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Evaluation of Digestible Lysine Needs for Male Broiler

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Abstract: This study was performed to evaluate the effect of feeding digestible lysine (Dig. Lys) levels in excess of NRC (1994) requirements in standard corn-soy rations from one to 42 d of age. A total of 960 one d old (Cobb 500) chicks were obtained from a commercial hatchery and randomly distributed in a randomized complete block design among 16 floor pens with four replicate pens/treatment. Diets were formulated to provide a minimum of 100% of NRC recommendation levels of amino acids other than Lys. Diets were fortified with L-lysine HCL to provide four levels of Lys (T1 = 100, T2 = 110, T3 = 120 and T4 = 130% of NRC recommendation). Birds received starter diet from 1-10 d, grower diet from 11-21 d and finisher diet from 22-42 d of age. Four pens of 60 male birds were fed one of the four experimental diets. There was a hierarchy of Lys requirements: the requirement for maximum gain was achieved when feeding T2 or T3, the requirement to maximize Breast Meat (BM) yield or Feed Conversion Ratio (FCR) was achieved when feeding T3; lastly, the requirement for percentage of minimum Abdominal Fat (AF) was achieved when feeding T4. Body weight, FCR and BM yield in birds responded quadratically while AF responded linearly to incremental Dig Lys levels. Thus, Lys requirement may be increased according to whatever criteria are desired.

Key words: Broiler, digestible lysine, body weight, breast yield, feed conversion

INTRODUCTION

Accurate estimates of Lys requirements of broilers are critical when trying to apply the "ideal protein" concept in formulating broiler diets (Han and Baker, 1994). In practical feeds the total sulfur amino acids, Lys and threonine are usually considered as the most limiting amino acids in practical feeding for growing chickens fed corn-soybean meal diets (Kidd, 2000). Lysine is considered as the second limiting amino acid and used as the reference to which all the other indispensable AAs are ratioed to achieve an ideal protein pattern (Samadi and Liebert, 2007). Baker (1997) reported that almost all of the Lys in the diet is used for protein accretion and that is the reason why it's considered as a reference amino acid.

The National Research Council (NRC, 1994) recommended that broilers receive 11.0, 10.0 and 8.5 g total Lys/kg of diet at 0-3, 3-6 and 6-8 wks of age, respectively, the recommendation of NRC (1994) is criticized to be too low. Modern broilers require higher dietary Lys concentrations to optimize performance and breast yield compared with the broilers of past years (Dozier *et al.*, 2009).

The body composition of broilers may be significantly affected by the amino acid Lys when administered at dietary levels higher than those required for maximum growth rate; this also results in an improved FCR. There is a hierarchy of requirements: the requirement for maximum gain is lower than that for BM yield, which in turn is lower than the requirement for FCR; however, the

requirement for percentage of minimum AF is the highest of all (Leclercq, 1998). Current research has focused on diets that exceed the NRC recommendation, with a specific interest in the changes effected by Dig Lys on the yield of BM (Corzo *et al.*, 2002; Kidd *et al.*, 2005; Cafe and Waldroup, 2006). Diet formulation on a digestible Lys basis should more consistently meet the animals' Lys requirements than those based on total Lys concentration.

The objective of the present study is to evaluate the response of male Cobb 500 strain to four levels of Dig Lys (100, 110, 120 and 130% of NRC) in standard corn-soy rations from 1-10, 11-21 and 22-42 d of age.

MATERIALS AND METHODS

Birds and management: A total of 960 one day old Cobb 500 broiler male chicks with an average body weight of 42.6 g were obtained from a commercial hatchery and randomly distributed among 16 floor pens with wood shavings with 60 chicks per pen. All chicks were vaccinated against Marek's disease, Newcastle and infectious bronchitis. Pen allocation in experiment was such that all pens had similar average the starting body weight. The birds were reared in a conventional poultry house with raised side windows. Each pen measured 0.09 m² floor space per broiler. The birds were maintained a 24 h light schedule.

