

Original Research Article

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Effects of Dietary Supplementation of Inorganic, Organic and Nano Selenium on Antioxidant Status of Giriraja Chicken

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ABSTRACT

An experiment was designed to investigate the effects of supplementing inorganic, organic and nano selenium on growth performance and antioxidant status of Giriraja chicken reared in deep litter system up to eight weeks of age. A total of 384 day old straight run chicks were equally divided into eight treatment groups with four replicates each. Control group (T₁) was fed with basal broiler diet (NRC, 1994) without added selenium supplementation in premix. Experimental groups T₂ and T₃ were given basal diet supplemented with Sodium selenite to contain selenium concentration of 150 ppb and 300 ppb, respectively in the experimental feed. Selenium concentration of 150 ppb and 300 ppb using selenium yeast is added to basal diet to form experimental diet for T₄ and T₅. Experimental groups T₆, T₇ and T₈ were fed on basal diet containing 50ppb, 150ppb and 300ppb selenium supplemented with nano selenium. Standard management and vaccination procedures were followed throughout the study. Weekly cumulative body weight, feed intake and overall cumulative FCR were similar between treatment groups. Antioxidant enzymes GPx, SOD level were increased in liver with selenium supplementation than the control group while catalase activity was improved by 300 ppb selenium supplementation irrespective of source. TBARS value recorded was significantly reduced in 150 ppb and 300 ppb organic and nano selenium supplemented groups.

Keywords

Anti-oxidant, GPx, SOD, CAT, TBARS, Giriraja, Nano selenium, Organic selenium

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Introduction

Selenium (Se) is an essential trace mineral having a number of biologically important roles in poultry such as regulation of antioxidants activities, improvement of immune functions, normal growth and body maintenance (Surai, 2002). Sodium selenite as an inorganic form is the most common source

of selenium used in animal feeds, whereas since 2000, dietary supplementation of organic selenium such as selenium enriched yeast and selenomethionine in poultry diets was legally allowed (FDA, 2000). Usually, the organic forms of Se have higher bioavailability and antioxidant properties than inorganic forms (Wang *et al.*, 2011). In addition, organic forms are less toxic and more environmental friendly

than inorganic forms (Kim and Mahan, 2001) and widely used as feed additive now a days.

Nanoparticles having size in nano scale at least in one dimension is getting importance as animal feed additive (Zhang *et al.*, 2001). But, nano-materials have different physical and chemical characteristics compared to the bulk material (Hartemann *et al.*, 2015). It is reported that nano-materials exhibit novel properties, like a high surface activity, a great specific surface area, a lot of surface active centers, and a high catalytic efficiency (Gao and Hiroshi, 2005). Due to both high surface reactivity and advantage of size effect, nanoparticle has been already used in pharmaceutical applications to increasing the bioavailability of drugs and targeting therapeutic agents to particular organs (Davda and Labhasetwar, 2002). It has been reported that nanoparticles showed new characteristics of transport and uptake and exhibited higher absorption efficiencies than their conventional counterparts (Zha *et al.*, 2008; Liao *et al.*, 2010).

The range between the minimum requirement and maximum tolerable level of Se is narrow. The NRC (1994) recommendations established a minimum level at 0.1 mg/kg of diet for Selenium supplementation for broilers. To ensure feed safety, maximum levels for Se in complete feeds of broilers have been set at 0.5 mg/kg in the European Union (2004). Diseases such as exudative diathesis and pancreatic fibrosis in chicks could be prevented by adequate dietary Se supplementation (Payne and Southern, 2005).

Selenium is an integral part of antioxidant enzyme Glutathione peroxidase (Rotruck *et al.*, 1973) which is having an important biochemical role in removal of free radicals formed as a result of metabolism in tissues. Free radicals formed react with oxygen to form peroxides, which serve as the entry

point to a multitude of reactions, producing numerous by-products and the decomposition of vital molecules and nutrients (Calabotta and Shermer, 1985). Antioxidant enzymes eliminate such free radicals before damage occurs (Sunde, 1994) to the molecules and to the tissues. Cai *et al.*, (2012) reported that dietary supplementation with nano-selenium (NS) at 0.3 to 0.5 mg/kg in broilers significantly improved oxidation resistance of liver and muscles.

