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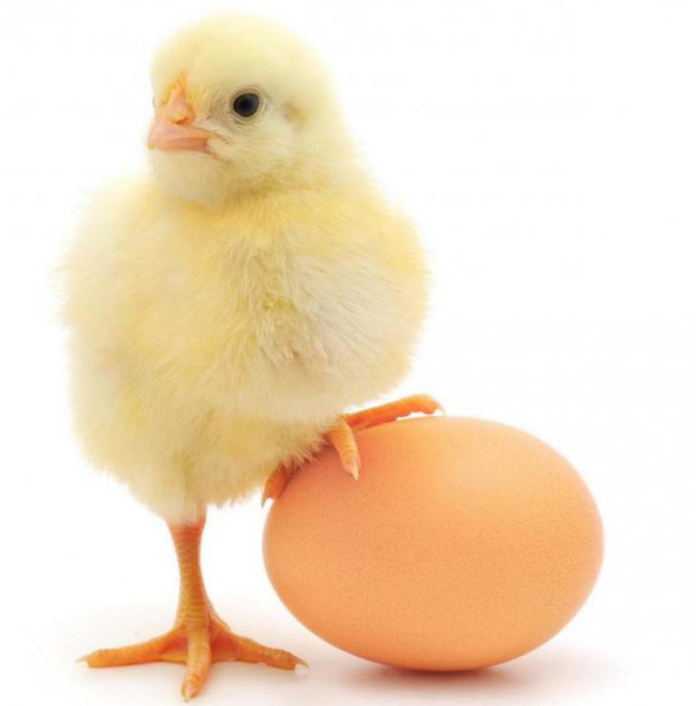

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Extent to Which Crude Protein May Be Reduced in Corn-soybean Meal Broiler Diets Through Amino Acid Supplementation[1]

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Abstract: This study was conducted to explore the extent to which CP can be reduced in corn-soybean meal broiler starter diets by amino acid supplementation while maintaining adequate performance. Using corn and soybean meal of known composition, diets were formulated by linear programming using a minimum of 107.5% of NRC (1994) amino acid recommendations. No minimum CP was required; as each amino acid became limiting crystalline sources were provided to meet the minimum specification. In sequence these were Met, Thr, Lys, Val, Ile, Arg, Phe, and Trp, resulting in eight treatments ranging from 16.61 to 22.48% CP. All diets contained 0.3% sodium bicarbonate and 0.2% aluminum hydroxide as a buffer and antacid. Three additional treatments utilized potassium sulfate to maintain a minimum dietary electrolyte balance (Na + K - Cl) of 250 meq/kg. Each treatment was fed to twelve replicate groups of six male broiler chicks from 1 to 21 d. Reducing CP below 20% while providing indispensable amino acids resulted in a significant reduction in body weight (BW) and feed conversion ratio (FCR). Crude protein content of freeze-dried carcasses declined and fat content increased as diet CP decreased. Feather content (actual weight or % of BW) was not affected until the CP was reduced to less than 18%. Maintaining dietary electrolyte balance at 250 meq/kg in reduced CP diets had no significant effect on any parameter.

Key words: Broilers, crude protein, nitrogen pollution, carcass content

Introduction

Over the course of time, amino acid supplements have been widely accepted for use in poultry industry. Beginning with the introduction of methionine in the 1950's, followed later by lysine and more recently threonine and tryptophan, nutritionists have incorporated these amino acids into poultry feeds to provide these essential amino acids. As a result, there has been a gradual reduction in the overall crude protein content of the diet. All indications point to further development of feed-grade amino acids that may result in a greater reduction in crude protein content.

One of the concerns facing the animal industry is that of waste disposal. Reduction of nitrogen and phosphorous in animal wastes will be one of the primary considerations in the diet formulation in the near future. This study was designed to explore the extent to which overall dietary crude protein may be reduced in a corn-soybean meal based poultry diet by amino acid supplementation while maintaining adequate performance.

