STUDTY OF TOXICOPATHOLOGICAL EFFECTS OF SODIUM DICHROMATE ON *Rattus norvegicus*

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(Received 28December 2014 ,Accepted 29 March 2015)

Keywords: Sodium dichromate, T2 , Rats

ABSTRACT

In order to evaluate the toxic pathological effect of sodium dichromate, a (30) rats were exposed to sodium dichromate in daily oral dose, these rats divided into 3 groups based on two doses T1 group (3mg/kg bw) and T2 group (9mg/kg bw) in addition to control group C were given distal water orally at the same time of experiment, The animals were sacrificed after (30 and 60) days of administration, then detected the toxicological effects of sodium dichromate by using body weight gain and pathological changes, the results showed significant decrease in body weight of T2 than T1 and control groups respectively. In addition, the pathological changes appear more sever at T2 than T1 compared with control group.

INTRODUCTION

Sodium dichromate is one of a number of inorganic compounds containing hexavalent chromium (CrVI) found in drinking water source supplies as a contaminant resulting from various industrial process including electroplating operations, leather tanning, and textile manufacturing [1]. Sodium dichromate also has many direct uses as an ingredient in the production of metal finishing: aids corrosion resistance, helps clean metal surfaces and promotes paint adhesion ; organic products: used as an oxidizing agent in the manufacture of products such as vitamin K and wax ; pigments: used in the manufacture of inorganic chromate pigments where it produces a range of light stable colors. Also some chromate grades are used as corrosion inhibitors in undercoats and primers; ceramics: used in the preparation of colored glass and ceramic glazes; textiles which used as a mordant for acidic dyes to

improve their color-fast properties chrome sulphate production. Chromium (VI) is more readily absorbed [2]. In the environment, Cr (III) is generally immobile in soil and is not very toxic to plants and animals [11], whereas Cr (VI) is both mobile and toxic. Chromium (VI) in solution exists as hydrochromate (HCrO4-), chromate (CrO42-), and dichromate (Cr2O72-) ionic species [12] and reacts over time to form Cr (III) [3].

Humans and animals localize chromium in the lung, liver, kidney, spleen, adrenals, plasma, bone marrow, and red blood cells (RBC) [4].There is no evidence that chromium is biotransformed, but Cr (VI) does undergo enzymatic reduction, resulting in the formation of reactive intermediates and Cr (III) [5]. The main routes for the excretion of chromium are via the kidneys/urine and the bile/feces [6].Animal studies show that Cr (VI) is generally more toxic than Cr (III), but neither oxidation state is very toxic by the oral route. Compounds of both Cr (VI) and Cr (III) have induced developmental effects in experimental animals that include neural tube defects, malformations, and fetal deaths [7].

MATERIALS AND METHODS

A thirty adult albino rats (*Rattus norvegicus*) of both sexes, (150-250) grams were used in this study. These rats divided randomly and equally into 3 groups in which each contained 10 rats, consist of T1 group dosed with sodium dichromate (3mg/kg B.W.) and T2 group (9mg/kg B.W.) while the control group was given distal water orally daily for 60 days. The animals were raised and bred in the animal house of College of Veterinary Medicine / Baghdad University where the research was done. The animals were kept in cages of ($20\times30\times50cm^3$) dimensions in average of three rats in each cage for one month before study for acclimatization in optimum conditions housing at (22±3ºC) with a (14/10) Hours (Light/Dark) cycle .Standard rodent diet (Commercial feed pellets) and drinking water were given every seven days. Sodium dichromate (sigma) is the chemical compound with the chemical formula Na2Cr2O7 was provided by the Biochemistry Dept./Veterinary Medicine Collage / University of Baghdad.

RESULTS AND DISCUSSION

Table-1 revealed variable values of body weight gain in rats of sodium dichromate (SDC) treated group T1(3mg/kg B.W.),T2 group (9mg/kg B.W.) and control group along the period of study. In SDC treated groups T1 ,T2 and control group at (zero day experiment/pretreatment) results were (147.8±1.9, 147.2±2.61,149.5±1.26) respectively, and at (30days treatment) group T2 were significantly($P<0.05$) less than in T1 and control group (171.6±1.24,178.4±1.22,214.2±2.62)respectively. Also at (60days post treatment) in animals of T2 were less than in T1 and control groups (187.7±2.42, 195.2±1.28 and 255.6±2.76) respectively.

