



The effect of Se-fortified wheat in feed on concentrations of selenium and GPx and SOD in blood of laying hens

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ABSTRACT

The aim of the study was to determine concentrations of selenium and enzymes GPx and SOD in blood of hens which were consuming feed with Se-fortified wheat. The study was conducted on 70 Tetra SL hybrid hens which were in the 40th week of production. Hens were fed with prepared mixtures for 26 days. After this period hens' blood was sampled for analysis of selenium and glutathione peroxidase (GPx) and superoxide dismutase (SOD) content. Treatments used in the study had no effect on weight of hens and egg production between examined groups ($P>0.05$). Selenium content in laying hens' blood of both groups was balanced and amounted for group A=0.1878 $\mu\text{g}/\text{kg}$ and for group B=0.1877 $\mu\text{g}/\text{kg}$ ($P>0.05$). The higher activity of the GPx enzyme was determined in blood of hens from experimental group B compared to group A (42935.3 U/l and 35675.5 U/l, respectively, $P<0.05$). SOD values showed similar trend as GPx values, with significantly higher ($P<0.01$) activity of this enzyme in blood of group B hens (0.9955 U/l) compared to group A hens (0.8101 U/l). Using wheat fortified with selenium in the diets for laying hens can affect a better supply of this microelement what was determined from the blood analysis.

(Keywords: wheat, laying hens, selenium, blood, enzymes)

INTRODUCTION

Selenium is an essential trace element for people and animals and it have to be taken with food. Selenium have multiple role in many biochemical processes in the body. Sufficient body supply with selenium will affect a better condition of immune, enzymatic and reproductive system (Surai, 2002). Selenium is a major component of the antioxidant defense mechanism in all living tissues because it is an integral part of a number of enzymes involved in cellular antioxidant defense (Tapiero *et al.*, 2003). Antioxidant system at the cellular level consists of three levels of defense. The first level of defense is responsible for preventing the formation of free radicals by removing their precursors. Defense at this level is provided by superoxide dismutase (SOD), glutathione peroxidase (GPx), catalase (CAT), glutathione, thioredoxin system and metals that make up proteins. The second level of defense is responsible for the prevention of chain breaking and its propagation, and it consists of vitamins A, C and E, carotenoids, ubiquinones, glutathione and uric acid. Lipases, peptidases, proteases, transferases, enzymes for DNA repair are parts of the third level of cellular protection and are responsible for cutting and repairing damaged parts of the molecule (Surai, 2006). Selenium is added to the poultry feed in two forms, inorganic and organic. A lot of

researchers have described better bioavailability of selenium if it is added to the feed in organic form (Skrivan *et al.*, 2006; Payne *et al.*, 2005; Rayman, 2004). Crop production aims to increase content of various trace elements in plants through fortification, which will then be consumed by animals in their diet in organic form.

The aim of this study was to determine concentrations of selenium and enzymes GPx and SOD in blood of hens that were fed mixtures with Se-fortified wheat.

MATERIAL AND METHODS

The study was conducted on 70 Tetra SL hybrid hens, divided into two groups (A and B). Hens were in the 40th week of production and during 26 days they were consuming mixtures in which 10% of corn was replaced with wheat. Wheat of Srpanjka variety was grown on calcareous soils with 7 different treatments in a randomized block design, and for feeding experiments wheat from two fertilization treatments was used, as follows: A=control without Se and B=foliar application of Se (10 g Se ha⁻¹). The composition of mixtures for hens is shown in *Table 1*. Analysis of selenium content in mixtures was made shortly before feeding trial. It was determined that mixture A contains 0.3059 mg Se/kg diet, and mixture B 0.5484 mg Se/kg diet. During the experimental period production of eggs was recorded, and weight of hens was controlled (beginning and end of the experiment).

At the end of the trial period 7 animals from each group were randomly selected for blood drawing in order to determine the concentration of selenium and status of enzymes glutathione peroxidase (GPx) and superoxide dismutase (SOD). Blood was drawn from the wing vein into sterile vacuum tubes BD Microtainer® SST™ (Becton, Dickinson and Company, USA). GPx concentration was determined from the whole blood and SOD concentration from serum of same animals. For enzyme determination commercial kits Randox Ransel RS 505 and Randox Ransod SD 125 (Laboratories Ltd, London, UK) were used, while selenium concentration in the blood was analyzed using a Perkin Elmer Optima 2100 DV device (Davidowski, 1993). Results of the research were analyzed by statistical software Statistica for Windows version 12.0 (StatSoft Inc., 2013).

RESULTS AND DISCUSSION

Table 2 shows the weight of laying hens used in the experiment, at the beginning and at the end of the experiment, and the production of eggs during the trial period. Treatments used in the experiment had no effect on the difference in weight of hens and egg production between two groups ($P>0.05$). In a study on the influence of fortified barley on performance of laying hens and their offspring Hassan (1990) reported that concentrations of selenium in fortified barley has no effect on the weight of laying hens, which is consistent with our results. Haug *et al.* (2008) in the study on the bioavailability of selenium from feed that contains wheat fortified with selenium on the performance and selenium content in muscle tissue of chickens, pointed out that the levels of selenium in feed had no effect on the weight of chickens, which is in accordance with our research. Gjorgovska *et al.* (2012) found statistically significant effect of organic selenium levels in feed for laying hens on egg production. Their results are not consistent with ours. Yoon *et al.* (2007) reported results consistent with ours and pointed out that the source but also the level of selenium in feed has no effect on the weight of chickens ($P>0.05$).

