

EFFECT OF DIETARY ENERGY, SYNTHETIC AMINO ACIDS,  
ENZYMES, ANTIBIOTICS, AND MOLTING METHOD  
ON COMMERCIAL LEGHORNS

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EFFECT OF DIETARY ENERGY, SYNTHETIC AMINO ACIDS,  
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ON COMMERCIAL LEGHORNS

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A Dissertation

Submitted to

the Graduate Faculty of

Auburn University

in Partial Fulfillment of the

Requirements for the

Degree of

Doctor of Philosophy

Auburn, Alabama  
August 7, 2006

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## VITA

Guangbing Wu, son of Zhengxi Wu and Honglian Xue was born May 12, 1973. He received his bachelor degree in Agriculture from Shanghai Fisheries University, Shanghai, China. After completion of his degree, he worked as technical manager in the aquaculture industry for two years and as a technical executive at Roche (China) Ltd. for two years. He entered master program under the direction of Dr. Allen D. Davis in Fisheries and Allied Aquaculture, Auburn University, and earned his master of science degree August of 2003. In July of 2003, he entered the Ph. D. Program in Poultry Science under the direction of Dr. David A. Roland, Sr. He will complete his Ph. D. by August of 2006 and plans a career as an animal nutritionist. He is happily married to Kun Yuan, daughter of Shiqun Yuan and Huafang, and is blessed with daughter Linda.

DISSERTATION ABSTRACT  
EFFECT OF DIETARY ENERGY, SYNTHETIC AMINO ACIDS,  
ENZYMES, ANTIBIOTICS, AND MOLTING METHOD  
ON COMMERCIAL LEGHORNS

Guangbing Wu

Doctor of Philosophy, August 7, 2006  
(M. S., Auburn University, Aug 2003)  
(B. S., Shanghai Fisheries University, July 1996)

177 Typed Pages

Directed by David A. Roland, Sr.

A series of experiments were conducted to investigate the effect of dietary energy, antibiotics, and molting method on performance, egg composition, egg solids, egg quality, and profits of current strains of commercial Leghorns. Two experiments were conducted to investigate the effect of adding synthetic amino acids on performance and nutrient utilization of commercial layers. One experiment was conducted to compare the effects of two sources of phytase on performance of commercial Leghorns.

The results showed that increasing dietary energy by the addition of poultry oil to a dietary energy level of 2,877 kcal ME/kg maximized egg weight during Phase I (from 21 to 36 wk of age). Based on feed conversion, increasing dietary energy to 2,864 kcal ME/kg by the addition of poultry oil gave optimal performance of laying hens during Phase II (from 40 to 51 wk of age). An ideal dietary energy level for optimal performance

could not be determined during early post-molt production period (from 70 to 81 wk of age). Based on improved feed conversion, a dietary energy of 2,846 kcal ME/kg gave optimal performance during second cycle phase 2 (from 86 to 95 wk of age). Based on body weight of hens, dietary energy level for optimal performance should be less than 2,936 kcal ME/kg during second cycle phase 3 (from 101 to 110 wk of age). There can be no fixed ideal dietary energy level for optimal profits, due to varying feed ingredient prices and egg price.

When protein level of a corn-soy diet is below 15% (supplying less than approximately 15 g protein/hen per d) or lysine intake is less than 720 mg/hen per d, the addition of synthetic lysine while maintaining a 0.75 Met+Cys/Lys ratio significantly improved 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. While antibiotic supplementation had no effect on performance, antibiotic supplementation significantly reduced body checks and dirty eggs, and increased yolk color, resulting in positive effects on egg quality of laying hens. The addition of phytase significantly increased egg production and egg mass of hens fed the phosphorus deficient diet (0.11% NPP) to levels that were similar to hens fed the control diet containing 0.38% NPP. There was no significant difference in performance between hens fed two phytases. Feeding a no salt diet can produce an acceptable long-term post-molt performance and egg quality, and may be considered as an effective alternative molt method for conventional feed withdrawal.

## ACKNOWLEDGEMENTS

The author would like to thank his major professor, Dr. Roland, for his three years of patient and helpful direction and support in completing this program. The author would like to thank Dr. Davis, Dr. Chiba, and Dr. Farmer for their help and suggestions in completing this program. Thanks are also due to Matilda Bryant and Dwain Holt for their assistance to the experiments.

The author wishes to express his love and gratitude to his wife Kun for her patience and support for the graduate study period. The author also wishes to express his appreciation to his loving child, father and mother, and brother for their moral support during the course of this program.

Style manual or journal used Journal of Poultry Science

Computer software used Microsoft word 2003 and SAS, 9.1

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## I. INTRODUCTION

The primary objective of poultry nutrition research is to obtain optimal performance and profits of broilers and layers, and to decrease potential pollution problems caused by feeding excess nutrients. It is important to know nutrient requirements for maximum performance of broilers and layers. However, in today's competitive market, it is even more important for producers to know requirements for maximum profits, which may or may not be the same as requirements for optimal performance. Optimizing nutrient utilization becomes a necessity, because improving nutrient utilization by supplementing synthetic amino acids, enzymes or fat to diets not only improves performance and profits, but also reduces nutrient waste burden on the environment.

Dietary energy is one of the most important and costly nutrients. There is a wide range of dietary energy levels (2,684–2,992 kcal ME/kg) currently being used by the egg industry. Part of the reason for this is that information is not available that would allow egg producers to know the ideal dietary energy level required for optimal performance and profits. A number of studies have been conducted to investigate dietary energy effect on feed intake of commercial Leghorns. Grobas et al. (1999) reported that an increase in dietary energy from 2,680 to 2,810 kcal ME/kg decreased feed intake by 4%. Harms et al. (2000) showed that hens fed diets containing 2,519 kcal ME/kg had 8.5% more

feed intake than hens fed diets containing 2,798 kcal ME/kg, and hens fed diets containing 3,078 kcal ME/kg had 3.0% less feed intake than hens fed diets containing 2,798 kcal ME/kg.

Many researchers have reported that increasing dietary energy by the addition of corn oil or poultry oil increased early egg weight (Keshavarz, 1995; Keshavarz and Nakajima, 1995; Harms et al., 2000; Bohnsack et al., 2002; Sohail et al., 2003). However, Summers and Leeson (1983) reported that egg weight was not changed by increasing dietary energy or supplementing with fat.

Feed intake and egg weight can significantly affect cost of production and profits. With the sharp increase in dietary energy prices that can occur and did during the past year, it is even more important for egg producers to have information available that would allow them to continually optimize dietary energy use. Although processed egg consumption such as liquid egg products and dried egg solids has steadily increased during the last 10 years, very few studies have been conducted to investigate the dietary energy effect on egg components and egg solids of current strains of Bovans White and Dekalb White. Therefore, a series of experiments (section II, III, IV, V, and VI) were conducted to determine the effect of dietary energy on performance, egg composition, and profits in Bovans White and Dekalb White hens.

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 (Harms et al., 1993). 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 (Barker, 2003; Vieira et al., 2004). 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/hen per d (Jensen et al., 1974; Latshaw, 1976; Nathanael and Sell, 1980; NRC, 1994; and Shutte and Swink, 1998). Some studies (Bateman et al., 2000; Yadalam et al., 2000; Yadalam, 2001) 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 the 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 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. (2003) and Liu et al. (2004) 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 (21-36 wk) of the first cycle hens and in the second cycle hens (70-80 wk), respectively. However, Liu et al. (2005) 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. Two experiments (section VII) were conducted to determine the effect of adding synthetic lysine while maintaining the Met+Cys/Lys ratio at 0.75 on performance and profits for commercial Leghorns.

Phosphorus is an essential mineral for laying hens in formation of eggshell and metabolism (Frost and Roland, 1991; Summers, 1995; Usayran and Balnave, 1995, Sohail and Roland, 2002). Only 20%-50% of plant-derived P is available to broilers, and the rest of P is in the form of phytate (myo-inositol hexaphosphate), which is poorly used by broilers (Ravindran et al. 1998). Ravindran et al. (1998) and Sebastian et al. (1998) reported that poultry cannot produce enough endogenous phytase to hydrolyze and release P from phytate. To meet dietary P requirements of laying hens, inorganic P such as dicalcium phosphate and monocalcium phosphate or exogenous phytase enzymes are commonly added to commercial corn-soy layer diets. However, inorganic P supplementation is not only expensive but also leads to environmental problems by over-supplementation. Excess P from the excreta of hens can easily add to the phosphorus loading of ground water, rivers, lakes, and oceans, and can contribute to eutrophication of aquatic systems and stimulate algae growth resulting in mortality of aquatic animals (Ryden et al., 1973).

Many researchers have demonstrated that phytase supplementation (from 100 to 2000 phytase unit (FTU)/kg of feed) to diets containing 0.1% dietary nonphytate phosphorus (NPP) has positive effects on egg production, egg mass, egg weight, egg specific gravity, bone ash, and eggshell quality by improving P utilization (Van der Klis et al., 1996; Gordon and Roland, 1997, 1998; Boiling et al., 2000a, b; Jalal and Scheideler, 2001; Roland et al., 2003; Keshavarz, 2003). Phytase supplementation

decreased P excretion in the manure and reduced the potential environmental problems (Jalal and Scheideler, 2001).

There are several commercial phytase products including Natuphos (BASF Corp., Mount Olive, NJ) and Ronozyme (Roche Vitamins, Parsippany, NJ) in the market. Natuphos<sup>®</sup> phytase originates from *Aspergillus niger*, and is extensively used in the poultry industry. Recently a new bacterial phytase Phyzyme (Danisco Animal Nutrition, Carol Stream, IL), which originates from the bacteria *Escherichia coli* and is produced by *Schizosacchromyces pombe*, has been introduced into the market. Phytases from different sources may have different biochemical and biophysical properties such as pH-activity profile and sensitiveness to pepsin, which can affect the *in vivo* bioefficacy of phytase.

Very little research has been conducted to evaluate the effect of the novel phytase Phyzyme<sup>®</sup> on commercial Leghorns fed corn-soy diets. An experiment (section VIII) was conducted to compare the effects of two sources of phytase (Phyzyme and Natuphos) on performance of commercial Leghorns fed corn-soy diets from 21 to 33 wk of age.

Antibiotics are substances that inhibit the growth of bacteria and related microorganism by interfering with essential metabolic functions. Antibiotic supplementation in feed increases weight gain and improves feed efficiency of broilers (Miller Publishing Co., 2000, Miles et al., 2006). However, antibiotics had no effect on body weight gain and feed utilization (Izat et al., 1990). Tylosin, an antibiotic, made naturally by the bacterium "*Streptomyces fradiae*" and acts to inhibit bacterial protein synthesis by inhibiting the 50S ribosome, a cellular structure of certain bacteria. Although Tylosin is widely used by poultry industry, there is very limited scientific literature about the effect of Tylosin on performance, egg component, and egg quality of laying hens.

Two experiments (section III and IV) were conducted to determine the effect of antibiotics on performance, egg component, and egg quality of laying hens.

Induced molting, an important management tool, has been widely used to rejuvenate laying hens for a second or third cycle of egg production by the egg industry in US for long time. Induced molting not only improved performance and eggshell quality, but also increased profits by optimizing the use of replacement pullets on commercial layer farms (Lee, 1982; Barker et al., 1983; Bell, 2003). At least 47% more hens would be required to keep houses full with the one-cycle egg production (Bell, 2003). The combination of feed withdrawal and light reduction is most widely used to induce molting in USA egg industry. In general, most producers today use some form of feed withdrawal for periods of 5 to 14 d (Bell and Kuney, 2004). Traditional feed removal method has received considerable attention relative to the animal welfare issues recently. The United Egg producers (UEP) Scientific Advisory Committee on Animal Welfare urged researchers and producers to work together to develop alternatives to feed withdrawal for molting (UEP, 2002). Large egg consumers such as McDonald's, Wendy's and Burger King stated that they would not purchase eggs from producers that use feed withdrawal in their molting programs (Egg industry, 2000; Smith, 2002).

Effectiveness of several nonfeed removal molting diets including low-sodium diets (Whitehead and Shannon, 1974; Begin and Johnson, 1976; Nesbeth et al., 1976a, b; Whitehead and Sharp, 1976; Monsi and Enos, 1977; Ross and Herrick, 1981; Harms 1981, 1983; Naber et al., 1984, Said et al., 1984), low-calcium diets (Gilbert et al., 1981), high-Zinc diets (Shippee et al., 1979; Berry and Brake, 1985; Bar et al., 2003), and low protein and low energy diets (Koelkebeck et al., 2001; Biggs et al., 2003) have been

evaluated. Naber et al. (1984) and Said et al. (1984) reported that low sodium diets can be effective in recycling hens for a second period of egg production. However, the low-salt diets decreased egg production and eggshell thickness in post-molt hens (Ross and Herrick, 1981). Different strains showed different performance after molt, compared to conventional feed withdrawal (Said et al., 1984; Bell and Kuney, 2004). In addition, there is little if any research concerning the effect of low-salt diets on post-molt egg composition and egg solids, which are important factors influencing profits of breaker egg markets. Therefore, it is necessary to have more knowledge in effect of no salt diet on long-term performance, egg quality, egg component, and egg solids in current strains of commercial Leghorns (Bovans and Dekalb) to determine acceptable alternatives for feed withdrawal method. Three experiments (section V and VI) were conducted to determine the effect of molting method on performance, egg components, egg solids, egg quality, and profits in Bovans white and Dekalb white hens in long-term post-molt production period.

II. EFFECT OF DIETARY ENERGY ON PERFORMANCE AND  
EGG COMPOSITION OF BOVANS AND DEKALB  
WHITE HENS DURING PHASE I

**ABSTRACT** A  $4 \times 2$  factorial experiment with four dietary energy levels (2,719, 2,798, 2,877 and 2,959 kcal ME/kg) and two strains (Bovans White and Dekalb White) was conducted to determine the effect of dietary energy on reproductive performance, egg composition, and profits of two strains of commercial leghorns. This experiment lasted 16 weeks. Bovans White hens (n = 768) and Dekalb White hens (n = 768) in Phase I (21 weeks of age) were randomly assigned into 8 treatments (16 replicates of 12 birds per treatment). Bovans White had significantly higher feed intake, egg production, egg mass, body weight, percent egg yolk, and Yolk/Albumen ratio than Dekalb White. Bovans White had significantly lower feed conversion, egg weight, egg specific gravity, percentage of albumen weight, percentage of shell weight, and Haugh unit than Dekalb White. When dietary energy increased from 2,719 to 2,956 kcal ME/kg, hens adjusted feed intake from 107.6 to 101.1 g/hen per day to achieve a constant energy intake so that the same amount of dietary energy (5.8 kcal) was used to produce 1 g of egg. Increasing dietary energy by the addition of poultry oil increased early egg weight, which was mostly due to increased yolk weight. Increasing dietary energy by the addition of poultry oil significantly decreased feed conversion and egg specific gravity, but had no effect on egg production, egg mass, body weight and mortality. Increasing dietary energy by the

addition of poultry oil to a 282 kcal ME/g lysine ratio maximized egg weight during Phase I. The energy/lysine ratio required for optimal profits varies with egg price and feed ingredient prices, which are variable.

*Key words:* strains, dietary energy, egg composition, egg weight, feed intake

## **INTRODUCTION**

A number of studies have been conducted to investigate dietary energy effect on feed intake of commercial Leghorns. Grobas et al. (1999) reported that an increase in dietary energy from 2,680 to 2,810 kcal ME/kg decreased feed intake by 4%. Harms et al. (2000) showed that hens fed diets containing 2,519 kcal ME/kg had 8.5% more feed intake than hens fed diets containing 2,798 kcal ME/kg, and hens fed diets containing 3,078 kcal ME/kg had 3.0% less feed intake than hens fed diets containing 2,798 kcal ME/kg.

Many researchers have reported that increasing dietary energy by the addition of corn oil or poultry oil increased early egg weight (Keshavarz, 1995; Keshavarz and Nakajima, 1995; Harms et al., 2000; Bohnsack et al., 2002; Sohail et al., 2003). However, Summers and Leeson (1983) reported that egg weight was not changed by increasing dietary energy or supplementing with fat.

There is a wide range of dietary energy levels (2,684–2,992 kcal ME/kg) currently being used by the egg industry. Part of the reason for this is that information is not available that would allow egg producers to know the ideal dietary energy level required for optimal performance and profits during phase I. Feed intake and egg weight can significantly affect cost of production and profits. With the sharp increase in dietary energy prices that can occur and did during the past year, it is even more important for

egg producers to have information available that would allow them to continually optimize dietary energy use. Although processed egg consumption such as liquid egg products and dried egg solids has steadily increased during last 10 years, no studies have been conducted to investigate the dietary energy effect on egg components of current strains of Bovans White and Dekalb White during Phase I.

The objective of this study was to determine the effect of dietary energy on performance, egg composition, and profits in Bovans White and Dekalb White hens during Phase I.

## **MATERIAL AND METHODS**

Four dietary energy levels (2,719, 2,798, 2,877 and 2,959 kcal ME/kg) and two strains (Bovans White and Dekalb White) in a 4 × 2 factorial arrangement were used. This experiment lasted 16 weeks. Ingredients and nutrient composition of experimental diets were shown in Table 1.

In this experiment, Bovans White hens (n = 768) and Dekalb White hens (n = 768) during Phase I (21 weeks of age) were randomly assigned into 8 treatments (16 replicates of 12 birds per treatment). Replicates were equally distributed into upper and lower cages to minimize cage level effect. Three hens were housed in a 40.6 × 45.7 cm<sup>2</sup> cage and five adjoining cages consisted of a replicate. All hens were housed in an environmentally-controlled house with temperature maintained at approximately 25.6°C. The house had controlled ventilation and lighting (16 h/d). All hens were supplied with feed and water *ad libitum*. Egg production was recorded daily, feed consumption and egg weight were recorded weekly, and egg specific gravity was recorded monthly. Egg weight and egg specific gravity were measured using all eggs produced during two

consecutive days. Egg specific gravity was determined using 11 gradient saline solutions varying in specific gravity from 1.060 to 1.100 with 0.005-unit increments (Holder and Bradford, 1979). Mortality was determined daily and the feed consumption was adjusted accordingly. Body weight was obtained by weighing 3 hens per group at the end of the experiment. Egg mass and feed conversion (g feed/g egg) were calculated from egg production, egg weight and feed consumption.

Egg components were measured using all eggs (n = 341) from four replicates of each treatment in the middle of experiment and all eggs (n = 301) from four replicates of each treatment at the end of experiment. Eggs were weighed and then broken. The yolks were separated from the albumen. Before determining the yolk weight, the chalazae was removed by forceps. Each yolk was rolled on a paper towel to remove adhering albumen. The shells were cleaned of any adhering albumen and dried for 5 days. Albumen weight was calculated by subtracting the weight of yolk and shell from the whole egg weight.

Three eggs from each replicate were collected at the end of experiment for measuring total solid. The yolk and albumen were mixed completely and 5-6 gram homogenate was pipetted into aluminum dish with weight recorded to 0.001 gram. The sample was dried in an oven for 24 h at 105 C (AOAC, 1990), and then weighed. Three eggs per replicate were used to analyze yolk and albumen solid. After yolk was separated from albumen, three yolks per replicate were mixed together. The albumens from three eggs per replicate were mixed together. The procedure for analyzing albumen and yolk solid was the same as the procedure for total egg solids content. Yolk color and Haugh unit were measured (3 eggs of each replicate) at the end of experiment by egg multi-tester EMT-5200 (Robotmation, Co., Ltd, Japan).

Data were analyzed by proc mixed procedures of Statistical Analysis System (SAS Institute, 2000) for a randomized complete block with a factorial treatment design. Dietary energy and strain were fixed, while blocks were random. The factorial treatment arrangement consisted of 4 dietary energy levels and 2 layer strains. The following model used to analyze data was as follows:

$$Y_{ijk} = \mu + \alpha_i + \beta_j + (\alpha\beta)_{ij} + P_k + \varepsilon_{ijk}$$

Where  $Y_{ijk}$  = individual observation,  $\mu$  = experimental mean,  $\alpha_i$  = dietary energy effect,  $\beta_j$  = layer strain effect,  $(\alpha\beta)_{ij}$  = interaction between dietary energy and strain,  $P_k$  = effect of block,  $\varepsilon_{ijk}$  = error component.

If differences in treatment means were detected by ANOVA, Duncan's Multiple Range Test was applied to separate means. Contrast statements were utilized to test for linear or quadratic dietary energy effects. A significance level of  $P \leq 0.05$  was used during analysis.

## **RESULTS AND DISCUSSION**

There was no interaction between dietary energy and strain on feed intake (Table 2). Dietary energy had a significant effect on feed intake. With increasing dietary energy levels feed intake linearly decreased from 107.6 to 101.1 g/hen/day, resulting in a net decrease of 6.5 g/hen/day or 6.0% of feed intake. An increase of 39 kcal ME/kg decreased feed intake by 1%. Calculations from the results of Grobas (1999) indicated that an increase of 33 kcal ME/kg decreased feed intake by 1%, which was very similar to our result. Strain significantly affected feed intake. Dekalb White hens had significantly lower feed intake than Bovans White hens. The decrease in feed intake in

Dekalb White hens was similar to that in Bovans White hens.

Strain had a significant effect on egg production and egg mass (Table 2). Bovans White hens had significantly higher egg production and egg mass than Dekalb White hens at all four energy levels. There was no significant dietary energy effect on egg production and/or egg mass. This result was consistent with that of Harms et al. (2000) who reported that egg production was not affected by dietary energy. There was no interaction between strain and dietary energy on egg production and/or egg mass.

There was no interaction between strain and dietary energy on egg weight (Table 3). Strain had a significant effect on egg weight. Dekalb White hens had significantly higher egg weight than Bovans White hens from 25 week of age. Dietary energy had a significant linear effect on egg weight during week 25 to 26, week 31 to 32, week 35 to 36, and overall average. This result was in agreement with that of Harms et al. (2000), Bohnsack et al. (2002), and Sohail et al. (2003), who observed that increasing dietary energy by the addition of corn oil or poultry oil had positive effects on early egg weight. With dietary energy levels increasing from 2,719 to 2,877 kcal ME/kg egg weight linearly increased from 60.85 to 61.47 g. However, a further increase in dietary energy levels from 2,877 to 2,956 kcal ME/kg had no additional effect on egg weight (Table 3). When dietary energy increased from 2,719 to 2,956 kcal ME/kg, hens adjusted feed intake from 107.6 to 101.1 g/hen per day to achieve a constant energy intake so that the same amount of dietary energy (5.8 kcal) was used to produce 1 g of egg (Table 4). Because nutrient contents, except dietary energy level in all four diets, were the same, nutrient intake such as protein, TSAA, and lysine linearly decreased with increasing dietary energy (Table 4). When methionine (TSAA) or lysine of the diets decreased, egg

weight decreased (Roland et al., 1992; Novak et al., 2004). The decrease of nutrient intake might explain why increasing dietary energy levels from 2,877 to 2,956 kcal ME/kg had no effect on egg weight and supports the hypothesis that there probably is an ideal energy/protein (lysine) ratio for optimal performance. Similarly, Bartov (1989) and Bartov and Plavnik (1998) reported that energy to protein ratio significantly affected feed intake, weight gain, breast meat yield, and feed efficiency of broiler chicks and concluded that there was an optimal energy to protein ratio for broilers, which may be below than NRC recommendation.

There are conflicting results on effect of linoleic acid on egg weight. NRC (1994) recommended that linoleic acid requirement for laying hens was 1.0%. Grobas et al. (1999) reported that linoleic acid (more than 1.15%) had no effect on egg weight. However, Scragg et al. (1987) reported that linoleic acid affected egg weight when linoleic acid content was more than 1%. In this trial, as dietary energy increased from 2,719 to 2,956 kcal ME/kg by the addition of poultry oil, linoleic acid increased from 1.21 to 2.28%. The increase of egg weight might be attributed to linoleic acid or poultry oil independent of linoleic acid content. Based on the results of this trial, it is difficult to distinguish the effects of linoleic acid and poultry oil independent of linoleic acid content on egg weight. More studies need to be conducted to determine the effect of linoleic acid on egg weight.

There were no interactions between dietary energy and strain on feed conversion, egg specific gravity, body weight, and mortality (Table 2). Feed conversion of Bovans White hens was significantly lower than that of Dekalb White hens. Dietary energy had a significant linear effect on feed conversion. As dietary energy increased from 2,719 to

2,956 kcal ME/kg, feed conversion linearly decreased from 2.14 to 1.97 (g feed/g egg), resulting in a net decrease of 7.9%. Egg specific gravity of Dekalb White hens was significantly higher than that of Bovans White at all four energy levels. Egg specific gravity in hens fed diets containing 2,719 and 2,798 kcal ME/kg dietary energy was significantly higher than that in hens fed diets with 2,877 and 2,956 kcal ME/kg dietary energy. This reduction of egg specific gravity can be attributed to increased egg weight and/or reduced calcium intake caused by increased dietary energy. These results are consistent with those of Sohail et al. (2003), who reported that the decrease of egg specific gravity was due to increased egg size caused by added fat. Bovans White hens had significantly higher body weight than Dekalb White hens. There was no dietary energy effect on hen body weight. Strain and dietary energy had no effect on mortality.

Dekalb White hens had significantly higher albumen weight, shell weight, percentage of albumen, and percentage of shell than Bovans White hens (Table 5). Bovans White hens had significantly higher percentage of yolk and Yolk/Albumen ratio than Dekalb White hens. These results suggest that eggs from some strains may be more desirable for further processing. Dietary energy had a significant linear effect on yolk weight (Table 5). As dietary energy increased from 2,719 to 2,956 kcal ME/kg, egg yolk increased from 15.95 to 16.39 g. These results suggest that the increase of egg weight was mainly due to increased yolk weight. Shell et al. (1987) hypothesized that because hepatic synthesis of lipoprotein by hens during early egg production is insufficient, exogenous fat might supply more lipids for egg yolk development. Haugh unit is a measure of egg quality (height of albumen/egg weight). Dekalb White hens had significantly higher Haugh unit than Bovans White hens (Table 6) possibly because of

higher egg weight and lower Yolk/Albumen ratio in Dekalb White hens. Haugh unit linearly decreased with increasing dietary energy. The reduction of Haugh unit might be due to increased egg weight and the effect of reduced amino acid intake on albumen. As dietary lysine intake decreased, percentage of albumen decreased (Novak et al., 2004).

