

COMPARATIVE EFFECT OF ANTIBIOTICS VIS-A-VIS FENUGREEK SEED POWDER (*TRIGONELLA FOENUM-GRÆCUM* L.) ON SERUM BIOCHEMICAL PROFILE OF BROILERS

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Abstract: The feeding trial of six weeks in broiler chicks (n=160) was conducted which were subjected to 4 treatments and designated as treatment T₀, T₁, T₂ and T₃ respectively. All the broiler chicks were fed with starter ration up to 21 days and finisher ration from 22 to 42 days of age as per BIS (1992) specification. The chicks fed with basal diet in control group (T₀), while chicks in treatment T₁ was fed with Antibiotics viz. Zinc bacitracin and Salinomycin @ 20 and 60 mg/kg of feed respectively, T₂ and T₃ were fed basal diet with Fenugreek seed powder @ 1.0 and 1.5 %, respectively. All the birds were given isocaloric and isonitrogenous diets throughout the experimental period. The data were analyzed using General Linear Model procedure of statistical package for social sciences (SPSS) 20th version and means were compared using Duncan's multiple range test (1955) and significance was considered at (P<0.05) level. The supplementation of fenugreek seed powder resulted in decreased level of total cholesterol and LDL, whereas HDL content was increased as compared to antibiotics and control group. The serum performance of birds at 1.5 per cent Fenugreek seed powder supplemented groups has been improved as compared to antibiotics (Zinc Bacitracin and Salinomycin) and control group.

Keywords: Broilers, Serum-Biochemical, Fenugreek, Antibiotics.

INTRODUCTION

The Indian poultry industry is thought to be 5000 years old, with a greater expansion from backyard chicken husbandry to the poultry industry over the last four decades. Poultry farming played an important role in reducing starvation, poverty and unemployment. The poultry business in India is a fast-growing sector that accounts for around 8 per cent of the country's GDP.

India is the world's third-largest producer of eggs and fifth-largest producer of chicken meat. In 2019, the total number of chickens in the country was 851.81 million, up 16.8 per cent from the previous census. In 2019, the total number of backyard poultry in the country is 317.07 million, while the total number of commercial poultry is 534.74 million. A total of 95.2 billion eggs and

5.3 million metric tonnes of poultry manure are generated.

Fenugreek (*Trigonella foenum-græceum* L.) is a well-known medicinal plant that grows naturally and is primarily grown in India, Pakistan and China. Fenugreek seeds contain a wide range of medicinal qualities, including hypoglycemic, anti-diabetic, anti-fertility, anti-cancer, anti-parasitic, anthelmintic, antibacterial, anti-inflammatory, antipyretic and antimicrobial capabilities (Bash *et al.*, 2003). It contains neurin, biotin, and trim ethylamine, which promote appetite by acting on the neurological system. (Al Habori *et al.*, 1998)

Abdul-Rahman (2012) investigated that feeding of Fenugreek seeds at 10 g/kg of diet in broiler breeder chicken significantly (p < 0.05) improved the packed cell volume percentage,

red blood count and Hb concentration and attributed this improvement in erythropoiesis to the enhancement of antioxidant activity in RBCs which decreases the production of free radicals that destroy Hb and cause haemolysis of RBCs where as Abbas (2010) found negative effects on feed intake and no effect on live weight. Apart from a range of beneficial effects including growth promoting, having 24 % CP and 3819 (ME) Kcal/Kg of energy and rich in vitamins and minerals, fenugreek can be regarded as a nutritious feed ingredient as well. El-Mallah *et al.* (2005) reported that 2% fenugreek in diets of turkey chicks significantly increased the digestibility of nitrogen free extract due to saponin present in fenugreek. Meanwhile, Al-Habori *et al.* (1998) found that fenugreek reduced the plasma cholesterol levels of rabbit.

