

## **THE ROLE OF THE ANTIOXIDANT - SELENOPYRAN IN THE STABILITY OF THE ANTIOXIDANT SYSTEM TO TOXIC EFFECT OF CADMIUM CHLORIDE\*\***

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**Abstract:** The purpose of the present work was, under conditions of the model experiment on rats, to tap the information about the features of short-term acclimatization of the antioxidant system in various organs to toxicity of cadmium at stages of an ontogenesis and about the preventive role of the antioxidant selenopyran in this process.

The obtained results showed the ontogenetic differences in the adaptive reactivity of the enzymatic antioxidant defense system in the most important organs and tissues of rats under conditions of oxidative stress induced by cadmium.

The ontogenetic differences of Se redistribution in a body under influence of cadmium administration were found. The discovered decrease of Se concentration in the liver of young animals and the increase of its concentration in the liver of old animals correlated positively with the changes of GPx activity. Preventive administration of selenopyran (9-phenyl-simmetrical octa-hydroselenoxanthene) to old animals reduced the oxidative stress intensity. Animals of all age groups showed higher selenium concentration in the tissues and the increase in the selenium-dependent GPx activity.

**Key words:** antioxidant, selenopyran (9-phenyl-simmetrical octa-hydroselenoxanthene), heavy metal cadmium, antioxidative defense, model animals.

## Introduction and literature review

Under conditions of the application of intensive technologies for animal breeding it is difficult to avoid influence of the polluted environment and contamination of feedstuffs with various pollutants, among which the special place belongs to the heavy metals. Except for direct toxicity, they possess a lot of unfavorable properties: allergenicity, carcinogenicity, provocation of oxidative damages and etc. These disruptions are holdback to full realization of genetically potential of high productive animals.

The major role in the stability of organism to oxidative disruptions plays the enzymes of antioxidative defense. The structure of its contents a big number of SH – groups (superoxide dismutase, glutathionreductase). Heavy metal cadmium react quickly with SH – groups (*Figueiredo-Pereira et al, 1998*) and replace zinc in the active centers of enzymes (*Vallee and Ulmer, 1972*). Thus leads to change of the antioxidative enzymatic activity.

For the prevention of this effect are used the antioxidants, before all selenium, which constitutes an essential component of antioxidative enzymes (glutathione peroxidase). The selenium-containing compound designed to supplement is expected to have the following properties hypotoxicity and biological availability of selenium in the composition. In this way, the organic selenium is more preferable, than sodium selenite.

The aim of the present work was the study of the protective effect of selenorganic compound - selenopyran under Cd induced oxidative stress in experimental animals.

## Material and methods

The experiment was conducted with 59 Wistar male rats at different ages: 60 days old, 90 days old and 24 months old. During the 7 days they received a standard for laboratory animals' diet. At each age the animals were divided in three groups: control and two experimental – treatment with cadmium (Cd) and with cadmium and selenopyran (Cd+Se). The animals from first experimental group during the last 4 days were obtained cadmium chloride in dose 4 mg/kg body weight into stomach by catheter. The animals from second experimental group from first day were obtained selenopyran in dose 0,21mg/kg body weight and during the last 4 days - cadmium chloride in dose 4 mg/kg body weight into stomach by catheter. Organic compound of selenium under the name of selenopyran was synthesized in 1981 by

Russian academician *A.F. Blinokhvatov*. Its chemical name is 9-phenyl-symmetrical octahydroselenoxanthene. The toxicity of it is more lower than sodium selenite ( $LD_{50}=1600\text{mg/kg}$  against  $LD_{50}=3,25\text{mg/kg}$ )

At 8-th day the biological material for analyzes was collected. The lipids peroxidation products, the activity of antioxidative enzymes and selenium content in organs and tissues were investigated.

The content of diene conjugates (DC) was estimated by *Placer* (1968) in modification of *Gavrilova et. al.* (4). Malondialdehyde(MDA) was detected by *Michara et.al.* (1980). For the estimation of glutathion reductase (GR) and glutathion peroxidase(GP) activity in animal tissues was used the methods of *Tilbotson* (1971) and *Mille* (1959) in modification of *Malcev et. al.* (1994); for super oxide dismutase activity(SOD) – method of *Niashikimi* (1972) in modification of *Malcev and Vasilev* (1994) for analyzer FP -901 («Labsystems»).

Selenium concentration in different organs was estimated by fluorometric method.

The obtained results are developed statistically by Student t-test.

## Results and discussion

Table 1 reflects the changes in selenium content in rat tissues after cadmium administration and addition of selenopyran. In young and old groups the toxic effect of cadmium leads to decrease of selenium in the investigated organs. Addition of selenopyran restored the selenium concentration. It needs to note that in all groups significantly increasing of Se content in testis was observed.

