HEPATOPROTECTIVE ACTIVITY OF ALCOHOLIC AND AQUEOUS EXCTRACTS OF WEDELIA CHINENSIS

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Summary

The hepatoprotective effect of alcoholic and aqueous extract of whole plant of *Wedelia chinensis* was monitored by estimating the serum transaminases (SGOT, SGPT), Serum alkaline phosphatase (SALP), total and direct bilirubin and liver weight of albino rat. The alcoholic extract at a dose level of 500 mg/kg was found to be more potent as compared to aqueous extract. The histopathology of rat liver was also done.

Keywords: Wedelia chinensis, carbontetrachloride induced hepatotoxicity, Histopathology

Introduction

There is a progressive increase in incidence of hepatic damage mainly due to the viral infection, hepatic chemicals (alcohol), and peroxides, toxin in food, pharmaceuticals, environmental pollutants and xenobiotics. There is hardly any remedy available in the modern system of medicine, including corticosteroids and immunosuppressive agents which bring about symptomatic relief supporting only the process of healing or liver regeneration. Hence increasing attention is being given to plant recommended for the treatment of hepatic disorders in the traditional system of medicine. In the context, we have assessed the hepatoprotective activity of alcoholic and aqueous extract of *Wedelia chinensis* against CCl₄ induced hepatotoxicity [1].

Wedelia chinensis (Asteraceae), a perennial herb, is one of the most commonly occurring plants in India. In Hindi, it is known as bhangra and pilabhangra [2]. It has a renowned position in Indian system of medicine and is used as antiinflammatory, anthelmentic, febrifuge and in various hepatic disorders like viral hepatitis [3].

Material and Methods

Plant material: The whole plant of *Wedelia chinensis* was collected from local area of New Delhi and authenticated by Dr. Anjula Pandey, National Herbarium of Cultivated Plant, National Bureau of plant genetic resource, New Delhi and the specimen voucher no. was **NHCP/NBPGR/2008/5/1947**.

Plant extract: The plant material was dried, reduced to moderately coarse powder and then about 200 gm materials were defatted with petroleum ether (60-80°C), alcohol (95%) and water. The extracts were dried under vacuum (yield 12.6%).

Preliminary Phytochemical Studies: The different extracts were then subjected to qualitative phytochemical screening for the identification of the phytoconstituents. Petroleum ether extract showed the presence of steroids, alcoholic extract showed the

presence of glycosides and alkaloids and aqueous extract showed positive test for glycosides and saponins. Alcoholic and aqueous extract of whole plant *Wedelia chinensis* at a dose level of 500mg/kg b.w. [4]. were used for monitoring the hepatoprotective activity.

Animals: Adult albino rats (200-250 gm b.w) were kept in polypropylene cages at an ambient temperature of $25^{\circ}\pm2^{\circ}C$ with 55-65% relative humidity and 12 h light/dark cycle. These animals had free access to water and normal laboratory diet. (Lipton India Limited).

The institutional animal ethics committee (IAEC) approved the use of animals for the present study, (Ethical clearance number: 711/02/a/CPCSEA).

Hepatoprotective activity: The animals were divided in four groups. Group I and II, served as Control and Carbon tetrachloride control, and received the vehicle (water: propylene glycol, 4:1) by gastric intubation once daily for 7 days. Group III and IV served as Treated and Standard and were given 1 ml suspension of alcoholic and aqueous extract of *Wedelia chinensis* at a dose level of 500 mg/kg b.w. and Silymarin at a dose level of 100 mg/kg b.w. [5]once daily for 7 days.

On the 8th day one hr after administration of the last dose of drug, the animals of groups II, III, IV were given an intraperitonial injection of CCl₄ (0.5 ml/kg b.w.). All the animals were then fasted for 24 hrs. After that they were anaesthetized and the blood was collected by cardiac puncture. The blood samples were allowed to coagulate at room temperature for one hour. Serum was separated by centrifugation at 4°C, 12000 rpm for 5 minutes [1].

Biochemical studies: The activity of Serum transaminases (SGOT, SGPT) were estimated by Reitman and Frankel method [6]. Serum bilirubin (total and direct) was determined by Melloy and Evelyn method [7]. Serum alkaline phosphate (SALP) level was also determined [8].

The histopathological study of rat liver was also done.

Statistical analysis: All the data obtained from the above studies were statistically evaluated and the significance of various treatments was calculated using student's t-test. A value of p<0.05 was considered significant as compared with control. [9].

Results

The results obtained from various parameters are summarized in the tables given below.

