



The Optimum Dietary Essential Amino Acid Pattern for Male Taiwan Country Chicks*

Hen-Wei Wei, Hsin-Mei Kuo, Wen-Zan Chiu and Bao-Ji Chen**

Department of Animal Science and Technology, National Taiwan University, Taiwan

ABSTRACT : The objective of this study was to estimate the optimum dietary essential amino acid pattern for male Taiwan country chicks. A series of experiments was conducted with chicks, 14 d of age, for 2 wks. A basal synthetic diet was established using a dose response test for all essential amino acids referring to the broiler requirements recommended by NRC (1994). Twelve chicks were sacrificed at the beginning to provide initial body nitrogen data, and every group of six birds received the basal diet or a diet with a deficiency in a single essential amino acid in twenty one treatments by intubation according to their daily metabolic body weight (MBW). Deposited body nitrogen was determined using comparative slaughtering. According to the daily intake from the limiting amino acid per unit of MBW and the body nitrogen accretion rates for every two deficient groups with the same limiting amino acid but at different levels, a corresponding straight line was computed for each essential amino acid to intersect with a horizontal line made by the body nitrogen accretion rate of the control group. The x coordinate of the intersection represented the daily requirement for growth plus maintenance based on MBW corresponding to the essential amino acid. The amino acid ratios can be considered as the optimum pattern of dietary essential amino acids. The results, expressed with respect to lysine = 100, were arginine 105, methionine 81, histidine 34, tryptophan 18, leucine 103, phenylalanine 135, isoleucine 69, threonine 65 and valine 79. This pattern could be utilized to compute the dietary requirements (g/kg feed) for all essential amino acids by multiplying by the requirement of a single essential amino acid cited from the literature. (**Key Words :** Taiwan Country Chicks, Essential Amino Acids, Body Nitrogen Accretion, Optimum Patterns)

INTRODUCTION

Taiwan country chickens are famous for their flavor (Cheng et al., 2008) and contributed approximately 60% of the Taiwan broiler industry value in 2007 (Agricultural statistics yearbook, 2007). The chickens are divided into two types, red or black, according to their feather color and characterized by a single comb, blue shank and a rich breast. These birds are not taken to market until 13 wk of age. Most studies on the nutrition of Taiwan country chickens focused on requirements for protein, metabolizable energy, calcium and phosphorous (Fan and Lee, 1984; Yu and Hsu, 1989; Yu et al., 1990; Shih et al., 1993; Huang et al., 2007) or on the effect of dietary inclusion of dehydrated food waste products on the growth performance (Chen et al., 2007). Only a handful of literature has concentrated upon

the requirement for individual amino acids, including lysine (Hsu and Lin, 1990), methionine (Hsu and Lin, 1989) and tryptophan (Lin and Hsu, 1994; Chen and Hsu, 1996). Establishing the optimum pattern of dietary essential amino acids will be an approach to attain the requirements (g/kg diet) of all essential amino acids for Taiwan country chickens because they would be computed by means of multiplying the optimum pattern by the requirement for a single essential amino acid resulting from dose-response trials. Wang and Fuller (1989) extended a method, published by Bender (1965) with rats, based on the removal of non-limiting amino acids with no effect on nitrogen retention. They set up the optimum dietary amino acid pattern for growing pigs. This method could not distinguish the requirements for maintenance and tissue protein accretion. Therefore, Fuller et al. (1989) designed consecutive trials to measure, simultaneously, requirements for maintenance and for body protein accretion for pigs. In addition, numerous literature reported on the accretion and maintenance requirements for individual essential amino acids for chicks (Baker, 1991; Burnham and Gous, 1992; Baker et al., 1996; Edwards et al., 1997; Kim et al., 1997a.

* The work was financially supported by the Council of Agriculture, Taiwan Government; research project number 91AS-1.1.3-AD-U2.

** Corresponding Author: Bao-Ji Chen. Tel: +886-233-664153, Fax: +886-227-324070, E-mail: bjchen@ntu.edu.tw

Received November 21, 2008; Accepted February 14, 2009

b, c; Edwards and Baker, 1999; Edwards et al., 1999; Sklan and Noy, 2005). After modifying the previous studies done on swine or chicks, a series of experiments were conducted to establish the optimum dietary essential amino acid pattern for red feather type male Taiwan country chicks to compute the requirements for individual essential amino acids and simultaneously set up requirements for maintenance and for growth.

MATERIALS AND METHODS

Animals

Day-old male chicks were purchased from a commercial hatchery and raised in electrically heated battery brooders with a wire floor (60×90 cm) holding 10 birds until 2 wk of age. The temperature in each brooder was maintained at 32±1°C at the beginning and then decreased by 2°C every 4 d. Water and a corn-soybean diet (Table 1) were supplied *ad libitum*.

