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Author	Su Hyun An[1] and Changsu Kong[1,2,3]
Affiliation	[1]Research Institute for Innovative Animal Science, Kyungpook National University, Sangju 37224, Republic of Korea [2]Department of Animal Science, Kyungpook National University, Sangju 37224, Republic of Korea [3]Department of Animal Science and Biotechnology, Kyungpook National University, Sangju 37224, Republic of Korea
ORCID (for more information, please visit https://orcid.org)	Su Hyun An (https://orcid.org/0000-0001-6236-6815) Changsu Kong (https://orcid.org/0000-0002-3876-6488)
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CORRESPONDING AUTHOR CONTACT INFORMATION

For the corresponding author (responsible for correspondence, proofreading, and reprints)	Fill in information in each box below
First name, middle initial, last name	Changsu Kong

Email address – this is where your proofs will be sent	changsukong@gmail.com
Secondary Email address	changsukong@knu.ac.kr
Address	Department of Animal Science, Kyungpook National University, Sangju 37224, Republic of Korea
Cell phone number	+82-10-3805-4776
Office phone number	+82-54-530-1225
Fax number	+82-54-530-1229

Abstract

An experiment involving 720 one-day-old male broilers (Ross 308) was conducted to investigate the effects of graded levels of crude protein and standardized ileal digestible (SID) amino acids (AA) on growth performance during the pre-starter period (0 to 7 d), and to compare the subsequent growth performance of birds fed a commercial diet in the later phase (8 to 28 d). On d 1, all birds were individually weighed and allocated to six groups with eight replicate pens (15 birds/pen). Broilers were fed diets containing six different dietary SID AA levels relative to the 90 to 115% requirement for 7 d. From d 8 to 28, birds were fed a commercial diet containing nutrient levels meeting their dietary requirements. The body weight gain (BWG; $p = 0.044$) and gain-to-feed ratio (G:F; $p = 0.005$) of birds increased quadratically, and feed intake of birds linearly increased with increasing dietary AA concentration during d 0 to 7. Following the transition to a commercial diet, body weight at 14, 21, and 28 d, BWG, feed intake, and G:F linearly increased ($p < 0.05$). From 22 to 28 days of age, BWG ($p = 0.001$) and feed intake ($p = 0.008$) of birds linearly increased compared to the 90% SID AA treatment, whereas G:F was not affected ($p = 0.088$) by dietary treatment. Overall, BWG and the growth rate of broilers aged 8 to 28 d also exhibited linear increments ($p < 0.01$) by the dietary AA concentrations in diets during the first week. The study findings confirm the influence of dietary AA concentrations on the growth performance of broilers in the first week after hatch, demonstrating that this impact persists in the later growth stage. Therefore, ensuring sufficient dietary AA intake during the first week of life can enhance performance in later stages of development in broiler chickens.

Keywords

Amino acid content, Standardized ileal digestible, Ideal amino acid ratio, Growth performance, Broiler

Introduction

Amino acids (AA) are essential nutrients involved in body protein accretion and the regulation of various physiological functions. It is well acknowledged that the growth performance of birds improves with increasing dietary digestible protein and AA concentrations [1-3]. Sufficient supply of dietary protein has been shown to increase duodenum weight, which facilitates body weight gain (BWG) in young broiler chickens [4]. Additionally, the growth potential of modern broilers has significantly improved through genetic selection by breeding companies. Consequently, optimal AA density might need continual adjustments to accommodate these advancements. Providing balanced AA supplementation in broiler diets is crucial to ensure sustainable modern broiler production and achieve associated economic and environmental benefits.

Recently, there has been a growing demand for lowering dietary crude protein (CP) concentrations by substituting soybean meal with alternative feed ingredients within the chicken meat industry. Consequently, there has been a corresponding rise in alternative protein sources, such as synthetic and crystalline amino acids, enabling the reduction of dietary CP. The mode of action for AA metabolism in the overall digestive process may vary depending on the type of AA supplements, whether protein-bound or non-bound proteins. This difference could also affect energy retention and nutrient availability. According to Beski et al. [5], supplementing poultry diets with synthetic AA improves feed conversion efficiency and reduces nitrogen excretion. The growth responses of birds may be influenced not only by dietary AA concentrations but also by the type of AA supplements. Several prior studies have investigated the optimal concentrations of dietary AA due to their significant role in enhancing broiler productivity [6-8]. These studies provide compelling evidence that higher levels of indispensable AA or protein intake facilitate productivity [8-12]. However, there is a paucity of information regarding the impact of dietary AA concentration on the growth rate of birds during the first week following hatching. Feeding regimens with varying protein levels or ingredient compositions alter broiler performance at each growth phase [13]; however, some studies have noted compensatory growth occurring as birds age [14]. In certain instances, the growth performance of birds can be affected by carry-over effects from prior dietary nutrient conditions, thus influencing subsequent growth stages.

