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Effect of Low Protein Diets with or without L-Tryptophan Supplementation on Growth and Carcass Characteristics in Commercial Broilers

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Abstract

The study was conducted to evaluate the performance of broilers fed with low protein diets supplemented by L-tryptophan. On cumulative basis, Body weight gain was significantly ($p<0.05$) reduced by step down of dietary crude protein levels of 1.5 per cent unit without L-tryptophan supplementation and L-tryptophan supplementation significantly ($p<0.05$) improved body weight gain, but not at the dietary crude protein reduction of 2.25 per cent unit. The feed consumption during the starter phase significantly ($p<0.05$) reduced by step down of dietary crude protein levels at 2.25 per cent unit without L-tryptophan supplementation and L-tryptophan supplementation significantly ($p<0.05$) improved the feed consumption. On cumulative basis, there was no significant difference in the feed conversion ratio up to 0.75 per cent unit with or without L-tryptophan supplementation and further dietary crude protein reduction resulted in inferior feed efficiency as compared to control group. Carcass yield, Breast muscle, Thigh muscle and Drumstick yield significantly ($p<0.05$) reduced by step down of dietary Crude protein levels at 2.25 per cent unit without L-tryptophan supplementation and L-tryptophan supplementation significantly ($p<0.05$) increased the carcass yield, breast muscle yield, thigh muscle yield and drumstick yield. Abdominal fat as a per cent of body weight significantly ($p<0.05$) increased by step down of dietary crude protein levels of 1.5 per cent unit without L-tryptophan supplementation and L-tryptophan supplementation significantly ($p<0.05$) reduced abdominal fat but the not at the crude protein reduction of 2.25 per cent unit.

Keywords: Broilers, Crude protein, Body weight gain, Feed consumption, Feed efficiency, Carcass characteristics

Broiler production in India has continuously faced challenges of providing optimum environment for maximum growth, production, disease control and finally the cost benefit ratio involved for a successful poultry husbandry practices. It is estimated that 25 per cent of the world's meat supply is derived from poultry. India being in tropical region is most congenial for poultry production. India is the 5th largest producer of poultry meat in the world, producing about 2.337 million tonnes of chicken meat annually. Poultry industry in India is growing at the rate of 8 to 15 per cent annually. The per capita availability of poultry meat is 2.15 kg as against the recommendation of the National Institute of Nutrition at 11 kg of meat per annum (Prabakaran, 2012). In order to sustain growth and profitability, it is becoming essential to create new innovative ways to stay competitive within the industry and decrease the cost of production as much as possible and at the same time to produce high quality products for consumers.

Feed is a major input cost in poultry accountable for more than 70 per cent of the production cost and it is becoming an issue of greater significance as the price of feed ingredients continue to rise. Protein is among the most expensive nutrient of the feed and broilers have high dietary CP requirements. Proteins and amino acids (AA) perform different functions such as biosynthesis of tissues and animal products. In poultry nutrition, the essential AA are off great concern and of which, tryptophan even though required in lesser quantity, still plays a vital role in tissue protein accretion especially when the dietary crude protein is reduced.

L-Tryptophan is a nutritionally essential AA for poultry and other mono gastric animals because it cannot be synthesized in the body. Tryptophan is a critical nutrient for protein synthesis. However, emerging evidence from recent studies shows that tryptophan and its metabolites (serotonin and melatonin) can regulate feed intake, reproduction, immunity, neurological function and anti-stress responses. Thus, adequate intake of this AA from the diet is crucial for growth, development and health of birds. Tryptophan reduces stressful behavior and provides a nutritional means to improve carcass quality high crude protein diets for broilers results in amino acid excess and elevated nitrogen excretion. Nitrogen retention efficiency may be increased if low crude protein broiler diets are supplemented with critical amino acids (CAA) in a pattern that matches maintenance and tissue accretion needs. In addition, lowering crude protein content in broiler diets may reduce feed cost, allow use of alternate feed stuffs and improve tolerance to heat stress. Poultry nutritionists have decreased the use of protein rich feed ingredients by supplementing critical AA such as DL-methionine, L-lysine and L-threonine which are widely used in poultry industry. Although the tryptophan requirement has been precisely established with many dose response studies with graded levels of tryptophan, yet there are many contradictory reports to state the extent of CP reduction when amino acids are balanced up to the level of L-tryptophan limiting and Hence, this study was conducted with the objectives to establish the level of CP reduction that can be achieved with supplementation of L-tryptophan in diets without compromising growth performance and carcass characteristics in commercial broilers.

Material and Methods

All experimental procedures and animal care were approved by Institutional Animal Ethics Committee (IAEC) of the Karnataka Veterinary, Animal and Fisheries Sciences University, Bidar.

Birds, Management and Experimental Design

Four hundred and sixty-two day-old Vencob Broiler Chicks of uniform weight were obtained from a Venkateshwara Hatcheries, wing banded, weighed and randomly assigned to seven dietary treatments according to a completely randomized design. Each treatment had six replications of 11 birds each. The broiler chicks were housed in deep litter system with paddy husk as litter material. Standard management practices were adopted during the experimental period with *adlibitum* access to feed and water for a period of forty-two days. Chicks were vaccinated as per the standard vaccination programme schedule commonly practised in India.

