

## Effects of Biotite V Supplementation on Growth Performance, Nutrients Digestibility and Serum and Meat Cholesterol in Broiler Chickens

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### Biotite V의 급여가 육계의 성장 능력, 영양소 소화율, 혈청 및 육내 콜레스테롤 함량에 미치는 영향

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**ABSTRACT** This study was conducted to investigate the effects of dietary supplementation of Biotite V on growth performance, nutrients digestibility and serum and meat cholesterol in broiler chickens. A total of four hundred and eighty Arbor Acre broilers (male, 2-d-old) were used in current feeding trial for 5 weeks. Broilers were allocated to three dietary treatments with eight replications per treatment and twenty broilers per pen according to a randomized complete block design. Dietary treatments included: 1) CON (basal diet), 2) BV200 (basal diet+200 mesh Biotite V 1.5%) and 3) BV325 (basal diet+325 mesh Biotite V 1.5%). During the first week of experiment, Biotite V increased weight gain in treatment groups compared to CON treatment ( $P<0.05$ ). Through the entire experimental period, weight gain, feed intake and feed/gain had no significant differences among the treatments ( $P>0.05$ ). Digestibilities of DM and N were also not affected when diets included Biotite V ( $P>0.05$ ). No effects were observed in Biotite V supplemented treatments compared to CON treatment on abdominal fat weight, meat and serum cholesterol concentrations ( $P>0.05$ ). In conclusion, supplementation of Biotite V in broiler diets has no influence on growth performance, nutrients digestibility and can not decrease the cholesterol concentrations in meat and serum.

(Key words: biotite V, growth performance, digestibility, cholesterol, broiler chickens)

## INTRODUCTION

The aluminosilicate compounds have been used as adsorbent in pigs and poultry for many years. It has been demonstrated that supplementation of hydrated sodium calcium aluminosilicates (zeolite, kaolinite and bentonite etc.) in diet or slurry can alleviate the environment pollution (Bernal and Lopez-Real, 1993; Cabuk et al., 2004). The mechanisms of these compounds are mainly related to their porosity properties which is called three-dimensional structures and highly ion selectivity and exchange capacity (Shurson et al., 1984; Moon et al., 1991). Moreover, a number of studies have also demonstrated that those aluminosilicates can bind some other toxin materials such as mycotoxins and aflatoxins (Davidson et al., 1987; Phillips et al.,

1988; Ramos and Hernandez, 1997) so that exert beneficial effects on animals.

Biotite V is such an aluminosilicate mineral product which is manufactured by Seobong Biobestech Co., Ltd. (Seoul, Korea). Previous researches suggested that Biotite V can increase growth performance, nutrients digestibility, meat cholesterol concentration and decrease fecal noxious gas emission (Kwon et al., 2003; Lee et al., 2003; Chen et al., 2005a; Chen et al., 2005b). Most of studies on Biotite V were conducted in pigs whereas its influences on poultry are not well documented until now. On the other hand, as the functions of Biotite V and aluminosilicates mainly originate from its special physical structure properties, the particle size should be also taken into account. Therefore, the objective of this study was to investigate the effects of Bio-

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tite V with different particle size on growth performance, nutrients digestibility and serum and meat cholesterol in broiler chickens.

## MATERIALS AND METHODS

### 1. Source and Compositions of Biotite V

The Biotite V used in the current experiment was manufactured by Seobong Biobestech Co., Ltd. (Seoul, Korea). The main compositions of this product were as follow: 61.90% SiO<sub>2</sub>, 23.19% Al<sub>2</sub>O<sub>3</sub>, 3.97% Fe<sub>2</sub>O<sub>3</sub> and 3.36% Na<sub>2</sub>O (Manufacturers specifications).

### 2. Experimental Design, Animals and Diets

Four hundred and eighty 2-d-old male broiler chickens were obtained from a commercial hatchery. Broiler chickens were randomly divided into three dietary treatments with eight replications per treatment and twenty broilers per pen. The experimental period lasted five weeks. Dietary treatments were as follow: 1) CON (basal diet), 2) BV200 (basal diet+200 mesh Biotite V 1.5%) and 3) BV325 (basal diet+325 mesh Biotite V 1.5%). All diets were formulated to meet or exceed the NRC (1994) requirements of broiler chickens. The compositions of the experimental diets are shown in Table 1. All diets were provided in mash form. Feed and water were available *ad libitum* throughout the experimental period.

