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Comparative Effects of Different Dietary Selenium Sources on Productive Performance, Antioxidative Properties And Immunity in Local Laying Hens Exposed to High Ambient Temperature

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ABSTRACT

The present study was carried out to compare the efficiency of different selenium (Se) sources [sodium selenite (SS), Se enriched yeast (Sel-Plex) and Nano-Se] on productive performance, lipid peroxidation, antioxidative status and immunity function of local chickens strain exposed to summer condition (high ambient temperature). One hundred and twenty birds, were randomly divided into 4 treatments: (1) control (basal diet without any supplementation of selenium), (2) SS (basal diet + 0.3 mg Se as SS / kg diet); (3) basal diet + 0.3 mg Nano-Se/ kg diet (Nano-Se); and (4) basal diet + 0.3 mg organic Se/ kg diet (Sel-Plex). The experimental diets are given to birds from 30 to 42 wk of age. Under hyperthermia, dietary 0.3 ppm Sel-Plex or Nano-Se improved feed conversion (g feed/g egg mass), egg production percentage and egg mass (g/d) ($P < 0.05$). Supplementing diets with Nano-Se increased total protein and globulin content compared with control diet. While, addition different sources of selenium significantly decreased malondialdehyde (MDA) content compared with treat (1) diet. Interestingly, under rise ambient temperature, the inclusion of 0.3 ppm Sel-Plex or Nano-Se in the chikens diet significantly enhanced the antioxidant enzyme (GSH-Px) activity being greater than 2-folds of the controls. Chickens fed diet supplemented with Sel-Plex or Nano-Se showed about 1.5-folds increase in seminal plasma (GSH-Px) compared with control diet. Dietary 0.3 ppm Sel-Plex or Nano-Se had a positive significant effect on Phagocytic activity (PA) and Phagocytic index (PI) on high ambient temperature. In conclusion, dietary 0.3 ppm organic Se or Nano-Se enhanced productive performance, antioxidative properties and immunity in chickens reared under heat stress conditions.

Keywords: hens; cockerel; Nano-Se; organic Se; antioxidative status; immunity.

INTRODUCTION

Heat stress is consider an important stressors negatively affecting in poultry industry leading to waste a lot of many each year. It has been noted that modern breeds are most sensitive to heat stress (HS) than first genotypes. Heat stress (high ambient temperature) decreases feed consumption , body weight gain, feed conversion, immune response, egg production and fertility of chickens (Siegel, 1995; Melesse *et al.,* 2011). Stress sensibility of poultry is a main problem faces present intensive industry of poultry. It is well known that high temperature (35±3^{o}C) joined with high humidity has a harmful effect on the chickens industry by decreasing flock fertility and production. McDaniel *et al.,* (1996) showed that fertility of broiler breeder males was highly reduced during high ambient temperatures, resulting in large annual financial losses. Several studies confirmed that heat stress significantly decreased egg production, egg mass and the index of spermatozoa quality, motility, viability and fertility; and the dead spermatozoa percentage was increased as a effect of HS (Ebeid, 2012; Kalamah, 2001). Similarly, the outcome of metabolic rates under stressors conditions, mortified levels of free radicals (ROS, reactive oxygen species) formed a formative leading to a lot of oxidative stress. Moreover, chicken and vertebrate spermatozoa show high levels of metabolic material such as (PUFA), which led them a mostly susceptible to oxidation by reactive oxygen species , specifically under stressors conditions in mammals (Aitken *et al.,* 1989) also, domestic poultry (Surai *et al.,* 1998a & b; Eid *et al.,* 2006). Production of free radicals in semen are involved in changes in spermatozoa membrane fluidity, DNA and RNA fragmentation, protein damage and, consequently, loss of spermatozoa motility and fertility (Lopes *et al.,* 1998; Sanocka and Kurpisz, 2004). Thus, ROS production and the antioxidant enzymes must have a balance between them to established and preserve spermatozoa viability and fertility (Surai, 2002; Ebeid , 2012).

