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HEMIDESMUS INDICUS (L.) R. BR.: AN OVERVIEW

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ABSTRACT

Since ancient times plants have been used as the primary source for medicinal purposes because of their bioactive compounds. *Hemidesmus indicus* a twining shrub with laticiferous branches which produce a wide range of bioactive compounds and their plant parts serve as a key component in traditional medicine preparations to treat diseases such as cancer, diabetes, fever, syphilis, leprosy, ulcer, skin disease and also used as a blood purifier, antivenom antidote against snakebites and scorpion stings. other-than medicinal application they are utilized in the production of cosmetics, nutraceuticals, confectionaries, and beverages. Because of these applications *H. indicus* is identified to be the most traded medicinal plant. Nowadays people are started to prefer herbal-based medicines (Ayurveda and Unani) because of no side-effects when compared to allopathy medicines. due to these events, the harvesting of *H. indicus* from their natural habitat is increasing. the plant is found to be grown in uncultivated lands and deciduous forests, so this highly valuable plant species is needed to be conserved and to ensure their protection from illegal harvesting. This review summarizes the studies carried out about the pharmacology, phytochemical composition, ethnomedicinal use, and the application of plant biotechnology techniques for the conservation of *H. indicus*.

Keywords **:** *Hemidesmus indicus*, ethnomedicinal use, conservation, 2-hydroxy-4-methoxybenzaldehyde (MBALD)

Introduction

The extracts obtained from herbal plants serves as a way of a new approach for treating various diseases, this new approach is widely followed in Asian countries like India since ancient times to treat humans and animals. It has been estimated that around 35000 to 70000 variety of plant species has been used for the production of cosmetics, medicines, and nutraceuticals. One such plant that has vast application for the production of cosmetics, perfumery, soft drinks, and baked foods, and also for the preparation of traditional medicines is *Hemidesmus indicus* (Sayyed *et al*., 2014; Kher *et al*., 2020). *Hemidesmus indicus* is also commonly known as Indian sarsaparilla or false sarsaparilla in English and also known by various names in different languages they are: Nannari (Tamil), Nannari, Naruninti, Narunenti, Narunari (Malayalam), Sugandhipala (Telugu) Namdaberu, and Sogadaberu (Kannada), Anantamul (Assamese and Bengali), Upalsari and Anantvel (Gujarati), Upsalan (Marathi), Onotomulo and Suguddimalo in (Oriya), Magrabu and Hindisalsa (Hindi), Anatmula, Dhavalasariva, Gopa, Gopakanya, Gopabandhu, Gopavalli, Gopi, Karala, Krishodari, Lata, Nagajihva, Sariva, Sugandha and Shgandhi (Sanskrit), Salsa (Urdu), Zaiyana (Arabic) Ushbanindi, Yasmine barri, Aushbahenindi (Persian) (George *et al*., 2008; Weissner, 2014; Banerji *et al*., 2017; Nandy *et al*., 2020a). It is a twining semi-erect or prostrate shrub that has laticiferous branches with purplish bark and woody roots that grow in mesophytic to semi-dry conditions, it can be found all over India (George *et al*., 2008; Austin, 2008).The plant was first placed under the family Asclepiadaceae and then transferred to Periplocaceae on basis of the pollinial characteristics and finally, it has been added to the family Apocynaceae on basis of phylogenetic reclassification (Austin, 2008; Banerjee and Ganguly, 2014; Banerji *et al*., 2014; Kher *et al*., 2020). *H. indicus* is a highly valuable multipurpose medicinal plant, that has various medicinal properties, it has been used by the native healers to treat nephritis, syphilis, sore mouth. The plant produces a variety of phytochemicals particularly the root part which is used as antipyretic, anti-diarrhoeal, astringent, blood purifier and also used to treat skin disease, ulcer, rheumatism, and leucorrhoea. The national medicinal plant board (NMPB) India has identified *H. indicus* as a highly traded medicinal plant because of its multipurpose nature (Austin, 2008; Aneja *et al*., 2008; Das and Bisht, 2012; Cheruvathur *et al*., 2013; Weissner, 2014; Kher *et al*., 2020). there are so many substitutes for *H. indicus* which produce similar kind of phytochemicals and has similar applications and medicinal properties are available they are *Cryptolepisbuchanani, Decalepishamiltoni, Ichnocarpus frutescens, Mondiawhitei, Swertia chirata, and Vallaris Solanaceae* (Austin, 2008; Rathi *et al*., 2017; Mishra *et al*., 2017; Nandy *et al*., 2020b).

This review is done to give a quick and brief view about *Hemidesmus indicus*, which includes geographical distribution, economic importance, and conservational need, phytochemical, pharmacological, and ethnobotanical studies

to understand the pharmacological activity of the bioactive compound produced by the plant and their usage by the locals for the treatment of various disease and illness. Due to the multipurpose nature of *H. indicus*, harvesting of the plant is increasing rapidly and that leads to the reduction of their natural population, because of these reasons certain protective measures are needed to be taken for the conservation of *H. indicus*. This review will be hoped to show various aspects of *H. indicus* and their need for conservation and provide sufficient information to the researchers.

