



Review Article

Phytochemical, morphological and pharmacological aspects of *Hybanthus enneaspermus*: A review

Abhishek Konar, Kübra Sağlam, Tahsina Tabia, Pranabesh Ghosh

Abstract

At this time, the world faces a critical challenge from various diseases due to high pollution and rapid industrial growth. Nowadays, in advanced treatment, various synthetic drugs are available. A synthetic drug not only cures diseases, but also has severe side effects on the human body and has high-cost value. A lot of examples reported that synthetic drugs are associated with side effects. As a result, Global Medicinal plants create huge demand due to low cost, fewer side effects. It has been continuously tried to minimize critical disorders in developed countries by utilizing secondary and primary plant bioactive phytochemical compounds. In this perspective, *Hybanthus enneaspermus* is an important multipurpose medicinal herb or a shrub. This plant belongs to the Violaceae family, commonly known as Ratanpuru, and has pink-colored flowers. It has been used as a medicinal herb since ancient times. The plant is used as an anti arthritic, anti-inflammatory, anti-microbial, anti-diabetic, antioxidant, and anti-ulcer. With potent antioxidant, nephroprotective, anti-inflammatory, antimicrobial, antidiabetic, anti-arthritic, anti-allergic, hepatoprotective, and anti-ulcer properties, *Hybanthus enneaspermus*'s pharmacological potential is summed up in this study. Numerous extracts exhibit notable bioactive qualities, such as ethanolic, aqueous, and ethyl acetate fractions. According to these results, *Hybanthus enneaspermus* has a variety of therapeutic uses and can be a useful natural resource for creating innovative herbal medications.

Keywords bioactive compounds, ethnobotany, medicinal plant, traditional uses

Introduction

Today, health problems are a major issue in developing countries [1-3]. In this time, common synthetic medicines are available like Aspirin, SGLT2 inhibitors, Clopidogrel, Diclofenac, Atorvastatin, Enoxaparin, Sulfasalazine, Ibuprofen, Metformin, Naproxen and Warfarin and are associated with various serious side effects of back pain, gastrointestinal, kidney, joint pain, bone fracture, folic acid deficiency, excessive bleeding, headaches, etc [4-8]. In the USA, about 8% of people are admitted to the hospital, and approximately 100,000 people die every year from the high toxicity of synthetic drugs [3]. Although many synthetic medicines serve Humanity, they are reported to have versatile side effects [3-4]. However, herbal medicinal/ homemade remedies are much safer and lower cost than synthetic drugs.

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Authors:

A. Konar ✉, T. Tabia, P. Ghosh
School of Life Sciences, Seacom Skills
University, Santiniketan, Bolpur,
Birbhum, West Bengal, India

K. Sağlam
Department of Food Processing, Food
Technology Program, Istanbul Gelisim
Vocational School, Istanbul Gelisim University,
Avcılar, Istanbul, Türkiye

✉ abhishek.konar60@gmail.com

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In addition to their therapeutic value and value as raw materials for the manufacture of novel and traditional medications, medicinal plants are a vital component of natural resources. In phytomedicine, herbal medicine is practiced according to the highest standards of giftedness and medical studies. The use of herbal drugs in deliberate holistic therapies is useful in almost every aspect of disease treatment [3-4]. That's why medicinal plants created a huge demand today [3]. Primarily, sources of natural biomolecules occur in various plants. Some medicinal plants are available in developing and impoverished nations, and rural people use them. In ancient India, medicinal plants were used to treat a variety of illnesses. Medicinal plants produce an infinite amount of phytochemicals that are extremely complex and potent. Globally, hundreds of naturally occurring bioactive substances have been identified and use [2-6].

