



EFFECTS OF POTASSIUM DICHROMATE ON HAEMATOLOGICAL PARAMETERS IN FEMALE AND MALE *Wistar albino* RATS

Adjroud, O.

Laboratory of Animal Physiology, Department of Biology Sciences, Batna University, Algeria

E-mail : o.adjroud@caramail.com

ABSTRACT:

Hexavalent chromium is a potent toxic agent. It has been found to be carcinogenic in human and animal. The purpose of the current work is to compare the effect of potassium dichromate ($K_2Cr_2O_7$) using variations in the dose, route of administration, and duration of exposure in male and female *wistar albino* rats with special focus on hematological parameters. $K_2Cr_2O_7$ was administered either in the drinking water with a dose of 30 mg/l for 20 consecutive days to male *wistar albino* rats, or as a single dose subcutaneously (s.c) at 10, 50 and 100 mg/Kg body weight (b w) to female *wistar albino* rats. Control groups received NaCl 0.9% (0.3 ml s.c), or drinking distilled water. Haematological parameters were recorded on day 3, 6, and 21 after subcutaneous exposure, or on day 10 and 20 after oral treatment. 10 mg/Kg b w of $K_2Cr_2O_7$ given subcutaneously induced during the first three days a marked decrease in the number of erythrocytes (-6%) of leucocytes (-30%) of platelets (-48%) and of hematocrit values (-15%), while the number of granulocytes is augmented (+124%) in comparison with control. Hemoglobin concentration and lymphocyte counts decreased markedly on day 6 after exposure. Chromium 50 mg/Kg b w, s.c mainly affected during the first three days the leucopoietic indices inducing leucopenia (-55%), lymphopenia (-57%), monocytosis (+104%), granulocytosis (+204%), and thrombocytosis (+38%) if compared with control, while the erythrocytic counts and hemoglobin concentration decreased from day 6 (-22%) and (-21%) respectively until day 21 (-41%) and (-36%) respectively, and hematocrit values decreased at the end of experiment (-36%) in comparison with control. The higher dose of chromium (100 mg/Kg b w, s.c) reduced during the first three days the number of erythrocytes (-20%), platelets (-20%), total leucocytes (-55%), lymphocytes (-59%) and augmented the number of monocytes (+56 %), and granulocytes (+166%), while on day 6 the number of platelets augmented (+27%) in comparison with control. In drinking water, 30 mg/l of chromium given to male *wistar albino* rats had no effect on all erythropoietic parameters studied with the exception of the elevation (+21%) in platelet counts at the end of exposure, while the number of lymphocytes and total leucocytes were significantly reduced on day 20 after exposure (-37%) and (-37%) respectively. Conversely, the number of granulocytes and monocytes markedly increased on day 10 after exposure (+42%) and (+22%) respectively if compared with control. Short-term exposures to low dose of $K_2Cr_2O_7$, s.c induce in female *wistar albino* rats erythrocytopenia, thrombocytopenia, leucopenia, lymphopaenia, granulocytosis, monocytosis, and a decrease in hematocrit values and hemoglobin concentration while in drinking water chromium was susceptible to affect in male rats the immune response inducing leucopenia, lymphopenia, monocytosis, and granulocytosis, while this oral route of exposure had no effect on erythropoietic parameters.

INTRODUCTION:

Hexavalent chromium Cr (VI) is the major terrestrial pollutant. It is widely used in various industries, including pigments for manufacturing and painting, metal plating and leather tanning. Cr (VI) ingested with food such as vegetables or meat and water is reduced to Cr (III) before entering the blood stream (Richelmi and Baldi, 1984 and Kerger *et al.*, 1997). Chromium enters the body through the lungs, gastrointestinal tract, and to a lower extent through skin (Corbett *et al.*, 1997; De Flora *et al.*, 2006 and Antonini *et al.*; 2007). It is known that oral intake including food and water is the major route of exposure to chromium for the general population. Regardless of route of exposure Cr(III) is poorly absorbed whereas Cr(VI) is more readily absorbed (O'Flaberty *et al.*, 2001; Cavalleri and Minoia, 1985). Cr (VI) can easily enter the cell through SO_4^{2-} and HPO_4^{2-} channels (Valko *et al.*, 2006) and remains here for the life of the cell (Costa *et al.*, 1996). After entering the cell Cr (VI) undergoes a chain reaction with production of Cr intermediates such as Cr (V) and Cr (IV) by cellular reductants such as ascorbic acid and riboflavin, glutathione and serum protein (Standeven and Wetterhahn, 1992). The reduced product binds to intracellular proteins, resulting in an elevation of total chromium in the blood cell for several weeks (Costa *et al.*, 1996). During this reduction process, Cr produces reactive oxygen species (ROS) (Manerikar *et al.*, 2008), and generates oxidative stress. This in turn is responsible for defective hematopoiesis (Bainy *et al.*, 1995). It was established that Cr (VI) is a strong oxidant which causes cellular dysfunction and cell death (Vasant *et al.*, 2001; Wang *et al.*, 2006; De Flora *et al.*, 2006; Lei *et al.*, 2008 and Meyers *et al.*, 2008). The routes of excretion of chromium are

via kidney/urine and bile/feces (Barceloux, 1999). The purpose of the current work is to compare the effect of potassium dichromate ($\text{K}_2\text{Cr}_2\text{O}_7$) using variations in the dose, route of administration and duration of exposure in male and female *wistar albino* rats with specially focus on hematological parameters.

MATERIALS AND METHODS:

Animals:

Adult female and male *wistar albino* rats (Pasteur Institute, Algiers) were kept in a lighting schedule of 12 h light: 12 h darkness at $22\pm 1^\circ\text{C}$ with free access for food and water. Rats were housed at five rats per cage.

Chemicals:

Potassium dichromate ($\text{K}_2\text{Cr}_2\text{O}_7$) was purchased from Sigma Aldrich Laborchemikalien GmbH; NaCl was purchased from panacreaac Qu mica Sa, diethyl ether Fisher Scientific (UK).

Experiments:

Each animal was anaesthetized with diethyl ether s.c., and was weighed before each experiment. The controlled groups and treated groups were injected s.c with 0.3 ml/rat of NaCl 0.9%, or drinking distilled water.

Potassium dichromate ($\text{K}_2\text{Cr}_2\text{O}_7$) was dissolved in sterile saline (NaCl 0.9%) and was given as a single s.c. at 10, 50 and 100 mg/kg body weight to female rats or 30 mg/l in drinking distilled water to male rats. Blood sample was collected on EDTA from jugular vein for haematological study on day 3, 6 and 21 after subcutaneous injection and on days 10 and 20 for oral route. The determination of haematological parameters was performed by Coulter Erma Inc PCE-21-ON.

Statistical analysis:

Data for each group of experiments (n=6) were statistically analysed by analysis of variance and expressed as mean \pm S.E.M. Significant differences between the treated group mean and its control group were performed by Student's "t" test. Differences were considered to be significant if $P < 0.05$. Data were analysed with Excel for windows, version 5.1, USA.

RESULTS:

1-Effects of $K_2Cr_2O_7$ on erythropoietic parameters on female and male wistar rats:

Effects on erythrocytic counts:

In the female *wistar albino* rats, 10 mg/Kg b w of subcutaneous chromium induced slight but significant decrease ($p < 0.05$) in the erythrocytic counts in comparison with control. This decrease became no significant from 6 to 21 days after treatment, while 50 mg/Kg b w decreased progressively the erythrocytic counts from 3 to 6 days by 10% and 22% respectively and reached a maximum of 40% on day 21 after exposure (Table 1). 100 mg/Kg b w of Cr induced a significant diminution in erythrocytic counts during the experiment period by 20%, 32% and 10% respectively in comparison with control. On the contrary, the oral route (30 mg/l $K_2Cr_2O_7$) had no effect on the number of erythrocytes in male rats (Table 2).

