control diet; P1=P0+ Se organic 0.2 ppm; P2=P0 + vitamin E 200 ppm;
 P3=P0 + combination of Se organic 0.2 ppm and vitamin E 200 ppm

CONCLUSION

Fortification diet with Se organic 0.2 ppm, vitamin E 200 ppm, and combination of vitamin E 200 ppm and Se organic 0.2 ppm did not affected the performances of kampong chickens, but increased Se content of meat.

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THE EFFECT OF FEEDING ZINC (Zn) AND VITAMIN E FORTIFIED DIETS ON DUCK EGG QUALITY STORED AT DIFFERENT TEMPERATURE DURING 21 DAYS

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ABSTRACT

Egg's yolk contain omega-3 and omega-6 fatty acid that easily oxidized during storage, so it needs to be protected. The objective of this study was to evaluate the effect of feeding Zn and vitamin E fortified diets on duck egg quality stored at different temperature during 21 days. This study used a completely randomized design (CRD) 15 treatment, 3 replication. The treatment were combination treatment diet (R1, R2, R3, R4, R5), stored temperature (T30 and T5) and stored periode (D0 and D21). The treatment diets were R1(control diet), R2(R1+40 IU of vitamin E), R3(R1+80 IU of vitamin E), R4 (R1+100 ppm ZnOrganic), and R5(R1+200 ppm ZnOrganic). Ninety duck eggs used in this study. Parameters observed were egg weight, the percentage of eggshell weight, albumen weight and yolk weight, *haugh unit*, yolk color score, and eggshell thickness. The results showed that fortification of Zn 200 ppm in the diet could maintain the egg quality stored at room temperature (30 °C) during 21 days. Fortification of vitamin E 80 IU in the diets was able to maintain the egg quality at refrigerator (5 °C) during 21 days. It concluded that fortification Zn and vitamin E in the diet could maintain the quality of duck egg stored during 21 days.

Key words: *Duck eggs, Physical quality egg, Storage, Vitamin E, Zn organic*

INTRODUCTION

Duck egg production in Indonesia in 2012-2014 were 275.938, 290. 369 and 297.074 tonnes per year, respectively, while egg production of laying hens were 1.139.949, 1.224.402, and 1.299.199 tons per year respectively (BPS 2015). Duck eggs contain 12.81 g of protein and 13.77 g of fat per 100 g egg (USDA 2015).

Eggs are good source of fatty acids, egg yolks contain omega-3 and omega-6 fatty acids as DHA (Docosahexaenoic Acid) and EPA(Docosahexaenoic Acid) derived from feed (Hartono *et al.*, 2008). Omega-3 and omega-6 fatty acids have a beneficial effect to prevent cardiovascular disease, cancer, alzheimer and schizophrenia (Simopoulos 2002). Unsaturated fatty acids susceptible to damage due to oxidation process during storage. Therefore, omega-3 and omega-6 fatty acids in duck eggs need to be protected by the addition of antioxidants in the diets. Antioxidants in biological systems has a role in counteracting free radicals that can resist oxidative damage, while antioxidants in the food system has a role for inhibiting the fat oxidation (Hartanto 2012).

Vitamin E and Zn can be added to ducks dietas natural antioxidants that can be metabolized and transferred into the egg. Vitamin E is fat soluble and as antioxidants has a role for breaking the chain of peroxide in membranes and protecting PUFAS (Poly Unsaturated Fatty Acids) from oxidation (Iswara 2009). Rink and Kirchner (2000) states that Zn act as antioxidants and protect the cells from the effects of oxidative damage. The objective of this study was to evaluate the effect of

feeding Zn and vitamin E fortified diets on duck egg quality stored at different temperature during 21 days.

MATERIALS AND METHODS

The diets were isocaloric (2850 kcal ME / kg) and isoprotein (16%) (Leeson and Summers, 2005). The composition and nutrients content in the experimental diets is presented in Table 1.

Table 1. The composition and nutrients content of control diet (R1), as fed

Feed Ingredient	(%)
Yellow corn	46
Rice bran	15.2
Soybean meal	19
Fish meal	8
Palm oil	3.5
Fish oil	1
CaCO ₃	6.5
NaCl	0.2
Premix	0.5
DL-Methionin	0.1
Total	100
Nutrient content *	
Crude protein (%)	15.9
Crude fiber (%)	10.3
Crude fat (%)	3:52
Ash (%)	12.5
Gross energy (kcal /kg)	3788

* Analysis at Laboratory of Nutrition and Feed Technology, Faculty of Animal Science, Bogor Agriculture University, 2014

A total of 90 of 20 weeks old ducks were randomly divided into five treatments. This study used a completely randomized design with 5 treatments and 3 replications. The treatments were combination of treatment diets (R1, R2, R3, R4, R5), storage temperature (T30 and T5) and storage period (D0 and D21). The treatment diets were R1(control diet), R2(R1+40 IU vitamin E), R3(R1+80 IU vitamin E), R4 (R1+100 ppm ZnOrganic), and R5(R1+200 ppm ZnOrganic). The ducks were given adaptation period for 2 weeks at the age of 20-22 weeks to introduce the treatment diet. The Pattern of diet adaptation was 75% of commercial ration (CR) and 25% treatment diet (TD) during 4 days, 50% CR and 50% TD during 4 days, 25% CR and 75% TD during 3 days and 0% CR and 100% TD for the last 3 days. Treatment diets were fed for 8 weeks at 22-30 weeks of age and the water was given *ad libitum* each day. Two eggs from each replication were separated to be stored for 0 days and 21 days (room temperature and refrigerator). After 21 days, the eggs were measured for quality of the eggs including egg weight (g), percentage of yolk weight (g), albumen weight (g), and egg shell weight (g), eggshell thickness (mm), yolk color score, and Haugh unit. Yolk color scores was measured by the Roche Yolk Colour Fan with scale numbers 1-15.

Haugh unit was obtained by calculating in the logarithm of the high of albumen and then transformed into a correction value of the function of egg weight (Wahju 1997).

$$HU = \text{Log } 100 (H + 7.57 - 1.7 \cdot W^{0.37})$$

Description: H = high of albumen; W = eggs weight

Data Analysis

The data were analyzed by analysis of variance. If there was a difference between treatments, the data were further analysed using Duncan's multiple range test according to Steel and Torrie (1993).

RESULTS AND DISCUSSION

Egg weights

Addition of vitamin E, organic Zn and storage of eggs at 5°C decreased egg weight significantly ($P < 0.05$). The effect of treatments on egg weight of ducks is presented in Table 2.

Table 2. Effect of treatment on egg weight

Treatments	Egg weight (g/egg)
R1D0	66.56 ± 1.95 ^{abc}
R2D0	73.74 ± 3.27 ^a
R3D0	68.34 ± 5.22 ^{abc}
R4D0	63.88 ± 2.04 ^{bc}
R5D0	69.93 ± 4.22 ^{ab}
R1T1D21	64.28 ± 4.85 ^{bc}
R2T1D21	65.42 ± 2.28 ^{abc}
R3T1D21	63.31 ± 5.22 ^{bc}
R4T1D21	54.73 ± 4.40 ^d
R5T1D21	64.62 ± 1.78 ^{abc}
R1T2D21	62.87 ± 5.15 ^{bcd}
R2T2D21	67.40 ± 0.80 ^{abc}
R3T2D21	60.13 ± 3.71 ^{cd}
R4T2D21	64.73 ± 10.01 ^{abc}
R5T2D21	64.97 ± 7.12 ^{abc}

Notes: mean in the same column with different superscripts differ significantly ($P < 0.05$)

R1D0 (diet without vitamin E and Organic Zinc and storage), R2D0 (R1+ 40 IU vitamin E, without storage), R3D0 (R1+ 80 IU vitamin E, without storage), R4D0 (R1+ 100 ppm Zn organic, without storage), R5D0 (R1+ 200 ppm Zn organic, without storage), R1T1D21 (R1+ storage of 21 days at a temperature of 29.29 -30.07 ° C), R2T1D21 (R1+ 40 IU vitamin E, Storage 21 days at a temperature of 29.29 -30.07 ° C), R3T1D21 (R1+ 80 IU vitamin E, storage of 21 days at a temperature of 29.29 -30.07 ° C), R4T1D21 (R1+ 100 ppm Zn organic, Storage 21 days at a temperature of 29.29 -30.07 ° C), R5T1D21 (R1+ 200 ppm Zn organic, Storage 21 days at a temperature of 29.29 ° -30.07 ° C), R1T2D21 (R1+ storage of 21 days at a temperature of 5°C), R2T2D21 (R1 + 40 IU vitamin E, Storage 21 days at a temperature of 5°C), R3T2D21 (R1+ 80 IU vitamin E, storage of 21 days at a temperature of 5°C), R4T2D21 (R1+ 100 ppm Zn organic, Storage 21 days at a temperature of 5°C), R5T2D21 (R1+ 200 ppm Zn organic, 21 days storage at 5°C temperature)

The average of eggs weight without storage was range from 63.88-73.74 g/egg. Storage of eggs for 21 days decreased eggs weight. Chukwuka *et al.* (2011) stated that the eggs quality was influenced by the management of housing and feeding, egg storage time and temperature.

Addition of 40 IU vitamin E in the diet without storage treatment resulted heavier eggs than the control diet (R1D0). The results showed that egg weight decreased after 21 days of storage both at refrigerator temperature and at room temperature. Egg storage at room temperature (29.29-30.07 °C) for 21 days decreased egg weight as much as 8.79%. Supplementation of 100 ppm Zn was not able to sustain the eggs weight stored during 21 days at room temperature (29.29 - 30.07 °C). According to Raji *et al.* (2009) that the storage for 28 days at the temperature of 32 °C and 5 °C decreased egg weight as much as 13.33% and 6.10%. The eggs stored at 30°C for 20 days would loss exosamine and hexose of ovomucin by approximately 50% and decrease sialic acid by 12% (Stadelman and Cotterill 1995). EFSA Panel on Biological Hazards (2014) stated that the the eggs weight reduced during storage due to loss of water vapor and carbon dioxide through pores of egg shell.

Eggshell Weights

Addition of vitamin E, organic Zn and eggs storage at 5°C and room temperature (29.29 -30.07 °C) did not affect the percentage of eggshell weight. The effect of treatments on shell weight and shell weight percentages was presented in Table 3.

Table 3. The effect of treatment of shell weight and shell weight percentages

Treatment	Eggshell weights (g)	Eggshell weight percentage (%)
R1D0	8.69 ± 0.54	13.05 ± 0.85
R2D0	8.74 ± 0.70	11.85 ± 0.86
R3D0	8:20 ± 0.23	12.03 ± 0.75
R4D0	8.33 ± 0.30	13.05 ± 0.88
R5D0	8.26 ± 0.39	11.82 ± 0.24
R1T1D21	7.62 ± 0.28	11.88 ± 0.70
R2T1D21	7.95 ± 0.35	12.15 ± 0.11
R3T1D21	7:20 ± 0.59	11.37 ± 0.00
R4T1D21	6.90 ± 1.13	12.66 ± 2.33
R5T1D21	7.60 ± 0.40	11.77 ± 0.82
R1T2D21	8.18 ± 0.54	13.03 ± 0.28
R2T2D21	8.30 ± 0.36	12.31 ± 0.44
R3T2D21	7.77 ± 0.84	12.92 ± 1.24
R4T2D21	7.53 ± 0.72	11.72 ± 0.76
R5T2D21	8.00 ± 0.95	12.40 ± 1.76

According to Namra *et al.* (2009), the decreasing of eggshell weight percentage can be due to antagonist interaction between Zn and Ca when given in high amounts. The addition of 200 ppm organic Zn in the diet in this study did not inhibit Ca metabolism and it was supported by previous study that the addition of 200 ppm organic Zn did not decrease the percentage of egg shell weight (Darmawan 2013). Standard of egg shell weight percentage is 12.0% (Suprijatna *et al.* 2005). The

percentage of egg shell weight stored at 5 °C was 11.72% -13.03%, while the percentage of egg shell weight stored at room temperature was 11.37% - 12.66%). Idowu *et al.* (2011) stated that the addition of various types of Zn could improve egg weight, shell thickness, shell weight and Haugh units.

Albumen Weight

Addition of vitamin E, organic Zn and eggs storage at 5°C and room temperature (29.29 -30.07°C) decreased the percentage of albumen weight significantly (P<0.05). Effect of treatments of albumen and albumen weight percentages is presented in Table 4.

Albumen weight without storage treatments was 34.41-37.06 g. According to Budiman and Rukmiasih (2007), the albumen weight was 33.96 ± 3.94 g, while according Nugraha *et al.* (2013) albumen weights ranged from 36.90-37.56 g. The addition of 100 ppm of organic Zn resulted the largest albumen weight percentage by 55.34%. Idowu *et al.* (2011) stated that the addition of 35 ppm Zn in the diet led to a decrease in the percentage of albumen weight than the control. Darmawan (2013) stated that addition of 200 ppm Zn in the diet produced albumen weight of 30.37 g (56.65%) - 32.32 g (55.88%).

Table 4. Effect of treatments of albumen and albumen weight percentage

Treatments	Albumen weight (g)	The percentage of Albumen (%)
R1D0	34.41 ± 2:24	51.65 ± 2.25 ^{abcd}
R2D0	36.94 ± 3.63	50.02 ± 2.84 ^{abcde}
R3D0	37.04 ± 0.81	54.35 ± 3.05 ^{ab}
R4D0	35.38 ± 2.71	55.34 ± 2.81 ^a
R5D0	37.06 ± 2.78	52.97 ± 1.08 ^{abc}
R1T1D21	28.27 ± 3.80	43.85 ± 2.66 ^{de}
R2T1D21	21.95 ± 0.95	33.61 ± 2.62 ^f
R3T1D21	29.40 ± 2:43	46.44 ± 0.00 ^{bcde}
R4T1D21	28.03 ± 4:11	51.53 ± 9.65 ^{abcd}
R5T1D21	27.20 ± 0.60	39.61 ± 1.97 ^e
R1T2D21	31.08 ± 3:22	49.40 ± 1.77 ^{abcde}
R2T2D21	30.33 ± 2:58	45.04 ± 4.36 ^{cde}
R3T2D21	28.07 ± 3.62	46.76 ± 6.57 ^{bcde}
R4T2D21	29.83 ± 8.62	45.60 ± 7.79 ^{cde}
R5T2D21	31.60 ± 3:58	48.66 ± 2.38 ^{abcde}

Notes: mean in the same column with different superscripts differ significantly (P <0.05)

The addition of 40 IU vitamin E and 200 ppm Zn has not been able to maintain (P >0.05) the albumen weight percentage stored for 21 days at room temperature. Raji *et al.* (2009) stated, that declining of albumen weight was caused by the increasing of storage temperature. The Storage leads to loss of CO₂ and water in albumen through eggshell pores and change the carbonic acid into carbon dioxide (Raji *et al.* 2009). Stadelman and Cotterill (1995) stated that the component of albumen was water, protein and ash which was 88%, 10,6% and 0,6% respectively.

Yolk Weight

Addition of vitamin E, organic Zn and eggs storage at 5°C and room temperature (29.29 -30.07°C) increased the percentage of yolk weight significantly (P<0.05). Effect

of the treatments of yolk weight and egg yolk weight percentage can be seen in Table 5.

Addition of 40 IU Vitamin E without storage resulted the largest percentage of yolk weight by 28.06 g ($38.13 \pm 3.14\%$), while the addition of 100 ppm Zn organic caused a smaller percentage of yolk weight ($31.61 \pm 2.45\%$) than the control. Suprijatna *et al.* (2005) stated that the percentage of the yolk weight was 35.4%. Bell and Weaver (2002) stated that the percentage of yolk weight was around 30% -32% of the egg weight. The addition of 30 ppm vitaminE and 0.15 ppm selenium in the diet could increase the yolk weight as much as 0.7%(Zduńczyk *et al.* 2013).

The yolk percentage of this study increased both of storage at room temperature and 5 ° C. The supplementation of 40 IU vitamin E and 200 ppm organic Zn with storage treatment for 21 days at room temperature caused the increasing of yolk weight. The increasing of yolk weight can be caused by the movement of water from the albumen into the yolk. According to Stadelman and Cotterill (1995) , water move from the albumen into the yolk during storage of eggs that will reduce the solid concentration in yolk. Rose *et al.* (1966) stated that solid concentration in the yolk (52.29%) stored at 4 ° C for one week decreased to 50.09%. Fromm (1966) in his research stated that the eggs stored at 24 ° C for 16 days led to decrease the yolk solid concentration of 53.5% to 49%. In addition, the increasing of yolk weight percentage can be caused by decreasing of albumen and egg shell weight. The content of the yolk was dominated by lipids (31.8% -35.5%) and protein (15.7% -16.6%), while the composition of inorganic elements in yolk was about 1.1% as ash (Stadelman and Cotterill 1995).

Table 5. Effect of the treatments of egg yolk weight and yolk weight percentage

Treatments	Yolk weight (g)	The yolk percentage (%)
R1D0	23.47 ± 1.80	35.29 ± 2.84 ^{def}
R2D0	28.06 ± 1.45	38.13 ± 3.14 ^{cdef}
R3D0	23.10 ± 4.25	33.61 ± 3.78 ^{ef}
R4D0	20.18 ± 1.44	31.61 ± 2.45 ^f
R5D0	24.61 ± 1.23	35.21 ± 0.85 ^{def}
R1T1D21	28.40 ± 1.14	44.27 ± 1.96 ^{bc}
R2T1D21	35.52 ± 2.88	54.23 ± 2.51 ^a
R3T1D21	26.71 ± 2.20	42.19 ± 0.00 ^{bcd}
R4T1D21	19.80 ± 7.23	35.82 ± 11.94 ^{cdef}
R5T1D21	30.70 ± 1.90	47.51 ± 2.54 ^{ab}
R1T2D21	23.60 ± 1.92	37.57 ± 1.86 ^{cdef}
R2T2D21	28.77 ± 3.16	42.65 ± 4.21 ^{bcd}
R3T2D21	24.30 ± 4.04	40.32 ± 5.45 ^{bcd}
R4T2D21	27.37 ± 4.80	42.68 ± 7.37 ^{bcd}
R5T2D21	25.37 ± 4.57	38.94 ± 3.97 ^{bcd}

Notes: mean in the same column with different superscripts differ significantly (P <0.05)

Haugh Unit

Addition of vitamin E, organic Zn and eggs storage at 5°C and room temperature (29.29 -30.07°C) decreased the value of Haugh units significantly (P<0.05). Effect of the treatments of the Haugh units presented in Table 6.

The results showed that supplementation of 200 ppm organic Zinc without storage resulted the highest Haugh unit value. According to Stadelman and Cotterill 1995, egg quality was divided into several categories, namely AA quality (Haugh unit value more than 72), A quality (Haugh unit value 60-72), and B quality (Haugh unit values less than 60). The Haugh unit value of the fresh egg was AA egg quality.

Addition of 200 ppm Zinc decreased as much as 7.78% Haugh unit value after being stored for 21 days at temperature of 5°C. Zduńczyk *et al.* (2013) stated that the addition of vitamin 30 ppm E increased the Haugh unit value of 5.02%, while Idowu *et al.* (2011) stated that the addition of 35 ppm Zinc increased the Haugh unit until 4.59%. The higher Haugh unit value showed the greater the quality of albumen protein (Stadelman and Cotterill 1995). Ovomucin was a type of protein found in albumen that contribute to make the structure of albumen.

Table 6. Effect of treatments of the Haugh unit value

Treatment	Haugh Unit
R1D0	91.21 ± 6.49 ^{ab}
R2D0	91.79 ± 1.96 ^{ab}
R3D0	93.52 ± 4.60 ^a
R4D0	91.14 ± 3.79 ^{abc}
R5D0	93.56 ± 3.49 ^a
R1T1D21	68.61 ± 6.28 ^{ef}
R2T1D21	48.06 ± 14.52 ^g
R3T1D21	69.48 ± 2.11 ^{abc}
R4T1D21	64.42 ± 2.12 ^f
R5T1D21	51.46 ± 18.93 ^g
R1T2D21	77.38 ± 7.20 ^{bcd}
R2T2D21	70.43 ± 8.46 ^{def}
R3T2D21	84.74 ± 6.44 ^{abcd}
R4T2D21	75.09 ± 1.83 ^{cdef}
R5T2D21	86.28 ± 5.48 ^{abc}

Notes: mean in the same column with different superscripts differ significantly (P <0.05)

Yolk Colour Scores

Addition of vitamin E, organic Zn and eggs storage at 5°C and room temperature (29.29 -30.07°C) decreased the yolk colour score significantly (P<0.05). Effect of treatment of yolk color scores is presented in Table 7. The results showed that the supplementation of Zn and vitamin E with 46% yellow corn in the diets resulted the yolk color score of 6.61-7.89. The feed Xanthophylls in the diets will lead the colour of orange or red yolk (Castaneda *et al.* 2005). Yellow corns contain xanthophylls about 17 mg/ kg (Moros *et al.* 2002). Eggs stored at 5 ° C for 21 days had lower yolk scores than the those at room temperature. The supplementation of 80 IU vitamin E without storage resulted in low yolk scores, but it could maintain yolk colour score after being stored for 21 days at 5°C as well as at room temperature.

Table 7. Effect of treatments of yolk color scores

Treatment	yolk colorscores
R1D0	7.72 ± 0.67 ^a
R2D0	7.61 ± 0.10 ^a
R3D0	6.61 ± 2.26 ^{ab}
R4D0	7.89 ± 0.38 ^a
R5D0	7.56 ± 0.63 ^a
R1T1D21	2.83 ± 1.04 ^{cd}
R2T1D21	1.75 ± 0.25 ^d
R3T1D21	6.50 ± 0.00 ^{ab}
R4T1D21	3.17 ± 0.58 ^{cd}
R5T1D21	3.50 ± 0.00 ^c
R1T2D21	7.50 ± 0.50 ^a
R2T2D21	7.67 ± 0.58 ^a
R3T2D21	5.33 ± 1.15 ^b
R4T2D21	5.67 ± 1.53 ^b
R5T2D21	6.67 ± 0.58 ^{ab}

Notes: mean in the same column with different superscripts differ significantly (P <0.05)

The yolk color scores that resulted by addition of 100 ppm and 200 ppm Zinc in the diet was not different from the control. The decreasing of yolk color score after a 21-day storage can be due to declining of yolk fat quality due to oxidation process. The storage of eggs caused the displacement of water from the albumen into the yolk and reduced lysozyme content and ruptured the vitelin membranes (Bieber et al. 2015). Yolk color scores was affected by the chemical structure of the xanthophylls, antioxidants and fat content in the feed (Stadelman and Cotterill 1995).

Eggshell Thickness

Addition of vitamin E, organic Zn and eggs storage at 5°C and room temperature (29.29 -30.07°C) decreased the eggshell thickness significantly (P<0.05). Effect of treatments of eggshell thickness is presented in Table 8.

Addition of 40 IU resulted the largest eggshell thickness (0.38 mm ± 0.02). The eggshell thickness that resulted by supplementation of 200 ppm organic Zinc was not different from the control, it was consistent with the study of Darmawan (2013) that the addition of 200 ppm organic Zinc did not interfere the calcium metabolism. The supplement of 100 ppm Zn organic with the storage treatment during 21 days at room temperature reduced the eggshell thickness. Zinc has an important role in the formation of egg shell and membrane cell, because Zn was a cofactor of enzyme for the formation of eggshell carbonate (Idowu et al. 2011). The results showed that the longer the eggs stored would reduce eggshell thickness because of carbonate ions evaporation during storage. Eggshell contains calcium carbonate (94%), magnesium carbonate (1%), calcium phosphate (1%), and organic materials such as proteins (4%) (Stadelman and Cotterill 1995).

Table 8. Effect of treatments of eggshell thickness (mm)

Treatments	Eggshell thickness (mm)
R1D0	0.36 ± 0.03 ^{ab}
R2D0	0.39 ± 0.02 ^a
R3D0	0.39 ± 0.00 ^a
R4D0	0.37 ± 0.05 ^{ab}
R5D0	0.37 ± 0.02 ^{ab}
R1T1D21	0.36 ± 0.01 ^{ab}
R2T1D21	0.35 ± 0.00 ^{abc}
R3T1D21	0.35 ± 0.00 ^{abc}
R4T1D21	0.30 ± 0.05 ^c
R5T1D21	0.35 ± 0.02 ^{abc}
R1T2D21	0.35 ± 0.05 ^{abc}
R2T2D21	0.39 ± 0.01 ^a
R3T2D21	0.36 ± 0.03 ^{ab}
R4T2D21	0.34 ± 0.03 ^{abc}
R5T2D21	0.33 ± 0.03 ^{bc}

Notes: mean in the same column with different superscripts differ significantly (P <0.05)

CONCLUSION

Adiition of 200 ppmZn or80 IU vitamin E in the diet could maintain the quality of duck egg stored during 21 days.

ACKOWLEGEMENT

This research was funded by Indonesian Ministry of Education and Culture, Directorate General of Higher Education with contract number: 77/IT3.11/LT/2014

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CARCASS CHARACTERISTICS AND INTESTINAL MORPHOLOGY OF MALE LAYING QUAILS AFFECTED BY ADMINISTERED INDIGENOUS PROBIOTICS LACTIC ACID BACTERIA

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ABSTRACT

This study was conducted to investigate the effect of indigenous probiotics Lactic Acid Bacteria (LAB) on carcass characteristics and intestinal morphology of male laying quails. Ninety two unvaccinated day old quails were assigned randomly into four treatment supplemented groups of mixed culture probiotics LAB *Lactobacillus murinus* (Ar3), *Streptococcus thermophilus* (Kd2), and *Pediococcus acidilactici* (Kp6). The fourth treatments were (T0) one group of unsupplemented birds as control and (T1), (T2), (T3) were supplemented orally with that those probiotics as much as 10^7 , 10^8 and 10^9 CFU/ml/bird/day respectively. All of treatment groups were replicated into four, with six birds each. The antibiotic-free diet was formulated and provided *ad libitum*. At the end of experiment, the euthanasia of 24 quails were measured to obtain data of slaughtered weight, carcass weight and carcass parts (breast, thigh, wing and back) and intestinal morphology. The data were analyzed by analysis of variance followed by Duncan's Multiple Range Test (DMRT). The results showed that the slaughtered weight, carcass weight and breast weight were significantly affected by probiotics Lactic Acid Bacteria ($P < 0,05$) and improved significantly of intestinal morphology of quails.

Keywords: *Probiotics lactic acid bacteria, Carcass characteristic, Intestinal morphology, Male laying quails*

INTRODUCTION

The ban on antibiotics as growth promoters has been a challenge for animal nutrition, increasing the need to find alternative methods to control intestinal health. There is currently a world trend to reduce the use of antibiotics in animal feed due to protect animal products from antibiotic residues, as well as the concern that some therapeutic treatments for human diseases might be jeopardized due to the appearance of resistant bacteria (Javadi *et al.*, 2012). Probiotics is alternative to antibiotics as growth promoters of broilers (Sri-Harimurti and Rahayu, 2010); and defined as living microorganism which given to animals, assist in the establishment of an intestinal microbial balance which is beneficial to the host and antagonistic to harmful microbes (Denly *et al.* 2003), and gut health resulting in greater intestinal enzyme activities and nutrient availability (Angel *et al.*, 2005; Willis and Reid, 2008). Potential beneficial effects of probiotics for farm animals such as greater resistance to infectious diseases, increased growth rate, improved feed conversion, improved digestion, better absorption of nutrients, improved carcass quality and less contamination (Tannock, 1999). Reported by Sri-Harimurti *et al.* (2014), that probiotics induced to enhancement of short chain fatty acids of male quails. The short chain fatty acids which are by products of bacterial fermentation stimulate the

proliferation of epithelial cells of the intestine (Ichikawa *et al.*, 1999 in Gunal *et al.*, 2006). The main objective was to study the carcass characteristics and intestinal morphology affected by administered indigenous probiotics Lactic Acid Bacteria (LAB).

MATERIALS AND METHODS

A total of 96 day old of male quails were used in this study conducted at Laboratory of Poultry Science, Faculty of Animal Science, Gadjah Mada University, Yogyakarta, Indonesia. Chicks were individually weighed and randomly divided into 4 treatment groups of Lactic Acid Bacteria probiotics mixture of *Lactobacillus murinus* (Ar3), *Streptococcus thermophilus* (Kd2), and *Pediococcus acidilactici* (Kp6) which were orally supplementation consisted of (T0) one group of unsupplemented and three groups of supplemented chickens as much as 10^7 (T1), 10^8 (T2) and 10^9 CFU/ml/bird/day (T3) respectively. All of treatment groups were replicated into 4, with 6 chickens each. The antibiotic-free diet was formulated and provided *ad libitum*. To study the carcass characteristics of birds, six birds from each treatment group were sacrificed on 6th week of age. Weight of carcass and cut up meat parts were calculated separately. To study the intestinal morphology, 4 cm segments of duodenum (from the top of one side loop to distal), jejunum and ileum (from Mickel diverticulum to distal) were removed, rinsed, and placed into 10% buffered formalin until further processing (histological process followed haematoxylin-eosin stained). Pictures of villus height and width, crypt depth were obtained with Olympus BX 51 microscope replenished by Olympus DP 12 projector in magnification 40 x, and used the monitor of JVC TMH 1750C. The data were analyzed using analysis of variance in a Completely Randomized Design and means were compared for significant differences by Duncan New Multiple Range Test.

RESULTS AND DISCUSSION

The result of same studies of supplementation of these indigenous lactic acid bacteria as probiotics to broiler has been showed that its benefited the host by stimulating appetite. Higher cumulative feed in take was inline with the greater live body weight or slaughtered weight(Sri-Harimurti and Widodo, 2014). Thus, higher slaughtered weightwas inline with the greater carcass weight and breast of male quail.

Result for carcass characteristics analyses shown in Table 1. Result for production parts parameters showed the all of the treatments not significantly affected on weight of thigh, wing and back. However slaughtered weight, carcass weight and breast of male quail were statistically significant ($P < 0.05$) from the control groups (unsupplemented probiotics). Those results were supported by Peyman *et al.* (2014) that supplementation of lactic acid, butyric acid and acetic acid significantly increased live body weight of male Japanese quail but not significantly affected on carcass weight, thigh, wing and back.

Regarding the intestinal morphology traits, the results as presented in Table 2 showed a positif effect of probiotics on villi height, villi width and crypt depth of duodenum, jejunum and ileum. That those were statistically significant ($P < 0.05$) from the control groups (unsupplemented probiotics).

Table 1. Effect of probiotics supplementation on slaughtered weight, carcass weight, carcass partsof male laying quails

	T0	T1	T2	T3
Slaughtered weight (g)	145.25±3.77 ^a	155.0±6.78 ^{ab}	177.25±5.38 ^c	162.25±6.80 ^b
Carcass weight (g)	97.0±1.41 ^a	106.0±5.60 ^b	107.75±3.86 ^b	111.0±8.98 ^b
Breast (g)	32.25±1.71 ^a	38.0±5.48 ^b	38.75±1.71 ^b	42.5±3.42 ^b
Thigh (g)	24.0±2.94	26.5±1.73	23.25±3.20	25.75±3.86
Wing (g)	10±0.8	10.75±2.21	11.75±1.50	12.75±2.62
Back (g)	30.75±2.21	30.75±3.50	34.0±0.81	30.0±3.56

^{abcd}Means with different superscripts columnwise differ significantly at (P< 0.05).

Table 2. Effect of probiotics supplementation on intestinal morphology of male laying quails

Segmentof intestine	Supplementation of probiotics treatments			
	T0	T1	T2	T3
Duodenum				
Villus height(µm)	271.3 ± 2.868 ^a	322.7 ± 1.846 ^b	324.8 ± 2.061 ^b	316.6 ± 10.199 ^b
Villuswidth(µm)	113.6 ± 4.336 ^a	138.6 ± 2.359 ^b	146.7 ± 16.635 ^b	145.3 ± 5.856 ^b
Crypt depth(µm)	61.3 ± 1.497 ^a	72.5 ± 6.250 ^b	69.9 ± 4.558 ^b	69.1 ± 4.578 ^b
Jejunum				
Villusheight(µm)	256.7 ± 2.521 ^a	293.2 ± 13.020 ^b	285.7 ± 11.274 ^b	288.4 ± 3.805 ^b
Villuswidth(µm)	64.0 ± 8.444 ^a	123.6 ± 10.141 ^b	134.7 ± 19.412 ^b	124.3 ± 15.658 ^b
Crypt depth(µm)	43.5 ± 1.979 ^a	65.1 ± 3.245 ^b	60.8 ± 5.302 ^b	67.0 ± 6.489 ^b
Ileum				
Villusheight(µm)	176.7 ± 3.897 ^a	244.5 ± 1.161 ^b	253.7 ± 9.477 ^b	246.5 ± 6.654 ^b
Villuswidth(µm)	59.9 ± 4.368 ^a	86.6 ± 5.942 ^b	85.9 ± 6.235 ^b	91.9 ± 3.549 ^b
Crypt depth(µm)	33,5 ± 1,927 ^a	55,7 ± 1,314 ^b	55,7 ± 3,560 ^b	55 ± 6,738 ^b

^{a, b}. Means with different superscripts columnwise differ significantly at (P< 0.05); T0: Control; T1: T2; T3 10⁷; 10⁸; 10⁹ CFU/ml/bird/day

Thus, it can be indicated that the improvement of intestinal histological changes associated with a higher ileal short-chain fatty acid production. The short chain fatty acids especially butyric acid stimulate the proliferation of epithelial cells of the intestine. Lactic acid bacteria do not produce butyric acid themselves, they increase butyric acid concentration in the gut indirectly, as they stimulate proliferation of butyric acid producing bacteria. This mechanism is called “cross-feeding mechanism” (Van Immerseel *et al.* 2009). Those reports were supported by Sri-Harimurti *et al.* (2014) that oral supplementation of mixture *Lactobacillus murinus* (Ar3), *Streptococcus thermophilus* (Kd2), and *Pediococcus acidilactici* (Kp6) as probiotics, increased the production of short chain fatty acids propionate and butyrate in the ileum of male quail.

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CHOLESTEROL, GLUKOSA AND URICACID IN BROILER CHICKENWITH DIETARY HERBS AS FEED ADDITIVE

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ABSTRACT

This research was aimed to reduce the blood and meat cholesterols of broiler and to know the glucose and uric acid. The research materials were 80 female day old chickens (DOC) of broiler MB 200 Platinum DOCs which were reared for 5 weeks at battery-postal cages and herbal mixture consisted of several herbal ingredients such as moringa leaves, noni fruit, turmeric, garlic and ginger which fermented with lactic acid bacteria (LAB). The research method was in vivo experimental using a completely randomized design (CRD) with 4 treatments, and each treatment was repeated 5 times, there were 4 broiler chickens at each repetition. Treatment consisted of R₀: Feed with herbal mixture addition of 0%; R₁: Feed with herbal mixture addition of 2%; R₂: Feed with herbal mixture addition of 4%; and R₃: Feed with herbal mixture addition of 6%. Data were analyzed using analysis of variance. The results showed that the use of herbal mixture in feed with different levels were significantly different (P<0.05) on blood and breast meat cholesterols but were not significant (P>0.05) on blood glucose and uric acid. The conclusion of this research was that the mixed herbs could be used in broiler feed up to 6% which could reduce blood and breast meat cholesterols; it could be used as feed additif which acts in maintain stamina and health of broiler.

Keywords: *Broiler, Cholesterol, Mixed herbs, Protein*

INTRODUCTION

The growth and health of broiler chicken are very much affected by the quality and quantity of the given feed. The quality of feed's nutrient not necessarily gives optimum growth effect so breeders usually give feed additives which can spur the growth. In the last five years, the government has begun to limit the use additives in the form of antibiotic. The continuous use of antibiotic can cause residue in the chicken meat. So, more natural alternatives should be found, one which can increase growth but still safe for human.

Feed additives like probiotics, vitamins, hormones, and herbal plants can be used as antibiotic alternatives as well as enhancer of chicken's appetite and health. Feeding chicken with herbs is aimed to improve animal health and stamina, growth stimulator, feed efficiency improvement, and the most important is reducing fat and cholesterol.

Herbs which are usually used for chicken including *Morinda citrifolia* L. fruit, *Andrographis paniculata* (Burm.f.) Wall. ex. Nees leaves, *Zingiber officinale* L. tuber, *Curcuma domestica* Valetton. tuber, *Languas galangal* Stuntz. tuber, *Curcuma xanthorrhiza* Roxb. tuber, *Piper betle* L. leaves, *Phaleria macrocarpa* Boer., *Kaempferia galanga* L. tuber, *Allium sativum* L. bulb etc. Herbs can be used as single or mixed preparations and served through drinking water and or feed (Shahid and Bhangar, 2006; Karadi *et al.*, 2006; Sharma *et al.*, 2013).

METHODS

The study used 100 broiler chickens. Cages used were cage group. Broiler chickens were placed in 20 different cages each cage filled with 4 broilers equipped with a tube feeder and a drink. The feed material used consisted of soybean meal, fish meal, lysine, methionine were purchased from *PT. Cheil Jedang Superfeed Indonesia*, corn, bran, coconut oil, topmix, lime, and fermeherbafit, consisting of 100 g turmeric, 100 g ginger, 50 g noni, 25 g garlic, 25 g moringa leaves, 10 g sugar and 100 ml water. Herbal ingredients are chopped and fermented with lactic acid bacteria (LAB), incubated in batch culture for 3 days. After dried at 40 ° C for 2 days and then mashed. Fermeherbafit nutrient content are 2200 kcal Metabolic Energy, 11.90% Crude Protein, 3.84% Crude Fat, 14.51% Crude Fiber, 0.07% Calcium and 1.4% Phosphorous (results of laboratory analysis of Animal Nutrition and Feed Science, 2014).

The study was conducted by using a completely randomized design (CRD), with four kinds of treatment i.e. R₀ = Control (fermeherbafit 0%), R₁ = fermeherbafit (2%), R₂ = fermeherbafit (4%), and R₃ = fermeherbafit (6%). Data were analyzed using analysis of variance (Steel and Torrie, 1993). The composition of feed is presented in Table 1.

Table 1. Nutrien Composition of Feed

Feedstuffs	R ₀	R ₁	R ₂	R ₃
	%			
Corn	50	50	50	50
Bran	15	13	11	9
Soybean Meal	20	20	20	20
Fish Meal	10	10	10	10
Coconut Oil	3.5	3.5	3.5	3.5
Premix	0.5	0.5	0.5	0.5
CaCO ₃	0.5	0.5	0.5	0.5
Methionin	0.25	0.25	0.25	0.25
Lysin	0.25	0.25	0.25	0.25
Mixed herbs**	0	2	4	6
Total	100	100	100	100
Nutrien contents				
Crude Protein (%)*	21.2	21.21	21.21	21.22
Metabolized energy (kcal/kg)*	3086	3086	3086	3086
Crude Fat (%)*	4.05	4.032	4.013	3.995
Crude Fiber (%)*	4.2	4.252	4.304	4.356
Ca (%)*	0.967	0.967	0.967	0.967
P (av) (%)*	0.5005	0.501	0.501	0.501
Lysin (%)*	0.7985	0.799	0.799	0.799
Methionin (%)*	0.545	0.545	0.545	0.545

Source: *) based on the calculation of table NRC (1994), **) The results of the analysis at the laboratory Animal Nutrition ad Feed Science Faculty of Animal Science Jenderal Soedirman University (2014).

Research was started from January 16th, 2015 until February 20th, 2015. The parameters measured in the study were cholesterol, glukosa and uric acid in blood broiler chicken. Blood cholesterol was determined by enzymatic activity of cholesterol oxidase para-amino phenazone (CHODPAP). Triglyceride was determined using enzymatic colorimetric method (DiaSys, Germany). Glucose and

Uric acid content calculation used calorimathic enzymatic test method with urease and PAP as catalist reaction indicators.

Statistical analyses:Data were analyzed using Nested ANOVA Completely Randomized Design (CRD). Any significant differences found between treatments, was then followed by the Honestly Significant Differences test (Steel and Torrie, 1994).

RESULTS AND DISCUSSION

Proximat analysis of mixed herbs consists of 12.29%protein, 3.08%lipid, 13.6%raw fiber, 62.51%BETN, 17681.34 ppm Fe mineral, 55.02 ppm Se mineral and 2242.92 ppm Zn mineral.

Blood, breast meat, leg meat and liver cholesterol profiles decreased following feeding with feed containing mixed herbs. Complete result was shown in Table 2.

Table 2. Average of blood, breast meat, leg meat and liver cholesterols of broiler

Treatment	Blood cholesterol (mg/dl)*	Glucose (mg/dl) ^{ns}	Uric Acid (mg/dl) ^{ns}
Control	111.80±17.02 ^a	174.60± 36.65	5.24±1.36
Mixed herbs 2% (w/w)	96.20±5.54 ^b	185.20±25.41	4.82±1.47
Mixed herbs 4% (w/w)	90.20±9.23 ^b	160.60±45.20	3.24±0.66
Mixed herbs 6 % (w/w)	89.20±12.76 ^b	177.00±38.40	3.98±1.07

Note ** = very significantly affected (P<0.01); * = significantly affected (P<0.05); ns = no significant. Different notation in the same column indicated significant different

Result of ANOVA indicated that the treatment very significantly affect (P<0.01) on the blood cholesterol, and however, the treatment did not significant affect (P>0.05) on blood glucose, and blood uric acids

Blood cholesterol content of broiler decreased 22.21% compared with average content of treatments were 89.20±12.76 mg/dl to 96.20±5.54 mg/dl and control was 111.80 to 17.02 mg/dl

Blood cholesterol content in this research was much lower than result from Murwani *et al.* (2011) which was 94.19 to 144.32 mg/dl. In addition, Paryad and Mahmoudi (2008) reported the blood cholesterol of broiler was 138.11 to 151.55 mg/dl, and 83.50 to 108.00 mg/dl in pullet (Bamidele and Adejumo, 2012).

Blood cholesterol of chicken fed with mixed herbs in their feed was lower thancontrol, and its reduction was in line with the content of mixed herbs in feed (Table 2.).It indicated that mixed herbs consumption up to 6% could serve as lipid and cholesterol contents reducer. Mixed herbs was used as feed additivewhich has bioactive substances that inhibits cholesterol development in two ways, those are 1) inhibits the rate of 3-hydroxy-3-methylglutaryle-Coenzyme A (HMG-CoA) reductase activity which is enzyme that inhibits lovasterol from feed lipid and metabolic lipid. As a results, the reduction in cholesterol development will occur, and 2) increase bile secretion which will bring cholesterol from intestine along with feces out from the body. The rest of cholesterol in tissues will be returned to liver and brought by HDL along with bile acid in intestine then will be released as excreted.

Roos and Katan (2000) mentioned that increasing bile acid will increase the cholesterol excretion resulting the reduction of cholesterol content in tissue.

Mixed herbs used in this study were moringa leaves, morinda fruit, curcuma tuber, turmeric tuber and garlic bulb. Morinda fruit contains digestive enzymes, vitamins and antibacterial substances such as antraquinone, acubin and alizarin, as well as nutrient such as protein (9.02%) and metabolic energy (3117 kcal/kg), xeronin and precursor xeronin. Proxeronin will be transformed to xeronin by proxeronase in the intestine, then absorbed by body cells to activate inactive proteins and control structure and shape of non-active cells. Scopoletin will bind serotonin which is a substance which usually cause narrowing the blood vessel and increasing blood pressure (Solomon, 1998). Scopoletin are found to have analgesic property and have an ability to control serotonin levels in the body and are responsible for most therapeutic activity of *M.citrifolia* (Wang and Su, 2001).

Glucose content in this research was 160.60 ± 45.20 to 185.20 ± 25.41 ml/dl, lower than result found by Bhatti *et al.* (2001) which was 231.82 ± 31.38 - 226 ± 15.20 ml/dl. Glucose in blood comes from feed carbohydrates metabolisms such as glucogenesis process and glycogenesis synthesis from liver and muscles glycogen, glucose regulation in blood through glycolysis, glycogenesis, glycogenolysis and gluconeogenesis.

Uric acid was synthesized in the liver and catalyzed by xanthine oxidase. Widhyari *et al.* (2009) reported the uric acid content in broiler was 8.33 to 9.0 mg/dl and Bhatti *et al.* (2001) was 4.16 ± 1.34 to 4.63 ± 1.88 mg/dl in normal chicken. Iriyanti *et al.* (2014) reported that in broiler fed with functional feed, the uric acid content was 4.53 to 7.67 mg/dl. Moringa leaves contains active substances such as saponin, tannin, flavonoid, alkaloid and terpenoid which could be used as antimicrobial substance (Bukar *et al.*, 2010; Fuglie, 2001; Kaloso *et al.*, 2010; Sato *et al.*, 2004; Cushine and Lamb, 2005). These active substances have mechanism in destructing bacterial cell membrane through increasing bacterial cell wall permeability and resulting lysis of bacterial cell (Esimone *et al.*, 2006).

Tannin works by binding to adhesion protein of bacterial cell which acts as bacterial surface receptor. The activity of tannin will reduce binding force and inhibit protein synthesis for cell wall development (Agnol *et al.*, 2003). Saponin is an active substance which produces lather in water as soap (Robinson, 1995). Saponin also has antibacterial activity. Saponin acts through destructing cytoplasm membrane which allow synergistic effect with tannin to destruct bacterial cell permeability. Saponin increases bacterial cell permeability and changes membrane function and structure resulting denaturation of protein membrane (Siswandono and Soekarjo, 1995). Saponin has molecule which is able to bind water and dissolves lipid, thus it could reduce cell surface and bacterial cell mortality.

CONCLUSION

The use of mixed herbs in broiler chicken's feed up to 6% could reduce to blood cholesterol level, but levels of the blood glucose and uric acid are relative same.

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OPTIMIZATION OF ORGANIC HERBS IN FEEDS AN EFFORT TO INCREASE BROILER CHICKENS PERFORMANCE

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ABSTRACT

This study aimed to optimize the function of organic herbs as an alternative to increase the productivity of broiler chickens, so as to overcome the problems of food, especially meat that is safe for consumption by the public. A hundred broiler chickens from PT Wonokoyo aged 1 day (DOC) were used in this research. Variables to be observed consisted of productivity of broiler chickens which include weight gain, feed consumption and feed conversion ratio. The method used in this research was experiment. Completely randomized design was used in this research. Treatment (P) of this study were: P0: feed without herbs, P1: feed with the addition of herbs ginger and turmeric, P2: feed with the addition of herbs turmeric, “*lemPUyang*”, vitamins, mixing ingredients and P3: feed with the addition of herbs: sand ginger, garlic, ginger, galangal, turmeric, ginger, betel leaf, cinnamon bark, vitamins, and mixing ingredients. Five replications were used in this research and every replication contained 5 chickens. Based on the results of the analysis, it could be concluded that the addition of organic herbs turned to be no significant effect on body weight gain, feed consumption and feed conversion ratio of broiler chickens. Suggestion for this research is that P3 treatment of herbal medicine should be given to determine a significant impact on the target specific organs although those treatments showed the same statistic number.

Keywords: *Feed, Organic herbs*

INTRODUCTION

Recently, the rearing of broiler chickens depends completely on the feed intake of prevention and treatment on diseases made by chemical ingredients. One positive side of giving the chemicals to broiler chickens into increase productivity and to maintain the health of broiler chickens. However, in contrast, there are many negative sides which are perceived incidentally by consumers. These chemicals, which are accumulated periodically in the chickens' body, will become a harmful residue to the consumers.

The emergence of the phenomenon of the insecurity of the broiler chicken meat attracts the consumers' attention and awareness to find alternatives to the consumption of the meat which is safe for body health. The meat must be free of factory-made chemicals which are totally harmful. Both feed intake and medicines for broiler chickens should always be safe from harmful chemicals in order to produce harmless and safe meat for consumption.

One helpful solution that can be done is by providing natural medicines for the broiler chickens to consume in the form of medical herbs that are definitely free from factory-made chemicals which are used comprehensively in the rearing of broiler chickens. However, it has been recognized that today, most of the medical herbs remain dangerous for human body because of the chemical use from the factory. Therefore, there should be a prominent separation between herbs with dangerous

chemical ingredients and natural herbs without anychemical ingredient known as organic herbs.

These organic herbs can serve as a substitute for vaccines or medicines to prevent and to treat broilers. Besides, it is also promising to increase the productivity of broiler chickens by means of increasing the broilers' appetite, increasing metabolism in their body, cleaning diseases in the digestive tract, stimulating hormones and enzymes as well as with a variety of other ways or performances.

Medical herbs constitute one of the local potentials which are easily found in many areas since they are planted deliberately by many people as wild plants that grow freely in the woods and they can be obtained at a cheap price. This will ensure the availability of natural medicines continuously with the effect of increasing the welfare of the people who seek for their life by supplying these medical herbs. If these medical herbs can function as an alternative for substituting the factory-made medicines, the level of dependence on medicines derived from foreign investment will no longer be found. As a result, food products can be derived from animal protein; especially broilers in which the availability is guaranteed in a sustainable manner with a stable and lower price for it can lessen production costs due to the use of organic herbs.

For the last decades, a lot of research has been conducted in order to obtain the optimal function of medical herbs. However, there are still many other kinds of medical herbs which have not been identified yet for the purpose of increasing the productivity or the health of the broiler chickens. For that reason, it seems to be an urgency to conduct a research about optimization of organic herbs in feed as an effort to increase broiler chickens' performance.”

The statement of the problem in this research was “Is there any effect of optimization of organic herbs in feed as an effort to increase broiler chickens' Performance?” and the purpose of this research was to analyze the effect of adding organic herbs in feed as an effort to increase broiler chickens' performance.

MATERIALS AND METHOD

This research used Quantitative method which was conducted in East Java Province. The data collection was done by experiment. Broiler chickens from PT Wonokoyo aged 1 day (DOC) as many as 100 were used in this research. Litter cage for chicken was used in this research.

Variables to be observed consisted of productivity of broiler chickens which included weight gain, feed consumption, and feed conversion ratio. This research used completely randomized design (CRD). Treatment (P) of this study were: P0: feed without herbs, P1: feed with the addition of herbs ginger and turmeric, P2: feed with the addition of herbs turmeric, “*lemduyang*”, vitamins, mixing ingredients and P3: feed with the addition of herbs: sand ginger, garlic, ginger, galangal, turmeric, ginger, betel leaf, cinnamon bark, vitamins, and mixing ingredients. Five replications were used in this research and every replication contained 5 chickens. The data was analyzed by analysis of variants.

RESULTS AND DISCUSSION

The result of the observation on the research about addition of organic herbs to the weight gain which has been done showed the average result P0 (21.80 g), P1 (19.00 g), P2 (21.16 g) and P3 (23.44 g). The result of variant analysis showed that the addition of organic herbs turned to be no significant effect ($P > 0.05$) on body weight gain of broiler chickens. This result indicated that there is no substantial difference between the treatment of herbs with or without control. These herbs have not shown any optimal potential yet from the substance of its ingredients to the target specific organs including the weight gain of broiler chickens. However, if it is seen from the average weight gain, it is shown that the herb is potential from the treatment P3 as an alternative herb that can be used because of the high average value.

Surprisingly, Saenab (2002) showed that giving herbs could increase the percentage of carcass on native chicken for about 64 %, with the weight gain on the body for 12 weeks as much as 1,065.5 g per head and 1,129.5 g per head. Herbs made from plain ingredients are helpful and effective to lessen the level of fatality on the chickens. If, in common, the fatality of the chicken reaches 4 heads, by means of the herbs, the fatality will only reach one head. Additionally, the use herbs can also lessen the smell of the chickens' feces, produce low fat meat, and give a pleasant color and a fresh aroma to the meat. A variety of these additional values practically opens up an opportunity to increase the income of the breeders. These herbs can be made and mixed from many ingredients that exist in our surrounding environment. The formulation consisted of sand ginger 1 kg, garlic 1 kg, ginger 0.5 kg, galangal 0.5 kg, turmeric 0.5 kg, *Curcuma zanthorrhiza* 0.5 kg, betel leaf 0.25 kg, and cinnamon bark 0.5 kg, a few leaves of *Phaleria macrocarpa*, M-Bio, and molasses (as source of energy). If molasses could not be found, it can be substituted by sugar.

A contradictory result is also shown by Hasbi and Sudirman (2012) in which the purpose of their research was to increase the productivity of native chicken by means of herbs as a feed supplement. The research was done using two treatments namely distinguishing between giving water (P0) and giving herbs (P1) to native chicken, by two replications, each replication consisted of 10 populations of chickens aged 6 weeks (grower phase), so that the total population was 20 chickens. Concentrate feed used was BP11-B. The results showed that giving herbal medicine with a dose of 30 cc L⁻¹ water contributed a real impact on chicken consumption, with an average of 1,976g, weight gain average was 464.5g, and feed conversion 4.3 kg to produce 1kg body weight, while the yield of feed consumption to the chickens without giving herbs reached 1,878g, an average weight gain reached only 362.5 g, and feed conversion reached 5.2 kg to produce 1kg of body weight.

The result of the observation on the impact of addition of the herbs to the feed consumption ratio which has been done before showed averagely P0 (41.73 g), P1 (45.49 g), P2 (46.12 g) and P3 (51.45 g). The result of variant analysis showed that the addition of organic herbs turned to be no significant effect ($P > 0.05$) on body weight gain of broiler chickens. This result indicated that there is no substantial difference between the treatment of herbs with or without control. These herbs have not shown any optimal potential yet from the substance of its ingredients to the target specific organs including the weight gain of broiler chickens. However, if it is seen from the average weight gain, it is shown that the herb is potential from the treatment P3 as an alternative herb that can be used because of the high average value.

According to Pramono (1994), galangal is beneficial as a booster for body warmth, painkillers and it also can increase the appetite. Sand ginger contains curcumin only about 0.006 percent; a low level of curcumin is incapable of affecting the optimal feed consumption of broilers. Galangal, when compared to other herbs, contains the lowest curcumin. Therefore, if we want to add galangal in feed consumption of the chickens, it becomes necessary to increase the dose in order that the production of broilers will be more optimal and it will also contribute a real effect on the consumption of broiler chickens. The result of the research done by Resnawati *et al.* (2002) found that the addition of galangal for about 0.02 percent up to 0.16 percent into the feed has no significant effect to the consumption of the feed containing energy for about 3130.5 kkal/kg and protein 20.33% given to the 5 week-old chicken.

In addition, Fais (2010) stated that basically alkaloids of garlic extract contain a toxin that is capable of inhibiting the growth of bacteria or causing the cell of the bacteria to experience lysis when it is exposed to the substance. He further explained that tannins inhibit proteolysis which functions to break down proteins into amino acids. It is then expected that the garlic should be able to increase appetite which will end up in the increasing of feed consumption.

Meanwhile, according to Nursal *et al.* (2006), ginger contains flavonoids, phenols, terpenoids. The benefit of the ginger is to stimulate the digestive glands. Likewise, galangal (*Alpinia galanga*) contains yellowish green essential oil that has distinctive smell. The taste is bitter and it makes the tongue cool. Galangal is beneficial to strengthen the stomach and bowels, improve digestion, and remove the mucus from the respiratory tract. Moreover, it can also treat headaches, chest pain, and increase appetite. Galangal seeds can also relieve colic or stomach pains, diarrhea, and nausea (Muhlizah, 1999). Both of these herbs should be able to improve the digestibility, which finally are able to increase the feed consumption.

The result of the observation on the impact of addition of the herbs to the feed consumption ratio which has been done showed averagely P0 (1.74), P1 (2.43), P2 (2.22) and P3 (2.21). The result of the variant analysis showed that the addition of organic herbs turned to be no significant effect ($P > 0.05$) on body weight gain of broiler chickens. This result indicated that there is no substantial difference between the treatment of herbs with or without control. These herbs have not shown any optimal potential yet from the substance of its ingredients to the target specific organs including feed conversion ratio of the broiler chickens.

According to Amrullah (2004), a good feed conversion ranges from 1.75 up to 2.00. Feed conversion rate is affected by three factors: the quality of feed, feeding techniques, and mortality. Feed conversion is closely related to the body weight gain; so the factors affecting the feed consumption and the body weight gain will also give a salient effect on the feed conversion. If different treatment ends up with the same body weight gain and feed consumption, it will produce the same feed conversion as well.

CONCLUSION AND SUGGESTIONS

Based on the results of the analysis, it could be concluded that the addition of organic herbs turned to be no significant effect on body weight gain, feed consumption, and feed conversion ratio of broiler chickens.

Suggestion for this research is that P3 treatment of herbal medicine should be given to determine a significant impact on the target specific organs although those treatments showed the same statistic number; and that further researches in relation to herbal medicine would be helpful to support this research.

ACKNOWLEDGEMENT

Special thanks to University of Muhammadiyah Malang for funding this research, University Farm for research place and all people who can't mention one by one.

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AN EXAMINATION ON PESTICIDE RESIDU IN RICE BRAN AND CORN FROM SEVERAL REGIONS IN EAST JAVA

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ABSTRACT

This study aimed at producing organic feed to overcome an issue of animal food safety, especially for broiler chicken consumed by society. In order to produce organic meat, organic feed which is free from synthetic chemical substances, including pesticide, is needed. An examination had been conducted on pesticide residue of rice bran and corn products from several regions in East Java, such as: Malang, Mojokerto, Bondowoso, and Sumenep. There were 10 types of pesticide being examined that were generated from three groups of organochlorines, organophosphates, and carbamate pesticides. The test was taken in PT. Angler BioChemLab, Surabaya, KAN accredited, ISO 17025:2005. The result showed that there was zero pesticide contamination for both rice bran and corn products; therefore, they are safe for animal consumption.

Keywords: *Pesticide, Rice bran, Corn, Organic*

INTRODUCTION

Broiler feed is made of several nutritious materials such as rice bran and corn as the energy resources for animal husbandry. Rice bran is the byproduct of rice grinding, making it possible to contain some pesticide during the rice cultivation period for pest and disease controls. Corn also experiences a phase where it needs some insecticide, allowing the crop to be freed from insects and diseases. Therefore, it is assumed that broiler has high potential to be contaminated by pesticide as the result of the contaminated food.

