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Growth Responses of Broiler Chicks to Different Levels of Threonine and Tryptophan in Chemically-Defined Diets

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In Experiment 1 three semi-purified diets were formulated using isolated soy protein and crystalline amino acids (AA) to contain three profiles of essential amino acids (EAA) based on NRC (1994; Diet 1), Illinois Ideal Chick Protein (IICP; Diet 2) and Blair *et al.* (1977; Diet 3), respectively, and were fed to male broiler chicks from 0 to 14 d of age. Mean body weight gain at 2 w of age was 132.7, 145.0 and 192.8 g ($P<0.05$) and mean daily feed intake 169.4, 173.9 and 240.4 g ($P<0.05$), respectively, for the NRC, Illinois and PRC diets. Mortality with Diet 3 was significantly higher ($P<0.05$) than with the other two diets, due mainly to an increased incidence of gizzard impaction. In Expt. 2, three levels (90%, 100% and 110% of NRC, 1994) of threonine (Thr) and tryptophan (Trp) were arranged in a 3×3 factorial manner in a modified Diet 3 to give nine purified diets containing crystalline AA as the only source of AA. Broiler chicks were fed a commercial diet during the first w of age and experimental diets were fed from 1 to 3 w of age. Weight gain was significantly lower ($P<0.05$) at the highest levels of both AA (110% Thr and 110% Trp) suggesting that that the diets became imbalanced at high levels of both Thr and Trp. The results of Experiment 2 suggest that dietary levels of Thr and Trp should be targeted at 65% of lysine (equivalent to 0.63% of digestible Thr) and 16% of lysine (equivalent to 0.16% of digestible Trp), respectively, for starting broilers. The results of this study suggest that the requirements for digestible Thr and Trp are no more than 7.2 and 1.8 g/kg diet, respectively.

Key words: broiler, chick, ideal protein, threonine, tryptophan.

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Introduction

The recent availability of feed grade threonine (Thr) and tryptophan (Trp) has increased the need for accurate estimates of the responses to these two potentially limiting AA in broiler diets. The National Research Council (NRC, 1994) recommended requirement values for 0–3 w-old broiler chicks are (air-dry basis): Thr 8.0 g/kg of the diet (3.48% of dietary crude protein) and Trp 2.0 g/kg of the diet (0.87% of dietary crude protein). However, these recommendations are based on published estimates of requirements that range from 5.0 to 8.5 g/kg Thr (2.8 to 3.7% of CP) and 1.4 to 2.8 g/kg Trp (0.78 to 1.2% of CP) (NRC, 1994).

Growth responses to EAA are best measured when the dietary AA are in good balance. The response pattern can then be used to estimate the dietary requirement. D'Mello and Lewis (1970) concluded that it is impossible to determine the chick's actual requirement for EAA unless the diet is in good AA balance, since the AA pattern of the diet will affect the chick's response to supplementation. The need for correct AA balance has led to the development of the ideal protein concept. Ideal protein is defined as the perfect AA profile or bal-

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ance in terms of dietary concentrations among the EAA to meet all AA requirements for a particular species or age without any excesses or deficiencies. In addition, formulating diets according to the ideal protein concept allows for the most efficient and economical use of dietary protein by maximizing nitrogen utilization and minimizing nitrogen excretion (Mack *et al.,* 1999).

Therefore, the objectives of the study were in Expt 1 to compare three dietary profiles of EAA in diets for broilers chicks that were targeted on ideal protein, i.e. NRC (NRC, 1994), Illinois Ideal Chick Protein (IICP) (Baker and Han, 1994) and PRC (Blair *et al.,* 1977); and in Expt 2 using the diet with the AA profile that supported best growth in Expt 1 to evaluate the responses of broiler chicks to graded levels of dietary Thr and Trp in order to provide estimates of requirement for these AAs.

Materials and Methods

Birds, Diets and Management

The guidelines of the Canadian Council on Animal Care were followed and protocols for the experiments were approved by The University of British Columbia Animal Care Committee.