In this view, *H. enneaspermus* (L.) F. Muell., is an important medicinal plant; it is an herb or a shrub that belongs to the Violaceae family. It is an ethnobotanical herb that is rare. This plant in the Yanadi and Santal tribes is known as "Ratanpurus". In the Ayurveda system called Sthalakamala. The "humpback flower" is another name for the flower. "Humpback flower" is a Greek word. This herb is used mostly to treat 'kapha' and 'pitta' problems in Ayurvedic literature. In the world of different regions, this plant belongs to 150 species. Ancient medicinal systems (Ayurveda & Siddha) mention their magical medicinal properties and to treat various critical diseases [8-14]. This review summarizes *H. enneaspermus* morphological characteristics, phytochemical composition, traditional uses, and phytopharmacological properties such as an anti arthritic, anti-inflammatory, anti-microbial activity, anti-diabetic, antioxidant, and anti-ulcer. It contains diverse phytochemicals responsible for strong antioxidant, anti-inflammatory, antimicrobial, antidiabetic, nephroprotective, hepatoprotective, and anti-ulcer activities. Various solvent extracts, including ethanolic and aqueous, exhibit significant bioactivity. Due to its broad pharmacological potential, *H. enneaspermus* is considered a valuable natural resource for developing effective and safe herbal medicines. This research aims to identify the bioactive compounds responsible for these effects and to establish *H. enneaspermus* as a promising natural source for developing effective, safe, and eco-friendly herbal medicine

Geographical distribution

In the Indian medicinal system, it is an essential plant. In tropical Asia, Taiwan, Africa, Australia, Ceylon, Guinea, and India, it grows as a tiny, suffrutescent perennial herb. Worldwide, there are over seventy species, the majority of which are found in America, ten in Australia, twenty in Africa and Madagascar, and five in Asia [14-16]. In India commonly found various state such as West Bengal, Andhra Pradesh, Bihar, Tamil Nadu, Gujarat, Uttarakhand, Kerala, Madhya Pradesh, Chhattisgarh, Maharashtra, Karnataka, Rajasthan, and Assam. The species grows in warm regions of the Deccan Peninsula in India along with other mimicking plants [9, 14].

Taxonomical classification [8-9]

Kingdom: Plantae

Phylum: Tracheophyta

Class: Magnoliopsida

Order: Malpighiales

Family: Violaceae

Genus: *Hybanthus*

Species: *Hybanthus enneaspermus*

Habitat

H. enneaspermus is an Annual or perennial herb. Most commonly found in the warmer parts of India and the semitropical and tropical regions of the world. It is commonly found in agricultural fields, along roadsides, along railroad lines, and in grassy areas [2, 9].

Characters of morphology

Root: Yellow color tap root system, branched, Slender. Roots are upto 10-12 cm long [9].

Stem: Stems are simple or branched, solid, and sweet-tasting. Stems are up to 17 cm in height [8-9].

Leaf: Leaves are alternate, small with lateral veins, 0.5-8.5 x 0.2-1.5 cm, elliptic to oblong-lanceolate. Leaves have a slightly sweet taste (Figure 1) [8-9].

Inflorescence: Flowers solitary [8-9].

Flower: Pink colour, Bracteate, bisexual flowers, zygomorphic hypogynous, 7 to 10 mm across. Flowers appear from July to November (Figure 1) [8-9].

Calyx: lanceolate sepals, sub-equal, polysepalous [8-9].

Corolla: Petals are five, unequal, polypetalous, imbricate, acuminate, regarding 3 to 5 millimetres long [8-9].

Androecium: Five Stamens, hypogynous, filaments very short, connate or free, connivent 2-4 millimetre long, two dorsal anthers in the anterior part [8-9].

Gynoecium: Stigma oblique, Superior, unilocular, ovule numerous, stigma capitates,

Fruit: Fruits are white in colour, capsule-shaped, and 5 mm thick [3, 8-9].

Seeds: It has many seeds, oval-ellipsoidal, swishy surfaces, longitudinal ribs, and is straight in embryonic development [8-9].

Pollination: Entomophilous pollination, i.e., by insects, cleistogamy pollination, i.e., self-pollination, or allogamy pollination, i.e., cross-pollination [1, 3, 8-11].



Figure 1. Whole plant part of *H. enneaspermus*

Anatomical aspects

Figure 2 shows the prominent anatomical features of *H. enneaspermus*, emphasizing important internal characteristics like vascular bundles, epidermal layers, cortex, and pith that support the physiological and therapeutic functions of the plant. Transverse Section of *H. enneaspermus* of leaf, stem, petiole, stomata, and root shows the following anatomical features:

Leaf: The epidermis leaf consists of elongated or polygonal cells with straight, compactly arranged cells containing mucilage [3, 8].

Stem: Transverse section of stem showing barrel shaped single layered epidermis, with thin cuticles, compactly arranged cells. Hypodermis consists of 1 to 2 layers collenchymatous and cortex layers of 5 to 6 layers [8].

