J.Animal and Poultry Prod., Mansoura Univ., Vol.7(10): 393- 401,2016

Effect of Some Sources of Antioxiadnts on The Productive and Reproductive Performance of Turkey Hens

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ABSTRACT

The objective of this study was to investigate the effect of using some sources of antioxidants on the productive and reproductive performance of turkeys. This study was carried out at Mehalet Mousa Animal Production Research Station, Kafr El-Sheikh, belonging to the Animal Production Research Institute, Agricultural Research Center, Ministry of Agriculture, Egypt. A total of 84 Bronze female turkeys, 32 weeks of age were used. The birds were randomly distributed into 7 experimental groups; each consisted of three equal replicates. The birds were individually housed in battery cages. The experimental period lasted 12 weeks (from 32 to 44 weeks of age). The birds were fed a yellow corn-soybean meal-based diet, supplemented with two antioxidants, zinc and selenium of three sources organic, inorganic and nano forms. The experimental groups were fed one of the following diets: (1) a basal diet without added zinc or selenium; (2) inorganic zinc (zinc oxide) 100 mg/kg diet; (3) organic zinc (zincmethionine) 100 mg/kg diet; (4) nano zinc (zinc oxide nanoparticles) 40 mg/kg diet; (5) inorganic selenium (sodium selenite) 0.3 mg/kg diet; (6) organic selenium (selenomethionine) 0.3 mg/kg diet; (7) nano selenium (selenium nano-particles) 0.15 mg/kg diet. The results obtained can be summarized as follows. Dietary supplementation with inorganic, organic and nano forms of zinc and selenium significantly ($P \leq 0.05$) increased serum levels of total protein, concentrations of blood haemoglobin, egg fertility, hatchability of eggs in turkey hens, and significantly ($P \leq 0.05$) improved feed conversion ratio as compared to that of the controls. Dietary supplementation with inorganic, organic and nano forms of zinc and selenium significantly ($P \leq 0.05$) decreased feed consumption, serum levels of total lipids and cholesterol as compared to those of the controls. Supplementation of organic and nano forms of zinc and addition of inorganic, organic and nano forms of selenium to diets of laying turkey hens significantly ($P \leq 0.05$) increased hen-day egg production rate and egg weight as compared to those of the controls. Dietary organic and nano forms of zinc and selenium of turkey hens caused significant increases ($P \leq 0.05$) in serum antibody titres against Newcastle disease virus and improved immunity, and exhibited a significant increase in relative weight of spleen and oviduct length compared with their control group. Turkey hens fed diets supplemented with organic and nano Se produced eggs with better albumen quality and egg shell quality as measured by Haugh units and egg shell thickness as compared to that of the control turkey hens. There were no significant differences among the different dietary treatments in relative weights of egg components, egg shape index, egg yolk index, yolk color score, relative weights of carcass yield, liver and heart of turkey hens. It can be concluded that dietary supplementation with organic and nano forms of zinc and selenium can induce beneficial effects on productive and reproductive performance of turkey hens.

INTRODUCTION

Free radicals are typically atoms containing one or more unpaired electrons. Most biologically-relevant free radicals are derived from oxygen and nitrogen, the so-called reactive oxygen species (ROS) and reactive nitrogen species (RNS). Both these elements are essential for life of organisms but in certain circumstances they are converted into free radicals which are highly unstable and their reactive capacity makes them capable of damaging biologically relevant molecules such as DNA, proteins, lipids or carbohydrates (Surai, 2002).

The formation of free radicals is a patho-biochemical mechanism involved in the initiation or progression of various diseases (Hogg, 1998). In livestock production, free radical generation and lipid peroxidation are responsible for the development of various diseases and decreasing the animal productivity, and product quality (McDowell, 2000).

The processes of oxidation and reduction are necessary in the biochemistry of the body. This gain or removal of an electron keeps many of the life processes working. As respiration occurs in animals, which is defined as the process from which cells derive energy in the form of ATP from the reactions of hydrogen and oxygen, they often produce various peroxides. These peroxides, including hydrogen peroxide, can be harmful to the body as they can lead to generation of free radicals, which can damage or destroy cells (Arthur, 2000).

Majority of trace minerals in commercial poultry diets are commonly supplemented in the form of inorganic salts, such as sulfates, oxides and carbonates, to provide levels of minerals that prevent clinical deficiencies and allow the birds to reach their genetic growth potential. Organic complexed mineral is a type of mineral linked to protein/peptide/amino acids that has a higher bioavailability than the inorganic salts (Swiatkiewicz *et al.* 2014).

Recently, nanotechnology and related products had rapid progress in different scientific areas. In fact this branch of science has fundamental effects on all aspects of life, and environment. Nanoscale of materials like nanosilver (as antimicrobial), nano-selenium and zinc oxide nanoparticles has an increasing attention because nano-formulation particulates exhibit a distinguishing quality such as a size, shape, large surface area, high surface activity, high catalytic efficiency, and strong adsorbing ability (Wijnhoven *et al.* 2009).

A lot of supplements is used in poultry diets to improve the growing and productive performance of laying hens. Zinc as an essential trace mineral has significant functions in the organism, probably because it is a co-factor of more than 200 enzymes. So, the presence of adequate zinc in the higher biological systems is important for normal development, maintenance and function of the immune system in poultry. One of the most significant roles of zinc is related to its antioxidant function and its participation in the antioxidant defense system of the body (Powell, 2000). Zinc deficiency provokes oxidative damage through the effects of free radical action and alters the status antioxidant enzymes and substances (Salgueiro *et al.* 2000).