Economic Feeding and Management Program developed by Roland et al. (1998, 2000) was used to calculate profits of different dietary energy levels at different egg and poultry oil prices. When poultry oil price was 22 cents/kg, maximum profits per dozen eggs were obtained in hens fed the diet containing 2,956 kcal ME/kg dietary energy at all three different egg prices (Table 7). However, when poultry oil price increased to 40 cents/kg, maximum profits were obtained in hens fed the diet containing 2,798 kcal ME/kg at all three egg prices. Because feed ingredient prices and egg price vary, there can be no fixed ideal energy/protein (lysine) ratio for optimal profits.

In conclusion, Bovans White had significantly higher feed intake, egg production, body weight, percent of egg yolk, and Yolk/Albumen ratio than Dekalb White. Bovans White had significantly lower egg weight, feed conversion, egg specific gravity, percentage of albumen weight, percentage of shell weight, and Haugh unit than Dekalb White. When dietary energy increased from 2,719 to 2,956 kcal ME/kg, hens adjusted feed intake from 107.6 to 101.1 g/hen per day to achieve a constant energy intake so that the same amount of dietary energy (5.8 kcal) was used to produce 1 g of egg. Increasing dietary energy by the addition of poultry oil increased early egg weight, which was mostly due to increased yolk weight. Increasing dietary energy by the addition of poultry oil significantly decreased feed conversion and egg specific gravity, but had no effect on egg production, egg mass, body weight and mortality. Increasing dietary energy by the

addition of poultry oil to a 282 kcal ME/g lysine ratio maximized egg weight during Phase I. The energy/lysine ratio required for optimal profits varies with egg price and feed ingredient price, which are variable.

#### **ACKNOWLEDGMENTS**

The authors thank Centurion Poultry, Inc., Lexington, GA, and Ridley Inc., Mankato, MN, for funding support of this research.

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**Table.** Ingredients and nutrient composition of experimental diets

Ingredient (%)	Diet 1	Diet 2	Diet 3	Diet 4
Corn	59.03	57.21	55.36	53.52
Soybean meal	29.28	29.43	29.57	29.72
CaCO <sub>3</sub>	6.98	6.98	6.97	6.97
Hardshell <sup>1</sup>	2.00	2.00	2.00	2.00
Dicalcium phosphate	1.68	1.69	1.70	1.71
Poultry oil	0.00	1.68	3.36	5.05
NaCl	0.38	0.39	0.39	0.39
Vitamin Premix <sup>2</sup>	0.25	0.25	0.25	0.25
Mineral premix <sup>3</sup>	0.25	0.25	0.25	0.25
DL-Methionine	0.14	0.14	0.15	0.15
Calculated analysis				
Crude Protein (%)	18.85	18.78	18.71	18.63
ME (kcal/kg)	2719	2798	2877	2956
Linoleic acid (%)	1.21	1.57	1.92	2.28
Calcium (%)	4.00	4.00	4.00	4.00
Available phosphorus (%)	0.42	0.42	0.42	0.42
Methionine (%)	0.44	0.44	0.44	0.44
Metionine+Cystine (%)	0.76	0.76	0.76	0.76
Lysine (%)	1.02	1.02	1.02	1.02

<sup>1</sup>Hardshell = large particle (passing US mesh #4 and retained by US mesh #6) CaCO<sub>3</sub> supplied by Franklin Industrial Minerals, Lowell, Florida.

<sup>2</sup>Provided per kilogram of diet: vitamin A (as retinyl acetate), 8,000 IU; cholecalciferol, 2,200 ICU; vitamin E (as DL- $\alpha$ -tocopheryl acetate), 8 IU; vitamin B<sub>12</sub>, 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<sub>1</sub> (thiamin mononitrate), 1 mg; pyridoxine, 2.2 mg; biotin, 0.05 mg; vitamin K (menadione sodium bisulfate complex), 2 mg.

<sup>3</sup>Provided per kilogram of diet: manganese, 65 mg; iodine, 1 mg; iron, 55 mg; copper, 6 mg; zinc, 55 mg; selenium, 0.3 mg.

**Table 2.** Influence of dietary energy on performance of Bovans White and Dekalb White (21 to 36 wk of age)

Factor		Feed intake (g/hen/day)	Egg production (%)	Egg mass (g egg/h/d)	Feed conversion (g feed/g egg)	Egg specific gravity (unit)	Body weight (kg)	Mortality (%)
Strain	Bovans	105.6 <sup>a</sup>	85.84 <sup>a</sup>	52.18 <sup>a</sup>	2.02 <sup>b</sup>	1.0887 <sup>b</sup>	1.62 <sup>a</sup>	0.076
	Dekalb	103.0 <sup>b</sup>	80.22 <sup>b</sup>	49.38 <sup>b</sup>	2.09 <sup>a</sup>	1.0909 <sup>a</sup>	1.58 <sup>b</sup>	0.086
Dietary energy (Kcal ME/kg)	2719	107.6 <sup>a</sup>	82.53	50.22	2.14 <sup>a</sup>	1.0903 <sup>a</sup>	1.58	0.086
	2798	105.4 <sup>b</sup>	83.25	50.75	2.08 <sup>b</sup>	1.0902 <sup>a</sup>	1.60	0.086
	2877	103.2 <sup>c</sup>	82.79	50.87	2.03 <sup>c</sup>	1.0894 <sup>b</sup>	1.60	0.086
	2956	101.1 <sup>d</sup>	83.54	51.29	1.97 <sup>d</sup>	1.0894 <sup>b</sup>	1.62	0.065
Pooled SEM		0.97	2.76	0.80	0.02	0.0004	0.06	0.0027
-----Probability-----								
Main effects and interactions								
Strain		0.0005	0.0001	0.0001	0.0001	0.0001	0.0117	0.8325
Dietary energy		0.0001	0.7396	0.6137	0.0001	0.0007	0.3087	0.8365
Strain × Energy		0.6614	0.6805	0.9390	0.8345	0.8392	0.9536	0.9199
Contrasts								
Energy linear		0.0001	0.4197	0.1939	0.0001	0.0002	0.0521	0.9247
Energy quadratic		0.9591	0.9893	0.9230	0.4348	0.9686	0.9447	0.5084

<sup>a-d</sup> Means within a column and under each main effect with no common superscripts differ significantly

**Table 3.** Influence of dietary energy on egg weight in Bovans White and Dekalb White

Factor		Egg weight (g)								
		21-22 wk	23-24 wk	25-26 wk	27-28 wk	29-30 wk	31-32 wk	33-34 wk	35-36 wk	Overall l
Strain	Bovans	51.76	56.82	58.95 <sup>b</sup>	60.59 <sup>b</sup>	62.44 <sup>b</sup>	63.26 <sup>b</sup>	63.14 <sup>b</sup>	62.85 <sup>b</sup>	60.80 <sup>b</sup>
	Dekalb	51.96	56.59	59.34 <sup>a</sup>	61.31 <sup>a</sup>	63.13 <sup>a</sup>	64.24 <sup>a</sup>	64.30 <sup>a</sup>	64.30 <sup>a</sup>	61.55 <sup>a</sup>
Dietary energy (Kcal ME/kg)	2719	51.72	56.65	58.95 <sup>ab</sup>	60.78	62.52	63.26 <sup>c</sup>	63.29	62.97 <sup>b</sup>	60.85 <sup>c</sup>
	2798	51.86	56.39	58.72 <sup>b</sup>	60.68	62.50	63.50 <sup>b</sup> <sub>c</sub>	63.76	63.56 <sup>a</sup> <sub>b</sub>	60.98 <sup>b</sup> <sub>c</sub>
	2877	51.72	56.94	59.47 <sup>a</sup>	61.26	63.15	64.22 <sup>a</sup>	63.89	63.81 <sup>a</sup>	61.47 <sup>a</sup>
	2956	52.01	56.82	59.43 <sup>a</sup>	61.09	62.96	64.03 <sup>a</sup> <sub>b</sub>	63.94	63.95 <sup>a</sup>	61.40 <sup>a</sup> <sub>b</sub>
Pooled SEM		0.71	0.45	0.38	0.36	0.42	0.41	0.47	0.46	0.35
-----Probability-----										
Main effects and interactions										
Strain		0.6787	0.3077	0.0389	0.0001	0.0016	0.0001	0.0001	0.0001	0.0001
Dietary energy		0.9310	0.3420	0.0117	0.0838	0.0770	0.0040	0.1869	0.0187	0.0286
Strain × Energy		0.9834	0.3092	0.3150	0.3046	0.7553	0.5541	0.6369	0.1856	0.6570
Contrasts										
Energy linear		0.6469	0.2966	0.0105	0.0585	0.0397	0.0015	0.0486	0.0027	0.0067
Energy quadratic		0.8351	0.7609	0.5901	0.8626	0.6974	0.2974	0.3604	0.3331	0.5762

<sup>a-c</sup> Means within a column and under each main effect with no common superscripts differ significantly

**Table 4.** Influence of dietary energy on nutrient intake for one gram egg of Bovans White and Dekalb White

Factor		Energy/Lysine ratio (kcal/g)	Nutrients used to produce 1 g egg			
			Dietary energy (Kcal)	Protein (g)	TSAA (mg)	Lysine (mg)
Strain	Bovans	278	5.7 <sup>b</sup>	0.38 <sup>b</sup>	15.39 <sup>b</sup>	20.65 <sup>b</sup>
	Dekalb	278	5.9 <sup>a</sup>	0.39 <sup>a</sup>	15.88 <sup>a</sup>	21.31 <sup>a</sup>
Dietary energy (Kcal ME/kg)	2719	267	5.8	0.40 <sup>a</sup>	16.29 <sup>a</sup>	21.87 <sup>a</sup>
	2798	274	5.8	0.39 <sup>b</sup>	15.80 <sup>b</sup>	21.20 <sup>b</sup>
	2877	282	5.8	0.38 <sup>c</sup>	15.44 <sup>c</sup>	20.72 <sup>c</sup>
	2956	290	5.8	0.37 <sup>d</sup>	15.00 <sup>d</sup>	20.12 <sup>d</sup>
Pooled SEM			0.054	0.004	0.158	0.212
-----Probability-----						
Main effects and interactions						
Strain			0.0001	0.0001	0.0001	0.0001
Dietary energy			0.9748	0.0001	0.0001	0.0001
Strain × Energy			0.6101	0.05619	0.5603	0.5614
Contrasts						
Energy linear			0.8347	0.0001	0.0001	0.0001
Energy quadratic			0.9743	0.9147	0.8110	0.8117

<sup>a-c</sup> Means within a column and under each main effect with no common superscripts differ significantly

**Table 5.** Influence of dietary energy on egg components in Bovans White and Dekalb White

Factor		Egg component weight (g)			% of egg components			Yolk:Albumen ratio
		Yolk	Albumen	Shell	Yolk	Albumen	Shell	
Strain	Bovans	16.31	40.64 <sup>b</sup>	5.76 <sup>b</sup>	26.05 <sup>a</sup>	64.76 <sup>b</sup>	9.20 <sup>b</sup>	0.404 <sup>a</sup>
	Dekalb	16.11	42.29 <sup>a</sup>	6.13 <sup>a</sup>	25.00 <sup>b</sup>	65.48 <sup>a</sup>	9.52 <sup>a</sup>	0.383 <sup>b</sup>
Dietary energy (Kcal ME/kg)	2719	15.95 <sup>b</sup>	41.58	5.95	25.15	65.46	9.39	0.386
	2798	16.09 <sup>ab</sup>	41.07	5.96	25.54	65.02	9.44	0.394
	2877	16.41 <sup>a</sup>	41.34	5.94	25.82	64.85	9.33	0.400
	2956	16.39 <sup>a</sup>	41.78	5.94	25.59	65.14	9.27	0.394
Pooled SEM		0.25	0.69	0.11	0.37	0.41	0.13	0.008
-----Probability-----								
Main effects and interactions								
Strain		0.1362	0.0003	0.0001	0.0001	0.0026	0.0001	0.0001
Dietary energy		0.0524	0.5173	0.9973	0.1218	0.2236	0.2923	0.1562
Strain × Energy		0.4279	0.1840	0.2364	0.1647	0.3645	0.5364	0.1929
Contrasts								
Energy linear		0.0110	0.5827	0.8847	0.0715	0.2309	0.1169	0.1127
Energy quadratic		0.5330	0.1882	0.9304	0.1142	0.0893	0.3824	0.1042

<sup>a-b</sup> Means within a column and under each main effect with no common superscripts differ significantly

**Table 6.** Influence of dietary energy on egg solids, haugh units, and yolk color in Bovans White and Dekalb White

Factor		% of solid			Haugh unit	Yolk color
		Whole egg	Albumen	Yolk		
Strain	Bovans	24.60	12.61	49.48	80.98 <sup>b</sup>	6.85
	Dekalb	23.53	12.57	49.20	83.49 <sup>a</sup>	7.02
Dietary energy (Kcal ME/kg)	2719	23.58	12.68	49.06	84.36 <sup>a</sup>	6.73
	2798	23.87	12.64	49.67	82.73 <sup>ab</sup>	7.23
	2877	23.95	12.54	49.52	80.37 <sup>c</sup>	6.83
	2956	24.87	12.49	49.13	81.46 <sup>bc</sup>	6.96
Pooled SEM		0.19	0.24	0.11	1.96	0.34
-----Probability-----						
Main effects and interactions						
Strain		0.0609	0.6636	0.4800	0.0020	0.2339
Dietary energy		0.4059	0.4131	0.6474	0.0040	0.0703
Strain × Energy		0.5086	0.0896	0.0176	0.7106	0.5540
Contrasts						
Energy linear		0.1213	0.0980	0.9667	0.0023	0.6407
Energy quadratic		0.5745	0.9100	0.2131	0.0900	0.1808

<sup>a-c</sup> Means within a column and under each main effect with no common superscripts differ significantly

**Table 7.** Influence of dietary energy and poultry oil price on profits<sup>1,2</sup>

	Dietary energy content (kcal ME/kg)			
	2719	2798	2877	2956
	Returns <sup>3</sup> (cents/dozen)			
High fat price (\$0.40/kg)				
High egg price	0.571	0.576	0.573	0.574
Moderate egg price	0.109	0.114	0.112	0.113
Low egg price	-0.143	-0.137	-0.138	-0.138
Low fat price (22 cent/kg)				
High egg price	0.571	0.578	0.581	0.586
Moderate egg price	0.110	0.116	0.120	0.125
Low egg price	-0.142	-0.135	-0.131	-0.125

<sup>1</sup> Corn price = \$0.12/kg, soy price = \$0.20/kg, CaCO<sub>3</sub> = \$0.03/kg, hard shell = \$0.03/kg, Dicalcium phosphate = \$0.27/kg, salt = \$0.06/kg, Vitamin premix = \$2.67/kg, mineral premix = \$0.59 cents/kg, DL-methionine = \$2.59 cents/kg.

<sup>2</sup> The egg price spread between medium and large eggs was 11 cents

<sup>3</sup>Returns (R) were calculated using the equation:  $R = \text{UBEP} - \text{NR} - \text{PC} - \text{FdC}$ , where UBEP = Urner Barry Egg Price, NR = nest run into package product delivered, PC = production cost, and FdC = feed cost, as described by Roland et al. (1998, 2000).

III. INFLUENCE OF DIETARY ENERGY AND ANTIBIOTIC  
ON PERFORMANCE, EGG COMPOSITION, EGG QUALITY  
IN BOVANS WHITE AND DEKALB WHITE HENS DURING PHASE 2

**ABSTRACT** A  $4 \times 2 \times 2$  factorial experiment of four dietary energy levels (2776, 2820, 2864, and 2908 kcal/kg) and two strains (Bovans White and Dekalb White) with and without Tylosin was conducted to determine the influence of dietary energy and antibiotic on performance, egg composition, and egg quality of two strains of commercial Leghorns during Phase II (from 40 to 51 week of age). This experiment lasted 12 weeks. Bovans White hens (n = 768) and Dekalb White hens (n = 768) at 40 week of age were randomly divided into 16 treatments (8 replicates of 12 birds per treatment). With increasing dietary energy hens linearly adjusted feed intake from 105.0 to 101.7 g/hen/day to achieve a constant energy intake so that the same amount of dietary energy (5.2 kcal) was used to produce 1 g of egg. With dietary energy levels increasing from 2,776 to 2,864 kcal ME/kg feed conversion linearly decreased from 1.89 to 1.79 g of feed/ g of egg. Based on feed conversion, increasing dietary energy to 2,864 kcal ME/kg by the addition of poultry oil might be sufficient for optimal performance of laying hens during Phase 2 (wk 40 to 51). There can be no fixed ideal dietary energy level for optimal profits during phase 2 (wk 40 to 51), due to varying feed ingredient prices and egg price. Increasing dietary energy had significant effects on percent of yolk, percent of albumen, Yolk/Albumen

ratio, and dirty eggs. Bovans White had significantly higher egg production, percent of egg yolk, Yolk/Albumen ratio, whole egg solids, and crack eggs than Dekalb White. Bovans white hens had significantly lower egg weight, feed conversion, egg specific gravity, percentage of shell, Haugh unit, shell thickness, and yolk color than Dekalb white hens. While Tylosin had no effect on feed intake, egg production, egg weight, feed conversion, and mortality, Tylosin supplementation significantly reduced body checks and increased yolk color, resulting positive effects on egg quality of laying hens.

*Key words:* strains, energy, Tylosin, egg composition, feed intake

## INTRODUCTION

Antibiotics are substances that inhibit the growth of bacteria and related microorganism by interfering with essential metabolic functions. Antibiotic supplementation in feed increases weight gain and improves feed efficiency of broilers (Miller Publishing Co., 2000, Miles et al., 2006). However, antibiotics had no effect on body weight gain and feed utilization (Izat et al., 1990). Tylosin, an antibiotic, made naturally by the bacterium “*Streptomyces fradiae*” and acts to inhibit bacterial protein synthesis by inhibiting the 50S ribosome, a cellular structure of certain bacteria. Although Tylosin is widely used by poultry industry, there is very limited scientific literature about the effect of Tylosin on performance, egg component, and egg quality of laying hens.

There is a wide range of dietary energy levels (2,684–2,992 kcal ME/kg) currently being used by the egg industry. Regulating dietary energy is believed to be one of the most effective ways to adjust feed intake of laying hens. Many researchers have reported that increasing dietary energy decreased feed intake (Grobas et al., 1999; Harms et al., 2000, Wu et al., 2005a) and improved feed conversion of laying hens (Wu et al., 2005a

and b). Increasing dietary energy by the addition of corn oil or poultry oil increased early egg weight (Keshavarz, 1995; Keshavarz and Nakajima, 1995; Harms et al., 2000; Bohnsack et al., 2002; Sohail et al., 2003). However, there are inconsistent results in effect of dietary energy on egg weight. Summers and Leeson (1983) reported that increasing dietary energy or supplementing with fat had no effect on egg weight. There is limited scientific information available that would allow egg producers to know the dietary energy level required for optimal performance during phase 2.

Although processed egg consumption such as liquid egg products and dried egg solids has steadily increased during last 10 years, no studies have been conducted to investigate the dietary energy effect on egg components of current strains of Bovans White and Dekalb White during Phase 2.

The objective of this experiment was to determine the effect of dietary energy and antibiotic on performance, egg composition, and egg quality in Bovans White and Dekalb White in Phase II (from 40 to 51 week of age).

## **MATERIAL AND METHODS**

This study was a  $4 \times 2 \times 2$  factorial arrangement of four dietary energy levels (2776, 2820, 2864, and 2908 kcal/kg) and two strains (Bovans White and Dekalb White) with and without Tylosin. Ingredients and nutrient composition of experimental diets were showed in Table 1. Feed samples were sent to Elanco Animal Health, Memphis, TN, USA, for Tylosin activity analysis. The average chemical analyzed value of Tylosin in the diets was 33.7 g/ton of feed, which was very close to expected value of 33 g/ton of feed.

In this experiment, Bovans White hens (n = 768) and Dekalb White hens (n =

768) at 40 week of age were randomly assigned into 16 treatments (8 replicates of 12 birds per treatment). The trial lasted 12 weeks. Replicates were equally distributed into upper and lower cages to minimize cage level effect. Three hens were housed in a 40.6 × 45.7 cm<sup>2</sup> cage and five adjoining cages consisted of a replicate. All hens were housed in an environmentally-controlled house with temperature maintained at approximately 25.6°C. The house had controlled ventilation and lighting (16 h/d). All hens were supplied with feed and water *ad libitum*. Egg production was recorded daily, feed consumption and egg weight were recorded weekly, and egg specific gravity was recorded monthly. Egg weight and egg specific gravity were measured using all eggs produced during two consecutive days. Egg specific gravity was determined using 11 gradient saline solutions varying in specific gravity from 1.060 to 1.100 with 0.005-unit increments (Holder and Bradford, 1979). Mortality was determined daily and the feed consumption was adjusted accordingly. Body weight was obtained by weighing 3 hens per group at the end of the experiment. Egg mass and feed conversion (g feed/g egg) were calculated from egg production, egg weight and feed consumption.

Dirty eggs, crack eggs, and body checkss were determined by using all eggs produced during two consecutive days at the middle and end of the experiment. Dirty eggs were scored as following: 0 = clean egg; 1 = dirty area (adhering manure or blood) less than 25% of egg (light dirty egg); 2 = dirty area (adhering manure or blood) more than 25% of egg (heavy dirty egg). Dirty egg score per replicate = [(1 × number of light dirty egg) + (2 × number of heavy dirty egg)]/total number of egg per replicate. A body check was determined by observing an egg having a ridged area in the shell under a light. A crack egg was determined by observing an egg having a broken or cracked shell under

a light.

Three eggs from each replicate were collected at the end of experiment for measuring egg component. Three eggs from each replicate were collected to measure whole egg solids in the middle and the end of experiment. Albumen and yolk solid were measured by using three eggs from each replicate in the middle and the end of experiment. The procedures for measuring egg components, whole egg solids, and albumen and yolk solid were the same as those of Wu et al. (2005). Yolk color and Haugh unit were measured (3 eggs of each replicate) at the end of experiment by egg multi-tester EMT-5200 (Robotmation, Co., Ltd, Japan).

Data were analyzed by proc mixed procedures of Statistical Analysis System (SAS Institute, 2000) for a randomized complete block with a factorial treatment design. The factorial treatment arrangement consisted of 4 dietary energy levels, 2 layer strains, and 2 Tylosin levels. Dietary energy, strain, and Tylosin were fixed, while blocks were random. The following model used to analyze data was as follows:

$$Y_{ijkl} = \mu + \alpha_i + \beta_j + \gamma_k + (\alpha\beta)_{ij} + (\alpha\gamma)_{ik} + (\beta\gamma)_{jk} + (\alpha\beta\gamma)_{ijk} + P_l + \varepsilon_{ijkl}$$

Where  $Y_{ijkl}$  = individual observation,  $\mu$  = experimental mean,  $\alpha_i$  = dietary energy effect,  $\beta_j$  = layer strain effect,  $\gamma_k$  = Tylosin effect,  $(\alpha\beta)_{ij}$  = interaction between dietary energy and strain,  $(\alpha\gamma)_{ik}$  = interaction between dietary energy and Tylosin,  $(\beta\gamma)_{jk}$  = interaction between Tylosin and strain,  $(\alpha\beta\gamma)_{ijk}$  = interactions among dietary energy, strain, and Tylosin,  $P_l$  = effect of block,  $\varepsilon_{ijkl}$  = error component.

If differences in treatment means were detected by ANOVA, Duncan's Multiple Range Test was applied to separate means. Contrast statements were utilized to test for

linear or quadratic dietary energy effects. A significance level of  $P \leq 0.05$  was used during analysis.

## **RESULTS AND DISCUSSION**

Strain had no significant effect on feed intake (Table 2). Dietary energy had a significant effect on feed intake. When dietary energy level increased from 2776 to 2908 kcal/kg, feed intake linearly decreased from 105.0 to 101.7 g/hen/day, resulting in a net decrease of 3.2% of feed intake. An increase of 36 kcal ME/kg dietary energy decreased feed intake by 1%, which was very similar to the results of Grobas (1999), who reported that an increase of 33 kcal ME/kg dietary energy decreased feed intake by 1%. When dietary energy increased from 2776 to 2908 kcal ME/kg, hens adjusted feed intake from 105.0 to 101.7 g/hen/day so that the same amount of dietary energy (5.2 kcal) was used to produce 1 g egg. There was no significant difference in feed intake between hens fed the diets without Tylosin and hens fed the diets supplemented with Tylosin.

Bovans White hens had significantly higher egg production than Dekalb White hens (Table 2). There was no significant dietary energy effect on egg production. This result was consistent with that of Harms et al. (2000) who reported that egg production was not affected by dietary energy. Hens fed the diets supplemented with Tylosin had constant numerically higher egg production than hens fed the diets without Tylosin from wk 1 to wk 12 ( $P < 0.09$ ).

Dekalb White hens had significantly higher egg weight than Bovans White hens (Table 2). There was no significant dietary energy effect on egg weight. While percentage yolk linearly increased with increasing dietary energy level, percentage albumen linearly decreased, which might be due to decreased nutrient intake caused by

decreased feed intake (Table 3). The increased yolk weight and decreased albumen weight might explain no response of egg weight to increasing dietary energy. Tylosin supplementation had no effect on egg weight. Egg mass was not affected by strain, dietary energy, or Tylosin supplementation.

Strain had no effect on feed conversion (Table 2). Dietary energy had a significant linear effect on feed conversion. With dietary energy levels increasing from 2,776 to 2,864 kcal ME/kg feed conversion significantly decreased from 1.89 to 1.81 g of feed/g of egg. However, a further increase in dietary energy levels from 2,864 to 2,908 kcal ME/kg had no additional effect on feed conversion. Therefore, increasing dietary energy to 2,864 kcal ME/kg by the addition of poultry oil might be sufficient for optimal performance of laying hens during phase 2 (wk 40 to 51). Similarly, Wu et al. (2005a) reported that increasing dietary energy to 2877 kcal ME/kg might obtain the optimal performance of laying hens during phase 1 (wk 21 to 36). Tylosin supplementation had no significant effect on feed conversion. Similarly, Izat et al. (1990) reported that antibiotics had no effect on feed conversion in broilers.

Strain had a significant effect on egg specific gravity (Table 2). Egg specific gravity of Dekalb White hens was significantly higher than that of Bovans White (Table 2) because percentage of shell and shell thickness of Dekalb White hens were significantly higher than those of Bovans White hens (Table 3 and 4). There were no significant effects of dietary energy on egg specific gravity, body weight, and mortality. Tylosin supplementation had no significant effect on egg specific gravity, body weight, or mortality.

