Meat Quality, Some Blood Profiles and Cecum Microbial Load in Broiler Fed With Effective Microorganisms, Turmeric (*Curcuma longa*) and Their Combination as Feed Additives

Chala Kinati Wakjira (ck2095@gmail.com)
Ambo University https://orcid.org/0000-0001-8782-2864
Negasi Ameha
Haramaya University
Meseret Girma
Haramaya University
Ajebu Nurfeta
Hawassa University

Research Article

Keywords: Broiler, Carcass quality, Economic efficiency, Probiotics, Turmeric powder

Posted Date: August 15th, 2022

DOI: https://doi.org/10.21203/rs.3.rs-1901893/v1

Abstract

A study was conducted to evaluate effect of feeding effective microorganisms (EM), turmeric (TP) and their combination (EM-TP) as natural feed additives on meat quality, some blood profiles and cecum bacterial load of broilers. A total of 192 chicks were assigned into one of four treatments control (CTL), 1ml/lit EM, 1% TP, 0.5 ml/lit EM and 0.5% TP (EM-TP) in a completely randomized design. Feed additives had higher ($P < 0.05$) b*(yellowness) value in breast and thigh meat color for TP and EM-TP than the other treatments. Shear force value is higher ($P < 0.05$) in CTL than the other in both breast and thigh muscles. The highest ($P < 0.05$) fat content was for CTL. The moisture, CP and ash of breast and thigh meat were similar ($P > 0.05$) except crude fat percentage which was higher ($P < 0.05$) for CTL group. High result ($P < 0.05$) in alkaline phosphatase (ALP) for EM and EM-TP while Cholesterol content, Low-Density Lipoprotein (LDL), and Triglycerides concentrations were decreased significantly ($P < 0.05$) by consumption of EM, T, P, and EM-TP than the control group. There were high ($P \leq 0.05$) population of total coliform count and E. coli bacteria were counted in CTL than the other treatment group. In conclusion, experimental additives can lower the crude fat in meat composition, blood cholesterol level, blood LDL cholesterol, triglyceride concentration and markedly reduced total coliform count and E. coli bacteria in the intestinal contents of broilers thereby improving gut efficiency and gut health of broiler chicken.

1. Introduction

The use of probiotics to competitively exclude the colonization of intestinal pathogens has been proposed for poultry, especially after some countries banned certain antibiotics being frequently included in rations as growth promoters (Salminen et al., 1998).

Many studies have found that including probiotic species such as *Lactobacillus, Streptococcus, Bacillus, Bifidobacterium, Enterococcus, Aspergillus, Candida*, and *Saccharomyces* in broiler nutrition improves growth performance, intestinal health, immune status, microbial load and meat characteristics like keeping quality, and sensory evaluation in broiler nutrition (Kabir, 2009). Probiotics are shown to beneficially affect the host animal by improving its intestinal microbial balance and prebiotics help the host by selectively promoting the growth and activity of one or a restricted number of bacteria in the colon (Kabir, 2009). It is suggested that such supplements alter the gut flora and increase intestinal absorption, which boosts overall performance (Sohail et al., 2012 and Caly et al., 2015).

The other feed additive, turmeric is a phytobiotic which is an essential medicinal herb that is used in chicken feed as a dietary supplement. Turmeric is the dried rhizome of *Curcuma longa* L., an herbaceous plant and native to South Asia. It is grown in tropical countries viz., Ethiopia, India, Pakistan, Myanmar, Chile, Peru,, etc. Turmeric (*Curcuma longa*) is one of exported spice in Ethiopia, Southwest Ethiopia produce this spice as a cash crop and many livelhoods is dependent on it for a living. India is the world biggest exporter and producers of turmeric, whereas, Ethiopia is the biggest exporters and producers of turmeric in Africa (Chaudhary et al., 2006). Turmeric's primary bioactive component is curcumin

(diferuloylmethane). It makes up 3–5% of the curcuminoids in turmeric rhizomes and is a powerful phenolic antioxidant (Stankovic, 2004; Jaggi, 2012). It improves broiler performances and endogenous digestive enzyme secretion, as well as triggering immunological responses and antibacterial and antioxidant properties in chicken (Burt, 2004; Khan et al., 2012). Antioxidant activity and blood biochemistry parameters are essential indicators of health and nutrient metabolism in an organism's body (Lokesh et al., 2012). The effects of turmeric powder on blood biochemistry markers and antioxidant capacity in broiler chickens have been documented in numerous studies (Emadi et al., 2007; Gowda et al., 2009; Daneshyar, 2012; Hussein, 2013). And also, turmeric powder significantly increased crude protein content in breast meat and significantly decreased ether extract in thigh meat in broilers supplemented with turmeric powder (7 g / kg) compared to the non-supplemented diet (Hussein, 2013).

Furthermore, previous research on the mode of action of phytobiotic substances and probiotic strains has shown the possibility of synergism between these classes of drugs and the performance of creatures (Hossain et al., 2012). The finding of Kim et al. (2007) indicated that plant extracts and *Lactobacillus spp*. maybe utilized as options to anti-microbial development promoters for the advancement of the development performance of broiler chicks. However, little information is available on the synergistic effects of probiotic and turmeric inclusion in broilers' diets on broiler meat quality and health benefits. Therefore, the objective of the present study was to assess the effect of effective microorganism, turmeric and their combination as an additive on meat quality, hematological parameters, serum chemistry and cecum bacterial load in broiler chickens.

2. Materials And Methods

2.1. Management of experimental birds

The experiment was conducted for 42 days. Before the beginning of the actual experiment, the experimental house was washed and disinfected; the floor was covered with *teff* straw at the depth of 7cm and disinfected using hydrogen peroxide thoroughly before placement of the birds. Each pen was equipped with a 250-watt infrared heat bulb.

A circular plastic feeder and waterer were placed in each of the pens a day before the placement of the birds. A total of one hundred- and ninety-two-day-old unsexed broiler chicks were purchased from Alama farm of Bishoftu, Ethiopia and transported to Haramaya University poultry farm. Water was available all the time and measured quantities of the experimental ration were provided on *ad-libitum* (~ 15.07% refusal) twice a day at 8:00 and 16:00 hrs. The refusals were recorded every morning for the determination of feed intakes. Bodyweight was measured at the beginning and a weekly interval during the experimental period and the end of the feeding trial using sensitive balance. The standard bio-security protocol was employed throughout the experimental period. The chicks were vaccinated against Newcastle disease (HB1) on day 7 and Lasota a booster dose on day 21 through eye drop

2.2. Dietary treatment and experimental design

The ingredients used for ration formulation were corn grain, wheat short, soybean meal, noug seed cake, turmeric, vitamin premix, di-calcium phosphate, limestone, salt, lysine, and methionine which were formulated as per the recommendation of the National Research Council (NRC, 1994). Corn grain, noug seed cake, and turmeric were hammer milled at 5mm sieve size and mixed based on a dry matter basis. lysine, methionine, di-calcium phosphate, and vitamin premix were added to the feed during mixing without hammer milling.

Adequate quantities of activated EM·1 packed in a plastic jar were obtained from Weljijie PLC located in Bishoftu, Ethiopia and transported to Haramaya University poultry farm and stored in dry place until used. The EM preparations used in this study were made following the guidelines prepared by EMROSA (2003), where activated EM·1 (1ml/liter) was added directly into chlorine-free clean drinking water which consists lactic acid bacteria (Lactobacillus and Pedicoccus) at 1 x 105 CFU/ml suspensions, yeast (Saccharomyces) at 2 x 106 CFU/ml suspension and fewer amounts of photosynthetic bacteria, actinomyces and other organisms (Matthew, 2002).