Furthermore, it is crucial to examine compensatory growth and carry-over effects during subsequent phases to fully comprehend the significance of dietary AA composition in the pre-starter phase.

Dietary nutrient levels in the early-stage broiler diet may influence growth in later growth phases. Particularly, growth responses in the early stages, driven by rapid protein synthesis and degradation, may be influenced by digestible dietary nutrient concentrations. Therefore, it is essential to include adequate amounts of dietary AA in the pre-starter diet to ensure the optimal growth and development of broiler chickens. Furthermore, the growth responses of modern broilers by two weeks of age account for over 20% of the entire production period, with the highest growth responses observed during the first week [15]. Birds provided with adequate quantities of dietary AAs for optimal growth during the early stages are likely to outperform those lacking sufficient AAs in their diets throughout all growth phases, consequently enhancing poultry production profitability while reducing feed costs.

The potential performance of the flock primarily depends on the inherent genotype; however, dietary nutrient content, environmental conditions [12], and the homogeneity of feed mixing [16] could also influence the harvest weight and flock uniformity. The influence of dietary AA concentrations on the growth responses of birds can be confirmed through the uniformity of flocks [10,17]. Uniformity in flocks is linked to flock performance and economic returns, as well as environmental waste. Moreover, maintaining a certain level of uniformity is crucial to maximize the productivity of meat-type broilers within flock populations.

This study aimed to determine the optimal nutritional concentration of AAs required for successful growth in male broiler chickens. Additionally, it aimed to assess the impact of various dietary levels of AAs on the growth rate of birds between 0 and 7 days old and their growth at subsequent stages.

Materials and Methods

The protocols for the present study were reviewed and approved by the Institutional Animal Care and Use Committee at Kyungpook National University (KNU 2017-0140).

Experimental diets

The experimental diets, based on corn-soybean meal (SBM), were formulated with standardized ileal digestible (SID) AA. These diets encompassed six levels of dietary SID AA, ranging from 90% to 115% of the expected requirements for each respective AA. Dietary Lys, Thr [18] and Met [19] recommendation levels (100% AA) were adjusted based on the results of our previous studies. To avoid any interactions between indispensable AA, levels of all dietary AA were adjusted to maintain a constant ratio to Lys. The ideal ratios for most AA (Arg, Ile, Leu, Cys, and Val) during the starter phase were obtained from the study of Hoehler et al. [20]. Ideal ratios for His, Phe, and Trp were obtained from Wu's [21] study. All experimental diets from d 0 to 7 were formulated to be isonitrogenous (22.5% of CP) by reducing the inclusion of glutamic acid, thereby enhancing the utilization of synthetic AA. Ten synthetic or crystalline AA and glutamic acid were supplemented to consider both the dietary indispensable and dispensable AA. We analyzed the nutrient compositions in feed ingredients, corn, SBM, as well as the commercial feeds for starter (d 8 to 21) and grower (d 21 to 28) phases used in the present study, as shown in Table 2. The amounts of minerals and vitamins in all experimental diets met the requirements for broilers reported by the NRC [22].

Animal and management

A total of 720-day-old male broilers (Ross 308) were used for the experiment. At the beginning of the experiment, all birds [initial body weight (BW): 46.5 g (SD = 2.91)] were individually tagged, weighed, and allocated to six experimental diets in a randomized complete block design, with initial BW as a blocking factor using a spreadsheet [23] to minimize the weight differences between each treatment. All birds were fed *ad libitum* with the experimental diets for 7 d. The experimental environment was controlled with continuous lighting, and the temperature was gradually reduced from 33 °C to 25 °C by d 28.

Growth performance measurements and chemical analysis

The BW of all birds and the remaining feed quantity were recorded upon discovering a deceased bird within a specific pen. This procedure was implemented to refine the growth performance data. To correct the data, we used the BW and estimated individual feed intake of the deceased birds, following the modifications suggested by Sung and Adeola [24]. The calculation for determining the metabolizable energy required for maintenance (expressed in kcal/d) was conducted following the equation proposed by Noblet et al. [25]: 136 kcal × $BW^{0.70}$.

