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A Review on Therapeutic Potential of *Artemisia nilagirica*

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Abstract

In the modern world of therapy, medicines are in major short of new treatments. It can take many years for a new drug to get through the research and development pipeline to be available in the market and the cost is enormous. And the growing drug resistance caused by the misuse of medications, has rendered several antibiotics and other life-saving drugs useless. Both these trends mean that scientists and pharmaceutical companies are urgently looking for new drug sources and are increasingly turning their eyes to traditional medicine. Major population (80%) in countries of Africa and Asia depends on traditional herbal treatments for their primary health care needs. When adopted outside of its traditional culture, traditional medicine is often called alternative medicine. *Artemisia nilagirica* which is locally known as 'Indian wormwood' belongs to the Asteraceae family and is considered against many ailments as it is possessed to have high content of biologically active molecules and essential oils. The plant has been used since centuries in antimicrobial, antifungal, antibacterial, antifilarial, insecticidal, antiulcer, anticancer, antioxidant and anti-asthmatic activity. This review indicates the collected information on the description of *in vitro* cultivation, phytochemical constituents and therapeutic potential of *A. nilagirica*. The pharmacological studies reported in the present review confirms the therapeutic value of *A. nilagirica*. Presences of wide range of chemical compounds indicate that the plant could serve as a potent material for the development of novel agents having good efficacy in various disorders in the coming years.

Keywords: *Artemisia nilagirica*; Medicinal plant; Therapeutic potential

Introduction

Plants are a significant group among all living organisms, which acts as support system for human beings as well as other living organisms. They have been supporting human civilization through the biologically active compounds which they contain. Plants being a source of medicine has been identified and used from ancient times. In west the use of plants and herbs as a source of medicine is growing, with approximately 40% of the population are reported to use it for the treatment of many diseases. There are about 45,000 species of plants in India are reported to have medicinal properties, with concentrated spots in the region of Eastern Himalayas, Western Ghats and Andaman & Nicobar Island. The officially documented plants with medicinal potential are few but traditional practitioners use more than 6000 plants. India is known as the botanical garden of the world, as it contributes the most to the medicinal herb production. In rural India, over 70 percent of the total population rely on the traditional source of medicine, the Ayurveda [1]. *Artemisia nilagirica* locally known as 'Indian wormwood' belongs to Asteraceae (family). It is a medicinal plant which has been reported to be used over 10 decades and is treating diseases and symptoms like, malaria, inflammation, diabetes, stress, depression, diabetes and many other microbial diseases. *A. nilagirica* is an aromatic herb composed of many volatile oils and biologically important compounds and the plant is perennial and is found mainly in the hilly areas of India [2].

There are many species of the genus Artemisia which are recognized and considered to have many therapeutical importance. The plant has been exploited to derive many biochemical molecules and essential oils which taken into consideration for the treatment of various diseases and ailments. *A. nilagirica* has been reported to have traditional as well as Ayurvedic and homeopathic medicinal uses. Artemisia species are composed of active components which has shown to possess properties to treat parasitic and helminthic diseases. Even after its discovery and exploitation from very long period the knowledge about its phytochemistry and phytotherapy is limited. *A. nilagirica* also has been reported to have efficiency against many neurological disorders, dermal infection and antifungal, antimicrobial, larvicidal, anti-inflammatory activities. The healing potential of this medicinal plant is so efficient that it is still recommended as Ayuvedic drug. It is used for the treatment of human parasites, animals and plants. This review indicates the collected information on the description of *in vitro* cultivation, phytochemical constituents and therapeutic potential of *A. nilagirica* [3].

Plant Description

The plant grows throughout the hilly regions of India. It is tall aromatic shrub. This medicinal herb is erect, hairy, often half-woody. The stems are leafy and branched. Leaves are alternate, large, ovate and lobbed, deeply pinnatisect with small stipule-like lobes at the base, pubescent above, ash-grey or white-tomentose beneath; upper most leaves are smaller, 3-fid or entire, lanceolate. The flowers are small and stand in long narrow clusters at the top of the stem, subglobose heads, in spicate or sub erect or horizontal panicled racemes [4]. They are brownish yellow in colour. Leaves and flowering tops are bitter, astringent and aromatic. The fruit are minute, bracts ovate or oblong. The percentage of oil constituents and the yield of oil vary with the distribution of the plant and also depend on the growth phases (Figure 1).

Figure 1: *A. nilagirica* (Area: Bhubaneswar, Odisha).

