Exploring the therapeutic potential of Vanda roxburghii in traditional Indian medicine

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Abstract

People have been captured by orchids for generations due to their extraordinary beauty and variety. In addition to being aesthetically pleasing, these plants are important in traditional Indian medicine. Because of its possible medicinal properties, the *Orchidaceae* family—which includes over 800 genera and 25,000 species worldwide—has attracted a lot of interest. *Vanda roxburghii*, often called *Rasna* and *Vanda tessellata*, is one of the family's most notable species. *V. roxburghii*'s wide spectrum of chemicals has led to its identification as a possible source of therapeutic qualities. Alkaloids, sitosterol, polyphenols, resins, saponins, flavonoids, tannins, fatty acids, and coloring agents are some of these substances. These substances have demonstrated encouraging pharmacological potential, suggesting that they may be used to treat a range of illnesses. Alkaloids and flavonoids found in *V. roxburghii* may help reduce pain and inflammation, while the plant's antibacterial and antimicrobial qualities may aid in the battle against bacterial and fungal diseases. Oxidative stress, a contributing component to chronic illnesses, may be countered by it due to its antioxidant qualities.

Key words: Ethanomedicinal properties, medicinal properties, orchids, Vanda roxburghii

INTRODUCTION

rchids are appreciated worldwide for their unique and beautiful flowers which hold a large commercial and floricultural value. Other than being ornamental, orchids are known to possess medicinal properties as well.[1] Of the various flowering plants, the Orchidaceae family has the greatest diversity, with over 800 genera and 25,000 species spread across the globe.^[2] This family has 186 genera, 1289 species, five subspecies, and 28 variations found throughout India's alpine, coastal, and island regions i.e. it is the most dominant orchid species.[3] India is very rich in orchid genetic resource.[4] The Eastern Himalayan and Peninsular regions are home to the majority of the family's diversity.^[5] A variety of orchids, including Vanda teres, Arundina, Aerides, and Vanda roxburghii, can be found in the forests of the eastern Himalayas.[6] The great significance of orchids lies in their flowers mostly because of their unique appearance and lengthy vase life. Approximately 8% of the global flower cultivation trade is made up of ornamental orchids, which are a multimillion-dollar industry in many Asian nations, such as China, the Philippines, Malaysia, Taiwan, Thailand, and

Singapore. [2] One of the widely cultivated orchids in Southeast Asia and the Indian subcontinent is the Genus *Vanda*. The name of *Vanda* came from an Indian language called Sanskrit, which means that people like these plants by their fragrance, color, and flower shape. [7] *Vanda* species is widely used for medicinal uses. *Vanda* ranks among the top five most popular genera with growers of tropical Asian orchids. *Vanda* is distributed from Himalaya [8] to Sri Lanka and across Southeast Asia to some Pacific Islands. [9] *Vanda* is also shown to possess antiproliferative effects against various types of cancers, including those from choriocarcinoma (cancer of germ cells), lung cancers, and stomach cancers. [10] Compounds commonly found in *Vanda* include bibenzyl derivatives (gigantols), phenanthrene derivatives, phenolic compounds, anthocyanins, alkaloids, steroids, and triterpenoids. [11]

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Received: 21-05-2024 **Revised:** 27-07-2024 **Accepted:** 12-08-2024 Vanda tessellata is the synonym for the *V. roxburghii*. [12] Is is also known as Rasna. [13] *V. roxburghii R.Br.* is considered as the source for Rasna in many of the Dravyaguna materia medica. [14] It has Tikta rasa, Ushna virya, and Katu vipak. It acts on Aamvat, Shula, and Shotha. [15] One orchid with several therapeutic qualities is *V. roxburghii*, one of the many plants used for medical purposes. [16] This orchid is epiphytic. Native American health systems like Ayurveda [17] and regional traditional medical practices have utilized vetiver plants. There have been reports on the many pharmacological characteristics of *V. roxburghii*, including its aphrodisiac, hepatoprotective, antidiarrheal, and anti-inflammatory effects. *V. tessellata* has shown an alkaloid, sitosterol, resin, saponin, polyphenol, flavonoids, tannins, fatty acids, coloring agents, etc. in the phytochemical screening process. [18]

