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Research Article

Acute and sub-chronic oral toxicity study of cadmium in Swiss albino mice

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ABSTRACT
Cadmium is an wide-ranging and non-biodegradable contaminant of great uneasiness to human health. In real-life scenarios, we are uncovered to combinations of chemicals rather than single elements, and it is of paramount standing to measure their toxicity. Cadmium is among the toxic and hazardous metals widely discrete in the environment at high levels. Cadmium is one of the furthermost substantial toxic heavy metals and likely for human exposure has mostly increased with the rise in this component's industrial procedure. In the present study to observe the consequence of Cadmium-induced toxicity in Swiss albino mice. Experimental groups received a single Oral Dose (100mg/kg) of Cd aqueous solution. Toxic effects of Cadmium was investigated on hematological and biochemical parameters in Swiss albino mice.. The present study shows that the level of (RBC) Red Blood Cells and (WBC) White Blood Cells were significantly decreased. This study showed a significant decrease ($P < 0.05$) in Total Cholesterol and Total Serum Protein and significantly increased Serum Creatinine and Total Blood Urea due to Cadmium exposure. It is concluded from the experimental study that cadmium toxicity increases hematological parameters and biochemical parameters in Swiss albino mice.

Keywords: Heavy metals, Cadmium, toxic effect, hematological parameters and Biochemical analysis.

INTRODUCTION

Environmental pollution, particularly by chemicals, is one of the greatest significant influences in destroying the biosphere components. Among all substances contaminates, heavy metals are considered potentially hazardous contaminants in the biosphere to human health (Feleafel and Mirdad, 2013). Heavy metals pollution characterizes a significant conservational problem due to metals' toxic effect and accumulation throughout the food chain leading to severe ecological and health issues (Mansour, 2014). This problem is even getting more serious worldwide, especially in developing countries (Sathawara et al., 2004; Radwan and Salama, 2006). An estimated 0.5-1.0 million people die prematurely each year due to heavy metal pollution (Kojima, 2001). These metals don't recognize a slightly biotic role in the body and are incredibly toxic, uniform at low stages (Robert and Clarkson, 2001; Jarup, 2003; Parthipan and Muniyan, 2013). Toxicity is definite as any detrimental effect of a chemical or a remedy on a target organism. Acute and sub-acute toxicities have been defining by a range of professionals. The Society for Economic Co-operation and Progress board of experts (OECD, 1980) describes serious toxicity as the opposing special things going on within a short time of direction of a single dose of a substance or numerous-dose given within 24 hours and substitute-acute toxicity as the adverse effects occurring as an outcome of the regular day-to-day dosing of chemical to investigational animals for fourteen days (Veerappan et al., 2007). Toxicity is a poisonous phrase, representative of the state of adverse effects caused by the interaction between toxicants and cells (Syahmi ARM et al., 2010). However, all substances are potential poisons since all of them can cause injury or death following excessive exposure (Duffus et al., 2006). On the other hand, all chemicals can be used undamaged if exposure of people or susceptible organisms to chemicals is kept below defined tolerable limits (IARC, 1993). Preliminary studies of acute toxicity should determine the suitable dose of a drug.

The effect of metal or multimetal exposure on the testis is of great concern as occupational exposure to certain metals results in impaired reproductive function (Onyenmechi J Afonne et al., 2002). Heavy metals similar to manganese, selenium, and zinc are vital for usual testicular function; however, mercury, cobalt, cadmium, and lead are toxic (Anderson MB et al., 1992). Cadmium (Cd) is one of the furthermost toxic heavy metals

