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Influence of Energy Level and Glycine Supplementation on Performance, Nutrient Digestibility and Egg Quality in Laying Hens

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ABSTRACT : Sixty four, 30-week-old, Lohmann Brown-Lite laying hens were randomly allocated to one of four treatments with eight replicates per treatment and two hens per replicate for a 10 week study. The control diet was a high energy (11.81 MJ/kg) diet and the moderate energy (11.39 MJ/kg) diets were formulated dropping the level of animal fat. The three moderate energy diets were fed either unsupplemented (0.0%) or supplemented with 0.05 or 0.10% glycine. There were no significant differences ($p>0.05$) in hen-day egg production, egg weight, feed intake or feed conversion between birds fed the unsupplemented moderate energy and high energy diets. Significant differences were detected concerning egg components and quality measurements as assessed by albumen percentage ($p = 0.02$), yolk weight ($p = 0.02$), yolk percentage ($p<0.01$), yolk to albumen ratio ($p<0.01$) and yolk color ($p = 0.01$) between birds fed the unsupplemented moderate and high energy diets. Glycine supplementation of the moderate energy diet linearly increased ($p<0.01$) egg weight and feed intake with no significant ($p>0.05$) effects on egg production or feed conversion. Glycine supplementation significantly increased egg content ($p<0.01$), albumen weight ($p<0.01$) and percentage ($p<0.01$) as well as yolk weight ($p<0.01$) while yolk percentage ($p = 0.04$), yolk to albumen ratio ($p = 0.01$) and egg shell percentage ($p<0.01$) were linearly decreased. Supplementation with glycine produced a tendency ($p = 0.09$) towards an increase in the percentage of large eggs (63-72.9 g) produced with a concomitant decrease in the percentage of small (below 53 g) eggs ($p = 0.09$). The overall results of this study indicate that glycine supplementation of laying hen rations has the potential to increase egg production and weight. These increases appeared to be mediated through increases in feed intake and the ileal digestibility of fat and energy. (**Key Words :** Laying Hens, Glycine, Egg Quality, Egg Production, Egg Weight, Ileal Digestibility)

INTRODUCTION

Amino acids are utilized by poultry to fulfill a diversity of functions. Amino acids are constituents of structural and protective tissues such as skin, feathers, and bone as well as soft tissues like organs and muscles (NRC, 1994). In addition, amino acids may serve a variety of metabolic functions and as precursors for many important non-protein body constituents. There are at least 20 amino acids in body proteins and all are physiologically essential (NRC, 1994). However, some amino acids can be synthesized by using carbon skeletons and amino groups derived from other amino acids present in the diet in excess of requirements. Amino acids synthesized in this manner are termed non-essential. Amino acids that cannot be synthesized or cannot be synthesized at a sufficient rate to permit optimal growth or reproduction are termed essential (NRC, 1994).

Although glycine has been categorized as a non-essential amino acid in poultry, it has been suggested that under certain circumstances there might be insufficient glycine to allow for maximum performance (Corzo et al., 2004; Dean et al., 2006; Powell et al., 2009; Waguespack et al., 2009). Glycine has a series of metabolic functions in addition to its usual role in protein synthesis. It is readily converted to the amino acid serine and is involved in the synthesis of purines (Rowe et al., 1978), the porphyrin moiety of heme groups (Shemin and Rittenberg, 1945), glutathione (te Braake et al., 2008) and creatine (Wyss and Kaddurah-Daouk, 2000). Additionally, glycine is an integral precursor of uric acid (Corozo et al., 2004) which is the excretory medium for 60-80% of urinary nitrogen in birds (Bondi, 1987). Glycine can also play a role in relation to bile salt metabolism (Powell et al., 2009).

The role of glycine in laying hens has not been widely studied. Therefore, the aim of the present study was to

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evaluate the effects of crystalline glycine supplementation on performance, interior and exterior egg quality, nutrient digestibility and relative organ weight in laying hens.

MATERIALS AND METHODS

All procedures used in this experiment were approved by the Animal Ethics Committee of Sungkyunkwan University (Suwon, Korea) and complied with the Guidelines for the Care and Use of Animals in Research published by the Korean Ministry for Food, Agriculture, Forestry and Fisheries (2008).

