Original Research

Efficacy of Herbal Choline as a Replacement of Synthetic Choline Chloride in Diets on Growth Performance of Broilers

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Abstract

The aim of the present study was to evaluate the efficacy of supplementation of herbal choline as a replacement of synthetic choline chloride in diets on performance of broiler chickens. 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 of age. The treatment group T1 (control) offered basal diet (BIS, 2007) without synthetic choline chloride-60% (CC) or herbal choline (HC), group T2 basal diet with choline chloride-60% @ 1 kg/ton of feed, groups T3, T4 and T5 offered basal diets with herbal choline @ 0.250, 0.350 and 0.500 kg/ton of feed, respectively. The weekly growth performance parameters were studied up to 6th week of age in broiler chickens. The mean weekly body weight and weight gain of birds in group T5 group (0.5 kg/ton herbal choline) was significantly ($p<0.05$) higher than all treatment groups. Whereas, the statistically significant ($p<0.05$) higher mean weekly body weight and weight gain was observed in treatment groups T2 and T4 as compared to control group T1 fed diet without supplementation of any choline chloride. The birds from treatment group T5 fed diet containing herbal choline at 0.500 kg/ton of feed showed significantly higher ($p<0.05$) cumulative feed consumption than treatment group T2 fed diet at 1 kg/ton of synthetic choline. The treatment groups T2, T4 and T5 showed significantly better ($p<0.05$) cumulative feed conversion ratio than groups T1 and T3 (herbal choline at 0.250 kg/ton of feed). The higher survivability was observed in groups T2 and T5 compared to all the treatment groups. The highest mortality was observed in control group T1 fed diets without supplementation of synthetic choline chloride or herbal choline in feed. The post mortem examination from group T1 revealed specific lesions which indicated that the birds were died due to fatty liver syndrome. The highest EPEF was observed in treatment group T5 receiving diet at 0.500 kg/ton of herbal choline followed by treatment groups T2, T4, T3 and T1. The overall performance of the birds under treatment group T5 was found to be better. It may be concluded that supplementation of herbal choline at 0.350 and 0.500 kg/ton of feed replaces synthetic choline chloride-60% routinely added at 1 kg/ton of broiler feed. Moreover, the supplementation of herbal choline at 0.500 kg/ton of feed was more beneficial in terms of improving the bird's performance as revealed by European performance efficiency factor.

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Key words: Broilers, Growth, Efficiency, Herbal Choline, Synthetic Choline Chloride

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Introduction

The use of high energy diets aim at shortening the rearing period may increase metabolic disorders such as fatty liver syndrome (FLS) in broiler chickens (Leeson *et al.*, 1995). Increased abdominal fat pad (Corduk *et al.*, 2007), the 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 (Buyse *et al.*, 2001). 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. Acetylcholine is an agent released at the termination of the parasympathetic nerves. With acetylcholine, there is transmission of nerve impulses from presynaptic to postsynaptic fibers of sympathetic and parasympathetic nervous systems. In addition choline plays an essential role in fat metabolism in the liver. 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). Choline is thus 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 also acts as a methyl group donor, after oxidized to betaine. Betaine can be used to convert homocysteine to methionine in the transmethylation pathway in the 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. Supplementation of choline in poultry ration is well established to improve growth, performance and carcass quality in broilers (Attia *et al.*, 2005). 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 (Chatterjee and Misra, 2004; Muthukumarasamy *et al.*, 2004; Gangane *et al.*, 2010). The present study was undertaken to evaluate the efficacy of herbal choline as a replacement of synthetic choline chloride in diets on performance of broiler chickens up to 42 days.

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Materials and Methods

Experimental Design

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 on deep litter housing system. The treatment group T1 (control) offered basal diet (BIS, 2007) without synthetic choline chloride-60% (SC) or herbal choline (HC), group T2 offered basal diet with choline chloride-60% @ 1 kg/ton of feed, groups T3, T4 and T5 offered basal diet with supplementation of herbal choline @ 0.250, 0.350 and 0.500 kg/ton of feed, respectively. The experimental design and the details of the dietary treatments are presented in Table 1.

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
Total Number of Birds				**300**

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 (B1) 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 national standards (BIS, 2007) for pre-starter, starter and finisher phases as presented in Table 2. All the diets were isocaloric and iso-nitrogenous. The treatment group T2 incorporated synthetic choline chloride 60% at 1 kg/ton of feed and treatment groups T3, T4 and T5 incorporated herbal choline at 0.250, 0.350 and 0.500 kg/ton of feed, respectively in the basal diet.