Zinc has numerous biological roles including DNA synthesis (Lieberman *et al.* 1963), cell division and multiplication (Rubin and Koide, 1973), protein metabolism (Forbes, 1984), the cell mediated immune response (Bertuzzi *et al.* 1998) and performance (Sadoval *et al.* 1999). Zinc is involved in boosting the immune system to disease outbreaks (Luecke *et al.* 1978).

Organic complexes of zinc have been proposed to be more available source of zinc for layer hens (Aliarabi *et al.* 2007). Zn-methionine had more bioavailability than inorganic zinc sources such as ZnO or $ZnSO_4{}^{\cdot}H_2O$ (Rahman *et al.* 2002). NRC (1994) recommended between 29-44 mg Zn/kg for laying strains of Leghorn-type laying hens. Higher Zn supplementation was found to be beneficial (Hudson *et al.* 2005). Ahmadi *et al.* (2014) found that the levels of zinc oxide nanoparticles from 60 to 90 mg/kg basal diet improved antioxidant state and serum enzymes of broiler chickens.

Selenium (Se) is a dietary essential trace mineral for poultry (NRC, 1994). Selenium is regarded as an essential trace element that exerts many functions in several biological processes in animals and birds. It is required for maintenance of growth, health, and physiological functions. It has been well-demonstrated to play vitally important roles in reproduction, immune function, antioxidant and redox reactions and muscle functions (Zhang *et al.* 2012). Supplementation of Se to diets is a common practice in poultry as well as in other animal species (Pavlovic *et al.* 2009). Selenium is required for maintenance of growth, health, and physiological functions.

Glutathione peroxidase (GSH-Px) has antioxidative action and contributes to the oxidative defense by catalyzing the reduction of hydrogen peroxide and lipid peroxides to less harmful hydroxides (Arthur, 2000). The activity level of this enzyme in the liver or plasma is indicative of the selenium supply to the organism; moreover, its antioxidant protection levels are affected by dietary Se status (Wang, 2009). Furthermore, Rotruck *et al.* (1973) showed that Se was essential for the proper function of the glutathione peroxidase enzyme. Combs G.F. and S.B. Combs (1986) indicated that inorganic sources of Se, such as sodium selenite or selenates, are passively absorbed, while organic sources, such as selenium yeast or selenomethionine, are actively absorbed via amino acid transport mechanisms. Organic Se is metabolized differently than inorganic Se (Sunde, 1997). Se-methionine is quickly absorbed with the consequence of higher blood levels in comparison to inorganic Se. This study aimed to investigate the effect of using some sources of antioxidants on the productive and reproductive performance of turkey hens.

MATERIALS AND METHODS

The experimental work of this study was performed at Mehalet Mousa Animal Production Research Station, Kafr El-Sheikh, belonging to the Animal Production Research Institute, Agricultural Research Center, Ministry of Agriculture, Egypt. This experiment was carried out to study the effect of using some sources of antioxidants on the productive and reproductive performance of turkey hens. A total of 84 Bronze female turkeys, 32 weeks of age were used. The birds were randomly distributed into 7 experimental groups, each treatment consisted of three equal replicates. The birds were individually housed in battery cages (65 × 50 cm) fitted with individual feeders and automatic nipple drinkers. The experimental period lasted 12 weeks (from 32 to 44 weeks of age). Birds were fed *ad libitum* and fresh water was available all the time, during the experimental period. Photoperiod was 16 hours daily during the experimental period. The feed ingredients and chemical composition are presented in Table 1. The experimental groups in this experiment were fed on one of the following

experimental diets:

T1: A basal diet (control group) without added Zn or Se.

T2: Basal diet + inorganic zinc (zinc oxide) 100 mg/kg diet.

T3: Basal diet + organic zinc (zincmethionine) 100 mg/kg diet.

T4: Basal diet + nano zinc (zinc oxide nanoparticles) 40 mg/kg diet.

T5: Basal diet + inorganic selenium (sodium selenite) 0.3 mg/kg diet.

T6: Basal diet + organic selenium (selenomethionine) 0.3 mg/kg diet.

T7: Basal diet + nano selenium (selenium nano particles) 0.15 mg/kg diet.

Egg weight and egg number and were recorded daily and feed intake was calculated weekly.Feed conversion (g feed/g egg) was also calculated on a weekly basis. Egg quality parameters were measured at the peak of egg production (38 weeks of age) during the experiment (10 eggs/ treatment). These parameters involved relative weight of egg components (yolk, albumen and shell) and some exterior and interior parameters of egg quality. Egg shell thickness was measured in mm using a micrometer. Egg shape index was calculated as the egg width times 100 divided by the egg length. Yolk index was also calculated as yolk height times 100 divided by yolk diameter. Haugh unit was measured according to the formula presented by Eisen *et al.* (1962). Yolk color score was determined by using a Roche yolk color fan.

Semen was collected from toms housed in floor pens and fed on the same experimental diets and artificially inseminated to turkey hens.The insemination was made using fresh semen as soon as possible after its collection. The artificial insemination was done once a week. All turkey hens were inseminated intra-vaginally. Raw semen was diluted at the rate of 1:1 with 0.9 %

NaCl. The insemination was performed by inserting 0.1 ml of the diluted semen with one millimeter tuberculin syringe into the vagina of each hen. Fertility and hatchability of eggs were made every 3 weeks of the experimental period. Fertility was calculated as the number of fertile eggs relative to total number of eggs set; while egg hatchability was calculated as the number of healthy hatched chicks relative to total number of eggs or to the total fertile eggs.

