Efficacy of Probiotics on Nutrient Intake and Egg Weight in Japanese Quail (*Coturnix coturnix japonica*)

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Abstract

This study aimed to determine the efficacy of lactic acid bacteria probiotics containing *Lactobacillus acidophilus* and *Lactococcus lactis* on nutrient consumption and egg weight of Japanese quail. A total of 120 females Japanese quails comprised four treatments and six replications. T0, T1, T2, and T3 treatment groups were administered *L. acidophilus* and *L. lactis* with concentrations of 0, 1, 2, and 3 mL/liter of drinking water. Consumption data were collected weekly, and egg weight data were collected daily during treatment. The data obtained were then analyzed statistically using analysis of variance (ANOVA) and continued with the Duncan multiple range test (p < 0.05). The results of this study showed that probiotics in drinking water reported significant differences (p < 0.05) compared between treatments (T1, T2, and T3) to reduce nutrient intake of organic matter, crude protein, crude fat, crude fiber and increase egg weight of quail. It can be concluded that the combination of *L. acidophilus* and *L. lactis* can reduce nutrient consumption but can increase the egg weight of Japanese quail.

Keywords: Coturnix coturnix japonica, Lactobacillus acidophilus, Lactococcus lactis, nutrient intake

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INTRODUCTION

The increasing demand for animal protein in Indonesia is in line with the increasing population in Indonesia. Poultry farming plays an important role in supplying animal protein needs, one of which is quail farming. Based on statistical data from the Directorate General of Animal Husbandry and Animal Health (2022), the quail population (Coturnix coturnix japonica) in Indonesia has increased every year. starting in 2019 with as many as 14,844,104 heads in 2020 with as many as 15,222,580 heads, and in 2021 with as many as 16,014,879 heads. It is inversely proportional to quail egg production in Indonesia, which has decreased; egg production in 2019 was 25,862 tons, in 2020, it was 24,648 tons, and in 2021 it was 24,269 tons.

The potential to develop quail farming is high, in line with the increasing demand for quail eggs. This must be balanced with the provision of high-quality feed to meet the nutritional needs of quails for optimal production. However, a challenge faced in quail farming is the relatively expensive cost of feed (Fathinah, 2018). In connection with this, efforts are needed to reduce feed consumption, accompanied by an increase in quail productivity. Probiotics are one of the innovations that can be done to reduce feed costs, improve production performance, and provide an alternative to antibiotic growth promoters (AGP) (Irawan, 2020; Lovela *et al.*, 2023). As is well known since 2006, the European Union and several other countries have banned the use of AGP in livestock (Wu *et al.*, 2021).

Probiotics in feed can increase enzymatic activity and help the digestive system to increase efficiency in the feed and digestibility of the feed (Lokapirnasari *et al.*, 2018). Probiotics are often derived from lactic acid bacteria, which are the bacterial group that ferments carbohydrates into lactic acid (Jiang *et al.*, 2021). The lactic acid bacteria commonly used that have been studied in include poultry Lactobacillus bulgaricus, Lactobacillus plantarum, Pediococcus pentosaceus (Yulianto et al., 2021; Purnama et al., 2021), Lactobacillus casei, Lactobacillus lactis, and Bifidobacterium spp. (Lokapirnasari et al., 2022; Agustono et al., 2022). Lactobacillus acidophilus and Lactococcus lactis (Lokapirnasari et al., 2023), L. casei and L. rhamnosus (Andriani et al., 2020).

Probiotics containing *L. acidophilus* and *L. lactis* have not yet been studied concerning nutrient intake i.e. organic matter, crude protein, crude fat, and crude fiber or egg production in quails. Therefore, this study aimed to prove the efficacy of probiotics on nutrient intake and egg weight in quails.

MATERIALS AND METHODS

Ethical Approval

This study has been declared ethically feasible by the Research Ethics Commission of Universitas Brawijaya (033-KEP-UB-2022). The treatment in this study used probiotics as feed additives that are safe to use, are thought to have positive and beneficial properties, and do not cause pain in experimental animals. Variable retrieval did not slaughter experimental animals.

