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SPARING EFFECT OF BETAINE SUPPLEMENTATION IN LOW METHIONINE LAYING HEN DIETS ON PRODUCTIVE PERFORMANCE AND EGG QUALITY

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ABSTRACT: This study evaluated the sparing effect of betaine supplementation to low methionine laying hen diets on productive performance, egg quality, and nutrient digestibility. A total of 250 Lohmann Brown laying hens, 25 weeks age were randomly distributed into 5 treatments of 50 hens in 5 replicates. Five tested diets were formulated T1: 0.45% methionine with no betaine supplementation which basal considered as a control diet, T2: 0.40% methionine with 0.75g betaine/kg, T3: 0.40% methionine with 1.5g/ betaine/kg, T4: 0.35% methionine with 0.75g betaine/kg, and T5: 0.35% methionine with 1.5g betaine/kg. Betaine supplementation to low methionine diets affected significantly ($P<0.05$) on FI, FBW and BWG. No significant differences ($P>0.05$) in EP, EW, EM, FCR, SW, AYW, HU, and EYC due to treatment effect. There was a significant difference ($P<0.05$) among groups on ST due to betaine supplementation to low methionine diet compared to no betaine control diet. Moreover, hens fed 0.35 and 0.40% methionine with betaine supplementation improved ($P<0.05$) immunoglobulin titre (Ig) compared to those fed on the control diet. Betaine supplementation to low methionine diets had significantly ($P<0.05$) affected on NB and all nutrients digestibility except CF digestibility compared to no betaine diet (T1). Methionine levels effect had effect ($P<0.05$) on FI, FCR, FBW, BWG, ST, EYC, and Ig titre. Decreasing methionine level up to 0.35% did not significantly ($P<0.05$) affected on all nutrients digestibility, except CP digestibility and NB which were affected significantly. Betaine levels effect significantly affected ($P<0.05$) on FI, HU, EYC, and Ig titre, while no effect ($P>0.05$) due to betaine levels on EP, EW, EM, FCR, FBW, BWG, SW, ST, and AYW. It could be concluded that betaine supplementation had a role-sparing effect with a low methionine diet especially at level 0.35% without any negative effect on laying productivity, egg quality, and immune response.

Keywords: betaine, laying hens, shell thickness, digestion, immunoglobulin titer

INTRODUCTION

Essential sulfur amino acid methionine plays critical roles in methylation reaction, protein accretion (Vazquez-Anon et al., 2006), productive performance promotion (Ai and Xie, 2005 and Zhan et al., 2006), and immune response enhancement (Zhang and Guo, 2008). Reducing the dietary methionine level from 0.36 to 0.23% in laying hen diets at 54 to 72 age weeks lead to lower egg production and smaller egg size (Keshavarz, 2003). Methionine supplementation as DL-Methionine, DL-2-hydroxy-4-methylthio butyric calcium, and coated DL-Methionine enhanced the production and egg quality of broiler breeders (Xiao et al., 2017). Many aspects of a methionine deficiency on avian metabolism have been studied such as its interaction with choline, betaine, folic acid, and vitamin B_{12} (Carew et al., 2003). Micronutrient betaine (chemically known as trimethylglycine) plays two roles in cells; stabilizing osmolyte and donating methyl group to other biomolecules (Craig, 2004). Betaine can potentially eliminate the need for creatine, methionine, and choline as methyl-group donors (Singh et al., 2015). Moreover, betaine donates the methyl radicals for the remethylation of homocysteine to methionine and to formulate creatine, carnitine, and phosphatidylcholine through the S-adenosyl methionine pathway. A methyl group is transferred from betaine to dimethylglycine through the enzyme betaine homocysteine methyl transferase. It also contains three methyl groups that allow effective spare of dietary methionine. Homocysteine forms methionine either through betaine or 5-methyl tetrahydrofolate (Alirezaei et al., 2011). Being a methyl group donor, betaine may sometimes replace methionine in a reaction with homocysteine (Maidin et al., 2021). Thus, methionine could be used more for its growth function (Paniz et al., 2005) and dietary betaine can be substitution by 30% of dietary methionine in broiler diets (Sahebi-Ala et al. 2021). Betaine supplementation in laying hen diets improved ($P<0.05$) egg production performance (Xing and Jiang, 2012). Moreover, enhanced significantly ($P<0.05$) egg laying rate, but did not affect ($P>0.05$) on feed intake and egg weight (Omer et al., 2020). Using betaine in poultry, fish, and pigs diets as feed additive (Eklund et al., 2005) and the reducing effects of heat stress in broiler chickens (Haldar et al., 2015). Adding betaine up to 1000mg/kg to Lohmann laying hens did not significantly ($P>0.05$) affected on egg production, feed intake, egg mass, shell density, shell strength, shell mass, shell thickness, and shell percentage (Maidin et al., 2021). Attia et al. (2016) showed that significant increased ($P<0.05$) of laying rate, egg mass, and feed intake of laying hens and improve the feed conversion ratio while, the egg weight was not significant affect for betaine supplementation (1g/kg) group compared to control laying hens at critical heat stress. The present study was to investigate the effect of dietary betaine supplementation with decreasing methionine levels on egg production, egg quality, immunoglobulin titer, and nutrients digestibility of Lohmann Brown laying hens. Therefore, a study was conducted to determine the best level of betaine in low-methionine diets to achieve sparing effect to realize optimum productive performance and egg quality of laying hens.