Table1:	Effect	of	Wedelia	chinensis	on	CCl4	Induced
Hepatoto	oxicity in	n Ra	its				

Parameters	Biochemical Parameters							
Groups				Bilirubin(mg/dl)				
	SGPT(IU/L)	SGOT(IU/L)	SALP(IU/L)	Total	Direct			
Group A	35.00±0.11	39.00±1.21	6.65±0.24	0.40 ± 0.008	0.14 ± 0.01			
Control								
Group B	218.23 ± 3.04^{c}	227.3±3.04 ^c	27.46±4.03 ^c	$8.96 \pm 0.41^{\circ}$	$1.29 \pm 0.01^{\circ}$			
Toxicant								
Group C	36.5±1.73	39.6±0.95	6.74±0.39	0.42 ± 0.02	0.15 ± 0.01			
Standard								
Group D	37.08±1.68	41.20±3.4	7.16±0.14	0.43±0.02	0.16 ± 0.04			
Ethanolic Extract								
(500mg/kg b.w.)								
Group D	38.4±2.10	43.6 ± 0.95^{b}	7.72 ± 0.38^{a}	0.43 ± 0.005^{b}	0.23 ± 0.03^{a}			
Aqueous Extract								
(500mg/kg b.w.)								

Values are expressed as mean ± SEM. (n=6) ^ap<0.05, ^bp<0.02, ^cp<0.001 as compared to control group

Groups	Liver Weight (gm)		
Group A Control	6.33±0.66		
Group B Toxicant	10.23±0.64b		
Group C Standard	7.11±0.14		
Group D Ethanolic Extract (500mg/kg b.w.)	7.92±0.38		
Group E Aqueous Extract (500mg/kg b.w.)	8.98±0.05a		

Table2: Effect of *Wedelia chinensis* on Liver weight in CCl₄ Induced Hepatotoxicity in Rats

Values are expressed as mean \pm SEM. (n=6) ${}^{a}p<0.02$, ${}^{b}p<0.01$ as compared to control group.

Effect of Wedelia chinensis on SGPT levels in CCl4 induced hepatotoxicity in rats





Effect of Wedelia chinensis on SGOT Levels in CCI4 Induced Hepatotoxicity in Rats







Effect of Wedelia chinensis On Total Bilirubin levels in CCl4 Induced Hepatotoxicity in Rats

Effect of Wedelia chinensis On Direct Bilirubin levels in CCl4 Induced Hepatotoxicity in Rats



Histopathological studies



Fig.1: Photomicrograph of liver from group animal control showing normal architecture and no necrosis and no cytoplasmic vacuolation (10 X)



Fig.2: Photomicrograph of liver from animal of toxic group treated with 1.25 ml/kg of CCl4 showing marked vacuolation and portal inflammation (10X)



Fig.3 Photomicrograph of liver from animal treated with CCl4 and silymarin showing diffuse vacuolation with interveining normal areas (10X)



Fig.4: Photomicrograph of liver from animal treated with CCl4 and alcoholic extract of *Wedelia chinensis* showing patchy hepatocyte vacuolation with regenerative activity and area of normal hepatocytes (10X)



Fig.5: Photomicrograph of liver from animal treated with CCl4 and aqueous extract of *Wedelia chinensis* showing patchy hepatocyte vacuolation with degenerative activity (10X)

Discussion

It is well established that CCl_4 induces hepatotoxicity by metabolic activation; therefore it selectively causes toxicity in liver cells maintaining semi-normal metabolic function. CCl_4 is bio-transformed by the cytochrome P450 in the endoplasmic reticulum to produce trichloromethyl free radical (. CCl_3). Trichloromethyl free radical when combined with cellular lipids and proteins in the presence of oxygen form trichloromethyl peroxyl radical, which may attack lipids on the membrane of endoplasmic reticulum faster than trichloromethyl free radical. Thus, trichloromethyl peroxyl free radical leads to elicit lipid peroxidation, the destruction of Ca^{++} homeostatis, and finally, results in cell death [10].

In the present study it was noted that the administration of CCl₄ increased the levels of SGOT, SGPT, and ALP bilirubin (total and direct). A significant reduction was observed in SGPT, SGOT, ALP, total and direct bilirubin levels in the groups treated

with silymarin and both alcoholic and aqueous extract of *Wedelia chinensis*. The enzyme levels were almost restored to the normal.

It was observed that the size of the liver was enlarged in CCl_4 intoxicated rats but it was normal in drug treated groups. A significant reduction in liver weight supports this finding.

Histopathological examination of the liver section of rats treated with toxicant showed necrosis and vacuolization. The rats treated with silymarin and extracts along with toxicant showed sign of protection against these toxicants to considerable extent as evident from the formation of normal hepatic cells and absence of necrosis and vacuoles.

The results were found comparable to Silymarin. Silymarin that is composite name of three flavanoids isolated from milk thistle silybum marinum and are used as hepatoprotectives against experimental hepatotoxicity of various chemicals including CCl₄ [11].

The present study revealed that among the two extracts tested, alcoholic extract at a dose level of 500 mg/kg was found to possess significant protective effect against hepatotoxicity induced by CCl_4 which may be attributed to the individual or combined action of phytoconstituents present in it.

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