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Influence of Different Levels of Certain Essential Amino Acids on the Performance, Egg Quality Criteria and Economics of Lohmann Brown Laying Hens

[1]M. Alagawany and [1]Kh. M. Mahrose
[1]Department of Poultry, Faculty of Agriculture, Zagazig University, Zagazig, Egypt
[2]University of Zimbabwe, Harare, Zimbabwe

Corresponding Author: M. Alagawany, Department of Poultry, Faculty of Agriculture, Zagazig University, Zagazig, Egypt

ABSTRACT

Improving productivity, reducing feed cost or increasing economical efficiency are main challenges in laying hen farms. Thus, the present experiment was conducted to study the effect of different levels of lysine and Total Sulfur Amino Acids (TSAA) on productive performance, egg quality and economical evaluation of Lohmann Brown laying hens. A total numbers of 144 hens aged from 34-50 weeks of age, were randomly divided into 9 experimental groups of 16 hens each. Each group was sub-divided into four replicates (4 hens per each). The experimental design was a 3×3 factorial arrangement with 3 levels of total lysine (0.74, 0.84 and 0.94%) and 3 levels of TSAA (0.67, 0.72 and 0.77%). Lohmann Brown hens consumed different levels of lysine, TSAA and their interaction showed non-significant differences in final body weight and body weight change at the end of experimental period. Feed consumption for hens fed 0.74 and 0.94% lysine significantly increased vs. those received 0.84% lysine diet during the experimental periods (34-38 and 38-42 weeks of age). Feeding different levels of lysine, TSAA and their interaction showed insignificant differences in productive performance criteria during the experiment. Egg quality was not affected by lysine levels except albumen percentage, Unit Surface Shell Weight (USSW) and yolk:albumen at 42 weeks as well as USSW and yolk diameter at 50 weeks of age. Egg market price, net revenue and economical feasibility were affected with different levels of lysine and TSAA, whereas, 0.84 and 0.72% of TSAA recorded the highest economical evaluation values, respectively. It can be conducted that, using 0.84% lysine and 0.72% TSAA was better to get the best productive performance and economical efficiency of Brown Lohmann hens throughout the production period of 18-34 weeks of age.

Key words: Layer, lysine, TSAA, productive performance, feed utilization, egg quality

INTRODUCTION

Synthetic lysine and methionine became available for poultry diets for over 60 years. This allowed poultry producers to formulate diets based on amino acids to match the bird's nutritional requirements, thereby reducing waste as well as cost. Feed and egg prices are the two major factors influencing profits but many producers did not alter the method of feed formulation or diets, given the changing dynamics of feed and egg prices. In order to establish a feeding program, one of the first things needed was to reconfirm that diets formulated based on amino acids were the most economically feasible. Hens fed rich in protein diets produced heavier eggs than hens fed diets formulated based on lysine (Sohail and Roland, 1997; Schutte *et al.*, 1988).

Efficiency of protein utilization in a diet depends on the amount, composition and digestibility of the amino acids that are contained therein. Methionine and lysine are generally the first and the next limiting amino acids in corn-soybean diets for laying hens (Liu *et al.*, 2005). Moreover, methionine is considered the first limiting amino acid in low protein corn-soybean meal diet for laying hens. Many studies have reported that the efficiency of protein utilization is increased by supplementation of methionine and lysine (Schutte *et al.*, 1994; Novak *et al.*, 2004). If one or two essential amino acids are deficient, the excess amino acids will be wasted as nitrogenous excretion. To use amino acids, efficiently the crystalline amino acids such as methionine and lysine can be added for balance. Liu *et al.* (2005) and Wu *et al.* (2005) reported that increasing lysine or TSAA had a positive effect on the performance of laying hens. It is important to know the TSAA and lysine requirements of laying hens. There are contradicting results in TSAA requirements of laying hens. NRC (1994) reported that white-egg laying hens with 100 g daily feed consumption required 300 and 580 mg methionine and TSAA in the diets daily per hen, respectively. Similarly, Rostango (1990) suggested that hens with daily feed consumption of 105 g required 327 methionine and 595 mg TSAA daily, respectively. Ahmad *et al.* (1997) reported that TSAA of 580-660 mg per hen per day had no effect on the performance of laying hens. However, Schutte *et al.* (1994) reported that the requirement for TSAA were ca. 740 mg per day per hen, of which ca. 440 mg was methionine. Cao *et al.* (1992) also reported that the requirement of methionine and TSAA were 424 and 785 mg per day per hen, respectively. In addition, there were differences in TSAA requirements estimated from different production parameters. Novak *et al.* (2004) reported that dietary TSAA levels for maximum egg production were 811 mg per day per hen while TSAA for feed efficiency was 699 mg per day per hen. To optimize profits, one must switch diets and the method of formulation as feed and egg prices and environmentally-linked (Roland *et al.*, 2000), many researchers reported that adding synthetic methionine to low protein diets was economical (Johnson and Fisher, 1958; Combs, 1962; Harms and Miles, 1988; Waldroup and Hellwig, 1995).

The present study, therefore, was designed to assess the effect of dietary lysine and TSAA and their interactions on egg production, feed utilization, egg quality and their economics in Lohmann Brown-egg type laying hens from 34-50 weeks of age.

MATERIALS AND METHODS

The present experiment was carried out at the Poultry Research Farm, Department of Poultry, Faculty of Agriculture, Zagazig University, Egypt. All experimental procedures were carried out according to the Local Experimental Animal Care Committee and approved by the ethics of our institutional committee of Department of Poultry, Faculty of Agriculture, Zagazig University, Zagazig, Egypt.

