Journal of Saudi Chemical Society (2013) **17**, 125–133

King Saud University

Journal of Saudi Chemical Society

www.ksu.edu.sa
www.sciencedirect.com

ORIGINAL ARTICLE

Chemical constituents and protective effect of *Ficus ingens* (Miq.) Miq. on carbon tetrachloride-induced acute liver damage in male Wistar albino rats

Abd El Raheim M. Donia [a,b,*], Gamal A. Soliman [c], Ahmed M. Zaghloul [a,d], Saleh I. Alqasoumi [e], Amani S. Awaad [f], Asmaa M. Radwan [g], Omer A. Basodan [e]

[a] *Pharmacognosy Department, College of Pharmacy, Salman Bin Abdulaziz University, Al-Kharj, Saudi Arabia*
[b] *Medicinal and Aromatic Plants Department, Desert Research Center, Cairo, Egypt*
[c] *Pharmacology Department, College of Pharmacy, Salman Bin Abdulaziz University, Al-Kharj, Saudi Arabia*
[d] *Pharmacognosy Department, College of Pharmacy, Mansoura University, Egypt*
[e] *Pharmacognosy Department, College of Pharmacy, King Saud University, Saudi Arabia*
[f] *Chemistry Department, College of Science, King Saud University, Saudi Arabia*
[g] *Botany and Microbiology Department, College of Science, Girls Branch, Al Azhar University, Cairo, Egypt*

Received 30 April 2012; accepted 29 May 2012
Available online 7 June 2012

KEYWORDS

Phytochemical studies;
Flavonoids;
Antharquinone;
Hepatoprotective effect

Abstract The aim of the present study was to investigate the chemical constituents and hepatoprotective effect of *Ficus ingens* (Miq.) Miq. (Moraceae) extract against carbon tetrachloride-induced acute liver damage in male Wistar albino rats. The ethanol extract of *F. ingens*, was subjected to phytochemical study. In addition, its acute and sub-chronic toxicities were assessed. Eight compounds were isolated from this plant and identified as β-sitosterol, β-sitosterol glucoside, chrysophanol, 7-hydroxy-2,5 dimethyl chromen-4-one, quercetin, Aloe emodin glucoside, rutin and Patuletin-3′-*O*-methyl-3-*O*-rutinoside. The structure elucidation was based on ^{1}H and ^{13}C NMR, proton–proton correlation spectroscopy (^{1}H–^{1}H Cosy), distortionless enhancement by polarization transfer (DEPT), Heteronuclear Multiple-Quantum Correlation (HMQC), and heteronuclear

* Corresponding author at: Pharmacognosy Department, College of Pharmacy, Salman Bin Abdulaziz University, Al-Kharj, Saudi Arabia. Tel.: +966 560019012.
E-mail address: donia22276@yahoo.com (A.M. Donia).

Peer review under responsibility of King Saud University.
http://dx.doi.org/10.1016/j.jscs.2012.05.008

Production and hosting by Elsevier

multiple bond correlations spectrum (HMBC). Hepatotoxicity induced with CCl_4 was evidenced by elevation of liver marker enzymes (ALT, AST, ALP and LDH) and TB content in serum. In addition, antioxidant enzymes were drastically inhibited with significant reduction of GSH and increased LPO in liver homogenate of CCl_4-intoxicated rats. Pre-treatment with *F. ingens* (200 and 400 mg/kg) and silymarin (50 mg/kg) avoided the changes observed in CCl_4-intoxicated rats. In conclusion, the ethanol extract of *F. ingens* showed protective activity against liver injury, which might be developed into a new hepatoprotective agent.

1. Introduction

Liver is a vital organ that has a wide range of functions, including detoxification, plasma protein synthesis, and production of biochemicals necessary for digestion. Damage to the liver inflicted by hepatotoxic agents is of grave consequence. Today, liver damage is one of very common aliment in the world resulting in serious debilities ranging from severe metabolic disorders to even mortality (Akilavalli et al., 2011). Various xenobiotics are known to cause hepatotoxicity; one among them is CCl_4 that may cause lipid peroxidation (Kodavanti et al., 1989; Demirdag et al., 2004). Many hepatoprotective herbal preparations have been recommended in alternative medicine for the treatment of liver diseases. Therefore, the search of a new natural hepatoprotective agent is of great interest.

