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Effect of betaine hydrochloride supplemented diets on growth and nutrient metabolizability of broilers

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Abstract
The present study was planned to assess the effect of different doses of betaine hydrochloride on the growth parameters and nutrient metabolizability of broilers. A total of 200 birds were allocated into five different treatment groups each having four replicates of ten birds in each. T_1 was taken as control while T_2, T_3, T_4, and T_5 were supplemented with 500, 1000, 1500 and 2000 mg/kg betaine hydrochloride in basal feed. A significant increase in body weight gain was observed during the 42 days trial in all betaine supplemented groups in comparison to control. The maximum body weight gain was found in group T_5 which received 2000 mg/kg betaine and differed significantly ($p<0.05$) from all other groups. The feed conversion efficiency of birds was most efficient in T_5 group among all treatment groups. The dry matter and nitrogen metabolizability was highest in group T_5 and differed significantly ($p<0.05$) from all other treatment groups. Similarly the gross energy metabolizability also significantly increased in all betaine supplemeted groups in comparison to control and was found to be highest in T_5 group. It was evident from this study that betaine supplementation @ 2000 mg/kg had the most profound effects on body growth and nutrient absorption rate of broilers.

Keywords: Betaine, feed conversion, metabolizability, weight gain

Introduction
Poultry meat is the fastest-growing component of global meat demand, and India, the world's second-largest developing country, is experiencing rapid growth in its poultry sector. The majority of poultry production in India comes from commercial sector which uses variety of feed supplements for efficient and economical production of poultry. One such feed supplement is betaine which is a trimethyl derivative of the amino acid glycine found naturally in some plants (e.g. beetroot) and animal tissues. Betaine has a number of different functions both at the gastrointestinal and metabolic level (Eklund *et al*., 2005) [7]. Betaine donates its labile methyl group which can be used in transmethylation reactions for synthesis of substances like carnitine and creatine. Therefore, dietary supplementation of betaine may reduce the requirement for other methyl group donors like methionine and choline. As a "methyl group donor", betaine could improve weight gain and feed efficiency (Hassan *et al*., 2005) [11]. It conserves the energy required for the Na+/K+ pump at high temperatures and this spared energy is used to fuel the growth of broilers (Ramus J., 2001) [15]. Betaine supplementation in feed or water may decrease the dehydration by facilitating water retention in the body and also it may contribute to maintain both the bird's energy balance and feed intake (Eklund *et al*., 2005) [7]. The osmoprotective properties of betaine may result in an increased proliferation of the intestinal structure which, in turn may have a positive impact on animal health and nutrient digestibility (Afrin *et al*., 2018) [1]. Keeping these points in mind this experiment was performed to analysis the effect of betaine supplementation on growth and nutrient metabolizability of broilers.

Materials and Methods
Ethical approval
The animal experiment was conducted in accordance with guidelines approved by the Institutional Animal Ethics Committee, 12/CPCSEA Dated 12.04.2022 in the Department of Animal Nutrition, Lala Lajpat Rai University of Veterinary & Animal Sciences, Hisar.

Experimental design
A study of 6 weeks duration was conducted on two hundred commercial broiler chicks randomly distributed into five treatment groups with four replicates of ten birds each.

The control group (T_1) was offered only basal diet to fulfill the metabolizable energy (ME) and crude protein requirements of broilers and formulated as per BIS (2007) [5] while groups T_2, T_3, T_4 and T_5 were supplemented with 500, 1000, 1500 and 2000 mg/kg betaine hydrochloride along with basal diet. The ingredients and chemical composition of the basal diet as analyzed as per AOAC (2013) [4] is presented in Table 1.

The birds were housed in deep litter system from day-old to 42 days of age following standard management practices. The birds were fed pre starter, starter and finisher diets for 1 to 7, 8 to 21 and 22 to 42 days of age, respectively. Feed and water were provided *ad-lib*. All the birds were vaccinated against Ranikhet disease on 4th day and IBD on 13th day of age. Feed intakes and body weight gain were recorded fortnightly. Feed conversion ratio (FCR) for each replicate was calculated as follows:

FCR = Total feed consumed (g)/Total body weight gain (g)

To study the balance of nitrogen and energy, a metabolism trial was conducted during 6th week of growth period. One bird was randomly selected from each replicate and transferred to metabolic cages. Thus, the feed residue and excreta voided were weighed and properly recorded for final calculations of the total daily feed consumption and excreta voided. Gross energy of oven dried feed was determined by standard procedure using Bomb Calorimeter.

Nitrogen retetion calculated as: (Nitrogen intake-Nitrogen excreted)/Nitrogen intake × 100

Similarly, dry matter metabolizability and gross energy metabolizability were also calculated. The data were statistically analyzed (Snedecor and Cochran, 1994) [18] and the means of different experimental groups were tested for statistical significance.