Animals and diets: One hundred forty-four Lohmann Brown hens at 34 week old were weighed individually, then randomly assigned and housed in wire laying cages until the end of the experiment. Birds were allotted into nine treatment groups each with four replicates (each group contains 16 hens). Hens were housed in conventional type cages with feed and water provided for *ad libitum* consumption. Hens were maintained on a 17-7 h light-dark cycle throughout the trial. Cage dimensions were 25×40×50 cm, equal (2,000 cm^2) to floor space. Vaccination and medical program were done according to the different stages of age under supervision of a licensed veterinarian. The composition and chemical analysis of the experimental diets are shown in Table 1.

Table 1: Chemical composition of the experimental diets

	Lysine levels (%)								
	0.74			0.84			0.94		
	TSAA levels (%)			TSAA levels (%)			TSAA levels (%)		
Parameters	0.67	0.72	0.77	0.67	0.72	0.77	0.67	0.72	0.77
Ingredients (%)									
Yellow corn	56.00	56.00	56.00	56.00	56.00	56.00	56.00	56.00	56.00
Soybean meal 44 (%)	15.60	15.60	15.60	15.50	15.50	15.54	15.60	15.60	15.60
Corn gluten meal 62 (%)	8.00	8.00	8.00	8.009	7.98	7.94	8.05	7.97	7.87
Wheat bran (%)	7.71	7.66	7.609	7.60	7.585	7.56	7.49	7.519	7.54
Cotton seed oil	2.00	2.00	2.00	2.00	2.00	2.08	2.00	2.00	2.00
Salt	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.30
Vit mineral premix[1]	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.30	0.30
Dicalcium phosphate	1.80	1.80	1.80	1.80	1.80	1.80	1.80	1.80	1.80
Limestone	8.21	8.21	8.21	8.30	8.30	8.30	8.20	8.20	8.20
Lysine	0.06	0.06	0.06	0.17	0.14	0.14	0.24	0.24	0.26
Dl-Methionine	0.02	0.071	0.121	0.021	0.071	0.13	0.02	0.071	0.13
Total	100.00	100.00	100.00	100.00	100.00	100.00	100.00	100.00	100.00
Chemical analysis[2]									
Crude protein	18.49	18.26	17.98	18.55	18.45	18.66	18.50	18.50	18.24
Ether extract	4.95	40.40	4.33	4.16	4.66	4.74	4.93	4.855	5.93
Crude fiber	3.30	0 3.20	2.91	3.00	2.80	2.92	3.04	3.12	3.20
Calculated composition[3]									
ME, Kcal/kg	2785.00	2785.00	2785.00	2802.00	2800.00	2808.00	2790.00	2790.00	2792.00
Crude protein (%)	18.00	18.00	18.00	18.07	18.08	18.03	18.03	18.03	18.02
Calcium (%)	3.64	3.64	3.64	3.67	3.67	3.67	3.65	3.65	3.65
Nonphytate P (%)	0.45	0.45	0.45	0.45	0.45	0.45	0.45	0.45	0.45
Lysine (%)	0.74	0.74	0.74	0.84	0.84	0.84	0.94	0.94	0.94
TSAA (%)	0.67	0.72	0.77	0.67	0.72	0.77	0.67	0.72	0.77
Total EAA	7.33	7.33	7.33	8.31	8.31	8.31	9.27	9.27	9.27
EAA% of CP	45.81	45.81	45.81	46.16	46.16	46.16	46.35	46.35	46.35
TSAA of % EAA	9.14	9.82	10.50	8.06	8.66	9.26	7.22	7.76	8.30
Price/ton diet, L.E[4]	2445.95	2465.20	2484.45	2467.31	2475.00	2509.81	2475.56	2510.56	2545.56

[1]Layer vitamin-minerals premix: Each 1 kg consists of Vit. A: 8000 IU, Vit. D_3: 1300 ICU, Vit. E: 5 mg, Vit. K: 2 mg, Vit. B_1: 2 mg, Vit. B_1: 0.7 mg, Vit. B_2: 3 mg, Vit. B_6: 1.5 mg, Vit. B_{12}: 7 mg, biotin 0.1 mg, Pantothenic acid: 6 g, Niacin: 20 g, Folic acid: 1 mg, Manganese: 60 mg, Zinc: 50 mg, Copper: 6 mg, Iodine: 1 mg, Selenium: 0.5 mg, Cobalt: 1 mg, [2]Chemical analysis according to AOAC (2005), [3]Calculated according to NRC (1994), [4]Caculated according to the price of feed ingredients when the experiment was started

Experimental design: Hens were randomly assigned to a 3×3 factorial design experiment, including three levels of total lysine (0.74, 0.84 and 0.94%) and three levels of total sulphur amino acids (0.67, 0.72 and 0.77%).

Experimental diets: The experimental diets were formulated to meet NRC (1994) nutrient recommendations. Diets were also supplemented with lysine to achieve the planned lysine levels and supplemented with DL-methionine to supply the required sulphur amino acids.

Data collection and calculation: Body weight was obtained individually at the initial 34 week of age and final 50 week of age. Body weight changes were calculated as the difference between the initial and the final body weights was obeserved. Feed consumption was recorded biweekly; the feed efficiency ratio (g egg/g feed) was calculated as the egg mass value divided by the amount of feed consumed. Egg weight and egg number were recorded daily to calculate the egg mass (g per hen per period studied). Egg mass was subsequently calculated.