The treatment rations were formulated using Feed win software to be isocaloric and isonitrogenous to meet the nutrient requirement standards for broilers (NRC, 1994). Accordingly, the starter treatment rations contained about 3000 kcal ME/kg DM and 22% crude protein, while the finisher treatment rations contained 3100 kcal ME/kg DM and 19% crude protein (Table 1). The starter phase was until 3 weeks of age while the finisher phase was from 4th up to 6th weeks of age.

Table 1
The proportion (%) of ingredients and chemical composition of experimental diets

Ingredients	Starter (1–3 weeks)				Finisher (4–6 weeks)			
	CTL	EM	TP	EM-TP	CTL	EM	TP	EM-TP
Maize	59	59	59	59	61.7	61.7	61.7	61.7
Wheat short	3	3	3	3	8	8	8	8
DL-methionine	0.5	0.5	0.5	0.5	0.1	0.1	0.1	0.1
Soybean meal	18	18	18	18	14	14	14	14
Noug seed cake	17	17	17	17	14	14	14	14
Vitamin premix	0.2	0.2	0.2	0.2	0.2	0.2	0.2	0.2
Salt	0.3	0.3	0.3	0.3	0.5	0.5	0.5	0.5
Limestone	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5
L-Lysine	1	1	1	1	0.5	0.5	0.5	0.5
Dicalcium phosphate	0.5	0.5	0.5	0.5	0.5	0.5	0.5	0.5
Total	100	100	100	100	100	100	100	100
Turmeric (g/kg)	0	0	1	0.5	0	0	1	0.5
EM (ml/L)	0	1	0	0.5	0	1	0	0.5
Composition (% DM basis)								
Crude protein	21.08	21.08	21.43	21.65	18.99	18.99	19.31	19.51
ME (kcal/kg DM)	2968	2968	3005	2992	3095	3095	3133	3119
Ether extract	4.10	4.1	4.51	4.35	4.31	4.31	4.43	4.28
Crude fiber	3.30	3.30	3.12	3.05	3.72	3.72	3.65	3.57
Ash	11.13	11.13	12.5	12.13	12.35	12.35	13.04	12.59
Calcium	1.23	1.23	1.79	1.02	1.23	1.23	1.04	1.05
Phosphorus	0.2	0.42	0.65	0.67	0.53	0.53	0.75	0.68

DM = dry matter; CTL *= Control EM = Effective Microorganisms; TP = Turmeric Powder;* CP = crude protein; ME = Metabolizable energy; *Ca = Calcium; P = phosphorus*

The chicks were assigned to four dietary treatments having CTL = control /no additive, EM = CTL + 1 ml/lit effective microorganisms, TP = CTL + 1% turmeric powder, EM-TP = CTL + combination of 0.5 ml/lit EM +

0.5% TP following a completely randomized design of 3 replications for each treatment. Treatment groups consisted of 48 birds which were randomly distributed to replicate groups (16 birds / replicate).

2.3. Data collection and measurements of parameters

2.3.1. Meat quality traits

Meat color

The meat color from the breast and thigh was determined according to the method of AMSA (2012) using a colour flex spectrophotometer (Hunter Lab Reston, VA, USA). The device was calibrated against black and white reference tiles prior to use. A total of three readings for each sample of L*(lightness), a* (redness), and b*(yellowness) were recorded and then the average value was calculated for each sample (Hunt 1980).

Cooking loss

Forty-five minutes post-slaughter, raw breast and thigh muscle (20–30 g) from the left side (to keep uniformity) of the carcass were cut, weighed and sealed in a plastic bag (30 microns) and cooked in a thermostatically controlled water bath (Fisher Scientific, Pittsburgh, PA) at 75°C for 45 minutes as described by Rizz et al. (2007). Then, the samples were cooled in running water for 15 minutes, dried with soft tissue, and weighed.

Cooking loss was calculated as sample weight before cooking minus sample weight after cooking ×100/sample weight before cooking (Petracci and Baéza 2009).

Meat pH

Meat pH was measured on the breast and thigh muscle of the slaughtered bird 24 h after slaughter using a portable digital pH meter (CRISON pH24, CRISON Instruments SA, Spain) with a piercing electrode. After every 20 measurements, the pH meter was calibrated with pH^4 and pH^7 standard buffer solutions (Ingold Messtechnik AG, Udorf, Switzerland).

Instrumental tenderness determination

Steak preparation

Each sample was thawed for 24 hours at room temperature (24-25^{o}C) for steaks preparation. The steaks were prepared according to procedures developed by AMSA (2015).

Determination of tenderness

The Warner-Bratzler shear force (WBSF) method was used to determine instrumental tenderness (AMSA, 1978). The steak was allowed to cool down to room temperature for about an hour to evaluate

instrumental tenderness using WBSF. After cooling, the steak was cut across the long axis putting the knife tip on the heavy connective tissue side (dorsal) and the handle of the knife on the ventral side to expose the fiber direction. Three to six cores were removed parallel with the muscle fibers. The muscle fibers were needed to run parallel with the core so that the shear was across the grain. The WBSF device was used to shear each core. The shear was across the middle (center) on each core. The peak values of WBSF were recorded in N (Newton) for each core. The average values for the three to six cores were taken for the determination of the value of each steak.

2.3.2. Sensory evaluation: The determined sensory parameters were juiciness, tenderness, flavor and overall acceptance. Skinless breast samples were frozen until cooking; the pieces were thawed at room temperature minced and cut into 2.5 cm cubes. The breast meat was cooked for 15 minutes on a pan. After cooking, the pieces were cooled to room temperature. The breast meats were evaluated following the sensory profile procedure (ISO, 2003). The panel consists of 10 trained panelists from Haramaya University Animal and Range Science staff members and post-graduate students. Panelists were instructed to chew and taste the meat and rinsed their mouths with bottled drinking water which was kept at room temperature between each sample and paused for 20 seconds before tasting the next sample.

A five-point category scale was used to evaluate the sensory characteristics of the products as follows:

Aroma and Flavor: very weak (1), weak (2), intermediate (3), strong (4), very strong (5). **Juiciness**: very juicy (1), juicy (2), intermediate (3), dry (4), very dry (5).

Overall acceptability

like very much (1), like (2), intermediate (3), dislike (4), dislike very much (5).

2.3.3. Chemical analysis of feed and meat

Representative samples of ingredients were analyzed for chemical composition before ration formulation (Table 2).

Table 2
Chemical composition of feed ingredients (% dry matter, except dry matter and metabolizable energy)

Feed ingredients	Chemical Composition							
	DM%	CP	EE	Ash	CF	Ca	P	ME (kcal/kg DM)
Corn grain	90.5	8.78	4.28	4.73	2.97	0.03	0.83	3736.3
Wheat short	91.0	15.0	3.84	5.02	9.87	0.19	0.78	2980.3
Soybean meal	93.75	39.68	8.53	6.37	6.04	0.34	0.66	3618.0
Noug seedcake	93.0	30.8	7.84	9.38	18.5	0.33	0.32	2314.3
Turmeric Powder	89.37	8.63	3.99	4.15	1.65	0.28	0.15	3852.4
DM: Dry mater CP: crude protein; EE: ether extract; CF: crude fiber; ME: Metabolizable energy; Ca: Calcium; P: phosphorus;								

The chemical analysis of breast and thigh muscle was carried out with out skin. The meat samples from breast and thighs of each slaughtered bird were minced separately, dried and homogenized and analyzed for moisture, crude protein, fat and ash content using standard methods (AOAC, 2011).

