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LITERATURE REVIEW

Phytochemical and Pharmacological Studies in *Justicia wynaadensis*, an Ethnomedicinal Plant: A Comprehensive Review

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Abstract

Justicia wynaadensis (Nees) T. Anderson., an ethno medicinal plant of western Ghats is of immense medicinal value. Commonly called Maddhu Thoppu, in Coorg (Kodagu) District of Karnataka, the plant is religiously used by the folkflore to gain immunity against local diseases. J. wynaadensis possesses a wide range of medicinal properties which also include anti-inflammatory, antimicrobial, animutagenic and antioxidant activity. Based on the medicinal properties, J. wynaadensis is a potential plant that that can used as a phyto drug. It has been observed that on 18th day of Aati masa or Kakkada, the medicinal properties of Justicia wynaadensis escalates its peak. Consumption of the plant on this day is believed to provide more health benefits. This review summarizes the literature based on pharmacological, phytochemical and medicinal uses of J. wynaadensis with the purpose of finding gaps demanding for future research and further investigating its potential as an herbal drug.

Keywords

Justicia wynaadensis, Phytochemistry, Antioxidant, Antimicrobial, Anti-inflammatory.

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1. Introduction:

The utilization of traditional plants as a substitute for orthodox medicines is in demand worldwide. Plants particularly folklore medicinal plants are being extensively exploited since thousands of years and utilized in various parts of the world as potential drugs and used in the treatment of local dieases *(Indira MN, 2022; Luzia KAML et al., 2017).* *Justicia wynaadensis* (Acanthaceae family), a subshrub is endemic to Western Ghats and South Malabar Hills in South India, in evergreen forests and on waste lands *(Gamble JS, 1967).* The plant is seen to grow a hedge plant and is cultivated mainly for its significant pharmacological properties. 'Maddhu thoppu', as known in the Kodava Clan, is believed to attain the medicinal property during the months July to August (Hindu calendar month of Karnataka or Adi). Ethno pharmacological survey in Kodagu district has revealed the use of *J. wynaadensis* in treating asthama and immunity *(Lingaraju D et al., 2013).* It is believed to attained maximum medicinal property during the harvest time in the month of August. The plant emits a pleasant aroma and unique flavor. The stem and leaves of *J. wynaadensis* are soaked in water and boiled, the deep purple colour extract thus obtained is used in the preparation of sweet dishes by the local community *(Sudha M et al., 2011).* The Kodava clan believe that on 18th day the plant will be filled with 18 kinds of medicinal properties. This customary practice is believed to ward off a number of diseases, promote immunity and good health throughout the year.

The Karuchair tribes of Tirunelli Forest, Kerala are known to use the plant against rheumatic inflammations *(Sujatha SS and Thrupthi N, 2020).* The current paper is an attempt to review the recent phytochemical and pharmacological significance of *J. wynaadensis.*

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Taxonomical Classification:

Kingdom : Plantae
Division : Tracheophyta
Order : Lamiales
Family : Acanthaceae
Sub family : Acanthoideae
Tribe : Justicieae
Genus : Justicia
Species : wynaadensis

Figure 1: *Justicia wynaadensis*

1.1 Botanical Aspects

J. wynaadensis has a slender stem measuring 2-3 m long with distinct nodes *(Figure 1)*. Leaves are simple, measuring 14-18 x 4-6 cm in dimension, petiolated and arranged oppositely. It is lanceolate, acuminate at both ends, glabrous with 6-8 pairs of veins. The petiole is 1-3.5 cm long. Flowers appear in distant pairs and are borne on drooping spikes 5-10 cm long, axillary and terminal. Rachis may be branched and pubescent. The bracts are hairy, ovate 3-5 mm long; sepals are linear, corolla is12 mm long with hairy throat; style is long with bifid stigma, ovary glabrous. The fruit or capsule is dark brown, 17 mm long, pubescent with ovoid seeds, oblique and minute. Anatomical studies have revealed the occurrence of a purple pigment in the middle cortex of the nodal region of the stem while a blue pigment was seen along upper and lower epidermis of the leaf. The cells of the internodal region are filled with chlorophyll pigment *(Patil, N et al., 2015).*

