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Full Length Research

Effect of soybean meal extract and *Lactobacillus plantarum* on fat digestibility, fatty meat and carcass weight in broiler

Lilik Krismiyanto[1], Vania Carissa Widiawati[2], Vitus Dwi Yunianto[1] and Mulyono[1]*

[1]Department of Animal Science, Faculty of Animal and Agricultural Sciences, Universitas Diponegoro, Semarang, Indonesia.
[2]Bachelor Study Program of Animal Science, Faculty of Animal and Agricultural Sciences, Universitas Diponegoro, Semarang, Indonesia.

*Corresponding author. Email: qmulyo@gmail.com

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ABSTRACT: The study aimed to evaluate the addition of soybean meal extract and *Lactobacillus plantarum* on fat digestibility, relative weight of abdominal fat, meat fat mass and carcass weight of broiler chickens. The experimental animals used were 8-day-old unsexed *Cobb* strain broilers as many as 192 birds with a body weight of 177.89±3.73 g. Soybean meal extract (SME) as prebiotic and *Lactobacillus plantarum* as probiotic. The research ration was prepared based on metabolizable energy of 3,034.58 kcal/kg and crude protein of 21.73%. The study was arranged using a completely randomized design (CRD) with 6 treatments and 4 replicates, each experimental unit was filled with 8 animals. The treatments applied include T_0 (basal ration); T_1 (basal ration + *Lactobacillus plantarum* 1.2%); T_2 (basal ration + SME 0.15%); T_3 (basal ration + SME 0.30%); T_4 (basal ration + SME 0.15% + *Lactobacillus plantarum* 1.2%); T_5 (basal ration + SME 0.30% + *Lactobacillus plantarum* 1.2%). Parameters measured included fat digestibility, relative weight of abdominal fat, meat fat mass and carcass weight. Data were processed using analysis of variance and Duncan's test in the SPSS version 16 program. The results showed that the addition of treatments to the ration had a significant effect ($p<0.05$) on fat digestibility, relative weight of abdominal fat, and carcass weight, but had no significant effect ($p>0.05$) on broiler meat fat mass. The conclusion is that the addition of 0.3% soybean meal extract and 1.2% *Lactobacillus plantarum* to the diet can reduce fat digestibility, relative weight of abdominal fat and increase carcass weight and produce the same meat fat mass.

Keywords: Broiler, carcass, fatty meat, *Lactobacillus plantarum,* soybean meal.

INTRODUCTION

Rapid growth in broilers requires a complete and balanced intake of nutrients. Ration is the main factor affecting body weight gain and fat content in broilers. Operational costs for rations reach 60-70% of all production costs (Boğa *et al*., 2022). The ration provided must meet nutrient needs so that daily body weight gain can increase. Efforts to increase productivity can provide natural feed additives in the ration. Natural feed additives added in the ration can be sourced from prebiotics and probiotics. Prebiotics as a substrate or food for beneficial bacteria, while probiotics as beneficial bacteria added exogenously to increase lactic acid bacteria (LAB). This study used prebiotics sourced from soybean meal extract (SME) and the probiotic used was *Lactobacillus plantarum*.

Prebiotics are feed ingredients that cannot be absorbed by the host but can have a good influence in improving animal health. Prebiotics act as a substrate for probiotics, so that beneficial bacteria can develop optimally (Wijaya *et*

al., 2017). The prebiotics used are sourced from soybean meal extract. Soybean meal extract contains soybean oligosaccharides (SOS) that enhance the growth of lactic acid bacteria (LAB) and inhibit the development of pathogenic bacteria. Oligosaccharides contained in soybean meal include raffinose 0.73 g/100 g, and stakiose 0.90 g/100 g (Krismaputri *et al*., 2016). Fermentation of SOS by bacteria in the intestine produces short chain fatty acids (SCFA) that reduce intestinal pH so that beneficial bacteria increase, while pathogenic bacteria decrease.

