Int.J.Curr.Microbiol.App.Sci (2017) *6*(12): 4224-4231

International Journal of Current Microbiology and Applied Sciences
ISSN: 2319-7706 **Volume 6 Number 12 (2017) pp. 4224-4231**
Journal homepage: http://www.ijcmas.com

Original Research Article https://doi.org/10.20546/ijcmas.2017.612.486

Influence of L-threonine Supplementation to Diets with Reduced Crude Protein on Feed Efficiency in Commercial Broilers

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Keywords

Broilers, Crude protein, Threonine, Feed consumption and feed conversion ratio.

Article Info

Accepted:
28 October 2017
Available Online:
10 December 2017

A B S T R A C T

The present study aimed to investigate the effect of reduced dietary crude protein (CP) and supplementation of threonine on feed efficiency in commercial broilers (CB). A total of 480 broiler chicks randomly assigned to 4 dietary treatments following completely randomized design, each treatment had six replications of 20 birds each. The control dietary formulation was prepared to meet requirement of nutrients and amino acids as per the NRC (1994). The dietary CP was reduced by 1, 2 and 3 percent unit to the standard recommendation in other 3 treatments, the treatment with reduced CP level were supplied with synthetic threonine to need desired level of threonine as in control and all amino acids were met as per the NRC (1994) by supplying the limiting amino acids. From overall study it was indicated that the weekly cumulative feed consumption was similar in 1 percent unit CP reduced diet with L-threonine supplementation compare to control diet but it was significantly decreased in 2 and 3 percent unit reduced levels of CP, whereas the weekly cumulative feed conversion ratio (FCR) significantly higher in 2 and 3 percent unit CP reduced diets, but FCR was significantly lower and better in 1 percent unit CP reduced diet with L-threonine supplementation and it was simillar to that of control diet.

Introduction

The main hurdle to future growth of poultry is the availability of feed, especially maize and soya at reasonable prices. The next requirement for the Indian poultry industry is increased productivity of feed. Maize and soybean meal are the key feed ingredients. The costs of these components are very crucial in sustainable operations of a poultry farm. Maize is the primary source of energy for the Indian poultry industry and constitutes 60% of the compound feed, while soybean is the primary source of protein and forms 30-35% of the feed.

Feed is a significant input cost in poultry accountable for more than 70% of the recurring expenditure. Protein is the most expensive nutrient of the feed and broilers have high dietary crude protein (CP) requirements. Proteins and amino acids perform different functions such as biosynthesis of tissues and animal products. All essential amino acids are equally necessary, and none of them are unimportant, those being relatively more crucial than others, are known as critical amino acids. Threonine is the third limiting amino acid after methionine and lysine in broiler chicken diets as the essential amino acids are of great importance in poultry nutrition (Han *et al.,* 1992).

High crude protein diets for broilers results in amino acids excess and elevated nitrogen excretion. Low crude protein broiler diet supplementation with crystalline amino acids may increase in a way that matches maintenance and tissue build up needs. Also, lowering crude protein content in broiler diets may reduce feed cost, allow the use of alternate feedstuffs and improve tolerance to heat stress (Kidd and Kerr, 1997). Poultry nutritionists have decreased the use of protein-rich feed ingredients by supplementing critical amino acids such as DL-methionine and L- lysine which is accepted widely for use in the poultry industry. The extent of decrease in crude protein without compromising on the bird performance remains subject to much debate (Kidd *et al.,* 1996).

In corn – soya based feedstuff for poultry, threonine is the third limiting amino acid in for broilers, and it becomes more limiting as crude protein decreases (Kidd, 2000). L-Threonine supplemented in low crude protein diets shown to support the same production that achieved in broilers fed high protein diets (Kidd *et al.,* 1997). Adequate threonine levels are needed to help optimum growth because it serves as an essential component of body protein and plays a vital role as the precursor of L-lysine and serine and additionally for excellent immune response and gastrointestinal mucin production (Kidd *et al.,* 1999).

Although the threonine requirement is established precisely with many dose-response studies with graded levels of threonine, yet there are many contradictory reports to state the extent of crude protein reduction with L-threonine supplementation. Considering the above the present study was taken up to evaluate the supplementation of L- threonine in the broiler diets by lowering crude protein on feed efficiency in commercial broilers.

Materials and Methods

A day-old four hundred and eighty straight run commercial Vencobb-400 broiler chicks of uniform body weight procured from the commercial hatchery were wing banded, weighed and randomly assigned to four dietary treatments following completely randomized design. Each treatment had six replications of 20 birds each. The broiler chicks were housed in deep litter system. Standard management practices were adopted during the experimental period. Threonine Chicks were vaccinated as per the standard vaccination schedule for commercial broilers practiced in India.

