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Lysine Requirement of Broiler Chickens Fed Low-density Diets under Tropical Conditions

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ABSTRACT : Two experiments were conducted to determine the lysine requirement of straight-run broiler chickens (Hubbard× Hubbard) during the period 4-21 (Exp. 1) and 21-40 (Exp. 2) days of age. Experiments were conducted during the summer months (June-August) in open-sided houses, thus exposing chicks to chronic heat stress. Daily min-max temperature averaged 28-40°C (Exp. 1) and 23-36°C (Exp. 2). Lysine deficient basal diets were formulated to contain low-nutrient-density, i.e., 2,700 kcal per kg ME, 18.6% CP (Exp. 1), and 2,750 kcal per kg ME, 17.1% CP (Exp. 2), to mimic typical commercial broiler diets used in Pakistan. Diets were supplemented with L-lysine HCl to provide total lysine level ranging from 0.85 to 1.10% (six increments) and 0.72 to 1.02% (six increments), respectively in Exp. 1 and 2. Live performance data were subjected to quadratic analysis and requirement was defined as the level achieving 95% of maximum or minimum values. Lysine requirements were found to be 0.98 and 0.97% total lysine, respectively, for gain and feed efficiency during 4-21 days, and 0.87% total lysine for both gain and feed efficiency during 21-40 days of age. Calculated on a digestible lysine basis, the estimates were 0.85 and 0.84%, respectively, for gain and feed efficiency during 4-21 days of age; and 0.75% for gain and feed efficiency during 21-40 days of age. (**Key Words** : Broilers, Heat-stress, Nutrient-density, Lysine Requirement)

INTRODUCTION

Lysine requirements of broiler chicks during different growth phases are well established (Han and Baker, 1994; Knowles and Southern, 1998; Mack et al., 1999; Baker et al., 2002; Corzo et al., 2003). Most of these studies are conducted under the "standard" conditions i.e. feeding high-nutrient-density basal diets (3,200 kcal per kg ME; 20-23% CP) to chickens kept under thermo-neutral conditions. But actual raising and feeding conditions can be much different from these "standard" conditions, which would affect lysine requirement. In Pakistan, for instance, broilers are reared on low-nutrient-density diets (2,700-2,750 kcal per kg ME; 17-19% CP). Furthermore, chickens are mostly reared in open-sided houses and are exposed to chronic heat stress during the summer season.

The objective of the present experiments was to determine the lysine requirement of broiler chickens fed low-nutrient-density diets and under severe heat stress conditions. The low-density diets are typically used in commercial broiler industry in Pakistan. Results thus obtained should be more readily applicable by the broiler industry in this region which shares similar feeding and raising conditions.

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MATERIALS AND METHODS

Experiment 1

The experiment was designed to evaluate lysine requirement of broiler chickens during 4-21 days of age, reared under chronic heat stress conditions. The experiment was conducted during the months of June-July 2004. Minimum-maximum temperature was recorded twice daily (8 a.m. and 6 p.m.) throughout the course of study. A lysine deficient basal diet was prepared, based on analyzed amino acid content of the main raw materials, to contain 0.85% total lysine (0.72% digestible lysine, Table 1). The basal diet was supplemented with 0, 0.064, 0.128, 0.192, 0.256 and 0.32% L-lysine HCl (79% lysine) to obtain total lysine levels of 0.85, 0.90, 0.95, 1.00, 1.05 and 1.10%, respectively. Corresponding digestible lysine levels were calculated to be 0.72, 0.77, 0.82, 0.87, 0.92 and 0.97%, respectively. Added crystalline lysine was assumed to be 100% bioavailable (Chung and Baker, 1992). Crude Protein

Table 1. Composition (% as is) of the lysine deficient basal diets

Ingredients	Experiment 1 (4-21 days)	Experiment 2 (21-40 days)
Corn	59	64
Soybean meal 46%	12.8	7.5
Canola meal 37%	10	10
Cottonseed meal 40%	2	2
Sunflower meal 34%	8	10
DL-methionine	0.14	0.120
L-threonine	0.082	0.105
Molasses	4.14	2.68
Limestone	0.5	0.8
Bone ash	2.6	2.0
Salt	0.15	0.18
Sodium bi-carbonate	0.1	0.12
Vitamin/mineral premix [A]	0.5	0.5
Calculated composition		
ME (kcal per kg)	2,700	2,750
CP (%)	18.6	17.1
Calcium (%)	0.90	0.90
Available phosphorus (%)	0.45	0.40
Lysine (%)	0.85	0.72
Methionine+cysteine (%)	0.81	0.76
Threonine (%)	0.78	0.74
Digestible lysine (%)	0.72	0.60

All other amino acids were set to meet or exceed the ideal ratios (relative to digestible lysine) as calculated with reference value of 0.97% digestible lysine for 4-21 days (Baker and Han. 1994) and 0.90% digestible lysine for 21-40 (Mack et al., 1999).