Lactobacillus plantarum as a probiotic has the advantage of other LAB, namely being able to survive until the final stage of food fermentation because it is more resistant to temperature and acidic pH (Winarti *et al*., 2019). *Lactobacillus plantarum* as a probiotic produces the enzyme bile salt hydrolase (BSH) that can deconjugate bile salts, where glycine and taurine are separated from steroids to produce free or deconjugated bile salts. The BSH enzyme produces deconjugated bile salts in the form of free cholic acid which is absorbed by the gut. Bile salts that return to the liver during enterohepatic circulation are reduced so that total cholesterol in the body is reduced along with a decrease in fat content in the body (Salehizadeh *et al*., 2019).

The addition of SME and *Lactobacillus plantarum* in broiler rations is utilized in the small intestine through the fermentation process. Fermentation of SME as a substrate by LAB produces metabolite products in the form of SCFA and lactic acid. The result of SCFA creates a low pH condition of the small intestine (acidic), so that the LAB population increases and pathogenic bacteria decrease. Increased LAB can produce BSH enzymes that deconjugate fats for low fat absorption (Harumdewi *et al*., 2018). The ability of LAB to deconjugate bile salts by the BSH enzyme produced by LAB results in fat not being emulsified. BSH enzymes work to deconjugate bile salts making fat unabsorbable and wasted with excreta (Tolnai *et al*., 2021). Low fat deposition can provide good meat quality and low fatty meat, thereby increasing carcass weight in broilers.

Based on the description above, this study has the novelty of the combination of soybean meal extract and *Lactobacillus plantarum* has not been tested on broiler chickens. Determination of the percentage level of soybean meal extract is based on the results of research by Krismaputri *et al*. (2016) while the addition of *Lactobacillus plantarum* level is based on the results of research by Abdurrahman *et al*. (2016).

MATERIALS AND METHODS

Livestock, materials and equipment

The experimental animals used were 8-day-old unsexed *Cobb* strain broiler chickens as many as 192 birds with an average body weight of 177.89 ± 3.73 g. The research ration was prepared from a mixture of ration ingredients listed in Table 1. Materials used for soybean meal extraction include soybean meal flour, distilled water, 92% ethanol, aluminum foil and fine filter paper. Equipment used for maintenance includes a 60-watt bulb lamp, thermohygrometer Alcohol Hisamatsu, digital scales and hanging scales with 1 g accuracy. Tools used for soybean meal extraction include a waterbath maskot, 1 liter beaker glass, stirrer, tray, measuring cup, mortar and digital balance with 1 g accuracy.

Table 1. Composition and nutrient content of research rations.

Feed stuff	Composition (in %)
Yellow corn	50.51
Pollard	16.74
Soybean meal	21.90
Meat bone meal	10.00
$CaCO_3$	0.30
Premix	0.25
Lysine	0.10
Methionine	0.20
Total	100.00
Nutrient content	
Metabolizable energy (kkal/kg)**	3034.58
Crude protein (%)*	21.73
Crude fat (%)*	3.40
Crude Fiber (%)*	4.42
Calcium (%)*	1.444
Phosphorus (%)*	0.73

Source: *Results of proximate and mineral analysis at the Animal Nutrition Science Laboratory, Faculty of Animal Husbandry and Agriculture, Diponegoro University (2024). ** Metabolizable energy is calculated using the Bolton formula (1967).

Methods

Preparation begins with the preparation of soybean meal extract based on the method of Krismaputri *et al*. (2016). The solvent used distilled water and ethanol in a ratio of 1:3 (v/v). Soybean meal flour was dissolved with ethanol in a ratio of 1:5 (b/v). The material was heated using a waterbath at 80°C, and stirred for 30 minutes then filtered to take the filtrate, then deposited in the refrigerator at 5°C for 24 hours. The precipitate was dried and then pulverized into flour. The amount of *Lactobacillus plantarum* given to chickens was 1 x 10^{10} log cfu/g.

The rearing stage of 192 broiler chickens lasted for 35 days. The rearing process begins with the process of chick in DOC whose weight is weighed as the initial weight of maintenance, then put in a cage per partition of 20 birds.