Table 1. Composition and calculated analysis of the basal diet used in this Study

Ingredients	%
Yellow corn	70.00
Soybean meal (44%CP)	11.50
Fish meal (65%CP)	10.00
Di calcium phosphate	2.00
Limestone	6.00
Salt (NaCl)	0.30
L-Lysine.Hcl	0.15
DL.Methionine	0.05
Total	100
Calculated analysis(As-fed basis; NRC, 1994)	
Crude protein %	17.72
Metabolizable Energy(kcal/kg diet)	2920

*** Supplied per kg of diet: Vit. A, 12000 IU; Vit. D_3, 2200 IU; Vit. E, 10 mg; Vit K_3, 2mg; Vit. B1, 1mg; Vit. B_2, 5mg; B_6, 1.5mg; B_{12}, 10 mg; Nicotinic acid, 30mg; Folic acid, 1mg, Pantothenic acid, 10mg; Biotin, 50 mg; Choline, 250mg; Copper, 10mg; Iron, 30mg; Manganese, 60mg; Iodine, 0.3 mg and Cobalt, 0.1mg.**

In this experiment, blood samples were collected from 44-week-old turkey hens (3 specimens per treatment) by venipuncture of the wing vein. Immediately after collection, each blood sample was transferred into two tubes, a heparinized tube and non-heparinized one. Heparinized whole blood samples were used for the determination of hemoglobin concentration according to Campbell (1995) and haematocrit value, as described by Hunsaker (1969).

The separated sera from non-heparinized blood samples were used for the colorimetric estimation of levels of glucose (Trinder, 1969), total protein (Henry *et al.* 1974), albumin (Doumas *et al.* 1971), cholesterol (Watson, 1960) and total lipids (Zollner and Kirsch, 1962), using commercial kits. The concentration of serum globulin was calculated by the difference between serum total protein and albumin.

Turkey hens were vaccinated against Newcastle disease virus by inactivated Newcastle vaccine at 30 weeks of age, serum samples from layers at 44 wks of age were used for determination of Newcastle disease virus antibodies titer, using methods described by (Liu *et al.*, 1999).

At the end of experimental period (44 wks old), 3 hens from each treatment were randomly selected and slaughtered for carcass evaluation. Carcass was eviscerated and head and shanks were removed. Carcass yield and some internal organs were expressed as percentage of live body weight. The oviduct length was also estimated.

Statistical analysis:

Data were statistically analyzed by one-way analysis of variance using the General Linear Model (GLM) procedures (SAS, 2003). Significant differences among means of treatments for different variables were detected by Duncan's multiple range test (Duncan, 1955). The following model was used to study the effect of inorganic, organic and nano forms of zinc or selenium on parameters investigated as follows:

$$Y_{ij} = \mu + T_i + e_{ij}$$

Where, Y_{ij} = an observation

μ = overall mean

T_i = treatments (i = 1, 2 … and 7)

e_{ij} = residual "random error".

RESULTS AND DISCUSSION

Productive performance of turkey hens:

Data on the effect of dietary organic, inorganic and nano forms of zinc and selenium on productive performance (in terms of feed intake, hen-day egg production rate, egg weight and feed conversion ratio) of turkey laying hens from 32 to 44 weeks of age are given in Table 2.

As presented in Table 2, the results indicated that turkey hens fed the diets supplemented with inorganic, organic and nano forms of zinc and selenium significantly ($P \leq 0.05$) decreased feed consumption compared with the control group during the whole experimental period. Supplementation of organic and nano forms of zinc and addition of inorganic, organic and nano forms of selenium to diets of laying turkey hens significantly ($P \leq 0.05$) increased hen-day egg production rate compared with that of the control group, during period from 32-44 weeks of age. The highest mean of hen-day egg production rate was achieved by turkey hens fed the organic Se-supplemented diet, followed by that of turkey hens fed the diet supplemented with nano Se, nano Zn, inorganic Se and organic Zn in a descending order, respectively. Turkey hens fed the diets supplemented with organic, inorganic and nano Se, and organic and nano Zn produced significantly heavier eggs ($P \leq 0.05$) compard with those of the control group. Dietary supplementation of inorganic, organic and nano forms of zinc and selenium to turkey hens significantly improved feed conversion ratio ($P \leq 0.05$) compard with the control group during the whole the experimental period. The best means of feed conversion ratio was attained by hens fed the diets supplemented with organic and nano forms of selenium compared with that of the control hens.

These results agree with the findings of Namra *et al.* (2009), who showed that the dietary addition of 50 mg Zn (from the zinc-methionine)/kg diet of laying Japanese quail significantly reduced feed intake compared with the control group. Also, Idowu *et al.* (2011) indicated that dietary supplementation with inorganic Zn at 140 mg/kg decreased significantly the feed consumption of laying birds as compared to their control ones.

