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# The Effect of Cobalt Chloride and Chromium Chloride on Development of Mouse Liver during Pregnancy

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**ABSTRACT:** Cobalt ($Co^{2+}$) is required as an essential trace element to a variety of metabolic functions such as the synthesis of vitamin B12 in mammals. It was proven that cobalt passes through the placenta to the fetus**.** Cobalt possesses toxic effect on embryo. Also, Chromium ($Cr^{3+}$) as an essential element for both animal and human nutrition has antioxidative capacity. In this study we examined the effects of exposure to cobalt chloride and chromium chloride on development of mouse liver during pregnancy. **Methods:** Pregnant mice received intraperitoneally injection of cobalt chloride and chromium chloride at the concentrations of Co10, Co40, Cr 50, Co10 Cr50, Co40 Cr50 mg/Kg bw on day 14 pregnancy. A group did not receive any treatment and served as control group. After pups delivery, female pups were sacrificed on days 21 and 60 (P21 and P60) and then liver were removed, weighed and examined. **Results:** Cobalt chloride groups (10 and 40 mg/kg bw) caused a significant decrease in body and liver weights at P21 and P60 ($p<0.05$**)**. Light microscope investigations showed that only Co40 mg/Kg bw decreased number of uninucleate hepatocytes and increased number of binucleate hepatocytes and kupffer cells at P21 and P60 ($p<0.05$**)**. In addition, size of hepatocyte and their nuclear increased at P21 and P60 only after exposure to Co40 mg/Kg bw ($p<0.05$**)**. Co-administration of Co40 mg/Kg bw and Cr 50 mg/Kg bw to mice showed mild histological changes at P21 and P60. Histologically, the Co40 mg/Kg bw administration induced degenerative and developmental changes such as congestion of central vein, fatty changes and pyknotic nuclei in the liver of mice at P21 and P60. **Conclusion:** Comparative pathology of treated groups show that cobalt chloride in high concentrations affects the development of liver and appears that chromium chloride ameliorates cobalt chloride hepatotoxicity but are not completely protective.

***Keywords*:** Cobalt Chloride, Chromium Chloride, Development, Liver, Mouse, Pregnancy.

## INTRODUCTION

Heavy metals are also known as pollutants of environment with deleterious effects on human health due to their wide use in various branches of industries. They exist everywhere, in the air, water and soils (Boguszewska and Pasternak, 2004). Environmental pollutants such as cobalt is a relatively rare trace element that is essential for the formation of vitamin $B_{12}$ (hydroxycobalamin). However, cobalt at higher concentrations is toxic to humans, terrestrial and aquatic animals, and plants. Although the general toxicity of cobalt has been proven, the molecular basis of toxicity is not clear yet. (Lison et al., 2001; De Boeck et al., 2003). at high doses, it exhibits diverse effects in several tissues like heart, spleen, kidney and liver (ATSDR, 2001).

The liver is a major organ involved in metabolic homeostasis as well as drug and chemical detoxification during the fetal period and after birth (Millward-Sadler, 1987; Sato et al, 2004 ). Due to its role in the transformation of environmental xenobiotics, the liver is at great risk of injury, where high intracellular concentrations of such compounds can be reached (Davies and Portmann, 1987). The first stage of liver development occurs at approximately embryonic 8.0 in the mouse and results in endoderm specification to a hepatic fate (Kaufman and Bard 1999), after several stages, by embryonic 14 hepatoblasts are differentiating into hepatocyte and biliary cells

(Houssaint 1980; Le Douarin 1975). Final maturation and development of the liver is gradual and continues into the postnatal period.. The adult liver has a characteristic histological architecture (Apte et al. 2007). During histological examination of the postnatal liver, this characteristic liver architecture and development of the liver continue for 1–2 weeks after birth after birth (Apte et al. 2007).

