Lysine and sulfur amino acid requirements of broiler chicks over short time periods within the starter phase

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Primary Audience: Production Managers, Nutritionists

SUMMARY

Six experiments were conducted to validate the hypothesis that Lys and SAA requirements decrease within the starter phase using 3-d periods from 2 to 11 d of age. In the first 3 experiments, 7 diets were generated by adding L-Lys to a lysine-deficient basal diet in 0.10% increments, ranging from 0.85 to 1.45% digestible Lys. In experiments 4 to 6, and 7 diets were generated by adding 0.07% increments of DL-methionine to a SAA-deficient diet to produce diets ranging from 0.63 to 1.04% DSAA. The linear broken line estimate for digestible Lys was 1.22, 1.17, and 1.16% for BWG and 1.31, 1.21, and 1.14% for FCR in experiments 1, 2, and 3, respectively. The linear broken line estimate for DSAA was 0.82, 0.81, and 0.94% for BWG and 0.82, 0.80, and 0.90 for FCR in experiments 4, 5, and 6, respectively. These results indicated that Lys requirements decreased linearly as hypothesized, however, the SAA requirements did not follow same pattern.

Key words: lysine, sulfur amino acids, requirement, broiler, broken line

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DESCRIPTION OF THE PROBLEM

The starter phase of production is a crucial stage of production requiring access to feed with high concentrations of dietary amino acids due to low feed intake. This increase in amino acids is important to support the growth and development of the systems and organs of the chicks needed to allow for the rapid muscle growth later in life (Schmidt et al., 2009). Dietary amino acid concentrations decrease as birds are transitioned to grower and finisher phases due to increased feed intake. Traditionally experiments have focused on requirement estimations that span an entire phase (Kalinowski et al., 2003; Garcia and Batal, 2005; Dozier et al., 2010; Cemin et al., 2017). However, Emmert and Baker (1997) introduced and validated the concept of using regression equations to predict short-term requirements within each phase to smooth out the large drops in requirements that would be seen during the transition from starter to grower and finisher diets which would ultimately result in dietary savings. In addition, feed disturbances could be minimized by avoiding large changes in diets needed to reduce cost from one phase to the next (Saleh et al., 1997). This was accomplished by changing diets every 1 to 2 d to follow the requirements of the birds more closely according to their age. However, the logistics of formulating and producing feed as well as the transportation of feed every 2 to 3 d would not be ideal or economically viable. To

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solve this issue, authors suggested producing a high- and low-nutrient dense diet and placing them in 2 different bins on the farm then mixing them in different proportions to meet the requirements of broilers based on the regression equations (Pope et al., 2002). However, this concept was put forth with the assumption that amino acid requirements will decrease within each phase without direct validation (Pope et al., 2002). Therefore, the objective of these experiments was to validate the hypothesis that sulfur amino acids (**SAA**) and lysine requirements decrease with age and to determine the Lys and SAA requirements within the starter phase (2−11 d of age) over 3-d periods.

MATERIALS AND METHODS

Diet Formulation

Two starter diets were formulated to be deficient in either Lys or SAA and used as a basal diet to create experimental diets. The Lys deficient diet was formulated to contain 0.85% digestible Lys using corn, corn gluten meal, DDGS, and soybean meal as the major amino acid containing ingredients (Tables 1 and 2). The SAA-deficient diet was formulated to 0.63% digestible SAA via corn, soybean meal, and poultry byproduct meal (Tables 3 and 4). The basal diets were formulated to be adequate in all essential nutrients according to Ross 708 broiler nutritional recommendations with the exception of Lys or SAA in experiments 1 to 3 and 4 to 6, respectively (Aviagen, 2018). The basal diet was then split into equal aliquots. Lysine-HCL was added at 0.10% increments to generate experimental diets that contained 0.85%, 0.95%, 1.05%, 1.15%, 1.25%, 1.35%, and 1.45% Lys and DL-Met were added to the basal diet at 0.07% increments to generate experimental diets that contained 0.63%, 0.70%, 0.77%, 0.83%, 0.90%, 0.97%, and 1.04% DSAA. Basal and experimental diets were sampled when diets were removed from the mixer and a composite feed sample was used for amino acid composition analysis (University of Missouri AESCL, Columbia, MO 65211). Additional starter diet was formulated and manufactured according to Ross 708 broiler nutritional recommendations (Aviagen, 2018) and used to maintain all chicks before they were transitioned to experimental diets. Diets in all experiments were manufactured in a mash form.