On the other hand, the current results disagree with the findings of Abdallah *et al.* (2014), who reported that supplemental dietary organic Zn to Gimmizah laying hens had no significant effect on their feed intake compared with the control birds. Also, Jing *et al.* (2015) showed that feed intake was not influenced by supplementation of different source of Se at 131-day-old brown laying hens. The different physiological effects of nano forms of Zn and Se might be related to differences in the absorption process and metabolic pathways. Nanoparticle shows new characteristics of transport and uptake and can exhibit higher absorption efficiencies (Liao *et al.*, 2010). They suggested that the superior performabce of nanoparticles may be attributed to their smaller particle size and larger surface area, increased mucosal permeability, improved intestinal absorption and tissue deposition.

The present results agree with those of Radwan *et al.* (2015), who showed that nano-Se supplementation in layer diets significantly increased egg production percentage compared with inorganic Se (sodium selenite) of Silver Montazah laying hens at 32 weeks of age. Similarly, Gjorgovska *et al.* (2012) who found that supplemental dietary organic selenium in layer diets increased egg production percentage.

Table 2. Effects of dietary inorganic, organic and nano forms of zinc and selenium on productive performance of turkey hens from 32 to 44 weeks of age

Dietary Treatments	Feed intake (g)	Hen-day Egg Production (%)	Egg weight (g)	Feed conversion ratio (g feed / g egg)
Control (Basal diet)	129.94^{a}	55.24^{f}	74.24^{d}	3.16^{a}
Basal diet + Inorganic Zn	127.59^{b}	55.66^{ef}	74.28^{d}	3.08^{b}
Basal diet + Organic Z	119.49^{e}	56.00^{de}	75.18^{c}	2.83^{c}
Basal diet + Nano Zn	117.66^{f}	62.12^{c}	75.46^{c}	2.50^{e}
Basal diet + Inorganic Se	126.18^{c}	56.33^{d}	81.88^{a}	2.73^{d}
Basal diet + Organic Se	121.16^{d}	70.09^{a}	80.70^{b}	2.13^{f}
Basal diet + Nano Se	117.88^{f}	69.10^{b}	81.36^{ab}	2.09^{f}
SEM	0.32	0.17	0.25	0.015
Significance	*	*	*	*

a-f: Means in the same column bearing different superscripts differ significantly (P≤0.05).
***: Significant at P≤0.05 SEM: Standard error of the means**

Also, Hanafy *et al.* (2009) reported that egg production percentage was significantly increased for hens fed organic Se supplementation of Bandarah local chickens at 56 and 60 weeks of age compared with the control group. Renema (2004) found that the addition of organic selenium source to the diet of layers improved egg production compared to inorganic Se and non supplemented treatments during the late laying period. Similarly, Bahakaim *et al.* (2014) showed that supplementation of organic zinc as zinc methionine of laying hens' diet significantly increased egg production of Golden Montazah laying hens at 24 weeks of age. Stanley *et al.* (2012) suggested that dietary supplementation with organic zinc (20 ppm/kg diet) significantly increased rate of egg production of White Leghorn laying hens at 78 weeks of age.

On the other hand, the present results disagree with those of Chantiratikul *et al.* (2008), who found that egg production was not affected by Se sources of Brown laying hens at 71 weeks of age. Cruz and Fernandez (2011) observed no differences in egg production percentage due to dietary inclusion of Se sources of Japanese quail. Attia *et al.* (2010) indicated that egg production percentage was not affected by dietary Se source.

The present results agree with those of Abdallah *et al.* (2014), who reported that hens fed diets supplemented with organic zinc at different levels produced the heaviest egg weights compared with those fed inorganic zinc-supplemented diet of Gimmizah laying hens. Renema (2004) found that the addition of organic selenium to the diet of layers improved egg weight compared to the non-supplemented treatments during the late laying period. Also, Sara *et al.* (2008) indicated that addition of organic Se in laying hen diets at 48-62 weeks of age improved egg weight. Stanley *et al.* (2004) showed that hens fed diet supplemented with organic Se significantly increased egg weight compared with the control group. Attia *et al.* (2010) indicated that the dietary addition of Se significantly increased egg weight compared with hens fed the control diet.

On the other hand, the present results disagree with those of Radwan *et al.* (2015), who showed that different levels of sodium selenite or nano-Se had no effect on egg weight of Silver Montazah laying hens. Similarly, Chantiratikul *et al.* (2008) found that egg weight was not affected by dietary Se sources of Brown laying hens. These results agree with those of Namra *et al.* (2009), who reported that the dietary addition of 50 mg Zn (from the zinc-methionine)/kg diet of Japanese quail hens achieved the best feed conversion ratio compared with the control diet. Osman and Ragab(2007) found that supplemental zinc-methionine at the level 0.3 g Zn Meth./kg diet of chicks gave the best feed conversion ratio during the period from 36 to 42 days of age. Also, Bahakaim *et al.* (2014) showed that supplemental organic zinc as Zn methionine (100 mg/kg) of laying hens gave the best feed conversion of Golden Montazah laying hens.

Kucuk *et al.* (2003) indicated that supplemental dietary zinc (30 mg/kg diet) as zinc sulphate to the basal diet improved feed conversion of broiler chicks. Ahmadi *et al.* (2014) showed that dietary addition of zinc oxide nanoparticles (ZONP) at levels of 60 mg/kg diet improved feed conversion ratio for the period from 1 to 21 days of age , than those of the control group. Also, our results agree with those of Zhou and Wang (2011), who indicated that feed conversion was improved by addition of 0.3 mg nano-Se/kg diets of Guangxi Yellow broilers. Similarly results were obtained by Radwan *et al.* (2015), who showed that dietary supplementation of Nano-Se to layer diets improved feed conversion ratio of Silver Montazah laying hens. They explained the positive effect of organic Se to that organic selenium could cross through intestine and enter into blood by active transport. The Selenomethionine from selenised yeast was well absorbed and was incorporated into body proteins in place of methionine. The extent of selenomethionine incorporation into proteins depends on the dosage and methionine status (Butler *et al.*, 1989).

