J. Sylhet Agril. Univ. 11(2): 29-34, 2024 *DOI: https://doi.org/10.3329/jsau.v11i2.82729*

ISSN: 2308-1597

Journal of the Sylhet Agricultural University

Journal home page: http://www.jsau.sau.ac.bd

Research Article

COMPARATIVE STUDY OF LICORICE POWDER, ANTIBIOTICS AND PROBIOTICS ON BODY WEIGHT AND BLOOD PROFILES OF BROILER CHICKEN

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Article info

Article history

Received: 19.09.2024

Accepted: 20.11.2024

Published: 30.12.2024

Keywords

Licorice powder, antibiotics, probiotics, body weight, blood profiles, broiler

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Abstract

The use of antibiotics in the broiler production has elevated concerns due to their potential negative impact on the health of human. Initial studies suggest that medicinal plants may offer a safer alternative, though limited research has been conducted in Bangladesh. The aims of the present experiment were to compare the impacts of licorice powder, antibiotics and probiotics on the growth, vital organs, and blood profiles of broiler. About 180-day-old *Lohman Meat* chicks were obtained from the Quality Hatchery Limited and equally divided into four groups with three replicates per group, and each replicate consisting fifteen chicks) using a rearing floor litter system. A basal diet was supplied during experiment; T0, control group- no feed additive supply, and treatment group; T1, - 1gm Licorice Powder (LP)/1L of drinking water (DW); T2- 1 gm of antibiotics/1L of DW and T3- 1gm of probiotics/1L of DW. Body weight of each individual birds was recorded weekly, FCR, organ weights, and carcass yields were calculated at the end of the experiment. Blood sample collected from birds on day 35 were analyzed for blood profiles. Data were analyzed using single-factor ANOVA at a 5% significance level. Body weight significantly varied by groups within each of defined days 14, 21, 28, and 35 ($p<0.05$), with the highest body weight gain in the T1 (licorice powder) group ($p>0.05$). The FCR was significantly lower in the T1 treated group and T3 treated group ($p<0.05$). There was no variation found in the change of various organ weight among treated and untreated group of broiler chicken. The hematological parameters were significantly greater in the treated group T3, followed by T1 and lower in the T2 groups compared to the control group T0 ($P>0.05$). The mean value of ALT was significantly lower in T3 (7. 16 u/l), followed by T2 (7.88 u/l) and T1 (7.94 u/l) than T0; ALP was lower in T1, T2, T3 respectively, total protein, cholesterol. The mean value of HDL was significant higher in T3 (68.35mg/dl) followed by T2 (60.31mg/dl) and T1(46.79mg/dl) than the T0 group ($p> 0.01$). The administration of licorice powder (T1) and probiotics (T3) resulted in positive impacts on body weight, organs weight and blood profiles of broiler chicks compared to antibiotics (T2). Licorice powder may be suggested as a suitable substitute to antibiotics in poultry production.

Introduction

Nowadays, access to wholesome food sources is becoming one of people's top worries as a result of the growing population. Proteins are essential for maintaining a healthy daily diet. In the meantime, meat from cattle and poultry is a crucial source of protein for human nutrition. The preference for poultry meat among consumers is increasing, driven by the rising incidence of cardiovascular diseases and diabetes linked to the overconsumption of unhealthy foods, such as red meat. Poultry has recently become one of the world's most popular animal proteins. Due to its affordability, rich nutrient content, adherence to various religious dietary restrictions, and widespread availability and acceptability. Hence, poultry business has advanced a lot in current years across the world. It is obvious that many issues, including sickness, have a negative impact on the profitability of the poultry sector (Alpatay, 2021).

Cite This Articale

Hossain MS, Rahman MM, Hasan MMI, Begum S and Islam S. 2024. Comparative Study of Licorice Powder, Antibiotics and Probiotics on Body Weight and Blood Profiles of Broiler Chicken. J. Sylhet Agril. Univ. 11(2): 29-34, 2024. https://doi.org./.....

In the poultry industry, antibiotics are used as a prophylactic to prevent diseases and maintain poultry health (Butaye *et al.,* 2003). Numerous farmers have started adding antibiotic as Antibiotic Growth Promoters (AGPs) to animal diets to improve feed conversion efficiency, control or kill harmful bacteria, and lower mortality rates (Suresh *et al*., 2018). Recent evidence indicates that this practice may threaten human health, as AGPs residues in chicken have been shown to disrupt intestinal microbiota and contribute to the development of the resistant bacteria against antibiotics (Ahmed et al., 2022). Technical boards in certain nations have therefore prohibited the practice of AGPs for non-medicinal purposes due to concerns about potential long-term impacts (Ajuwon, 2016; Castanon, 2007).

