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Gut morphology, immunocompetence, blood and meat quality profile of broiler chickens fed turmeric (*Curcuma longa*) oil as an antibiotic substitute

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ABSTRACT

The present investigation was carried out to study the antibiotic efficacy of turmeric oil in commercial broiler chicken. Day-old broiler chicks (144) were randomly divided into six treatment groups with four replicates of six chicks each. The experimental diets were corn – soya meal basal diet (T_1), basal diet + antibiotic (500 ppm oxytetracycline-T_2), basal diet + 0.025% turmeric oil (T_3), 0.050% turmeric oil (T_4), 0.075% turmeric oil (T_5) and 0.100% turmeric oil (T_6) respectively. The birds were reared under standard managemental practices for 42 days. The levels of serum total protein, albumin, globulin, serum total cholesterol, HDL cholesterol, LDL cholesterol, triglycerides and breast meat cholesterol were not influenced either by the supplementation of antibiotic or by the supplementation of turmeric oil at graded levels. Neither antibiotic nor turmeric oil supplementation had any significant effect on the gut microbial population (total microbes, *Lactobacillus, E. coli* and *Clostridium*) and length of ileal villi of broiler chicken. Turmeric oil supplementation at 0.025 and 0.050% levels led to significant reduction in crypt depth than that of antibiotic supplemented group. Dietary supplementation of turmeric oil at 0.025 and 0.100% levels showed higher profit (₹ 0.32 and 1.03/kg live weight gain respectively) when compared to antibiotic supplemented group. It could therefore be concluded that supplementation of turmeric oil at 0.025% level instead of antibiotic feed additive would produce similar effect on serum lipid profile, meat quality, gut microbial load and intestinal morphology in broiler chicken.

Key words: Antibiotic, Commercial broilers, Gut morphology, Immunocompetence, Turmeric oil

The use of antibiotic growth promoters in poultry industry has been seriously criticized by policy makers and consumers because of the development of microbial resistance to these products and the potential harmful effects on public health. Consequently, in 2006, European Union banned use of antibiotics as feed additives because of its residual effects in animal tissues and subsequently leading to antimicrobial resistance in human beings. This in turn caused major complications related to decreasing animal welfare and increasing economic losses (Eevuri and Putturu 2013). On the other hand, there was increasing public and government pressure in several countries to search for natural alternatives to antibiotics. Hence, prebiotics, probiotics, synbiotics, herbs, spices and essential oils have been investigated as an alternative to antibiotics because of their antibacterial, antioxidant, digestive and metabolic enhancing effects (Durrani *et al.* 2006). Botanical compounds have been shown to be potential alternatives to antibiotics for poultry production (Nasir and Grashorn 2010, Nouzarian *et al.* 2011). Turmeric, a member of the Zingiberaceae family, contains different bioactive compounds, such as curcumin, demethoxycurcumin, bisdemethoxycurcumin and tetrahydrocurcuminoids (Sadeghi *et al.* 2012). The active component, viz. curcuminoids in the form of oil has not been studied much. Hence, an investigation was carried out by including different levels of turmeric oil in the diet to study the gut morphology, blood bio-chemical parameters, immuno-competence and meat quality of broilers.

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MATERIALS AND METHODS

The biological trial was carried out as per the Institutional Animal Ethics Committee's (IAEC) approved schedule (Permission No.: 323/GOReBi/S/02/2016/CPCSEA). The biological experiment was conducted with day-old broiler chicks (144) in the deep litter house of Animal Nutrition Department, Veterinary College and Research Institute, Namakkal. The chicks were wing banded, weighed individually and assigned randomly to twenty four groups of six chicks each and allocated to six dietary treatments with four replicates each in a completely randomized design. Each replicate had even number of male and female chicks. The experimental broiler prestarter, starter and finisher diets were formulated as per the Vencobb standard, 2014. The ingredient and nutrient composition of broiler prestarter,

starter and finisher diets are presented in Table 1. The prestarter, starter and finisher diets were fed to birds from 1 to 14, 15 to 28 and 29 to 42 days of age, respectively.

