EFFECT OF BLACK PEPPER POWDER SUPPLEMENTATION ON BLOOD AND SERUM PROFILE IN BROILER BIRDS

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Received: 16.11.2021; Accepted: 27.10.2022

ABSTRACT

A feeding trial was conducted with 300 broiler birds to study the effect of dietary supplementation of black pepper at various levels. The birds were distributed according to completely randomized design in 6 treatment groups with five replicates, hence there were 10 birds in each replicate. The Experiment lasted for 42 days and included 3 phases; pre-starter phase (0-7d), starter phase (7-21d) and finisher phase (21-42d). The negative control group (T1) was offered basal diet containing maize- soybean meal which was formulated as per specifications of Bureau of Indian Standards, 2007 to accomplish the metabolizable energy (ME) and crude protein requirements of broilers. The T2 group was kept as control group and offered basal diet with antibiotic while T3, T4, T5 and T6 groups were offered 0.25, 0.5, 0.75 and 1% black pepper powder respectively along with basal diet. On completion of feeding trial, one bird from each replicate was selected randomly for sampling. The blood was collected in EDTA vials for blood parameters and serum was collected separately to analyse the serum biochemistry. Highest Hb, TEC and TLC contents were found in T4 group (10.49%) followed by T2 (10.34%) and was significantly (P<0.05) higher than negative control group (9.42%). TEC and TLC contents were also highest in T4 group. Cholesterol, triglycerides and LDL levels were significantly (P<0.05) reduced and HDL levels were significantly (P<0.05) improved in black pepper powder supplemented groups as compared to negative control and control group. The highest values of HDL were found in T4 group. Thus, black pepper powder supplementation led to improved blood and serum profile in terms of increased Hb, TEC and TLC and decreased cholesterol, triglycerides and LDL.

Keywords: Black pepper powder, Broilers, Serum cholesterol, Triglycerides

How to cite: Gunjan, Sihag, S., Walia, R. and Swaroop, R. (2022). Effect of black pepper powder supplementation on blood and serum profile in broiler birds. *Haryana Vet.* **61(2)**: 247-249.

There is scarcity of feed in terms of quantity as well as quality, it is essential to make maximum utilization of the available feed. The concept of feed additives is introduced here. Feed additives are ingredients or combination of ingredients used in micro-quantities in diet to fulfill the specific needs (Reddy, 2016). The advancement of biotechnology has enhanced and eased the availability of these feed additives.

The most common antimicrobial feed additives being used are antibiotics. But their exorbitant use has raised many concerns which has become focus of interest for the scientific community now. The most common alternatives under consideration are phytogenics or phytobiotics. Phytogenics are plant extracts which possess various pharmacological activities viz. antimicrobial, antioxidant, anti-inflammatory, digestive stimulant (Akyildiz and Denli, 2016). Terpenoids, phenolics, glycosides and alkaloids are minor metabolites contained in plant extracts as alcohols, aldehydes, ketones, esters, ethers, and lactones (Parganiha et al., 2011). Black pepper is a spice being used routinely in an Indian kitchen. It belongs to Piperaceae family. It constitutes many active components such as piperine, cupsaeesin, cupsisin and cupsantine (Barad et al., 2017). Piperine has been shown to improve the therapeutic efficacy of a variety of drugs and nutrients by increasing oral bioavailability and inhibiting a

number of metabolising enzymes and it also increases the vaccine efficacy (Johnson *et al.*, 2011). Various studies have proven that it enhances feed intake, body weight gain and other growth parameters are improved significantly (P< 0.05).

MATERIAL AND METHODS

Three hundred, one day old broiler chicks, were purchased from a local commercial hatchery. Randomly distributed into six dietary treatments with five replicates per treatment and each replicate has ten birds hence there were 30 subgroups. Basal ration was formulated as per BIS (2007) to fulfill the metabolizable energy (ME) and crude protein requirements of birds. The composition of various diets are listed in table 1. The first group was kept as negative control (T1) and given a basal diet without antibiotics, while the second (T2) received a basal diet with antibiotics and kept as control group, while in the third (T3), fourth (T4), fifth (T5) and sixth (T6) groups antibiotic was substituted with 0.25, 0.5, 0.75, and 1% black pepper powder in the diet, respectively. The birds in the study were raised in a deep litter system. The feed intake, body weights and FCR was recorded weekly. At the end of feeding trial (42 days), one bird from each replicate was selected randomly for metabolic trial and sampling. Blood samples were collected from the slaughtered birds. About 2 ml of blood was collected from each bird via

