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Effect of probiotic (*Saccharomyces cerevisiae*) on performance of broiler chicks

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Abstract

One hundred, one-day-old male broiler chicks (Faobrow CD), were randomly assigned to 5 treatments (20 birds/treatment). Treatment groups were; Control diet without yeast addition (treatment 1), baker yeast 0.5% (treatment 2), baker yeast 1% (treatment 3), baker yeast 1.5% (treatment 4), baker yeast 2 % (treatment 5). Chicks were reared for 21 days. Body weight, feed consumption and feed conversion were weekly determined. All chicks were scarified at the end of 21 days and blood samples were taken from brachial vein for blood analysis, including total RBCS, hemoglobin, Packed cell volume, total white blood cells, differential leukocyte count. Serum samples were taken and assayed for estimation of total protein, triglycerides, cholesterol, calcium, uric acid, glucose, ALT and AST enzymes. Chicks in all treatments were killed at the end of the experiment by cervical dislocation, and liver, gizzard, heart, spleen, proventriculus, bursa of Fabricius, pancreas and kidney were collected, weighed, and calculated as a percentage of body weight. Body weight gain for the entire period (3 weeks) were significantly ($P<0.05$) increased in the treatments 3, 4 and 5, when yeast was added at a rate of 1, 1.5 and 2%, compared with the other treatments (1 and 2). These birds also had a significantly ($P<0.05$) higher feed intake and feed conversion ratio than others. There was no significant difference in the relative organs weights in all treatment groups. No significant changes in hematological parameters, except in hemoglobin improvement at the highest Sc inclusion rate of 2%. No differences were recorded in the percentages and absolute numbers of different WBCs in all treatment groups. Addition of yeast at a rate of 1, 1.5 and 2 %, were significantly ($P<0.05$) increase levels of total serum protein and glucose, compared with other treatments. All yeast additive levels were responsible for significant ($P<0.05$) reduction in serum triglycerides, but only at highest level (2%) for serum cholesterol, compared with other treatments. No effect on serum calcium and uric acid levels, and ALT, AST serum activities, were observed by the addition of all Sc additives levels, compared with control treatment.

Keywords: Probiotic, Chicken, Performance.
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تأثير استخدام خميرة الخبز الجافة (*Saccharomyces cerevisiae*) كمعزز حيوي في أداء أفراخ فروج اللحم

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الخلاصة

وزع ١٠٠ من ذكور أفراخ فروج اللحم نوع فاوبرو بعمر يوم واحد على خمسة مجاميع وبواقع ٢٠ فرخ لكل معاملة. وهذه المجاميع كالأتي: عليقه سيطرة (المجموعة الأولى)؛ إضافة 0.5% خميرة الخبز الجافة (المجموعة الثانية)؛ إضافة ١ % من الخميرة (المجموعة الثالثة)؛ إضافة 1.5 % من الخميرة (المجموعة الرابعة؛ إضافة ٢% من الخميرة (المجموعة الخامسة). تم تربية الأفراخ لمدة ٢١ يوم وحسبت كل من معدل الزيادة الوزنية و استهلاك العليقة و معامل التحويل الغذائي أسبوعيا. أخذت عينات دم بعد انتهاء

التجربة لقياسات الدم وقياسات مصل الدم المختلفة. تم بعد خلع الرقبة لطيور المجاميع المختلفة وأخذت مختلف الأعضاء الداخلية لحساب وزنها النسبي. أوضحت النتائج أن إضافة ١، ١,٥ و ٢% من خميرة الخبز الجافة قد أدى إلى تحسن معنوي (P<0.05) في معدل الزيادة الوزنية,استهلاك العليقة ومعامل التحويل الغذائي مقارنة مع المجاميع الأخرى. لم يكن هناك فروق معنوية بين جميع المجاميع في الوزن النسبي للأعضاء الداخلية. لم يكن هناك تأثير معنوي على مختلف مقاييس الدم عدا الزيادة في مستوى الهيموغلوبين عند إضافة الخميرة بمستوى ٢%. لم يكن هناك فروقا معنوية في النسب المئوية والأعداد المطلقة لخلايا الدم البيض بين كافة المعاملات. سجلت زيادة معنوية (P<0.05) في مستوى الكلوكوز والبروتين في مصل الدم وانخفاض معنوي (P<0.05) في مستوى الترايكلايسيريدات عند إضافة الخميرة بالمستويات ١، ١,٥ و ٢% ; وكذلك انخفاض معنوي (P<0.05) في مستوى الكولسترول ولكن عند إضافة الخميرة عند مستوى ٢%. لم يسجل تأثير معنوي على مستوى ALT, AST في مصل الدم عند المقارنة بمجموعة السيطرة. .