Petiole: T.S. of the petiole showed a convex outline. Single-layered epidermis consists of tubular-shaped cells, trichomes, and thin cuticles [3].

Stomata: The stomata measure 19–26 mm in width and 28–31 mm in length. There are anisocytic stomata dispersed throughout the epidermis, but they are more plentiful on the abaxial surface than the adaxial surface [3, 8].

Root: The Phellem and cortex of a root are uniformly narrow. In the cortex, there is parenchyma, tangential cells, and secondary phloem [3, 8].

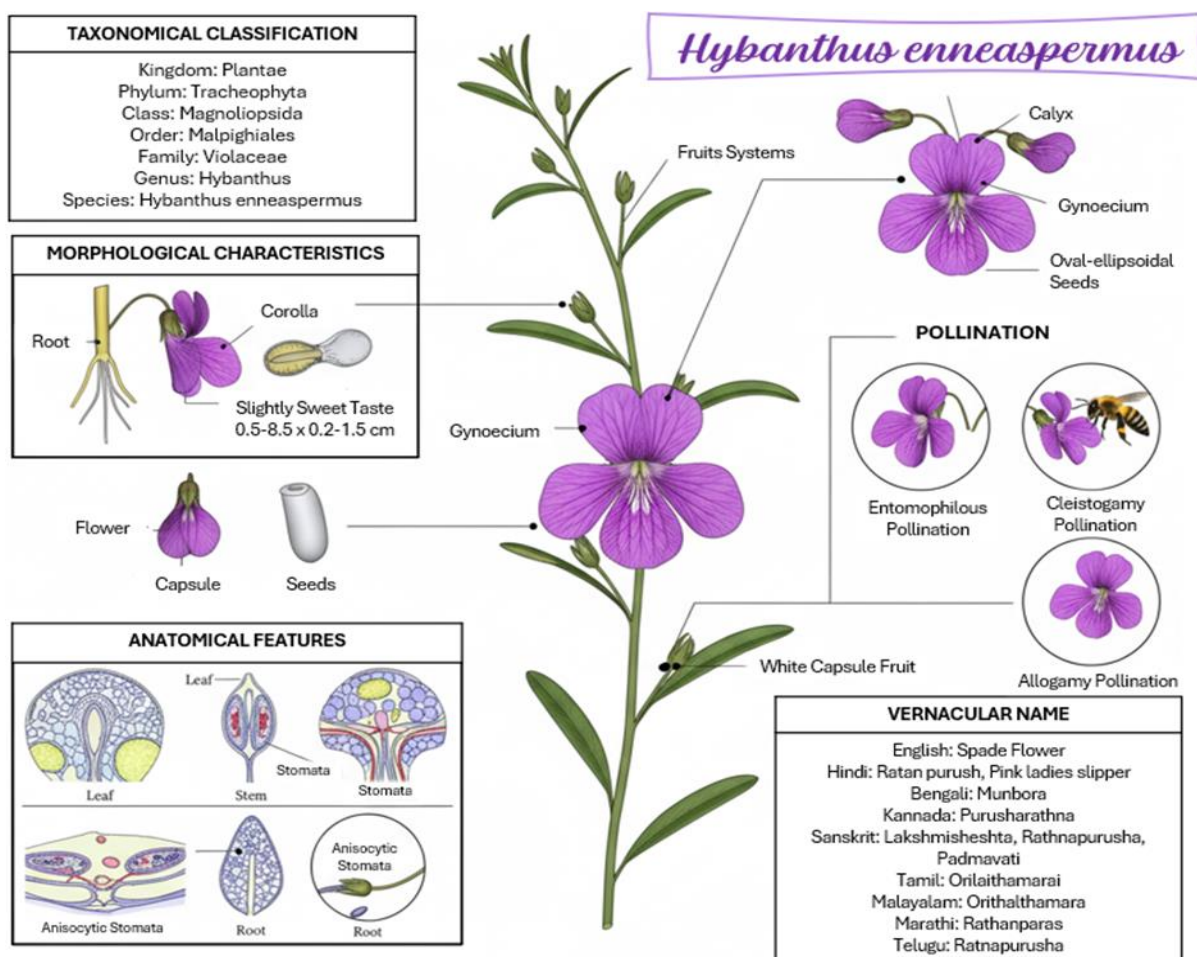


Figure 2. Anatomy, taxonomy, morphology, and vernacular names of the *Hybanthus enneaspermus* plant



