TUBER PRODUCTION OF YAM BEAN (*Pachyrhizus* spp.) DUE TO SINK-REPRODUCTIVE PRUNING

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ABSTRACT

Yam bean (*Pachyrizhus*spp) can be used as new alternative food resource. To increase yam bean production including tuber quality traits, can be acquired troughintraspesific and interspesific crosses between *P. erosus* and *P. ahipa*. Simultaneous sink-reproductive pruning treatment is conducted to increased the tuber production, because the treatment can divert assimilate flow to tuber which consequently change the tuber dimensions.

Materials used in this experiment were 9 yam bean genotypes collection of A. Karuniawan (Plant Breeding Laboratory, Padjadjaran University). Nine genotypes consisted of three genotypes of *P. Erosus*, one genotype of *P. Ahipa*, two genotypes of *P. erosus* intraspecific hybrid and three genotypes resulted from interspecific between *P. erosus* and *P. ahipa*. The field trial was conducted at experimental field of Faculty of Agriculture, Padjadjaran University Jatinangor located at an altitude of 753 m above sea level with Inceptisols soil type and type C rainfall according to Schmidt-Fergusson. The experiment was conducted from August 2009 until March 2010.

The experiment was arranged in split plot design which was repeated twice. The main plot consisted of two treatments: without sink-reproductive pruning and with sink-reproductive pruning. The main plot was divided into nine subplots based on genotypes. Characters observed were tuber fresh weight (g), tuber dry matter (%), total starch content (% WB), and protein content of tuber (% WB).

Pruning affected the increasing of tuber fresh weight. Interspesific hybrid genotype AC 216-139 d x EC 550 produced the highest value for dry matter content. The highest starch and protein content contained on *P. ahipa* genotype AC 216-139 d. There was interaction between pruning and genotype on dry matter content, which the best result shown by AC 216-139 d with pruning treatment. The genotype which was good for food resource material with high starch and protein production was intraspecific crosses genotype x B-56/CJ EC 550.

Keywords: *yield, sink-reproductive pruning, yam bean.*

INTRODUCTION

Yam bean is a plant originated from Mexico, Central America and South America. This plant consists of five species, three of which have been cultivated and two of them are wild species. Cultivated species are *Pachyrhizuserosus*, *P. ahipa*, and *P. tuberosus*. Meanwhile, for the wild species are *P.panamensis* and *P. ferrugineus*[6]. Until currently, yam bean (*Pachyrhizus*spp) is categorized as one of the neglected and underutilized by the International Plant Genetic Resources Institute (IPGRI). In Indonesia, this plant is a minor crop that has not been a priority for development, meanwhile, it is widely cultivated in Sumatra, Java, Southeast Nusa, Sulawesi, Bali and Kalimantan.

The role of tuberous plant which is also a legume crop such as yam bean (*Pachyrizhus*spp) will be more strategic in food security, including Indonesia. Yam bean is a plant which produces both starch and proteins in quite high level. Thus, yam bean can be used as an alternative source of starch with high protein content [6]. This is related to the efforts in fulfill the alternative sources of protein-rich starch as food. FAO (1979) has given attention to the importance of the use of tuberous legume as well as starch and protein sources, especially in areas experiencing the deficiencies of starch and protein fulfillment.

Average productivity of yam bean tuber in Indonesia remains in low level. In the western part of Indonesia, the average production ranges from 10 ton ha^{-1}-70 ton ha^{-1} and in the eastern parts of Indonesia ranged from 10 ton 10 ton ha^{-1}-36 ton ha^{-1}[3]. While in Mexico, the yam bean productivity can reach 100 ton ha^{-1}-145 ton ha^{-1}. In the Philippines, productivity of yam bean about 80 ton ha^{-1}-90 ton ha^{-1} [3], and in Thailand's average productivity of yam 60 ton ha^{-1}-90 ton ha^{-1}[5]. From the above data, it is shown that Indonesia yam bean productivity is lower compared with other yam bean producer countries.

Yam bean has been recommended as an alternative flour production material[1]. In Indonesia yam bean has potential to be further developed as an alternative source of starch and high protein. This can be seen from the analysis of 100 g of fresh yam bean which has starch content at 2.1 g - 10.7 g and 1 g - 2.2 g protein. The lowest water content contained in *P. tuberosus* which is 70% -74%, followed by *P. ahipa* that is equal to 74% -76%, then *P. erosus* by 78% -94% [6]. Based on the assumption that the average results of 35 t ha-1, dry weight ranges from 6% - 22% per 100 g of fresh yam bean tuber, the starch content of 50% and protein 10% dry matter, the content of starch and protein produced by yam bean per hectare is 1.05 ton – 3.85 ton starch and 0.21 ton – 0.77 ton protein [4]. It makes yam bean as a potential source of starch and protein.

One disadvantage of starch yam bean tuber use is the high water content, so that although the tuber has great size but the starch and protein levels possibly will be low due to the low dry matter content. To broaden genetic diversity and increase the amount of dry matter, Plant Breeding Laboratory, Padjadjaran University in Bandung did intraspecific and interspecific hybridization between *P. erosus* and *P. ahipa*. All *Pachyrhizus* spp. species are diploid plants with chromosome number n = 11. The same chromosome number makes compatible crosses (no obstacle in the process of pollination).

Besides plant breeding, the potential of yam can be improved by developing appropriate cultivation techniques. One of the frequent cultivation techniques are sink-reproductive pruning [6]. Sink-reproductive pruning is the cutting of reproductive parts of plants, which is beginning at the first flower bud appearance. Sink-reproductive pruning aimed to reduce competition of photosynthates distribution between the sink-reproductive and tuber. Therefore, sink-reproductive pruning will shift assimilate to tuber. Flower bud pruning on the yam bean can increase 30%-70% of tuber fresh weight [4].

This study aimed to test the simultaneous effect of sink-reproductive pruning on tuber production, also study on starch and protein content of yam bean tuber. Further, expectantly there will be new genotypes with better tuber productions with high starch and protein contents.

MATERIAL AND METHODE

Materials used in this experiment were 9 yam bean genotypes collection of A. Karuniawan (Plant Breeding Laboratory, Padjadjaran University). Nine genotypes consisted of three genotypes of *P. Erosus*, one genotype of *P. Ahipa*, two genotypes of *P. erosus* intraspecific hybrid and three genotypes resulted from interspecific between *P. erosus* and *P. ahipa*.

The method used was experiment method. The field trial was conducted at experimental field of Faculty of Agriculture, Padjadjaran University Jatinangor located at an altitude of 753 m above sea level with Inceptisols soil type and type

C rainfall according to Schmidt-Fergusson. The experiment was conducted from August 2009 until March 2010.

The experiment was arranged in split plot design which was repeated twice. The main plot consisted of two treatments: without sink-reproductive pruning and with sink-reproductive pruning. The main plot was divided into nine subplots based on genotypes. Each subplot size was 2 m x 3 m with spacing 50 cm x 50 cm between plants and 100 cm between subplots. Fertilizer recommendations are given referring to the sweet potato cultivation in dry land.

Sink-reproductive application treatment was carried out after 50% of plants per plot had entered the flowering phase (R5) [6], it was done once a week until the harvest time. Harvesting of tuber was approximately 180 days (six months) after planting.

Characters observed were weight of tubers (g), tuber dry matter content (%), total starch content (% WB), and total protein content (%WB). Yam bean tuber starch content was analyzed using Luff Schoorl method and protein content was analyzed using Micro-Kjeldahl Method. Quality traits test was conducted at Yield Physiology Laboratory, Vegetable Research Institute, Lembang West Bandung.

Data were analyzed using statistical analysis of variance (anova). Comparison of the mean value was done using the least significant difference test (LSD) on the real level 5%.

