



Research article

Effect of L-arginine and L-lysine supplementation in low-protein feeds on the growth performance of Native chickens in the starter phase

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Abstract

Some feeds that are used as protein sources, such as fish meal and soybean meal, have a relatively high cost. Therefore, synthetic amino acids, such as arginine and lysine, can be used as alternative protein sources. This study determined the effect of L-arginine and L-lysine in low-protein feed on the growth performance of one- to six-week-old Native chickens. A total of 250 one-week-old Native chickens mixed sex were divided into five treatments and five replications based on a completely randomized design. Each replication consisted of 10 chicks. The treatments were T0 (0.46% L-arginine + 0.42% L-lysine + 19% CP), T1 (0.56% L-arginine + 0.51% L-lysine + 18% CP), T2 (0.66% L-arginine + 0.60% L-lysine + 17% CP), T3 (0.76% L-arginine + 0.69% L-lysine + 16% CP), and T4 (0.86% L-arginine + 0.78% L-lysine + 15% CP). The treatment T2 had the highest feed consumption (612.81 ± 4.82 g/chick/6 weeks), while treatment T3 had the highest body weight and body weight gain rate (296.08 ± 1.09 g/chick and 265.48 ± 1.57 g/chick/6 weeks, respectively). The best feed conversion ratio was seen for treatment T3 (2.29 ± 0.40) while treatment T3 had the highest carcass weight and carcass percentage (177.57 ± 2.04 g/chick and 59.97 ± 0.55 %/chick, respectively). Statistical analysis showed that the treatment had a significant effect on feed consumption ($P < 0.05$), body weight, weight gain, feed conversion ratio, carcass weight, and carcass percentage ($P < 0.01$). The increase in L-arginine and L-lysine of 0.76 and 0.69% from 0.46 and 0.42%, respectively, suppressed the use of CP by 19.00% to 16.00% in the starter phase.

Keywords: Amino acids, Carcass, Crude protein, Native chicken performance

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INTRODUCTION

Protein is the important thing on native chicken's growth, and the protein source ingredients usually fish meal and soybean meal. Seventy percent of the cost of producing native chicken lies in the feeds, and the most expensive part of the feeds is protein source ingredients. Therefore, alternative low-cost protein sources, such as synthetic amino acids, are necessary. The use of essential amino acids, such as methionine, lysine, threonine, and tryptophan, in Native chickens has been reported as a standard requirement (Lisnahan and Nahak, 2020). Additionally, one of the essential amino acids in metabolism is arginine. Arginine functions as a trigger for nitric oxide, which is a compound that plays a role in improving blood circulation to carry nutrients and functions as a growth stimulant (He et al., 2021). Balnave and Barke (2019) stated that arginine together with glycine and methionine are precursors in forming creatine.

Arginine, along with cysteine, glutamine, leucine, proline, and tryptophan, are amino acids that regulate key metabolic pathways that are required for maintenance, growth, reproduction, and immunity (Furukawa et al., 2021). These amino acids maximize feed efficiency and protein aggregation, reduce adiposity, and enhance livestock health. Toprak et al. (2021) described arginine as one of the most versatile amino acids in animal cells. It is required for the synthesis of several compounds, such as ornithine, polyamines (spermidine, spermine, and putrescine), proline, creatine, protein, nitric oxide, and citrulline. Arginine also plays a role in increasing the release of insulin, growth hormone, and IGF-I in the bloodstream (Birmani et al., 2019; Tsugawa et al., 2019; Yu et al., 2022). Poultry cannot synthesize arginine and lysine and therefore depend on the supply of amino acids in feed.

Arginine and lysine are necessary for Native chickens. The high demand for arginine and lysine is indicated by a lack of endogenous arginine and lysine synthesis and the high rate of protein deposition due to rapid growth in the starter and grower phases, especially for muscle formation. Lysine plays a significant role in the manufacturing of proteins in muscles and other body tissues, along with vitamin C, to form collagen and other body connective tissues (cartilage and joints) and is used in low protein feed (Lisnahan et al., 2022). Castro et al. (2019) stated that deficiency of the amino acid lysine in feed could increase feed intake, lower feed efficiency and uric acid excretion, and decrease nitrogen retention.

