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RESEARCH ARTICLE

Hepatoprotective effect of ethanolic extract of *Curcuma caesia* rhizomes in rats on paracetamol induced liver cirrhosis

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ABSTRACT

The objective of the study to evaluate the hepatoprotective effect of ethanolic extract of *Curcuma caesia* rhizomes in Wistar rats on paracetamol induced liver. Albino wistar rats were separated into five groups (n=6). Ethanolic extract of *Curcuma caesia* rhizomes was prepared and evaluated for its hepatoprotective efficacy against paracetamol (PCM) (2g/kg p.o.) induced hepatotoxicity in rats. Silymarin (100mg/kg p.o.) was used as standard. Levels of Serum glutamic pyruvate transaminase (SGPT), Serum glutamic oxaloacetic transaminase (SGOT), alkaline phosphatase (ALP), and total protein was evaluated along with histopathological investigation in various experimental groups of rats. Present study suggested thatthe protective effect of ethanolic extract of *Curcuma caesia* against paracetamol induced hepatic damage.

Keywords: *Curcuma caesia*, Hepatoprotective effect, paracetamol, Histopathological study, cirrhosis

Introduction

Cirrhosis is a problem of liver disease which is associated with loss of liver cells and unchangeable scarring of the liver[1]. Hepatic fibrosis is extremely exuberant wound healing in which excessive connective tissue builds up in the liver[2]. This inflammatory response is accompanied by limited deposition of extra cellular matrix (ECM), so that if the regeneration of dying cells fails during persistent liver injury, hepatocytes are replaced by abundant ECM, including fibrillar collagen, depending on the origin of injury[3].

Hepatotoxicity resulting from liver damage which plays a pivotal role in amendable various physiological processes in the body, such as metabolism, secretion and storage. It has great capacity to detoxify toxic substances and synthesize useful principles[4]. Paracetamol promote development of free radicals and subsequent lipid peroxidation damages the membranes of liver cells and organelles andcausing the swelling and necrosis of hepatocytes. Paracetamol is widely used an antipyretic and analgesic, but higher doses produced acute liver damage[5]. Various newly advanced drugs have been used for treatment of liver diseases; however, these drugs produced destructive side effects. For that purpose, further research on herbs that could potentially substitute the chemical-based drugs is very crucial as several medicinal herbs have been shows hepatoprotective properties and offers a comprehensive coverage for the treatment of virtually every manifestation of liver dysfunction[6]. Therefore, this study has been conducted to evaluate the hepatoprotective effect of ethanolic extract of *Curcuma caesia* rhizomes in rats on paracetamol induced liver cirrhosis.

Materials and Methods

Drugs & Chemicals

Paracetamol (Sigma chemicals, USA) and silymarin (Micro labs, India) was used in present study. All other chemicals and other biochemical used in the experiments were of analytical grade from different firms.

Collection of plant material

The plant was purchased from local market of Bhopal. The plants were collected in the month of September to November from local market of Bhopal, India.

Acute toxicity studies

Acute oral toxicity was conducted according to the method of Organisation for Economic Co-operation and Development (OECD)[7-9]. Animals were kept fasting providing only water, ethanolic extract of whole plant *Curcuma caesia* (ECC) (50, 100, 150, 200, 300 mg/kg/day) was administered orally for 4 days of six groups of rats (n=6) and the animals were kept under observation for

mortality as well as any behavioural changes for evaluation of a possible hepatoprotective effect.

Experimental designs

Group-I: Normal control (0.5% CMC 1 ml/kg, p.o.)

Group-II: Paracetamol (PCM, 2 g/kg p.o.)

Group-III: *Curcuma caesia* (100 mg/kg, p.o.) + PCM (2 g/kg, p.o.)

Group-IV: *Curcuma caesia* (200 mg/kg, p.o.) + PCM (2 g/kg, p.o.)

Group-V: Silymarin (100 mg/kg, p.o.) + PCM (2 g/kg, p.o.)