V. tessellata (Roxb.) Hook. Ex G. Don. (syn. V. roxburghii R. Br.), of the family Orchidaceae is popularly known as gray orchid. It is a widely cultivated horticultural and epiphytic orchid that forms huge clumps on tree trunks or rocks in thick forests and sacred groves. V. tessellata were observed frequently on the trees, such as Mangifera indica and Diospyros melanoxylon growing along the roads in the periphery of the lagoon and adjacent villages.[19] It is found all throughout Bangladesh.[20] It grows on guava, blackberry, and mango trees throughout Bangladesh.[21] Traditional Indian medicine, Ayurveda, provides a detailed description of the therapeutic characteristics of *V. roxburghii*. [22] There is a traditional story about this plant that claims its root can reduce inflammation. Historically, the root of *V. roxburghii* has been utilized to cure a variety of conditions, including piles, hiccups, bronchitis, fever, and dyspepsia.^[23] In addition, the root is applied externally to treat nervous system illnesses, rheumatism, and related conditions. [24] To reduce fever, the leaves are crushed, and the body is covered in their paste; The juice is placed into the ear to treat otitis. Numerous active chemicals were found in V. roxburghii through phytochemical research. These included melanin, 2,7,7-tri methyl bicyclo-heptanes having aphrodisiac and antifungal properties, as well as heptacosane and octacosanol. Significant pharmacological properties of the plant have also been found, including wound healing, [15] anti-oxidant, anti-inflammatory, anti-arthritic, anti-bacterial activity, anti-fungal activity, hepatoprotective activities, and anti-cancer activities.^[25] We have already documented the antinociceptive properties of methanol and water-based extracts derived from *V. roxburghii* leaves.^[23] The plant's root is claimed to offer anti-inflammatory, anti-bronchitis, antirheumatic, anti-abdominal, anti-tremor, anti-pyretic^[26] and anti-infectious properties against tuberculosis and bacterial infections, among other purposes in traditional medicine.[27] Also the plant possesses some anti-fungal activity. [28] To lower fever, the crushed leaves are administered topically as a paste; the juice is then given topically to treat otitis and other inflammatory disorders.[29]

Furthermore, the study shows that the species of *V. roxburghii* has shown a detailed sequential role with respect to

micro-morpho-anatomical developments in the tissue culture raised plantlets of *V. roxburghii* toward altered environmental conditions for their epiphytic adaptations.^[30] The petroleum ether extract of *V. roxburghii* shows the Hepatoprotective activity.^[31]

Reosto is a polyherbal formulation and is a rich source of phytoestrogens and calcium. Reosto is recommended for the management of postmenopausal osteoporosis and contains the powder of *V. roxburghii*.^[32]

A particular histochemical staining methodology and structural advancements are employed in the investigation to detect cell wall depositions in *V. tessellata* epiphytic adaptations. It offers precise visualization of the location of the cell wall by light microscopy, as well as the ability to determine mechanical support and water absorption and retention capacity. Significant variations in the characteristics of the cell wall composition are visible at different developmental stages; roots produced *in vitro* exhibit less deposition.^[33]

MORPHOLOGY

V. roxburghii having stem 1-2 feet, climbing leaves 6-8 inches, narrow complicate, peduncle 6-8 inch., sepals and petals yellowish green or bluish except from clathrate brown nerves, margins white, spur conical.[34] V. Roxburghii is a perennial epiphytic orchid that grows to a height of 30–60 cm. It has a sturdy stem covered in a sheath and scandent leaves, a raceme-like inflorescence, and a flower cluster of five to twelve. The sepals are yellow and tessellated with brown lines, the petals are yellow with brown spots that are shorter than the sepals, the lip is purple, the column is white and 0.9 cm long, and the capsule is ribbed and 7.5–9 cm long. The monopodial orchid *V. roxburghii* is found in tropical regions, such as India, Nepal, China, Bangladesh, Sri Lanka, and Myanmar, with an elevation range of 200-600 m. Twice a year, in early summer (April-May) and early winter, the plant blooms.[35]

PHARMACOGNOSTIC EVALUATION OF VANDA ROXBURGHI

Microscopic Characteristics

The leaf's outline is V-shaped, and its cuticles are $5-9~\mu m$ thick on both surfaces. Its epidermis is uniseriate, parenchymatous, and its layer of densely walled fibers is uniseriate. Amphistomatous stomata can be either paracytic or anisocytic, and they can be actinocytic, paracytic, or anisocytic on the abaxial side. Chloroplast density, homogeneity, and lack of differentiation characterize the mesophyll.

The circular stem has a cuticle that is $3-4\,\mu m$ thick, a uniseriate epidermis, and a hypodermis that is 2-3 layers thick. Ground tissue is parenchymatous, including triangular air gaps enclosed by circular to elongated oval cells. They include raphides, vascular bundles, and water-storage cells with thick walls.

The plant features a velamen made up of polygonal cells, circular roots, an exodermis-containing cortex, and a 21–24-layer cortex with water-storing and thin-walled parenchymatous cells. There are sporadic thin-walled passage cells dotted throughout the uniseriate, O-thickened endodermis. There is a vascular cylinder and pith on the stele.^[36]

The synonyms and taxonomical arrangement for *V. roxburghii* are shown in Table 1.