Broiler Management

All animal procedures were approved by the Institutional Animal Care and Use Committee at Virginia Tech (Blacksburg, VA). Six independent experiments were conducted for estimating the Lys and DSAA requirements from 2 to 5, 5 to 8, and 8 to 11 d of age. In each experiment, 350 male Hubbard × Ross 708 chicks were weighed, sorted by body weight into 3 body weight ranges to minimize differences in initial cage body weight across treatments. Diets were randomly assigned to the cages with 10 replicates of 5 birds in each battery cage. Chicks were provided ad libitum access to experimental feed and water. Temperature was maintained according to breeder specifications based on the age of the birds which ranged from 30°C at placement to 24°C at 11 d of age (Aviagen, 2018). Continuous lighting was provided from 0 to 3 d of age, and then the lighting was adjusted to provide 20 h of light and 4 h of darkness from 3 to 11 d of age according to the commercial management guide (Ross 708 management guide). Health checks occurred twice daily when any mortality was removed from the cage, weighed, and recorded.

Data and Sample Collection

Individual body weight and pen feed offered and refused were measured at the beginning and end of each experiment. Body weight gain and feed intake were calculated by the difference between final and initial body and feeder weight, respectively. Body weight gain and feed intake were used to calculate mortality corrected feed conversion ratio (**FCRm**) by adding the cage mortality body weight gain to cage bird body weight gain. At the end of each

Table 1. Composition of lysine-deficient basal diets fed to broiler chicks from 2 to 5, 5 to 8, and 8 to 11 d of age, experiments 1 to 3.[1]

Ingredient	Inclusion (%)	Nutrient profile	Formulated (%)	Analyzed[2] (%)
Corn	60.53	Crude protein	22.72	21.72
Soybean meal (47.5%)	22.49	ME (kcal/kg)	3030	—
Corn gluten meal	7.45	Calcium	0.90	—
DDGS	5.00	Available P	0.45	—
Soy oil	0.20	Digestible Met	0.60	0.63 (0.59)
Sodium chloride	0.20	Digestible Cys	0.28	0.35 (0.28)
Sodium bicarbonate	0.10	Digestible SAA	0.88	0.98 (0.89)
L-arginine	0.14	Digestible Lys	0.85	0.92 (0.81)
DL-methionine	0.26	Digestible His	0.48	0.56 (0.51)
L-lysine-HCl	0.00	Digestible Trp	0.19	0.24 (0.21)
L-threonine	0.13	Digestible Thr	0.77	0.83 (0.71)
Limestone	0.95	Digestible Arg	1.24	1.22 (1.15)
Dicalcium phosphate	1.82	Digestible Iso	0.79	0.93 (0.83)
Choline chloride (60%)	0.10			
Vitamin and mineral premix[3]	0.63			

[1]L-lysine was added at 0.10% increments to generate experimental diets that contained 0.85%, 0.95%, 1.05%, 1.15%, 1.25%, 1.35%, and 1.45% lysine.

[2]Calculated amino acid values are reported as total dietary amino acid (digestible amino acid) and analyzed amino acid values are reported as total dietary amino acid. Analyzed total amino acid values were converted to digestible amino acid values using AminoDat Software (Version 5, 2016) by multiplying the digestibility coefficient of each ingredient by the amount of amino acid provided by that ingredient in the diet.

[3]Provided per kg of diet: vitamin A, 1,320,000 IU; vitamin D3, 440,000 ICU; vitamin E, 2,860 IU; menadione, 176 mg; biotin, 6.6 mg; vitamin B12, 1.9 mg; choline, 71.5 g; niacin, 6.6 mg; pantothenic acid, 1.8 g; selenium, 40 mg; riboflavin, 880 mg; Cu, 4.4 g; Fe, 45 g; I, 135 mg; Mn, 44 g; Zn, 44 g; Co, 4.4 g.

experiment, birds were euthanized using cervical dislocation and the breast muscle of all remaining birds was removed and weighed. The breast weight was expressed as a relative breast weight to body weight percentage.