Effects on hematocrit values:

On the other hand, the subcutaneous administration of $K_2Cr_2O_7$ at graded doses (10, 50 and 100 mg/kg b w) had no effect on the hematocrit values during the first three days after treatment while, on day 6 after exposure, 10 and 100 mg/Kg b w doses significantly decreased the hematocrit values by 20% and

16% respectively in comparison with the control, whereas, on day 21 after treatment, the hematocrit values were significantly reduced by 23% only with 50 mg/Kg b w in comparison with control (Table 1). 30 mg/l of orally $K_2Cr_2O_7$ had no effect on the hematocrit values in male *wistar albino* rats compared to the control (Table 2).

Effects on hemoglobin concentrations:

Similarly, the concentration of hemoglobin is slightly but not significantly decreased during the first three days after exposure to the graded doses of subcutaneously $K_2Cr_2O_7$, the decrease was highly significant from 6 to 21 days after treatment with the lower dose by 37% and middle dose by 24% respectively, compared to control, while, the higher dose decreased markedly the hemoglobin concentration by 23% only on day 21 after treatment compared with control (Table 1). In male *wistar albino* rats the oral route induced a negligible decrease in the hemoglobin concentration only on day 20 of treatment (Table 2).

Effects on blood platelets:

The graded doses of chromium induced a significant decrease in the number of blood platelets during the first three days after treatment by about 48%, 38% and 20% respectively, and on day 21 with 50 mg/Kg b w, of chromium sc, compared to control group in female *wistar albino* rats. While on day 6 the chromium induced a slight increase in platelet counts with the lower dose (+11%) and middle dose (+37%). This elevation in the number of platelets was highly significant (+27%) on day 21 after subcutaneous administration in comparison with control as shown in table 1. Similarly, in male *wistar albino* rats, 30 mg/l of chromium added to drinking water increased

progressively the platelet counts (+21%) on day 20 compared to control as shown in table (2).

2-Effects of $K_2Cr_2O_7$ on leucopoietic parameters on female and male wistar rats:

Effects on total leukocyte counts:

A significant decrease in the number of leukocytes was immediately observed during the first three days after exposure to graded doses of subcutaneous $K_2Cr_2O_7$ (10, 50, 100 mg/Kg b w) by 6%, 55% and 55% respectively in comparison with control. This decrease was maintained on day 6 with the middle and high doses by 47%, 20% and 76% respectively, while on day 21 a marked increase in the number of leukocytes was observed with the middle dose by 39% and the highest by 30% compared to 6 days after treatment (Table 3). In drinking water 30 mg/l $K_2Cr_2O_7$ significantly decreased the leukocyte counts from 10 to 20 days after treatment (Table 3).

Effects on lymphocyte counts:

10 mg/Kg b w of $K_2Cr_2O_7$ induced a significant decrease in the lymphocyte counts by 47% only on day 6 after subcutaneous treatment in female rats, while 50 and 100 mg/Kg b w, s.c immediately provoked a significant decrease on day 3 after exposure by 57% and 59% respectively. This diminution was only maintained with high dose by 27% on day 21 after treatment in comparison with control (Table 3). Male rats having received 30 mg/l of $K_2Cr_2O_7$ orally in drinking water showed a slight but not significant decrease in the lymphocyte numbers on day 10 after treatment,

this decrease became significant on day 21 and attained 37% in comparison with control (Table 4).

Effects on monocyte counts:

In female rats the monocyte counts augmented slightly but not significantly during exposure period with the lower dose of $K_2Cr_2O_7$ s.c, while 50 and 100 mg/Kg b w, s.c induced immediately a progressive increase in the number of monocytes by 104% and 56% respectively on day 3 and by 349% and 200% on day 6 after treatment, while on day 21 this increase was only maintained with 100 mg/Kg b w by 119% in comparison with control (table 3). In male rats 30 mg/l of $K_2Cr_2O_7$ in drinking water induced a marked increase in the monocyte counts by 424% on day 10 after treatment which disappeared on day 21 in comparison with control (Table 4).