On d 21 and 28, the body weight, feed supply, and leftovers per cage were recorded for each individual bird. This information was used to determine the BWG and feed intake. Subsequently, the gain-to-feed ratio (G:F) was calculated utilizing the data on BWG and feed intake. Furthermore, analyses were conducted to determine the dry matter, crude protein, and AA compositions in the experimental diets. The samples were ground using a Cyclotec Mill (CT293 cyclotec, Foss, Denmark) through a 1.0-mm screen for chemical analysis. The feed ingredients and samples of the commercial and experimental diets were dried at 135 °C for 2 h [AOAC [26]; method 930.15]. Additionally, dried corn, SBM, and samples of the commercial and experimental diets samples were analyzed for their AA composition [AOAC [26]; method 982.30 E (a, b, c)].

Statistical analysis

All data for each ingredient were analyzed using the GLM procedure in SAS software (SAS Inst. Inc., Cary, NC, USA). Data for the CP and AA concentrations were examined using one-way ANOVA and considered as the fixed effect. The interquartile range (IQR) was used to identify and eliminate outliers; data points with values exceeding 1.5 times the IQR were deemed outliers. The average flock uniformity of treatments was determined by calculating the coefficient of variation (CV) of BW. The dietary SID Met concentrations in the pre-starter diet were considered a fixed variable, whereas the block (replicate) was considered a random variable. Means for each treatment were computed using the least square means. Orthogonoal polynomial contrast coefficients were used to test the linear and quadratic effects of increasing AA in diets. The experimental unit was a pen, and statistical significance was set at $p < 0.05$.

Results

Throughout the experimental period, all birds remained healthy. Mortality rates were recorded daily for each cage, totaling 2.5% over the entire growth period. Analysis of AA concentrations in experimental diets revealed a similar calculated pattern (Table 3).

The isonitrogenous and consistent ideal protein ratio diets across the dietary treatments were used in this study. The proportions of protein-bound AA, in this case, corn and SBM, were fixed, and quantities of non-bound AA were increased to meet the target dietary AA concentrations. Therefore, the total indispensable AA increased from 8.15 to 9.87%, and dispensable AA decreased from 16.11 to 12.87%, as expected (Table 3). In this context, linear improvements ($p < 0.01$) in BWG and G:F in birds fed diets ranging from 90% AA to 115% AA at first 7 days (Table 4) could be attributed to the increased absolute AA intake and the amount of indispensable AA derived from non-bound AA.

The results regarding the effects of dietary CP and AA concentrations on growth are presented in Table 4. During d 0 to 7, an increase in dietary AA concentration showed both linear and quadratic effects on the BW at d 7 ($p = 0.043$), BWG ($p = 0.044$), and G:F ($p = 0.005$). Furthermore, the feed intake of birds exhibited a linear increase ($p < 0.01$) with increasing dietary AA concentration from 90% AA to 115% AA. At d 7, the BW ranged from 133 (90% AA) to 154 g (115% AA), lower than the Ross 308 broiler standard (Aviagen [42]; 189 g) across all treatments. In addition, the estimated CV in the present study did not align with changes in dietary AA density during the overall growth periods, although CV values remained below 10% (Tables 4 and 5). The growth responses showed linear increment ($p < 0.01$) at above 100% AA, however, the range of dietary AA (from 90% AA to 115% AA) might be sufficient to confirm the influence of dietary AA density on growth performance in the early stages of bird development.

The linearly improved growth responses were observed ($p < 0.05$) in birds fed diets increasing dietary AA concentration from 90% to 115% AA during the pre-starter phase across all feeding periods, exept for BWG during d 8 to 14 and G:F during d 22 to 28 (Tables 5 and 6).

