

Full Length Research Paper

Effects of dietary probiotic, prebiotic and butyric acid glycerides on performance and serum composition in broiler chickens

K. Taherpour¹, H. Moravej^{1*}, M. Shivazad¹, M. Adibmoradi² and B. Yakhchali³

¹Department of Animal Science, Faculty of Agriculture, University of Tehran, Karaj, Iran.

²Department of Basic science, Faculty of veterinary medicine, University of Tehran, Tehran, Iran.

³National Institutes for Genetic Engineering and Biotechnology (NIGEB). Tehran, Iran.

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An experiment on ROSS 308 male broilers was conducted to evaluate probiotic (Primalac), prebiotic (Fermacto) and butyric acid glycerides (Baby C₄) on broiler performance and serum composition. Seven hundred and four day-old broilers were randomly distributed in a 2×2×2 factorial arrangement with two levels of probiotic, prebiotic and butyric acid glycerides. Eight treatments with four replicates each with 22 birds per pen were used. Three-way interaction between dietary treatments were observed for final body weight (P<0.05), feed intake (P<0.01) and feed conversion ratio (P<0.01) in the experiment. Body weight, feed intake and feed conversion ratio between supplementary treatments and control group were significantly different (P<0.01). Three-way interaction between dietary treatments were observed for total cholesterol (P<0.05), LDL (P<0.01), cholesterol/HDL ratio and HDL/LDL ratio (P<0.01) in the experiment. Total cholesterol concentration, LDL, HDL/LDL ratio and cholesterol/HDL ratio between supplementary treatments and control group were significantly different (P<0.05 and P<0.01). Serum triglyceride, HDL and VLDL cholesterol concentrations were not significantly different among dietary treatment when compared to control group (P>0.05). In conclusion, dietary supplementation improved the body weight, feed conversion ratio, HDL/LDL ratio and decreased feed intake, total cholesterol, LDL cholesterol and cholesterol/HDL ratio in the serum of broiler chickens.

Key words: Probiotic, prebiotic, butyric acid glycerides, serum cholesterol, broiler.

INTRODUCTION

Due to growing concerns about antibiotic resistance and the potential for a ban for antibiotic growth promoters in many countries, there is an increasing interest in finding alternatives to antibiotics in poultry production (Patterson and Burkholder, 2003; Chichlowski et al., 2007). Some possible alternatives to antibiotics for growth promotion and improvement of feed efficiency in domestic avian species are dietary supplementation of probiotic, prebiotic and organic acids (Jin et al., 1997; Biggs et al., 2007; Xu et al., 2006). Probiotic, which means "for life" in Greek (Gibson and Fuller, 2000), has been defined as a live microbial feed supplement which beneficially affects the host animal by improving its intestinal balance, stimula-

ting synthesis vitamins of the B-group, improving immunity system, preventing harmful microorganism, providing digestive enzymes and increasing of production of volatile fatty acids (Fuller, 1989; Rolf, 2000). In broiler nutrition, probiotics have a beneficial effect on broiler performance (Zulkflia et al., 2000; Kabir et al., 2004; Gil et al., 2005), improvement in feed conversion (Yeo and Kim, 1997), reduction in mortality (Kumprecht, 1998) and depress serum cholesterol (Mohan et al., 1996; Kurtoglu et al., 2004). There are also conflicting studies; probiotic have been shown not to improve weight gain, feed conversion ratio and to reduce mortality (Jin et al., 1997).

Prebiotics are food ingredients that selectively stimulate the activity and/or growth of endogenous bacteria such as Bifidobacteria and Lactobacilli, which benefit the host (Gibson and Roberfroid., 1995). They are short chain carbohydrate that is non-digestible by animal enzyme

*Corresponding author. E-mail: hmoraveg@ut.ac.ir.

(Quigly et al., 1999). Several studies have shown that administering prebiotics can improve weight gain, feed intake and feed conversion in broiler (Waldroup et al., 1993; Grimes et al., 1997; Wu et al., 1999; Xu et al., 2003; Pelicano et al., 2004; Rodrigues et al., 2005). However, some reports indicate that prebiotic supplementation did not effect in body weight gain, feed intake or feed conversion (Stanczuk et al., 2005). Prebiotic supplementation may also depress cholesterol concentration in blood and egg yolk (Mohan et al., 1995; Jin et al., 1998; Li et al., 2007).

