Egypt. Poult. Sci. Vol (28) (I): (103-124)

THE EFFECT OF DIETARY TOTAL SULFUR AMINO ACIDS, LYSINE, VALINE AND THREONINE SUPPLEMENTATION LEVELS ON LAYING HEN PERFORMANCE AND EGG QUALITY MEASUREMENTS

By

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Received: **21/1/2008** Accepted: **17/2/2007**

Abstract: *One hundred and twenty Bovans brown hens (58-wk of age) were used in order to evaluate the effect of different dietary supplemented TSAA (add as Met), lysine (Lys), valine (Val), and threonine (Thr) amino acid supplementation levels on laying hen performance and egg quality measurements. Hens were randomly assigned to 10 treatments (12 hens in each, assigned to 4 replicates). Feed and water were supplied ad-libitum with 16 hr of light per day. Corn-soybean meal basal diet was formulated to cover the nutrient requirements recommended for the breed and served as the control diet (T1; contains 680, 810, 775 and 618 mg/H/D of TSAA, Lys, Val and Thr, respectively). TSAA, as DL-Met, L-Lys, L-Val and L-Thr were added to the control diet at levels of 110 and 120% (of each amino acid) of the breed AA requirements to form treatments from T2 to T9 as following: 750 and 820 mg TSAA/H/D for T2 & T3; 890 and 970 mg Lys/H/D for T4 & T5; 850 and 930 mg Val/H/D for T6 & T7; 680 and 740 mg Thr/H/D for T8 & T9. Treatment 10 (All) represents the combined effect of 110% of each added AA (750, 890, 850 and 680 mg/H/D for TSAA, Lys, Val and Thr, respectively). Final body weight (BW), Egg production (EP), egg weight (EW), Egg mass (EM), feed intake (FI), feed conversion ratio (FCR) and egg quality & composition analysis were determined.*

The results indicated that TSAA (750 mg/H/D) optimized EW, EP, EM and FCR while FI was minimized. This means that 750 mg/H/D is the optimum dietary TSAA for maximizing egg parameters and FCR. Lys (890 mg/H/D), Thr (680 mg/H/D) and All (110% of all AAs) treatments resulted in relatively better EW, EP, EM and FCR than the control, Lys (970 mg/H/D), Thr (740 mg/H/D) and Val supplementations. A significant reduction in final BW was resulted from Val (930 mg/H/D) and Thr (680 *mg/H/D) compared with Lys (970 mg/H/D) and TSAA (750 mg/H/D) treatments.*

Egg shell thickness was not affected by the different treatments while egg shell percentage significantly increased by increasing TSAA (750 & 820 mg/H/D), Lys (970 mg/H/D), Val (850 & 930 mg/H/D) and Thr (740 mg/H/D) levels over that of the control. No significant differences were noticed either among the amino acids supplemented treatments or between these treatments and the control in each of the egg yolk percentage, albumen percentage and albumen yolk ratios. Egg protein, solids and ash were increased while egg ether extract was decreased as dietary TSAA, Lys, Val, and Thr increased. The highest relative economical efficiency (REE; 124%) was recorded for TSAA (750 mg/H/D) treatment while the lowest (90%) was recorded for Val (930 mg/H/D). So, amino acids supplementation to corn-soybean diets is recommended to improve performance and egg quality of laying hens.

INTRODUCTION

Egg production industry is facing various problems that need to be solved, such as reduction of egg production, egg quality and meat of spent hens during the laying cycle phases.

Methionine (Met) is the first limiting AA in corn-soybean meal poultry diets followed by lysine (Lys) and threonine (Thr). These limiting AAs are commercially available at economical prices; their utilization is beneficial for increasing layer facilities profit by optimizing egg production, composition and quality. Shafer et al. (1998) indicated that supplementation of layer hen diets with Met significantly increased albumen component and yolk mass yield. In the second production cycle of brown-egg layer hens, increasing level of dietary total sulfur amino acids (TSAA) improved egg weight (EW) and feed intake (FI) found by Barbosa et al., (1999a). Togashi et al. (2002) showed that the use of 0.582% and 0.569% TSAA in laying hen diets improved EW and egg mass (EM), respectively. However, Ahmad and Roland (2003) found little or no effect on egg production (EP) or FI when various levels of TSAA were supplemented to laying hen diet. Dietary TSAA levels for maximum EP and feed conversion ratio (FCR) were near 811 and 699 mg/H/D whereas these of EW were closer to 877 and 779 mg/H/D for phases I and II, respectively (Novak et al., 2004). On the other hand, El-Husseiny et al. (2005) reported that Met (0.40and 0.50%) had no significant effect on laying hen performance and egg quality.

Scheideler et al. (1996) reported that increasing Lys from 500 to 1000 mg /H/D increased EP parameters during late production cycle of Leghorn hens. The increment of Lys from 677 to 1154 or 1613 mg/H/D did not affect FI and EP. Significant increases in egg and albumen weight were noticed with increased Lys intake from 42 to 64 wks of age for Hy-Line W-36 hens (Prochaska et al., 1996). Besides, Barbosa et al. (1999b) reported that the brown-egg layer hens (second production cycle) had good response to Lys supplementation where, EW, EM, and FCR were improved by Lys supplementation. Similarly, Balnave et al. (2000) found that the daily Lys requirement for maximum EP was 975 mg/H/D. FI was reduced by increasing dietary Lys while Lys intake, egg shell breaking strength and albumen weight were increased. Novak et al. (2004) concluded that the dietary levels of Lys for optimizing EP are close to 860 and 715 mg /H/D for phases I and II, respectively.

