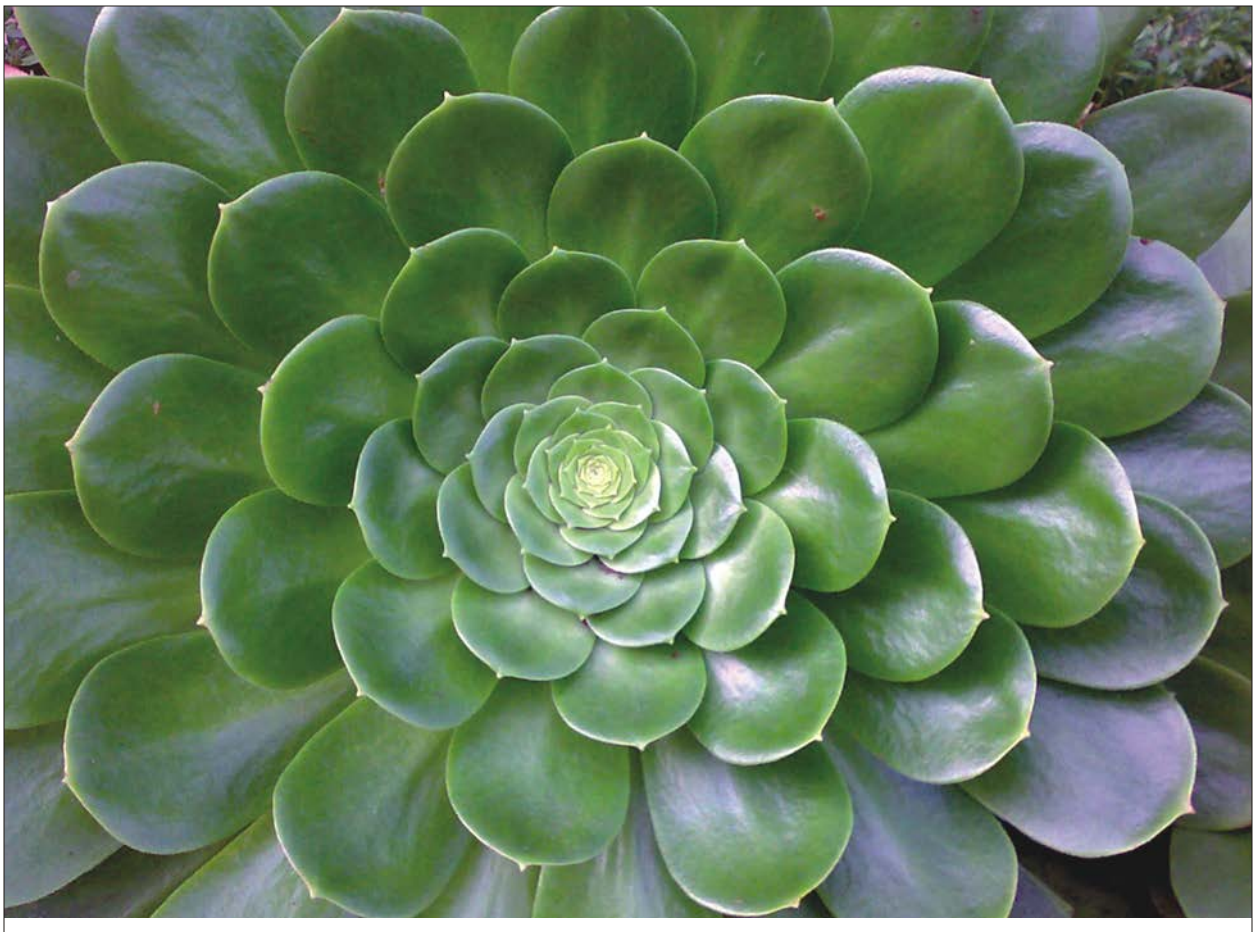

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Coscinium fenestratum (Gaertn.) Colebr.-A Review on this Rare, Critically Endangered and Highly-Traded Medicinal Species

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Abstract: *Coscinium fenestratum* (Fam. Menispermaceae), a critically endangered and highly-traded medicinal plant, has become very important in recent years due to its rarity and huge demand in the medicinal plant sector. This review throws light into the various research aspects undertaken in the past and present in the area of botany, pharmacology, chemistry, agrotechnology, harvesting practices and biotechnology along with its medicinal, ethnobotanical and other uses.

