



The Addition Probiotic as Feed Additive on Intestinal Characteristic of Cross Breed Chickens

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Abstract. The purpose of this study was to evaluate the effect of adding various levels of liquid probiotics as feed additive on the intestinal characteristics of cross-breed chickens. The material was used 180 non-sexing cross-breed chickens maintained for 60 days. This research method was a field experiment using a completely randomized design consisting of 5 treatments and 4 replications. The treatments were T0 = control feed, T1 = control feed + 0.25% liquid probiotic, T2 = control feed + 0.5% liquid probiotic, T3 = control feed + 0.75% liquid probiotic, T4 = control feed + 1% liquid probiotic. Data were analyzed by analysis of variance and if there is a significant effect will be tested with DMRT. The measured variable was digesta viscosity, number of ileal villi, length, and depth of the crypt. The results showed that probiotics had a very significant effect ($P < 0.01$) on the viscosity and number of villi, but had no significant effect ($P > 0.05$) on the length of the ileal villi, and the depth of the crypt. It concluded that the addition of 0.75% liquid probiotics as a feed additive can give the best result to improve the intestinal characteristics of cross-breed chicken.

Keywords: characteristic · cross breed · chicken · intestinal

1 Introduction

Chicken meat is one of the most popular livestock products that can be used for animal protein needs. This can be seen in meat production in Indonesia, which dominated by chicken at 79%, followed by beef at 11%, pork at 5%, and other meat at 5%. Broiler and layer farms are the most preferred choice for chicken farmers in Indonesia, but some farmers choose native chickens with the consideration that they can produce meat which tasty, savory, not mushy, and low in fat [1]. Native chickens tend not to develop due to slow and low meat production. This problem triggers the innovation of crossbreed chickens between male chickens (Bangkok) and female chickens which aims to produce meat with a pure native chickens taste, but the maintenance period is relatively faster. These crossbreed chickens are often known as *Joper* chickens. The key to the success of a farm is the focus of farmers in managing the cage, feed, and breed, which of the three factors, feed occupies the highest component in the success of livestock production.

Cultivation of crossbreed chickens must be supported by proper feeding in order to produce quality meat by adjusting the needs of chickens. The nutritional needs of chickens will have a positive impact on the metabolic process in the chicken's body so that it can produce optimally [2]. One of the efforts to increase the utilization of nutrients currently widely applied is the addition of feed additives.

Feed additives are not included in the feed ingredients that are added to promote optimal livestock growth, increasing productivity, and production quality. So far, the livestock industry used a lot of feed additives like AGP (Antibiotic Growth Promoters) which are considered to be able to increase the efficiency of feed use with the principle of reducing pathogenic bacteria in the digestive tract, so that it will increase the performance and the productivity optimally [3]. Giving antibiotics to livestock can also be dangerous for humans who consume by causing allergic reactions, even poisoning is possible [4]. The Indonesian government has officially banned the use of AGP as feed additive since January 1, 2018, as stipulated in the Minister of Agriculture Number 14/2017 Article 16, both in the form of products and raw materials for veterinary drugs mixed in feed. With the prohibition on the use of AGP, research was conducted to find alternatives to increase livestock productivity without causing negative impacts for both livestock and consumers. Several alternative feed additives that have been developed in the livestock industry, especially in the poultry sector are probiotics, prebiotics, enzymes, organic acids, and phytobiotics [5]. Probiotics are often used as feed additives because of their easy application.

Probiotics are one of the feed additives with microorganisms that can increase the growth and efficiency in animal feed without causing absorption of probiotic components in the body, so there will be no residues and no mutations occur in livestock [6]. The addition of probiotics is considered to improve the digestive tract. For this reason, seeing the success of probiotic performance and health in the digestive tract, it can be done by observing the characteristics of the intestine. Intestinal characteristics are related to the production performance of livestock. Based on the background above, this study focused on the effect of adding liquid probiotics as feed additive on intestinal characteristics of crossbreed chickens to increase livestock productivity. This research could be improved the green economy through of improving nutrition for the human by utilizing probiotics as an poultry feed additive.

2 Materials and Method

2.1 Research Location

The research was conducted from 22 September to 28 November 2021 at UD. Berlin Farm, Maguan Village, Ngajum District, Malang Regency. Histopathic preparations were made at Institut Bioscience Universitas Brawijaya. Viscosity measurement is carried out at the Feed Laboratory of Universitas Brawijaya. Measurements of the number of villi, length of villi, and depth of crypts were carried out at the Biomole Laboratory of the Faculty of Mathematics and Natural Sciences, Universitas Brawijaya.