Exp. 1

Ten chicks weighing 242.4±23.0 g were allotted into individual brooders at 2 wk old. Feed (Table 1) and water

Table 1. The practical diet used in the first trial

Ingredients	g/kg
Corn (dent yellow grain)	448.40
Soybean meal (Solvent extracted)	389.70
Fish meal	30.00
Choline chloride (50%)	0.50
Soybean oil	88.90
Calcium phosphate (dibasic)	16.00
Calcium carbonate	14.70
Sodium chloride, iodized	4.80
Premix ¹	5.00
Methionine	2.00
Chemical composition	
ME (Mcal/kg) ²	3.2
Moisture (%)	12.27
Crude protein (%)	23.26

¹ Supplying (per kilogram): retinol 4,500 IU, cholecalciferol 600 IU, α -tocopherol 20 IU, menadione (50%) 2.0 mg, thiamin 2.22 mg, riboflavin 3.96 mg, nicotinic acid 38.5 mg, cyanocobalamin (0.1%) 50 mg, folic acid (98%) 0.62 mg, biotin 8.25 mg, pyridoxine-HCl 4.68 mg, d-pantothenate-hemi-calcium 30.03 mg, manganese sulfate (monohydrate) 0.161 g, sodium selenite (pentahydrate) 0.16 mg, zinc sulfate (heptahydrate) 68.9 mg, ferrous sulfate (pentahydrate) 71.5 mg.

² Calculated

were provided *ad libitum*. Daily food intake and body

Table 2. Experimental diets used in the second trial

Ingredients	Treatment (%) ¹				
	60	80	100	120	140
	g/kg				
Cellulose	86.99	97.48	98.31	97.76	108.25
Casein	58.00	58.00	58.00	58.00	58.00
Gelatin	59.00	59.00	59.00	59.00	59.00
Glucose	444.87	443.96	459.69	477.82	476.91
Choline chloride (50%)	3.84	3.84	3.84	3.84	3.84
Soybean oil	71.00	71.00	64.00	56.00	56.00
Premix ²	70.00	70.00	70.00	70.00	70.00
Threonine	1.30	2.90	4.50	6.10	7.7
Glutamic acid	197.97	169.56	141.16	122.75	84.34
Valine	0.31	2.11	3.91	5.71	7.51
Methionine	3.25	5.13	7.01	88.9	10.77
Isoleucine	1.21	2.81	4.41	6.01	7.61
Leucine	0.36	2.76	5.16	7.56	9.96
Phenylalanine	0.48	3.16	5.84	8.52	11.20
Lysine HCl	0.00	2.75	5.50	8.25	11.00
Histidine	0.03	0.73	1.43	2.13	2.83
Arginine	0.94	3.97	6.99	10.02	13.04
Tryptophan	0.45	0.85	1.25	1.65	2.05
Chemical composition					
ME (Mcal/kg) ³	3.2	3.2	3.2	3.2	3.2
Moisture (%)	6.08	6.37	6.60	6.67	7.15
Crude protein (%)	23.26	23.73	23.55	23.64	23.09

¹ The percentage of requirements recommended by NRC (1994) for all the essential amino acids for broilers.

² Supplying (per kilogram): retinol 4,500 IU, cholecalciferol 600 IU, α -tocopherol 20 IU, menadione (50%) 2.0 mg, thiamin 2.22 mg, riboflavin 3.96 mg, nicotinic acid 38.5 mg, cyanocobalamin (0.1%) 50 mg, folic acid (98%) 0.62 mg, biotin 8.25 mg, pyridoxine-HCl 4.68 mg, d-pantothenate-hemi-calcium 30.03 mg, calcium carbonate 21.44 g, calcium phosphate (dibasic) 8.24 g, sodium bicarbonate 0.4 mg, potassium bicarbonate 0.4 mg, sodium chloride (iodized) 3.52 g, potassium chloride 0.14 mg, potassium phosphate (dibasic) 6.84 mg, potassium iodate 0.51 mg, cupric sulfate (pentahydrate) 34.93 mg, manganese sulfate (monohydrate) 0.207 g, sodium selenite (pentahydrate) 0.37 mg, zinc sulfate (heptahydrate) 0.194 g, ferrous sulfate (pentahydrate) 0.443 g, magnesium sulfate (pentahydrate) 6.69 g.

³ Calculated.

weight were recorded to estimate metabolizable energy (k) for daily maintenance plus growth based on MBW in 2-4 wk of age. The k was computed using the equation, $I \times ME = k \times (BW)^{0.75}$, in which I is intake and ME expresses the metabolizable energy of feed while $(BW)^{0.75}$ represents MBW.

Exp. 2

One hundred chicks weighing 243.2 ± 20.1 g were randomly distributed into brooders of four birds each at 2 wk of age. Water and feed (Table 2) were provided *ad libitum* for 2 wk. The percentages of all essential amino acids in five diets were designed as 60, 80, 100, 120 or 140% referring to the broiler requirements suggested by the NRC (1994). At the end of the experiment body weight and feed intake were recorded for calculating feed efficiency.

