Arch. Pharm. Res. Vol.21, No.3, pp. 223-230, 1998

Antihepatotoxic Potential of *Trianthema portulacastrum* in Carbon Tetrachloride-induced Chronic Hepatocellular Injury in Mice: Reflection in Haematological, Histological and Biochemical Characteristics

Animesh Mandal, Ranajit Karmakar, Subrata Bandyopadhyay and Malay Chatterjee

Department of Pharmaceutical Technology, Division of Biochemistry, Jadavpur University, Calcutta 700-032, INDIA

(Received April 2, 1997)

The effect of an ethanolic extract of the plant *Trianthema portulacastrum* L. on the CCl_4-induced chronic hepatocellular damage of Swiss albino mice has been investigated. The normal mice received olive oil (0.2 ml/mouse) for five weeks. The CCl_4 control mice, on the other hand, received CCl_4 (0.05 ml/mouse) in olive oil for five weeks. The extract was administered at the dose of 100 mg/kg or 150 mg/kg for five weeks by gastric intubation in addition to CCl_4 treatment. The CCl_4 administraction alone caused hepatocellular necrosis, severe anemia, leucopaenia, lymphocytopaenia, neutrophilia, eosinophilia and haemoglobinaemia along with the alterations of plasma albumin and globulin. The administration of plant extract (at 100 or 150 mg/kg) restored the CCl_4-induced alterations of the haematological parameters to the normal level. The extract of *T. portulacastrum* elicited a marked protection against CCl_4-induced hepatotoxicity as indicated by the several haematological parameters, related indices of formed elements, and different fractions of plasma protein. We also observed the dose-dependent antihepatotoxic effect of the extraction on these mice. The 150 mg/kg of extract was found to be more effective in normalizing the toxic effects of CCl_4 on the above parameters of mice. These results suggest that the hepatoprotective effect of *T. portulacastrum* could be caused by its critical involvement in modulating several factors associated with erythropoiesis, and the boosting of general immunity of the host.

Key words : *Trianthema portulacastrum*, Antihepatotoxicity, Haematological parameters, Plasma protein

INTRODUCTION

The plant *Trianthema portulacastrum* L. of Aizoaceae family is a prostrate, glabrous, succulent which can be found in the most part of India, both in agriculatural and waste land (Chadha, 1976). It is popularly used both as a food and in the indigenous medicine in India and Philippines (Chadha, 1976; Kirtikar and Basu, 1975). The taste of the plant, i.e. hot and bitter, stomachic, laxative, alexiteric and has been attributed with alterative, antiulcer, cardioprotective and abortifacient properties (Kivtikar and Basu, 1975; Wahid and Siddqui, 1961; Fazal and Razzact, 1978). The herb is also recommended for the relief of the liver damage, the decoction of this herb is considered as a popular antidote to alcohol (Chadha, 1976). Crude extract of the roots and leaves of this plant have been investigated for their general pharmacodynamic properties, i.e. their actions on the respiration, smooth and skeletal muscles, blood pressure and isolated heart preparations (Gupta and Bhaskaran, 1976). The ethanol extract of the whole plant has been reported to manifest potent antibacterial, analgesic and antiinflammatory activities (Vohora *et al.*, 1983).

Recently, we have documented a morked antihepatotoxic activity of an ethanol extract of the whole plant (excluding the roots) against acute hepatocellular injury induced by alcohol and carbon tetrachloride (CCl_4) in mice (Bishayee *et al.*, 1996). The extract exerted a significant protection against hepatic lipid peroxidation, an elevation of hepatic glutathione (GSH) and the restoration of a number of hepato-specific enzymes (Bishayee *et al.*, 1996). In CCl_4-induced chronic liver damage in mice, we have also found the beneficial response of the extract which has been fairly reflected in the normalization of altered serum transaminases

Correspondence to: Malay Chatterjee, Department of Pharmaceutical Technology, Division of Biochemistry, Jadavpur University, Calcutta 700-032, INDIA

as well as the restoration of enzymes of plasma membrane, microsomal, lysosomal and cytoplasmic fractions of hepatic tissue (Mandal *et al.*, 1997a). In order to understand the underlying biochemical mechanism of this plant we have measured the level of hepatic lipid peroxidation, the modulation of GSH and the activities of several antioxidant defence enzymes (Mandal *et al.*, 1997b). We observed the hepatoprotective activity of *T. portulacastrum* is mediated by the reduction of hepatic lipid peroxidation, increment of GSH along with alterations of other enzymic activities.

Our present communication embodies the effect of the ethanol extract of *T. portulacastrum* on the haematological status, haematopoietic system and plasma protein levels of mice during CCl_4-evoked chronic hepatocellular injury in order to delineate the probable involvement of these factors in the antihepatotoxic responses of this herb.

MATERIALS AND METHODS

Extraction

Fresh plants of *T. portulacastrum* were collected from the locality of Memari in Burdwan district (West Bengal, Indai). The plant was identified by Dr. M. Sanjappa, Deputy Director, Central National Hebarium, Botanical Survey of India, Howrah, W. B., India and voucher specimen (No. JU/Pharm/Biochem/Sp. 03) was deposited in it. The extraction of the plant material has already been described in detail in our previous report (Bishayee *et al.*, 1996). In short, the air-dried and pulverized plant material was extracted overnight using a Soxhlet-extractor with petroleum ether (b.p. 60~80°C). After discarding the petroleum ether extract, the residue was successively extracted with benzene, chloroform and acetone. The residue was finally extracted with ethanol (95% v/v) and the ethanol extract was saved. On the completion of evaporation of ethanol extract, the material (yield 4.6% w/w) was stored at 4°C. The extract was dissolved in distilled water just prior to use for animal feeding.

