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RESEARCH PAPER

Influence of Phytobiotics and Organic Acid on Nutrient Utilization Efficiency and Carcass Characteristics of Broiler

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Abstract

A total of 120-day old straight run broiler chicks (cobb 500) were randomly assigned into four groups having three replications in each. In control group (To) only mash feed (ME- 3028 kcal/kg, CP- 22.72%) was supplied and in other groups (TN, TM, TC), control + 1% dried Neem leaf powder, Control + 1% dried Moringa leaf powder and control + 1% Citric acid were supplied respectively. The entire period for feeding trial was 28 days. At last day of the feeding trial blood and meat samples were collected. The body weight gain of broilers was observed as the highest in Tc group ($p<0.05$). Feed conversion ratio (FCR) and Metabolizable energy (ME) intake per gram gain (4.63) was lower ($p < 0.05$) in TM than the other groups of birds. Energy efficiency ratio in TM and protein efficiency ratio in TN was higher ($p < 0.05$) than other groups of birds. CP content was high ($p < 0.05$) in TN. Drip loss and cooking loss was lower ($p < 0.05$) in TM than other groups. Based on the findings of the present experiment, it could be concluded that, supplementation of dried Moringa leaf powder has stimulating effect on nutrient utilization and carcass characteristics of broiler and useful phytobiotics for safe and high-quality broiler meat production.

Introduction

The increasing trends of microbial resistance and accumulation of residues in broiler meat due to the use of synthetic antibiotic growth promoters in broiler feed has a bad effect on human health. Broiler producers of most low and middle-income countries use different types of antibiotic growth promoter indiscriminately, which also hampers animal welfare. But it is very important to increase the production of animal protein like broiler to cope with a huge demand. Moreover, producers have become more interested in producing broiler meat using antibiotic growth promoters, as they enhance the production rate as well as reduce the production cost. Nowadays consumers are more interested in safe broiler meat and the market for safe broiler is increasing day by day. So, it is very important to search a new alternative to synthetic antibiotic growth promoters and in that situation, phytogenic feed additive could be one of the best solutions (Ghalamkari *et al.,* 2012; David *et al.,* 2016). Moreover, phytogenic feed additives are non-toxic, natural, free from residual effects (El-Hack *et al.,* 2020; Magdalena *et al.,* 2021) and also enhance digestibility by stimulating the digestive enzyme secretion, boosting up immunity as well as show antiviral effects (Oh *et al.,* 2013; Ognik *et al.,* 2020). Non-nutritive feed additives like antibiotics used in sub therapeutic level in animal production create antimicrobial resistance in human due to the production of residues in animal products (Durrani *et al.,* 2008). Medicinal herb enhances the feed quality and maximizes carcass output in broiler (Ali *et al.,* 2019). Different types of phytobiotics have gained popularity nowadays.

Neem is one the best phytobiotics and many parts of this plants is used for medication. Neem leaves have quercetin, nimbosterol, liminoids which enhance broiler growth as well as feed utilization efficiency

(Prasannabalaji, 2012). Moringa leaves are also responsible for better growth performance of broiler as well as have some effects on meat color and quality (Naji, 2013) which is one of most consumers` preferences. Organic acid like citric acid exhibit similar effects of phytobiotics in broiler (Khan *et al.,* 2016) and enhances broiler performance through modulating gut homeostasis (Sabour, 2019). So, the present experiment was conducted to explore the effects of medicinal herb and organic acid on nutrient utilization efficiency and carcass characteristics of broiler.

Materials and Methods

Feeding trial

Feeding trial was conducted in Shahjalal animal Nutrition Field laboratory, Bangladesh Agricultural University, Bangladesh. A total of 120-day old chicks (cobb 500) were purchased from local distributor and chicks were randomly distributed into four groups having three replications in each group (10 chicks in each replication). Mash feed (Table 1) was offered to chicks which was prepared following NRC (1994).

