

Ideal Amino Acid Profile for 28-to-34-Week-Old Laying Hens

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Kristjan Bregendahl,¹ assistant professor;
Stacey Roberts, graduate research assistant;
Brian Kerr, research leader, USDA/ARS;
Dirk Hoehler, director of technical services,
Evonik Degussa Corporation

Summary and Implications

The ideal amino acid (AA) profile employs the concept that, whereas absolute AA requirements change due to genetic or environmental factors, the ratios among them are only slightly affected. Thus, once the ideal AA profile has been determined, the requirement for a single AA (i.e., lysine) can be determined experimentally for a given field situation and the requirements for all the other AA calculated from the ideal ratios. Seven separate experiments were conducted with laying hens to determine the ideal ratio of arginine, isoleucine, methionine, methionine+cystine, threonine, tryptophan, and valine relative to lysine for maximal egg mass. The assays were conducted simultaneously using the same basal diet to which crystalline AA were added to create the graded level of the respective assay AA and to ensure that the assayed AA was first limiting. Hens were fed the assay diets from 26 to 34 weeks of age, with the first 2 weeks considered a depletion period. Egg production was recorded daily and egg weight was determined weekly on eggs collected over 48 hours; egg mass was calculated as egg production \times egg weight. The requirement for each AA was determined using the broken-line regression method. Consumption of arginine did not affect egg mass, thus an ideal arginine:lysine ratio could not be determined. The ideal AA ratio for maximum egg mass for 28-to-34-week-old laying hens was isoleucine 79%, methionine 47%, methionine+cystine 94%, threonine 77%, tryptophan 22%, and valine 93% on a true digestible basis relative to lysine.

Introduction

Amino acid requirements for laying hens are published by the National Research Council.² However, the experiments upon which these requirements are based are dated and do not account for the genetic progress of laying hens in the last 12 or more years. Amino acid requirements have been reported since the publication of the National

Research Council requirements. However, these experiments have been conducted for 1 AA at a time, performed under different experimental conditions and with different basal diets, genetic lines of hens, feed consumption rates, dietary energy contents, ambient temperature, cage space, and ages of laying hens, all of which influence the AA requirements.

Because multiple factors affect AA requirements, requirements determined under experimental conditions may not be applicable under field conditions. The solution to obtaining reliable AA requirements is therefore *not* to determine the AA requirements, but rather to determine the ideal AA profile for laying hens. The ideal AA profile employs the concept that, while AA requirements change drastically due to genetic or environmental factors, the ratio among them is only slightly affected. Thus, once the ideal AA profile has been determined, the requirement for a single AA (e.g., lysine) can be determined experimentally for a given field situation and the requirement for all the other AA calculated. Such an approach has been adopted with success by the swine industry and is finding use in the broiler industry as well.

The objective of Experiment 1 was to investigate responses of laying hens, 28 to 34 weeks of age, to graded dietary inclusions of the essential AA arginine, isoleucine, lysine, methionine, threonine, tryptophan, and valine, in order to determine the ideal AA ratios of the assayed AA relative to lysine. In Experiment 2, the objective was to confirm the lysine, methionine, and methionine+cystine requirements (and, therefore, the ideal methionine:lysine and methionine+cystine:lysine ratios) determined in Experiment 1.

Material and Methods

Housing and Management

A total of 1,008 white single-comb Leghorn-type hens (Hy-Line W-36), 26 weeks of age, were housed 2 per cage (619 cm²/hen) in wire-bottomed cages (Chore-Time, Milford, IN), each equipped with a plastic self-feeder and a nipple drinker. The photoperiod was 16 h of light and 8 h of darkness and the hens had free access to feed and water at all times. Prior to Experiment 1 and between Experiments 1 and 2, the hens were fed and managed in accordance with the Hy-Line W-36 Commercial Management Guide. All procedures relating to the use of live animals were approved by the Iowa State University Institutional Animal Care and Use Committee.

¹To whom correspondence should be addressed:
kristjan@iastate.edu

²National Research Council. 1994. Nutrient requirements of poultry. 9th ed. Natl. Acad. Press, Washington, DC.

