



## Effect of gelatin supplementation on growth performance and blood metabolites of broiler chickens fed diets varying in crude protein



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### ABSTRACT

An experiment was conducted to investigate the effects of gelatin inclusion on growth performance, blood metabolites and carcass characteristics of broiler chickens fed diets varying in crude protein (CP) concentration in a 42-d study. Four concentrations of gelatin (0, 30, 60, and 90 g/kg) and 2 concentrations of CP in the starter (220 and 200 g/kg) and grower (195 and 182 g/kg) periods were evaluated in a 2 × 4 factorial arrangement of treatments. Each diet was fed to 5 replicate pens of 15 male broiler chickens (Ross 308) from d 0 to 42. Digestibility coefficients of amino acid (AA) of gelatin were in the range of 0.90–0.95. Gly, Pro, Ala, and Glu were the major AA in gelatin. There were interactions between dietary CP content and gelatin concentration for feed intake (FI;  $P=0.016$ ), body weight gain (BWG;  $P < 0.001$ ) and feed conversion ratio (FCR;  $P < 0.001$ ) during the starter (d 0–21), grower (d 21–42), and the overall experiment period (d 0–42). In the low CP diets, gelatin inclusion had a greater negative effect on BWG and FCR compared with those fed the normal CP diets over the course of the experiment (gelatin × CP,  $P < 0.001$ ). The contrasts showed that the increase in dietary gelatin content decreased BWG and FI, whereas FCR increased with higher dietary gelatin content from d 0 to 42. The CP × gelatin interactions showed that gelatin addition to the low CP diets increased plasma concentrations of glucose (gelatin × CP,  $P < 0.01$ ) and uric acid (gelatin × CP,  $P < 0.001$ ), and decreased N retention (gelatin × CP,  $P < 0.001$ ) to a greater extent than was the case for the normal CP diets at d 42. The CP × gelatin interactions indicated that gelatin inclusion in the low CP diets decreased carcass yield (gelatin × CP,  $P < 0.01$ ) and abdominal fat (gelatin × CP,  $P < 0.001$ ), and increased heart weight (gelatin × CP,  $P < 0.01$ ) to a greater extent than was the case for the normal CP diets. The present findings showed that gelatin inclusion had a greater negative effect on growth performance and carcass yield in broiler chickens that received the low CP diets compared with those fed the normal CP diet over the course of the study. Also, increasing dietary gelatin content depressed growth rate and N retention.

### 1. Introduction

Meeting the amino acid (AA) requirements of broiler chickens constitutes a large portion of the cost associated with poultry production. Thus, reduction of dietary crude protein (CP) concentrations can lead to considerable cost savings depending on the cost of the various ingredients in the diet (Berres et al., 2010; Khalaji et al., 2016). The ability to decrease dietary CP can also result in lower N excretion (Donsbough et al., 2010; Jiang et al., 2005) and allows for the use of a wider range of feedstuffs (Aftab et al., 2006) if low CP diets are fortified with crystalline AA in a pattern that matches maintenance and tissue accretion requirements. Many different strategies have been assessed to improve growth performance of broiler chickens fed on the low CP diets. These strategies include addition of triammonium citrate or L-Glu to provide nonspecific N (Bregendahl et al., 2002), addition of

indispensable AA (IDAA) or dispensable AA (DAA) to meet or exceed NRC (1994) recommendations (Dean et al., 2006) and also the use of exogenous proteases to enhance the digestibility of AA (Cowieson and Roos, 2014). Dispensable and indispensable AA supplementation to reduced CP diets has been demonstrated to be relatively successful (Ospina-Rojas et al., 2012; Waguespack et al., 2009). The inclusion of L-Glu has been reported to improve growth performance of broiler chickens consuming low protein diets (Berres et al., 2010). In addition, because of its important role, especially in reduced CP diets, Gly may be considered to be a conditionally IDAA in poultry nutrition (Corzo et al., 2009; Powell et al., 2009). Waguespack et al. (2009) demonstrated that the inclusion of Gly to a normal CP diet (220 g/kg of diet) resulted in improved feed conversion ratio (FCR). The addition of Gly to a reduced CP diet (160 g/kg) has been reported to support similar growth performance in broiler chickens fed a diet containing 220 g CP/kg

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(Dean et al., 2006).

