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The relative weight of internal organs and digestive tract in native chickens age 12 weeks that are given various levels of BSF larvae meal (*Hermetia illucens* L) in the ration

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Abstract. The feed consumed is one factor that influences the development and anatomy of the internal organs and the digestive tract. This study was conducted to determine the effect of BSF larvae meal on the ratio of internal organ weight in native chickens. According to a completely randomized design, one hundred and forty o*ne-day-old chicken* (DOC) were randomly assigned to one of five treatments with four replications for each treatment. The treatment were: P0 = basal ration + 15% fish meal + 0% BSF larvae meal, P1 = basal ration + 11.25% fish meal + 3.75% BSF larvae meal , P2 = basal ration + fish meal 7.5 % + 7.5% BSF larvae meal , P3 = basal ration + 3.75% fish meal + 11.25% BSF larvae meal , P4 = basal ration + 0% fish meal + 15% BSF larvae meal . The results showed that P3 (3.75% fish meal + 11.25% BSF larvae) had a significant effect ($P<0.05$) on the percentage of heart weight and the percentage of the small intestine weight. Also, the treatment tends to increase the percentage of caecum weight. However, the treatment did not affect the liver, gizzard, pancreas, spleen, and colon weight. In conclusion, the use of BSF larvae meal does not negatively affect the internal organs and digestive tract of native chickens.

1. Introduction

The native chickens are chickens that have adapted to the tropical environment of Indonesia. It is generally maintained freely around the house, with extensive monitoring traditionally looking for its feed to grow irregularly. Native chicken is one of the livestock that produces output in the form of meat and eggs that have a distinctive taste and texture and high adaptability to the environment and more resistant to disease. One of the weaknesses of the village chickens is their low productivity. To increase the potential of village chickens, there needs to be improved maintenance management and improvement of genetic quality by crossing with chickens that have high [1].

Efforts to improve the genetic quality of native chickens through crosses are expected to be obtained offspring with high productivity, adaptability with the right environment, and disease resistance. However, the resulting genetic quality will be able to appear optimal when followed by improved feed quality. In its maintenance, chicken cattle require a cost to feed more than 70% of the total production cost [2]. Fish meal is one of the main feed ingredients of protein sources in the rations of native chickens. At this time, the price of fishmeal is relatively high and limited, which encourages imports. Based on these conditions, efforts to find alternative feed ingredients for protein sources are significant.

The use of alternative feed ingredients is expected to reduce feed costs. Black Soldier Fly (BSF) is one of the insects whose characteristics and nutrient content are studied. The BSF or maggot larvae form the first cycle (larva) of the black soldier fly, which then metamorphoses into adult flies. The percentage of BSF larvae's nutritional content is generally relatively high, which is 44.26% protein and 29.65% fat. BSF larvae are rich in various types of antimicrobial peptide (AMP), which have inhibitory activity against multiple types of pathogenic microorganisms [3]. The values of amino acids, fatty acids, and minerals in the larvae are also not inferior to other protein sources. The BSF larvae can be used as alternative feed ingredients for protein sources to replace fish meal.

The food consumed is one factor that influences the development and anatomy of the internal organs and digestive tract in poultry. Utilization of manure meals from the degradation of black fly larvae up to 15% produces the same egg weight, yolk weight, and egg mass in native chickens [4]. The presence of crude fiber in the ration can increase the value of gizzard [5]. Choi *et al.*, (2012) stated that AMP-A3 as much as 60–90 mg kg-1 has the same effect as avilamycin 15 mg/kg on performance, productivity, immunity, intestinal microorganism composition, and small intestine morphology of broiler Ross 305 broilers [6]. The digestive tract organ is essential in converting food into meat and eggs, which have high nutritional value. Therefore, it is necessary to research this study using BSF larvae meal to the relative weight of the internal organs and digestive tract of native chickens.

2. Material and Methods

2.1. Experimental design and feeding

The study used 140 DOC native chickens and an average bodyweight of 28.74±1.58 g was placed in 20 colony-shaped cages with a size of 80 x 60 x 50 cm, each cage containing seven chickens. The trial design used was a completely randomized design (CRD) with five treatments and four replications, consisting of 7 chickens. The treatment ration given was formulated as follows: P0 = basal ration + 100% fish meal (15% in ration) + 0% BSF larva meal (0% in ration), P1 = basal ration + 75% fish meal (11.25% in rations) + 25% BSF larva meal (3.75% in rations), P2 = basal rations + 50% fishmeal (7.5% in rations) + 50% BSF larvae flour (7.5% in rations), P3 = basal ration + 25% fish meal (3.75% in ration) + 75% BSF larva meal (11.25% in ration), P4 = basal ration + 0% fish meal (0% in ration) + 100% BSF larvae meal (15% in ration). The BSF larvae used in this study were BSF larvae that had been dried and ground into a meal. The BSF larvae that have become meal are mixed with other feed ingredients. The feed given in this study was a starter phase in the form of commercial feed for two weeks from commercial feed as an adaptation period, then continued with basal feed for the grower and finisher phases. Rations are given from one day to twelve weeks of age every morning and evening according to each maintenance phase's needs, while drinking water is given adlibitum. The composition and chemical composition of the ration is presented in table 1.