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

Ingredients (%)	Pre-starter	Starter	Finisher
Maize	52.18	53.44	57.93
Soybean meal	40.7	38.2	32.9
Vegetable oil	3.1	4.4	5.25
Dicalcium phosphate (DCP)	1.8	1.8	1.8
Limestone powder (LSP)	1.2	1.2	1.2
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	0.2	0.18	0.15
L-Lysine	0.05	0.03	0.02
Toxin binder (UTPP)	0.1	0.1	0.1
Coccidiostat	0.05	0.05	0.05
Sodium bicarbonate	0.15	0.15	0.15
Total	**100**	**100**	**100**
Nutrient composition (% DM)			
Metabolizable energy (kcal/kg)	3003.83	3103.04	3202.76
Crude protein (%)	23.01	22	20.02
Ether extract (%)	5.48	6.8	7.78
Crude fiber (%)	4.21	4.09	3.88
Calcium (%)	1.01	1	0.98
Total phosphorus (%)	0.7	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

**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. A12.50MIU, 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.*

Data Collection

Data was collected on weekly weight changes determined by weighing the birds on weekly basis and replicate wise weight gain was calculated by subtracting the weight of the previous week from that of the current week. The feed intake was determined by subtracting the left-over feed from the feed offered, while feed conversion ratio was calculated as average feed intake divided by average weight gain taking into consideration of mortality, if any. The European Production Efficiency Factor (EPEF) was calculated based on live weight (LW), livability (LA), slaughter age (SA) and feed conversion ratio (FCR) using formula as described by Marcu *et al.* (2013). EPEF) = (Livability (LA) (%) x Live weight (kg))/ (Slaughter age (days) x FCR) x 100

Statistical Analysis

Data generated was subjected to statistical analysis by using Complete Randomized Design and the treatment means were compared by Critical Differences (Snedecor and Cochran, 1994).

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Result and Discussion

Live Body Weight and Cumulative Weight Gain

The various treatments had significant ($p<0.05$) effect on mean weekly live body weights of birds. The mean weekly body weights and cumulative weight gain of birds in group T5 fed diet containing herbal choline at 0.500 kg/ton of feed was significantly ($p<0.05$) higher than all treatment groups (Table 3).

Table 3: Weekly live body weight and cumulative weight gain of broilers fed different levels of herbal choline

Age (weeks)	Treatment Groups				
	T1 (Basal diet without SC/HC)	T2 (SC @ 1 kg/ton)	T3 (HC @ 0.250 kg/ton)	T4 (HC @ 0.350 kg/ton)	T5 (HC @ 0.500 kg/ton)
	Live Body Weights (g/bird)				
Day-old	44.63±0.69	44.7±0.24	44.43 ±1.62	44.83±0.41	45.27±0.27
I	145.63±0.74	151.97±3.09	156.85±2.16	155.07±2.12	158.28±3.06
II	413.93±0.55	434.63±2.75	436.80±4.69	437.17±5.44	451.00±5.46
II	875.30±6.40	906.57±10.39	903.23±11.92	901.39±0.53	928.23±5.32
IV	1447.97±11.14	1497.05±22.42	1492.37±14.93	1497.84±10.55	1551.43±5.53
V	2157.35±24.96	2192.83±50.86	2168.01±37.63	2210.36±29.71	2265.38±34.02
VI	2785.02±43.76	2817.65±54.34	2812.02±49.77	2843.49±32.61	2919.96±28.02
Pooled mean	1124.26±217.54^{a}	1149.35±220.23^{b}	1144.82± 218.80^{ab}	1155.74± 221.98^{b}	1188.51± 227.93^{c}
CD	23.496*				
	Cumulative Weight Gain (g/bird)				
I	101.00±0.60	107.20±2.91	112.42±3.73	110.23±1.97	113.02±2.79
II	369.30±0.50	389.87 2.57	392.37±6.23	392.34±5.28	405.73±5.21
II	830.67±5.85	861.80±10.41	858.80±10.64	856.56±0.17	882.97± 5.32
IV	1403.34±11.60	1452.28±22.50	1447.93±13.51	1453.01±10.15	1506.17±5.77
V	2112.72 ±25.58	2148.07±50.94	2123.58±36.35	2165.53±29.33	2220.12±34.19
VI	2740.38 ±44.39	2772.89±54.50	2767.59±48.59	2798.66±32.21	2874.69±28.08
Pooled mean	1259.57±227.2^{a}	1288.68±229.38^{b}	1283.78±227.72^{a}	1296.05±231.33^{b}	1333.78±237.40^{c}
CD	27.265*				