RESULTS AND DISSCUSSION

1. Result

TABLE 1

ANALYSIS OF VARIANCE RESULT FOR TUBER FRESH WEIGHT, TUBER DRY MASS, TOTAL STARCH CONTENT AND TOTAL PROTEIN CONTENT OF NINE GENOTYPES YAM BEAN

Variable	F		
	Pruning	Genotype	Interaction PxG
Tuber fresh weight (g)	2742.35*	1.20	0.73
Tuber dry mass (%)	1.12	3.90*	9.43**
Total starch content (%)	0.12	17.02**	2.60
Total protein content (%)	128.27	3.99*	0.77

Explanation : * significant on alpha 0,05 Ftable F(0,05) F(0,01)
** significant on alpha 0,01 Pruning : 161 4062
Genotypes : 2.77 4.30
Interaction PxG : 2.77 4.30

Based on analysis of variance (Table 1), main factor, pruning treatment gave significant different. Genotypes as the subplot factor, gave significant different on all tuber quality traits observed. There was at least one genotype which gave different value on tuber dry mass and total starch content characters, in fact, total protein content showed highly significant result. Sink-reproductive pruning and genotypes interacted on tuber dry mass in highly significant difference.

TABLE 2
PRUNING EFFECT ON YAM BEAN TUBER FRESH WEIGHT.

Treatments	Tuber fresh weight g
Non pruning	122.48 a
Pruning	348.04 b

Exp: Numbers followed by the same letters are not significantly different on alpha 0.05 LSD test

Tabel 2 shows least significant difference (LSD) test on tuber fresh weight. It proved that sink-reproductive pruning had higher value. Non pruning treatment only resulted 122.48 g,whereas, sink-reproductive pruning 348.04 g.The ratio for this result was more than two times.

TABLE 3.
EFFECT OF GENOTYPES ON YAM BEAN TUBER DRY MASS, TOTAL STARCH CONTENT, TOTAL PROTEIN CONTENT

Genotip	Dry mass content		Total starch content		Total protein content	
	---%---					
EC 550	6.11	ab	1.07	A	0.94	a
EC 033	5.36	a	0.71	A	1.17	a
B-56 / CJ	7.33	cd	1.63	Ab	1.21	a
AC 216-139 d	8.86	fg	6.73	E	1.89	d
EC 550 x AC 216-139 d	7.50	de	0.87	A	1.01	a
EC 033 x B-56 / CJ	8.82	f	1.69	D	1.80	cd
AC 216-139 d x B-56 / CJ	6.60	bc	1.51	Bc	1.51	ab
EC 550 x B-56 / CJ	8.35	ef	1.74	Cd	1.56	b
EC 550 x AC 208-72h	10.27	g	2.66	De	1.72	bc

Exp: Numbers followed by the same letters are not significantly different on alpha 0.05 LSD test

Table 3 defines LSD test for effect of genotypes for dry mass content, total starch content and total protein content. Interspesific hybrid genotype between *P.*

erosus and *P. ahipa* (EC 550 x AC 208-72h) had the highest value on dry mass content. On total starch content character, the best result was showed by *P. ahipa* species, AC 216-139 d genotype, then followed by EC 550 x AC 208-72h genotype which had the highest on dry mass content character. Almost the whole hybrids intraspesific and interspesific genotypes were higher than Indonesian native yam bean, B-56 / CJ also another *P. erosus* genotypes EC 550 and EC 033 on total protein content.

TABLE 4

INTERACTION BETWEEN GENOTYPES x SINK-REPRODUCTIVE PRUNING ON TUBER DRY MASS CONTENT

Genotypes	Tuber dry mass	
	Non-pruning	Pruning
EC 550	8.51 B B	3.71 a A
EC 033	6.04 ab B	4.68 ab A
B-56 / CJ	7.65 B B	7.01 de A
AC 216-139 d	1.38 A A	16.34 h B
EC 550 x AC 216-139 d	8.78 B B	6.22 c A
EC 033 x B-56 / CJ	10.42 B B	7.23 e A
AC 216-139 d x B-56 / CJ	9.70 B B	3.50 a A
EC 550 x B-56 / CJ	8.34 B A	8.37 f A
EC 550 x AC 208-72h	11.71 B B	8.84 g A

Exp: Numbers followed by the same letters are not significantly different on alpha 0.05 LSD test

There was interaction between genotypes x sink-reproductive pruning on tuber dry mass content, it is shown on Table 4. On non pruning treatment, only *P. ahipa*AC 216-139 d which gave smallest mean value for tuber dry mass character. While, contrast result was shown on sink-reproductive pruning, AC 216-139 d genotype had the highest value than other genotypes, then followed by interspesific hybrid genotype EC 550 x AC 208-72h.

TABLE 5

YAM BEAN TUBER, DRY MATTER, STARCH AND PROTEIN PRODUCTION

Genotypes-pruning	Tuber	Dry matter	Starch (WB)	Protein (WB)
	----ton/ha---			
EC 550	73.43	2.72	0.46	0.41
EC 033	72.26	3.38	0.07	0.68
B-56 / CJ	88.70	6.21	1.53	0.78
AC 216-139 d	19.38	3.17	1.23	0.28
EC 550 x AC 216-139 d	61.86	3.85	0.93	0.59
EC 033 x B-56 / CJ	40.36	2.92	0.72	0.50
AC 216-139 d x B-56 / CJ	103.44	3.62	1.19	0.86
EC 550 x B-56 / CJ	97.53	8.16	2.45	1.36
EC 550 x AC 208-72h	69.53	6.14	2.01	0.96

WB = Wet Basic

Table 5 contains production of yam bean tuber for dry matter, starch and protein. To select the best yam bean genotype for industry material is based on yam bean production. These values were got from yield character on 200.000 plants density for one hectare area as mentioned in methodology.

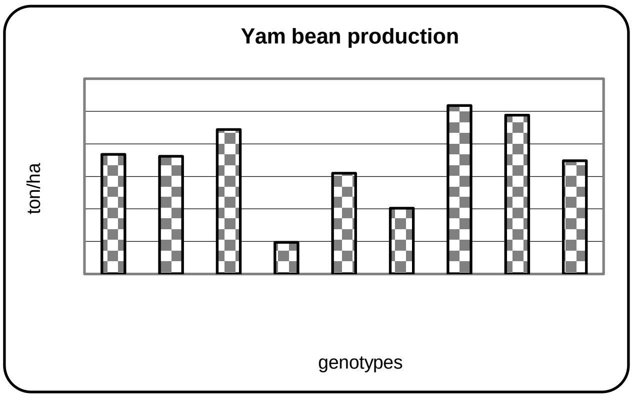

Fig. 1. Yam bean production on nine genotypes

In figure 1, the highest yam bean production was shown by interspesific hybrid genotype AC 216 d x B-56/CJ, then followed by intraspesifc hybrid genotype EC 550 x B-56/CJ. The smallest value was shown by *P. ahipa* genotype AC 216-139 d.

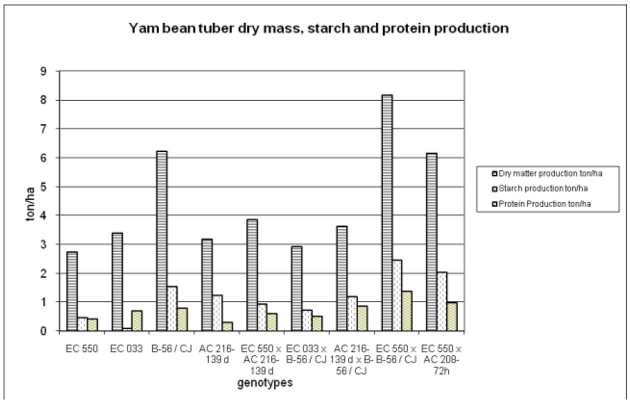

Fig. 2 Yam bean tuber dry mass, starch and protein content

On Figure 2, tuber production based on tuber quality aspects such dry matter, starch and protein production. The value was taken from the value characters multiplied by yam bean production per hectare. Therefore, the higest value was gained byintraspesifc hybrid genotype EC 550 x B-56/CJ, then followed by interspesific hybrid genotype AC 216 d x B-56/CJ. The smallest value was shown by *P. erosus*genotype EC 550 for dry matter production, EC 033 for starch production, and *P.ahipa* genotype AC 216-139 d for protein production.