1. 2. Phytochemistry

Secondary metabolites such as alkaloids, flavonoids, alkaloids, tannins have been reported in the methanol extract of J. wynaadensis *(Abhishek M et al., 2014)*. GC-MS and HPLC analysis of the plant extract has also revealed the presence of various phyto-constituents *(Ponnamma SU and Manjunath K, 2012; Vandana CD and Shanti KN, 2017)*. Dihydrocoumarin, phytol and palmitic acid are the major phyto-constituents along with other compounds such as linoleic acid, stearic acid, squalene, campesterol and stigmasterol. In vitro induced callus derived from leaves and stem explants have revealed the occurrence of a number of phytochemicals when subjected to GC-MS analysis *(Vandana CD, 2018)*. GC-MS study has ascertained the medicinal usage of J. wynaadensis by the local community. These secondary metabolites are known to exhibit antimicrobial, anti-carcinogenic, anti-inflammatory, antioxidant, hypercholesterolemic, antimutagenic and wound healing properties *(Saini and Gangwar, 2023)*.

2. Pharmacological and *in vitro* Studies:

2.1. Antioxidant activities

The phytochemicals such as phenolics, flavonoids and alkaloids have been quantitatively evaluated in the methanolic extract of root, stem and leaves of *J. wynaadensis (John B et al., 2013; John B et al., 2014)*. According to few reports, the total flavonoid content in the plant was estimated to be 178 and 76 µg Quercetin Equivalent (QE) / g, while total phenolics recorded was 0.65 ± 0.08 mg and 0.16 ± 0.1 mg of (GAE)/ g weight of leaf and stem respectively. The antioxidant activity in the leaves was reported as 1.94 mg AAE/ g and that in stem was found to be 1.90 mg AAE/g. The catalase as well as peroxidase activity were also estimated. The potential antioxidant activity is due to the reducing capacity of a compound. The leaves and stem extract of *J. wynaadensis* exhibited a significant reducing effect *(Sudha M et al., 2011)*. Flavonoid was detected in few endophytic strains isolated from *J. wynaadensis*. The high antioxidant property could be correlated to the presence of flavonoids and phenolics. The catalase and peroxidase enzyme activities may also contribute to its scavenging activity. Similar studies on the antioxidant activity of the leaf extracts of *J. wynaadensis* were observed and reported *(Sahana Chavan et al., 2019).*

2.2. Anti-Inflammatory activity

Significant anti-inflammatory activity of plant *J. wynaadensis* comparable to positive control diclofenac has been reported. Oral administration of the alcoholic extracts of *J. wynaadensis* is known to reduce the edema induced by carrageenan by 40 % and 44 % respectively *(Vidyabharathi BP, 2012).*

2.3. Cytotoxicity activity

The leaves *J. wynaadensis* reportedly shows *in vitro* cytotoxicity. The cold aqueous extract of *J. wynaadensis* was found to exhibit maximum cytotoxicity with IC50 value of 20.76 µg/ml and 28.46 µg/ml against the two cancer cell lines namely MCF7 (Breast cancer cell line) and HCT116 (Colorectal cancer cell line) respectively *(Vandana CD and Shanti KN, 2017)*. The aqueous leaf extract of the plant is reported to possess potential genoprotective and cytoprotective properties. Significant results were obtained in a mouse bone marrow micronucleus (MN) assay that was performed to determine the protective effect of Justicia extract against genotoxicity *(Vishma BL et al., 2016)*. GC-MS results of aqueous and methanolic extract of callus and *in vitro* propagated leaf of *J. wynaadensis* have indicated the presence of compounds that show anticancer activity. *In silico* studies of phytocompounds with anticancer activity from *in vivo* and *in vitro* extracts of *J. wynaadensis* has also been experimented *(Vandana CD et al., 2020).*

2.4. Antibacterial activity

The methanol extract of *J. wynaadensis* was found effective against *Klebsiella pneumoniae* MTCC 3384 *(Ponnamma, SU and Manjunath K, 2015)*. The antibacterial activity could be attributed to the presence of volatile components such as phytol and to the fatty acids such as linoleic acid, myristic acid, palmitic acid, and stearic acid present in the plant extract. An effective dose of 3,3′,4′-Trihydroxyflavone (range 32 µg/ml to 1.2 mg/ml) a bioactive compound obtained from the chloroform HPLC fraction of the plant extract could be exploited for the treatment of various infections and used as a potential drug against the bacteria causing diabetic wound and urinary tract infection *(Dorin Dsouza and Lakshmidevi Nanjaiah, 2017).* Flavonoid 7 (3,3′,4′-trihydroxyflavone) extracted from the leaves of *J. wynaadensis* exhibited antimicrobial activity against *Enterocytes faecalis, S. aureus, Enterobacter aerogenes, S. epidermidis* and *K. pneumonia* (*Carneiro et al., 2023).* The leaf extract of *J. wynaadensis* along with the roots of *Aristolochia indica* are known to possess significant anti-biofim activity against *P. aeruginosa,* an opportunistic nosocomial pathogenic bacteria *(Farhan Z et al., 2016)*. A total of nine fungal endophytes belonging to six genera were isolated and identified from *J. wynaadensis,* which exhibited antioxidative and antibacterial activity.