Dietary Treatments

A basal diet was formulated using corn, soybean meal, and meat and bone meal to meet or exceed all nutrient requirements, except amino acids. In Experiment 1, the basal diet was mixed in 2 separate 2,722-kg batches in a horizontal mixer at a commercial feed mill (Kent Feeds, Altoona, IA) and bagged in 22.6-kg bags according to batch number. In Experiment 2, a basal diet was mixed in a vertical mixer in 3 separate 500-kg batches at the Iowa State University Poultry Science Research Center. Representative samples of the basal-diet batches were pooled within experiment and analyzed for AA content by ion-exchange chromatography and for true AA digestibility by the cecectomized rooster assay. The crystalline AA added to the basal diet were assumed to be 100% true digestible.

The 35 assay diets in Experiment 1 were formulated with equal parts of each of the 2 basal-diet batches plus a mixture of cornstarch, K_2CO_3 to maintain a similar dietary electrolyte balance among all diets, and crystalline AA to create 5 equally spaced graded inclusions of each of the 7 assayed AA, such that diet 3 provided the assayed AA at the estimated requirement (Table 1). In Experiment 2, only the responses to lysine and methionine were determined; thus, there were 10 assay diets formulated with equal parts of each of the 3 basal-diet batches as described for Experiment 1 (Table 1). Crystalline AA, other than the one assayed, were added to all diets to assure that the assayed AA was first-limiting in any given assay diet.

Data Collection

Hens were offered free access to the assay diets from 26 to 34 weeks of age (Experiment 1) or from 50 to 58 weeks of age (Experiment 2), with the first 2 weeks of each experiment considered a depletion period. Thus, only data from the last 6 weeks of the experiments were used in the statistical analyses.

Egg production was recorded daily and feed consumption (determined as feed disappearance) was measured weekly throughout the 8-week-long experiments. Consumption of the assay AA (mg/day) was calculated from the mean daily feed consumption (g/day) over the last 6 weeks of each experiment and the dietary true digestible AA content (%). This latter content was calculated from the analyzed total AA content of the basal diet multiplied by the analyzed digestibility coefficient plus the inclusion of crystalline AA. Once every week, eggs collected over a 48-hour period were weighed and the egg mass calculated by multiplying the week's egg-production rate by the egg weight.

Statistical Analyses

The requirement for each assayed AA was determined in a randomized complete block design with 5 dietary treatments (i.e., 5 levels of the assayed AA) and 12 blocks.

The cage location within the barn served as the blocking criterion and the experimental unit was 1 cage containing 2 hens. The requirements for digestible AA were calculated with the single-slope broken-line regression model with the consumption of the assayed AA (mg/day) as the independent variable. Block was not included in the broken-line regression model. Feed consumption data were analyzed by analysis of variance (ANOVA) with block and dietary treatment as the independent variables; treatment effects were separated using linear, quadratic, and cubic orthogonal polynomial contrasts.

Results

The responses to consumption of arginine, isoleucine, lysine, methionine, methionine+cystine, threonine, tryptophan, and valine in Experiment 1 are shown in Figure 1. Hens fed the 5 arginine assay diets consumed between 574 and 843 mg/day of true digestible arginine, yet there were no responses to consumption of arginine. The responses to consumption of lysine, methionine, and methionine+cystine in Experiment 2 are shown in Figure 2. Hens fed the graded levels of AA generally responded by increasing the feed consumption in a linear or curvilinear matter (Table 2). The requirements for true digestible AA for maximal egg production, egg weight, and egg mass, calculated using the broken-line regression method are shown in Table 3, whereas the ideal AA profile for maximal egg mass are shown in Table 4.

Discussion

The AA requirements, used to calculate the ideal AA profile, were determined with the broken-line regression model. This method is considered the best for obtaining the ideal ratios among AA, whereas curvilinear models, such as exponential or quadratic curve fitting, are better suited to establish the AA requirements for optimal performance. Typically, the broken-line regression method results in lower AA requirements than when a non-linear curve fitting is applied to the same dataset. However, the broken-line regression model has the advantages of a clearly defined breakpoint (i.e., the requirement) at a dietary AA consumption that marginally limits performance, both necessary to determine the ideal AA profile. Although the absolute AA requirements (mg/day) are reported herein, they are only valid for the particular hens in the particular experimental settings in the present study and should not necessarily be used in commercial settings, especially because they were determined using the broken-line method, and not a curvilinear model.