The present results agree with the findings of Payne and Southern (2005), who found that mortality rate of broilers was not affected by addition of Se source (from organic or inorganic Se) or level of supplementation (0 or 0.30 mg/kg).

On the other hand, Stanley *et al.* (2012) found that dietary supplementation with organic Zinc (Bio-Plex®) at a level of 20 ppm significantly decreased rate of mortality of White Leghorn laying hens at 78 weeks old.

Egg Components and Egg Quality:

The present results showed that there were no significant differences among dietary treatments with respect to relative weights of egg albumen, egg yolk, egg shell, egg shape index, egg yolk index or yolk color score (Table 3).

The current results indicated that turkey hens fed diets supplemented with organic and nano Se produced eggs with better albumen quality as measured by Haugh units. No significant differences were observed in Haugh units among the other experimental groups of turkey hens (Table 3).

Supplementation of organic and nano Se to the diets of turkey hens significantly (P≤0.05) increased egg shell thickness and thus, improved egg shell quality as compared to that of the control turkey hens. No significant differences were observed in egg shell thickness among the other experimental groups of turkey hens (Table 3).

The current results agree with those of Abdallah *et al.* (2014), who indicated that supplemental dietary organic Se caused significantly higher egg shell thickness of Gimmizah laying hens as compared with those for inorganic form. In addition, Idowu *et al.* (2011) found that egg quality parameters were not significantly influenced by Zn sources except Haugh units values of laying hens. The present results agree also with those of Hanafy *et al.* (2009), who reported that egg quality (Haugh unit, and shell thickness) were significantly increased with supplemental dietary organic Se than control group of Bandarah laying hens. Similarly, Paton *et al.* (2000) indicated that supplemental dietary organic selenium improved egg shell strength at 80-week-old Babcock layers. In addition, Gajcevic *et al.* (2009) found that dietary supplementation of organic selenium increased Haugh units values compared with eggs produced of hens fed the control diet. Also, Aliarabi *et al.* (2007) showed that weights of egg components and shell thickness were not significantly influenced by supplementation of organic zinc of the diets but values of Haugh units was higher in the groups received organic zinc than that of hens fed the control diet. Similarly, Cruz and Fernandez (2011) indicated that there were no significant differences in yolk index, albumen index and egg shell weight of Japanese quails by dietary supplementation with Se. Gjorgovska *et al.* (2012) showed that the percentages of yolk, albumen and shell egg were not affected by different sources or levels of supplemental Se.

Table 3. Effects of dietary inorganic, organic and nano forms of zinc and selenium on egg components and egg quality of 38 weeks old turkey hens

Dietary treatments	Egg Components (%)			Egg quality parameters				
	Egg albumen %	EggYolk %	Egg shell %	Egg shape index%	Egg yolk index%	Haugh units	Shell thickness (mm)	Yolk Color score
Control (Basal diet)	52.69	30.92	16.37	74.04	53.09	89.62[c]	0.377[b]	7.50
Basal diet + Inorganic Zn	52.54	30.85	16.58	73.69	52.55	90.40[c]	0.383[b]	7.60
Basal diet + Organic Zn	52.89	30.18	16.91	76.66	53.82	91.11[abc]	0.385[b]	7.30
Basal diet + Nano Zn	53.18	30.25	16.54	77.15	52.95	91.02[abc]	0.389[b]	7.40
Basal diet + Inorganic Se	53.38	30.14	16.45	74.76	52.82	92.92[abc]	0.377[b]	7.40
Basal diet + Organic Se	52.25	29.42	18.30	78.34	52.94	96.49[a]	0.419[a]	7.50
Basal diet + Nano Se	52.05	30.16	17.75	78.02	52.23	95.58[ab]	0.419[a]	7.40
SEM	0.94	1.16	1.76	1.46	1.27	1.74	0.007	0.24
Significance	NS	NS	NS	NS	NS	*	*	NS

a-c: Means in the same column bearing different superscripts differ significantly (P ≤0.05).
NS: Not significant *: significant at P≤0.05 SEM: Standard error of the means

On the other hand, the present results disagree with those of Bahakaim *et al.* (2014), who showed that albumen index and Haugh units were significantly affected by supplementation of zinc methionine (100 mg/kg diet) of Golden Montazah laying hens but shell thickness significantly improved by addition of organic Zn in diet. Similarly, Attia *et al.* (2010) found that dietary supplementation of organic or inorganic selenium had no significant effect on any traits of egg quality. Also, Chantiratikul *et al.* (2008) found that Haugh units and egg shell thickness were not changed by source of Se addition of Brown laying hens.