Chemical Analysis of the Feed Ingredients and Feed Samples

Prior to formulating the diets, all the basal ingredients and the feed samples used in the experimental diets were analysed for crude protein, ether extract, crude fibre and ash content as per the standard procedures. The AA composition of the ingredients used in formulation of the experimental diets were analysed at Evonik SEA Pte. Ltd., Singapore and the AA composition of the experimental diets was calculated based on the analysed AA composition of ingredients. A commercial feed grade L-tryptophan procured from standard supplier was prior tested for purity in reference laboratory and its contribution for CP and energy value were taken into consideration while formulating the test diets.

Experimental Diets

The experimental period was divided into three phases *viz.,* pre-starter (0-14 days), starter (15-28 days) and finisher (29-42 days) and the respective diet was offered in mash form. The control (T_1) was formulated to meet the minimum requirement. The dietary CP was reduced by 0.75 per cent unit in treatment groups (T_2 & T_3), 1.5 per cent unit in treatment groups (T_4 & T_5) and 2.25 per cent unit in treatment groups (T_6 & T_7). For treatment groups T_3, T_5 and T_7, L-tryptophan was supplemented to match the levels present in the control group. The tryptophan to lysine ratio was maintained in all the test diets to meet the minimum requirement except for negative control groups (T_2, T_4 & T_6). The ingredient, nutrient and amino acid composition under pre-starter, starter and finisher diets is presented in Tables 1, 2, 3, 4, 5 and 6, respectively.

TABLE 1

Per cent Ingredient composition of pre-starter diets (0-14 days of experiment)

Ingredients	Treatments						
	T_1	T_2	T_3	T_4	T_5	T_6	T_7
Maize (yellow)	54.62	56.01	56.01	57.40	57.40	58.83	58.85
Soybean meal	34.98	32.37	32.34	29.71	29.65	27.02	26.93
Meat cum bone meal	5.00	5.00	5.00	5.00	5.00	5.00	5.00
Rice Polish	0.0	1.50	1.50	3.00	3.00	4.50	4.50
Vegetable oil	2.80	2.40	2.40	2.00	2.00	1.60	1.60
[1]DCP	0.75	0.75	0.75	0.75	0.75	0.75	0.75
[2]LSP	0.35	0.35	0.35	0.35	0.35	0.35	0.35
Salt	0.40	0.40	0.40	0.40	0.40	0.40	0.40
Sodium Bicarbonate	0.05	0.05	0.05	0.05	0.05	0.05	0.05
*TM Mix	0.100	0.100	0.100	0.100	0.100	0.100	0.100
Potassium carbonate	-	0.05	0.05	0.12	0.12	0.18	0.185
Lysine	0.090	0.170	0.170	0.251	0.252	0.333	0.335
DL-Methionine	0.292	0.314	0.314	0.336	0.337	0.359	0.360
L-Threonine	0.028	0.064	0.064	0.099	0.100	0.136	0.138
Valine	0.00	0.00	0.00	0.016	0.017	0.058	0.060
Isoleucine	0.00	0.00	0.00	0.015	0.016	0.059	0.061
Tryptophan	0.00	0.00	0.013	0.00	0.026	0.00	0.039
**Additives	0.60	0.60	0.60	0.60	0.60	0.60	0.60
Total	100.00	100.00	100.00	100.00	100.00	100.00	100.00

*Trace mineral mixture: Fe - 90000 ppm, I - 2000 ppm, Cu - 15000 ppm, Mn - 90000ppm, Zn - 80000 ppm, Se - 300 ppm.

[1] DCP : Dicalcium phosphate ; [2] LSP : Limestone powder

**Additives : Vit A-10mIU, D_3-2.0 mIU, E-30.0g, C-50 g, B_1-2.0g, B_2-10.0g, B_6-3.0g, B_{12}-0.015, Niacin-30.0g, Calcium-D-Pantothenate 15.0g, Biotin - 0.10g, Folic Acid - 2.0g and Vit - K-4.0g; Herbal Liver stimulant - 1700g; Semduramicin - 30.0g; Tetracyclin-30.00g; a commercial Toxin binder-200

Growth Performance

Cumulative body weights of individual birds were recorded at the end of each phase. Average phase-wise feed consumption for each replicate group was calculated by subtracting the residual feed from the total feed provided at the end of each phase and the overall cumulative feed consumption for experimental period of 42 days was calculated from the data of three phases. Feed efficiency was calculated at the end of each phase and at the end of the experimental period. Mortality was monitored twice daily throughout the experimental period. Birds that died were weighed and the weight was used to adjust the feed conversion (FCR = total feed consumed ÷ (weight of the dead birds + weight gain of the live birds).

Carcass Characteristics

At the end of the experiment (6 weeks), two birds per replicate were slaughtered by severing jugular vein and carotid artery on one side of the neck, allowed to bleed for 1-2 minutes and scalding at 54°C for 2 minutes in dunking scalder and de-feathered mechanically for 30-60 seconds in a rotary drum picker. The birds were dressed by cutting the head at atlanto-occipital joint, leg at hock joint and the carcass was eviscerated by making a slit opening at the abdominal area. The dressing percentage was calculated to determine the carcass characteristics including internal organs *viz*., heart without pericardium, liver without gall bladder and gizzard without inner mucous membrane of the bird (giblets weight).