Exp. 3

Twelve chicks with body weight around 234.6 ± 15.8 g were sacrificed at the beginning to provide initial body nitrogen data for comparative slaughtering and then one hundred and twenty six chicks weighing 238.3 ± 14.2 g were randomly assigned into twenty one treatments at the end of wk 2. Every six chicks received a basal diet or a diet deficient in a single essential amino acid (Table 3) by intubation according to their daily MBW while water was provided *ad libitum*. Rations were partitioned into four parts and each part was supplied to the chicks every 6 h. Before feeding, the diets were combined with water at a ratio of 5 to 2. Fourteen days later, the chicks were sacrificed using CO_2 asphyxiation and transferred to glass trays after overnight fasting and then weighed. A midline incision was made from the anus to the pharynx to open the abdominal cavity. After oven-drying at $105^\circ C$ for 36 h, the cadaver

was weighed, chopped and then ground in a coffee grinder. The residual moisture content of the ground samples was determined by oven-drying at $100^\circ C$ for 4 h (Wei and Fuller, 2006). The ground samples were then subjected to the Kjeldahl method for analyzing nitrogen content (AOAC, 1984). Feed samples containing approximately 10 mg nitrogen each combined with 2 ml of 12.5 mM norleucine solution serving as an external standard were hydrolyzed in 6 N hydrochloric acid at $110^\circ C$ for 24 h. After being filtered, quantified, rotary-evaporated and reconstituted, hydrolysate samples were analyzed using an amino-acid analyzer (Beckman 6300) for the limiting amino acid content to ensure that the diets were prepared properly. The methionine and control group feed samples were preoxidized using performic acid before hydrolysis for quantifying methionine (Moore, 1963). The tryptophan content of the tryptophan-deficient and control diets was determined separately using HPLC (Gilson 805) following alkaline hydrolysis at $120^\circ C$ for 16 h using 4.1 M lithium hydroxide (Langer, 1995).

Statistical analysis

Apart from t-test comparison with Microsoft Excel 2003, most of data were statistically analyzed using the SAS (SAS, 1988) GLM procedure and differences among groups were determined using Duncan's multiple-range test after subjected to ANOVA. A level of $p < 0.05$ was used as the criterion for statistical significance.

RESULTS

Exp. 1

According to the daily feed intake and body weight of individual chicks, the mean estimate (k) of metabolizable

Table 3. Experimental diets used in the third trial

Treatment	Control ¹	Lysine	Arginine	Methionine	Histidine	Tryptophan	Leucine	Phenylalanine	Isoleucine	Threonine	Valine
Ingredients	g/kg										
Cellulose	96	93	92	93	93	95	95	95	95	95	92
Glucose	438.65	438.98	439.18	430.05	427.38	439.72	439.73	436.31	435.55	439.2	439.1
Choline chloride (50%)	3.84	3.84	3.84	3.84	3.84	3.84	3.84	3.84	3.84	3.84	3.84
Soybean oil	56	56	56	56	56	56	56	56	56	56	56
Premix ²	70	70	70	70	70	70	70	70	70	70	70
Threonine	9.84	9.84	9.84	9.84	9.84	9.84	9.84	9.84	9.84	9.84	9.84
Glutamic acid	226.78	223.84	235.95	244.86	249.01	231.37	232.43	231.93	233.11	228.27	228.61
Valine	11.07	11.07	11.07	11.07	11.07	11.07	11.07	11.07	11.07	11.07	11.07
Methionine	11.07	11.07	11.07	11.07	11.07	6.41	5.34	11.07	11.07	11.07	11.07
Isoleucine	9.84	9.84	9.84	9.84	9.84	9.84	9.84	9.84	9.84	9.84	9.84
Leucine	14.76	14.76	14.76	14.76	14.76	14.76	14.76	14.76	14.76	14.76	14.76
Phenylalanine	16.48	16.48	16.48	16.48	16.48	16.48	16.48	16.48	16.48	16.48	16.48
Lysine	13.53	9.14	7.83	13.53	13.53	13.53	13.53	13.53	13.53	13.53	13.53
Histidine	4.31	4.31	4.31	4.31	4.31	4.31	2.49	2.08	4.31	4.31	4.31
Arginine	15.38	15.38	15.38	8.90	7.42	15.38	15.38	15.38	15.38	15.38	15.38
Tryptophan	2.46	2.46	2.46	2.46	2.46	2.46	2.46	2.46	2.46	2.46	2.46
Chemical composition											
ME (Mcal/kg) ³	3.2	3.2	3.2	3.2	3.2	3.2	3.2	3.2	3.2	3.2	3.2
Moisture (%)	7.18	6.42	6.74	6.40	6.82	7.11	7.01	7.02	7.08	6.93	7.06
Crude protein (%)	23.82	23.09	24.73	22.25	23.01	23.20	23.52	23.93	24.13	23.07	23.28
Limiting amino acid (g/kg)	-	9.05	7.66	9.07	7.31	6.72	5.59	2.58	2.00	1.47	1.22

¹ The 123% of requirements recommended by NRC (1994) for all the essential amino acids for broilers.

² The same as that of the Table 2. ³ Calculated.

Table 4. The mean estimate (*k*) of metabolic energy for daily maintenance plus growth based on metabolic body weight in 2-4 wk of age

Age	2-4 week Means*±SD (n = 10)
Body weight (g) ¹	465.6±41.2
Intake (g/bird/d)	47.1±6.9
<i>k</i> (kcal ME/kg BW ^{0.75} per day)	272.2±22.1

* Calculated from the average data of each bird for fourteen days.