Effects on granulocyte counts:

The number of granulocytes immediately augmented during the first three days after treatment with the graded doses of chromium administered subcutaneously to female rats by 124% (10 mg/Kg b w), 204% (50 mg) and 166% (100 mg) respectively. This increase was maintained from day 6 and was about 142%, 201% and 234% respectively, until day 21 with 10 mg/kg by about 46% and the higher dose by 48% compared to control values (Table 3). In drinking water, 30 mg/l of chromium induced a marked increase by 22% in the granulocyte counts on day 10 after treatment. This effect disappeared on day 21 after exposure compared to control values (Table 4).

Table (1): Effects of subcutaneous Chromium hexavalent erythropoiesis in female Wistar albino rats

Parameters		Control (n=6)	10mg/Kg (s.c)	50 mg/Kg (s.c)	100 mg/Kg (s.c)
Erythrocytes counts	Day 3	7.29 ± 0.38	6.84±0.043*	6.53±0.53	5.86±0.42*

(x10 ⁶ /mm ³)	Day 6	7.39 ± 0.28	5.74±1.37	5.79±0.10 *	5.03±0.45*
	Day 21	7.22±0.25	6.74±1.31	4.26±0.551**	6.46±0.57
Hematocrit values (%)	Day 3	39.88 ±4.18	34.04±1.04	36.5±1	36.16±3.2
	Day 6	38.36 ±1.63	30.54±0.48 *	35.07±0.4	31.98±2.8*
	Day 21	38.4 ±1.42	37.44±0.6	24.55±4.6**	34.95±1.97*
Haemoglobin concentrations (dl)	Day 3	16.40 ± 0.98	14.46±1.98	15.15±1.65	15.03±0.83
	Day 6	16.30 ± 0.67	10.34±0.16 *	12.90±0.81 *	16.40±0.98
	Day 21	16.20±0.42	12.20±0.21*	8.92±1.27**	12.46±0.73*
Platelets counts /x10 ³ mm ³	Day 3	1457 ± 102.32	758.40±316.8*	895.5±65.32*	1160.60±13.02*
	Day 6	168.33±45.65	1297±83.8*	1598.25±494.8	1489.66±181*
	Day 21	1114±27.9	1191.8±83	701.5±271.67*	1164.83±88

Each value erythropoiesis or body weight represents the mean ± SEM 6 rats per group **p<0.01, *p<0.05 compared with control value, student's t test.

Table (2): Effects of oral Chromium hexavalent on erythropoiesis in male Wistar albino rats

Parameters		Control (n = 6)	30 (mg/l)
Erythrocytes counts (x10 ⁶ /mm ³)	Day 10	6.95 ± 0.26	6.85±0.41
	Day 20	7.04±0.33	7.06±0.24
Hematocrit values (%)	Day 10	38.51 ±1.15	38.15±1.15
	Day 20	38.16±1.3	37.52±0.91
Haemoglobin concentrations (dl)	Day 10	13.33 ±0.39	13.2 ±0.37
	Day 20	13.6 ±0.43	12.66 ±0.35
Platelets counts X10 ³ /mm ³	Day 10	842.33±123	993.83 ±82.62
	Day 20	1080.83±123	1309.33±80*

Each value erythropoiesis or body weight represents the mean±SEM 6 rats per group *p<0.05 compared with control value, student's t test.