Experiment 1

A total of 108 day-old male (Peterson×Arbor Acres) broiler chicks, vaccinated against Marek's disease, was obtained from a local commercial hatchery and placed into pens of 9 chicks with similar mean initial body weight in battery brooder units (Petersime Incubators Co., Gettysburg, OH, USA) with 23 h of light daily. Individual body weights were recorded at the start of the experiment and weekly thereafter for 14 d. Feed and water were available ad libitum during the 14-d experiment. Feed intake per pen was recorded weekly.

Three semi-purified diets with different AA profiles (Table 1) were used. All three diets were formulated to contain 12 g digestible lysine per kg, as suggested by Baker and Han (1994). The other AA were included in the proportions suggested by the NRC (1994), Baker and Han (1994) and by Blair *et al.* (1977), respectively. All diets were made isonitrogenous (36.8 g N/kg) by adjusting the level of L-glutamic acid, and isoenergetic at 14.64 MJ ME/kg. There were 4 replications (9 chicks per replication) for each dietary treatment.

Cornstarch and corn oil served as the main energy sources, and isolated soy protein (NURISH 1500, Protein Technologies International, St. Louis, MO, USA) and crystalline AA were used as the only sources of AA. The vitamin and mineral mixture was based on one used by Baker and Han (1994) supplemented with $NaHCO_3$ and $Al(OH)_3$, as suggested by Blair *et al.* (1977). All diets contained 30 g/kg cellulose (Solka-Floc), as a fibre source. Extracted hardwood sawdust was used as an additional fibre source in the diet developed by Blair *et al.* (1977), therefore it was included in Diet 3. The hardwood sawdust was ground to pass through a 2 mm screen (Tyler Standard Screen Scale, Ohio, USA), then extracted with dichloromethane for 24 h and dried at air temperature before use.

Isolated protein was used at such levels that the need for supplementation with crystalline AA was minimized. The isolated soy protein contained 832 g crude protein per kg and the following AA (g/kg, air-dry basis): alanine (35.0), arginine (62.0), aspartic acid (96.0), cystine (10.0), glutamic acid (157.0), glycine (34.0), histidine (22.0), isoleucine (40.0), leucine (67.0), lysine (52.0), methionine (30.0), phenylalanine (43.0), proline (42.0), serine (43.0), Thr (31.0), Trp (11.0), tyrosine (31.0), and valine (42.0).

All AA were supplied as L-isomers except methionine, which was supplied as the DL-isomer. Free-base forms of AA were used with the exception of lysine which was provided as the hydrochloride. Feed-grade sources were used for lysine HCl (788.0 g/kg), Thr (985.0 g/kg), and methionine (980.0 g/kg); the remaining AA were of pharmaceutical grade. The true digestibilities of AA in the isolated soy protein and free AA were assumed to be 100% (Chung and Baker, 1992).

Experiment 2

A total of 180 male day-old broiler chicks (Peterson×Arbor Acres), vaccinated against Marek's disease, was obtained from a commercial hatchery. The chicks were placed randomly in groups of 10 in battery brooder units, similar to those used in Expt 1, with 23 h of light daily. All chicks were wing-banded at day-old and were fed a commercial broiler starter diet for the first w. On the morning of d 8, all chicks were weighed individually. Groups of 5 chicks of similar mean body weight were randomly assigned to battery pens. Individual body weight and group feed intake were measured weekly for

Table 1. **Experiment 1: Compositions and amino acid contents of diets fed to broilers from hatching to 14 days of age**

