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Effects of Different Sources and Levels of Selenium on Performance, Thyroid Function and Antioxidant Status in Stressed Broiler Chickens

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Abstract: The effects of adding different sources and levels of selenium on performance, thyroid function and antioxidant status in stressed broiler chickens were evaluated. Stress was induced by supplementation with 20 mg/kg Corticosterone (CORT) in basal diet. Total 144 Avian broilers were used for 2 weeks from 14 days of age, which were randomly distributed to 6 groups. One of these groups was fed with basal diets and the remaining with basal diet supplemented with CORT. Meanwhile, diets used in CORT groups were supplemented with 0, 0.1 or 0.4 mg/kg sodium selenite or yeast selenium, respectively. The results showed that dietary CORT resulted in significant suppression of growth and thyroid function, increase in creatine kinase activity and uric acid concentration in serum, indicating that CORT induced stress of broilers. In addition, the balance between pro-oxidants and antioxidants was disturbed by CORT administration, resulting in oxidative injury in the body. Supplementation of dietary selenium improved feed efficiency, promoted conversion of thyroxine (T4) to triiodothyronine (T3), minimized the changes of blood biochemical parameters; furthermore, it elevated antioxidases activities and decreased lipid peroxidation products in stressed broilers. The effects of adding 0.4 mg/kg selenium were better than 0.1 mg/kg. Although there was no significant difference between two selenium sources, it was conclude that high level of inorganic or organic selenium was able to attenuate stress and oxidative injury due to exogenous CORT administration, especially organic yeast selenium.

Key words: Selenium, corticosterone, broiler, stress, oxidative injury

INTRODUCTION

Selenium has been considered as an essential trace element that plays important roles in growth, immune function, productivity and anti-stress. It is well known that the form of supplemental selenium includes inorganic and organic selenium and the recommendation concentration of selenium in broiler diets is 0.15 mg/kg (NRC, 1994). The majority of pervious studies investigated the effects of adding different selenium source in broiler chickens just in normal condition. However, in modern large-scale poultry production, there are many factors, such as temperature, density, immune stimulation, transport and feed quality *et al.*, which could cause stress of broiler chickens easily, resulting in growth inhibition and oxidative injury in the body.

In stress condition, selenium requirements of broilers may change, whereas, the effects of different selenium sources in stressed broiler are not clear yet. Recently, some studies reported that exogenous Corticosterone (CORT) administration would significantly increase CORT concentration in blood and induce stress or oxidative stress in broiler chickens (Taniguchi *et al.*, 1997; Eid *et al.*, 2003; Lin *et al.*, 2004).

In the present study, the objective was to evaluate the effects of different sources and levels of selenium on performance, thyroid function and antioxidant status in stressed broiler chickens induced by dietary corticosterone administration.

MATERIALS AND METHODS

Animals and management: One hundred and forty-four 14-day old Avian broiler chickens were used in this experiment. All chickens kept in pens were given feed and water *ad libitum* with 24 h light and acquired immunity according to usual ways. This study was approved by the Committee of Sichuan Agricultural University for Animal Care and Use.

Experimental design and diets: Chickens were randomly allotted into 6 treatments with 3 replicates of 8 birds each. Briefly, the control group (Group I) was given the basal diet; the stressed group (Group II) was given the basal diet supplemented with 20 mg/kg corticosterone (Sigma, = 92%, USA); the groups III, IV, V and IV were fed the diets with same content of corticosterone and 0.1 or 0.4 mg/kg sodium selenite or yeast selenium, respectively (Table 1). The basal diet was formulated according to nutrient requirements of poultry (NRC, 1994) (Table 2). The experiment lasted 14 days.

