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Effects of Dietary Yeast (*Saccharomyces cerevisiae*) Components on Growth Performance, Ileal Morphology and Serum Cholesterol in Male Broiler Chickens

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효모(*Saccharomyces cerevisiae*)의 급여가 육계의 생산성, 장내 융모 발달 및 혈청 콜레스테롤에 미치는 효과

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ABSTRACT An experiment was conducted to investigate whether dietary yeast (*Saccharomyces cerevisiae*, SC) and its' structural components, i.e., yeast cell-extract (YE) and yeast cell-wall (CW) could influence growth performance, ileal morphology and serum lipids of male broiler chickens. There were four dietary treatments, each consisting of 6 replicates (10 birds per replicate). Chickens were fed a corn-soybean meal base control diet and diets containing SC (0.5%), YE (0.25%) and CW (0.25%), respectively for 5-wk-experimental period. Dietary SC, YE and CW versus the control diet did not affect growth performance of male broiler chickens. Ileal morphology as to villus height, crypt depth and villus:crypt ratio of birds fed on the control diet was not significant from those fed on diets rich in SC, YE and CW, respectively. Dietary SC significantly lowered ($P<0.05$) serum total cholesterol by on average 19.7% as compared to the control group. In addition, chickens fed on diets with either YE or CW lowered serum cholesterol by on average 15.3 and 12.5%, respectively as compared to the control albeit that the former only reached statistical significance. In conclusion, our study observed the hypocholesterolemic effect of SC in male broiler chickens. Moreover, YE, i.e., an extract of intracellular components of SC contains active molecules that are responsible for lowering serum cholesterol concentrations, but their identification at the molecular level needs to be assessed.

(Key words: *Saccharomyces cerevisiae*, growth performance, cholesterol metabolism, broiler chicken)

INTRODUCTION

It is common practice to add antibiotics to poultry diets to improve chicken health, productivity and meat quality. However, it is generally accepted that the use of antibiotics may potentially affect human health due to their ubiquitous presence (Levy, 1987; Schwarz et al., 2001). This may result in proliferation of antibiotics-insensitive bacteria and thus a decrease in the therapeutic effectiveness of antibiotics used to treat a variety of bacterial infections in humans. This threat to human health has urged European countries to ban antibiotics, and alternatives to antibiotics are currently being encouraged (World Health Organization; cited by Humphrey et al., 2002).

Dietary *Saccharomyces cerevisiae* (SC) has been known to improve growth performance of broiler chickens (Valdivie, 1975; Stanley et al., 1993; Onifade et al., 1999b) and poults (Bradley et al., 1994), of which observation could substantiate its use as an alternative to in-feed antibiotics under the current intensive poultry production system. In addition, cholesterol metabolism was affected by feeding diets containing SC to

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chickens (Stanley et al., 1993). Though dietary SC has the effect on growth performance of broiler chickens, the underlying mechanism(s) are not well understood.

In this study, we used SC and two SC structural components i.e., yeast cell-extract (YE) and yeast cell-wall (CW) to see whether dietary SC could improve growth performance of broilers and whether the observed effect of SC could be also seen by feeding either YE or CW. The information obtained in this study would be of significant value further to disclose the effects of SC on growth performance of broiler chickens. Ileal morphology as to villus height, crypt depth and villus : crypt ratio was measured as dietary SC would presumably have effect on ileal environment. In addition, serum lipids were also monitored in this study.

MATERIALS AND METHODS

1. Animals, Diets and Experimental Design

240 day-old male broiler chicks (Ross) were obtained from a local hatchery. They were weighed on arrival and allotted to wire-bottomed, suspended cages in an environmentally controlled facility. Continuous lighting was used throughout the experimental period. Room temperature with the cages was set at 34℃ at beginning and gradually decreased to 25℃ at 3 weeks and kept constant thereafter. There were four dietary treatments, each consisting of 6 replicates. A replicate was identical to cage with 10 birds so that each treatment had 60 chicks. Starter and finisher diets were formulated and used as a control diet (Table 1). To compose an experimental diet, SC[1)] was added to the control diet to reach at the concentration of 0.5% at the expense of soybean meal. In addition, CW and YE[2)] were introduced to the control diet to reach 0.25% in diets, and the former was substituted with soybean meal whereas the latter with corn. All diets used in this study did not contain exogenous cholesterol. Feeding periods were 0~21 days (starter) and 21~35 days (finisher). Diet and water were provided *ad libitum*.