1. Geographical Distribution

Hemidesmus indicus grows in the plains and to an altitude of 600m in semi-dry to mesophytic conditions. it is found widely throughout India precisely from Gangetic plains to eastwards of Bengal, Sundarbans, Assam, and different locations of central, western, and southern parts of India. It is usually seen in deciduous forests, uncultivated lands, and hedges. Other than India it is also reported to be found in Sri Lanka, Pakistan, Iran, Bangladesh, Moluccas, and Malaysia (Austin, 2008; George *et al*., 2008; Banerjee and Ganguly, 2014; lalrinpuia *et al*., 2017; S. Nandy *et al*., 2020a; Nandy *et al*., 2020b). (Fig. 1) Shows the geographical distribution of *Hemidesmus indicus*.

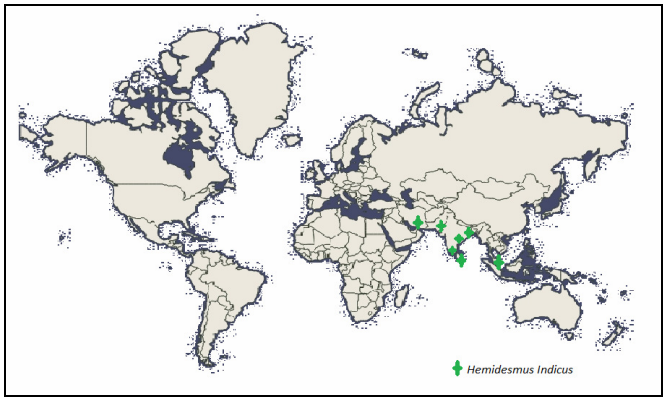

Fig 1. Geographical distribution of *H. indicus*

2. Phytochemical Composition of *H. indicus*

2.1. Root

The chemical constituents of the root are essential oil composed of 80% 2-hydroxy-4-methoxybenzaldehyde (MBALD) and also contains2-hydroxy-4-methoxy acetophenone, 2-hydroxy-4-methoxybenzoic acid, 3-hydroxy-4-methoxybenzaldehyde, 4-hydrobenzaldehyde, 4-hydroxy-3-methoxybenzoic acid, aromadendrene, methyl 2-hydroxy-4-methoxybenzoate, α-muurolol, (E)-nerolidol, α-bisabolol, β-sitosterol,2-methoxyphenol, 3-methoxyphenol, 2-phenylethyl cinnamate, methyl salicylate, (E,Z)-2,6-nonadienal, (E)-2,(Z)-6-decadienal, terpinen-4-ol, Salicylaldehyde, limonene, α-terpinyl acetate,16-dehydropregnenolone, amyl cinnamate, benzophenone, benzyl benzoate, borneol, dihydrocarvyl acetate, camphor, coumarins, coumarinolignoids, Flavonoids, glycosides, hemidesmin-1, hemidesmin-2, hydroquinone, polyphenols, steroids, Tannins, terpenoids,triterpenes, the alkaloids from the root are extracted by hexane, ethyl acetate,ledol, nerolidol,linalyl acetate, isocaryophyllene, lupeol acetate (Chatterjee and Bhattacharya, 1955; Heble and Chadha, 1978; Sreekumar *et al*., 1998; Nagarajan *et al*., 2001; Jirovetz *et al*., 2002; Das *et al*., 2003; Mary *et al*., 2003; Chatterjee *et al*., 2006; Fimognari *et al*., 2011; Kundu *et al*., 2012; Ferruzzi *et al*., 2013; Nagat *et al*., 2016; Turrini *et al*., 2018)

2.2. Stem

3-keto-lup-12-ene-21→28-olide, Δ^{12}-dehydrolupanyl-3β-acetate, Δ^{12}-dehydrolupeol acetate,γ- lactone, desmicine, desinine, emidine, hemidine, hemidescine, hemisine, hexadecenoic acid, indicine, Keto-triterpenoids, lupanone,medidesmine, sitosterol, 2-hydroxy-4-methoxybenzaldehyde, 3-hydroxy-4-methoxybenzaldehyde, 4-hydroxy-3-methoxybenzaldehyde (Oberai *et al*., 1985; Prakash *et al*., 1991; Gupta *et al*., 1992; Chandra *et al*., 1994; Deepak *et al*., 1997)

2.3. Leaf

Alkaloids, flavonoids, glycosides, hyperoside, phenols, rutin, saponins, steroids, sterols, Tannins, terpenoids, triterpenoids, (Subramanian and Nair, 1968; Dhanalakshmi, *et al*., 2018)

2.4. Flower

Flavonol glycosides (isoquercitrin, rutin, hyperoside) (Subramanian and Nair, 1968; Nandy *et al*., 2020)