Key words: *Coscinium fenestratum*, *daruharidra*, critically endangered, medicinal plant, review, berberine

INTRODUCTION

The use of *Coscinium fenestratum* (Gaertn.) Colebr. as a source of the important ayurvedic raw drug *daruharidra* dates back to several centuries. Its stem has long been used in South India and Sri lanka as a yellow dye and bitter tonic and has found its way to Europe under the name False Calumba or Tree Turmeric. It is a dioecious, large, woody climber of the family Menispermaceae, a more or less primitive group, indigenous to the Indo-Malayan region (Fig. 1). In India, it is restricted to the Western Ghats, mostly in the high rainfall wet evergreen forests, moist evergreen, semi-evergreen and semi-deciduous forests at 500-750 m altitude (Kolammal, 1978; Sumy *et al*., 2000; Mohanan and Sivadasan, 2002). It grows well in humus rich soil having good drainage and areas having more than 2000 mm rainfall with an annual mean temperature of 27°C (Ravikumar and Ved, 2000; Sumy *et al*., 2000). Dried stem and root form the useful parts (Anonymous, 2005; Nambiar *et al*., 2000). The slow growing liana takes 15 years to reach its reproductive stage. But due to its huge demand for industrial consumption, it gets chopped down before it is fit to regenerate and also the traders directly engage tribals and other collectors for the supply of raw drug. Combination of rampant destruction of the forests along with over exploitation of the species for the raw drug market and very slow rate of regeneration has seriously depleted its population in the wild, making conservational measures very urgent (Tushar and Udayan, 2005). The threat status of this species has been assessed as Critically Endangered for Karnataka, Kerala and Tamil Nadu in India, due to more than 80% decline in the wild populations over the last 30 years (Ravikumar and Ved, 2000). Hence, this species is now banned from export by the Ministry of Commerce (vide notification No. 47 (PN)/92-97 dated 30 March 1994) (www.mtnforum.org). Wild populations of *Coscinium fenestratum* have also been important in Vietnam for berberine extraction to produce drugs since the beginning of the 1980s. They have been under heavy pressure since then and listed one among the total of 22 species in the red data book of Vietnam. In India and Sri Lanka, *Coscinium fenestratum* has already been listed as an endangered species (Agusta, 2003; Tran and Ziegler, 2001).

Distribution

Geographical: India, Sri Lanka, Thailand, Cambodia, Vietnam, Thailand, Peninsular Malaysia, Sumatra, Bangka, western Java and Borneo (Sharma *et al*., 1993; Agusta, 2003).

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Fig. 1: Photographs of *Coscinium fenestratum* (Gaertn.) Colebr

Regional: States of Kerala, Karnataka and Tamil Nadu (http://www.frlht-india.org). In Kerala, it occurs in Thiruvananthapuram, Thrissur, Malappuram, Palakkad, Kollam, Idukki, Wayanad (Nambiar *et al*., 2000), Kannur (Udayan *et al*., 2004) and Kozhikode districts.

Synonyms

Menispermum fenestratum Gaertn., *Coscinium peltatum* Merr., *C. wallichianum* Miers, *C. maingayi* Pierre, *C. usitatum* Pierre, *C. blumeanum* Miers var. *epeltatum* Boerl., *C. wightianum* Miers, *C. miosepalum* Diels, *C. fenestratum* var. *macrophyllum* Yamamoto, *C. fenestratum* var. *ovalifolium* Yamamoto, *Pereiria medica* Lindl (Forman, 1978; http://www.forest.go.th).

In India, vernacularly it is known as Haldigach (Bengali), Tree turmeric, False calumba, Columbo weed (English), Jhar-I-haldi (Hindi), Maradashina, Maramanjali (Kannada), Maramanjal, Manjavalli (Malayalam), Jhade-halade, Venivel (Marathi), Darvi, *daruharidra*, Pitadaru (Sanskrit), Venivel, Bangwellgetta (Sinhalese), Maramanjal, Manjalkodi, Atturam, Kadari, Udaravi, Pasamantram, Imalam (Tamil), Manu pasupu (Telugu) (Chopra *et al*., 1956; Caius, 1992; Sharma *et al*., 1993; Nambiar *et al*., 2000; Sumy *et al*., 2000; Anonymous, 2001).

Vernacular names in East are Vang dang (Cambodia), Akar kuning, kunyit-kunyit, kunyit-kunyit babi, tol, kupak, kopak (Malaya), Akar kunyit (Bangka), Akar kuning (Java), Abang asuh, binap kokop, upak-upak (Borneo), Perawan, dipang (Sarawak), Upak-upak (East Kalimantan), Khruea hen (North-eastern Thailand), Khamin khruea (South-eastern Thailand) (Agusta, 2003; http://www.arcbc.org.ph).

