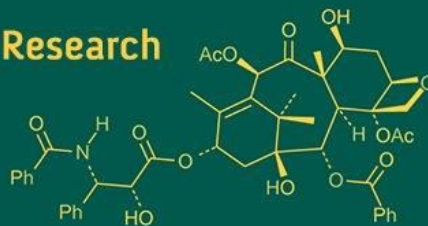
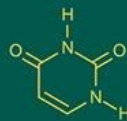
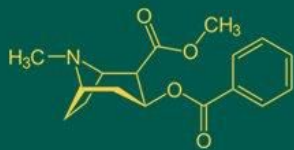


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Effects of different levels of choline chloride supplementation on the growth performance, digestibility of nutrients and haematological profile of broiler birds

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Abstract

The study was conducted to investigate the effect of supplementing different levels of choline chloride on the performance of (Vencobb 400) broilers. Two hundred day old chicks from a single hatch and average body weights (47.62 ± 0.29 g) were allocated randomly into 5 treatment groups (T₁, T₂, T₃, T₄ and T₅) with 40 birds in each group. Standard broiler pre-starter, starter and finisher rations as per Bureau of Indian Standard (BIS), 2007 was prepared and fed to the T₁ (control) group. All the birds were fed iso-caloric and iso-nitrogenous with or without choline chloride supplementation. The remaining four groups were fed standard rations incorporated with choline chloride at 250, 500, 750 and 1000 mg per kg of feed for T₂, T₃, T₄ and T₅ groups respectively. During the feeding trial of 42 days (6 weeks), no significant difference ($p > 0.05$) was observed in the weekly feed intake and total feed intake among the different treatment groups. Significant difference ($p < 0.05$) was observed on the 4th week and higher significant difference ($p < 0.01$) was observed on the 5th and 6th week on the weekly body weights of the experimental broiler birds. Average daily gain showed significant difference ($p < 0.01$) and feed conversion efficiency also showed significant difference ($p < 0.05$) among the different treatment groups. There was no significant difference ($p > 0.05$) observed due to supplementation of choline chloride. Haematological parameters revealed no significant difference ($p > 0.05$) in the hemoglobin, PCV, RBC, WBC and monocyte counts while the lymphocyte count showed high significant difference ($P < 0.01$) on the 42nd day of blood collection. It was concluded that choline chloride could be supplemented to broiler ration preferably at 1000 mg per kg of feed for better performance.

Keywords: Broilers, choline chloride, growth performance, nutrient digestibility, haematology profile

Introduction

Choline is an essential nutrient considered part of the B-complex vitamins and is available in forms like choline chloride for dietary supplementation. Choline has many biological functions such as its role as the precursor and component of lecithin and sphingomyelin, which are found in all cell membranes and membranes of most cell organelles (Rucker *et al.*, 2007) ^[1]. Choline is able to convert excess fat from the liver into lecithin and increase the utilisation of fatty acids from the liver (McDonald *et al.*, 2010) ^[2]. Choline is also necessary for the nervous system as it is a precursor to the neurotransmitter acetylcholine and also has a metabolic role, where it act as a source of labile methyl groups (Combs, 2008) ^[3]. Zeisel and da Costa (2009) ^[4] reported that the de novo synthesis of choline is possible but is not sufficient to meet the requirements. This implies that a dietary source of choline is necessary to meet the overall requirements of a chicken.

Many studies have suggested supplementation of choline at higher levels than the recommended levels of BIS (2007) ^[5], which is currently recommended at 500 mg/kg of feed, for the better health and production in broilers. An updated definition of choline requirements may contribute to minimizing feed costs because currently used levels vary considerably between feed manufacturers.

Gregg *et al.* (2023) ^[6] conducted an experiment on modern broiler chickens and found significant reduction ($p < 0.05$) in feed intake in finisher and overall period and also reported improved FCR in broilers fed increasing levels of choline chloride per kg of feed.