Introduction

A popular alternative to the use of antibiotics has been the use of probiotics which have been used in poultry for "competitive/exclusion" of bacterial pathogens (1). The positive effects of probiotics on animals can result either from a direct nutritional effect of the probiotic, or a health effect, with probiotics acting as bioregulators of the intestinal microflora and reinforcing the host's natural altitude defenses. There have been numerous studies in humans and animals on the ability of probiotics to change the types and numbers of gut microflora (2-4). Gong et al. (5) define probiotics as health-promoting bacteria inhabiting the gastrointestinal tract of humans and animals. Exactly how supplemental dietary microbial products function in the digestive system is not known, but some suggested mechanism are that they: 1) provide nutrients, 2) aid in digesting foods, and 3) inhibit harmful bacteria (6). Since Tortuero (7) found that Lactobacillus cultures improve broiler growth, many investigations (8,9) have been conducted to determine the effects of probiotic bacteria, mainly the lactic acid bacteria (LAB), on the performance of domestic animals, especially poultry species. Supplementing broilers with microbial cultures provides beneficial bacteria to aid in nutrient absorption and enhance the microbial balance in the avian digestive tract. Therefore, probiotics are used to get rid of stress-induced abnormalities in the gastrointestinal tract, thus normalizing gut activity (10). Some reports (11,12) showed that additional benefits can be gained by supplementing broiler diets with probiotics as feed additives. Cavazzoni et al. (13) found that feeding probiotic supplements based on Bacillus coagulans enhanced the growth rate of broilers. The objective of this study was to determine the effects Saccharomyces cerevisiae as a dietary probiotic source, on broiler performance, haematological profile and blood biochemistry.

Materials and methods

Chicks and diet

One hundred, one-day-old male broiler chicks (Faobrow CD), were obtained from a commercial hatchery, individually weight, wing-banded and housed in experiment room and continuous fluorescent lighting. The birds were randomly assigned to 5 treatment groups (20 birds/each). Control diet without yeast addition (treatment 1), baker yeast 0.5% (by 3.44x108 CFU/g of Saccharomyces cerevisiae concentration for 37C0 for 3 days) (treatment 2), baker yeast 1% (treatment 3), baker yeast 1.5% (treatment 4), baker yeast 2% (treatment 5). Feed and water were provided for ad libitum consumption. Chicks were reared in individual wire cages for 21 days and fed a typical broiler diet with 22.0 % crude protein and 2950 metabolizable energy (Kcal/kg). Diets were designed to satisfy the recommendations of the NRC (14).

Performance

Body weight gain, feed consumption and feed conversion ratio were weekly determined. Dead birds were daily recorded.

Hematological parameters

All chicks were scarified at the end of 21 days and blood samples were taken from brachial vein for blood analysis, including total RBCS, hemoglobin, Packed cell volume, total white blood cells, differential leukocyte count, according to (15).

Serum biochemistry

Serum samples were taken and stored under (-20 ºC) until assayed for estimation of total protein, triglycerides, cholesterol, calcium, uric acid, glucose, ALT and AST enzymes using standard kits (BioMereux, France; Randox, England; Biomaghreb, USA).

Relative organs weight

Chicks in all treatments were killed at the end of the experiment by cervical dislocation, and liver, gizzard, heart, spleen, proventriculus, bursa of Fabricius, pancreas and kidney were collected, weighed, and calculated as a percentage of life body weight.

Statistical analysis

All experimental data were subjected to the analysis of variance (16). Least square means were compared by Duncan's multiple range test. All statements of differences were based on significance of P<0.05.

Results

Data presented in Table 1 showed the effect of graded levels of the yeast, on body weight gain, feed consumption and feed conversion in all treated chicks. Body weight gain for the entire period (3 weeks) were significantly (P<0.05) increased in the treatments 3, 4 and 5, when Sc was added at a rate of 1, 1.5 and 2%, as compared with the other treatments. Best results were seen in treatments 4 and 5. Moreover, these birds also had significantly higher feed intake and feed conversion ratio than others (P<0.05). In all treatments no mortality was recorded (Table 1).

No any adverse effects on various organs relative weights was noted in birds of all treatments (Table 2).

