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Comparison of Different Ideal Amino Acid Ratios in Male and Female Broiler Chickens of 21 to 42 Days of Age

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An amino acid bioassay was carried out to compare responses of male and female broiler chicks to different ideal amino acid (AA) ratios from 21 to 42 d of age. The ideal ratios were used to calculate AA requirements were IICP (Illinois Ideal Chick Protein), NRC (1994), RPAN (Rhone Poulenc Animal Nutrition) and feedstuff AA ratios which were compared with each other and with a positive control diet. Indispensable AAS were rationed to lysine according to requirement ratios in tested profiles, with digestible lysine set at 0.85 and 0.78 for male and female, respectively. Experimental period began at day 21 and lasted in 42 d of age. Diets for all profiles contained 3200 kcal AME_n/kg, and L-glutamic acid was used to make all diets equal in crude protein at 14.25% of the diet. Chicks fed diets formulated with RPAN had significantly lower weight gain and feed efficiency than IICP, NRC and feedstuff which is probably consequence of high Lys: Leu ratio in this profile. There were no significant differences in feed intake, weight gain and feed: gain among chicks fed IICP, NRC (1994) and feedstuff. Since IICP ratios in the case of all AAS (except Met+Cys in NRC) are lower than or close to the ratios in NRC (1994) and feedstuff, it can be concluded that IICP ideal ratios is sufficient for supporting maximal weight gain and feed efficiency. Chicks fed diets which their AA requirements calculated by feedstuff ratios had higher breast meat yield than IICP and NRC (1994) which may be due to higher Lys: Met+Cys ratio in this profile. Results of this experiment suggest that although male and female broiler chicks have different body composition and consequently different AA requirements, but they respond similarly to different ideal AA ratios.

Key words: amino acid profile, broiler, ideal protein

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Introduction

Growing concerns about the environmental impact of animal production may lead to addition of environmental factors such as nitrogen and phosphorus pollution as parameters in feed formulation schemes in the future. Since ideal protein (or ideal amino acid) concept provide a precise ratio of AAS and minimizes nitrogen excretion, it can play an integral role in precision nutrition. AA requirements can be influenced by a variety of dietary (e.g., protein level, energy level, presence of protease inhibitors), environmental (e.g., disease, crowding, feeder space, heat and cold stress) and genetic factors (e.g., sex and capacity for lean vs. fat growth). According to ideal amino acid ratios, although amino acid requirements change due to mentioned factors, ideal ratios, unlike to requirements, would not change. In this concept lysine was chosen as reference AA and all the other AAS expressed as a percentage of lysine. Lysine was chosen because it is used almost exclusively for protein accretion after absorption (contrary to the other amino acids), it is a limiting AA in reduced protein corn-soybean meal broiler diets, and the analysis for Lys is uncomplicated (Baker, 1997; Emmert and Baker, 1997; Baker and Han, 1994). Most of the works has been done, on ideal AA ratios with young birds from hatching to about 3 weeks of age whereas the database for broilers from third to sixth weeks of age is still rather limited. With respect to lack of accurate requirement informations for most AAS in the later stages, using ideal ratios for predicting AA requirements may be more beneficial. When birds advance in age (or weight), both AA requirements and ideal ratios would change with aging. This was assumed primarily because it seemed probable that as birds age, maintenance needs for AA such as methionine, cystine, theronine, tryptophan, valine, arginine and isoleucine would increase faster than maintenance needs for lysine (Emmert and Baker, 1997). Maintenance needs as a percentage of total AA requirements are minimal (probably 3 to 6%) for young birds, but they increase substantially as birds advance in age and weight. However, research using pigs and rats has long led to the belief that the percentage of lysine required for maintenance is very small and does not increase substantially as birds grow and age. Thus the ideal ratios of those amino acids contributing significantly to maintenance (e.g. methionine, cystine, theronine etc)

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apparently increase with birds' age. We previously (Taherkhani *et al.,* 2005) compared different ideal AA ratios during the second and third weeks posthatch, but there has been not made any comparison in the later stage of chicks' life, so the main objective of this study was to compare different ideal AA ratios in male and female (because of different body composition of male and female) broiler chicks in the period of third to sixth weeks posthatch.