Discussion

Indispensable and dispensable AA

Dietary total indispensable AA (synthetic and crystalline AA) to the dispensable AA (glutamic acid) ratio increased from 0.22 (90% AA) to 0.84 (115% AA). This may help reduce the impact of different dietary protein levels and diminish the potential limitations in protein accretion caused by varying ideal protein ratios. The differences in digestion and absorption between protein-bound and non-bound AA are well documented by the previous literatures [27,28], however, in this study, the protein accretion might be explained by increased AA concentrations derived from non-bound AA. The average standardized ileal AA digestibility of protein-bound AA, specifically corn and SBM used in this study, was determined to be 87.1% and 88.7%, respectively, based on data from three previous studies [29-31]. In contrast, non-bound AA were found to exhibit nearly 100% digestibility [32,33]. Consequently, it is commonly assumed in the practical formulation of broiler diets that the digestibility of non-bound amino acids is 100%. Furthermore, Selle et al. [34] demonstrated that variations in AA digestibility can result in imbalances at protein synthesis sites. In addition, previous research has investigated determining the ideal protein ratios for young broilers within the first three weeks of life [21,22,35]. Compared to these studies, the protein ratio employed in this study proves to be adequate in meeting the indispensable AA needs of birds in their first week of life. Nonetheless, it is important to note that the ideal protein concept, which solely considers indispensable AA, is not entirely effective in supporting the synthesis of AA within animal cells de novo [36]. In this regard, there has been a growing emphasis on providing appropriate ratios and sufficient amounts of both indispensable and dispensable AA in order to achieve efficient protein accumulation within the animal body [36]. An excess of dietary glutamic acid can serve as a nutritional resource by providing a precursor for the synthesis of amino acids in cases where dietary amino acids are lacking. Eariler studies have suggested the use of large amounts of glutamic acid (10 to 13 times the level of Lys) to supply indispensable AA [37,38]. However, this study provided dietary glutamic acid at relatively lower levels, ranging from 4 (115% AA) to 7 (90% AA) times the level of Lys. This contrasts with the higher levels used in previous studies. Unfortunately, in this study, the ideal protein ratio only considered indispensable AA, with L-glutamic acid being supplemented to meet the demands of dispensable AA and adjust to maintain the dietary protein level at 22.5% across the experimental diets. The addition of L-glutamic acid from 8.50% (90% AA) to 4.87% (115% AA) may be excessive for young broiler chicks, considering the recommended optimal amino acid ratios from Texas A&M University [21]. The reduced growth performance observed in birds fed diets with less than 100% AA may be attributed to the low ratio of indispensable to dispensable AA in the diet. Difference of approximately 20 grams in BW of birds at 7 days was observed between the treatment groups receiving 115% AA and 90% AA. This discrepancy could be resolved within a single day at this age. Furthermore, at this age, the voluntary feed intake of birds is neglible because of thier underdeveloped gastrointestinal tracts. As a result, the authors consider it challenging to conclusively determine the impcat of elevated dietary L-glutamic acid on the findings of this study. Body weight was observed to increase as dietary glutamic acid concentration decreased, although this outcome was predominantly might attributed by the dietary indispensable AA concentration rather than the glutamic acid concentration. It is important to note that dispensable AA cannot be converted into indispensable AA, while indispensable AA can be exchanged with dispensable AA during catabolism.

Dietary AA concentration

It is now widely acknowledged that the dietary concentrations of AA in broiler diets may act as a limiting factor in early-stage growth [39]. Previous studies have demonstrated varying growth rates of broilers in response to absolute dietary AA concentrations [6,7,40,41]. The diet (100% AA) utilized in this study had a Lys concentration of 1.36%, which exceed the dietary SID Lys concentration (%) reported by Rostagno et al. [43] for 7-day-old with low-standard performance male broilers weighing 194 g (1.34%). However, the recorded BW of the 7-day-old broilers in this study was similar to that at five days old, as noted by Rostagno et al. [43]. The estimated SID Lys required for male broilers at five days old was 1.35%, similar to the dietary Lys content in the 100% AA diet. Additionally, Dozier III et al. [7] reported that the dietary Lys for Ross high-yielding broilers at 7 d of age is 1.36%, estimated through regression equations. However, Met, the first-limiting AA in a corn-SBM-based diet, had the lowest percentage (0.47%) compared to the values of 0.55% recommended by Rostagno et al. [43] and

0.62% recommended by Dozier III et al. [7]. Based on the values from these literatures, the 100% AA diet (0.47% Met) employed in this study may be insufficient to achieve maximum growth responses.

