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Effects of Dietary Garlic Supplementation on Performance and HMG-CoA Reductase in Broiler Chicks

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육계사료내 마늘의 첨가가 육계의 생산성과 HMG-CoA Reductase에 미치는 영향

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ABSTRACT

This study was conducted to determine the effect of dietary garlic supplementation on the growing performance and activity of 3-hydroxy-3-methylglutaryl coenzyme A (HMG-CoA) reductase in broiler chicks from 3 to 5 wk post hatching. Fifty chicks were divided into 5 groups with 10 replicates per treatment and placed in a wire battery cage. Five levels of dietary garlic(0, 0.1, 0.3, 0.6 and 1.0%) were provided in an one way analysis. Feed and water were given ad libitum. Feed intake, weight gain and feed conversion rate(FCR) were not affected by the garlic supplementations. The HMG-CoA reductase activity decreased significantly($P<0.05$) with the supplementation of garlic powder, compared to the garlic free group. As the dietary garlic level was increased, chicks showed decreased lipid contents in liver and blood serum. The results of this study indicate that blood cholesterol of chicks fed garlic supplemented diet might be reduced by inhibition of HMG-CoA reductase activity.

(Key words: garlic, HMG-CoA reductase, cholesterol, liver, serum, broilers)

INTRODUCTION

Various types of garlic were used in studies such as garlic powder (Sharma et al., 1979), garlic oil and paste (Qureshi et al., 1983) in laying hen, and garlic fraction in rats (Chang and John, 1980). Hypocholesterolemic and antiatherosclerotic effects of garlic have been widely studied (Itokawa et al., 1973, Krichevsky, 1975, Jain, 1976, 1977, 1978, Chi et al., 1982). The magnitude of this action ranges from a modest 14% lowering of serum cholesterol in humans (Brodia, 1981) to a lowering of 80% in cholesterol-fed hypercholesterolemic rabbits(Arora and Arora, 1981). On the other hand, Ahmad (1986) reported that chicks fed the 3.8% garlic containing diet showed significant decr-

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eases (71~83%) in hepatic HMG-CoA reductase (3-hydroxy-3-methylglutaryl coenzyme reductase : E.C. 1.1.1.34) activity. It is well known that the agents responsible for hypocholesterolemia and an increase in fibrinolytic activity of blood play an inhibitory role in the pathogenesis and development of experimental atherosclerosis, and heart disease (Bordia, 1981, Fenwick and Hanley, 1985). A Chick seems to be a proper model to study atherosclerotic changes, because lipoproteins of the bird are more closely related to those of human (Shih, 1982). Therefore, the objective of this investigation was to determine the effcet of dietary garlic powder on performance, HMG-CoA reductase activity and lipid contents in broiler chicks.

MATERIALS AND METHODS

1. Animals and diets

Day-old male broiler chicks (Abor Acres) were used in this experiment. All chicks were reared with a experimental diet and had free access to water in the electrical heated battery brooder where temperature was maintained at 37℃. Artificial lights were provided 24 hrs a day until the chicks were one week old, and then all the birds were moved to a portable wire battery. Room temperature was maintained at 25±3℃ and light was on 24 hrs daily until chicks reached two weeks of age. Thereafter, all the birds were weighed individually and divided into 5 groups of 10 chicks each. The average body weight of each group was made to be equal. They were reared with experimental diets until five weeks of age. Basal diet was formulated to contain 20% crude protein and 3,160 kcal/kg (Table 1). Five levels of dietary garlic (0%, 0.1%, 0.3%, 0.6% and 1.0%) were fed in an one way analysis design.

Table 1. Composition of basal diet

Item	%
Ingredients	
Yellow corn 8%	61.69
Gluten meal 60%	10.42
Soybean meal 44%	18.08
Canola	1.96
Calcium carbonate	0.62
Tricalcium phosphate	2.09
Salt	0.38
Choline Cl	0.11
Animal fat	3.90
Lysine HCl	0.30
DL-Methionine	0.12
Vitamin-Mineral premix[1]	0.19
Salinomysin 60	0.10
Zn bacitracin	0.05
Calculated analysis	
Crude Protein	20.00
Crude Fat	6.80
Crude Fiber	2.60
Ca	0.90
P	0.70
ME(kcal/kg)	3,160

[1] Vitamin-mineral mixture provided following nutrients per kg of diet : vit. A, 12,000 IU; vit. D_3, 2,400 IU; vit. E, 7.2 IU; vit. K, 1.44 mg; vit. B_1, 0.96 mg; vit. B_2, 3.84 mg; vit. B_6 2.88 mg; vit. B_{12}, 7.2 mg; pantothenic acid, 5.76 mg; niacin, 24 mg; choline 240 mg; folic acid, 0.48 mg; I, 0.72 mg; Fe, 57.6 mg; Mn, 84 mg; Zn, 62.4 mg; Cu, 24 mg; Co, 0.48 mg.