betaine, laying hens, shell thickness, digestion, immunoglobulin titer

MATERIAL AND METHODS

The experiment, the laboratories work and feed mixing processing were conducted at Animal Production Department, Faculty of Agriculture, Cairo University, Giza, Egypt

Birds and management

Two hundred and fifty Lohmann Brown laying hens (25 week age) were randomly distributed into 5 treatments of 50 hens in 5 replicates of 10 hens each. Birds were housed in battery cages, kept under similar environmental and managerial conditions throughout the whole experimental period (16 weeks). Feed and water were offered *ad-libitum* throughout the experimental period from 25 to 40 week of age, under a total of 16 hours of light per day regimen.

Diets

The experimental diets were formulated to meet the nutrient requirements according to recommended allowances of the Lohmann Brown breed. The control diet was formulated to contain 2800 kcal ME/kg, 18.50% CP, and 0.45% methionine without betaine supplementation. Accordingly, a total of 5 experimental diets were formulated (2 methionine levels × 2 betaine levels = 4 + control diet = 5 diets). The treatments were control diet (T1), 0.40% methionine and 0.75g betaine/kg (T2), 0.40% methionine and 1.5g betaine/kg (T3), 0.35% methionine and 0.75g betaine/kg (T4), and 0.35% methionine and 1.5g betaine/kg (T5). The dietary betaine supplementation was added as a Betafin S6 source (Danisco Animal Nutrition, Finnfeeds Finland Limited, Sokeritehtaantie, Naantali, Finland). All experimental diets and their calculated chemical analysis (Table 1) were formulated to be isocaloric (2800 kcal ME/kg) and isonitrogenous (18.5% CP) to meet all the essential nutrient requirements for laying hens (NRC, 1994).

Measurements

At the beginning of trial, all birds of each treatment were weighed and at the end of experimental period to calculate final body weight gain (BWG). Feed intake (g/hen/day), egg production (hen-day %), egg mass (g) were calculated every 4 weeks interval during the experimental period. Egg production (EP), egg weight (EW), and feed intake (FI) were used to calculate the amount of feed (kg) which was required to produce one kg of eggs per hen to calculate feed conversion ratio (FCR; kg. feed : kg. eggs). Egg shell thickness (ST) was determined using a dial pipe gauge digital. Haugh units were calculated based on the height of albumen determined by a micrometer and egg weight (Eisen et al., 1962). Egg yolk visual color was determined by Laroch's fan with 15 degrees of egg yolk color (Haughman Laroch's fan). Serum total immunoglobulin titres were determined according to Zipp et al. (1983).

Digestion coefficient and nitrogen balance

At the end of experimental period, 40 weeks of age, a total of 60 hens 12 from each treatment were randomly taken for carrying digestion trial to estimate the nutrients digestibility, and nitrogen balance. Excreta were collected quantitatively every 24 hours, during a three days collection period. Nutrients proximate analysis of the feed and dried excreta was determined by A.O.A.C. (2000) and Faecal nitrogen by Jakobsen et al. (1960).