Carry-over effect

Following hatching, the development process of the birds' digestive organs commences as they initially adapt to solid feed and use it as a nutrient source rather than nutrients stored in the yolk. Particularly during this phase, the voluntary feed intake of young chicks is negligible compared to that of older, growing birds, making them susceptible to influence related to energy or AA intake [15]. However, as indicated by the present study, differences in BWG and G:F persisted despite improved voluntary feed intake in later stages of growth. From d 8 to 28, during which the birds were fed commercial diets following a seven-day experimental feeding period, the preceding dietary AA concentration affected both BW and feed efficiency. We hypothesized that growth responses, negatively impacted by lower AA density in the first week of age, would be rectified during the re-alimentation period with a commercial diet. Although the commercial starter diet utilized in this study contained elevated levels of indispensable (Met, Phe, Thr, and Trp) and all dispensable AA (except for Cys and Glu) when compared to the experimental diets containing 105% AA, our findings demonstrate that comparable growth patterns were observed during the entire grow-out period. Additionally, at d 28, the BW of groups fed 100% AA or dietary AA amounts exceeding their nutrient requirements ranged from 1,523 g to 1,618 g (Ross 308 male broiler standard is 1,576 g [42]).

Several studies have investigated the impact of different feeding strategies on the growth rate and carcass traits of birds in subsequent growth phases following the provision of diets with varying levels of dietary AA density [6,17,44-47]. Corzo et al. [17] found that birds exhibited increased growth rates when fed a diet with higher AA density, noting that concentrating high AA density in the early growth stage yielded greater benefits for subsequent growth. These findings align with the results of the present study. In contrast to our findings, some studies have observed compensatory growth in birds following re-alimentation [15, 44]. Certain research reports suggest that extending the feeding period of diets with lower dietary AA levels in later growth stages might mitigate the adverse effects of lower AA density in preceding phases. Eits et al. [44] also noted no evidence of a nutritional carry-over effect from previous feeding on the proportion of carcass weight in overall gain. Noy and Skaln [15] evaluated the long-term effects of feeding different dietary fat and CP concentrations in the early post-hatch period. They followed a feeding regimen similar to that in the present study, wherein birds were fed for 7 days post-hatching and then provided with the same commercial diet until they reached market weight (d 41). They found that differences in growth attributed to feeding varying dietary protein levels during the first 7 days had almost disappeared by day 18, with no significant differences observed thereafter. In the present study, during periods of feeding with the commercial diet, birds consumed similar amounts of feed, except for a week prior to the conclusion of the experiment. This implies that providing chicks with optimal dietary AA during the early growth stage may be crucial for ensuring performance on slaughter day. However, if birds were nourished for prolonged durations (as they approach slaughter weight), it is plausible that they may have exhibited compensatory growth. This phenomenon can be attributed to their augmented voluntary feed intake, which arises from expanding their digestive tract capacity as they mature. Because theoretical maximum feed intake would be set by the capacity of the digestive stystem. Unfortunately, the duration of this study was limited to 28 days, which prevented us from confirming the long-term impact of dietary AA density on growth during the later stages of feeding. Therefore, additional research is required to establish the extent of the compensatory growth responses in subsequent stages, considering growth potential of modern broilers. Such insights would be valuable additions to the feeding regimen programs for modern broilers to improve the growth responses as well as meat quality.

Flock body weight uniformity

Evidence suggests that uniformity improves with the provision of high levels of a limiting AA or protein [9,10]. Corzo et al. [17] noted an enhancement in flock uniformity in terms of BW as dietary AA concentration increased. Lemme [9] similarly observed a reduction in the coefficient of variation (CV) of BW and breast weight of broilers with an increase in dietary DL-Met concentration. Various factors, including animal, nutritional, and environmental conditions, influence BW uniformity. Among these factors, a deficiency of dietary AAs, such as limiting AAs, results in poor growth uniformity [10]. Vasdal et al. [48] proposed that the expected level of flock weight uniformity in broilers should be below

10% CV. Additionally, Berhe and Gous [10] and Gous [12] reported a marked increase in bird growth variation when poor-quality feed is provided. Berhe and Gous [10] found that birds fed a high-protein diet exhibited lower variability in BW at 21 d of age compared to those fed a low-protein diet (11.3% in low protein vs. 9.8% in high protein). However, the estimated CV in the present study did not align with changes in dietary AA density during the overall growth periods, although CV values remained below 10%. Furthermore, we observed no differences in variability between birds fed experimental diets and those fed commercial diets. This difference could be attributed to the dietary ingredients and nutrient compositions of the experimental diets. In this study, experimental diets were administered for a 7-day feeding period, with a fixed dietary protein level of 22.5%, and AA density adjusted by supplementing nine synthetic indispensable AA, as well as L-Cys and glutamic acid as dispensable AA. Conversely, previous studies predominantly utilized two or three synthetic AA, primarily Met, Cys, Lys, and Thr, or feed-grade commercial AA. Notably, improvements in growth responses have been primarily observed when birds are fed diets substituting plant protein sources with synthetic AA. The reasons for the improved growth responses in the subsequent growth periods remain unclear, but the composition of dietary protein sources (protein-bound or non-bound AA) may contribute to the observed variations in the studies.