The use of arginine must be balanced with lysine. If excessive arginine is followed by an increase in lysine or vice versa, then it will result in antagonism (He et al., 2021). Furthermore, when growth inhibition is caused by the antagonism of a single amino acid, it can be corrected by amino acids that are antagonistic to that single amino acid. Oliveira et al. (2022) stated that excessive lysine markedly increases the need for arginine, and if lysine is even slightly greater than that required in comparison with arginine, it causes stunted growth. However, excessive arginine affected feeds are low in lysine content (Balnave and Barke, 2019).

The standard balance of arginine and lysine in starter and grower phase chickens has been reported by [Lisnahan et al. \(2022\)](#), namely, 1.10. Based on this standard, the use of crude protein can be reduced to decrease feed costs while maintaining the growth performance of Native chickens. Hence, this study determined the effect of increasing L-arginine and L-lysine in low-crude protein feeds on the growth of starter-phase chickens.

MATERIALS AND METHODS

Ethical approval

The study protocol was approved by the animal ethics committee of the Animal Husbandry Program Study, Faculty of Agriculture, University of Timor, Indonesia, with 12/UN60.1/SR/2022 number recommendation, April 25, 2022.

Study period and location

This research was conducted in Sasi village, Kefamenanu. It was held for two months starting from May to June, 2022. Laboratory analysis was conducted at the Laboratory of Faculty of Agriculture, University of Timor, Kefamenanu, East Nusa Tenggara, Indonesia.

Animal and feed preparation

The research materials consisted of 250 Native chickens a-day-old with initial body weight was 30.56 ± 0.58 g, 25-litre cages measuring 2 x 1 x 0.5 metres (10 chicks/cage), 25 feed containers (2 kg/container), 25 drinking water containers (1 litre/container), incandescent lamps (Each cage was given the intensity of lighting with a 40-watt incandescent lamp), and O thirst scales with a capacity of 5 kg and an accuracy of 0.01 g. The feed ingredients were yellow corn, rice bran, soybean meal, fish meal, di-calcium-phosphate, premix vitamins, Dl-methionine, L-lysine, L-arginine, L-threonine, and L-tryptophan. The feed ingredients are mixed according to the treatment in crumble form.

Dietary treatment and feeding duration

This study employed a completely randomized design with five treatments and five replications. Each replicate consisted of ten Native chickens. The treatments are as follows:

T_0 : 0.46% L-arginine + 0.42% L-lysine + 19% CP

T_1 : 0.56% L-arginine + 0.51% L-lysine + 18% CP

T_2 : 0.66% L-arginine + 0.60% L-lysine + 17% CP

T_3 : 0.76% L-arginine + 0.69% L-lysine + 16% CP

T_4 : 0.86% L-arginine + 0.78% L-lysine + 15% CP

Feed on the T_0 treatment was based on the recommendation of [Lisnahan et al. \(2022\)](#). Feeding is done 3 times a day, every 07.00; 11.00 and 15.00.

Data collection and analysis

The measured parameters were body weight, weight gain, feed consumption, feed conversion ratio, carcass weight, and carcass percentage. Weighing of chickens was carried out every week for 6 weeks. Weight gain is the difference between final body weight minus initial body weight. Feed consumption data is obtained every day, namely the feed given minus the remaining feed. Feed conversion ratio is feed consumption divided by weight gain. The carcass weight was obtained after deducting blood, feathers, head and neck, legs, internal organs (digestive tract, liver, heart, spleen, pancreas, bile), and abdominal fat. Carcass percentage is obtained from carcass weight divided by body weight multiplied by 100%. Samples for carcass weight were selected from the average weight of chickens, namely 2 chickens per cage (1 male and 1 female).

Data obtained were analysed by an analysis of variance based on the Completely Randomized Design (CRD) and Duncan's test (IBM SPSS Statistics 25). The mathematical formula for Completely Randomized Design is $Y_{ij} = \mu + \alpha_i + \varepsilon_{ij}$.