Considering these facts, a study was conducted to evaluate the growth performance and antioxidant status of Giriraja birds fed on varying levels of inorganic, organic and nano selenium up to eight weeks.

Materials and Methods

A total of 384 day old straight run Giriraja chicks were procured from Department of Poultry Science, Veterinary College, Bangalore and wing banded, weighed and randomly assigned to eight treatment groups with four replicates having 12 chicks in each group, in a randomized block design.

Giriraja birds are crossbred dual purpose chicken developed for backyard rearing by Department of Poultry Science, Veterinary College, Bangalore which have wide acceptance among farmers in India and in many developing countries. Giriraja birds reared on broiler feed are marketed at eight weeks for meat purpose, while birds fed on chick mash and layer feed or birds reared in backyard system by scavenging are used for egg production.

The chicks were reared in deep litter system with all standard managerial practices till eight weeks of age. Birds were immunized against Newcastle disease on day 7 and 21 and against Infectious Bursal Disease on day 14 and 28, respectively.

Experimental broiler diet in mash form with uniform particle size was formulated as per NRC (1994) specifications with varying selenium concentration as described in Table 1. Chicks were fed with experimental broiler prestarter feed till 21 days followed by experimental broiler starter feed from 22 to 42 days and experimental broiler finisher feed from 43 to 56 days of age. Continuous uniform lighting was ensured in poultry house with natural and artificial lights during entire period of study. All the procedures and methods followed in this study were approved by Institutional Animal Ethics Committee of KVASU, Bidar.

Cumulative body weight and cumulative feed intake was recorded at the end of 3rd, 6th and 8th weeks of age and cumulative feed conversion ratio (FCR) was calculated. Glutathione peroxidase activity (GPx), Superoxide dismutase activity (SOD), Catalase activity (CAT) in liver tissue and Thiobarbituric acid reactive substance (TBARS) value in breast muscle was estimated after slaughter of birds. Procedures are detailed in brief below,

Tissue preparation: Immediately after sacrificing the animals, the tissues were processed for the estimation of activity of antioxidant enzymes as per the method of Bruce and Baudry (1995). Ten per cent liver homogenate was made by homogenising 1 g liver tissue in 10 ml of ice cold 0.05 M phosphate buffer (pH 7.4). The liver homogenate was centrifuged at 1500 g for 1 hour at 4°C and the supernatant obtained was used for the estimation of total proteins, superoxide dismutase, catalase and glutathione peroxidase levels. Protein content of the tissue was determined by the method described by Lowry *et al.*, (1951).

SOD activity: Superoxide dismutase was determined by the method described by Marklund and Marklund (1974). Superoxide

anion is an intermediate in the auto-oxidation of pyrogallol which occurs at pH 8.2. The ability of SOD to inhibit the auto-oxidation of pyrogallol at pH 8.2 provides the basis for enzymatic activity. To 0.5 ml of tissue homogenate, 0.25 ml of ethanol and 0.15 ml of chloroform were added and mechanically shaken for 15 min. Then the contents were centrifuged at 13,000 g for 15 min at 4 °C. The supernatant was carefully separated and used for the test. OD value of assay mixture consisted of 2 ml of 0.1M TrisHCl, 0.5 ml of homogenate, 1.5 ml of distilled water and 0.5 ml of pyrogallol was taken for 3 min at 420 nm wavelength.