2. Discussion

In this study, sink-reproductive pruning gave higher tuber fresh weight trait for whole genotypes. Sink-reproductive pruning of flower bud removal diverts assimilate distribution into tuber storage sinks (Ho, 1988). The increased flow of assimilate to the tuber, consequential on the change in dimensions. In addition, the increased of assimilate flow also affect tuber fresh weight.

Interspesific hybrid genotype between *P. erosus* and *P. ahipa* (EC 550 x AC 208-72h) had the highest value on dry mass content. On total starch content character, the utmost result was showed by *P. ahipa* species, AC 216-139 d genotype, then followed by EC 550 x AC 208-72h genotype which had the highest on dry mass content character. Almost the whole hybrids intraspesific and interspesific genotypes were higher than Indonesian native yam bean, B-56 / CJ also another *P. erosus* genotypes EC 550 and EC 033 on total protein content.

In this study, pruning did not affect the qualitative character of the percentage of dry matter, starch and protein content. Yam bean production increased as a result of pruning, this practice did not affect the percentage of soluble sugar content and dry matter percentage. But there are variations of these traits on genotypes were evaluated caused by genetic factors [7]. This variation can be seen in the character of dry matter, starch and protein content in this study.

P.ahipa has a dry matter contained 24% -26% dry matter, while *P. erosus* contained 6%-22% dry matter depend on environment [6]. In this study, *P. ahipa* genotype AC 216-139 d got a second on dry matter, the highest dry matter occupied by interspesific hybrid genotype EC 550 x AC 208-72h, 10.27% dry matter content, and contrary with the elder EC 550 which had only 6.11% of dry matter. Therefore, the cross was succeed to improve the dry matter character.

P.erosus contains 2.1% -10.7% starch [6]. Starch content of *P. ahipa* generally higher than *P. erosus*[2]. In the present study showed that the highest starch content contained in *P. ahipa* genotype AC 216-139 d is 6.73%, followed by hybrid genotype interspesific AC 216-139d x EC 550, 2.66%. Whereas other genotypes had starch content started from 0.71% - 1.69%. This also showed that the crossing able to alter the character of starch.

*P. erosus*contained higher protein than *P.ahipa*, both of which were planted in Benin[2]. In this study, the protein content of hybridization genotypes generally was higher than that of yam genotypes of *P. erosus* but not to *P. Ahipa*. This was likely influenced by different planting conditions, so that gene expression on protein characters was different with statement above [2]. The highest protein content was gained by *P. ahipa*, then followed by EC 033 x B-56 / CJ and AC 216-139d x EC 550. The smallest value was attained by EC 550.

P. erosus is the most stable species in production, also it produces the prime number for yield [2]. *P. ahipa* is known as species with higher dry matter and starch content but not for protein content. Therefore, hybridization using those species is expected to gain the new genotype in high production amount with better quality. This study noted that intraspesific and interpesific crosses produced a new genotype with higher production amount with better quality character for dry matter, starch and protein than the elders.

CONCLUSION

Pruning affected on tuber fresh weight for all genotypes observed. Interspesific hybrid genotype AC 216-139 d x EC 550 produced the highest value for dry matter content. The highest starch and protein content contained on *P. ahipa* genotype AC 216-139 d. There was interaction between pruning and genotype on dry matter content, which the best result shown by AC 216-139 d with pruning treatment.

The main material used food resource is starch with high protein content. Therefore, characters which become the basis of determining best genotype must be balanced between the yield and qualitative character. Thus, although the genotype AC 216-139 d has the highest starch and protein content, but not it was supported by yield character.

The genotype which was good for a alternative foodstuffs had to have high starch and protein suppoted by high tuber production. Therefore, it can be concluded that the genotype of yam bean EC 550 x B-56/CJ which get sink - reproductive pruning treatment is the best genotypes as new food alternatives resources.

REFERENCES

deMelo, E.P., N. Krieger, and T.L.M. Stamford. *Physchochemical properties of Jacatupe (PachyrhizuserosusL. Urban) starch*. *Starch 46: 245 247,* 1994.

Kale. *Studies on Nutritional and Processing Properties of Storage Roots of Different Yam Bean*(*Pachyrhizusspp*) and Wild Mung Bean (*Vignavexillata*) Species. (Disertation) CuvillierVerlagGoettingen, Universitaet of Goettingen, Germany. 2006.

Karuniawan, *A. Cultivation Status and Genetic Diversity of Yam Bean (Pachyrhizuserosus) in Indonesia*. (Disertation) CuvillierVerlagGoettingen, Universitaet of Goettingen,Germany. 2004.

Nusifera, Sdan A. Karuniawan. *Stability of 16 Genotypes Yam Bean (Pachyrhizus erosus L. Urban) Tuber Dry Matter Content Stabilitas Kadar in Jatinangor West Java Based AMMIModel*. *Zuriat,* Vol. 18 No. 1, 2007.

Ratanadilok, N., K. Suriyawan, and S. Thanaisawanrayangkura. 1998. *Yam bean (PachyrhizuserosusL. Urban) and its economic potential*. *In* :Sørensen, M., J.E. Estrella, O.J. Hamann and S.A. Rios Ruiz (Eds.). Proceedings of the 2nd International Symposium on Tuberous Legumes, Celaya, Guanajuato, Mexico 5 – 8 August 1996. Copenhagen, Denmark.

Sørensen, M. *Yam Bean Pachyrhizus DC.Promoting the Conservation and Use of Underutilized and Neglected Crops*.IPGRI.Rome, 1996.

Zanklan, A.S. *Agronomic Performance and Genetic Diversity of The Root Crop Yam Bean (Pachyrhizus Spp.) under West African Conditions*. (Dissertation).CuvillierVerlag Gottingen, Uniersitaet of Goettingen Germany, 2003.

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THE EFFECT OF MILK FERMENTED WITH PROBIOTIC BACTERIA SUPPLEMENTATION ON HAEMATOLOGIC CONDITION OF BROILER

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ABSTRACT

The effect of probiotic supplementation in fermened milk on broiler hematologic condition, (the number of erythrocyte, hemoglobin and hematocryte value),used 100 broilers, and used Completely Randomized Design, with four treatments:R0=as control, R1= 0,5% probiotic, R2 = 1,25% probiotic, R3= 2,00% probiotic, which was five times repeated. The resultsshowed that theprobiotic supplementation up to1,25 percent, had no significantcy(p>0.05) on hematologic condition, but 2 percent probiotic supplementation has significantly increased the hematologic condition.

Key Words :*Probiotic, Haematologic, Erythrocyte, Haemoglobine, Haematocrite Value*

INTRODUCTION

Probiotic is classically defined as a microbial dietary supplement that beneficially affect the host through its effects in the intestinal tract . This definition however, was initially intended for use with animal feed. Probiotic microorganisms that have a favourable influence on physiological processes of the host by their effect on the intestinal flora may play a role in improving human health(Ericson and Nail, 2000). Erythrocytes contained hemoglobin that carries oxygen from the lungs and will be released into the tissue, and also binding carbon dioxide in tissue that will be brought to the organ secretions. Hemoglobine functions such as respiration and blood pigment as a buffer in the blood system, which is closely related to the ability of blood to carry oxygen. Non-pathogenic microbes such as *Lactobacillus bulgaricus*and *Streptococcus thermophillus* are capable of producing lactic acid and some amino acids and vitamins produced by microbes, and also as a precursor for the formation hemoglobin.

Microbes that classified as lactic acid bacteria (LAB) have a high microbial activity due to the resulting product will inhibit the growth of pathogenic bacteria that can damaged the cell membrane permiability and ended with the destruction of the cell wall, resulting in the release of hemoglobin from the cell. Fe has a large influence on the formation of blood hemoglobin. Fermented milk contains complete mineral,and will absorbed in the small intestine,and because has a low molecular weight, the compounds are soluble in water. Calcium and phosphorus are essential for the growth of bones and teeth, while iron in building muscle, skin and eggs of red blood cells, stimulates nerves, maintaining muscle elasticity and maintain osmotic pressure (Surono, 2004). Organic acids produced by lactic acid bacteria, mainly lactic acid and acetic acid to help stimulate activities of gastric the rate of passage become slower, and cause increased absorption of nutrients as a result of metabolic processes in the body and substance formation of red blood cells and hemoglobin will increase.