Broilers were housed in stainless steel battery brooders with concrete floors covered by clean rice bran. The temperature was maintained at 33±1 °C in the first week and decreased 3 °C per week until 24 °C. Artificial light was provided 24 h/d by the use of fluorescent lights. Initially, there were 2 additional broilers in each pen for compensating early chick mortality during the first few days post-hatch.

### 3. Sampling and Measurements

Broilers were weighed by pen at 1, 21 and 35 day of age. Feed intake was also recorded during each period. The average feed conversion per pen was calculated by dividing the total feed consumption during the experimental period.

One week before the ending of the experiment, ten broilers were randomly selected from each treatment and caged according to treatments for determining nutrients digestibility. Diets

**Table 1.** Diet composition(as-fed basis)

Ingredients(%)	Starter <sup>1</sup>	Finisher <sup>1</sup>
Corn	55.67	63.21
Soybean meal (CP 48%)	28.25	24.61
Corn gluten meal (CP 60%)	6.50	3.50
Soybean oil	5.50	4.89
Tricalcium phosphate	2.46	2.29
Limestone	0.89	0.75
Salt	0.20	0.20
DL-methionine	0.07	0.07
L-lysine-HCl	0.06	0.08
Vitamin premix <sup>2</sup>	0.20	0.20
Trace mineral premix <sup>3</sup>	0.20	0.20
Chemical composition <sup>4</sup>		
ME(kcal/kg)	3,100	3,050
Crude protein(%)	22.00	19.00
Lysine(%)	1.10	1.00
Ca(%)	1.00	0.90
P(%)	0.80	0.75

<sup>1</sup> Starter diets, provided for 0~3 weeks; finisher diets, provided for 4~5 weeks.

<sup>2</sup> Provided per kg of diet: 15,000 IU of vitamin A, 3,750 IU of vitamin D<sub>3</sub>, 37.5 mg of vitamin E, 2.55 mg of vitamin K<sub>3</sub>, 3 mg of vitamin B<sub>1</sub>, 7.5 mg of vitamin B<sub>2</sub>, 4.5 mg of vitamin B<sub>6</sub>, 24 mg of vitamin B<sub>12</sub>, 51 mg of niacin, 1.5 mg of folic acid, 126 mg of biotin and 13.5 mg of pantothenic acid.

<sup>3</sup> Provided per kg of diet: 37.5 mg of Zn, 37.5 mg of Mn, 37.5 mg of Fe, 3.75 mg of Cu, 0.83 mg of I, 62.5 mg of S and 0.23 mg of Se.

<sup>4</sup> Calculated values.

for those broilers were mixed with chromium oxide (Cr<sub>2</sub>O<sub>3</sub>) at the level of 0.20%. Fecal samples were collected at the end of experiment and stored in refrigerator at -20°C until further analysis. Before analysis digestibility of DM and N, fecal samples were dried in a forced-air drying oven (70 °C) for 72 h and then finely ground. Subsequently, fecal samples, together with feed samples, were analyzed according to the AOAC procedures (AOAC, 1995). Chromium was analyzed by UV absorption spectrophotometry (Shimadzu, UV-1201, Japan).

At the end of the experiment, 10 broilers were randomly selected from each treatment and blood samples were collected from wing vein using sterilized injector. Samples were transferred into vacuum tube (Becton Dickinson Vacutainer Systems, Franklin Lakes, NJ) immediately and stored in refrigerator at  $-4^{\circ}\text{C}$ . Then, samples were centrifuged at  $2,000\times g$  ( $4^{\circ}\text{C}$ ) for 30 min and serum was separated. The total, LDL and HDL cholesterol concentrations in serum were determined in an automatic analyzer (Hitachi 747, Japan) by direct enzymatic kits (Boehringer Mannheim, Germany).