Experimental design, birds and housing

Sixty four, 30-week-old, Lohmann Brown-Lite laying hens (Join Farm, Songtan, Korea), with an average laying rate of 99.3±5.84%, were randomly allocated to one of four treatments with eight replicates per treatment and two hens per replicate (Table 1). The control diet was a high energy (11.81 MJ/kg) diet formulated with corn, wheat and animal fat as energy sources and with soybean meal, canola meal, corn gluten meal and meat and bone meal providing supplementary protein. Three moderate energy (11.39 MJ/kg) diets were formulated using similar ingredients but dropping the level of animal fat from 2.99 to 0.90%. With the exception of energy, all nutrients met or exceeded the nutrient requirements (Table 2) suggested in the Lohmann Brown Management Guide (Lohmann Tierzucht GmbH, 2008). The higher energy diet was formulated with a MEn level approximately 0.42 MJ/kg higher than the level recommended by Lohmann while the moderate energy diets were formulated with MEn levels approximately 0.21 MJ/kg lower than the level recommended by Lohmann. The three moderate energy diets were fed either unsupplemented (0%) or supplemented with 0.05 or 0.10% glycine (G7126, Sigma-Aldrich, St Louis, MO). The unsupplemented high energy diet provided 0.88% glycine while the unsupplemented moderate energy diet provided 0.87% glycine. The experimental diets were fed in mash form throughout the 10 week study.

All hens were housed in a windowless and environmentally controlled room with the room temperature kept at 21-23°C and the light cycle set at 16 h of light (incandescent lighting, 10 lux) and 8 h dark. The cages were galvanized metal wire (approximately 43×35×50 cm) in double-decker rows providing 750 cm^2 per hen. For this experiment, one upper deck of cages and one lower deck of cages were used. Each cage had a nipple waterer.

Feed and water were available *ad libitum* throughout the experiment. A continuous, plastic feed trough was divided by replicate to ensure that the hens were not able to consume feed assigned to the adjoining replicate. Feed consumption was measured on a weekly basis.

Sampling and analyses

A wire egg collector was installed in the front of each cage to prevent eggs from separate replicates from being mixed. Eggs produced by each replicate were collected and

Table 1. Ingredient composition of diets fed to determine the effects of glycine supplementation on the performance of laying hens (% as fed)

Energy level	High	Moderate		
Glycine level (%)	0.00	0.00	0.05	0.10
Corn	50.02	52.50	52.45	52.40
Soybean meal	22.34	21.94	21.94	21.94
Wheat	10.00	10.00	10.00	10.00
Canola meal	1.50	1.50	1.50	1.50
Meat and bone meal	1.50	1.50	1.50	1.50
Corn gluten meal	1.00	1.00	1.00	1.00
Limestone	8.56	8.56	8.56	8.56
Oyster shell	1.40	1.40	1.40	1.40
Animal fat	2.99	0.90	0.90	0.90
Vitamin-mineral premix[a]	0.23	0.23	0.23	0.23
Salt	0.20	0.20	0.20	0.20
Sodium bicarbonate	0.06	0.06	0.06	0.06
Phytase	0.05	0.05	0.05	0.05
Methionine	0.09	0.09	0.09	0.09
L-lysine HCl	0.04	0.04	0.04	0.04
Glycine	0.00	0.00	0.05	0.10

[1] Provided the following nutrients per kg diet: vitamin A, 12,000 IU; vitamin D_3, 3,500 IU; vitamin E, 30 IU; vitamin K_3, 3.0 mg; thiamine, 3.0 mg; riboflavin, 7.0 mg; pyridoxine, 5.0 mg; vitamin B_{12}, 0.025 mg; niacin, 40.0 mg; pantothenic acid, 10 mg; folic acid, 1.0 mg; biotin, 0.15 mg; Fe, 75.0 mg; Zn, 97.5 mg; Mn, 97.5 mg; Cu, 7.5 mg; I, 1.5 mg; Se, 0.2 mg.