Study Period and Location

This study was conducted for 35 days (January–February 2021). The quails were reared in the breeding farm located at a quail farm in Kediri Regency. Proximate analysis of the feed and variables examination was conducted at the Laboratory of Animal Nutrition, Division of Animal Husbandry, Department of Veterinary Science, Faculty of Veterinary Medicine, Universitas Airlangga.

Experimental Animal

This study used 120-week-old quails. The study materials used in this study included a probiotic combination containing *L. acidophilus* and *L. lactis* with a concentration of 1.2×10^8 CFU/mL.

This study was conducted for 35 days, one week of adaptation, and 28 days of treatment. The quails were randomized into four treatments and six replications, respectively i.e. (T0) Control, (T1) *L. acidophilus* and *L. lactis* 1 mL/liter of drinking water, (T2) *L. acidophilus* and *L. lactis* 2 mL/liter of drinking water, (T3) *L. acidophilus* and *L. lactis* 3 mL/liter of drinking water.

The quails were adapted for one week with gradual doses of probiotics. The probiotic solution was mixed into their drinking water and stirred evenly. The preparation of the probiotic solution took place each morning before administration. Feeding was conducted twice a day at 7 a.m. and 4 p.m. The New Hope SQ101 commercial feed, provided in crumbles, was given in an amount tailored to meet the daily nutritional requirements of the quails while drinking water was made available ad libitum. This study ensured that variables such as temperature, humidity, lighting, and feed quality were controlled and kept consistent during the study period.

Data Collection

Data collection was carried out on feed consumption, and data obtained were compiled as an average for four weeks of treatment. Nutrient consumption data was calculated using the following formula (Karwanti et al., 2023): Dry matter intake (g) = feed intake (g) \times feed dry Organic matter intake (g) = feed intake (g) \times feed organic matter (%) \times feed dry matter (%) Crude protein intake (g) = feed intake (g) \times feed crude protein $(\%) \times$ feed dry matter (%) (3) Crude fat intake $(g) = feed intake (g) \times feed$ crude fat $(\%) \times$ feed dry matter (%)(4) Crude fibre intake (g) = feed intake $(g) \times feed$ crude fibre $(\%) \times$ feed dry matter (%)(5)

Statistical Analysis

The data obtained were analyzed using the analysis of variance (ANOVA) method, then followed by Duncan's multiple range test with p < 0.05. The data analysis process was entirely using SPSS 26.0 for the Windows program.

RESULTS AND DISCUSSION

The results of statistical analysis showed that there were significant differences (p < 0.05) in organic matter intake, and crude protein intake in each control treatment. Nutrient intake in this study in order from the lowest was T3, T2, T1, and T0. The organic matter intake was reported 15.51–16.49 g/quail/day, crude protein intake (4.45–4.73 g/quail/day), crude fat intake (1.16–1.23 g/quail/day), and crude fiber intake (0.77–0.82 g/quail/day), respectively (Table 1).

Variables	Organic matter intake (g/quail/day)	Crude protein intake (g/quail/day)	Crude fat intake (g/quail/day)	Crude fiber intake (g/quail/day)
T0	$16.49^d\pm0.04$	$4.73^{d} \pm 0.01$	$1.23^{d} \pm 0.01$	$0.82^d\pm0.00$
T1	$16.25^{\circ} \pm 0.18$	$4.66^{c} \pm 0.05$	$1.21^{\circ} \pm 0.01$	$0.81^{\circ} \pm 0.01$
T2	$15.85^b\pm0.08$	$4.55^{\text{b}}\pm0.02$	$1.18^{\text{b}} \pm 0.01$	$0.79^{b}\pm0.00$
Т3	$15.51^{a}\pm0.12$	$4.45^{\text{a}}\pm0.03$	$1.16^{\mathrm{a}} \pm 0.01$	$0.77^{\mathrm{a}} \pm 0.01$

^{a,b,c,d} Different superscripts in the same column showed significant differences (p < 0.05).

