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National Animal Production Research Institute
Ahmadu Bello University
P.M.B 1096, Shika-Zaria,
Kaduna State, Nigeria.
Email:japr@napri.gov.ng Url:journals.napri.gov.ng

EFFECTS OF SERIAL DIETARY SUPPLEMENT OF SELENIUM AND VITAMIN E ON PERFORMANCE OF LAYING HENS, EGG QUALITY ATTRIBUTES AND DEPOSITION OF NUTRIENTS IN THE EGGS

Jemiseye, F. O. and *Ogunwole, O. A.

Agricultural Biochemistry and Nutrition Unit, Department of Animal Science, University of Ibadan, Ibadan, Nigeria

**Corresponding Author's E-Mail: droaogunwole@gmail.com*

ABSTRACT

Effects of serial dietary supplement of selenium and vitamin E on performance of laying hens, egg quality attributes and deposition of selenium and vitamin E in eggs were evaluated. ISA brown pullets (n=192) at week 31 of age, weighing 1.38±0.11kg were assigned to six experimental diets. Basal diet (T1) was without supplemental selenium and vitamin E while diets T2, T3 and T4, contained 0.5, 1.0 and 1.5 mg/kg selenium, respectively. Other diets (T5 and T6) were supplemented with 20 and 40 mg/kg vitamin E, respectively. Each treatment comprised four replicates with eight birds per replicate allotted in a completely randomized design. Hen day egg production of hens on T2 (47.23 %) was highest. Other indices of performance were not significantly ($p<0.05$) affected by supplemental selenium and vitamin E. There was no effect ($p>0.05$) of treatment on egg length and shell thickness. However, egg weight, egg diameter, shell weight and shell ratio differed significantly ($p<0.05$). Hens on T6 had the highest egg weight (62.56 g) and egg diameter (44.22 mm). Eggs from hens on T1 had highest shell weight of 6.03 g and shell ratio of 10.50 among others. Supplemental selenium and vitamin E had no significant ($p>0.05$) effect on albumen weight, albumen diameter, yolk height, yolk colour, and yolk albumen ratio. However, albumen length and yolk weight significantly ($p<0.05$) increased with T2 (110.11) and T6 (15.13), respectively. Haugh unit were higher for eggs of hens on T4 (76.85) and T5 (80.04). Vitamin E deposition in eggs increased significantly ($p<0.05$) with dietary vitamin E from 1.08 mg/100 g in T1 to 1.26 mg/100 g in T6. Lower supplemental selenium enhanced egg production while increased supplemental vitamin E improved egg quality parameters and vitamin E deposition in egg.

Keywords: Hens laying performance, Supplemental vitamin E and selenium, Vitamin deposition, Egg quality attributes

INTRODUCTION

Vitamin E is a lipid soluble part of biological membranes which is considered a major antioxidant in protecting cells and tissues from oxidative damage induced by free radicals (Gallo-Torres, 1972). Selenium is also known as a powerful antioxidant whose metabolic role is in its function at the active site of the enzyme glutathione peroxidase which protects the cells from free radicals damage and lipid peroxides (Combs and Combs, 1986).

Selenium and vitamin E are antioxidants that play important roles in maintenance of health in laying hens, improve productivity and reproductive characteristics (Surai, 2000). Jiang *et al*. (1994) supplemented 400 IU DL-α-tocopheryl acetate in diet for laying hens and reported that 390 mg/kg α-tocopherol was deposited in the egg yolk. There was a record of increased yolk selenium content with increasing selenium level in the diet of laying chickens (Aljamal, 2011).