[A] Provided per kg of diet: vitamin A, 8,000 IU; cholecalciferol, 2,000 ICU; vitamin E, 30 mg; manadione, 2 mg; riboflavin, 5.5 mg; pantothenic acid, 13 mg; niacin, 36 mg; choline, 500 mg; vitamin B_{12}, 0.02 mg; folic acid, 0.5 mg; thiamin, 1 mg; pyridoxine, 2.2 mg; biotin, 0.05 mg; ethoxiquin, 125 mg; Mn, 65 mg; Fe, 55 mg; Cu, 6 mg; Zn; 55 mg.

(CP) contributed by L-lysine HCl was ignored and not corrected between diets of the supplemental series, resulting in a slight increase in actual diet CP with each increment of L-lysine HCl. All other amino acids in the basal diet were set to meet or exceed the Ideal Protein ratios (Baker and Han. 1994) calculated at 0.97% digestible lysine (the maximum level of digestible lysine used in this experiment) in order to ensure that none of the amino acids, except lysine, was deficient in any of the test diets. With each increment of L-lysine HCL, dietary level of salt was reduced (from 0.15% to 0.04% in increments of 0.022%) and sodium bi-carbonate was increased (from 0.10% to 0.27% in increments of 0.034%) to maintain similar dietary electrolyte balance (Na+K-Cl) among all test diets.

Exp. 1 was conducted in two houses, situated in the same vicinity and managed in a similar way. A total of 1,500 day-old straight-run chicks (Hubbard×Hubbard) were reared on a common starter diet for the first 4-days of age. At the 4th day of age, feed was removed for eight hours and chicks were weighed individually. On the basis of similar body weights, a total of 840 chicks was selected and randomly divided into 42 floor pens (20 chicks per pen) so that average initial body weight and range were alike among all pens. Individual pen measurement was 6×3.5 ft^2. Out of the total 42 pens, 24 were in House-I and 18 were in House-II. Each of the six experimental diets was then randomly assigned to 7 pens (4 each in House-I and 3 each in house-II). Birds were offered feed and water for *ad-libitum* intake, and 24 h light was provided. The experiment was conducted during 4-21 days of age. At Day 21, feed intake, body weight gain, mortality, and FCR were recorded on a pen basis.

Experiment 2

The experiment was conducted during the month of August 2004. Daily low-high temperature was recorded at 8 a.m. and 6 p.m., respectively, throughout the course of study. A practical-type lysine deficient basal diet was prepared, based on analyzed amino acid content of the main raw materials, to contain 0.72% total lysine (0.6% digestible lysine). Composition of the lysine deficient basal diet is given in Table 1. The basal diet was supplemented with 0, 0.076, 0.152, 0.228, 0.304 and 0.380% L-lysine HCL to obtain total lysine levels of 0.72, 0.78, 0.84, 0.90, 0.96 and 1.02%, respectively. Corresponding digestible lysine levels were calculated to be 0.60, 0.66, 0.72, 0.78, 0.84 and 0.90%, respectively. All other amino acids in basal diet were set to meet or exceed the Ideal Protein ratios (Mack et al., 1999) calculated at 0.90% digestible lysine (the maximum level of digestible lysine used in the supplemental series) in order to ensure that the response of lysine should not be restricted by the deficiency of other amino acids. With each increment of L-lysine HCL, dietary level of salt was reduced (from 0.18% to 0.06% in increments of 0.024%) and sodium bi-carbonate was increased (from 0.12% to 0.28% in increments of 0.032%) to maintain similar dietary electrolyte balance (Na+K-Cl) among all test diets.

A total of 1,000 day-old straight-run chicks (Hubbard×Hubbard) were reared on a common starter diet for first 21-days of age. At the 21st day of age, feed was removed for eight hours and chicks were weighed individually. On the basis of similar body weights, a total of 480 chicks was selected and randomly divided into 24 floor pens (20 chicks per pen) so that average initial body weight and range were alike among all pens. Individual pen measurement was 6×3.5 ft^2. Each of the experimental diets was then randomly assigned to 4 pens. Birds were offered feed and water for *ad-libitum* intake, and 24 h light was provided. At 40 days of age, feed intake, body weight gain, FCR, and mortality were recorded on a pen basis.