Chemical analysis: Samples of diets were analyzed for their content of nitrogen by the Kjeldahl method 984.13 (AOAC, 2005) and the protein contents were calculated using the multiplication factor of 6.25, ether extract by the method 920.39A (AOAC, 2005) and crude fiber by the method 978.10 (AOAC, 2005).

Egg quality criteria: Egg components were measured using 5 eggs from each treatment replicate monthly during the experimental period. Egg length, width and weight were measured. The yolks were separated from albumen and the shell of the egg was cleaned of any adhering albumen. Egg components were expressed as percentage of egg weight. Egg shape index was computed by the ratio of egg width to the length (Awosanya *et al*., 1998). Yolk index was calculated according to Funk *et al*. (1958), as yolk height divided by yolk diameter (mm):

$$\text{Unit Surface Shell Weight (USSW)} = \frac{\text{Egg weight (mg)}}{\text{Egg surface area (cm}^2\text{)}}$$

where, egg surface area (S) in cm^2 is $3.9782W^{0.75056}$ and W is egg weight (mg). Haugh units were calculated according to Eisen *et al*. (1962) by using the following equation:

$$\text{Haugh Units (HU)} = 100 \log (H+7.57-1.7\ W^{0.37})$$

where, H is height of the albumen (mm) and W is egg weight (g). Egg quality measurements were determined according to Romanoff and Romanoff (1963) method. Egg shell thickness was measured in µm using a micrometer.

Economical analysis: The economical efficiency of egg production was calculated from the input-output analysis and calculated according to the price of the experimental diets and egg production during the year, 2013. The values of economical efficiency were calculated as the net revenue per unit of total cost (Zeweil *et al*., 1996).

Statistical analysis: Data was statistically analyzed on a 3×3 factorial design basis according to Snedecor and Cochran (1982) using the following model:

$$Y_{ijk} = \mu+A_i+S_j+AS_{ij}+e_{ijk}$$

where, Y_{ijk} the is observation, µ is the overall mean, A_i is effect of lysine level (i = 0.74, 0.84 and 0.94%), S_j is the effect of TSAA level (j = 0.67, 0.72 and 0.77%), AS_{ij} is the interaction effect between levels of lysine and TSAA level (j = 1, 2..... and 9) and $e_{(ijk)}$ is random error. Differences among means within the same factor were tested using Duncan's New Multiple Range test (Duncan, 1955).

RESULTS AND DISCUSSION

Live body weight and body weight changes: Lohmann Brown hens consuming different levels of lysine, TSAA and their interaction showed non-significant differences in final body weight at the end of experimental period (Table 2). These results might be due to the adequacy of low lysine and TSAA in the diets to maintain body weight. Similar results were reported by Hassan *et al.* (2003), Abdalla *et al.* (2005) and Zeweil *et al.* (2011) who stated that insignificant difference in the overall mean body weight for hens fed different methionine levels. Bouyeh and Gevorgian (2011) found that various levels of lysine and methionine had no effect on body weight of laying hens after peak production. In contrast, Narvaez-Solarte *et al.* (2005) reported that Leghorn hens fed 0.484% of TSAA had the lowest body weight gain, while hens fed 0.684% had the highest body weight change during the period from 22 up to 38 weeks of age.

Feed utilization: Results in Table 3 indicated that feed consumption during the experimental periods 34-38 and 38-42 weeks of age. The feed intake of hens in periods 38-38 and 38-42 weeks of age were significantly greater ($p\leq 0.05$) for hens fed the low (0.74%) and high (0.94%) as compared to 0.84% of lysine. However, the reverse was occurred at the period 42-46 weeks, where the 0.84% lysine level resulted in significantly ($p\leq 0.05$) higher feed intake as compared to the low or high levels. At the period from 42-46 and 46-50 weeks of age, feed consumption significantly ($p\leq 0.05$) increased with decreasing TSAA level. Contrary, dietary TSAA had no effect on feed consumption during the other study periods (Table 3).

Table 2: Live body weight and body weight change of Lohmann laying hens as affected by lysine, total sulfur amino acids levels and their interaction

Items	Initial body weight (g h^{-1}) At 34 week of age	Final body weight (g h^{-1}) At 50 week of age	Change of body weight (g) At 50 week of age
Lysine (%)			
0.74	1796.87	1859.85	62.98
0.84	1778.45	1828.52	50.066
0.94	1826.91	1765.41	-61.50
TSAA (%)			
0.67	1805.37	1862.10	56.73
0.72	1780.12	1793.60	13.48
0.77	1816.75	1798.08	-18.67
Lysine×TSAA (%)			
0.74×0.67	1824.93	1969.99	145.06
0.74×0.72	1770.43	1765.41	-5.02
0.74×0.77	1775.25	1844.16	48.91
0.74×0.67	1754.55	1761.33	7.20
0.84×0.72	1773.00	1875.83	102.83
0.84×0.77	1808.25	1848.41	40.16
0.84×0.67	1837.06	1854.99	17.93
0.94×0.72	1796.93	1739.58	-57.35
0.94×0.77	1846.75	1701.66	-145.08
SEM	17.65	18.88	78.61
Two-way ANOVA (probability)			
Lysine	0.5092	0.3972	0.7231
TSAA	0.4940	0.6041	0.2432
Lysine×TSAA	0.9121	0.6219	0.1772

