

Blood parameters and productivity of broilers fed ration composed of microparticle protein with the addition of *Lactobacillus* sp.

L. T. Wulandari, N. Suthama* and B. Sukamto

Faculty of Animal and Agricultural Sciences, Diponegoro University,
Tembalang Campus, Semarang 50275- Indonesia

*Corresponding E-mail: nsuthama@gmail.com

Received April 13, 2018; Accepted June 23, 2018

ABSTRAK

Penelitian bertujuan untuk mengkaji efektivitas penambahan *Lactobacillus* sp. pada ransum dengan protein mikropartikel terhadap status darah dan produktivitas broiler. Sebanyak 192 ekor broiler umur 21 hari, bobot badan 481 ± 67 g dibagi dalam rancangan acak lengkap (RAL) dengan 8 perlakuan dan 4 ulangan (@ 6 ekor) dan diberi perlakuan selama 3 minggu. Perlakuan sebagai berikut : T0: ransum protein non-mikro 21%, T1: ransum protein non-mikro 18%, T2: ransum protein mikropartikel 21%, T3: ransum protein mikropartikel 18%, T4: T0 + *Lactobacillus* sp. 1,2 mL, T5: T1 + *Lactobacillus* sp. 1,2 mL, T6: T2 + *Lactobacillus* sp. 1,2 mL, dan T7: T3 + *Lactobacillus* sp. 1,2 mL. Parameter yang diamati yaitu total bakteri asam laktat dan *Coliform*, pH, kolesterol darah, *low density lipoprotein* (LDL), *high density lipoprotein* (HDL), kolesterol daging, bobot daging, konsumsi ransum, dan pertambahan bobot badan (PBB). Data dianalisis ragam dan dilanjutkan dengan uji Duncan taraf 5%. Hasil penelitian menunjukkan bahwa ransum T7 nyata ($P < 0,05$) menurunkan kolesterol darah, LDL, dan kolesterol daging, tetapi meningkatkan HDL, bobot daging, dan PBB. Kolesterol darah, LDL, dan kolesterol daging nyata ($P < 0,05$) paling rendah akibat perlakuan T7. Kesimpulan, pemberian protein mikropartikel 18% dengan penambahan *Lactobacillus* sp. 1,2 mL dapat memperbaiki status darah dan meningkatkan produktivitas broiler

Kata Kunci: ayam broiler, profil darah, *Lactobacillus* sp., protein mikropartikel, produktivitas

ABSTRACT

The study was aimed to evaluate the effectiveness of feeding dietary microparticles protein added with *Lactobacillus* sp. on blood parameters and broiler productivity. A total of 192 birds of 21 days old broiler with initial body weight of 481 ± 67 g were divided into 8 treatments and 4 replications (6 birds each) of a completely randomized design (CRD). Dietary treatments were T0: 21% intact protein ration, T1: 18% intact protein ration, T2: 21% microparticle protein ration, T3: 18% microparticle protein ration, T4: T0 + 1.2 mL *Lactobacillus* sp, T5: T1 + 1.2 mL *Lactobacillus* sp., T6: T2 + 1.2 mL *Lactobacillus* sp., and T7: T3 + 1.2 mL *Lactobacillus* sp. Parameters observed were total lactic acid bacteria and *Coliform*, pH, blood cholesterol, low density lipoprotein (LDL), high density lipoprotein (HDL), meat cholesterol, meat weight, feed consumption, and body weight gain (BWG). Data were analyzed using analysis of variance and followed by Duncan test ($P < 0.05$). The results showed that T7 treatment significantly ($P < 0.05$) decreased blood cholesterol, LDL, and meat cholesterol, but increased HDL, meat weight, and BWG. Blood cholesterol, LDL, and meat cholesterol were indicated significantly ($P < 0.05$) lowest values due to T7 treatment. In conclusion, feeding 18% microparticle protein with addition of 1.2 mL *Lactobacillus* sp. can improves blood status and increases broiler productivity.

Keywords: broiler chicken, blood profiles, *Lactobacillus* sp., microparticles protein, productivity

INTRODUCTION

Along with the advanced technology and information, public awareness of the importance of health and nutritional value of animal protein increased. Broiler is a livestock commodity that can produce meat to meet the needs of animal protein. Consumer concerns are related to livestock products that contain high fat such as broiler as fast growing animals also have high fat deposition. The rapid broiler growth should be provided high-protein ration content, but since protein source ingredients are high in price, thus, feed cost also tend to be expensive. Feed cost is the biggest portion of productivity components, which covers about 60-70% of total cost. Considering feed cost in broiler production comprises the largest portion, an effective effort is needed in order to improve productive efficiency (Thirumalaisamy *et al.*, 2016). Reducing dietary protein content is one way that can be applied to reduce feed cost. However, the consequence of providing low protein ration has an impact on the lack of protein supply for broilers. To anticipate those problems, it needs to develop an alternative way to increase, at least maintaining, protein utilization. Processing intact protein ingredients to become microparticle is an attempt to reduce protein diet that can be a possible practical technology to improve protein availability (Suthama and Wibawa, 2016), and amino acids digestibility (Suthama and Wibawa, 2018).

