



## Effect of Different Levels of Nano-selenium on Performance, Blood Parameters, Immunity and Carcass Characteristics of Broiler Chickens

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### Abstract

The aim of the present study was to test the hypothesis that nano-selenium inclusion in broilers' diets can improve productivity and metabolic functions of broilers. Feed and water were provided *ad libitum*. A total of 180 one-day old male Ross 308 chicks were randomly assigned to six groups based on a completely randomized design, each with three replicates of 10 birds. One of the groups served as the control (CON) and was given a basal diet without further dietary supplementation, whereas the other five groups were offered the same starter and grower diets further supplemented with dietary nano-selenium (NS) at 0.1 mg/kg of feed (NS1), 0.2 mg/kg of feed (NS2), 0.3 mg/kg of feed (NS3), 0.4 mg/kg of feed (NS4), and 0.5 mg/kg of feed (NS5). Nano-selenium dietary supplementation significantly improved weight gain and feed conversion ratio in starter (1<sup>st</sup>-21<sup>st</sup> day), grower (22<sup>nd</sup>-42<sup>nd</sup> day) and whole (1<sup>st</sup>-42<sup>nd</sup>) periods of experiment ( $P < 0.05$ ). At the same time, energy and protein utilization was more efficient in NS supplemented groups than the control ( $P < 0.05$ ). Breast and drumsticks percentages had higher values in the NS supplemented birds than the control ( $P < 0.05$ ), while abdominal fat percentage had lower values in the NS supplemented birds than the control ( $P < 0.05$ ). Significant differences in relative weight of testes were observed between treatments ( $P < 0.05$ ). Glucose and total protein concentrations in blood plasma were not significantly different among the experimental groups ( $P > 0.05$ ). While, albumin levels in blood were decreased and anti-Newcastle disease hemagglutination-inhibition titer was increased after the dietary supplementation with the nano-selenium ( $P < 0.05$ ). As conclusion, the current study demonstrated that the supplementation of nano-selenium in broiler diets could improve growth performance, carcass components and immune function, without negative effects on internal organs, and other carcass parameters and gastrointestinal parts.

### Introduction

Selenium is one of essential minerals required for optimal growth and productivity in birds. It supports multiple functions related to poultry production, fertility, and disease prevention. Selenium - as an integral part of the enzyme glutathione peroxidase- serves as an

antioxidant enzyme and helps to control levels of hydrogen peroxide and lipid peroxides. These metabolites are produced during normal metabolic activity (Rotruck *et al.*, 1973; Canoğullari *et al.*, 2010; Baylan *et al.*, 2011). Van Beirendonck *et al.* (2016) found there is a relation between selenomethionine content in dietary

selenium sources and selenium deposition in broiler muscle tissue. Rao *et al.* (2016) stated supplementing organic forms of selenium had positive effect on performance, anti-oxidant and immune responses in broiler chicken reared in tropical summer. There are some evidences on positive effects of organic selenium on hen performance and productivity of broiler breeders (Rajashree *et al.*, 2014). Meanwhile there is a relationship between fatty acids profile of meat from broiler chickens supplemented with inorganic or organic selenium (del Puerto *et al.*, 2017).

The maximum amount for selenium in diets has been set at 0.5 mg/kg based on the European Union (2004) recommendation to ensure feed safety. The bioavailability of selenium is largely correlated with its physical form. Nano-selenium (NS) has attracted widespread attention nowadays, since nanometer particulates exhibit novel characteristics such as large surface, excellent surface activity, good catalytic efficiency, high absorbing ability, and low toxicity (Wang *et al.*, 2007; Zhang *et al.*, 2008). Some data exist that evaluate growth performance parameters of commercial broilers fed with NS supplemented diets (Wang *et al.*, 2007; Wang, 2009; Cai *et al.*, 2012; Hu *et al.*, 2012; Mahmoud *et al.*, 2016; Moghaddam *et al.*, 2017). However more researches are necessary for determination of its optimal doses in different fields. The aim of the present study was therefore to test the hypothesis that NS inclusion in broilers' diets can improve productivity and metabolic functions of broilers.

