THE EFFECT OF DEUTERIUM DEPLETED WATER ON THE OSMOTIC RESISTANCE IN CADMIUM CLORIDE INTOXICATION

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Summary

The aim of this study was to show the effect of deuterium depleted water (DDW) treatement on the osmotic resistance and haemoglobin values in cadmium chloride intoxicated rats. The intoxication with cadmium chloride induced oxidative stress to rats; the oxidative stress is revealed at the membranes' level through the modification of their fragility. It was observed that DDW administration (preventive and also as treatement after the cadmium chloride intoxication in rats) has the property to increase the red blood cells membrane's permeability, bringing it back to the initial value at the end of the experiment.

Key words: rats, deuterium depleted water, red blood cells fragility, cadmium cloride.

Water is a fundamental factor of all the biological systems.Any changes of deuterium concentration (in water) can lead to important changes in cells.There are many studies witch proved that the decreasing of deuterium concentration influences many fundamental processes in cells.

Cd appears in atmosphere by many reasons: industrial and cars pollution and even cigarette smoke. It is an dangerous environmental polluting agent with carcinogen effect (15). There are several studies about the oxidative damages produced by cadmium(1,9,10). Cadmium produces oxidative stress indirectly through the inactivation of thiol group (SH), inhibiting the antioxidant defence (8, 14).The acute cadmium intoxication in single dose induce the lipid peroxidation and has the implications on red blood cells'membranes (4).

Materials and methods

The experiment was carried out on 48 adults Wistar male rats with a body weight of 250- 300 g. They were divided in four groups. All groups received the same dry food (60% corny blend, 30% corn and 10% sun flower) and were maintained in good physiological conditions.They were treated after the following protocol:

I-control group received natural drinking water (with a deuterium content of 150 ppm) ad libitum during 61 days.

II-received DDW (with a deuterium content of 30 ppm) ad libitum during 61 days.

III-received drinking water during 30 days, in the 31 day of the experiment a single dose of $CdCl_2$ (20 ppm Cd/Kg b.w.) was administrated by gastric tubing and another 30 days, drinking water ad libitum was administrated.

IV-pre-treated with DDW ad libitum during 30 days, in the 31 day of the experiment a single dose of $CdCl_2$ (20 ppm Cd/Kg b.w.) was administrated by gastric tubing and another 30 days DDW was administrated.

The blood was collected on heparine, by cardiac punction, under general narcosis, after 31 days from the bigining of the experiment (24 hours after the cadmium cloride intoxication of IIIa and IVa groups) and after 61 days (at the end of the experiment-IIIb and IVb groups).The osmotic resistance and haemoglobin were determined by colorimetric methods using an UV-VIS Shimadzu spectrophotometer (11).

Results and discussions

In this study at the III group the minimum osmotic resistance decreased due the cadmium chloride intoxication from 0.550% NaCl (in physiological limits according to Csopp et al cited by Ghergariu, 2000) (5) to 0.650% NaCl after 24 hours from the cadmium's administration (IIIa group) and maintened to the same value until the 61^{th} day (IIIb group), shown an increasing of erytrocytes' fragility during the experiment.This results are in accordance with previous studies (12).The average of minimum osmotic resistance in the group pre-treated with DDW (II) is in physiological limits according to Czopp et al cited by Ghergariu(5), decreased with 4. 54% compared to II group after the intoxication (IVa group), but it brought back to normal values until the 61^{th} day (IVb group).

At the cadmium intoxicated group who received drinking water (IIIa group) the maximum osmotic resistance decreased to 0.385% NaCl (from 0.300% NaCl at the control) after 31 days from the bigining of the experiment and increased to 0.350% NaCl after 61 days (IIIb group) been in physiological limits according to Hoffman et al cited by Ghergariu 2000)(5).