Animals were divided into five groups of 6 animals each. The first group received 0.5% CMC 1 ml/kg for one week (control). The group II received 0.5% CMC 1 ml/kg for one week (positive control). The groups III, IV and V received 100 mg/kg and 200 mg/kg of ethanolic extract of whole plant C. caesia and silymarin (100 mg/kg p.o.) respectively once a day for seven days. On the fifth day, after the administration of the respective treatments, all the animals of groups II, III, IV and V were administered with PCM 2 g/kg orally. After 7 days animals were anaesthetized with ether for collection of blood from retro orbital plexus, and then sacrificed under ether anaesthesia for the removal of liver. Various biochemical analyses were carried out[10].

Biochemical Evaluation in Serum

Serum glutamic pyruvate transaminase (SGPT), Serum glutamic oxaloacetic transaminase (SGOT), Alkaline phosphatase (ALP), Total protein concentration and total bilirubin was estimated by using commercial kits as per the manufacturer instructions[11].

Histopathology of liver

Liver tissues of rat was removed and washed with normal saline. The cleared tissue was fixed in 10% natural buffered formalin solution (pH 7.0-7.2). After proper fixation tissue was processed for dehydration in ascending grade of ethanol, clearing with toluene, followed by impregnation in paraffin wax, then sections of 5 ì in thickness was cut with help of semi-automatic rotary microtome. Sections were stained with haematoxylin. All the sections of the tissues were examined under microscope for the analyzing the altered architecture due to the liver tissue due to PCM challenge and improved liver architecture due to pre-treatment with test extracts and standard drug[12]. These were examined under the microscope for histopathological changes such as congestion, hemorrhage, necrosis, inflammation, Infiltration, kuffer cells and sinusoids and photographs were taken.

Results and Discussions

Histopathological changes of liver are given in Fig.1. Histology of the liver sections of normal control animals showed normal liver architecture with well brought out central vein well-preserved

cytoplasm and prominent nucleus and nucleolus (Fig. 3A). The liver samples of control animals showed feathery degeneration, micro and macro cellular fatty changes, and inflammatory cells around portal tract (Fig. 3B). The control + standard (Silymarin) treated animals also showed a good protection from inducer changes in the liver (Fig. 3C). The histopathological examination clearly revealed that the hepatic cells, central vein, and portal triad were almost normal in 100 mg/kg and 200 mg/kg dose of ECC administered groups in contrast to group which received inducer but with mild Kupffer cell hyperplasia (Fig. 3D& 3E).

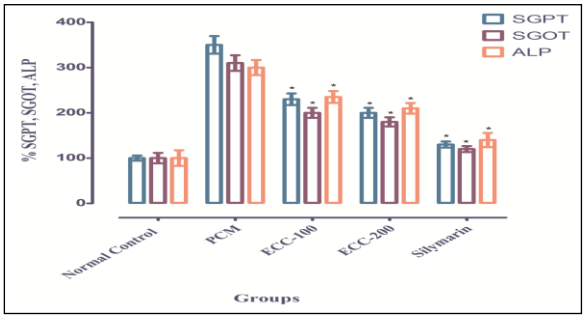
Conclusion

Hepatoprotective activity of the ethanolic extract (100 & 200mg/kg) of Curcuma caesia was studied. Inthis study, the drug treated animals showed decreased serum enzymes like SGPT, SGOT. This confirms the protective effect of ethanolic extract of Curcuma caesia against paracetamol induced hepatic damage.

Table 1: Effect of pre-treatment of ECC and Silymarin on %SGPT, SGOT and ALP levels in paracetamol induced hepatotoxicity in rats.

