Evaluation of the Use of Date Seed Flour in Feed on the Small Intestine Characteristics of Broiler

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ABSTRACT: The purpose of this study was to determine and evaluate the use of date seed flour in feed on the small intestine characteristics of broilers. This study used 200 Day Old Chick (DOC) broilers with type MB 202. This study used a field experiment method with a Completely Randomized Design (CRD) consisting of 5 treatments and 4 replications. The treatments used were P0: basal feed without the use of date seed flour (DSF), P1: basal feed using 2.5% DSF, P2: basal feed using 5% DSF, P3: basal feed using 7.5% DSF, P4: basal feed using 10% DSF. The variables measured in this study were the number of villi, villi height, villi surface area, crypt depth, digesta pH, and digesta viscosity. The data obtained during the study were processed using Microsoft Excel software and analyzed using analysis of variance (ANOVA) and continued with Duncan's Multiple Range Test if the results obtained were significantly different (P<0.05) or very significantly different (P<0.01). This study gave results that were not significantly different (P>0.05) on the number of villi, villi height, villi surface area, crypt depth, and digesta pH. However, this study was able to give very significantly different results (P<0.01) in digesta viscosity. The conclusion of this study is the use of date seed flour in feed up to 10% was able to give good results on the number of villi, villi height, villi surface area, crypt depth, and digesta pH of the small intestine of broilers, but it has not been able to give good results on the digesta viscosity of the small intestine of broilers.

Keywords: Broiler; Date seed flour: small intestine characteristics

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INTRODUCTION

Today, businesses in the livestock sector are starting to develop. This is evidenced by an increase in domestic meat consumption. One of the livestock that has the main product in the form of meat and the most favored by the community is the broiler. According to data from the Directorate General of Livestock and Animal Health of the Indonesian Ministry of Agriculture (2021), the consumption of broiler meat per capita per year in 2019 was 5.683 kg and increased to 6,048 kg in 2020. The increase related to meat consumption must be balanced with the availability of meat so that to meet the availability of meat, an effort is needed to increase the population of broilers.

Important aspects in broiler development are breeding, feeding, and rearing management. Breeding is the main determinant of broiler productivity because the productivity of an animal is greatly influenced by genetics, so superior breeds are needed. The ability of broilers to produce meat very quickly and efficiently is influenced by the main factor in the form of superior breeds (Tangendjaja, 2014). In rearing broilers with superior breeds, it is also necessary to support appropriate feed nutrition to optimize livestock productivity.

Feed is a supporting factor for breeds and is an important aspect of the livestock business because it is related to production costs. Production costs derived from the feed are the largest component that reaching 60-70%. This is what causes livestock business actors to pay extra attention to the feed provided to produce low costs. Most of the farmers in Indonesia use commercial feed where the price is still very volatile, even in certain areas where commercial feed is so scarce that there are farmers who use food waste that can affect the health of livestock.

Therefore, an alternative is needed by exploring the potential of conventional and non-conventional feed ingredients that have good quality as animal feed ingredients at low prices and are available in sufficient quantities to meet livestock needs. Feed ingredients can be said to be of good quality if they contain nutrients that can have a positive effect on livestock growth and productivity. One of the feed ingredients that can be used as animal feed is date seeds.

Date seeds are a by-product of processing date fruits which still have good nutritional content as animal feed ingredients. Date seeds are rich in mineral content and have a high carbohydrate content as an energy source for livestock. The content of carbohydrates in date seeds is 72 - 73.4%, protein 5 - 6.5%, and fat 9.9 - 13.6%, and date seeds have components of 6.10 - 11.47% of the total dates (Sendikadisnu et al., 2020).

Date seeds contain flavonoids which act as antioxidants in the body of livestock which will affect the physiological condition of the digestive tract, especially the small intestine. The increase in villi in the intestine can occur due to the influence of flavonoid compounds which cause the absorption surface to become wider so that nutrient absorption becomes more optimal (Mistiani et al., 2020). Broilers given the addition of date seeds in the feed can increase antioxidants that function as immunity in broilers (El-Far et al., 2016).

This shows that date seeds can be used as animal feed ingredients because the nutritional content contained in them can have a positive effect on livestock. In the use of date seeds as animal feed ingredients, it is necessary to carry out a processing process into flour which includes separating the seeds from the fruit flesh, drying, and milling. The use of date seed flour (DSF) in broiler feed is expected to increase the nutritional content of the feed and make a good contribution to improving the physiological condition of the small intestine.