Both dietary energy and Tylosin supplementation had no significant effects on

whole egg solids, albumen solid and yolk solid (Table 3). Dekalb White hens had significantly lower whole egg solids than Bovans White hens. The higher whole egg solids of Bovans White hens was due to higher Yolk/Albumen ratio of Dekalb White hens. These results suggest that eggs of some strains may be more desirable for further processing. Dietary energy had a linear effect on percent yolk, percent albumen, and Yolk/Albumen ratio. As energy content increased from 2776 to 2908 kcal/kg, percentage egg yolk and Yolk/Albumen ratio increased possibly because hens can use available exogenous fat as lipid for egg yolk formation. Scheideler and Froning (1996) and Scheideler et al. (1998) reported that hens can deposit dietary lipid into egg yolk and change the composition of yolk lipid. Tylosin had no effect on percent yolk, percent albumen, percent shell or Yolk/Albumen ratio.

Dekalb White hens had significantly lower crack egg than Bovans White hens possibly because shell thickness of Dekalb White hens was significantly higher than that of Bovans White hens (Table 4). Compared to Bovans White hens, Dekalb White hens had significantly higher body checks, which might be due to higher egg size of Dekalb White hens. Haugh unit of Dekalb White hens was significantly higher than that of Bovans White hens possibly because of higher egg weight and lower Yolk/Albumen ratio of Dekalb White hens. Dekalb White hens had significantly higher yolk color than Bovans White hens. Hens fed the diets containing 2776 kcal/kg produced significantly more dirty eggs than hens fed the diets containing other three dietary energy levels. Body checks of hens fed the diets supplemented with Tylosin was significant lower than that of hens fed the diets without Tylosin.

Hens fed the diets supplemented with Tylosin had significantly higher yolk color than hens fed the diets without Tylosin. The improvement of yolk color of hens fed the diets supplemented with antibiotic may be due to better absorption of pigments caused by change of gut microflora and physical change of gastrointestinal tract. Antibiotics can control and limit the growth of some species of pathogenic bacteria (Feller et al., 1984). The decreased gut microflora decrease nutrient absorption by decreasing gastrointestinal tract thickness and decreasing nutrient competition of microflora with host (Ravindran et al., 1984; Apajalahti et al., 2004). In addition, physical changes, including thinner gastrointestinal tract and increased number of villi per unit length, in hens given antibiotics resulted in improved performance. Body checks of hens fed the diets supplemented with Tylosin was significant lower than that of hens fed the diets without Tylosin. A body check was an egg whose shell cracked inside the hen's body and repaired by additional calcium deposited over the crack, resulting in a ridged area. Antibiotic supplementation might make hens calmer, resulting in less hen activity. Roland (1984) suggested that hen activity during the critical period of shell calcification could cause body checks.

The Economic Feeding and Management Program developed by Roland et al. (1998, 2000) was used to calculate profits of different dietary energy levels at poultry oil prices. When poultry oil price were \$0.20/kg or 0.30/kg, maximum profits per dozen eggs were obtained in hens fed the diet containing 2,864 kcal ME/kg of dietary energy (Table 5). However, when poultry oil price increased to \$0.40/kg, maximum profit was obtained in hens fed the diet containing 2,776 kcal ME/kg of dietary energy. Because feed

ingredient prices and egg price often vary, there can be no fixed ideal dietary energy level for optimal profits during phase II (wk 40 to 51).

In conclusion, increasing dietary energy by the addition of poultry oil linearly decreased feed intake. When dietary energy linearly increased from 2776 to 2908 kcal/kg, hens adjusted feed intake from 105.0 to 101.7 g/hen/day so that the same amount of dietary energy (5.2 kcal) was used to produce 1 g egg. Based on feed conversion, increasing dietary energy to 2,864 kcal ME/kg might be sufficient for optimal performance of laying hens during Phase 2 (from 40 to 51 week of age). There can be no fixed ideal dietary energy level for optimal profits during phase 2 (wk 40 to 51), due to varying feed ingredient prices and egg price. Increasing dietary energy by the addition of poultry oil had a significant effect on percent yolk, percent albumen, Yolk/Albumen ratio, and dirty eggs. Bovans White had significantly higher egg production, percent yolk, Yolk/Albumen ratio and crack eggs than Dekalb White. Bovans White had significantly lower egg weight, feed conversion, egg specific gravity, percentage of shell, body checks, Haugh unit, shell thickness, and yolk color than Dekalb White. While Tylosin had no effect on feed intake, egg production, egg weight, feed conversion, and mortality, Tylosin supplementation significantly reduced body checks and increased yolk color, resulting in positive effects on egg quality of laying hens.

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**Table 1. Ingredients and nutrient composition of experimental diets**

Ingredient	Diet 1	Diet 2	Diet 3	Diet 4	Diet 5	Diet 6	Diet 7	Diet 8
Corn (%)	64.59	64.59	63.55	63.55	62.53	62.53	61.51	61.51
Soybean meal (%)	23.83	23.83	23.92	23.92	24.00	24.00	24.08	24.08
CaCO <sub>3</sub> (%)	7.12	7.12	7.12	7.12	7.11	7.11	7.11	7.11
Hardshell <sup>1</sup> (%)	2.00	2.00	2.00	2.00	2.00	2.00	2.00	2.00
Dicalcium phosphate (%)	1.50	1.50	1.50	1.50	1.50	1.50	1.51	1.51
Poultry oil (%)	0.00	0.00	0.95	0.95	1.89	1.89	2.82	2.82
NaCl (%)	0.39	0.39	0.39	0.39	0.39	0.39	0.39	0.39
Vitamin Premix <sup>2</sup> (%)	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
Mineral premix <sup>3</sup> (%)	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
DL-Methionine (%)	0.08	0.08	0.08	0.08	0.08	0.08	0.09	0.09
Tylosin <sup>4</sup>	-	+	-	+	-	+	-	+
Calculated analysis								
Crude protein (%)	16.65	16.65	16.61	16.61	16.57	16.57	16.53	16.53
ME (kcal/kg)	2776	2776	2820	2820	2864	2864	2908	2908
Ca (%)	4.00	4.00	4.00	4.00	4.00	4.00	4.00	4.00
Available phosphorus (%)	0.38	0.38	0.38	0.38	0.38	0.38	0.38	0.38
Methionine (%)	0.36	0.36	0.36	0.36	0.36	0.36	0.36	0.36
Methionine+Cystine (%)	0.65	0.65	0.65	0.65	0.65	0.65	0.65	0.65
Lysine (%)	0.87	0.87	0.87	0.87	0.87	0.87	0.87	0.87

<sup>1</sup>Hardshell = large particle (passing US mesh #4 and retained by US mesh #6) CaCO<sub>3</sub> supplied by Franklin Industrial Minerals, Lowell, Florida.

<sup>2</sup>Provided per kilogram of diet: vitamin A (as retinyl acetate), 8,000 IU; cholecalciferol, 2,200 ICU; vitamin E (as DL- $\alpha$ -tocopheryl acetate), 8 IU; vitamin B<sub>12</sub>, 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<sub>1</sub> (thiamin mononitrate), 1 mg; pyridoxine, 2.2 mg; biotin, 0.05 mg; vitamin K (menadione sodium bisulfate complex), 2 mg.

<sup>3</sup>Provided per kilogram of diet: manganese, 65 mg; iodine, 1 mg; iron, 55 mg; copper, 6 mg; zinc, 55 mg; selenium, 0.3 mg.

<sup>4</sup>Elanco Animal Health, Memphis, TN

**Table 2.** Influence of dietary energy and Tylosin on performance of Bovans White and Dekalb White from 40 to 51 wk of age

Factor		Feed intake (g/hen per day)	Energy intake (Kcal ME/g egg)	Egg production (%)	Egg weight (g)	Egg mass (g egg/hen per d)	Feed conversion (g of feed/of g egg)	Egg specific gravity (unit)	Body weight (kg)	Mortality (%)
Strain	Bovans	103.3	5.2	89.98 <sup>a</sup>	62.77 <sup>b</sup>	56.48	1.83	1.0814 <sup>b</sup>	1.62	1.95
	Dekalb	104.3	5.2	88.22 <sup>b</sup>	64.37 <sup>a</sup>	56.79	1.84	1.0835 <sup>a</sup>	1.61	1.59
Dietary energy (Kcal/kg)	2776	105.0 <sup>a</sup>	5.2	88.15	63.11	55.63	1.89 <sup>a</sup>	1.0825	1.59	2.36
	2820	104.8 <sup>a</sup>	5.2	89.26	63.56	56.72	1.85 <sup>b</sup>	1.0828	1.62	1.30
	2864	103.8 <sup>ab</sup>	5.2	89.77	63.93	57.38	1.81 <sup>c</sup>	1.0823	1.62	1.59
	2908	101.7 <sup>b</sup>	5.2	89.22	63.67	56.81	1.79 <sup>c</sup>	1.0823	1.61	1.85
Tylosin	-	103.6	5.2	88.51	63.68	56.35	1.84	1.0824	1.61	1.85
	+	104.0	5.2	89.69	63.46	56.92	1.83	1.0826	1.61	1.69
Pooled SEM		1.54	0.05	1.40	0.54	1.07	0.02	0.0007	0.42	0.98
-----Probability-----										
Main effects and interactions										
Strain		NS	NS	0.0135	0.0001	NS	NS	0.0001	NS	NS
Dietary energy		0.0129	NS	NS	0.0852	NS	0.0001	NS	NS	NS
Tylosin		NS	NS	0.0949	NS	NS	NS	NS	NS	NS
Strain × Energy		NS	NS	NS	NS	NS	NS	NS	NS	NS
Strain × Tylosin		NS	NS	NS	NS	NS	NS	NS	NS	NS
Energy × Tylosin		NS	NS	NS	NS	NS	NS	NS	NS	NS
Str. × Ene. × Tylosin		NS	NS	NS	NS	NS	NS	NS	NS	NS
Contrasts										
Energy linear		0.0022	NS	NS	0.0463	0.0852	0.0001	NS	NS	NS
Energy quadratic		NS	NS	NS	NS	NS	NS	NS	NS	NS

<sup>a-c</sup>Means within a column and under each main effect with no common superscripts differ significantly

**Table 3.** Influence of dietary energy and Tylosin on egg solids and egg component in Bovans White and Dekalb White from 40 to 51 wk of age

Factor		% of solid			% of egg components			Yolk:Albumen ratio
		Whole egg	Albumen	Yolk	Yolk	Albumen	Shell	
Strain	Bovans	24.31 <sup>a</sup>	12.70	48.56	28.05 <sup>a</sup>	62.77	9.19 <sup>b</sup>	0.453 <sup>a</sup>
	Dekalb	24.00 <sup>b</sup>	12.52	48.52	27.42 <sup>b</sup>	63.00	9.59 <sup>a</sup>	0.440 <sup>b</sup>
Dietary energy (Kcal/kg)	2776	24.08	12.71	48.63	27.43 <sup>b</sup>	63.28 <sup>a</sup>	9.29	0.439 <sup>c</sup>
	2820	24.23	12.54	48.53	27.45 <sup>b</sup>	63.18 <sup>ab</sup>	9.38	0.440 <sup>bc</sup>
	2864	24.28	12.66	48.47	28.06 <sup>a</sup>	62.48 <sup>bc</sup>	9.47	0.453 <sup>ba</sup>
	2908	24.04	12.53	48.52	28.00 <sup>a</sup>	62.60 <sup>b</sup>	9.40	0.453 <sup>a</sup>
Tylosin	-	24.08	12.63	48.56	27.79	62.82	9.40	0.447
	+	24.23	12.59	48.52	27.68	62.95	9.37	0.445
Pooled SEM		0.13	0.06	0.03	0.27	0.30	0.08	0.006
ANOVA		-----Probability-----						
Main effects and interactions								
Strain		0.0012	0.0644	NS	0.0020	NS	0.0001	0.0080
Dietary energy		NS	NS	NS	0.0337	0.0195	NS	0.0485
Tylosin		0.0947	NS	NS	NS	NS	NS	NS
Strain × Energy		NS	NS	NS	NS	NS	NS	NS
Strain × Tylosin		NS	NS	NS	NS	NS	NS	NS
Energy × Tylosin		NS	NS	NS	NS	NS	NS	NS
Str. × En. × Tyl		NS	NS	NS	NS	NS	NS	NS
Contrasts								
Energy linear		NS	NS	NS	0.0096	0.0057	NS	0.0105
Energy quadratic		0.0406	NS	NS	NS	NS	NS	NS

<sup>a-c</sup>Means within a column and under each main effect with no common superscripts differ significantly

**Table 4.** Influence of dietary energy and Tylosin on egg quality in Bovans White and Dekalb White from 40 to 51 wk of age

Factor		Dirty egg scores	Crack egg (%)	Body checks (%)	Shell thickness (mm)	Haugh unit	Yolk color
Strain	Bovans	0.27	0.07 <sup>a</sup>	0.05 <sup>b</sup>	0.371 <sup>b</sup>	72.71 <sup>b</sup>	5.95 <sup>b</sup>
	Dekalb	0.31	0.02 <sup>b</sup>	0.10 <sup>a</sup>	0.385 <sup>a</sup>	75.58 <sup>a</sup>	6.15 <sup>a</sup>
Dietary energy (Kcal/kg)	2776	0.37 <sup>a</sup>	0.04	0.12	0.376	73.79	6.06
	2820	0.29 <sup>b</sup>	0.04	0.06	0.376	73.91	5.98
	2864	0.26 <sup>b</sup>	0.04	0.08	0.380	74.31	6.09
	2908	0.25 <sup>b</sup>	0.05	0.09	0.379	74.46	6.07
Tylosin	-	0.29	0.05	0.11 <sup>a</sup>	0.379	73.68	5.97 <sup>b</sup>
	+	0.29	0.04	0.06 <sup>b</sup>	0.377	74.52	6.13 <sup>a</sup>
Pooled SEM		0.06	0.03	0.04	0.014	0.72	0.15
-----Probability-----							
Main effects and interactions							
Strain		NS	0.0019	0.0025	0.0011	0.0018	0.0097
Dietary energy		0.0286	NS	NS	NS	NS	NS
Tylosin		NS	NS	0.0332	NS	NS	0.0324
Strain × Energy		NS	NS	NS	NS	NS	NS
Strain × Tylosin		NS	NS	NS	NS	NS	NS
Energy × Tylosin		NS	NS	NS	NS	NS	NS
Str. × En. × Tyl.		NS	NS	NS	NS	NS	NS
Contrasts							
Energy linear		0.0051	NS	NS	NS	NS	NS
Energy quadratic		NS	NS	0.0720	NS	NS	NS

<sup>a-b</sup>Means within a column and under each main effect with no common superscripts differ significantly

**Table 5.** Influence of dietary energy and poultry oil price on profits<sup>1,2</sup> from 40 to 51 wk of age

	Dietary energy content (kcal ME/kg)			
	2776	2820	2864	2908
		Returns <sup>3</sup> (\$/dozen)		
High fat price (\$0.40/kg)	0.176	0.175	0.175	0.173
Medium fat price (\$0.30/kg)	0.176	0.178	0.180	0.181
Low fat price (\$0.20/kg)	0.176	0.177	0.178	0.177

<sup>1</sup> Corn price = \$0.12/kg, soy price = \$0.20/kg, CaCO<sub>3</sub> = \$0.03/kg, hard shell = \$0.03/kg, Dicalcium phosphate = \$0.27 cents/kg, salt = \$0.06/kg, Vitamin premix = \$2.67/kg, mineral premix = \$0.59/kg, DL-methionine = \$2.59/kg.

<sup>2</sup> The egg price spread between medium and large eggs was 11 cents

<sup>3</sup> Returns (R) were calculated using the equation:  $R = \text{UBEP} - \text{NR} - \text{PC} - \text{FdC}$ , where UBEP = Urner Barry Egg Price, NR = nest run into package product delivered, PC = production cost, and FdC = feed cost, as described by Roland et al. (1998, 2000).

IV. INFLUENCE OF DIETARY ENERGY AND ANTIBIOTICS ON  
PERFORMANCE, EGG SOLIDS, AND EGG QUALITY IN BOVANS  
WHITE AND DEKALB WHITE HENS DURING PHASE 3

**ABSTRACT** A  $4 \times 2 \times 2$  factorial arrangement of four dietary energy levels (2,776, 2,820, 2,864, and 2,908 kcal/kg) and two strains (Bovans White and Dekalb White) with and without Tylosin was conducted to determine the influence of dietary energy and antibiotic on performance, egg solids, and egg quality of two strains of commercial Leghorns during Phase 3 (from 55 to 64 week of age). This experiment lasted 10 weeks. Bovans White hens (n = 768) and Dekalb White hens (n = 768) at 55 week of age were randomly divided into 16 treatments (8 replicates of 12 birds per treatment). Bovans hens had significantly higher egg production than Dekalb hens, while Bovans hens had significantly lower egg weight, egg specific gravity and shell weight than Dekalb hens. Increasing dietary energy by the addition of poultry oil had no significant effect on performance, egg solids, and eggshell quality. An ideal dietary energy level for optimal performance could not be determined for laying hens during phase 3 (from wk 55 to 64 wk of age). There can be no fixed ideal dietary energy level for optimal profits during phase 3, due to varying feed ingredient prices and egg price. While Tylosin had no effect on performance and egg solids, Tylosin supplementation significantly reduced dirty eggs, resulting in a positive effect on egg quality of laying hens.

*Key words:* strains, energy, Tylosin, feed intake

## INTRODUCTION

Dietary energy is one of the most important nutrients in layer diets. Dietary energy is contributed from cereals such as corn and soy or supplemental fat such as poultry oil and corn oil. Dietary energy can significantly affect the cost of production because increasing dietary energy by the addition of poultry oil or corn oil can significantly decrease feed intake (Grobas et al., 1999; Harms et al., 2000, Wu et al., 2005a). Protein (amino acid) intake significantly decreased with increasing dietary energy (Wu et al., 2005a). Decreased protein and amino acid intake can decrease the performance of laying hens (Roland et al., 1992; Novak et al., 2004; Wu et al., 2005b). There might be an ideal energy to protein ratio for optimal performance during phase 1 (from 21 to 36 wk of age) (Wu et al., 2005a). As hens age, protein requirements of hens decrease so that more corn and less soy are used in the corn-soy diets resulting in higher dietary energy level. Increasing dietary energy by the addition of poultry oil or corn oil to low protein diets may not significantly decrease feed intake because high dietary energy level in the low protein diets may be sufficient for energy requirements of hens. It is necessary to have better understanding on the response of feed intake to dietary energy during phase 3.

Egg weight can significantly affect the profits of laying hens. Summers and Leeson (1993) and Jalal et al. (2006) reported that increasing dietary energy or supplementing with fat had no effect on egg weight. However, there are inconsistent results in effect of dietary energy on egg weight. Increasing dietary energy by the addition of corn oil or poultry oil increased egg weight (Keshavarz, 1995; Keshavarz and

Nakajima, 1995; Harms et al., 2000; Bohnsack et al., 2002; Sohail et al., 2003, Wu et al., 2005).

Increasing dietary energy by the addition of poultry oil significantly affected egg component (Wu et al., 2005). The change in percent egg components may affect egg solids. Although breaker egg industry has steadily increased during last 10 years, few experiments have been conducted to investigate the dietary energy effect on egg solids of current strains of Bovans White and Dekalb White during Phase 3. There is a wide range of dietary energy levels (2,684–2,992 kcal ME/kg) currently being used by the egg industry. However, there is limited information about the ideal dietary energy level for optimal performance and profits of commercial Leghorns during phase 3. It is necessary to have better understanding in optimizing dietary energy to improve performance and profits of laying hens during phase 3.

Antibiotics are substances that inhibit the growth of bacteria and related microorganism by interfering with essential metabolic functions. Antibiotic supplementation in feed increases weight gain and improves feed efficiency of broilers (Miller Publishing Co., 2000, Miles et al., 2006). However, antibiotics had no effect on body weight gain and feed utilization (Izat et al., 1990). Tylosin, an antibiotic, made naturally by the bacterium "*Streptomyces fradiae*" and acts to inhibit bacterial protein synthesis by inhibiting the 50S ribosome, a cellular structure of certain bacteria. Although Tylosin is widely used by poultry industry, there is very limited scientific literature about the effect of Tylosin on performance, egg component, and egg quality of laying hens.

The objective of this experiment was to determine the effect of dietary energy and antibiotic on performance, egg composition, and egg quality in Bovans White and Dekalb

White in Phase 3 (from 55 to 64 wk of age).

## MATERIAL AND METHODS

This study was a  $4 \times 2 \times 2$  factorial arrangement of four dietary energy levels (2,776, 2,820, 2,864, and 2,908 kcal/kg) and two strains (Bovans White and Dekalb White) with and without Tylosin. Ingredients and nutrient composition of experimental diets were showed in Table 1. Feed samples were sent to Elanco Animal Health, Memphis, TN, USA, for Tylosin activity analysis. The average chemical analyzed value of Tylosin in the diets was 33.7 g/ton of feed, which was very close to the expected value of 33 g/ton of feed.

In this experiment, Bovans White hens (n = 768) and Dekalb White hens (n = 768) at 55 week of age were randomly assigned into 16 treatments (16 replicates of 12 birds per treatment). The trial lasted 10 weeks. Replicates were equally distributed into upper and lower cages to minimize cage level effect. Three hens were housed in a  $40.6 \times 45.7$  cm<sup>2</sup> cage and five adjoining cages consisted of a replicate. All hens were housed in an environmentally-controlled house with temperature maintained as close to 25.6°C as possible. However, because the experiment was conducted during winter month, the house temperature was less than 25.6°C. The house had controlled ventilation and lighting (16 h/d). All hens were supplied with feed and water *ad libitum*. Egg production was recorded daily, feed consumption was recorded weekly, egg weight was recorded biweekly, and egg specific gravity was recorded monthly. Egg weight and egg specific gravity were measured using all eggs produced during two consecutive days. Egg specific gravity was determined using 11 gradient saline solutions varying in specific gravity from 1.060 to 1.100 with 0.005-unit increments (Holder and Bradford, 1979). Mortality was

determined daily and the feed consumption was adjusted accordingly. Body weight was obtained by weighing 3 hens per replicate at the end of the experiment. Egg mass and feed conversion (g feed/g egg) were calculated from egg production, egg weight and feed consumption.

Dirty eggs and crack eggs were determined by using all eggs produced during two consecutive days at the middle and end of the experiment. Dirty eggs were scored as following: 0 = clean egg; 1 = dirty area (adhering manure or blood) less than 25% of egg (light dirty egg); 2 = dirty area (adhering manure or blood) more than 25% of egg (heavy dirty egg). Dirty egg score per replicate =  $[(1 \times \text{number of light dirty egg}) + (2 \times \text{number of heavy dirty egg})] / \text{total number of egg per replicate}$ . A crack egg was determined by observing an egg having a broken or cracked shell under a light.

Three eggs from each replicate were collected at the end of experiment for measuring egg component. Three eggs from each replicate were collected to measure whole egg solid in the middle and the end of experiment. Albumen and yolk solid were measured by using three eggs from each replicate in the middle and the end of experiment. The procedures for measuring egg components, whole egg solid, and albumen and yolk solid were the same as those of Wu et al. (2005). Yolk color and Haugh unit were measured (3 eggs of each replicate) at the end of experiment by egg multi-tester EMT-5200 (Robotmation, Co., Ltd, Japan).

Data were analyzed by proc mixed procedures of Statistical Analysis System (SAS Institute, 2000) for a randomized complete block with a factorial treatment design. The factorial treatment arrangement consisted of 4 dietary energy levels, 2 layer strains, and 2 Tylosin levels. Dietary energy, strain, and Tylosin were fixed, while blocks were

random. The following model used to analyze data was as follows:

$$Y_{ijkl} = \mu + \alpha_i + \beta_j + \gamma_k + (\alpha\beta)_{ij} + (\alpha\gamma)_{ik} + (\beta\gamma)_{jk} + (\alpha\beta\gamma)_{ijk} + P_1 + \varepsilon_{ijkl}$$

Where  $Y_{ijkl}$  = individual observation,  $\mu$  = experimental mean,  $\alpha_i$  = dietary energy effect,  $\beta_j$  = layer strain effect,  $\gamma_k$  = Tylosin effect,  $(\alpha\beta)_{ij}$  = interaction between dietary energy and strain,  $(\alpha\gamma)_{ik}$  = interaction between dietary energy and Tylosin,  $(\beta\gamma)_{jk}$  = interaction between Tylosin and strain,  $(\alpha\beta\gamma)_{ijk}$  = interactions among dietary energy, strain, and Tylosin,  $P_1$  = effect of block,  $\varepsilon_{ijkl}$  = error component.

If differences in treatment means were detected by ANOVA, Duncan's Multiple Range Test was applied to separate means. Contrast statements were utilized to test for linear or quadratic dietary energy effects. A significance level of  $P \leq 0.05$  was used during analysis.

## RESULTS AND DISCUSSION

There was no significant interaction on all parameters among dietary energy, strain, and Tylosin (Table 2). Feed intake of Bovans hens was similar to that of Dekalb hens. Increasing dietary energy by the addition of poultry oil had no significant effect on feed intake. This was in agreement with that of Jalal et al. (2006), who reported that there was no significant effect of dietary energy on feed intake. This is inconsistent with that of Wu et al. (2005a), who reported that feed intake linearly decreased as dietary energy increased. This might be due to the smaller gap between dietary energy levels (approximately 44 kcal ME/kg) in this experiment, compared to that (approximately 80 kcal ME/kg) of Wu et al. (2005a). In addition, because this experiment was conducted in the winter, hens might have consumed as much as feed as possible to keep body

temperature. Energy effect on feed intake might be offset by low environmental temperature. There was no significant effect of Tylosin on feed intake.

Bovans hens had significantly higher egg production than Dekalb hens (Table 2). There was no significant dietary energy effect on egg production. This result was in agreement with that of Harms et al. (2000), Wu et al., (2005c), and Jalal et al. (2006), who reported that dietary energy had no effect on egg production. Antibiotic supplementation had no significant effect on egg production. This result was in agreement with that of Wu et al., (2005c), who reported there was no significant difference on egg production between hens fed the diets without antibiotics and hens fed the diets supplemented with antibiotics.