GPx activity: Glutathione peroxidase was determined by the method described by Rotruck *et al.*, (1973). About 0.5 ml of 0.4 M phosphate buffer was taken and 0.1 ml of sodium azide, 0.2 ml of 4 mM GSH, 0.5 ml tissue homogenate, 0.1 ml of 30 mM H₂O₂ were added and the volume was made up to 2 ml with distilled water. It was incubated for 10 min in room temperature and the reaction was terminated by the addition of 0.5 ml 10 percent TCA and centrifuged at 4 °C for 10 min at 1500 rpm. To determine the residual glutathione content, the supernatant was removed and to this 4 ml of 0.3 M disodium hydrogen phosphate and 1 ml dithiobis-nitrobenzoic acid (DTNB) (0.004%) reagent were added. The color developed was read at 412 nm against the reagent blank containing only phosphate solution and DTNB reagent.

CAT activity: Catalase was estimated by the method of Caliborne (1985). Catalase activity was determined by monitoring the decrease in absorbance spectrophotometrically at 240 nm due to decomposition of H₂O₂. The difference in extinction coefficient per unit time was a measure of the catalase activity. To 0.2 ml of homogenate, 1 ml of 30 mM H₂O₂ was added and the OD value was taken at 240 nm at an interval of 1 min for 3 min. Blank used

contained 0.2 ml of distilled water plus 1 ml of 30 mM H₂O₂.

TBARS: Thiobarbituric acid reactive substances (TBARS) value was estimated as per Witte *et al.*, (1970) with slight modifications. Ten gram of sample was triturated with 25 mL 20% trichloroacetic acid (TCA) in 2 M orthophosphoric acid solution for 2 min. The content was taken into beaker and rinsed with 25 mL of water, mixed well and filtered through Whatman No. 1 filter paper. Three millilitres of TCA filtrate was mixed with equal volume of thiobarbituric acid (TBA) reagent (0.005 M) and stored for 16 hours in adark room at 27⁰C. Optical density (OD) value was recorded at 532 nm wavelength. TBARS value was calculated as mg malondialdehyde /kg of sample by multiplying OD with K factor (5.2).

Statistical analysis: To determine the statistical significance, ANOVA technique described by Snedecor and Cochran (1994) was used. Differences between the means were tested using Duncan's Multiple Range Test (Duncan, 1995) at 5 per cent confidence level.

Results and Discussion

Body weight

Supplementation of Selenium as SS, SY and NS did not influence cumulative body weight between treatments and control group on day 21, 42 and 56 of the study (Table 2). The results obtained in this study was in agreement with the results of Srimongkol *et al.*, (2004), Niu *et al.*, (2009), Da Silva *et al.*, (2010), Cai *et al.*, (2012), Tayeb and Quader (2012), Rao *et al.*, (2013), Chen *et al.*, (2014), Habibian *et al.*, (2014) and Dalia *et al.*, (2017) who observed no significant change in body weight of chicken due to dietary supplementation of organic selenium comparison to inorganic selenium. Similarly, supplementation of nano

selenium did not produce any significant difference in cumulative body weight of Giriraja birds which was in agreement with Moghaddam *et al.*, (2017), Gangadoo *et al.*, (2018) and Li *et al.*, (2018) who reported comparable body weight for broiler birds supplemented with organic and nano selenium in feed.

In contradictory to the results obtained in the present study, Ozkan *et al.*, (2007), Upton *et al.*, (2008), Yang *et al.*, (2012), Rajashree *et al.*, (2014), Selim *et al.*, (2015), Xu *et al.*, (2015), Wang *et al.*, (2016), Ravindran and Elliott (2017) and Zia *et al.*, (2017) reported increased body weight for broilers supplemented with organic source of selenium compared to inorganic source.

Zhou and Wang (2011), Bagheri *et al.*, (2015), Mahmoud *et al.*, (2016) and Bakhshalinejad *et al.*, (2018) noticed higher body weight gain in chicken supplemented with nano selenium compared to other sources.

Feed intake

As shown in Table 3, cumulative feed intake on day 21, 42 and 56 of the experiment was not affected by selenium supplementation. Results in the experiment was in agreement with the results published by Niu *et al.*, (2009), Da Silva *et al.*, (2010), Cai *et al.*, (2012), Tayeb and Quader (2012), Chen *et al.*, (2014), Habibian *et al.*, (2014), Gogmen *et al.*, (2016), Dalia *et al.*, (2017), Moghaddam *et al.*, (2017) and Li *et al.*, (2018) who observed no significant difference in feed intake of chicken due to selenium supplementation.