El-Deep *et al*., (2016) found with broiler that, ROS was prevented by natural antioxidants , which have a vital role in cells by decresing free radicals and avoiding lipid peroxidation . Surai, 2002 demonstrated that, antioxidant system has many antioxidant enzymes, like (GSH-Px), (SOD) and (CAT). Antioxidative properties was activating by the role of Se, which participation in the active site of the enzyme GSH-Px in blood, liver, seminal yolk extract and edible tissues (El-Deep *et al*., 2016 ; Ebeid, 2012 ; Yoon *et al.,* 2007) antioxidant enzyme has a serves helps to control levels of lipids peroxides and ROS (El-Deep *et al*., 2016 ; Arthur, 2000) leading to enhancing the immunity in many species (Rayman, 2004; Ebeid *et al.,* 2013). Several previous studies observed that addition Se was influence on growth performance, egg production, semen and quality of meat and antioxidant levels in semen, blood and egg yolk of chickens (El-Deep *et al*., 2016 ; Ebeid , 2012 ; Wang and Xu, 2008). The Se bioavailability is associated with its forms. Inorganic informs for example sodium selenite (Na_2SeO_3; SS) , also organic forms like Selenium enriched yeast (SY) and selenomethionine (Sel-Plex) are widely used in poultry field (Surai, 2002). Previous studies suggested that inorganic Se is more toxic than organic Se (Doucha *et al.,* 2009). Nanotechnology holds wide promises for medication and nutrition recently in poultry field. Newly studies notice that nano-elemental like (Nano-Se) has a high bioavailability, low toxicity , best catalytic efficiency, and high ability to adsorbant compared with selenite and organic Se in chickens (El-Deep *et al*., 2016 ; Wang *et al.,* 2009), rats (Jia *et al.,* 2005 and Wang *et al.,* 2007), sheep and goat (Shi *et al.,* 2011a,b). However, according to our knowledge, data available on antioxidant status on semen, eggs and blood constants of hens exposed to heat-stress using Nano-Se is still limited. For that, the objective of these experimental was compared the efficiency of dissimilar Se sources (SS, SY and Nano-Se) on growth, egg production, lipids peroxidation, ant oxidative status

and immunity function of laying hens exposed to hot climate conditions.

MATERIALS AND METHODS

Birds, the conditions of housing and environmental: A total of 30, 42-week-old hens and cockerels Inshas (Egyptian local strain) were chosen from a flock of about 120 birds ((4 treatment × 3 rep. × 10 birds (8 hens + 2 cockerels)) to obtain randomly selected from the flock of farm with similar production and weight , also spermatozoa count (2.14± 0.15 × 10^9/ml). Individual battery cages have birds , which housed in open system farm under a sixteen hour light: eight hour dark according to lighting schedule. The experiment was carried out under Egyptian conditions (from June to August) it means hot climate . In period of theses study, the daily temperature mean was ranged between 33 to 36ºC and relative humidity average between 60 to 70% in inside the farm. The experiment was perfect according to institutional guidelines for roles of animal use.

The experimental design: All birds were provided *ad libitum* with water and the experimental feeds were subedited to be iso nitrogenous and iso caloric to meet the nutrients need requirements (Table 1) according to Agriculture Ministry Decree (1996). Birds were divided to the 4 experimental treatments (30 birds in each treatment): (1) control (basal diet); (2) SS (basal diet + 0.3 mg Se as (Na_2SeO_3) sodium selenite (SS) / kg diet),which produced by Sigma- Co., Japan; (3) Nano-Se (basal diet + 0.3 mg Nono-Se/ kg diet) Selenium nanoparticles 100 to 500 nm were provided by Eszenyi *et al.*, (2011) ; and (4) Sel-Plex (basal diet + 0.3 mg organic Se/ kg diet) source of selenomethionine was obtained from Alltech Inc, Tokyo . , the basal diet don't have any Se addition , also the ingredients of the feed have a low Se , which are used to performed basal diet (calculated, 0.072 ppm).

Measurements: Daily egg weight and number were recorded for each hen and feed intake was recorded every week. Egg production were calculated for monthly intervals during the production period as number of egg /hen/ all period for replicates then, it calculated the meam of each treatment in whole period. Egg mass equaled egg number × mean egg weight. Feed conversion (FC) = gram feed/ gram egg mass). At the last week of experimental quality of eggs were determined, in which 6 eggs were randomly taken from each treatment (2 eggs from each replicate). Each egg was broken to calculate egg shell, yolk and albumen percentages . Shell thickness (nn) of egg was measured by a micrometer. Yolk index and egg shape index records were calculated according to Sauter *et al*. (1951); according to Haugh (1937); Kotaiah and Mohapatra, (1974) and Eisen *et al.* (1962) haugh unit was measured. The samples of blood were collected for every hen in tubes containing heparin then divided in two parts ; first part of blood sample was centrifuged flowed that kept plasma samples at -20°C for chemical analyses of total lipids, total protein, alb, globulin and AST were estimated in plasma by colorimetric methods using marketable kits, following the steps as labeled by manufactures.

Table 1. Composition and calculated chemical analysis of the basal diet.

Ingredients		%
Yellow corn		63.23
Soybean meal (44%)		23.10
Limestone		8.36
Corn gluten meal (62%)		3.15
Mono – Ca – P		1.43
Salt (NaCl)		0.37
Vit. & Min. Mix.[1]		0.30
DL-Met. 98%		0.06
Total		***100***
Calculated chemical analysis[2] :		
CP	%	17.019
ME	Kcal /kg	2751
CF	%	3.203
EE	%	2.817
Ca	%	3.404
Available P	%	0.430
Lys.	%	0.863
Met.	%	0.387
Met. + Cys.	%	0.676
Na	%	0.162
Se	ppm	0.072