The experimental commercial broiler diet was divided into three phases *i.e.,* pre- starter, starter and finisher phases. The control (T_1) dietary formulation was prepared to meet the minimum requirement of nutrients and amino acids as per the NRC (1994). The dietary crude protein was reduced by one per cent unit in $T_{2,}$ two percent unit in T_3 and three per cent unit in T_4 to the standard recommendations (Table 1). The treatments with reduced protein level were supplied with synthetic threonine to meet desired level of threonine as in control group. In all lowered dietary protein treatments, the minimum requirement of all amino acids were met as per the NRC (1994) by supplying the limiting amino acids. Threonine to lysine ratios was maintained in all the treatments to meet the minimum requirement as per Baker (1994). The ingredient, nutrient and amino acid compositions of the diet are presented in Table 2, 3 and 4 respectively. Body weight, feed consumption and feed conversion ratio were recorded at the end of each week. The data collected in the study was stored in MS excel and analyzed using Statistical Package for Social Science (SPSS). The data pertaining to body weight feed consumption and feed conversion ratio parameters for the trial were subjected to statistical analysis by one way analysis of variance (ANOVA). The statistical analysis was done at five per cent level of significance ($P \leq 0.05$). Significant mean difference between the treatments was determined by using Duncan's new multiple range tests.

Results and Discussion

The effect of supplementation of L-threonine by lowering dietary crude protein (CP) levels on the mean body weight, feed consumption and feed conversion ratio (FCR) of commercial broilers from day one to sixth week of age on weekly cumulative basis were significantly influenced among the various treatment groups presented in Table 5, 6 and 7 respectively and shown in Figure 1.

Reduction of CP in the diet by 2 and 3 percent unit (T_3 and T_4) showed a significant reduction in the feed intake as compared to that of the control diet (T_1). On cumulative basis, there was no significant variation in feed consumption in 1 per cent unit reduced CP group with L-threonine supplementation (T_2) as compared to that of the control group (Table 6). The results were in close agreement with the findings of Han *et al.,* (1992) who found no differences in feed intake of broilers when the CP content of the diet was decreased. Bartov and Plavnik (1998) also reported no difference in feed intake of broilers fed diets with different CP levels. Similar results were also obtained by Kidd *et al.,* (1996) and Ramarao *et al.,* (2011). This is probably due to requirements in terms of CP in accordance with the principle of achieving the correct balance of dietary amino acids but up to 1 percent unit reduction of CP did not affect the feed intake and performance. Present study results are on par with Bade *et al.,* (2014) was observed that reduction of protein content in the diet by 1 percent unit than recommended level coupled with supplementation of threonine did not affect the feed consumption of birds, rather resulted in achieving marginally better body weights for similar feed consumption.

The results of the present study were also in accordance of Zaefarian *et al.,* (2008), Ahmed (2014) and Anand (2015) they concluded that reduction of CP more than 1.5% unit in the broiler diets will decrease the feed intake as compared to that of the high protein control diet. However, on the contrary Ardekani and Chamani (2012) who reported that supplementation of L- threonine in 16% CP diets resulted in highest feed consumption with better growth performance as compared to 20% crude protein diets.

Table.1 Experimental diets formulated for different growth phases of commercial broilers

Experimental diets	Pre-Starter (1 – 14 days)		Starter (15 – 28 days)		Finisher (29 – 42 days)	
	CP %	Threonine	CP %	Threonine	CP %	Threonine
T_1	23.00	Basal level	21.00	Basal level	19.00	Basal level
T_2	22.00	0.046%	20.00	0.051%	18.00	0.040%
T_3	21.00	0.094%	19.00	0.095%	17.00	0.095%
T_4	20.00	0.145%	18.00	0.140%	16.00	0.140%

CP- Crude Protein

L-threonine was supplemented to meet basal threonine level of 0.86% in pre-starter, 0.76% in starter and 0.70% in finisher rations.