Since, cobalt is transferred from mothers to the fetus via the placenta and to the neonates via the milk (Van Bruwaene et al., 1984; Byczkowski et al., 1994; Kratchler et al., 1998). Therefore, late pregnancy and early postnatal are critical periods to pups intoxication in liver (Domingo et al. 1985). Also, Cobalt is suspected to be toxic to many cell types, including neural cells (Yang et al., 2004) and can induce cell death by apoptosis and necrosis (Huk et al., 2004). Cobalt can cause DNA fragmentation (Zou et al., 2001), activation of caspases (Zou et al., 2002) and increases production of reactive oxygen species (ROS) leading to oxidative stress (Olivieri et al., 2001). These free radicals may lead to cellular damage when the rate of their generation overcomes the rate of their decomposition by antioxidant defense systems, such as superoxide dismutase (SOD), catalase (CAT), or reduced Glutathione (GSH) (Di Mascio et al., 1991; Mates et al., 1999; Datta et al., 2000).

On the other hand, the supplementation of antioxidants can be considered as the alternative method for therapy. In fact, several studies demonstrated that the cellular antioxidant activity is reinforced by the presence of dietary antioxidants (Kiefer et al., 2004; Khan et al., 2005). Accordingly, interest has recently grown in the role of natural antioxidants used as a strategy to prevent oxidative damage as a factor in the pathophysiology of various health disorders (Heikal et al., 2011; Kalender et al., 2010). Among antioxidants, trace mineral trivalent chromium ($Cr^{3+}$) is an essential nutrient involved in the regulation of carbohydrate, lipid, and protein metabolism via an enhancement of insulin action (Anderson, 1986, 1989, 1993). Mammals need $Cr^{3+}$ to maintain balanced glucose metabolism (Mertz, 1975), and thus chromium may facilitate insulin action (Nielsen, 1993, Vincent, 1999), and has an anabolic function (Evans, 1989). In addition to its beneficial effects on type 2 diabetes mellitus-related diseases, trivalent chromium protects against acute lethal hepatic damage induced by carbon tetrachloride, at least in part, through the scavenging of trichloromethyl radicals (Tezuka et al. 1991a,b).

Due to the toxic effects of cobalt chloride in high doses on the liver development, therefore the present study aimed to evaluate the hepatoprotective effect of chromium chloride when co-administered to female mice during late pregnancy using developmental alterations and histological findings as criteria in their offspring.

## MATERIALS AND METHODS

### *Animals and experimental design*

Experiments were performed on NMRI female mice weighing about 30 gr. Mice were housed at temperature 23±2°C and relative humidity of 40% with a 12-hour light and 12-hour dark cycle in the animal house , and received a standard mice chow with water ad libitum. After 1 week of acclimatization to the laboratory conditions, male and female mice were housed by pairs in each cage. Pregnant mice were inspected daily by the presence of the vaginal plug, which indicated day zero of pregnancy. Thirty-six Pregnant mice were randomly divided into six groups of 6 each: (1) did not receive any substance. (2) received 10 mg/kg bw cobalt chloride (3) received 40 mg/kg bw cobalt chloride (4) received 50 mg/kg bw chromium chloride (5) received 10 mg/kg bw cobalt chloride and 50 mg/kg bw chromium chloride (6) received 40 mg/kg bw cobalt chloride and 50 mg/kg bw chromium chloride intraperitoneally on day12-14 pregnancy. After pups delivery, female pups were divided into 2 groups: (1) 21days (36 pups), (2) 60days (36 pups), Each group were divided into 6 subgroups of 6 each that their mothers were administered Cobalt Chloride and Chromium Chloride intraperitoneally Treatments 1-6, respectively, at the concentrations of (1) 0, (2) 10 mg/kg bw cobalt chloride, (3) 40 mg/kg bw cobalt chloride (4) 50 mg/kg bw chromium chloride (5) 10 mg/kg bw cobalt chloride and 50 mg/kg bw chromium chloride (6) 40 mg/kg bw cobalt chloride and 50 mg/kg bw chromium chloride on day12-14 pregnancy, then pups were sacrificed on days 21 and 60 and livers were rapidly excised, weighed and processed for histological assays.