[1]**Supplied per kg of the diet: Vit. A 10000 IU; Vit. D_3 2000 IU; Vit. E 10 mg; Vit.K_3 1 mg; Vit.B_1 1 mg; Vit. B_2 5 mg; Vit. B_6 1.5 mg; Vit. B_{12} 0.01 mg; Niacin 30 mg; Pantothenic acid 10 mg; Folic acid 1mg; Biotin 0.05 mg; Choline 250 mg; Mn, 60 mg; Cu, 4 mg; Zn, 50 mg; Fe, 30 mg; I, 0.3 mg; Co, 0.1 mg.** [2] **Analysis of ingredients calculated according to NRC (1994)**

The complete blood samples kept at 4°C then directly determined activity of glutathioneperoxidase (GSH-Px) and phagocytosis of polymorphonuclear. Phagocytosis of polymorphonuclear cells using Candida albicans was performed. In plastic tube, the following aliquots were mixed: 100 µl fetal calf serum, 100 µl heat Killed Candida albicans (5×10^6/ml) and 100 µl blood. The tubes was mixed and incubated at 37° C for 30 minutes, after which it was centrifuged for 5 min at 1000 rpm. The supernatants were discarded leaving a droplet in to which the sediment was re-suspended. Smears were prepared from the deposit, dried in the air, fixed with methyl alcohol and stained with Giemsa stain. One hundred heterophils were examined and the number of heterophils ingesting Candida was counted and expressed as percentage.

Phagocytic activity (PA) = Percentage of phagocytic cells containing yeast cells.

$$\text{Phagocytic index (PI)} = \frac{\text{Number of yeast cells phagocytized}}{\text{Number of phagocytic cells}}$$

According to Lake and Stewart, 1978 samples of semen were collected weekly. At the last 4th weeks of period study, The cockerels semen samples of were analysed. Semen samples were centrifuged at 2500g for 20 min. At -20 C supernatant was stored to analysis. The volume (ml) of the ejaculate was estimated by using a haemocytometer (Lake and Stewart, 1978). Spermatozoa motility percentages were analyzed according to (Ebeid, 2009; Słowinska *et al.,* 2011). Spermatozoa motility according to Melrose and Laing, (1970). Viability was evaluated as the percentage of spermatozoa (Lake and Stewart, 1978). Calcium (Ca^{++}) in plasma of semen was estimated by the method of

Gindler and King (1972) .Lipids peroxidation in the plasma of blood and semen were estimated the oxidative stress index thiobarbituric acid reactive substance (TBARS) in particular malondialdehyde (MDA)as described by Richard *et al.,* (1992). The GSH-Px in the plasma of semen were evaluated according to Paglia and Valentine, (1967) and Koracevic *et al.,* (2001), by use kits produced by Bio-diagnostic, Egypt.

Statistical analysis: Data were subjected to ANOVA using the general linear models procedure of SPSS®. Significant variance among treatment averages were compared by Duncan's new Multiple-Range Test (Duncan, 1955) at $P<0.05$.

RESULTS AND DISCUSSION

Productive performance: Climatic stress, especially high ambient temperature, effects on productive performance. The effects of Se source on live body weight change (g), feed intake (g/d) and feed conversion ratio (g feed/g egg mass) are shown in Table 2. The live body weight change (g), feed intake (g/d) were insignificantly affected by addition of all types of Se source. The effect of Se source in productive performance at the all experimental time are found in Table 2. Nano-Se Supplementation in hens diets significantly enhanced egg production percentage (%) , egg mass and feed conversion (FC) when compared with T2 (SS) or control groups, while Nano-Se and organic selenium (Sel-Plex) were insignificantly in other productive performance traits and both of them were decreased the side effects of hot climate on productive performance. The enhancement due to Se supplementation in diets, Se is one of an important extra issue for the key enzyme of 5-deiodinase. The importance of iodothyronine deiodinase enzymes , which was switch the pro-hormone (inactive form) thyroxine (T4) to new active form triiodothyronine (T3). Triiodothyronine is a master hormone which controls in growth by controlling energy of body's and muscles anabolism (Arthur *et al*., 1999; Preter, 2000). Also, GSH-Px has in it's structure selenium. The increase ROS has led to reductions in production (Underwood and Suttle, 1999). Hens receiving 0.3 ppm of Nano-Se significantly had the best value in both egg production percentage and egg mass by 19.60%, 8.56%, respectively while improved feed conversion ratio by 4.05% compared to SS diet. Egg weight, feed intake and body weight of birds were not affect by Se source in all groups (Table 2).

Table 2. Effect of different selenium sources on productive performance parameters of laying hens from 30 to 42 weeks of age.

Treatments	Live body weight change (g)	Feed intake (g/d)	Feed conversion (g feed/g egg mass)	Egg production (%)	Egg weight (g)	Egg mass (g/d)
Control	266±6.01	117.55±1.50	3.55^{a}±0.06	60.05^{c}±0.59	47.50±0.12	33.66^{c}±0.33
SS	267±6.10	118.75±0.95	3.45^{a}±0.07	61.21^{c}±0.86	47.85±0.11	33.99^{c}±0.35
Nano-Se	280±7.05	120.35±1.09	3.31^{b}±0.04	73.21^{a}±0.98	48.01±0.15	36.90^{a}±0.35
Sel-Plex	274±5.12	119.40±0.98	3.43^{b}±0.04	70.08^{b}±1.02	47.99±0.20	35.89^{ab}±0.34
Significant	NS	NS	**	**	NS	**

a-c: Means in the same column followed by different letters are significantly different. NS= not significant and ** (P≤0.01).
SS= sodium selenite.