2.3.4. Hematology and serum biochemical indices

At the end of the experiment, blood samples (5 ml each) were collected from the wing vein of 12 hens from each treatment and analyzed for hematology and chemistry. 2.5 ml of blood was drawn into EDTA (Ethylene Diamine tetra acetic acid) tube for the determination of total red blood cell (RBC), hemoglobin (Hb), packed cell volume (PCV), white blood cell (WBC) total protein (TP), and serum cholesterol concentration. The RBC and WBC were determined by using an improved Neubauer hemocytometer chamber (Dacie and Lewis, 1991). Hemoglobin concentration was determined by using Actin hematin methods. The packed cell volume (PCV) was determined by microhematocrit (capillary) tubes method and centrifuged at 3000 revolutions per minute (rpm) for 5 minutes and reading on hematocrit reader. Finally, serum was harvested from blood collected in a plain tube which was transferred to an Endorphin tube and stored at -20^{o}C and analyzed for serum chemistry parameters (serum total protein and albumin, total cholesterol count, HDL-C, and LDL-C) with an automated chemistry analyzer (Douglas et al., 2010). The globulin value was determined by the difference between serum total protein and albumin (Doumas et al., 1981).

The results were calculated as follows:

Mean corpuscular volume (MCV), $= \frac{PCV}{RBC} x 10,$

Mean corpuscular haemoglobin concentration (MCHC) $= \frac{Hb}{PCV} x 100,$

Mean corpuscular haemoglobin (MCH) was computed as = $\frac{Hb}{RBC}x10,$ (Irizaary-Rovira, 2004). While the remaining 2.5 ml was drawn in a plain tube and left to coagulate. Serum was separated after centrifugation at 3,000 rpm x g for 15 min and stored at -20°C until used. Serum alanine aminotransferase (ALT), aspartate aminotransferase (AST), alkaline phosphatase (ALP) activities, cholesterol, glucose and triglycerides concentrations were measured by using enzyme/ buffer and substrate kits. Total serum immunoglobulin concentration was determined by serum zinc sulfate turbidity test by reading the optical density of the test and the control separately at 545 nm using a spectrophotometer (Mondesire, 2004).

2.3.5. Cecum bacterial load determination

The effect of dietary treatment on caecal bacterial load was assessed by carrying out the total bacterial count in the caecal feces of experimental birds used for carcass determination. A fecal sample from each treatment replicate was aseptically obtained and diluted serially using 2% buffered peptone water until a 10^{-8} ratio was obtained. Then, 0.1 ml of each dilution ratio was plated on plate count agar (Merck, Modderfontein, South Africa) using the spread plate method which was incubated at 37°C for 24 h (Cheesbrough, 1985) for the total coliform count. Eosin Methylene Blue (EMB) agar was used for *E. coli*, which were incubated at 37°C from 8 to 24 h (Collee et al., 1989). Then plates were counted between 24 and 48 h after inoculation (Collee et al., 1989; Cheesbrough, 1985). The counted data were transformed to log10 cfu/ml.

2.4. Statistical Analysis

Data was analyzed using the general linear model procedure of statistical analysis systems software (SAS, 2009). Differences between treatment means were separated using Duncan's multiple range tests. The following model was used for data analysis. $Yij = \mu + Ti + e_{ij}$, Where: Y_{ij} = represents the j^{th} observation in the i^{th} treatment level, μ = overall mean, Ti = treatment effect and e_{ij} = random error.

3. Results

3.1. Meat quality characteristics and proximate composition

Feed additives had no significant ($p > 0.05$) influence on L*(lightness), and a* (redness) of meat (Table 3). The b*(yellowness) value of breast and thigh meat was higher ($P < 0.05$) for TP and EM-TP treatment group than in the other treatments, while the lowest ($P < 0.05$) value was for the CTL group. The pH and cooking loss were similar ($P > 0.05$) among treatments. Shear force value for CT was higher ($P < 0.05$) than TP and EM-TP group for breast muscle. The highest ($P < 0.05$) shear force was for the CLT group for thigh muscle. The highest ($P < 0.05$) fat content was for the control group. The moisture, CP, and ash contents of breast and thigh meat were not significantly different among treatments.

Table 3. Meat quality trait and proximate composition of chicken breast and thigh in chicks supplemented with effective microorganisms, turmeric and its combination as feed additives

Parameters		Treatment				SEM	P-Value
		CTL	EM	TP	EM-TP		
Color parameter							
L*	breast	47.54	46.48	46.73	48.12	0.852	0.504
	thigh	47.56	45.85	46.27	47.12	0.803	0.463
a*	breast	4.95	4.53	4.86	5.17	0.563	0.875
	thigh	6.28	4.91	5.97	5.87	0.422	0.197
b*	breast	10.13^{c}	11.24^{b}	12.68^{a}	12.59^{a}	0.205	0.000
	thigh	11.13^{c}	12.11^{b}	13.35^{a}	13.26^{a}	0.273	0.004
pH	breast	6.05	6.26	6.34	6.36	0.308	0.883
	thigh	6.03	6.27	6.36	6.38	0.304	0.841
Cooking Loss	breast	14.57	16.16	17.5	18.27	3.860	0.909
	thigh	15.9	16.82	18.03	18.8	3.640	0.943
Shear force (N)	breast	18.93^{a}	15.24^{ab}	17.56^{bc}	14.74^{c}	0.855	0.026
	thigh	20.64^{a}	15.66^{b}	17.34^{bc}	14.66^{c}	0.816	0.004
Proximate composition							
Moisture	breast	73.71	74.76	75.25	73.07	0.610	0.120
	thigh	74.71	75.76	76.25	73.73	0.622	0.081
Protein	breast	22.37	23.55	22.7	23.24	0.630	0.570
	thigh	21.42	23.45	22.64	23.34	0.664	0.197
Crude fat	breast	3.9^{a}	1.09^{b}	1.83^{b}	1.52^{b}	0.500	0.020
	thigh	4.70^{a}	2.32^{b}	1.75^{b}	1.61^{b}	0.4644	0.005
Ash	breast	1.35	1.13	1.42	1.18	0.220	0.770
	thigh	0.89	21.81	0.72	0.77	10.549	0.444

^{abc} Within a row means with different letters are significantly different at $P < 0.05$.

L* = lightness, a*= redness, b*= yellowness, SEM = Standard error of the mean.

3.2. Sensory evaluation

There was no significant difference (P > 0.05) among treatment in broilers fed different feed additives (Fig. 1).

3.3. Hematology and serum biochemical indices

The effects of supplementation of effective microorganisms, turmeric powder and a combination of effective microorganisms and turmeric powder, as an additive, on hematological and serum biochemical parameters in broiler chicken are presented in Tables 4 and 5, respectively. The results showed that there were no significant effects of additives on all hematological parameters of the blood among the experimental groups (Table 4). The highest (P < 0.05) ALP was for EM and EM + TP while the lowest (P < 0.05) was for TP. Cholesterol, LDL, and triglyceride concentrations for the control group were the highest (P < 0.05) while the additives had similar (P < 0.05) values among each other.

Table 4
Hematological traits of broiler chickens fed with effective microorganisms, turmeric and its combination as feed additives

Items	Dietary supplementations				SEM	p value
	Control	EM	TP	EM-TP		
Hemoglobin(Hb) (g/dL)	10.65	10.45	10.70	10.02	0.309	0.437
RBCx10^6/µl	2.70	2.63	2.70	2.50	0.109	0.447
WBC x 10^3/µl	28.33	28.33	26.17	29.50	3.969	0.727
PCV (%)	34.00	33.33	35.00	32.00	1.184	0.394
MCV (fl)	126.23	127.18	128.85	129.72	1.627	0.466
MCH (pg)	39.58	39.87	39.55	40.60	0.730	0.726
MCHC (%)	31.32	31.32	30.68	31.32	0.353	0.526

RBC = Red Blood Cell, WBC = White Blood Cell, Packed cell volume; MCV = Mean cell volume; MCH = Mean cell hemoglobin; MCHC = Mean cell hemoglobin concentration.