Animals

Male Swiss albino mice weighing 25~30 g were purchased from the Indian Institute of Chemical Biology (Calcutta, India). The mice were kept in a cage and maintained under the standard conditions as follows: 23±1°C, 65±10% relative humidity and 12-h light/12-h dark cycle. All animals were fed with commercial diet (Hindustan Lever Ltd., Mumbai, India) and tap water *ad libitum* during the entire term of our study.

Experimental design

Following an acclimatization period of one week, the animals were divided into four different groups taking six mice in each. Group A animals (vehicle control) were given distilled water *per os* at 1 ml/kg once daily for successive seven weeks and olive oil (0.2 ml/mouse) three times a week (every other day) for the last five weeks through a stomach tube 1 h following distilled water feeding. Group B animals (CCl_4 control) were administered distilled water in similar way for consecutive seven weeks and 0.2 ml of 20% CCl_4 (Merck, Darmstadt, Germany) in olive oil per mouse (CCl_4:0.05 ml) three times a week (every other day) for the last five weeks through gastric intubation according to published regimen (Okazaki *et al.*, 1985) 1 h after distilled water feeding. Group C and D (experimental groups) were treated with the ethanol extract of *T. portulacastrum per os* at 100 and 150 mg/kg respectively once daily for successive seven weeks and with CCl_4 for the last five weeks 1 h following the plant extract treatment as stated above. All animals from each group were fasted overnight prior to sacrifice.

Haematological assessment

At the end of the seventh week, blood from mice of different groups were taken by heparinised syringe directly through cardiac puncture. All samplings were performed between 10:00 and 11:00 h in order to avoid diurnal variation of the parameters observed in this study. Red blood cells (RBC) were isolated by centrifugation at 600 g for 10 min followed by two-fold washing with 0.15 M NaCl and recentrifugation under the same condition. Total counts of RBC and white blood cells (WBC) were determined with the help of haemocytometer. Differential counts of WBC (e.g. neutrophils, eosinophils, basophils, lymphocytes and monocytes) were carried out by staining the WBC with Leishman's stain. The determination of erythrocyte sedimentation rate (ESR) was performed by the standard physiological method. haemoglobin (Hb) was quantitiated by cyanohaemoglobin colorimetric method as per standard technique. Packed cell volume (PCV) was estimated by centrifugation the blood sample in heparinised capillary tube at 11,000 g for 6 min. All measurements were repeated. Mean corpuscular volume (MCV), mean corpuscular haemoglobin concentration (MCHC) were calculated using the following formula:

$$\text{MCV } (\text{um}^3) = \frac{\text{PCV (ml/L)}}{\text{RBC } (10^6/\text{mm}^3)}$$

$$\text{MCH (pg)} = \frac{\text{Hb (g/dl)}}{\text{RBC } (10^6/\text{mm}^3)}$$

$$\text{MCHC (\%)} = \frac{\text{Hb (g/dl)}}{\text{PCV (ml/dl)}} \times 100$$

Estimation of plasma protein

Total plasma protein and two plasma protein fractions e.g. albumin and globulin were assayed using the routine technique (Lowry *et al.*, 1951). Before the estimations were carried out, albumin and globulin were separated by salt fractionation as described elsewhere (Mandal *et al.*, 1993)

Histological study

Portions of liver tissue were collected immediately after sacrifice in 10% buffered formalin for fixation. The tissue was embedded in paraffin (60~62°C). Sections of 5 µm were cut, streched on grease free slides, deparaffinised in xylene and rehydrated through 100, 90, 70, 50 and 30% ethanol and water. Serial sections were taken for staining with haematoxylin and eosin (H and E) stains by conventional technique. Any hepatocellular lesions observed in H and E staining were recognised by an Adcon-5591 photomicroscope. All slides were examined without prior knowledge of the treatment given to the animals from which the specimen under investigation was taken.

Statistical analysis

The obtained data were analysed using Student's *t*-test. $p<0.05$, $p<0.01$ and $p<0.001$ were considered levels of significant.

RESULTS

The administration of CCl_4 significantly decreased the total RBC count (Fig. 1; $p<0.001$), Hb concentration (Fig. 3; $p<0.01$), PCV (Fig. 4; $p<0.001$), MCH (Fig. 6; $p<0.001$) and MCHC (Fig. 7; $p<0.001$) in group B mice with concomitant significant increase in values of ESR (Fig. 2; $p<0.001$) and MCV (Fig. 5; $p<0.001$) when the comparisons were made with the normal animals (group A) in relation to each parameters. The CCl_4 was found to have significantly altered total RBC count along with the RBC related parameters (formed elements) in group B animals, whereas the effect of *T. portulacastrum* was evaluated, a significant increase ($p<0.05$) in RBC count was noticed in group C animals and a more higher significant increment ($p<0.01$) was recorded in group D mice as compared to the CCl_4-treated control mice (Group B). In case of ESR, the administration of *T. portulacastrum* plant extract exhibited an insignificant inhibition in the rate of sedimentation in group C animals but group D mice showed a significant fall ($p<0.01$) of ESR in comparison with group B counterparts. A significant depletion ($p<0.001$) in Hb concentration caused by CCl_4 in group B mice was also found to restore towards normalization ($p<0.001$) by the plant extract in groups C and D. Sim-