Materials and Methods

Diet formulation: Diets were formulated based on several assumptions. First, we assumed that the NRC (1994) recommended amino acid levels were adequate. Secondly, we assumed that commercial nutritionists would add a reasonable margin of safety to these recommendations. We also assumed, based upon studies with crystalline amino acid diets, that diets high in supplemental amino acids would benefit from supplementation with sodium bicarbonate and aluminum hydroxide as a buffer and antacid (Dean and Scott, 1965). With that in mind, diets were formulated to meet a minimum of 107.5% of the NRC (1994) amino acid recommendations for the 0 to 21-d broiler. All diets were supplemented with 0.3% sodium bicarbonate and 0.2% aluminum hydroxide as a buffer and antacid. No CP minimum was specified; diets were formulated with amino acid supplementation offered in turn to provide the minimum specification as each amino acid became limiting. In sequence these were Met, Thr, Lys, Val, Ile, Arg, Phe, and Trp. This resulted in a total of eight dietary treatments (Table 1). Three additional treatments were added in which aliquots of diets F, G, and H were supplemented with potassium sulfate to increase the (Na + K) - Cl ratio to approximately 250 meq/kg as suggested by Mongin (1981). Each of these 11

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Table 1: Composition (g/kg) of diets with reduced crude protein levels

	CP (%) and 1st limiting amino acid of each diet							
	A	B	C	D	E	F	G	H
	22.48	21.18	20.93	20.55	20.32	18.10	18.06	16.61
	Met	Thr	Lys	Val	Ile	Arg	Phe	Trp
Ingredients (g/kg)								
Yellow corn	477.80	518.29	526.31	539.06	546.90	639.33	641.10	706.39
Soybean meal (47.5%)	401.27	365.48	358.17	346.43	339.15	250.58	248.87	185.06
Poultry oil	79.46	73.52	72.29	70.25	68.92	52.89	52.54	39.52
Dicalcium phosphate	17.56	17.81	17.86	17.95	18.00	18.66	18.67	19.15
Limestone	10.69	10.78	10.80	10.83	10.85	11.06	11.07	11.22
Sodium bicarbonate	3.00	3.00	3.00	3.00	3.00	3.00	3.00	3.00
Salt	2.52	2.51	2.51	2.51	2.51	2.51	2.51	2.51
Broiler vitamins[1]	2.00	2.00	2.00	2.00	2.00	2.00	2.00	2.00
Aluminum hydroxide	2.00	2.00	2.00	2.00	2.00	2.00	2.00	2.00
Trace minerals[2]	1.00	1.00	1.00	1.00	1.00	1.00	1.00	1.00
DL methionine	2.70	3.06	3.14	3.26	3.34	4.26	4.28	4.95
L-Threonine	0.00	0.55	0.67	0.85	0.97	2.36	2.38	3.39
L-Lysine Hcl	0.00	0.00	0.25	0.65	0.89	3.91	3.97	6.15
L-Valine	0.00	0.00	0.00	0.21	0.34	1.95	1.98	3.14
L-Isoleucine	0.00	0.00	0.00	0.00	0.13	1.74	1.77	2.94
L-Arginine	0.00	0.00	0.00	0.00	0.00	2.75	2.80	4.79
L-Phenylalanine	0.00	0.00	0.00	0.00	0.00	0.00	0.06	2.36
L-Tryptophan	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.43

[1]Provides per kg of diet: vitamin A (from vitamin A acetate) 7714 IU; cholecalciferol 2204 IU; vitamin E (from dl-alpha tocopheryl acetate) 16.53 IU; vitamin B_{12} 0.013mg; riboflavin 6.6 mg; niacin 39 mg; pantothenic acid 10 mg; choline 465 mg; menadione (from menadione dimethylpyrimidinol) 1.5 mg; folic acid 0.9 mg; thiamin (from thiamine mononitrate) 1.54 mg; pyridoxine (from pyridoxine hydrochloride) 2.76 mg; d-biotin 0.066 mg; ethoxyquin 125 mg; Se 0.1 mg. [2]Provides per kg of diet: Mn (from $MnSO_4 \cdot H_2O$) 100 mg; Zn (from $ZnSO_4 \cdot 7\,H_2O$) 100 mg; Fe (from $FeSO_4 \cdot 7\,H_2O$) 50 mg; Cu (from $CuSO_4 \cdot 5\,H_2O$) 10 mg; I (from $Ca\,(IO_3)_2 \cdot H_2O$) 1 mg.

treatments was fed to 12 replicate pens of six male chicks of a commercial broiler strain. Diets were pelleted and crumbled to enhance feed intake.

Chicks and Housing: Day-old male chicks of a commercial broiler strain[4] were obtained from a local hatchery where they had been vaccinated *in ovo* for Marek's virus and had received vaccinations for Newcastle Disease and Infectious Bronchitis post hatch via a coarse spray. They were randomly assigned to compartments in electrically heated brooders with raised wire floors. Six chicks were placed in each of 132 compartments. The test diets and tap water were provided for ad libitum consumption. Continuous 24 hr fluorescent lighting was provided.