2.5. Wound Healing Activity

3,3′, 4′-Trihydroxyflavone isolated from *J. wynaadensis* showed significant wound healing properties against pathogens associated with diabetes. Rats when treated with 25 and 50 mg/kg BW dose of 3,3′,4′-Trihydroxyflavone exhibited a higher contraction of wound than when treated with normal, diabetic control, and standard drug, framycetin *(Dorin D and Lakshmidevi N, 2017)*. A marked increase in the phyto-constituents and salicylic acid during the monsoons is associated with the health benefits and dermatological curative pro*perties of J. wynaadensis (Karunakaran et al., 2022).*

2.6. Anti cholesterol activity

It was reported that the plant extract of *J. wynaadensis* reduces the concentration of cellular cholesterol and cholesteryl ester. Reports indicate an inhibitory effect on the uptake of ox-LDL by human macrophage cell line. The ethanol extracts of the plant could also be used for the treatment of hypercholesterolemia and atherosclerosis (Subbiah MTR and Norman EJ, 2002).

2.7. In vitro Studies

A rapid method for in vitro regeneration of shoots from the nodal explants of Justicia has been reported on MS media supplemented with 3 mg/l BAP *(Ponnamma S. and Manjunath K, 2012)*. BAP is known to initiate multiple shoot regeneration in Justicia. *In vitro* callus was induced from the leaves and stem of J. wynaadensis on MS media supplemented with different combinations of auxins and cytokinins. 2mg/L 2, 4-D was found ideal for callus induction from stem explants. The aqueous and methanol extracts of the callus and in vitro plant leaf extracts revealed the presence of various phytochemicals when subjected to GC-MS analysis *(Vandana CD et al., 2018).*

2.8. As a food colorant

HPTLC fingerprint profile has been established for the separation of color components which suggests its usage as a natural food colorant *(Nigudkar M et ai., 2014)*. Microwave extraction method was used to examine the microscopic structure and test the stability of the colour extracted from *J. wynaadensis*. Experiments revealed that the purple extract of the plant was sensitive to light, low temperature (4 °C), high temperature (≥50 °C). The extract showed stability at alkaline pH (6.0 – 8.0). These studies further validate the usage of *J. wynaadensis* as a food colorant in neutral or slightly alkaline bakery products *(Patii, N et al., 2015).*

2.9 Synthesis of nanoparticles

The bio-synthesized ZnO-NPs possess significant biological activities. The synthesized nanoparticles were hexagonal wurtzite shaped and exhibited high purity. FT-IR studies confirmed that the ZnO-NPs were free from moisture and also proved the capping of the phytochemicals during the bio-synthesis process. The antimitotic assay reportedly showed inhibition in cell division upon treatment with ZnO-NPs *(Hemanth Kumar NK et al., 2019).* The synthesized silver nanoparticle from leaf extract also exhibited antibacterial, anticancer activity (Cell line A549), anti-inflammatory and anti-diabetic activities *(Lava MB et al., 2021).*

3. Conclusion

The review has brought out the significant research work on various phytocomponents and medicinal properties of *J. wynaadensis* which claims and validates its usage in traditional medicines. Isolation of nanoparticles from Justicia also has proved that it is a good alternative for the synthesis of green nanoparticles and can be significantly used in medicine and industrial application. Studies have indicated that the extracts of in vitro propagated J*. wynaadensis* is rich in bioactive secondary metabolites, hence this technique of micropropagation can be used as an alternative method to mass propagate the plant and obtain more number of plants within a short period. However, there is not enough systematic data on its therapeutic applications. There is still need for significant research on the green synthesis and pharmacological effects of the species. Future studies could aid in the

extraction and isolation of potential bioactive compounds to be used in pharmaceuticals.

Conflicts of Interest:
The authors declare no conflicts of interest

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