Test	Result (Methanolic extract)	References
Alkaloids	++	[5,6]
Amino acids	++	[6]
Carbohydrates	+	[6]
Flavonoids	++	[5-8]
Glycosides	+	[5]
Phenol	++	[6]
Phlobatannins	-	[6]
Quinines	+	[6]
Saponin	+	[6-8]
Tannins	++	[5,6]
Terpenoids	++	[5-8]
Volatile oils	+	[6]

Table 1: Phytochemical screening of *A. nilagirica* extracts [(++) abundant, (+) present, (–) absent].

Phytochemical Screening

The therapeutic potential of herbs or medicinal plants come from its phytochemical constituents. The common phytochemicals that causes effective results on human health care are flavonoids, alkaloids, tannins, glycosides etc. The phytochemical screening of *A. nilagirica* have shown presence of many biologically active chemical compounds such as alkaloids, tannins, flavonoids, glycosides and terpenoids [5]. Another analysis by Ahameethunisa et al. have shown the presence of major chemical compounds such as flavonoids, alkaloids, phenol, tannins, amino acids, quinines and terpenoids. Tannins were reported to be present only in ethanol, diethyl ether and methanol extracts. Volatile oils were present in methanol, hexane and petroleum ether. In hexane and petroleum ether extracts, phlobatannins was present. Also, saponins and amino acid were present in ethanol and methanol extracts with carbohydrates exclusively present in methanol extract. Interestingly, hydrolysable tannins were absent in all the extracts [6]. Another qualitative analysis of the Artemisia species was performed by HPLC, GC-MS and NMR and presence of various secondary metabolites like flavonoids, terpenoids, saponins and polysaccharides were confirmed [7,8] (Table 1).

Bacterial culture	Zone of inhibition (mm diameter)	References
Erwinia sp.	12 ± 0.5	[9]
X. Campestris	12 ± 0.0	
P. syringae	11 ± 0.0	
C. michiganense	12 ± 0.0	
E. coli	22 ± 0.1	[10]
Y. enterocolitica	12 ± 0.0	[9]
B. subtilis	8 ± 0.0	
E. faccalis	-	
K. pneumonia	8 ± 0.0	
S. typhi	10 ± 0.0	
S. aureus	-	
E. acrogens	11 ± 1.0	
P. valgaris	12 ± 1.0	
P. aeruginosa	12 ± 0.0	
S. flexneri	10 ± 1.0	

Table 2: Antibacterial activity- Zone of inhibition (mm diameter) [(±) - mean standard deviation of triplicates, Concentration of extract - 5 mg/disk, (-) - No zone of inhibition observed].

Antibacterial Activity

Medicinal plants possess many therapeutic activities among which antibacterial properties play a major role. In modern world, use of medicines such as antibiotics are causing resistance towards many pathogenic bacteria. Hence there is a need of developing new drugs from herbal or medicinal plant as they contain many bioactive chemical compounds which may have antibacterial properties. Previously many research reports have shown the potential insecticidal [9] and antibacterial activity of *A. nilagirica*. A study used agar disk diffusion method to study the antibacterial activity of *A. nilagirica*. In this method 15 different strains of bacteria were used, to which the extracts of the plant were given, and the activity was observed. The Minimum Inhibitory Concentration (MIC) of *A. nilagirica* extract were performed using two-fold agar dilution method at concentrations ranging from 32 to 512 µg/ml. The result showed positive result for inhibitory activity of gram-positive and gram-negative bacteria except for *Staphylococcus aureus, Enterococcus faecalis* and *Klebsiella pneumoniae*. Various extracts of the plant were used but in that the hexane extract was found to be most effective against all pathogens with lowest MIC of 32 µg/ml, but the methanol extract exhibited the highest inhibition activity against *Escherichia coli*, *Yersinia enterocolitica*, *Salmonella typhi*, *Enterobacter aerogenes*, *Proteus vulgaris*, *Pseudomonas aeruginosa* (32 µg/ml), *Bacillus subtilis* (64

µg/ml) and *Shigella flaxneri* (128 µg/ml) [9]. In another study *A. nilagirica* have shown significant effect against *E. coli* strain with 22 ± 0.1 mm inhibition zone (diameter) [10] (Table 2).

Constituents	R.I. (Retention Indices)	References
α-Thujene	922	[13]
Thujanol	1138	[14]
α-Thujone	1104	
β-Thujone	1116	
Camphor	1145	
Camphene	955	
α-Pinene	941	
β-Pinene	978	[13]
α-Terpinene	1020	[14]
Y-Terpinene	1064	
Myrtenol	1199	[13]
Myrcenone	1142	
p-Cymene	1028	[14]
Limonene	1033	
1,8-Cineole	1035	
Artemisia ketone	1054	[13]
α-Cubebene	1351	
α-Copaene	1379	
β- Caryophyllene	1419	[14]
β- Cedrene	1421	
Y-Curcumene	1480	
α-Cadinene	1538	
Y-Cadinene	1511	
δ-Cadinene	1524	
β-Eudesmol	1634	
Y-Eudesmol	1651	
α-Eudesmol	1654	

Table 3: Essential oils in *A. nilagirica*.