The most frequent method to enrich eggs with both vitamins and minerals is to use high level of these nutrients in the diet (Laudadio *et al*., 2015). Fortification of eggs with vitamin E is an acceptable practice as a result of its antioxidative properties for all species (Mohiti-Asli *et al*., 2008). Increased level of vitamin E in laying hens diet resulted in higher egg vitamin E level, increased egg production as well as laying duration (Gjorgovska *et al*., 2012). Similarly, egg selenium level was reported to increase with increased dietary selenium supplementation (Gjorgovska *et al*., 2012). It has been reported that selenium and vitamin E can also be deposited in the egg of laying hens (Laudadio *et al*., 2015). Vitamin E deposition in egg yolk is a reflection of its level in the breeder diet which varies with selenium supplementation (Surai, 2000). Zduńczyk *et al*. (2013) reported that increase in the levels of vitamin E and selenium in the diet from 30 to 60 mg/kg and 0.15 to 0.30 mg/kg, respectively, showed no effect on the number of eggs laid, average egg weight and yolk to albumen ratio. Scheideler *et al*. (2010) reported non-beneficial effects of vitamin E and selenium on feed intake or egg production variables when 0.55 of 0.75 ppm of selenium and 50, 100 or 150 IU/KG vitamin E were added to laying hens diet. Vitamin E or Selenium enriched yeast alone or in combination was discovered to be effective in improving the laying performance and egg quality of laying hens (El-Mallah *et al.,* 2011).

There have been conflicting reports on the levels of either vitamin E or selenium that could elicit appropriate performance and egg quality attributes. Thus, this study was aimed at assessing the effect of dietary supplementations of vitamin E and selenium on egg production, egg quality and the deposition of selenium and vitamin E in eggs.

MATERIALS AND METHODS

Experimental site and animal allotment

The experiment was carried out at the Poultry Unit of the Teaching and Research Farm, University of Ibadan, Ibadan, Nigeria. A total of 192 ISA Brown hens weighing 1.38 ±0.11kg at week 31 of age were used for the study. They were initially raised on a basal ration for two weeks and subsequently assigned to six experimental diets with two weeks of adjustment period in a completely randomized design. Each treatment was replicated four times with eight birds per replicate. The experiment lasted for ten weeks.

Data collection

Eggs were collected daily and recorded; hen day egg production was calculated as the number of eggs laid divided by number of chickens at the period of data collection. Feed intake was calculated by subtracting left over from feed offered to the chickens. Eggs were weighed using electronic top loading scale (JS-B LCD® Display Scale). Egg mass was calculated from the percent hen day egg production and average egg weight. Weight (g) of the eggs and its shell were determined using an electronic top loading scale (JS-B LCD® Display Scale). Length with diameter of the eggs was measured with electronic digital Vernier caliper. The shell thickness (mm) with the membranes was also measured with electronic digital Vernier caliper and the average was determined after measurement at the sharp, broad and the middle part of the egg (Monira *et al.,* 2003). Shell ratio was assessed through the formulae: Shell ratio (%) = (Shell weight / Egg Weight) x 100 (Scott and Silverside, 2000).

An opening was created close to the sharp portion of the egg thereby allowing albumen and egg yolk passage into a flat plate. The yolk was carefully removed from the albumen with a spoon and thereafter placed in separate petri dish for measurement using an electronic top loading scale. Yolk and albumen lengths as well as diameters were measured using a

Vernier caliper. Albumen and yolk height was measured by putting the pointed tip of the Vernier caliper in the middle of the albumen and yolk, height of the fluid on the tip was marked and read on the digital screen of the Vernier caliper. Yolk colour was accessed with DSM Yolk Colour Fan which has colour scale from 1-15 (Monira *et al.,* 2003).

Haugh Unit score was calculated from the egg weight and albumen height, using the equation:

HU = 100 log (H – 1.7w0.37 + 7.6)

Where: HU – Haugh Unit, H – observed height of albumen in mm, w – weight of egg in g (Haugh, 1937). Egg samples (n=3 per treatment) were collected and analyzed for vitamin E and selenium content of the yolk according to the procedure of AOAC (2006).

Statistical analyses

Data were subjected to descriptive statistics, polynomial regression and analysis of variance (SAS, 2001), while means were separated using Duncan's multiple range option of the same software at $\alpha_{0.05}$.

