Available online at www.sciencedirect.com
SCIENCE DIRECT®
Journal of Ethnopharmacology 97 (2005) 521–525
Journal of ETHNO-PHARMACOLOGY
www.elsevier.com/locate/jethpharm

Free radical scavenging and hepatoprotective activity of jigrine against galactosamine induced hepatopathy in rats

Abul K. Najmi [a,*], K.K. Pillai [a], S.N. Pal [a], M. Aqil [b]

[a] *Department of Pharmacology, Faculty of Pharmacy, Jamia Hamdard (Hamdard University), New Delhi 110 062, India*
[b] *Department of Pharmaceutics, Faculty of Pharmacy, Jamia Hamdard (Hamdard University), New Delhi 110 062, India*

Received 17 February 2004; received in revised form 3 November 2004; accepted 16 December 2004

Abstract

Jigrine a polypharmaceutical herbal hepatoprotective formulation containing aqueous extracts of 14 medicinal plants is used in Indian system of medicine (Unani). Jigrine was evaluated for its hepatoprotective activity against galactosamine induced hepatopathy in rats. Galactosamine induced hepatotoxicity resembles human viral hepatitis. Biochemical parameters like AST, ALT and urea in serum, TBARS and glutathione in liver and whole blood glutathione were estimated to assess liver function. DPPH-free radical scavenging activity of jigrine was also evaluated. Biochemical data exhibited significant hepatoprotective activity of jigrine against galactosamine. Silymarin used as reference standard also exhibited significant hepatoprotective activity against galactosamine. The biochemical observations were supplemented with histopathological examination of rat liver sections.

Keywords: Jigrine; Unani medicine; Galactosamine; Hepatopathy; Silymarin; DPPH

1. Introduction

Jigrine is a polypharmaceutical herbal formulation containing aqueous extracts of 14 medicinal plants used for liver ailments (Table 1). Few studies are reported for its formulation (Najmi et al., 2002), safety evaluation (Valecha et al., 1990), mechanism of hepatoprotective action (Vivek et al., 1994; Karunakar et al., 1997a; Aftab et al., 1999) and anti-inflammatory activity (Karunakar et al., 1997b). Hepatoprotective and anti-inflammatory effects of some of the individual ingredients of jigrine are also reported in literature (Jindal et al., 1975; Sadique et al., 1987; Chawla et al., 1992; Gilani and Aftab, 1992; Gilani et al., 1993; Reddy et al., 1993; Gilani and Janbaz, 1994; Pandey et al., 1994; Sultana et al., 1995; Zafar and Ali, 1998).

The present study is designed to investigate the DPPH-free radical scavenging activity, hepatoprotective and antioxidant activity of jigrine against galactosamine induced hepatotoxicity in rats.

2. Material and methods

2.1. Drugs and chemicals

Jigrine was provided by Hamdard (Wakf) Labs., Ghaziabad, India. Jigrine contains aqueous extract of 14 medicinal plants (Table 1). These constituent plants were collected and authenticated by taxonomist at the Hamdard (Wakf) Labs. The extract was prepared from the freshly collected plant materials. Silymarin was purchased from Micro Labs., Holar, TN, India. Galactosamine was procured from SRL, India. All the biochemicals and chemicals used were of analytical grade.

* Corresponding author. Fax: +91 11 2605 9663.
E-mail addresses: aknajmi@hotmail.com, aknajmi@jamiahamdard.ac.in (A.K. Najmi).

0378-8741/$ – see front matter © 2005 Elsevier Ireland Ltd. All rights reserved.
doi:10.1016/j.jep.2004.12.016

Table 1
Medicinal plant ingredients of jigrine (a unani polypharmaceutical herbal formulation)