The group pre-treated with DDW had the maximum osmotic resistance with 8,33% lower than the control group (I), situated between the physiological limits (Hoffman et al, Czopp et al cited by Ghergariu 2000)(5). The installation of the oxidative stress induced by cadmium chloride's administration was observed by a decreasing of the maximum osmotic resistance to 0.400% NaCl (IVa group- 24 hours after cadmium chloride intoxication) compared to the control (0.300% NaCl) and after 61 days the same values like in IIIb group (in physiological limits) (5).

From the point of view of maximum red blood cells' resistence do not appear significant differences between the groups treated with DDW (II) or natural drinking water (I).The variations of the minimum osmotic resistence results that DDW offer a protection for the erytrocitary membrane against the oxidative

damages like the increase of its fragility. The cause for this increase of red blood cells' membrane's fragility can be the lipid peroxidation or membrane transport disturbances produced by cadmium (4,5,13).This experiment reveled the antioxidant effect of deuterium depleted water on red blood cells' membrane.

In our study in the 30th day the haemoglobin average value of II group were lower with 1.39% then in control. After 24 hours from the cadmium chloride's administration the haemoglobin had an important decreasing in bought groups (IIIa and IVa) according with other previous studies (2,3).

At the cadmium intoxicated group and treated with DDW for 30 days (IVb group) there was registered a significant increasing of the haemoglobin (104.21% compared to the control and 105.67% compared to the II group).At the cadmium intoxicated group who received drinking water (IIIb group) the increase of the haemoglobin was more slowly (98.7% compared to control after 61 days).

Table1.

Minimal and maximal osmotic resistence and haemoglobin average values

	I	II	III_a	III_b	IV_a	IV_b
	H_2O	DDW	$H_2O+CdCl_2$	$H_2O+CdCl_2+H_2O$	$DDW+CdCl_2$	$DDW+CdCl_2+DDW$
R_{min} (%NaCl)	0,550±0.025	0,550±0.15	0,650±0.05	0,650±0.05	0,575±0.015	0,550±0.025
R_{max} (%NaCl)	0,300±0.025	0,325±0.025	0,385±0.015	0,350±0.015	0,400±0.025	0,350±0.015
Hb (g/dl)	10,82±0,34	10,67±0,157	10,52±0,256	10,68±0,245	10.54±0.285	11,275±0,45

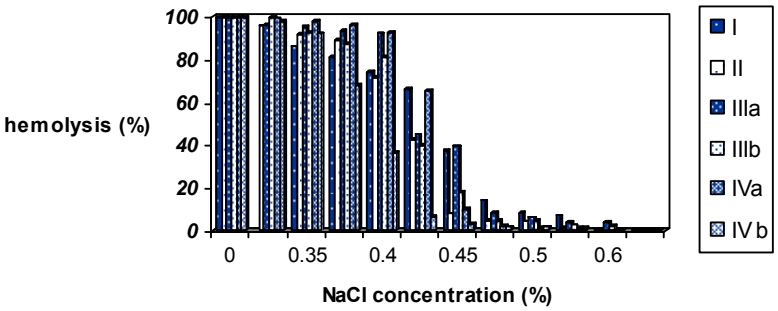

Fig.1 Hemolysis level in hipotonic solutions of NaCl in the control and experimental groups
(I-H_2O, II-DDW, IIIa- $H_2O+CdCl_2$, IIIb- $H_2O+CdCl_2+H_2O$, IVa-$DDW+CdCl_2$, IVb- $DDW+CdCl_2+DDW$)

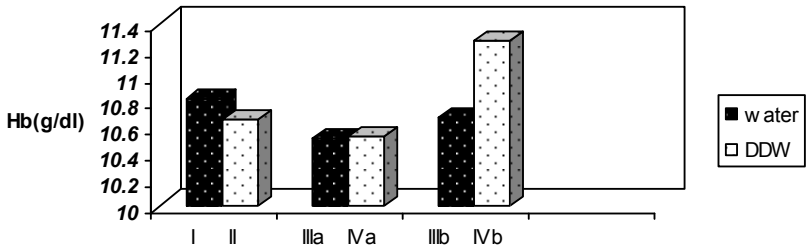

Fig.2 The haemoglobin variation in the experimental groups (I-H_2O, II-DDW, IIIa- H_2O+$CdCl_2$, IIIb- H_2O+$CdCl_2$ +H_2O, IVa-DDW+$CdCl_2$, IVb-DDW+$CdCl_2$ +DDW)