The intent with the experimental design of the current study was to have 2 of the 5 assay diets supply the assayed AA below the estimated requirement, 1 diet supply the assayed AA at the estimated requirement, and 2 diets supply the assayed AA above the estimated requirement. However,

the requirements for lysine and methionine were overestimated in Experiment 1, such that only 1 assay diet supplied the assayed AA below the observed requirement. In other words, only 1 of the 5 assay diets was deficient in the assayed AA, lowering the confidence in the calculated AA requirement needed to calculate the ideal AA profile. Therefore, the lysine and methionine requirements were reevaluated in Experiment 2 with the same hens using the same methodology as in Experiment 1, albeit with lower dietary contents of the assayed AA. Consequently, the breakpoints for lysine, methionine, and methionine+cystine were better defined in Experiment 2. Despite the differences in the age of the hens between the 2 experiments, the requirements for true digestible lysine, methionine, and methionine+cystine were similar. Accordingly, the ideal methionine:lysine ratios (47 and 52% in Experiments 1 and 2, respectively) and methionine+cystine:lysine ratios (94 and 96% in Experiments 1 and 2, respectively) corresponded fairly well between the 2 experiments, indicating that the ratios for methionine and methionine+cystine determined in Experiment 1 were acceptable. Hence, the ideal AA profile for 28-to-34-week-old laying hens was calculated from the true digestible AA requirement for maximal egg mass from Experiment 1.

The ideal AA ratio for maximum egg mass for 28-to-34-week-old laying hens was isoleucine 79%, methionine 47%, methionine+cystine 94%, threonine 77%, tryptophan 22%, and valine 93% on a true digestible basis relative to lysine. The ideal AA profile determined in this study indicated that laying hens need less true digestible isoleucine and valine and more true digestible methionine and threonine in relation to lysine than that suggested by Coon and Zhang³ and that calculated from requirements published by the National Research Council.² The ideal isoleucine:lysine ratio observed in the present study corresponded well with that calculated from AA recommendations by the Dutch Centraal Veevoederbureau.⁴

In addition, the determined ideal AA profile agrees well with the profile calculated from AA recommendations suggested by Leeson and Summers⁵ for 32-to-45-week-old hens and is similar to that reported by Jais et al.⁶ with the exception of tryptophan and valine. The ideal methionine:lysine and methionine+cystine:lysine ratios in the present study were higher than those reported by the National Research Council,² but agree well with the ratios suggested by the Centraal Veevoederbureau⁴ and by Leeson and Summers⁵ for 32-to-45-week-old hens. If the lowest true digestible arginine consumption observed in Experiment 1 (i.e., 574 mg/day) is accepted as meeting or exceeding the requirement of the hen for arginine, the ideal arginine:lysine ratio was then no higher than 107%, similar to the 101% calculated from the National Research Council² arginine and lysine recommendations, and less than the 130% recommended by Coon and Zhang³ (Table 3).

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³Coon, C., and B. Zhang. 1999. Ideal amino acid profile for layers examined. *Feedstuffs* 71(14):13–15, 31.

⁴Centraal Veevoederbureau. 1996. Amino zurenbehoefte van leghennen en vleeskuikens [Amino acid requirements for laying hens and broiler chickens]. Documentation Report nr. 18 (in Dutch), Centraal Veevoederbureau, Lelystad, The Netherlands.

⁵Leeson, S., and J. D. Summers. 2005. *Commercial poultry production*. 3rd ed. University Books, Guelph, ON.

⁶Jais, C., F. X. Roth, and M. Kirchgessner. 1995. The determination of the optimum ratio between the essential amino acids in laying hen diets. *Arch. Geflügelk.* 59:292–302.