Table 5
Serum biochemical traits of broiler chickens fed on effective microorganisms, turmeric and its combination as feed additives

Parameter	Dietary supplementations				SEM	p-value
	Control	EM	TP	EM + TP		
ALT (u/l)	19.60	19.32	19.82	19.32	0.857	0.969
AST (u/l)	131.50	129.67	130.67	128.83	4.619	0.978
ALP (u/l)	918.2^{b}	1060.7^{a}	743^{c}	1051.8^{a}	34.773	0.000
Cholestrol (mg/dl)	173.62^{a}	141.43^{b}	147.53^{b}	141.1^{b}	6.595	0.024
LDL (mg/dl)	89.93^{a}	72.00^{b}	77.65^{b}	72.73^{b}	3.363	0.019
HDL (mg/dl)	42.02	44.42	40.10	39.60	2.706	0.603
Glucose (mg/dl)	229.33	224.67	223.00	240.17	8.040	0.472
Triglycerides concentration (mg/dl)	57.18^{a}	43.867^{b}	38.3^{b}	39.917^{b}	3.119	0.010
Total serum protein (g/dl)	3.93	3.98	3.95	3.95	0.087	0.981
Albumin (g/dl)	2.82	2.87	2.80	2.78	0.079	0.889
Globulin (g/dl)	1.14	1.16	1.21	1.22	0.032	0.298
Urea (mg/dl)	13.15	12.93	13.35	14.20	0.902	0.772
Creatinine (mg/dl)	0.63	0.59	0.56	0.60	0.046	0.684

^{abc} Within a row means with different letters are significantly different at $P < 0.05$.

ALT = Alanine aminotransferase, AST = aspartate aminotransferase, ALP = alkaline phosphatase, LDL = Low Density Lipoprotein, HDL = High Density Lipoprotein

3.4. Cecum bacterial load

The effect of supplementation of EM, TP, and EM-TP on caecal Total Coliform Count (TCC) and *E. coli* in broiler chicken is presented in Fig. 1. There were high ($P \leq 0.05$) populations of total coliform and *E.coli* bacteria in CTL than the other treatment groups.

There were a reduction ($P \leq 0.05$) in total coliform count and *E. coli* bacteria in chicks fed EM ($8.48*10^5$cfu/ml and $8.12*10^5$cfu/ml), TP ($6.33*10^5$cfu/ml and $5.92*10^5$cfu/ml) and EM-TP ($4.55*10^5$cfu/ml and $4.5*10^5$cfu/ml) as compared with the control ($17.48*10^5$cfu/ml and $16.68*10^5$cfu/ml) respectively. However, there were no significant ($P \geq 0.05$) differences among additives (EM, TP and EM-TP) for total coliform and *E. coli* bacteria.

[ab] Means with different lowercase superscript letters are significant differences among treatments ($p < 0.05$).

cfu = coliform forming unit, CTL = Control, EM = Effective microorganism, TP = Turmeric Powder, EM-TP = Combination of Effective microorganism and Turmeric Powder, E. coli = *Escherichia coli*, T. C. Count = *Total* coliform Count

Figure 2. The effect of additives on caecal microbial load counts for total coliform and *E. coli* bacteria

4. Discussion

4.1. Broiler meat quality characteristics and Proximate analysis

Color is one of the main indicators of the quality of most feeds (Smith et al., 2002). The L* value is the main parameter that determines poultry meat color. It is an important meat quality trait as it affects consumer acceptability of meat (Adeyemi et al., 2016). The meat color in the current experiment is within the normal range ($46 < L^* < 53$) of breast and thigh meat color reported by Zhang and Barbut (2005). However, Abdulla *et al*. (2017) reported higher L (lightness), a (redness) and b (yellowness) values of breast muscles from Cobb 500 chickens supplemented with probiotic containing Bacillus subtilis during the rearing period, compared to control birds. The possible reason for the high b* yellowness color in the current experiment for TP and EM-TP could be due to the presence of bio-active compounds in turmeric feed additive, which could alter muscle pigmentation (Ruby, 1995). Additionally, the study of Faria et al. (2009) indicated that the ingestion of a larger amount of feed rich in carotenoids by growing chickens provides a greater intensity of yellow color in the meat, resulting in a higher b* value. A similar study result by Kanani et al. (2017) indicated that turmeric and cinnamon powders elevated the water-holding capacity and pH.

The similar pH values among the treatments in the current study, indicates that they were closer to neutral, resulting in similar lightness (L*) and redness (a*) values for all treatment groups. This could be because experimental birds did not experience transportation stress and were subjected to the same pre-slaughter management circumstances as commercial broiler production (Souza et al., 2011). These factors may have contributed to the pH levels being similar among the treatments in this study.

Effective microorganisms, TP, and their combination did not affect mean breast and thigh cooing loss. Cooking loss is a measurement of how much water is lost during cooking due to shrinkage. The pH and lipid content of the tested muscle can affect cooking loss readings (Souza, 2004). The finding indicates that additives used in the current experiment can be used since they do not have any effect on cooking loss.

Warner-Bratzler Shear Force (WBSF) is one of the most commonly used instruments in estimating meat tenderness and texture quality of poultry meat, whereby the higher WBSF values are associated with less

tender poultry meat (Zhuang et al., 2008). Therefore, the less tender meat for both breast and thigh meat observed in the CTL group agrees with the results observed by Zhang et al. (2005), who investigated the effects of *Saccharomyces cerevisiae* cell components on meat quality of male broilers. The shear forces determined in cooked breast and thigh muscle in experimental groups was lower as compared with the control in the current experiment. Contrary to the current result Peter et al. (2015) indicated that higher shear force value of breast and thigh muscle was observed in probiotic supplemented than the other group. This difference might be due to the effectiveness of a probiotic application which may depend on many factors (Patterson and Burkholder, 2003), such as species composition and viability, administration level, application method, frequency of application, overall diet, bird age, overall farm hygiene, and environmental stress factors (Zhou et al., 2010).

The lower crude fat percentage of the thigh and breast meat in those fed additives in the current study agreed with the finding of Pietras (2001) who reported that crude fat and total cholesterol content tended to decrease in the meat of chickens given probiotics (Lactobacillus acidophilus and Streptococcus faecium). Addition of turmeric at a rate of 3 g/kg feed reduced the meat fat content and increased the carcass quality of broilers (Al-Sultan, 2003, Samarasinghe et al. 2003, Emadi and Kermanshahi, 2006). The probable reason for fat and cholesterol content reduction could be due to the presence of polyunsaturated fatty acids and the antioxidant content of turmeric.

4.2. Sensory evaluation of broilers breast meat

The results of sensory evaluation in the current experiment revealed that feeding of effective microorganisms and turmeric did not affect color, flavor, or smell in chicken meat which is in agreement with the study of AL-Sultan (2003) who reported that turmeric did not induce any abnormal flavor, color, or smell in broilers breast meat. According to Liu *et al.* (2012), probiotic supplementation to a chick's diet resulted in a higher degree of satisfaction in flavor pleasantness scores for the consumer compared with the control.

4.3. Effect of effective microorganisms and turmeric feed additives on hematological and biochemical indices

Hematological and serum biochemical parameters are useful indicators of the physiological responses of animals to the diet they are consuming (Madubuike and Ekenyem, 2006). In the current study, EM, TP and EM-TP supplementation had a similar effect on blood hematological traits, which were found to be within the normal ranges (RBC: 2.5–3.5 x106 µl, PCV: 22–35%, Hb: 7–13 g/dl, WBC: 12–30 x 103µl, MCV: 90–140 fL, MCH: 33–47 pg/cell and MCHC: 26–35 g/dl) for healthy broiler chickens (Wakenel, 2010). Moreover, Ibrahim (2012) reported that probiotic supplementation did not affect blood hematological traits. The current findings are consistent with those of Emadi et al. (2007), who found that adding turmeric to broiler hematocrit levels at days had no significant effect.