Means in the same column within each classification bearing different letters are significantly different ($p<0.05$ or 0.01)

Table 3: Feed consumption of Lohmann laying hens as affected by lysine, total sulfur amino acids levels and their interaction during the experimental periods

	Feed consumption (g h^{-1} day^{-1})				
Items	34-38 week	38-42 week	42-46 week	46-50 week	34-50 week
Lysine (%)					
0.74	94.62^a	89.42^a	102.88^b	113.35	100.07
0.84	84.57^b	87.59^b	105.31^a	111.27	97.19
094	98.61^a	90.97^a	101.12^b	111.19	100.47
TSAA (%)					
0.67	94.97	88.23	105.77^a	116.59^a	101.39
0.72	90.36	90.23	102.68^{ab}	110.33^b	98.47
0.77	92.20	89.52	100.86^b	108.90^b	97.87
Lysine×TSAA (%)					
0.74×0.67	95.72	88.97	103.39	118.31	101.60
0.74×0.72	92.85	91.19	102.56	114.51	100.28
0.74×0.77	95.28	88.13	102.69	107.22	98.33
0.74×0.67	88.43	85.85	109.25	115.36	99.72
0.84×0.72	80.11	87.61	104.15	105.47	94.09
0.84×0.77	85.18	89.30	103.53	112.99	97.75
0.84×0.67	100.76	89.89	104.67	116.10	102.85
0.94×0.72	98.93	91.90	102.33	111.00	101.04
0.94×0.77	96.15	91.13	96.36	106.48	97.53
SEM	3.18	1.12	1.93	3.60	1.78
Two-way ANOVA (probability)					
Lysine	0.0029	0.0017	0.0245	0.5602	0.7784
TSAA	0.2228	0.1362	0.0354	0.0445	0.5758
Lysine×TSAA	0.5747	0.2382	0.2425	0.6657	0.6871

Means in the same column within each classification bearing different letters are significantly different ($p<0.05$)

The higher level of dietary lysine had significantly ($p\leq0.05$) the higher values of feed efficiency during 42-46 weeks of age compared to other periods (Table 4), whereas feed efficiency ratio in this period was significantly ($p\leq0.05$) increased gradually with increasing dietary lysine from 0.74-0.84 and 0.94% for Lohmann hens. The improvement of feed consumption and feed efficiency ratio may be due to the increase of egg mass and might be attributed to more balanced amino acids. The results obtained coincided with Bateman *et al.* (2008) who found that lysine had an increased ($p\leq0.05$) effect on feed utilization at 26 and 27 weeks of age, whereas feed consumption accentuated when as the lysine level was increased from 0.79-0.97%. On the contrary, Prochaska *et al.* (1996) demonstrated that incorporation of supplemental lysine had no significant effect on feed consumption of hens fed diets containing 0.72, 0.89 and 1.15% lysine.

The feed efficiency ratio of laying hens was not significantly affected by increasing dietary TSAA except over the experimental period from 46-50 weeks-old, where the best value of feed efficiency ratio (0.397 g egg/g feed) was achieved with hens consuming the intermediated level of TSAA versus other levels during this period. Our results coincided with those found by Novak *et al.* (2004) who reported that feed efficiency was not significantly improved with increased TSAA intake during the whole experimental period which is in accordance with other research reports (Bertram and Schutte, 1992; Baiao *et al.*, 1999; Bateman *et al.*, 2008).

Table 4: Feed efficiency ratio of Lohmann laying hens as affected by lysine, total sulfur amino acids levels and their interaction during the experimental periods

	Feed efficiency (g egg/g feed)				
Items	34-38 week	38-42 week	42-46 week	46-50 week	34-50 week
Lysine (%)					
0.74	0.485	0.432	0.364[c]	0.337	0.405
0.84	0.536	0.413	0.385[b]	0.363	0.424
094	0.485	0.381	0.420[a]	0.389	0.419
TSAA (%)					
0.67	0.466	0.414	0.391	0.334[c]	0.4021
0.72	0.530	0.416	0.379	0.397[a]	0.431
0.77	0.510	0.396	0.399	0.359[b]	0.416
Lysine×TSAA (%)					
0.74×0.67	0.447	0.472	0.352	0.317	0.397
0.74×0.72	0.521	0.429	0.370	0.365	0.423
0.74×0.77	0.488	0.394	0.371	0.329	0.396
0.74×0.67	0.469	0.376	0.376	0.296	0.379
0.84×0.72	0.552	0.401	0.362	0.431	0.436
0.84×0.77	0.588	0.464	0.416	0.363	0.457
0.84×0.67	0.482	0.395	0.446	0.387	0.429
0.94×0.72	0.517	0.419	0.405	0.396	0.435
0.94×0.77	0.455	0.330	0.409	0.385	0.394
SEM	0.03	0.04	0.02	0.02	0.02
Two-way ANOVA (probability)					
Lysine	0.1750	0.3761	0.0263	0.3352	0.1247
TSAA	0.3342	0.2353	0.5874	0.0487	0.0874
Lysine×TSAA	0.4945	0.4857	0.6687	0.2354	0.2245

Means in the same column within each classification bearing different letters are significantly different ($p<0.05$)

Contradicting results, by Hassan *et al.* (2003) and Koreleski and Swiatkiewicz (2010) demonstrated that dietary methionine significantly improved feed intake and feed conversion per kilogram of eggs.

