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Hepatoprotective Activity of *Momordica diocia Roxb* Fruits in CCl₄-Induced Hepatotoxicity in Rats

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Abstract

Fruits, leaves and tuberous roots of *Momordicia diocia* are used in India as a folk remedy for treatment of a wide range of disorders. The objective of this study was to evaluate the hepatprotective activity of the fruits of *Momordicia diocia* by preparing different extracts and the resultant extract were screened for the hepatprotective activity. The ethyl acetate and ethanolic extracts of *Momordicia diocia* fruits were prepared and subjected for phytochemical screening and tested for their hepatprotective activity in CCl4-induced hepatotoxicity in rats. Phytochemical screening showed positive test for steroids, triterpenoids and glycosides (Etoh extract). The ethyl acetate and ethanolic extracts showed a significant hepatprotective activity at a dose of 200 mg/kg (p.o.). The results suggested that *Momordica diocia diocia* fruits possess potential hepatprotective activity against CCl4 hepatotoxicity possibly by antioxidant activity.

Keywords: Hepatprotective activity; *Momordicia diocia* fruits; *Received:* October 10, 2010; *Accepted:* March 15, 2011.

1. Introduction

Momordica diocia Roxb.Ex. Wild. (*Cucurbitaceae*) is a perennial dioiceous climber with tuberous roots found throughout India from Himalayas to Ceylon, up to an attitude of 1,500 m. The plant is sometimes found growing wild and is common in hedges. It is often cultivated for its fruits, which are used as vegetables [1]. The whole plant is used for the treatment of eye diseases, poisoning and fever [2]. Juice of the root is stimulant, astringent and antiseptic, and tubers are used in cases of bleeding piles and similar infections. Plant is claimed to be expectorant, analgesic and soothing agent. Root is also used to stop bleeding from piles, as an expectorant and also in urinary and bowel complaints [3]. Fruits, leaves and tuberous roots used in India as a folk remedy for diabetes [4]. The aqueous extract of root has spermicidal activity and anthelemintic activity [5]. The roots are reported to possess moderate antimicrobial activity and poor antifungal activity [4] and postcoital antifertility activity [6].

Phytochemical investigation has revealed the presence of traces of alkaloids and ascorbic acid in fruits. Lectins, β -sitosterol, saponinglycosides, triterpens of ursolic acid, hederagenin, oleanolic acid, α -spiranosterol,

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steraic acid, gypsogenin, momodicaursenol, two novel aliphatic constituents [4, 7-9].

To the best of our knowledge no report is available on the hepatprotective activity of *M*. *diocia* fruits. Hence the present study was undertaken to verify and evaluate the hepatprotective activity of fruits of *M*. *diocia* by preparing various extracts and the resultant extracts was screened for the hepatprotective activity.

2. Materials and methods

2.1. Plant material

Fresh fruits of *M. dioica* were collected during July-August, 2007 and identified and authenticated by Dr Gajendra Roa, Department of botany, Regional Research Institute, Jayanagar, Bangalore. An authenticated specimen was deposited in the herbarium with voucher no: 43525 for further reference.

2.2. Extraction and isolation

Fresh fruits of M. dioica (5 kg) were crushed and extracted successively with ethyl acetate and ethanol in a soxhlet extractor and yield was 0.42, and 0.58%, respectively, on dried weight basis.

2.3. Phytochemical screening of various extracts

Freshly prepared extracts were tested for the presence of alkaloids, steroid and triterpenoids and their glycosides, tannins, flavonoids and their glycosides, carbohydrates, and cardiac glycosides using standard procedure [9].

2.4. Test animals

Male Wistar rats weighing 190-210 g were used in the experiment. They were maintained in standard environmental conditions of temperature (25 ± 2 °C) relative humidity ($55\pm10\%$) and 12 h dark/light cycle. They were fed with standard diet (Hindustan Lever, India) and water *ad libitum*. The animals were divided into five groups of six rats each.

Group 1: served as control and received the vehicle (1 ml/kg/day of 1% (w/v) gum tragacanth p.o. for 14 days). Group II and V received 0.1 ml/kg/day of CCl_4 i.p. for 14 days; Group III animals received the standard drug silymarin [8] (Ranbaxy lab, dewas) in the dose of 100 mg/kg/day, p.o. for 14 days, while the ethyl acetate and ethanolic extracts of *M. diocia* were administered to Groups IV and V in the dose of 200 mg/kg/day, p.o., respectively, for 14 days.