Table (1): Effects of SDC in Toxicology experiment for both male and female rats.

Dosed	Days of treatment		
	zero day of treatment	30 days of treatment	60 days of treatment
T1 3mg/kg b.w	147.8±1.9 a C	178.4±1.22 c B	195.2±1.28 c A
T2 9mg/kg b.w	147.2±2.61 a C	171.6±1.24 b B	187.7±2.42 bA
Control (C)	149.5±1.26 aC	214.2±2.62 aB	255.6±2.76 aA

T1/ 30days and 60 days: rat dosed SDC30 days and 60 days

T2/30days ,60days: rat dosed 9mg/kg B.W. SDC for 30 days and 60days.

C/30daysand 60days: rat dosed distilled water for 30 days and 60days.

*** LSD=5.4 * (P<0.05).**

Pathological results

Clinical signs

The following clinical symptoms were recorded after sub chronic administration of SDC to T1,T2 groups which showed different symptoms(depression, slow movement (Fig. 1 and 2). In addition to anorexia, body weight loss, diarrhea, loss of hair (Alopecia), ruffled hair Fig. (1) the symptoms recorded were more sever in T2 than T1group and appear earlier compared with control one that showed no symptoms during the course of experiment.

Figure (1): Rat (groupT1/30days) with rough skin, loss and ruffled hair.

Figure (2): Rat (groupT2/60days); dull, and uncoordinated.

The histopathological changes were:

T1/ 30days group

Spleen: Their histological sections showed sever hemorrhage in red and white pulp with moderate hyperplasia of lymphoid follicles, presence of brown pigment (hemosedrin) engulfed by macrophage or diffusely (Figure-3).

Kidney: The cortical tissue showed; vacuolar degeneration of renal tubular lining epithelial cells sometimes appeared as star-shape, in other section most of the tubular lumen filled with hyaline casts and others were dilated (figure-4).The glomeruli showed distension of bowman´s space, atrophy of few of them and periglomerular of MNCs infiltration & perivascular cuffing also noted. In addition to pyelitis;

characterized by sloughing of pelvic mucosal transitional epithelia,infiltration of inflammatory cells and sever hemorrhage with congestion of blood vessels (Figure-4)

Liver: Showed degenerative changes of hepatocytes which appeared enlarged and swollen with narrowing of the sinusoidal spaces, congestion of hepatic blood vessels and central vein in addition to the thickening of portal areas due to congestion of blood vessel (portal vein) with perivascular infilteration of inflammertary cells mainly mononucller cells found also in lumen of congested blood vessel (Figure -5).

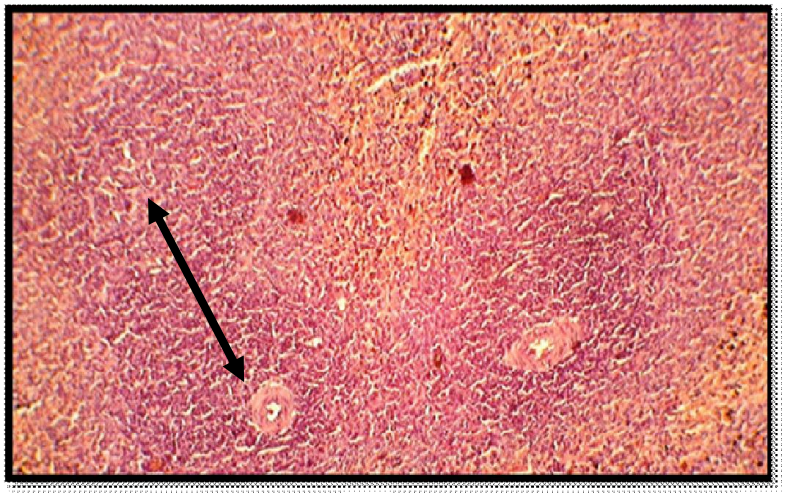


Figure (3): Histopathological section of spleen in (3mg /kg b.w/30days); moderate lymphoid hyperplasia (H&E stain, 40X).