Related Taxon

Coscinium blumeanum Miers ex Hook.f. & Thoms., reported from peninsular Thailand, Penang and Pangkor Island of Peninsular Malaysia has a very restricted distribution. The name *C. blumeanum* has often been wrongly applied to *Coscinium fenestratum* and the uses reported in the literature for *C. blumeanum* probably refer to *C. fenestratum* (Agusta, 2003). It is used against boils (http://www.arcbc.org.ph).

Morphology

A large dioecious liana up to 10 m long, with yellow wood and sap. The stem and root slices are hard and woody. Wood is yellowish-brown in colour externally and yellow internally. The drug occurs in large woody, cylindrical, straight pieces, sometimes as much as 10 cm in diameter (Anonymous, 2001; http://www.ibiblio.org). Branchlets terete, brown tomentose, later glabrescent with disciform petiole-scars. Leaves simple, alternate, exstipulate, broadly ovate, rounded, truncate or shallowly cordate at base, acuminate at apex, 10-32×8-22 cm, glabrescent above, hoary yellowish-white tomentellous beneath, thinly coriaceous; main nerves 5-7, palmate, with 2 pairs of distal lateral nerves; midrib and other main nerves sunken, whitish tomentose beneath; petioles 3-16 cm long, conspicuously swollen at both ends, geniculate at base, inserted up to 0.8 cm above basal margin on the lamina of leaf; stipules absent. Inflorescence globose heads on 1-3 cm long peduncles, of 5-11 cm long racemes, 6-7 mm across, supra-axillary or on old leafless stems; bracts subulate, villous, closely pressed on the calyx, those of peduncles small, 4-5 mm long. Flowers unisexual, small, yellowish or whitish; sepals 9, in 3 whorls, imbricate, densely sericeous-pilose; petals absent; Male flowers: sessile or shortly pedicellate, ca. 1 mm long; sepals broadly elliptic to obovate, 1.5-2 mm long, densely sericeous outside, glabrous inside, yellow; outer ones 3-6, broadly elliptic, 1-1.5 mm long; inner ones 3-6, spreading, 1.5-2 mm long, yellow. Stamens 6, outer 3 free and inner connate to the middle; filaments ca. 1 mm long; anthers small, oval, adnate, outer ones one-celled, inner ones two-celled; pollen grains oblate-spheroidal or rarely spheroidal, triporate having reticulate tectum and lumina with fine granules (Ferguson, 1978). Female flowers: 3-6 free, subglobose carpels with slender subulate recurved or filiform styles, staminodes 6. Drupes 1-3, on globose, 7-8 mm in diam. gynophore, subglobose, 2.6-3 cm across, brown, orange or yellow, tomentellous; pericarp woody when dry, ca. 1 mm thick; endocarp bony, 2.2-2.5 cm in diam., with persistent calyx. Seeds whitish, subglobose with divaricate, much folded and divided cotyledons; hollow within, peltate, enclosing condyle; endosperm present. Embryo having very thin divaricate cotyledons with an irregular margin and a small superior radicle. Flowering season is from November-December (Sharma *et al*., 1993; Kolammal, 1978). Rema Shree *et al*. (2006) has conducted detailed micromorphological studies on the leaf and revealed that the dense tomentum composed of two types of trichomes in the abaxial surface of *C. fenestratum* is a striking feature that distinguishes this species from other genera of the tribe *Coscinieae*. The study conducted at the author's lab confirmed the chromosome number as 2n = 16.

Population studies of *C. fenestratum* revealed that they survive and regenerate naturally in disturbed habitats compared to undisturbed forest (Kathriarachchi *et al*., 2004). Assessment of genetic diversity in *Coscinium fenestratum* using RAPD markers revealed that there was less genetic diversity between the populations (Narasimhan *et al*., 2006). FRLHT, Bangalore, in collaboration with the Forest department has established a Medicinal Plant Conservation Area (MPCA) at Kulamavu in Idukki district of Kerala exclusively for the conservation of this endangered species.