	DIET[1]					
	Amino acid profile based on					
Ingredient g/kg	NRC		Illinois		PRC	
Corn starch	544.4		527.1		381.6	
Isolated soy protein[2]	157.1		125.7		168.0	
Amino acid mixture	36.9		40.0		38.6	
L-glutamic acid	106.4		149.7		92.6	
Corn oil	58.5		60.8		122.5	
Cellulose[3]	30.0		30.0		30.0	
Mineral-vitamin mixture[4]	66.7		66.7		66.7	
Extracted sawdust	0		0		100.0	
Amino acid	g/kg	% relative to lysine	g/kg	% relative to lysine	g/kg	% relative to lysine
Lysine	12.0	100	12.0	100	12.0	100
Arginine	13.68	114	12.6	105	14.4	120
Histidine	3.84	32	3.84	32	4.8	40
Isoleucine	8.76	73	8.04	67	9.6	80
Leucine	13.08	109	13.08	109	16.8	140
Methionine	5.52	46	4.32	36	6.6	55
Cystine	4.32	36	4.32	36	1.68	14
Phenylalanine	7.92	66	6.6	55	8.4	70
Tyrosine	6.72	56	6.0	50	8.4	70
Threonine	8.76	73	8.04	67	8.4	70
Tryptophan	2.16	18	1.92	16	2.4	20
Valine	9.84	82	9.24	77	10.32	86
Glycine	13.68	114	7.8	65	12.0	100
Proline	6.6	55	5.28	44	8.4	70
Calculated analysis						
ME MJ/kg	14.64		14.64		14.64	
N g/kg	36.8		36.8		36.8	
Ca g/kg	12.0		12.0		12.0	
P available g/kg	6.6		6.6		6.6	

[1] Based, respectively, on NRC (1994); Baker and Han (1994); Blair *et al.* (1977).
[2] NURISH 1500, Protein Technologies International, St. Louis, MO, USA.
[3] Solka-Floc, International Fiber Corporation, North Tonawanda, NY 14120, USA.
[4] Supplied per kg: 2 g choline chloride (100%), 20 mg thiamin HCl, 10 mg riboflavin, 30 mg calcium pantothenate, 50 mg niacin, 6 mg pyridoxine HCl, 4 mg folacin, 0.6 mg biotin, 0.04 mg cobalamin, 100 mg inositol, 2 mg para-aminobenzoic acid, 5,200 IU vitamin A, 600 ICU vitamin D_3, 20 IU vitamin E, 2 mg menaphthone, 125 mg ethoxyquin (Santoquin, Monsanto Agricultural, St. Louis, MO, USA), 5 g NaCl, 33 g $Ca_3(PO_4)_2$, 0.6 g MgO, 4.5 g K_2CO_3, 5 g $NaHCO_3$, 5 g Al $(OH)_3$, 650 mg $MnSO_4$. H_20, 50 mg ZnO, 567 mg $FeSO_4$. $7H_2O$, 20 mg CuO_4. $5H_2O$, 0.4 mg Na_2SeO_3, 40 mg KI, 1 mg $CoSO_4$. $7H_2O$, 9 mg H_3BO3, 9 mg Na_2MoO_4. $2H_2O$.

each of 14 d.

A total of 9 diets was fed to the birds from 7–21 d of age (Table 2), with 4 replications of 5 birds per diet. Cornstarch and corn oil served as the main energy sources, with crystalline AA as the only source of AA. The intention was to provide exact details of the AA contents of the diets, and to aid in the interpretation of results. All AA were supplied as L-isomers except methionine, which was supplied as the DL-isomer. Free-base forms of AA were used, with the exception of lysine which was provided as the hydrochloride. Feed-grade sources were used for lysine HCl (788.0 g/kg), Thr (985.0 g/kg), and methionine (980.0 g/kg); the remaining AA were of pharmaceutical grade. The true digestibility of the crystalline AA was assumed to be 100% (Izquierdo *et al.,* 1988; Chung and Baker, 1992; Zhang and Parsons, 1993). All diets were made isonitrogenous (38.3 g N/kg) by adjusting the level of L-glutamic acid. The dietary ME level was set at 15.25 MJ/kg, to provide the same ME/N ratio as in Expt 1, and the digestible lysine level was set at 11.0 g/kg.