Statistics

Data were processed utilizing SAS software (SAS, 2004) implementing the General Linear Model procedures (PROC

GLM) of SAS. Data were tested by one-way analysis of variance for treatments, methionine, and betaine effects. Means differences were obtained using a least squares means statement, if ANOVA was significant, means were then separated by Duncan's multiple range test (Duncan, 1955) and differences were significant at (P≤0.05).

RESULTS AND DISCUSSION

Laying hen's performance

Statistical analysis for laying hens performance is illustrated in Table 2. There were significant (P<0.05) differences between low methionine diets with betaine supplementation versus control group for final body weight (FBW; P<0.0001), body weight gain (BWG; P<0.0001), and feed intake (FI; P<0.0001). The laying hens fed T4 diet gave the highest values of FBW (1900g), BWG (432g), and FI (117.74g), while those fed T3 recorded the lowest values of FBW (1800g) and BWG (332g) compared with the other groups including control diet (T1) which less consumed FI (107.93g/hen/day). No significant difference (P>0.05) between the control diet and T3 group in FI was observed. Laying hens fed low methionine diets with betaine supplementation (T2-T5) significantly (P<0.001) consumed more feed than those fed control diet (T1). No significant differences (P>0.05) among low methionine diets compared to control methionine group in egg production (EP; P=0.708), egg weight (EW; P=0.730), egg mass (EM; P=0.842), and feed conversion ratio (FCR; P=0.177). The observations in current study for Lohmann Brown laying hens performance were somewhat consistent with Park and Rya (2011) observed that EP, EW, and EM increased (P>0.05) with laying hens fed 0.6g/kg betaine, also FI and FCR were not significantly (P>0.05) affected due to betaine supplementation. Laying rate, EM, FCR, and BWG improved (P<0.05) and FI affected (P<0.05) with laying hens fed on diets containing betaine up to 1g/kg compared to control group, while, EW was not affected significantly (Attia et al., 2016). Likewise, FI and BW were significantly (P<0.05) increased with 1, 1.5, and 2 g/kg betaine supplementation to broiler diets and FCR improved (P<0.05) with 2g/kg diet (Chand et al., 2017). Broilers BWG and FI increased linearly (P<0.05) with betaine supplementation at levels 0.50, 1.00 and 2.00g/kg diet, while no significant (P>0.05) effect on FCR was observed (Liu et al., 2019). At 4 weeks of starter period for laying hens, BW and FI increased (P<0.05) and uniformity improved (P>0.05) with laying fed 12 and 18 mg betaine/kg diet (Wahid et al., 2021). Moreover, BWG and FI were significantly (P<0.05) increased with broilers fed 2g/kg betaine compared with those fed basal diet without methionine addition (Savaram et al., 2022). However, laying hens fed 0.7 and 1.5g/kg dietary betaine increased (P<0.05) daily eggs number and improved (P>0.05) EW and EM (Gudev et al., 2011). In quails fed diets including 0.6 and 1.2 g/kg dietary betaine improved (P<0.05) EP, EM, and FCR, increased (P<0.05) FI, and decreased (P<0.05) EW (Ratriyanto et al., 2017). Recently, no significant (P>0.05) differences in FI, FBW, and EW, while EP improved (P<0.05) due to 5g/kg betaine supplementation to laying hen diets (Omer et al., 2020). In the current study, the improvement in BWG with low methionine (0.35 and 0.40%) and different levels of betaine (0.75 and 1.50g/kg) due to increased nutrient digestibility (Table 4) and important role of betaine as a methyl donor in many reactions including

DNA and RNA methylation and protein synthesis, besides the methyl groups sparing function (Savaram et al., 2022). Hens receiving the low methionine diet with betaine supplementation consumed significantly (P<0.0001) more feed than those receiving the control diet. Hens increased FI to meet their increased methionine requirement. Increasing dietary supplementation of methionine slightly decreased the amount of feed consumed in control group.