Antioxidant activity and blood biochemistry parameters are essential indicators of health and nutrient metabolism in an organism's body (Lokesh et al., 2012). All the serum biochemical parameters were

within the normal range (AST = 70–220 U/L, ALP = 568–8831 U/L, ALT = 19–50 U/L, Total protein = 2.5–4.5 g/dl, Uric acid = 1.9–12.5 mg/dl, Total Cholesterol = 87–192 mg/dl, Triglycerides = 45.7–172 mg/dl, Albumin = 1.17–2.74 g/dl, Glucose = 200–500 mg/dl, LDL = 80–120 mg/dl, HDL = 35–86 mg/dl, Globulin = 0.5–1.8 g/dl, Creatinine = 0.55–0.95 mg/dl) (Clinical diagnostic division, 1990; Meluzzi et al., 1992; Lumeij, 1997; Thrall, 2007) in the current experiment. The finding in the current study agrees with those of Yazhini et al. (2018), Siadati et al. (2017), and Iqramu et al. (2017), who reported that supplementing poultry with probiotics reduced serum total cholesterol levels. Following probiotic administration to broilers, the reduction of cholesterol and fat content in the breast and thigh meat was observed (Hossain et al., 2012). Lactic acid bacteria are also a probiotic which reduces the cholesterol level by assimilating endogenous or exogenous originated cholesterol in the intestinal tract (Gilliland,1989), reducing or inhibiting the expression levels of Niemann-Pick C1-like 1a protein, expressed on the surface of enterocytes, which reduces the cholesterol absorption (Huang and Zheng 2010). On the other hand, Shirisha et al. (2017) observed no significant difference in total cholesterol levels between probiotic-supplemented and control birds.

The lower low-density lipoprotein concentration in the treatments with additives in the current experiment concur with the findings of Kalavathy et al. (2010) who reported that probiotic supplementation decreased serum low-density lipoprotein level and but had no significant effect on serum high density lipoprotein level in broiler chickens. Consistent with the current finding Ashayerizadeh et al. (2011) reported that supplementation of probiotics did not affect serum high-density lipoprotein concentration. Supplementation of probiotic bacteria to broiler chicken altered the lipoprotein metabolism of birds favorably with a more pronounced reduction in the total cholesterol and low density lipoprotein cholesterol concentration. Moreover, Kermanshahi and Rasi (2006) reported that turmeric rhizome at 0.05–0.2% levels decreased low-density lipoprotein in layer breed. Probably the reason that the lower result of low density lipoprotien in TP treatment could be due to turmeric effect in the regulation of alkaline phosphatase (Upadrasta and Madempudi, 2016; Khalesi, 2014) and lactate dehydrogenase in the broiler chickens' blood (Kermanshahi and Rasi, 2006). Hosseini-Vashan et al. (2012) reported significant reductions in the activities of alanine aminotransferase, aspartate aminotransferase and alkaline phosphatase enzymes in broiler chickens fed 0.4% and 0.8% turmeric powder under heat stress. In contrast to the current study, Kumari et al. (2007) and Mehala & Moorthy (2008) reported that the activity of some liver enzymes, such as alkaline phosphatase and low density lipoprotien, and glucose concentrations did not change with the dietary inclusion of turmeric powder in all the treated broiler chicken groups. The variation with the current study might be due to the dose effect of turmeric.

Regarding the concentration of triglycerides, the present study agrees with the findings of Al-Saad et al. (2014), Kalavathy et al. (2010), Ashayerizadeh et al. (2014), Abeer *et al*. (2015) and Iqramu et al. (2017), who reported a significant decrease in serum triglycerides level in broiler chickens fed probiotic as an additive. Similarly, the finding of Nouzarian et al. (2011) reported that triglyceride concentration of the serum was markedly reduced by turmeric powder inclusion in the diet in comparison with the control diet. This phenomenon may be due to the lowering hepatic lipogenesis effect of turmeric powder, because

triglycerides are produced in the liver by hepatic lipogenesis and are secreted into the plasma (Lanza-Jacoby, 1986; Herzberg and Rogerson, 1988).

4.4. Antibacterial effects of effective microorganism and turmeric

Reduction in the total coliform count and *E. coli* bacteria indicated that there was variation among the treatment which is supported by the idea of EMROSA (2006) which revealed that EM is self-sterilizing (pH between 3.4 to 3.7); therefore, pathogens cannot survive in EM. The volatile fatty acids produced by probiotic bacteria are lipophilic penetrating the bacterial cell wall and producing H^+ ions, which in turn destroy the internal physiology of the bacterial cell (Kuruti et al., 2017). It can be concluded that the use of probiotics as an additive affects the balance of caecal volatile fatty acids, which in turn exert an antimicrobial effect in poultry. Similarly, the finding of Deniz et al. (2011) indicated that dietary probiotic supplementation has markedly raised the bacillus population in the caecum while the enterococci and coliform populations were significantly reduced compared to the non-supplemented control. Fuller (1977) also, found that host-specific Lactobacillus strains were able to decrease *E. coli* in the crop and small intestine.

The study of Fitoni et al. (2013) indicated that turmeric with a curcumin active compound could inhibit the growth of coliform bacteria with a total of 108 cfu colonies compared with treatment without turmeric that contained more than 300 cfu of coliform bacteria. The curcumin antibacterial mechanism was that these particles entered the bacteria cell walls by completely damaging the cell walls so that it resulting in cell death (Bhawana et al., 2011).

5. Conclusion

In conclusion, the findings of this study revealed that dietary feed additives (EM, TP, and EM-TP) significantly reduced crude fat in meat, blood total cholesterol, blood low density lipoprotein, triglyceride concentration, total coliform count and *E. coli* in the intestinal contents of Cobb 500 broilers, resulting in improved gut efficiency and gut health in broiler chicken.

Declarations

Ethical Approval

The protocols for this experiment, use, and care of broilers were carried out in accordance with the consent of the newly established (but not yet fully functional they were on the process) Animal Care and Use Committee of Haramaya University, Ethiopia

Consent to Participate and Publish

This study with the consent of the Haramaya University ethics committee which was a newly established committee not yet fulfilling the formal guidelines they are in the process to have the formal guideline for approval.

Funding

"This work was supported by Ministry of education(no Grant numbers) and Ethiopian Institute of Agricultural Research (no grant no.) which was funded indirectly through the institutional process without author's contact."

Competing Interests

"The authors have no relevant financial or non-financial interests to disclose."

Author Contributions

Chala Kinati was the one who came up with the idea and wrote the paper. The manuscript was critically edited for key intellectual content by Negasi Ameha, Meseret Girma, and Ajebu Nurfeta, who also authorized the final version for publication.

Availability of data and materials

The datasets used and/or analysed during the current study are available from the corresponding author on reasonable request.

References

1. Abeer ESM, Mosaad SA (2015) Effect of dietary probiotic and/or prebiotic supplementation on growth performance, carcass traits and some serum biochemical alterations in broiler chicken. J Anim Sci Adv 5(11):1480–1492
2. Adeyemi KD, Shittu RM, Sabow AB, Ahmed AA, Karim R, Karsani SA, Sazili AQ (2016) Comparison of myofibrillar protein degradation, antioxidant profile, fatty acids, metmyoglobin reducing activity, physicochemical properties and sensory attributes of gluteus medius and infraspinatus muscles in goats. J Anim Sci Technol 5:1–17
3. Al-Saad S, Abbod M, Abo Yones A (2014) Effect of some growth promoters on blood hematology and serum composition of broiler chickens. Int J Agricultural Res 9(5):265–270
4. AL-Sultan. The effect of *Curcuma longa* (turmeric) feeds additive on the overall performance of broiler chickens.International Journal of Poultry Science, 2(5):351–353
5. AMSA (American Meat Science Association) (2012) Meat color measurement guidelines. (AMSA). Champaign, Illinois, USA)
6. AMSA (American Meat Science Association) (2015) Research guidelines for cookery, sensory evaluation, and instrumental tenderness measurements of meat, 2nd edn. Champaign, Illinois, USA

