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Effects of Cobalt(II) Compounds on Some Hematological Parameters in Developing Mice

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The effect of chronic exposure to cobalt(II) depends on the type of compound used, dose, time duration as well as on the age of the experimental animals. Day18 mice showed to be more sensitive to Co(II) treatment. Higher hemoglobin content was measured in samples treated with $CoCl_2$ compared to those with CoEDTA. Higher plasma Fe concentration was measured in samples of mice treated with Co-EDTA which corresponded to low hemoglobin content.

Key words: cobalt(II) compounds, mouse blood plasma, hemoglobin, iron.

Introduction

As inorganic and complex compounds (with organic ligand) cobalt(II) is used as nutritional supplement, preservative, in drinks, as therapeutic agent for treating different diseases, etc. Cobalt(II) accumulates in organs such as kidney, liver, spleen, heart, stomach, intestines, muscle, brain and testes [1]. Exposure to this metal also causes allergic contact dermatitis, diseases of the upper respiratory tract, etc. [7]. Young animals (rats and guinea pigs) have 3- to 15-fold greater absorption than adult animals (aged 200 days or more). Water-soluble cobalt compounds exhibit greater absorption than non-water-soluble forms but absorption is species dependent [9]. Cobalt(II) chloride ($CoCl_2$) is a water soluble, hypoxia-mimicking agent. Ethylenediamine tetraacetic acid (EDTA) is a widespread organic pollutant. It is a powerful antioxidant and due its ability to bind metals it is used in chelation therapy. $CoCl_2$ oral treatment of patients with refractory anemia increased bone marrow erythropoietic activity, hematocrit and mean cell volume [3]. Topashka-Ancheva et al. determined that consuming food containing industrial dust with cobalt induces changes in hemoglobin, hematocrit, in red and white blood cell counts [8]. Iron (II), on the other hand, is incorporated in the heme complex which is an essential component of the oxygen carrier proteins such as hemoglobin. The daily requirement of iron is about 25 mg. Approximately about 80% of total body Fe is

incorporated into hemoglobin. Studies show strong relationship between cobalt blood and serum concentrations and iron status, having low cobalt concentration in case of high body iron stores [2].

The *aim* of the present work is to study the effects of cobalt(II) compounds – $CoCl_2$ and Co-EDTA on some hematological parameters in developing mice.

Material and Methods

Animal model

Pregnant balb/c mice in late gestation were subjected to cobalt chloride ($CoCl_2.6H_2O$) or Co-EDTA treatment at daily doses of 75 mg/kg or 125 mg/kg. Cobalt(II) compounds were dissolved and obtained from drinking tap water. Animals were fed a standard diet and had access to food *ad libitum*. Mice were maintained in the institute's animal house at 23°C ± 2°C and 12:12 h light-dark cycle in individual standard hard bottom polypropylene cages to ensure that all experimental animals obtained the required dose $CoCl_2$. The newborn pups were sacrificed on days 18, 25 and 30 which correspond to different stages of development. Mice were weighed weekly and the experimental cobalt concentration was adjusted accordingly. Whole blood samples were obtained, centrifuged and plasma was stored at –20°C until further analysis. Plasma samples were used for measuring hemoglobin (Hb) and iron (Fe) concentration. Hb concentration was determined by hemiglobincianide method (HiCN). Plasma iron concentration (transferrin-bound iron) was measured using spectrophotometric bathophenanthroline method [5]. The Fe content was calculated using standard solution of iron (II) (Sentinel).