Kaninathan *et al.* (2023) ^[7] reported most ($p < 0.01$) average daily gain (ADG) and average daily feed intake (ADFI) and lowest ($P < 0.01$) FCR when 1000 mg choline chloride (60%) was added to the basal diet of broiler chickens. They also reported improved ADG and FCR when choline chloride was added to a fatty-liver induction diet. Many research articles are not available on the effect of graded levels of choline chloride on the digestibility of nutrients and haematological profile. The observations by Igwe *et al.* (2015) ^[8] showed that there were significant differences ($P < 0.05$) in the RBC, WBC, lymphocyte, monocyte, and basophil counts while there were no significant differences in the PCV, Hb, MCV, MCHC, MCH and eosinophil count. Considering all the potential beneficial effects of choline supplementation for the poultry production, the present study was conducted to study the effects of supplementing different levels of choline chloride on the growth performance as well as haematological profile of Vencobb 400 broiler birds.

Materials and Method

Location and period of study

The study was conducted in the livestock farm of the College of Veterinary science & Animal Husbandry, Selesih, Aizawl, Mizoram. It was conducted from first week of November and the feeding trial was carried out for 42 days (6weeks).

Approval of Institutional Animal Ethics Committee (IAEC): The study was approved by Institutional Animal Ethics Committee (registered under CPCSEA) of College of Veterinary Science and Animal Husbandry, Central Agricultural University (Imphal), Selesih, Aizawl, Mizoram (Registration number and date of registration.1476/GO/Re/SL/2011/CCSEA, 29/07/2024).

Animals and experimental design

Two hundred day old chicks (Vencobb 400) from a single hatch and average body weights (47.62 ± 0.29 g) were allocated randomly into 5 treatment groups (T_1 , T_2 , T_3 , T_4 and T_5) and housed in a well-ventilated animal shed. Birds were raised under deep litter system of management. The birds were fed two times daily with broiler pre-starter ration (1-7 days of age), broiler starter ration (8-21 days of age) and broiler finisher ration (22-42 days of age). Fresh drinking water was provided at all times. The duration of the experiment was 42 days (6weeks). Body weight of the broiler birds and feed intake were determined every week. Average daily gain (ADG) and Feed conversion efficiency of experimental broiler birds were also determined.

Experimental diets

Choline chloride of 98% purity was used for this experimental study. The first treatment group (T_1) was fed the standard basal diet as per BIS (2007) ^[5] without any choline chloride supplementation and served as the control group. Birds in T_2 were fed standard feed supplemented with choline chloride at the rate of 250 mg/kg of feed. Likewise, birds T_3 , T_4 and T_5 were fed standard feed supplemented with choline chloride at the rates of 500

mg/kg, 750 mg/kg and 1000 mg/kg respectively.

Chemical analysis

The representative samples of the concentrate feeds, residuals of feeds as well as faeces were analysed as per the methods described by AOAC (2000) ^[9]. The parameters calculated were dry matter (DM; method no. 934.01), total ash (TA; method no. 942.05), crude protein (CP; Kjeldahl nitrogen $\times 6.25$; method no. 976.05), ether extract (EE; method no. 920.39) and crude fibre (CF; method no. 962.09). The nitrogen free extract contents in samples were calculated by subtracting CP, total ash, EE and CF from DM content.

Feeding Trial

The complete duration of feeding trial was 42 days (6 weeks). The daily feed intake was calculated by recording the daily feed offered and residual feed. The body weight of the broiler birds were taken at day old and at 7 days interval during the experimental period of 42 days.

Metabolic trial

At the end of the feeding trial, a 3 days long metabolic trial was conducted. Two days adaptation period was given to the selected representative birds in the metabolic cages before the actual metabolic trial. The feed offered and residue left within 24 hours were recorded for each bird to estimate daily feed intake. Faeces voided during 24 hours was collected and weighed for each bird under metabolic trial. These samples were analysed for dry matter content.

Haematological analysis

On the 21st day and 42nd day of the feeding trial, blood was collected aseptically from the jugular/wing vein of the broiler birds using a 5ml scalp vein needle and kept in an EDTA vial to prevent blood clotting. Proper care was taken in order to prevent any stress to the birds. Haematological variables were determined from whole blood using MS4S2 automatic hematology analyser following standard procedure of the manufacturer (HD Consortium India Pvt Ltd., New Delhi, India).

Statistical Analyses

The data obtained were analysed by one-way analysis of variance (ANOVA) in a completely randomized design using SPSS software. Significance was declared when probability values were $p \leq 0.05$ and $0.05 > p \leq 0.10$ was declared as trend. The results are presented as the mean $\pm$ SEM and compared using Duncan's multiple range test.