7. AMSA (American Meat Science Association) (1978) Guidelines for Cookery and Sensory Evaluation of Meat. Amer. Meat Sci. Assoc. and Natl. Live Stock and Meat Board, Chicago, IL
8. AOAC (2011) Official methods of analytical chemist, 18th edn. published by AOAC International, North Frederick Avenue, Gaithersburg, Maryland, USA
9. Ashayerizadeh A, Dabiri N, Mirzadeh KH, Ghorbani MR (2011) Effect of dietary supplementation of probiotic and prebiotic on growth indices and serum biochemical parameters of broiler chickens. J Cell Anim Biology 5:152–156
10. Ashayerizadeh O, Dastar B, Samadi F, Khomeiri M, Yamchi A, Zerehdaran S (2014) Comparison between the effects of two multi-strain probiotics and antibiotics on growth performance, carcass characteristics, gastrointestinal microbial population and serum biochemical values of broiler chickens. J Anim Sci 3(4):110–119
11. Bhawana RK, Basniwal H, Buttar S, Jain VK, Jain N (2011) Curcumin nanoparticles: preparation, characterization, and antimicrobial study. J Agric Food Chem 59:2056–2061
12. Bressan MC (1998) Efeito dos fatores pré e pós-abate sobre a qualidade da carne de peito de frango [tese]. Campinas (SP):Faculdade de Engenharia de Alimentos, Universidade Estadual de Campinas
13. Burt S (2004) Essential oils: their antibacterial properties and potential applications in foods – a review. Int J Food Microbiol 94:223–253
14. Caly DL, D'Inca R, Auclair E, Drider D (2015) Alternatives to antibiotics to prevent necrotic enteritis in broiler chickens: A microbiologist's perspective. *Frontiers in Microbiology*, 6: P.1336
15. Castellini C, Mugnai C, Dal Bosco A (2002) Effect of organic production system on broiler carcass and meat quality. Meat Sci 60(3):219–225
16. Chaudhary AS, Sachan SK, Singh RL (2006) Studies on varietal performance of turmeric (Curcuma longa L.). Indian J Crop Sci 1:189–190
17. Cheesbrough M (1985) Medical laboratory manual for tropical countries. Microbiology 2:400–480
18. Clinical diagnostic division (1990) Veterinary reference guide. Eastman Kodak Company, Rochester
19. Collee JG, Duguid JP, Fraser AG, Marmion BP (1989) Practical medical microbiology, 2nd edn. Churchill Livingstone, New York, pp 456–479
20. Dacie JV, Lewis SM (1991) Practical Haematology. 7th Edition, Churchil Livingstone, Edinburgh, 54–79
21. Daneshyar M (2012) The effect of dietary turmeric on antioxidant properties of thigh meat in broiler chickens after slaughter. Anim Sci J 83:599–604
22. Deniz G, Orman A, Cetinkaya F, Gencoglu H, Meral Y, Turkmen I (2011) Effects of Probiotic (Bacillus Subtilis DSM 17299) Supplementation on the Caecal Microflora and Performance in Broiler. Revue Med Veterinary 162(11):538–545
23. Douglas J, Weiss K, Wardrop J (2010) Veterinary Hemathology. 6th edition, Blackwell Publishing Ltd. Esteghamat

24. Doumas BT, Bayso DD, Carter RJ, Peters T, Schaffer R (1981) Determination of total serum protein. Clin Chem 27:1642–1643
25. Fitoni CN, Asri MT, Hidayat MT (2013) Pengaruh pemanasan filtrat rimpang kunyit (Curcuma longa) terhadap pertumbuhan koloni bakteri coliform secara in vitro. Lentera Biology 2(3):217–221
26. Emadi M, Kermanshahi H (2006) Effect of turmeric rhizome powder on performance and carcass characteristics of broiler chickens. Int J Poult Sci 5:1069–1072
27. Emadi M, Kermanshahi H, Maroufyan E (2007) Effect of varying levels of turmeric rhizome powder on some blood parameters of broiler chickens fed corn-soybean meal-based diets. Int J Poult Sci 6:345–348
28. EMROSA (Effective microorganisms research organization of South Africa) (2003)Emrosa Inc., South Africa. DOI: https://www.researchgate.net/publication/239850224
29. EMROSA (Effective Microorganisms research organization of South Africa) (2006) User's manual. Emrosa, inc. South Africa
30. Faria PB, Bressan MC, Souza XR, Rodrigues EC, Cardoso GP, Gama LT (2009) Composic¸a˜o proximal e qualidade da carne de frangos das linhagens Paraı´so Pedreˆs e Pescoc¸o Pelado (Proximal composition and meat quality of broilers of "Paraı´so Pedreˆs" and "Pescoc¸o Pelado"). Revis Bras Zootec 10.1590/S1516- 35982009001200023
31. Fuller R (1977) The importance of lactobacillus in maintaining normal microbial balance in the crop. Br Poult Sci 18(1):85–94
32. Gilliland SE (1989) Acidophilus milk-products a review of potential benefits to consumers. J Dairy Sci 72:2483–2495
33. Gowda NK, Ledoux DR, Rottinghaus GE, Bermudez AJ, Chen YC (2009) Antioxidant efficacy of curcuminoids from turmeric (*Curcuma longa L.*) powder in broiler chickens fed diets containing aflatoxin B1. Br J Nutr 102:1629–1634
34. Gur S, Turgat-Bubk D, Gur N (2006) Antimicrobial activates and some fatty acids of turmeric, ginger root and linseed are used in the treatment of infectious diseases. World J Agricultural Sci 2:439–442
35. Herzberg GR, Rogerson M (1988) Hepatic fatty acid synthesis and triglyceride secretion in rats fed fructose- or glucose-based diets containing corn oil, tallow or marine oil. J Nutr 118:1061–1067
36. Hossain Elias GM, Kim SK, Lee, Chul Ju Y (2012) Growth Performance, Meat Yield, Oxidative Stability, and Fatty Acid Composition of Meat from Broilers Fed Diets Supplemented with a Medicinal Plant and Probiotics. Asian-Australian J Anim Sci 25(8):1159–1168
37. Hosseini-Vashan SJ, Golian A, Yaghobfar A, Zarban A, Afzali N, Esmaeilinasab P (2012) Antioxidant status, immune system, blood metabolites and carcass characteristic of broiler chickens fed turmeric rhizome powder under heat stress. Afr J Biotechnol 11:16118–16125
38. Huang Y, Zheng Y (2010) The probiotic Lactobacillus acidophilus reduces cholesterol absorption through the down-regulation of Niemann-Pick C1-like 1 in Caco-2 cells. Br J Nutr 103:473–478