Gelatin is a purified animal protein from skin, bones, and connective tissues produced by selective hydrolysis (Boomgaard and Baker, 1972). Gelatin proteins are the major components of connective tissue, skin, and cartilage in human and animal body (Khalaji et al., 2016). Gelatin extracted from cow skins is potentially a valuable protein source in poultry diets with digestibility coefficient (DC) of 0.90 (Khalaji et al., 2016; Reutersward, 1984). Gelatin contains 750–850 g CP/kg with a good amino acid balance, and Gly, Ala, Pro, and Glu account for two-third of its polypeptide chain (Boomgaard and Baker, 1972). Fasina et al. (2007) reported that dietary inclusion of gelatin up to 20 g/kg of diet improved body weight gain (BWG) compared with the diet containing no gelatin.

The lack of adequate synthesis of DAA such as Gly, Pro, Ser, and Glu, and also the increased IDAA to DAA ratio are main factors which can result in lower performance in reduced CP diets (Aftab et al., 2006). The AA profile of gelatin (Fasina et al., 2007; Khalaji et al., 2016) may make it suitable to permit reductions in CP without the commensurate reductions in performance that have been previously observed. The current study was conducted to investigate the effect of different dietary gelatin and CP concentrations and their interactions on growth performance, blood metabolites, and N retention of broiler chickens during different stages of growth.

## 2. Materials and methods

### 2.1. Broiler chickens and management

All procedures including the use of broiler chickens, management, and care were in compliance with the European parliament and the European Council Directive regulations on the protection of animals used for scientific purposes (2010/63/EU). A total of 600 male broiler chickens (Ross 308; Aviagen, Huntsville, AL) were obtained from a commercial hatchery after being vaccinated against Newcastle disease and infectious bronchitis and used in a 42-d experiment. The house was composed of insulated steel trusses with longitudinal sidewall inlets in both sides (1 m height from the floor and 0.5 m width). Broiler chickens were housed in 40 floor pens (2.25 m<sup>2</sup> per pen) containing wood shavings. Each of the floor pens was equipped with a tube feeder and a bell drinker. Broiler chickens had unlimited access to mash diets and fresh water during the experiment. During the study, the lighting program consisted of 23 h light: 1 h dark except during d 1 post-hatch when lighting was continuous. Temperature was maintained at 32 °C at placement followed by a 3 °C decrease each week to achieve 20–21 °C by using thermostatically controlled heaters, fans, and adjustable sidewall inlets.

### 2.2. Determination of apparent ileal digestibility (AID) of gelatin

The apparent ileal digestibility (AID) of AA of gelatin was determined. This ingredient has a high variability in nutrient concentration and digestibility so digestibility values were determined to improve the accuracy of diet formulation. A total of 72 one-day-old broiler chickens were housed randomly in 6 battery cages for a 3-wk period. Broiler chickens were fed a corn-soybean meal based starter mash diet (13.35 MJ ME/kg; 12.1 g Lys/kg) which its AA were supplied based on ideal protein concept. At 21 d of age, chickens were weighed and randomized into 12 battery cages. Six cages of 6 broiler chickens each were allocated to each of the 2 dietary treatments (a reference diet and a test diet; Table 1). The test diet was produced by mixing 600 g of the reference diet, and the 400 g of the gelatin as described by Widyaratne and Drew (2011). The reference diet contained 4 g chromic oxide/kg as an indigestible marker. At 28 d of age, the broiler chickens were slaughtered by cervical dislocation and the intestinal tracts were immediately excised. The distal half of the ileum samples (from Meckel's diverticulum to 2 cm proximal to the ileocecal junction) was

