**Centella asiatica: The paragon of medicinal values under threat**

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**Abstract**

*Centella asiatica* is well known medicinal plants in herbal trades of India. It is commonly known as Brahmi, Gotu kola, mentioned as one of the oldest herb in medicinal systems. The diverse range of pharmacological properties validates by scientific studies with relevant to ethnomedicinal uses of Centella. The plant shows its potential as respiratory disorders, hepatic, spasmodic, cardiovascular and especially neural disorders. The literature on chemical constituents in every parts from root to leaves can prove more valuable pharmaceuticals for human health. Rapid advancement in the era of biotechnology with such type of herbs as trouser horse and create boon to pharmaceutical-industry. Various ethnomedicinal reports on local and traditional home remedies, proliferation of books, theories of medicinal practitioners were reported every years validates by documentation and research every year. In this review, approach was made to document the pharmacological and pharmaceutical uses. Numerous health care products using Centella formulations are already available in the market but always chance of new drug derivatives from a plant using phytochemicals will be there.

**Keywords:** perennial, pharmacology, phytochemicals, neuroprotectant

**Introduction**

The treatment using plant parts based upon traditional herbal formulation was well known all over the world. The passage of this knowledge from generation to generation via medicinal practitioners, amchis, vaids, saints using spiritual thoughts, devasising myths, sacred methodologies draws attentions of ethnobotanist. The claims of these community were well documented from ancient documents and literature confined in Charaka Samhita, Sushruta Samhita written more than 2000 years ago. Several reports from Vedic periods considered this *jalbrahmias* miracle elixirs of life mentioned in Ayurveda, homeopathic, siddha and amachis tradition of India [1].

The *Centella asiatica* is a frost-tender perennial plant in commonly known as Tiger grass, Indian panniywot or Hydrocotyl asiaticae. It is belonging to family Umbelliferae or Apiaceae. It is dominated in Asian countries but its distribution is also abundant in Mexico and eastern South America. The pioneer herb has a long medical history and used from Vedic periods and also used by Aryans in middle of second millennium for the treatment of specifically mental disorders. In south region, the elephants are fed upon it to increase the longevity among beast. The function of aerial part of *C. asiatica* explained in different Indian medicine systems The well fertile loosely arranged sandy loam soil preferred the best growth of plants. Many spiritual historical methodologies are connected to the plant. It is also known as ‘Brahmi’ means wisdom goddess. It is also known as ageless herb because it shows remarkable properties of skin rejuvenations and wound healing. The species composition prefers the present in plentifully humid, grassland and marshy regions adjoining to cultivated fields. The cultivation is well supported by monsoon periods but having proper drainage systems. The knowledge of allopathic medicine is necessary but the escalating faith in local herbal medicines increases having fewer side effects.

Although efficiency and versatility of this herb will always be unreachable and unpredictable whereas scientific documentation continues work in such direction. The momentous shifts in exploring the therapeutic potential via pharmaceutical industries based on clinical trials, mechanism of action based research, along with safety issues in term of informations. The present paper highlights the therapeutic uses, chemical constituents, pharmaceutical uses, conservation status and threat using standard and reliable research published on CAM-PubMed, Natural Medicine Comprehensive Database (NMCD), NCBI and Pubmed.

**Botanical description**

The herbaceous herb is small, prostrate and creeping stoloniferous flexible stem, the trailing herb having two different types of structures at nodes, the dorsal side of stem node occupied by pink glabrous structures and the ventral portion of nodes consists of tiny rooted structures. Leaves are fleshy, reniform to orbicular and dentate in shape, arranges in clusters of 2 or 3 attached to main stem by a long 2-10cm rounded pedicels that lacks structural modification on surface. The leaves shape is resembling with the frog back so the plant named as mandukparni in Sanskrit language. The flowers are sessile and ovoid in shape. The runners lie along the ground and long leaves with their scoloped edges rise above on long reddish petioles. The insignificant greenish- to pinkish-white flowers are borne in dense umbels on separate stems in the summer [2]. The plant is slightly pungent but lack aromaticity, whereas some species grows is fallow land lack grazing activities having more pungent leaves. The whole plant parts provide medicinal value and trending in properties of high blood pressure, wound healing, leprosy, CNS, utero-relaxant actions, depression, memory enhancement, emotional disorders, promoting longevity,
revitalizing of nerves and brain cells in relevant to mind disorders [3].