Short-chain fatty acids such as butyrate are also considered as potential alternatives to antibiotic growth promoter (Dierick et al., 2002; Lesson et al., 2005; Van Immerseel, et al., 2005). Dietary butyric acid had no effect on body weight or weight gain in starter, grower and finisher periods, but birds consumed less feed intake when diets were supplementation with butyrate relative to the control birds (Lesson, et al., 2005).

There are similar effects of probiotic, prebiotic and butyric acid glycerides on the performance of broiler chickens and there are a commercial varieties of these supplementation available and producers are presented with a challenge to choose the most suitable supplementation to reduce their costs of diet and production. Therefore, the objective of this study was to investigate the interaction effects of supplementation of probiotic (premalac), prebiotic (Fermacto) and butyric acid glycerides (Baby c4) on the performance and blood chemistry of broiler chickens under commercial conditions.

MATERIALS AND METHODS

Experimental animals

Seven hundred and four day-old male broilers chicks of commercial strain (Ross 308) were randomly divided into 8 treatments each composed of 88 birds. Birds in each treatment were placed in 4 pens, each containing 22 birds. All birds were raised on floored pen (2.5 × 1.25 M), in an environmentally controlled house with continuous light (10 to 20 lux) and had access to feed and water *ad libitum*. The house temperature was maintained at 33°C for the first 3 d, after that temperature was gradually reduced by 3°C per week until reaching 24°C; this temperature was maintained until the end of the 42 d of experiment. The trial was conducted in 3 periods consisting of the starter period (1 to 10 day), grower period (11 to 28 day) and finisher period (29 to 42 day).

Experimental design and diets

Birds were distributed in a completely randomized block design with 2×2 factorial arrangement. The composition and nutrient analysis of basal diet are shown in Table 1. The basal diet was a typical corn- soybean diet as mash form that was formulated to meet Ross nutrient requirements for starter grower, and finisher growth periods (Aviagen, 2004). Commercial probiotic (Primalac), Prebiotic (Fermacto), and Butyric acid glycerides (Baby C4) were used according to manufacture's instruction. Probiotic was added to the diet at 0.9 kg/ton in the starter, 0.45 kg/ton in the grower and 0.225 kg/ton in the finisher periods. Prebiotic was added to the diet at 1.8 kg/ton in the starter, and 1 kg/ton until slaughter age (42 d). Butyric acid glycerides

were added to the diet at 3 and 2 kg/ton in the starter and grower periods respectively (1 to 28 d). Thus, the experimental diets consisted of:

Basal diet unsupplemented or control diet (T₁)
 Basal diet + Probiotic (T₂ or Pro)
 Basal diet + Prebiotic (T₃ or Pre)
 Basal diet + butyric acid glycerides (T₄ or But)
 Basal diet + Probiotic + Prebiotic (T₅ or Pro×Pre)
 Basal diet + Probiotic+ butyric acid glycerides (T₆ or Pro× But)
 Basal diet + Prebiotic + butyric acid glycerides (T₇ or Pre× But)
 Basal diet + Probiotic + Prebiotic + butyric acid glycerides (T₈ or Pro× Pre× But)

Performance parameters

Chickens were weighed at 1, 10, 28 and 42 d on Pen basis to determine weight gain. Feed consumed was recorded daily, the uneaten discarded, and feed conversion ratio (FCR) was calculated (total feed : total gain). Mortality was recorded as it occurred and percentage mortality was determined at the end of the study.

Measurement of serum indices

On 42 day of experimental period, 3 ml of blood was collected from brachial vein from one bird of each pen (from four birds of each treatment). Serum was isolated by centrifugation at 3,000×g for 10 min. The serum concentrations of total triglyceride, cholesterol, high-density lipoprotein (HDL) cholesterol, low-density lipoprotein (LDL), cholesterol/ HDL and HDL/ LDL ratio in serum samples were analyzed by an automatic biochemical analyzer (Clima, Ral. Co, Spain), following the instructions of the corresponding reagent kit (Pars Azmon Co., Iran). VLDL cholesterol was calculated from triglycerides by divided the factor 5 (Arun et al., 2006). The LDL cholesterol was calculated by using the formula: LDL cholesterol = Total cholesterol - HDL cholesterol - VLDL cholesterol.