Materials and Methods

General procedures

400 feathers sexed male and female broiler chicks (Ross 308) were fed a commercial corn soybean meal (3200 kcal AME_n/kg, 1.4% total Lys and 1.02% TSAA) from hatching to day 21 posthatching. Then after being subjected to an overnight period of feed withdrawal, chicks were weighed individually and randomly assigned to battery pens so that pens have equal initial weight and weight distribution. Experiment started at 21 d of age and lasted in 42 d of age. Feed intake and weight gain were measured for each pen at day 42 posthatch and then feed: gain was calculated. At day 42 ten birds per each treatment were obtained for measuring carcass dissections (i.e. abdominal fat, breast meat yield and liver weight).

Diet

AA requirements were estimated by using 1) digestible Lys requirement of 0.85 and 0.78 for male and female, respectively which were reported by Han and Baker (1994) and 2) ideal AA ratios in tested profile including IICP (Illinois Ideal Chick Protein), NRC (1994), RPAN (Rhone Poulenc Animal Nutrition) and Feedstuff AA ratios (Table 1). Except for NRC (1994), all the other ratios depicted, are based on true digestible levels of dietary AA. AA content of corn and soybean meal were analyzed by HPLC and then digestible content of AA was calculated using RPAN AA digestibility coefficient. Synthetic AAS (12 AA) were used to exactly meet the AA requirements in each profile. All AAS supplied as L-isomers except methionine, which was supplied as DL-isomers.

Feed-grade sources were used for lysine HCL (98.5%) and methionine (99%); the remaining AAS were pharmaceutical grade. The true digestibility of free AA was assumed to be 100%. All the diets were kept isonitrogenous (by varying levels of glutamate) and isocaloric (3200 kcal AME_n/kg of diet). Also, when profiles were compared lysine level remained constant. All the diets were checked to have at least 0.3% of diet proline and 0.6% of diet glycin. A positive control diet was also formulated using NRC (1994) recommendations (3200 kcal ME, 1.1% Tot Lys and 0.9% SAA). The diets used in this bioassay are presented in Table 2. Four battery pens of ten chicks were fed one of four different profiles or positive control diet in both sexes.

Statistical procedures

Pens were identified as the experimental units. Weight gain (WG), feed intake, feed: gain and carcass dissection data were statistically evaluated by the analysis of variance procedure of SAS software (1996), involving a factorial arrangement of main factor (AA ratios and sex) in a randomized complete design. Significant differences between means were separated by the GLM procedure of SAS software (1996). Statistical significance was considered $P < 0.05$.

Table 1. Amino acid ratios in IICP (Illinois ideal chick protein), NRC 1994, RPAN 1993 (Rhone Poulenc Animal Nutrition) and Feedstuff (1999)

Amino Acid	IICP	NRC	RPAN	Feedstuff
Lys	100	100	100	100
Met+Cys	75	72	81	80
Met	37	38	48	46
Thr	62[1]	74	67	62
Val	80	82	85	89
Arg	108	110	108	110
Trp	17	18	19	18
Ile	69	73	75	75
Leu	109	109	144	116
His	35	32	35[2]	32
Phe+Tyr	105	122	105[2]	128

[1] Baker *et al.* (2002) reported 100:55.7 for Lys: Thr for the second and third weeks posthatch, with respect to this matter that Lys: Thr ratio may increase only slightly with advancing age (Baker, 2003), we used 62 instead of 70 for Lys: Thr in IICP.

[2] RPAN did not specify requirements for this AA, so the ratios used in IICP was used also for RPAN.

Results and Discussion

Feed intake, feed: gain and WG are presented in Table 3. Results of this study show that broiler chicks fed positive control diet had higher feed intake, WG and better feed: gain than those received low protein, AA-supplemented diets in both male and female (Table 3). There are numerous studies have compared practical diets with low crude protein diets supplemented with crystalline AAS. Edmonds *et al.* (1985) found that performance of young chickens was not maintained when protein was decreased from 245 to 160 g/kg diet with appropriate AAS supplementation. Moreover, Pinchasov *et al.* (1990) found a decline in performance of young chick with the decrease in dietary protein which did not compensate by AA supplementation. Holsheimer and Janssen (1991) and Kerr and Kidd (1999) came to similar conclusions. Lower performance in low protein AA-supplemented diets observed here may be mainly attributed to higher feed intake in positive control diet (Table 3) due to better texture and palatability of such conventional diet than purified and semi-purified diets.

