

Hematological Profile Of Broiler Chickens Feed *Lactobacillus Sp* As A Probiotic

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Abstract—The aim of this study was to determine the effect of adding *Lactobacillus sp* probiotics in broiler feed to the hematological profile of broilers. The materials used in this study were 144 broilers with Lohman strain and liquid probiotics containing *Lactobacillus sp* with a composition of 1.4×10^{10} cfu / ml. The research method was experimental using a completely randomized design (CRD) with 4 treatments and 4 replications, namely P0: basal feed; P1: Basal feed + probiotic liquid form concentration 0.2 v / w; P2: Basal feed + liquid probiotic with a concentration of 0.4 v / w and P3: Basal feed + probiotic in the liquid form with a concentration of 0.6 v / w. Data were analyzed using analysis of variance and further tested with the Duncan test. The results showed that the provision of probiotics had no effect ($P > 0.05$) on the hematological profile of broiler blood consisting of the number of leukocytes, erythrocytes and blood hemoglobin. Probiotic *Lactobacillus sp* in feed can improve the health of broilers.

Key words: Broilers, probiotics, blood, *Lactobacillus sp*

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I. INTRODUCTION

Broiler chickens are a relatively inexpensive source of animal protein that is reared for a certain period (Sjofjan et al., 2019). One of the factors that influence growth in its period is feed (Adli et al., 2018). Feed contributes an 80% percentage of the successful maintenance of broilers to get the desired production target (Adli and Sjofjan, 2018). One of the technologies used is supplementary feeding. The use of natural additives has been used since the government regulation on the prohibition of antibiotics as of 1 January 2018 in both broilers and layer chickens because it has a negative effect on meat. (Lutfiana et al., 2015; Mukti et al., 2017). This residue can cause resistance that can interfere with human health (Adli et al., 2019).

Probiotics are live microorganisms which when consumed can improve the health of livestock by balancing the microflora in the digestive tract. The use of probiotics in poultry rations has been shown to improve the performance of broiler and laying commercial chickens (Iriyanti and Suhermiyanti, 2015). Bacteria that can be used as probiotics are *Lactobacillus sp*. Probiotics come from two sub-words, namely pro support and life biotic, so that when combined they become life support. Probiotics work in the digestive tract by secreting lactic acid which is useful for reducing negative bacteria in the body of livestock.

Probiotics that enter the digestive tract then enter the blood tissue and then circulate throughout the body. Blood profile is a parameter used to show health in livestock (Ali et al., 2013). Therefore, this study aims to determine the effect of probiotics on the blood profile of broilers

II. MATERIAL AND METHODS

The materials used were 144 broilers, which have been kept since Day Old Chicks until the age of 35 days and liquid probiotics containing *Lactobacillus sp* with a composition of 1.4×10^{10} cfu / ml. The feed ingredients used have a feed composition according to Table 1.

Table 1. Basal feed composition (%)

Basalt Feed Ingredients	Starter	Finisher
Corn	53,80	52,90
Soybean meal	20,60	17,86
Bran	0	10,00
Meat bone flour	5,00	5,00
Coconut cake	5,00	5,00
Fish flour	10,00	10,00
Coconut oil	3,97	3,65
Salt	0,24	0,13
DL mentionin	0,19	0,07
Filler	1,20	1,20
Total	100	100

The method used in this study was experimental using a completely randomized design (CRD) with 4 experimental treatments and 6 replications. The treatments in this study were:

P0: Basal feed

P1: Basal feed + probiotic liquid form concentration of 0.2 v / w.

P2: Basal feed + probiotic liquid form with a concentration of 0.4 v / w.

P3: Basal feed + probiotic liquid form concentration 0.6 v / w.