Blood parameters of turkey hens:

Data on the effect of dietary organic, inorganic and nano zinc and selenium on blood biochemical and haematological parameters of 44-week–old turkey hens are shown in Table 4. The present results showed that no significant differences were observed in

serum glucose levels by addition inorganic, organic and nano forms of zinc and selenium compared than controls, and the highest value of serum level of glucose for hens was achieved by hens fed diet supplemented with organic Se and the lowest value of serum level of glucose for hens was achieved by hens fed diet supplemented with inorganic Zn. Serum levels of total protein and concentrations of blood haemoglobin for hens fed diets supplemented with inorganic, organic and nano forms of zinc and selenium were significantly higher (P≤0.05) than those of the controls. Serum levels of albumin for hens fed diet supplemented with organic Zn was significantly higher (P≤0.05) than those of the controls. No significant differences were observed in serum albumin levels among the remaining experimental groups of turkey hens. Serum globulin concentrations of hens fed the diets supplemented with nano Zn and nano Se were significantly higher (P≤0.05) than those of the control group, however, other experimental groups of hens exhibited insignificantly comparable levels of globulin to that of the controls. Dietary addition of inorganic, organic and nano forms of zinc and selenium for hens caused significant reductions (P≤0.05) in serum levels of total lipids and serum cholesterol levels as compared to that of the controls. Dietary supplementation with inorganic and nano Se resulted in significant increases (P≤0.05) in blood haematocrite values (%) of turkey hens compared with that of the controls.

Dietary organic and nano forms of zinc and selenium of turkey hens caused significant increases (P≤0.05) in serum antibody titres against Newcastle disease virus (NDV) as compared to that of the controls; other experimental groups of hens exhibited insignificantly comparable levels of serum antibody titres against NDV to that of the controls. So, these results indicated that addition of organic and nano forms of zinc and selenium as an adjuvant to Newcastle disease vaccine played an important role to improve the level of immunity, as measured by antibodies titers in serum for hens.

The present results agree with those of Hassan *et al.* (2003), who found that supplemental dietary zinc methionine to the basal diet of Mandarah laying hens significantly increased serum total protein and albumin as compared to the control group. Ibs and Rink (2003) showed that total protein and albumin were significantly increased when organic zinc was added to the diet of birds. Bahakaim *et al.* (2014) indicated that supplemental dietary zinc methionine (100 mg/kg diet) to the basal diet of Golden Montazah laying hens significantly increased total protein and albumin and improved albumin : globulin ratio. El-Sheikh *et al.* (2010) showed that supplemental dietary organic selenium at 0.2 and 0.3 ppm significantly increased the blood level of hemoglobin.

Table 4. Effects of dietary inorganic, organic and nano forms of zinc and selenium on blood biochemical and hematological parameters of turkey hens at 44 weeks of age

Dietary treatments	Glucose (mg/dl)	Total protein (g/dl)	Albumin (g/dl)	Globulin (g/dl)	Total Lipids (g/dl)	Cholesterol (mg/dl)	Hemoglobin (g/dl)	Haematocrit (%)	NDV titre
Control (Basal diet)	354.66 ^{abc}	4.53 ^{d}	2.74 ^{b}	1.78 ^{b}	3.60 ^{a}	295.00 ^{a}	9.28 ^{d}	35.60 ^{b}	9.05 ^{c}
Basal diet+ Inorganic Zn	348.33 ^{c}	5.26 ^{bc}	3.18 ^{ab}	2.08 ^{b}	2.81 ^{b}	247.33 ^{b}	10.03 ^{c}	34.18 ^{b}	8.52 ^{c}
Basal diet + Organic Zn	358.66 ^{abc}	5.71 ^{ab}	3.50 ^{a}	2.21 ^{b}	1.87 ^{d}	185.00 ^{cd}	11.30 ^{b}	34.20 ^{b}	11.80 ^{b}
Basal diet + Nano Zn	364.33 ^{ab}	5.88 ^{a}	2.90 ^{ab}	2.98 ^{a}	1.55 ^{e}	177.33 ^{cd}	11.47 ^{b}	34.90 ^{b}	13.11 ^{a}
Basal diet + Inorganic Se	352.00 ^{bc}	5.03 ^{c}	2.66 ^{b}	2.37 ^{b}	2.53 ^{c}	239.66 ^{b}	10.13 ^{c}	37.68 ^{a}	9.23 ^{c}
Basal diet + Organic Se	366.33 ^{a}	5.48 ^{abc}	3.21 ^{ab}	2.27 ^{b}	1.73 ^{de}	187.66 ^{c}	11.59 ^{ab}	34.69 ^{b}	11.74 ^{b}
Basal diet + Nano Se	364.33 ^{ab}	5.85 ^{a}	2.86 ^{ab}	2.98 ^{a}	1.52 ^{e}	176.33 ^{d}	11.85 ^{a}	38.93 ^{a}	13.28 ^{a}
SEM	4.17	0.14	0.21	0.18	0.08	3.46	0.11	0.48	0.33
Significance	*	*	*	*	*	*	*	*	*

a-e: Means in the same column bearing different superscripts differ significantly (P ≤0.05).
***: Significant at P≤0.05 SEM: Standard error of the means NDV: Newcastle disease virus**

These results agree also with those reported by Radwan *et al.* (2015)), who found that supplemental dietary Nano-Se significantly decreased blood plasma levels of cholesterol and total lipids compared with that in hens fed the control diet of Silver Montazah laying hens. Attia *et al.* (2010) observed that blood plasma levels of cholesterol and total lipids were significantly decreased when inorganic or organic selenium were added to the diet of laying hens compared with those in hens fed the control diet. Sahoo *et al.* (2014) showed that dietary organic and nano zinc supplementation (15 ppm and 0.06 ppm respectively) to the basal diet of broiler birds significantly decreased plasma cholesterol concentration as compared to the control group.