Statistical Analysis

Digestible Lys and SAA requirements were estimated using both the linear and quadratic broken line models based on body weight gain,

Table 2. Formulated and analyzed lysine content of experimental starter diets fed to male broiler chicks over the 2 to 5, 5 to 8, and 8 to 11 d periods, experiment 1 to 3.[1]

	Formulated		Analyzed	
Diet	Total Lys (%)	Digestible Lys (%)	Total Lys (%)	Digestible Lys[2] (%)
Corn-SBM-CGM basal	0.96	0.85	0.92	0.81
Basal + 0.10% L-Lys	1.06	0.95	0.89	0.79
Basal + 0.20% L-Lys	1.16	1.05	1.01	0.89
Basal + 0.30% L-Lys	1.26	1.15	1.26	1.15
Basal + 0.40% L-Lys	1.36	1.25	1.33	1.18
Basal + 0.50% L-Lys	1.46	1.35	1.44	1.28
Basal + 0.60% L-Lys	1.56	1.45	1.47	1.31

[1]L-lysine was added at 0.10% increments to generate experimental diets that contained 0.85%, 0.95%, 1.05%, 1.15%, 1.25%, 1.35%, and 1.45% lysine.

[2]Calculated amino acid values are reported as total dietary amino acid (digestible amino acid) and analyzed amino acid values are reported as total dietary amino acid. Analyzed total amino acid values were converted to digestible amino acid values using AminoDat Software (Version 5, 2016) by multiplying the digestibility coefficient of each ingredient by the amount of amino acid provided by that ingredient in the diet.

Table 3. Composition of DSAA-deficient basal diets fed to male broiler chicks from 2 to 5, 5 to 8, and 8 to 11 d of age, experiments 4 to 6.[1]

Ingredient	Inclusion (%)	Nutrient profile	Formulated[2] (%)	Analyzed (%)
Corn	56.52	Crude protein	22.20	21.87
Soybean meal (48%)	35.66	ME (kcal/kg)	3030	—
Poultry byproduct meal	2.00	Calcium	0.90	—
Soy oil	1.85	Available P	0.45	—
Sodium chloride	0.15	Digestible Met	0.34	0.37 (0.33)
Sodium bicarbonate	0.27	Digestible Cys	0.29	0.34 (0.30)
DL-methionine	0.02	Digestible SAA	0.63	0.71 (0.63)
L-lysine-HCl	0.15	Digestible Lys	1.28	1.37 (1.25)
L-threonine	0.06	Digestible His	0.54	0.56 (0.51)
Limestone	0.88	Digestible Trp	0.24	0.28 (0.24)
Dicalcium phosphate	1.70	Digestible Thr	0.77	0.89 (0.79)
Choline chloride	0.10	Digestible Arg	1.39	1.50 (1.41)
Vitamin and mineral premix[3]	0.63	Digestible Iso	0.85	0.91 (0.86)

[1]DL-Met was added to the basal diet at 0.07% increments to generate experimental diets that contained 0.63%, 0.70%, 0.77%, 0.83%, 0.90%, 0.97%, and 1.04% DSAA.

[2]Calculated amino acid values are reported as total dietary amino acid (digestible amino acid) and analyzed amino acid values are reported as total dietary amino acid. Analyzed total amino acid values were converted to digestible amino acid values using AminoDat Software (Version 5, 2016) by multiplying the digestibility coefficient of each ingredient by the amount of amino acid provided by that ingredient in the diet.

[3]Provided per kg of diet: vitamin A, 1,320,000 IU; vitamin D3, 440,000 ICU; vitamin E, 2860 IU; menadione, 176 mg; biotin, 6.6 mg; vitamin B12, 1.9 mg; choline, 71.5 g; niacin, 6.6 mg; pantothenic acid, 1.8 g; selenium, 40 mg; riboflavin, 880 mg; Cu, 4.4 g; Fe, 45 g; I, 135 mg; Mn, 44 g; Zn, 44 g; Co, 4.4 g.

FCRm, and relative breast weight. Regression analysis was analyzed by JMP nonlinear model option of JMP 14 (SAS Institute Inc, Cary, NC) with formulated digestible Lys or SAA concentration as the independent variable. Digestible amino acid values were calculated using AminoDat Software (Version 5, 2016) by multiplying the digestibility coefficient of each ingredient by the amount of amino acid provided by that ingredient in the diet. When a number is provided with a plus-minus sign, the second number presented is a pooled SEM.

