



Original Research Article

Dietary sources and levels of selenium supplements affect growth performance, carcass yield, meat quality and tissue selenium deposition in broilers



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ABSTRACT

This study examined the effects of sources and levels of selenium (Se) on performance, carcass parts yield, meat quality and tissue Se concentration in broilers. A total of 960 one-day-old male broilers were divided into 8 treatments in a 4 × 2 factorial arrangement. Chicks were penned in groups of 20 with 6 pens per group. Selenium sources were sodium selenite (SS), Se enriched yeast (SY), DL-selenomethionine (SM) and nano-selenium (NS) and dietary supplemental Se levels were 0.1 and 0.3 mg/kg diet. The average daily gain (ADG), average daily feed intake (ADFI), feed:gain ratio, mortality, and carcass parts yield were not affected by dietary treatments. The level of 0.3 mg/kg Se decreased lightness and increased yellowness of the breast and thighs ($P < 0.001$). Nano-selenium improved yellowness, redness and meat quality ($P < 0.05$). The interactive effects of sources and the levels of Se affected Se retention ($P < 0.001$). Inorganic Se showed poor retention compared to other sources of Se; and NS showed equal retention with the organic sources. With consideration to meat quality responses, NS had a more significant positive effect compared to SS as an inorganic source of Se. Overall, NS and organic sources of Se resulted in better meat quality compared with the inorganic source. Moreover, the highest Se retention percentage was achieved by supplementation of NS followed by organic sources at 0.1 mg/kg compared to SS.

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1. Introduction

Among essential trace elements in animal nutrition, selenium (Se) plays vital roles in animal health and productivity (Yoon et al., 2007; Zhou and Wang, 2011). There are 4 common sources of Se supplements in the animal diet: inorganic Se such as sodium selenite (SS) and organic Se such as Se-enriched yeast (SY), DL-selenomethionine (SM) and nano-selenium (NS).

Sodium selenite is the most widely used source in poultry diets to meet the Se requirement (Perić et al., 2009). Since the FDA (US Food and Drug Administration) approved usage of SY in the poultry diets in 2001, SY products have developed rapidly and it is claimed to have higher effectiveness than inorganic sources in poultry (Yoon et al., 2007). Selenomethionine is the primary form of Se in feed ingredients such as cereal grains and oilseed meal. The SM includes almost 50% of the Se in cereal grains (Ševčíková et al., 2006). Also, SM is the only Se-amino acid chelate that is nonspecifically incorporated into tissue proteins in place of methionine, allowing increased Se reserves in the organism (Schrauzer, 2000). Schrauzer (2000) suggested that SM is the most effective form of Se, and Wang et al. (2011) observed that application of SM improves tissues Se deposition in broilers. Nano-selenium is considered as a novel form of Se, exhibiting high absorption ability, surface activity, catalytic efficiency, and low toxicity (Wang et al., 2007). It has been reported that nanoparticle shows new characteristics of transport, uptake and exhibit higher absorption efficiencies (Davda and

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Labhasetwar, 2002). Higher bioavailability, subsequent rates of accumulation and lower toxicity are the main advantages of organic Se sources compared to inorganic (Wang et al., 2011).

Animal and poultry feed require Se supplementation to ensure health, efficient performance and good meat quality. An insufficient Se supply has negative effects on the performance of chickens (Bakhshalinejad et al., 2018). It is well understood that the level of Se in poultry feed varies widely depending on the plant ingredients provided in the ration and the Se characteristics of the soils in which the ingredients were grown. Under current practice, the recommended Se concentration in broiler diet ranges from 0.1 (NRC, 1994) to 0.3 mg/kg (FDA, 2004).

Selenium supplementation can be used for enriching meat and eggs, allowing higher human intake of Se (Perić et al., 2009). Meat products are one of the main sources of Se for human, which show high bioavailability. Therefore, meat products enriched in Se can have a great nutritional benefit for human (NRC, 2005).

There is little information in how various sources of Se behave at different levels when incorporated into broiler diets on meat quality as well as Se retention and accumulation in tissues. Thus, this study aimed to investigate the effect of various forms and levels of dietary Se on growth performance, carcass yield, meat quality and tissues Se deposition in Ross 308 broiler chickens.

2. Materials and methods

2.1. Birds and management

This project was approved by the Institution Animal Care and Use Committee at the Ferdowsi University of Mashhad, and the animal trial was conducted in accordance with the National Institutes of Health Guidelines for the care and use of experimental animals.

A total of 960 one-day-old broiler chicks (Ross 308) were obtained from a local hatchery and wing tagged, weighed and

allocated into 8 dietary treatments with 6 replicates of 20 male chicks each. The mash feed and fresh water were provided for *ad libitum* consumption throughout the experiment. Each floor pen of 1.5 m × 1.5 m × 0.8 m (length × width × height) was equipped with 6 nipple drinkers and a tube feeder, and clean hardwood shavings were used as litter. Environmental temperature was set at 33 °C on d 1 and lowered stepwise to 21 °C. For the first 7 d, light was on 24L:0D, and then lighting was lowered stepwise to 21L:3D. The birds were housed in a tunnel-ventilated building. Mortality and feed intake (FI) were recorded daily.