Materials and Methods

Present study was carried out at Poultry Unit, Veterinary Polyclinic and A.I. Centre, Department of Animal Husbandry and Dairy Science, Mahatma Phule Krishi Vidyapeeth, Rahuri, Maharashtra State. Located at 19.392677 N and 74.648827 E at an altitude of 455 meter. Minimum and maximum ambient temperature range from 12^o C to 15^o C in winter and 33^o C to 38^o C in summer with annual rainfall of 455 mm. The experiment was conducted in July - August during the year 2021-22. The experimental chicks were all raised in a deep litter system with rice husk as a litter material in a well-ventilated house under the same management and environmental conditions. For the first three weeks of life, proper brooding of chicks was accomplished by giving adequate heat and light using electric lamps in each group.

The fenugreek seed powder (FSP) was procured from Medicinal and Aromatic Plant Unit, Department of Agricultural Botany, Mahatma Phule Krishi Vidyapeeth, Rahuri as feed supplement and mixed in commercial broiler feed manufactured by Baramati Agro industries as per different treatment levels.

Selection of Experimental Chicks

For the present study 160 chicks of day old age, commercial broiler chicks of Vencob strain were

procured from Vyankateshwara Hatcheries Pvt. Limited, Pune, Maharashtra. On arrival, chicks were weighed and distributed randomly in to 4 groups viz. T₀, T₁, T₂ and T₃ with 40 chicks in each treatment as replicates, on equal weight basis.

Treatment Details

The dietary treatments are as follows,

- T0 Basal Diet (Control)
- T1 Basal Diet + Antibiotic (Zinc Bacitracin 20 mg/kg and Salinomycin 60 mg/kg)
- T2 Basal Diet + Fenugreek seed powder @1.0 % of feed.
- T3 Basal Diet + Fenugreek seed powder @1.5 % of feed.

Proximate Composition of Experimental Broiler Ration

It was observed that experimental broiler rations contained adequate nutrients for growth as per BIS (1992). The proximate composition of experimental Pre-starter, starter ration and finisher ration is given in Table 1. The crude protein and metabolizable energy contain in pre-starter, starter and finisher ration as 23.63, 21.49, 20.24 and 2963.25, 3073.2, 3186.5 kcal/ kg, respectively.

OBSERVATIONS RECORDED

Body Weight

At the end of each week, total numbers of birds were collectively weighed replicate wise and data were recorded. However, in next week same birds were weighed to obtain the body weight gain. Likewise the data were recorded to calculate gain in weight in each replicate.

METHODS OF ANALYSIS

Lipid profile estimation

The lipid profile includes following constituents.

Total serum cholesterol (TC), triglycerides (TG), low density lipoprotein (LDL) and high density lipoprotein (HDL) which were determined in serum using biochemical analysis.

Total blood cholesterol

The blood cholesterol was estimated by the procedure of Godkar (1994). Same is narrated as follows.

Procedure : Test tubes were labelled as blank (B), standard (S) and test (T) for estimating serum total cholesterol. The further procedure was as follows.

Reagent	Serum (T)	Standard (S)	Blank (B)
-	0.01 ml	0.01 ml	-
Cholesterol reagent	1.0 ml	1.0 ml	1.0 ml

One ml cholesterol reagent was taken for each test tube. Then 0.01 ml cholesterol standard was taken in (S) test tube and also 0.01 ml serum was taken in (T) test tube. Then it was mixed well and incubated at 37°C for five minutes. After incubation absorbance of test (T) and standard (S) was measured against blank (B) on calorimeter at 510 nm. Serum total cholesterol was calculated by using following formula.

$$\text{Serum cholesterol (mg / dl)} = \frac{\text{Absorbance of sample (T)}}{\text{Absorbance of standard (S)}} \times 200$$

Where,

200 = Standard Concentration

Serum triglycerides

The serum triglyceride was estimated by the procedure of Godkar (1994) as follows.

Procedure: Test tubes were labelled as blank (B), standard (S) and test (T) for estimating serum total cholesterol. The further procedure was as follows.