Anatomy

Transverse section of young stem reveals single layered epidermis with certain ridges at regular intervals and is fully covered with very long uniseriate, multicellular tuft of hairs. Cortex consists of rectangular and polyhedral, thin walled, 4-6 layered collenchymatous cells consisting of very prominent yellowish bands of hard stone cells with crystals inside. Thirty to thirty five vascular bundles are arranged in a broken ring. Just above each vascular bundle, arches of 15-18 layered sclerenchymatous cells with lysigenous cavities are present opposite to the phloem in definite patches. In between the arches, 2-4 layers of chlorenchymatous tissue are present. The 1-2 layered interfascicular cambium originates in between the bundles, in line with the fascicular cambium, resulting in a ring of 2-6 layered cambium. The primary phloem cells are found below these arches. Secondary phloem, consisting of large sieve tubes and companion cells is present above the secondary xylem forming a cup like structure. Primary xylem consists of vessel elements with pitted thickening, annular and spiral tracheids and fibres. Outermost zone is cork, composed of 20-30 rows of moderately thick-walled cells with inclusions. Anomalous secondary thickening with fissured xylem and continuous vascular cambium is observed in the mature stem. Wood region consists of xylem strips, composed of large sized vessels, fibres, axial parenchyma and tracheids that are alternate to the thick walled medullary rays. Broad medullary rays, consisting of 30-40 layers and lying alternate to the conductive tissues, occupy almost half of the xylem region. Pith is very prominent and parenchymatous consisting of thick walled and closely packed cells towards the periphery and large, circular to polyhedral and comparatively thin walled cells with inter cellular spaces towards the centre. Starch grains are smaller and present throughout the pith and medullary rays. A group of yellow coloured stone cells are also present in the center of the pith (Nambiar *et al*., 2000; Rema Shree *et al*., 2005). The stem of *Coscinium* can be easily distinguished from that of *Berberis* by large vessels in the wood, bright greenish yellow colour with open, porous structures and devoid of crenate rings. Wood is lighter and softer than *Berberis* sp. (Anonymous, 2001).

Outermost region of the root consists of 10-13 rows of thick walled cells. The cortex is rectangular and thin walled with oil drops in certain cells. Inner boundary of the cortex forms a wavy band of several arches composed of yellow coloured thick walled cells composed of longer, elongated stone cells that form major portion of the band in addition to a few short sclerenchymatous cells. Below these arches some crushed secondary and primary phloem are seen. Wood consists of large sized vessels, xylem parenchyma and thick walled fibres. Medullary rays consist of multiseriate radially elongated thick walled cells, seen in between the secondary xylem region. Pith is not discriminable in old roots (Nambiar *et al*., 2000).

Transverse section of petiole shows single layered epidermis with thin cuticle and multicellular and uniseriate trichomes. Cortical region is parenchymatous with vascular bundles in a ring and a sclerenchymatous bundle cap above each bundle. In the centre of the petiole, parenchymatous cells are large and loosely arranged (Nambiar *et al*., 2000).

Transverse section of the leaf midrib shows thick and thin-walled rosette cells. Epidermis is single layered with lower region possessing large number of multicellular and uniseriate trichomes. The epidermal cells are tangentially elongated with straight anticlinal and smooth periclinal walls with

cuticle. The mesophyll consists of 1-2 layered, thick walled, highly chlorophyllous palisade tissue and 2-4 layered, thin walled and spongy tissue with abundant intercellular spaces. Vascular bundle is encircled by a wavy ring of 2-10 layers of sclerenchymatous tissue. Collenchyma and parenchyma cells are present wherein the latter is mostly filled with yellow coloured berberine deposits. Stomata are Ranunculaceous type. Stomatal index is 29.31, palisade ratio is 3.81 and vein-islet number is 29.31 (Nambiar *et al*., 2000; Remashree *et al*., 2006). The leaf anatomy of the tribe *Coscinieae* has been described by Wilkinson (1978).

Powder colour of stem and root is yellow and of leaf is dark brown. It has a bitter taste without any characteristic odour (Pinho *et al*., 1992; Nambiar *et al*., 2000; Anonymous, 2005).

Quantitative Standards

Root

Foreign matter: Not more than 1%, Ash: Not more than 2%, Acid insoluble ash: Not more than 0.4%, Ethanol soluble extractive: Not less than 11%, Water soluble extractive: Not less than 10% .

Stem

Foreign matter: Not more than 1%, Ash: Not more than 3%, Acid insoluble ash: Not more than 2%, Ethanol soluble extractive: Not less than 3%, Water soluble extractive: Not less than 8% (Anonymous, 2005).