The results obtained in this study revealed that the interaction between lysine and TSAA had no significant effect on all feed utilization measurements through the different experimental periods (Table 3 and 4). Similar findings were obtained by Bateman *et al.* (2008) who stated that hens consuming 0.97% lysine with 0.66% TSAA had better feed consumption during the period from 21-35 weeks of age, although feed conversion was not significantly affected by the interaction.

Productive performance: Feeding Lohmann laying hens different levels of lysine, TSAA and their interactions showed insignificant differences in egg number criteria during the experimental periods (Table 5). Egg weight was significantly ($p\leq 0.05$) improved (61.05 and 63.02 g) by lysine intake (0.84%) during 42-46 and 46-50 week of age, respectively. Egg mass also was significantly ($p\leq 0.05$) increased linearly with increasing lysine intake during period 46-50 and 34-50 weeks of age (Table 5, 6 and 7). Similar results obtained by Bouyeh and Gevorgian (2011) who documented that 0.76% lysine or more (10 and 20% above NRC (1994), recommendation) in Hy-line W-36 diet

Table 5: Egg weight of Lohmann laying hens as affected by lysine, total sulfur amino acids levels and their interaction

	Egg weight (g)				
Items	34-38 week	38-42 week	42-46 week	46-50 week	34-50 week
Lysine (%)					
0.74	56.86	55.90	56.04[b]	57.28[b]	56.52
0.84	58.25	56.70	61.05[a]	63.02[a]	59.75
094	60.84	57.05	59.73[a]	61.70[a]	59.83
TSAA (%)					
0.67	56.74	57.40	58.80	60.65	58.39
0.72	58.45	56.14	58.20	61.40	58.54
0.77	60.76	56.11	59.83	59.95	59.16
Lysine×TSAA (%)					
0.74×0.67	54.70	56.20	57.10	57.66	56.41
0.74×0.72	56.13	55.76	54.23	55.00	55.28
0.74×0.77	59.76	55.73	56.80	59.20	57.87
0.74×0.67	58.23	60.80	59.93	62.36	60.33
0.84×0.72	57.76	53.83	62.53	64.86	59.74
0.84×0.77	58.76	55.46	60.70	61.83	59.18
0.84×0.67	57.30	55.20	59.36	61.93	58.44
0.94×0.72	61.46	58.83	57.83	64.33	60.61
0.94×0.77	63.76	57.13	62.00	58.83	60.43
SEM	2.00	2.54	2.03	2.01	1.97
Two-way ANOVA (probability)					
Lysine	0.0854	0.0987	0.04578	0.0354	0.2589
TSAA	0.0789	0.0987	0.2654	0.0698	0.2647
Lysine×TSAA	0.2245	0.2354	0.2587	0.3547	0.5879

Means in the same column within each classification bearing different letters are significantly different ($p<0.05$)

led to significantly ($p\leq0.05$) higher egg production and egg output. Contrary, Schutte and Smink (1998) indicated that egg production was not significantly ($p\leq0.05$) affected by the different lysine (0.65, 0.69, 0.73, 0.77, 0.81, 0.85, 0.89 and 93%) levels in white Leghorn diets. On the same context, Sohail *et al.* (2003) found that adding 0.097% additional synthetic lysine to corn-soy diets formulated based on lysine containing 0.79-0.97% lysine had no significant influence on egg production for commercial Leghorn.

The TSAA of diets did not significantly affect egg mass during the studied periods except the last period (46-50 week-old), where the moderate level of TSAA recorded the highest value (1225.17 g) compared to other groups. This result is well accepted since egg mass is calculated as egg number multiplied by egg weight; therefore, egg mass values depend mainly upon egg number and egg weight values which were not affected by dietary TSAA levels. Similar results were reported by Alagawany *et al.* (2011) concluded that egg number and egg mass did not differ significantly due to TSAA levels except those of the trail period (22-34 weeks of age) that differed significantly ($p\leq0.05$), where birds received 0.72% of TSAA recorded the highest values for the two traits. These results disagreed with Liu *et al.* (2005) and Wu *et al.* (2005) who reported that increasing dietary levels of TSAA improved egg production and egg mass. Low egg production for hens fed low TSAA levels can partially be attributed to amino acids imbalance which attenuates

Table 6: Egg number of Lohmann laying hens as affected by lysine, total sulfur amino acids levels and their interaction during the experimental periods

	Egg numbers				
Items	34-38 week	38-42 week	42-46 week	46-50 week	34-50 week
Lysine (%)					
0.74	23.16	19.95	19.27	18.99	81.36
0.84	22.50	18.79	20.77	20.16	82.20
094	23.48	17.37	20.60	21.21	82.64
TSAA (%)					
0.67	21.79	18.87	20.77	19.27	80.68
0.72	24.00	19.45	19.99	21.65	85.08
0.77	23.35	17.79	19.88	19.44	80.44
Lysine×TSAA (%)					
0.74×0.67	22.12	22.25	18.66	18.83	81.84
0.74×0.72	24.00	20.12	19.99	20.99	85.08
0.74×0.77	23.37	17.50	19.16	17.16	77.16
0.74×0.67	20.00	16.75	21.33	17.49	75.56
0.84×0.72	22.62	18.87	19.49	22.32	83.28
0.84×0.77	24.87	20.75	21.49	20.66	87.76
0.84×0.67	23.25	17.62	22.32	21.49	84.68
0.94×0.72	25.37	19.37	20.49	21.66	86.88
0.94×0.77	21.82	15.12	18.99	20.49	76.40
SEM	0.55	0.65	1.05	1.44	1.71
Two-way ANOVA (probability)					
Lysine	0.0897	0.0879	0.1245	0.2245	0.1658
TSAA	0.0988	0.0798	0.1135	0.3254	0.1687
Lysine×TSAA	0.1245	0.2234	0.3254	0.5647	0.2658