Diets and experimental design: In order to test the effectiveness of Dig Lys in levels higher than that in NRC

(1994) on the performance of male broiler chicken, birds were divided into 4 groups: T1 = Control diet (NRC Lys level); T2 = T1+10% Lys; T3 = T1+20% Lys; T4 = T1+30% Lys, [starter diets had 9.9, 10.9, 11.88 and 12.87; grower diet had 9.0, 9.9, 10.8 and 11.7; finisher diet had 7.65, 8.42, 9.18 and 9.95 g/kg Dig Lys, respectively]. Total Lys values and digestibility coefficients were obtained from an established feedstuffs database (Rhone Poulenc Animal Nutrition, 1993; Jiang, 1999; Degussa, 2005) for ingredients used in the present study. Digestibility coefficients were applied to the calculated total Lys values to obtain calculated Lys digestibility of feedstuffs for formulation purposes. Graduations of dietary Lys were achieved through the addition of L-lysine HCl at the expense of cornstarch. Isocaloric and isonitrogenous corn-soybean meal diets were formulated; isonitrogenous diets were achieved through the substitution of corn for soybean meal. Use of corn gluten meal in grower and finisher periods allowed for reduction in Lys content so it can match the NRC requirements for both periods (Table 1). The experimental diets were formulated to meet NRC (1994) nutrient requirements for broiler, in particular the recommendations for Cobb 500 strain. Feed and water were provided *ad libitum*. Crude protein was analyzed according to the procedures established by the Association of Official Analytical Chemists (AOAC, 1990) for corn, soybean meal and corn gluten meal and values were adjusted in the nutrient matrix before formulating the diets.

Measurements: Body weight and feed consumption were recorded at the end of each period by pen and feed conversion was computed at 10, 22 and 42 d of age. Cumulative feed intake and body weight were measured at the end of the experiment. At 42 d of age, two males per pen were selected for processing (8 birds/treatment). Birds consumed water before processing, but feed was withdrawn 10 hr before processing. After euthanasia, feather, heads, neck and shanks were removed and the remaining carcasses were dissected and were weighed. The percentages of eviscerated was calculated on the basis of live weight while the breast and abdominal fat were calculated on the basis of dressed weight. Then breast, thigh and drumstick were deboned and total meat percentage was calculated.

Statistical analysis: Treatments were distributed following randomized complete block design, in which each experimental diet was fed to 4 replicate pens. The experimental unit was the pen mean. Data were analyzed by analysis of variance procedures appropriate for a randomized complete block design, using General Linear Model procedure of the Statistical Analysis System (SAS, 2002-2003). When significant differences among treatments were found, means were separated using LSD test and orthogonal contrasts (Kuehl, 2000). Statistical significance was assessed at ($p < 0.05$).

Table 1: Composition of control diet fed to broilers from 1-42 days

Ingredients (g/kg)	Starter*	Grower*	Finisher*
Corn	596.60	621.70	655.20
Soybean meal (48% CP)	334.00	291.00	225.00
Corn gluten meal (60% CP)	0.00	15.00	46.00
Soy oil	20.00	28.00	32.00
Salt	3.00	3.00	3.00
Limestone	6.20	5.40	6.00
Di Calcium phosphate	25.50	24.50	23.00
DL-methionine	2.30	1.00	0.20
Cornstarch	5.00	5.00	5.00
Vitamin-mineral premix ¹	3.00	3.00	3.00
Sodium bicarbonate	2.90	1.50	1.00
Amprol	0.50	0.50	0.00
Choline chloride	1.00	0.50	0.50
Calculated analysis			
ME (kcal/kg)	3000.00	3100.00	3200.00
CP (%)	21.00	20.00	19.00
Dig. Lysine ² (%)	0.99	0.90	0.77
Methionine + Cystine (%)	0.90	0.75	0.67
Methionine (%)	0.54	0.40	0.34
Calcium (%)	1.00	0.95	0.90
Phosphorus, nonphytate (%)	0.50	0.48	0.45

*Time of feeding the diets was 1-10 d for starter, 11-22 for grower and 23-42 d for finisher.

¹Vitamin-mineral mix provide per kg of diet: Vitamin A, 17500 IU; Cholecalciferol, 5000 IU; Vitamin E, 25 IU; Vitamin B₁₂, 0.03 mg; Riboflavin, 15 mg; Niacin, 75 mg; D-pantothenic acid, 25 mg; Choline, 705.5 mg; Menadione, 5 mg; Folic acid, 1.5 mg; Pyridoxine, 6.25 mg; Thiamine, 3.03 mg; D-biotin, 0.127 mg. Manganese, 120 mg; Zinc, 100 mg; Copper, 10 mg; Iodine, 2.5 mg; Calcium, 135 mg; Iron, 75 mg; Selenium, 0.15 mg.