## Materials and Methods

### Animals and treatments

This study was conducted at a commercial poultry farm at Rasht, Iran equipped to necessary facilities and conditions for research. A total of 180 one-day old male (Ross 308) chicks were individually randomly assigned to six groups, each with three replicates of 10 birds based on a completely randomized design. Each replicate was housed in a floor pen (1 m × 1 m). Thermo-neutral ambient temperature was maintained in accordance to standard brooding practices and adapted to the birds rearing stages (Aviagen, 2014). Light regime was regulated as follows: 23 h light and 1 h dark (1<sup>st</sup>-7<sup>th</sup> day), 20 h

light and 4 h dark (8<sup>th</sup>-39<sup>th</sup> day), and 22 h light and 2 h dark (40<sup>th</sup>-42<sup>nd</sup> day). The birds were vaccinated as water drinking against infectious Bronchitis virus (IBV) (H120; 1<sup>st</sup> day of age), Newcastle disease (8<sup>th</sup> and 21<sup>st</sup> day of age), influenza (1<sup>st</sup> day of age) and Gumboro disease (IBD071IR; 14<sup>th</sup> and 23<sup>rd</sup> day of age).

Feed and water were provided *ad libitum* in chute feeders and conical drinkers, apart from the 1<sup>st</sup> week when feeder trays were used. One of the groups served as the control (CON) and was given a basal diet without further dietary supplementation, whereas the other five groups were offered the same starter and grower diets further supplemented either with dietary NS (Farzanegan Co, Iran) at 0.1 mg/kg of feed (NS1), or 0.2 mg/kg of feed (NS2), 0.3 mg/kg of feed (NS3), 0.4 mg/kg of feed (NS4), 0.5 mg/kg of feed (NS5). Diets were formulated according to Ross manual recommendations and offered as mash form. Table 1 shows the ingredients and the composition of the basal (control) starter and grower diets used in the present experiment.

### Performance and carcass characteristics

Body weight and feed intake were weekly recorded grouply for each pen. Feed conversion ratio, energy intake, energy efficiency ratio, protein intake, and protein efficiency ratio were further calculated based on conventional protocols (Aziz-Mousavi *et al.*, 2015). Carcass characteristics measured based on Poorghasemi *et al.* (2013). Briefly, at the age of 42<sup>nd</sup> day, one chick per replicate (three chicks per treatment) was selected close to mean body weight of replicate, fasted for 4h. Feet were separated from the carcass in the tibio-tarsal joint. Weights of carcass parts, abdominal fat, internal organs (liver, thymus, heart, lungs, kidneys, pancreas, testes), and gastrointestinal tract characteristics (crop, proventriculus, gizzard, right and left cecum) were measured. The length, width and wall thickness of left and right cecum were also recorded. Total weight of all dissected parts and the weights of various segments of the digestive tract were expressed as a percentage of carcass, according to the following formula: [(weight of component(s)/carcass weight) × 100].

The procedures used in the present study were approved by the Ethic Committee of the Islamic Azad University, and was conducted in respect to the International Guidelines for research involving animals.

**Table 1.** Feed ingredients and nutrient analysis of the basal diet (% , unless mentioned)

Ingredients	Starter (1 <sup>st</sup> -21 <sup>st</sup> day of age)	Grower (22 <sup>nd</sup> -42 <sup>nd</sup> day of age)
Corn	58.6	61.6
Soybean meal (44% CP)	36.2	33.5
Soybean oil	1.40	1.50
Calcium Carbonate	0.80	0.90
Gluten meal	0.65	0.16
Dicalcium Phosphate	1.30	1.15
NaCl	0.25	0.32
Mineral premix*	0.25	0.25
Vitamin premix**	0.25	0.25
DL-Methionine	0.25	0.30
L-Lysine hydrochloride	0.05	0.07
<i>Nutrient analysis</i>		
ME (Kcal/kg)	2950	3000
Crude Protein	21.0	20.0
Calcium	0.95	0.90
Available Phosphorus	0.47	0.45
Sodium	0.17	0.15
Chloride	0.18	0.17
Lysine	1.12	1.05
Methionine	0.48	0.45
Methionine + Cystine	0.80	0.75
Threonine	0.74	0.70