Yeast at its four levels 0.5, 1, 1.5, and 2%, did not adversely influence blood erythrocyte counts, hemato-crit, mean corpuscular volume, mean corpuscular hemo-globin, mean corpuscular hemoglobin concentration, except a significant (P<0.05) improving in hemoglobin parameter at the highest yeast rate of 2%. In general the best results among all treatments were noticed when higher yeast level (2%) was added (Table 3).

No reduction in total WBCs were recorded in chicks of all treatment groups with or without yeast at its four graded levels. No change in the percentages of heterophils, lymphocytes, monocytes, eosinophils and basophiles was noticed in all treatment groups fed yeast (Table 4), and so their absolute numbers (Table 5), compared with those of the control group.

Table 1: The effect of yeast on body weight gain, feed consumption and feed conversion of broilers at 3 weeks of age.

Treatments	yeast %	Body weight gain(g)				Feed consumption (g)	Feed conversion ratio (g/g)	Mortality %
		1-7 d	8-14 d	15-21d	1-21 d			
1	0	57±2.0 a	104±3.0 a	140±2.0 c	301±3.5 cd	410±15.8 c	1.362±0.117 b	0
2	0.5	57.6±2.4 a	105±2.6 a	139.4±2.4c	302±2.2b cd	411±17.0b c	1.360±0.106 ab	0
3	1	58.1±1.9 a	108.6±2.5a	142.9±3.9bc	309.6±3.2 b	421±18.2 b	1.359±0.102ab	0
4	1.5	58.6±2.4 a	110.4±2.9a	149.3±3.3ab	318.3±2.8a	430±18.7 a	1.350±0.114 a	0
5	2	60.2±2.2 a	111±2.8 a	152.7±2.7a	323.9±2.1a	434±18.8 a	1.339±0.112 a	0

Table 2: Effect of yeast on relative weights of different internal organs in birds at 3 weeks of age.

Treatments	yeast %	Relative organs weight (g/100g)							
		Liver	Gizzard	Heart	Spleen	Proventriculus	Bursa of Fabricius	Pancreas	Kidney
1	0	2.845± 0.08	3.924± 0.15ab	0.503± 0.02ab	0.032± 0.02	0.583± 0.02	0.289± 0.01	0.433± 0.02	0.700± 0.05
2	0.5	2.871± 0.01	4.002± 0.08a	0.542± 0.04a	0.036± 0.01	0.575± 0.03	0.290± 0.05	0.434± 0.09	0.711± 0.02
3	1	2.861± 0.02	4.050± 0.24a	0.552± 0.02a	0.034± 0.02	0.576± 0.01	0.291± 0.05	0.436± 0.01	0.714± 0.11
4	1.5	2.891± 0.02	3.990± 0.04a	0.514± 0.09ab	0.036± 0.01	0.539± 0.04	0.293± 0.03	0.437± 0.12	0.716± 0.01
5	2	2.885± 0.09	3.982± 0.17a	0.505± 0.06ab	0.033± 0.06	0.565± 0.14	0.297± 0.09	0.439± 0.05	0.722± 0.06

Addition of yeast at a rate of 1, 1.5 and 2 %, were significantly ($P<0.05$) increase the level of serum total protein, compared with other treatments. All yeast additive levels (0.5, 1, 1.5 and 2%) were responsible for significant ($P<0.05$) reduction of serum triglycerides, but only at high (2%) level for serum cholesterol, compared with other treatments. No effect on serum calcium levels, were observed by the addition of all yeast additives levels, compared with control treatment (Table 6).

Addition of yeast at a rate of 1, 1.5 and 2 %, were significantly ($P<0.05$) increase the level of serum glucose, compared with other treatments. No effect on serum uric acid levels, and ALT, AST serum activities, were observed by the addition of all yeast additives levels, compared with control treatment (Table 7).

Table 3: Effect of yeast on hematological parameters in chicks at 3 weeks.