Feeding microparticle protein with low protein ration can be anticipated by the addition of probiotics *Lactobacillus* sp. that can utilize dietary carbohydrate source, especially low molecule weight carbohydrate, by fermentation, and results short chain fatty acids (SCFA) which acid bring about the decreased intestinal pH condition. Thus, such condition stimulate the increase in lactic acid bacteria (LAB) and decrease pathogenic bacteria. Lactic acid bacteria (LAB) can produce bile salt hydrolase (BSH), an enzyme that can work in the changing process of conjugated bile salt become deconjugated form. Probiotics were able to increase the growth of LAB to produce BSH enzyme and lowered blood cholesterol (Ooi and Liong, 2010). The increase in LAB population with low pathogenic bacterial number is the indication of the better intestinal health which provides an impact on the improvement of nutrients digestibility. Feeding microparticle protein composed diet was supposed to increase protein utilization due to the

improved digestibility as previously reported (Suthama and Wibawa, 2017), and attributable to the better meat production characteristics (Abdurrahman *et al.*, 2016b).

Probiotics are beneficial microorganisms that can provide positive effects on intestinal health by creating acidic condition through the improvement of microflora balance (Getachew, 2016). The work of *Lactobacillus* sp. as probiotic is expected to support the productivity of broiler when microparticle diet is fed. Previous research has shown that dietary inclusion of probiotics improved the productivity of broiler (Jin *et al.*, 1998). The advantages of bacterial strains such as *Lactobacillus* have the ability in lowering cholesterol via the activity of BSH and increase intestinal homeostasis (Grunewald, 1982). Microparticle protein is more effectively utilized by broiler supported by the contribution of probiotics *Lactobacillus* sp. in improving intestinal health. Study concerning the combining effect of feeding microparticle protein and *Lactobacillus* sp. on lipid profile, meat quality, and body weight gain in broiler chicken have never been previously conducted. Therefore, it is necessary to elucidate blood lipid parameters and productivity of broilers fed a combination of microparticle protein diet and the addition of *Lactobacillus* sp.

MATERIALS AND METHODS

Experimental Animal and Feed

Experimental animals were 192 birds of 21-day sex-separated broiler with the ratio of male and female was 50 and 50 (initial body weight was 481 ± 67 g), and given dietary treatment for 3 weeks, from day 21 to day 42. Rations and drinking water were available *ad libitum*. The basal ration was composed of ground corn, rice bran, fish meal, soybean meal, premix, and calcium carbonate (CaCO₃). Intact fish meal and soybean meal were processed to become microparticle based on the method of Jambrak *et al.* as modified by Suthama and Wibawa (2016). Intact and microparticle protein-composed feeds were created as basal feed added with probiotic *Lactobacillus* sp. at 1.2 mL (10^8 cfu/mL). Table 1 indicated detail composition of experimental feeds.

Treatment and Experimental Design

The study was assigned in a completely randomized design (CRD) with 8 treatments and 4

Table 1. Composition and Nutrient Content of Experimental Feed

Feedstuff	Feed Composition			
	Intact Protein (21%)	Intact Protein (18%)	Microparticle Protein (21%)	Microparticle Protein (18%)
Ground corn	48.00	50.50	48.00	50.50
Rice bran	14.00	20.00	14.00	20.00
Intact soybean meal	27.00	21.00	0.00	0.00
Microparticle soybean meal	0.00	0.00	27.00	21.00
Intact fish meal	10.00	7.50	0.00	0.00
Microparticle fish meal	0.00	0.00	10.00	7.50
CaCO ₃	0.50	0.50	0.50	0.50
Premix	0.50	0.50	0.50	0.50
Total	100	100	100	100
Nutrient Content* (%)				
Metabolizable energy (kcal/kg)**	2978.41	2948.32	2978.41	2948.32
Crude protein	21.29	18.12	21.29	18.12
Ether extract	2.81	2.70	2.81	2.70
Crude fiber	4.27	4.77	4.27	4.77
Calcium	1.03	0.88	1.03	0.88
Phosphorus	0.65	0.61	0.65	0.61
Methionine***	0.45	0.39	0.45	0.39
Lysine***	1.37	1.12	1.37	1.12
Arginine***	1.51	1.28	1.51	1.28

