RUNNING TITLE: METHIONINE+CYSTEINE/LYSINE RATIO

TITLE: Effects of Added Synthetic Lysine on Second Cycle Commercial Leghorns While Maintaining the Methionine+Cysteine/Lysine Ratio at 0.75

AUTHORS: G. Wu, Z. Liu, M.M. Bryant, R. A. Voitle, and D. A. Roland, Sr. [1]

Department of Poultry Science, Auburn University, Auburn, AL, 36849

SPECIFIC SECTION: METABOLISM AND NUTRITION

SUMMARY

Two experiments were conducted to determine the influence of synthetic lysine on performance and profits of commercial Leghorns while maintaining a 0.75 Met+Cys/Lys ratio in a corn-soy diet. In Experiment 1, a 3 × 4 factorial experiment with three protein levels (16.01, 14.86, and 14.42%) and four added synthetic lysine levels (0.000, 0.030, 0.059, and 0.088%) was conducted with Hy-line W-36 laying hens (n = 1440, 85 wk old) for 8 weeks. In Experiment 2, a 3 × 3 factorial experiment with three protein levels (14.87, 14.44, and 14.00%) and three added synthetic lysine levels (0.000, 0.030, and 0.059%) was conducted with Hy-line W-36 laying hens (n = 1080; 98 wk old) for 12 weeks. Results of Experiment 1 showed that there were no significant effect of adding synthetic lysine while maintaining a 0.75 Met+Cys/Lys ratio on egg production, egg mass, feed intake, feed conversion, egg specific gravity and final body weight. Results of Experiment 2 showed that the addition of synthetic lysine while maintaining a 0.75 Met+Cys/Lys ratio had significant effects on egg production, egg mass, feed intake, feed conversion, and egg specific gravity. In conclusion, when protein level of a corn-soy diet is below 15% (supplying less than approximately 15 g protein/h/d) or lysine intake is less than 720 mg/h/d, the addition of synthetic lysine while maintaining a 0.75 Met+Cys/Lys ratio can significantly improve performance of laying hens. Although adding synthetic lysine to diets containing less than approximately 15% protein can

improve performance and profits depending on value of performance improvements gained and cost of protein and lysine, these results give no indication as to whether or not diets containing less than 15% protein would be economical.

Key words: Hens, Synthetic lysine, Met+Cys/Lys ratio, Protein, Performance

DESCIPTION OF PROBLEM

The amino acid composition of diets is generally considered to affect the efficiency of protein utilization. Methionine is the first limiting amino acid in corn-soybean diets, followed by lysine [1]. The ideal protein concept is often used in the formulation of diets. This concept assumed that all amino acids are in balance and are equally limiting [2, 3]. Although the absolute requirements of amino acids may change in different practical situations, the ratios between amino acids should remain stable. Therefore, lysine is often used as the reference amino acid and the other essential amino acids are calculated by using the respective ratio to lysine.

Reports on the lysine requirement of laying hens varied from 650 to 900 mg/h/d [4, 5, 6, 7, 8]. Some studies [9, 10, 11] demonstrated that the optimal Methionine+Cystine/Lysine (Met + Cys/Lys) ratio for laying hens was 0.75. If the protein level of diets changes, the natural lysine content also varies. When protein content of a typical corn-soybean diet decreases from 19% to 14%, lysine content decreases from 1.02% to 0.69% while methionine level decreases from 0.29% to 0.24%. The sharp reduction in lysine level in relation to the drop in methionine suggests that addition of synthetic lysine to diets while maintaining the optimal Met+Cys/Lys ratio may have an effect on the performance of laying hens. However, Few studies have been conducted to investigate influence of the addition of synthetic lysine on the performance of laying hens while maintaining the optimal Met+Cys/Lys ratio, and results of these studies were inconsistent. Sohail et al. [12] and Liu et al. [13] reported that the addition of synthetic lysine while maintaining the optimal Met+Cys/Lys ratio had no effect on egg production, feed intake, egg weight or egg specific gravity of layers in Phase I of the first cycle (21-

37 wk) and in the second cycle (70-80 wk) respectively. However, Liu et al. [14] reported that there were significant effects of added synthetic lysine while maintaining the optimal Met+Cys/Lys ratio on egg production, egg mass, and egg weight of laying hens in Phase II of the first cycle.