### *Histological assay*

Fresh portions of the left and medial lobes of the liver from each group were cut rapidly, fixed in neutral buffered formalin (10%), specimens were dehydrated, embedded in paraffin, sectioned at 5 um and stained with hematoxylin and eosin for histological examination (Bancroft and Stevens 1986).

### *Morphometric assay*

For morphometric analysis of H&E stained sections, hepatocyte and nuclei diameter and area measurements were performed using an Olympus research microscope connected to a Dino Capture Camera. The outline of each hepatocyte and nuclei was measured after taking an image with a 40X objective. Four randomly chosen section of each sample, incorporating both left and medial lobes, and ten randomly chosen fields/sections were considered for

measurement, so that a minimum of fifty hepatocytes per sample were measured (Shen et al., 2000). the counting of uni- and bi-nucleate cells and kupffer cells in five randomly chosen fields/sections were performed using an Olympus research microscope with a 40X objective and then other parameters obtained from these data with SPSS software.

***Statistical analysis***

Results were expressed as mean and standard errors of the means (SE) for treatment groups. Statistical analyses were performed with One-Way ANOVA and independent sample T-test for comparisons using of the Statistical Package for Social Sciences (SPSS Inc, Chicago, IL, USA) Version 17 software. P values of less than 0.05 were considered to indicate statistical significance.

## RESULTS AND DISCUSSION

***RESULTS***

***Body weight and liver weights***

Data of body weights and liver weights of both groups subjected to different treatments are shown in (Table 1) it was observed that treatment 3 achieved significant decreases (One-Way ANOVA; P<0.05) in body weights and liver weights compared to treatment 1 and other treatments. treatments 4,5 in comparison with treatment 1 were not significant. Also, the body weight and liver weights of treatments 2,6 significantly (One-Way ANOVA; P<0.05) lower than those of treatment 1, and the treatment 6 slightly restored these parameters. These results were similar in both groups.

Table1. Body and liver weights of different treatment groups.

| Groups | Treatments | body weight (gr) | Liver weight (gr) |
|---|---|---|---|
| Group1 | T1 | 12.66≠0.42[a] | 0.637≠0.049[a] |
| | T2 | 10.50≠0.34[b] | 0.501≠0.048[b] |
| | T3 | 8.16≠0.65[c] | 0.389≠0.012[c] |
| | T4 | 12.50≠0.34[a] | 0.621≠0.023[a] |
| | T5 | 12.33≠0.21[a] | 0.619≠0.035[a] |
| | T6 | 10.66≠0.21[b] | 0.503≠0.009[b] |
| Group2 | T1 | 26.33≠0.49[a] | 2.02≠0.40[a] |
| | T2 | 21≠0.36[b] | 1.21≠0.21[b] |
| | T3 | 17.16≠0.30[c] | 0.81≠0.08[c] |
| | T4 | 25.16≠0.47[a] | 1.78≠0.32[a] |
| | T5 | 25.50≠0.42[a] | 1.80≠0.48[a] |
| | T6 | 21.16≠0.54[b] | 1.27≠0.36[b] |

Group1: 21 days. Group2: 60 days.

T1,T2,T3 , T4,T5& T6: Treatment with 0 , co 10mg/kg wb/, co40 mg/kg wb,cr 50 mg/kg wb ,co10 mg/kg & cr5o mg/kg wb and co40 mg/kg & cro 50 mg/kg wb ;respectively.

Values are expressed as mean ± SEM a/d the mean difference is significant vs T1

***Morphometric observations***

The mean number of uninucleate hepatocytes had shown a decrease in treatments 2,3,6 and was significant in treatments 2,3,6 in comparison with treatment 1 (One-Way ANOVA; P<0.05), treatments 4,5 in comparison with treatment 1 were not significant, but was significant increase in treatments 4,5 in comparison with treatments 2,3,6 (One-Way ANOVA; P<0.05), These results were similar in both groups (Table2).