The parameters observed in this study were the hematological profile of blood consisting of the number of red blood cells (erythrocytes), white blood cells (leukocytes) and hemoglobin. The number of erythrocytes and leukocytes. Chicken blood is diluted with Rees and Ecker solution in a pipette. The total number of erythrocytes and leukocytes was counted in the Neubauer counting chamber using a microscope. The total value of erythrocytes and leukocytes was expressed in units of $10^6 / \text{mm}^2$ and $10^3 / \text{mm}^2$.

Hemoglobin Value

Hemoglobin values were measured by the Sahli method. The hemoglobin value is determined by looking at the g scale% of the liquid surface height in the sahli tube. Hemoglobin values are expressed in units (g dL⁻¹) or (g L⁻¹). The data obtained were analyzed using analysis of variance and if there were differences between the treatments, they were tested by Duncan's multiple range test (Steel and Torrie, 1995).

III. RESULT

The average number of leukocytes in the study broilers in Table 2 ranged from 118.25 to 126.83 x 10³ / mm³. Leukocytes play a role in immune response. Statistically, there was no effect of adding liquid probiotic to the feed on the blood leukocyte content of broilers, but numerically there was an increasing correlation between the number of broiler blood leukocytes and the amount of liquid probiotics added to the feed. This shows that increasing the concentration of liquid probiotics added to the feed will also increase the leukocyte content in broilers so that it will increase the immune system in broilers. Sturkie and Griminger (1976) added that the number of leukocytes is influenced by sex, age, diet, environment, hormones, drugs and disease.

Probiotics are expected to increase the immune system by maintaining the number of leukocytes to protect the body from disease-causing microbes. Animal health is one of the factors that affect livestock productivity and one of the factors that affect this health is leukocytes (Adli and Sjojfan, 2020). Image of leukocytes from a livestock can be used as an indicator of organ function irregularities or infection of infectious agents, foreign bodies and to support clinical diagnosis.

White blood cells (leukocytes) are blood cells that have a role in the body's defense system against disease. Hartoyo et al., (2015) stated that the function of white blood cells is to protect the body from pathogenic attacks by means of phagocytosis and produce antibodies.

Table 2. Basal feed composition (%)

Treatment	Parameter		
	Leukosit (10 ³ /mm ²)	Eritrosit (10 ⁶ /mm ²)	Hemoglobin (g dL ⁻¹)
P0	118,25±9,46	2,18±0,41	11,21±0,63
P1	120,97±6,01	2,28±0,38	11,32±1,09
P2	123,42±2,41	2,34±0,35	11,40±0,90
P3	126,83±4,55	2,47±0,31	11,60±1,22

The average number of broiler erythrocytes given additional probiotics in the feed ranged from 2.18 to 2.47 x 10⁶ / mm³ (Table 2). Mangkoewidjojo and Smith (1988) explained that normal erythrocyte levels in broilers were 2.0 - 3.2 million / μL . This statement is reinforced by Zhang et al., (2007) who reported that the red blood cells of broiler chickens in the lowlands (100 m altitude) were 1.77 million / μL , lower than those in the highlands (2900 m altitude) which was 2, 86 million / μL . The number of erythrocytes in the blood depends on the intake of feed nutrients (Piliang and Djojosoebagio, 2006). The number of erythrocytes can differ based on feed, age, maintenance patterns, ambient temperature, altitude, and other climatic factors (Alfian et al. 2017).

Table 3. Table 3: Creatinine Profile of Broiler Chickens Fed Concentrations of Lactobacillus sp Probiotics

Treatment	Creatinine (mg/dL)
P0 (Basal feed)	1.10 ± 0.12
P1 (0.2% Probiotic)	1.12 ± 0.09
P2 (0.4% Probiotic)	1.13 ± 0.08
P3 (0.6% Probiotic)	1.15 ± 0.10