Egg weight of Bovans hens was significantly lower than that of Dekalb hens (Table 2). Increasing dietary energy by the addition of poultry oil had no significant effect on egg weight during phase 3 (from 55 to 64 wk of age). Egg weight of hens fed the diet without supplemental fat was numerically lower than that of hens fed the diets supplemented with fat ( $P \leq 0.0577$ ). Similarly, Wu et al. (2005d) reported that there was no response of egg weight to dietary energy for laying hens during phase 2 (from 40 to 51 wk of age). However, Wu et al. (2005a) reported that egg weight linearly increased with increasing dietary energy for laying hens during phase 1 (from 21 to 36 wk of age) and suggested that the increase of egg weight was mainly due to increased yolk weight for young hens during peak production. Sell et al. (1987) hypothesized that hens during early egg production might need more exogenous fat to supply lipids for egg yolk development. Therefore, hens might have the ability to synthesize enough lipoprotein for

yolk formation after early egg production. Tylosin supplementation had no significant effect on egg weight.

There was no significant difference on egg mass and feed conversion between Bovans hens and Dekalb hens (Table 2). Increasing dietary energy had no significant effect on egg mass and feed conversion. As dietary energy increased from 2,776 to 2,908 kcal ME/kg, feed conversion numerically decreased from 1.95 to 1.88, resulting in a 3.6% improvement of feed conversion. When dietary energy increased from 2,776 to 2,908 kcal ME/kg, hens adjusted feed intake, according to egg yield, to achieve a constant energy intake so that the similar quantities of dietary energy (5.4-5.5 kcal) were used to produce 1 g of egg. There was no significant effect of Tylosin on egg mass or feed conversion. Similarly, Izat et al. (1990) reported that antibiotics had no effect on feed conversion in broilers. Increasing dietary energy by the addition of poultry oil had no significant effect on body weight and mortality in both strains. There was no significant difference on body weight and mortality between hens fed the diets without Tylosin and hens fed the diets with Tylosin. Because increasing dietary energy by the addition of poultry oil had no significant effect on feed intake, egg weight, and feed conversion, an ideal dietary energy level for optimal performance could not be determined for laying hens during phase 3 (from wk 55 to 64 wk of age).

Either strain or Tylosin had no significant effect on whole egg solid, albumen solid, yolk solid, Haugh unit, or yolk color (Table 3). There was no significant dietary energy effect on whole egg solid, albumen solid, and yolk solid. Similar results were reported by Wu et al. (2005a). Increasing dietary energy by the addition of poultry oil had no significant effect on Haugh unit and yolk color.

Strain had a significant effect on egg specific gravity (Table 4). Egg specific gravity of Dekalb hens was significantly higher than that of Bovans hens because Dekalb hens had significantly higher shell weight than Bovans hens (Table 4). Similar results were reported by Wu et al. (2005a). These results suggest that Deklab hens have better eggshell quality than Bovans hens. Increasing dietary energy by the addition of poultry oil had no significant effect on egg specific gravity, shell weight, shell thickness, dirty eggs and crack eggs. Tylosin supplementation had no effect on egg specific gravity, shell weight, shell thickness, and crack eggs. Hens fed the diets supplemented with Tylosin had significantly lower dirty eggs than hens fed the diets with Tylosin. Dirty eggs were significantly increased by wet excreta (Smith et al., 2000), which can be caused by gastrointestinal disorders like diarrhea in hens. Tylosin supplementation might have prevented and decreased gastrointestinal disorders in hens, resulting in less dirty eggs.

The Economic Feeding and Management Program developed by Roland et al. (1998, 2000) was used to calculate profits of different dietary energy levels at poultry oil prices. When poultry oil price were \$0.22/kg, maximum profits per dozen eggs were obtained in hens fed the diet containing 2,908 kcal ME/kg of dietary energy (Table 5). However, when poultry oil price increased to \$0.40/kg, maximum profit was obtained in hens fed the diet containing 2,776 kcal ME/kg of dietary energy. Because feed ingredient prices and egg price often vary, there can be no fixed ideal dietary energy level for optimal profits during phase 3 (wk 55 to 64).

In conclusion, Bovans hens had significantly higher egg production than Dekalb hens, while Bovans hens had significantly lower egg weight, egg specific gravity and shell weight than Dekalb hens. Increasing dietary energy by the addition of poultry oil

had no significant effect on performance, egg solids, and eggshell quality. An ideal dietary energy level for optimal performance could not be determined for laying hens during phase 3 (from wk 55 to 64 wk of age). There can be no fixed ideal dietary energy level for optimal profits during phase 3, due to varying feed ingredient prices and egg price. While Tylosin had no effect on performance and egg solids, Tylosin supplementation significantly reduced dirty eggs, resulting in a positive effect on egg quality of laying hens.

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**Table 1.** Ingredients and nutrient composition of experimental diets

Ingredient	Diet 1	Diet 2	Diet 3	Diet 4	Diet 5	Diet 6	Diet 7	Diet 8
Corn (%)	65.70	65.70	64.68	64.68	63.66	63.66	62.63	62.63
Soybean meal (%)	22.41	22.41	22.49	22.49	22.57	22.57	22.66	22.66
CaCO <sub>3</sub> (%)	7.57	7.57	7.57	7.57	7.57	7.57	7.56	7.56
Hardshell <sup>1</sup> (%)	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	1.40	1.40	1.41	1.41
Poultry oil (%)	0.00	0.00	0.93	0.93	1.87	1.87	2.80	2.80
NaCl (%)	0.36	0.36	0.36	0.36	0.36	0.36	0.36	0.36
Vitamin Premix <sup>2</sup> (%)	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
Mineral premix <sup>3</sup> (%)	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
DL-Methionine (%)	0.06	0.06	0.07	0.07	0.07	0.07	0.07	0.07
Tylosin	-	+	-	+	-	+	-	+
Calculated analysis								
Crude protein (%)	16.05	16.05	16.01	16.01	15.97	15.97	15.93	15.93
ME (kcal/kg)	2776	2776	2820	2820	2864	2864	2908	2908
Ca (%)	4.15	4.15	4.15	4.15	4.15	4.15	4.15	4.15
Available phosphorus (%)	0.36	0.36	0.36	0.36	0.36	0.36	0.36	0.36
Methionine (%)	0.33	0.33	0.34	0.34	0.34	0.34	0.34	0.34
Metionine+Cystine (%)	0.62	0.62	0.62	0.62	0.62	0.62	0.62	0.62
Lysine (%)	0.83	0.83	0.83	0.83	0.83	0.83	0.83	0.83

<sup>1</sup>Hardshell = large particle (passing US mesh #4 and retained by US mesh #6) CaCO<sub>3</sub> supplied by Franklin Industrial Minerals, Lowell, Florida.

<sup>2</sup>Provided per kilogram of diet: vitamin A (as retinyl acetate), 8,000 IU; cholecalciferol, 2,200 ICU; vitamin E (as DL- $\alpha$ -tocopheryl acetate), 8 IU; vitamin B<sub>12</sub>, 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<sub>1</sub> (thiamin mononitrate), 1 mg; pyridoxine, 2.2 mg; biotin, 0.05 mg; vitamin K (menadione sodium bisulfate complex), 2 mg.

<sup>3</sup>Provided per kilogram of diet: manganese, 65 mg; iodine, 1 mg; iron, 55 mg; copper, 6 mg; zinc, 55 mg; selenium, 0.3 mg.

<sup>4</sup>Elanco Animal Health, Memphis, TN

**Table 2.** Influence of dietary energy and Tylosin on performance of Bovans White and Dekalb White from 55 to 64 wk of age

Factor		Feed intake (g/hen per day)	Egg produc- tion (%)	Egg weight (g)	Egg mass (g egg/hen per d)	Feed conversion (g of feed/of g egg)	Energy intake (Kcal ME/g egg)	Body weight (kg)	Mort ality (%)
Strain	Bovans	109.7	88.5 <sup>a</sup>	64.80 <sup>b</sup>	57.35	1.91	5.4	1.48	3.17
	Dekalb	109.9	85.6 <sup>b</sup>	66.64 <sup>a</sup>	57.04	1.93	5.5	1.53	3.81
Dietary energy (Kcal/kg)	2776	110.8	87.3	65.19	56.91	1.95	5.4	1.50	3.27
	2820	110.6	86.4	65.85	56.89	1.94	5.5	1.51	4.02
	2864	108.2	86.6	65.68	57.88	1.90	5.4	1.50	3.69
	2908	109.7	88.1	66.16	58.29	1.88	5.5	1.50	2.99
Tyosin	-	109.9	87.2	65.84	57.41	1.91	5.4	1.52	2.96
	+	109.7	87.0	65.60	57.07	1.92	5.5	1.51	4.03
Pooled SEM		1.72	1.92	0.50	1.32	0.05	0.12	0.08	0.17
-----Probability-----									
Main effects and interactions									
Strain		NS	0.0114	0.0001	NS	NS	NS	NS	NS
Dietary energy		NS	NS	0.0577	NS	NS	NS	NS	NS
Tylosin		NS	NS	NS	NS	NS	NS	NS	NS
Strain × Energy		NS	NS	NS	NS	NS	NS	NS	NS
Strain × Tylosin		NS	NS	NS	NS	NS	NS	NS	NS
Energy × Tylosin		NS	NS	NS	NS	NS	NS	NS	NS
Str. × Ene. × Tyl		NS	NS	NS	NS	NS	NS	NS	NS

<sup>a-c</sup>Means within a column and under each main effect with no common superscripts differ significantly

**Table 3.** Influence of dietary energy and Tylosin on egg solid, Haugh unit, and yolk color in Bovans White and Dekalb White from 55 to 64 wk of age

Factor		% of solid			Haugh unit	Yolk color
		Whole egg	Albumen	Yolk		
Strain	Bovans	23.74	11.92	50.60	71.53	6.55
	Dekalb	23.66	11.89	50.70	71.37	6.75
Dietary energy (Kcal/kg)	2776	23.67	11.92	50.38	73.94	6.83
	2820	23.64	11.86	50.58	71.94	6.63
	2864	23.65	11.84	50.78	69.17	6.71
	2908	23.83	12.01	50.87	70.74	6.44
Tylosin	-	23.88	11.92	50.64	71.69	6.64
	+	23.52	11.89	50.66	71.21	6.67
Pooled SEM		0.28	0.15	0.89	3.19	0.22
ANOVA		-----Probability-----				
Main effects and interactions						
Strain		NS	NS	NS	NS	NS
Dietary energy		NS	NS	NS	NS	NS
Tylosin		NS	NS	NS	NS	NS
Strain × Energy		NS	NS	NS	NS	NS
Strain × Tylosin		NS	NS	NS	NS	NS
Energy × Tylosin		NS	NS	NS	NS	NS
Str. × Ene. × Tylosin		NS	NS	NS	NS	NS

<sup>a-c</sup>Means within a column and under each main effect with no common superscripts differ significantly

**Table 4.** Influence of dietary energy and Tylosin on egg quality in Bovans White and Dekalb White from 55 to 64 wk of age

Factor		Egg specific gravity (unit)	Shell weight (g)	Shell thickness (mm)	Dirty egg scores	Crack egg (%)
Strain	Bovans	1.0827 <sup>b</sup>	5.82 <sup>b</sup>	0.376	0.17	0.019
	Dekalb	1.0854 <sup>a</sup>	6.06 <sup>a</sup>	0.382	0.23	0.018
Dietary energy (Kcal/kg)	2776	1.0846	5.88	0.378	0.24	0.024
	2820	1.0838	5.95	0.379	0.18	0.016
	2864	1.0838	5.93	0.379	0.19	0.017
	2908	1.0840	5.99	0.381	0.19	0.017
Tylosin	-	1.0842	5.97	0.382	0.23 <sup>a</sup>	0.021
	+	1.0839	5.91	0.376	0.17 <sup>b</sup>	0.016
Pooled SEM		0.0005	0.23	0.016	0.06	0.017
-----Probability-----						
Main effects and interactions						
Strain		0.0001	0.0479	NS	NS	NS
Dietary energy		NS	NS	NS	NS	NS
Tylosin		NS	NS	NS	0.0422	NS
Strain × Energy		NS	NS	NS	NS	NS
Strain × Tylosin		NS	NS	NS	NS	NS
Energy × Tylosin		NS	NS	NS	NS	NS
Str. × Ene. × Tylosin		NS	NS	NS	NS	NS

<sup>a-b</sup>Means within a column and under each main effect with no common superscripts differ significantly

**Table 5.** Influence of dietary energy and poultry oil price on profits<sup>1,2</sup> from 55 to 64 wk of age

	Dietary energy content (kcal ME/kg)			
	2776	2820	2864	2908
	Returns <sup>3</sup> (\$/dozen)			
High fat price (\$0.40/kg)	0.085	0.081	0.083	0.082
Low fat price (\$0.22/kg)	0.086	0.084	0.088	0.089

<sup>1</sup> Corn price = \$0.12/kg, soy price = \$0.20/kg, CaCO<sub>3</sub> = \$0.03/kg, hard shell = \$0.03/kg, Dicalcium phosphate = \$0.27 cents/kg, salt = \$0.06/kg, Vitamin premix = \$2.67/kg, mineral premix = \$0.59/kg, DL-methionine = \$2.59/kg.

<sup>2</sup> The egg price spread between medium and large eggs was 11 cents

<sup>3</sup> Returns (R) were calculated using the equation:  $R = \text{UBEP} - \text{NR} - \text{PC} - \text{FdC}$ , where UBEP = Uner Barry Egg Price, NR = nest run into package product delivered, PC = production cost, and FdC = feed cost, as described by Roland et al. (1998, 2000).

V. EFFECT OF MOLTING METHOD AND DIETARY ENERGY  
ON POST-MOLT PERFORMANCE, EGG COMPOSITION, EGG SOLIDS,  
AND EGG QUALITY OF BOVANS AND DEKALB WHITE  
HENS DURING SECOND CYCLE PHASE 1

**ABSTRACT:** A  $4 \times 2 \times 2$  factorial experiment of four dietary energy levels (2,767, 2,806, 2,846, 2,886 kcal ME/kg), two molting methods (feed withdrawal and no salt diet), and two strains (Bovans White and Dekalb White) was conducted to determine the effect of dietary energy and molting method on post-molt performance of two strains of commercial Leghorns. Before molt, either Bovans hens or Dekalb hens were randomly divided into two groups. Feed was withdrawn from half of the hens (66 wk of age) for 9 days. Bovans and Dekalb hens lost 32.8% and 32.9% body weight, respectively. A molt feed was fed from day 10 to 28. The other half of hens was fed a no salt diet for 28 days. Bovans and Dekalb hens lost 16.4% and 17.8% body weight at the end of molt, respectively. After molt, Bovans White hens (n = 720) and Dekalb White hens (n = 720) at 70 wk of age were randomly divided into 16 treatments (6 replicates of 15 birds per treatment). Bovans hens had significantly higher egg production and lower egg weight and egg specific gravity than Dekalb hens. With increasing dietary energy hens adjusted feed intake to achieve a constant energy intake so that the similar quantities of dietary energy (5.8-5.9 kcal) were used to produce 1 g of egg. Dietary energy had a significant

effect on yolk color. Dietary energy had no significant effect on feed intake, egg production, egg weight, egg mass, feed conversion, body weight, mortality, percent of egg components, percent of egg solids, egg specific gravity, and Haugh unit after molt. An ideal dietary energy level for optimal post-molt performance could not be determined, and there can be no fixed ideal dietary energy level for optimal profits for hens during second cycle phase 1 (from 70 to 81 wk of age). There were no significant differences in feed intake, egg weight, egg mass, feed conversion, body weight, and mortality between hens molted by feed withdrawal and hens molted by no salt diet. Egg production of hens molted by feed withdrawal was significantly higher than that of hens molted by no salt diet in wk 6, 7, 9, 10, and 11. Hens molted by feed withdrawal had significantly higher egg specific gravity than hens molted by no salt diet during wk 8. There were no significant differences in overall average egg production and egg specific gravity due to molting method during second cycle phase 1 (from 70 to 81 wk of age). Other than slightly reduced egg production and egg specific gravity, feeding no salt diet to induce molt could be used as an alternative for conventional feed withdrawal method.

*Key words:* strains, dietary energy, molting method

## **INTRODUCTION**

Induced molting has been widely used to revive the productivity of aging laying hens by the egg industry in US for nearly a century. Induced molting increased egg production, renewed feather, and improved egg quality including egg specific gravity and shell thickness (Lee, 1982; Barker et al., 1983; Bell, 2003). Induced molting not only improves performance, but also extends the economically useful life of laying hens from 80 wk to 110 wk or even to 140 wk of age, which reduce the quantity of replacement

pullets. Feed withdrawal with light reduction is most widely used to induce molting in USA. In recent years, induced molting by conventional feed withdrawal has been vigorously criticized by animal welfare groups. The United Egg producers (UEP) Scientific Advisory Committee on Animal Welfare urged researchers and producers to work together to develop alternatives to feed withdrawal for molting (UEP, 2002).

Effectiveness of several nonfeed removal molting diets including low-sodium diets (Whitehead and Shannon, 1974; Begin and Johnson, 1976; Nesbeth et al., 1976a, b; Whitehead and Sharp, 1976; Monsi and Enos, 1977; Ross and Herrick, 1981; Harms 1981, 1983; Naber et al., 1984, Said et al., 1984), low-calcium diets (Gilbert et al., 1981), high-Zinc diets (Shippee et al., 1979; Berry and Brake, 1985; Bar et al., 2003), and low protein and low energy diets (Koelkebeck et al., 2001; Biggs et al., 2003) have been evaluated. Naber et al. (1984) and Said et al. (1984) reported that low sodium diets can be effective in recycling hens for a second period of egg production. However, the low-salt diets decreased egg production and eggshell thickness during post-molt egg production (Ross and Herrick, 1981). Different strains showed different performance after molt, compared to conventional feed withdrawal (Said et al., 1984; Bell and Kuney, 2004). In addition, there is no or little research in effect of no salt diets on egg composition and egg solids, which are important factors influencing profits of breaker egg market. Therefore, it is necessary to have more knowledge concerning the effect of no salt diets on performance, egg quality, egg component, and egg solids in current strains of commercial Leghorns (Bovans and Dekalb) to determine acceptable alternatives for conventional feed withdrawal method.

Dietary energy by the addition of poultry oil or corn oil had significant effect on feed intake (Grobas et al., 1999; Harms et al., 2000; Wu et al., 2005a, b, c), egg weight (Keshavarz., 1995; Keshavarz and Nakajima, 1995; Harms et al., 2000; Bohnsack et al., 2002; Sohail et al., 2003; Wu et al., 2005a), feed conversion (Wu et al., 2005a, b), and egg specific gravity (Wu et al., 2005a). However, Summers and Leeson (1993) and Jalal et al. (2006) reported that dietary energy had no effect on feed intake and egg weight. Feed intake and egg weight can significantly affect profits of egg producers. As hens age, protein requirement decreases so that more corn and less soybean meal are used in the diets, which result in more energy in the diets for older hens. Increasing dietary energy in the elder hens may not decrease feed intake as well as in young hens. There is very limited literature information on effect of dietary energy on performance, egg composition, egg solids, egg quality, and profits of post-molt hens. It is necessary to have a better understanding concerning how to optimize the use of dietary energy to obtain optimal performance and profits of post-molt laying hens.

The objective of this study is to determine the effect of molting method and dietary energy on performance, egg component, egg solids, egg quality, and profits in Bovans White and Dekalb White hens during second cycle phase 1 (wk 70 to 81 of age).

## **MATERIAL AND METHODS**

This experiment was a  $4 \times 2 \times 2$  factorial arrangement of four dietary energy levels (2,767, 2,806, 2,846, and 2,886 kcal ME/kg), two molting methods (feed withdrawal and no salt diet), and two strains (Bovans White and Dekalb White). The experiment lasted 12 weeks. Ingredients and nutrient composition of experimental diets were shown in Table 1. From wk 70 to 75, four experimental diets contained 17.3%

protein level. From wk 76 to 81, protein level in four diets was decreased to 15.3% to decrease egg weight, because egg weight from wk 70 to 75 was higher than recommended egg weight of Bovans White Management Guide (Anonymous, 2003).

Feed was withdrawn from half of the hens (66 wk of age) for 9 days. Bovans and Dekalb hens lost 32.8% and 32.9% body weight, respectively. A molt feed was fed from day 10 to 28 (Table 1). The other half of hens was fed a no salt diet for 28 days (Table 1). Bovans and Dekalb hens lost 16.4% and 17.8% body weight at the end of molt, respectively. Hens molted by feed withdrawal stopped laying by day 4, while hens molted by no salt diet did not stop laying until day 17. After molt, Bovans White hens (n = 720) and Dekalb White hens (n = 720) at 70 wk of age were randomly divided into 16 treatments (6 replicates of 15 birds per treatment). Replicates were equally distributed into upper and lower cages to minimize cage level effect. Three hens were housed in a  $40.6 \times 45.7 \text{ cm}^2$  cage and five adjoining cages consisted of a replicate. All hens were housed in an environmentally-controlled house with temperature maintained close to  $25.6^\circ\text{C}$  as possible. However, because the experiment was conducted during winter month, the house temperature was less than  $25.6^\circ\text{C}$ . The light period was reduced from 16 hr to 8 hr daily from day 1. On the 29<sup>th</sup> day, the light period was returned to 16 hr daily. All hens were supplied with feed and water *ad libitum*. Egg production was recorded daily, feed consumption was recorded weekly, and egg weight was recorded biweekly, and egg specific gravity was recorded monthly. Egg weight and egg specific gravity were measured using all eggs produced during two consecutive days. Egg specific gravity was determined using 11 gradient saline solutions varying in specific gravity from 1.060 to 1.100 with 0.005-unit increments (Holder and Bradford, 1979). Mortality was

determined daily and the feed consumption was adjusted accordingly. Body weight was obtained by weighing 3 hens per replicate at the end of the experiment. Egg mass and feed conversion (g feed/g egg) were calculated from egg production, egg weight and feed consumption.

Egg components were measured using two eggs from each replicate in the middle and the end of experiment. Two eggs from each replicate were collected to measure whole egg solids in the middle and the end of experiment. Albumen and yolk solid were measured by using two eggs from each replicate in the middle and the end of experiment. The procedures for measuring egg components, whole egg solids, and albumen and yolk solid were the same as those of Wu et al. (2005). Yolk color and Haugh unit were measured (3 eggs of each replicate) at the end of experiment by egg multi-tester EMT-5200 (Robotmation, Co., Ltd, Tokyo, Japan).

Data were analyzed by proc mixed procedures of Statistical Analysis System (SAS Institute, 2000) for a randomized complete block with a factorial treatment design. Dietary energy, molting method, and strain were fixed, while blocks were random. The factorial treatment arrangement consisted of 4 dietary energy levels, 2 molting methods, and 2 layer strains. The following model used to analyze data was as follows:

$$Y_{ijkl} = \mu + \alpha_i + \beta_j + \gamma_k + (\alpha\beta)_{ij} + (\beta\gamma)_{jk} + (\alpha\gamma)_{ik} + (\alpha\beta\gamma)_{ijk} + P_l + \varepsilon_{ijkl}$$

Where  $Y_{ijkl}$  = individual observation,  $\mu$  = experimental mean,  $\alpha_i$  = dietary energy effect,  $\beta_j$  = molting method,  $\gamma_k$  = layer strain effect,  $(\alpha\beta)_{ij}$  = interaction between dietary energy and molting method,  $(\beta\gamma)_{jk}$  = interaction between molting method and strain,  $(\alpha\gamma)_{ik}$  = interaction between dietary energy and strain,  $(\alpha\beta\gamma)_{ijk}$  = interaction among dietary energy, molting method, and strain,  $P_k$  = effect of block,  $\varepsilon_{ijkl}$  = error component.

If differences in treatment means were detected by ANOVA, Duncan's Multiple Range Test was applied to separate means. A significance level of  $P \leq 0.05$  was used during analysis.

## **RESULTS AND DISCUSSION**

There was no interaction among strain, dietary energy, and molting method in feed intake, egg production, egg weight, egg mass, feed conversion, energy intake, body weight, and mortality. Strain had no significant effect on feed intake (Table 2). With increasing dietary energy, feed intake did not linearly decrease. This is inconsistent with that of Wu et al. (2005a), who reported that feed intake linearly decreased as dietary energy increased. This might be due to the smaller gap between dietary energy levels (approximately 40 kcal ME/kg) in this experiment, compared to that (approximately 80 kcal ME/kg) of Wu et al. (2005a). In addition, because the experiment was conducted during winter, hens might over-consume the diet, which caused over energy intake of hens. Although feed intake of hens molted by feed withdrawal was significantly higher than that of hens molted by no salt diet in wk 2, 5, and 9, there was no significant difference in overall feed intake between hens molted by feed withdrawal and hens molted by no salt diet. This result was very similar to that of Naber et al. (1984).

Bovans hens had significant higher egg production, and lower egg weight than Dekalb hens (Table 2). Dietary energy had no significant effect on egg production (Table 2). This was consistent with that of Wu et al. (2005b, and c) and Jalal et al. (2006), who reported that there was no significant effect of dietary energy level on egg production (Table 2). Dietary energy had no significant effect on egg weight (Table 2), which is inconsistent with that of Wu et al. (2005a), who reported that egg weight linearly

increased with increasing dietary energy. Wu et al. (2005a) suggested that the increase of egg weight was mainly due to increased yolk weight for young hens. Sell et al. (1987) hypothesized that hens during early egg production might need more exogenous fat to supply lipids for egg yolk development. Percent egg yolk did not increase with increasing dietary energy by the addition of poultry oil (Table 3). This suggested that older hens might have the ability to synthesize enough lipoprotein so that they did not need more exogenous fat to supply lipids for egg yolk development.

Egg production of hens molted by feed withdrawal was numerically lower than that of hens molted by no salt diet in wk 1 and 2, and then increased quickly from wk 3 (Figure 1). Hens molted by feed withdrawal had significantly higher egg production than hens molted by no salt diet in wk 6, 7, 9, 10, and 11 (Figure 1). However, there was no significant difference in overall average egg production between hen molted by feed withdrawal and hens molted by no salt diet (Table 2). Similarly, Said et al. (1983) and Naber et al. (1984) reported that molting method had no effect on egg production. In contrast, Ross and Herrick (1984) reported that hens molted by feed withdrawal had significant higher egg production than hens molted by no salt diet. The difference among researchers might be due to higher sodium content in no salt diet of Ross and Herrick (1981).