Table 4. Formulated and analyzed sulfur amino acid content of experimental starter diets fed to broiler chicks over the 2 to 5, 5 to 8, and 8 to 11 d periods, experiment 4 to 6.[1]

	Formulated		Analyzed	
Diet	Total SAA (%)	Digestible SAA (%)	Total SAA (%)	Digestible SAA[2] (%)
Corn-SBM-PBM basal	0.72	0.63	0.73	0.64
Basal + 0.07% DL-Met	0.79	0.70	0.85	0.76
Basal + 0.14% DL-Met	0.86	0.77	0.90	0.80
Basal + 0.21% DL-Met	0.93	0.83	0.94	0.84
Basal + 0.28% DL-Met	0.98	0.90	1.06	0.95
Basal + 0.35% DL-Met	1.05	0.97	1.13	1.01
Basal + 0.42% DL-Met	1.12	1.04	1.21	1.08

[1]DL-Met was added in 0.07% increments to generate experimental diets that contained 0.63%, 0.70%, 0.77%, 0.83%, 0.90%, 0.97%, and 1.04% DSAA.

[2]Calculated amino acid values are reported as total dietary amino acid (digestible amino acid) and analyzed amino acid values are reported as total dietary amino acid. Analyzed total amino acid values were converted to digestible amino acid values using AminoDat Software (Version 5, 2016) by multiplying the digestibility coefficient of each ingredient by the amount of amino acid provided by that ingredient in the diet.

Table 5. Lysine requirements (mg/d and %) of Hubbard × Ross 708 broiler chicks estimated using linear and quadratic broken line models based on body weight gain (BWG) from 2 to 5, 5 to 8, and 8 to 11 d of age, Experiments 1 to 3.[1]

BWG (g)	Linear broken line			Quadratic broken line		
	Requirement		R^2	Requirement		R^2
	(%)[2]	(mg/d)		(%)[3]	(mg/d)	
2–5 d	1.22	610	0.79	DNC[4]	DNC[4]	—
5–8 d	1.17	772	0.87	1.35	891	0.94
8–11 d	1.16	1,283	0.72	1.29	1,427	0.96
Labadan et al. (2001)						
0–14 d[5]	1.19[6]	—	—	—	—	—
Cemin et al. (2017)						
0–12 d[7]	1.09	—	0.87	1.17	—	0.88
Dozier and Payne (2012)						
0–7 d[8]	—	—	—	1.35[6]	—	—
0–7 d[9]	—	—	—	1.27[6]	—	—
0–14 d[8]	—	—	—	1.27[6]	—	—
0–14 d[9]	—	—	—	1.18[6]	—	—

[1]Requirements were estimated based on 10 cages of 5 broilers per cage which were fed 7 diets containing that contained 0.85%, 0.95%, 1.05%, 1.15%, 1.25%, 1.35%, and 1.45% lysine concentrations.
[2]95% confidence intervals for linear broken line were 1.12 to 1.32, 1.12 to 1.22, and 1.13 to 1.23 for experiment 1, 2, and 3, respectively.
[3]95% confidence intervals for quadratic broken line were 1.30 to 1.40, and 1.25 to 1.33 for experiment 2 and 3, respectively.
[4]Did Not Converge - The response did not solve for the broken line model.
[5]Male Ross.
[6]R^2 was not reported.
[7]Male Cobb.
[8]Female Ross.
[9]Female Cobb.

RESULTS AND DISCUSSION

Lysine Requirement

Chicks utilized in this experiment were from young broiler breeder hens and weighed approximately 37.5 g/bird when they reached our facilities after transportation from the hatchery. There were no differences in initial BW ($P > 0.05$) of broiler chicks in experiments 1 to 3, but were between 1 and 2 d below breed expectations at start (Aviagen, 2019). This reduced body weight at the start is most likely due to small weights at hatch. In experiment 1, birds weighed 53 ± 0.22 g at the start of the experiment at 2 d. At 5 d, when experiment 2 started, birds weighed 103.0 ± 0.53. Finally, in experiment 3, 8-day-old birds weighed 168 ± 1.7. Final BW for each experiment was 90 on d 5 in experiment 1, 155 on d 8 in experiment 2 and 260 g/bird on d 11 in experiment 3. With the context of the low starting body weights, final body weights at the end of the 3 experiments were within 1 to 2 d of expected body weight in comparison to the performance objectives for Ross 708 broilers (Aviagen, 2019). This suggests that despite the low starting body weight, chicks were still growing as expected over the experimental period. The linear broken line model resulted in estimates of 1.22, 1.17, and 1.16% of digestible lysine to maximize BWG from 2 to 5, 5 to 8, and 8 to 11 d of age, respectively (Table 5). Requirements for FCR were estimated at 1.32, 1.21, and 1.14% or 660.2, 798.6, and 1,261.3 mg/d digestible lysine from 2 to 5, 5 to 8, and 8 to 11 d of age, respectively (Table 6). The linear broken line model did not result in estimates for maximum breast weight from 2 to 5 d of age; however, the requirements from 5 to 8 and 8 to 11 d of age were 1.13 and 1.17% or 751.4 and 1,294.4 mg/d of digestible lysine (Table 7).

