THE EFFECT OF DEUTERUM DEPLETED WATER ON SOME HEPATIC ENZYMES’ ACTIVITY IN RATS INTOXICATED WITH CHROMIUM (VI)


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Abstract
The aim of these study was to show the effect of deuterium depleted water (DDW) (30 ppm deuterium) on the activity of some hepatic enzymes on rats intoxicated with chromium (VI). We used Wistar rats intoxicated with a single dose of K₂Cr₂O₇ - 20 ppm Cr/body weight. Is known that the chromium (VI) affects especially the liver. The hepatocyte status is depicted by the alanin aminotransferase (ALT) and aspartat aminotransferase (AST) activities. The effect of chromium (VI) was maintained for 60 days for all groups. The 90th days treatment with DDW help the liver to recover.

Key words: Chromium- Deuterium depleted water- rat- hepatic enzymes

INTRODUCTION
The chromium is an essential oligoelement. The organism needs small quantities of chromium and this can be taken normally by food (bread, potatoes, fresh fruits and vegetables, liver, black pepper, etc.). The chromium has an important role in the glucose’s metabolism, but the chromium salts are toxic for organism. The chromium toxicity depends on the oxidation stage and its solubility. Cr(VI) is more toxic than Cr(III) (Trivedi B., Saxena D.K.,1989). The chromium (VI) has toxic effects on liver and kidneis (Trivedi B., Saxena D.K.,1989, Sutherland, J. E. et al., 2000).

Deuterium depleted water (DDW) is water with a low deuterium content. If the tap water contains about 140-160 ppm deuterium, the DDW contain less then 80 ppm deuterium (in this study DDW contains 30 ppm deuterium). DDW is obtained at Drobeta Turnu Severin and Ramnicu Valcea, Romania.

There are many studies on deuterium depleted water, studies who prove that the DDW has a special influence on the cells and tissues development, has antioxidiant properties, influence the whole organism (Hăulică I., et. al., 2002, Somlyai G.,et. al., 1998, Somlyai G., 2001).
Deuterium depleted water has similar properties with the water from the inside of the cell of the living organism (Hăulică I., et. al., 2002).

MATERIAL AND METHOD

This experiment was carried on 96 Wistar female rats, adults, with a body weight of 200-260g, divided in 8 groups. The rats were maintained in good physiological conditions. The fodder received was a mixture of cereals (40% wheat, 40% corn, 20% sunflower), mixture which insures the nutritional and energetic value for the animals during the experiment, according to a corresponding diet.

L1 $\text{H}_2\text{O}$ - 60 days
L2 DDW (30 ppm) -60 days
L3 $\text{H}_2\text{O}$ - 30 days $\text{K}_2\text{Cr}_2\text{O}_7$
L4 DDW (30 ppm) -30 days $\text{K}_2\text{Cr}_2\text{O}_7$
L5 $\text{H}_2\text{O}$ - 30 days $\text{K}_2\text{Cr}_2\text{O}_7$ $\text{H}_2\text{O}$ - 30 days
L6 DDW (30 ppm) -30 days $\text{K}_2\text{Cr}_2\text{O}_7$ DDW (30 ppm) - 30 days
L7 $\text{H}_2\text{O}$ - 30 days $\text{K}_2\text{Cr}_2\text{O}_7$ $\text{H}_2\text{O}$ - 60 days
L8 DDW (30 ppm) -30 days $\text{K}_2\text{Cr}_2\text{O}_7$ DDW (30 ppm) - 60 days

A single doze of $\text{K}_2\text{Cr}_2\text{O}_7$ (20 ppm Cr(VI)/body weight) was administrated by gastric tubing in the 30$^{th}$ day. 24 hour after the chromium administration the blood were collected (on heparin), by cardiac punction from all groups. New samples were collected 30 respectively 60 days after the chromium intoxication from L3, L4, L5, L6 respectively L7 and L8.