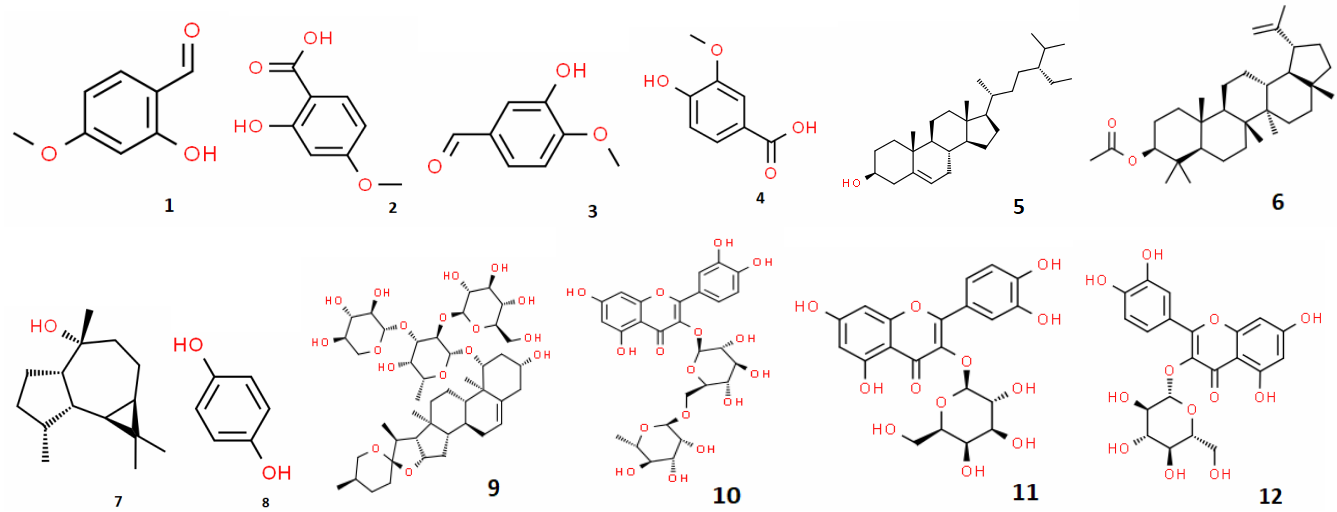

Fig. 2 : Chemical Structure of some phytochemicals found in *Hemidesmus indicus.*

1. 2-Hydroxy-4-Methoxybenzaldehyde; **2.** 2-Hydroxy-4-Methoxybenzoic acid; **3.** 3-Hydroxy-4-Methoxybenzaldehyde; **4.** 4-Hydroxy-3-Methoxybenzoic acid; **5.** β-sitosterol; **6.** Lupeol Acetate; **7.** Ledol; **8.** Hydroquinone; **9.** Saponin; **10.** Rutin; **11.**Hyperoside; **12.** Isoquercetrin.

3. Pharmacological Studies

Hemidesmus indicus possess various pharmacological activity because of its diverse phytochemical nature, so it has a long history for medicinal usage in the traditional system of medicine since ancient times. This section will show some of the major pharmacological aspects of *H. indicus*.

3.1. Anti-inflammation, Anti-nociceptive and Anti-pyrectic

The inflammation induced by Vipera venom in albino mice is effectively neutralized by methanolic extracts of 2-hydroxy-4-methoxy benzoic acid from the root of *H. indicus* and it reduced the body temperature in yeast-induced pyrexia in rats (Alam and Gomes, 1998). The methanolic extract of the root (100, 200, and 400 mg/kg orally) showed anti-inflammatory activity against carrageenan-induced rat paw edema and antipyretic activity (100 mg/kg) against brewer's yeast-induced pyrexia in rats (Lakshman *et al*., 2006). The ethanolic *H. indicus* root extract showed a dose-dependent antinociceptive activity in acetic acid (Writhing test) and formalin (paw licking test)-induced nociception in mice, the root extract is administered orally at a dose range of 25, 50, and 100 mg/kg which blocked both neurogenic and inflammatory pain (Verma *et al*., 2005). The anti-inflammatory activity of the carrageenan-induced hind paw model in rats is exhibited by the hydroalcoholic extracts of *Capsicum annum* and *H. indicus* at a dose level of 100 mg/kg (Vijayalakshmi *et al*., 2010). The methanolic crude extract (MHI) and methanolic extract (ME1) from the crude (MHI) of *H. indicus* root show reduced inflammation activity induced by *Salmonella typhimurium*, in which rat ileum infected with ME1 treated *S. typhimurium* and the rat pre-administered with MHI extract followed by infection with wild *S. typhimurium* exhibits inhibition of type three secretory proteins encoded by SPI-1 (involved in invasion and enteritis) and SPI-2 (involved in intracellular survival and multiplication) (Das and Devaraj, 2009).

