RESEARCH ARTICLE

Received on: 12/05/2015
Accepted on: 30/06/2015
Published on: 22/08/2015

Corresponding Author
Sandeep Kaur
Department of Zoology and Environmental Sciences
Punjabi University, Patiala, India-147002.
Email:
noori_rainu@yahoo.co.in

QR Code for Mobile users

Conflict of Interest: None Declared !

Assessment of Biochemical Alterations Induced by Acute and Chronic Doses of Cadmium in Albino Mice

Sandeep Kaur and Suman Sharma
Department of Zoology and Environmental Sciences
Punjabi University, Patiala, India-147002.

ABSTRACT

Cadmium is an extremely hazardous heavy metal, which is widely distributed throughout the biosphere due to increase in industrialization. Its accumulation in blood affects various organs and causes many clinical dysfunctions. The main purpose of this study was to evaluate the toxic effects of different doses of cadmium (Cd) on liver function through biochemical analysis. For this, animals were divided into three groups. Group 1: Control group. Group 2: received a single dose (acute dose) of 2mg/kg body weight of $CdCl_2$ intraperitoneally where autopsies were done on 1, 7, 15 and 30 days post treatment. Group 3: received 0.01mg/kg body weight/day (chronic dose) of $CdCl_2$ intraperitoneally daily for 15 and 30 days. It was observed that Cd led to significant increase in the level of serum cholesterol, LDL-c, VLDL-c and triglycerides on 15 and 30 days post treatment in both acute and chronic dose treated mice as compared to control. Similarly, in the case of transaminases, activity of SGPT was found to be increased significantly ($P<0.05$) in cadmium treated groups (2 and 3) compared to control mice but SGOT was increased significantly ($P<0.01$) only in chronic dose treated mice. Hence, the present study shows that chronic doses of cadmium to mice showed more deleterious results.

Keywords: Cadmium, hepatotoxic, lipid profile and transaminases.

Introduction

Cadmium (Cd) is a ubiquitous environmental pollutant and is widely distributed in the environment due to extensive use of cadmium based products. It has special importance due to its long half life and it can threaten human health both through environmental and occupational exposure[1].

Under normal conditions, the intake of Cd depends on the Cd concentration in natural sources such as air, land and water which usually does not exceed 20mg/day[2]. Consumption of contaminated food due to excessive use of phosphate fertilizers[3] and contaminated water through galvanized pipes have become the major ways of human exposure to cadmium[4,5]. Moreover, in many countries, contamination of rivers and adjoining seas by Cd and other heavy metals have been reported because of excessive discharge of the waste liquid matter from industrial sites and residual sludges of fertilizers and pesticides[6]. The first reports of the severe health problems due to Cd intoxication arose in 1940s in Japan, where the Itai-Itai disease was endemic and its major reported symptoms were bone and renal damage, which were caused by eating Cd-polluted rice[7]. International Agency for Research on Cancer (IARC) has classified Cd as "category I" human carcinogen[8,9]. Cadmium can also lead to bone defects, high blood pressure, myocardiac dysfunctions, proteinuria and pulmonary oedema[10].

The liver and kidneys are among the major target organs of cadmium accumulation and intoxication[11, 12], including testes and ovaries. Approximately, half of the cadmium absorbed systematically accumulates rapidly in the liver, resulting in reduced availability of the cadmium to the other organs such as kidneys and testes, which are more sensitive to its toxic actions[13].

Hence, the aim of the present study was to evaluate and compare the hepatotoxic effects of different doses of cadmium by considering biochemical analysis in albino mice.

Materials and Methods

Chemical

Cadmium chloride ($CdCl_2$) was purchased from S.D. Fine –Chemical Limited, Mumbai, India. The lethal median dose (LD_{50}) of cadmium in mice was considered to be 3.2mg/kg body weight[14]. Hence, an acute dose of 2mg/kg body weight (0.60 LD_{50}) and chronic dose of 0.01mg/kg body weight (0.032 LD_{50}) were selected for this study.

Experimental animals

Albino mice weighing 23-25g were procured from Central Research Institute, Kasauli (H.P, India). They were kept and acclimatized to the laboratory conditions for 15 days under optimal conditions of temperature and light (25±1ºC, 12h light-dark cycle). They were fed standard mice feed (purchased from Ashirwad Private Limited, Chandigarh) and water ad libitum. The animals were handled with humane care in accordance with the guidelines of the 'Committee for the Purpose of Control and Supervision of Experiments on Animals (CPCSEA)', India and all experimentation procedures were approved by Institutional Animal

Ethical Committee (Reg No. 107/1999/CPCSEA/2009-03).