Table 2. **Experiment 2: Compositions and calculated analyses of diets (g/kg) fed to broilers from 7-21 days of age**

	DIET								
	1	2	3	4	5	6	7	8	9
Ingredient									
Corn starch	524.6	524.6	524.6	524.6	524.6	524.6	524.6	524.6	524.6
AA mixture[1]	115.4	116.5	117.5	115.8	116.8	117.8	116.1	117.2	118.1
Lglutamic acid	119.8	118.7	117.7	119.4	118.4	117.4	119.1	118.0	117.1
Corn oil	100.0	100.0	100.0	100.0	100.0	100.0	100.0	100.0	100.0
Cellulose (Solka-Floc)	30.0	30.0	30.0	30.0	30.0	30.0	30.0	30.0	30.0
Mineralvitamin mixture[2]	60.2	60.2	60.2	60.2	60.2	60.2	60.2	60.2	60.2
Extracted sawdust	50.0	50.0	50.0	50.0	50.0	50.0	50.0	50.0	50.0
AA Levels g/kg (% relative to Lys)									
Lysine	11.0(100)	11.0(100)	11.0(100)	11.0(100)	11.0(100)	11.0(100)	11.0(100)	11.0(100)	11.0(100)
Threonine	7.2(65)	8.0(73)	8.8(80)	7.2(65)	8.0(73)	8.8(80)	7.2(65)	8.0(73)	8.8(80)
Tryptophan	1.8(16)	1.8(16)	1.8(16)	2.0(18)	2.0(18)	2.0(18)	2.2(20)	2.2(20)	2.2(20)
Arginine	13.2(120)	13.2(120)	13.2(120)	13.2(120)	13.2(120)	13.2(120)	13.2(120)	13.2(120)	13.2(120)
Histidine	4.4(40)	4.4(40)	4.4(40)	4.4(40)	4.4(40)	4.4(40)	4.4(40)	4.4(40)	4.4(40)
Isoleucine	8.8(80)	8.8(80)	8.8(80)	8.8(80)	8.8(80)	8.8(80)	8.8(80)	8.8(80)	8.8(80)
Leucine	15.4(140)	15.4(140)	15.4(140)	15.4(140)	15.4(140)	15.4(140)	15.4(140)	15.4(140)	15.4(140)
Methionine	6.1(55)	6.1(55)	6.1(55)	6.1(55)	6.1(55)	6.1(55)	6.1(55)	6.1(55)	6.1(55)
Cystine	1.5(14)	1.5(14)	1.5(14)	1.5(14)	1.5(14)	1.5(14)	1.5(14)	1.5(14)	1.5(14)
Phenylalanine	7.7(70)	7.7(70)	7.7(70)	7.7(70)	7.7(70)	7.7(70)	7.7(70)	7.7(70)	7.7(70)
Tyrosine	7.7(70)	7.7(70)	7.7(70)	7.7(70)	7.7(70)	7.7(70)	7.7(70)	7.7(70)	7.7(70)
Valine	9.5(86)	9.5(86)	9.5(86)	9.5(86)	9.5(86)	9.5(86)	9.5(86)	9.5(86)	9.5(86)
Glycine	11.0(100)	11.0(100)	11.0(100)	11.0(100)	11.0(100)	11.0(100)	11.0(100)	11.0(100)	11.0(100)
Proline	7.7(70)	7.7(70)	7.7(70)	7.7(70)	7.7(70)	7.7(70)	7.7(70)	7.7(70)	7.7(70)
Calculated analysis									
ME MJ/kg	15.25	15.25	15.25	15.25	15.25	15.25	15.25	15.25	15.25
N g/kg	38.3	38.3	38.3	38.3	38.3	38.3	38.3	38.3	38.3
Ca g/kg	12.0	12.0	12.0	12.0	12.0	12.0	12.0	12.0	12.0
P available g/kg	6.6	6.6	6.6	6.6	6.6	6.6	6.6	6.6	6.6

[1] Supplied the amino acids shown. Amounts of amino acids except threonine and tryptophan were the same in all diets.

[2] Provided the same amounts of mineral and vitamins as in Expt 1 (Table 1).

The digestible lysine level was reduced from 12.0 g/kg used in Expt 1 to 11.0 g/kg, since several reports indicated that the lysine requirement of 1-week-old broilers was around 11.0 g/kg (Han and Baker, 1991, 1993; Vazquez and Pesti, 1997).