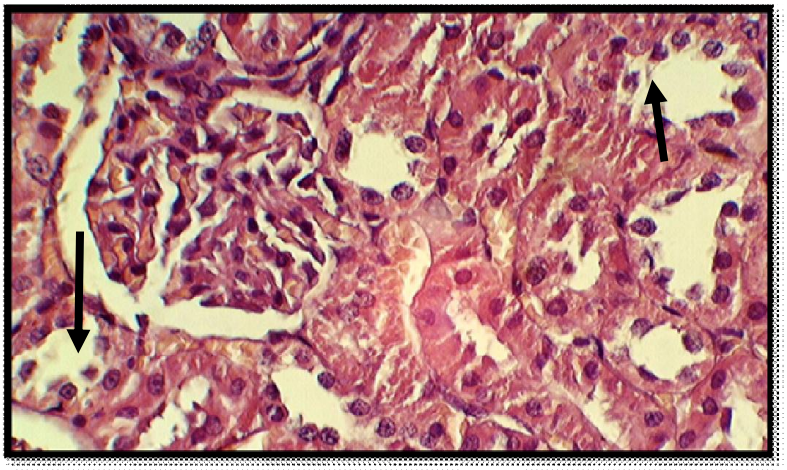


Figure (4): histopathological section in kidney of (3mg /kg B.W/30days). The cortical tissue showed cellular degeneration of renal tubular epithelium (40x).

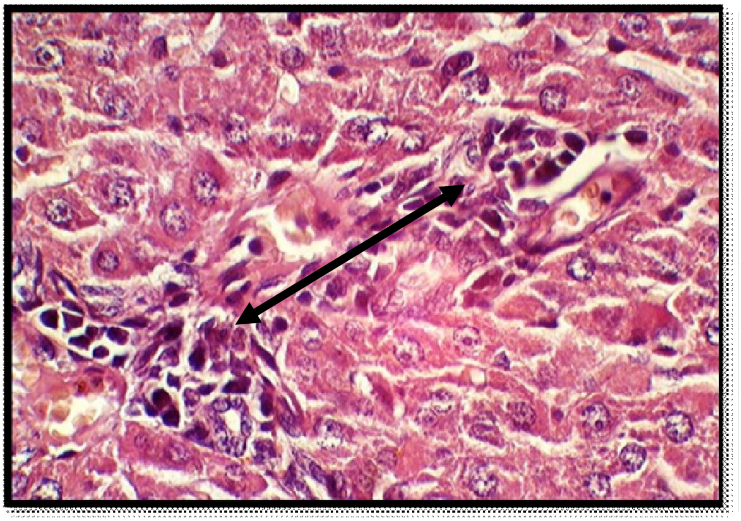


Figure (5): Histopathological section in liver of (3mg /kg b.w/30days) showed enlargment of hepatocytes and thickening of portal area due to congestion of blood vessel with infiltration of inflammatory cells mainly mononuclear cells (40x) .

T2: 9mg/kg B.W./30 days)

Spleen: There was a moderate lymphoid hyperplasia of white pulp and thickening of blood vessel wall due to hydropic degeneration of endothelial cells of tunica intenia and hypertrophy of tunica media, hemorrhage in red pulp, hemosiderine pigment engulfed by macrophage (Figure-6).

Liver: The histopathological lesions of showed few inflammatory cells (MNCs) in portal area and congestion of blood vessels with some inflammatory cells inside it, dilated sinusoid, focal area of necrosis and infiltration of inflammatory cells (granulomatous lesion which characterized by necrotic center surrounded by lymphocytes, macrophage and giant cells (Figure-7).

Kidney: The cortical part of kidney showed acute cell swelling of the epithelial lining cells of proximal tubules, in other section there were atrophic glomerular tufts and great destination of bowman´s space in addition to congestion of blood vessels and edema (Figure-8). Pyelitis also noted characterized by infiltration of (MNCs) in pelvic walls (sloughing of transitional epithelium and congestion of blood vessels, other section showed presence of hyaline casts in the collecting tubules and heavy infiltration of perivascular with periglomerular infiltration of inflammatory cells.