The present results agree with the report of Nadia Radwan *et al*. (2015) who indicated that increasing the level of Se supplementation does not affect feed intake of hens fed various concentrations (0 to 0.25 ppm) of Nano-Se. On the other hand, addition Nano-Se by 0.30 ppm significantly enhanced FC (Zhou and Wang, 2011; Cai *et al*., 2012). The percentage of egg production was improved by addition organic selenium in layer feed (Gjorgovska *et al*., 2012; Nadia Radwan *et al*., (2015)). Also, Attia *et al*. (2010) observed that egg weight and egg mass significantly increased and feed conversion ratio improved by selenium supplementation when compared with hens fed the control diet. Nevertheless, Se source has not affected in the egg production percentage (Leeson *et al*., 2008; Reis *et al*., 2009; Attia *et al*., 2010).

Egg quality: The both external and internal egg quality were not affected by Se source, except of Yolk color of egg , shell thickness and percentage which were significantly affected by Se source of hens exposed to high ambient temperature (Table 3). Addition Selenium by 0.30 ppm (Nano-Se or Sel-Plex) significantly enhanced Yolk color, thickness and percentage of shell. The highest value of Yolk color, thickness and percentage of shell were recorded in layer hens fed diets with 0.30 ppm of Nano-Se flowed by hens fed Sel-Plex compared with control. While, Haugh unit, egg yolk and egg shape index were insignificantly increased by Se source. The highest value was recorded for 0.30 ppm of Nano-Se flowed by Sel-Plex group as compared with control diet. The addition of Nano-Se and Sel-Plex to control diet ameliorated the adverse effect of high ambient temperature. Nadia Radwan *et al*. (2015) and Attia *et al*. (2010) were found that addition inorganic (SS) or organic selenium in birds diets, also had no significantly affected on all traits of both egg quality, showing that selenium content of the basal diet was enough to support egg production of good quality. On the other hand, Gjorgovska *et al*. (2012) indicated that Se sources not affected in ratio of albumin , yolk and shell egg. As well as, Paton *et al*. (2000) reported that addition 0.3 ppm selenium (inorganic or organic) had insignificant on Haugh unit when compared with control group. Conversely, Gajcevic *et al*. (2009) indicated that hens fed a diet with organic selenium had higher Haugh unit record than eggs in control group.

Table 3. Effect of different selenium sources on some egg quality (Means ± S.E).

Treatment	Yolk color	Shell thickness (mm)	Haugh unit	Egg yolk index%	Egg shell %	Egg yolk %	Egg albumin %	Egg shape index
Control	5.5^{b}±0.21	0.370^{b}±0.01	78.20±0.81	44.55±0.25	11.41^{c}±0.28	31.00±0.31	55.90±0.60	75.36±0.86
SS	5.3^{b}±0.30	0.361^{c}±0.01	78.14±1.15	44.45±0.30	11.36^{c}±0.35	31.00±0.25	56.30±0.44	75.21±1.00
Nano-Se	5.8^{a}±0.08	0.382^{a}±0.01	78.70±0.86	45.00±0.46	11.75^{b}±0.25	31.00±0.22	55.80±0.48	75.55±0.76
Sel-Plex	5.4^{b}±0.36	0.380^{a}±0.01	78.60±0.16	44.80±0.26	11.85^{b}±0.38	31.20±0.18	56.70±0.70	75.78±0.76
Significant	*	*	NS	NS	*	NS	NS	NS

a-c: Means in the same column followed by different letters are significantly different. NS= not significant and *(P≤0.05). SS= sodium selenite.

Biochemical parameters: The effect of different dietary Se sources in the liver functions (Table 4), Nano-Se and Sel-Plex supplementation caused a significant reduction in plasma total lipid. The greatest reduction was observed when addition 0.3 ppm of Nano-Se to diet was insignificant effect as compared to 0.30 ppm Sel-Plex. The reason of this reduction may be due to vital role of Se in dominant effects of thyroid (T3) hormone on fat metabolism (Masukawa *et al*., 1983). Hypercholesterolemia have been found to be related with Se deficiency was related to improve 3-hydroxy-3-methylglutary CoA reductase the microsomes of liver activity (Nassier *et al*., 1997). The GSH-Px is activity by selenium which plays a vital role as an antioxidant enzymes may be affective on lowering cholesterol.

Table 4. Effect of different selenium sources on some constituents of blood.