*Means bearing different superscripts within a row differ significantly. *P<0.05, CD-Critical difference.*

Whereas, the treatment groups T2 and T4 showed significantly ($p<0.05$) higher mean weekly body weights and weight gain than control group T1 fed diet without supplementation of herbal choline or synthetic chloride. The lower dose of herbal choline i.e. group T3 fed diet having 0.250 kg/ton also showed higher body weight but the difference was statistically non-significant compared to control group. The treatment group T2 and group T4 observed significantly higher ($p<0.05$) cumulative weight gain than control group T1 and T3 receiving diet with herbal choline at 0.250 kg/ton of feed. The treatment group T1 showed numerically lower cumulative weight gain than group T3, but difference was statistically non-significant (Table 3). In the present experiment, the data revealed that the birds from the group T1 receiving diet without herbal/synthetic choline chloride recorded the lowest live body weights among all treatment groups. The supplementation of herbal choline at 0.350 and 0.500 kg per ton of feed found to be effective in replacing the synthetic choline 60% at 1 kg/ton of feed in broiler diets in terms of live body weight and

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weight gain. These results are in agreement with Jadhav *et al.* (2008) reported that the weekly body weights were significantly ($p<0.05$) enhanced by supplementation of synthetic choline and herbal choline as compared to control i.e. low choline feed. Similarly, Sapcota *et al.* (2007) observed that that the supplementation of herbal growth promoters improved weight gain and FCR in broilers. The results of the present study showed that, regardless of the level of synthetic choline chloride and herbal source of choline used in the diets, the response in broiler performance was the same. This confirms previous observations that the herbal source of choline, produced from selected plants with high content of choline in esterified form and high bioavailability, can be an important alternative for synthetic choline chloride in corn-soybean meal diets for broilers from 0 to 42 days of age. Similarly, Kathirvelan *et al.* (2013) reported that the body weight of control group receiving diet without choline was significantly ($p<0.05$) less as compared to birds receiving diet with choline. They also concluded that supplementation of natural choline @1.0 kg/ton in broiler diet may be replaced synthetic choline (1kg/ton of feed) without affecting body weight gain.

Cumulative Feed Consumption

The birds from treatment group T5 received diet with herbal choline at 0.500 kg/ton of feed showed significantly higher ($p<0.05$) cumulative feed consumption than treatment group T2 (Table 4). The statistical difference was non-significant among treatment groups T1, T2, T3 and T4 for cumulative feed consumption. Higher feed intake was observed in group fed herbal choline at 0.500 kg/ton of feed as compared to groups supplemented with synthetic choline at 1 kg/ton of feed. However, the feed intake in all other treatment groups was comparable. Similarly, Sharma *et al.* (2015) reported that the feed intake in birds fed diets supplemented with herbal choline at 500gm per ton of feed was higher than birds fed diets supplemented with synthetic choline-60% at 1000 gm per ton of feed at 42nd day. Earlier, Waldroup *et al.* (2006) who observed that the choline supplemented in diet found to improve feed intake in broilers. The marginal improvement in feed intake observed in Bio-choline groups could be due to its plant ingredients containing a broad spectrum of vitamins, acids and alkaloids among many other active compounds mainly supposed to increase bile flow and improve feed intake. Natural biotin in Biocholine, lecithin and related constituents in lecithin extract might have enhanced the assimilation of dietary nutrients (Cengiz *et al.*, 2012). Increased weight gain with no change in feed intake and slight improvements in FCR by experimental diets suggested that lipotropic activities (especially Bio choline and choline chloride) improve the utilization of energy in the diet.

Cumulative FCR

The data revealed that the treatment groups T2, T4 and T5 showed significantly ($p<0.05$) better cumulative feed conversion ratio as compared to control group T1 receiving diet without synthetic choline or herbal choline and T3 receiving diet with herbal choline at 0.250 kg per ton of feed (Table 4).

