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Replacement of Synthetic Choline Chloride by Herbal Choline in Diets on Liver Function Enzymes, Carcass Traits and Economics of Broilers

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ABSTRACT

The present study was conducted to evaluate the replacement of synthetic choline chloride by herbal choline in diets on liver function enzymes, carcass traits and economics of broiler production. Three hundred straight-run Cobb-400 day-old broiler chicks were randomly distributed into five treatment groups having three replicates having 20 chicks in each and reared up to 6 weeks. The treatment group T1 (control) offered basal diet (BIS, 2007), T2 offered basal diet with choline chloride-60% @ 1 kg/ton of feed, groups T3, T4 and T5 offered basal diets with herbal choline @ 0.25, 0.35 and 0.50 kg/ton of feed, respectively. The SGPT, SGOT and ALP were significantly ($P<0.01$) reduced in treatment group T5 as compared to control group. There was non-significant difference for blood loss, feather loss, edible carcass yield and abdominal fat percent in all treatment groups. The net profit expressed in terms of rupees per kg live weight in treatment groups was increased by 18.35% (T2), 14.37% (T3), 21.05% (T4), and 38.55% (T5) over the control group (T1). It may be concluded that the herbal choline supplemented at dose levels of 0.35 kg/ton and 0.5 kg/ton of feed was found to be useful to improve the overall performance due to better growth resulting into increase in profit margin of broiler production replacing synthetic choline chloride (1kg/ton) in broiler diets. Moreover, the supplementation of herbal choline at 0.5 kg/ton of feed was more beneficial in terms of improved liver protection, carcass traits and economics of broiler production.

Keywords: Broilers, Liver enzymes, Carcass, Economics, Herbal choline

The use of high energy diets aimed at shortening the rearing period may increase metabolic disorders such as fatty liver syndrome (FLS) in broiler chickens and condition that generally affects fast growing broilers and caged layers with an inadequate chance to move and exercise freely (Jiang *et al.,* 2013). Increased abdominal fat pad (Corduk *et al.,* 2007), incidence of leg problems (Van Emous *et al.,* 2015) and hypertension (Gopi *et al.,* 2014) are some other detrimental responses associated with high energy diets. Choline is a beta-hydroxy ethyl trimethyl ammonium hydroxide. It is essential for the formation of acetylcholine, a substance that makes possible the transmission of nerve impulses. It prevents abnormal accumulation of fat (fatty livers) by promoting its transport as lecithin or by increasing the utilization of fatty acids in the liver itself (Xu *et al.,* 2010). The supplementation of choline chloride diet improved weight gain and feed conversion efficiency in quail (Alagawany *et al.,* 2015) and the symptoms of choline deficiency include reduced growth, fatty infiltration of liver and perosis in chicks (McDonald, 2011). Therefore, choline is referred as a "lipotropic" factor due to its function of acting on fat metabolism by hastening removal or decreasing deposition of fat in liver.

Choline is a rediscovered critical amino acid for poultry and usually added to poultry diets in the form of synthetic choline chloride. However, synthetic choline chloride has several drawbacks. It is highly hygroscopic and the acceleration of oxidative loss of vitamins in the diet, and the formation of tri-methylamine in the gastrointestinal tract of the birds (Zeisel *et al.,* 1989). Synthetic choline chloride is very less absorbed from intestine. Choline is

also present in plants in the phosphatidylcholine form, free choline and sphingomyelin. Currently, there are natural products, produced from selected plants, with high content of choline in esterified form and with high bioavailability, which may be an important alternative to synthetic choline chloride. Many researchers have shown that these products can replace choline chloride in diets for poultry (Muthukumarasamy *et al*., 2004; Gangane *et al.,* 2010). Sharma and Ranjan (2015) also reported that the supplementations of herbal choline can replace synthetic choline and biotin as evident by the comparable bird's growth, viability and hypocholesterolemeic effect. The present study was undertaken to evaluate the effect of replacement of synthetic choline chloride by herbal choline in diets on liver function enzymes, carcass traits and economics of broiler production.

MATERIALS AND METHODS

Ethical approval

The biological experiment was carried out as per the Institute Animal Ethics Committee's (IAEC) approved.

