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Dietary arginine supplementation enhances the growth performance and immune status of broiler chickens

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ABSTRACT

This experiment was conducted to investigate the effects of dietary Arg on growth performance and immunity in broiler chickens. Three hundred 1-d-old broiler chickens were divided into 5 treatments with 6 cages and 10 chickens per cage. The broiler chickens in the 5 treatments were fed with the basal diet (Arg-deficient diet, 8.6% Arg in diet from 0 to 21 d and 6.8% Arg in diet from 21 to 42 d) supplemented with 0%, 0.45%, 0.90%, 1.35%, and 1.80% Arg, respectively. The effects of dietary Arg on performance and immune status were assessed. The results showed that, with increasing of dietary Arg content, body weight was increased quadratically on d 21 ($P < 0.01$) and 42 ($P < 0.01$). The daily weight gain ($P < 0.01$) and feed conversion ($P < 0.05$) were improved quadratically during the entire period. In addition, with increasing of dietary Arg content, serum concentrations of growth hormone ($P < 0.01$), insulin-like growth factors-1 ($P < 0.01$), insulin ($P < 0.01$), IgA ($P < 0.01$), and IFN-γ ($P < 0.01$), as well as thymus weight ($P < 0.05$) of broiler chickens, were improved quadratically on d 21; and lymphocyte proliferation ($P < 0.01$), the antibody titers to newcastle disease (linear, $P < 0.01$; quadratic, $P < 0.01$), and serum IgM concentration ($P < 0.05$) were increased linearly or quadratically on d 42. The results indicated that incorporating Arg in excess of the 1994 NRC requirement improves the growth and immunity of broiler chickens.

1. Introduction

Arginine (Arg) is one of the natural amino acids, classified as a semi-dispensable or conditionally indispensable amino acid for mammals, but an indispensable amino acid for poultry, because poultry cannot synthesize Arg and are completely dependent on the diet as source of Arg to meet their needs for protein synthesis and other functions (Khajali and Wideman, 2010). Past research has clearly demonstrated the importance of providing broiler chicks adequate dietary Arg to support growth performance and immune response (Burton and Waldroup, 1979; Cuca and Jensen, 1990). But in general, the content of Arg in diet was not high enough to support the maximum growth rate and immune function for animals (Masoud et al., 2014; Munir et al., 2009; Perez-Carbajal et al., 2010; Ruiz-Feria et al., 2001). The NRC (1994) estimated the Arg requirement of broiler chickens to be 1.25% from 0 to 3 wk, 1.10% from 3 to 6 wk, and 1.00% from 6 to 8 wk of age (NRC, 1994). Previous reports suggested indicated that Arg supplementation beyond the NRC (1994) recommendations improved productivity of broiler chickens (Fernandes et al., 2009; Ruiz-Feria, 2009). Kidd et al. (2001) reported that supplementing broiler chicks diets with 0.2% Arg beyond NRC (1994) requirement resulted in improved growth performance. The recognition of the importance of Arg for broiler chickens health and production has resulted in practices of supplementing broiler chickens diets with content of Arg exceeding those recommended by the NRC (1994). In addition to the requirement for growth, Arg has been shown recently in several studies to have beneficial effects on the immune status of animals (de Jonge et al., 2002; Kidd et al., 2001; Kwak et al., 1999; Ochoa et al., 2001; Tan et al., 2009). However, much excess Arg had negative effect on growth and health of animals (Edmonds et al., 1987; Southern and Baker, 1982; Tani et al., 1990).

Up to now, research about the optimal supplemental dosage of Arg in diets, which can cause the best growth performance and immunity of broiler chickens, is sparse. Growth responses of young chicks as affected by dietary Arg should be studied further. Therefore, the aim of the present study was to evaluate the effect of Arg on growth performance, endocrine and immune status of broiler chickens to test the hypothesis that feeding increasing content of Arg could quadratically improve the growth performance and immunity of broiler chickens.

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2. Materials and methods

The experiment was carried out in the Experimental Farms of the Inner Mongolia Agricultural University in China and received prior approval from the Animal Care and Use Committee of this Institution.

2.1. Broiler chickens, diets and experimental design

Three hundred 1-d-old mixed-sex Arbor Acre broiler chicks (mean initial weight was 47 g per broiler chick) were randomly allotted to 5 dietary treatments with 6 cages in each treatment, 10 broiler chicks (5 cockerels and 5 pullets) in each cage. There was no significant difference in initial body weight among treatments. The present study contained the following 5 treatments: the basal diet (Arg-deficient without supplementation), the basal diet plus 0.45% L-Arg (achieve nutrient Arg content of 1994 NRC), the basal diet plus 0.90% L-Arg, the basal diet plus 1.35% L-Arg, and the basal diet plus 1.80% L-Arg. An initial basal diet was mixed to contain a formulated Arg of 0.86% in diet of 0–21 d and 0.68% in diet of 21–42 d. Experimental diets were formulated according to the different growth stages of broiler chickens 0–21 d and 21–42 d). The main raw materials were corn and corn gluten meal. Various experimental diets were prepared by adding different doses of Arg in basal diet. The basal diet met the nutrient requirements suggested by NRC (1994) and Agricultural Industry Standards of PR China (Feed Standard of Chicken, NY/T 33-2004) except for lack of Arg, and the basal diet contained normal content of Arg (Table 1). The final diets were obtained by mixing 98% of the basal ration with a premix containing different concentrations of Arg. Diet analyses for Arg concentration was determined by HPLC methods (Zheng et al., 2010) and crude protein (N × 6.25) according to the methods of the AOAC (2000).