in the atmosphere. Besides its salts, cadmium chloride, Cadmium is amongst the essential air and water pollutants. They have been used extensively by copper, lead, and zinc smelter alkaline accumulator, paint, and plastic industries (M.A.EI-Hady et al., 1995). Their toxicity has been extensively studied and described. Cadmium acquaintance causes impairment to live organs, particularly in humans, leading to Itai-Itai disease (B.T. Emmerson, 1970; Feriberg et al., 1986). Also, all Cd mixtures have been classified as human carcinogenic fundamentals (IARC, 1993). The primary sources of exposure to these metals are working with them through oral or dermal contact. Still, it occurs mainly through inhalation (RB Kalahasthi et al., 2006), cigarette smoking, and alcoholic beverages (L.Jarup et al., 1998). The adverse effects of Cadmium involve oxidative harm in tissues. This effect is considered an early sign of its toxicity and has been linked to carcinogenesis (Waalkes MP, 2000). Cadmium (Cd) everywhere, heavy metal and an environmental pollutant, found in soil, water, and air.

Cadmium (Cd) represents a carcinogenic metal (WHO, 1992), and it is a severe ecological and industrial pollutant. Industrial emissions, cigarette smoking, and fertilization are symbols of an essential cadmium revelation source for humans. In the body, cadmium accumulates mainly in the reproductive tissues, kidneys, liver, etc. (Valko M et al., 2006). Cadmium mixtures have been shown to wield toxic and carcinogenic properties in humans and experimental animals (Misra RR et al., 1998). Cadmium has a high effect inducing toxicity in Skeleton muscles in edematous emphysema, lungs, osteomalacia, osteoporosis, hemorrhage, brain edema, and blood-brain barrier disruption (Patra Rc et al., 2011; Shukla A et al., 1996). Though, Cadmium likely critical results in cerebral microvessel thrombosis and its association to oxidative stress and systemic inflammation has not been reported. Critical toxicity studies' primary objective is to classify a single dose causing significant adverse effects or life-threatening toxicity, often estimating the minimum dose causing lethality.

MATERIAL AND METHODS

Chemicals: Cadmium and all the chemicals used in the experiments were of analytical mark and purchased from Himedia Laboratories Private Limited. (Mumbai, India).

Animals: Swiss albino mice (6-7 week old) weighing almost 25-30 gm were stored in polypropylene cages (3 animals each cage). The animals were kept throughout the experiment for full accommodation in an air-conditioned animal room (25± 2ºC) under a 12 h light/dark cycle. The animals had allowed access to water and standard pellet diet. All experimental process was accomplished in accordance with the recommendations found in the Guide for the Care and Use of Laboratory Animals (Refer) and permitted by the institutional Animal House of Biyani Girls College, Jaipur and Use Committee of the Jayoti Vidyapeeth Women's University of Jaipur. Institutional ethical rules were also followed in all Experiments.

Induction of Cadmium in mice: Cadmium was induced in Swiss albino mice by feeding 100 mg/kg body weight dose solution in water for 5 week that was prepared every day.

Experimental design: The mice were divided into two sets including of 3 animals in separately group as follow:-

Group I: - Normal (Control mice), received fed with standard pellet diet and water.

Group II: - Cadmium fed mice, fed with 100mg/kg body weight aqueous solution for 35 days.

Experimental procedure:

Bodyweight Estimation: All animals were weighed every week until the end of the experimental protocol.

Biochemical Estimation: The experiments were carried out for 5 weeks. Throughout the experiment blood sample were obtained after the overnight from the tail vein of all the animals. Blood was left to lump and centrifuge at 5000 rpm for 15 min. at 4°C for separating the serum which was frozen and stored at -20°C until biochemical analysis like serum cholesterol (Zak's Method, 1957), total serum protein (Lowry et al., 1951); serum creatinine (Brod J et al., 1948 & Maheswari C et al., 2013) and blood urea level (Natelson S et al., 1951 & Aseervatham J et al., 2010) was also analysis.

Statistical analysis:

All results are presented as mean±SEM. To regulate the significant variances between the two groups were considered using student t- test. *P* value of less than 0.05 were measured to be significant. All evaluates were completed using IBM SPSS Statistics 21.

RESULTS

Cadmium when bio accrued from dissimilar sources, leads to numerous pathological circumstances. (Hounkpatin et al., 2013) highlighted that the maximum significant tissues in the human body are blood where metabolic variations are reproduced. Consequently, any differences in the parameters of blood are regarded as the most consistent indicators of toxic

belongings of chemicals, heavy metals and drugs etc.