Statistical analysis

All data were analyzed by ANOVA using the SAS (SAS Institute, 2000) GLM program. Treatment means were portioned by LSMEAN analysis. The model is:

$$Y_{ijkl} = \mu + A_i + B_j + C_k + (AB)_{ij} + (AC)_{ik} + (BC)_{jk} + (ABC)_{ijk} + Block_L + e_{ijkl}$$

Where Y is the observed response, μ is the overall mean, A_i is the effect of Probiotic, B_j is the effect of Prebiotic, C_k is the effect of butyric acid glycerides AB_{ij}, AC_{ik}, and ABC_{ijk} interaction between the effect of two or three factors, block_L is the effect of block e_{ijkl} and is the error.

RESULTS AND DISCUSSION

The final body weight, feed intake and feed conversion ratio of chicks is shown in Table 2. Three-way interaction between dietary treatments were observed for body weight (P<0.05), feed intake (P<0.01) and feed conversion ratio (P<0.01) in the experiment. The result showed that all the treatments except the treatment containing only prebiotic have better final body weight in compare with control treatment. These results are in agreement with those of Runho et al. (1997), Jin et al. (1998) and

Table 1. Composition of the basal diet fed to broilers.

Ingredient and Composition	Starter (0-10d)	Grower (11-28d)	Finisher (29-42d)
Ingredient % as fed			
Corn	49.03	59.6	65.99
Wheat	5.58	5.00	5.00
Soybean meal (44% CP)	26.86	16.05	10.12
Gluten	10.00	11.48	11.50
Soybean oil	3.50	3.34	3.09
Limestone	1.45	1.23	1
Dicalcium phosphate	1.95	1.8	1.83
Salt (NaCl)	0.36	0.36	0.36
Vitamin Permixon ¹	0.25	0.25	0.25
Mineral Permixon ²	0.25	0.25	0.25
L-Lysine	0.52	0.58	0.57
DL-Methionine	0.25	0.06	0.04
Nutritional content (calculated)			
ME, Kcal/kg	3010	3150	3200
CP	23.00	20.00	18.00
Calcium	1.00	0.90	0.90
Available phosphorus	0.50	0.45	0.45
Lys	1.41	1.16	1.05
Met and Cystine	1.09	0.81	0.78

vitamin premix the following per kilogram of complete feed: Vitamin A, 4,500 IU (retinyl acetate); cholecalciferol, 1000 IU; vitamin E, 25 IU; vitamin B₁₂, 0.02 mg; menadione, 1.5 mg; riboflavin, 3 mg; thiamin 1.5 mg; pantothenic acid, 5 mg; niacin, 20 mg; choline, 150 mg; folic acid, 0.5 mg; biotin, 0.5 mg; and pyridoxine, 2.5 mg.

The mineral premix supplied the following per kilogram of complete feed: Manganese, 60 g; zinc, 40 mg; iron, 80 mg; copper, 8 mg; selenium, 0.2 mg; iodine, 0.8 mg; and cobalt, 0.4 mg.

Table 2. Effect of dietary treatment on the performance of broiler chickens at 42 d age.

Treatment	Levels			Body weight	Feed intake	Feed conversion ratio
	Pro	Pre	But			
Pro ^x Pre ^x But	0	0	0	1910.01 ^b	3824.90 ^a	2.00 ^a
	1	0	0	2037.89 ^a	3464.12 ^{bc}	1.70 ^b
	0	1	0	1971.61 ^{ab}	3386.38 ^{bc}	1.72 ^b
	0	0	1	2063.83 ^a	3404.92 ^{bc}	1.65 ^b
	1	1	0	2037.80 ^a	3506.53 ^{bc}	1.72 ^b
	1	0	1	2009.10 ^a	3345.11 ^c	1.66 ^b
	0	1	1	2027.26 ^a	3595.50 ^b	1.77 ^b
	1	1	1	2015.30 ^a	3361.92 ^{bc}	1.68 ^b
	SE			31.35	71.6	0.03
P- value			0.05	0.004	0.0003	

^{a-c} Means with in columns with different superscript differ significantly (P<0.05; P<0.001). ¹PRO= Probiotic; PRE= Prebiotic; and BUT= Butyric acid glycerides.

