



Efficacy of Vincholine (A Herbal Choline) in Improving the Performance of Commercial Broilers

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Abstract

Choline is an essential micronutrient for the fast growing commercial broilers. It plays crucial role in mobilization of excess fat accumulated in the adipocytes and instigates lipid metabolism. This yields huge energy that supports rapid growth of the birds. Choline chloride is routinely used as a feed additive in poultry ration. However, it exhibits many demerits. Vincholine, a natural choline, formulated by Vinayak Ingredients (India) Pvt. Ltd. is an alternative to the synthetic choline chloride.

A trial was conducted in broiler birds (n = 420) randomized into 7 treatment groups including one control, one choline chloride (1000 g/tonne), three Vincholine (150, 250 and 300 g/tonne), and two competitor (400 and 750 g/tonne) groups. The experiment was continued for 42 days. The results showed that Vincholine treatment significantly improved the body weight and FCR of the birds as compared to the control group. The carcass quality was better in Vincholine group as compared to the control. The serum biochemistry data showed that Vincholine treatment ensured optimum triglyceride, cholesterol and ALT level among the birds indicating enhanced fat mobilization and metabolism. The liver histopathology data revealed superior results in Vincholine group than control. In the present trial, Vincholine 300 g/tonne feed was found most efficient in all growth performances of the birds.

In conclusion, Vincholine is a promising herbal feed supplement and an excellent alternative to the synthetic choline chloride.

Keywords: Commercial Broiler; Fat metabolism; Fat mobilization; Performance trait; Vincholine Herbal Choline

Introduction

Poultry industry has a considerable role in the global food chain [23]. It is one of the promising sectors in the agri-business platform. Commercial broiler production has emerged as a smart farming business because of its higher return on investment [6,13]. Due to rapid growth rate and low feed conversion efficiency (FCR), broiler chickens are a vital source of protein for human consumption.

According to Watt Executive Guide, 2015, India stands 7th in chicken meat production worldwide [32]. As per the statistical data of 2017, India produces 4.2 million tonnes of broiler per annum [28,30].

In commercial broilers choline plays pivotal role in regulating the lipid metabolism. Choline is a constituent of the vitamin B complex. It is the fundamental constituent in phospholipid synthesis

[10]. It forms acetylcholine, the molecule required for the neuro-transmission. Choline, as a part of very-low-density lipoprotein (VLDL), facilitates lipid mobilization out of hepatic tissue and prevents fatty liver syndrome [22]. It plays a significant role as a lipotropic factor; thus helps to reduce the fat deposition in the body. It is a donor of methyl group that helps to perform various cellular functions [19]. Inadequate supply of choline in diet causes interrupted fat metabolism leading to accretion of excess fat in abdomen and visceral tissue. Choline deficiency produces fatty changes in liver and the incidences of perosis among the young birds [22]. Hence, the delayed growth and poor performance among the broiler birds noticed.

Adding synthetic choline chloride in ration is a common practice in broiler farming. However, synthetic choline has evidenced many demerits and low margin of safety. It is highly hygroscopic and corrosive in nature. It shows poor absorption rate and loss of vitamins [22]. It causes accumulation of trimethylamine (TMA) in the gut of the bird developing a fishy taint in the edible tissues of the broiler [8]. Moreover, supplementation of choline chloride in poultry diet upsurges its manufacturing cost. Hence, an alternative to the synthetic choline chloride is being extensively investigated. Many herbs possess choline-like property and may exert outstanding effects when added to the ration of the birds [22].

Myriads of literature reported the hepatoprotective and anti-lipogenic potential of a number of medicinal plants or plant-based formulations [5]. They play crucial role as antioxidant and counteract with the harmful reactive oxygen species (ROS) produced in tissues. They enable the cellular metabolism and maintain homeostasis. They facilitate in conversing the fatty change of liver by lipid mobilization [12]. Previous reports suggest that herbs may exert choline-like effect in removing the excess abdominal fat and prevent obesity [9,22]. Therefore, herbs may serve as natural source of choline and improve performance of the broiler birds by supporting body weight gain, optimum FCR, and enhanced muscle mass [9,16,22]. The present trial was conducted to evaluate the impact of Vincholine, a natural choline formulation prepared by Vinayak Ingredients (India) Pvt. Ltd, at various concentration levels in commercial broiler birds. The performance parameters like growth, dressing percentages, organ weights, serum biochemistry and liver histopathology etc. were evaluated.