When comparisons were made among chicks fed diets formulated with different ideal amino acid ratios, the most significant ($P < 0.05$) differences observed between RPAN and the other ratios. RPAN had lowest feed intake and WG ($P < 0.05$) and it had also an impaired gain: feed ($P < 0.05$) relative to all the other ratios in both sexes. It seems

Table 2. Composition of experimental diets

Feed Ingredient (%)	IICP		NRC		Feedstuff		RPAN	
	Male	Female	Male	Female	Male	Female	Male	Female
Corn	76.19	78.08	76.38	76.25	75.43	76.63	80.13	80.88
Soybean Meal	11.19	7.85	11.19	7.85	12.03	7.85	9.85	4.84
Dicalcioum Phosphate	2.04	2.38	2.04	2.38	2.03	2.38	2.04	2.10
Oyster shell	1.36	1.23	1.37	1.23	1.37	1.46	1.38	1.38
Sun Flower Oil	1.50	1.50	1.50	0.92	1.70	1.50	0.70	—
$NaHCO_3$	0.56	0.74	0.66	0.66	0.66	0.70	0.66	0.66
Min. premix[1]	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.30
Vit. premix[2]	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.30
L-Glutamate	2.42	4.93	1.39	3.69	2	4.55	0.81	3.86
L-Lys.HCL	0.87	0.85	0.86	0.85	0.84	0.85	0.84	0.93
DL-Met	0.42	0.39	0.49	0.45	0.45	0.41	0.48	0.48
L-Thr	0.65	0.35	0.79	0.72	0.38	0.39	0.28	0.64
L-Arg	0.68	0.65	0.77	0.74	0.69	0.68	0.63	0.83
L-Leu	0.59	0.57	0.56	0.56	0.51	0.51	0.43	1.04
L-Val	0.39	0.38	0.44	0.43	0.43	0.43	0.40	0.49
L-Ile	0.23	0.21	0.35	0.31	0.28	0.27	0.42	0.39
L-Phe	0.06	0.06	0.24	0.22	0.28	0.3	0.02	0.21
L-Gly	0.19	0.32	0.18	0.31	0.16	0.32	0.16	0.4
L-Trp	0.07	0.07	0.09	0.09	0.11	0.11	0.10	0.11
L-His	0.11	0.11	0.08	0.08	0.05	0.05	0.10	0.16
Calculated Nutrient, % (unless noted otherwise)								
ME (kcal/kg)	3200	3200	3200	3200	3200	3200	3200	3200
CP	16.25	16.25	16.25	16.25	16.25	16.25	16.25	16.25
Ca	1	1	1	1	1	1	1	1
Ava. Phosphorus	0.45	0.45	0.45	0.45	0.45	0.50	0.45	0.45
Cl	0.25	0.20	0.20	0.20	0.20	0.20	0.20	0.21
Na	0.20	0.22	0.20	0.20	0.20	0.21	0.20	0.20
Lys	0.85	0.78	0.85	0.78	0.85	0.78	0.85	0.78
Met+Cys	0.64	0.58	0.61	0.58	0.68	0.62	0.68	0.63
Met	0.49	0.45	0.43	0.45	0.49	0.46	0.51	0.49
Thr	0.53	0.48	0.63	0.58	0.53	0.48	0.57	0.53
Trp	0.15	0.13	0.15	0.13	0.15	0.14	0.16	0.14
Arg	0.92	0.84	0.93	0.84	0.93	0.86	0.91	0.84
Ile	0.59	0.54	0.62	0.54	0.64	0.58	0.63	0.58
Leu	0.93	0.85	0.93	0.85	0.99	0.90	1.22	1.12
Val	0.68	0.62	0.70	0.62	0.76	0.70	0.72	0.66
His	0.30	0.27	0.27	0.25	0.27	0.25	0.30	0.27
Phe+Tyr	0.89	0.82	1.03	0.95	1.15	1.05	1.22	0.82
Phe	0.55	0.50	0.63	0.50	0.73	0.69	0.73	0.50
Gly+Ser	1.12	1.12	1.12	1.12	1.12	1.12	0.93	0.86
Gly	0.89	0.95	0.78	0.95	0.73	0.86	0.60	0.70