Conclusions

A long term treatment with the deuterium depleted water has antioxidant properties on the red blood cells membrane's fragility beeing in accordance with other previous studies (6,7).

The deuterium depleted water treatment helped to repair the damages produced by cadmium on rats' haemoglobin.

References

1.**D. Caisova, V.Eybl** 1986 „Effect of Cd^{2+} ,In^{3+} ,Ce^{3+} ,Tb^{3+} ,Eu^{3+} and Gd^{3+} on lipid peroxidation and glutathion level in liver of mice and rats", Biologia (Bratislava) 41, 1211-1219.

2.**C. Chaw,** American J. Clin. Nutr.,1979,32,1066-1081.

3.**Chişu Iuliana, Olariu Lucia, Tulcan Camelia**, Lucr. St."Actualități în patologia şi creşterea animalelor domestice „ 2002, vol.57-58, Cluj-Napoca.

4.**V. Eybl, Dana Kotyzova, J. Koutensky**,2006 „Comparative study of natural antioxidants –curcumin, resveratol and melatonin-in cadmium-induced oxidative damage in mice, Toxicology 225, 150-156.

5.**S. Ghergariu, A. Pop, L. Kadar, Marina Spînu,** „Manual de laborator clinic veterinar",Ed. All Educational, Bucureşti, 2000.

6.**I. Haulica, M. Poculea, I. Stefanescu, Gh. Titescu, M. Todiraş, W. Bild** 1998 „Effects of Heavy and Deuterium Depleted Water on Vascular Reactivity", Romanian Journal of Physiology (Physiological Science), vol.35, nr.12, 25-32.

7.**I. Haulica, I. Stefanescu, W. Bild, GH. Titescu, R. Tamaian, Veronica Nastasa, T. Ionita** „Research Concerning Physiological effects of

Deuterium Depleted Water on Unspecific Immunity", 2002 Conferința INGIMED, ed. IIIa, Bucureşti.
8. **K.S.**,**Kasprzak**, 2002 „Oxidative DNA and protein damage în metal induced toxicity and carcinogenesis", Free Radical Biol. Med. 32, 958-967.
9. **D. Manca, A.C. Ricard, B. Trottier, G. Chevalier,** 1991 „Studies on lipid peroxidation in rat tissues following administration of low and moderate doses of cadmium chloride"Toxicology 67,303-323.
10. **S. Sarkar , P. Yadav, R. Trivedi, A. K. Bansal, D. Bhatnagar**,1995 „Cadmium-induced lipid peroxidation and the status of the antioxidant system in rat tissues", J. Trace Elem. Med. Biol. 9, 144-149.
11. **Letiția Stana**, „Biofizică Metode de laborator", Ed. Solnes, Timişoara, 2006.
12. **Letiția Stana, Iuliana Chişu, Mihaela Petcu, Lucia Olariu, Camelia Tulcan**, „Rezistența membranei eritrocitare la şobolani intoxicați cu tetraclorură de carbon şi clorura de cadmiu", Lucr.Şt. Medicină Veterinară, vol.XXXVI, 2003, Timişoara.
13. **V. Stroescu**, „Bazele farmacologice ale practicii medicale", Ed. Med., Bucuresti, 1998.
14. **M.P.Waalkes,** 2003 „Cadmium carcinogenesis", Mutat.Rez. 533, 107-120.
15. International Agency for Research on Cancer Monographs, 1993 „Cadmium", vol.58.IARC Press, Lyon, pp.119-238.