Table 1: Ingredients and Chemical Composition (% DM Basis) of Experimental Diets in Different Growth Phases of Broiler Chicks

Ingredients (Kg/100 kg)	Pre-starter	Starter	Finisher
Maize	50	52.64	58.2
Soybean Meal	25	22.36	16.8
Fish Meal	6	6	6
Groundnut cake	10	10	10
Vegetable Oil	4.67	5.48	5.9
Mineral Mixture	2	2	2
Common salt	0.5	0.5	0.5
Chemical Composition (% DMB)			
Crude Protein	22.79	21.81	20.10
Ether Extract	7.78	8.38	8.61
Crude Fibre	4.89	4.45	4.15
Ash	9.83	9.51	8.92
Nitrogen Free Extract	43.09	43.51	45.74
Metabolizable Energy (Kcal/Kg)	3050.67	3194.64	3390.56

Results and Discussion

Growth performance

The mean body weight gain during first fortnight (0-14d) increased significantly ($p<0.05$) among all betaine supplemented groups in comparison to control. Treatment group T_5 showed the maximum weight gain however there was no significant ($p>0.05$) difference between groups T_3, T_4 and T_5. During the second fortnight period (15-28d) the body weight gain of T_3, T_4 and T_5 increased significantly ($p<0.05$) from T_2 and T_1 (control) and was highest in T_5 group (769.60 g). Again in the third recording (29-42d) the T_5 group showed the maximum (1189.47 g) mean values of body weight gain in comparison to control. During the overall period (0-42d) betaine supplementation resulted in significant ($p<0.05$) body weight gain of all betaine supplemented groups in comparison to control and ranged from 2037.47 g (T_1) to 2257.49 g (T_5) as can be seen in data presented in Table 2. Betaine supplementation @ 2000 mg/kg proved to be most effective in gaining weight among all dietary treatments. Our results are in agreement with the findings of other researchers (Chand *et al*., 2017; Amer *et al*., 2018; Liu *et al*., 2019 and Akther & Avi., 2023) [6, 3, 12, 2] that betaine @ 2000 mg/kg basal diet proved to be most effective for broilers in gaining body weight in comparison to control and treatment groups containing lower doses of betaine. The increase in body weight gain resulted due to potential effect of betaine to improve the digestibility of specific nutrients (Eklund *et al*., 2006) [8] and higher energy-protein metabolism due to methyl group donor function (Eklund *et al*., 2005) [7]. Higher body weight gain has also been reported elsewhere by supplementing betaine at a level of 0.65% in diet (Garcia *et al*., 1999; Eklund *et al*., 2005; El-Husseiny *et al*., 2007; Nofal *et al*., 2015) [10, 7, 9, 13] thus supporting our research findings.

Table 2: Body weight gain (g) in broiler chicks during different growth periods under different dietary treatments

Treatments	Mean Body Weight Gain(g)			
	0 to 14d	15 to 28d	29 to 42d	0 to 42d
T_1	251.54^{a}±6.5	731.75^{a}±13.7	1051.45^{a}±12.04	2034.74^{a}±11.42
T_2	286.40^{b}±1.25	739.70^{a}±18.93	1091.24^{ab}±15.12	2117.36^{b}±17.35
T_3	296.20^{c}±1.1	748.22^{b}±9.23	1140.34^{b}±18.21	2184.81^{bc}±24.68
T_4	298.40^{c}±3.35	769.60^{bc}±14.41	1189.47^{bc}±21.34	2257.49^{c}±27.31
T_5	298.93^{c}±3.59	790.37^{c}±16.94	1244.37^{c}±13.27	2333.67^{d}±19.37

Means bearing different superscripts in a column differ significantly ($p<0.0$)

Data pertaining to feed intake given in Table 3 shows that broilers consumed more feed in betaine suppmented groups in comparison to control (T_1). During the first two weeks the feed intake was highest (447.90 g) in group T5 followed by T_4, T_3, T_2 and T_1. The same trend was seen in next two fortnightly observations upto 42 days trial completion. The overall (0-42d) feed intake was significantly ($p<0.05$) higher in T_5 group (3684.43 g) in comparison to rest of the treatment groups. Similar results were reported in other studies where supplementation of 0.2% betaine resulted higher feed intake in supplemented group compared with control (Rafeeq *et al*., 2011; Nofal *et al*., 2015; Chand *et al*.,2017[6]; Amer *et al*.,2018[3] and Liu *et al*., 2019) [14, 13, 6, 3, 12].

Table 3: Feed intake (g) during different growth periods in broiler chicks under different dietary treatments

Treatments	Feed Intake (g)			
	0 to 14d	15 to 28d	29 to 42d	0 to 42d
T_1	$425.35^{a}\pm6.62$	$1222.02^{a}\pm8.53$	$1843.01^{a}\pm18.65$	$3490.38^{a}\pm16.37$
T_2	$434.12^{b}\pm5.84$	$1265.77^{b}\pm5.74$	$1840.05^{a}\pm24.65$	$3539.94^{b}\pm28.45$
T_3	$433.31^{b}\pm6.14$	$1262.77^{b}\pm9.45$	$1855.01^{ab}\pm25.52$	$3551.09^{b}\pm34.24$
T_4	$436.24^{b}\pm5.34$	$1278.31^{c}\pm6.34$	$1886.27^{b}\pm23.87$	$3600.82^{c}\pm18.41$
T_5	$447.90^{c}\pm4.24$	$1281.86^{c}\pm6.31$	$1954.67^{c}\pm22.34$	$3684.43^{d}\pm24.08$