2.2. Measurement

The study was conducted until the chickens were twelve weeks old, and at the end of the study, one chicken per treatment replication. Previously, the chickens were fasted for about 3 hours and still given drinking water ad libitum, was continued weighing to determine the live weight, then slaughtering and taking internal and digestive organs (liver, heart, gizzard, pancreas, spleen, small intestine, caecum, and colon). Organs that have been taken and cleaned of the remaining dirt and weighed with analytical scales. The data obtained is calculated using the following formula [7]:

$$\text{The relative weight of organ} = \frac{\text{Weight of organ}}{\text{live weight}} \times 100\%$$

2.3. Statistical analysis

Data were analyzed using analysis of variances (ANOVA) according to a completely randomized design. The significant effects of treatments were further determined using *Duncan's Multiple Range Test* at a 5% level of significance [8].

Table 1. Feedstuff and chemical composition of the experimental diets.

Feedstuff composition (%)	Treatment				
	P0	P1	P2	P3	P4
Maize	64	64	64	64	64
Fine rice bran	12.5	12.5	12.5	12.5	12.5
Soybean meal	1	1	1	1	1
Fish meal	15	11.25	7.5	3.75	0
BSF larvae meal	0	3.75	7.5	11.25	15
Coconut cake meal	2.5	2.5	2.5	2.5	2.5
Vegetable oil	2	2	2	2	2
CaCO3	2	2	2	2	2
Premix	1	1	1	1	1
Total	100	100	100	100	100
Nutrien composition*)					
Protein (%)	18.46	18.44	18.42	18.39	18.37
Fat (%)	3.31	3.79	4.27	4.75	5.24
Crude fiber (%)	7.11	7.43	7.76	8.09	8.42
Ca (%)	0.99	0.97	0.96	0.94	0.93
P (%)	0.91	0.81	0.71	0.6	0.5
Ash (%)	8.48	7.96	7.44	6.92	6.4
BETN (%)	61.56	59.33	59.06	58.79	58.53
ME**(kkal/kg)	3063,16	3090,54	3117,92	3145,30	3172,68

*) Calculated based on the results of the analysis and calculation of proximate analysis table data at the Laboratory of Animal Feed Chemistry and Nutrition, Faculty of Animal Husbandry, Universitas Hasanuddin. **) Metabolic Energy (EM) value is calculated based on the Balton formula: EM = 40.81 x (0.87 (PK + (2.25 x LK) + BETN) + 2.5

3. Result and discussion

In this study, what is meant by internal organs are the liver, heart, gizzard, pancreas, spleen, small intestine, caecum, and colon. All are converted into percent of live weight and are presented in Table 2. Based on the weight of internal organs obtained, it can be seen that the organ weight in each treatment as a whole is still within normal weight limits and is yet following its live weight without being influenced by the presence of treatment rations. The percentage of internal organ weight varies and can be affected by type, age, size, and animal [9].

The liver has a detoxification function carried out by liver enzymes, namely by converting potentially harmful substances into physiologically inactive substances [10]. It can see that the average percentage of liver organs ranges from 1.94±0.32% to 2.40±0.32%. This is following the expressed weight of a heart that is, ranging from 1.70 to 2.80% of live weight [10,11]. An increase in the percentage of liver weight is thought to increase liver activity caused by the secretion of bile and breaking down excess protein into uric acid. An enlarged liver indicates increased work, and the cause is neutralizing toxins. Ressang (1984) reported that the liver functions as a detoxification of toxins, and if the liver is abnormal, it is characterized by an enlargement or reduction of the liver [9]. The observation results showed no swelling and changes in liver color so that the liver condition was normal. This indicates that the liver works typically, not affected by the use of BSF larvae meal. The work of BSF larvae meal will mainly affect environmental conditions in the small intestine.

Table 2. Percentage of organ weight in native chickens aged 12 weeks.