Table 1: Composition of ration for different dietary group

Ingredient	Amount (%)
Maize	46.50
Protein concentrate	8.50
Rice Polish	10.00
Soybean Meal	29.00
DCP	1.50
Soybean Oil	3.00
Salt	0.50
Lysine	0.25
DL-Methionine	0.25
Vit-Mineral premix	0.50
Total	100.00
Nutrient Composition (Calculated)	
ME (Kcal/kg)	3028
CP (%)	22.72
CF (%)	4.85
Ca (%)	0.68
P (%)	0.31

Vitamin-mineral premix composition: Vitamin A 12,500,000 IU, Vitamin D_3 2,500,000 IU, Vitamin E 20,000 mg, Vitamin K_3 4,000 mg, Iron 40,000 mg Vitamin B_1 2,500 mg, Vitamin B_2 5,000 mg, Vitamin B_6 4,000 mg Nicotinic acid 40,000 mcg, Pantothenic acid 12,500 mg, Vitamin B_{12} 12,000 mcg, Folic acid 800 mg, Biotin 100 mg, Cobalt 400 mg, Copper 10,000 mg, Iodine 400 mg, Manganese 60,000 mg, Zinc 50,000 mg, Selenium 150 mg, Di-Calcium Phosphate 380 gm

The feed for all groups of birds were iso caloric and iso nitrogenous. Moringa (*Moringa olifera*), Neem (*Azadiracta indica*) dried leaf powder and citric acid were purchased locally. In Control group (T_o) only the mash feed was supplied and in other groups, control + 1% dried Neem leaf powder, Control + 1% dried Moringa leaf powder and control + 1% ascorbic acid were supplied in T_N, T_M, T_C group respectively. Birds of all replications were reared in separate pens and that was assigned unbiasedly. Saw dust were used as litter material. Feeding trial was conducted for 28 days and feed and water was supplied ad-libitum throughout the feeding trial. At the age of 4th, 11th and 19th days, ND, IBD and ND booster vaccines were provided through eye. Throughout the feeding trial period, body weight and feed intake were collected on weekly basis.

Collection of samples and analysis

At 29th day of the feeding trial blood samples (5 ml) were collected from unbiasedly selected birds from each replication and centrifuged at 6000 rpm for 10 minutes for serum separation. Separated serum were stored at -20^o C for further analysis of blood metabolites. Then birds were slaughtered and collected meat samples from breast and thigh and preserved the last day of the feeding trial. The parameters include total protein, albumin, globulin, creatinine, urea, calcium, phosphorus, total cholesterol, HDL and LDL in blood were analyzed using commercially available kits following the method of kit manufacturer. Proximate analysis of thigh and breast meat were performed following the method established by AOAC (1995). p^H of meat was measured within 30 minutes after slaughter (initial p^H) and after 24 hours of storage (p^Hu). Moreover, energy efficiency ratio was calculated according to Kamran (2008) as gram of weight gain× 100/total ME intake and protein efficiency ratio was measured following the formula of McDonald (1995).

Statistical analysis

All data were subjected to analysis of variance (ANOVA) in a Completely Randomized Design (CRD) to test the significance of treatment effects and comparison of treatment mean was performed using Tukey's HSD test. SPSS statistical analysis software (SPSS Inc. Chicago, IL, USA) was used for all statistical analysis. Comparison of means was carried out at 5% level of significance ($p < 0.05$).

Results

Nutrient Utilization

Weight gain, FCR, Protein efficiency ratio, Energy efficiency ratio, ME intake per gram gain and CP intake per gram gain was significantly affected in the present experiment. Weight gain was high in T_C group ($p < 0.05$). FCR (1.60) and ME intake per gram gain (4.63) was lower ($p < 0.05$) in T_M group than the other groups of birds (Table 2). Energy efficiency ratio (22.13) in T_M group and Protein efficiency ratio (2.80) in T_N group was higher ($p < 0.05$) than other groups of birds.

Meat quality

In case of breast muscle, significant difference was found in dry matter, crude protein, drip loss and

cooking loss (Table 3). Dry matter was significantly high (29.02%) in neem leaf supplemented groups (T_N). concentration of glucose in blood was high ($p < 0.05$) in control group of birds than others.