The study was conducted using a completely randomized design (CRD) with 6 treatments and 4 replications, each experimental unit was filled with 8 birds. The treatments that will be applied are as follows:

T_0: basal ration
T_1: basal ration + *Lactobacillus plantarum* 1.2%
T_2: basal ration + SME 0.15%
T_3: basal ration + SME 0.30%
T_4: basal ration + SME 0.15% + *Lactobacillus plantarum* 1.2%
T_5: basal ration + SME 0.30% + *Lactobacillus plantarum* 1.2%

The treatment (*Lactobacillus plantarum* or SME or SME+ *Lactobacillus plantarum*) is given every morning by giving 20% of the ration consumption, if the ration has run out, the basal ration is given without treatment.

Data collection in this study included fat digestibility, abdominal fat weight, meat fat mass, and carcass weight. Calculation of fat digestibility begins with the total collection of excreta in the final week of maintenance which is carried out for 4 days. The excreta collected during the total collection treatment was sprayed using 0.1 N HCL every 2 hours to prevent nitrogen evaporation. The excreta that has been collected is cleaned of dirt and chicken feathers and then weighed to determine the wet weight of the excreta and then dried in the sun and weighed to determine the dry weight of the excreta. The dried excreta was then homogenized using a blender, then the excreta was used as a test sample for fat content using a soxhlet with the aim of knowing the fat content in the excreta. Fat digestibility was calculated using the formula of Moningkey *et al*. (2019) as follows:

$$\text{Fat Digestibility (\%)} = \frac{\text{(fat intake-amount of excreta fat)}}{\text{Fat intake}} \text{x } 100\%$$

Abdominal fat is measured by weighing the fat in the abdominal cavity and digestive organs including the small intestine, gizzard, abdominal tissue and abdominal cavity wall and then calculated by the formula of Krismiyanto *et al*. (2020) as follows:

$$\text{RWAF} = \frac{\text{abdominal fat weight (g)}}{\text{live weight (g)}} \times 100\ \%$$

Where: RWAF = Relative weight of abdominal fat.

Meat fat mass can be determined from the calculation of meat fat content and meat weight. Meat fat content is measured using a soxhlet device with meat samples on the breast and thigh that have been mashed. Meat weight is obtained through weighing at the time of chicken slaughter. Meat fat mass is calculated by the formula according to Mentari *et al*. (2014) as follows:

Meat fat mass (g) = meat fat content (%) x meat weight (g)

The carcasses measured were chicken body parts without blood, feathers, legs, head, neck and all contents of the abdominal cavity except liver, gizzard and heart. The resulting carcasses were weighed using a digital scale.

Data were analyzed using analysis of variance (ANOVA) applying SPSS 16.0 statistical software. Duncan's multiple range tests examined the differences between the treatments′ means. The significance level was $p<0.05$.

RESULTS AND DISCUSSION

Fat digestibility

The results of analysis of variance showed that the addition of soybean meal extract (SME) and *Lactobacillus plantarum* in the ration had a significant effect ($p<0.05$) on the fat digestibility of broiler chickens (Table 2). Treatment T_5 showed the lowest results ($p<0.05$) compared to other treatments due to the oligosaccharide content in SME which is food for LAB. The amount of *Lactobacillus plantarum* given can increase the population of beneficial bacteria by utilizing substrates from SME through fermentation to produce metabolite products such as short chain fatty acids (SCFA) and lactic acid. The increased LAB population can produce the enzyme bile salt hydrolase (BSH). The BSH enzyme works in the process of bile salt deconjugation, causing conjugated bile salts to be converted into deconjugated bile salts. BSH enzymes deconjugate bile salts so that fat is less absorbed by the small intestine (Kirana *et al*., 2017). Bile salts that return to the liver during enterohepatic circulation are reduced so that the total fat in the body is reduced. Krismiyanto *et al*. (2021) stated that lactic acid bacteria produce BSH enzymes that can deconjugate bile salts so that fat cannot be absorbed and is wasted with excreta.