Vernacular name

We provide a list of the various vernacular names mentioned in various books in the following list [11-13]:

English: Spade Flower

Hindi: Ratan purush, Pink ladies slipper

Bengali: Munbora

Kannada: Purusharathna

Sanskrit: Lakshmisheshta, Rathnapurusha, Padmavati

Tamil: Orilaithamarai

Malayalam: Orithalthamara

Marathi: Rathanparas

Telugu: Ratnapurusha

Traditional uses

Several illnesses and disorders have been shown to benefit from *H. enneaspermus* treatment [8]. Traditionally, it has been used to treat conditions such as blood trouble, cholera, asthma, epilepsy, vomiting, inflammation, dysuria, leucorrhea, diarrhea, and sterility [11, 16]. A general tonic is derived from the entire plant for pregnant women [8, 11, 17]. Roots are used as an infusion for UTI (Urinary-tract-infection). Leaves are applied externally to treat lesions in traditional medicine. Yanadi tribes use the leaf and fruit to treat scorpion bites and cobra bites [8, 18-21]. Roots are used as an infusion for UTI (Urinary-tract-infection). Leaves are applied externally to treat lesions in traditional medicine. Yanadi tribes use the leaf and fruit to treat scorpion bites and cobra bites [8, 18]. Furthermore, it treats skin conditions, gastrointestinal disorders, epilepsy, asthma, coughs, colds, and fevers. Whole plant pastes are used to treat cough. An oral treatment for diabetes and sexual energy is administered for four to five months using the whole plant's juice combined with cow's milk [21-23].

Combinations of bioactive

Numerous bioactive phytochemicals have been identified in *H. enneaspermus*. The leaf and entire plant contain high levels of secondary metabolites. This plant contains flavones, dipeptides, β -sitosterol, triterpenes, Catechin, phenols, and sitosterol [8, 9]. Polyphenolic chemicals in flavonoids are antibacterial, anti-inflammatory, protect cells from oxidative damage, and are strong anti-cancer agents. Among the 17 amino acids found in the leaf aqueous extract, leucine was the most abundant [9, 24]. This plant contains a significant amount of Fe and Mn [25].

A paper chromatographic experiment has demonstrated that *H. enneaspermus* contains several types of sugars, flavonoids, and amino acids [26]. Various types of research papers have confirmed that this plant contains essential bioactive molecules such as amino acids, sugars, tannins, and anthraquinones present as this plant by paper chromatography [9]. Savithramma et al., [27] phytochemical screening was conducted using 5 different solvents to identify phenols, tannins, saponins, flavonoids, alkaloids, terpenoids, glycosides, cardiac glycosides, coumarins, and steroids in floral extracts [27-28].

Phytochemical studies of Indian medicinal plants should be conducted since these secondary metabolites contribute to their therapeutic properties [29]. Another paper reported that Metformin, 2,3-dihydro-3, Menthol, Theobromine, Tetradecanoic acid, 2,6,6-trimethyl-, Catechin, 9,12,15-Octadecatrienoic acid, and Kaempferol [9, 30]. In an article published by Awobajo et al., [31], the aqueous extract of leaf contained flavonoids, cardiac glycosides, cyanogenic glycosides, anthraquinone glycosides, saponins, tannins, and hexose sugars [31].

According to the GC-MS analysis, there are over ten chemicals, which include 2-piperdinone, cedarn-diol, monolinoleo glycerol trimethyl silyl ether, phytol, and D-mannitol [1]. Nectar of *H.*

enneaspermus contains amino acids like glutamic, valine, and leucine [32-33]. Among the metabolites present in *H. enneaspermus* flowers, tannin was more abundant than alkaloids, steroids, phenol, flavonoids, saponins, and terpenoids [34]. Another article reported that the flower extract of this plant contains nanoparticles such as Ag and Au NP, phytochemicals of steroids, terpenoids, and alkaloids [2, 9]. Figure 3, shows a phytochemical illustration of *H. enneaspermus*, showing the major bioactive compounds reported from the plant.