Egg weight of hens molted by feed withdrawal was significantly lower than that of hens molted by no salt diet in wk1, and then increased quickly from wk 2 (Figure 2). Hens molted by feed withdrawal had significantly higher egg weight than hens molted by no salt diet in wk4. From wk 5, there was no significant difference between hens molted by feed withdrawal and hens molted by no salt diet. Overall average egg weight of hens

molted by feed withdrawal was the same as that of hens molted by no salt diets (Table 2). This result was in agreement with those of Ross and Herrick (1981), Said et al. (1983) and Naber et al. (1984), who reported that there was no significant difference in egg weight due to molting method.

There was no significant effect of strain on egg mass (Table 2). Dietary energy had no significant effect on egg mass. This result was in agreement with that of Wu et al. (2005a, b, and c), who reported that there was no significant effect of dietary energy on egg mass. Strain had no significant effect on feed conversion. As dietary energy increased from 2,767 to 2,886 kcal ME/kg, feed conversion numerically improved from 2.13 to 2.05, resulting in a 3.8% improvement of feed conversion. When dietary energy increased from 2,767 to 2,886 kcal ME/kg, hens adjusted feed intake to achieve a constant energy intake so that the similar quantities of dietary energy (5.8-5.9 kcal) were used to produce 1 g of egg. Because increasing dietary energy by addition of poultry oil had no significant effect on feed intake, egg mass and feed conversion, an ideal dietary energy level for optimal post-molt performance could not be determined. This conclusion was different from that of Wu et al. (2005a), who reported that there may be an ideal energy/protein ratio for optimal performance during phase 1 (wk 70 to 81 of age), probably because egg weight linearly increased and feed intake linearly decreased with increasing dietary energy in the experiment of Wu et al. (2005a).

Hens molted by feed withdrawal had the same feed conversion and egg mass as hens molted by no salt diet. In addition, hens molted by feed withdrawal used the same quantity of dietary energy (5.8 kcal) to produce 1 g egg (Table 2). This suggests that molting method has no effect on efficiency of dietary energy utilization. Strain or dietary

energy had no influence on body weight and mortality (Table 2). There was no significant difference in body weight of hens and mortality between hens molted by feed withdrawal and hens molted by no salt diet. Similar result was reported by Naber et al. (1984) and Said et al. (1984). However, Ross and Herrick (1981) reported that hens molted by no salt diet had significantly higher mortality than hens molted by feed withdrawal.

There was no interaction among strain, dietary energy, and molting method in percent of yolk, albumen, and shell, percent of whole egg solids, yolk solid, and albumen solid, egg specific gravity, Haugh unit, and yolk color (Table 3). While Bovans hens had similar percent of yolk and albumen as Dekalb hens, Bovans hens had significantly lower percent of shell and egg specific gravity than Dekalb hens. These results and those of Wu et al. (2005) suggest that Dekalb hens had better eggshell quality than Bovans hens. Bovans hens had similar percent of whole egg solids, yolk solid, and albumen solid as Dekalb hens. Increasing dietary energy had no effect on egg specific gravity (Table 3) probably because egg weight did not increase with increasing dietary energy. Dietary energy had no effect on percent of yolk, albumen, and shell and percent of whole egg solids, yolk solid, albumen solid. These results were in agreement with Wu et al. (2005a), who reported that there was no significant effect of dietary energy on egg component and egg solids, which are very important factors influencing profits of breaker egg industry.

Percent of yolk, albumen, and shell and percent of whole egg solids, yolk solid, and albumen solid of hens molted by feed withdrawal were similar to those of hens molted by no salt diet (Table 3). Molting method had no effect on post-molt egg composition and egg solids. Although hens molted by feed withdrawal had significantly

higher egg specific gravity than hens molted by no salt diet during wk 8, there was no significant difference in overall average egg specific gravity due to molting method. In addition, hens molted by feed withdrawal had the same percent of eggshell as hens molted by no salt diet. These results and the results of Ross and Herrick (1981), Naber et al. (1984), and Said et al. (1984) suggested molting methods had no significant effect on eggshell quality.

Dekalb hens had significantly higher Haugh unit than Bovans hens. Similar result was reported by Wu et al. (2005a). Dietary energy had no effect on Haugh unit. Hens molted by feed withdrawal had similar Haugh unit as hens molted by no salt diet. This result was in agreement with that of Ross and Herrick (1981) and Said et al. (1984), who reported that there was no significant difference in Haugh unit due to molting methods. Either strain or molting method had no significant effect on yolk color. Increasing dietary energy had a significant effect on yolk color. Yolk color linearly increased with increasing dietary energy. This might be due to that poultry oil contained certain level of pigments. With increasing poultry oil, more pigments were included in the diets.

The Economic Feeding and Management Program developed by Roland et al. (1998, 2000) was used to calculate profits of different dietary energy levels at poultry oil prices. When poultry oil price were \$0.26/kg, maximum profits per dozen eggs were obtained in hens fed the diet containing 2,846 kcal ME/kg of dietary energy (Table 4). However, when poultry oil price increased to \$0.40/kg, maximum profits was obtained in hens fed the diet containing 2,767 kcal ME/kg of dietary energy. Because the ideal dietary energy level for optimal performance may not exist for post-molt hens and feed

ingredient prices and egg price often vary, there can be no fixed ideal dietary energy level for optimal profits for post-molt hens.

In conclusion, Dekalb hens had significantly higher egg weight, percent of eggshell, egg specific gravity, and Haugh unit than Bovans hens, while Dekalb hens had significantly lower egg production than Bovans hens. There was a linear response of yolk color to increased dietary energy. Increasing dietary energy by the addition of poultry oil had no significant effect on feed intake, egg production, egg weight, egg mass, feed conversion, body weight, mortality, percent of egg components, percent of egg solids, egg specific gravity, and Haugh unit after molt. An ideal dietary energy level for optimal post-molt performance could not be determined, and there can be no fixed ideal dietary energy level for optimal profits for hens during second cycle phase 1 (from 70 to 81 wk of age). Egg production of hens molted by feed withdrawal was significantly higher than that of hens molted by no salt diet in wk 6, 7, 9, 10, and 11. Hens molted by feed withdrawal had significantly higher egg specific gravity than hens molted by no salt diet during wk 8. There were no significant differences in overall average egg production and egg specific gravity due to molting method during second cycle phase 1 (from 70 to 81 wk of age). Other than slightly reduced egg production and egg specific gravity, feeding no salt diet to induce molt could be used as an alternative for conventional feed withdrawal method.

#### **ACKNOWLEDGMENTS**

The authors thank Centurion Poultry, Inc, Lexington, GA, and Ridley Inc., Mankato, MN, for funding support of this research.

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**Table 1.** Ingredients and nutrient composition of experimental diets

Ingredient	No salt diet	Molt diet following feed withdrawal	Post-molt diets							
			Wk 70 to 75				Wk 76 to 81			
			Diet 1	Diet 2	Diet 3	Diet 4	Diet 1	Diet 2	Diet 3	Diet 4
Corn (%)	73.12	72.78	62.42	61.50	60.58	59.66	66.29	65.38	64.46	63.54
Soybean meal (%)	20.02	20.51	25.67	25.75	25.82	25.89	20.86	20.94	21.01	21.08
CaCO <sub>3</sub> (%)	4.13	4.02	4.99	4.99	4.99	4.98	6.42	6.42	6.41	6.41
Hardshell <sup>1</sup> (%)	0.00	0.00	4.00	4.00	4.00	4.00	4.00	4.00	4.00	4.00
Dical. phosphate (%)	1.39	1.38	1.70	1.71	1.71	1.71	1.52	1.52	1.53	1.53
Poultry oil (%)	0.25	0.25	0.25	1.09	1.94	2.78	0.00	0.83	1.68	2.52
NaCl (%)	0.00	0.38	0.36	0.36	0.36	0.36	0.36	0.36	0.36	0.37
Vitamin Premix <sup>2</sup> (%)	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
Mineral premix <sup>3</sup> (%)	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
DL-Methionine (%)	0.19	0.05	0.10	0.10	0.10	0.11	0.05	0.05	0.06	0.06
Calculated analysis										
Crude protein (%)	15.9	15.8	17.3	17.3	17.3	17.3	15.3	15.3	15.3	15.3
ME (kcal/kg)	2989	2996	2767	2806	2846	2886	2767	2806	2846	2886
Ca (%)	2.00	2.00	4.00	4.00	4.00	4.00	4.50	4.50	4.50	4.50
A. P. (%)	0.36	0.36	0.42	0.42	0.42	0.42	0.38	0.38	0.38	0.38
Sodium	0.02	0.18	0.17	0.17	0.17	0.17	0.17	0.17	0.17	0.17
Methionine (%)	0.47	0.44	0.38	0.38	0.38	0.38	0.31	0.31	0.31	0.31
Met+Cys (%)	0.73	0.73	0.69	0.69	0.69	0.69	0.59	0.59	0.59	0.59
Lysine (%)	0.80	0.79	0.92	0.92	0.92	0.92	0.78	0.78	0.78	0.78

<sup>1</sup>Hardshell = large particle (passing US mesh #4 and retained by US mesh #6) CaCO<sub>3</sub> supplied by Franklin Industrial Minerals, Lowell, Florida.

<sup>2</sup>Provided per kilogram of diet: vitamin A (as retinyl acetate), 8,000 IU; cholecalciferol, 2,200 ICU; vitamin E (as DL- $\alpha$ -tocopheryl acetate), 8 IU; vitamin B<sub>12</sub>, 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<sub>1</sub> (thiamin mononitrate), 1 mg; pyridoxine, 2.2 mg; biotin, 0.05 mg; vitamin K (menadione sodium bisulfate complex), 2 mg.

<sup>3</sup>Provided per kilogram of diet: manganese, 65 mg; iodine, 1 mg; iron, 55 mg; copper, 6 mg; zinc, 55 mg; selenium, 0.3 mg.

**Table 2.** Influence of dietary energy and molting method on post-molt performance of Bovans white and Dekalb white hens from 70 to 81 wk of age

Factor		Feed intake (g/hen per day)	Egg producti on (%)	Egg weight (g)	Egg mass (g egg/hen per d)	Feed conversion (g of feed/of g egg)	Energy intake (Kcal ME/g egg)	Body weight (kg)	Mort ality (%)
Strain	Bovans	112.5	79.9	68.6	54.8	2.06	5.80	1.76	2.93
	Dekalb	112.4	76.9	69.8	53.4	2.10	5.92	1.76	2.49
Dietary energy (Kcal/kg)	2767	112.8	77.1	69.0	53.1	2.13	5.88	1.81	3.88
	2806	114.2	79.1	69.4	54.9	2.08	5.84	1.76	2.36
	2846	111.5	79.2	68.7	54.4	2.05	5.83	1.70	2.87
	2886	111.3	78.3	69.6	54.5	2.05	5.90	1.77	1.75
Molt regimen	NS	111.4	77.7	69.2	53.7	2.08	5.86	1.78	1.95
	FW	113.5	79.2	69.2	54.8	2.08	5.86	1.74	3.48
Pooled SEM		2.27	1.82	0.49	1.52	0.05	0.14	0.15	1.74
-----Probability-----									
Main effects and interactions									
Strain		NS	0.0014	0.0001	NS	NS	NS	NS	NS
Dietary energy		NS	NS	NS	NS	NS	NS	NS	NS
Molt regimen		NS	NS	NS	NS	NS	NS	NS	NS
Strain × Energy		NS	NS	NS	NS	NS	NS	NS	NS
Strain × Molt		NS	NS	NS	NS	NS	NS	NS	NS
Energy × Molt		NS	NS	NS	NS	NS	NS	NS	NS
Str. × Ener. × Molt		NS	NS	NS	NS	NS	NS	NS	NS

<sup>a-c</sup>Means within a column and under each main effect with no common superscripts differ significantly

**Table 3.** Influence of dietary energy and molting method on post-molt egg component, egg solids, and egg quality in Bovans white and Dekalb white hens from 70 to 81 wk of age

Factor		% of egg component			% of egg solids			Egg quality		
		Yolk	Albumen	Shell	Whole egg	Yolk	Albumen	Egg specific gravity (unit)	Haugh unit	Yolk color
Strain	Bovans	26.57	64.31	9.10	23.01	50.46	11.67	1.0836	69.08	6.44
	Dekalb	26.20	64.31	9.51	23.50	50.31	11.72	1.0856	72.65	6.52
Dietary energy (Kcal/kg)	2767	26.73	64.02	9.27	23.43	50.34	11.70	1.0850	70.59	6.36
	2806	25.92	64.94	9.14	23.28	50.45	11.60	1.0846	71.40	6.41
	2846	26.80	64.22	9.37	23.54	50.30	11.78	1.0843	71.40	6.63
	2886	27.07	64.05	9.45	22.76	50.45	11.71	1.0845	70.05	6.52
Molt regimen	NS	26.51	64.18	9.30	23.46	50.37	11.62	1.0844	70.25	6.45
	FW	26.26	64.43	9.31	23.05	50.40	11.78	1.0848	71.47	6.50
Pooled SEM		0.35	0.40	0.18	0.77	0.15	0.14	0.0006	2.09	0.14
-----Probability-----										
Main effects and interactions										
Strain		NS	NS	0.0021	NS	NS	NS	0.0001	0.0010	NS
Dietary energy		NS	NS	NS	NS	NS	NS	NS	NS	0.0428
Molt regimen		NS	NS	NS	NS	NS	NS	NS	NS	NS
Strain × Energy		NS	NS	NS	NS	NS	NS	NS	NS	NS
Strain × Molt		NS	NS	NS	NS	NS	NS	NS	NS	NS
Energy × Molt		NS	NS	NS	NS	NS	NS	NS	NS	NS
Str. × En. × Molt		NS	NS	NS	NS	NS	NS	NS	NS	NS

<sup>a-c</sup>Means within a column and under each main effect with no common superscripts differ significantly

**Table 4.** Influence of dietary energy and poultry oil price on profits<sup>1,2</sup>

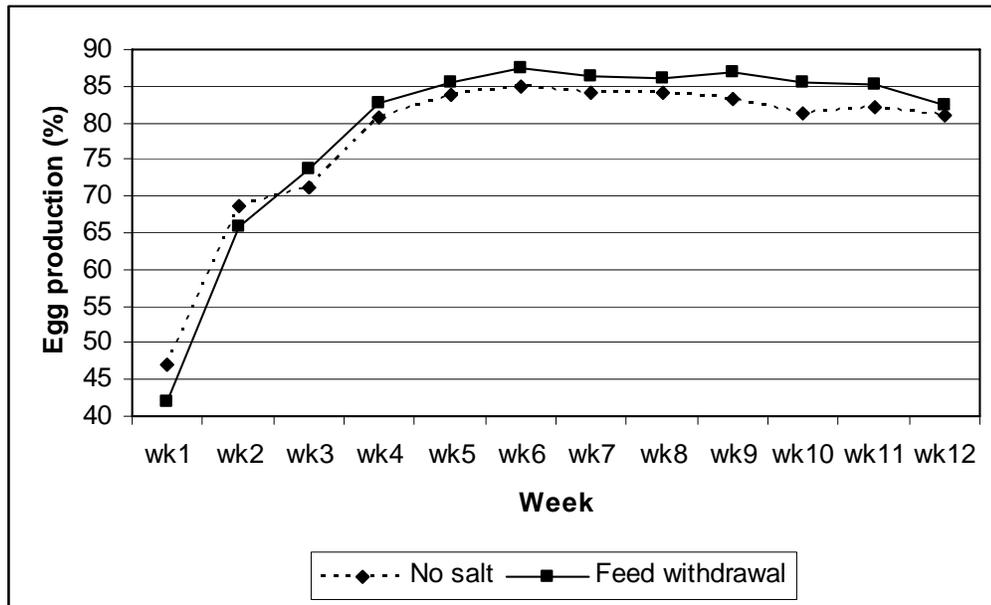
	Dietary energy content (kcal ME/kg)			
	2767	2806	2846	2886
		Returns <sup>3</sup> (\$/dozen)		
High fat price (\$0.40/kg)	0.158	0.156	0.157	0.150
Low fat price (\$0.26/kg)	0.158	0.158	0.162	0.157

<sup>1</sup> Corn price = \$0.09/kg, soy price = \$0.20/kg, CaCO<sub>3</sub> = \$0.03/kg, hard shell = \$0.03/kg, Dicalcium phosphate = \$0.27 cents/kg, salt = \$0.06/kg, Vitamin premix = \$2.67/kg, mineral premix = \$0.59/kg, DL-methionine = \$2.59/kg.

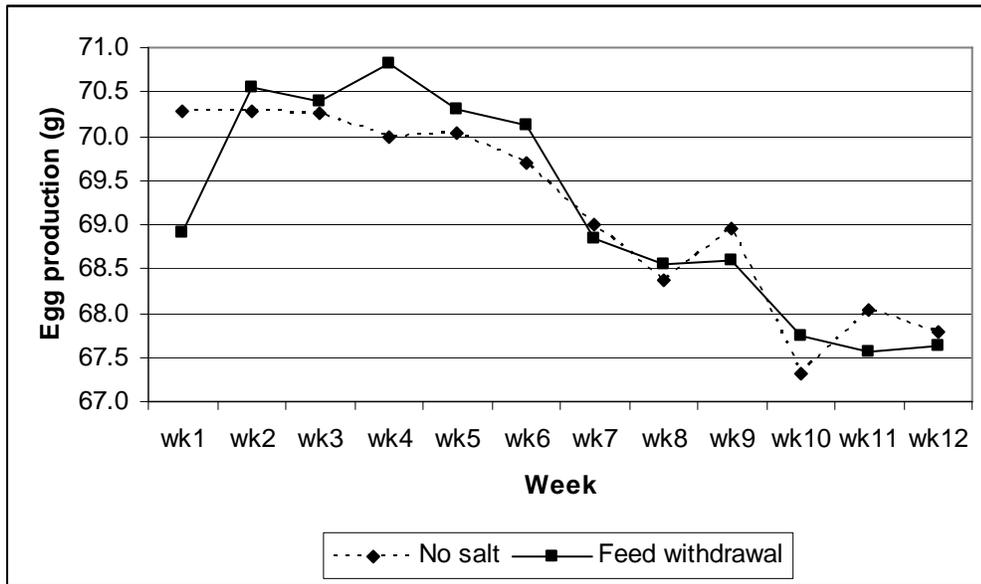
<sup>2</sup> The egg price spread between medium and large eggs was 11 cents

<sup>3</sup> Returns (R) were calculated using the equation:  $R = \text{UBEP} - \text{NR} - \text{PC} - \text{FdC}$ , where UBEP = Uner Barry g Price, NR = nest run into package product delivered, PC = production cost, and FdC = feed cost, as described by Roland et al. (1998, 2000).

**FIGURE 1.** The effect of molting method on egg production of laying hens from 70 to 81 wk of age



**FIGURE 2.** The effect of molting method on egg weight of laying hens from 70 to 81 wk of age



VI. EFFECT OF MOLTING METHOD AND DIETARY ENERGY ON  
POST-MOLT PERFORMANCE, EGG COMPOSITION, EGG SOLIDS, AND EGG  
QUALITY OF BOVANS AND DEKALB WHITE HENS  
DURING SECOND CYCLE PHASE 2 AND 3

**ABSTRACT** Two  $4 \times 2 \times 2$  factorial experiments of four dietary energy levels, two molting methods (feed withdrawal and no salt diet), and two strains (Bovans White and Dekalb White) were conducted to determine the effect of dietary energy and molting method on long-term post-molt performance of two strains of commercial Leghorns. Before molt, either Bovans hens or Dekalb hens were randomly divided into two groups. Feed was withdrawn from half of the hens (66 wk of age) for 9 days. Bovans and Dekalb hens lost 32.8% and 32.9% body weight, respectively. A molt feed was fed from day 10 to 28. The other half of hens was fed a no salt diet for 28 days. Bovans and Dekalb hens lost 16.4% and 17.8% body weight at the end of molt, respectively. In both Experiment 1 and 2, Bovans White hens (n = 576) and Dekalb White hens (n = 576) were randomly divided into 16 treatments (6 replicates of 12 birds per treatment). Experiment 1 lasted from 86 to 95 wk of age, and Experiment 2 lasted from wk 101 to 110 of age. Bovans white hens had significantly higher egg production than Dekalb white hens while Bovans white hens had significantly lower egg weight, percent of eggshell, and egg specific gravity than Dekalb white hens. Increasing dietary energy had no significant effect on all

parameters, except feed conversion, during second cycle phase 2 (86 to 95 wk of age). Increasing dietary energy linearly improved feed conversion. Increasing dietary energy had no effect on all parameters, except body weight of hens, during second cycle phase 3 (101 to 110 wk of age). Based on improved feed conversion, dietary energy of 2,846 kcal ME/kg might be enough for optimal performance during second cycle phase 2. Based on body weight of hens, dietary energy level for optimal performance should be less than 2,936 kcal ME/kg during second cycle phase 3. There can be no fixed ideal dietary energy level for optimal profits for post-molt egg production. Molting method had no effect on egg production and egg mass during the early and middle stage of post-molt production period. However, hens molted by feed withdrawal had significantly higher egg production and egg mass during latter stage of post-molt production period, compared to hens molted by no salt diet. Although hens molted by feed withdrawal had numerically higher egg specific gravity than hens molted by no salt diet, there was no significant difference in egg specific gravity due to molting method. Molting method had no significant effect on whole egg solids, albumen solid, yolk solid, Haugh unit and yolk color. If egg producers are forced to give up the conventional withdrawal molting method by animal welfare organizations, they may lose some egg production, feed conversion, and egg specific gravity. Therefore, feeding no salt diet will produce reasonable long-term post-molt performance and egg shell quality, rather than optimal performance and egg shell quality.

*Key words:* strains, dietary energy, molting method

## INTRODUCTION

Induced molting, an important management tool, has been widely used to rejuvenate laying hens for a second or third cycle of egg production by the egg industry in US for long time. Induced molting not only improved performance and egg shell quality, but also increased profits by optimizing the use of replacement pullets on commercial layer farms (Lee, 1982; Barker et al., 1983; Bell, 2003). 47% more hens would be required to keep houses full with the one-cycle egg production (Bell, 2003). The combination of feed withdrawal and light reduction is most widely used to induce molting in USA egg industry. In general, most producers today use some form of feed withdrawal for periods of 5 to 14 d (Bell and Kuney, 2004). Traditional feed removal method has received considerable attention relative to the animal welfare issues recently. The United Egg producers (UEP) Scientific Advisory Committee on Animal Welfare urged researchers and producers to work together to develop alternatives to feed withdrawal for molting (UEP, 2002). Large egg consumers such as McDonald's, Wendy's and Burger King stated that they would not purchase eggs from producers that use feed withdrawal in their molting programs (Egg industry, 2000; Smith, 2002).

Effectiveness of several nonfeed removal molting diets including low-sodium diets (Whitehead and Shannon, 1974; Begin and Johnson, 1976; Nesbeth et al., 1976a, b; Whitehead and Sharp, 1976; Monsi and Enos, 1977; Ross and Herrick, 1981; Harms 1981, 1983; Naber et al., 1984, Said et al., 1984), low-calcium diets (Gilbert et al., 1981), high-Zinc diets (Shippee et al., 1979; Berry and Brake, 1985; Bar et al., 2003), and low protein and low energy diets (Koelkebeck et al., 2001; Biggs et al., 2003) have been evaluated. Naber et al. (1984) and Said et al. (1984) reported that low sodium diets can be

effective in recycling hens for a second period of egg production. However, the low-salt diets decreased egg production and eggshell thickness in post-molt hens (Ross and Herrick, 1981). Different strains showed different performance after molt, compared to conventional feed withdrawal (Said et al., 1984; Bell and Kuney, 2004). In addition, there is little research on effect of low-salt diets on post-molt egg composition and egg solids, which are important factors influencing profits of breaker egg market. Wu et al., (2006) reported that there was no significant difference in performance, egg composition, egg solids, and egg quality between hens molted by feed withdrawal and hens molted by no salt diet during the early stage of post-molt egg production (wk 70 to 81). However, molting method may affect long-term performance, egg composition, egg solids, and egg quality. Therefore, it is necessary to have more knowledge on effect of no salt diet on long-term performance, egg quality, egg component, and egg solids in current strains of commercial Leghorns (Bovans and Dekalb) to determine acceptable alternatives for feed withdrawal method.

Feed intake (Grobas et al., 1999; Harms et al., 2000; Wu et al., 2005a, b, c) significantly decreased with increasing dietary energy or supplemental fat. However, Summers and Leeson (1993), Jalal et al. (2006), and Wu et al. (2006) reported that there was no significant effect of dietary energy on feed intake. Decreased feed intake might have a big impact on cost of production. If feed intake cannot be linearly decreased by increased energy, increasing dietary energy by the addition of fat may not be economical. In addition, egg weight increased with increasing dietary energy (Keshavarz., 1995; Keshavarz and Nakajima, 1995; Harms et al., 2000; Bohnsack et al., 2002; Sohail et al., 2003; Wu et al., 2005a). However, Wu et al. (2005c and 2006) and Jalal et al. (2006)

reported that there was no response of egg weight to increasing dietary energy by the addition of fat. Egg weight is also an important factor, which can affect profits. There is very limited literature information on effect of dietary energy on performance, egg composition, egg solids, egg quality, and profits in post-molt egg production. It is necessary to have a better understanding on how to optimize the use of dietary energy to get optimal performance and profits of post-molt laying hens.

The objective of this study is to determine the effect of molting method and dietary energy on performance, egg component, egg solids, egg quality, and profits in Bovans white and Dekalb white hens in long-term post-molt production period.

## **MATERIAL AND METHODS**

Two experiments of  $4 \times 2 \times 2$  factorial arrangement of four dietary energy levels, two molting methods (feed withdrawal and low salt diet), and two strains (Bovans White and Dekalb White) were conducted. Experiment 1 lasted from wk 86 to 95, and Experiment 2 lasted from wk 101 to 110. Ingredients and nutrient composition of experimental diets were shown in Table 1.

