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HEPATOPROTECTIVE EFFECT OF *POLYGONUM PERSICARIA* (LINN.) AQUEOUS EXTRACT ON CARBON TETRACHLORIDE INDUCED HEPATOTOXICITY IN ALBINO RATS

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ABSTRACT

The role of *Polygonum persicaria* was investigated for the prevention of CCl_4 induced liver damage. Twenty albino wistar rats were allotted to five groups *i.e.* control, CCl_4 induced hepatotoxicity and hepatotoxicity with *Polygonum persicaria* treated with 200 and 400 mg/kg body weight. Rats were scarified after 14 days. Toxicity was performed using 12 rats. They were randomly divided into three groups *i.e.* control and treated with 200 and 400 mg/kg *Polygonum persicaria* and one group were treated with standard silymarin. Blood samples were collected for biochemical analysis. Mean serum AST, ALT, ALP levels and TB concentration, were significantly increased in CCl_4 induced hepatotoxic group of rats compared to the control (P<0.001). However significant reduction in these parameters were found in groups treated with *Polygonum persicaria*. Anaemia was evident in the group received CCl_4 . The severe fatty changes in the livers of rats caused by CCl_4 were decreased in the treated groups. Toxicity evaluation of similar doses of the plant revealed no alteration in the parameters measured above except in the higher dose few scattered fatty changes in the liver was present. Liver injury was confirmed by the histological changes.

Keywords: CCl₄, Hepatoprotective, *Polygonum persicaria*, Silymarin, Biochemical analysis, Histopathology.

1. INTRODUCTION

Liver is probably the biggest organ in human body and the main site for extreme digestion and discharge. So it has an amazing job in the support, execution and controlling homeostasis of the body. It is associated with practically all the biochemical pathways to development, battle against infection, supplement supply, vitality arrangement and proliferation [1]. The significant elements of the liver are carbohydrate, protein and fat digestion, detoxification, discharge of bile and capacity of nutrient. In this way, to keep up a solid liver is a vital factor for in general wellbeing and prosperity. Be that as it may, it is ceaselessly and variedly presented to ecological poisons, and manhandled by poor medication propensities, and liquor and endorsed and over-thecounter medication which can in the end lead to different liver affliction like hepatitis, cirrhosis and alcoholic liver ailment [2, 3]. Accordingly liver sicknesses are a portion of the deadly ailment on the planet today. They represent a genuine test to global general wellbeing. Current medications have little to offer for mitigation of hepatic illnesses and it is essentially the plant based arrangements which are utilized for their treatment of liver issue. In any case, there are very little medications accessible for the treatment of liver issue [4, 5].

Along these lines, numerous people cures from plant starting point are tried for its potential cancer prevention agent and hepatoprotective liver harm in trial creature model. Carbon tetrachloride (CCl_4) initiated hepato-toxicity model is generally utilized for the investigation of hepatoprotective impacts of medications and plant removes [6, 7].

Polygonum persicaria Linn. (commonly called ladys thumb) is a herbaceous flowering plant belonging to family Polygonaceae. This tall perennial herb grows in moist, shady areas on higher ground in the North of India (Punjab, Kashmir, Sikkim and Himmalyan Region), England, Southern Scotland, Europe, central Asia and west of the Rockies in North America, It flowers in May and June. The plant may be propagated by division of the root stock. The rhizome is odorless, but powerfully astringent in taste, as it contains tannin to the extent of 21 percent. The bitter taste is due to anti-nutritional factors such as alkaloids, saponins, tannins and glycosides [8].

2. MATERIAL AND METHODS

2.1. Plant material

The fresh plant material *polygonum persicaria* was collected from Lethpora, Pampora Kashmir near Jhelum river and was identified by undersigned at centre for biodiversity and Taxonomy, department of Botany, University of Kashmir. With voucher specimen Herbarium No. 2925-(KASH).The dried plant materials were then pulverized into coarse powder in a grinding machine.

2.2. Preparation of aqueous extract

The prepared powder weighing 500 g was macerated in 1000 ml of distilled water for 72h. It was then filtered and filtrate was dried in Petri dishes and concentrated to dark green residue by heating at 40°C, till complete evaporation of water was achieved.

2.3. Animals

Male wistar rats $(130\pm10g)$ were used for the study. The animals were housed in well ventilated cages and given standard rat chow and water *ad libitum*. The animals were maintained at a controlled condition of temperature of 26-28°C with a 12 h light: 12 h dark cycle. Experiments were done according to OECD guidelines, after getting the approval of the Institute's Animal Ethics Committee (IAEC), to Pinnacle Biomedical Research Institute (PBRI) Bhopal, India (Reg. No.1824/PO/Ere/S/15/CPCSEA). Animals were treated and cared for in accordance with the guidelines recommended by the Committee for the Purpose of Control and Supervision of Experiments on Animals (CPCSEA), Govt. of India.

