



Effect of supplementation of probiotics and prebiotics on growth, lipid profile and slaughter traits of coloured broilers during heat stress

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ABSTRACT

Supplementation of probiotics and/or prebiotics to coloured broilers during summer on growth, serum lipid profile and carcass traits were studied. Day-old coloured broiler chicks (180) were randomly distributed into 4 groups with 3 replicates having 15 chicks in each. The 4 dietary treatments such as T₁ -control group fed with basal diet, T₂ - basal diet+ probiotic @ 10 g/q feed, T₃-basal diet + prebiotic (MOS) @ 4,000 ppm and T₄- basal diet+ probiotic @ 10 g/q and prebiotic @ 4,000 ppm. The supplementation was continued from 0 to 56 days of age during natural peak summer period. Significantly better growth rate and FCR was observed in the group supplemented with both pro- and pre-biotics than control but no difference of feed intake was observed. There was significant reduction of lipid profile like triglycerides, LDL and VLDL levels of all the supplemented groups. No difference of any of the carcass traits was observed. It is concluded that the feeding of both probiotics and prebiotics in combination to the coloured broilers during summer improved growth performance and decreased serum triglycerides levels.

Key words: Broiler, Growth, Heat stress, Lipid profile, Prebiotics, Probiotics

With the ban on sub therapeutic antibiotic usage in most countries, there is increasing interest in finding alternatives to antibiotics for poultry production. Probiotics and prebiotics are two of several approaches that have potential to reduce enteric diseases in poultry, increases growth and quality of the poultry products. Combination of probiotics and prebiotics is known as synbiotics where the efficacy of probiotics is enhanced by the inclusion of the specific prebiotic. The studies on supplementation of probiotics and prebiotics in diet of chicken mostly focused on growth, feed efficiency (Semaskaite *et al.* 2006, Vicente *et al.* 2007), reduction of serum cholesterol (Jin *et al.* 1998, Kurtoglu *et al.* 2004, Banday and Pampori 2005 and Panda *et al.* 2006) at different levels of supplementation and under different managerial and environmental conditions. However the results obtained have been inconsistent and variable. Hence the present study was undertaken to investigate the influence of probiotic, prebiotic alone and combination on the growth

performance, serum lipid profile and carcass characteristics of the coloured broilers under hot and humid condition.

MATERIALS AND METHODS

Birds, diets and management: Day-old coloured broiler chicks (180) were randomly distributed into 4 groups with 3 replicates having 15 chicks in each. The control group (T₁) was fed with basal diet without any supplementation; T₂ - basal diet+ probiotic @ 10 g/q feed; T₃-basal diet + prebiotic manno oligo saccharide (MOS), @ 4,000 ppm and T₄- basal diet+ probiotic @ 10 g/q and prebiotic @ 4,000 ppm. The supplementation was continued from 0 day to 8 weeks of age which was started from the 15 April and continued to 11 June which is the summer of this region. A commercial multi-species probiotic preparation, containing multi-species of microbes of bacterial, fungal and yeast origin and the prebiotic manno oligo saccharide (MOS) extracted from cell wall of *Saccharomyces cerevisiae* were used. Chicks of each replicate were assigned to separate cage of linear battery brooder. The chicks were fed starter ration up to 21 days and finisher ration from 22 to 56 days of age as per the BIS (1992) recommendations. Antibiotics and coccidiostats were not added to feed in order to avoid interference. Feed and water was provided *ad lib*. Daily feed given was weighed and at the weekend individual body weight and feed residue of each replicate was recorded. The chicks were vaccinated against New Castle disease with NewCastle disease vaccine LaSota strain on 7 and 21 days of age.

Temperature and humidity: The maximum and minimum

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temperature during the 8 weeks experimental period ranged from 35.81 to 39.36°C (average 36.15°C) and 24.51 to 26.85°C (average 25.92°C), respectively. The maximum and minimum relative humidity during the experimental period ranged from 88.4 to 92.5% (average 90.98%) and 50.9 to 61.9% (average 56.47%), respectively.

Sample collection and analytical procedure: Samples of experimental feeds were analyzed for proximate principles (AOAC 2005). Weekly group replicate average body weight, feed consumption and FCR was calculated. Blood samples were collected from 12 birds of each treatment at the end of the experiment (8 weeks) without addition of anticoagulant and serum collected from them were utilized for analysis of total protein, triglyceride, cholesterol, LDL, HDL and VIDL cholesterol in spectrophotometer by using kit. Similarly at the end of experiment 6 birds from each treatment (1 male and 1 female from each replicate) were taken for estimation of the carcass characteristics like dressed weight, giblet weight, eviscerated weight and abdominal fat% in the department of Livestock products technology.