TABLE 2

Calculated nutrient and amino acid composition (%) of the pre-starter experimental diets

Nutrient	Treatments						
	T_1	T_2	T_3	T_4	T_5	T_6	T_7
Crude Protein	23.000	22.252	22.251	21.500	21.501	20.752	20.751
ME Kcal/Kg	2976	2977	2977	2978	2979	2981	2982
Calcium	0.992	0.986	0.986	0.980	0.980	0.973	0.973
Total Phosphorus	0.747	0.749	0.749	0.752	0.752	0.754	0.754
Available Phosphorus	0.452	0.451	0.451	0.450	0.450	0.449	0.449
Fat	2.849	3.072	3.072	3.295	3.295	3.518	3.518
Linoleic Acid	1.371	1.447	1.447	1.523	1.523	1.599	1.600
CrudeFibre	3.270	3.389	3.389	3.508	3.507	3.625	3.623
Amino acid composition							
Lysine	1.265	1.265	1.265	1.265	1.265	1.265	1.265
Methionine	0.576	0.586	0.586	0.596	0.597	0.607	0.607
Methionine +Cysteine	0.914	0.914	0.914	0.914	0.914	0.914	0.914
Threonine	0.851	0.852	0.851	0.851	0.851	0.851	0.851
Valine	1.040	1.002	1.001	0.977	0.977	0.977	0.977
Isoleucine	0.917	0.875	0.875	0.847	0.847	0.847	0.847
Tryptophan	0.244	0.232	0.244	0.220	0.244	0.208	0.244
Arginine	1.550	1.483	1.482	1.414	1.413	1.345	1.342
Histidine	0.587	0.564	0.564	0.542	0.541	0.519	0.518
Leucine	1.867	1.807	1.806	1.745	1.744	1.682	1.680
Phenyl Alanine	1.094	1.049	1.048	1.002	1.001	0.955	0.953
Tyrosine	0.677	0.650	0.649	0.622	0.621	0.593	0.592
Serine	1.017	0.972	0.972	0.927	0.926	0.880	0.879

Dressing percentage (Carcass yield), abdominal fat (AF) percentage, breast muscle, thigh muscle and drumstick yield were recorded on absolute weight and were also expressed as per cent of carcass weight. While randomly choosing the birds for carcass characteristics, the birds were selected closer to the replicate average weight and those without the visual signs of abnormalities were selected. The data pertaining to the BW, FI, FCR, carcass yield, breast muscle, thigh muscle, drumstick yields, giblet weight and abdominal fat per cent, were subjected to statistical analysis by one-way analysis of variance (ANOVA) using Graph Pad Prism (version 5.01) statistical software. The significant mean differences between the treatment groups were determined at a level of $p<0.05$ using Tukey's Multiple Comparison Test.

RESULTS AND DISCUSSION

1. Growth Performance

a) Body Weight Gain

The body weight gain (BWG), feed intake (FI) and feed conversion ratio (FCR) of the broilers fed with reduced CP diets supplemented with L-tryptophan are presented in Table 7. During pre-starter, starter and finisher phases, there was no significant ($p >0.05$) reduction in the BWG up to 1.5 per cent unit reduced Crude protein (CP) group (T_5) with and without the supplementation of L-tryptophan as compared to that of the higher CP diets (19-23 per cent) control group

TABLE 3

Per cent ingredient composition of starter diets (14-28 days of experiment)

Nutrient	Treatments						
	T_1	T_2	T_3	T_4	T_5	T_6	T_7
Maize (yellow)	60.15	61.58	61.58	63.14	63.12	64.50	64.55
Soybean meal	29.36	26.72	26.70	24.00	23.95	21.30	21.22
MBM	5.00	5.00	5.00	5.00	5.00	5.00	5.00
Rice Polish	0.00	1.50	1.50	3.00	3.00	4.50	4.50
Vegetable Oil	3.00	2.60	2.60	2.10	2.12	1.70	1.70
DCP	0.50	0.50	0.50	0.50	0.515	0.52	0.52
LSP	0.40	0.40	0.40	0.40	0.40	0.42	0.42
Common Salt	0.40	0.40	0.40	0.40	0.40	0.40	0.40
Sodium Bicarbonate	0.05	0.05	0.05	0.05	0.05	0.05	0.05
TM Mix	0.10	0.10	0.10	0.10	0.10	0.10	0.10
Potassium carbonate	-	0.05	0.05	0.12	0.12	0.18	0.185
Lysine	0.170	0.198	0.198	0.281	0.283	0.363	0.366
DL-Methionine	0.283	0.305	0.306	0.328	0.329	0.351	0.352
Threonine (98%)	0.058	0.094	0.095	0.131	0.132	0.168	0.170
Valine	0.00	0.017	0.018	0.060	0.061	0.102	0.104
Isoleucine	0.00	0.017	0.016	0.061	0.063	0.108	0.108
Tryptophan	0.00	0.00	0.130	0.00	0.260	0.00	0.400
Total	100.00	100.00	100.00	100.00	100.00	100.00	100.00

*Trace mineral mixture: Fe-90000ppm, I–2000ppm, Cu–15000ppm, Mn–90000ppm, Zn-80000ppm, Se–300ppm