Methionine level had affect (P<0.05) on FI (P<0.0001), FCR (P=0.038), FBW (P<0.0001), and BWG (P<0.0001), while no effect (P>0.05) on EP (P=0.313), EW (P=0.446), and EM (P=0.480). The FCR improved (P>0.05) with 0.27%methionine laying hens group in comparison with those fed 0.46%methionine diet, while EP, EM and EW significantly (P<0.05) decreased (Liu et al., 2022). Methionine supplementation at level 0.51% in laying hen diets had significantly (P<0.05) lower EW while, EM and FI significantly (P<0.05) increased compared to control diet (Moghadam et al., 2021).

Betaine supplementation to low methionine diets significantly affected (P<0.05) on FI, FBW and BWG, however, no significant effect (P>0.05) on laying hens EP, EW, EM, and FCR. No effect (P>0.05) due to betaine levels on EP (P=0.436), EW (P=0.354), EM (P=0.724), FCR (0.334), FBW (P=0.439), and BWG (P=0.439), while FI (P=0.049) significantly affected. Somewhat, there was insignificantly (P>0.05) increased in FBW (P=0.91) and decreased in EW (P=0.310) due to betaine supplementation up to 0.05g/kg in laying hen diets (Abobaker et al., 2017).

Egg quality and immunoglobulin titre

Statistical analysis for egg quality is presented in Table 3. For external egg quality, no significant (P>0.05) effect of betaine supplementation to low methionine diets on shell weight (SW; P=0.306) between dietary betaine groups and control group (T1, no betaine supplementation). A significant difference (P<0.05) among groups on shell thickness (ST; P=0.007) due to betaine supplementation to low methionine diet compared to no betaine control diet. Shell thickness significantly decreased (P<0.05) by adding 1.50g/kg betaine to 0.40% methionine diet (T3), but no significant difference in ST between the other groups and control group. For internal egg quality, no significant differences (P>0.05) in albumin and yolk weight (AYW; P=0.484), Haugh unit (HU; P=0.084), and egg yolk color (EYC; P=0.076) between low methionine diets with betaine supplementation and control group. Moreover, AYW and HU insignificantly (P>0.05) improved due to betaine supplementation in all low methionine diets (T2 – T5) compared to no betaine supplementation diet (T1, control group). The results in egg quality inhere were confirmed by Gudev et al. (2011) showed that the SW, AYW, HU, and EYC score did not significantly (P>0.05) affected by 0.7 and 1.5g/kg betaine supplementation in laying hen diets, while ST improved (P<0.05) in betaine groups. In quails, egg ST, egg shell breaking strength, albumin height, HU, and EYC were not significantly (P>0.05) affected by 0.6g/kg betaine supplementation (Park and Rya, 2011) and yolk and albumen percentage and egg shell percentage were not affected (P>0.05) by 0.0, 0.7, 1.4 and 2.1g/kg betaine supplementation (Ratriyanto et al., 2018). However, supplementation 0.6 and 1.2 g betaine/kg to quail diets improved (P<0.05) yolk weight and SW and decreased (P<0.05) albumin weight, while betaine supplementation did not significantly improved ST (Ratriyanto et al., 2017). In current study, added betaine at levels 0.75 and 1.5g/kg to 0.35% low methionine diets improved ST due to the calcium losses on the low methionine diet (0.35%) and were lower than the intermediate (0.40%) and control methionine (0.45%) diets. High egg ST quality due to high calcium intake because of high feed intake in 0.35% low methionine diets with 0.75 and 1.5g betaine/kg diet (Table 2). Moreover, the reason for reduced shell quality is due to increased egg size, distribution a constant amount of shell over larger egg.

Methionine level significantly affected (P<0.05) on ST (P=0.005), however, no effect (P>0.05) on SW (P=0.085). Moreover, methionine addition had significantly (P<0.05) effect on EYC (P=0.048), while no difference in both AYW (P=0.233) and HU (P=0.440). As the same trend, methionine supplementation at levels 0.51% decreased (P<0.05) ST, while HU had not affected (P>0.05) due to methionine addition to laying hen diets (Moghadam et al., 2021). The ST was significantly (P<0.05) lower with 0.27% methionine group compared to control group (0.46% methionine), while HU and EYC hadn't affected (P>0.05) by methionine levels (Liu et al., 2022).