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brachial wing vein puncture using sterilized syringes and 5 ml scalp vein needle set into vacutainer containing EDTA for hematology and in non-heparinised tubes for serum. The samples were centrifuged at 3000 rpm for 15 minute and serum obtained was stored at -20°C until analysis. Serum samples were analysed for different serum variables like total cholesterol, triglyceride, HDL (high density lipoprotein) and LDL (low density lipoprotein) using autoanalyser. The results were evaluated using the general linear model protocol of the statistical package for social sciences 20th edition (SPSS) and the means were compared using Duncan's multiple range test (DMRT) with significance set at P<0.05.

RESULTS AND DISCUSSIONS

The mean values of results of blood analysis are shown in Table 2. Mean values of Hb (g/dl) ranged from 9.42 g/dl (T1) to 10.49 g/dl (T4) and findings of present investigation exposed that hemoglobin level in control group of broilers and broilers fed ration supplemented with 0.50% BP were at par, however, significantly higher (P<0.05) as compared to negative control, T3 (0.25% BP) and T6 (1.00% BP). Mean values of RBC (red blood cell), PCV and WBC (white blood cell) were found significantly (P<0.05) higher in 0.5% (T4) and 0.75% (T5) black pepper supplemented group as compared to other treatments, antibiotic supplemented group and negative control group. The increase in PCV, Hb, RBC content of the blood of the birds fed the test ingredient is an indication of improved oxygen carrying capacity of the cell which translated to a better availability of nutrient to the birds consequently affecting their well being. The present data are in harmony with those obtained by Singh et al. (2018); who reported that hemoglobin levels were increased in birds fed with black pepper supplemented diets as compared to control group. Barad et al. (2016) have shown in their study that Hb, PCV, TEC and TLC are numerically improved but there are non-significant differences obtained by feeding coriander seeds, turmeric and black pepper in diet. Similarly, the present findings are corroborated with earlier research of Sidhu et al. (2018), who reported that feeding black pepper @ 0.5% in diet can significantly (p<0.05) increase the Hb and PCV levels. On the other hand, Shahverdi et al. (2013) concluded that hemoglobin and PCV levels were lowered due to feeding of pepper in diet. Al-Kassie et al. (2011) also showed that treatment groups fed black pepper had significantly (P<0.05) lowered RBC, PCV and Hb as compared with the control group, but had no significant effect on WBC among treatments.

The mean values of results of serum analysis are shown in table 3. Total cholesterol concentration in serum

Table 1. Ingredient composition of basal diets fed to broilers during different phases of growth

Ingredient (kg/100 of feed)	Pre starter	Starter	Finisher
Maize	53.0	53.0	57.0
Soybean meal	21.0	18.5	16.5
Groundnut cake	13.0	14.5	14.0
Fish meal	7.0	7.0	5.0
Vegetable oil	4.0	5.0	5.5
Mineral mixture	2.0	2.0	2.0
Total	100.0	100.0	100.0

of broilers under dietary treatments T4 (0.50% BP), T5 (0.75% BP) and T6 (1.00% BP) did not differ significantly among themselves, but were significantly (P<0.05) low as compare to control groups T1 & T2 as well supplemental group T3 (0.25% BP). Serum cholesterol of dietary treatment group T4 was lowest and negative control group T1 being highest. The serum triglycerides level of different dietary treatments supplemented with either level of black pepper were statistically lower (P<0.05) compared to control groups T1 (negative control) and T2 (positive control). As the black pepper supplementation level increased from 0.25 to 1.00%, serum triglyceride concentration decreased.

Dietary supplementation of black pepper powder significantly (P<0.05) improved the HDL value over the control and negative control group. The serum HDL concentration increased as the level of black pepper increased up to 0.75 % level. Serum LDL levels of dietary treatments T4, T5 and T6 had significantly (P<0.05) lower serum LDL level that negative control T1 and positive control T2. Serum LDL in 1.0% black pepper powder supplemented group T6 was lowest and negative control group T1 being highest. The result of the study revealed that replacement of antibiotics with black pepper in the diet of broilers increased serum HDL concentration and decreased LDL concentration. The inclusion of black pepper @ 0.50% of ration in the diets of broiler chickens elicited no adverse effect on hematological and serum biochemical parameters, thus, can be used as replacement for antibiotics. These results were in agreement with findings of Shahverdi et al. (2013) who reported that feeding pepper significantly (P<0.05) affected triglycerides and cholesterol levels of blood. They explained it that the pungent compound of piper nigrum especially piperine increases the production of saliva and gastric secretions (Herati and Marjuki, 2011). Also free gallbladder acids attach to bacteria and fibers and this can increase the excretion of them. Deconjugation of gallbladder acids in small intestine can affect control of serum cholesterol, while deconjugated acids are not capable to solve and absorb fatty acids as conjugated acids. As a consequence,