²Diets were fortified with L-lysine HCl to provide four levels of Lys (control, 110, 120 and 130% of the control) for each period. Dietary lysine levels were achieved through the addition of L-lysine HCl at the expense of cornstarch

RESULTS

Feed consumption and feed efficiency: Weight gain, feed intake and FCR for each period are shown in Table 2. At 10 d of age, significant differences in body weight and FCR were observed ($p < 0.001$); birds received T2 gained more weight compared to control diet with no further increase at T3 or T4. No significant difference in feed intake was observed between treatments. Birds that received the T2 or T3 had a better FCR compared to T1. At 22 d, birds received T2 gained more body weight compared to those received T4 or T1 and similar gain compared to those received T3 ($p < 0.001$). Moreover, feed intake was higher in birds received T2 or T4 compared to those received T1 or T3 ($p < 0.01$). FCR at this period was highest for birds received T1 followed by those received T4 then T2, the best FCR was achieved in birds received T3 ($p < 0.001$). At 42 d of age, significant differences in body weight gain and FCR were observed ($p < 0.01$, 0.001, respectively); Birds received T2, gained more weight compared to those received T1 or T4, similar gain was observed between T2 and T3; T3 and T4; T1 and T4. Lys level had no significant effect on feed intake at this period ($p > 0.05$). In addition, Lys improved FCR in birds received T3 followed by T2 then T4, with the worst FCR for birds received T1 ($p < 0.001$).

Table 2: Weight gain, Feed Intake (FI) and Feed Conversion Ratio (FCR) of broiler chickens given experimental diets at 10, 22 and 42d

	Treatment				SEM	p*
	T1	T2	T3	T4		
Performance at 10 d						
Gain (g)	251 ^c	287 ^a	283 ^{ab}	278 ^b	±2.46	***
FI (g)	257	266	262	265	±2.17	NS
FCR (g:g)	1.024 ^a	0.924 ^c	0.926 ^c	0.952 ^b	±0.006	***
Performance at 22 d						
Gain (g)	623 ^c	678 ^a	671 ^{ab}	671 ^b	±5.01	***
FI (g)	924 ^b	948 ^a	915 ^b	953 ^a	±7.24	**
FCR (g:g)	1.482 ^a	1.398 ^c	1.363 ^d	1.449 ^b	±0.008	***
Performance at 42 d						
Gain (g)	1891 ^c	2010 ^a	1995 ^{ab}	1942 ^{bc}	±18.9	**
FI (g)	3462	3528	3445	3510	±24.5	NS
FCR (g:g)	1.831 ^a	1.755 ^c	1.727 ^d	1.808 ^b	±0.008	***

^{abcd}Means in the row with different superscripts differ significantly (*p<0.05, **p<0.01, ***p<0.001, NS: Not significant). SEM = Standard Error of the Mean

Table 3: Cumulative live weight (BW), feed intake, Feed Conversion Ratio (FC) and mortality of broiler chickens given experimental diets from 1-42 d

	Treatment						Regression		
	T1	T2	T3	T4	SEM	p*	Linear	Quad.	Cubic
Cumulative performance (1-42 d)									
BW (g)	2523 ^c	2667 ^a	2666 ^a	2603 ^b	±16.9	***	*	***	NS
Feed (g)	4644 ^b	4741 ^a	4622 ^b	4727 ^a	±25.5	*	NS	NS	**
FC (g:g)	1.847 ^a	1.764 ^c	1.733 ^d	1.819 ^b	±0.005	***	**	***	*
Mortality	2.15	2.13	2.08	2.00	±0.074	NS	NS	NS	NS

^{abcd}Means in the row with different superscripts differ significantly (*p<0.05, **p<0.01, ***p<0.001, NS: Not significant). SEM = Standard Error of the Mean

Table 4: Effect of different treatments on parts yield as percentages of broiler dressed weight at 42 d

	Treatment						Regression		
	T1	T2	T3	T4	SEM	p*	Linear	Quad.	Cubic
	(%)								
Eviscerated ¹	70.43	70.49	70.5	70.51	±0.26	NS	NS	NS	NS
Breast	20.87 ^d	21.21 ^b	21.41 ^a	21.04 ^c	±0.053	***	*	***	NS
Total meat ²	35.2 ^c	35.67 ^a	35.82 ^a	35.43 ^b	±0.07	***	*	***	NS
Abdom. fat	2.51 ^a	2.48 ^{ab}	2.44 ^b	2.36 ^c	±0.011	***	***	*	NS

¹Eviscerated % = eviscerated carcass, without neck, abdominal fat and internal organs, as percentage of live weight.

²Total meat = sum of breast meat, thigh and drumstick deboned without skin, as percentage of live weight.

