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A MINI REVIEW ON AN IMPORTANT MEDICINAL PLANT *CENTELLA ASIATICA*

Arpita Roy

M. Tech - Industrial Biotechnology, Department of Biotechnology, Delhi Technological University, New Delhi - 110042, India.

ABSTRACT: In the Ayurvedic system of medicine, *Centella asiatica* (gotu kola) is one of the important rejuvenating herbs for nerve and brain cells and is believed to be capable of increasing intelligence, longevity, and memory. It contains several active constituents, and the most important bioactive compounds are triterpenoid saponins, including asiaticoside, centelloside, madecassoside, and asiatic acid. It possesses anti-leprotic, anti-viral, anti-bacterial, anti-tumor activities. Due to its medicinal importance, this plant is being overexploited and to conserve this plant, it is necessary to micropropagate this plant in a laboratory with high biomass yield.

Keywords: *Centella asiatica*, Asiaticoside, Pharmacological activity, *In-vitro* studies

Correspondence to Author:

Arpita Roy

M. Tech - Industrial Biotechnology, Department of Biotechnology, Delhi Technological University, New Delhi - 110042, India.

E-mail: arbt2014@gmail.com

INTRODUCTION: Medicinal plants are the traditional source of many pharmaceutically important compounds. In recent times, they are utilized by the pharmaceutical companies for the preparation of several formulations. In the present time, there has been an increase in the use of herbal products around the world¹. In the last 20 years, about 28% of new chemical compounds that are launched into the market are accounted for by the natural products². World Health Organisation (WHO) also stated that more than 80% of the world's population relies on herbal medicines³. Importance of medicinal plants is due to the presence of specific chemical compounds that produce a physiological effect on the human body.

These bioactive chemical constituents of plants include saponin, flavonoids, alkaloids, sterols, tannins, phenols⁴. Medicinal plant-based drugs have an advantage over the other drugs because they are simple, effective and offer a broad spectrum of activity. Furthermore, they have very less adverse side effects as compared to chemotherapeutic drugs⁵. India is rich in medicinal plant diversity, and since the ancient times use of drugs of herbal origin is prevalent in the traditional system of medicines such as *Ayurveda* and *Unani*. There are about 426 biomes which comprise of different habitat diversity that gives rise to the richest centers for plant genetic resources in the world⁶. Out of 18,665 flowering species, only about 3000 plants have been used for the various formulations in the classic system of medicines such as *Ayurveda*, *Siddha* and *Unani*⁷.

Centella asiatica is one of the important traditional medicinal plant belonging to family Apiaceae and commonly known as 'Gotu kol,' 'Indian Pennywort' or 'Mandookaparni' in India.

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It is an important perennial medicinal herb found in the tropical and subtropical countries like India, Sri Lanka and Bangladesh. *C. asiatica* contains several triterpenes, saponins like asiaticoside, asiatic acid, sapogenins, madecassic acid, vellarin, adecassoside, glycosides and centelloside⁸.

In India, it grows up to an altitude of 600-1800 meters above the sea level^{9, 10} on moist, clayey or sandy soils forming a dense green carpet. *Centella asiatica* has a glabrous stem and long petiolated fleshy leaves rooting at nodes. Flowers are greenish to pinkish white and borne in dense umbels. Seeds are pumpkin shaped. It spreads by producing new plants on above ground runners. New plants can be separated from parent plant once they have taken roots. Leaves contain a high amount of triterpenoids¹¹. It possesses several important properties like antileprotic, antistress, antifeedant, anti-tuberculosis activities, wound - healing properties^{12, 13}, antibacterial, atherosclerosis, and fungicidal activity¹⁴.

It is used in the treatment of leprosy, wound, cancer, fever, allergies¹⁵, abscesses, asthma, catarrh, convulsions, dysentery, eczema, gonorrhoea, hypertension, bronchitis, headache, jaundice, pleuritis, rheumatism, ulcers, spasms, tuberculosis, urethritis, etc.¹⁶ Leaves of this plant are rich in vitamin B and C. and minerals such as magnesium, potassium, calcium, phosphorus and aluminum¹⁷.

It is also used as a brain tonic and blood purifier¹⁸. *C. asiatica* contains various flavonoids which include quercetin and kaempferol, rutin and naringin¹⁹. Due to its medicinal importance, this plant is overexploited, and there is a decline in the population of the *Centella asiatica*. International Union for Conservation of Nature and Natural resources (IUCN) listed it as threatened plant and endangered species²⁰.