Ficus is a genus belonging to family Moraceae, it comprises about 850 species of woody trees, shrubs, vines, epiphytes, and hemiepiphyte. *F. ingens* is an evergreen deciduous tree up to 10 m height, occasionally higher, with a rounded or spreading crown and with a spread of up to 30 m wide. The plant grows in various habitats including Saudi Arabia. All the parts have milky latex when broken. Fruits are found on the tree usually throughout the year but peaking in summer (Myburgh et al., 1994).

Many active compounds were isolated from *Ficus benghalensis* bark; 20-tetratriaconthene-2-one, 6 heptatriacontene-10-one, pentatriacontan-5-one, β-sitosterol, β-D-glucoside and meso inositol (Mousa et al., 1994). In addition, the fruit extract of *F. benghalensis* exhibited antitumor activity (Joy et al., 2001), while the methanol extract of *F. benghalensis* possesses antioxidant activity (Yadav et al., 2011). *Ficus sycomorus* extracts are used in Folk medicine in the treatment of infertility and sterility in humans (Malgras, 1992; Pakia et al., 2003; Kone and Atindehou, 2008). *Ficus capensis* extract was used for treatment of azoospermia (Gelfand et al., 1985). *Ficus asperifolia* extract has been reported to have an estrogenic effect in female rats (Watcho et al., 2009).

2. Materials and methods

2.1. Plant material

The aerial parts of *Ficus ingens* (Miq.) Miq. were freshly collected from Tabouk area-KSA, during Summer 2010. The collected plant was kindly authenticated by Prof. Dr. Abd El Naser El-Gifri, Prof. of Taxonomy, Salman bin AbdulAziz University, KSA. A voucher specimen has been deposited in the herbarium of the Pharmacognosy department, Salman bin AbdulAziz University. The plant was dried under shade and then ground to fine powder.

2.2. Extraction

One kg of the dried powder was extracted by percolation in 70% aqueous ethanol for 72 h. The combined ethanol extracts were concentrated under reduced pressure at a temperature not exceeding 45 °C. The extract was fractionated using silica gel column chromatography (350 g) and gradiently eluted with chloroform containing increasing proportions of methanol. Fractions (85, 100 mL each) were collected and monitored by TLC (silica gel, chloroform–methanol). Similar fractions were combined together to obtain 5 groups. Each group was reapplied to silica gel column eluted with chloroform containing gradually increasing proportions of methanol. Further purification was carried out using Sephadex LH-20 columns to afford compounds **1**–**8**.

2.3. Acid hydrolysis

Two mg of compounds 2, 6, 7 and 8 was dissolved in 2 mL of methanol: water (1:1, v/v), mixed with 1 mL of 2 N HCl, and refluxed at 60 °C for 3 h. The aglycone moiety was subsequently extracted with ethyl acetate. The aqueous phase was neutralized with silver oxide then filtered. The filtrate was used for identification of the sugar moiety (Stahl, 1969).

2.4. Apparatus

Proton (^1H) and carbon 13 (^{13}C NMR) spectra were recorded on Bruker VX500 NMR spectrometer operating at 500 and 125 MHz respectively. ^1H–13C correlations were established by using HMQC and HMBC pulse sequences respectively. ^1H–1H correlations were determined by double quantum filtered COSY.

2.5. Experimental animals

Male Wistar albino rats (160–180 g) and albino mice of both sexes (27–30 g) were maintained in the Laboratory Animal Unit of the College of Pharmacy, Salman Bin Abdulaziz University. They were housed in polypropylene cages and fed with standard chow diet and water *ad libitum*. The animals were exposed to alternate cycle of 12 h of darkness and light. Male rats were used because of their constant metabolism compared to the variation in the female physiology. Animals were allowed to adapt to the laboratory environment for one week before experimentation. The care and handling of the animals were in accordance with the internationally accepted standard guidelines and were approved by an institutional review board.