Experimental design and management

Three hundred straight run 'Cobb-400' day-old broiler chicks were randomly distributed into five treatment groups having three replicates of 20 chicks in each and reared up to 6 weeks of age. The treatment group T1 (control) offered basal diet (BIS, 2007) without synthetic choline chloride-60% (SC) or herbal choline (HC). The experimental design and the details of the dietary treatments are presented in Table 1.

The standard and uniform managemental practices were followed for all treatment groups throughout the experimental period. The birds were offered *ad-lib* fresh and clean drinking water throughout the experiment. The immunization against Ranikhet Disease (Lasota strain) and Infectious Bursal Disease (IBD standard strain) vaccination was carried out on 7th and 14th day, respectively, followed by booster doses on 18th day and 24th day through drinking water.

Procurement of ingredients and feed formulation

The good quality feed ingredients were procured from local market for preparation of experimental diets. The herbal choline was procured from *M/s. Vamso Biotec Pvt. Ltd., Gurgaon, Haryana, INDIA*. The rations were formulated as per BIS, 2007 for pre-starter, starter and finisher phases (Table 2). All the diets were isocaloric and iso-nitrogenous. The *ad-lib* feeding was provided during the experimental period.

Liver function enzyme tests

Two blood samples from each replicate i.e. 6 birds from each treatment group were collected on 21st and 42nd day of experiment by random selection and separated serum was subjected to blood biochemical test, alanine aminotransferase (ALT/SGPT), aspartate aminotransferase (AST/SGOT), alkaline phosphatase (ALP) by using standard commercial kits (*Cogent, SPAN diagnostic Ltd, INDIA*) as per (Kumar and Krupakaran, 2014).

Carcass traits

Two birds from each replicate of six birds from each treatment were selected at the end of 6th week of age. The birds were fasted for a period of twelve hours prior to slaughter. These birds were slaughtered using the standard procedure. Carcass traits such as blood loos, feather loss, edible carcass yield and abdominal fat pad weight were measured. The percent blood loss and feather loss relative to live body weight were calculated by using following equations.

Blood loss (%) =

$$\frac{\text{Live body weight} - \text{Body weight after bleeding}}{\text{Live body weight}} \times 100$$

Feather loss (%) =

$$\frac{\text{Body weight after bleeding} - \text{Body weight after de-feathering}}{\text{Live body weight}} \times 100$$

Economic of broiler production

During this study, attempts were made to calculate the economics of broiler production under different treatment groups. The economics of broiler production was worked out by considering the prevailing prices of different inputs and sale price of broilers in local market. The cost of day

Table 1: Details of the dietary treatments

Treatment groups	Treatment details	No. of replicates / treatment	No. of birds / replicate	No. of birds/ treatment
T1	Control - (basal diet without herbal or synthetic choline-60%)	3	20	60
T2	Basal diet + Synthetic choline chloride-60% @ 1 kg/ton of feed	3	20	60
T3	Basal diet + herbal choline @ 0.250 kg/ton of feed	3	20	60
T4	Basal diet + herbal choline @ 0.350 kg/ton of feed	3	20	60
T5	Basal diet + herbal choline @ 0.500 kg/ton of feed	3	20	60

old chick, feed, medication, vaccination, litter and other overheads were considered while calculating the cost of production. All other cost components of production i.e. cost of chick, medicines, vaccine and other overhead were taken as constant for all the treatment groups. Gross profit per bird was calculated by subtracting the cost of production per bird from the price fetched per bird after selling it in the local market on live weight basis.