Oxidation reactions can occur in body cells and damage the body, especially the cell membrane. One of the important body of the cell membrane is the erythrocyte membrane. Oxidation reaction on erythrocyte membranes will result in damaging erythrocytes, will eventually lower the body resistance and response to disease. Erythrocyte damage would cause metabolic disorders at least will affect the productivity of livestock. The damage can be inhibited by the ability of lactic acid bacteria which can suppress the growth of various gram-positive bacteria and gram negative. Suppression process is influenced by the production of hydrogen peroxide inhibits the growth of pathogenic bacteria through the powerful influence of oxide on bacterial cell or through the destruction of the basic molecular structure of nucleic acids and proteins of cells and production of special proteins called bacteriocin.

Previous studies in rabbit which are given yoghurt containing *Lactobacillus bulgaricus and Streptococcus thermophillus* 2.00% of body weight, can increase the number of erythrocytes, hemoglobin, and hematocrit values significantly. Studies conducted on rats using of yoghurt containing *Lactobacillus bulgaricus, Streptococcus thermophillus, Lactobacillus acidophilus, and*

Bifidobacterium of 1.25% and 2.00% of body weight can increase the number of erythrocytes, hemoglobin and hematocrit values.

MATERIALS AND METHODS

Bacteria Strain: Two bacteria used in this research, are *Lactobacillus bulgaricus*and *Streptococcus thermophilus* as yoghurt culture starter.

Animal : one hundred broiler cp 707, day old chick, the treatment given from day old chicks until six weeks.

Experimental Design, broiler were randomly devided into 4t reatments groups with 5 replications. The treatments consisted of probiotic yoghurt, R0=control , R1= control + 0,5% probiotic, R2 = control + 1,25% probiotic, R3= control + 2,00% probiotic

Data were analysed in a Completely Randomized Design (CRD). Further analysis for significantly was conducted using Duncan Test, the parameter observed were : the number of erythrocyte, hemoglobin, and hematocrit value.

RESULT AND DISCUSSIONS

Table 1, The results of the effect of treatment with addition of probiotic (Lactobacillus bulgaricus, Streptococcus thermophillus) on the number of erythrocyte, hemoglobin, and haematocrit.

Tabel 1. Effect of the treatment on total erythrocyte,haemoglobine, hematocryte value

Treatment	Erythrocyte	Hemoglobine	haematocryte
	(x 10^6) alls/mm^3	g/ml	%
Ro	2,27	8,10	31,4
R1	2,32	8,38	31,6
R2	2,88	9,01	32,4
R3	3,04	9,22	35,5

Notes :

R0 : control

R1 : control +0,5% from body weight

R2 : control +1,25% from body weight

R3 : control +2,00% from body weight

Based on Table 1, R3 treatment has total erythrocyte, significantly higher compared with the treatment R0 and R1, while R2 and R3 has not significancy. The highest hemoglobin levels achieved in R3, that significantly higher than R0 and R1, while R2 and R3 showed no significancy. The highest hematocrit value achieved by the R3 treatment was significantly; compared with the treatment of R0, R1 and R2.

Lactobacillus produce metabolite on increased the size of new cells,and will influence the body weight gain, and also improved metabolism*Lactobacillus bulgaricus* and *Streptococcusthermophillus, Lactobacillus Acidophilus*it can increase the elasticitythe cellular membranes thereby improving cell membranes, which in turn will improved the ability of erythrocytes to maintain the integrity of the membranes.

According, to 1997 Lovita, 2005,the organic acids produced by lactic acid bacteria such as lactic acid and acetic acid can protect the cell membranes and damaged other subcellular oxidation reaction by the peroxide bond.

Lactic acid bacteria which can increase the elasticity of cell membranes so that it will produce better cell membrane, which in turn will improve the ability in maintaining erythrocyte membrane integrity (Ganong, 1985). Other factors that affect their ability to both bacteria, will survives in the lower acid to base environment (Fuller, 1992). The addition of yoghurt are still within the criteria of normal erythrocytes. Acetic acid , some amino acids and vitamins produced by microbes, is a precursor to the formation of hemoglobin.

Acetic acid is inferred that changing in the cycle become keto glutarat kreb-alpha acid, and then binded the ketoglutarat two-alpha acid with one molecule of glycine to form pyrole compounds. The next four pyrole compounds to form compounds protoporfirin. One compound, known as n protoporfirin III, when binded with iron will form a molecule hem. Finally, four molecules bind one end of the globin molecule, the formation of hemoglobine, which in turn will increased the amount of hemoglobin (Guyton, 1985).

According to Swenson (1970), increasing in hemoglobin and erythrocyte number will be in line with the increase in hematocrit value because there is a positive relationship between the three components.The overall results indicated that administration of probiotic (Lactobacillus bulgaricus and Streptococcus thermophilus) on the dose up to 2% of body weight, will increase the amount of value erythrocyte, hemoglobin and hematocrit, still in the normal range.

REFERENCES

Aseltin, M.S. 1992. *Biotechnology in the Feed Industry*. Fifth Edition. Oxford and I B H Publishing Co., New Rank Test Delhi. Bombay-Calcuta. 20-30.

Baldy, C.M. 1995. *Phatology Fisiologi*. PenerbitBuku Kedokteran. Jakarta. 224-231.

Bottazzi, V. 1983.*Other Fermented Dairy Products In Biotechnology A Comprehensive Treatise in 8 Volumes. Food and Feed Production With Microorganisms*. Editor H.J. Rehm and G. Reed.Volume 5.VerlagChemie. Florida. Basel. 317-360.

Brown, E.M. 1989. *HistologiVeteriner*. Edisi ke-3.Penerbit UI Press.Jakartt. 108-114

Surono, Inggrid S. 2004. *Probiotik Susu Fermentasi dan Kesehatan*. YAPMMI. Jakarta

Guyton, A.C. 1961. *Medical Physiology*. 2nd Edition. WB Sanders Company. Philadelphia. **Review of Textbook of Medical Physiology**, April 21,2006 by Darrell Wu (Chicago, IL United States)

Lovita, A. 2005. *Bakteri Probiotik Sebagai Starter Dan Implikasi Efeknya Terhadap Kualitas Yoghurt, Ekosistem Saluran Pencernaan Dan Biokimia Darah Mencit*. Disertasi Program Pascasarjana Universitas Padjadjaran. Bandung.

Swenson, M.J., 1977 Dukes Physiology Of Domestic Animal. 10 thEd. Comstock

Publishing Associate a Division of Cornell University Press. Ithechaand Londonhttp://www.vscht.cz/kch/galerie/lact.htm#Lactdelb. Diakses tanggal 25 Februari 2008.

THE EFFECT OF CLOVE OIL DOSAGE ON POPULATION OF *Callosobruchus maculatus* F., Seed VIABILITY AND VIGOR OF TWO SOYBEAN CV AFTER THREE MONTHS STORAGE DURATION[1]

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ABSTRACT

The objective of the experiment was to study the effect of clove oil dosage on controlling storage weevil *C maculatus* F., and maintaining seed quality including seed viability and seed vigor of two soybean cv after three months storage duration. The experiment was conducted in Laboratory of Seed Technology ,Faculty of Agriculture Padjadjaran University, Jatinangor, Sumedang, West Java, from March until May 2009.

The experiment design used was Split Plot Design consist of two factors and replicated three times. The main factor was cultivar of soybean, were Anjasmoro (big size seeds) and Sindoro (medium size seeds). The sub factor was bio protectant of clove oil dosage, which consist of five levels, were : without clove oil, 0.5 mL kg^{-1} soybean seed, 1.0 mL kg^{-1} , 1.5 mL kg^{-1} soybean seed, and 2.0 mL kg^{-1} soybean seed. All of the experiment consist of 45 units experiment , where every experiment unit used 75 g of seed. Seeds and five pairs of storage weevil were fill in plastic cup and placed in plastic container during three months storage.

