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Hepatoprotective effect of leaves of aqueous ethanol extract of *Cestrum nocturnum* against paracetamol-induced hepatotoxicity

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Abstract

The hepatoprotective activities of *Cestrum nocturnum* (Queen of Night) was evaluated against the paracetamol induced hepatotoxicity in the mice. Aqueous ethanol (30:70) extract of plant was obtained by maceration. Results showed that aqueous ethanol extract of *C. nocturnum* (250 and 500 mg/kg) produced significant ($p<0.05$) hepatoprotective activities against paracetamol induced liver injury in *Swiss* albino mice. Histopathalogical studied of liver further supported the hepatoprotective effects of *C. notrunum.* Phytochemical screening showed the presence of alkaloids, flavonoids, saponins, terpenes, phenolic compounds, carbohydrates and volatile oils. Most of the flavonoids have hepatoprotective activity. Therefore, the hepatoprotective activity of *C. nocturnum* may be due to the presence of flavonoids and phenolic components. It was concluded from the present study that aqueous ethanol extract of leaves of *C. nocturnum* has hepatoprotective activity against the paracetamol induced hepatotoxicity in albino mice.

Introduction

Liver is most vital organ of the body. It play important role in the metabolisms of carbohydrate, protein and fat and detoxification of toxic chemicals and xenobiotics, secretion of bile and storage of many vitamins. A healthy liver is a sign of good health (Subramonium et al., 1999). Certain infections, drugs, hepatotoxins, environmental factors, viruses, bacteria and alcohol use cause liver injury. Hepatic injury is caused by the production of reactive oxygen free radical during biochemical reactions (Ali et al., 2004). Scavenging of free radicals by anti-oxidants reduced the process of liver cells fibrosis (Raja et al., 2006). Many plants have been identified as hepatoprotective like *Trianthema decandra* (Balamurugan and Muthusamy, 2008), *Cocculus hirsutus* (Thakare et al., 2009), *Carica papaya* (Sadeque and Begum, 2010), *Carissa spinarum* (Hegde and Joshi, 2010), *Convolvulus arvensis* (Ali et al., 2013), *Dodonaea viscosa* (Khan et al., 2013), *Trichodesma sedgwickianum* (Saboo et al., 2013), *Ofpomoea staphylina* (Bag and Mumtaz, 2013) and Khamira Gaozaban Ambri Jadwar Ood Saleeb Wala (Akhtar et al., 2013).

Cestrum nocturnum (Family Solanaceae) commonly known as "Queen of Night" is used as anti-inflammatory, analgesic (Mazumder et al., 2010), antimicrobial (Khan et al., 2011), antiepileptic (Perez et al., 2008), anticancer (Zhong et al., 2008), insecticidal (Savchenko et al., 2000), local anesthetic (Zeng et al., 2002), and CNS depressant (Zeng et al., 2003). Important phytoconstituents e.g. flavonoids, alkaloids and phenols have been reported in this plant (Prasad et al., 2013). Most of the flavonoids have hepatoprotective activity (Ali et al., 2013). The plant has been proved to be anti-oxidant (Al-Reza et al., 2010). Phytochemical profile and anti-

oxidant activity of the plant appealed us to scientifically evaluate its hepatoprotective potential.

Materials and Methods

The plant material

The leaves of *C. nocturnum* were collected from the Lahore (Pakistan) in the month of March 2013. The plant was authenticated by the Dr. Mansoor Hameed, Department of Botany, University of Agriculture, Faisalabad.

The leaves were cleaned with tap water and dried under shade for one week. They were grinded into coarse powder by a mechanical grinder. The coarse powder (1,000 g) was then subject to maceration with 500 mL aqueous ethanol (70:30) for seven days in air tight vessels with occasional shaking at room temperature. The macerates were then filtered through Whatman filter paper No. 1 and filtrates were concentrated in the rotary evaporator at 35°C and at high pressure. The concentrated extract were then stored at 4°C in dark ambered colored glass containers.

Experimental protocols

Swiss albino mice were used for the study, of either sex weighing between 25-30 g. They were fed on standard diet and water *ad libitum.* The study protocol was approved by the Ethical Review Committee, GC University Faisalabad.