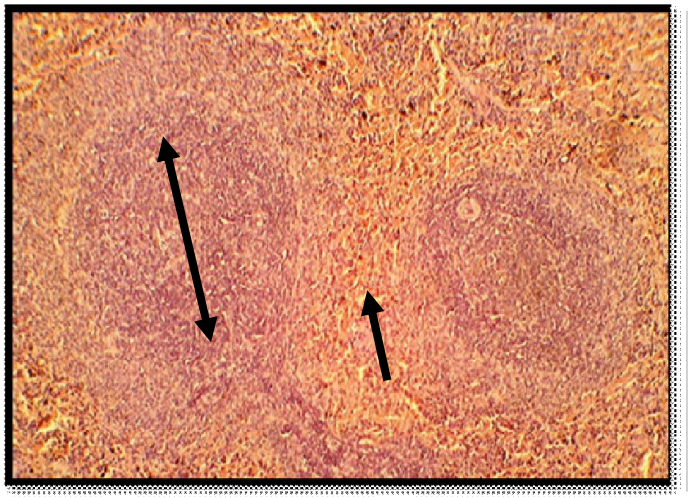


Figure (6): Histopathological section of spleen (9mg/kg b.w/30days); in white pulp appeared moderate lymphoid reactive hyperplasia and sever hemorrhage in red pulp (H&E stain, 10X).

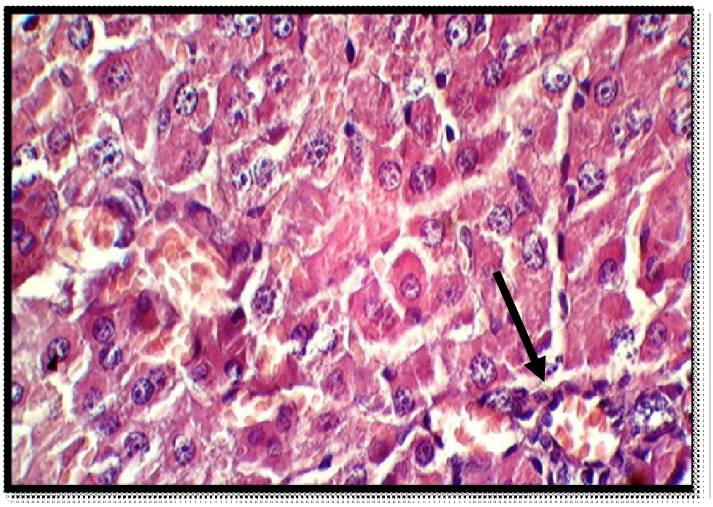


Figure (7): Histopathological section of liver 9mg/kg B.w/30days showed prevasculer mononuclear cells (MNCs) manily consist of (lymphocyte,macrophage). 40x H&E

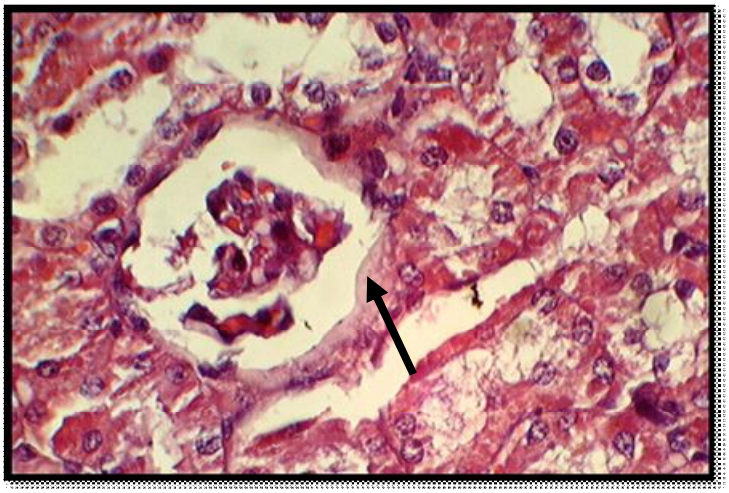


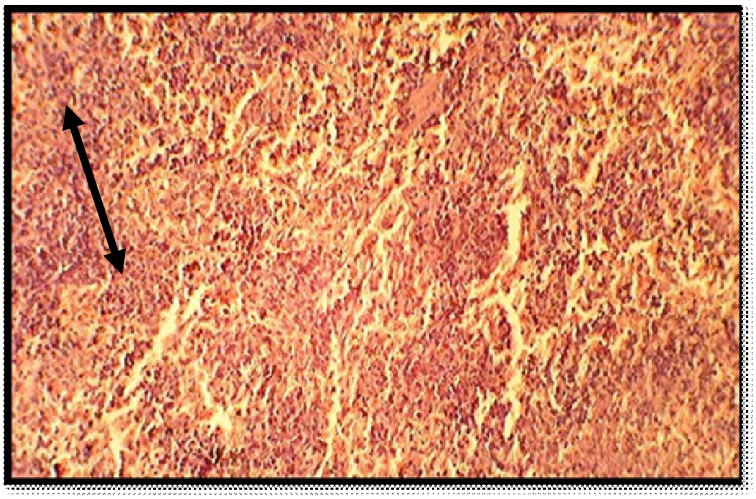
Figure (8): Histopathological section of kidney (9mg/kg B.w/30days). showed atrophy of glomerular tuft, increase bowman´s space ,with hyalinized capsular wall and necrosis of epithelial lining cells of proximal tubules 40x H&E.