Table.2 Ingredient composition of pre-starter, starter and finisher diets

Ingredient (%)	Pre-starter diets (1-14 days)				Starter diets (15-28 days)				Finisher diets (29-42 days)			
	T_1	T_2	T_3	T_4	T_1	T_2	T_3	T_4	T_1	T_2	T_3	T_4
Maize	49.50	50.85	53.00	55.05	52.42	54.29	57.40	59.50	57.02	59.80	61.95	63.77
Soybean meal	41.40	38.00	34.45	31.07	36.50	33.05	29.80	26.40	31.20	27.85	24.37	20.90
Rice polish	0.00	2.20	4.01	5.46	00.00	1.80	2.15	3.67	0.00	1.00	2.70	4.60
Di-calcium phosphate	1.95	1.95	1.95	1.95	2.20	2.20	2.20	2.20	2.20	2.20	2.20	2.20
Limestone powder	1.30	1.30	1.30	1.30	1.30	1.30	1.30	1.30	1.30	1.30	1.30	1.30
Salt	0.40	0.40	0.40	0.40	0.40	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	0.05	0.05	0.05	0.05	0.05
***Trace mineral mixture**	0.10	0.10	0.10	0.10	0.10	0.10	0.10	0.10	0.10	0.10	0.10	0.10
Vegetable oil	4.50	4.10	3.50	3.20	6.20	5.80	5.40	5.00	6.90	6.30	5.75	5.30
Lysine (98%)	0.012	0.110	0.215	0.325	0.000	0.110	0.215	0.320	0.015	0.110	0.220	0.330
DL- Methionine (99%)	0.288	0.311	0.332	0.355	0.232	0.260	0.295	0.320	0.210	0.240	0.270	0.300
L-Threonine (98%)	0.00	0.046	0.094	0.145	0.00	0.051	0.095	0.140	0.00	0.040	0.095	0.140
**** Additives**	0.60	0.60	0.60	0.60	0.60	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**	**100.00**	**100.00**	**100.00**	**100.00**	**100.00**

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

**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-2000g.

Table.3 Nutrient compositions calculated for pre-starter, starter and finisher diets

Ingredient (%)	Pre-starter diets				Starter diets				Finisher diets			
	T_1	T_2	T_3	T_4	T_1	T_2	T_3	T_4	T_1	T_2	T_3	T_4
Crude protein (%)	23.02	22.05	21.01	20.00	21.04	20.10	19.02	18.01	19.04	18.03	17.03	16.02
Metabolizable Energy (Kcal/kg)	2999	3001	3007	3003	3104	3108	3106	3104	3204	3201	3205	3202
Calcium (%)	1.02	1.01	1.00	0.99	1.06	1.05	1.04	1.03	1.04	1.04	1.03	1.02
Total phosphorous (%)	0.726	0.731	0.734	0.733	0.747	0.749	0.741	0.741	0.727	0.724	0.726	0.728
Available phosphorous (%)	0.463	0.463	0.461	0.460	0.499	0.498	0.495	0.494	0.493	0.492	0.490	0.489
Fat (%)	6.99	6.90	6.59	6.52	8.74	8.61	8.33	8.17	9.54	9.13	8.85	8.68
Linoleic acid (%)	2.36	2.37	2.32	2.33	2.83	2.83	2.79	2.78	3.07	3.01	2.96	2.95
Crude fibre%	4.73	4.79	4.86	4.89	4.52	4.58	4.52	4.56	4.33	4.33	4.39	4.46

* Amino acid composition calculated based on ingredients analysed

Table.4 Amino acid compositions calculated for pre-starter, starter and finisher diets

Ingredient (%)	Pre-starter diets				Starter diets				Finisher diets			
	T_1	T_2	T_3	T_4	T_1	T_2	T_3	T_4	T_1	T_2	T_3	T_4
Lysine	1.266	1.263	1.261	1.264	1.131	1.134	1.134	1.133	1.009	1.001	1.003	1.006
Methionine	0.577	0.585	0.592	0.600	0.505	0.517	0.535	0.545	0.463	0.477	0.491	0.505
Methionine + Cysteine	0.931	0.926	0.919	0.912	0.833	0.831	0.835	0.831	0.765	0.766	0.766	0.766
Threonine	0.860	0.861	0.861	0.865	0.765	0.760	0.762	0.763	0.708	0.702	0.709	0.707
Valine	1.062	1.013	0.960	0.908	0.973	0.920	0.867	0.815	0.880	0.828	0.776	0.724
Isoleucine	0.975	0.921	0.864	0.809	0.886	0.830	0.775	0.720	0.793	0.738	0.682	0.626
Leucine	1.907	1.828	1.747	1.667	1.774	1.693	1.618	1.539	1.642	1.567	1.486	1.405
Tryptophan	0.270	0.254	0.238	0.222	0.243	0.227	0.211	0.195	0.215	0.199	0.183	0.167
Arginine	1.583	1.498	1.406	1.316	1.432	1.342	1.249	1.159	1.273	1.182	1.091	1.001
Tyrosine	0.898	0.852	0.803	0.754	0.817	0.769	0.720	0.671	0.733	0.685	0.636	0.587
Serine	1.335	1.262	1.185	1.110	1.212	1.136	1.061	0.986	1.084	1.009	0.933	0.857
Histidine	0.616	0.587	0.557	0.527	0.565	0.535	0.506	0.476	0.514	0.485	0.455	0.425
Phenylalanine	1.150	1.091	1.029	0.969	1.051	0.990	0.930	0.870	0.948	0.889	0.828	0.767
Glycine	0.961	0.901	0.841	0.783	0.878	0.818	0.767	0.709	0.792	0.737	0.678	0.618
Glycine + Serine	2.296	2.164	2.026	1.893	2.090	1.955	1.828	1.694	1.876	1.747	1.611	1.475