Biggs et al. (2007) and on the contrary Leeson et al. (2005); Chichlowski et al. (2007) and Mount et al. (2007). Inconsistent results of using probiotic, prebiotic and organic acid may be due to differences in methods of preparing the supplement, different in dietary nutrient levels, different experimental condition or different sex of

birds. Our research demonstrated that dietary treatments decreased feed intake significantly compared with control diet (P<0.05; P<0.01) and the treatments containing probiotic + butyric acid glycerides and Probiotic + Prebiotic have the lowest and highest feed intake among dietary treatments, respectively. Several studies con-

Table 3. Effect of dietary treatment on the blood chemical parameters of broiler chicken at 42 d age (Mg/dL).

Treatment	Levels			Total Cholesterol	HDL	LDL	VLDL	Chol./HDL	HDL/LDL	Triglycerides
	Pro	Pre	But							
But×Pre×Pro	0	0	0	170.50 ^a	56.12	77.13 ^a	37.25	3.19 ^a	0.75 ^c	151.25
	1	0	0	128.00 ^b	72.21	30.04 ^{bc}	25.75	1.77 ^b	2.47 ^{bc}	128.75
	0	1	0	121.25 ^b	58.52	35.93 ^b	26.80	2.08 ^b	2.2 ^{bc}	134.00
	0	0	1	141.50 ^b	68.13	43.47 ^b	29.90	2.08 ^b	1.59 ^c	149.5
	1	1	0	92.00 ^c	62.13	9.53 ^d	20.35	1.49 ^b	6.72 ^a	101.75
	1	0	1	125.00 ^b	63.09	36.81 ^b	25.10	2.01 ^b	2.25 ^{bc}	125.50
	0	1	1	100.25 ^c	58.50	17.40 ^{cd}	24.35	1.75 ^b	3.49 ^b	121.75
	1	1	1	136.00 ^b	70.21	43.20 ^b	22.60	1.96 ^b	1.78 ^{bc}	113.00
	P-value			0.001	0.19	0.001	0.096	0.001	0.001	0.65
	SE			6.93	4.72	6.02	3.65	0.19	0.55	19.40

^{a-d}Means with in columns with different superscript differ significantly (P<0.05; P<0.001).

¹ PRO= Probiotic; PRE= Prebiotic; BUT= Butyric acids glycerides.

ducted to evaluate the feeding of probiotic and butyric acid showed that feed intake decreased in broiler chickens (Zulkifli et al., 2000; Lesson, et al., 2005). Feed conversion ratio was improved significantly for the broilers fed all dietary treatments compared with control diet (P<0.05; P<0.01). Numerous studies showed that feed conversion improved with addition of probiotic, prebiotic and organic acid in broiler diet (Yeo and Kim, 1997; Gil et al., 2005; Rodrigues et al., 2005). Probiotic, prebiotic and organic acids maintained a better microbial environment in digestive tract of birds by reducing the number of pathogenic microbes. This enhanced digestion, absorption and efficiency of utilization of feed (Khaksefidi and Ghoorchi, 2006; Chicholowski and et al., 2007).

The other results of our study demonstrated there was non-significant decrease in feed intake and feed conversion ratio by prebiotic treatment compared with control diet. This results in agreement with those of several studies noted that addition of prebiotic to broiler diet had no significant influence on feed intake and feed conversion (Sohail et al., 2002; Chicholowski et al., 2007; Mountzouris et al., 2007). Mortality for all groups was within the expected range and there was no significant difference in mortality of all treatments. The serum concentrations of triglycerides, total cholesterol, HDL, LDL and VLDL cholesterol, cholesterol/HDL and HDL/LDL ratio, in dietary treatments at the end of the experiment (42d) are presented in Table 3. Three-way interaction between dietary treatments were observed for total cholesterol (P<0.05), LDL (P<0.01), cholesterol/HDL and HDL/LDL ratio (P<0.01). There were not any significant differences on serum triglyceride, HDL and VLDL concentrations of birds fed with all treatments. The serum total cholesterol concentrations were significantly reduced by dietary treatments compared to the control group (P<0.05 or P<0.01), and the treatment containing pro × pre and pre × but showed the lowest cholesterol among all dietary treatments (P<0.05). There was no significant difference on serum HDL cholesterol