The objective of Experiment 1 and Experiment 2 was to determine the influence of adding synthetic lysine while maintaining the Met+Cys/Lys ratio at 0.75 on performance and profits for commercial Leghorns in the second cycle (85-93 wk and 98-110 wk).

MATERIAL AND METHODS

In Experiment 1, a 3 × 4 factorial experiment with three protein levels (16.01, 14.86, and 14.42%) and four added synthetic lysine levels (0.000, 0.030, 0.059, and 0.088%) (Table 1) was conducted. Twelve diets were formulated based on lysine instead of protein, and Met + Cys/Lys ratio was maintained at 0.75 (Table 1). The diets were fed for eight weeks. In Experiment 2, a 3×3 factorial experiment with three protein levels (14.87, 14.44, and 14.00%) and three added synthetic lysine levels (0.000, 0.030, and 0.059%) was conducted (Table 2). Nine diets were formulated based on lysine instead of protein, and Met+Cys/Lys was maintained at 0.75 (Table 2). The diets were fed for 12 weeks. The feed samples were sent for nutrient analysis[1].

In Experiment 1, Hy-line W-36 hens (n = 1440) in the second cycle (85 week old) were randomly divided into 12 treatments (8 replicates of 15 hens per treatment). In Experiment 2, Hy-line W-36 hens (n = 1080) in the second cycle (98 week old) were randomly divided into 9 treatments (8 replicates of 15 hens per treatment). Replicates were equally distributed into upper and lower cage levels to minimize cage level effect. Three hens were housed in a 40.6 cm × 45.7 cm cage and five adjoining cages consisted of a group. All hens were housed in an environmentally controlled house with temperature maintained at approximately 25.6°C (21.1°C during the night and 28.9°C during the day). The house had controlled ventilation and lighting (16 hr/day). All hens were supplied with feed and water *ad libitum*. Egg production was recorded daily, feed consumption was recorded weekly, and egg specific gravity and egg weight was recorded bi-weekly. Egg weight and egg specific gravity were measured using all eggs produced during two consecutive days. Egg specific gravity was determined using 11 gradient

[1] Degussa Corporation, GA

86 saline solutions varying in specific gravity from 1.060 to 1.100 incremented by 0.005-
87 unit increments [15]. Mortality was determined daily and egg production and feed
88 consumption were adjusted to a hen-day basis. Body weight was obtained by weighing 3
89 hens per group at the end of the experiment. Egg mass and feed conversion (g feed/g egg)
90 were calculated from egg production, egg weight and feed consumption

91 Data was analyzed by factorial design in the General Linear Models (GLM) in
92 SAS/STAT [16]. The statistical model was:

93 $$Y_{ijk} = \mu + P_i + L_j + PL_{ij} + e_{ijk}$$

94 where Y_{ijk} is the *k*-th response for the treatment combination P_iL_j, μ is the overall
95 mean, P_i is protein level effect, L_j is added synthetic lysine effect, PL_{ij} is the interaction
96 of protein level and added synthetic lysine, e_{ijk} is the random error. If differences in
97 treatment means were detected by ANOVA, Duncan's Multiple Range Test was applied
98 to separate means. Statements of statistical significance are based on a probability of ($P \leq$
99 0.05). Contrast statements were utilized to test for added lysine linear or quadratic effects.

100

RESULTS AND DISCUSSION

Experiment 1

The total mortality was 0.7% (10 hens out of 1,440), and neither protein nor lysine had a significant effect on mortality (Table 3). There were no interactions between protein and added synthetic lysine on feed intake, egg production, egg weight, egg mass, feed conversion, mortality and final body weight (Table 3). There were significant protein effects on feed intake, egg weight, egg mass, egg specific gravity, and body weight (Table 3). Feed consumption in hens fed the diet containing 16.01% protein was similar to that of hens fed the diet containing14.86% protein, but was significantly higher than that of hens fed the diet containing 14.42% protein. Egg mass and egg weight of hens fed the diet containing 14.89% protein were similar to those of hens fed the diet containing14.42% protein, and both were significantly lower than those of the diet containing 16.01% protein. Hens fed the diet containing 14.42% protein had significant higher egg specific gravity and lower body weight than hens fed the diets containing 14.86% and 16.01% protein.

