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Relative toxicity of dietary free gossypol concentration in ducklings from 1 to 21 d of age

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ABSTRACT

The aim of this study was to investigate the toxic effects of gossypol from cottonseed meal on the growth performance, serum characteristics and tissue gossypol residues in Shanshui White Ducklings. A total of 750 1-d-old ducklings (375 males and 375 females) was randomly divided into 5 dietary treatments with 5 replicate pens per treatment and 30 birds in each pen (15 males and 15 females). The experimental diets were formulated with raw cottonseed meal, de gossypol cottonseed meal and cottonseed powder as free gossypol (FG) sources. The final FG concentrations in the 5 diets were 0, 80, 160, 240 and 320 mg/kg feed. Feeding the diet with 320 mg FG/kg decreased ($P < 0.0003$) body weight (BW), average daily gain (ADG) and average daily feed intake (ADFI) of ducklings during the periods of d 1–10 and 1–21, and increased ($P < 0.0003$) feed: gain ratio (F:G) of ducklings during the period of d 1–10. Gossypol concentration in liver and muscle of ducklings measured at d 10 and 21 linearly increased ($P < 0.001$) with the increase of dietary FG levels. Notably, the diet containing 320 mg FG/kg increased ($P < 0.05$) liver index and serum alanine aminotransferase and aspartate aminotransferase activities at d 10, as well as decreased ($P < 0.05$) thymus index and serum total protein and albumin contents at d 10 and the body protein deposition at d 21. Our results indicated that ducklings fed the diet with 320 mg FG/kg resulted in an accumulation of gossypol in liver and then induced hepatic damage in the ducklings at d 10. Subsequently, the serum total protein concentrations and body protein deposition were reduced and growth rate were depressed in the ducklings at d 10 and 21 (or from d 10 to 21). Therefore, dietary FG concentration should be controlled at below 320 mg/kg to prevent an economic loss from the toxicity of FG in Shanshui White ducklings from 1 to 21 d of age.

1. Introduction

In the recent years, the growth of poultry production is limited by the increasing demand and high cost of feed protein sources in the developing countries, such as soybean meal or fish meal (Kanyinji and Sichangwa, 2014). Cottonseed meal (CSM), as a relatively rich sources in protein (30–50% dry basis) and amino acids, is used as a cheaper partial replacement of soybean meal in the poultry diets (Swiatkiewicz et al., 2016). Many studies showed that ducks, especially for local domestic ducks, can make a better use of non-

Abbreviations: ADFI, average daily feed intake; ADG, average daily gain; ALP, alkaline phosphatase; ALT, alanine aminotransferase; AST, aspartate aminotransferase; BW, body weight; Ca, calcium; CP, crude protein; CSM, cottonseed meal; CSP, cottonseed powder; DM, dry matter; F:G, feed:gain ratio; FG, free gossypol; P, phosphorus; ME, metabolizable energy; TP, total protein

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conventional raw materials including the CSM by product of cottonseed oil extraction in Asian countries. Thus, utilization of CSM in duck diets becomes more intensive than in broiler diets (Tai and Tai, 2001). However, the main concern about the usage of CSM in poultry feed is the free gossypol (FG) presence (Nagalakshmi et al., 2007). Free gossypol as a polyphenolic compound reacts with free epsilon amino groups of lysine and iron and induces the formation of protein-gossypol and iron-gossypol complexes, respectively (Abou-Donia, 1976). The chelated compounds generated from the reaction could reduce protein and iron digestibility and have negative effects on growth performance and physiological status in broilers, laying hens and ducks (Swiatkiewicz et al., 2016). Some researchers reported that feeding the increase of FG concentration in CSM diets reduced body weight gain and feed intake and decreased hemoglobin in blood or total protein content in serum of broilers (el Boushy and Raterink, 1989; Henry et al., 2001; Lordelo et al., 2005). However, not all researchers found that diets containing FG had the effects on the above aspects (Gamboa et al., 2001). These inconsistent results could be due to the differences of the FG sources (types of CSM or CSM processing) and levels (associated with the tolerance or toxicity of the animals used) in the experimental diets (Heywang and Bird, 1955; Narain et al., 1960). Broilers and laying hens are particularly sensitive to the FG over the tolerance level, which the tissues are damaged due to the accumulation of gossypol (Lordelo et al., 2005, 2007). However, the tolerance levels of FG for broilers vary widely (90–1000 mg/kg diet) depending on the age and strain of birds (Panigrahi and Morris, 1991) and the protein content and quality (Narain et al., 1960) and presence of minerals especially for the iron content in the diet (Waldeoup and Goodner, 1973). In comparison with broilers, the tolerance level of meat ducklings to FG and/or the relative toxicity of FG to meat ducklings in the diet have not been well studied and evaluated. With the increase in using CSM as a protein source in duckling feed, it is very important and necessary for diet formulators and poultry producers to know the limitations using CSM as protein sources. Therefore, the objective of this study was to investigate the toxic effects of dietary FG from the raw CSM, CSM on de gossypol and cottonseed powder (CSP) sources on the growth performance, serum characteristics and tissue gossypol residues in Shanshui White Ducklings from 1 to 21 d of age.