In the present study, the Red Blood cell (RBC) and White Blood cell (WBC) of Swiss albino mice subjected to Cadmium treatments are shown in Table 1 and Figure 1. During the 5 week of experiment, the White Blood cell (WBC) and Red Blood cell (RBC) of Swiss albino mice group (9.06±0.001; 8.08±0.004) slightly significantly decreased from the first week until the end of the experiment when compared with control group (3.79±0.001; 3.53±0.009) ($p<0.05$).

The effect of Cadmium on body weight of mice group during the experimental period are represented in Figure 2; Table 2. The results showed the body weight Cd-treated mice group (20.07±0.01) significantly decreased with respect to the control group (31.17±0.08) ($P<0.05$).

Table 1: The effect of cadmium on WBC (white blood cell) and RBC (red blood cell) in control and experimental mice

Days	WBC		RBC	
	Group- I	Group-II	Group- I	Group-II
	Control Mice	Cd Treated Mice (100mg/kg)	Control Mice	Cd Treated Mice (100mg/kg)
0 Days	9.67±0.005	9.65±0.003	8.98±0.001	8.92±0.003
7 Days	9.98±0.004	6.98±0.001	8.07±0.003	7.67±0.002
14 Days	8.36±0.001	5.35±0.098	9.04±0.001	6.63±0.001
21 Days	8.99±0.004	4.98±0.007	8.98±0.001	5.78±0.002
28 Days	9.12±0.001	3.88±0.009*	8.07±0.003	4.09±0.007
35 Days	9.06±0.001	3.79±0.001*	8.08±0.004	3.53±0.009*

Values are shown as the mean±SEM. Values are statistically significant at * $p<0.05$ when compared to normal/control group.

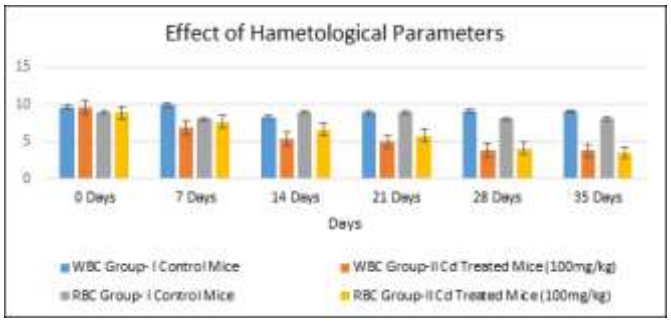

Fig.1: showing the effect of cadmium on WBC (white blood cell) and RBC (red blood cell) in control and experimental mice

Table 2: The effect of cadmium on Body weight in control and experimental mice

Days	Group- I	Group-II
	Control Mice	Cd Treated Mice (100mg/kg)
0 Days	32.45±0.09	32.56±0.09
7 Days	32.56±0.06	28.05±0.04
14 Days	30.98±0.05	26.09±0.03
21 Days	31.08±0.07	23.12±0.02
28 Days	32.98±0.07	22.09±0.02
35 Days	31.17±0.08	20.07±0.01*

Values are shown as the mean±SEM. Values are statistically significant at * $p<0.05$ when compared to normal/control group.

Fig.2: Showing the effect of cadmium on body weight in control and experimental mice

Treatment with Cd caused showed a significant decrease of cholesterol (70.09±0.11) with ($p<0.05$) compared to the control group (87.09±0.24). Meanwhile the concentration of serum total protein was reduced (2.11±0.001) ($p < 0.05$) as compared to control group (7.13±0.009). Table 3; Figure 3. The results showed the serum creatinine Cd-treated mice group significantly increase (6.76±0.21) with respect to the control group (0.98±0.08) ($P<0.05$). cadmium induced mice were found to have significantly elevated blood urea level (57.09±0.28) with the respect to the control group (38.99±0.13) ($p<0.05$) Table 4; Figure 4.