39. Hunt MC(1980) Meat color measurements. Proceedings of the Meat Conference of the American Meat Science Association. Purdue University, Lafayette, Indiana. p. 41–46
40. Hussein SN (2013) Effect of turmeric (*Curcuma longa*) powder on growth performance, carcass traits, meat quality, and serum biochemical parameters in broilers. J Adv Biomedical Pathobiology Res 3:25–32
41. Ibrahim A(2012) Hematological and some biochemical values of indigenous chickens in
42. Al-Ahsa, Saudi Arabia during the summer season.Asian Journal of Poultry Science, 6(4):138–145
43. Iqramu MH, Nazim A, Mohammad AM (2017) Comparative analysis of body weight and serum biochemistry in broilers supplemented with some selected probiotics and antibiotic growth promoters. J Adv Veterinary Anim Res 4:288–294
44. Irizaary-Rovira AR (2004) Avian and reptilian clinical pathology (Avian hematology & biochemical analysis). Section XI. In: Cowell RL (ed) Veterinary Clinical Pathology Secrets. Elsevier Inc., St. Louis, MO, USA, pp 282–313
45. ISO (International Standards Organization) (2003) ISO 13299. General Guidance for Establishing a Sensory Profile. ISO, Geneva, Switzerland
46. Jaggi L (2012) Turmeric, curcumin and our life: A review. Bull Environ Pharmacol Life Sci 7:11–17
47. Kabir SML (2009) The role of probiotics in the poultry industry. Int J Mol Sci 10:3531–3546
48. Baghban KP, Daneshyar M, Aliakbarlu J, Hamian F (2017) Effect of Dietary Turmeric and Cinnamon Powders on Meat Quality and Lipid Peroxidation of Broiler Chicken under Heat Stress Condition. Veterinary Res forum 8(2):163–169
49. Khalesi S, Sun J, Buys N, Jayasinghe R (2014) Effect of probiotics on blood pressure. A Systematic review and meta-analysis of randomized, controlled trials. Hypertension 70:1–13
50. Khan RU, Rahiman ZU, Javed I, Muhammad F (2012) Effect of vitamins, probiotics and protein on semen traits in post-molt male broiler breeders. Anim Reprod Sci 135:85–90
51. Kalavathy R, Abdullah N, Jalaludin S, Ho YW (2010) Effects of *Lactobacillus cultures* on growth performance, abdominal fat deposition, serum lipids and weight of organs of broiler chickens. Br Poult Sci 44:139–144
52. Kermanshahi H, and Riasi A (2006) Effect of turmeric rhizome powder (Curcuma longa) and soluble NSP degrading enzyme on some blood parameters of laying hens. Int J Poult Sci 5(5):494–498
53. Kim DW, Kim SH, Yu DJ, Kang GH, Kim JH, Kang HG, Jang BG, Na JC, Suh OS, Jang IS, Lee KS (2007) Effects of single or mixed supplements of plant extract, fermented medicinal plants and Lactobacillus on growth performance in broilers. Korean J Poult Sci 34:187–196
54. Kumari P, Gupta MK, Ranjan R, Singh KK, Yadava R (2007) Curcuma longa as a feed additive in broiler birds and its pathophysiological effects. Indian J Exp Biol 45:272–277
55. Kuruti K, Nakkasunchi S, Begum S, Juntupally S, Anupoju GR (2017) Apid generation of volatile fatty acids (VFA) through anaerobic acidification of livestock organic waste at low hydraulic residence time (HRT). Bioresour Technol 238:188–193

56. Lanza-Jacoby S (1986) Effect of continuous and discontinuous intravenous or intragastric total parenteral nutrition in rats on serum lipids, liver lipids and liver lipogenic rates. J Nutr 116:733–741
57. Liu Xiaolu H, Lv YL, Xu Q, Yin C, Zhang K, Wang P, Jiye Hu (2012) Growth Performance and Meat Quality of Broiler Chickens Supplemented with Bacillus Licheniformis in Drinking Water. Asian-Australian J Anim Sci 25(5):682–689
58. Lokesh G, Ananthanarayan SR, Murthy VN (2012) Changes in the activity of digestive enzymes in response to chemical mutagen diethyl sulfate in the silkworm, Bombyx mori L. (Lepidoptera: Bombycidae). Asian J Appl Sci 5:431–437
59. Lumeij JT(1997) Avian Clinical Biochemistry.In: KANEKO, J.J.; HARVEY, J.W.; BRUSS, M.L. Clinical Biochemistry of Domestic Animals 5th edition. SanDiego, Academic Press, p. 932
60. Madubuike FN, Ekenyem BU (2006) Haematology and serum biochemistry characteristics of broiler chicks fed varying dietary levels of Ipomoea asarifolia leaf meal. Int J Poult Sci 5(1):9–12
61. Matthew W(2002) Effective Microorganisms (EM) Evaluated for Poultry Production and Research. Available at http://www.emturkey.com.tr/eskisite/TR/dosya/1-371/h/ effective-microorganisms-em.pdf
62. McLandsborough LA (2004) Food Microbiology Laboratory. CRC Press, Boca Raton, FL
63. Mehala C, Moorthy M (2008) Effect of Aloe vera and *Curcuma longa* (Turmeric) on carcass characteristics and biochemical parameters of broilers. Int J Poult Sci 7:857–861
64. Meluzzi A, Primiceri G, Giordani R, Fabris G (1992) Determination of blood constituents reference values in broilers. Poult Sci 71(2):337–345
65. Mondesire RR (2004) Immunodiagnostics and other emerging diagnostic techniques. In: Thrall MA, Baker DC, Campbell TW, DeNicola D, Fettman MJ, Lassen ED, Rebar A, Weiser G (eds) Veterinary hematology and clinical chemistry. Lippincott Williams and Wilkins, Philadelphia, pp 55–68
66. Nouzarian R, Tabeidian SA, Toghyani M, Ghalamkari G, Toghyani M (2011) Effect of turmeric powder on performance, carcass traits, humoral immune responses, and serum metabolites in broiler chickens. J Anim Feed Sci 20(3):389–400
67. NRC (National Research Council) (1994) Nutrient requirements of poultry. 9th rev. Ed. National Academy Press, Washington, DC. 1994
68. Raskar SS, Bhagat DJ, Agare HR, Desai BG, Chorage NT (2019) Effect of Feeding Turmeric (*Curcuma Longa*) Powder on the Meat Quality of Broilers. Pharma Innov J 8(1):52–55
69. Rizz C, Marangon A, Chiericat GM (2007) Effect of Genotype on Slaughtering Performance and Meat Physical and Sensory Characteristics of Organic Laying Chicken. Poult Sci 86:128–135
70. Ruby AJ, Kuttan G, Dinesh Babu K, Rajasekharan KN, Kuttan R (1995) Antitumor and antioxidant activity of natural curcuminoids. Cancer Lett 94:79–83
71. Patterson JA, Burkholder KM (2003) Application of prebiotics and probiotics in poultry production. Poult Sci 82(4):627–631

72. Peter Haščík L, Trembecká M, Bobko J, Čuboň O, Bučko JT (2015) Evaluation of meat quality after application of different feed additives in diet of broiler chickens. Potravinarstvo® Sci J Food Ind 9(1):174–182. https://doi.org/10.5219/429
73. Petracci M, Baéza E(2009) Harmonization of methodology of assessment of poultry meat quality features. Working paper of WPSA Working Group 5 Poultry Meat July 2009
74. Pietras M (2001) The effect of probiotics on selected blood and meat parameters of broiler chickens. J Anim Feed Sci 10(2):297–302
75. Salminen S, von Wright A, Morelli L, Martean P, Brassart D, de Vos WM, Fonden R, Matills-Sandholm T (1998) Demonstration of safety of probiotic-A Review. Int J Food Microbiol 44:93–106
76. Samarasinghe K, Wenk C, Silva KSFT, Gunasekera JMDM (2003) Turmeric *Curcuma longa* root powder and mannan oligosaccharides as alternatives to antibiotics in broiler chicken diets. Asian Australian Journal of Animal Science 16:1495–1500
77. Shirisha R, Raju K, Sai MVLN, Ravinder RV (2017) Effect of dietary supplementation of probiotic (problend) on Immune Status, biochemical profile and *E. coli* counts in commercial broiler chicken. J Anim Res 7:717–721
78. Siadati SA, Ebrahimnezhad Y, Gh SJ, Shayegh J (2017) Evaluation of the probiotic potential of some native Lactobacillus strains on the growth performance and serum biochemical parameters of Japanese quails (Coturnix Coturnix japonica) during the rearing period. Brazilian J Poult Sci 19:399–408
79. Smith D, Lyon C, Lyon B (2002) The effect of age, dietary carbohydrate source, and feed withdrawal on broiler breast fillet colour. Poult Sci 81:1584–1588
80. Sohail M, Hume M, Byrd J, Nisbet D, Ijaz A, Sohail A, Shabbir M, Rehman H (2012) Effect of supplementation of prebiotic mannan-oligosaccharides and probiotic mixture on growth performance of broilers subjected to chronic heat stress. Poult Sci 91:2235–2240
81. Souza XR(2004) Cartoon characters, carne quality and composição lipídica de frangos de corte criados em sistemas de produção caipira e convencional (tese). Federalidade de Lavras (MG).
82. Souza Xisto R, De, Ressan MC, Peter, Faria (2011) Proximate Composition and Meat Quality of Broilers Reared under Different Production Systems Proximate Composition and Meat Quality of Broilers Reared under Different Production Systems. Brazilian J Poult Sci 13(1):15–20
83. Stankovic I (2004) Curcumin. Chemical and Technical Assessment (CTA). FAO
84. Thrall MA (2007) Hematologia e bioquímica clínica veterinária.Philadelphia. Lippincott, Williams &Wilkins, São Paulo: Roca, p 582
85. Upadrasta A, Madempudi RS (2016) Probiotics and blood pressure: Current insights. Integr Blood Press Control 9:33–42
86. Wakenell PS (2010) Hematology of Chickens and Turkeys. In: Weiss DJ, Wardrop KJ (eds) Schalm's Veterinary Hematology, 6 edn. Blackwell Publishing Company, USA, pp 958–966

