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Reevaluation of the Digestible Lysine Requirement of Arian Male Broiler Chicks by Different Diets with Cottonseed Meal

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ABSTRACT

An experiment was conducted to reevaluate the digestible lysine requirement of Arian male broilers by comparing the performance of chicks fed different levels of cottonseed meal (CSM) on a total amino acid (AA) versus a digestible AA basis. Four hundred and thirty two (432) Arian male broiler chicks were allotted to 18 treatments with four replicates of six chicks each in a completely randomized block design in a factorial arrangement with two factors (two levels of digestible lysine × nine different diets). The first of the eighteen treatments was the corn and soybean meal control diet based on previously determined digestible lysine requirement for maximum body weight gain. Diets 2 to 5 contained 5, 10, 15 or 20% CSM, respectively and were formulated to contain levels of total AA equivalent to those in the first treatment. Diets 6 to 9 contained the same levels of CSM as diets 2 to 5 but the diets were formulated to be equal in digestible AA content to the first treatment. Treatments 10 to 18 were the same as treatments 1 to 9, but on the basis of determined digestible lysine requirement for maximum feed efficiency. Formulating diets containing CSM on a digestible AA basis resulted in an improvement in chick performance compared to those formulated on a total AA basis (P<0.01). The lessening of chick performance with diets formulated on a total AA basis compared to a digestible AA basis, indicated that the previously determined digestible lysine requirement for the Arian broiler strain may be correct.

Keywords: Broiler, Corn-soybean meal, Cottonseed meal, Digestible amino acid, Total amino acid.

INTRODUCTION

The results of previous experiments by the authors indicated that the digestible lysine requirements of Arian male broilers to achieve maximum body weight gain and feed efficiency in the starter period were 1.075 and 1.179 (percentage of diet) respectively (Zaghari *et al*., 2002). These results indicated that the lysine requirement of Arian male broilers for maximum body weight gain and feed efficiency were higher than those of values reported for other strains (Han and Baker, 1991 and 1993; Baker, 1994). Nevertheless accuracy of estimates is critical for applying data in practice to broiler diet formulation. Therefore CSM was used as an alternative feed ingredient with a low digestibility of AA for practical evaluation of the determined digestible lysine requirements of Arian male broilers by comparing the performance of chicks fed diets formulated on a total AA versus a digestible AA basis. The low digestibility of AA in CSM, particularly lysine, has several implications (NRC, 1994). Lysine can become unavailable because of binding to gossypol, a polyphenolic compound that is located inside the pigment glands of cotton seeds (Ryan *et al*., 1986). This compound can form a Schiff's base with the epsilon amino

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group of lysine and render it undigestible. It has been shown that the adverse effects of gossypol can be alleviated by adding $FeSo_4$ to CSM in a 2:1 ratio of iron weight to free gossypol to form a harmless iron gossypol complex (Clawson and Smith, 1966). In addition, processing CSM involves much heat and pressure to break the seed and extract as much oil as possible. Thus there is the possibility of forming maillard compounds (Mauron, 1981) between the reducing sugars in CSM and AA, particularly lysine. The problem of low digestibility of AA should be resolved by obtaining accurate values in the dietary formulation to compensate for deficiencies in CSM.

The primary objective of this research was to evaluate the determined digestible lysine requirement of Arian male broilers for maximum body weight gain and feed efficiency in the starter period using CSM diets formulated on a total AA *vs* a digestible AA basis.

MATERIALS AND METHODS

Ingredient Analyses

The corn and soybean meal (SBM) used in the experimental diets were from the same batch as previous experiments (Zaghari *et al*., 2002). Corn, SBM and CSM were first analyzed for dry matter (DM), crude protein (CP), ether extract (EE) and crude fiber (CF). After proximate analysis, the feed ingredients were analyzed for amino acids (except tryptophan). The amino acid concentrations of three feedstuffs (Table 1) except for total sulfur AA were determined after acid hydrolysis, whereas total sulfur amino acid content was determined after performic acid oxidation and acid hydrolysis (AOAC, 1990) followed by ion-exchange chromatography. Analysis of the free gossypol in the used CSM was conducted elsewhere using high performance liquid chromatography (Fernandez *et al*., 1995).