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Table 4: Weekly cumulative feed consumption, feed conversion ratio (FCR), Mortality and EPEF of broilers under different dietary treatments

Age (weeks)	Treatment Groups				
	T1 (Basal diet without SC/HC)	T2 (SC @ 1 kg/ton)	T3 (HC @ 0.250 kg/ton)	T4 (HC @ 0.350 kg/ton)	T5 (HC @ 0.500 kg/ton)
	Cumulative Feed Consumption (g/bird)				
I	127.47±5.84	132.63±5.89	146.83±3.60	134.07±2.80	134.50±7.52
II	532.72±6.52	527.13±5.97	565.40±4.49	538.74±9.07	544.87±6.72
II	1223.79±13.72	1225.87±15.96	1250.78±14.73	1204.83±11.27	1240.00±19.25
IV	2219.01±16.64	2215.47±37.89	2226.87±23.25	2220.25±15.36	2267.20±16.23
V	3472.72±39.63	3448.52±62.80	3432.96±49.41	3477.00±42.98	3511.42±31.10
VI	4816.72±46.17	4749.58±92.06	4775.63±53.44	4800.24±52.63	4845.52±49.75
Pooled mean	2065.40±400.92^{ab}	2049.87±395.74^{a}	2066.41±394.18^{ab}	2062.52±399.85^{ab}	2090.58±403.26^{b}
CD	37.836*				
	Cumulative FCR				
I	1.26±0.06	1.24±0.07	1.31±0.07	1.22±0.02	1.19±0.06
II	1.44±0.02	1.35±0.02	1.44±0.03	1.37±0.03	1.34±0.01
II	1.47±0.01	1.42±0.00	1.46±0.03	1.41±0.01	1.40±0.02
IV	1.58±0.02	1.53±0.01	1.54±0.02	1.53±0.02	1.51±0.02
V	1.64±0.02	1.61±0.01	1.62±0.02	1.61±0.01	1.58±0.01
VI	1.76±0.02	1.71±0.01	1.73±0.02	1.72±0.01	1.69±0.01
Pooled mean	1.53±0.04^{b}	1.48±0.04^{a}	1.52±0.03^{b}	1.47±0.04^{a}	1.45±0.04^{a}
CD	0.033*				
	Mortality (%)				
Up to 42 days	5	1.67	3.33	3.33	1.67
	European Production Efficiency Factor (EPEF)				
42nd Day	352.52	379.01	369.04	375.53	399.27

*Means bearing different superscripts within a row differ significantly. * p<0.05, CD-Critical difference.*

The treatment groups T4 and T5 receiving diet with herbal choline at 0.350 and 0.500 kg/ton of feed, respectively, showed numerically better cumulative feed conversion ratio as compared to group T2 receiving diet with synthetic choline chloride at 1 kg/ ton of feed, but the difference was statistically non-significant. The treatment group receiving diet at 0.500 kg/ton of feed showed better feed conversion ratio than all treatment groups. However, the treatment groups supplemented with herbal choline at 0.350, 0.500 kg/ton and synthetic choline at 1 kg/ ton of feed showed improved FCR than non-supplemented choline group. Further, these observations also matched with the findings Bhanja *et al.* (2007) who stated that the herbal choline supplemented pullets produced significantly ($p<0.021$) more percent hen day eggs (84.72) along with significantly ($p<0.013$) better feed efficiency (1.34 vs 1.54) as compared to pullets fed with commercial feed grade choline. Similarly, Khosravinia *et al.* (2015) the inclusion of Bio choline and lecithin extract in the diet reported to significantly improve daily weight gain and FCR in broilers. Moreover, Giovani *et al.* (2017) reported that the biocholine supplementation has improved the FCR during the period of 14-28 days over synthetic choline chloride in broilers.

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Mortality (%)

The mortality percent in treatment groups T1, T2, T3, T4 and T5 was 5.00, 1.67, 3.33, 3.33 and 1.67, respectively (Table 4). The highest mortality was observed in treatment group T1 (control) diet without supplementation of synthetic choline chloride or herbal choline in feed. The post mortem examination from group T1 revealed general obesity with an enlarged, fatty liver and becomes soft and easily damaged which was suggestive of fatty liver syndrome. On microscopic observation sections revealed hepatocytes containing variably sized diffuse vacuoles of small, medium and large sizes with varying degree fat globules (Fig. 1). The higher survivability was observed in groups T2 and T5 as compared to all treatment groups. These findings are suggestive of beneficial effects of supplementation of herbal choline at 0.500 kg/ton of feed and synthetic choline chloride at 1 kg/ton of feed for improving the survivability rate in broiler chickens. Similarly, it was observed that birds fed diet with supplementatal choline/herbal source of choline offers complete protection to the liver (Sharma *et al*., 2015; Castro *et al.*, 2011; Reis *et al.*, 2012). The results in the present study are in confirmation with Lombardi *et al.* (1968) stating that choline deficient rats suffered from fatty liver due to an impaired release of hepatic triglycerides into plasma indicating the role of choline in regulating lipid metabolism and the results of the present study further confirms the same.