Methods

Birds

Day old broiler birds (n = 420) were procured from a reputed hatchery and maintained at the facility of Omega Laboratories,

Satara, India. The care and handling of the birds were performed according to the national and international guidelines. Standard brooding, feeding and watering procedures were carried out from day old till the end of trial. All birds were vaccinated with an appropriate schedule as mentioned in table 1.

Table 1: Vaccination schedule.

Vaccine	Time
Lasota, 1 drop by intraocular route	day 7, 21
IBD vaccine, by intraocular route	day 14

Diet

All birds received the diet in 2 experimental phases like Starter and Finisher. The diet was formulated according to the description of table 2.

RM	Ingredient	Starter	Finisher
	MAIZE (9% MOISTURE)	551.7	621.7
	HIPRO SOYA 49%	366.5	301.4
	SOYA/ RB OIL (FFA<5, PV<4)	35	40
	LSP	10	8
	DCP	16	10.7
	SODA	2.5	2.8
	Salt	2.8	2.2
Additives	VITAMIN PREMIX (VITAMIX)	0.55	0.5
	PREMIX	0.5	0.5
	ANTI OXIDANT (ENDOX DRY)	0.15	0.15
	CHOLINE CHLORIDE/ VINCHOLINE (HERBAL CHOLINE) **	00	00
	TOXIN BINDER (AVATOX)	1	1
	EMULSIFIER (LIPTOX XT)	0.5	0.45
	ANTICOCCIDIALS- MADURA+ NICARB (GLAVITRO)	0.6	0.6
	LYSINE SULPHATE	3	2
	D L METHIONINE	3.2	2.5
	L THREONINE	1.5	1
	BMD 10%	0.5	0.5
	PHYTASE 5000 FTU (QUANTUM BLUE)	0.15	0.15
	PROTEASE WITH NSPase (GLAVENZA DUO)	0.35	0.35
	MOULD INHIBITOR (MOLDZAP)	0.6	0.6
	LIVER TONIC (LIV52)	0.5	0.5
	BETAINE HCL	0.7	0.7
	ACIDIFIER	1	1
ANTIDIARRHOEAL/GUT MODULATOR	0.5	0.5	
	TOTAL	1000	1000

Table 2: Basal diet formula for control groups.

** required quantity was added in the respective groups as per the trial protocol.

Study design

The broiler birds obtained from a reputed hatchery and housed in the poultry research station of Omega Laboratories at Satara, Maharashtra, India. The birds were quarantined and acclimatized prior to initiation of the experiment. The experimental room was maintained with standard temperature, relative humidity and light and dark cycle as per guideline throughout the study period. The

birds were provided with chlorinated drinking water and poultry mash feed ad libitum and tagged with appropriate identification number. The broiler birds (n = 420) were randomized and allotted into 7 treatment groups including one basal diet group. The treatment groups 2 to 7 were provided with different levels of choline chloride/herbal choline in the entire study (table 3). Each group had 60 birds. The experiment was continued for 42 days.

Groups	Treatment 1	Treatment 2	Treatment 3	Treatment 4	Treatment 5	Treatment 6	Treatment 7
	Basal diet	Vincholine	Vincholine	Vincholine	Choline Chloride 60%	Competitor 1	Competitor 2
	No Choline	150 g/tonne	250 g/tonne	300 g/tonne	1000 g/tonne	400 g/tonne	750 g/tonne
No of birds/replicate	20	20	20	20	20	20	20
No of replicates	3	3	3	3	3	3	3
Total no of birds	60	60	60	60	60	60	60

Table 3: Experimental design.

Assessment parameters

Weekly average body weight, feed consumption, feed conversion ratio (FCR), clinical signs, morbidity and mortality of the broiler birds were examined and recorded. Feed consumption was calculated by deduction of feed residue (remaining at the end of week) from the feed offered. The FCR was calculated by using the formula:

$$FCR = \frac{\text{Total feed consumed}}{\text{Final weight} - \text{initial weight}}$$

The postmortem examination of the birds died during experiment was carried out and the findings were recorded. At the end of 42 days, From each experimental group, 10 birds were sacrificed for further analysis. The blood sample from the birds was collected aseptically in clot activator tubes and serum was separated for the evaluation of biochemical parameters like cholesterol, triglycerides, total proteins, albumin, globulins, AST, ALT and BUN and creatinine. Liver samples were collected and stored in 10% formalin for histopathological examination. Dressing percentages, weights of thigh, weights of breast and weights of liver of the birds were measured and recorded.

