Effects of L-threonine and L-tryptophan supplementation on the body weight and internal organs weight of native chickens aged 14 weeks

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Abstract. The aim of this study was to determine the effect of L-threonine and L-tryptophan supplementation on the body weight and internal organs weight of native chickens aged 14 weeks. The experiment used 112 six-week old native chickens. They were allocated to four treatments and four replicates. The four dietary treatments were: (1) T₀ (control); (2) T₁ (supplementation of 0.25% L-threonine and 0.07% L-tryptophan); (3) T₂ (supplementation of 0.58% L-threonine and 0.14% L-tryptophan); and (4) T₃ (supplementation of 0.85% L-threonine and 0.20% L-tryptophan). The result showed that the body weight of T₀, T₁, T₂ and T₃ were 1044.28, 1081.91, 1115.95 and 1151.34 g/bird, respectively. The liver, pancreas, and gizzard weights and the intestinal length were 2.56, 2.66, 2.81 and 2.91 g/bird; 3.90, 4.17, 4.43 and 4.46 g/bird; 26.00, 28.63, 30.26 and 31.28 g/bird; 124.80, 131.90, 137.88 and 139.73 cm/bird, respectively. The result indicated that the body weight, liver, gizzard, and intestinal length affected by level of L-threonine and L-tryptophan (P<0.01). It was concluded that supplementation with 0.85% L-threonine and 0.20% L-tryptophan to the diet of native chickens resulted in optimal body weight and internal organs weight at 14 weeks.

1. Introduction

All feed formulations with alternative ingredients must be based on digestible amino acids. Amino acids are important in poultry nutrition not only for protein deposition, but also for other metabolic functions, such as improving immune and gut function. Recent advances in poultry industry have been made by supplementing synthetic amino acids such as L-lysine, DL-methionine, L-threonine and L-tryptophan, L-valine in commercial poultry birds to reduce the feed cost. As result, the poultry nutritionists may reduce the crude protein (CP) level while maintaining the bird performance and profitability [1]. It has been reported that feed feeding with 2,971 kcal kg⁻¹ metabolizable energy (ME) with 15.26% CP, 1.47% Ca and 0.59% P supplemented with 0.25% methionine and 0.60% lysine during the grower phase increased native chicken productivity during growing phase [2]. Threonine and tryptophan are the third and the fourth critical of amino acids after methionine and lysine.

Threonine plays a role in bone formation, controlling the health, and immune system of the body, liver, and nerves, maintaining body proteins, and involved in digestive function and fat metabolism. Tryptophan functions as a serotonin trigger, stimulates niacin production, increases appetite and feed efficiency, stimulates growth and increases body weight [3]. Working with native chickens, Lisnahan et al. [2] reported growth performance of native chickens in the grower phase fed methionine and...
lysine-supplemented cafeteria standard feed. The purpose of this study was to determine the effect of L-threonine and L-tryptophan supplementation on body weight and internal organ weight of native chickens.

2. Material and methods

2.1. Experimental birds and management of experimental diets

This research was conducted in Sasi, Kefamenanu, East Nusa Tenggara from March to May 2019. One hundred and twelve six-week old native chickens were used in this experiment. They were randomly assigned to the four diets in a completely randomized design with four replicates. The four treatments were: (1) T0 (control feed without supplementation of L-tryptophan and L-threonine; (2) T1 (supplementation of 0.07% L-tryptophan and 0.25% L-threonine; T2 (3) (supplementation 0.14% L-tryptophan and 0.57% L-threonine) and (4) T3 (supplementation of 0.20% L-tryptophan and 0.85% L-threonine). The feed ingredients and nutrient composition of each treatment are shown in Table 1 [4]. Feed and water were offered ad libitum during the study.

2.2. Data collection and analysis

Parameters observed were body weight, liver, pancreas, gizzard, and small intestine weights and intestinal length. The data obtained were analyzed by the analysis of variance [5].

| Table 1. Composition (%) and nutrient content (%DM) of experiment diets during the growth phase (6-14 weeks). |
|---------------------------------------------------------------|
| **Ingredients** | **Treatments (%)** | **T0** | **T1** | **T2** | **T3** |
| Yellow corn | 50.50 | 50.50 | 50.50 | 50.50 |
| Rice bran | 36.48 | 36.15 | 35.76 | 35.42 |
| Soybean meal | 5.18 | 5.18 | 5.18 | 5.18 |
| Fish meal | 6.48 | 6.48 | 6.48 | 6.48 |
| Limestone | 0.30 | 0.30 | 0.30 | 0.30 |
| Vitamin premix | 0.29 | 0.29 | 0.29 | 0.29 |
| DL-methionine | 0.23 | 0.23 | 0.23 | 0.23 |
| L-lysine HCl | 0.55 | 0.55 | 0.55 | 0.55 |
| L-Threonine | 0.25 | 0.25 | 0.58 | 0.85 |
| L-Tryptophan | 0.07 | 0.14 | 0.20 |
| Total | 100.00 | 100.00 | 100.00 | 100.00 |
| **Calculated analysis** |  | 2949.15 | 2941.77 | 2933.11 | 1925.54 |
| Metabolized energy (kcal kg⁻¹) | 15.34 | 15.30 | 15.26 | 15.22 |
| Crude protein (%) | 5.93 | 5.90 | 5.87 | 5.84 |
| Ether extract (%) | 8.01 | 7.98 | 7.94 | 7.90 |
| Ash (%) | 7.31 | 7.26 | 7.20 | 7.15 |
| Crude fiber (%) | 0.25 | 0.25 | 0.25 | 0.25 |
| Methionine (%) | 0.60 | 0.60 | 0.60 | 0.60 |
| Lysine (%) | 0.05 | 0.30 | 0.63 | 0.90 |
| Threonine (%) | 0.03 | 0.10 | 0.17 | 0.23 |
| Tryptophan (%) | 1.24 | 1.24 | 1.23 | 1.23 |
| Phosphorus (%) | 0.59 | 0.59 | 0.58 | 0.58 |