The result of experiment showed that there were interaction effect between cultivars and dosage of clove oil on seed damage percentage after three months storage, but there were no intercation effect on another parameters was observed. Sindoro has better storability than Anjasmoro, as indicated by weevil population, seed damage percentage, germination capacity, vigor index and seed moisture content. Dosage of clove oil 5 mL/kg soybean seed showed better affected on suppression of storage weevil and maintaning seed viabilty and vigor.

Key words : Clove oil, *C maculatus*, Seed viability and Seed Vigor.

INTRODUCTION

Soybean is one of the third important food crop in Indonesia after rice and corn. Soybean seed contain approimately 400 g kg $^{-1}$ protein and 200 g kg^{-1} oil (Fehr,1987 cited by Egli and Crafts- Brandener, 1996) and is used as a source of edible vegetable oil, forage enrichment , and many traditional food product. Most of Indonesian feeds of traditional food product made from soybean seeds.

Soybean needs seriously handling for full fil domestic consumption, because has economic strategic value. Demand of soybean grain showed indicated increasing paralel with population growth . Until now domestic production still lower than national demand. To full fill domestic demand, Indonesia government policy imported soybean grain until 750 metric ton – 1 million ton per year from several country (BPS, 2010).

One of the effort to full fill domestic demand for increasing soybean yield productivity used was high quality seeds following by cultivated technology application and extensification cultivated area. Since 90's decade Indonesian Agriculture Department has strategic issue to support food strengthening program particularly for increasing used high quality seeds. Which government policy recognized with the six of seed program to solved the problem in seed quality stock during planting time. The six of seed program government policy include the approriate number of high seed quality , appropriate seed quality, appropriate specific variety , appropriate planting time, approriate agroclimat zone ,and approapriate price properly. But , until now a lot of farmer used non certified seed, because in several region has gap between planting time with stock of seed. Other reason , farmer considered seed price still expensive.

Seed storage is one of solution to solve the problem lack of high quality stock during planting time, usually during dry season. But some of weakness of soy bean seed is has short longevity in bad storage system . Soybean food reserve are rich of protein and lipids could generate fast to deteriorate if seed placed in bad storage . To avoid this problem needs appropriate storage technology , particularly in pest storage cotrol without decreased seed viability or vigor.

Seed deterioration during storage duration affected by seed properties characters and environment storage condition include biotic and abiotic factors. Seed properties were seed coat characteristic, food reserve chemical composition, seed viability and vigor , moisture content , and genetics (ortodox or recalsitrant) . Abiotic factor were temperature, relative humidity and light intensity during storage, while biotics factor affected seed viability during storage is insect and fungi (Šimic *et al* ., 2006 ; Copeland and McDonald,2004 ; Justice and Bass,2002) .

One of the alternative way to maintaning seed viablity and vigor during storage is to controlling growth of pest storage. Seed moisture content adjustment could effective to protect pest and microorganism invasion. Moisture content 8-9 % inhibit insect and microorganism invasion, while moisture content between 12 % - 18 % not safe for seed storage , seed is easy invasion by insect and fungi (Imdad, 1999). Moisture content safe for storage of food crop storage and horticulture is 9 – 12 % (Direktorat Jendral Tanaman Pangan dan Hortikultura, Direktorat Perbenihan, 2000 ; Interntional Seed Testing Association, 2003) .

In tropical and sub tropical region were three species of *Callosobrochus i.e. C maculatus* F. is one of the storage pest usually invasion soybean seed during storage, *C chinensis* invasion mung bean seed and *C analis* (F) invasion tunggak bean (Anwary *et al*., 2004). Imago active in day with life cycle between 21-30 days average at room temperature 30 ^{o} C and Relative Humidity 80 -85 % (Kalshoven, 1981). Primary food reserve was destroyed by *C maculatus* caused significant decreasing of seed viability and vigor capacity due to germination inhibition and produce abnormal seedling. Pest population development can be inhibit by sintetic protectant, but had negative impact on environment safety. One of effort to solve this problem used natural protectant or bio protectant made from clove oil. Clove oil is volatile oil contain 70-85 % eugenol , which oil vapour have long period aromatic (Guenther, 1990). Kardinan statement (2000) eugenol from clove oil was effective to control *Stegobium paniceum.* The importance of bio protectant not only capable to control pest storage , but able to maintanance seed viability and vigor.

Several experiment result showed proved bio protectant made from clove oil can to supressing pest storage. Sri Dewi Kartika *et al* (2006) and Sumadi *et al* (2009) proven clove oil 2.5 mL kg $^{-1}$ good effect to controlling *Sitophillus zeamais* and maintaining corn seed viability until three months storage. Other experiment study about the effect of clove oil on soybean seed Wilis cultivar , showed 1.5 mL kg^{-1} seed had good effect. Rika Meilasari (2000) proven was used clove oil 1 mL 100^{-1} g seed capable to supress *C* *maculatus* develepment without decreasing mung bean seed viability after 4 months storage period.

Negative effect on seed viability can occurred if use higher concentration of clove oil. Seed storage experiment by Rita Enggreni (2000) showed that clove oil 5 mL kg^{-1} seed were reduced soy bean seed viability until 50 % compare with control treatment (without clove oil) after two months storage. How ever storage insect population can reduce until 0 %, while at control treatment insect population increase until 40 % . Other experiment about negative effect used higher dosage clove oil 20 mL kg^{-1} sorghum seeds caused severe seed viability after three months storage (Julia Wingantini, 2005). Also, experiment by Sri Dewi Kartika *et al* (2006) showed that > 5 mL kg^{-1} with direct application caused corn seed deterioration .

Seed response to protectant factor depend on dosage and specific characteristic of seed coat. Seed with thicker seed coat more resist than seed with thinner seed coat. Each soy bean variety have specific chemical composition , seed coat physical properties , seed size and seed coat colour (Tekrony *et al*., 1987). Variation of seed size depend on cultivar. In Indonesia , soybean seed size classification consist of three categories based on 100 grains weight , ie . small size (6-10 g), medium size (10 – 12 g) and large size (> 13 g). According to cultivar description , Anjasmoro as large size and Sindoro as medium size categories. Seed coat of larger seed size thinner and more permeable than the small one (Monorahardjo *et al* , 1993 cited by Sumadi , 1997) . While seed large size have better vigor (Tekrony *et al* .,1987).

According to explanation some information coud be conclude that the effect of clove oil for controlling storage weevil and maintaining seed viability and vigor after three months storage duration depend on clove oil dosage and soybean seed cultivar . While, soybean seed viability and vigor could be maintanance after three months storage if application by appropriate dosage of clove oil. So, the result of experiment could be done to completed information about seed bank strategic.

MATERIAL AND METHODS

The experiment was conducted at Seed Technology Laboratory, Faculty of Agriculture, except for determine protein content at Chemical Analysis Laboratory , Faculty of Mathemetics and Science, Padjadjaran University , Jatinangor , since March 2009 until May 2009.

Two seed cultivar of *Glycine max* L. were used, one from the Balitbiogen and the other from IPB (Bogor Agriculture Institute) , Clove oil were use from P.T. Indrasari Jakarta which contain 70 -85 % eugenol. *C maculatus* imago obtain from SEAMEO , Bogor, and *kertas merang* for germination substrat, plastics sheet with size 30 x 20 cm.

The experiment was arranged in a split plot design with three replications. Two levels of soybean cultivar i.e Anjasmoro (large size seed) and Sindoro (medium size seed) were place in the main plot and five levels of clove oil dosage i.e no clove oil application as control , 0.5 mL kg $^{-1}$ seed , 1.0 mL kg $^{-1}$ seed , 1.5 mL kg $^{-1}$ seed , and 2.0 mL kg $^{-1}$ seed as sub plot. Data were analyzed statistically and mean were separated by DMRT.