In sex mature (adult) group cadmium treatment leads to decrease of selenium in blood plasma, but to significantly increase of it concentration in testis (table 1).

Intragastral cadmium administration has affected the activity of both hepatic and intestinal enzymes as well as concentration of lipids peroxidation products in these organs (Fig.1 and 2). Considerable decrease of glutathionreductase activity was observed in both young and adult rats( $P<0,05$ ).The experiment has revealed a rapid adaptive response of glutathioneperoxidase (GPx) turnover systems in adult animals. On the contrary, the GPx activity decreased considerably in young rats ( $P<0,01$ ). The activity of superoxide dismutase has a tendency to increase in all groups in small intestine and in adult and old animals in liver. The concentration of lipids peroxidation products (MDA and DC) in small intestine and liver of

all groups increase after cadmium administration.

**Table 1. The change of selen concentration in rat issues after toxically effect of cadmium chloride and addition of selenopyran.**

Age of animals	Parametrs	Se in testis's	Se in liver	Se in blood plasma
		ng/g	ng/g	ng/ml
Young (60 days)	Control, n=8	81,0±8,5	912±77	185±16
	Cd, n=8	61*±3	488***±68	182±11
	Cd+ Selenopyran, n=7	†90**±7	†819*±118	†287**±25
Sex mature (Adult) (90 days)	Control, n=6	82±3	562±42	399±24
	Cd, n=6	117***±6	635±65	375±31
	Cd+ Selenopyran, n=6	147±36	698±44	369±23
Old (24 months)	Control, n=6	93±4	751±12	666±12
	Cd, n=6	74**±4	814±40	373***±24
	Cd+ Selenopyran, n=6	†95***±2	948±50	378±35

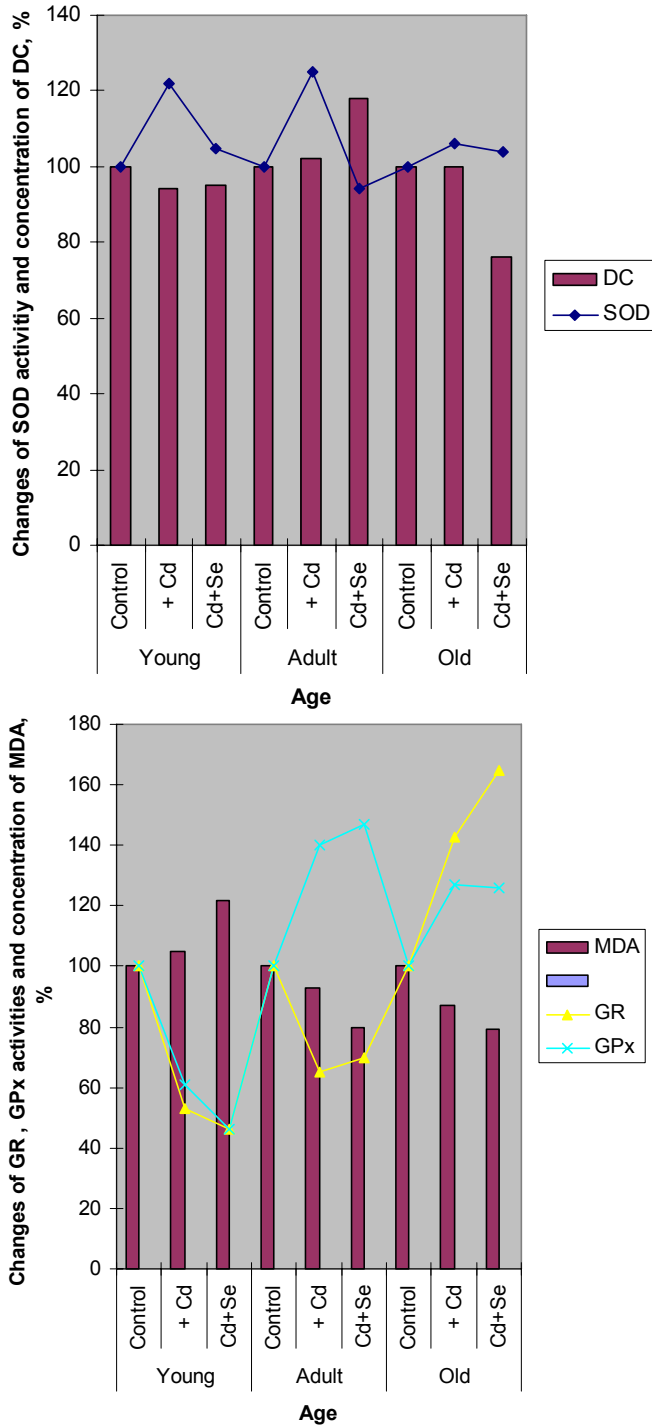
\* - differences between groups,  $p < 0.05$