Treatment	Total lipids (g/dl)	Total protein (g/dl)	Albumin (g/d)	Globulin (g/d)	A/g Ratio	Serum AST (IU/L)
Control	17.0^{a}±1.17	5.71^{c}±0.27	2.40±0.03	3.31^{c}±0.09	0.72^{a}±0.01	120.9^{a}±0.87
SS	16.1^{a}±0.29	6.28^{b}±0.16	2.69±0.04	3.59^{b}±0.28	0.74^{a}±0.11	129.5^{a}±0.34
Nano-Se	13.22^{b}±0.33	7.19^{a}±0.21	2.2±0.07	4.99^{a}±0.33	0.44^{c}±0.10	110.3^{b}±0.91
Sel-Plex	14.63^{b}±0.54	5.99^{bc}±0.11	2.38±0.09	3.61^{b}±0.32	0.65^{b}±0.10	113.4^{b}±0.49
Significant	**	**	NS	**	**	*

a-c: Means in the same column followed by different letters are significantly different. NS= not significant, *(P≤0.05) and ** (P≤0.01). SS= sodium selenite.

The effects of dietary Nano-Se on blood constituents are presented in Table 4. Total protein was increased by dietary all types of Se source, Nano-Se recorded the highest total protein value followed by Sel-Plex. The same trend was obtained with A/g ratio, the highest value of globulin was obtained by using 0.3ppm of Nano-Se while, there were insignificant differences between albumin values recorded by different groups. It seem that Nano-Se and Sel-Plex has also been proven beneficial in alleviating the side effects of hot climate.

The Se source had significantly affect in AST enzyme. The birds fed either Nano-Se or Sel-Plex recorded lower values of AST enzyme compared with control group. On the other hand, the birds fed SS recorded the highest value as compared with all groups. The results are disagreement with that reported by Yang *et al*. (2012) who observed that selenium (organic or inorganic) didn't affected in liver enzymes. Moreover, Mohapatra *et al*. (2014) found also liver enzymes were insignificantly between sodium selenite (0.3 ppm) and Nano-Se (0.15, 0.30 and 0.60 ppm) treated.

Results are presented in the Table 5. Under hot temperature, addition Nano-Se, Sel-Plex and SS had a positive (P≤0.05) effect on spermatozoa count and motility. The spermatozoa counts of experimental groups with supplementary Nano-Se and Sel-Plex were similar and substantially higher than the control group followed by SS group. Semen samples from basal chickens contained a significant increased percentage of dead spermatozoa than any of the other groups. Inclusion of Se source reduced the percentage of dead spermatozoa under hot climate conditions and had no effect on ejaculate volume compared with control (Table 5). Insignificant affect of dietary treatment on seminal plasma Ca^{++} concentration was detected.

Table 5. Mean (± SE) volume of semen, concentration, motility and percentage of dead spermatazoa in cockerels fed on diets with or without dietary supplementary SS, Nano-Se and Sel-Plex.

Treatments	Semen Volume (ml)	Spermatazoa Total ($\times 10^{9}$/ml)	Motility (score)	Dead (%)	Seminal plasma Ca ++(mg/100ml)
Control	0.31 ± 0.01	1.87^{c} ± 0.17	4.10^{b} ± 0.18	16.17^{a} ± 0.68	9.65 ± 0.4
SS	0.32 ± 0.01	2.84^{b} ± 0.18	4.35^{b} ± 0.17	8.63^{b} ± 0.81	9.68 ± 0.4
Nano-Se	0.31 ± 0.01	2.95^{a} ± 0.17	4.98^{a} ± 0.19	6.01^{b} ± 0.74	9.69 ± 0.3
Sel-Plex	0.32 ± 0.01	2.91^{a} ± 0.17	4.97^{a} ± 0.18	6.82^{b} ± 0.79	9.72 ± 0.3
Significant	NS	**	**	*	NS

a-c: Means in the same column followed by different letters are significantly different. NS= not significant, *(P≤0.05) and ** (P≤0.01). SS= sodium selenite.

Dietary supplementation ameliorated the sids effects of hot climate on lipids peroxidation and also , antioxidative properties in birds semen (Figure). Dietary supplementation with Se source decreased TBARS values (Figure, A1,2), of antioxidants reduced the TBARS in seminal and blood plasma, respectively . (Figure, B1, 2). Supplementary dietary Se source showed a positive effect on antioxidant properties as measured by TBARS and GSH-Px activity in seminal and blood plasma, respectively. Under hot climate

conditions, the inclusion of 0.3 ppm Nano- Se or 0.3 ppm Sel-Plex, markedly enhanced the plasma of semen GSH-Px activity by more than two-fold as compared with controls in seminal and blood plasma, respectively (Figure, B1,2).