Higher feed intake in dietary selenium supplemented groups were reported by Yang *et al.*, (2012), Rajashree *et al.*, (2014), Bagheri *et al.*, (2015), Ravindran and Elliott (2017) and Zia *et al.*, (2017) which was in opposition to the present study.

Feed conversion ratio

Supplementation of selenium did not affect cumulative FCR in Giriraja birds (Table 4) which was in agreement with the study conducted by Tayeb and Quader (2012) and Da Silva *et al.*, (2010) who reported that the dietary supplementation of inorganic and organic sources of selenium at varying levels resulted in no significant differences in feed conversion ratio in broiler chicken. Similarly research conducted by utilising various levels of sodium selenite and organic selenium by Rao *et al.*, 2013 (0, 0.1, 0.2, 0.3 and 0.4 mg/kg diet), Rajashree *et al.*, 2014 (0.25 and 0.5 mg/kg), Dalia *et al.*, 2017 (0.3 mg/kg) and Ravindran and Elliott, 2017 (0.3 mg/kg) reported no significant difference in FCR between treatment groups. Habibian *et al.*, (2014) reported similar FCR for broiler birds supplemented with 0, 0.5 and 1 mg/kg Se in feed reared in thermo neutral and heat stressed environment.

Study on FCR in broilers as influenced by nano selenium supplementation at 0.0, 0.3, 0.5, 1.0, or 2.0 mg/kg to the basal diet (Cai *et al.*, 2012) revealed no significant difference between treatments. Moghaddam *et al.*, (2017) and Li *et al.*, (2018) supplemented 0.3 mg Se/kg as organic selenium and nano selenium in chicken feed and observed no significant difference in FCR.

Against the current study, improved FCR was reported in earlier experiments by supplementing inorganic and organic selenium to basal diet at 0.05, 0.15 and 0.25 mg/kg level (Wang *et al.*, 2016) and at 0.3 mg/kg level (Zia *et al.*, 2017) to the chicken feed. Similarly, Roch *et al.*, (2000), Swain *et al.*, (2000), Choct *et al.*, (2004), Naylor *et al.*, (2009), Niu *et al.*, (2009) reported improved FCR of broilers supplemented with organic source of selenium than inorganic selenium supplemented groups and control group.

Earlier studies reported improved FCR in broiler chicken supplemented with nano selenium supplemented at 0.3 mg/kg diet (Zhou and Wang, 2011) and at 0.5 mg/kg (Bagheri *et al.*, 2015) level in feed. Selim *et al.*, (2015) revealed better FCR for organic Se (0.15, 0.3 mg/kg) and nano Se (0.3 mg/kg) supplemented groups than 0.15 mg/kg inorganic selenium supplemented group.

Mahmoud *et al.*, (2016) noted improved FCR in heat stressed birds by supplementing nano selenium at 0.3 mg/kg level in feed, while in another study by Safdari-Rostamabad *et al.*, (2017) supplementation of 1.2 mg/kg Nano selenium alleviated the adverse effects of heat shock on the FCR of heat stressed-broilers. In the present study, birds were not subjected to any additional stress condition which may resulted in similar FCR in Giriraja birds.

Antioxidant status of birds

Effect of supplementation of sodium selenite, selenium yeast and nano selenium on antioxidant status of birds measured through activity of SOD, GPx, CAT and TBARS value (Table 5) was differed significantly ($P \leq 0.05$) between treatments. Activity of SOD was significantly ($P \leq 0.05$) higher in all selenium enriched groups than the non-supplemented control group.

Activity of SOD was similar among inorganic, organic and nano selenium supplemented groups. GPx activity was significantly ($P \leq 0.05$) improved by supplementation of sodium selenite at 300 ppb level, selenium yeast and nano selenium at 150 and 300 ppb level than control group. Activity of catalase enzyme was significantly increased in 300ppb sodium selenite, selenium yeast and nano selenium supplemented groups. TBARS value was decreased significantly ($P \leq 0.05$) by supplementation of selenium yeast and nano selenium at 150 and 300 ppb level.