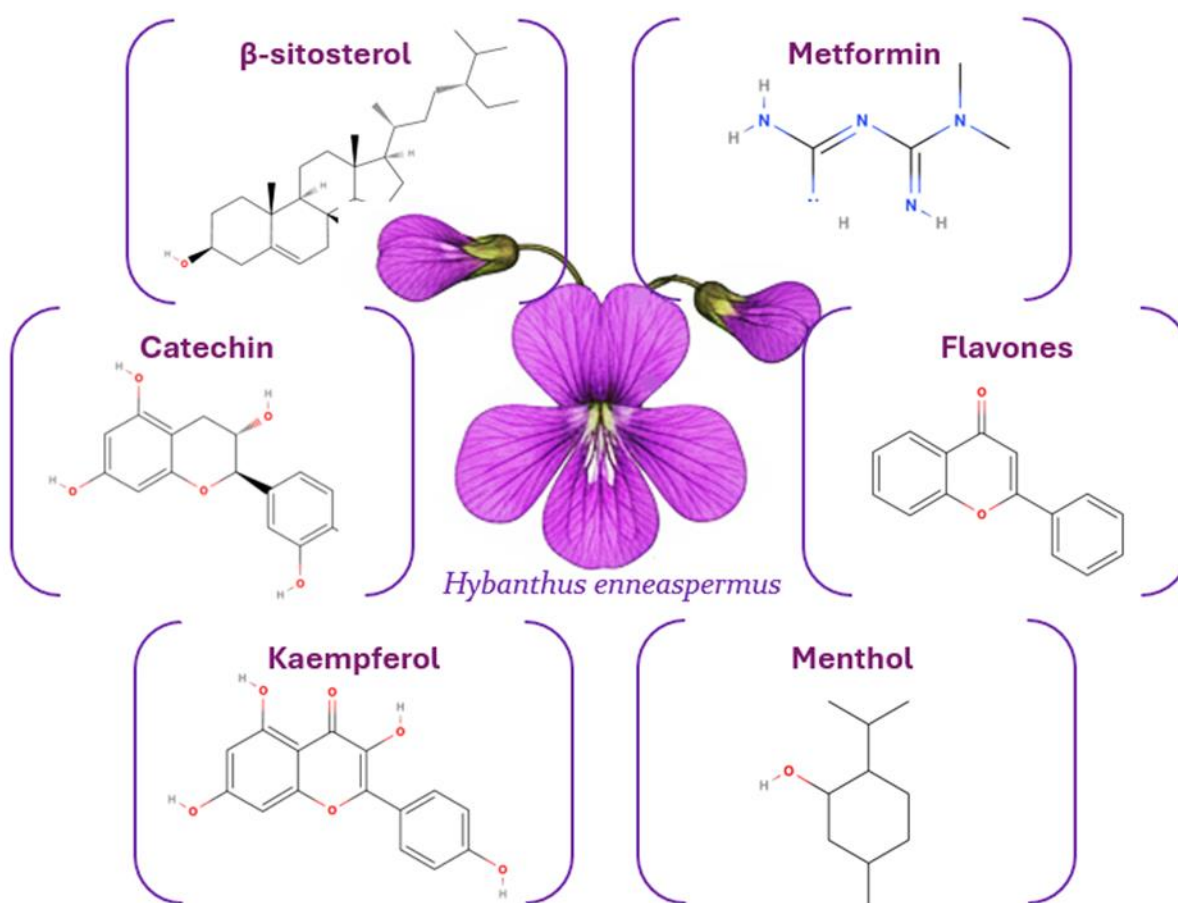


Figure 3. Phytochemical illustration of *Hybanthus enneaspermus*, showing the major bioactive compounds reported from the plant

Pharmacological aspects

Plants are the main sources of medicine for our daily lives. In today's healthcare industry, herbal remedies are taking on a prominent role. In accordance with WHO estimates, one-eighth of the global population uses herbal remedies in some aspect of basic healthcare without experiencing any side effects. A variety of bioactive substances possessing established antioxidant properties, including flavonoids, alkaloids, phenolic compounds, and saponins [21, 35-40]. *H. Enneaspermus* has high popularity due to its multiple disease treatments. This plant has occupied huge amounts of medicinal value in famous medicinal books. National and international researchers reported to have reported antioxidant, antimicrobial, and neuroprotective activities [8, 29]. The notable therapeutic effects of *H. Enneaspermus* are shown in Figure 4.