Before molt, either Bovans hens or Dekalb hens were randomly divided into two groups. Feed was withdrawn from half of the hens (66 wk of age) for 9 days. Bovans and Dekalb hens lost 32.8% and 32.9% body weight, respectively. A molt feed was fed from day 10 to 28 (Table 1). The other half of hens was fed a low salt diet for 28 days (Table 1). Bovans and Dekalb hens lost 16.4% and 17.8% body weight at the end of molt, respectively. Hens molted by feed withdrawal stopped laying by day 4, while hens molted by no salt diet did not stop laying until day 17. In both experiments, Bovans White hens ( $n = 576$ ) and Dekalb White hens ( $n = 576$ ) were randomly divided into 16

treatments (6 replicates of 12 birds per treatment). Replicates were equally distributed into upper and lower cages to minimize cage level effect. Three hens were housed in a  $40.6 \times 45.7 \text{ cm}^2$  cage and five adjoining cages consisted of a replicate. All hens were housed in an environmentally-controlled house with temperature maintained at approximately  $25.6^\circ\text{C}$ . The light period was reduced from 16 hr to 8 hr daily from day 1. On the 29<sup>th</sup> day, the light period was returned to 16 hr daily. All hens were supplied with feed and water *ad libitum*. Egg production was recorded daily, feed consumption was recorded weekly, and egg weight was recorded biweekly, and egg specific gravity was recorded monthly. Egg weight and egg specific gravity were measured using all eggs produced during two consecutive days. Egg specific gravity was determined using 11 gradient saline solutions varying in specific gravity from 1.060 to 1.100 with 0.005-unit increments (Holder and Bradford, 1979). Mortality was determined daily and the feed consumption was adjusted accordingly. Body weight was obtained by weighing 3 hens per group at the end of the experiment. Egg mass and feed conversion (g feed/g egg) were calculated from egg production, egg weight and feed consumption.

Egg components were measured using two eggs from each replicate in the middle and the end of experiment. Two eggs from each replicate were collected to measure whole egg solids in the middle and the end of experiment. Albumen and yolk solid were measured by using two eggs from each replicate in the middle and the end of experiment. The procedures for measuring egg components, whole egg solids, and albumen and yolk solid were the same as those of Wu et al. (2005). Yolk color and Haugh unit were measured (3 eggs of each replicate) at the end of experiment by egg multi-tester EMT-5200 (Robotmation, Co., Ltd, Tokyo, Japan).

Data were analyzed by proc mixed procedures of Statistical Analysis System (SAS Institute, 2000) for a randomized complete block with a factorial treatment design. Dietary energy, molting method, and strain were fixed, while blocks were random. The factorial treatment arrangement consisted of 4 dietary energy levels, 2 molting methods, and 2 layer strains. The following model used to analyze data was as follows:

$$Y_{ijkl} = \mu + \alpha_i + \beta_j + \gamma_k + (\alpha\beta)_{ij} + (\beta\gamma)_{jk} + (\alpha\gamma)_{ik} + (\alpha\beta\gamma)_{ijk} + P_l + \epsilon_{ijkl}$$

Where  $Y_{ijkl}$  = individual observation,  $\mu$  = experimental mean,  $\alpha_i$  = dietary energy effect,  $\beta_j$  = molting method,  $\gamma_k$  = layer strain effect,  $(\alpha\beta)_{ij}$  = interaction between dietary energy and molting method,  $(\beta\gamma)_{jk}$  = interaction between molting method and strain,  $(\alpha\gamma)_{ik}$  = interaction between dietary energy and strain,  $(\alpha\beta\gamma)_{ijk}$  = interaction among dietary energy, molting method, and strain,  $P_k$  = effect of block,  $\epsilon_{ijkl}$  = error component.

If differences in treatment means were detected by ANOVA, Duncan's Multiple Range Test was applied to separate means. A significance level of  $P \leq 0.05$  was used during analysis.

## **RESULTS AND DISCUSSION**

There were no interactions among strain, dietary energy, and molting method in all parameters during second cycle phase 2 (86 to 95 wk of age) and phase 3 (101 to 110 wk of age) (Table 2, 3, 4, and 5). Bovans hens had significantly greater egg production than Dekalb hens during second cycle phase 2 and 3 (Table 2 and 4). Egg mass and feed conversion of Bovans hens were significantly better than Dekalb hens during second cycle phase 2. Bovans hens used significantly less dietary energy to produce 1 g egg, compared to Dekalb hens. Dekalb hens had significantly higher egg weight during second

cycle phase 2, and numerically higher egg weight during second cycle phase 3 ( $P \leq 0.089$ ) than Bovans hens. Dekalb hens had significantly higher percent of eggshell and egg specific gravity than Bovans hens during second cycle phase 2 and 3 (Table 3 and 5). These results were in agreement with those Wu et al. (2005a and b), who reported Bovans hens had significantly higher egg production, egg mass, and lower egg weight than Dekalb hens. There was no significant difference in percent of yolk and albumen, percent of whole solid, Haugh unit and yolk color between Bovans hens and Dekalb hens during second cycle phase 2 and 3 (Table 3 and 5). Breaker egg industry may get more total egg solids in Bovans hens because of higher egg yield of Bovans hens.

Increasing dietary energy by the addition of poultry oil had no significant effect on feed intake during second cycle phase 2 (wk 86 to 95 of age) and phase 3 (101 to 110 wk of age) (Table 2 and 4). Similarly, Wu et al. (2006) reported that there was no effect of dietary energy on feed intake during second cycle phase 1 (70 to 81 wk of age). However, these results were inconsistent with that of Wu et al. (2005a), who reported that feed intake linearly decreased as dietary energy increased. This might be due to the smaller gap between two levels of dietary energy (approximately 40 and 46 kcal ME/kg in second cycle phase 2 and 3, respectively), compared to that (approximately 80 kcal ME/kg) of Wu et al. (2005a). Dietary energy had no effect on egg production, egg mass, and mortality in second cycle phase 2 and 3 (Table 2 and 4). These results were in agreement with those of Wu et al. (2005a and c; 2006) and Jalal et al. (2006), who reported that there was no significant effect of dietary energy level on egg production.

Dietary energy had no significant effect on egg weight during second cycle phase 2 (86 to 95 wk of age) and phase 3 (101 to 110 wk of age) (Table 2 and 4). Similarly, Wu

et al. (2005c; 2006) reported that there was no significant effect of egg weight to increasing dietary energy during first cycle phase 2 (40 to 51 wk of age) and second cycle phase 1 (70 to 81 wk of age), respectively. However, Wu et al. (2005a) reported that egg weight linearly increased with increasing dietary energy from 21 to 36 wk of age, and suggested that the increase of egg weight was mainly due to increased yolk weight for young hens. There was no response of percent of yolk to increasing dietary energy during second cycle phase 2 and 3 (Table 3 and 5). Young hens might not have enough ability to produce enough lipoprotein and need more exogenous fat to supply lipids for egg yolk development in the early stage of egg production (Shell et al., 1987). This might explain why there was no response of egg weight to increasing dietary energy for hens after first cycle phase 1.

Increasing dietary energy by the addition of poultry oil had a linear effect on feed conversion during in second cycle phase 2 (wk 86 to 95) (Table 2). Increasing dietary energy from 2,767 to 2,846 kcal ME/kg improved feed conversion from 1.97 to 1.88, resulting in a 4.6% improvement in feed conversion. Further increasing dietary energy from 2,846 to 2,886 kcal ME/kg had no improvement in feed conversion. This suggested dietary energy of 2,846 kcal ME/kg might be enough for optimal performance during second cycle phase 2 (86 to 95 wk of age). Similarly, Wu et al. (2005) reported that hens fed the diet containing dietary energy of 2,877 kcal ME/kg obtained optimal performance. Dietary energy had no significant effect on feed conversion in second cycle phase 3 (101 to 110 wk of age) (Table 4). Hens fed the diet containing 2,936 kcal ME/kg had significantly higher body weight than hens fed the diets with other dietary energy levels. This suggested that dietary energy level of 2,936 kcal ME/kg might provide more

dietary energy than the optimal energy level for performance. Because egg mass (approximately 48 g per hen daily) of hens during second cycle phase 3 (101 to 110 wk of age) were less than that (approximately 52 g per hen daily) of hens during second cycle phase 2 (86 to 95 wk of age), less energy were used to produce egg so that excess energy was used to produce body fat. This might explain body weight of hens fed 2,936 kcal ME/kg was significantly higher than that of hens fed other dietary levels. The dietary energy level for optimal performance should be less than 2936 kcal ME/kg for hens during second cycle phase 3 (101 to 110 wk of age).

The Economic Feeding and Management Program developed by Roland et al. (1998, 2000) was used to calculate profits of different dietary energy levels at poultry oil prices. Maximum profits per dozen eggs were obtained in hens fed the diet containing 2,806 kcal ME/kg of dietary energy in both poultry oil prices during second cycle phase 2 (86 to 95 wk of age) (Table 6). When poultry oil price were \$0.26/kg, maximum profits per dozen eggs were obtained in hens fed the diet containing 2,890 kcal ME/kg of dietary energy during second cycle phase 3 (101 to 110 wk of age) (Table 7). However, when poultry oil price increased to \$0.40/kg, maximum profits was obtained in hens fed the diet containing 2,797 kcal ME/kg of dietary energy during second cycle phase 3. Because feed ingredient prices and egg price often vary, there can be no fixed ideal dietary energy level for optimal profits for post-molt hens.

Although there was no linear response of feed intake to increasing dietary energy, hens did adjust feed intake to achieve a constant energy intake so that the similar quantities of dietary energy (5.3 to 5.5 kcal in second cycle phase 2 and 6.1 to 6.4 kcal in second cycle phase 3, respectively) were used to produce to 1 g egg (Table 2 and 4).

Increasing dietary energy had no effect on percent of percent of yolk, albumen, and shell, percent of whole egg solids, yolk solid, and albumen solid, egg specific gravity, Haugh unit, and yolk color in second cycle phase 2 (86 to 95 wk of age) and phase 3 (wk 101 to 110 of age). Similarly, Wu et al. (2006) reported there was no significant effect of dietary energy on egg composition, egg solids, and egg quality during second cycle phase 1 (70 to 81 wk of age).

There was no significant difference in feed intake and egg weight between hens molted by feed withdrawal and hens molted by low salt diet during second cycle phase 2 (wk 86 to 95 of age) and phase 3 (101 to 110 wk of age) (Table 2 and 4). Similarly, Wu et al. (2006) reported that molting method had no significant effect on feed intake and egg weight during second cycle phase 1 (70 to 81 wk of age). These results were consistent with Ross and Herrick (1984), Said et al. (1983), and Naber et al. (1984), who reported that there was no significant difference in feed intake and egg weight, due to molting method. Hens molted with feed withdrawal had numerically higher egg production and egg mass than hens molted by no salt diet during second cycle phase 2 ( $P \leq 0.0782$  and  $P \leq 0.0873$ , respectively) (86 to 95 wk of age). Hens molted by feed withdrawal had significantly higher egg production and egg mass than hens molted by no salt diet during second cycle phase 3 (101 to 110 wk of age). Wu et al. (2006) reported that there was no significant difference in egg production and egg mass during second cycle phase 1 (70-81 wk of age), due to molting method. These results and Wu et al. (2006) suggested that molting method had no effect on egg production and egg mass during the early and middle stage of post-molt egg production. However, hens molted by feed withdrawal had higher egg production and egg mass during later stage of post-molt

egg production, compared to hens molted by no salt diet. Similarly, difference in egg production between hens molted by no salt diet or low nutrient diet and hens molted by feed withdrawal increased with age (Ross and Herrick, 1979; Biggs, 2003). This might be due to different body weight loss of hens molted by different molting methods, which leads to different ovarian regression. Hens molted by feed withdrawal lost 32.9% body weight, while hens molted by no salt diet lost 17.1% body weight during molting. Barker et al. (1983) reported that a body weight loss of approximately 27 to 31% obtained optimal post-molt performance.

There was no significant difference in feed conversion and energy intake during second cycle phase 2 (86 to 95 wk of age) (Table 2). Hens molted by feed withdrawal had significantly better feed conversion and used significantly less dietary energy to produce egg than hens molted by no salt diet during second cycle phase 3 (101 to 110 wk of age) (Table 4). The improved feed efficiency of hens molted by feed withdrawal might be due to higher egg production of hens molted by feed withdrawal, compared to that of hens molted by no salt diet. Hens molted by feed withdrawal had significantly higher percent of yolk and less percent of albumen than hens molted by hens molted by no salt diet during second cycle phase 2 (86 to 95 wk of age). The effect of molting method on egg specific gravity approached significance ( $P \leq 0.0563$ ) during second cycle phase 2. There was no significant difference in egg specific gravity during second cycle phase 3. Wu et al. (2005) reported there was no significant difference in egg specific gravity during second cycle phase 1. Similar results were reported by Ross and Herrick (1979), Naber et al. (1984), and Biggs et al. (2003). Based on the results of this study and Ross and Herrick (1979), Naber et al. (1984), and Biggs et al. (2003), it was concluded that

although hens molted by feed withdrawal had numerically higher egg specific gravity than hens molted by no salt diet, there was no significant difference in egg specific gravity due to molting method. There was no significant difference in percent of shell, percent of whole egg solids, yolk solid, and albumen solid, Haugh unit, and yolk color between hens molted by feed withdrawal and hens molted by no salt diet during second cycle phase 2 and 3 (Table 3 and 4). Similarly, Wu et al. (2006) reported that molting method had no effect on egg solids, Haugh unit, and yolk color. These results were in agreement with those of Ross and Herrick (1979), Naber et al. (1984), and Biggs et al. (2003), who reported that molting method had no effect on Haugh unit. Therefore, molting method had no significant effect on egg solids and egg quality during post-molt egg production.

In conclusion, Bovans white hens had significantly higher egg production than Dekalb white hens while Bovans white hens had significantly lower egg weight, percent of eggshell, and egg specific gravity than Dekalb white hens. Increasing dietary energy had no significant effect on all parameters, except feed conversion, during second cycle phase 2 (86 to 95 wk of age). Increasing dietary energy linearly improved feed conversion. Increasing dietary energy had no effect all parameters, except body weight of hens, during second cycle phase 3 (101 to 110 wk of age). Based on improved feed conversion, dietary energy of 2,846 kcal ME/kg might be enough for optimal performance during second cycle phase 2 (86 to 95 wk of age). Based on body weight of hens, dietary energy level for optimal performance should be less than 2,936 kcal ME/kg during second cycle phase 3 (101 to 110 wk of age). There can be no fixed ideal dietary energy level for optimal profits for post-molt egg production. Molting method had no effect on egg

production and egg mass during the early and middle stage of post-molt production period. However, hens molted by feed withdrawal had significantly higher egg production and egg mass during latter stage of post-molt production period, compared to hens molted by no salt diet. Although hens molted by feed withdrawal had numerically higher egg specific gravity than hens molted by no salt diet, there was no significant difference in egg specific gravity due to molting method. Molting method had no significant effect on whole egg solids, albumen solid, yolk solid, Haugh unit and yolk color. If egg producers are forced to give up the conventional withdrawal molting method by animal welfare organizations, they may lose some egg production, feed conversion, and egg specific gravity. Therefore, feeding no salt diet will produce reasonable long-term post-molt performance and eggshell quality, rather than optimal performance and egg shell quality.

#### **ACKNOWLEDGMENTS**

The authors thank Centurion Poultry, Inc, Lexington, GA, and Ridley Inc., Mankato, MN, for funding support of this research.

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**Table 1.** Ingredients and nutrient composition of experimental diets

Ingredient	No salt diet	Molt diet following g feed withdrawal	Post-molt diets							
			Experiment 1 (wk 86 to 95)				Experiment 2 (wk 101 to 110)			
			Diet 1	Diet 2	Diet 3	Diet 4	Diet 1	Diet 2	Diet 3	Diet 4
Corn (%)	73.12	72.78	65.41	64.50	63.57	62.65	67.62	66.52	65.43	64.34
Soybean meal (%)	20.02	20.51	22.43	22.51	22.58	22.66	20.59	20.68	20.77	20.85
CaCO <sub>3</sub> (%)	0.00	4.02	5.83	5.83	5.82	5.82	5.70	5.69	5.69	5.69
Hardshell <sup>1</sup> (%)	4.13	0.00	4.00	4.00	4.00	4.00	4.00	4.00	4.00	4.00
Dical. phosphate (%)	1.39	1.38	1.40	1.40	1.41	1.41	1.19	1.19	1.20	1.20
Poultry oil (%)	0.25	0.25	0.00	0.84	1.68	2.53	0.00	1.00	2.00	3.00
NaCl (%)	0.00	0.38	0.36	0.36	0.36	0.36	0.36	0.36	0.36	0.36
Vitamin Premix <sup>2</sup> (%)	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
Mineral premix <sup>3</sup> (%)	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
DL-Methionine (%)	0.19	0.05	0.07	0.07	0.07	0.07	0.05	0.05	0.05	0.05
Calculated analysis										
Crude protein (%)	15.90	15.77	16.04	16.00	15.97	15.93	15.32	15.28	15.23	15.19
ME (kcal/kg)	2989	2996	2767	2806	2846	2885	2797	2843	2890	2938
Ca (%)	2.00	2.00	4.25	4.25	4.25	4.25	4.15	4.15	4.15	4.15
A. P. (%)	0.36	0.36	0.36	0.36	0.36	0.36	0.32	0.32	0.32	0.32
Sodium	0.02	0.18	0.17	0.17	0.17	0.17	0.17	0.17	0.17	0.17
Methionine (%)	0.47	0.44	0.33	0.33	0.33	0.33	0.31	0.31	0.31	0.31
Met+Cys (%)	0.73	0.73	0.62	0.62	0.62	0.62	0.58	0.58	0.58	0.58
Lysine (%)	0.80	0.79	0.83	0.83	0.83	0.83	0.78	0.78	0.78	0.78

<sup>1</sup>Hardshell = large particle (passing US mesh #4 and retained by US mesh #6) CaCO<sub>3</sub> supplied by Franklin Industrial Minerals, Lowell, Florida.

<sup>2</sup>Provided per kilogram of diet: vitamin A (as retinyl acetate), 8,000 IU; cholecalciferol, 2,200 ICU; vitamin E (as DL- $\alpha$ -tocopheryl acetate), 8 IU; vitamin B<sub>12</sub>, 0.02 mg; riboflavin, 5.5 mg; D-calcium pantothenic acid, 13 mg; niacin, 36 mg; holine, 500 mg; folic acid, 0.5 mg; vitamin B<sub>1</sub> (thiamin mononitrate), 1 mg; pyridoxine, 2.2 mg; biotin, 0.05 mg; vitamin K (menadione sodium bisulfate complex), 2 mg.

<sup>3</sup>Provided per kilogram of diet: manganese, 65 mg; iodine, 1 mg; iron, 55 mg; copper, 6 mg; zinc, 55 mg; selenium, 0.3 mg.

**Table 2.** Influence of dietary energy and molting method on post-molt performance of Bovans White and Dekalb White hens during second cycle phase 2 (86 to 95 wk of age) (Experiment 1)

Factor		Feed intake (g/hen per day)	Egg production (%)	Egg weight (g)	Egg mass (g egg/hen per d)	Feed conversion (g of feed/of g egg)	Energy intake (Kcal ME/g egg)	Body weight (kg)	Mortality (%)
Strain	Bovans	96.80	78.31 <sup>a</sup>	66.10 <sup>b</sup>	51.76 <sup>a</sup>	1.87 <sup>b</sup>	5.22 <sup>b</sup>	1.75	2.01
	Dekalb	96.83	74.46 <sup>b</sup>	67.28 <sup>a</sup>	50.10 <sup>b</sup>	1.94 <sup>a</sup>	5.54 <sup>a</sup>	1.73	3.09
Dietary energy (Kcal/kg)	2767	98.79	75.50	66.62	50.31	1.97 <sup>a</sup>	5.51	1.73	3.93
	2806	96.16	76.12	66.68	50.73	1.90 <sup>ab</sup>	5.33	1.71	2.78
	2846	95.85	76.62	66.57	50.99	1.88 <sup>b</sup>	5.36	1.76	2.50
	2886	96.46	77.28	66.88	51.68	1.87 <sup>b</sup>	5.31	1.78	1.00
Molt regimen	NS	95.65	75.36	66.86	50.25	1.91	5.39	1.74	1.87
	FW	97.99	77.40	66.52	51.61	1.90	5.38	1.74	3.23
Pooled SEM		2.45	2.29	0.69	1.54	0.05	0.14	0.15	2.00
-----Probability-----									
Main effects and interactions									
Strain		NS	0.0012	0.0010	0.0386	0.0223	0.0001	NS	NS
Dietary energy		NS	NS	NS	NS	0.0497	NS	NS	NS
Molt method		0.0625	0.0782	NS	0.0873	NS	NS	NS	NS
Strain × Energy		NS	NS	NS	NS	NS	NS	NS	NS
Strain × Molt		NS	NS	NS	NS	NS	NS	NS	NS
Energy × Molt		NS	NS	NS	NS	NS	NS	NS	NS
Str. × Ener. × Molt		NS	NS	NS	NS	NS	NS	NS	NS

<sup>a-c</sup>Means within a column and under each main effect with no common superscripts differ significantly

**Table 3.** Influence of dietary energy and molting method on egg component, egg solids, and egg quality in post-molt Bovans White and Dekalb White hens during second cycle phase 2 (86 to 95 wk of age) (Experiment 1)

Factor		% of egg component			% of egg solids			Egg quality		
		Yolk	Albumen	Shell	Whole egg	Yolk	Albumen	Egg specific gravity (unit)	Haugh unit	Yolk color
Strain	Bovans	26.12	65.01	8.81 <sup>b</sup>	22.88	50.85	11.42	1.0807 <sup>b</sup>	72.55	5.98
	Dekalb	25.69	65.15	9.17 <sup>a</sup>	22.84	50.58	11.59	1.0831 <sup>a</sup>	72.60	5.94
Dietary energy (Kcal/kg)	2767	25.83	65.16	8.88	22.92	50.81	11.37	1.0824	73.37	6.08
	2806	25.89	65.18	8.93	22.48	51.01	11.49	1.0820	71.20	5.87
	2846	25.75	65.19	9.06	22.72	50.62	11.58	1.0821	73.23	5.93
	2886	26.15	64.78	9.08	23.32	50.41	11.60	1.0810	72.50	5.95
Molt regimen	NS	25.47 <sup>b</sup>	65.60 <sup>a</sup>	8.93	22.79	50.72	11.54	1.0815	72.80	6.01
	FW	26.34 <sup>a</sup>	64.56 <sup>b</sup>	9.05	22.93	50.71	11.48	1.0823	72.35	5.90
Pooled SEM		0.80	0.93	0.25	0.38	0.24	0.37	0.0008	2.25	0.15
-----Probability-----										
Main effects and interactions										
Strain		NS	NS	0.0057	NS	NS	NS	0.0001	NS	NS
Dietary energy		NS	NS	NS	NS	NS	NS	0.0788	NS	NS
Molt method		0.0330	0.0280	NS	NS	NS	NS	0.0563	NS	NS
Strain × Energy		NS	NS	NS	NS	NS	NS	NS	NS	NS
Strain × Molt		NS	NS	NS	NS	NS	NS	NS	NS	NS
Energy × Molt		NS	NS	NS	NS	NS	NS	NS	NS	NS
Str. × Ener. × Molt		NS	NS	NS	NS	NS	NS	NS	NS	NS

<sup>a-c</sup>Means within a column and under each main effect with no common superscripts differ significantly

**Table 4.** Influence of dietary energy and molting method on post-molt performance of Bovans White and Dekalb White hens during second cycle phase 2 (101 to 110 wk of age) (Experiment 2)

Factor		Feed intake (g/hen per day)	Egg production (%)	Egg weight (g)	Egg mass (g egg/hen per d)	Feed conversion (g of feed/of g egg)	Energy intake (Kcal ME/g egg)	Body weight (kg)	Mortality (%)
Strain	Bovans	103.78	72.91	66.48	48.47	2.15	6.07 <sup>b</sup>	1.77	1.93
	Dekalb	102.54	70.01	67.09	46.95	2.19	6.38 <sup>a</sup>	1.73	2.53
Dietary energy (Kcal/kg)	2797	103.84	70.56	66.26	46.75	2.23	6.35	1.71 <sup>a</sup>	2.51
	2844	103.20	70.70	66.43	46.94	2.21	6.28	1.72 <sup>a</sup>	2.54
	2890	103.61	72.75	67.19	48.87	2.13	6.15	1.73 <sup>a</sup>	1.74
	2936	102.01	71.83	67.25	48.29	2.12	6.12	1.83 <sup>b</sup>	2.12
Molt regimen	NS	102.57	69.79 <sup>b</sup>	66.95	46.65 <sup>b</sup>	2.21	6.34 <sup>a</sup>	1.74	2.45
	FW	103.76	73.13 <sup>a</sup>	66.62	48.78 <sup>a</sup>	2.13	6.11 <sup>b</sup>	1.75	2.01
Pooled SEM		1.87	2.80	0.71	1.98	0.08	0.22	0.14	1.74
-----Probability-----									
Main effects and interactions									
Strain		NS	0.0414	0.0897	NS	NS	0.0064	NS	NS
Dietary energy		NS	NS	NS	NS	0.0949	NS	0.0380	NS
Molt method		NS	0.0197	NS	0.0396	0.0483	0.0462	NS	NS
Strain × Energy		NS	NS	NS	NS	NS	NS	NS	NS
Strain × Molt		NS	NS	NS	NS	NS	NS	NS	NS
Energy × Molt		NS	NS	NS	NS	NS	NS	NS	NS
Str. × Ener. × Molt		NS	NS	NS	NS	NS	NS	NS	NS

<sup>a-c</sup>Means within a column and under each main effect with no common superscripts differ significantly

**Table 5.** Influence of dietary energy and molting method on egg component, egg solids, and egg quality in post-molt Bovans White and Dekalb White hens during second cycle phase 2 (101 to 110 wk of age) (Experiment 2)

Factor		% of egg component			% of egg solids			Egg quality		
		Yolk	Albumen	Shell	Whole egg	Yolk	Albumen	Egg specific gravity (unit)	Haugh unit	Yolk color
Strain	Bovans	28.10	63.28	8.62	24.17	50.35	11.95	1.0789 <sup>b</sup>	68.14	6.34
	Dekalb	27.94	62.84	9.22	23.82	50.19	12.08	1.0816 <sup>a</sup>	69.61	6.55
Dietary energy (Kcal/kg)	2797	27.66	63.24	9.10	23.98	49.88	12.02	1.0809	69.02	6.38
	2844	28.36	62.68	8.96	24.00	50.57	11.94	1.0802	68.48	6.41
	2890	28.03	63.30	8.68	23.98	50.30	12.14	1.0797	70.98	6.57
	2936	28.03	63.01	8.97	24.01	50.30	11.98	1.0802	67.02	6.41
Molt regimen	NS	27.81	63.44	8.75	23.92	50.34	11.92	1.0800	69.03	6.45
	FW	28.22	62.69	9.09	24.07	50.20	12.11	1.0804	68.72	6.44
Pooled SEM		0.71	0.82	0.38	0.52	0.38	0.24	0.0009	2.25	0.15
-----Probability-----										
Main effects and interactions										
Strain		NS	NS	0.0024	NS	NS	NS	0.0001	NS	NS
Dietary energy		NS	NS	NS	NS	NS	NS	NS	NS	NS
Molt method		NS	0.0691	0.0734	NS	NS	NS	NS	NS	NS
Strain × Energy		NS	NS	NS	NS	NS	NS	NS	NS	NS
Strain × Molt		NS	NS	NS	NS	NS	NS	NS	NS	NS
Energy × Molt		NS	NS	NS	NS	NS	NS	NS	NS	NS
Str. × Ener. × Molt		NS	NS	NS	NS	NS	NS	NS	NS	NS

<sup>a-c</sup>Means within a column and under each main effect with no common superscripts differ significantly

**Table 6.** Influence of dietary energy and poultry oil price on profits<sup>1,2</sup> in Experiment 1 (86 to 95 wk of age)

	Dietary energy content (kcal ME/kg)			
	2767	2806	2846	2886
	Returns <sup>3</sup> (cents/dozen)			
High fat price (\$0.40/kg)	0.196	0.198	0.195	0.193
Low fat price (\$0.26/kg)	0.196	0.200	0.198	0.198

<sup>1</sup> Corn price = \$0.09/kg, soy price = \$0.19/kg, CaCO<sub>3</sub> = \$0.03/kg, hard shell = \$0.03/kg, Dicalcium phosphate = \$0.27 cents/kg, salt = \$0.06/kg, Vitamin premix = \$2.67/kg, mineral premix = \$0.59/kg, DL-methionine = \$2.59/kg.