Table 2: Growth performance of birds

Parameter	T_0	T_N	T_M	T_C	*p*-value
Body Weight gain	1371^{b}±15	1326^{c}±25	1390^{b}±29	1430^{a}±35	0.021
Feed intake	2413±40	2150±35	2224±28	2384±32	0.210
FCR	1.76^{a}±0.11	1.62^{b}±0.15	1.60^{b}±0.11	1.66^{ab}±0.14	0.014
Protein efficiency ratio	2.55^{b}±0.32	2.80^{a}±0.12	2.78^{ab}±0.18	2.73^{ab}±0.22	0.001
Energy efficiency ratio	18.13^{c}±2.12	19.26^{a}±3.02	22.13^{ab}±2.52	20.68^{b}±2.31	0.033
ME intake per gram gain (kj/g)	5.27^{a}±0.24	4.70^{b}±0.36	4.63^{b}±0.39	4.84^{ab}±0.11	0.009
CP intake per gram gain (g)	0.43^{a}±0.05	0.36^{b}±0.03	0.35^{b}±0.6	0.37^{ab}±0.9	0.019

T_0 = Control group, T_N = Control+ 1% dried Neem leaf powder, T_M = Control+ 1% dried Moringa leaf powder, T_C = Control + 1% Citric acid

^{abc} means bearing dissimilar superscript in same row differ significantly at the level of 5%

Among the four groups of birds, crude protein content was high ($p < 0.05$) in T_N. Like breast muscle, dry matter and crude protein content was high in T_N groups of birds. But ether extract content was significantly lower in citric acid supplemented group of birds in both breast and thigh muscle. Drip loss and cooking loss was lower ($p < 0.05$) in T_M than other groups of birds.

Discussion

Nutrient Utilization Efficiency

Medicinal plant has beneficial effects on broiler growth performance. In this experiment, weight gain of broiler was high in T_C. Demirel (2012) stated that supplementation of citric acid had increased body

Table 3: Carcass characteristics

	T_0	T_N	T_M	T_C	*p*-value
Breast muscle:					
P^H (Initial)	6.20±0.63	6.05±0.32	5.70±0.65	5.78±0.45	0.342
P^Hu (after 24 h)	5.80±0.51	5.97±0.25	5.71±0.33	5.70±0.21	0.411
Dry matter (%)	27.75^{b}±2.2	29.02^{a}±2.4	27.02^{c}±1.89	26.30^{d}±1.7	0.023
Crude protein (% DM)	25.22^{b}±2.4	26.51^{a}±2.4	24.98^{c}±1.5	24.55^{c}±1.69	0.002
Ether extract (% DM)	1.22^{a}±0.12	0.95^{c}±0.09	1.01^{c}±0.05	1.12^{b}±0.02	0.030
Ash (%DM)	1.08±0.12	1.19±0.32	1.15±0.09	1.14±0.08	0.532
Water holding capacity	57±4.03	66±5.12	68±7.01	65±3.22	0.222
Drip loss	15^{a}±2.12	13^{b}±1.65	9^{ab}±2.03	11^{ab}±3.11	0.011
Cooking loss	33^{a}±3.05	32^{ab}±5.18	28^{b}±2.12	30^{b}±2.03	0.001
Thigh muscle:					
P^H (Initial)	5.99±0.53	5.87±0.23	5.18±0.48	5.43±0.54	0.443
P^Hu (after 24 h)	5.70±0.16	5.81±0.31	5.17±0.29	5.33±0.25	0.087
Dry matter (%)	23.56^{c}±2.6	24.22^{a}±1.5	23.86^{b}±1.8	24.03^{ab}±1.7	0.002
Crude protein (% DM)	20.76^{b}±1.8	21.89^{a}±1.6	21.07^{ab}±1.5	21.45^{a}±1.32	0.034
Ether extract (% DM)	1.77^{a}±0.12	1.51^{b}±0.09	1.68^{a}±0.3	1.50^{b}±0.08	0.003
Ash (%DM)	0.95±0.01	1.05±0.08	1.12±0.08	1.19±0.06	0.731
Water holding capacity	59±3.07	63±6.09	65±4.11	61±2.04	0.882
Drip loss	13^{a}±2.10	11^{b}±1.01	10^{ab}±1.20	14^{a}±1.80	0.003
Cooking loss	36^{a}±3.05	30^{ab}±5.04	26^{b}±2.15	33^{b}±2.07	0.047