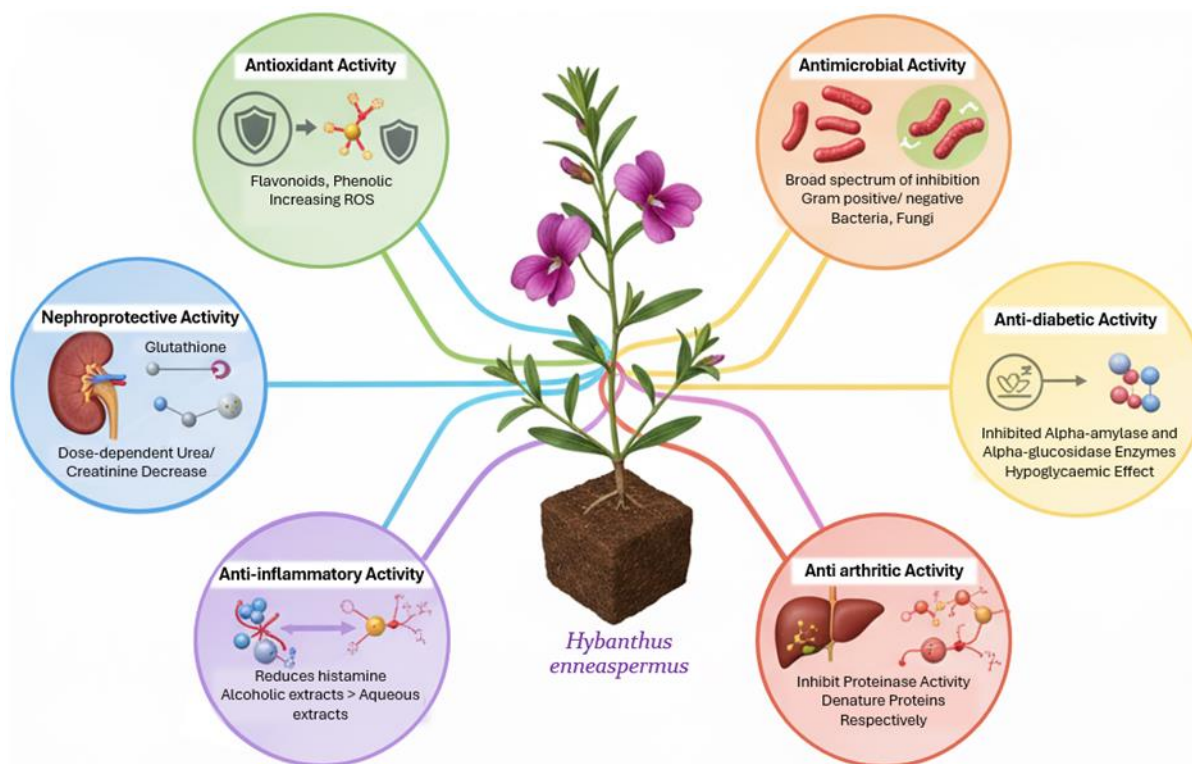


Figure 4. Pharmacological Aspects of *Hybanthus Enneaspermus*

Antioxidant activity

In this time, many food industries have been focused on naturally occurring antioxidants due to high toxic properties such as BHA (Butylated hydroxyl anisole) and BHT (Butylated hydroxyl toluene). Mainly, naturally occurring antioxidants are found in various plant parts [2]. The presence of phytoconstituents may contribute to *H. enneaspermus* strong free radical scavenging activity [8, 37]. In higher medicinal plants, a variety of pathways are developed, which determine how much reactive oxygen species are produced (ROS), increasing ROS, metabolites such as lipids, proteins, and nucleic acids are destroyed. In plants, antioxidant enzymes are activated to detoxify reactive oxygen species. Secondary metabolites of plant, which are abundant in many medicinal plants, are powerful antioxidants that can be useful in adsorbing and neutralizing free radicals [41]. There is evidence that plant extracts of *H. enneaspermus* are capable of scavenging free radicals with notable effects on TBARS, DPPH, ABTS, superoxide, nitric oxide, and ferric chloride [8, 13]. An extract of leaves was tested in vitro for its antioxidant properties. Five radical scavengers were tested: DPPH, hydrogen peroxide, reducing power, and nitric oxide radical scavengers, it was concluded that leaves possess strong antioxidant properties [42]. The antioxidant activity of this plant was assessed in vitro using hydrogen peroxide scavenging, DPPH, reduction power, nitric oxide, petroleum ether, chloroform, ethyl acetate, and aqueous fractions. Among the various studied fractions, ethyl acetate had the highest antioxidant property, reducing power, nitric oxide, hydrogen peroxide, and DPPH radical scavenging activity percentage. According to this study, the ethyl acetate fraction exhibited considerable antioxidant activity. Another researcher reported that the ethyl acetate fraction of this plant has high antioxidant properties [2, 43-45].