Chick Assays

One day-old commercial male broiler chicks (Arian strain) were individually weighed and wing banded and were assigned to diets and battery cages. The initial body weight and weight range was 39.5±1(gr). The chicks were housed in thermostatically controlled batteries with raised wire floors from 0 to 21 days old. Feed and water were supplied *ad libitum* and uniform light was provided 24 hours per day. Diets were formulated on the basis of the analyzed values. Digestible amino acids were calculated using the digestibility coefficients of Rhone-Poulenc (1993) and by assuming 100% digestibility of crystalline amino acids (Chung and Baker, 1992). The apparent metabolizable energy value of

Table 1. Analysis of crude protein and amino acid composition of feed ingredients, as fed basis.

Amino acid	Corn	Cottonseed meal	Soybean meal
		--------------------- % ----------------------	
Crude protein	8.16	25.45	46.31
Lysine	0.25	1.08	2.82
Methionine	0.16	0.47	0.63
Cystine	0.18	0.39	0.74
Threonine	0.3	0.88	1.82
Arginine	0.39	3.41	3.52
Isoleucine	0.27	0.87	2.15
Leucine	1.03	1.45	3.64
Valine	0.37	1.20	2.24
Histidine	0.24	0.69	1.30
Phenylalanine	0.41	1.22	2.47

CSM corrected for zero nitrogen retention (ME_n) was equal to 3333.69 Kcal/Kg which was calculated by using the following equation: $ME_n = 21.26DM + 47.13EE - 30.85CF$ (NRC, 1994). To avoid any adverse effect from free gossypol in CSM, $FeSo_4.7H_2o$ was added to CSM to achieve a 2:1 ratio of iron weight to free gossypol before mixing the experimental diets (Husby and Kroening, 1971), an amount of $FeSo_4.7H_2o$ equivalent to the average amount of iron, added to the control corn-SBM diet. On day 21, all chicks were weighed individually and their pen feed intake was measured. Growth performance was evaluated by average body weight gain (ABWG), average daily feed intake (ADFI) and gain: feed (G:F) ratio.

Experimental Design

In this experiment, 432 male chicks were randomly allotted to 18 treatments of four replicates of six chicks per replicate based on a completely randomized block design in a factorial arrangement with two factors (two levels of digestible lysine × nine different diets). Battery cages consisted four decks, and each deck was identified as a block. The first of the eighteen treatments was the corn-SBM control diet based on previously determined digestible lysine requirements for maximum body weight gain (Zaghari *et al*., 2002). Diets 2 to 5 contained 5, 10, 15 or 20% CSM, respectively, and were formulated to contain levels of total AA equivalent to those in the first treatment. Diets 6 to 9 contained the same levels of CSM as diets 2 to 5 but the diets were formulated to be equal in digestible AA content to the first treatment (Table 2). Treatments 10 to 18 were similar to treatments 1 to 9 but on the basis of determined digestible lysine requirement for maximum feed efficiency (Zaghari *et al*., 2002) (Table 3). The control diet met an ideal amino acid ratio as recommended by Illinois (Baker, 1994) for all other indispensable amino acids for chicks from 0 to 21 days old as determined for the levels of 1.075 and 1.179% digestible lysine.

Statistical Analysis

Data were subjected to ANOVA procedures appropriate for completely randomized block designs (Steel and Torrie,1980) and analyzed as a 2 × 9 factorial, with main effects of digestible lysine requirement basis (body weight gain *vs* feed efficiency) and diet (1 to 9), using algorithms generated by the SAS Institute (1985). Differences between individual treatment means were assessed using the Duncan multiple range test (Steel and Torrie, 1980).