Table 4: Methods used to assess various parameters.

	Parameters	Method used
1	Serum ALT IU/L	Merill Biochemistry analyser
2	Serum AST IU/L	
3	Serum Cholesterol mg/dl	
4	Total serum Protein g/dl	
5	Serum Albumin g/dl	
6	Serum globulin g/dl	
7	Serum BUN mg/dl	
8	Serum creatinine mg/ml	
9	Organ weights- liver, thigh, breast	Digital weighing balance
10	Liver histopathology	H&E stain- paraffin embedding technique
11	Statistical analysis	Dunnett’s test for comparing several treatments

Statistical analysis

At the end of the trials, the raw study data were compiled and expressed as Mean. The standard error of mean (SEM) was calculated. Dunnett’s test was used for comparing the treatment groups. $p < 0.05$ was accepted as statistically significant value.

Result

Growth performance

In the present study, the broiler birds (n = 420) were assigned into 7 treatment groups. The birds of treatment group 1 received only basal diet whereas, other groups obtained various quantity of herbal choline or synthetic choline chloride added in their basal

diet. The study was commenced while the birds were day old and continued up to 6 weeks of their age. The body weight of the birds was measured weekly interval till end of the study and FCR value was calculated.

At 6th week of age, the birds of treatment group 4 showed significantly higher body weight as compared to the birds of rest of the groups (table 5, figure 1A). FCR values (table 6, figure 1B) were significantly lower in group 3 and 4 as compared to other groups. The treatment group 1 revealed least body weight and noticeably high FCR. However, there was no difference in the liveability percentage of the birds. In each group 1% mortality was observed (Table 7).

	Treatment 1	Treatment 2	Treatment 3	Treatment 4	Treatment 5	Treatment 6	Treatment 7
Week 6 body weights	2419.55 ^f	2594.67 ^{2e}	2789.55 ^b	2847.21 ^{7a}	2730.13 ^{3c}	2668.06 ^{7d}	2770.13 ^{6b}
SEM	46.23	41.43	38.55	40.54	51.65	49.51	46.66

Table 5: Body weights (6th week).

*Body weights were significantly higher in group Treatment 4 as compared to all other groups. Treatment 1 group showed significantly lower body weights than the other treatment groups.

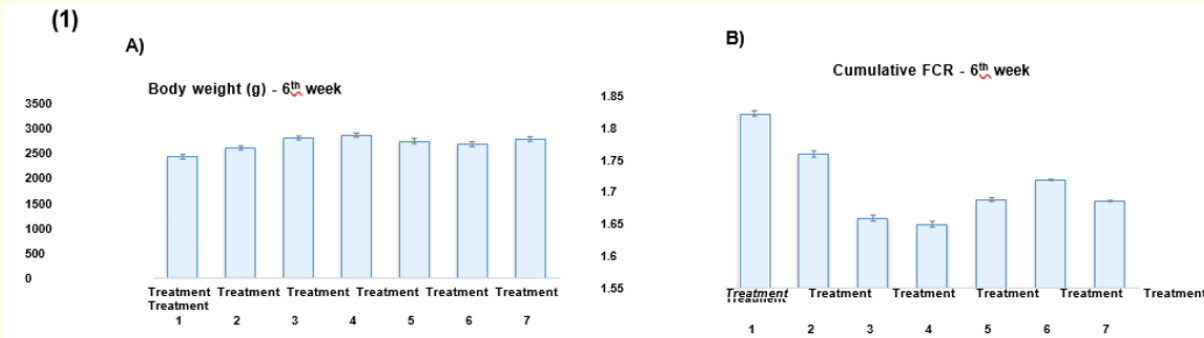


Figure 1: Histograms showing Body weights (A) and FCR (B) of the broilers (6th week) at various treatment groups.

	Treatment 1	Treatment 2	Treatment 3	Treatment 4	Treatment 5	Treatment 6	Treatment 7
FCR Week 6	1.822653 ^e	1.759375 ^d	1.659407 ^a	1.650032 ^a	1.688562 ^b	1.719222 ^c	1.686199 ^b
SEM	0.004258	0.004866	0.00468	0.004586	0.003134	0.000918	0.000947

Table 6: Cumulative FCR (6th week).