Table 2. Egg weight (g) evaluation after	probiotics administration in Japanese q	uail
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Variables	Egg weight (g)
T0	$10.31^{a} \pm 0.03$
T1	$10.41^{b} \pm 0.04$
T2	$10.48^{\circ}\pm0.01$
Т3	$10.50^{\circ} \pm 0.03$

 a,b,c Different superscripts in the same column showed significant differences (p < 0.05).

The results of statistical analysis showed that there was a significant difference (p < 0.05) in egg weight between control treatments. Duncan's multiple range test showed that egg weight in T2 and T3 treatment showed no significant difference (p > 0.05) (Table 2). The order of egg weight in this study was T3 with an egg weight of 10.50 g, T2 weighing 10.48 g, T1 weighing 10.41 g, and T0 weighing 10.31 g.

In the treatment of organic matter intake, crude protein intake, crude fat intake, and crude fiber intake showed lower results when compared to the control group. It was due to decreased feed consumption after treatment, so nutrient consumption also decreased (Wardiana et al., 2021). The provision of probiotics caused a decrease in nutrient consumption. Probiotics, as beneficial microorganisms, antipossess inflammatory effects that can reduce inflammation in the intestines. thereby influencing the health, structure, and function of the intestinal lining, including villi (Forte et al., 2016; Abdurrahman et al., 2022). The function of the villi is to absorb nutrients. Goblet cells in

intestinal villi have a role in secreting mucus that is useful as an antimicrobial, accelerating transportation through the intestinal epithelium, maintaining the integrity of the intestinal epithelium, and preventing the entry of pathogens into the intestine (Forder *et al.*, 2007; Agustono *et al.*, 2019; Reynolds *et al.*, 2020).

Khattab et al. (2021) also explained that the administration of probiotics in the diet can enhance feed utilization, thereby improving digestive enzyme activity and feed nutrient consumption. The accumulation of nutrients in tissues depends on feed consumption, intestinal absorption, and metabolism (Pavlidis et al., 2007; Wideman et al., 2013). Probiotic administration can lower pH by producing organic acids, making the digestive tract acidic (Faiqoh et al., 2023). This inhibits the growth of pathogenic bacteria and enhances non-pathogenic bacteria, maintaining the balance of normal flora in the poultry digestive tract (Ouwehand, 2016).

L. lactis bacteria produce antibacterial compounds, namely nisin, which have a bacteriocin effect against pathogenic bacteria

(Khelissa *et al.*, 2021). *L. acidophilus* bacteria produce two components of bacteriocin, namely bacteriocin B and acidolin (Hermawati *et al.*, 2020), which work to inhibit the development of pathogenic bacteria by lowering the pH in the digestive tract (Gao *et al.*, 2022). Probiotics, especially *L. acidophilus*, play a role in the balance of microbiota in the digestive tract and intestinal health, thus affecting feed consumption, digestibility, nutrient absorption, and increasing feed conversion ratio and feed efficiency (Wu *et al.*, 2021; Utomo *et al.*, 2022; Chandra *et al.*, 2022).

In this study, there was a decrease in nutrient intake in both organic matter intake, crude protein intake, crude fat intake, and crude fiber intake, but the production of egg weight showed a significant increase compared to the control (T0). The weight of eggs produced from the study was in the normal range of 10.31-10.50 g. Egg weight increases with the increasing age of quails and reaches maximum weight when approaching the end of the egg-laying period. It follows Daikwo et al. (2014), which state that the egg weight of quails was between 8-10 g with an average of 10 g/egg (about 8% of the body weight of the female quail). Some factors that cause variations in egg weight include feed, management, strain, age, nutrient intake, and diseases (Hanusoya et al., 2015; Sokołowicz et al., 2018; Fikri et al., 2024). The main factor that most affects eggs' weight is the amount of feed consumed with high protein (16%). The higher the feed protein given, the higher the weight of eggs produced by laying birds (Agro et al., 2013; Agustono et al., 2024).

