

HISTOPATHOLOGICAL EFFECT INDUCED BY DIMETHOATE ON LIVER OF ALBINO RATS

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ABSTRACT

Pesticide hazards have been accentuated by the sharp rise in their agricultural, industrial and domestic use. The extensive use of dimethoate poses a health hazard to animals and humans because of its persistence in soil and crops. Our study investigated the toxic effects of the dimethoate at different dose level, an organophosphorous pesticide, on organ like liver of albino rat. Dimethoate was administered orally for one month at doses 1/20th, 1/40th and 1/80th on the basis of LD₅₀. Half of the experimental animals were sacrificed and the rest were left to study the inbuilt capacity of the animals to metabolize the toxicant. Treated mice were showed many histopathological alterations in liver like, vacuolation, blood congestion, and high lymphocytic infiltration around the central vein, enlargement of hepatic sinusoids, hepatocellular damage, necrosis, and increase in number of kuffer cells, lessions and hemorrhage. However, these alterations were recovered in the next set of experimental animals showing the inbuilt capacity of the organism to metabolize the toxicant.

Keywords: Dimethoate, Histopathology, Albino rat.

I. INTRODUCTION

The environmental pollution is one of the most serious problems that faces mankind in this century. There are many types of pollutants that interfere with our-life both directly and indirectly. Furthermore, potential future hazards to human health and wildlife can be created by residues from some long-lived pesticides that may build up in the food chain and cause widespread contamination of the environment (El-Sebae, 1993; Zaahkhouk et al., 2000). Nowadays, the hazards of pesticides have been accentuated by the sharp rise in their use by farmers, industrialists and householders alike. Pesticide poisoning resulting in death occurs mainly in developing countries where the consumption of these products is high (Banerjee et al., 1999). Organophosphate (OP) compounds are the most toxic pesticides causing environmental pollution and potential health hazards (Agdi et al., 2000). Being biologically active compounds, pesticides have been thoroughly tested for safety and usefulness prior to their agricultural use. However their misuse may cause drastic effects to humans, and the environment (GIFAP, 1984).

Dimethoate, are the widely used systemic pesticides in agriculture against a wide range of insects, mites and fungal diseases of fruits, vegetables, or namental plants and field crops as both systemic and contact pesticides and are also used indoor to control houseflies (Meister, 1992; Farag et al., 2006; Brkic et al., 2008 and Muthuvivegandave et al., 2011). The irrational and excessive use of these pesticides plays a crucial role in the occurrence of many disease affecting plants, animals and human (Zaahkhouk et al., 2000 and Al-Haj et al., 2005). The liver is the primary organ involved in xenobiotic metabolism and is a major target organ of

chemicals and drugs. Hepatotoxicity is therefore an important end point in the evaluation of the effect of a particular xenobiotic. Clinical chemistry and histopathological evaluations are commonly used methods for detecting organ specific effects related to chemical exposure (Travlos *et al.*, 1996; Crissmann *et al.*, 2004). Dimethoate has acute oral toxicity of 150mg/kg by in mice and has been classified as moderately hazardous (IPCS/WHO., 2001; Kidd, *et al.*, 1991). Also, Poet *et al.*, 2003, studied the effect of two organophosphorous pesticide on the hepatic and intestinal metabolism in the rat and found that the metabolizing enzymes responsible for both the bioactivities and detoxification are present in the small intestine at lower level than the liver but still significant. Also, Bulusu and ChakravartyIndira *et al.*, 1992, studied nucleic acid and protein propel in normal and malnourished rat liver on exposure to organophosphorous group of pesticides and found that were affected in both. Moreover it has been proven to accumulate in the testis where it persisted for weeks even after oral administration was stopped (Afifi *et al.*, 1991) and inhibited steroidogenesis primarily by blocking transcription of the steroidogenic acute regulatory (STAR) gene (Walsh 2000). Few studies have been made on the histopathological effects of dimethoate (Thangavel *et al.*, 1994; Persis *et al.*, 2001). The present studies have been aimed to investigate the histopathological effects of the dimethoate (an organophosphorous pesticide) that is widely used in some agricultural areas in India on some organ of albino rat.

II. MATERIALS AND METHODS

2.1 Chemicals and Plant extract

All chemicals were purchased from SDFCL (SD Fine-Chem Ltd) and SRL (Sissco Research Laboratories Pvt Ltd). All chemical used were either of analytical grade or the highest purity commercially available. Standard rat feed was purchased from Ashirwad Industries, Mohali, India.