RESULTS

Dietary inclusion of 10 to 20% CSM on a total AA and weight gain lysine requirement basis depressed chick weight gain and feed efficiency ($P<0.01$) in comparison to the corn-SBM control diet (Table 4). However, when the diets were formulated according to the requirement for maximum feed efficiency, the dietary inclusion of 20% CSM on a total AA basis depressed chick weight gain and feed efficiency in comparison to the corn-SBM control diet ($P<0.01$). In both cases, formulating diets containing CSM on a digestible AA basis resulted in improved chick performance compared to those achieved from formulation on a total AA basis ($P<0.01$). No significant effect was observed on average daily feed intake ($P>0.05$).

Dietary formulation on the basis of lysine requirement for maximum feed efficiency *vs* the maximum weight gain had no effect on average body weight gain and feed efficiency ($P>0.05$) but resulted numerically in an 0.8% improvement in feed efficiency. No significant interaction was observed between these two factors and chick performance.

DISCUSSION

The results of the present study further confirmed an earlier study by Fernandez *et al*. (1995) that formulation of diets contain-

Table 2. Composition of experimental diets containing cottonseed meal, according to the digestible lysine requirement for maximum body weight gain.

Ingredients and analysis	Control diet	Total AA basis				Digestible AA basis			
	Treatments								
	1	2	3	4	5	6	7	8	9
Corn	51.022	48.939	46.827	44.716	42.624	48.979	46.937	44.866	42.814
Soybean meal	37.27	34.85	32.41	29.97	27.51	34.76	32.25	29.74	27.23
Cottonseed meal	0	5	10	15	20	5	10	15	20
Corn oil	7.40	6.91	6.43	5.94	5.44	6.89	6.39	5.89	5.39
Oyster shell	1.56	1.57	1.57	1.58	1.59	1.57	1.58	1.58	1.59
Monocalcium phosphate	1.54	1.53	1.51	1.50	1.48	1.53	1.51	1.50	1.49
NaCl	0.47	0.46	0.46	0.46	0.45	0.46	0.46	0.46	0.45
Vitamin premix [a]	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
Mineral premix [b]	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
$FeSo_4.7H_2o$	0.028	0.011	0.023	0.034	0.046	0.011	0.023	0.034	0.046
DL-Met	0.18	0.17	0.17	0.17	0.17	0.19	0.19	0.2	0.21
L.Lysine	0.04	0.06	0.09	0.11	0.14	0.09	0.13	0.18	0.22
L.Thr			0.01	0.02	0.03	0.02	0.03	0.05	0.06
L.Ile					0.02				
Analysis									
ME_n Kcal/Kg	3200	3200	3200	3200	3200	3200	3200	3200	3200
CP %	21.57	21.57	21.57	21.57	21.57	21.57	21.57	21.57	21.57
Na %	0.2	0.2	0.2	0.2	0.2	0.2	0.2	0.2	0.2
Calcium %	1	1	1	1	1	1	1	1	1
Available phosphorous %	0.45	0.45	0.45	0.45	0.45	0.45	0.45	0.45	0.45
Crud fiber	3.7	4.7	5.7	6.8	7.8	4.7	5.7	6.8	7.8
Total Lys %	1.21	1.21	1.21	1.21	1.21	1.22	1.23	1.25	1.26
Total Met+Cys %	0.86	0.86	0.86	0.86	0.86	0.87	0.88	0.89	0.89
Total Thr %	0.83	0.83	0.83	0.83	0.83	0.83	0.84	0.85	0.86
Total Trp %	0.26	0.26	0.26	0.26	0.26	0.26	0.26	0.26	0.26
Total Ile %	0.93	0.92	0.91	0.90	0.90	0.92	0.90	0.89	0.87
Total Leu %	1.88	1.84	1.80	1.77	1.73	1.84	1.80	1.76	1.72
Total Arg %	1.51	1.58	1.66	1.74	1.81	1.58	1.65	1.73	1.80
Total Val %	1.02	1.02	1.01	1.01	1.01	1.02	1.01	1.01	1.00
Total Phe+Tyr %	1.95	1.92	1.90	1.88	1.86	1.92	1.90	1.87	1.85
Total His %	0.60	0.60	0.60	0.60	0.59	0.60	0.60	0.59	0.59
Digestible Lys %	1.075	1.05	1.045	1.03	1.01	1.075	1.075	1.075	1.075
Digestible Met+Cys %	0.77	0.76	0.75	0.74	0.73	0.77	0.77	0.77	0.77
Digestible Thr %	0.72	0.71	0.70	0.69	0.68	0.72	0.72	0.72	0.72
Digestible Trp %	0.22	0.22	0.21	0.21	0.21	0.22	0.22	0.21	0.21
Digestible Ile %	0.84	0.82	0.80	0.78	0.77	0.82	0.79	0.77	0.75
Digestible Leu %	1.72	1.67	1.62	1.57	1.52	1.67	1.62	1.57	1.52
Digestible Arg %	1.41	1.47	1.53	1.59	1.65	1.47	1.53	1.58	1.64
Digestible Val %	0.90	0.89	0.87	0.86	0.85	0.88	0.87	0.86	0.85
Digestible Phe+Tyr %	1.79	1.76	1.72	1.69	1.66	1.75	1.72	1.68	1.65
Digestible His %	0.55	0.54	0.54	0.53	0.52	0.54	054	0.53	0.52
Free Gossypol %		0.0011	0.0023	0.0034	0.0046	0.0011	0.0023	0.0034	0.0046