concentrations of birds fed dietary treatments compared to the control group (P>0.05). Serum LDL cholesterol concentrations were significantly reduced by dietary treatments compared to the control group (P<0.05 or P<0.01) and the treatment containing pro × pre and pre × but showed the lowest LDL among all dietary treatments (P<0.05). Serum VLDL cholesterol concentrations were not significantly reduced by dietary treatments when compared to control group (P>0.05). Similar cholesterol depressing effect due to probiotic supplementation in broiler chicken was observed by Joy and Samuel (1997). In addition, in some study, probiotic supplementation reduced the serum LDL cholesterol (kalavathy et al., 2003) and triglycerides (Santos et al., 1995) in broiler chicken. The finding of our study and previous studies indicated that feeding of probiotic has a cholesterol depressing effect in broiler chicken. Besides, it is reported that some of the microorganisms present in the probiotic preparation could utilize the cholesterol present in the gastro intestinal tract for their own metabolism, thus, reduce to absorption the amount cholesterol (Nelson and Gilland, 1984; mohan et al., 1995). Lactobacillus, which has a high bile salt hydrolytic activity, is responsible for deconjugation of bile salts (Saron, 2003). Deconjugated bile acid are less soluble at low pH and less absorb in the intestine and is more likely to be excreted in feces (Klaver and van der meer, 1993). In addition, probiotic microorganism inhibits hydroxymethyl- glutaryl- coenzyme A, an enzyme involved in the cholesterol synthesis (Fukushima and Nakon, 1995). The most important mechanism by which prebiotic eliminates cholesterol would likely be through reducing lipid absorption in intestine by binding bile acids, which results in increased cholesterol elimination and hepatic synthesis of new bile acid (Zhang et al., 2003).

On the other hand, Salma et al. (2007) have shown that cholesterol concentration in thigh and breast muscle of the broilers had a positive correlation with the change of the cholesterol contents in serum. Thus, it is expected

that with decreasing of serum cholesterol, the amount of meat cholesterol is tending to decrease too. In view of dietary health, food that contains these supplementations can help in reducing the occurrence of cardiovascular heart diseases. Therefore, these results led to the conclusion that there were similar improvements on performance of birds fed diets with single supplementation of probiotic, prebiotic and butyric acid glycerides as well as their combination, and their performance was improved better than birds fed control diet, and choice preference of supplementation should be based on its economic value. In addition to, it is possible that all supplementation diets can help in reducing the occurrence of cardiovascular heart diseases in consumers.