To produce organic broiler meat, chicken needs to be given organic food from organic farms. As stated in Indonesian National Standard (SNI) 6729: 2003, any kind of livestock product can be claimed as an organic product if 80% (based on its dry weight measurement) of its non-ruminant materials are derived from organic resource. Rice bran and corn contribute to the highest percentage of food for broiler feed which potentially has high pesticide contamination. It is necessary to select organic rice bran and corn from organic farm as well as to conduct pesticide residue test done by reliable national accredited laboratory to make sure that the rice bran and corn are free from pesticide residue and safe to be consumed.

MATERIALS AND METHODS

A survey on organic farming was conducted by internet observation on accredited operator data of rice and spices commodity. In addition, a direct visit had been done to several regions as the centers for organic farming in East Java : Jabung-Malang, Bondowoso, Mojokerto, Sumenep, and the LeSOS (Seloliman Organic Certification Institution) in Mojokerto. The samples of rice bran and corn

were taken from several regions that were appointed as the centers for organic farming in East Java; they were in Jabung-Malang, Bondowoso, Mojokerto, and Sumenep. Those areas were chosen since they fulfilled some criteria for organic farming such as: crop rotation, the use of high yielding varieties, the implementation of intercropping or poly-culture cultivation system, and zero chemical yet biological application to control plant pests. The pesticide residue examination in the rice bran and corn samples was conducted in the testing laboratory of PT. Angler BioChemLab, Surabaya. Data were analyzed descriptively.

RESULTS AND DISCUSSION

The main feed as energy resource in broiler feed consists of rice bran and corn taken from several areas of central organic farming producers in East Java. The organic farming criteria according to the Indonesian National Standard 6729: 2003 required an organic farming to have minimum external input materials without the usage of synthetic fertilizer and pesticide. Therefore, in the organic farming, the product is merely grown from and based on natural substances. It avoids and/or limits the use of synthetic chemical substance, such as chemical and factory made fertilizers, synthetic pesticide, herbicide, growth regulator substance, and some other additives in food material for livestock growing.

According to Kristianingrum (2009), pesticide is defined as any form of chemical substance and other materials as well as microorganism and virus utilized to control the spread of diseases. Pesticide is majorly used in farming industry for the purpose of pest and disease controls; they are utilized to kill or prevent plant diseases in order to maximize the harvesting product. The effect of pesticide excessive use can be classified into two categories, direct effect and indirect effect for food product being consumed by human and animals. The pesticide is detected in the form of pesticide residue in food materials from crops.

The Pest and Disease Control act by using pesticide is seen to be the most practical, economical and efficient; however, it gives negative effect in the form of pesticide residue, not to mention another effect of environmental pollution problem. The results of pesticide tests in rice bran and corn are presented in Table 1 and Table 2.

All the tested samples gave ND results which mean not detected, and that the pesticide level is below the RL (Reporting Limit) of 0.025 ppm. All rice bran and corn samples were declared to be safe for further broiler's consumption. As stated by Anonymous (2011), the negative effect of pesticide for human is the occurrence of acute poisoning that might occur from direct skin contamination, inhalation, mouth and alimentary tract and if it reaches certain level, it may cause death. Poisoning, apart from the contamination level, can also be determined by the different levels of pesticide toxicity between one to another formula. Chronic poisoning, such as in the case of carcinogenic, teratogenic, onkogenic, mutagenic, heart damage, kidney damage, and so on, can also occur when people consume certain pesticide contaminated products.

Cattle or pet poisoning may happen upon the direct use of cattle or pet pesticide to control the growth of ecto-parasites. In the less direct manner, poisoning occurs when pesticide is applied to control rodent by spraying or giving rodenticides with baits. Due to farmer ignorance, the baits are eaten by chicken, ducks, and other

livestock. The other indirect way is by spraying the pesticide to kill weeds which is also one of the cattle's feeds.

In this study, the types of pesticides being tested were DDT (*dichloro diphenyl trichloroethane*), lindane and heptachlor, classified into *organochlorines* (OC) group, CPM (*chlorpyrifos metil*) and diazinon, grouped into *organophosphates* (OP), as well as carbofuran that belongs to carbamate type. The underpinning test towards pesticide is based on the research conducted by Indraningsih and Sani (2004), stating that the prohibition to use OC type in farming industry triggers the use of OP and carbamate pesticide types to control pests and diseases. The most widely use organophosphates (OP) pesticide in Indonesia are diazinon and CPM where we can detect their residue on soil or land, agricultural waste and livestock products. From the carbamate group, carbofuran is the highest in usage frequency treated as herbicide.

Organochlorines is an illegal type of pesticide determined under the Decree of Agriculture Minister No. 434.1/ktps/TP.270/7/2001, Article 6. This clause considers the danger of OC's long unraveling period which has stable and non-decomposed character in soil. Indraningsih and Widiastuti (1998) mention that the soil absorption of pesticide residue is influenced by several factors, such as: the ability of pesticide absorption by the soil particles, the existence of rain water to wash the soil, the soil water evaporation, the ability of soil degradation by microorganism, as well as decomposition by means of physicochemical and sunrays. In addition, the characteristics of each pesticide play their role in determining the length of their decomposition. For example, organochlorine has high resistance towards water and difficult to evaporate; therefore, it is hard to be degraded.

“Total DDT” is commonly used to refer to the sum of all components; DDT, DDE, and DDD (*dichlorodiphenyldichloroethane*). DDT characteristics are: (1) non-degraded towards photolysis, both biologically or chemically, (2) containing halogen, commonly chlorine, (3) very low solubility, (4) high solubility towards fat, (5) semi volatile, (6) can be moved by the wind over long distances in the air, (7) biocumulative, (8) biomagnificative (toxicity increases along the food chain).

As one of the persistent chemical compounds, DDT is difficult to be degraded into simpler compounds. When DDT enters the food chain, it reaches eight years of decomposed period, which means, one time consumption of DDT needs eight years of degrading and decomposing time allotment. When it is given to animal, DDT will be accumulated in fat tissues and inside the liver.

The existence of 200 ppm DDT in the broiler body will cause liver swelling. *DichlorodiphenylDichloro Ethylene* (DDE), as a form of DDT's metabolite as much as 20-30 ppm will trigger eggshell thinning for 15-25% and it would also decrease eggs' hatchability. The symptoms of acute poisoning in chicken due to DDT contamination may result in chicken paralysis, tremor, convulsive seizures, and staggering. In the chronic phase, there will be heart and adrenal gland malfunctioning. It also gives carcinogenic, teratogenic, mutagenic, and immune-suppressive effects on chicken (Indraningsih and Widiastuti, 1998). The symptoms of acute poisoning in human are tremor, headache, drowsiness, and vomiting. The chronic poisoning effects as a result of DDT contamination are in the form of liver cell destruction, kidney problem, neurodegenerative system disorder, and issues in immune and reproduction systems. The chronic poisoning effects for poultry can be clearly observed in eggshell thinning and demasculinisation.

The study report proposed by Mutiatikum and Sukmayati (2010) affirms that some amount of organochlorine pesticide was found in rice product produced in Surabaya; the substances were lindane compounds (0.0075 mg/kg), heptachlor (0.0349 mg/kg), and from carbamate group in the form of Carbofuran (0.02 mg/kg). Although they were detected in small amount, organochlorine type of pesticide should be given more attention, especially by its BMR since it has long decomposed lifespan, possesses stable characteristic inside the soil also in very long time.

The types of pesticide included in the organophosphates family are: *Azinophosmethyl, Chloryfos, Demeton Methyl, Dichlorovos, Dimethoat, Disulfoton, Ethion, Palathion, Malathion, Parathion, Diazinone, Chlorpyrifos*. The last two substances have been tested in the organic farming for rice bran and corn plants. The test results are negative; both samples are safe and free from diazinon or *chlorpyrifosmetil* (CPM). Therefore, they can be safely consumed as broilers' staple feed.

For human, organophosphate disrupts the work of cholinesterase enzyme which functions to break acetylcholine into choline and acetic acid. Acetylcholine is secreted by the tips of nerve to the other following nerve system, and then being processed inside *Central Nervous System (CNS)*, which later on initiates certain movement coordinated by human brain. If human body is repeatedly exposed to the organophosphates in the long run human mechanism, the work of cholinesterase enzyme would be interfered, resulting in severe nerve system disorder. Organophosphate pesticide enters human body by means of digestion organs or digestion, respiratory tract or inhalation and through the exposed skin surface or penetration (Priyanto, 2009).

Priyanto (2009) further explains, organophosphate poisoning commonly occurs on farmers as the result of their reckless usage without obeying the indicative measurement given by the producers in order to safely control plant diseases; they mostly prefer to use cover blanket system that is to give or spray pesticide in any condition both in the pests' absence and presence. The uncontrolled usage of pesticide is likely to impact the farmer's health and environment in general.

Organophosphates and Carbamate pesticides may trigger acute poisoning with the symptoms as follows: neck choking feeling, dizziness, weak feeling, staggering, pupil or iris narrowing, blurred vision, tremor, sometimes muscle spasm, restlessness and decreased consciousness, nausea feeling, vomiting, stomach spasm, diarrhea, excessive sweat secretion, tightness and a sense of fullness in chest, flu, cough with phlegm, and excessive salivating. The cause of the symptoms can only be detected after 12 hours resulting in the slowing down of heart beat and inability to control urinating or bowel moving.

Astuti and Juliawati (2009) mention that organophosphate and carbamate inhibit the work of acetyl cholinesterase enzyme (AChE) resulting in the accumulation of acetylcholine (ACh). Acetylcholine accumulated in the Central Nervous System (CNS) would induce tremor, in-coordination, convulsive seizures, and so on. The affinity power of this insecticide should be able to bind AChE enzyme so that acetylcholine, that is supposed to transfer stimulation impulses from pre to post synapses (*neurotransmitter*), works twice harder because it cannot be diverged by AChE enzyme. Several kinds of organophosphates are soluble in water, causing systemic poisoning for insects and mammals. This group has more stable inhibitor creating more dangerous effect on living creatures, whereas carbamate has

less stable or freer (reversible) inhibitor making it less harmful to creatures and environment.

Kristianingrum (2009) confirms that BMR (Residue Minimum Limit) is the minimum allowed amount of residue in any marketed food materials, which is stated in mg/kg of food materials (bpj, ppm). According to Atmawidjaja (1986, in Kristianingrum, 2009), the residue minimum limit for Diazinon in corn should be 0.1 mg/kg. In this study, the researcher did not find any amount of Diazinon in the whole rice bran and corn samples.

Mutiatikum and Sukmayati (2009) assert that the amount of pesticide in rice product is still high in percentage and the pesticide residue in the farming products available in Indonesian National Standard Program (RSNI 2) still adopts Codex Alimentaris Commission (CAC). Less regulation is applied for pesticide BMR in the rice planting time. Rice BMR has to be low, because rice is a staple food consumed by human and animals in very large amount. Pesticide in rice product will be accumulated in a long period of time, causing some health problems. Rice is the source of carbohydrate, and the average per-capita daily consumptions based on its food group in 2002 were: for West Java (278.88 gram), Central Java (223.77 gram), and East Java (233.51 gram), thus, the average consumption was 245.39 gram.

Further, it is explained that apart from BMR, ADI (Acceptable Daily Intake) has to be considered. ADI is the estimated number of compound types of pesticides in foods that if it is consumed everyday for the rest of human life, it will not contribute any bad effect or health risk. Carbofuran detected in rice is 0.029-0.0755 mg/kg; it is considered low in amount, because its BMR is 0.2 mg/kg, and its ADI is 0.01 mg/kg/day. The calculation result of BMR in accordance with the daily average rice consumption in Indonesia shows the number of 0.02 mg/kg; however, since rice is the main commodity consumed in Indonesia, the BMR ratio should thoroughly be re-investigated more. It needs to be adjusted to the rice consumption pattern for Indonesians. BMR re-calculation result suitable for Indonesian rice consumption pattern per-capita and per-day is 245.38 gram smaller than that of the standard BMR.

CONCLUSION

There is no pesticide residue detected on rice bran and corn plantation products from several organic farming centers in East Java, such as: Jabung-Malang, Bondowoso, Mojokerto, and Sumenep.

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THE TREND OF CORN AVAILABILITY AS POULTRY FEED IN EAST JAVA

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ABSTRACT

Feed is one of the important components in poultry industry. The main problem to feed energy sources, especially corn, is continuity of availability, especially during the dry season, in addition to varying product quality. Comprehensive, macro, and sustainable problem solving is absolutely necessary to overcome the problems of corn in Indonesia, especially in East Java. In order that the attempt is successful, it would require the availability of corn as feed ingredient trend in East Java. This trend will be useful to look at the condition of corn as feed ingredients in East Java during the past few years. The results of these trends can also be extrapolated to predict the trend of corn production in the future in East Java. Furthermore, by the results of these trends it can be planned development of poultry feed ingredients (corn) better. This study specifically aimed at looking at the trend of the availability of feed corn in the area of East Java. Formulation of the problem is how the trend of corn is feed availability in East Java. The method used in this study was a survey and study of literature. The research approach was a quantitative approach. This study was conducted in the province of East Java. The data used in this study were primary and secondary data. Informants in this study were those in the official agencies in the city/regency, while the key informants were officials in Animal Husbandry Department in East Java, which could provide information related to the problem. The determination of informants was done purposively with the criteria of the competent authorities with the data of farms. Furthermore, the taking of informants was done by employing snowball technique. The data were collected through observation, in-depth interview, and expert discussions. The required data included quantitative and qualitative data. Data analysis employed correlation regression analysis and then was extrapolated to see the trend of the potential availability of corn in the future in East Java. The conclusion was the trend of the availability of corn as poultry feed in East Java was experiencing a rise from year to year with a tendency to rise linearly. East Java provincial government is suggested to strive harder to increase the production of corn for poultry feed because there was a tendency in recent years for a decline in production

Keywords: *Corn, Feed, Poultry*

INTRODUCTION

Feed is one of the important components in poultry industry. The imported poultry feed ingredients is the cause for the annihilation in poultry industry because the budget sets for this feed reaches 70% for broilers and 90% for laying hens.

The attempt to fulfill either vegetable protein or animal protein of feed ingredients still becomes a major problem. Soybean meal which is one of the main components for poultry feed cannot be optimally produced in Indonesia because soybean is categorized as a subtropical plant. Furthermore, soybean production is mainly aimed for human. To meet the demand, our government needs to import soybeans. Likewise, the necessity for fish flour as feed mixture can only be fulfilled by our government by importing it.

Corn, rice bran, cassava, and oil as the energy source feed ingredients can be in long term fulfilled by our government. In the contrary, our government needs to import feed material as the source of protein. The main issue of feed energy sources, especially corn, is not only on its availability continuity but also its various product quality. That is not the only issue our government needs to deal with since after crop treatment, for example the drying process and storing process somehow create another problem since they have not been seriously solved. Meanwhile, those are the main reasons of the rarity of corn during dry season as well as the cause of quality variations.

A comprehensive, macro, and sustainable solution is of urgency to overcome the problems of corn in Indonesia, especially in East Java. In order that the attempt is successful, it would require the availability of corn as feed ingredient trend in East Java. This trend will be useful to look at the condition of corn as feed ingredients in East Java during the past few years. The results of these trends can also be extrapolated to predict the trend of corn production in the future in East Java. Furthermore, by the results of these trends it can be planned development of poultry feed ingredients (corn) better. This study specifically aimed at looking at the trend of the availability of feed corn in the area of East Java.

RESEARCH METHODOLOGY

The methodology used in this research was survey method and literature study. The research approach used was quantitative approach. This research was supported by the documents related to poultry feed ingredients and the result of this research would be used for formulating the trend.

This research was conducted in East Java Province by considering that in East Java there is a potential corn that can be used for feed poultry. This corn has not been optimally treated and needs specific mapping effort in order to elevate the feed security which later on can develop into the food security.

The data used in this research were primary and secondary data. Primary data covered the variables in transferring model mapping of corn as poultry feed which was seen from geographical dimension such as the corn potential as poultry feed ingredients, the potential of poultry livestock, and the necessity of corn as poultry feed ingredients. Those variables will help to know the effort to optimize and to increase corn production as poultry feed in East Java to elevate the feed security which later on can develop into the food security. Meanwhile, the secondary data were gained from data documentation about corn from related institutions, such as Animal Husbandry Institutions in city/district in East Java or Animal Husbandry Institution in East Java Province.

Informants in this study were those in the official agencies in the city/regency, while the key informants were officials in Animal Husbandry Department in East Java, which could provide information related to the problem. The determination of informants was done purposively with the criteria of the competent authorities with the data of farms. Furthermore, the taking of informants was done by employing snowball technique.

Data collection was done by doing observation, in-depth interview, and discussion among experts. The required data were qualitative and quantitative data. The qualitative data were mainly used to investigate the corn potential as poultry

feed ingredients in city/district in East Java, while qualitative data were used to investigate the availability range of poultry feed ingredients in every area.

The data were analyzed by using correlation regression analysis which later were extrapolated to detect the trend of the potential availability of corn in the future in East Java. The next phase that could be done after doing some previous phases was analyzing the data using the relevant theory, interpreting them, and concluding the data based on the problems.

FINDINGS AND DISCUSSION

Based on the data from Central Agency on Statistics (BPS), it was found out that there were various fluctuations in corn production every year for the past six years. The data taken from 2008 to 2013 can be seen in the table 4.1 below:

Table 4.1. The Corn Production in East Java Province

No	Year	Production (Ton)
1	2008	5053107
2	2009	5266720
3	2010	5587318
4	2011	5443705
5	2012	6295301
6	2013	5741833

Source: BPS 2014

Based on the above table, it can be seen that corn fluctuation production is not irregular. At first, the production tended to stably increase with a positive trend. However, there was a confusing situation for all parties because of the irregular production mainly in 2010, 2100, 2012, and 2013. In the last mentioned year, the production tended to decrease. The observers of agriculture production attempted to find the answer to this problem. Some of them blamed on the government policies. While the others arraigned on the global economy condition, such as global warming and so forth. Therefore, the possible alternative to overcome this problem was by doing a simple linier regression analysis as can be seen in this equation:

$$Y = 4,926089 + 182450.286 X$$

Where: correlation coefficient(r) = 0.791 and determination correlation is (r^2) = 0.626

Based on that equation, the tendency of increasing corn production every year can be clearly seen. Likewise, there was a strong correlation between year and corn production as much as 79.1% which showed that there was indeed a strong correlation. Meanwhile, the effect of year on corn production was pictured in determination coefficient value as much as 62.6% which then can be concluded that the year played a relatively great role in corn production. Yet, we need to assure whether we need to test this linier regression analysis. The suitable test for it is Test F.

The test result showed that there was a visible, and thus the linier regression can be applied. The tendency of corn production rising in East Java cannot be separated from the attempt of every party to increase the sustainability of corn

production. The policies issued by the government facilitated the corn plantation. Corn farmers were enthusiastic to plant corn because of its promising prospects. In other areas, corn sellers, suppliers, field elucidators and other related parties experienced the same excitement as corn farmers.

This aligns with the statement of East Java governance which declares that East Java is well known for its national granary, including corn which has a surplus as much as 3.7 million tons. Since the corn production in East Java is always over production every year, the production can be exported to some areas outside of East Java that needs approximately 3.7 millions of corn per year. The corn consumption of East Java inhabitant is merely 2.5 million tons; to emphasize, the surplus is 3.7 million tons. (www.jatimprov.go.id, 2014).

East Java Province is renowned for its name as a national granary, especially corn. This is proven by the corn production in East Java in 2012 that reached 6.30 million tons. The dry corn maize increased as much as 0.85 million tons or 15.64 percents. It was certainly different from the corn production in 2011 which was only as much as 5.44 million tons of dry corn maize based on the calculation of Fixed Value (ATAP). The corn production dramatically increased in 2012 or 30 percents more than that of national corn production contribution which last year reached as much as 19.38 million tons. The raising of corn production in East Java in 2012 happened because there was an increase in the harvested area of 28.46 thousand hectares or 2.36 percents. The level of productivity also increased as much as 5.87 quintals/hectares or 12.98 percents. There were five districts in East Java that dominantly produced corn in 2012. However, there were some changes compared to 2011. Last year, Bangkalan belonged to the best five ranks in producing corn; but it is now replaced by Lamongan. The big five districts of which areas were dominantly wide in 2012 were: Sumenep (142.13 thousand hectares or 11.53 percents), Tuban (92.44 thousand hectares or 7.50 percents), Sampang (84.24 thousand hectares or 6.83 percents), Probolinggo (70.50 thousand hectares or 5.72 percents), and Lamongan Regency (59.54 thousand hectares or 4.83 percents).

The prediction of decreasing corn production in 2014 made the poultry feed businessman predict the necessity of imported corn as much as 3.6 million tons. The imported corn volume would raise as much as 500,000 tons. In 2013, Indonesia imported corn as much as 3.1 million tons. It shows that the necessity of imported corn for poultry feed industry will elevate every year. In 2012, the imported corn realization was only as much as 1.7 million tons. The volume projection of imported corn itself refers to the production capacity of 65 poultry feed domestic industries and the estimation of corn production that was released by Central Agency on Statistics BPS. The contribution of corn for poultry feed components is 50-55%. Despite the target set by the government that corn production this year is as much as 20.82 million tons, it increases as much as 12.48% compared to that of 2013 production. Unfortunately, the target cannot be required considering the fact that corn production every year tends to decrease. In 2013, national corn production decreased as much as 880,000 tons or 4.52%. The need of corn for poultry feed industry in 2014 would reach 7.5 million tons (www.kabarbisnis.com, 2014).

In attempt to elevate corn production, Madura is likely to supply corn as much as 1.8 million tons for national poultry feed material. It is supported by dry field and the level of land fertility which play an important role in corn plantation. The changes of global climate benefits corn farmers since it can increase the productivity

of corn. The majority of corns are planted in such dry lands. Our government identifies that the potential land for corn plantation in Madura reaches as much as 300,000 tons. If 1 hectare (ha) land can produce 5 tons of corn, the harvested corn will be as much as 1.5 million tons. That assumption will significantly grow bigger if the productivity level per hectare is as much as 6 tons, so that the potential production that can be gained is as much as 1.8 million tons. The chance for corn supply in Madura for poultry feed material industry has grown up. Unfortunately, it was not supported by well-built infrastructures; as a consequence, the distribution cannot yet be optimized.

The need of corn for poultry feed industry is 7.5 million tons/year. Our corn farmers are supposed to meet the number because the corn production is over 19 million tons. Coming to our surprise, The Association of Poultry Feed Industries (GPMT) claimed that we need to import corn for poultry feed because of low corn quality and availability. Only in 2013, GPMT imported corn as much as 2.8 million tons. Meanwhile, we imported corn as much as 1.7 million tons last year.

The corn production in East Java contributed 40% to total national production within 1.3 million ha farming areas. Among those farming areas, 75% is a dry land. The data from *Directorate General of Food Crops stated that the farming realization for planting season (MT) in 2012/2013 (October 2012-April 2013) reached 2.87 million ha. It was dramatically decreasing as much as 239,000 ha (8.35%) compared to that of the same period last year. If the farming targets between MT 2012/2013 and MT 2013 in the same period are compared, the demand of corn as much as 349,846 Ha (only 89.13%) cannot be fulfilled. Only in October-November that the farming target was fulfilled: October was 102.6% and November was 108.1%. Meanwhile, in December-April, the target could not yet be fulfilled. (www.kabarbisnis.com, 2014).*

The national corn need for poultry feed was 7.5 million tons. Meanwhile, the corn production in East Java was about 6 million tons. There were around 2.5 million tons for the human food need; while the rest of them (3.5 million tons) were utilized for poultry feed. It showed that East Java contributed around 55% of the need of national poultry feed.

CONCLUSION

The trend of corn availability as poultry feed in East Java is increasing from year to year linearly.

SUGGESTIONS

The government of East Java Province must work harder to escalate corn production for poultry feed because there is a decreasing tendency in the last year of production.

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THE EFFECT OF DIETARY VITAMIN E AND ZINC (Zn) LEVELS ON PERFORMANCE OF LAYING DUCKS

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ABSTRACT

Vitamin E and Zn are necessary for preventing free radical damage to phospholipid membranes, enzymes and other important molecules. The objective of this study was to evaluate the effect of dietary vitamin E and Zn levels on performance of laying duck egg. A total of 90 ducks of 22 weeks old were randomly divided into 15 experimental units by assigning a completely randomized design with 5 treatments and 3 replications. The treatment diets were R1(control diet), R2(R1+40 IU vitamin E), R3(R1+80 IU vitamin E), R4 (R1+100 ppm Zn Organic), and R5(R1+200 ppm ZnOrganic). Ninety duck eggs used in this study. The experiment was carried out for 8 wk. Parameters observed were egg production, feed conversion ratio, egg weight and consumption. The results showed that increasing of vitamin E from 40 IU to 80 IU and Zn from 100 ppm to 200 ppm did not effect the consumption and egg weight. Otherwise, the dietary 80 IU Vitamin E and 100 ppm organic Zn decreased significantly feed conversion ratio and increased significantly egg production. It concluded that dietary of 80 IU vitamin E or 100 ppm organic Zn could increase the performance of laying ducks

Key words: duck eggs, performance, organic Zn, vitamin E,

INTRODUCTION

Duck egg production in Indonesia was lower and fluctuated than laying egg production. It was 297.074 tonnes, while egg production of laying hens was 1.299.199 tons in the year of 2014 (BPS 2015). According to Ismoyowati & Suswoyo (2011), egg production of conventional-reared laying ducks was only about 26.9%-41.3%. However, egg production can be improved by improving dietary quality and increasing nutritional availabilities.

Palm oil and fish oil are used as source of omega-3 and omega-6 fatty acids. Eicosapentaenoic acid (EPA) and arachidonic acid (AA) are precursors of prostaglandins that have an important role in several aspects of reproduction, such as ovulation, estrus and embryo survival (Nava *et al.*, 2011). Eicosapentaenoic acid (EPA) and arachidonic acid (AA) are formed from ω -3 and ω -6 fatty acid. The use of omega-3 and omega-6 fatty acids on laying ducks ration can be combined with Zn and vitamin E supplementation. The presence of Zn is very beneficial to the metabolic transformation of ω -3 and ω -6 fatty acid to prostaglandins that play a role in the reproductive system (Bhowmik *et al.*, 2010). Meanwhile, Vitamin E is fat soluble and as antioxidants has a role for breaking the chain of peroxide in membranes and protecting PUFAS (Poly Unsaturated Fatty Acids) from oxidation (Lamid, 1995). The objective of this study was to evaluate the effect of dietary organic Zn and vitamin E on performance of laying duck.

MATERIALS AND METHODS

Ninety laying duck of 20 weeks old used in this study were randomly divided into five treatments. This study used a completely randomized design with 5

treatments and 3 replications. The diets were isocaloric (2850 kcal ME / kg) and isoprotein (16%) (Leeson and Summers, 2005). The composition and nutrients content in the experimental diets is presented in Table 1.

The treatment diets were R1(control diet), R2(R1+40 IU vitamin E), R3(R1+80 IU vitamin E), R4 (R1+100 ppm Organic Zn), and R5(R1+200 ppm Organic Zn). The ducks were given adaptation period for 2 weeks at the age of 20-22 weeks to introduce the treatment diet. The Pattern of diet adaptation was 75% of commercial ration (CR) and 25% treatment diet (TD) during 4 days, 50% CR and 50% TD during 4 days, 25% CR and 75% TD during 3 days and 0% CR and 100% TD for the last 3 days. Treatment diets were fed for 8 weeks at 22-30 weeks of age and the water was given *ad libitum* each day. Egg weight and egg production were recorded daily, feed intake and feed conversion ratio were recorded weekly. Egg production performance was expressed as a percentage of egg production. Feed intake was recorded on a weekly basis.

The data were subjected to analysis of variance, and when significant effect at treatment was found, Duncan’s Multiple Range Test was used to determine the significant difference among mean values.

Table 1. The composition and nutrients content of control diet (R1), as fed

Feed Ingredient	(%)
Yellow corn	46
Rice bran	15.2
Soybean meal	19
Fish meal	8
Palm oil	3.5
Fish oil	1
CaCO ₃	6.5
NaCl	0.2
Premix	0.5
DL-Methionin	0.1
Total	100
Nutrient content *	
Crude protein (%)	15.9
Crude fiber (%)	10.3
Crude fat (%)	3.52
Ash (%)	12.5
Gross energy (kcal /kg)	3788

* Analysis at Laboratory of Nutrition and Feed Technology ,Faculty of Animal Science ,Bogor Agriculture University, 2014

RESULTS AND DISCUSSION

The performance of laying duck is summarized in Table 2. There were no significant differences in feed consumption and egg weight. However, egg production and feed conversion ratio were affected by dietary treatments.

Table 2. Average of feed intake, egg weight and feed conversion ratio of laying ducks

Parameters	Treatments				
	R1	R2	R3	R4	R5
Feed Intake(g/duck/day)	167,18±1,59	167,96±3,08	170,56±1,64	168,87±2,58	166,92±3,94
Egg Weight (g/egg)	71,17±2,42	69,54±1,77	68,77±2,55	69,71±4,21	71,61±2,60
Feed Conversion Ratio	8,20 ± 0.34 ^a	6,66 ± 0.72 ^b	6,96 ± 0.64 ^b	6,85 ± 0.43 ^b	6,99 ± 0.66 ^b

Notes: mean in the same raw with different superscripts differ significantly (P <0.05)

R1(diet without Vitamin E and organic Zn), R2(R1+40 IU vitamin E), R3(R1+80 IU vitamin E), R4 (R1+100 ppm Organic Zn), and R5(R1+200 ppm Organic Zn).

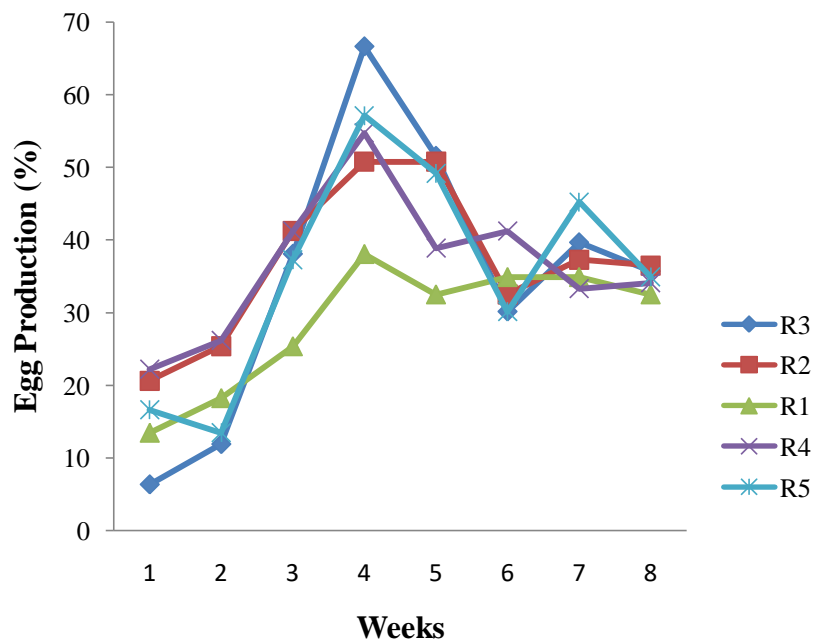
Based on Table 2, the average feed intake of ducks ranged from 166.9 to 167.98g / duck/ day and the treatment did not effect the feed intake. This was probably due to the shape feed similarity and the level of oil in diets which resulted equal palatability. Ferket and Gernat (2006) stated that feed intake was influenced by the shape of rations, color and ration palatability. Darmwan *et al.*, (2013) reported that diets without oil decreased feed intake significantly because of dusty texture. Feed intake of the study was smaller than managed by the farmers in general, which is about 160 to 200 g/duck/day (Sasongko, 2010). Chen and Shen (1989) reported that Tsaiya, Taiwan local ducks with the body size was almost the similar to Bantul duck, showed the results of feed intake ranged from 205 to 225 g/duck /day with feed on mash form. This condition was probably due to the high ambient temperature. The temperature in the house was 33,3-36,7 °C by day and it was higher than the recommended thermo neutral zone of 18-24°C for poultry in the tropical regions (Holik, 2009). Wilson *et al.* (1980) stated that ideal temperature for ducks and laying hens between 18.3 to 25.5°C and feed intake, production and quality of eggs greatly reduced at 30-32°C. According to Scoot and Dean (1991) that the feed intake decreased whereas the water consumption increase at temperature 32-35°C.

Egg weight of the study ranged from 69.54-71.61 g / egg. It was greater than the finding of Darmawan *et al.* (2013) who reported that egg weight of ducks was 56.60 g with 22,89 g/duck/day protein consumption. The difference was possibly due to the high protein consumption. Protein consumption in the present study was about 26.75 g/bird/day. Ketaren & Prasetyo (2002) reported that protein consumption on 20-43 wk of age was 26.65 g/bird/d. According to Leeson and Summers (2005), protein and/or amino acids (especially methionine) are the most nutrients that have important role in controlling the size of eggs. Fish oil in the diet did not affect egg size but affect the composition of the fat content in the yolk (Suripta and Astuti, 2006). In another study, Bozkurt *et al.* (2009) showed that essential oil supplementation in diet did not affect egg weight of broiler breeders.

Feed conversion resulting from this research is 6.66-8.20 (Table 2). The feed conversion is bigger than reported by Zubaidah (2001) that feed conversion for

laying duck on 28 wk of age was 5.55 - 6.70. Feed conversion in this study also bigger than reported by Darmawan et al., (2013) that using balance of omega-3 and omega-6 = 1: 4.5 and 200ppm organic Zn. Addition of Vitamin E and Organic Zn decreased significantly ($P < 0.05$) feed conversion ratio. It was caused by the increasing of egg production, while feed intakes were not significantly different. According to Leeson and Summers (2005), factors that affect feed conversion are eggs production, energy and nutrient content of the feed, body weight, and temperature.

Picture 1. Percentage of Egg Production



R1(diet without Vitamin E and organic Zn), R2(R1+40 IU vitamin E), R3(R1+80 IU vitamin E), R4 (R1+100 ppm Organic Zn), and R5(R1+200 ppm Organic Zn).

The treatments increased Egg production of laying ducks during eight weeks (picture 1). Addition of vitamin E and organic Zn increased significantly ($P < 0.05$) egg production. According to Ashok and Sushil (2005), the relationship of vitamin E with reproduction is the role of vitamin E as an antioxidant that can prevent the fat oxidation. Vitamin E is fat soluble and as antioxidants has a role for breaking the chain of peroxide in membranes and protecting PUFAS (Poly Unsaturated Fatty Acids) from oxidation (Lamid, 1995). Use of 100 ppm to 200 ppm organic Zn was also able to increase the eggs production. The presence of Zn is very beneficial to the metabolic transformation of ω -3 and ω -6 fatty acid to prostaglandins that play a role in the reproductive system (Bhowmik *et al.*, 2010). Prostaglandins increase the secretion of follicle stimulating hormone (FSH) and luteinizing hormone (LH) from the anterior pituitary. The follicle stimulating hormone will stimulate the follicle growth, while the luteinizing hormone will stimulate the ovulation process (Yuwanta, 2004). Bachri *et al.* (2006) reported that the used of 112.5 mg Zn increased egg production significantly because the Zn mineral can activate amino peptidase and carboxypeptidase enzymes and provide sufficient amino acids for the

formation of the egg. Darmawan et al.,(2013) reported that diets containing the ratio of ω -3 : ω -6 = 1 : 3 and 200 ppm organic Zn produced highest egg production significantly ($P < 0.05$).

CONCLUSION

Addition of 100 ppm Zn or 80 IU vitamin E in the diet can increase the performance of laying duck on 22-30 wk of age.

ACKNOWLEDGEMENT

This research was funded by Indonesian Ministry of Education and Culture, Directorate General of Higher Education with contract number: 77 /IT3.11/LT/2014.

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THE EFFECTS OF LENGTH OF FEEDING AND LEVEL OF CRUDE FIBER ON CARCASS QUALITY AND SERUM CHOLESTEROL OF BROILER CHICKEN

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ABSTRACT

The objective of this study was to obtain the effect of length of feeding (LF) and level of crude fiber (LCF) on carcass quality of broiler. This research was used Completely Randomized Design (CRD), Factorial pattern 4 x 3. Factor A were 4 LF of feeds, namely: A₁ (4 days); A₂ (6 days); A₃ (8 days); and A₄ (16 days). Factor B were 3 LCF of feeds, namely: B₁ (5%); B₂ (7.5%); and B₃ (10%). Each treatment was repeated 2 times. Three weeks of age of 120 broiler chickens were used in this research. Variables which measured were carcass quality as weight of carcass, abdominal fat weight, and serum cholesterol. The experiment data were analyzed using ANOVA, and HSD test was then performed to showed the difference between treatments. The analysis of variance showed that the treatment has not gave an interaction between factor A and B (P>0.01) on carcass weight, and abdominal fat weight (P<0.05). The treatment no affected for serum cholesterol. The treatment as length of feeding (Factor A) has effects significantly (P<0.05) on carcass weight, and abdominal fat weight (P<0.05). Combination of length of feeding and level of crude fiber caused not affected in carcass weight, abdominal fat, and serum cholesterol. However length of feeding tend to decreasing carcass weight and abdominal fat content.

Keywords: *Feed restriction, Carcass quality.*

INTRODUCTION

Development of livestock production, especially meat consumption pattern seen from the people of Indonesia experienced the dilemma, on one hand meat consumption per capita is still low, on the other hand there is a certain tendency of consumers to limit consumption of poultry meat due to fat content which is considered a negative effect on food quality (cholesterolphobia). Recommendations on food to live healthy is starting lively populous in various developed countries in the world and even the world organization board such as WHO, has conveyed a message for people to consume less fat and cholesterol, and consume a lot of starch and fiber. The issue is certainly a very big challenge for the development of livestock production in the century now and the future. Similarly, due to bird flu outbreaks also affect the consumption of chicken meat, especially because there are allegations of the population of bacteria, especially pathogenic microorganisms such as salmonella in the digestive organs can be contaminated in chicken meat during the cutting process. This issue is certainly a challenge for expert animal husbandry in order to develop a leaner supply products in order to produce cattle with low fat content carcass but have high edible portion of meat as a source of safety food for consumers. Compensatory growth is genetic potential of livestock as part of manivestasi hyperplasia and hypertrophy cell at a particular phase or period of

growth with environmental factors. Feed restriction or limitation of feed intake can be applied in the livestock business in a period / phase specific pertubuhan for two purposes of improving feed efficiency and meat quality. Feed consumption restriction has dimension offeed intake reduction (reduction of total consumption) and reduction eather quantityor qualityof feed. Time and duration of feed restriction both in quantity and quality will affect the response of livestock through compensation of its growth. If the concept application is made in accordance with the of animal growing period which fed both quantity and quality that fit with the needs of animal, the compensation growth will give a positive response to both feed efficiency and carcass quality as well as to the characteristics of digestive organs.

Meat chickens are a very strategic commodity livestock for food security including animal protein needs of the Indonesian people because it has several advantages over other meat commodities. Chicken meat has a high taste so preferably ranging from children to adults, high biological value, price is relatively cheap, so affordable by almost all social strata. On business side, broiler farm is relatively easy to better control the business scale and investment management. Another benefit of chicken meat commodity that is such ofmeal can be found very wide ranging from households, restaurants through the high prestige hotels where the chicken meat dish is offeredin various menus, and it can be found wherever at home or abroad. Most of all, the duration time of growing chicken meat is very short, in which within 4 weeks (28 days) it has reached an ideal weight so it is very effective selling produce meat.

METHODS

The research experiment was conducted in Faculty of Animal Husbandry Sam Ratulangi University, starting from June to August, 2015.

Research Materials

Animal Experiments: This study was used 3 weeks of age of 120 broiler chickensstrain CP 707 from PT. Charoen Pokphand Indonesia.

Experiment Feed: The experiment feeds which used for the study were of commercial feed CP 11 and CP 12 where the content of nutrients are given in Table 1.

Tabel 1. Nutrients Composition in Commercial Feed

Nutrients	CP 11	CP 12
Water (%)	13,00	13,00
Protein (%)	21,00 - 23,00	19,00 - 21,00
Energy (kcal)	2.961	3.180
Fiber (%)	5,00	5,00
Ash (%)	7,00	7,00
Calcium (%)	0,90	0,90
Phosphorus (%)	0,60	0,60

Source: PT. Chareon Pokphand Indonesia

24 units of the battery cage system were used in this study. Each cage unit was occupied 5 heads of chickens and equipped with dining and drinking plastic cups. Other equipment used were knives, plastic bags, buckets, cutter, rulers, scales and gauges.

Research Methods

This research was used Completely Randomized Design (CRD), Factorial pattern 4 x 3. Factor A were 4 levels, namely: A₁(4 days); A₂ (6 days); A₃(8 days); and A₄ (16 days). Factor B were 3 levels of feeds, namely: B₁ (5%); B₂ (7.5%); and B₃(10%). Each treatment was repeated 2 times. Experimental unit in this study amounted to 24. Each treatment consisted of 5 chickens. This study used 120 broiler chickens that were selected from 200 chickens to fit gain weight uniformity. The feed that used was a commercial feed.

Research Procedure

Rearing chickens were conducted during 42 days which included 21 day starter period and 21 day finisher period. During the starter period chickens were kept in a litter cage. At 21 days of age the weight of chicken were measured to take the initial body weight of the study. Chicken with equal weight were placed randomly into 6 treatments and identity was then labeled in each cage. On day of 21 the amount of chickens feed intake was measured as a benchmark for the treatment of feed restriction. Within 24, 28, 32, and 36 days feed restriction treatments were carried out, then later on the day of 37 to 41 the chicken fed *ad libitum*. Feed intake was observed everyday by measured given amount of each feed treatments subtracted by its residual. On the day of 41 the experiment chickens were fasted for 12 hours, where then weighed to determine the final body weight of the study. At the end of the study the day of 42 the chickens were slaughtered to be taken the carcass. Research variables were carcass weight, abdominal fat and serum cholesterol

Data Analysis Method

This research data analyzed by analysis of variance (Kusriningrum, 2008). The difference among treatment were determined by Least Significant Difference (LSD) test (LSD Fisher).

RESULT AND DISCUSSION

Effect of treatment on carcass weight

The averages of carcass weight for each treatment were shown in Table 3. It can be seen the average carcass weight in this experiment was 1654.32-1804.03 gram. head⁻¹, in which nearly the whole treatments were decreased in carcass weight. Results of the analysis of variance showed that the combination of length of feeding and crude fiber content in the diet was not significantly different interactions. The analysis of variance showed that the length of feeding were significantly (P<0.05) effected on carcass weight. The LSD test showed that the 4-day long giving significantly different from the 8, 12 and 16 days. Carcass weight is influenced by live weight, so the weight of living will followed by a large carcass weight as well. Wahju (1992) stated that the high carcass weight is supported by the end of the live weight. Furthermore, Resnawati (2004) explains that the resulting carcass weight is influenced by several factors such as age, sex, weight pieces, big and body conformation, fat content, quality and quantity of rations and strains maintained.

Table 3 The averages of carcass weight, abdominal fat, and serum cholesterol.

Treatment	Carcass weight (g.head ⁻¹)	Abdominal Fat (g.head ⁻¹)	Serum Cholesterol (mg/dl)
A ₁ B ₁	1682.63	24.15	130.63
A ₁ B ₂	1710.64	25.39	126.38
A ₁ B ₃	1765.64	29.68	126.88
A ₂ B ₁	1732.00	23.96	143.50
A ₂ B ₂	1804.03	21.27	134.50
A ₂ B ₃	1765.64	30.91	119.00
A ₃ B ₁	1678.65	31.48	121.50
A ₃ B ₂	1654.32	22.17	131.00
A ₃ B ₃	1740.00	29.45	125.00
A ₄ B ₁	1660.00	23.99	118.00
A ₄ B ₂	1710.00	22.14	129.00
A ₄ B ₃	1770.07	23.97	142.00

Notes: The different superscription the same row was significantly differences ($P < 0.05$)

Effect of treatment on abdominal fat

The research data in Table 3 shows the average weight of abdominal fat chicken experiment were 21.27 - 31.48 g.head⁻¹. Results of the analysis showed that the combination treatment of length of feeding and crude fiber content in the diet was not significantly different in their interactions affect abdominal fat weight of the broiler. For a long treatment rationing indicate a difference, treatment of crude fiber content in the diet affects weight of abdominal fat is not significant ($P > .05$). For a long treatment rationing, using LSD test showed that the 4-day long giving significantly different from the 8, 12 and 16 days.

According to Fontana *et al.* (1993), abdominal fat will rise to the chickens fed diets with low protein and high-energy rations. The excess energy will be stored as fat in the tissues. One part of the body that is used to store fat by chickens is part around the abdomen. Fat content in broiler chicken carcass is required to give a good appearance on the cuts and to improve the quality of the meat, however if too much it will damage the quality of meat (Amrullah, 2004). The amount of energy consumed in excess will be stored in fat cells. One of place where the fat is accumulation is in the abdominal area. Abdominal fat accumulation is considered as a waste of food energy that resulted in carcass shrinkage.

Effect of treatment on serum cholesterol

The averages of serum cholesterol levels can be seen in Table 3. The data results showed the highest serum cholesterol levels in the treatment of A₂B₁ was 143.50 mg/dl and the lowest in the treatment of A₄B₁ was 118.00 mg/dl. Results of the analysis showed that the combination treatment of length of feeding and crude fiber content in the diet have not significantly different affect the serum cholesterol.

CONCLUSION

Combination of length of feeding and level of crude fiber caused not affected in carcass weight, abdominal fat, and serum cholesterol. However length of feeding tend to decreasing carcass weight and abdominal fat content.

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CRICKET AND PUPA MEAL AS SOURCE OF PROTEIN IN PRE AND POST WEANING LAMB DIET TO EVALUATE HEMATOLOGY AND BLOOD METABOLITE PROFILES

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ABSTRACT

Evidence has accrued over the past 30 years that provides strong support and justification for the sustainable use of insects as a means to produce *protein* for use as *feed livestock, poultry, and aquacultured species*. Cricket meal with 48% protein and pupa meal with 43%, are an alternative source of protein as a substitute of soyabean meal for the pre ruminant diet. The aim of this research was to assess the use of cricket and pupa meal to evaluate blood metabolite profiles of pre and post weaning lamb in a Completely Randomized Design. This research consists of two experiments, first by using twelve pre-weaning lambs which divided into four different milk replacer treatments (control by suckling their mother =C, cow milk=M, cricket meal =CM and pupa meal=PM). Meanwhile another twelve post-weaning lambs were fed with control diet (C), concentrate containing 50% cricket meal (C-50) and concentrate containing 100% cricket meal (C-100). The parameters measured were hematology profiles, plasma glucose, plasma protein and BUN. Result showed that there were no significant differences of hematological parameters of pre-weaning lamb administered with milk replacer containing cow milk, pupa and cricket meal compared to control. Meanwhile the result of post-weaning lamb showed that also there were no significant difference of hematological parameters and blood glucose, protein and BUN. It was concluded that cricket and pupa meal could be used as protein source to support hematological and metabolite profiles of pre and post weaning lamb without any different with control treatment.

Keywords: *cricket meal, hematology, metabolite parameters, pupae meal, pre and post lamb*

INTRODUCTION

Sheep is one of small ruminant which have potential to produce meat. Garut ewes are prolific animal with litter size around 1.77 head per ewes (Inounu, 1996). Sheep population in 2010 was 10,725 heads and increased by 2014 became 15,715 heads (BPS, 2014). Problem with high population is how to overcome the feed. It was reported that around 100 000 ton dry matter concentrate and 7.5 million ton dry matter forage per year were needed to produce sheep in Indonesia (Directorate General Livestock and Animal Health Services, 2011). Nutrient requirement for post weaning lamb or growing sheep with ADG 100 g/d is around 14% CP and 52.50% TDN (NRC, 2007). Source of protein is comes from plant protein and animal protein. Price of animal protein is quite expensive and mostly from import. One of plant protein for concentrate is soybean meal with crude protein content around 49%. Dendi (2012) reported that utilization of 15% soybean meal in the ration has increased performance of growing lamb, but the feed cost was also high. It should be there is other alternative protein source even from plant or animal protein with low price, high quality and easy to produce.

Evidence has accrued over the past 30 years that provides strong support and justification for the sustainable use of insects as a means to produce *protein* for use as *feed livestock, poultry, and aquacultured species*. Cricket is insect with high protein content (48.84% - 56.02%), fat (24.41% - 32.84%) and gross energy 4610 Kal/kg (Sinaga et al, 2010; Novianti, 2003). Utilization of cricket meal for pre-weaning lamb should be limited due to the chitin content (8.70%) where can decrease the digestibility of nutrient. Meanwhile another animal protein source is pupae meal from silkworm with protein content around 55% (Tomotake, 2010). Both of the protein insects are the alternative ingredients to be used either as milk replacer or concentrate for pre and post weaning lamb.

Problem with triplet or quart birth was the limitation of milk quantity from the ewes. Milk production of the ewes not always relate to the number of lamb birth. Tiesnamurtiet *al.* (2002) reported that milk production of garut ewes with single and twin litter size has only 10% by different. So it is needs to help this problem in order to increase sheep population. Milk replacer is liquid diet for pre-weaning lambs with high quality of nutrient content. FAO (2011) reported that milk replacer has 24 % of protein, 22 % of fat and 1.2 % of calcium.

The objective of this research was to assess the use of cricket and pupa meal to evaluate blood metabolite profiles of pre and post weaning lamb.

MATERIALS AND METHODS

This research was consist of two experiments, first by using twelve pre-weaning lambs which divided into four different milk replacer (MR) treatments (control by suckling their mother =C, cow milk=M, cricket meal =CM and pupa meal=PM). The lambs were born from ewes with litter size duplet, one for control and the other lambs divided into three different MR treatments. The MR made from based ingredients such skim milk, powder egg yolk, cream milk, gluten meal, fish oil, minerals and vitamins. Dry matter MR was calculated from 3% of BW and diluted into 250 ml warm water, mixed it and gave two times a day (morning and afternoon).

Meanwhile another twelve post-weaning lambs were fed with control diet (C), concentrate containing 50% cricket meal (C-50) and concentrate containing 100% cricket meal (C-100). The parameter measured for both study were hematology profiles, plasma glucose, plasma protein and BUN. Blood sample was collected at the end of this experiment through jugular vein by using 5 mL syringe. Hematology measurements were done according to standard procedure. All metabolite nutrient parameters were analysed by using KIT with catalog of glucose was no. 112191, triglyseride was no. 116392 and protein was no. 157092. A Completely Randomized Design was used to analyze all data with ANOVA and for further differences of mean treatment was test by DUNCAN (Steel and Torrie, 1995).

RESULT AND DISCUSSION

Result showed that there were no significant difference of hematological parameters of pre-weaning lamb administered with milk replacer containing cow milk, pupa and cricket meal compared to control. It showed that MR containing cricket meal and pupae meal and also 100% of cow milk replaced ewes milk were not affected to the hematological status or good healthy condition. Weiss and Wardrop (2010) reported that the normal lamb hematology have 9-14 trillion/mm³ of

RBC, 9-15 g.dL⁻¹ of hemoglobin, 27-45% PCV and 4-12 million/mm³ of WBC. The hematology profiles of pre-weaning lamb fed containing cricket meal as shown at Table 1.

Table 1. Hematology profile of pre-weaning lamb fed cricket meal

Parameters	C	M	CM	PM
RBC (x 10 ⁶ .mm ⁻³)	7.54 ± 1.48	7.55 ± 1.19	7.37 ± 1.33	7.23 ± 2.76
Hemoglobin (g.dL ⁻¹)	11.63 ± 0.90	10.27 ± 0.70	9.87 ± 0.31	9.93 ± 1.33
PCV (%)	28.67 ± 2.08	28.67 ± 2.52	27.83 ± 2.57	27.25 ± 0.20
WBC (x 10 ³ mm ⁻³)	12.73 ± 1.95	12.92 ± 1.83	15.27 ± 1.00	9.85 ± 0.21

C = control suckling from their mother; M= MR 100% of cow milk; CM = MR containing cricket meal and PM= MR containing pupae meal

Metabolite parameters which relate to nutrient status of pre-weaning lambs such glucose, triglyceride and total protein were same in all treatment. Cow milk, MR with cricket meal and MR with pupae meal have same effect to the nutrient status of pre-weaning lambs compare to control, except triglyceride. This mean that pupae and cricket meal in part of milk replacer ingredient would not give any disturbance to the metabolite status. Kaneko *et al.* (1989) reported that normal blood glucose in lamb around 50-100 mg/dL. Meanwhile Smith and Mangkoewidjojo (1988) reported that the protein content in healthy lamb blood plasma were 6.0 – 7.59 g dl⁻¹. Result of blood triglyceride showed that there was a significance difference (P<0.05) due to the treatment, where milk replacer containing pupae meal was the highest. Pupae meal has high fat content (32%). Gani *et al.*(2013) reported that the normal blood triglyceride in lamb is around 26-145 mg/dL. It showed that the lamb has normal triglyceride concentration so far.

Table 2. Glucose, triglyceride and total protein plasma of pre-weaning lamb fed cricket meal

Parameters	C	M	CM	PM
Glucose (mg/dL)	118.18 ± 14.93	126.32 ± 19.31	144.01 ± 14.62	134.94 ± 46.30
Triglyceride (mg/dL)	47.51 ± 5.70b	26.62 ± 3.76b	39.30 ± 7.09b	134.33 ± 49.25a
Total protein (mg/dL)	5.52 ± 0.49	6.03 ± 0.50	6.14 ± 0.71	6.04 ± 0.21

C = control suckling from their mother; M= MR 100% of cow milk; CM = MR containing cricket meal and PM= MR containing pupae meal

The result of post-weaning lamb or growing lamb showed that there were no significant difference of hematological parameters, it means that the utilization of cricket meals as substitute of soybean meal has no effect to the health condition of the animals (Table 3). The PCV value was significant different due to the treatment, where milk replacer containing soybean meal has higher than other treatments (P<0.05). Percentage of PCV has correlation with water intake. More protein intake

will increase higher water intake which will dilute the blood in the body so that the high water intake will reduce percentage of PCV. Astuti et al. (2008) reported that lamb blood profile which suckling from their mother by grazing at tropical rain forest have 7.57 trillion/mm³ of RBC, 7.21 g.dL⁻¹ hemoglobin and 28.10% of PCV.

Table 3 Hematology profile of post-weaning lamb fed cricket meal

Parameters	MR (SB)	MR (SB+CM)	MR (CM)
RBC (x 10 ⁶ mm ⁻³)	9.43 ±1.38	9.11 ± 0.49	9.09 ± 0.23
Hemoglobin (g.dL ⁻¹)	11.23 ± 0.87	11.13 ± 0.98	11.05± 0.41
PCV (%)	30.75 ± 1.71b	28.25 ± 2.06a	27.25 ± 0.50a
WBC (x 10 ³ mm ⁻³)	12.38 ±1.07	13.76 ± 0.91	13.98 ± 2.12
- Netrophil (%)	39.32 ± 5.58	39.75 ± 1.50	41.05 ± 2.31
- Eosinophil (%)	3.66 ±1.50	6.25 ± 1.26	5.23 ± 0.98
-Lymphosit (%)	52.89 ± 6.14	49.25 ± 0.96	49.49 ± 2.73
- Monosit (%)	2.91 ± 0.19	3.25 ± 0.50	2.74 ± 0.51
- Basophil (%)	1.22 ± 0.48	1.50 ± 0.58	1.49 ± 0.57

MR (SB)= milk replacer containing soybean meal; MR (SB+CM) = milk replacer containing soybean meal and cricket meal; MR (CM) = milk replacer containing cricket meal

Data statistic analysis of metabolite status of post weaning lamb fed different level of cricket meal same in all treatments (Table 4). Again, the result showed that cricket meal in the ration could substitute soybean meal until 100% without any problem to the nutrient absorption and status of metabolite glucose and protein. Some chitin from cricket meal will reduce the absorption due to the low digestibility. Wang *et al.* (2005) reported that in 100 g of cricket meal has protein contain 58.30% and chitin around 8.70% which can disturbance to the nutrient absorption.

Protein which is absorpted through the blood system can be as ammonia, albumin or globulin (Frandsen 1992). This protein will distribute to whole body for support nutrient requirement of the animal. So it can be determine that protein status in blood will relate to protein intake. This also happen for all other nutrients status in the body.

Table 4. Glucose and total protein plasma of post-weaning lamb fed cricket meal

Parameters	MR (SB)	MR (SB+CM)	MR (CM)
Glucose (mg/dL)	66.22 ± 7.52	58.68 ± 8.70	58.04 ± 7.98
Total protein (mg/dL)	6.76 ± 0.24a	8.32 ± 1.29b	6.09 ± 0.62a

MR (SB)= milk replacer containing soybean meal; MR (SB+CM) = milk replacer containing soybean meal and cricket meal; MR (CM) = milk replacer containing cricket meal.

CONCLUSION

It was concluded that cricket and pupa meal could be used as protein source to support hematological and metabolite profiles of pre and post weaning lamb without any different with control treatment.

ACKNOWLEDGEMENT

This research was supported by private budget collaboration from Author, co-author and under-graduate students involved. I would like to thanks to The Insect Team who have already work hard for finishing this project.