The addition of synthetic lysine while maintaining a 0.75 Met+Cys/Lys ratio had no significant effect on egg production, egg mass, feed intake, feed conversion, egg specific gravity and final body weight of hens (Table 3). This result was in agreement with those of Sohail et al. [12] and Liu et al. [13], who observed that adding synthetic lysine while maintaining an optimal Met+Cys/Lys ratio had no influence on performance of laying hens. In Experiment 1, lysine intake of hens fed the 16.01, 14.86, and 14.42% protein diets without added synthetic lysine were 849, 767, and 730 mg/h/d respectively, which was higher than NRC recommended lysine requirements (690 mg/h/d) (Table 5)

[7]. Calculation of data from Liu et al. [13] showed that lysine intakes of hens fed the 17.52, 16.24, and 15.22% protein diets without added synthetic lysine were 949, 835, and 741 mg/h/d respectively. Calculation of data from Sohail et al. [12] showed that lysine intakes of hens fed the 18.04, 17.34, 16.65, 16.10, and 15.54 % protein diets without added synthetic lysine were 787, 754, 719, 683, and 640 mg/h/d respectively.

Experiment 2

Feed consumption: There was no interaction between protein and added synthetic lysine on feed intake (Table 4). Feed consumption was not significantly affected by protein. Adding synthetic lysine while maintaining a 0.75 Met+Cys/Lys ratio had a significant effect on feed intake. Feed consumption of hens fed the diets supplemented with 0.030% lysine was similar to that of hens fed the diets supplemented with 0.059% lysine, but was significantly higher than that of hens fed the diets without added synthetic lysine.

Egg production: There was no interaction between protein and added synthetic lysine on egg production (Table 4). Protein effect was not significant. Adding synthetic lysine while maintaining a 0.75 Met+Cys/Lys ratio showed a significant linear effect on egg production. As added synthetic lysine increased from 0.000% to 0.059%, egg production linearly increased from 66.91% to 70.45%.

Egg mass: No interaction between protein and added synthetic lysine on egg mass was observed (Table 4). Protein effect on egg mass approaching significance was observed ($P < 0.08$). Hens fed 14.87% protein diets had the highest overall average egg mass (46.12 g/h/d). Adding synthetic lysine while maintaining a 0.75 Met+Cys/Lys ratio had a significant linear effect on egg mass. As added synthetic lysine increased from

147 0.000% to 0.059%, egg mass linearly increased from 43.57 to 46.04 g, resulting a net
148 increase of 2.47 g

149 ***Egg weight:*** There was no interaction between protein and added synthetic lysine
150 on egg weight (Table 4). There was no significant protein effect on egg weight. Adding
151 synthetic lysine while maintaining a 0.75 Met+Cys/Lys ratio had no significant effect on
152 egg weight.

153 ***Feed conversion:*** No interaction between protein and added synthetic lysine on
154 feed conversion were observed (Table 4). Protein effect on feed conversion was not
155 significant. Adding synthetic lysine while maintaining a 0.75 Met+Cys/Lys ratio had a
156 significant linear effect on feed conversion. As dietary lysine increased, feed conversion
157 linearly decreased from 2.16 to 2.06.

158 ***Egg specific gravity, final body weight, and mortality****:* There was no interaction
159 between protein and added synthetic lysine on egg specific gravity, body weight or
160 mortality (Table 4). Protein effects on specific gravity and mortality were not significant.
161 Adding synthetic lysine while maintaining a 0.75 Met+Cys/Lys ratio had a significant
162 effect on egg specific gravity. Egg specific gravity of hens fed the diets supplemented
163 with 0.059% lysine was similar to that of hens fed the diets supplemented with 0.030%
164 lysine, and both were significantly lower than that of hens fed the diets without added
165 lysine. Total mortality was 1.9% (20 hens out of 1,080), and mortality was not
166 significantly affected by protein or added synthetic lysine (Table 4).

167 In Experiment 2, the addition of synthetic lysine while maintaining a 0.75
168 Met+Cys/Lys ratio had significant effects on feed intake, egg production, egg mass, feed
169 conversion, and egg specific gravity (Table 4). These results was consistent with those of

Liu et al. [14], who reported that adding synthetic lysine while maintaining a 0.75 Met+Cys/Lys ratio had a significant effect on performance of laying hens. In Experiment 2, lysine intakes of hens fed the 14.87, 14.44 and 14.00% protein diets without added synthetic lysine were 710, 686, and 629 mg/h/d respectively, which were close to or lower than NRC recommended lysine intake value (690 mg/h/d) (Table 5) [7]. Calculation of data from Liu et al. [14] showed that lysine intakes of hens fed the 14.3 and 13.6% protein diets without added synthetic lysine were 689 and 592 mg/h/d respectively.