Nephro-protective activity

Several herbal remedies can treat kidney damage, one of the most serious health problems. Chronic renal failures, including acute kidney failure, have doubled in prevalence over the past 15 years. Nephrotoxicity is a condition that occurs when the body becomes exposed to a poison or medicine [46-47]. A cisplatin-induced kidney damage model in rats was used to assess the efficacy of alcoholic and aqueous extracts at dose levels of 250 and 500 mg·kg⁻¹. Increased glutathione levels, glutathione S-transferase activity, and superoxide dismutase activity were observed in kidney homogenate, resulting in the prevention of lipid peroxidation. Concentrations of blood urea and serum creatinine were decreased in a dose-dependent manner by the extracts [48].

Anti-inflammatory activity

The anti-inflammatory properties of *H. enneaspermus* alcoholic and aqueous extracts were evaluated against acute inflammation caused by carageenan, histamine, and 5-HT at dosages of 250 and 500 mg kg⁻¹. As well as reducing inflammation, the extracts reduce 5-HT levels and histamine levels. Alcoholic extracts had a greater and noticeable impact than aqueous extracts [49-50].

Antimicrobial activity

The diseases that are caused primarily by microorganisms are equally damaging to people and animals [51]. The antimicrobial activity of *H. enneaspermus* extracts in chloroform, petroleum ether, benzene, and methanol was evaluated against five Gram-negative bacteria, including *E. coli*, *E. aerogens*, *B. subtilis*, and two Gram-positive bacteria, *S. aureus* and *B. subtilis*. The extracts were shown to have similar antimicrobial activity to antibiotics when they were tested at 300 micrograms per disc [9]. Additionally, disc diffusion and micro broth dilution tests were used to evaluate the effects of extracts entire plant material on *C. tropicalis*, *C. Krusei*, and *C. albicans*. This showed, in contrast to the petroleum ether extract, which had a moderate impact, the ethanolic extract of this had a considerable and broad spectrum of inhibition. However, chloroform and aqueous solutions have strong antimicrobial activity [52]. Moreover, disc-diffusion and micro broth dilution assays were performed on ethanolic, aqueous, petroleum ether, and chloroform extracts of the whole plant of this plant, and these were tested against *C. albicans*, and *C. tropicalis*. From the obtained study, the ethanolic extract of this plant has a stronger inhibition spectrum than the petroleum ether extract. However, compared to other fractions, chloroform and aqueous extracts were less active [53]. Besides its antiviral properties, this herb also has other benefits. An anti-HIV efficacy test was performed on *H. enneaspermus* using methanol and hexane extracts. As part of its evaluation of its anti-HIV activity, *H. enneaspermus* leaf extract is now tested for reverse transcriptase inhibition activity. Excellent extraction results were achieved in methanol and hexane, but no inhibition effect of chloroform and petroleum [9, 54]. Besides, other researchers discovered that solvent extracts of this plant have a unique property against urinary tract infections [9].

Anti-diabetic activity

Worldwide wide diabetes mellitus (DM) is a common disorder of humans. Throughout the world, people suffer from diabetes mellitus due to malfunctions in insulin production. The kidneys, eyes, heart, and neurological systems may be affected in these cases [11]. Anti-diabetic activity of aqueous and methanolic extract of leaf was measured using 80 and 180 mg·kg⁻¹, in Diabetic rats. After 12 hours, the result showed a hypoglycaemic effect of the diabetic rate [8]. Hypoglycaemic effects of *H. enneaspermus* are dose-dependent; it is used as a hypoglycaemic agent, and it has the property to cure diabetes mellitus [9]. According to Vennila and Pavithra, *H. enneaspermus* extracts inhibited alpha-amylase and alpha-glucosidase enzymes [55]. Patel et al., [2], reported *H. enneaspermus* extract demonstrated anti-diabetic properties in rat hemidiaphragms [2]. In another study, it was established



that alcoholic extract increased body glucose absorption in the diabetic rat hemidiaphragm than in the control group [35, 56].

