Original Research

Effect of Dietary Supplementation of Synthetic L-Lysine and DL-Methionine on Growth Performance, Nutrient Utilization and Immunity in Commercial Broiler Chicken

Bornalee Handique*, Gunaram Saikia, Runjun Dowarah, Bibeka Nanda Saikia and Joga Dev Mahanta[1]

Department of Animal Nutrition, College of Veterinary Science, Assam Agricultural University, Khanapara, Guwahati-781022, Assam, INDIA

[1]Department of Poultry Science

***Corresponding author:** bornalee16@gmail.com

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Abstract

An experiment was conducted to investigate the effect of synthetic L- lysine and DL-methionine on growth performance, nutrient utilization and humoral immunity in commercial broilers. 144 Day old broiler chicks (Vencobb 400) were randomly divided into four groups (3 replicates of 12 each) and basal diet supplemented with synthetic L-lysine @ 0, 20, 30 and 40% and DL-methionine @ 0, 30, 40 and 50% higher than BIS (2007) recommendation for 42 days. Supplementation of synthetic L-lysine and DL-methionine improved (P<0.001) final body weight and gain in weight at finisher phase as compared to control. The best feed conversion ratio (FCR) and protein efficiency ratio (PER) was observed in T_2 group supplemented with 20% lysine and 30% methionine. Higher (P<0.05) digestibility of crude protein and fibre and nitrogen retention was observed by supplementation of L-lysine and DL-methionine in broiler chicken. Supplementation of synthetic L- lysine and DL-methionine improved humoral immune response in the experimental birds with higher antibody titre.

Key words: Broiler, DL-Methionine, Growth Performance, Immunity, L-Lysine, Nutrient Utilization

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Introduction

Due to increase in world population there is a serious problem in providing food for all people. Poultry meat is the source of animal protein for fight against malnutrition and to overcome the problem of food scarcity. Therefore, in recent year's poultry farming is the profitable and fastest growing food industry in

animal husbandry sector. Preparation of balanced poultry feeds requires various feed ingredients, among all the feed ingredients protein rich feed ingredients cost higher. Cereal grains are deficient in critical amino acids like lysine, methionine, threonine and tryptophan (Smith, 2001). However, the animal protein supplements such as fish and meat meal though rich in these essential amino acids but their use in poultry ration are not affordable for their short supply, very expensive and restriction for use due to adulteration. Quality of the protein is more important than the quantity of protein used. The major problem facing poultry farmers is high cost of protein feed resources but it can be overcome by supply the right quantity and quality of protein *i.e.* amino acids in the diets. Since, amino acids are responsible for protein synthesis in the body which is the primary constituents of structural and productive tissues such as skin, feathers, bone matrix, ligaments, soft tissues and different organs. Lysine is essential for protein synthesis, regulation of cell division, calcium absorption, mobilization of fatty acid into the mitochondria and helps in enhancing overall growth performance, breast meat yield, efficiency of feed utilization, carcass protein retention and reduces fat deposition. Methionine plays roles in poultry such as involve in protein synthesis, acts as methyl donor for cellular metabolism and formation of coenzyme S-adenosylmethionine involves in polyamine synthesis, act as sulphur donor and as lipotropic agent. Considering the above facts present experiment was conducted to study the effect of dietary supplementation of synthetic L-lysine and DL-methionine in excess of BIS (2007) recommendation for broiler chicken.

Materials and Methods

This experiment was conducted in the experimental poultry shed, Department of Animal Nutrition, College of Veterinary Science, Assam Agricultural University, Khanapara, Guwahati-781022, Assam . A total of 144 day old broiler chicks (Vencobb strain 400) randomly distributed into 4 treatment groups (3 replicates of 12 chicks each) using randomized block design *viz.*, T_0-basal diet (control), T_1, T_2 and T_3 were offered the basal diet supplemented with synthetic L- lysine @20, 30 and 40 percent and DL- methionine at 30, 40 and 50 percent in excess of the total BIS (2007) requirement. The basal diet was prepared as per BIS (2007) without supplementation of L-lysine and DL-methionine (Table 1). All the chicks were reared in deep litter system under similar environmental conditions (temperature 20-27^{o}C and relative humidity 78-88%) and had access to *ab libitum* feed and water during the entire experimental period. The chicks were vaccinated for Ranikhet disease with Lasota strain (F1) vaccine on 7th day and 14th day of age Infectious Bursal Disease (IBD) vaccine (dose as per company's recommendation). Performance of the broiler in respect of weekly feed intake, digestibility of nutrients, weekly body weight and body weight gain, feed conversion ratio (FCR), protein efficiency ratio (PER) were calculated.