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Table 1. Dietary true digestible amino acid content of the assayed amino acid (as-is basis).^{1,2}

Amino acid assayed	Assay diet				
	1 ³	2	3	4	5
	----- % -----				
Experiment 1					
Arginine	0.69	0.75	0.81	0.87	0.93
Isoleucine	0.37	0.50	0.64	0.78	0.92
Lysine	0.51	0.69	0.87	1.05	1.24
Methionine	0.19	0.41	0.63	0.85	1.07
Methionine+cystine ⁴	0.35	0.57	0.78	1.01	1.22
Threonine	0.38	0.45	0.53	0.60	0.67
Tryptophan	0.09	0.13	0.17	0.21	0.25
Valine	0.47	0.61	0.74	0.88	1.02
Experiment 2					
Lysine	0.34	0.46	0.59	0.72	0.84
Methionine	0.13	0.25	0.37	0.48	0.60
Methionine+cystine ⁴	0.36	0.47	0.59	0.71	0.83

¹Calculated from the analyzed total amino acid content of the basal diet multiplied by the analyzed digestibility coefficient plus the inclusion of crystalline amino acids (the latter assumed 100% digestible).

²Crystalline amino acids, other than the one assayed, were added to all diets to assure that the assayed amino acid was first-limiting in the particular assay diet.

³Basal diet

⁴The responses to methionine+cystine were evaluated in the methionine assay; graded levels of methionine+cystine were created by addition of methionine (not methionine+cystine) to the basal diet.

Table 2. Feed consumption of hens fed the amino acid-supplemented basal diet.¹

Amino acid assayed	Diet					SEM ²
	1	2	3	4	5	
	----- g/day -----					
Experiment 1						
Arginine ³	89.0	87.9	82.1	83.1	80.6	1.6
Isoleucine ^{3,4,5}	55.7	79.8	85.2	80.9	79.7	1.9
Lysine ^{4,5}	77.8	89.0	84.6	80.5	81.4	1.7
Methionine ^{3,4,5}	62.7	86.3	81.6	81.5	82.7	1.8
Threonine ^{4,5}	82.6	90.5	85.2	82.8	82.2	1.8
Tryptophan ^{3,4}	61.6	74.7	87.9	80.9	82.5	2.0
Valine ^{3,4,5}	53.8	83.3	85.1	86.7	81.8	1.3
Experiment 2						
Lysine ^{3,4}	78.2	91.9	94.2	97.3	90.2	1.9
Methionine ^{3,4,5}	60.8	92.3	97.5	92.3	91.5	2.1

¹The data for methionine and methionine+cystine were from the same data set in each experiment; feed consumption rates for methionine and methionine+cystine were therefore equal.

²Pooled standard error of the mean (n = 12).

³Linear effect ($P < 0.05$).

⁴Quadratic effect ($P < 0.05$).

⁵Cubic effect ($P < 0.05$).

Table 3. Dietary true digestible amino acid requirements and associated maximal production values derived from the broken-line regression analysis of egg production data for laying hens 28 to 34 weeks of age.^{1,2}

Amino acid	Egg production		Egg weight		Egg mass	
	Requirement	Maximal egg production at requirement	Requirement	Maximal egg weight at requirement	Requirement	Maximal egg mass at requirement
	mg/day	%	mg/day	g	mg/day	g/day
Arginine ³	ND	–	ND	–	ND	–
Isoleucine	427	90.5	394	53.2	426	48.1
Lysine	482	92.3	649	53.3	538	49.1
Methionine	347	92.2	143	53.2	253	48.7
Methionine+cystine ³	479	92.4	264	53.2	506	49.2
Threonine	400	93.3	418	53.4	414	49.8
Tryptophan ³	119	91.7	ND	–	120	48.7
Valine	493	93.5	517	53.2	501	49.4

¹Data from Figure 1.

²Amino acid requirement data should only be used for determination of the ideal amino acid profile (see text).

³ND, not determined (a broken-line regression could not be fitted to the data, $P > 0.05$).

⁴Graded levels of methionine+cystine were created by addition of methionine (not methionine+cystine).

Table 4. Ideal amino acid profiles¹ for laying hens determined in the present study (28 to 34 wk of age) and calculated from reported amino acid requirements.