Measurements: Birds were group weighed by pen at 1 and 21 d. Feed consumption during the period was determined by weighing the feed container at the start and end of the study. Mortality was checked twice daily; birds that died were weighed with the weight used to adjust feed conversion [FCR = total feed consumed ÷ (weight of live birds + weight of dead birds)]. At 18 d, feces were collected for one day on aluminum foil and placed in a freezer. The samples were then freeze-dried, ground using a 2 mm screen, and allowed to equilibrate to ambient temperature and humidity. Moisture and nitrogen content of the samples were then determined. Mixed diets were analyzed for crude protein, total amino acids, supplemental amino acids, sodium, chloride, and potassium content.

Samples of birds (two per pen) from each diet were subjected to an eight hr fast (water available during fast) and then processed. Weight of the birds was determined just prior to slaughter, after feather removal (to estimate weight of feathers), and after evisceration. Weight of viscera and abdominal fat pad was determined (total viscera weight; weight of heart, liver,

[4]Cobb 500. Cobb-Vantress, Inc., Siloam Springs AR 72761.

Table 2: Nutrient analysis of diets with reduced crude protein levels

	Dietary code and 1st limiting amino acid of each diet							
	A	B	C	D	E	F	G	H
Nutrient (%)	Met	Thr	Lys	Val	Ile	Arg	Phe	Trp
Crude protein (C)[1]	22.48	21.18	20.93	20.55	20.32	18.10	18.06	16.61
Crude Protein (A)[2]	23.70	22.60	23.40	19.70	21.50	18.60	18.20	15.60
Met (C)	0.61	0.63	0.63	0.64	0.64	0.69	0.68	0.72
Met (A)	0.58	0.60	0.59	0.64	0.62	0.68	0.66	0.71
Lys (C)	1.28	1.18	1.18	1.18	1.18	1.18	1.18	1.18
Lys (A)	1.29	1.16	1.28	1.19	1.17	1.19	1.14	1.15
Trp (C)	0.32	0.29	0.29	0.28	0.28	0.22	0.22	0.22
Trp (A)	0.30	0.29	0.30	0.27	0.27	0.21	0.21	0.20
Thr (C)	0.86	0.86	0.86	0.86	0.86	0.86	0.86	0.86
Thr (A)	0.89	0.90	0.95	0.93	0.93	0.89	0.87	0.88
Ile (C)	0.96	0.89	0.88	0.86	0.86	0.86	0.86	0.86
Ile (A)	0.99	0.97	1.01	0.94	0.94	0.92	0.89	0.88
His (C)	0.60	0.56	0.56	0.55	0.54	0.45	0.38	0.38
His (A)	0.60	0.57	0.61	0.58	0.56	0.47	0.46	0.39
Val (C)	1.04	0.98	0.97	0.97	0.98	0.97	0.97	0.97
Val (A)	1.14	1.14	1.14	1.10	1.09	1.10	1.04	1.04
Leu (C)	1.90	1.80	1.78	1.75	1.73	1.49	1.48	1.30
Leu (A)	1.90	1.77	1.92	1.78	1.77	1.58	1.47	1.31
Arg (C)	1.53	1.42	1.40	1.37	1.34	1.34	1.34	1.34
Arg (A)	1.12	1.41	1.63	1.41	1.41	1.35	1.33	1.30
Phe (C)	1.08	1.01	1.00	0.98	0.96	0.79	0.79	0.90
Phe (A)	1.13	1.05	1.16	1.08	1.07	0.88	0.83	0.94
Gly (C)	0.94	0.88	0.87	0.85	0.84	0.69	0.68	0.57
Gly (A)	0.95	0.92	0.96	0.92	0.90	0.74	0.70	0.60
Ser (C)	1.13	1.06	1.05	1.02	1.00	0.82	0.82	0.68
Ser (A)	1.13	1.08	1.13	1.07	1.07	0.89	0.85	0.71
Met + Cys (C)	0.97	0.97	0.97	0.97	0.97	0.97	0.97	0.97
Met + Cys (A)	0.92	0.93	0.93	0.97	0.94	0.97	0.93	0.95
Gly + Ser (C)	2.07	1.94	1.92	1.89	1.84	1.51	1.50	1.25
Gly + Ser (A)	2.08	2.00	2.09	1.99	1.97	1.63	1.55	1.31
Phe + Tyr (C)	1.98	1.85	1.83	1.78	1.76	1.44	1.44	1.44
Phe + Tyr (A)	1.90	1.77	1.95	1.79	1.78	1.43	1.38	1.40
Na + K - Cl, meq/kg	274.3	258.9	254.4	247.2	242.7	187.8	186.7	146.9

[1]Calculated value. [2]Analyzed value.

and gizzard; weight of small intestine). The defeathered, eviscerated carcass was frozen, ground, and sub-samples freeze-dried. The freeze-dried samples were analyzed for nitrogen and fat content.