*Analyzed at the Laboratory of Nutrition and Feed Science, Faculty of Animal and Agricultural Sciences, Diponegoro University (2016)

**Calculated based on the formula of Carpenter and Clegg (1965)

***Based on Table of National Research Council (1994)

replications (6 birds each). Treatments applied in this study were as follows: 21% intact protein ration (T0), 18% intact protein ration (T1), 21% microparticle protein ration (T2), 18% microparticle protein ration (T3), 21% intact protein ration + 1.2 mL *Lactobacillus* sp. (T4), 18% intact protein ration + 1.2 mL *Lactobacillus* sp. (T5), 21% microparticle protein ration + 1.2 mL *Lactobacillus* sp. (T6), and 18% microparticle protein ration + 1.2 mL *Lactobacillus* sp. (T7).

Parameters and Statistical Analysis

Parameters observed were intestinal LAB and *Coliform* counts, blood cholesterol, LDL, HDL, meat cholesterol, feed consumption and productivity based on meat weight and

cummulative body weight gain (BWG). Blood samples of about 2 ml was taken on day 42 through the brachial vein and collected in the test tube containing ethylene diamine tetraacetic acid (EDTA), and temporary was stored in a cooling box for further analysis. Meat weight was measured and meat sample was obtained from all components of the carcass, and meat sample was then mixed and homogenized prior to analysis. Meat cholesterol was analyzed according to *Lieberman Burchard* method using spectrophotometer at 550 nm wavelength. Determination of blood cholesterol, LDL, and HDL were performed based on the methods of enzymatic- colorimetric.

Feed consumption and cummulative body

weight gain (BWG) were measured during 3 weeks, from day 21 to 42. Feed consumption (g/bird) was measured during 3 weeks of observation. Body weight gain (g/bird) was obtained from the difference between final body weight and initial body weight. Potential hydrogen (pH) was measured from the digesta using pH meter. Intestinal LAB and *Coliform* populations were measured from duodenum and jejunum using media deman Rogosa Sharpe (MRS) and eosin methylene blue agar (EMBA). Planting process was proceeded with total plate count method according to Fardiaz (1993) with the formula namely, total colony (cfu/g) = total colony x 1/dilution factor per plate. LAB and *Coliform* counts were determined from duodenum and jejunum based on the consideration that the most nutrients absorption was undergone in the upper intestinal segments (Rinttila and Apajalahti, 2013) under the interference effects of the bacteria, while the absorptive rate in the ileum was very low since digestive enzyme activities have reduced (Rehman *et al.*, 2007). Data were analyzed using analysis of variance, and continued to Duncan test at 5% probability level (Steel and Torrie, 1981).

RESULTS AND DISCUSSION

Blood Cholesterol, Low Density Lipoprotein (LDL), and High Density Lipoprotein (HDL)

Treatments of feeding 21 and 18% intact protein as well as microparticles protein without or with the addition of *Lactobacillus* sp. on blood cholesterol, LDL, and HDL are presented in Table 3. Feeding microparticle protein with the addition of *Lactobacillus* sp. was significantly ($P < 0.05$) decreased blood cholesterol, LDL, but increased HDL. Blood cholesterol seemed to be closely related to both LDL and HDL. The decrease in blood cholesterol could be connected with the contribution of soybean oligosaccharide (SOS) function as prebiotic derived from soybean meal microparticle which can be readily fermented by *Lactobacillus* sp. in the digestive tract. The mechanism is discussed in the following paragraph.

Feeding microparticle protein ensures the more effective access of digestive enzymes due to the smaller particle size. Small to medium particle size of feed would be beneficial for digestive tract development, nutrients digestibility, and growth performance (Gabriel *et al.*, 2003; Mingbins *et al.*, 2015) when it was offered in pelleted form.