Medicinal plants have earned widespread recognition for their numerous beneficial uses in poultry, animal, and human health (Fukai *et al*., 2002a). Medicinal plant has shown potential as replacements to AGPs in poultry diets. Licorice (*Glycyrrhiza glabra),* acts as therapeutic remedy for over four thousand years (Fuhman *et al.,* 2002). Studies have shown that licorice possesses antimicrobial, antioxidant, anti-inflammatory properties, anti-nephritic, and free-radical binder properties (Sato *et al*., 2000; Fuhman *et al.,* 2002; Fukai *et al.,* 2002a). These studies suggested that Low fat oxidizers (LFO) helped reduce growth by lowering the mass of fat in humans. Aoki *et al*. (2007) demonstrated that LFO decreased growth and white adipose tissue of abdominal in mice on fat-rich diets. Additionally, it was reported that licorice hydrophobic oil reduced abdominal fat in both humans and mice (Nakagawa *et al.,* 2004). Plant powdered oils comprising terpenoids may improve poultry health and productivity (Wallace et al., 2010). While the impacts of licorice powder have been shown in vitro and in laboratory animals (Yin et al., 2022) , its impact on poultry performance has yet to be documented. There were no previous studies in Bangladesh to assess the usefulness of licorice on poultry --- Therefore, the aims of this study were to compare the impacts of licorice powder with antibiotics and probiotics on the body, organs weight and blood profiles in broiler chicken.

Materials and Methods

Experimental birds and management

A 180 broiler chicks (*Lohman-Meat)* were purchased from a reputed commercial hatchery Ltd and divided into four groups with three replications, which consisted of fifteen chicks per replication. The T0, control group was given a basal diet, while T1, treatment was supplied with licorice powder (1gm/L of DW, T2 was supplemented with Ciprofloxacin at 1.0gm/L of DW, and T3 was given probiotics at 1gm/L of DW. The *ad-libitum* feeding schedule included a commercial starter diet with 23% crude protein and 3100 KCal ME/kg of diet, and 21% CP and 3050 KCal ME/kg of grower diet. All birds reared in a floor with proper maintained of humidity (55-58%) and temperature (20-22^{0}C) in conventional open housing system and proper vaccination schedule was followed such as BCRDV, ND IBD and IB as well.

Determination of body weights, organs weights and carcass yields

Body weight was measured every seven days throughout 35 days of the experiment and feed conversion ratio was calculated after the end of the experiment. On day 35, three broilers from each replicate were selected simple randomization, weighed and then humane slaughtered. After that, all feathers, feet's, shanks, and viscera were removed from those broilers, and dressed and weighed. Dressing percentage were calculated by following the standard formula (Ref) . Lastly, each vital organ like liver, lungs, and heart were weighed.

Assessment of blood profiles

At the end of the trial, about 2mL of blood sample was drowned in the wing vein of each of five randomly selected broiler chickens per replicate and kept it into Ethylene Diamine Tetra Acetic Acid (EDTA) containing blood tube for hematological assessment, were analyzed using an Auto Veterinary Hematology Analyzer (PE 7010 VET) at the department of Physiology at Sylhet Agricultural University. Additionally, Serum samples were separated from the blood samples obtained and used for biochemical assessment by using a semi-automated chemistry analyzer (Model-), following the instructions provided with the specific reagent kits.

Statistical analysis

Data obtained were entered into MS excel sheet Data sorting, cleaning and coding were done here before transporting to IBM SPSS Statistics 22 software statistical analysis. One-way ANOVA was performed to assess the variation among groups. The results were as mean, min, max, 95% confidence interval and p value (5% level of significance)

Results

Body weight gain and FCR

The mean value of body weight was no significant variation on day 1 and 7, as shown in Table 1. However, the growth of broiler had a significant variation among groups on the other comparative days (14, 21, 28, 35) ($p<0.05\%$). The greater body weight gain was recorded in the licorice powder supplemented group than in the probiotics supplemented group or the antibiotic-treated group of broiler chickens ($p<0.05\%$) (Table 1).