It is considered as a brain tonic. It is also used for the treatment of asthma, bronchitis, elephantiasis, dropsy, gastric catarrh, kidney troubles, leucorrhoea, skin disease and urethritis²¹ with antibacterial, anti-feedent, anti-filarial, anti-stress, anti-tuberculosis activities and wound healing properties^{22, 23}. Anonymous, 1992 reported that *C. asiatica* plants contain the following glycosides: asiaticoside, indocentelloside, brahminoside,

brahmoside, thankuniside, and isothankuniside. Asiaticoside is useful in the treatment of leprosy and tuberculosis. The plant shows good therapeutic effects on peptic ulcers.

Chemical Constituent: *Centella asiatica* contains various chemical constituents which includes triterpenoid saponins (asiaticoside, masecassoside, asiatic acid and medecassic acid)^{24, 25}, polyacetylenes²⁶, flavones²⁷, phytosterols and lipid²⁸ alkaloids, volatile oils, etc.²⁹ Bosse *et al.*, (1979)³⁰ reported the amount of four active compounds which are asiatic acid (29-30%), madecassic acid (29-30%), madecassoside (1%) and asiaticoside (40%). They are the biologically active compounds and have potential to be promoted as commercial products (Indu Bala and Ng, 1999). In addition they also contain total phenolics about 23000mg/100gm^{31, 32}.

Saponins: Various saponins have been isolated from this plant which includes Asiaticoside, Madecassoside, Brahmoside, Centelloside, Thakuniside, etc.³³

Asiaticoside: Asiaticoside was first isolated from leaves of *Centella asiatica* more than fifty six years ago by Polonoski (1951)³⁴. It is one of the principle terpenoids of this plant; it acts as antibacterial and fungicidal agents against pathogens and fungi (Hausen, 1993)³⁵. It helps in collagen I synthesis in human (Bonte F, Dumas M, Chaudagne C and Maybeck A (1994)³⁶, Influence of Asiatic acid, madecassic acid and asiaticoside on human collagen I synthesis, Planta Med.

It is clinically used as a wound healing agent in combination with madecassic and asiatic acids (Hausen, 1993)³⁷.

Asiatic Acid: It is a pentacycluc triterpene compounds and the aglycone of asiaticoside. It exhibits bioactive efficacy³⁸ and also known to control cell division in human melanoma, heptoma cells and cytotoxic activity on fibroblast cells³⁹. It shows protective activity against UV induced photoaging, wound healing, induces cell cycle arrest, and anti-proliferative effects on human breast, gastric, and urine cancer cells⁴⁰.

Madecassoside: It is a glycoside that acts as a strong anti-inflammatory agent⁴¹.

Madecassic Acid: Its wound healing property has been attributed to its ability to stimulate collagen synthesis⁴².

Triterpenic Acids: Several pentacyclic triterpenic acids have been isolated and characterized from this plant. They occur either in a free state or as aglycones of the naturally occurring saponins. *e.g.*, asiatic acid, madasiatic acid, brahmie acid, isobrahmic acid, thankunic acid, betulic acid, centoic acid, centellic acid, 6b-Hydroxiasiatric acid & terminolic acid⁴³.

Phytosterols: The plant is reported to possess Stigmasterol, Campesterol, Beta-sitosterol, and stigmasterol-b-D-glucopyranoside⁴⁴. Chowdhary *et al.*, (2014)⁴⁵ reported the estimation of stigmasterol, and it is a useful substance in the medication of Alzheimer's disease. The stigmasterol obtained from best source is 0.0582%.

In a study, a patent published related to the role of stigmasterol in the treatment of Alzheimer's disease. In this invention, the use of stigmasterol is for the treatment of amyloidosis of beta-amyloid peptide and Alzheimer's diseases⁴⁶.

Thus the study of stigmasterol content in *Centella asiatica* is essential. Phytosterols have the ability to reduce cholesterol levels, and this was first demonstrated in humans in 1953^{47, 48}. Phytosterols were subsequently marketed as a pharmaceutical under the name Cytellin as to treat the elevated cholesterol^{49, 50} reported that phytosterols have potential to inhibit stomach, lung, breast and ovarian cancers. Phytosterols also have the potential to reduce the elevated triglyceride levels, which is a risk factor for cardiovascular diseases⁵¹.

It was found that the level of triglycerides reduced by 14% by supplementing 1.6 g/day of plant sterols in a fermented milk beverage for six weeks⁵². The proposed mechanism behind the triglyceride-lowering effect of phytosterols is the reduction in triglyceride-rich very low-density lipoprotein particles produced by liver⁵³.