*FCR values were significantly lower in group Treatment 3 and 4 as compared to all other groups. Treatment 1 group showed significantly higher FCR than the other treatment groups.

	Treatment 1	Treatment 2	Treatment 3	Treatment 4	Treatment 5	Treatment 6	Treatment 7
Mortality	1	1	1	1	1	1	1
Findings	Nonspecific septicaemia and toxaemia due to yolk sac retention in first week						

Table 7: Mortality %.

Dressing percentages and organ weights

Dressing percentage and organ weights are the vital parameters indicating the performances of the broiler birds. In the recent trial, the factors like dressing percentage, thigh weight, breast weight, abdominal fat and liver weight were measured and compared among the groups (figure 2).

As shown in figure 2A, the dressing percentage was noticeably lower in treatment group 1 as compared to the birds of other treatment groups.

Thigh weights of the broiler birds (figure 2B) were significantly higher in treatment groups 4, 5, 6 and 7 as compared to the treatment groups 1, 2 and 3. The study data showed lowest value of the thigh weight in treatment group 1.

The broiler birds of the treatment group 7 showed maximum breast weights as presented in figure 2C. The breast weight was lowest among the birds of the treatment group 1.

Further, the birds assigned for treatment 1 revealed maximum abdominal fat and least liver weight (figure 2D and E).

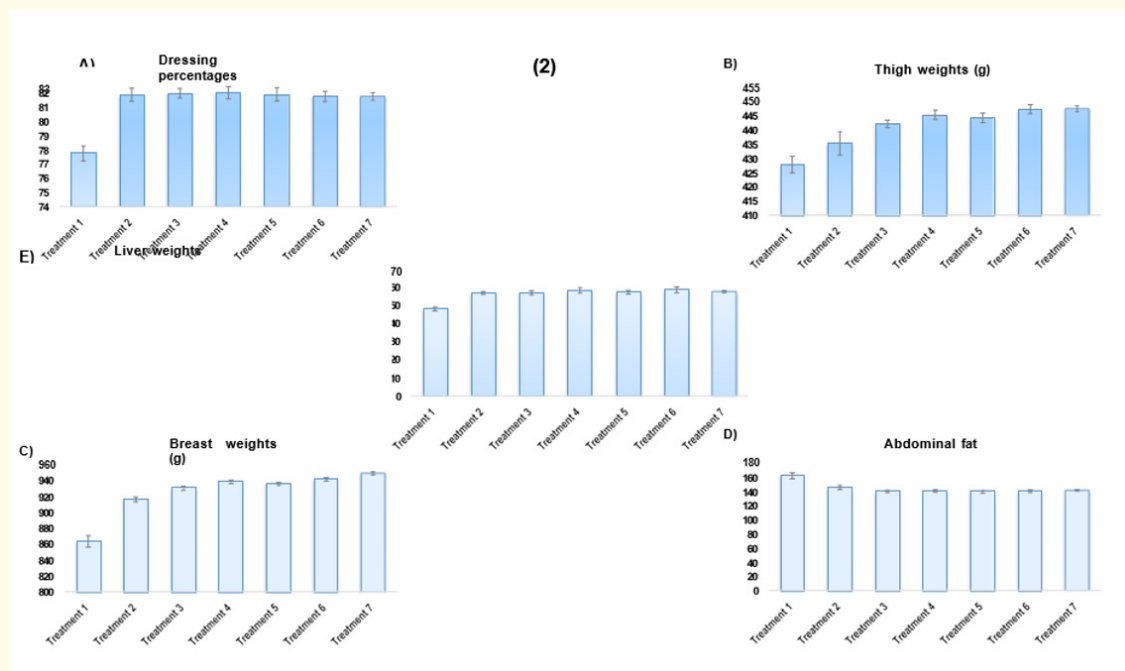


Figure 2: Histograms showing A) Dressing percentage, B) Thigh weight, C) Breast weight, D) Abdominal fat percentage and E) Liver weight of the broilers at different treatment levels.

Serum biochemistry

At the end, 10 birds from each group were randomly selected and sacrificed to perform serum biochemistry. The parameters like cholesterol, triglycerides, total proteins, albumin, globulins, AST, ALT, BUN and creatinine were assessed (figure 3).

Treatment group 1 showed significantly lower total protein, albumin and globulin levels as compared to the birds of all other groups. AST, ALT, cholesterol, triglyceride and BUN values were maximum in treatment group 1 as compared to all other groups.