The birds were housed in deep litter system and reared under standard managemental practices. The chicks were fed *ad lib.* with weighed quantity of experimental diets and had free access to water. At the end of 42 days of age, eight birds from each treatment were randomly selected and slaughtered. Whole blood was collected during slaughter to evaluate the serum biochemical profile. Breast and thigh muscles were collected for cholesterol and meat quality studies and preserved at –18°C until further analysis. The antibody titre against Newcastle disease vaccination in the serum was detected by Haemagglutination inhibition test. From the slaughtered birds, the small intestine was removed and milked out. The intestine samples were incised from jejunum portion and preserved in 10% buffered formalin for morphological studies. The whole small intestinal contents were removed for gut microbial assay. The economics of raising broilers up to 42 days with different levels of turmeric oil supplementation was calculated based on the actual cost of feed per kg weight gain. The data collected on various parameters were statistically analyzed as per the method of Snedecor and Cochran (1989) and the mean of different experimental groups were tested for statistical significance by Duncan's multiple range test (Duncan 1955).

Table 1. Ingredient and nutrient composition (% DM) of broiler diets

Ingredients (%)	Pre-starter (0–14 days)	Starter (15–28 days)	Finisher (29–39 days)
Maize	54.1	57.4	58.3
Soybean meal	38.8	36.5	31.0
Rice bran oil	2.9	3.6	4.9
Calcite	2.2	1.0	1.0
Dicalcium phosphate	1.4	1.0	0.9
Salt	0.36	0.05	0.10
L-Lysine hydrochloride	254	192	164
DL methionine	323	277	500
Additives[#] (g)	460	460	460
Nutrients* (%)			
Crude protein	22.46	21.79	19.57
Crude fibre	2.74	2.65	2.62
Ether extract	6.49	6.43	7.31
Total ash	6.20	6.77	6.40
Nitrogen free extract	52.63	52.44	53.91
Calcium	0.94	0.88	0.85
Total phosphorus	0.64	0.56	0.51
Available phosphorus*	0.45	0.46	0.44
Lysine*	1.25	1.34	1.17
Methionine*	0.61	0.64	0.62
Metabolizable energy (Kcal/kg)*	3000	3067	3189

[#]Additives added 460 g/100 kg containing Vit. AB_2D_3K[1], 20; Vitamin-B complex[2], 20; Coccidiostat[3], 50; Toxin binder[4], 50; Liver stimulant[5], 50; Lysoforte[6], 50; Vit. E[7], 10; Endox dry[8], 10; Choline chloride[9], 100 and Trace minerals[10], 100 g. [1]Supplied per kg of diet: Vit. A, 16,500 IU; Vit. B_2, 10 mg; Vit. D_3, 3,200 IU and Vit. K, 2 mg. [2]Supplied per kg of diet: Thiamin, 4 mg; Pyridoxine, 8 mg; Cyanocobalamine, 40 mcg; Vitamin E, 40 mg; Niacin, 60 mg; Calcium D pantothanate, 40 mg; Folic acid, 4 mg. [3]Coccidiostat added at 0.5 g/kg of feed supplied 125 mg of Salinomycin. [10]Supplied per kg of diet: Mn, 54 mg; Zn, 52 mg; Fe, 20 mg; I, 2 mg; Cu, 2 mg; Co, 1 mg. *Calculated value

RESULTS AND DISCUSSION

Serum profile: Effect of turmeric oil supplementation on serum biochemical profile and breast meat cholesterol is presented in Table 2. The level of serum total protein, albumin and globulin were not influenced either by the supplementation of antibiotic or by the supplementation of turmeric oil at graded levels. This suggests that neither antibiotic nor turmeric oil influences the protein nutrition status or protein metabolism of birds. Similar observations were reported in studies with turmeric powder by Ahmedi (2010), Abou-Elkhair *et al.* (2014) and Fallah and Mirzaei (2016). Supplementation of antibiotic (OTC at 50 ppm level) or turmeric oil did not have any significant effect on serum total cholesterol, HDL cholesterol, LDL cholesterol, triglycerides and breast meat cholesterol when compared to antibiotic free control. Similar observations were also recorded by Namagirilakshmi *et al*. (2010), Mehala and Moorthy (2008) and Abou-Elkhair *et al*. (2014) in broilers fed with turmeric powder.

Meat quality: Effect of turmeric oil supplementation on meat pH, water holding capacity (WHC), cooking loss and shear force value of broiler meat is presented in Table 3. Supplementation of either antibiotic or turmeric oil had no influence on pH of the meat of the treatment birds when compared to control. Similar study was conducted by Daneshyar *et al*. (2011) and Wang *et al*. (2015) using turmeric rhizome extract (TRE) and they also observed that dietary supplementation of TRE had no significant effect on the pH of thigh muscles.