Experimental diets

Basal diet was supplemented with selenium and vitamin E as follows: T1-Basal diet, T2-Basal diet with 0.5 mg selenium/kg, T3- Basal diet with 1.0 mg selenium/kg, T4- Basal diet with 1.5 mg selenium/kg, T5-Basal diet with 20 mg vitamin E/kg, T6-Basal diet with 40 mg vitamin E/kg*)*. Feed and water were supplied *ad libitum* to the chickens. Dietary formulation has been documented (Jemiseye, 2018; Jemiseye and Ogunwole, 2018) Details of the dietary feed composition are shown in shown Table 1.

Table 1: Composition of basal experimental diet fed to laying hens

Ingredient	% inclusion (g/100gDM)
Corn	50.00
Soya bean meal	22.00
Wheat offal	11.00
Palm kernel cake	11.24
Oyster shell	3.03
Di-calcium phosphate	1.70
Vitamin-mineral premix	0.25
Table salt	0.30
Mycofix ®	0.15
Biotronics	0.30
DL-Methionine	0.15
L-lysine	0.15
Total	**100.00**
Calculated nutrients	
ME (Kcal/kg)	2702.90
Crude protein (%)	17.39
Crude fibre (%)	5.25
Methionine (%)	0.40
Lysine (%)	0.98
Calcium (%)	1.80
Available phosphorus (%)	0.52

Premix - Vitamin A–10,000,000IU, Vitamin D3–500IU, Vitamin E–40,000mg, Vitamin K–2,000mg, Vitamin B1–1,500mg, Vitamin B2–4,000mg, Vitamin B6–40,000mg, Vitamin B12–200mg, Niacin–40,000mg,Panthothenic–10,000mg,Folic–1,000mg, Biotin–100mg, Choline Chloride–300,000mg,Manganese–80,000mg, Zinc–60,000mg,Iron–40,000mg,Copper–80,000mg, Iodine–800mg, Selenium–200mg, Cobalt–300mg, Antioxidant–100,000mg*

RESULTS AND DISCUSSION

Performance of laying hens fed diets supplemented with selenium and vitamin E

Table 2 shows the performance of laying chickens fed diets supplemented with selenium and vitamin E. Feed intake, egg weight and egg mass were not affected by dietary supplementations of vitamin E and selenium. Hen day egg production varied with supplemental selenium and vitamin E. The HDEP of 85.02% in chickens on T2 was similar (p>0.05) to 77.83 % in those on T1, 74.73% in T3, 75.30 % in T5 and 70.82 % in T6 but significantly higher than 67.40 % in T4.

The increased egg production from 67.40% in hens on vitamin E supplementation to 85.02% due to supplemental selenium agreed with the observation of Mohiti-Asli *et al.* (2010), on increased egg production in single comb white leghorn hens raised at temperature of 24 ^{0}C. No significant differences (p>0.05) were observed in feed intake, egg weight or egg mass, regardless of the levels of supplemental selenium or vitamin E supplementation in this trial.

Adebiyi *et al*. (2014) reported that the performance of turkey in respect of feed intake and body weight gain were not affected significantly by the dietary treatment of vitamin E and selenium. Scheideler *et al.* (2010) also observed no significant effects of varying dietary levels of supplemental α-tocopherol or selenium on feed intake, feed efficiency, egg weight, or hen body weight. However, Ciftci *et al*. (2005) noted an increased egg weight with vitamin E supplementation.

Table 2: Performance of laying hens fed diets supplemented with selenium and vitamin E

Parameters	T1	T2	T3	T4	T5	T6	SEM
Hen day egg production (%)	77.83^{ab}	85.02^{a}	74.76^{ab}	67.40^{b}	75.30^{ab}	70.82^{ab}	2.14
Feed intake (g)	99.99	100.99	100.97	99.39	96.26	100.38	1.19
Egg weight (g)	60.15	62.89	58.01	62.68	64.79	61.76	1.05
Egg mass	46.69	53.25	43.55	42.34	49.19	43.92	1.65