In this study the addition of lactic acid bacteria probiotics could reduce nutrient intake but not with their egg weight. It is in line with the study of Lokapirnasari *et al.* (2019), which showed that probiotic supplementation of lactic acid bacteria could be used to improve the quality of the external and internal eggs of Japanese quail. It is in line with the study by Karwanti *et al.* (2023), which suggests that adding the probiotic *Lactobacillus* spp. can improve gut health and contribute to increased production. Probiotics can reduce pathogenic bacteria in the gastrointestinal tract, which can cause an increase in the optimal absorption of food substances (Amara and Shibl, 2013; Pratama *et al.*, 2021). Probiotics also optimize the absorption of feed nutrients, enabling the fulfillment of nutritional and energy requirements with only a small amount of consumed feed (Lokapirnasari *et al.*, 2019).

CONCLUSION

It can be concluded that feeding probiotics containing *L. acidophilus* and *L. lactis* in drinking water reduces nutrient intake but can increase egg weight. The administration of probiotics in drinking water at a dose of 2–3 mL/liter of drinking water can be applied by breeders to improve quail performance so that the quality obtained is maximized.

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AUTHORS' CONTRIBUTIONS

WPL: Conceptualization, Methodology. MAA, ALPK, ARL, SH, SHW, AA: Formal Analysis. WPL, ALPK, ARL: Project Administration. WPL, ALPK, ARL: Writing Original-Review & Editing. All authors have reviewed, and approved the read, final manuscript.

COMPETING INTERESTS

The authors declare that they have no competing interests.

REFERENCES

Abdurrahman, F., Soeharsono, S., & Soepranianondo, K. (2022). Study of Performance Index and Business Analysis on Chicken Infected by *Escherichia coli* with Probiotic Provision of Lactic Acid Bacteria. *Jurnal Medik Veteriner*, 5(1), 74–80.

- Agro, L. B., Tristiarti, & Mangisah, I. (2013). Kualitas Fisik Telur Ayam Arab Petelur Fase I dengan Berbagai Level Azolla Microphylla. *Animal Agricultural Journal*, 2(1), 445–457.
- Agustono, B., Al Arif, M. A., Yunita, M. N., Purnama, M. T. E., & Ulkhaq, F. (2019).
 Bioactivity of digestive enzymes and histological descriptions of jejunum of broilers supplemented with sunflower seed flour (*Helianthus Annuus* L). *Indian Veterinary Journal*, 96(8), 12–15.
- Agustono, B., Lokapirnasari, W. P., Yunita, M. N., Kinanti, R. N., Cesa, A. E., & Windria, S. (2022). Efficacy of Dietary Supplementary Probiotics as Substitutes for Antibiotic Growth Promoters During the Starter Period on Growth Performances, Carcass Traits, and Immune Organs of Male Layer Chicken. *Veterinary World*, 15, 324–330.
- Agustono, B., Abidah Alfirdausy, S., Warsito, S.
 H., Yunita, M. N., Lokapirnasari, W. P., Purnama, M. T. E., Chhetri, S., Windria, S., & Rahman, M. A. (2024). Influence of *Mauritia flexuosa* L. on Broiler Carcass Mass and Digestive Organs Temperature-Exposed Body Mass. *The Indian Veterinary Journal*, 101(2), 10–14.
- Amara, A. A., & Shibl, A. (2013). Role of Probiotics in Health Improvement, Infection Control and Disease Treatment and Management. *Saudi Pharmaceutical Journal*, 23, 107–114.
- Andriani, A. D., Lokapirnasari, W. P., Karimah,
 B., Hidanah, S., & Al-Arif, M. A. (2020).
 Potency of Probiotic on Broiler Growth
 Performance and Economics Analysis. *Indian Journal of Animal Sciences*, 90(8), 1140–1145.
- Chandra, E. H., Lokapirnasari, W. P., Hidanah, S., Al-Arif, M. A., Yuniarti, W. M., & Luqman, E. M. (2022). Probiotic Potential of Lactic Acid Bacteria on Feed Efficiency, Weight and Carcass Percentage in Ducks. Jurnal Medik Veteriner, 5(1), 69–73.
- Daikwo, I. S., Dim, N., & Momoh, M. O. (2014). Genetic Parameters of Some Egg Production