CHEMICAL CONSTITUENTS OF V. ROXBURGHII

The plant contains long-chain aliphatic substances, resins, fatty oils, tannins, β - and γ -sitosterol, alkaloids, glycosides, and coloring agents. In addition there are components including 17- β hydroxy-14,20-epoxy-1-oxo-[22R],^[37] heptacosane, octacosanol and acetyl tetracosyl ferulate^[38] and also the 17-ketosteroid as anti-microbial agent.^[39] Furthermore, there is a compound called O- β -D-glucopyranosyl-3 β -[O]-5,24 with melanin and dienolide and alkyl perulate.^[40] Steroids are known to have inflammatory properties. Research has shown that β -sitosterol possesses qualities that can help reduce inflammation and fever.^[41]

Roots contain tetracosyl ferrulate and β -sitosterol-D-glucoside [1, 4]. Heptacosane (C27H56) containing traces of the higher homologue (C29H60) and octacosanol (C28H58O)

Table 1: The synonyms and taxonomical arrangement of *Vanda roxburghii*

Taxonomical classification		Synonyms	
Kingdom	Plantae	Language	Common names of drug
Subkingdom	Tracheobionta	Sanskrit	Rasna, Vandaka
Superdivision	Spermatophyta	Bengali	Rasna
Division	Magnoliophyta	Gujarati	Rasna
Class	Liliopsida	Hindi	Vanda
Order	Orchidales	Kannada	Banda Nike
Family	Orchidaceae	Telugu	Van Danika
Genus	Vanda		
Species	Roxburghii		

containing traces of higher homologue (C30H62O), (C32H66O) were identified.^[42]

TRADITIONAL USES

Some plants, such as V. roxburghii and V. tessellata have been documented for their medicinal value.[43] V. Roxburghii extract has shown effects, in promoting wound healing in rats.[22] It also helps to enhance contractions and acts as a bronchodilator aids digestion and purifies the blood. This extract is commonly used for treating conditions such, as Edema, fever, [44] gout, rheumatic diseases, [45] asthma, and stomach discomfort.[46] The Unani and Ayurvedic medical traditions also mention the plant's therapeutic qualities. The plant contains an amount of compounds and exhibits strong antioxidant properties. In addition, it shows promise, in inhibiting cholinesterase activity.^[47] Furthermore, *V. tessellata*, one of the most recognized orchids of Ayurveda advocated for its use as an alexiteric, diseases of abdomen, hiccough, and is known to heal tremor as per Ayurveda. V. tessellata was also used as a laxative by Unani practitioners.[48]

The traditional use of V. roxburghii root in Bangladesh includes its use as a brain tonic and for treating nervous system disorders, such as Alzheimer's disease. Native Americans have historically relied on Vanda for its anti-inflammatory properties. Indian Vanda has demonstrated antiproliferative effects on malignancies, such as choriocarcinoma, lung, and stomach cancers. The plant is also used to relieve conditions, such as sprains, lumbago, back pain, earaches, blisters on the scalp, toothache, piles, and bronchitis.[10] In addition, it is employed in the treatment of bone fractures, utilizing a typical procedure that involves using a paste prepared from Phoenix loureiroi's sensitive buds and aerial roots to plaster fractures. In Nagpur, India, Vanda leaves are ground into a paste and used to treat otitis media and fever, [49] while a root compound decoction is administered for various conditions, such as mental disorders, ear infections, bone fractures, [50] fever, and even snake bites.[51] V. tessellata was used by FH1 for treatment of rheumatic pain. The anti-arthritic action of the plant has been reported.[52] Powdered form of roots is considered as an antidote for poisoning. The whole plant is used in sciatica.^[53] The root of *V. tessellata* is also traditionally used as an antidote for scorpion stings and a remedy for bronchitis and rheumatism.^[54]

PHARMACOLOGICAL ACTION FOR WOUND HEALING ACTIVITY

Wound healing properties of the extract of *V. roxburghii* is investigated, as it is reported in Ayurveda as a strong candidate of medicinal plant used in anti-inflammatory, antiarthritic, and in the treatment of otitis externa.^[55]

Hossain *et al.* Evaluated the extract of *V. roxburghii* for wound healing^[56] studies utilizing the excision wound model in rats. After being thoroughly washed and allowed to air dry, utilizing a typical procedure that involves using a paste prepared from *P. loureiroi's* sensitive buds and aerial roots to plaster fractures. After filtering the materials, the clear filtrate was utilized in these investigations. Every day, fresh paste was made ready for use. Twenty inbred female Sprague Dawley rats (n = 20) in good health, weighing between 180 and 200 g, were used in the study.

The animals were anesthetized with intravenous ketamine hydrochloride before and during the creation of wounds. On the shaved dorsal fur, 2.5 cm long and 0.2 cm deep incisions were created using toothed forceps, a surgical blade, and pointed scissors. Ten days passed as the wounds remained open. The control group received topical application of carboxymethyl cellulose (CMC 1%), while the experimental group was treated with *V. roxburghii* leaf extract (150 mg/kg) dissolved in water. Granulation tissue started to develop on the wounds on the 11th day; these were removed and weighed. The tissue was dried, and its dry weight and protein content were measured. Hydroxyproline and hexosamines in the tissue were determined using acid hydrolysate. On the 1st, 5th, and 11th days, the areas of the wounds were measured using transparencies and a permanent marker with recordings and measurements on graph paper for both groups.