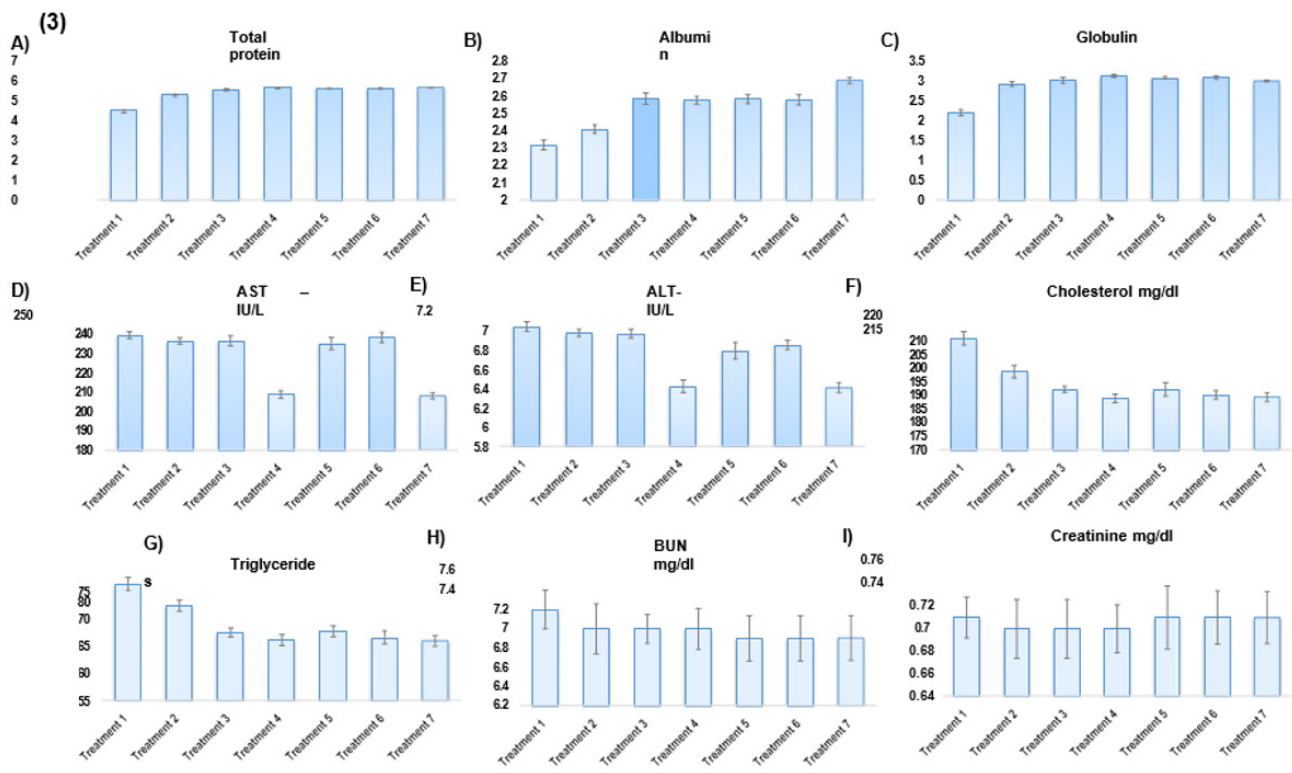


Figure 3: Serum biochemical parameters of the broiler birds allotted in various treatment groups.

The results showed that, the treatment groups 4 and 7 had significantly lower AST and ALT values than the serum samples of other groups. However, the treatment groups 4, 6 and 7 showed significantly lower cholesterol values than the birds of the remaining groups. The birds of the treatment groups 4 and 7 showed significantly lower triglyceride levels.

Treatment 1 group showed significantly higher BUN values than the other groups. There is no significant difference amongst the treatment groups for the creatinine levels. Although, the values were within the normal physiological ranges.

Histopathology of liver

The histopathology was performed with the liver samples of the birds (n = 10) sacrificed at the end of the trial. The study data exhibited that, the fatty change scores and the hepatocyte necrosis scores were significantly higher in treatment group 1 as compared to other groups, Treatment 4, 6 and 7 showed significantly lower fatty change scores of hepatocytes. The birds of the treatment groups 3, 4, 5, 6 and 7 showed significantly lower hepatocyte necrosis scores. The histopathological findings of liver was graphically presented in histograms in figure 4.