Ishibashi et al. (1998) found that FI and EM were increased as dietary Thr increased during period of 29-39 wks of age. Martinez-Amezcua et al. (1999) reported that EP, EW and FCR improved as Thr level over that of NRC increased (when fed to post-peak hens 60-80 wks of age). Similarly, Faria et al. (2002) mentioned that supplementation of layer hen diets with graded level of Thr (0.35-0.58%) increased EP, EM and daily Thr intake. In addition, Khalifah and Abdalla (2005) indicated that supplemented graded levels of Thr from 5.19 to 6.29 g/kg diet to Gimizah layer (40-48 wks of age) diets had no significant effect on EP and FI. However, EW, EM and FCR improved significantly with increasing dietary Thr level up to 6.07 g/kg diet.

The combination of Lys, Met and/or Thr in layer hen diets did not affect EP, FI or egg yield (Koelkebeck et al., 1991). However, Okazaki et al. (1994) used diets containing Lys, Met and Thr (130 and 160% of NRC), the performance of hens was slightly superior compared to that of the other groups fed on 85, 100, and 115% of NRC. AA requirements calculated from EP and EM were 105-118% during 30 to 60 week of age. EW increased with increasing dietary AA level. Recently, Azazi et al. (2006) reported that the addition of 0.05 or 0.10% Met and 0.12% Lys to Bovans brown commercial laying hen diets (16.0% CP) significantly improved EP, EM, FI, FCR and economical efficiency. However, no significant effects were observed in BW and egg quality.

The margin between required and excess valine (Val) is wide in laying hen diets. Harms and Russel (2001) reported that EP, EW and egg component were increased with increasing Val level from 0.525 to 0.700% in hen diets. In addition, Peganova and Eder (2002) showed that dietary Val (up to 1.06%) did not cause any depression in laying hens performance. However, 1.36% Val reduced FC and EM by 5-10%.

The objective of this study was to evaluate the effect of supplementation TSAA (add as Met), Lys, Val and Thr amino acids to laying hen diets on production performance, egg quality measurements, egg composition analysis and economical efficiency.

MATERIALS AND METHODS

The present work was conducted at the poultry experimental farm, College of Agriculture, Suez Canal University, Ismailia, Egypt. One hundred and twenty Bovans brown hens (58 wks of age) were used in order to evaluate the effect of different dietary TSAA (add as Met), Lys, Val, and Thr levels on hen performance, egg quality parameters and economic efficiency. Hens, close in body weight and have high egg production%, were weighed and randomly assigned to 10 treatments (12 birds per treatment). Each dietary treatment was replicated four times (3 birds per replicate). Hens were kept under similar conditions in a semi-open layer house in battery cages with raised wire floors. Each replicate was represented by one cage (48 ×36 × 32 cm). Replicates were randomly and equally distributed into upper and lower cages to minimize cage effect. During the experimental period (8 weeks), 16 hr of light: 8 hr dark were provided daily. Hens in each replicate shared a feed trough and had access to cups for drinking water. All hens were supplied with food and water *ad-libitum*. General management was applied according to the breed management guide.

Corn-soybean meal basal diet was formulated to cover the nutrient requirements recommended for the breed and served as the control diet (T1; contains 680, 810, 775 and 618 mg/H/D of TSAA, Lys, Val and Thr, respectively). TSAA, as DL-Met, L-Lys, L-Val and L-Thr were added to the control diet at levels of 110 and 120% (of each amino acid) of the breed AA requirements to form treatments from T2 to T9 as following: 750 and 820 mg TSAA/H/D for T2 & T3; 890 and 970 mg Lys/H/D for T4 & T5; 850 and 930 mg Val/H/D for T6 & T7; 680 and 740 mg Thr/H/D for T8 & T9. Treatment 10 (All) represents the combined effect of 110% of each added AA (750, 890, 850 and 680 mg/H/D for TSAA, Lys, Val and Thr, respectively). Diets were formulated to be Iso-Caloric. The composition of the formulated diet is illustrated in Table (1).

Average body weight (BW) was obtained by weighing three hens per replicate at the beginning (58 wks of age; initial BW) and at the end (66 wks of age; final BW) of the experimental period. Egg production (EP) and egg weight (EW) were recorded daily while feed intake (FI) was recorded at the end of the experimental period. The daily FI (g/H/D), EW (g/H/D) and hen/day EP percentage were calculated at the end of the experimental period. Egg mass (EM; g/H/D) was calculated by multiplying the number of eggs per hen by the average egg weight during the study. Feed conversion ratio (FCR; g of feed/g of EM) was calculated by using the average FI (g/H/D) divided by EM (g/ H/D).

All eggs produced during three consecutive days from each replicate were collected in the middle (4^{th} week) and the end (8^{th} week) of the experimental period for measuring egg quality. Egg dimension (width and length) were measured using digital caliper. Eggs were weighed and broken out. The following traits were measured:

- Percentage of egg yolk, albumen and shell weight relative to egg weight.
- Egg shape and yolk index (Carter, 1968).
- Shell thickness (mm; including shell membranes).
- Proximate analysis of egg components (A.O.A.C., 1990).

Economical efficiency (EE) and relative economical efficiency (REE) of egg production were calculated according to the experimental diets price, eggs produced and body weight changes at the end of the experimental period.

Data were subjected to analysis of variance (Steel and Torrie, 1980) as a completely randomized design, using the General Linear Models (GLM) procedures of SAS (SAS Institute, 1998). Duncan's multiple range test (Duncan, 1955) was used to determine the differences between treatment means.