(T_1). This may be due to the fact that 1.5 per cent reduced CP diet provide adequate minimum levels of amino acids needed to support tissue protein accretion and growth and avoiding the excess of EAA and NEAA which is noticed in high CP (19-23 per cent) diets. The excess of AA is catabolized and excreted as uric acid which costs metabolic loss of 2-14 mol of ATP, otherwise utilized for growth and production. The energy saved in low CP diets supplemented with EAA may be better utilized for growth and better amino acid balance as well as ratios between essential and non-essential amino acids for maximizing the protein tissue accretion as opposed to their less efficient use as an energy source could have been the reason for improved performance similar to that of higher CP (19-23 per cent) control group. At 1.5 per cent unit reduced CP with L-tryptophan supplementation may be matching the bird's limiting AA requirements up to the level of tryptophan for maintenance and tissue protein accretion in order to obtain optimum performance similar to that of the control group having higher CP. Low CP diets with balanced EAA improve N efficiency utilization, reduce N excretion and reduce the level of ammonia in the litter (Basavantha *et.al.,* 2015). Increased retention of nutrients and N retention might be responsible for similar performance in low crude protein diets as compared to those fed higher CP diets (Ramarao *et al*., 2011). The present findings were in corroboration with the studies conducted by Ramarao *et al*., 2011 and Abudabos & Ajumaah, 2012.

The results indicated that L-tryptophan can be supplemented by reducing the dietary crude protein by 2.25 per cent unit during pre-starter and starter phases of feeding. The results obtained are in

TABLE 4

Calculated nutrient and amino acid composition (%) of the starter experimental diets

Nutrient	Treatments						
	T_1	T_2	T_3	T_4	T_5	T_6	T_7
Crude Protein	21.002	20.251	20.254	19.501	19.500	18.751	18.750
ME Kcal/Kg	3050	3052	3052	3049	3051	3050	3052
Calcium	0.788	0.781	0.781	0.775	0.778	0.780	0.779
Total Phosphorus	0.685	0.688	0.688	0.690	0.692	0.696	0.695
Available Phosphorus	0.404	0.403	0.403	0.402	0.404	0.404	0.404
Sodium	0.218	0.218	0.218	0.218	0.218	0.218	0.218
Chloride	0.287	0.287	0.287	0.288	0.288	0.289	0.289
Crude Fat	3.076	3.299	3.299	3.526	3.525	3.748	3.748
Linoleic Acid	1.460	1.536	1.536	1.614	1.614	1.690	1.690
Crude Fibre	3.224	3.342	3.342	3.462	3.460	3.578	3.576
Amino acid composition							
Lysine	1.150	1.150	1.150	1.150	1.150	1.150	1.150
Methionine	0.548	0.558	0.559	0.569	0.569	0.579	0.580
Met + Cys	0.863	0.863	0.863	0.863	0.863	0.863	0.863
Threonine	0.806	0.806	0.806	0.806	0.806	0.806	0.806
Valine	0.946	0.923	0.923	0.923	0.923	0.923	0.923
Isoleucine	0.821	0.778	0.778	0.735	0.734	0.691	0.690
Tryptophan	0.217	0.205	0.217	0.192	0.217	0.180	0.217
Arginine	1.385	1.317	1.316	1.247	1.245	1.177	1.174
Histidine	0.534	0.511	0.511	0.488	0.488	0.465	0.464
Leucine	1.736	1.674	1.674	1.612	1.610	1.549	1.546
P.Alanine	0.989	0.943	0.943	0.896	0.894	0.848	0.846

accordance with Corzo *et al*., 2005a indicating tryptophan which is required in lesser proportion still plays a role in limiting protein synthesis and the needs are high for tissue protein accretion leading to optimum performance.

However, during finisher phase, L-tryptophan addition ($T_{3,}$ T_5 & T_7) did not show any significant reduction in body weight gain as compared to control group (T_1) and corresponding negative control groups at 0.75, 1.5 and 2.25 protein reduction groups (T_2, T_4 and $T_{6)}$. In the finisher phase, there was no significant difference observed among all the treatment groups, due to relatively higher feed intake to compensate for the nutrient deficiency of ration for requirement and supply of amino acids and resulted in higher performance on par with the control group but for the poor feed efficiency. The above findings in finisher phase were in accordance with Liu Suo Zhu, 2007.

On cumulative basis, CP reduction of 1.5 per cent unit and above resulted in significantly (P<0.05) poor BWG. It was interesting to note that, the L-tryptophan addition significantly (P<0.05) improved the BWG *vis-à-vis* respective negative controls without L-tryptophan supplementation.

The reduction of the BWG in T_4 and T_6 could be due to reduction of feed intake significantly (p<0.05) reduced feed intake and feed efficiency leading to

TABLE 5

Per cent ingredient composition of finisher diets (29-42 days of experiment)

Ingredients	Treatments						
	T_1	T_2	T_3	T_4	T_5	T_6	T_7
Maize (yellow)	63.66	65.05	65.05	66.45	66.47	67.87	67.90
Soyabean meal	24.55	21.93	21.91	19.26	19.21	16.56	16.48
Meat cum bone meal	5.00	5.00	5.00	5.00	5.00	5.00	5.00
Rice Polish	0.00	1.5	1.50	3.00	3.00	4.50	4.50
Vegetable Oil	4.2	3.80	3.80	3.40	3.40	3.00	3.00
Dicalcium phosphate	0.70	0.70	0.70	0.70	0.70	0.70	0.70
Limestone powder	0.50	0.50	0.50	0.50	0.50	0.50	0.50
Salt	0.40	0.40	0.40	0.40	0.40	0.40	0.40
SodiumBicarbonatee	0.05	0.05	0.05	0.05	0.05	0.05	0.05
TM Mixture	0.100	0.100	0.100	0.100	0.100	0.100	0.100
Potassium carbonate	-	0.05	0.05	0.12	0.12	0.18	0.185
Lysine	0.082	0.161	0.162	0.243	0.244	0.325	0.328
Methionine	0.210	0.232	0.232	0.255	0.255	0.278	0.278
Threonine	0.025	0.061	0.061	0.097	0.098	0.1340	0.135
Valine	0.00	0.000	0.000	0.022	0.023	0.066	0.068
Isoleucine	0.000	0.000	0.000	0.045	0.046	0.092	0.094
Tryptophan	0.00	0.000	0.013	0.000	0.026	0.000	0.039
Additives	0.600	0.600	0.600	0.600	0.600	0.600	0.600
Total	100.00	100.00	100.00	100.00	100.00	100.00	100.00