2.2 Animals

The female albino rats aging 9-12 weeks and weighing 140-170 grams were procured from the Department of Livestock Production and Management, GADVASU, Ludhiana and were maintained in laboratory conditions with standard pelleted rat feed and water provided *ad libitum*. The animals were kept in a room in which the humidity and temperature were environmentally controlled. All methods and procedures of animal handling during research were conducted in accordance with the guidelines of Committee for the Purpose of Control and Supervision of Experiments on Animals (CPCSEA), India and experiments conducted in the present study were duly approved by Institutional Animal Ethics Committee (IAEC), Guru Angad Dev Veterinary and Animal Sciences University, Ludhiana vide letter no IAEC/2014/125-53 dated 13/08/2014.

2.3 Experimental design

Female albino rats were acclimatized for ten days in laboratory conditions and were divided into eight groups with six rats in each group.

Group I (Control) animals received feed + distilled water, olive oil and served as control

Group II 1/20th of dimethoate by oral intubation

Group III 1/40th of dimethoate by oral intubation

Group IV 1/80th of dimethoate by oral intubation

GroupV Olive oil for 15 days and left without any treatment for next 15 days then sacrificed.

GroupVI 1/20th of dimethoate for 15 days and left without any treatment for next 15 days then sacrificed.

Group VII 1/40th of dimethoate for 15 days and left without any treatment for next 15 days then sacrificed.

Group VIII 1/80th of dimethoate for 15 days and left without any treatment for next 15 days then sacrificed.

At the end of 15 days, mice from exposed Groups (I, II, III & IV) were anesthetized and dissected, liver was removed and fixed for histopathological investigations using 10% neutral formal saline. Tissue was processed by routine histological techniques, sectioned at 6µm, stained with hematoxylin and eosin. Finally stained sections were examined under the high magnifying microscope and subsequently micrographs were taken. At the end of experimental period, rat from (Group V, VI, VII & VIII) were taken in batches dissected and examined for histopathological investigations.

III. RESULTS

3.1 Histopathological Study of Liver:

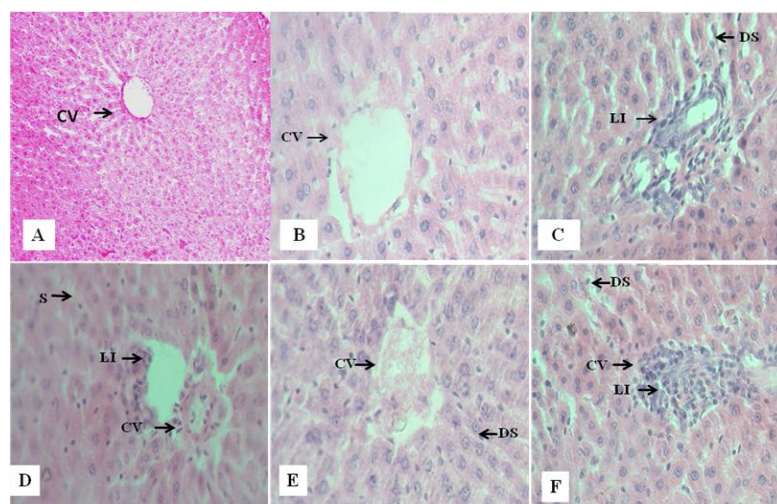
Sections from the liver were prepared from both control and treated mice and were examined under the high magnifying light microscope. The microscopic observations were reported and the intensity of changes was tabulated in Table 1, and presented by the Figs. 1. These changes include the presence of more endothelial cells scattered among the hepatocytes and some vacuolation in the liver texture (Fig A to F). Some sections of liver from mice of treatment groups (I to III) treated with dimethoate pesticides showed rupture in some hepatocytes, many other sections of the treatment sets showed dense lymphocytic infiltration round the central vein and dark stained hepatocytic nuclei indicating cell pycnosis. A number of hepatocytes were found to contain shrunken nuclei, while the liver sections belonging to the metabolized groups showed minor changes represented by group V to VIII.

Table-I Histopathological Changes observed in Liver

Sr. No	Parameter	I	II	III	IV	V	VI	VII	VIII
1	Congestion	+	++	++	+	+	++	+	+
2	Cell rupture	No	Mild	Moderate	Moderate	No	Moderate	No	No
3	Vacuolation	+	+++	++	++	No	+	No	No
4	Lymphocytic infiltration	Occasional	++	++	+	No	++	+	+
5	Nuclear death	No	++	+	+	No	No	No	No
6	Hemorrhage	No	++	+	+	No	periportal round cell collection	No	No
7	Enlargement of control & portal veins	Mild	Moderate	Moderate	Moderate	No	No	No	No