¹ The body weight (gram) of 2 (initial), 3 or 4 (final) wk of age was 242.4±23.0, 424.5±39.7 or 667.3±61.1, respectively.

energy for daily maintenance plus growth based on MBW was calculated as 272.2±22.1 kcal for 2-4 wk of age (Table 4), and this datum would be cited as a constant to compute daily feed intake for each chick in Exp 3.

Exp. 2

The growth performance including body weight gain, intake and feed efficiency of chicks receiving diets containing no less than 100% of all essential amino acid requirements for broilers recommended by the NRC (1994) was significantly better than those fed 80 or 60% (Table 5).

A quadratic equation acquired by the regression of feed efficiency on the different percentages of total essential amino acid requirements was as $y = 0.000266x^2 - 0.065464x + 5.506857$ ($r^2 = 0.997$) and a point of inflection was computed at (123, 1.48) so that the individual essential amino acid levels of the control diet prepared in Exp 3 were set at 123% of the corresponding requirements recommended by the NRC (1994).

Exp. 3

The linear regression of body nitrogen mass on body weight of the initial chicks produced an equation, $y = 0.03232x - 1.11019$ ($r^2 = 0.901$), for estimating the initial body nitrogen data of the twenty one groups and their retention rates of body nitrogen were computed and shown in Table 6. The x coordinate of an intersection for each straight line, resulting from the daily intake of the limiting amino acid and the retention rates for body nitrogen based on MBW of every two deficient groups with the same limiting amino acid but at different levels, with a horizontal line, from the datum of the control group, was shown in Table 6 representing the daily requirement for the corresponding essential amino acid based on MBW for

Table 5. The effect of percentages of all essential amino acid requirements recommended by NRC (1994) on the growth performances of male Taiwan country chicks

Treatment (%) ¹	60	80	100	120	140
Ingredients	Means*±SD				
Body weight gain (g/d) n = 20	10.4±3.8 ^a	18.0±4.3 ^b	27.1±3.3 ^c	27.7±3.7 ^c	28.2±4.0 ^c
Intake (g/bird/d) n = 5	26.1±3.8 ^a	34.7±3.9 ^b	43.6±3.2 ^c	42.0±1.2 ^c	43.3±4.3 ^c
Feed efficiency n = 5	2.55±0.25 ^a	1.95±0.15 ^b	1.61±0.12 ^c	1.52±0.08 ^c	1.54±0.04 ^c

¹ The same as that of the Table 2. * Calculated from the average data of each bird for fourteen days.

The initial body weight (2 wk of age) was 243.2±20.1 gram.

Table 6. The effects of individual limiting amino acids on the body nitrogen retention rates of male Taiwan country chicks

Limiting amino acid	Intake (limiting amino acid g/kg BW ^{0.75} per d) n = 6		Retention rate (nitrogen g/kg BW ^{0.75} per d) n = 6		Slope	X-intercept	X-coordinate of intersection ¹
	Level 1	Level 2	Level 1	Level 2			
	Lysine	0.77	0.65	1.47±0.06			
Arginine	0.77	0.62	1.39±0.13	1.09±0.09*	2.02	0.082	0.87
Methionine	0.57	0.48	1.32±0.08	1.08±0.07*	2.55	0.052	0.67
Histidine	0.22	0.17	1.24±0.12	0.95±0.16*	5.76	0.005	0.28
Tryptophan	0.13	0.10	1.35±0.06	1.11±0.15*	11.18	0.004	0.15
Leucine	0.73	0.61	1.33±0.05	1.10±0.04*	2.02	0.070	0.86
Phenylalanine	0.83	0.67	1.16±0.10	0.93±0.11*	1.46	0.032	1.12
Isoleucine	0.49	0.40	1.34±0.09	1.07±0.07*	3.08	0.053	0.57
Threonine	0.49	0.41	1.42±0.08	1.17±0.11*	3.25	0.051	0.54
Valine	0.54	0.46	1.30±0.05	1.10±0.07*	2.53	0.027	0.65

* Significantly different from the level 1.

¹ The intersection point of a straight line, resulting from the two deficient groups with the same limiting amino acid but at different levels, with a horizontal line, $y = 1.58$, from the control group.

Table 7. The optimum patterns of dietary essential amino acids for growth plus maintenance (relative to Lys) for Taiwan country chicks compared with data cited from literature

	Breed	Lysine	Arginine	Sulfur amino acids	Histidine	Tryptophan	Leucine	Aromatic amino acids	Isoleucine	Threonine	Valine
Growth plus maintenance current study	Male Taiwan country chicks	100	105	81	34	18	103	135	69	65	79
Baker and Han (1994)	New Hampshire\ Columbian male chicks	100	105	72	32	16	109	105	67	67	77
Baker et al. (2002)	New Hampshire\ Columbian Plymouth Rock male chicks	100				16.6			61.4	55.7	77.5
NRC (1994)	Broilers	100	114	82	32	18	109	122	73	73	82
Sklan and Noy (2005)	Male Ross\ Ross broilers	100 ¹	113		29		103	107	90	73	86
		100 ²	133		18		123	119	91	87	100

¹ For the second week after hatching. ² For the third week after hatching.

growth plus maintenance. The x-intercept and the reciprocal of slope for each straight line stood for the respective requirements for maintenance and for growth corresponding to the examined amino acid (Table 6). Ratios among essential amino acids for the x coordinate of the intersection, the x-intercept and the reciprocal of the slope can be considered the optimum patterns for growth plus maintenance, for maintenance and for growth, respectively. The result expressed with respect to lysine is shown in Table 7 and 9.