2.2. Experimental design and diets

A completely randomized design involving a 4 × 2 factorial arrangement of treatments was used in this study. Four supplemental Se sources were SS (Sigma–Aldrich Chemical Co., St. Louis, MO), SM (Sigma–Aldrich Chemical Co., St. Louis, MO), SY (SelPlex 2000, Alltech Inc., Nicholasville, KY) and NS, and 2 supplemental Se levels were 0.1 and 0.3 mg/kg diet.

A corn–soybean meal basal diet (Table 1) was formulated to meet the Ross 308 recommendations (Aviagen International, 2014) with regard to the all requirements except Se. The Se levels from different sources were supplemented into the basal diet to form the aforementioned experimental treatments. The analyzed Se concentrations of the diets are listed in Table 2. The feeding program was considered in 3 phases including starter, grower and finisher that fed from 1 to 10 d, 11 to 24 d and 25 to 42 d, respectively.

The SS (contained 41.7% Se on the basis of analysis) was used as the inorganic Se source, SY (contained 22.4% Se on the basis of analysis) and SM (contained 37.8% Se on the basis of analysis) were used as the organic Se sources. The NS contained 32.9% Se on the basis of analysis. The NS was prepared according to the method described by Wang et al. (2007).

The Se concentrations in feed samples were measured by hydride generation–atomic absorption spectrophotometry (HG–AAS;

Table 1
Composition of the experimental basal diet.

Item	Starter (1–10 d)	Grower (11–24 d)	Finisher (25–42 d)
Ingredients, g/kg			
Corn grain	510.4	540.7	588.5
Soybean meal	421.5	384.7	331.5
Soybean oil	23.8	35.0	43.8
Limestone	14.4	13.3	12.3
Dicalcium phosphate	15.2	13.2	11.3
Sodium chloride	3.0	3.0	3.0
Vitamin premix ¹	2.5	2.5	2.5
Mineral premix ²	2.5	2.5	2.5
DL-methionine	4.2	3.6	3.3
L-threonine	0.7	0.3	0.1
L-lysine · HCL	1.9	1.3	1.3
Nutritive value, calculated, g/kg			
ME, kcal/kg	3,000	3,100	3,200
CP	230	215	195
Ca	9.6	8.7	7.8
Available P	4.8	4.3	3.9
Lysine	14.4	12.9	11.5
Methionine	7.7	7.0	6.4
Methionine + Cysteine	10.8	9.9	9.0
Threonine	9.7	8.8	7.8
Nutritive value, analyzed, g/kg			
DM	884.2	882.9	881.7
Crude ash	57.7	53.1	47.8
Selenium, mg/kg	0.09	0.11	0.15

¹ Provides per kilogram of diets: vitamin A (retinol), 12,000 IU; vitamin D₃ (cholecalciferol), 5,000 IU; vitamin E (DL- α -tocopheryl acetate), 66.0 IU; vitamin K₃ (menadione), 2.65 mg; vitamin B₁ (thiamin), 2.97 mg; vitamin B₂ (riboflavin), 8.0 mg; vitamin B₃ (niacin), 57.42 mg; vitamin B₅ (pantothenic acid), 17.86 mg; vitamin B₆ (pyridoxine), 4.45 mg; vitamin B₉ (folic acid), 1.9 mg; vitamin B₁₂ (cyanocobalamin), 0.02 mg; vitamin H₂ (biotin), 0.18 mg; choline chloride, 487.5 mg; antioxidant 1.0 mg.

² Provides per kilogram of diets: Mn (manganese oxide), 120.6 mg; Zn (zinc oxide), 105.0 mg; Fe (iron carbonate), 20.5 mg; Cu (copper sulfate), 16.1 mg; I (calcium iodate), 1.2 mg; choline chloride, 474.0 mg.

Table 2
Dietary treatments and supplemented and analyzed concentration of selenium in diets (mg/kg).

Treatment	Supplemented values		Analyzed values ¹		
	Sources	Levels	Starter	Grower	Finisher
1	SS	0.1	0.19	0.21	0.25
2	SS	0.3	0.38	0.41	0.45
3	SY	0.1	0.19	0.21	0.24
4	SY	0.3	0.37	0.42	0.43
5	SM	0.1	0.18	0.20	0.23
6	SM	0.3	0.40	0.42	0.44
7	NS	0.1	0.19	0.22	0.24
8	NS	0.3	0.39	0.40	0.46

SS = sodium selenite; SY = selenium enriched yeast; SM = DL-selenomethionine; NS = nano-selenium.

¹ Selenium concentrations of dietary treatment were analyzed with hydride generation-atomic absorption spectrophotometry (Tinggi, 2003).

AA6501, Shimadzu Ltd., Japan) according to the methods described by Tinggi (2003). Feed samples were analyzed for dry matter and crude ash according to standard procedures of Association of Official Analytical Chemists (AOAC International, 2000). Feed samples were analyzed in triplicate.

2.3. Production performance

Birds of each replicate were weighed by digital balance (model GF 400, A&D, Weighing, CA) at the beginning and end of each feeding phases. The FI was calculated by considering the difference between given and consumed feed at the end of each feeding phases and Se intake calculated according to FI collected data and Se concentration. Based on these data average daily gain (ADG), average daily feed intake (ADFI) and feed:gain ratio were calculated. Moreover, mortality of the birds was recorded and weighed daily to adjust the FI, accordingly.