Different protein levels of diets or lysine intakes of hens might explain the inconsistent results among Experiment 1, Experiment 2 and other studies [12, 13, 14]. Compared to the high protein diets, the low protein diets contained less natural lysine in ingredients (Table 1 and 2). Hens fed the low protein diets consumed less feed intake than hens fed the high protein diets (Table 3 and 4). As protein level decreases, lysine intake of hens decreases. In addition, feed intake, which can be affected by many factors such as energy level of diets and environmental temperatures, may affect lysine intake of laying hens. Because feed intake in Experiment 1 was higher than that in Experiment 2, lysine intake in Experiment 1 was higher than that in Experiment 2 (Table 5). Results of Experiment 2 and Liu et al. [14], which showed significant effects of adding synthetic lysine while maintaining a 0.75 Met+Cys/Lys ratio on performance of laying hens, differed from those of Experiment 1, Sohail et al. [12] and Liu et al. [13] possibly because lysine intake or protein level in Experiment 1, Sohail et al. [12] and Liu et al. [13] was higher than that in Experiment 2 and Liu et al. [14]. Therefore, when protein level of a corn-soy diet is below 15% or lysine intake is less than 720 mg/h/d, which was

similar to NRC recommended lysine value, the addition of synthetic lysine while maintaining a 0.75 Met+Cys/Lys ratio significantly improved performance of laying hens.

Economic feeding and management developed by Roland et al. [17, 18] was used to evaluate profits of different added synthetic lysine levels at different protein levels. In Experiment 2, profits in hens fed the 14.00 % protein diet supplemented with 0.030% or 0.059% lysine was equal to or superior to profits of hens fed the 14.87% protein diet without added synthetic lysine (Table 6). Therefore, the addition of synthetic lysine while maintaining a 0.75 Met+Cys/Lys ratio may have a positive influence on profits depending upon ingredient prices and value of performance improvements obtained.

CONSLUSIONS AND APPLICATIONS

1. When protein level of a corn-soy diet is below 15% (supplying less than approximately 15 g protein/h/d) or lysine intake is less than 720 mg/h/d, the addition of synthetic lysine while maintaining a 0.75 Met+Cys/Lys ratio can significantly improve performance of laying hens.
2. Although adding synthetic lysine to diets containing less than approximately 15% protein can improve performance and profits depending on value of performance improvements gained and cost of protein and lysine, these results give no indication as to whether or not diets containing less than 15% protein would be economical.

ACKNOWLEGMENTS

The authors thank Ridley Inc., Mankato, MN**,** for funding support of this experiment.

REFERENCES AND NOTES

1. Harms, R. H. and F. J. Ivey, 1993. Performance of commercial laying hens fed various supplemental amino acids in a corn-soy meal diet. J. Appl. Poult. Res., 2:273-282.

2. Barker, D. H. 2003. Ideal amino acid patterns for broiler chicks. Pages 223-225 in amino acids in animal nutrition, 2nd ed. J. P. F. D'Mello, ed. CABI Publishers, Wallingford, UK.

3. Vieira, S. L., A. Lemme, D. B. Goldenberg, and I. Brugalli. 2004. Response of growing broiler to diets with increased sulfur amino acids to lysine ratios at two dietary protein levels, Poult. Sci. 83:1307-1313.

4. Jensen, L. S., C. H. Chang, and L. Falen. 1974. Response to lysine supplementation by laying hens fed practical diets. Poult. Sci. 53:1387-1391.

5. Latshaw, J. W. 1976. Lysine requirement of hens fed diets with corn as the major cereal gain. Poult. Sci. 55:2348-2353.

6. Nathanael, A. S., and J. L. Sell. 1980. Quantitative measurements of the lysine requirement of the laying hen. Poult. Sci. 59:594-597.

7. National Research Council, 1994. Nutrient Requirement for Poultry. 9th rev. ed. National Academy Press, Washington, DC.

8. Shutte, J. B., and W. Swink. 1998. Requirement of the laying hen for apparent fecal digestible lysine. Poult. Sci. 77:697-701.

9. Bateman, A., M. Bryant and D. A. Roland Sr., 2000. Optimal M+C/Lysine ratio for first cycle Phase I commercial Leghorns. Poult. Sci. 79 (Suppl. 1):92. (Abstr.)