RESULTS AND DISCUSSION

Laying hen performance

The effect of experimental treatments on body weight, egg production traits, feed intake and feed conversion ratio are illustrated in Table (2).

1- Body weight (BW):

No significant differences were noticed on the initial BW of the laying hens (Table 2). An increase in final BW was observed with hens fed TSAA (750 & 820 mg), Lys (890 & 970 mg) and Val (850 mg) supplementations compared with the initial BW and the control treatment. However, a reduction in final BW occurred with hens fed Val (930 mg), Thr (680 & 740 mg) and All (110% of all AAs) supplementations. There was a significant difference ($P \leq 0.05$) between the reduction in final BW resulted with Val (930 mg) treatment and either BW of TSAA (750 mg) and Lys (970 mg) treatments. Data stated herein partially agree with some previously published researches. Okazaki et al. (1994) reported that the performance of hens on diets containing Met, Lys or Thr at levels 130 and 160% of NRC was slightly superior to that of 85, 100 or 115% of NRC. In addition, Novak et al. (2004) noted that hen weight gain improved ($P \leq 0.03$) by increased Lys. Increasing TSAA intake up to 689 mg/H/D increased hen weight gain but gain decreased at the highest intake (Novak et al., 2004). In the contrary, Sohail et al. (2002) and Togashi et al. (2002) reported that TSAA had no significant effect on BW. In addition, Amaefule et al. (2004) and El-Husseiny et al. (2005) found that Met had no effect on body weight gain. Similarly, Azazi et al. (2006) noted no significant differences in BW of Bovans brown laying hens fed diets supplemented with Met and Lys. The reduction in final BW observed herein with Val (930 mg) was in contrast with data of Peganova and Eder (2002) who reported that dietary Val concentrations up to 1.06% did not cause any depression in the laying performance. The performance depressant effect of Val (930 mg) might be due to antagonism between Val and the other branched-chain amino acids (isoleucine and leucine).

2- Egg production traits:

Egg production. As shown in Table (2), it is obvious that EP of hens fed dietary TSAA (750 mg), Lys (890 mg), Thr (740 mg) and All (110% of all AAs) were significantly ($P \leq 0.05$) higher than those fed Val (930 mg) supplemented diet. No significant differences were noticed either among the rest of the treatments or between the treatments and the control. Thr (740 mg) and Lys (890 mg) supplementations used in this study resulted in high EP preceded by TSAA (750 mg) treatment.

Egg weight. Similar to the EP data, TSAA (750 mg) treatment resulted in a significantly ($P \leq 0.05$) higher EW than the control, Lys (970 mg) and Thr (740 mg) treatments. Also, TSAA (820 mg), Lys (890 mg), Val (850 & 930 mg), Thr (680 mg) and All (110% of all AAs) treatments presented higher EW than the control but the data failed to show any significant differences. On the other hand, Lys (970 mg) followed by Thr (740 mg) treatments illustrated the lowest EW values in comparison with the rest of the treatments.

Egg mass. TSAA (750 mg) treatment was significantly superior in improving EM over the control, Lys (970 mg) and Val (850 & 930 mg) treatments. This effect was related to the high EP and EW resulted with TSAA (750 mg) treatment. No significant differences in EM were noticed between the control, TSAA (820 mg), Lys (890 mg), Val (850 & 930 mg) and All (110% of all AAs) treatments.

3- Feed Intake (FI) and feed conversion ratio (FCR):

In comparison with the control, TSAA (820 mg) and Thr (740 mg) treatments, hens treated with Lys (970 mg) had the significant lowest FI value followed by Thr (680 mg), Val (850 mg) and TSAA (750 mg) treatments. Dietary amino acids supplementation, TSAA (750 mg) followed by Thr (680 mg), resulted in significantly superior FCR than the control diet. In addition, TSAA (750 mg) treatment showed significantly best FCR than the Val (930 mg) treatment. This result may be attributed to the slight decrease in FI and the increase in EM. No significant differences were noticed either among the rest of the treatments or between these treatments and the control.

Improving EP, EW, EM and FCR by increasing studied amino acids above the NRC (1994) recommendation agreed partially with results of Prochaska et al. (1996), Martinz-Amezcua et al. (1999), Faria et al. (2002), Novak et al. (2004) and Azazi et al. (2006). The current TSAA recommendation (580 mg/H/D) listed in the NRC (1994) may be below than what is required for optimal production performance in laying hens under current condition. The data reported herein (Table 2) showed that TSAA (750 mg/H/D) optimized EP, EW, EM and FCR while FI was minimized. This means that 750 mg/H/D is the optimum dietary level of TSAA for maximizing egg parameters and FCR. Similarly, Zollitsch et al. (1993) and Harms and Russel (1998) stated that increasing TSAA resulted in a positive linear response of hens EW. In consistent to the data stated herein, Roland et al. (1994), Barbosa et al. (1999a), Sohail et al. (2002) and Togashi et al. (2002) reported that increasing TSAA levels in layer diets improved EW, FI, EM and/or EP.

However, Ahmad and Roland (2003) reported little or no effect on EP when various levels of TSAA were fed to laying hens, whereas increasing TSAA level significantly increased EW. In addition, Novak et al.

(2004) showed that although FI and EM were not significantly affected by dietary TSAA, combining the decrease in FI with the increase in EM significantly improved FCR (g of feed /g of EM) as TSAA intake increased. They stated that dietary TSAA level for maximum EP and FCR was near 699 mg/H/D but that of EW may be closer to 779 mg/H/D for phase II. Abd-Elsamee (2005) stated that Met up to 0.55% in hen diet significantly improved EP and EW but FI did not affect compared with Met 0.42%. Azazi et al. (2006) reported that EP, FI, FCR and EM were significantly improved by supplementing Met (0.05 and 0.1%) and Lys (0.12%) to 16.0% CP corn-soybean meal layer diets. The highest EW was obtained with 17.6% CP diet supplemented with 0.05% Met.