The mean of other parameters such as number of binucleate hepatocytes and kupffer cells, binucleate/all hepatocytes ratio, diameter and area of hepatocytes and nuclei, area of cytoplasm and nuclear/cytoplasmic ratio had shown a increase in treatment 3 in both groups, and was significant in treatment 3 in comparison with treatment 1 (One-Way ANOVA; P<0.05) (Tables 2, 3, 4 ).

The results presented in Tables 2, 3, 4 demonstrate the mean number of kupffer cells, diameter and area of hepatocytes and area of cytoplasm in both groups were not significant between treatments 2,4,5 and treatment 1, but was significant decrease in treatments 2,4,5 in comparison with treatments 3,6 (One-Way ANOVA; P<0.05).

The data presented in Tables 2, 3, 4 show the mean number of binucleate hepatocytes, binucleate/all hepatocytes ratio, diameter and area of nuclei, and nuclear/cytoplasmic ratio in both groups were not significant between treatments 4,5 and treatment 1, but was significant decrease in treatments 4,5 in comparison with treatments 2,3,6 (One-Way ANOVA; P<0.05).

The mean of all parameters in group1 had shown a significant difference in comparing with group2 (T-test; $P<0.05$).

Table 2. The mean number of uninucleate hepatocytes, binucleate hepatocytes, kupffer cells and binucleate/all hepatocytes ratio in liver of different treatment groups.

| Groups | Treatments | number of uninucleate hepatocytes | number of binucleate hepatocytes | binucleate/all hepatocytes ratio | number of kupffer cells |
|---|---|---|---|---|---|
| Group1 | T1 | 125.62±2.47[a] | 6.25± 0.26[a] | 0.05± 0.002[a] | 7.35±0.28[a] |
| | T2 | 120.04±1.19[b] | 8.62± 0.31[b] | 0.07± 0.002[b] | 8± 0.34[a] |
| | T3 | 86.04± 1.67[c] | 14.37 ± 0.68[c] | 0.16± 0.007[c] | 12.95± 0.54[b] |
| | T4 | 129.41±1.66[a] | 6.62± 0.31[a] | 0.05 ± 0.002[a] | 7.04± 0.30[a] |
| | T5 | 127.58± 2.30[a] | 6.83± 0.36[a] | 0.05± 0.003[a] | 7.58± 0.34[a] |
| | T6 | 108.29± 1.88[d] | 10.41± 0.50[d] | 0.08± 0.004[d] | 9.70± 0.49[c] |
| Group2 | T1 | 77.58± 0.63[a] | 8.75± 0.32[a] | 0.11± 0.004[a] | 9± 0.49[a] |
| | T2 | 71.41± 1.88[b] | 9.87± 0.53[b] | 0.13± 0.007[b] | 9.37± 0.46[a] |
| | T3 | 45.08± 0.80[c] | 15.75± 0.61[c] | 0.35± 0.01[c] | 13.04± 0.46[b] |
| | T4 | 80.08± 1.16[a] | 7.58± 0.44[a] | 0.09± 0.005[a] | 8.04± 0.35[a] |
| | T5 | 78.25 ± 1.55[a] | 7.91± 0.42[a] | 0.10±0.006[a] | 8.20± 0.34[a] |
| | T6 | 59.66± 0.53[d] | 11.20± 0.43[d] | 0.18 ± 0.007[d] | 10.41± 0.42[c] |

Group1: 21 days. Group2: 60 days.

T1,T2,T3 , T4,T5& T6: Treatment with 0 , co 10mg/kg wb/, co40 mg/kg wb,cr 50 mg/kg wb ,co10 mg/kg & cr5o mg/kg wb and co40 mg/kg & cro 50 mg/kg wb ;respectively.