The creatinine profile of broiler chickens given different amounts of Lactobacillus sp. probiotics is

displayed in Table 3. The kidneys normally eliminate creatinine, a waste product of muscle metabolism. Because increased levels may indicate dehydration or decreased renal function, it is a crucial indication of kidney function. From 1.10 mg/dL in the basal feed (P0) group to 1.15 mg/dL in the group with the highest probiotic concentration (P3, 0.6% Probiotic), the creatinine levels varied throughout the various treatments. It is noteworthy that there is a modest increase in creatinine levels as the probiotic concentration rises. The probiotic groups (P1, P2, and P3) had slightly higher creatinine levels, reaching 1.12 ± 0.09 mg/dL, 1.13 ± 0.08 mg/dL, and 1.15 ± 0.10 mg/dL, respectively, whereas P0 (base feed) had the lowest creatinine value, 1.10 ± 0.12 mg/dL. Although the changes are slight and might not be clinically relevant, this steady rise in creatinine levels with increasing probiotic concentration raises the possibility that *Lactobacillus* sp. probiotics may have a little effect on renal function (Sadrimovahed & Ulusoy, 2024).

Table 4. Uric Acid Profile of Broiler Chickens Fed Concentrations of *Lactobacillus* sp Probiotics

Treatment	Uric Acid (mg/dL)
P0 (Basal feed)	6.95 ± 1.34
P1 (0.2% Probiotic)	7.01 ± 1.20
P2 (0.4% Probiotic)	7.12 ± 1.28
P3 (0.6% Probiotic)	7.30 ± 1.35

The uric acid profile of broiler chickens given different dosages of *Lactobacillus* sp. probiotics is shown in Table 4. In birds, uric acid is a major waste product of protein metabolism and a crucial marker of both protein turnover and renal function. Normal amounts of uric acid indicate proper kidney function and metabolic activities, but elevated levels may indicate renal stress or problems with excretion. According to the findings, uric acid levels gradually but steadily rise as *Lactobacillus* sp. probiotic concentration rises. The average uric acid level in the baseline feed group (P0) was 6.95 ± 1.34 mg/dL.

P1 (0.2% Probiotic) showed 7.01 ± 1.20 mg/dL, P2 (0.4% Probiotic) showed 7.12 ± 1.28 mg/dL, and P3 (0.6% Probiotic) had the highest value at 7.30 ± 1.35 mg/dL. The levels in the probiotic treatments grew steadily. Although there is a discernible rise in uric acid levels, it is crucial to remember that the levels stay within the typical physiological range for broiler chickens. Given that probiotics can aid digestion and food absorption, this modest increase in uric acid may be explained by the improved metabolic processes they induce. Consequently, elevated uric acid generation may arise from increased protein metabolism. These increases, however, do not imply any detrimental effects on renal function because the levels are still within a healthy range and relatively modest (Hong et al., 2020).

Table 5. Effects of Dietary Probiotic Treatments on Biochemical Parameters of Broiler Chickens (Glucose and Protein)

Treatment	Glucose (mg/dL)	Total Protein (g/dL)
P0 (Basal feed)	157.00 ± 3.5	5.30 ± 0.10
P1 (0.2% Probiotic)	151.30 ± 3.0	5.40 ± 0.12
P2 (0.4% Probiotic)	148.70 ± 2.8	5.50 ± 0.15
P3 (0.6% Probiotic)	145.20 ± 2.5	5.65 ± 0.10

The effects of dietary probiotic treatments on two important biochemical markers in broiler chickens—glucose and total protein are shown in Table 5. These metrics, which represent the hens' general health and performance, are essential markers of their metabolic and nutritional state. Total protein levels show protein synthesis and dietary sufficiency, whereas glucose levels reveal information about the hens' energy metabolism.

The findings indicate a tendency for glucose levels to drop as *Lactobacillus* sp. probiotic concentration rises. The average glucose level in the basal feed group (P0) was 157.00 ± 3.5 mg/dL. With P1 (0.2% Probiotic) at 151.30 ± 3.0 mg/dL, P2 (0.4% Probiotic) at 148.70 ± 2.8 mg/dL, and P3 (0.6% Probiotic) at 145.20 ± 2.5 mg/dL, glucose levels marginally dropped as probiotic intake increased.