The blood samples were collected under general narcosis (with chloroform) and the investigation were carried out with the approval of the Local Ethics Committee according the Romanian law 205/2004, art.7,18,22 and the regulation no.143/400/2002 and 37/2002, concerning with the protection of vertebrate animals used for experimental and other scientific purposes.
Alanin aminotransferase (ALT) and aspartat aminotransferase (AST) were determined in plasma by colorimetric methods (Ghergariu S. et al., 2000). DDW was obtained in accordance with a contract between the Faculty of Veterinary Medicine of Timisoara and the heavy water plant ROMAG, Drobeta-Turnu-Severin, Romania. The data are presented as means ± S.D. values. ANOVA, TTest, MINITAB and nonparametric test Mann-Whitney were used to analyse mean difference between experimental groups and for each parameter separately and between the groups.

RESULTS AND DISCUSSIONS

After 30 days from the beginning of the experiment the ALT’s activity was maintained in normal limits for the L2 groups, treated with DDW (30 ppm).

At L3, 24 hour after chromium intoxication the ALT’s activity significant decreased (p < 0.001) compared to the control (L1) and also to the L2 (treated with DDW) (83.7%). This value indicate an important affectation of the liver according to the scientific data (Kim H.Y. et al., 2004).

At L4 group, after 30 days of pre-treatment with DDW, the chromium intoxication after 24 hour made the ALT’s activity to decrease insignificant, to the 21.11± 5.82 UI, a little lower then the physiological values. The affectation of the liver is insignificant at the DDW pre-treated groups. 90 days treatment with DDW improved the ALT’s activity value (45.12± 7.88 UI) (p<0.05).This value is in physiological limits according the scientific paper (Ghergariu S. et al., 2000, Meingassner J.K., Schmook F.P., 1992). The results are presented in table 1 and figure 1.

The AST’s activity recorded significant lower values (p < 0.001) (8.508±0.28 U/l) at L3 groups (intoxicated with chromium and untreated, 24 hour after intoxication) compare to the control. In the pre-treated group (L4) the AST’s activity was significant higher even of the control (26.448± 21.923 U/l) (p<0.001).

After 60 days of treatment with DDW the AST’s activity values showed that the chromium intoxication affected the liver yet. After 90 days of treatment with DDW the values of AST activity were similar with those of the scientific paper (Ghergariu S. et al., 2000, Meingassner J.K., Schmook F.P., 1992), significant increased (p<0.001). The results are presented in table 1 and figure 2.
Table 1.

The mean values of alanin aminotransferase and aspartat aminotransferase in rats intoxicated with chromium (VI) in single dose and treated with DDW

<table>
<thead>
<tr>
<th></th>
<th>ALT (U/l)</th>
<th>AST (U/l)</th>
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<tr>
<td>L1 (H₂O)</td>
<td>23.885± 1.202</td>
<td>18.08± 1.571</td>
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<tr>
<td>L2 (DDW 30)</td>
<td>30.69± 5.917***</td>
<td>16.81± 1.149**</td>
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<tr>
<td>L3 (H₂O + Cr)</td>
<td>4.988± 0.159***</td>
<td>8.50± 0.281***</td>
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<tr>
<td>L4 (DDW + Cr)</td>
<td>21.10± 5.825</td>
<td>26.4± 1.923***</td>
</tr>
<tr>
<td>L5 (H₂O + Cr + H₂O)</td>
<td>4.36± 0.377**</td>
<td>11.2± 2.420***</td>
</tr>
<tr>
<td>L6 (DDW + Cr + DDW)</td>
<td>26.16± 2.498</td>
<td>14.42± 7.999</td>
</tr>
<tr>
<td>L7 (H₂O + Cr + H₂O60)</td>
<td>50.455± 13.827***</td>
<td>89.23± 4.827***</td>
</tr>
<tr>
<td>L8 (DDW + Cr + DDW60)</td>
<td>45.117± 7.880**</td>
<td>81.4± 8.039***</td>
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Mean ± S.D.; n= 12 animals/group, * p> 0.05, ** p< 0.05, *** p< 0.01

Figure 1. The mean values of ALT’s activity in rats intoxicated with chromium in single dose and treated with DDW
Figure 2. The mean values of AST’s activity in rats intoxicated with chromium in single dose and treated with DDW

CONCLUSIONS

The chromium maintained its toxic effects even after 60 days from the intoxication.

After 90 days of treatment with DDW (30 ppm) the ALT and AST’s activity reached similar level to those recorded in the scientific paper.

The DDW pre-treatment (30 days) protects the liver from the chromium toxicity and have an important scavenger role.

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