Anti arthritic activity

Symptoms of arthritis include inflammation of a joint or joints. Two common types of arthritis are Osteoarthritis and rheumatoid arthritis. Symptoms of both diseases include joint pain and stiffness due to cartilage breakdown [46]. That's why Anti-arthritis drugs are extremely important. Rheumatoid arthritis is largely caused by protein denaturation, which can cause autoantigen overproduction. Various doses of Au NPs were shown to inhibit proteinase activity and denature proteins, respectively 50 g/ml to 150 g/ml. Kamatchi et al., [57], reported Ag and Au NPs showed a strong inhibitory effect on protein denaturation and proteinase activity, their values was 72.24 ± 0.46 and 70.25 ± 0.36 [56-57]. Researchers evaluated Freund's adjuvant-induced arthritis in rats after thirty days of medication at $500 \text{ mg} \cdot \text{kg}^{-1}$, p.o. with alcoholic and aqueous substances of the entire plant, and they finally concluded that both alcoholic and aqueous had the same anti-arthritic property [58].

Anti-allergic activity

Chemical dust, pollen for example, trigger an allergic reaction when they come into contact with the immune system. Physiological reactions to allergic substances can result in a number of diseases [11]. Several studies have demonstrated that flavonoids are anti-anaphylactic and anti-asthmatic. *H. enneaspermus* is also known to have anti-allergic and analgesic properties. Therefore, these chemical moieties may be responsible for *H. enneaspermus* anti-allergic actions. In this study, petroleum ether and ethanol extracts were tested using three techniques: heated plate, tail plunge, and tail flick [9, 59]. Another researcher anti-allergic property was found in the ethanolic extract of *H. enneaspermus* [22].

Hepatoprotective activity

Bhanu et al., [60] examined ethanolic substances of the leaf for hepatoprotective activity at the dose of $\text{mg} \cdot \text{kg}^{-1}$, p.o. in male albino mice for seven days. They estimated important liver enzymes such as superoxide dismutase, lipid peroxidase, and glutathione S-transferase, and they finally concluded that ethanolic extract had a hepato-protective effect [49, 60-61]. Several altered enzymes of liver were restored by *H. enneaspermus* extract, indicating its hepato-protective properties [9].

Anti-ulcer activity

In ancient times, many medicinal plants were used to treat gastric disorders. Medicinal plants contain a wide variety of novel compounds that researchers can use as medications or as biochemical instruments to study disease. A major cause of ulceration is excessive gastric juice and pepsin secretion because of an imbalance between the gastric aggressive factor, oxidative stress, pepsin activity and mucosal defence factor [62-63]. Sahoo et al., 2006 [52] studied extracted alcoholic and aqueous of this whole plant and studied anti-ulcer properties in rats using pyloric ligation (PL) induced gastric ulcer model, and this research showed that a higher pH of gastric juice was found to indicate that alcoholic extract better neutralizes acids. Alcoholic extract also reduces acidity. In conclusion, alcoholic and aqueous extract of the entire plant have anti-ulcer and anti-secretory properties with regulate pepsin and gastric acid secretion activity [19, 6].

Conclusion

The presence of essential phyto-constituents of *H. enneaspermus* makes the plant useful for treating different critical disorders. Therefore, this review will be adjuvant to researchers who have

an interest in examining *H. enneaspermus*. Due to the powerful therapeutic potential and low cost with no side effect of bioactive compounds, natural medicines are gaining in popularity worldwide. The Indian and other cultures have long used traditional ethnomedicines to treat a wide range of ailments, including *H. enneaspermus*. The plants documented in this article are scientifically reported for different pharmacological properties in animal models. As a result, this study provided different products that could be used in a variety of medical and healthcare settings, such as anti arthritic, anti-inflammatory, anti-microbial activity, anti-diabetic, anti-oxidant, and anti-ulcer. As a result of the available data, *H. enneaspermus* can be explored, and its potential in medical care can be explored.

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