**Table 1**

Ingredients and nutrient composition of the reference diet (as-fed basis).<sup>a</sup>

Item	Content
Ingredients (g/kg)	
Corn	540.0
Soybean meal	400.0
Gelatin (test ingredient)	0
Soybean oil	9.7
Choline chloride	1.0
Vitamin-mineral premix <sup>b</sup>	5.0
Dicalcium phosphate	21.2
Limestone	15.3
Sodium chloride	3.8
Chromic oxide	4.0
Calculated nutrients (g/kg unless noted)	
ME <sub>n</sub> (MJ/kg)	12.74
Crude protein	181.0
Lys	10.4
Met + Cys	6.9
Ca	10.9
Non-phytate P	5.2

<sup>a</sup> The test diet was produced by mixing 600 g of the reference diet and the 400 g of the gelatin as described by Widyaratne and Drew (2011).

<sup>b</sup> Provided per kilogram of diet: vitamin A (trans-retinyl acetate), 10,000 IU; vitamin D3 (cholecalciferol), 4500 IU; vitamin E (all-rac-tocopherol acetate), 25 mg; vitamin K (bisulfate menadione complex), 3.2 mg; thiamine, 3.0 mg; riboflavin, 12.0 mg; pantothenic acid (D-calcium pantothenate), 12.8 mg; niacin, 55.64 mg; pyridoxine, 4.52 mg; folic acid, 2.2 mg; vitamin B12 (cyanocobalamin), 0.017 mg; biotin, 0.2 mg; choline (choline chloride), 600 mg; antioxidant, 100 mg; Fe (FeSO<sub>4</sub>·H<sub>2</sub>O), 100 mg; Mn (MnSO<sub>4</sub>·H<sub>2</sub>O), 120 mg; Cu (CuSO<sub>4</sub>·5H<sub>2</sub>O), 15 mg; Se (Na<sub>2</sub>SeO<sub>3</sub>), 0.30 mg; Zn (ZnSO<sub>4</sub>·H<sub>2</sub>O), 100 mg, and I (KI), 1 mg.

collected. The digesta samples of the 6 broiler chickens of each cage were pooled at the sampling time and mixed with 40 g formic acid/kg. The pooled samples were kept at –20 °C until analysis. The AID coefficients of AAs for the gelatin were calculated by the method of Ten Doeschate et al. (1993). The standardized ileal digestibility (SID) of AA was calculated from AID coefficients using the method of Yin et al. (2002), based on the mean endogenous AA composition of basal endogenous protein (Kadim et al., 2002). The AA composition and AID and SID coefficients of AA of the gelatin calculated by correcting the digestibility coefficient (DC) of the test diet for the DC of the reference diet are presented in Table 4. The major AA of gelatin were Gly, Pro, Ala, and Glu. The SID coefficients of AA of the gelatin were in the range of 0.90–0.95.

### 2.3. Experimental diets

The experimental diets were based on corn and soybean meal and formulated to meet broiler chicken (Ross 308) requirements (Aviagen, 2009) with the exception of CP in the low CP diets (Tables 2 and 3). The study was performed in a completely randomized design with 2 × 4 factorial arrangement of treatments and 5 replicates (15 broiler chickens per replicate). Two CP concentrations (200 and 220 g/kg, and 195 and 182 g/kg during the starter and grower phases, respectively) and 4 gelatin concentrations (0, 30, 60, and 90 g/kg of diet) were evaluated from d 0 to 42 of the study. Dietary treatments were formulated for the starter and grower period (d 0–21 and d 21–42, respectively) and were calculated to be isoenergetic. Also, all experimental diets were formulated to have the same levels of the IDAA including Lys, Met, Thr, and Trp. A gelatin product, which was mainly composed of cow skin, was used in the current study and analyzed for chemical composition before diet formulation (Table 4).

### 2.4. Measurement and laboratory analysis

After arrival, broiler chickens were weighed and allocated to experimental treatments to minimize body weight differences between

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