Results and Discussion

Analysed Nutrient content of Experimental diets

The chemical composition of pre-starter, starter and finisher experimental diets fed to the different groups of experimental broiler birds are depicted in Table 1, 2 and 3 respectively. As per BIS, 2007 ^[5] specification, all the formulated feeds for the different groups were made iso-nitrogenous and iso-caloric for broiler pre-starter, starter and finisher diets respectively.

Table 1: Formulated and chemical composition of experimental broiler pre-starter diet

Ingredients (kg)	T-1	T-2	T-3	T-4	T-5
Maize crush	50	50	50	50	50
Rice Polish	5	5	5	5	5
Soyabean Meal	30.7	30.7	30.7	30.7	30.7
Fish Meal	7.5	7.5	7.5	7.5	7.5
Vegetable Oil	3.75	3.75	3.75	3.75	3.75
Choline chloride	0	0.025	0.05	0.075	0.1
DCP	0.75	0.75	0.75	0.75	0.7
Limestone Powder	0.7	0.68	0.65	0.65	0.65
Coccidiostat	0.04	0.04	0.04	0.04	0.04
Toxin Binder	0.03	0.03	0.03	0.03	0.03
Vitamin Premix	0.02	0.02	0.02	0.02	0.02
Antioxidant	0.01	0.01	0.01	0.01	0.01
Mineral Mixture	1	1	1	1	1
Iodised Salt	0.5	0.5	0.5	0.5	0.5
Total	100	100	100	100	100
Chemical composition (% DM basis)					
Dry matter (%)	89.56	88.92	89.43	89.32	89.26
Crude protein (%)	23.16	23.09	23.11	23.05	23.12
Ether extract (%)	3.73	3.12	3.49	3.30	3.27
Crude fibre (%)	3.50	3.51	3.50	3.48	3.47
Total ash (%)	9.39	9.36	9.64	9.59	9.07
Nitrogen free extract (%)	60.22	60.92	60.26	60.58	61.07
Calcium (%)	1.08	1.04	1.34	1.03	1.19
Phosphorus (%)	0.73	0.96	0.94	0.88	0.79
ME (kcal/kg)*	2996.9	2996.9	2996.9	2996.9	2996.9
Lysine (%)*	1.33	1.33	1.33	1.33	1.33
Methionine (%)*	0.53	0.53	0.53	0.53	0.53
Threonine (%)*	0.30	0.30	0.30	0.30	0.30

* Calculated value

Table 2: Formulated and chemical composition of experimental broiler starter diet

Ingredients (kg)	T-1	T-2	T-3	T-4	T-5
Maize crush	53	53	53	52.97	53
Rice Polish	5	5	5	5	5
Soyabean Meal	29.25	29.25	29.25	29.25	29.25
Fish Meal	5.5	5.5	5.5	5.5	5.5
Vegetable Oil	4.5	4.5	4.5	4.5	4.5
Choline chloride	0	0.025	0.05	0.075	0.1
DCP	0.5	0.5	0.5	0.5	0.5
Limestone Powder	0.65	0.65	0.6	0.6	0.55
Coccidiostat	0.04	0.04	0.04	0.04	0.04
Toxin Binder	0.03	0.03	0.03	0.03	0.03
Vitamin Premix	0.02	0.02	0.02	0.02	0.02
Antioxidant	0.01	0.01	0.01	0.01	0.01
Mineral Mixture	1	1	1	1	1
Iodised Salt	0.5	0.5	0.5	0.5	0.5
Total	100	100	100	100	100
Chemical composition (% DM basis)					
Dry Matter (%)	89.93	89.59	90.13	89.95	89.55
Crude protein (%)	22.24	22.21	22.23	22.25	22.20
Ether extract (%)	4.20	3.89	3.66	4.01	4.01
Crude fibre (%)	2.49	2.30	2.20	2.50	2.49
Total ash (%)	7.22	7.55	7.92	7.86	7.21
Nitrogen free extract (%)	63.85	64.05	63.99	63.38	64.09
Calcium (%)	1.47	1.34	1.08	1.19	1.52
Phosphorus (%)	0.99	0.90	0.77	0.80	0.78
ME (kcal/kg)*	3099.12	3099.12	3099.12	3099.12	3099.12
Lysine (%)*	1.21	1.21	1.21	1.21	1.21
Methionine (%)*	0.49	0.49	0.49	0.49	0.49
Threonine (%)*	0.28	0.28	0.28	0.28	0.28