Table 3: The effect of cadmium on Total cholesterol and Total serum protein in control and experimental animal.

Days	Total Cholesterol		Serum Protein	
	Group- I	Group-II	Group- I	Group-II
	Control Mice	Cd Treated Mice (100mg/kg)	Control Mice	Cd Treated Mice (100mg/kg)
0 Day	87.12±0.23	88.12±0.31	6.78±0.006	7.1±0.009
7 Day	87.34±0.24	79.55±0.21	7.01±0.009	6.12±0.006
14 Day	88.18±0.33	76.05±0.18	7.12±0.007	4.78±0.004
21 Day	89.06±0.34	75.16±0.18	6.99±0.006	4.01±0.004
28 Day	87.98±0.24	71.97±0.11	7.11±0.009	3.06±0.002
35 Day	87.09±0.24	70.09±0.11*	7.13±0.009	2.11±0.001*

Data are shown as the mean±SEM. Values are statistically significant at * $p<0.05$ as compared to normal/control group.

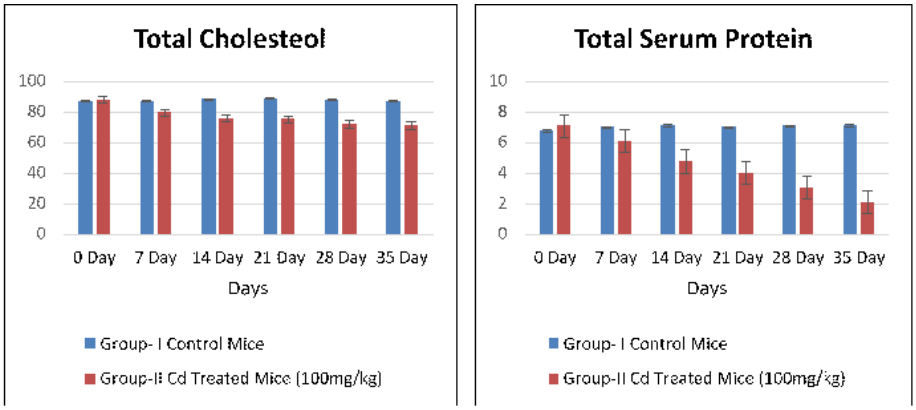

Fig.3: Showing the effect of cadmium on Total cholesterol and Total serum protein in control and experimental mice.

Table 4: The effect of cadmium on Serum creatinine and Blood urea in control and experimental animal

Days	Serum Creatinine		Blood Urea	
	Group- I	Group-II	Group- I	Group-II
	Control Mice	Cd Treated Mice (100mg/kg)	Control Mice	Cd Treated Mice (100mg/kg)
0 Day	1.09±0.12	0.98±0.08	39.98±0.13	37.97± 0.13
7 Day	0.92±0.07	1.19±0.12	37.16±0.11	41.85±0.19

14 Day	1.02±0.13	1.98±0.14	38.88±0.12	52.19±0.22
21 Day	0.98±0.08	2.97±0.17	39.78±0.13	54.89±0.25
28 Day	0.99±0.08	4.89±0.19	38.09±0.11	58.98±0.28
35 Day	0.98±0.08	6.76±0.21*	38.99±0.13	57.09±0.28*

Data are shown as the mean±SEM. Values are statistically significant at * $p<0.05$ as compared to normal/control group.

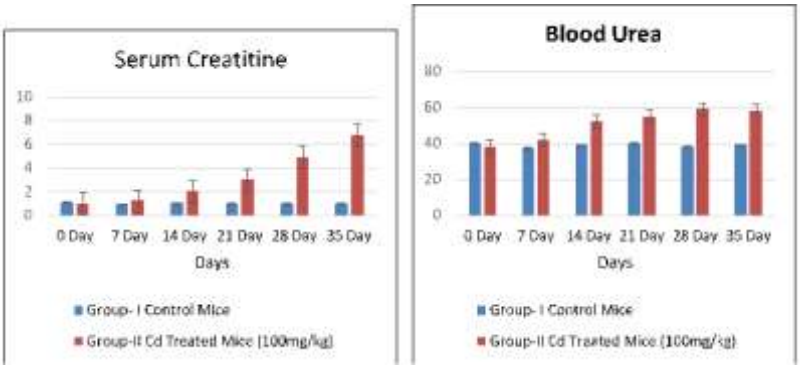

Fig.4: Showing the effect of cadmium on Serum Creatinine and Blood Urea in control and experimental mice