Experimental design: Forty two mice were divided into three groups of six animals in each. Animals of group 1 were received distilled water and kept as control. Animals of group 2 were administrated a single dose (acute dose) of 2mg/kg body weight of $CdCl_2$ intraperitoneally and were sacrificed on 1, 7, 15 and 30 days post treatment. Mice of Group 3 were further divided into two sub groups and 0.01mg/kg body weight (chronic dose) of $CdCl_2$ were injected for 15 and 30 days and autopsies were done on respective days.

Biochemical analysis: On the day of autopsy, the blood sample was collected in separate eppendorf tubes from each mouse under ether anesthesia. The blood sample was centrifuged (3000 rmp at 2°C for 15 minutes) and serum was collected in fresh separate clean tubes. Serum was then stored at -20 °C for later use for biochemical analysis:

Lipid profile: Quantitative estimation of cholesterol concentration in serum was determined by Cholesterol oxidase–peroxidase/ Phosphotungstate method using commercially available kit (Reckon Diagnostics Private Limited, Baroda, India). Triglycerides were determined by Glycerol-3-phosphate oxidase-Peroxidase method by using kit (Medsource Ozone Biomedicals Private Limited, New Delhi, India). High density lipoprotein-cholesterol (HDL-c) was determined by standard method (kit supplied by Transasia Bio-Medicals, Baddi, India). Very low density lipoprotein-cholesterol (VLDL-c) was determined by using the formula: VLDL-c (mg/dl) = Triglycerides (mg/dl)/ 5. Low density lipoprotein-cholesterol (LDL-c) was determined indirectly by Friedewald's formula LDL-c (mg/dl) = Total cholesterol–HDL-c–(0.20 x triglycerides).

Serum transaminases activity: The estimation of serum transaminases including serum alanine transaminases (ALT) or glutamic pyruvate transaminases (GPT) and aspartate transaminases (AST) or glutamic oxaloacetate transaminase (GOT), which were determined according to IFCC (International Federation of Clinical Chemistry and Laboratory Medicine) on autoanalyzer by using biochemical kits procured from Transasia Bio-Medicals Limited., Baddi, India.

Statistical analysis: The values are expressed as Mean±Standard Error. The data was analyzed by using One Way ANOVA followed by post hoc comparison with the level of significance at $P<0.05$.

RESULT

During experimental period, no mortality was recorded in any of the exposed groups. However, mice treated with $CdCl_2$ (group 2 and 3) showed mild signs of toxicity such as reduced feed intake, indigestion, anxiety, dizziness, fatigue, aggression, ruffled hair coat and marked weight loss whereas controls appeared normal.

Tables 1 and 2 depict the lipid profile of experimental groups. The Cholesterol, LDL-c, VLDL-c and triglyceride level was also found to be significantly increased in both acute and chronic dose treated mice in comparison to control group. Although, the elevation of lipid profile was pronounced at 15 and 30 days post treatment in both groups (2 and 3) of Cd treated mice (Table 1 and 2), more devastating results ($P<0.001$) were obtained in chronic dose treated mice (group 3). In contrast, HDL-c level in cadmium treated mice showed significant reduction in comparison to control mice. But in chronic dose treated mice, the reduction was more prominent ($P<0.001$) on 30 days post treatment.

In the case of serum transaminases activity, the glutamic pyruvate transaminases (GPT) was found to be increased significantly ($P<0.05$) in both acute and chronic dose treated mice (at 15 and 30 days post treatment, Fig.1 and 2) as compared to control group but serum glutamic oxaloacetate transaminase (GOT) was increased statistically significant ($P<0.01$) only in chronic dose treated mice (Fig. 2).

Parameters (mg/dl)	Autopsy days				
	Control	1	7	15	30
Cholesterol	85.30±1.59	88.13±0.78	89.33±0.89	94.00±1.29*	96.33±0.99*
HDL-c	56.47±0.53	54.62±0.42	50.25±1.19*	48.16±1.31*	46.83±1.63*
LDL-c	7.66±1.89	10.35±0.74	17.08±1.76*	15.83±1.49	18.69±2.57*
VLDL-c	21.17±0.60	23.16±0.47	26.67±0.56*	29.99±0.51*	30.80±0.72*
Triglycerides	105.85±3.00	115.80±2.35	133.35±2.81*	149.95±2.55*	154.00±3.60*