Table 2. Chemical composition of diets fed to determine the effects of glycine supplementation on the performance of laying hens (% as fed)[1]

Energy level	High	Moderate	Moderate	Moderate
Glycine level (%)	0	0	0.05	0.10
Dry matter	92.99	92.20	92.26	92.15
MEn (MJ/kg)	11.81	11.39	11.39	11.39
Ether extract	5.85	4.05	4.07	4.12
Ash	13.79	12.00	12.23	12.03
Calcium	4.28	4.21	4.18	4.12
Total phosphorus	0.53	0.49	0.50	0.49
Crude protein	17.56	16.94	17.08	16.99
Arginine	1.14	1.06	1.01	0.98
Histidine	0.42	0.40	0.39	0.37
Isoleucine	0.66	0.62	0.58	0.55
Leucine	1.43	1.39	1.32	1.28
Lysine	0.86	0.79	0.76	0.73
Methionine+cystine	0.59	0.59	0.58	0.60
Phenylalanine	0.74	0.71	0.67	0.64
Threonine	0.66	0.62	0.60	0.58
Valine	0.78	0.75	0.73	0.69
Glycine	0.88	0.87	0.93	1.00
Glycine+serine	1.78	1.70	1.78	1.87

[1] With the exception of ME, all data are the results of a chemical analysis conducted in triplicate.

weighed every day. Egg weight (g of egg/hen per day) and feed conversion (g of feed/g of egg) were calculated from egg production, egg weight, and feed consumption.

Egg components (the percentages of albumen, yolk and shell) were measured using two eggs of average weight obtained from each cage on the last two days of each week. Yolk color and Haugh units were measured using an Egg Multi-Tester EMT-5200 (Robotmation Co. Ltd., Tokyo, Japan). HU were calculated from the records of egg weight and albumen height using the formula: $HU = 100 \log10 (H - 1.7 W^{0.37}+7.56)$, where H = height of the albumen (mm), and W = egg weight (g). The yolks were separated from the tester tray (yolk, albumen and tray) using a Teflon spoon. Before the yolk weight was determined, the chalaza was removed by spatula. Albumen weight was calculated by subtracting the weight of the tester tray from the remaining egg contents. The egg shells were weighed without drying. Total egg content was calculated (% egg production×(daily yolk weight+daily albumen weight) from egg production, yolk weight and albumen weight. Eggs from each cage were sorted into four size groups (very large, large, medium, and small) according to the European Union Marketing Standards (European Commission, 2008).

At the end of the 10-week experiment, all diets were removed from the feeders and replaced with similar diets containing the indigestible marker chromic oxide (0.4%). Seven days later, all birds in each replicate were weighed and euthanized using CO_2. The gastrointestinal tract, proventriculus, gizzard, ceca, liver and the pancreas were removed aseptically and any digesta present was removed. After digesta emptying, each organ was weighed. Empty body weight was also determined. The weight of the organs was expressed relative to live body weight.

All of the ileal digesta between the yolk sac and the terminal ileum were scraped and flushed with deionised water from the intestinal tract immediately after slaughter. Ileal digesta from the two hens in each replicate were freeze dried, mixed together and ground through a 1.0 mm mesh screen before analyses.

The feed and digesta were analyzed in triplicate according to the methods of the AOAC (1995). Analyses were conducted for moisture (Method 930.15), ether extract (Method 920.39), crude protein (Method 984.13), ash (Method 942.05) and ether extract (Method 920.39). Calcium was determined by a Shimazu AA625 Atomic Absorption Spectrophotometer (Shimazu, Kyoto, Japan), and phosphorus was analyzed using a UV-vis. Spectrophotometer (Hitachi, Tokyo, Japan). Amino acid analysis was performed using a L8500-Hitachi Amino Acid Analyzer (Hitachi, Tokyo, Japan) after hydrolysis for 24 h in 6 N HCl. Performic acid (85%) hydrolysis was performed for analysis of sulfur-containing amino acids. Chromic oxide was determined by the method of Fenton and Fenton (1979). Digestibility coefficients for nutrients were calculated using the equations for the indicator method described by Schneider and Flatt (1975).