2.2 Research Materials

This research used 180 non-sexed crossbreeds DOC (Day Old Chick), colony and open house cages, starter and finisher feed, feed additive in form of liquid probiotics with the composition of Lactic Acid Bacteria and *Bacillus* sp. 2.58×10^9 CFU/ml, chicken vaccines and medicines, as well as complementary equipment for the cage.

2.3 Research Methods

The method used is the in vivo experimental method of feed using a completely randomized design (CRD) in a unidirectional pattern with 1 treatment factor, that is the addition of commercial liquid probiotics (containing Lactic Acid Bacteria and *Bacillus* sp. 2.58×10^9 CFU/ml) in feed according to the treatment, with the difference in level as much as 0%, 0.25%, 0.5%, 0.75%, and 1%. The study consisted of 5 treatments with each treatment consisting of 4 replications so there were 20 experimental units. Each replication consisted of 9 chickens. The treatment includes:

T0 = Control Feed

T1 = Control Feed + 0.25% Probiotic

T2 = Control Feed + 0.5% Probiotic

T3 = Control Feed + 0.75% Probiotic

T4 = Control Feed + 1% Probiotic

2.4 Research Variable

The variables measured in this study are:

1. Viscosity

Measured in the area of the ileum of a freshly cut chicken. Digesta viscosity was measured by diluting 1 gram of digesta in the small intestine with distilled water to a volume of 10 ml. The solution was centrifuged at 3000 rpm for 5–10 min., then the supernatant liquid from the centrifugation was separated for viscosity measurement using a viscometer.

2. The number of villi, the length of the villi, and the depth of the crypts of the small intestine

The villi preparations were analyzed with DIC Olympus BX51TF light microscope that was connected to the optilab application. Measurements of the number of villi, length of villi, and depth of crypts were carried out using the Image Raster application whose magnification was adjusted at the time of observation

2.5 Data Analysis

The data obtained were analyzed using ANOVA on CRD with the help of Microsoft Excel. If the results of the analysis obtained data are significantly different or very real, then proceed with Duncan's Multiple RangeTest (DMRT).

3 Results and Discussion

The results of this observation and statistical analysis on the small intestine of crossbreed chickens including viscosity, number of villi, length of villi, and depth of crypts given 5 treatments can be seen in Table 1.

Table 1. Viscosity, number of villi, length of villi, and depth of crypts in the digestive tract of crossbreed chickens

Treatments	Variables			
	Viscosity (cP)	Number of Villi (per transversal cut)	Length of Villi (μm)	Depth of Crypt (μm)
T0	8.25 ± 0.96^a	38.09 ± 1.16^a	511.08 ± 65.56	121.95 ± 14.05
T1	13.50 ± 1.73^b	42.09 ± 3.36^a	566.99 ± 102.38	125.94 ± 12.63
T2	13.00 ± 2.16^b	42.48 ± 2.15^a	572.22 ± 44.47	127.58 ± 17.48
T3	12.25 ± 0.50^b	51.00 ± 2.33^b	545.48 ± 27.45	125.45 ± 8.36
T4	11.25 ± 0.96^b	47.75 ± 2.85^b	645.31 ± 73.08	129.31 ± 9.46

a: average redness; b: average yellowness

3.1 Effect of Addition Probiotics as Feed Additive on Viscosity

Table 1 shows the results of the average viscosity of crossbreed chickens based on the field studies with control feeding and the addition of liquid probiotics as feed additives that have observed in the laboratory. Based on this study, the results of data analysis showed that the addition of liquid probiotics to feed had a very significant effect ($P < 0.01$) with the average viscosity from the lowest to the highest was T0 (8.25 ± 0.96), T4 (11.25 ± 0.96), T3 (12.25 ± 0.50), T2 (13.00 ± 2.16), and T1 (13.50 ± 1.73).

The highest viscosity was 13.50 at T1 chicken with the addition of 0.25% probiotics, while the lowest was 8.25 at T0 or without the addition of probiotics. Probiotics are considered to be able to increase viscosity because of their ability to produce enzymes. This is following research conducted by Sjojfan, *et al.*, [7] who reported that an increase in digesta viscosity was evidenced the reshuffle of NSP (Non-Starch Polysaccharide) content in the feed so that it has an impact on viscosity digestion. The probiotic bacteria *Bacillus* sp. can produce digestive enzymes such as protease and amylase that can help digestion, and produce short-chain organic acids that have antimicrobial properties [8]. Increased digesta viscosity will cause the digesta rate to be slow and allow for an increase in the digestive process and absorption of nutrients more effectively, so the availability of nutrients for the synthesis of body tissues increases. Meanwhile, if the digesta viscosity is low, the digesta rate will be faster and allow a decrease in the digestive process and absorption of food substances. This decrease in absorption activity causes the number and length of the villi of the small intestine of chicken not to develop properly [7].

