

Hepatoprotective activity of ethanolic extract of *Eclipta alba* in albino rats

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ABSTRACT

200 mg/kg body weight of isolated fraction of *Eclipta alba* was found to be hepatoprotective against CCl₄ induced hepato damage in rats. The experimental protocol was performed as per CPCSEA guide lines.

Key words: Hepatoprotective, *Eclipta alba*, hepatotoxicity, carbontetrachloride.

INTRODUCTION

Eclipta alba (L.) Hassk has been widely used in India for the traditional treatment of liver disorders (Chopra *et al.*, 1966; Mehra and Handa, 1968). An Ayurvedic drug from *Eclipta alba* has been found to be quite beneficial for the treatment of jaundice in children (Dixit and Achar, 1981). Its effect on Na⁺/K⁺ ATPase in hepatic injury has also been studied (Mogre *et al.*, 1981). In vitro immunoinactivation of surface antigen of hepatitis B virus (HBsAg) by *Eclipta alba* has been reported (Thyagarajan *et al.*, 1982). In guinea pigs, hepatoprotective properties of liquid extract from fresh *Eclipta alba* leaves against acute carbontetrachloride (CCl₄) liver damage have been demonstrated (Khin *et al.*, 1978). *Eclipta alba* powder has been found to contraractan increase in liver weight hepatic lipid peroxidation liver glutamyl transpeptidase, serum alanine transferase, serum alkaline phosphatase and serum albumin to globulin ratio induced in rats in vivo by CCl₄ (Chandra *et al.*, 1987). This plant showed antihepatotoxic activity in assays using CCl₄ galactosamine and phalloidin – induced cytotoxicity in cultured rat hepatocytes (Wagner *et al.*, 1986).

MATERIAL AND METHODS

Plant material

Fresh Plant of *Eclipta alba* were collected in August 2006 from the local surround of Vidisha, Madhya Pradesh, India. Dr. P.N. Shrivastava performed taxonomic identification and the voucher specimen was deposited in the herbarium of our laboratory for future reference (vide access no. 32).

Preparation of extracts

The plants were washed thoroughly with tap water and air dried in shade at room temperature. They were then mechanically powdered and sieved. 1000gm of powdered plant material was extracted with ethanolic soxhlation and dried in a rotary evaporator at 40°C. Another 500gm of the powdered plant material was decocted in a 1000 ml of water. The liquid aqueous extract obtained was concentrated in vacuume at 40°C. The extractive yield were found to be 12.5 % and 17.36% for ethanolic and equous extract of *Eclipta alba*, respectively.

Preliminary phytochemical screening:

A preliminary phytochemical screening was carried out for the extracts employing the

standard procedure to reveal the presence of alkaloids, steroids, terpenes, flavonoid, saponins, tannins, glycosides, carbohydrates and proteins.

Animals:

Twenty four albino rats of wistar albino rats weighing 100-200 gm were obtained from laboratory of college. The animals were housed in polypropylene cages and maintained in controlled temperature ($27\pm 2^{\circ}\text{C}$) and light cycle (12 h light and 12 h dark). They were fed with pellets of Golden feed, New Delhi. Water was supplied *libitum*. They were given a week time to get acclimatized with the laboratory conditions. Approval for the study was obtained from the institutional animal ethical committee (IAEC) Reg. No. 804/03/CA/CPCSEA.

Acute toxicity studies

Liver damage was induced in rats by administering carbon tetrachloride subcutaneously in the lower abdomen in a suspension of liquid paraffin in the ratio 1:2 v/v at the dose of 1 ml CCl_4 was administered twice a week, on every first and fourth day of all the 13 weeks.

Treatment schedule

Rats were divided into six groups of 6 rats each as follows. Group I animals served as control, group II animals constituted hepatotoxic rats which received liquid paraffin+ CCl_4 twice a week for 13 week period. Group III, IV and V were the treated group which received liquid paraffin+ CCl_4 +*Eclipta alba* at 50mg, 100mg and 200mg/kg body weight. Group VI received the CCl_4 and Silymerin a standard hepatoprotective drug at 100mg/kg body weight. The rats were received 1 ml water at the dose of 70 mg/kg body weight for 3 months period. All the group were given fresh food daily at 10:30 am and then measuring the food intake by recording the body weight of rats in each group. Animals were kept starved overnight on 90th day. On the next day the blood was collected by making an incision by jugular vein to collect the blood. The liver tissue was dissected by blotting of the blood washed in saline.

Biochemical estimation

Serum was prepared from the collected blood and subjected to biochemical estimation of different parameters like aspartate aminotransferase (AST), alanine aminotransferase

(ALT), alkaline phosphatase (ALP), total lipid, triglyceride, phosphatase and cholesterol. Liver homogenates were also subjected to various biochemical estimation.

Histopathology:

A portion of liver tissue in each group was fixed in 10% formalin (formalin diluted to 10% with normal saline) and proceeded for histopathology. After paraffin embedding block, were stained with Haematoxylin and Eosin and examined under microscope. A few photomicrographs of representative types were also taken. (Fig.1) .

Statistical analysis

One way analysis of variance (ANOVA) followed by Scheffes test was applied for determining the statistical significance of difference in enzymes, protein and lipids levels between different group. The level of significance was set at 0.05.