[a] Vitamin premix provided the following per kilogram of diet: Vitamin A, 9000IU; Cholecalciferol, 2000IU; Vitamin E, 18IU; Vitamin k3, 4mg; Vitamin B12, 0.015mg; Biotin, 0.15mg; Folacin, 1mg; Niacin, 30mg; Pantothenic acid, 25mg; Pyridoxine, 2.9mg; Riboflavine, 6.6mg; Thiamine 1.8mg.
[b] Mineral premix provided the following per kilogram of diet: Copper(as cupric sulfate 5H20),10mg; Iodin (as calciumiodate), 0.99mg; Iron(as ferrous sulfate 7H20), 50mg; Manganese (as manganese oxide), 99mg; Selenium (as sodium selenite), 0.2mg; Zinc (as zinc oxid), 84mg.

ing 20% CSM on a true digestible AA basis was superior to formulation on a total AA basis.

The explanation of this observation was probably the lower AA digestibility of the CSM compared with corn or SBM. Thus the

Table 3. Composition of experimental diets containing cottonseed meal, according to the digestible lysine requirement for maximum feed efficiency.

Ingredients And Analysis	Control diet	Total AA basis				Digestible AA basis			
		treatments							
	10	11	12	13	14	15	16	17	18
Corn	50.302	48.219	46.117	44.026	41.954	48.329	46.286	44.286	42.304
Soybean meal	37.65	35.25	32.81	30.37	27.91	35.1	32.56	30.0	27.40
Cottonseed meal	0	5	10	15	20	5	10	15	20
Corn oil	7.47	6.98	6.49	5.99	5.49	6.95	6.43	5.9	5.36
Oyster shell	1.56	1.57	1.57	1.58	1.59	1.57	1.57	1.58	1.59
Monocalcium phosphate	1.54	1.52	1.51	1.49	1.48	1.52	1.51	1.50	1.49
NaCl	0.47	0.46	0.46	0.46	0.45	0.46	0.46	0.46	0.45
Vitamin premix	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
Mineral premix	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
$FeSo_4.7H_2o$	0.028	0.011	0.023	0.034	0.046	0.011	0.023	0.034	0.046
DL-Met	0.25	0.24	0.24	0.24	0.23	0.26	0.27	0.28	0.28
L.Lysine	0.17	0.18	0.20	0.23	0.25	0.21	0.26	0.30	0.35
L.Thr	0.07	0.07	0.08	0.08	0.09	0.08	0.10	0.11	0.13
L.Ile					0.01			0.01	0.04
L.Val						0.01	0.03	0.04	0.06
Analysis									
ME_n Kcal/Kg	3200	3200	3200	3200	3200	3200	3200	3200	3200
CP %	21.89	21.89	21.89	21.89	21.89	21.89	21.89	21.89	21.89
Na %	0.2	0.2	0.2	0.2	0.2	0.2	0.2	0.2	0.2
Calcium %	1	1	1	1	1	1	1	1	1
Available phosphorous %	0.45	0.45	0.45	0.45	0.45	0.45	0.45	0.45	0.45
Crud fiber	3.7	4.7	5.8	6.8	7.8	4.7	5.8	6.8	7.8