The diets were formulated to contain graded levels of Thr and Trp, representing 90%, 100% and 110% of the NRC (1994) recommendations for Thr and Trp. The previous experiment (Expt 1) showed that Diet 3 gave the best growth response. Therefore it was used as the basis of the diets in this experiment, with the levels of Thr and Trp adjusted to 90%, 100% and 110% of the NRC (1994) recommendations. The levels of Thr and of Trp were arranged in a 3×3 factorial manner (Table 2). The levels of other EAA were as in Diet 3 used in Expt 1. Extracted sawdust was included in all diets as a fibre source, in addition to cellulose. An inclusion rate of 50 g/kg was used, based on the results of Expt. 1. Feed and water were offered *ad libitum* during the experiment.

Statistical Analyses

Experiment 1:

Data were subjected to analysis of variance (ANOVA) procedures appropriate for a completely randomized design, using the General Linear Models (GLM) procedure of SAS software (SAS Institute, 1996). Mortality data were subjected to arcsine conversion prior to statistical analysis. Where treatments were found to be significantly different, Tukey's multiple range test (Snedecor and Cochran, 1980) was used to determine the significant differences between treatment least-square means.

Experiment 2:

Data were analyzed as in Expt. 1, for a 3×3 factorial design.

Results

Experiment 1

Live body weight of the birds fed the PRC diet was significantly higher ($P<0.05$) at d 7 and 14 than with the other two diets (Table 3). No significant differences in body weight at d 7 and 14 were found between the groups fed the NRC and Illinois diets. Body weight gain and feed intake followed the same pattern. Feed conversion efficiency was not significantly different among birds fed the three diets. Overall, during the 14 d study birds fed the PRC diet consumed significantly more feed ($P<0.05$) and gained significantly more weight ($P<0.05$) than those fed the other two diets.

An average of 5.8 % birds died during the experiment, mainly from impacted crops and yolk-sac infections. Mortality was higher ($P<0.05$) with the PRC diet, which was attributed to the inclusion level of sawdust.

Experiment 2

Growth performance during d 7–14

Level of dietary Thr and Trp did not significantly ($P>0.05$) affect any growth parameter (Table 4).

Growth performance during d 14–21

Body weight was not affected significantly by dietary Thr level, however it was significantly reduced ($P<0.05$) as dietary Trp was raised to 110% of the NRC recommendation (Table 4). Weight gain was significantly reduced ($P<0.05$) when Thr or Trp levels were raised to 110% of the NRC recommendation. Feed intake was reduced as Trp level was increased to the highest level ($P<0.05$), but was unaffected by dietary Thr level. Feed conversion efficiency was not affected significantly by treatment.

Interactions ($P<0.05$) between Thr and Trp on body weight at d 21 and weight gain during d 14–21 and overall weight gain were observed (Table 5). Body weight at 21 d of age was significantly lower ($P<0.05$) in birds fed diets containing 110% Trp when Thr level reached 110% of the NRC (1994) recommendation and it was significantly lower ($P<0.05$) in birds fed diets containing 110% Thr when Trp level reached 110% of the NRC (1994) recommendation.

Diets based on Trp at 100% or 110% of the NRC (1994) recommendations gave significantly reduced ($P<0.05$) weight gain during d 14–21 and live body weight at 21 d when the Thr level was 100% or 110% of the NRC (1994) recommendation.

Overall growth performance (d 7–21)

Overall weight gain was not significantly affected ($P>0.05$) by dietary Thr level but was decreased significantly ($P<0.05$) as the level of Trp increased (Table 4). A treatment interaction on overall body weight gain was observed (Table 5). Overall weight gain was significantly reduced ($P<0.05$) when the diets contained 100% of Thr and 110% of Trp or 110% of Thr and 110% of Trp relative to the NRC (1994) recommendations. Overall weight gain was significantly reduced ($P<0.05$) at the highest level of Thr when the dietary level of Trp was the highest. Neither feed intake nor gain/feed ratio was affected significantly ($P>0.05$) by treatment.