* Calculated value

Table 3: Formulated and chemical composition of experimental broiler finisher diet

Ingredients (kg)	T-1	T-2	T-3	T-4	T-5
Maize crush	58.75	58.75	58.75	58.75	58.75
Rice Polish	3.5	3.5	3.5	3.5	3.45
Soyabean Meal	24	24	24	24	24
Fish Meal	6	6	6	6	6
Vegetable Oil	5.25	5.25	5.25	5.25	5.25
Choline chloride	0	0.025	0.05	0.075	0.1
DCP	0.5	0.5	0.5	0.45	0.45
Limestone Powder	0.4	0.4	0.4	0.4	0.4
Coccidiostat	0.04	0.04	0.04	0.04	0.04
Toxin Binder	0.03	0.03	0.03	0.03	0.03
Vitamin Premix	0.02	0.02	0.02	0.02	0.02
Antioxidant	0.01	0.01	0.01	0.01	0.01
Mineral Mixture	1	1	1	1	1
Iodised Salt	0.5	0.5	0.5	0.5	0.5
Total	100	100	100	100	100
Chemical composition (% DM basis)					
Dry matter (%)	90.31	91.05	90.91	91.36	90.75
Crude protein (%)	20.16	20.13	20.15	20.14	20.12
Ether extract (%)	4.10	4.17	4.18	4.13	4.12
Crude fibre (%)	3.04	2.91	2.86	3.01	3.07
Total ash (%)	7.10	6.76	6.81	6.75	7.03
Nitrogen free extract (%)	65.60	66.03	66.00	65.97	65.66
Calcium (%)	1.24	1.46	1.69	1.30	1.79
Phosphorus (%)	0.76	0.82	0.96	0.79	0.92
ME (kcal/kg)*	3197.08	3197.08	3197.08	3197.08	3197.08
Lysine (%)*	0.99	0.99	0.99	0.99	0.99
Methionine (%)*	0.46	0.46	0.46	0.46	0.46
Threonine (%)*	0.26	0.26	0.26	0.26	0.26

* Calculated value

Feed intake

The average weekly feed intake by the experimental broiler birds was recorded (Table 4). Statistical analysis revealed no significant difference ($p>0.05$) on the weekly feed intake among the different treatment groups throughout the feeding trial. The present study was supported by Gregg *et al.*

(2022) ^[10] where they observed no significant difference ($p>0.05$) on the feed intake among the broiler chickens when increasing levels (0, 400, 800, 1200, 1600 and 2000 mg) of choline chloride per kg of feed was added to the different experimental diets.

Table 4: Weekly feed intake (g/bird) by the experimental broiler birds of different groups

Age	T-1	T-2	T-3	T-4	T-5	P Value
1 st week	91.77±3.84	92.08±2.35	90.98±4.10	91.13±1.97	96.60±4.65	0.782 ^{NS}
2 nd week	187.03±1.90	188.16±0.94	187.11±1.88	189.03±0.53	188.14±0.71	0.809 ^{NS}
3 rd week	444.18±5.40	446.29±9.35	444.49±8.78	449.80±6.15	450.43±8.71	0.965 ^{NS}
4 th week	683.95±4.06	683.29±3.25	698.32±4.43	696.76±8.28	691.77±2.57	0.144 ^{NS}
5 th week	795.88±7.98	791.90±12.44	796.60±11.63	806.72±3.40	800.95±1.73	0.783 ^{NS}
6 th week	1058.67±9.56	1064.39±2.21	1054.98±8.46	1060.18±2.04	1057.53±0.52	0.838 ^{NS}

NS=non-significant

Growth performance

The average weekly body weight of the experimental broiler birds was recorded (Table 5). There was no significant difference ($p>0.05$) observed from the initial body weight of the experimental birds till the 3rd week of the feeding trial. However, there was significant difference ($p<0.05$) observed on the 4th week, as well as higher significant difference ($p<0.01$) was observed on the 5th and 6th weeks. The average daily gain was significant difference ($p<0.01$) among the different treatment groups and feed conversion efficiency was significantly better ($p<0.05$) in T₄ and T₅ upon statistical analysis (Table 6). The lowest values were observed for T₁ (control) group while the highest values were observed for the T group which was supplemented with the most amount of choline chloride for this experiment at 1000 mg per kg of feed. Our observations