In conclusion, birds fed diets with increasing AA density (over 100% AA) exhibited linearly greater body weight and improved feed efficiency during the early stages of life (d 0 to 7). This study demonstrates that feeding chicks diets with varying AA densities during the first week after hatching may persist in similar growth patterns in older birds. Increasing the dietary AA density from 90% to 115% of the recommended AA intake from d 0 to 7 linearly affected BW and feed efficiency until 28 days of age.

Competing Interests

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Author's Contributions

Conceptualization: An SH, Kong C.

Data curation: An SH, Kong C.

Formal analysis: An SH.

Methodology: An SH, Kong C.

Software: An SH, Kong C.

Validation: An SH.

Investigation: Kong C.

Writing - original draft: An SH, Kong C.

Writing - review & editing: An SH, Kong C.

Ethics approval and consent to participate

The protocols for the present study were reviewed and approved by the Institutional Animal Care and Use Committee at Kyungpook National University (KNU 2017-0140).

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Tables

Table 1. Ingredient composition of the experimental diets for broilers aged 0 to 7 d, on an as-fed basis

Item	Standardized ileal digestible amino acids concentrations relative to lysine, %					
	90	95	100	105	110	115
Ingredient, %						
Corn (8.8% crude protein)	37.05	37.05	37.05	37.05	37.05	37.05
Soybean meal (43% crude protein)	26.70	26.70	26.70	26.70	26.70	26.70
Corn-starch	19.27	19.49	19.74	20.01	20.27	20.61
Glutamic acid	8.50	7.82	7.11	6.39	5.67	4.87
Soybean oil	2.00	2.00	2.00	2.00	2.00	2.00
L-lysine-HCl (78.8%)	0.46	0.54	0.62	0.70	0.77	0.85
DL-methionine	0.21	0.23	0.26	0.28	0.31	0.33
L-threonine	0.19	0.23	0.26	0.30	0.34	0.38
L-arginine	0.20	0.26	0.32	0.39	0.45	0.51
L-cysteine	0.27	0.30	0.32	0.35	0.37	0.40
L-histidine	0.03	0.05	0.07	0.09	0.11	0.14
L-isoleucine	0.18	0.23	0.27	0.31	0.35	0.40
L-leucine	0.06	0.12	0.19	0.26	0.32	0.39
L-phenylalanine	0.00	0.04	0.08	0.11	0.15	0.19
L-valine	0.26	0.31	0.36	0.41	0.46	0.51
Limestone	1.30	1.30	1.30	1.30	1.30	1.30
Monocalcium phosphate	1.93	1.93	1.93	1.93	1.93	1.93
Salt	0.30	0.30	0.30	0.30	0.30	0.30
Sodium bicarbonate	0.40	0.40	0.40	0.40	0.40	0.40
Vitamin premix[1]	0.20	0.20	0.20	0.20	0.20	0.20
Mineral premix[2]	0.20	0.20	0.20	0.20	0.20	0.20
Choline chloride	0.11	0.11	0.11	0.11	0.11	0.11
Total	100.0	100.0	100.0	100.0	100.0	100.0
Calculated composition, %						
MEn[3], kcal/kg	3,158	3,167	3,176	3,185	3,194	3,204
Crude protein	22.5	22.5	22.5	22.5	22.5	22.5
Calcium	1.0	1.0	1.0	1.0	1.0	1.0
Non-phytate Phosphorus	0.45	0.45	0.45	0.45	0.45	0.45
SID amino acids, %						
Arginine	1.14	1.20	1.26	1.33	1.39	1.45
Histidine	0.39	0.41	0.43	0.46	0.48	0.50
Isoleucine	0.76	0.80	0.84	0.89	0.93	0.97
Leucine	1.19	1.26	1.33	1.39	1.46	1.53
Lysine	1.12	1.18	1.24	1.30	1.36	1.43
Methionine	0.42	0.45	0.47	0.49	0.52	0.54
Cysteine	0.45	0.47	0.50	0.52	0.55	0.57
Phenylalanine	0.67	0.71	0.74	0.78	0.82	0.86
Threonine	0.67	0.70	0.74	0.78	0.81	0.85
Tryptophan	0.17	0.18	0.19	0.20	0.21	0.22
Valine	0.88	0.93	0.98	1.03	1.08	1.13

[1]Supplies the following per kilogram of diet: vitamin A, 24,000 IU; vitamin D_3, 8,000 IU; vitamin E, 160 mg/kg; vitamin K_3, 8 mg/kg; vitamin B_1, 8 mg/kg; vitamin B_2, 20 mg/kg; vitamin B_6, 12 mg/kg; pantothenic acid, 40 mg/kg; folic acid, 4 mg/kg; niacin, 12 mg/kg.