Table 2: Ingredient (%) and nutrient composition of basal diet for different growth phases

Ingredient (%)	Pre-starter	Starter	Finisher
Maize	52.18	53.44	57.93
Soybean meal	40.70	38.20	32.90
Vegetable oil	3.10	4.40	5.25
Dicalcium phosphate (DCP)	1.80	1.80	1.80
Limestone powder (LSP)	1.20	1.20	1.20
Salt	0.27	0.25	0.25
Trace mineral mixture*	0.15	0.15	0.15
Vitamin premix**	0.05	0.05	0.05
DL-Methionine	0.20	0.18	0.15
L-Lysine	0.05	0.03	0.02
Toxin binder (UTPP)	0.10	0.10	0.10
Coccidiostat	0.05	0.05	0.05
Sodium bicarbonate	0.15	0.15	0.15
TOTAL	**100.00**	**100.00**	**100.00**
Nutrient composition (% DM)			
Metabolizable energy (kcal/kg)	3003.83	3103.04	3202.76
Crude protein (%)	23.01	22.000	20.02
Ether extract (%)	5.48	6.80	7.78
Crude fiber (%)	4.21	4.09	3.88
Calcium (%)	1.01	1.00	0.98
Total phosphorus (%)	0.70	0.69	0.67
Available phosphorus (%)	0.42	0.42	0.42
Total lysine (%)	1.31	1.22	1.08
Total methionine (%)	0.55	0.51	0.46

The treatment group T2 incorporated synthetic choline chloride 60% at 1 kg/ton of feed and treatment groups incorporated herbal choline at 0.250 kg/ton (T3), 0.350 kg/ton (T4) and 0.500 kg/ton (T5) of feed in the basal diet.

*Trace Mineral Mixture: - Each kg contains: Copper-15g, Iodine-2g, Iron-90g, Manganese-100g, Selenium-0.3g and Zinc-80g; **Vitamin Premix: -Each 500g contains: Vit. A-12.50MIU, Vit. D3-2.50 MIU, Vit. E-12g, Vit. K-1.50g, Thiamine (B1)-1.50g, Riboflavin (B2)-5g, Pyridoxine (B6)-2g, Cyanocobalamin (B12)-0.015g, Niacin-15g, Cal D Pantothenate-10g and Folic acid-0.50g.

Statistical Analysis

Data generated from different treatment groups was analyzed for statistical significance using completely randomized design (Snedecor and Cochran, 1994) and the treatment means were compared by critical differences.

RESULTS AND DISCUSSION

Liver function enzymes

The serum biochemical parameters of liver marker enzymes SGPT, SGOT and ALP on 21st and 42nd day of age in different treatment groups have been presented in the Table 3.

On 21st day, the mean values of liver marker enzyme SGPT, SGOT and ALP were significantly ($P<0.01$) decreased in groups T2, T4 and T5 in comparison with groups T1 and T3. The results suggested that the birds fed diet with supplementation of herbal choline (@ 0.350 or 0.500 kg/ ton of feed) or synthetic choline chloride (@1 kg/ton of feed) in broilers showed decreased liver enzymes indicating hepatoprotective property of the choline. Thus, the use of herbal choline and synthetic choline protected the liver function as evident by normalization of SGPT, SGOT and ALP enzyme levels in broilers. The statistical analysis of data revealed that the SGPT, SGOT and ALP levels in treatment group T5 supplemented with herbal

Table 3: Liver function enzymes on 21st and 42nd days in broilers fed with different levels of herbal choline

Treatment Groups	ALT/SGPT (IU/L)		AST/SGOT (IU/L)		ALP (IU/L)	
	21 day	42 day	21 day	42 day	21 day	42 day
T1	$31.51^{a} \pm 2.11$	$38.12^{a} \pm 2.41$	$279.50^{a} \pm 11.35$	$318.58^{a} \pm 20.52$	$52.83^{a} \pm 4.24$	$58.36^{ab} \pm 2.76$
T2	$23.30^{b} \pm 1.07$	$30.13^{ab} \pm 3.47$	$224.83^{b} \pm 9.39$	$265.95^{a} \pm 31.46$	$36.50^{bc} \pm 2.36$	$51.85^{b} \pm 3.24$
T3	$29.54^{a} \pm 1.50$	$39.61^{a} \pm 6.31$	$272.50^{a} \pm 7.66$	$276.66^{a} \pm 34.70$	$39.83^{bc} \pm 1.64$	$60.70^{a} \pm 2.06$
T4	$19.37^{b} \pm 1.48$	$21.56^{bc} \pm 1.56$	$230.33^{b} \pm 12.00$	$149.37^{b} \pm 17.29$	$43.50^{b} \pm 3.10$	$53.98^{ab} \pm 1.90$
T5	$13.59^{c} \pm 1.40$	$19.45^{c} \pm 1.64$	$196.33^{c} \pm 6.93$	$112.92^{b} \pm 9.04$	$34.83^{c} \pm 2.06$	$33.34^{c} \pm 2.97$
CD	6.101**	13.963**	38.127**	96.474**	11.155**	10.402**
CV%	16.163	29.147	9.844	26.683	16.705	12.518