Trace mineral mixture: Fe-90000ppm, I–2000ppm, Cu–15000ppm, Mn–90000ppm, Zn-80000ppm, Se–300ppm

inadequate levels of NEAA in the diet of the birds for which the requirement is not yet known or there could be imbalance of amino acid requirement on digestibility basis leading to altered ratio of EAA to NEAA. Supplementation of L-tryptophan may provide optimum amino acid balance leading to adequate nitrogen pool to provide for the synthesis of non-essential amino acids and balance among the certain essential amino acids which might have resulted in higher performance similar to that of the 0.75 per cent unit reduced crude protein group with supplementation of L-tryptophan (T_3).

The effects of the L-tryptophan deficiency observed in the study are in accordance with the observations of the previous studies (Corzo *et al*., 2005b and Hsia *et al*., 2005).

The results indicated that supplementation of L-tryptophan in optimizing the growth performance of broilers fed low protein diets as compared to the birds fed higher CP had the beneficial effect but was restricted to reduction of 1.5 to 2.25 per cent crude protein during different feeding phases of the experimental period.

On the contrary, Aftab, 2006; Sterling *et al*., 2005; Waldroup *et al*., 2005 and Jiang *et al*., 2005 reported significantly lower performance of broilers fed low CP diets although the levels of essential AA were maintained according to the requirements.

The variation observed in the experiment upon the extent of the level of protein reduction in optimizing broiler performance may be attributed to the fact that the differences in the digestibility coefficient of the

TABLE 6
Calculated nutrient and amino acid composition (%) of the finisher diets

Nutrient	Treatments						
	T_1	T_2	T_3	T_4	T_5	T_6	T_7
Crude protein	19.001	18.251	18.253	17.500	17.500	16.752	16.750
ME Kcal/Kg	3153	3154	3154	3156	3157	3159	3160
Calcium	0.850	0.844	0.844	0.837	0.837	0.831	0.831
Total Phosphorus	0.699	0.702	0.701	0.704	0.704	0.706	0.706
Available Phosphorus %	0.431	0.431	0.431	0.430	0.430	0.429	0.429
Sodium	0.217	0.217	0.217	0.217	0.217	0.218	0.218
Chloride	0.287	0.287	0.287	0.288	0.288	0.289	0.289
Fat	3.135	3.358	3.358	3.581	3.581	3.804	3.805
Linoleic Acid	1.508	1.584	1.584	1.660	1.661	1.737	1.737
Crude Fibre	3.142	3.261	3.260	3.379	3.378	3.496	3.494
Amino acid composition							
Lysine	1.000	1.000	1.000	1.000	1.000	1.000	1.000
Methionine	0.462	0.472	0.472	0.483	0.482	0.493	0.493
Methionine+ Cysteine	0.752	0.752	0.752	0.752	0.752	0.752	0.752
Threonine	0.702	0.702	0.702	0.702	0.702	0.702	0.702
Valine	0.859	0.820	0.820	0.802	0.802	0.803	0.802
Isoleucine	0.734	0.692	0.692	0.690	0.690	0.690	0.690
Tryptophan	0.191	0.179	0.191	0.167	0.191	0.154	0.191
Arginine	1.238	1.170	1.170	1.102	1.100	1.032	1.029
Histidine	0.485	0.463	0.463	0.440	0.440	0.417	0.416
Leucine	1.609	1.548	1.548	1.486	1.485	1.423	1.421
Phenyl Alanine	0.893	0.847	0.847	0.801	0.800	0.753	0.752
Tyrosine	0.546	0.518	0.518	0.490	0.490	0.462	0.461

amino acids in the various ingredients chosen for the experiment, deficiency of amino acid when formulated on digestibility basis, birds age, strain, genetic makeup (genotypes) and of the bird with the different growth rates, rate of feathering of the broilers, environmental, management factors and their interactions.

b) Feed Intake

The Feed intake (FI) with L-tryptophan addition was altered at all phases except at finisher phase. During pre-starter and starter phase, the FI significantly (P<0.05) reduced on CP reduction at 2.25 per cent unit as compared to control group (T1). Interestingly with L-tryptophan supplementation, the FI improved (P>0.05) on par with the control at 2.25 per cent unit CP reduction at the end of pre-starter and starter phases. During cumulative basis, FI significantly (P<0.05) reduced at 2.25 per cent CP reduced group without tryptophan supplementation. However, with the tryptophan addition (T_7) it showed improved FI and was on par with the control group (T_1)

The results obtained in the study were in accordance with Hsia *et al*., 2005 who reported no significant differences in feed intake at reduced CP diet with tryptophan levels at 0.198, 0.228 and 0.258 and CP levels of 18.8 per cent during pre-starter and starter phase (0-21 days).