Based on this description, it is necessary to research evaluating the use of DSF in feeding on the characteristics of the small intestine of broilers to know and evaluate the use of DSF in feeding on the characteristics of the small intestine of broilers which include the number of villi, villi height, villous surface area, crypt depth, digesta pH, and digesta viscosity.

MATERIALS AND METHODS Materials

The material used in this study were 200 Day Old Chick (DOC) broilers type MB 202 that were undifferentiated by sex with an average body weight of 45.03 ± 3.31 with a coefficient of the diversity of 7.35%. The cage used was an open house cage consisting of 20 plots with the size of each plot of 130 cm x 100 cm x 60 cm. Each plot is filled with 10 broilers. The base of the cage is in the form of rice husks and each plot is equipped with a place for feeding and drinking.

The equipment used includes feed, drinking, digital scales with a capacity of 10 kg with an accuracy of 1 g, a 10 Watt bulb as lighting, stationery, thermo-hygrometer, brooder, Liquified Petroleum Gas (LPG), and cage cleaning equipment such as broom, bucket, and disinfectant sprayer. The materials used include feed ingredients including DSF, corn, concentrate, and bran, as well as chemicals including 0.01% physiological NaCl and 10% formalin. The feed used in this study was self-mixed. The nutrient content of the feed ingredients can be seen in Table 1.

The composition of the treatment feed and the nutrient content based on the calculations are presented in Table 2.

Methods

The method used in this study was a field experiment using a completely randomized design (CRD) consisting of 5 treatments and 4 replications, each replication consisting of 10 broilers. The treatments used include:

- P0 = Basal feed without the use of DSF
- P1 = Basal feed using 2.5% DSF
- P2 = Basal feed using 5% DSF
- P3 = Basal feed using 7.5% DSF
- P4 = Basal feed using 10% DSF

Research Procedure

The initial stage of this study began with the preparation of feed ingredients and preparation of feed, preparation of cages and equipment and research support materials, and continued with the preparation of broiler DOC livestock.

Maintenance of broilers is carried out for 35 days, starting from the starter phase at the age of 1-21 days with a brooding period at the age of 1-14 days, followed by the finisher phase at the age of 22-35 days. During the rearing period, broilers were fed and watered ad libitum 2 times a day, at 07.00 WIB and 16.00 WIB. After the broilers were 35 days old, harvesting was done by cutting 1 broiler in each replication. Next, surgery is performed to separate the digestive tract.

Sampling was carried out by cutting the small intestine along 3-5 cm in the ileum, followed by removing the digesta and accommodating in a film pot as much as 1 g for pH testing and 1 g for viscosity testing. Next, another 3-5 cm cut was made in the ileum at a distance of 5 cm from the caeca junction, the digesta was removed using a syringe filled with physiological NaCl solution slowly, then the sample is put into a film pot filled with 10% formalin solution to make preparations. Preparations were made by cutting the small intestine with a thickness of 4 µm using a microtome and placing it on a slide for staining using the Haemoxylin-eosin (HE) method (Ranjan, et al., 2016).

Observations were made on the number of villi, villous height, basal width, and apical width of the villi to calculate the surface area of the villi, as well as the depth of the crypt on the preparations using a DIC BX51TF Olympus light microscope connected to the Cytovision software (Natsir, et al., 2013). The surface area of the villi is calculated based on the formula of Iji et al., (2001) in Kusuma et al., (2020) namely (b + c) / c x a (a = villous height, b = villous basal width, c = villous apical width).

Nutrient content*	Corn	Concentrate	Bran ^a	Date seed flour ^s
Metabolizable energy (Kcal/kg)	3370	2400	2800	2983,865
Crude protein (%)	8,6	41	8	5,31
Crude fat (%)	3,9	4	8	4,43
Crude fiber (%)	2	6	12	8,97
Ash (%)	3,66	13	8	10,11
Calcium (%)	0,02	2,85	0,12	
Phosphor (%)	0,01	1,5	0,21	

 Table 1. Nutrient Content in Feed Ingredients

*Based on 100% dry matter. a: based on the standard content of feed ingredients. b: commercial feed: KBR PT. Japfa Comfeed Indonesia, Tbk. c: the results of the analysis at the Laboratory of Animal Nutrition and Feed, Faculty of Animal Science, Brawijaya University, Malang, Indonesia.