The mice were divided into five groups. Each group contained five mice (Sabir and Rocha, 2008). One group received distill water (5 mL/kg, p.o) for 9 days once daily and served as normal control. Another group received water (5 mL/kg, p.o) for 9 days once daily and paracetamol (250 mg/kg, p.o) after three hours daily. Third group was administered standard drug silymarin (50 mg/kg, p.o) for 9 days once daily and paracetamol (250 mg/kg, p.o) after three hours daily. Fourth group of mice was administered alcohlic extract of *C. nocturnum* (250 mg/kg, p.o) for 9 days once daily and administered paracetamol (250 mg/kg, p.o) after three hours daily (Grish et al., 2009). The last group of mice were administered alcohlic extract of *C. nocturnum* (250 mg/kg, p.o) for 9 days once daily and three hours administered paracetamol (250 mg/kg, p.o).

Biochemical studies: Animals were sacrificed and blood was collected to estimate liver function tests, e.g. ALT, AST, ALP and total bilirubin.

Histopathological studies

Swiss albino mice were anesthetized by using chloroform and sacrificed. Livers were isolated from each animal and preserved in 10% formalin. The liver tissues were stained with hematoxylin. The prepared slides were than observed under the microscope.

Phytochemical screening

The preliminary phytochemical screening of various active compounds was accomplished by methods used by Farhan et al. (2012).

Statistical analysis

The values were expressed as mean ± SEM. The statistical analysis was carried out by one way analysis of variance (ANOVA). $p < 0.05$ was considered significant.

Results

Paracetamol increased the normal level of liver enzymes (AST, ALT, ALP and TB) which was decreased to normal level by treatment with silymarin (Table I). Similarly, treatment of mice with two doses of ethanol extract of *C. nocturnum* significantly decreased the elevated level of liver enzymes caused by paracetamol. Histopathological studied of liver supported the hepatoprotective effects of *C. notrunum* (Figure 1).

Discussion

The results indicated that hydroalcoholic extract of plant showed significant hepatoprotective effect. Silymarin treated group brought the elevated to the normal level which was used as standard drug. The results were further supported by the histopathological

Table I

Effect of aqueous ethanol (70:30) extract of *Cestrum nocturnum* on liver enzymes and total bilirubin

Groups	AST (IU/L)	ALT (IU/L)	ALP (IU/L)	TB (g/dL)
Normal (Distilled water)	26.2 ± 3.8	30.8 ± 5.1	124.8 ± 35.9	0.25 ± 0.1
Paracetamol (250 mg/kg)	105.8 ± 9.7	94.6 ± 7.4	296.8 ± 16.8	1.3 ± 0.1
Silymarin (50 mg/kg) + Paracetamol (250 mg/kg)	56.8 ± 4.8[b]	50 ± 6.5[b]	185.0 ± 6.8[b]	0.4 ± 0.1[b]
Ethanol extract (250 mg/kg) + Paracetamol (250 mg/kg)	70.2 ± 3.7[b]	65.4 ± 4.3[a]	235.4 ± 8.0*	0.9 ± 0.1[a]
Ethanol extract (500 mg/kg) + Paracetamol (250 mg/kg)	67.4 ± 6.46[b]	61.2 ± 13.06[a]	220.4 ± 21.66**	0.89 ± 0.07[a]