Chemical compounds
Start from the differences among geography, climate, varietal, extraction and analysis method favored the difference in the phytochemicals composition. *Centella asiatica* has more than 70 chemicals including terpenes, alkaloids, flavanoids, essential oils, amino acids, minerals, carbohydrates and vitamins.

1. Triterpenes and their glycosides
A number of glycosides have been reported from *Centella asiatica* including Asiaticoside, Medecassoside, Brahmoside, Centelloside and Thakuniside. The important biological compounds are Asiatic acid, Asiatricoside, Medacassic acid and Medecassoside. These triterpenes are the important biomarkers component for quality assessment. Asiatricoside along with Medacassic acid and Asiatic acids used as wound healing agent by stimulating the collagen I synthesis in human. Several pentacyclictriterpenic acids are found as free or aglycones as naturally occurring saponins such as Asiatic acid, Betulic acid, Brahmic acid, centelic acid, centoic acid, isobrahmic acid, 6b-Hydroxiasiatic acid, madiastic acid and terminolic acid [5, 6].

2. Flavonoids
A potential source of flavonoids include apigenin, catechin, farnesol, kaempherol, quercitin, naringin, rutin, 3-glucosylquercetin, 3-glucosylkaepferol and 7-glucosylkaempferol. Kaempherol and Quercitin glycosides and astragalin are the flavones derivatives observed in the *C. asiatica*.[7,8]

3. Volatile oils
Caryophyllene, elemene and famesol, X-humulene, germacrene-D, bicycloelemene are the abundant essential oils observed in the plant. Monoterpenoid hydrocarbons, oxygenated terpenoids, sesquerpenoid and oxygenated sesquerpenoids and sulphidesesquerpenoids are the essential oil observed in the experimental study.

4. Phytosterol
Among phytosterols including beta-sitosterol, campesterol, stigmasterol and sterol-b-D-glucopyranoside are reported in this plant help to reduce the breast, ovarian, stomach and lung cancers.

5. Other Phytocstituents
It is quite rich in Vitamin A, Vitamin B1, Vitamin B2, Vitamin C, niacin and carotene. Moreover ash has calcium, chloride, iron, magnesium, phosphate, potassium and sulphate. Alanine, glutamate and serine are the most frequent amino acids besides the other 17 amino acids have been observed in the various parts of *C. asiatica* leaves have volatile fatty acids such as lignoceric, linoleic, linolenic, oleic, palmitic and steric acids and hydrocetyl.

Traditional Uses
*C. asiatica* is valuable plant known from very prehistoric time for its immense medicinal potential. It is considered as valuable traditional plant practiced in diverse traditional systems such as Ayurvedic, Unani and folk remedies. German author establishes pioneer systematic documentation of *C. asiatica* and provided evidences for the uses from centuries in South East Asia and India [9].

Medicinal Uses
1. Cognitive treatment
The extract of *C.asiatica* was help to improve dentitytarborization, differentiation of synopsis and cognitive treatment. Asiatic acid is effective in supplement for proper functioning of stroke and restore synapatial memory and cell proliferation. It is also effective in cognitive associated disorders such as oxidative stress and mitochondria dysfunction. The extract of *C. asiatica* and effect of acute administratation on rats for memory improvement and 30 mg/kg significantly elevate learning ability. The asiaticoside maintains calcium homeostasis, enhance free radical scavenging and decrease Ache inhibitory activity. It enhances the calcium homeostasis, mitochondorial regeneration and activity of genes responsible for antioxidant production. In an experiment conducted with the plant extract shows that increase the differentiation of synaptic, impairment of neural cell differentiation and dendritic arborization help to cope up with memory syndrome and reduce the neurological defects associated with hippocampal regions. In rat the plant aqueous extract was found to enhance activity of cyclic AMP response element binding protein phosphorylation level was enhanced in primary culture of rat embryonic cortical cells be benefited in Alzheimer disease and significantly memory improvement.