The measurement of the wound area indicated a decrease, in the group that received the extract compared to the control group who were treated with CMC in water. The test group also showed increases in both dry weights of the granulation tissue. In the test group, the obtained granulation tissue had somewhat higher protein content. Furthermore, an increase in the levels of hydroxyproline and hexosamine were noted in the animal tissue within the experimental group.^[34]

ANTIOXIDANT PROPERTY

Prakash *et al.* explained about the antioxidant property of *V. tessellata*. About 500 g of *V. tessellata*^[57] powdered leaves were put in the Soxhlet apparatus and extracted using ethanol and CHCl₃ in turn until the extraction process was finished. *V. tessellata* also possesses the anti-oxidant property in petroleum ether extract that can be evaluated using *in vitro* method. After the hot extract was filtered, the resulting extract was vacuum-distilled at low pressure to eliminate all traces of the solvent, and it was then dried in a desiccator.

The investigation's goal was to assess the antidepressant and antioxidant attributes of extracts from *V. tessellata* leaves. The rats were split up into 4 groups, with 6 rats in each. Saline, imipramine, *V. tessellata* ethanol extracts, and a forced swim test were administered to the mice. Before the study, the animals were left hanging on a table for 60 min. In comparison to chloroform extracts, the ethanol extracts

demonstrated the highest total polyphenol concentration, according to the data. The extracts' antioxidant qualities can be utilized to treat and prevent a number of illnesses linked to free radical damage. Most plants have the ability to scavenge radicals because they contain flavonoids, which contain hydroxyl. Scavenging and chelating reactions are the ways in which flavonoids work. The main results of TPC and TFC of the extract of *V. tessellata* leaves showed that the extract had antioxidant properties. The ethanol extract has a higher content of flavonoids and polyphenols than the CHCl₃ extracts. The *V. tessellata* ethanol extract was chosen to examine its potential antidepressant effects. The study emphasizes the significance of plant antioxidant activity and the potential medical applications of these compounds.^[59]

Uddin *et al.* conducted a study to assess the antioxidant potential of several extracts of *V. roxburghii*, with a particular emphasis on scavenging hydroxyl radicals and inhibiting lipid peroxidation. With an absorbance of 1.39, the chloroform extract exhibited the maximum activity at 100 μg/mL. In DPPH radical scavenging, the IC₅₀ values for catechin and chloroform extract were 4.55 μg/mL and 5.76 μg/mL, correspondingly. In addition, the most effective method of preventing brain lipid peroxidation was the chloroform extract. Because *V. roxburghii* contains polyphenols and may have antioxidant properties, it has long been used in Bangladesh to treat Alzheimer's disease.^[47]

ANTI-INFLAMMATORY ACTIVITY

It was discovered that a number of orchid family members had anti-inflammatory properties.^[60] *V. roxburghii* has demonstrated significant anti-inflammatory action^[61] in carrageenan-induced edema in rats and mice.^[62] The plant root, *V. roxburghii* roots, was dried into a coarse powder and steeped in methanol for 5 days to produce the extract of crude methanol and its CHCl₃ fraction. The study used Swiss albino mice for the experiment. After filtering and evaporating the crude methanol extract, 28.37 g of dry extract were obtained. To isolate the compounds, the chloroform fraction was separated using chloroform and then put through column chromatography. The molecules that were separated out were identified as dihydroxyconiferyl dihydro-p-coumarate, gigantol, syringaldehyde, and vanillin.

There were seven groups of mice, each including four animals. The mice were carefully weighed, and the doses for the test samples were calculated. The mice were injected with a carrageenan suspension that had been diluted with usual saline, and the test materials were dissolved in 1% Tween 80 in regular saline. The anti-inflammatory activity was expressed by computing the percentage of inhibition of paw edema. The methanolic extract of *V. roxburghii* roots had a chloroform-soluble fraction from which four compounds were extracted. The Rf values of the compounds were 0.5, 0.6, 0.9, and 0.76. The substances were identified as syringaldehyde, gigantol,

dihydroxyconiferyl dihydrocoumarate, and vanillin. At a dosage of 100 mg/kg, gigantel showed the greatest effectiveness of all the drugs, with edema inhibition ranging from 26.2% to 41.2%. At the same dosage, edema inhibition ranges for syringaldehyde, vanillin, and dihydroconiferyl dihydro-p-coumarate were 14.1–30.1%, 13.7–31.2%, and 12–33.2%, respectively. Among the compounds isolated from the chloroform fraction, gigantol was shown to be the most potent anti-inflammatory agent. Vanillin and syringaldehyde have been shown in earlier research to possess anti-inflammatory and antioxidant qualities.^[2]