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Table 1. Ingredients and composition of the basal diet[1]

Ingredients	Starter (0~3wk)	Finisher (4~5wk)
	---------------- g/kg ----------------	
Corn	592.6	646.3
Soybean meal (44 %)	205.2	189.2
Corn gluten meal	94.3	71.7
Rapeseed meal	50.0	30.0
Soybean oil	20.0	30.0
Tricalcium phosphate	17.8	12.7
Limestone	8.8	10.8
Salt	4.0	4.0
DL-methionine (50 %)	3.4	1.6
L-lysine HCl	1.9	1.7
Vitamin premix[1]	1.0	1.0
Mineral premix[2]	1.0	1.0
Total (%)	1000	1000
Calculated chemical composition;		
ME (kcal/kg)	3,100	3,100
CP	215.0	190.0
Ca	10.0	9.0
Available P	4.5	3.5
Methionine	5.0	3.8
Lysine	11.0	10.0

[1] Provided followings per kilogram of diet: vitamin A, 5,500 IU; vitamin D_3, 1,100 IU; vitamin E, 11 IU; vitamin B_{12}, 0.0066 mg; riboflavin, 4.4 mg; pantothenic acid, 11 mg (Ca-pantothenate: 11.96 mg); choline, 190.96 mg (choline chloride 220 mg); menadione, 1.1 mg (menadione sodium bisulfite complex 3.33 mg); folic acid, 0.55 mg; pyridoxine, 2.2 mg (pyridoxine hydorchloride, 2.67 mg); biotin, 0.11 mg; thiamin, 2.2 mg (thiamin mononitrate 2.40 mg); ethoxyquin, 125 mg.

[2] Provided followings per kilogram of diet: Mn, 120 mg; Zn, 100 mg; Fe, 60 mg; Cu, 10 mg; I, 0.46 mg; and Ca, min: 150 mg, max: 180 mg.

2. Measurements

All chicks were weighed by pen on a weekly basis. Feed intake by pen was monitored weekly and used to calculate feed:gain ratio. At the end of feeding trial, blood from one chicken per cage was collected from wing vein into a vacuum tube and kept at room temperature. Serum was obtained by gentle centrifugation and stored at -20℃ before required for lipid analysis. Serum total and high-density-lipoprotein (HDL) cholesterol, and triglycerides were measured as described earlier (Yeom et al., 1999) on an automatic analyzer[3]. Low-density-lipoprotein (LDL) cholesterol was obtained from the formula as proposed by Friedwald et al. (1972). Immediately after blood sampling, birds were humanely killed by cervical dislocation. A 3-cm long segment posterior to ileo-cecal value was then sampled and fixed in 10% neutral buffered formalin for the measurement of ileal morphology. The samples fixed in formalin were cut and embedded in paraffin wax. Sections cut at 4~5 μm were stained with hematoxylin and eosin. Each slide was measured for villus height (μm) and crypt depth (μm) from 59 microscopic fields using an image analysis system. Thereafter, villus:crypt ratios were calculated.

3. Statistical Analysis

All data obtained in this study were evaluated by one-way analysis of variance (Steel and Torrie 1980). When significant treatment effects were disclosed, statistically significant differences among treatment means were identified by the multiple range test of Duncan (1955). The level of statistical significance was pre-set at $P<0.05$.

RESULTS

1. Growth Performance and Ileal Morphology

This study was completed without problems. There was no mortality occurred throughout the 5-wk-experimental periods. Table 2 presents growth performance of broiler chickens fed a control diet and diets enriched with either SC, YE or CW, respectively for 35 days. At 3 weeks, weight gains of birds fed on the diet containing SC did not differ from the no-added control counterparts. Both YE and CW failed to affect weight gains of broiler chickens. Feed intakes and feed:gain ratios at 3 weeks of age were not affected by dietary treatments. Furthermore, no significant impact of SC, YE and CW on growth performance that measured at either 3~5 weeks or 0~5

Table 2. Growth performance of male broiler chickens fed diets containing yeast (*Saccharomyces cerevisiae*, SC), yeast cell-extract (YE) and yeast cell-wall (CW) for 35 days