10. Yadalam, S. S., 2001. Influence of M+C/L ration, calcium, phosphorus and phytase on performance and profit of commercial Leghorns. M. S. Thesis, Auburn Unversity, Auburn, AL.

11. Yadalam, S. S., M. M. Bryant and D. A. Roland, Sr., 2000. Optimal M+C/Lysine ratio for first cycle Phase I commercial Leghorns. Poult. Sci. 79 (Suppl. 1):92. (Abstr.)

12. Sohail S. S., M. M. Bryant, D. A. Roland, Sr. 2003. Influence of adding synthetic lysine in corn-soy diets for commercial leghorns. Int. J. Poult. Sci. 2(5):335-340.

13. Liu, Z., G. Wu, M. M. Bryant and D. A. Roland, Sr., 2004b. Influence of added synthetic lysine for first Phase second cycle commercial leghorns with the methionine+cysteine/lysine ratio maintained at 0.75. Int. J. Poult. Sci. 3:220-227.

14. Liu, Z., G. Wu, M.M. Bryant, D. A. Roland, Sr. 2005. Influence of added synthetic lysine in low protein diets with the methionine+cystine /lysine ration maintained at 0.75. J. Appl. Poult. Res. (In press).

15. Holder, D. P., and M. V. Bradford. 1979. Relationship of specific gravity of chicken eggs to number of cracked eggs and percent shell. Poultry Sci. 58:250-251.

16. SAS institute. 2000. SAS/STAT User's Guide. SAS Institute Inc., Cary, NC.

17. Roland, D. A. Sr., M. M. Bryant, J. X. Zhang, D. A. Roland, Jr., S. K. Rao and J. Self, 1998. Econometric feeding and management 1. Maximizing profits in Hy-line W-36 hens by optimizing total amino acid intake and environmental temperature. J. Appl. Poult. Res. 7:403-411.

18. Roland, D. A. Sr., M. M. Bryant, J. X. Zhang, D. A. Roland, Jr., and J. Self.

2000. Econometric feeding and management of commercial Leghorns: Optimizing profits using new technology. Pages 463-472 in Egg Nutrition and Biotechnology. J. S. Sim, S. Nakai, and W. Guenter ed. CABI Publishing, CAB International.

TABLE 1. Ingredient and nutrient content of the experimental diets (Experiment 1)

Ingredient (%)	Diet 1	Diet 2	Diet 3	Diet 4	Diet 5	Diet 6	Diet 7	Diet 8	Diet 9	Diet 10	Diet 11	Diet 12
Corn	64.61	64.54	64.48	64.42	68.17	68.10	68.04	67.98	69.50	69.44	69.37	69.31
Soybean meal	22.50	22.50	22.51	22.51	19.55	19.55	19.56	19.56	18.44	18.45	18.45	18.45
$CaCO_3$	7.18	7.18	7.18	7.18	7.19	7.19	7.19	7.19	7.19	7.19	7.19	7.19
Hardshell[1]	2.00	2.00	2.00	2.00	2.00	2.00	2.00	2.00	2.00	2.00	2.00	2.00
Dicalcium phosphate	1.40	1.40	1.40	1.40	1.41	1.41	1.41	1.41	1.42	1.42	1.42	1.42
Poultry oil	1.36	1.36	1.36	1.36	0.76	0.76	0.76	0.76	0.54	0.54	0.53	0.53
NaCl	0.39	0.39	0.39	0.39	0.39	0.39	0.39	0.39	0.39	0.39	0.39	0.39
Vitamin Premix[2]	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
Mineral premix[3]	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
DL-Methionine[4]	0.07	0.09	0.11	0.13	0.04	0.06	0.08	0.10	0.02	0.05	0.07	0.09
L-lysine[5]	0.00	0.04	0.08	0.11	0.00	0.04	0.08	0.11	0.00	0.04	0.08	0.11
Calculated analysis												
CP (%)	16.01	16.05	16.10	16.14	14.86	14.90	14.95	14.99	14.42	14.47	14.52	14.56
ME (kcal/kg)	2852	2852	2852	2852	2852	2852	2852	2852	2852	2852	2852	2852
Ca (%)	4.00	4.00	4.00	4.00	4.00	4.00	4.00	4.00	4.00	4.00	4.00	4.00
Available phosphorus (%)	0.36	0.36	0.36	0.36	0.36	0.36	0.36	0.36	0.36	0.36	0.36	0.36
Methionine (%)	0.34	0.36	0.38	0.40	0.29	0.31	0.34	0.36	0.27	0.30	0.32	0.34
Metionine+Cystine (%)	0.62	0.64	0.67	0.69	0.56	0.58	0.61	0.63	0.54	0.56	0.58	0.61
Lysine (%)	0.83	0.86	0.89	0.92	0.75	0.78	0.81	0.84	0.72	0.75	0.78	0.81
Experimental analysis												
Methionine (%)	0.32	0.34	0.36	0.38	0.27	0.28	0.3	0.34	0.25	0.27	0.29	0.32
Metionine+Cystine (%)	0.58	0.59	0.62	0.63	0.51	0.52	0.53	0.56	0.48	0.50	0.53	0.55
Lysine (%)	0.81	0.82	0.88	0.90	0.69	0.72	0.74	0.77	0.68	0.71	0.77	0.79

