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Ethnopharmacology and toxicology of threatened tuberous plant genus *Ceropegia* sp. L.: A review

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Abstract

Ceropegia L. belongs to the family Asclepiadaceae, native to Africa, Southern Asia and Australia. It is a botanically a curious genus, mainly distributed in Western Ghats. The genus comprises of 200 species found throughout the world is mainly distributed in subtropical and tropical Asia. In India 55 species are reported, in which 28 are endemic to Peninsular India. *Ceropegia* species have sound ethnomedicinal values and is an important component in several indigenous drug preparations. The pharmacological importance of the genus is mainly due to the presence of 'cerpegin' a pyridine alkaloid, apart from the different potential phytoconstituents such as steroids, terpenoids, anthocyanins, anthracene glycosides, coumarins, flavonoids, fatty acids, phenolic compounds, alkaloids and carotenoids. The main objective of this review is to cover its botanical characterization, distribution, indigenous and traditional uses, ethnopharmacology and toxicology studies of reported species of *Ceropegia*.

Keywords: *Ceropegia*, steroids, ethnopharmacology, Cerpegin

1. Introduction

Ceropegia genus with 200 species are present throughout the world, also has distribution in tropical and subtropical Asian countries. In India 55 species have been reported, among them, 28 are endemic to peninsular India and 16 species of them are included in the Red Data Book (Nayar and Sastry, 1987) [32]. Documentation of 56 species, two subspecies and three varieties for India has been done (Karthikeyan *et al.* 2009) [25], among them a few species are edible and few are domesticated as ornamental plants (Mabberley, 1997)⁴. These species of *Ceropegia* have many common names snake creeper, lantern flower, parasol flower, string of hearts, bushman's pipe, parachute flower, wine-glass vine, necklace vine and rosary vine. Several *Ceropegia* species are included under the categories of rare, endangered, vulnerable, extinct and threatened plants (Jagtap and Singh, 1999) [14]. Several workers attracted towards the *Ceropegia* genus in recent years due to its potent medicinal properties and rarity of its species. There are various reasons for threatened status of *Ceropegia* species such as severe anthropogenic pressures on the forest land, overexploitation of *Ceropegia* species for their tubers by humans and various animals and hence these species needs special attention and requires proper conservation strategies.

2. Botany and distribution

The genus *Ceropegia* L. belonging to the family Asclepiadaceae is a botanically curious genus with largest number of species native to Africa, Southern Asia and Australia. In India these species are mainly distributed in the Western Ghats (Jagtap and Singh, 1999) [14]. *Ceropegia* L. shows a variety of growth habits such as erect herbs, climbers; rarely sub shrubs, leafless succulents and non-succulent twiners. Check-list of 55 *Ceropegia* species, including status of taxa, phenology, and habit, importance of the conservational strategies, Indian and World distribution has been summarised (Ramamurthy *et al.* 2012) [38]. Most of the species are producing tuberous roots whereas few have fascicled roots (Surveswaran *et al.* 2009) [42]. Different *Ceropegia* species with their diagnostic features and distribution in India are shown in Table 1.

3. Traditional uses and preparations

Ceropegia species have sound and numerous ethnomedicinal values and hence is an important component in several indigenous drug preparations (Ayurveda).