RESULTS

Body weight gain

The production of one- to six-week-old Native chickens is presented in Table 1, and development of body weight presented in Figure 1. The highest average body weight and weight gain of six-week-old Native chickens was T_3 . An analysis of variance showed that the treatment had a significant effect on the weight gain of Native chickens ($P < 0.01$). The body weight started to increase at T_1 with a level of 0.56% L-arginine + 0.51% L-lysine with a 1% decrease in CP from 19% to 18%. This weight gain was 1.17%. The body weight continued to increase when L-arginine and L-lysine were upgraded to 0.76% and 0.69% at 16% CP (T_3) by 1.00% compared to T_2 . When CP was reduced to 15%, with the addition of L-arginine and L-lysine at 0.86 and 0.78% (T_4), respectively, the body weight decreases by 4.46% compared to T_3 .

Table 1 Composition and nutrient content of the treatment feeds

Ingredients	Treatments (%)				
	T ₀	T ₁	T ₂	T ₃	T ₄
Yellow corn	58.61	60.46	62.33	64.18	66.04
Rice bran	17.00	17.00	17.00	17.00	17.00
Fishmeal	10.66	9.62	8.56	7.52	6.47
Soybean meal	10.00	9.00	8.00	7.00	6.00
Vitamin Premix	0.40	0.40	0.40	0.40	0.40
Dl-methionine ¹	0.40	0.40	0.40	0.40	0.40
L-tryptophan ²	0.35	0.35	0.35	0.35	0.35
L-threonine ²	0.70	0.70	0.70	0.70	0.70
L-lysine ³	0.42	0.51	0.60	0.69	0.78
L-arginine ³	0.46	0.56	0.66	0.76	0.86
Dicalcium-phosphate	1.00	1.00	1.00	1.00	1.00
Total	100.00	100.00	100.00	100.00	100.00
Calculated nutrients					
Metabolized energy (kcal/kg) ⁴	3019.54	3039.48	3060.21	3079.78	3099.93
Crude protein ⁴	19.00	18.00	17.00	16.00	15.00
Ether extract ⁴	5.27	5.28	5.28	5.28	5.28
Ash ⁴	7.91	7.48	7.04	6.61	6.18
Crude fibre ⁴	5.16	5.04	4.93	4.81	4.70
Methionine ⁵	0.44	0.43	0.43	0.43	0.42
Tryptophan ⁵	0.50	0.50	0.50	0.50	0.50
Threonine ⁵	1.00	1.00	1.00	1.00	1.00
Lysine ⁵	0.82	0.91	1.00	1.09	1.18
Arginine ⁵	0.90	1.00	1.10	1.20	1.30
Calcium ¹	1.50	1.50	1.50	1.50	1.50
Phosphorus ¹	0.60	0.60	0.60	0.60	0.60

Note: ¹Recommendation of [Lisnahan et al. \(2017a\)](#); ²Recommendation of [Lisnahan and Nahak \(2020\)](#); ³Recommendation of [Lisnahan et al. \(2022\)](#); ⁴Calculated based on the analysis results of the Biochemistry Laboratory, Faculty of Animal Science, UGM [Yogyakarta, 2017](#); ⁵Calculated based on the analysis results of the Organic Chemistry Laboratory, Faculty of Mathematics and Natural Sciences, UGM [Yogyakarta, 2018](#); T₀ is based on the arginine-lysine ratio (1.10) and a CP of 19.00% ([Lisnahan et al. 2022](#)); T₀ (0.46% l-arginine + 0.42% l-lysine + 19% CP); T₁ (supplementation of 0.56% l-arginine + 0.51% l-lysine + 18% CP); T₂ (supplementation 0.66% l-arginine + 0.60% l-lysine + 17% CP); T₃ (supplementation of 0.76% l-arginine + 0.69% l-lysine + 16% CP); and T₄ (supplementation of 0.86% l-arginine + 0.78% l-lysine + 15% CP). CP in fishmeal 59.74%, soybean meal 54.26%, yellow corn 9.15%, rice bran 11.06%; methionine in fishmeal 0.014%, soybean meal 0.335%; lysine in fishmeal 0.015%, soybean meal 0.621%.