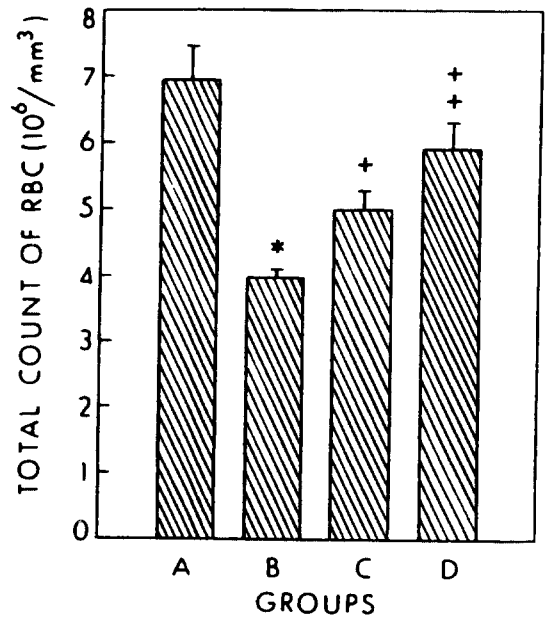

Fig. 1. Effect of *T. portulacastrum* on the total count of red blood corpuscles (RBC) of CCl_4-induced chronic hepatocellular damage in mice. Data represent±SEM of 6 mice. A=Normal mice; B=CCl_4-induced control mice; C=CCl_4+*T. portulacastrum* (100 mg/kg); D=CCl_4+*T. portulacastrum* (150 mg/kg): *=$p<0.001$ (comparison between groups A and B); +=$p<0.05$ (comparison between groups B and C); ++=$p<0.01$ (comparison between groups B and D).

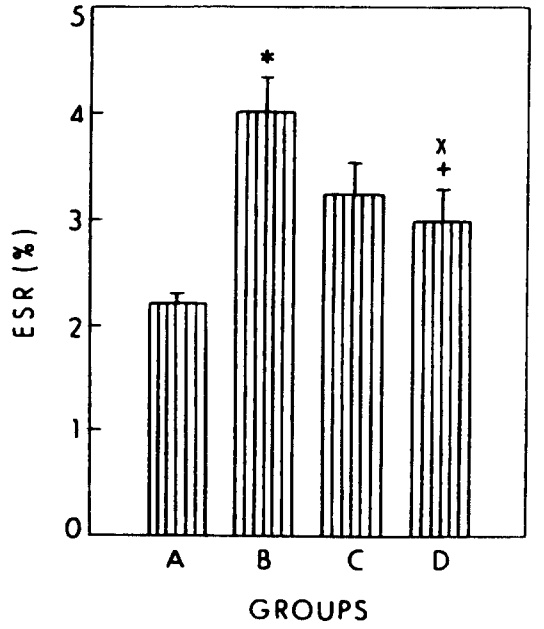

Fig. 2. Effect of *T. portulacastrum* on the erythrocyte sedimentation rate (ESR) of CCl_4-induced chronic hepatocellular damage in mice. Data represent±SEM of 6 mice. A=Normal mice; B=CCl_4-induced control mice; C=CCl_4+*T. portulacastrum* (100 mg/kg); D=CCl_4+*T. portulacastrum* (150 mg/kg): *=$p<0.001$ (comparison between groups A and B); +=$p<0.03$ (comparison between groups B and D); x $p<0.05$ (comparison between groups A and D).

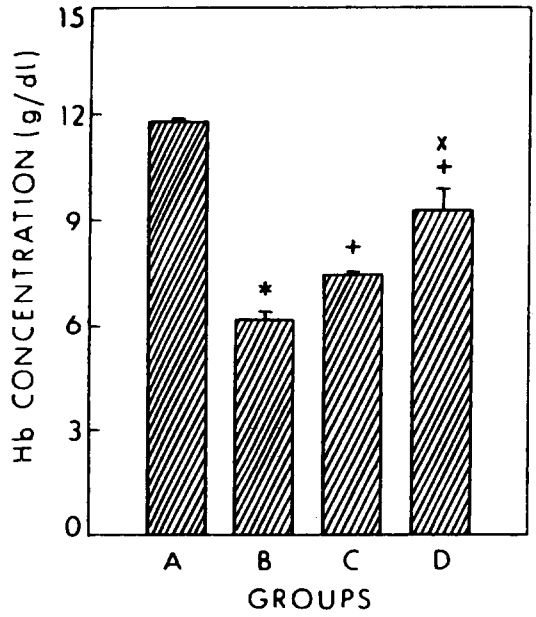

Fig. 3. Effect of *T. portulacastrum* on the haemoglobin (Hb) concentration of CCl_4-induced chronic hepatocellular damage in mice. Data represent±SEM of 6 mice. A=Normal mice; B=CCl_4-induced control mice; C=CCl_4+*T. portulacastrum* (100 mg/kg); D=CCl_4+*T. portulacastrum* (150 mg/kg): *= p<0.01 (comparison between groups A and B); +=p<0.001 (comparison between groups B and C; groups B and D); x= p<0.01 (comparison between groups A and D).