Means bearing different superscripts within a column differ significantly. **P<0.01, CD-Critical difference, CV-Coefficient of variance. ALT/SGPT-Alanine aminotransferase, AST/SGOT- Aspartate aminotransferase, ALP-Alkaline phosphatise.

Table 4: Blood loss, feather loss, edible carcass yield percentage and abdominal fat percentage of broilers fed different levels of herbal choline

Treatment groups	Blood loss (%)	Feather loss (%)	Edible carcass yield (%)	Abdominal fat (%)
T1	3.37 ± 0.25	4.65 ± 0.15	76.95 ± 0.31	1.60 ± 0.16
T2	3.40 ± 0.50	4.80 ± 0.41	76.53 ± 0.89	1.46 ± 0.21
T3	3.47 ± 0.31	4.82 ± 0.21	75.63 ± 0.97	1.53 ± 0.10
T4	3.31 ± 0.15	4.94 ± 0.24	76.01 ± 0.55	1.49 ± 0.15
T5	3.26 ± 0.11	4.53 ± 0.30	77.02 ± 0.46	1.48 ± 0.08
CD	NS	NS	NS	NS
CV %	21.516	14.188	2.200	23.532

NS- Non-significant, CD-Critical difference, CV-Coefficient of variance.

choline @ 0.500 kg/ton of feed showed significant (P<0.01) decrease as compared to groups T1 and T2 at 42nd day. The supplementation of herbal choline @ 0.500 kg/ton of feed in broilers better protected the liver function as evidenced by lowered SGPT, SGOT and ALP enzyme levels as compared to other groups. These findings are in accordance with Jadhav *et al.* (2008) who reported that there was numerical decrease in SGOT and SGPT in synthetic choline and herbal choline supplemented groups when compared with control. Rath *et al.* (2017) observed that the serum enzymes SGPT (ALT) and SGOT (AST) were significantly lower in the choline treated groups. The results obtained also corroborates with those of Das *et al.* (2011) who reported significantly lower serum SGPT and SGOT levels in the broiler birds fed diet containing synthetic choline chloride.

Carcass traits

The carcass traits such as percent blood loss, feather loss, edible carcass yield and abdominal fat were non-significant in all treatment groups (Table 4). The treatment group T5 showed numerically lower percent blood loss and feather loss as compared to all treatment groups but the difference was non-significant. The edible carcass yield was numerically higher in treatment group T5 supplemented with 0.500 kg/ton of herbal choline in feed but statistical difference was non-significant. From these findings, the supplementation of herbal choline at 0.500 kg/ton of herbal choline in feed improved carcass traits in broilers for economical purpose. These results were in accordance with Khosravinia *et al.* (2015) who also reported that the birds fed on diets with and without lipotropic agents did not influence their carcass yield.

Similarly, Devegowda *et al.* (2011) also reported that broilers supplemented with vegetal source of choline in the feed showed a reduction of fat in the abdomen and liver. Rodelas *et al.* (2011) showed that, except for the overall feed consumption, feed efficiency and dressing percentage, the other performance parameters did not differ significantly among dietary treatment supplemented with choline chloride or biocholine with or without combination with herbal vitamin E and C.