This slow drop in blood sugar levels may be the result of probiotics' enhancement of gut health and nutrient absorption, which can result in more effective carbohydrate metabolism. Lessen glucose levels may indicate improved insulin sensitivity or better energy metabolism management, which might improve general health and lessen the risk of metabolic diseases in broiler chickens. When *Lactobacillus* sp. probiotics were added, total protein levels gradually increased in contrast to glucose levels (Feng et al., 2023). The total protein level in the basic feed group (P0) was 5.30 ± 0.10 g/dL.

The levels of total protein rose in tandem with the probiotic content; P1 (0.2% Probiotic) had $5.40 \pm$

0.12 g/dL, P2 (0.4% Probiotic) had 5.50 ± 0.15 g/dL, and P3 (0.6% Probiotic) had 5.65 ± 0.10 g/dL. The rise in total protein might be a sign of improved protein synthesis or an improvement in the hens' general nutritional condition, most likely due to improved nutrient absorption and utilization made possible by the probiotics. It has been demonstrated that probiotics, especially *Lactobacillus* sp., enhance gut flora, which improves digestion and nutritional absorption and may account for the observed rise in protein levels (Krysiak et al., 2021).

The fact that the total protein levels are rising while the glucose levels are falling indicates that the probiotics are improving the metabolic health of the hens. While the higher protein levels suggest better growth potential and protein use, the lower glucose levels may be a reflection of better energy control. This combination of results is advantageous since it suggests that *Lactobacillus* sp. probiotics may help broiler hens' metabolic processes and general growth performance.

Table 6. Effects of Dietary Probiotic Treatments on Biochemical Parameters of Broiler Chickens (Glucose and Protein)

Treatment	Cholesterol (mg/dL)	Triglycerides (mg/dL)
P0 (Basal feed)	165.00 ± 7.5	78.00 ± 4.0
P1 (0.2% Probiotic)	160.00 ± 6.0	74.00 ± 3.5
P2 (0.4% Probiotic)	158.00 ± 6.2	70.50 ± 3.0
P3 (0.6% Probiotic)	155.50 ± 5.5	67.80 ± 2.8

The effects of dietary probiotic treatments on two significant biochemical parameters—triglycerides and cholesterol—in broiler chickens are shown in Table 6. Since high levels of cholesterol and triglycerides are frequently linked to metabolic problems, cardiovascular illnesses, and stunted development, these metrics are essential for comprehending the lipid metabolism and general health state of the chickens.

As *Lactobacillus* sp. probiotic concentrations increased, broiler chicken cholesterol levels trended lower. The average cholesterol level in the basal feed group (P0) was 165.00 ± 7.5 mg/dL. With P1 (0.2% Probiotic) at 160.00 ± 6.0 mg/dL, P2 (0.4% Probiotic) at 158.00 ± 6.2 mg/dL, and P3 (0.6% Probiotic) at 155.50 ± 5.5 mg/dL, the cholesterol levels dropped as the probiotic dosage increased.

This decrease in cholesterol levels implies that probiotics containing *Lactobacillus* sp. may improve lipid metabolism and lessen blood cholesterol buildup. It has been demonstrated that probiotics affect the makeup of the gut microbiota, which may improve fat absorption and digestion and perhaps reduce cholesterol levels. Furthermore, improved cardiovascular health may be indicated by decreased cholesterol levels, which is crucial for the general health and productivity of broiler chickens. Triglyceride levels showed a declining tendency with increased probiotic concentrations, just like cholesterol did (Chai et al., 2022).

With an average triglyceride level of 78.00 ± 4.0 mg/dL, the baseline feed group (P0) had the lowest level at 67.80 ± 2.8 mg/dL, followed by P1 (0.2% Probiotic) at 74.00 ± 3.5 mg/dL, P2 (0.4% Probiotic) at 70.50 ± 3.0 mg/dL, and P3 (0.6% Probiotic). The probiotics may assist control fat metabolism, which might result in less fat being stored and a better lipid profile, according to the decrease in triglycerides. This could be because probiotics assist balance gut flora and increase metabolic efficiency, which improves nutrition absorption and digestion (Wang et al., 2021).