TABLE 7

Effect of supplementation of L-tryptophan in low crude protein diets on the growth performance of broilers

Nutrient	Treatments								p value
	T_1	T_2	T_3	T_4	T_5	T_6	T_7	SEM [2]	
Days 0-14									
BWG [3] (g)	285.97 [a]	276.64 [ab]	291.52 [a]	268.97 [ab]	280.01 [ab]	249.55 [b]	267.89 [ab]	3.349	0.0120
FI [4] (g)	343.30 [ab]	334.94 [ab]	351.44 [a]	329.64 [ab]	341.60 [ab]	307.03 [b]	327.97 [ab]	4.049	0.0823
FCR [5]	1.200	1.216	1.207	1.224	1.218	1.230	1.224	0.0029	.0751
Days 15-28									
BWG (g)	714.34 [a]	687.73 [ab]	718.63 [a]	666.12 [ab]	702.60 [a]	637.27 [b]	684.47 [ab]	6.262	0.0018
FI (g)	1061.79 [a]	1034.97 [ab]	1073.15 [a]	1008.3 [ab]	1053.51 [a]	959.15 [b]	1044.7 [ab]	8.885	0.0045
FCR	1.483 [a]	1.501 [bc]	1.491 [ab]	1.517 [d]	1.510 [cd]	1.529 [e]	1.520 [de]	0.0025	<0.001
Days 29-42									
BWG (g)	1052.33	1031.67	1035.72	985.65	1027.18	991.70	999.29	6.565	0.0324
FI (g)	2102.93	2068.37	2061.53	2063.62	2096.78	2070.68	2044.99	10.85	0.8300
FCR	2.018 [a]	2.030 [ab]	2.026 [ab]	2.049 [cd]	2.041 [bc]	2.065 [e]	2.057 [de]	0.0027	<0.001
Days 0-42									
BWG (g)	2052.65 [a]	1996.05 [abc]	2045.88 [ab]	1920.75 [cd]	2009.80 [abc]	1878.51 [d]	1951.66 [bcd]	12.26	0.032
FI (g)	3508.03 [a]	3438.29 [ab]	3486.12 [ab]	3401.55 [ab]	3497.90 [abc]	3336.86 [b]	3417.67 [ab]	15.22	0.021
FCR	1.715 [a]	1.732 [ab]	1.717 [a]	1.748 [bc]	1.740 [bc]	1.772 [d]	1.750 [c]	0.0032	<0.001

[1] Control diet was formulated to meet minimum NRC Recommendations (1994)
[2] Standard error of the means. Data is the mean of six replicate pens of the birds within each treatment
[3] Body weight gain
[4] Feed intake
[5] Feed conversion ratio
Values in the same row bearing unlike superscripts are different ($p<0.05$)

TABLE 8

Effect of tryptophan on the carcass characteristics and organometry of the birds fed with different treatment diets at the end of the experimental period

Treatment	Carcass		Breast		Thigh		Drumstick		Abdominal fat		Liver	Gizzard	Heart
	Yield (g)	% of CW	Yield (g)	% of CW	Yield (g)	% of CW	Yield (g)	% of CW	Yield (g)	% of CW	% of live body weight		
T_1	1510 ^{a}	72.74	324.7 ^{a}	21.50	203 ^{a}	13.44 ^{a}	152.1 ^{a}	10.07 ^{a}	39.32	1.89 ^{a}	2.019	1.975	0.4561
T_2	1442 ^{ab}	72.43	309.1 ^{ab}	21.43	192 ^{ab}	13.31 ^{ab}	142.7 ^{ab}	9.89 ^{a}	38.95	1.96 ^{ab}	2.010	1.993	0.4651
T_3	1519 ^{a}	72.68	327.6 ^{a}	21.56	203 ^{a}	13.36 ^{a}	152.1 ^{a}	10.01 ^{a}	39.77	1.90 ^{a}	1.969	1.976	0.4591
T_4	1406 ^{ab}	71.95	295.6 ^{ab}	21.02	181 ^{ab}	12.87 ^{abc}	131.4 ^{ab}	9.34 ^{ab}	42.15	2.16 ^{b}	2.017	2.015	0.4711
T_5	1473 ^{ab}	72.35	312.3 ^{ab}	21.20	193 ^{ab}	13.10 ^{abc}	140.2 ^{ab}	9.51 ^{ab}	41.91	2.06 ^{ab}	2.004	1.983	0.4671
T_6	1383 ^{b}	71.68	283.9 ^{b}	20.52	171 ^{b}	12.36 ^{c}	122.8 ^{b}	8.87 ^{b}	41.96	2.18 ^{b}	2.019	2.026	0.4731
T_7	1429 ^{ab}	72.14	295.2 ^{ab}	20.65	179 ^{ab}	12.52 ^{bc}	131.5 ^{ab}	9.20 ^{b}	42.95	2.17 ^{b}	2.010	2.019	0.424
SEM	11.90	0.119	3.571	0.171	2.547	0.103	2.249	0.109	0.392	0.015	0.008	0.008	0.008
p value	0.009	0.172	0.004	0.129	0.002	0.033	0.001	0.003	0.256	0.011	0.678	0.433	0.996

Values in the same column with unlike superscripts are different ($p<0.05$)

However, no significant (p>0.05) difference were observed in the feed intake during the finisher phase among all the treatment groups. Similar findings in the finisher phase were observed by Hsia *et al*., 2005. They also attributed to the fact that 0.154 per cent tryptophan present in the finisher diet in the 2.25 per cent unit reduced CP group may be enough for the feed intake but not enough for the amino acid balance.