After collected blood samples, broilers were killed by cervical dislocation. The abdominal fat samples were collected and weighted. Thigh meat samples without skin were also taken. Then, samples were chopped, ground and frozen at  $-20^{\circ}\text{C}$  until further analysis. For analysis meat cholesterol concentration, samples were thawed and extracted with 2:1 chloroform:methanol (Bligh and Dyer, 1959). The total cholesterol concentration of thigh meat was determined according the method of Allain et al. (1974), as modified by Sale et al. (1984).

#### 4. Statistical Analyses

Statistical analyses were performed by using GLM (General Linear Models) procedure in a completely randomized block design with the SAS software program (SAS Institute, 1996). Pen was considered as the experimental unit and replication was served as a random effect. The model included the effects of block (replication) and treatment. Statements of statistical significance were based on  $P<0.05$ .

## RESULTS

Effects of Biotite V on growth performance in broilers are presented in Table 2. From 1 to 7 d of age, broilers BW were increased significantly at the sequence of CON, BV200 and BV325 ( $P<0.05$ ). Feed intake was increased slightly (114 vs 104) in BV325 treatment without statistical difference ( $P>0.05$ ). Feed/gain was not affected among the treatments. During 7 to 21 d of age, weight gain, feed intake and feed/gain were not affected by the addition of Biotite V. During 21 to 35 d of age, weight gain and feed intake increased numerically in BV treatments, however, there were no significant differences ( $P>0.05$ ).

**Table 2.** Effects of Biotite V on growth performance in broiler chickens

Items	CON <sup>1</sup>	BV200 <sup>1</sup>	BV325 <sup>1</sup>	SE <sup>2</sup>
0~7 days				
Weight gain(g)	73 <sup>c</sup>	81 <sup>b</sup>	86 <sup>a</sup>	2
Feed intake(g)	104	106	114	3
Feed/gain	1.42	1.31	1.33	0.44
7~21 days				
Weight gain(g)	525	524	538	14
Feed intake(g)	751	750	757	24
Feed/gain	1.43	1.43	1.41	0.06
21~35 days				
Weight gain(g)	893	912	910	29
Feed intake(g)	1,685	1,707	1,634	108
Feed/gain	1.89	1.87	1.80	0.09
0~35 days				
Weight gain(g)	1,491	1,517	1,534	35
Feed intake(g)	2,540	2,563	2,505	119
Feed/gain	1.70	1.69	1.63	0.05

<sup>1</sup> Abbreviations: CON, basal diet; BV200, basal diet+200 mesh Biotite V 1.5%; BV325, basal diet+325 mesh Biotite V 1.5%.

<sup>2</sup> Pooled standard error.

<sup>a-c</sup> Means in the same row with different superscripts differ ( $P<0.05$ ).

Feed/gain was lower in BV325 treatment without significant difference ( $P>0.05$ ). Through all the experimental period, no significantly differences were observed in weight gain, feed intake and feed/gain among the treatments ( $P>0.05$ ).

Effects of Biotite V on nutrients digestibility in broilers are showed in Table 3. No effect was observed on DM digestibility when diet added different particle size of Biotite V ( $P>0.05$ ). N digestibility was increased about 4% (71.27% vs 68.41%) in BV325 treatment compared to CON treatment, however, there was no significant difference ( $P>0.05$ ).

Table 4 shows the effects of Biotite V supplementation on abdominal fat weight and thigh meat cholesterol concentration in broilers. No significant difference was observed in abdominal fat weight when diets included Biotite V ( $P>0.05$ ). Thigh meat

**Table 3.** Effects of Biotite V on nutrients digestibility in broiler chickens

Items(%)	CON <sup>1</sup>	BV200 <sup>1</sup>	BV325 <sup>1</sup>	SE <sup>2</sup>
DM	80.94	81.10	82.06	0.42
N	68.41	68.93	71.27	0.85

<sup>1</sup> Abbreviations: CON, basal diet; BV200, basal diet+200 mesh Biotite V 1.5%; BV325, basal diet+325 mesh Biotite V 1.5%

<sup>2</sup> Pooled standard error.

**Table 4.** Effects of Biotite V on abdominal fat weight and thigh meat cholesterol in broiler chickens

Items	CON	BV200 <sup>1</sup>	BV325 <sup>1</sup>	SE <sup>2</sup>
Abdominal fat weight(g)	30.27	28.68	26.81	1.44
Thigh meat cholesterol (mg/dL)	125.65	121.10	110.20	5.26

<sup>1</sup> Abbreviations: CON, basal diet; BV200, basal diet+200 mesh Biotite V 1.5%; BV325, basal diet+325 mesh Biotite V 1.5%.