Table 1: Comparison of serum lipid profile in control and cadmium treated mice (acute dose).** Significance at *P*<0.001, * P<0.01, ° P<0.05HDL: High-density lipoprotein; VLDL: Very-low-density lipoprotein; LDL: Low-density lipoprotein

Parameters (mg/dl)	Autopsy days		
	Control	1	7
Cholesterol	85.30±1.59	96.97±0.94**	99.46±0.79**
HDL-c	56.45±0.53	47.00±2.02*	44.50±3.77**
LDL-c	7.66±1.89	21.27±1.67**	24.8±1.98**
VLDL-c	21.17±0.60	28.70±0.80**	30.16±0.73**
Triglycerides	105.85±3.00	143.5±4.02**	168.83±3.64**

Table 2: Comparison of lipid profile in control and cadmium treated mice (chronic dose).* Significance at P<0.01, ** P<0.001
HDL: High-density lipoprotein; VLDL: Very-low-density lipoprotein; LDL: Low-density lipoprotein.

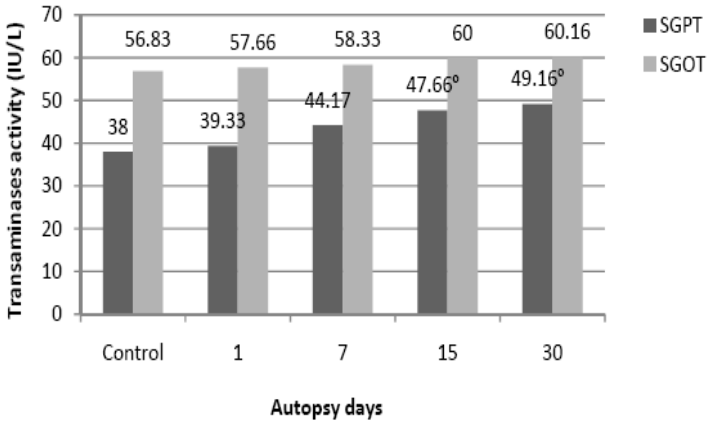

Figure1: Comparison of serum transaminases in control and cadmium treated mice (acute dose).
º Significant variations at P<0.05. SGOT: Serum glutamic pyruvate transaminases; SGPT: Serum glutamic oxaloacetate transaminases

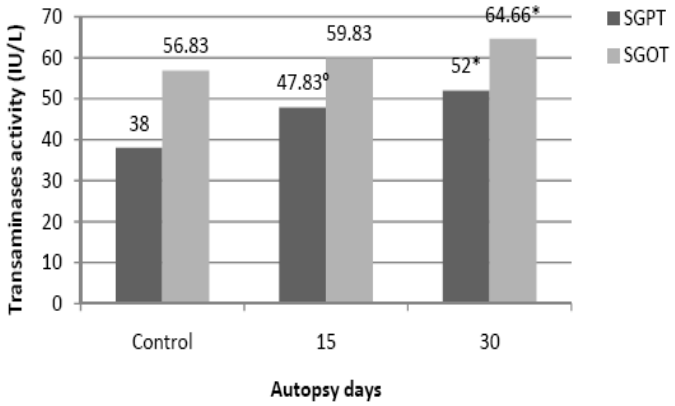

Figure-2: Comparison of transaminases in control and cadmium treated mice (chronic dose).
º Significant variations at P<0.05 and * P<0.01. SGOT: Serum glutamic pyruvate transaminases; SGPT: Serum glutamic oxaloacetate transaminase

Discussion

The liver is a major target organ for toxicity of xenobiotics and drugs, because most of the orally ingested chemicals and drugs are initially metabolized in liver into toxic or non toxic intermediates. Excessive exposure to hepatotoxins may induce severe liver injury characterized by abnormality of hepatic function[15]. In this study, we evaluated the effect of cadmium exposure on lipids and transaminases activity in mice, which act as biomarkers of liver damage and thus used for estimation of liver diseases with some degree of intact liver function.

During present study, cadmium exposure caused a dose-dependent increase in plasma Cholesterol and triglyceride levels which is thus associated with increased concentration of the triglycerides-rich lipoproteins i.e. LDL+VLDL fraction. Further, a significant increase in SGPT in liver of mice treated with acute and chronic dose was observed but SGOT increased significantly only in chronic dose treated mice. These results are in confirmation with the results of various studies [16,17,18,19].