Medicinal Uses

The medicinally active compound is berberine, an isoquinoline alkaloid with numerous bioactivities (Birdsall and Kelly, 1997). The drug is useful in vitiated conditions of *kapha* and *vata*, inflammations, wounds, ulcers, jaundice, burns, skin diseases, abdominal disorders, diabetes, fever and general debility (Warrier *et al*., 1994; Agusta, 2003). An infusion, tincture and concentrated liquor are also prepared to wash wounds and skin rashes (http://www.ibiblio.org). Stem pieces are boiled and one cup is given in case of a fresh, deep cut, being the most common use against tetanus. It purifies the blood (http://www.sinharaja.4t.com). Decoction of stem is given internally in cases of bites from monkeys, snakes, brahmin-lizards and geckos (Caius, 1992). The root bark is used for dressing wounds, ulcers and in cutaneous leishmaniasis (Anonymous, 2001). It is known to treat influenza and eye diseases. Simply boiling the pieces and bathing with the water relieves body pain. *Coscinium* is also used to treat bleeding piles and excessive bleeding during menstruation. For snakebite poisoning, paste of *Coscinium* and turmeric is applied. For quick healing of ulcers, *Coscinium* powder is applied after mixing with ghee (http://www.island.lk). Many traditional healers of Chhattisgarh use the bark in their treatments. A combination of the bark and honey is taken internally for the treatment of jaundice. Bark is also used in the treatment of leucorrhoea and other gynaecological troubles. According to them its aqueous extract is more useful but due to non-availability of fresh bark, a decoction by boiling the bark in water is used by taking it in empty stomach daily morning (http://www.botanical.com). It has also been applied in a complex decoction after childbirth in Peninsular Malaysia. In Vietnam, tablets made from crude alcoholic *C. fenestratum* extracts are prescribed to cure dysentery (Agusta, 2003). Old parts or roots is crushed and boiled for drinking against colic and stomachache (Tran and Ziegler, 2001). Plant is used in the treatment of fractures (Asolkar *et al*., 1992; Anonymous, 2001). Infusion of *C. fenestratum* is used in bath tubs, in facial creams as an antiseptic and as a common home remedy by the mothers in Sri Lanka (http://www.ayurvedainsrilanka.com). Diabedrink (Diabetic Food Supplement) for Diabetic patients is an ideal drink for people who need supplementary care along with medical treatment for Diabetes Mellitus (http://www.fuzing.com). Rhumatone is used in the treatment of chronic musculo-skeletal disorders such as arthritis, rheumatism, gout, fibromyalgia and in general massage therapy

(http://www.araliya.com). *Coscinium* is also useful in cholera and gastroenteritis (http://www.ikisan.com). It is used in over 62 ayurvedic preparations like Aswagandharishtam, Khadirarishtam, Anuthailam, Katakakhadiradi kashayam, Elaneer kuzhampu, Mahapanchagavyam etc (Nambiar *et al*., 2000). Other preparations made include Infusum Coscinii, I.C.A.-infusion of *Coscinium*. Liquor Coscinii Concentratus, I.C.A.-concentrated solution of *Coscinium* and Tinctura Coscinii, I.C.A.-tincture of *Coscinium* (http://www.ibiblio.org).

Safety Aspects and Dosage

Traditionally prescribed dose of 1-3 g of root powder is considered safe (Anonymous, 2005)

Toxicology

The wood is considered to be toxic causing vomiting, diarrhoea and cramps (Warrier *et al*., 1994). But the water extract of *C. fenestratum* showed negative results against acute and subchronic toxicity tests (Wongcome *et al*., 2007).

Uses (General)

The yellow dye obtained from the wood of *Coscinium fenestratum* has been used in traditional fabric dyeing in Malaysia (Warrier *et al*., 1994). *Coscinium* is an active ingredient of Sri Lanka's ayurvedic shampoo Apsara Venivel and soap Aspara Calumba wood used to enhance the skin complexion (http://www.dailymirror.lk). It is also used in ayurvedic bath soap, bath oil and shower oil under the trade name Araliya (http://www.araliya.com), in Paspanguwa, an ayurvedic general tea along with *Zingiber officinale* Rosc., *Coriandrum sativum* L., *Oldenlandia corymbosa* L. and *Solanum surattense* Burm.f. (http://www.drmel.co.nz) and in Samahan, a concentrated, water soluble preparation of selected medicinal plants that belongs to the ancient remedy that assists the immune system known in Sri Lanka as Peyava. Over 3 million sachets are being safely used every month by Sri Lankans and are exported to the UK, USA, Australia, New Zealand, Germany, Switzerland, India, Singapore and the Middle East (http://www.linknaturalproducts.com). The fluorescent property of berberine has been made use for the conservation of world's oldest document Dunhaung Diamond sutra, obtained from Caves of China, by dyeing these documents with it (http://plant-tc.coafes.umn.edu). *Coscinium* is used intensively by villagers for the production of medicinal beverages (Gunathilake and Gunathilake, 1980). Swallowing the juice obtained from chewing the roots, before drinking, removes the effects of intoxication (Forman, 1978).