On the other hand, the data reported here showed that Lys (890 mg/H/D), Thr (680 mg /H/D) and All (110% of all AAs) treatments resulted in relatively better EW, EP, EM and FCR than the control, Lys (970 mg/H/D), Thr (740 mg/H/D) and Val treatments (Table 2). In partial agreement to that, Scheideler et al. (1996) stated a positive linear response to increasing dietary Lys from 500 to 1000 mg/H/D for all egg production parameters in White Leghorn hens. Maximal EP and EM are maintained with Lys intake of 900 mg/H/D during the later egg production cycle, which agree with our findings. In accordance, Scheideler et al. (1996) and Novak and Scheideler (1998) reported that the current Lys requirement (680 mg/H/D) listed in the NRC (1994) may be lower than what is required for optimal production performance in hens. Authors recommended that Lys intake should be 850 to 900 mg/H/D for optimal EM ,EP and FCR, almost similar to the level used in our study. Novak et al. (2004) also stated that dietary Lys at 816 mg/H/D for phase II optimized EW and FCR while EP was not affected by dietary Lys level. This finding is very close to what was reported by Balnave et al. (2000) who mentioned that maximum EP and minimum FI was obtained by 975 mg Lys. However, Prochaska et al. (1996) reported that overall FI and EP were not affected by increasing dietary Lys from 677 to 1154 or 1613 mg/H/D but significantly increased EW during 42-64 wks of age.

In agreement with present data, Martinz-Amezcua et al. (1999) stated that EP, EW and FCR of hens from 60 to 72 wks of age were significantly improved as Thr level was increased above the NRC (1994) recommended level. In addition, Faria et al. (2002) mentioned that EP and EM were significantly increased with increasing Thr level. Also, Khalifah and Abdalla (2005) illustrated that Thr level from 5.19 to 6.29 g/kg diet had no effect on EP. However, EW and EM were significantly improved with increasing dietary Thr level up to 6.07 g/kg diet (equal to 802 mg/H/D) and then decreased with increasing Thr level. On the contrary, Ishibashi et al. (1998) found that EM and FI increased as Thr level increased and then decreased linearly as dietary Thr increased. Also Schutte (1998) recommended 460 mg/H/D Thr for maximum EM which is below the levels used herein and very close to the NRC (1994) level for Thr.

Valine levels used in this work (Table 2) numerically increased EW and FCR and reduced FI compared with the control treatment. However, EP and EM did not improve. Harms and Russel (2001) reported that EP and EW were increased by Val supplementation. Peganova and Eder (2002) reported that optimum daily EM was reached at 0.685% Val level. Dietary Val concentration up to 1.06% did not cause any depression in the laying performance.

Egg quality measurements

Table (3) shows the effect of supplemental amino acids on external {shell (%), shell thickness (mm) and egg shape index (%)} and internal {albumen (%), yolk (%), albumen yolk ratio and yolk index (%)} egg quality measurements.

1- External egg quality:

Egg shell (%). Some numerical increases in egg shell percentage were noticed as a result of different treatments and resulted in significantly ($P \leq 0.05$) higher values by TSAA (750 & 820 mg), Lys (970 mg), Val (850 & 930 mg) and Thr (740 mg) than the control. However, differences between the different amino acid supplementations were not significant.

Egg shell thickness. No significant differences were noticed in shell thickness either among the different treatments or between the treatments and the control.

Egg shape index (%). Supplementations of TSAA (750 & 820 mg), Val (930 mg), Thr (740 mg) and All (110% of all AAs) resulted in significantly higher egg index values than the control.

2- Internal egg quality:

Egg albumen percentage, yolk percentage and albumen yolk ratio: No significant differences were noticed either between the supplementation treatments or among these treatments and the control in each of the egg albumen percentage, yolk percentage and albumen yolk ratio values.

Egg yolk index: No significant differences were noticed in yolk index between the control and the TSAA, Lys and Thr (680 mg) treatments. However, Val (850 & 930 mg), Thr (740 mg) and All (110% of all AAs) treatments showed significantly lower yolk index values than that of the control.

3- Egg composition analysis

Data in Table (4) show the effect of experimental treatments on egg composition analysis as percentages (egg solids, ether extract, crude protein and ash).

Egg solids (%): It was higher over the control as a result of TSAA (820 mg), Lys (890 mg), Val (850 mg) and Thr (740 mg) treatments. In addition, TSAA (820 mg) treatment resulted in higher egg solids (%) value over TSAA (750 mg) and Val (930 mg) treatments. No significant differences in egg solids values were noticed between the rest of the treatments.

Egg ether extract (%): Supplementation of Lys (970 mg), Thr (740 mg) and All (110% of all AAs) resulted in significantly lower egg ether extract (%) than the control, TSAA (820 mg) and Val (930 mg) treatments. No significant differences in egg ether extract values were noticed between the rest of the treatments.

Egg crude protein (%): Supplementation of Thr (740 mg) significantly increased egg protein (%) than those of the control and the rest treatments. TSAA (750 & 820 mg), Val (930 mg), Thr (680 mg) and All (110% of all AAs) treated hens had numerically higher egg protein (%) than those of the control, Lys, Val (850 mg).