Table.1 Description of the experimental groups, sources and levels of selenium supplemented

Experiment group	Experiment diet	Levels of selenium
T ₁	Control	Basal diet
T ₂	SS150	Basal diet + SS
T ₃	SS300	Basal diet + SS
T ₄	SY150	Basal diet + SY
T ₅	SY300	Basal diet + SY
T ₆	NS50	Basal diet + NS
T ₇	NS150	Basal diet + NS
T ₈	NS300	Basal diet + NS

SS- Sodium Selenite, SY- Selenium Yeast, NS- Nano Selenium

Table.2 Effect of supplementation of SS, SY and NS on weekly cumulative body weight (g) of Giriraja chicken

Treatment groups		Mean body weight (g)			
		Day 1	Day 21	Day 42	Day 56
T ₁	Control	40.36±0.81	335.36±6.21	991.52±9.97	1445.25±15.70
T ₂	SS150	40.39±0.39	336.65±6.71	984.70±9.85	1453.64±25.36
T ₃	SS300	40.09±0.30	345.26±4.71	1007.58±18.05	1472.78±24.04
T ₄	SY150	40.06±0.66	356.60±6.43	1027.64±18.17	1509.85±30.32
T ₅	SY300	39.80±0.23	356.34±3.51	1038.59±14.58	1528.06±18.34
T ₆	NS50	39.46±0.70	334.79±7.57	988.64±10.85	1453.67±25.72
T ₇	NS150	39.67±0.84	351.75±8.53	1019.72±20.38	1483.77±38.39
T ₈	NS300	39.97±0.15	352.34±4.80	1039.92±12.43	1507.36±13.66

Table.3 Effect of supplementation of SS, SY and NS on cumulative feed intake (g) of Giriraja chicken

Treatment groups		Cumulative feed intake (g)		
		Day 21	Day 42	Day 56
T ₁	Control	494.08±5.28	1859.13±3.24	3056.00±1.48
T ₂	SS150	477.63±11.35	1854.56±3.58	3052.40±1.68
T ₃	SS300	490.56±9.47	1853.69±2.03	3051.35±1.56
T ₄	SY150	485.23±7.96	1858.31±2.81	3052.94±1.27
T ₅	SY300	489.71±6.77	1858.79±2.08	3052.92±1.94
T ₆	NS50	500.98±3.00	1859.63±0.92	3053.54±1.28
T ₇	NS150	493.54±9.72	1856.02±2.69	3052.31±2.66
T ₈	NS300	479.33±11.52	1858.06±3.08	3052.65±2.57

Table.4 Effect of supplementation of SS, SY and NS on cumulative feed conversion ratio of Giriraja chicken

Treatment groups		Cumulative feed conversion ratio		
		Day 21	Day 42	Day 56
T ₁	Control	1.68±0.05	1.96±0.02	2.18±0.02
T ₂	SS150	1.62±0.06	1.97±0.02	2.17±0.04
T ₃	SS300	1.61±0.03	1.92±0.03	2.13±0.04
T ₄	SY150	1.54±0.04	1.88±0.03	2.08±0.04
T ₅	SY300	1.55±0.03	1.86±0.03	2.05±0.02
T ₆	NS50	1.70±0.04	1.96±0.02	2.16±0.04
T ₇	NS150	1.59±0.05	1.90±0.04	2.12±0.05
T ₈	NS300	1.54±0.04	1.87±0.02	2.08±0.02