<sup>2</sup> The egg price spread between medium and large eggs was 11 cents

<sup>3</sup> Returns (R) were calculated using the equation:  $R = \text{UBEP} - \text{NR} - \text{PC} - \text{FdC}$ , where UBEP = Uner Barry Egg Price, NR = nest run into package product delivered, PC = production cost, and FdC = feed cost, as described by Roland et al. (1998, 2000).

**Table 7.** Influence of dietary energy and poultry oil price on profits<sup>1,2</sup> in Experiment 2 (101 to 110 wk of age)

	Dietary energy content (kcal ME/kg)			
	2797	2844	2890	2936
	Returns <sup>3</sup> (cents/dozen)			
High fat price (\$0.40/kg)	0.143	0.140	0.142	0.137
Low fat price (\$0.26/kg)	0.143	0.142	0.146	0.144

<sup>1</sup> Corn price = \$0.09/kg, soy price = \$0.24/kg, CaCO<sub>3</sub> = \$0.03/kg, hard shell = \$0.03/kg, Dicalcium phosphate = \$0.27 cents/kg, salt = \$0.06/kg, Vitamin premix = \$2.67/kg, mineral premix = \$0.59/kg, DL-methionine = \$2.59/kg.

<sup>2</sup> The egg price spread between medium and large eggs was 11 cents

<sup>3</sup> Returns (R) were calculated using the equation:  $R = \text{UBEP} - \text{NR} - \text{PC} - \text{FdC}$ , where UBEP = Uner Barry Egg Price, NR = nest run into package product delivered, PC = production cost, and FdC = feed cost, as described by Roland et al. (1998, 2000).

VII. EFFECTS OF ADDED SYNTHETIC LYSINE ON SECOND CYCLE  
COMMERCIAL LEGHORNS WHILE MAINTAINING THE  
METHIONINE+CYSTEINE/LYSINE RATIO AT 0.75

**ABSTRACT** Two experiments were conducted to determine the effect 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, three protein levels (16.0, 14.9, and 14.4%) and four added synthetic lysine levels (0.000, 0.030, 0.059, and 0.088%) in a 3 × 4 factorial arrangement were used. Hy-line W-36 laying hens (n = 1440, 85 wk old) were fed for 8 weeks. In Experiment 2, three protein levels (14.9, 14.4, and 14.0%) and three added synthetic lysine levels (0.000, 0.030, and 0.059%) in a 3 × 3 factorial arrangement were used. Hy-line W-36 laying hens (n = 1080; 98 wk old) were fed for 12 weeks. The results of Experiment 1 showed that there was no significant effect of adding synthetic lysine while maintaining a 0.75 Met+Cys/Lys ratio on performance of laying hens. The results of Experiment 2 showed that the addition of synthetic lysine while maintaining a 0.75 Met+Cys/Lys ratio had significant effects on feed intake, egg production, egg mass, 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/hen per d) or lysine intake is less than 720 mg/hen per 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

## INTRODUCTION

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 (Harms et al., 1993). 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 (Barker, 2003; Vieira et al., 2004). 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/hen per d (Jensen et al., 1974; Latshaw, 1976; Nathanael and Sell, 1980; NRC, 1994; and Shutte and Swink, 1998). Some studies (Bateman et al., 2000; Yadalam et al., 2000; Yadalam, 2001) 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 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. (2003) and Liu et al. (2004) 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. (2005) 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 this study was to determine the effect 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**

Two experiments were conducted in which the diets were formulated based on lysine instead of protein, and Met + Cys/Lys ratio was maintained at 0.75. In Experiment 1, three protein levels (16.0, 14.9, and 14.4%) and four added synthetic lysine levels (0.000, 0.030, 0.059, and 0.088%) in a 3 × 4 factorial arrangement were used (Table 1). The diets were fed for eight weeks. In Experiment 2, three protein levels (14.9, 14.4, and 14.0%) and three added synthetic lysine levels (0.000, 0.030, and 0.059%) in a 3×3 factorial arrangement were used (Table 2). The diets were fed for 12 weeks. Feed

samples of two experiments were analyzed for amino acids<sup>1</sup>.

In Experiment 1, Hy-line W-36 hens (n = 1440) in the second cycle (85 wk old) were randomly assigned 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 wk old) were randomly assigned 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 × 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 biweekly. Egg weight and egg specific gravity were measured using all eggs produced during two consecutive days. Egg specific gravity was determined using 11 gradient saline solutions varying in specific gravity from 1.060 to 1.100 with 0.005-unit increments (Holder and Bradford, 1979). Mortality was determined daily and egg production and feed consumption were adjusted to a hen-day basis. Body weight was obtained by weighing 3 hens per group at the end of the experiment.

Data was analyzed as a factorial arrangement of treatment using the General Linear Models (GLM) in SAS/STAT (SAS Institute, 2000). If differences in treatment means were detected by ANOVA, Duncan's Multiple Range Test was applied to separate means. Statements of statistical significance are based on a probability of ( $P \leq 0.05$ ).

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<sup>1</sup> Degussa Corporation, Allendale, NJ 07401

Contrast statements were utilized to test for added lysine linear or quadratic effects.

## **RESULTS**

### ***Experiment 1***

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. There were significant protein effects on feed intake, egg weight, egg mass, egg specific gravity, and body weight. Feed consumption of hens fed the diet containing 16.0% protein was similar to that of hens fed the diet containing 14.9% protein, but was significantly higher than that of hens fed the diet containing 14.4% protein. Egg mass and egg weight of hens fed the diet containing 14.9% protein were similar to those of hens fed the diet containing 14.4% protein, and both were significantly lower than those of the diet containing 16.0% protein. Hens fed the diet containing 14.4% protein had significant higher egg specific gravity and lower body weight than hens fed the diets containing 14.9% and 16.0% protein. Adding synthetic lysine while maintaining a 0.75 Met+Cys/Lys ratio had no significant effect on feed intake, egg production, egg weight, egg mass, feed conversion, egg specific gravity, final body weight, and mortality of laying hens.

### ***Experiment 2***

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, and both were significantly higher than that of hens fed the diets without added synthetic lysine.

Protein effect on egg weight and egg mass was approaching significant ( $P < 0.1$ ) (Table 4). Adding synthetic lysine while maintaining a 0.75 Met+Cys/Lys ratio showed a significant linear effect on egg production and egg mass, but had no significant effect on egg weight. There was no interaction between protein and added synthetic lysine on egg production, egg weight, and egg mass.

There was no significant protein effect on feed conversion (Table 4). Adding synthetic lysine while maintaining a 0.75 Met+Cys/Lys ratio had a linear effect on feed conversion. As dietary lysine increased, feed conversion linearly decreased from 2.16 to 2.06. No interaction between protein and added synthetic lysine on feed conversion were observed

There was no interaction between protein and added synthetic lysine on egg specific gravity, body weight or mortality (Table 4). There was no significant protein effect on specific gravity and mortality. Egg specific gravity of hens fed the diets supplemented with 0.059% lysine was similar to that of hens fed the diets supplemented with 0.030% lysine, and both were significantly lower than that of hens fed the diets without added lysine. Total mortality was 1.9% (20 hens out of 1,080), and mortality was not significantly affected by protein or added synthetic lysine.

## **DISCUSSION**

In Experiment 1, the addition of synthetic lysine while maintaining a 0.75 Met+Cys/Lys ratio had no effect on feed intake, egg production, egg weight, egg mass, feed conversion, egg specific gravity, final body weight, and mortality of laying hens

(Table 3). This result was in agreement with those of Sohail et al. (2003) and Liu et al. (2004), who observed that adding synthetic lysine while maintaining an optimal Met+Cys/Lys ratio had no influence on performance of laying hens. Lysine intake of hens fed the 16.0, 14.9, and 14.4% protein diets without added synthetic lysine were 849, 767, and 730 mg/h/d, respectively, which were higher than NRC (1994) recommended lysine requirements (690 mg/hen per d) (Table 5). Calculation of data from Liu et al. (2004) showed that lysine intakes of hens fed the 17.5, 16.2, and 15.2% protein diets without added synthetic lysine were 949, 835, and 741 mg/hen per d, respectively. Calculation of data from Sohail et al. (2003) showed that lysine intakes of hens fed the 18.0, 17.3, 16.7, 16.1, and 15.5 % protein diets without added synthetic lysine were 787, 754, 719, 683, and 640 mg/hen per d, respectively.

In Experiment 2, the addition of synthetic lysine while maintaining a 0.75 Met+Cys/Lys ratio had significant effects on feed intake, egg production, egg mass, feed conversion, and egg specific gravity (Table 4). These results were consistent with those of Liu et al. (2005), 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 14.9, 14.4 and 14.0% protein diets without added synthetic lysine were 710, 686, and 629 mg/hen per d, respectively, which were close to or lower than NRC (1994) recommended lysine intake value (690 mg/hen per d) (Table 5) . Calculation of data from Liu et al. (2005) showed that lysine intakes of hens fed the 14.3 and 13.6% protein diets without added synthetic lysine were 689 and 592 mg/hen per 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 (Sohail et al., 2003; Liu et al., 2004, and Liu et al., 2005). 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 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). The results of Experiment 2 and Liu et al. (2005) differed from those of Experiment 1, Sohail et al. (2003) and Liu et al. (2004) 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. (2005). Therefore, when protein level of a corn-soy diet is below 15% or lysine intake is less than 720 mg/hen per d, which was similar to NRC (1994) 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 Program developed by Roland et al. (1998; 2000) was used to evaluate profits of different added synthetic lysine levels at different protein levels. In Experiment 2, profits in hens fed the 14.0 % protein diet supplemented with 0.030% or 0.059% lysine was equal to or superior to profits of hens fed the 14.9% 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.

In conclusion, when protein level of a corn-soy diet is below 15% (supplying less than approximately 15 g protein/hen per d) or lysine intake is less than 720 mg/hen per 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.

#### **ACKNOWLEDGMENTS**

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

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

Ingredient (%)	Diet											
	1	2	3	4	5	6	7	8	9	10	11	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 <sub>3</sub>	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 <sup>1</sup>	2.00	2.00	2.00	2.00	2.00	2.00	2.00	2.00	2.00	2.00	2.00	2.00
Dical. 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 <sup>2</sup>	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 <sup>3</sup>	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
D,L-Methionine <sup>4</sup>	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 <sup>5</sup>	0.00	0.04	0.08	0.11	0.00	0.04	0.08	0.11	0.00	0.04	0.08	0.11
Cal. analysis												
CP (%)	16.0	16.1	16.1	16.1	14.9	14.9	15.0	15.0	14.4	14.5	14.5	14.5
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
A. P. (%)	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
Met+Cys (%)	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
Chemical analysis												
Methionine (%)	0.33	0.34	0.36	0.39	0.28	0.29	0.31	0.34	0.25	0.27	0.30	0.33
Met+Cys (%)	0.59	0.60	0.63	0.64	0.51	0.51	0.54	0.56	0.48	0.50	0.53	0.56
Lysine (%)	0.83	0.84	0.91	0.89	0.73	0.70	0.75	0.78	0.69	0.72	0.78	0.81

<sup>1</sup>Hardshell = large particle (passing US mesh #4 and retained by US mesh #6) CaCO<sub>3</sub> supplied by Franklin Industrial Minerals, Lowell, Florida.

<sup>2</sup>Provided per kilogram of diet: vitamin A (as retinyl acetate), 8,000 IU; cholecalciferol, 2,200 ICU; vitamin E (as D,L- $\alpha$ -tocopheryl acetate), 8 IU; vitamin B<sub>12</sub>, 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<sub>1</sub> (thiamin mononitrate), 1 mg; pyridoxine, 2.2 mg; d-biotin, 0.05 mg; vitamin K(menadione sodium bisulfate complex), 2 mg.

<sup>3</sup>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.

<sup>4</sup>D,L-methionine calculated as 99.7%.

<sup>5</sup>L-lysine 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 <sub>3</sub>	7.37	7.37	7.37	7.38	7.37	7.37	7.38	7.38	7.38
Hardshell <sup>1</sup>	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 <sup>2</sup>	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
Mineral premix <sup>3</sup>	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25
DL-Methionine	0.03	0.06	0.08	0.02	0.05	0.07	0.01	0.03	0.06
L-lysine <sup>4</sup>	0.00	0.04	0.08	0.00	0.04	0.08	0.00	0.04	0.08
Calculated analysis									
CP (%)	14.9	14.9	15.0	14.4	14.5	14.5	14.0	14.1	14.1
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
A. P. (%)	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
Met+Cys(%)	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
Chemical analysis									
Methionine (%)	0.27	0.29	0.30	0.26	0.29	0.30	0.26	0.27	0.27
Met+Cys (%)	0.52	0.55	0.55	0.50	0.54	0.55	0.51	0.51	0.50
Lysine (%)	0.73	0.79	0.77	0.72	0.77	0.79	0.72	0.74	0.72

<sup>1</sup>Hardshell = large particle (passing US mesh #4 and retained by US mesh #6) CaCO<sub>3</sub> supplied by Franklin Industrial Minerals, Lowell, Florida.

<sup>2</sup>Provided per kilogram of diet: vitamin A, 8,000 IU; cholecalciferol, 2,200 ICU; vitamin E, 8 IU; vitamin B<sub>12</sub>, 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<sub>1</sub> (thiamin mononitrate), 1 mg; pyridoxine, 2.2 mg; biotin, 0.05 mg; vitamin K (menadione sodium bisulfate complex), 2 mg.

<sup>3</sup>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.

<sup>4</sup>L-lysine calculated as 78.6%.

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 feed/hen per d)	Egg production (%)	Egg weight (g)	Egg mass (g egg/hen per d)	Feed conversion (g of feed/g of egg)	Egg specific gravity (unit)	Body weight (kg)	Mortality (%)
Protein	16.01%	103.62 <sup>a</sup>	77.57	66.88 <sup>a</sup>	51.87 <sup>a</sup>	2.00	1.0767 <sup>a</sup>	1.69 <sup>a</sup>	0.63
	14.86%	102.12 <sup>ab</sup>	75.63	65.92 <sup>b</sup>	49.87 <sup>b</sup>	2.05	1.0769 <sup>a</sup>	1.67 <sup>a</sup>	0.21
	14.42%	101.18 <sup>b</sup>	75.28	65.48 <sup>b</sup>	49.30 <sup>b</sup>	2.06	1.0775 <sup>b</sup>	1.55 <sup>b</sup>	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
Main effects and interaction		-----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

\*means the supplemental lysine level

<sup>a-b</sup> Means within a column and under each main effect with no common superscripts differ significantly

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 per d)	Egg production (%)	Egg weight (g)	Egg mass (g egg /hen per d)	Feed conversion (g of feed/g of egg)	Egg specific gravity (unit)	Body weight (kg)	Mort (%)
Protein	14.87%	95.57	70.20	65.70	46.12	2.08	1.0723	1.68 <sup>a</sup>	0.83
	14.44%	95.70	68.77	65.26	44.88	2.14	1.0720	1.66 <sup>a</sup>	2.22
	14.00%	93.92	68.47	64.68	44.32	2.13	1.0723	1.59 <sup>b</sup>	2.50
Lysine*	0.000%	93.71 <sup>b</sup>	66.91 <sup>b</sup>	65.05	43.57 <sup>b</sup>	2.16 <sup>a</sup>	1.0729 <sup>a</sup>	1.65	1.94
	0.030%	96.65 <sup>a</sup>	70.09 <sup>a</sup>	65.26	45.71 <sup>a</sup>	2.12 <sup>ab</sup>	1.0716 <sup>b</sup>	1.63	2.50
	0.059%	94.82 <sup>ab</sup>	70.45 <sup>a</sup>	65.33	46.04 <sup>a</sup>	2.06 <sup>b</sup>	1.0721 <sup>b</sup>	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
-----Probability-----									
Main effects and interaction									
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

\*means the supplemental lysine level

<sup>a-b</sup> Means within a column and under each main effect with no common superscripts differ significantly

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/hen per d)	Diet	Protein level (%)	Lysine level (%)	Lysine intake (mg/hen per d)
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				

TABLE 6. Influence of protein level and added synthetic lysine on profits (Experiment 2)<sup>1,2</sup>

	Added synthetic lysine level (%)		
	0.000	0.030	0.059
	Returns <sup>3</sup> (cents/dozen)		
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

<sup>1</sup> Corn price = \$5.52/100 lb, soy price = \$9.20/100 lb, CaCO<sub>3</sub> = \$1.50/100 lb, hard shell = \$1.50/100 lb, Dicalcium phosphate = \$12.10/100 lb, poultry oil = \$13.50/100 lb, salt = \$2.83/100 lb, Vitamin premix = \$121.00/100 lb, mineral premix = \$27.00/100 lb, DL-methionine = \$117.60/100 lb.

<sup>2</sup> The egg price spread between medium and large eggs was 11 cents

<sup>3</sup> Returns (R) were calculated using the equation:  $R = \text{UBEP} - \text{NR} - \text{PC} - \text{FdC}$ , where UBEP = Uerner Barry Egg Price, NR = nest run into package product delivered, PC = production cost, and FdC = feed cost, as described by Roland et al. (1998, 2000)

VIII. COMPARISON OF NATUPHOS AND PHYZYME AS PHYTASE  
SOURCES FOR COMMERCIAL LAYERS FED CORN-SOY DIET

**ABSTRACT** The objective of this experiment was to compare the effects of two sources of phytase on performance of commercial Leghorns fed corn-soy diets. Seven diets were fed to Hy-line W-36 hens (n = 840, 8 replicates of 15 hens per treatment) from 21 to 33 wk of age. The treatments consisted of a control diet containing 0.38% nonphytate phosphorus (NPP) and a 2 × 3 factorial arrangement of two dietary NPP concentrations (0.11 and 0.26%) with two phytase sources (Natuphos<sup>®</sup>, Phyzyme<sup>®</sup>) and without phytase. Dietary NPP had significant effects on feed intake, NPP intake, total P intake, egg production, egg weight, egg mass, egg specific gravity, and excreta P. The addition of Phyzyme<sup>®</sup> or Natuphos<sup>®</sup> significantly increased egg production and egg mass of hens fed the deficient-phosphorus diet (0.11% NPP) to levels that were similar to hens fed the control diet containing 0.38% NPP. Feed intake of hens fed the diets supplemented with Phyzyme<sup>®</sup> or Natuphos<sup>®</sup> was significantly less than that of hens fed the control diet containing 0.38% NPP. Phyzyme<sup>®</sup> or Natuphos<sup>®</sup> supplementation in the diets containing 0.11% NPP had significantly reduced excreta P of the control diet (approximately 58% and 54%, respectively) with no adverse effect on egg production and egg mass. There were no significant differences in feed intake, NPP intake, total P intake, egg production, egg weight, egg mass, feed conversion, egg specific gravity, mortality, body weight, and

excreta P between the diets supplemented with Natuphos<sup>®</sup> and the diets supplemented with Phyzyme<sup>®</sup>. In conclusion, Phyzyme<sup>®</sup> had the same positive effects on performance of commercial Leghorns fed corn-soy diets as Natuphos<sup>®</sup>.

*Key words:* Hens, phytase, NPP

## **INTRODUCTION**

Phosphorus is an essential mineral for laying hens in formation of eggshell and metabolism (Frost and Roland, 1991; Summers, 1995; Usayran and Balnave, 1995, Sohail and Roland, 2002). Only 20%-50% of plant-derived P is available to broilers, and the rest of P is in the form of phytate (myo-inositol hexaphosphate), which is poorly used by broilers (Ravindran et al. 1998). Ravindran et al. (1998) and Sebatian et al. (1998) reported that poultry can not produce enough endogenous phytase to hydrolyze and release P from phytate. To meet dietary P requirement of laying hens, inorganic P such as dicalcium phosphate and monocalcium phosphate or exogenous phytase enzymes are commonly added to commercial corn-soy layer diets. However, inorganic P supplementation is not only expensive but also leads to environmental problems by over-supplementation. Excess P from the excreta of hens can easily add to the phosphorus loading of ground water, rivers, lakes, and oceans, and can contribute to eutrophication of aquatic systems and stimulate algae growth resulting in mortality of aquatic animals (Ryden et al., 1973).

Many researchers have demonstrated that phytase supplementation (from 100 to 2000 phytase unit (FTU)/kg of feed) to diets containing 0.1% dietary nonphytate phosphorus (NPP) has positive effects on egg production, egg mass, egg weight, egg specific gravity, bone ash, and eggshell quality by improving P utilization (Van der Klis

et al., 1996; Gordon and Roland, 1997, 1998; Boiling et al., 2000a, b; Jalal and Scheideler, 2001; Roland et al., 2003; Keshavarz, 2003). Phytase supplementation decreased P excretion in the manure and reduced the potential environmental problems (Jalal and Scheideler, 2001).

There are several commercial phytase products including Natuphos<sup>® 2</sup> and Ronozyme<sup>® 3</sup> in the market. Natuphos<sup>®</sup> phytase originates from *Aspergillus niger*, and is extensively used in the poultry industry. Recently a new bacterial phytase Phyzyme<sup>® 4</sup>, which originates from the bacteria *Escherichia coli* and is produced by *Schizosacchromyces pombe*, has been introduced into market. Phytases from different sources may have different biochemical and biophysical properties such as pH-activity profile and sensitiveness to pepsin, which can affect the *in vivo* bioefficacy of phytase.

Very little research has been conducted to evaluate the effect of the novel phytase Phyzyme<sup>®</sup> on commercial Leghorns fed corn-soy diets. The objective of this experiment was to compare the effects of two sources of phytase (Phyzyme<sup>®</sup> and Natuphos<sup>®</sup>) on performance of commercial Leghorns fed corn-soy diets from 21 to 33 wk of age.

## MATERIAL AND METHODS

Seven diets were fed to Hy-line W-36 hens from 21 to 33 wk of age. The treatments consisted of a control diet containing 0.38% nonphytate phosphorus (NPP) and a 2 × 3 factorial arrangement of two dietary NPP concentrations (0.11 and 0.26%) with two phytase sources (Natuphos<sup>®</sup>, Phyzyme<sup>®</sup>) and without phytase (Table1). The diets were mixed twice and the feed samples of each mix were analyzed for calcium and

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<sup>2</sup> BASF Corp., Mount Olive, NJ.

<sup>3</sup> Roche Vitamins, Parsippany, NJ.

<sup>4</sup> Danisco Animal Nutrition, Carol Stream, IL.

P concentrations according to AOAC procedures (1984) by Experimental Station Chemical Laboratories, University of Missouri-Columbia.

One phytase unit is defined as the amount of enzyme activity that liberates 1 mmol of inorganic P/min from a 0.5 mM Na-phytate solution at pH 5.5 and 37°C. Phytases from two different sources (Natuphos<sup>®</sup> 600 produced by BASF and Phyzyme<sup>®</sup> XP 5000G produced by Danisco Animal Nutrition) were supplemented at 300 FTU/kg feed in diets at two different P concentrations. The phytase contents of Natuphos<sup>®</sup> and Phyzyme<sup>®</sup> in the premixes and diets were analyzed by Danisco Animal Nutrition to confirm enzyme activity.

Hy-line W-36 hens (n = 840) at 21 weeks of age were randomly assigned into 7 treatments (8 replicates of 15 hens per treatment). Replicates were equally distributed into upper and lower cages to minimize cage level effect. Three hens were housed in a 40.6 × 45.7 cm<sup>2</sup> cage and five adjoining cages consisted of a replicate. 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). Pullets were moved into the house at 18 wk of age. Light was increased by 15 minutes per week from 12 hours per day to 16 hours per day. All hens were supplied with feed and water *ad libitum*. Egg production was recorded daily, egg weight and feed consumption were recorded weekly, and egg specific gravity was recorded bi-weekly. Egg weight and egg specific gravity were measured using all eggs produced during two consecutive days. Feed intake was determined by subtracting the ending feed weight of each trough (each replicate) from the beginning feed weight weekly. Egg specific gravity was determined using 11 gradient saline solutions varying in specific gravity from 1.060 to 1.100

incremented with 0.005-unit increments (Holder and Bradford, 1979). At the middle and end of the experiment, excreta samples were obtained (three replicates per treatment) by placing a pan under cages for 24 hour. Excreta samples were then dried for 48 hour at 100 C and were analyzed for P concentration according to AOAC procedures (1984) by Experimental Station Chemical Laboratories, University of Missouri-Columbia. Mortality was determined daily and egg production and feed consumption were adjusted to a hen-day basis. Body weight was obtained by randomly weighing 3 hens per group at the beginning and the end of the experiment. Mean value and standard deviation of the beginning body weight and egg production was  $1.41 \pm 0.12$  kg and  $44.8 \pm 9.4$  %, respectively.