2.4. Carcass yield

After 12-h fasting at the end of the experiment (42 d of age), 24 birds were randomly selected from each treatment (3 birds per replicate pen) and killed by cervical dislocation and then bleed. Afterwards, the liver, gizzard, heart, kidneys, pancreas, spleen, total abdominal fat, thigh and breast muscles were immediately collected and weighed. The liver, kidneys, thigh and breast muscles were frozen at -20°C for further meat quality and Se concentration analyses. In all considered parameters, the skin was removed and bone-in the part.

2.5. Blood collection and analysis

Twenty-four birds were selected (3 birds per replicate pen) and blood samples were taken from the vena brachialis at the end of the experiment (42 d of age). Blood samples were spin at $3,000 \times g$ for 10 min under 4°C and were analyzed for Se concentrations by the described method of Wang et al. (2011).

2.6. Meat color measurement

Color of the right sides of the breast and thigh muscle were determined using Minolta Chroma Meter (model CR-410, Minolta Chroma Meter, Osaka, Japan) within 4 h after slaughtering with natural exposure at 25°C room temperature (Yang et al., 2012). The CIE (International Commission on Illumination) values for lightness

(L*), redness (a*) and yellowness (b*) were used for expressing of color measurement.

2.7. Drip loss and cooking loss measurements

Frozen thigh and breast samples (right sides of the carcass) were trimmed to $2\text{ cm} \times 1\text{ cm} \times 1\text{ cm}$ (length \times width \times thickness), placed in a plastic bag and fastened to avoid evaporation and left at 4°C , and the final weight was determined at 24 h postmortem. Percentage of drip loss was calculated by $100 \times (\text{initial muscle weight} - \text{final muscle weight}) / \text{initial muscle weight}$ as described by Akbari Moghaddam Kakhki et al. (2017).

Frozen thigh and breast samples (left sides of the carcass) were thawed at 2°C overnight. The thawed thigh and breasts were weighed and baked in a preheated oven at 176°C . Internal chicken thigh and breast temperatures were determined using thermocouples to reach the final internal temperature of 77°C . Cooked thigh and breasts were cooled to ambient temperature (20°C), patted dry with a paper towel, and reweighed. The formula of Schilling et al. (2010) as $100 \times [(\text{initial weight} - \text{final weight}) / \text{initial weight}]$ was used to calculate cooking loss.

2.8. Muscle pH measurement

At 24-h postmortem, right side of breast and thigh muscles were used for pH measurement using a pH meter (model 691 Laboratory pH Meter, Metrohm Co, Herisau, Switzerland) instrument at a depth of 2.5 cm below the surface, as described by Akbari Moghaddam Kakhki et al. (2017).

2.9. Shear force measurement

Tenderness was assessed using an objective texture procedure described by Schilling et al. (2010). Breast and thigh samples that were used for cooking loss determination were used for shear force (N) measurement. Two adjacent $2\text{ cm} \times 1\text{ cm} \times 1\text{ cm}$ (length \times width \times thickness) strips were cut from the cooked breast. Each strip was sheared once perpendicular to the muscle fibers by using a Warner-Bratzler shear attachment mounted on an Instron Tinius Olsen Testing Machine (model H5KS, Tinius Olsen Co, Horsham, PA) equipped with a maximum 50-N load cell and a crosshead speed of 200 mm/min. The mean was calculated for each thigh and breast meats.

2.10. Determination of mineral concentration in meat and excreta

Total excreta collection was performed on 39 through 42 d of age to determine Se apparent retention according to a modified method of Bourdillon et al. (1990). For each cage, total excreta were weighed and collected daily during the 3-d collection period. Feathers and feed particles were carefully removed from the excreta, pooled and stored at -20°C until subsequent analysis. In addition, approximately 10 g of liver, kidney and hand-deboned thigh and breast muscles samples were mixed and finely ground. The concentrations of Se in tissues and excreta samples were determined according to the method described by Tinggi (2003) using a hydride generation-atomic absorption spectrophotometry (HG-AAS; AA6501, Shimadzu Ltd., Japan). The Se apparent retention in microgram was calculated by subtracting total Se intake from the output Se of the excreta samples in each replicate. The Se apparent retention (%) was calculated according to the following formula: $100 \times [(\text{Se intake} - \text{Se excreted}) / \text{Se intake}]$ according to the method described by Yoon et al. (2007).

2.11. Statistical analysis

Data were analyzed as a 4 × 2 (source × level) factorial arrangement of treatments by two-way analysis of variance with a model including the main effects of Se source, Se level and their interaction using the general linear model (GLM) procedure of the SAS 9.4 (SAS Institute Inc, 2009). The pen was considered the experimental unit. When an effect was significant ($P < 0.05$), means were compared by a Tukey multiple-range test.

3. Results

3.1. Growth performance

As shown in Table 3, NS sources compared to SS and supplementation of Se at 0.3 mg/kg compared to 0.1 mg/kg resulted in higher ADG and lower mortality in each and the whole experimental period but not significantly ($P > 0.05$). Moreover, the Se sources or levels had no significant effects on ADFI and feed:gain ratio ($P > 0.05$) in each experimental period. No interactions between Se source and level were observed in ADG, ADFI, feed:gain ratio and mortality ($P > 0.05$) in each and the whole experimental period.