Table 6. Lysine requirements (mg/d and %) of Hubbard × Ross 708 broiler chickens estimated using linear and quadratic broken line models based on mortality corrected feed conversion ratio (FCRm) from 2 to 5, 5 to 8, and 8 to 11 d, experiments 1 to 3.[1]

FCRm (g:g)	Linear broken line			Quadratic broken line		
	Requirement		R^2	Requirement		R^2
	(%)[2]	(mg/d)		(%)[3]	(mg/d)	
2−5 d	1.32	600	0.72	DNC[4]	DNC[4]	—
5−8 d	1.21	772	0.84	1.27	810	0.94
8− 11 d	1.14	1,283	0.81	1.27	1,429	0.92
Labadan et al. (2001)						
0−14 d[5]	1.12[6]	—	—	—	—	—
Cemin et al. (2017)						
0−12 d[7]	1.08	—	0.74	1.16	—	0.87
Dozier and Payne (2012)						
0−7 d[8]	—	—	—	1.38[6]	—	—
0−7 d[9]	—	—	—	DNC[4]	DNC[4]	—
0−14 d[8]	—	—	—	DNC[4]	DNC[4]	—
0−14 d[9]	—	—	—	1.26[6]	—	—

[1]Requirements were estimated based on 10 cages of 5 broilers per cage which were fed 7 diets containing that contained 0.85%, 0.95%, 1.05%, 1.15%, 1.25%, 1.35%, and 1.45% lysine concentrations.
[2]95% confidence intervals for linear broken line were 1.21 to 1.43, 1.12 to 1.30, and 1.06 to 1.22 for experiment 1, 2, and 3, respectively.
[3]95% confidence intervals for quadratic broken line were 1.12 to 1.42, and 1.18 to 1.36 for experiment 2 and 3, respectively.
[4]Did not converge - The response did not solve for the broken line model.
[5]Male Ross.
[6]R^2 was not reported.
[7]Male Cobb.
[8]Female Ross.
[9]Female Cobb.

Requirement estimates for digestible Lys for BWG, and FCR increased with age on mg/d basis, however when expressed on a percentage of diet basis, requirement estimates were reduced with chick age. These data validate the assumption of previous authors that digestible Lys requirements decrease with age when expressed on a dietary basis (Pope et al., 2002).

Table 7. Lysine requirements (mg/d and %) of Hubbard × Ross 708 broiler chickens estimated using linear and quadratic broken line models based on relative breast weight from 2 to 5, 5 to 8, and 8 to 11 d, experiments 1 to 3.[1]

Breast weight	Linear broken line			Quadratic broken line		
	Requirement		R^2	Requirement		R^2
	(%)[2]	(mg/d)		(%)[3]	(mg/d)	
2−5 d	DNC[4]	DNC[4]	—	DNC[4]	DNC[4]	—
5−8 d	1.13	751	0.86	1.27	844	0.95
8−11 d	1.17	772	0.69	1.44	950	0.99
Labadan et al. (2001)						
0−14 d[5]	1.23[6]	—	—	—	—	—

[1]Requirements were estimated based on 10 cages of 5 broilers per cage which were fed 7 diets containing that contained 0.85%, 0.95%, 1.05%, 1.15%, 1.25%, 1.35%, and 1.45% lysine concentrations.
[2]95% confidence intervals for linear broken line were 1.08 to 1.18, and 1.11 to 1.23 for experiment 2 and 3, respectively.
[3]95% confidence intervals for quadratic broken line were 1.21 to 1.33, and 1.34 to 1.54 for experiment 2 and 3, respectively
[4]Did not converge - The response did not solve for the broken line model.
[5]Male Ross.
[6]R^2 was not reported.

