ANNALES
UNIVERSITATIS MARIAE CURIE-SKŁODOWSKA
LUBLIN — POLONIA
VOL. LVII, N 1, 17 SECTIO D 2002

Department of General Chemistry, Medical University of Lublin

KAZIMIERZ PASTERNAK, EWA BIELAK

Influence of time period of cadmium intoxication on the concentrations of ascorbic acid selenium in certain tissues and blood serum of rats

Cadmium is a harmful substance. It mainly enters the organism through the alimentary tract and by way of respiration. Most cadmium is found in the kidneys, liver and bones. Cadmium is stored in the liver by low molecular-weight proteins – metallothioneins – which are characterized by relatively high cystein content. If cadmium levels exceed the metallothioneins' binding ability it can then threaten various cells (12, 15). Cadmium's toxic activity is associated first of all with its influence on the metabolism and function of many important bioelements. Interactions between these bioelements and cadmium can occur at various stages such as absorption, transport and excretion – important biological functions of the cell. The fact that cadmium and calcium have similar electron structures can influence absorption of calcium from the alimentary canal, which can lead to reduced biogenic amine synthesis.

Cadmium is a biological antagonist to zinc. Cadmium/zinc interactions can interfere in those metabolic processes that are zinc regulated, mainly the cellular production of DNA, RNA, proteins and enzymes. In the event of acute toxicity, an increase in peroxidase dysmutase has been observed in lungs and liver (14). In cases of chronic toxicity, this enzyme's activity has been observed to be decreased or unchanged. Cadmium absorption can be influenced by the presence of calcium, iron, zinc, copper, manganese and selenium as well as vitamins C and D (2). Supplementation with these bioelements and vitamins can limit dangerous effects of cadmium on the organism. Ascorbic acid can bind toxic cadmium ions in the form of complexes and remove these complexes from the organism (4).

Selenium can prevent the cadmium-influenced degeneration of the cell nucleus. In erythrocytes and certain tissues, glutathione peroxide, which contains selenium as a prosthetic group, is responsible for the morphological and functional changes caused by cadmium (1). Negative effects of cadmium on a series of metabolic processes and reports

concerning the protective effects of selenium and vitamin C have led us to conduct experiments attempting to elucidate the extent to which cadmium (depending on the concentration and period of intoxication) influences the concentrations of vitamin C and selenium in blood serum and certain tissues of experimental animals.

MATERIAL AND METHODS

Our experiments were conducted on two-month-old male Wistar rats weighing 120–150 g. The animals were divided into experimental groups. The I, II, IV and V groups received a cadmium chloride solution in their drinking water (groups I and IV – 3900mg/dm^3; groups II and V – 100 mg/dm^3). Groups III and VI served as controls – these animals received redistilled water. All animals were fed dry LSM animal feed and were watered *ad libitum*. The animals in groups I – III were sacrificed after 3 weeks. The animals in groups IV – VI were sacrificed after 6 weeks. All the animals were sacrificed by peritoneal ketamine administration. Blood and tissue samples were taken for analysis. Serum was obtained from the blood and the respective tissue samples were homogenized. Modified Kyaw spectrophotometric method was used in order to determine vitamin C concentrations in serum and homogenate eluents (13). In three of the selected tissues (liver, kidney, thigh muscle) selenium concentrations were determined by atomic absorption spectrophotometry (AAS) 7. The results were analyzed by Student t-test and Cochran-Cox method assuming $p< 0.05$ as significant.

RESULTS

The results obtained indicate that cadmium intoxication reduces vitamin C concentration in blood serum and in selected tissues (Fig. 1, Tab. 1).

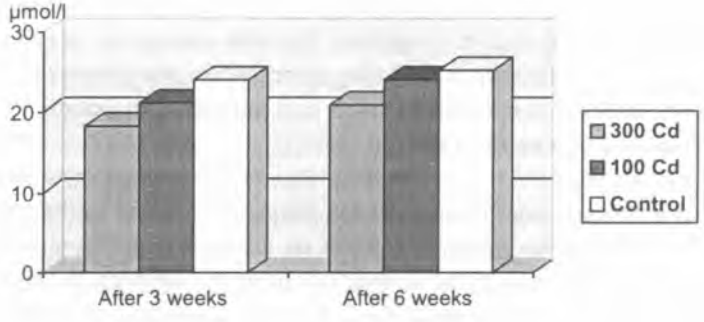

Fig. 1. Cadmium influence on ascorbic acid concentration in blood serum
*Results are an average of six experiments

In the case of blood serum, the decrease in vitamin C concentration was greater when the concentration of cadmium increased. However, no major changes in vitamin C concentration occurred when intoxication periods varied.