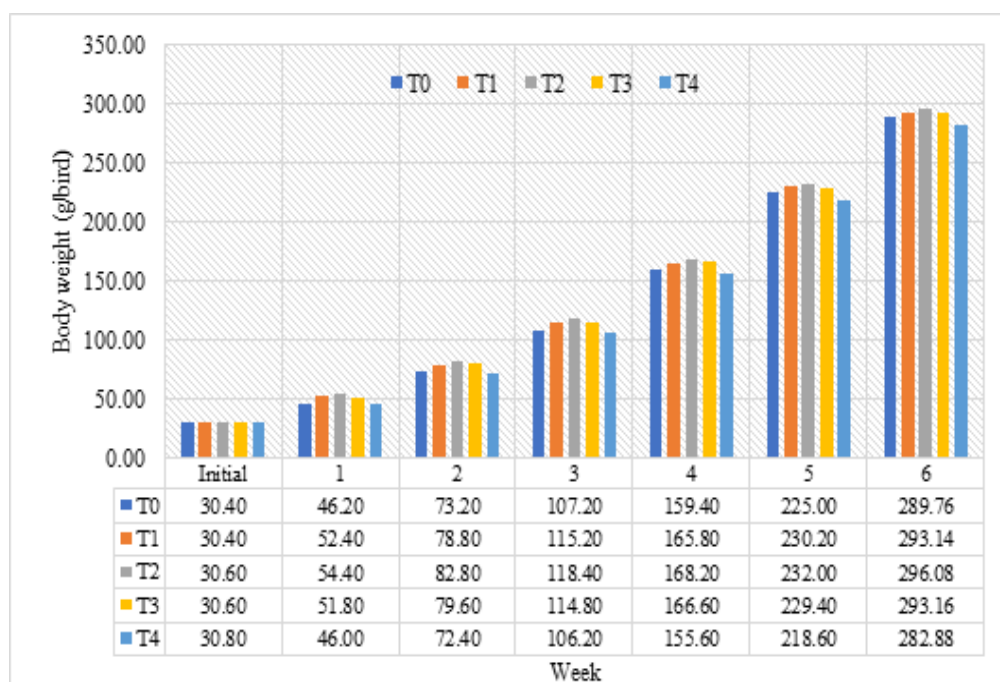


Figure 1 Development body weight of Native chicken at the starter phase.

Feed intake

The development of Native chickens feed consumption is presented in [Figure 2](#). The highest and lowest average consumption of Native chicken feed was at T_2 and T_0 , respectively ([Table 2](#)). An analysis of variance showed that the treatment had a significant effect on the consumption of Native chicken feed ($P < 0.05$). Supplementation with T_1 increased the consumption of Native chicken feed by 1.13%. When 0.66% L-arginine + 0.60% L-lysine + 17% CP (T_2) increased to 0.90%, the feed consumption continued to increase by 0.79% compared to T_1 , and this effect was the same at T_3 . The feed consumption decreased at T_4 by 0.91% compared to T_3 .