Statistical analysis: The data obtained from the study were statistically analyzed according to Snedecor and Cochran (1994). The data were also analyzed for analysis of variance (ANOVA) and DMR test (Duncan 1955) was used to test the difference between treatment means wherever necessary.

RESULTS AND DISCUSSION

Body weight, feed intake and FCR: No significant effect on body weight gain was observed up to 4 weeks in any of the treatment but during 5–8th week T₄ has higher body weight than control (Table 1). Higher total body weight was recorded in both T₄ and T₂ groups than control and T₃ at the end of the experiment. Significantly better FCR was observed in the group supplemented with both pro and prebiotics than control but no difference of feed intake was observed. The mortality that occurred during second and third week of rearing is very less and the post mortem findings showed some signs of heat stroke which was non significant among treatments.

The significant improvement of the body weight of the birds supplemented with probiotic + prebiotics during the

later phase in the present study might be attributed to a better microbial environment in the gut and higher enzymatic activity which in turn have enhanced digestion, absorption and utilization of feed (Panda *et al.* 2000, Shilpa *et al.* 2007, Apata 2008). The average maximum temperature and humidity during the period of study was 36.15°C and 90.98% respectively. Under heat stress conditions (36±1°C) Zulkifli *et al.* (2000) also found improvement in body weight, weight gain, higher feed intake and lower feed efficiency in the commercial broiler in comparison under normal temperature by feeding *Lactobacillus* culture but antibiotic did not prove to be beneficial in comparison to probiotic in this heat stress condition. No significant increase in body weight of broilers by supplementation of prebiotic (MOS) was observed, which is in agreement with the reports of Elangovan *et al.* (2005), Albino *et al.* (2006) and McCann *et al.* (2006). The feed conversion efficiency of the birds fed different strains of probiotics and /or with antibiotic and prebiotic was found to be better as described by Goh and Hwang (1999), Kalavathy *et al.* (2003), Flemming (2005) and Freitast (2005) Vicente *et al.* (2007) and Apata (2008). In the present experiment the FCR was little higher side with respect to others findings, because of the nature of the breed (coloured bird) and hot and humid condition during the experimental period. Zulkifli *et al.* (2000) reported less body weight gain and poor feed efficiency of the Hubbard broilers in heat stress conditions.

Lipid profile: The cholesterol levels (Table 2) in supplemented groups (T₂, T₃ and T₄) were significantly (P<0.01) lower than the control (T₁) though the decrease in level was maximum in T₄ followed by T₃ and in T₂ group. Triglycerides are fatty acid esters of glycerol. The free fatty acids are utilized for esterification with glycerol in liver and adipose tissue to contribute triglyceride level of blood. Liver synthesizes triglyceride for VLDL and its transport where as tissue triglyceride is meant for storage. All the birds of supplemented groups had significantly (P<0.01) lower level of triglycerides. The level of serum triglyceride in T₃ group was lower than the T₂ but not significantly different where as T₄ groups showed significant decreased triglycerides level than both T₂ and T₃.

Supplementation of probiotic and /or prebiotic decreased

Table 1. Body weight, feed consumption and FCR of birds supplemented with probiotics and/or prebiotics

Groups	Body weight (g)			Cumulative feed consumption (g)			FCR		
	0–4 wk	5–8 wk	0–8 wk	0–4 wk	5–8 wk	0–8 wk	0–4 wk	5–8 wk	0–8 wk
T ₁	506.57 ±6.73	1068.41 ^b ±13.18	1574.37 ^b ±15.49	1156.11 ±11.26	3043.61 ±37.38	4199.72 ±64.50	2.29 ±0.05	2.85 ^a ±0.06	2.67 ^a ±0.03
T ₂	521.76 ±6.16	1134.67 ^{ab} ±15.87	1656.43 ^a ±17.43	1158.05 ±17.67	3176.94 ±41.08	4335 ±72.60	2.23 ±0.03	2.79 ^{ab} ±0.05	2.62 ^a ±0.05
T ₃	518.36 ±7.20	1105.77 ^{ab} ±14.88	1624.13 ^b ±16.53	1170.27 ±15.98	3075 ±38.38	4245.28 ±71.20	2.28 ±0.06	2.78 ^{ab} ±0.07	2.61 ^a ±0.04
T ₄	526.14 ±8.54	1159.32 ^a ±15.75	1685.46 ^a ±18.34	1160.69 ±14.78	3006.11 ±40.87	4166.8 ±69.20	2.21 ±0.01	2.59 ^b 0.03	2.47 ^b ±0.02

Mean with same superscript within a column does not differ statistically (P<0.01).

the HDL cholesterol in the birds but the decrease was statistically nonsignificant. The calculated concentration of VLDL cholesterol were found 1/5th of triglycerides concentration, the birds in all the groups exhibited similar trend as that of triglycerides. So all the treated groups T₂, T₃ and T₄ showed significantly ($P < 0.01$) lower VLDL cholesterol than that of control (T₁). Whereas the group T₄ which was supplemented with both probiotics and prebiotics showed significantly lower VLDL than either of the supplementation.