The addition of probiotics into the diet was able to increase digestibility of nutrients by adjusting the appropriate pH conditions of digestive tract to digest nutrients (Hyden, 2000). The finding of Musa *et al.* (2006) showed a positive correlation between blood lipid profiles and probiotics treatment. The phenomenon in this study indicated that feeding microparticle protein with the addition of *Lactobacillus* sp. was possible to decrease blood lipid profiles such as blood cholesterol and LDL. Process of making microparticle protein source ingredients, especially soybean meal, provided also the change in its condition of oligosaccharide that function as simple prebiotic carbohydrate called as soybean oligosaccharide. Soybean oligosaccharide (SOS) derived from soybean meal can be fermented by additional *Lactobacillus* sp. working together with beneficial endogenous LAB to produce metabolic substance known as SCFA. Production of SCFA facilitated intestinal environment to be acidic condition, and consequently promoting the activity of LAB to be better growth and development, on the other hand, the activity of pathogenic bacteria such as *Coliform* was inhibited (Table 2). Dietary inclusion of soybean oligosaccharide was reported to have beneficial effect on the increase in LAB, and can be connected with the improved protein digestibility and meat quality (Suthama *et al.*, 2018).

The product of SCFA brought about the decreased intestinal pH which can affect the reduction of pathogenic bacteria and the increase in lactic acid bacteria (Dzirkova *et al.*, 2005). The decrease in circulating cholesterol could be the impact of the increase in SCFA production. SCFA were the main products of fermentative activity of beneficial bacteria such as lactic acid bacteria (LAB) in the poultry digestive tract (Fiordaliso *et al.*, 1995). The increasing number of LAB that can produce BSH enzyme is associated with the decreased blood cholesterol. The measurement of BSH enzyme was not performed in the present study, but it might deconjugate bile salts that can not hydrolyze dietary fat as described by previous researchers (Begley *et al.*, 2006; Fajrih *et al.*, 2014). The deconjugated bile salt has low effectiveness on fat emulsion in the small intestine, therefore, the rate of fat absorption to be low. In addition, the more deconjugated bile salts are formed the more need for new bile salts formation that requires cholesterol as raw materials and leads to be lower blood cholesterol concentration. Moser and Savage (2001) stated

Table 2. Populations of Intestinal Lactic Acid Bacteria (LAB) and *Coliform*, and pH in Broiler Fed Microparticle Protein Diet with Inclusion of *Lactobacillus* sp.

Parameters	LAB (10^8 cfu/g)	<i>Coliform</i> (10^6 cfu/g)	Intestinal pH
T0	1.42±0.28 ^b	4.31±0.34 ^a	6.20±0.23 ^a
T1	1.63±0.29 ^b	2.85±0.40 ^b	5.98±0.17 ^{ab}
T2	1.54±0.18 ^b	2.38±0.28 ^{bc}	5.78±0.08 ^{bc}
T3	2.55±0.48 ^a	2.91±0.48 ^b	6.00±0.15 ^{ab}
T4	2.50±0.63 ^a	2.61±0.18 ^{bc}	5.60±0.17 ^c
T5	2.14±0.29 ^{ab}	2.53±0.35 ^{bc}	5.75±0.28 ^{bc}
T6	2.00±0.15 ^{ab}	2.23±0.04 ^c	5.60±0.15 ^c
T7	2.55±0.54 ^a	2.23±0.34 ^c	5.45±0.08 ^c

^{a-c}Values in the same column followed by different superscript show significantly difference (P<0.05)

that the liver tried to synthesize bile salt to replace cholesterol lost along with the excreta. BSH enzyme activity was known to work in the process of bile salts deconjugation and could help to decrease cholesterol and increase intestinal homeostasis that also allowed the mechanism of cholesterol reshuffle or degradation by intestinal microbes (Grunewald, 1982).

The mechanism of decreased blood cholesterol is suggested to correlate with LDL and HDL levels. LDL levels decreased due to the production of SCFA by LAB activity, and on the contrary, HDL increased significantly. Gropper *et al.* (2018) stated that the concentration of blood cholesterol affects both LDL and HDL. LDL plays an important contribution in releasing cholesterol in the transportation metabolism of the liver into the target tissues, so that decreased blood LDL. The increase in cholesterol was followed by an increase in LDL. As it has been known that LDL is a carrier of cholesterol from the liver to the tissues. In contrast, the increased HDL was followed by the decreased blood cholesterol (Daniels *et al.*, 2009). Cholesterol was circulated by blood in the form of LDL and HDL which were function as component of cholesterol transporter (Hirakawa, 2005). The mechanism of LDL reduction is closely related to low blood cholesterol, as well as the activity of lactic acid bacteria (LAB) in producing BSH enzyme. It has been previously discussed that BSH enzyme causes bile salts to be deconjugated. Deconjugated bile salt is less efficient to emulsify

fats, and caused low absorbed fat indicated by low LDL as found in the present study. LDL and HDL are the two types of plasma lipoproteins that function to transport blood cholesterol as an indication that concentration in the blood was greatly influenced by the amount of cholesterol synthesis (Diestchy, 2003).