Data analysis: Pen means served as the experimental unit. Data were subjected to analysis of variance (SAS Institute, 1991) as a single factor arrangement using the General Linear Models procedure. The means for treatments showing significant differences in the analysis of variance were compared using the least significant difference procedure. The data of fecal nitrogen content were subjected to regression analysis using the PROC REG function of SAS (SAS institute, 1991). All statements of significance are based on the 5% level of probability.

Results and Discussion

Analyzed values for CP and individual amino acids were in good agreement with calculated values (Table 2). The effect of reduced CP levels on live performance and carcass characteristics are summarized in Table 3. The CP level had a significant effect on body weight, feed conversion ratio, and carcass composition, but not on mortality. Reducing CP below 20% while providing recommended levels of indispensable amino acids resulted in a significant reduction in BW and increased FCR. Further growth retardation was observed when dietary CP was decreased lower than 18%. The FCR remained unchanged when dietary CP reduced from 18

Table 3: Effects of diets with reduced levels of crude protein on body weight, feed conversion ratio (FCR), mortality, and carcass content of male broiler chicks

Diet	Limiting Amino acid	Crude Protein (%)	21 d BW (g)	0-21 d FCR (g:g)	0-21 d mortality (%)	Carcass composition[1]	
						Crude protein (%)	Crude fat (%)
A	Met	22.48	779^{a}	1.304^{c}	0.00	60.3^{a}	28.6^{d}
B	Thr	21.18	760^{ab}	1.304^{c}	3.33	58.9^{ab}	30.6^{bcd}
C	Lys	20.93	747^{abc}	1.331^{c}	1.66	58.6^{ab}	31.4^{b}
D	Val	20.55	764^{ab}	1.297^{c}	3.33	58.4^{ab}	31.3^{bc}
E	Ile	20.32	724^{abc}	1.350^{c}	1.66	56.3^{bc}	31.4^{b}
F	Arg	18.10	710^{bc}	1.414^{b}	1.66	54.2^{c}	34.8^{a}
G	Phe	18.06	718^{bc}	1.416^{b}	0.00	53.1^{d}	36.2^{a}
H	Trp	16.61	637^{d}	1.446^{ab}	0.00	51.6^{d}	36.5^{a}
I	As F + K2	18.10	700^{c}	1.444^{ab}	1.66	52.4^{d}	36.0^{a}
J	As G + K	18.06	718^{bc}	1.416^{b}	1.66	53.5^{cd}	35.0^{a}
K	As H + K	16.61	6.24^{d}	1.487^{a}	1.66	50.4^{d}	38.8^{a}
Probability > F			0.0001	0.0001	0.91	0.0005	0.0001
CV			9.19	4.15	2.573	2.86	3.42

[1]Percent of freeze-dried carcass. [2]Potassium added to provide dietary electrolyte balance (Na + K) - Cl) of 250. Meq/kg. [3]CV of transformed means. [abcd]Means in column with unlike superscripts differ significantly ($P \leq 0.05$).

Table 4: Effect of diets with reduced crude protein levels on development of feathers and carcass components

Diet	Limiting Amino acid	Crude Protein (%)	Carcass components: Feathers		Small Intestine		Gizzard		Abdominal Fat pad	
			Weight (g)	% of BW	Weight (g)	% of BW	Weight (g)	% of BW	Weight (g)	% of BW
A	Met	22.48	75.3^{a}	9.67^{a}	41.8^{a}	5.48	21.8^{a}	2.82	6.7^{b}	0.79^{c}
B	Thr	21.18	75.7^{a}	10.10^{a}	38.3^{ab}	5.13	21.8^{a}	2.92	6.6^{b}	0.88^{bc}
C	Lys	20.93	75.1^{a}	9.82^{a}	39.6^{a}	5.23	21.7^{a}	2.83	7.1^{b}	0.95^{bc}
D	Val	20.55	72.2^{a}	9.73^{a}	40.2^{a}	5.33	22.0^{a}	2.83	7.3^{b}	0.98^{bc}
E	Ile	20.32	75.2^{a}	9.51^{a}	36.7^{b}	5.01	23.0^{a}	3.12	7.2^{b}	0.92^{bc}
F	Arg	18.10	71.0^{a}	9.70^{a}	37.5^{ab}	5.25	20.6^{ab}	2.88	6.2^{b}	0.99^{abc}
G	Phe	18.06	72.8^{a}	9.91^{a}	40.7^{a}	5.66	20.0^{bc}	2.83	9.4^{a}	1.32^{a}
H	Trp	16.61	56.7^{b}	8.31^{b}	34.3^{b}	5.25	19.0^{c}	2.90	7.4^{b}	1.12^{ab}
Probability > F			0.0001	0.001	0.008	0.70	0.01	0.10	0.01	0.0001
CV			18.08	11.91	18.29	16.17	16.57	11.99	36.00	34.86