The T_4 treatment showed the digestibility of crude fat was not significantly different ($p>0.05$) with the T_3 and T_1 treatments due to the addition of SME and *Lactobacillus plantarum* at that level has not been able to produce enough substrate to increase the LAB population optimally. Krismiyanto *et al*. (2023) stated that prebiotics added to the ration can produce SCFA, changing the acidic atmosphere in the digestive tract so that the LAB population increases and pathogenic bacteria decrease. However, the T_4 treatment showed a significant difference ($p<0.05$) with the T_5 treatment due to the lower amount of SME addition. Fat digestibility can decrease influenced by the addition of SME which potentially contains soybean oligosaccharides which are utilized by LAB to lower pH and inhibit the work of lipase. Harumdewi *et al*. (2018) stated that soybean meal containing SOS can be utilized by LAB and increase SCFA to produce a lower pH in the digestive tract which reduces the performance of lipase enzymes so that fat digestibility decreases.

Treatment T_4 with the addition of 0.15% SME and 1.2%

Table 2. Fat digestibility, meat fat mass, relative weight of abdominal fat and carcass weight.

Treatments	T0	T1	T2	T3	T4	T5
Fat digestibility (%)	79.96[a]	78.16[ab]	79.34[a]	77. 9[ab]	74.96[b]	68.27[c]
Meat fat mass (mg)	12.2	11.6	11.4	11.5	11.3	10.5
Relative weight of abdominal fat (%)	1.02[a]	0.96[ab]	0.96[ab]	0.86[b]	0.69[c]	0.67[c]
Carcass Weight (g)	863.00[d]	893.25[cd]	927.00[c]	1019.00[b]	1051.50[b]	1096.50[a]

Different superscripts on mean values indicate significant differences ($p<0.05$).

Lactobacillus plantarum produced lower fat digestibility than treatments T_0 and T_2. The addition of SME 0.15% and *L. plantarum* 1.2% has a connection that SME is utilized as a substrate for *Lactobacillus plantarum*, thus producing metabolite products and a high LAB population. Because the T_2 treatment (SME 0.15%) or single addition has not been able to increase the endogenous LAB population. The results of research by Rochman *et al*. (2019) stated that the addition of a level of 0.3% combination of dayak onion bulb extract or *Lactobacillus acidophilus* was able to reduce crude fat digestibility in broiler chickens. The combination of prebiotics and probiotics can create an acidic atmosphere in the digestive tract, this contributes to the growth of LAB compared to single addition because if the addition of substrate without the addition of LAB will not cause the fermentation process and does not produce SCFA enzymes.

The T_0 and T_2 treatments were not significantly different ($p>0.05$) from the T_1 and T_3 treatments due to the addition of *Lactobacillus plantarum* alone or not together. The same pH environment and LAB population support the same amount of BSH production. The relationship between pH, LAB and BSH production affects fat absorption. The T_0 treatment does not have the addition of SME or *Lactobacillus plantarum*, but the T_0 ration contains soybean meal, it is also possible that oligosaccharides can be utilized although not as optimally as SME. Oligosaccharides in soybean meal cannot be optimally utilized because it does not go through the extraction process. Extraction is the process of separating a substance from its mixture using an appropriate solvent (Abubakar *et al*., 2020). The extraction process is carried out so that it is easier for livestock to absorb the nutrient content in soybean meal because the nutrient content is maximized. The addition of SME or *Lactobacillus plantarum* alone does not maximize LAB in producing BSH, resulting in high fat absorption. A high LAB population can increase the production of BSH enzymes to deconjugate bile salts so that the digestibility and absorption of fat decreases (Krismiyanto *et al*., 2023).