3.2. Carcass yield

Neither carcass yield nor carcass yield components such as breast and thigh muscles, liver, gizzard, heart, kidney, pancreas, spleen and abdominal fat of broilers was affected by different levels ($P > 0.05$) or sources ($P > 0.05$) of Se at 42 d of age (Table 4). No interactions between Se source and level were observed among their parameters ($P > 0.05$).

3.3. Meat color

There was no interaction between the sources and levels of Se on the breast and thigh color ($P > 0.05$; Table 5). The Se sources had significant effects on redness and yellowness of the breast and thigh ($P < 0.05$). The red and yellow color degree of the breast and thigh were significantly higher in the birds fed diets supplemented with NS source than in those fed diets supplemented with SS

($P < 0.05$). However, lightness of the breast and thigh muscles was not affected by different sources of Se ($P > 0.05$). Supplementation of Se at 0.3 mg/kg diet decreased lightness of the breast ($P = 0.011$) and thigh ($P = 0.001$) muscles, in concomitant with an increase in yellowness of the breast ($P = 0.038$) and thigh ($P = 0.001$). However, levels of Se had no significant effect on the redness of the breast and thigh muscles ($P > 0.05$).

Based on the contrast comparison, there was a difference ($P < 0.05$) between inorganic sources of Se (SS) and NS in terms of redness and yellowness of the breast and thigh muscles, in which birds fed the diet supplemented with NS had higher values of their parameters.

3.4. Meat quality

There was no interaction between the sources and levels of Se in pH, drip loss and shear force in the breast and thigh muscles ($P > 0.05$, Table 6). However, cooking loss was significantly affected by the interactive effect of Se sources and levels ($P < 0.05$). Nano-selenium supplementation at both levels and SY at 0.3 mg/kg resulted in lower cooking loss in the breast and thigh muscles compared to SS at both levels and SM at 0.1 mg/kg ($P < 0.05$). Supplemental Se in the forms of SY and NS led to lower drip loss and cooking loss in the breast muscle compared to SS ($P < 0.05$). Birds fed diets supplemented with NS had lower drip loss and cooking loss in the thigh muscle than those fed SS or SM diets ($P = 0.001$). Shear force was increased in birds fed diets supplemented with SS or SM compared to those fed diet supplemented with NS ($P < 0.05$). Supplemental levels of Se had no remarkably impact on measured meat quality parameters ($P > 0.05$).

Based on the contrast comparison, there was a difference ($P < 0.05$) between organic sources of Se and NS in terms of drip loss, cooking loss and shear force of breast muscle, in which birds fed diets supplemented with organic sources of Se (SM or SY) had higher values of their parameters. Moreover, broilers fed SS as inorganic sources of Se had higher ($P < 0.05$) cooking loss and shear force than those fed NS sources.

3.5. Tissues mineral deposition

However, there was no interaction between the sources and levels of Se in the Se concentration of the liver ($P > 0.05$, Table 7),

Table 3
Effect of different dietary levels and sources of selenium on growth performances of broilers from 1 to 42 d of age.¹

Item	ADG, g/d per bird				ADFI, g/d per bird				Feed:Gain ratio, g/g				Mortality, %
	1–10 d	11–24 d	25–42 d	1–42 d	1–10 d	11–24 d	25–42 d	1–42 d	1–10 d	11–24 d	25–42 d	1–42 d	
Se source													
SS	24.2	56.4	84.1	58.5	30.1	84.0	166.1	101.2	1.24	1.49	1.97	1.73	4.6
SM	24.3	56.9	84.9	59.1	30.0	84.0	166.6	101.3	1.23	1.48	1.96	1.72	4.4
SY	24.3	56.9	85.0	59.1	30.3	84.1	166.2	101.2	1.25	1.48	1.95	1.71	5.1
NS	24.4	57.1	85.0	59.2	30.3	84.2	167.5	101.8	1.24	1.48	1.97	1.72	3.6
SEM	0.650	0.972	1.125	0.989	0.304	0.552	0.891	1.150	0.025	0.090	0.123	0.101	0.951
Se level, mg/kg													
0.1	24.2	56.6	84.6	58.8	30.2	83.9	166.4	101.3	1.25	1.48	1.97	1.72	4.6
0.3	24.4	57.0	84.9	59.1	30.2	84.1	166.8	101.5	1.24	1.48	1.96	1.72	4.2
SEM	0.450	0.722	1.020	0.871	0.215	0.408	0.677	1.005	0.017	0.081	0.102	0.985	0.719
Source of variation, P-value													
Se source	0.405	0.199	0.107	0.251	0.937	0.630	0.062	0.218	0.912	0.327	0.207	0.131	0.128
Se level	0.089	0.064	0.640	0.098	0.942	0.135	0.391	0.186	0.531	0.304	0.553	0.211	0.361
Se source × Se level	0.219	0.371	0.421	0.874	0.333	0.844	0.268	0.117	0.149	0.708	0.308	0.303	0.955
Contrast, P-value													
Inorganic vs. Organic ²	0.405	0.069	0.780	0.910	0.979	0.935	0.498	0.472	0.756	0.090	0.068	0.089	0.764
NS vs. SY and SM	0.069	0.449	0.098	0.801	0.770	0.239	0.320	0.770	0.919	0.916	0.160	0.306	0.041
NS vs. SS	0.219	0.371	0.421	0.875	0.784	0.275	0.160	0.404	0.721	0.119	0.703	0.256	0.125

ADG = average daily gain; ADFI = average daily feed intake; SS = sodium selenite; SM = DL-selenomethionine; SY = selenium enriched yeast; NS = nano-selenium.