Table.5 Effect of supplementation of SS, SY and NS on Antioxidant status of bird

Treatment groups		Liver			Breast Muscle
		SOD	GPx	CAT	TBARS
T ₁	Control	17.45±0.29 ^b	39.99±1.90 ^b	8.41±0.95 ^b	0.99±0.10 ^a
T ₂	SS150	18.53±0.57 ^a	43.92±0.62 ^{ab}	9.56±0.39 ^{ab}	0.95±0.11 ^a
T ₃	SS300	19.32±0.34 ^a	47.12±2.02 ^a	11.52±0.46 ^a	0.81±0.11 ^{ab}
T ₄	SY150	19.24±0.53 ^a	46.54±3.77 ^a	10.41±0.56 ^{ab}	0.59±0.11 ^b
T ₅	SY300	19.45±0.34 ^a	48.17±1.84 ^a	11.67±0.42 ^a	0.56±0.07 ^b
T ₆	NS50	19.36±0.29 ^a	44.27±0.57 ^{ab}	10.50±1.10 ^{ab}	0.89±0.04 ^{ab}
T ₇	NS150	19.38±0.13 ^a	49.47±2.11 ^a	10.65±0.42 ^{ab}	0.56±0.16 ^b
T ₈	NS300	19.24±0.29 ^a	48.67±2.07 ^a	11.91±1.17 ^a	0.55±0.14 ^b

Units: SOD: U/min/mg of protein, GPx: μ moles of Glutathione utilised/min/mg of protein, CAT: μ moles of H₂O₂/min/mg of protein, TBARS: g/Kg MDA

Selenium supplementation as sodium selenite, selenium yeast and nano selenium resulted in increased activity of SOD, GPx and CAT than non-supplemented control group. Enzyme activity was higher in all 300 ppb selenium enriched feed supplemented groups irrespective of source. Selenium supplementation at 50 and 150 ppm concentration resulted in comparable antioxidant enzyme activity in sodium selenite, selenium yeast and nano selenium supplemented groups. Increased activity of SOD, GPx and CAT was consistent with decreased MDA concentration (TBARS value) in treatment groups which was in agreement with Rao *et al.*, (2013) who stated

decreased Lipid peroxidation in plasma along with linearly increased activities of glutathione peroxidase and catalase in blood.

In agreement with the current study, Dalia *et al.*, (2017) reported significant elevation in liver GPx and CAT activity in selenium supplemented treatment groups, with a substantial reduction in malondialdehyde concentration. But enzyme activity was similar between inorganic and organic selenium supplemented groups which were in line with the current study. Earlier studies by Wang *et al.*, (2011), Ozkan *et al.*, (2007), Jiang *et al.*, (2009), Zoidis *et al.*, (2010), Upton *et al.*, (2009), Chen *et al.*, (2013),

Rajashree *et al.*, (2014), Edens and Sefton (2016), Gogmen *et al.*, (2016) and Bakhshalinejad *et al.*, (2018) observed increased glutathione peroxidase activity in various tissues of broiler such as liver, breast muscle, blood and serum as influenced by supplementation of organic sources of selenium.

Chen *et al.*, (2014) noted significantly higher liver GPx activity in selenium yeast supplemented group than that sodium selenite supplemented group, while Wang *et al.*, (2011), in line with current study, observed similar activity in these two groups. In contrary, Edens, (2002) reported a steady state release of selenium from organic selenium for incorporation into the glutathione peroxidase antioxidant system which resulted in increased GPx activity. Similarly, Yang *et al.*, (2012) stated 155.83% higher serum glutathione peroxidase activity in 0.3 mg/kg organic selenium supplemented 42 day old broiler chicken than that in 0.3 mg/kg inorganic selenium supplemented group.

Wang *et al.*, (2009) stated that the GPx activity influences the oxidation state of myofibrillar protein and reduce the drip loss by improving the cell membranes integrity. The meat quality is influenced by pre slaughter GPx concentration in live animals and by anti-oxidative stability of carcass (Renerre *et al.*, 1996) which can be effected by selenium supplementation (Liao *et al.*, 2012). Correspondingly, reduced drip loss was noticed in selenium supplemented groups having increased GPx concentration in the current study.