T1: (3mg/60days males and females):

Spleen: there was moderate lymphoid hyperplasia of white pulp, hemorrhage in white pulp with hemosiderine pigment engulfed by macrophages or free.(Figure -10).

Kidney: perivascular infiltration of MNCs in the interstitial tissue between proximal renal tubules with congestion of blood vessels and edema. Acute cell swelling of tubular epithelial cells, congestion of renal blood vessels and glomerular tuft with vacuolar degeneration , some tubules showed necrosis and hyalinzation. (Figuer -11,12).

Liver: there were congestion of blood vessels and sinusoidal capillaries and portal blood vessels (dilated and filled with blood), few to moderate perivascular cuffing (mainly lymphocytic infiltration) in other section focal aggregation of MNCs were noted. The hepatocytes showed vacuolar degeneration (Figuer -13,14).



Figuer-9: **Spleen section (T1/60days) showed with moderate lymphoid hyperplasia of white pulp .10X H&E stain.**

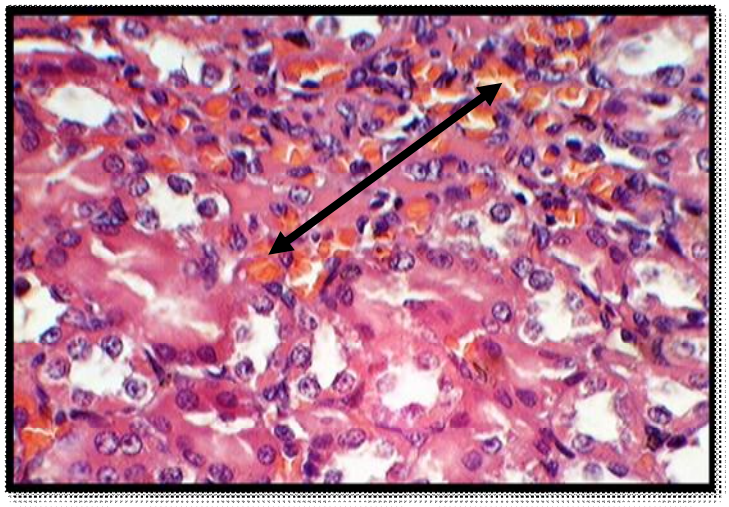


Figure -10: kidney section(T1/60days) showed: congestion of blood vessels and few infiltration of MNCs in interstitial tissue, degenerative lining epithelial cells of tubules also noted, (40x H&E stain,)

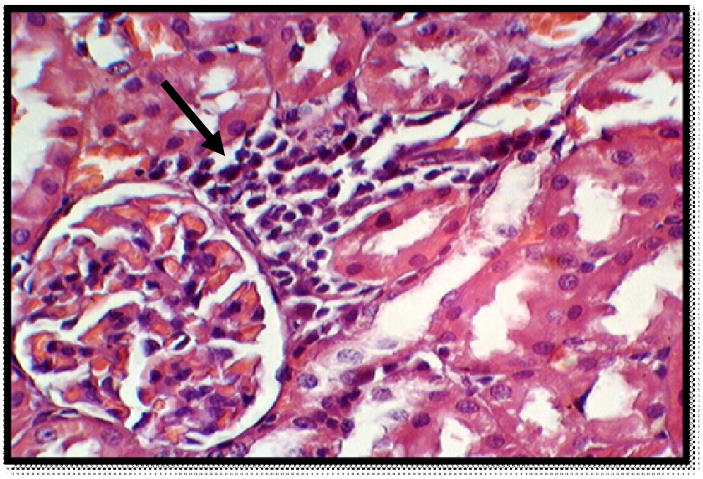


Figure-11: Kidney section (T1/60days) showed periglomerular infiltration of MNCs. (40X, H&E stain).