$^{a, b,}$ means in the same row with different superscripts are significantly different (P<0.05). SEM-standard error of mean,T1-Control, T2-0.5 mg/kg selenium, T3-1.0 mg/kg selenium, T4-1.5 mg/kg selenium, T5-20 mg/kg vitamin E,T6-40 mg/kg vitamin E

External and internal quality characteristics of eggs from hens fed supplemental selenium and vitamin E

External quality characteristics of eggs from chickens fed varying supplemental levels of selenium and vitamin E in the diets are presented in Tables 3 and 4. Egg length and shell thickness were not significantly (p>0.05) affected by dietary supplements. There was significant influence (p<0.05) of supplemental selenium and vitamin E on egg weight, egg diameter, shell weight, and shell ratio. Hens on T6 produced eggs with higher (p<0.05) weight (62.56 g) and diameter compared to others. Shell weight (6.03 g) and shell ratio (10.50) of eggs from hens on T1 was higher and similar (p>0.05) to 5.70 g and 9.77 obtained for shell weight and shell ratio of eggs in hens on T5.

Internal quality characteristics of eggs from hens fed supplemental selenium and vitamin E is shown in Table 4. The dietary supplements did not affect (p>0.05) albumen weight, albumen diameter, yolk height, yolk colour and yolk weight to albumen weight but varied significantly ($p<0.05$) with albumen length, albumen height, yolk weight, yolk diameter, yolk ratio, and Haugh unit. The albumen length of eggs from hens on T2 (110.11 mm) was similar to those on T1 (102.43 mm) and T6 (101.02 mm) but significantly higher ($p<0.05$) than those on T4 (90.77 mm) and T5 (84.82 mm). Higher albumen height was obtained from eggs produced by hens on T5 (6.45 mm) and T6 (6.02 mm) than those on T1 (3.76 mm). Hens on T6 (15.13 g) produced similar ($p>0.05$) egg yolk weight with those on T3 (14.70 g) and T5 (14.23 g) but higher than those on T2 (12.66 g). Eggs from hens on T6 (42.62 mm) and T3 (42.23 mm) had increased yolk diameter similar to those on T1 (40.81 mm) and T5 (41.36 mm), while yolk diameter reduced in eggs from T2 (39.46 mm) and T4 (39.83 mm). Yolk ratio was significantly higher ($p<0.05$) in hens on T3 (25.19) than T1 (22.92) and T4 (24.17). Haugh unit of 80.04 and 76.85, recorded in eggs from hens on T5 and T4 were similar ($p>0.05$) to those from T2 (69.35), T3 (68.53) and T6 (75.34) but significantly higher ($p<0.05$) than 56.91 in T1.

Increased egg weight recorded with vitamin E supplementation in this trial could have resulted from increased yolk weight, albumen height and yolk diameter. The egg size increased as hen ages unless nutritionally manipulated. Increase in egg weight of birds fed supplemental vitamin E at 40 mg/kg suggests the positive effects of vitamin E on egg size at the mid laying phase. However, results from this trial negated the findings of Zduńczyk *et al.* (2013) who demonstrated that egg production and egg weight were not significantly affected by the dietary levels of vitamin E. Reduced egg weight obtained from hens on dietary selenium supplementation conformed to the findings of Mohiti-Asli (2010) on reported reduction in egg production in hens exposed to high temperature.

A quality egg must be above 60 Haugh units (HU) and eggs with Haugh units lower than 30 are not meant to be consumed as a table egg (Faris *et al.*, 2011). The HU will decrease with increasing bird age, with HU decreasing by around 1.5 to 2 units (Coutts and Wilson, 1990) for each month in lay. The mechanism behind the decrease in HU is that carbon dioxide which is lost from the egg contents by diffusion and resultant rise in the pH of the albumen correlates with the increased shell porosity and surface area (Williams, 1992; Brake *et al*., 1997).

Higher Haugh unit recorded indicated superior albumin in egg of hens fed T4 and T5. Elevated Haugh unit from hens on diets containing 1.5mg/kg and 20mg/kg selenium suggests a positive effect of antioxidant nutrients on absorption and protection of fat-soluble compounds.