The CCl_4 and silymarin or the extracts were administered concomitantly to the respective groups.

All of the animals were sacrificed on the 14th day under light ether anesthesia. The blood samples for each animal were collected separately in sterilized dry centrifuge tubes by carotid bleeding and allowed to coagulate for 30 min at 37 °C.

The clear serum was separated at 2500 rpm for 10 min. and subjected to biochemical investigation Viz, total bilirubin [10], total protein [11], serum alanine transaminase, asparatate transaminase [12], and alkaline phosphatase [13].

Results of Biochemical estimations are reported as Mean±SEM of six animal in each group. The data were subjected to One-way ANOVA followed by Tukey's multiple comparisons test, and p<0.001 was considered statistically significant.

3. Results

Phytochemical screening gave positive tests for steroids, triterpenoids, and glycosides (Eth Ac extract) and steroids, triterpenoids and their glycosides (Ethanolic extract).

The effect of ethyl acetate and ethanolic extract of *M. diocia* in CCl_4 induced liver damage in rats with reference to biochemical changes in serum is shown in Table 1. The CCl_4 treated control groups showed significant

Group (N)	Total bilirubin	Total protein	AST (IU/L)	ALT (IU/L)	ALP (IU/L)
	(mg/dl)	(gm%)			
Control (1% w/v gum	0.44±0.01	9.44±0.02	153.52±1.43	54.08±1.16	174.99±1.80
trangacanth, p.o.) CCl4 (0.1 ml/kg/day, i.p.) "+Silymarin(100 mg	2.45±0.01*	5.93±0.01	2213.50±32.79*	1413.00±1.99*	444.33±1.56*
/kg/day, p.o.) "+Ethyl acetate extract (200 mg/ kg/day, p.o.)	0.54±0.01† 0.93±0.02 ^{†@}	8.82±0.01† 7.40±0.02 ^{†@}	208.50±2.17 † 242.17±0.02 ^{†@}	75.18±1.17† 193.36±149 ^{†@}	184.40±1.16† 244.30±1.91 ^{†@}
"+Ethanolic extract (200 mg/kg/day, p.o.)	$0.60{\pm}0.01^{\dagger@}$	8.41±0.02 ^{†@}	221.67±2.59 ^{†@}	125.06±1.27 ^{†@}	204.43±1.64 ^{†@}

Tabel 1. Effect of ethyl acetate and ethanolic extract	ct of fruits of M. diocia in	CCl4 induced hepatotoxicity in rats.
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Values are expressed as Mean±SEM. n=6 in each group, $p \le 0.01$ compared to the control group. $p \le 0.01$ compared to the CCl₄-treated group. $p \le 0.01$ compared to the CCl₄ + silymarin treated group.

increase in serum total bilirubin, alanine transaminase, asparatate transaminase and alkaline phosphatase and a decrease in total protein indicating liver injury caused by CCl_4 , where as animals treated with ethyl acetate and ethanolic extract exhibited a decrease in total bilirubin, alanine transaminase, asparatate transaminase and alkaline phosphatase along with a significant increase in total protein.

4. Discussion

The present investigation indicated that both the extracts of M. dioica fruits provide significant protection against CCl_4 induced hepatotoxicity in rats. CCl_4 is widely used as a hepatotoxin in experimental studies. CCl_4 is biotransformed by the Cytochrome P450 system to produce the trochloromethyl free radical, which in turn covalently binds to cell membranes and organelles to elicit lipid peroxidation [14]. Further, it has been found that several phytoconstituents have the ability to induce microsomal enzymes either by accelerating the excretion of CCl_4 or by inhibition of lipid peroxidation induced by CCl_4 [15].

Phytoconstituents like flavonoids, triterpenoids, saponins, and alkaloids are known to posse's hepatprotective activity, and phytochemical investigations of ethyl acetate and ethanolic extract of fruits of M. dioica revealed the presence of alkaloids, saponins, glycosides, triterpenoids and tannins [16-20].

The present study revealed that among the two extract tested, ethanolic extract of fruits of M. diocia found to posses a significant protective against hepatotoxicity induced by CCl_4 which may be attributed to the individual or combined action of phytochemical present in it. However, further investigation is needed for identification of the active compound(s) responsible for hepatprotective activity.

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