2.4. Experimental design for hepatoprotective activity

Animals were divided into five groups of six rats each. Group I served as normal received only the vehicle (5% gum acacia; 1 ml/kg; *p.o.*) and Group II as a toxic control, (received only CCl₄. Group III animals were treated with standard silymarin at an oral dose of 100 mg/kg and group IV-V received the aqueous extract of *Polygonum persicaria* at an oral dose of 200 and 400 mg/kg, as a fine suspension of 5% aqueous gum acacia. The treatment was continued for 14 days, once daily. On the day of 14 for groups III-V, 30 min post-dose of extract administration animals received CCl₄ at the dose of 1.5 ml/kg (1:1 of CCl₄ in olive oil *i.p.*).

2.5. Chemicals and reagents

The chemicals and reagents were procured from Sigma-Aldrich Chemicals USA. Remaining analytical grade chemicals and estimation kits were purchased from commercial sources and used.

2.6. Biochemical analysis

The rats were anesthetized with ether and blood samples were collected in tubes for biochemical analysis by the retro-oribital puncture method. Blood samples were centrifuged for 10min at 3000rpm to separate the serum. After blood collection, the animals were sacrificed using ether anaesthesia and the liver tissue was harvested for biochemical using standard techniques [9].

2.7. Histopathology

After the experimental period animals were decapitated, the liver expelled quickly, cut, and washed in saline. The tissues of liver were fixed in 10% formalin and embedded in paraffin wax. Sections of 4-5 microns thickness were made using rotary microtome. The pieces of liver were prepared and installed in paraffin wax. Sections were taken and stained with eosin and hematoxylin and photomicrographs were shot for histopathological observation [10].

2.8. Statistical analysis

All values were expressed as mean \pm SD (n=6 in each group). One-way ANOVA was applied to test for the significance of biochemical data of the different groups. Significance is set at p<0.001.

3. RESULTS

The present study had been attempted to demonstrate the role of hepatoprotective activity of crude aqueous extract of plant material of Polygonum Persicaria belonging to the family Polygonaceae in carbon tetrachloride induced hepatotoxicity at different doses. The animals treated with toxic doses of carbon tetrachloride had markedly elevated values of the serum ALT, AST, ALP and total bilirubin compared to normal rats, indicating acute hepatocellular damage. The toxic effect of CCl₄ was controlled in the animals treated with P. Persicaria (200 and 400 mg/kg, p.o.) (Table 1). Serum enzyme values in the animals pretreated with aqueous extract of Polygonum Persicaria (200 and 400 mg/kg; p.o) were significantly (p < 0.001) lower than those of toxic control values and except for ALP. ALT, AST, total bilirubin serum enzyme values in treated animals were similar to the normal control values. The effects of the aqueous extract of *Polygonum persicaria* were comparable to that of standard silymarin activity.

3.1. Histological Studies

Histopathological examination of liver sections of control group showed normal cellular architecture with distinct hepatic cells, sinusoidal spaces and central vein (Fig.1). Disarrangement of normal hepatic cells with centrilobular necrosis, vacuolization of cytoplasm and fatty degeneration were observed in CCl_4 intoxicated animals (Fig. 2). The liver sections of the rats treated with aqueous extract of *Polygonum Persicaria* and silymarin followed by CCl_4 intoxication showed a sign of protection as it was evident by the absence of necrosis and vacuoles (Fig. 3, 4 and 5).

Table 1: Effect of pretreatment with aqueous extract of *Polygonum Persicaria* (P.P) & Silymarin on CCl₄-induced hepatotoxicity in rats.

Groups	ALT (IU/L)	AST (IU/L)	ALP (IU/L)	TB (mg/dl)
Normal Control	54.35±5.81	139.7±11.41	140.35 ± 6.82	0.50 ± 0.09
CCl ₄ Toxic	209.41±10.78	287.53 ± 59.87	345.28±75.43	2.04 ± 0.27
$P.P.200 \text{mg/ml}+CCl_4$	119.7±49.06	221.73±16.27	254.74±37.28	1.49±0.08
$P.P.400 \text{mg/ml}+\text{CCl}_4$	83.73±10.68	184.48±20.99	184.78 ± 25.71	0.81 ± 0.07
Silymarin-100 mg/kg+CCl ₄	60.88 ± 5.29	150.25 ± 15.20	151.16±11.63	0.55 ± 0.08