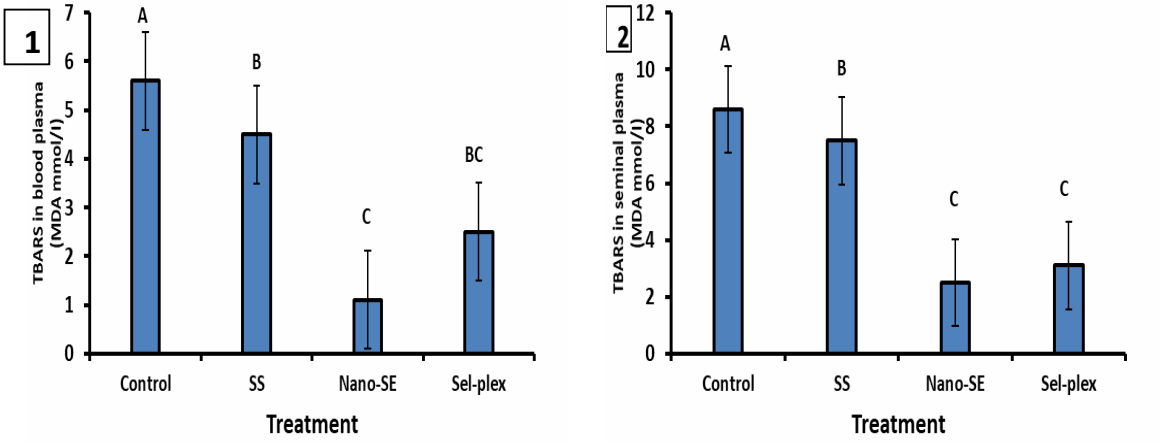

Fig. A. Effect of different sources of dietary Se on blood MDA (1) and seminal MDA (2) contents in chickens exposed to high ambient temperatures (means ± SE).

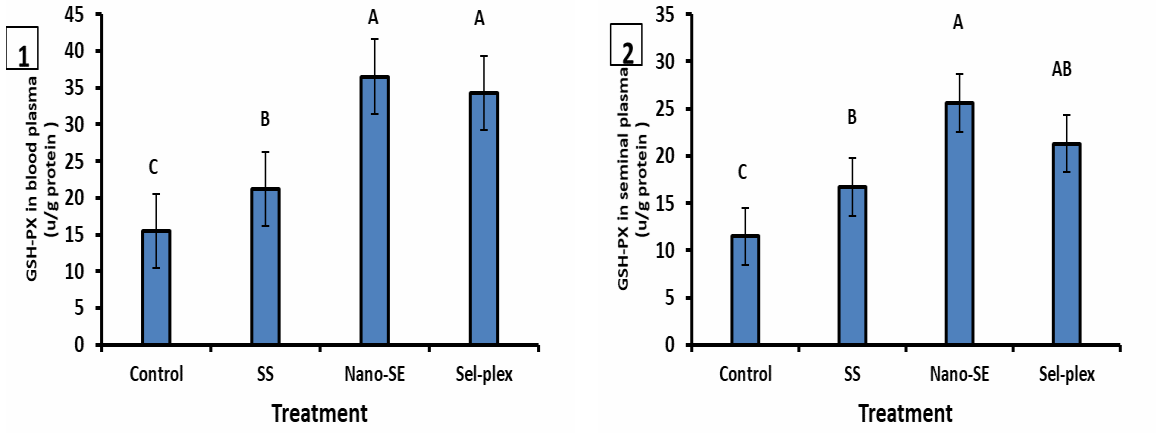

Fig. B. Effect of different sources of dietary Se on blood GSH-PX (1) and seminal GSH-PX (2) contents in chickens exposed to high ambient temperatures (means ± SE).

Heat stress, (summer Egypt) high ambient temperature, affects semen quality characteristics in chickens and subsequently the flock fertility (McDaniel *et al.,* 1995 & 1996; Kalamah, 2001). Moreover, it was documented that stress could improve lipids peroxidation in both the plasma of semen and the spermatozoa membranes , the results in reduction of the spermatozoa count (Surai *et al.,* 1998a & b; Eid *et al.,* 2006). Results of during study show the positive effects of Nano-Se and organic Se supplementation on semen quality characteristic, inclusive spermatozoa count, motility and the percentage of dead spermatozoa under high ambient temperature (Table5). Recently results are agreement with other results which indicated that organic Se (Ebeid, 2009) led to enhance semen quality in cockerels by increasing concentrations of spermatozoa and cell viability under stress conditions. Several studies have also reported improvements in spermatozoa quantity and quality with supplemental Se in poultry (Hanafy *et al.,* 2009; Lin *et al.,* 2005) and in mammals (Yousef, 2010; Yue *et al.,* 2010). Se supplementation plays a vital role in improving the semen quality in cockerels, especially under stressful conditions. Mammalian (Kelso *et al.,* 1997) and avian (Surai *et al.,* 1997 & 1998b) spermatozoa are characterized by highly increase concentrations of PUFA in the phospholipids that are oversensitive to peroxidative damages, and are considered to be a cause of male sub fertility (Aitken, 1994). Consequently, an effective antioxidant system such as (GPX) is required to keep spermatozoa membranes against ROS and peroxidative damages (Eid *et al.,* 2006; Ebeid, 2009; Ebeid, 2012).