On cumulative basis, there was significant reduction in feed consumption in 2.25 per cent unit reduced CP group without L-tryptophan supplementation (T_7) as compared to that of the control group (T_1). The results indicated that feed intake has significantly affected by the lower level of tryptophan at 0.208 levels at pre-starter phase, 0.180 levels at starter phase and 0.154 levels at finisher phase cumulatively affect the feed intake at the end of the experiment (0-42 days). The research findings are in accordance with Corzo *et al*., 2005c. Supplementation of L-tryptophan showed an improvement in the feed intake similar to that of the control group (T_1). The results of the present experiment indicates that bird's feed intake is sensitive to the moderate deficiency of tryptophan in low crude protein diets and upon addition of L-tryptophan improved the feed intake which perhaps indicates that tryptophan has important role in regulating feed intake since tryptophan is precursor of neuro transmitter, serotonin which is formed in the brain and influences the feed intake of the birds. It has been virtually observed that the birds consuming low levels of tryptophan exhibited abnormal behaviour characterized by feed spillage (Corzo *et al*., 2005d). This observation is partially explained by the fact that the birds were searching for a missing nutrient (Tryptophan).

On the contrary, 1.5 per cent unit reduced CP group without L-tryptophan supplementation having the tryptophan content of 0.22, 0.192 and 0.167 per cent in pre-starter, starter and finisher phase did not significantly (p>0.05) affect the feed intake. Probably, the mild deficiency of L-tryptophan did not affect the feed intake significantly (p>0.05) at the end of the experiment on cumulative basis. This indicates that the tryptophan requirement for the feed intake may be sufficient to control feed intake but not for the body weight gain and feed to gain ratio as it significantly reduced as compared to the control group (T_1). However, tryptophan supplementation in same level of CP group showed non-significant increase improvement in the feed intake.

c) Feed Conversion Ratio (FCR)

FCR did not differ among all the treatment groups as compared to control. The findings are in accordance with Campos *et al*., 2010 who reported no significant difference in feed conversion ratio calculated on digestible tryptophan to lysine ratio.

However, during starter phase, the better FCR observed in control (T_1) significantly (P<0.05) surpassed treatments T_4 and above. The CP reduction significantly (P<0.05) depressed FCR at and beyond 0.75 per cent unit, which was improved on par to control with tryptophan supplementation only at lower level of CP reduction (T_3) but not with further CP reduction.

During starter phase, there was significantly lower feed efficiency at 0.75 per cent reduced CP group without L-tryptophan supplementation group (T_2) due to relatively higher feed intake as compared to that of control group (T_1). Supplementation of L-tryptophan improved the feed to gain ratio up to 0.75 per cent reduced CP group (T_3) similar to that of the control group (T_1) indicating that the birds are sensitive to mild deficiency of tryptophan at starter phase. However, requirement of tryptophan may be higher for feed efficiency than that of requirement for feed intake and body weight gain at starter phase as the lower feed efficiency in starter phase up to 2.25 per cent unit reduced CP groups showed wider feed conversion ratios compared to the control group (T_1).

At finisher phase and on cumulative basis, depressed FCR was seen at 1.5 and 2.25 per cent unit's protein reduction as compared to that of the control group. However, tryptophan supplementation did not improve as that of the control group.

At finisher phase, there was no significant (p>0.05) difference in the FCR up to 0.75 per cent unit reduced

CP group with or without L-tryptophan supplementation because the birds could meet their requirement through increased feed intake as compensatory mechanism to meet the amino acid requirement and sustained growth has occurred which lead to non-significant improvement in the feed efficiency indicating mild deficiency of tryptophan resulted in the higher intake as a compensatory mechanism to meet the nutrient requirements.

Further, CP reduction by 1.5 per cent unit with and without L-tryptophan supplementation group (T_4 & T_5) showed significantly ($p<0.05$) poorer feed efficiency as compared to optimum CP control group (T_1). Tryptophan supplementation did not improve the feed efficiency as that of the control group (T_1). This lower FCR might be due to the result of the birds fed low CP diets consumed relatively more feed compared to that of higher CP control group and grew slowly which in turn may be due to deficiency of tryptophan and other CAA and so the efficiency of feed utilization has decreased.

Groups with CP reduction by 2.25 per cent unit with and without L-tryptophan supplementation groups (T_6 & T_7) showed significantly ($p<0.05$) lower feed efficiency as compared to optimum CP control group (T_1) during starter and finisher phase. Upon supplementation of L-tryptophan (T_7) at 2.25 per cent unit CP reduction level, feed conversion ratio improved to that of 1.5 per cent reduced CP with or without L-tryptophan supplementation (T_4 & T_5) during starter phase but could not improve as that of the control (T_1) or 0.75 per cent unit reduced CP (T_2 & T_3) during starter phase. During finisher phase, supplementation of L-tryptophan (T_7) showed improvement in the feed conversion ratio to that of 1.5 per cent unit reduced CP without L-tryptophan supplementation (T_4) but could not improve as that of the control (T_1). This further supports the hypothesis, that the requirement of tryptophan is higher for feed conversion (Rosa *et al.*, 2001) as compared to that of feed intake as well as body weight gain.