2. Antimicrobial
The *Casia* extract exhibits various antimicrobial activities and inhibitory effect at very mild concentration [10]. In an experiment conducted on two different strains of gram positive staphylococcus bacteria observed when essential oils given at different ranges. The *Bacillus cereus* and *Listeria monocytogenes* two negative strains when observed under different environment, the antibacterial activity of *C. asiatica* is enhanced when tested under stress condition. Methanolic extract of *C. asiatica* leaves shows inhibitory effect on *Escherichia coli*, *Staphylococcus aureus* and *Staphylococcus aureus*. The high dose of extract increase memory potential and coordinates the event related to neurogenesis [11].

3. Anti-inflammation
The triterpenoids extract containing centaloids of *C. asiatica* used to evaluate the potential in anti-inflammation responses for oncological experiments. The different concentrations of methanolic extract and diclofinac sodium were used but extract shows maximum stability in membrane at specific dose concentration. In another study the extract of *C. asiatica* is effective against EDMA as it regulate the activity of hepatic enzymes glutathione and superoxide dismutase [12, 13].

4. Anti-oxidant
Ageing is the major concern in all organisms initiated with the production of ROS (Reactive oxygen species) and free radicals and overcome by scavenging mechanisms. In Sprague-Dawley rats the plant extract of *C.asiatica* cause detoxify in ROS species by lowering the activity level of superoxide dismutase [14]. In an experiment conducted on comparative evaluation of polyphenols,
between the butylhydroxyanisole (synthetic antioxidant) and *C. asiatica* (natural antioxidant), the result significantly favors the higher effect of herb [15, 16].

5. Wound Healing

The extract analysis containing Asiatic acid, madecassic acid and asiaicoide shows positive effects in wound healing. It enhances the activity of peptidichydroxyproline content cause strengthening of collagen biosynthesis, epithelialization and its tensile strength. The alcoholic extract demonstrate the stability of collagen their cross-linkage, its maturation, aldehyde and acids, increase cellular proliferation, DNA and protein content. The experiment conducted in rat treatment for shows that the faster wound healing evidenced from increasing collagen amount applied orally in gel form. The cell layer fibrinonecin, extracellular matrix formation and fibroblast proliferation appears to be promotes the wound treatment. Moreover, fibroblasts and keratinocytes proved its importance in dermal and skin diseases. An increase in DNA, protein, and collagen content of granulation tissues was observed on supplementation of extract of *C. asiatica* resulting in collagen synthesis and cellular proliferation at wound site (under plagiarm). The extract of *C. asiatica* considerably enhanced the wound breach power in incision model when compared to controls, wound contraction rate was noticeably enhanced as compared to control wounds (p<0.001), and wounds epithelized faster when treated with an extract of CA. Rats treated with extract showed a better tensile strength of wound after 7 days of wound infliction when compared with control. A study on ethanol-induced gastric lesion oral administration of CE (0.05, 0.25, and 0.50 g/kg) before ethanol administration considerably lowered mucosal myeloperoxidase activity checked gastric lesions formation (58% to 82% reduction) and in a dose-dependent [17].

6. Neurotoxic and Memory Booster

The level of dopamine, 5-ht and norepinephrine will be decrease in albino mice brain under in vitro condition. The hydroalcoholic extract of *C. asiatica* was tested and it inhibits the acetylcholine with 50% inhibition rate at dose of 150ug/ml. Experimental finding of *C. asiatica* under various concentrations against tyrosinase, acetycholine shows inhibitory effect. The asiaticosides under phospholipase A provides positive responses against neural disorders. In mice the effect of increasing memorizing ability observed during the doses examined under 200 to 1000 mg/kg. The regeneration of neurons and its dendritic arborization will be pointed out at dose of 6ml/kg for 6 weeks in mice under in vitro condition. It shows the protective effect against oxidative stress induced by mitochondrial dysfunction. Similarly, the *C.asiatica* extract containing Asiatic acid that increase blood permeability in neural cell and effective against cerebral ischemia disorders [18].

7. Hepatoprotective

The effect of methanolic extract of *C.asiatica* was evaluated for hepatic concentrations of interleukin-1β, diabetes mellitus, MCP-1, and tumor necrosis factor alpha in diabetic control. In enhanced fibrosis of liver tissues in rats by mass intralobular degeneration, perportal bridging necrosis and focal necrosis. *Centella asiatica* extracts contains saponins regulate proper liver function in rats by

Restoring the production of cytokine. It shows hepatoprotective activity when checked against Ccl4-induced liver injury probably due to the presence of asiaticoside. Conventionally, Asiatic acid protects liver injuries and used to get rid of hepatic disorders therefore, could be an array of health-care for hepatoprotective [19].