¹ Growth performance data are means of 6 pens with 20 chickens per each.

² Inorganic source of selenium was sodium selenite vs. organic sources of selenium including SM and selenium enriched yeast.

Table 4
Effect of different dietary levels and sources of selenium on carcass characteristics of broilers at 42 d of age (%).^{1, 2}

Item	Carcass yield	Breast muscle	Thigh muscle	Liver	Gizzard	Heart	Kidney	Pancreas	Spleen	Abdominal fat
Se source										
SS	72.50	28.20	17.62	2.13	1.27	0.45	0.93	0.22	0.15	1.48
SM	72.68	28.27	17.67	2.13	1.27	0.45	0.93	0.22	0.15	1.47
SY	72.90	28.35	17.72	2.14	1.27	0.46	0.93	0.23	0.15	1.46
NS	73.15	28.45	17.78	2.15	1.28	0.45	0.95	0.23	0.15	1.47
SEM	1.025	0.891	0.689	0.101	0.012	0.009	0.011	0.051	0.019	0.121
Se level, mg/kg										
0.1	72.76	28.29	17.68	2.14	1.27	0.46	0.93	0.22	0.15	1.47
0.3	72.86	28.34	17.71	2.14	1.27	0.45	0.93	0.22	0.15	1.48
SEM	0.987	0.719	0.545	0.098	0.009	0.007	0.010	0.035	0.012	0.109
Source of variation, <i>P</i> -value										
Se source	0.342	0.914	0.985	0.001	0.073	0.699	0.098	0.368	0.822	0.081
Se level	0.681	0.873	0.932	0.109	0.298	0.515	0.228	0.606	0.667	0.502
Se source × Se level	0.999	1.000	1.000	0.643	0.232	0.731	0.984	0.846	0.995	0.886
Contrast, <i>P</i> -value										
Inorganic vs. Organic ³	0.373	0.728	0.852	0.859	0.594	0.276	0.097	0.387	0.517	0.093
NS vs. SY and SM	0.267	0.658	0.817	0.358	0.065	0.943	0.998	0.785	0.463	0.787
NS vs. SS	0.087	0.494	0.718	0.842	0.070	0.254	0.149	0.352	0.745	0.610

SS = sodium selenite; SM = DL-selenomethionine; SY = selenium enriched yeast; NS = nano-selenium.

¹ Carcass characteristics data are means of 6 pens with 4 sacrificed broilers per each pen.

² In all considered parameters, skin was removed and bone-in the part and percentages of BW at slaughter.

³ Inorganic source of selenium was sodium selenite vs. organic sources of selenium including SM and selenium enriched yeast.

Table 5
Effect of different dietary levels and sources of selenium on meat color of broilers at 42 d of age.¹

Item	Breast muscle			Thigh muscle		
	L* (lightness)	a* (redness)	b* (yellowness)	L* (lightness)	a* (redness)	b* (yellowness)
Se source						
SS	47.0	3.48 ^b	8.73 ^b	50.1	5.50 ^b	9.63 ^b
SM	46.8	3.50 ^{ab}	8.75 ^{ab}	49.8	5.52 ^{ab}	9.76 ^{ab}
SY	46.5	3.51 ^{ab}	8.77 ^{ab}	49.6	5.53 ^{ab}	9.79 ^{ab}
NS	46.3	3.59 ^a	8.84 ^a	49.4	5.61 ^a	9.82 ^a
SEM	0.095	0.033	0.023	0.082	0.010	0.033
Se level, mg/kg						
0.1	47.7 ^a	3.50	8.76 ^b	50.4 ^a	5.53	9.67 ^b
0.3	46.6 ^b	3.51	8.82 ^a	49.6 ^b	5.53	9.78 ^a
SEM	0.059	0.012	0.012	0.047	0.008	0.010
Source of variation, <i>P</i> -value						
Se source	0.325	0.001	0.001	0.549	0.001	<0.001
Se level	0.011	0.101	0.038	0.001	0.131	0.001
Se source × Se level	0.999	0.959	0.648	0.999	0.881	0.558
Contrast, <i>P</i> -value						
Inorganic vs. Organic ²	0.235	0.154	0.125	0.109	0.143	0.741
NS vs. SY and SM	0.541	0.325	0.179	0.480	0.713	0.379
NS vs. SS	0.120	<0.001	<0.001	0.091	<0.001	<0.001

SS = sodium selenite; SM = DL-selenomethionine; SY = selenium enriched yeast; NS = nano-selenium.

^{a, b} Values within the same column with different superscript letters differ ($P < 0.05$).

¹ Meat color data are means of duplicated analysis of 24 samples per each treatment.

² Inorganic source of selenium was sodium selenite vs. organic sources of selenium including SM and selenium enriched yeast.

but Se concentration in the serum, kidney, breast and thigh muscles was affected by the interactive effects of the sources and levels of Se ($P < 0.05$). Nano-selenium, SY and SM supplementation at the level of 0.3 mg/kg resulted in higher Se concentration in the thigh muscle and serum compared to other treatments ($P < 0.05$), and NS and SY supplementation at the level of 0.3 mg/kg also resulted in higher Se concentration in the breast muscle compared to other treatments ($P < 0.05$). Supplemental Se in the forms of NS led to higher ($P = 0.001$) Se concentration in the breast and thigh muscles compared to other sources of Se. The highest Se concentration in the liver and kidney was observed in birds fed SS and SM, and those fed diets supplemented with NS showed the lowest Se concentration ($P < 0.05$). Supplemental Se in the forms of NS or SY led to higher ($P = 0.001$) Se concentration in the serum compared to SS or SM. An increase in the supplemental level of Se at 0.3 mg/kg

enhanced Se concentration in the serum, liver, kidney, breast and thigh muscles ($P < 0.05$).

