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Antidiabetic activity of alcoholic stem extract of *Coscinium fenestratum* in streptozotocin-nicotinamide induced type 2 diabetic rats

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Abstract

The antidiabetic potential of the alcoholic stem extract of *Coscinium fenestratum* Colebr. (Menispermaceae), a medicinal plant widely used in the traditional Ayurveda and Siddha systems of medicine for the treatment of diabetes mellitus was evaluated in the STZ-nicotinamide induced type 2 diabetic model. Graded doses of the alcoholic stem extract were administered to normal and experimental diabetic rats for 12 days. Significant ($p < 0.05$) reduction in fasting blood glucose levels were observed in the normal as well as in the treated diabetic animals. Serum insulin levels were not stimulated in the animals treated with the extract. In addition, changes in body weight, serum lipid profiles, thiobarbituric acid reactive substance levels, glycosylated hemoglobin and liver glycogen levels assessed in the extract treated diabetic rats were compared with diabetic control and normal animals. Significant results were observed in the estimated parameters, thereby justifying the use of the plant in the indigenous system of medicine.

Keywords: *Coscinium fenestratum*; Menispermaceae; Streptozotocin-nicotinamide induced diabetes; Antidiabetic activity

1. Introduction

Coscinium fenestratum Colebr. (Menispermaceae), commonly known as, 'tree turmeric', is widely distributed in the Western Ghats (Tamilnadu and Kerala, India) and Ceylon. It is a woody climbing shrub with cylindrical stem, externally yellowish brown and internally yellow in colour. Its stem has often been used as a substitute for berberis, but it can be readily distinguished by the presence of large vessels in the wood, absence of annual rings and the crenate ring of sclerenchyma beneath the cortex. The stem yields a yellow dye, which is used either alone or in combination with turmeric and other colouring materials (The Wealth of India, 1950). The roots of *Coscinium fenestratum* contain alkaloids berlambine, dihydroberlambine, 12, 13-dihydro-8-oxo berberine, tetrahydroberberine, oxyberberine and noroxy hydrastinine (Datta et al., 1988; Malhotra et al., 1989).

It has been reported that the stem extract possesses hypotensive action (Singh et al., 1990). The infusion and tincture preparation of stem is widely used in the traditional Ayurvedic system for the treatment of diabetes mellitus (Varier, 1994). In the Siddha system of medicine, the powdered stems are dissolved in milk and given to the diabetic patients (Chinnaiah, 2002). The rural people of Kanyakumari District, Tamilnadu, India use the decoction of the stem for the treatment of diabetes (Kalavincela, 1998). The preliminary screening of this plant for its antidiabetic potential revealed that it has good antidiabetic activity (Mahapatra, 1997). With this in mind, the present study aimed to investigate the antidiabetic activity of *Coscinium fenestratum* using streptozotocin-nicotinamide induced type 2 diabetic rats.

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2. Materials and methods

2.1. Plant material

The stems of the plant material *Coscinium fenestratum* were purchased from Jogappa Shanbag Ayurvedic Store, Udupi, Karnataka, India during the month of August 2003. The plant was authenticated by Dr. Gopalakrishna Bhat, Department of Botany, Poorna Prajna College, Udupi, Karnataka, India. The voucher specimen (PP 526) has been deposited in the Department of Pharmacognosy, Manipal College of Pharmaceutical Sciences, Manipal, India.

2.2. Preliminary phytochemical screening

Preliminary phytochemical screening (Kokate, 1994; Harborne, 1998) revealed the presence of alkaloids, saponins, steroids, phenolic substances and carbohydrates. The alkaloid berberine was isolated from the alcoholic extract (yield 3.7%). The berberine content of the alcoholic extract of *Coscinium fenestratum* was estimated by HPTLC and was found to be 4.06%.

2.3. Preparation of alcoholic extract

About 160 g of stem powder was taken in the soxhlet extractor and extracted with ethanol for 72 h. The solvent was recovered by distillation in vacuo and the residue (yield 18 g) was stored in dessicator and used for subsequent experiments.

2.4. Animals

Healthy adult male Wistar albino rats between 2 and 3 months of age and weighing about 250–300 g were used for the study. The animals were housed in polypropylene cages, maintained under standard conditions (12 h light: 2 h dark cycle; $25 \pm 30\,^{\circ}C$; 35–60% humidity). They were fed with standard rat pellet diet (Hindustan Lever Ltd., Mumbai, India) and water ad libitum. The Institutional Animal Ethical Committee of KMC, Manipal, India (IAEC/KMC/03/2003-04), approved the study.

2.5. Acute toxicity studies

Healthy adult Wistar albino rats of either sex, starved overnight were divided into four groups ($n = 6$) and were orally fed with the alcoholic extract in increasing dose levels of 100, 500, 1000 and 3000 mg/kg body weight (Ghosh, 1984). The rats were observed continuously for 2 h for behavioral, neurological and autonomic profiles and after 24 and 72 h for any lethality (Turner, 1965).

2.6. Oral glucose tolerance test

The oral glucose tolerance test (Bonner-weir, 1988) was performed in overnight fasted (18 h) normal animals. Rats divided into four groups ($n = 6$) were administered 2% gum acacia solution, alcoholic extract (250 mg/kg), alcoholic extract (500 mg/kg) and glibenclamide (0.25 mg/kg), respectively. Glucose (2 g/kg) was fed 30 min after the administration of extracts. Blood was withdrawn from the retro orbital sinus under ether inhalation (to minimize the distress) at 0, 30, 60, 90 and 120 min of extract administration. The fasting blood glucose levels were estimated by glucose oxidase–peroxidase reactive strips (Accu-chek, Roche Diagnostics, USA).

2.7. Normoglycaemic study

For normoglycaemic study, rats were divided into four groups ($n = 6$) and were administered 2% gum acacia solution, alcoholic extract (250 mg/kg), alcoholic extract (500 mg/kg) and glibenclamide (0.25 mg/kg), respectively. The blood glucose levels were estimated on days 0, 5 and 12.

2.8. Induction of non-insulin dependent diabetes mellitus

NIDDM was induced (Masiello et al., 1998) in overnight fasted animals by a single intraperitoneal injection of 60 mg/kg STZ (Sigma Aldrich, Germany), 15 min after the i.p. administration of 120 mg/kg nicotinamide (Qualigens Fine Chemicals, Division of Glaxo, Mumbai, India). Hyperglycemia was confirmed by the elevated glucose level in the blood, determined at 72 h and then on day 7 after injection. The rats found with permanent NIDDM were used for antidiabetic study.

2.9. Experimental design

The diabetic animals, divided into four groups ($n = 6$) were administered 2% gum acacia solution, alcoholic extract (250 mg/kg), alcoholic extract (500 mg/kg) and glibenclamide (0.25 mg/kg), respectively, for 12 days. The fasting blood glucose levels were estimated on days 0, 5 and 12.

The effects of administration of the alcoholic extract on diabetic rats were estimated on the 12th day after the animals were sacrificed by decapitation. Serum insulin levels, serum lipid profiles, liver glycogen levels (Nicholas, 1956), Glycosylated hemoglobin levels, thio barbituric acid reactive substance levels (Ohkawa et al., 1979) and changes in body weight were assessed in the diabetic animals treated with extracts and compared with diabetic control and normal animals.

Serum insulin levels were estimated by a Radio Immuno Assay Kit issued by the Board of Radiation and Isotope Research, Bhaba Atomic Research Centre (BARC), Mumbai, India. Serum lipid profiles and glycosylated hemoglobin levels (turbidimetric inhibition immuno assay) were estimated by using an auto analyzer (Hitachi 912).