The functionsof selenium in the cytoplasm is destroy peroxides (Surai, 2002) suggesting synergism between both in the antioxidative system. In addition, Se is necessary for the development of spermatozoa and spermatozoa production, via their effects on increasing testosterone concentrations (Kaur and Bansal, 2004). Collectively, these studies clearly indicate that addition natural antioxidant is essential for normal spermatozoa development, integrity and protection, and are likely to be involved in improving semen quality traits under HS conditions. Males supplemented with different Se

source were characterized by better resistance to hot climate conditions and this was reflected by best motility and better concentration (Table5). Furthermore, on the biochemical rate, the defensive effect of Nano-Se and Sel-Plex were related to a decrease in lipids peroxidation in blood and semen (Figure A1,2). It is well know that lipids are one of the vital component of semen, participate to cellmembrane structure, metabolism and the capability to capacitate and fertility of oocytes (Surai, 2002). The highly concentrations of PUFA within the lipids fraction require the presence of an effective antioxidant system enzymes to protect against per oxidative damages, and associated spermatozoa dysfunction and male infertility (Aitken, 1994; Surai *et al.,* 1997). Therefore, it could be expected that decreasing the concentration of TBARS in seminal plasma would be associated with enhanced spermatozoa motility and concentration under stressors conditions. In Figure A, the TBARS as an index of lipids peroxidation, the 0.3 mg of Nano-Se and organic Se provided the greatest protective effect against lipid peroxidation in plasma of semen.

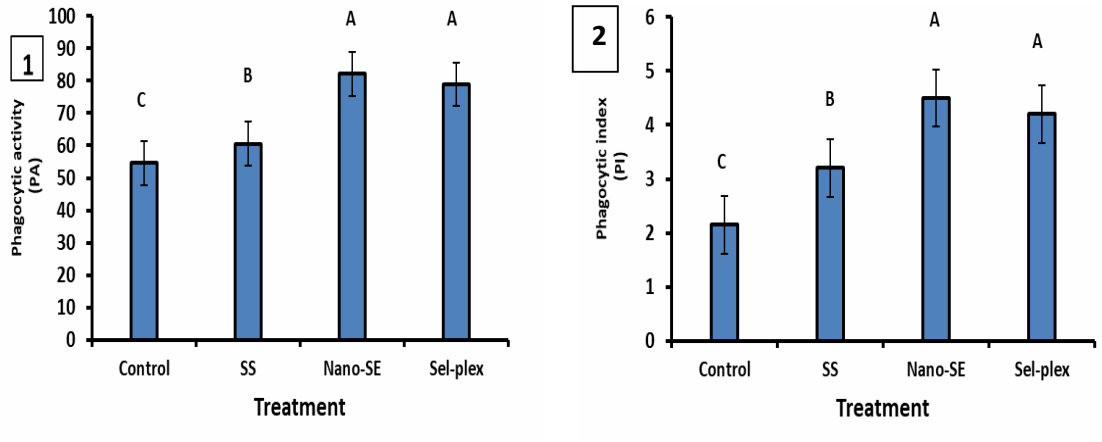

Fig. C. Effect of different sources of dietary Se on Phagocytic activity and Phagocytic index (2) contents in chickens exposed to high ambient temperatures (means ± SE).

Lipids peroxides was detoxification by GSH-Px enzyme involved selenium, which plays a great role in that which can appear in birds spermatozoa (Froman and Thurston, 1981). In birds and spermatozoa, nearly 60% and 40% of the Se dependent form of active GSH-Px in semen spermatozoa was observed in plasma of semen (Surai *et al.,* 1998b). Additionally, it was demonstrated that the inclusion of Nano-Se and Sel-Plex (0.3 mg/kg) in the male diet significantly increased Se-dependent GSH-Px activity, which is correlating with better protection against lipid peroxidation in semen under all climate conditions (Surai *et al.,* 1998a) specially ,under heat stress conditions (Ebeid, 2009).

The effects of dietary different sources of Se on Phagocytic activity (PA) and Phagocytic index (PI) the immune response are shown in Figure C1, 2 . Dietary 0.3 ppm Sel-Plex or Nano-Se had positive significant effect on PA and PI of chickens ($P < 0.05$) under high ambient temperature. Under hot climate conditions, the PA and PI were greater in dietary 0.3 ppm Sel-Plex or Nano-Se compared as control and SS ($P < 0.05$).

As presented in Fig. C1,2, dietary Se supplementation had a immunostimulating effect (PA and PI) of birds subjected to heat stress and our results support with regared to the antioxidative parameters were discussed above. The results from the current trial are in agreement with several studies (Niu *et al.,* 2009; Liao *et al.,* 2012; Habibian *et al.,* 2014) which indicated that addition organic Se (sel plex) improved titer of antibody when chickens were exposed to stress conditions. Virtually, there is an evidence that Se addition led to give more health advantages on the immunity response and reduced inflammation (Rayman, 2004). Rooke *et al. (*2004) indicated that Se is involved in many of immune functions, at all levels, like reduction of glucocorticoids (indicator for immunity disorder), minimizing levels and duration of inflammatory infections, arrangement the function of T lymphocytes cell and T killer cells and activation of IL-2. Recently, Zamani Moghaddam *et al.* (2017) observed an improvement in total anti-sheep red blood cell (SRBC) in birds fed on a diet including with Nano-Se at 0.3 mg/kg compared to those fed on a diet supplemented with SS

CONCLUSION

Based on the data presented on this study and taking into account the antioxidant interactions, it could be assumed that dietary 0.3 ppm organic Se or Nano-Se might be involved in enhancing productive performance, egg quality, semen quality, antioxidative properties and immunity in hens reared under hot climate conditions.