Sample collection and analysis: The feed intake of chickens was measured weekly. At the end of the experiment, chickens in each pen were weighed as a group and blood samples were collected from win vein of 2 birds of each replicate. The serum was separated by centrifugation at 3000 rpm for 10 min. Supernatant

Table 1: Experimental design

Items	Group I (Control)	II	III	IV	V	VI
Se level (mg/kg)	0	0	0.1	0.4	0.1	0.4
Se source			Na_2SeO_3	Na_2SeO_3	Yeast-Se	Yeast-Se
Corticosterone	-	+	+	+	+	+

Note: "-" denotes no supplementation of corticosterone with the basal diet; "+" denotes supplementation of that

Table 2: Composition and nutrient levels of the basal diet

Ingredients (%)		Nutrient levels	
Maize	60.00	Metabolic energy (Mcal/kg)	3.02
Soybean meal	33.05	Crude protein (%)[b]	19.14
Rapeseed oil	3.20	Lysine (%)	1.01
Calcium carbonate	1.39	Methionine (%)	0.43
Dicalcium phosphate	1.26	Methionine + cystine (%)	0.70
DL-methionine	0.15	Calcium (%)	0.90
Salt	0.3	Available phosphonium (%)	0.35
Choline chloride	0.15	Selenium (mg/kg)[b]	0.04
Premix[a]	0.5		

(a) Provided per kilogram of complete feed: vitamin A 18 000IU, vitamin D_3 4 000IU, vitamin E 40IU, vitamin K_3 4.8 mg, vitamin B_1 4 mg, vitamin B_2 12 mg, vitamin B_6 7.2 mg, vitamin B_{12} 0.032 mg, niacin 60 mg, Ca-pantothenic acid 20 mg, folic acid 2 mg, biotin 0.16 mg and copper 8 mg, zinc 40 mg, iron 80 mg, manganese 60 mg, iodine 0.35 mg, (b) Analysis data

was gathered into Eppendorf tubes respectively and then immediately stored at -20°C until analysis.

The concentration of 3,5,3'-triiodothyronine (T3), thyroxine (T4) and Corticosterone (CORT) in serum were determined by Radioimmune Assay (RIA) and Enzyme-linked Immunosorbent Assay (ELISA) using commercial kits (3V Bioengineering group Co., Ltd, Shandong, China and Rapidbio, CA, USA), respectively.

Activities of Gluthathione Peroxidase (GPX), Superoxide Dismutase (SOD), Catalase (CAT), Alanine Aminotransferase (ALT) and Creatine Kinase (CK) and content of Malondialdehyde (MDA), Total Antioxidant Capacity (T-AOC), Glucose (GLU) and Uric Acid (UA) in serum were dertermined by standard assay kits (Jiancheng Bioengineering Institute, Nanjing, China).

Statistical analysis: Data was analyzed by one-way ANOVA and mean values were compared by multiple range test (Duncan) with SPSS 13.0. Statistical significance was accepted at $p<0.05$.

RESULTS

Performance and serum corticosterone concentration: The growth performance and serum corticosterone concentration of broilers were shown in Table 3. Dietary CORT significantly decreased ADG ($p<0.01$) and increased G:F ($p<0.01$), whereas supplementation of different levels of selenium improved daily gains and feed conversion ratio in broilers and the effect of 0.4 mg/kg Yeast-Se was the best. But there were no differences between selenium sources. The different dietary treatment also did not affect ADFI among all groups.

As for serum corticosterone concentration, CORT diet group was significantly higher than the control group ($p<0.05$) and supplementation of selenium minimized this tendency. Meanwhile, the concentrations declined as selenium level increased, although still higher than the control group ($p>0.05$) and Yeast-Se was slightly better than Na_2SeO_3 ($p>0.05$).

Thyroid hormone: In Table 4, the concentrations of thyroid hormone in serum of broilers were presented. Compared with the control group, dietary CORT treatments decreased T3 ($p<0.01$), T4 ($p>0.05$) and T3:T4 ($p>0.05$). However, as selenium level increased, T3 and T3:T4 increased obviously and T4 trended to decrease. There was no significant difference in selenium sources; however, 0.4 mg/kg Yeast-Se group had higher T3 and T3:T4 than the other CORT groups.

Serum biochemical parameters: In Table 5, biochemical parameters in serum of broilers were shown. The activities of ALT ($p>0.05$) and CK ($p<0.05$) in the CORT groups were higher than those in the control group, so was UA content ($p<0.05$). Supplementation of different sources and levels of selenium decreased those parameters, especially 0.4 mg/kg Yeast-Se group ($p<0.05$), compared with CORT group. There was no difference in the concentration of GLU among all groups, but GLU trended to increase in broilers consuming CORT diet without selenium.