Supplementation of antibiotic or turmeric oil had no significant effect on cooking loss, shear force value and water holding capacity of meat of the treatment birds when compared to control. However, Wang *et al.* (2015) in his study with TRE, observed a significant decrease in drip loss of thigh muscle in a dose-dependent manner indicating that dietary supplementation of TRE could improve the antioxidant capacity of broilers, thereby subsequently maintaining the integrity of the cell membrane and increasing the water holding capacity. Neither antibiotic nor turmeric oil had any significant effect on tyrosine and thiobarbituric acid value of meat of the experimental birds when compared to control. This observation was not in tandem with the findings of Daneshyar *et al.* (2012), who observed lower serum malondialdehyde (MDA) content and mortality in broilers fed with 5 g/kg turmeric rhizome powder.

Gut microbial load: Effect of turmeric oil supplementation on intestinal microbial load of broilers is presented in Table 4. Supplementation of oxytetracycline

Table 2. Effect of turmeric oil supplementation on serum protein, lipid profile and breast meat cholesterol of broilers

Treatment group	Total protein (g/dl)	Albumin (g/dl)	Globulin (g/dl)	Total cholesterol (mg/dl)	HDL (mg/dl)	LDL (mg/dl)	Triglycerides (mg/dl)	Breast meat cholesterol (mg/dl)
T_1 (Control)	5.2±0.16	3.5±0.17	1.7±0.08	131.8±3.98	51.9±1.81	79.8±2.27	133.1±2.15	80.75±2.07
T_2 (Control + OTC @ 50 ppm)	4.9±0.46	3.3±0.51	1.6±0.07	129.7±5.24	49.3±2.30	80.4±4.21	132.3±3.66	81.12±2.01
T_3 (Control + 0.025% turmeric oil)	5.2±0.23	3.5±0.29	1.7±0.10	124.3±1.51	50.0±0.93	74.2±0.61	130.3±1.63	78.87±2.03
T_4 (Control + 0.050% turmeric oil)	5.1±0.30	3.4±0.30	1.7±0.06	125.5±3.03	49.2±0.68	76.3±2.70	132.2±3.59	82.12±2.01
T_5 (Control + 0.075% turmeric oil)	5.0±0.15	3.3±0.21	1.7±0.07	123.1±0.53	49.7±0.54	73.3±0.59	134.5±1.86	79.37±2.08
T_6 (Control + 0.100% turmeric oil)	5.3±0.39	3.2±0.42	1.8±0.06	124.8±1.13	49.3±0.27	75.5±1.18	132.7±2.18	78.87±2.04
P value	0.700	0.790	0.842	0.303	0.762	0.191	0.919	0.601

Each value in the table is mean of eight observations. OTC, Oxytetracycline.

Table 3. Effect of turmeric oil supplementation on meat quality of broilers

Treatment group	Cooking loss (%)	Shear force value (kg of force/cm^2)	Meat pH	Water holding capacity (%)	Tyrosine value (mg/g)	Thiobarbituric acid value (mg/g)
T_1 (Control)	22.80±1.01	1.12±0.10	6.13±0.04	62.6±0.51	23.11±0.52	0.695±0.05
T_2 (Control + OTC @ 50 ppm)	22.31±1.21	1.21±0.11	6.13±0.04	63.0±0.33	22.50±0.31	0.674±0.03
T_3 (Control + 0.025% turmeric oil)	22.34±2.02	1.14±0.10	6.17±0.04	62.7±0.52	23.00±0.44	0.674±0.01
T_4 (Control + 0.050% turmeric oil)	24.73±1.50	1.20±0.13	6.18±0.04	63.1±0.50	23.01±0.72	0.671±0.04
T_5 (Control + 0.075% turmeric oil)	22.51±1.22	1.12±0.17	6.12±0.04	63.4±0.44	22.91±0.51	0.685±0.02
T_6 (Control + 0.100% turmeric oil)	22.68±1.10	1.21±0.11	6.17±0.04	63.2±0.32	23.82±0.34	0.689±0.01
P value	0.587	0.118	0.839	0.992	0.928	0.616

OTC, Oxytetracycline. Each value in the table is mean of eight observations.