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Table 1: Percent ingredient and nutrient composition of the experimental basal diet

Attributes	Starter (Kg)	Finisher (Kg)
Percent Ingredient Composition		
Maize	51	54.3
De-oiled rice polish	5.5	7.2
De-oiled GNC	15	10
Soybean meal	23.5	22.5
Vegetable oil	3	4
Mineral mixture	1.5	1.5
Salt	0.5	0.5
Nutrient Composition on DM basis (%)		
Dry matter	92.23	92.09
Crude protein	22.08	20.01
Crude fibre	5.78	5.1
Ether extract	4.2	4.3
NFE	54.79	57.59
Total ash	13.15	13
*ME(Kcal/kg)	3130	3202
*Lysine	1.2	1
*Methionine	0.5	0.45

**Calculated values (N.B. Vitamin premix (Vitablend vit A, B2, D3, K) was added @ 20 g per quintal of diet in both starter and finisher diet. Mineral mixture contained calcium 25%, Phosphorus 5%, Sodium chloride 23%, Iodine 10 ppm, Copper 100 ppm, Manganese 2000 ppm and Cobalt 10 ppm.*

At the end of the feeding trial (after 42 days), a metabolism trial was conducted for five days (2 days adaptation and 3 days of collection) to assess the digestibility and balance of nutrients. Humoral immune response was also measured at the end of the feeding trial. The data's were analyzed as a randomized complete block design using GLM procedures of SPSS (version 17.0 for Windows; SPSS, Chicago, III., U.S.A). Difference among treatment means were determined by one way and two-way ANOVA and compared according to Duncan's multiple range test (Duncan, 1995).

Results and Discussion

Growth Performance

The change of body weights in supplemented groups (T_1, T_2 and T_3) were significantly higher ($P<0.001$) as compared to control group (T_0) during 4-6 weeks of feeding (Table 2). Whereas, non-significant ($P>0.05$) effect had been observed in average body weight change among the experimental groups during 0-3 weeks of age. Similarly, gain in weight (g/bird) from 0-3 weeks was also did not show any significant effect due to supplementation of synthetic L- lysine and DL-methionine in the diet (Table 2). The present findings were accordance with the report of Kalbande *et al.* (2009) observed in broilers fed on different level of synthetic methionine supplemented diets. Batal and Dale (2006) reported that the benefit of increase lysine levels may not be seen at early stage of growth but at the end of the finisher phase. Higher ($P<0.001$) total

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gain in weight was observed in supplemented groups (T_1, T_2 and T_3) as compared to control (T_0) whereas, T_2 and T_3 groups had better ($P<0.001$) total gain (0-6 weeks) among the treatment groups. It might be due to the effect of balance of both lysine and methionine and their combined effect in improving feed and nutrient utilization and more availability of lysine and methionine for protein synthesis. This corroborates with the finding of Bowa *et al.* (2008) reported that consumption of lysine and methionine resulted in significant increase in weight gain. Feed intake in the broilers did not showed any significant difference among the experimental groups during the entire experimental period. Contrary to the present findings, Pillai *et al.* (2006) reported higher ($P<0.05$) feed intake with addition of high level of methionine in the broiler ration. However, there was a slight increase of feed intake at high level of lysine and methionine supplemented groups that might be the improvement of the quality of feed due to amino acid balance. The observed values of the total feed consumption by the experimental birds were comparable to the values reported by Kalbande *et al.* (2009), Ahmed and Abbas (2011 and 2015) and Zhai *et al.* (2016).