Total Lys %	1.31	1.31	1.31	1.31	1.31	1.33	1.34	1.35	1.36
Total Met+Cys %	0.93	0.93	0.93	0.93	0.93	0.94	0.95	0.96	0.97
Total Thr %	0.90	0.90	0.90	0.90	0.90	0.90	0.91	0.92	0.93
Total Trp %	0.26	0.26	0.26	0.26	0.26	0.26	0.26	0.26	0.26
Total Ile %	0.94	0.93	0.91	0.90	0.90	0.92	0.91	0.90	0.91
Total Leu %	1.88	1.85	1.80	1.77	1.74	1.84	1.80	1.76	1.72
Total Arg %	1.52	1.59	1.67	1.75	1.82	1.59	1.66	1.74	1.81
Total Val %	1.02	1.02	1.02	1.02	1.02	1.03	1.04	1.05	1.06
Total Phe+Tyr %	1.96	1.94	1.91	1.89	1.87	1.93	1.90	1.88	1.85
Total His %	0.61	0.60	0.60	0.60	0.60	0.60	0.06	0.60	0.59
Digestible Lys %	1.179	1.15	1.14	1.13	1.11	1.179	1.179	1.179	1.179
Digestible Met+Cys %	0.84	0.83	0.82	0.81	0.80	0.84	0.84	0.84	0.84
Digestible Thr %	0.78	0.78	0.77	0.76	0.75	0.78	0.78	0.78	0.78
Digestible Trp %	0.22	0.22	0.22	0.21	0.21	0.22	0.21	0.21	0.21
Digestible Ile %	0.85	0.83	0.80	0.78	0.77	082	0.80	0.78	0.78
Digestible Leu %	1.73	1.67	1.63	1.58	1.53	1.67	1.62	1.57	1.52
Digestible Arg %	1.42	1.48	1.54	1.60	1.66	1.48	1.53	1.59	1.64
Digestible Val %	0.90	0.89	0.88	0.87	0.86	0.90	0.90	0.90	0.90
Digestible Phe+Tyr %	1.80	1.77	1.73	1.70	1.67	1.76	1.73	1.69	1.65
Digestible His %	0.55	0.55	0.54	0.53	0.53	0.54	0.54	0.53	0.52
Free Gossypol %		0.0011	0.0023	0.0034	0.0046	0.0011	0.0023	0.0034	0.0046

digestible AA levels in the diet decreased as the level of CSM increased. For example dietary lysine digestibility decreased about 3.5% as the CSM level increased from 0 to 20%. Previous studies on several by-product ingredients (Rostagno *et al*., 1995 and Michele *et al*., 1999) have also shown that formulation of poultry diets on a digestible AA basis is superior to formulation on a total AA basis when using ingredients that have AA digestibilities lower than those in corn- and SBM- based diets.

The dietary inclusion of CSM up to 20% had no effect on feed intake. However, Fernandez *et al*. (1995) reported that higher levels of CSM in the diet decreased feed intake from between 30 to 40%. This decrease in feed intake might be due to the presence of some detrimental factors in

Table 4. The effects of different diets containing cottonseed meal formulated on a digestible and total AA basis, on the performance of Arian male broiler chicks.