Values are expressed as mean ± SEM a/d the mean difference is significant vs T1 .

Table 3. The mean diameter of hepatocytes and nuclei in liver of different treatment groups.

| Groups | Treatments | diameter of hepatocytes (um) | diameter of nuclei (um) |
|---|---|---|---|
| Group1 | T1 | 19.37±0.06[a] | 6.53±0.01[a] |
| | T2 | 19.41±0.04 [a] | 6.60±0.009[b] |
| | T3 | 20.26±0.04[b] | 7.07±0.0.01[c] |
| | T4 | 19.34±0.04[a] | 6.48±0.009[a] |
| | T5 | 19.44±0.03[a] | 6.50±0.01[a] |
| | T6 | 19.57±0.04[c] | 6.71±0.01[d] |
| Group2 | T1 | 23.68±0.10[a] | 7.89±0.05[a] |
| | T2 | 23.73±0.04[a] | 8.09±0.02[b] |
| | T3 | 24.62±0.04[b] | 8.71±0.02[c] |
| | T4 | 23.65±0.06[a] | 7.86±0.03[a] |
| | T5 | 23.75±0.04[a] | 7.88±0.03[a] |
| | T6 | 23.97±0.06[c] | 8.21±0.02[d] |

Group1: 21 days. Group2: 60 days.

T1,T2,T3 , T4,T5& T6: Treatment with 0 , co 10mg/kg wb/, co40 mg/kg wb,cr 50 mg/kg wb ,co10 mg/kg & cr5o mg/kg wb and co40 mg/kg & cro 50 mg/kg wb ;respectively.

Values are expressed as mean ± SEM a/d the mean difference is significant vs T1

Table 4. The mean area of hepatocytes, nuclei, cytoplasm and nuclear/cytoplasmic ratio in liver of different treatment groups

| Groups | Treatments | area of hepatocytes ($um^2$ ) | area of nuclei ($um^2$ ) | area of cytoplasm ($um^2$ ) | nuclear/cytoplasmic ratio |
|---|---|---|---|---|---|
| Group1 | T1 | 296.90±1.97[a] | 33.66±0.19[a] | 262.60±1.97[a] | 0.1299±0.001[a] |
| | T2 | 296.92±1.36[a] | 34.29±0.09[b] | 263.260±1.33[a] | 0.1324±0.0007[b] |
| | T3 | 323.58±1.40[b] | 39.40±0.16[c] | 284.18±1.40[b] | 0.1409±0.0009[c] |
| | T4 | 294.70±1.33[a] | 33.21±0.09[a] | 261.59±1.30[a] | 0.1282±0.0006[a] |
| | T5 | 297.53±1.19[a] | 33.32±0.15[a] | 264.21±1.15[a] | 0.1279±0.0007[a] |
| | T6 | 301.89±1.35[c] | 35.57±0.16[d] | 270.31±1.30[c] | 0.1351±0.0008[b] |
| Group2 | T1 | 442.95±3.82[a] | 49.86±0.67[a] | 392.27±3.64[a] | 0.1375±0.001[a] |
| | T2 | 443.79±1.70[a] | 51.68±0.28[b] | 393.93±1.65[a] | 0.1404±0.0008[b] |
| | T3 | 477.04±1.71[b] | 59.89±0.37[c] | 417.15±1.56[b] | 0.1487±0.0009[c] |
| | T4 | 440.94±2.53[a] | 49.08±0.49[a] | 391.85±2.55[a] | 0.1363±0.001[a] |
| | T5 | 443.97±1.81[a] | 49.14±0.37[a] | 394.83±1.79[a] | 0.1359±0.001[a] |
| | T6 | 452.42±2.27[c] | 53.16±0.34[d] | 403.26±2.18[c] | 0.1433±0.001[b] |

Group1: 21 days. Group2: 60 days.