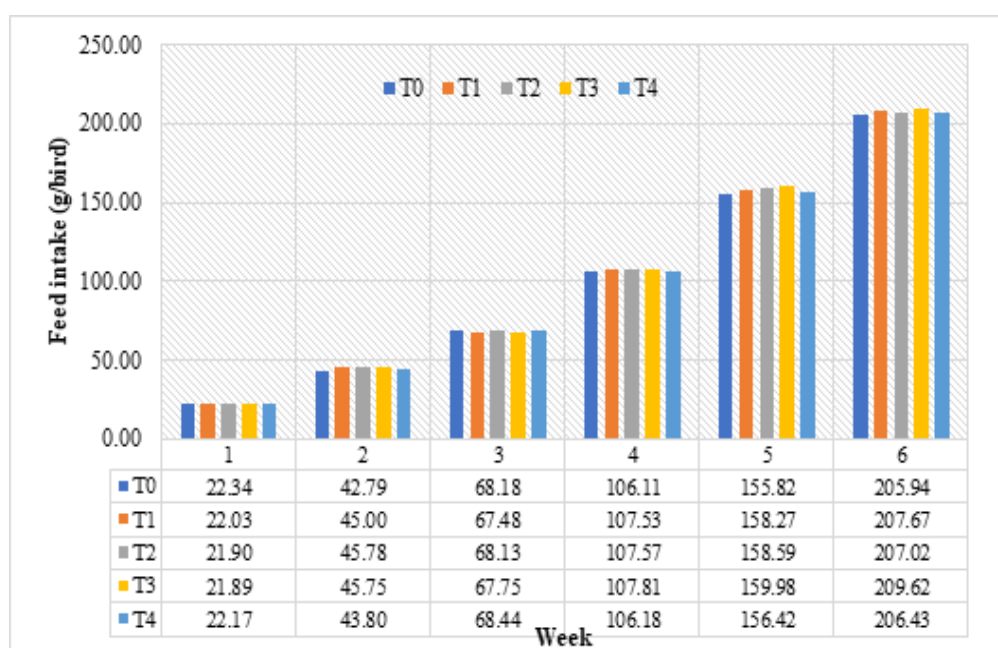


Figure 2 Development feed consumption of Native chicken at the starter phase.

Table 2 The performance of one- to six-week-old Native chickens (\pm standard deviation)

Parameter	Treatments				
	T ₀	T ₁	T ₂	T ₃	T ₄
Body weight (g/chick)	289.76 \pm 1.04 ^c	293.14 \pm 0.49 ^b	293.16 \pm 1.76 ^b	296.08 \pm 1.09 ^a	282.88 \pm 3.74 ^d
Weight gain (g/chick/6 weeks)	259.36 \pm 0.97 ^c	262.74 \pm 0.83 ^b	262.56 \pm 2.03 ^b	265.48 \pm 1.57 ^a	252.08 \pm 3.13 ^d
Feed consumption (g/chick/6 weeks)	601.19 \pm 2.77 ^d	607.98 \pm 4.29 ^{bc}	612.81 \pm 4.82 ^a	609.00 \pm 8.75 ^{ab}	603.43 \pm 4.53 ^{cd}
Feed conversion	2.32 \pm 0.10 ^{bc}	2.31 \pm 0.10 ^{bc}	2.33 \pm 0.10 ^b	2.29 \pm 0.40 ^c	2.40 \pm 0.20 ^a
Carcass weight (g/chick)	168.23 \pm 2.43 ^c	172.65 \pm 1.51 ^b	173.11 \pm 1.47 ^b	177.57 \pm 2.04 ^a	164.13 \pm 2.57 ^d
Carcass percentage (%/chick)	58.06 \pm 0.66 ^c	58.90 \pm 0.47 ^b	59.05 \pm 0.67 ^b	59.97 \pm 0.55 ^a	58.02 \pm 0.23 ^c

Note: a, b, c, and d superscripts show that there is a significant difference in responses ($P < 0.05$). T₀ (0.46% l-arginine + 0.42% l-lysine + 19% CP); T₁ (supplementation of 0.56% l-arginine + 0.51% l-lysine + 18% CP); T₂ (supplementation 0.66% l-arginine + 0.60% l-lysine + 17% CP); T₃ (supplementation of 0.76% l-arginine + 0.69% l-lysine + 16% CP); and T₄ (supplementation of 0.86% l-arginine + 0.78% l-lysine + 15% CP).

Feed conversion ratio

The highest and lowest average feed conversions ratio of Native chicken were at T₄ and T₃, respectively (Table 2). An analysis of variance showed that the treatment had a significant effect on the feed conversion ratio of Native chicken ($P < 0.01$). Duncan's test demonstrated that the response at T₀, T₁, and T₂ was not significant. However, if the supplementation occurred with T₃, the feed conversion ratio could decrease by 1.72% compared to T₂. At the highest L-arginine and L-lysine levels and the lowest CP (T₄), the feed conversion ratio increased by 4.80%.