T_0 = Control group, T_N = Control+ 1% dried Neem leaf powder, T_M = Control+ 1% dried Moringa leaf powder, T_C = Control + 1% Citric acid

^{abc} means bearing dissimilar superscript in same row differ significantly at the level of 5%

Blood profile

Creatinine level was significantly high in control group of birds (Table 4) and lower level was observed in T_M. Total protein (4.40 g/dl) and globulin (2.91 g/dl) were high ($p < 0.05$) in T_M where albumin (2.48 g/dl) were higher in T_N. Calcium and phosphorus level were Cholesterol and glucose level were significantly affected by the inclusion of dried leaf powder and citric acid in the diet. Total cholesterol, LDL as well as LDL/HDL ratio were lower ($p < 0.05$) in T_M. Moreover,

weight gain of broiler and also mentioned that supplementation of citric acid at a level of 3 % can enhance the feed conversion ratio. Moreover, citric acid is an organic acid which create a suitable gut environment for growth enhancing bacteria by reducing the pathogenic bacteria that improve the feed utilization efficiency of broiler (Baghban-Kanani *et al.,* 2019; Hasan *et al.,* 2016). But FCR, ME intake per gram of gain, CP intake per gram of gain were lower and energy efficiency ratio was lower in T_M. Osama

(2020) also found better body weight gain supplementing Moringa seed powder that supports the results of the present experiment. Another researcher, Banjo (2012) conducted an experiment supplementing different levels of moringa leaf (1%, 2% and 3%) and found better body weight gain at 2% moringa leaf supplementation. Inclusion of extruded hemp in broiler feed increases the growth performance by enhancing the availability of certain enzymes for proper nutrient utilization (khan, 2010). Medicinal plant has antioxidant properties, some important amino acids which enhances the production of certain enzymes that increase energy and protein efficiency ratio in broiler. Alpha linoleic acid is high in moringa (Moyo, 2011) and for enhancement in carcass yield, ascorbic and tocopherol of moringa may be responsible (Hekmat *et al.,* 2015; Khan *et al.,* 2012).

capacity in $T_{M.}$ Moreover, decreasing trend in p^H of broiler meat after 24 hours of storage is very high in control and T_C and lower in neem and moringa supplemented group of birds. Faster reduction in p^H makes the meat dry and pale may be due to the reduction of water holding capacity of meat that`s why water holding capacity is high and drip loss and cooking loss is low in T_N and $T_{M.}$ Micronutrient, essential oil and antioxidant of neem increase the utilization of feed protein which ultimately enhances the protein content of broiler meat in $T_{N.}$

Medicinal herb supplementation affects the blood metabolites of broiler and many herbs can reduce the total cholesterol, LDL and trigger HDL production. According to khan *et al.,* 2012, herb can alter the cholesterol production process in liver and convert the cholesterol into bile acid through limiting