Egg ash (%): Supplementation of TSAA (750 & 820 mg), Lys (890 mg), Val (930 mg) and Thr (740 mg) resulted in numerically higher egg ash (%) than those of the control and the rest of the treatments but the data failed to show any significant differences.

Data reported herein showed that egg shell quality (percentage and thickness) was affected by EW. When EW was increased egg shell quality was decreased, this may be resulted from increased EW without an increase in shell production. Also, increasing the level of studied supplemented amino acids increased egg shell percentage and thickness. Fraser et al. (1998) reported that the foundation of shell consists of protein matrix and it may be possible that increasing the amino acid intake my influence protein synthesis of shell membranes. There have been mixed reports on the effect of TSAA and Lys on egg shell quality. Shafer et al. (1996) noted no effect of TSAA levels on egg shell weight or its percentage when TSAA intake increased from 624 to 822 mg/H/D. Scheideler and Elliot (1998) and Novak et al. (2004) mentioned a similar lack of response of shell percentage to TSAA or Lys intake. On the other hand, Balnave et al. (2000) found that egg shell percentage increased with increasing Lys concentration. They also noted an improvement in egg shell breaking strength as Lys concentration increased to 975 mg/H/D. While Summers et al. (1991) noted that egg size was improved by adding extra Lys, Sohail et al. (1999) stated that egg size did not improve by increasing Lys level.

In the contrary to the data reported herein, Larbier (1973) noted increments in yolk weight as Thr and Lys supplementation increased. Also, Prochaska and Carey (1993) and Prochaska et al. (1996) noticed increases in albumen weight, albumen percentage and albumen protein content with increasing Lys intake. However, yolk protein, total albumen and yolk solids were not significantly affected by Lys addition. Whereas, Scheideler et al. (1996) noted significant increments in egg albumen weight with increasing Lys level from 500 to 1000 mg/H/D at phase II. In agreement with that, Balnave et al. (2000) and Novak et al. (2004) mentioned that wet and dry albumen percentages were increased by the increment in Lys whereas dry yolk percentage was decreased.

The data reported herein showed increments in egg protein, egg solids and egg ash, while egg ether extract was decreased as dietary TSAA, Lys, Val, and Thr increased. Other researchers (Shafer et al., 1996) found a significant increases in albumen & yolk weight and solids when feeding 822 mg TSAA/H/D compared with 624 mg during the later egg production cycle (52 – 60 wks of age). However, Scheideler and Elliot (1998) and Novak et al. (2004) reported no effect of increasing TSAA from 520 to 800 mg/H/D on albumen or yolk percentage, which in consistent to our finding. In the contrary, Shafer et al. (1998) stated increases in albumen component yield, total solids and protein content when hens were supplied with 815 or 849 mg TSAA/H/D compared with 718 mg (partially agreed with our data). In support to that, Novak et al. (2004) noted that hens fed 811 and 699 mg TSAA produced eggs with the greatest yolk solids with no effect on albumen or yolk percentages which agreed with the data reported herein.

Economical efficiency (EE) and relative economical efficiency (REE)

Calculation of the economical efficiency (EE) and the relative economical efficiency (REE) per hen of different formulated diets are summarized in Table (5).

The percentages of the economical efficiency for treatments relative to the control ranged from 90 to 124%. The highest REE (124%) was recorded for TSAA (750 mg) treatment, followed by Thr (680 mg), Lys (890 & 970 mg) and Val (850 mg) treatments. This effect may be due to the high egg numbers and BW gain produced by hen fed these diets. TSAA (820 mg), Thr (740 mg) and All (110% of all AAs) supplementations showed REE comparable to the control diet. The lowest REE (90%) was recorded by hens fed Val (930 mg). This may be due to the low egg numbers produced and the losses of BW. In agreement with that, Khalifah and Abdalla (2005) found that increasing dietary Thr level in layer diet resulted in increase EE and REE, with best values from Thr (6.07 g/kg diet) supplementation. Similarly, Azazi et al. (2006) reported that the addition of Met and Lys improved EE of Bovans brown hens. On the other hand, El-Husseiny et al. (2005) stated that increasing Met in Lohmann brown hen diets had little or no effect on the REE.

Inclusion of TSAA, Lys, Val and Thr or their combination improved Bovans brown laying hen performance and egg quality. This means that amino acids deficiency rather than protein per se may be responsible for low egg parameters and quality. However, there are a lot of differences in the amino acids concentrations used by different researchers for laying hens. These discrepancies may be due to factors affecting the requirements such as strain, age of hens, EP rate, dietary crude protein levels, dietary amino acids levels and/or the environment around the hens.

Table (1): Ingredients and calculated composition of the control basal diet.

Ingredients	%
Yellow corn	62.07
Soybean meal (44% CP)	25.49
Di-calcium phosphate	1.57
Limestone	8.75
Salt	0.30
Vit. and Min. Mixture*	0.30
DL-Methionine	0.13
Vegetable oil	1.39
Total	100.00
Calculated composition:	
ME(Kcal/kg)	2775.20
Crude protein	16.62
Calcium	3.80
Available phosphorus	0.41
Methionine + Cystine (TSAA)	0.680
Lysine	0.810
Valine	0.775
Threonine	0.618

*Each kg of Vit. & Min. Mixture contains: Vit A 4000000 IU, Vit D_3 400000 IU, Vit E 4 g, Vit K_3 400 mg, Vit B_1 400 mg, Vit B_2 2 g, Vit B_6 600 mg, Vit B_{12} 4 mg, Pantothenic acid 4 g, Niacin 12 g, Biotin 20 mg, Folic acid 400 mg, Choline chloride 240 g, Iron 12 g, Manganese 24 g, Zinc 20 g, Copper 2 g, Cobalt 40 mg, Iodine 120 mg and Selenium 40 mg. $CaCo_3$ was used as carrier.