Table 2.	Composition	of Feed	Treatment	and (Content	of Nutrients	Based on	Calculations
				a	-			

Starter phase							
Feed ingredients	Composition (%)						
	P0	P1	P2	P3	P4		
Corn	60	57,25	54,49	51,74	48,98		
Concentrate	40	40,25	40,51	40,76	41,02		
Date seed flour	0	2,5	5	7,5	10		
Total	100	100	100	100	100		
	Nutrient con	tent*					
Metabolizable energy (Kcal/kg)	2982,00	2969,88	2957,77	2945,65	2933,54		
Crude protein (%)	21,56	21,56	21,56	21,56	21,56		
Crude fat (%)	3,94	3,95	3,97	3,98	3,99		
Crude fiber (%)	3,60	3,78	3,97	4,15	4,34		
Ash (%)	7,40	7,58	7,77	7,95	8,14		
Finisher phase							
Feed ingredients		Con	position (%	6)			
	P0	P1	P2	P3	P4		
Corn	56	53,15	50,40	47,65	44,89		
Concentrate	35	35,26	35,51	35,76	36,02		
Bran	9	9,09	9,09	9,09	9,09		
Date seed flour	0	2,5	5	7,5	10		
Total	100	100	100	100	100		
Nutrient content*							
Metabolizable energy (Kcal/kg)	2983,70	2971,10	2971,10	2971,10	2971,10		
Crude protein (%)	19,89	19,89	19,89	19,89	19,89		
Crude fat (%)	4,30	4,32	4,32	4,32	4,32		
Crude fiber (%)	4,30	4,49	4,49	4,49	4,49		
Ash (%)	7,32	7,51	7,51	7,51	7,51		

*Based on the calculations in Table 1.

Data Analysis

The data obtained during the study were tabulated and calculated using Microsoft Excel software and analyzed using analysis of variance (ANOVA), if the results obtained were significantly different (P<0.05) or very significantly different (P<0.01) then followed by Duncan's Multiple Distance Test (Steel and Torrie, 1995).

The effect of using DSF in feed on the characteristics of the small intestine of broilers which include the number of villi, villi height, villous surface area, crypt depth, digesta pH, and digesta viscosity in broilers during the study can be seen in Table 3.

Effect of DSF use on the number of villi

The absorption of food substances that occurs in the digestive tract can be influenced by the number of villi in the intestine because the intestinal villi function as a place for absorption of food substances. Based on the data presented in Table 3, the of average number villi produced sequentially from the highest to the lowest is P4 (66±2.75), P3 (63±9.15), P2 (63±1.89), P1 (62 ±6.83), P0 (54±7.55). The average number of villi produced in the study showed that the higher the use of DSF in the feed, the higher the number of villi. This is thought to be caused by the presence of flavonoid compounds in DSF which can inhibit the growth of pathogenic bacteria to maintain the physiological condition of the small intestine.

This is following the results of research by Alyileili et al., (2020) that broilers fed with degraded date seeds as

much as 5% and 10% were able to reduce the population of pathogenic bacteria such as Salmonella spp., Campylobacter spp., Shigella spp., and Escherichia coli. The results of statistical analysis showed that the treatment had no significant effect (P>0.05) on the number of small intestinal villi. This indicates that the use of DSF in feed has a positive effect on the number of villi in the small intestine of broilers. The increase in the number of villi produced in this study illustrates that the process of absorption of nutrients can run optimally. This is following the statement of Djunaidi et al., (2020) that the number of villi in the intestine will affect the absorption of nutrients so that absorption will run optimally when supported by a high number of villi. The highest number of villi in this study was at P4 with the use of DSF as much as 10% in the feed. This shows that to increase the number of villi in the small intestine, DSF up to 10% can be used in broiler feed.

Table 3. Effect of 7	Treatment on	Small	Intestine	Characteristics	of Broilers
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Treatments	Number of villi	Villi height (µm)	Villi surface area (µm ²)	Crypt depth (µm)	Digesta pH	Digesta viscosity (mPaS)
PO	54,00±7,55	340,96±21,26	961,37±87,30	132,32±10,18	6,50±0,28	110±0,00 ^A
P1	62,00±6,83	353,17±53,04	965,90±335,42	132,65±11,96	6,55±0,30	$110,25\pm0,50^{B}$
P2	63,00±1,89	381,74±33,44	993,22±107,97	145,39±13,71	6,61±0,23	$110\pm0,00^{A}$
P3	63,00±9,15	388,81±23,09	1077,59±210,54	148,51±9,51	6,71±0,25	$111\pm0,00^{\circ}$
P4	66,00±2,75	407,87±14,76	1043,64±153,29	149,62±11,38	6,57±0,21	111±0,00 ^C

Note: Different superscripts (A-C) in the same column indicate a very significant difference (P < 0.01).