Relative weight of abdominal fat

The results of the analysis of variance of the addition of SME and *Lactobacillus plantarum* to the ration had a significant effect ($p<0.05$) on the relative weight of abdominal fat of broiler chickens. T_4 and T_5 treatments were significantly different ($p<0.05$) lower than T_0, T_1, T_2 and T_3 treatments. This is in line with low fat digestibility and carcass fat. Low fat digestibility affects the fat deposited in the form of adipose fat or as low meat fat mass so that the relative weight of the abdominal fat produced is low. Abdominal fat has a positive correlation with total carcass fat, the higher the abdominal fat content, the higher the broiler carcass fat content and vice versa (Salam *et al*., 2017). Although the T_4 and T_5 treatments were significantly different from the other treatments, the T_4 treatment was not significantly different ($p>0.05$) from the T_5 treatment. Pathogenic bacteria cannot survive at acidic pH, so it can increase the number of LAB in the intestine. A decrease in pH can inhibit the growth of pathogenic bacteria such as *Salmonella*, because pathogenic bacteria tend not to survive in acidic conditions (Adisti *et al*., 2018). A high LAB population can increase the production of BSH enzymes to deconjugate bile salts so that absorbed fat decreases. An increase in LAB results in an increase in the enzyme bile salt hydrolase, which plays a role in deconjugating bile salts, resulting in a decrease in fat absorption (Istiqomah *et al*., 2020).

The T_3 treatment was not significantly different ($p>0.05$) with the T_1 and T_2 treatments, this is in accordance with the supporting data showing the LAB population, coliform, and pH in the small intestine showed results that were not significantly different. The raffinose and stakiose compounds contained in SME are not effectively utilized by beneficial bacteria in the digestive tract to produce lactic acid. Dwiputra *et al*. (2020) stated that lactic acid bacteria can cause the pH condition of the digestive tract to become acidic and inhibit the function of the lipase enzyme in digesting fat. The provision of SME with a level of 0.30% which is not balanced with the provision of *Lactobacillus plantarum* has not been able to produce enough BSH enzymes so that fat absorption is low. Low fat absorption can bring fat to be deposited into tissues or organs, so that not much fat is stored in the form of abdominal fat. Kirana *et al*. (2017) stated that high LAB populations produce BSH enzymes that can deconjugate bile salts which have low fat emulsion effectiveness so that fat is wasted with excreta and fat deposition decreases.

Meat fat mass

The results of analysis of variance showed that the addition of SME and *Lactobacillus plantarum* in the ration had no significant effect (p>0.05) on broiler meat fat mass (Table 2). The results of the analysis of the addition of SME and *Lactobacillus plantarum* had no significant effect (p>0.05) on broiler meat fat mass can be assumed due to the influence of endogenous lactic acid bacteria (LAB) in the digestive tract of chickens. Lactic acid bacteria are known to produce natural statins that can inhibit fat formation in the liver. Cavallini *et al*. (2009) stated that LAB produce statins, which are 3-hydroxy-3-methyl-glutaryl-KoA reductase (HMG-KoA reductase) inhibitors that work in the biosynthesis of fat, cholesterol, blood lipoproteins and blood triglycerides. Natural statins can reduce the activity of the enzyme acetylcoa carboxylase (ACC) which works in the synthesis of fatty acids in the liver. Ulupi and Sumantri (2015) stated that the ACC enzyme in the liver works in converting acetyl-CoA into malonyl-CoA, which is then synthesized into palmitate (long chain fatty acid). If ACC enzyme activity decreases, fatty acid synthesis in the liver also decreases.

The addition of SME did not significantly affect the fat mass of broiler meat. Krismaputri *et al*. (2016) stated that the addition of prebiotic soybean oligosaccharide (SOS) from soybean meal extract and soybean skin extract at the level of 0.15 and 0.3% had no significant effect (p>0.05) on meat fat mass in broiler chickens. The absorbed fat is mostly stored in the form of abdominal fat (Table 2). Fat deposition in meat tends to be the same and does not result in decreased meat fat quality. Fat in meat comes from excess feed energy that is stored in the form of fat (Fouad and El-Senousey, 2014). This study shows that fat reserves contained in the body are stored in abdominal fat, so the mass of meat fat has no significant effect. Factors that influence the high and low fat content of meat are influenced by several factors. Fat content in meat is influenced by genetics, sex, and age of the animal (Hidayati *et al*., 2016).