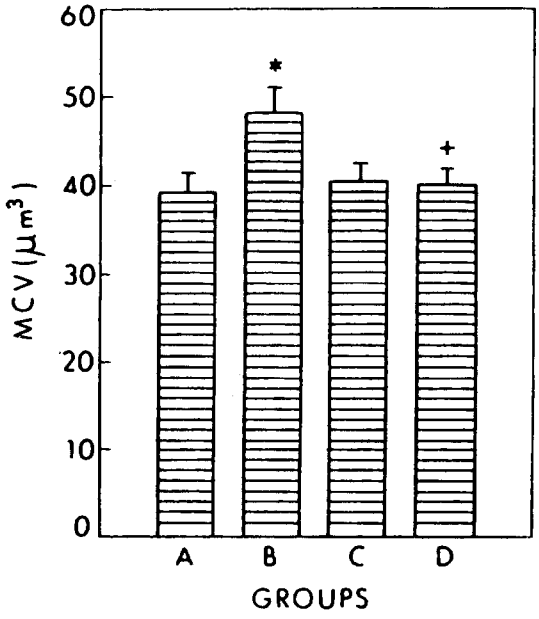

Fig. 5. Effect of *T. portulacastrum* on the mena corpuscular volume (MCV) of CCl_4-induced chronic hepatocellular damage in mice. Data represent±SEM of 6 mice. A=Normal mice; B=CCl_4-induced control mice; C=CCl_4+*T. portulacastrum* (100 mg/kg); D=CCl_4+*T. portulacastrum* (150 mg/kg): *=p<0.01 (comparison between groups A and B); +=p<0.05 (comparison between groups B and D).

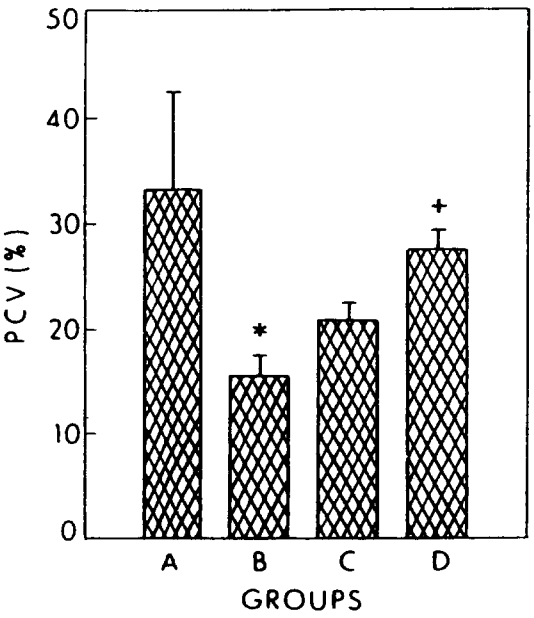

Fig. 4. Effect of *T. portulacastrum* on the packed cell volume (PCV) of CCl_4-induced chronic hepatocellular damage in mice. Data represent±SEM of 6 mice. A=Normal mice; B=CCl_4-induced control mice; C=CCl_4+*T. portulacastrum* (100 mg/kg); D=CCl_4+*T. portulacastrum* (150 mg/kg): *=p<0.001 (comparison between groups A and B); +=p<0.05 (comparison between groups B and D).

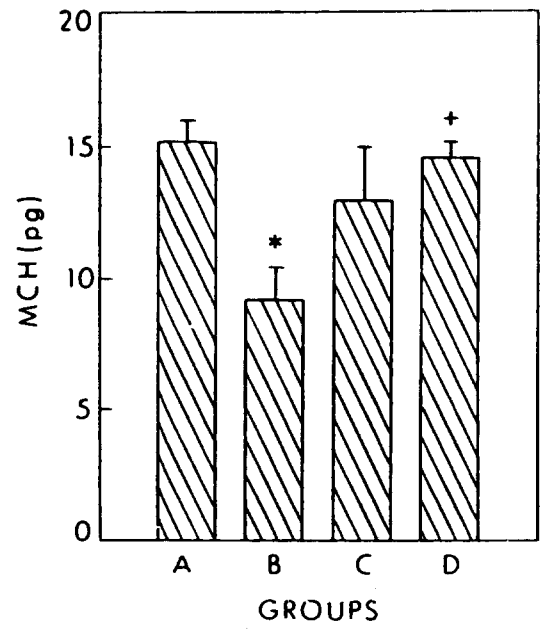

Fig. 6. Effect of *T. portulacastrum* on the mean corpuscular haemoglobin (MCH) of CCl_4-induced chronic hepatocellular damage in mice. Data represent±SEM of 6 mice. A= Normal mice; B=CCl_4-induced control mice; C=CCl_4+*T. portulacastrum* (100 mg/kg); D=CCl_4+*T. portulacastrum* (150 mg/kg): *=p<0.01 (comparison between groups A and B); += p<0.01 (comparison between groups B and D).