Traits in Japanese Quail in a Tropical Environment. *Journal of Agriculture and Veterinary Science*, 7(9), 39–42.

- Direktorat Jenderal Peternakan dan Kesehatan Hewan. (2022). Data Statistik Peternakan dan Kesehatan Hewan. Kementerian Pertanian Republik Indonesia, Jakarta.
- Faiqoh, B. E., Lamid, M., Damayanti, R., Chusniati, S., Al Arif, M. A., Warsito, S. H., Lestari, T. D., Raharjo, H. M., Rehman, S., & Hussain, M. A. (2023). Effect of Probiotic Administration of *Bacillus subtilis* and *Bacillus coagulans* Isolate on Growth Performance in Broiler Chicken. *Jurnal Medik Veteriner*, 6(3), 410-417.
- Fathinah, A. (2018). Pengaruh Penambahan Probiotik sebagai Pengganti Antibiotic Growth Promoter terhadap Analisis Usaha Burung Puyuh. [Thesis]. Fakultas Kedokteran Hewan. Universitas Airlangga: Surabaya.
- Fikri, F., Purnomo, A., Chhetri, S., Purnama, M. T. E., & Çalışkan, H. (2024). Effects of black soldier fly (*Hermetia illucens*) larvae meal on production performance, egg quality, and physiological properties in laying hens: A meta-analysis. *Veterinary World*, 17(8), 1904–1913.
- Forder, R. E. A., Howarth, G. S., Tivey, D. R., & Hughes, R. J. (2007). Bacterial Modulation of Small Intestinal Goblet Cells and Mucin Composition During Early Posthatch Development of Poultry. *Journal of Poultry Science*, 86, 2396–2403.
- Forte, C., Acuti, G., Manuali, E., Proietti, P. C., Pavone, S., Trabalza-Marinucci, M., Moscati, L., Onofri, A., Lorenzetti, C., & Franciosini, M. P. (2016). Effect of Two Probiotics on Microflora, Morphology, and Morphomrtry of Gut in Organic Laying Hens. *Journal of Poultry Science*, 95, 2528– 2535.
- Gao, H., Li, Xin., Chen, X., Hai, D., Wei, C., Zhang, L., & Li, P. (2022). The Functional Roles of *Lactobacillus acidophilus* in Different Physiological and Pathological Processes. *Journal of Microbiology and Biotechnology*, 32(10), 1226–1233.