DISCUSSION

It is important to establish the requirements for individual essential amino acids for raising Taiwan country chicks considering that less nitrogen will be excreted and more nitrogen will be deposited when the dietary levels for individual essential amino acids are close to their corresponding requirements. The current study determined the requirements for all essential amino acids simultaneously. In the past only a handful of literature focused on the requirements for individual amino acids, including lysine (Hsu and Lin, 1990), methionine (Hsu and Lin, 1989) and tryptophan (Lin and Hsu, 1994; Chen and Hsu, 1996). The methodology chosen for measuring these essential amino acids was based on dose-response reactions and feeding *ad libitum*. Because the anterior piriform cortex of the brain would detect a decline in the levels of free limiting amino acids in plasma, release neurotransmitters to regulate ventromedial hypothalamus and then lead to a decrease in intake (Gietzen et al., 1998), chicks receiving diets deficient in essential amino acids showed lower feed intake and poorer responses, i.e. the depressed growth performance resulted from the limiting amino acid but also deficiencies in other nutrients. This would affect the regression coefficient and the x-coordinate of an intersection of two regressed straight lines when a broken line model was introduced. Although Edwards and Baker (1999) adopted another approach by maintaining all other

essential amino acids at minimized excess levels that were 15% as an ideal pattern above various doses of sulfur amino acids to reduce the impact on intake due to amino acid imbalance, their data showed that feed intake decreased linearly as sulfur amino acid levels decreased. Intubation was applied in the current study to make sure every chick could receive sufficient feed according to its daily MBW. This technique is time and labor consuming and confined the number of treatment groups and chicks per group while the intake depression resulting from limiting amino acids could be avoided. This was also observed in a previous work conducted on pigs (Chang and Wei, 2005).

The current daily requirements based on MBW for growth plus maintenance for each essential amino acid was derived from the x coordinate value of an intersection of a corresponding straight line from two deficient groups and the horizontal line from the control group. This design was modified from the work done on swine by Wang and Fuller (1989) in which 20% of an essential amino acid was removed from a diet with all other essential amino acids in excess and nitrogen retention and amino acid intake were related to compute the requirements for individual essential amino acids for the growing pigs receiving the control diet. Although Gahl et al. (1995) criticized this approach neglecting diminishing returns which were observed in the experiments conducted in growing rats (Gahl et al., 1991) and swine (Gahl et al., 1995), a number of studies demonstrated that the relationships between the deposition of protein and/or limiting amino acids over a wide range of dose levels and the protein and/or limiting amino acids intake were linear models regardless if the animals were pigs (Batterham et al., 1990; Chung and Baker, 1992; Bikker et al., 1994; Adeola, 1995), rats (Hegsted and Neff, 1970) or chicks (Baker, 1991; Burnham and Gous, 1992; Baker et al., 1996; Edwards et al., 1997; Kim et al., 1997a, b, c; Edwards and Baker, 1999; Edwards et al., 1999; Sklan and Noy, 2005). Based on the linear models demonstrated and the afore-mentioned intubation limitation, only two deficient levels for individual essential amino acids were

Table 8. The dietary requirements of essential amino acids based on percentage in feed computed from the multiplication of the optimum pattern by the dietary requirement of lysine, methionine or tryptophan resulting from dose-response trials conducted by feeding *ad lib.* for male Taiwan country chicks

Essential amino acid (g/kg)	Lysine	Arginine	Sulfur amino acids	Histidine	Tryptophan	Leucine	Aromatic amino acids	Isoleucine	Threonine	Valine
Current study based on lysine (Hsu and Lin, 1990)	10.8	11.3	8.8	3.6	1.9	11.2	14.6	7.4	7.0	8.5
Current study based on sulfur amino acids (Hsu and Lin, 1989)	11.5	12.1	9.4	3.9	2.0	11.9	15.6	7.9	7.5	9.1
Current study based on tryptophan (Lin and Hsu, 1994)	11.9	12.5	9.7	4.0	2.1	12.3	16.0	8.2	7.7	9.4
NRC (1994) for broilers	11.0	12.5	9.0	3.5	2.0	12.0	13.4	8.0	8.0	9.0

conducted in the current study.