[2]Supplies the following per kilogram of diet: Fe, 120 mg/kg; Cu, 320 mg/kg; Zn, 200 mg/kg; Mn, 240 mg/kg; Co, 2 mg/kg; Se, 0.6 mg/kg; I, 2.5 mg/ kg.

[3]MEn=Nitrogen-corrected metabolizable energy.

Table 2. Analyzed nutrient concentrations (%) in feed ingredients and commercial diets used in the present study

Item	Feed ingredients		Commercial diets	
	Corn	Soybean meal	Starter	Grower
Dry matter	87.0	87.6	89.1	88.8
Crude protein	8.8	42.7	20.7	19.3
Crude fat	4.8	1.9	6.8	6.1
Crude fiber	2.0	3.9	3.1	3.0
Ash	1.6	6.0	5.5	4.7
Indispensable amino acid				
Arginine	0.43	3.21	1.22	1.11
Histidine	0.25	1.08	0.45	0.43
Isoleucine	0.29	1.94	0.74	0.68
Leucine	0.99	3.45	1.62	1.45
Lysine	0.30	2.68	1.40	1.19
Methionine	0.19	0.61	0.65	0.56
Phenylalanine	0.43	2.26	0.91	0.85
Threonine	0.35	1.76	0.98	0.90
Tryptophan	0.04	0.47	0.20	0.19
Valine	0.44	2.09	0.97	0.95
Dispensable amino acid				
Alanine	0.60	1.98	1.06	0.91
Aspartic acid	0.61	4.91	1.62	1.54
Cysteine	0.19	0.73	0.39	0.40
Glutamic acid	1.65	7.70	3.40	3.37
Glycine	0.35	1.91	1.12	0.92
Proline	0.82	2.28	1.45	1.57
Serine	0.45	2.29	1.05	1.03
Tyrosine	0.25	1.47	0.54	0.43

Table 3. Analyzed nutrient concentrations (%) in the experimental diets

Item	Standardized ileal digestible amino acids concentrations relative to lysine, %					
	90	95	100	105	110	115
Dry matter	89.4	89.3	89.5	89.5	88.9	88.9
Crude protein	22.2	22.1	22.2	22.4	21.6	21.4
Crude fat	4.0	4.2	4.4	4.3	4.2	4.2
Crude fiber	1.9	1.9	1.8	1.7	1.9	1.8
Ash	6.0	6.0	6.1	6.1	6.0	6.0
Indispensable amino acid						
Arginine	1.20	1.29	1.35	1.41	1.43	1.45
Histidine	0.41	0.44	0.47	0.48	0.49	0.51
Isoleucine	0.83	0.89	0.95	0.92	1.13	1.03
Leucine	1.34	1.48	1.49	1.62	1.57	1.66
Lysine	1.23	1.29	1.36	1.41	1.39	1.45
Methionine	0.44	0.48	0.48	0.50	0.50	0.53
Phenylalanine	0.77	0.84	0.86	0.89	0.92	0.92
Threonine	0.78	0.82	0.86	0.88	0.93	0.95
Tryptophan	0.14	0.15	0.15	0.16	0.16	0.18
Valine	1.01	1.05	1.10	1.15	1.17	1.19
Dispensable amino acid						
Alanine	0.76	0.79	0.76	0.76	0.74	0.72
Aspartic acid	1.59	1.64	1.59	1.58	1.59	1.51
Cysteine	0.52	0.55	0.55	0.56	0.55	0.59
Glutamic acid	10.35	9.63	9.57	8.69	7.95	7.27
Glycine	0.63	0.66	0.65	0.65	0.64	0.62
Proline	1.00	1.01	0.98	1.00	0.97	0.96
Serine	0.81	0.84	0.81	0.81	0.81	0.78
Tyrosine	0.45	0.47	0.46	0.44	0.44	0.42

Table 4. Growth performance of broilers fed with varying concentrations of dietary amino acids from 0 to 7 d of age[1]