DISCUSSION

Cadmium is an actual toxic metal and an eco-friendly and industrial pollutant current in soil, water, air, and food (Cinar, 2003; Kaplan et al., 2011). It's identified to mark several organs like liver, tests, bones and kidney in human beings and experimental animals (Tokamure et al., 2006; Diana, 2008; Mohamed et al., 2014). The results found from the current study showed that when male mice are exposed to different Cadmium doses, it causes significant differences in most of the hematological parameters and Biochemical analysis.

The current study result revealed a significant decrease in WBC and RBC of treated mice with Cadmium compared with the control group.

It has been reported that RBC and WBC count significantly decreased in the swiss albino mice administered $CdCl_2$ (Saggu S. et al., 2019). A similar result has been reported by M.A.R Sarkar 2013, who found a significant decrease in WBC and RBC count in swiss albino mice exposed to Cadmium. (Ashraf M.M. Sharaf et al., 2017) have also shown that RBC count reduced significantly in $CdCl_2$ treated Rats. These results coincide with those obtained by (Hounkpatin A.S.Y. 2012). They indicated that the RBC count significantly decreased in Wistar Rats Exposed to the Cd concentration comparatively to the control group. These results have been shown by other authors (Veena S. et al.,2011; Lavicoli I. et al.,2003) conducted on mice poisoned with lead.

In the present study, Cadmium fed mice showed a significant and consistent decrease in body weight at different intervals through the experiment period compared to the control group. Other studies have been described in swiss albino mice that significantly decrease body weight (M.A.R Sarkar et al., 2013; Kumari, A. and Sharma, S.,2020). The decrease in body weight content in the Cd-treated group agrees with other authors' work (Naima, L. and Zine, K., 2012; Ashraf M.M. Sharaf et al., 2017). The observed rats' decreased weight gain in this study is consistent with some previously published reports (Horiguchi et al., 1996). Our study cadmium treated mice showed a significant decrease in Cholesterol, serum protein, and increase serum creatinine and blood urea level compared to the control group. Previous studies showed a substantial reduction in total serum protein level and increase urea and creatinine levels in albino rats (Kshirsagar M et al. 2015; Ashraf M. M. Sharaf et al., 2017). The decrease in cholesterol content in the Cd-treated group agrees with other authors' work (Kshirsagar M et al., 2015; Purohit RK et al., 2007; Sharma S, and Vijaya P, 2015; Sharma S, and Vijaya P, 2015; Chakrawarti A et al., 2010). Reduced protein content was observed in the Cd-treated group. Some authors also reported a similar trend in their research findings (Sharma S, and Vijaya P, 2015; Sharma S, and Vijaya P, 2015; Chakrawarti A et al., 2010; El-Demerdash FM et al., 2004; Kaoud HA et al., 2010; Somade PM et al., 2014; Babaknejad N et al., 2016). The decrease in serum total protein of Cd-treated mice might be due to changes in protein synthesis and metabolism (Kshirsagar M et al. 2015; Dostal et al., 1989; Das and Dasgupta, 2000). This result agrees with other findings (Yousuf, 2002 and Naima, L. and Zine, K., 2012).

CONCLUSION

These findings suggest that cadmium present in the environment and in particular in foodstuffs of first necessity cause of hematological disturbances in the blood. Based on the explained data, we conclude that the present investigation also showed that cadmium chloride has significant effect on the abnormalities of hematological parameters and biochemical analysis.

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Conflict of Interest: Nil

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