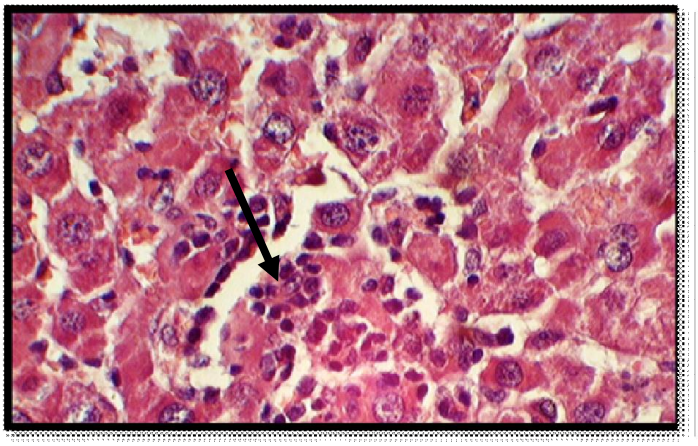


Figure -12: liver (T1/60days)showed: small granulomatous lesion consiste from necrotic cells and aggregation of MNCs .The hypatocytes showed vacuolar degeneration.(40x .H&E stain).

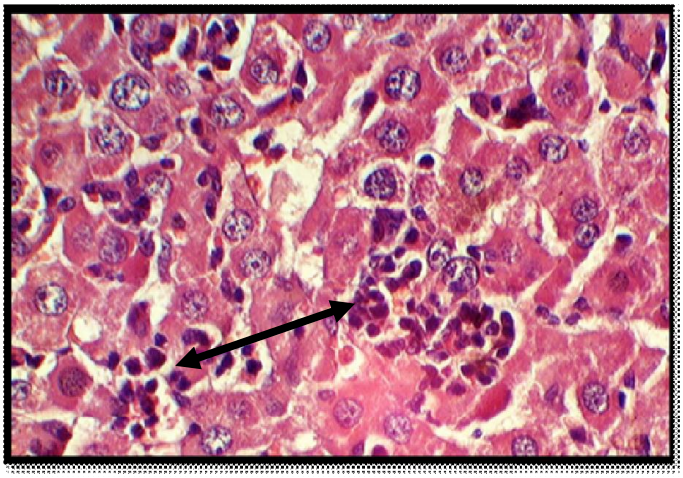


Figure-13: Liver **(T1/60 days) showed multiple small aggregation of MNCs and necrotic hepatocytes. (40X ,H&E stain).**

T2: 9mg/60days males and females);

Spleen: There was depletion of lymphoid follicles in white pulp also congestion and dilation of sinuse blood vessels, hemorrhage in the red pulp with hemosidrene pigment which appeared brown in color free in tissue and engulghed by macrophages. In other section moderate hyperplasia of malpujian corpusles noted Figuer-15 and16.
Liver; The hepatocytes showed necrosis and focal aggregation of MNCsin liver parenchyma seen, congested blood vessels and edema also seen causes narrowing of the sinosoidal cavities.Figuer-17.

Kidney: The lining epithelial cells of renal tubules appeared with acute cell swelling, congestion of blood vessels and edema, infiltration of inflammatory cells .figure -18.

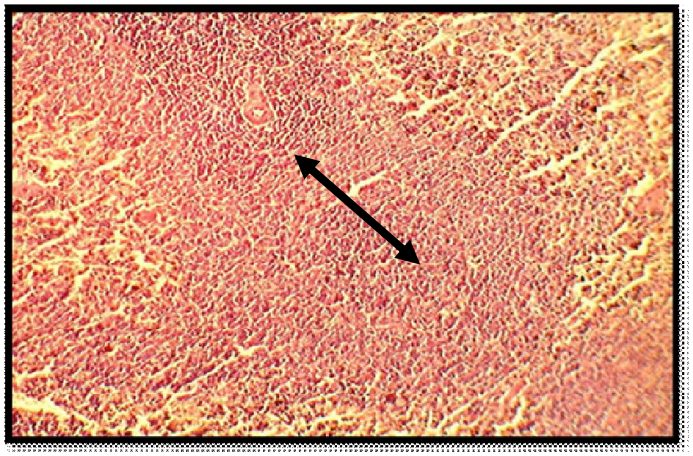


Figure-14: **Spleen T2/60days showed moderate hyperplasia of lymphoid follicles and sever hemorrhage in red pulp. (10X H&E stain).**