2. Materials and methods

All experimental diets were formulated using the raw CSM, CSM on de gossypol and CSP as FG sources were used to formulate the experimental diets. The raw CSM was obtained from a commercial oil processing plant without any detoxication (Wenfeng Trade Co., Ltd., Xinjiang, China). The CSM on de gossypol was obtained from a commercial a biotech company using the extrusion processing (Zhongmian Ziguang Biological Technology Co. Ltd., Beijing, China). The CSP was prepared by crushing and screening cottonseed through 20 meshes in our lab. The CSM, CSM on de gossypol and CSP contained 42.5, 50.0 and 38.6% CP and 623, 475, and 3878 mg/kg FG based on analyzed values, respectively.

2.1. Animals and diets

All experimental procedures were approved by the Institutional Animal Care and Use Committee of South China Agricultural University. A total of 750 1-d-old Shanshui White ducklings (375 males and 375 females) was randomly divided into dietary 5 treatments with 5 replicate pens per treatment and 30 ducks in each pen (15 males and 15 females). Treatment pens were arranged following a completely randomized design. The diets were corn-soybean based diets with or without the addition of the CSM, CSM on de gossypol and CSP. The FG concentrations of the diets were 0, 80, 160, 240, and 320 mg/kg (diets 1–5). These diets were isonitrogenous and isocaloric and formulated to meet or exceed nutrient requirements of ducks according to NRC (1994). The composition and nutrient levels for experimental diets were listed in Table 1. The FG concentrations of all diets were analyzed after formulated. All birds were allowed ad libitum access to feed and water in an environmentally controlled house with a 24 h constant light schedule. Ducklings were weighed and feed consumption was recorded by each replicate pen at 10 and 21 d of age after 12 h feed withdrawal. The ADG, ADFI, and feed: gain ratio (F:G) were calculated accordingly.

2.2. Sample collections and preparations

At 10 and 21 d of age, blood and tissue samples were collected from 6 ducklings (3 males and 3 females) based on the average BW per pen. The blood samples were collected via a bronchial vein (3.5 mL/bird), and immediately placed on ice. The serums were obtained after the blood samples were centrifuged at 2,000 × g for 15 min at 4 °C. Serums from the 6 birds in each pen was pooled in one sample with an equal volume, and then were stored at −20 °C until analysis. Birds were then killed by cervical dislocation, and liver, kidney, breast muscle, spleen, bursa of Fabricius, and thymus were removed and weighed. The organ index were calculated as organ weight (g): BW (kg) ratio. A portion of liver and breast muscle were collected and packed in ice, and then were stored at −20 °C for FG analysis. As described previously (Crespo and Esteve-Garcia, 2002), 2 ducklings (1 male and 1 female) at the start and end of experiment per replicate were randomly selected and euthanized without loss of blood, freed of gastrointestinal contents without loss of feathers. The birds were weighed and homogenized in a blade cutter for 3 min, freeze-dried and powdered. The freeze-dried samples of 2 birds from each replicate were combined as single one to determine total body protein and energy. Protein and energy deposition were calculated by difference of protein and energy contents between final and initial body, and protein deposition efficiencies were calculated as percentage of protein and energy intakes.

2.3. Sample analyses

Contents of CP, Ca and total P of in feed ingredients, diets and the freeze-dried body samples were determined using the methods