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INCREASING PRODUCTIVITY DAIRY GOAT BY FEEDING WAFER SUPPLEMENT CONTAINING LAMTORO AND KALIANDRA LEAVES

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ABSTRACT

Productivity is very important factor to support reproductive of dairy goat. One way of increasing productivities of dairy goat is by improving the production and reproduction, breeding, and availability of high quality feeds. The aim of this experiment was to determine the influence of feeding wafer supplement containing lamtoro dan kaliandra leaves on productivity of dairy goat. This research was conducted at Laboratory of Feed Industry, Faculty of Animal Science, Bogor Agricultural University, Indonesia; whereas productivity test was conducted at Cordeo Farm, Bogor from February to April 2015. The experimental design used was Randomized Block Design with four treatments and three replications. Wafer feed supplement containing lamtoro and kaliandra leaves. The treatments were formulated as follow: R0= conventional feed without supplement; R1= RO + 5 % feed supplement; R2= RO + 10% feed supplement; and R3= RO +15% feed supplement. The results indicated that treatments gaveno significant effect ($P>0.05$) on final body weight, daily weight gain, and feed efficiency. Meanwhile, treatments significantly affected ($P<0.05$) group body weight, and ewes with a small body weight was more responsive to feed supplements. Feed supplement containing lamtoro and kaliandra leaves significantly ($P<0.05$) increased udder and teats length of ewes, accelerated estrus and pregnancy rates. It can be concluded that the dairy goats consumed 15% feed supplement containing lamtoro and kaliandra leavesgrew faster than the other treatments and increased udder and teatslength, period of estrus, and pregnancy rate of ewes.

Keywords: *Feed supplement, Body weight, Dairy goat, Feed efficiency, Productivity*

INTRODUCTION

Goat as one of the livestock in Indonesia has contributed significantly to the income of small farmers. Goats used as a sideline business because producing and marketing of meat, milk, leather and feces are relatively easy. The female goat is called a “doe” or “nanny.” When she is between the age of 6 to 12 months she is sometimes referred to as a “doeling.” The doe can reach puberty between 4 to 12 months of age. However, overfeeding or underfeeding the goat can hinder puberty as well as her reproductive performance. A lack of adequate nutrition will subsequently hinder lactation (Jakes, 2007). In order to increase the goat productivity is by improving the performance of production and reproduction, breeding, and high quality feed. Managemen at the farm is still traditional. Problems often occur in livestock raising goat is a health problem and limited livestock feed.

The major constraints of ruminant feed are as follows: low quality of forage; the level of palatability and digestibility is low. Therefore, it is necessary to develop suitable technologies to produce ruminant feed which is more durable, easier to handle, more convenient to distribute and are available in all seasons (Retnani *et*

al.,2013a). During the rainy season, forage is abundant, but during the dry season forage very little or none so that the goat can decrease its productivity (Retnani *et al.*,2013b).

To solve this problem is making forage into wafer feed. A pressing technology can make the feed products into a wafer form. The wafer feed must contain energy; mineral; vitamin and protein that are needed by animal to increase productivity (Retnani *et al.*,2010a). Wafer of feed supplement is made of fiber, especially fresh green forage as a replacement for ruminants in order to utilize the fiber when the quality and quantity of forage decreased (Retnani *et al.*, 2013a).

MATERIALS AND METHODS

The experiment used 12 heads of dairy goat. The dairy goat were females between the age of 6 to 12 months with average initial body weight around 25.54± 2.6 kg. The experimental were maintained individually cages. The experimental maintained into individually cages. The ration used consisted of two types conventional feed and wafer feed. Nutrient content of wafer feed (% dry matter) are presented on Table 1.

Table 1. Nutrient Content of Wafer Feed (% Dry Matter)

Ingredient	Water content	Ash	Crude protein	Crude fiber	Crude fat	NFE
Nutrient	6.98	8.30	33.73	10.45	5.87	41.65

Laboratory Analysis of Feed Science and Technology (2015)

The process of wafer of feed supplement production was conducted by chopping, drying, mixing, preassing, heating and forming with temperature of 100⁰C for 10 minutes to get wafer of feed supplement and then being cooled in room temperature (Retnani *et al.*, 2014). Figure 1 showed that diagram process of wafer feed production.

Experimental Design

The experimental design used Randomized Block Design with four treatments and three replications. The treatments were level of wafer of supplement i.e R0= conventional feed , R1=R0 + 5 % wafer of feed supplement containing lamtoro and kaliandra leaves, R2=R0 + 10% wafer of feed supplement containing lamtoro and kaliandra leaves, R3=R0 +15% wafer of feed supplement containing lamtoro and kaliandra leaves. Conventional feed were forage and concentrate. The data was analyzed with the analysis of variance (ANOVA) and T-Test for data of the length udder and the teats of goat, and the differences among treatments were examined with duncantest (Steel and Torrie, 1993).The parameters measured were body weight, daily weight gain, feed conversion, the length udder and teats of goat.

The variables that would be measured were:

- (i) Body weight was calculated by body weight goat were weighed every 2 weeks during maintenance
- (ii) Daily weight gain was calculated by:

$$\text{Daily weight gain (g/head/day)} = \frac{\text{Final body weight again (g)} - \text{initial body weight gain (g)}}{\text{During the research (days)}}$$

(iii) Feed conversion was calculated from feed consumption (kg) divided by body weight again (kg).

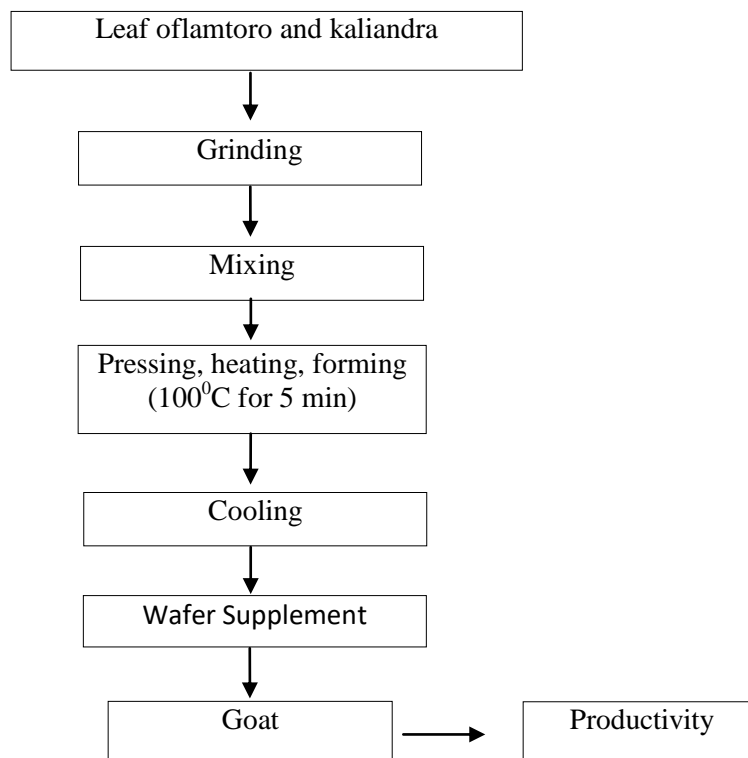


Figure 1. Diagram Process of Wafer Feed Production (Retnani *et al.*, 2013b)

RESULTS AND DISCUSSION

Physical form of wafer feed was compact with size 3x5x5 cm³. The texture of wafer feed was rough with coarse particle size. During the process making wafer feed by heating and pressing did not decline the nutritional quality. Wafer feed had physical characteristics which were water content 11.71 %, water absorption 96.02%, water activity 0.81 and density 0.97 g/cm³ (Retnani *et al.*, 2014). In this research used conventional feed. Conventional feed were forage and concentrate. Nutrient content of conventional feed (% dry matter) are presented on Table 2.

The result showed that wafer supplement treatment did not significant (P>0.05) on daily weight gain of goat. However, feeding treatment was significant effect (P<0.05) on block of daily weight gain. Daily weight gain (g/head/day) was shown on Table 3. It was mean, wafer feed given to the goat that have a medium body weight produced higher daily weight gain (104.23 g/head/day) compared with the other treatments.

Table 2. Nutrient Content of Conventional Feed (% Dry Matter)

Ingredient	Water content	Ash	Crude protein	Crude fiber	Crude fat	NFE
Forage	11.12	12.81	15.17	3.26	24.45	44.31
Concentrate	5.43	4.24	14.87	4.55	27.76	48.579

Laboratory Analysis of Feed Science and Technology (2015)

Tabel 3. Daily Weight Gain (g/head/day)

Treatment	Block			Average
	B1	B2	B3	
R0	66.90	121.43	90.00	97.28± 27.37
R1	102.86	102.86	99.29	89.60± 19.94
R2	97.86	88.10	69.52	85.16± 14.39
R3	112.62	108.10	95.24	105.31± 9.01
Average	95.06±19.7 ^a	104.23±14.9 ^b	80.36±14.37 ^c	

R0= conventional feed , R1=R0 + 5 % wafer of feed supplement containing lamtoro and kaliandra leaves, R2=R0 + 10% wafer of feed supplement containing lamtoro and kaliandra leaves, R3=R0 +15% wafer of feed supplement containing lamtoro and kaliandra leaves.B1 (Small body weight block), B2 (medium body weight block), B3 (Big body weight)

Dailyweight gainin this studyranged from66.90-121.43g/head/day. The treatment of R3(R0 +15% wafer of feed supplement containing lamtoro and kaliandra leaves)was highest daily weighth gain thanwith other treatments.According to NRC (1985),daily weight gain was influenced by several factors, i.e. the total consumption of protein, sex, age, genetic, environmental, physiological condition of livestock and management. According to Handoyono (2004) that dailyweight gain of goat was 88,2g/head/day. Setiawan dan Tanius (2005) states that the daily weight gain ofgoat was 100 g/head/day

The result showed that wafer suplement treatment did not significant (P>0.05) on average final body weight of goat. However, feeding treatment was significant effect(P<0.05) on block of body weight.Final body weight of goat (kg/head) was shown on Table 4.

Table 4. Final Body Weight of Goat (kg/head)

Treatment	Block			Average
	B1	B2	B3	
R0	27.70	31.03	32.26	30.33± 2.35
R1	26.39	29.99	34.68	30.35± 4.16
R2	31.13	32.42	31.43	31.48± 0.38
R3	27.52	33.28	34.08	31.63± 3.58
Average	28.19±2.05 ^a	31.55±1.39 ^b	33.11±1.52 ^c	

R0= conventional feed , R1=R0 + 5 % wafer of feed supplement containing lamtoro and kaliandra leaves, R2=R0 + 10% wafer of feed supplement containing lamtoro and kaliandra leaves, R3=R0 +15% wafer of feed supplement containing lamtoro and kaliandra leaves.B1 (Small body weight block), B2 (medium body weight block), B3 (Big body weight)

Treatment of R3 (R0 +15% wafer of feed supplement containing lamtoro and kaliandra leaves) have average body weight of the highest compared to the other treatments.Goat were fed by conventional fed had final body weight 30.33 kg, meanwhile goat were fed15% wafer of feed supplement containing lamtoro and kaliandra leaves31.63 kg or 4.11% higher than conventional. According Markel and Subandryo (1997) body weight goat can reach 35 kg.

Feed conversion was affected by feed quality, digesttibility value, and efficiency. Increase in feed quality will improve body weight gain, so feed conversion value will decrease, meaning that the application of feed is efficient (Pond *et al.*, 1995). Feed conversion depends on dry matter intake and body weight gain.The result showed that wafer suplement treatment did not significant

($P > 0.05$) on feed conversion with average of feed conversion value were 7.04 ± 0.76 until 8.64 ± 1.59 . Feed conversion was not significant in this research caused by insignificant increase in body weight gain.

By feeding wafer of feed supplement containing lamtoro and kaliandra leaves had significantly effected ($P < 0.05$) on increasing the length udder and teats of goat. Goats were fed by 15% wafer of feed supplement containing lamtoro and kaliandra leaves had the length udder of goat longer than the other treatments.

Table 5. The length udder of goat (cm)

Treatment	Length Udder	
	Before Treatment	After Treatment
R0	11.00 ± 1.00^a	11.67 ± 0.58^a
R1	12.67 ± 2.52^b	12.33 ± 1.58^b
R2	11.33 ± 1.53^c	12.67 ± 0.58^c
R3	9.33 ± 0.58^d	12.67 ± 1.15^c

R0= conventional feed , R1=R0 + 5 % wafer of feed supplement containing lamtoro and kaliandra leaves, R2=R0 + 10% wafer of feed supplement containing lamtoro and kaliandra leaves, R3=R0 +15% wafer of feed supplement containing lamtoro and kaliandra leaves.

Udder length of goat is varies, usually ranging from 10-20 cm, and the teats of goat range 5-10 cm (Tedjowati, 1988). The teats goat presented in Table 6. The range of teats goat before treatment in the study is 5-6 cm and after treatment were 6-7 cm.

Table 6. The teats of goat (cm)

Treatment	Teats	
	Before Treatment	After Treatment
R0	6.00 ± 0.00^a	6.33 ± 0.47^a
R1	5.00 ± 1.00^b	5.67 ± 0.58^b
R2	5.33 ± 1.00^c	6.00 ± 1.00^c
R3	4.67 ± 0.57^d	6.67 ± 0.57^c

R0= conventional feed , R1=R0 + 5 % wafer of feed supplement containing lamtoro and kaliandra leaves, R2=R0 + 10% wafer of feed supplement containing lamtoro and kaliandra leaves, R3=R0 +15% wafer of feed supplement containing lamtoro and kaliandra leaves

By feeding wafer of feed supplement containing lamtoro and kaliandra leaves accelerating estrus and pregnancy rates. Goat reaches age puberty around 5-6 months. Signs of estrus goat was discharge clear mucus and watery during estrus that form the crystallization patterning such as ferns, the mucus becomes thick white mass of cells that contain many elements horned (Davendra and Burns, 1994). According Davendra and Burns (1994), Setiadi (1987) the length of the estrous cycle of goat was 20.25 days with a range of 7-27 days.

Table 7. Signs estrus of goat

Treatment	Weeks							
	1	2	3	4	5	6	7	8
R0U1	-	-	Estrus					
R0U2	-	-	-	-	-	-	-	-
R0U3	-	-	-	-	-	-	-	-
R1U1	-	Estrus						
R1U2	-	Estrus				pregnant		
R1U3	-	-	-	-	-	-	-	-
R2U1	-	Estrus						
R2U2	-	-	-	-	-	-	-	-
R2U3	-	Estrus						
R3U1	-	Estrus						
R3U2	-	Estrus						
R3U3	-	-	-	-	-	-	-	-

R0= conventional feed , R1=R0 + 5 % wafer of feed supplement containing lamtoro and kaliandra leaves, R2=R0 + 10% wafer of feed supplement containing lamtoro and kaliandra leaves, R3=R0 +15% wafer of feed supplement containing lamtoro and kaliandra leaves

CONCLUSION

Goats were given 15% wafer of feed supplement containing lamtoro and kaliandra leaves had the length udder and teats of goat longer than the other treatments. Descriptively, there were accelerating estrus and pregnancy rates by feeding wafer of feed supplement containing lamtoro and kaliandra leaves.

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EFFECT THE ADDITION OF CALCIUM SOAP CANOLA AND FLAXSEED OILS ON VOLATILE FATTY ACID PROFILES IN THE *in vitro* FERMENTATION

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ABSTRACT

Supplementation of vegetable oils as a source of unsaturated fatty acid in ruminant diets could improve the production and quality of beef meat. However, the utilization of vegetable oils as a source of unsaturated fatty acids needed to be protected to avoid bio hydrogenation process by rumen bacteria which convert to saturated fatty acid otherwise. Calcium soap (Ca soap) is one of protection agent of unsaturated fatty acids that is easier and cheaper to be applied. This experiment was designed to evaluate the effectiveness of calcium soap of Canola oil (high linoleic acid content) and flaxseed oil (high in linolenic acid) on fatty acid profile at 4 and 8 hours fermentation. The experimental design was conducted in a Randomized Block Design with 3 treatments and 4 blocks as replications. The treatments were formulated as follow: control diet (forage:concentrate = 60:40), diet with Ca soap-canola oil 6%, and diet with Ca soap-flaxseed oil 6%. Variables observed were rumen fatty acid profile at 4 h fermentation for volatile fatty acid (VFA) concentration and at 0, 4, 8 h fermentation for fatty acid profile. The result showed that Ca soap-oils increased total VFA production and proportion of propionate. The addition of Ca soap-canola oil and flaxseed oil improved total VFA up to 25% and 34%, respectively, compared to control diet. It can be concluded that utilization of Ca soap-flaxseed oil in the diet decreased acetate proportion, increased propionate proportion and reduced acetate propionate ratio (A/P) compared to the control and Ca soap-canola oil treatments.

Keywords: *Volatile fatty acid, Ca soap, Canola oil, Flaxseed oil, Rumen fermentation.*

INTRODUCTION

Supplementation of vegetable oils unsaturated fatty acid in ruminant feed could improve the production and quality of beef meat. However, the using of vegetable oils as unsaturated fatty acids sources need to be protected to avoid bio hydrogenation process by rumen bacteria which convert to saturated fatty acid. Feed ingredients derived from vegetable fat contains a lot of unsaturated fatty acids, such as oleic, linoleic and linolenic acid that will undergo biohydrogenation process or saturation reaction massively in the rumen which could increase the saturated fatty acids in the rumen. Moreover, vegetable oil also is a source of alternative energy needed by cattle. However, the use of vegetable oil as ruminant feed must to be restricted because of some disadvantage effect in the rumen system. According to Bunting (1996), when the oil is given above 5% in the ruminant diets might disrupt the microbial population in the rumen and reduce the ability of ruminants in the feed digestion.

To overcome this problem, the addition of vegetable oil need to be protected to avoid bio hydrogenation process by rumen bacteria. Calcium soap (Ca soap) was protection method of unsaturated fatty acids that can be easy and cheap to be applied.

Calcium soaps is a chemical process to produces soap from fat and alkali materials known as saponification and added with calcium chloride.

Vegetable oil which contain a lot of unsaturated fatty acids include flaxseed oil and canola oil. According to Carter (1993), flaxseed contains 32-45% oil, which is 51-55% alpha - linolenic acid (Omega 3 family), 15-18% linoleic (Omega 6). Moreover, canola oil contain 60% oleic, 20% linoleic and 10%linolenic acid (Holländer et al., 2012). Our previous research showed that calcium soap of flaxseed oil or canola oil addition at level 4% in the concentrate feed have higher endurance from biohydrogenation process in the in vitro rumen fermentation and increased volatile fatty acid production compared with calcium soap from sesame oil (Suharti et al., 2014; Hidayah et al., 2014).

Based on our previous results, this research aimed to assess the influence of calcium soaps canola oil or flaxseed oil at the higher level i.e 6% in the ruminant concentrate feed on rumen microbe population and volatile fatty acid *in vitro*.

MATERIALS AND METHODS

***In vitro* Fermentation.** The canola oil was obtained from CV. MH. Farm Bogor and flaxseed oil were produced by Green Tosca. Calcium soap from these two kinds of vegetable oils was made according to method by Kumar *et al.*, (2006). *In vitro* fermentation was conducted according to Tilley and Terry method (1963). The rumen fluid for this experiment was obtained before morning feeding from the rumen of fistulated Ongole crossbred beef cattle using commercial concentrate and elephant grass as substrate. The substrate for in vitro frementation contained forage and concentrate mixture (cassava by product, wheat pollard, soybean meal, coconut cake meal, molasses, CaCO₃, premix, urea, and oil) with 15-17 %CP and 69-74% TDN (Table 1). The design of experiment was completely block design with 3 treatments were T1 = 60% elephant grass : 40% concentrate (Control/C), T2 = C + 6% ca-soap canola oil and T3=C + Ca-Soap Flaxseed oil. Volatile fatty acid profile, protozoa and bacteria population were measured from liquid sample taken at 4 h incubation. Volatile fatty acid analysis was performed by using Gas Chromatography (GC 8A, Shimadzu Crop., column SP-1200, 1% H₃PO₄ on 80/100 Cromosorb WAW). Protozoa counting under a microscope and bacteria population measurement by using roller-tube method (Ogimoto & Imai, 1981).

Table 1. Nutrient composition of in vitro fermentation substrate supplemented with Ca-soap Canola Oil or Flaxseed oil

Nutrient (%)	Treatment		
	Control (C)	C + 6% Ca-Soap Canola oil	C + 6% Ca-Soap Flaxseed oil
Dry Matter	86.08	81.04	81.04
Ash	5.49	5.77	5.78
Crude Protein	13.55	14.50	14.44
Crude Fat	4.96	10.02	9.20
Crude Fiber	14.10	14.99	14.93
TDN	72.39	72.37	72.53

Note :1. Control = 60% Elephant grass + 40% concentrate mix. 2. Estimation of TDN by Hartadi (1980) formula. $TDN = 92.464 - (3.338 \times CF) - (6.945 \times EE) - (0.762 \times \text{Beta-N}) + (1.115 \times CP) + (0.031 \times CF^2) - (0.133 \times EE^2) + (0.036 \times CF \times \text{Beta-N}) + (0.207 \times EE \times \text{Beta-N}) + (0.1 \times EE \times CP) - (0.022 \times EE \times CP)$

Statistical Analysis. Rumen microbe data were analyzed by ANOVA using the GLM procedures (SPSS 13.0 for windows, 2004. The differences among all treatments were separated by Duncan’s multiple range test was used to compare the means of the treatments. Data of volatile fatty acid analyzed by descriptive.

RESULT AND DISCUSSION

Rumen Microbe population

The addition of Ca-Soap canola oil or flaxseed oil did not affect bacteria and protozoa population (Table 2). This result indicating that the use of high unsaturated fatty acid protected with calcium soap did not alter the growth of rumen microbe. As we know that some of unsaturated fatty acid might toxic for rumen microbe and the use of high level of fat sources will interfere the activity of rumen microbe in the fiber digestion. Tanuwiria (2011) reported that the increase of unsaturated fatty acid (C18) content in the ration will decrease the bacteria population.

Table 3. Effect of the addition of Ca-soap Canola oil or Flaxseed oil on Bacteria and Protozoa population in the in vitro fermentation

Treatment	Bacteria population (Log cfu/ ml)	Protozoa population (Log cell/ml)
Control (C)	6.68 ± 0.76	3.95 ± 0.13
C + 6% Ca-Soap Canola oil	6.27 ± 0.25	3.85 ± 0.26
C + 6% Ca-Soap Canola oil	6.27 ± 0.18	3.94 ± 0.13

The addition of Ca-soap of Canola oil or flaxseed oil did not inhibit the growth of protozoa and bacteria population indicating the effectivity of calcium soap method in the fatty acid protection. Moreover, the toxic effect of unsaturated fatty acid on rumen microbe could be reduced and did not alter activity of rumen microbe in the feed degradation and fermentation.

Previous research reported the effect of non protected fatty acid on rumen protozoa. Sondakh *et al.* (2012) reported that the feed which contain medium chain fatty acid (MFCA) up to 1.5% significant decreased protozoa population. Lauric acid in the MCFA have defaunating effect and caused the decreasing of protozoa population. Machmuller (2006) suggested that MCFA could inhibit the growth and activity of ciliate protozoa (*Entodinium spp.*) and gram positive archaea as lauric acid might increased the sensitivity of cell wall of rumen microbe.

The lauric acid content in the Ca-soap canola oil or flaxseed oil were fairly high, approximately 2.9% and 1.48%, respectively. However, the addition of Ca-soap canola oil or flaxseed oil did affect the protozoa population because oil which protected by Ca-soap technology will stable in the rumen that have neutral pH (6-7) and fatty acid compound (including lauric acid) will inert so did not alter rumen microbe.

Volatile Fatty Acid (VFA) Profile

The addition of Ca-soap canola oil and flaxseed oil could increased the total VFA production compared with control treatment. Moreover, the use of Ca-soap flaxseed oil resulting the highest total VFA production, propionate proportion and C2/C3 ratio (Table 3). The increasing of VFA production might be caused by the glycerol content in the ca-soap product as a result of lipid hydrolysis. Glycerol will

be convert to be VFA by rumen microbe as an energy sources of ruminant. Sakinah (2005) suggested that the indicator of energy fulfillment of ruminant was the high of VFA production.

The addition of Ca-soap canola oil or flaxseed oil increased propionate proportion, decreased acetate and butyrate proportion as well as decreased propionate/acetate (C₂/C₃) ratio.

Table 3. Effect of the addition of Ca-soap Canola oil or Flaxseed oil on volatile fatty acid profile

Parameters	Treatments		
	control	C + 6% Ca-Soap Canola oil	C + 6% Ca-Soap Flaxseed oil
total VFA (mM)	36.53	45.57	49.01
Proportional VFA (mM/100 mM)			
Acetate (C ₂)	67.24	65.22	65.69
Propionate (C ₃)	22.10	22.42	25.84
Isobutirate + Butirate (C ₄)	9.77	10.57	7.84
Isovalerate + Valerate (C ₅)	0.88	1.78	0.63
C ₂ : C ₃	3.04	2.91	2.54
Methane Estimation (mM)*	10.27	12.49	12.54

Note: Methane (mM) = 0.45 C₂ – 0.275 C₃ + 0.40 C₄ (Moss *et al.* 2000)

The higher level of propionate proportion indicating that the availability of energy sources increased because propionate has a role as major energy sources of beef cattle. However, the addition of Ca-soap canola oil and flaxseed oil increased the estimation of methane production. The increasing of methane gas production inline with the increasing of total VFA production. As we know, the side effect of rumen fermentation activity improvement will produce the high level of gas production including methane. Moreover, the increasing of methane production might be due to the inhibition of biohydrogenation process in the ca-soap treatment. The biohydrogenation process using hydrogen which available in the rumen, thus the inhibition of biohydrogenation process will increase the availability of hydrogen and stimulating for methane production. Jalc *et al.* (2007) reported the reducing of propionate proportion and the increasing of acetate proportion with the addition of oleic acid. However, the addition of alpha linolenic acid increased propionate proportion and decreased acetate propionate ratio.

CONCLUSIONS

The addition of Ca-soap canola oil or flaxseed oil did not interfere the growth of rumen microbes and could stimulate the fermentation activity. The use of Ca-soap flaxseed oil increased Total VFA production and propionate proportion, but decreased acetate proportion and C₂/C₃ ratio

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THE EFFECT OF SUPER FORMULA SILAGE QUALITY IN RATION ON DAIRY COWS PERFORMANCES AT TRADITIONAL DAIRY FARMING

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ABSTRACT

An effort to sustain quality feed supply for dairy cattle at traditional dairy farming has been done by using super formula silage technique on traditional dairy farmer best feeding practice and silage technology application. This experiment was aimed to compare cattle performances (body condition score, milk production, and qualities) at three different types of feeding practices: super formula based on fresh forage (R1), ration based on conventional silage (R2), and ration based on super formula silage (R3). Nine lactating cows at early-mid lactation cycles have been used. The results showed that R3 produced higher milk production compared to R2 and R1 (19.73, 18.1 and 11.73 l h⁻¹.d⁻¹), but R1 produced higher milk fat and total solid compared to R3 and R2. The higher milk component secreted at R3 could not be balanced by the nutrient intake of the cows resulted in lower BCS. In conclusion, the super formula silage superior in milk production and quality, but still unsustainable for long term nutrient supply and body storage.

Keywords: *Lactating dairy cows, milk, ration, silage, traditional feeding system*

INTRODUCTION

Indonesian domestic demand for milk increased 10%/year during 2000 to 2006, faster than local farmer ability to produce the milk which were only increase 2.5%/year (Ahmad & Hermiyetri, 2007). The conditions lowered domestic milk contribution from 30% (before 2005) to 18% by the end of 2012. The contribution will continue to shrink if there is no significant intervention will be made.

Indonesian dairy farming, like most of dairy farming in South-East Asia, previously were developed as government support to improve social welfare because its labor intensive, thus provide many employment opportunities on farm, milk transportation and processing (Moran, 2005). In average, the farmer owned 0.44 ha land and grew high production but low quality grasses such as Napier grass to provide 63.7% of forage required by their 6.07 AU cattle (Survey Nasional Sapi Perah, 2012). The rest (about one third) of the requirements were fulfilled from natural grasses and agricultural by product (Despalet *et al.*, 2014^a) which were collected along roadsides, rice paddies bank (Moran, 2005), and horticulture land surrounding the farms, purchased collectively or individually. The available grasses contained lower nutrition than the dairy cattle's need (Despalet *et al.* 2014^{b,c}). Their availabilities and qualities were seasonally fluctuated (Despalet *et al.* 2013).

In order to provide continuous supply of sufficient nutrients for lactating dairy cows, therefore, the forage should be preserved and supplemented with effective concentrates (Moran, 2005). One of the forage preserving techniques is ensilage. Ensiling preserves forage through spontaneous fermentation of forage water soluble carbohydrate (WSC) into lactic acid by epiphytic lactic acid bacteria (LAB) under *anaerob* condition. Silage quality was determined by fermentative and aerobic stability characteristics which were influenced by forage condition, harvesting

condition and ensiling technique. Forage condition factors included moisture and water soluble carbohydrate contents which varied by species and variety (Mironet *al.*, 2007), age (Montemayoret *al.*, 2000; Mironet *al.*, 2006; Nadeau, 2007) and planting condition (Dawoet *al.*, 2007).

Although silage could be used as an alternative strategy to sustain quality supply of nutrients for dairy cattle, its utilization in tropical area especially by smallholder farmers were still limited. It might be caused by lack of knowledge (Rangnekar, 2000), lack of capital (Nakamanee, 2000), complicated process, limited value added, low animal genetic potential (Rangnekar, 2000), high cost (Raza, 2000), lack of planning and forage availability as well as equipment (Rangnekar, 2000). According to Woolford (1984), tropical legume and grass were less ideal for silage making because their low WSC but high moisture contents, high buffering capacity which lead to high dry matter and protein degradation during ensiling. High fiber content, temperature and sunlight intensity during storage were some other drawback factors to the silage application (Elferinket *al.*, 2000).

Silage might be difficult to be applied in tropical countries, however, it becomes urgent nowadays because of the difficulties in collecting natural grass after high land conversion and lack of family worker. On the other hand, seasonal byproducts became more available (Nakamanee, 2000) commercially.

Through several projects, silage forage have been introduced (Despalet *al.*, 2013) to several farmer members of dairy cooperatives in Indonesia. To overcome several limitation and barriers in applying silage forage for smallholder dairy cattle, some modifications have been applied as suggested by Mannetje (2000), such as smaller silo capacity, simplification ensiling technique, reduce investments cost, utilize local materials, faster return on investment as suggested by Nakamanee (2000) as well as co-finance system which help farmer at early stage. Several farmers in several cooperatives have been trying to make silage and included them in daily dairy rations. So far, the farmers making silage based on available materials and less considered on the silage quality.

Through serials research of dairy farming in highland and lowland areas of Indonesia, several best practices formula of dairy rations have been identified and tested. The rations based on Napier grass and cooperative made concentrate. For the high yielding cows, the farmers supplement the basic ration with tofu and cassava waste which are become difficult to be reached at reasonable price by farmer. The ingredients are thought to be the only feed that could support long lasting high production cows, therefore, some farmers still used it. To help farmer in overcome dependencies to tofu and cassava waste, supplement super formula silage have been introduced.

The objectives of this study was to evaluate the impact of utilizing super-formula silage in daily rations on nutrient consumptions and digestibilities as well as dairy cattle performances.

MATERIALS AND METHODS

The research have been done on February 2014 to April 2015. Research have been conducted at KPSBU cooperative demo farm in Lembang and PAU-IPB laboratory. Twelve lactating cows with average bodyweight 495.24 ± 44.70 kg have been grouped into 3 groups according to their lactation period (early, mid and late) and have been kept individually and fed ration according to the treatments.

Two type of rations (supplement tofu and cassava waste (R1) and silage super-formula (R2)) have been offered 3% of the cows body weight in two times equal interval feeding frequencies. Ingredients and nutrient compositions of each ration are shown in Table 1.

Table 1. Ration Ingredients and nutrient compositions

Ingredients	R1	R2
	------(%)-----	
Napier grass	37.15	19.41
Concentrate (Mako)	42.46	61.83
Tofu waste	11.60	
Cassava waste	17.58	
Super formula silage		18.74
Total	100	100
Nutrient contents (%DM)		
Ash	13.11	12.33
Organic matter	86.89	87.67
Crude Protein	17.67	18.14
Lipid	3.85	4.17
Crude Fiber	20.25	18.82
NFE	45.11	46.53

The cows were observed for 6 weeks (5 weeks preliminary and 1 week collecting periods). During the observation periods, feeds offered and refusal were weighted twice daily, sampled, dried and composited for proximate compositions analyzing. Milk produced were measured and sampled for morning and afternoon milking, and analyzed for its fat, solid non-fat (SNF), lactose, protein, and milk density using *Lacto-scan*. Body weight were measured at the beginning and the end of the observation periods two determine the gains. Body condition were scored based on fat deposit on backbone, loin and rump. The score were ranged from 1 (skinny) to 5 (too fat) according to Berry *et al.* (2007) procedure.

During collection period, the cows manure were scored according to their consistencies, color and manure screening (Wells 2013). Feces and urine were collected according to Naiket *al* (2014) procedures. The total of feces were weighted, sampled, dried and analyzed for their dry matter and organic matter contents. Urine were collected in 150 l plastic containers. Sample of feces and urine collected were 10% of the total feces and urine weight measured.

The experiments were designed using complete block design with period of lactation as a block. T-test were used to compare the amount of feed and nutrients consumed, digest as well as the cows performances (Steel dan Torrie 1995).

RESULTS AND DISCUSSIONS

The amount of feed and nutrient intakes observed and digestibilities measured in this experiment were shown in Table 2. The results showed that there were no significant impact of the type of the rations on feed and nutrient intakes except for NFE which was slightly higher on the rations contained the silage. They showed that

the rations offered in this experiment of similar qualities. However, their dry matter, organic matter and protein digestions by the cows showed that the silage supplementation were higher than the tofu and cassava waste which resulted higher in TDN values of the rations.

For the most Indonesian dairy farmer, tofu and cassava waste are thought to be the only ingredients for protein and energy supplements to achieve higher milk production. The ingredient however, is too difficult to reach by the farmer at reasonable price per their nutrient units. The cooperative are trying to exchange the used of the waste with other alternatives ingredients. Providing the same quality feeds but better digestibility such as the super-formula silage might be an alternative to the cooperative effort. With the silage technique, farmer can provide higher quality feed for their cows less depending on weather condition (Cavallarin *et al.* 2005).

Table 2. Nutrient Intake (kg/head/day)

Nutrient	Intake			Digestibilities		
	R1	R2	T-test	R1	R2	T-test
Dry Matter	15.03	15.08	0.896	71.51	73.05	0.023
Organic Matter	13.04	13.22	0.855	72.67	74.68	0.044
Crude Protein	2.67	2.74	0.523	77.75	78.39	0.037
Ether Extract	0.58	0.63	0.402	90.98	94.54	0.537
Crude Fiber	3.08	2.84	0.064	65.56	66.54	0.367
NFE	6.80	7.05	0.042	71.93	74.15	0.268
TDN				71.76	73.43	0.034

The higher digestibilities of R2 might be caused by LAB fermentation activity during ensiling which produced acid, weaken hydrogen bond and partly breakdown ligno-(hemi) cellulosic linkages and made other nutrients were more accessible to ruminal microbe fermentation and post ruminal enzymatic digestion. With this condition, it is hoped that R2 will give better impact on the cow's performances because of their higher nutrient available for metabolic and synthesis.

Dairy cow performances on the two type of the rations were shown in Table 3. The results showed that there were no significant dairy performance between the two types of the ration. The R2 cow's tent to produce higher milk production, total solid and milk fat content, and lower milk rejected percentage. The R1 treated cows however, tent to produce higher milk density and solid non-fat (lactose and protein). The higher total solid in R2 were caused by increasing milk fat content of the milk. The higher milk fat in the R2 treated cows milk might be caused by higher feed fiber digested which produced higher acetic acid proportion in the rumen and provide better fat milk precursor for milk fat synthesis. Higher milk fat content in dairy cows fed with silage were also found by Argelet *et al.* (2000), Veliket *et al.* (2000), Nugroho *et al.* (2015), Zahera *et al.* (2015) and Lestari *et al.* (2015).

Improvement of cow's performances tent to occur not only in milk production and milk fat component but also in body weight gain and score and manure score. The improvement of body weight and body score are hoping to be seen in the next lactation period because milking cows store reserves as body tissue for later use as energy sources (Moran, 2005).

Table 4. Dairy cow performances

Parameters	R1	R2	T-test
Milk Production (L/head/day)			
Early research	17.64	18.14	0.874
End research	18.15	18.60	0.976
Rejected (%)	0.51	0.46	0.483
Milk quality (%)			
Fat	2.98	3.74	0.101
Density	1027.92	1026.53	0.313
Lactose	4.28	4.12	0.611
Protein	2.84	2.78	0.482
Solid non fat	7.77	7.63	0.903
Total solid	10.75	11.36	0.347
Body Weight (kg/h)			
Early research	500.11	490.38	0.166
End research	500.00	501.97	0.252
Body Weight Gain	-0.11	11.59	0.115
Body Conditioning Score			
Early research	2.88	2.83	0.754
End research	2.71	2.96	0.331
Alteration	-0.17	0.13	0.243
Manure Score			
Early research	2.88	2.41	0.057
End research	2.59	2.58	0.039

Improvement of manure score were also seen in R2, although the score are still less ideal for dairy cows. The desire score for manure is around 3 with description consistency like porridge, soft pile of 40 – 50 mm high, have several concentric ring and small depression in the middle (Moran, 2005). The manure score is one of indicator to highlight problem with feeding management. Suddenly change of feed composition by giving higher concentrate percentage for this research purpose, reduce manure score as frequently seen in early period of lactation.

CONCLUSIONS

From this research it is concluded that tofu and cassava waste could be replaced by super-formula silage which resulted similar feed and nutrient consumption, higher digestibility and cow’s performances and provide better energy storage to overcome negative energy balance for the next cycle. Super-formula silage provide more sustainable supply of nutrients for high yielding cows in compare to tofu and cassava waste.

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SUBSTITUTION OF COMMERCIAL FEED WITH BEAN SPROUT WASTE ON FEED EFFICIENCY OF GROWING LOCAL MALE RABBITS

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ABSTRACT

Composition and quality of feed are two factors that can affect the digestibility of nutrients in rabbits. Various ingredients have been used in different types of animal feed, including waste from bean sprouts processing. Bean sprout waste is the residual from bean sprouts processing consisted of green peel and bean sprout fraction which is not consumed by human. This research was conducted to evaluate daily feed consumption, performance (daily weight gain), digestibility, and protein efficiency ratio (PER) of rabbits. A total of 12 local male rabbits aged 12 weeks with an initial body weight about 972 ± 156 g were used in this research. Treatments were formulated as follows: commercial pellets 100% as control (R0), 85% R0 with 15% bean sprouts waste (R1), 70% R0 with 30% bean sprouts waste (R2), and 55% R0 with 45% bean sprouts waste (R3). This research was conducted over 8 weeks period. Research results showed that treatments did not significantly ($P > 0.05$) affect consumption of dry matter, organic matter, and crude protein; but significantly ($P < 0.01$) affected crude fat consumption. Average crude fat consumption was: R0 = 2.33 ± 0.52 g, R1 = 1.98 ± 0.50 g, R2 = 2.93 ± 0.09 g, and R3 = 1.08 ± 0.36 g. Likewise, the treatments did not significantly ($P > 0.05$) affect daily weight gain, digestibility of dry matter, organic matter, and crude protein but significantly ($P < 0.05$) affected crude fat digestibility. The average digestibility rate of crude fat was: R0 = $83.30 \pm 4.61\%$, R1 = $84.81 \pm 5.49\%$, R2 = $85.18 \pm 2.98\%$, and R3 = $68.98 \pm 8.38\%$. Treatments also significantly ($P < 0.05$) decreased protein efficiency ratio (PER). Average PER in this study was: R0 = 1.34 ± 0.29 ; R1 = 0.92 ± 0.22 ; R2 = 1.01 ± 0.05 , and R3 = 0.94 ± 0.10 . It can be concluded that utilization of bean sprouts waste at level of 45% can be used as a feed source for rabbits, although it decreases crude fat consumption, crude fat digestibility, and protein efficiency ratio (PER).

Keywords: *Local male rabbit, Bean sprouts, Nutrient digestibility*

INTRODUCTION

The requirement of animal protein in Indonesian people is increasing every year along with the increase of population. People need animal protein source in adequate amount, short acquisition times and good quality. Rabbits can be one of alternative options of animal protein source because it has fast production characteristics and can give birth 4-8 times a year. The other superiority of rabbits is the reproduction cost not too expensive, and rabbit's meat also contains cholesterol lower than the other meats. Moreover, rabbits are potentially to develop a meat production livestock like the other livestock producing meat (lambs, goats, and beef cattle) because the production level is moderately high.

The attention to diets that fed to rabbits become one of priority factors in supporting the rabbit's reproduction. The success of farming is determined by the quality of feed. The fact in the field, many farmers feed the animals without looking at

the quality, quantity and technique in feeding that cause the productivity become less optimal even occurring disadvantage.

Moreover, the availability of feed stuff now facing various constraint. This case is caused by the change of farmland function become housing and industry that reducing the chance of planting forages as a main feed of livestock. Feeding full concentrate also will increase the production cost. The present ration cost of rabbits is moderately expensive because of manufacturer production still less and more tends to rabbit pet ration. To settle the problem need to find an alternative feed supplement that contains proper nutrients, continuously available, and price that can suppress the production cost. The utilization of bean sprouts waste can be a choice. The peel of bean sprouts and the other part are a waste from bean sprouts. The availability is many because not used by people and the nutrient contained is high enough. The result from a survey by Rahayuet *al.* (2010) inform that total production of bean sprouts in Bogor around 6.5 ton/day and had a chance to produce bean sprouts waste around 1.5 ton/day. Bean sprouts waste also had nutrient value that good enough, namely 65.35% water, 7.35% ash, 1.17% fat, 13.62% protein and 49.44% crude fiber contained.

Utilization of a waste become one of feed product that reduce environment pollution load. Bean sprouts waste also can be utilized as a cheap feed stuff source, easily to obtained and not competitive with human needs. This research will conduct a trial of effect of substitution complete feed with bean sprouts waste in rabbits ration, considering the use of bean sprouts waste as rabbits feed still utilized unlike the utilization on ruminants yet.

This research is aimed to know the effect of substitution commercial feed with bean sprouts waste on the efficiency of ration utilization in growing local male rabbits.

MATERIALS AND METHOD

Location and Time

This research was conducted from February to April 2012. The research located in Field Laboratory of Small Ruminants Production Science block B and Laboratory of Feed Science and Technology, Animal Science Faculty, Bogor Agricultural University. Pellets for rabbits was made in CV. Indofeed.

Materials

Animals

Animals used in this research were 12 local male rabbits with 12 weeks of age and weight about 972 ± 156 g. Feed was given as a complete ration pellets.

Cages and Equipment

Cages used in this research were 12 individual cages made from bamboo with size 50x50x50 cm. Each cage had feeding and drinking equipment and feces collecting tools that placed lower on front side in order to let the feces go down to the feces collecting tools. The other equipment that used was digital scale, thermometer for room, digital camera, labels paper, and cleaning tools.

Ration

The ration was commercial complete ration mixed with different level of bean sprouts waste. Complete ration consisted of corns, rice brans, wheat brans, soybean meals, coconut meals, molasses, forages, anti-molds, anti-oxidants, minerals and

vitamins. Feeds and waters was provided ad libitum. Nutrients composition on materials used can be seen on Table. 3 and nutrients composition on treatment ration on Table 4.

Table 3. Nutrients composition of ration materials (100% DM basis)

Analysis	Bean Sprouts waste	Commercial ration
Dry Matter	22.91	88.12
Ash	3.09	9.66
Crude Protein	14.73	19.13
Crude Fiber	42.27	20.09
Crude Fat	0.11	3.37
Nitrogen free extract	39.80	47.75

Source: Proximate results from Laboratory of Feed Science and Technology (2012)

Table 4. Nutrients composition on treatment ration (100% DM basis)

Analysis	R0	R1	R2	R3
Dry Matter	88.12	85.82	85.83	84.76
Ash	9.66	9.02	7.92	7.03
Crude Protein	19.13	17.94	16.54	15.95
Crude Fiber	20.09	25.08	26.89	30.49
Crude Fat	3.37	2.71	2.81	1.13
Nitrogen free extract	47.75	42.25	45.84	45.40

Source: Proximate results from Laboratory of Feed Science and Technology (2012)

Procedure

Cage Preparation

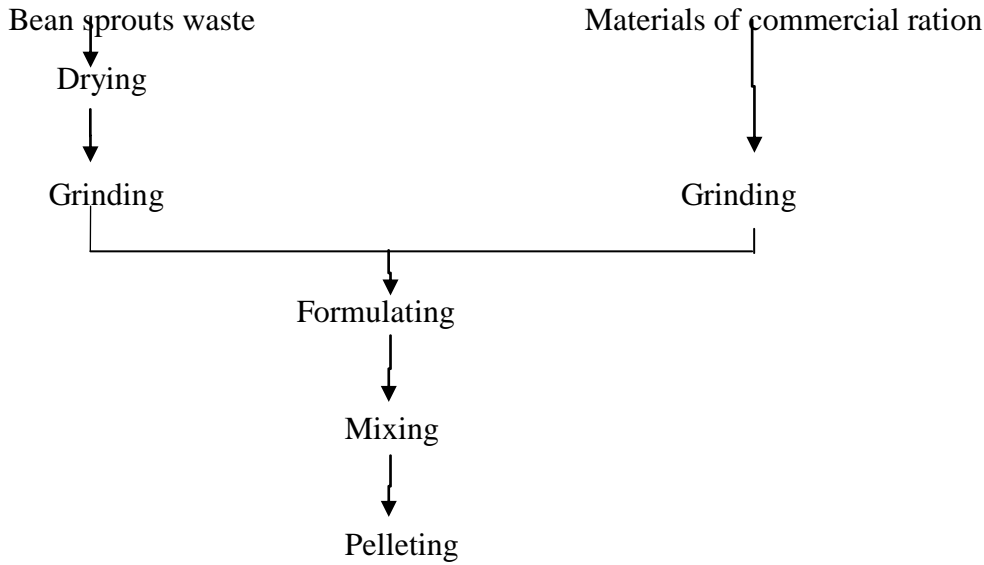
Cage preparation did with repairing the cage to keep the rabbits comfort. Then, the cage was flushed with water, brushed with detergent, and flushed again with water. Disinfectant also given to the cage to clear the odor and prevent dangerous microbe residual from the last maintenance period.

Complete Ration Production

Bean sprouts were collected and dried, then the dried bean sprouts were finely ground. Materials from complete rations also ground and mixed according to the treatments and filled in mixer for mixing process. After all the materials were homogenously mixed, the mixed ration were filled into pellet machine for pelleting process. Machine was cleaned each times the changes of producing different ration. Manufacturing process of complete ration can be seen on Pictures 2.

Maintenance

Maintenance was did for 8 weeks consists of 2 weeks adaptation period and 6 weeks treatments. During the maintenance, rabbits were placed in individual cages. The rabbits were weighed in the beginning of maintenance and weighed again once a week to know the change of daily weight gain. Feed and water were provided ad libitum every day in the morning and afternoon, measuring the feed residue was did in the next day to know the daily consumption. Cleaning the feeding and drinking equipment was did every day to keep the rabbits health.

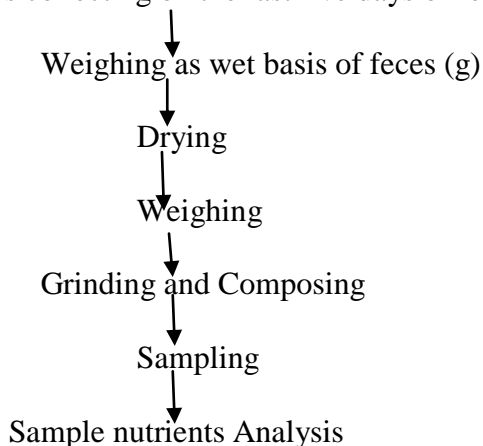


Picture 2. Manufacturing Process of Complete Ration Pellets

Measuring Nutrients Digestibility

According to Mc. Donald et al. (2002), collecting the feces was done for five days to know the nutrients contain on feces and calculating the levels of nutrients digestibility. On the research, feces collecting was did for five days in the last week of maintenance. The feces were collected daily and keep not mixed with urine. Feces weighed, dried with solar thermal and weighed again after dried. Feces were composed and ground, the sample was taken for laboratory analyzed. Then, the sample of feces was analyzed with proximate analysis to know the nutrient contained. Nutrients contained analyzed were dry matter, ash, crude protein and crude fat. The scheme of collecting process of feces sample for analysis can be seen on Pictures 3.

Feces collecting on the last five days of research



Pictures 3. Collecting Feces for Analysis

1. Dry Matter Measuring (AOAC, 1990)

3 g of sample were weighed and dried on 105⁰C Oven for 8 hours. The sample was taken out and put into Desicator/Exicator, 15 minutes later the sample was weighed. Percentage of dry matter contain can be known after the moisture contain calculated with calculating formula:

$$\% \text{ Moisture} = \frac{\text{Early sample weight (g)} - \text{Oven Sample Weight (g)}}{\text{Early sample weight (g)}} \times 100\%$$

$$\% \text{ Dry Matter} = 100\% - \% \text{ Moisture}$$

2. Ash/Minerals Measuring (AOAC, 1990)

3 g of sample were dried in tanure on 800⁰C temperature for 1 hour until being white grey. Sample was taken out and put into exicator, 15 minutes later sample was weighed. Calculating the percentage of Minerals/ash contain was:

$$\% \text{ Ash} = \frac{\text{Early sample weight (g)} - \text{Tenure Sample Weight (g)}}{\text{Early sample weight (g)}} \times 100\%$$

3. Crude Protein Measuring (AOAC, 1990)

0.25 g of samples were weighed, then put into 30 mlKjeldhal tube. Then, 1 spoon of catalis (NaOH, Na₂SO₄, CuSO₄ and Selen). Add 20 ml of 97% H₂SO₄ .Samples were boiled (destruction) for 1 to 1.5 hours until the liquid becomes clear, then cooled. Kjeldahl tube’s content were transferred into a distillation equipment before filled with 10-15 ml water. Then, the tube was washed and rinsed for 5-6 times with 5-10 ml water. Washing water was put into the distillation equipment and add 100 ml of 40% NaOH solution. Erlenmeyer tube was prepared, then filled with 10 ml of 0.1 N H₂SO₄ and 4-5 drops of thymol blue (a mixture of 2 parts of 0.2% methyl red in alcohol with 1 part of 0.2% methylene blue in alcohol). The Erlenmeyer tube was placed under the condenser. The condenser tube’s ends must be submerged in a solution of 0.1 N H₂SO₄ which was added with thymol blue. Furthermore, the distillation was did to obtain approximately 15 ml of distillate in Erlenmeyer. Distillate from the distillation subsequently titrated with 0.2 N HCl standard solution until the endpoint was marked by a color changed from dark blue to green bluish. Protein content determined by following equation:

$$\% \text{ PK} = \frac{(\text{ml sample} - \text{ml blanko}) \times N \text{ NaOH} \times 14 \times 6.25}{\text{sample (mg)}} \times 100\%$$

4. Crude Fat Measuring (AOAC, 1990)

Fat tube was dried in a dryer at 105-110 ⁰C of temperature for 1 hour, then cooled in desiccator and then weighed. Approximately, 1 g sample was wrapped in filter paper and treated cotton, and put in soxhlet extraction tool that already contains diethyl ether. Reflux carried out for 5 hours and the solvent was distilled in fat tube. Furthermore, fat tube containing the extracted fat was heated in an oven at 105 ⁰C of temperature. Once dried until reach the constant weight and cooled in a desiccator, the tube with fat were wighed. The fat contain was determined by following formula:

$$\% \text{ LK} = \frac{\text{Fat tube after extraction (g)} - \text{Empty fat tube (g)}}{\text{early sample weight (g)}} \times 100\%$$

Data Analysis and Design

Treatments

This research used four different feeding treatments with three replications:

- R0 = 100% commercial ration (control)
- R1 = 85% R0 + 15 Bean sprouts waste
- R2 = 70% R0 + 30% Bean sprouts waste
- R3 = 55% R0 + 45% Bean sprouts waste

Experimental Design

The experimental design used was a randomize block design with four treatments and three groups. The group was based on the rabbit body weight, there were small, medium, and large weight. Mathematical models of experimental design was:

$$Y_{ij} = \mu + \tau_i + \beta_j + \epsilon_{ij}$$

Information:

Y_{ij} : Observation on the treatment to-i and the group to-j

μ : The general average score from observation

τ_i : The treatment effect to-i

β_j : The grouping effect to-j

ϵ_{ij} : The random effect on treatment to-I and group to-j

Data Analysis

The data obtained from observation were analyzed using Analyses of Variance (ANOVA), if different between treatments then tested with Duncan (Steel and Torrie, 1993).

Variables Measured

Consumption (g)

Consumption is the amount of feed that enters into livestock body and were used to meet the basal requirements and animal production purposes. Daily feed intake was calculated by reduction between the feed was given with residual feed (g/head/day). Feed nutrients intake was calculated by multiplying the total consumption with feed nutrient percentage in ration. Feed nutrients intake measured were dry matter, organic matter, crude protein and crude fat intake, calculated by following formula:

$$\text{Feed nutrients intake (g/head/day)} = \text{consumption (g)} \times \% \text{ feed nutrients}$$

Digestibility (%)

Ration digestibility measured were, dry matter, organic matter, crude protein and crude fat with following formula:

$$\text{DM digestibility (\%)} = \frac{\text{Ration DM intake (g)} - \text{feces DM (g)}}{\text{Ration DM intake (g)}} \times 100\%$$

$$\text{OM digestibility (\%)} = \frac{\text{Ration OM intake (g)} - \text{feces OM (g)}}{\text{Ration OM intake (g)}} \times 100\%$$

$$\text{CP digestibility (\%)} = \frac{\text{Ration CP intake (g)} - \text{feces CP (g)}}{\text{Ration CP intake (g)}} \times 100\%$$

$$\text{CF digestibility (\%)} = \frac{\text{Ration CF intake (g)} - \text{feces CF (g)}}{\text{Ration CF intake (g)}} \times 100\%$$

Information:

$$\text{DM intake (g)} = (\text{Ration fed} - \text{residual ration}) \times \% \text{ Ration DM}$$

$$\text{OM intake (g)} = (\text{Ration fed} - \text{residual ration}) \times \% \text{ Ration OM}$$

$$\text{CP intake (g)} = (\text{Ration fed} - \text{residual ration}) \times \% \text{ Ration CP}$$

$$\text{CF intake (g)} = (\text{Ration fed} - \text{residual ration}) \times \% \text{ Ration CF}$$

$$\text{Feces DM (g)} = \text{Feces excreted} \times \% \text{ Feces DM}$$

$$\text{Feces OM (g)} = \text{Feces excreted} \times \% \text{ Feces OM}$$

$$\text{Feces CP (g)} = \text{Feces excreted} \times \% \text{ Feces CP}$$

$$\text{Feces CF (g)} = \text{Feces excreted} \times \% \text{ Feces CF}$$

Daily Weight Gain (g/head/day)

Daily weight gain was the increased of rabbit body weight (g/head/day). The increase in weight can be determined by weighing the live weight of rabbits each week by calculating the difference of final week body weight with early week body weight, calculated by following formula:

$$\text{Daily weight gain (g/head/day)} = \frac{\text{final BW maintenance (g)} - \text{early BW (g)}}{\text{The duration of treatments (day)}} \times 100\%$$

Protein Efficiency Ratio (PER)

Protein efficiency ratio (PER) was calculated from daily weight gain divided by feed protein intake during the research, by following formula:

$$\text{Protein Efficiency Ratio (PER)} = \frac{\text{Daily Weight Gain (g)}}{\text{Ration CP intake (g)}}$$

RESULTS AND DISCUSSION

Consumption

Average dry matter, organic matter, crude protein and crude fat intake can be seen on Table 5. Based on the results of ANOVA, show that the group does not provide significant effect ($P > 0.05$) on dry matter, organic matter, crude protein and crude fat intake.

Table 5. Average of Dry Matter, Organic Matter, Crude Protein, and Crude Fat Intake of Rabbits

Intake (g/head/day)	Treatments			
	R0	R1	R2	R3
Dry Matter	69.13 ± 15.38	72.82 ± 18.48	104.41 ± 3.15	95.26 ± 31.13
Organic Matter	62.45 ± 13.90	66.26 ± 16.81	96.13 ± 2.90	88.56 ± 29.87
Crude Protein	13.23 ± 2.94	13.07 ± 3.32	17.27 ± 0.52	15.19 ± 5.12
Crude Fat	2.33 ± 0.52 ^{AB}	1.98 ± 0.50 ^B	2.93 ± 0.09 ^A	1.08 ± 0.36 ^C

Information: small alphabet of superscript value different on the same line show significantly very different ($P < 0.01$). 100% Commercial Complete Pellet Ration (R0), 85% R0 + 15% Bean Sprouts Waste (R1), 70% R0 + 30% Bean Sprouts Waste (R2), 55% R0 + 45% Bean Sprouts Waste (R3).

DM Intake

ANOVA results also show that the use of bean sprouts wasted on rabbit ration do not provide significant effect ($P > 0.05$) on dry matter intake. The higher use of bean sprouts waste in ration tends to increase dry matter intake. The consumption tends to increase on the ration mixed with bean sprouts waste was suspected because the bean sprouts waste had a higher palatability than the commercial complete ration. Bean sprouts waste can be used as forage feed because it has a high fiber content and

good protein content, meanwhile grain feed usually use as concentrate feed on rabbits. De Blas et al. (1981) stated that the ration dry matter intake increased by 2.97 g/head/day for every one percent increase in the fiber content. High crude fiber content in bean sprouts waste ration will speed up the flow rate of feed in the digestive tract. The faster flow rate of feed in the digestive tract, the digestive process become less than optimal. Undigested feed will quickly excreted and cause livestock consume the new feed is increasing due to the digestive tract was empty. Hoover and Heitmann (1972) report that rabbits were fed with ration contains Acid Detergent Fiber (ADF) 29.40% had a flow rate higher than ration contains 14.73% ADF with each time comparison 6.21 hours and 7.01 hours.