Traits	Treatments[1]			
	Control	SC	YE	CW
0~3 weeks				
Weight gain, g	635.8±30.0	654.4±24.7	646.5±16.8	665.9±27.9
Feed intake, g	1182.9±26.2	1158±39.4	1174.7±35.4	1183.6±38.8
Feed:gain, g:g	1.86±0.12	1.77±0.04	1.82±0.08	1.78±0.07
3~5 weeks				
Weight gain, g	1002.2±43.6	1012.0±29.5	997.1±49.6	1025.7±46.9
Feed intake, g	1808.9±30.2	1776.2±39.5	1788.0±31.2	1828.0±61.9
Feed:gain, g:g	1.81±0.08	1.76±0.05	1.80±0.08	1.79±0.11
0~5 weeks				
Weight gain, g	1637.5±70.0	1666.5±45.4	1643.1±63.1	1691.5±69.1
Feed intake, g	2952.7±30.2	2920.0±39.5	2931.8±31.2	2972.0±61.9
Feed:gain, g:g	1.81±0.08	1.75±0.04	1.79±0.06	1.76±0.08

[1] Values are expressed as means (±SD) of six replicates per dietary treatment.

3) Hitachi 7150, Hitachi Medical Co., Japan

weeks was seen. Group-mean weight-gains ranged between from 997 g to 1,026 g at 3~5 weeks and from 1,638 g to 1,692 g at 0~5 weeks. Feed:gain ratios did not differ between dietary treatments, the ratios being 1.75~1.86 in all ages.

Table 3 shows that ileal villi lengths of birds fed on the control diet were not different ($P>0.05$) from those fed on the diets rich in SC, YE and CW, respectively. The crypt depth appeared to range from 72.4 to 77.2 μm without reaching statistical significance between dietary treatments. Consequently, villus:crypt ratios were not significant between dietary treatments, the ratios being 8.1 to 8.7.

2. Cholesterol Metabolism

Table 4 presents serum lipid profiles of male broiler chickens fed diets containing either SC, YE or CW, respectively for 35 days. It was apparent that feeding SC to broiler chickens significantly lowered ($P<0.05$) serum total cholesterol by on average 19.7% as compared to the control. In addition, chickens fed on the diets containing either YE or CW lowered serum cholesterol by on average 15.3 and 12.5%, respectively when compared to those fed on the control diet albeit that the former only reached statistical significance. Serum triglycerides were lowest for the chickens fed the SC-added diet and highest for the control-fed group, but the group differences did not reach statistical significance. HDL cholesterol was not affected ($P>0.05$) by dietary treatments and ranged from 87 to 90 mg/dL of serum (Table 4). On the other hand, the ratio of HDL cholesterol to total cholesterol was significantly higher in the SC-fed group versus no-added group whereas the ratio showed intermediate for the CW and YE groups. Supplementation of SC into the control diet significantly lowered serum LDL cholesterol by on average 32.3%. Chickens fed on the diets containing CW and YE lowered LDL cholesterol by on average 9.2% and 23.5%, respectively as compared to those fed the control diet, however the reduction was not of statistical significance.

Table 3. Villus length, crypt depth and villus/crypt ratio of male broiler chickens fed diets containing yeast (*Saccaromyces cerevisiae,* SC), yeast cell-extract (YE) and yeast cell-wall (CW) for 35 days

Traits	Treatments[1]			
	CO	SC	YE	CW
Villus length, μm	620.9±30.7	650.6±106.1	598.6±70.3	645.4±87.2
Crypt depth, μm	77.2± 5.3	76.2±14.7	72.4±12.3	76.6±11.6
Villus : crypt ratio	8.1± 0.6	8.7± 1.5	8.4± 1.1	8.5± 1.2

[1] Each value is the mean±SD of 59 microscopic fields per segment with 6 birds per treatment.

Table 4. Serum lipids of male broiler chickens fed diets containing yeast (SC), yeast cell-extract (YE) and yeast cell-wall (CW) for 35 days

Traits	Treatments[1]			
	Control	SC	YE	CW
Total cholesterol, mg/dL	184.0±12.8[a]	147.8±17.6[b]	155.8± 8.3[b]	161.0±11.8[ab]
Triglycerides, mg/dL	104.8±15.8	74.7±29.6	83.5±15.4	75.0±16.6
HDL cholesterol, mg/dL	90.1± 2.5	88.1± 6.3	86.7± 3.4	89.4± 4.2
LDL cholesterol, mg/dL	50.2±12.9[a]	34.0± 7.5[b]	38.4± 7.0[ab]	45.6± 6.1[ab]
HDL cholesterol, % of total	49.1± 3.4[b]	60.0± 5.7[a]	55.7± 2.7[ab]	55.7± 2.8[ab]

[1] Values are expressed as means (±SD) of six replicates per dietary treatment.

[a,b] Means in a row with no common superscripts differ significantly ($P<0.05$).