	Standardized ileal digestible amino acids concentrations relative to lysine, %						RMSE[1]	*P*-values	
Item	90	95	100	105	110	115		Linear	Quadratic
Number of observations	(8)	(6)	(7)	(8)	(8)	(8)			
Body weight, g									
Initial BW, g	46.5	45.5	46.5	46.5	46.5	46.5	0.03	0.251	0.591
BW at d 7, g	133	137	144	148	154	154	3.9	< 0.01	0.043
CV	5.3	6.1	6.9	5.4	3.2	3.8			
BWG, g/bird	86.6	90.8	97.7	101.7	107.9	107.3	3.89	< 0.01	0.044
Feed intake, g/bird	90.8	92.5	97.4	98.7	102.7	105.7	3.65	< 0.01	0.777
Gain-to-feed ratio, g/kg	0.95	0.98	1.00	1.03	1.05	1.02	0.032	< 0.01	0.005

BW = Body weight; CV = coefficient of variation; BWG = body weight gain.

[1]RMSE = Root mean square error.

Table 5. Growth performance of broilers fed commercial diets from 8 to 28 d of age[1]

	Standardized ileal digestible amino acids concentrations relative to lysine, %						RMSE[1]	*P*-values	
Item	90	95	100	105	110	115		Linear	Quadratic
Number of observations	(8)	(6)	(7)	(8)	(8)	(8)			
Body weight, g									
d 14, g	404	408	426	430	436	439	13.45	< 0.01	0.286
CV	4.8	6.9	5.2	3.0	3.1	4.2			
d 21, g	887	892	930	926	934	945	28.8	< 0.01	0.362
CV	5.4	6.2	3.6	2.4	2.5	3.0			
d 28, g	1,523	1,560	1,597	1,598	1,618	1,647	48.69	< 0.01	0.480
CV	3.0	4.8	5.1	2.4	2.7	3.0			
BWG, g/bird									
d 8 to 14	271	271	281	281	276	285	14.2	0.062	0.781
d 15 to 21	484	482	503	496	499	505	18.7	0.011	0.612
d 22 to 28	636	666	672.2	673.5	682.8	699.8	32.94	0.001	0.635
Feed intake, g/bird									
d 8 to 14	373	384	368	352	365	346	23.1	0.006	0.669
d 15 to 21	707	690	671	660	661	663	37.7	0.008	0.172
d 22 to 28	964	1,011	1,025	1,009	1,012	1,037	40.88	0.008	0.268
Gain-to-feed ratio, g/kg									
d 8 to 14	0.73	0.71	0.77	0.8	0.76	0.82	0.064	0.002	1.000
d 15 to 21	0.69	0.7	0.75	0.75	0.76	0.76	0.041	<0.01	0.087
d 22 to 28	0.66	0.66	0.66	0.67	0.67	0.67	0.024	0.088	0.594

CV = coefficient of variation; BWG = body weight gain.

[1]RMSE = Root mean square error.

[2]All the birds were provided with a starter diet for seven days, from day 8 to day 14. After that, they were given a grower diet for 14 days until they reached 28 days of age.

Table 6 Growth performance of broilers fed with varying concentrations of dietary amino acids from 8 to 28 d of age and overall experimental period[1]

	Standardized ileal digestible amino acids concentrations relative to lysine, % requirement, %						RMSE[1]	*p*-values	
Item	90	95	100	105	110	115		Linear	Quadratic
Number of observations	(8)	(6)	(7)	(8)	(8)	(8)			
BWG, g/bird									
d 8 to 28	1391	1421	1455	1451	1457	1490	47.3	< 0.01	0.538
Overall	1477	1511	1553	1553	1565	1598	48.4	< 0.01	0.445
Feed intake, g/bird									
d 8 to 28	2044	2082	2064	2021	2038	2046	62.7	0.394	0.961
Overall	2134	2175	2161	2119	2141	2152	63.9	0.769	0.976
Gain-to-feed ratio, g/kg									
d 8 to 28	0.68	0.68	0.71	0.72	0.72	0.73	0.023	< 0.01	0.534
Overall	0.69	0.69	0.72	0.73	0.73	0.74	0.022	< 0.01	0.418

BW = Body weight; CV = coefficient of variation; BWG = body weight gain.

[1]RMSE = Root mean square error.

[2]All the birds were provided with a starter diet for seven days, from day 8 to day 14. After that, they were given a grower diet for 14 days until they reached 28 days of age.