[abc]Means in columns with unlike superscripts differ significantly ($P \leq 0.05$)

to 16% while theoretically all essential amino acids were adequate. Crude protein content of freeze-dried carcasses declined and fat content increased as diet CP decreased. Maintaining the dietary electrolyte balance at 250 meq/kg failed to show any significant benefit on any parameters examined except for abdominal fat (Table 3). Dietary CP also had significant impact on feather development, viscera development, and fat pad formation (Table 4). Feather content (actual weight or % of BW) was not significantly affected until the CP was reduced to less than 18%. Though the percentage of small intestine and gizzard was unaffected by dietary CP, their weight decreased significantly when dietary CP was reduced below 18%. The abdominal fat pad, expressed as a percentage of BW, was significantly increased with the reduction of dietary CP. Neither the weight nor the percentage of BW of other internal organs such as heart and liver was affected by dietary CP level (data not listed).

Fecal nitrogen content significantly decreased in a linear manner with the reduction of dietary CP with the dietary CP ranging from 16.61 to 22.48% (Fig. 1). No higher order than linear regression was demonstrated to fit the data. The regression model is as follows:

$$Y = 0.299X - 1.949$$

where Y is fecal nitrogen content (%); X is the dietary CP (%). For every one percent decrease of dietary CP, there was about 0.3% less nitrogen excreted in the feces. As a result, a 13% reduction of nitrogen excretion was observed without affecting growth performance, which is

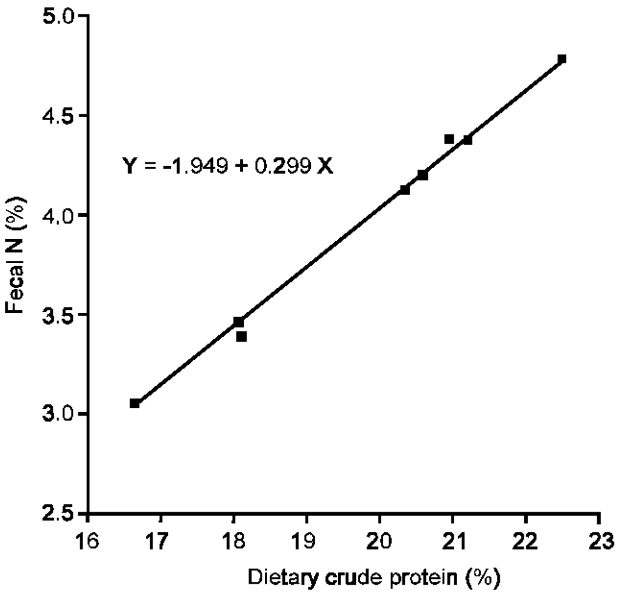

Fig. 1: Observed (■) and estimated (—) fecal N content from broilers fed diets varying in crude protein content

in close agreement with Schutte (1994). He concluded that in broiler chick diets based on corn-soybean meal with adequate lysine and methionine, the protein level could be reduced 1.5 to 2% and the nitrogen excretion would be reduced by 15-20%.

Reducing dietary CP to 20% while satisfying all the essential amino acid requirements (NRC, 1994) seemed to a be threshold point for starting broiler chickens under the conditions of this study, in agreement with previous reports (Pinchasov *et al.*, 1990; Schutte, 1994). There are many factors that may contribute to the failure of obtaining equivalent performance, including reduced levels of potassium and/or altered ionic balance, lack of sufficient nitrogen pool to provide for synthesis of "nonessential" amino acids, and imbalance among certain amino acids. However, maintaining DEB at 250 meq/kg by adding potassium sulfate failed to overcome the growth retardation at lower CP levels. Therefore an acid-base imbalance would not appear to be an explanation for the reduced performance.

Increased carcass and abdominal fat was a problem associated with these lower CP diets fortified with crystalline amino acids, in agreement with Lipstein *et al.* (1975); Fancher and Jensen (1989); Parr and Summers (1991). One of the explanations for this phenomena is associated with the widening of the energy:protein ratio (Bartov *et al.*, 1974; Summers and Leeson, 1979). In this study, the energy: protein ratio ranged from 142 to 190 when dietary CP was reduced from 22.48 to 16.61%. Further work is needed to identify specific problems associated with use of diets low in crude protein.

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