Bhilla tribal group in Northwest Maharashtra region are using *Ceropegia hirsuta* fresh root for treatment of stomach disorders (Kambale *et al.* 2008) [21]. In India, practise of taking a teaspoon of tuber paste daily once for 3 days is reported to be effective for treating the stomach related problems (Reddy *et al.* 2009) [39]. The whole plant extract used as an antidote for snack bite (Suresh and Paulsamy, 2010) [41] and fleshy stem is used for treatments of stomach and gastric disorders in traditional and folk medicines. The combination of goat milk with crushed stems of *Ceropegia juncea* is taken orally for three days to cure decess ulcer by Paliyan/Paliyar tribe in Sirumalai hills of Southern India (Karuppuswamy, 2007) [26]. Starchy edible tubers with medicinal properties gained economic importance (Jagtap and Singh, 1999) [14]. Fresh tubers are boiled before consumption to remove bitterness. Tuberos roots are rich sources of starch, sugar, gum, albuminods and medicinally potent alkaloid 'cerpegin' (Anonymous, 1980) [2] used as blood purifier and as nutritive tonic. Kani, the tribal group from Kerala-Tamil Nadu border used *Ceropegia spirallis* corm extract for purification of blood and in treating syphilis (Das *et al.* 2008) [9]. The alkaloid cerpegin extracted from the tubers of *Ceropegia* is a valuable constituent in the traditional Indian drug preparations against various ailments especially dysentery, diarrhoea and to cure sneezing, cold and eye diseases in Bihar region and also the seed paste has been used for treatment of Deafness (Kirtikar and Basu, 1935) [28]. *Ceropegia bulbosa* var. *bulbosa* Roxb. Tuber paste is applied on the inflammation of skin and decoction taken to get rid of urinary bladder stone. Aqueous extract of *C. bulbosa* contains steroids, polyphenols, sugars and potassium (Khare, 2007) [27]. Tubers of *Ceropegia hirsuta* Weight Arn. are eaten raw or taken in the form of decoction for stomach ache, dysentery, diarrhoea and warm tuber paste is used for treatment of viral infections (Deshmukh and Rathod, 2013) [10]. Raw leaf and flower extracts of *C. pusilla* is used for the treatment of nervous weakness in India (Rajan *et al.* 2005) [37]. The whole plant extract of *C. ciliata* Wight is used for treatment of fever while *C. oculata* raw tubers are used for treatment of fever in children and stomach ache (Jagtap *et al.* 2006) [15].

4. Chemical constituents

Tuberos roots of *Ceropegia* L. are sources of relatively rare alkaloid 'cerpegin' has been elucidated as 1,1,5-trimethylfuro-[3,4-c]pyridine-3,4(1H,5H)-dione [1, 2] and identified and isolated from *Ceropegia juncea* as new pyridine type alkaloid that, possess a wide range of properties such as anti-inflammatory, analgesic, tranquilizer anti-ulcer, mast-cell stabilizing, hepatoprotective, antipyretic, local anaesthetic and antiulcer (Adibatti *et al.* 1991) [1]. *Cerpegia* is the only naturally occurred example of bicyclic furo [3,4-c] pyridine ring system, a convenient method was developed for pyridine alkaloid 'cerpegin' and its various new C-1 and N-5 derivatives in high yields (Hovhannisyanyan, 2014) [12]. Presence of different phytoconstituents such as steroids, terpenoids, anthocyanins, anthracene glycosides, coumarins, flavonoids, fatty acids, phenolic compounds, alkaloids, carotenoids have been reported in *Ceropegia juncea* (Karayil and Veeraiah,