[a]p<0.05; [b]p<0.01

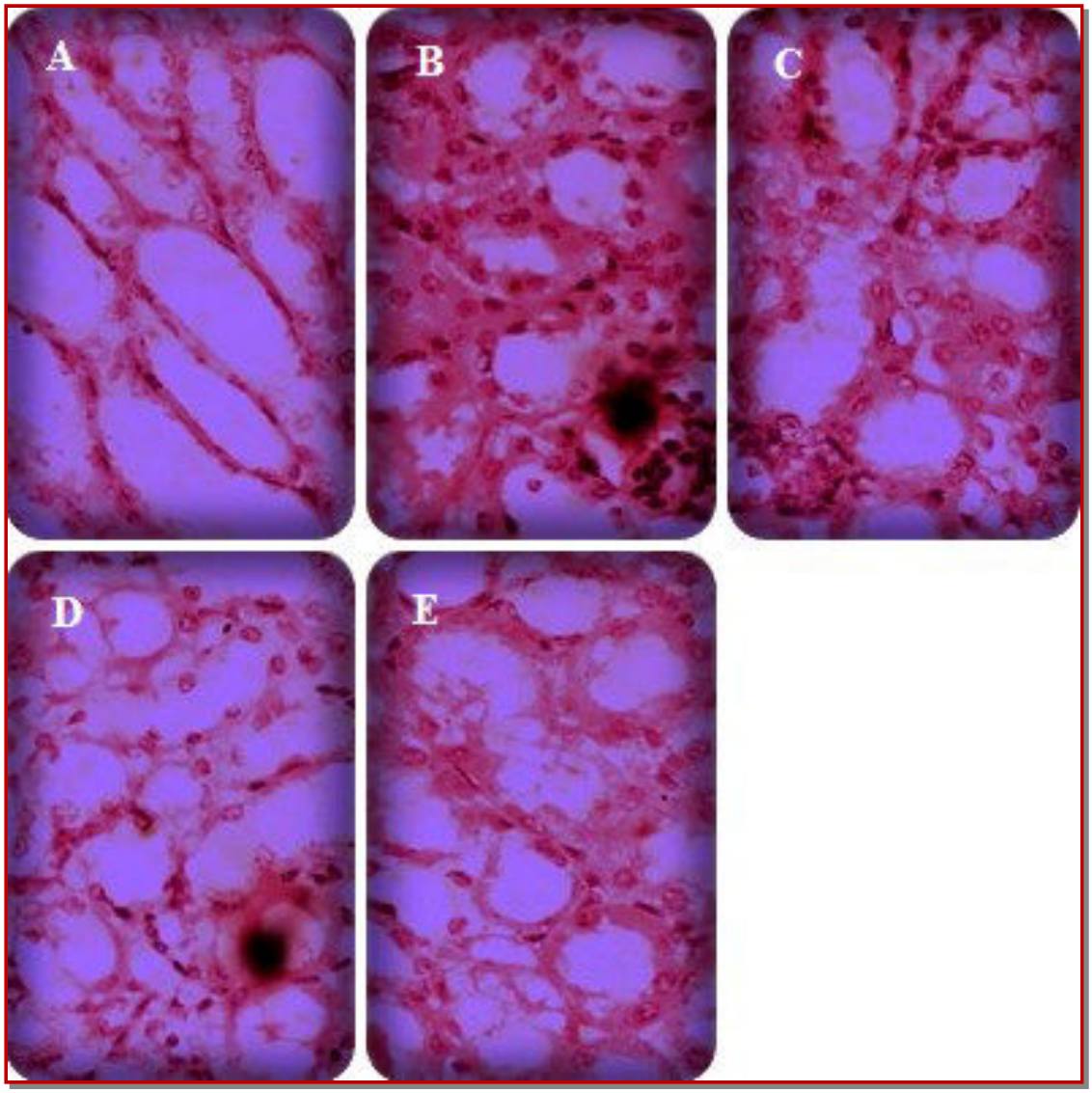

Figure 1: Histopathological pictures of (A) Normal hepatocytes; (B) Paracetamol treated group, marked inflammation, necrosis, sinusoidal constrictions and ballooning; (C) Silymarin-treated group, improvement in necrosis, inflammation, ballooning and moderate dilatation of sinusoids; (D) Extract 250 mg/kg treated group, mild inflammation ballooning and moderate sinusoidal dilatation; (E) Extract 500 mg/kg treated group, moderated inflammation, mild ballooning, mild necrosis and moderate sinusoidal dilatation

examination. The group treated with paracetamol showed the inflammation, necrosis and blooming. While the animal treated with plant extract showed less damage to the liver cells when compared with the paracetamol treated group. However animal group treated with silymarin showed no blooming, cell necrosis and mild inflammation.

Phytochemical analysis of *C. nocturnum* showed the presence of flavonoids, alkaloids, tannins, cardiac glycosides, carbohydrates, terpenoids, phenolic compound, and volatile oils. The same constituents have also been identified by Prasad et al. (2013). Most of the flavonoids have hepatoprotective activity (Ali et al., 2013). Therefore, the hepatoprotective activity of *C. nocturnum* may be due to the presence of flavonoids and phenolic components.

It was concluded from the present study that aqueous ethanol extract of leaves of *C. nocturnum* has hepatoprotective activity against the paracetamol-induced hepatotoxicity in albino mice.

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