8	Hepatocellular damage	No	++	++	+	No	Perivesicular few hepatocytes focally	No	No
9	Increase in the number of kuffer cells	No	+++	++	+	No	mild	No	No
10	Necrosis	No	++	+	+	No	Focal area show necrosis	No	No
11	Subcapsular stimulation of reticular endothelium	No	++	+	+	No	No	No	No
12	Enlargement of hepatic sinusoids	No	+++	++	++	No	Mild	No	No
13	Parenchymatous cells showing cytoplasmic vacuolization & degeneration in nuclei	No	++	+	+	No	No	No	No
14	Hepatocytes which may show variation in lipid Like bodies of various sizes	Normal	+++	++	+	No	Mild	No	No

Figure 1 Showing Liver sections from different treatment groups of Albino rat



IV. DISCUSSION

The animal model used in this study was used to assess the adverse effects of pesticides on laboratory animals (Costa et al., 1989). While, investigating the histopathological effects of dimethoate pesticides on Liver of albino rat, the results showed mild to severe effects on the organ studied. Some significant results were found while investigating the histopathological parameters. The Histology section of Liver showed to be more severely affected by all the doses of dimethoate. The changes reported include lymphocytic infiltration, congestion, nuclear death, enlargement of hepatic sinusoids, hepatocellular damage, parenchymatous cells showing cytoplasmic vacuolation and degeneration in nuclei. To a bit extent, cell rupture, increase in kuffer cell number, hemorrhage and some hepatocytes which showed variation in lipid like bodies of various sizes were also observed. The role of liver in xenobiotic metabolism is very essential as it contains a large number of enzymes responsible for xenobiotic metabolism. Accordingly as per its role it is more susceptible to chemical injury (Sharkoori, Alam & Ali., 1990). Section through the liver showed chronic inflammation, congestion which could lead to jaundice and death of hepatocyte (necrosis). In an earlier study, Khogali et al. (2005), reported Dimethoate-induced hepatic pycnosis, vacuolation, blood congestion and high lymphatic infiltration around the central vein; Thangevel *et al.*, 1994 reported the toxic effect of an organophosphate pesticide dimecron and acarbamate fungicide cumin on the histology of liver showed the destruction of hepatocytes and protrusion of nuclei. While oral administration of Carbosulfan at 48 mg/kg/day for 5, 10, 20 and 30 days in albino mice resulted in dilation of central vein and sinusoids between hypertrophied hepatocytes, vacuolization and hyalinization of hepatocytes with loss of radial arrangement, suggesting that the insecticide had adverse effects on liver functions leading to histological and physiological impairments (Ksheerasagar and Kaliwal, 2006). The lymphocytic infiltration observed in some controlled mice might be due to the groundnut oil or due to the hygienic condition other than the chemical effects. Liver tissues from mice treated with high doses of the dimethoate showed hepatocytic nuclear death or cell pycnosis. Similar effects were reported by Singh et al., (1994). The evidences of liver damage like cord disarray hypertrophy and disintegration of hepatocytes showing different sizes of nuclei, lymphocytic infiltration, in addition to sinusoidal blood congestion and hemorrhage was also reported by many investigators using various chemicals and toxicants with different animals including fishes (Persis and Kalairasi, 2001; Abdu Rabou., 1996; Shakoori., 1992; Khogali 1989). Recently, El-bendary et al. (2014) evaluated histopathological abnormalities associated with Profenifos and Chlorpyrifos exposure in male albino mice where liver showed hepatic cell damage with degenerative changes including congestion of blood vessels, vacuolar degeneration of hepatic cells, focal infiltration and mononuclear cells, dilated central vein and other hepatic blood vessels, necrosis of hepatic cells, disorganized with the formation of denoid structure and hepatocytomelagy; whereas kidney showed hemorrhage, oedema, necrosis and glomeruli shrinkage. Since, the present feed treatments of albino rat with Dimithoate clearly demonstrated significantly changes in the histopathology of hepatic tissue, it could be possible that prolonged exposure to this insecticide in man may play a significant role in aggravating such diseases as chronic liver failure. In addition, structural changes to hepatic tissue such as haemorrhage, congestion, vacuolation and erosion may also lead to acute liver damage. The present results could thus be exploited as a potential biomarker of common insecticide toxicity in human beings.

V. CONCLUSION

The present findings clearly demonstrate that Dimethoate is capable of inducing dosedependent morphometric i.e. histopathological change in the liver of the exposed rat. According to these data, it is suggested that systemic insecticide like Dimethoate exposure might cause hazardous effects, especially at high doses, to man and environment. For field and domestic uses of this insecticide, quantities and mode of usage need to be strictly monitored to minimize the possibility of its exposure to non-target organisms including human beings. This can be achieved through public health education to make people aware of the hazardous effects of this chemical. Due attention also is to be paid for a delayed period of field application of this insecticide to avoid its possible adverse effects to consumers, who should be warned of the potential risk of Dimethoate contamination of food and drinking water in the country.

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