Reagent	Serum (T)	Standard (S)	Blank (B)
-	0.01 ml	0.01 ml	-
Triglyceride reagent	1.0 ml	1.0 ml	1.0 ml

One ml triglyceride reagent was taken for each test tube. Then 0.01 ml triglyceride standard was taken in (S) test tube and also 0.01 ml serum was added in (T) test tube. Then it was mixed well and incubated at 37°C for five minutes. After incubation absorbance of test (T) and standard (S) was measured against blank (B) on calorimeter at 510 nm. Serum triglyceride was calculated by using following formula.

$$\text{Serum triglyceride (mg / dl)} = \frac{\text{Absorbance of sample (T)}}{\text{Absorbance of standard (S)}} \times 200$$

Where,

200 = Standard Concentration

Serum HDL cholesterol

The serum HDL cholesterol was estimated by the procedure of Richmond (1973) as follows.

Step 1 : Precipitation of HDL and LDL

The 0.5 ml serums were taken in each test tube and added HDL precipitating reagent was pipette into a clean dry centrifuge tube. It was mixed well and allowed to stand at room temperature for five minutes, then centrifuge at 4000 rpm for 10 minutes to obtain a clear supernatant.

Step 2 : Assay of HDL cholesterol

Test tubes were taken and labelled as blank (B), standard (S) and test (TH). The further procedure was as below.

Reagent	Serum (T)	Standard (S)	Blank (B)
-	0.05 ml	0.05 ml	-
Cholesterol working solution reagent	1.0 ml	1.0 ml	1.0 ml

Take 1.0 ml cholesterol working solution reagent in each test tube. Then add 0.05 ml HDL standard in (S) test tube. Add also 0.05 ml supernatant solution in (TH) test tube. Then it was mixed well and incubated at 37°C for five minutes. After incubation absorbance of test (TH) and standard (S) was measured against blank (B) on a calorimeter at 510 nm. Serum HDL cholesterol was calculated by using following formula.

$$\text{Serum HDL cholesterol (mg / dl)} = \frac{\text{Absorbance of sample (TH)}}{\text{Absorbance of standard (S)}} \times 100$$

Where,

100 = Standard Concentration

LDL cholesterol

The LDL cholesterol was calculated by using Friedwald's formula (Friedwald's *et al.*, 1972) as follows.

LDL cholesterol (mg/dl) = Total cholesterol - (HDL cholesterol)

Statistical Analysis

The data were analyzed using General Linear Model procedure of statistical package for social sciences (SPSS) 20th version and comparison of means tested using Duncan's multiple range test (1955) and significance was considered at ($P < 0.05$).

RESULTS AND DISCUSSION

Present study was conducted to investigate the comparative effects of fenugreek seed powder and antibiotic on broilers performance and Serum-biochemical changes. Table 2 represents the data Serum parameters (%) due to different dietary treatments of fenugreek seed powder and antibiotic supplementation during experimental period. The data also presented graphically in Fig. 1.

The serum parameters data indicated significant difference in the serum traits (%) among treatment groups. The serum-Biochemical constituents like glucose, cholesterol, triglyceride, high density lipoprotein (HDL), low density lipoprotein (LDL) was estimated at the end of experiment and the results are presented in Table. 2. corresponding analysis carried out in CRD and is depicted in figure 1.

The level of glucose at 42nd day in T_0 , T_1 , T_2 and T_3 were 123.33, 137.25, 144.58 and 151.26 mg/dl respectively. The level of glucose in T_0 was significantly lower than T_2 , T_3 and T_1 . Higher glucose level found in T_3 (151.26 mg/dl), T_2 (144.58 mg/dl) and lowest level of glucose was found in T_1 (123.33 mg/dl). Result indicates that the concentration of glucose is increases as the quantity of feed additive (fenugreek seed powder) increases.