Botanical name	Common name	Unani name	Family	Part used
Cichorium intybus Linn	Chicory	Tukhme, kasni	Compositeae	Leave
Tamarix dioica Roxb	Tamarisk	Jhau	Tamaricaceae	Leave
Solanum nigrum Linn	Black nightshade	Makoh	Solanaceae	Fruit
Rheum emodi wall	Indian rhubarb	Revand chini	Polygonaceae	Rhizome
Rubia cordifolia Linn	Indian madder	Majeeth	Rubiaceae	Root
Vitex negundo Linn	Nisinda	Sambhalu	Verbenaceae	Whole shrub
Cassia occidentalis Linn	Coffee senna	Kasaundi	Caesalpiniaceae	Leave
Foeniculum vulgare Mill	Fennel	Sonf	Umbellifereae	Fruit
Cuscuta reflexa Roxb	Amarvella	Tukhme, kasoos	Convolvulaceae	Seed
Careya arborea Roxb	Wild guava	Baokhamba	Barringtoniaceae	Fruit
Phyllanthus niruri Linn & Hook	Jaramla	Bhui amla	Euphorbiaceae	Leave
Plantago major Linn	Isphagol	Bartang	Plantaginaceae	Leave
Rosa damascena Linn	Damask rose	Gul-e-surkh	Rosaceae	Flower
Solanum xanthocarpum Schrad & wendl.	Yellow berries, nightshade	Katheli	Solanaceae	Root, fruit

2.2. Animals

Wistar strain albino rats (weighing 150–200 g) were used for the study. Animals were supplied by Central Animal House Facility of Hamdard University and kept under standard laboratory conditions in 12 h light–dark cycle at 25 ± 2 °C. Animals were provided with pellet diet (Lipton, India) and water ad libitum.

2.3. Experimental protocol

Rats were randomly divided into four groups of six animals each. Group I served as normal control and received normal saline for 21 days. Group II served as toxic control and received normal saline (1 ml/kg, p.o.) for 21 days. Groups III and IV were prophylactically treated with jigrine (1 ml/kg, p.o.) and silymarin (25 mg/kg, p.o.) for 21 days, respectively. Groups II, III and IV also received galactosamine (400 mg/kg, i.p.) on 21st day (Mitra et al., 2000). After 24 h of galactosamine administration blood was collected from tail vein under light ether anaesthesia. Immediately, after blood withdrawal all the groups were sacrificed. Liver samples were also collected for histological and biochemical estimations. The blood samples were allowed to clot for 30–40 min. Serum was separated by centrifugation at 37 °C and was used for estimation of various biochemical parameters. Liver samples collected were washed with chilled normal saline, weighed and 10% (w/v) liver homogenates were made in ice cold 0.15 M KCl solution using motor driven Teflon pestle. All the procedures carried out on animals were approved by institutional animal ethics committee (JHAEC).

2.4. Assessment of liver function

2.4.1. Assay of serum transaminases and urea

The activities of serum aspartate transaminase (AST), alanine transaminase (ALT) (Reitman and Frankel, 1957) and serum urea concentration (Teitz, 1976) were estimated by the reported procedures.

2.4.2. Determination of reduced glutathione

Blood and liver glutathione were estimated by 5,5-dithiobis-2-nitrobenzoic acid (DTNB) according to the method of Ellman (1959) and expressed as mg% and μmol/g of protein, respectively.

2.4.3. Liver TBARS and protein estimation

TBARS was used as an index of lipid peroxidation and measured by the modified method of Ohkawa et al. (1979). Total protein in the tissue homogenate was also estimated (Lowry et al., 1951). The levels of TBARS were expressed as nmol MDA/mg protein.

2.5. Histological studies

Livers were quickly removed and preserved in neutral buffered formalin. Histological liver sections were prepared as described previously by Luna (1968).

2.6. DPPH-free radical scavenging activity

DPPH-free radical scavenging activity was measured according to the previously described procedure with slight modification (Braca et al., 2002). Briefly, different methanolic dilutions of jigrine were mixed with equal volumes of 0.004% MeOH solution of DPPH. The resulting solutions were thoroughly mixed and absorbance was measured at 520 nm after keeping the tubes in dark for 20 min. The scavenging activity was determined by comparing the absorbance with that of control containing equal volumes of DPPH solution and methanol.

2.7. Data analysis

Results are expressed as mean ± S.E.M. Total variation present in a set of data was estimated by one-way analysis of variance (ANOVA) followed by Dunnet's post-hoc test. $P < 0.05$ was considered significant.