Table 2. Mean values of hematological parameters of birds under different dietary treatments

Treatments	Hemoglobin g/dl	RBC×10 ⁶ /μ1	WBC×10 ³ /μ1	PCV%
	9.42 ^d ±0.04	1.88 ^{bc} ±0.04	26.91°±0.08	28.38°±0.13
T_2	$10.34^{ab} \pm 0.03$	$1.97^{b}\pm0.02$	28.27 ^b ±0.02	29.99 ^b ±0.09
T_3	$9.94^{\circ}\pm0.12$	$1.94^{b}\pm0.01$	$27.63^{\text{bc}}\!\!\pm\!0.04$	$29.96^{b}\pm0.20$
T_4	$10.49^a \pm 0.06$	$2.15^a \pm 0.01$	$28.96^{a}\pm0.03$	$31.54^a \pm 0.07$
T_5	$10.21^{b} \pm 0.05$	$1.96^{\rm b} \pm 0.02$	27.98 ^b ±0.05	$29.82^{bc} \pm 0.13$
T_6	$9.84^{\circ} \pm 0.07$	$1.86^{\circ}\pm0.02$	$26.95^{\circ} \pm 0.08$	$27.13^{d} \pm 0.50$

Table 3. Mean values of total cholesterol, triglycerides, HDL and LDL content in serum of broilers under different dietary treatments

Treatments	Cholesterol (mg/dl)	Triglycerides (mg/dl)	HDL (mg/dl)	LDL (mg/dl)
$\overline{T_1}$	136.20°±2.58	118.60°±2.30	46.83 ^b ±0.89	69.10 ^a ±1.94
T_2	$133.80^{a}\pm3.96$	$113.60^{a}\pm1.81$	48.12 ^b ±1.05	$66.84^{ab}{\pm}1.54$
T_3	125.60 ^b ±3.36	$105.60^{b} \pm 1.78$	55.63°±2.12	$68.88^{a}\pm2.14$
T_4	$111.40^{\circ}\pm2.70$	90.60°±1.30	58.71°±0.95	$64.76^{bc} \pm 1.79$
T_5	$115.00^{\circ}\pm2.91$	$92.00^{\circ} \pm 1.14$	59.01 ^a ±1.11	62.35°±1.23
T_6	114.80°±3.34	$91.40^{\circ} \pm 1.67$	$57.62^a \pm 4.86$	$58.60^{d} \pm 1.30$

they prevent from absorption of cholesterol. Ghaedi et al. (2014) concluded that Triglyceride and cholesterol concentration decreased in black pepper supplemented group while HDL increased in the same group. Myandoab and Mansoub (2011) have revealed a significant reduction in levels of cholesterol and triglycerides on feeding of Piper nigrum in diet and the main reason of decrease in level of cholesterol and triglycerides in blood of quails is substances like carvacrol and tymol which are present in herbs. Al-Kassie et al. (2011) also showed lowered cholesterol and H/L ratio. H/L ratio is indicator of stress hence feeding black pepper in diet reduces stress level in birds. The findings of our results are also supported by Mansoub (2011) who have shown lowered cholesterol, triglycerides, LDL and increased HDL levels. Unlikely, Abou-Elkhair et al. (2014) reported that the concentration of serum albumin, cholesterol and GOT were not significantly different compared to those of the control group but the studied supplement combinations (coriander, turmeric and black pepper) were effective in decreasing serum total triglycerides.

CONCLUSION

Hence, as an interpretation of these results, it can be inferred that 0.5%Black pepper powder can be an effective substitute of antibiotic growth promoter in broiler ration that resulted in increased hemoglobin content, erythrocyte count, lowered cholesterol, trigycerides, LDL and increased HDL content in that group as compared to negative control

and control group. The main active components of black pepper such as piperine stimulate digestive enzymes secretion and increase flow of saliva that can improve feed efficiency and overall health, also increased enzymatic activity causes less absorption of fatty acids leading to lowered cholesterol.

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