8. Anti-diabetes

The blood glucose level of rat is 32.6%, 38.8%, and 29.9% after the administration of leaves extract of *C. asiatica* of 250, 500, and 1000 mg/kg respectively [20]. Similarly blood glucose level are 29.4%, 32.8%, 33.6%, and 35.7% at dose of 50, 100, 200, and 400 mg/kg respectively after the administration of *C. asiatica* extract [21]. Plant extract allows to lowers the glucose absorption and impedes the effect of alpha amyrase and intestinal disaccharides [22]. Asiatic acid reported to be declines the blood glucose level by promoting islets fibrosis by preventing the islets dysfunction [23]. Moreover it allows the restore the beta cell mass [24].

9. Against cancer

Aqueous extract of *C. asiatica* had inhibitory effect on the cell lines of rat glioma (C6), mouse melanoma (B16F1) and human breast cancer (MDA MB-231) with IC50 values of 698.0, 648.0, and 1000.0 µg per mL respectively [25]. *C. asiatica* methanolic extract impede the MCF-7 cell lines by inducing the apoptosis as causing DNA breaks, nuclear condensation and loss of membrane potential of mitochondria [26]. The concentration of 0.1% of *C. asiatica* juice causes the damage to DNA and programmed cell death of human HepG2 cell lines. Asiatic acid having anti-proliferative activity and thus inhibits the tumor volume and weight in the lung cancer [27]. It also cause apoptosis and decline the viability of melanoma SK-MEL-2 cells. Asiaticoside impedes the mouse lung fibroblast (L-929) cells of B16F10. It declines the 50% viability of ovarian cancer cells with treatment of 40ug/ml Asiatic acid and cell cycle arrest at G0/G1 phase with 7-10 folds increased apoptosis [28].

10. Against depression

*C. asiatica* is showing potential antidepressant activity. In the forced swimming test, total triterpenes and imipramine cause to fall in the stillness duration and regulation of amino acid level. Moreover the serotonin, norepinepherine, dopamine and their derivatives level have been intensified as well declined cortisone level in the serum [29, 30]. A reversed swing in the behavior and physiology in the OBX induced depression has been developed due to the standardized extract administration in rats [31, 32]. Treatment with two composition (asatic acid and asatic + midazolam), Sprague-Dawley rat emitting significantly good open arm time, maximum speed and time in the forced swim test.

11. Skin care

3, 5-O-dicaffeoyl-4-omalonalquinic acid, derivative of caffeoyl has been intensified in vitro in order to nullify the 300-330 nm range of UV light by mean of absorption. Consequently, UV damages to skin, skin ageing and collagen breakdown has been reduced [33]. Study revealing that *C. asiatica* operates on the connective tissues of vascular wall and reduce the capillary filtration to improve the microcirculation to lessen the skin ageing [34]. Asiaticoside targets to promote the adhesion of initial
skin cell, migration of skin cell for wound healing and intensify the human dermal fibroblasts [35].

12. Against ulcer

Asiaticoside in the C. asiatica extract extensively lessens the stress induced ulcers in the rat. Reduction in the gastric ulcer was observed to be dose dependent and coincides with GABA level in dose dependent manner [36]. A study involves the administration of fresh juice of plant significantly provide protection against the ulcer by toughening the defensive factors of mucosa [37]. Inhibition of leucocytes infiltration and protection of gastric walls has been reported in the treated rat with C. asiatica.

13. Against epilepsy

Traditionally herb is used as anticonvulsant and anxiolytic as it increase the GABA cerebral level. C. asiatica extract reduced the PTZ-kindled seizures and improve the learning deficit. The aqueous extract of C. asiatica acts as an adjuvant to anti-epileptic drugs to prevent the cognitive impairment. A study of oral administration of C. asiatica extract against the pentylenetetrazol-induced convulsions, strychnine induced opisthotonus tonic convulsion and pentylenetetrazol-kindled seizures [38]. Moreover, reduction in spontaneous motor activity and lipid peroxidation products along with the potentiation of pentobrbitone sleeping time and diazepam withdrawl-induced hyperactivity has been reported.