Means in the same column within each classification bearing different letters are significantly different ($p<0.05$)

protein synthesis; inhibits absorption and increase catabolism of essential amino acids (Ahmad and Roland, 2003). There were no statistical ($p\leq 0.05$ or 0.01) differences due to interaction effect at all ages for egg weight, egg number or egg mass. These results were contrary to those obtained by Bateman *et al.* (2008) who found that high dietary levels of lysine (0.97%) and low level of TSAA (0.69%) recorded the heaviest egg weight, while lower lysine levels (0.79%) with TSAA level recorded the more height egg weight. Moreover, egg production was significantly ($p\leq 0.01$) affected by interaction between lysine and methionine during the period from 24-34 week of age, whereas that 0.97% lysine with 0.81% TSAA recorded the best values at all ages. Also, Murray *et al.* (1998) found that addition of synthetic amino acids like lysine and methionine at high levels to the diet can stimulate insulin secretion from pancreas by aggregation in plasma which in turn releases amino acids from stored sources and leads to protein synthesis.

Egg quality criteria: Table 8 and 9 show that, all of egg quality measurements were not significantly ($p\leq 0.05$ or 0.01) affected by various levels of dietary lysine except (albumen percent, USSW and yolk: albumen ratio) and (USSW and yolk diameter) at 42 and 50 weeks of age, respectively. Albumen percent and USSW were significantly ($p\leq 0.01$) increased by increasing lysine

Table 7: Egg mass of Lohmann Laying hens as affected by lysine, total sulfur amino acids levels and their interaction

	Egg mass (g)				
Items	34-38 week	38-42 week	42-46 week	46-50 week	34-50 week
Lysine (%)					
0.74	1288.19	1080.89	1050.16	1051.72[c]	4470.96[c]
0.84	1251.35	1016.88	1135.34	1119.48[b]	4523.04[b]
094	1349.62	972.12	1188.36	1214.41[a]	4724.52[a]
TSAA (%)					
0.67	1227.00	1024.06	1158.96	1085.32[b]	4495.36
0.72	1349.56	1053.35	1089.34	1225.17[a]	4717.44
0.77	1312.60	992.48	1125.53	1075.12[b]	4505.72
Lysine×TSAA (%)					
0.74×0.67	1200.86	1171.98	1018.77	1058.66	4450.28
0.74×0.72	1356.90	1096.77	1061.78	1171.19	4686.64
0.74×0.77	1306.80	973.92	1069.94	925.32	4276.00
0.74×0.67	1118.63	906.06	1151.20	939.05	4112.00
0.84×0.72	1231.07	984.16	1044.95	1270.85	4531.04
0.84×0.77	1404.35	1160.43	1209.87	1148.54	4923.20
0.84×0.67	1361.51	994.14	1306.92	1258.24	4920.00
0.94×0.72	1460.71	1079.12	1161.37	1233.48	4934.68
0.94×0.77	1226.65	843.10	1096.78	1151.51	4318.04
SEM	82.03	97.53	60.69	77.12	58.65
Two-way ANOVA (probability)					
Lysine	0.08970	0.1254	0.5478	0.04578	0.0215
TSAA	0.09825	0.1354	0.6547	0.03587	0.5689
Lysine×TSAA	0.12540	0.2587	0.8974	0.07890	0.6879

Means in the same column within each classification bearing different letters are significantly different ($p<0.05$)

levels in Lohmann layer hen diets, while yolk:albumen ratio and yolk diameter decreased with increasing lysine levels. These results agreed with those obtained by Abd El-Maksoud *et al.* (2011) who revealed that shell weight, egg shell thickness and albumen weight were not significantly ($p\leq0.05$) affected by amino acid supplementation, while the shape index (%) was improved with amino acids supplementation.

Regardless of lysine level, albumen percentage, yolk: Albumen ratio and yolk index were significantly ($p\leq0.05$ or 0.01) affected by TSAA levels (Table 8 and 9). On the other hand, other egg quality criteria were not significantly affected by TSAA intake at 42 and 50 weeks of age. Similar results reported by Koreleski and Swiatkiewicz (2010) indicated that eggshell characteristics were unchanged when methionine content in the diet was increased. Also, Schutte *et al.* (1994) reported significant improvements in egg weight when lower levels of TSAA were used in graded increments. These results coincided with those found by Novak *et al.* (2006) who showed that Haugh unit was not affected during the first cycle of production by different levels of TSAA. Liu *et al.* (2005) and Wu *et al.* (2005) found that increasing TSAA levels increased egg weight while Haugh unit reduced with increasing TSAA levels.