Wang *et al.*, (2011) and Chen *et al.*, (2013) noted significant increase in SOD activity with dietary supplementation of selenium. Yang *et al.*, (2012), Dalia *et al.*, (2017) and Bakhshalinejad *et al.*, (2018) stated that

organic and inorganic source of selenium had no obvious effect on SOD which was opposed by Chen *et al.*, (2014) who observed increased SOD activity in organic selenium supplemented group. Broiler chicken fed on selenium deficient diet in an experiment conducted by Cao *et al.*, (2016) revealed significantly reduced SOD and CAT activity in the arteries and veins. But CAT activity reported by Ozkan *et al.*, (2007) was similar between selenium supplemented groups and control.

Mahmoud *et al.*, (2016) revealed higher GPx, SOD and catalase activity in birds supplemented with nano selenium. In a trial conducted by Cai *et al.*, (2012) by supplementing nano selenium to at 0.3 mg/kg level showed increased glutathione peroxidase activity compared with the birds receiving 0 and 2.0 mg/kg of nano selenium and was similar to the birds supplemented with 0.3, 0.5, and 1.0 mg/ kg of nano-Selenium. Supplementation of various sources of selenium to chicken, by Mohapatra *et al.*, (2014) revealed higher GPx activity than non-supplemented group and nano selenium improved GPx and SOD activity than sodium selenite supplemented groups. Similarly, Safdari-Rostamabad *et al.*, (2017), Qu *et al.*, (2017) and Li *et al.*, (2018) observed linearly increased the activity of glutathione peroxidase by dietary supplementation of nano selenium. Contrary to the result obtained for nano selenium supplemented group Li *et al.*, (2018) noticed no significant change in activity of SOD and catalase enzyme between treatments and control.

In accordance with the results obtained many studies conducted earlier revealed significant reduction in TBARS value in terms of malondialdehyde concentration in breast meat as influenced by selenium supplementation. Edens and Sefton (2016), Dalia *et al.*, (2017) and Bakhshalinejad *et al.*, (2018) obtained

significantly lesser malondialdehyde concentration for Selenium supplemented group than negative control group. Significant decrease in TBARS value as influenced by selenium yeast than sodium selenite was reported by Sevcikova *et al.*, (2006), Jokic *et al.*, (2009), Hosseini-Mansoub (2011), Taulescu *et al.*, (2011), Wang *et al.*, (2011), Boiago *et al.*, (2014), Chen *et al.*, (2014), Delles *et al.*, (2014), Salami *et al.*, (2015) and Ravindran and Elliott (2017). Significantly decreased TBARS value was recorded at 0.15mg/kg selenium supplementation by Hooge (2007) while TBARS value noted for 0.3 mg/kg organic selenium was similar to control group. While, Ozkan *et al.*, (2007) and Kralik *et al.*, (2013) did not observe any change in TBARS value due to selenium supplementation.

Earlier studies using nano selenium was also in agreement with current trial results. A similar study conducted by Visha *et al.*, (2017) and Li *et al.*, (2018) reported that dietary supplementation of selenium yeast or selenomethionine and nano selenium significantly decrease the concentration of malondialdehyde in serum compared with the sodium selenite group. Cai *et al.*, (2012) and Moghaddam *et al.*, (2017) conducted a study by supplementing nano selenium as a feed supplement in broiler chicken and reported significant reduction in MDA concentration in liver compared to the control.

Feeding nano selenium to broilers reared at high ambient temperature (35°C) significantly alleviated the negative effects of heat stress indicated by reduced level of the malondialdehyde content in liver and breast muscle (Mahmoud *et al.*, 2016). Contrary to these studies, Selim *et al.*, (2015) stated that liver and thigh muscle MDA concentration is not affected by inclusion of organic, inorganic and nano forms of selenium in broiler feed at 0.15 and 0.30 mg/Kg level.

Supplementation of SS, SY and NS up to 300 ppb level did not influence body weight, feed intake and FCR of Giriraja dual purpose chicken reared in deep litter system. Higher levels of selenium (300 ppb) irrespective source improved liver antioxidant enzyme level. Selenium yeast and nano selenium at 150 ppb and 300 ppb level in feed significantly reduced MDA concentration in breast muscle.

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