\*Supplied per Kg of mixture: 1,081 mg *trans*-retinol; 20 mg cholecalciferol; 4 mg  $\alpha$ -tocopherol acetate; 800 mg menadione; 720 mg thiamine; 2,640 mg riboflavin; 4,000 mg niacin; 12,000 mg calcium pantothenate acid; 1,200 mg pyridoxine; 400 mg folic acid; 6 mg cyanocobalamin; 40 mg biotin; 100,000 mg choline; 40,000 mg antioxidant.

\*\* Supplied per Kg of mixture: 39,680 mg manganese; 20,000 mg iron; 33,880 mg zinc; 4,000 mg copper; 400 mg iodine; 80 mg selenium; 1 mg excipient.

### Plasma metabolites and hepatic enzymes

Blood constitutes measured based on Jahanpour et al. (2013). Briefly, at the end of the experiment (42<sup>nd</sup> day) one bird from each replicate (three chicks per treatment) was randomly selected for blood sampling. Collection of blood was performed early in the morning to minimize the circadian variations in the examined plasma parameters. Feed was also removed for a period of 4 h before sampling for the same reason. Blood samples (5 mL/bird) were collected from the wing vein (*Vena cutaneaulnaris*) into tubes coated with 10 mg of the anticoagulant ethylenediaminetetraacetic acid (EDTA). Samples were rapidly transferred to the laboratory (within 2 h of collection), centrifuged at 3000  $\times$  g for 10 min at room temperature and blood plasma was stored at -20°C for further analyses. Blood parameters determined in the present study were glucose (Glu), albumin (Al), and total protein (TP) by using a Roche Cobas Integra autoanalyzer (Roche Diagnostics, GmbH, Mannheim, Germany). Their analysis was performed using commercial kits (Pars Azmoon Co., Tehran, Iran), according to the manufacturer's instructions, as described in

previous studies (Nahavandinejad et al., 2014; Shabani et al., 2015).

### Immune competency

One bird per replicate (three birds per treatment) was randomly selected and blood samples were collected from the brachial vein at the 26<sup>th</sup> day of age. Serum was separated by centrifugation (3000  $\times$  g for 15 min) and was stored at -20°C until further analyses. Response to the Newcastle lentogenic vaccine (vaccine titres) was assessed based on the haemagglutination inhibition (HI) assays following the procedure described in previous works (Seidavi et al., 2014; Ebrahimi et al., 2015).

### Statistical analysis

Shapiro-Wilks test confirmed the normal distribution of data and were therefore analyzed according to a completely randomized experimental design involving six treatments by using the General Linear Model procedures of the IBM SPSS Statistics 21 software for Windows® (SPSS, 1997). Significant differences were assessed via Tukey's post hoc test at 0.05 significance level and the results are presented as means  $\pm$  standard error of means (SEM).

## Results and Discussion

As shown in Table 2, nano-selenium dietary supplementation significantly improved weight gain and feed conversion ratio in starter, grower, and whole periods of experiment. These findings can be due to higher requirement of broilers to selenium. At the same time, energy and protein utilization was more efficient in NS supplemented groups than the control. In fact, selenium have some roles in energy metabolism

(Hawkes and Keim, 2003), and these findings are predictable. Improved daily weight gain and feed conversion ratio were also observed in avian broilers (Wang and Xu, 2008), or Guangxi Yellow chickens (Zhou and Wang, 2011) fed a diet supplemented with nano-selenium at the doses of 0.2-0.5 mg/kg. It could be due higher absorption/utilization of selenium compared to CON group.