The requirement estimates for breast weight increased from 5 to 8 d in comparison to 8 to 11 d of age which could indicate that the lysine digested earlier in life was used to drive structural development not maximization of breast weight. The linear-broken line Lys requirements of male Ross broiler chicks from 0 to 14 d of age were estimated at 1.28, 1.21, and 1.32% total Lys or 1.19, 1.12, and 1.23% digestible Lys for BWG, FCR and breast weight, respectively (Labadan et al., 2001). These results are similar to estimates for BWG in the current experiment. The Lys requirement estimates for FCR were similar, but slightly lower than the requirements reported in the current experiment. Breast weight requirements from 1 to 14 d were higher than the currently reported requirements, which are consistent with the idea that older birds may have a higher Lys requirement for breast weight when compared to birds early in life. Previously, Cobb 500 broilers were raised in floor pens from 0 to 12 d of age resulting in digestible Lys requirements of 1.09 and 1.08% digestible Lys for BWG and FCR, respectively. These estimates were lower than those found in the current experiment or other reports for Ross strains (Cemin et al., 2017). This might suggest that broiler strains do have differences in digestible Lys requirements.

Results from the quadratic broken line analysis resulted in similar responses as the linear broken line model where the requirements of digestible Lys decreased from experiment 2 to 3 for BWG and FCR, respectively. No requirement was estimated for experiment 1 for all parameters for the quadratic broken line model due to a lack of quadratic response. Overall, the quadratic broken line resulted in higher estimates for all parameters compared to the linear broken line analysis. Additionally, the quadratic broken line analysis resulted in a better fit (R^2) compared to the linear broken line analysis. The Lys requirement of 0 to 7 and 0 to 14 d of age female Ross 708 and Cobb 500 broilers were estimated using quadratic broken line models (Dozier and Payne, 2012). The digestible Lys requirement was estimated at 1.35 and 1.27% for 0 to 7 and 0 to 14 d of age BWG for Ross broilers similar to the quadratic-broken

Table 8. Digestible sulfur amino acid requirements (mg/d and %) of Hubbard × Ross 708 broiler chickens estimated using linear and quadratic broken line models based on body weight gain (BWG) from 2 to 5, 5 to 8, and 8 to 11 d, experiments 4 to 6.[1]

	Linear broken line			Quadratic broken line		
	Requirement		R^2	Requirement		R^2
BWG (g)	(%)[2]	(mg/d)		(%)[3]	(mg/d)	
2−5 d	0.82	156	0.80	0.85	173	0.95
5−8 d	0.81	253	0.86	0.91	288	0.94
8−11 d	0.94	321	0.74	0.98	406	0.96
Sklan and Noy (2003)						
0−7 d[4]	0.82	—	0.96	—	—	—
Garcia and Batal (2005)						
0−7 d[5]	0.82[6]	—	—	—	—	—
Goulart et al. (2011)						
0−7 d[5]	—	—	—	0.87[6]	183	—
Kalinowski et al. (2003)						
0−21 d[4]	—	—	—	0.89[6]	—	—

[1]Requirements were estimated based on 10 cages of 5 broilers per cage which were fed 7 diets containing that contained 0.63%, 0.70%, 0.77%, 0.83%, 0.90%, 0.97%, and 1.04% DSAA concentrations.

[2]95% confidence intervals for linear broken line were 0.79 to 0.85, 0.79 to 0.83, and 0.88 to 1.00 for experiment 4, 5, and 6, respectively.

[3]95% confidence intervals for quadratic broken line were 0.78 to 0.86, 0.86 to 0.96, and 0.90 to 1.06 for experiment 4, 5, and 6, respectively.

[4]Male Ross 308.

[5]Male Cobb 500.

[6]R^2 was not reported.

Table 9. Digestible sulfur amino acid requirements (mg/d and %) of Hubbard × Ross 708 broiler chickens estimated using linear and quadratic broken line models based on mortality corrected feed conversion ratio (FCRm) from 2 to 5, 5 to 8, and 8 to 11 d, experiments 4 to 6[1].