RESULTS

Total biochemical parameters were recorded in the present study. These include AST, ALT, ALP, TP, triglyceride, cholesterol and phospholipids regarding AST in the blood serum the control value were 22.87 ± 0.02 were achieved when higher dose of *Eclipta alba* 200 mg was given to the hepatotoxic rats. Values were little less as compared to the Silymerin. Regarding ALT the higher doses of *Eclipta alba* showed a significant increase as compared to the control group. Regarding ALP the value remained to be a bit higher as compared to the control group and Silymerin treatment. As regards the total proteins the 200 mg dose of *Eclipta alba* brings the protein value at most equal to the normal rats. As regards the total lipid the value got increased in 50 mg dose but at higher dose, it got decreased which is still much higher than the total lipid values in the control rats. Regarding triglycerides the value was a little higher in 200 mg/kg body weight dose as compared to the control group. Cholesterol value came to be nearer to the control group of animals. There was a considerable increase in the phospholipids of the plant extract treated group VIth serum as compared to the group I which recorded only LP (liquid paraffin). Thus, it is quite clear that, total lipid, cholesterol and phospholipids were found to be increased against

Table 1: Protective effect of Eclipta alba whole plant extract and Silymerin on different biochemical parameters in the serum of rats

Parameter	Group - I LP only	Group - II LP ± CCl4	Group - III LP±CCl4±EA 50 mg/kg b.w.	Group - IV LP±CCl4±EA 100 mg/kg b.w.	Group - V LP±CCl4±EA 200 mg/kg b.w.	Group - VI CCl4±Silymer 100 mg/kg b.w.
Aspartate transaminase%	22.87±0.02	34.78±1.34	34.10±1.06	28.26±1.08	24.72±1.08*	25.8±1.02
Alanine transaminase%	27.62±1.02	72.12±4.11	69.02±2.14	40.12±2.12	35.21±1.16*	26.82±1.16
Alkaline phosphatase%	74.42±3.14	139.12±5.0	120.12±4.0	99.20±3.57	79.02±3.24*	75.12±3.52
Total protein (g/100ml serum)	6.74±0.74	4.32±0.19	4.30±0.18	5.14±0.15	6.41±0.21*	6.68±0.42
Total lipid (g/100ml serum)	198.65±5.62	302.66±8.73	282.22±8.73	246.89±8.73	215.98±8.12**	9.41±0.84
Tryglycrides (g/100ml serum)	8.21±0.82	14.84±1.34	13.46±1.24	11.24±1.02	10.12±0.89*	9.41±0.84
Cholesterol (g/100ml serum)	64.86±3.36	99.61±4.46	82.16±4.26	75.42±3.87	69.15±3.57*	65.75±3.57
Phospholipid (g/100ml serum)	122.65±8.91	240.76±12.88	240.62±12.88	192.26±11.46	180.62±11.02**	199.56±10.02

*P<0.50 as compared to group - I ** P <0.10 as compared to group - II Value are mean ± SE from 6 animals of each group.

Table 2: Effect of Eclipta alba on different biochemical parameters in the liver of rats.

Parameter	Group - I LP only	Group - II LP ± CCl4	Group - III LP±CCl4±EA 50 mg/kg b.w.	Group - IV LP±CCl4±EA 100 mg/kg b.w.	Group - V LP±CCl4±EA 200 mg/kg b.w.	Group - VI CCl4±Silymer 100 mg/kg b.w.
Total Protein (g/100ml serum)	8.14±0.28	5.04±0.21	5.86±0.38	6.96±0.22	7.99±0.22	7.42±0.24
Total Lipid (g/100ml serum)	7268±618	9982±436	9012±402	8124±492	7608±514	7204±346
Tryglycrides (g/100ml serum)	2238±84.3	3329±78.6	29.92±68.2	2546±88.6	2278±64.6	2256±78.3
Cholesterol (g/100ml serum)	1289±63.2	2438±74.8	2183±81.2	1496±88.6	1316±91.2	1396±68.6
Phospholipid (g/100ml serum)	3680±461	3721±354	3720±354	3718±486	3714±496	3718±485

*P<0.50 as compared to group - I Value are mean ± SE from 6 animals of each group.

the control group 1st. Rest parameters were found to be all most normal level when three different doses of the compound of the *Eclipta alba* were given to the CCl₄ induced hepatotoxic rat.

DISCUSSION

The results in the present study indicate that 200 mg/kg body weight dose of the plant extract was able to reduce major elevated biochemical parameters due to the changes associated with CCl₄ induced liver damage in the experimental rats. The level of total protein was found to be at normal level after the treatment. Similar results have been observed by Pattanayak and Priyashree (2008) which have noticed such changes in the hepatotoxic animal when treated with the extract from *Dendrophoe falcata*. Similarly, level of triglyceride, total lipid and cholesterol was found to be decreased after the treatment. Vanukumar and Latha (2002) have also reported that when liver cell plasma membrane is damaged, a variety of enzymes normally located in the cytosol which are released in blood stream which causes hepatocellular changes. Treatment of CCl₄ increases the level of total protein ,

cholesterol in the liver. But it was noticed that the recovery beginning as soon as the plant extract was given. The phospholipids content in serum registered a significant like that of liver showed a deamination in CCl₄ administered group. A histopathological study of the liver further suggest the hepatoprotective efficacy of *Eclipta alba* extracts (Fig. 1). Kumar & Mishra (2008) have also reported hepatoprotective effect of *Pergularia demia* ethanol extract.

The observations of these dose, clearly indicate the involvement of hepatoprotective active principles in *Eclipta alba*. The detail phytochemistry of the active hepatoprotective principles is still awaited.

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