* Amino acid composition calculated based on ingredients analysed

Table.5 Weekly cumulative body weight (g/bird) of commercial broilers as influenced by dietary supplementation of L-threonine by reducing the levels of crude protein

Treatment	Crude protein reduction (%)	Weekly body weight (g)					
		I	II	III	IV	V	VI
T_1	Control	163.85[a]	456.95[a]	845.93[a]	1327.46[a]	1839.23[a]	2345.26[a]
T_2	1.0	165.49[a]	454.33[a]	839.67[a]	1323.63[a]	1821.46[a]	2337.92[a]
T_3	2.0	155.18[b]	431.57[b]	802.34[b]	1217.86[b]	1676.34[b]	2151.09[b]
T_4	3.0	150.21[b]	404.54[c]	742.24[c]	1111.75[c]	1550.23[c]	2005.92[c]
S.Em. ±		**2.84**	**5.55**	**9.62**	**28.69**	**22.86**	**16.59**
C.D. at 5%		**8.38**	**16.38**	**28.37**	**84.64**	**67.43**	**48.95**
F value		**6.49***	**19.26***	**24.49***	**12.75***	**35.24***	**96.57***

Note: The pairs with different superscripts are significant at 5%

Table.6 Cumulative feed consumption (g/bird) of commercial broilers as influenced by dietary supplementation of L-threonine by reducing the levels of crude protein

Treatment	Crude protein reduction (%)	Weekly feed consumption (g)					
		I	II	III	IV	V	VI
T_1	Control	138.15 [ab]	519.01 [a]	1075.65 [a]	1816.27 [a]	2743.23 [a]	3975.32 [a]
T_2	1.0	141.23 [a]	511.73 [b]	1056.26 [b]	1803.22 [b]	2725.72 [a]	3970.72 [a]
T_3	2.0	138.28 [ab]	512.45 [b]	1039.53 [c]	1761.87 [c]	2664.70 [b]	3894.71 [b]
T_4	3.0	136.05 [b]	504.29 [c]	996.55 [d]	1684.05 [d]	2570.05 [c]	3794.54 [c]
S.Em. ±		**1.07**	**2.03**	**3.20**	**4.35**	**6.81**	**7.49**
C.D. at 5%		**3.17**	**5.99**	**9.45**	**12.83**	**20.10**	**22.10**
F value		**3.93***	**8.79***	**110.64***	**187.43***	**131.75***	**127.83***

Note: The pairs with different superscripts are significant at 5%

Table.7 Weekly cumulative feed conversion ratio of commercial broilers as influenced by dietary supplementation of L-threonine by reducing the levels of crude protein

Treatment	Crude protein reduction (%)	Weekly feed conversion ratio					
		I	II	III	IV	V	VI
T_1	Control	0.84 [b]	1.14 [bc]	1.27 [bc]	1.37 [b]	1.49 [c]	1.70 [c]
T_2	1.0	0.85 [b]	1.13 [c]	1.26 [c]	1.38 [b]	1.50 [c]	1.71 [c]
T_3	2.0	0.89 [a]	1.18 [ab]	1.30 [b]	1.45 [ab]	1.59 [b]	1.81 [b]
T_4	3.0	0.91 [a]	1.22 [a]	1.34 [a]	1.52 [a]	1.65 [a]	1.89 [a]
S.Em. ±		**0.02**	**0.02**	**0.01**	**0.03**	**0.02**	**0.01**
C.D. at 5%		**0.05**	**0.05**	**0.04**	**0.10**	**0.06**	**0.04**
F value		**3.79***	**7.59***	**7.78***	**4.56***	**14.84***	**56.86***