were in agreement with Khose *et al.* (2018) ^[11], who observed that supplementing synthetic choline chloride to broilers at 1000 mg/kg produced higher body weight than the control group throughout the rearing period. Similarly, Ghasura *et al.* (2021) ^[12] saw significant increase ($p<0.05$) in overall body weight of broilers at 1000 g/t of choline chloride supplementation in the diet. This study was also supported by Hossain *et al.* (2014) ^[13], who observed that weight gain differed ($p<0.05$) among the treatment groups with lowest weight gain reported for the unsupplemented group. Similarly, Farina *et al.* (2017) ^[14] found higher overall weight gain in choline chloride supplemented groups compared to the control group. Recent study by Kaninathan *et al.* (2023) ^[7] recorded better overall FCR in broilers supplemented with 1000mg/kg of choline chloride.

Table 5: Weekly body weight (g/bird) of the experimental broiler birds of different group

Age	T-1	T-2	T-3	T-4	T-5	P Value
Initial body wt.	47.42±0.46	47.88±0.31	47.59±0.30	47.67±0.21	47.53±0.17	0.863 ^{NS}
1 st week	121.36±0.85	120.51±0.83	121.64±0.90	121.09±0.82	122.32±1.03	0.691 ^{NS}
2 nd week	229.32±6.88	232.29±2.63	233.59±2.69	237.72±5.39	249.07±5.65	0.094 ^{NS}
3 rd week	503.00±8.27	512.50±17.61	521.22±18.22	526.16±15.51	553.06±7.26	0.186 ^{NS}
4 th week	871.00±14.09 ^b	905.50±14.96 ^a	897.95±6.53 ^{ab}	914.77±11.22 ^a	923.87±2.00 ^a	0.035 [*]
5 th week	1261.65±4.09 ^c	1334.00±14.05 ^{ab}	1318.25±16.27 ^{ab}	1294.30±12.10 ^{bc}	1350.92±19.77 ^a	0.004 ^{**}
6 th week	1757.10±14.68 ^c	1768.50±16.49 ^{bc}	1765.62±15.84 ^{bc}	1812.30±15.12 ^{ab}	1839.90±14.25 ^a	0.006 ^{**}

Means bearing different superscripts in a row differ significantly

[NS = non-significant, * = $P < 0.05$, ** = $P < 0.01$]

Table 6: Effect of different levels of choline chloride supplementation on feed intake, average daily gain and feed conversion efficiency during experimental period (42 days)

Attributes	T-1	T-2	T-3	T-4	T-5	P Value
Initial body weight (g)	47.42 ± 0.46	47.88 ± 0.31	47.59 ± 0.30	47.67 ± 0.21	47.53 ± 0.17	0.863 ^{NS}
Final body weight (g)	1757.10 ± 14.68 ^c	1768.50 ± 16.49 ^{bc}	1765.62 ± 15.84 ^{bc}	1812.30 ± 15.12 ^{ab}	1839.90 ± 14.25 ^a	0.006 ^{**}
Body weight gain (g)	284.95 ± 2.42 ^c	286.77 ± 2.71 ^{bc}	286.34 ± 2.63 ^{bc}	294.10 ± 2.55 ^{ab}	298.73 ± 2.36 ^a	0.006 ^{**}
Average daily gain (g/day)	40.71 ± 0.35 ^c	40.97 ± 0.39 ^{bc}	40.91 ± 0.37 ^{bc}	42.01 ± 0.36 ^{ab}	42.67 ± 0.34 ^a	0.006 ^{**}
Total feed intake (g)	3261.49 ± 14.31	3266.11 ± 18.93	3265.93 ± 16.85	3293.62 ± 17.38	3291.98 ± 10.41	0.453 ^{NS}
Feed conversion efficiency	1.78 ± 0.02 ^a	1.77 ± 0.02 ^a	1.77 ± 0.01 ^a	1.74 ± 0.01 ^{ab}	1.70 ± 0.01 ^b	0.024 [*]

Means bearing different superscripts in a row differ significantly

[NS = non-significant, * = $P < 0.05$, ** = $P < 0.01$]

Digestibility of nutrients

From the perusal of Table 7, it was observed that the digestibility of nutrients of the experimental broiler birds did not show significant differences ($p > 0.05$) among the different treatment groups during the metabolic trial. Our observations align with studies by Vilas (2023) [15] who did

not find appreciable increase in digestibility of nutrients in choline supplemented broiler birds. Similarly, Chitithoti *et al.* (2024) [16] found no significant difference in the digestibility of nutrients when synthetic choline chloride was added in the diet of broilers.