Conservation Threat

The modern biotechnological propagation and extraction techniques to C. asiatica can be harnessed for secondary metabolite extraction. According to the reports of Export and Import Bank of India Centella asiatica is one of the important medicinal plants in the International market of medicinal Plant Trade. However, the wild stock of this plant species has been markedly depleted, because of its large scale and unrestricted exploitation coupled with limited cultivation and insufficient attempts for its replacement has been made. Moreover, now it has been listed as Threatened plant species by the International Union for Conservation of Nature and Natural Resources (IUCN), and also as an endangered species [39].

Table 1: Properties and mode of action via usage of Centella asiatica

<table>
<thead>
<tr>
<th>S. no.</th>
<th>Property</th>
<th>Mechanism of action</th>
<th>Study conducted</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>Cognitive treatment</td>
<td>Enhance synaptic differentiation and dendritic arborization</td>
<td>30 mg/kg acute administration on rats for memory improvement</td>
<td>Prakash et al. 2017 [45]</td>
</tr>
<tr>
<td>2.</td>
<td>Anti-inflammation</td>
<td>Inhibition of cyclooxygenase-2, interleukins (IL-6, IL-1β), cytokine tumor necrosis factor</td>
<td>Madecassoside inhibit collagen II-induced arthritis in mice</td>
<td>Sabaragamuwa et al., 2018 [46]</td>
</tr>
<tr>
<td>3.</td>
<td>Anti-oxidant</td>
<td>Reduction of hydroperoxides, inactivation of free radicals, chelation of metal ions</td>
<td>Oral application of crude methanol extract of C. asiatica to lymphoma bearing mice significantly increased the superoxide dismutase (SOD), catalase and glutathione peroxidase (GSHPx)</td>
<td>Roy and Bharadvaja 2017 [47]</td>
</tr>
<tr>
<td>4.</td>
<td>Wound Healing</td>
<td>By increasing migration rate of skin cell, enhancing the initial skin cell adhesion and inducing an increase in the number of normal human dermal fibroblasts</td>
<td>Ethanolic leaf extract of C. asiatica induce wound healing in Wistar albino rats</td>
<td>Kant et al., 2019 [48]</td>
</tr>
<tr>
<td>5.</td>
<td>Memory Booster</td>
<td>increasing the level of norepinephrine, dopamine and 5-HT in the brain</td>
<td>aqueous extract of the leaves of the Centella asiatica revitalize the brain and nervous system thus exhibit significant effect on learning and memory process</td>
<td>Sabaragamuwa et al., 2018 [46]; Chirmoya et al., 2019 [49]</td>
</tr>
<tr>
<td>6.</td>
<td>Hepatoprotective</td>
<td>Enhanced fibrosis in liver tissue</td>
<td>Extract of C. asiatica shows positive result in curing chronic hepatic disorder.</td>
<td>Kant et al., 2019 [48]</td>
</tr>
<tr>
<td>7.</td>
<td>Anti-diabetes</td>
<td>Lower the blood glucose level and fasten the wound recovery</td>
<td>Ethanolic and methanolic extracts of C. asiatica had shown significant protection and lowered the blood glucose levels to normal in glucose tolerance test carried out in the alloxan induced diabetic rats</td>
<td>Roy and Bharadvaja 2017 [47]</td>
</tr>
<tr>
<td>8.</td>
<td>Against cancer</td>
<td>Inducing apoptosis in cancerous cells</td>
<td>oral administration C. asiatica (500 and 1000 mg/kg) exhibits significant decrease in the level of tumor incidence, weight, cumulative number of papilloma</td>
<td>Kant et al., 2019 [48]</td>
</tr>
<tr>
<td>9.</td>
<td>Against depression</td>
<td>Reducing cortisone level</td>
<td>Triterpenoid saponins present in the plant exhibit antidepressant activity by reducing corticosterone level in serum</td>
<td>Gohil et al., 2016 [34]</td>
</tr>
<tr>
<td>10.</td>
<td>Skin care</td>
<td>Stimulate the synthesis of skin aging inhibitor type I collagen in human dermal fibroblasts cells</td>
<td>Aqueous extract as nanoencapsulated form reduced the expression of metalloproteinase in UV radiated cells and inhibited hyaluronidase expression in mouse skin</td>