Coscinium is used as a substitute/adulterant for Calumba (*Jateorhiza palmata* (Lam.) Miers), a climbing plant indigenous to Portuguese East Africa and growing freely in forests near the Zambesi (Nambiar *et al*., 2000; http://www.ibiblio.org). The dried root is insect resistant (Anonymous, 2001). The fruits are eaten by animals such as orangutans, gibbons and macaques and birds (Anonymous, 2001; Agusta, 2003).

Ethnobotany

Stem pieces are used against rheumatism, jaundice and skin diseases by the Oorali tribes of Idukki district and Kaadar tribes of Thrissur district (Udayan *et al*., 2005a,b). The traditional healers of Gandai region use the powdered bark in treating eye troubles, both internally as well as externally. Internally it is used in combination with other herbs and externally they prepare a paste by mixing bark with cow milk and apply it. According to them, the external application removes the extra heat from eyes (http://www.botanical.com). In Malaysia, the entire plant is boiled in water to make a tea and stem is used for thinning and yellowing skin (Kulip, 2003). It is also reported to be a constituent of Malayan arrow and dart poison, used by *sakais* (Caius, 1992).

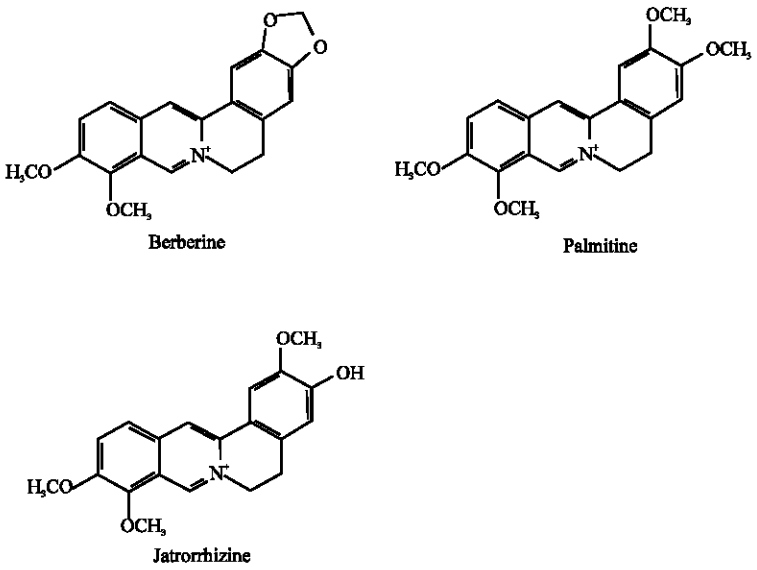

Fig. 2: Chemical structures of major compounds present in *Coscinium fenestratum*

Chemical Composition

The major alkaloids are yellow crystalline berberine, protoberberine and jatrorrhizine (Fig. 2). Many other alkaloids, mainly of the protoberberine type, isolated from stem and root are magnoflorine, berberrubine, thalifendine, palmitine and oxyberberine (Katti and Shintre, 1930; Child and Nathanael, 1943; Varier and Pillai, 1943; Siwon *et al*., 1980; Jayaweera, 1982; Malhotra *et al*., 1989; Pinho *et al*., 1992; Agusta, 2003; Anonymous, 2005). The stem and root also contain ceryl-alcohol, saponin, hentriacontane, sitosterol, palmitic acid, oleic acid and sitosterol glucoside (Katti and Shintre, 1930; Siwon *et al*., 1980; Anonymous, 2001). The total extractives are: petroleum ether 2.1%, ether 2.5%, alcohol 6.9%, chloroform 3.4%, acetone 4.1%, benzene 2.7% and water 5.1% (Kolammal, 1978; http://www.ibiblio.org). Other compounds reported from the stem and root are N,N-dimethyllindacarpine (Siwon *et al*., 1980), oxypalmitine, (-)-8-oxotetrahydrothalifendine, (-)-8-oxoisocorypalmine, (-)-8-oxothaicanine, (-)-8-oxo-3-hydroxy-2,4,9,10-tetramethoxyberberine, (-)-8-oxocanadine (Pinho *et al*., 1992), 12,13-dihydro-8-oxoberberine,5,6,13,13a-tetrahydro-9,10,dimethoxydibenzo(a,g) 1,3-benzodioxolo(5,6a) quinalizine-8-one, stigmasterol (Malhotra *et al*., 1989), berlambine, dihydroberlambine and noroxyhydrastinine (Datta *et al*., 1987).