Treatments	Yeast %	Erythrocyte count (x106/mm3)	Hemoglobin g %	Hematocrit %	Mean corpuscular volume (µm3)	Mean corpuscular hemoglobin (pg)	Mean corpuscular hemoglobin concentration (%)
1	0	2.63±0.01ab	7.1±0.1b	30.7±0.7ab	116.730±2.227a	26.996±0.277ab	23.127±0.206ab
2	0.5	2.60±0.40ab	7.0±0.3b	29.5±0.4bc	113.461±22.448ab	26.923±1.580ab	23.728±1.32ab
3	1	2.81±0.08ab	8.3±1.0ab	31.0±2.0ab	110.320±10.320ab	29.537±4.365ab	26.774±1.602ab
4	1.5	2.72±0.12ab	7.5±0.5b	30.2±2.2ab	111.029±3.337ab	27.573±0.65ab	24.834±0.166a
5	2	2.85±0.20 a	9.2±0.6 a	34.0±0.6 a	119.290±12.785 a	32.280±0.172 a	27.05±2.479 a

Table 4: Effect of yeast on Total leucocyte counts, and their percentages in growing chicks at 3 weeks of age.

Treatments	yeast %	Leucocytes count (10^3/mm^3)	Heterophils	Lymphocytes	Monocytes	Eosinophils	Basophiles
1	0	25.0±0.10	28±4.45	62±6.72	6±2.54 a	2±0.59	2±0.07
2	0.5	24.7±0.70	27±5.20	64±8.35	6±1.34 a	1±1.00	2±0.70
3	1	23.9±0.90	25±9.00	72±8.25	1±0.77 ab	1±0.55	1±0.98
4	1.5	24.5±0.50	24±3.80	69±1.90	5±1.03 ab	1±0.90	1±0.90
5	2	27.6±0.60	30±8.00	61±1.05	6±1.41 a	2±0.20	1±0.80

Table 5: Effect of yeast on absolute number of different leucocytes in growing chicks at 3 weeks of age.

Treatments	yeast %	Heterophils	Lymphocytes	Monocytes	Eosinophils	Basophiles
1	0	7.00±1.00	15.50±1.500	1.50±0.500a	0.50±0.001a	0.50±0.010a
2	0.5	6.67±1.081	15.80±1.480	1.48±0.042a	0.25±0.005ab	0.49±0.240a
3	1	5.97±1.860	17.21±1.195	0.24±0.009ab	0.24±0.009ab	0.24±0.021ab
4	1.5	5.88±0.615	16.90±0.105	1.22±0.475a	0.24±0.005ab	0.25±0.005ab
5	2	8.26±0.990	16.84±0.096	1.66±0.633a	0.55±0.270a	0.28±0.006ab

Table 6: Effect of yeast on serum total protein, triglycerides, cholesterol and calcium levels of broiler chicks at 3 weeks of age.

Treatments	Yeast %	Total protein (g/100ml)	Triglycerides (mg/100ml)	Cholesterol (mg/100ml)	Calcium (mg/100ml)
1	0	3.33±0.03 b	424.78±4.21 a	199.71±6.53 a	11.23±0.14 a
2	0.5	3.41±0.08 b	328.46±2.89 b	179.83±3.69 ab	11.19±0.13 ab
3	1	3.40±0.02 a	295.65±0.35 c	176.14±4.55 ab	10.59±0.16 ab
4	1.5	3.78±0.09 a	282.58±3.03 c	180.09±1.73 ab	10.80±0.36 ab
5	2	4.05±0.03 a	297.57±0.68 c	168.83±4.33 b	11.33±0.03 a

Table 7: Effect of Sc addition on serum uric acid, glucose levels, and ALT and AST serum activities of broiler chicks at 3 weeks of age.

Treatments	Yeast %	Uric acid (mg/100ml)	Glucose (mg/100ml)	ALT (IU/L)	AST (IU/L)
1	0	8.40±0.84 a	299.2±2.05 b	15.91±1.4	45.50±4.5
2	0.5	6.65±0.01 ab	298.1±3.48 b	18.48±4.2	46.02±0.9
3	1	6.86±1.01 ab	311.1±3.27 a	18.83±5.9	50.18±11.9
4	1.5	6.40±1.41 ab	326.2±2.45 a	21.04±6.8	63.53±18.5
5	2	7.20±0.21 ab	307.8±2.69 a	17.94±1.6	45.01±1.7