- Hanusova, E., Hrnčár, C., Hanus, A., & Oravcova, M. (2015). Effect of Breed on Some Parameters of Egg Quality in Laying Hens. *Journal of Acta Fytotechnica et Zootechnica*, 18, 12–24.
- Hermawati, A. N., Aryati, & Isnaeni. (2020). Inhibitory Activity of Probiotic Milk Against *Eschericia coli ATCC 6538* and *Staphylococcus aureus ATCC 8739. Berkala Ilmiah Kimia Farmasi*, 7(1), 14–16.
- Irawan, P. I. (2020). Potensi Probiotik Bakteri Asam Laktat Terhadap Performa Produksi dan Analisis Usaha Pada Ayam Petelur. [Thesis]. Fakultas Kedokteran Hewan. Universitas Airlangga: Surabaya.
- Jiang, S., Lingzhi, C., & Longxian, L. (2021). Pediococcus pentosaceus, a Future Additive or Probiotic Candidate. Journal of Microbiology Cell Factories, 20(45), 1–14.
- Karwanti, N. W., Arumdani, D. F., Yulianto, A.
 B., Marbun, T. D., Sherasiya, A., Al-Arif, M.
 A., Lamid, M., & Lokapirnasari, W. P.
 (2023). Efficacy of *Moringa oleifera* Lam.
 Extracts and *Pediococcus pentosaceus*, *Lactobacillus acidophilus*, *Lactobacillus plantarum* probiotic during starter period on growth performance of male broiler chicken. *F1000Research*, 12, 215.
- Khattab, A. A., El Basuini, M. F., El-Ratel, I. T., & Fouda, S. F. (2021). Dietary probiotics as a strategy for improving growth performance, intestinal efficacy, immunity, and antioxidant capacity of white Pekin ducks fed with different levels of CP. *Poultry Science*, 100(3), 100898.
- Khelissa, S., Chihib, N, E., & Gharsallaoui, A. (2021). Conditions of Nisin Production by *Lactococcus lactis subsp. lactis* and Its Main Uses as a Food Preservative. *Archives of Microbiology*, 203, 465–480.
- Lokapirnasari, W. P., Al Arif, M. A., Maslachah, L., Chandra, E. H., Utomo, G. S. M., & Yulianto, A. B. (2022). Effect of Combination of Probiotics and *Moringa oleifera* Leaf Extract on Nutrients Intake in Ducks. *Jurnal Medik Veteriner*, 5(2), 241– 246.

- Lokapirnasari, W. P., Al Arif, M. A., Maslachah,
 L., Kirana, A. L. P., Suryandari, A.,
 Yulianto, A. B., & Sherasiya, A. (2023). The
 Potency of *Lactobacillus acidophillus* and *L. lactis* Probiotics and *Guazuma ulmifolia*Lam. Extract as Feed Additives with
 Different Application Times to Improve
 Nutrient Intake and Feed Efficiency in *Coturnix coturnix japonica* Females. *Journal of Animal and Feed Sciences*, 32(1),
 59–67.
- Lokapirnasari, W. P., Al Arif, M. A., Soeharsono., Fathinah, A., Najwan, R., Wardhani, H. C. P., Noorrahman, N. F., Huda, K., Ulfah, N., & Yulianto, A. B. (2019). Improves in External and Internal Egg Quality of Japanese Quail (*Coturnix coturnix japonica*) by Giving Lactic Acid Bacteria as Alternative Antibiotic Growth Promoter. *Iranian Journal of Microbiology*, 11(5), 406–411.
- Lokapirnasari, W. P., Hidanah, S., Suharsono., Fathinah, A., Dewi, A.R., Adriani, A.D., Karimah, B., Nuhajati, T., Soepraniandono, K., & Lamid, M. (2018). Probiotics on HDL, LDL, Cholesterol and Total Protein of Egg's Quail (*Coturnix coturnix japonica*). Journal of Applied Environmenta and Biological Science, 8(1).
- Lokapirnasari, W. P., Lamid, M., Kurnijasanti, R., Teriyanto, N., Kartika, A. T., Chandra, E. H., & Yulianto, A. B. (2020). Supplementation of Synbiotic Content of *Moringa oleifera* extract and Lactobacillus to Improve Growth Performance in Starter Phase Diet of Broiler Chicken. *Tropical Journal of Natural Product Research*, 4(12), 1096–1100.
- Lokapirnasari, W. P., Pribadi, T. B., Al Arif, M. A., Soeharsono., Hidanah, S., Harijani, N., Najwan, R., Huda, K., Wardhani, H. C., Rahman, N. F., & Yulianto, A. B. (2019).
 Potency of Probiotics *Bifidobacterium sp.* and *Lactobacillus casei* to Improve Growth Performance and Business Analysis in Organic Laying Hens. *Veterinary World*, 12(6), 860.