Parameter (%)	Treatment					P-Value
	P0	P1	P2	P3	P4	
Liver	1.94±0.32	2.33±0.29	2.14±0.26	2.40±0.32	2.26±0.32	0.27
Heart	0.38±0.08^{a}	0.50±0.03^{b}	0.42±0.02^{ab}	0.51±0.07^{b}	0.48±0.10^{a}	0.01
Gizzard	2.65±0.36	2.71±0.15	2.77±0.07	2.89±0.26	2.75±0.58	0.88
Pancreas	0.19±0.02	0.21±0.07	0.26±0.03	0.29±0.08	0.24±0.02	0.14
Spleen	0.16±0.02	0.29±0.06	0.27±0.09	0.31±0.10	0.25±0.13	0.22
Small intestine	2.20±0.14^{a}	2.40±0.36^{ab}	2.62±0.03^{b}	3.13±0.06^{b}	2.66±0.13^{c}	0.00
Caecum	0.55±0.10	0.55±0.11	0.58±0.07	0.60±0.11	0.56±0.08	0.07
Colon	0.18±0.04	0.20±0.03	0.21±0.02	0.19±0.04	0.23±0.12	0.41

^{ab}Different superscripts on the same row showed significant differences (P<0.05).

The results of the analysis of variance showed that the use of BSF larvae meal in native chicken feed significantly (P<0.05) increased heart weight in treatment P3 compared to other treatments, this indicates that one form of adaptation of native chickens to the use of BSF larvae meal substituted with a fish meal in the ration. The research results show that the average heart weight percentage ranges from 0.38–0.51%. These results are still within the average heart weight range as [11] that the percentage of chicken heart weight ranges from 0.42%–0.75% of live weight. Heart swelling in P3 treatment was caused by the presence of a higher intake of antinutrients than controls. Ressang (1984) stated that if the blood contains toxins and antinutrients, there will be an increase in excessive contractions, causing swelling of the heart. The heart's size is very much influenced by the type, age, size, and activity of the animal and has a great power to adjust in its body [9].

Based on the research results, it can be seen that the average gizzard weight ranges from 2.65–2.89% of the live weight and is above the stated gizzard weight range. Putnam (1991) stated that the weight of broiler gizzards at the age of 42 days was about 1.60–2.30% of the live weight [11]. The results of the analysis of variance showed that the P3 treatment (3.75% fish meal + 11.25% BSF larva meal) did not affect the gizzard weight percentage of native chickens. The lowest gizzard weight was obtained in treatment P0 (2.65±0.36%), and the heaviest was obtained in treatment P3 (2.89±0.26%). Giving BSF larvae meal did not harm gizzard work. This means that the use of BSF larvae meal in the feed does not ease the gizzard workload. The gizzard is an organ that has unique muscles which play a significant role in the mechanical digestion of feed. The gizzard functions to shrink feed particles mechanically and have a thick muscle layer. The factors affecting gizzard are the size of the livestock and the type of feed consumed. During this research, the feed used is a meal with high enough crude fiber so that the gizzard muscles work more actively, and the longer they get thicker so the the gizzard size is more significant. The gizzard weight was influenced by livestock size, ration typesetting, and feeding phase. If the ration given has high crude fiber content, the gizzard work will be more massive and increase the gizzard's size and weight [5].

The pancreas is located in the small intestine between the folds of the duodenum. The mean percentage of the pancreas weight produced ranged from 0.19 to 0.29% of live weight. This result is higher than research that the average percentage of poultry pancreas weight is around 0.25–0.40% of live weight [12]. Based on the research data, the use of BSF larvae meal did not affect the pancreas (table 2). This shows that the use of BSF larva meal with fish meal substitution does not harm the pancreas' work. One of the pancreas' functions in poultry is to secrete pancreatic juice in the digestion of starches, fats, and proteins by its enzymes. The ration's crude protein content affects the work of the pancreas to secrete the enzymes trypsin, amylase, and lipase into the small intestine. Apart from its function in secreting pancreatic juice, the pancreas also functions to secrete the hormone insulin. Increased weight of the pancreas, which produces lipolytic, amylolytic, and proteolytic enzymes, is one form of adaptation to meet digestive enzymes [13]. The factors that affect the percentage of pancreatic weight are genetic, behavioural, and environmental factors. The poultry pancreas' two main functions are related to the use of ration energy, namely exocrine and endocrine. The exocrine function is to supply enzymes that digest carbohydrates, proteins, and fats into the small intestine and then carry it to the duodenum and secrete amylase, trypsin, and lipase enzymes to receive carbohydrates, proteins, and fats.