The gradual rise in serum cholesterol level could probably be due to cadmium induced changes in the gene expression of some hepatic enzymes such as hydroxyl-methyl-glutaryl Co A reductase (HMG-CoA), which in turn depresses LDL receptors gene expression resulting in reduced endocytosis of cholesterol-rich LDL into the cell[20]. Besides this, the elevated serum triglycerides may attribute to hypoactivity of lipoprotein lipase in blood vessels, which helps in breaking up triglycerides into fatty acid and glycerol[21].

According to Ghorbe[22] rise in the level of transaminases in serum may be due to increased cellular basal metabolic rate, irritability and destructive changes of liver. Thus, increased level of SGPT and SGOT indicate necrotic lesions in the liver[23], which is also supported by histopathological alterations observed in liver of heavy metals treated animals in previous reports[24, 25, 26]. The biochemical changes observed in Cd treated mice during present study may also arise due to Cd induced oxidative stress by generating superoxide, hydroxyl and nitric oxide radicals via fenton reaction[27] result in lipid peroxidation which alters the fluidity of the cell membrane and DNA[28,29]. Hence, increased concentration of lipids and serum transaminases in the present study may partly be attributed to oxidative damage of hepatocytes.

In this study cadmium chloride caused dose and time dependent hepatotoxicity. It is observed that daily exposure to a low dose of cadmium (chronic dose) exhibited more deleterious effects to hepatic tissue than acute dose (single dose) and thus can cause serious health hazards to almost all the organisms of

the biosphere including human beings. Therefore, the extensive toxic potential of the cadmium has warranted the humanity to restrict undesired abuse of the metal so as to save the planet from its dangerous clutches.

Acknowledgement

The authors gratefully acknowledge the facilities provided by Department of Zoology and Environmental Sciences, Punjabi University, Patiala to pursue the research work.