- Lordelo, M., Fernandes, E., Bessa, R. J. B., & Alves, S. P. (2016). Quality of Eggs from Different Laying Hen Production Systems, from Indigenous Breeds and Specialty Eggs. *Journal of Poultry Science*, 96, 1485–1491.
- Lovela, A. R., Lokapirnasari, W. P., Warsito, S. H., Prasinta, R., Hapsari, T., & Andriani, A. (2023). The Quality of Japanese Quail Eggs After Administration of *Bifidobacterium* sp. and *Guazuma ulmifolia* Leaf Extract. *Jurnal Medik Veteriner*, 6(1), 132–136.
- Ouwehand, A. C., Forssten, S., Hibberd, A. A., Lyra A., & Stahl, B. (2016). Probiotic Approach to Prevent Antibiotic Resistance. *Annals of Medicine Journal*, 48, 246–255.
- Pavlidis, H. O., Balog, J. M., Stamps, L. K., Hughes, J. D., Huff, W. E., Anthony, N. B. (2007). Divergent Selection for Ascites Incidence in Chickens. *Journal of Poultry Science*, 86, 2517–2529.
- Pratama, H. S., Lokapirnasari, W. P., Soeharsono, S., Al-Arif, M. A., Harijani, N., & Hidanah, S. (2021). Effect of probiotics *Bacillus subtilis* on feed efficiency and egg mass of laying hens. *Jurnal Medik Veteriner*, 4(1), 37–41.
- Purnama, M. T. E., Ernanda, E. P., Fikri, F., Purnomo, A., Khairani, S., & Chhetri, S. (2021). Effects of dietary supplementation with breadfruit leaf powder on growth performance, meat quality, and antioxidative activity in Japanese quail, *Veterinary World*, 14 (7), 1946–1953.
- Reynolds, K. L., Cloft, S. E., & Wong, E. A. (2020). Changes with Age in Density of Goblet Cells in the Small Intestine of Broiler Chicks. *Journal of Poultry Science*, 99, 2342–2348.

- Sokołowicz, Z., Krawczyk, J., & Dykiel, M. (2018). The Effect of the Type of Alternative Housing System, Genotype and Age of Laying Hens on Egg Quality. *Journal of Annals Animal Science*, 18(2), 541–556.
- Utomo, G. S. M., Hidanah, S., Al Arif, M. A., Lokapirnasari, W. P., & Yuniarti, W. M. (2022). Business Analysis of Probiotic Administration of Lactic Acid Bacteria on The Performance of Kampung Super Chicken. Jurnal Medik Veteriner, 5(1), 87– 93.
- Wardiana, N. I., Lokapirnasari, W. P., Harijani, N., Al-Arif, M. A., & Ardianto, A. (2021). *Bacillus subtilis* probiotics in chicken feed improve egg quality with differences in shelf life. *Jurnal Medik Veteriner*, 4(1), 8–13.
- Wideman, R. F., Rhoads, D. D., Erf, G. F., & Anthony, N. B. (2013). Pulmonary Arterial Hypertension (Ascites Syndrome) in Broilers: A Review. *Journal of Poultry Science*, 92, 64–83.
- Wu, Z., Yang, K., Zhang, A., Chang, W., Zheng, A., Chen, Z., Cai, H., & Liu, G. (2021).
 Effects of *Lactobacillus acidophilus* on the Growth Performance, Immune Response, and Intestinal Barrier Function of Broiler Chickens Challenged with *Escherichia coli* 0157. *Journal of Poultry Science*, 100, 101323.
- Yulianto, A. B., Suwanti, L. T., Widiyatno, T. V., Suwarno, S., Yunus, M., Tyasningsih, W., Hidanah, S., Sjofjan, O., & Lokapirnasari, W. P. (2021). Probiotic *Pediococcus pentosaceus ABY 118* to Modulation of ChIFN-γ and ChIL-10 in Broilers Infected by *Eimeria tenella* oocyst. *Veterinary Medicine International Journal*, 1, 1–6.