**Table 2.** Growth performance parameters as affected by the different levels of dietary nano-selenium supplementation

Groups*	Feed intake (g/day)	Weight gain (g/day)	Feed Conversion Ratio	Energy Intake (kcal/day)**	Energy Efficiency Ratio (kcal/g)**	Protein Intake (g/day)**	Protein Efficiency Ratio**
Starter period (1 <sup>st</sup> -21 <sup>st</sup> days of age)							
CON	47.667	31.841 <sup>c</sup>	1.497 <sup>a</sup>	144.192	4.528 <sup>a</sup>	10.963	0.344 <sup>a</sup>
NS1	47.302	32.381 <sup>bc</sup>	1.461 <sup>b</sup>	143.087	4.420 <sup>b</sup>	10.879	0.336 <sup>b</sup>
NS2	46.063	33.063 <sup>abc</sup>	1.393 <sup>d</sup>	139.342	4.215 <sup>d</sup>	10.595	0.320 <sup>d</sup>
NS3	47.683	34.317 <sup>bc</sup>	1.390 <sup>d</sup>	144.240	4.204 <sup>d</sup>	10.967	0.320 <sup>d</sup>
NS4	48.444	34.698 <sup>a</sup>	1.396 <sup>d</sup>	146.544	4.223 <sup>d</sup>	11.142	0.321 <sup>d</sup>
NS5	47.556	33.476 <sup>abc</sup>	1.421 <sup>c</sup>	143.856	4.297 <sup>c</sup>	10.938	0.327 <sup>c</sup>
P-value	0.530	0.058	<0.001	0.530	<0.001	0.530	<0.001
SEM	0.837	0.640	0.004	2.533	0.013	0.193	0.001
Grower period (22 <sup>nd</sup> -42 <sup>nd</sup> days of age)							
CON	159.476	73.317 <sup>d</sup>	2.175 <sup>a</sup>	502.350	6.852 <sup>a</sup>	33.490	0.457 <sup>a</sup>
NS1	160.556	76.254 <sup>c</sup>	2.106 <sup>b</sup>	505.750	6.632 <sup>b</sup>	33.717	0.442 <sup>b</sup>
NS2	163.111	78.968 <sup>abc</sup>	2.065 <sup>cd</sup>	513.800	6.506 <sup>cd</sup>	34.253	0.434 <sup>cd</sup>
NS3	164.889	80.524 <sup>a</sup>	2.047 <sup>d</sup>	519.400	6.449 <sup>d</sup>	34.627	0.430 <sup>d</sup>
NS4	164.016	79.476 <sup>ab</sup>	2.064 <sup>cd</sup>	516.650	6.501 <sup>cd</sup>	34.443	0.433 <sup>cd</sup>
NS5	162.254	77.587 <sup>bc</sup>	2.091 <sup>bc</sup>	511.100	6.587 <sup>bc</sup>	34.073	0.439 <sup>bc</sup>
P-value	0.454	0.001	<0.001	0.454	<0.001	0.454	<0.001
SEM	2.054	0.859	0.011	6.470	0.034	0.431	0.002
Whole period (1 <sup>st</sup> -42 <sup>nd</sup> days of age)							
CON	103.571	52.579 <sup>d</sup>	1.970 <sup>a</sup>	326.250	6.205 <sup>a</sup>	21.750	0.414 <sup>d</sup>
NS1	103.929	54.317 <sup>c</sup>	1.913 <sup>b</sup>	327.375	6.027 <sup>b</sup>	21.825	0.402 <sup>c</sup>
NS2	104.587	56.016 <sup>ab</sup>	1.867 <sup>cd</sup>	329.450	5.881 <sup>cd</sup>	21.963	0.392 <sup>cd</sup>
NS3	106.286	57.421 <sup>a</sup>	1.851 <sup>d</sup>	334.800	5.830 <sup>d</sup>	22.320	0.389 <sup>d</sup>
NS4	106.230	57.087 <sup>a</sup>	1.861 <sup>d</sup>	334.625	5.862 <sup>d</sup>	22.308	0.391 <sup>d</sup>
NS5	104.905	55.532 <sup>bc</sup>	1.889 <sup>bc</sup>	330.450	5.951 <sup>bc</sup>	22.030	0.397 <sup>bc</sup>
P-value	0.373	0.000	0.000	0.373	0.000	0.373	0.000
SEM	1.048	0.455	0.009	3.301	0.027	0.220	0.002

Means within each column with no common superscript differ significantly at  $P < 0.05$ .