Table 2: Effect of synthetic L-lysine and DL-methionine on growth performance in commercial broiler chicken

Attributes	Dietary Treatment				SEM	P value
	T_0	T_1	T_2	T_3		
Average Body Weight						
0 day	42.89±0.57	42.89± 0.55	43.33±0.50	42.17±0.49	0.26	0.464
0-3 week	644.80±4.20	646.80±13.20	647.70±11.9	668.80±12.70	12.7	0.53
4-6 week	1754^{a}±23.90	1843^{b}±26.80	1963^{b}±26.90	1925^{b}±23.60	14.2	<0.001
Gain in Weight (g/bird)						
0-3 week	306.50±9.42	308.70±9.52	321.00±9.52	332.80±8.66	4.68	0.166
4-6 week	424.20^{a}±8.00	414.10^{a}±9.28	501.30^{c}±9.50	458.60^{b}±9.15	5.29	<0.001
Total gain	1713.2^{a}±23.9	1799.9^{b}±26.8	1923.4^{c}±27.0	1882.6^{c}±23.5	14.2	<0.001
Feed Intake (g/bird)						
0-3 week	857.5±29.4	857.1±8.07	900.1±28.9	981.4±31.4	23.9	0.052
4-6 week	2433.5±17.4	2407.3±5.58	2453.0±8.58	2426.9±17.5	9.44	0.145
0-6 week	3291±21.0	3214±25.1	3353±37.8	3408±14.1	73.17	0.881
FCR						
0-3 week	1.55±0.12	1.39±0.17	1.41±0.22	1.39±0.06	0.08	0.424
4-6 week	2.18^{b}±0.01	2.01^{b}±0.01	1.86^{a}±0.01	1.92^{b}±0.01	0.05	<0.001
0-6 week	1.92^{b}±0.05	1.79^{b}±0.03	1.75^{a}±0.01	1.81^{b}±0.11	0.13	0.042
PER						
0-3 week	3.69±1.01	3.99±1.29	3.69±0.30	2.52±0.21	0.36	0.76
4-6 week	2.18^{a}±0.01	2.34^{b}±0.15	2.48^{c}±0.01	2.36^{b}±0.10	0.05	<0.001
0-6 week	2.93±0.37	3.16±0.36	3.08±0.11	2.94±0.24	0.12	0.925

*^{abc}*Mean values with different superscripts within row differ significantly*

The feed conversion ratio (FCR) was narrowest in 0-3 weeks of age in the all four experimental groups but wider in 4-6 weeks of age (Table 2). T_2 group showed the best ($P<0.05$) FCR as compared to other groups during 4-6 weeks and 0-6 weeks of the experimental period. It might be due to better effect of lysine and

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methionine supplementation in nutrient utilization resulting highest body weight gain of broilers in this group where effective balance of lysine and methionine in the diet. Similar pattern of findings were reported by Osti and Pandey (2004). Numerically similar value of FCR in broilers were reported by Kalbande *et al.* (2009), Kanduri *et al.* (2013), Ahmed and Abbas (2015) by feeding diet supplemented with synthetic and herbal amino acids.

Protein efficiency ratio (PER) during the whole experimental period in all the treatments were decreased along with the increase of age (Table 2). Significant differences (P<0.001) were found in 4-6 weeks of age; where supplemental groups (T_1, T_2.and T_3) showed significantly better (P<0.001) PER than control group. Present findings were comparable to the experiment reported by Ahmed and Abbas (2011) where broiler diets were supplemented with methionine at higher level (120 and 130%) than NRC recommendation. The better PER in T_2 group might be due to better nutrient utilization of crude protein digestibility and higher nitrogen retention. This result was in agreement with the findings of Onu *et al.* (2010) where supplementation of synthetic lysine and methionine give better result as compare to un-supplemented group.

Nutrients Digestibility and Balance

Apparent dry matter and CP digestibility was better (P<0.05) in supplemented groups compared to control (Table 3).

