



Anti-Cancer Activity of Ophiorrhiza Species Endemic to Southern Western Ghats: A Review

Abdul Jaleel.K¹ Malarkodi Velraj^{2*}

¹College of Pharmaceutical Sciences, Government Medical College, Kozhikode, Kerala, India

²School of Pharmaceutical Sciences, VELS Institute of Science, Technology & Advanced Studies (VISTAS), Pallavaram, Chennai, Tamil Nadu, India

Abstract

Cancer is still a challenge to medical science. The enormous human suffering and pain necessitate more efforts to find better treatment possibilities. Camptothecin (CPT) and its analogues are highly promising alkaloids in cancer treatment. Plants in genus *Ophiorrhiza* shows the presence of significant amount of camptothecin. *Ophiorrhiza mungos* is traditionally used in anticancer treatment in Ayurveda. Anthraquinone fraction isolated from in vitro cultures of *Ophiorrhiza rugosa* var *decumbens* also shown antitumor activity. Presence of Forty seven species and nine varieties of *Ophiorrhiza* reported in Indian subcontinent. Out of this, sixteen species and three varieties are reported from Southern Western Ghats. Most of these species are very rare and endemic to Southern Western Ghats. Relevant research studies in these plants are reviewed in this article.

Keywords: Cancer, Medicinal plants, Camptothecin, *Ophiorrhiza* species

INTRODUCTION

For time immemorial, plant materials have been used in treatment of human ailments. Extensive research has undertaken in plant kingdom to discover effective drug for treatment of various cancer diseases. Although the number of successful plant drugs are very less, are very effective and commonly used in chemotherapy of cancer. Over 60 percent of drugs employed in current cancer chemotherapy are derived from plants and microorganism. A good cancer chemotherapeutic drug should kill or incapacitate cancer cells without causing high damage to normal dividing cells. Cancer cells are characterized by uncontrolled proliferation due to altered cell signaling pathways.

Very few drugs currently being used for the treatment of cancer were discovered on the basis of rational structural design. Most of the drugs have been discovered as a result of empiricism, serendipity, or large-scale evaluation (screening) programs[1]. Discovery of vincristine and vinblastine in 1950s was a turning point followed by discovery of podophyllotoxins. United states national cancer institute conducted an extensive plant screening program in 1960 which led to discovery of taxanes and camptothecins[2]. In the cancer drug discovery program, a paradigm based on ethnobotanical and ethnopharmacological data would be more economical and beneficial for identifying potential anticancer molecules than mass screening of plant species.

Monoterpene indole alkaloid camptothecine(CPT) is a potent anticancer agent first isolated and reported from extracts of Chinese tree *Camptotheca acuminata* (Nyssaceae) by M E Wall and M C Wani in 1958 [3]. Thereafter camptothecins were isolated from plants of different families mainly *Ophiorrhiza mungos* and *Nothapodytes nimmoniana*. Due to very low solubility of camptothecin, water soluble sodium salt were used in initial trials which caused unpredictable toxicity and suspension of the trials. Limited solubility of camptothecin prompted search for more water soluble natural derivatives resulted in isolation of 10-hydroxy-camptothecin and 9-Methoxy

camptothecin[4]. Water soluble semisynthetic camptothecin analogues with an intact lactone ring such as Irinotecan (Campostar) and Topotecan (Hycantin) are drugs currently used in ovarian and colorectal carcinoma. Other semi synthetic derivatives of camptothecins are under clinical trial are Chimmitecan, Exaletacan mysylate, Cosetican, Gimatecan etc. Manufacturing of camptothecin based drugs mostly depend on natural sources. The estimated turnover of camptothecin derivatives in 2004 was one thousand million US dollars [5].

The genus *Ophiorrhiza* is a potent natural source for camptothecin. Presence of forty seven species and nine varieties of *Ophiorrhiza* reported in Indian subcontinent [6]. Out of this, sixteen species and three varieties are reported from Southern Western Ghats [7]. Roots of *Ophiorrhiza mungos* is traditionally used for cancer treatment in Ayurveda[8]. Five species of genus *Ophiorrhiza* endemic to Southern Western Ghats of Kerala are reviewed. Plants selected for this study are *Ophiorrhiza mungos*, *Ophiorrhiza rugosa*, *Ophiorrhiza eriantha*, *Ophiorrhiza trichocarpon* and *Ophiorrhiza shendurunii*.

OPHIORRHIZA MANGOS

Ophiorrhiza mangos belong to family Rubiaceae, commonly known as Mongoose plant. Vernacular names are Sarhati (Hindi), Keerippundu (Tamil), Avilpori (Malayalam). Plant is a suffretescent herb, 45-60 meter height. Leaves are elliptic-lanceolate, acuminate; long attenuate at base, Flowers white, in sub-umbellate cymes. Fruits compressed, coriaceous, seeds numerous angular, pale brown.

In Ayurveda Roots of the plant is used for treatment of cancer, useful in snake poison, leprosy etc[8]. Roots are useful in the treatment of cancer[9]. Roots are considered as remedy against bites of venomous snakes, mad dogs etc[10]. Alkaloids camptothecin and 10-methoxy camptothecin isolated from root extracts [11]. Leaves shows the presence of Luteolin-7-O-Glucoside [12].