Antioxidant status: The effect of adding selenium on antioxidant status of broilers was shown in Table 6. The activities of GPX, SOD and T-AOC in serum were reduced by dietary CORT treatment and increased by adding selenium. Their activities in broilers consuming CORT diets supplemented 0.4 mg/kg Se were highest in all groups, whereas Yeast-Se was superior to Na_2SeO_3 ($p>0.05$). There was no difference in serum CAT activity among all groups. Compared with the

Table 3: Effect of adding selenium on performance and serum corticosterone concentration of stressed broilers

Items	Group I	II	III	IV	V	VI	SEM
Average Daily Gains (ADG) (g)	41.25^{A}	23.23^{B}	24.03^{B}	25.22^{B}	24.70^{B}	25.46^{B}	1.09
Average Daily Feed Intake (ADFI) (g)	85.79	83.30	81.89	83.02	82.14	81.93	1.80
Gain:Feed ratio (G:F)	2.08^{Aa}	3.60^{Bc}	3.41^{Bbc}	3.29^{Bbc}	3.33^{Bbc}	3.22^{Bb}	0.16
Corticosterone (nmol/l)	9.48^{a}	16.90^{b}	14.45^{ab}	12.61^{ab}	13.06^{ab}	11.93^{ab}	2.66

Mean values in a row with different superscript small letter are significant difference (p<0.05), those with the different superscript capital letter are significant difference (p<0.01)

Table 4: Effect of adding selenium on serum thyroid hormone of stressed broilers

Items	Group I	II	III	IV	V	VI	SEM
T3 (ng/ml)	0.80^{De}	0.61^{Aa}	0.64^{ABab}	0.70^{BCcd}	0.67^{ABbc}	0.75^{CDd}	0.02
T4 (ng/ml)	41.97^{Bb}	35.25^{ABab}	32.43^{ABa}	29.96^{Aa}	31.99^{ABa}	29.30^{Aa}	3.38
T3:T4 ratio	0.019^{ABab}	0.018^{Aa}	0.020^{ABab}	0.023^{ABbc}	0.021^{ABabc}	0.026^{Bc}	0.002

Mean values in a row with different superscript small letter are significant difference (p<0.05), those with the different superscript capital letter are significant difference (p<0.01)

Table 5: Effect of adding selenium on biochemical indices in serum of stressed broilers

Items	GroupI	II	III	IV	V	VI	SEM
ALT (IU/l)	2.67^{ABbc}	3.67^{Bc}	2.00^{ABab}	2.00^{ABab}	1.33^{Aab}	1.00^{Aa}	0.66
GLU (mmol/l)	13.01	14.36	13.20	13.01	12.51	13.75	0.85
UA (ìmol/l)	170.00^{a}	236.67^{b}	212.33^{ab}	185.57^{ab}	210.00^{ab}	168.67^{a}	23.96
CK (U/ml)	1.60^{a}	2.65^{b}	2.12^{ab}	1.80^{ab}	1.98^{ab}	1.66^{a}	0.38

Mean values in a row with different superscript small letter are significant difference (p<0.05), those with the different superscript capital letter are significant difference (p<0.01)

Table 6: Effect of adding selenium on antioxidant status in serum of stressed broilers

Items	Group I	II	III	IV	V	VI	SEM
GPX (U)	533.93^{Aa}	432.83^{ABa}	729.77^{BCb}	959.51^{CDc}	747.20^{BCb}	1018.80^{Dc}	82.20
SOD (U/ml)	63.73^{ABa}	55.86^{Aa}	84.73^{BCb}	98.44^{CDbc}	91.82^{CDb}	113.17^{Dc}	8.63
CAT (U/ml)	3.72	3.20	3.57	3.74	3.54	4.14	1.21
T-AOC (U/ml)	8.43^{ab}	6.94^{a}	12.25^{abc}	14.71^{bc}	12.55^{abc}	15.51^{c}	2.89
MDA (nmol/ml)	7.35^{a}	15.23^{b}	10.38^{ab}	8.59^{a}	9.55^{ab}	8.43^{a}	2.54

Mean values in a row with different superscript small letter are significant difference (p<0.05), those with the different superscript capital letter are significant difference (p<0.01)

control group, the content of MDA increased (p<0.05) in broilers consuming CORT diet, but alleviated by supplementation with selenium, especially at 0.4 mg/kg (p<0.05). The source of selenium did not affect MDA (p>0.05).