No significant differences (P>0.05) due to betaine effect on SW (P=0.143) and ST (P=0.176). There were significant differences (P<0.05) in HU (P=0.034) and EYC (P=0.048), while no difference (P>0.05) in AYW (P=0.182) due to betaine effect up to 0.75 g/kg. Moreover, betaine supplementation at level 0.05g/kg had not significantly (P>0.05) affected on yolk weight (P=0.450) in laying hens group (Abobaker et al., 2017).

Statistical analysis for immunoglobulin titre (Ig) is illustrated in Table 3. Treatment effect had significantly (P<0.05) on Ig titre (P<0.0001). Hens fed 0.35 and 0.40% methionine with betaine supplementation improved Ig titre compared to those fed on control diet (no betaine supplementation). Moreover, there were significant differences in Ig titre due to be methionine effect (P=0.005) and betaine supplementation (P=0.005). The improvement in Ig titre due to betaine supplementation with different methionine levels in present study were similar to those found by Chand et al. (2017) antibody titer was improved (P<0.05) with broilers fed 1.0, 1.5 and 2.0g/kg betaine groups. Betaine supplementation in poultry diet has several biological benefit in lipid metabolism, anti-oxidation, anti-inflammation, and immunity (Zhang et al., 2016 and Wang et al., 2020). Lymphocyte proliferation was improved (P<0.05) with broilers fed 2g/kg betaine diet when compared to those fed basal diet without methionine addition (Savaram et al., 2022). Serum total Ig decreased (P<0.05) with increasing dietary methionine concentrations in Brown layers diets (Balnave, 2000).The improvement of immune response of laying hens might be due to increased digestibility and utilization of methionine (Augustine and Danforth, 1999) and other nutrients like carotenoids, lysine, protein, and fat (Remus et al., 1995), which are known to influence immune responses (Latshaw, 1991). Moreover, through its methyl donating property, betaine might have spared the dietary methionine from methyl donor function and the dietary methionine was available for other vital functions like protein synthesis and immune modulation (Kidd et al., 1997).

Nutrients digestibility and nitrogen balance

Statistical analysis for nutrients digestibility and nitrogen balance (NB) are listed in Table 4. Betaine supplementation to low methionine diets (T2 – T5) had significant ($P<0.05$) affected on nutrients digestibility of DM ($P=0.006$), OM ($P=0.009$), CP ($P<0.0001$), EE ($P<0.0001$), and NFE ($P=0.002$) compared to no betaine diet (T1). No significant difference in CF digestibility ($P=0.114$) due to treatment effect. The laying hens fed T5 tested diet were the highest ($P<0.05$) DM digestibility while, those fed T3 diet were the lowest ($P<0.05$) value of OM%. Crude protein digestibility was significantly ($P<0.05$) decreased with T2 (97.07%) and T3 (97.29%) but, not significantly differences between T4 and T5 in comparison with T1 group. The betaine laying hens groups were increased ($P>0.05$) in CF% digestibility when compared to T1 group. The EE digestibility was increased ($P<0.05$) in T2 group by 0.27%, but decreased ($P<0.05$) with T3 and T4 by 2.63 and 0.46%, respectively. No significant difference between T1 and T5 in EE digestibility. Laying hens in T4 group significantly increased ($P<0.05$) NFE digestibility by 2.60% compared to those in T1 control group while, the other betaine groups were not significantly affected. Nitrogen balance significantly ($P<0.0001$) increased in T4 and T5, but T3 group not affected ($P>0.05$) when compared to the control group (T1). Decreasing methionine level up to 0.35% not significantly ($P>0.05$) on all nutrients digestibility, except CP digestibility ($P<0.0001$) and NB ($P<0.0001$) which affected significantly. Betaine supplementation didn’t significantly ($P>0.05$) effect on of DM ($P=0.068$), CP ($P=0.499$), CF ($P=0.533$), and EE ($P=0.072$) digestibility and NB ($P=0.085$), while digestibility of OM ($P=0.008$) and NFE ($P=0.010$) affected significantly. Nutrients digestibility of DM, CP, EE, and CF, were significantly ($P<0.05$) differences with quail fed diets containing 0.6 and 1.2 g/kg dietary betaine, while NFE digestibility was not significantly affected (Ratriyanto et al., 2017). However, digestibility of DM, EE, CF and NFE digestibility not affected ($P<0.05$) by betaine supplementation up to 1g/kg in laying hen diets (Attia et al., 2016) Moreover, digestibility of DM and CP didn’t significantly ($P>0.05$) differ among broiler diets due to betaine supplementation up to 1.2 g/kg with different levels of dietary CP (Park and Kim, 2019). The CF digestibility not significantly ($P<0.05$) affected, while nitrogen retention was significantly ($P<0.05$) increased with 0.50, 1.00, and 2.00 g/kg betaine supplementation in broiler diets (Liu et al. (2019). The improvement of nutrients digestibility might be due to the role of betaine which has osmoprotective properties that help to increase proliferation of intestinal structure (Eklund et al., 2005).