3.2. Anti-cancer

The polyherbal decoction composed of *Nigella sativa* (seeds), *Hemidesmus indicus* (root), and *Smilax glabra* (rhizome) mediates anti-hepatocarcinogenic effects through an Anti-inflammatory mechanism. Oral administration of decoction to the C3H mice injected with diethylnitrosamine (DEN) inhibits the activation of nuclear factor kappa B (NF-κB) and it is due to the inactivation of the TNF-dependent IKKβ activity by the components responsible for anti-inflammatory activity in the decoction (Galhena *et al*., 2012). Iddamaldeniya *et al*., 2006 reported that the long-term administration of decoction composed of *N. sativa*, *H. indicus* and *S. glabra* to the rat inhibited the DEN induced Glutathione S-transferase-P (GST-P) along with carcinogen-mediated development of overt tumors and histopathological changes that lead to tumor development. Chemopreventive potential of *H. indicus* was assessed by 7,12-dimethyl-benz[a]anthracene and 12-O-tetra-decanoyl 13-phorbol acetate (TPA) promoted murine skin carcinogenesis, topical application of *H. indicus* exhibits significant protection against cutaneous tumorigenesis. Plant extract administration at a dose range of 1.5 and 3.0 mg/kg body weight in acetone prior to the TPA treatment resulted in significant inhibition of oxidative stress and it also resulted in reduced level of lipid peroxidation and restoration of depleted levels of glutathione and reduced antioxidant enzyme activity (Sultana *et al*., 2003).

3.3. Antivenom

Inhibition of viper venom-induced hemorrhagic and coagulation in albino mice has been achieved by using the methanolic extracts of *H. indicus* and the compound responsible for the inhibition activity has been identified to be 2-hydroxy-4-methoxy benzoic acid (Alam *et al*., 1994). The viper venom neutralization using *H. indicus* and *Pluchea indica* done by Alam *et al*., 1996 reported that the *H. indicus* provides maximum protection against viper venom than *P. indica*. The chemical compound other than 2-hydroxy-4-methoxy benzoic acid which showed effective venom neutralization activity against venoms of *Daboia russellii* and *Najakaouthia* is lupeol acetate (LA) which is extracted by the methanolic extracts of *H. indicus* root by Chatterjee *et al*. (2006), they also reported that LA showed neutralization against the hemorrhagic, defibrinogenatingedema and phospholipase A2 (PLA2) induced by the activity of *Daboia russellii* venom, LA also showed neutralization of cardiotoxic, apnoea and PLA2 activity of *Najakaouthia* but it failed to provide protection against neurotoxic activity of the *Najakaouthia* venom in the albino mice. Alam and Gomes (1998) produced antiserum from rabbits using *Viperarussellii* venom, the rabbits are introduced with 0.7 mg *Viperarussellii* venom along with 2-hydroxy-4-methoxy benzoic acid (200 mg/kg, s.c) the produced antiserum showed neutralization of *Viperarussellii* along with *Echiscarinatus*, *Najakaouthia* and *Ophiophagus hannah* venom-induced lethality in albino mice.

3.4. Anti-diarrhoeal

The methanolic root extract of *H. indicus* at the dose of 1500 mg/kg showed a more effective reduction of diarrhea than the drug Lomotil which is used to treat diarrhea (Das *et al*., 2003). Evans *et al*., 2004 proved the injection of water extracts of *H. indicus* into the jejunal sac of rat increase the absorption of water and Na^+ and K^+ ions from the sac, they also suggested that the incorporation of *H. indicus* root powder or water extract in oral rehydrating solution may increase its anti-diarrhoeal activity.

3.5. Anti-leprotic

2% concentration of *H. indicus* aqueous extract drug delayed the proliferation of *Mycobacterium leprae* in mice,the prevention is achieved by delaying cutaneous hypersensitivity stimulation and the dose of plant drug which showed effective reaction is 100 mg/kg^{-1} (Gupta, 1981).

3.6. Anti-ulcer

Austin and Jegadeesan, 2003 reported that the roots harvested during flowering season expressed better anti-ulcer property and the root extracts react via mucoprotective activity and selective inhibition of prostaglandin. Ethanolic extracts of *H. indicus* whole plant administered orally in two concentrations (200 and 400 mg/kg) against indomethacin-

induced ulcer model in Wistar rats showed significant anti-ulcer activity through cytoprotective activity or by strengthening of the gastric mucosa that enhances mucosal defense (Vishali *et al*., 2011). Indomethacin induced peptic ulcers in Wister Albino rats are treated using aqueous (500 mg/kg) and alcohol (100 mg/kg) extracts of roots, in that alcohol root extracts showed effective ulcer healing property than the aqueous root extract (Bharadwaj and Nayak, 2013).

3.7. Antidiabetic

The diabetic effects (glycosuria, hyperglycemia, polyphagia, polydipsia) induced by administration of alloxan (150 mg/kg) in Wister albino rat can be reversed by aqueous root extract of *H. indicus* via antihyperglycemic activity by increasing the secretion of insulin or enhanced transport of blood glucose to peripheral tissue. An increase in lipid levels in the serum by alloxan administration can also be reduced by antihyperlipidemic action of root extract, which serves as a defense mechanism against atherosclerosis development (Sowmia and Kokilavani, 2007). Oral administration of (500 mg/kg) aqueous root extract reduces blood glucose level in 5 hours and 12 weeks administration changes the level of insulin, glycosylated hemoglobin, total cholesterol, High-Density Lipoprotein-cholesterol, Low-Density Lipoprotein-cholesterol, triglycerides, alkaline phosphatase, aspartate transaminase, γ-Glutamyl transferase and creatine kinase to normal level was observed in streptozotocin-induced diabetic rats. The same result was observed in the administration of 2-hydroxy-4-methoxy benzoic acid extracted from the root at a dose of 500mg/kg in streptozotocin-induced diabetic rats (Mahalingam and Kannabiran, 2009a; 2009b).