The optimum dietary essential amino acid pattern expressed with respect to lysine in the current study was similar to that observed by Baker and Han (1994), except for higher values for methionine and phenylalanine in the current pattern (Table 7). The chicks tested in both experiments belonged to slow-growing breeds. If the dietary level for individual essential amino acids recommended by the NRC (1994) for fast-growing broilers was converted into a pattern relative to lysine, the values for arginine, isoleucine, threonine and valine were higher in comparison with the pattern reported by Baker and Han (1994) and the current study. In addition, the datum for methionine in the NRC pattern was similar while the phenylalanine value was smaller compared with the current pattern. A similar tendency was also observed in converted patterns originating from broiler data reported by Sklan and Noy (2005). Although the daily requirements for individual essential amino acids for growth plus maintenance based on MBW were set up in the current study, those data must be transformed into dietary requirements based on the percentage in feed in practice so that the optimum pattern must be multiplied by the requirement for a single essential amino acid resulting from dose-response trials. The requirements (g/kg feed) for all essential amino acids were computed and shown in Table 8. Discrepancies exist, however, in the results from the multiplication of the current pattern using the dietary requirement for lysine, methionine or tryptophan cited from the literature (Hsu and Lin, 1989; 1990; Lin and Hsu, 1994). This is probably due to the previously mentioned inevitable inaccuracy in dose-response methodology conducted by feeding *ad libitum*. Differences also occurred between the dietary data cited from the NRC (1994) for fast-growing broilers and the data calculated using the slow-growing Taiwan country chicks. Although Han and Baker (1991) reported that the

requirement for lysine based on dietary percentage for a slow-growing strain was roughly the same as that for fast-growing one, the optimum dietary essential amino acid pattern for broilers computed from the data observed by Sklan and Noy (2005) was different from that for slow-growing breeds reported by Baker and Han (1994) and the current study. Even the same broilers at different stages needed different optimum patterns (Table 7) according to the converted data cited from the same report (Sklan and Noy, 2005). Fuller et al. (1989) were of the opinion that this requirement type concerning the sum of growth and maintenance can only be considered strictly applicable to one particular rate of growth, i.e. if the requirements for individual essential amino acids for any accretion rate for whole body protein need predicting, the respective requirements for individual essential amino acids for growth and for maintenance must be set up simultaneously and these two data recombined for any given rate for protein retention. This was conducted in the current study and the requirement for lysine, for example, was computed as 536.1 mg if the body protein accretion rate of Taiwan country chickens weighing 1 kg was set at 6.25 g/MBW ($\text{kg}^{0.75}$) per day.

The differences in the maintenance requirements for individual essential amino acids existed within the literature or the current study and literature (Table 9). One interpretation would be different response criteria. The approach and response criterion used in this study were based on the concept that when a single amino acid is deficient in the diet the body nitrogen accretion rate is directly associated with that one amino acid (Fuller et al., 1989). Gahl et al. (1995) deemed that the accretion rate of a limiting amino acid was more appropriate than the body nitrogen retention or nitrogen balance to reflect dose-response effects. Edwards and Baker (1999) supported such an opinion as well. Gahl et al. (1995) also mentioned,

Table 9. Daily requirements for maintenance or for growth based on metabolic body weight (MBW) for Taiwan country chicks compared with data cited from literature

	Breed	Lysine	Arginine	Sulfur amino acids	Histidine	Tryptophan	Leucine	Aromatic amino acids	Isoleucine	Threonine	Valine
Growth (mg/protein accreted g)	Male Taiwan country chicks	80.0	79.3	62.8	27.8	14.3	79.3	109.5	51.9	49.2	63.3
Maintenance (mg/MBW kg ^{0.75} per day)	Male Taiwan country chicks	36.1	81.9	51.8	4.7	4.2	70.0	31.6	52.6	50.9	26.9
	Edwards et al. (1999) ^{1,2} ; Edwards and Baker (1999) ³ ; Edwards et al. (1997) ² ; Baker et al. (1996) ²	45 ¹	6.9 ²							39.2 ²	32.4 ²
	Peterson/Hubbard male broilers ³			9.4 ³							
	Skalan and Noy (2005)	32	28		11		29	27	31	22	23
	Kim et al. (1997a, b, c)	344		37		29					
	Leveille and Fisher (1959, 1960); Leveille et al. (1960)	0*	81*	58*	0*	10*	81*	57*	73*	82*	82*
Maintenance (mg/BW kg per day)	Cockerels								60		
	Burnham and Grous (1992)										

* Data cited from Fuller et al. (1989) originating from Leveille and Fisher (1959, 1960) and Leveille et al. (1960).

however, that nitrogen gain may be a second best choice considering the limitations of the necessary expense and time for analyzing amino acids. Another reason would be different chicken breeds. Edwards et al. (1999) compared the lysine maintenance requirements between two strains and observed that the chicks growing more slowly needed more lysine based on MBW for maintenance. Similarly, the sulfur amino acids (Edwards and Baker, 1999) and threonine (Edwards et al., 1997) required for maintenance for fast-growing broilers were less in comparison with the current data except for valine (Baker et al., 1996). Compared to another fast-growing broiler data reported by Skalan and Noy (2005), the Taiwan country chicks needed more individual essential amino acids for maintenance except for histidine.