All of the experiment consist of 2 x 5 x 3 x3 packet. Each a packet of used 75 g seed was fill in plastic cup and covered by aluminium foil , and then were placed in each container and store under room temperature for one, two and three months respectively. Before storage, seed protect by clove oil with indirect application use small cotton and place at site corner of plastic cup (Sumadi, 2006) and were invested five pairs of *C maculatus* in plastic cup.

One week prior storage were tested seed quality .ie. germination percentage, seed moisture content , vigor index , 100 grains seed weight, and protein content . Each month after storage seed quality was measured. Parameter will be measured after storage include pest population, seed damage by insect, moisture content,weight of 100 seeds , germination rate, electric conductivity value and vigor index.

Seed analysis . Seed quality testing based on ISTA procedure with some modification :

a. Seed moisture content measured by basic or oven methods which seeds drying at 130 ^{o}C during 50 menits . Five grams seed weighing (Fresh weght), followed by drying used oven at 130 ^{o} C during 50 menits and weighing with electrical balance (dry weight). Seed moisture content = FW – DW /FW x 100 %. Each treatment replicated three times.

b. Germination rate was tested by rolled paper test and placed in germinator cabinet. Seedling measured at First Day Count (FDC) at 5th and Last Day Count (LDC) at 8th . Calculation and expression of result are expressed as percentage by Sum of normal seedling divide seed number was tested by 100 %.

c. Seed vigor were determined by vigor index and electric conductivity value.

   IV = G5/D5 + ...G8/D8. Vigor Index showed that speed of germination, which value close to maximum (10) as good quality.

RESULT AND DISCUSSION

The physical and physiological state of seeds as internal factor was greatly influence their life span or seeds storability (Copeland and McDonald, 2004). Seeds that have been broken, cracked, or even bruised deteriorate more rapidly than undamage seeds (McDonald, 1985 and Prietley, 1986). Other seed physical properties affected rapid of deterioration was moisture content and seed size. Even without physical symptoms, seed may be physiologically impaired and become susceptible to rapid deterioration. For instance , seed vigor state before storage affected deterioration rate.

The means of seed quality including moisture content, germination rate or germination capacity, and vigor before storage presented in Table 1. According to result of prior test seed quality, Anjasmoro and Sindoro cultivar seeds have good physical and physiological standard. Prior of storage, the moisture content , germination capacity, and vigor index both of cultivars seed as good quality based on certitification standard. Harrington (1972) cited by Copeland and McDonald (2004) have recomendation which moisture content for long duration storage ortodox seed does not above 14 or below 5 %. Seeds store at moisture content above 14% begin to exhibit increased respiration, heating and fungal invasion that destroy seed viability more rapidly , while below 5 % cause seed membran structure hasten seed deterioration.

Table 1. Mean of Seed Quality before Storage

Seed Properties Characters	**Anjasmoro**	**Sindoro**
Moisture Content (%)	9.62	8.5
Weight of 100 seeds (g)	13.7	8.73
Germination Rate (%)	96.0	98.0
Value of Vigor Index	9.6	9.68
Protein Content (%)	40.31	39.30

Note : maximum value of Vigor index in this case is 10

Protein content as one of the internal factor were influence seed storability. One of protein properties is hydrophilic to vapour water arround of seed. Where soybean seed as one of the legume family with rich protein content usually have short storage longevity. Exception under well storage control , while sealed storage methods at low temperature and low relative humidity able to maintain seed quality.

The analysis of variance result showed that no significant effect of interaction between soybean cultivar and clove oil dosage on weevil population, weight of 100 seeds and moisture content after three months storage. Both of treatment between cultivar and clove oil dosage showed that was independent effect respectively (Table 2).

Table 2. Weevil Population, and 100 grains weight , and moisture content after three

months storage duration

Treatment	**Weevil Population**	**Weight of 100 seeds (g)**	**Moisture Content (%)**

Soybean Cultivar			
Anjasmoro	0.33 b	14.40 b	10.36 a
Sindoro	0.00 a	8.91 a	10.66 a
Clove oil dosage			
m_0 (0.0 mL kg^{-1})	0.33 b	11.50 a	10.66 a
m_1 (0.5 mL kg^{-1})	0.00 a	11.61 a	9.62 a
m_2 (1.0 mL kg^{-1})	0.00 a	11.40 a	10.23 a
m_3 (1.5 mL kg^{-1})	0.00 a	12.28 a	11.00 b
m_4 (2.0 mL kg^{-1})	0.00 a	11.49 a	11.05 b

After storage weevil population indicated decrease compare when fisrt invested before storage. Even some of them all of weevil was invested are died. Probably died of weevil in all of treatment are stress in the new place. Anjasmoro seed size bigger than Sindoro seed size .Seed size significantly affected on weevil population , which *C maculatus* like invasion the big one. Development of weevil at place contain the bigger seed size like Anjasmoro cultivar were condusive. The bigger size showed that more succeptable to *C maculatus* invasion than the small one. How ever no significant affected on moisture content , although increasing moisture content was occurred compare before storage. After three months storage duration moisture content both of seed was increase between 0.74 % - 2.16 % . Although,increasing of seed moisture content still under limited for safe storage.

Seed damage was affected by cultivar and clove oil dosage together. Infleunce of cultivar character particularly seed coat thickness depend on clove oil dosage (Table 3). Increasing of clove oil application showed supressing of pest invasion on seed both cultivar.

Table 3. Seed damage percentage

Kultivar	Seed damage percentage (%)				
	m_0	m_1	m_2	m_3	m_4
Anjasmoro	4.00 b	2.67 b	0.00 a	0.67 a	0.00 a
	D	C	A	B	A
Sindoro	2.00 a	0.00 a	0.00 a	0.00 a	0.00 a
	B	A	A	A	A

Effect of clove oil dosage on seed damage percentage by weevil depend on cultivar properties. While size of Anjasmoro seed bigger than Sindoro was suspected have thinner seed coat than the small one .Sukarman dan Raharjo (2000) were report their experiment soybean seed with small size have well storability than seed with medium size. Performance indicator as implication of cultivar properties and clove oil application showed in physiological character (Table 4).

Table 4. Seed Germination Rate, Vigor Idex and Electtic Conductivity

Treatment	Seed Germination (%)	Vigor Idex	Electric Conductivity (μ S g^{-1})
Soybean Cultivar			
Anjasmoro	87.07 a	8.55 a	3.35 a
Sindoro	96.67 b	9.57 b	5.15 b

Clove oil dosage			
m 0 (0.0 mL kg-1)	92.00 ab	9.03 b	4.32 bc
m 1 (0.5 mL kg-1)	88.00 a	8.71 a	3.53 a
m 2 (1.0 mL kg-1)	90.00 ab	8.82 ab	4.07 b
m 3 (1.5 mL kg-1)	93.33 b	9.24 bc	4.56 cd
m 4 (2.0 mL kg-1)	96.00 b	9.50 c	4.78 d

Influence on seed physiological of cultivar character and clove oil are independent. According to value of germination rate and vigor index , sindoro have storability potential better than Anjasmoro.How ever both of physiological character still good quality, germination rate more than 80 % as ISTA standard for certified seed. Also, vigor index is high category . In this case some inconsitency result showed EC value. Good seed have membrane integrity better than bad seed quality with low value , while EC Sindoro seed higher than Anjasmoro. Integrity membrane can determine by measure of electrolite leakage after soaking (Viera *et al,* 2001 ; Copeland and MacDonald, 2004). EC value was measured by EC meter, value of EC indicate degree of membrane destroyed as high of fatty acid content (Tatipata *et al*, 2004 : Tatipata, 2010).

Influence of clove oil on germination rate, vigor index and EC depend on dosage. Which dosage increasing until 1.5 mL kg^{-1} gave germination rate and vigor was increased compare with the lower one , but no significant increase of germination rate and vigor index if dosage add until 2.0 mL kg^{-1} . Like cultivar character, some inconsistent result showed at EC data was observed. Even EC data inconsistency , but all of value from seed testing showed that seed quality still high after storage. This indicate application of clove oil as bio protectant until 2 mL kg^{-1} able to control pest invasion and maintain seed viability and vigor.