Carcass weight

The results of the analysis of variance of the addition of SME and *Lactobacillus plantarum* to the ration had a significant effect (p<0.05) on the carcass weight of broiler chickens (Table 2). Treatment T_5 was significantly (p<0.05) higher than treatments T_0, T_1, T_2, T_3, and T_4. This was influenced by the relative weight of abdominal fat which was lower than the other treatments. Low abdominal fat proves that the nutrients absorbed by the body into meat are higher. Abdominal fat has a close relationship with carcass weight, if abdominal fat decreases then carcass weight increases due to the absence of excess energy so that there is no accumulation of abdominal fat and vice versa (Pahlepi *et al*., 2015). The carcass weight produced is in line with the relative weight of abdominal fat produced as well. The lower the relative weight of abdominal fat, the better the carcass produced (Wang *et al*., 2022).

Treatment T_4 was not significantly different (p>0.05) from treatment T_3, but treatment T_4 was significantly different (p<0.05), higher than treatments T_0, T_1, and T_2. This is because the amount of LAB and pH in these treatments are the same. The SCFA content produced as a result of SOS prebiotic fermentation by LAB in the digestive tract of chickens can improve intestinal morphology to create an acidic condition. The acidic condition in the digestive tract results in limited lipase enzyme activity, resulting in decreased fat digestion and body fat formation (Syafrizal *et al*., 2018). High SCFA content can reduce pH in the small intestine, thereby increasing protein digestibility and affecting the increase in broiler carcass weight (Setyoko *et al*., 2020).

The T_2 treatment was not significantly different (p>0.05) from the T_1 treatment, but the T_2 treatment was significantly different (p<0.05), higher than the T_0 treatment. Protein is a nutrient needed by livestock for body tissue growth. Protein contained in the body has been absorbed by the body, so the body will form meat (Saputra *et al*., 2016). High protein digestibility causes the live weight of broilers to increase, thereby increasing carcass weight. High ration protein digestibility so that meat growth is also maximized. Rochman *et al*. (2019) stated that the addition of a combination of noni fruit extract and *Lactobacillus acidophilus* in broiler rations can increase protein digestibility to 84.44%. Optimal protein absorption has an impact on livestock productivity, especially on their high carcass weight and low fat.

The T_0 treatment was not significantly different (p>0.05) from the T_1 treatment, but the T_0 treatment was significantly different (p<0.05), lower than the T_2, T_3, T_4 and T_5 treatments. This was due to the absence of probiotics, so the bacteria in the T_0 treatment did not work optimally in digesting and absorbing nutrients. Probiotics can improve the performance of digestive enzymes in breaking down and absorbing nutrients, so that they are used for tissue growth and increasing body weight (Astuti *et al*., 2015).

Conclusion

The addition of 0.3% soybean meal extract and 1.2% *Lactobacillus plantarum* to the diet can reduce fat digestibility, relative weight of abdominal fat and increase carcass weight and produce the same meat fat mass. The addition of soybean meal extract and *Lactobacillus plantarum* can be applied on an industrial scale livestock farming.

CONFLICT OF INTEREST

The authors declare no competing interests.

REFERENCES

Abdurrahman, Z. H., Pramono, Y. B., & Suthama, N. (2016). Meat characteristic of crossbred local chicken fed inulin of dahlia tuber and Lactobacillus sp. *Media Peternakan*, 39(2), 112-118.

Abubakar, A. R., & Haque, M. (2020). Preparation of medicinal plants: Basic extraction and fractionation procedures for experimental purposes. *Journal of Pharmacy and Bioallied Sciences*, 12(1), 1-10.

Adisti, R., Setiawan, D., Zakiatulyaqin, Z., & Purnomosidi, M. (2018). The effect of native orange water (*Citrus microcarpa*) in drink water on broiler meat fat. Scientific Journal Encompassing All Area of Animal Science, 18(1), 18-23.