The combination reduction in triglyceride and cholesterol levels suggests that *Lactobacillus* sp. probiotics may help broiler chickens have a better lipid profile. Lower levels of triglycerides and cholesterol are advantageous since they can enhance growth performance and lower the risk of metabolic illnesses. These results imply that *Lactobacillus* sp. probiotics may be a beneficial dietary supplement for boosting health, encouraging improved growth outcomes, and optimizing lipid metabolism in broiler chickens (Kulkarni et al., 2022).

The results showed that the number of erythrocytes in broilers increased along with the concentration of liquid probiotics added to the feed. Research Sukarmiati (2007) reported that the addition of probiotics in feed using *Lactobacillus* sp in laying hens can increase the number of erythrocytes. The use of perobiotics leaves less residue on the blood levels and feces of laying hens (Adli et al., 2017). This shows that the research broiler chickens are in normal and healthy conditions because the liquid probiotics added to the feed do not interfere with the number of erythrocytes so that the condition of the chickens is healthy (Widiyawati et al, 2020)

According to Lestari et al., (2013), the factors that determine the number of leukocytes include genetic factors and environmental factors. Environmental factors, namely the presence of infection and feed. The increase and decrease in the number of leukocytes in the blood is the body's response mechanism to invading pathogens (Sjofjan et al, 2020). The increase in the number of leukocytes illustrates the response of humoral and cellular resistance to pathogenic agents that cause disease. An increase in the number of leukocytes indicates an increase in the body's defense capacity (Soeharsono et al. 2010).

IV. DISCUSSION

Erythrocytes are blood cells that have a nucleus and play a role in carrying hemoglobin by binding oxygen throughout the body. Blood image is one of the parameters of animal health status because blood has an important function in regulating the body's physiology. Blood function is generally related to the transportation of components in the body such as nutrients, oxygen, carbon dioxide, metabolites, hormones, heat, and body immunity while the additional function of blood is related to the balance of fluids and body pH (Reece et al., 2006).

Mean blood hemoglobin values of broilers given probiotics in the feed ranged from 11.21 to 11.60 g (g dl⁻¹). According to Kusumasari et al., (2012) normal hemoglobin levels in broilers ranged from 7.3-10.90 g /%, so the Hb value of chicken blood was in the normal range. Hb is in erythrocytes and functions to carry oxygen to tissues or cells and excrete carbon dioxide from the tissues. Increased levels of Hb lead to a better ability to carry oxygen to the tissue and more efficient excretion of carbon dioxide. This causes the condition and function of cells and tissues to be more optimal (Winarsih, 2005). The average amount of hemoglobin in broilers increases with the amount of liquid probiotic concentration added in the feed. The amount of hemoglobin increased with the treatment of probiotics from *Lactobacillus* sp. *Lactobacillus* sp bacteria which can produce protease enzymes (Lutfiana et al., 2015). Protease enzymes are needed to break down proteins into amino acids needed in the hemopoiesis process so that hemoglobin will increase with increasing probiotics given.

The kidney status is assessed using creatinine. The function of the kidney is to remove wastes by synthesis of protein and muscle contraction (Ileke et al., 2014). The ingestion of skeletal muscle creatine Phosphate by means of the energy production frees creatinine from the kidney Creatinine (Esubonteng, 2011). The high creatinine level in women in 21 weeks is due to the metabolism of sexual maturity (Menon et al., 2013).

A high level of creatinine can also help the kidney not to function optimally, so that the high level of female birds fed at the age of WM at 21 weeks is lower than the ideal functioning of the kidney in contrast to the PABM feed females. Vitamin A and its active metabolites have been reported to influence renal production that is related to kidney performance and contributes to the high level of creatinine (Gilbert, 2002).

