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RESEARCH ARTICLE

Histochemical Effects of Carbohydrates on Liver of Mancozeb Induced Albino Mice

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Abstract

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Pesticides are both a boon and bane for civilization. Along with solving the problem of food crisis for the growing population it causes serious health hazards. The current study observes the histochemical effects with different doses of mancozeb widely used against diseases of field crops, fruits and ornamentals (Worthing, 1991) in the liver of albino mice. Though it has low acute toxicity; yet its breakdown product ethylene thiourea causes major toxicological concern and was found to have toxic effects in number of experimental animals. Three groups of Swiss albino mice were used wherein distilled water at 0.5 ml per mice per day orally was used in group I as control while the same volume of mancozeb dissolved in distilled water at a dose level of 4.2 mg/kg/bw and 6.4 mg/kg/bw were administered consecutively for 6 days a week for 4 weeks in group II and III respectively. The histochemical analysis of the samples revealed significant decrease in the intensity of stain for glucose content with severity in group III as compared to the control values. The results of the present work indicate marked alterations in the content of glucose in mancozeb exposed albino mice.

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INTRODUCTION

The use of pesticides has increased alarmingly nowadays. It has proved to be boon as well as bane for us. It causes serious health hazards much of which is still not known to the farmers who use them. There has been increasing concern about the effects of carbamates and mancozeb in human and experimental animals. Mancozeb, is a polymeric complex of zinc and manganese salt of ethylene bis dithiocarbamate group (EDBC). It is commonly used for foliar application and seed- treatment in agriculture (Worthing, 1991). Despite its low acute toxicity mancozeb has been shown to produce significant toxicological effects on thyroid and gonads of rats and chromosomes of bone marrow cells in mice (Kurinny, 1972 and Kackar, 1997a and b). Mancozeb significantly decreases blood glucose, causes anemia and induces pathological changes in liver, kidney, spleen, and heart (Hore, 1997). Reproductive toxicity in animals has also been reported by various workers (Bindali and Kaliwal, 2002). Pesticide-induced changes in the liver can be regarded as an index for the identification of pollution stress. The liver can store more toxicants than any of the tissues in the body. This increases in the occurrence of adverse effects in the liver and it provides toxicologists a significant site for investigation (Patel, et al., 2001).

The present study was chosen to evaluate the biochemical effects in liver of albino mice by mancozeb. Few studies have been reported on the histochemical effects of mancozeb.

MATERIALS AND METHODOLOGY

Test material

Mancozeb (commercial grade 75% wettable powder) was made available from Indofil Chemical Company, Mumbai and dissolved in distilled water for oral administration at a dose of 4.2 mg/kg bw and 6.7 mg/kg bw.

Treatment of animals

Mice weighing between (25-30gm) were selected randomly and kept in individual cages with 12:12 hour light and dark cycle and at room temperature of 26 ± 10^{0} C. Synthetic pellet diets and water were provided ad libitum. All experiments were performed in compliance with guide for care and use of laboratory animals (National Research Council, 1985). Animals were divided into three groups with 6 mice in each group. The first group was used as control while the second and the third groups were administered mancozeb.

The mice of control group I were given distilled water orally for 6 days a week for 30 days while group II mice received mancozeb at a dose level of 4.2 mg/kg bw/day (MCZ D1) and group III mice were administered 6.7 mg/kg bw/day (MCZ D2) in equivalent volume of 0.5 ml in the same procedure for 4 weeks. **Histochemical Assays**

For the histochemical study, mice from group I, II and III after 15th and 30th day of treatment were sacrificed and dissected to remove the liver tissues. After washing in saline they were preserved in 10% formalin and after routine procedure paraffin blocks were made. Sections were made from the blocks and routine procedure was followed for slide preparation and stained in PAS stain for carbohydrates.

RESULTS AND DISCUSSION:

Histochemical changes

The sections from both control and mancozeb treated mice were examined under light microscope. The intensities of stain were recorded in table 1. Sections from the liver of control mice showed high protein and glucose content while those from group II treated with 4.2 mg/kg/bw showed moderate intensity and group III treated with 6.7 mg/kg/bw showed least intensity. The intensity of staining was used for comparing the carbohydrate contents present in the liver cells of the normal mice with treated mice at different concentrations.

The PAS reaction in control showed normal carbohydrate/ glycogen content as pink granules in liver parenchyma and hepatocytes. Group II showed depletion in the carbohydrate content and some hepatocytes showed PAS negative reaction while some showed PAS positive reaction on day 15. The intensity was further decreased on day 30. In group III the depletion of carbohydrate content was higher than group II and on day 30 the decline as highest.



Fig: 2. (A) Photomicrograph of liver section of control mice showing reddish deeply stained granules due to PAS positive reaction, CV: central vein (B) Photomicrograph of liver section of Gr II 15 days showing few to moderate glycogen content, pinkish granules and PAS reaction, (C) Photomicrograph of liver section of Gr II 30 days showing few glycogen content and moderate PAS reaction, IN: infiltration of inflammatory cells (D) Photomicrograph of liver section of Gr III 15 days showing very few glycogen content and minimum PAS reaction, DCV: degenerated central vein (E) Photomicrograph of liver section of Gr III 30 days showing almost negative PAS reaction, DH: degenerated hepatocytes

Carbohydrates are essential constituents of the food of animals as it the source of energy. The present study shows decrease in glucose content in blood. Some disruption in glucokinase activities and glycogenesis process were seen during toxic intoxication. Abdel Raheem, et al., (1986) reported glycogen depletion in liver of rodenticide –treated rats along with increased glycogenolysis. Similar findings were also reported by Ebaid, et al., (2007). Ksheerasagar and Kaliwal, (2010) observed that treatment with mancozeb led to decrease in protein and glycogen content in testes of mice.

In the present study, hepatic carbohydrate content significantly decreased after treating the mice with mancozeb. Singh and Srivastav, (1981) mentioned that, the carbohydrates decreased as a result of exposure to sublethal concentration of a mixture of aldrin and formothion. Krapagaganapthy, et al., (1988) reported that the carbohydrate content of liver in Colisa lalia showed progressive decrease in staining intensity to PAS at all exposure period when treated at sublethal (0.04 mg/l) and median lethal (0.09 mg/l) concentrations of lindane. This is possibly utilization of carbohydrate to greater extent during the stressful condition. Khare and Singh, (2004)

observed histochemical changes by way of reduction in the carbohydrate contents in the gill of the fish, Nandus nandus exposed to sublethal concentration of endosulfan and carbaryl for one month. Further, the change in the histochemical contents indicates their rapid utilization to provide excess energy in order to cope with stressful conditions.

CONCLUSION

The liver is the most important target organ in a vertebrate body, as it is the chief metabolic and detoxification centre. Many reports indicate that it is the organ with the highest concentration of the pesticide and with the greatest damage or impairment due to pesticide treatment. Since all toxins pass through liver during detoxification and hence manifest the highest toxin. Thus, mancozeb exposure leads to histochemical alterations during two week exposure period and highly significant changes were observed during four weeks exposure period. Similar interpretation was confirmed by others authors on fungicides within the same class (Ugazio, et al .1985). The results of the present study suggest mancozeb has adverse effects on liver functions. The study reveals mancozeb might have affected cell metabolism and cellular defence mechanism and detoxification system in liver.

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