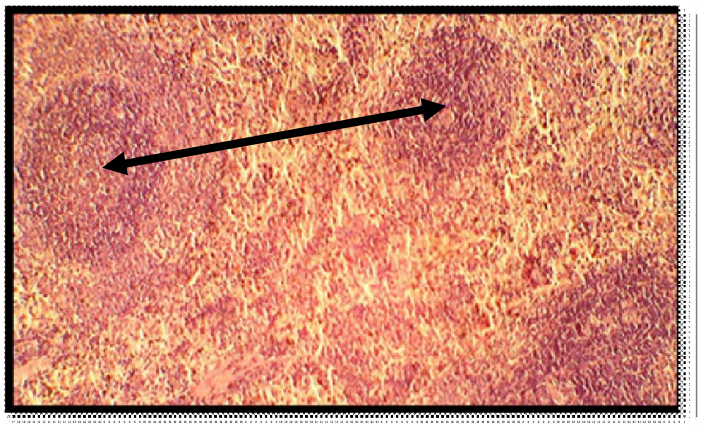
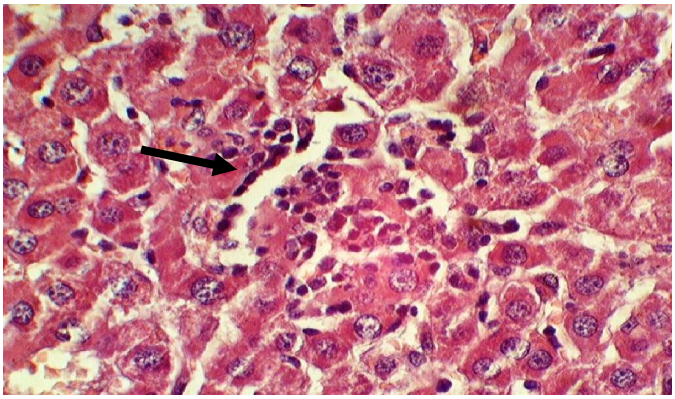
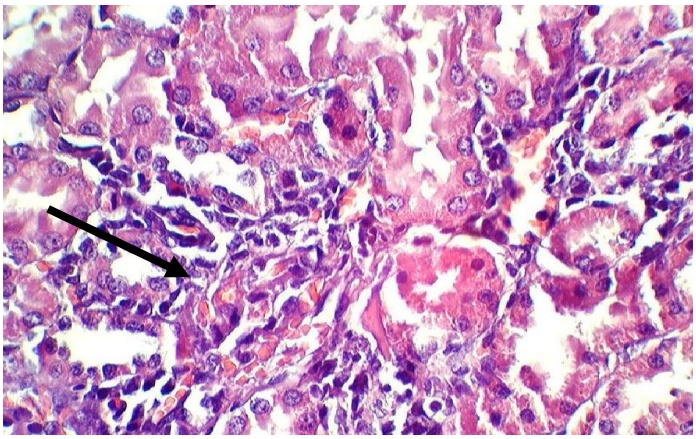


Figure-15: Spleen T2/60days showed moderate depletion of lymphoid follicles and sever hemorrhage in red pulp (10X H&E stain).



Figuer-16: liver section T2/60days with necrosis and focal aggregation of MNC's in liver.40X H&E stain.



Figuer- 17: kidney T2/60days showed lining epithelial cells of renal tubules appeared with acute cell swelling, congestion of blood vessels and edema, infiltration of inflammatory cells .40X H&E stain.

These clinical symptoms were agreed with **[8]**who believed that sodium dichromate cause immediate burning of mouth and throat, sever damage to the gut, heart, liver and kidneys. Chromium compounds are extensively spread in the body due to greater affinity of Cr(VI) to cross plasma membranes of the cell and because chromium (VI) unstable in the body and reduced to chromium (V), (IV) and (III) by endogenous substances such as ascorbate and glutathione which it is thought that the toxicity of chromium may resulted from damage to cellular components during this process through the generation of free radicals and due to depletion of main body antioxidants (glutathione and acerbate)**[9].** In Iraq ,at (Qarmat Ali facility) the reporter**[10]** said that chemical(sodium dichromate) was used as an antirust coating for pipes that supply water to the oil fields. After the 2003 US-led invasion, later the chemical was found strewn around the facility and its grounds. The mean clinical signs were seen

on soldiers were continuous bloody noses, spitting up of blood, coughing, irritation of the noses, eyes, throat, and lungs, shortness of breath.