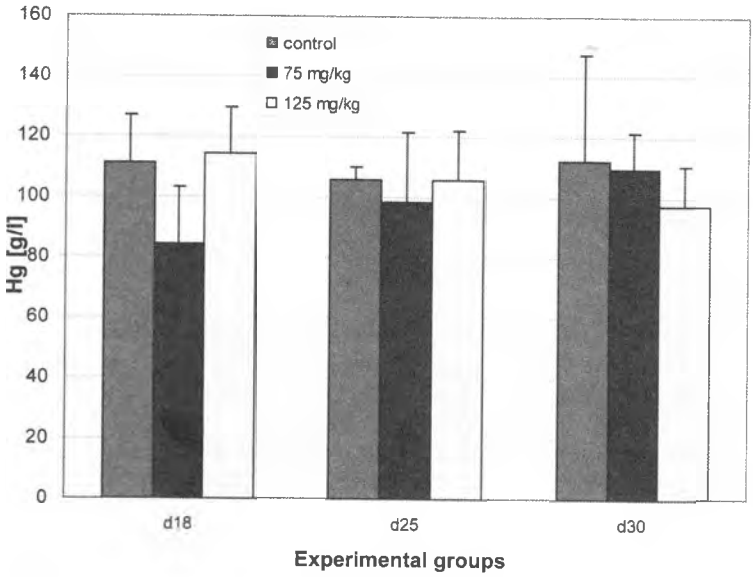
Statistical analysis

The obtained results are presented as mean value ± SD. Statistical significance between the experimental groups was determined using Student's *t*-test. Difference was considered significant at $p<0.05$.

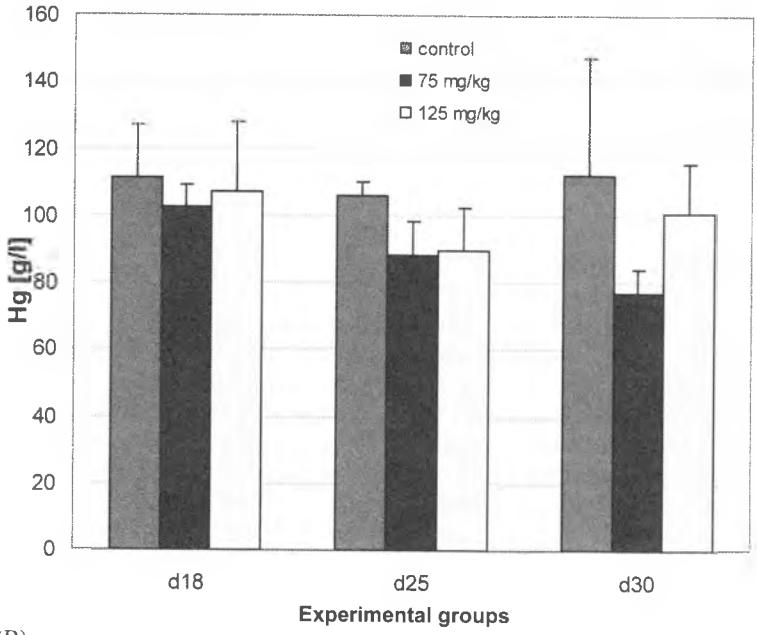
Results and Discussion

Results showed that high dose (125 mg/kg) of $CoCl_2$ induced an increase in Hb concentration in d18 mice. Treatment with low dose (75 mg/kg) reduced Hb concentration in all experimental groups. When CoEDTA was used Hb content was lower than the control samples in both cases – with low and high dose treatment. Results showed that high doses of $CoCl_2$ enhanced hemoglobin biosynthesis compared to CoEDTA.

Plasma Fe concentration was lower in samples of d18 and d25 mice treated with high dose $CoCl_2$ compared to the same age mice treated with the low dose. Higher Fe content was measured in sampled treated either with low or high dose CoEDTA which corresponded to the lower Hb concentration measured. Elevated iron levels are also a sign for changes in the liver function, i.e. reduced levels of hepcidin, etc. Cobalt and iron are shown to compete with close affinity constant for transferrin [4]. Thus, cobalt competes with iron for incorporation in the heme moiety which may be a possible explanation for the increased plasma Fe content [6].