The level of cholesterol at 42nd day in T_0 , T_1 , T_2 and T_3 were 134.52, 149.25, 136.96 and 146.48 mg/dl respectively. The level of cholesterol in T_0 , T_1 , T_2 and T_3 was non-significant but different T_0 (134.52 mg/dl) was lower cholesterol level followed by T_2 (136.96 mg/dl), T_3 (146.48 mg/dl) and T_1 (149.25 mg/dl). Which indicates that the concentration of cholesterol is decreases in 1% of inclusion level of fenugreek seed powder. Similar observation were also found by Bhale (2015) who reported that the concentration of cholesterol was decreased as the quantity of feed

additive (germinated fenugreek seed powder) increases. The results of lipid fractions show a decrease in serum cholesterol confirming the hypocholesterolemic properties of fenugreek seeds (Khadr and Abdel Fattah 2007)

The level serum triglyceride at 42nd day in T_0 , T_1 , T_2 and T_3 were 209.66, 192.59, 139.44 and 105.79 mg/dl, respectively. Triglyceride values are significant and the highest value in T_0 (209.66 mg/dl) and lowest value in T_3 (105.79 mg/dl) i. e. control group without supplemented fenugreek seed powder had significantly ($P < 0.01$) higher level of serum triglyceride.

The level of HDL cholesterol at 42nd day in T_0 , T_1 , T_2 and T_3 were 66.35, 87.36, 81.24 and 76.44 mg/dl respectively. HDL highest value in T_1 (87.36 mg/dl) with antibiotics i.e Zinc Bacitracin 20 mg and Salinomycin 60 mg and lowest value in T_0 (66.35 mg/dl) in control group. The values of HDL clearly showed that all supplemented fenugreek seed powder treatment have high values than control.

The level of LDL cholesterol at 42nd day in T_0 , T_1 , T_2 and T_3 were 23, 32.58, 35.25 and 36.96 mg/dl, respectively. The level of LDL cholesterol in T_0 , T_1 , T_2 and T_3 was non-significant. T_3 (36.96 mg/dl) was higher LDL level followed by T_2 (35.25 mg/dl), T_1 (32.58 mg/dl) and lowest level of LDL was found in T_0 (23.00 mg/dl).

The level of LDL / HDL ratio at 42nd day in T_0 , T_1 , T_2 and T_3 were 0.34, 0.37, 0.43 and 0.48, respectively. LDL / HDL ratio highest value in T_3 (0.48) in 1.5 per cent level of fenugreek seed powder and lowest value in T_0 (0.34) in control group. Which is proves the positive effect of fenugreek seed powder in serum lipid profile.

Duru *et al.* (2013) also reported the same finding and stated that if the level of Fenugreek seed increases above 20 g/kg broiler diet causes decreases triglyceride and cholesterol level but increases the LDL, cholesterol and glucose.

Al-Habori (1998) found that fenugreek and its extract reduced the levels of cholesterol, triglycerides, and Low density lipoprotein (LDL-cholesterol) with no effect on high density lipoprotein (HDL-cholesterol). This selective reduction in LDL-cholesterol results in the improvement of the ratio of HDL- cholesterol to LDL-cholesterol. Whereas Safaei *et al.* (2013)