Statistical analysis

Feed intake and total body weight gain was recorded at 21 days (Exp. 1) or 40 days (Exp. 2) posthatch and were used to calculate the body weight gain and FCR for

Table 2. Feed intake, body weight gain, and FCR for Experiment 1

Total lysine (%)	Digestible lysine (%)	Feed intake (g per bird)	weight gain (g per bird)	FCR
0.85	0.72	743±65.7[a]	281±18.2[a]	2.64±0.22[a]
0.9	0.77	927±38.6[b]	454±16.2[b]	2.04±0.06[b]
0.95	0.82	990±37.2[c]	528±19.4[c]	1.88±0.06[b]
1.00	0.87	968±21.3b[c]	515±7.8[c]	1.88±0.05[b]
1.05	0.92	967±35.1b[c]	519±27.0[c]	1.86±0.09[b]
1.10	0.97	983±23.5b[c]	528±19.5b[c]	1.86±0.09[b]
Pooled SEM		14.97	7.14	0.042

Values represent mean of 7 replicate pens (20 chicks per pen) during 4-21 days of age.

[a-c] Means in a column with no common superscript differ significantly ($p<0.05$).

Table 3. Feed intake, body weight gain, and FCR for Experiment 2

Total lysine (%)	Digestible lysine (%)	Feed intake (g per bird)	Weight gain (g per bird)	FCR
0.72	0.60	1,585±108[a]	521.8±40.3[a]	3.04±0.042[a]
0.78	0.66	2,123±109[b]	832.3±61.8[b]	2.55±0.085[b]
0.84	0.72	2,193±60.1[b]	881.3±43.7[b]	2.49±0.064[b]
0.90	0.78	2,233±53.3[b]	894.0±41.7[b]	2.50±0.081[b]
0.96	0.84	2,246±62.9[b]	921.8±41.6[b]	2.44±0.052[b]
1.02	0.90	2,159±130[b]	859.5±89.6[b]	2.51±0.136[b]
Pooled SEM		46.0	28.03	0.041

Values represent mean of 4 replicate pens (20 chicks per pen) during 21-40 days of age.

[a-b] Means in a column with no common superscript differ significantly ($p<0.05$).

Table 4. Summary of the estimated lysine requirement from quadratic response curves (Exp. 1 and 2).

Age (days)	Variable	Quadratic response curve	R-sq	Estimated requirement[1]
4-21	Gain	$-7,857.1 X^2+16,131 X-7,730.5$	0.91	0.98 (0.85)[2]
4-21	FCR	$25.756 X^2-52.763 X+28.80$	0.91	0.97 (0.84)[2]
21-42	Gain	$-9,666.7 X^2+17,758 X-7,212.8$	0.91	0.87 (0.75)[2]
21-42	FCR	$13.966 X^2-25.712 X+14.239$	0.88	0.87 (0.75)[2]

[1] Calculated at 95% of the maximum response. [2] Values in parenthesis represent the digestible amino acids.

[R-sq] Coefficient of determination.

respective experiments; mortality was recorded daily and the weight of dead birds was used to correct the FCR. Data regarding feed intake, gain, FCR and mortality were subjected to Analysis of Variance according to a Completely Randomized Design, using Minitab. The differences for all parameters were tested according to the following statistical model:

$$Y_{ij} = u+T_i+e_{ij}$$

Where,

Y_{ij} equals the variance associated with a parameter,

u is the overall mean,

T_i is the treatment effect, and e_{ij} is the error term.

Pens were treated as experimental units and where the differences between treatments reached the conventional level of significance ($p<0.05$) mean separation was calculated using the Tukey's test.

Quadratic regression analysis ($Y = a+bx+cx^2$) was fitted to calculate the optimum lysine requirement for gain and FCR; 0.95 of the maximum or minimum response was considered as "requirement" in each case.

RESULTS AND DISCUSSION

The test diets of the present study closely mimicked practical-type broiler diets used in Pakistan in terms of choice of raw materials and nutrient density. Average daily low-high temperature was found to be 28-40°C in Exp. 1. and 23-36°C in Exp. 2. These temperatures were stressful for birds, as observed by severe panting and very low feed intakes during the hot hours of the day.