Amino acid	Present study ²	NRC (1994) ³	Jais et al. (1995) ⁴	CVB (1996) ⁵	Coon and Zhang (1999) ⁶	Leeson and Summers (2005) ⁶
Lysine	100	100	100	100	100	100
Arginine	– ⁸	101	82	–	130	103
Isoleucine	79	94	76	79	86	79
Methionine	47	43	44	50	49	51
Methionine+cystine	94	84	–	93	81	88
Threonine	77	68	76	66	73	80
Tryptophan	22	23	16	19	20	21
Valine	93	101	64	86	102	89

¹Lysine requirement set at 100%.

²Based on true digestible requirements for maximal egg mass in Experiment 1 (Table 3).

³Based on total amino acid requirements (National Research Council. 1994. Nutrient requirements of poultry. 9th ed. Natl. Acad. Press, Washington, DC).

⁴Based on N balance (Jais, C., F. X. Roth, and M. Kirchgessner. 1995. The determination of the optimum ratio between the essential amino acids in laying hen diets. Arch. Geflügelk. 59:292–302).

⁵Based on digestible amino acid requirements (Centraal Veevoederbureau. 1996. Amino-zurenbehoefte van leghennen en vleeskuikens [Amino acid requirements for laying hens and broiler chickens]. Documentation Report nr. 18 (in Dutch), Centraal Veevoederbureau, Lelystad, The Netherlands).

⁶Based on digestible amino acid requirements (Coon, C., and B. Zhang. 1999. Ideal amino acid profile for layers examined. Feedstuffs 71(14):13–15, 31)

⁷Based on total amino acid requirements for 32-to-45-wk-old laying hens (Leeson, S., and J. D. Summers. 2005. Commercial poultry production. 3rd ed. University Books, Guelph, ON).

⁸The Arg:Lys ratio is estimated to be 107 or less; see text.