FCR (g:g)	Linear broken line			Quadratic broken line		
	Requirement		R^2	Requirement		R^2
	(%)[2]	(mg/d)		(%)[3]	(mg/d)	
2−5 d	0.82	156	0.74	0.87	170	0.94
5−8 d	0.80	248	0.72	0.95	279	0.94
8−11 d	0.90	303	0.71	0.96	338	0.92
Sklan and Noy (2003)						
0−7 d[4]	0.82	—	0.92	—	—	—
Garcia and Batal (2005)						
0−7 d[5]	0.84[6]	—	—	—	—	—
Goulart et al. (2011)						
0−7 d[5]	—	—	—	0.87[6]	183	—
Kalinowski et al. (2003)						
0−21 d[4]	—	—	—	0.89[6]	—	—

[1]Requirements were estimated based on 10 cages of 5 broilers per cage which were fed 7 diets containing that contained 0.63%, 0.70%, 0.77%, 0.83%, 0.90%, 0.97%, and 1.04% DSAA concentrations.

[2]95% confidence intervals for linear broken line were 0.78 to 0.86, 0.76 to 0.84, and 0.82 to 0.98 for experiment 4, 5, and 6, respectively.

[3]95% confidence intervals for quadratic broken line were 0.82 to 0.92, 0.90 to 1.00, and 0.88 to 1.04 for experiment 4, 5, and 6, respectively.

[4]Male Ross 308.

[5]Male Cobb 500.

[6]R^2 was not reported.

line estimates reported in the current experiment. Female Cobb broilers had lower digestible Lys requirements of 1.27 and 1.18% for BWG in comparison to the current experiment and previous data. This again supports the idea that there are differences in the efficiency of the utilization of Lys between strains. Similarly, the digestible Lys requirement for FCR was estimated at 1.27% for 5 to 8 and 8 to 11 d but was not estimable at 2 to 5 d of age. As expected these estimated from the quadratic broken line were higher than estimates from the linear broken line. Furthermore, these results were similar to the estimates reported in Cobb female broilers in a previous experiment from 0 to 14 d of age but lower than the estimated requirements for the Ross female broilers (Dozier and Payne, 2012).

DSAA Requirement

Chicks utilized in this experiment were from young broiler breeder hens and weighed approximately 38.5 g/bird when they reached our facilities after transportation from the hatchery. There were no differences in initial BW ($P > 0.05$) of broiler chicks in experiments 1 to 3, but were between 1 and 2 d below breed expectations at start (Aviagen, 2019). This reduced body weight at the start is most likely due to small weights at hatch. In experiment 1, birds weighed 58 ± 0.27 g at the start of the experiment at 2 d. At 5 d, when experiment 2 started, birds weighed 108 ± 0.53. Finally, in experiment 3, 8-day-old birds weighed 165 ± 1.7. Final BW for each experiment was 105 on d 5 in experiment 1, 174 on d 8 in experiment 2 and 260 g/bird on d 11 in experiment 3. With the context of the low starting body weights, final body weights at the end of the 3 experiments were within 1 to 2 d of expected body weight in comparison to the performance objectives for Ross 708 broilers (Aviagen, 2019). This suggests that despite the low starting body weight, chicks were still growing as expected over the experimental period. The linear broken line model resulted in estimates of 0.82, 0.81, and 0.94% of DSAA to maximize BWG from 2 to 5, 5 to 8, and 8 to 11 d of age, respectively (Table 8). Requirements for FCR were

Table 10. Digestible sulfur amino acid requirements (mg/d and %) of Hubbard × Ross 708 broiler chickens estimated using linear and quadratic broken line models based on relative breast weight from 2 to 5, 5 to 8, and 8 to 11 d, experiments 4 to 6.[1,2]

	Linear broken line			Quadratic broken line		
	Requirement[3]		R^2	Requiremen[4]		R^2
Breast weight (%)	(%)	(mg/d)		(%)	(mg/d)	
2−5 d	0.84	160	0.74	0.97	203	0.94
5−8 d	0.82	260	0.79	0.95	312	0.95
8−11 d	0.98	331	0.81	1.17	503	0.99

[1]Requirements were estimated based on 10 cages of 5 broilers per cage which were fed 7 diets containing that contained 0.63%, 0.70%, 0.77%, 0.83%, 0.90%, 0.97%, and 1.04% DSAA concentrations.

[2]No previous breast weight data have been reported over this timeframe of broiler development.

[3]95% confidence intervals for linear broken line were 0.78 to 0.90, 0.78 to 0.86, and 0.90 to 1.06 for experiment 4, 5, and 6, respectively.