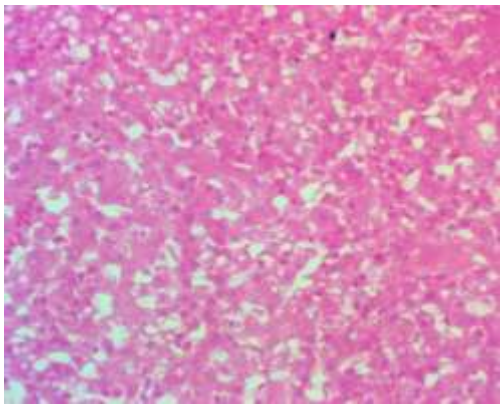

Fig.1: Liver showing various sizes vacuoles with fat droplets. European Production Efficiency Factor (EPEF)

The EPEF has been considered for evaluating the broiler performance index under different dietary treatments. The highest EPEF was observed in treatment group T5 receiving diet at 0.500 kg/ton of herbal choline followed by treatment groups T2, T4, T3 and T1 (Table 4). Thus, the overall performance of the birds under treatment group T5 was found to be better. The performance efficiency assessment by calculating EPEF was positively influenced by the supplementation of herbal choline 0.500 Kg/ton of feed as compared to all other treatment groups. Khosravinia *et al.* (2015) also indicated that the performance efficiency index was improved in the birds fed with biocholine.

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Conclusion

It may be concluded that supplementation of herbal choline at 0.350 and 0.500 kg/ton of feed replaces synthetic choline chloride-60% routinely added at 1 kg/ton of broiler feed. Moreover, the supplementation of herbal choline at 0.500 kg/ton of feed was more beneficial in terms of improving the bird's performance as revealed by European performance efficiency factor.

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References

1. Attia, W.A., El-Ganzory E.H. and Hassan R.A. 2005. Growth, carcass quality & serum constituents of slow growing chicks as affected by Betaine addition to and diet containing on different levels of choline. *J. Poult. Sci.*, 4(11):840-850.
2. Bhanja, S.K., Agarwal S.K., Elangovan A.V., Bhattacharyaya and Verma J. 2007. Proceed. XXIV IPSACON, 25-27th April, Ludhiana, 109:5-49.
3. BIS, Bureau of Indian Standards 2007. Poultry Feeds Specification. (5thRevision), Manak Bhavan, 9, Bahadur Shah Zafar Marg, New Deli-11 110002.
4. Buyse, J., Janssens G.P. and Decuypere E. 2001. The effects of dietary L-carnitine supplementation on the performance, organ weights and circulating hormone and metabolite concentrations of broiler chickens reared under a normal or low temperature schedule. *British Poultry Science*, 42: 230–241.
5. Castro, S.F., Fortes, B. D. A., Carvalho J.C.C., Bertechini A. G., Queiroz L.F.B. and Garcia Jr. A.A.P. 2011. Relacao metionina e dietetic sorbe o desempenho de codornas japonesas (*Coturnix Coturnix Japonica*) em postura. *Ciencia Animal Brasileira*, 12 (4): 635-641.
6. Cengiz, O., Hess J.B. and Bilgili S.F. 2012. Dietary biotin supplementation does not alleviate the development of footpad dermatitis in broiler chickens. *Journal of Applied Poultry Research*, 21: 764-769.
7. Chatterjee, S. and Misra S.K. 2004. Efficacy of herbal biocholine in controlling fatty liver syndrome in commercial broilers on high metabolic energy diet. *Fhytomedica*, 5: 37-39.
8. Corduk, M., Ceylan N. and Ildiz F. 2007. Effects of dietary energy density and L-carnitine supplementation on growth performance, carcass traits and blood parameters of broiler chickens. *South African Journal of Animal Science*, 37: 65–73.
9. Gangane, G.R., Gaikwad N.Z., Ravikanth K. and Maini S. 2010. The comparative effects of synthetic choline and herbal choline on hepatic lipid metabolism in broilers. *Veterinary World*, 3(7): 318-320.
10. Giovani Farina, Alexandre de Mello Kessler, Patricia Diniz Ebling, Fabio Ritter Marx, Ricardo Cesar and Andrea Machado Leal Ribeiro 2017. Performance of broilers fed different dietary choline sources and levels. *Ciencia Animal Brasileira*, 18: 1-14,
11. Gopi, M., Purushothaman M.R. and Chandrasekaran D. 2014. Effect of dietary coenzyme Q10 supplementation on the growth rate, carcass characters and cost effectiveness of broiler fed with three energy levels. *Springer Plus*, 3: 518.