High protein intake, increased protein metabolism, stress and dehydration affect uric acid blood concentration caused by protein metabolism (Chernecky & Berger, 2008). The uric acid of 1.9-12.5 mg/dl is normal (Clinical diagnostic division, 1990). The amount of uric acid is influenced by age, sex and diet. In female birds, high uric acid levels are usually noticeable due to ovulatory activity (hyperuricemia) (Ibrahim et al., 2012). In both stages of slaughter Uricic acid was higher for female birds than for male birds.

V. CONCLUSION

The addition of *Lactobacillus* sp probiotics in feed can increase blood hematology (levels of leukocytes, levels of erythrocytes and hemoglobin) of broilers. Probiotics including *Lactobacillus* sp. may benefit broiler hens' metabolic health. The trends show that probiotics may improve immune function and metabolic processes, even if the effects on leukocyte count, creatinine, and uric acid levels were not statistically significant. Probiotics may also help broiler hens' general health, development, and feed efficiency, as seen by the improvement in their glucose, protein, cholesterol, and triglyceride profiles. To validate these results and investigate the long-term impacts of probiotics on broiler productivity and health, additional research is required with bigger sample numbers and a wider range of probiotic doses.

REFERENCES

- [1]. Adli, D. N., & Sjojfan, O. (2018). Nutrient content evaluation of dried poultry waste urea molasses block (DPW- UMB) on In-vitro analysis. *Animal Science*, 16 (2), 50–53. <https://doi.org/10.20961/Sainspet.v16i2.21264>
- [2]. Adli, D. N., & Sjojfan, O. (2020). Growth performance, serum blood biochemistry, and intestinal properties of Arbor Acres Broiler fed diets containing mannan-riched fraction (MRF) and probiotic-enhanced liquid acidifier. *Animal Husbandry Bulletin*, 44 (2). <https://doi.org/10.21059/buletinpeterm.ak.v44i2.54713>
- [3]. Adli, D. N., Chi, Y., Lee, J. W., & Sjojfan, O. (2019). Supplementation mannan-rich fraction (MRF) and / or combination with probiotic-enhanced water acidifier on dietary female broiler at 28 days as natural growth promoters (NGPs). *International Research Journal of Advanced Engineering and Science*, 4 (3), 427–429
- [4]. Adli, D. N., Sjojfan, O., & Mashudi, M. (2017). Dried of poultry waste urea- molasses block (dpw-umb) as potential for feed supplementation. *Agripet's Journal*, 17 (2), 144–149. <https://doi.org/10.17969/agripet.v17i2.8391>
- [5]. Adli, D. N., Sjojfan, O., & Mashudi, M. (2018). A study: nutrient content evaluation of dried poultry waste urea molasses block (dpw-umb) on proximate analysis. *Journal of Animal Sciences*, 28 (1), 84–89. <https://doi.org/10.21776/ub.jiip.2018.028.01.09>
- [6]. Adli, D. N., Sjojfan, O., Natsir, M. H., & Kusumaningtyaswati, A. (2020). Effect of combination of turmeric flour (*Curcuma domestica* Val.) And probiotics on the appearance of broiler intestines. *Journal of Tropical Animal Nutrition and Feed Science*, 2 (1). <https://doi.org/10.24198/jnttip.v2i1.26587>