A change in egg quality can be affected by many factors, including stress, age, and diet of the hen. In a trial of Pappas *et al*. (2005), eggs from pre peak and peak production were stored for 2 weeks under typical conditions. The HU, pH, shell characteristics, egg components, weight, Se content, and fatty acid profile were thereafter measured. Albumen HU decreased with storage, although eggs from high Se treatments had greater HU compared with the low Se treatments (Scott and Silversides, 2000). Patton *et al.* (2000) reported that sodium selenite or selenium yeast supplementation of 0.3 ppm had no effect on HU compared with eggs from hens on basal diet. Zduńczyk *et al*. (2013) however, reported that different dietary levels of selenium and vitamin E had no significant effect on the yolk to albumen ratio and egg protein quality.

In Figure 1, there was a positive linear relationship between selenium supplementation and Haugh unit of eggs. The relationship between supplemental dietary selenium and Haugh unit is shown in Figure 1 and represented by the equation:

$y = -4.1333x^2 + 18x + 58.03$…… ($R^2 = 0.3718$)…………………………………....1

This indicated that the higher the dietary supplement of selenium, the higher the Haugh unit. Also, supplemental dietary vitamin E correlated negatively and quadratically with Haugh unit in Figure 2. The supplemental dietary vitamin which engendered optimum Hu was 26 mg/kg vitamin E and is represented in equation 2 below;

$y = -0.0348x^2 + 1.8529x + 56.91$…………… ($R^2 = 0.6114$)……………………………….. 2

Table 3: External quality characteristics of eggs from hens fed supplemental selenium and vitamin E

Parameters	T1	T2	T3	T4	T5	T6	SEM
Egg weight (g)	57.36[ab]	55.26[b]	58.46[b]	54.73[b]	58.36[ab]	62.56[a]	0.94
Egg length(mm)	56.26	54.59	55.72	54.78	56.36	57.37	0.41
Egg diameter(mm)	42.75[ab]	42.25[b]	43.49[ab]	42.37[b]	43.20[ab]	44.22[a]	0.23
Shell thickness(mm)	0.38	0.41	0.39	0.39	0.41	0.40	0.00
Shell weight (g)	6.03[a]	5.30[b]	5.56[ab]	5.13[b]	5.70[ab]	5.73[ab]	0.09
Shell ratio	10.50[a]	9.63[ab]	9.56[ab]	9.38[ab]	9.77[ab]	9.18[b]	0.15

[a, b,] means in the same row with different superscripts are significantly different (P<0.05). SEM-standard error of mean,T1-Control, T2-0.5 mg/kg selenium, T3-1.0 mg/kg selenium, T4-1.5 mg/kg selenium, T5-20 mg/kg vitamin E,T6-40 mg/kg vitamin E

Table 4: Internal quality characteristics of eggs from hens fed supplemental selenium and vitamin E

Parameters	T1	T2	T3	T4	T5	T6	SEM
Albumen weight (g)	35.86	37.56	36.00	31.59	35.63	38.56	0.16
Albumen length (mm)	102.43[ab]	110.11[a]	99.77[abc]	90.77[cb]	84.82[c]	101.02[ab]	2.60
Albumen diameter (mm)	78.35	82.60	80.86	67.10	66.70	81.19	2.38
Albumen height (mm)	3.76[b]	4.76[ab]	5.06[ab]	5.86[ab]	6.45[a]	6.02[a]	0.31
Yolk weight (g)	13.13[bc]	12.66[c]	14.70[ab]	13.23[bc]	14.23[ab]	15.13[a]	0.27
Yolk height (mm)	3.76	4.76	5.06	5.86	6.45	6.02	0.35
Yolk diameter (mm)	40.81[ab]	39.46[b]	42.23[a]	39.83[b]	41.36[ab]	42.62[a]	0.36
Yolk colour	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Yolk ratio	22.92[b]	23.56[ab]	25.19[a]	24.17[ab]	24.41[ab]	23.22[ab]	0.29
Yolk W: Albumen W	0.37	0.35	0.40	0.42	0.40	0.39	0.01
Haugh unit	56.91[b]	69.35[ab]	68.53[ab]	76.85[a]	80.04[a]	75.34[ab]	2.72