Table 1. Effect of Licorice powder, antibiotic and probiotics on Body Weight and FCR

Body wt (Kg)	Control	Treatment (Mean± SD)			P- value
	T0	T1 (Licorice Powder)	T2 (Antibiotics)	T3 (Probiotics)	
14th days	0.50 ± .039	0.78 ± 0.060	0.74 ± 0.063	0.76 ± 0.064	0.001
21 Days	0.95 ± 0.04	1.30 ± 0.089	1.27 ± 0.088	1.32 ± 0.11	0.001
28days	1.43 ± 0.08	1.93 ± 0.108	1.86 ± 0.132	1.83 ± 0.136	0.001
35days	1.78 ± 0.14	2.13 ± 0.277	2.12 ± 0.23	2.06 ± 0.191	0.001
FCR (%)	1.7875	1.57	1.62	1.5775	0.001

Carcass and organs weight

The findings of the effects of licorice powder, antibiotic and probiotics on organs and carcass weight of broiler have been showed in Table 2. The mean value of carcass weight differed in each of treated groups compared to control groups. The highest value of the liver weight was recorded in the T2 (antibiotic) followed by T3 (probiotics) and T1 (licorice powder) groups. However, the mean value of heart and lungs weight was found higher in the T1 (licorice powder) than the T2 (antibiotics) and T3 (probiotic) groups.

Table 2. Effect of licorice powder, antibiotic and probiotics on carcass weight and organ's weight of broiler chicken

Organ Name	Control	Treatment (Mean ± SD)			P value
	T0	T1 (Licorice Powder)	T2 (Antibiotics)	T3 (Probiotics)	
Carcass wt.(kg)	1.34±0.058	1.54±0.11	1.5±0.1	1.46±0.058	0.8
Liver (gm)	51.69± 3.84	52.72± 5.91	56.21±7.26	56.212±5.24	0.48
Heart (gm)	13.66±2.51	14.01±3.04	15.33±1.154	15.66±2.30	0.4
Lungs (gm)	13.33±1.52	14.66±2.51	11.66±2.089	13.33±3.51	0.7

Blood profiles

The impacts of licorice powder, antibiotics and probiotics on hematology of broiler have been showed in Table-3. The mean value of Total Erythrocyte Count (TEC) was significantly greater in T3 treated groups followed by T1, while lower in T2 treated groups, compared to control groups. The most value of TEC had in T3 group added in diets, whereas the least value noted in T2 treated groups. Hemoglobin concentration of blood was significantly greater in T3 groups followed by T1 group, compared to T0 groups. The percentage of Hematocrit (HCT) was considerably higher in treated groups related to control groups.

Table 3. Effect of Licorice powder, antibiotic and probiotics on hematological parameters of broiler

Parameters	Control T0	Treatment ((Mean ± SD)			P-value
		T1(Licorice Powder)	T2 (Antibiotics)	T3 (Probiotics)	
TEC (x10^6 µl)	2.486±0.162	3.273±0.080	2.446±0.102	3.86±0.404	0.001
TLC (x 10^3 µl)	115.926±3.98	142.866±2.04	133.666±6.07	145.2±2.75	0.001
Hb (gm/dl)	10.166±0.51	11.133±0.305	10.026±0.32	11.646±0.706	0.05
HCT (%)	28.566±1.101	41.366±1.861	38.446±1.422	45.976±4.694	0.001

The average value of biochemical parameters has been showed in Table-4. The average value of Alanine Transaminase (ALT), Alkaline phosphatase (ALP), Total protein (TP), Cholesterol, and High-density lipoprotein (HDL) presented significant variation (P>0.05) among groups. The results revealed a decrease in ALT among the treated groups compared to untreated groups. The mean value of alkaline phosphatase was least in T1 and T2 treated group, while the highest value was noted in T3 group compared to T0 group. The value of serum total protein was remarkably greater in T3, whereas lower in T1 and T2 group. Moreover, in case of cholesterol, it was significantly lower in T3 groups, followed by T2 groups and T1 groups compared to T0 group. In addition, HDL found the significant higher in T3 groups than other groups of broilers.