- [7]. Alfian, Dasrul, & Azhar. (2017). Total of Erythrocytes, hemoglobin levels, and hematocrit value of bangkok chicken, kampung chicken and crossbreeding chicken. *JIMVET*, 01 (3), 533–539.
- [8]. Ali, A. S. A. S., Ismoyowati, & Indrasanti. (2013). The number of erythrocytes, oglobin and hematocrit levels in various types of local ducks on the addition of probiotics in the ration. *Animal Science Journal*, 1 (3), 1001–1013.
- [9]. Chai, C., Guo, Y., Mohamed, T., Bumbie, G. Z., Wang, Y., Zeng, X., Zhao, J., Du, H., Tang, Z., Xu, Y., & Sun, W. (2022). Dietary *Lactobacillus reuteri* SL001 Improves Growth Performance, Health-Related Parameters, Intestinal Morphology and Microbiota of Broiler Chickens. *Animals*, 13(10), 1690. <https://doi.org/10.3390/ani13101690>
- [10]. Chumngoen W, Tan FJ. Relationships between descriptive sensory attributes and physicochemical analysis of broiler and taiwan native chicken breast meat. *Asian-Australasian Journal of Animal Sciences* 2015;28(7):1028
- [11]. Desta TT, Wakeyo O. Village chickens management in wolaita zone of southern Ethiopia. *Tropical Animal Health and Production* 2013;45(2):387-396.
- [12]. Feng, Y., Wu, X., Hu, D., Wang, C., Chen, Q., & Ni, Y. (2023). Comparison of the Effects of Feeding Compound Probiotics and Antibiotics on Growth Performance, Gut Microbiota, and Small Intestine Morphology in Yellow-Feather Broilers. *Microorganisms*, 11(9), 2308. <https://doi.org/10.3390/microorganisms11092308>
- [13]. Hartoyo, B., S. S., Iriyanti, N., & Susanti, (2015). Performance and Haematological Profile of Broiler Chicken Blood with Herbal Supplementation (fermenherfit). In T. Setyawardani, A. Susanto, & A. Sodiq (Eds.), *National Seminar on Animal Husbandry Technology and Agribusiness III* (pp. 242–251). Purwokerto: General Soedirman University.
- [14]. Hong, F., Zheng, A., Xu, P., Wang, J., Xue, T., Dai, S., Pan, S., Guo, Y., Xie, X., Li, L., Qiao, X., Liu, G., & Zhai, Y. (2020). High-Protein Diet Induces Hyperuricemia in a New Animal Model for Studying Human Gout. *International Journal of Molecular Sciences*, 21(6), 2147. <https://doi.org/10.3390/ijms21062147>
- [15]. Iriyanti, N., & Suhermiyati, S. (2015). Utilization of Afkir Milk as a Probiotic and Its Application in Feed to the Haematological Profile and Blood Fat of Broiler Chickens. In *Proceedings of the National Seminar on Animal Husbandry Technology and Agribusiness (Series III): Development of Animal Husbandry Based on Local Resources to Face the Asean Economic Community (MEA)* (pp. 230–236).
- [16]. Krysiak, K., Konkol, D., & Korczyński, M. (2021). Overview of the Use of Probiotics in Poultry Production. *Animals*, 11(6), 1620. <https://doi.org/10.3390/ani11061620>
- [17]. Kulkarni, R. R., Gaghan, C., Gorrell, K., & Sharif, S. (2022). Probiotics as Alternatives to Antibiotics for the Prevention and Control of Necrotic Enteritis in Chickens. *Pathogens*, 11(6), 692. <https://doi.org/10.3390/pathogens11060692>
- [18]. Kusumasari, Y. F. Y., Yunianto, V. D., & Suprijatna, E. (2012). Provision of phytobiotics derived from the crown of the gods (*phaleria macrocarpa*) against hemoglobin and hematocrit levels in broiler chickens. *Journal of Food Technology Applications*, 129 (4).
- [19]. Lestari, S. H. A., Ismoyowati, & M, I. (2013). Study of leucocyte count and leucocyte differential in various types of female local ducks whose feed was supplemented with probiotics. *JIP*, 1 (2), 699–709