Phytochemical Studies

Berberine is present in all parts of the plant, the higher percentage reported in old stem and old roots. Quantitative analysis of berberine was done in the samples from stem pieces by HPLC (Narasimhan and Nair, 2004b). TLC identity test and TLC-densitometric method was standardised for quantitating berberine in the methanolic extracts of stem and root. The samples analysed contained 1-2% (w/w) berberine (Anonymous, 2005). Maceration with 80% ethanol gave the highest content of berberine in the extract. TLC of the extracts from different methods showed a similar pattern (Rojsanga *et al*., 2006). An enzyme tetrahydro berberine oxidase (THB) involved in final step of berberine biosynthesis, has been partially purified from the plant tissue and cell cultures. Supplementation of copper sulphate in the production medium also showed increased activity of the enzyme along with increase in berberine production (Anonymous, 2001).

Pharmacological Studies

Hypoglycemic activity was exhibited by the alcoholic stem extract of *Coscinium fenestratum* for the treatment of diabetes mellitus evaluated in streptozotocin-nicotinamide induced type 2 diabetic rats (Mahapatra, 1997; Punitha *et al*., 2005; Shirwaikar *et al*., 2005a) and also by the aqueous stem extract in non-insulin dependent diabetic rats (Shirwaikar *et al*., 2005b). A 50% ethanolic extract of the stem material has been found to possess hypotensive action in anaesthetised dogs, rats and guinea pigs in a dose-related pattern (Singh *et al*., 1990). The water extract from *C. fenestratum* is effective in reducing blood pressure in anesthetized normotensive rats (Wongcome *et al*., 2007). Antioxidant effect of methanolic extract of the stem powder was studied by Venukumar and Latha (2002) in carbon tetrachloride-intoxicated rat liver and by Punitha *et al*. (2005) in streptozotocin-nicotinamide induced type 2 diabetic rats. Anti-hepatotoxic activity of methanolic extract of the stem was confirmed against carbon tetrachloride-induced hepatopathy in rats (Venukumar and Latha, 2004). Antinociceptive effects on mouse formalin test have also been studied (Chitra *et al*., 2004). Methanolic extract had the strongest *in vitro* antiplasmodial activity with EC(50) value of 0 .5 μg mL^{-1}, inhibiting the growth of the chloroquine-resistant *Plasmodium falciparum* strain FCR-3 with EC(50) values less than 10 μg mL^{-1} (Tran *et al*., 2003). The polar components of *C. fenestratum* are cytotoxic against laryngeal cancer cell lines (http://pharm.swu.ac.th). Neurotoxicity of the stem has been studied by Wattanathorn *et al*. (2006). The alcoholic stem extract also possesses good hypolipidemic activity (http://www.freshpatents.com).

Methanolic, methanol-water and water extracts of *C. fenestratum* was tested for their antiproliferative activities against human HT-1080 fibrosarcoma cells. Among these methanol extracts and methanol-water (1:1) extract exhibited antiproliferative activities in a concentration-dependent manner. *C. fenestratum* showed selective activity against lung carcinoma and lung metastatic cell lines, A549, LLC and B16-BL6. Characteristic morphological change and DNA fragmentation indicated the antiproliferative activity to be due to the induction of apoptosis (Ueda *et al*., 2002).

The ethanolic extract of *C. fenestratum* significantly suppressed *in vitro* anti-herpes simplex virus type 1 (HSV-1) plaque formation in Vero cells (Ekalaksananan, 2006). Extracts of *Coscinium* showed strong antifeedant activity against the fourth instar larvae of Mexican bean beetle, *Epilachna varivestis* Muls., Coccinellidae (Jayasinghe *et al*., 2003). The aqueous and alcoholic extracts of the stem exhibited antibiotic and antimicrobial activities (Ray and Majumdar, 1976; Jayaweera, 1982; Anonymous, 2001). Selective inhibitory action on *Clostridium tetani* was observed at a concentration of 6.25 mg mL^{-1} (Anonymous, 2005). Nair *et al*. (2005) reported that the antibacterial activity of *C. fenestratum* is mainly due to the presence of berberine. Pharmacological screening of an aqueous methanol (1:1) extract showed convulsant activity. In clinical tests in Vietnam, the extract also showed distinct activity on *Staphylococcus aureus* and *Streptococcus hemolyticus*, which may cause inflammation and infection especially in women after childbirth. The pharmacological effects of berberine have been fairly well investigated. It has been found active against a number of gram-positive as well as gram-negative bacteria and also against a number of fungi. It was also effective against experimentally induced intestinal amoebiasis in rats and showed growth inhibition of Ehrlich and lymphoma ascites tumour cells. Berberine is also present in high concentrations in other *Menispermaceae species*, e.g., in *Arcangelisia flava* (L.) Merr., which is used for similar complaints as *C. fenestratum* (Agusta, 2003).