Based on the consumption data on Table 5 , it seen a decrease of dry matter intake in the ration with bean sprouts waste level of 45% (R3), although statistically not show a significantly difference with the higher composition of bean sprouts waste in ration. The decrease of dry matter intake on ration with bean sprouts waste level of 45% suspected due to bean sprouts waste physical nature is bulky and cause rabbits tend to quickly feel full because the digestive tract is full. Moreover, the quickly filled digestive tract can also be caused by rabbit's stomach capacity which is not too big. This conditions will suppress appetite and reducing dry matter intake. The average of dry matter intake based on body weight in this research ranged from 5.10% until 7.29% with total average is 6.37% of live weight. It shows that dry matter intake of this research has been meet the needs of dry matter on rabbits based on Irlbeck (2001), rabbits will eat about 5% of body weight. Meeting the needs of livestock became a very important concern and need to make it suitable with the needs in order to the livestock can grow well.

Organic Matter Intake

The results of ANOVA also show that the use of bean sprouts waste in rabbits ration do not provide a significant effect ($P>0.05$) on organic matter intake. It shows that the use of bean sprouts waste until 45% in rabbits ration have an organic intake equal with commercial complete ration. Organic matter intake of bean sprouts waste ration equals with commercial complete ration caused by organic matter content in bean sprouts waste ration is lower than commercial complete ration so that the increasing of dry matter intake will not cause organic matter intake increase. Organic matter intake data on Table 5 statistically not show a significant differences but provide the increasing pattern of consumption which is equal with dry matter intake, organic matter intake tends to increase on ration with the higher bean sprouts waste level in ration ($P<0.20$). This case is caused by organic matter intake is the largest part of dry matter intake, so the dry matter intake tends to increase on ration with high beans sprouts waste content also will cause organic matter tends to increase with the increasing of bean sprouts waste composition in ration.

Based on the average of organic matter intake data on Table 5, the average of organic matter intake from all treatments is 78.35 g/head/day. The results on this research is higher than the report from Pinheiro *et al.* (2009), that is 57% g/head/ day with complete ration high fiber content treatment. The high intake of organic matter in this research caused by the dry matter along the research also high. The average of dry matter intake in this research is 85.40% g/head/day higher than the results of Pinheiro *et al.* (2009) research, 63 g/head/day.

Crude Protein Intake

The result of ANOVA also show that the use of bean sprouts waste in rabbits ration do not provide significant effect ($P>0.05$) on crude protein intake. The average of crude protein intake in this research can be seen on Table 5 range from 13.07-17.27 g/head/day. The result of this research is higher than reported by Lestari (2004), range from 7.93-10.29 g/head/day with complete ration mixed with soybean seed coat. Crude protein intake in this research is higher due to the average dry matter intake in bean sprouts waste ration range from 69.13-104.41 g/head/day higher than complete ration mixed with soybean seed coat which is 49.14%-58.19%. Moreover, bean sprouts waste also has a good quality, it has 14.73% protein content higher than protein content of soybean seed coat based on Lestari (2004) is 11.62%.

Crude Fat Intake

The use of bean sprouts waste in rabbits ration provide a very significant effect ($P<0.01$) on crude fat intake. Crude fat intake of rabbits was fed with bean sprouts waste level of 30% (R2) on ration, significantly higher than ration with bean sprouts waste level of 15% (R1) and 45% (R3), meanwhile crude fat intake on commercial complete ration (R0) is not significantly different to ration with bean sprouts waste level of 15% (R1) and 30% (R2). Bean sprouts waste level of 30% (R2) in ration with 2.81% crude fat content lower than commercial complete ration (R0) which is 3.37%, however results same fat intake. The equal fat intake caused by dry matter intake that tends to increase in ration with bean sprouts waste level of 30% (R2) than the commercial complete ration (R0). The higher consumption, nutrient content consumption also increase. Crude fat intake on ration with bean sprouts waste level of 15% (R1) lower than ration with bean sprouts waste level of 30% (R2) caused by crude fat intake and content of ration are low, meanwhile crude fat intake in ration with bean sprouts waste level of 15% (R1) equal with commercial complete ration (R0) because it has a higher intake although crude fat content of ration is low.

Digestibility

Digestibility level is a value that can describe nutrients absorbed in livestock body. Many factors can affect the level of feed digestibility in livestock. According to Campbell *et al.* (2003), the factors that affect the ration digestibility including the physical form of feed, feed flow rate as it passes through the digestive system and nutrient composition of feed. Digestibility average percentage of dry matter, organic matter, crude protein and crude fat can be seen on Table 6. Based on the results of ANOVA show that group does not provide significant effect ($P>0.05$) on dry matter, organic matter, crude protein and crude fat digestibility.

Dry Matter Digestibility

The results of ANOVA show that the use of bean sprouts waste in rabbits ration do not provide significant effect ($P>0.05$) on dry matter digestibility. This case show that the use of 15%, 30%, and 45% level of bean sprouts waste in rabbits ration have digestibility level of dry matter equal with commercial complete feed. Dry matter digestibility is equal caused by high feed flow rate in ration mixed with bean sprouts waste so the consumption tends to increase not significant to increase dry matter digestibility. The higher crude fiber content in ration with bean sprouts waste will increase consumption and influence the retention time of feed in digestive tract. This case is causing feed flow rate increase and the chance to be degraded also become less.

Table 6. Average Percentage of Dry Matter, Organic Matter, Crude Protein and Crude Fat Digestibility of Rabbits

Digestibility (%)	Treatments			
	R0	R1	R2	R3
Dry Matter	59.79 ± 4.67	57.88 ± 5.90	51.71 ± 6.59	51.64 ± 3.20
Organic Matter	60.54 ± 5.50	56.53 ± 5.91	49.28 ± 6.76	48.90 ± 5.59
Crude Protein	70.34 ± 3.85	69.09 ± 3.22	61.05 ± 3.27	61.79 ± 4.18
Crude Fat	83.30 ± 4.61 ^a	84.81 ± 5.49 ^a	85.18 ± 2.98 ^a	68.98 ± 8.38 ^b

Information: small alphabet of superscript value different on the same line means significantly different (P<0.05). 100% Commercial Complete Pellet Ration (R0), 85% R0 + 15% Bean Sprouts Waste (R1), 70% R0 + 30% Bean Sprouts Waste (R2), 55% R0 + 45% Bean Sprouts Waste (R3).

Based on average data of dry matter digestibility on Table 6, average digestibility of dry matter from all treatments is 55.26%. Dry matter digestibility in this research lower than reported by Zulharman (2010), which is 59.48% in local male rabbits which fed with complete ration silage with 23% ration crude fiber content. Dry matter digestibility on this research is lower because crude fiber content of bean sprouts waste is higher with the average of all treatments is 25.63%. High crude fiber content in ration caused feed quickly go out from digestive tract so feed not completely digested and decrease the dry matter digestibility. The result of this research still in normal level of dry matter digestibility on pellet ration shaped mixed with bean sprouts waste based on Cheeke (1987) statement, dry matter digestibility of rabbits fed with complete ration pellet shaped is 45%. This case show that ration mixed with bean sprouts waste had digestibility level which is good enough. The higher digestibility will increase dry matter intake of ration and higher adequate of feed nutrient.

Organic Matter Digestibility

The results of ANOVA also show that the use of bean sprouts waste in rabbits ration do not provide significant effect (P>0.05) on organic matter digestibility. Ration mixed with 15%, 30% and 45% level of bean sprouts waste do not effect organic matter digestibility. This case show that the use of bean sprouts waste in rabbits ration had organic matter digestibility equal with commercial complete ration. Organic matter digestibility will be in a line with dry matter digestibility, the same dry matter digestibility between commercial complete ration with ration mixed with 15%, 30%, 45% level of bean sprouts waste cause organic matter digestibility also not significantly different. Organic matter is the largest component in dry matter, including crude protein, crude fat, crude fiber, and Nitrogen Free Extract. The increasing of dry matter digestibility in ration mixed with bean sprouts waste will increase the digestibility of organic matter conversely.

Based on the data of organic matter digestibility on Table 6, average digestibility of organic matter from all treatment is 53.81%. Organic matter digestibility in this research is lower than reported by Zulharman (2010), which is 61.66% in local male rabbits fed with complete ration silage with 23% crude fiber content of feed. The low organic matter digestibility in this research is suspected because the nutrients content in ration. Bean sprouts waste ration contains fiber higher than complete ration silage. The average of crude fiber in ration in this research is higher, which is 25.63%. High crude fiberin ration will cause feed no

longer feed stuck in digestive tract and decrease organic matter digestibility. The small capacity of stomach also will not allow rabbits to retain amount large of feed so the high fiber of feed will go out quickly and decrease digestibility. Moreover, Rabbits include the hindgut fermenters, the rear digestive tract has an important role in digestive system of rabbits such as caecum and colon.

Crude Protein Digestibility

The results of ANOVA also show that the use of bean sprouts waste in rabbits ration do not provide significant effect ($P>0.05$) on crude protein digestibility. The use of 15%, 30%, and 45% level of bean sprouts waste in rabbits ration do not provide significant effect ($P>0.05$) on crude protein digestibility of rabbits. The lower crude protein content in ration with the increase used of bean sprouts waste in ration do not influence the crude protein digestibility. This case suspected due to the nature of coprophagy in rabbits. Rabbits consume again the soft feces which high of water, nitrogen and electrolyte content. This characteristic allow rabbits to utilize protein properly from bean sprouts waste ration with high crude fiber contain. Feces that consumed will help bacteria in the rear of digestive tract produce microbial protein qualified and increase protein digestibility, although in high fiber ration. This case appropriate with the statement of Blakely and Bade (1991) who said that coprophagy allow the rabbits to completely utilize bacterial digestion in the rear of digestive tract, convert forages protein to high quality microbial protein, synthesize vitamin B, and break down the cellulose or fiber into a useful energy. Green peas had a similar amino acid structure with soy beans. The better quality of protein with complete and balance amino acid contents, the higher digestibility of protein. Green peas had high enough protein digestibility value however protein digestibility is effected by tripsin inhibitor present. The decreasing of protein digestibility in ration mixed with bean sprouts waste is suspected by tripsin inhibitor effect, however there is no difference about digestibility level in ration with higher level of bean sprouts waste. This case caused by coprophagy nature so rabbits may utilize protein from bean sprouts waste ration as well as commercial complete ration in lower protein content and high crude fiber. Gracia *et al.* (1993) stated that crude protein digestibility is effected by crude fiber and protein level of ration.

Based on the data of average crude protein digestibility on Table 6, average crude protein digestibility of all treatments is 65.57%. Crude protein digestibility in this research had a digestibility level not far difference compare with the research of Carabano *et al.* (1997), amount 67.30% with Rabbits feed alfafa based and ration protein is higher as much as 23.77%. Feed composition could be one of factors that determines protein digestibility to a certain feed. Crude protein digestibility on ration mixed with bean sprouts waste is good enough can caused by protein content of ration meets the needs of rabbits, that is range from 15.96%-19.13%. Based on NRC (1977), crude protein needs of growing rabbits is 16%.

Crude Fat Digestibility

The use of bean sprouts waste on rabbits ration provide significant effect ($P>0.05$) to crude fat digestibility. The use of bean sprouts waste until 45% in rabbits ration effected crude fat digestibility. The results in this research show that ration with 45% bean sprouts waste level (R3) had crude fat digestibility lower than ration with 15% bean sprouts waste level (R1), 30% (R2) and commercial complete ration (R0), however there is no significant differences of crude fat digestibility on ration

with 15% beans sprouts waste level (R1), 30% (R2) and commercial complete ration (R0). The low crude fat digestibility of ration with 45% bean sprouts waste level (R3) is caused by low fat contents in ration (1.13%) and high crude fiber (30.49%). High crude fiber affect metabolism process in digestive tract, the higher fiber contents will cause feed particle pushed away from digestive tract and more energy needed to break down the crude fiber become a simple compound. The low energy in body cause digestive enzyme production include lipase enzyme decrease and reduce the ration’s crude fat digestibility. The same crude fat digestibility between commercial complete ration, ration with 15% and 30% bean sprouts waste level are suspected because rabbits still tolerant with each ration’s fiber content are 20.09%, 25.08% and 26.89%. Energy inside the body still adequate to produce digestive enzyme so the ration’s fat content can be utilized properly inside the body. Templeton (1968) stated that the optimal crude fiber level for rabbits ranged from 20%-27%.

The average of crude fiber in this research ranged from 68.98%-85.18% (Table 6). The result of this research that reported by Yahya (2002) is ranged from 86.9%-89.15% with complete ration mixed with soybeans seed. The low crude fat digestibility on this research is caused by low crude fat contents of ration treatment and higher crude fiber. Mathius *et al.* (2001) stated that feed stuff digestibility effected by livestock’s age, feeding level, processing and feeding method, feed compositions and feed nutrients level contained. The lower feed nutrients content in ration, the lower feed nutrients digestibility.

Coefficient fat digestibility on this research had a higher value than the other feed nutrients. Ration mixed with bean sprouts waste is ration from forages and grains that generally are unsaturated fat. Fat will hydrolyzed by lipase and produce glycerol and fatty acid. Rabbits will become active to digest fat from feed in digestive tract and then utilized as energy. De Blas and Wiseman (2010) stated that fat digestibility on rabbits had a same value with others nutrients, however it has variation depend on age. Fat digestibility in lactating rabbits is bigger than growing rabbits are amount 64% and 58%.

Body Weight Gain

Livestock growth can be seen from body weight that produce. A high body weight gain show that livestock is growing well. Smith and Mangkoewidjojo (1988), stated that growth rate of 8 until 26 weeks rabbits is 100-150 g/week. The average of rabbits weight gain can be seen on Table 7.

Table 7. Average of weight gain in Rabbits

Variable	Treatments			
	R0	R1	R2	R3
PBB (g/head/day)	17.14 ± 1.45	12.14 ± 5.16	17.40 ± 0.70	14.21 ± 4.62
Information:	100% Commercial Complete Pellet Ration (R0), 85% R0 + 15% Bean Sprouts Waste (R1), 70% R0 + 30% Bean Sprouts Waste (R2), 55% R0 + 45% Bean Sprouts Waste (R3).			

Based on the results of ANOVA show that group do not provide significant effect ($P > 0.05$) to body weight gain. The results of ANOVA also show that the used of bean sprouts waste at 15%, 30% and 45% level provide body weight gain same as the used of commercial complete ration. Consumption that tends to increase on bean sprouts waste ration than commercial complete ration do not affect body weight gain.

This case is suspected by the effect of tripsin inhibitor contained in bean sprouts waste that originally from green peals as a main material to produce bean sprouts. Tripsin inhibitor will bind tripsin enzyme (protease) and decrease tripsin enzyme activity. The reduction of tripsin enzyme function causing protein from bean sprouts waste cannot be break down completely become a simple molecule such as amino acid and cannot be absorbed by small intestine and utilized properly by the body for growing. Protein digestibility from green peals is very effected by protein inhibitor presence (Bressani *et al.*, 1982).

Nitrogen Free Extract content in bean sprouts waste ration that lower than commercial ration also can caused no effect on increasing consumption to body weight gain. Moreover, the higher crude fiber content with increasing of bean sprouts waste use will suppress the retention time of feed in digestive tract. Feed is quickly pushed away so feed nutrients do not utilized properly by the livestock body. The high feed flow rate in digestive tract also will affecting protein digestibility, proteins does not utilized properly by the livestock body. Low total of digested nutrients such as crude fiber and this nitrogen free extract causing the energy inside the body unavailable in adequate amount and proteins that available are utilized to meet the needs. The degradation process of proteins become energy can affect the rabbits growing.

The average of body weight gain in this research ranged from 12.14-17.40 (g/head/day). The results of this research still in a normal range based on Cheeke (1987) who stated that rabbits growing in tropical area about 10-20 g/day. Ration mixed with bean sprouts waste do not affect body weight gain although the consumption is increase with higher bean sprouts waste level. A high crude fiber content in bean sprouts waste ration causing the feed flow rate increase and allow the rabbits to consume more to meet the needs. Fekete (1984), stated that rabbits have an ability to adapt the consumption with the energy required. Moreover, antinutrient like tripsin inhibitor that suspected still contained in bean sprouts waste affect the metabolism activity in the body, so the nutrients that contained in ration unutilized properly by the body to grow.

Protein Efficiency Ratio (PER)

Protein Efficiency Ratio (PER) counted from body weight gain divided with feed proteins consumption. PER is used to know the effect of protein on body weight gain. The higher PER value, the better effect of protein consumption to livestock body weight gain. The average of Protein Efficiency Ratio (PER) can be seen on Table 8.

Table 8. The Average of Protein Efficiency Ratio (PER)

Variable	Treatments			
	R0	R1	R2	R3
PER	1.34 ± 0.29 ^a	0.92 ± 0.22 ^b	1.01 ± 0.05 ^b	0.94 ± 0.10 ^b

Information: value with small alphabet of superscript different on the same line means significantly different (P<0.05). 100% Commercial Complete Pellet Ration (R0), 85% R0 + 15% Bean Sprouts Waste (R1), 70% R0 + 30% Bean Sprouts Waste (R2), 55% R0 + 45% Bean Sprouts Waste (R3).

Based on the results of ANOVA show that group do not provide significant effect (P>0.05) to Protein Efficiency Ratio (PER), however the use of bean sprouts waste on rabbits ration affect Protein Efficiency Ratio (PER). The use of bean

sprouts waste in ration at 15%, 30% and 45% level have the same PER. Protein consumption in commercial ration have more efficient effect on body weight gain. This case caused by consumption that tends lower on commercial complete ration than bean sprouts waste ration. Commercial complete ration (R0) with protein of ration is 19.13% and dry matter intake is 69.13 ± 15.38 g/head/day can the same body weight gain as ration with bean sprouts waste treatment on higher consumption. Low PER in bean sprouts waste ration is suspected by there is still tripsin inhibitor contained from green peals so protein less utilized properly compared with commercial complete ration. Moreover, bean sprouts waste contains crude protein lower than commercial complete ration so the increasing of bean sprouts waste use in ration, decrease crude protein content in bean sprouts waste ration.

The effect of antinutrient become one of constraints in utilize bean sprouts waste as a feed stuff. According to nutrients, green peals as a main material to produce bean sprouts waste have a high protein content and amino acid structure resemble to soybeans amino acid structure, but then this nutrients less utilized because the effect of antinutrient contained in green peal. Antinutrient like tripsin inhibitor will affect the nutrient digestibility such as proteins. Some methods to reduce the antinutrients content is by giving treatments on that green peals by soaking, germinating and maturing (Belinda 2009). Producing green peals become a bean sprout is expected can reducing the effect of tripsin inhibitor from green peal. Based on the results that reported by Gervani and Theophillus (1980) that PER from green peal effected by processing, germinating and frying method can reducing PER value, whereas boiling will increasing the PER of green peal. Green peals processing by germinating produce PER amount 1.19. The higher PER value means the more efficient livestock using proteins and at last will affect the growing process.

Saenap (2011) stated that green peals waste have energy content amount 3737 kcal/kg. The energy content of this bean sprouts waste is moderately high, however high crude fiber content causing flow rate in digestive tract also high so the digestive process become less optimal. Low energy that available in the livestock body causing the body break down the reserve energy. Deficiency of energy also will causing available protein converted to energy so the role of protein will reduce. Moreover, other antinutrients content such as tripsin inhibitor can causing nutrients absorbance inside the body. This case is suspected as a cause of the low PER in complete ration mixed with bean sprouts waste. Based on NRC (1977), growing rabbits need energy 2500 kcal/kg and protein 16%. Balance of protein and energy in ration need to consider to meet the needs of rabbits adequate appropriate with the requirements (livestock age, type and species along with livestock production). Moreover, antinutrients contained in feed stuff also had to be considered in order not to influential on nutrient digestive process.

CONCLUSION

The utilization of bean sprouts waste at 15%, 30% and 45% in rabbits ration provide the equal effect with commercial complete ration on consumption and digestibility of dry matter, organic matter, and crude protein however affect the consumption and digestibility of crude fat. Bean sprouts waste ration treatment also not affect the body weight gain but decrease the value of Protein Efficiency Ratio (PER). PER value on bean sprouts waste ration treatment is lower than commercial

complete ration but there is no difference in PER value between ration with bean sprouts waste at 15%, 30% and 45% level.

SUGGESTION

Following research need to conduct about the level of bean sprouts waste utilization in rabbits ration. Bean sprouts waste can be used as one of alternative feed stuff in rabbits.

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THE EFFECT OF DIFFERENT ENERGY LEVELS IN THE DIETS ON MATING ACCELERATION OF LOCAL SHEEP

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ABSTRACT

The objective of this research was to find the appropriate ration energy levels to determine the puberty time of ewes. The study used 12 offspring female sheep obtained from crossing Jonggol ewes of Animal Science Teaching and Research Unit (JASTRU) Faculty of Animal Science IPB with Garut male sheep. The animal age was approximately 2-3 months with the average body weight 9.79 ± 1.97 kg. The treatments were consisted of 3 rations which have different TDN level, namely: T1= 65% TDN, T2=70% TDN and T3= 75% TDN. Completely randomized design was used in this experiment. The measured parameters were feed intake, average daily gain, and the first mating age and body weight. The results indicated that the ration energy levels did not significantly affected on the feed intake, average daily gain, as well as the first mating age and body weight. The age of first mating was 180-203 days with body weight at 18-22 kg. It is concluded that the TDN level between 65% -75% has similar response to accelerate the mating of local sheep.

Keywords: *Energy, acceleration, Mating, Local sheep*

INTRODUCTION

Estrus is one of the important things that will be affected on the success rate of mating sheep. It will indirectly impact on the reproductive efficiency and development of local sheep. The improvement of parent reproduction aspects are not only improve the efficiency of biological livestock, but also increase the production efficiency of livestock business (Dickerson, 1996). Smith and Akinbamijo (2000) stated that there are four main factors that determine reproductive performance of ruminants in the tropics, there are genetic, physical environment, nutrition and management. The levels of energy and protein in the ration will affect the success rate of reproduction. Callaghan et al (2000) suggested that the level of energy consumption will influence to the systemic of hormonal concentrations and follicular fluid. Kusina et al (2001) examined the provision of ration with three different energy levels, namely: low, medium and high on Mashona goats, it showed the reducing expression of estrus, conception, fecundity and twinning rates in goats that consume ration with low energy. Koyuncu and Canbolat (2009) stated that the level of dietary energy supplement at pre-mating period can have a beneficial effect and can be practiced to improve the reproductive performance of ewes. The information of optimal energy content in ration for local sheep and its influence to the speed of estrus and first mating have not been widely available. The objective of this research was to find the appropriate ration energy levels to determine the puberty time of local ewes.

MATERIAL AND METHODS

Animals and diets

Twelve female local sheep used in the experiment with an initial age of 2-3 months old and bodyweight 9.79 ± 1.97 kg. Animals were housed on individual cages. The female lambs were fed diets containing the different level of TDN, namely T1=65% TDN, T2=70% TDN and T3=75% TDN. The Ingredient composition of the experimental diets are presented on Table 1 and nutrient compositions of the diets shown in Table 2. Sheep were freely allowed to feed and water. The feed was given twice at around 06.00 pm and 14.00 pm. Concentrates and forage were given separately. Body weight gain was obtained by weighing scale every two weeks.

Puberty detection and mating in female lambs

The Estrus detection was carried out after the lamb brood stock achieve 60% of mature body weight (13-18 kg), or the age of 5-6 months. Estrus detection was done by using a ram teaser. The teaser was released in the cage after feeding in the morning and afternoon. Based on the teaser detection, the female sheep were removed from individual cages to make sure whether female sheep were estrus. The female sheep was already considered estrus in case it was silent when it was ridden by teaser during mating. The onset of first estrus was used as an indicator for the onset of puberty. Date of onset at the first estrus was recorded for each female lamb and considered as an indicator for pubertal age. After the mating date has recorded, the sheep were immediately weighed to determine the body weight at the time of mating.

Table 1. The Ingredient composition of experimental diets (% dry matter)

	Experimental diets		
	T1	T2	T3
	-----%-----		
Native grass	40	40	30
Yellow corn	11	7.4	32
Cassava meal	14.1	15	12
Coconut meal	31.1	31	21
Urea	0.4	1.0	1.1
Crude palm oil	0.0	2.0	2.2
Mollases	0.0	2.0	1.0
CaCO ₃	2.9	2.0	1.0
Dicalcium Phosphate	0.0	0.2	0.3
NaCl	0.3	0.2	0.1
Premix	0.2	0.2	0.1

Experimental Design

Completely randomized design (CRD) with 3 treatments and 4 replications was used on this experiment. The treatments were: T1 = ration with TDN 65%, T2 = ration with TDN 70% and T3 = ration with TDN 75%.

Parameters

The measured parameters were feed intake, nutrient intakes, average daily gain, feed efficiency, proportion of weight change, as well as the age and body weight of first mating.

Table 2. The nutrient composition of experimental diets based on dry matter

Nutrient	Experimental diets		
	T1	T2	T3
Dry Matter (%)	89.37	88.62	88.37
Ash (%)	9.15	10.38	7.01
Ether Extract (%)	8.42	10.43	7.36
Crude Protein (%)	14.60	17.90	16.32
Crude Fiber (%)	13.44	13.86	11.35
Calcium (%)	1.29	0.85	0.65
Phosphorus (%)	0.11	0.06	0.12
TDN (%)	67	70	73

Note: Analysis by Laboratory of Science and Feed Technology, IPB (2011)

Statistical analysis

Data were analyzed using ANOVA (Analysis of Variance / ANOVA). If the analysis results showed the significant differences, it will be tested using Orthogonal Contrast (Steel and Torrie, 1993).

RESULT AND DISCUSSION

The consumption of Grass, concentrate, dry matter and nutrients on local sheep with different level energy diets are showed in Table 3.

Table 3. The grass, concentrate, total dry matter and nutrients intakes on local sheep with different level of energy ration

Parameters	Experimental diets		
	T1	T2	T3
Grass (g/h/day)	149.85	151.50	122.27
Concentrate (g/h/day)	300.4	312.37	394.94
Total Dry matter (g/h/day)	450.29 ± 100.76	463.87 ± 70.30	517.21 ± 115.13
Dry matter intake (%BW)	3.2	3.26	3.49
tio of Roughage:concentrate	33:67	32 : 68	24 : 76
Crude fat (g/h/day)	39.43	51.27	36.41
Crude protein (g/h/day)	67.08	86.84	86.55
Crude fiber (g/h/day)	55.63	59.00	53.16
Calcium (g/h/day)	6.32	4.24	3.55
P (g/h/day)	0.55	0.29	0.63
TDN (g/h/day)	306.53	333.32	390.51

The energy levels of ration had not significant different affected on the consumption of grass, concentrate and total dry matter. The amount of DM intake ration was ranged from 450.29 to 517.21 g / head / day, or 3.20% - 3.49% of body weight. . This is due to the way of sheep to obtain the feed was based on the physical nature and composition of the feed nutrients which are similar. Parakkasi (1999) stated that the factors affecting feed intake of dry material in ruminants included physical nature and chemical composition of feed.Result is still in line with the recommendation of NRC (1985), that the sheep weighing 10-20 kg requires dry matter on 3-5% of body weight. The ration with 65%, 70% and 75% of TDN content could provide the sufficient dry matter to growth ewes

Consumption of crude protein (CP) and TDN of treatment T1 is lower than T2 and T3 of treatment, although not statistically significant. T2 and T3 of treatment have the PK consumption value almost equal, but the value of TDN consumption is lower than T3. Total consumption of Digestible nutrients is similar to the research conducted by Rianto et al. (2006), and lower than the NRC recommendations (1985) the sheep weighing 10-20 kg live crude protein should consume approximately 127-167 g / head / day and TDN of about 400-800 g / head / day. The consumption of crude protein and TDN on this research is lower than the recommendation standard requirements of NRC.

Table 4. Mean (\pm SE) of initial body weight and final body weight at first mating

Parameters	Experimental diets		
	T1	T2	T3
Number of lamb	4	4	4
Initial body weight (kg)	9.75 \pm 2.90	9.87 \pm 1,70	9.75 \pm 1,71
Average. body weight gain (g/h/day)	82.74 \pm 10.85	90.33 \pm 21.99	104.87 \pm 13.04
Feed efficiency	0.18 \pm 0.17	0.19 \pm 0.11	0.20 \pm 0.10
Proportion of weight change (%)	88	101	126
Body weight at first mating (kg)	18.37 \pm 3,27	19.87 \pm 1,93	22 \pm 2.94
The age of first matting (mounth)	6.2 \pm 0.39	6.6 \pm 0.55	6.8 \pm 0.12

The data showed (Table 4) that T1 and T2 and T3 have not significantly different on average body weight at puberty. However, average body weight at puberty tended to be slightly increased with the increasing of energy level on ration. The achieved sheep daily gain on this research similar to Prawoto et al. (2001) that found the improvement of body weight gain on local sheep reaches 57-132 g / head. Chelikani et al.(2003) stated that both of average daily gain and body weight are contributed to 96% of the variation in attaining puberty. Shirley et al. (2001) found that the heavier ewes tended to produce more growth hormone (GH) attained puberty earlier than lighter weight ewes. Wells et al. (2003) suggested that thyroxin (T4) may be an indicator of onset of puberty because T4 rises gradually from the low concentrations just before the onset of breeding season to the peak concentrations just before the transition to the estrus. EI-Saidy et al. (2008) suggested that female lambs growing at the faster rates exhibited their first estrus and most likely conceive at a younger age than ewe lambs growing at slower rate. Furthermore, Ionel et al. (2012) claimed that in general, the onset of puberty takes place in most of the ewes not only at the age of 6–9 months, but also at 3–4 months in breeds that mature earlier (much later than 18–20 months). The difference of energy content in the ration did not give a different effect on the age of first matting. This suggests that the levels of energy ration at 65% TDN is sufficient to accelerate the puberty of local sheep.

CONCLUSION

It is concluded that the TDN level between 65% -75% has similar response to accelerated the mating of local sheep.

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FEED AND NUTRIENT INTAKES OF MARES (*Equus caballus*) GIVEN FARMER’S FEED IN PRIMA FIT FARM BOGOR

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ABSTRACT

Prima Fit Farm in Bogor is a commercial farm keeping race horses with good genetic potential. Some of the milk produced is sold as traditional medicine with high price and this is good opportunity. Feed consumed has great impact on animal production, but there is limited information about feed and nutrient intakes of mares eating feeds provided by farmer. Therefore, this study was conducted to determine effect of feed given by farmer on feed and nutrient intake of mares. Three months observation was carried out; feeds given were field grass, wheat bran, and commercial pellet (Vital). Lactating mares used were two Thoroughbred descendant and three local breeds. Variables measured were feed and nutrient intakes, and body weight estimated from the chest width and body length; data were analysed descriptively. Results showed that Thoroughbred descendant mares had heavier body weight than local breed mares. Mares consumed all commercial pellets given (3.00 kg/head/day) with field grass and wheat bran gave anintakes of 7.68 and 2.20 kg/head/day. Average total dry matter intake was 6.37 kg/ head/day (86.76 kg/metabolic body weight/day). Based on metabolic body weight, Thoroughbred descendant mares consumed less feed and nutrients than local breed mares. It is concluded that differences between the breeds occurred in field grass and wheat bran intakes related to its body weight, but each nutrient was consumed at similar level on the basis of DM intake.

Keywords: *Feed, Nutrient, Intake, Mare*

INTRODUCTION

Prima Fit Farm in Bogor is a commercial farm keeping race horses with good genetic potential. The horses kept in Prima Fit Farm are Indonesian local horse, and Indonesian race horses having Thoroughbred blood descendant. These Indonesian race horse (Thoroughbred descendat horse) are similar to those kept in Pamulang Equestrian Centre (Destiawan, 2010). The horses in Prima Fit Farm are maintained under intensive management (cages, health, feeding, *et cetera*). Each horse is kept in an individual cage; the mares that had been given birth are kept with the foals until weaning or until the foal reaching one year old. Some of the milk produced are given to the foals, but some are sold as traditional medicine with high price and this is a good opportunity. As traditional medicine, mare milk is believed to have bioactive compound that can cure diseases relating to gastrointestinal tract, lungs, renal, kidney, anemia, avitaminosis, *et cetera* (Hermawati, 2005); casein in mare milk was easier to be digested than that in cow milk preventing any problems in digestive tract (Morel, 2003).

Keeping the mares under intensive management also means that the farmer has done a good feeding management practice by giving feeds in a scheduled time and in

a certain ratio or weight according to the condition of the mares, i.e. non pregnant, pregnant, non lactating and lactating mares. This feeding management becomes an important factor controlling mare performances (Huntington, 2012). Feed intake, the horse breed and body condition of mare are important factors affecting milk production and composition (Sudarwanto *et al.*, 1998; Legowo, 2002; Huntington, 2012). Although feed consumed has great impact on animal production, there is a limited information about feed and nutrient intakes of mares eating feeds provided by a farmer. Therefore, this study is conducted to determine effect of the amount of feeds given by farmer on feed and nutrient intakes of mares.

MATERIALS AND METHODS

Materials

There were five lactating mares at the time of observation that were three local mares and two Thoroughbred - local crossbred mares (Thoroughbred descendant). The local mares were one Sandel horse, and two Tokol horses (a and b). The two crossbred mares were the third generation crossbred horse (G3) and Indonesian race horse (kuda pacu Indonesia, KPI). Lactation period of all mares were in the middle and end of lactating period (Table 2); no data available for the beginning of lactating period.

Feeds consisted of field grass, pollard and a commercial concentrate in pellet form with 1.5 cm length and 0.5 cm diameter (Vital brand). This pellet composed of corn, barley, pollard, alfalfa, oats, bran, soybean meal, corn gluten meal (CGM), molasses, vegetable oil, salt, trace minerals, vitamin and mannan oligosacharride. Nutrient composition was analysed following proximate analysis, and data were compared with those of Sutardi (1981), Hernawati (2011) for the field grass and pollard, and the label (Vital) for the pellet (Table 1).

Nutrient composition for field grass and pollard were similar to those of Sutarde (1981) and Hernawati (2011). However, results of proximate analysis indicate that the pellet contained less crude protein and crude fibre, but were high in other nutrient contents than those shown in the label. Vital pellet in this experiment was drier, but was less in crude protein content compared to those analysed by Destiawan (2010), i.e. 87.45% dry matter and 12.16% crude protein contents. TDN and DE for horse was estimated using McDowell *et al.* (1974) formula (in Destiawan, 2010). Field grass had lower energy content than pollard and pellet. Vital pellet had greater energy content than those calculated by Destiawan (2010), 1.97 Mcal DE/kg; this could be due differences in nutrient contents of different batches of Vital pellet.

Procedures

The observation was conducted for three months from June up to August 2012 in Prima Fit, Bogor. Feed nutrient composition was analysed in Pusat Antar University (Inter University Centre, Life Science), Institut Pertanian Bogor, Bogor.

Drinking water was provided *ad libitum*. Grasses were given in the morning (08.30 am) and evening (07.00 pm). Concentrate (pellet) and pollard were given in the morning (05.00 am) and afternoon (03.30 pm); pellet and pollard were mixed with the water before given to the mares. Mares were milked twice a day at 10.00 am and 02.30 pm, but the mares were kept with the foals in an individual cage.

Variables measured were feed intake and body weight and metabolic body weight (MBW). These variables were measured following these procedures :

1. Feed intake were determined by subtracting feed given with feed residue for each type of feeds. Feeds given and its residue were measured using feed balance. Dry matter (DM) intake was calculated from fresh intake multiplied by DM content of each feed, and was then added to obtain total DM intake. Each nutrient intake was calculated from DM intake of each feed which was multiplied by each nutrient content of each feed. Total nutrient intake was obtained by adding the nutrient intakes of field grass, pollard and pellet. Data of total nutrient intake divided by DM intake could be used to indicate percentage of nutrient consumed by the mares.

2. Body weight was calculated based on the chest circumference (hearth girth) and body length. Chest circumference (hearth girth) was measured using measuring band, and body length was measured with measuring stick. Body weight (kg) was calculated following Pilliner (1992) formula : $\{(chest\ circumference)^2 \times body\ length\} / 8717$. Metabolic body weight (MBW) was calculated using this formula : $(body\ weight)^{0.75}$; the MBW data were used to calculate nutrient requirement for maintenance.

Experimental design and data analysis

No experimental design was applied. However, data were analysed descriptively determining the average and standard deviation (Steel and Torrie, 1993).

Tabel 1. Nutrient composition of feed given to mare on dry matter basis

Nutrient composition (%)	Field grass		Pollard		Commercial pellet	
	1)	2)	3)	4)	5)	6)
Dry matter (% fresh)	23.18	24.40	87.52	88.50	88.81	-
Ash	11.94	14.50	5.36	5.90	11.43	9.00
Organic matter	88.06	85.54	94.64	94.04	88.57	79.50
Crude protein	8.53	8.20	13.07	18.46	10.42	11.50
Ether extract	1.67	1.44	3.75	3.88	5.66	2.00
Crude fibre	24.29	31.70	7.93	9.70	11.27	14.50
Nitrogen free extract (NFE)	53.56	44.20	69.89	62.00	61.22	51.50
TDN (%) ⁷⁾	46.32	45.85	56.35	59.36	68.62	53.16
DE (Mcal/kg) ⁷⁾	1.86	1.85	2.23	2.34	2.68	2.11

1), 3), 5) Proximate analysis from the laboratory of PAU (Ilmu Hayati) IPB, 2) Sutardi (1981), 4)Hernawati (2011), 6)Label of commercial pellet (Vital), 7) Estimated from the formula of McDowell *et al.* (1974) for horse in Destiawan (2010): $TDN (\%) = 52,476 + 0,189 (CF) + 3,010 (EE) - 0,723 (NFE) + 1,590 (CP) - 0,013 (CF)^2 + 0,564 (EE)^2 + 0,006 (CF) (NFE) + 0,114 (EE) (NFE) - 0,302 (EE) (CP) - 0,106 (EE)^2 (CP)$. $DE (Mcal/kg) = 0,0365 \times TDN (\%) + 0,172$

RESULTS AND DISCUSSIONS

Mares need nutrients to meet requirements for maintenance, daily activity, reproduction and lactation (Pond *et al.*, 1996; Destiawan, 2010; Huntington, 2012). Feeding needs to fulfill those requirement. The amounts of feeds given were determined by body weight; therefore, it is necessary to determine body weight of mares to be able to estimate its nutrient requirement which related to mare physiological condition (Pond *et al.*, 1996; Huntington, 2012). Nutrient requirement

of lactating mares were also determined by milk production and composition (McDonald *et al.*, 2002).

Table 2 shows that the crossbred and local breed mares had passed their peak milk production; mostly the mares were in the middle of lactation period (3 - 7 months) with milk produced was about 0.57 kg/day for Tokol b (3 month lactation) and 0.74 kg/day for G3 (7 month lactation). The amounts of milk produced could be greater than those measured, the small amounts of milk yield were because of part of the milk was consumed by the foal which was kept with the mare in the same cage. In fact, mare could produced milk per day as much as 3-4% body weight in the first 2 months of lactation, then reduced to 2% body weight in the late lactation with 20 - 25% crude protein content on DMbasis (McDonald *et al.*, 2002; Huntington, 2012). These milk productions indicate that mares secreted significant amount of nutrients (energy, protein, minerals and vitamins) in milks; consequently, lactating mares needed high amounts of nutrients to produce milk, to recover from foaling stress and to rebreed, and good feeding management became important (Huntington, 2012; Anderson, 1994).

Crossbred mares were 5 - 6 years old; two of the local breed had the same age, but one local breed (Tokol b) was the oldest. The crossbred mares had greater body weight and MBW meaning that the crossbred mares had greater body size than those of local breed. Local breed were the result of crossbreeding between local pony horse with Arabian horse (grading up). Within the crossbred mares, G3 were smaller than KPI. Differences in body size of the two crossbred could be due to differences in the blood ratio between Thoroughbred and local breed. G3 had smaller blood ratio of Thoroughbred and greater local breed (87.5% : 12.5%) than KPI (93.75% : 6.25%); G3 was the crossbred between G2 mare with Thoroughbred stallion; KPI was the fourth generation (G4) produced from crossbreeding between local Thoroughbred mare (T4L) and local Thoroughbred stallion (T4L) (Soehardjono, 1990).

Tabel 2. Feed intake and body weight of lactating mare

Horse breed	Lactating month	Age (year)	Body weight (kg)	MBW (kg)	Feed intake (kg/head/day)		
					Grass	Pollard	Pellet
G3	5,6,7	5	393.52	88.35	8.62	2.50	3.00
KPI	4,5,6	6	410.36	91.17	8.65	2.50	3.00
Tokol(a)	4,5,6	3	254.08	63.64	7.00	2.00	3.00
Tokol(b)	3	10	227.17	58.51	7.00	2.00	3.00
Sandel	4,5,6	3	305.67	73.10	7.13	2.00	3.00
Ave			318.16	74.95	7.68	2.20	3.00
± sd			81.73	14.53	0.87	0.27	0.00

First lactation for G3, KPI, Tokol(a) and Sandel. Fourth lactation for Tokol(b)

As a consequence of differences in body weight, crossbred mares consumed more fresh grasses and pollard than local breed (Table 2). However, pollard (2.50 kg/head/day for the crossbred, and 2.00 kg/head/day for the local breed) and pellet were given at the same amounts. The amount of grass given in this experiment was greater, but was lesser in the amount of concentrates than those found by Destiawan (2010) for crossbred lactating mares (494.6 ± 5.9 kg body weight, at 4 - 6 months of lactation period) that were given 6 kg Pangola grass, and commercial concentrates (3

kg Haras and 4 kg Vital); the mares were still allowed to graze the pasture to fulfill lack of grass consumed in Pamulang Equestrian Centre. The amount of forage consumed in this experiment was lower than that observed by Lawrence (1998). Lawrence (1998) found that grass intake was 9.5 - 13 kg/day for Thoroughbred and Quarter horses, but concentrates were eaten at a similar amount (3 - 4 kg/day). Differences could be due to differences in mare breed and its body weight as well as physiological condition and activity (Pond *et al.*, 1996; McDonald *et al.*, 2002; Tulung, 2015).

On the basis of DM intake (Table 3), the ratio of grass consumed was smaller than those of pollard and pellet; this could be due to the moisture content of grass that were lesser than those of pollard and pellet (Table 1). With the average DM intake was 6.37 kg/head/day, the average ratios of grass, pollard and pellet were 28, 30 and 42%. For the crossbred mares, total DM intake was 6.8 kg/head/day, and the ratios of grass, pollard and pellet were 29, 32 and 39%; for the local breed mares, total DM intake was 6.0 kg/head/day with the ratios of grass, pollard and pellet were 27, 29 and 44%. The data indicates that crossbred mares had greater ratio of grass and pollard intakes, but was lower in pellet ratio than the local mares. Although grasses were consumed at a smaller ratio, this caused variations in total DM intakes. Tulung (2015) also found that, with total DM intake was 12.23 kg in Indonesian race horse, corn was the most preferable feed consumed (38.01% of total DM intake) followed with grass, grain, bran, soybean and green bean; this means that the ratio of grass eaten by the horse was smaller than the corn. Corn, bran, and soybean were not given as single feeds in this experiment, but those feeds were parts of the pellet. Mende *et al.* (2015) also found that Indonesian race horse in Minahasa consumed 11.344 kg DM with the ratio between grass and local feed concentrate was 32.03% and 67.97%, and 11.849 kg DM with the ratio between grass and import feed concentrate was 30.88% and 69.12%. The data show that the horse consumed less grass on DM basis than the concentrate. This was because of horses are non-ruminant herbivorous animals known as hindgut fermenters in which feed (nutrient) fermentation by microbes occurs in the lower gastrointestinal tract; as hindgut fermenter, mares had limitation in consuming and using the grass (Pond *et al.*, 1996; McDonald *et al.*, 2002). DM intake varied among the horses and was affected by the age and physiological condition of the horse (Huntington, 2012). In Pamulang Equestrian Centre having crossbred horse, a 5 months old foal consumed 5.1 kg DM, stallions at different ages, young, stallion and old, had 7.8, 8.7 and 7.8 kg DM intake, and the equestrian stallion was capable of consuming 6.9 kg DM; however, no data were available for the grazing mares and its foals (Destiawan, 2010).

When the grass DM intake was standardised on the basis of body weight, the average percentage of grass consumed for crossbred mares was 0.50% and for local breed mare was 0.63% (Table 3). These percentages were lower than that suggested by Pond *et al.* (1996). Horses needed to be fed with forages at least 1% DM per body weight; this amount was required to maintain normal microbial function; forages were also needed to provide energy, protein, vitamins and minerals (Pond *et al.*, 1996; McDonald *et al.*, 2002). NRC (1989) in Huntington (2012) also indicated that mares, both in early and late lactations, per body weight, needed 10 - 2.0% forage; the early lactation mares required 1.0 - 2.0% concentrates, this concentrate amount could be reduced to 0.5 - 1.5% for the late lactation mares; as a result, the ratio between forage and concentrate was 50 : 50% of diet for mares in early

lactation, and 65 : 35% of diet for mares in late lactation. The greater ratio of concentrate given in this study and Destiawan (2010) result could be due to the moisture content and nutrient composition of grass that were lesser than those of concentrate diets (Table 1). The farmer then gave more concentrate to meet nutrient requirement. However, it is still necessary to suggest the farmer to give more grasses to the mares; but the amount of grass given should be based on mares condition (body weight and physiological condition) and ability of mares in using the grass as mares are non-ruminant herbivorous with hindgut fermenter (Pond *et al.*, 1996; McDonald *et al.*, 2002). Forage (grass), especially good quality forage, were still required as major nutrient sources, and used for preventing digestive problems such as colic or founder, and discouraging undesirable vices, such as cribbing and tail chewing (Anderson, 1994).

Table 3 also demonstrates that mares with heavier body weight consumed DM in greater amount than mares with lighter body weight. However, a reverse condition was observed when the intake was standardised on MBW basis; total DM intakes of crossbred mares were lower than those of local breed mares. These data could indicate that DM requirement for maintenance was greater in local breed mares than in crossbred mares. DM intakes as well as energy and nutrient intakes were affected by MBW, and other factors such as feed nutrient composition and digestibility, and workload, especially in race horse, milk production (Tulung, 2012; Huntington, 2012).

Table 3. Feed dry matter intake and its ratio

Horse breed	Dry matter intake (kg/head/day)				Dry matter intake (% BW/day)				Total dry matter intake (g/kg MBW/day)
	Grass	Pollard	Pellet	Total	Grass	Pollard	Pellet	Total	
G3	2.00	2.19	2.66	6.85	0.51	0.56	0.68	1.74	77.54
KPI	2.01	2.19	2.66	6.86	0.49	0.53	0.65	1.67	75.22
Tokol(a)	1.62	1.75	2.66	6.04	0.64	0.69	1.05	2.38	94.87
Tokol(b)	1.62	1.75	2.66	6.04	0.71	0.77	1.17	2.66	103.18
Sandel	1.65	1.75	2.66	6.07	0.54	0.57	0.87	1.98	83.00
Ave	1.78	1.93	2.66	6.37	0.58	0.62	0.88	2.09	86.76
± sd	0.20	0.24	0.00	0.44	0.10	0.10	0.23	0.42	11.92

As it was found in total DM intakes, the nutrient and DE intakes (kg/head/day) of crossbred mares were greater than those of local breed mares, but no differences in nutrient and DE intakes within each breed (Table 4). In term of MBW, nutrient and energy intakes of crossbred mares were lower than those of local breed mares with no differences in nutrient intakes within each breed (Table 5). However, Tokol mares consumed more nutrients and energy than Sandel mare. These indicate that mares with greater body weight had lower MBW causing lower nutrient and energy intakes, and these mares needed smaller amounts of nutrients and energy for meeting its maintenance requirement than mares with lower body weight and higher MBW (Pond *et al.*, 1996).

Table 4. Total nutrient and energy intakes per head

Horse breed	Total intakes (kg/head/day)						Total DE intake (Mcal/head/day)
	Ash	Organic matter	Crude protein	Ether extract	Crude fibre	NFE	
G3	0.660	0.619	0.734	0.266	0.959	4.230	15.74
KPI	0.661	0.620	0.735	0.267	0.961	4.234	15.75
Tokol(a)	0.592	0.545	0.645	0.244	0.833	3.724	14.06
Tokol(b)	0.592	0.545	0.645	0.244	0.833	3.724	14.06
Sandel	0.595	0.547	0.647	0.244	0.841	3.740	14.12
Ave	0.620	0.575	0.681	0.253	0.885	3.930	14.75
± sd	0.037	0.405	0.049	0.012	0.068	0.275	0.91

Table 5. Total nutrient and energy intakes on the basis of metabolic body weight

Horse breed	Total intakes (g/kg metabolic body weight/day)						Total DE intake (Mcal/kg metabolic body weight/day)
	Ash	Organic matter	Crude protein	Ether extract	Crude fibre	NFE	
G3	7.47	70.06	8.31	3.02	10.86	47.88	17.81
KPI	7.25	67.96	8.06	2.92	10.54	46.44	17.27
Tokol(a)	9.30	85.56	10.13	3.83	13.09	58.51	22.10
Tokol(b)	10.12	93.06	11.02	4.16	14.24	63.63	24.03
Sandel	8.15	74.85	8.86	3.34	11.50	51.16	19.31
Ave	8.46	78.30	9.27	3.45	12.05	53.52	20.11
± sd	1.22	10.69	1.26	0.53	1.57	7.33	2.89

Table 6 demonstrates percentage of nutrient and energy consumed per DM intake for lactating mares. Data show that no differences in percentage of all nutrients and energy consumed per DM intake between crossbred and local breed mares. Therefore, the percentage of nutrient consumed by the mares per DM intake were 9.74%, 90.26%, 10.69%, 3.97%, 13.89% and 61.70%, respectively for ash, OM, crude protein, ether extract, crude fibre and NFE, and the energy was 2.32 Mcal DE/kg DM intake. The crude protein level was little bit lesser, but the energy level was slightly greater than those suggested by Destiawan (2010) for crossbred lactating mares with 494.6 kg body weight at 4 - 6 months of lactation period. i.e. 11.66% crude protein and 2.09 Mcal DE/kg DM intake. Differences in energy and crude protein required by the mares in this experiment and Destiawan (2010) were due to differences in mare breed in which mares in Destiawan (2010) observation were mostly crossbred and Thoroughbred, body weight and milk production (Huntington, 2012). The percentage of crude protein consumed in this observation was above that required for maintenance (7.2%), but was less than that for mare in the early lactation stage (13%); mare at the last gestation period need protein at levels of 10 - 11% (Pond *et al.*, 1996). The crude protein level could still be lowered than that required for lactation at the last lactating month. Huntington (2012) suggested to provide ration with 13 - 15% crude protein with, at least, 1.5% forages per body weight and total intakes as much as 3 - 7 kg for lactating mares. In comparison to those found by Tulung (2012) with the Indonesian race horses, these horse consumed more DM (12.23 kg DM), ether extract (4.10%) and crude fibre (18.33%), but similar

percentage of crude protein (10.77%) with 0.3% Ca and 0.48% P. Differences between the two results could be due to differences in horse breed, metabolic body weight, physiological condition of the horse and milk production and composition. The data in percentage of nutrient consumed per DM intake could be used as a basis for estimating nutrient requirement by the lactating mares in the present condition. Once an appropriate nutrient requirement has been determined for the lactating mares, rations with suitable nutrient contents could be formulated using local feeds with known nutrient composition. The new diet formula (composition) is then suggested to the farmer to improve feeding the lactating mares.

Table 6. Percentage of nutrient and energy consumed per dry matter intake of lactating mares

Horse breed	Percentage of intakes (% DM intake)						Total DE intake (Mcal/kg DM intake)
	Ash	Organic matter	Crude protein	Ether extract	Crude fibre	NFE	
G3	9.64	90.36	10.72	3.89	14.00	61.75	2.30
KPI	9.64	90.36	10.71	3.89	14.01	61.75	2.30
Tokol(a)	9.81	90.19	10.68	4.04	13.80	61.67	2.33
Tokol(b)	9.81	90.19	10.68	4.04	13.80	61.67	2.33
Sandel	9.82	90.18	10.67	4.02	13.85	61.63	2.33
Ave	9.74	90.26	10.69	3.97	13.89	61.70	2.32
± sd	0.09	0.09	0.02	0.08	0.11	0.05	0.02

CONCLUSIONS

In Prima Fit Farm, Bogor, differences between the breeds occurred in field grass and pollard intakes due to differences in its body weight. However, each nutrients were consumed at similar levels on DM basis. Mares consumed all commercial pellets given (3.00 kg/head/day) with field grass and pollard intakes were 7.68 and 2.20 kg/head/day. Average total DM intake was 6.37 kg/head/day (86.76 g/metabolic body weight/day), and percentage of nutrient consumed per DM intake were 9.74%, 90.26%, 10.69%, 3.97%, 13.89% and 61.70%, respectively for ash, OM, crude protein, ether extract, crude fibre and NFE with the energy was 2.32 Mcal DE/kg DM intake.

ACKNOWLEDGMENT

Haji Dwi Susanto is thanked for allowing the authors to conduct the study in Prima Fit Farm, Bogor from June up to August 2012.

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NUTRIENT DIGESTIBILITY AND PHYSIOLOGY STATUS RESPONSE OF FEED RESTRICTION BLIGON MALE GOAT

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ABSTRACT

This study aimed to determine the digestibility of nutrients and physiological status of Bligon goats treated by restricting feed. Six Bligon goats aged of 12 months with an average body weight of 23.6 kg were fed on peanut roughes (*Rendeng*) and concentrate feeds. Goats were divided into 2 treatment groups. Three goats for the control treatment were fed diets based on dry matter (DM) requirements 3.5% of body weight, and three goats for the feed restriction treatment were fed diets with 50% reduction of DM requirement. Feed treatment was conducted for 35 days. The variables measured were: dry matter (DM) digestibility, organic matter (OM) digestibility, body temperature, respiration frequency, and heart rate. Data were analyzed by independent samples T-test. The results showed that feed restriction significantly reduced ($P < 0.05$) DM and OM digestibility, but gave not significantly ($P > 0.05$) effect on body temperature, frequency of respiration, and heart rate of Bligon goats.

Keywords: *Bligon goats, Feed restriction, Digestibility, Physiological status.*

INTRODUCTION

Goats are small ruminants which well known in Indonesia. Human population increases that lead increasing meat demand. Goat productivity should be maintained to satisfy meat demand, especially Bligon goats as meat production purpose. Bligon goat (Java Randu) is crossing between Kacang goat with Ettawa crossbreed (PE).

Feed availability especially forages is common problems in raising goats or cattle, abundant in wet/ rainy season but scarcity in dry season. Tropical country as in Indonesia has two season where the rainy season usually abundant of forage production, while during dry season forage production decreases (Aryanto *et al.*, 2013). Forage fodder is one integral requirement in the livestock development, especially ruminants (Suwignyo *et al.*, 2012).

Animal feed is important and needs special attention in the business of raising livestock since it becomes factor that determine the growth and productivity of livestock. Feed supply in the term of quantity and quality is important in order to obtain optimal results. Diet for housed livestock fully controlled by human including the type, amount and time period of feed offered (Tomaszewska *et al.*, 1991). Feed offered for livestock will also follow the pattern of feed availability in every season (Aryanto *et al.*, 2013). Farmers will provide feed that is less than usual (feed restriction) when feed availability decreased. Expected feed restrictions is reducing the number of feed offered but it does not interfere metabolic processes and animal health, even can increase the absorption of nutrients because the rate of digesta be slower.

Feed restriction increases the value of the organic matter digestibility, gross energy and crude protein in sheep (Kamalzadeh and Aouladrabiei, 2009) but decrease reproduction status, delaying ovulation and growth (Aboelmaaty *et al.*, 2008). Kacang goat and PE with feed restriction treatment, has lower body temperature below the normal, lower heartbeat and respiration but still within the normal range (Aryanto, 2012).

There still very few studies or research done with Bligon goat, particularly concern effects of feed restriction on feed consumption, digestibility and physiological status of the goat. This very important to observe response of animal on performance / productivity status including physiology status that might be caused by feed restriction management.

MATERIALS AND METHODS

Material

Six ± 12 months Bligon male goats with average body weight of 23,6 kg divided into two group, goat control (G₀) and feed restriction goat feeding trial (G₁). Individual cages stall type was used in this trial. Diets offered consist of 60% peanut roughages (*rendeng*) purchased from farmers in the region Banguntapan, Bantul, Yogyakarta and 40% commercial pelleted concentrate “Gemuk A[®]” (DM based). Diets provided is calculated based on the nutrient requirement (3,5% dry matter of body weight) for control goat (G₀), whereas 50% deduction for feed restriction goat feeding trial (G₁). Drinking water was given ad libitum for both groups. Chemical feed composition in this study are presented in Table 1.

Table 1. Chemical feed composition of peanut roughages and concentrate

Feed ingredients ^{*)}	Chemical composition (%)					
	Dry matter (DM)	Ash	Ether extract (EE)	Crude Protein (CP)	Crude Fiber (CF)	Extract Materials Without Nitrogen (EMWN)
peanut roughages (<i>rendeng</i>)	21.08	14.63	7.56	16.09	24.86	36.,86
concentrate	86.09	9.89	2.34	16.04	10.63	61.10

^{*)} Samples were analyzed at Forages and Pasture Laboratory, Faculty of Animal Science UGM

The equipment were used in this study consisted of forage chopper, balance scales for weighing cattle brands model EB9872 Camry, capacity of 150 kg with a sensitivity of 100 g. Balance scales brands Camry models EK3650/EK3651 capacity of 5 kg with a sensitivity of 1 g is used for weighing feed, feed samples, food remains and feces. Wiley mill with 1 mm screen sieve to grind samples of feed, food remains and feces. Digital analytical balance brands denver instrument XL 410 with a capacity of 410 g and 0,001 g sensitivity that was used for weigh the feed sample analysis. A set of proximate analysis equipment and digital thermometer to measure the rectal temperature goats.