Conclusion and application

Based on the significant interactions among dietary methionine and betaine in the current study, the data indicated that using 0.75 and 1.5g/kg betaine supplementation with low levels of methionine (0.35 and 0.40%) in feeding Lohmann Brown laying has improved productive performance, egg quality, immunoglobulin titre, and nutrients digestibility. These positive effects may be attributed to the methionine sparing effect as a result of dietary supplementation of betaine when added to low methionine diets (0.35 and 0.40%). This means that an inadequate methionine supply to the diet can be compensated by an increase in betaine supplementation. Furthermore, decreasing dietary methionine with betaine supplementation up to 1.5g/kg resulted in a better nitrogen balance and lower nitrogen excretion which in turn would have a positive impact on the environment. Consequently, environmental contamination with nitrogen and its by-products may be reduced with the ultimate benefit of producing more friendly environment and healthier products.

Table (1): Composition and chemical analysis of basal diets

Items	Methionine levels		
	0.45%*	**0.40%**	**0.35%**
Ingredients			
Yellow corn	48.94	49.42	48.71
Wheat bran	5.03	4.64	5.15
Vegetable oil	4.55	4.43	4.63
Soybean meal (44%)	30.79	30.87	30.92
Limestone	8.28	8.28	8.28
Dicalcium phosphate	1.65	1.65	1.65
Salts (NaCl)	0.30	0.30	0.30
Vitamin and mineral premix**	0.30	0.30	0.30
DL-methionine	0.16	0.11	0.06
Total	100.00	100.00	100.00
Calculated analysis			
Crude protein (%)	18.50	18.50	18.50
Metabolizable energy (kcal/kg)	2800	2800	2800
Total methionine (%)	0.45	0.40	0.35
Methionine + Cysteine %	0.75	0.70	0.65
Lysine (%)	1.00	1.00	1.00
Calcium (%)	3.60	3.60	3.60
Available phosphorus (%)	0.45	0.45	0.45

*0.45% methionine diet considered as a control diet.

**Vitamin and mineral premix at 0.3% of the diet supplies the following per kg of the diet: vit.A 8000000 I.U., vit.D_3 2400000 I.U., vit. E 14000 mg, vit. K_3 2000 mg, vit. B_1 2000 mg, vit.B_2 5000 mg, vit.B_6 2000 mg, vit.B_{12} 10 mg, pantothenic acid 8000 mg, niacin 27000 mg, folic acid 500 mg, biotin 34 mg, manganese 54 g, zinc 50 g, iron 27 g, copper 7 g, iodine 2 g, selenium 0.2 g, cobalt 0.20 g