Table 4. Effect of Licorice powder, antibiotic and probiotics on biochemical parameters

Parameters	Control T0	Treatments (Mean ± SD)			P-value
		T1 (Licorice Powder)	T2 (Antibiotics)	T3 (Probiotics)	
Alanine Transaminase (U/L)	21.31±1.83	7.94±1.348	7.88±2.90	7.16±2.104	0.001
Alkaline Phosphatase (U/L)	238.96±12.023	231.92±13.55	231.48±16.55	315.26±68.18	0.05
Total Protein (gm/d)	2.72±0.126	3.60±0.071	3.90±0.154	2.94±0.163	0.001
Cholesterol (mg/dL)	129.03±8.670	120.62±8.39	108.74±10.06	93.89±5.715	0.05
HDL (mg/dL)	31.61±4.448	46.79±4.453	60.31±6.050	68.35±4.678	0.001

Discussion

The impacts of of licorice powder, antibiotics and probiotics were assessed on body weight gain, organ weight, and blood profiles in boiler chicken. The highest body weight gain was detected in the licorice powder supplemented group, then the probiotics supplemented group, and the antibiotic-treated group of broiler chickens. These results correspond to number earlier studies in Bangladesh (?) (Rashidi *et al*.,2019; Naser *et al*., 2017; Shah *et al*., 2021; Ramlucken *et al*., 2020 and Samli *et al*., 2007). Better FCR found in T1, T2 and T3 which are supported by (Zulkifli *et al.,* 2000 and Ahmed *et al*., 2014). However, some studies found poorer FCRs with probiotics and antibiotics (Bai et al. 2013).

There were no significant variations the average value of carcass and organ weights between treated groups and untreated group of broiler chicken (Table 2). Similar results were noted in other studies (Sarangi *et al.,* 2016; Awad *et al*., 2009). However, a study was observed by (Hussein and Selim, 2018) shown that a probiotic cocktail with yeast increased body weight and carcass output better than a single.

The mean values of TEC, TLC, Hb concentration, and HCT showed a significant increase in T3 followed by T1 and T2 treated groups ($p<0.05$), that is similar with findings from other studies by (Al-Kassie *et al.,* 2008 and Nosrati *et al.,* 2017). Additionally, the TLC count was significantly greater among groups ($p<0.05$). These results collectively advocate that probiotics enhance poultry immunity and better health as a whole, as reported by Shah *et al*., 2021.

We observed significant reductions in ALT and total cholesterol levels, with notable differences between the groups, while the mean value of HDL increased. These findings are fully aligned by Li *et al.,* 2011 and Ahmadi, 2011) on cholesterol, who reported significant decreases in plasma cholesterol levels in ducks nourished *B. subtilis* and in broilers supplied the yeast. Since licorice extract contains substantial amounts of flavonoids and ascorbic acid, its addition to the diet led to increased HDL concentration and an improved HDL/LDL ratio in the blood, as noted by Naser *et al*., 2017). Moreover, supplementing broilers' drinking water with 0.4% Licorice Extract increased plasma HDL levels while reducing alanine aminotransferase (ALT) levels ($p<0.05$), as reported by Salary *et al.* 2014).

One of the main limitations of this study is environmental factors such as temperature, humidity, feed composition, and management practices can affect the results. We used to standardize environmental conditions, use controlled housing, and ensure uniform feeding programs across all treatment groups and a randomized block design to lessen environmental bias. Another constraint was the use of antibiotics in broilers may contribute to antibiotic resistance, which poses a threat to human and animal health and to solve it by proper monitor and analyze antibiotic resistance patterns and compare them with the effectiveness of probiotics and licorice as alternatives. Established scientific guidelines were followed for dosage application. Ethical guidelines were followed for animal research, use minimal stress techniques, and train personnel in humane handling methods.

While probiotics and licorice are considered natural alternatives to antibiotics, their cost-effectiveness for large-scale poultry farming remains uncertain. We did not conduct cost-benefit analysis to determine if the use of licorice powder and probiotics can provide economic advantages over antibiotics.

Though the study provides valuable insights into alternatives to antibiotics for enhancing broiler performance, limitations such as environmental variability, dosage optimization, antibiotic resistance risks, and economic feasibility must be carefully considered to draw accurate and applicable conclusions.

Conclusion

Among these treatments, licorice powder and probiotics showed the most progressive impact on the broiler production (insert some significant findings here), and they could be effective replacements to antibiotic growth promoters in poultry feed, particularly for farmers in Bangladesh, where licorice powder is readily available and convenient. Future research exploring the combination of licorice powder with other probiotics to assess their impact on chick development will offer further insights into their overall effectiveness.

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