Carcass weight

The highest and lowest average carcass weights and percentages of six-week-old Native chickens were at T₃ and T₄, respectively. An analysis of variance indicated that the treatment had a significant effect on carcass weight and its percentage of Native chickens ($P < 0.01$). Supplementation with T₁ increased the carcass weight and its percentage by 2.63% and 1.45%, respectively, compared to T₀. The same response occurred in carcass weight and its percentage at T₂. The highest responses were at T₃, namely, 2.58% and 1.56% compared to T₂. At the lowest CP level (15%), although L-arginine and L-lysine increased by 0.86 and 0.78%, the carcass weight and its percentage decreased by 7.57% and 3.25%, respectively, compared to T₃.

DISCUSSION

Arginine and lysine supplementation in feeds affects feed consumption and the body weight of Native chickens. Feed consumption decreased if both amino acids were too low at high CP (19%) or conversely at low CP (15%). At 17% CP, the feed consumption of Native chicken was at its maximum, while the maximum body weight and weight gain occurred at 16% CP with 0.76% L-arginine + 0.69% L-lysine. The average feed consumption of Native chicken was 612.81 ± 4.82 g/chick. Sirathonpong et al. (2019) reported in broilers that there is a correlation between the use of arginine and lysine in enhancing feed consumption and body weight. Furthermore, increased lysine in the feeds boosts the need for arginine or vice versa. There is antagonism between lysine and arginine if the balance is small (Balnave and Barke, 2019). Lisnahan and Nahak (2019) reported that the feed consumption of Native chicken supplemented with methionine, lysine, tryptophan, and threonine (without L-arginine) was 124.18 g/chick/week for the starter phase (aged 1-6 weeks). Furthermore, it was reported that the feed consumption of Native chickens in the grower phase (age 7-14 weeks) was 50.10 g/chick/day. Arginine plays a principal role in different metabolic, pathophysiological, and immunological processes in poultry due to the lack of a functional urea cycle (Birmani et al., 2019). Native chickens cannot synthesize endogenous arginine, and thus, it is considered an essential amino acid. Chickens depend exclusively on dietary sources of arginine, and hence, proper amounts should be provided in feeds (Teng et al., 2021). In this study, increasing L-arginine + L-lysine from 0.46% + 0.42% to 0.66% + 0.60% could reduce the use of feed CP by 2.00% from 19.00% to 17.00%. This is related to the cost of feed, such as fish and soybean meal, which are high-priced. Moreover, the treatment in this study can reduce cage pollution, such as ammonia, if the chickens are fed high CP feeds.

Body weight is related to feed consumption. In this case, body weight increased with increasing L-arginine + L-lysine from 0.46% + 0.42% to 0.76% + 0.69%. This response decreased CP usage by 3% from 19.00% to 16.00%. This showed that there was an effect of arginine and lysine on the body weight of Native chickens. The two amino acids contribute to the efficient use of feed CP. However, at CP levels lower than 16.00%, the high levels of L-arginine and L-lysine had a nonoptimal impact on the body weight of Native chickens. Any deficiency or excess of these two amino acids negatively affects plasma and muscle amino acid concentrations and growth performance (Balnave and Barke, 2019). If the CP feed is high, it must be balanced with arginine and lysine. Excessive CP feed and the imbalance of arginine and lysine will interfere with kidney function. This effect is indicated more by excessive lysine (low arginine-lysine ratio) than by excessive arginine (high arginine-lysine ratio) (Sirathonpong et al., 2019). It is reported that excessive dietary lysine will not affect the digestibility or absorption of arginine but primarily inhibit renal reabsorption and stimulate renal arginase activity (Balnave and Barke, 2019).

In feeds, when lysine levels are high and arginine levels are low, kidney arginase activity escalates, and consequently, arginine degrades (Balnave and Barke, 2019; He et al., 2021). As a result, less arginine is available for protein synthesis and arginine-dependent biological functions. Excessive

lysine reduces arginine-glycine amidinotransferase activity (Furukawa et al., 2021; Teng et al., 2021) and catalyses the first step of creatine synthesis and suppresses muscle creatine concentrations (Chrystal et al., 2021). Antagonism between arginine and lysine appears more in birds that are unable to synthesize arginine endogenously because of a lack of carbamoyl phosphate synthetase I and the low renal activity of argininosuccinate synthetase, argininosuccinate lyase, and ornithine transcarbamylase (Balnave and Barke, 2019). Therefore, arginine is an essential amino acid and its needs must be met from feed to support the growth of Native chickens. According to the (Oliveira et al., 2022), the optimal arginine-lysine ratio in broilers should be 1.06 during the first three weeks, and 1.00 from three to six weeks. Balnave and Barke (2019) suggest that the optimal arginine-lysine ratio should range from 0.90 to 1.18.