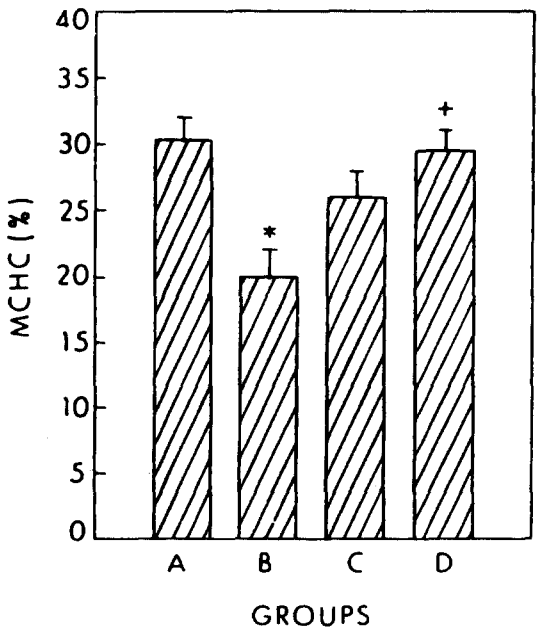

Fig. 7. Effect of *T. portulacastrum* on the mean corpuscular haemoglobin concentration (MCHC) and CCl_4-induced chronic hepatocellular damage in mice. Data represent±SEM of 6 mice. A=Normal mice; B=CCl_4-induced control mice; C=CCl_4+*T. portulacastrum* (100 mg/kg); D=CCl_4+*T. portulacastrum* (150 mg/kg): *=p<0.001 (comparison between groups A and B); +=p<0.01 (comparison between groups B and D).

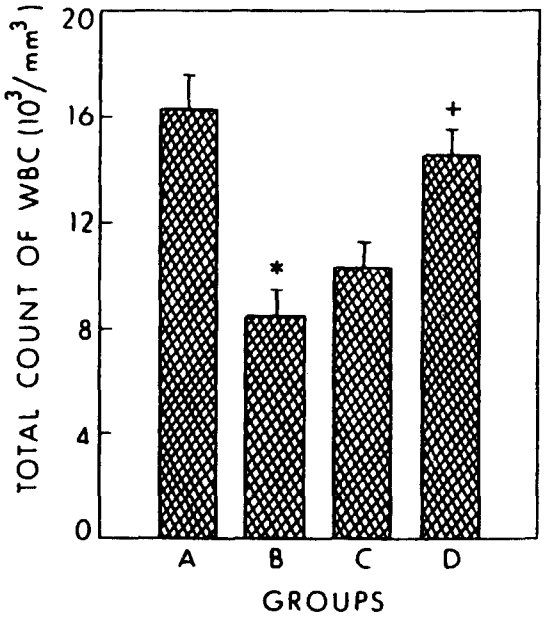

Fig. 8. Effect of *T. portulacastrum* on the total count of white blood corpuscles (WBC) of CCl_4-induced chronic hepatocellular damage in mice. Data represent±SEM of 6 mice. A=Normal mice; B=CCl_4-induced control mice; C=CCl_4+*T. portulacastrum* (100 mg/kg); D=CCl_4+*T. portulacastrum* (150 mg/kg): *=p<0.001 (comparison between groups A and B); + =p<0.001 (comparison between groups B and D).

ilar type of response was observed in PCV of plant extract treated groups. The significantly increased corpuscular volume by CCl_4 administration in group B animals was lowered (p<0.05) in group D animals. The response of MCH and MCHC towards CCl_4 and extract administration featured the same type as found in both Hb concentration and PCV. Total WBC count as in Fig. 8 illustrated a severe leucopaenia in group B mice compared with group A mice. Although the plant extract caused an insignificant elevation in leucocyte population of group C animals, the group D mice on the other hand, showed a further increment (p<0.01) in count towards the normalization.

Whilst the CCl_4 treatment caused a highly significant decrease (p<0.001) in lymphocyte count (24.5%) from the normal value (75.2%) in group A animals (Table I), the neutrophil value was found to show a highly significant elevation (69.9%) from the value (19.2%) of vehicle control mice. The monocyte count did not revela any effect following either CCl_4 or the plant extract treatment. But eosinophils on the other hand, exhibited a significant elevation (p<0.001) after CCl_4 administration. While the *T. portulacastrum* was administered in group C animals, the lymphocyte value increased (p<0.01) but a further increment was also recorded in group D mice (p<0.001) from CCl_4-induced depressed value in group B counterpart. In contrast, in case of neutrophil count, the plant extract was found to decrease the value in addition to highest depletion (p<0.001) in group D mice when compartison was made with the group B individuals. Similarly, the same type of response as recorded in neutrophil value was

Table I. Effect of *Trianthema portulacastrum* on the differential count of leucocytes (%) during CCl_4-induced chronic hepatocellular damage

Groups	Lym	Neu	Mon	Eos
A	75.20±8.26	19.20±2.10	4.38±1.20	1.22±0.80
B	24.50±2.21***	69.90±8.20***	3.31±1.50^{ns}	2.27±0.10***
C	40.00±3.23^{a}	54.20±7.80^{ns}	4.00±1.50^{ns}	0.80±0.50^{a}
D	58.50±5.64^{b}	35.20±8.80^{c+}	4.50±0.90^{ns}	1.80±0.90^{b}

Each value represents the mean±SEM of 10 animals
***p<0.001 (comparison between groups A and B); a=p<0.01 (comparison between groups B and C); b=p<0.01 (comparison between groups B and D); c=p<0.05 (comparison between groups B and D); +=p<0.05 (comparison between groups A and D); ns=Not significant.