The current study established an optimum dietary pattern concerning essential amino acids for Taiwan country chicks. These data could be transformed into dietary requirements based on percentage (g/kg feed) by multiplying the requirement for a single essential amino acid resulting from dose-response trials conducted by intubation feeding. If the different body protein accretion rates are considered, the respective requirements for growth and for maintenance can be combined and converted into dietary requirements based on percentage by feed intake.

ACKNOWLEDGMENT

The authors are grateful to Dr. Zhirong Jiang and Ajinomoto Co. for their kindness in providing crystalline amino acids and we are indebted to Prof. W. C. Chang

(National Taiwan University) for his technique assistance in measuring amino acids.

REFERENCES

- Adeola, O. 1995. Dietary lysine and threonine utilization by young pigs: Efficiency for carcass growth. *Can. J. Anim. Sci.* 75:445-452.
- Agricultural statistics yearbook 2007. Council of Agriculture, Executive Yuan, Taiwan Government, Taipei, Taiwan.
- AOAC 1984. Official methods of analysis, 14th edn. Association of Official Analytical Chemists, Arlington, Virginia.
- Baker, D. H. 1991. Partitioning nutrients for growth and other metabolic functions: Efficiency and priority considerations. *Poult. Sci.* 70:1797-1805.
- Baker, D. H. and Y. Han. 1994. Ideal amino acid profile for chicks during the first three weeks posthatching. *Poult. Sci.* 73:1441-1447.
- Baker, D. H., S. R. Fernandez, C. M. Parsons, H. M. Edwards, III, J. L. Emmert and D. M. Webel. 1996. Maintenance requirement of valine and efficiency of its use above maintenance for accretion of whole-body valine and protein in young chicks. *J. Nutr.* 126:1844-1851.
- 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.
- Batterham, E. S., L. M. Andersen, D. R. Baigent and E. White. 1990. Utilization of ileal digestible amino acids by growing pigs: Effects of dietary lysine concentration on efficiency of lysine retention. *Br. J. Nutr.* 64:81-94.
- Bender, A. E. 1965. The balancing of amino acid mixtures and proteins. *Proceedings of the Nutrition Society* 24:190-197.
- Bikker, P., M. W. A. Verstegen, R. G. Campbell and B. Kemp. 1994.