No birds died or had to be removed during the experiment.

Discussion

This study showed significant responses to increasing levels of dietary Thr and Trp in AA-based

Table 3. **Experiment 1: Effect of diets based on different amino acid profiles on growth performance of male broilers from 1-14 d of age**

DIET	Live body weight (g)			Body weight gain (g)			Feed intake (g/bird)			Feed conversion efficiency (gain/feed)			Mortality %*		
	Start	Day 7	Day 14	Day 7	Day 14	Overall	Day 7	Day 14	Overall	Day 7	Day 14	Overall	Day 7	Day 14	Overall
NRC[1]	41.8	86.3[b]	175[b]	44.3[b]	88.4[b]	132.7[b]	53.9[b]	115.5[b]	169.4[b]	0.82	0.74	0.77	4.8[b]	8.1	6.5
Illinois	40.3	82.6[b]	185[b]	41.7[b]	103.3[b]	145.0[b]	48.2[b]	125.6[b]	173.9[b]	0.85	0.82	0.83	0[a]	11.03	5.5
PRC	40.7	103.5[a]	233[a]	61.2[a]	131.5[a]	192.8[a]	64.2[a]	176.2[a]	240.4[a]	0.95	0.75	0.79	4.8[b]	23.5	14.2
Mean	40.9	90.8	198	49.1	107.7	156.8	55.4	139.1	194.6	0.87	0.77	0.80	3.2	14.3	5.8
SEM[2]	0.4	2.0	9.7	2.2	7.2	9.9	2.4	9.2	10.7	0.03	0.02	0.03	0.96	2.96	1.91

[1] As in Table 1.
[2] Standard error of the mean; data represent means of four replications of nine chicks.
[a, b] Treatment means with different superscripts within a column are significantly different at $P<0.05$.
* Data were analyzed and presented on arcsine transformed values.

Table 4. **Experiment 2: Effect of dietary level of threonine and tryptophan on growth performance of male broilers from 7–21 days of age**

DIET	Live body weight[1] (g)		Body weight gain (g)			Feed intake (g/bird)			Feed conversion efficiency (gain/feed)		
	D 14	D 21	D 7–14	D 14–21	Overall	D 7–14	D 14–21	Overall	D 7–14	D 14–21	Overall
Threonine											
90% NRC	273.3	451.4	134.4	178.3[a]	312.5	203.3	211.3	414.7	0.66	0.85	0.75
100% NRC	268.3	440.4	135.9	172.4[a]	308.4	196.4	201.0	397.1	0.69	0.87	0.77
110% NRC	277.7	434.5	142.3	156.6[b]	298.9	197.3	179.3	376.6	0.72	0.85	0.79
Tryptophan											
90% NRC	279.8	462.8[a]	142.4	183.1[a]	325.3[a]	198.9	206.1[a]	405.1	0.72	0.90	0.80
100% NRC	270.1	449.8[a]	137.8	179.4[a]	317.2[a]	202.1	213.6[a]	415.6	0.68	0.84	0.76
110% NRC	269.3	413.7[b]	132.5	144.7[b]	277.3[b]	196.0	171.9[b]	367.9	0.68	0.83	0.76
Overall mean	273.1	442.1	137.6	168.7	306.5	199.1	197.2	396.2	0.69	0.85	0.77
Pooled SEM[2]	2.9	5.6	2.0	3.9	5.1	2.2	8.3	10.0	0.01	0.01	0.02
Factorial effects[3]											
Threonine (Thr)	NS	NS	NS	*	NS	NS	NS	NS	NS	NS	NS
Tryptophan (Trp)	NS	*	NS	*	*	NS	*	NS	NS	NS	NS
Thr×Trp	NS	*	NS	*	*	NS	NS	NS	NS	NS	NS

[1] Mean body weight at the start of the experiment (day 7) was 135 g.
[2] Pooled standard error of the mean; data represent means of four replications of five chicks.
[3] NS=not significant (P > 0.05) or *=significant at $P<0.05$.
[a,b] Treatment means with different superscripts within a column and within a treatment group are significantly different at $P<0.05$.