2014; Kalimuthu and Prabakaran, 2013) [12, 23]. The phenolic compounds such as gallic acid, vanillin, catechol and ferulic acid, flavonoids and their antioxidant capacity were identified and reported from various extracts of three species of *Ceropegia* such as *Ceropegia spiralis*, *Ceropegia panchganiensis* and *Ceropegia evansii* from the Western Ghats of India (Chavan *et al.* 2013) [7]. *Ceropegia pusilla* tubers were screened for secondary metabolites and confirmed the presence of glycosides, alkaloids, flavonoids, tannins, saponin, triterpenes phenol and steroids through phytochemical screening and GC-MS analysis (Kalimuthu and Prabakaran, 2013) [16]. In North Central Nigeria, *C. deightonii* plant used for the treatment of various infectious diseases was screened for phytochemicals and revealed the presence of tannins, flavonoids, steroids, terpenes, saponins, anthraquinones, cardiac glycosides and triterpenes, glycosides are seems to be rare in *Ceropegia* (Awolowo, 2014) [3]. *C. bulbosa* tuberos roots consists of polyphenols, steroids, fats, albuminoids, sugars, potassium and the active constituent an alkaloid cerpegin (Monika *et al.* 2012) [31]. Tubers and leaves of different *Ceropegia* species are screened for ascorbic acid content and found that ascorbic acid content was highest in tubers of *C. hirsute* and lowest in *C. bulbosa* leaves, concluded that leaves contained higher ascorbic acid than the tubers. A novel coumarin, (E)-5-(4-methyl-2-oxo-2H-chromen-8-yl)-2-(4-methylbenzyl)-3-oxopent-4-enitrile was isolated from *Ceropegia juncea* (Karayil *et al.* 2014) [24]. Coumarins have attracted the attention in recent years due to their diverse properties such as hepatoprotective, anti-inflammatory, antioxidant, antithrombotic, anti allergic and anti-carcinogenic activities (Karayil *et al.* 2014) [24].

5. Pharmacological activities

Ceropegia species has been subjected to various pharmacological investigations by various researchers. The pharmacological importance of the genus *Ceropegia* L. is mainly due to the presence of 'cerpegin', a pyridine alkaloid.

5.1. Antimicrobial activity

Studies on antibacterial activity of three *Ceropegia* species such as *C. spiralis*, *C. juncea* and *C. candelabrum* var. *candelabrum* on human pathogens *Klebsiella pneumoniae*, *Staphylococcus aureus*, *Escherichia coli* and *Pseudomonas* has been carried out, findings showed that ethanol extract from the *In-vitro* plant showed a higher antibacterial activity as compared to chloroform and aqueous extracts⁴⁰. Antimicrobial activity was assayed with whole plant extract of *Ceropegia pusilla* against five species of bacteria (*Staphylococcus aureus*, *Escherichia coli*, *Klebsiella pneumoniae*, *Shigella sonnei*, *Bacillus* sp.) and four species of fungi (*Candida albicans*, *Aspergillus* sp., *Penicillium* sp., *Mucor* sp.) by agar well diffusion method⁴¹. The crude extract of *C. deightonii* showed antimicrobial activity against pathogens *Staphylococcus aureus*; *Streptococcus faecalis*; *Escherichia coli*; *Shigella dysenteriae* and *Candida albicans*⁵. Leaf extract of *C. thwaitesii* used for the plant mediated synthesis of AgNPs (silver nanoparticles) showed antimicrobial activity against *S. typhii* and *B. subtilis*⁴².

Table 1: Different *Ceropegia* species with their diagnostic characters and distribution in India