Results of Exp. 1 are given in Table 2. Analysis of variance showed that the house effect was non-significant for each of the parameters studied, so the data were combined. Feed intake, body weight gain, and FCR were significantly ($p<0.05$) affected by dietary treatments. Weight gain, feed intake, and feed efficiency improved quadratically ($p<0.05$) with incremental dietary lysine addition. Quadratic regression analysis (Figure 1; Table 4) revealed a requirement (95% of maximum or minimum values) of 0.98% and 0.97% total lysine (0.85 and 0.84%

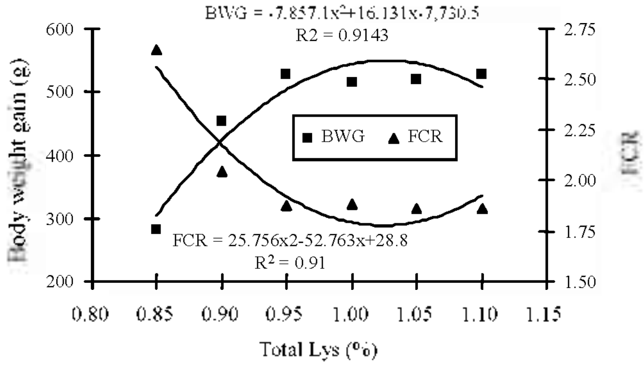

Figure 1. Body weight gain (BWG) and feed conversion ratio (FCR) of broilers fed progressive levels of dietary lysine during 4-21 days of age (Exp. 1) subjected to quadratic analysis.

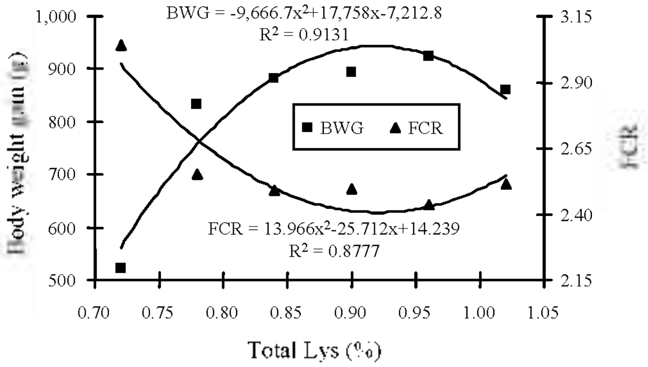

Figure 2. Body weight gain (BWG) and feed conversion ratio (FCR) of broilers fed progressive levels of dietary lysine during 21-40 days of age (Exp. 2) subjected to quadratic analysis.

digestible lysine) for body weight gain and feed efficiency, respectively.

Similar trends were observed in Exp. 2. Feed intake, body weight gain, and feed efficiency were significantly ($p<0.05$) depressed by the basal (un-supplemented) diet. Supplementation of increments of lysine to the basal diet improved feed intake, gain, and FCR quadratically ($p<0.05$). Quadratic regression analysis (Figure 2; Table 4) revealed a requirement (95% of maximum or minimum values) of 0.87% total lysine (0.75% digestible lysine) for both body weight gain and feed efficiency. Overall mortality did not exceed 3% in each experiment, and no treatment effect was found (data not shown).

The poor live weight performance of broilers fed the basal diets and the responses to supplemental L-lysine indicated that the basal diets were severely deficient in lysine (Tables 2 and 3). The performance depression could, at least in part, be attributed to the low feed intake as a result of feeding diets with amino acid imbalances (Byung-chul Park, 2006). Requirement of lysine for gain and feed efficiency were similar in each experiment. Others have reported a higher requirement for feed efficiency than for gain (Han and Baker, 1994; Baker et al., 2002). Leclercq (1998) has shown that there is a hierarchy of lysine requirement i.e., the requirement for maximum gain is lower than that for breast meat yield, which is, in turn, lower than that for feed efficiency; and lastly the requirement for minimum abdominal fat percentage is the highest. On the other hand, other amino acids like threonine, valine, isoleucine and tryptophan, do not show differential requirement for gain vs. feed efficiency (Leclercq, 1998; Baker et al., 2002).

We studied the lysine requirement using the low-nutrient-density diets under heat-stress conditions. Thus, a direct comparison of the requirement established in this study with those reported by others is difficult; as dietary levels of ME and/or CP could affect the absolute requirement of amino acids (Gong et al., 2005). Most such studies used basal diets that contain around 3,200 kcal per kg ME or even higher. However, if we scale the lysine requirement as a proportion of ME, the body weight gain and feed efficiency seems to optimize at ratios of 3.63 g total lysine (or 3.15 g digestible lysine) per Mcal ME in Exp. 1, and 3.16 g total lysine (or 2.73 g digestible lysine) per Mcal ME in Exp. 2. Similar calculations based on lysine and ME requirements outlined by NRC (1994) revealed a corresponding figure of 3.44 g total lysine (or 3.10 g digestible lysine) per Mcal ME for 4-21 days of age, assuming 90% lysine digestibility in standard corn-soy based diets (Parsons, 1991). For the grower phase (21-42 days), NRC (1994) suggested a value of 3.13 g total lysine (or 2.82 g digestible lysine) per Mcal ME. It seems that for both phases, our results, expressed as digestible lysine per Mcal ME, were similar to the values listed by NRC (1994), even though our study was conducted under feeding and housing conditions which were very different from those studies that NRC (1994) was based on.