Treatment	Dose	SGPT (%)	SGOT (%)	ALP (%)
Normal Control	0.5% CMC 1 ml/kg	100.0 ± 5.74	100.0 ± 11.55	100.0 ± 17.32
Paracetamol (PCM)	2 g/kg	350.0 ± 19.44	310.0 ± 17.22	300.0 ± 16.66
Curcuma caesia (ECC)	100 mg/kg	230.0 ± 12.77***	200.0 ± 11.11***	235.0 ± 13.05***
Curcuma caesia (ECC)	200 mg/kg	200.0 ±11.10***	180.0 ±10.00***	210.0 ± 11.66***
Silymarin	100 mg/kg	130.0 ± 6.66***	120.0 ± 6.67***	140.0 ± 15.55***

Values are expressed as the mean ± SEM of six observations. ***P<0.001 vs. control treatment (One-way ANOVA followed by Dunnett's test)



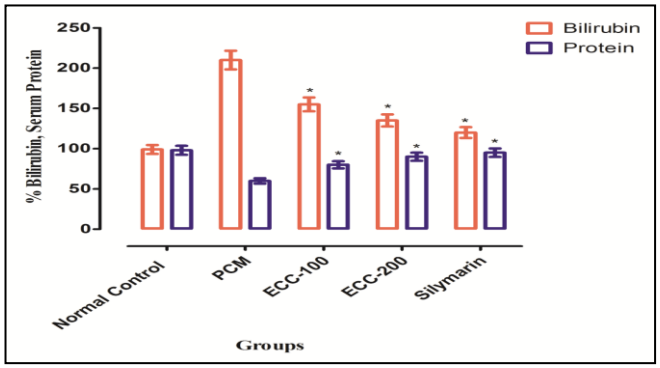
Values are expressed as the mean ± SEM of six observations. *P<0.001 vs. control treatment (One-way ANOVA followed by Dunnett's test)

Fig. 1 Effect of pretreatment of ECC and Silymarin on %SGPT, SGOT and ALP levels in paracetamol induced hepatotoxicity in rats.

Table 2: Effect of pretreatment of ECC and Silymarin on % serum bilirubin and protein levels in paracetamol induced hepatotoxicity in rats.

Treatment	Dose	Serum Bilirubin (%)	Serum Protein(%)
Normal Control	0.5% CMC 1 ml/kg	99.0 ± 5.50	98.0 ± 5.54
Paracetamol (PCM)	2 g/kg	210.0 ± 11.60	60.0 ± 3.33
Curcuma caesia (ECC)	100 mg/kg	155.0 ± 8.61***	80.0 ± 4.44***
Curcuma caesia (ECC)	200 mg/kg	135.0 ± 7.50***	90.0 ± 5.00***
Silymarin	100 mg/kg	120.0 ± 6.67***	95.0 ± 5.27***

Values are expressed as the mean ± SEM of six observations. ***P<0.001 vs. control treatment (One-way ANOVA followed by Dunnett's test)



Values are expressed as the mean ± SEM of six observations. *P<0.001 vs. control treatment (One-way ANOVA followed by Dunnett's test)

Fig. 2 Effect of pretreatment of ECC and Silymarin on % serum bilirubin and protein levels in paracetamol induced hepatotoxicity in rats.

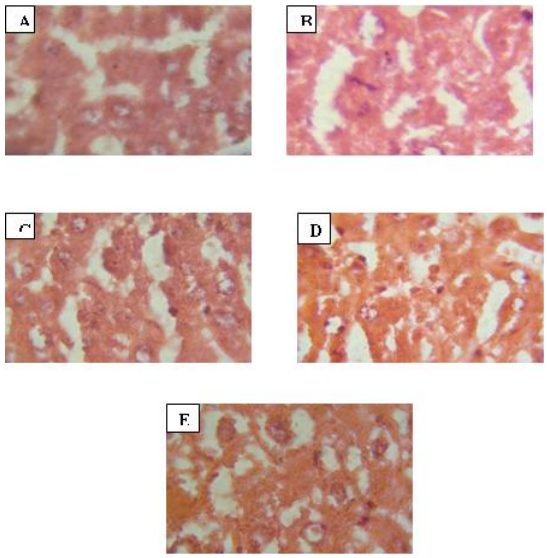


Fig.3 Light microscopic analysis of rat liver sections of normal rats and treated with drug administration. (A) normal; (B) PCM; (C) Silymarin; (D) ECC-100; (E) ECC-200

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