87. Yazhini P, Visha P, Selvaraj P, Vasanthakumar P and ChandranV (2018) Dietary encapsulated probiotic effect on broiler serum biochemical parameters. Veterinary World 11(9):1344–1348
88. Zhang L, Barbut S (2005) Effects of regular and modified starches on cooked PSE, normal and DFD chicken breast meat batters. Poult Sci 84(5):789–796
89. Zhuang H, Savage EM, Smith DP, Berrang ME (2008) Effect of Dry-Air Chilling on Warner-Bratzler Shear Force and Water-Holding Capacity of Broiler Breast Meat Deboned Four Hours Postmortem. Int J Poult Sci 7(8):743–748
90. Zhou X, Wang Y, Gu Q, Li W (2010) Effect of dietary probiotic, Bacillus coagulants, on growth performance, chemical composition, and meat quality of Guangxi Yellow chicken. Poult Sci 89(3):588–593

Figures

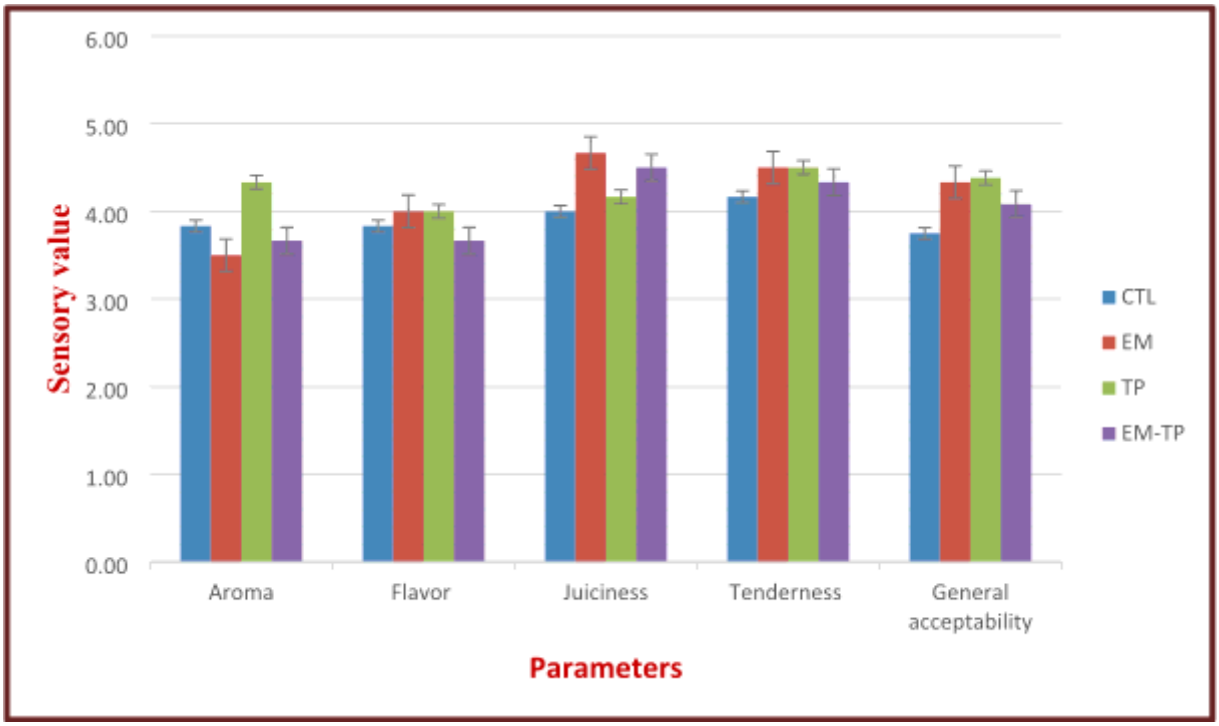

Figure 1

Sensory evaluation of the effect of different feed additives on broiler breast meat

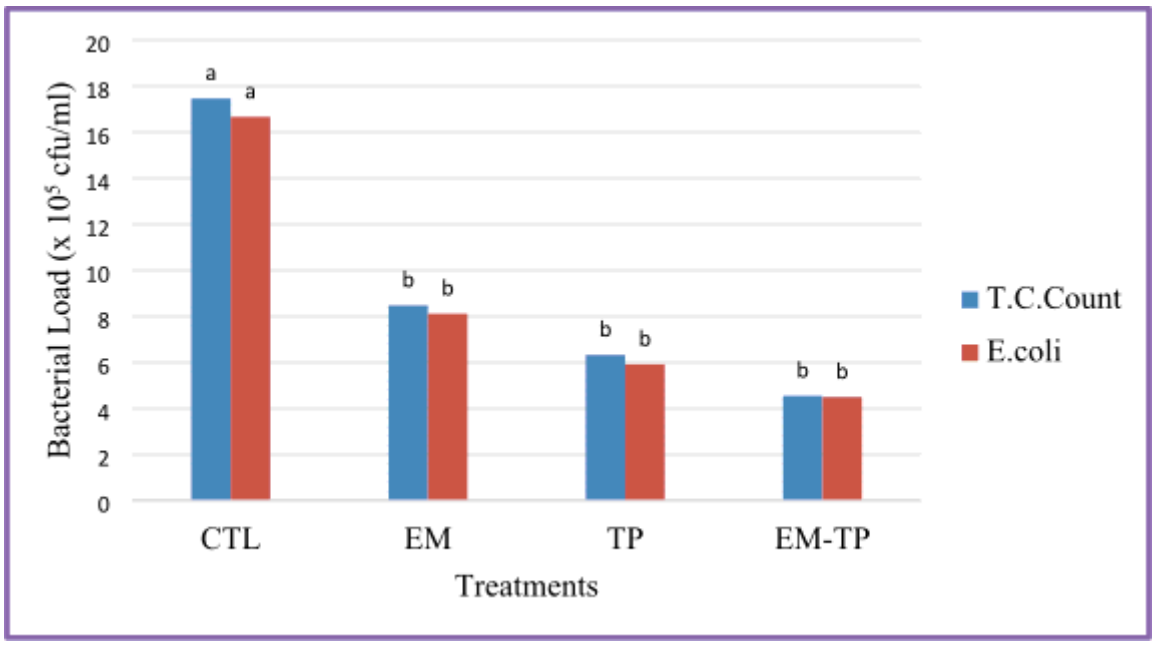

Figure 2

The effect of additives on caecal microbial load counts for total coliform and *E. coli* bacteria