Table 3. Effects of energy level and glycine supplementation on the performance of laying hens

Energy level	High	Moderate			SEM	p-values		
Glycine level (%)	0.00	0.00	0.05	0.10		Energy level	Glycine level	
							Linear	Quadratic
Hen weight at start (g)	1,957	1,951	1,946	1,959	19.3	0.83	0.72	0.63
Hen weight at end (g)	1,959	1,948	1,940	1,980	48.7	0.88	0.67	0.71
Egg production (%)	97.6	97.5	98.3	99.5	0.82	0.85	0.07	0.92
Egg weight (g/hen/d)	58.7	58.2	59.3	62.7	0.64	0.62	<0.01	0.16
Feed intake (g/hen/d)	111.8	111.7	112.5	118.4	0.96	0.96	<0.01	0.03
Feed conversion (g feed/g egg)	1.91	1.93	1.90	1.90	0.03	0.53	0.56	0.54

Statistical analysis

Data were analyzed as a randomized block design (Snedecor and Cochran, 1989) using the appropriate Analysis of Variance procedures of Statistix (1996). Hens were blocked on the basis of hen-day egg production during the adaptation period and the cage was considered the experimental unit for all analyses. The model included the effects of replication (i.e., block), treatment, and replication ×treatment (error). The significance of differences between means was determined by the Least Significant Difference (LSD) method for data at the level of $\alpha = 0.05$. Polynomial contrasts were constructed to determine the nature of response variables to increasing levels of supplemental glycine and energy level.

RESULTS

There were no significant differences in hen-day egg production, egg weight, feed intake or feed conversion between birds fed the unsupplemented moderate and high energy diets (Table 3). Glycine supplementation of the moderate energy diet linearly increased ($p<0.01$) egg weight and feed intake with no significant effects on egg production or feed conversion.

The effects of energy level and glycine supplementation on egg components and egg quality are presented in Table 4. Significant differences were detected concerning egg components and quality measurements as assessed by albumen percentage ($p = 0.02$), yolk weight ($p = 0.02$), yolk percentage ($p<0.01$), yolk to albumen ratio ($p<0.01$) and yolk color ($p = 0.01$) between birds fed the unsupplemented moderate energy and high energy diets. Egg content, albumen weight, egg shell weight and percentage as well as HU were not significantly influenced by energy level ($p>0.05$). Glycine supplementation of the moderate energy diet significantly increased egg content ($p<0.01$), albumen weight ($p<0.01$) and percentage ($p<0.01$) as well as yolk weight ($p<0.01$) while yolk percentage ($p = 0.04$), yolk to albumen ratio ($p = 0.01$) and egg shell percentage ($p<0.01$) were linearly decreased. Egg shell weight, yolk color and Haugh units were unaffected by level of glycine supplementation ($p>0.05$).

The effects of energy level and glycine supplementation on nutrient digestibility are presented in Table 5. Energy level had no effect on the ileal digestibility of any of the parameters measured ($p>0.05$). Supplementation with glycine significantly increased fat ($p<0.01$) and energy ($p<0.01$) digestibility while the ileal digestibility of organic matter, crude protein and all of the essential amino acids

Table 4. Effects of energy level and glycine supplementation on egg components and egg quality of laying hens

Energy level	High	Moderate			SEM	p-values		
Glycine level (%)	0.00	0.00	0.05	0.10		Energy level	Glycine level	
							Linear	Quadratic
Weight of egg contents (g)	50.8	50.5	51.5	54.6	0.57	0.60	<0.01	0.18
Weight of egg albumen (g)	35.3	35.1	36.1	38.7	0.44	0.68	<0.01	0.12
Percentage albumen (%)	60.6	59.8	60.8	61.7	0.24	0.02	<0.01	0.38
Weight of egg yolk (g)	15.7	15.1	15.4	15.9	0.19	0.02	<0.01	0.75
Percentage yolk (%)	26.8	26.0	26.1	25.4	0.19	<0.01	0.04	0.17
Yolk to albumen ratio (%)	45.0	43.1	43.0	41.4	0.47	<0.01	0.01	0.23
Weight of egg shell (g)	7.8	7.8	7.8	8.1	0.10	0.86	0.09	0.14
Percentage shell (%)	13.4	13.5	13.1	12.9	0.12	0.72	<0.01	0.67
Yolk color	7.2	6.9	7.0	7.0	0.09	0.01	0.12	0.84
Haugh units	86.1	84.8	84.0	84.3	0.85	0.28	0.64	0.60