Lysine requirement basis	CSM	Dietary formulation method	Body weight-gain/bird	Gain:Feed	Average daily feed intake/bird
	(%)		(g)	(g:g)	(g)
	0	Digestible AA	621.8[a]	0.721[ab]	40.6
	5	Digestible AA	634.8[a]	0.752[a]	39.0
	10	Digestible AA	621.0[a]	0.715[ab]	41.1
	15	Digestible AA	630.2[a]	0.714[ab]	40.0
Weight gain	20	Digestible AA	627.0[a]	0.729[ab]	40.8
	5	Total AA	633.7[a]	0.732[ab]	41.1
	10	Total AA	580.8[b]	0.695[bc]	39.7
	15	Total AA	578.6[b]	0.698[b]	38.0
	20	Total AA	573.6[b]	0.647[c]	42.1
	Mean		611.3	0.711	40.3
	SE		5.6	0.004	0.466
	P-Values		0.01	0.01	Ns
	0	Digestible AA	622.0[ab]	0.747[a]	38.3
	5	Digestible AA	640.1[a]	0.715[ab]	41.7
	10	Digestible AA	618.8[ab]	0.714[ab]	40.8
	15	Digestible AA	621.1[ab]	0.729[ab]	39.7
Feed efficiency	20	Digestible AA	619.5[ab]	0.725[ab]	40.0
	5	Total AA	616.6[ab]	0.716[ab]	40.9
	10	Total AA	624.8[ab]	0.722[ab]	41.1
	15	Total AA	595.0[bc]	0.701[ab]	40.0
	20	Total AA	576.1[c]	0.687[b]	39.8
	Mean		614.9	0.717	40.3
	SE		5.9	0.004	0.464
	P-Values		0.01	0.01	ns

Means within a column with different superscript differ significantly ($P<0.05$).
Ns: Not significant.

CSM, such as the high amount of fiber, gossypol, or cyclopropene fatty acids. However, these factors wouldn't be a problem in the present study because all diets were formulated to be isocaloric, iron sulfate was added to neutralize the gossypol and cyclopropene fatty acids should be low because they are almost totally eliminated from the cottonseeds during the oil extraction process (Phelps *et al*., 1965).

Under the conditions of this study, the depression of chick performance with diets formulated on a total AA basis compared to a digestible AA basis, indicated that the previously determined digestible lysine requirement for Arian chicks is correct (Zaghari *et al*., 2002). If the levels of AA in diets, especially lysine, were higher than the requirement, we would not observe any differences in chick performance between the two dietary formulation methods.

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ارزیابی مجدد لیزین قابل هضم مورد نیاز جوجه های گوشتی نر آرین توسط جیره‌های حاوی کنجاله تخم پنبه

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چکیده

در یک آزمایش با مقایسه عملکرد جوجه هایی که سطوح مختلف کنجاله تخم پنبه را مصرف نموده و جیره آنها بر اساس معیار آمینو اسیدهای کل و قابل هضم متوازن گردیده بود، نیاز لیزین قابل هضم جوجه گوشتی نر سویه آرین مورد ارزیابی مجدد قرار گرفت. در این پژوهش تعداد ۴۳۲ قطعه جوجه نر از سویه آرین به ۱۸ تیمار در ۴ تکرار و ۶ مشاهده در هر تکرار در قالب طرح بلوک کامل تصادفی بصورت فاکتوریل(۲ سطح لیزین قابل هضم×۹ جیره) اختصاص داده شد.نتایج نشان داد که افزودن کنجاله تخم پنبه به جیره هایی که آمینو اسید های محتوی آنها بر اساس آمینو اسید های کل و نیاز لیزین قابل هضم برای صفت وزن بدن و بازده غذایی متوازن شده بود موجب کاهش وزن بدن و بازده غذایی در مقایسه با جیره کنترل شد(P< ۰/۰۱). صرف نظر از صفت مورد نظر در تامین لیزین مورد نیاز، متوازن نمودن آمینو اسیدهای خوراک بر اساس آمینواسیدهای قابل هضم نسبت به آمینو اسیدهای کل موجب بهبود عملکرد جوجه ها شد(P< ۰/۰۱). بهبود عملکرد جوجه ها با جیره هایی که آمینواسیدهای محتوی آ نها بر اساس آمینو اسیدهای قابل هضم نسبت به آمینواسیدهای کل متوازن شده بود حاکی از صحت بر آورد نیاز لیزین قابل هضم این سویه در آزمایشهای قبلی بود.