REFERENCES

- Arun KP, Savaram VR, Mantena VLNR, Sharma, SR (2006). Dietary supplementation of lactobacillus Sporogenes on performance and serum biochemical- lipid profile of broiler chickens. The J. Poult. Sci. 43: 235-240.
- Aviagen (2004). Ross broiler performance objective guides. www.ross.intl. Aviagen.com/ include. Asp?sec=371& con=371.
- Biggs P, Parsons CM, Fahey GC (2007). The effects of several oligosaccharides on growth performance, nutrient digestibilities, and cecal microbial populations in young chicks. Poult. Sci. 86: 2327-2336.
- Chichlowski M, Croom J, McBride BW, Daniel L, Davis G, Kaci MD (2007). Direct-fed microbial primals and Salinomycin modulate whole-body and intestinal oxygen consumption and intestinal mucosal cytokine production in the broiler chick. Poult. Sci. 86: 1100-1106.
- Dierick NA, Decuypere JA, Molly K, Vanbeek E, Evanderbeke (2002). The combined use of triacylglycerols containing medium-chain fatty acids and exogenous lipolytic enzymes as an alternative for nutritional antibiotics in piglet nutrition. Livest. Prod. Sci. 75:129-142.
- Fukushima M, Nakano M (1995). The effect of probiotic on faecal and liver lipid classes in rats. Br. J. Nutr., 73: 701-710.
- Fuller R (1989). Probiotics in man and animals. J. Appl. Bacteriol. 66: 365-378.
- Gibson GR, Roberfroid MB (1995). Dietary modulation of the human colonic Microbiota- Introducing the concept of prebiotics. J. Nutr. 125: 1401-1412.
- Gil De Los Santos JR, Storch OB, Gil-Turnes C (2005) Bacillus Cereus Var. Toyoi and Saccharomyces boulardii increased feed efficiency in broilers infected with Salmonella Enteritidis. Br. Poult. Sci. 46: 494-497.
- Grimes JL, Maurice DV, Lightseg SF, Lopez JG, (1997). The effect of dietary fermento on laying hen Performance. J. Appl. Poult. Res. 6: 399-403.
- Jin LZ, Ho YW, Abdullah N, Jalaludin S (1998). Growth Performance, intestinal microbial population and serum cholesterol of broilers fed diets containing Lactobacillus Cultures. Poult. Sci. 77: 1259-1265.
- Jin LZ, Ho YW, Abdullah N, Jalaludin S (1997). Probiotics in poultry: Mode of action. Worlds Poult. Sci. J. 53:351-368.
- Joy AD, Samuel JJ (1997). Effect of Probiotic supplementation on the performance of broilers. J. Vet. Anim. Sci. 28-10-14.
- Kabir SML, Rahman MM, Rahman Mb, Ahmed SU (2004). The dynamic of probiotics on growth performance and immune response in broiler. Int. J. Poult. Sci. 3: 361-364.
- Kalavathy R, Abdullah N, Jalaludin S, YWH (2003). Effect of lactobacillus cultures on growth performance, abdominal fat deposition, serum lipid and weight of organs of broiler chickens. Br. Poult. Sci., 44: 139-144.
- Khaksefidi A, Ghoorchi T (2006). Effect of probiotic on performance and immunocompetence in broiler chicks. J. Poult. Sci., 43:296-300.
- Klaver FAM, Van der Meer R (1993). The assumed assimilation of cholesterol by lactobacillus and Bifidobacterium bifidum is due to their bile salt deconjugating activity. Applied Environ. Microbiol. 59: 1120-1124.
- Kumprecht I, Zobac P (1998). The effect of probiotic preparation containing Saccharomyces Cerevisiae and Enterococcus Faecium in diet with different level of B- vitamins on chicken broiler performance. Zivocisna Vgropa 43: 63-70.
- Kurtoglu V, Kurtoglu F, Seker E, Coskum B, Balevi T, Polat ES (2004). Effect of probiotic supplementation on laying hen diets on yield performance and serum and egg yolk cholesterol. Food Addit. Contam. 21: (9): 817-823.
- Leeson S, Namkung H, Antongiovanni M, Lee EH (2005). Effect of butyric acid on the performance and carcass yield of broiler chickens. Poult. Sci. 84: 1418-1422.
- Li X, Liu L, Li K, Hao K, Xu C (2007). Effect of fructooligosaccharides and antibiotics on laying performance of chickens and cholesterol content of egg yolk. Br. Poult. Sci. 48(2): 185-189.
- Mohan B, Kadirvel R, Bhaskaran M, Natarajan A (1995). Effect of probiotic supplementation on serum/ yolk cholesterol and on egg shell thickness in layers. Br. Poult. Sci. 36: 779-803.