[4]95% confidence intervals for quadratic broken line were 0.91 to 1.03, 0.90 to 1.00, and 1.03 to 1.31 for experiment 4, 5, and 6, respectively.

estimated at 0.82, 0.80, and 0.90 or 156.6, 248.5, and 303.8 mg/d for 2 to 5, 5 to 8, and 8 to 11 d of age, respectively (Table 9). The requirements for breast weight were estimated at 0.84, 0.82, and 0.98 or 160.4, 260.8, and 331.8 mg/d for 2 to 5, 5 to 8, and 8 to 11 d of age, respectively (Table 10). A study investigating the SAA requirement in male Ross broilers found estimated the requirement at 0.91 % Total SAA or 0.82% digestible SAA for BWG and FCR using a linear broken line analysis (Sklan and Noy, 2003). Similarly, Garcia and Batal (2005) found a similar requirement in 2 experiments estimated at 0.82 and 0.84% for BWG and FCR using a linear broken line model in Cobb 500 male broilers from 0 to 7 d of age. These estimates are similar to the results of this experiment for BWG from 2 to 5 and 5 to 8 d of age but slightly higher when FCR requirements are considered. These results are somewhat surprising as it does not appear that Met requirements for BWG have increased over this time period in the past 15 to 20 yr of genetic selection. A requirement of 0.87% DSAA was found using a quadratic polynomial model for optimizing BWG and FCR utilizing Cobb 500 broiler from 0 to 7 d of age (Goulart et al., 2011). A study utilizing the quadratic polynomial model found a comparable requirement of 0.89% in Ross 308 fast feathering male broilers from 0 to 21 d of age (Kalinowski et al., 2003). The quadratic polynomial model results in a higher estimation of requirements compared to the linear or quadratic broken line used in our experiment which would explain the variation in estimates. The quadratic broken line estimates for DSAA requirements expressed on a dietary basis increased when 2 to 5 d estimates were compared to 5 to 8 d estimates for BWG and FCR which was inconsistent with the linear broken line estimates. Although consistent with other reports, quadratic broken line estimates resulted in better overall fit for the data compared to the linear broken line (Sarsour et al., 2021).

In the current set of experiments, digestible SAA requirement estimates for BWG, FCRm, and relative breast weight increased with age on a mg/d basis, however when expressed on a percentage of the diet basis, the expected decrease was observed going from 2 to 5 d of age to 5 to 8 d of age. DSAA requirements increased from 8 to 11 d of age. These increased DSAA requirements from 8 to 11 d of age generally correspond with the feathering of chicks at this time and could be related to the role that sulfur amino acids play in feather production. Previous authors have reported a 0.05% increase in the DSAA requirement of fast vs. slow feathering Ross broilers from 0 to 3 d of age (Kalinowski et al., 2003). Furthermore, Zeng and others (2015) reported that by increasing the DSAA concentration in the diet from 0.55% to 0.81%, there was an increase in feather coverage in 28- and 35-day-old Pekin ducks. The current results indicate that DSAA requirements do not decrease over the starter period and at least for DSAA the assumption that increased feed intake will reduce dietary amino acid needs over the starter period do not seem to hold true. This would indicate that before assumptions are made on the responses of specific amino acids requirements within a feeding phase, validation needs to be done due to the utilization of some amino acids for functions besides direct growth that might be driving or contributing to the requirement.

CONCLUSION AND APPLICATIONS

1. The linear broken line lysine requirement was estimated to be 1.22, 1.17, and 1.16% for BWG and 1.31, 1.21, and 1.14% for FCR from 2 to 5, 5 to 8, and 8 to 11 d of age, respectively.
2. Requirements for dietary lysine decrease with increased age and the increase in feed intake associated with older and larger chicks, validating the idea that within a feeding phase, dietary lysine requirement would decrease as chicks' age.
3. The linear broken line DSAA requirement was estimated at 0.82, 0.81, and 0.94% for BWG and 0.82, 0.80, and 0.90 for FCR from 2 to 5, 5 to 8, and 8 to 11 d of age, respectively.
4. Requirements for dietary DSAA do not decrease with increased age within the starter period and do not follow the same responses as dietary lysine.
5. The idea of further refining feeding periods to more closely follow biological requirements within a feeding phase is still viable, but further validation of specific amino acid requirements should be completed due to potential nongrowth related factors.

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DISCLOSURES

Albaraa Sarsour and Mike Persia declare no conflict of interest.

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