[1] Provided (per kilogram of diet): $Ca_3(PO4)_2$, 28.0 g; K_2HPO_4, 9.0 g; NaCl, 8.89 g; $MgSO_4$ $7H_2O$, 3.5 g; $ZnCO_3$, 0.10 g; $CaCO_3$, 3.0 g; $MnSO_4$ H_2O, 0.65 g; $FeSO_4$ $7H_2O$, 0.42 g; KI, 40 mg; $CuSO_4$ $5H_2O$, 20 mg; Na_2MoO_4 $2H_2O$, 9 mg; H_3BO_3, 9 mg; $CoSO_4$ $7H_2O$, 1 mg; Na_2SeO_3, 0.22 mg.

[2] Provided (per kilogram of diet): thiamin HCL, 20 mg; niacin, 50 mg; riboflavin, 10 mg; D-Ca-pantothenate, 30 mg; vitamin B12, 0.04 mg; pyridoxine HCL, 6 mg; D-biotin, 0.6 mg; folic acid, 4 mg; menadione dimethylpyrimidinol bisulfate, 2 mg; cholecalciferol, 15 μg; retinyl acetate, 1,789 μg; ascorbic acid, 250 mg.

that inappropriate ratio of Lys: Leu in RPAN amino acid profile (100:144) which is too high in comparison to the other profile is the main and primary cause of lower performance in chicks fed this treatment in both sexes. Adverse effects of AA imbalance on the utilization of nutrient are well documented. Accounts of AA imbalances conventionally focus on the growth-depressing effects in animals (Harper, 1964; Tews *et al.,* 1979). However, it has been consistently recorded that a predisposing factor is a rapid and marked reduction in food intake. Thus Harper and Rogers (1965) reported that rats fed an imbalanced diet reduced their food intake within 3–6 h. These results implied that the depression in food intake was the primary event responsible for ensuing retardation of growth. It expected that with AA imbalance, protein utilization efficiency decrease dramatically. Experiments in rats indicated detrimental effects of AA imbalance on utilization of first limiting AA. Excess AA contributes to AA im-

Table 3. Performance of broiler chicks fed experimental diets

Sex	Ideal ratio	Feed intake (kg/bird)	Feed: Gain	Weight gain (kg)
Male	IICP	2.61±0.467[b]	2.18±0.27[b]	1.20±0.12[b]
	NRC	2.46±0.272[b]	2.22±0.40[b]	1.11±0.03[b]
	Feedstuff	2.49±0.027[b]	2.21±0.18[b]	1.13±0.09[b]
	RPAN	2.11±0.250[c]	2.38±0.32[a]	0.89±0.08[c]
	Control	2.86±0.100[a]	1.91±0.06[c]	1.49±0.02[a]
	Mean	2.49	2.38	1.08
	SE	0.101	0.110	0.066
Female	IICP	1.92±0.24[b]	2.22±0.11[b]	0.85±0.11[b]
	NRC	2.01±0.23[b]	2.37±0.09[ab]	0.84±0.10[b]
	Feedstuff	1.94±0.20[b]	2.31±0.21[ab]	0.83±0.07[b]
	RPAN	1.22±0.12[c]	2.62±0.33[a]	0.47±0.05[c]
	Control	2.70±0.02[a]	2.04±0.09[c]	1.32±0.06[a]
	Mean	1.96	2.31	0.866
	SE	0.110	0.050	0.064

[a–c] Significant differences among treatments within a row ($P<0.05$).

Table 4. Effect of different ideal ratios on carcass dissections

Sex	Ideal ratio	Breast meat[1]	Abdominal fat[1]	Liver weight[1]
Male	IICP	20.64±3.5[c]	2.70±0.03	2.39±0.27[b]
	NRC	20.63±2.0[c]	2.41±0.70	2.37±0.16[b]
	Feedstuff	23.04±1.6[b]	1.74±0.67	2.91±0.47[b]
	RPAN	21.79±2.2[bc]	1.86±0.79	3.37±0.75[a]
	Control	25.98±2.2[a]	1.29±0.65	2.58±0.29[b]
	Mean	22.41	2.00	2.72
	SE	0.43	0.11	0.08
Female	IICP	18.14±1.5[d]	1.44±0.78[a]	2.55±0.15[b]
	NRC	20.57±1.4[bc]	2.43±0.57[a]	2.92±0.37[b]
	Feedstuff	21.9 ±3.5[b]	2.17±0.91[a]	2.58±0.47[b]
	RPAN	19.71±3.9[c]	1.86±0.78[ab]	3.47±0.47[a]
	Control	26.02±2.1[a]	1.61±0.59[b]	2.51±0.45[b]
	Mean	21.26	2.10	2.58
	SE	0.53	0.11	0.07