Essential Oil Composition and Antifungal Activity

Many medicinal plants have high concentration of chemical compounds such as flavonoids, terpenes, coumarins and other phytochemicals also. These can be considered as reason for antifungal properties in such plants [11]. Fungal infection in recent times are showing high rates specially among immunosuppressed patients, patients undergoing organ transplants, some other stem cell based therapies and among those suffering from endemic diseases like AIDS and cancer [12].

A study reported the presence of 41 essential oils and other chemical constituents in *A. nilagirica* which was demonstrated by GC-MS and GC-FID methods. In *A. nilagirica* these 41 chemical compounds are 95.9% of total composition of plant [13]. In another study to analyse the chemical composition of *A. nilagirica* and derive their antifungal properties, it has been shown that the plant contained monoterpenoids (79.91%) and sesquiterpenoids (18.25%). The major constituents of these oil were α-Thujone (36.35%), β-thujone (9.37%), germacrene D (6.32%), 4-terpineol (6.31%), β-caryophyllene (5.43%), camphene (5.47%) and borneol (4.12%). The study was performed by using Gas Chromatography- Mass Spectrometry. A relation between the essential oil and antifungal activity of plant was reported. The essential oil exhibited significant antifungal activity against *Rhizoctonia solani* (ED_{50}, 85.75 mg L^{-1}), *Sclerotium rolfsii* (ED_{50}, 87.63 mg L^{-1}) and *Macrophomina phaseolina* (ED_{50}, 93.23 mg L^{-1}) (ED- Effective dose) [14]. The study indicated that *A. nilagirica* is composed of biologically important oils, which can be used to control phytopathogenic fungi infesting agricultural crops and commodities.

Another study showed the precence of essential oils in *A. nilagirica* which were found to possess anti-dermatophytic activity. The minimum inhibitory concentration of the oil was found to be 200 ppm. The oils were fungistatic in nature and had a broad fungitoxic spectrum [15] (Table 3).

Antiulcer Activity

Peptic ulcer is among major concerns in the modern world due to emergence of unhealthy food and food contamination. This disease effects about millions of people, and has high chances to affect almost every individual throughout their life time [16]. *H. pylori* is one of the causal organism of duodenal ulcers and it causes an inflammatory action in the gastric mucosa by increasing the production of cytokines and ultimately causing ulcer formation [17].

A. nilagirica contains many essential oils as mentioned earlier. In a previous study it has been reported that these oils can have antiulcer activity. Limonene is used as flavoring agent in foods and beverages also it is considered as a low toxic component [18]. Hence, studies have shown the gastroprotective activity of this essential oil which is a major constituent of *A. nilagirica* [19-21].

In an *in vivo* study done on rats, it was observed that *A. nilagirica* treatment on ulcer induced rats caused protection against the disease and also there was a significant increase in the mucus content [22].

Antioxidant and Anticancer Activity

Antioxidant activity refers to the potential of a plant to scavenge free radicals that causes cancer and other related mutation. A study shows that the essential oils of *A. nilagirica* possess high DPPH radical scavenging activity (IC50- 26.11 ± 1.04) [23]. Suseela et al. investigated the *A. nilagirica* extracts to show lipid peroxidation inhibition activity. The study also indicated the presence of high total phenolic and flavonoid content (69.71 ± 1.7 mg gallic acid equivalent/g of dry material and 28.41 ± 0.6 mg quercetin equivalent/mg of dry material, respectively). Hence the results indicated the substantial antioxidant activity [24].

A. nilagirica have a significant effect on anticancer and antioxidant activity, which was studied on cancer induced mice. These studies indicate that the anticancer and antioxidant effect of Ethanolic extract of *A. nilagirica* may be due to free radical quenching properties of

sesquiterpene lactones and flavanoid present in ethanolic extract [25]. Devmurari et al. also evaluated methanolic extract of *A. nilagirica* and reported anticancer activity albino mice [26].