[1]Hardshell = large particle (passing US mesh #4 and retained by US mesh #6) $CaCO_3$ supplied by Franklin Industrial Minerals, Lowell, Florida.

[2]Provided per kilogram of diet: vitamin A (as retinyl acetate), 8,000 IU; cholecalciferol, 2,200 ICU; vitamin E (as $_{DL}$-α-tocopheryl acetate), 8 IU; vitamin B_{12}, 0.02 mg; riboflavin, 5.5 mg; $_{D}$-calcium pantothenic acid, 13 mg; niacin, 36 mg; choline, 500 mg; folic acid, 0.5 mg; vitamin B_1 (thiamin mononitrate), 1 mg; pyridoxine, 2.2 mg; d-biotin, 0.05 mg; vitamin K (menadione sodium bisulfate complex), 2 mg.

[3]Provided per kilogram of diet: manganese, 65 mg; iodine, 1 mg; ferrous carbonate, 55 mg; copper oxide, 6 mg; zinc oxide, 55 mg; sodium selenium, 0.3 mg.

[4] DL-methionine calculated as 99.7%.

[5] L-ysince calculated as 78.6%.

TABLE 2. Ingredient and nutrient content of the experimental diets (Experiment 2)

Ingredient (%)	Diet 1	Diet 2	Diet 3	Diet 4	Diet 5	Diet 6	Diet 7	Diet 8	Diet 9
Corn	68.44	68.37	68.31	69.77	69.71	69.64	71.11	71.03	70.96
Soybean meal	19.53	19.53	19.54	18.42	18.42	18.43	17.31	17.33	17.34
$CaCO_3$	7.37	7.37	7.37	7.38	7.37	7.37	7.38	7.38	7.38
Hardshell[1]	2.00	2.00	2.00	2.00	2.00	2.00	2.00	2.00	2.00
Dicalcium phosphate	1.09	1.09	1.09	1.09	1.09	1.09	1.10	1.10	1.10
Poultry oil	0.66	0.66	0.65	0.43	0.43	0.43	0.21	0.21	0.21
NaCl	0.39	0.39	0.39	0.39	0.39	0.39	0.39	0.39	0.39
Vitamin Premix[2]	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
Mineral premix[3]	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
DL-Methionine[4]	0.03	0.06	0.08	0.02	0.05	0.07	0.01	0.03	0.06
L-lysine[5]	0.00	0.04	0.08	0.00	0.04	0.08	0.00	0.04	0.08
Calculated analysis									
CP (%)	14.87	14.91	14.96	14.44	14.48	14.53	14.00	14.06	14.10
ME (kcal/kg)	2852	2852	2852	2852	2852	2852	2852	2852	2852
Ca (%)	4.00	4.00	4.00	4.00	4.00	4.00	4.00	4.00	4.00
Available phosphorus (%)	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.30
Methionine (%)	0.29	0.31	0.34	0.27	0.30	0.32	0.26	0.28	0.30
Metionine+Cystine (%)	0.56	0.58	0.61	0.54	0.56	0.58	0.52	0.54	0.56
Lysine (%)	0.75	0.78	0.81	0.72	0.75	0.78	0.69	0.72	0.75
Experimental analysis									
Methionine (%)									
Metionine+Cystine (%)									
Lysine (%)									

[1]Hardshell = large particle (passing US mesh #4 and retained by US mesh #6) $CaCO_3$ supplied by Franklin Industrial Minerals, Lowell, Florida.