T1,T2,T3 , T4,T5& T6: Treatment with 0 , co 10mg/kg wb/, co40 mg/kg wb,cr 50 mg/kg wb ,co10 mg/kg & cr5o mg/kg wb and co40 mg/kg & cro 50 mg/kg wb ;respectively.

Values are expressed as mean ± SEM a/d the mean difference is significant vs T1

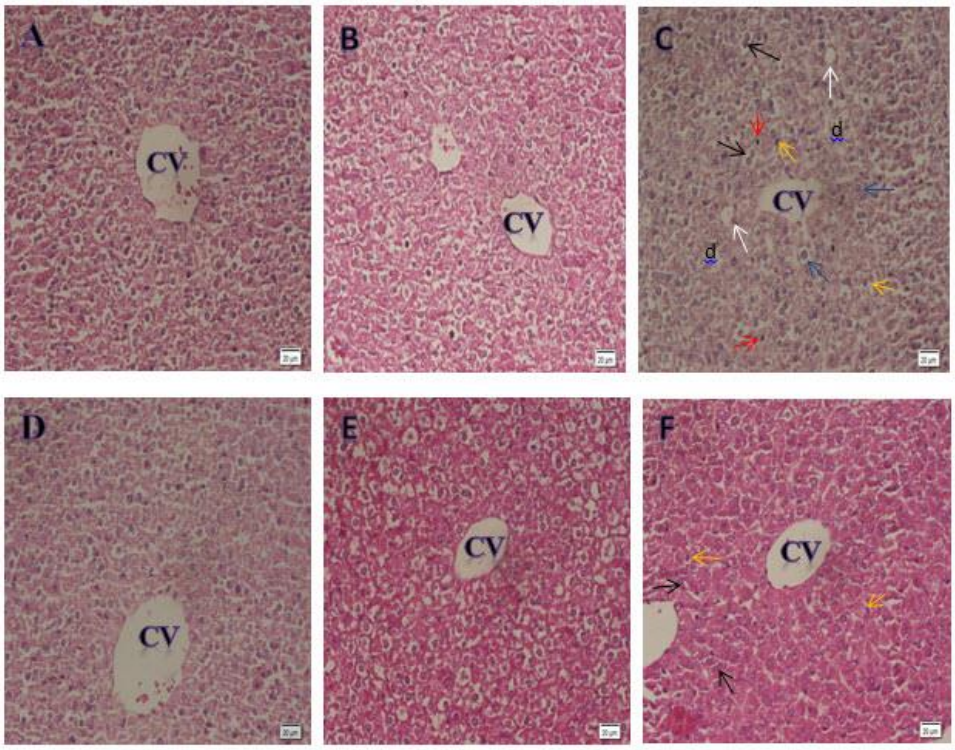


Figure 1. Photomicrograph of H&E stained sections of liver from treatment mice 21 days showing : (A) Treatment 1: normal structure. (B, D and E) Treatment 2,4,5: almost the normal appearance of hepatocytes around the central vein (cv). (C) Treatment 3: fatty changes (white arrow), degeneration (d) in hepatocytes, increasing in kupffer cell (orange arrow) between the degenerated hepatocytes, binucleation (black arrow) , apparent increase in nuclei size (blue arrow), pyknotic nuclei (red arrow), congestion of central vein. (F) Treatment 6: a mild degree of lesions, binucleation and kupffer cells proliferation.