Table 5. Effects of energy level and glycine supplementation on egg quality and egg size for laying hens

Energy level	High	Moderate			SEM	p-values		
Glycine level (%)	0.00	0.00	0.05	0.10		Energy level	Glycine level	
							Linear	Quadratic
Normal eggs (%)[1]	48.95	52.43	49.57	45.18	8.43	0.79	0.54	0.94
Egg grade (%)								
Very large (above 70 g)	0.31	0.11	0.11	0.21	0.15	0.40	0.52	0.72
Large (63-72.9 g)	25.60	25.74	26.68	48.94	8.94	0.99	0.09	0.36
Medium (53-62.9 g)	69.90	61.87	70.24	50.33	8.67	0.40	0.37	0.21
Small (below 53 g)	4.09	12.28	2.98	0.52	4.25	0.34	0.09	0.56

[1] Without stains, cage marks, pimples, pinholes, misshapen, sandpaper or rough shells, body-cracked, mottled or glassy shells, gross cracks, hairline cracks, star cracks and thin-shelled eggs, fly marks or flat-sided eggs.

were unaffected by supplementation with glycine (p>0.05).

The percentage of normal eggs (i.e. eggs without stains, cage marks, pimples, pinholes, misshapen or thin shells, as well as cracks) was not affected (p>0.05) by energy level or supplementation with glycine (Table 6). In addition, egg grade was not influenced by energy level. However, there was a linear trend (p = 0.09) towards a greater percentage of large size eggs (63-72.9 g) and a linear reduction (p = 0.09) in the percentage of small size eggs (below 53 g) as a result of glycine supplementation.

The effects of energy level and glycine supplementation on the relative weight of the organs are shown in Table 7. Neither energy level nor glycine supplementation had any significant effects on empty body weight or the relative weights of the gastrointestinal tract, proventriculus, gizzard, ceca, liver or pancreas (p>0.05).

DISCUSSION

There were no significant differences in feed intake, feed conversion, hen-day egg production or egg weight between birds fed the unsupplemented moderate and high energy diets. A number of studies have been conducted to investigate the effects of dietary energy level on the feed intake of laying hens and it is generally accepted that hens adjust their feed intake to meet their energy requirements (Wu et al., 2005; Van Krimpen et al., 2007; Valkonen et al., 2008; Gunawardana et al., 2009). For example, Grobas et al. (1999) reported that a decrease in dietary energy content from 11.79 to 11.21 MJ of ME/kg diet increased feed intake by 4% while Harms et al. (2000) showed that hens fed diets containing 10.54 MJ of ME/kg had a 8.5% higher feed intake than hens fed diets containing 11.71 MJ of ME/kg.

Table 6. Effects of energy level and glycine supplementation on ileal digestibility (%) of nutrients for laying hens

Energy level	High	Moderate			SEM	p-values		
Glycine level (%)	0.00	0.00	0.05	0.10		Energy level	Glycine level	
							Linear	Quadratic
Organic matter	93.57	93.56	93.11	94.15	0.87	0.98	0.71	0.37
Crude protein	92.78	92.70	92.48	92.59	0.65	0.95	0.82	0.81
Ether extract	92.00	91.30	92.55	93.25	0.36	0.30	<0.01	0.91
Energy	93.18	93.01	93.24	94.86	0.33	0.72	<0.01	0.04
Essential amino acids								
Arginine	94.87	95.26	94.86	95.25	0.57	0.64	0.85	0.52
Histidine	93.90	94.50	93.95	94.61	0.68	0.56	0.96	0.43
Isoleucine	93.30	93.68	93.05	93.82	0.74	0.73	0.96	0.40
Leucine	94.66	95.02	94.60	95.21	0.55	0.65	0.99	0.38
Lysine	92.23	93.03	92.42	92.88	0.88	0.54	0.78	0.60
Methionine	93.70	95.00	94.15	95.59	0.81	0.15	0.77	0.24
Cysteine	84.48	90.11	88.95	93.95	2.20	0.16	0.29	0.16
Phenylalanine	94.19	94.51	94.12	94.64	0.61	0.63	0.92	0.47
Threonine	90.99	91.78	90.98	91.83	1.03	0.57	0.90	0.49
Valine	90.88	92.08	91.22	92.14	0.94	0.44	0.92	0.42
Glycine	93.17	93.97	93.19	94.04	0.77	0.19	0.91	0.38