Table 7: Nutrient digestibility of the experimental broiler birds of different groups

Digestibility co-efficient (%)	T-1	T-2	T-3	T-4	T-5	P Value
Dry matter	74.83±0.94	75.51±1.69	75.08±1.39	75.51±1.20	75.72±1.98	0.262 ^{NS}
Crude protein	73.81±1.84	78.53±5.33	78.88±6.14	79.86±0.86	80.75±3.91	0.390 ^{NS}
Crude fibre	44.27±1.10	45.63±5.65	44.83±3.31	45.11±2.25	47.23±3.98	0.422 ^{NS}
Ether extract	77.23±3.60	78.37±4.35	78.48±6.78	79.75±2.03	80.09±3.22	0.786 ^{NS}
Nitrogen free extract	82.99±2.11	83.77±2.94	79.46±2.09	80.90±2.62	81.59±3.09	0.062 ^{NS}

NS = non-significant

Haematological profiles

Statistical analysis showed significant difference ($p < 0.01$) on the lymphocyte count on the 42nd day (Table 8). The lymphocyte count progressively increase across the treatment groups. Maia *et al.*, 2025 [17] had reported that choline can positively influence lymphocytes as it supports phosphatidylcholine dependent membrane synthesis,

regulates immune cell development through methylation pathways, and enhances cholinergic signalling involved in lymphocyte activation. In accordance with our observations, Khose *et al.* (2018) [11] reported significant increase ($p < 0.01$) in lymphocyte % with synthetic choline chloride supplementation than control group in broilers on the 42nd day of rearing.

Table 8: Effect of different levels of choline chloride supplemented diets on haematological profile in experimental broiler birds

Blood	T-1	T-2	T-3	T-4	T-5	P Value	Significance
Hemoglobin (g/dl)							
21 st day	13.87 ± 0.72	13.67 ± 0.40	12.70 ± 0.30	12.82 ± 0.45	13.27 ± 0.13	0.304	NS
42 nd day	11.57 ± 0.80	13.15 ± 0.48	12.82 ± 1.14	11.15 ± 0.33	12.20 ± 0.69	0.326	NS
PCV (%)							
21 st day	24.07 ± 2.93	20.42 ± 1.02	19.07 ± 0.84	17.60 ± 1.59	21.02 ± 1.91	0.186	NS
42 nd day	26.92 ± 2.44	28.37 ± 1.42	25.90 ± 2.00	26.00 ± 1.18	27.82 ± 1.90	0.838	NS
RBC (x10⁶/μl)							
21 st day	2.02 ± 0.20	1.71 ± 0.09	1.65 ± 0.10	1.46 ± 0.14	1.78 ± 0.18	0.183	NS
42 nd day	2.25 ± 0.21	1.96 ± 0.05	1.87 ± 0.11	1.66 ± 0.16	1.97 ± 0.18	0.167	NS
WBC (x10³/μl)							
21 st day	26.50 ± 0.84	21.82 ± 0.86	23.67 ± 1.16	23.57 ± 1.04	23.63 ± 1.17	0.071	NS
42 nd day	21.67 ± 0.50	22.66 ± 0.81	22.84 ± 0.97	22.76 ± 1.24	23.27 ± 0.90	0.797	NS
Lymphocyte (%)							
21 st day	26.05 ± 1.08	30.05 ± 1.77	30.25 ± 2.42	31.25 ± 1.34	31.57 ± 0.86	0.159	NS
42 nd day	27.45 ± 0.99 ^b	32.45 ± 1.34 ^a	33.55 ± 1.30 ^a	34.07 ± 1.15 ^a	34.52 ± 1.32 ^a	0.006	**

Monocyte (%)							
21 st day	2.52 ± 0.05	2.80 ± 0.12	2.70 ± 0.17	2.85 ± 0.09	2.90 ± 0.04	0.145	NS
42 nd day	2.67 ± 0.05	2.87 ± 0.12	2.70 ± 0.16	2.60 ± 0.09	2.87 ± 0.11	0.346	NS

Means bearing different superscripts in a row differ significantly

[NS = non-significant, * = $P < 0.05$, ** = $P < 0.01$]

Conclusion

Based on the data observed on this present study, it could be conclude that choline chloride at different levels (250, 500, 750 and 1000 mg/kg of feed) did not have any adverse effects on the growth performance, nutrient digestibility as well as haematological profile of broiler birds. Incorporation of broiler feed with choline chloride could be carried out, preferably at 1000 mg/kg of feed for better performance since better results were observed at this level.