[a, b, c] means in the same row with different superscripts are significantly different (P<0.05). SEM-standard error of mean,T1-Control, T2-0.5 mg/kg selenium, T3-1.0 mg/kg selenium, T4-1.5 mg/kg selenium, T5-20 mg/kg vitamin E,T6-40 mg/kg vitamin E

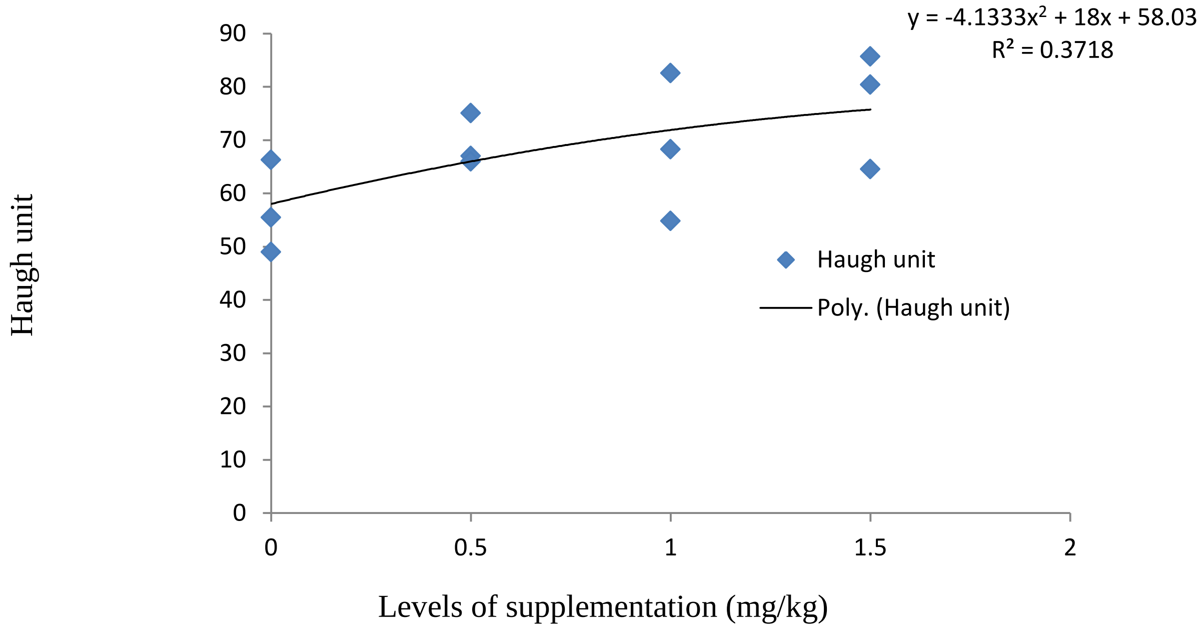

Figure 1: Effect of the relationship between dietary selenium supplementation and egg Haugh unit