A study on the anti-inflammatory^[17] effects of aqueous extracts from V. roxburghii and Commiphora wightii was carried out by Shekhawat and Sisodia After the extracts were made and analyzed, it was discovered that when rat hind paw edema was induced using carrageenan, V. roxburghii extract outperformed the conventional drug indomethacin in terms of anti-inflammatory activity management. After being extracted from C. wightii and V. roxburghii leaves, the gum resin was coarsely powdered and shade-dried before being passed through a 40-mesh filter. After that, the extract was dried using a rotary evaporator or water bath. The final extract was labeled, sealed, and placed in an amber-colored bottle. The percentage yield (w/w) was then computed. Four groups of six rats each—Control, Standard, C. wightii, and V. roxburghii were randomly assigned. Oral administration of 0.3% CMC was administered to the control group, whereas C. wightii (100 mg/kg), V. roxburghii (100 mg/kg), and the usual medication (Indomethacin, 10 mg/kg) were administered to the other three groups. The outcomes demonstrated that both extracts were more effective in treating carrageenan-induced hind paw edema than the recommended drug, indomethacin. These results offer strong proof of the anti-inflammatory properties of C. wightii and V. roxburghii. [63]

ANTI-NOCICEPTIVE ACTIVITY

According to a study by Uddin et al., V. roxburghii's root possesses less toxic and anti-nociceptive qualities. The substance whether produced from plants or not, may be useful in treating pain. Methanol was used to extract the powdered V. roxburghii root after it had been air-dried. The outcome extract was separated into petroleum ether, CHCL₃, ethyl acetate, and water extracts after being concentrated under vacuum. For the experiment, adult Swiss albino mice were employed. Alkaloids, glycosides, flavonoids, phenols, saponins, tannins, and steroids were examined in the extracts. In addition, using three distinct mouse models, the study looked at the root's anti-nociceptive qualities. The Formalin test examined pain responsiveness, whereas the acetic acidinduced writhing test measured anti-nociceptive action. Central analgesic activity was assessed using the hot plate test. To evaluate each root extract's potential for cytotoxicity against brine shrimp nauplii, in vitro tests were carried out. The findings demonstrated a strong anti-nociceptive effect of the root extracts on the nauplii, offering important new information on the possible anti-nociceptive qualities of *V. roxburghii* and its possible uses in pain relief therapies.^[46]

The study by Chowdhury et al. evaluated the cytotoxic and antinociceptive properties of methanol and waterbased V. tessellata leaf extracts. The leaves were ground into powder and soaked in methanol for a week, yielding 1.6% w/w (VTM) and 1.2% w/w (VTA). Swiss albino mice were used to test the extract's effects. The extract showed a low toxicity profile, with there include flavonoids, tannins, phenolic chemicals, and saponins. The writhing test caused by acetic acid in mice showed a dose-dependent reduction in pain, with the highest inhibition observed at higher doses. In a dose-dependent way, the leaves' antinociceptive activity lengthened the latency duration of thermal stimuli elicited by hot water. Methanol and aqueous extracts had LC₅₀ values of 574.32 and 430.41 µg/mL, respectively. According to the study's findings, V. roxburghii leaves have low cytotoxicity and a strong, dose-dependent analgesic effect that has been supported by a number of pain models. The research validates the traditional therapeutic application of *V. roxburghii* for the relief of pain conditions and calls for more investigation to isolate and identify the plant's bioactive constituents.^[64]

ANTI-DEPRESSANT ACTIVITY

Prakash *et al.* studied the antidepressant properties of *V. roxburghii* leaves extracted with ethanol and chloroform after drying and powdering. The extract underwent filtration, distillation, and desiccator drying. The study found that the ethanol extract significantly shortened the animals' periods of immobility compared to the control group. The leaves also demonstrated good protection and dose-dependent antidepressant efficacy. The antioxidant qualities of the leaves confirm their antidepressant action, as oxidative stress is a major issue in depression patients. The polyphenol and flavonoid components of the leaves play a crucial role in controlling depressive disorders. The study provides valuable insights into the potential of *V. roxburghii* leaves in treating depression.^[59]

ANTI-MITOTIC ACTIVITY

Using plant-based model systems, Parmar *et al.* studied the antimitotic activity of plant extracts from different herbs, plants, and trees in India. They made aqueous extracts from a variety of plant parts, including the fruit, bark, seed, root, stem, and leaf. Water was used as the adverse model and vincristine as the constructive model. In the *Allium* test, *Allium cepa* was cultivated in water under carefully monitored laboratory conditions, and onions were subjected to aqueous plant extracts containing 40 mg/mL for a whole day. A formula was used to compute the mitotic index. The National Center for Cell Science in Pune, Maharashtra, India is where the human

colorectal cancer (HCT-116) cell line was obtained. HCT-116 cells that were growing exponentially were used in the investigation, along with doses of *Terminalia bellirica* (1:250 and 1:100) and DMSO as a 24-h vehicle control system. To evaluate the inhibitory effect on cell proliferation, the MTT test was utilized.