Table 5. **Experiment 2: Significant interactions of dietary levels of threonine and tryptophan on growth performance of male broilers from 7–21 days of age**

DIET	Live body weight[1] at 21 d (g)				Gain in body weight 14–21 d (g)				Gain in body weight 7–21 d (g)			
	Tryptophan				Tryptophan				Tryptophan			
Threonine	90% NRC	100% NRC	110% NRC	Mean	90% NRC	100% NRC	110% NRC	Mean	90% NRC	100% NRC	110% NRC	Mean
90% NRC	471.8	457	425.5	451.4	201	181.3	152.8	178.3[a]	336.8	322	312.5	312.5
100% NRC	460	453	408.5	440.4	182.8	182.8	151.3	172.4[a]	325	318	308.4	308.4
110% NRC	457	439.8	406.5	434.5	166.3	175.5	128.5	156.6[b]	322	304.8	298.9	298.9
Mean	462.8[a]	449.8[a]	413.7[b]	442.1	183.1[a]	179.4[a]	144.7[b]	168.7	325.3[a]	317.2[a]	277.3[b]	306.5

[1] Mean body weight at the start of the experiment (day 7) was 135 g.
[a,b] Treatment means with different superscripts within a treatment group are significantly different at $P<0.05$.

diets containing 28.3 g N/kg. Gain in body weight was higher when the diet contained 7.2 or 8.0 g/kg Thr than with 8.8 g/kg Thr. Body weight, body weight gain and feed intake were higher with 1.8 or 2.0 g/kg Trp than with 2.2 g/kg Trp. The depression in growth performance was particularly apparent when Thr and Trp were increased simultaneously. The diets used were based on crystalline AA, which are known to be highly digestible in the chick. Therefore the dietary contents of AA can be regarded as digestible AA. The results of this study suggest that the requirements for digestible Thr and Trp are no more than 7.2 and 1.8 g/kg diet, respectively.

Mortality with Diet 3 In Experiment 1 was significantly higher ($P<0.05$) than with the other two diets, due mainly to an increased incidence of gizzard impaction. This effect was attributed to the high inclusion rate of extracted sawdust to improve the texture of the diet. Mortality was in the acceptable range in Experiment 2 when the inclusion rate of extracted sawdust was reduced.

The NRC (1994) estimates of requirement are Thr 8.0 g/kg and Trp 2.0 g/kg diet. These estimates are based on corn/soy diets with an estimated AA digestibility of about 90% (Rhodimet Nutrition Guide, 1993; NRC, 1994). In terms of digestible Thr and Trp the NRC estimates of requirement can therefore be calculated as 7.2 and 1.8 g/kg diet, respectively. Our estimates of requirements for digestible Thr and Trp agree with these estimates.

Other recent estimates of Thr and Trp requirements have been reported by Baker *et al.* (2002), using male cross-bred chicks fed diets based on corn gluten meal and crystalline AA. The digestible Thr and Trp values were estimated at 5.9–6.0 and 1.8 g/kg, respectively. Their estimated Trp requirement agrees with our findings. Their estimated Thr requirement is much lower than the NRC (1994) requirement and is outside the range of values tested in our study.

These results can be compared with other recent findings. Holsheimer *et al.* (1994) reported an improvement in weight gain and feed efficiency of young broiler chicks when dietary Thr was increased to 7.25 g/kg. Kidd *et al.* (1996) found that Thr levels ranging from 7.36 to 8.96 g/kg failed to improve the weight gain of 1–21 d-old broiler chicks. Using a milo-soybean meal-corn gluten meal basal diet, Yamazaki *et al.* (1997) concluded that 13 wk-old broiler chicks require a dietary level of 6.5 g/kg digestible Thr, slightly lower than the estimate obtained in our study. Smith and Waldroup (1988) concluded that a diet containing 1.6 g/kg Trp met the requirement of the growing chick for Trp. On the other hand, several researchers have reported a higher requirement value for Trp (Abebe and Morris, 1990; Parr and Summers, 1991; Han *et al.,* 1991), some researchers reporting requirement values of 2.25–2.4 g/kg diet (Thomas *et al.,* 1992; Rhodimet Nutrition Guide, 1993; Austic, 1994). Corzo *et al.* (2005) suggested a requirement value of 2.0–2.2 g Trp per kg diet for broiler chicks and Hsia *et al.* (2005) suggested 1.98 g per kg diet.