Based on the contrast comparison, significant differences ($P < 0.05$) between the effect of organic or inorganic Se sources and NS were observed in Se concentration in the tissues in which birds fed diets supplemented with NS had a higher of Se concentration in the breast and thigh muscles and a lower concentration of Se concentration in the liver and kidney.

3.6. Selenium retention

The effect of different dietary Se sources and levels on Se retention of broiler chickens at 42 d of age are shown in Table 7. The value and percentage of Se retention were influenced by the interactive effects of the sources and levels of Se ($P < 0.05$). Nano-

Table 6
Effect of different dietary levels and sources of selenium on meat quality of broilers at 42 d of age.^{1,2}

Item	Breast muscle				Thigh muscle			
	pH	Drip loss, %	Cooking loss, %	Shear force, N	pH	Drip loss, %	Cooking loss, %	Shear force, N
0.1 mg/kg SS	5.90	4.46	26.46 ^a	34.36	6.59	0.95	32.58 ^a	25.13
0.3 mg/kg SS	5.89	4.45	26.15 ^a	34.25	6.57	0.93	32.20 ^a	25.06
0.1 mg/kg SM	5.88	3.96	26.35 ^a	34.22	6.55	0.93	32.58 ^a	25.01
0.3 mg/kg SM	5.86	3.92	26.09 ^{ab}	34.09	6.52	0.91	32.27 ^a	24.93
0.1 mg/kg SY	5.85	3.58	25.89 ^b	34.02	6.46	0.89	32.01 ^{ab}	24.89
0.3 mg/kg SY	5.85	3.57	25.58 ^{bc}	33.89	6.33	0.88	31.63 ^b	24.79
0.1 mg/kg NS	5.84	3.18	25.32 ^c	33.82	6.20	0.86	31.31 ^{bc}	24.74
0.3 mg/kg NS	5.81	3.17	24.80 ^c	33.72	6.10	0.85	30.67 ^c	24.66
SEM	0.099	0.041	0.055	0.045	0.079	0.037	0.080	0.039
Se source								
SS	5.90	4.45 ^a	26.31 ^a	34.31 ^a	6.57	0.94 ^a	32.39 ^a	25.09 ^a
SM	5.87	3.94 ^{ab}	26.22 ^{ab}	34.15 ^a	6.54	0.92 ^a	32.42 ^a	24.97 ^a
SY	5.85	3.58 ^{bc}	25.73 ^{bc}	33.95 ^{ab}	6.39	0.88 ^{ab}	31.82 ^{ab}	24.84 ^{ab}
NS	5.82	3.18 ^c	25.06 ^c	33.77 ^b	6.15	0.85 ^b	30.99 ^b	24.70 ^b
SEM	0.052	0.008	0.028	0.021	0.028	0.015	0.044	0.028
Se level, mg/kg								
0.1	5.87	3.80	26.00	34.10	6.45	0.90	32.12	24.94
0.3	5.84	3.78	25.66	33.69	6.38	0.89	31.69	24.86
SEM	0.047	0.006	0.018	0.015	0.020	0.009	0.031	0.020
Source of variation, <i>P</i> -value								
Se source	0.095	<0.001	0.001	<0.001	0.131	0.001	<0.001	0.001
Se level	0.141	0.561	0.387	0.425	0.716	0.377	0.626	0.189
Se source × Se level	0.502	0.506	<0.001	0.913	0.452	0.902	0.001	0.967
Contrast, <i>P</i> -value								
Inorganic vs. Organic ³	0.076	0.001	0.091	0.154	0.395	0.756	0.119	0.377
NS vs. SY and SM	0.421	0.031	0.001	<0.001	0.191	0.030	0.001	0.102
NS vs. SS	0.914	<0.001	<0.001	<0.001	0.082	<0.001	<0.001	<0.001

SS = sodium selenite; SM = DL-selenomethionine; SY = selenium enriched yeast; NS = nano-selenium.

^{a-c} Values within the same column with different superscript letters differ ($P < 0.05$).¹ Meat quality characteristics data are means of duplicated analysis of 24 samples per each treatment.² Measurements were performed at 24-h post mortem.³ Inorganic source of selenium was sodium selenite vs. organic sources of selenium including SM and selenium enriched yeast.**Table 7**
Effect of different dietary levels and sources of selenium on selenium concentration in tissues and Se retention of broilers at 42 d of age.¹