Method

Preparation phase. Six goats were randomized into two treatment groups, three heads as a control (G₀) and three tail treatment of feed restriction (G₁). In the preparation phase was done for cleaning and repairing cages, feed bank and

drinking water storage, as well as the installation of feces container for digestibility sample collection.

Introduction phase. Goats were initially weighed before start the trial. Goats were placed in the individual stall cages in order to feed management purposes. Adaptation period for animal was performed for 1 month. Diet was given twice a day, 06.30 in the morning with half portion of and a half the rest was given in at 16.00 in the afternoon. After adaptation period, followed by a trial period of 35 days by feeding in accordance with the treatment.

In vivo phase. Feed In the *in vivo* digestibility trials were collected feed given, feed remains and feces samples. Consumption sample was collected 10% of the rest from both peanut roughages and concentrate feed per day, put into weighed paper bag prior to dried, grounded and laboratory analyzed. Total collections was done during the last 7 days in the trial feeding period. Feed remains and feces samples in that day was collected, weighed, recorded, put into weighed paper bag. Those samples were dried in the 55°C oven for 4 days, milled using a wiley mill with 1 mm screen sieve then proximate analyzed according to AOAC (2005). Physiological data was taken at the end of the collection feeding trial period.

Variables observed

Nutrient composition. Feed given samples were proximate analyzed (AOAC, 2005) of dry matter (DM), organic matter (OM), ether extract (EE), crude fiber (CF), crude protein (CP), extracts material without nitrogen (EMWN), and total digestible Nutrient (TDN). Rest feed and feces samples were analyzed of dry matter (DM) and organic matter (OM).

Nutrients consumption. Nutrient consumption is the amount of feed given nutrient deducted by the amount of nutrients in feed remains. Nutrient consumption: consumption of dry matter (DM), organic matter (OM), and total digestible nutrients (TDN).

Nutrient digestibility. Nutrient digestibility was determine from number of nutrients afford digested / absorbed in the digestive tract of cattle. Nutrient digestibility: dry matter (DM) and organic matter (OM). Nutrient digestibility formula according to Tillman *et al.* (1998):

$$\text{Nutrient digestibility} = \frac{\text{nutrient intake} - \text{nutrient feces}}{\text{nutrient intake}} \times 100\%$$

Frequency of respiration. Respiration frequency was done with count the number of goat breath (inhale and exhale) for 1 minute then repeated 3 times. The results was the average.

Heart rate (pulsus). Heart rate (pulsus) measurement in goats was done by palpated the pulsation of artery femoralis, done for 1 minute for 3 times then averaged.

Body temperature. Body temperature measurement was done by using a thermometer digital. Zeroed the thermometer, inserted into the rectum then read the scale, done for 5 minute for 3 times then averaged.

Statistical analysis

Nutrient digestibility data obtained were arranged based on completely randomized design. Data of nutrients consumption and physiological status obtained were analyzed by using Independent samples T-Test with the help of personal computer software Statistical Product and Service Solutions (SPSS) version 20.0.

RESULT AND DISCUSSION

Nutrient Consumption

The amount of nutrient consumption of each goat were calculated based on the deduction of nutrient content in feed given and residual feed (g/head/day), can be seen in Table 2.

Table 2. Nutrients consumption of Bligon goats (g/head/day)

Variable	Goat	
	Control	Restriction 50%
Consumption DM (g)	676.07 ± 14.759 ^b	372.30 ± 53.075 ^a
Consumption OM (g)	639.38 ± 15.885 ^b	349.88 ± 49.354 ^a
TDN (g)	461.60 ± 10.229 ^b	253.99 ± 36.174 ^a

^{a, b} different superscript in the same row shows the difference (P<0.05)

Based on the statistical analysis showed that feed restriction significantly (P <0.05) reduce dry matter intake (DM), organic matter (OM), and total digestible nutrients (TDN) in Bligon goats. It was quite natural since only 50% feed provided for animal, finally will affect to consumption.

Dry matter intake

Dry matter consumption of control Bligon goats showed significantly higher (P<0.05) compared with feed restriction Bligon. G₀ consumed DM 676.07 g/head/day higher than G₁ 372.30 g/head/day. Kearl (1982), states that dry matter intake for basic living goat weighing 20-25 kilograms of between 540-640 grams. Based on the reference, DM consumed by G₀ has met the requirement, but not for G₁. The amount of DM consumed by G₁ did not meet basic living needs. There some Factors affected nutrient consumption as if number of feed, feed intake, digestion rate of feedstuffs in the digestive tract, the spending rate of rest feed consumed and nutrient quality of the diet offered (Tillman *et al.*, 1998).

Organic Materials Consumption

Statistical analysis showed that the OM consumption of G₀ significantly higher (P<0.05) than G₁. OM consumption of G₀ was higher (639.38 g/head/day) higher than G₁ consumption (349.88 g/head/day). Organic matter (OM) consumption was influenced by DM consumption. So that the OM consumption of G₀ was higher than G₁. This accordances with Sutardi (1980), stated that OM closely related with DM, because OM is part of DM. Decreasing DM consumption will be followed by decreasing OM consumption as well and vice versa.

Consumption of total digestible nutrients

TDN value of the consentrat and peanut roughages (based on computation formula Hartadi *et al.*, 2005) were 71.11% and 65.96%. Hartadi *et al.* (2005) stated that TDN of peanut roughages (*rendeng*) about 65.00%. While, Meianto (2009) stated that TDN concentrates Bligon goat consisting of rice bran, pollard, kleci, soybean meal, and mineral about 71.67% TDN.

Data % TDN obtained then be used to compute TDN consumption of feed. TDN consumption fo G₀ was significantly (P<0.05) higher (461.60 g/head/day) compare with TDN consumption of G₁ (253.99 g/head/day) as seen in Table 2.

Perry (1984) stated that TDN requirement for 23 kg body weight of goat for maintenance status is 292.9 g/head/day. Based on the result, G₀ has met the TDN

requirement, but not the G_1 . Aside of intake level, TDN consumption could be influenced by nutrients quality of the feed due to total digestible nutrients is the number of energy that can be derived from the digestible nutrient content of feed (Siregar, 1994).

Nutrient Feed Digestibility

In vivo Feed digestibility trial was done to study the response of goat either in the *ad libitum* phase or feed restriction phase. Digestibility values measured were DM digestibility and OM digestibility. The results of digestibility analysis of DM and OM in both group of goat can be seen in Table 3.

Based on the results showed that feed restriction significantly ($P < 0.05$) reduce DM and OM digestibility in Bligon goats.

Table 3. Nutrient digestibility on Bligon goat (%)

Variable	Goat	
	Control	Restriction 50%
Digestibility BK	74.39 ± 1.597 ^b	71.27 ± 0.886 ^a
Digestibility OM	79.42 ± 1.540 ^b	76.34 ± 0.239 ^a

^{a, b}Superscript on the same row shows the difference ($P < 0.05$)

Dry matter digestibility

Table 3 showed that DM digestibility of G_0 (74.39%) was significantly higher ($P < 0.05$) compare with G_1 (71,27%). Tillman *et al.* (1998) stated that the highest digestibility obtained in the consumption with restriction which is given lower than basic life need. However in this study, the restricted feeding obtained digestibility value lower compared with control Bligon goat which is given feed based on their needs. This may be caused by number and duration of restriction. Rumen microbes can not grow optimally in the excessive feed restriction, so that feed digestibility will not optimal. Research done by Aryanto (2012) resulted that DM digestibility in Kacang goat during restriction phase was lower than in the *ad libitum* phase. Van Soest (1994), stated that factors that affect feed digestibility are animal species, age, feed treatment, crude fiber and lignin, the influence of feed association, nutrient deficiency, feed composition, the physical form of feed, feed level, frequency of feeding and drink, plant age and feed retention in the rumen.

Organic matter digestibility

Table 3 showed that OM digestibility of G_0 (79.42%) was significantly higher ($P < 0.05$) compare with G_1 (76.34%). Digestibility of organic matter is closely related to the dry matter digestibility. Organic matter associated with DM digestibility values, OM digestibility will be higher than the value of the dry matter digestibility. Research done by Aryanto (2012) resulted that OM digestibility in Kacang goat during restriction phase was lower than in the *ad libitum* phase. Kamalzadeh and Auoladrabiei (2009) stated that OM digestibility of Sangsari sheep with feed restriction was higher than Sangsari sheep given *ad libitum* feed. The difference results of study may be due to the different types and breeds of animal, probably will also affect to different digestibility response. Other factors that might affect the digestibility are feed composition, crude protein, fat, marrow composition, preparation of feed, animal factor, and the number of feed (Tillman *et al.*, 1998).

Status Physiology

Measurements of physiological status was done to study the response of animal either in the ad libitum phase or feed restriction phase. Measured physiological status include body temperature, respiration frequency and heart rate (pulsus). The results of body temperature, respiration frequency and heart rate (pulsus) measurement in the G₀ and G₁ can be seen in Table 4.

Table 4 showed that body temperature, respiration frequency and heart rate (pulsus) did not affected by treatment.

Table 4. Status physiology result in the G₀ and G₁

Variable	Goat	
	Control	Restriction 50%
Body temperature (°C) ^{ns}	38.60 ± 0.200	38.23 ± 0.305
Respiration (x/) ^{ns}	30.67 ± 5.033	26.00 ± 4.000
Heart rate (pulsus) (x/minute) ^{ns}	77.33 ± 9.237	76.00 ± 10.583

^{ns} non significant

Body Temperature

Body temperature was not significantly different between G₀ and G₁. This indicated that feed restriction still in the limit tolerances / can be accepted by the goat. Table 4 showed that Body temperature of G₀ and G₁ still in the normal condition. Triakoso *et al.* (2011) stated that normal Body temperature ranged from 38.6°C to 40.2°C. However, Body temperature of G₀ bit lower. This may be one response of feed restriction, where the goat was in unusual situation, allowing the temperature changes to unusual situation. Isroli *et al.* (2011) stated that the changing of animal body temperature can be affected by the heat which produced from the number of feed consumed. Heat of the body is produced by metabolic activity (Swenson and Reece, 1993). Ganong (2002) stated that some chemical reactions in the body produced heat, as well as at mealtime.

Frequency of Respiration

Frequency of respiration was not significantly different between G₀ and G₁ respectively 30.67 and 26 times/minute. Table 4 showed that frequency of respiration of G₀ and G₁ still in the normal range value. Smith (1988) stated that the normal range of respiration frequency of goats ranged from 26 to 54 times/minute. The frequency of respiration was also depend on the amount of food consumed. Mushawwir (2010) stated that in order to transform feed into energy, animals need oxygen through the process of respiration. The lower number of animal feed intake, the needs of oxygen to convert feed into energy also lower, so that the frequency of respiration also lower. Factors that may affect the frequency of respiratory according to Swenson and Reece (1993) were body size, age, muscle movement, ambient temperature, pregnancy, and full digestive.

Heart Rate (pulsus)

The heart rate was not significantly different between G₀ and G₁ respectively 77.33 and 76 times/minute. Table 4 showed that heart rate of G₀ and G₁ still in the normal range value. Frandson (1992) stated that normal heart rate of goat ranged from 70 to 135 beats/minute. Heart rate associated with metabolic rate. Feed that was eaten can increase metabolic rate (Ganong, 2002). Reducing the number of feed consumption will reduce to rapidity of metabolic rate, thereby heart rate also slower.

CONCLUSION

Based on the research done, it can be concluded that the restriction of feed (feed restriction) significantly affected the nutrient consumption (DM and OM), feed digestibility (DM and OM) of Bligon goats but did not affect to physiological status. Bligon goats was given only 50% of the requirement (DM basic), has lower consumption and digestibility (DM and OM) compare with full feed goats. However, Body temperature, respiration frequency and heart rate (pulsus) did not differ between both treatment.

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SUPPLEMENTATION OF HEIT - CHROSE INTO DAIRY COW DIET IMPROVES IN VITRO RUMEN FERMENTATION

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ABSTRACT

The objective of the study was to evaluate the effect of Heit-Chrose (HC) supplementation in the diet on *in vitro* ruminal fermentation of dairy cattle. HC is a feed supplement containing allicin and organic minerals (Se, Cr, and Zn). This research was conducted experimentally using Completely Randomized Design with 6 treatments and 4 replications. The treatments were: CTL = dairy cattle feed (CP 15.38%, CF 23.38%, TDN 61.26%); OM = CTL+ organic minerals (0.3 ppm Se + 0.15 ppm Cr + 40 ppm Zinc-lysinate) + 0 ppm of HC, HC-15 = CTL+ 15 ppm HC; HC-30 = CTL+ 30 ppm of HC; HC-45 = CTL+ 45 ppm of HC; HC-60 = CTL + 60 ppm of HC. Data were analyzed using analysis of variance with SPSS version. HC supplementation increased DMD, OMD, VFA, and reduced total gas and protozoa count. HC supplementation greater than 30 ppm did not effectively improve ruminal fermentation. Supplementation at 30 ppm HC to dairy cow diet was the correct level to improve the efficiency of rumen fermentation.

Keywords: *Heit-chrose, Ruminal fermentation, Dairy cattle.*

INTRODUCTION

Manipulation of rumen ecosystem aims to improve the digestibility of crude fiber in feed, reduce methane production and nitrogen excretion by ruminants. This is done to improve performance and is the most important goal of animal nutritionists (Patra et al., 2006). Extracts of plants with high concentrations of metabolites are the best way to achieve one or more of these goals (Teferedegne, 2000; Wanapat *et al.*, 2008). Garlic has the complete content such as allicin (C₆H₁₀S₂O), diallyl sulfide (C₆H₁₀S), diallyl disulfide (C₆H₁₀S₂), allyl mercaptan (C₃H₆S), and other contents (Lawson, 1996). These components can manipulate rumen fermentation such as reducing the proportion of acetate and propionate and increasing butyrate, as inhibitors of methanogenesis and lowering the ratio of CH₄: VFA (Busquet *et al.*, 2005). Kim et al. (2009) reported that garlic skin had a 7-fold greater the polyphenol content than that of garlic bulbs, among others, allicin, which plays a role in the decline of methanogens. Feed fermentation in the rumen will result in a decreased availability of mineral (Prayitno and Widiyastuti, 2010). Micro minerals like Selenium, Chromium, and Zinc can improve the efficiency of rumen fermentation (Prayitno et al., 2013). Selenium supplementation of 0.3 ppm + 1.5 ppm Chromium + 40 ppm Zinc in dairy cattle feed improve feed efficiency although methanogens that are formed are still in the level of 25.4 mM (Prayitno and Widiyastuti, 2010). The purpose of this study was to assess the effect of supplementation with Heit Chrose in dairy cattle feed on the efficiency of *in vitro* rumen fermentation.

MATERIALS AND METHODS

The experiment was conducted by using a completely randomized design (CRD), with 6 treatments and 4 replications . Feed consisting of 60 % forage (Elephant grass 95 % and 5 % Gliricidia leaves) and concentrate (coconut meal, soybean meal, pollard, cassava, minerals, CGF (Corn Gluten Feed), and CGM (Corn Gluten Meal). Forage was dried in the sun and then mashed. The research was conducted experimentally by in vitro method of Tilley and Terry (1963). Inoculum source came from dairy cows, taken via mouth. The tested treatments were Control : dairy cow Feed (13.8 % CP; 25.7 % CF; 58.6 % TDN); HC-0 : Control + organic minerals (0.3 ppm Selenium + 1.5 ppm Chromium + 40 ppm Zink- lysinat) + 0 ppm HC; HC-15 : Control + 15 ppm of HC ; HC-30: Control + 30 ppm of HC; HC-45: Control + 45 ppm of HC; HC-60 : Control + organic mineral + 60 ppm extracts of garlic skin. The data were analyzed using analysis of variance (Anova) followed by Honestly Significant Difference (HSD) test (Steel and Torrie, 1995). The variables to be measured were digestibility of dry matter (DMD), digestibility of organic matter (DOM), VFA, NH₃, total gas (Menke and Steingass, 1988), the population of bacteria and protozoa (Ogimoto and Imai, 1981) and NDF digestibility.

RESULTS AND DISCUSSIONS

The results showed that there was a highly significant effect ($P < 0.01$) of the supplementation treatments on the DMD and the OMD. Therresults of the study are shown in Table 1. The value of dry matter and organic matter can be used as an indicator of the ease of feeding the rumen microbes that were degraded by digestive enzymes and digested by pascarumen. The higher the DMD, the higher the OMD, and the higher the chances of nutrients that can be used both for livestock production and other activities. This study showed that supplementation with HC could improve rumen microbial activity.

Table 1. The Effects garlic peel extract on DMD, OMD, total VFA, NH₃, protozoa count, bacterial count, total gas production, and NDF digestibility

Parameter	CNT	HC-0	HC-15	HC-30	HC-45	HC-60
DMD	34.09±1.7 ^a	37.85±1.58 ^{abc}	39.32±2.59 ^c	34.13±1.43 ^{ab}	35.5±0.57 ^{abc}	35.48±2.10 ^{abc}
OMD	32.02±1.45 ^a	35.57±2.01 ^{abc}	37.22±0.93 ^c	32.05±4.03 ^{ab}	33.4±2.09 ^{abc}	33.28±0.75 ^{abc}
NDFD	31.33±2.14 ^{ab}	34.71±2.12 ^{ab}	36.58±2.01 ^b	31.67±0.76 ^{ab}	31.12±2.84 ^{ab}	29.43±1.46 ^a
pH	7±0.00 ^b	7±0.00 ^b	6.9±0.00 ^a	6.97±0.06 ^b	7±0.00 ^b	7±0.00 ^b
Total VFA	147.5±24.24 ^{ab}	165±6.00 ^{abc}	180.5±12.26 ^{bcd}	209±24.30 ^{bd}	174.5±12.15 ^{bcd}	132±9.93 ^a
Amonia mM	14.68±1.59	14.72±1.652	14.20±1.00	13.20±1.579	16.88±1.439	15.58±1.895
Gas Total	27.71±3.68 ^c	24.28±2.13 ^{abc}	22.48±1.21 ^{abc}	21.06±2.04 ^a	21.63±5.58 ^{ab}	24.34±1.6 ^{abc}
Protozoa (10 ⁶ /ml)	2.05±0.503 ^b	1.46±0.498 ^{ab}	0.94±0.423 ^a	1.18±0.323 ^{ab}	1.35±0.270 ^{ab}	1.34±0.518 ^{ab}
Bacteria (log 10/0.05g DM)	10.28±0.119	10.24±0.106	10.32±0.103	10.39±0.122	10.52±0.044	10.50±0.112

Notes : Values in the some row with similar letter (s) are not significantly different ($P > 0.05$)

Total VFA

The results showed that there was a highly significant effect ($P < 0.01$) of the treatment given. The mean concentration of Volatile Fatty Acids (VFA) in total treatment feed ranged between 132-209 mM. The value had exceeded the value of the adequacy of VFA. Sutardi *et al.* (1977) states that the VFA concentration range that is sufficient for rumen microbial growth is 80-160 mM. Supplementation of HC in the substrate significantly affected the concentration of total VFA through increased DMD and DMO so there were more dietary available nutrients to be converted into C2 (acetate), C3 (propionate), and C4 (butyrate) through the process of carbohydrate metabolism. According to Van Soest (1994), the VFA is the most important source of metabolic energy for ruminants and the source of the carbon chain for microbial synthesis because VFA is able to supply 55-60% of the energy needed by cattle.

This study showed HC supplementation increased the production of VFA, or manipulated the fermentation process to increase efficiency. VFA production increase occurred with supplementation of 30 ppm HC. VFA production is important to know the process of fermentation of carbohydrates and related to the productivity of livestock because most of the VFA in the rumen derived from the fermentation of feed carbohydrate (Hungate, 1966). At ration treatment, HC-30 had the highest total VFA, this treatment was thought to contain non-structural carbohydrate (starch) more than the other treatments so as to contribute to the fermentation of feed, resulting in higher VFA production. This is in line with the statement of Jouany (1994) that the non-structural carbohydrates contribute VFA production that is higher than structural carbohydrates.

Ammonia

The results showed that the treatment affected ($P < 0.05$) the concentration of ammonia. It showed that supplementation of HC in the dairy affected microbial activity in the rumen metabolism of proteins to produce ammonia. The feed average concentrations of ammonia ranged from 13-16 mM. The values had exceeded the value of the adequacy of ammonia in the rumen. One of the things that can cause high concentrations of NH_3 in each treatment is not for the absorption of ammonia in *in vitro* systems, that cause the accumulation of ammonia in the syringe, there is a residual ammonia, which is not used in the synthesis of microbial protein together with the carbon framework derived from the fermentation of carbohydrates, and can also come from donations of protein lysis from microbes. The results in Table 1 show that the average values of ammonia on the successive treatments were 14.72 ± 1.652 (HC-0), 14.20 ± 1.000 (HC-15), 13.02 ± 1.579 (HC-30), 16.88 ± 1.439 (HC-45), 15.58 ± 1.895 (HC-0). The research of Kurniawati (2009) reports the concentration of NH_3 generated from all treatments with the addition of lerak extracts (*Sapindus rarak*) ranged between 15.85 to 16.31 mM and the values were still optimal for rumen microbial growth. The NH_3 concentration of less than 3.57 mM may inhibit rumen microbial growth (McDonald *et al.*, 1995).

Total Gas

The results showed that extracts of garlic skin affected rumen fermentation of liquid phase when the feed of dairy cows substrate contained adequate organic minerals. Gas production had values that varied and tended to be lower than in control (HC-0 and HC-15) (Table 1). The highest gas production occurred in HC-60

treatment. The total gas was a result of fermented feed, especially organic materials into VFA performed by rumen microbes. The amount of gas produced showed high or low digestibility of the feed. High production of gas indicated inefficiency usage of feed, thereby, increased the gas production. The little amount of gas produced in this study showed that fermented organic materials were used for microbial protein synthesis (Van Soest, 1994).

Protozoa Population

Protozoa population in these treatments had values that varied and tended to be lower than the control, except for R2. This indicated that the saponins in HC (20:13%) could break down the cell membrane of protozoa (Prayitno et al., 2013). On the other hand, the HC could inhibit the formation of methane, which was of allixin origin that inhibited the enzyme, HMG-CoA reductase (Busquet et al., 2005). As a result, the availability of H₂ in the rumen was not used for the synthesis of methane, but for the synthesis of propionate (Prayitno et al., 2013).

The results of analysis of variance showed that the treatment affected highly significantly (P <0.01) on the protozoa population. The highest average value on protozoa population was reached by the control feed (R0) is 3.80 ± 0.32 (10⁶ cells / ml rumen fluid) and the lowest average was achieved by control feed + 45 ppm HC (HC-45), 2.35 ± 0.37 (10⁶ cells / ml rumen fluid). Protozoa population declined due to reduced population of methanogens that are symbiotic with protozoa due to saponins and allixin role of HC. Kongmun et al. (2011) adds, the saponin contained in the flour of garlic oil can reduce methane bacteria, therefore, the protozoa population decreases.

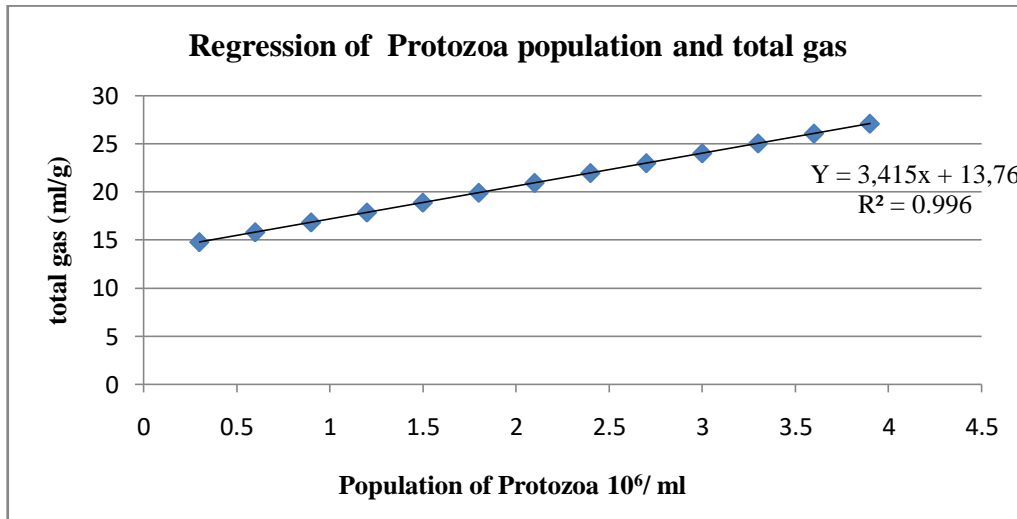


Figure 1. Relationship between Protozoa Population and Production of Total Gas

Total Bacteria

The results showed that the treatment affected highly significantly (P <0.01) on total rumen bacteria. Increased total rumen bacteria presumably because of the feed treatment that was capable of providing nutrients for the growth of the microbes. In addition, it could be affected by the presence of declined protozoa population due to inhibition of methanogenic activity that was in symbiotic with protozoa (Newbold et

al, 1995). The average values of total bacteria ranged from 10.32 to 10.52 (log 10/0.05 gr DM). The mean of total bacteria from all treatments is shown in Table 1. Based on Table 1, treatment HC-60 with a level of 60 ppm HC had a value of highest total bacteria, thus the active ingredient in allicin and organosulfur of garlic efficiently inhibited the performance of the enzyme of HMG - CoA in suppressing the methanogenic population, so that feed was used more efficiently and total rumen bacteria increased. A research by Kongmun et al., (2011) showed administration of 50 grams of garlic powder on buffaloes were able to increase the total cellulolytic bacteria by 42.2% in the 4-hour observation. The increase of fiber degrading bacteria population will result in an increase in rumen fermentation products. The increasing population of microbes in the rumen will increase the amount of microbial protein.

NDF Digestibility

The results showed a decline in NDF digestibility significantly ($P < 0.01$) in table 1. NDF digestibility in this study ranged between 29.43 % - 36.58 %. NDF digestibility was highest in the treatment of HC-15 amounted to 36.58 % and the lowest for the treatment NDF digestibility HC-60 amounted to 29.43 %. The high digestibility of NDF in the treatment of R2 assumed to be was due to the ability of the rumen microbes to degrade the fiber fraction. This study showed that supplementations of dairy cows feed with HC to the extent of 30 ppm were able to increase the efficiency of rumen fermentation. This condition occurred presumably because of the increased rumen microbial activity and decreased production of CH₄. Heit Chrose with active ingredient of allicin and saponins and Se, Cr, and Zn minerals allegedly was able to improve rumen fermentation by way of decreasing the activity of methanogens, lowering population of protozoa and increasing the population of bacteria.

CONCLUSION

Heit Chrose supplementation in feed of dairy cows by in vitro is able to increase the efficiency of rumen fermentation, as indicated by the increasing feed digestibility, VFA and decreasing total gas production and protozoa populations.

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THE EFFECT OF BUFFER ADDITION ON ANAEROBIC MEDIUM WITH CARBOXYL-METHYL-CELLULOSE ASCARBONE SOURCES AND CELLULASE INDUSERON RUMEN CELLULOLYTICMICROORGANISMS DEGRADATION

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ABSTRACT

This research was aimed to study of the effect of buffer addition on rumen cellulolytic microorganisms degradation of anaerobic medium that consists of carboxyl-methyl-cellulose (CMC) as carbon source and cellulase induser. Cellulase is an enzyme which acts in cellulose degradation into simple glucose. Liquid medium MRS with a pH of 3.8 was added to CMC 1% as a carbon source as well as cellulase induser, and rumen liquid as a source of microbes. The treatment was NaHCO₃ buffer addition in the liquid medium with the level of addition of 0%, 0.1%, 0.2%, 0.5%, 0.75%, and 1%. The microbe growth was identified using Hungate tube for 24 hours at 39°C with 3 replications. At the end of fermentation process, the observation was conducted for microbe protein concentration, a residual of reducing sugar, and carbohydrates total. Data were analyzed using one way anova. If there is significant result due to buffer addition, Duncan's new multiple range test (DMRT) was used to compare means. The results showed that supplementation of buffer NaHCO₃ in the medium CMC as a carbon source and cellulase inducer gave significant ($P < 0.05$) result towards microbes protein concentration, residual of reducing sugar, and decreasing carbohydrates total ($P < 0.05$). It can be concluded that the addition of NaHCO₃ buffer will increase the medium pH and the activity of rumen cellulolytic bacteria will be affected because there was an increase in microbes protein number, carbohydrates total, and residual of reducing sugar.

Keywords: *CMC, Rumen microbial cellulolytic, pH, Buffer*

INTRODUCTION

People awareness towards the importance of animal protein consumption (e.g. meat, egg, and milk) increases along with increment of scientific knowledge and income. Animal protein becomes an important role as the basic to improve human resource development quality. The requirement of animal protein is fulfilled domestically, though most of the need of meat and milk in Indonesia is still fulfilled overseas. Because of that, there is a need to increase the production from ruminant livestock.

Ruminant livestock depends on the forage feedstuff. Forage productivity has fluctuation trend, and is abundant in rainy season and lack in dry season. It often happens in a populated area of cattle. The main problem in the cattle production development in Indonesia is the difficulty to fulfil the stock of feed continuously, in terms of quality and number. Cattle who is only given with forage will have low productivity so that they need to be fed by high quality of feed such as concentrates or silages. The provision of high quality concentrates or silages will enhance cattle's

development, so that the approximate gain will be reached in a short time. However, the provision of high quality concentrates or silages in massive number will result bad effect because it can cause acidosis. Acidosis happens if the cattle consumes silage or concentrate with high soluble carbohydrates source. Carbohydrates fermentation occurs rapidly inside the rumen and provides lactic acid in a big number which makes a great acid difference in the rumen spontaneously so that the rumen takes a risk to get acidosis. Beauchemin (2007) stated that feed with high energy consists of low neutral detergent fiber (NDF) and high starch. Starch source is often processed through a way to optimize the availability of starch inside rumen and fiber source is easily digested because it is given in small particle forms. The effect from those feed is this kind of feed which is easily fermented in the rumen and it makes a lack of crude matters which are needed to maximize the digestibility and the flow of saliva buffer to the rumen. The result shows that rumen pH decreases along with increasing acidosis risk.

The handling of agricultural waste can biologically be conducted by using enzyme like cellulase. Cellulase is an extracellular enzyme which contains of *endo*- β -1.4-gluconase, *exo*- β -1.4-gluconase (*aviselase*, *selobiohidrolase*, C1 cellulose) and β -1.4-glucosidase or *selobiase*. However, this research aims to find out the effect of pH towards cellulose enzyme activity. The relationship between enzyme activities is presented in the Figure 1.

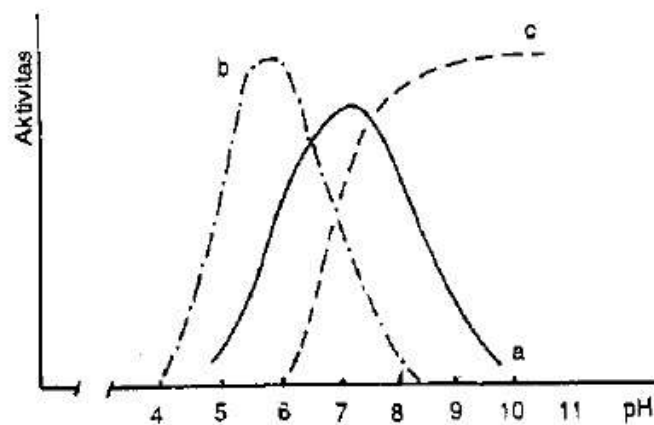


Figure 1. The effect of pH fluctuation towards enzymes activity

Based on the figure above: (a) the activity curve is generally served that optimal pH has the bell form, (b) the optimum pH number depends on the enzyme and this reliance can be more or less sharp, (c) for some enzymes, the activity isn't depended on the pH.

MATERIAL AND METHODS

Materials

The instruments which were used in this research were autoclave, oven, spectrophotometer GENESYS 20, water bath, Sartorius analytical scale with maximum capacity 160g and sensitivity 0.0001 g, pH meter, vortex mixer, tubes, Erlenmeyer, and *eppendorf*.

Materials which are used in this research were aquades, bacteria inoculation reagent, reagent for microbe protein concentration test with Lowry methods, reagent

for residual sugar with Nelson and Somogyi methods, and reagent for total carbohydrates test with Antron methods.

Methods

Bacteria inoculation. 200 ml medium which contained of 30 ml mineral I, 30 ml mineral II, 0.2 ml 0.1% resazurin, 50 ml aquades, 0.1 g 1% CMC, 1 g yeast extract, 6.66 ml 12% Na₂CO₃, and 3.34 ml 0.1% *cysteine-HCl*. Liquid Medium except 1% CMC substrate washomogenized by using stirrer and medium pH and wasjustified at 3.8 by adding acetate glacial acid. 1% CMC is separated in the tubes and diluted by adding aquades. Medium and substrate weresterilized with autoclave. After that, the medium is added with 1% CMC substrate and rumen liquid as microbe source. The medium is boiled and added by 3% cysteine-HCl and 12% Na₂CO₃. Afterward, it wasflown with CO₂ until it reaches anaerobe situation which is marked by the change of medium color from pink until yellow. As the following step, Erlenmeyer wascovered by paper and brown color paper.

Degree of Acidity or pH.The pH number wasprovided from fermentation result after 24 hours incubation.

Microbe Protein. Rumen liquid from the result of microbe growth wasrotated at 3000 rpm for 15 minutes. The supernatant wastaken and putinto *ependorf*. The supernatant was centrifuged at 10.000 rpm for 15 minutes. Precipitates from centrifugation wereseparated for the determination of microbe protein concentration using Lowry method. Precipitates from preparation sample were added by 1 N NaOH and then boiled at 90°C for 10 minutes. 0.5 ml sample was put into the tubes and added by 2.5 Lowry B, and, then, homogenized by vortex and left for 10 minutes. After that, 0.25 ml Lowry A was added into it and it wasleft at room temperature for 30 minutes. The collected results wereread by using spectrophotometer with wavelength 750 nm. Protein concentration wascalculated by using equation: $Y = 2.507 x + 0.082$ in which X is protein concentration and Y is absorbent.

Residual sugar. Sampel which was separated from its protein by using Zn hydroxide and Barium sulfate provides filtrate which did not contain reduced substance except glucose. Zn-barium filtrate washeated with Cu-alkali reagent and, then, added by colorizing reagent *arsenomolibdat*. Filtrate color was compared to the standard. At first, protein wasprecipitated by 0.5 ml sample + 1.5 ml Aquades + 1.5 ml Ba(OH)₂ 0.3N + 1.5 ml ZnSO₄ 5% and homogenized together. The sample wassentrifuged at 3000 rpm for 20 minutes. Supernatant was used to determine glucose concentration. The determination of residual sugar usedNelson and Somogyi method. Firstly, two tubes are set, in which one wasadded by 1 ml sample and the other one is added by 1 ml aquades as blank. The two tubes werereadded by 1 ml Nelson reagent and heated in the 100°C water bath for 20 minutes. The samples are taken and cooled down in the water flow at room temperature (25°C). After that, those tubes werereadded by 1 ml arsenomolibdat and homogenized using vortex. The liquid wasread at wavelength 540nm. The glucose resulted in reduced concentration can be calculated byusing equation $X = Y - 0.063/0.003$ in which X was residual sugar and Y is absorbent.

Total Carbohydrates. Supernatant which wasseparated from protein was used to determine total carbohydrates. The determination of total carbohydrates used Antron method. Firstly, there were two tubes. One tube is added by 1 ml sample and the other one is added by 1 ml Aquades as blank. The two tubes are added by 1 ml Antron

reagent and heated at 100°C water bath for 20 minutes. The samples are cooled down in the water flow and at room temperature (25°C). The samples are read with spectrophotometer with the wavelength 540 nm. The residual sugar is calculated with the equation: $X = Y - 0.15/0.007$ where X is total carbohydrates concentration and Y is absorbent.

RESULTS AND DISCUSSION

Enzyme is protein compound which can catalyze all chemical reaction in biology system. Enzyme works by attaching at the molecular surface and reacting together so that it can accelerate the reaction process. Lehninger (1982) stated that catalyzed activity of enzyme in the cell is set partially by the change of environmental medium acidity. The pH environmental number can affect the enzyme activity speed to catalyze the reaction. It is affected by hydrogen concentration affect 3-D structure enzyme and the activity. Every enzyme has optimum pH which is unique; the pH number can affect the maximum activity. pH optimum enzyme is not always the same as the normal environmental pH, and with pH which is more or less than the optimum pH. At the optimum pH, the 3-D structure enzyme can attach the substrates at the most conducive point. If the hydrogen concentration changes from optimal concentration, enzyme activity will progressively be lost until the enzyme is not functioned anymore.

The first step to know is that the microbe growth and enzyme production are conducted by Optical Density (OD) to expect the cells number in each cellulolytic inoculums. Compositions of medium to produce cellulase enzyme are KH_2PO_4 , $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$, $(\text{NH}_4)_2\text{SO}_4$, $\text{CaCl}_2 \cdot 2\text{H}_2\text{O}$, $\text{CoCl}_2 \cdot 2\text{H}_2\text{O}$, urea, $\text{FeSO}_4 \cdot 7\text{H}_2\text{O}$, $\text{MnSO}_4 \cdot \text{H}_2\text{O}$ and $\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$ and CMC as carbon source and cellulase inducer in which each of those substances has function in cellulase enzyme activity.

Every cellulolytic bacteria gains complex cellulase enzymes which are different and which depend on the genes and the kind of carbon source which is used. In this research, all the inoculums grow in CMC liquid medium and contain of CMC as carbon source and inducer which has trouble because of the low acidity by adding acetate acid until the pH reaches 3.8 and, then, it is added by NaHCO_3 as buffer with proportion 0%, 0.1%, 0.2%, 0.5%, 0.75%, and 1%. Those were conducted to prove that enzyme activity was affected by pH. The pH medium was one important factor for microorganism growth and fermentation product production.

Optimum pH enzyme has high stability. The same enzyme has different optimum pH because it depends on the enzyme source. Winarno (1986) stated that the change of enzyme activity because of the change of ionized enzyme, substrate or complex enzyme-substrate, and the change in the enhancement ability and the effect of reaction rates. In general, enzyme shows maximum activity at some rate which is called as optimum pH. Keidane and Birgele (2003) stated that, with normal pH for proteinase and peptidase, enzyme are 5.5 to 7.0, cellulase is between 6.2 to 7.0, deamination pH is between 6.5 to 7.0.

Yeast extract is added in the medium for triggering cell growth in the first phase. After glucose in the medium is run out, the bacteria will use carbon source from cellulose by synthesizing cellulase enzyme. CMC at the medium was a substrate and also inducer to provide cellulase enzyme. In addition, CMC can be used for cellulolytic bacteria as the main carbon source to provide glucose. Apriani et al (2014) informed that inducer is a substance which is needed to induce the gene so that

there is genes expression, which is exactly like Jacob-Monod theory about induced enzyme. CMC can be used by cellulolytic bacteria as main carbon source to produce glucose. From this research, the collected data such as pH, residual sugar, microbe protein and total carbohydrates are presented on the Table 1.

Table 1. The results of CMC incubation towards different level of NaHCO₃ buffer

Treatments	pH	Residual sugar (mg/ml)	Microbe protein ^{ns} (mg/ml)	Total Carbohydrates (mg/ml)
0%	5.51 ± 0.36 ^a	56.78 ± 3.10 ^b	2.43± 0.24	67.71 ± 1.97 ^c
0.1%	5.97 ± 0.01 ^b	50.00 ± 7.53 ^{a,b}	2.81± 0.11	42.40 ± 5.99 ^a
0.2%	6.43 ± 0.06 ^c	42.61 ± 8.07 ^a	2.59 ± 0.51	37.21± 7.62 ^a
0.5%	6.78 ± 0.18 ^d	40.44 ± 3.18 ^a	2.84 ± 0.50	37.83 ± 3.73 ^a
0.75%	6.90 ± 0.12 ^d	42.99 ± 3.21 ^a	1.99 ± 0.38	53.16 ± 7.73 ^b
1%	7.56 ± 0.12 ^e	59.39± 2.42 ^b	2.28 ± 0.27	62.45 ± 3.78 ^{b,c}

^{a,b,c,d,e} different superscript at the same row shows significant difference (P<0.05)

^{ns} non-significant difference (P>0.05)

Activity of endo-1.4-β-*glucanase* enzyme can affect the environmental pH medium (Pometto III and Crawford, 1986). From the incubation process, pH medium tendsto change to base. This might happen because there is product accumulation which is simple residual sugar that provides from hydrolyzing cellulose. The change in pH number in the incubation process is affected by enzyme activity because the change in pH can change catalytic groups and enzyme conformation, and because of those, ionic character from carboxyl group and amine group from the enzyme will easily be affected by the pH.

From this research, the optimum cellulase enzyme activity is at 6.4 to 6.9. If the enzyme activity is higher, the residual sugar will also be higher. The enzyme activity is affected by pH, because of the ionic character from carboxyl group and amine group which can easily be affected by the pH. The change in pH medium or the unsuitable pH will make catalytic group and enzyme conformation changed. In addition, the change in the pH will make enzyme denaturation and the loss of enzyme activity. Based on that, it can be said that the acidity degree of medium can affect the enzyme activity.

The lowest residual sugar concentration and total carbohydrates from the treatments werethe samples which werereadded by buffer as much as 0.2%, 0.5%, and 0.75%. From those treatments, pH medium was6.4 to 6.9. If the residual sugar and carbohydrates total werelower after 24 hours fermentation, it showed that there washigh activity of microbes because there wasmore glucose which wasused by the microbe. If there is higher number in rumen microbe, the microbe protein concentration will also be higher. From this research, it showed that microbe protein concentration had non-significant difference (P>0.05), and, probably, because the high microbe activity wasnot expected to affect the microbe protein number.

CONCLUSION

From this research, it can be concluded that supplementation of NaHCO₃ buffer affected the medium pH. The collected data such as microbe protein, residual

sugar, and carbohydrates total were used in the fermentation process so that optimum degradation of cellulase enzyme could happen at pH range from 6.4 to 6.9.

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INDONESIAN LOCAL GOAT PRODUCTIVITY AND INCOME OVER FEED COST FED SUPPLEMENTED UREA PALM SUGAR BLOCK

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ABSTRACT

The role of ruminant microorganism using urea palm sugar block (UPSB) supplement in Indonesian local goat productivity was not fully studied. The objective of this study was to evaluate the utilization of agricultural waste product in the form of UPSB supplement on Indonesian Local goat productivity and income over feed cost of household farmers. Fresh local grass (*Brachiaria mutica*) was fed *ad libitum* without UPSB supplement (as control (R₀)), fresh local grass (*Brachiaria mutica*) *ad libitum* with UPSB supplement level of 75 g per day (R₇₅), fresh local grass (*Brachiaria mutica*) *ad libitum* with UPSB supplement level of 100 g per day (R₁₀₀), fresh local grass (*Brachiaria mutica*) *ad libitum* with UPSB supplement level of 125 g per day (R₁₂₅), and fresh local grass (*Brachiaria mutica*) *ad libitum* with UPSB supplement level of 150 g per day (R₁₅₀). 2(5x5) Double Latin Square Design with the period of 10 days for preliminary period and 15 days for data collection. Results showed that treatments of R₁₀₀, R₁₂₅, and R₁₅₀ significantly ($P<0.05$) increased production of the Indonesian local male goats as reflected linearly by higher average daily gain (0.121 kg to 0.127 kg) compared with control ration (R₀) and treatment of R₇₅ of 0.112 kg and 0.117 kg, respectively. There was a significant ($p<0.05$) different on average daily gain for bucks to be greater than that of ewes (0.120 kg and 0.110 kg), respectively. In addition, the income over feed cost (IOFC) increased linearly for about 381 IDR/head/day (bucks) and about 192 IDR/head/day (ewes) at the treatment of R₁₅₀ compared with those fed control ration (R₀). There was a significant tendency ($p<0.01$) of IOFC for bucks (9,225 IDR/head/day) to be greater than that of ewes (8,362 IDR/head/day).

Keywords: *Indonesian local goat, Productivity, Supplement, Urea palm sugar block.*

INTRODUCTION

Indonesian local goats in rural areas were owned by household farmers. They have adapted to harsh environment under hot and humid climate as well as low-quality feed to produce meat with high quality (Devendra and Burns 1994). The local goats play a role for increasing income of smallholder animal agriculture in North Sulawesi province of Indonesia. However, they are not intensively managed by adding feed supplement using local feed ingredients. Animal growth in developed farm system was generally measured by average daily gain; in addition, body size was generally detected by increase of chest girth and body length (Afolayan et al., 2006; Bozkurt 2006; Ozkaya and Bozkurt 2008; Fajemilehin and Salako 2008).

In the dry season, natural pasture decreases in nutritive value and improved grasses cannot grow. Therefore, it is important to find an alternative feeding system because purchased supplements are too expensive for poor farmers (Silivong and Preston 2015). The Indonesian local goats are raised by smallholder under traditional management using local grass around coconut plantation and opened grass field areas. Strategy to increase Indonesian local cattle productivity was utilization of the potential local agricultural waste product in form of the urea palm sugar block

(UPSB) supplement. The UPSB supplement was composed of easily fermented carbohydrate (waste product of local palm red sugar), fermented Nitrogen source (urea), undegradated protein source (fish meal, coconut meal), long chain fatty acid source (rice bran, coconut meal), and other mineral source (zeolit or CaCO₃). These nutrient contents were formulated in UPSB supplement that are supporting efficiently growth and development of microorganism in animal rumen (Merchen and Titgemeyer 1992; Gerson et al 1985).

Growth of Indonesian local goats fed 7 percents dry matter per kg body weight increased nutrient consumption per head per day compared with those of 5 percents dry matter per kg body weight (Suranindyah et al 2013). Growth of small ruminant animal (Ettawah crossbred goat) supplemented with the urea palm sugar block increased average daily gain (ADG) of 150.67 g compared to those of unsupplemented ration as control treatment of 93.1 g (Budiarso and Papatungan 2001). The objective of this study was to evaluate the utilization of agricultural waste product in form of the urea palm sugar block (UPSB) supplement on the Indonesian local goat production and income over feed cost of household farmers.

MATERIALS AND METHODS

Animal location and measurement

This study was conducted at Wori village, Wori district of North Minahasa Regency, North Sulawesi province of Indonesia, during 125 days of data collections divided into four periods (November 2014 to February 2015). This North Minahasa regency is categorized as agricultural areas with altitude of 50-100 m above sea level. It is characterized by hot and humid climate (28-32°C of temperature and 70-80 percents of humidity). Each period of treatment was used for resting period as preliminary treatment (treatment adaptation) taking of 10 days and data collection period taking of 15 days. Treatments involved the Indonesia local goats of five bucks and five ewes at the ages of 18 to 24 months old with the weight averages of 14 ±0.6 kg and 12±0.7 kg live weight (LW), respectively. Each animal was raised in each pen (0.75 x 1 m). Animals were weighed directly using the indicator digital electrical scale with capacity of 2000 kg.

Formulation of urea palm sugar block (UPSB) feed supplement

Formulation of the urea palm sugar block (UPSB) feed supplement was applied in the study location using ingredient of all local agricultural waste products with the composition and procedures as follows: (1) Material feed ingredients were weighed with the compositions of waste palm red sugar (50%), urea (4%), rice bran waste product (26%), coconut meal waste product (9%), animal bone meal waste product (6%), salt (2%), cattle mineral (3%); (2) Waste palm red sugar was added with fresh water (ratio of 1:2); (3) Rice bran waste product, coconut meal waste product, animal bone meal waste product, salt and cattle mineral were homogeneously mixed; (4) The mixtures of waste palm red sugar and water were gradually filled into the mixtures in the third procedure by gradually shaking them to form homogeneous batter and heated above fire heater for about 3 to 4 minutes, and thereafter fire heater was turned off; (5) Batter of urea palm sugar block was weighed in four formulation weights of 75 g, 100 g, 125 g and 150 g and wrapped into plastic bag. (6) The urea palm sugar block batter wrapped in the plastic bag was pressed by pressing tool for five minutes to form UPSB feed supplement.

Ration treatment

Rations fed to animals were formulated as follows:

- R₀ : Fresh Local grass (*Brachiaria mutica*) *ad libitum* without urea palm sugar block
R₇₅ : Fresh Local grass (*B. mutica*) *ad libitum* + 75 g of urea palm sugar block
R₁₀₀ : Fresh Local grass (*B. mutica*) *ad libitum* + 100 g of urea palm sugar block
R₁₂₅ : Fresh Local grass (*B. mutica*) *ad libitum* + 125 g of urea palm sugar block
R₁₅₀ : Fresh Local grass (*B. mutica*) *ad libitum* + 150 g of urea palm sugar block

Treatments were applied using double Latin Square design of 2(5x5) with the period of 10 days for preliminary period and 15 days for data collection. Fresh local grass (*Brachiaria mutica*) was prior to being fed to animals weighed and chopped (Figure 1). Drinking fresh water was provided *ad libitum* to the trial animals. Feed supplement of UPBS was fed daily to animals based on treatments (75g, 100g, 125g, 150g) per animal per day.

Data collection was done during fifteen days after finishing preliminary period (animal adaptation for each ration treatment) of 10 days. Samples of ration (fresh local grass) and UPSB feed supplement were collected at the initial and end of the study in each period of treatment. The left over grass feeding was collected daily. Daily difference between feed consumed and the left over grass feeding (ration) was defined as the animal feed consumption. Animal feed consumption was converted into dry matter consumption (kg unit) per animal per day. Table 1 gives nutrient compositions of the trial animal ration. Nutrient compositions of the ration treatment were presented in Table 2.

Variables observed

Variables observed in the trial included:

- Dry matter consumption of ration in gram per animal per day (g/animal/day), calculated as daily difference between feed consumption and the left over ration, and then converted into dry matter consumption.
- Average daily gain (ADG) in gram per animal per day (g/animal/day), calculated as difference between animal live weights at the end and at initial period of research divided by fifteen days of data collection.
- Feed conversion, calculated as ratio between dry matter consumption of ration (g/animal/day) and the ADG (g/animal/day).
- Income over feed cost (IOFC), calculated as difference between price of ADG in the unit of Indonesian rupiah (IDR) and daily feed consumption costs of local grass (dry matter weight) and UPSB supplement, all in IDR per animal per day (IDR/animal/day).

Statistical analysis

Data were analyzed using Analysis of variance (ANOVA) (Steel and Torrie 1980). Dry matter consumption, ADG, feed conversion and nutrient digestibility were included as dependent variable, while five treatments levels of urea palm sugar block were included as independent variables in the ANOVA model (Steel and Torrie 1980). Data were analyzed using the Insert Function Procedure of the related statistical category in datasheet of Microsoft Office Excel (2007). The significant difference in the model of treatments was tested using *honestly significant difference*,

while differences between variable averages in animal sex were tested using pair *t*-test (Byrkit 1987).

Table 1. Nutrient contents the ingredient ration of trial animals

Ingredient*	Nutrient contents (%)					Gross Energy (kcal/ kg)
	Crude Protein	Fat	Crude fibre	Calcium	Phosphorus	
Local grass (<i>B. mutica</i>)	13.42	3.07	31.54	0.44	0.19	3290
Rice bran waste product	10.12	7.98	18.72	1.06	8.97	2400
Coconut meal waste product	19.44	11.2	12.5	0.79	0.85	2400
Waste palm red sugar	0.63	0.11	0.26	0.12	0.12	3872

*) Results of laboratory analysis of the Faculty of Animal Science, Sam Ratulangi University, Manado, Indonesia (2014).

Table 2. Nutrient compositions of the ration treatment

Ration treatments	Dry matter (%)	Protein (%)	Energy (kcal/kg)
<i>Brachiaria mutica</i> (R ₀)	18.21	11.01	2864
<i>B. mutica</i> + 75 g of urea palm sugar block (R ₇₅)	31.1	12.4	2852
<i>B. mutica</i> + 100 g of urea palm sugar block (R ₁₀₀)	36.9	13.6	2907
<i>B. mutica</i> + 125 g of urea palm sugar block (R ₁₂₅)	41.8	14.2	2961
<i>B. mutica</i> + 150 g of urea palm sugar block (R ₁₅₀)	43.3	14.9	2979
Urea palm sugar block	80.97	5.21	3657

*) Results of laboratory analysis of the Faculty of Animal Science, Sam Ratulangi University, Manado, Indonesia (2014).

RESULTS AND DISCUSSION

Dried matter consumption

Data of dry matter consumption was calculated on the basis of the total dry matter consumption of local grass (*Brachiaria mutica*) and feed supplement of urea palm sugar block as presented in Table 3. Results of ANOVA showed that animals used in this study fed dry matter consumption with the average values ranging from 0.659 to 0.692 kg. However, the periods of data collection of dry matter consumption in animals were considered significant at $P < 0.05$. Increase of dry matter consumption in each additional period of treatment might be due to linear increasing development of animal growth. In focused case of treatment, it showed that animal's dry matter consumption of the local grass (*Brachiaria mutica*) without urea palm sugar block feed supplement (R₀) of 0.66 kg was not significantly different ($P > 0.05$) from that of the local grass (*Brachiaria mutica*) with UPSB feed supplement of 150 g (R₁₅₀) of 0.68 kg. This might indicate that the urea palm sugar block feed supplement of 150 g was not able to increase animal's appetite in dry matter consumption.

In this study, increasing levels of the urea palm sugar block feed supplement of 100 to 150 g tended to increase animal's appetite in dry matter consumption, ranging from 0.67 to 0.68 kg. Higher level of the urea palm sugar block feed supplement, ranging from 100 to 150 g in animal rumen might stimulate the development of ruminal microorganism in decomposing high crude fiber of grass (Elliot and Armstrong 1982; Nolan et al 1989; Merchen and Titgemeyer 1992). This study of UPSB feed supplement composed of urea, rice bran, coconut meal, palm red sugar

and other minerals might stimulate animal’s appetite to consume dry matter in ration. This study was in agreement with studies reported by Van Soest (1991) and Klusmeyer et al (1991).

The average consumptions of dry matter of animal in this study, ranging from 0.66 to 0.69 kg at body weight average of 21 to 26 kg are lower than those of dry matter consumption of goat at body weight of 37 to 42 kg, ranging from 1.587 to 1.764 kg per head per day (Suranindyah et al., 2013). In this study, there was a significant tendency ($P<0.05$) of dry matter consumption for bucks to be greater than that of ewes, (0.684 kg vs 0.671 kg), respectively (Table 3).

Table 3. Averages of dry matter consumption of local grass (*Brachiaria mutica*), daily gain, and feed conversion of animals in each treatment of the urea palm sugar block supplement (kg/animal/day)

Average of dry matter consumption in each treatments						
Sex	R ₀	R ₇₅	R ₁₀₀	R ₁₂₅	R ₁₅₀	Average
M	0.677 ± 0.007	0.678 ± 0.007	0.685 ± 0.006	0.687 ± 0.005	0.693 ± 0.006	0.684±0.007 ^y
F	0.659 ± 0.009	0.667 ± 0.011	0.673 ± 0.006	0.677 ± 0.007	0.681 ± 0.009	0.671±0.009 ^z
(P =0.000844)						
Average of animal daily gain in each treatments						
Sex	R ₀	R ₇₅	R ₁₀₀	R ₁₂₅	R ₁₅₀	Average
M	0.112 ± 0.002 ^a	0.117 ± 0.004 ^{ab}	0.121 ± 0.005 ^b	0.124 ± 0.007 ^b	0.127 ± 0.007 ^b	0.120±0.006 ^y
F	0.104 ± 0.002 ^a	0.109 ± 0.003 ^{ab}	0.111 ± 0.004 ^b	0.111 ± 0.003 ^b	0.117 ± 0.003 ^b	0.110± 0.005 ^z
(P =0.000433)						
Average of feed conversion in each treatments						
Sex	R ₀	R ₇₅	R ₁₀₀	R ₁₂₅	R ₁₅₀	Average
M	6.045 ± 0.10 ^a	6.002 ± 0.11 ^a	5.756 ± 0.08 ^b	5.678 ± 0.09 ^b	5.727± 0.08 ^b	5.842±0.169 ^y
F	6.046 ± 0.11 ^a	6.064 ± 0.13 ^a	5.956 ± 0.06 ^b	5.786 ± 0.06 ^b	5.820± 0.07 ^b	5.934±0.127 ^z
(P = 0.04606)						

M (male) = Bucks; F (female) = ewes;

^{ab} Means in the same row are different at $p<0.05$

^{y,z} Means in the same column are different at $p<0.05$

Average daily gain

Results of variance analysis showed that animals used in this study performed ADG values ranging from 0.112 to 0.127 kg in sbucks and 0.104 to 0.117 kg in ewes (Table 3). In this study, increasing levels of the urea palm sugar block feed supplement of 100 to 150 g increased animal’s ADG significantly ($p< 0.05$), ranging from 0.121 to 0.127 kg in bucks and 0.111 kg to 0.117 kg in ewes compared to ADG for control ration (R₀) and R₇₅ of 0.112 to 0.117 kg in bucks and 0.104 to 0.109 kg in ewes (Table 3).

In this study, it is clear that higher levels of the UPSB feed supplement, ranging from 100 to 150 g stimulated development of ruminal microorganism in decomposing high crude fiber of grass (Nolan et al 1989; Merchen and Titgemeyer 1992). High rating decomposition of crude fiber by ruminal microorganisms caused increasing animal’s appetite in dry matter consumption reflecting to higher ADG. The microorganism playing role in the bio degradation process of grass crude fiber in the ruminants was cellulosic bacteria (Gerson et al 1985). In this study, there was a significant tendency ($p=0.000433$) of ADG for bucks to be greater than that of ewes in the treatment application (Table 3).