Table 4: Blood Metabolites

Parameter	T_0	T_N	T_M	T_C	*p*-value
Creatinine (mg/dl)	0.28 ±0.02	0.23±0.003	0.18±0.05	0.20±0.03	0.076
Total protein (g/dl)	3.90^{c}±0.12	4.20^{b}±0.21	4.40^{a}±0.19	4.28^{b}±0.25	0.001
Albumin (g/dl)	1.59^{bc}±0.23	2.48^{a}±0.15	2.08^{b}±0.19	1.24^{c}±0.14	0.004
Globulin (g/dl)	2.17^{b}±0.32	2.12^{b}±0.12	2.91^{a}±0.12	1.67^{c}±1.09	0.036
Phosphorus (mg/dl)	7.06^{c}±0.71	8.20^{b}±1.11	9.38^{a}±1.22	8.24^{b}±0.89	0.025
Calcium (mg/dl)	4.27^{b}±1.10	4.35^{b} ±2.2	4.76^{a} ±1.6	4.15^{c}±1.10	0.033
Urea (mg/dl)	8.04±0.69	6.19±0.59	5.32±0.62	6.05±0.23	0.092
Uric acid (mg/dl)	4.75±0.71	4.28±0.94	4.25±0.61	4.36±0.32	0.065
Total Cholesterol (mg/dl)	102.9^{a} ±9.12	92.85^{b} ±8.35	76.18^{c}±2.73	85.9^{bc} ±6.57	0.002
HDL (mg/dl)	90.23±5.53	82.75±6.21	95.61±3.98	86.66±6.11	0.068
LDL (mg/dl)	79.34^{a}±4.76	65.67^{b}±3.55	35.55^{c}±4.11	45.48^{bc}±3.05	0.002
LDL/HDL	0.88^{a}±0.22	0.80^{a}±0.17	0.37^{c}±0.05	0.52^{b}±0.08	0.001
Glucose (mg/dl)	170.43^{a}±0.58	150.65^{b}±0.33	135.71^{bc}±0.21	140.51^{c}±0.71	0.041

T_0 = Control group, T_N = Control+ 1% dried Neem leaf powder, T_M = Control+ 1% dried Moringa leaf powder, T_C = Control + 1% Citric acid

^{abc} means bearing dissimilar superscript in same row differ significantly at the level of 5%

Carcass Quality and Blood Metabolites

Meat p^H is correlated to meat color and this is an important factor for consumer preference (Kostadinovic *et al.,* 2015). Low meat p^H is responsible for acidic meat as well as dark, firm and dry meat results from higher p^H of meat (Laudadio *et al.,* 2011). Moreover, according to Tashla (2019) normal p^H for broiler meat is 5.6 to 6.1 and in the present experiment, p^H is within the range but have no significant effects of medicinal herb and organic acid supplementation on meat p^H. In the present experiment, p^H level is not significantly affected by the supplementation of medicinal herb and citric acid but numerically lower in $T_{M,}$ but water holding capacity is high in both thigh and breast meat than control. The reason behind that may be the p^H value of meat was within the range. Puvaca (2011) stated that, lower p^H which means acidic meat have lower water holding capacity which supports the result of the present experiment. Young (2003) also reported that antioxidant enhance water holding capacity of meat and in this experiment, antioxidant present in moringa leaf may be responsible for higher water holding

the action of HMG-CoA reductase and fatty acid synthase. Furthermore, Balami (2018) also found low total cholesterol, triglycerides and LDL in broiler blood serum where moringa is supplemented which is similar to our findings. High concentration of polyphenol, flavonoid, phenolic compound in Moringa shows hypercholesterolaemic effects (Verma *et al.,* 2009) and high fiber of moringa also limit absorption of triglycerides and cholesterol from intestinal tracts (Mandal *et al.,* 2014). Flavonoid and alkaloids of hemp also helps in lowering the LDL cholesterol (Ramadan *et al.,* 2007). Moringa can prevent the catabolism of protein by limiting the secretion corticosterone which ultimately enhances protein level in blood (Luqman *et al.,* 2012). The increase in serum protein level is the reflection of maximum metabolism of feed protein (Sirvydis *et al.,* 2006) and Teye (2013) was found more serum protein level in broiler than control group when supplementing moringa leaf. The relation between serum urea and protein concentration is vice versa. When the concentration of serum protein is high, urea level becomes low and the reason behind that is the efficient absorption and utilization of dietary protein

that may due to the presence of micronutrient in moringa (Hussein *et al.,* 2019). Moringa is a rich source of mineral and protein and for this reason, calcium and phosphorus level in blood serum is higher in $T_{M.}$

Conclusion

The most important challenge in present broiler industry is to produce safe broiler, fulfilment of the consumer preference along with the reduction of production cost as well as cope up the demand of broiler meat. In this situation, inclusion of phytobiotics in broiler ration is very impressive. Phytobiotics like neem and moringa leaf can be used in broiler feed for the safe and more nutritious broiler meat.

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