DISCUSSION

The elevation of blood corticosterone concentration could be considered as the signal of suffered stress (Moberg and Mench, 2000). In present study, it was clear that the corticosterone concentration in serum significantly elevated; meanwhile, the growth performance significantly declined, reflected in decrease in daily gains and feed efficiency in broilers consuming CORT diet, indicating that dietary CORT induced stress of birds, in accordance with the results of previous studies (Taniguchi *et al.*, 1997; Eid *et al.*, 2003; Lin *et al.*, 2004; Virden *et al.*, 2007). In addition, dietary CORT disturbed secretion of thyroid hormone, causing decrease in serum T3, T4 and T3:T4 ratio, similar to the studies of Darras *et al.* (1996) and Sui (2005). Thyroid hormone could be responsible for anabolic metabolism of nutrients in animal growth and He *et al.* (2000) demonstrated that selenium improved growth of broilers via thyroid hormone status. Stress induced by CORT suppressed thyroid function, then decreased muscle protein synthesis and enhanced proteolysis, resulting in growth inhibition of broilers eventually. However, as adding level of selenium elevated, feed conversion rate of stressed broilers trended to improve, indicating that high level of dietary selenium could partly counteract growth inhibition caused by stress. Our results were similar to the study of piglets suffered oxidative stress induced by diquat administration (Yuan, 2007). This role of selenium might be partly associated with thyroid hormone, because high level of selenium could be an inhibitor of corticosterone secretion to reduce the damage of thyroid function. Meanwhile, selenoproteins in thyroid, such as glutathione peroxidase and thioredoxin reductase, were able to remove excessive H_2O_2 generated from thyroid follicle by stress, so as to maintain the integrity of the structure and function of thyroid (Ekholm and Bjorkman, 1997). Moreover, liver 5' deiodinase I, as another selenoprotein, was responsible for conversion of T4-T3 in the body. Supplementation of selenium promoted production of biologically active T3 by elevating deiodinase I activity (Arthur *et al*., 1990).

The serum creatine kinase activity would rise when damage of skeletal muscle and heart tissue and destruction of the membrane barrier by various factors. In present study, the CK activity significantly increased in CORT treatment group, in line with the reports of Malherios *et al*. (2003) and Lin *et al*. (2004). However, it decreased obviously in stressed broilers as adding level of selenium increased; indicating high level of selenium was able to alleviate tissue injury due to stress. Uric acid was the end product of protein metabolism and also considered as a non-enzymatic antioxidant in birds, especially under chronic stress condition (Watanabe *et al*., 1997). In present study, the UA concentration in serum was increased by dietary CORT, whereas decreased by supplied with selenium. Selenium could enhance enzymatic antioxidants to improve antioxidant capacity in body and meanwhile protect the muscle cell membrane to inhibit protein catabolism, bringing about decrease in UA together, which benefited the recovery of growth of stressed broilers. In addition, we also observed the increase in ALT activity in stressed broilers, suggesting possible liver injure. After adding selenium, the decrease in ALT activity indicated that selenium might be involved in protection of liver cell and metabolism of amino acids, but which needs further study. It was surprised that the level of GLU in serum did not rise, possibly due to relative later time of blood collection and the different dose of dietary CORT, compared with some studies (Sui, 2005; Jiang, 2007).

In addition, the significant increase in serum concentration of MDA in chickens consuming CORT diet, indicating oxidative stress, in line with pervious reports (Ohtsuka *et al*., 1998; Eid *et al*., 2003; Lin *et al*., 2004). Accompanied by an increase in lipid peroxidation products, the activities of GPX, SOD and CAT and T-AOC in serum declined in CORT treatment group since augmented formation of Reactive Oxygen Species (ROS) counteracted plenty of antioxidases in the body, however, which was changed by supplementation of selenium. GPX, the active site of which is incorporated by selenium via selenocysteine, together with SOD and CAT constitute the enzymatic antioxidant system of the body, against toxic to hydrogen peroxide, eliminating free radical and protecting cell membrane. High level of selenium enhanced antioxidant capacity and relieved oxidative stress, reflected in elevation of antioxidases activities and decrease in lipid peroxidation products.