Table 1
Composition of experimental diets (air-dry basis).

Ingredients, %	Starter 0–21 d	Grower 21–42 d
Corn	62.00	68.46
Corn gluten meal	18.50	19.00
Soybean meal	6.50	–
Wheat bran	7.20	7.48
Limestone	1.51	1.41
Dicalcium phosphate	1.90	1.59
Methionine	0.16	–
Lysine	0.75	0.60
Threonine	0.05	0.06
Tryptophan	0.06	0.05
Sodium chloride	0.37	0.35
Premix[a]	1.00	1.00
Total	100.0	100.0
Composition[b]		
Metabolic energy (MJ/kg)	12.49	12.81
Crude protein (%)	21.51	19.18
Calcium (%)	1.04	0.91
Available phosphorus (%)	0.46	0.40
Lysine (%)	1.28	0.98
Digestible lysine (%)	1.14	0.88
Methionine (%)	0.56	0.38
Arginine (%)	0.86	0.68
Digestible arginine (%)	0.79	0.63
Threonine (%)	0.84	0.75
Methionine + Cystine (%)	0.92	0.76
Tryptophan (%)	0.22	0.20
Leucine (%)	1.26	1.11

[a] Premix provided the following per kilogram of diet: Fe (as ferrous sulfate) 95 mg; Zn (as zinc sulfate) 75 mg; Mn (as manganese sulfate) 75 mg; Cu (as copper sulfate) 10 mg; I (as calcium iodate) 0.6 mg; Se (as sodium selenite) 0.3 mg; Vitamin A 6000 IU; Vitamin D 1250 IU; Vitamin E 15 mg; Vitamin K 2.2 mg; Vitamin B_1 1.5 mg; Vitamin B_2 8.0 mg; Vitamin B_6 2.5 mg; Vitamin B_{12} 0.011 mg; Niacin 44 mg; D-pantothenic acid 11 mg; Folic acid 0.9 mg; Biotin 0.11 mg; Choline 1100 mg.

[b] Crude protein and arginine are measured values, and other nutrient content are calculated values.

All experimental broiler chickens were reared in metal cages (0.8 m × 0.8 m × 0.5 m). Feed and water were supplied ad libitum for all of the broiler chickens during the whole experimental period. The trial lasted 6 wks and was divided into the first period of the experiment 0–21 d) and the second period of treatment 21–42 d). Infrared heating device was installed in the broiler chicken house to maintain the temperature automatically. Room temperature was set at approximately 32 °C in the first wk, and then reduced by 3 °C weekly until reaching 20 °C, which was preserved until the end of the experiment. And 23-h light and 1-h dark illumination schedule was kept. Vaccinations to newcastle disease virus (Harbin Pharmaceutical Group Bio-vaccine Co., Ltd. Harbin, China) were administered at d 14 via spray.

2.2. Sampling procedures and measurements

2.2.1. Growth performance

During the experiment, body weight of broiler chickens were measured on d 0, 21 and 42, and feed consumption was recorded in the course of the whole experiment for each cage, daily weight gain and daily feed intake calculated as follows: *Daily weight gain* = (*FBW-IBW*)/($m \times n$) and *Daily feed intake* = $FC/(m \times n)$, where *FBW* is final body weight of d 21 or d 42, *IBW* is initial body weight of d 0 or d 21, *FC* is total feed consumption of each cage during the starter or grower, m is the trial days of starter and grower period and n is the number of broiler chickens in each cage. The mortality was registered. Feed conversion rates calculated subsequently and corrected for mortality. Body weights of dead and culled broiler chickens were included for corrected body weight gain in feed conversion calculations.

2.2.2. Blood sample collection and measurements

At d 21 and 42 and after 12 h feed restriction, 3 representative cockerels and 3 pullets of the body weight close to the respective treatment mean were chosen from each treatment group on the morning. Blood sample (5 mL) was collected by venipuncture of the wing vein. Serum was frozen until hormones, immunoglobulins, cytokines and antibody titers for newcastle disease virus could be performed. At the same time, the other blood sample was collected into a tube with anticoagulant for analysis of lymphocyte proliferation. Serum was separated from whole blood by centrifugation at 1500 × *g* and 4 °C for 15 min and stored at − 20 °C until analysis of serum immune indexes and other biochemical analysis.

Hormones including growth hormone (GH), insulin-like growth factors-1 (IGF-1) and insulin in serum were assayed using commercially available kits (Beijing Sino-uk Institute of Biological Technology, Beijing, China), according to the manufacturer's instructions. The serum concentrations of IgA, IgG, IgM, IL-4 and IFN-γ were measured using commercial ELISA kits (Nanjing Jiancheng Bioengineering Institute, Nanjing, China) following the manufacturer's instructions. Antibody titers to newcastle disease were determined according to the method of Hu and Liu (1997). Lymphocyte proliferation was measured according to the method of Mosmann et al. (1986). Mononuclear phagocytic index was determined according to the method of Lin (1999).

2.2.3. Lymphoid organ weight

Broiler chickens were slaughtered after blood samples were collected, lymphoid organ (thymus, spleen, and bursa of fabricius) were collected from each broiler chicken. Adherent fat was removed from these tissues, and the tissues were weighed individually. Lymphoid organ was expressed in percentage relative to the individual broiler chicken's body weight.

2.3. Statistical analysis

Data were analyzed using the General Linear Model (GLM)