It is probably more exact to state requirements in terms of dietary crude protein (CP), in accordance with the principle of achieving the correct balance of dietary AA. Thus the NRC (1994) estimates (which are based on a dietary CP level of 230 g/kg for starting broilers) can be expressed as 3.13 and 0.78% digestible Thr and Trp, respectively, of dietary CP. The corresponding estimates from our study are 4.04 and 1.01%, respectively. These values are not directly comparable, however, since practical diets based on regular feedstuffs usually contain an excess of some AA. Purified diets, especially those based on crystalline AAs, usually contain minimal excesses of AAs.

Other recent estimates of Thr requirement expressed as a percentage of protein have been reported as 4.53% (Holsheimer *et al.,* 1994) and 3.96–4.13% (Koide and Ishibashi, 1995). Boomgaardt and Baker (1971) reported that when Trp requirement was expressed as percent of the diet, the requirement increased with dietary protein level. However, when expressed as a % of CP, the requirement remained constant at 0.87% over five different CP levels. Smith and Waldroup (1988) reported a Trp requirement value of 0.80% of CP. Abebe and Morris (1990) reported a value of 1.2% CP. Austic (1994) and Leeson and Summers (1997) suggested a value of 1.1% and 0.9% of CP, respectively.

In seeking to achieve the ideal balance of dietary AAs, several authors have suggested that Lys be used as the reference AA (e.g. Baker, 1997). Levels of other AAs can then be expressed relative to Lys. Baker *et al.* (2002) calculated that the ideal ratios of digestible Thr and Trp to Lys are 55.7 and 16.6%, respectively. The corresponding ratios from our results are 65 and 16%, respectively. There is, therefore, good agreement on Trp in these findings. Our suggested Thr: Lys ratio is higher, due possibly to the fact that Baker *et al.* (2002) used a wider range of Thr values than in our study. Another possible explanation is that the body weight gain obtained in our study was higher than in the Baker *et al.* (2002) study; 22.7 g/d compared with 20.7 g/d. A Thr: Lys ratio of 62% was reported by Austic (1994).

Compared to the AA profiles in the diets based on NRC (1994) and the Illinois group (IICP) (Expt 1), the diet based on the PRC profile contained a higher ratio of most EAA relative to Lys except total SAA, Thr and glycine. The diet based on this AA profile gave the best weight gain and the chicks consumed significantly more ($P < 0.05$) of this diet throughout the 2w study than birds fed the other two diets. This diet has been listed by the NRC (1984, 1994) as a standard reference diet. It was based on the recommendations of Scott *et al.* (1982), Bolton and Blair (1974) and Lee and Blair (1974). The higher voluntary feed intake, and the higher growth rate, found with this diet are attributed to a more optimal AA profile since all diets were formulated to contain the same ME and N levels. It is well known that dietary AA balance is important in maximizing feed consumption of diets

(D'Mello, 1993; 1994). The extracted sawdust in the PRC-based diet may have improved the texture and acceptability of the diet, but this does not fully explain the greater feed intake. Further studies should be conducted on the role of fibre in purified diets for chicks.

As stated above, the PRC-based diet (Blair *et al.,* 1977) has been listed as a standard reference diet by the NRC (1994), although the fibre level may require modification. The inclusion of a fibre source such as extracted sawdust appears to improve the texture of the purified diet. The results of Expt 2 suggest that Diet 1 used in this experiment is also a satisfactory reference diet for broiler chicks.

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