As for selenium sources, there was no significant difference between sodium selenite and yeast selenium. It seemed that the effect of anti-stress of selenium had nothing to do with the form of selenium, but the effects of organic yeast selenium were superior to inorganic selenium slightly. In any case, supplementation with high level of selenium could attenuate stress and oxidative injury caused by dietary CORT.

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REFERENCES

Arthur, J.R., F. Nicol and G.J. Beckett, 1990. Hepatic iodothyronine 5'2-deiodinase?: The role of selenium. Biochem. J., 272: 537-542.

Darras, V.M., S.P. Kotanen, K.L. Geris, L.R. Berghman and E.R. Hqhn, 1996. Plasma thyroid hormone levels and iodothyronine deiodinase activity following an acute glucocorticoid challenge in embryonic compared with posthatch chickens. Gen. Comp. Endocrinol., 104: 203-212.

Eid, Y.Z., A. Ohtsuka and K. Hayashi, 2003. Tea polyphenols reduce glucocorticoids induced growth inhibition and oxidative stress in broiler chickens. Br. Poult. Sci., 44: 127-132.

Ekholm, R. and U. Bjorkman, 1997. Glutathione peroxidase degrades intracellular hydrogen peroxide and thereby inhibits intracellular protein iodination in thyroid epithelium. Endocrinol., 138: 2871-2878.

He, J.H., A. Ohtsuka and K. Hayash, 2000. Selenium influences growth via thyroid hormone status in broiler chickens. Br. J. Nutr., 84: 727-732.

Jiang, K.J., 2007. Effects of stress and dietary energy level on growth and lipid metabolism of broiler chickens. Master's thesis. Shandong Agricaltual University, Tai'an, China.

Lin, H., E. Decuypere and J. Buyse, 2004. Oxidative stress induced by corticosterone administration in broiler chickens (Gallus gallus domesticus)1. Chronic exposure. Comparative Biochem. and Physiol., 139: 737-744.

Malherios, R.D., V.M. Moraes, A. Collin, E. Decuypere and J. Buyse, 2003. Free diet selection by broiler as influenced by dietary macronutrient ratio and corticosterone supplementation.1. Diet selection, organ weights and plasma metabolites. Poult. Sci., 82: 123-131.

Moberg, G.P. and J.A. Mench, 2000. The Biology of Animal Stress: Basic Princilies and implications for animal welfare. CABI Publishing: 3-6.

National Research Council, 1994. Nutrient Requirements of Poultry. 9th Rev. Ed. National Academy Press, Washington, DC.

Ohtsuka, A., T. Ohtani, H. Horiguchi and K. Hayashi, 1998. Vitamin E reduces glucocorticoids induced growth inhibition and lipid peroxidation in rats. J. Nutri. Sci. Vitaninl., 44: 237-247.

Sui, S.J., 2005. Effects of stress and energy type of diets on meat quality of broilers. Master's thesis. Shandong Agricaltual University, Tai'an, China.

Taniguchi, N., A. Ohtsuka and K. Hayashi, 1997. Effect of dietary corticosterone and vitamin E on growth and oxidative stress in broiler chickens. Anim. Sci. J., 70: 195-200.

Virden, W.S., M.S. Lilburn, J.P. Thaxton, A. Corzo, D. Hoehler and M.T. Kidd, 2007. The effect of corticosterone induced stress on amino acid digestibility in Ross broiler. Poult. Sci., 86: 338-342.

Watanabe, C., C.Y. Kima and H. Satoha, 1997. Tissue-specific modification of selenium concentration by acute and chronic dexamethasone administration in mice. Br. J. Nutr., 78: 501-508.

Yuan, S.B., 2007. Impacts of oxidative stress on piglets and the anti-stress effect and mechanism of selenium. Doctorial thesis. Sichuan Agricaltual University, Ya'an, China.