3.8. Diuretics

The aqueous (200 mg/kg) and ethanolic (400 mg/kg) *H. indicus* root extracts are administered orally to the dehydrated rats and both extracts showed a significant increase in urine output in higher doses, the diuresis induced by aqueous extract (400 mg/kg) in 5 hours showed the similar result of the drugs frusemide and hydrochlorothiazide and also reveals the increase in urinary Na^+ and K^+ levels (Gadge and Jalalpure, 2011).

3.9. Anti-Microbial

The aqueous (hot and cold) and solvent (acetone, chloroform, methanol) extracts of flowering and vegetative seasonal samples are tested against the human isolates of *Helicobacter pylori* in disc diffusion method, among different extracts only chloroform extracts of both flowering and vegetative season sample extracts showed maximum zone of inhibition (vegetative 8mm and flowering 11 mm) and analyzed further for comparison of minimal inhibitory concentration (75 µg/disc) and minimal lethal concentration (100 µg/disc) with the known concentration of antibiotics (Austin *et al*., 2003). Das and Devaraj, 2006 performed antienterobacterial activity using chloroform (CHI), methanol (MHI), and fatty substance removed methanol extracts (ME1) against different enterobacterial strains using different methods (disc diffusion, agar well diffusion, modified agar well diffusion, and swab methods). In the disc diffusion method, ME1 extract showed more potent inhibition action against both standard and clinical isolates of *S. typhimurium*, in overall ME1 found to more efficient than MHI followed by CHI might be due to inefficient diffusion. Different extracts of *H. indicus* (petroleum ether, chloroform, methanol, ethanol, and aqueous) are tested for antibacterial activity against 15 human pathogenic microorganisms using standard disc diffusion methods. Moderate inhibition was exhibited by the extracts with the MIC (minimum inhibitory compositions) range from 6.20 to 10.65 mg when compared to standard antibiotics (ampicillin, tetracycline, and chloramphenicol) which exhibits MIC from 9.5 to 21.6 mg/ml) (Sujatha and Anusha, 2010). The ethanolic extracts of *H. indicus* tested for the antibacterial activity against *Escherichia coli* by colony-forming assay unit and the effect of extract activity on the bacterial membrane by fluorescence-activated cell sorting and scanning electron microscope. The bacterial killing was observed with 400 and 500 µg/ml concentration of the extract (Saritha *et al*., 2015).

4. The Economical importance and Conservation

From the economic point of view, there is an increasing demand for *H. indicus*, the reason is due to the ability of *H. indicus* to produce a variety of phytochemicals having pharmacological activity, those phytochemicals are required in the preparation of medicine in pharmaceutical industries, production of cosmetics, and serves as a flavoring agent in food products like confectionery and baked foods and in soft drinks, because of these applications *H. indicus* has been identified as most traded medicinal plants (Sreekumar *et al*., 1998; Kher *et al*., 2020). The production of cosmetics and related herbal products using H. indicus has been reported in detail by Nandy *et al*., 2020a. Especially the root part has high demand, the roots are extensively used in traditional medicine (Ayurveda and Unani) as antipyretic, anti-diarrheal, and as a blood purifier and also recommended for skin disease, syphilis, fever, bronchitis, asthma, kidney, and urinary disorders and rheumatism. Apart from this *H. indicus* has antivenom activity against scorpion sting and snakebite (Austin, 2008; Rathi *et al*., 2017). 2-hydroxy 4-methoxy benzaldehyde (MBALD) an isomer of vanillin which is accumulated in the root of *H. indicus* is used as a flavoring agent and it is used to make soft drinks and beverages, it is used to prepare a health drink by the name Nannarisharbat or Rayalaseema sharbat, these health drinks are considered to provide relief from constipation and acidity and it is quite popular in south India, other than economical usage the phytochemical constituents of *H. indicus* root has the potential to purify the water contaminated with industrial wastes (Raju and Ramana, 2011; Rathi *et al*., 2017; Dandekar *et al*., 2018). Because of these properties, root harvesting is increasing rapidly, it also includes the collection of roots from the reproductively immature plants. per year 1,614 tonnes of roots are harvested mostly from the wild. *H. indicus* is a slow-growing plant and the yield of root biomass is low, the collection of root affects the natural regeneration of plants (Patnaik and Debata, 1996; Raju and Ramana, 2011; Goyal *et al*., 2015; Nandy *et al*., 2020b).Considering the commercial importance for increasing demand on *H. indicus* and threats posed on their existence in natural habitat, several conservative methods have been adopted. the methods like in vitro production of secondary metabolites, in vitro cryo-banking and genetic transformational studies to re-introduce critically endangered or endangered species, production of synthetic seeds and to reduce the pressure on natural habitats, DNA bar-coding for identification and conservation, in-vitro micropropagation to induce organogenesis and somatic embryogenesis from callus induction (Patnaik and Debata, 1996; Austin, 2008;