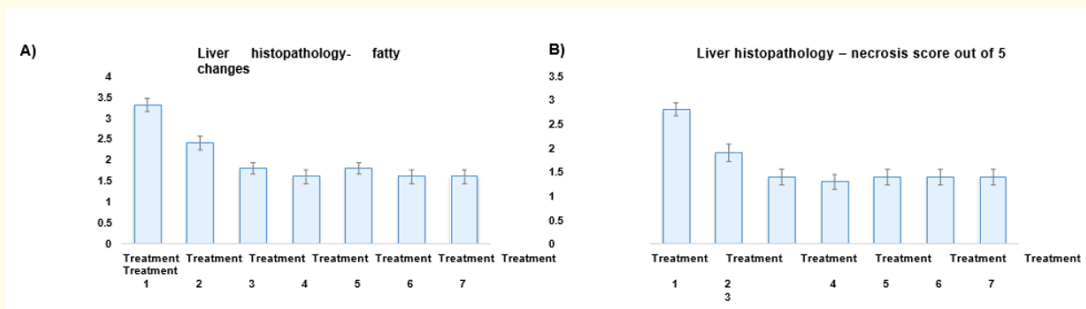


Figure 4: Histopathological interpretation of liver tissue of the broiler birds at various treatment levels.

Discussion

Choline is an essential micro-nutrient required for growth and performance of the broilers. Since birds cannot synthesize sufficient amount of choline, broiler ration is made with substantial amount of choline supplement [19,21]. In spite of a number of advantages, synthetic choline chloride unveils many demerits like hygroscopicity, corrosiveness, TMA residue in edible tissue etc. [25]. Vinayak Ingredients (India) Pvt. Ltd. has manufactured a natural choline formulation, Vincholine as an alternative to the synthetic choline. Vincholine is an excellent product that regulates the lipid metabolism and plays significant role in mobilization of excess abdominal fat. Lipid metabolism undergoes an important role in poultry birds as it is a huge source of energy. Fat breaks down into fatty acid which enters into the mitochondrial beta-oxidation pathway and releases energy [27]. The adipose tissue is commonly known as the fat depot of the body. The enzyme lipase catalyzes the fat metabolism and helps to mobilize the fat from its storage to other tissues for energy production [31]. Thus, it prevents obesity and fatty changes of liver. Synthetic choline chloride is a requisite feed additive in commercial broiler ration that facilitates in diminution of the hepatic and abdominal fat and improves the performance of the birds [14]. Vincholine is a natural replacement of synthetic choline chloride and free from any adverse reactions. The recent trial corroborated the efficacy of herbal feed additive Vincholine in commercial broiler birds (Cobb 430 Y). Here birds were randomized into 7 groups including control, Vincholine (n = 3, 150, 250 and 300 g/tonne), choline chloride 60% (1000 g/tonne), and competitors or marketed reference products (n = 2, 400 and 750 g/tonne). The trial was initiated in day old chicks and continued for 42 days. At 6th week of age the birds having diet supplemented with Vincholine showed maximum body weight and optimum FCR value. The

mortality among the experimental broiler birds of all treatment groups observed in the first week of the trial due to the nonspecific septicaemia and toxemia as a result of the yolk sac retention. The trial data revealed that the broiler birds receiving ration comprising of 300 g/tonne Vincholine showed the superior performance amongst the trial groups including competitors and synthetic choline chloride as well.

The serum biochemical parameter ALT is a significant marker of hepatic function. Elevated level of ALT indicates the accumulation of fat in liver causing hepatic inflammation [24]. In the recent study, optimum ALT level in Vincholine treated group denotes effective mobilization of fat from the adipose tissue of the birds and trimming down of accumulated fat in hepatic tissue. Further, fatty acid is stored as triglycerides after esterifying with glycerol and serves as the lipid depot for future utilization. Cholesterol is another major constituent of lipid either absorbed through dietary food or synthesized in liver. Inefficient lipid metabolism causes abnormally increased level of serum triglyceride and cholesterol [7]. Our study report revealed optimum triglyceride and cholesterol level in Vincholine group of the experimental birds as compared to the control. This ensures efficient lipid metabolism among the broilers after Vincholine supplementation in their ration. The histopathological findings of hepatic tissue evidenced the results corresponding to the serum biochemical analysis. The fatty changes of liver and hepatocyte necrosis were lesser in Vincholine added group as compared to the untreated control birds. The carcass quality of the birds demonstrated significant changes in Vincholine supplemented groups as compared to the untreated control. The dressing percentage, thigh, breast and liver weight were significantly higher in birds received Vincholine feed supplement as compared to the