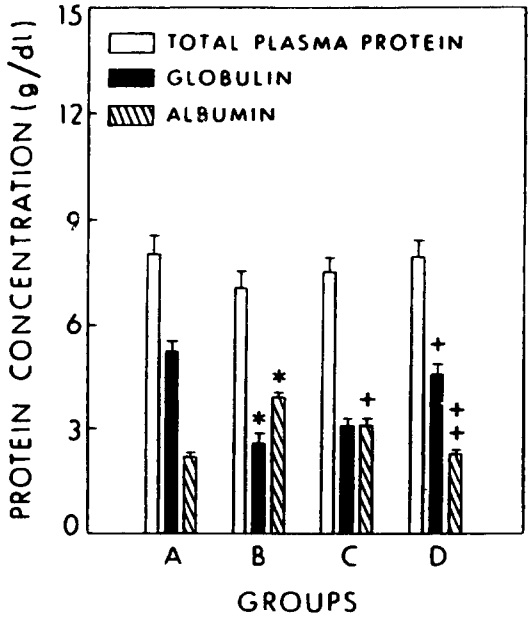

Fig. 9. Effect of *T. portulacastrum* on the plasma protein concentration of CCl_4-induced chronic hepatocellular damage in mice. Data represent±SEM of 6 mice. A=Normal mice; B=CCl_4-induced control mice; C=CCl_4+*T. portulacastrum* (100 mg/kg); D=CCl_4+*T. portulacastrum* (150 mg/kg): *=p<0.001 (comparison between groups A and B); +=p<0.01 (comparison between groups B and C); ++=p<0.001 (com- parison between groups B and D).

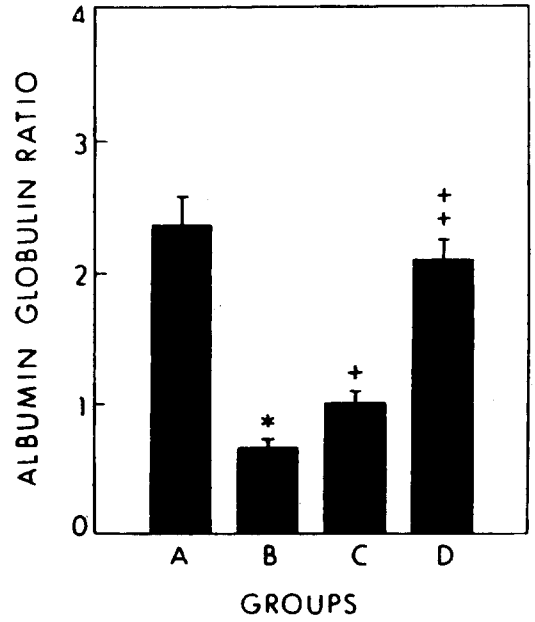

Fig. 10. Effect of *T. portulacastrum* on the albumin: globulin ratio of CCl_4-induced chronic hepatocellular damage in mice. Data represent±SEM of 6 mice. A=Normal mice; B=CCl_4-induced control mice; C=CCl_4+*T. portulacastrum* (100 mg/kg); D=CCl_4+*T. portulacastrum* (150 mg/kg): *=p<0.001 (comparison between groups A and B); +p<0.05 (comparison between groups B and C); ++=p<0.001 (comparison between groups B and D).

found in eosinophil count in different treatment schedules.

As depicted in Fig. 9, neither CCl_4 nor the plant extract could alter the total plasma protein concentration but an inverse relation was revealed between albumin and globulin concentrations excepting in group C animals where both the protein fractions showed more or less the same value. The treatment of animals with CCl_4 induced diminution in albumin concentration ($p<0.001$). A reverse relationship due to the extract administration was recorded between both the fractions in group D individuals. The altered values that recorded in group D mice were found to indicate the changes towards the normalization. The ratio of albumin to globulin (Fig. 10) decreased in CCl_4-treated regimen (group B) but the plant extract caused a slight uplift ($p<0.05$) of the depressed value in group C animals and a further increment towards the normal value in group D mice in comparison with the group B counterparts. When the value of group D animals of each and every parameters was compared with normal mice (group A), ESR (Fig. 2; $p<0.05$), Hb (Fig. 3; $p<0.01$) exhibited significant differences exepting the others.

Histopathology of liver showed drastic damages due to CCl_4administration (Fig. 12) when comparison was made with normal hepatic histology (Fig. 11). The treatment was found to cause extensive vacuolization of cytoplasm with enucleation of cells. The cytoplasm became eosinophilic and the nuclear membrane was found to exhibit crenation. The plant extract at the dose of 100 mg/kg showed a moderate improvement in cellular architecture (Fig. 13) over the CCl_4-induced hepatic damage. But in the higher dose (150 mg/kg) a better improvement was observed as evidenced from the considerable reduction in the hepatocellular necrosis. The vacuoles in cytoplasm formed due to CCl_4-treatment were still present but less in number as compared to the normal counterpart. The crenated nuclear envelope in CCl_4 control animals were found to be nearly regular in 150 mg/kg plant extract treated group and the beneficial effects of the extract were further documented from normal hepatic cord, less necrosis, less fatty degeneration etc.