To quantify cell viability, the study also employed an assay for the exclusion of trypan blue dye. Annexin V/PI staining was utilized to determine the percentage of plating efficiency and to assess apoptosis. The researchers discovered that *T. bellirica*, *Bauhinia variegata*, *V. roxburghii*, *Cassia angustifolia*, and *Aconitum heterophyllum* were the most effective in inhibiting germination using the *Vigna radiata* seed germination inhibition assay. *C. angustifolia*, *V. roxburghii*, *B. variegata*, *T. bellirica*, and *A. heterophyllum* aqueous extracts considerably reduced the mitotic index in the *A. cepa* root tip assay and entirely inhibited seed germination in the *V. radiata* assay.

T. bellirica was selected for additional testing because the MTT test confirmed its high efficacy among these extracts. Morphological examination of the HCT-116 cells showed significant morphological changes brought on by T. bellirica. [65]

ANTI-CONVULSANT PROPERTY

A study on the anticonvulsant properties of *V. roxburghii* root extract (SVR) on seizures in mice generated chemically and electrically was carried out by Pathan and Ambavade To evaluate toxicity, SVR extract was administered to the mice at dosages of 30, 100, 300, 1000, 2000, and 5000 mg/kg. For the Pentylenetetrazole (PTZ), Maximal Electroshock (MES), and Picrotoxin (PTX)-induced convulsion models, the animals were split up into groups of six mice each. Oral administration of *V. roxburghii* (SVR) extract at doses up to 2000 mg/kg has been shown to be safe and did not result in any fatalities. Mice given a single intraperitoneal dose of PTZ showed convulsions; however, SVR treatments at dosages of 25, 50, and 100 mg/kg did not appreciably change the animals' latency to THLE. SVR, on the other hand, reduced mortality to 50% and 83.33% at 50 and 100 mg/kg, respectively.

Mice given a single subcutaneous injection of PTX showed signs of convulsions. SVR (100 mg/kg) significantly reduced the incidence of THLE and markedly extended latency to clonic convulsions without having a significant effect on mortality. SVR (50 mg/kg), dramatically decreased THLE incidence while having no discernible effect on death or clonic convulsions. There was no significant effect of SVR (25 mg/kg) on death, THLE incidence, or latency to clonic convulsions.

In conclusion, *V. roxburghii* has anticonvulsant action against convulsions in mice generated by PTX, MES, and PTZ.^[66]

ANTIMICROBIAL ACTIVITY

Gupta et al. conducted an in vitro study to examine the antibacterial activity of solvent extracts of *V. tessellata* roots. The plant material was gathered from the Southern Ravalli Hills of Rajasthan, near the Phulwari Ki Naal Wildlife Sanctuary. Bacterial isolates from Candida albicans, Aspergillus niger, Proteus mirabilis, Staphylococcus aureus, Klebsiella pneumoniae, and Escherichia coli were used in the investigation. The roots were broken up into little bits, ground, and dried in the shade. Petroleum ether, CHCl, ethyl acetate, acetone, CH,OH, and hexane were among the organic solvents that were utilized in the Soxhlet procedure to extract the coarse particles. The extracts were utilized as an antibacterial agent after being distilled out at atmospheric pressure over a water bath. The antibacterial action of the crude extracts was evaluated using the agar-well diffusion technique. The findings demonstrated the extracts' strong antibacterial efficacy against both bacteria and fungus. The study emphasizes V. tessellata roots' potential as an antibacterial agent.

Using the Akinpelu and Kolawole methods, the study calculated the minimum bactericidal concentration (MBC) and minimum inhibitory concentration (MIC) of plant extracts from the roots of V. tessellata. At different doses, the crude extracts were added to nutritional agar that had been pre-sterilized for fungi and to Sabouraud Dextrose Agar for bacteria. After that, the extracts were incubated for a maximum of 72 h at 25°C for fungus and 24 h at 37°C for bacteria. The extract may be bacteriostatic at lower dosages but bactericidal at higher concentrations, as suggested by the fact that the MIC values were lower than the MBC values. The extract with the highest % extractive value was CH₂OH, followed by CHCl₂, acetone, petroleum ether, and ethyl acetate. The findings imply that the extracts in acetone and ethyl acetate showed a good zone of inhibition, perhaps as a result of the solvent-insoluble nature of the active component. The extractable material's activity level was also greatly affected by the extraction solvents, indicating that organic extracts exhibited more antibacterial activity. With a 17-28 mm Zone of Inhibition, the compounds VR-1 and VR-2 that were isolated from V. roxburghii exhibited potent antibacterial activity.[24]

Bakul Bhattacharjee *et al.* conducted a study on the antimicrobial activity of *V. tessellata* plant extracts from Rajshahi, Bangladesh. The plant was used as a source for extract preparation, and the extracts were extracted using organic solvents. The extracts were tested on clinical strains of bacteria and fungal organisms, including *Penicillium* sp., *Rhizopus* sp., and *A. niger*. The study found that the chloroform extract showed the highest inhibition against pathogenic bacteria, while the hexane extract had the second-highest zone. The plant extract also showed antimitotic activity against tested fungal isolates. Bioactive substances including alkaloids, terpenoids, flavonoids, phenols,

tannins, steroids, and glycosides were found, according to the phytochemical activities. One possible option for bioprospecting for antibiotic and antifungal medications was the chloroform extract.^[67]