Item	Selenium concentration					Se retention	
	Breast muscle, µg/g	Thigh muscle, µg/g	Liver, µg/g	Kidney, µg/g	Serum, µg/L	µg	%
0.1 mg/kg SS	1.36 ^c	1.05 ^d	3.37	2.36 ^d	1.03 ^c	11.38 ^g	84.84 ^c
0.3 mg/kg SS	4.08 ^b	3.07 ^b	7.11	6.10 ^a	1.29 ^b	31.88 ^c	83.59 ^c
0.1 mg/kg SM	1.45 ^c	1.15 ^d	3.95	2.27 ^{de}	1.06 ^c	13.72 ^f	88.44 ^b
0.3 mg/kg SM	4.09 ^b	3.28 ^{ab}	6.92	5.91 ^a	1.40 ^{ab}	34.24 ^b	85.13 ^{bc}
0.1 mg/kg SY	1.49 ^c	1.36 ^c	3.19	2.18 ^{de}	1.16 ^{bc}	12.05 ^{fg}	88.52 ^b
0.3 mg/kg SY	4.20 ^{ab}	3.39 ^a	6.63	5.62 ^b	1.47 ^a	35.51 ^{ab}	86.11 ^{bc}
0.1 mg/kg NS	1.58 ^c	1.57 ^c	3.09	2.08 ^e	1.27 ^b	17.44 ^d	94.06 ^a
0.3 mg/kg NS	4.30 ^a	3.50 ^a	6.15	5.13 ^c	1.68 ^a	36.80 ^a	87.04 ^b
SEM	0.029	0.033	0.135	0.032	0.023	1.001	1.132
Se source							
SS	2.72 ^b	2.06 ^b	5.24 ^a	4.23 ^a	1.16 ^b	19.74 ^c	84.21 ^c
SM	2.77 ^b	2.21 ^b	5.43 ^a	4.09 ^{ab}	1.23 ^{ab}	22.18 ^b	86.78 ^{bc}
SY	2.84 ^{ab}	2.38 ^{ab}	4.91 ^b	3.90 ^b	1.32 ^a	22.09 ^b	87.31 ^b
NS	2.95 ^a	2.54 ^c	4.62 ^c	3.61 ^c	1.47 ^a	25.58 ^a	90.55 ^a
SEM	0.011	0.009	0.116	0.012	0.017	0.852	0.988
Se level, mg/kg							
0.1	1.47 ^b	1.28 ^b	3.40 ^b	2.22 ^b	1.13 ^b	12.89 ^b	88.96 ^a
0.3	4.17 ^a	3.31 ^a	6.70 ^a	5.69 ^a	1.46 ^a	31.91 ^a	85.47 ^b
SEM	0.010	0.006	0.101	0.009	0.010	0.452	0.684
Source of variation, <i>P</i> -value							
Se source	0.001	0.001	0.007	0.001	0.001	0.001	<0.001
Se level	<0.001	<0.001	<0.001	0.001	0.003	0.003	<0.001
Se source × Se level	<0.001	<0.001	0.063	<0.001	0.001	<0.001	<0.001
Contrast, <i>P</i> -value							
Inorganic vs. Organic ²	0.641	0.038	0.158	0.001	0.002	0.001	0.005
NS vs. SY and SM	0.009	0.004	0.010	0.001	0.138	<0.001	<0.001
NS vs. SS	<0.001	<0.001	0.001	<0.001	<0.001	<0.001	<0.001

SS = sodium selenite; SM = DL-selenomethionine; SY = selenium enriched yeast; NS = nano-selenium.

^{a-g} Values within the same column with different superscript letters differ ($P < 0.05$).¹ Values are the means of 6 replicates with 4 broilers per replicate.² Inorganic source of selenium was sodium selenite vs. organic sources of selenium including SM and selenium enriched yeast.

selenium had the highest retention (% and μg), followed by organic sources and SS in which the increase in supplemental level increased the amount of preserved Se ($P < 0.05$). An increase in the supplemental level of Se at 0.3 mg/kg improved Se retention value ($P = 0.003$) but decreased the percentage of Se retention ($P < 0.001$).

Based on the contrast comparison, there was a considerable difference between organic sources of Se (SY and SM) and inorganic Se sources (SS) ($P < 0.05$). Furthermore, the result of NS supplementation significantly differed from SY and SM in the percent of Se apparent retention ($P < 0.001$).

4. Discussion

Selenium supplementation did not influence ($P > 0.05$) the ADG, ADFI, mortality, and feed:gain ratio of broiler chickens during the experimental period. These results agreed with those of Bakhshalinejad et al. (2018), who reported no significant effect of the Se sources and levels on the growth performance of broilers during the first 21 d of life. Several other studies on broilers concluded that Se source (SS, SY, SM, and NS) did not influence the growth parameters under the controlled condition (Payne and Southern, 2005; Yoon et al., 2007; Niu et al., 2009; Perić et al., 2009). In our experimental conditions, the amount of Se in the basal diet (0.09 to 0.15 mg Se/kg of feed) appeared sufficient to maintain growth performance of broilers.

Ineffective supplementation of Se by different sources and levels on carcass, breast, thigh muscles yields and relative organs weight was in agreement with the findings of Downs et al. (2000) and Payne and Southern (2005) who did not observe any differences in carcass, breast and thigh muscles yields in broilers fed diets supplemented with SS or organic Se. The present results are in line with the study of Cai et al. (2012) who reported no significant effect of NS on the weights of carcass parts in broilers.

Meat color as an essential quality attribute for consumers is affected by several factors including pH, myoglobin concentration, nitrites, etc. (King and Whyte, 2006). Some studies have shown that Se could significantly improve serum glutathione peroxidase activity, enhance oxidation resistance, effectively prevent the myoglobin or oxymyoglobin from being oxidized to metmyoglobin, deepen the muscle chroma, and improve meat color of broilers (Cai et al., 2012; Oliveira et al., 2014). Yang et al. (2012) reported that broilers fed diets supplemented with SY as an organic source at 0.3 mg/kg had a higher redness in their breast and thigh compared with those fed SS.