Tafur S et al studied antiviral and anticancer activity of roots of *Ophiorrhiza mungos*[11]. P Antonisamy et al studied gastroprotective efficacy and mechanism of luteolin-o-glucoside isolated from *Ophiorrhiza mungos* on different ulcer models using rats and concluded that luteolin-o-glucoside is effective for indomethacin and ethanol induced gastric injury in rats[13].Basker et al studied the cancer chemoprotective potential of Luteolin-7-O-glucoside isolated from the leaves of *Ophiorrhiza mungos* against for different cell lines(COLO 320 DM,AGS,MCF-7 and A549) and the result of the study shows that Luteolin-O-glucoside can be used as a potent anticancer drug for colon carcinogenesis[12]. Anaswara KS et al studied anti-Snake venom activity of water and reported that aqueous root have potential to neutralize the viper venom induced lethality and hemorrhage in chick embryo model [14].Napagoda M T et al reported 70% methanol extract of leaves shown excellent protective effect on skin from UV rays, have displayed SPF value greater than 25 [15].

Since it is a good natural source for camptothecin and its endemic nature, many tissue culture studies are done to develop methods for commercial production of secondary metabolites. A.G.Namdeo et al reported a method for micropropagation and production of camptothecin from in vitro plants of *Ophiorrhiza mungos* and established a protocol for rapid proliferation of roots and organogenesis [16].HPLC analysis in the above study shows that production of camptothecin in vitro is higher than naturally grown plants. Deepthi.S and Satheeshkumar K reported significant elicitation of camptothecin production by using jasmonic acid in cell suspension culture [17]. Jose B et al established in vitro mass multiplication of plants through seedling (shoot) cultures of *Ophiorrhiza mungos*[18]. Deepthi S et al studied the effect of major nutrients, growth regulators and inoculum size on enhanced growth and camptothecin production in adventitious root cultures of *Ophiorrhiza mungos* [19]. Deepthi S et al studied influence of Silver nitrate and Yeast extract on cell growth, camptothecin accumulation and cell viability and reported a significant increase in biomass and camptothecin production [20].

Ophiorrhiza mungos var. *angustifolia* is a variety of *Ophiorrhiza mungos*, Rajan et al performed a HPTLC densitometry study and demonstrated the presence of high amount of camptothecin in this plant compared to other species of *Ophiorrhiza*[4]. Krishnan JJ et al developed a method for in vitro mass multiplication of *Ophiorrhiza mungos* L.var.*angustifolia* [21].

OPHIORRHIZA RUGOSA

Ophiorrhiza rugosa is commonly known as wrinkled-leaf snake root or dwarf *Ophiorrhiza*. Vernacular names includes Jari ful, Kalashona (Bengali). Plant is a Perennial herb and some times it is annual. It is a weak plant up to 2 feet tall. Leaves have nearly equal pairs and leaf stalk is 0.5 to 1.5cm. Stems are pilosulous to velvety. Flowers are tubular-funnel shaped. Corolla 6-9mm, white or pale pink and is hairy inside [22]. Two varieties of the species are *Ophiorrhiza rugosa* var.*prostrata* and *Ophiorrhiza rugosa* var.*decumbens*.

OPHIORRHIZA RUGOSA VAR.PROSTRATA

Ophiorrhiza rugosa var.*prostrata* is a dicot procumbent herb grows up to 20-30cm high, Leaves ovate, obtusely acute at apex, rounded at base, petiole 1-2 cm long, stipule triangular, hairy. Cymes 1-2 cm across, congested, peduncle 5 cm long. Flowers minute, white; calyx 1 mm long, lobes ovate; corolla 3 mm long, white. Capsule 2.5 mm across, compressed; seeds 3-5, rugose, folded [7].

Gharpure G et al studied the variation of camptothecin content in root, stem, young leaves, old leaves and fruits of *Ophiorrhiza rugosa* var.*prostrata* [5].Quantification of camptothecin is done by RP-HPLC and found that roots contain significantly high level of camptothecin than stem and leaves. They also studied the seasonal variation of camptothecin content. Quantification of camptothecin content by HPTLC densitometry method was performed by Rajan et al [4].

OPHIORRHIZA RUGOSA VAR.DECUMBENS

Ophiorrhiza rugosa is a herb, grows about 7-30 cm height, with branched decumbent stem. Leaves opposite, elliptic or narrow elliptic, obtuse or acute at apex, narrowed at base, glabrous above, brownish green beneath with 6-9 lateral nerves on either side. Inflorescence 1-1.5 cm across, peduncles 2.5-4 cm long, puberulous. Flowers 5-7 mm long. Corolla 4.2-6 mm long, infundibuliform, puberulous outside, villous at the middle within. Seeds are glabrous brown [7].