Astuti, F. K., Busono, W., & Sjofjan, O. (2015). Effect of adding liquid probiotics to feed on production performance in broilers. Indonesian *Journal of Environment and Sustainable Development*, 6(2), 99-104.

Boğa, M., Avcı, B. C., & Kılıç, H. N. (2022). Evaluation of some commercial food rations in terms of chemical composition, methane production, net energy and organic substance digestibility. *Turkish Journal of Agriculture-Food Science and Technology*, *10*(6), 1095-1101.

Bolton, W. (1967). *Poultry nutrition*. HMSO, London. MAFF Buletin. No 174.

Cavallini, D. C., Bedani, R., Bomdespacho, L. Q., Vendramini, R. C., & Rossi, E. A. (2009). Effects of probiotic bacteria, isoflavones and simvastatin on lipid profile and atherosclerosis in cholesterol-fed rabbits: a randomized double-blind study. *Lipids in Health and Disease*, *8*, 1-8.

Dwiputra, R., Adriani, L., & Permana, R. (2020). The effect of probiotic addition based on milk, soy milk and mung bean milk in the ransum on cholesterol and triglyceride blood of broiler chicken. *Journal of Tropical Animal Nutrition Feed Science*, *2*(3), 135-144.

Fouad, A. M., & El-Senousey, H. K. (2014). Nutritional factors affecting abdominal fat deposition in poultry: a review. *Asian-Australasian Journal of Animal Sciences*, *27*(7), 1057-1068.

Harumdewi, E., Suthama N. & Mangisah I. (2018). Effect of feeding microparticles protein diet and probiotics on fat digestibility and meat fat deposition in broiler chicken. *Jurnal Sain Peternakan Indonesia*, *13*(3), 258-264.

Hidayati, N. N., Yuniwarti, E. Y. W., & Isdadiyanto, S. (2016). Comparison of meat quality of magelang duck, pengging duck and tegal duck. *Bioma*, 18(2), 56-63.

Istiqomah, L., Sakti, A. A., Sofyan, A., Herdian, H. & Anggraeni, A. S. (2020). Cholesterol-lowering activity of lactic acid bacteria and yeast when used as probiotics in laying quail (*Coturnix coturnix Japonica*). *Journal of the Indonesian Tropical Animal Agriculture*, *45*(4), 305-319.

Kirana, N. G. P. S., Bidura, I. G. N. G., & Puspani E. (2017). Effect of using *Saccharomyces* sp. fermented tofu pulp in rations on broiler blood fat and cholesterol distribution. Journal of Tropical Animal Science, *1*(1), 105-119.

Krismaputri, M. E., Suthama, N., & Pramono, Y. B. (2016). Pemberian *soybean oligosaccharides* dari ekstrak bungkil dan kulit kedelai terhadap pH usus, populasi *E. coli*, dan PBBH pada broiler. *Mediagro*, 12(2), 20-25.

Krismiyanto, L., Suthama N. & Mangisah I. (2020). Pemanfaatan sumber minyak berbeda terhadap kecernaan lemak dan kualitas daging ayam broiler. *Journal of Tropical Animal Science and Technology, 7*(1), 77-81.

Krismiyanto, L., Suthama, N., & Wahyuni, H. I. (2021). Population of small intestine bacteria and performance of native chicken-leghorn crossbreed duo to feeding of dahlia tuber extract). *Scientific Journal Encompassing All Area of Animal Science,* 21(2), 157-164.

Krismiyanto, L., Yunianto, V. D., Mangisah, I., & Crismawati, R. (2023). Feeding rations supplemented with acidifiers, inulin and papain enzymes on fatty meat and carcass weight of grower-phase male turkeys. *Journal of Tropical Animal Production,* 24(2), 71-83.

Mentari, A. S., Mahfudz, L. D., & Suthama, N. (2014). Protein and fat meat mass in broiler chikens which given finggerroot (*Boesenbergia pandurata* ROXB.) powder in ration. *Animal Agriculture Journal*, 2(4), 148-160.