Table 7. Effects of energy level and glycine supplementation on the relative weight (% of BW) of organs in laying hens

Energy level	High	Moderate			SEM	p-values		
Glycine level (%)	0.00	0.00	0.05	0.10		Energy level	Glycine level	
							Linear	Quadratic
Empty body weight[1]	80.28	80.13	79.94	80.55	0.57	0.90	0.61	0.42
Gastrointestinal tract[2]	6.56	6.68	6.72	6.53	0.15	0.58	0.40	0.39
Proventriculus	0.31	0.32	0.32	0.32	0.01	0.33	0.54	0.72
Gizzard	1.27	1.28	1.26	1.32	0.04	0.99	0.47	0.38
Ceca	0.35	0.36	0.37	0.34	0.02	0.39	0.28	0.52
Liver	2.55	2.55	2.55	2.50	0.13	0.96	0.95	0.81
Pancreas	0.13	0.14	0.14	0.13	0.01	0.47	0.69	0.92

[1] Without the liver and the gastrointestinal tract and its contents. [2] From the end of the crop to the anus, including digesta content.

However, in the present study, there was no significant difference in feed consumption between birds fed the high and moderate energy diets. Our results agree with those of Jalal et al. (2006, 2007) who reported no significant effects of dietary energy level on feed intake. The fact that no significant effect of dietary energy level on feed intake was found in the present experiment suggests that the energy level of the moderate energy diet was adequate to meet the needs of the laying hens under the experimental conditions used in the present study.

The results of studies on the effects of dietary energy on egg laying rate are conflicting. For example, Mathlouthi et al. (2002) reported increased laying rates at an energy content of 11.52 MJ of ME/kg of feed compared with 11.10 MJ of ME/kg of feed while Ciftci et al. (2003) found that decreasing the energy content of the feed from 11.51 to 11.05 MJ of ME/kg increased the laying rate of hens from 86.4 to 88.3%. In the present study, egg production was similar between birds fed the moderate and high energy diets which agrees with the findings of van Krimpen et al. (2007, 2008) and provides further support for the assertion that the energy level of the moderate energy diet was adequate to meet the needs of the laying hens under the experimental conditions used in the present study.

Many researchers have reported that increasing dietary energy level increases egg weight (Keshavarz, 1995; Keshavarz and Nakajima, 1995; Harms et al., 2000; Bohnsack et al., 2002; Sohail et al., 2003; Wu et al., 2005). However, in the present experiment, there was no significant effect of energy level on egg weight. Our findings agree with the results of Summers and Leeson (1993) who reported that egg weight was not changed by increasing dietary energy. According to Bish et al. (1985) differences in the body weight of layers may partly account for the observation of an increase in egg weight as a result of increasing feed energy content. In the present experiment, there was no difference in the weight of the hens fed the moderate and high energy diets and therefore an increase in egg weight would be less likely to be observed.

In the present study, there was a trend towards a linear increase in egg production with increasing levels of glycine supplementation. This finding was somewhat surprising since the rate of production exceeded 95% for all treatments. As a result, there was a very small window of opportunity for glycine to increase the rate of egg production. In commercial poultry production, peak production usually occurs when birds reach 24 to 26 weeks of age and production steadily declines until the flock is taken out of production at approximately 76 weeks of age (Bell, 2002). The present study was conducted relatively close to the period of peak production and it would be interesting to repeat the study to determine whether or not glycine supplementation has further beneficial effects on the productivity of laying hens during the later stages of the production cycle.

One potential explanation for the increase in egg production due to glycine supplementation could be the fact that feed intake showed a significant linear increase with increasing levels of glycine supplementation resulting in a greater supply of nutrients available for egg production. Glycine might be expected to increase the feed intake of laying hens as it is a sweet-tasting crystalline solid (Zeng et al., 1991) and is sold as a sweetener/taste enhancer for the human market. In the present study, increases in feed intake were observed when glycine was supplemented at 0.05 and 0.10% of the diet. However, care should be taken to ensure that glycine is not supplemented at too high a level as previous research has actually show reductions in feed intake when glycine was supplemented in chicken diets at levels as high as 3% of the diet (Cave, 1978; 1983).