Table (3): Effects of subcutaneous Chromium hexavalent on leucopoiesis in female Wistar albino rats

Parameters		Control (n = 6)	10 mg/Kg (s.c)	50 mg/Kg (s.c)	100 mg/Kg (s.c)
Leucocytes counts/(mm ³)	Day 3	10433.33 ±2157	7320±894.02*	4650±851*	4700±596.93*
	Day 6	11566.66 ±96.45	6180±995	9225±551 *	2730±246.41*
	Day 21	11400±1137.24	9520±1073.19	15492±1888*	14836.6±2153.9*
Lymphocytes /(mm ³)	Day 3	8905 ±515.96	8580±1177.4	3850±526.6*	3633.33±348.8*
	Day 6	8153.33 ±403.55	6020±815.78 *	7750±526.6	5950±1285
	Day 21	8026.66 ±588	8920±960.72	8275±994	8133±92.62
Monocytes /(mm ³)	Day 3	276.5±76.77	376±101.58	565.5±184.3*	433.33±123*
	Day 6	288.5 ±144.31	420±167.33	1295±505.25*	866.66±66.66*
	Day 21	288.5±144.31	266±102.81	575±280.7	633.33±36.6*
Granulocytes /(mm ³)	Day 3	151.66±11.83	340±75.03*	462.5±77.89 *	403.33±107.51*
	Day 6	165±98.8	400±50 *	497.5±2.89*	551.66±58.97*
	Day 21	153.33±45.52	224±122.3	160±38.63	226.66±26.39

Each value leucopoiesis represents the mean±SEM 6 rats per group.

*p<0.05 compared with control value, student's t test.

Table (4): Effects of oral Chromium hexavalent on leucopoiesis in male Wistar albino rats

Parameters		Control (n = 6)	30 (mg/l)
Leucocytes/(mm ³)	Day 10	11013±482	9633±1096
	Day 20	11233±1364	7066.66±803.51*

Lymphocytes/(mm ³)	Day 10	9283.33±487.46	7700±1025.32
	Day 20	9216.66±709.26	5816±505.28*
Monocytes/(mm ³)	Day 10	250±68.13	1311±82.21 *
	Day 20	231.66±40.14	250±37.51
Granulocytes/(mm ³)	Day 10	506.83 ±3.59	616.66±11.35 *
	Day 20	466.66±73.22	533.33±119.2

Each value leucocytes represents the mean ± SEM 6 rats per group

*p<0.05 compared with control value, student's t test.

DISCUSSION:

The present study demonstrated that in female *wistar albino* rats, subcutaneous lower dose of hexavalent chromium affected immediately the erythropoietic parameters indicating anemia. In fact, the reduction in the number of erythrocytes, of the hematocrit values and platelet counts was immediately observed during the first three days after exposure to the lower dose of chromium, while hemoglobin concentrations decreased between day 6 and day 21. The middle dose on the contrary, later declined the number of erythrocytes, the hematocrit values and hemoglobin concentrations between day 6 and day 21, while the platelet number decreased only during the first three days after subcutaneous exposure. The higher dose decreased immediately the number of erythrocytes during the first six days, and the hematocrit values decreased only on day 6 while hemoglobin concentrations diminished at the end of exposure and platelet counts only on day 3 after exposure to subcutaneous treatment. We have also observed that on day 6, the graded doses of chromium used in the present study tend to augment progressively the number of platelets.

Short-term exposures to low concentrations of chromium inducing a decrease in erythropoietic indices were reported in fishes (Vutukuru, 2005) and in mice (Shrivastava *et al.*, 2005). This anemia could be due to iron deficiency and consequently to its reduced use