Table 4. Effect of turmeric oil supplementation on intestinal microbial load ($\log_{10}$ cfu/g) of broiler chicken

Treatment group	Total microbial count (10^9)	*Lactobacillus* (10^4)	*E. coli* (10^4)	*Clostridium* (10^1)	Haemagglutination inhibition titre
T_1 (Control)	11.03±0.31	4.12±0.12	6.25±0.07	4.86±0.50	Nil
T_2 (Control + OTC @ 50 ppm)	10.87±0.25	4.28±0.35	5.89±0.04	5.12±0.22	Nil
T_3 (Control + 0.025% turmeric oil)	10.92±0.07	4.48±0.12	6.18±0.07	5.22±0.15	Nil
T_4 (Control + 0.025% turmeric oil)	10.70±0.62	4.31±0.09	6.01±0.07	5.20±0.16	Nil
T_5 (Control + 0.075% turmeric oil)	11.14±0.49	4.66±0.10	5.91±0.06	5.33±0.12	Nil
T_6 (Control + 0.100% turmeric oil)	10.63±0.66	4.01±0.08	6.29±0.07	4.77±0.31	Nil
P value	0.801	0.639	0.147	0.935	0.993

OTC, Oxytetracycline. Each value is the mean of eight observations.

at the rate of 50 ppm or turmeric oil at graded levels had no significant effect on gut total bacterial count, *E. coli, Lactobacillus* and *Clostridium* of broiler chicken when compared to antibiotic free control group. Similar non-significant observations were also reported by Sarica *et al*. (2005) for total bacterial count and Pokhrel *et al.* (2012) for *Escherichia coli* count, when turmeric powder and ethanolic extract of coriander and turmeric was supplemented respectively to broiler diets. On the contrary, Namagirilakshmi *et al.* (2010) and Nayaka *et al*. (2013) documented significant reduction in the intestinal total microbial count. Faghani *et al*. (2014) observed marked increase in the intestinal *lactobacillus* population and decrease in *E. coli* population in turmeric extract supplemented broilers as compared to control.

Gut morphology: Effect of turmeric oil supplementation on gut morphology of broilers is presented in Table 5. Supplementing either antibiotic at 50 ppm level or turmeric oil at graded levels had no significant effect on ileal villi length of broilers as compared to control. This was in disagreement with the findings of Namagirilakshmi *et al.* (2010) and Hady *et al.* (2016) who observed significant increase in intestinal villi length when the broilers were supplemented with turmeric powder. Rajput *et al*. (2013) also reported that dietary supplementation of curcumin increased the length of intestinal villi.

Table 5. Effect of turmeric oil supplementation on gut morphology of broiler chicken.

Treatment group	Villi length (µm)	Crypt depth (µm)
T_1 (Control)	1010.332±27.986	164.487^{a}± 10.851
T_2 (Control + OTC@ 50 ppm)	963.403±28.541	205.710^{b}± 26.032
T_3 (Control + 0.025% turmeric oil)	1007.990±97.714	158.368^{a}± 9.590
T_4 (Control + 0.050% turmeric oil)	1068.573±56.206	147.787^{a}± 5.797
T_5 (Control + 0.075% turmeric oil)	1052.190±23.802	187.206^{ab}± 7.575
T_6 (Control + 0.100% turmeric oil)	1055.428±24.111	211.890^{b}± 6.718
P value	0.963	0.003**

OTC, Oxytetracycline. Each value is the mean of eight observations. ^{ab}Means with different superscripts in a column differ significantly **(P<0.01).

The ileal crypt depth was markedly higher in antibiotic supplemented group when compared to antibiotic free control group. Dietary supplementation of turmeric oil at the rate of 0.025 and 0.050% levels showed significantly lower (P<0.01) crypt depth values when compared to antibiotic supplemented group. However, the ileal crypt depth of birds fed with 0.075 and 0.100% levels of turmeric oil was comparable to that of antibiotic supplemented birds. Similar observation with turmeric powder was reported by Hady *et al.* (2016). However, Rajput *et al.* (2013) documented significant reduction in crypt depth of broilers fed with curcumin at 200 mg/kg feed.

Dietary supplementation of antibiotic increased the production cost over control group by ₹ 1.27/kg live weight gain. Among the different turmeric oil supplemented groups, 0.025 and 0.100% groups had higher profit/kg live weight gain (₹ 0.32 and 1.03 respectively) when compared to antibiotic supplemented group. Similar observation was reported by Chaudhary *et al.* (2015) in whose study 0.50% level of herbal mixture (equal concentrations of *Curcuma longa, Emblica officinalis and Nigella sativa*) showed higher profit/kg live body weight (₹ 0.96) over antibiotic supplemented group.

The results of the present investigation revealed that the effect of graded levels of turmeric oil on serum lipid profile, meat cholesterol, gut microbial population and intestinal morphology of birds was as good as antibiotic supplementation.

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