Bhatnagar et al. found that V. tessellata, Vitex negundo, and Holarrhena antidysenterica are popular as medicine among tribal communities of the Mayurbhanj adjoining area. These tribal groups use plant bark paste and leaf juice against dyspepsia and dysentery. The study found that the extracts of leaf and bark showed significant zone of inhibition against various human pathogens, including Cyanobacterium, S. aureus, Vibrio cholera, Shigella flexneri, E. coli, Pseudomonas syringae, Klebsiella sp., Citrobacter freundii, Proteus vulgaris, Acetobacter, and Salmonella enterica Typhi. The ethanolic plant leaf extract showed the highest inhibition in both V. tessellata and V. negundo, but in the case of *H. antidysenterica*, the highest zone of inhibition was observed in the n-hexane extract. The highest zone of inhibition was found in S. flexneri, Vibrio cholera, and E. coli with zones of inhibitions of 50-56, 40-56, and 43-53 mm, respectively. The bark paste of *V. tessellata* contains alkaloids, tannins, and arthroquinones, which might be responsible for the inhibition zone of the human pathogen Vibrio cholerae. In V. negundo, the bark is mainly used for dysentery, with the bark paste being co-related to the presence of terpenoids used in n-hexane and ethanol extract. In conclusion, the ethnobotanical survey reveals the potential of these plants as potential sources for new drugs, potentially effective against dysentery and combating antimicrobial resistance.[16]

PROTECTIVE EFFECTS OF V. TESSELLATA LEAVES

The hepatoprotective and nephroprotective qualities of *V. tessellata* leaves, as well as the toxicity of methotrexate in rats, were the subjects of an investigation by Chaudhuri *et al.* According to the study, the extract reduced methotrexate-induced liver and kidney damage in rats. This finding raises the possibility that methotrexate's side effects might be lessened without sacrificing its therapeutic benefits by utilizing *V. tessellata* leaf ethanol extract. This may enhance the therapeutic effectiveness of toxicities caused by methotrexate on several organ systems.^[6]

NEURO-PROTECTIVE EFFECT

Using a rat model, Yash conducted the study to assess the neuroprotective properties of *V. roxburghii* against Alzheimer's disease. The findings demonstrated that the ethanolic extracts raised levels of antioxidant enzymes, reduced lipid peroxidation, and enhanced behavioral activity and spatial navigation tasks. These results imply that this plant could be useful as Alzheimer's disease therapies.^[68]

ANALGESIC ACTIVITY

The analgesic effect of the V. tessellata (Roxb.) Hook leaf ethyl acetate extract was assessed in a study by Kamalutheen on mice. The plant material was gathered, dried, cleaned, and then ground into tiny fragments. Organic solvents were used to extract the extracts, which were then distilled. The 2% gum acacia solution in distilled water was used to dilute the dry extract. The study employed adult Swiss albino mice and adult albino rats. The night before the experiment, the animals were split up into eight groups and given a nighttime fast. The Karber's approach was used to dose gum acacia extracts administered intraperitoneally. At 24, 48, and 72 h, the animals were checked for alterations in behavior and signs of mortality. Even after 72 h, no deaths were reported despite the fact that experimental animals reacted to the drug's ethyl acetate extract at a minimal dosage of 200 mg/kg, according to the toxicity research. The analgesic action was measured using a tail flick device. The results show that the highest amount of ethyl acetate extract that can be consumed safely by an individual is 3 g/kg body weight. The leaves of V. roxburghii were extracted using ethyl acetate, and the results showed a dose-dependent analgesic impact as well as significant analgesic effectiveness against thermally uncomfortable stimuli.[69]

Shekhawat and Sisodia looked at the plant *V. roxburghii*'s potential as an analgesic. Using analytical grade solvents, the gum resin from *V. roxburghii* leaves was collected, ground into a powder, and extracted. The extract was kept for pharmacological research and phytochemical analysis in an amber-colored container. Four experimental groups of rats were randomly assigned, and the control group's only treatment was 0.3% CMC.