Table (2): Effect of experimental diets on laying hens performance

Items	EP (%)	FI (g/hen/day)	EW (g)	EM (g of egg/ hen/day)	FCR (g feed : g egg)	FBW (g)	BWG (g)
T1 (0.45% Methionine + 0.00 betaine g/kg)	90.58	107.93[d]	58.61	53.10	2.03	1850.00[c]	382.00[c]
T2 (0.40% Methionine + 0.75 betaine g/kg)	87.12	110.73[c]	59.81	52.11	2.13	1845.00[d]	377.00[d]
T3 (0.40% Methionine +1.5 betaine g/kg)	86.21	108.46[d]	60.58	52.23	2.08	1800.00[e]	332.00[e]
T4 (0.35% Methionine + 0.75 betaine g/kg)	83.68	117.74[a]	59.43	49.73	2.37	1900.00[a]	432.00[a]
T5 (0.35 % Methionine + 1.5 betaine g/kg)	83.77	114.14[b]	60.20	50.43	2.26	1890.00[b]	422.00[b]
SEM	3.40	0.33	1.00	2.16	0.12	0.00	0.00
P-value							
Treatment effect	0.708	<0.0001	0.730	0.842	0.177	<0.0001	<0.0001
Methionine effect	0.313	<0.0001	0.446	0.480	0.038	<0.0001	<0.0001
Betaine effect	0.436	0.049	0.354	0.724	0.334	0.439	0.439

T: treatment, EP: Egg production, FI: feed intake, EW: egg weight, FCR: feed conversion ratio, FBW: final body weight, BWG: body weight, FBW: final body weight, FI: feed intake, FCR: feed conversion ratio, P-value: probability value
a., b…and e mean values with different superscript letters in the same column are significantly different (P<0.05).

Table (3): Effect of experimental diets on egg quality and Immunoglobulin titre

Items	External egg quality		Internal egg quality			Ig titre
	SW (g)	ST (µm)	AYW (g)	HU	EYC	
T1 (0.45% Methionine + 0.00 betaine g/kg)	6.55	0.42[a]	52.06	64.69	8.16	5.00[b]
T2 (0.40% Methionine + 0.75 betaine g/kg)	6.09	0.39[ab]	53.72	66.20	7.67	6.00[a]
T3 (0.40% Methionine +1.5 betaine g/kg)	6.03	0.38[b]	54.54	64.14	7.92	5.00[b]
T4 (0.35% Methionine + 0.75 betaine g/kg)	6.31	0.42[a]	53.12	68.06	7.92	6.00[a]
T5 (0.35 % Methionine + 1.5 betaine g/kg)	6.20	0.41[a]	54.01	64.73	7.83	6.00[a]
SEM	0.16	0.01	0.90	0.90	0.10	0.00
P-value						
Treatment effect	0.306	0.007	0.484	0.084	0.076	<0.0001
Methionine effect	0.085	0.005	0.233	0.440	0.048	0.005
Betaine effect	0.143	0.176	0.182	0.034	0.048	0.005

T: treatment, SW: Shell weight, ST: Shell thickness, AYW: Albumin and yolk weight, HU: Haugh unit, EYC: Egg yolk color, Ig: Immunoglobulin titre. P-value: probability value
a and b mean values with different superscript letters in the same column are significantly different (P<0.05).

Table (4): Effect of experimental diets on nutrients digestibility and nitrogen balance

Items	Digestibility coefficient of nutrients (%)						Nitrogen balance
	DM	OM	CP	CF	EE	NFE	
T1 (0.45 Methionine + 0.00 Betaine g/kg)	72.52[b]	81.79[ab]	98.12[a]	28.13	88.82[b]	82.15[bc]	61.82[c]
T2 (0.40 Methionine + 0.75 Betaine g/kg)	71.93[b]	82.99[a]	97.07[c]	28.51	89.06[a]	82.94[b]	64.01[b]
T3 (0.40 Methionine +1.5 Betaine g/kg)	72.44[b]	80.47[c]	97.29[b]	27.80	86.48[d]	82.44[bc]	62.37[c]
T4 (0.35 Methionine + 0.75 Betaine g/kg)	72.35[b]	82.38[ab]	98.25[a]	28.71	88.41[c]	84.28[a]	67.43[a]
T5 (0.35 Methionine + 1.5 Betaine g/kg)	73.68[a]	81.60[bc]	98.22[a]	28.92	88.77[b]	81.24[c]	67.69[a]
SEM	0.11	0.23	0.04	0.18	0.03	0.24	0.23
P-value							
Treatment effect	0.006	0.009	<0.0001	0.114	<0.0001	0.002	<0.0001
Methionine effect	0.118	0.923	<0.0001	0.093	0.210	0.772	<0.0001
Betaine effect	0.068	0.008	0.499	0.533	0.072	0.010	0.085

T: treatment, DM: dry matter, OM: organic matter, CP: crude protein, CF: crude fiber, EE: ether extract, NFE: nitrogen free extract. P-value: probability value

a, b, c and d mean values with different superscript letters in the same column are significantly different (P<0.05).