[2]Provided per kilogram of diet: vitamin A, 8,000 IU; cholecalciferol, 2,200 ICU; vitamin E, 8 IU; vitamin B_{12}, 0.02 mg; riboflavin, 5.5 mg; $_D$-calcium pantothenic acid, 13 mg; niacin, 36 mg; choline, 500 mg; folic acid, 0.5 mg; vitamin B_1 (thiamin mononitrate), 1 mg; pyridoxine, 2.2 mg; biotin, 0.05 mg; vitamin K (menadione sodium bisulfate complex), 2 mg.

[3]Provided per kilogram of diet: manganese, 65 mg; iodine, 1 mg; ferrous carbonate, 55 mg; copper oxide, 6 mg; zinc oxide, 55 mg; sodium selenium, 0.3 mg.

[4] DL-methionine calculated as 99.7%.

[5] L-lysince calculated as 78.6%.

284 TABLE 3. Effect of protein and added synthetic lysine on performance of commercial Leghorns (85 to 93 wk of age) in Experiment 1

Factor		Feed intake (g/hen/day)	Egg production (%)	Egg weight (g)	Egg mass (g)	Feed conversion (g feed/g egg)	Egg specific gravity (unit)	Body weight (kg)	Mortality (%)
Protein	16.01%	103.62[a]	77.57	66.88[a]	51.87[a]	2.00	1.0767[a]	1.69[a]	0.63
	14.86%	102.12[ab]	75.63	65.92[b]	49.87[b]	2.05	1.0769[a]	1.67[a]	0.21
	14.42%	101.18[b]	75.28	65.48[b]	49.30[b]	2.06	1.0775[b]	1.55[b]	1.46
Lysine*	0.000%	102.00	75.64	65.84	49.82	2.05	1.0772	1.68	0.28
	0.030%	103.27	77.26	65.93	50.94	2.03	1.0770	1.62	0.56
	0.059%	101.72	75.93	66.53	50.52	2.02	1.0769	1.64	1.94
	0.088%	102.24	75.81	66.08	50.11	2.05	1.0770	1.62	0.28
16.01% Pro	0.000% Lys	102.32	76.98	67.04	51.63	1.99	1.0761	1.78	0.00
16.01% Pro	0.030% Lys	103.58	78.11	66.44	51.89	2.00	1.0767	1.74	0.83
16.01% Pro	0.059% Lys	104.44	77.96	67.40	52.52	1.99	1.0769	1.61	1.67
16.01% Pro	0.088% Lys	104.15	77.22	66.64	51.45	2.03	1.0770	1.64	0.00
14.86% Pro	0.000% Lys	102.30	74.95	65.27	48.94	2.09	1.0779	1.58	0.00
14.86% Pro	0.030% Lys	103.88	76.86	66.40	51.03	2.04	1.0774	1.64	0.83
14.86% Pro	0.059% Lys	100.30	76.13	66.29	50.47	1.99	1.0762	1.75	0.00
14.86% Pro	0.088% Lys	101.98	74.59	65.73	49.04	2.09	1.0763	1.72	0.00
14.42% Pro	0.000% Lys	101.37	74.98	65.22	48.90	2.08	1.0778	1.69	0.83
14.42% Pro	0.030% Lys	102.34	76.81	64.94	49.91	2.05	1.0770	1.46	0.00
14.42% Pro	0.059% Lys	100.41	73.72	65.89	48.57	2.07	1.0775	1.55	4.17
14.42% Pro	0.088% Lys	100.60	75.62	65.88	49.84	2.02	1.0777	1.52	0.83
Pooled SEM		0.93	1.44	0.52	1.08	0.027	0.0004	0.05	0.64
Two-way ANOVA		----------Probability----------							
Protein		0.0413	0.0584	0.0011	0.0029	0.1008	0.0119	0.0348	0.1565
Lysine		0.5092	0.4940	0.3972	0.6041	0.7313	0.6945	0.7231	0.0935
Protein × Lysine		0.6518	0.9121	0.6219	0.8456	0.5593	0.0211	0.1772	0.2432

285 *means the supplemental lysine level

286 [a-b] Means within a column and under each main effect with no common superscripts differ significantly

287

288

289 TABLE 4. Effect of protein and added synthetic lysine on performance of commercial Leghorns (98 to 110 wk of age) in Experiment 2