\*\* - differences between groups,  $p < 0.01$

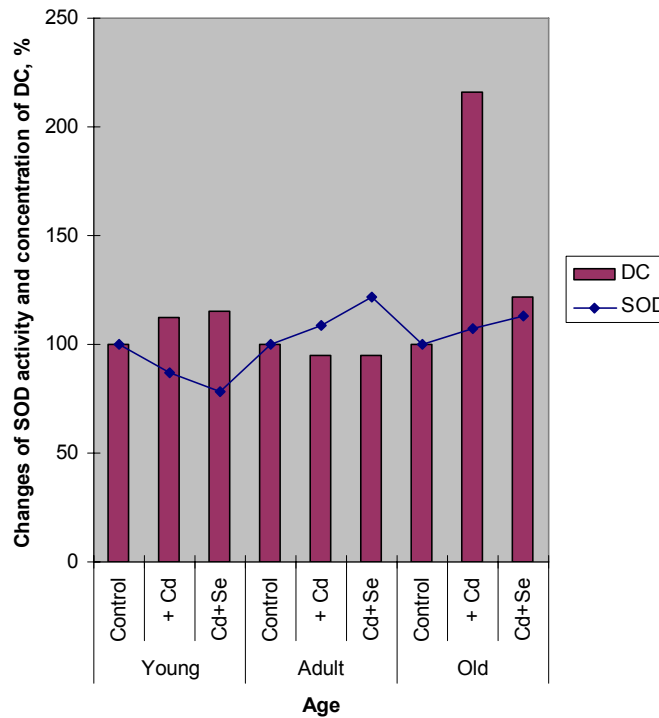
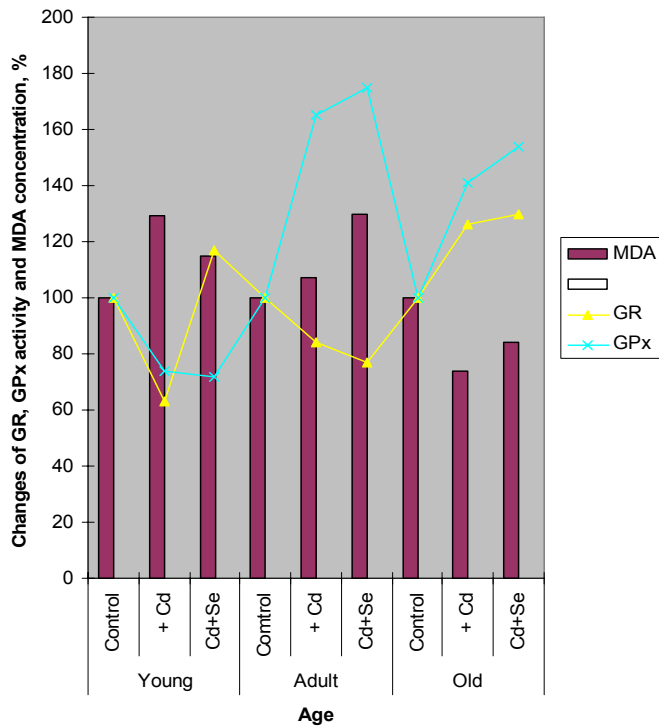
\*\*\* - differences between groups,  $p < 0.001$ ;

† - differences between groups «Cd» and «Cd+Selenopyran»

**Fig.1** Changes of enzymatic activity and concentration of the lipids peroxidation products in small intestine of control and experimental rats



**Fig.2** Changes of enzymatic activity and concentration of the lipids peroxidation products in liver of control and experimental rats



Selenopyran pretreatment had enhanced the GPx and GR intestinal and hepatic activity in both adult and old rats, whereas in young rats only hepatic activity of GR increases (Fig.2). The most preventive role of selenopyran against oxidative stress was expressed in the digestive organs of old animals. It reduces significantly the concentration of diene conjugates ( $P<0,001$ ).