Economics of broiler production

The economics of broiler production of the trial was worked out by considering the price of inputs prevalent at the time of experiment in the market. The cost of synthetic choline chloride @ ₹ 100 per kg and cost of herbal choline @ ₹ 80 per kg were considered for calculating the cost of production for preparation of broiler feeds. The cost of production (₹ /kg) live weight for the treatments groups were ₹ 65.97 (T1), 64.68 (T2), 64.96 (T3), 64.49 (T4) and 63.26 (T5). The net profit rupees per kg live weight was 7.03 (T1), 8.32 (T2), 8.04 (T3), 8.51 (T4) and 9.74 (T5) in treatment groups (Table 5). The net profit expressed in rupees per kg live weight in treatment groups was increased by 18.35% (T2), 14.37% (T3), 21.05% (T4) and 38.55% (T5) over the control group (T1) received diet without synthetic choline chloride/herbal choline. However, the herbal choline supplementation has resulted to increase the net profit per kg live weight in treatment groups by 2.28% (T4) and 17.07% (T4) over the treatment group T2 received diet with synthetic choline chloride at the rate 1kg per ton of feed.

From these results it is revealed that, the treatment group T5 fed diet containing herbal choline @ 0.50 kg/ ton of feed was economically more beneficial among all the treatment groups. The findings are in accordance with Reddy and Bayram (2011) who reported that the supplemental cost of

Table 5: Economics of broiler production fed different levels of herbal choline

Sl. No.	Parameters	Treatment groups				
		T1	T2	T3	T4	T5
1	Chick cost (₹/chick)	38	38	38	38	38
2	Feed intake (g/bird)					
	1. Pre-starter	300.00	300.00	300.00	300.00	300.00
	2. Starter	700.00	700.00	700.00	700.00	700.00
	3. Finisher	3816.72	3749.58	3775.63	3800.24	3845.52
3	Feed price per kg (₹/kg)					
	1. Pre-starter	28.39	28.48	28.41	28.42	28.43
	2. Starter	28.62	28.71	28.64	28.65	28.66
	3. Finisher	28.08	28.17	28.10	28.11	28.12
4	Feed cost per bird (₹/bird)					
	1. Pre-starter	8.52	8.54	8.52	8.53	8.53
	2. Starter	20.03	20.10	20.05	20.05	20.06
	3. Finisher	107.16	105.62	106.09	106.81	108.13
5	Total feeding cost (₹/bird)	135.72	134.26	134.66	135.39	136.72
6	Miscellaneous cost (₹/bird)	10	10	10	10	10
7	Cost of production (₹/bird)	183.72	182.26	182.66	183.39	184.72
8	Cost of production (₹/kg) live weight	65.97	64.68	64.96	64.49	63.26
9	Body weight at the end of 6^{th} week (g/bird)	2785.02	2817.65	2812.02	2843.49	2919.96
10	Cumulative feed conversion ratio at 6^{th} week	1.76	1.71	1.73	1.72	1.69
11	Return on sale @ ₹ 73 per kg body weight	203.31	205.69	205.28	207.57	213.16
12	Net profit (₹/bird)	19.59	23.43	22.62	24.19	28.44
13	Net profit (₹/kg) live weight	7.03	8.32	8.04	8.51	9.74

Bio-choline is lesser than synthetic choline chloride with better performance and livability. Further, they observed that the final cost of production was more economical in Bio-choline supplemented birds as compared to synthetic choline chloride supplemented birds. It indicates that the herbal choline can successfully replace synthetic choline chloride from broiler ration with substantial reduction in cost of production. Jadhav *et al.* (2008) reported that the economic impact analysis also revealed that the birds fed polyherbal formulations recorded higher net returns when compared with control as well as synthetic choline inclusion. In their experiment, the supplementation of both synthetic and polyherbal formulation resulted in overall improvement of broiler performance, economy, nutrient balance and biochemical parameters when compared with control and birds fed with synthetic choline. Similarly, Rodelas *et al.* (2011) indicated that supplementation of broiler diet with biocholine in combination with herbal vitamins C or E significantly reduced feed consumption, but significantly improved feed efficiency of the broilers and increased income over feed and chick cost.

CONCLUSION

It may be concluded that the herbal choline supplemented at dose levels of 0.35 kg/ton and 0.5 kg/ton of feed was found to be useful to improve the overall performance due to better growth resulting into increase in profit margin of broiler production replacing synthetic choline chloride (1kg/ton) in broiler diets. Moreover, the supplementation of herbal choline at 0.5 kg/ton of feed was more beneficial in terms of improved liver protection, carcass traits and economics of broiler production.

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