Meanwhile, the endocrine regulates nutrition and uses energy absorbed by the body in the digestive process [14,15].

The results of the analysis of variance showed that the use of BSF larvae meal did not affect the spleen weight percentage of native chickens (table 2). The average percentage of spleen weight produced ranged from 0.16–0.31% of live weight. This condition is higher than the percentage of spleen weight according to [11], namely 0.18%–0.23% of live weight. Ressang (1984) stated in his research, the spleen functions in forming lymphocytes and is associated with the formation of antibodies that will change in size if there are toxic or anti-nutritional substances [9]. This shows that the use of BSF larvae meal in the ration does not harm the work of the spleen organs. As the spleen functions as a blood filter and iron storage to be reused in haemoglobin synthesis [16].

Based on the data presented in table 2, the small intestine relative weight results ranged from 2.20–3.13%. The results of the analysis of variance showed that the use of BSF larvae meal substituted with the fish meal had a significant effect ($P<0.05$) on the percentage of the small intestine weight of native chickens compared to other treatments. It is suspected that BSF larvae meal has high crude fiber, thus contributing to increasing the fiber content in the treated feed. It may also be due to the addition of intestinal villi in digesting antinutrients in the use of BSF larvae meal to substitute fish meal so that the intestinal weight increases. According to Amrullah (2004), this change in the digestive tract's size is also followed by the number of intestinal villi and the ability to secrete digestive enzymes. The same nutrient absorption ability will follow the same digestive tract size. The small intestine's size is influenced by the small intestine's capacity to digest and absorb food substances. The small intestine functions to hydrolyze simple carbohydrates to be absorbed by the body as an energy source, while crude fiber cannot be degraded [5]. The P0 treatment of the small intestine more easily absorbs food substances. The intestine works lighter in absorption than treatment with the use of BSF larvae substituted in fish meal. The weight of the intestine indicates that the intestine is working harder to absorb nutrients. The use of BSF larva meal in the treatment P1, P2, P3, and P4, which in the ration contains high enough crude fiber, can help in the digestion of crude fiber. Besides, there is an antimicrobial peptide (AMP) in BSF larvae, which has inhibitory activity on various types of pathogenic microorganisms. Park et al (2014) and BSF larvae also contain monolaurin compounds derived from lauric acid, which are antibacterial [3], which work effectively against *Staphylococcus aureus* and *Escherichia coli* found in the digestive tract of poultry so that absorption of nutrients in the intestine is not inhibited [15].

The caecum is an organ of the digestive tract that functions as a microbial digestion area to digest nutrients that are not absorbed in the small intestine, especially fiber and nitrogen. Based on the data presented in table 2, it was obtained that the relative weight results for caecum ranged from 0.55 to 0.60%, this result was higher than reported. Delman and Brown (1992) that is 0.46–0.49% of the live weight [16]. Caecum weight is influenced by the amount of feed and the amount of fiber not absorbed in the small intestine [14]. The results showed that the P3 treatment (3.75% fish meal + 11.25% BSF larvae meal) tended ($P = 0.07$) to increase in caecum weight relative to the control (P0). This indicates that the increase in fiber affects the function of the caecum. In non-ruminant livestock, caecum can better utilize fiber. Skřivanová et al (2014) reported that the increase in caecum weight was due to increased digestion activity of nutrients that are not absorbed in the small intestine due to reduced digestibility of feed in the intestine [17].

Table 2 shows that the use of BSF larvae meal in the P4 treatment (15% BSF larva meal + 0% fish meal) did not affect the colon's relative weight compared to the control (P0). The average relative weight of the colon ranges from 0.18–0.23%. This result is below the range reported by Sharifi et al., (2012) that is 0.3–0.43% of live weight [18]. This indicates that the ration given does not improve the colon's performance in the absorption of food substances, especially crude fiber. The large intestine is the organ for distributing food waste from the small intestine to the cloaca and the absorption site for water and minerals. Low colon activity causes the low relative weight of the large intestine to below. Sumiati and Sumirat (2003) in his research reported that the absorption of crude fiber in the large intestine took place in small amounts so that the increase in the weight of the colon of native chickens did not affect [19].

4. Conclusion

The use of BSF larva meal as a substitute for fish meal in native chicken rations had a significant effect ($P<0.05$) on the percentage of heart and small intestine weight. Besides, the treatment tends to increase the percentage of caecum weight. However, the treatment did not affect the percentage of liver, gizzard, pancreas, spleen, and colon weight. The use of BSF larvae meal did not harm the relative weight of internal organs and the digestive tract of native chickens.

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