Table (2): Effect of dietary TSAA, lysine, valine and threonine on performance measurements of Bovans brown layer hens (mean ± SE).

T #	Treatment description (mg/H/D)	Body weight (g)		Egg			Feed	
		Initial	Final	production (%/H)	weight (g/H/D)	mass (g/H/D)	intake (g/H/D)	conversion (g feed/g EM)
1	*Control*[1]	1845±2.96	1841±34.4^{abc}	87.63±2.86^{ab}	64.94±1.12^{bc}	56.91±2.20^{bc}	111.4±0.38^{a}	1.97±0.07^{a}
	TSAA:							
2	750	1850±9.79	1899±25.2^{ab}	91.25±1.54^{a}	67.38±1.34^{a}	61.49±1.67^{a}	107.7±0.92^{ab}	1.76±0.04^{c}
3	820	1845±1.85	1860±46.8^{abc}	89.11±1.60^{ab}	66.19±0.52^{ab}	58.97±1.02^{ab}	111.9±0.51^{a}	1.90±0.04^{abc}
	Lysine:							
4	890	1849±1.56	1889±16.1^{abc}	90.55±1.40^{a}	65.74±0.23^{ab}	59.53±0.81^{ab}	109.7±0.88^{ab}	1.85±0.04^{abc}
5	970	1747±2.95	1910±39.7^{a}	87.04±1.17^{ab}	63.23±0.37^{c}	55.03±0.66^{c}	105.0±2.53^{b}	1.91±0.04^{abc}
	Valine:							
6	850	1849±2.57	1882±34.7^{abc}	87.21±1.62^{ab}	66.41±1.04^{ab}	57.93±1.55^{bc}	107.2±2.42^{ab}	1.85±0.04^{abc}
7	930	1846±4.31	1789±27.1^{c}	85.29±0.98^{b}	65.61±0.36^{ab}	55.97±0.84^{bc}	108.1±3.11^{ab}	1.93±0.07^{ab}
	Threonine:							
8	680	1841±2.75	1803±17.5b^{c}	89.56±0.12^{ab}	66.03±0.44^{ab}	59.14±0.40^{ab}	106.3±2.28^{ab}	1.80±0.05^{bc}
9	740	1844±2.99	1827±15.0^{abc}	91.23±0.97^{a}	64.98±0.18^{bc}	59.27±0.72^{ab}	110.8±1.31^{a}	1.87±0.02^{abc}
10	*All*[2]	1845±4.32	1836±20.6^{abc}	90.06±0.36^{a}	65.07±0.04^{abc}	58.60±0.22^{ab}	109.0±1.13^{ab}	1.86±0.02^{abc}
	Overall means	1846±1.24	1854±10.3	88.89±0.51	65.56±0.26	58.28±0.43	108.7±0.61	1.87±0.02

a-c Numbers with different superscript in each column are significantly different at $P \le 0.05$.

[1] Contains 680, 810, 775 and 618 mg/H/D of TSAA, Lys, Val and Thr, respectively.

[2] Contains 750, 890, 850 and 680 mg/H/D of TSAA, Lys, Val and Thr, respectively.

Table (3): Effect of dietary TSAA, lysine, valine and threonine on egg quality measurements of Bovans brown layer hens (mean ± SE).

T #	Treatment description (mg/H/D)	External egg quality			Internal egg quality			
		Shell (%)	Shell thickness (mm)	Shape Index (%)	Albumen (%)	Yolk (%)	Albumen Yolk ratio	Yolk index (%)
1	*Control*[1]	10.20±0.12[b]	0.50±0.01	74.74±0.23[c]	63.45 ± 0.49	26.35±0.45	2.42±0.07	59.45±0.59[a]
	TSAA:							
2	750	10.99±0.21[a]	0.50±0.01	76.53± 0.31[ab]	62.51 ± 0.59	26.50±0.66	2.37±0.08	48.86±0.34[ab]
3	820	11.04±0.18[a]	0.52±0.01	77.45± 0.63[a]	61.93 ± 0.33	27.04±0.27	2.30±0.03	49.74±0.74[ab]
	Lysine:							
4	890	10.62±0.14[ab]	0.49±0.01	76.29± 0.35[abc]	63.36 ± 0.89	26.03±1.00	2.46±0.13	48.53±0.93[ab]
5	970	10.83±0.11[a]	0.49±0.01	75.67± 0.63[bc]	63.61 ± 0.16	25.56±0.24	2.50±0.03	48.77±0.75[ab]
	Valine:							
6	850	10.97±0.18[a]	0.50±0.01	76.27± 0.60[abc]	63.11 ± 0.41	25.93±0.50	2.44±0.06	47.89±0.61b
7	930	11.02±0.21[a]	0.51±0.01	76.72± 0.33[ab]	62.91 ± 0.21	26.08±0.28	2.42±0.03	47.84±0.41[b]
	Threonine:							
8	680	10.70±0.30[ab]	0.49±0.02	75.95± 0.64[abc]	62.91 ± 0.61	26.40±0.57	2.39±0.07	49.12±0.31[ab]
9	740	10.91±0.10[a]	0.50±0.01	76.34± 0.54[ab]	62.69 ± 0.89	26.41±0.97	2.40±0.13	47,81±0.96[b]
10	*All*[2]	10.59±0.17[ab]	0.49±0.01	76.45± 0.29[ab]	63.55 ± 0.45	25.86±0.28	2.47±0.04	48.05±0.84[b]
	Overall means	10.79±0.07	0.50±0.003	76.25± 0.17	63.00 ± 0.17	26.22±0.18	2.42±0.02	48.70±0.23

[a-c] Numbers with different superscript in each column are significantly different at $P \le 0.05$.