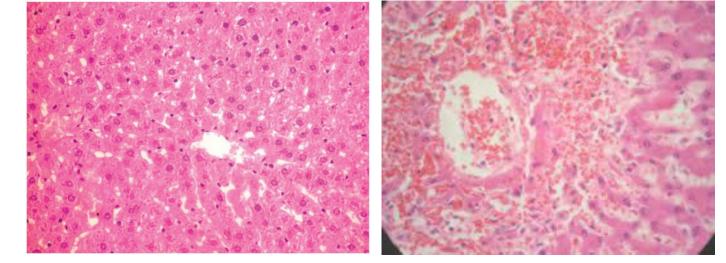


Fig.1: Normal Group I

Fig.2: CCl₄group-II

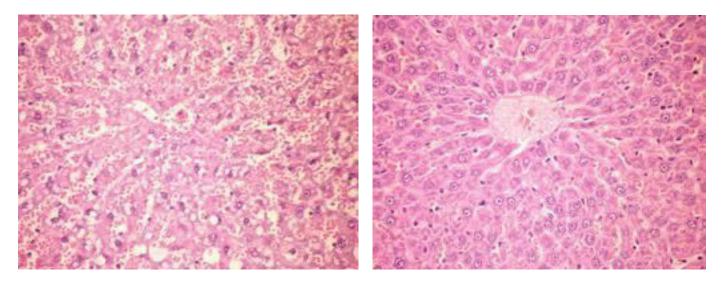


Fig.3: Silymarin + CCl₄ group-III

Fig.4: P.P 200mg/kg+ CCl₄ group-IV

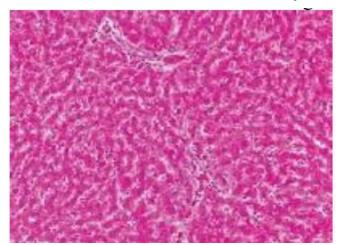


Fig.5: P.P 400mg/kg+ CCl₄ group-V

4. DISCUSSION

In general, CCl_4 has been broadly utilized as a model to study hepatic harm and utilized as a marker of defensive action of newfound medications [11]. In the present investigation, CCl_4 was utilized as a poisonous operator and the hepatoprotective impact of *Polygonum Persicaria* was considered against the CCl_4 -prompted hepatotoxicity. The degree of poisonous quality was assessed by biochemical markers like AST, ALT, ALP and total bulirubin levels in serum; and histopathological investigations of liver tissues. The present investigation reports the potential hepatoprotective action of *Polygonum Persicaria* against hepatic damage delivered by CCl_4 in Wistar albino rats.

It is outstanding that CCl_4 is biotransformed by the Cyt P450 framework to create a without trichloromethyl free radical, which further experiences decrease to shape a trichloromethyl peroxyl radical, which lastly prompts cell demise [11,12]. Subsequently, spillage of enormous amounts of compounds into the circulatory system is frequently connected with monstrous rot of the liver [13, 14]. Our examination validates past discoveries and inspired a huge increment in the exercises of serum catalysts (AST, ALT, and ALP in serum); Total bilirubin, on introduction of rats to CCl_4 , showing significant hepatocellular damage [13, 15].

Treatment with *Polygonum persicaria* reversed the changes produced by CCl₄. *Polygonum persicaria* significantly decreased the levels of above-mentioned biochemical markers to near normal levels.

As expected, silymarin kept up typical design with insignificant wounds and preferred security than *Polygonum persicaria* alone. Interminable treatment with silymarin likewise outlined stamped recuperation in serum compound levels and oxidative parameters in liver tissue. Photomicrograph reveals that the *Polygonum persicaria* functions as a hepatoprotectant. The histopathological profile reveals a significant harm in the toxic group. In this way, it obviously expresses that, poisonous quality is expected to both of the above instruments. Concurrent treatment of the extract and silymarin with CCl_4 displays less harm to the hepatic cells when contrasted with the rats treated with the lethal category.

The *Polygonum persicaria* showed a significant result. Previous reports show flavonoids and steroids are responsible for hepatoprotective effect [16-19]. The plant contains numerous bioactive substances, for example, flavonoids (vitexin, isovitexin, orientin, and isorientin), anthraquinones, especially in roots (emodin and chrysophanol), carotenoids, nutrients (particularly nutrient C), proteins, lipids, and natural acids. Flavonoids present in the plant might be liable for the stamped hepatoprotective impacts, as saw in the present investigation.

5. CONCLUSION

With the aid of enzyme levels and histopathological studies of rat liver, we can conclude that *Polygonum persicaria* has potent hepatoprotective action against CCl_4 -induced hepatic damage in rats. The present study, thus, justifies the traditional use of *Polygonum persicaria* in the treatment of liver diseases and also points out that *Polygonum persicaria* warrants future detailed investigation as a promising hepatoprotective agent.

6. ACKNOWLEDGEMENTS

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Conflict of Interest

The authors declare that; there is no conflict of interest.

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