Several experiment used clove oil was proved by Rika Meilasari (2000) on control of *C maculatus* development without decreasing mung bean seed viability after 4 months storage period, Sri Dewi Kartika *et al* (2006) and Sumadi *et al* (2009) on control of *Sitophillus zeamais* and maintaining corn seed viability until three months storage. Other result experiment report by Zainal *et al* (2010) that is clove oil abble to inhibit growth of *Clavibacter michiganensis* on tomato seed.

CONCLUSSION

Interaction between soybean cultivar and clove oil dosage had significant effect on seed damage, while there were no siginificant effect on another variable that were observed. Sindoro has better storability than Anjasmoro seed, it has indicated by weevil population, seed damage percentage, germination rate, vigor index and seed moisture content. The clove oil 1.5 mL kg^{-1} soybean seed indicated better influence on decreased weevil population, seed damage percentage, maintaining germination rate of soy bean seed, and vigor index.

According conclussion result was sugested following to study about the effect of weevil number, kind of packaging material, and longer storage duration under variation room condition .

REFFERENCES

Anwari, M., Rudi Iswanto, Y. Afriana dan N .Rachmasari. 2004. Evaluasi ketahanan beberapa genotipe kacang hijau terhadap hama gudang *Callosobrochus chinensis.* Prosiding Seminar Kinerja Penelitian Mendukung Agribisnis kacanag-kacangan dan Umbi-umbian. Puslitbangtan. Bogor. 220-225

Bewley.,J.D .,and M.Black. 1982. Seed Physiology and Biochemistry. In relation to germination.
Vol 2. Springer-Verlag. Berlin. Heidelberg, New York.

Copeland, L.O and M.B. McDonald. 2004. Principles of Seed Science and Technology. 4 th
edition. Kluwer Academic Publishers. Boston,Dordrecht, London.

Locher, R and P. Bucheli, 1998. Comapration of Soluble Sugar Degradation in Soy bean Under
Simulated Tropical Storage Conditions. Crop.Sci. 38 : 1229-1235

Meilasari, R. 2000. Penggunaan protektan botanis untuk mengendalikan hama gudang
Callosobrohus maculatus F dan mempertahankan viabilitas benih kacang hijau
selama penyimapanan. Skripsi Jurusan BDP . Faperta IPB. Bogor (tidak
dipblikasikan).

Purwanti**,** S. 2004. Kajian suhu ruang simpan terhadap kualitas benih kedelaia hitam dan kuning.
Ilmu Pertanian Vol. 11 No.1, 2004 : 22-31

Šimic, B., A. Sudaric, I. Liovic, I. Kalinovic, V. Rozman and J. Cosic. 2007. Influence of storage
condition on seed quality of maize, soybean and sunflower.

Sukarman dan M. Rahardjo. 2000. Karakter Fisik, Kimia dan Fisiologis Benih Beberapa
Varietas Kedelai. *Buletin Plasma Nutfah* 6 (2) : 31-36.
.

Sumadi, 1997. Toleransi kultivar kedelai dengan kapasitas wadah yang berbeda terhadap cekaman air selama periode pembentukan polong sampai pengisian biji. Laporan penelitian. Universitas Padjadjaran (tidak dipublikasikan).

______, Anne Nuraini dan Sri patmawati. 2008. Pengaruh kombinasi kadar air awal benih dan dosis minyak cengkeh dalam mengendalikan hama *Callosobrochus maculatus*. F untuk memperptahankan Viabilitas dan Vigor benih kedelai (*Glycine max* L) selama periode simpan tiga bulan. Prosiding Seminar Inovasi Teknologi. Balitkabi. Departemen Pertanian. Malang. Hal 411-417

______, Anne Nuraini dan Ivan Rendi Mustofa. 2008. Pengaruh dosis minyak cengkeh terhadap viabilitas dan vigor benih jagung (*Zea mays* L) pada berbagai investasi hama *Sitophillus zeamais* motsch setelah penyimpanan. Prosiding Seminar Nasional dan Workshop Perbenihan dan Kelembagaan. Faperta UPN- Forum Perbenihan DIY. Yogyakarta. Hal III.34-40

Tatipata, A., Prapto Yudono, Aziz P dan Woerjono. 2004. Kajian Aspek Fisiologi dan Biokimia Deteriorasi Benih Kedelai (*Glycine max* L) dalam Penyimpanan. Jurnal Ilmu Pertanian. Vol 11 : 76-87

Tatipata, A. 2010. Perubahan asam lemak selama penyimpanan benih kedelai (*Glycine max* L.Merr) dan hubungannya dengan viabilitas benih. J. Agon.Ind.Vol.XXXVIII (1) : 30-35

Tekrony, D.M., D.B. Egli and G.M. White. 1987. Seed Production and Technology. In . J.R. Wilcox (ed.) Soybean : Improvement, Production and Uses. Second Edition. Am. Soc. of Agron.,Inc. Crop. Sci .Soc .Am.Inc, Soil Sci.Soc.Am, Inc.

Viera. R.D. ; D.M. Tekrony ; D.B. Egli and M. Rucker. 2001. *Electrical conductivity of Soybean seeds sfter storage in several environments.* Seed Science and Technology., 29. 599-608.

Zainal, A., A. Anwar, S. Ilyas, Sudarsono dan Giyanto. 2010. Efektivitas Ekstrak Tumbuhan untuk mengeliminasi *Clavibacter michiganensis* pada benih tomat. J. Agon.Ind.Vol.XXXVIII (1) : 52-59

THE EFFECTS OF *Lactobacillus bulgaricus* and *Streptococcus thermophilus*

AS RATION SUPPLEMENT ON BROILER CARCASS WEIGHT, CARCASS FAT CONTENT AND THE SERUM CHOLESTEROL CARCASS CONTENT.

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ABSTRACT

Hundred final stock Arbor Acres were used in this experiments, using Completely Randomized Design (CRD) with four treatments : R-0 - as control (0.0% *Lactobacillus bulgaricus* and *Streptococcus thermophilus*), R-1 (1.0% *Lactobacillus bulgaricus* and *Streptococcus thermophilus),* R-2 (1.5% *Lactobacillus bulgaricus* and *Streptococcus thermophilus*), and R-3 (2.0% *Lactobacillus bulgaricus* and *Streptococcus thermophilus*); which was five times repeated. Results indicated that R-3 (ration that has supplemented with 2.0% *Lactobacillus bulgaricus* and *Streptococcus thermophilus*), has the highest carcass weight but the carcass fat content and the serum cholesterol carcass content was the lowest.

Keywords : *Lactobacillus bulgaricus* and *Streptococcus thermophilus*, broiler carcass, carcass fat content and serum cholesterol carcass content.

INTRODUCTION

Using probiotic, will significantly improved the body weight of the broilers. Carcass quality and blood contituents were determined and the administration of probiotic affected positively the body weight, feed intake and feed conversion rate by 7.7 and 8.1%, respectively (P> 0.05) compared to the control group. Lower concentration of serum cholesterol and triglycerides were observed in the treatment group. The probiotic addition reduced the fat content of the chicken meat (Ignatova, et al, 2009). The effect of probiotic started at two weeks of age. The differences in the body weight became greater towards the end of the trial periods. The birds fed on probiotic level 1 g/kg exhibited higher body weights among groups at all times of the trial. Lengkey, et al (2009) was isolated heterofermentative *Lactobacillus* species from raw poultry meat. Khaksefidi and Ghoorchi, 2006 report that probiotic supplemented to the birds improve the body weight and daily weight gain of broiler. Mohan et al (1996) reported that the beneficial effect of probiotic on chicken occurred only after the 4th week of growth. Edens (2003), reported that the inclusion of desirable microorganisms (probiotics) in the diet allows rapid development of beneficial bacteria in the digestive tract of the host, improving its performance. According to Arun et al (2006), the probiotic supplementation of *Lactobacillus sporogenes* at 100 mg/kg diet will reduced the serum total cholesterol and the triglycerides were reduced significantly. The significant reduction in serum cholesterol of broiler chickens fed probiotic supplemented diet could be attributed to reduced absorption and/or synthesis of cholesterol in the gastro-intestinal tract by probiotic supplementation (Mohan, et al, 1996)

Chickens fed with various level of probiotic showed a significant decrease in cholesterol concentrations when compared to the control group. Ignatova et al (2009) reported that the probiotic supplementation reduced the serum cholesterol and triglyceride significantly. Mohan et al (1996) mentioned that chickens that received 75, 100, and 125 mg probiotic/kg diets had lower serum cholesterol content (93.3 mg/100 ml) compared to the control birds (132.2 mg/100 ml). According to Abdulrahim et al, 1996; *Lactobacillus acidophilus* reduces the cholesterol in the blood by deconjugating bile salts in the intestine, thereby preventing them from acting as precursors in cholesterol synthesis.