Feed conversion

Results of variance analysis showed that animals used in this study performed feed conversion average values ranging from 5.73 to 6.04 in bucks and from 5.82 to 6.05 in ewes (Table 3). In addition, the periods of data collection of feed conversion in each animal were considered not significant. The feed conversions in all periods within males and females were not significant. In addition, there was no high tendency of feed conversion between males and females within period. In this study, increasing levels of the urea palm sugar block feed supplement of 100 to 150 g decreased significantly ($p < 0.05$) animal's feed conversion, ranging from 5.73 to 5.76 in bucks and 5.82 to 5.96 in ewes compared with feed conversion for control ration (R_0) and R_{75} with 6.00 to 6.04 in bucks and 6.05 to 6.06 in steers, respectively (Table 3). There was significant tendency ($p = 0.04606$) of feed conversion for treatment within ewes (5.93) to be greater than treatment in bucks (5.84) as shown in Table 3.

The feed conversion for animals on a ration of local grass (*Brachiaria mutica*) with the UPSB feed supplement (R_{100}) was 5.76 for bucks, indicating that implementation of UPSB supplement of 100 g consumption of animal could change 5.76 kg of grass dry matter into 1 kg meat product. On the other hand, animals fed the ration of local grass without implementation UPSB supplement (R_0), required 6.04 kg dry matter to produce 1 kg meat product as descriptively shown in Figures 3 and Figure 4.

Income over feed cost (IOFC)

Income over feed cost was calculated as difference between price of ADG in the unit of Indonesian rupiah (IDR) and daily feed consumption costs of local grass (dry matter weight) and UPSB supplement, all in the IDR per animal per day (IDR/animal/day). Based on the consumption costs of local grass (*Brachiaria mutica*) added by the UPSB supplement and prices of animal live weight and ADG, there were significant tendency of bucks to be greater than ewes for some variables of grass dry matter consumption ($p = 0.000844$), feed cost of grass dry matter consumption ($p = 0.000788$), price of ADG ($p = 0.000433$), feed costs of grass dry matter consumption + UPSB consumption ($p = 0.000788$), and IOFC ($p = 0.000502$). Therefore, the IOFC in bucks was more efficient of ten percents than that of ewes as calculated using the IOFC of both animal sex base averages as shown in Table 4. Economical analysis in terms of IOFC showed that animals fed UPSB at level of 100 to 150 g per animal per day produced IOFC ranging from IDR 9,233 to 9,446 compared to those without UPSB supplement of IDR 9,065; increasing by about 381 IDR/animal/day in bucks and about 192 IDR/animal/day in ewes compared to those fed control ration (without UPSB) as descriptively shown in Figure 1 and Figure 2.

Table 4. Income over feed cost (IOFC) derived from consumption costs of local grass (*Brachiaria mutica*) and the UPSB supplement and prices of animal live weight and average daily gain.

Cost and price components	Sex	Treatments					Average
		R ₀	R ₇₅	R ₁₀₀	R ₁₂₅	R ₁₅₀	
Grass dry matter consumption (GDMC) (kg/animal/day)	M	0.677	0.678	0.685	0.687	0.693	0.684±0.007 ^y
	F	0.659	0.667	0.673	0.677	0.681	0.671±0.009 ^z (<i>p</i> = 0.000844)
Feed cost of GDMC (IDR/animal/day)	M	1,015	1,017	1,027	1,030	1,039	1,026± 9.84 ^y
	F	988	1,000	1,009	1,015	1,021	1,007 ± 12.97 ^z (<i>p</i> = 0.000788)
Feed cost of Urea Palm Sugar Block consumption (UPSB-C) (IDR/animal/day)	M	0	473	630	788	945	-
	F	0	473	630	788	945	-
Average daily gain (ADG) (kg/animal/day)	M	0.112	0.117	0.121	0.124	0.127	0.120±0.006 ^y
	F	0.104	0.109	0.111	0.111	0.117	0.110± 0.005 ^z (<i>p</i> = 0.000433)
Price of animal live weight (IDR/kg)	M	90,000	90,000	90,000	90,000	90,000	-
	F	90,000	90,000	90,000	90,000	90,000	-
Price of ADG (IDR/animal/day)	M	10,080	10,530	10,890	11,160	11,430	10,818 ± 530.16 ^y
	F	9,360	9,810	9,990	9,990	10,530	9,936± 420.21 ^z (<i>p</i> = 0.000433)
Feed costs of GDMC + UPSB-C	M	1,015	1,490	1,657	1,818	1,984	1,593± 371.58 ^y
	F	988	1,473	1,639	1,803	1,966	1,574 ± 375.47 ^z (<i>p</i> = 0.000788)
Income Over Feed Cost (IDR/animal/day)	M	9,065	9,040	9,233	9,342	9,446	9,225 ± 174.94 ^y
	F	8,372	8,337	8,351	8,187	8,564	8,362 ± 134.42 ^z (<i>p</i> = 0.000502)

Note: M (male) = Bucks; F (female) = Ewes;

Ingredients and processing costs of 1 kg UPSB = IDR 6,300.-;

Price of 20 kg local grass (*B. mutica*) with 20% dry matter content = IDR 6,000.- or

Price of 1 kg grass dry matter = IDR 1,500.-

^{y,z}Means within the same column are different at *p*<0.05

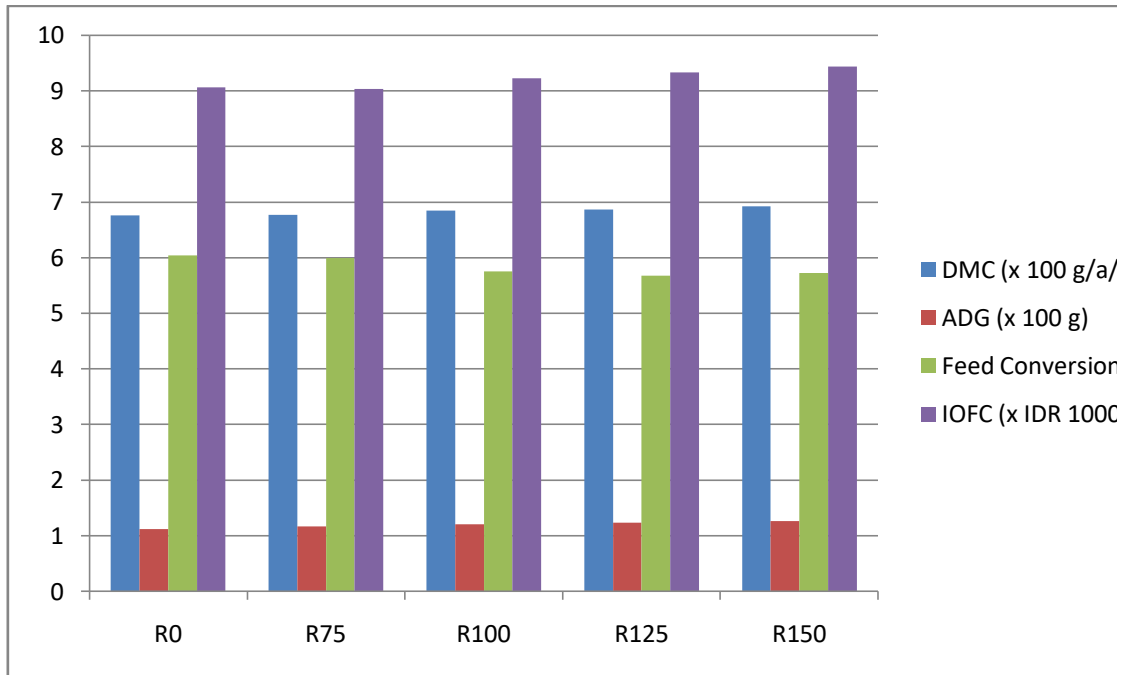


Figure 1. Supplementation effect of urea palm sugar block on the dry matter consumption (DMC), average daily gain (ADG), feed conversion, and income over feed cost (IOFC) in bucks

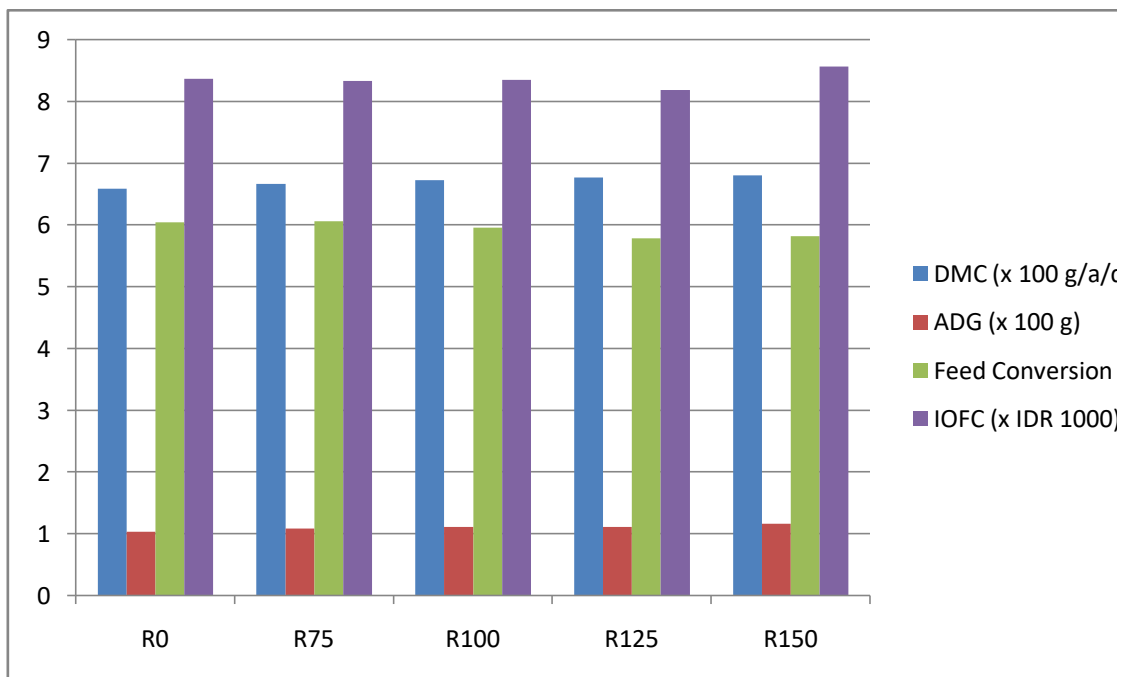


Figure 2. Supplementation effect of urea palm sugar block on the dried matter consumption (DMC), average daily gain (ADG), feed conversion and income over feed cost (IOFC) in ewes

CONCLUSIONS

- Supplementation of the agricultural waste product formulated in the urea palm sugar block (UPSB) of 100 to 150 grams significantly increased production of the Indonesian local goats as shown by higher average daily gain (ADG) of 111 to 127 g compared to the ADG of control ration without UPSB and 75 g of UPSB with 104 to 117 grams.
- Goats fed UPSB supplement at level of 100 to 150 g per animal per day produced IOFC ranging from IDR 9,233 to 9,446 compared to those without UPSB supplement of IDR 9,065; increasing by about 381 IDR/animal/day in bucks and about 192 IDR/animal/day in ewes compared to those fed control ration (without UPSB) and ration with 75 g UPSB.

ACKNOWLEDGEMENT

The financial support of the Ministry of Education and Culture, Republic of Indonesia through their Research Finance Program is gratefully acknowledged. The authors also acknowledge Refly Momongan and his farmer group members at Wori village, district of Wori, North Minahasa Regency, North Sulawesi Province of Indonesia for their assistance in animal data collection.

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EFFECT OF FERMENTED COCOA POD HUSK LEVEL IN CONCENTRATE TOWARD ETAWAH GRADE SEMEN QUALITY

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ABSTRACT

The goat farming is expected to make contributions of meat self-sufficient program because goat population in Indonesia is quite high. National data for goat population in Indonesia was 19,216,410 heads in 2014. The purpose of study was to determine the effect of fermented CPH supplementation level in concentrate toward semen quality of Etawah Grade Buck (EGB). This research was conducted from August to October, 2011 at experiment pen and Feeds Science Laboratory, Faculty of Animal Science, Universitas Gadjah, Yogyakarta. Semen was collected from 4 heads of EGB with about 2 years old and weight approximately of 24 kg. Concentrate was main material of CPH fermentation with composition P0 0, P1 20%, P2 40% and P3 60% from total of ration. The composer was used by the *Aspergillus niger* BPT with density 9.1×10^7 of colony/g as many as 1% of CPH material (on dry matter). Semen collection was done every 1 time/week and to be done five times semen collection. An examination was consisting of volume, color, smell, consistency, and the pH. Meanwhile, for microscopic examination was motion of mass, individual, motility, viability of spermatozoa. pH of P0, P1, P2 and P3 were 6.95, 7.03, 7.50 and 7.79, respectively. Mass movement of P0, P1, P2 and P3 were +++, +++, ++ and ++, while viability of P0, P1, P2 and P3 were 79.57%, 79.47%, 59.97% and 51.13%, respectively. It was concluded that addition of CPH fermentation in concentrates up to 20% for of EGB has not affect to feed consumption but it has affect to decreasing of semen quality in both of microscopic and microscopic.

INTRODUCTION

The meat self-sufficient program although ended in the era of previous the government (2014) but the achievement of meat self-sufficient program based on domestic resources are be continued by the present government through a program broader scale namely development in an area. One of the objectives of this program was acceleration of increases in population through improving the reproductive system of beef cattle. The goat farming is expected to make contributions of meat self-sufficient program because goat population in Indonesia is quite high. National data for goat population in Indonesia on 2014 about 19,216,410 heads, which it is the highest population in Central Java reached 4,014,570 heads, while the population highest in the Sulawesi Island is South Sulawesi reached 643,359 heads and followed by Central Sulawesi about 615,200 heads (DGLAHR., 2014).

Increasing in population of the goat is through improving the reproductive system particularly the semen quality of semen. The good quality of semen has high fertilization when he has mating to ewe with high of pregnancy. One of the factors can affect on the goat semen quality with giving feed quality. The preparation of feed quality can use an agricultural waste that it is abundant available during harvest season. The cocoa pod husk (CPH) is an alternative as main matter for ruminants concentrate. The CPH is always available on farmers cocoa plantation which it is can be harvested throughout the year (Prabowo *et al.*, 2014).

The benefits value of CPH as a fodder can be increased by reducing of fraction fiberlike hemicellulose, cellulose and lignin (Munier et al., 2014). CPH has low crude protein content but it has high crude fiber content about 9.14 and 35.74%, respectively, while content of fraction fibers like neutral detergent fiber (NDF) and acid detergent fiber (ADF) are 58.78% and 47.04%, respectively (Alemawor et al., 2009), hemicellulose, cellulose and lignin are 6.01%, 35.33% and 38.78%, respectively (Laconi, 1998).

The decline of CPH fiber content fraction is fermentation methods using microorganism in both of aerobic or anaerobic conditions. Feeding of concentrates to Etawah Grade Buck (EGB) can improve the semen quality. The semen quality is determined by the ejaculate volume, color, pH, consistency, viability, motility abnormality and concentration of spermatozo (Husin et al., 2007). The purpose of study was to determine the effect of fermented CPH supplementation level in concentrate toward semen quality of Etawah Grade Buck (EGB).

MATERIALS AND METHODS

This research was conducted from August to October, 2011 at pen experiment and Feeds Science Laboratory, The Faculty of Animal Science, Universitas Gadjah, Yogyakarta. Equipments were used in this research such as artificial vagina (minitub), microscope, object glasses, glass cover, test tube, pipet, Neubauer haemocytometer improved, tissue paper, universal of pH indicator (Merck), hand counter, refrigerator, ice container, thermometer, hammer mill. Materials were used EGB semen, sterile water warm, vaseline, eosin, physiological NaCl 0.3 %, alcohol 70 %, feed, vitamins and drugs.

Semen was collected from 4 heads of EGB with about 2 years old and weight approximately of 24 kg. The health reproductive condition of animals should be normal, before feeding trials were given worm drug. Concentrates were given from the CPH with a measure that was enumerated 1x5 cm, which it was fermented in aerobic condition. The composer was used by the *Aspergillus niger* BPT with density 9.1×10^7 of colony/g as many as 1% of CPH material (on dry matter).

The CPH was fermented in seven days. Drying of CPH after fermentation process was 3-5 days depend on irradiating the sun. CPH dry was grounded using hammer mill by the size of screen in 5 mm. Feed was given to animals in form of fresh forage for King grass (*Pennisetum hybrid*) and concentrate which it was the portion as follows:

Table 1. The Portion Feeding Trials

Feed materials	Dietary treatments			
	P ₀	P ₁	P ₂	P ₃
King grass (<i>Pennisetum hybrid</i>) (g)	3,350	2,605	1,860	1,116
Groats soybean (g)	90	26	13	6
Fermented CPH (g)	-	124	297	482

The total of concentrates protein in concentrate was 12%. Feed was given every day to animals of 4.0% from live weight on dry matter basis (NRC, 1981). Concentrates were enriched by the addition of 2.0% minerals and 0.5% salt from total of concentrates material. Feeding trials were done twice a day, morning at 08.00AM and 03.00 pm, started feeding of concentrates and to be continued by King

grass chopping. Adaptation period of feed trial was done 15 days before started of the research. Water drinking was given unlimited (*ad-libitum*).

The semen collection was done using artificial vagina already cleaned, sterile and dried. Warm water with the temperature 45° C incorporated into the artificial vagina, then in the interior of the vagina artificial mouth was lubricated by vaseline. Teaser prepared was used to increasing the libido and get the good quality and to be done false mounting (Toelihere, 1981). Semen collection was done every 1 time/week and to be done five times semen collection. Semen collection was done started at 09.00AM after feeding. The frequency of collection was three times for each individual treatment. Semen was collected to be sheltered in tube inside and put it in a ice container and taken to laboratory for next analysis. Variable was observed in microscopic examination. An examination consisting of volume, color, smell, consistency, and the pH. Meanwhile, for microscopic examination was the motions of masses, individuals, motility, viability of spermatozoa according to procedures of Hardijanto et al. (2008).

Motility of spermatozoa or impulse spermatozoa was determined in the mass movement and the movement of individuals. Based on the results of the assessment of the mass movement of semen quality can be determined as follows: a) excellent (+++), which it is looks great waves, thick, dark, and many, like wisps of active black clouds move fast moving place; b) good (++), when visible waves of small, thin, sparse, less clear, and moving slowly; c) fair (+), if not visible waves but rather just individual movements active progressive; d) (N, necrospermia or 0) if only little or no movement of the individual.

Semen concentration was the number of spermatozoa per ml of semen. Directly calculation with hemocytometer that was by using a method of neubauer calculating room. The concentration of spermatozoa in the five chambered calculated according to a diagonal direction using a microscope with an enlargement 40x10. The formula to calculate the concentration of spermatozoa:

$$Y \times \frac{400}{80} \times \frac{200}{0.1} = Y \times 10,000 \text{ mm}^3 = Y \times 10,000,000/\text{ml}$$

Y = Totally of observation spermatozoa in 5 big box
 400 = Totally of small box in calculation room
 80 = Totally of small box in 5 big box
 200 = Dilution of 200 times
 0.1 = Volume of calculation box (mm³)

The percentage of life spermatozoa was a drop of a substance with eosin in one glass object sterile and one small drops sperm added and mixed evenly, to be observed under a microscope with 40x10 enlargement. Calculation of 100 to 200 semen cell and the best 500 semen cells (Toelihere, 1981) using hand counter. The life spermatozoa head was not absorb color but it was dead absorb a red color after that the percentage of the life spermatozoa to be determined.

The percentage of spermatozoa abnormally was after counting of spermatozoa concentration and this semen was used to calculate the percentage semen abnormally. Spermatozoa was calculated up to 200 cells on one or several views under the microscope 40x10 with enlargement, then to be determined the percentage of spermatozoa abnormally.

RESULTS AND DISCUSSION

Feed Consumption

The totally of feed can be consumed by animals in the first day and eighthday of the feeding trial is presented in table 2.

Table 2. Feed consumption

	Dietary treatments			
	P ₀	P ₁	P ₂	P ₃
Feed consumed* (daily):				
King grass (g)	2,727.47	2,366.45	1,654.90	1,116.00
Groats soybean(g)	90.00	26.00	13.00	6.00
Fermented CPH (g)	-	61,90	50,38	141,01
Feed consumed** (daily):				
King grass (g)	3,069.66	2,538.79	1,825,91	1,116.00
Groats soybean(g)	90.00	26.00	13.00	6.00
Fermented CPH (g)	-	80,15	197,71	298,39

* Feed collected in first day of feeding trial.

**Feed collected in eighthday of feeding trial.

Feeding trial of P₀ as a comparison that is not granted an additional CPH fermentation in concentrate so that the portion of the King grass more as basal feed and fiber source. While the P₁, P₂ and P₃ a bit more given the grass King because given additive of CPH fermentation as fiber source. The addition of groats soybeans were all treatments as the main protein source in the concentrate. According to Hartadi et al. (2005); Cordesse (1990), groats soybeans contain crude protein reached 41.3-41.7%. The addition of groats soybeans in rations on this research can fulfill the standards of the crude protein needs of animal growing about 12%.

The high feed consumption on P₀ was caused by feeding only from King grass and soybean groats. The results of observations during the research, soybean groats it was a favorite goat so that it can be spent in a short period of time that is around 5-10 minutes every time of feeding. The factors affect the feed consumption is the very nature of feed such as particles size, texture and scent. According to Preston and Leng (1987) that scent was an important factor of feed material. The animal can refused material feed given without feel because it was not fond of the scent. The other factors that were also affect of feed consumption like body sizes of animal, increasing feed consumption in line with increasing the size of animal (Zain, 2009).

Groats soybeans provided can be spent starting the first day until the eighth day collection of fodder though mixed with CPH fermentation, animal first was eat more groats soybeans and to be continued eating of CPH fermentation. This case is caused by the goats have selective properties in consuming the feed through the senses of smell so it takes longer for the adaptation of the new type feed is given. The selective nature of the goat in consuming the feed associated with the tendency of choosing a feed that is nonfibrous. According to Huston (1998) that was the goats have the ability to choose the type of feed to be consumed and preferred types of feed without fibrous.

Characteristic of Semen Macroscopic

The test semen quality under macroscopic examination such as semen of volume, color, smell, consistency, and the degree of the acidity or pH. The result of the observation of macroscopic in of the characteristics of fresh EGB semen in this research is still categories good and in the normal range of especially on P₀ and P₁,

while P2 and P3 relatively less than the normal range of such as are presented in table 3.

The semen volume of EGB on P0 and P1 were 0.90 ml and 0.87 ml, respectively, these volume are still in the normal range, relatively the same with the volume of EGB semen was reported by Tambing et al. (2001) with average 0.95 ml. The volume of goat semen has varies every collecting with range of 0.5-1.5 ml (Devendra and Burns, 1994). Semen color is still in the normal range which it is creamy to yellowish on all feed treatment, but if the semen color is white to pale and it would affect on other semen characteristics. The semen consistency for P0 and P1 had moderately viscous to viscous, while P3 the semen quality was relatively low marked with dilute consistency. Color, consistency and concentration have links to each others. The semen color more faded, and the spermatozoa concentration become decrease and to be followed by more dilute of semen (Mahmilia et al., 2006).

Table 3. Average of macroscopic characteristic of EGB semen

Semen characteristic	Dietary treatments			
	P ₀	P ₁	P ₂	P ₃
Volume (ml)	0.90	0.87	0.43	0.37
Color	cream-yellowish	cream-yellowish	cream-yellowish	yellowish
Odor	khas	khas	khas	khas
Consistency	moderately viscous-viscous	moderately viscous-viscous	dilute-moderately viscous	dilute
pH	6.95	7.03	7.50	7.79

Note: Analyzed in the Reproduction laboratory, Animal Science Faculty, UGM, 2011

Semen pH of P0 and P1 were 6.95 and 7.03, respectively, semen pH in this research is still in the normal range to the EGB. According to Bearden Fuquay (1984), good quality of semen had a pH with a little acid from 7,0 and in average of 6.7. Semen pH in this research is closed to previous studies of semen pH for EGB. The results of previous studies reported that pH ranges about 6.85-7.13 for semen EGB (Tambing et al., 2000; Tambing et al., 2001). P2 and p3 had pH above the range of goat pH in both of 7.50 and 7.79 which is leads to the alkalis. The reaction of alkalis in spermatozoa is often associated with the low quality and low fertility (Perry, 1969).

The low semen quality in macroscopic in P2 and P3 had relation with feed consumption because during feed consumption was not optimal especially feed concentrates which was the more portion of CPH fermentation. P1 had low portion of CPH fermentation and higher portion of soybean groats in concentrate, while P0 had not give CPH fermentation and given higher portion of soybean groats. Groats soybean is a source of high protein that affects on the formation of the semen quality. The analysis of chemical composition was groats soybean having high crude protein content reached 33.07%, to be supported by organic matter 89.93% and crude fat 7.72%. The chemical composition of fresh semen plasma had high protein content about 214 mg/100 ml (Tambing et al., 2003) or 4.59% as component in semen plasm.

Characteristic of Semen Microscopic

Microscopic examination of goat semen is massmotions, individualmotions, concentration, motility, viabilitas and abnormality spermatozoa. Table 4 is show average of characteristic for EGB semen microscopic.

Table 4. Average of microscopic characteristic of EGB semen

Semen characteristic	Dietary treatments			
	P ₀	P ₁	P ₂	P ₃
Mass movement	+++	+++	++	++
Motility (%)	78.33	76.67	51.67	46.67
Concentration (ml)	2.40 x	2.24 x	1.01 x	0.96 x
Viability (%)	10 ⁹	10 ⁹	10 ⁹	10 ⁹
Abnormality (%)	79.57	79.47	59.97	51.13
	5.59	5.93	10.63	16.77

Analyzed in the Reproduction laboratory, Animal Science Faculty, UGM, 2011

The mass movement of EGB semen of P₀ about +++ and P₁ +++, these treatment were includes the categories excellent marked with visible large waves, thick, dark, and many, like wisps of active black clouds move fast moving place. It is supported with the high motility of the sperm in P₀ and P₁ was 78.33% and 76.67%, this condition indicates that the individual movement of sperm is quite progressive. It is powered by a low sperm abnormality in P₀, P₁ and P₂ about 5% 5.93% and 10.63%, respectively, but P₃ had more over from standard recomendation reached 16.77%. The semen concentration in all the feed treatments was still in the normal range of 0.96-2.40 x 10⁶/ml, but it is still lower than the research results Tambing et al. (2001), 2.94 x 10⁶/ml. Hafez (1987) recommend that the terms of semendeluted that it may had mass movement, individual movement more over 65%, abnormality spermatozoa in range 14-15%. While Toelihere (1981) reported that the minimum standards for the semen quality which can be used for artificial insemination (AI) was the minimum to contain 500 million cells/ml/mass movement/ejakulate, as well as 50% in borh of semen viability and motility. The characteristics of EGB semen on this research especially P₀ P₁ and P₂ can be used to the AI program.

CONCLUSION

Based on the results of this research it can be concluded up as follows:

1. The addition of CPH fermentation in concentrates up to 20% for of EGB has not affect to the feed consumption.
2. The addition of CPH fermentation in concentrates up to 20% for of EGB has not decreasing of the semen quality both in microscopic and microscopic.

ACKNOWLEDGEMENTS

The authors gratefully acknowledge to Mr. Wartono and Isnawan as Staff in Reproduction Laboratory, Animal Science Faculty, Universitas Gadjah Mada Yogyakarta for their colaboration and helpull during sperm collection and sperm quality analysis in Reproduction Laboratory.

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THE EFFECTIVENESS OF PADDY FIELD MUD ON CULTIVATION OF *AZOLLA PINNATA* AS A HIGH PROTEIN FORAGE

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ABSTRACT

Aquatic macrophytes, such as *Azolla pinnata* has a wide range of adaptation in growth and fast yield time. This plant has high nitrogen fixation ability and potency as high protein forage. The aim of this research was to measure production potency and protein content on several media. This research was designed using factorial completely randomized design with two factors. Factor A was paddy field mud 10% and factor B was media (control, *hoagland*, and *hyponex* 0.1%). The measured parameters were dissolved N absorption, doubling time, cover area, biomass production, and protein content. Data were analyzed using ANOVA and the results showed that *Azolla pinnata* has high dissolved N absorption ($\geq 97.51\%$). Additional of paddy field mud and kinds of media treatments significantly shortened doubling time, cover area, biomass production, and protein content compared with control (without paddy field mud) ($p < 0.05$).

Keywords : Aquatic macrophytes, *Azolla pinnata*, high protein forage

INTRODUCTION

Forages are the main sources of feed for ruminant that determine livestock performance and productivity. Forage production is high at rainy season and low at dry season with relatively lower forage quality. The land use status for pasture was limited because of the other transformation. So that, it is necessary to improve aquatic plants potency as a potential aquatic area utilization in Indonesia.

Azolla pinnata is an aquatic fern which is potential as an alternative forage source of protein and mineral. Protein content of *Azolla pinnata* is 21–37% (Khan 1988; Alalade and Iyayi 2006) and protein digestibility is 84% from total crude protein. Essential amino acid (lysine and methionine) on *Azolla pinnata* is quite high with the content of lysine and methionine of 0.98% and 0.34% respectively (Alalade and Iyayi 2006). *Azolla* is rich in macro and micromineral such as Fe, Ca, P, Mg, Cu, and Mn (NDDDB 2012), so that it has potential as a mineral supplement for ruminant (Parashuramulu *et al.* 2013).

Azolla pinnata has a short yield time (7–20 days) and a fast fresh biomass production of 390 tons per hectare a year (Ferentinos *et al.* 2002). *Azolla* has many benefits such as a biofertilizer, human food (Pabby *et al.* 2003), ruminant feed, fish food, herb (Mithraja *et al.* 2011), biogas production, and phyto remediation (Kempenet *et al.* 2013). The development of aquatic plant (*Azolla*) as an alternative forage source has a high potential to support performance and productivity of livestock. Lately, the study about *Azolla pinnata* based on media type was limited. The aim of this research is to measure production potency and protein content on several media.

MATERIALS AND METHODS

The materials used on this research consists of culture box (36×28.5×10 cm³), scale, pH meter, oven, cooling box, and furnace. The materials used were *Azolla pinnata* R Br, water, *hyponex*, *hoagland* media, and paddy field mud.

Methods of this research was designed using factorial completely randomized design with two factors. Factor A was paddy field mud 10% and factor B was media (control, hoagland, and hyponex 0,1%). The measured parameter were dissolved N media absorption, doubling time cover area, biomass production and protein content. Data were analyzed using Anova.

RESULTS AND DISCUSSION

Azolla pinnata R Br is an aquatic plant that lives well on stagnant water, such as paddy field, swamp, and lake. Mutualism symbiotic between *Azolla* plant with *Cyanobacteria* such as *Anabaena azolla* because this plant has a high growth ability (Arifin 2003). *Anabaena azollae* has a high nitrogen fixation ability from the air (van Reine and Trono 2001). The ability to grow fast and good quality improve the potential of this plant as a cultivate crop for feed. The environment condition such as the plantation media, is one of the factor that determines productivity and quality of *Azollapinnata*.

Nitrogen absorption effectivity of *Azollapinnata* on culture media

The results of N absorption from culture media showed that absorption value for each treatment was high, that is more than 97.51% from total dissolved nitrogen in media (Table 1). This high absorption value is not fully used by plant to grow. The total nitrogen content in media could be lost through several factors, such as plant immobilization, evaporation during N mineralization, and N-nitrate denitrification. The total amount of nitrogen that is absorbed by plant will affect productivity and quality of plant. Nitrogen plays an important role in forming all protein, chlorophyle, coenzyme, amino acids, and also growth hormones like cytokinin and auxin (Hanafiah 2010). A plant needs nitrogen supply at every stage of growth, moreover at early stage of growth. Cedergreen and Madsen (2002) said that aquatic plant has an ability on absorbing N element in the form of NH^{4+} and NO^{3-} through its roots and leaves.

Table 1. The absorption of N media in *Azollapinnata* culture

Media	Without paddy field mud	With paddy field mud
 %	
Control	98.38	97.51
Hoagland	99.61	99.52
Hyponex	98.67	98.77

Doubling timecover area (CA) of *Azolla pinnata* based on culture media

Azolla plant's CA doubling time is influenced by the characteristics of the planting media. Additional treatment of paddy field mud and fertilizer showed significant effect ($p < 0.01$) to the doubling time of CA as presented in Table 2. The addition of paddy field mud and manure effectively accelerate *Azolla pinnata* doubling time from 6.38 days to 3–4 days. This doubling time acceleration is reasonable because the addition of fertilizer and paddy field mud improves nutrients adequacy for plants in its metabolism process. Hardjowigeno (1995) stated that the plant productivity is very determined by the soil nutrient status and the use of

fertilizer, so the nutrient supply is adequately well. *Azolla pinnata* has replication capability in 3–10 days (Hasan and Chakrabarty 2009).

Table 2. Doubling time cover area *Azollapinnata* based on culture media

Media	without paddy field mud		with paddy field mud	
 day			
Control	6.38 ±	0.29B	4.24 ±	0.13A
Hoagland	4.06 ±	0.04A	3.91 ±	0.3A
Hyponex	4.34 ±	0.13A	4.61 ±	0.34A

Biomass production of *Azolla pinnata* base in culture media

The results showed that the addition of paddy field mud and fertilizer to the *Azolla* culture media significantly increased ($p < 0.01$) its biomass production (Table 3). The best increase in biomass production is in the Hoagland media, both with paddy field mud or without. This indicates that the nutrient adequacy in Hoagland media is better than the others. According to Taiz and Zeiger (2002) that Hoagland has a complete nutritional content consisting of macro nutrients and micronutrients. Prihantoro *et al.* (2014) stating that *Lemnaminor* productivity is optimal when added with fertilizer that contains a complete macro- and micro-minerals needed for plant growth.

Table 3. Fresh biomass production rate of *Azollapinnata* based on culture media

Media	without paddy field mud		with paddy field mud	
 g			
Control	15.31 ±	3.31C	36.64 ±	13.21AB
Hoagland	61.52 ±	6.17A	50.46 ±	12.37A
Hyponex	43.12 ±	9.55AB	44.33 ±	16.66AB

Protein content of *Azollapinnata* based on culture media

The results showed that the addition of paddy field mud and fertilizer significantly ($p < 0.05$) increase the protein content of *Azolla pinnata* (Table 4). Hoagland and hyponex fertilizer treatment is significantly ($p < 0.05$) better in improving the crude protein content of *Azolla pinnata* compared to control. This increase of protein value illustrates the plant’s high effectiveness in using plant nutrient component from the media that will have a positive impact on improving the protein content. Hoagland and hyponex are fertilizers that contain nutrients designed for leaves and roots growth where macro-micro nutrients are available to plants so that can be directly utilized by the plant. According to Parman (2007) that increased nutrients uptake will contribute to an increase in plant protein synthesis which resulted in increased protein content in plant tissues

Table 4. Crude protein content of *Azollapinnata* based on culture media

Media	without paddy field mud		with paddy field mud	
 %			
Control	14.86 ±	0.84c	19.22 ±	0.61b
Hoagland	27.19 ±	0.05a	22.24 ±	2.35ab
Hyponex	27.56 ±	2.87a	27.59 ±	0.37a

CONCLUSION

Azollapinnata has high dissolved N absorption ($\geq 97.51\%$), Additional of paddy field mud and kinds of media treatments significantly shortened doubling time cover area, biomass production and protein content compared with control.

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THE EFFECT OF DIFFERENT TYPES OF ORGANIC POTS AS PLANTING MEDIUMS ON GROWTH AND PRODUCTIVITY OF COWPEA (*Vigna unguiculata*) PLANTED IN SANDY LAND

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ABSTRACT

The study was done to determine the growth and productivity of cowpea planted in sandy land using organic pots as planting medium. Five types of organic pots were made from chicken excreta (COP), goat (GOP) and cows (SOP) manure composts, biogas sludge (SOP), and leaf compost (LOP). These organic pots were filled with soil before 2 cowpea seeds were planted. Three blocks of sandy land (for replication) were used in this study. Each block was randomly divided into 6 plots and 5 type of organic pots and a control (without pot) were placed in the plot. Plant height and number of leaf were observed weekly during 8 weeks. Root nodules were weighed after separated from root, and forages were measured after harvesting of the plant at the end of the study. Data were analyzed using analyse of variance for completely randomized block design and the difference between means were tested using Duncan's Multiple Range Test. The results showed that plant height (54.20 cm) and weight of root nodule (2.12 g) cowpea planted in COP were significantly higher ($P < 0.05$) than that other organic pots. However the types of organic pots had no significantly effect on number of leaf and plant production. Plant height (58.20 cm), number of leaf (134.41), dry matter production (41.21 g), organic material production (39.18 g), and crude protein production (5.65 g) cowpea planted in sandy land in block II were significant higher ($P < 0.05$) than that in other blocks. In general organic pot made from chicken manure (COP) was better than the other organic pots. It can be concluded that use of organic pots as planting mediums in the sandy land can improve the growth and productivity of cowpea.

Keywords: Cowpea, Sandy land, Organic pot, Planting medium, Growth and productivity

INTRODUCTION

Limiting factors of utilisation sandy land were loose consistency of soil structure, low water holding capacity, porous and plant nutrients deficiency due to sand content exceed from 95%, and low productivity for growing plant in sandy land (Rinsema, 1986). Low productivity of plants cultivated in sandy land could be improved by organic fertilizer application, watering arrangements, and the use of organic pot. Organic pot was a pot made from organic compost material that was easily decomposed by soil microorganisms, and contained soil nutrients which were be able to be utilised as plants nutrients (Budi *et al.*, 2011).

The use of organic pots in sandy land was able to improve the plant growth environment, water and nutrients availabilities for the plants root developement. Organic compost has advantages on soil structure and water holding capacity of sandy land (Indriani, 2011). Partoyo (2005) reported that addition clay and animal compost in sandy land improve soil quality.

Cowpea (*Vigna unguiculata*) was one of drought resistant legumes that was able to be grown in dry or sandy land. As animal feed cowpea forage harvested at 60 and 70 days respectively contained 17.29 and 16.11% crude protein, and the dry matter

production was 2.04 and 2.08 ton/ha . However, *in vitro* dry matter digestibility was significantly higher for cowpea forage harvested at 60 days (67.17%) than that harvested at 70 days (61.77%) (Kotten et al., 2004). The dry matter production was going to be higher if cowpea planted as intercropping with sudan grass (Kotten et al., 2007).

This study was conducted to determine the effect of organic pot utilisation as plant growth medium on the productivity of cowpea planted in the sandy land. The utilisation of organic pot was one of efforts how did improve sandy land productivity.

MATERIALS AND METHODS

Three blocks of sandy land (as replicates) of 6 x 4 m² in Agriculture Training, Research and Development Center Gadjah Mada University located in Berbah Sleman Yogyakarta. Each block was randomly divided into 6 plots of 75 x 300cm² with plot space of 20 cm. Fives types of organic pots made from various composts those were chicken, goat, and cattle manure, biogas sludge and compost of plants waste (leaves, small branch, twig, straw, husk, etc.) and controls (without organic pot) were used in this study. The dimension of organic pot was 17 cm of height and 22 cm of diameter, with 1,864 cm³ of medium capacity. Each pot was filled fully with soil as plant medium, buried in a hole as a level in the sandy land with 60 cm of planting space (between pot). Two seeds of cowpea were planted (local variety) 5 cm under the land surface. Chemical fertilizers were SP 36 contained 36% of P₂O₅ and KCl contained 60% of K₂O were applicated 2-week after planting as a level of 10 g of SP 36/pots and 6 g KCl/pot, and only one plant was maintained on each pot. The plant watering was done every 2 days with 1.300ml of tap water in each pot. Plant growth as plant height and number of leaf were measured every week from second week to eighth week. Plant harvesting was done 70 days after planting, by cutting a whole of plant at 3-5cm above soil level. After harvesting, plant forage was separated with seed and root part, and weighed of each part. Sample of forage was dried in oven of 50°C for 5 days, ground using wiley mill with 1 mm of screen diameter for dry and organic matter, and crude protein analyses (AOAC, 1975).

Data of plant height, number of leaf and root nodules, and forage production were analyzed by analysis of variance of Randomized Complete Block Design (RCBD), and differences between means were tested using Duncan's test (Astuti, 1980).

RESULTS AND DISCUSSION

Plant Growth

Plant height

The plant growth was measured weekly through plants height during 8 weeks. The result in Table 1. showed that the plant height of cowpea planted in sandy land using differents type of organic pot as planting medium respectively was biogas *sludge* (55.87 cm), cattle compost (55.07cm), leaves compost (54.33cm), chicken excreta compost (54.20 cm), goat compost (59.60cm), and control without organic pot (50.60cm).

There was no significantly differents between type of organic pots on plant height. However, cowpea planted in the sandy land of block II (58.20 cm) was significantly higher (P<0.05) compared to that of the block III (52.17 cm). Plant height in the block I (54.47 cm) was not significantly different than that in the block

II, as well as in the block III (Table 1). The plant height measured up to 8 weeks-old of plants, therefore the plant nutrient needs was still able be met from the soil and P and K fertilizer added, most of the plants had not been able to use nutrients in organic pot . The soil characteristics in this study was dominated by sand fraction of fine to coarse sand, so that the soil had a coarse texture that affected soil nutrients availability (Darmawidjaja, 1997). Availability of nutrients in the soil affected plant growth and production. According Winarto and Kasno (1998) cowpea plant height was about 51.4 cm varied from 12.6 to 122 cm.

Table 1. Plant height (cm) of cowpea grown in sandy land using diferents type organic pot as planting medium

Type of pot made from compost of	Block of land			Mean ^{ns}
	I	II	III	
Biogas sludge	46.40±4.67	65.20±11.05	56.00±9.69	55.87±11.45
Cattle	60.40±4.98	51.00±15.59	53.80±12.21	55.07±11.65
Leaves	59.60±9.39	49.80±4.71	53.60±5.94	54.33±7.69
Chicken excreta	55.40±8.47	54.60±6.69	52.60±1.67	54.20±5.97
Goat	51.00±7.00	69.60±9.76	58.20±3.03	59.60±10.33
Control (no pot)	54.00±7.18	59.00±13.17	38.80±3.49	50.60±12.11
Mean	54.47±8.15 ^{pq}	58.20±12.29 ^q	52.17±9.07 ^p	

^{p, q} Different superscripts in the same row indicated significant differences (P<0.05)

^{ns} : non significant

Number of leaf

Number of leaf (Table 2) of cowpea grown on sandy land using planting medium of organic pots made from chicken excreta compost (122.80/plant) was significantly higher (P<0,05) than cattle manure (88.20/plant) and controls (85.27/plant), however, there were no significant different compared to biogas sludge (110.80/plant), leaves (104.27/plant), and goats manure compost (122.80/plant). The chicken excreta compost contained more N and P than cattle and goat manure and leaf compost. Nitrogen was a limiting nutrient for plant growth (Rinsema, 1986).

Table 2. Number of leaf of cowpea grown in sandy land using diferents type organic pot as planting medium

Type of pot made from compost of	Block of land			Mean ^{ns}
	I	II	III	
Biogas sludge	114.20	141.80	76.40	110.80 ^{ab}
Cattle	77.80	113.00	73.80	88.20 ^a
Leaves	108.80	113.80	90.20	104.27 ^{ab}
Chicken excreta	103.80	175.60	89.00	122.80 ^b
Goat	89.80	135.60	91.00	105.47 ^{ab}
Control (no pot)	87.00	125.20	43.60	85.27 ^a
Mean	96.90 ^q	134.41 ^r	77.33 ^p	

^{a, b, c} Different superscripts in the same column indicated significant differences (P<0.05)

^{p, q, r} Different superscripts in the same row indicated significant differences (P<0.05)

In the Table 2 showed that number of leaf of cowpea planted on sandy land of block I (96.90/plant) was higher (P <0.05) compared with cowpea planted on block III (77.33/plant) and number of leaf of cowpea planted on sandy land in block III (134.41/plant) was significantly higher than Block I and III. The plant growth was

affected by the soil fertility, environment conditions, and genetic factors. Increasing the nutrients quantity in the soil lead to increase in the of nutrients content in plants and plant growth (Foth, 1998).

Cowpea production

Production of dry matter

The dry matter production of cowpea grown on sandy land using organic pots made from biogas sludge, cattle manure, leaf compost, chicken excreta compost, goats manure, and control/no pot consecutively was 39.24 g, 31.95 g, 35.19 g, 34.63 g, 33.28 g, and 23.71 g/plant (Table 3). There no significant differences was detected between types of organic pots on dry matter production of cowpea.

Table 3. Dry matter production (g/plant) of cowpea grown in sandy land using differents type organic pot as planting medium

Type of pot made from compost of	Block of land			Mean ^{ns}
	I	II	III	
Biogas sludge	30.33±14.16	41.05±19.53	46.33±22.51	39.24±18.93
Cattle	33.61±8.94	45.14±21.45	17.10±10.27	31.95±18.06
Leaves	38.92±23.81	44.67±25.76	21.97±8.41	35.19±21.71
Chicken excreta	35.16±10.14	53.43±17.33	15.30±7.83	34.63±19.81
Goat	32.57±24.39	34.96±16.90	32.31±21.38	33.28±19.59
Control (no pot)	33.46±11.11	28.13±19.96	9.56±7.39	23.71±16.65
Mean	34.01±15.40 _q	41.23±20.25 ^a	23.76±18.14 ^P	

^{P, q} Different superscripts in the same row indicate significant differences (P<0.05)

^{ns} : non significant

The dry matter production (Table 3) of cowpea grown on sandy land of block III (23.76 g/plant) was significantly lower (P<0.05) compared with the block I of land (34.01 g/plant) and block II (41.23 g/plant). The difference in response might be caused by factors fertilizing plant species, stage of growth, the amount and timing of fertilizer application, soil nutrient levels, and climate (Rinsema, 1986). According Muhadjir (1988), dry matter production of plants was the balance between photosynthesis and respiration. The rate of physiological processes varied depend on the plant organ, age, climate conditions and plant cultivation.

Production of organic matter

The organic matter production (Table 4) of cowpea grown on sandy land using organic pots made from biogas sludge, cattle manure, leaf compost, chicken excreta compost, goats manure, and control/no pot consecutively was 37.45 g, 30.41 g, 33.60 g, 32.84 g, 31.62 g, and 22.73 g/plant. Organic matter was like dry matter production of cowpea grown in a control looked like lower than the others, but statistically not significant.

There no significant differences was detected between types of organic pots (Table 3). Organic matter production (Table 4) of cowpea grown on sandy land of block III (22.80 g/plant) was significantly lower (P<0.05) compared to the block I (32.35 g/plant) and block II (39.18 g/plant). Application of N fertilizer increased organic matter production, since photosynthesis rate improved tahan protein, carbohydrates, fats and vitamins formed increased (Tillman et al., 1998). Nutrients

from photosynthetic yield were accumulated and then transformed into organic matter during plant growth (Rosmarkam and Nasih, 2002).

Table 4. Organic matter production (g/plant) of cowpea grown in sandy land using different types of organic pot as planting medium

Type of pot made from compost of	Block of land			Mean ^{ns}
	I	II	III	
Biogas sludge	28.99±13.54	38.92±18.51	44.46±21.59	37.45±18.09
Cattle	31.50±8.37	43.26±20.56	16.47±9.89	30.41±17.25
Leaves	37.08±22.68	42.33±24.41	21.39±8.19	33.60±20.52
Chicken excreta	33.28±9.59	50.96±16.53	14.28±7.27	32.84±18.97
Goat	30.98±23.20	32.81±15.86	31.09±20.57	31.62±18.64
Control (no pot)	32.26±10.71	26.80±19.02	9.14±7.06	22.73±15.6
Mean	32.35±14.66 ^q	39.18±19.29 ^q	22.80±17.44 ^p	

^{p, q} Different superscripts in the same row indicate significant differences (P<0.05)

^{ns} : non significant

Crude protein production

The crude protein production (Table 5) of cowpea grown on sandy land using organic pots made from biogas sludge, cattle manure, leaf compost, chicken excreta compost, goats manure, and control/no pot consecutively was 5.27 g, 4.51 g, 4.53 g, 4.64 g, 4.54 g, and 3.27 g/plant. There were no significant differences detected on crude protein productions between types of organic pots (Table 5).

The effect of block on sandy land field was significant on crude protein production of cowpea (Table 5). Statistical analysis showed that the crude protein production of cowpea plants grown on land block III (3.11 g/plant) was lower (P<0.05) compared with the block I of land (4.64 g/plant) and block II (5.65 g/plant). There was no significant difference on crude protein production among the block I and the block II of sandy land. Soil type in this study was regosol contained 1.4% of organic matter, 0.06% of nitrogen, and 6.41% of water and clay, so this was rather limited for agriculture utilization (Brady, 1982). The composition of plant was affected by the soil, environment conditions, and genetic factors. Increasing the nutrients quantity in the soil could lead to an increase in the percentage of nutrients in plants (Foth, 1998).

Table 5. Crude protein production (g/plant) of cowpea grown in sandy land using different types of organic pot as planting medium

Type of pot made from compost of	Block of land			Mean ^{ns}
	I	II	III	
Biogas sludge	3.95±1.84	5.78±2.75	6.09±2.95	5.27±2.56
Cattle	5.27±1.40	6.05±2.87	2.21±1.32	4.51±2.52
Leaves	4.97±3.04	5.89±3.40	2.73±1.04	4.53±2.85
Chicken excreta	4.84±1.39	7.18±2.33	1.90±0.97	4.64±2.71
Goat	4.56±3.41	4.94±2.39	4.25±2.81	4.58±2.70
Control (no pot)	4.26±1.41	4.05±2.87	1.50±0.76	3.27±2.18
Mean	4.64±2.09 ^q	5.65±2.72 ^q	3.11±2.35 ^p	

^{p, q} Different superscripts in the same row indicate significant differences (P<0.05)

^{ns} : non significant

Root nodule

Weight of root nodules of cowpea grown on sandy land using planting medium of organic pots made from biogas sludge (0.40 g/plant), cattle manure (0.98 g/plant) and controls (0.63 g/plant) were significantly lower ($P<0,05$) than leaves compost (1.85 g/plant), chicken excreta compost (2.12 g/plant) and goats manure (1.86 g/plant) showed in Table 6. There were no statistically differences between biogas sludge, cattle manure and controls, as well as between leaves, chicken excreta and goats manure compost. Number of legume root nodules and wight have often been positively correlated to the amount of N fixed (Handarson and Danson, 1993).

Table 6. The mean weight of root nodules (g/plant) of cowpea planted in sandy land using differents type organic pot as planting medium

Type of pot made from compost of	Block of land			Mean ^{ns}
	I	II	III	
Biogas sludge	0.30±0.28	0.32±0.04	0.58±0.80	0.40±0.47 ^a
Cattle	1.56±0.49	1.04±0.39	0.34±0.05	0.98±0.61 ^a
Leaves	1.66±0.83	0.54±0.16	3.36±0.91	1.85±1.37 ^b
Chicken excreta	3.58±3.00	2.34±1.80	0.46±0.27	2.12±2.30 ^b
Goat	2.78±1.22	0.34±0.13	2.48±1.80	1.86±1.61 ^b
Control (no pot)	1.88±0.98	0.80±0.08	1.24±0.11	0.63±0.77 ^a
Mean	1.88±1.69 ^q	0.81±1.01 ^p	1.24±1.48 ^p	

^{a, b} Different superscripts in the same column indicate significant differences ($P<0.05$)

^{p, q} Different superscripts in the same row indicate significant differences ($P<0.05$)

In the Table 6. showed that weight of cowpea root nodules planted on sandy land of block I (1.88 g/plant) was higher ($P<0.05$) compared with cowpea planted on block II (0.81 g/plants) and block III (1.24 g/plant). Nodule formation is the result of a symbiotic rhizobium bacteria and the legume roots. This symbiotic is a relationship between legumes root and rhizobium bacteria will establish if rhizobium strains capable to infect root legumes and then forming legume nodules. Nodule formation is the result of a symbiotic rhizobium bacteria and legume roots. Response of legumes to Rhizobium inoculation depends mainly on the amount of natural rhizobium can form root nodules on these plants (Reksohadiprojo, 1985).

CONCLUSION and SUGESSTION

Based on the results of this study it can be concluded that use of organic pots as planting mediums in the sandy land can improve the growth and productivity of cowpea. Organic pot made from chicken manure (COP) was better than another organic pots. Generally, the block II of sandy land in Agriculture Training, Research and Development Center Gadjah Mada University has better land fertility indicated by higher plant height, number of leaf and forages production.

It can be suggested that the organic pot made from chicken excreta and goats manure are potential plant medium which can be used for improving crop productivity and soil fertility especially in sandy land.

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STUDY ON PROTEIN EFFICIENCY RATIO, *IN VIVO* AND *IN VITRO* PROTEIN DIGESTIBILITY OF SELECTED PROTEIN FOOD

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ABSTRACT

This study determine the protein efficiency ratio (PER) and protein digestibility of selected protein foods. The selected protein sources were quail, muscovy duck, eel and tuna which was used as treatment group with casein and milk powder as reference formulations. The selected samples were analyzed for proximate analysis, protein quality, protein digestibility and amino acids content. The rats that were fed with quail meat protein (389.56 ± 36.38 g) indicated the highest mean of increased body weight (73.14 ± 18.55 g) while rats fed with milk protein (353.38 ± 33.04 g) showed the lowest mean increase in body weight (54.52 ± 14.59 g). From our study, One-way ANOVA test reported that only rats that were fed with milk and quail showed significant differences ($p < 0.05$) with mean body weight gain. However, both treatments showed no significant difference ($p > 0.05$) in total food intake. As for the PER value, milk revealed the highest PER value (2.61), followed by eel (2.26), quail (1.56), muscovy duck (1.47), casein (0.94) and tuna mackerel (0.67). As for the *in vivo* apparent protein digestibility test, casein showed the highest digestibility value (94.59) compared to tuna mackerel (94.23), while eel (89.47) showed the lowest. As for the *in vitro* digestibility analyses, Casein indicated the highest value for *in vitro* digestibility (110.25 ± 8.61). Casein (795.99 mg/g protein) and eel (787.41 mg/g protein) contained higher amino acids (essential amino acid and non-essential amino acids) compared to the other four samples. It was found that eel had the highest amino acid content amino acid score compared to the other samples. The major amino acids in all the selected samples were aspartate, glutamate and lysine. In conclusion, our study revealed that eel had the highest protein quality while tuna mackerel displayed the highest digestibility.

Key words: *protein efficiency ratio, in vivo, in vitro protein digestibility, protein food*

INTRODUCTION

Formulated diet plays an important role as the source of nutrients and protein is recognized as one of the most important dietary components (Goytortua-Bores *et al.* 2006). Protein quality can be classified as high and low quality protein. Low quality protein does not contain all essential amino acids required for use in protein synthesis whereas high quality proteins contain most of the essential amino acids that are needed for the functioning of human body systems. Plant proteins are often considered to be of lower quality than animal proteins because they have a lower content of certain essential amino acids. Nevertheless, protein from either source provides amino acids to humans as important materials for protein synthesis and as a source of energy. Generally, protein from animal foods for example dairy products, eggs, meats, fish and poultry are higher quality than protein from plant foods. During the past 50 years, growing concerns about food quality have led scientists to look for methods of measuring and defining the quality of proteins and the officially approved assay for protein nutritional quality is the rat based Protein Efficiency Ratio (PER) bioassay (AOAC, 1980). The rat PER assay is easy to conduct and has

been used extensively. The PER is the standard used by the U.S. food industry to evaluate the quality of protein in foods and also used to calculate the U.S. Recommended Daily Allowance (USRDA) for protein shown on food tables in the United States (Endres, 2001).

The Protein Efficiency Ratio (PER) is a measure of protein quality which is usually used to calculate protein quality by putting young animals on diets at 10% protein by weight with various test proteins and monitoring their growth. Osborne et al. (1971) observed that young rat fed with certain proteins gained little weight and ate little protein whereas those which were fed better quality protein gained more weight and consumed more protein. In an attempt to compensate for the difference in food intake, they calculated PER formulation based on gain in weight per gram of protein eaten.

In this study, the protein quality of formulated diet of selected high protein food sources such as Muscovy duck, eel, quail, tuna were evaluated using *in vivo* rat bioassay of Protein Efficiency Ratio (PER), as well as *in-vivo* digestibility with milk powder and casein as reference.

MATERIALS AND METHODS

Proximate analysis

Proximate analysis of samples is determined according to standard methods described by the Association of Official Analytical Chemists (AOAC 2000). A proximate analysis of samples was shown in Table 1. Nitrogen is determined using the micro Kjeldahl procedure. The oven method is used to determine moisture and ash content. Soxhlet method is used to determine fat content.

Rat Diet Preparation

All dried samples were grinded using a powder grinder with 5mm grinder plate. Diet formulation was done using procedures for PER as outlined by AOAC 1984 with casein and milk powder as reference protein. Other components included in the diet are ash mix (USP XVII), vitamin mix AOAC (CA 40055), corn starch, cellulose, sucrose and cooking palm oil (BURUH). Calculation of ingredient in diet formulation is based on the proximate analyses of the test protein. Each type of diet formulation (tuna mackerel, quail, eel, muscovy duck, milk and casein) were fed to 5 male and 5 female rats (Sprague-Dawley Strain) obtained from the animal laboratory at Universiti Kebangsaan Malaysia Bangi, Selangor.

Rat bioassay

Approximately 4 weeks were taken to conduct the protein quality study for each samples to determine PER and *in vivo* apparent protein digestibility. The 28 days rats are placed in individual cages and randomly assigned by treatment to individual cages. Prior to feeding the experimental diets, the rats are placed on an adaptation diet for 3 days period.

PER Assay

Food and water were supplied *ad libitum*. Body weight was recorded for 0 day and every two days for 28 days. For determination of feed intake, feces and the spilled feed were collected daily, dried in oven (100°C) for an hour, and then analyzed for moisture content before weighing.