Cultivation Practices (Agrotechnology)

Seed viability levels, optimum seed storage conditions, suitable seed germination methods and vegetative propagation techniques were carried out in this species (http://www.pgis.lk). The plant regenerates from stumps of old plants and also through seeds, but the rate of regeneration is found to

be extremely low (Harinarayanan *et al*., 1994). Both freshly collected seeds and those stored failed to germinate even after pretreatments (puncturing the testa, acid, hot, cold water treatments, etc). Seeds have a dormancy period of 6 months and about 200 days are taken for 90% germination. Incubating the seeds at 30°C and 80% relative humidity, 90% germination is obtained in 60 days. The fresh stem cuttings of pencil size are suitable for vegetative propagation. Cuttings of about 15 cm length are dipped in IAA 500 ppm for 24 h. Vegetative buds appear after 2 weeks that produce nodes and internodes. Within a month the shoots attain a length of 45 cm. After one month of growth, the cuttings produce 1-2 roots and after one more month the plants can be transplanted to larger containers for hardening. The seedlings or sprouted cuttings thus developed must be planted in pits at a distance of 1-1.5 m away from the trunk of the tree that will provide support. Irrigation may be provided depending on climate and soil conditions. About 360 seeds constitute 1 kg (Nambiar *et al*., 2000; Sumy *et al*., 2000).

Tissue Culture

Preliminary studies on *in vitro* multiplication of *Coscinium fenestratum* was carried out by Nair and Seeni (2003). A protocol for obtaining berberine-producing callus and cell suspension cultures were established in this plant from the petiole segments. Among the auxins tested, highest yield of berberine (5.79 mg/30 mL; 4.14 times to that of control) was obtained with 4 mg L^{-1} of NAA, while the best cell growth (214.43 mg dry wt., 1.96 times to that of control) was observed in the presence of 2 mg L^{-1} of 2,4-D (Narasimhan and Nair, 2004a). Intercellular berberine and berberine released into liquid media were studied by Narasimhan and Nair (2004b).

Harvesting and Post Harvest Handling (Semi-processing)

The root, stem and branches are collected as and when required from the forest. The mature liana stem are cut 50 cm above the base leaving short stump for coppicing. Three yearly harvesting cycles are ideal (Ekanayake *et al*., 2004). They are dried for a few weeks and used or sold in the market. The dried stems are not liable to insect or pest attack and can be stored for long periods without any loss to the medicinal properties. In preparing them for use, the stem is first scraped to remove the outer corky rind and then gently beaten with wooden mallets till the useful part separates out. Wood is not ordinarily used for medicine (Kolammal, 1978). But nowadays the entire stem is cut into pieces and used along with the root. For obtaining the yellow dye, the wood is broken into pieces, steeped in water, crushed in rice-husking machine and squeezed. The dye is used either alone or in combination with turmeric (Anonymous, 1998).

Adulterants/Substitutes

In North India, different species of *Berberis* viz., *B. aristata* DC., *B. asiatica* Roxb., *B. lycium* Royle, *B. vulgaris* L., supposed to possess more or less similar properties are being used as *daruharidra*. The physicians of South India use *Coscinium fenestratum* as the source, which is often adulterated with all the above species, *Mahonia leschenaultii* (Wall. ex Wight & Arn.) Takeda ex Gamble, *Berberis tinctoria* Lesch., *Anamirta cocculus* (L.) Wight & Arn. (Mooss, 1983; Menon, 2003; Anonymous, 2005) and also vice versa.

Requirement and Market Price

Annual requirement of *Coscinium fenestratum* in the traditional medicine sector of Sri Lanka is 54 ton (IUCN, 2001) and that of Kerala state in India is 143 ton.

Retail market price: Rs. 60/- per kg of dried stem

Intellectual Property Rights

An US Patent 20060018978 was recorded on the use of *Coscinium fenestratum* in a pharmaceutical product. Its aim was to develop a pharmaceutical product of medicinal herbal origin for maintaining good health in a healthy person as well as in diabetic patient (http://www.freshpatents.com).

CONCLUSIONS

Many useful pharmacological activities have been proved for the alkaloids of *Coscinium fenestratum*. Hence, biotechnological approaches have to be sought for the large scale production of the alkaloids such as berberine. Conservation of this very important and rare medicinal plant is possible only through generating awareness among the raw drug collectors and the multiplication of the plant species through tissue culture and conventional propagation methods. This will finally lower the pressure on wild populations of the very important endangered medicinal species.

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