\* CON: control, without supplementation, NS1: supplemented with nano-selenium at 0.1 mg/kg DM of feed, 0.2 mg/kg of feed (NS2), 0.3 mg/kg of feed (NS3), 0.4 mg/kg of feed (NS4), 0.5 mg/kg of feed (NS5).

\*\*Calculated based on Aziz-Mousavi *et al.*, (2015).

The results of the present study are also in accordance with that of Sevcikova *et al.* (2006) and Dlouha *et al.* (2008), since they declared an improvement in body weight due to the selenium dietary supplementation at the level of 0.3 mg/kg. Wang and Xu (2008) also observed an improvement of feed conversion ratio after the dietary supplementation with Se at the level

of 0.2 mg/kg both in the form of sodium selenite and selenium yeast. At the same time, Hu *et al.* (2012) found that average daily gain and gain/feed intake ratio increased linearly and quadratically as the level of nano-selenium increased in the diet from 0.15 to 1.20 mg/kg. On the other hand, Cai *et al.* (2012) indicated no significant differences in weight gain, feed

intake, and feed conversion ratio in broilers fed diets supplemented with 0.3 to 2.0 mg nano-selenium per kg of diet. Similar results have also been shown by Ryu *et al.* (2005) even when the supplemental level was 8 mg/kg.

Breast and drumsticks percentages had higher values in the NS supplemented birds than the control, while adominal fat percentage had lower values in the NS supplemented birds than the control (Table 3). No differences among

the experimental groups were found in the weights of edible organs (liver, heart and gizzard) (Table 3), non edible organs (lungs, kidneys, pancreas, testes, crop, proventriculus, right and left cecum) (Table 4), length, width and diameter of the right and left cecum) (Table 5), and thymus (Table 6). However, significant differences in relative weight of testes were observed between treatments (Table 4).

**Table 3.** Relative weight of carcass components as affected by the different levels of dietary nano-selenium supplementation (% of live body weight)

Group s*	Breast	Drumsticks (thighs)	Wings	Abdominal fat	Liver	Heart	Gizzard
CON	20.904 <sup>b</sup>	11.907 <sup>b</sup>	6.006	1.912 <sup>a</sup>	2.615	0.510	1.743
NS1	22.319 <sup>a</sup>	12.919 <sup>a</sup>	6.316	1.075 <sup>bc</sup>	2.596	0.623	1.858
NS2	22.369 <sup>a</sup>	12.940 <sup>a</sup>	5.722	1.662 <sup>ab</sup>	2.654	0.696	1.946
NS3	22.422 <sup>a</sup>	12.923 <sup>a</sup>	3.254	0.991 <sup>c</sup>	2.324	0.489	1.882
NS4	22.406 <sup>a</sup>	12.911 <sup>a</sup>	5.279	1.182 <sup>bc</sup>	2.788	0.607	1.586
NS5	22.414 <sup>a</sup>	12.935 <sup>a</sup>	5.877	0.968 <sup>c</sup>	2.341	0.606	1.951
P-value	< 0.0001	< 0.0001	0.181	0.015	0.392	0.601	0.956
SEM	0.167	0.036	0.819	0.185	0.172	0.088	0.314

Means within each column with no common superscript differ significantly at  $P < 0.05$ .

\* CON: control, without supplementation, NS1: supplemented with nano-selenium at 0.1 mg/kg DM of feed, 0.2 mg/kg of feed (NS2), 0.3 mg/kg of feed (NS3), 0.4 mg/kg of feed (NS4), 0.5 mg/kg of feed (NS5).