Meat Cholesterol, Meat Weight, Feed Consumption, and Body Weight Gain

Meat cholesterol of either 21% or 18% dietary proteins both in the form of intact and microparticle added with *Lactobacillus* sp. were significantly (P<0.05) decreased (Table 4) and lower as compared to control group. In contrast, that of 18% intact and microparticle protein diets without *Lactobacillus* sp. were significantly (P<0.05) increased, but that of 21% microparticle protein diet was similar to control group. A decrease in blood cholesterol (Table 3) provided an impact on low cholesterol mobilization that can be deposited into the meat. Roberfroid *et al.* (2010) reported that intestinal microbes, especially beneficial bacteria such as *Lactobacillus* and *Bifidobacteria*, play important role in the formation of SCFA. The phenomenon found in the present study was supported by the increased LAB population and the decreased intestinal pH (Table 2) which were the indications of the increasing dietary lipid breakdown. Some previous studies (Sudha *et al.*, 2009; Kumar *et al.*, 2013; Rai *et al.*, 2013) revealed that the presence of beneficial bacteria could increase intestinal

homeostatis and allowing the process of destruction or degradation of cholesterol performed by intestinal microorganisms via the conversion of cholesterol into bile salts so that circulating cholesterol levels decreased. The increase in LAB population promotes the production of BSH enzyme which can inhibit fat absorption in general and cholesterol in particular, and finally decreased meat cholesterol.

As it has been previously discussed that the work of BSH enzymes caused bile salt to be deconjugated and this form of bile salt have low activity on lipid emulsion in the intestine and mostly excreted via excreta. Other possibility of the decreased cholesterol meat was due to the need of cholesterol for bile salt forming compound. Abdurrahman *et al.* (2016a) found that the decreased fat and cholesterol meat in crossbred local chickens were due to feeding effect of probiotics *Lactobacillus* sp. combined with prebiotic inulin. Similar results as previously reported (Fajrih *et al.* 2014) that feeding dietary inclusion of prebiotic inulin could improve performance and meat quality of crossbred local chickens. Weight of meat in T5 to T7 treatments significantly increased due to dietary inclusion effect of probiotic *Lactobacillus* sp. (Table 4). The increased meat weight was also found in T2 treatment, but meat weight in T1 and T3 treatments were similar to that in control group (T0). These results can be assumed that the addition of *Lactobacillus* sp. was more pronounced irrespective of dietary protein forms

and feed consumption (Table 4). However, the interesting result found in the present study was that the higher cumulative body weight gain (with similar feed consumption), especially in T7 (Table 4), was achieved with *Lactobacillus* sp. inclusion into low dietary protein (18%, Table 1). This phenomenon was due to the beneficial effect of probiotic *Lactobacillus* sp. although with lower protein supply since feed consumption was the same.

The increase in the amount of LAB in treatments of T7 was higher than that in treatments T0 to T2 treatments (Table 2), resulted healthier digestive tract condition and brought about better nutrient, especially protein, digestion and absorption. Wong *et al.* (2006) stated that short chain fatty acids (SCFA), mainly acetate, propionate, and butyrate, could reduce the production of toxins by bacteria and improve the morphology of the intestinal wall, and they were also able to reduce the colonization of pathogenic bacteria (Pourabedin and Zhao, 2015). If the balance of intestinal microflora can be maintained stable it should brought about the increase in the host's body health leading to improve the performance of broilers. In case of this study, it was characterized by the increase in meat weight and body weight gain with the same amount of feed consumption (Table 4).

The low intestinal pH in T4 to T7 treatments inhibited the growth of pathogenic bacteria as indicated by the decreased number of *Coliform* (Table 2). Similarly, the addition of prebiotics can

Table 3. Blood Cholesterol, Low Density Lipoprotein (LDL) and High Density Lipoprotein (HDL) in Broiler Fed Microparticle Protein Diet with Inclusion of *Lactobacillus* sp.