control. In contrast, the abdominal fat level was significantly lower in Vincholine group as compared to the birds of control group. Eventually, the broiler birds treated with Vincholine at a concentration of 300 g/tonne feed showed superior results in all growth performances in the trial. This results evidenced that Vincholine was effective in mobilization of accumulated fat from adipose tissue and viscera and helped to transport excess fat for muscle development. This led to higher weight gain and enhanced performance of the broiler birds. In accordance with previous reports scientists claim that many herbal formulations may exert choline-like property and help to lessen the deposited abdominal and hepatic fat when supplemented in broiler ration [1,20]. Hence, herbs may exhibit significant role as lipotropic factor. In agreement with earlier research findings, our trial data evidenced the fact of lipid mobilization of herbal formulation Vincholine. Choline is an important nutrient that triggers transport of accumulated fat from hepatic tissue to the extrahepatic space and instigates lipid metabolism. This yields huge energy which helps to build-up muscle protein resulting in body weight gain and rapid growth of the birds [11]. Consequently, superior performance trait is acquired. This phenomenon is widely accepted by the scientists and nutritionists. Choline chloride is an excellent feed additive that prevents fatty liver disease and induces lipolysis [2]. However, the proof of detrimental effects of synthetic choline chloride has inspired the investigators to find out choline from vegetable sources. Various literatures reported the potential effect of plants in lipid metabolism and hepatoprotection [15,29]. Herbal active ingredients play considerable role in reducing triglyceride and cholesterol level promoting growth and performance of the broiler birds [3,4]. *In vivo* study in rodents showed that plant-derived polyphenols may improve the acetaminophen-induced hepatotoxic condition [26]. Studies showed that polyherbal formulation may exert anti-lipogenic effect in methionine-choline deficiency (MCD) diet-induced hepatosteatosis in rodents [5]. Therefore, herbs may contribute significantly in lipolysis and hepatoprotection influencing the boosting up of overall performance of the broilers. Our experiment data are in concordance with the previous findings.

Conclusion

In a nutshell, Vincholine is a marvellous feed supplement derived from the natural source and efficiently promotes growth, performance and improvises the zootechnical parameters of the

broiler birds. It acts in declining the carcass fat content and increment of muscular growth attributing to better lipid mobilization and metabolism. Vincholine at a concentration of 300 g/ton of feed in broiler deficient in choline-diet showed superior efficacy in overall growth performances of the broilers. Further research is needed to explore the molecular pathway through which Vincholine proves its potential effect.

Bibliography

1. Abdel-Wareth A A and Lohakare J. "Bioactive Lipid Compounds as Eco-Friendly Agents in the Diets of Broiler Chicks for Sustainable Production and Health Status". *Veterinary Sciences* 10.10 (2023): 612.
2. Azadmanesh V and Jahanian R. "Effect of supplemental lipotropic factors on performance, immune responses, serum metabolites and liver health in broiler chicks fed on high-energy diets". *Animal Feed Science and Technology* 195 (2014): 92-100.
3. Betancourt L., et al. "Effects of Colombian oregano essential oil (*Lippia origanoides* Kunth) and *Eimeria* species on broiler production and cecal microbiota". *Poultry Science* 98.10 (2019): 4777-4786.
4. Brenes A and Roura E. "Essential oils in poultry nutrition: Main effects and modes of action". *Animal Feed Science and Technology* 158.1-2 (2010): 1-14.
5. Chandrasekaran PR., et al. "Evaluation of lipotropic effect of herbal formulation on hepatic fat accumulation in rats fed with methionine-choline deficient diet". *Pharmacognosy Magazine* 15.66 (2019).
6. Chatterjee RN and Rajkumar U. "An overview of poultry production in India". *Indian Journal of Animal Health* 54.2 (2015): 89-108.
7. Cox RA and García-Palmieri MR. "Cholesterol, Triglycerides, and Associated Lipoproteins". In: Walker HK, Hall WD, Hurst JW, editors. *Clinical Methods: The History, Physical, and Laboratory Examinations*. 3rd edition. Boston: Butterworths; 1990. Chapter 31.