1) General preparation

At the end of experimental period, all chicks were weighed invidually and then used as described below.

(1) Serum

Blood samples were collected for the measurement of various lipid fractions by cutting an artery in the cervical region. Blood samples were

centrifuged at 150×g for 15 min. The supernatant were removed into test tubes and then stored at -30℃ until further analysis.

(2) Liver

The chicks were killed by cervical dislocation. Their livers were immediately removed, washed with saline, and was divided into two parts. One part was placed in 8 ml of ice-cold 0.25 M sucrose solution which contained 1 mM EDTA-2Na for the measurement of various lipid fractions. The other part was used for the measurement of HMG-CoA reductase activity. The liver preparation was made according to the procedure of Shefer et al. (1972) with slight modification. Livers were cut with scissors, homogenized and centrifuged (Model RS-18, Tomy Seiko) at 600×g at 4℃ for 15 min. Their supernatants were recentrifuged (Model 65P, RP 40-705 rotor, Hitachikoki) at 105,000×g at 4℃ for 60 min and the precipitates were rehomogenized in 0.25 M sucrose solution containing 10 mM dithiotheritol (DTT), 56 mM $MgCl_2$ and trypsin inhibitor (1mg/7.5ml). This enzyme solutions were used for measurement of the HMG-CoA reductase activity in the liver. All subsequent operations were carried out at 4℃.

2) Assay of HMG-CoA reductase activity and lipid fractions.

(1) HMG-CoA reductase activity

HMG-CoA reductase activity in chick liver was measured by the method of Shefer et al. (1972). Protein contents of the solution used for enzyme assay were determined by the method of Lowry et al. (1951). Enzyme activities were expressed as pmole of substrate converted to product per minute per mg protein at 38℃.

(2) Lipid content in chick liver and serum

The contents of various lipid fractions in the liver and serum were analyzed by the method of Tanaka et al. (1979).

The data were analyzed by the one-way lay out design of the analysis of variance. Significant differences among treatments were deteremined using Duncan's multiple range test (Duncan, 1955).

RESULTS AND DISCUSSION

Feed intakes, body weight gains, and weights of liver and abdominal fat of chicks are shown in Table 2. Although there were no statistically significant differences, feed intake was lowest in the 0.3% garlic diet group and the body weight gain was lowest when chicks were fed the 0.1% garlic diet. However, liver and abdominal fat weights of chicks fed garlic-free diet were higher than those of chicks fed garlic supplemented diets. Some workers(Qureshi et al., 1983, Chang and Johnson, 1980) reported that garlic supplementation in diets decreased the feed intake because of specific odor of garlic. However, in the present investigation, supplementation of garlic powder did not affect the broiler performance, which was in agreement with results of other studies with chicks(Sharma et al., 1979, Reddy et al., 1991). This discrepancy may be due to differences in type and quantity of the garlic and in species and age of animals.

The HMG-CoA reductase activities of chicks fed the 0.3%, 0.6% and 1.0% garlic diet were significantly lower ($P<0.05$) than those of chicks fed garlic free diet(Table 2), indiating that HMG-CoA reductase activity was reduced significantly($P<0.05$) when chicks were fed diets containing garlic more than 0.3%. Although there were no significant differences among the garlic diet groups, the activity dec-

Table 2. Effect of dietary supplemental garlic on feed intake, weight gain, feed conversion ratio(FCR), liver(%), abdominal fat(AF; %) to body weight, HMG-CoA reductase activity of broiler chicks from 3 to 5 weeks[1)]