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التأثير المقارن لاستخدام مصادر مختلفة من السيلينيوم فى العليقة على الأداء الإنتاجى و خصائص مضادات الاكسدة و المناعة للدجاج البياض المحلى المربى تحت ظروف الجو الحار.

محمود حمزه الديب ، محمد شعبان حسن ، محمد حسنى عصر ، خليل عبد الجليل عطية و محمد احمد محمد سيد

معهد بحوث الإنتاج الحيوانى - مركز البحوث الزراعية – الدقى – الجيزة - مصر

اجريت هذه الدراسة بهدف دراسة تأثير استخدام مصادر مختلفة من السيلينيوم (صوديوم سيلينيت – خميرة سيلينيوم (سيلبليكس منتج تجارى مصدر للسيلينيوم العضوى) - نانو سيلينيوم) على الأداء الإنتاجى (وزن الجسم المكتسب والعلف المستهلك والكفاءة التحويلية) وحالة مضادات الاكسدة ووظيفة المناعة وبعض مقاييس الدم وصفات السائل المنوى وذلك على الطيور المرباة تحت ظروف الجو الحار.تم استخدام عدد120 طائرمن سلالة انشاص عمر30 أسبوع (96 دجاجة و24ديك) تم تقسيم هذه الطيور عشوائياً إلى 4 مجاميع بكل مجموعة 3 مكررات وبكل مكرر 10 طيور(8 دجاجات + 2 ديك) أى (4 مجاميع × 3 مكررات × 10 طيور(8 دجاجات + 2 ديك)). وكانت المعاملات التجريبية كالتالى:المعاملة الاولى : عليقة أساسية (كنترول بدون إضافة أى مصدر سيلينيوم). المعاملة الثانية : عليقة أساسية + 0,3 ملليجرام سيلينيوم (صوديوم سيلينيت) / كجم علف.المعاملة الثالثة : عليقة أساسية + 0,3 ملليجرام نانو سيلينيوم / كجم علف.المعاملة الرابعة : عليقة أساسية + 0,3 ملليجرام سيلينيوم عضوى / كجم علف.واستمرت فترة التجربة من عمر 30 وحتى 42 اسبوع.وكانت أهم النتائج المتحصل عليها كما يلى: أدت إضافة 0,3 جزء فى المليون من السيلينيوم العضوى (سيلبلكس) أو النانو سيلينيوم إلى تحسن معنوى فى الكفاءة التحويلية للغذاء والنسبة المئويةلإنتاج البيض وكتلة البيض مقارنة بالكنترول أو إضافة الصوديوم سيلينيت. بينما لم يكن هناك فروق معنوية فى متوسط كمية الغذاء المأكول والتغير فى وزن الجسم الحى.أدت إضافة 0,3 جزء فى المليون من السيلينيوم العضوى أو النانو سيلينيوم إلى انخفاض معنوى فى مستوى الليبيدات الكلية والـ AST فى بلازما الدم مقارنة بالكنترول أو إضافة الصوديوم سيلينيت. ادت إضافة المصادر المختلفة من السيلينيوم إلى زيادة معنوية فى مستوى البروتينات الكلية والجلوبيولين فى بلازما الدم مقارنة بالكنترول، وكانت أعلى زيادة عند إضافة 0,3 جزء فى المليون من النانو سيلينيوم حيث بلغت 25,91، 50,75 % على الترتيب.أدت إضافة 0,3 جزء فى المليون من السيلينيوم العضوى أو النانو سيلينيوم إلى انخفاض معنوى فى مستوى المالونالدهيد MDA بينما زاد معنوياً الجلوتاثيون بيروكسيديز GSH-PX فى بلازما كل من الدم والسائل المنوى مقارنة بالكنترول. أدت إضافة 0,3 جزء فى المليون من السيلينيوم العضوى أو النانو سيلينيوم إلى حدوث تأثيرإيجابى ومعنوى فى كل من الـ Phagocytic activity (PA) والـ Phagocytic index (PI)وعموماً يمكن القول أن استخدام مستوى 0,3 جزء فى المليون من السيلينيوم العضوى أوالنانو سيلينيوم أدى إلى تحسين فى الأداء الإنتاجى وصفات السائل المنوى والخصائص المضادة للأكسدة ووظيفة المناعة فى الدجاج البياض المربى تحت ظروف الجو الحار.