2. Carcass Characteristics

In the carcass traits (Table 8), the carcass fresh weight significantly ($P<0.05$) decreased in T_6 in comparison with control, while the others remained non-significant with T_1 as well as T_7. However, the dressing per cent and relative weights of giblet organs were not influenced by the treatments. The breast yield (weight) was significantly decreased with CP reduction, at 2.25 per cent unit. Addition of L-tryptophan improved breast weight at CP reduction of 2.25 per cent unit. But the per cent carcass weight did not show any significant difference. A similar pattern of variation was observed for thigh and drum stick weights as well as their percent to carcass weights. The absolute abdominal fat (AF) weight did not show any significant difference among all the treatment groups. However, AF expressed as a per cent of body weight on relative basis, a different trend of variation was observed. AF as a percent of body weight showed significant increase at 1.5 and 2.25 percent unit reduced protein groups (T_4 & T_6) as compared to control group (T_1). Tryptophan addition showed improvement at 1.5 per cent protein reduced group but did not show any improvement at 2.25 percent unit protein reduced group as compared to control group. Further CP reduction at 2.25 per cent unit significantly ($P<0.05$) increased AF production. However, AF was not alleviated by supplementation of L-tryptophan. The relative weights of giblet organs, liver, heart and gizzard were not affected by dietary treatments.

The results of our study indicate that the carcass yield are influenced by the tryptophan content in the diet which is at the lowest level in T_6 group having 0.208, 0.180 and 0.154 per cent, respectively which may be attributed to imbalance of amino acids in the treatment diet. Also diets containing low dietary protein may limit muscle protein synthesis by reduction of myogenic gene transcription and myofibril size (Duclos, 2005 and Tesseraud *et al.*, 2006).

The results obtained are in accordance with the findings of Hsia *et al.*, 2005. On the contrary, Abdallah *et al.* (2009) concluded that supplementation of L-tryptophan in low crude protein diets had no significant ($p<0.05$)

effect on carcass weight. The CP reduction of 2.25 per cent unit without L-tryptophan supplementation (T_6) significantly ($p<0.05$) reduced cumulative BWG which upon tryptophan supplementation significantly ($p<0.05$) improved to equate to that of 0.75 per cent reduced CP group with or without L-tryptophan supplementation groups (T_2 & T_3) but could not improve as that of the control group (T_1). This explains the magnitude of effect of tryptophan supplementation on tissue protein accretion, which is reflected as total body mass yield on absolute weight basis including increased yields in breast muscle, thigh muscle and drumstick which contributed to higher carcass yield. However, carcass yield on relative basis, expressed as a per cent of BW did not differ statistically ($p<0.05$) among all treatment groups.

The CP reduction of 2.25 per cent unit without L-tryptophan supplementation group (T_6) significantly reduced cumulative BWG which upon tryptophan supplementation significantly ($p>0.05$) improved to equate to that of 0.75 per cent unit reduced CP with or without L-tryptophan supplementation group (T_2 & T_3). This explains the magnitude of effect of tryptophan supplementation on tissue protein accretion, which is reflected in the Breast mscle yield (BMY), Thigh muscle yield (TMY) and Drumstick yield (DY) in the present study. The research findings are in accordance with Hsia *et al*., (2005). Contrary findings have been reported by Abdallah *et al*. (2009) which states that supplementation of L-tryptophan in low crude protein diets had no significant ($p>0.05$) effect on breast muscle yield. This reduction of BMY, TMY and DY at 2.25 per cent reduced CP may be attributed to the deficiency of tryptophan and further imbalance of amino acids at the lower CP diet groups. However, supplementation of L-tryptophan improved the BMY because of the better amino acid balancing and probably the requirement of tryptophan may be higher than 0.208, 0.180 and 0.154 per cent during pre-starter, starter and finisher phases, respectively for optimum BMY at the end of the experimental period.

The AF was increased in low CP diets which indeed reflect the altered energy and protein ratio and more specifically energy and AA ratios. Since a constant energy was maintained in all test diets and subsequently relatively more amount of feed was consumed per live kg weight in low CP diets than control group birds which resulted in excess energy intake per kg live weight gain and excess energy which in turn got deposited as abdominal fat. Similar effects have been observed in other studies. Thus, the increased fat deposition in birds receiving low CP diets reported herein may be a result of lower energy expenditure and less amino acid nitrogen catabolism into uric acid as reported by Basavantha *et.al.,* 2015.

Dietary CP could be reduced by 1.5 per cent units with supplementation of L-tryptophan without affecting BWG at the end of 42 days in commercial broilers. FI do not get affected in the commercial broilers up to 2.25 per cent unit reduction in the dietary CP levels with supplementation of L-tryptophan in the broiler diets. FCR is not affected in the commercial broilers up to 0.75 per cent unit reduction in the CP levels with supplementation of L-tryptophan. L-tryptophan supplementation significantly ($p<0.05$) increased carcass yield, BMY, TMY and DY in CP reduction of 2.25 per cent unit. L-tryptophan supplementation significantly ($p<0.05$) reduced AF at CP reduction of 1.5 per cent but the not at the CP reduction of 2.25 per cent unit. Giblet weights *viz.,* heart without pericardium, liver without gall bladder and gizzard without inner mucous membrane expressed as per cent of body weight was unaffected by various dietary CP levels with or without L-tryptophan supplementation.

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