While**[11]** showed that the oral administration of Cr(VI) at a dose of chronic and sub–chronic experiment does not appear to resulted in significant toxicological effects. While some studies had reported minimal or transient changes in body weight gain, hematological indices and the immune system. Other showed significant changes in hematological and immunity system **[12,13]**

The results of histopathological changes as seen in figures were agreed with **[14]** who reported necrosis in glomerulus, moderate necrosis in Bowman's capsule, necrosis in collecting tubule, degenerative changes in glomerulus and necrosis in haemopoietic tissue were observed in the fishes exposed to chromium in both acute and chronic periods. All these changes were more pronounced in 30 days chromium exposed fish**.[15]**reported that Cr (VI) could involve the kidney in many ways and cause different changes in it. Also **[16]** found that Cr(VI) could cause damage to kidney and liver due to its per oxidative damage which lead to reduced hepatic and kidney function that can be seen by the area of necrosis in kidney. And hepatocellular apoptosis after ingestion of water contaminated with high levels of Cr (VI).**[17,18]** reported that the hepatocytes appeared polyhedral in shape with no vacuolated cytoplasm, containing basophilic granules and central rounded vesicular nuclei with in treated liver rats which showed severe liver damage, including focal necrosis, congestion of blood vessels, increased lymphocytic infiltration around blood vessels, pyknotic nuclei, karyolysis, proliferation of Kupffer cells and bile ductless, and there were numerous vacuoles and sinusoids as well as distortion of hepatic cells .

[19] showed at their study the changes of spleen after oral administration of Cr (VI) were enlargement of the capsule and depletion of the red pulp cells, accompanied by an increase in macrophages. 24 h after injection. Partial restoration of red pulp was noted after 48 h.While **[20]**found no histological changes of spleen in all treated groups after oral administration of Cr(VI) . Degenerative, and necrotic changes were observed in brain, liver, and kidney. That what results of **[21]**showed after subchronic exposure to the metal mixture affected general health of male rats by altering the functional and structural integrity of kidney, liver, and brain.

Conclusions

we concluded he body that Sodium dichromate effected the body weights of rats at dose 9mg/kg B.W. more than that at 3mg/kg b.w. and that related with the clinical signs appeared (anorexia, body weight loss, diarrhea, loss of hair (Alopecia), ruffled hair) which were more sever atT2 that T1 group compare to control one. Also the histological changes at different organs (spleen, liver, kidney) were summarized by depletion of lymphoid follicles in spleen with hemosidrene pigment which appeared brown in color free in tissue and engulghed by macrophages. While hepatocytes showed necrosis and focal aggregation of MNCsin liver parenchyma , perivascular infiltration of MNC's in the interstitial tissue between proximal renal tubules with congestion of blood vessels and edema. Acute cell swelling of tubular epithelial cells, congestion of renal blood vessels and glomerular tuft with vacuolar degeneration , some tubules showed necrosis and hyalinization.

دراسه التاثيرات السميه المرضيه لثنائي كرومات الصوديوم في الجرذان البيضاء

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الخلاصه

يهدف البحث دراسه التاثير السمي والتغيرات النسيجيه (للكبد والطحال والكلى) لمادة ثنائي كرومات الصوديوم في الجرذان البيضاء حيث تم استخدام ٣٠ جرذي جرعت فمويا ويوميا بمادة ثنائي كرومات الصوديوم حيث قسمت لثلاث مجاميع اعتمادا على الجرع مجموعه الاولى T1 (٣ملغم/كغم وزن الجسم) والمجموعه الثانيه T2 (٩ ملغم/كغم وزن الجسم) بالاضافه لمجموعه السيطرة C التي جرعت الماء المقطر طيله فترة التجربه وتم التضحيه بالحيوانات بعد(٣٠-٦٠) يوم من التجربه وقد سجلت تغييرات في الوزن لمجموعه T2 مقارنه بمجموعتي التجربه الاولى والسيطرة وكذلك سجلت تغييرات نسيجيه وعيانيه لمجاميع التجربه مقارنه بمجموعه السيطرة .

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