In table 2 are shown the changes of concentration of lipids peroxidation products in blood plasma. Significantly differences were observed only in concentrations of DC in young and old animals. Selenopyran pretreatment decreases both the concentrations of DC and MDA in old animals

**Table 2. Concentration of lipid peroxidation products in blood plasma of control and experimental rats**

Age	Parameters	Diene conjugates (DC)	Malondialdehyde (MDA)
		nmol/ml	nmol/ml
Young (60 days)	C, n=8	4,31±0,44	11,7±0,52
	Cd, n=8	6,15**±0,29	10,9±0,5
	Cd+SeP, n=7	5,65±0,25	11,1±0,5
Adult (90 days)	C, n=6	3,98±0,29	10,5±0,7
	Cd, n=6	3,29±0,16	8,79±0,43
	Cd+SeP, n=6	3,35±0,13	7,51±0,65
Old (24 months)	C, n=6	2,75±0,23	4,16±0,27
	Cd, n=6	5,78***±0,168	4,77±0,09
	Cd+SeP, n=6	† 4,34*** ±0,08	† 4,01*** ±0,04

\* - differences between groups,  $p<0.05$

\*\* - differences between groups,  $p<0.01$

\*\*\* - differences between groups,  $p<0.001$ ;

† - differences between groups «Cd» and «Cd+Selenopyran»

On the age- depend and organs specific stability of antioxidant system to toxically administrations of different chemicals pointed out a lot of authors (Anisimov *et al.*, 1999; Samanta *et al.*, 1999; Saadet *et al.*, 2000; Youdim

and Deans, 2000). Selenium helped enhance the function of the body's antioxidants and rid the body of potentially damaging agents. Our results are in compliance with data of Chow and Chen (1980) that the selenium intake enhancement more the activity of GPx in adult rats than in young. More often the activity of GR didn't change.

Obtained results are in agreement with results of other authors investigated the dietary effect of organic selenium under different stress agents - heat stress, arsenic poisoning, cadmium intake (Modi et al, 2007; Kamel and Edens, 2003; Flora et al.,1982; 1999; Andersen and Nielsen, 1994). In all cases the organic selenium stimulate the activity of GPx antioxidant system in different organs - blood, liver. The effect of selenium could be reinforce with intake of vitamin E (Ebru and Mesut , 2003).

## Conclusion

The age differences of Se redistribution in a body under influence of cadmium administration were found. The discovered decrease of Se concentration in the liver of young animals and the increase of its concentration in the liver of old animals correlated positively with the changes of GPx activity. In sex mature (adult) animals after cadmium treatment and selenopyran pretreatment the higher concentration of Se in testis was detected compared with other animal groups. Thus point out importance of testis at this stage of ontogenesis.

In young animals pretreatment of selenopyran under cadmium administration in this dose didn't increase significantly the activity of antioxidative defense enzymes in small intestines and liver.

In adult animals rapid adaptive response of glutathioneperoxidase (GPx) to cadmium treatment was observed. Activity of this enzyme was increased more after pretreatment with selenopyran.

The most power preventive effect of selenopyran against oxidative stress in all investigated organs of old animals was observed. The activity of SOD, GR and GPx was increased and concentration of lipid peroxidation products was decreased.

On the base of the obtained results we could suggest the use of selenopyran in animal feeding as a good source of selenium, which is needed for selenium proteins construction and the stability of antioxidant system.



## **ULOGA ANTIOKSIDANTA - SELENOPIRANA NA STABILNOST ANTIOKSIDANTSKOG SISTEMA NA TOKSIČNI EFEKAT KADMIJUMHLORIDA**

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### **Rezime**

Svrha ovog rada je bila da se u ogledu na pacovima dođe do informacija o odlikama kratkoročne aklimatizacije antioksidativnog sistema u različitim organima na toksičnost kadmijuma u stadijumima ontogeneze I o preventivnoj ulozi antioksidanta selenopirana u procesu.

Ogled je izveden na 59 Wistar mužjaka pacova različitog uzrasta: 60 dana, 90 dana i 24 meseca. Eksperimentalne životinje su intragastralno dobijale kadmijumhlorid I organsko jedinjenje selen -selenopiran. Osmog dana eksperimenta je sakupljan biološki material za analize. Proizvodi peroksidacije lipida, aktivnost antioksidativnih enzima i sadržaj seleno u organima i tkivima su analizirani.

Dobijeni rezultati su pokazali ontogenetske razlike u adaptivnoj reakciji enzimskog antioksidantskog sistema odbrane u najvažnijim organima I tkivima pacova u uslovima oksidativnog stresa izazvanog kadmijumom.

Utvrđene su ontogenetske razlike redistribucije Se u telu pod uticajem davanja kadmijuma. Otkriveno smanjenje koncentracije Se u jetri mlađih životinja i povećanje koncentracije u jetri starijih životinja je bilo u pozitivnoj korelaciji sa promenama GPx aktivnosti. Preventivno davanje selenopirana (9-fenil-simetrični okta-hidroselenoksanten) starijim životinjama je smanjilo intenzitet oksidativnog stresa. Životinje svih starosnih grupa su pokazale više koncentracije seleno u tkivima i GPx aktivnosti koja zavisi od seleno.

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