The present results agree also with those of Kidd *et al.* (1996), who indicated that addition of zinc-methionine to the diet of laying hens may improve the immune system and augment disease resistance. However, zinc-methionine supplementation has beneficial effects on turkey macrophage function and humoral immunity (Kidd *et al.* 1992). Wang (2009) found that antibody titre against Newcastle disease increased by dietary nano-Se supplementation at levels between 0.15 and 1.2 mg/kg diet as compared to the un-supplemented controls.

Egg Fertility and Hatchability for turkey hens:

As presented in Table 5, percentages of egg fertility, hatchability of total eggs and hatchability of fertile eggs achieved by hens fed diets supplemented with inorganic, organic and nano forms of zinc and selenium were significantly higher (P≤0.05) than those of the controls.

Table 5. Effects of dietary inorganic, organic and nano forms of zinc and selenium on egg fertility and hatchability of turkey hens during the experimental period from 32-44 weeks of age

Dietary treatments	Egg Fertility (%)	Hatchability of total Eggs (%)	Hatchability of fertile Eggs (%)
Control (Basal diet)	93.22 ^{d}	88.38 ^{c}	94.78 ^{b}
Basal diet + Inorganic Zn	95.56 ^{bc}	92.09 ^{b}	96.36 ^{a}
Basal diet + Organic Zn	97.48 ^{a}	94.73 ^{a}	97.17 ^{a}
Basal diet + Nano Zn	97.76 ^{a}	94.77 ^{a}	96.93 ^{a}
Basal diet + Inorganic Se	95.44 ^{c}	91.89 ^{b}	96.27 ^{a}
Basal diet + Organic Se	97.18 ^{ab}	94.05 ^{ab}	96.77 ^{a}
Basal diet + Nano Se	97.14 ^{ab}	94.30 ^{ab}	97.06 ^{a}
SEM	0.52	0.79	0.39
Significance	*	*	*

a-d: Means in the same column bearing different superscripts differ significantly (P ≤0.05).
***: Significant at P≤0.05 SEM: Standard error of the means**

These results agree with those reported by Abdel-Galil and Abdel Samad (2004), who showed that dietary supplementation of Zn (100 mg/kg diet) improved fertility rate. Amen and Al-Daraji (2011a) found that 180 mg zinc/kg diet improved performance and hatchability traits of brown parent stock layers. Namra *et al.* (2009) showed that supplemental dietary 50 mg Zn-methionine /kg diet improved fertility of laying Japanese quail. Similarly, Amen and Al-Daraji (2011b) showed that Cobb 500 broiler breeders fed diets supplemented with 50, 75 or 100 mg Zn/kg of diet significantly surpassed the control group as concerns percentages of fertility, hatchability of total eggs and hatchability of fertile eggs, and the best values of these three traits were recorded with birds received the diet supplemented with 100 mg Zn/kg of diet.

The present results agree also with those of Agate *et al.* (2000), who observed that dietary organic Se supplementation of laying hen diets improved the environment of the sperm storage tubules in the hen's oviduct, allowing sperm to live longer, increasing the length of time sperm can be stored. Hanafy *et al.* (2009) indicated that supplemental dietary organic Se of laying hens increased the hatchability of fertile eggs. The presence of antioxidants (zinc and selenium) could partially interfere with oxidative protein denaturation and would improve digestibility of nutrients, feed efficiency and may have a role in egg formation. The hatching process is considered to be a time of oxidative stress. Therefore, improved antioxidant defences during embryonic development could potentially increase the hatchability of eggs.

Selected slaughter measurements:

As given in Table 6, the results showed that there were no significant differences in relative weight of carcass yield, liver and heart of turkey hens among different dietary treatments. The results showed also that turkey hens fed inorganic, organic and nano form of Zn and organic and nano form of Se caused a significant increase in relative weights of ovary and oviduct of turkey hens. The results displayed also that turkey hens fed organic and nano forms of Zn and Se-supplemented diets exhibited a significant increase in relative weight of spleen and oviduct length compared with their control group (Table 6).

The present results agree with those of Hanafy *et al.* (2009), who indicated that relative weights of ovary and spleen were significantly increased by addition of organic Se to diets of Bandarah laying hens from 40 to 60 weeks of age. However, Rama Rao *et al.* (2013) found that relative weight of liver and spleen were not affected by addition of organic Se to broiler diets. Also, Osman and Ragab (2007) found no significant effect of dietary supplementation of zinc-methionine on carcass traits (liver, gizzard, spleen and heart) of Hubbard broiler chicks. Namra *et al.* (2008) found that supplementation of different sources of dietary Zn had no significant effect on most of the carcass traits examined Japanese quail chicks.