All the supplemented groups (T₂, T₃, T₄) had significantly ($P < 0.01$) decreased level of LDL cholesterol from that of control birds and the decrease percent was highest by 32.03% in group T₄ followed by 30.21% in T₃ and by 29.46 in T₂ than control. The LDL cholesterol in T₃ and T₄ were significantly lower than the control (T₁) as well as probiotic (T₂) fed groups.

The cholesterol, HDL and LDL of control group were agreed to the reported ranges (Jin *et al.* 1998, Kannan *et al.* 2005, Banday *et al.* 2005, Panda *et al.* 2006, Musa *et al.* 2007). However, the VLDL and triglycerides content of control birds were higher than the earlier findings (Kannan *et al.* 2005, Panda *et al.* 2006, Musa *et al.* 2007). The variation in normal total cholesterol level may be attributed to the difference in age / fat content of diet / breed characteristics. Owing to breed characteristics, increase utilization of fatty acids for esterification in liver leads to higher triglycerides level in blood, which in turn compel the birds to synthesize higher VLDL for transport of triglycerides to tissue.

Supplementation of probiotic and prebiotic separately and in combination resulted significant decrease in serum cholesterol level with a higher decrease (13.10%) in probiotic+prebiotic (T₄) group. In the experiment probiotic supplementation reduced total cholesterol by 10.21%, which is in good agreement with the reports of Mott *et al.* (1973) in germ free pigs, Grunewald (1982) in rats, Mohan *et al.* (1996), Jin *et al.* (1998), Kalavathy *et al.* (2003), Kim *et al.* (2003), Kurtoglu *et al.* (2004), Banday *et al.* (2005) and Panda *et al.* (2006) in poultry. Reduced level of cholesterol by 12.10% in prebiotic supplemented group corroborates with the findings of Kannan *et al.* (2005), Gilliland *et al.* (1985) and Li *et al.* (2007). It is attributed to oral supplementation of oligosaccharides and polysaccharides which reduces blood cholesterol through various mechanisms by reducing its absorption and synthesis.

Some of the microorganisms present in probiotic preparations could utilize cholesterol present in gut for their own metabolism (Nelson and Gilliland 1984). *Lactobacillus* species in probiotic reduces intestinal pH there by cholesterol absorption is decreased and more cholesterol is excreted in faeces (Klaver and van der Meer 1993). *Aspergillus oryzae* and *Lactobacillus* increases synthesis of bile salts from cholesterol, thereby decreasing the cholesterol level in blood (Abdulrahim *et al.* 1996, Surono 2003). They also inhibit hydroxy methyl glutaryl coenzyme A, the enzyme required for cholesterol synthesis which adds

to the reason of low cholesterol synthesis (Fukushima and Nakano 1995). Decreased cholesterol level by supplementation of probiotic might be due to the aforesaid reasons in the present study.

Further, prebiotics used in this experiment is a manooligosaccharide, so it might be the reason of decreased cholesterol level in the birds of T₃. In addition prebiotics promote the *Lactobacillus* count of GI tract and thereby increases the cholesterol assimilation by these bacteria to reduce its level in blood. Since prebiotic has got a positive effect on *Lactobacillus* and *Aspergillus* species of probiotics, their increased count reduces cholesterol absorption and increases incorporation in the cell membrane synthesis (Gilliland *et al.* 1985, Kannan *et al.* 2005 and Li *et al.* 2007). However, the highest decrease percent in cholesterol level in T₄ might be due to the synergistic effect of probiotic and prebiotic and due to incorporation of multispecies bacteria, yeast and fungus in probiotic. These species may help in some alternate way to reduce the cholesterol content in GI tract / increased cholesterol utilization by themselves / inhibit cholesterol synthesis by which significant decreased cholesterol was recorded in group T₄ birds.