MATERIAL AND METHODE

The Chickens are Broiler Arbo Acres 707 doc, but for these experiments, after 4 weeks old. The broiler carcass weight, fat content in the carcass, serum cholesterol carcass content was detect at 4 weeks old, using Completely Randomized Design (CRD).

RESULTS AND DISCUSSIONS

Probiotic effects on Broiler Carcass Weight

In Table 1, there is the results of probiotic supplementation on broiler carcass weight.

Table 1. The effect of *Lactobacillus bulgaricus* and *Streptococcus thermophilus* on Broiler Carcass Weight (g)

		Treatment	(%)		Total
Repetition	R-0	R-1	R-2	R-3	
1	909.55	997.64	999.98	1058.92	
2	915.45	993.70	1000.77	1062.28	
3	909.45	999.06	1001.05	1060.90	
4	910.15	992.24	1001.05	1081.40	
5	911.15	995.66	1001.00	1041.00	
Total	4555.75	4978.30	5003.85	5304.50	19842.40
Average	911.15	995.66	1000.77	1060.90	

From Table 1, it can be noticed that the probiotic treatments (R-1; R-2 and R-3), showed more broiler carcass weight than control group (R-0). These results showed that the probiotic supplemented groups improved the body weight and get more carcass weight than the control groups, that feed no probiotic in the diet. It means that the beneficial effect of probiotic on chicken ration. The improvement in the body weight in this study may be due to the increased efficiency of digestion and nutrient absorbtion processed due to precence of the probiotic bacteria. As a consequence, there is an improvement in the intestinal environment, increasing the efficiency of digestion and nutrient absorption processes.

According to Ignatova et. al (2009), the administration of probiotic affected positively body weight ($p<0.001$), feed intake and feed conversion rate by 7.7 and 8.1%, respectively ($p>0.05$) compared to the control group.

Effects of Probiotics on Fat Content in the Carcass.

Table 2, shows that the fat content in the carcass of the chickens that have fed with probiotic diet, get less fat content than the carcass from the control diet. The probiotic supplementation showed less fat content in the carcass, in proportion to the content of probiotic in the diet. The carcass from the chickens that fed with no probiotic in the ration have the highest fat content in the carcass. And the group with more probiotic in the diet, have the less fat content in the carcass, because the lactic acid in the probiotic will lower the pH in digestive tracts and inhibit the growing of other bacteria, especially the harmful bacteria. And also the energy from the carbohidrat was convert into lactic acid, so the fat formation in the carcass will decreased. The triglycerides were reduced by dietary supplementation of probiotic in the ration. Similar results were reported by Arum et al (2006) who found that triglyceride were reduced significantly by dietary supplementation of probiotic containing *Lactobacillus sporogenus* at 100 mg per kg diet.

Table 2. Fat content in the carcass that fed on rations containing different concentration of probiotic (g/kg feed)

		Treatment	(%)		Total
Repetition	R-0	R-1	R-2	R-3	
1	4.92	4.32	4.67	2.21	
2	5.87	3.68	3.76	2.25	
3	5.91	3.80	2.82	2.66	
4	4.74	3.78	4.09	4.05	
5	3.61	4.90	3.08	3.13	
Total	25.05	20.40	18.45	14.30	78.20
Average	5.01	4.08	3.69	2.86	

Effect of probiotic in the serum cholesterol carcass content.

Table 3, shows the serum cholesterol carcass content of the chickens that have fed with probiotic diet.

Table 3. Serum cholesterol carcass content of the chickens that fed on rations containing different concentration of probiotic (mg/g carcass)

		Treatment	(%)		
Repetition	R-0	R-1	R-2	R-3	Total
1	117	112	114	109	
2	123	108	100	113	
3	113	114	108	103	
4	130	134	103	100	
5	127	102	115	105	
Total	610	570	540	530	2250
Averages	122	114	108	106	

Table 3 clearly demonstrated that the probiotic in the diet will reduced the serum cholesterol carcass content. Chickens that received probiotic had lower serum cholesterol, (106, 108 and 114 mg/100 g carcass) compared to 122 mg/100 g carcass, from the control birds. Chickens that received probiotic *Lactobacillus* has found to have a high bile salt hydrolytic activity, which is responsible for deconjugation of bile salt. Similar results were reported by Arun et al (2006), who found that serum total cholesterol and triglycerides were reduced significantly by dietary supplementation of probiotic containing *Lactobacillus sporogenes* at 100 mg per kg diet.

Mohan et al (1996) mentioned that chickens that received 75, 100, and 125 mg probiotic/kg diets had lower serum cholesterol content (93.3 mg/100 ml) compared to the control birds

(132.2 mg/100 ml). The significant reduction in serum cholesterol of broiler chickens that fed probiotic supplemented diet could be attributed to reduced absorption and/or synthesis of cholesterol in the gastro-intestinal tract by probiotic supplementation (Mohan et al, 1995). According to Ignatova et al (2009), the probiotic supplementation reduced the serum cholesterol and triglyceride significantly.

CONCLUSIONS

The probiotic supplementation in the diet, incearsed the body weight, because it was increased feed intake and improved feed utilization.

The probiotic supplementation also reduced the serum cholesterol and triglyceride significantly.

REFERENCES

Abdulrahim, S. M., M. S. Y. Haddadin, E. A. R. Hashlamoun and R. K. Robinson. 1996. The influence of *Lactobacillus acidophilus* and *bacitracin* on layer performance of chickens and cholesterol content of plasma and egg yolk, British Poultry Science 341 (1996), p. 346.

Arun, K. P., Rama Rao, V. Savarm, V. L. N. Raju Mantena and R. Sharma Sita. 2006. Dietary supplementation of *Lactobacillus sporogenes* on performance and serum biochemico-lipid profile of broiler chickens. Journal of Poultry Science 43 (2006). pp. 235 – 240.

Eden, F. W., 2003. An alternative for antibiotic use in poultry probiotics. Brazilian Journal of Poultry Science 5 (2003), pp. 75 – 97.

Ignatova, M., V. Sredkova, V. Marasheva. 2009. Effect of Dietary Inclusion of Probiotic on Chickens Performance and Some Blood Indices. Journal for the Improvement of Animal Husbandry. Biotechnology in Animal Husbandry. Zemun – Belgrade. pp. 1079 - 1085

Khaksefidi, A and T. Ghoorchi, 2006. Effect of probiotic on performance and immunocompetence in broiler chicks. Journal of Poultry Science 43 (2006), pp. 296 – 300.

Lengkey, H. A. W., R. L. Balia, I. Togoe, B. A. Tasbac, and M. Ludong. 2009. Isolation and Identification of Lactic Acid Bacteria From Raw Poultry Meat. Journal for the Improvement of Animal Husbandry. Biotechnology in Animal Husbandry. Zemun – Belgrade. pp. 1071 – 1077.

Mohan, B., R. Kadirvel, M. Bhaskaran and M. Natarajan. 1995. Effect of probiotic supplementation on serum/yolk cholesterol and on egg shell thikness in layers. British Poultry Science 36 (1995). pp 799 – 803.

Mohan, B., R. Kadirvel, M. Natarajan and M. Bhaskaran. 1996. Effect of probiotic supplementation on growth, nitrogen utilization and serum cholesterol in broilers. British Poultry Science 37 (1996), pp. 799 – 803.