Table1. Cadmium influence on tissue ascorbic acid

Tested tissues	Ascorbic acid concentrations g/g of tissue					
	after 3 weeks of intoxication			after 6 weeks of intoxication		
	group I	group II	group III	group IV	group V	group VI
Liver	*192.0±44.2	*267.7± 42.9	334.6±56.9	*183.7±34.9	*209.6±37.7	244.8±51.4
Kidney	66.9±13.4	64.6±16.8	69.7±13.2	63.2±15.2	62.3±13.1	65.3±14.4
Spleen	60.6±13.3	68.1±!7.1	65.0±13.7	65.8±17.1	68.3±16.4	66.6±18.0
Heart	*53.7±19.4	60.6±13.3	69.0±16.6	*41.6±10.8	55.5±12.8	57.7±14.4
Brain	*22.8±5.3	25.2±5.7	29.9±7.5	*20.7±5.4	26.1±5.0	29.2±6.4
Femoral muscle	*19.5±4.9	21.1±4.0	26.3±6.8	*18.6±5.1	19.4±3.7	27.1±6.2

Group I and IV - cadmium 300 mg/dm^3, group II and V - cadmium 100 mg/dm^3, group III and VI - control.

The greatest drop in vitamin C levels occurred in the liver tissue. In the case of this tissue, higher cadmium concentrations and longer intoxication periods very clearly cause decrease in levels of vitamin C. These results were statistically significant when both of the experimental cadmium concentrations were administered for 3- and 6-week intoxication periods. In kidney and spleen, no vitamin C concentration changes were observed in the tested animals. In the case of heart, thigh muscle and brain tissues, only greater cadmium concentrations caused a statistically significant decrease in vitamin C concentrations. Longer periods of intoxication resulted in slight changes in vitamin C levels. This may be due to the fact that the rat has the ability to synthesize this vitamin. Intoxication may cause an increase in vitamin C synthesis, therefore changes in its concentration took place only in certain tissues when cadmium concentration was increased.

Our results indicate that cadmium levels and periods of intoxication both influence tissue selenium concentrations (Table 2).

Decreased selenium levels were most obvious in kidney tissue and were statistically significant at both lower and higher cadmium doses administered to the animals. In liver, selenium concentration was mainly dependent on the level of cadmium administered and was statistically significant at higher concentrations of this element. In thigh muscle, selenium concentration reductions were dependent on cadmium concentration levels and length of intoxication periods. The period of intoxication influenced selenium concentra-

Table 2. Cadmium influence on selenium concentration in certain rat's tissues

Tested tissues	After 3 weeks of intoxication			After 6 weeks of intoxication		
	group I	group II	group III	group IV	group V	group VI
Liver	*189± 36	560±42	580±48	*108±24	506±40	573±46
Kidney	*150±28	*845±73	1073±96	*102±18	*330±38	840±82
Femoral muscle	*108±24	*175±28	243±49	*104±30	*155±34	220±46

Group I and IV – cadmium 300 mg/dm^3, group II and V - cadmium 100 mg/dm^3, group III and VI - control.

tion levels to a small extent in all of the tested tissues. This perhaps was associated with the presence of adaptation mechanisms in the affected animals.

DISCUSSION

According to the literature, kidney tissue is especially sensitive to cadmium activity and accumulation of this element leads to renal tubule dysfunction (9). In experimental animals with cadmium-induced liver and kidney damage, altered zinc and copper metabolism has been observed, which is the earliest indicator of cadmium's nephrotoxicity (6). Cadmium intoxication decreases the activity levels of various enzymes. This process is reduced to a certain extent by supplementation. For example, in the presence of magnesium, cadmium reduced amino-acyl tRNA synthetase activity (11).

Cadmium causes liver and kidney damage. Damage to these organs leads to increased levels of alanine aminotransferase and malate dehydrogenase and proteins. In the case of both of these tissues, cadmium caused an increase in lipid peroxidase. Administration of antioxidants along with cadmium in the first phase of intoxication protected the animals, to a great extent, from inflammation and poisoning (14).

Vitamin C decreases the toxic effects of cadmium, which is demonstrated by lipid peroxidase assays (5). In cases of cadmium intoxication, increased lipid peroxidation was observed, however administration of cadmium together with vitamin C resulted in significantly reduced lipid peroxidation.

During the course of cadmium intoxication other elements play a significant role in its activity, either increasing or eliminating it (2, 3, 8).

Ognjanovic has observed results similar to ours (10). Furthermore, he also assayed the activities of vitamin E and antioxidative enzymes in the livers of cadmium-intoxicated rats, selenium-intoxicated rats as well as rats intoxicated with both cadmium and selenium together. He stated that cadmium intoxication resulted in reduced SOD, catalase and peroxidase activities as well as decreased vitamin E concentrations. Sele-

nium acted as an antagonist and increased SO and catalase activities as well as vitamin C and E concentrations. In cases of simultaneous cadmium/selenium administration, cadmium activity was significantly limited.