Treatments	Cholesterol	LDL	HDL
	----- (mg/dl) -----		
T0	135.76±2.05 ^a	112.43±15.10 ^a	21.28±1.99 ^c
T1	135.12±1.51 ^a	126.59±11.73 ^a	23.70±1.65 ^c
T2	117.36±4.11 ^b	120.07 ± 9.67 ^a	20.77±0.79 ^c
T3	123.96±8.41 ^b	115.20±12.60 ^a	25.55±2.14 ^{bc}
T4	124.15±9.97 ^b	108.37±16.20 ^a	28.10±3.23 ^{bc}
T5	121.16±1.29 ^b	103.30±16.10 ^a	36.00±7.83 ^a
T6	122.22±7.85 ^b	103.61±16.90 ^a	32.28±2.70 ^{ab}
T7	118.06±4.61 ^b	77.32±11.30 ^b	35.69±7.03 ^a

^{a-c}Values in the same collumn followed by different superscript show significantly difference (P<0.05)

Table 4. Table 4. Meat Cholesterol, Meat Weight, Cummulative Feed Consumption and Body Weight Gain in Broiler Fed Microparticle Protein Diet with Inclusion of *Lactobacillus* sp.

Treatments	Meat Cholesterol (mg/100g)	Meat Weight (g)	Cummulative Feed Consumption (g/bird)	Cummulative Body Weight Gain (g/bird)
T0	110.20±1.39 ^c	319.69±4.81 ^{cd}	1230.45±80.73	548.75±48.56 ^b
T1	115.33±1.57 ^{ab}	309.78±6.02 ^d	1127.29±27.12	558.75±49.54 ^b
T2	113.44±3.42 ^{bc}	334.35±2.83 ^{ab}	1164.72±53.93	563.25±43.16 ^b
T3	118.42±1.02 ^a	310.33±5.12 ^d	1119.94±31.30	618.75±28.83 ^{ab}
T4	84.63±3.83 ^d	324.33±5.17 ^{bc}	1244.05±78.24	612.25±36.47 ^{ab}
T5	83.40±1.84 ^d	343.56±4.35 ^a	1170.38±34.58	636.25±59.88 ^{ab}
T6	82.17±0.89 ^d	341.89±10.5 ^a	1188.71±48.24	638.75±58.45 ^{ab}
T7	73.28±4.82 ^c	340.19±6.89 ^a	1162.83±93.56	679.25±73.56 ^a

^{a-c}Values in the same collumn followed by different superscript indicate significantly difference (P<0.05)

increase lactic acid bacteria and decrease the population of pathogenic bacteria (Krismiyanto *et al.*, 2014). The improved intestinal bacteria balance in the present study could be achieved due to the decrease in pathogenic bacteria (*Coliform*) as previously described. Therefore, better condition of microbial balance affected the health of the chicken gastrointestinal tract causing the increased production performance (Liu *et al.*, 2012). LAB can create better environment of the intestine by extending the active enterocyte cell's period, so that intestine performance getting better (Markovic *et al.*, 2009). In connection with the present study, this phenomenon was closely related to the increased intestinal work to digest and absorp nutrients and concomitant with the support of feeding microparticle protein diet. Therefore, it was greatly possible to have an impact on the improvement of nutrient supply, especially protein, and ultimately brought about the increase in either meat weight or body weight gain (Table 4). Gibson *et al.* (2011) reported that the provision of probiotics was able to promote the growth and activity of beneficial bacteria but inhibit pathogenic bacteria and lead to the stimulation of immune response which was followed by the increased host's body health. This condition brought about the increased meat weight in T5 to T7 treatments. Similar result was found in broiler fed diet with inclusion of prebiotic soybean oligosaccharide (SOS) that the

increased lactic acid bacteria population brought about the improved intestinal health and presumably digestive enzymes activity as well (Suthama *et al.*, 2018). The increased enzyme activity was indicated by the improved protein digestibility as a manifestation of higher availability of protein supply for protein deposition. Protein supply acts as an important function on protein deposition which is affected by the increased intestinal health status in particular, indicated by high LAB counts and low *Coliform* number (Table 2), and higher body resistance in general, revealed by lower heterophyl-lymphocyte ratio (H/L ratio) (Suthama and Wibawa, unpublished data). Therefore, the interrelationship of metabolic phenomenon and physiological parameters as described above was an important determinant for the increase in either meat weight or body weight gain.

CONCLUSION

It can be concluded that feeding 18% microparticle protein-composed ration with the addition of *Lactobacillus* sp. at 1.2 mL decreases blood bad lipid and meat cholesterol, and increases meat weight and body weight of broiler.

REFERENCES

Abdurrahman, Z.H., Y. B. Pramono and N.