8. D'souza P and Selvam R. "Evaluation of polyherbal formulation in broilers fed high energy diet: Implications on zootechnical parameters, fat accretion, and serum L-carnitine levels". *Journal of Advanced Veterinary and Animal Research* 9.1 (2022): 166.
9. Ding X., et al. "The effects of plant extracts on lipid metabolism of chickens—A review". *Animal Bioscience* 36.5 (2023): 679.
10. Farina G., et al. "Performance of broilers fed different dietary choline sources and levels". *Ciência Animal Brasileira* 18 (2017): e37633.
11. Fouad AM., et al. "Nutritional requirements of meat-type and egg-type ducks: what do we know?". *Journal of Animal Science and Biotechnology* 9 (2018): 1-11.
12. Gangane G R., et al. "The Comparative effects of synthetic choline and herbal choline on hepatic lipid metabolism in broilers". *Veterinary World* 3.7 (2010): 318.
13. Goswami A. Understanding The Economics Of Broiler Chicken Production, Poultry Trends, May, (2021).
14. Hossain ME and Das GB. "Effects of supplemental choline on deposition of cardiac, hepatic and abdominal fat in broiler" (2014).
15. Kannan N., et al. "Protective Effect of Acacia nilotica (L.) against Acetaminophen-Induced Hepatocellular Damage in Wistar Rat". *Advances in Pharmacological and Pharmaceutical Sciences* 2013.1 (2013): 987692.
16. Kathirvelan C., et al. "Effect of replacement of synthetic choline with herbal choline on growth performance of broilers". *Indian Journal of Animal Nutrition* 30.2 (2013): 184-187.
17. Khose K K., et al. "Efficacy of herbal choline as a replacement of synthetic choline chloride in diets on growth performance of broilers". *Journal of Livestock Research* 8.10 (2018): 313-322.
18. Khose K., et al. "Replacement of synthetic choline chloride by herbal choline in diets on liver function enzymes, carcass traits and economics of broilers". *Journal of Animal Research* 9.1 (2019): 87-93.
19. Parsons CM and Leeper RW. "Choline and methionine supplementation of layer diets varying in protein content". *Poultry Science* 63.8 (1984): 1604-1609.
20. Petrolli T G., et al. "Vegetable choline as a replacement for choline chloride in broiler feed". *Acta Scientiarum. Animal Sciences* 43 (2021): e53265.
21. Pour H A., et al. "Effect of choline on performance quality of non-ruminant". *Entomology and Applied Science Letters* 1.1-2014 (2014): 14-18.
22. Ramalho de Lima M., et al. "Choline supplementation: Impact on broiler chicken performance, steatosis, and economic viability from 1 to 42 days". *Plos one* 19.3 (2024): e0295488.
23. Ravindran V. "The role of poultry in human nutrition". In: *POULTRY DEVELOPMENT REVIEW* (2013): 66-69.
24. Ruhl C E and Everhart JE. "Determinants of the association of overweight with elevated serum alanine aminotransferase activity in the United States". *Gastroenterology* 124.1 (2003): 71-79.
25. Selvam R., et al. "Evaluation of polyherbal formulation and synthetic choline chloride on choline deficiency model in broilers: Implications on zootechnical parameters, serum biochemistry and liver histopathology". *Asian - Australasian Journal of Animal Science* 31.11 (2018): 1795-1806.
26. Subramanya S B., et al. "Therapeutic potential of plants and plant derived phytochemicals against acetaminophen-induced liver injury". *International Journal of Molecular Sciences* 19.12 (2018): 3776.
27. Talley J T and Mohiuddin S S. "Biochemistry, fatty acid oxidation". (2020).
28. Team Pashudhan Praharee. "Status of poultry production in India". Pashudhan Praharee. November, 2019. www.pashudhanpraharee.com (2019).
29. Tranchida F., et al. "Metabolomic and lipidomic analysis of serum samples following curcuma longa extract supplementation in high-fructose and saturated fat fed rats". *PLoS One* 10.8 (2015): e0135948.
30. Vetrivel S and Chandrakumarmangalam S. "Status of Poultry in India". *Brazilian Journal of Poultry Science* 1981 (2013).
31. Wang S., et al. "Lipolysis and the integrated physiology of lipid energy metabolism". *Molecular Genetics and Metabolism* 95.3 (2008): 117-126.
32. Watt Executive Guide. Poultry Trends (2015).