Effect of DSF use on the villi height

Another factor that can affect the absorption of nutrients is the height of the small intestinal villi. According to Ibrahim (2008), the potential and capacity of the small intestine in digesting and absorbing food substances are influenced by the physical size of the intestine which varies greatly. Based on the data presented in Table 3, the average villous heights produced sequentially from the highest to the lowest are P4 (407.87±14.76 m), P3 (388.81±23.09 P2 $(381.74 \pm$ 33.44 m), m). P1

 $(353.17\pm53.04 \text{ m})$, P0 $(340.96\pm21.26 \text{ m})$. The average villi height produced in the study increased in line with the increase in the use of DSF in feed. The increase in villi height indicates that the potential and absorption capacity of the small intestine is greater.

This is comparable with the statement of Awad et al., (2008) in Jamilah et al., (2014) that an increase in villi height in the small intestine of broilers has a positive correlation with an increase in digestive function and absorption function because the wider the absorption area, the smoother the transportation system. nutrients throughout the body that benefit the host. The results of statistical analysis showed that the treatment had no significant effect (P>0.05) on the height of the small intestine villi.

This shows that the use of DSF in feed has a positive effect on the height of the villi of the small intestine of broilers. The level of villi in the intestine can be influenced by various factors, one of which is the growth of pathogenic bacteria. The decreased colony of pathogenic bacteria will affect the increase in the height of the small intestinal villi, and vice versa, if the colony of pathogenic bacteria increases it will inhibit the growth of the small intestine villi height (Jamilah, et al., 2014). The highest villi height in this study was at P4 with the use of DSF as much as 10% in the feed. This shows that to increase the height of the villi in the small intestine, up to 10% DSF can be used in broiler feed.

Effect of DSF use on the villi surface area

Based on the data presented in Table 3, the average villous surface area produced sequentially from the highest to the lowest is (1043.64±153.29 P4 m2), P3 (1077.59±210.54 m2), P2 (993.22 ±107.95 (965.90 ± 335.42) m2). P1 m2). **P0** (961.37±87.30 m2). The average villous surface area produced in the study was directly proportional to the use of DSF in feed because more use of DSF resulted in a wider villi surface. This shows that the use of DSF in feed can improve the performance of the small intestine so that the potential for nutrients that can be absorbed is greater. This is following the opinion of Kusuma et al., (2020) that the larger the surface area of the small intestinal villi, the higher the intestine's ability to digest the food that has been consumed.

The results of statistical analysis showed that the treatment had no significant effect (P>0.05) on the villous surface area. This indicates that the use of DSF in feed has a positive effect on the surface area of the villi of the small intestine of broilers. The highest villi surface area in this study was at P4 with the use of DSF as much as 10% in the feed. This is in line with the increase in villi height produced because the surface area of the villi is affected by the villi height. The higher the villi indicate the wider surface and the greater the opportunity for nutrient absorption (Choct, 2009 in Hidayat et al., 2020).

Effect of DSF use on the crypt depth

The absorption of nutrients in the small intestine can also be affected by the depth of the crypt. The size of the depth of the crypts starting from the bottom of the villi to the surface of the villi influences the ability to absorb nutrients. Based on the data presented in Table 3, the average depth of the crypts generated sequentially from the highest to the lowest is P4 (149.62 ± 11.38 m), P3 (148.51 ± 9.51 m), P2 (145.39 ± 13.71 m), P1 (132.65 ± 11.96 m), P0 (132.32 ± 10.18 m).

The resulting average crypt depth indicates that the higher the use of DSF in feed can increase the crypt depth. The resulting crypt depth has a positive correlation with villous height. This is comparable to the statement put forward by Sjofjan and Adli (2020) that the increase in crypt depth is in line with the increase in villi height. This indicates that the use of DSF can have a positive effect because it can increase the ability to absorb nutrients so that it can optimize the growth of broilers. The results of statistical analysis showed that the treatment had no significant effect (P>0.05) on the crypt depth of the small intestine.