[1] Contains 680, 810, 775 and 618 mg/H/D of TSAA, Lys, Val and Thr, respectively

[2] Contains 750, 890, 850 and 680 of TSAA, Lys, Val and Thr, respectively

Table (4): Effect of dietary TSAA, lysine, valine and threonine on egg composition analysis of Bovans brown layer hens (mean ± SE).

T #	Treatment description (mg/H/D)	Egg composition (%)			
		Solids	Crude fat	Crude protein	Ash
1	*Control*[1]	23.19 ± 0.39^{c}	12.27 ± 0.18^{ab}	11.18 ± 0.20^{b}	**1.12 ± 0.05**
	TSAA:				
2	750	24.02 ± 0.48^{bc}	11.83 ± 0.19^{bcd}	11.65 ± 0.11^{b}	**1.15 ± 0.03**
3	820	25.74 ± 0.39^{a}	12.39 ± 0.09^{a}	11.67 ± 0.15^{b}	**1.17 ± 0.02**
	Lysine:				
4	890	24.82 ± 0.14^{ab}	12.15 ± 0.07^{abc}	11.39 ± 0.13^{b}	**1.16 ± 0.04**
5	970	24.28 ± 0.18^{abc}	11.69 ± 0.04^{cd}	11.45 ± 0.25^{b}	**1.06 ± 0.03**
	Valine:				
6	850	24.90 ± 0.72^{ab}	11.98 ± 0.08^{abcd}	11.37 ± 0.31^{b}	**1.12 ± 0.03**
7	930	24.02 ± 0.27^{bc}	12.45 ± 0.07^{a}	11.70 ± 0.07^{b}	**1.16 ± 0.08**
	Threonine:				
8	680	24.42 ± 0.76^{abc}	11.83 ± 0.28^{bcd}	11.76 ± 0.37^{b}	**1.11 ± 0.08**
9	740	25.25 ± 0.65^{ab}	11.64 ± 0.11^{d}	12.51 ± 0.09^{a}	**1.15 ± 0.02**
10	*All*[2]	24.19 ± 0.24^{abc}	11.78 ± 0.12^{cd}	11.51 ± 0.22^{b}	**1.09 ± 0.03**
	Overall means	**24.48 ± 0.17**	12.00 ± 0.06	**11.62 ± 0.08**	1.13 ± 0.01

a-d Numbers with different superscript in each column are significantly different at $P \leq 0.05$.

[1] Contains 680, 810, 775 and 618 mg/H/D of TSAA, Lys, Val and Thr, respectively.

[2] Contains 750, 890, 850 and 680 mg/H/D of TSAA, Lys, Val and Thr, respectively.

Table (5): Effect of dietary TSAA, lysine, valine and threonine on economical efficiency of Bovans brown layer hens.

T #	Treatment description (mg/H/D)	Feed			Eggs		Body weight		Total revenue (L.E)[5]	Net revenue /H (L.E)[6]	EE	REE[7] (%)
		Total intake (kg/H)	Price /kg (L.E)	Total cost (L.E)	Total number /H	Total price/H (L.E)[3]	BW Change (kg/H)	Total price (L.E)[4]				
1	*Control*[1]	6.240	1.336	8.336	49.07	14.721	-0.004	-0.036	14.685	6.749	0.76	100
	TSAA:											
2	750	6.028	1.348	8.123	51.10	15.330	0.049	0.441	15.771	7.649	0.94	124
3	820	6.265	1.359	8.514	49.90	14.970	0.015	0.135	15.105	6.591	0.77	102
	Lysine:											
4	890	6.143	1.347	8.272	50.71	15.213	0.040	0.360	15.673	7.301	0.88	116
5	970	5.881	1.357	7.982	48.74	14.622	0.062	0.558	15.180	7.198	0.90	118
	Valine:											
6	850	6.005	1.346	8.085	48.84	14.652	0.033	0.297	14.949	6.864	0.85	112
7	930	6.051	1.357	8.210	47.77	14.331	-0.058	-0.522	13.809	5.560	0.68	90
	Threonine:											
	680	5.952	1.344	7.999	50.15	15.045	0.038	0.342	15.387	7.388	0.92	121
9	740	6.207	1.352	8.393	51.09	15.327	-0.017	-0.153	15.174	6.781	0.81	106
10	*All*[2]	6.104	1.377	8.403	50.43	15.129	-0.010	-0.090	15.039	6.636	0.79	104

[1] Contains 680, 808, 736 and 618 mg/H/D of TSAA, Lys, Val and Thr, respectively.
[2] Contains 750, 890, 854 and 680 mg/H/D of TSAA, Lys, Val and Thr, respectively.
[3] Price of each egg = 0.30 L.E. at the time of experimental period [4] Price of 1 kg live body weight = 9.00 L.E.
[5] Total revenue = total price of eggs/hen ± price of live body weight change/hen
[6] Net revenue = total revenue - total feed cost/hen
[7] Relative economical efficiency (%) assuming the control treatment = 100%.