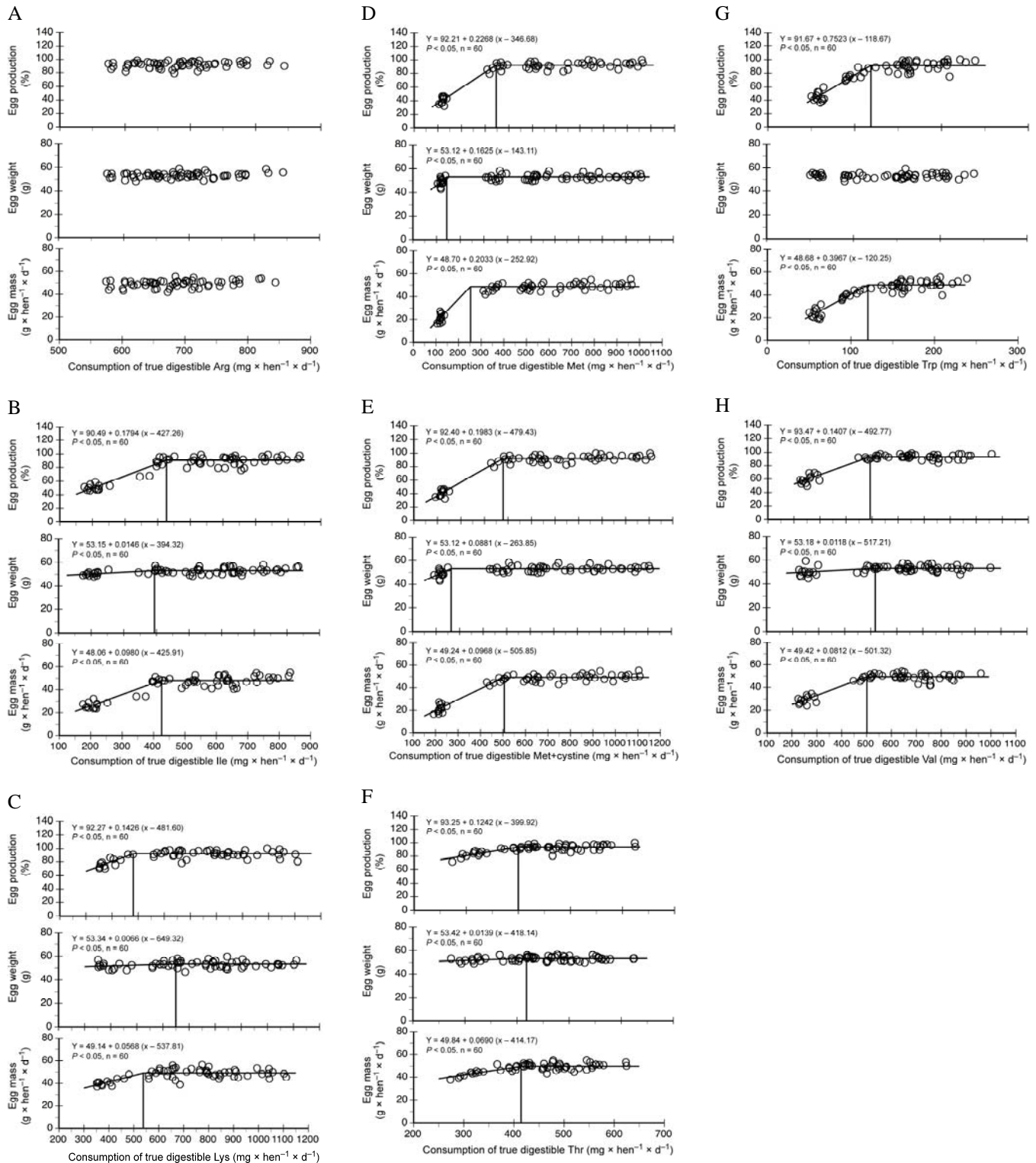


Figure 1. Responses to consumption of true digestible arginine (A), isoleucine (B), lysine (C), methionine (D), methionine+cystine (E), threonine (F), tryptophan (G), and valine (H) in Experiment 1 (hens at 28 to 34 weeks of age) and associated broken-line regressions. Each dot (o) represents data collected over 6 week from 1 cage containing 2 hens.

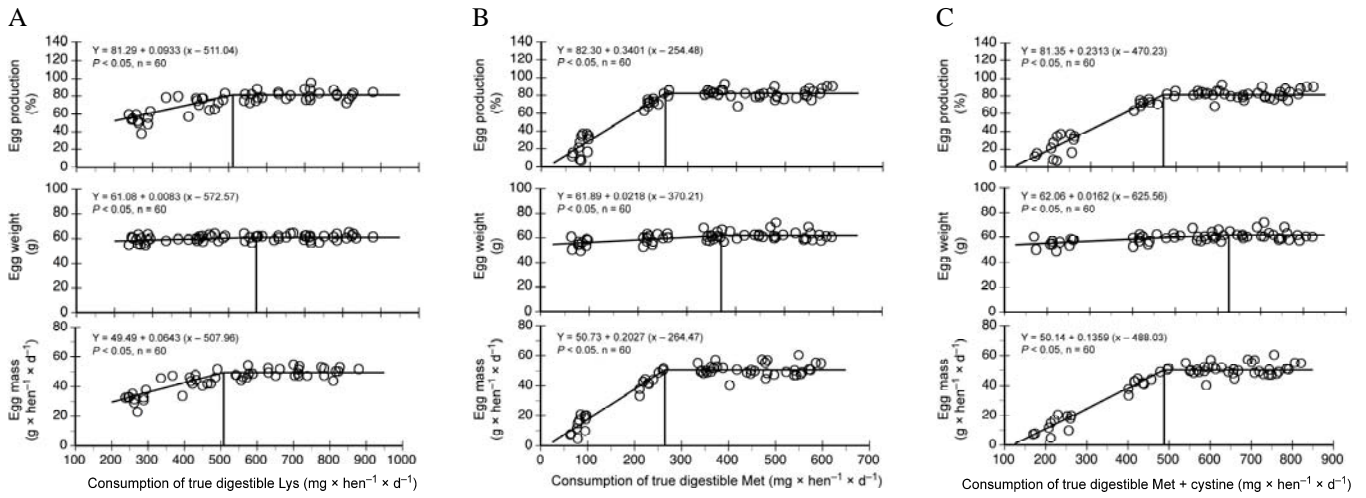


Figure 2. Responses to consumption of true digestible lysine (A), methionine (B), and methionine+cystine (B) in Experiment 2 (hens at 52 to 58 weeks of age) and associated broken-line regressions. Each dot (o) represents data collected over 6 week from 1 cage containing 2 hens.