<sup>2</sup> Pooled standard error.

cholesterol concentration was decreased numerically (125.65 vs. 110.20) in BV325 treatment compared to CON treatment without statistical difference ( $P>0.05$ ).

Effects of Biotite V on serum cholesterol concentrations are presented in Table 5. No significant differences were observed in total cholesterol, HDL-cholesterol, LDL-cholesterol, as well as triglyceride concentrations among the treatments ( $P>0.05$ ).

## DISCUSSION

From the current study, the addition of Biotite V in broiler diets significantly increased weight gain in early period (0~7 d of age) whereas no marked improvement was observed in subsequent period (7~21 and 21~35 d of age). Previous studies regarding the use of aluminosilicate minerals on growth performance in broilers are various. Elliot and Edwards (1991) found an improvement of feed efficiency when broilers feed diets added natural zeolite. However, Amon et al. (1997) reported that dietary addition of zeolite had no effect on body weight gain and feed efficiency. Aluminosilicate materials are complex and widely diverse family (Kubena et al., 1998), therefore, this

**Table 5.** Effects of Biotite V on serum cholesterol concentrations in broiler chickens

Items(mg/dL)	CON <sup>1</sup>	BV200 <sup>1</sup>	BV325 <sup>1</sup>	SE <sup>2</sup>
Total cholesterol	140.25	136.00	140.60	10.51
Triglyceride	23.00	22.83	26.00	4.24
HDL-cholesterol	105.00	101.33	107.20	7.82
LDL-cholesterol	30.65	30.10	28.20	3.36

<sup>1</sup> Abbreviations: CON, basal diet; BV200, basal diet+200 mesh Biotite V 1.5%; BV325, basal diet+325 mesh Biotite V 1.5%.

<sup>2</sup> Pooled standard error.

may be one of the reasons that diverse results were obtained from different studies. Interestingly, Coffey and Pilkington (1989) using sodium zeolite-A in diet observed improvement in weight gain and feed efficiency in weanling pigs, but no effects were observed in their growing and finishing pigs experiments. It seems that these aluminosilicate additives exert their functions in early age rather than later growth phase. Ward et al. (1991) suggested that the related mechanism may due to a potential interaction with antibiotic in diet. As diets used in current experiment didn't include antibiotics, therefore, this may lead to inconsistent results with other studies which used diets included antibiotics. Moreover, a large number of studies reported that aluminosilicate compounds diminished many of the adverse effects which are caused by toxin materials such as aflatoxins in poultry (Kubena et al., 1998; Teleg et al., 2004). In the review of Ramos et al. (1996), they suggested that dietary addition of hydrated sodium calcium aluminosilicate may induce the formation of a stable complex between the sorbent and toxins, which consequently reduce the bioavailability of those toxins and exert positive influences on growth. The improvement of weight gain obtained in our experiment may due to the decrease of above mentioned factors. However, it should be noted that even though we found an improvement in the early age of broilers, this effect is too slightly to influence the entire growth performance.

According to the report which was conducted by Xia et al. (2004), they suggested that copper-bearing montmorillonite (aluminosilicate clay) can increase the nutrients digestibility by improving digestive enzyme activities in broilers. This observation was in agreement with some other reports (Ouhida, et al., 2000; Alzueta et al., 2002). Most previous studies about Biotite

V in nutrients digestibility were conducted in pigs and the results were not consistent. According to early study which was conducted by Chen et al. (2005a), supplementation of 0.2% Biotite V in diets increased nutrients digestibility of finishing pigs. Similar, Kwon et al. (2003) found an improvement in nutrients digestibility when growing pigs were fed diet with Biotite V. Lee et al. (2003) also observed improvement in nutrients digestibility in growing pigs, however, such effect was not found in their nursery pigs experiment. In the present study, we evaluated nutrients digestibility when broiler diet included Biotite V and didn't observe any effect. This result was similar with Thacker (2003), who reported that nutrients digestibility was not improved by the addition of Biotite V in growing-finishing pigs.