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الملخص العربي

تأثير إضافة مستويات غذائية من الأحماض الأمينية الكبريتية والليسين والڤالين والثريونين على أداء الدجاج البياض وقياسات جودة البيضة

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أستخدمت في هذه الدراسة ١٢٠ دجاجة بياضة من سلالة بوفانز البني (عمر ٥٨ اسبوعا) لتقييم تأثير إضافة مستويات غذائية من الأحماض الأمينية الكبريتية (مضافة فى صورة مثيونين) والليسين والڤالين والثريونين على اداء الدجاج البياض وقياسات جودة البيضة.

وزعت الدجاجات عشوائيا في ١٠ معاملات كل معاملة ١٢ دجاجة موزعة على 4 مكررات. تم تقديم الغذاء والماء بحرية حتى الشبع مع ١٦ ساعة إضاءة في اليوم. تم تجهيز عليقة قاعدية من الذرة وفول الصويا بحيث تفي بالإحتياجات الغذائية طبقا لتوصيات دليل تربية السلالة وأستخدمت كعليقة مقارنة (الكنترول؛ معاملة ١: وتحتوي على ٦٨٠ و٨١٠ و٧٧٥ و٦١٨ مجم/دجاجة/يوم من الأحماض الأمينية الكبريتية والليسين والڤالين والثريونين على التوالي) . تم إضافة الأحماض الأمينية الكبريتية (فى صورة مثيونين) والليسين والڤالين والثريونين إلى عليقة الكنترول بنسبة ١١٠ و١٢٠% من الإحتياجات الغذائية لهذه الأحماض الأمينية لكل حامض أمينى لتكون المعاملات من ٢ الى ٩

كما يلى:

معاملة ٢ و ٣ وتحتوي على ٧٥٠ و٨٢٠ مجم/دجاجة/يوم من الأحماض الأمينية الكبريتية على التوالي.

معاملة ٤ و ٥ وتحتوي على ٨٩٠ و٩٧٠ مجم/دجاجة/يوم من الليسين على التوالي.

معاملة ٦ و٧ وتحتوي على ٨٥٠ و٩٣٠ مجم/دجاجة/يوم من الڤالين على التوالي.

معاملة ٨ و ٩ وتحتوي على ٦٨٠ و٧٤٠ مجم/دجاجة/يوم من الثريونين على التوالي.

معاملة ١٠ وتمثل التأثير التجميعي للمستوى ١١٠ % من كل حامض أميني مضاف (تحتوي على ٧٥٠ و٨٩٠ و٨٥٠ و٦٨٠ مجم/دجاجة/يوم من الأحماض الأمينية الكبريتية والليسين والڤالين والثريونين على التوالي).

تم قياس وزن الجسم النهائي وإنتاج البيض ووزن البيض وكتلة البيض والغذاء الماكول والكفاءة التحويلية للغذاء وجودة وتحليل تركيب البيضة.

وقد أوضحت النتائج أن :

مستوى الأحماض الأمينية الكبريتية ٧٥٠ مجم/دجاجة/يوم حسن وزن البيض وإنتاج البيض وكتلة البيض والكفاءة التحويلية للغذاء مع تقليل الغذاء المأكول. وهذا يعني أن ٧٥٠ مجم/دجاجة/يوم من الأحماض الأمينية الكبريتية هو المستوى الأمثل لتعظيم قياسات البيض والكفاءة التحويلية للغذاء.

كما نتج عن إضافة الليسين ٨٩٠ مجم/دجاجة/يوم والثريونين ٦٨٠ مجم/دجاجة/يوم ومعاملة التأثير التجميعي للأحماض الأمينية (معاملة ١٠) تحسن نسبي في وزن البيض وإنتاج البيض وكتلة

البيض والكفاءة التحويلية للغذاء مقارنة بالكنترول والليسين ٩٧٠ مجم/دجاجة/يوم والثريونين ٧٤٠ مجم/دجاجة/يوم والفالين بمستوييه.

أيضاً نتج عن إضافة الفالين ٩٣٠ مجم/دجاجة/يوم والثريونين ٦٨٠ مجم/دجاجة/يوم إنخفاض معنوي في وزن الجسم النهائي مقارنة بالليسين ٩٧٠ مجم/دجاجة/يوم والأحماض الأمينية الكبريتية ٧٥٠ مجم/دجاجة/يوم والتي أدت إلى زيادة وزن الجسم النهائي.

لم يتأثر سمك قشرة البيضة بأي من المعاملات بينما زادت النسبة المئوية للقشرة معنويا بزيادة الأحماض الأمينية الكبريتية ٧٥٠ و٨٢٠ مجم/دجاجة/يوم والليسين ٩٧٠ مجم/دجاجة/يوم والفالين ٨٥٠ و٩٣٠ مجم/دجاجة/يوم والثريونين ٧٤٠ مجم/دجاجة/يوم مقارنة بالكنترول.

لم توجد أي فروق معنوية سواء بين المعاملات وبعضها او بين المعاملات والكنترول لكلا من النسبة المئوية للألبيومين والنسبة المئوية للصفار ونسبة الألبيومين إلى الصفار. زادت نسبة البروتين والجوامد والرماد للبيضة بينما قلت نسبة مستخلص الأثير فى البيضة بزيادة الأحماض الأمينية الكبريتية والليسين والفالين والثريونين. سجلت الأحماض الأمينية الكبريتية ٧٥٠ مجم/دجاجة/يوم أعلى كفاءة إقتصادية نسبية (١٢٤%) بينما سجل الفالين أقل كفاءة إقتصادية نسبية (٩٠%).

أوصت النتائج أن إضافة الأحماض الأمينية إلى علائق الذرة وفول الصويا حسن من أداء الدجاج البياض وقياسات جودة البيضة.