Nitrogen Containing Constituents: An alkaloid hydrocotylin, C₂₂H₃₃O₈N, melting point 110-12 °C, has been isolated from this plant with 0.0016% yield. The plant also yields glycine, aspartic acid, glutamic acid, alanine and phenylalanine⁵⁴.

Flavonoids: It was found that leaves contain 3-glucosyl quercetin, 3-glucosylkaempferol and 7-glucosyl kaempferol⁵⁵.

Pharmacological Activities:

Wound Healing: Titrated extract of *Centella asiatica* which consist of a mixture of three triterpenes (asiaticoside, asiatic acid, and madecassic acid) stimulates glycosaminoglycan and collagen synthesis in rats⁵⁶. Asiaticoside and asiatic acid were more active than madecassic acid in wound healing; thus, it appears to be an effective treatment of wound healing disturbances⁵⁷.

Central Nervous System: Mook-Jung *et al.*, (1999)⁵⁸ reported that asiaticoside derivatives reduce or inhibit H₂O₂ induced cell death and lower the intracellular free radical concentration and protect against the effects of beta-amyloid neurotoxicity. *Centella asiatica* extract was found to increase brain GABA levels⁵⁹.

Memory Enhancing: Aqueous extract of *Centella asiatica* showed a significant effect in memory enhancement. This positive effect is due to the presence of brahminoside, brahmie acid, and brahmoside in plant^{60, 61}. In a study, different doses of fresh leaf juice of *Centella asiatica* were given to seven-day-old neonatal rats for different time periods. These rats were then subjected to the spatial learning and passive avoidance tests along with the age-matched normal and saline control rats. Results showed that there was an improvement in spatial learning performance and enhanced memory retention in neonatal rats treated with higher doses. These results indicate that fresh leaf juice of *Centella asiatica* enhances memory retention⁶².

Antibacterial: Anonymous, 1988 reported that asiaticoside was an active agent against *Bacillus leprae*, *Mycobacterium tuberculosis*, and *Entamoeba histolytica*. Methanolic extract of *Centella asiatica* showed inhibition zone against *V. alginolyticus*, *V. vulnificus*, and *Streptococcus sp.*⁶². Sankar *et al.*, (2010)⁶³ reported that methanolic extract of *Centella asiatica* showed antibacterial activity against three Vibrio species that are *V. harveyi*, *V. alginolyticus*, and *V. parahaemolyticus* but acetone, chloroform and hexane extract was not showed antibacterial activity against these species.

Antioxidant: In a study, it was reported that asiaticoside significantly increased the levels of catalase, superoxide dismutase, glutathione peroxidase, ascorbic acid and vitamin E in excision type cutaneous wounds in rats. The level of antioxidant activity was highest during the initial stages of treatment⁶⁴.

Hepatitis: It was found that titrated extract of *Centella asiatica* helps in the improvement in chronic hepatic disorders⁶⁵.

Cardiovascular: Montecchchio *et al.*, (1991)⁶⁶ reported that three-week treatment of triterpene fraction of *Centella asiatica* in clients with the post-phlebotic syndrome reduced the number of circulating endothelial cells as compared to the normal one. Cesarone *et al.*, (1992)⁶⁷ reported that in a clinical trial *Centella asiatica* extract found to be efficacious in the treatment of reducing ankle, venous insufficiency, foot swelling, edema, improving capillary filtration rate and microcirculatory parameters.

Neuroprotective Effects: Ramanathan *et al.*, (2007)⁶⁸ reported that *Centella asiatica* extract protects monosodium glutamate-induced neurodegeneration. Water extract of *Centella asiatica* showed neuroprotective efficacy against 3-nitropropionic acid-induced oxidative stress in brain of prepubertal mice enhanced glutathione levels, antioxidant defenses in brain regions^{69,70}.

Anti-diabetic: Chauhan *et al.*, (2010) reported that triterpenic fraction of *Centella asiatica* is useful in diabetic microangiopathy by improving the microcirculation and decreasing the capillary permeability. Also, triterpenoid fraction of *Centella asiatica* protects against the deterioration. Methanolic and ethanolic extracts had shown significant protection and lowered blood glucose levels to normal glucose levels in tolerance test.

In-vitro Studies: In recent years, there has been an increased interest in *in-vitro* culture techniques which offer a viable tool for mass multiplication and germplasm conservation of rare, endangered and threatened medicinal plants. Therefore, it is important to develop an efficient micropropagation technique for *C. asiatica* to rapidly disseminate superior clones once they are identified. Tissue culture techniques can play an important role in the

clonal propagation of elite clones and germplasm conservation of *C. asiatica*. Shoot regeneration from leaf derived callus and stem segments of *C. Asiatica* were reported. All parts of the plant have been used as the explant source. Nodal segments of mature plants have been however used in most cases.