Moningkey, A. F., Wolayan, F. R., Rahasia C. A., & Regar, M. N. (2019). Digestibility of organic matter, crude fiber and crude fat in broiler diets fed with pumpkin (*Cucurbita moschata*) waste meal. *Animal Science Review,* 39(2), 257-265.

Pahlepi, R., Hafid H., & Indi, A. (2015). Final weight of carcass percentage and abdominal fat of broiler chickens with betel leaf extract (*Piper betle* L.) in drinking water. *Journal of Tropical Animal Sicence and Technology*, 2(3), 1-7.

Rochman, F., Yuanita, I., Wahyuni, H. I. & Suthama, N. (2019). Effect of dayak onion bulb extract (*Eleutherine palmifolia*) combined with probiotics (*Lactobacillus acidophilus*) on fatty liver in broilers. Seminar Nasional Pengelolaan Sumber Daya Alam Berkesinambungan di Kawasan Gunung Berapi. Pp. 1-4.

Salam, S., Fatahilah A., Sunarti D. & Isroli I. (2017). Carcass weight and abdominal fat of broiler chickens fed black cumin (*Nigella sativa*) meal in the diet during the summer season. *Sains Peternakan*, 11(2), 84-90.

Salehizadeh, M., Modarressi, M. H., Mousavi, S. N., & Ebrahimi, M. T. (2019). Effects of probiotic lactic acid bacteria on growth performance, carcass characteristics, hematological indices, humoral immunity, and IGF-I gene expression in broiler chicken. *Tropical Animal Health And Production*, *51*, 2279-2286.

Saputra, Y. A., Mangisah I. & Sukamto B. (2016). Effect of onion skin meal addition on crude protein digestibility, body weight gain and carcass percentage of Mojosari ducklings. *Indonesian Journal of Animal Science*, 26(1), 29-36.

Setyoko, H., Sukamto, B., Wahyono, F., & Krismiyanto, L. (2020). Crude fat digestibility and carcass weight of broiler chickens due to the addition of noni fruit extract (*Morinda citrifolia* L.) and *Lactobacillus acidophilus*. E-Prosiding Seminar Nasional Ilmu Peternakan Terapan. Pp. 134-142.

Syafrizal, S., Nurliana N. & Sugito S. (2018). The administration effect of soybean dregs and palm kernel meal fermented with *Aspergillus niger* on fat and cholesterol levels of broiler meat. *Scientific Journal Encompassing All Area of Animal Science*, *18*(2), 74-82.

Tolnai, E., Fauszt, P., Fidler, G., Pesti-Asboth, G., Szilagyi, E., Stagel, A., Konya, J., Szabo, J., Stundl L., Babinszky, L., Remenyik, J., Biro, S., & Paholcsek, M. (2021). Nutraceuticals induced changes in the broiler gastrointestinal tract microbiota.

American Society for Microbiology, 6(2), 11-24.
Ulupi, N., & Sumantri, C. (2015). The role of triglyceride lipase, fatty acid synthase and fatty acid binding protein gene groups on broiler fat metabolism. *Wartazoa*, *25*(1), 15-22.
Wang, R. L., Chen, J. W., Gooneratne, R., He, X. P., Huang J. Z., & Zhao, Z. H. (2022). Effects of varied molecular weight of chitosan oligosaccharides on growth performance, carcass trait, meat quality, and fat metabolism in indigenous yellow-feathered chickens. *Journal of Applied Poultry Research, 31*(1), 1-10.
Wijaya. Y., Suprijatna E. & Kismiati S. (2017). Utilization of herbal industry waste and lactic acid bacteria (*Lactobacillus* sp.) as synbiotic for interior quality of laying hen eggs. Indonesian *Journal of Animal Science,* 19(2), 47- 54.
Winarti, W., Mahfudz L. M., Sunarti D. & Setyaningrum S. (2019). Weight of proventriculus, gizzard, caecum, rectum and length caecum and rectum of broilers due to addition ofsynbiotic from inulin extract of gembili tuber and *Lactobacillus plantarum* in feed. *Journal of Agricultural Sciences and Veterinery, 8*(2), 151-164.