Statistical analyses of data were performed by using the general linear models (GLM) procedure of SAS (SAS Institute, 2000). A  $2 \times 3$  factorial arrangement of two dietary NPP concentrations (0.11 and 0.26%) with two phytase sources (Natuphos<sup>®</sup>, Phyzyme<sup>®</sup>) and without phytase was used to analyze the main effects of dietary NPP and phytase and their interactions (Diet 1 to 6). If differences in treatment means were detected by ANOVA, orthogonal contrasts were applied to separate means. Four preplanned additional nonorthogonal contrasts were also carried out to compare control diet and several specific diets. Statements of statistical significance are based on a probability of ( $P \leq 0.05$ ).

## **RESULTS AND DISCUSSION**

A significant interaction between dietary NPP and phytase on feed intake was observed (Table 2). Dietary NPP had a significant effect on feed intake of hens fed the diets without phytase from week 1 to the end of trial. As dietary NPP concentration

increased from 0.11 to 0.26 in the diets without phytase, feed intake significantly increased from 84.21 to 92.08 g feed/hen per d, resulting in a 9.35% increase of feed intake. The addition of phytase to the diets containing 0.11% NPP had a significant effect on feed intake, while the addition of phytase to the diets with 0.26% NPP had no effect on feed intake. The addition of phytase prevented the decline of feed intake of hens fed the phosphorus-deficient diets (0.11% NPP). These results are in agreement with those of Gordon and Roland (1997), Jalal and Scheideler (2001), and Roland et al. (2003), who reported that the addition of phytase to diets containing 0.1% NPP significantly increased feed intake. Feed intake of hens fed the diets supplemented with Natuphos<sup>®</sup> was similar to that of hens fed the diets supplemented with Phyzyme<sup>®</sup> at both NPP concentrations. Feed intake of hens fed the diets supplemented with Phyzyme<sup>®</sup> or Natuphos<sup>®</sup> was significantly lower than that of hens fed the control diet containing 0.38% NPP. There was no significant difference in feed intake between the control diet containing 0.38% NPP and the diet containing 0.26% NPP without phytase. Hens fed the diets containing 0.11% NPP had significantly lower feed intake than hens fed the control diet containing 0.38% NPP.

A significant interaction between dietary NPP and phytase was observed on NPP intake (Table 2). Dietary NPP concentration had a significant effect on NPP intake. When dietary NPP was 0.11%, NPP intake of hens ranged from 93 to 101 mg/hen per d, which was much lower than dietary NPP requirement of hens (250 mg/hen per d) (NRC, 1994). NPP intake of hens fed the diets containing 0.26% NPP was around 240 mg/hen per d, which was close to NRC value of 250 mg/hen per day (NRC, 1994). A wide range of NPP from 290 to 470 mg/hen per d is used in the industry (Roland, 1994). The NRC

recommended value of dietary NPP has declined from 350 mg/hen per d (NRC, 1984) to 250 mg/hen per d (NRC, 1994). Sohail and Roland (2002) reported that dietary NPP requirement of young laying hens for maximum performance ranged from 250 to 325 mg/hen per d and a higher margin of safety for dietary P might be necessary. Dietary NPP intake in hens fed the control diet containing 0.38 NPP was 358 mg/hen per d, which was close to NRC value of 350 mg/hen per d (NRC, 1984). Total P intake of hens fed the diets containing 0.11% NPP was significantly lower than that of hens fed the diets containing 0.26% NPP and hens fed the control diet containing 0.38 % NPP (Table 2).

A significant interaction between dietary NPP and phytase on egg production was observed (Table 2). Egg production significantly increased when dietary NPP of the diets without phytase increased from 0.11 to 0.26%. Phytase supplementation in diets containing 0.11% NPP significantly improved egg production from week 3 to the end of trial, while phytase supplementation in diets containing 0.26% NPP had no effect on egg production. The different egg production response to phytase supplementation can be attributed to that 0.26% NPP concentration in the diets or 241 mg NPP intake/hen per d has fulfilled the requirement of laying hens, and 0.11% NPP concentration in the diets or 93 mg NPP intake/hen per d was not enough for laying hens. There was no significant difference in egg production between Natuphos<sup>®</sup> and Phyzyme<sup>®</sup> at both dietary NPP concentrations. Egg production of hens fed the diets supplemented with Natuphos<sup>®</sup> or Phyzyme<sup>®</sup> was similar to that of hens fed the control diet containing 0.38% NPP. These results are consistent to those of Gordon and Roland (1997, 1998), Jalal and Scheideler (2001), and Roland et al. (2003), who reported that supplementing diets containing 0.1% NPP with phytase significantly increased egg production to the level of hens fed

adequate-phosphorus diets. Egg production of hens fed the control diet containing 0.38% NPP was similar to that of hens fed the diet containing 0.26% NPP without phytase. Hens fed the control diet containing 0.38% NPP had significantly higher egg production than hens fed the phosphorus-deficient diet (0.11% NPP).

Phytase supplementation had no significant effect on egg weight (Table 2). Egg weight in hens fed the diets supplemented with Natuphos<sup>®</sup> was similar to that in hens fed the diets with Phyzyme<sup>®</sup>. Dietary NPP had a significant effect on egg weight. Egg weight significantly increased when dietary NPP increased from 0.11% to 0.26%. There were no significant differences in egg weight between the control diet containing 0.38% NPP and the diets supplemented with Natuphos<sup>®</sup> or Phyzyme<sup>®</sup>.

A significant interaction between dietary NPP and phytase on egg mass was observed (Table 2). As dietary NPP increased from 0.11 to 0.26 in the diets without phytase, egg mass significantly increased from 42.17 to 48.29 g egg/hen per d, resulting in a 14.51% increase of egg mass. Phytase supplementation significantly increased egg mass in the diets containing 0.11% NPP, but had no effect on egg mass in the diets containing 0.26% NPP. Egg mass in hens fed the diets supplemented with Natuphos<sup>®</sup> was similar to that in hens fed the diets with Phyzyme<sup>®</sup>. There were no significant difference between the control diet containing 0.38 % NPP and the diets supplemented with Phyzyme<sup>®</sup> or Natuphos<sup>®</sup>. Similarly, Jalal and Scheideler (2001) reported that phytase supplementation significantly increased egg mass of hens fed 0.10% NPP diet. Egg mass of hens fed the control diet was significantly higher than that of hens fed the phosphorus-deficient diet without phytase (0.11% NPP), but was similar to that of hens fed the diet containing 0.26% NPP without phytase.

Dietary NPP had no significant effect on feed conversion (Table 3). Phytase effect on feed conversion was approaching significant ( $P < 0.08$ ). There was no significant difference in feed conversion between Natuphos<sup>®</sup> and Phyzyme<sup>®</sup>. Feed conversion of hens fed the diets supplemented with Phyzyme<sup>®</sup> was significantly lower than that of hens fed the control diet containing 0.38% NPP.

Although feed intake of hens fed the diets supplemented with Phyzyme<sup>®</sup> or Natuphos<sup>®</sup> was significantly less than that of hens fed the control diet, there were no significant differences in egg mass and egg production between the control diet containing 0.38% NPP and the diets supplemented with Phyzyme<sup>®</sup> or Natuphos<sup>®</sup>. Also, feed conversion of hens fed the diets supplemented with Phyzyme<sup>®</sup> was significantly lower than that of hens fed the control diet. Similarly, Roland et al. (2003) reported that even though less feed was consumed, there was no difference in egg production between adequate-phosphorus diets and the deficient-phosphorus diets supplemented with phytase. Therefore, phytase<sup>®</sup> supplementation might have improved not only phosphorus availability but also the availabilities of some other nutrients such as energy and amino acids. This conclusion was supported by those of Namkung and Lesson (1999), who reported that phytase supplementation improved AME and digestibilities for some amino acids such as Val and Ile in broilers.

Dietary NPP had a significant effect on egg specific gravity (Table 3). As dietary NPP concentration increased from 0.11% to 0.26%, egg specific gravity significantly decreased. Phytase had no significant effect on egg specific gravity. Egg specific gravity in hens fed the diets supplemented with Natuphos<sup>®</sup> was similar to that in hens fed the diets with Phyzyme<sup>®</sup>. Egg specific gravity of hens fed the diets supplemented with

Phyzyme<sup>®</sup> was significantly higher than that of hens fed the control diet. Both dietary NPP and phytase had no effect on final body weight and mortality (Table 3).

Dietary NPP had significant effect on excreta P content (Table 3). When NPP concentration decreased from 0.38% to 0.11%, a 58% or 54% reduction in excreta P was obtained by supplementing the diets containing 0.11% NPP with Phyzyme<sup>®</sup> or Natuphos<sup>®</sup>, respectively. These results are in agreement with those of Boling et al. (2000b), who reported that phytase supplementation decreased excreta P concentration approximately 50%. Phytase supplementation can greatly reduce potential P environmental pollution problems caused by inorganic P in feed.

In conclusion, the addition of Phyzyme<sup>®</sup> or Natuphos<sup>®</sup> significantly increased egg production and egg mass of hens fed the deficient-phosphorus diet (0.11% NPP) to levels that were similar to hens fed the control diet containing 0.38% NPP. Feed intake of hens fed the diets supplemented with Phyzyme<sup>®</sup> or Natuphos<sup>®</sup> was significantly less than that of hens fed the control diet containing 0.38% NPP. Feed conversion and egg specific gravity of hens fed the diets supplemented with Phyzyme<sup>®</sup> were significantly better than those of hens fed the control diet. Phyzyme<sup>®</sup> or Natuphos<sup>®</sup> supplementation in the diets containing 0.11% NPP had significantly reduced excreta P of the control diet (approximately 58% and 54%, respectively) with no adverse effect on egg production and egg mass. There were no significant differences in feed intake, NPP intake, total P intake, egg production, egg weight, egg mass, feed conversion, egg specific gravity, mortality, body weight, and excreta P between the diets supplemented with Natuphos<sup>®</sup> and the diets supplemented with Phyzyme<sup>®</sup>. Phyzyme<sup>®</sup> had the same positive effects on performance of commercial Leghorns fed corn-soy diets as Natuphos<sup>®</sup>.

## **ACKNOWLEDGMENTS**

The authors thank Danisco Animal Nutrition, Carol Stream, IL, for funding support of this research.

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TABLE 1. Ingredient and nutrient content of the experimental diets

Ingredient	Diet 1	Diet 2	Diet 3	Diet 4	Diet 5	Diet 6	Control diet
Corn (%)	62.54	62.54	62.54	61.84	61.84	61.84	61.30
Soybean meal (%)	25.66	25.66	25.66	25.72	25.72	25.72	25.76
CaCO <sub>3</sub> (%)	7.95	7.95	7.95	7.48	7.48	7.48	7.11
Hardshell <sup>1</sup> (%)	2.00	2.00	2.00	2.00	2.00	2.00	2.00
Dicalcium phosphate (%)	0.00	0.00	0.00	0.84	0.84	0.84	1.49
Poultry oil (%)	0.88	0.88	0.88	1.15	1.15	1.15	1.36
Salt (%)	0.38	0.38	0.38	0.38	0.38	0.38	0.38
Vitamin Premix <sup>2</sup> (%)	0.25	0.25	0.25	0.25	0.25	0.25	0.25
Mineral premix <sup>3</sup> (%)	0.25	0.25	0.25	0.25	0.25	0.25	0.25
DL-methionine (%)	0.10	0.10	0.10	0.10	0.10	0.10	0.10
Natuphos <sup>®4</sup> (FTU/kg of feed)		300			300		
Phyzyme <sup>®5</sup> (FTU/kg of feed)			300			300	
Calculated analysis							
Crude protein (%)	17.38	17.38	17.38	17.35	17.35	17.35	17.33
ME (kcal/kg)	2816	2816	2816	2816	2816	2816	2816
Ca (%)	4.00	4.00	4.00	4.00	4.00	4.00	4.00
Total P (%)	0.32	0.32	0.32	0.47	0.47	0.47	0.59
Nonphytate P (%)	0.11	0.11	0.11	0.26	0.26	0.26	0.38
Methionine+Cystine (%)	0.69	0.69	0.69	0.69	0.69	0.69	0.69
Lysine (%)	0.92	0.92	0.92	0.92	0.92	0.92	0.92
Chemical analysis							
Ca (%)	3.78	4.89	4.57	4.27	4.44	4.39	4.84
Total P (%)	0.36	0.35	0.34	0.53	0.54	0.51	0.68

<sup>1</sup>Hardshell = large particle (passing US mesh #4 and retained by US mesh #6) CaCO<sub>3</sub> supplied by Franklin Industrial Minerals, Lowell, Florida.

<sup>2</sup>Provided per kilogram of diet: vitamin A (as retinyl acetate), 8,000 IU; cholecalciferol, 2,200 ICU; vitamin E (as DL- $\alpha$ -tocopheryl acetate), 8 IU; vitamin B<sub>12</sub>, 0.02 mg; riboflavin, 5.5 mg; D-pantothenic acid, 13 mg; niacin, 36 mg; choline, 500 mg; folic acid, 0.5 mg; vitamin B<sub>1</sub> (thiamin mononitrate), 1 mg; pyridoxine, 2.2 mg; d-biotin, 0.05 mg; vitamin K (menadione sodium bisulfate complex), 2 mg.

<sup>3</sup>Provided per kilogram of diet: manganese, 65 mg; iodine, 1 mg; iron, 55 mg; copper, 6 mg; zinc, 55 mg; selenium, 0.3 mg.

<sup>4</sup>BASF Corp., Mount Olive, NJ.

<sup>5</sup>Danisco Animal Nutrition, Carol Stream, IL.

TABLE 2. Effects of dietary NPP and phytase on performance of Hy-line W-36 hens from 21 to 33 wk of age

	NPP (%)	Phytase <sup>1</sup>	Feed intake (g feed/hen per d)	NPP intake <sup>2</sup> (mg/hen per day)	Total P intake <sup>3</sup> (mg/hen per day)	Egg production (%)	Egg weight (g)	Egg mass (g egg/hen per d)
Diet 1	0.11	0	84.2 <sup>b</sup>	93 <sup>d</sup>	303 <sup>d</sup>	78.1 <sup>b</sup>	54.03	42.17 <sup>b</sup>
Diet 2	0.11	Natuphos <sup>®</sup>	92.0 <sup>a</sup>	101 <sup>c</sup>	322 <sup>c</sup>	89.0 <sup>a</sup>	53.77	47.86 <sup>a</sup>
Diet 3	0.11	Phyzyme <sup>®</sup>	91.0 <sup>a</sup>	100 <sup>c</sup>	309 <sup>d</sup>	87.8 <sup>a</sup>	54.12	47.51 <sup>a</sup>
Diet 4	0.26	0	92.8 <sup>a</sup>	241 <sup>a</sup>	487 <sup>a</sup>	88.7 <sup>a</sup>	54.48	48.29 <sup>a</sup>
Diet 5	0.26	Natuphos <sup>®</sup>	90.4 <sup>a</sup>	235 <sup>b</sup>	483 <sup>a</sup>	86.3 <sup>a</sup>	54.27	46.88 <sup>a</sup>
Diet 6	0.26	Phyzyme <sup>®</sup>	92.5 <sup>a</sup>	241 <sup>a</sup>	472 <sup>b</sup>	89.1 <sup>a</sup>	54.74	48.76 <sup>a</sup>
Main effect (Diet 1 to 6)								
NPP	0.11		89.1	98	312	84.9	53.97 <sup>b</sup>	45.84
	0.26		91.9	239	481	88.0	54.49 <sup>a</sup>	47.98
Phytase		0	88.5	167	395	83.4	54.25	45.23
		Natuphos <sup>®</sup>	91.2	168	402	87.7	54.02	47.37
		Phyzyme <sup>®</sup>	91.8	171	391	88.4	54.43	48.13
Control diet	0.38	0	94.2	358	641	87.5	54.58	47.77
Pooled SEM			0.58	1.19	3.61	1.09	0.26	0.63
			-----Probability-----					
NPP main effect			0.0006	0.0001	0.0001	0.0010	0.0154	0.0001
Phytase main effect			0.0025	0.0568	0.0072	0.0001	NS	0.0001
NPP × Phytase			0.0001	0.0002	0.0056	0.0001	NS	0.0001
Control vs. Diets with Natuphos <sup>®</sup>			0.0069	0.0001	0.0001	NS	NS	NS
Control vs. Diets with Phyzyme <sup>®</sup>			0.0233	0.0001	0.0001	NS	NS	NS
Control vs. Diet 1			0.0001	0.0001	0.0001	0.0001	NS	0.0001
Control vs. Diet 4			NS	0.0001	0.0001	NS	NS	NS

<sup>1</sup>Both Natuphos<sup>®</sup> and Phyzyme<sup>®</sup> supplemented at 300 FTU/kg of feed.

<sup>2</sup>NPP intake was based on calculated dietary NPP concentration.

<sup>3</sup>Total P intake was based on chemical analyzed dietary P concentration.

TABLE 3. Effects of dietary NPP and phytase on performance of Hy-line W-36 hens from 21 to 33 wk of age

	NPP (%)	Phytase <sup>1</sup>	Feed conversion (g of feed/g of egg)	Egg specific gravity (unit)	Mortality (%)	Final body weight (kg)	P content in excreta (%)
Diet 1	0.11	0	2.00	1.0884 <sup>a</sup>	0.00	1.44	0.94 <sup>b</sup>
Diet 2	0.11	Natuphos <sup>®</sup>	1.92	1.0872 <sup>b</sup>	0.83	1.47	0.98 <sup>b</sup>
Diet 3	0.11	Phyzyme <sup>®</sup>	1.92	1.0876 <sup>ab</sup>	0.00	1.48	0.88 <sup>b</sup>
Diet 4	0.26	0	1.92	1.0868 <sup>b</sup>	0.00	1.52	1.63 <sup>a</sup>
Diet 5	0.26	Natuphos <sup>®</sup>	1.93	1.0866 <sup>b</sup>	1.67	1.45	1.62 <sup>a</sup>
Diet 6	0.26	Phyzyme <sup>®</sup>	1.90	1.0867 <sup>b</sup>	0.83	1.54	1.67 <sup>a</sup>
Main effect (Diet 1 to 6)							
NPP	0.11		1.95	1.0877 <sup>a</sup>	0.28	1.46	0.93 <sup>b</sup>
	0.26		1.92	1.0867 <sup>b</sup>	0.83	1.50	1.64 <sup>a</sup>
Phytase		0	1.96	1.0876	0.00	1.48	1.28
		Natuphos <sup>®</sup>	1.93	1.0869	1.25	1.46	1.30
		Phyzyme <sup>®</sup>	1.91	1.0871	0.42	1.51	1.28
Control diet	0.38	0	1.97	1.0862	0.83	1.48	2.11
Pooled SEM			0.015	0.0004	0.59	0.02	0.03
-----Probability-----							
NPP main effect			NS	0.0001	NS	NS	0.0001
Phytase main effect			0.0769	0.0578	NS	NS	NS
NPP × Phytase			NS	NS	NS	NS	NS
Control vs. Diets with Natuphos <sup>®</sup>			NS	0.0729	NS	NS	0.0001
Control vs. Diets with Phyzyme <sup>®</sup>			0.0269	0.0144	NS	NS	0.0001
Control vs. Diet 1			NS	0.0001	NS	NS	0.0001
Control vs. Diet 4			NS	NS	NS	NS	0.0001

<sup>a-b</sup>Means within a column with no common superscripts differ significantly.

<sup>1</sup>Both Natuphos<sup>®</sup> and Phyzyme<sup>®</sup> supplemented at 300 FTU/kg of feed

## IX. CONCLUSIONS

### Dietary Energy Experiment 1 (section II)

Increasing dietary energy by the addition of poultry oil linearly decreased feed intake. When dietary energy increased from 2,719 to 2,956 kcal ME/kg, hens adjusted feed intake from 107.6 to 101.1 g/hen per day to achieve a constant energy intake so that the same amount of dietary energy (5.8 kcal) was used to produce 1 g of egg. Increasing dietary energy by the addition of poultry oil increased early egg weight, which was mostly due to increased yolk weight. Increasing dietary energy by the addition of poultry oil significantly decreased feed conversion and egg specific gravity, but had no effect on egg production, egg mass, body weight and mortality. Increasing dietary energy by the addition of poultry oil to a dietary energy level of 2,877 kcal ME/kg maximized egg weight during Phase I (from 21 to 36 wk of age). There can be no fixed ideal dietary energy level for optimal profits, due to varying feed ingredient prices and egg price.

### Dietary Energy Experiment 2 (section III)

Increasing dietary energy contents by the addition of poultry oil linearly decreased feed intake and improved feed conversion. When dietary energy linearly increased from 2776 to 2908 kcal/kg, hens adjusted feed intake from 105.0 to 101.7 g/hen/day so that the same amount of dietary energy (5.2 kcal) was used to produce 1 g

egg. Based on feed conversion, increasing dietary energy to 2,864 kcal ME/kg might be sufficient for optimal performance of laying hens during Phase 2 (from 40 to 51 week of age). There can be no fixed ideal dietary energy level for optimal profits during phase 2 (from wk 40 to 51 of age), due to varying feed ingredient prices and egg price. Increasing dietary energy by the addition of poultry oil had significant effects on percent yolk, percent albumen, Yolk/Albumen ratio, and percent dirty eggs.

#### Dietary Energy Experiment 3 (section IV)

Increasing dietary energy by the addition of poultry oil had no significant effect on significant effect on feed intake, egg production, egg weight, egg mass, feed conversion, body weight, mortality, percent of egg solids, egg specific gravity, shell weight, shell thickness, dirty eggs, crack eggs, Haugh unit, and yolk color. An ideal dietary energy level for optimal performance could not be determined for laying hens during phase 3 (from wk 55 to 64 wk of age). There can be no fixed ideal dietary energy level for optimal profits during phase 3, due to varying feed ingredient prices and egg price.

#### Dietary Energy Experiment 4 (section V)

There was a linear response of yolk color to increased dietary energy. Increasing dietary energy by the addition of poultry oil had no significant effect on feed intake, egg production, egg weight, egg mass, feed conversion, body weight, mortality, percent of egg components, percent of egg solids, egg specific gravity, and Haugh unit after molt. With increasing dietary energy hens adjusted feed intake to achieve a constant energy

intake so that the similar quantities of dietary energy (5.8-5.9 kcal) were used to produce 1 g of egg. The ideal dietary energy/protein (lysine) ratio for optimal post-molt performance could not be determined, and there can be no fixed ideal dietary energy level for optimal profits for hens during second cycle phase 1 (from 70 to 81 wk of age).

#### Dietary Energy Experiment 5 and 6 (section VI)

Increasing dietary energy had no significant effect on all parameters, except feed conversion, during second cycle phase 2 (from 86 to 95 wk of age). Increasing dietary energy linearly improved feed conversion. Increasing dietary energy had no effect all parameters, except body weight of hens, during second cycle phase 3 (from 101 to 110 wk of age). Based on improved feed conversion, dietary energy of 2,846 kcal ME/kg might be enough for optimal performance during second cycle phase 2. Based on body weight of hens, dietary energy level for optimal performance should be less than 2,936 kcal ME/kg during second cycle phase 3. There can be no fixed ideal dietary energy level for optimal profits for post-molt egg production.

#### Synthetic Amino Acid Experiments 1 and 2 (section VII)

When protein level of a corn-soy diet is below 15% (supplying less than approximately 15 g protein/hen per d) or lysine intake is less than 720 mg/hen per 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.

#### Phytase Experiments 1 (section VIII)

The addition of Phyzyme<sup>®</sup> or Natuphos<sup>®</sup> significantly increased egg production and egg mass of hens fed the deficient-phosphorus diet (0.11% NPP) to levels that were similar to hens fed the control diet containing 0.38% NPP. Feed intake of hens fed the diets supplemented with Phyzyme<sup>®</sup> or Natuphos<sup>®</sup> was significantly less than that of hens fed the control diet containing 0.38% NPP. Feed conversion and egg specific gravity of hens fed the diets supplemented with Phyzyme<sup>®</sup> were significantly better than those of hens fed the control diet. Phyzyme<sup>®</sup> or Natuphos<sup>®</sup> supplementation in the diets containing 0.11% NPP had significantly reduced excreta P of the control diet (approximately 58% and 54%, respectively) with no adverse effect on egg production and egg mass. There were no significant differences in feed intake, NPP intake, total P intake, egg production, egg weight, egg mass, feed conversion, egg specific gravity, mortality, body weight, and excreta P between the diets supplemented with Natuphos<sup>®</sup> and the diets supplemented with Phyzyme<sup>®</sup>. Phyzyme<sup>®</sup> had the same positive effects on performance of commercial Leghorns fed corn-soy diets as Natuphos<sup>®</sup>.

#### Antibiotic Experiments 1 and 2 (section III and IV)

While Tylosin had no effect on feed intake, egg production, egg weight, feed conversion, mortality, percent of egg component, percent of egg solids, and Haugh unit, Tylosin supplementation significantly reduced body checks and increased yolk color,

resulting positive effects on egg quality of laying hens during phase 2 (from 40 to 51 wk of age). While Tylosin had no effect on performance and percent of egg solids, Tylosin supplementation significantly reduced dirty eggs, resulting in a positive effect on eggshell quality of laying hens during phase 3 (from 55 to 64 wk of age).

#### Molting Experiments 1 (section V)

Egg production of hens molted by feed withdrawal was significantly higher than that of hens molted by no salt diet in wk 6, 7, 9, 10, and 11. Hens molted by feed withdrawal had significantly higher egg specific gravity than hens molted by no salt diet during wk 8. There were no significant differences in overall average egg production and egg specific gravity due to molting method during second cycle phase 1 (from 70 to 81 wk of age). Other than slightly reduced egg production and egg specific gravity, feeding no salt diet to induce molt could be used as an alternative for conventional feed withdrawal method.

#### Molting Experiments 2 and 3 (section VI)

Molting method had no effect on egg production and egg mass during the early (from 70 to 81 wk of age) and middle (from 86 to 95 wk of age) stage of post-molt production period. However, hens molted by feed withdrawal had significantly higher egg production and egg mass during latter stage (from 101 to 110 wk of age) of post-molt production period, compared to hens molted by no salt diet. Although hens molted by feed withdrawal had numerically higher egg specific gravity than hens molted by no salt diet, there was no significant difference in egg specific gravity due to molting method.

Molting method had no significant effect on whole egg solids, albumen solid, yolk solid, Haugh unit and yolk color. Therefore, feeding no salt diet can produce acceptable long-term post-molt performance and egg quality, and may be considered as an effective alternative molt method for conventional feed withdrawal.

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