- Suthama. 2016a. Feeding effect of inulin derived from dahlia tuber combined with *Lactobacillus* sp. on meat protein mass of crossbred kampong chicken. *J. Indonesian Trop. Anim. Agric.* 41(1): 37-44.
- Abdurrahman, Z.H., Y. B. Pramono and N. Suthama. 2016b. Meat characteristic of crossbred local chicken fed inulin of dahlia tuber and *Lactobacillus* sp. *Med. Pet.* 39(2): 112-118.
- Begley, M., C. Hill and C.G.M. Gahan. 2006. Bile salt hydrolase activity in probiotics. *Appl. Environ. Microbiol.* 72(3): 1729-1738.
- Carpenter, K.J. and K.M. Clegg. 1965. 1965. The metabolizable energy of poultry feeding stuffs in relation to their chemical composition. *J. Sci. Food Agric.* 7(1): 45-51.
- Daniels, K. M. Killinger, J. J. Michal, R. W. Wright and Z. Jiang. 2009. Lipoproteins, cholesterol homeostasis and cardiac health. *Int. J. Bio. Sci.* 5(5): 474-488.
- Dietschy, J. M. 2003. How cholesterol metabolism and transport present novel targets for lipid treatment. *Adv. Stud. Med.* 3(4): 5319-5323.
- Dzirkova, B.G. Dongowski, E. Gebhardt and A. Habel. 2005. The composition of dietary fibre rich extrudates from oat affect bile acid binding and fermentation in vitro. *J. Food. Chem.* 90(1): 181-192.
- Fajrih, N., N. Suthama and V. D. Yunianto. 2014. Body resistance and productive performance of crossbred local chicken fed inulin of dahlia tubers. *Med. Pet.* 37(2): 108-144.
- Fardiaz, S. 1993. Analisis Mikrobiologi Pangan. Raja Grafindo Persada, Jakarta.
- Fiordaliso, M., N. Kok, J. P. Desager, F. Goethals, D. Deboyser, M. Roberfroid and N. Delzenne. 1995. Dietary oligofructose lowers triglycerides, phospholipids and cholesterol in serum and very low density lipoprotein of rat. *Lipids* 30(2): 163-167.
- Gabriel, I., S. Mallet and M. Leconte. 2003. Differences in the digestive tract characteristics of broiler chickens fed on complete pelleted diet or on whole wheat added to pelleted protein concentrate. *Br. Poult. Sci.* 44(1): 283-290.
- Getachew, T. 2016. A Review on effects of probiotic supplementation in poultry performance and cholesterol levels of egg and meat. *J. World Poult. Res.* 6(1): 31-36.
- Gibson, G.R., H.M. Probert, J.A.E. Van Loo and M.B. Roberfroid. 2004. Dietary modulation of the human colonic microbiota: updating the concept of prebiotics. *Nutr. Res. Rev.* 17(1): 257-263.
- Gropper, S. S., J. L. Smith and T. P. Carr. 2018. *Advanced Nutrition and Human Metabolism*. 7th Ed. Wadsworth.
- Grunewald, K. K. 1982. Serum cholesterol level in rats fed skim milk fermented by *Lactobacillus acidophilus*. *Appl. Environ. Microbiol.* 49(1): 377-381.
- Hirakawa, B. 2005. Cholesterol. *Encyclopedia of Toxicology*. 2nd Ed., Elsevier Inc. Amsterdam.
- Hyden. M. 2000. Protected acid additives. *Feed Int.* 21(7):14-17
- Jin, L. Z., Y. W. Ho, N. Abdullah and S. Jalaludin. 1998. Growth performance, intestinal microbial populations, and serum cholesterol of broilers fed diets containing *Lactobacillus* cultures. *Poult. Sci.* 77(1): 1259-1265.
- Krismiyanto, L., N. Suthama and H. I. Wahyuni. 2014. Feeding effect of inulin derived from *Dahlia variabilis* tuber on intestinal microbes in starter period of crossbred native chickens. *J. Indonesian Trop. Anim. Agric.* 39(4):217-223.
- Kumar, R., H. Rajkumar, M. Kumar, S. R. Varikuti, R. Athimamula, M. Shujuddin, R. Ramagoni and N. Kondapalli. 2013. Molecular cloning, characterization and heterologous expression of bile salt hydrolase (BSH) from *Lactobacillus fermentum* NCDO394. *Mol. Biol. Rep.* 40(1): 5057-5066.
- Liu, X., H. Yan, L. Lv, Q. Xu, C. Yin, K. Zhang, P. Wang and J. Hu. 2012. Growth performance and meat quality of broiler chickens supplemented with *Bacillus licheniformis* in drinking water. *Asian-Aust. J. Anim. Sci.* 25(1): 682-689.
- Markovic, R., D. Sefer, M. Krstic and B. Petrujkic. 2009. Effect of different growth promoters on broiler performance and gut morphology. *Arch. Med. Vet.* 41(1): 163-169.
- Mingbin, L.V., N.L. Yan, Z. Wang, S. An S, M. Wu and L.V. Zunzhou. 2015. Effects of feed form and feed particle size on growth performance, carcass characteristics and digestive tract development of broilers. *Anim. Nutr.* 1(1): 252 -256.