References

- Rucker RB, Zemleni J, McCormick DB, Suttie JW. Handbook of Vitamins. 4th ed. Boca Raton: Taylor and Francis Group; 2007.
- McDonald P, Edwards RA, Greenhalgh JFD, Morgan CA, Sinclair LA, Wilkinson RG. Animal Nutrition. 7th ed. Harlow: Pearson; 2010.
- Combs GF Jr. The Vitamins: Fundamental Aspects in Nutrition and Health. 3rd rev. ed. San Diego: Academic Press; 2008.
- Zeisel SH, da Costa KA. Choline: an essential nutrient for public health. Nutrition Reviews. 2009;67(11):615-623. doi: 10.1111/j.1753-4887.2009.00246.x
- B.I.S. Poultry Feeds Specification, IS: 1374-2007. 5th Revision Edn. New Delhi: Manak Bhaban; 2007.
- Gregg CR, Hutson BL, Flees JJ, Lowman ZS, Estes KA, Starkey JD, *et al.* Evaluation of increasing concentrations of supplemental choline chloride on modern broiler chicken growth performance and carcass characteristics. Animals. 2023;13(9):1445. doi: 10.3390/ani13091445
- Kaninathan A, Subramaniyam S, Marimuthu S, D'Souza P. Impact of polygenic formulation on performance and fatty liver disease of broiler chickens. Online Journal of Animal Feed Research. 2023;13(2):89-96.
- Igwe IR, Okonkwo CJ, Uzoukwu UG, Onyenegecha CO. The effect of choline chloride on the performance of broiler chickens. Annual Research & Review in Biology. 2015;8(3):1-8.
- AOAC. Official Methods of Analysis. 15th ed. Arlington (VA): Association of Official Analytical Chemists; 2000.
- Gregg CR, Tejeda OJ, Spencer LF, Calderon AJ, Bourassa DV, Starkey JD, *et al.* Effect of dietary choline chloride supplementation on growth performance and carcass characteristics of broiler chickens reared to 32 days of age. Poultry. 2022;1(2):66-73.
- Khose KK, Manwar SJ, Sedmake ED, Ingole RS, Rathod PR. Supplementation of herbal choline in diets on haematological indices and development of vital organ in broilers. Journal of Entomology and Zoology Studies. 2018;6(6):1054-1058.
- Ghasura AS, Savaliya FP, Rajpura RM, Patel AB, Bhagora NJ, Patel NM, *et al.* Effect of dietary supplementation of choline from different sources on performance and economics of commercial broiler chicken. Indian Journal of Veterinary Science and Biotechnology. 2021;17(2):81-86.
- Hossain ME, Das GB, Hasan MM, Shaikat AH, Bari ASM. The effect of choline chloride supplementation on performance parameters and carcass characteristics of broiler. Iranian Journal of Applied Animal Science. 2014;4(2):373-378.
- Farina G, Kessler ADM, Ebling PD, Marx FR, Cesar R, Ribeiro AML, *et al.* Performance of broilers fed different dietary choline sources and levels. Ciência Animal Brasileira. 2017;18:1-14.
- Vilas DM. Effect of dietary supplementation of herbal choline on performance of broiler chicken. MVSc. Thesis. Nagpur: Maharashtra Animal and Fishery Sciences University; 2023.
- Chitithoti AK, Thullimalli S, Murikipudi NS, Metta M, Ganguly B. Evaluation of the effect of synthetic and herbal choline supplements on haemato-biochemistry, redox balance and nutrient digestibility in broilers. Journal of Animal Research. 2024;14(2):107-114.
- Maia C, Fung CW, Sanchez-Lopez E. Choline in immunity: a key regulator of immune cell activation and function. Frontiers in Immunology. 2025;16:1617077. doi: 10.3389/fimmu.2025.1617077