Raveendran et al reported antitumor activities of an anthraquinone fraction isolated from in vitro cultures of *Ophiorrhiza rugosa* var.*decumbens* [23]. Roja G developed a method for micropropagation and production of camptothecin from in vitro plants of *Ophiorrhiza rugosa* var.*decumbens* [24]. Kamble S et al developed an in vitro culture method for production of camptothecin by hairy roots and regenerated transformed shoots of *Ophiorrhiza rugosa* var.*decumbens* [25]. *Agrobacterium rhizogenes* strain LBA9402 were used for the induction of hairy roots for the above study. Gopalakrishnan R et al isolated camptothecin from multiple shoot cultures of *Ophiorrhiza rugosa* var. *decumbens* and demonstrated cytotoxic activity on cell lines [26]. Rajan et al selected the plant for camptothecin quantification by using HPTLC densitometry [4].

OPHIORRHIZA TRICHOCARPOS BLUME

Ophiorrhiza trichocarpos blume is a perennial herb, grows about 15 cm height with erect stem. Leaves ovate, acute at apex and base, glabrous or sericeous above, pubescent on nerves below, pale green on drying with 6-7 lateral nerves. Inflorescence axillary or terminal dichotomously branched cyme, densely flowered, peduncle 1- 3.5 cm long, elongating up to 5.5 cm in fruiting. 6 - 10 mm long white Flowers. Calyx lobes , ovate, acute, pubescent. Corolla , tubular, lobes 1.5 - 1.75 × 1 - 1.25 mm, ovate, acute. Glabrous, irregularly angular seeds. Flowering and fruiting season is from May to October[27].

Ophiorrhiza trichocarpos blume is a new species reported from Southern Western Ghats of India by Sibi C V et al 201[27]. HPTLC based quantification of camptothecin is

studied by Rajan et al [4]. Sibi C V et al demonstrated the subculture of *Ophiorrhiza trichocarpose blume* for prolonged time by alternating media strength in each subculture, resulted in substantial increase in shoot production and biomass[28].

Ophiorrhiza shendurunii

Ophiorrhiza shendurunii shows high degree of endemicity in Southern Western Ghats. Khan AES et al reported this new species of *Ophiorrhiza* in 1998 from Kollam district of Kerala[29]. The specific name *shendurunii* is after the renowned wild life sanctuary Shendurunii. It is an undershrub with erect stem, branched, internodes 7-10 cm long. Leaves 8-12 x 3-5 cm, ovate to elliptic lanceolate and terminal inflorescence. It is an allied species to *Ophiorrhiza eriantha* but differs in having fewer lateral nerves in the leaves, pubescent inflorescence, glabrous bracts and bracteoles, corolla hairy above the base and papillate upwards within and pilose capsules. Flowering and fruiting throughout the year [29].

Phytochemical investigation of hexane and chloroform extracts of *Ophiorrhiza shendurunii* has done and one new pentacyclic triterpenoid fatty acid ester, lupan-20-ol-3(β)-yl hexa decanoate along with 8 compounds (lupan-20-ol-3(β)-yl acetate, Olean-18-en-3(β)-yl hexa decanoate, dotriacontanoic acid, stigmasterol, rubiadin, nonadecanoic acid, palmitic acid and camptothecin) [30]. HPTLC based quantification of camptothecin is studied by Rajan et al [4].

Ophiorrhiza eriantha

Ophiorrhiza eriantha is an erect subshrub, leaves to 12x5 cm, elliptic, apex acute to acuminate, petiole to 3 cm, stipules 0.8 cm, ovate acuminate. Chymes axillary and terminal. Flowers are white [7]. HPTLC based quantification of camptothecin is studied by Rajan et al [4]. Jose P et al developed a method for micropropagation of *Ophiorrhiza eriantha* Wight. through leaf explant cultures [31].

Other endemic species of *Ophiorrhiza* found in Southern Western Ghats like *Ophiorrhiza pectinata*, *Ophiorrhiza barberi*, *Ophiorrhiza caudata*, *Ophiorrhiza grandiflora*, *Ophiorrhiza nairii* are also selected for HPTLC-densitometry study by Rajan R et al[4]. This study reports *Ophiorrhiza barberi*, *Ophiorrhiza caudata*, *Ophiorrhiza nairii* showed zero or non detectable levels of camptothecin. Baiju E.C et al studied in vitro antioxidant and antibacterial activities of *Ophiorrhiza pectinata* and reported significant antioxidant and antibacterial activity in *Ophiorrhiza pectinata*[32].

New species of *Ophiorrhiza* like *Ophiorrhiza jacobii* and *Ophiorrhiza sahyadriensis* are discovered by Hareesh V.S et al from southern western ghats[33]. Anti cancer studies of these new species are yet to be carried out.

CONCLUSION

Ophiorrhiza mungos is traditionally used as anticancer agent in ayurvedic treatment. It is also having many other traditional uses. Most of the plants in Genus *Ophiorrhiza* shown the presence of camptothecin. The quantity of active constituents and secondary metabolites present in plants may vary due to geographical factors, climatic

conditions, time or period of collection of plant, abiotic stress etc. So camptothecin content and other active components in different species of *Ophiorrhiza* may vary according to these factors also. Some plants reviewed in this article are not explored in detail and are promising candidates for further anticancer studies.

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