for hemoglobin synthesis. Red blood cell chromium is currently considered the best indicator of hexavalent chromium exposure (Costa *et al.*, 1996). It was reported earlier that Cr (VI) can penetrate rapidly the membrane of erythrocyte and enter the cell and accumulates in erythrocytes of exposed workers (Lewalter *et al.*, 1985; Minoia and Cavalleri, 1998 and Stridsklev *et al.*, 2004). The accumulation of Cr (VI) induced micronucleus frequency in erythrocytes of adult mice and their fetuses after intraperitoneal injection of Cr (VI) (De Flora *et al.*, 2006, 2008) and caused DNA-protein crosslink formation in erythrocytes of fishes (Kuykendall *et al.*, 2006). Furthermore, into the erythrocyte, Cr (VI) was bound to beta-chain of hemoglobin (Barceloux, 1999) which could explain the depletion of hemoglobin concentrations observed in the present study. On the other hand, the diminution in hemoglobin concentrations could be probably due to structural alteration of heme which disturbs hemoglobin synthesis, and also to the inhibition of the enzyme system involved in the synthesis of hemoglobin as earlier suggested with other heavy metals (Burden *et al.*, 1998., Gurer *et al.*, 1998). Dichromate potassium in drinking water had no effect on the number of erythrocytes and hematocrit levels in male *wistar albino* rats. This is in accordance with a study on mice, in which Cr (VI) with drinking water does not induce any clastogenic effect on hematopoietic cells of adult mice and their fetuses (De Flora *et al.*, 2006). This route of

exposure is widely believed to cause much less toxicity than other route exposures, because ingested Cr(VI) is converted to inactive trivalent chromium in stomach (Paustenbach *et al.*, 2003 and De Flora *et al.*, 2006). The diminution in platelets counts induced with graded doses of chromium subcutaneously on day 3 after exposure could be due to the presence of infection as observed in mice after inoculation with Dengue virus (Shrivastava *et al.*, 2005). On the contrary, the augmentation on platelets values induced by Chromium on day 6 subcutaneously or at the end of experiment in drinking water also reported in mice (Shrivastava *et al.*, 2005) suggested the presence of inflammatory case. Furthermore, our results demonstrated that chromium dichromate in drinking water or administered subcutaneously to male or female rats is susceptible to perturb immune response. Indeed, leucopenia and lymphopenia observed on day 3 and 6 after subcutaneous Cr (VI) administration or on day 20 in drinking water were also observed in mice (Shrivastava *et al.*, 2005) and in fishes (Steinhagen *et al.* 1984 and Arunkumar *et al.*1986). It was reported that Cr (VI) easily enters in physiological membranes and is actively transported into cells and remains here for the life of the cell. In persons occupationally exposed to Cr (VI), the determination of Cr (VI) showed a significant increase in chromium levels in the lymphocytes (Lukanova *et al.*1996). Furthermore, the depletion of lymphocytes has also been reported *in vivo* in patients with metallic prostheses and has been correlated with elevated chromium levels in blood (Raghunathan *et al.*, 2009). Cr (VI) induced in human to it exposed an apoptosis of blood lymphocytes (Vasant *et al.*, 2001) and significantly reduced the lymphocytes size (Geetha *et al.*, 2005). Cr (VI) in contact with biological compounds may lead to peroxidation

of biological compounds that are present in the cell or on its surface. In effect, some negative changes such as cell membranes damaged due to peroxidation of unsaturated fatty acids or inhibition of both mitochondrial transmembrane potential in rat lymphocytes (Geetha *et al.*, 2005) may occur and could explain the reduced lymphocyte and leukocyte counts. On the other hand, it was reported that Cr (VI) is genotoxic. Several studies reported that the one major lesion associated with Cr (VI) is the DNA damage in the intact lymphocytes (Costa *et al.*, 1996). Incubation of human lymphocytes with Cr (VI) resulted in a dose-dependent increase in DNA strand break. This is also detected in the rat peripheral lymphocytes (Gao *et al.*, 1992). Furthermore, the decrease in the lymphocyte counts in our rats, which received Cr (VI) in drinking water during three weeks, could be due to the increase in the formation of DNA-Protein- crosslinks reported in the rat blood lymphocytes (Coogan *et al.*,1991) and in the exposed population (Taioli *et al.*1994) or during *in vitro* or *in vivo* exposure (Manerikar *et al.* 2008). The formation of DNA lesions induced by Cr (VI) may result from the implication of the enhanced reactive oxygen species (ROS) and hydrogen peroxide in the human lymphocytes (Aziak and Kowalik, 2000 and Geetha *et al.* 2005) and the decrease in glutathione levels and inhibition of proliferation of lymphocytes (Geetha *et al.* 2002). On the contrary, the present study showed that subcutaneous administration of potassium dichromate in female rats or in male 10 days after exposure in drinking water augmented the number of monocytes and granulocytes. Similar findings have been reported in fish (Arunkumar *et al.*, 2000) and in mice (Shrivastava *et al.*, 2005) exposed to Cr (VI) with drinking water or in rats exposed to atmosphere containing Cr (VI) (Cohen *et al.*, 1998). Moreover, chronic