[1] Yields of Breast meat, Abdominal fat and Liver weight are as percentage of eviscerated carcass.
[a–d] Significant differences among treatments within a row ($P<0.05$).

balance, stimulate AA catabolism pathway which will result in AA, especially, limiting AA catabolism (Kumta *et al.,* 1962). D'Mello and Lewis (1970) reported that supplementation of diet with excess Leu, inhibited response of chicks to first limiting AA, namely Met, and decreased efficiency of protein utilization. The result obtained herein is in agreement with our previous finding (Taherkhani *et al.,* 2005) in which high levels of dietary Leu due to high Lys: Leu ratio (100:150) caused the same reduction in feed intake, WG and feed efficiency during the second and third weeks posthatch.

There were no significant difference among the IICP, NRC (1994) and Feedstuff in any of performance criteria (i.e. feed intake, feed: gain and WG). With respect to lack of significant difference among these AAS profiles and either with regard to lower levels of majority of ideal AAS ratios in IICP (except Lys: Met+Cys in NRC) relative to NRC and Feedstuff, it can be concluded that some of the ideal ratios in NRC and feedstuff may be over-estimated for supporting maximum WG and feed efficiency and can be decreased to the levels recommended by IICP without any negative effect. We recommend that the 100:75 is the optimum ideal ratio for Lys: Met+Cys which may be overestimated in Feedstuff (100:80). The ratio reported for Lys: Met+Cys by Mack *et al.* (1999) and CVB (1996) also support this idea. Mack *et al.* (1999) came to the ratio of 100:75 for Lys: Met+Cys for the period of 20 to 40 d of age and dutch bureau of livestock feeding (1996) reported 100:73 for the period of hatching to 42 d of age.

Based upon the results obtained in Baker *et al.* (2002) experiments which report an ideal ratio of 55.7 for Thr for the period of second to third weeks posthatch and taking account that it should increase only slightly with advancing in age (Baker, 2003) and either with respect to ratio reported by Mack *et al.* (1999) and dutch bureau of livestock feeding (1996) (63 and 65 respectively), it can be argued that Lys: Thr ratio in NRC (1994) (100:74) is too high and can be decreased. The same argument can be made about the ratios of Phe+Tyr in both NRC (1994) and Feedstuff, Val in Feedstuff and Ile in NRC and Feedstuff.

Breast meat yield (BMY) did not respond in the same way as WG and feed: gain. BMY (Table 4) was significantly ($P<0.05$) higher in Feedstuff in comparison to IICP and NRC (1994). It is well documented that AA requirement for supporting maximal breast meat is higher than those required for maximize weight gain. Schutte and Pack (1995) found the TSAA (total sulfur AA) requirement for growing broilers was 0.05 percent greater for BMY and feed conversion in comparison to that for growth. Han and Baker (1994) also found a higher lysine requirement for broiler BMY and FG in comparison to that for body weight. In a study examining the threonine requirement of male turkeys approximately 0.06 percent more threonine was required for BMY (Lehmann *et al.,* 1997). Consideration in sulfur AA and threonine ratios (Table 1) will reveal that the higher ratio of Lys: Met+Cys is the main cause of higher BMY in Feedstuff and that the ratio of sulfur AA in IICP and NRC (1994) may be under-estimated for maximize BMY.

According to Table 4 liver weight in RPAN was significantly higher than those in IICP, NRC (1994) and Feedstuff in both males and females. Higher liver weight observed in RPAN in our study may be attributed to higher level of Leu. Harper and Roger (1965) found that surplus AA arriving in portal circulation after consumption of an imbalanced diet stimulate synthesis or suppress break down of protein in the liver (both will result in increase in liver weight) leading to greater retention of the limiting AA. Greater liver weight is usually associated with lower feed efficiency observed in our study in RPAN (Table 3).

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