- Moser, S.A. and D.C. Savage. 2001. Bile salt hydrolase activity and resistance to toxicity of conjugated bile salt are unrelated properties in *Lactobacilli*. *J. App. Environ. Microbiol.* 67(1): 3476-3480.
- Musa H. H., G. H. Chen, K. H. Wang, B. C. Li, D. M. Mekki, J. T. Shu and H. P. Ju. 2006. Relation between serum cholesterol level, lipoprotein concentration and carcass characteristics in genetically lean and fat chicken breeds. *J. Biol. Sci.* 6(1): 616-620.
- National Research Council. 1994. Nutrient Requirements of Poultry. 9th Ed. National Academy Press, Washington DC.
- Ooi, L.G. and M. T. Liong. 2010. Cholesterol-lowering effects of probiotics and prebiotics: A Review of in vivo and in vitro. *Int. J. Mol. Sci.* 11(6): 2499-2522.
- Pourabedin, M. and X. Zhao. 2015. Prebiotics and gut microbiota in chickens. *FEMS Microbiol. Lett.* 362(15):1-8.
- Rai, A. K., P. Debetto and F. D. Sala. 2013. Molecular regulation of cholesterol metabolism: HDL-based intervention through drugs and diet. *Indian J. Exp. Biol.* 51(1): 885-894.
- Rehman, H. U., W. Vahjen, W. A. Awad and J. Zentek. 2007. Indigenous bacteria and bacterial metabolic products in the gastrointestinal tract of broiler chickens. *Arch. Anim. Nutr.* 61(1): 319-335.
- Rinttila. T. and J. Apajalahti. 2013. Intestinal microbiota and metabolites - implications for broiler chicken health and performance. *J. Appl. Poult. Res.* 22(1): 647-658.
- Roberfroid, M., G. R. Gibson, L. Hoyles, A. L. McCartney, R. Rastall, I. Rowland, D. Wolvers, B. Watzl, H. Szajewska, B. Stahl, F. Guarner, F. Respondek, K. Whelan, V. Coxam, M. J. Davicco, L. Leotoing, Y. Wittrant, N. M. Delzenne, P. D. Cani, A. M. Neyrinck and A. Meheust. 2010. Prebiotic effects: metabolic and health benefits. *Br. J. Nutr.* 104(1): S1-S63.
- Steel, R.G.D. and J.H. Torrie 1981. Principles and Procedures of Statistics: A Biometrical Approach. McGraw-Hill Book Company, New York, Toronto, London.
- Sudha, M. R., P. Chauhan, K. Dixit, S. Babu and K. Jamil. 2009. Probiotics as complementary therapy for hypercholesterolemia. *Biol. Med.* 1(1): 1-13.
- Suthama, N. and P. J. Wibawa. 2016. Digestibility evaluation of microparticle protein derived from fish meal and soybean meal in broiler chicken. Proc. International Seminar: The 3rd Animal Production International Seminar (3rd APIS), and The 3rd ASEAN Regional Conference on Animal Production (3rd ARCAP). Malang, October 19-21, 2016.
- Suthama, N. and P. J. Wibawa. 2017. Feeding Microparticle Protein-Composed Diet on Protease Activity and Protein Utilization in Broiler Chickens. Proc. The Sixth International Conference on Sustainable Animal Agriculture for Developing Countries. Batu, Malang, October 16-19, 2017.
- Suthama, N. and P. J. Wibawa. 2018. Amino acids digestibility of pelleted microparticle protein of fish meal and soybean meal in broiler chickens. *J. Indonesian Trop. Anim. Agric.* 43(2):169-176.
- Suthama, N., Y. B. Pramono, and B. Sukanto. 2018. Improvement of broiler meat quality due to dietary inclusion of soybean oligosaccharide derived from soybean meal extract. *Earth Environ. Sci.* 102: 012009.
- Thirumalaisamy, G., J. Muralidharan, S. Senthilkumar, R. Hema Sayee and M. Priyadharsini. 2016. Cost-effective feeding of poultry. *Int. J. Sci., Environ. Technol.* 5(6): 3997-4005.
- Wong, J. M., R. De Souza, C. W. Kendall, A. Eman and D. J. Jenkins. 2006. Colonic health: fermentation and short chain fatty acids. *J. Clin. Gastro.* 40(3): 235-243.