 DOI 10.5455/ijlr.20180402101714

12. Jadhav, N.V., Nagbhushana V., Shivi Maini and Kartikesh S.M. 2008. An Evaluation of comparative effects of feeding synthetic and herbal choline on broiler performance, nutrient balance and serum activities. *Veterinary World*, 1(10): 306-309.
13. Kathirvelan, C., Chandrasekaran D., Vasanthakumar P. and Purushothaman M.R. 2013. Effect of replacement of synthetic choline with herbal choline on growth performance of broilers. *Indian Journal of Animal Nutrition*, 30(2): 184-187.
14. Khosravinia, H., Chethen P.S., Umakantha B. and Nourmohammadi R. 2015. Effects of lipotropic products on productive performance, liver lipid and enzymes activity in broiler chickens. *Poultry Science Journal*, 3 (2): 113-120.
15. Leeson, S. and Summers J.D. 2005. Commercial Poultry Nutrition, third ed. Nottingham Univ. Press, Nottinghan, UK.
16. Lombardi, B., Pani P. and Schlunk F.F. 1968. Choline deficiency fatty liver: impaired release of hepatic triglycerides. *Journal of Lipid Research*, 9: 437-446.
17. Marcu, A., Vacaru-Opris I., Dumitrescu G., Ciochina L.P., Marcu A., Nicula M. 2013. The influence of genetics on economic efficiency of broiler chickens growth. *Animal Science and Biotechnologies*, 46(2): 339-346.
18. Muthukumarasamy B., Sahu B. K., Swain R. K. and Samantaray D. P. 2004. Studies on the effect of biocholine supplementation in commercial broilers. *Indian Journal of Poultry Science*, 39(3):246-251.
19. Reis, S.R., Barreto, S.L.B., Paula E., Muniz J.C.L., Viana G.S.V. and Mencalha R. (2012) Niveis de suplememtacao de calina na dieta de codornas japonesas em postura. *Revista Brasileira de Agropecuaria Sustentavel*, 2 (1): 118-123.
20. Sapcota, D., Baruah K.K. and Upadhyay T.N. 2007. Proceed. XXIV IPSACON, 25-27th April, Ludhiana, 112: 5-56.
21. Sharma, A. and Ranjan S. 2015. Effect of herbal and chemically synthetic choline on physio-biochemical characteristic of chicks. *Journal of Global Biosciences*, 4(6): 2537-2542.
22. Snedecor G.W. and Cochran W.G. (1994) Statistical methods, 8th edition. Journal of Educational and Behavioral Statistics. 19(3): 304-307.
23. Van Emous, R.A., Kwakkel R.P., Van Krimpen M.M. and Hendriks W.H. 2015. Effects of dietary protein levels during rearing and dietary energy levels during lay on body composition and reproduction in broiler breeder females. *Poultry Science*, 94: 1030-1042.
24. Waldroup, P.W., Motl M.A., Yan F. and Fritts C.A. 2006. Effects of betaine and choline on response to methionine supplementation to broiler diets formulated to industry standards. *Journal of Applied Poultry Research*, 15: 58–71.
25. Xu C.F., Yu C.H., Xu L., Sa X.Y. and Li Y.M. 2010. Hypouricemic therapy: a novel potential therapeutic option for nonalcoholic fatty liver disease. Hepatology, in press. doi: 10.1002/hep.23798.
26. Zeisel S.H., Dacosta K.A., Youssef M. and Hensey S. 1989. Conversion of dietary choline to trimethylamine and dimethylamine in rats: dose-response relationship. *Journal Nutrition*, 119:800-804.

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