<i>Ceropegia</i> species	Plant characters	Distribution in India	References
<i>C. juncea</i> Roxb.	Fleshy twining herb with small fleshy and lance-shaped leaves almost looks like leafless. Flowers are goglet shaped and borne in umbels with yellow, green and purple colors.	Idukki and Palakkad district of Kerala	Boomibalagam <i>et al.</i> 2015 [6]
<i>C. bulbosa</i> Roxb.	Inflorescence is axillary cyme with 4-10 flowers about 1.5-2 cm. Tuberos roots are round, flattened and stem is heavily twinning type, fleshy and smooth in immature stage and gets harder, woody when it get fully matured ²⁵ .	Thar Desert	Phulwaria <i>et al.</i> [34]
<i>C. attenuata</i> Hook.	Linear or linear to lanceolate leaves, bracteate cyme, produces globose tubers.	Nasik district of Maharashtra	Yadav <i>et al.</i> 2006 [45]
<i>C. anjanerica</i> Malpure, Kamble and Yadav	Erect perennial herb, tuberous rootstock, tubers 2-5cm diameter, stem terete usually branched, leaves opposite decussate, flowers solitary, axillary or extra axillary	Anjaneri hill in Nasik district of Maharashtra	Yadav <i>et al.</i> 2006 [45]
<i>C. concanensi</i> Kambale, Chandore Yadav	Corolla lobes are shorter than corolla tube, corolla tube is abruptly dilated towards base, and outer corona is sparsely hairy	Sindhudurg and Kolhapur districts of Maharashtra	Kambale <i>et al.</i> 2012 [20]
<i>C. hirsuta</i> Weight and Arn.	Perennial twiner, with broad, elliptic to ovate petiolate leaves covered with hairs	Degaon, Maharashtra	Deshmukh and Rathod, 2013 [16]
<i>C. mahabalei</i> Hemadri Ansari	Tricotyledony plant, produces carbohydrates rich tubers	Ralegaon Hills about 10 km West of Junnar, Pune District, Maharashtra	Mishra and Singh, 2001 [30]
<i>C. bhatii</i> Yadav Shendage	Perennial twining herbs, producing axillary or extra axillary cymes with 2-2.5 cm diameter tubers	Malebennur Ghat of Davangere distr., Karnataka	Yadav and Shendage, 2010 [44]
<i>C. noorjahaniae</i> Ansari	Plant is 2.7 cm long, glabrous and attractive corolla	Satara district of Maharashtra	Chavan <i>et al.</i> 2014 [8]
<i>C. woodii</i> Schltr.	Popularly known as rosary vine with pinkish or purplish trailing stems and tubers are more or less similar to potatoes	Kalimpong, West Bengal	
<i>C. omissa</i> H. Huber	Perennial twining herb, stems slender, cylindrical, branched. Leaves are simple, opposite. Inflorescences extra-axillary, cymose, pedunculate.	Tamil Nadu and Kerala	Venu <i>et al.</i> 2017 [43]
<i>C. ravikumariana</i> Kambale Gnanasek.	Perennial herb, stems are prostrate often twining usually branched, glabrous purple to greenish. Leaves simple, opposite-decussate. Inflorescences extra-axillary cyme. Tuberous rootstock.	Tamil Nadu (Tirunelveli district)	Kambale and Gnanasekaran, 2016 [19]
<i>C. metziana</i> Miq	Twinning herb, stems pubescent at nodes, leaves ovate, ovate-oblong, peduncles glabrous or hairy	Kerala (Palakkad, Idukki, Pathanamthitta, Kollam, Thrissur, Wayanad, Kannu districts)	Indian Biodiversity Portal [13]
<i>C. decaisneana</i> Wight	Twinning herb, tuberous roots, leaves linear lanceolate, inflorescence axillary-cyme	Kerala (Idukki, Malappuram, Palakkad, Wayanad districts)	Indian Biodiversity Portal [13]
<i>C. huberi</i> Ansari	Perennial tuberous twiner, stems branched, glabrous. Leaves petiolate; inflorescence lateral or subumbellate cyme	Maharashtra (Kolhapur, Ratnagiri and Satara districts)	
<i>C. jainii</i> Ansari Kulkarni	Erect herb producing tuberous roots; stem is slender producing opposite linear to lanceshaped leaves	-	
<i>C. rollae</i> Hemadri	Erect and Broad leaves, cymose inflorescences, tuberous roots	Western Ghats ranges of Maharashtra	Kamble <i>et al.</i> 2016 [35]
<i>C. oculata</i> Hook.	Tuberous twinning herb, stem terete, unbranched, leaves opposite, Flowers in axillary, peduncled, 2-4-flowered umbellate cymes	Satpuda hill ranges of Nandurbar district, Maharashtra	Punekar <i>et al.</i> , 2006 [36]