Factor		Feed intake (g/hen/day)	Egg production (%)	Egg weight (g)	Egg mass (g/hen/day)	Feed conversion (g feed/g egg)	Egg specific gravity (unit)	Body weight (kg)	Mortality (%)
Protein	14.87%	95.57	70.20	65.70	46.12	2.08	1.0723	1.68[a]	0.83
	14.44%	95.70	68.77	65.26	44.88	2.14	1.0720	1.66[a]	2.22
	14.00%	93.92	68.47	64.68	44.32	2.13	1.0723	1.59[b]	2.50
Lysine*	0.000%	93.71[b]	66.91[b]	65.05	43.57[b]	2.16[a]	1.0729[a]	1.65	1.94
	0.030%	96.65[a]	70.09[a]	65.26	45.71[a]	2.12[ab]	1.0716[b]	1.63	2.50
	0.059%	94.82[ab]	70.45[a]	65.33	46.04[a]	2.06[b]	1.0721[b]	1.64	1.11
14.87% Pro	0.000% Lys	94.61	67.70	65.65	44.47	2.13	1.0724	1.68	1.67
14.87% Pro	0.030% Lys	97.31	72.13	65.71	47.34	2.06	1.0721	1.70	0.83
14.87% Pro	0.059% Lys	94.80	70.77	65.75	46.55	2.04	1.0724	1.64	0.00
14.44% Pro	0.000% Lys	95.30	67.53	65.06	43.95	2.17	1.0729	1.71	1.67
14.44% Pro	0.030% Lys	97.72	69.14	65.29	45.10	2.17	1.0709	1.61	4.17
14.44% Pro	0.059% Lys	94.06	69.64	65.43	45.60	2.07	1.0722	1.66	0.83
14.00% Pro	0.000% Lys	91.23	65.51	64.44	42.30	2.17	1.0735	1.57	2.50
14.00% Pro	0.030% Lys	94.91	68.99	64.79	44.69	2.13	1.0719	1.58	2.50
14.00% Pro	0.059% Lys	95.62	70.92	64.80	45.97	2.09	1.0716	1.63	2.50
Pooled SEM		0.88	1.32	0.57	0.99	0.03	0.0004	0.027	0.8731
Two-way ANOVA		----------Probability----------							
Main effects									
Protein		0.1669	0.2382	0.0960	0.0835	0.1161	0.7109	0.0263	0.1750
Lysine		0.0245	0.0027	0.8205	0.0065	0.0113	0.0050	0.7858	0.3342
Protein × Lysine		0.2228	0.5747	0.9988	0.7316	0.7877	0.2118	0.2754	0.4945
Contrasts									
Lysine linear		0.2833	0.0017	0.5489	0.0034	0.0031	0.3899	0.7712	0.3761
Lysine quadratic		0.0116	0.1362	0.8550	0.2033	0.7683	0.4099	0.5324	0.2353

290 *means the supplemental lysine level

291 [a-b] Means within a column and under each main effect with no common superscripts differ significantly

292 TABLE 5. Influence of adding synthetic lysine on lysine intake of laying hens

Experiment 1				Experiment 2			
Diet	Protein level (%)	Lysine level (%)	Lysine intake (mg/h/day)	Diet	Protein level (%)	Lysine level (%)	Lysine intake (mg/h/day)
1	16.01	0.000	849	1	14.87	0.000	710
2	16.01	0.030	891	2	14.87	0.030	759
3	16.01	0.059	930	3	14.87	0.059	768
4	16.01	0.088	958				
5	14.86	0.000	767	4	14.44	0.000	686
6	14.86	0.030	810	5	14.44	0.030	733
7	14.86	0.059	812	6	14.44	0.059	734
8	14.86	0.088	857				
9	14.42	0.000	730	7	14.00	0.000	629
10	14.42	0.030	768	8	14.00	0.030	683
11	14.42	0.059	783	9	14.00	0.059	717
12	14.42	0.088	815				

293

294 TABLE 6. Influence of protein level and added synthetic lysine[1] on profits[2] (Experiment 2)

Returns[3] (cents/dozen)	Added synthetic lysine level (%)		
	0.000	0.030	0.059
Protein 14.87 (%)	14.44	15.10	15.06
Protein 14.44 (%)	14.08	13.97	14.94
Protein 14.00 (%)	14.30	14.54	14.62

295 [1] Synthetic lysine price was $95/100lb

296 [2] The egg price spread between medium and large eggs was 11 cents

297 [3]Returns (R) were calculated using the equation: R = UBEP - NR -PC - FdC, where UBEP = Urner Barry Egg Price, NR = nest run
298 into package product delivered, PC = production cost, and FdC = feed cost, as described by Roland et al. [17].