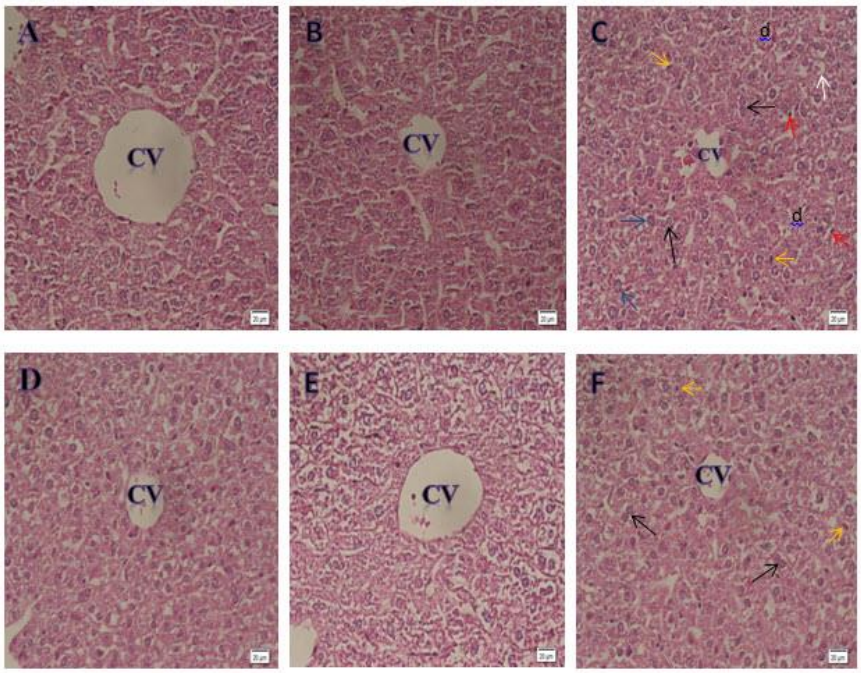


Figure 2. Photomicrograph of H&E stained sections of liver from treatment mice 60 days showing : (A) Treatment 1: normal structure. (B, D and E) Treatment 2,4,5: almost the normal appearance of hepatocytes around the central vein (cv). (C) Treatment 3: fatty changes (white arrow), degeneration (d) in hepatocytes, increasing in kupffer cell (orange arrow) between the degenerated hepatocytes, binucleation (black arrow) , apparent increase in nuclei size (blue arrow), pyknotic nuclei (red arrow), congestion of central vein. (F) Treatment 6: a mild degree of lesions, binucleation and kupffer cells proliferation

***DISCUSSION***

As a class of toxic factors, metals are a concern of the highest priority for human exposure. Exposure to an excessive amount of cobalt can have deleterious effects on the human body several tissues like liver (ATSDR, 2001). The late fetal, the neonatal and the late suckling period have been identified as periods of enzymatic differentiation in the liver, hence the importance of cytological changes during these periods. Exposure to toxic stimuli during this period of functional and structural heterogeneity can adversely influence various morphological and functional parameters, thereby increasing morbidity (Alexander et al., 1997). In addition, It is well known that maternal nutritional status has a significant influence on embryonic development and can be an important modulator of the toxicity induced by several agents (Keen et al., 1997). Experimental and clinical evidence have shown that several antioxidants have an impact on health outcomes following metals' exposure (Tarasub et al, 2012).

This study showed hepatotoxicity of cobalt 40 mg/kg bw and the effect of antioxidants agents such as trivalent chromium for modulation this toxicity. the antioxidant effect of chromium has been demonstrated in some experimental and clinical studies (Cheng et al. 2004; Atac et al. 2006).

In our experimental study, exposure of female mice to cobalt 40 mg/kg bw during pregnancy period induced a marked decrease in Body and liver weights of their offspring. These findings were consistent with previous reports, which demonstrated that exposure of female rats to cobalt during pregnancy and early postnatal periods induced a marked decrease in food consumption and in water intake associated with a concomitant decrease in body and liver weights of their offspring (Garoui et al., 2011). Our results suggest that perinatal exposure of adult female mice to cobalt 40 mg/kg bw delays the growth of their pups due to the transfer of $Co^{2+}$ through both placenta and milk. Furthermore, growth perturbations of pups could be explained either by a malabsorption of nutrients from the gastrointestinal tract and an impaired feed conversion efficiency as reported by (Ball and Chhabra, 1981). On the other hand, cobalt 40 mg/kg bw may induce oxidative stress leading to generation of free radicals and alterations in antioxidant status or ROS which cause metabolic disorder and weight loss. For this reason, treatment with antioxidants (trivalent chromium) and free radical scavengers can decrease the oxidative stress and improve metabolic process of cobalt treated mice and induced a partial recovery of body and liver weights of offspring. These results are consistent with many previous investigators with antioxidants such as propolis, vitamin E and selenium (Garoui et al., 2012; El-Desoky et al., 2012).