^{abcd}Means in the row with different superscripts differ significantly (*p<0.05, **p<0.01, ***p<0.001, NS: Not significant). SEM = Standard Error of the mean

Cumulative body weight, feed intake and FCR from 1-42 d of age are shown in Table 3. A quadratic effect of Dig Lys levels on cumulative body weight was observed in this experiment (p<0.001). Birds received T2 or T3 had higher body weight compared to those fed T4 (p<0.001), while the lowest body weight was obtained from birds fed T1. Birds responded cubically (p<0.001) to incremental Dig Lys level. Cumulative feed intake was significantly different between treatments (p<0.01), birds fed T1 and T4 had a similar feed intake, they consumed more feed compared to T1 or T4. A significant quadratic effect was observed for cumulative FCR (p<0.001). Cumulative FCR was significantly affected by Lys level (p<0.0001) such that birds fed T3 had the best FCR followed by T2, T4 and T1. Mortality was not affected by treatment in this experiment.

Carcass characteristics: The mean percentage of carcass characteristics is documented in Table 4. Eviscerated percentage was not affected by dietary treatment. A quadratic response of Dig Lys was observed for BM yield. A higher percentage of BM yield was obtained from chicken that were fed T3 diet compared to all other treatments (p<0.001) while the lowest yields were obtained from birds fed T1. A quadratic trend was observed for total meat as a response of Dig Lys. Total meat percentage was significantly higher for birds who received T2 or T3 compared to T1 or T4 (p<0.001). A linear trend was observed for AF as a result of Lys levels. Abdominal fat was significantly higher for birds received T1 compared to all other treatments (p<0.001); while birds fed T4 had the lowest AF percent.

DISCUSSION

The results revealed a significant improvement in FCR at 10, 22 and 42 d of age for birds fed higher Dig Lys than that recommended by NRC (1994) up to 120% and no further improvement in FCR was obtained by increasing Dig Lys level to 130% of the NRC, suggesting a negative effect when feeding such a high level of Lys. Similarly, Latshaw (1993) reported that chicks given 13.9 or 14.4 g Lys/kg diet from 0-21 d of age were negatively affected by these levels. Based on the results obtained from this experiment for the cumulative FCR, it is recommended to have 10.9, 10.8 and 9.18 g Dig Lys/kg diet for starter, grower and finisher to optimize cumulative FCR. The result agrees with Cafe and Waldroup (2006) who found that 110 and 120% Lys level of the diet improved FCR for Cobb strain at 16 and 42 d of age, respectively. Han and Baker (1991) determined requirements of 8-21 day old Hubbard x Hubbard strain to be not greater than 12.1 g Dig Lys/kg of the diet for maximal FCR which was higher than that reported in this experiment. Kidd *et al.* (1997) reported that increasing dietary lys from 11.0-12.0 g/kg from 1-18 d in Ross x Ross broilers improved FCR and increasing Lys to 105% of NRC was needed to optimize the 18-54 d FCR. Labadan *et al.* (2001) also determined that Lys requirements for (Ross x Avian) strain broilers up to 2 weeks of age to be 12.1 g/kg diet. However, Dastar *et al.* (2005) reported a lower Dig Lys requirement (10.4 g/kg) of Ross male broiler based on broken-line model from 6-16 d. Rostagno *et al.* (2007) reported that the Dig Lys requirements based on FCR for Cobb 500 male broilers as 11.6 and 10.4 g/kg (front 10-21 and 22-35 d of age, respectively). Recently, Berri *et al.* (2008) reported a significant FCR in male Ross 308 by increasing Dig Lys from 8.3-9.3 g/kg between 21 and 42 d of age. The improvement in FCR in this experiment could be explained by the improvement in body weight gain at 10, 22 and 42 d of age and cumulative body weight from 1-42 d of age for birds received 110 or 120% of the NRC. The positive effect in FCR by feeding 120% Lys from 23-42 d of age occurred as a result in improvement in body weight gain rather than feed intake since feed intake was not significant at this period. This result agrees with finding of Corzo *et al.* (2002) and Berri *et al.* (2008). Based on the results obtained by this experiment it is recommended to have no more than 10.9, 9.9 and 8.42 g Dig Lys/kg diet for the starter, grower and finisher diets, respectively to optimize body weight. Lys increased feed intake at 110 and 130% but not at 120%. These findings suggested that improved body weight obtained with digestible Lys addition may be the result of high digestibility and more availability of synthetic lys at 120% of the NRC. The results of this experiment showed clearly that the requirement for cumulative body gain (110% of the NRC) was less than that for cumulative FCR (120% of the NRC). Same effect of Lys on FCR and