Similar to our observation, it has been reported that sources and levels of Se had no effects on 24-h pH in the breast or thigh meats of broiler chickens (Perić et al., 2009; Wang et al., 2011; Oliveira et al., 2014). However, other studies have shown that the use of organic sources promotes an increase in pH of chicken and pork in relation to SS (Boiago et al., 2014; Calvo et al., 2017). Calvo et al. (2017) observed a positive correlation between the higher pH values for pork, which is contrary to the present study. The ability of muscle proteins to absorb water and hold it within the cells is of paramount importance to meat quality. Supplementation of Se by organic sources and NS led to a reduction in drip and cooking loss, which can be explained by their higher bioavailability compared with inorganic Se, and another hypothesis is the nanoparticle and organic source of Se are more effective in combating oxidation and hence preserving cell membranes. Mahan et al. (1999) concluded that inorganic Se might act as a tissue desolator pro-oxidant. In previous studies, Zhou and Wang (2011) showed that birds fed diets supplemented with NS at 0.3 mg/kg had 45.10% lower drip loss due to the improved integrity of cell membranes compared to those fed diets supplemented with organic source. Our results are consistent

with those of Perić et al. (2009) and Boiago et al. (2014), who verified a better action of organic sources of Se on the maintenance of muscle cell integrity and promoting lower drip and cooking loss. Similarly, Naylor and Choct (2000) and Yang et al. (2012) reported an improvement in cooking loss of the breast muscle in response to supplementation 0.3 mg/kg of Se from SY source. In the current study, supplementation of Se by SY and NS could improve physicochemical characteristics of the meat. However, SM did not affect the meat quality compared to SS.

The response of Se deposition in tissues was consistent with the concept that organic Se tends to be deposited more than inorganic Se does in slow turnover tissues, such as breast and thigh muscles (Schrauzer, 2003). Our results indicated that the Se concentration in breast and thigh muscles by SY and NS source was higher than in SS, and agreed with other researchers (Payne and Southern, 2005; Zhou and Wang, 2011; Hu et al., 2012). It is likely that organic sources of Se, such as SY, can be absorbed by active transport and nonspecifically incorporated into proteins in place of Met, and is preferentially absorbed and utilized by the body over inorganic Se (Schrauzer, 2003). Oliveira et al. (2014) showed that SY was retained at higher concentrations in the duodenum, jejunum, and ileum than SS, and Downs et al. (2000) found out that the Se content of the breast muscle is 2.2 times more in broilers fed diet supplemented with the organic Se supplementation than that with the inorganic Se. Selenomethionine and SY can be transformed to selenocysteine through the trans-selenation pathway and then be lysed by β - and γ -lyase to selenide (Suzuki, 2005). In addition, SM can be utilized for the synthesis of proteins without the body distinguishing. Thus, organic sources of Se (SM and SY) might be easily utilized in the tissue than SS (Suzuki, 2005). Similar to our result, it has been reported that Se concentration in the liver (Zhou and Wang, 2011) and blood (Cai et al., 2012) is dependent on the supplemental level of Se, and Se concentration in kidney and liver also increased linearly with an increase in dietary Se concentration (Echevarria et al., 1988). Spears et al. (2003) observed that hepatic Se content was increased in broilers fed diets contained Zn-SM compared to those fed a diet with SS. However, Cantor et al. (1982) reported no difference in hepatic Se concentration in pullets fed SS or SM supplemented diets.

Biofortification of meat with utilization of nanotechnology is one of the recently developed way to improve meat quality and their retention rate is considered to be a criterion for mineral utilization in animals (Liao et al., 2010). Different absorption and metabolic pathways can be attributed to the different retention rate of various sources of Se (Zeng, 2009). In our study, the highest Se retention in percentage and microgram resulted from supplementation of 0.1 and 0.3 mg/kg NS, respectively. Modest organic sources and nanoparticles of Se supplementation can easily saturate selenoenzymes, and thereby markedly increasing the retention of Se (Zeng, 2009). Higher excretion via the urine leads to less efficiency of Se retaining. Choct and Naylor (2004) observed higher excreted Se in 38-d male broilers fed diets supplemented with SS compared to those fed an organic Se source (a commercial product). It has been reported that nanoparticles show new characteristics of transport and uptake, and exhibit higher absorption efficiencies which can explain the higher retention of NS (Liao et al., 2010).

5. Conclusion

In conclusion, the results from this study showed that Se is an essential micronutrient in improving meat quality. The different sources and supplemental levels of Se could affect meat color. The physicochemical characteristics of the meat were mainly influenced by sources of Se. The organic sources (SY or SM) and NS resulted in a better meat quality compared with the inorganic

source (SS). The interaction between the sources and levels of Se dramatically influenced its accumulation in tissues and retention. The highest retention percentage was achieved by the supplementation of NS followed by organic sources at 0.1 mg/kg compared to SS. Thus, more researches are needed to study interactive effects of different sources of Se and a broader range of supplemental Se on meat quality and antioxidant enzymes activity.

Conflict of interest

None of the authors has personal conflict of interest.

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