<td>Zahara et al., 2014 [50]</td>
</tr>
<tr>
<td>11.</td>
<td>Against ulcer</td>
<td>dose dependent reduction of gastric ulceration was associated with a dose dependent increase of the GABA level in the brain</td>
<td>protection of gastric mucosa and inhibition of leucocytes infiltration of gastric wall in rats pre-treated with Centella asiatica extract</td>
<td>Kant et al., 2019 [48]</td>
</tr>
<tr>
<td>12.</td>
<td>Against epilepsy</td>
<td>reduced the PTZ-kindled seizures</td>
<td>oral administration of C. asiatica extract against the pentylenetetrazol-induced convulsions, strychnine induced</td>
<td>Gohil et al., 2017 [34]</td>
</tr>
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<td>S. no.</td>
<td>Region</td>
<td>Medicinal systems</td>
<td>Uses</td>
<td>References</td>
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</tr>
<tr>
<td>1</td>
<td>Indian medicinal system</td>
<td>Sushruta Samhita</td>
<td>Wound healing, leprosy, lupus, varicose ulcers, eczema, psoriasis, diarrhoea, fever, amenorrhea and diseases of female genitourinary tract</td>
<td>Brinkhaus et al., 2000 [9]; Sabaragamuwa et al., 2018 [46]</td>
</tr>
<tr>
<td>2</td>
<td>Java and other Indonesian Islands and Madagascar</td>
<td>Folk medicine</td>
<td>Traditional healers</td>
<td>Brinkhaus et al., 2000 [9]</td>
</tr>
<tr>
<td>3</td>
<td>Island of Mauritius</td>
<td>Folk medicine</td>
<td>Leprosy</td>
<td>Brinkhaus et al., 2000 [9]</td>
</tr>
<tr>
<td>4</td>
<td>Indian Ayurveda</td>
<td>Ayurveda traditional medicine</td>
<td>Mild and chronic diseases</td>
<td>Das, 2011 [52]</td>
</tr>
<tr>
<td>5</td>
<td>Sri Lankan and Indian</td>
<td>Ayurvedic traditional medicine</td>
<td>Skin diseases, syphilis, rheumatism, mental illness, epilepsy, hysteria, dehydration and leprosy</td>
<td>Matsuda, et al., 2001 [8]</td>
</tr>
<tr>
<td>7</td>
<td>Southeast Asian</td>
<td>Traditional</td>
<td>Prompt bladder activity, for physical and mental exhaustion, diarrhoea, eye diseases, inflammation, asthma, and high blood pressure</td>
<td>Matsuda et al., 2001 [8]</td>
</tr>
<tr>
<td>8</td>
<td>Java and Malay peninsula</td>
<td>-</td>
<td>heal wounds and constituent in brain tonics to treat cognitive retardation</td>
<td>Kartnig, 1988 [54]</td>
</tr>
<tr>
<td>9</td>
<td>Europe</td>
<td>-</td>
<td>externally (powder, cream and ointment) and internally (tablet, tonic) used cosmetic, and medicinal preparations</td>
<td>European Medicines Agency, 2010 [53]</td>
</tr>
<tr>
<td>10</td>
<td>North America</td>
<td>-</td>
<td>to treat fever, stomach upset, scrofula (a form of tuberculosis) and syphilis, as an antidote for organophosphorus pesticide-induced toxicity</td>
<td>Bandara et al, 2011 [55]</td>
</tr>
</tbody>
</table>

Table 2: Distribution status, importance and enlisted uses explained in different medicinal system
Fig 1: Diverse role of *C. asiatica* in disease ailments.
**Conclusion**

_Centella asiatica_ has been used as traditional medicines for wide range of diseases from ancient times. The herb is very well recognized for memory reinforcement. The plant is used in multipurpose disease treatment such as dysentery, skin, Rheumatism leprosy, syphilitic ulcer, fever, psoriasis, and bowel complaints, nervous and blood diseases. The Leaves are enriched with ascorbic acid and other secondary metabolites accelerate nervous functioning. It improves mental weakness, intelligence and longevity when given with fenugreek and black pepper in dose specific manner explained in Charaka Samhita. Due to its very less toxic effects, it is popularly used as natural products and act as potential herb in numerous healthcare applications. For conservation of this medicinally important plant population, techniques like tissue culture should be well established. It is not only economically affordable method, it also enhance plant population in very short time period.

**References**