The PER is calculated using the formula:

$$\text{PER} = \frac{\text{Increase in body weight (g)}}{\text{Weight of protein consumed (g)}}$$

In Vivo apparent protein digestibility (AD)

Fecal output and food consumption data were recorded daily for eight (day 10-18) of the 28 days. This is to determine the *in vivo* protein AD. It was calculated as follow:

$$\text{In vivo AD (\%)} = \frac{\text{N in diet (g)} - \text{N in feces (g)} \times 100}{\text{N in diet (g)}}$$

In Vitro Protein Digestibility

The *in vitro* protein digestibility of protein food sample was measured using the AOAC method (1984) with casein as a reference. The enzyme use is mixture of papain and bromeline (50:50) was prepared by add 10ml of water. Both solutions were kept in ice until ready for use.

Measurements of pH reduction by the Enzymes system

A reference solution containing 6.25 mg/ml protein casein was prepared. Sample weight (depending on protein content) was placed in test tube (25 x 95 mm) containing 10 ml distilled water. This solution was kept in ice for protein hydration for 1 hour. The protein solution was adjusted to pH 8 at 37 degree. Solution A and B were also adjusted to reach pH 8 at 37 degree, and then returned to the ice bath. Beginning with the casein standard solution, 1 ml of solution A was added to the reaction vessel. After exactly 10 minutes of addition of solution A, 1 ml of solution B was added and the tube transferred to the 55 degree water bath. At 19 minutes, the tube was transferred immediately to the reaction vessel and the pH was measured at exactly 20 min. The pH of standard casein should read 6.4. The same procedure was repeated for the protein test sample and the pH values (X) read exactly at 20 minutes.

$$\text{In vitro apparent digestibility} = 234.84 - 22.56 (X)$$

Amino Acid

For amino acid analysis, three methods have been employed in order to obtain all amino acid composition. Duplicate samples were hydrolyzed with 6NHCl hydrolysate, performic acid hydrolysate and tryptophan method.

Statistical Analyses

All statistical computations and mean \pm SD was performed by Statistical Packaged for Social Sciences (SPSS) version 21.0.

RESULTS AND DISCUSSION

Proximate analyses

Data on proximate of all 6 samples were shown in Table 4.1. The data indicated that the protein content of eel (98%) was highest followed by tuna mackerel (77.2%) and quail (73.7%). Otherwise, the lowest protein content of all samples was Casein (51%). Compared to the result from Babji and Letchumanan (1989), the percentage of protein in Casein was higher (83.56 ± 0.88). This may because of the casein that used in this study was old stock and affect the nutrient content. Therefore milk protein was used as a second control in this study.

A study from Jensen *et al.* (1990) shows that percent composition of average fish meal is 72% , which is lower than protein content in Tuna mackerel and eel from this study. This indicates that tuna mackerel and eel have higher protein content compared to standard protein content for fish. A study report by Ogunmola *et al.* (2013) shows that protein content in local chicken (50.95%) and Turkey (67.59%) are lower than protein content in muscovy duck (71.20%) and quail (73.7%). This result indicated that muscovy duck and quail are good source of protein, compared to chicken. Protein helps to maintain body’s structure, and have important function to build and repair tissues. Plus, it can fight infection, and transport oxygen from the lungs to the body tissue, by the action of chemical messenger through the chemical reaction in the body (Ogunmola *et al.* 2013). From this present study, protein from fish group (tuna mackerel and eel) has higher protein content than poultry group which indicate that fish is better source of protein compared to poultry.

Table 1. Proximate analysis of food samples

Protein food	% Protein	% Fat	% Moisture	% Ash
Milk	68.90 ± 6.22	3.00 ± 1.40	7.60 ± 0.00	2.90 ± 0.14
Casein	51.00 ± 38.47	1.90 ± 0.00	9.70 ± 0.70	2.60 ± 1.27
Tuna Mackerel	77.20 ± 1.13	5.00 ± 2.80	10.10 ± 0.70	5.80 ± 0.42
Muscovy duck	71.20 ± 0.00	12.40 ± 0.70	12.00 ± 0.00	4.20 ± 0.99
Eel	98.00 ± 0.00	2.50 ± 0.70	10.50 ± 0.70	4.00 ± 0.23
Quail	73.70 ± 0.18	15.30 ± 0.10	6.10 ± 1.41	4.20 ± 0.40

Rat bioassay

Through the study, all rats survived and gain positive body weight. Based on table 4.2, the rats fed with quail diet (73.14 ± 18.55g) shown highest body weight gain and followed by eel diet group (68.43 ± 13.27g). The lowest mean body weight (54.52 ± 14.59g) was recorded in the rat fed of milk. The other three diet showed no significant difference (p>0.05) for mean body weight.

Table 2. Body weight gain of rats fed with formulated diet

Treatment	N	Total Food Intake (g)	Increased in Body Weight (g)
Milk	10	353.38 ± 33.04 ^a	54.52 ± 14.59 ^a
Casein	10	366.62 ± 41.90 ^a	65.19 ± 18.34 ^{ab}
Tuna Mackerel	10	388.08 ± 30.11 ^a	68.43 ± 13.27 ^{ab}
Muscovy duck	10	388.46 ± 34.02 ^a	57.21 ± 26.51 ^{ab}
Eel	10	381.47 ± 40.03 ^a	64.34 ± 8.79 ^{ab}
Quail	10	389.56 ± 36.38 ^a	73.14 ± 18.55 ^b

Protein Efficiency Ratio

PER data for rats fed with treatment diets of casein, milk, eel, tuna mackerel, muscovy duck and quail were shown in table 4.3. The results of PER values obtained from the study were 2.61, 2.26, 1.56, 1.47, 0.94 and 0.67 for milk, eel, quail,

muscovy duck, casein and tuna mackerel respectively. The milk diets showed the highest PER value while tuna mackerel displayed the lowest PER value as being shown by graph 4.1. Generally, the PER value of food samples were compared to a standard value of casein protein which is 2.7. Any value that exceeds 2.7 is considered to be an excellent protein source (Hoffman *et al.* 2004). In this study, the PER value of milk (2.61) is approximately near the standard value. The PER value for milk protein was higher than casein indicated the higher quality of the protein. Eel formulated diet has higher PER value (2.26) compared to caseinated diet (0.94). The PER of casein in this study was lower when compared to the standard PER value of casein. This might be due to the lactic acid casein which had a reduced content of sulfur-containing amino acid, methionine. Besides that, the PER of caseinated diet should be higher than eel. Babji *et al.* (2007) reported that PER value of majority fish-based diet: canned sardine (2.48); anchovy (2.46); marckerel (2.34) were lower than casein (3.14).

Table 3. PER values of Milk, Casein, Eel, Tuna Mackerel, Muscovy duck and Quail.

Source of Protein	Weight Gain Rats (g/rat) (Mean ± S.D.)	Total Feed Intake (g/rat/28 days) (g ± S.D)	% Protein in Feed (N x 6.25)	Protein Consumed (g/rat/28 days) (g ± S.D.)	PER	Adj PER
Milk	54.52 ± 14.59 ^a	353.38 ± 33.04 ^a	5.88	20.85 ± 1.95 ^a	2.61	6.94
casein	65.19 ± 18.34 ^{ab}	366.62 ± 41.90 ^a	19.19	69.66 ± 7.96 ^a	0.94	-
Eel	68.43 ± 13.27 ^{ab}	388.08 ± 30.11 ^a	7.75	30.27 ± 2.35 ^a	2.26	6.01
Tuna Mackerel	57.21 ± 26.51 ^{ab}	388.46 ± 34.02 ^a	22.06	85.46 ± 7.49 ^a	0.67	1.78
Muscovy duck	64.34 ± 8.79 ^{ab}	381.47 ± 40.03 ^a	11.44	43.64 ± 4.58 ^a	1.47	3.91
Quail	73.14 ± 18.55 ^b	389.56 ± 36.38 ^a	12.13	46.75 ± 4.37 ^a	1.56	4.15

As for tuna mackerel, the PER value in this study was 0.67. The result obtained showed the difference in value might be due to different practices of method used during diet preparation. The diet preparation also plays an important role in determining the PER value. The tuna mackerel had undergone heat treatment for drying purpose prior diet preparation. Thus, the protein quality could have been denatured by exposure to high heating temperature, thus affecting protein quality (Babji *et al.* 2007). Protein consumed in tuna mackerel rat diet was the highest while milk was the lowest protein consumed by rats but no significant difference showed by each diet ($p > 0.05$). As stated by Mercer *et al.* (1981), the rats in low protein diet increase their food intake per 100g weight to 11.53g. As protein concentration increases, food intake decreases to about 7.4g per 100g weight. This relationship indicates that there is some factor which limits the rat’s ability to increase food intake to obtain more protein.

Apparent protein digestibility (APD)

Table 4. showed *in vivo* apparent protein digestibility (APD) of casein, milk, eel, muscovy duck, tuna mackerel and quail. Casein showed the highest percentage

of digestibility (94.59%) followed by tuna mackerel (94.23%), muscovy duck (92.59%), quail (89.66%), eel (89.47) and milk displayed the lowest percentage of digestibility (84.62%). Study from Albreksten *et al.* (2006) showed that fish meal has high quality protein produces higher AD value compared to a vegetable meal which has low quality protein.

In the *in vivo* technique, nitrogen value was used to calculate the apparent digestibility. Furthermore, in the digestive system of rat, there several kind of enzymes to hydrolyze different type of food component such as carbohydrate, fat, and protein. Thus, the nitrogen value may come from other component rather than protein. Therefore, it contributes with the digestibility of the food sample. Besides, there are some factors such as control of temperature and pH might affect the result. Which, if the pH *in vitro* system is lower or above the optimum pH, the digestibility might not occur due to the inactive enzyme. Similarly, if the temperature is higher than 37⁰C, it may cause the enzyme to inactive. Based on table 4. casein showed high in *in vitro* and *in vivo* digestibility. Diet containing casein as the source of protein was significantly more digestible than those prepared with the eel mixture. The greater extent of digestibility of the casein diet resulted in a higher retention of dietary nitrogen as indicated by the lower fecal nitrogen output of the rats consuming the diet (Mensa-Wilmot *et al.* 2001).

Table 4. *In vivo* and *In vitro* protein digestibility of different protein source

Source of protein	% <i>In vitro</i> Digestibility	% <i>In vivo</i> Apparent Digestibility (AD)
Casein	110.25 ± 8.61	94.59
Milk	81.32 ± 2.08	84.62
Eel	82.56 ± 0.00	89.47
Quail	80.76 ± 0.32	89.66
Muscovy duck	NIL	92.59
Tuna mackerel	NIL	94.23

As comparison, the values obtained from *in vitro* technique were lower compared with the value determined by the *in vivo* rat assay. Similar results were reported by other investigators (Babji et al. 1980; Bodwell et al. 1980). This because, the availability of enzymes present in the *in vivo* technique was different from *in vitro* technique. The enzymes used in this study were from plant-based enzyme which are papain + bromelain, and flavourzyme. Thus, the mechanism of enzymes on protein digestibility *in vitro* might be different compared to *in vivo* apparent digestibility.

Amino acid analysis

From the table 6. it shows that casein and eel had higher total amino acid compared to the other four samples which is quail, muscovy duck, tuna mackerel and milk powder. The major amino acids in quail, eel, tuna mackerel and muscovy duck were aspartate (ranging from 59.61-80.86mg), glutamate (ranging from 91.98-122.82mg) and lysine (ranging from 51.06 to 69.17mg in 1g of protein). Mulia (2008) and CFCD (2002) reported that swamp eel are abundant in aspartate, glutamate and lysine which correspond to the result of this work.

Glycine, arginine and alanine in the four analyzed food protein is higher ($p < 0.05$) than in milk and casein. Glycine, which is one of the major components of human skin collagen, together with other amino acids such as alanine form a polypeptide that have regenerative effects (Heimann, 1982). Whereas arginine plays an important role in ammonia removal, immune function, and hormone release. It is also the precursor for biological synthesis of nitric oxide which plays important roles in neurotransmission, blood clotting, and maintenance of blood pressure (Sarma *et al.* 2013).

High quality proteins are readily digestible and contain the dietary essential amino acids (EAA) in quantities that correspond to human requirements (WHO 2007). By comparing the freshwater (eel) and saltwater fish (tuna mackerel), it shows that eel has higher protein quality compared to tuna mackerel because it contain higher ($p < 0.05$) dietary EAA threonine, methionine, lysine and phenylalanine.

Table 4.6 Amino acid compositions of quail, eel, tuna mackerel, Muscovy duck, casein and milk powder (m/g protein) in dry weight basis.

Amino acid	Sample					
	Quail	Eel	Tuna mackerel	Muscovy duck	Casein	Milk powder
Hydroxyproline	3.19 ^{ab} ±0.43	14.44 ^c ±2.67	2.72 ^{ab} ±0.14	10.08 ^{bc} ±7.29	0.00 ^a	0.00 ^a
Aspartate	72.67 ^d ±4.49	80.86 ^e ±3.23	65.19 ^c ±2.23	59.61 ^{bc} ±0.90	55.69 ^b ±1.96	44.94 ^a ±2.45
Serine	27.42 ^c ±1.64	30.68 ^c ±1.30	21.98 ^b ±1.03	23.60 ^b ±0.16	41.04 ^d ±1.77	15.75 ^a ±1.60
Glutamate	110.78 ^c ±6.14	122.82 ^d ±4.24	91.98 ^b ±3.59	96.42 ^b ±0.04	166.13 ^c ±6.76	65.17 ^a ±3.96
Glycine	30.43 ^b ±0.93	55.74 ^c ±6.66	31.28 ^b ±0.69	36.14 ^b ±10.43	11.07 ^a ±2.73	6.68 ^a ±0.62
Histidine*	20.73 ^{bc} ±1.18	18.18 ^{bc} ±0.66	41.48 ^d ±2.22	15.77 ^{ab} ±0.24	27.99 ^c ±9.40	6.95 ^a ±0.56
Arginine	46.78 ^d ±2.06	53.63 ^e ±1.99	40.01 ^c ±2.19	42.70 ^{cd} ±1.80	26.16 ^b ±1.27	8.81 ^a ±0.62
Threonine*	31.50 ^{de} ±2.30	34.64 ^e ±1.26	27.34 ^{bc} ±1.03	26.55 ^b ±0.71	30.56 ^{cd} ±1.59	20.06 ^a ±1.84
Alanine	35.05 ^b ±1.70	45.38 ^c ±2.98	34.06 ^b ±0.81	35.44 ^b ±2.75	21.17 ^a ±0.14	17.93 ^a ±0.65
Proline	25.53 ^{ab} ±0.66	39.32 ^c ±4.63	25.99 ^{ab} ±0.91	30.08 ^{bc} ±6.68	79.61 ^d ±4.86	18.07 ^a ±1.58
Tyrosine	21.17 ^b ±1.27	26.05 ^c ±0.23	22.33 ^b ±0.79	23.34 ^{bc} ±1.15	45.26 ^d ±2.28	15.59 ^a ±0.61
Valine*	35.91 ^c ±2.01	37.33 ^c ±1.17	36.92 ^c ±1.40	31.26 ^b ±0.85	51.45 ^d ±3.02	21.03 ^a ±1.49
Methionine*	21.05 ^{abc} ±1.33	25.18 ^{bc} ±3.08	11.19 ^a ±11.55	27.00 ^c ±0.59	23.06 ^{abc} ±2.71	12.13 ^{ab} ±2.84
Lysine*	66.53 ^d ±5.15	69.17 ^d ±1.45	53.38 ^{bc} ±2.05	51.06 ^b ±2.50	58.86 ^c ±2.76	37.76 ^a ±1.29
Isoleucine*	35.62 ^c ±2.33	36.62 ^{cd} ±0.86	32.68 ^{bc} ±0.98	29.54 ^b ±1.32	39.93 ^{cd} ±2.24	22.29 ^a ±1.50
Leucine*	55.88 ^c ±3.25	56.07 ^c ±0.83	51.25 ^{bc} ±1.36	48.25 ^{ab} ±1.77	69.25 ^d ±3.93	43.85 ^a ±2.31
Phenylalanine*	29.11 ^{bc} ±1.79	31.37 ^c ±1.10	27.71 ^b ±0.59	25.93 ^b ±0.45	40.41 ^d ±2.26	13.45 ^a ±0.85
Cysteine	6.50 ^b ±0.43	5.92 ^b ±0.79	4.85 ^{ab} ±0.34	6.80 ^{bc} ±0.23	2.58 ^a ±0.26	9.16 ^c ±2.18
Tryptophan*	3.93 ^b ±0.47	4.01 ^b ±0.81	3.77 ^b ±1.06	4.73 ^b ±0.60	5.77 ^b ±1.78	0.78 ^a ±0.11
Total essential amino acid (EAA)	300.26	312.57	285.72	260.09	347.28	178.30
Total non-essential amino acid (NEAA)	379.52	474.84	340.39	364.21	448.71	202.10
Protein content (%)	73.70	98.0	77.20	71.20	51.0	68.90

* Essential amino acid

(a-e) Means in the same row with unlike superscript differ significantly ($p < 0.05$)

In clinical nutrition, threonine is used for treating various nervous system disorders (Hyland, 2007), methionine is used for treating liver disorders, improving wound healing, and treating depression, alcoholism, allergies, asthma, copper poisoning, radiation side effects, schizophrenia, drug withdrawal and Parkinson’s disease (Mischoulon & Fava 2007) and lysine is an EAA which is extensively required for optimal growth and its deficiency leads to immunodeficiency (Chen *et al.* 2003). However, amount of EAA in eel is comparable with quail ($p>0.05$). Therefore, eel and quail can be recommended as alternate sources of protein in this country instead of overconsumption of chicken, beef and other food fishes.

CONCLUSIONS

Among the 4 treatment group, eel showed the highest value of Protein Efficiency Ratio, but relatively lower than milk, reflecting lower protein quality from the standard reference. While for *in vivo* and *in vitro* digestibility, casein displayed the highest value, and milk displayed the lowest value for *in vivo* digestibility. In the rat bioassay, all rats gained positive body weight and rats fed with quail diet had the highest mean body weight compared to others. It was found that eel had the highest amino acid content and the amino acid score compared to quail, muscovy duck and tuna mackerel.

ACKNOWLEDGEMENT

The authors would like to thanks all the officers and staffs from Faculty Science and Technology, Universiti Kebangsaan Malaysia which have been involved throughout the study for all assistance and helps.

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FERMENTATION QUALITY OF LAB-SCALE CASSAVA LEAVES (*MANIHOT ESCULENTA* CRANTZ.) SILAGE WITH GLYCEROL AND/OR CHESNUT TANNIN ADDITIVES

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ABSTRACT

Cassava leaves have a big potential to be valuable protein supplement for ruminant and a better way to maintain its nutritious composition is by ensilage. This present study aimed to evaluate the quality fermentation of cassava leaves silage with glycerol and chestnut tannin extract additives. Cassava leaves was ensiled using four different types of additive on the laboratory-scale silo. They were control (without additive, S0), 3% DM glycerol (SG), 3% DM chestnut tannin (ST), and mix of 3% DM glycerol and 3% DM chestnut tannin (SGT). Cassava leaves ensiled in the three different batches during 4 weeks period. Silage quality were evaluated on week 0 and 4. Data were analyzed using general linear model (GLM) procedure then significant means were separated using Duncan multiple range F-test. Every single cassava leaves silages on week 4 in this study showed well-preserved quality. It depends on high proportion of lactic acid (73.5% from total acid), not detected butyric acid and low concentration of N-NH₃ although the pH value slightly higher. We consider that the fermentation characteristics of cassava leaves ensilages with glycerol and/or chestnut tannin additives have values that in a range of a good silage properties.

Keywords: *cassava leaves, chestnut tannin, glycerol, silage*

INTRODUCTION

Cassava leaves (*Manihot esculenta* Crantz) productivity has a big potential to be valuable protein supplement for ruminant. However, due to the abundant availability when crop, cassava leaves requires a storage technology to maintain its nutritious composition. The one of better way is by ensilage. The mechanism principle of ensilage is to ferment materials in anaerobic condition so that lactic acid bacteria (LAB) will growth and produce acidic environment at pH 4 or lower to suppress and even stop the activity of proteolytic enzymes and harmful microorganisms (McDonald *et al.* 1991; Napasirth *et al.* 2015). However, the ensiling process of cassava leaves required additives because its low content of WSC and higher protein. Additives used in this present study were glycerol and chestnut tannin extract.

The glycerol usage as a silage additive associated with high glycerol production cause the development of the biodiesel industry in order to meet the increasing global demand for renewable and sustainable energy. Generally, this industry produce crude glycerol 10-20% of the total volume of biodiesel produced (Quispe *et al.* 2013). Glycerol has been recognized safely used for animal feed (FDA

2006). Many countries in the European Union, USA, Australia and even Thailand have begun to adapt glycerol as a feed substitution for energy source. Other studies have been published and mentioned that glycerol supplementation in ruminants does not cause adverse effect on rumen fermentation characteristics (Lee *et al.* 2011), cattle performance and carcass characteristics (Ramos & Kerley 2012) as well as the production and composition milk (Donkin *et al.* 2009). Glycerol supplementation is generally used by mixed in the ration and known that glycerol has never been used as silage additives. We tried to explore the glycerol usage for ruminant supplementation in another form as silage additives on cassava leaves.

Chestnut tannin extract is one type of hydrolysable tannins from temperate plant and had been widely used either as an additive in ration and in silage. Tannins able to complexes with protein and resistant on proteolytic activity either in silos (Salawu *et al.* 1999) as well as in the rumen (Messman *et al.* 1996). Tobacco *et al.* (2006) also used as an additive hydrolysable tannins on alfalfa silage and proven to reduce protein degradation during storage. The objective of this study was to evaluate the fermentation quality of cassava leaves silage with glycerol and chestnut tannin extract addition.

MATERIALS AND METHODS

Silage Preparation

Cassava leaves (*Manihot esculenta* Crantz) obtained from the Selangor Wholesale Market, Malaysia. Cassava leaves silage were made with separated the stems and leaves then chopped manually into a length of 4-5 cm, homogenized, divided into four parts, then each was sprayed with or without silage additives using a mini hand-sprayer. The additives were control (without additives, S0), 3% DM glycerol (SG), 3% DM chestnut tannin (ST), and mix of 3% DM glycerol and 3% DM chestnut tannin (SGT). Each additives were dissolved first in distilled water prior to 20 ml kg⁻¹ of fresh leaves. Cassava leaves that have been mixed with or without additives were filled into mini polyethylene silos with a capacity of 450 cc and stored in a room with a temperature of about 28-31°C until 4 weeks. This experimental design was arranged in randomized complete block design with triple replicates per treatment.

Chemical Analysis

Fermentation products were determined from silage fluid extraction according to method of Ridla and Uchida (1998) with a slight modification. Wet silage (25 g) was blended with 100 mL distilled water then filtered with Whatman filter paper (No. 1). The pH was measured immediately with a glass electrode pH meter (Mettler Toledo), and ammonia-N concentration was determined by phenate method as modification from Parsons *et al.* (1984) using spectrophotometer. The organic acid contents were measured by gas chromatography (GC, Agilent 69890N Series Gas Chromatography System from Agilent Technologies, USA that equipped with a flame ionization detector and packed column 5% Thermon-3000) methods as described by Cottyn and Bouque (1968) which modified by Minato and Kudo (pers. Comn). The samples of cassava leaves silages were oven-dried at 60°C for 48 h then ground to pass through a 1 mm screen. Dry matter (DM), ether extract (EE) and crude ash were analyzed according to AOAC (2005). The organic matter (OM) was calculated as the weight loss upon ashing. Crude protein (CP) was analyzed according to Dumas combustion method (Gerhardt Dumatherm, Malaysia). The neutral detergent fiber

(NDF) and acid detergent fiber (ADF) were analyzed by the methods of Van Soest *et al.* (1991). Nutrient composition of fresh matter cassava leaves were shown on Table 1.

Table1. Nutrient composition of fresh cassava leaves

Nutrient composition	(% DM)
Dry matter	29.30
Organic matter	92.59
Crude protein	33.97
Ether extract	5.30
NDF	46.89
ADF	21.99

Statistical Analysis

Cassava leaves was ensiled using four different types of additive on the laboratory-scale silo. They were control (without additive, S0), 3% DM glycerol (SG), 3% DM chesnut tannin (ST), and mix of 3% DM glycerol and 3% DM chesnut tannin (SGT). Data on the chemical composition and fermentation products of cassava leaves silage were analyzed using general linear model (GLM) procedure of SPSS 16.0 then the significant means were separated using Duncan multiple range F-test ($P < 0.05$).

RESULTS AND DISCUSSION

Fermentation Characteristics of Cassava Leaves Silage

Fermentation characteristics of cassava leaves silage (Table 2) with or without additives showed well-preserved quality. Some parameters on the fermentation characteristics may become a main determinant for the indicator of silage quality includes pH value, the percentage of organic acids produced and proteolytic activity occurred. However, the main indicator in this present study was shown by a high proportion of lactic acid. Lactic acid proportion of cassava leaves silage showed not significant ($P > 0.05$) between treatments that approximately 73.5% ($37 \text{ g kg}^{-1} \text{ DM}$) of total acids on average. The value was slightly higher than a proportion range according to Kung Jr (2010) and Ward and de Ondarza (2008), which is about 65-70% of the total acid or about 3-6% DM (Seglar 2003). Lactic acid concentration in cassava leaves silage with or without molasses showed a lower value was 9.5-9.9 $\text{g kg}^{-1} \text{ DM}$ (Man & Wiktorsson 2002).

Main activity of lactic acid bacteria (LAB) is produce lactic acid that decreasing pH value in the end product, tolerant to acid itself in order to improve the silage quality through rapidly growth and dominate other microorganisms but not metabolize organic acids or proteins (McDonald *et al.* 1991). Well-preserved quality of cassava leaves silage was also supported with no detection of butyric acid and low concentration of acetic and propionic acid. Good quality of silage indicated by the content of acetic, propionic and butyric acid in the range of $< 3\%$, $< 0.5\%$ and $< 0.1\%$, respectively (Ward & de Ondarza 2008). Acetic and propionic acid produced during the fermentation generally to maintain aerobic phase but if the proportion is more than 3%, it indicated inefficiency of heterofermentatif fermentation (Seglar 2003). Contrary, butyric acid indicated a nutritional damage due to secondary fermentation of clostridial activity (McDonald *et al.* 1991). The secondary fermentation produced

nitrogenous end product and not palatable such as amines and amides (Seglar 2003). It may reduce silage nutrient significantly.

Table 2. Fermentation characteristics of cassava leaves silage with and without additives

Fermentation characteristics	Treatment				SEM	P-value
	S0	SG	ST	SGT		
Lactic acid(g kg ⁻¹ DM)	41.01	37.24	31.60	38.39	6.417	0.960
C ₂ (g kg ⁻¹ DM)	10.36	15.80	5.36	6.45	1.692	0.224
C ₃ (g kg ⁻¹ DM)	0.32	0.24	0.54	0.22	0.031	0.178
C ₄ (g kg ⁻¹ DM)	nd	Nd	nd	Nd	-	-
pH value	4.86 ^b	4.70 ^a	5.05 ^c	4.79 ^{ab}	0.019	<0.001
N-NH ₃ (g kg ⁻¹ TN)	15.01 ^a	15.06 ^a	14.42 ^a	20.81 ^b	0.275	<0.001

Note: Means in the same row with different superscript differ significantly ($P < 0.05$), C₂: acetic acid, C₃: propionic acid, C₄: butyric acid, N-NH₃: nitrogen ammonia, TN: total nitrogen, DM: dry matter, S0: without additives, SG: with glycerol 3% DM, ST: with chesnut tannin extract 3% DM, SGT: with glycerol 3% DM and chesnut tannin extract 3% DM, SEM: standart error mean.

Characteristics of the well-preserved cassava leaves silage were also shown by N-NH₃ concentrations. N-NH₃ concentrations influenced by level of additives treatment and showed a significantly highest value of SGT. However, that value was not enough to prove the existence of nutrients damage in the silage because it was still lower than the concentration standard on good quality of silage. N-NH₃ concentration should be <3% for grass silage and <8% for alfalfa or legume silage to indicate well-preserved quality (McDonald *et al.* 1991). The production of N-NH₃ showed the breakdown of amino acids (deamination) which occur due to the proteolysis. The proteolysis activity proved to be reduced with using low level (4% DM) of hydrolysable-chesnut tannins as an additive in alfalfa silage (Tabacco *et al.* 2006) because the tannins have ability to protect protein through a stable complexes at pH 3.5-7.5 (Barry & McNabb 1999).

Organic acids produced inside cassava leaves silage reflected to its pH value but showed a negative correlation. The pH value of silage in this present study showed a relatively higher value than the standard of good quality silage. The final pH value at S0, SG, ST and SGT were 4.86, 4.70, 5.05 and 4.79, respectively. The pH value on final product of silage proved well-preserved quality when it can achieve low pH about 3.8-4.0 to 4.3-4.5 for silage maize and legume silage in a period of about 6 weeks (Ward & de Ondarza 2008). The declining of pH value may be able to be continued when the shelf life of silage be extended, such as the results of Man and Wiktorsson (2002) which showed that the pH value of cassava leaves silage on 2 months (4.38) continued to decline until 4 months (4.21).

The final pH value of cassava leaves silage were relatively high (> 4.5) were allegedly caused by buffering capacity so it can not decline maximumly. Buffering capacity on silage generally occurred when materials have a high CP and alkali content. Alkali content represented by mineral content. Silage of leguminose particularly alfalfa showed a high value of buffering capacity due to contained a high calcium content so that some lactic acid produced will be neutralized (Toth *et al.* 1956). It was also occurred in cassava leaves silage due to mineral content especially calcium on cassava leaves about 14.5 g kg⁻¹ which was slightly lower than calcium in alfalfa (15.0 g kg⁻¹) (Ravindran 1993) beside its high CP content. The value of

buffering capacity was not determined in this study but it may be detected from lower activity of Clostridial fermentation due to lower percentage of N-NH₃ produced and a slightly changed of CP content from fresh matter.

Nutrient Composition of Cassava Leaves Silage

Nutrient composition of cassava leaves silage included dry matter (DM), organic matter (OM), crude protein (CP), ether extract (EE), neutral detergent fiber (NDF) and acid detergent fiber (ADF) were shown on Table 3. Generally, nutrient composition of cassava leaves silage showed not significant value between treatments except DM content. Dry matter (DM) content of cassava leaves silage showed a significantly increase ($P < 0.001$) between treatments starting from S0, SG as same as SGT then ST. It is caused by the form of each additives. However, DM content of each silages were included in a range of DM content on good quality silage. It were between 250-350g kg⁻¹ (McDonald *et al.* 1991). The declining of OM and CP content of cassava leaves silage as much as 0.15-0.40% DM and 0.6-1.0% DM from fresh matter, respectively. They were associated with a lower nutrient loss in the silages due to the activity of epiphytic microorganisms including LAB contained in cassava leaves. CP content of cassava leaves silage in this present study were included in the range of 29.3-38.6% DM (Yeoh and Chew 1976) depending on the variety and age of plants. Contrary, the increasing of EE content from fresh matter related to the increasing of silage DM content towards increasing of silage EE proportion.

Table 3. Nutrient composition of cassava leaves silage with and without additives

Nutrient composition (% DM)	Treatment				SEM	P-value
	S0	SG	ST	SGT		
Dry matter	30.28 ^a	31.49 ^b	32.65 ^c	31.70 ^b	0.149	<0.001
Organic matter	92.38	92.44	92.20	92.38	0.038	0.125
Crude protein	33.38	33.32	32.89	32.96	0.130	0.442
Ether extract	5.76	5.64	5.62	5.55	0.102	0.908
NDF	38.94	37.10	39.73	38.94	0.358	0.197
ADF	21.65	22.08	20.95	21.02	0.247	0.345

Note: Means in the same row with different superscript differ significantly ($P < 0.05$)

NDF: neutral detergent fiber, ADF: acid detergent fiber, DM: dry matter, S0: without additives, SG: with glycerol 3% DM, ST: with chesnut tannin extract 3% DM, SGT: with glycerol 3% DM and chesnut tannin extract 3% DM, SEM: standart error mean.

NDF content of silage decreased in a range between 7-9% from fresh matter while the declining ADF content was lower than 1%. The ensiled process of cassava leaves without additives capable of reducing NDF content as much as 8% from fresh matter but not occurred in ADF content (Man & Wiktorsson 2002), while Morrison (1979) reported that 10-20% of the hemicelluloses were solubilized during the 150 day ensiling period. The reduction of NDF content during ensiling associated with fiber components relatively hemicelluloses were easy hydrolyzed by acid (Jatkauskas & Vrotniakiene 2006) so that its carbon chain can be used as a energy source for LAB growth. In addition, McDonald *et al.* (1991) reported that more than half of hemicelluloses contained in the materials can be degraded either by enzymes

hemicellulase origin plants, bacteria hemicellulases or through hydrolysis by organic acids produced during fermentation. The fiber reduction as NDF-ADF content is expected to increase ruminant digestibility.

CONCLUSION

The ensiling process of cassava leaves with or without glycerol and/or chestnut tannin extract additives showed well-preserved quality for 4 weeks fermentation. It was indicated by a high lactic acid produced, not detected butyric acid, low concentration of N-NH₃ and low percentage of nutrient loss.

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THE EFFECT OF FERMENTATION DURATION OF AREN (*Arenga pinnata* Merr) STEM WASTE ON CHEMICAL COMPOSITION AND *IN VITRO* DIGESTIBILITY

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ABSTRACT

This research was conducted to determine the effect of fermentation duration of aren stem waste on chemical composition and *in vitro* digestibility. In the beginning of fermentation, 0.3 g biostarter and 1.2 g urea was added to 300 g aren stem waste. Fermentation was conducted anaerobically at room temperature for 10, 20, 30, 40 d with three replicates for each treatment. The samples of fermented aren stem waste were analyzed physical quality (texture, odor, color, growth of fungi and pH), chemical composition, and *in vitro* digestibility. The data obtained were analyzed variance following one-way design, and continued by Duncan's new multiple range tests for any differences between treatments. Results showed that fermentation gave soft texture, more acidic odor, and dark brown color than fresh aren stem waste. The fermentation decreased pH of aren stem waste up to less than 4.2 ($P < 0.01$) at 20, 30, and 40 d (4.2, 3.82 and 4.17). The chemical composition of fermented aren stem waste increased after a longer fermentation time. While crude fiber content decreased when a longer fermentation time with the lowest value 31.57% ($P < 0.01$) at 40 d of fermentation time. After 30 d of fermentation, dry matter, organic matter, and crude fiber digestibility increased 12.84%, 17.18%, and 20.91% respectively compared to 10 d of fermentation time ($P < 0.01$). It could be concluded that 30 d was the most effective of fermentation duration of aren stem waste which increased chemical composition and *in vitro* digestibility.

Key Words: *Aren stem waste, Fermentation duration, Chemical composition, In vitro digestibility*

INTRODUCTION

Arenga pinnata is a natural forest species that originates from the Palmae family, it is known as a fast growing palm that is able to reach maturity within 10 years (Ishaka *et al.*, 2013). It has also an economically important resource that is exploited for multiple purposes. The most important use is tapping of sap from the inflorescence for sugar, wine or vinegar and extraction of starch from the stem for sago (Pongsattayapipat and Barfod, 20015). This plant has not been cultivated in a commercial scale, whereas most farmers either utilize it directly from the forest or those which are grown in their backyard.

All parts of the plant can be utilized and converted into a variety of products such as sugar sap, starch, *kolang-kaling* (endosperm), or fibers (Soeseno, 2000). The usage of this plant is not limited only for consumable products but in terms of conservation it helps to prevent erosion, improve soil macro conditions, improve soil porosity, and trapping rainwater (Devi *et al.*, 2014).

Sugar palm produces starch from its trunk, the starch can be used to make biodegradable polymer. sugar palm starch has been used traditionally by the locals to make flour, noodles, 'mee hoon' and animal feeds until now (Ishaka *et al.*, 2013). According to Singhal *et al.* (2008) from the sago starch extraction process from sago stem, it will produce waste named sago pith waste (SPW), which contains high fibres and starch. These wastes not only serves as animal feed but also to solve

the environment problem of agricultural waste accumulation. Manufacturers revealed that approximately one tonne of SPW is formed from every tonne of sago starch produced (dry weight basis). However, SPW has not been utilised by manufacturers but is usually combined with the waste water and discharged into the river. Meaning that about the same quantity of SPW was dumped into the river. Lai *et al* (2013) reported that moisture content analysis showed that the fresh SPW contains 82%, by weight of moisture. Starch contents of the SPW range between 58 and 67% (dry weight basis). In view of the fact that SPW contains high percentage of starch, it is believed that the waste can be used as an animal feed. Fermentation of SPW is conducted to preserve and increase its quality. Therefore the purpose of the experiment was to obtain the effective time an aren stem waste took to ferment which resulted the best fermented aren stem waste quality.

MATERIAL AND METHODS

Experimental Design

Experimental design was arranged in a one way design, with the main factors being fermentation duration (10, 20, 30, 40 d). Fermentation experiments were separately conducted for each treatment with three replicates. Air-dried aren stem waste was utilized as substrate for solid state fermentation.

Fermentation of aren stem waste

Aren stem waste was dried under the sun to achieve the moisture content less than 40%. Mixed 300 g air-dried aren stem waste with 0.1% of biostarter and 0.4% of urea (w/w). Then all treatments were added with distilled water to achieve the final water content of fermentation by 45%.

The final water content of fermentation was 45% for all treatments by adding distilled water. Fermentation was conducted anaerobically at room temperature for 10, 20, 30, 40 d with three replicates for each treatment. At the end of the incubation period, physical quality and pH was determined in each fermentation duration. Then, sample was collected, dried at 55°C for 72 h, ground through a 1-mm screen Wiley mill and analyzed for chemical composition as well as *in vitro* digestibility.

Measurement of Fermentation Parameters

Physical quality. Physical quality of the samples was analyzed including texture, odor, color, growth of fungi.

pH of fermentation. pH of fermented aren stem waste was immediately recorded using a pH meter at the end of each the fermentation duration.

Chemical composition. Fermented aren stem waste was analyzed for chemical composition including dry matter (DM), organic matter (OM), crude fiber (CF), crude protein (CP), ether extract (EE), and nitrogen free extract (NFE) following to AOAC methods (2005). These analysis were conducted for original and fermented sample of aren stem waste to know the effect of fermentation on chemical composition and *in vitro* digestibility.

***In vitro* digestibility.** *In vitro* digestibility of the samples was determined including dry matter digestibility (IVDMD), organic matter digestibility (IVOMD), and crude fiber digestibility (IVCFD), and used was similar to that described by Tilley and Terry (1963), but only in the first step. Short-term *in vitro* incubations were carried out with rumen liquor from a fistulated Ongole Cross Breed, which was withdrawn before the morning feeding and was squeezed through four layers of

surgical gauze into an Erlenmeyer flask. The rumen fluid was then mixed with buffer (McDougall) in ratio 1:4 as a medium. Whereas the temperature and pH of the mixture were maintained at 39C and 6.9, respectively. While anaerobic conditions were maintained during fermentation by initial flushing of the tubes with CO₂. Approximately 0.25 g of ground air-dried substrate (fermented aren stem waste from different fermentation duration) of known chemical composition previously was introduced in a 50 ml rubber cap glass vessel. The volume of medium was added up to 25 ml and the vessel flushed with CO₂, capped with a one-way valve and incubated in water-bath for 48 h at 39C. All vessels were swirled at 0 h and third daily thereafter. Blanks were run in triplicate throughout the incubation and digestion process. After 48 h, the contents of the vessel filtered through crussible layered by glass wool and washed with boiled water. The residues were analyzed for DM, OM, and CF to determine IVDMD, IVOMD, and IVCFD, respectively. *In vitro* digestibility was calculated as original weight of sample (before incubation) minus dried-residue weight (after incubation) divided by original sample weight. To derive *in vitro* digestibility percentage, these values were then multiplied by 100.

Statistical Analysis

Statistical analysis of all data was performed by one way design with SPSS 10 software to determine the effects of fermentation duration. All statements of significance were based on a probability of less than 0.05. The differences of mean value were analyzed by Duncan’s new multiple range test (Rosner, 1990).

RESULT AND DISCUSSION

Fermentation characteristics and its chemical composition

The chemical composition including DW, OM, CP, CF, EE, and NFE of aren stem waste was 16.94%, 97.81%, 1.62%, 38.83%, 0.54%, and 56.82%, respectively. Fermented aren stem waste had low pH value (i.e., 3.82-4.77), longer fermentation duration got lower pH value, as well as resulted good texture, acid odour, dark colour, and no-fungi growth.

The length of fermentation affected chemical composition of fermented aren stem waste. The longer fermentation time up to 40 d increased its nutrient quality, as presented in Table 1, it increased DM, OM, CP, EE (P<0.01), and NFE (P<0.05), but decreased CF (P<0.01). The fermentation duration up to 40 d decreased CF 16.72% compared to 10 d. The decrease of the CF content was due to biostarter had both cellulases and xylanases activity, those were 0.001 U/mg and 0.308 U/mg respectively.

According to (Koukiekolo *et al.* (2005) cellulose and hemicelluloses are the major components of plant cell walls. Cellulolytic and xylanolytic enzymes degrade plant biomass to more valuable products. In nature, a variety of bacteria and fungi produce cellulases and xylanases to hydrolyze these insoluble polysaccharides to soluble oligomers, and subsequently to monomers (Paripok *et al.*, 2010). The end-product of this plant biomass degradation will be continually bio-converted by lactic acid bacteria which form lactic acid which also decrease pH value. The obtained results agree with previous results reported by Hanim *et al.* (2011) who obtained CF content. *A. Pinnata* stem waste decreased when fermented with *Aspergillus niger* which produced cellulases.

Table 1. Effect of fermentation duration on chemical composition of fermented aren stem waste

Parameters (%)	Fermentation duration (d)			
	10	20	30	40
Dry matter ^{**}	38.88 ^a	46.85 ^b	46.25 ^c	52.58 ^d
Organic matter ^{**}	95.76 ^a	97.28 ^c	96.50 ^b	95.85 ^a
Crude protein ^{**}	3.93 ^a	4.97 ^{bc}	4.78 ^b	5.30 ^c
Crude fiber ^{**}	37.91 ^c	36.80 ^c	34.62 ^b	31.57 ^a
Ether extract ^{**}	2.76 ^a	3.81 ^b	4.01 ^c	5.01 ^d
NFE [*]	51.16 ^x	51.70 ^{xy}	53.09 ^{yz}	53.97 ^z

* P<0.05

** P<0.01

In vitro digestibility

In vitro digestibility including IVCFD, IVOMD, and IVDMD of aren stem waste was 12.39%, 21.84%, and 23.98%, respectively.

Table 2. Effect of *A. niger* addition on *in vitro* digestibility of fermented aren stem waste

Parameters (%)	Fermentation duration (d)			
	10	20	30	40
IVCFD ^{**}	12.77 ^a	13.23 ^a	15.44 ^b	16.60 ^b
IVOMD ^{**}	21.89 ^a	22.71 ^a	25.65 ^a	27.74 ^c
IVDMD ^{**}	28.74 ^a	29.10 ^a	32.43 ^b	35.84 ^c

** P<0.01

Fermentation process improved *in vitro* digestibility of aren stem waste. As shown in Table 2, the longer fermentation duration increased its digestibility (P<0.01). The increase in IVCFD resulted by fermentation process, because the biostarter produced cellulase and xylanase that degraded fiber, as showed in Table 1 that CF content decreased when aren stem waste was fermented with biostarter and longer fermentation time. The increase in IVCFD at 40 d fermentation time was 23.07% compared to 10 d. This result was agreed with previous study by Hanim *et al.* (2011) showed that fermentation of aren stem waste using *A. niger* enhanced its *in vitro* digestibility.

CONCLUSION

The most effective of fermentation duration of aren stem waste which enhanced chemical composition and *in vitro* digestibility was 30 d.

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THE EFFECT OF DIFFERENT BINDER ON PELLET QUALITY

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ABSTRACT

This research was conducted to find out the effect of different binder on pellet quality without steaming process production. The experiment used rice bran, corn, soybean meal, pollard, *Dried Destilated Grain with Soluble*(DDGS), and molasses and cassava meal as binder. The feed ingredients were ground by Screen mesh Tyler No. 14. The level of binder was 5% and 10% (%DM), respectively. Feed ingredients, binder, and water were mixed to reach 50% DM, thereafter start to process on pellet production using pelletizer with 9 mm diameter of die. After pellet processing, pellet was dried using oven 60° C to reach 14% water content. Pellet was observed by physical and chemical analyzer, there were durability, hardness, compactness, texture, colour, homogeneity, and proximate analyze. Data was analyzed by Complete Randomized Block Design. Results showed that feed ingredients have similarity on Modulus of Uniformity. Modulus of Fineness showed all feed ingredients were meal. Different binder and level of binder did not change chemical quality, otherwise physical quality showed that pellet was compact, hard, homogen, but showed different colour as the binder used. Pellet hardness was not significant different. Pellet quality was good, different binder and level of binder did not change physical and chemical quality.

Keyword: *binder, pellet, quality*

INTRODUCTION

Pellet is agglomerated feed formed by compacting and forcing through die openings by a mechanical process (Parker, 1988). The advantages feed pellet are reducing feed waste, preventing a decomposition of feed components, improving palatability and shortening eating time, reducing the feed selection, increasing feed efficiency and animal performance, and easier of feed handling.

Variables that affect pellet processing is feed moisture and materials. Ziggers (2004) stated that the ration consists of maize or soy bean is difficult to produce good physical quality (durability), but when binder added into the pellet ingredients will improve the physical quality of pellet. Ensminger and Olentine (1978) stated that in order to produce pellets with good texture can use 5-15% of dry matter feed ingredients of molasses as binder. Feed components on pellets formed and bound with binder will cause a compact pellet structure. Feed ingredients are very important on the good quality of pellet processing. The binder (eg, starch and molasses), protein, fiber, minerals, and fats from ingredients will affect pellet quality. Pellet quality can be measured by chemically, physically and biologically. Pellet physical qualities consist of durability, hardness, appearance, texture, color, uniformity, and compactness (Parker, 1988; Thomas, 1998).

MATERIALS AND METHODS

Materials

This study used rice bran, corn, soybean meal, *Dried Destilated Grain with Soluble* (DDGS), and pollard. Cassava strach and molases were used as binder.

Methods

All feed components were ground (Screen mesh tyker No. 14) to achieve same size and mixed with a mixer until homogene. Rations were mixed with the binder with a level of 5% and 10% (basis dry matter): ration with cassava strach binder 5% and 10% level and ration with molasses binder 5 % and 10% level. Each ration was mixed with water to achieve 50% of water content. The ration were put in a pellet machine with a 9 mm die size, then were dried to achieve 14% of moisture content. Pellets were analyzed proximate analysis (AOAC, 1985) to determine the chemical quality, physical quality (hardness, cohesiveness, texture, color and uniformity) (Parker, 1988; Thomas, 1998). The data obtained were analyzed using analysis of variance with Randomized Complete Block Design.

RESULTS AND DISCUSSION

Modulus of Uniformity and Modulus of Fineness

The value of Modulus of Uniformity and Modulus of Fineness of the feed materials that are made into pellet is presented on the Table 1 below.

Table 1 . Modulus of Uniformity and Modulus of Fineness

Feed Materials	Modulus of Uniformity			Modulus of Fineness
	Coarse	Medium	Fine	
DDGS	9.09	0.42	0.48	flour
Pollard	7.70	0.95	1.40	flour
Corn starch	6.95	1.55	1.55	flour
Soybean Meal	9.10	0.35	0.50	flour
Bran	6.40	1.30	2.35	flour

The process of pellet making is strongly influenced by its constituent materials. To obtain a good quality pellet, an even pellet size is needed. Modulus of uniformity and fineness are measured by using the Shieve and Shaker tool to test the uniformity (Modulus of Uniformity) of pellets material. The results obtained were DDGS, pollard, corn starch, soybean meal, and bran had the same size, in which most of them were coarse, and then followed by medium, and fine. While Modulus of Finenes showed that all materials used was in form of flour.

Chemical Compositions of Pellet

The chemical compositions of pellet that is produced by a different binder is presented on the Table 2.

Table 2. The chemical compositions of pellet produced by a different binder *

Treatment	Chemical Composition (%DM) ^{ns}					
	DM	Ash	CP	EE	CF	NFE
Tapioca 5%	94.42	6.20	11.90	7.20	9.28	65.42
Tapioca 10%	94.40	6.01	10.95	6.48	9.32	67.24
Molasses 5%	95.00	6.49	11.77	7.33	9.76	64.65
Molasses 10%	94.58	6.67	11.74	6.99	9.66	64.94

*Analysis conducted on the Lab. of Animal Feed Technology, Faculty of Animal Science UGM 2010

Based on the proximate analysis, all treatments did not show any significant difference to the chemical compositions. Pellet produced by 5% and 10% of tapioca binder and 5% and 10% of molasses binder resulted in dry matter 94,42%, 94,40%,

95,00%, and 94,58%, respectively; ash 6,20%, 6,01%, 6,49%, and 6,67% respectively; crude protein 11,90%, 10,95%, 11,77%, and 11,74% respectively; crude fat 7,20%, 6,48%, 7,33%, and 6,99% respectively; crude fiber 9,28%, 9,32%, 9,76%, and 9,66% respectively; and NFE 65,42%, 67,24%, 64,65%, and 64,94% respectively. The treatment using different binder of tapioca 5 and 10% and molasses 5 and 10% did not change the chemical compositions of pellet. Mixing process was done manually and did not use the mixer owned by the laboratory of FAS due to its very large capacity. However, from the results of the proximate analysis, it can be said that the mixing process can be done well and homogeneous.

Physical quality of pellet

The physical quality of pellet produced by different binder is on the Table 3 below.

Table 3. Physical quality of pellet produced by different binder

Treatment	Physical Quality				
	compactness	texture	color	uniformity	Hardness (kg/cm ²) ^{ns}
Tapioca 5%	Compact	Coarse enough	Whitish brown	uniform	3.99
Tapioca 10%	Compact	Coarse enough	Whitish brown	uniform	4.10
Molasses 5%	Compact	Coarse enough	brown	uniform	3.54
Molasses 10%	Compact	Coarse enough	Dark brown	uniform	4.76

The physical qualities of the pellets cover the compactness, texture, color, and uniformity that are seen by some panelists while the hardness was measured by using a tool for measuring the tenderness of meat (Warner Blaztler). Pellet given tapioca binders 5 and 10%, and molasses 5 and 10%, showed the same compactness, pellets that use materials with the same softness produce a compact pellet. Texture of pellets in all treatments also showed the same hardness after heated in the oven until the water levels reached 14%, this is in accordance with the value of hardness which also showed no difference, namely 3.99 kg/cm², 4.10 kg/cm², 3.54 kg/cm², and 4.76 kg/cm², respectively. A good pellet with a diameter of 9 mm has a hardness of 6.5 so that it's still necessary to increase the pellet's level of hardness, for example by the reducing the quantity of water added. The pellet hardness is also greatly influenced by its constituent materials; the pellets containing high starch will certainly have a high hardness due to the gelatinization. Materials and binder will form a physical-chemical bond during the pelleting process so that it will affect the quality of the pellet (Anonymous, 2010).

The physical quality of pellets is influenced by: 1. The component of constituent materials, especially protein, starch, crude fiber and fat. Materials that are rich in protein will be plastic when they are exposed to the heat, so that they are easy to mold and could improve the appearance of the pellets. Crude fiber is difficult to be molded, but on the other hand, in an adequate amount, it could be a reinforcing material for the pellet. Fat can reduce the dusty nature, but if the amount is excessive it could cause clumps of material in the mixer; 2. The material condition before

molding, the humidity, temperature and particle size, in which the more extensive the particle size (the finer the material), it will expand the area of contact between the particles, so that the bonds between the particles become stronger; 3. The instrument used, for example, the speed of the mold rotation and the material catch. A high composition of crude fiber could cause a difficulty in the molding process, but it gives a positive effect to the pellet hardness (Tjokrokoesoemo, 1986 in Sutrisno, et al. (1994).

A pelleted complete feed for calves with a diameter of 5 cm expired milk powder binder of 5 and 10% resulted in the hardness 4.35 and 4.32 kg/cm² (Budhi et al, 2009). While Andriyani et al., (2010), mentions that the pellet hardness of fermented rice straw with dextrin binder 15%, 20% and 25% resulted in 3.95; 4.23; and 4.77 kg/cm² respectively. Patrick and Schaible (1979) explain that the binder or adhesive is crucial to form the pellet hardness. The harder the pellet, the higher the durability.

The results of the pellets will have the same uniformity, although the pellet length has not be cut with the same size as the cutting tool is not installed, the size of the pellets can be said to be relatively the same. While the die used in the research was a 9 mm die.

The pellet with 5% molasses binders produces a brown color whereas that of 10% produces a dark brown color. Pellets with tapioca binder 5 and 10%, produces the whitish brown color. These color effects are caused by the color of binders used. Molasses that contain 25-40% of sucrose and 12-25% of reducing sugar with a total sugar content of 50-60% have a brownish black color that affects the color of pellets. Tapioca has a white color because of the high starch content so that when it is used as binder, it also affects the color of pellets (Anonymous, 2010).

CONCLUSION

The quality of the pellet resulted in the research was quite good, the use of different types and levels of binder did not cause any changes in the physical and chemical quality and quantity of pellets.

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ENHANCING NUTRITIVE VALUE OF JATROPA LEAVES AS ANIMAL FEED BY VARIOUS PROCESSING METHOD

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ABSTRACT

The main aim of this study was to investigate the characteristics of chemical and nutritive value of *Jatropha* leaves that is associated with its potential as animal feed by determine the quality of various processing method of *Jatropha* leaves (dried, steamed, fermentation and hydrolysis). The research was done in two periods. Firstly, laboratory experiment using proximate analysis and *in vitro* gas production method with our treatments, namely T1 = Non processing of *Jatropha* leaves (control), T2 = fermented *Jatropha* leaves using tempeh yeast, T3 = Steamed *Jatropha* leaves, T4 = hydrolyzed *Jatropha* leaves using NaOH. Parameters measured were: 1). yield of gas (Makkar, 1995), 2). Parameters b and c, 3). Dry matter (DM) and organic matter (OM) residue digestibility, 4). Metabolizable Energy (ME), Nett Energy (NE), Microbial protein (MP) and Organic matter digestibility (OMD) (Menke and Steingass, 1988). Secondly, *in-vivo* experiment using male Boer goat with parameters : intake, digestibility, daily weight gain, urea and glucose blood. The first experiment results showed that there was slightly increase on nutritive value of T2 in crude protein (CP) of *Jatropha* leaves then the other treatment. The results of *in vitro* method showed that the yield of gas T1 has the highest value of gas production, DM and OM digestibility, b and c parameter, ME, NE, MP and OMD compared to other treatments. This result indicates that the non processing leaves of *Jatropha* treatment was the best treatment viewed from *in vitro* gas production method. The results of second experiment showed that increasing the level of dried *Jatropha* leaves on complete feed as a supplement feed cause decreasing of intake, digestibility, and daily weight ($P < 0.05$) and there were no significant effect to urea and glucose blood ($P > 0.05$) but the trends was got lower. In general, It can be concluded that *Jatropha* leaves can not as a supplement feed for goat because of its secondary metabolit contents.

Key words : *Jatropha curcas. L*, *in vitro*, *in vivo*, processing, nutritive value

INTRODUCTION

One of the alternative energy sources that can be renewable are plant-based fuel called of biofuels (biodiesel). One of the plants that can be used as raw material for biodiesel is seeds of *Jatropha* (*Jatropha curcas* , *L*) . *Jatropha* seeds based farming is considered quite beneficial, because its also produces waste of leaves that is considered useful as green manure. For the purpose of ruminant feed, *Jatropha* leaves is less palatable (not preferred), besides that this leaves suspected of containing poison of "krusin" (Berenbaum, 2010) which may be quite harmful to livestock. In addition *Jatropha* leaves contains anti nutrients such as tannins (Butler and Bailey, 1973). Until now, no research related to *Jatropha* leaves for ruminant feed. Yusuf (2008) reported that the leaves of *Jatropha* contains high crude protein (19.95 %), low crude fiber (16.66 %) and the *in-vitro* digestibility of 59.40 %. Based on the chemical characteristics and nutritional information, *Jatropha* leaves have the potential for ruminant feed.

Based on the above description, it have been conducted a study to determine the nutritive value of leaves of *Jatropha* (*Jatropha curcas L.*) using *in vitro* gas production method and *in vivo* experiment.

METHODOLOGY

The methodology of the research in current study were done in two stage, laboratories method namely in vitro gas production and in vivo method.

Experiment 1 : In Vitro

This research aimed to determine the quality of Jatropha on various processing (dried, steamed, fermentation and hydrolysis) of Jatropha leaves using in vitro gas production method . The material of research used for in vitro experiment were : 1). Jatropha (*Jatropha curcas* L.) , 2 . tempeh yeasts , 3) . Na₂CO₃ , 4). Rumen fluid. Chemical characteristics of Jatropha forage was also determined. Four treatments, namely T1 = Non processing of Jatropha leaves (control) T2 = fermented Jatropha leaves using tempe yeast, T3 = Steamed Jatropha leaves, T4 = hydrolyzed Jatropha leaves using NaOH. Parameters measured were: 1). yield of gas (Makkar, 1995) 2). Parameters b and c , 3). DM and OM residue digestibility, 4).ME, NE, MP and OMD (Menke and Steingass,1988). Data obtained from this study were analyzed using analysis of randomized block design.