Srivastava and Iqbal, (1999), reported use of nodal segments in *Ammi majus* L., nodal explants of *B. monnieri* were propagated *in-vitro* using liquid shake cultures nodal explants were also used for *Eclipta alba* shoot tip, nodal and internodal segments were reported in *Phyllanthus amarus*. Stem and leaf explants of greenhouse grown plants were used for the regeneration from callus cultures of *Centella asiatica*. Banerjee *et al.*, (1999), used 5-6-month-old glasshouse-grown plants of *Centella* for *in-vitro* multiplication from leaf explants. Tiwari *et al.*, (2000) have reported micropropagation of *Centella* using nodal segments. Explants were collected from natural stands where it grows luxuriously around the marshy fringes of a pond.

Full strength Murashige & Skoog medium (1962) has been used for most of the herbaceous species. Multiple shoots were obtained from shoot tips (1-2 cm) derived from field-grown plants of *Bacopa monnieri* in Murashige and Skoog medium supplemented with 0.5 mg/l BAP within 6 days of culture, whereas in the case of *Paederia foetida* and *Centella asiatica* multiple shoots were obtained from field-grown plants in MS medium supplemented with 1.0 mg/l BAP within 7 days of culture.

Supplementation of Plant Growth Regulators such as 0.3 mg / l BAP and 0.2 mg / l kinetin have been found to show a good response of shoot proliferation in *Withania somnifera* with the regeneration of 85%. Full strength MS media with growth regulators such as 0.5 mg/l of BAP in combination with 0.01 mg/l NAA has been reported to give optimum results in *Utleria salicifolia*. Banerjee *et al.*, (1999) reported that in *Centella* initial sprouting required the presence of BAP (2 mg/l) and IBA (0.1 mg/l), however for multiple shoot induction a higher concentration of BAP (3.0 mg/l) and a lower concentration of NAA (0.05 mg/l) is required.

Tiwari *et al.*, (2000) reported that the bud break was dependent on BAP, the synergistic combination of 22.2mM BAP and 2.68mM NAA gave optimum result for the shoot number *i.e.* 4 to 5 shoots per node as well as for optimum frequency, *i.e.* 91%. Das *et al.*, (2008) reported that MS media fortified with 4.0 mg/l BAP + 0.1 mg/l NAA showed average 10.2 ± 0.38 shoots per explants.

Status of *Centella asiatica*: The annual requirement of *Centella asiatica* was around 12,700 tonnes of dry biomass valued at rupees 1.5 billion. National Medicinal Plants Board, Government of India, has projected a combined demand of Centella and Brahmi of 6621.8 MT with an annual growth rate of 20.1% till the year 2004-05. This requirement is rising sharply given the popularity of mandukaparni-based drugs. Because of the large scale and unrestricted exploitation of this natural resource to meet ever increasing demand by the Indian pharmaceutical industry which is coupled with the limited cultivation and insufficient attempts for its replenishments, the wild stock of this medicinal important plant species has been markedly depleted and it is listed as a threatened species by International Union for Conservation of Nature and National Resources an endangered species. Further, there is some evidence of genotypic differences in the glycoside content and medicinal properties.

Side Effects and Toxicity: Alcoholic extracts of *Centella asiatica* have shown no toxicity at doses of 350 mg/kg when given to rats. Reported adverse effects include GI upset and nausea. Topical use of the extract has led to reports of rash. Three cases of jaundice with elevated liver enzymes were reported in Argentina following dosing of Centella. Patients had taken Centella (standardization and dose unknown) for 20-60 days and recovered on discontinuation of the herb.

CONCLUSION: *Centella asiatica* has been used for many years for the treatment of various diseases. A significant amount of work has been done on its pharmacological activity and possible applications of chemical compounds from the whole part of the plant. The present review shows that *Centella asiatica* contains several different phytochemicals like saponins, glycosides,

phytosterols, flavonoids, *etc.* Out of these phytochemicals saponins, *i.e.* asiaticoside is an important one who is responsible for the different pharmacological activity.

It was found that asiaticoside is the main phytochemical which helps in the wound healing activity. *Centella asiatica* also helps in the improvement of neurodegenerative disorder, *i.e.* Alzheimer's as it improves the memory. So it can be utilized as a drug for the treatment of neurodegenerative disorders. *In-vitro* propagation shows that the plant has maximum growth in MS media with different concentration of auxins and cytokinins. Most commonly used cytokinin is BAP (1-4 mg/l) for the shooting multiplication. In future micropropagation and cryopreservation can be used for the conservation of the plant.

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