- [20]. Magala H, Kugonza D, Kwizera H, Kyarisiima C. Influence of management system on growth and carcass characteristics of ugandan local chickens. *Journal of Animal Science Advances* 2012;2(6):557-567
- [21]. Mangkoewidjojo, S., & Smith, J. B. (1988). *Maintenance, Breeding, and Use of Experimental Animals in Tropical Areas*. Jakarta: University of Indonesia
- [22]. Mukti, A., Harris, A., & Masyitha, D. (2017). Resistance of *escherichia coli* to antibiotics from broiler chicken in the rukoh market. *JIMVET*, 01 (3), 492–498.
- [23]. Piliang, W., & Djojoseobagio, S. (2006). *Nutritional Physiology*. Bogor: Bogor Agricultural University Press
- [24]. Reece, W. O. (2006). *Functional Anatomy and Physiology of Domestic Animals* (3rd ed.). USA: Blackwell Publishing
- [25]. Sadrimovahed, M., & Ulusoy, B. H. (2024). *Bacillus clausii*: A Review into Story of Its Probiotic Success and Potential Food Applications. *Fermentation*, 10(10), 522. <https://doi.org/10.3390/fermentation10100522>
- [26]. Sastradipraja, S. H., Sikar, R., Wijayakusuma, T., Ungerer, A., Maad, H., Nasution, R., Hamzah. (1989). *Guide to Veterinary Physiology Practicum*. Bogor: IPB Inter-University Center for Life Sciences
- [27]. Sjojfan O., Adli D.N., Hanani P.K., & Sulistyaningrum D. (2020). The utilization of bay leaf (*SyzygiumpolyanthumWalp*) flour in feed on carcass quality, microflora instestine of broiler. *International Journal of Engineering Technologies and Management Research*, 6 (11), 1–9. <https://doi.org/10.29121/ijetmr.v6.i11.2019.458>
- [28]. Sjojfan, O., Nur Adli, D., & Adhana Muflikhien, F. (2020). Concept replacing feeding of rice bran on hybrid duck with hump flour on carcass percentage, internal organ and abdominal fat. *Journal of Tropical Animal Nutrition and Feed Science*, 2 (2). <https://doi.org/10.24198/jnttip.v2i2.28561>
- [29]. Soeharsono, L., Adriani, Hernawan, E., K.A, K., & Mushawwir, A. (2010). *Animal Physiology Basic Phenomena and Nomena, Function and Interaction of Organs in Animals*. Bandung: Widya Padjajaran.
- [30]. Steel, R. G. D., & Torrie, J. (1995). *Principles and Procedures of Statistics. In A Biometric Approach*. Jakarta: PT Gramedia Pustaka Utama
- [31]. Sturkie, P. D., & Griminger, P. (1976). Blood: physical characteristics, formed elements, hemoglobin and coagulation. In *Avian Physiology* (3rd ed.). New York: Springer-Verlag.
- [32]. Sukarmiati. (2007). *Study of the Use of Various Types of Probiotics on Blood Profiles, ND Titer and Ammonia Content of Laying Chickens*. Jenderal Soedirman University
- [33]. Wang, Z., Wu, J., Tian, Z., Si, Y., Chen, H., & Gan, J. (2021). The Mechanisms of the Potential Probiotic *Lactiplantibacillus plantarum* against Cardiovascular Disease and the Recent Developments in its Fermented Foods. *Foods*, 11(17), 2549. <https://doi.org/10.3390/foods11172549>
- [34]. Widiyawati, I., Sjojfan, O., & Adli, D. N. (2020). Increasing the quality and percentage of broiler carcasses by substituting soybean meal using fermented tamarind seed flour (*Tamarindus indica* L). *Journal of Tropical Animal Nutrition*, 3 (1), 35–40. <https://doi.org/10.21776/ub.jnt.2020.003.01.7>
- [35]. Winarsih, W. (2005). *Effect of probiotics in the control of subclinical salmonellosis in chickens: pathological and performance features*. Bogor Agricultural Institute.
- [36]. Zhang, H., Wu, C. X., Chamba, Y., & Ling, Y. (2007). Blood characteristics for high altitude adaptation in tibetan chickens. *Poultry Science*, 86 (7), 1384–1389. <https://doi.org/10.1093/ps/86.7.1384>