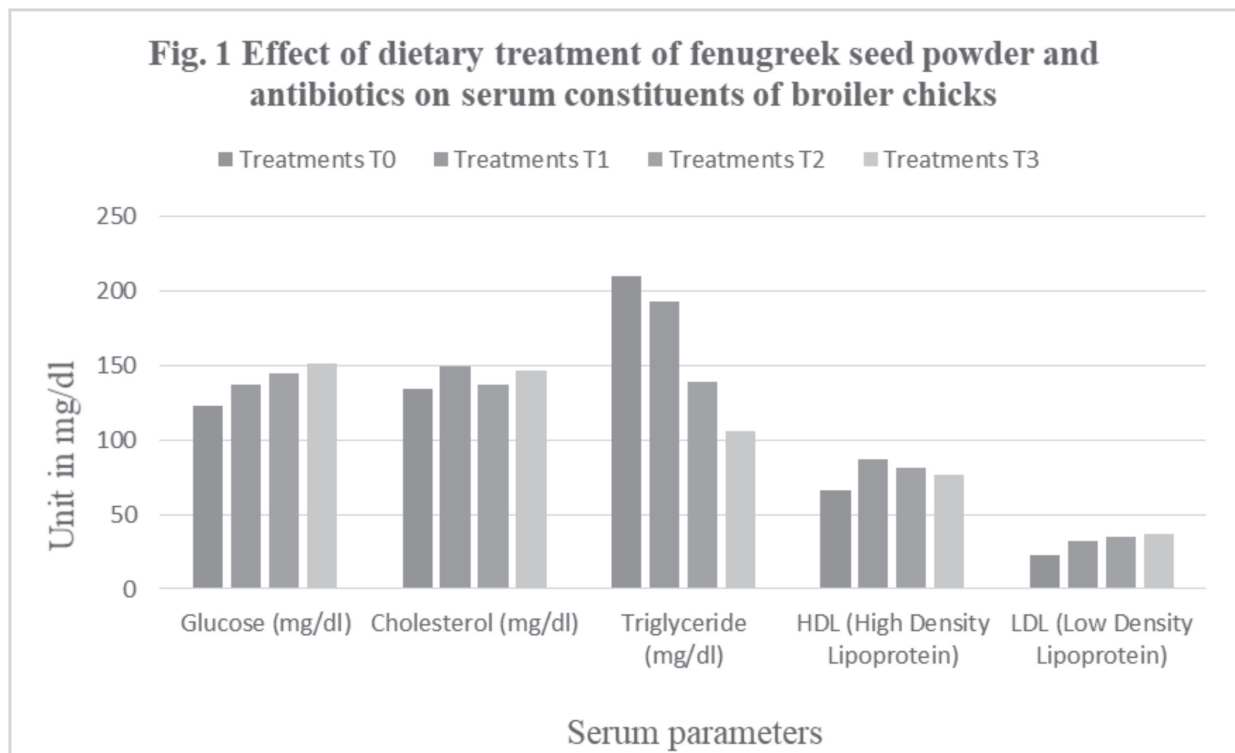
reported that effects of fenugreek extract in drinking water on some blood parameter like triglyceride, cholesterol, and glucose levels were significantly different among the treatments when compared to the control group.

Table 1: Proximate chemical composition of experimental broiler ration (% DM basis)

Sr. No.	Nutrients	Per cent in ration		
		Pre-starter	Starter	Finisher
1	Crude Protein	23.63	21.49	20.24
2	Crude Fibre	3.49	3.44	3.78
3	Ether Extract	4.38	5.39	5.49
4	Total Ash	6.59	6.41	5.89
5	Acid Insoluble Ash	1.29	1.59	1.62
6	Nitrogen Free Extract	60.44	63.37	64.45
7	Metabolizable Energy (Kcal/Kg)	2963.25	3073.2	3186.5
8	E/P Ratio	126.26:1	142.34:1	153.54:1

Table 2: Effect of different levels of dietary treatments on serum constituents of broiler chicks

Parameters	Treatments				SE	CD
	T ₀	T ₁	T ₂	T ₃		
Glucose (mg/dl)	123.33	137.25	144.58	151.26	3.42	10.89
Cholesterol (mg/dl)	134.52	149.25	136.96	146.48	7.68	NS
Triglyceride (mg/dl)	209.66	192.59	139.44	105.79	32.76	101.69
HDL (High Density Lipoprotein)	66.35	87.36	81.24	76.44	4.83	13.88
LDL (Low Density Lipoprotein)	23	32.58	35.25	36.96	2.78	NS
LDL/HDL Ratio	0.34	0.37	0.43	0.48	0.57	NS



CONCLUSION

According to the trial carried out earlier, it is valuable to use fenugreek as a growth promoter or feed supplement and alternative to antibiotics in commercial broiler production. The conclusions can be drawn from this project that the addition of fenugreek seed powder in broiler ration gradually decreased serum cholesterol,

triglyceride and increased the glucose level and improve the LDL/HDL ratio whereas antibiotic inclusion in feed resulted in increase in cholesterol, triglyceride and HDL.

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