exposure to these low clinically relevant concentrations of Cr (VI) induced a potent adaptive response with elevated glutathione-S-transferase expression and increased activities and expression of reactive oxygen scavengers, superoxide dismutases, catalase and glutathione peroxidase and temporal increases in reduced glutathione levels, glutathione reductase activity, and glutamate cysteine ligase expression (Raghunathan *et al.*, 2009). Monocytes were more susceptible to the toxicity of the metal. Indeed, chromium used in prostheses enhanced the human blood monocyte/macrophage proliferation and significantly increased the level of interleukin-1 α , interleukin-1 β , and TNF- α (Lee *et al.*, 1997 and Wang *et al.*, 1996).

CONCLUSION:

The interesting finding in the present study is that short-term exposures to a low dose of K₂Cr₂O₇, s.c induces in female *wistar albino* rats erythrocytopenia, thrombocytopenia, leucopenia, lymphopaenia, granulocytosis, monocytosis and a decrease in hematocrit values and hemoglobin concentrations, on the other hand, in drinking water chromium is susceptible to affect the immune response and induces leucopenia, lymphopenia, monocytosis, and granulocytosis. In male *wistar albino* rats, oral route of exposure had no effect on erythropoietic parameters.

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تأثير ديكرومات البوتاسيوم على مؤشرات الدم عند إناث وذكور الجرذان ألبينو وستار عجروود وناسة

مخبر فيزيولوجيا الحيوان - قسم العلوم البيولوجيا- جامعة باتنة - الجزائر

يعتبر الكروم سداسي التكافؤ من أهم السموم الفتاكة حيث يتسبب غالباً في إصابة الإنسان والحيوان بداء السرطان. وفي هذه الدراسة تمت مقارنة تأثيرات ديكرومات البوتاسيوم ($K_2Cr_2O_7$) على المؤشرات الدموية لإناث وذكور الجرذان ألبينو وستار، وذلك باستعمال العديد من الجرعات والحقن المختلفة مع تغيير مدد المعايين من تجربة لأخرى. ومن جهة تم إعطاء ذكور الجرذان جرعة ٣٠ ملليجرام/لتر من ($K_2Cr_2O_7$) عن طريق ماء الشرب خلال ٢٠ يوم على التوالي، ومن جهة أخرى تم حقن إناث الجرذان بحقن تحت الجلد بجرعات متزايدة من (١٠-٥٠، ١٠٠ ملليجرام/كجم). أما المجموعة الضابطة فعولجت بكلوريد الصوديوم ٠.٩% (٠.٣ مل) عن طريق الحقن تحت الجلد أو بالماء المقطر عن طريق الشرب.

المؤشرات الدموية تم قياسها في اليوم الثالث، اليوم السادس، واليوم الحادي والعشرين عن طريق المعالجة تحت الجلد، أما بالنسبة للمعالجة عن طريق ماء الشرب فتم قياسها في اليوم العاشر واليوم العشرين من العلاج. أثبت النتائج أن ١٠ ملليجرام/كجم التي تم حقنها تحت الجلد أحدثت انخفاضاً سريعاً ومعنوياً في الثالث أيام الأولى في عدد الكريات الحمراء بنسبة ٦%، بنسبة ٣٠% في عدد الكريات البيضاء، ٤٨% في عدد الصفائح الدموية، كما سجلت نسبة الخلايا المصمتة انخفاضاً بنسبة ١٥%، أما عدد كريات الدم البيضاء المحببة فسجلت ارتفاعاً بنسبة ١٢٤% مقارنة بضابط التجربة، كما أوضحت أيضاً تركيز الهيموجلوبين إلى جانب عدد اللمفاويات انخفاضاً محسوساً في اليوم السادس بعد المعالجة.