Means bearing different superscripts in a column differ significantly ($p<0.05$)

Data presented in Table 4 shows that the feed conversion ratio during 0-14 days of trial dropped significantly ($p<0.05$) in betaine supplemented groups in comparison to control (T_1). The FCR values of T_3, T_4 and T_5 had no significant differences while differed significantly from T_2 and control (T_1). In the second fortnight (15-28d) the FCR values of T_5 were lowest and differed significantly ($p<0.05$) from other groups. During day 29 to 42 the FCR of betaine supplemented groups further decreased and differed significantly from control (T_1). However, groups T_4 and T_5 having lowest FCR values differed non significantly ($p>0.05$) with each other. In the overall growth period (0-42d) the FCR ranged from 1.71 (T_1) to 1.57 (T_5) and was most efficient in treatment group T_5 supplemented with 2000 mg/kg betaine (1.57) while differing non significantly with group T_4 (1.59). These results are in line with the findings of other researchers (Chand *et al*., 2017; Sun *et al*., 2019 and Liu *et al*., 2019) [6, 12] that betaine supplementation by 2000 mg/kg diet had significant reduction in FCR of broilers. Similarly, El-Husseiny *et al*. (2007) [9] revealed that addition of betaine at the levels of 0.75 g/kg in diet significantly improved FCR as compared to the control group. However, in contrast to present study, very little or no effect of betaine on FCR was detected in broiler birds in other studies (Rostagno and Pack, 1996; Zulkifli *et al*., 2004 and Amer *et al*., 2018) [16, 20, 3].

Table 4: Feed conversion ratio during different growth periods in broiler chicks under different dietary treatments

Treatments	Feed conversion ratio			
	0 to 14d	15 to 28d	29 to 42d	0 to 42d
T_1	$1.69^{c}\pm0.02$	$1.66^{b}\pm0.02$	$1.75^{d}\pm0.03$	$1.71^{c}\pm0.02$
T_2	$1.51^{b}\pm0.02$	$1.71^{c}\pm0.02$	$1.68^{c}\pm0.04$	$1.67^{bc}\pm0.03$
T_3	$1.46^{a}\pm0.01$	$1.68^{b}\pm0.03$	$1.62^{b}\pm0.05$	$1.63^{b}\pm0.03$
T_4	$1.46^{a}\pm0.02$	$1.66^{b}\pm0.01$	$1.58^{a}\pm0.03$	$1.59^{a}\pm0.01$
T_5	$1.49^{a}\pm0.01$	$1.62^{a}\pm0.03$	$1.57^{a}\pm0.01$	$1.57^{a}\pm0.01$

Means bearing different superscripts in a column differ significantly ($p<0.05$)

Nutrient Metabolizability

The percent mean values (Table 5) of dry matter metabolizability which ranged from 70.53% (T_1) to 73.84% (T_5) increased gradually as the level of betaine was increased among treatment groups and was found maximum in T_5 group which differed significantly ($p<0.05$) from rest of the betaine supplemented groups and control(T_1). Similarly, the mean values of nitrogen and gross energy metabolizability percentage was found to be significantly ($p<0.05$) improved in T_5 group supplemented with 2000 mg/kg betaine in comparison to control (T_1) and other groups. Findings of Liu *et al*., 2019 [12] are in support of our findings who stated that nitrogen retention by broilers in heat stressed condition linearly improved ($p<0.05$) by graded dietary betaine levels (500, 1000, 2000 mg/kg). The improvements in metabolizability of nutrients may be due to because betaine improves the structural and functional characteristics of intestinal epithelia which results in better absorption of the nutrients (Sakomura *et al*., 2013) [17].

Table 5: Mean values of nutrients metabolizability in broiler chicks under different dietary treatments

Treatment	DM Metabolizability (%)	Nitrogen Metabolizability (%)	GE Metabolizability (%)
T_1	$70.53^{a}\pm0.11$	$71.00^{a}\pm0.29$	$69.65^{a}\pm0.11$
T_2	$71.78^{b}\pm0.12$	$72.48^{b}\pm0.25$	$70.88^{b}\pm0.15$
T_3	$72.95^{c}\pm0.16$	$72.81^{b}\pm0.24$	$71.15^{b}\pm0.22$
T_4	$73.03^{c}\pm0.10$	$72.84^{b}\pm0.14$	$71.35^{bc}\pm0.15$
T_5	$73.84^{d}\pm0.17$	$73.65^{c}\pm0.24$	$71.93^{c}\pm0.23$

Means bearing different superscripts in a column differ significantly ($p<0.05$)

Conclusion

In this study betaine improved the overall growth performance of broiler birds. The highest improvement in body weight gain, feed intake, FCR and nutrient metabolizability were observed in 2000 mg/kg betaine supplemented group in comparison to control and other groups. Thus, this study suggests that betaine at an inclusion level of 2000 mg/kg feed can be recommended as a potential feed supplement in broilers diet for better growth results.

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