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Short communication

Effects of putrescine supplementation on growth performance, blood lipids and immune response in broiler chickens fed methionine deficient diet

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ABSTRACT

The polyamines including putrescine (PUT), spemidine (SPD) and spermine (SPM) have been shown to play an important role in basic cellular processes. Methionine (Met) and arginine are required for polyamine biosynthesis. This experiment was carried out to study the effects of Met deficient diet with or without supplemental PUT on the performance, blood lipids and glucose and humoral immunity in broiler chickens. A total of 192 day-old chicks were allocated into factorial arrangement (2 × 2) of four dietary treatments including two Met levels (Normal and Low) and two PUT levels (0 and 0.03%). Body weight and feed intake were measured weekly. At the age of 28 d, 10 birds per treatment were challenged with infectious bursal disease vaccine orally (10 dosage/bird, 2 ml). Blood samples of the challenged birds were collected for antibody determination and white blood cell count. Blood cholesterol, triglyceride, glucose, hematocrit and protein of non-challenged birds were measured at the ages of 24, 33 and 40 d. Low Met decreased body weight gain, protein efficiency ratio and feed conversion ratio ($P<0.05$). In grower period, energy efficiency ratio was improved by PUT supplementation in the chicks fed with normal Met level. Feed conversion ratio was badly affected ($P<0.05$) by low Met during grower period. PUT decreased body weight gain of chickens fed low Met significantly ($P<0.05$), indicating the importance of adequate Met level when high dietary PUT is offered. Met deficient diet decreased plasma protein level significantly ($P<0.05$). Five days post challenged, PUT supplementation elevated antibody level significantly ($P<0.05$) in the chicks fed normal level of Met. However, Met deficient diet in challenged chicks, caused significantly ($P<0.05$) lower monocyte ratio. In conclusion, growth performance declined due to Met deficiency, particularly during starter period (0–21 d) and when dietary PUT was supplemented. PUT showed the potential to increase blood antibody level and better dietary energy efficiency ratio.

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1. Introduction

The polyamines (PA) including putrescine (PUT), spermidine (SPD) and spermine (SPM) are essential components required for cell growth and function (Igarashi and Kashiwagi, 2000). PA promotes anabolic process like synthesis of DNA, RNA, protein, and increase amino acid uptake (Pegg, 1986). Biosynthesis of these polycationic compounds requires arginine and methionine (Met). These two amino acids are also important in cell division, protein synthesis and tissue growth (Seiler, 1992). Met serves the body as methyl group donor for methylation reactions of DNA and other molecules (Hoffman, 1984). Met (Shini et al., 2005) and Arg (Tayade et al., 2006) are two amino acids involving in immune regulatory action. Met is significantly affecting humoral immune system and the rate of white blood cells (Mirzaaghatabar et al., 2011). Moreover, Met deficiency is responsible for cell cycle arrest and increased of apoptosis in spleen development, influencing immune system (Bangyuan et al., 2012). In contrast, Rubin et al. (2007) demonstrated that low dietary Met while deteriorating BWG and FCR, and had no effect on the humoral immunity of chicks. Beside this dietary, Met involves in hypercholesterolaemia via enhanced hepatic cholesterol (Chol) synthesis (Hirche et al., 2006). In the meantime Kalbande et al. (2009) reported that low Met diets cause higher blood Chol and triglyceride (TG) in three weeks old broilers.

This information shows that Met as a methyl donor is involved in polyamine biosynthesis, immune response and blood lipids level. Therefore it is hypothesized that interaction between dietary polyamine and Met-deficient might be effective on immune response and blood lipids and protein. The purpose of current study was to investigate the effects of PUT supplementation in Met-deficient diets on growth performance, blood lipids and immune response of broiler chickens. This study has been performed in accordance with the ethical standards of the Ethics Committee of Animal Utilization of the University Putra Malaysia (UPM).

2. Materials and method

2.1. Experimental birds and diet

A total of 192 male day old chicks (Cobb 500) were weighed and wing-banded individually and divided into 4 dietary treatments. The treatments were factorial combination of two Met levels (Normal and Low) and two supplemental PUT (0 and 0.03%). 0.03% Met applied in current study was based on the previous study (Hashemi, 2013) that contributed best growth performance in young broilers. Met levels were 0.54 and 0.35% in starter, and 0.40 and 0.29% in grower period. All diets were formulated iso-caloric and iso-nitrogenous. Each treatment consisted of six replicates with eight chicks per replicate. The chicks had free access to experimental diets and water throughout the experimental period.

2.2. Parameters measured

Body weight (BW) and feed intake (FI) were measured weekly. Feed conversion ratio (FCR), body weight gain (BWG), energy efficiency ratio (EER) and protein efficiency ratio (PER) were calculated accordingly. FCR was calculated as daily feed intake per daily BWG. PER and EER were calculated as daily BWG (g)/protein intake (g) and daily BWG (g)/1000 cal intake of energy, respectively.

Blood samples of unchallenged chicks (4 chicks/treatment) at the ages of 24, 33 and 40 d were collected and subjected to hematocrit (HCT) using micro hematocrit method and serum protein using refractometer (ATAGO refractometer, Japan). Blood cholesterol (Chol), triglyceride (TG) and Glucose (Glu) were measured by commercial diagnostic kits (RANDOX Laboratory Ltd, UK) based on an enzymatic method.

At 28 d of age, 10 birds from each treatment were randomly selected and removed to other cages and kept inside a ventilated chamber. The chicks were challenged with infectious bursal disease (IBD) vaccine through oral administration (10 doses per bird, two ml). The IBD vaccine was obtained from Vaksindo Satwa Nusantara, Indonesia. Normal vaccination program was conducted for these birds using combined Newcastle disease (ND, Hitcher B_1 strain) and infectious bronchitis (IB, H120 strain) at 7 and 21 d of age (1 dose per birds). Blood samples were taken from the left wing vein of the birds before challenged (at 24 d of age), five days post challenged (at 33 d of age) and 11 days post challenged (at 40 d of age). Serum of challenged chicks was used to determine antibody titter against IBD and ND using ELISA kit (BioChek Poultry Immunoassays, Catalog Code CK113). Blood films were then made from both challenged and unchallenged chicks for white blood cell (WBC) count, using a microscope with 400 × magnification. Eosin and hematoxylin were used for blood staining.

2.3. Statistical analysis

The data were statistically analyzed using General Linear Model procedure of SAS software (SAS, 1999). Duncan's multiple-range test (Duncan, 1955) was used to ascertain differences among treatment means. The statistical model used was $Y_{ij} = \mu + P_i + M_j + PM_{ij} + E_{ij}$, where Y_{ij}, response variables observed from each replicate or individual birds, μ, the overall mean, P_i, the effect of dietary PUT, M_j = the effect of dietary Met, PM_{ij} = the effect due to interactions between dietary PUT and Met and E_{ij} = the statistical error.