CONCLUSIONS

1. Cadmium intoxication caused reductions in vitamin C concentrations in blood serum and tested tissues. These changes were dependent on the intensity of intoxication and type of tissue tested. Period of intoxication influenced vitamin C concentration to a small extent.

2. Selenium tissue concentration decreased under the influence of cadmium in a manner dependent on the intensity and period of intoxication.

REFERENCES

1. Böck A.: Selenium Proteins Containing Selenocysteine. Encyclopedia of Inorganic Chemistry, 3700, 1994.
2. Brzóska M. et al.: Interakcje kadmu z wybranymi biopierwiastkami. Terapia, 7, 28, 1997.
3. Brzóska M. et al.: Disturbances in calcium metabolism in rats after oral cadmium intoxication. Acta Pol. Toxicol., 5(1), 91, 1997.
4. Cieślak-Golonka M.: Właściwości chemiczne i biologiczne kwasu L-askorbinowego. Wiad. Chem., 49, 9, 525, 1994.
5. Gupta P., Kar A.: Role of ascorbic acid in cadmium-induced thyroid dysfunction and lipid peroxidation. J. Appl. Toxicol., 18(5), 317, 1994.
6. Hałatek T., Chmielnicka J.: Dynamika wydalania wysoko i niskocząsteczkowych białek wiążących cynk i miedź z moczem szczurów narażonych na kadm. Acta Pol. Toxicol., 1, Supl. 1, 64, 1994.
7. Marczenko Z., Balcerzak M.: Spektrofotometryczne metody w analizie nieorganicznej. Wyd. PWN, Warszawa 1998.
8. Minta M. et al.: Embriotoksyczność selenu i kadmu w badaniach na chomiku złocistym. Acta Pol. Toxicol., 1, Supl. 1, 62, 1993.
9. Nagyova A. et al.: Histopathological evidence of vitamin C protection against Cd-nephrotoxicity in guinea pigs. Exp. Toxicol. Pathol., 46 (1), 11, 1994.
10. Ognjanovic B. et al.: The effects of selenium on the antioxidant defense system in the liver of rats exposed to cadmium. Physiol. Res., 44 (5), 293, 1995.
11. Pasternak K.: Wpływ kadmu na proces aktywacji aminokwasów w wątrobie, mózgu i nerce szczura. Streszczenia XXXV Zjazdu PTBioch. Olsztyn 13-16 IX, 194, 1999.

12. Roesijadi G.: Metallothionein and its role in toxic metal regulation, Comp. Biochem. Physiol., 113C, 117, 1996.
13. Rutkowski M., Grzegorczyk K.: Kolorymetryczne oznaczanie stężenia witaminy C w osoczu krwi przy użyciu odczynnika fosforowolframowego – modyfikacja metody Kyaw. Diagn. Lab., 34, 511, 1998.
14. Shaikh Z. A. et al.: Oxidative stress as a mechanism of chronic cadium induced hepatoxicity and renal toxicity and protection by antioxidants. Toxicol. Appl. Pharmacol., 154 (3), 256, 1999.
15. Sitarek K. et al.: Badania rozwoju fizycznego i behawioralnego potomstwa szczurów narażonych w czasie ciąży i laktacji na kadm, Acta Pol. Toxicol., 1, Supl. 1, 63, 1993.

2001.06.15

SUMMARY

Ascorbic acid and selenium are important antioxidants for the reactive forms of oxygen. The purpose of these experiments was to determine the influence of various concentrations of cadmium on ascorbic acid and selenium concentrations in certain tissues of rats. The results showed that cadmium, added to rats' feed, influenced the concentration of ascorbic acid in blood serum and tested rats' tissues. Cadmium influenced the concentration of selenium in certain tissues as well. Furthermore, it has been demonstrated that ascorbic acid and selenium concentrations are dependent on the type of tissue tested as well as on the size of cadmium concentrations and length of the experiments.

Wpływ czasu intoksykacji kadmem na stężenie witaminy C i selenu w wybranych tkankach i surowicy szczurów

Kwas askorbinowy i selen spełniają ważną rolę jako antyoksydanty w unieczynnianiu reaktywnych form tlenu. Celem przeprowadzonych doświadczeń było wykazanie wpływu różnych stężeń kadmu na stężenie kwasu askorbinowego i selenu w wybranych tkankach szczurów. Uzyskane wyniki wykazały, że kadm podawany w pożywieniu wpływał na stężenie kwasu askorbinowego w surowicy krwi i badanych tkankach szczurów. Kadm wpływał również na tkankowe stężenie selenu w wybranych tkankach. Stężenia kwasu askorbinowego i selenu zależały od rodzaju badanej tkanki, jak również od stężenia podawanego kadmu oraz czasu intoksykacji.