Although total egg weight was not significantly affected by energy level, the percentage of the egg weight that comprised the albumen and the yolk as well as yolk weight and yolk to albumen ratio was significantly lower for the unsupplemented moderate energy diet than the high energy diet. The fact that yolk weight was affected to a greater

extent than albumen weight agrees with the finding of Wu et al. (2005). The principle difference in the high and moderate energy diets was that the high energy diet was formulated using 2.99% animal fat while the moderate energy diet was formulated using 0.9% animal fat. Sell et al. (1987) hypothesized that during early lay, laying hens have inadequate hepatic synthesis of lipoprotein for egg yolk development and that by providing exogenous fat, more lipids might be available for egg yolk development.

Egg weight was linearly increased as a result of increases in glycine supplementation. The increase in egg weight appeared to result from increases in both albumen and yolk weight both of which increased linearly with increases in glycine supplementation. The increase in egg weight can most likely be attributed to the increase in feed intake caused by glycine supplementation resulting in more nutrients being available for albumen and yolk development. Research in humans has suggested that glycine plays an important role in the control of the release of growth hormone (Eklund et al., 2005). Responses to increased growth hormone levels are typically an increased efficiency of protein synthesis (Dean et al., 2006) which may also help to explain the increase in albumen and egg yolk weight observed due to glycine supplementation.

An interesting facet of the present study was the fact that the ileal digestibility of ether extract and energy were linearly increased as a result of increases in glycine supplementation. Glycine is a precursor for the production of bile salts (Powell et al., 2009). Primary bile acids are synthesized from cholesterol in the hepatocytes of the liver, and are then conjugated with glycine or taurine in the small intestine to form bile salts. The bile salts play an essential role in digestion and absorption of dietary fat and other fat soluble nutrients (Gomez and Polin, 1976; Stamp and Jenkins, 2008). These results of this study are in agreement with those of Fedde et al. (1960) and Alzawqari et al. (2010), who reported a linear increase in fat digestibility in birds fed diets supplemented with glycine.

In this study, higher dietary energy level did not affect egg shell quality or grade of egg. This agrees with the findings of Grobas et al. (1999) who studied laying hens from 22 to 75 weeks of age and reported that egg shell quality was not affected by dietary energy levels.

In the present study, supplementation with glycine resulted in a tendency for an increase in the percentage of large eggs (63-72.9 g) with a concomitant decrease in the percentage of small (below 53 g) eggs. As many countries pay a premium for larger sized eggs (Food and Agriculture Organization of the United Nations, 2003), there may be an economic incentive for producers to utilize glycine as a means of increasing the percentage of large size eggs produced.

The overall results of this study indicate that glycine supplementation of laying hen rations has the potential to increase egg production and weight. These increases appeared to be mediated through increases in feed intake and the ileal digestibility of fat and energy. Another potential mechanism through which glycine might act to increase egg production and weight is by modulating the inflammatory response. In a recent study, glycine supplementation reduced mRNA expression of pro-inflammatory cytokines such as interleukin-1, interleukin-6 and tumor necrosis factor-α following lipopolysaccharide injection in broiler chickens (Takahashi et al., 2008). Such a response would result in a fewer nutrients being directed towards generation of an immune response and a greater percentage available for productive purposes. If a similar mechanism existed in laying hens, it might explain the increases in productivity observed.

CONCLUSIONS

The addition of 0.05 and 0.10% glycine to laying hen rations providing 11.39 MJ/kg of ME tended to increase egg production and significantly increased egg weight. These increases appeared to be mediated through increases in feed intake and the ileal digestibility of fat and energy. Supplementation with glycine resulted in a tendency for an increase in the percentage of large eggs (63-72.9 g) produced with a concomitant decrease in the percentage of small (below 53 g) eggs. As many countries pay a premium for larger sized eggs, there may be an economic incentive for producers to utilize glycine as a means of increasing the percentage of large size eggs produced.

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