**Table 4.** Relative weight of non edible organs as affected by the different levels of dietary nano-selenium supplementation (% of live body weight)

Groups*	Lungs	Kidneys	Pancreas	Testes	Crop	Proventriculus	Right cecum	Left cecum
CON	0.299	0.516	0.211	0.072 <sup>a</sup>	0.715	0.381	0.296	0.596
NS1	0.261	0.506	0.247	0.054 <sup>b</sup>	0.701	0.412	0.253	0.303
NS2	0.295	0.520	0.238	0.076 <sup>a</sup>	0.494	0.399	0.256	0.269
NS3	0.259	0.631	0.298	0.067 <sup>ab</sup>	0.989	0.400	0.333	0.370
NS4	0.284	0.549	0.201	0.067 <sup>ab</sup>	0.791	0.505	0.275	0.368
NS5	0.259	0.624	0.268	0.060 <sup>ab</sup>	0.462	0.375	0.255	0.437
P-value	0.687	0.726	0.114	0.011	0.749	0.787	0.262	0.282
SEM	0.024	0.075	0.024	0.774	0.268	0.069	0.026	0.097

Means within each column with no common superscript differ significantly at  $P < 0.05$ .

\* CON: control, without supplementation, NS1: supplemented with nano-selenium at 0.1 mg/kg DM of feed, 0.2 mg/kg of feed (NS2), 0.3 mg/kg of feed (NS3), 0.4 mg/kg of feed (NS4), 0.5 mg/kg of feed (NS5).

**Table 5.** Length, width and diameter of right and left cecum as affected by the different levels of dietary nano-selenium supplementation (mm)

Groups*	Right cecum			Left cecum		
	length	width	diameter	length	width	diameter
CON	15.333	7.983	0.193	15.370	3.603	0.217
NS1	17.667	8.527	0.280	16.600	4.027	0.360
NS2	16.333	12.040	0.290	15.837	2.877	0.353
NS3	16.333	9.720	0.420	16.717	6.213	0.283
NS4	17.000	10.810	0.260	16.420	4.587	0.283
NS5	16.333	7.900	0.257	16.277	5.003	0.290
P-value	0.729	0.197	0.155	0.791	0.433	0.593
SEM	1.045	1.265	0.053	0.743	1.136	0.061

Means within each column with no common superscript differ significantly at  $P < 0.05$ .

\* CON: control, without supplementation, NS1: supplemented with nano-selenium at 0.1 mg/kg DM of feed, 0.2 mg/kg of feed (NS2), 0.3 mg/kg of feed (NS3), 0.4 mg/kg of feed (NS4), 0.5 mg/kg of feed (NS5).

Although there is some report about relationship between selenium and gastrointestinal characteristics (Wang *et al.*, 2013) and carcass components (Naik *et al.*, 2015; Konieczka *et al.*, 2015), however scarce literature exists regarding the effect of nano-selenium dietary supplementation on the above parameters in broilers. However, these findings can clarify the effects of nano-selenium on gastrointestinal segment's function and output. Zhang *et al.*, (2016) demonstrated selenium deficiency affects the mRNA expression of inflammatory factors and selenoprotein genes in the kidneys of broiler chicks. Zhang *et al.* (2017) revealed a disbalance of calcium regulation-related genes in broiler hearts induced by selenium deficiency. Cao *et al.* (2017) found an inflammatory response occurs in veins of broiler chickens treated with a selenium deficiency diet. Sevcikova *et al.* (2006) demonstrated no effect of Se dietary supplementation in form of Se-yeast or Se-Chlorella (0.3 mg/kg) on the weights of breast, thigh, liver, giblets and abdominal fat. The results of the present study are in agreement with that of Downs *et al.*, (2000) and Payne and Southern (2005), who showed that carcass traits were not affected by selenium addition (sodium selenite or selenium-enriched yeast) in the diets of broilers. Cai *et al.*, (2012) and Chen *et al.*, (2014) found no effect of selenium addition (nano-selenium or sodium selenite/selenium-enriched yeast, respectively) on the weights of bursa of Fabricius, thymus and spleen. Shirsat *et al.*, (2016) found protective role of biogenic

selenium nanoparticles in immunological and oxidative stress generated by enrofloxacin in broiler chicken. Overall, there is not enough evidence for positive effects of supra concentrations of selenium on broiler immunity (Swain *et al.*, 2000). Dalia *et al.* (2017) found an effect of dietary bacterial organic selenium on growth performance, antioxidant capacity, and selenoproteins gene expression in broiler chickens. Colnago *et al.* (1994) demonstrated high doses of selenium could increase the leukocyte numbers and improve immunity to coccidiosis in chickens. Immunoglobulins contains selenium at disulfide bonds, hence the optimum levels of selenium can develop the immunity functions (Burton *et al.*, 1977).