5.2. Antioxidant activity

DPPH (1,1-diphenyl-2-picryl hydrazyl) radical scavenging activity, ferric reducing antioxidant power (FRAP) as well as metal chelating ability of major phenolics compounds such as gallic acid, vanillin, catechol and ferulic acids from leaves of *Ceropegia* species such as *C. spiralis*, *C. panchganiensis* and *C. evansii* has been reported [23]. The leaves of *C. bulbosa* showed the highest superoxide dismutase activity [38]. *C. juncea* has possessed various potent secondary metabolites such as tannins, flavonoids and many polyphenolic compounds. Ethanolic leaf extracts of *C. juncea* showed gastroprotective and antioxidant activities in rats due to presence of polyphenolics compounds [43]. *In vivo* tuber extract

of *C. pusilla* demonstrated antioxidant properties with 1, -diphenyl-2-picryl- hydrazyl free radical scavenging assay and reducing power activity, anti-angiogenesis effect was studied by Chicken Egg Chorioallantoic Membrane (CAM) assay. Results showed better DPPH, reducing power activity and inhibited angiogenesis by blocking normal vascularisation [44].

5.3. Anti-urolithiatic activity

The alkaloid 'cerpegin' isolated from *C. bulbosa* was tested for anti-urolithiatic activity and showed maximum dissolution of calcium oxalate and calcium phosphate stones (Monika *et al.* 2012) [31].

5.4. Anti-cancer activity

A comparative studies on *in vivo* and *in vitro* tuber extracts of *C. pusilla* confirmed antiproliferative property against HeLa cancer cell line (Kalimuthu *et al.* 2014) [17]. As well as the three *Ceropegia* species which were *C. spiralis*, *C. juncea* and *C. candelabrum*, screened for anti-cancer activity and confirmed the potent anticancer effect of ethyl acetate fraction of *C. spiralis* against cell lines namely HCT-118 (Colon cancer cell) (Binish and Mary Suja, 2015) [4].

5.5. Toxicological studies

Cytotoxicity effects was demonstrated by three *in vitro* propagated *Ceropegia* species such as *C. spiralis*, *C. juncea* and *C. candelabrum* against HCT-118 Colon cancer cell (Binish and Mary Suja, 2015) [4]. Different stages of cytotoxicity such as cell rounding, aggregation, shrinkage and cell death depending on the concentration of the *in vitro* and *in vivo* tuber ethanolic extracts of *C. pusilla* has been observed (Kalimuthu *et al.* 2014) [17]. At the concentration of 50µg/ml, there was evidenced increased number of death cells, 85.0%, 82.0% respectively. Acute-toxicity studies has been carried out on Swiss Albino mice by using ethanolic leaf extract of *C. juncea* (administered orally at the dose of 1000, 2000 and 3000 mg/kg body weight to animals), results showed no changes in normal behaviour pattern and no sign of toxicity and mortality. It was confirmed that the animals were safe up to a maximum dose of 3000 mg/kg body weight (Paras *et al.* 2011) [33] and total alkaloidal fraction of *C. juncea* was devoid of side effects as noted out by the sub-acute toxicity studies (Adibatti *et al.* 1991) [1].

6. Conclusion

The *Ceropegia* species are being good sources of diverse physiological bioactive compounds such as glycosides, alkaloids, flavonoids, tannins, saponins, triterpenes, phenol, and steroids with wide range of therapeutically active properties *viz.*, anti-inflammatory, anti-oxidant, anti-microbial, anti-cancer and anti-urolithiatic. The pharmacological importance of the genus *Ceropegia* is mainly due to presence of 'cerpegin', which is a rare naturally occurring pyridinone alkaloid. The isolation and purification of potent bioactive principle 'cerpegin' can aid in cancer treatment and also it acts as a potent "therapeutic agent". This review highlights the indigenous and traditional uses of *Ceropegia* species by different tribal groups in India such as Bhillia Tribe, Kani Tribe and Paliyan/Paliyar Tribe for curing of different ailments and also this crop offers opportunities for development of wide range of life saving medicines, value-added products, nutraceuticals and food applications to enhance various health benefits.

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