- Mohan B, Kadirvel R, Bhaskaran M, Natra Jan A (1996). Effect of probiotic supplementation on growth, nitrogen utilization and serum cholesterol in broilers. Br. Poult. Sci. 37: 395-401.
- Mountzouris KC, Tsirtsikos P, Kalamara E, Nitsch S, schatzmayr G, Flegeros K (2007). Evaluation of the efficacy of a Probiotic containing Lactobacillus, Bifidobacterium, Enterocococcus, and Pediococcus strains in promoting broiler performance and modulating cecal microflora composition and metabolic activities. Poult. Sci. 86: 309-317.
- Nelson CR, Gilliland SE (1984). Cholesterol uptake by lactobacillus acidophilus. J. Dairy Sci. p: 67 (Supplement).
- Patterson JA, Burkholder K (2003). Application of prebiotics and probiotics in poultry production. Poult. Sci. 82:627-631.
- Pelicano ERL, de Souza PA, de Souza HBA, Leonel FR, Zeola NMBL, Boiago MM (2004). Productive traits of broiler chickens fed diet containing different growth promoters. Rev. Bras. Zootec. 31: 177-182.
- Quigly ME, Hudson GJ, Englst HN (1999.) Determination of resistant short-chain carbohydrates (non-digestible oligosaccharides) using gas- liquid chromatography. Food. Chem. 65: 381-390.
- Rodrigues TA, Sartor C, Higgins SE, Wolfenden AD, Bielke LR, Pixley CM, Sutton L, Tellez G, Hargis M (2005). Effect of Aspergillus meal prebiotic (fermacto) on performance of broiler chickens in the starter phase and fed low protein diets. J. Appl. Poult. Res. 14: 665-669.
- Rolf RE (2000). The role of probiotic cultures in the control of gastrointestinal health. J. Nutr. 130: 396-402.
- Runho RC, Sakomura NK, Kuana S, Banzatto D, Junoqueria OM, Stringhini JH (1997) Use do acid organic (acid Furmico) has racoes de frangos de corte, Revista Brasileira de Zootecnia, 26: 1183-1191.
- Salma U, Miha AG, Make T, Nishimura M, Tsujii H (2007). Effect of dietary Rhodobacter Capsulatus on cholesterol concentration and fatty acid composition in broiler meat. Poult. Sci. 86: 1920-1926.
- Santos U, Tanaka K, Ohtani S (1995). Effect of dried Bacillus subtilis culture on growth, body composition and hepatic lipogenic enzyme activity in female broiler chicks. Br. J. Nutr., 74: 523-529.
- SAS Institute (1999-2000). SAS user's Guide. Release 8.2 Ed. SAS institute Inc., Cary, Nc.
- Sohail SS, Bryant MM, Voitle RA, Roland DA (2002). Influence of calsporin on commercial leghorns. J. App. Poult. Res., 11: 379-387.
- Stanczuk J, Zdunczyk Z, Juskiwicz J, Jankowski J (2005). Indices of response of young turkeys to diets containing mannanoligosaccharide or inulin. Vet. Zootech. 31: 98-101.
- Sarono S (2003). In vitro probiotic preparation of indigenous dadih lactic acid bacteria. Asian- Australian J. Ani. Sci., 16: 726-731.
- Van Immerseel F, Boyen F, Gantois I, Timbermont L, Bohez L, Pasmans F, Haesebrouck F, Ducatelle R (2005). Supplementation of coated butyric acid in the feed reduces colonization and shedding of salmonella in poultry. Poult. Sci. 84: 1851-1856.
- Waldroup AL, Skinner JT, Hierholzer RE, Waldroup PW (1993). An evaluation of fructooligosaccharide in diets for broiler chickens and effects on salmonella contamination of carcasses. Poult. Sci. 72: 643-650.
- Wu TX, Dai XJ, Wu LY (1999). Effects of fructooligosaccharide on the

- broiler production. *Acta Agric. Zhejiangensis* 11:85-87.
- Xu C-L, Ji C, Ma Q, Hao K, Jin ZY, Li K (2006). Effect of a dried bacillus culture on egg quality. *Poult. Sci.* 85: 364-368.
- Yeo J, Kim K (1997). Effect of feeding diets containing an antibiotic, a Probiotic, or yucca extract on growth and intestinal urease activity in broiler chicks. *Poult. Sci.* 76: 381-385.
- Zhang WF, Li DF, Lu WQ, Yi GF (2003). Effect of isomalto-oligosaccharides on broiler performance and intestinal micro flora. *Poult. Sci.* 82: 657-663.
- Zulkifli I, Abdullah N, Azrin NM, Ho YW (2000). Growth performance and immune response of two commercial broiler strains fed diets containing *Lactobacillus* cultures and oxytetracycline under heat stress conditions. *Br. Poult. Sci.* 41: 593-597.