As Biotite V exert its effect mainly due to the physical structure and properties, the particle size should be considered as important as other factors such as addition level. Small particle size can extend the surface areas, so that the supplemented materials may be more evenly distributed in digestive tract and have more frequent access to digesta contents (Edrington et al., 1996). Current experiment compared two particle sizes (200 mesh vs. 325 mesh) of Biotite V. Obtained result that small particle size (325 mesh) improved weight gain in early age of broiler was in agreement with above mention standpoint.

Previous literatures are limited regarding the changes of meat and serum cholesterol by aluminosilicate agents. The results from this study indicated that Biotite V can't significantly reduce cholesterol concentrations in thigh meat and serum. In contrast to present study, Kwon et al. (2003) reported that Biotite V can reduce serum cholesterol concentration in finishing pigs. However, they didn't explain the relative mechanism of this effect. According to the report of Ponte et al. (2004), meat cholesterol can be obtained from both diet and de novo synthesize. The synthetic pathway is under feedback control from dietary cholesterol. That means when cholesterol intake is low, de novo synthesize can also produce sufficient cholesterol. Due to this reason, they suggested that only a direct alteration of cholesterol biosynthetic pathway can result an alteration of final cholesterol concentration in broiler meat. It has been well documented by *in vivo* experiment that aluminosilicate minerals have the ability of binding toxic materials such as aflatoxin. Aflatoxin can accumulate lipids in tissue especially

in liver (Huff et al., 1986; Teleg et al., 2004) and such effect may be due to the inhibition of lipid transport (Tung, et al., 1972) or from interference of lipogenesis (Donaldson et al., 1972). Therefore, aluminosilicate minerals may indirectly affect the lipids metabolism by inhibiting the activity of aflatoxin, subsequently result an alternation of tissue cholesterol concentration. However, the current study didn't support this hypothesis. Konjufca et al. (1997) suggested that plasma cholesterol which belongs to the "fast turnover cholesterol pool" (Field et al., 1960; Chobanian and Hollander, 1962) is easier to be affect compared to muscle cholesterol. Even though, our study also didn't find any alternation of cholesterol in serum. Therefore, it is rather reluctant to confirm that Biotite V can decrease serum and meat cholesterol concentration.

## IMPLICATIONS

From current experiment, no beneficial effects on growth performance, nutrients digestibility, abdominal fat and serum and meat cholesterol concentrations were observed by the supplementation of Biotite V in broiler diets. Whether biotite V is effective in preventing the toxicity material such as aflatoxin should be investigated in further research.

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## 적 요

본 시험은 육계 사료내 Biotite V의 첨가가 육계의 성장 능력, 영양소 소화율, 혈청 및 육내 콜레스테롤 함량에 미치는 영향을 조사하기 위하여 실시하였다. 총 480수의 2 일령된 Arbor Acre Broiler(♂) 병아리를 공시하였고 사양시험은 5주간 실시하였다. 시험 설계는 육계 전기 사료로서 1) CON (basal diet), 2) BV200 (basal diet+200 mesh Biotite V 1.5%) 및 3) BV325 (basal diet+325 mesh Biotite V 1.5%)를 첨가하여

제조하였다. 처리당 8반복, 반복당 20수씩 완전 임의 배치하였다. 개시시부터 1주까지의 증체량에서는 Biotite V를 첨가한 처리구가 대조구에 비하여 유의적인 증가를 보였다 ( $P < 0.05$ ). 전체 시험기간 동안의 증체율, 사료 섭취량과 사료 효율에 있어서는 Biotite V를 첨가한 처리구간에 유의적인 차이를 보이지 않았다 ( $P > 0.05$ ). 건물과 질소 소화율에서는 처리구간의 차이를 보이지 않았다 ( $P > 0.05$ ). Biotite V 첨가구에서 복강내 지방 무게, 혈청 과 육내 콜레스테롤 함량이 대조구에 비해 유의적인 차이는 없었다 ( $P > 0.05$ ). 결론적으로, 육계 사료내 Biotite V의 첨가는 성장율, 영양소 소화율, 혈청 및 육내 콜레스테롤 함량에 영향을 미치지 않았다.

(색인어: Biotite V, 성장능력, 소화율, 콜레스테롤, 육계)

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