Glucose and total protein concentrations in blood plasma were not significantly different among the experimental groups (Table 6). Glucose is affected by vitamin E and selenium somewhat, since vitamin E and selenium decrease the cellular oxidative stress, so it preserve the beta cells of liver as glucose regulator. On the other hand, albumin levels in blood were decreased except for NS5 which is due to dehydration during sampling period, and only NS3 and NS5 indicated significant increases compared to the control for anti-Newcastle disease hemagglutination-inhibition titer after the dietary supplementation with the nano-selenium (Table 6). There was no significant difference between other treatments and the control.

**Table 6.** Plasma constituents, relative weight of organ related with immune system, and anti-Newcastle disease hemagglutination-inhibition titers as affected by the different levels of dietary nano selenium supplementation

Groups*	Glucose (mg/dL)	Albumin (g/dL)	Total protein (g/dL)	Thymus (% of LBW)**	ND titer (log 10)***
CON	81.843	1.281 <sup>a</sup>	3.697	0.189	3.333 <sup>b</sup>
NS1	95.490	0.069 <sup>b</sup>	3.503	0.593	3.667 <sup>b</sup>
NS2	52.933	0.102 <sup>b</sup>	4.093	0.633	5.333 <sup>ab</sup>
NS3	125.947	0.090 <sup>b</sup>	3.500	0.170	6.333 <sup>a</sup>
NS4	65.980	0.154 <sup>b</sup>	3.397	1.083	5.333 <sup>ab</sup>
NS5	169.507	1.172 <sup>ab</sup>	2.610	0.169	6.333 <sup>a</sup>
P-value	0.123	0.057	0.414	0.312	0.018
SEM	29.011	0.338	0.467	0.318	0.624

Means within each column with no common superscript differ significantly at  $P < 0.05$ .

\* CON: control, without supplementation, NS1: supplemented with nano-selenium at 0.1 mg/kg DM of feed, 0.2 mg/kg of feed (NS2), 0.3 mg/kg of feed (NS3), 0.4 mg/kg of feed (NS4), 0.5 mg/kg of feed (NS5).

\*\* Relative weight of thymus (% of live body weight); \*\*\* Anti-Newcastle disease hemagglutination-inhibition at 26<sup>th</sup> day of age.

Selenium can improve plasma lipoproteins i.e. decline the LDL-c (low density lipoprotein-cholesterol), cholesterol and plasma triglycerides, and increase HDL-c (high density lipoprotein-cholesterol) cholesterol (Iizuka *et al.*, 2001). Shen and Sevanian (2001) found the selenium deficiency led to glutathione destroy under improvement of macrophage activity, and the other hand it increase the gamma glutamyl cysteine synthetase, hence inhibit glutathione synthetase enzyme. So, the oxidized LDL-c will increase hydroperoxide lipids and aldehydes and then destroy the lipids. LDL-c acetate increases the gamma glutamyl cysteine synthetase, and hence increases glutathione and glutathione peroxidase (Holovska *et al.*, 2003). Meanwhile, selenium deficiency result to free radicals production and there radicals affect on malondialdehyde, and so increase the plasma cholesterol (Kuklinski *et al.*, 1991). Similarly, in a previous study, antibody levels of IgM were increased in groups fed using 0.3-1.0 mg/kg of

nano-selenium and chicks supplemented 0.30 mg/kg of nano-selenium had the highest IgG and IgM titres (Cai *et al.*, 2012). On the other hand, no effect of Se dietary supplementation on the content of blood immunoglobulins was found by Chen *et al.* (2014). These finding can be due higher absorption of nano-particles of selenium against organic/non organic selenium.

### Conclusion

The current study demonstrated that the supplementation of nano-selenium in broiler diets could improve growth performance parameters and immune function, without negative effects on internal organs, carcass parameters and gastrointestinal parts. It is recommend to use higher doses until a negative effect is expected in next studies.

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