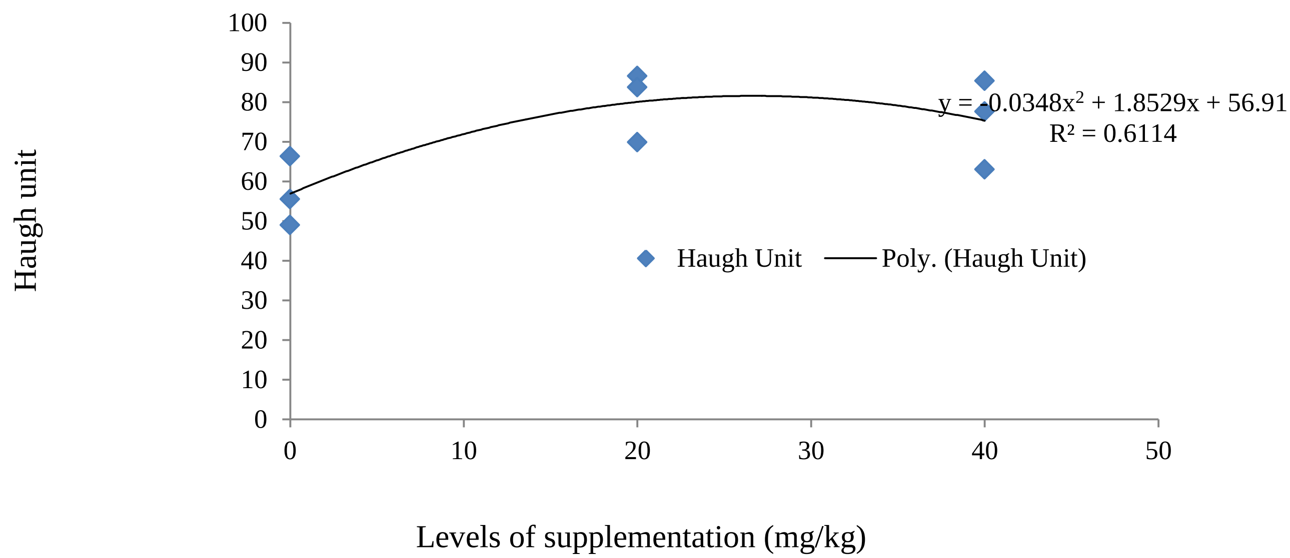

Figure 2: Effect of the relationship between dietary vitamin E supplementation and eggs Haugh unit

Selenium and vitamin E deposition in eggs of hens fed supplemental selenium and vitamin E

Table 5 shows selenium and vitamin E deposition in eggs of hens fed diets containing supplemental levels of selenium and vitamin E. Supplemental selenium showed no influence (p>0.05) on selenium deposition in the eggs. Supplemental vitamin E influenced (p<0.05) vitamin E deposition in eggs. Highest vitamin E deposition was recorded in eggs from hens on T6 similar to values obtained in those on T2 (1.12), T3 (1.13), T4 and T5 (1.13) (1.14). A decreased egg vitamin E deposition was observed in eggs from hens on T1 (1.08).

The increased vitamin E deposition in the yolk with increasing dietary supplement of vitamin E corroborates the findings of Jiang *et al*. (1994) that a linear relationship exists between dietary DL-α tocopheryl acetate level and egg yolk concentration of α-tocopherol. Conversely, Utterback *et al*. (2005) reported that the use of organic selenium in laying hen diets was very effective for increasing the selenium content of eggs. The authors further reported that dietary supplement of organic selenium for laying hens resulted in higher egg selenium concentrations than in eggs of hens on basal diets. Scheideler *et al.* (2010) documented a linear increase in yolk tocopherol with vitamin E supplementation and that yolk selenium content increased with dietary selenium supplementation. They also noted that deposition was more efficient when organic source of selenium was fed compared with inorganic.

Table 5: Selenium and vitamin E deposition in eggs of hens fed supplemental selenium and vitamin E

Parameters	T1	T2	T3	T4	T5	T6	SEM
Selenium (µg/100g)	1.72	1.94	1.73	1.80	1.86	1.78	0.04
Vitamin E (mg/100g)	1.08^{b}	1.12^{ab}	1.13^{ab}	1.14^{ab}	1.13^{ab}	1.26^{a}	0.20

a, b, means in the same row with different superscripts are significantly different (P<0.05). SEM-standard error of mean,T1-Control, T2-0.5 mg/kg selenium, T3-1.0 mg/kg selenium, T4-1.5 mg/kg selenium, T5-20 mg/kg vitamin E,T6-40 mg/kg vitamin E

CONCLUSION

The quality attributes of eggs were enhanced by dietary supplement of vitamin E and selenium. Selenium supplementation in the diet at 0.5 mg/kg increased hen day egg production while 40 mg/kg supplemental vitamin E improved egg quality parameters and egg vitamin E deposition.

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