3. Results and discussion

The results of this study indicated that the body weight and internal organ weight of native chickens at 14 weeks of age were affected by treatments (Table 2). Supplementation of 0.85% at T3 yielded the highest body weight compared to other treatments. In addition, essential amino acids threonine and tryptophan in the feed also stimulated faster growth. Rapid growth must be stimulated with an increase
in the content of threonine and tryptophan. Body weight of native chickens at the age of 14 weeks with the addition of DL-methionine and L-lysine (without L-threonine and L-tryptophan) in the feed was 1,064.00 g/bird [2]. In this study, with the addition of 0.20% L-tryptophan and 0.85% L-threonine, the body weight reached 1,151.34 g/bird. This findings is in accordance with previous study where threonine had a significant effect on feed intake and body weight [3]. Threonine levels significantly affected on feed conversion. Supplementation of L-threonine and L-tryptophan generated a better chicken performance since they are the third limiting amino acids after methionine and lysine [1,6].

**Table 2.** Average of body weight and digestive organs of native chicken at 14th week of age.

| Parameters                  | T0            | T1            | T2            | T3            |
|-----------------------------|---------------|---------------|---------------|---------------|
| Body weight (g/bird)        | 1044.28±21.03 | 1081.91±5.49  | 1115.95±14.97 | 1151.34±28.38 |
| Liver (g/bird)              | 27.29±1.13    | 28.78±0.90    | 30.79±1.44    | 32.75±0.74    |
| Liver (%)                   | 2.61±0.12c    | 2.66±0.09bc   | 2.76±0.15ab   | 2.85±0.12a    |
| Pancreas (g/bird)           | 3.90±0.21     | 4.17±0.19     | 4.43±0.25     | 4.46±0.71     |
| Pancreas (%)                | 0.37±0.01     | 0.39±0.02     | 0.40±0.02     | 0.39±0.06     |
| Gizzard (g/bird)            | 28.00±1.2c    | 28.63±1.14b   | 30.26±0.46b   | 31.28±1.36a   |
| Gizzard (%)                 | 2.68±0.13bc   | 2.65±0.10b    | 2.71±0.03b    | 2.72±0.15a    |
| Intestine length (cm/bird)  | 124.80±6.09c  | 131.90±2.67b  | 137.88±2.95a  | 139.73±1.09a  |
| Small intestine weight (g/bird) | 36.44±1.62c | 38.02±1.12bc  | 39.84±0.88b   | 42.16±1.09a   |
| Small intestine (%)         | 3.42±0.19b    | 3.52±0.12b    | 3.63±0.12ab   | 3.75±0.14a    |

Superscript on the same line indicates significant difference (p<0.01)

Addition of 0.23 - 0.31% tryptophan in the feed increased broiler chicken weight [7] which positively correlated with the weight of internal organs namely gizzard, liver and intestine. Supplementation of threonine and tryptophan caused a more balance of essential amino acids needed. Balanced nutrient made better growth including gizzard, liver, and intestinal growth. The larger internal organs caused more feed to be consumed, digested, and absorbed. Gizzard was part of the digestive tract functioning as a mechanical stomach. Consumed feed and digestive juice from the salivary glands and proventriculus pass into the gizzard for grinding, mixing, and mashing [8]. Previous observation showed that the gizzard weight of native chickens was 4.20% of the body weight [2]. One of the important roles of the liver in the process of digestion is producing bile sap which is channeled into the duodenum. Feed that enters the duodenum stimulates the gallbladder to shrink and expel bile sap into the duodenum which can help fat absorption by the small intestine [9]. The liver percentage of broiler chickens increased following the supplementation of 0.2% tryptophan in feed [10] and addition of threonine levels at aged 42 days [11]. In our previous works, we found that the percentage of pancreas in native chickens was 0.39% of the body weight [2]. According to Cahyono et al. [12], pancreatic weight was 0.28% of body weight. Pancreas secrete trypsin, amylase, and lipase to the intestinal duodenum. The intestine is a major part of the digestion and absorption of nutrients.

The intestine length of native chickens at 12-week-old was 129.10 cm [11]. Significant differences were found in the interaction of crude protein and threonine for the relative weight and length of the duodenum and jejunum [8]. After methionine and lysine, threonine and tryptophan have a very important role in the metabolism and synthesis of the body proteins. Threonine maintains a balance of proteins and amino acids, maintains intestinal function and improves immune responses [1,3], while tryptophan plays a role in the process of protein biosynthesis, constituents of muscle mass and stimulates the immune response, enhancing the immune system, maintaining internal proteins to be stable, inducing growth and increasing antibodies, serotonin precursors and controlling aggressiveness [3,4,7,10]. The use of tryptophan along with lysine controls stress, increases palatability and body weight and decreases feed conversion [12]. Tryptophan can regulate feed intake, behavior, growth, immunity, protein synthesis and intestinal integrity of livestock [13]. Tryptophan deficiency affects body protein synthesis, but also damages the synthesis of important neurotransmitters such as...
serotonin and melatonin. High tryptophan levels have a positive effect on systemic immune responses and growth performance in poultry [14]. The poultry feed that deficiency of niacin, tryptophan plays a role in reducing depression in the weight gain and feed intake[15].

4. Conclusion
Supplementation with 0.85% L-threonine and 0.20% L-tryptophan on the feed of native chickens aged 6-14 weeks resulted in optimal body weight and internal organs weight.

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