Cheruvathur *et al*., 2013; Thomas and Hoshino, 2016; Mishra *et al*., 2017; B.B. Kundu *et al*., 2020).The production of synthetic seed from the nodal cuttings of *H. indicus* using two concentrations (3 and 4%) of sodium alginate and 100mM calcium chloride (CaCl2) which ensures somatic embryogenesis and long-term storage (Chervathur *et al*., 2013). The micropropagation of nodal explants of *H. indicus* in the full, half, and quarter strength MS medium (Murashige and Skoog, 1962) shows the emergence of 0.2 – 0.5 mm bud in 2 weeks which showed better results when it is compared to other explants and the composition of growth regulators 2.22 µM BA and 1.07 µM NAA in the half-strength MS medium shows the best result for shoot initiation within 5-6 weeks which are reported by Sreekumar *et al*., 2000 (Thomas and Hoshino, 2016). Production of secondary metabolites can be done by initiating cell culture without sacrificing the plant and it also can be enhanced. The production of 2-hydroxy-4-methoxy benzaldehyde which is the principal component in the root of *H. indicus* can be produced by initiating the root culture (Sreekumar *et al*., 1998). Production of 2-hydroxy-4-methoxy benzaldehyde (MBALD) in the root is coupled with the synthesis of phenylalanine ammonia-lyase (PAL) enzyme through the phenylpropanoid pathway, it is confirmed by suppressing the activity of PAL using aminooxy acetic acid. The bio-synthesis can be elicited by chitosan which activates defense or stress response in the plant and results in the production of secondary metabolites (Chakraborty *et al*., 2008; S.Nandy *et al*., 2020a). Secondary metabolite production can also be enhanced by initiating hairy root culture upon incubating explants with *Agrobacterium rhizogenes*, hairy roots are bio-synthetically and genetically stable and it can be used as a source for the production of secondary metabolites and it can be improved by the external application of methyl jasmonate and chitosan (Sreekumar *et al*., 1998; Sevon and Caldentey, 2002; Chakraborty *et al*., 2008; Pistelli *et al*., 2010; Swathi *et al*., 2019). The plant is mainly harvested for the vast medicinal properties of the root, so the conservation technique is specifically focusing on in-vitro culturing of roots and to increase their efficiency for producing secondary metabolites in higher quantities.

Conclusion

By going through all these studies, it clearly proves that *H. indicus* is one such irreplaceable ethnomedicinal plant that has a vast application over pharmaceuticals, nutraceuticals, cosmetics, confectionery, and beverages. An ethnomedicinal study indicates their multipurpose use in the preparation of traditional medicine and to treat various ailments from common infections to life-threatening diseases like cancer. because of all these applications, *H. indicus* is facing a rapid decline in their natural population due to over-harvesting. *H. indicus* is an indigenous plant species found in Indian sub-Continents, so the conservation of the plant becomes the first and foremost priority. the conservation can be established by approaching genetic engineering and plant tissue culture techniques, by which the whole plant can be regenerated in in-vitro condition, and their efficiency to produce phytochemical can be increased using gene modification of the pathways responsible for the production. Production of synthetic seeds from the nodal cuttings by which somatic embryogenesis and organogenesis are induced, it can be preserved for a long time. Other plant species that possess similar characteristics like *D. hamiltonii* can be used as a substitute in medicinal and other preparations. In recent times people started to prefer traditional medicines (Ayurveda and Unani) over allopathy medicine, due to no side-effects in traditional medicine, so there is a better scope for *H. indicus* in the future and their importance along with other medicinal plants will be established.

Ethnobotanical studies:

Serial No	Plant part Used	District/ State/Country	Local Name	Ethnomedicinal Use	Indication for Preparation	References
1	Root, Leaves	Odisha, India	Anantamul	One of the ingredients for preparation of prophylactic remedy for malaria	Extraction of juice from leaf and root	Nagendrappa *et al*., 2013
2	Root	Uttara kannada district, Karnataka, India	Sugandhi	Treatment of leucorrhoea during menstrual period	Ground with *Mimosa pudica L.*	Bhandary *et al*., 1995
3	Root	Uttara kannada district, Karnataka, India.	Sugandhi	Body tonic	Powdered root with cow's milk	Bhandary *et al*., 1995
4	Root	Chanduli district, Uttar Pradesh, india.	Anantamul	Eczema, scabies and ringworm infection	Boiling of pounded dry roots in coconut oil	Singh and Singh, 2009
5		Orissa	Anatamula	Diarrhoea		Dash and Padhy, 2006
6	Root	Sothern part of Tamil Nadu, India	Anantamul	Snakebite	Decoction of root.	Samy *et al*., 2008;
7	Root	India	Anantamul	Snakebite	Decoction of root.	Makhija and Khamar, 2010
8	Root	Sitamata wildlife sanctuary, Rajasthan, India	Garmali	Gonorhoea, antidote.	Powdered root.	Jain *et al*., 2005