The digestible lysine requirement for 21-42 days of age for body weight gain as shown by Han and Baker (1994) was 0.85% and 0.78%, respectively, for male and female broilers. Average of both values (mixed sexes) was 0.82% digestible lysine. The test diets contained 3,200 kcal per kg ME, so the digestible lysine requirement was 2.56 g per Mcal ME, a value that was lower than our estimate of 2.73 g per Mcal ME for broiler chickens of 21-40 days of age. This difference could be partially explained by different regression techniques used to calculate the requirements. Han and Baker (1994) used broken-line regression which underestimates the requirement compared with quadratic regression analysis (Morris, 1989; Mack et al., 1999; Baker et al., 2002). It was also possible that the chronic high ambient temperature during the finishing period increased lysine requirement of the birds, as demonstrated by Corzo et al. (2003).

It seems from the above discussion that the idea of specifying lysine requirement of broilers as fixed ratios of lysine (more precisely digestible lysine) to ME may hold some promise. This concept would simplify broiler feed formulation technique, particularly for regions where nutrient densities of commercial feeds varied greatly from the "standard" (or NRC recommended) levels. Once the digestible lysine to ME ratios were established, specifications on all other essential amino acids could be deduced according to Ideal Protein Ratios (Baker and Han, 1994; Mack et al., 1999; Baker et al., 2002).

REFERENCES

Baker, D. H., A. B. Batal, T. M. Parr, N. R. Augspurger and C. M. Parsons. 2002. Ideal ratio (relative to lysine) of tryptophan, threonine, isoleucine and valine for chicks during the second and third weeks posthatch. Poult. Sci. 81:485-494.

Baker, D. H. and Y. Han. 1994. Ideal amino acid profile for broiler chicks during first three weeks post hatching. Poult. Sci. 73:1441-1447.

Byung-chul Park. 2006. Amino acid imbalance-biochemical and mechanism and nutritional aspect. Asian-Aust. J. Anim. Sci. 19(9):1361-1368.

Chung, T. K. and D. H. Baker. 1992. Apparent and true digestibility of a crystalline amino acid mixture and of casein comparison of values obtained with ileal-cannulated pigs and cecetomized cockerels. J. Anim. Sci. 70:3781-3790.

Corzo, A., E. T. Moran and D. Hoehler. 2003. Lysine needs of summer-reared male broilers form six to eight weeks of age. Poult. Sci. 82:1602-1607.

Gong, L. M., C. H. Lai, S. Y. Qiao, Defa Li, Y. X. Ma and Y. L. Lin. 2005. Growth performance, carcas cheracteristics, nutirent digestibility, and serum biochemical parameters of broilers fed low protien diets supplemented with various ratios of threonine to lysine. Asian-Aust. J. Anim. Sci. 18(8):1164-1170.

Han, Y. and D. H. Baker. 1994. Digestible lysine requirement of male and female broiler chicks during the period three to six weeks posthatching. Poult. Sci. 73:1739-1745.

Knowles, T. A. and L. L. Southern. 1998. The lysine requirement and ratio of total sulfur containing amino acids to lysine fir chicks fed adequate or inadequate lysine. Poult. Sci. 77:564-569.

Leclercq, B. 1998. Specific effect of lysine on broiler production: Comparison with threonine and valine. Poult. Sci. 77:118-123.

Mack, S., D. Bercovici, G. De Groote, B. Leclercq, M. Pack, J. B. Schutte and Van Cauwenberghe. 1999. Ideal amino acid profile and dietary lysine specifications for broiler chickens from 20 to 40 days of age. Br. Poult. Sci. 40:257-265.

Morris, T. R. 1989. The interpretation of response data from animal feeding trials. In: Recent Developments in Poultry Nutrition (Ed. D. J. A. Cole and W. Heresign). Butterworhts, UK.

National Research Council. 1994. Nutrient requirements of poultry. 9th rev. ed. National Academy Press, Washington, DC.

Parsons, C. M. 1991. Amino acid digestibility in feedstuffs for poultry: Feedstuff evaluation and requirements. Biokyowa Technical Review No. pp. 1:1-15.