3.2 Effect of Addition Probiotics as Feed Additive on Number of Villi

Table 1 shows the results of the average number of villi of crossbreeds based on field studies that have been observed in the laboratory. Based on this study, the results of data analysis showed that the addition of liquid probiotics to feed had a very significant effect ($P < 0.01$) with the average number of villi from the lowest to the highest was T0 (38.09 ± 1.16), T1 (42.09 ± 3.36), T2 (42.48 ± 2.15), T4 (47.75 ± 2.85), T3 (51.00 ± 2.33). The highest value of the number of villi was 51.00 at T3 chicken with 0.75% probiotic addition, while the lowest value was 38.09 at T0 or without the addition of probiotics. The increase in villi was due to the presence of probiotic fermentation by bacteria which produces short-chain fatty acids, helping in expanding the absorption and multiplication of intestinal epithelial cells and protecting the villi from damage by reducing and preventing pathogenic infection by producing antimicrobials on the walls of the digestive tract so the villi can grow maximum properly [9]. This is following research by Hidayat *et al.*, [10] stated that the lactic acid bacteria produce butyric acid that has been shown to increase villi in quail. The villi in the small intestine play a role in the absorption of nutrients. The increase in the number of villi proves that there is an increase in the digestive process and absorption of food in the intestines of the ileum so that it can increase livestock productivity. Pertiwi *et al.*, [11] state that indicators of a healthy chicken digestive tract can be seen from the development of body weight and length of the digestive tract as well as the optimal development of intestinal villi.

3.3 Effect of Addition Probiotics as Feed Additive on Length of Villi

Based on the research, the results of the data analysis in Table 1 show that the addition of liquid probiotics to the feed had no significant effect ($P > 0.05$) with the average length of villi from the lowest to the highest was T0 (511.08 ± 65.56), T3 (545.48 ± 27.45), T1 (566.99 ± 102.38), T2 (572.22 ± 44.47), then T4 (645.31 ± 73.08). The highest value of the length of villi was 645.31 at T4 chicken with 1% probiotic addition, while the lowest value was 511.08 at T0 or without the addition of probiotics. This condition is suspected because the absorption of food substances containing probiotics has not been absorbed optimally. In contrast to observations by Hidayat, *et al.*, [10] who reported that giving probiotics in the form of lactic acid bacteria to quail had a significant effect on the length of villi because the feed consumed was well absorbed, thus reflecting that the status of the livestock was in a healthy condition. The increase in the length of the villi reflects the maximum absorption of nutrients. Based on the results of the study, T4 had a higher villi length than the control or other treatments because it was suspected that the chickens in that treatment could absorb feed better than the others. This is following Kompang [12] who reported that broiler chickens given *Bacillus* sp. have longer villi with intestinal surface area to absorb nutrients more widely than those receiving AGP. Pathogenic microbes found in probiotics will block pathogens with their adhesion to the intestinal mucosa, by increasing immunity and increasing nutrient absorption. Priastoto, *et al.*, [13] added to their research that macroscopically probiotics can cause the intestinal size to become longer, and microscopically it can affect the density and length of the villi so that the absorption of nutrients in chickens is judged to be better by consuming less ration.

3.4 Effect of Addition Probiotics as Feed Additive on Depth of Crypt

Based on research, the results of data analysis in Table 1 show that the addition of liquid probiotics to feed has no significant effect ($P > 0.05$) with the average crypt depth from the lowest to the highest was T0 (121.95 ± 14.05), T3 (125.45 ± 8.36), T1 (125.94 ± 12.63), T2 (127.58 ± 17.48), then T4 (129.31 ± 9.46). The highest crypt depth value was 129.31 at T4 chicken with 1% probiotic addition, while the lowest value was 121.95 at T0 or without the addition of probiotics. This is thought to be correlated with the length of the villi which in the results of the analysis also does not have a significant effect. In addition, the stressed condition influenced the nutrients in the feed are not absorbed properly. In contrast to Sen, et al., [14] who reported that administration of *Bacillus subtilis* could increase the ratio of villi height to crypt depth in the duodenum and ileum of broiler chickens. The higher depth of the crypt made nutrients are digested and absorbed which ultimately affects the growth of body organs [15].

4 Conclusion

Based on the results of the study, it can be concluded that the addition of liquid probiotics as a feed additive with an optimal level of 0.75% gave the best results in improving viscosity, number of villi, length of villi, and depth of crypts. Improving quality of villi could be enhanced the absorption of the nutrients. Then it could be improved the green economy in the future.

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