Table 6. Effects of dietary inorganic, organic and nano forms of zinc and selenium on relative weights of carcass yield and vital internal organs of 44-week-old turkey hens

Dietary Treatments	Carcass yield %	Liver %	Spleen %	Heart %	Ovary %	Oviduct %	Oviduct length (cm)
Control (Basal diet)	72.59	2.82	0.113^{d}	0.40	2.40^{b}	2.34^{c}	62.66^{bc}
Basal diet + Inorganic Zn	72.98	2.56	0.115^{bcd}	0.39	2.63^{a}	2.65^{b}	64.33^{b}
Basal diet + Organic Zn	73.46	2.39	0.122^{ab}	0.39	2.73^{a}	3.03^{a}	74.66^{a}
Basal diet + Nano Zn	73.38	2.58	0.120^{abc}	0.38	2.65^{a}	2.96^{a}	72.00^{a}
Basal diet + Inorganic Se	72.73	2.73	0.114^{cd}	0.39	2.39^{b}	2.21^{c}	60.66^{c}
Basal diet + Organic Se	73.19	2.69	0.125^{a}	0.39	2.70^{a}	2.91^{a}	72.66^{a}
Basal diet + Nano Se	73.18	2.55	0.123^{a}	0.39	2.71^{a}	2.84^{ab}	72.33^{a}
SEM	0.45	0.17	0.002	0.01	0.07	0.07	0.89
Significance	NS	NS	*	NS	*	*	*

a-d: Means in the same column bearing different superscripts differ significantly ($P \leq 0.05$).
***: Significant at $P \leq 0.05$ NS: Not significant SEM: Standard error of the means**

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تأثير بعض مصادر مضادات الاكسدة على الآداء الانتاجى والتناسلى لإناث الرومى

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الهدف من هذه الدراسة هو تقييم تأثير إستخدام بعض مصادر مضادات الاكسدة على الاداء الانتاجى والتناسلى للرومى . أجريت هذه الدراسة فى محطة بحوث الانتاج الحيوانى بمحله موسى ، كفرالشيخ التابعة لمعهد بحوث الانتاج الحيوانى ، مركز البحوث الزراعية ، وزارة الزراعة ، مصر . إستخدم فى هذه الدراسة عدد ٨٤ طائر (دجاجة) من سلالة الرومى البرونز ، عمر٣٢ إسبوع . وزعت الطيور بطريقة عشوائية الى ٧ معاملات تجريبية ، أحتوت كل معاملة على ٣ مكررات متساوية . تم تسكين الطيور بطريقة فردية فى بطاريات. إستغرقت التجربة ١٢ أسبوع من عمر٣٢- ٤٤ أسبوع . غذيت الطيور بعليقة أساسية (كنترول) مكونة من أذرة صفراء وكسب فول صويا مضاف اليها نوعين من مضادات الاكسدة هما الزنك والسيلينيوم فى ثلاثة صور (العضوية ؛ الغيرعضوية و النانو). وكانت المعاملات الغذائية كالتالى: المعاملة الاولى عبارة عن عليقة كنترول بدون إضافات للزنك أوالسلينيوم ؛ الثانية زنك غير عضوى (١٠٠ مجم/ كجم عليقة) ؛ الثالثة زنك عضوى (١٠٠ مجم/ كجم عليقة) ؛ الرابعة نانو زنك (٤٠ مجم / كجم عليقة) ؛الخامسة سلينيوم غيرعضوى (٠.٣ مجم/ كجم عليقة) ؛ السادسة سلينيوم عضوى (٠.٣ مجم/ كجم عليقة) ؛ السابعة نانو سلينيوم (٠.١٥ مجم/ كجم عليقة) وكان أهم النتائج المتحصل عليها كالتالى :- أعطت المعاملات المضاف اليها الزنك والسلينيوم فى الثلاث صور (الغير عضوية والعضوية والنانو) زيادة معنوية ($P\leq0.05$) فى تركيزات كل من البروتينات الكلية و الهيموجلوبين فى بلازما الدم ، نسبة الخصوبة و الفقس للبيض وتحسن معنوى فى معدل التحويل الغذائى لاناث الرومى مقارنة بالاناث فى مجموعة الكنترول.- أعطت المعاملات المضاف اليها كل من الزنك والسلينيوم فى الثلاث صور (الغير عضوية والعضوية والنانو) إنخفاض معنوي ($P\leq0.05$) فى معدل استهلاك العلف، تركيز مستوى الليبيدات الكلية والكوليسترول فى بلازما الدم لاناث الرومى مقارنة بالاناث فى مجموعة الكنترول.- أعطت المعاملات المضاف اليها الزنك فى الصور العضوية والنانو والسلينيوم فى الثلاث صور (الغير عضوية والعضوية والنانو) تحسن معنوي ($P\leq0.05$) فى معدل إنتاج البيض اليومى ووزن البيض لاناث الرومى مقارنة بالاناث فى مجموعة الكنترول.- أعطت المعاملات المضاف اليها الزنك والسلينيوم فى الصور العضوية والنانو تحسن معنوى فى مستوى الاستجابة المناعية ضد مرض النيوكاسل وبالتالى زيادة مناعة الجسم ، زيادة معنوية فى الوزن النسبى للطحال وطول قناة البيض لاناث الرومى مقارنة بالاناث فى مجموعة الكنترول.- أعطت المعاملات المضاف اليها السلينيوم فى الصور العضوية والنانو تحسن معنوى فى جودة البياض المقاس من خلال وحدات هاو، سمك قشرة البيض لاناث الرومى مقارنة بالاناث فى مجموعة الكنترول. - لم تكن هناك فروق معنوية بين مختلف المعاملات التجربية فى الوزن النسبى للمكونات الداخلية للبيضة ، دليل قشرة البيضة ، دليل الصفار ، لون القشرة ، الوزن النسبى للذبيحة والقلب والكبد.يستنتج من النتائج المتحصل عليها إن إضافة الزنك والسلينيوم فى الصور العضوية والنانو إلى العليقة يمكن أن يحدث تأثيرات إيجابية على الاداء الآنتاجى والتناسلى لإناث الرومى.