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الملخص العربى

التأثيرالتعويضي لإضافة البيتايين إلى علائق الدجاج البياض المنخضة المثيونين على الأداء الإنتاجي وجودة البيضة

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تقيم هذه الدراسة تأثير إضافة البيتايين الى علائق الدجاج البياض على الأداء الإنتاجي و جودة البيضة ومعاملات هضم المركبات الغذائية. قسمت 250 دجاجه لوهمان بني عمر 25 أسبوع عشوائيا الى 5 مجاميع تجريبية إحتوت كل مجموعة على 50 دجاجه والتي قسمت الى 5 مكررات. تم عمل خمس علائق تجريبية ،العليقة الاولى تحتوي على 0,45% مثيونين بدون إضافة بيتايين و هي العليقة الأساسية (الكنترول)، العليقة الثانية إحتوت على 0,40% مثيونيين بالإضافة الى 0,75 جم /كجم بيتايين، العليقة الثالثة تحتوي على 0,40% مثيونين و 1,5جم/كجم بيتايين، العليقة الرابعة إحتوت على 0,35% مثيونين و 0,75جم/كجم بيتايين و العليقة الخامسة إحتوت على 0,35% مثيونين و 1,5جم/كجم بيتايين. إضافة البيتايين الى العلائق منخفضة المثيونين أثرت معنويا على وزن الجسم النهائي، ومعدل الزيادة في وزن الجسم والماكول. ولم يتاثر معنويا كلا من وزن القشرة ووزن البياض والصفار و HU و ولون الصفار بين العلائق المنخفضة المثيونين والمضاف اليها البيتايين والمجموعة الكونترول. ظهر تأثر معنوي بين المجاميع في سمك القشرة للعلائق المحتوية على البيتايين ومنخفضة المثيونين مقارنة بعدم وجود البيتايين في العليقة الكونترول. وجد ان أن الدجاج المغذى على 0,35 و 0,40% ميثيونيين مع إضافة البيتايين حسنت من تيترإميونوجلوبيولين مقارنة بالمجموعة المغذاه على العليقة الكونترول. إضافة البيتايين الى العلائق منخفضة المثيونين أثرت معنويا على معامل هضم كل من المادة الجافة ، المادة العضوية، البروتين الخام، مستخلص الإيثير ومستخلص خالى الأزوت مقارنة بالعليقة غير المحتوية على البيتايين (المجموعة الكنترول). مستويات المثيونين أثرت معنويا على كل من المأكول، معامل التحويل الغذائي، وزن الجسم النهائي، متوسط الزيادة في وزن الجسم ، سمك القشرة، لون صفار البيضة، تيتر الايميونوجلوبيولين. إنخفاض المثيونين الى 0,35% لم يؤثر معنويا على معاملات هضم المركبات الغذائية فيما عدا معامل هضم البروتين الخام وميزان النيتروجين حيث أظهر تأثيرا معنويا. مستويات البيتايين أثرت معنويا على المأكول و HU ، لون صفار البيضة وتيتر الإميونوجلوبيولين بينما لم تظهر مستويات البيتايين تأثير على إنتاج البيض، وزن البيضة، حجم البيضة، معامل التحويل الغذائي، معدل الزيادة الوزنية و وزن القشرة، سمك القشرة ووزن البياض والصفار. نستنتج أن إضافة البيتايين كان له دور في التاثير التعويضي لنقص المثيونين في العلائق خاصة عند مستوى 0,35% بدون أي تأثير سلبي على إنتاج البيض، جودة البيضة والإستجابة المناعية.