DISCUSSION

From the results of our present observation, it is quite clear that the extract of the plant *T. portulacastrum*, offers a significant protection of the adverse effect caused by the chronic CCl_4 treatment on different haematological parameters. This finding presumably supports our previous investigations regarding the hepatoprotective role of the plant extract (Bishayee *et al.*,

1996; Mandal *et al.*, 1997a and 1997b). Moreover, an important outcome of the present study is that the higher dose of the plant extract (150 mg/kg) was found to be more effective in normalizing the adverse toxic effects of CCl_4 on different haematological parameters and related indices and confirms the fact that a relatively long-term exposure of *T. portulacastrum* extract affords a greater beneficial effect against toxicant-induced hepatocellular damage.

Since the changes associated with CCl_4-induced liver damages are similar to that of acute viral hepatitis (Robenstein, 1962), CCl_4-mediated hepatotoxicity was employed in present observation. The toxic properties of CCl_4 are caused by the cleavage of the carbon chlorine bond and formation of highly reactive free moieties (CCl_3). This highly trichloromethyl free radical then attacks polyunsaturated fatty acids in the cytoplasmic reticulum and results in the formation of lipid radical. The lipid radical readily reacts with molecular oxygen to produce peroxy radical initiating lipid peroxidation which is considered to be major factor influencing the breakdown and turnover of biomembranes and eventually cause liver injury (Recknagel and Glenda, 1973; Cheeseman *et al.*, 1995).

It is more or less established that the haematological toxicity that observed by peripheral cell counting is not sufficient to indicate a change in quality and quantity of precursor cells in bone marrow rather than to reflect toxicity to more mature haemopoietic tissue (Schofield, 1986). But it is still the way to determine the cytotoxicity on haematological indices (Van der Wilt *et al.*, 1992). As the number of blood cells are essential for normal functioning of immue systems (Schofield, 1986) which helps to determine the end point toxicity.

In present investigation, the CCl_4-treatment resulted in an anemia which was due to depletion in the number of RBC along with the Hb concentration. The destruction in red cells reflects the failure in hepatocellular functions and the various morphological abnormalities in RBCs are probably due to the change in the membrane cholesterol and phospholipid content and/or ratio (Scherlock and Dooley, 1993). The plant extract exerted the beneficial effects in increasing the Hb level with concomitant elevation of related indices (*viz.* MCH and MCHC) which were found to be depressed in the CCl_4-treated regimen. Now, the exact mechanism of extract mediated induction in Hb concentration is not clearly known. It probably influences the process of haem synthesis along with the erythropoietic mecahnism as shown by elevated RBC values in plant extract-treated series.

The CCl_4-induced leucopaenia is associated with cirrhosis and mainly polymorphonuclear leucocytes are greatly affected (Scherlocle and Dooley, 1993). The increased neutrophilia and abnormal mitotic activity are evident from the appearance of abnormal and aberrant forms of leucocytes. The reversal of the process of leucopaenia particularly in higher dose (150 mg/kg) of the extract-treated regimen could be effected through a alteration of lymphoid-myeloid ratio. The normalization of leucocyte count with concomitant retainment of normal Hb concentration with other related indices following *T. portulacastrum* extract treatment in CCl_4-treated group supports the idea of great importance of the extract in modulating the factors by immunohaemopoietic mechanism.

The liver is known to have an important role in the synthesis of plasma protein and different globulins. The CCl_4 has been reported to interefere the tepatic protein synthesis (Magee, 1966; Dianzini, 1979). In our study, the CCl_4 induced a significant alteration in plasma albumin and plasma globulin level with no noticeable change in total plasma protein level. The low level of albumin following CCl_4 administration may be due to reduced synthesis of albumin (Bishayee *et al.*, 1997). The Kupffer cells of liver of reticulo-endothelial system are well known for the antigenicity as they phagocytose many antigens. However, in hepatic injury, the activity of Kupffer cells diminished and many antigens have a low chance of being phagocytosed (Schosield, 1986). So, the high level of globulin may be due to the fact that foreign materials have a high chance of remaining in contact with antibody forming cells. The alterations of albumin and globulin levels by the extract towards the normalization indicates the repairment of impaired protein synthesis by CCl_4 and a simultaneous decrease in globulin fraction suggests that the plant extract improves the functional status of Kupffer cells. From this study, *T. portulacastrum* may be considered as an antihepatotoxic agent because of its role on the alteration of protein synthesis with concurrent improvement of phagocytic activity of Kupffer cells of the liver. However, it warrants further detailed studies to be undertaken in the near future.

The plant extract alters the hepatic architecture towards the normalization along with the normalization of altered hepatic enzyme levels (Mandal *et al.*, 1997b) that strongly points out the possible recovery role over CCl_4-induced hepatic damage. From our previous and present study has been documented that CCl_4 induced centrolobular necrosis, extensive vacuolization in cytoplasm, isolation foci of eosinophilic cells and crenation of nuclear membrane. The above mentioned adverse effects of CCl_4 on hepatic architecture may be due to diminution of endogenous GSH level because GSH has known as important protective biomolecule against CCl_4-induced hepatocellular injury. The elevated lipid peroxidation, on the other hand, may also be responsible for the derrangement of the nuclear envelope (Bishayee *et al.*, 1996). In our present study, we have

also noticed that altered haematological parameters are stabilized by *T. portulacastum* extract which is a clear indication of improvement of functional status of hepatocytes. From ESR and Hb evaluations, it may be concluded that the administration of *T. portulacastum* (150 mg/kg) restored the injured liver towards normalization but from the evaluations of remaining parameters, it may be said that the extract normalized the injured liver.