إن حقن ٥٠ ملليجرام/كجم من الكروم تحت الجلد تسبب في اليوم الثالث في حدوث أضرار بليغة في مستوى مؤشرات تنشئة الكريات البيضاء، كما خفضت الكريات بنسبة ٥٥%، والكريات اللمفاوية بنسبة ٥٧% مع ارتفاع كريات الدم أحادية النواة بنسبة ١٠٤% زيادة على ارتفاع تكاثر الكريات البيضاء المحببة بنسبة ٢٠٤%، كما عرف تكاثر الصفائح الدموية بنسبة ٣٨% مقارنة بضابط التجربة، كما سجلنا في اليوم السادس انخفاضاً في عدد الكريات الحمراء وتركيز الهيموجلوبين على التوالي بنسبة ٢٢%، ٢٢% أما في اليوم الواحد والعشرين فعرف هو أيضاً انخفاضاً على التوالي بنسبة ٤١%، ٣٦% أما نسبة الخلايا المصمتة انخفضت بنسبة ٣٦% فقط في نهاية التجربة.

أما بالنسبة لجرعة ١٠٠ ملليجرام/كجم من الكروم فتم حقنها تحت الجلد فخفضت بشكل واضح خلال الأيام الثلاثة الأولى في عدد الكريات الحمراء بنسبة ٢٠% والصفائح الدموية بنسبة ٢٠%، وإجمالي الكريات البيضاء بنسبة ٥٥%، والكريات اللمفاوية بنسبة ٥٩%، كما ارتفع عدد كريات الدم البيضاء بنسبة ٥٦%، وكريات الدم البيضاء المحببة بنسبة ١٦٦%، كما ارتفع عدد الصفائح الدموية في اليوم السادس ب ٢٧% مقارنة بضابط التجربة.

لم تؤثر إضافة ٣٠ ملليجرام/لتر من الكروم في ماء الشرب على تنشئة كريات الدم باستثناء تسببها في ارتفاع عدد صفائح الدم في نهاية التجربة، إلا أن عدد الكريات البيضاء والكريات اللمفاوية انخفضت بشكل محسوس على التوالي بنسبة ٣٧%، ٣٧% في اليوم العشرين بعد التجربة، وخلافاً لذلك ارتفعت كريات الدم البيضاء المحببة والكريات البيضاء خصوصاً في اليوم العاشر بعد المعالجة على التوالي بنسبة ٤٢%، ٢٢% مقارنة بضابط التجربة.

إن حقن الجرذة ألبينو وستار تحت الجلد بجرعة ضعيفة من الكروم على المدى القصير يتسبب في انخفاض في عدد الكريات الحمراء والكريات الدم البيضاء وعدد الصفائح الدموية مع ارتفاع عدد كريات الدم البيضاء أحادية النواة وتكاثر كريات الدم البيضاء المحببة وانخفاض نسبة الخلايا المصمتة وتركيز الهيموجلوبين، أما شرب الجرذ ألبينو وستار لجرعة الكروم في ماء الشرب يمكن أن يسبب له أضرار على مستوى خلايا المناعة، والتي تتسبب في انخفاض عدد الكريات البيضاء واللمفاوية وتكاثر كريات الدم البيضاء أحادية النواة والمحببة. وإن الحقن عن طريق الشرب ليس له تأثير على مؤشرات تنشئة كريات الدم عند الجرذان ألبينو وستار.