Table 1

Composition and chemical analysis of the mixture for laying hens

Ingredient, %	¹A and B
Corn	40,75
Triticale	6,60
Wheat	10,00
Soybean meal	18,33
Toasted soy	8,33
Sunflower meal	1,66
Alfalfa	1,00
Calcium granules	8,13
Monocalcium	1,58
Yeast	0,50
Salt	0,33
Mineral Detox	0,25
Probiotic Pro Bio	0,05
Methionine	0,25
Premix ²	0,58
Soybean oil	1,66
Total	100,00
³Chemical analysis of the mixture (g/kg)	
Moisture	87
Crude protein	190
Crude fiber	40
Ash	139
Fat	52
Ca	41

¹in mixtures 10% of corn was replaced with wheat as follows: A=control without selenium fortification and B=wheat fortified with 10 g Se ha⁻¹

²Premix smixture K, content in 1 kg: vitamin A 200000 UI, vitamin D₃ 500000 UI, vitamin E 10000 mg, vitamin K₃ 600 mg, vitamin B₁ 400 mg, vitamin B₂ 1000 mg, vitamin B₆ 1000 mg, vitamin B₁₂ 3000 µg, vitamin C 4000 mg, vitamin H 12 mg, vitamin B₃ 8000 mg, vitamin B₅ 2400 mg, vitamin B₉ 150 mg, vitamin B₄ 100000 mg, iodine 200 mg, manganese 18000 mg, zinc 14000 mg, cobalt 30 mg, iron 12000mg, copper 1600 mg, inorganic selenium 50 mg, calcium 238 g, phytase 100000 FYT, canthaxanthin 500 mg, beta-apo-beta-carotenoic acid 300 mg, antioxidant (butyl hydroxytoluene) 20000 mg

³Reference methods used for chemical analysis of feed: HRN ISO 6496:200; HRN EN ISO 5983-2:2010; HRN EN ISO 6865:2001, modified according to the instructions FOSS Fiber Cap manual; HRN ISO 5984:2004; HRN ISO 6492:2001, modified according to the instructions of the extraction system ANKOM XT15; RU-5.4.2-11 (internal method)

Stressful situations in mammals, such as starvation and intensive production, are associated with changes in the antioxidant defense of the body and it can vary according to the species, while values in the blood are not correlated with those determined in the organs (*Wohaieb and Godin, 1987*), because of particular role of the metabolism of certain organs. Unlike mammals, birds have a higher body temperature and more intense metabolism and therefore they need much more oxygen. For this reason birds are exposed to stronger oxidative stress. Adding selenium to poultry feed in intensive farming has a positive effect on the increase in antioxidant activity.

Table 2

Weight of hens and egg production

Production indicators	A	B
Hens weight (g)	($\bar{x} \pm \text{sd}$)	($\bar{x} \pm \text{sd}$)
Experiment beginning	2081,60±144,82	2062,68±133,65
Experiment end	2159,91±230,38	2083,14±129,66
Egg production (pcs.)	882	880

Table 3 shows the effect of selenium levels in feed for laying hens on concentration of selenium and enzymes GPx and SOD in blood. It is evident that the level of selenium in mixtures for laying hens has no effect on the concentration of selenium in the blood ($P>0.05$). A higher concentration of the enzyme GPx was found in hens in group B compared to group A (42935.3 U/l and 35675.5 U/l, respectively, $P<0.05$). SOD values showed similar trend as GPx values, with significantly higher ($P<0.01$) values of this enzyme in the blood of group B hens (0.9955 U/l) compared to group A hens (0.8101 U/l). *Gajčević et al.* (2009.) point out that higher level of selenium in the diet (0.4 ppm) has a positive effect on GPx activity in blood of laying hens ($P<0.05$). Numerous studies related to the use of selenium in poultry feed represent a connection to research of the impact of selenium sources (inorganic or organic) on different production traits in animals. At the conclusion of each of the studies better results were obtained using an organic form of selenium. *Mahmound and Edens* (2003), who were investigating the impact of different sources of selenium on the biochemical changes in the blood of hens, found a higher GPx activity in the blood of birds that consumed selenium from organic sources in the diet. Positive experiences of using organic selenium compared to inorganic selenium in poultry feed mentioned also *Leng et al.* (2003), who highlighted the positive correlation between levels of selenium in feed and GPx activity in blood of broiler chickens.

Table 3

Influence of selenium level in feed on concentrations of selenium and GPx and SOD enzymes in blood of laying hens

Indicator	A ($\bar{x} \pm \text{sd}$)	B ($\bar{x} \pm \text{sd}$)
Se ($\mu\text{g}/\text{kg}$)	0,1878±0,008	0,1877±0,006
GPx (U/l)	35675,5±5713,4 ^b	42935,3±3589,7 ^a
SOD (U/l)	0,8101±0,088 ^B	0,9955±0,058 ^A

Values in rows marked with ^{A,B} exponents differ at $P<0,001$ level, and those marked with ^{a,b} differ at $P<0,05$ level.

Hassan (1990) stated that activity of GPx enzyme was significantly reduced ($P<0.01$) in hens with a deficit of selenium in feed. In accordance with our results *Yoon et al.* (2007) state that increased levels of organic selenium in feed can significantly ($P<0.05$) increase concentration of selenium and GPx enzymes in the blood of chickens.

CONCLUSIONS

From the research results it can be concluded that the use of Se-fortified wheat in feed for laying hens influenced greater supply of body with selenium, which resulted in significantly higher levels of GPx and SOD enzymes in blood of hens from experimental group B compared with group A ($P < 0.05$ and $P < 0.01$). In order to obtain even better results it is necessary to expand the research, and to add fortified corn as well as a greater proportion of fortified wheat into laying hens' feed.

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