References

1. Julin B, Wolk A, Johansson JE, Andersson SO, Andren O, Akesson A. Dietary cadmium exposure and prostate cancer incidence. A population-based prospect cohort study. British Journal of Cancer. 2012; 107(5): 895-900.
2. Tsalev DL. Atomic Absorption Spectrometry in Occuapational and Environmental Health Practice. CRC Press, Florida. 1993; Vol 1, chap 2: pp 105-112.
3. Duruibe JO, Ogwuegbu MOC, Egwurugwu JN. Heavy metal pollution and human biotoxic effects. International Journal of Physical Sciences. 2007; 2(5): 112-118.
4. World Health Organisation (WHO). 1992. Environment Health Criteria, 134, Cadmium. World Health Organisation IPCS, Geneva.
5. Satarug S, Garrett S, Sens MA, Sen DA. Cadmium, environmental exposure and health outcomes. Environmental Health Perspectives. 2010; 118(2):182-190.
6. Pentyala S, Ruggeri J, Veerraju A, Yu Z, Bhatia A, Desaiah D, Vig P. Microsomal Ca^{2+} flux modulation as an indicator of heavy metal toxicity. Indian Journal of Experimental Biology. 2010; 48: 737–743.
7. Nomiyana K, Nomiyana H. Cadmium-induced renal dysfunction: new mechanism, treatment and prevention. Journal of Trace Elements in Experimental Medicine. 1998; 11: 275-288.
8. Singh P, Mogra P, Bano H, Sankhla V, Deora K, Javeria, S. Protective and preventive effects of curcumin against cadmium chloride induced gastrointestinal toxicity in Swiss albino mice. World Journal of Science and Technology. 2012; 2(12):10-17.
9. Nair AR, De Gheselle O, Smeets K, Kerkhove EV, Cuypers A. Cadmium-induced pathologies: Where is the oxidative balance lost (or not)? International Journal of Molecular Sciences. 2013; 14(3): 6116-6143.
10. Al-Attar AM. Vitamin E attenuates liver injury induced by exposure to lead, mercury, cadmium and copper in albino mice**.** Saudi Journal of Biological Sciences. 2011; 18(4): 395–400.
11. Jihen EH, Imed M, Fatima H, Abdelhamid K. Protective effects of Selenium (Se) and Zinc (Zn) on cadmium toxicity in the liver and kidneys of the rats: Histology and cadmium accumulation. Food and Chemical Toxicology. 2008; 46(11): 3522-3527.
12. Haouem S, EI-Hani AH. Effects of cadmium on lipid peroxidation and on some antioxidants in the liver, kidneys and testes of rats given diet containing cadmium-polluted radish bulb. Journal of Toxicologic Pathology. 2013; 26(4): 359-364.
13. Ige SF, Akhigbe RE, Edeogho O, Ajao FO, Owolabi OQ. Hepatoprotective activities of allium cepa in cadmium-treated rats. International Journal of Pharmacology and Pharmaceutical Sciences. 2011; 3(5): 60-63.
14. Ivanoviene L, Sadauskeine I, Lesauskaite V, Stapulionis R, Ivanov L. Induction of apoptosis by cadmium chloride in mouse liver. Biologija. 2004; 2: 42.
15. Higuchi H, Gores GJ. Mechanisms of liver injury: an overview. Current Molecular Medicine. 2003; (3): 483-490.
16. EI-Demerdash FM, Yousef IM, Kedwany FS, Bagdadi HH. Cadmium-induced changes in lipid peroxidation, blood hematology, biochemical parameters and semen quality of male rats: protective role of vitamin E and beta-carotene. Food and Chemical Toxicology. 2004; 42(10): 1563-1571.
17. Pari L, Murugavel P. Role of diallytetrasulfide in ameliorating the cadmium induced biochemical changes in rats. Environment Toxicology and Pharmacology. 2005; 20(5): 493-500.
18. Yadav N, Khandelwal S. Therapeutic efficacy of Picroliv in chronic cadmium toxicity. Food and Chemical Toxicology. 2009; 47(4):871-879.
19. Elayat W, Bakheelf MS. Effect of chronic lead toxicity on liver and kidney functions. Journal of Medical and Laboratory Sciences. 2010; 1(2): 29-36.
20. Kojima M, Masui T, Nemoto K, Degawa M. Lead nitrate-induced development of hypercholesterolemia in rats: sterol independent gene regulation of hepatic enzymes responsible for cholesterol homeostasis. Toxicology Letters. 2004; 154(1-2):35-44.
21. Terasawa Y, Ladha Z, Leonard SW, Morrow JD, Newland D, Sanan D, Traber MG, Farese RV. Jr. Increased artherosclerosis in hyperlipidemic mice deficient in alpha-tocopherol transfer protein and vitamin E. Proceedings of National Academy of Sciences USA. 2000; 97(25): 13830-13834.
22. Ghorbe F, Boujelbene M, Makni-Ayadi F, Guermazi F, Kammoum A, Murat JC. Effect of chronic lead exposure on kidney function in male and female rats: Determination of a lead exposure biomarker. Archives of Physiology and Biochemistry. 2001; 109: 457-463.
23. EI-Shenawy SM, Hassan NS. Comparative evaluation of the protective effect of selenium and garlic against liver and kidney damage induced by mercury chloride in the rats. Pharmacology Reports. 2008; 60(2): 199-208.
24. Mani U, Parsad AK, Suresh KV, Lal K, Kanojia RK, Chaudhari BP, Murthy RC. Effect of fly ash inhalation on biochemical and histomorphological changes in rat liver. Ecotoxicology Environmental Safety. 2007; 68(1):126-33.
25. Renugadevi J, Prabu SM. Cadmium induced hepatotoxicity in rats and protective effect of naringenin. Experimental Toxicology and Pathology. 2010; 62(2): 171-181.
26. Sharma S, Kaur S, Kaur K. Histopathological and histometric effects of cadmium on liver of albino mice. Biochemical and Cellular Archives. 2013; 13(1):47-51.
27. Watanabe M, Henmi K, Ogawa K, Suzuki T. Cadmium-dependent generation of reactive oxygen species and mitochondrial DNA breaks in photosynthetic and non-photosynthetic strains of Euglena gracilis. Comparative. Biochemistry and Physiology c-Toxicology and Pharmacology. 2003; 134: 227-234.
28. Zama D, Meraihi Z, Tebibel S, Benayssa W, Benayache FS. Chlorpyrifos-induced oxidative stress and tissue damage in the liver, kidney, brain and fetus in pregnant rats: the protective role of the butanolic extract of Paronychie argentea L. Indian Journal of Pharmacology. 2007; 39: 145-150.
29. Mohammed SI. The antioxidant and hepato-protective effects of aqueous Zingiber officinale extract against cadmium bromide toxicity in female wistar rats (Rattus norvegicus) Zanco. Journal of Pure and Applied Sciences. 2011; 23(1): 12.

Cite this article as:

Sandeep Kaur and Suman Sharma. Assessment of Biochemical Alterations Induced by Acute and Chronic Doses of Cadmium in Albino Mice, Asian Journal of Pharmacology and Toxicology; 2015; 1-4.