The results of our present study, thus establish the fact that an ethanol extract of *T. portulacastum* when given at a dose of 150 mg/kg *per os* has a unique beneficial effect on CCl_4-induced toxic effects on various haematological parameters. It has a unique antihepatotoxic potency which has subsequently confirmed by histopathological of hepatic tissue. It is also more or less evident that the beneficial influences are due to the critical involvement in modulating several factors associated with erythropoiesis along with the boosting of general immunity of the host.

ACKNOWLEDGEMENTS

The authors are grateful to the Council of Scientific and Industrial Research (CSIR), ICMR Govt. of India, for providing their financial support.

REFERENCES CITED

Bishayee, A., Karmakar, R., Mandal, A., Kundu, S. N. and Chatterjee, M., Vanadium-mediated chemoprotection against chemical rat hepatocarcinogenesis: Reflection in haematological and histological characteristics. *Eur. J. Cancer Preven.*, 6, 58-71 (1997).

Bishayee, A., Mandal, A. and Chatterjee, M., Prevention of alcohol-Carbon tetrachloride-induced signs of early hepatotoxicity in mice by *Trianthema portulacastrum* L. *Phytomedicine*, 3, 155-161 (1996).

Chadha, Y. R., *The Wealth of India- Raw Materials*, Vol. 10, Council of Scientific and Industrial Research, New Delhi, p. 281 (1976).

Cheeseman, K. H., Albano, E. F., Tomast, A. and Slater, T.F., Biochemical studies on metabolic activation of halogenated alkalines. *Environ. Health Presp.*, 64, 85-102 (1995).

Dianzini, M. U., Reaction of the liver injury. In: *Toxic Injury of the Liver.* (Farber, E. and Fisher, M. M. eds.) Part A, Marcel Dekker, Inc., New York, p. 281 (1979).

Fazal, U. and Razzack, M. A., *A Handbook of Common Remedies in Unani Medicine,* Central Council for Research in Unani Medicine, New Delhi, p. 3 (1978).

Gupta, S. K. and Bhaskaran, M., *Ind. J. Pharm.*, 38, 38-41 (1976).

Kirtikar, K. R. and Basu, B. D., *Indian Medicinal Plants*, Vol. 2, Lalit Mohan Basu, Alahabad, p. 1180 (1975).

Lowry, O. H., Rosenbrough, N. J., Farr, A. L. and Randall, R. J., Protein measurement with Folin phenol reagent. *J. Biol. Chem.,* 193, 265-275 (1951).

Mandal, A., Bishayee, A. and Chatterjee, M., *Trianthema portulacastrum* affords antihepatotoxicity activity against carbon tetrachloride-induced chronic liver damages in mice: Reflection in subcellular levels. *Phytother. Res.*, 11, 216-221 (1997a).

Mandal, A., Bishayee, A. and Chatterjee, M., *Trianthema portulacastrum* reverses lipid peroxidation, glutathione status and activities of related antioxidant enzyme in carbon tetrachloride-induced chronic liver damage in mice. *Phytomedicine*, 4, 239-244 (1997b).

Mandal, P. K., Bandyopadhyay, S., A. and Chatterjee, M., Stimulation of tissue repari by *Mikania cordata* root extract in carbon tetrachloride-induced liver injury in mice. *Phytother Res.*, 7, 103-107 (1993).

Magee, P. N., Toxic liver necrosis. *Lab. Invest.*, 15, 111-116 (1966).

Okazaki, M., Furuya, E., Kasahara, T. and Sakamoto, K., Function of reticuloendothelial system on CCl_4-induced liver injury in mice. *Jpn. J. Pharmacol.*, 39, 503-514 (1985).

Recknagel, R. O., and Glede, E. A., Jr., Carbon tetrachloride hepatotoxicity: an example of lethal cleavage. *CRC Crit. Rev. Toxicol.*, 2, 263-297 (1973).

Robenstein, D., Epinephrine release and liver glycogen levels after catbon tetrachloride administration. *Am. J. Physiol.*, 203, 1033-1037 (1962).

Schofield, R., Assessment of cytotoxic injury to bone marrow. *Br. J. Cancer*, 53, 115-125 (1986).

Sherlock, S. and Dooley, J., *Disease of the Liver and Biliary System*, 9th Edn., Blackwell Scientific Publications, p. 44 (1993).

Van der Wilt, C. L., Van Laor, J. A. M., Gyergyay, F., Smid, K. and Peters, G. J., Biochemical modification of the toxicity and the anti-tumour effect of 5-fluorouracil and cis- of platinum by WR-2721 in mice. Eur. J. Cancer, 28A, 2017-2028 (1992).

Vohora, S. B., Shah, S. A., Naqvi, S. A. H., Ahmed, S. and Khan, M. S. Y., Studies on *Trianthema portulacastrum. Planted Med.*j, 47, 106-108 (1983).

Wahid, A. and Siddiqui, H. H., *A Survey of Drugss*, 2nd Edn., Institute of History of Medicine and Medical Research, New Delhi, p. 110 (1961).