Our results showed that cobalt 40 mg/kg bw caused a increase in number of binucleate cells. Binucleation seen in this study might represent a consequence of cell injury and is a sort of chromosomes hyperplasia which is usually seen in regenerating cells (Gerlyng et al., 2008). It has been also reported that acytokinesis or the defective cytokinesis of mononuclear cells or the fusion of two mononuclear cells was suggested as the cause of the binucleation, which could be important cells in the genesis of tetraploid hepatocytes. Liver cell polyploidation is considered an original physiological process: cells pass through a binucleation step which enables them to increase metabolic output and constitutes an alternative to cell division (Guidotti et al., 2003).

In the present study, cobalt 40 mg/kg bw exposure resulted in a decrease in number of uninucleate hepatocytes and increase binucleate/all hepatocytes ratio. This is in partial agreement with another study (Altunkaynak et al., 2009) which reported Rats fed high-fat diets have shown higher numerical density of binucleate hepatocytes, presumably as a compensatory response to the decreased number of uninucleate hepatocytes following necrotic changes.

The data of the present study showed that cobalt 40 mg/kg bw activates the phagocytic activity of the sinusoidal cells by increasing the number of Kupffer cells. Similar findings were reported by other investigators (Jarrar and Taib., 2012). This might be a result of increased autophagy throughout the hepatic tissue to help in removing the accumulated cobalt and its metabolites where lysosomes are involved in the intracellular breakdown into small metabolic products. The produced Kupffer cells hyperplasia might be correlated with the amount of injury to the hepatic tissue induced by cobalt intoxication and represents a defense mechanism of detoxification and might be contributed to hepatic oxidative stress (Neyrinck, 2004).

The results of this study showed that cobalt 40 mg/kg bw may make some changes such as increasing in the diameter and area of hepatocytes and nuclei, which can be the result of hyperactivity of the hepatocytes and nuclei due to damage. With regard to this subject, the other scientists have reported that exposure of postnatal rats to sodium arsenite caused a increase in the diameter and area of hepatocytes and nuclei (Bhattacharya et al, 2012).

The current study demonstrated that cobalt 40 mg/kg bw administration caused fatty changes, degeneration in hepatocytes, increasing in Kupffer cell between the degenerated hepatocytes, binucleation, pyknotic nuclei, congestion of central vein. These observations indicated marker changes in the overall histoarchitecture of liver in response to cobalt 40 mg/kg bw, which could be due to its toxic effects primarily by the generation of reactive oxygen species causing damage to the various membrane components of the cell. Our results are supported by other studies conducted on other metals and toxins (Tarasub et al, 2012, Heikal T et al, 2012, El-Desoky G et al, 2012).

The co-treatment of trivalent chromium improved the histological alterations induced by cobalt 40 mg/kg bw, which could be attributed to the antiradical/antioxidant efficacy of this metal. Moreover, these results are in good accordance with those obtained by other studies which have postulated the beneficial role of trivalent chromium and other antioxidants on histological changes (Combs GF et al ,1984, McPherson, 1994, Chen W, 2009).

## CONCLUSION

This study demonstrates that administration of cobalt chloride (40 mg/kg bw) to female mice during late pregnancy period leads to hepatic toxicity which represents as developmental and histolgical changes in their offspring. Administration of chromium chloride as potent antioxidant can partially quench the deleterious effects of chronic toxicity of cobalt chloride.

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