Feed conversion ratio is the ratio between feed consumed and weight gain. Low feed conversion indicates better feed efficiency. Feed conversion ratio in this study was correlated with body weight and feed consumption. The best feed conversion ratio, weight gain, and feed consumption were found at 0.76% L-arginine + 0.69% L-lysine with a CP level of 16%. This shows that the utilization of synthetic amino acids, such as L-arginine and L-lysine, can reduce the use of CP feed. Based on this study, Native chickens in the starter phase consume feed with a CP of 16% if L-arginine and L-lysine are sufficient. Barekattain et al. (2021) reported that for broilers, a high arginine-lysine ratio has the best impact on feed efficiency. Castro et al. (2019) suggested that more arginine is required for feed conversion ratio than for broiler live weight gain. They further stated that live weight and feed conversion ratio were optimized at 1.24% and 1.31% arginine, respectively. Barekattain et al. (2019) explained that the efficiency of arginine use in poultry decreased linearly with increasing lysine levels. This is in contrast to broilers with fast growth rates and low feed conversion ratio, as in previous studies. In Native chickens, genetic growth is slow with high feed conversion ratio. Hence, the use of CP feed in Native chickens based on this study is only 17.00% or 16.00% with the use of L-arginine and L-lysine, respectively. This CP level is sufficient to maximize feed consumption and weight gain as well as suppress feed conversion ratio.

The largest carcass composition in Native chickens is the chest and thigh muscles. The chest muscles become heavier and are mainly determined by feed consumption, especially the balance of amino acids that make up the muscles. Arginine and lysine significantly contribute to the carcass weight of Native chickens. For feed consumption, weight gain, and feed conversion ratio, the use of L-arginine and L-lysine can reduce the use of feed CP. Tolerance to the use of the lowest CP feed is 16.00% CP with L-arginine and L-lysine at 0.76% and 0.69%, respectively. Furthermore, if the CP of feed is only 15.00%, even though the L-arginine and L-lysine are elevated, it will negatively impact carcass weight and its percentage of Native chickens. Dietary arginine determines the carcass component of Native chickens. Arginine is involved in the synthesis of polyamines related to cell division, protein synthesis, tissue growth (Yu et al., 2022), and intestinal development (Monavvar et al., 2020). The tissue polyamine concentrations in poultry are highly responsive to the manipulation of the amino acid arginine. Due to the lack of a functional urea cycle, poultry relies on exogenous sources of arginine to form ornithine, which in mammals, is derived from glutamic acid (Tsugawa et al., 2019). Furukawa

et al. (2021) reported that a lack of polyamines inhibits the proliferation, migration, and apoptosis of intestinal cells.

Lisnahan et al. (2022) reported that the amino acid composition in the body of six-day-old Native chicks showed that the arginine content was 110% higher than the lysine content. This indicates that arginine in Native chickens needs to be increased. L-arginine can be converted to citrulline and nitric oxide by the enzyme nitric oxide synthase (Birmani et al., 2019; He et al. 2021). Nitric oxide has shown vasodilator properties (Oliveira et al, 2022) and elevated blood flow to the muscles, especially the chest and thighs. Balnave and Barke (2019) reported that chest muscle thickness was significantly increased by increasing arginine and lysine levels in feed.

CONCLUSION

Supplementation with 0.76% L-arginine and 0.69% L-lysine can boost the body weight gain, feed consumption, feed conversion ratio, carcass weight, and carcass percentage of Native chickens in the starter phase (one to six weeks), and it can reduce the use of feed CP by 2 to 3%, namely, from 19.00% to 16.00–17.00% CP.

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CONFLICT OF INTEREST

The authors declare no conflict of interest.

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