Garlic (%)	Feed intake	Weight gain	FCR (feed/gain)	Liver	AF	HMG-CoA reductase
	g			%		p mole/mg
0	1950	1020	1.91	2.59±0.18	2.85±0.26	64.6±6.5[a]
0.1	1980	990	2.00	2.28±0.13	2.65±0.26	50.5±8.4[ab]
0.3	1800	1030	1.75	2.44±0.14	2.73±0.21	35.1±5.9[b]
0.6	1930	1010	1.91	2.48±0.19	2.74±0.10	29.0±5.8[b]
1.0	1900	1020	1.86	2.27±0.09	2.50±0.13	30.7±4.1[b]

[a,b] Mean±SE within a row with no common superscript differ significantly ($P<0.05$).

reased as the garlic content in the diet increased. The reduced HMG-CoA reductase activity by dietary garlic was also observed by other workers previously(Qureshi et al., 1983a, 1983b, Reddy et al., 1991, Noller, 1965). The organic disulfide found in the garlic oil is a good acceptor of hydrogen and its biological function may be due partly to its reaction with thiol group of substrates, reduced pyridine nucleotide (Augusti, 1974, 1977), and inhibited G-6-P and G-6-PDH (Jain, 1975). Furthermore, it was reported that the disulfide in garlic lowered blood cholesterol (Itokawa et al., 1973, Qureshi et al., 1983b).

The variations of lipid contents in liver and serum of chicks fed the garlic supplemented diets are given in Table 3. Triglyceride content of the liver of the 1.0% garlic diet group was significantly lower than those of other groups. However, the dietary garlic lowered hepatic free cholesterol and phospholipid contents significantly($P<0.05$). The serum triglyceride contents were lowered significantly($P<0.05$) by supplementing garlic powder in the diets. On the other hand, free cholesterol, cholesterol ester, phospholipid contents were not significantly different among treatments, even though their values appeared to decrease with the garlic supplementation. This study revealed that garlic powder decreased the level of lipid in the liver and serum. According to Augusti (1974), in rats, the lipid lowering effect of garlic is due to

Table 3. The contents of lipid fractions in the liver and serum of chicks fed the garlic containing diets

Garlic	Liver[1]			Serum[1]			
(%)	TG	FC	PL	TG	FC	CE	PL
	mg/g			mg/100ml			
0	57.3±5.6[a]	1.51±0.04[a]	27.8±1.5[a]	86.1±5.9[a]	20.6±0.9	150±3.6	285±7.4
0.1	55.3±5.8[a]	1.41±0.10[b]	26.6±1.0[b]	66.0±5.3[b]	18.4±0.7	136±5.5	260±9.5
0.3	56.4±10.4[a]	1.37±0.09[b]	25.9±1.5[b]	66.4±2.3[b]	16.9±0.5	137±5.2	264±10.2
0.6	51.5±5.8[a]	1.36±0.10[b]	25.6±1.7[b]	67.6±7.0[b]	17.8±0.4	140±5.0	270±8.6
1.0	34.6±4.8[b]	1.33±0.06[b]	23.5±0.6[b]	62.6±4.5[b]	17.0±0.4	135±5.8	247±8.0

[1] TG : Triglyceride, FC : Free cholesterol, CE : Cholesterol ester PL : Phospholipid

[a,b] Mean±SE within a row with no common superscript differ significantly($P<0.05$).

the decrease in the levels of triglyceride and free cholesterol. In this investigation, the hypolipidemic effect of garlic in the serum was greater than that in the liver, indicating that garlic supplementation in diet lowered the content of lipid mainly because the HMG-CoA reductase activity was significantly reduced.

적 요

본 연구는 사료내 마늘의 첨가가 육용계의 생산성 및 3-hydroxy-3-methylglutaryl coenzyme A (HMG-CoA) 환원효소에 미치는 영향을 구명하고자 실시하였다. 3주령의 육용계(Abor Acres) 50수를 처리구당 10수씩 마늘 0%, 0.3%, 0.6%, 1.0%를 첨가한 5개 처리구에 배치하여 사료와 물은 자유채식시켰다. 사료섭취량, 증체량, 사료효율에 있어서 마늘 첨가에 의한 영향은 없었다. HMG-CoA 환원효소의 활성 및 간장과 혈청내 지질함량은 마늘 첨가에 의해 유의하게 감소되었다. 따라서 본연구결과 마늘이 첨가된 사료를 급여한 육계의 혈액내 콜레스테롤의 감소는 HMG-CoA 환원효소 활성의 저하에 기인되는 것으로 사료된다.

(색인: 마늘, HMG-CoA 환원효소, 콜레스테롤, 육계)

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