9	Root	Mayurbhanj district, Orissa, India	Anatamula	Skin disease	Powdered root	Rout *et al*., 2009
10	Leaf	Mayurbhanj district, Orissa, India	Anatamula	Piles	Pasted leaf	Rout *et al*., 2009
11	Root	Mayurbhanj district, Orissa, India	Anatamula	Gout and joint pain	Boiling of crushed roots for 10-15 minutes with mustard oil	Rout *et al*., 2009
12	Stem	Sri-lanka	Eramusu	Skin disease		Arseculeratne *et al*., 1985
13	Root	Malkangiri district, Orissa,	Chirmar	Toothache	Root paste	Pattanaik *et al*., 2007
14		Virudhunagar district, Tamil Nadu, India	Nannari	Dermatological ailments		Mutheeswaran *et al*., 2011
15	Root	Meerut district, Uttar Pradesh, India	Gurmar	To treat swellings, blood purifier, and skin disease	Root paste and root decoction	Tomar, 2009
16	Root		Anatamuli	Edema induced by Russell's viper bite		Gupta and Peshin, 2012
17	Leaves	Bogra district, Bangladesh	Anantamul	Urinary tract infection		Hossan *et al*., 2010
18	Root	Sundargarh district, Orissa, India	Anantamul	Stomach ache	Root decoction	Mukherjee and Namhata., 2010
19	Whole plant	Kancheepuram district, Tamil Nadu, India	Nannari	Body coolants	Juice extraction from whole plant	Muthu *et al*., 2006
20	Whole plant	Madurai district, Tamil Nadu, India	Nannari	Fever	Decoction of whole plant	Ignatchimuthu *et al*., 2006
21	Root	Orissa, India	Dudhili	Increase lactation	Root powder along with stem bark of *Syzygiumcumini in water*	Das *et al*., 2003
22	Root	Orissa, India	Sugandhi	Excess Menstruation	Root decoction	Das *et al*., 2003
23	Root	Orissa, India	Sugandhi	Itching	Root paste	Das *et al*., 2003
24	Whole plant	Shimoga district, Karnataka, India	Sogadaeberu	Stomach ache	Unboiled cow's milk with whole plant paste	Mahishi *et al*., 2005
25	Root	Coimbatore district, Tamil Nadu, India	Nannari	Body coolant	Root decoction	Kumar, p *et al*., 2007
26		Tirunelveli district, Tamil Nadu, India	Nannari	Venereal diseases and stomach ache	Prepared along with *Alternanthera sessilis, Hibiscus rosasinensis, Centella asiatica, Pavetta indica, Vetiveriazizanioides*and consumed with cow's milk	Ayyanar and ignatchimuthu, 2011
27	Leaf	Erode district, Tamil Nadu, India	Nannari	Stomach ache and improper digestion	Root and leaf decoction	Revathi and parimelazhagan, 2010
28	Leaf	Eastern Himalayan zone of Arunachal Pradesh, India	Mamephul	Bone fracture	Leaf paste	Tangjang *et al*., 2011
29	Root	Magura district, Bangladesh	Anontomool	To increase sperm production		Rahmatullah *et al*., 2010
30	Root	Southern districts of Tamil Nadu, India	Nannari	Cooling beverages	Root powder and sugar solution in water	Rajendran *et al*., 2008
31		Odisha, India	antamula	Joint pains	oil	Panda, 2014
32	Root	Purulia district, West Bengal, India	Anantamul	Antidote for snake bite	Root paste	Chakraborty and Bhattacharjee, 2006