Note: The pairs with different superscripts are significant at 5%

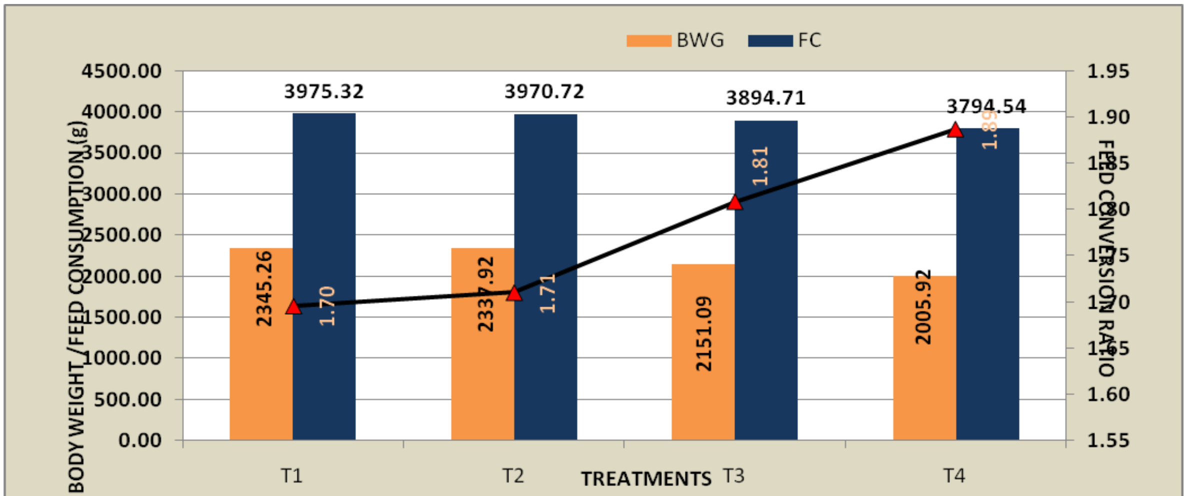

Fig. 1: The influence of supplementation of L-threonine on growth performance parameters in reduced CP diets of various treatments on cumulative period (1-42d) in commercial broilers

There was no significant difference in cummulative FCR up to 1 percent unit CP reduced diet fed birds with L-threonine supplementation (T_2) as compared to that of control diet (T_1) but on further 2 and 3 percent unit reduced CP diets (T_3 andT_4) FCR was significantly poorer from the end of the first week to sixth week as compare to control diet (T_1) except at the end of third and fourth week 2 precent unit reduced CP diet (T_3) did not showed significant difference in FCR compare to that of control diet (T_1). However, 3 percent unit reduced CP diet fed birds had poor FCR during entire experimental period compare to that of control diet (T_1) (Table 7).

Marginally better FCR ratio with threonine supplementation observed in this study was in agreement with the findings of Kidd *et al.*, (1999) and Bade *et al.*, (2014). The findings of the trial were also in agreement with the observations of Abbasi *et al.*, (2014) the feed conversion ratio indicated better values in birds fed on diets containing 95% of crude protein level supplemented with 120% threonine of the standard recommended levels. Further, FCR values are in agreement with findings of Anand (2015) reported that there was no significant difference in feed conversion ratio up to 1.5% unit reduced CP with L-threonine supplementation as compare to control diet, but FCR was significantly poorer when the CP level in the diets decreased at 2.25%

According to Ahmed (2014) control diet had better FCR but comparable FCR was also observed in 0.5 and 1% reduced CP diet fed broilers but 1.5% reduced CP diets showed poor FCR,these reports resembles the present study that 2 and 3percent unit CP reduced diet fed birds showed poor FCR. Contrary to our findings in this experiment Hai and Blaha (1998) found no difference in FCR when the protein level of the isocaloric diets was reduced from 20 to 16% crude protein and supplemented with essential amino acids. Further, results were contrary to the observations of Han *et al.*, (1992), Kerr and Kidd (1999b), Araujo *et al.*, (2004) and Rezaei *et al.*, (2004) who found no differences in FCR of the birds when low crude protein diets supplemented with amino acids according to an ideal amino acid ratio.

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How to cite this article:

Ashok Madapurada, Neeraj, Ramesh Pandey, Sushma, G. Ananda Manegar and Krishnamurthy, T.N. 2017. Influence of L-threonine Supplementation to Diets with Reduced Crude Protein on Feed Efficiency in Commercial Broilers. *Int.J.Curr.Microbiol.App.Sci.* 6(12): 4224-4231.
doi: https://doi.org/10.20546/ijcmas.2017.612.486