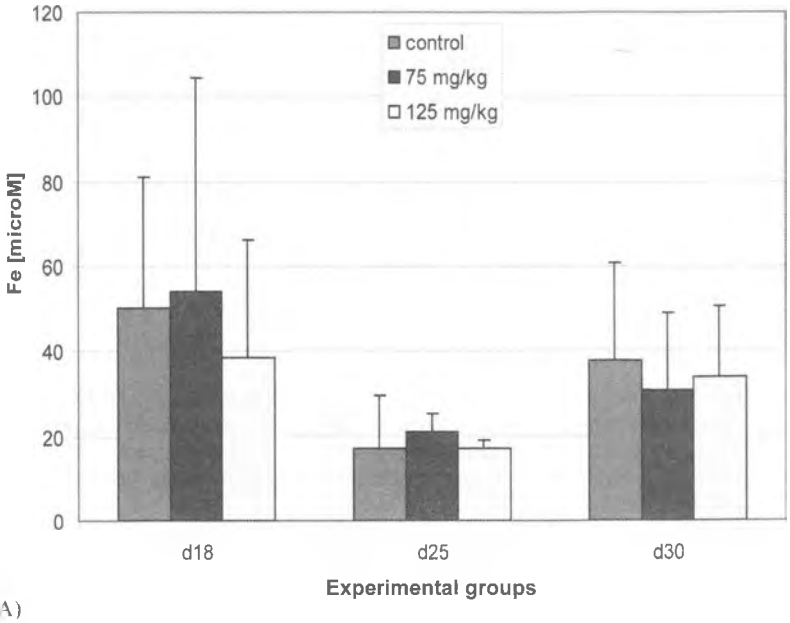


(A)

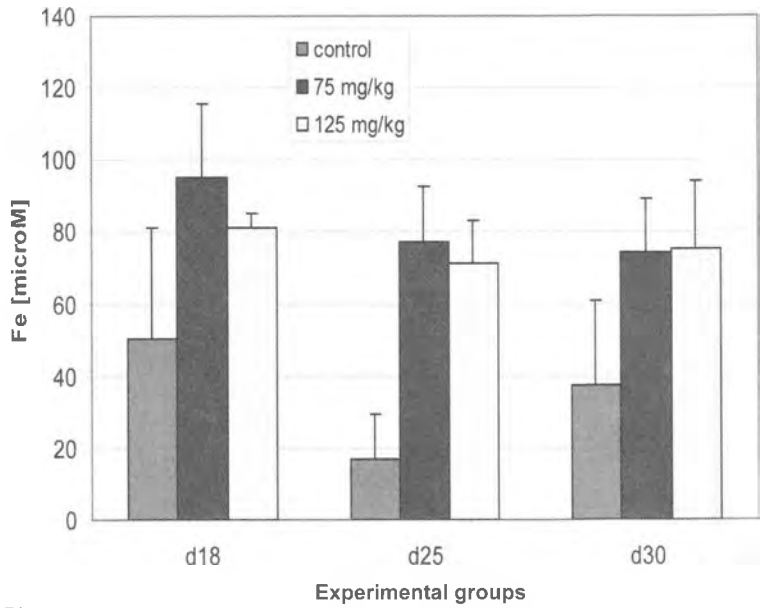


(B)

Fig. 1. Hemoglobin (Hb) content in blood plasma of mice treated with low or high daily doses of $CoCl_2$ (A), and CoEDTA (B)



(A)



(B)

Fig. 2. Iron (Fe) content in blood plasma of mice treated with low or high daily doses of $CoCl_2$ (A), and CoEDTA (B)

Conclusions

The effect of chronic exposure to cobalt(II) depends on the type of compound used, dose, time duration as well as on the age of the experimental animals. d18 mice are more sensitive to Co(II) treatment. High doses of $CoCl_2$ enhanced hemoglobin biosynthesis compared to CoEDTA. Higher plasma Fe concentration was measured in samples of mice treated with Co-EDTA which corresponded to low Hb concentration.

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