Table 3: Effect of synthetic L-lysine and DL-methionine on digestibility and balance nutrient in broiler chicken

Attributes	Dietary Treatment				SEM	P value
	T_0	T_1	T_2	T_3		
Nutrient Digestibility						
DM	64.74^{a}±1.09	76.97^{b}±0.85	77.90^{b}±2.43	75.43^{b}±0.98	1.05	0.004
CP	61.16^{a}±1.77	64.99^{b}±1.67	71.79^{c}±1.07	63.06^{b}±3.16	1.64	0.003
CF	21.95±0.28	21.97±0.20	24.73±0.11	22.11±0.10	0.1	0.899
EE	64.76±4.19	72.64±5.11	73.75±3.16	68.69±5.68	2.79	0.06
NFE	53.36±1.77	52.05±1.35	54.03±0.94	51.97±0.81	0.605	0.672
Nutrient Balance						
Nitrogen	60.93^{a}±93	64.99^{b}±0.68	71.78^{c}±0.69	63.06^{b}±0.56	0.51	0.01
Calcium	51.19±0.89	51.93±0.55	51.37±0.84	51.38±0.94	0.37	0.925
Phosphorus	50.38±1.89	50.64±1.06	50.41±0.43	50.33±1.86	0.65	0.999

^{abc}*Means with different superscripts within the same row differ significantly.*

It might be due to better feed utilization by the bird due to adequate balance of amino acid in the diet. T_2 group had better (P<0.05) digestibility amongst the supplemental groups. Non-significant (P>0.05) differences were observed between the experimental groups in respect of EE, CF and NFE digestibility in the commercial broilers (Table 2). The present study was in agreement of finding of Onu *et al.* (2010).

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Higher (P>0.05) CF digestibility in T_2 was due to increase efficiency of utilization of energy and protein providing optimum environment for bacterial growth in the large intestine. Similar digestibility co-efficient of CF in broiler were reported by Sonowal (2008). There was significant (P<0.05) difference in percent nitrogen retention among the experimental groups and the values were higher (P<0.05) in T_2 group as compared to other groups. It might be due to digestive environment creating by adequate balance of amino acid. Present findings were similar with findings of Gorman and Balnave, 1995; Si *et al*., 2001 and Xu *et al*., 2003. The retention of both calcium and phosphorus were found to be non-significant (P>0.05) among the experimental groups for entire experimental period (Table 3). Similar finding was reported by Ogunwole *et al.*, 2017.

Humoral Immune Response

The humoral immunity (HI) was expressed as the antibody response to sheep erythrocytes (SRBC) deploying HA test. Antibody titre value (log_2 titre) was maximum on 5th day of post inoculation (Table 4).

Table 4: Effect of synthetic L-lysine and DL-methionine on antibody titre (log_2 titre) against sheep RBC in broilers chicken

Treatment	Period			Significant		
	3d	5d	Mean ±SE	T	P	TXP
T_0	0.90±0.14	1.05±0.15	0.98^{a}±0.17	0.014	0.141	0.089
T_1	1.50±0.28	1.35±0.15	1.43^{b}±0.22			
T_2	1.20±0.20	1.80±0.00	1.50^{b}±0.36			
T_3	1.20±0.00	1.20±0.00	1.20^{ab}±0.16			
Mean ±SE	1.20±0.09	1.35±0.11				

*^{ab}*Means in the same column bearing different superscripts differ significantly*

The supplemented groups had showed higher (P=0.014) antibody titre in comparison to control (T_0), however T_1 and T_2 groups had higher (P=0.014) immune response than T_3 group. It might be due to lysine and methionine constructively affects the humoral immune response. Similar findings were reported by Shini *et al*. (2005) and Bouyeh (2012) by diet supplemented with lysine and Methionine (as TSAA) more than NRC (1994) recommendation.

Conclusion

Dietary supplementation of synthetic L-lysine and DL-methionine @ 30 and 40% respectively influenced the growth performance, utilization of dry matter and crude protein, balance of nitrogen and humoral immune status in broiler chicken. This clearly suggests that supplementation of L-lysine and DL-methionine could be strategy to increase performance and immunity in broilers. However, further studies were required to ascertain the observed findings in the present study.

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