- Digestible lysine requirement of gilts with high genetic potential for lean gain, in relation to the level of energy intake. *J. Anim. Sci.* 72:1744-1753.
- Burnham, D. and R. M. Gous. 1992. Isoleucine requirements of the chicken: requirement for maintenance. *Br. Poult. Sci.* 33:59-69.
- Chang, Y. M. and H. W. Wei. 2005. The effects of dietary lysine deficiency on muscle protein turnover in postweanling pigs. *Asian-Aust. J. Anim. Sci.* 18:1326-1335.
- Chen, J. M. and J. C. Hsu. 1996. Effects of dietary tryptophan and niacin levels on growth performance of Taiwan country chicks during 0 to 4 weeks of age. *J. Chin. Soc. Anim. Sci.* 25 (Suppl.):142(Abstr.).
- Chen, K. L., H. J. Chang, C. K. Yang, S. H. You, H. D. Jenq and B. Yu. 2007. Effect of dietary inclusion of dehydrated food waste products on Taiwan native chicken. *Asian-Aust. J. Anim. Sci.* 20:754-760.
- Cheng, F. Y., C. W. Huang, T. C. Wan, Y. T. Liu, L. C. Lin and C. Y. Lou Chyr. 2008. Effect of free-range farming on carcass and meat qualities of black-feathered Taiwan native chicken. *Asian-Aust. J. Anim. Sci.* 21:1201-1206.
- Chung, T. K. and D. H. Baker. 1992. Efficiency of dietary methionine utilization by young pigs. *J. Nutr.* 122:1862-1869.
- Edwards, H. M., III, D. H. Baker, S. R. Fernandez and C. M. Parsons. 1997. Maintenance threonine requirement and efficiency of its use for accretion of whole-body threonine and protein in young chicks. *Br. J. Nutr.* 78:111-119.
- Edwards, H. M., III, and D. H. Baker. 1999. Maintenance sulfur amino acid requirements of young chicks and efficiency of their use for accretion of whole-body sulfur amino acids and protein. *Poult. Sci.* 78:1418-1423.
- Edwards, H. M., III, S. R. Fernandez and D. H. Baker. 1999. Maintenance lysine requirement of young chicks and efficiency of its use for accretion of whole-body lysine and protein. *Poult. Sci.* 78:1412-1417.
- Fan, Y. K. and Y. P. Lee. 1984. The effects of dietary nutrient density and protein energy ratio on the performances of the three varieties of meat-type chicken in Taiwan. *J. Chin. Soc. Anim. Sci.* 13:1-12.
- Fuller, M. F., R. McWilliam, T. C. Wang and L. R. Giles. 1989. The optimum dietary amino acid pattern for growing pigs: 2. Requirements for maintenance and for tissue protein accretion. *Br. J. Nutr.* 62:255-267.
- Gahl, M. J., M. D. Finke, T. D. Crenshaw and N. J. Benevenga. 1991. Use of a four-parameter logistic equation to evaluate the response of growing rats to ten levels of each indispensable amino acid. *J. Nutr.* 121:1720-1729.
- Gahl, M. J., T. D. Crenshaw and N. J. Benevenga. 1995. Diminishing returns in weight, nitrogen, and lysine gain of pigs fed six levels of lysine from three supplemental sources. *J. Anim. Sci.* 73:3177-3187.
- Gietzen, D. W., L. F. Erecius and Q. R. Rogers. 1998. Neurochemical changes after imbalanced diets suggest a brain circuit mediating anorectic responses to amino acid deficiency in rats. *J. Nutr.* 128:771-781.
- Han, Y. and D. H. Baker. 1991. Lysine requirements of fast- and slow-growing broiler chicks. *Poult. Sci.* 70:2108-2114.
- Hegsted, D. M. and R. Neff. 1970. Efficiency of protein utilization in young rats at various levels of intake. *J. Nutr.* 100:1173-1180.
- Hsu, A. and R. P. Lin. 1989. Sulfur amino acid requirement of small type country chick in Taiwan. *J. Chin. Soc. Anim. Sci.* 18:13-20.
- Hsu, A. and R. P. Lin. 1990. Lysine requirement of small type country chicken in Taiwan. Proceeding of the 4th Conference of WPSA. Far East and South Pacific Federation. P37. Taipei, Taiwan, ROC.
- Huang, C. C., C. C. Hsieh and S. H. Chiang. 2007. Estimating the energy partitioning of Taiwanese native chickens by mathematical model. *Anim. Feed Sci. Technol.* 134:189-197.
- Kim, J. H., W. T. Cho, C. J. Yang, I. S. Shin and I. K. Han. 1997a. Partition of amino acids for maintenance and growth of broilers I. Lysine. *Asian-Aust. J. Anim. Sci.* 10:178-184.
- Kim, J. H., W. T. Cho, C. J. Yang, I. S. Shin and I. K. Han. 1997b. Partition of amino acids for maintenance and growth of broilers II. Methionine. *Asian-Aust. J. Anim. Sci.* 10:277-283.
- Kim, J. H., W. T. Cho, C. J. Yang, I. S. Shin and I. K. Han. 1997c. Partition of amino acids for maintenance and growth of broilers III. tryptophan. *Asian-Aust. J. Anim. Sci.* 10:284-288.
- Langer, S. 1995. Amino acid interactions in pigs. Ph. D. Thesis, University of Aberdeen, Scotland, UK.
- Leveille, G. A. and H. Fisher. 1959. Amino acid requirements for maintenance in the adult rooster: II. The requirements for glutamic acid, histidine, lysine and arginine. *J. Nutr.* 69:289-294.
- Leveille, G. A. and H. Fisher. 1960. Amino acid requirements for maintenance in the adult rooster: III. The requirements for leucine, isoleucine, valine and threonine, with reference also to the isomers of valine, threonine and isoleucine. *J. Nutr.* 70:135-140.
- Leveille, G. A., R. Shapiro and H. Fisher. 1960. Amino acid requirements for maintenance in the adult rooster: IV. The requirements for methionine, cystine, phenylalanine, tyrosine and tryptophan; the adequacy of the determined requirements. *J. Nutr.* 72:8-15.
- Lin, Y. F. and A. Hsu. 1994. Tryptophan requirement of country chicks in Taiwan. *J. Chin. Soc. Anim. Sci.* 23:121-126.
- Moore, S. 1963. On the determination of cystine as cysteic acid. *J. boil. Chem.* 238:235-237.
- National Research Council 1994. Nutrient Requirements of Poultry, 9th ed. Natl. Acad. Sci., Washington DC.
- SAS Institute, Inc. 1988. SAS/STAT User's guide. Version 6.03 edn. SAS Institute Inc., Cary, North Carolina.
- Shih, B. L., J. C. Hsu, Y. K. Fan, P. W. S. Chiou and B. Yu. 1993. The effects of dietary calcium levels on the growth performance of Taiwan country chickens during 0 to 4 weeks of age. *J. Chin. Soc. Anim. Sci.* 22:119-130.
- Sklan, D. and Y. Noy. 2005. Direct determination of optimal amino acid intake for maintenance and growth in broilers. *Poult. Sci.* 84:412-418.
- Wang, T. C. and M. F. Fuller. 1989. The optimum amino acid pattern for growing pigs: 1. experiments by amino acid deletion. *Br. J. Nutr.* 62:77-89.
- Wei, H. W. and M. Fuller. 2006. Dietary deficiencies of single amino acids: whole-body amino acid composition of adult rats. *Arch. Anim. Nutr.* 60:119-130.

- Yu, W. H. and A. Hsu. 1989. Protein and energy requirements of small type country chickens in Taiwan. *J. Chin. Soc. Anim. Sci.* 18:1-11.
- Yu, W. H., R. P. Lin and A. Hsu. 1990. Calcium and available phosphorus requirements of small type country chickens in Taiwan. *J. Chin. Soc. Anim. Sci.* 19:107-116.