Antiasthmatic Activity

Asthma is one of the most common respiratory disease which is caused because of exposure to common allergens present in environment [27-29]. In children asthma can be caused by viral infection of respiratory tract or common cold [30,31]. Reactive oxygen species are associated with the pathogenesis of asthma by inducing bronchial hyper-reactivity and promoting histamine release from mast cells and mucus secretion from airway epithelial cells [32]. Also, free radicals can be considered as potent causative agents of several diseases like bronchial asthma. Free oxygen radicals cause damage to all biological membranes by attacking their protein, lipids, nucleic acids and glycoconjugates [33].

A. nilagirica was given as an antiasthmatic drug to the ovalbumin suspension ingested Wistar rats. The rats shoed an efficient therapeutic property which was confirmed by low WBC count (monocytes and neutrophils). It also decreased the production of nitrate ion hence, minimizing the lung inflammation of asthmatic animal models and relieving bronchial congestion when compared to ovalbumin treated group. The *in vivo* antiasthmatic study, the aqueous plant extracts of *A. nilagirica* at a concentration of 200 mg/kg showed significant activity when compared to positive control (ketotifen) [34].

Larvicidal Activity

Mosquitoes and other parasites and insects can transmit major diseases and ultimately causes millions of deaths every year. Also to our concern, these are getting resistant to synthetic market drugs over the time. Hence, there is a need of alternative form of drug like herbal or Ayurvedic to avoid such kind of concerns.

A. nilagirica as already has been discussed to contain many essential oils and other chemical compounds, based on this factor a study was conducted to examine the larvicidal activity of the plant. In this study the plant extract was proved significant against the action of Aedesal bopictus [35]. Verma et al. also performed one preliminary study on the plant which also showed effective result [36].

In Vitro Cultivation

A. nilagirica as already has been discussed has many proven and unproven therapeutic potential. The first highly effective anti-malarial drug was extracted from *A. nilagirica*. Artemisinin is the drug which is recommended worldwide against malaria [37]. The plant is found in the hilly regions of India and few other Asian countries. But due to their numerous potentials they can be cultivated *in vitro*. Habitat destruction for agricultural purposes and low seed viability is a major concern for the propagation of these plants. For mass multiplication and propagation of this plant tissue culture techniques should be utilized [38,39]. Based on the previous comment a reliable *in vitro* plant culture method has been developed for the *A. nilagirica* (Indian Wormwood). The protocol suggests use of MS medium supplemented with 2.5 µM 6 -benzylaminopurine +7.5 µM 2- isopentenyl adenine for shoot regeneration from nodal derived callus of the plant. Whereas, for rooting from the shoots, the shoots were cultured with quarter strength MS medium supplemented with 10 µM Indole-3-butyric acid. Morphological, cytological studies and ISSR marker analysis showed genetic stability of regenerants. This has important scope for mass scale production of this efficient medicinal plant which could be used for plantation and also as a stock material for pharmaceutical uses [40].

Conclusion

A. nilagirica is widely distributed throughout Asian, especially in India, China, Japan and Afghanistan. In India it is found in the hilly regions of Sikkim, western ghats etc. The plant has many evidences from past for its therapeutic uses in parasitic infections, bacterial and fungal diseases, antiulcer, antinflammation, anti-asthmatic activity etc. It is reported to contain essential oils, sesquiterpene lactones, coumarins and acetylenes and others including many phytochemicals like alkaloids, phenols, flavonoids, and terpenes, which have wide range of pharmaceutical uses. The biochemical and pharmacological studies in the present review confirms the therapeutic efficiency of *A. nilagirica*. It is an important source of various types of compounds with diverse chemical structures as well as pharmacological properties and presence of such a wide range of chemical compounds indicate that the plant could serve as a "lead" for the development of novel agents having good efficacy in various disorders in the coming years. And as the plant is having such therapeutic value, they can get overexploited by the pharmaceutical companies and hence the review suggests the effective way of mass propagation of plant by using plant tissue culture technique.

Future Perspectives

The herb *A. nilagirica* is widely used in Ayurveda for the treatment of various disorders and the results shown are significant. But most of the medicinal effects which has been observed are not yet studied pre-clinically and researchers are not focusing on the effects like antiulcer or antidiarrhoeal activity. In literatures most of the activities of the plant were evaluated using the leaf as well as the extracts of flowering meristems. The root of the plant also has a well-known medicinal activity; it was used in Ayurvedic medicine for the treatment of various disorders, but it is not yet used for studying the pharmacological activities using animal models. Hence, *in vivo* experiments need to be performed by using the root extract of the plant as the history or traditional usage suggests high potential. Furthermore, the development of natural antimicrobials will help to decrease the negative effects of synthetic drugs. Fractionation and characterization of these active compounds will be the future work to investigate. Additionally, the *in vitro* culture and genetic modification of the plant can prevent over exploitation and also may produce novel and more potential variety.

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