DISCUSSION

1. Growth Performance and Ileal Morphology

Adding SC, YE and CW into a control diet did not affect growth performance of male broiler chickens in all ages. In reality, our study is unexpected in the light of previous studies showing that dietary SC improved growth performance of broiler chickens (Valdivie, 1975; Stanley et al., 1993; Onifade and Babatunde, 1996; Onifade et al., 1999b; Zhang, 2004). The discrepancy between our study and others (Valdivie, 1975; Stanley et al., 1993; Onifade and Babatunde, 1996; Onifade et al., 1999b; Zhang, 2004) is not readily understood, but environment may play a role by attenuating the effect of SC, YE and CW, if any, on growth performance. This assumption is reinforced by the observation that all dietary treatments failed to affect ileal morphology. None the less, it should be remembered that dietary SC has been clearly shown the growth-stimulating effect in broiler chickens by study of Zhang (2004), which the experimental condition was identical to that used in this study.

2. Cholesterol Metabolism

Dietary SC has been known to lower cholesterol concentrations in chickens (Stanley et al., 1993) and rabbits (Onifade et al., 1999a), being inclusion levels ranged between 0.1~0.3% in diets. As reported earlier (Stanley et al., 1993), dietary SC significantly lowered serum cholesterol of broiler chickens that were fed for 35 days in this study. It is also clear from our study that SC-induced reduction in serum cholesterol was closely related to subsequent reduction in LDL cholesterol. On the other hand, HDL cholesterol was not changed by dietary treatments, thus favoring higher percentage of HDL cholesterol in chickens fed on the SC-treated diet. Our study showed that hypocholesterolemic component(s) in SC can be attributed to either CW or YE, albeit that the latter was more effective in reducing serum cholesterol. The reduction in serum cholesterol by YE was closely associated with LDL cholesterol.

CW contains β-glucan that is known to affect cholesterol metabolism in broiler chickens Fadel et al., 1987; Peterson and Qureshi, 1997) and in human (Nicolosi et al., 1999). Soluble fibers such as beta-glucan are known to increase intestinal viscosity which interferes fat and cholesterol absorption (Lee et al., 2004). It can be postulated that CW-effect on cholesterol metabolism would be diminished as our experimental diets contained no-added cholesterol. YE is soluble in water and consists of essential amino acids, vitamins and minerals. Especially, YE contains niacin at 410 mg/kg and Ca-pantothenate at 180 mg/kg as main vitamin components (Zhang, 2004). This concentration would provide a surplus of 10.3 mg of niacin and of 4.5 mg of Ca-pantothenate to the diet rich in YE versus no-added control diet on a kg basis. Whether these extra vitamins could modulate cholesterol metabolism in broiler chickens needs to be answered.

In conclusion, the present study failed to confirm the growth-stimulating effect of SC in male broiler chickens. Our study, however, observed the hypocholesterolemic effect of SC with male broiler chickens. Moreover, YE, i.e., an extract of intracellular components of SC, contains active molecules that are responsible for lowering serum cholesterol concentrations, but their identification at the molecular level needs to be assessed.

적 요

효모(*Saccharomyces cerevisiae, SC*)의 구성성분인 yeast cell-extract(YE)과 yeast cell-wall(CW)이 육계의 생산성, 장내 융모 발달 및 혈청 콜레스테롤에 어떠한 영향을 미치는지 알아보고자 사양실험을 실시하였다. 육용 수평아리(Ross) 240수를 4처리 6반복, 반복당 10수를 공시하였다. 옥수수 및 대두박 위주 기본사료에 SC, YE 그리고 CW를 각각 0.5, 0.25 그리고 0.25%로 첨가한 실험사료를 제조하여 육계에 5주간 급여하였다. 효모의 급여는 육계의 생산성에 어떠한 영향을 미치지 못하였으며, villus height, crypt depth 및 villus:crypt ratio 역시 처리간 유의성은 발견되지 않았다. SC를 급여받은 육계의 혈청 콜레스테롤은 대조구에 비하여 19.7% 감소($P<0.05$)를 나타내었다. 특히, 효모성분 중 YE가 혈청 콜레스테롤을 유의적으로 낮추었다($P<0.05$). 결론적으로, 사료내 효모의 첨가는 육계의 혈청 콜레스테롤을 유의적으로 감소시켰으며($P<0.05$), 콜레스테롤을 감소시키는 성분은 효모의 세포내용물에 포함되어 있는 것으로 본 연구결과 알 수 있었다.

(색인 : 육계, 생산성, 효모, 콜레스테롤대사)

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