Experiment 2 : In Vivo

The aim of this second experiment was to determine the effect of adding dried jatropha leaves to performance of Boer goat. There were 4 treatments and replicated 4 times. P1 = Tebon jagung (ad lib) + 300 g Concentrate (C); (Control), P2 = Tebon jagung (ad lib) + 280 g C + 20 g dried Jatropha leaves, P3 = Tebon jagung (ad lib) + 260 g C + 40 g dried Jatropha leaves , P4 = Tebon jagung (ad lib) + 240 g C + 60 g dried Jatropha leaves. Parameters measured were : 1) Intake, 2) Dry Matter and Organic Matter Digestibility,3) Average Daily Gain and 4) BUN and blood glucose. Data obtained from this study were analyzed using analysis of randomized block design.

RESULTS AND DISCUSSION

Experiment 1 Chemical characteristics of Jatropha leaves

The results of the proximate analysis, ADF, NDF, hemisellulosa, and cellulose, as well as the content of anti-nutritional substances such as tannins and saponins of Jatropha can be seen in Table 1.

Table 1. Nutrient content of dry leaves of *Jatropha curcas*

Nutrient content	(%)
Dry material (DM)	84.08
Crude Protein (CP)	24.55
Crude Fat (CF)	2.92
Crude Fiber (CF)	15.23
ash	18.46
Organic materials (OM)	81.54
ADF	30.71
NDF	33.25
tannin	14.34
saponins	4.68

Table 1 shows that the CP content of Jatropha leaf was quite high, as much as 24.55%, so that Jatropha can be classified as protein sources of forages. However, this leaves has constrain due to high ADF and tannin as much as 30.71% and 14.34% respectively. The content of DM, OM and CP of processed Jatropha leaves

analyzed in the Laboratory of Animal Nutrition, Faculty of Animal Husbandry Brawijaya University can be seen in Table 2.

Table 2. Nutrient content of processed Jatropha leaves

Treatments	DM* (%)	OM* (%)	CP* (%)
Non processing (control) (T1)	91,74	82,76	24,55
fermented Jatropha leaves (T2)	89,74	81,32	25,47
Steamed Jatropha leaves (T3)	88,46	82,38	24,36
hydrolyzed Jatropha Leaves (T4)	89,27	83,54	23,98

Table 2 shows that CP of fermented jatropha leaves increased slightly, but the steamed processing and hydrolysis virtually no change. This implies that various processing was not increase nutrient content of jatropha leaves.

***In vitro* gas production**

The results of the *in-vitro* gas production of processed jatropha leaves in different incubation periods up to 72 hours are presented in Table 3.

Table 3. *In-vitro* gas production for all treatment of processed jatropha leaves in different incubation period until 72jam

Treatment	Gas Production (ml/500 mg DM)						
	2	4	8	12	24	48	72
Control (T1)	2,46	5,74	13,60	27,02	49,71	60,93	65,11
fermented Jatropha leaves (T2)	2,89	5,35	11,14	16,58	31,59	36,99	39,16
Steamed Jatropha leaves (T3)	1,80	4,50	13,67	28,17	46,13	55,38	59,23
hydrolyzed Jatropha Leaves (T4)	0,57	2,46	3,61	6,73	24,58	38,59	43,66

Table 3 shows that Jatropha leaf (without treatment =control) has the highest gas production compared to the other treatment. However fermented Jatropha leaf treatment which is expected having high potential to be fermented in the rumen, the value of gas production are below steamed treatment .

The average value of gas production potential is changed as fermented organic matter (OM) potential (b) and gas production rate is translated as fermentation rate per hour (c) of feed that can be seen in Table 4 below. Statistical analysis results showed that the various processing Jatropha leaves has significant (P < 0.01) effect on parameters of a, b and c. Table 4 shows that Control treatment has b and c value higher than other treatments. In addition fermentation treatment was not able to increase its potential to be easily fermented. Thus Jatropha leaves without processing is still better in terms of gas production parameters as well as the value of a, b and c.

After knowing the value of a, b and c, it could be made a graph in according with the equation exponential formula (Orskov, 1994) which is also a relationship graphic between the gas production of feed with various incubation periods.

Table 4 . Parameters of a, b, and c of *in vitro* gas production of all treatments with different incubation period until 72 hours

Treatment	Gas production parameters		
	a (ml)*	b(ml)**	c(ml/hour)**
Control	-1.20793 ^a	74.58418 ^a	0.0458 ^a
fermented Jatropha leaves	-1.57328 ^{ab}	43.37074 ^c	0.0522 ^{ab}
Steamed Jatropha leaves	-4.3148 ^d	68.21703 ^{ab}	0.0522 ^{ab}
hydrolyzed Jatropha Leaves	-2.82376 ^c	66.00394 ^b	0.0205 ^c

* Different superscript in the same column showed highly significant differences (P < 0.01)

Based on Table 3 and Figure 1 shows that the gas production of feed for all treatments tended to increase rapidly after 24 hours of incubation up to 72 hours. This suggests that the availability of organic matter (OM) feed is still quite a lot to be fermented by rumen microbes and the possibility after 72 hours of incubation the graph will tend ramps . Table 5 and graphs of Figure 1 shows that the gas production of non treatment of Jatropha was the highest followed by the steamed, hydrolyzed and fermented processing.

Digestibility

The average value of digestibility of DM and OM of residual *in vitro* gas production can be seen in Table 5 .

Table 5 . The average value of digestibility of DM (DMD) and OM (OMD) of residual *in vitro* gas production of all treatments of jatropha leaves a with different incubation periods up to 72 hours

Treatments	DMD (%)	OMD (%)
Control	28.89 ^a	43.15 ^a
fermented Jatropha leaves	11.36 ^c	31.93 ^b
Steamed Jatropha leaves	24.59 ^b	43.05 ^a
hydrolyzed Jatropha Leaves	27.85 ^{ab}	45.21 ^a

* Different superscript in the same column showed highly significant differences (P < 0.01)

Statistical analysis results showed that the various processing of Jatropha leaves had significant (P < 0.01) effect on the digestibility of DM and OM. Table 5 shows that the non processing treatment (control) has digestibility of DM and OM value higher (P < 0.01) than the other treatments (except hydrolyzed OMD) and the lowest digestibility of DM and OM was fermentation treatment. Fermented treatment that having high CP content was also low in digestibility of DM and OM, where this result is in line with the results of gas production and the value of b and c. High value of digestibility of DM and OM hydrolysis treatment, probably due to hydrolysis of Na₂OH can break the bonds of lignin and cellulose so that digestibility was increased. Fermentation treatment of digestibility of DM and OM were low, probably due to the increased content of mycelia (from yeast tempeh) that is difficult to digest.

ME, NE, and MP

The values of ME, NE, MP and OMD of all treatments can be seen in Table 6. Table 6 showed that the value of ME, NE, OMD and MP of non processing treatment (without treatment) was the highest (P < 0.01) followed by steamed, fermented and hydrolyzed treatment. MP (microbial protein) value of non

processing (dry) is also have the highest value. These values are in line with the gas production and digestibility of non processing but for hydrolyzed treatment has the lowest value. Based on the result of the research it could be said that the processing applied to the leaves of Jatropha does not have a positive impact, and non processing Jatropha leaves could be chosen viewed on practical and economically.

Table 6 . ME , NE , OMD and MP of all treatment of Jatropha leaves with various incubation periods up to 72 hours

Treatment	ME (MJ/kgDM)	NE (MJ/kgDM)	OMD (%)	MP (g/kgOMD)
Control	5.30 ^a	4.03 ^a	43.08 ^a	51.96 ^a
fermented Jatropha leaves	4.16 ^c	3.40 ^b	36.63 ^b	44.19 ^b
Steamed Jatropha leaves	5.07 ^a	3.90 ^a	41.80 ^a	50.42 ^a
hydrolyzed Jatropha Leaves	3.72 ^d	3.16 ^b	34.14 ^b	41.18 ^b

* Different superscript in the same column showed highly significant differences (P <0.01)

Experiment 2

Based on the result of Experiment 1, research was continued with in vivo experiment which is used dried Jatropha leaves as supplement feed for male Boer goat, and the result can be seen in Table 7. Table 7 showed that average DMI all the treatments were 3.36 % until 3.76 % of bodyweight. It was convenient with Soebarinoto et. al (1990) that DMI ruminant were 2 – 4 % from bodyweight. It can be said that feed treatment in these research already fulfill the animal needs. The result also showed that there was a tend to decrease of DMI along with the increase of Jatropha leaves as a supplement feed. It could be happened because of the bitter and a little bit sticky of jatropha leaves were affected the palatability.

The result of Covariance analysis showed that initial bodyweight did not give influence to digestibility (P>0.05), but Variance analysis indicated significant influence (P<0.05) to DMD and OMD. The data described that P2 was not different statistically with P1 (control), it could be decided that Jatropha leaves as supplement feed best in level 20 g/head/day (P2). Its also supported with data of BUN, blood glucose and ADG in Table 8. Although All treatments were tend to decrease, but P2 has the same superscript with P1, it means still the best feed treatment among other treatments.

Table 7. Average Intake and Digestibility of male Boer goat fed Jatropha leaves as supplement feed

Treatments	Intake (g/head/day)		
	DM	OM	CP
P1	815.00 ± 64.23 ^a	653.50 ± 68.90 ^a	93.70 ± 4.39 ^a
P2	798.80 ± 69.29 ^{ab}	645.19 ± 54.46 ^{ab}	92.37 ± 6.78 ^{ab}
P3	732.74 ± 59.58 ^b	596.62 ± 51.38 ^b	85.94 ± 5.32 ^b
P4	750.20 ± 48.78 ^b	610.30 ± 59.43 ^b	87.68 ± 7.89 ^b
		Digestibility (%)	
P1	68.35 ± 12.13 ^a	59.25 ± 11.49 ^a	
P2	66.89 ± 9.27 ^a	57.54 ± 11.54 ^a	
P3	61.58 ± 9.34 ^{ab}	53.52 ± 13.43 ^{ab}	
P4	58.23 ± 9.83 ^b	50.37 ± 16.67 ^b	

^{a-b} Different superscript in the same column showed significant differences (P <0.05)

Table 8 showed that BUN, Blood glucose and ADG tend to decrease with the increasing of jatropha leaves as supplement feed. It can caused by highly tannin content of Jatropha leaves that can inhibit the fermentation in the rumen, disturb nutrient utilization and influence the palatability.

Table 8. Average BUN, blood glucose and average daily gain (ADG)

Treatments	BUN (mg/dl)	Blood glucose (mg/dl)	ADG (g/head/day)
P1	24.35 ± 1.39 ^a	52.32 ± 3.49 ^a	81.56 ± 7.69 ^a
P2	21.64 ± 2.58 ^a	49.87 ± 2.87 ^a	77.48 ± 6.53 ^a
P3	20.34 ± 2.78 ^a	50.23 ± 3.64 ^a	74.38 ± 5.37 ^{ab}
P4	20.59 ± 1.38 ^a	50.68 ± 3.59 ^a	71.25 ± 8.19 ^b

^{a-b} Different superscript in the same column showed significant differences (P <0.05)

CONCLUSION

Based on the test of the *in vitro* gas production method showed that Jatropha leaf without treatment has the highest value of gas production, digestibility of DM, OM, the value of b and c, the value of ME, NE, OMD and MP compared other treatment. This indicates that the dried leaves of Jatropha treatment was the best treatment viewed from *in vitro* gas production method.

The results of second experiment showed that increasing the level of dried Jatropha leaves on complete feed as a supplement feed cause decreasing of intake, digestibility, and daily weight (P<0.05) and there were no significant effect to urea and glucose blood (P>0.05) but the trends was got lower. In general, It can be concluded that Jatropha leaves can not as a supplement feed for goat because of its secondary metabolit contents. It need another research for processing of Jatropha leaves, like making silage of jatropha leaves with BAL, etc.

Acknowledgement

The main authors is gratefully acknowledge to the the Directorate General of Higher Education (DIKTI) the Ministry of Education and Culture (KEMENDIKBUD) the Government of Indonesia Republic and and Brawijaya University (UB) for financial support.

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PHYSIOLOGICAL AND NUTRITIONAL STATUS OF THE PIGS RAISED IN A HIGH ENVIRONMENTAL TEMPERATURE

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ABSTRACT

Negative effect of hot and humid environmental temperatures has been elaborated in many places in the world. Yet, little is known about the effects of a high environmental temperature on physiological and nutritional status of the pigs. The present study was designed to characterize physiological and nutritional status of the pigs by comparing highland (as a control) and coastal area represented the high environmental temperature. Fifteen pigs each (15 pairs of littermates) were assigned to one of two treatments: control (CT) or higher environmental temperature (HET). Feed intake was paired between treatment groups. Each variable was averaged and means were statistically analyzed using t test. Research results showed that HET increased respiratory rate (47.0 vs. 23.0 breath/minute; $P < 0.01$), rectal temperature (39.2 vs. 37.4°C; $P < 0.05$). HET pigs increased water consumption (6.8 vs. 5.2 liter; $P < 0.05$), decreased daily feed consumption (2.80 vs. 3.40 kg), and daily gain (0.65 vs. 0.54 kg). Plasma osmolality was reduced in HET pigs compared with CT (292.0 vs. 287.0 mOsmol/kg, $P < 0.05$). Pigs raised in the coastal area recovered during the cool overnight period as indicated by a reduction in respiratory rate and rectal temperature in the morning. The result demonstrates that high environmental temperature alters some physiological status of the pigs.

Keywords: *Pigs, Physiology, Nutrition, Highland area, Coastal area.*

INTRODUCTION

High environmental temperatures have negative effects on animal performance. Under hot conditions, pigs respond differently compared to other animals. Some physiological characteristics of pigs are unique and cause them to respond differently. They do not sweat effectively. Pigs rely on evaporative heat loss and the respiratory tract is the only channel for controlling evaporative heat loss.

The development of a strategy to deal with thermal stress must recognize these physiological limitations. Heat stress induces physiological alterations, such as a disturbance of acid-base balance and increased heart rate, mean blood pressure, etc (Umboh, 1993). Discussion of acid-base balance in pigs must consider other aspects of the physical environment such as heat stress. Heat stress also alters nutrient balance, such as increased mineral loss. Therefore, the composition of the diet should reflect the effect of heat stress on nutrient balance (Patience, 1990; Patience, *et al.*, 2003).

Much of our present knowledge regarding the physiological responses and quantitative effects of a given environment on pig performance is based on investigations with constant temperatures. Generally speaking, pigs are not raised under constant temperatures. Pigs raised in confinement may be subjected to variations in air temperature caused by inadequate mechanisms for temperature control, building design, fluctuating animal heat losses or even changes in outside temperatures (Umboh and Tulung, 2001). An improved understanding of animal : environment interactions will

lead to accurate prediction of the performance level of animals exposed to fluctuating environmental temperatures and thereby help producers in decision-making processes.

The primary objective of the present study was to characterize the physiological and nutritional status of the pigs raised in a high environmental temperature.

MATERIALS AND METHODS

Animals, diet, and management

Thirty Duroc x Spotted Polland China gilts (15 pairs of littermates) were assigned to one of two treatments: control (CAT) or higher environmental temperature (HET) and placed in individual metabolic crates to facilitate the separate collection of faeces. The diet, based on corn, rice bran, copra meal, and fish meal, was formulated (Table 1) to meet or exceed all requirements for growing swine as defined on a percentage basis by the NRC (1988). The diet was offered to the pigs as a mass. Initial body weight of the pigs was 25 ± 4 kg (* mean \pm range) for CAT and 25 ± 5 kg for HET pigs. The animals were placed in individual metabolism crates and adjusted to their new surroundings over a 5-d period and then the diet was introduced. The experiment was initiated following a 5-d acclimation period. After the 30-d treatment period, the experiment was terminated. The experiment was conducted from July to August at the highest monthly temperature throughout the year.

Temperature treatment

Control (CAT) treatment in this experiment represented by highland area of Tomohon (about 800 m above sea level) and high environmental temperature (HET) represented by coastal area around Manado (0-100 m above sea level).

Data recording and measurement

Room temperature and relative humidity. Both room temperature and relative humidity were recorded at 0700, 1430 and 1900 daily. A hand-held thermohygrometer (Cole Parmer, Model 37200-00) was used to measure room temperature and relative humidity. Measurements were taken at the height of the pigs in the metabolism crates.

Table 1. Composition of experimental diets (g/kg as fed).

<u>Ingredients</u>	<u>(%)</u>
Yellow corn	50.0
Rice bran	24.0
Copra meal	10.0
Fish meal	14.0
Salt	1.0
Mineral + vitamin premix	1.0
<u>Nutrients, assayed</u>	100
Dry matter	91.05
Protein (%)	15.75
Crude Fiber (%)	8.40
Ether extract (%)	7.42
Ca (%)	0.56
P (%)	0.57
Cl (%)	0.03
Digestible Energy (DE) (Kkal/kg)	3230.40

Feed and water intake.

Feed and water intake were recorded during the total experimental period. Pigs in both CAT and HET regime were given ad libitum access to feed between 0700 and 1900. At the time of feeding, the unconsumed water was removed and weighed and feed was provided dry. The net quantity of feed and water consumed was recorded. The design of the trough in the metabolism crate essentially eliminated wastage of feed or water. Trays placed below the crates revealed insignificant wastage.

Rectal temperature.

Rectal temperatures were recorded at 0700, 1430 and 1900, using a digi-sense hand-held thermometer (Cole Parmer, Model L-05985-80); the probe was inserted 7 to 10 cm deep into the rectum, and temperatures were recorded.

Respiration rate

Respiration rates were determined by counting flank movements and recorded as frequency per minute. Three measurements were determined during each collection period (0700, 1430, and 1900) and averaged.

Osmolality

Urine osmolality were determined using an Osmometer (5500 Vapour Pressure Osmometer, Wescor, Inc., Logan, Utah, USA). Ten microliters of urine were pipetted, using a Wescormicropipetter (fixed volume), onto a solute-free paper disc in a circular sample holder. The sample holder was then conveyed into the instrument by means of a slide where it is locked. Locking initiated the automatic measurement sequence. The readout gave the osmolality values of the urine in mOsmol/kg.

Statistical analysis

All data were analyzed using the GLM Procedure (SAS Institute, 1989). A Tukey's test (Steel and Torrie, 1990) was used to examine the treatment differences. Differences between treatment means were considered significant when $P < 0.05$.

RESULTS AND DISCUSSION

The air temperatures in the CAT and HET rooms (barns) were recorded over the course of the experiment. The average temperature in the CAT rooms (barns) was about constant at 21.6⁰C (18.0⁰C in the morning, 27.0⁰C in the noon, and 20.0⁰C in the evening). The average temperature in the HET rooms (barns) was 24.8⁰C (20.3⁰C in the morning, 32.0⁰C in the noon, and 22.0⁰C in the evening). The relative humidity in the CAT and HET rooms was measured. It is estimated to have been between 60.0-80.0% in both CAT and HET rooms (barns).

Respiratory rate and rectal temperature as affected by different environmental temperatures are shown in Table 2. Respiratory rates for HET pigs were higher than CAT pigs (47.0 vs 23 breath/min., $P < 0.05$). Rectal temperatures were also elevated in HET pigs compared to CAT pigs (39.7 vs 38.1⁰C, $P < 0.05$). Table 3 summarizes the effects of different environmental temperature on the average respiration rate and rectal temperature at 0700, 1330, and 1900 h. No differences in respiration rate and rectal temperature at 0700 h were found between HET and CAT pigs. Respiration rate and rectal temperature were all elevated in HET pigs compared to CAT pigs at 1330 and 1900 h ($P < 0.05$). HET pigs had lower plasma osmolality (287.7 vs 290.7 mOsmol/kg, $P < 0.05$) compared to CAT pigs. HET pigs had lower plasma osmolality compared with CAT pigs at 1900 h from day 1 to 30 ($P < 0.05$). No differences in plasma osmolality between control and HET pigs were observed at 0700 h and 1430 h.

Daily water consumption, feed consumption, and daily gain as affected by different environmental temperatures are shown in Table 4. Daily water consumption in HET pigs were higher than CAT pigs (6.80 vs 5.20 l, $P < 0.05$). Daily feed consumption and daily gain were reduced in HET pigs compared to CAT pigs (3.40 vs 2.80 kg, and 0.65 vs 0.54 kg respectively, $P < 0.05$).

Table 2. Effect of different environmental temperature on respiration rate and rectal temperature in pigs.

Parameter	Treatment		Sign. Level [#]
	CAT	HET	
Respiration Rate (breaths/min) [§]	23.0	47.0	0.01
Rectal Temperature (□ C) [§]	37.5	39.1	0.05

[#] Treatment effects.

[§] Values are averaged from 3 collection times daily (0700 h, 1330 h, and 1900 h) from day 0 to 30.

Table 3. Time and treatment effects on respiration rate, rectal temperature, and heart rate in pigs.

Parameter	Time (h)	Treatment		Sign. Level [§]
		CAT	HET	
RR (br/min) [#]	0700	14.0	20.0	0.17
	1330	40.0	96.0	0.01
	1900	15.0	24.0	0.01
RT (□ C) [#]	0700	37.1	38.2	0.80
	1330	38.0	40.2	0.01
	1900	37.5	39.0	0.01
Osmolality (mOsmol/kg)	0700	292.0	290.0	0.16
	1430	289.0	287.0	0.08
	1900	291.0	286.0	0.05

[§] Treatment effects

[#] RR = Respiration rate, RT = rectal temperature.

Table 4. Effect of Effect of different environmental temperature on daily water consumption, daily feed consumption, and daily gain in pigs.

Parameter	Treatment		Sign. Level [#]
	CAT	HET	
Water consumption (kg/d)	5.20	6.80	0.05
Feed consumption (kg/d)	3.40	2.80	0.05
Daily gain (kg/d)	0.65	0.54	0.05

[#] Treatment effects

The present study was designed to determine the effect of high environmental temperature on the physiological and nutritional status of the pig. To achieve this objective comparisons were made between animals maintained under hot environmental temperature represented by coastal area of Manado and those kept in thermoneutral (control) conditions represented by highland area of Tomohon. It was estimated that 21.0°C was a thermoneutral zone for growing swine at the standard feeding level. However, Geuyen *et al.* (1984) presented data which indicated that the lower border of thermoneutrality may be as high as 19 to 20°C for growing swine at

standard feeding levels. NRC (1981) and Curtis (1985) pointed out that the environmental temperature range of 18 to 21⁰C generally has been found to be optimal for growing-finishing pigs in terms of productive performance. In this study a control temperature of about 21.0⁰C was chosen to represent a thermoneutral environment. Defining the concept of thermoneutrality, NRC (1981) and Yousef (1985) stated that within the range of effective ambient temperature (EAT) over which the body temperature remains normal, sweating and panting do not occur and metabolic heat production remains at a minimum. In the middle range of these environmental conditions, pigs show signs of thermal comfort. In our study, the control pigs were maintained within a thermoneutral environment as indicated by the respiration rate and rectal temperature (Table 2 and 3) which remained normal and constant. Panting did not occur in the control animals and they were more comfortable compared to the HET pigs during the experiment. Pigs in the CAT room spent their time mostly resting and sleeping and no sign of restlessness were observed during the experiment. In contrast, during periods of hottest day in coastal area (HET), pigs could be observed to alter laying patterns and exhibit a high degree of restlessness.

The temperature of 32.0⁰C (HET) in this experiment was sufficient to increase respiration rate and rectal temperature (Table 2 and 3). These findings suggest that thermal balance was different between CAT and HET pigs. These results are in agreement with those of Flowers *et al.* (1989) and Giles *et al.* (1991). The increase in rectal temperature found under HET conditions emphasizes the degree of heat stress. In these climatic conditions sensible heat losses (by radiation, convection, and conduction) are very low because ambient temperature is comparable to the body temperature and consequently evaporation is the main process for eliminating excess body heat (Christon, 1988).

Compared to many mammalian and avian species, the pig is poorly equipped to handle high environmental temperature stress. The respiratory tract is the major channel to dissipate the heat load from the body. As a result, the pig has to increase respiration rates when the body temperature increases. In the present study, respiration rates increased in HET pigs compared to CAT pigs (Table 2 and 3). The increase in respiration rate is similar to the results reported by Black *et al.*, (1986) who found that pigs increase their evaporative heat loss mainly through an increase in respiration rate. The increase in respiration rate is associated with an increase in body temperature.

Respiration rate and rectal temperature were higher in HET pigs compared with CAT pigs at 1430 and 1900 h (hot period). No significant differences were found between CAT and HET pigs at 0700 h (cool period). These results are in agreement with those of Lopez *et al.* (1991); Giles *et al.* (1991) and Lorsch *et al.* (1991) who found that respiration rate and rectal temperature generally showed a similar diurnal rhythm or nyctohemeral pattern indicating some degree of environmental/heat stress during the hot part of the day. The results also showed that pigs recovered during the cool period overnight as indicated by the lower respiration rate and rectal temperature at 0700 from day 1 to day 30.

Osmolality, which is a measure of all of dissolved solutes in an aqueous solution (Brenner *et al.*, 1987), was lower for HET pigs compared to CAT pigs. A decrease in plasma osmolality can occur following a loss of electrolytes or a gain of water, or both. In the present study, no evident of electrolytes loss since we did not measure net loss of solutes (electrolytes) by analysing plasma constituents. This indicates that the lower osmolality for HET pigs found in the present study was not due

to the loss of electrolytes but merely a result of net water gain in the HET pigs compared with CAT pigs (6.8 vs 5.2 kg/d, $P < 0.05$). In rats, Okuno *et al.* (1988) found that excessive water consumption correlated with a dilution of body fluid and lower plasma osmolality. The significant decrease in osmolality for HET pigs was only shown at 1900 and no significant differences were found between HET and CAT pigs at 0700 and 1330 h (Table 3). The results suggest that the effect of high environmental temperature in pigs raised in coastal area on blood osmolality can only be seen during the peak hot period (at 1900 h measurements). An explanation is that the pigs recovered during the cool night and morning period. Lower osmolality in HET pigs compared to control pigs at 1900 h reflected the change in the pig's drinking pattern. HET pigs consumed more water in the day time (hot period) than in the cool period of the day.

In summary, high environmental temperature increased rectal temperature, respiration rate, daily water consumption, and reduced daily feed consumption and daily gain in pigs. Pigs had to respond behaviourally when exposed to an increase in ambient temperature. HET pigs recovered during the cool overnight period as indicated by a reduction in respiration rates and rectal temperatures at morning (0700 h) measurement. The results demonstrate that high environmental temperature alters some of the physiological states.

CONCLUSION

High environmental temperature in the coastal area of Manado alters the physiological state, decreases daily feed intake and daily gain, and increases daily water intake of the pigs.

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EFFECT OF UTILIZATION OF VIRGIN COCONUT OIL (VCO) IN THE DIET ON PIG PERFORMANCE

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ABSTRACT

Virgin coconut oil (VCO) is one type of coconut oil that has recently gained a lot of attention due to various claimed medicinal values, such as antioxidant, antimicrobial, antiviral, antihypercholesterol, and antithrombotic activities. Yet, little is known about the effect of utilization of VCO in animal diets. The present study was designed to elaborate the effect of utilization of VCO in the diets on pig performances. The experiment was conducted using 20 castrated male pigs aged 6-7 weeks. The data were analyzed according to the linear model procedure for ANOVA appropriate for Randomized Block Design with 5 treatments and four replications. Treatments were: R0 = 100% control diet + 0% VCO; R1 = 99.5% control diet + 0.5% VCO; R2 = 99% control diet + 1% VCO; R3 = 98.5% control diet + 1.5% VCO; and R4 = 98% control diet + 2% VCO. Parameters measured were: energy, protein, calcium, and phosphorus digestibility; feed consumption, water consumption, daily gain and feed efficiency. The results showed that the utilization of VCO up to 2% in the diets had no effect ($P > 0.05$) on daily feed consumption, daily gain, water consumption, feed efficiency; and digestibility of energy, protein, calcium, and phosphorus. It can be concluded that utilization of VCO up to 2% in the diets has no significant influence on pig performance.

Keywords: *Virgin coconut oil (VCO), Diets, Pig performance, Nutrient digestibility.*

INTRODUCTION

The feed cost is the most important cost in pig production and energy represents the greatest proportion of this cost. Due to their high energy value, being approximately 2.25 times that of carbohydrates, the use of fats and oils in diets for pigs is of great importance. One of the commonly used fat sources in pig feed is vegetable fat sources, such as coconut oil or virgin coconut oil (VCO). Virgin coconut oil (VCO) is made by the milk extracted from the mature kernel of fresh coconuts. It is termed as virgin because it is 100% pure as in white, it does not undergo any hydrogenation or any heating process. It is unrefined, unbleached and has not undergone any deodorizing process. Unlike other coconut oil that is extracted from copra through heat, cold pressed VCO does not possess that rancid odor, but fresh aroma of coconut oil. Cold pressed VCO has longer shelf-life of more than two years. Coconut oil is a saturated fat made up primarily of medium chain fatty acids. Also known as medium chain triglycerides (MCTs), medium chain fatty acids are known to increase metabolism.

Fats and oils are important dietary ingredients in animal production owing to their high energy value. Furthermore, the fatty acid pattern of the dietary lipids is reflected in the fatty acid profile of animal products (Jørgensen et al. 1996). However, the lipids used in domesticated animal diets are extremely diverse in chemical structure, which may influence digestibility and energy value.

Yet, little is known about the effect of utilization of VCO in animal diets. The present study was conducted to determine the inclusion of virgin coconut oil (VCO) in the diet on pig performances.

MATERIALS AND METHODS

Animals and diets

Twenty castrated Duroc X Spotted Poland China pigs were obtained from local breeding farm. The pigs were housed individually in pens and had free access to water and a self-formulated grower diet. The basal diet, based on yellow corn, copra meal, fish meal, and cassava meal was formulated (Table 1) to meet or exceed all requirements for growing swine as defined on a percentage basis by the NRC (1988). The diet was offered to the pigs as a mass. The treatments were formulated as follow: R0 = 100% basal diet without VCO; R1 = 99.5 basal diet + 0.5% VCO; R2 = 99.0% basal diet + 1.0% VCO; R3 = 98.5% basal diet + 1.5% VCO; and R4 = 98.0% basal diet + 2.0% VCO. Fresh water was provided in the trough ad libitum until the time of feeding. At feeding, unconsumed water was removed from the trough and weighed. The animals were placed in individual metabolism crates and adjusted to their new surroundings over a 7-d period and then the diet was introduced. The experiment was initiated following a 7-d acclimation period. After the 60-d treatment period, the experiment was terminated. The chemical composition of the experimental diets is presented in Table 1.

Table 1. Composition of experimental diets (g/kg as fed)

<u>Ingredients</u>	
Yellow Corn	570.00
Copra meal	180.00
Fish meal	130.00
Cassava meal	100.00
Vitamin Premix	10.00
Mineral Premix [#]	10.00
<u>Nutrients, assayed</u>	
Dry Matter (%)	89.64
Protein (%)	16.07
Ash (%)	4.91
Crude fiber (%)	4.72
Calcium (%)	10.45
Phosphorus (%)	7.20
Digestible energy (DE) (kcal/kg)	3210.40

The experiment was conducted according to a 5 x 4 Completely Randomized Block Design. The mean body weights of the pigs were 13.5±3.5 kg at the start and 30.5±4.0 kg at the conclusion of the experiment. Total collection method was employed for the determination of the digestibilities of nutrients and energy. Each pig was fitted with a fecal bag in order to minimize contamination during feces collection period. Feces were collected according to standard procedures and were oven dried at 60°C for 72 hours immediately after collection.

Analytical methods

Feces were dried, pooled within pig and diet and ground. Dry matter was determined according to AOAC (1990) methods. Protein (N x 6.25) was measured by the Kjeldahl method using a Kjell-Foss 16200 autoanalyzer (Foss Electric, Hillerød, Denmark). Gross energy was determined with an IKA-C 400 bomb calorimeter. Parameters measured were: energy, protein, calcium, and phosphorus digestibility; feed consumption, water consumption, daily gain and feed efficiency.

Statistical analyses

ANOVA was performed according to a 5 x 4 Completely Randomized Block Design following procedures described by Steel and Torrie (1980). Where appropriate, treatment means were compared using the Tukey’s test (Steel and Torrie 1980). Treatment means were considered statistically different at $P < 0.05$. The calculations were performed with a statistical computer program (SAS/STAT Version 6.10; SAS Institute, Cary, NC).

RESULTS AND DISCUSSION

Energy, protein, calcium, and phosphorus digestibility; feed consumption, water consumption, daily gain and feed efficiency as affected by different level of VCO in pigs are shown in Table 2.

Table 2. Effect of different VCO level in the diets on energy, protein, calcium, and phosphorus digestibility; feed consumption, water consumption, daily gain, and feed efficiency in pigs.

Digestibility (%)	Treatments				
	RO	R1	R2	R3	R4
Energy	82.40	83.08	79.06	77.80	81.03
Protein	85.31	84.05	83.90	82.32	83.80
Calcium (Ca)	70.11	69.24	68.54	68.84	69.33
Phosphorus (P)	64.88	66.02	65.13	65.51	64.88
Daily Feed consumption	2.86	2.84	2.82	2.82	2.81
Water consumption (l)	4.02	4.39	4.17	4.42	4.25
Daily gain	0,86	0,85	0,84	0,84	0,83
Feed efficiency ratio	0,28	0,29	0,29	0,28	0,28

Energy, protein, calcium (Ca), and phosphorus (P) digestibility data in the present study (Table 2) did not significantly ($P > 0.05$) affected by the inclusion of VCO up to 2% in pig diets. Daily feed consumption, water intake, daily gain, and feed efficiency ratio were all also not significantly ($P > 0.05$) affected by the inclusion of VCO in pig diets. We proposed that coconut oil or VCO which is rich in energy would suppress pigs’ feed consumption as the inclusion of coconut oil or VCO in the diet is increased. Carandang (2005) stated that VCO mostly consisted of fat which is characterized by mediumchain fatty acid (MCFA) needed to increase body metabolism and supplies some energy to the animals. Santos et al.,(2005) pointed out that VCO consisted of 48-50% lauric acid, a medium chain fatty acid with C-12 that has an ability to increase the rate of metabolism and absorption of nutrients in the body. Theoretically, the inclusion of VCO in the present study should also increase energy digestibility and other parameters measured. In fact, all parameters measured

were all not significantly ($P > 0.05$) affected by inclusion of VCO in pig diets. We assumed that this discrepancy was more affected by the environmental temperature where this study was conducted. The environmental temperature during the trial was on the range of 22-27⁰C in the morning-afternoon and 16-22⁰C in the evening-morning. These environmental temperature range are considered as a thermoneutral zone of pigs at that age (grower–finisher), so that inclusion of VCO in the diets could not be able to affect energy and other nutrients metabolism. VCO is also known as a source of low heat increment energy and adding VCO in pig diets could lower heat production in the hot environment. It might be different when the present study was conducted in the hotter environmental temperature. Unchanged digestibility of energy, protein, Ca, and P in the present study can also be influenced by palatability factor that can be proven by studying blood glucose level since the increase in blood glucose should directly affect feed consumption. When blood glucose level in the cell is satisfied, then feed consumption decreases because energy is still available in the body (Fife, 2001).

CONCLUSION

It can be concluded that energy, protein, calcium, and phosphorus digestibility; feed consumption, daily gain, and feed efficiency of pigs are not affected by the inclusion of virgin coconut oil (VCO) in pig diets.

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CONSUMPTION AND DIGESTIBILITY OF DRY MATTER AND ORGANIC MATTER IN RUMINANT FEED USING METHANOGENIC INHIBITOR SUBSTRATE

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ABSTRACT

The aim of this research is to investigate effects using different level of medium chain fatty acids (MCFA) as methanogenic inhibitor substrate to evaluate consumption and digestibility of dry matter and organic matter by in- vivo method. Three different proportion of medium chain fatty acids, namely R0: 0% MCFA, R1: 1% MCFA, R2: 1.5% MCFA in 100% dry matter. Each treatment consisted of four sheep. The sheep were kept for three months. The data were analyzed using a variance analysis (completely randomized design), followed by Duncan's new multiple range test (DMRT) for the significant means. The result indicated that R0, R1 and R2 treatment were not different significantly ($p>0.05$) on consumption dry matter and organic matter, while digestibility of dry matter and organic matter was significantly ($p<0.05$). Supplementation with medium chain fatty acids 1% - 1.5% can used in ruminant as methanogenic inhibitor feed because increasing digestibility dry matter and organic matter.

Keywords: *consumption, digestibility, methanogenic inhibitor, ruminant*

INTRODUCTION

Methane gas is one of the fermentation products of feed material by rumen microbes. Many nutritionists of ruminant trying to reduce methane production, because they felt responsible for the contribution of livestock to atmospheric pollution by methane as one of the pollutants that have always been related with the destruction of ozone and global warming (Moss *et al.*, 2000). But the interest of the nutrition experts have been in the energy utilization of feed. The amount of methane emission showed not efficiency of feed utilization by ruminant (Yusiati *et al.*, 2002). Johnson and Johnson (1995) states that 6% of energy intake is lost in the form of methane gas. According to Beauchemin dan Mc Gin (2006), cattle was lost 6% of energy consumption total as methane, meanwhile fattening cattle was lost 3,5% energy consumption total as methane. This is the main issue how to exploit the energy of feed that is not used in the formation of methane but used to improve the productivity of ruminants. There are two mechanisms to inhibit the production of methane that is weakening protozoan (defaunation) and the other is hydrogen zink. Several studies have shown that the role of medium chain fatty acids (MCFA) able to reduce methane gas in the rumen fermentation *in vitro*. According to Sondakh *et al.* (2012b) that the content of 1% MCFA able to reduce methane in the feed of 14.33% and if MCFA was increased to 1.5%, methane content decreased again to 25.30% in the *in vitro* fermentation. The decreasing in methane production due to the reduced number of rumen protozoa. Furthermore Sondakh *et al.* (2012b) stated that the number of protozoa decreased 29.84% when the feed given MCFA 1.5%. Thus,

MCFA is considered to function as a protozoa defaunation agent. In addition, the role of MCFA also able to increase the propionate (Sondakhet *al.*, 2012b). There is competition between propionic acid-forming microbes and microbial methane-forming in getting hydrogen.

Livestock productivity is determined by the quality and quantity of nutrients consumed. Quality of nutrients for ruminants is also influenced by the existence of rumen microbial available. With the decrease in methane production caused the decrease of the number of protozoa. It will affect the degradation of the feed and will certainly affect also on consumption and digestibility of the feed value. A decrease in the amount of methane and protozoa in the rumen fermentation after being given the supplement MCFA and the effect on feed intake and digestibility is an interesting part to study.

MATERIAL AND METHOD

Animal. Twelve male sheep approximately 1 year old with an initial liveweight of 16-17 kg were kept in individual cages shaped stage in three months and, were randomly divided into three groups ration treatment. Each group consisted of four sheep.

Feed. Feed used were consisted of forage and concentrate in the ratio 60:40. Forages used were elephant grass(*Pennisetum purpureum*), while the concentrate used were coconut cake, soybean cake and rice bran with different compositions for each treatment. The experiment consisted of three ration treatments, namely, (I) Ration treatment was containing MCFA 0%, (II) Ration treatment was containing MCFA 1.0% and (III) ration treatment was containing MCFA 1.5%. According to the results of previous studies feeding trial II and III able to reduce methane gas to 14.33% and 25.30% (Sondakh *et al.*, 2012b). For a clearer treatment of the experiment can be seen in Table 1.

Table 1. The composition of the nutrient content of the ration experiment, fat content and MCFA in coconut cake from each ration treatment

Variable	MCFA (%)		
	0	1.0	1.5
Feed materials (%)			
Elephant grass	60.00	60.00	60.00
Rice bran	17.00	6.00	2.00
Soybean cake	23.00	20.00	17.00
Coconut cake	0.00	14.00	21.00
The composition of nutrient			
Crude Protein	17.08	17.28	17.01
Crude Fat	5.93	5.07	5.27
CrudeFiber	23.27	23.29	23.41
Extract Non Nitrogen	42.34	42.35	43.28
Ash	10.46	9.84	9.96
MCFA	0	1.0	1.5

The method of research

Animal that have been weighed initial body weight was kept in individual cages and given fed every day at 08.00 and 15.00 *ad libitum*. Before feeding, the feed first was weighed and then the next day was weighed feed remains being and was

done during the study. Research was conducted for twelve weeks, consists of two weeks of preliminary and ten weeks for collection.

In the sixth week was conducted a study sample collection consisted of samples of feed and residual feed, feces and urine samples. Sample collection was done every day for six weeks. Faeces taken during the collection period was collected then weighed and taken as much as 5% to be stored in the refrigerator 1⁰C, as the sample. Samples of feed remains were collected for each individual, was dried and milled with a milling machine that was equipped with a filter with diametre 1 mm. Results of mill were inserted into a plastic bag for proximate analysis namely, the dry matter (DM), organic matter (OM), crude protein (CP). Faeces shelters result was mixed for 3 minutes and then used for the determination of DM, OM, CP. The variables observed in this study was consumption, digestibility of feed and body weight.

Data Analysis

The data were analyzed using a variance analyses (completely randomized design), followed by Duncan’s new multiple range test (DMRT) for the significant means) (Steel and Torrie, 1980).

RESULT AND DISCUSSION

Consumption of dry matter (DM), organic matter (OM) and protein

Table 2. The average of consumption dry matter, organic matter and protein on sheeps getting feed with MCFA

Feed intake (g/animal/day)	MCFA		
	0%	1.0%	1.5%
Dry matter ^{ns}	879.31 ± 60.60	891.25 ± 57.70	935.80 ± 13.79
Organic matter ^{ns}	779.24 ± 53.23	784.22 ± 50.77	832.58 ± 12.27
Crude protein ^{ns}	150.19 ± 5.36	154.01 ± 5.33	159,18 ± 2.34

The average consumption of dry matter (DM) in the treatment of MCFA 0, 1.0, and 1.5% were 879.31 ± 60.60, 891.25 ± 57.70, and 935.80 ± 13.79 g/animal/day. Based on the analysis of variance is known that the treatment of feed containing MCFA up to 1.5% provides non significant on consumption of dry matter (DM), organic matter (OM) and crude protein (CP). The maximum value of feed consumption largely depends on the balance of nutrients in the gastrointestinal (Preston and Leng, 1984). This is due to the nutritional needs of a major stimulus delivered to the hypothalamus as the hunger center. Furthermore, Preston and Leng (1984) states that the feed nutrient imbalance will affect feed intake. The balance of nutrients in the diet will affect rumen fermentation, which in turn will affect feed intake (Tilmann *et al.*, 1998). Not influential MCFA content in the feed up to 1.5% due to the possibility of feed given already comply the balance of nutrients available in the ration for each treatment has complied the needs of the sheep.

Digestibility of nutrients

Table 3. The average of digestibility of dry matter, organic matter and protein on sheeps getting feed with MCFA

Digestibility (%)	MCFA (%)		
	0	1.0	1.5
Dry matter	61.14 ^a ± 4.21	69.23 ^b ± 5.92	73.92 ^b ± 2.44
Organic matter	66.44 ^a ± 3.46	73.12 ^b ± 5.25	77.37 ^b ± 1.78
Protein	66.72 ^a ± 2.53	73.78 ^b ± 4.10	75.79 ^b ± 2.51

^{a,b}Different superscript on the same line differ significantly (P <0.05).

Digestibility of nutrients which consists of dry matter (DM), organic matter (OM) and crude protein can be seen in Table 3. Judging from the existing data on the table, it seems that the MCFA to give effect to the value of DM, organic matter BO and protein can be seen in Table 3.

MCFA content of 1.0% significant effect (P <0.05) when compared to DM digestibility of feed that does not contain MCFA, while DM on feed digestibility values with the MCFA content of 1.0 and 1.5% showed non significantly. It also applies to the OM and protein digestibility. Degradation of DM, OM weredetermined by the rumen microbial activity. Rumen microbial activity is strongly influenced by the availability of sufficient VFA and NH₃ as a source of energy and N for rumen microbial (Utomo, 2001). According to Sondakhet *al.* (2012b), feed containing MCFA 1% and 1.5% were able to increase the amount of NH₃ and VFA. The increasing in the number of NH₃ causes an increasing in the amount of microbial protein synthesis, which It will increase the degradation of DM and OM. Utomo(2001) stated that digestibility of nutrients in ruminants is strongly influenced by the content of crude fiber and rumen microbial activity, especially cellulotic bacteria.

Average of daily gain (ADG) and feed conversion

Average daily gain (ADG) and feed conversion can be seen in Table 4.

Table 4. The average of daily gain and feed conversion on sheeps getting feed with MCFA

Variables	MCFA (%)		
	0	1,0	1,5
Average daily gain (g)	102.59 ^a ± 4.83	116.77 ^b ± 7.10	119.08 ^b ± 6.03
Feed conversion (g)	8.61 ^b ± 0.58	7.48 ^a ± 0.53	7.87 ^a ± 0.33

^{a,b}Different superscript on the same line differ significantly (P <0.05).

Based on the analysis of variance showed that the feed containing MCFA give significant on ADG (P <0.05).MCFA content in the feed yield in an increase of body weight daily. The content of MCFA 1.0% in the diet caused an increase ADG from 102.59 to 116 g / animal / day (increase 12.15%), if MCFA raised up to 1.5%

in feed, ADG will increase to 15.93%, while the content of MCFA from 1.0% to 1.5% give no effect on ADG. The increase of ADG due to the influence of the MCFA. According Sondakhet *al.* (2012b), MCFA content in the feed plays a role in increasing the levels of propionic acid in the rumen fluid. Propionic acid will prevent ruminalbiohydrogenation. It means the saturated fatty acids are straight immediately into the small intestine and it will be absorbed without ruminalbiohydrogenation (Sondakhet *al.*, 2012a). Most of the essential fatty acids will be transported to the body tissues for regulating of intracellular metabolism to activate cellular enzymes that catalyze the biosynthesis process for growth (Ashes et al., 1995). Unsaturated fatty acids also supported the increasing of gluconeogenesis from propionic acid. The increasing of glucose will stimulate insulin secretion, which further increases the permeability of the cell membrane, thereby increasing the entry of glucose and amino acids into the cell for protein biosynthesis. The process of protein biosynthesis are reminded by the role of second messenger through intracellular enzymes activity (Ashes et al., 1995). These phenomena can be seen in the increase in hyperplasia and hypertrophy of cell tissues through increased muscle mass, as seen in the ADG.

Feed conversion of this research can be seen in Table 4. Based on the analysis of variance showed that the addition of MCFA in rations showed the significant effect ($P < 0.05$) on feed conversion. Treatment of feed by using MCFA 1.0 and 1.5% is more efficient in determining the growth of sheep when compared to without MCFA. Feed conversion value in this study shows the value of the effectiveness in the use of feed containing MCFA. These results are highly correlated with the value of ADG. Feed conversion is the amount of feed intake given to raise the unit body weight.

CONCLUSION

Supplementation with medium chain fatty acids 1% - 1.5% can be used in sheep as methanogenic inhibitor feed because increasing nutrient digestibility, increasing average daily gain and lower feed conversion value.

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THE EFFECT OF *IN VITRO* DIGESTION ON THE ANTIOXIDANT ACTIVITY OF TROPICAL FRUITS PEELS

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ABSTRACT

The objective of this study was to investigate the effect of antioxidant activity of tropical fruit peels on the *in vitro* digestion. The peels used in this experiment were matoa, salak and soursop fruits. All samples were measured the DPPH (1, 1-diphenyl-2-picrylhydrazyl) radical scavenging activity, lipid peroxidation and total phenolic content in the undigested and digested at *in vitro* gastrointestinal digestion. The result in the undigested of samples showed that salak peel (89%) was the highest of DPPH radical scavenging activity, soursop peel (0.21 μmol MDA/mg linoleic acid) was the lowest lipid peroxidation concentration, and matoa peel (7.5 mg GAE/g sample) and salak peel (6.9 mg GAE/g sample) were the highest total phenolic level. After digestion, the result of DPPH radical scavenging activity in soursop and matoa peel was increased after digestion, whereas the result in salak peel was decreased. Increased lipid peroxidation concentrations were observed in matoa, salak, and soursop peel. Increased total phenolic contents were found in matoa and soursop peel, whereas decreased total phenolic contents were detected in salak peel. The antioxidant capacity of the soursop and matoa peels remained high throughout the *in vitro* digestion and these have potent for used as feed additive for animals.

Keywords: fruit peel, gastrointestinal digestion, antioxidant, lipid peroxidation, total phenolic

INTRODUCTION

Feed supplementation containing antioxidant compounds can increase the quality and quantity of livestock production and improve the livestock health status. Antioxidants derived from plants and fruits have phytochemical substances such as α -tocopherol, β -carotene, ascorbic acid, flavonoids, carotenoids, anthocyanins, phenolic compounds, zinc and selenium. These compounds were rich in the fruits peel. Chamorro et al. (2015) reported that supplementation of grape pomace to chick had increase polyunsaturated fatty acids content in meat and prevented lipid peroxidation in meat. And also mangosteen peel powder in swamp buffaloes fed can increase propionic acid production and reduce methane production in the rumen (Wanapat et al., 2014).

On the basis of that observation, there is the possibility of fruit peels of salak, soursop and matoa containing antioxidant compounds that can be used as feed additive. These fruits are easily found in Indonesia. There is no studies that have reported their antioxidant activity and polyphenols content. Furthermore, many studies determined the antioxidant capacities of fruits but the antioxidant activity in the gastrointestinal system was not taken into consideration. The *in vitro* digestion has been used often to simulate gastrointestinal conditions can be considered since

they are relatively simple when compared to the *in vivo* models, besides being safe and do not present ethical restrictions. Then, the purpose of this study was to investigate the effect of antioxidant activity after *in vitro* digestion of tropical fruit peels using DPPH, lipid peroxidation and total phenolic content.

MATERIALS AND METHODS

Sample that were used in this experiment are peels of Sirsak fruit (*Annona muricata* L.), Matoa fruit (*Pometia pinnata*), and Salak fruit (*Salacca zalacca*). The samples were obtained in February 2014. All samples were dried at 50°C for 12 to 24 hours and were ground to a fine powder. All samples were extracted by QuEChERS method (Sato *et al.*, 2015).

All samples were measured total phenolic content, analysis of DPPH radical scavenging activity and analysis lipid peroxidation. Total phenolic content was determined using the procedure of Folin-Ciocalteu (Asada & Tamura, 2012) with slight modification. The free radical scavenging activity of the extracts was measured using the DPPH (1, 1-diphenyl-2-picrylhydrazyl) radical scavenging method described by Zhu *et al.* (2014). Lipid peroxidation inhibition was measured by using a thiobarbituric acid (TBA) method (Tamura and Yamagami, 1994).

Gastrointestinal in vitro digestion

Based on the methodology was described by Yonekura *et al.* (2014), with a little modification. Samples dried powders were mixed with 5 ml of pepsin buffer solution in 25 ml conical flask (1600 unit/ml pepsin, 3.6 mM CaCl₂, 12 mM KCl, 1.5 mM MgCl₂.6H₂O, 49 mM NaCl, 6.4 mM KH₂PO₄). The mixture was acidified to pH 2.5 with 2 M HCl and the mixture was placed in a shaker bath at 37°C for 1 hour. After step-wise gastric digestion, intestinal digestion was performed with the addition of 5 ml of pancreatin-bile solution (0.682 g of pancreatin and 0.062 g bile extract in 155 ml of 0.1 M NaHCO₃) and the pH of solution was adjusted to 6.8 with 2 M HCl. The mixture was incubated in a shaker bath at 37°C for 2 hour. The samples were filtered and analyzed again by DPPH radical scavenging activity, lipid peroxidation and total phenolic.

Statistical analysis

All data are expressed as mean ± standard deviation (SD) in triplicate at least. For comparisons between samples, data was analysed by ANOVA and Duncan test (SPSS, version 16.0). A probability of 5% or less was accepted as statistically significant.

RESULTS AND DISCUSSION

All samples were subjected to an *in vitro* digestion procedure designed to stimulate *in vivo* digestion. The digestion process directly affects the composition of the extracts depending on the simulation of the physiological conditions and the sequence of events within the gastrointestinal tract.

Table 1 showed that there was significant (P<0.05) difference between the undigested values and digested of tropical fruit peels in the DPPH radical scavenging activity, lipid peroxidation, total phenolic content. At a concentration 50 µg/ml and 10 µg/ml, the DPPH radical scavenging activity of matoa peel (70.2-81.8%) and soursop peel (35.0-75.3%) exhibited a significant increase in the undigested and digested fruit peels. Pavan *et al.* (2014) stated that pH and enzymatic interactions during digestion could cause the increase of antioxidant activity.

Table 1. DPPH radical scavenging activity, lipid peroxidation inhibition, and total phenolic undigested and digested tropical fruit peels

Sample name	Parameter	<i>In vitro</i> digestion	
		undigested	Digested
Soursop peel (50 µg/ml)	DPHH radical scavenging activity (%)	35.0 ± 0.4 ^{aX}	75.3 ± 3.1 ^{bY}
Matoa peel (50 µg/ml)		70.2 ± 0.3 ^{aY}	81.8 ± 0.3 ^{bZ}
Salak peel (50 µg/ml)		89.1 ± 1.3 ^{bZ}	32.5 ± 1.5 ^{aX}
Soursop peel (10 µg/ml)	DPHH radical scavenging activity (%)	9.5 ± 0.4 ^{aX}	25.8 ± 1.4 ^{bZ}
Matoa peel (10 µg/ml)		19.3 ± 0.9 ^{aY}	24.7 ± 0.7 ^{bY}
Salak peel (10 µg/ml)		28.5 ± 3.1 ^{bZ}	14.7 ± 3.1 ^{aX}
Soursop peel	Lipid peroxidation (µmol MDA/mg linoleic acid)	0.21 ± 0.02 ^{aX}	0.65 ± 0.06 ^{bX}
Matoa peel		0.61 ± 0.06 ^{aZ}	0.97 ± 0.03 ^{bZ}
Salak peel		0.26 ± 0.01 ^{aY}	0.91 ± 0.04 ^{bY}
Soursop peel	Total phenolic (mg GAE/g sample)	2.0 ± 0.2 ^{aX}	14.2 ± 1.3 ^{bY}
Matoa peel		7.5 ± 0.7 ^{aY}	18.6 ± 1.9 ^{bZ}
Salak peel		6.9 ± 0.7 ^{aY}	5.9 ± 0.7 ^{aX}

Each value represents mean ± SD. Means with different small letters in a row are significantly different ($p < 0.05$) between the undigested and digested. Means with different big letters in a column are significantly different ($p < 0.05$) between the samples tested. DPPH radical scavenging activity (n=3), lipid peroxidation (n=6); total phenolic, Total phenolic (n= 4); MDA = Malondialdehyde; GAE = gallic acid equivalent.

The lipid peroxidation of salak peel and soursop peel in the undigested was the lowest concentration 0.21 and 0.26 µmol/mg linoleic acid, respectively. After *in vitro* digestion all fruit peels showed a significant increase in the lipid peroxidation and soursop peel (0.65 µmol MDA/mg linoleic acid) resulted the lowest concentration for lipid peroxidation compared matoa peel and salak peel. Dietary 10% pomace grape to chick that containing total phenolic (0.19 g GAE/100 g dry matter) resulted the lowest lipid peroxidation value 0.055 µg MDA/g meat after storage for 1 days compared control.

The salak peel exhibited a significant decrease ($P < 0.05$) in the total phenolic concentration: 6.9 – 5.9 mg GAE/ g sample after *in vitro* digestion. A similar result was found in papaya and araticum extract after digested (Pavan *et al.*, 2014). The TPC values after *in vitro* digestion showed that matoa peel (18.6 mg GAE/g sample) had the highest value of TPC followed by soursop peel and salak peel. Total phenolic contents were found increasing in ileal and excreta of chick after feeding 10% pomace grape (Chamorro *et al.*, 2015). The increasing of total phenolic after digested could be caused by releasing of polyphenols and changing their structural form which have affects their chemical and functional properties (Bhatt and Patel, 2013).

CONCLUSIONS

The result showed that the *in vitro* digestion process was made variation effects in test of total phenolic, lipid peroxidation and antioxidant activity in the fruit peels. Prior to *in vitro* digestion, salak peel was the most potent antioxidant containing fruit peel of all samples analysed. However, after *in vitro* digestion process the antioxidant activity in salak peel was reduced. Soursop peel and matoa

peel has a good result after *in vitro* digestion. This study concluded that soursop peel and matoa peel have high bioavailability of antioxidant activity and these could be used as feed additive for animals.

ACKNOWLEDGEMENT

This research was completed at Department of Applied Biological Science, Faculty of Agriculture, Kagawa University, Japan. And also it was granted by Japan Student Services Organization (JASSO) Scholarship.

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**EGG CHARACTERISTICS AND BLOOD PROFILES OF LAYER CHICKEN
CONSUMED VITAMIN E IN THE DIET ON HEAT STRESS.**

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ABSTRACT

Vitamin E is primary antioxidants to preventing lipid peroxidation damage. This study was conducted to determine egg characteristics and blood profiles of layer which consumed vitamin E in commercial ration. Eggs stored for 7 days. This study was using randomized assign design with 3 treatments and 10 replicates. The variables observed were egg characteristics (egg index, Haugh Unit (HU), egg weight, albumen weight, shell weight, shell thickness, egg number, follicle number, yolk color score, *malondialdehyde* (MDA), and blood profiles (hemoglobin, hematocrit, erythrocyte, leukocyte, and N/L ratio). This study was used 30 of 45 weeks old of layer chickens, Isa Brown strain. The treatments were M₁ = commercial diet and maintain at AC room, M₂ = commercial diet + 200 mg/kg and maintain under heat stress, M₃ = commercial diet and maintain in under heat stress. Result shows that 200 mg/kg vitamin E supplementation increasing HU on eggs stored for 7 days, and decreasing MDA value of egg. That also decreasing leukocyte and reduced ratio of N/L on the state of heat stress. A decrease in the characteristics of eggs and blood profiles due to heat stress can be overcome with vitamin E supplementation in feed.

Keywords: *vitamin E, egg characteristics, blood profile, heat stress*