This shows that the nutritional content of DSF does not have a bad effect on the physiological conditions of the small intestine, especially the depth of the crypts, because crypts play an important role in the replacement of epithelial cells in the villi. This is following the statement of Hu et al., (2012) in Hidayat et al., (2020) that crypts have an important role in villous renewal because the stem cell population continues to divide throughout life, thus enabling the replacement of villous epithelial cells. The more epithelial cells of the small intestine, the wider the surface and the greater the number of villi so that the weight of the small intestine will be heavier (Mistiani, et al., 2020).

Effect of DSF use on the digesta pH

The process of digestion and absorption of food substances that occur in the small intestine is influenced by the growth of bacteria, both in the form of pathogenic bacteria and non-pathogenic bacteria. One of the factors related to the growth of bacteria in the small intestine is the pH of the digesta. Based on the data presented in Table 3, the average digesta pH produced sequentially from the highest to P3 lowest is (6.71±0.25). **P**2 the $(6.61 \pm 0.23),$ P4 (6.57±0, 21). **P1** (6.55±0.30), P0 (6.50±0.28). The average digesta pH value from P0 to P3 increased but decreased again at P4. This indicates that the use of DSF of as much as 10% in the feed can produce a lower pH of the small intestine digesta than the use of DSF of as much as 5% and 7.5%. The low pH of digesta in the small intestine indicates that the balance of microorganisms can be maintained by increasing the growth of Lactic Acid Bacteria (LAB) and suppressing the growth of pathogenic bacteria (Yulianti et al., 2020).

The results of statistical analysis showed that the treatment had no significant effect (P>0.05) on the pH of the small intestine digesta. This is presumably because the flavonoid compounds contained in DSF affect the acidity level of the small intestine so that it can maintain a balance of bacterial growth. The high carbohydrate content in DSF also contributes to producing a low digesta pH value by increasing the growth of non-pathogenic bacteria in the small intestine because carbohydrates are non-pathogenic needed by bacteria, especially LAB in the metabolic process. LAB requires carbohydrates, especially soluble carbohydrates that will be used for metabolic processes and are used as an energy source for LAB (Widodo et al., 2015).

This indicates that the use of DSF up to 10% in feed can produce a low digesta pH so that it can improve the physiological condition of the small intestine and can increase the number of villi and villi height. Effect of DSF use on the digesta viscosity The important role of digesta pH in the digestion and absorption of nutrients correlates with the high and low viscosity of the digesta. High digesta viscosity is caused by the lower pH value so that the digesta rate runs slowly which can improve digestion in the stomach and intestines (Cahyaningsih et al., 2013). Based on the data presented in Table 3, the average digesta viscosity produced sequentially from the highest to the lowest is P4 (111±0.00 mPaS), P3 (111±0.00 mPaS), P1 (110.25±0.50 mPaS), P2 (110±0.00 mPaS), P0 (110±0.00 mPaS). The average digesta viscosity produced in each treatment gave results that were not much different and the increase in the use of DSF in feed was able to provide an increasing digesta viscosity.

This is thought to be due to DSF's high fiber content so that it can increase digesta viscosity. This is equivalent to the research results of Tabbook et al., (2006) that fiber in dates can increase the viscosity of digesta in the ileum. This increase in digesta viscosity indicates that the digestive process of feed is not optimal, and vice versa if the digesta viscosity is low, it indicates that the feed can be digested well in the small intestine. The results of statistical analysis showed that the treatment had a very significant effect (P<0.01) on the viscosity of the small intestine digesta.

This shows that the content of food substances in DSF, especially crude fiber content can affect the condition of digesta in the small intestine. Crude fiber which is high enough in DSF can cause the digestive process to be hampered because enzymes in digestion are not able to digest high crude fiber, causing the digesta viscosity to increase. The use of date seed flour in feed up to 10% was able to give good results on the number of villi, villi height, villous surface area, crypt depth, and pH of the digesta of the small intestine of broilers, but it has not been able to give good results on the viscosity of the digesta of the small intestine of broilers.

CONCLUSIONS

The use of date seed flour in feed up to 10% was able to give good results on the number of villi, villi height, villous surface area, crypt depth, and pH of the digesta of the small intestine of broilers, but it has not been able to give good results on the viscosity of the digesta of the small intestine of broilers.

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