The interaction between lysine and TSAA levels indicated that yolk percentage and yolk:albumen ratio at an age of 42 week, as well as yolk index and diameter at 50 weeks were

Table 8: Egg quality criteria of Lohmann laying hens as affected by lysine, total sulfur amino acids levels and their interaction at 42 week of age

Items and levels	Albumein (%)	Yolk (%)	Shell (%)	Haugh unit	USSW	Yolk index	Yolk: Albumein ratio	Shell thickness (mm)	Yolk diameter (mm)
Lysine (%)									
0.74	61.21[b]	25.48	13.28	87.49	3.85[b]	52.11	0.416[a]	0.439	36.42
0.84	63.00[a]	24.36	12.61	86.70	3.94[a]	51.45	0.388[b]	0.429	37.03
0.94	62.82[a]	24.30	12.84	86.08	3.94[a]	50.52	0.387[b]	0.437	37.19
TSAA (%)									
0.67	62.08[b]	25.06	12.83	87.07	3.90	51.16	0.404[a]	0.440	37.25
0.72	61.49[b]	25.15	13.33	86.28	3.91	51.14	0.409	0.434	36.82
0.77	63.46[a]	23.93	12.58	86.93	3.92	51.78	0.378[b]	0.430	36.56
Lysine×TSAA (%)									
0.74×0.67	61.00	26.18[a]	12.79	88.32	3.87	50.01	0.429[a]	0.443	37.36
0.74×0.72	59.65	26.03[a]	14.30	84.87	3.83	52.13	0.434[a]	0.443	35.73
0.74×0.77	62.98	24.24[b]	12.75	89.28	3.87	54.18	0.385[c]	0.430	36.18
0.84×0.67	60.89	25.48[ab]	13.60	86.67	3.92	52.23	0.419[a]	0.437	37.22
0.84×0.72	62.46	25.37[ab]	12.15	85.59	3.96	51.57	0.406[a]	0.433	37.36
0.84×0.77	65.67	22.23[c]	12.08	87.85	3.94	50.55	0.339[c]	0.417	36.51
0.94×0.67	64.37	23.51[c]	12.09	86.23	3.91	51.24	0.365[b]	0.440	37.19
0.94×0.72	62.36	24.07[b]	13.54	88.37	3.95	49.71	0.386[b]	0.427	37.37
0.94×0.77	61.74	25.32[ab]	12.91	83.65	3.96	50.62	0.410[a]	0.443	37.00
SEM	0.55	0.630	0.47	0.62	0.03	2.00	0.010	0.010	0.65
Two-way ANOVA (probability)									
Lysine	0.0054	0.07890	0.5487	0.2548	0.0057	0.3972	0.0413	0.1772	0.1568
TSAA	0.0087	0.09865	0.5657	0.2547	0.0897	0.6041	0.0211	0.2784	0.2432
Lysine×TSAA	0.0987	0.00570	0.7852	0.4457	0.0963	0.7867	0.0065	0.3658	0.3586

Means in the same column within each classification bearing different letters are significantly different ($p<0.05$)

significantly ($p\leq 0.05$) influenced by the tested levels of lysine and TSAA in Lohmann brown hens diet. In contrast, there were no differences for other egg quality criteria at all studied ages. Egg quality criteria results were affected by lysine and TSAA combinations indicated that using 0.74 or 0.84% lysine with 0.67% TSAA recorded the highest values (yolk percentage, yolk: albumen ratio and yolk index) by comparison with others combination. Also decreases of amino acid levels in the diet suggest diet price and decreasing of N pollution so if one or two essential amino acids are increased over the nutrient requirement, losses in amino acids will occur as N excretion. To efficiently use excess amino acids, the crystalline amino acids such as DL-methionine and lysine can be added to the diets to balance the amino acid ideal profile. Novak *et al.* (2004), Liu *et al.* (2005) and Wu *et al.* (2005) reported that increasing lysine and TSAA had positive effect on performance of laying hens.

Economical evaluation: Data presented in Table 10 showed that, according to economical evaluation analysis; egg market price and net return were linearly increased from 49.17 and 20.57-49.76 and 21.77 and to 51.96 and 22.71 LE (Egyptian pounds) with increasing dietary lysine intake from 0.74 to 0.84 to 0.94%, respectively. The diet containing 0.84% lysine achieved the best values of economical efficiency (77.77%) compared to other diets during the whole experimental period.

Table 9: Egg quality criteria of Lohmann laying hens as affected by lysine, total sulfur amino acids levels and their interaction at 50 week of age

Items	Albumein (%)	Yolk (%)	Shell (%)	Haugh unit	USSW	Yolk index	Yolk: Albumein ratio	Shell thickness (mm)	Yolk diameter (mm)
Lysine (%)									
0.74	62.68	26.02	12.76	81.44	3.88[b]	45.68	0.417	0.389	41.56[a]
0.84	62.11	29.74	12.76	81.55	3.97[a]	44.66	0.412	0.408	38.06[b]
0.94	61.70	25.88	12.69	78.58	3.95[a]	46.49	0.420	0.407	38.35[b]
TSAA (%)									
0.67	61.58	26.48	12.11	78.39	3.93	45.98[ab]	0.432	0.391	39.00
0.72	61.49	25.82	12.91	82.00	3.95	42.15[b]	0.419	0.407	39.59
0.77	63.42	29.34	13.18	81.18	3.92	48.71[a]	0.398	0.406	39.38
Lysine×TSAA (%)									
0.74×0.67	60.57	27.27	12.14	78.52	3.89	46.12[a]	0.452	0.370	38.61[b]
0.74×0.72	60.23	25.83	13.89	81.75	3.84	44.34[ab]	0.423	0.410	41.90[a]
0.74×0.77	67.25	24.94	12.25	84.06	3.91	46.59[ab]	0.375	0.378	44.18[a]
0.84×0.67	62.16	26.02	12.08	79.03	3.96	48.78[a]	0.421	0.413	38.29[b]
0.84× 0.72	62.29	25.500	12.94	86.33	4.00	38.11[b]	0.411	0.403	37.85[b]
0.84× 0.77	61.88	37.72	13.26	79.29	3.95	47.11[a]	0.404	0.407	38.06[b]
0.94×0.67	62.03	26.17	12.11	77.64	3.95	45.98[ab]	0.424	0.390	40.11[b]
0.94× 0.72	61.95	26.11	11.91	77.92	3.99	42.15[b]	0.422	0.407	39.03[b]
0.94× 0.77	61.13	25.35	14.04	80.18	3.91	48.71[a]	0.415	0.423	35.91[c]
SEM	1.77	3.44	0.67	4.40	0.04	2.81	0.030	0.010	1.05
Two-way ANOVA (probability)									
Lysine	0.4940	0.2118	0.2033	0.3899	0.04587	0.0986	0.5684	0.2527	0.0365
TSAA	0.5874	0.2354	0.1925	0.4099	0.0987	0.0098	0.4578	0.2789	0.0687
Lysine×TSAA	0.6584	0.2658	0.2687	0.5178	0.1568	0.0487	0.6587	0.3254	0.0065