Oral supplementation of probiotic, prebiotic separately and in combination significantly decreased triglyceride, VLDL and LDL cholesterol in all the treated groups where as HDL cholesterol did not vary significantly among the groups. Our results corroborates with earlier findings (Santoso *et al.* 1995, Kalavathy *et al.* 2003, Kannan *et al.* 2005 and Panda *et al.* 2006). Increased *Lactobacillus* species in gut lowers the pH and thereby absorption and assimilation of fat decreases. Due to low availability of free fatty acids in blood, triglycerides synthesis is decreased which in turn decreases VLDL cholesterol level in serum. As cholesterol contributes 75% to LDL cholesterol, low serum cholesterol level by supplementation of probiotics and prebiotics resulted significantly lower level of LDL cholesterol. In addition a non significant reduction in HDL cholesterol level may be due to inhibition of cholesterol synthesis by probiotics in the tissue which is reflected in the form of low HDL cholesterol in the treated birds.

Slaughter traits: The average eviscerated weight of control (T₁), probiotic (T₂), prebiotic (T₃) and pro+prebiotic (T₄) supplemented groups were observed as 72.16, 71.82, 72.35 and 71.74% of the live weight respectively (Table 3). The values of the different groups did not differ significantly. The average per cent dressed weight of the birds of different groups ranged from 75.46 to 76.58 without significant difference. Also the percent weight of giblet and abdominal fat of the birds of different groups did not differ significantly.

The observed eviscerated weight, dressed weight and giblet weight of the birds was within the range of 68.0 to 72.6%, 67.0 to 88.0% and 4.5 to 6.68%, respectively, as reported earlier (Panda *et al.* 2000, Banday *et al.* 2005, Kannan *et al.* 2005, Takahashi *et al.* 2005, Das *et al.* 2005, Elangovan *et al.* 2005, Shilpa *et al.* 2007), which did not

Table 2. Lipid profile of birds supplemented with probiotics and/or prebiotics

Treatments	Serum total cholesterol (mg/dl)	Serum triglyceride (mg/dl)	HDL cholesterol (mg/dl)	VLDL* cholesterol (mg/dl)	LDL* cholesterol (mg/dl)
T ₁	125.33 ^a ±1.02	102.91 ^a ±0.85	61.91 ±1.069	20.58 ^a ±0.17	42.83 ^a ±0.65
T ₂	112.25 ^b ±1.04	100.16 ^b ±0.98	60.08 ±0.96	20.03 ^b ±0.19	32.14 ^b ±0.60
T ₃	110.16 ^b ±1.01	98.08 ^b ±0.99	60.66 ±1.49	19.61 ^b ±0.19	29.89 ^c ±1.14
T ₄	108.91 ^b ±1.12	95.25 ^c ±0.96	60.75 ±0.93	19.05 ^c ±0.18	29.11 ^c ±0.64
CD	3.95 ^{**}	2.64 ^{**}	2.18 ^{NS}	0.530 ^{**}	2.20 ^{**}

Means with same superscripts within a column do not differ significantly (P<0.01). * Calculated value.

Table 3. Slaughter traits of birds supplemented with probiotics and/or prebiotics

Treatments	Live wt (g)	Dressed wt. (%)	Giblet wt. (%)	Evs. wt. (%)	Abdominal fat (%)
T ₁	1666.84 ±23.88	76.58 ±3.41	4.65 ±0.10	72.16 ±1.22	3.1 ±0.04
T ₂	1694.65 ±27.66	75.46 ±2.68	4.41 ±0.01	71.82 ±1.09	3.21 ±0.06
T ₃	1669.42 ±22.87	76.14 ±0.86	4.36 ±0.13	72.35 ±0.64	2.99 ±0.04
T ₄	1719.4 ±34.33	76.32 ±0.76	4.54 ±0.09	71.74 ±1.05	3.13 ±0.09
CD	No	7.78	0.37	3.55	0.26

vary significantly in comparison to control groups. The estimated weight of abdominal fat of birds in control group and different treatment groups did not differ significantly, which is in good agreement with other reports (Elangovan *et al.* 2005, Takahashi *et al.* 2005, Bandey *et al.* 2005). But the observed values (2.99 to 3.21%) for all the groups were higher than the reported range of 1.33 to 2.28% of live weight (Elangovan *et al.* 2005 and Kannan *et al.* 2005). This might be due to the age of slaughter of birds at 56 days of age in our experiment with respect to 42 days in others. Also the birds were fed finisher ration containing high energy from fourth week to the end of eighth weeks of age which was reflected as higher serum triglycerides and VLDL cholesterol (Table 2) and abdominal fat. Hermier *et al.* (1989) also reported that the growth of adipose tissue in birds depends directly upon the VLDL triglyceride level.

From the results of the present study, it is concluded that supplementing both probiotic and prebiotic to the coloured broilers during the heat stress condition had beneficial effect on growth performance, lowered serum triglyceride, VLDL and LDL levels. Further study is required to investigate the suitable combination of probiotic and prebiotics for a better synergistic and symbiotic action in the birds.

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