33	Root	Theni district, Tamil Nadu, India	Nannari	Body cooling	Decoction	Ignacimuthu *et al*., 2008
34	Leaf and root	Cuddalore district, Tamil Nadu, India	Nannari	Blood purification	Extracts of leaf and root	Anbarashan and Padmavathy, 2010
35	Whole plant	Raigarh district, Chhatisgarh, India	Dudhi bel	High fever	Small pieces of plant material stung in a thread for five days	Jain and Singh, 2010
36	Root	Sirumalai, Tamil Nadu, India	Nannari	Ulcer	Root decoction	Karuppusamy, 2007
37	Root	Visakhapatnam district, Andhra Pradesh, India	Sugandhipala	Rheumatism	Root powder in water	Bapuji and Ratnam, 2009
38	Root and leaves	Ayyakarkoil, Tamil Nadu, India	Nannari	Bronchial asthma, fever, wounds and leukoderma	Root decoction and leaf paste	Rajendran *et al*., 2003
39	Root	Andhra Pradesh, India	Sugandapala	Blood purification	Root decoction	Reddy *et al*., 1988
40	Root and whole plant	Bastar district, Madhya Pradesh, India	Sugandhiapala	Stomach ache and fever	Chewing of root for stomach ache and whole plant crushed in water for fever	Jain, 1965
41	Root	Madurai district, Tamil Nadu, India	Nannari	Menorrhagia	Root paste with water or cow's milk	Ignacimuthu *et al*., 2006
42	Leaf	Madurai district, Tamil Nadu, India	Nannari	Stomach ache	Fresh leaves are taken orally	Ignacimuthu *et al*., 2006
43	Root	Yercaud hills of eastern ghats, Tamil Nadu, India	Nannari	Rheumatism	Powdered root with water	Parthipan *et al*., 2011
44	Root	Kotia hills Vizianagaram district, Andhra Pradesh, India	Sugandhipala	Snakebite	Root paste along with A*lliumsativum*	Naidu *et al*., 2013
45	Root	Salem district, Tamil Nadu, India	Nannari	Snakebite	Root decoction	Alagesaboopathi, 2013
46	Root	Kaniyakumari district, Tamil Nadu, India	Nannari	Body coolant and mouth ulcers	Root extracts	Uma and Parthipan, 2015
47	Root	Kouthalai of Tirunelveli hills, Tamil Nadu, India	Nannari	Increase the production of sperm	Root powder along with the fruits of *Calophylluminophyllum, Diospyros ebenum, Terminalia chebula, Terminalia bellirica and Phyllanthus emblica*and with honey	Ayyanar and Ignacimuthu, 2005
48	Whole plant	Shenbagathope in Virudhunagar district, Tamil Nadu, India	Nannari	Fever, leprosy and scorpion sting		Shanmugam *et al*., 2009
49	Leaf, root and stem	Thoppampatti, Dindigul district, Tamil Nadu, India	Nannari	Blood purifier, syphilis, leucorrhea, galactagogue, diarrhea, febrifuge alterative and abscess	Decoction and orally	Sivasankari *et al*., 2014
50	Root and whole plant	Silent valley, Kerala, India	Nannari	Jaundice, cooling agent and body pain	Juice, paste and decoction	MorvinYabesh, J.E., *et al*., 2014

Methods for Conservation:

S.No	Techniques	Explant used	Optimum Concentration	Observed result	References
1.	Micropropagation	Nodal segments	2.22 μM BA and 1.07 μM NAA; 9.8 M IBA	Shoots reached a length of 7–8 cm in 5-6 weeks.	Sreekumar *et al*., 2000

				Root initiation was achieved easily in quarter salt strength medium with less traces or no callusing, 9.8 M IBA induced maximum formation of roots without callusing.	
2.	In vitro regeneration	Leaf and nodal segments	1.0, 3.0, 5.0 mgL^{-1} concentration of IAA, NAA, IBA; 1.0, 3.0, 5.0 mgL^{-1} concentration of 2,4,5-T, Kn and BAP; 0.5 mgL^{-1} IBA and Kn.	Different composition of PGR's showed effective callus induction, shoot differentiation and rhizogenesis from shoot. 95% of the Produced plantlets showed vigorous growth upon acclimatization.	Shanmugapriya and Sivakumar, 2011
3.	Root culture initiation	Nodal segments	2 mg IBA l^{-1}; 2 mg IBA l^{-1} and 4% sucrose	2 mg IBA $^{l-1}$ in Half strength MS media produced 10-12 roots in 10 days. Supplementation of 2 mg IBA l^{-1} and 4% sucrose in Gamborg *et al*., medium produced 550 mg root and 0.18% MBALD in root after 30 days.	Sreekumar *et al*., 1998
4.	In vitro regeneration	Nodal segment	10 µM BA and 5 µMKn	Showed presence of lupeol in regenerated shoots and the concentration is slightly higher than wild shoots.	Pathak *et al*., 2017
5.	Micropropagation	Nodal segment	1.15 µMKn and 0.054 NAA; 7.35 µM IBA and 1.15 µMKn	95% frequency with shoot multiplication rate of 8.2 ± 0.4 shoot/explant. Better rooting response are seen in shoots from subcultures than the primary cultures.	Patnaik and Debata., 1996
6.	In vitro propagation	Nodal segment	0.1 mgl^{-1} NAA and 2.0 mg l^{-1} BAP; 1.5 mg l^{-1} IBA	Bud break within 4 days after inoculation and maximum production of root is observed. Regenerated plants show 85% survival rate	Saha *et al*., 2003
7.	Synthetic seeds	Somatic embryos initiated from nodal cuttings	4 µM BA and 1.5 µM GA_3; 3% sodium alginate and 75 mM $CaCl_2$	Germination of synthetic seeds after 120 days of storage at 4℃ was observed and 92% success rate in survival	Cheruvathur *et al*., 2013
8.	PAL mediated biosynthesis of MBALD	Root	200 mg/L chitosan	Activity of PAL can be increased upon chitosan treatment which increases the accumulation of phenolic compounds in the root	Chakraborty *et al*., 2008

2,4,5-T, 2,4,5- Trichloro phenoxy acetic acid; **BA,** N^6-benzyladenine; **BAP,** Benzyl Amino Purine; **GA_3**, Gibberellic acid; **IAA,** Indole-3- Acetic acid; **IBA,** Indole-3-Butyric Acid; **Kn,** Kinetin; **NAA,** α-naphthalene acetic acid; **PAL,** Phenylalanine ammonia-lyase.

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