Means in the same column within each classification bearing different letters are significantly different ($p<0.05$)

The improvement of the net revenue and economical evaluation criteria for Lohmann laying hens which consumed high levels of lysine may be due to an increase in egg production and egg weight during the experimental period and might be attributed to 0.84% of lysine, also, due to the increase of feed consumption and egg mass during this period (Table 10). These results disagreed with Abd El-Maksoud *et al*. (2011) who found that hens fed diet supplemented with lysine recoded better values than those fed diets without adding lysine or methionine.

Egg market price, net revenue and economic feasibility were changed with different levels of TSAA, whereas, 0.72% of TSAA recorded the highest economical evaluation values (Table 10). These results revealed that 0.72% of TSAA in Lohmann layer diets was adequate to maximize economical evaluation criteria during or after the peak production. In addition, increasing the TSAA percentage in layer diets over the nutrient requirement for Lohmann hens elevated the price per kilogram diet and nitrogeneous excretion. These results coincided with those found by Abd El-Maksoud *et al*. (2011) who stated that the highest value of economic efficiency and relative economic efficiency had been recorded for hens fed a diet supplemented with methionine. On the other hand, Zeweil *et al*. (2011) reported that adding 0.27% methionine recorded the best value for economic efficiency (62.4%) compared with other levels for laying hens during the period from 28-48 week of age. The economical efficiency was not affected with the different levels of dietary TSAA throughout the experimental period.

Table 10: Economical evaluation of Lohmann laying hens as affected by lysine, total sulfur amino acids levels and their interaction during whole period 34-50 week of age

Variables and levels	Total feed intake (kg)	Cost of 1 kg feed (LE)	Total feed cost (LE)[A]	Total egg mass (kg)	Egg market price (LE)[B]	Net return (LE)[C]	Economical efficiency (%)[D]
Lysine effect (%)							
0.74	11.608	2.464	28.60	4.470	49.17	20.57	71.92
0.84	11.274	2.483	27.99	4.523	49.76	21.77	77.77
0.94	11.654	2.510	29.25	4.724	51.96	22.71	77.64
TSAA effect (%)							
0.67	11.761	2.462	28.95	4.495	49.44	20.49	70.77
0.72	11.422	2.483	28.36	4.717	51.88	23.52	82.93
0.77	11.352	2.512	28.51	4.505	49.55	21.04	73.79
Interaction (Lycine×TSAA)							
0.74×0.67	11.762	2.445	28.75	4.450	48.95	20.20	70.26
0.74×0.72	11.632	2.465	28.67	4.686	51.54	22.87	79.77
0.74×0.77	11.406	2.484	28.33	4.276	47.03	18.70	66.00
0.74×0.67	11.567	2.467	28.53	4.112	45.23	16.70	58.53
0.84×0.72	10.914	2.475	27.01	4.531	49.84	22.83	84.52
0.84×0.77	11.339	2.509	28.44	4.923	54.15	25.71	90.40
0.84×0.67	11.930	2.475	29.52	4.920	54.12	24.60	83.33
0.94×0.72	11.720	2.510	29.41	4.934	54.27	24.86	84.52
0.94× 0.77	11.313	2.545	28.79	4.318	47.49	18.70	64.95

A: Total feed cost = Feed intake×cost of kg, B: Egg market price = Total egg mass×cost of kg egg (11 LE), C: Net return = Difference between egg market price and total feed cost, D: Economic efficiency = Net return/total cost×100

The results of the economical evaluation indicated that the best net return (25.71 LE) and economic efficiency (90.40%) were recorded by Lohmann hens fed 0.84% lysine with 0.77% TSAA during the period from 34-50 week of age. These results agreed with El-Maksoud *et al*. (2011) who found that hens fed diet supplemented with lysine and methionine had recorded better values than those having diet without adding lysine or methionine.

CONCLUSION

Discounting of essential amino acids especially lysine and methionine could play a significant role in reducing feed cost or increasing economical efficiency. From the nutritional and economical aspects of view it can be conducted that, using 0.84% lysine and 0.72% TSAA was enough to get the best productive performance and economical efficiency of Brown Lohmann hens. Thus, dietary level of 0.84% lysine with 0.72% TSAA is recommended for feeding Lohmann hens throughout the whole period of 18-34 weeks of age.

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