Discussion

In this experiment baker yeast (*Saccharomyces cerevisiae)* supplementation of broilers, to the level of 1, 1.5 and 2%, were significantly (P<0.05), increase the body weight gain, feed consumption and feed conversion efficiency, compared with control group and group added to its diet 0.5 bakers yeast. No negative effects were exert on the addition of *Saccharomyces cerevisiae* at all inclusion levels on internal body organs nor the hematological picture as compared with control group. The beneficial effect of *Saccharomyces cerevisiae* is attributed to the fact that it is a naturally rich source of proteins, minerals and B-complex vitamins (17). It is well known that yeast culture, and its cell wall extract containing 1,3-1,6 D-glucan and Mannan-oligosaccharide are the important natural growth promoters for modern livestock and poultry production (18). The advantages of these promoters over the traditional antibiotic growth promoters are 1) no withdrawal time, 2) no residual effect, and 3) no causes of microbial mutation (19). *Saccharomyces cerevisiae* is considered as one of the live microorganisms probiotic that, when administered through the digestive tract, have a positive impact on the hosts health through its direct nutritional effect (20). Field reports (21,22) have suggested that probiotic supplementation improved performance of broilers. The different mechanisms of probiotic action suggested are; nutritional effect by regulation of metabolic reactions that produces toxic substances; stimulation of endogenous enzymes and by production of vitamins or antimicrobial substances (23). Moreover, *Saccharomyces cerevisiae* could act as bioregulator of the intestinal micro flora and reinforcing the host natural defenses, through the sanitary effect by increasing the colonization resistance and stimulation of the immune response (24). These effects were largely reflected by using mannan Oligosaccharide**,** the naturally derived extract from the cell wall of *Saccharomyces cerevisiae*. This oligosaccharide content is approxi-mately 50% of the carbohydrate fraction and improved body weight gain in broiler chickens and that this effect can be attributed to the trophic effect of this product on the intestinal mucosa, because it increases villus height, particularly during the first 7 days of the chickens life (25). Mortalities were not reported in all groups of this experiment. This observation could be in accordance with that mention mannan Oligosaccharides used to control pathogenic scours of all kinds in livestock caused by salmonella, and E. coli etc (26,27). Mannan-oligosaccharides are thought to block the attachment of pathogenic bacteria to the animal's intestine and colonization that may result in disease, while acting as a nutrient to other beneficial bacteria. It is also thought to stimulate the animal's immune system, thereby further reducing the risk of disease (27)**.**. Oyofo et al. (28), observed that the adherence of *Salmonella typhimurium* to enterocytes of the small intestine of chicks, in vitro, was inhibited in the presence of mannose. Later, they found that inclusion of mannose in the drinking water of chicks reduced *S. typhimurium* colonization of the cecum (29,30). Furthermore, feeding MOS to chickens improved the morphology of the small intestine, as evidenced by increased Goblet cell numbers, reduced crypt depth, and greater villus width (31).

In our study, it is evident that addition of *Saccharomyces cerevisiae* at a rate of 1.5, 2 and 2.5 was responsible for a significant (P<0.05) increase of totalserum protein and glucose levels, compared with control group and groups fed low level of *Saccharomyces cerevisiae* 0.5 and 1%. In the contrary a very interesting and a healthy to the consumer, reduction in levels of both serum triglycerides and cholesterol. These beneficial effects of *Saccharomyces cerevisiae* Probiotic supplementation has been shown to reduce the cholesterol concentration were reported in egg yolk by (32) and serum in chicken (33,34). Recent report suggested that feeding of chicory beta fructans – an oligosaccharide, a prebiotic, reduced the serum cholesterol and abdominal fat of broiler chicken (35). Oral administration of probiotics has been shown to significantly reduce cholesterol levels by as much as 22 to 33% (36) or prevent elevated cholesterol levels in mice fed a fat-enriched diet (37). other possible mechanisms include assimilation of cholesterol by the bacteria, binding of cholesterol to the bacterial cell walls of *Lactobacillus*, (38). Gilliland *et al.* (39) suggested that the Prebiotic supplementation could have enhanced the Lactobacilli count. Similar results have been reported by others (33,40) and a similar hypocholesterolaemic effect was observed in broiler chicken supplemented with beta fructans from chicory as a source of Prebiotic (35). MOS is also considered as substrate for lactic acid producing bacteria like *Lactobacillus spp.* and *Bifidobacterium bifidum* (41). Increasing level of MOS also increase the CFU of this lactic acid producing bacteria (42). Gilliland *et al*. (39) hypothesized that some *Lactobacillus spp.* are able to incorporate cholesterol into the cellular membrane of the organism, thus, cholesterol assimilation by *Lactobacillus* in turn reduce cholesterol absorption in the system. The lower level of serum triglyceride might be due to increased level of lactic acid producing bacteria in the gut of broiler chicken. The results of Santoso *et al.* (43) reported that supplementation of *B.Subtilis* in broiler diets decreased triglycerides in the serum.

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