body weight has been observed by a number of authors. In another experiment, Han and Baker (1994) reported that the requirement for FCR from three to six weeks of age was 11.55 g Dig Lys/kg of feed, which was higher than the requirement for gain (10.14 g/kg). Similarly, Leclercq (1998) demonstrated that the requirement for FCR was always higher than that for gain, the broken-line model resulted in 9.24 g total Lys/kg of feed for gain and 10.10 for FCR; in the monomolecular model the requirements were 9.69 and 11.84 g/kg, respectively. Zaghari *et al.* (2002) reported that Dig Lys requirement for maximum body weight gain from 6-21 d of age was 10.75 and 10.49 g/kg for males and females, respectively. The Lys requirement for maximum FCR was 11.79 and 11.49 g/kg for males and females, respectively.

Breast muscle provides the greatest portion of edible meat in broilers (Acar *et al.*, 1993) and the contribution of BM to total carcass meat is extensive (Halvorson and Jacobsen, 1970). Changes induced by Lys can be considered meaningful in terms of carcass quality and consumer preferences (Moran and Bilgili, 1990). The level of digestible Lys needed in this experiment to optimize BM yield and total meat was 120% of the NRC requirements (11.88, 10.8 and 9.18 g digestible Lys/kg diet for starter, grower and finisher diets, respectively) which was higher than that needed for optimal body weight and similar to that needed to maximize FCR. The Dig Lys requirements to optimize BM yield at 6 wks old was close to the values of 8.7 g/kg (Han and Baker, 1994), 9.2 g/kg (Labadan *et al.*, 2001) and 9.3 g/kg (Berri *et al.*, 2008). Lysine is known to exhibit specific effects on carcass composition of broilers; it increases protein synthesis and decreases protein degradation in chicks (Kidd and Fancher, 2001). Muscle protein is high in Lys, which accounts for the higher Lys requirement when BM yield is used as the basis for setting the lys requirement as compared to weight gain (Hickling *et al.*, 1990; Moran and Bilgili, 1990; Schutte and Pack, 1996; Leclercq, 1998).

Several authors have shown that Lys supplementation at higher levels results in specific and significant effects on body composition, especially in the case of BM yield (Sibbald and Wolynetz, 1986; Hickling *et al.*, 1990; Moran and Bilgili, 1990; Han and Baker, 1993, 1994; Holsheimer and Ruesink, 1998; Quentin *et al.*, 2005). However, Lys can also modify fat deposition, as demonstrated in this experiment; the Dig Lys requirement to minimize AF was 130% of the NRC requirement (12.87, 11.7 and 9.95 g Dig Lys/kg diet for starter, grower and finisher diets, respectively) which was higher than that needed to maximize body weight, BM yield and FCR. Deposits of fat in the abdominal region are considered a waste by the poultry industry, abdominal fat is not only a loss, but also it represents an added expense for the processing effluent treatment.

The result of this study is also supported by the findings of Ojano-Dirain and Waldroup (2002) who found that increasing the dietary Lys levels from 10.03-11.2 g/kg significantly improved BM and reduced AF in broilers from 3-6 wk of age reared under moderate heat stress. They also reported that BM yield *Pectoralis major* and *Pectoralis minor* increased linearly in response to increased dietary Lys levels while AF decreased linearly. Cafe and Waldroup (2006) found that at 35 d of age, BM yield of birds fed 120% of NRC (1994) Lys was significantly greater than that of birds fed 100%. At 42 d of age, there was a significant reduction in AF in birds fed 120% of NRC (1994) lysine as compared to those fed 100%. Tesseraud *et al.* (2009) examined the effect of feeding birds with alternations of diets varying in Lys content on the expression of genes related to proteolysis in chicken *pectoralis*. They suggested that timing-sensitive regulation of proteolysis in chicken *pectoralis* according to Lys level and a high metabolism capacity to compensate for changes in Lys supply.

Conclusion: The effect of Lys on performance and yields of broiler chickens indicate that increasing Lys intake above the level required for maximum live weight gain causes significant changes in body composition by increasing BM yield and decreasing percentage of AF. These effects, as well as Lys's general effect on growth rate, have a beneficial effect on the FCR. Thus, Lys requirement may be increased according to whatever criteria are desired: the requirement for maximum gain is lower than that for BM yield or FCR, which in turn is lower than the Lys requirement for minimum percentage of AF. An existing matrix of total Lys requirements should be converted to Dig Lys rather than total Lys. Diet formulation on a Dig Lys basis should be more consistently meet the animals' Lys requirements than those based on total Lys concentration.

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