Acetic acid was injected intraperitoneally into the rats, and during 10 min of observation, the number of writhes the rats exhibited was recorded. The % inhibition was calculated as the average writhes in the control group minus the writhes in the drug extract group divided by the writhes in the control group times 100%. Rats were divided into four groups: Group I served as the control group, Group II as the standard group, and Groups III and IV received oral treatments with extracts from C. wightii and V. roxburghii. C. wightii and V. roxburghii extracts considerably accelerated the rats' response times, according to Eddy's hot plate test. In the rats' acetic acid-induced writhing test and Eddy's hot plate method, the results demonstrated that both C. wightii and V. roxburghii extracts demonstrated strong analgesic effectiveness versus conventional medicines, such as indomethacin and tramadol.[70]

ANTI-BACTERIAL ACTIVITY

Ramana et al. conducted an investigation on the epiphytic roots and leaves of V. tessellata from Penchalikona to

evaluate the antibacterial activity of the disc diffusion method *in vitro*. To create plant extracts, the leaves and roots were separated, sterilized, and then dried for a period of 40 days. For *in vitro* antimicrobial investigations, they were subsequently pulverized and dissolved in methanol, ethyl acetate, and n-hexane. Using a Soxhlet extraction apparatus, a stock solution of extract was created and diluted with various quantities. After being impregnated into Mueller Hinton Agar plates, the antibiotic discs were tested in triplicate and then incubated for 24 h at 37°C. Merely methanol extract was utilized to prepare the negative control.

Using the disk diffusion method, the study assessed the antibacterial action of *V. tessellata* (NV01) leaf and root extracts against a variety of pathogens, including as *Pseudomonas aeruginosa*, *Bacillus subtilis*, and *E. coli*. According to the findings, *Acampe praemorsa* and its ecotype caused a larger zone of inhibition on all four bacterial strains, but *V. tessellata* was the most efficient. All four isolates were inhibited by ethyl acetate leaf extracts of *V. tessellata*, *A. praemorsa*, and its ecotype. While *V. tessellata* ecotype inhibited *S. aureus*, *V. tessellata* and its ecotype demonstrated efficacy against *E. coli* and *P. aeruginosa*.^[71]

Behera et al. studied the Antibacterial activities of crude extracts of orchid species (V. tessellata) obtained by four different solvents were studied against some clinically significant human pathogens. The result revealed that all crude extracts showed antibacterial activity in varying degrees inhibiting at least one or more test pathogens. Among the solvents, di-ethyl ether extracts showed significant antibacterial activity against all the test pathogens followed by butanolic, chloroform, and methanolic extracts. The MIC value of different extracts ranged from 3.5 to 25 mg/mL. The results indicated that the crude extracts were bactericidal in action. The antibiogram pattern of the pathogens revealed multiple antibiotic resistance indexes of 40-60%. The activity of different extracts was compared with standard antibiotics, in terms of zones of sensitivity. The findings suggest that ethnomedicinal orchids could be used as an alternative source of the therapeutic agent in the near future. [60]

Khasim *et al.* studied the plant *V. tessellata*, for its antibacterial properties, which grows in West Bengal's Hijli Forest, India, was employed. Before being combined with solvents including benzene, chloroform, acetone, methanol, ethanol, and water, the plant components were cleaned, dried, and pulverized. After filtering and evaporating the extracts, a semi-solid residue was produced. Both Gram-positive and Gram-negative bacteria were used. A first examination of the phytochemicals showed that the plant extract ethanolic had a good zone of inhibition against the tested bacteria. While water extract exhibited reduced effectiveness against all tested microorganisms, other extracts showed action against *Micrococcus luteus*. There were secondary metabolites present, including phenols, tannins, alkaloids, terpenoids, flavonoids, steroids, and glycosides. The plant has the

potential to be used in folklore medicine and bioprospecting for antibiotics. Additional research on phytochemicals is required to identify the specific kind of component exhibiting antibacterial properties.^[72]

Disc diffusion and dilution techniques were used to investigate the antibacterial activity of root extracts from V. tessellata in vitro. The roots underwent separation, surface sterilization, three rounds of washing in sterilized distilled water, 40 days of shade drying, and powdering. Three distinct solvents were used to dissolve the powder: methanol, ethyl acetate, and n-hexane. Two stages were used to test the extracts' antibacterial activity: the disc-diffusion method for antibacterial activity and the dilution method for antibacterial activity (MIC). Using dried powdered plant materials, a stock solution of extract was prepared for the disc diffusion procedure, and it was diluted with varying quantities. After that, the extracts were injected onto sterile, blank discs with a diameter of 6 mm, and they were incubated for 24 h at 37°C. Using the dilution procedure, the root extracts' secondary antibacterial activity and MIC were ascertained. The MIC of the examined root extracts that prevented the bacterium under test from growing visibly was noted. E. coli and P. aeruginosa, two Gram-negative bacteria, were successfully combatted by methanolic root extracts of V. tessellata. According to earlier research, V. tessellata showed antibacterial activity against each of the four bacterial strains. Prior research is supported by V. tessellata's superior effectiveness against the two Gram-negative bacteria.

In summary, root extracts from *V. tessellata* have demonstrated potential for both antibacterial activity and the treatment of a variety of bacterial illnesses.^[73]

APHRODISIAC ACTIVITY

Subramoniam *et al.* investigated the effects of an alcohol extract from *V. tessellata* flower on aphrodisiac activity (10) in mice. The extract increased blood nitric oxide levels in mice in a way that depends on concentration and time. The aphrodisiac compound (AC) was isolated from the AF and activated corpus cavernosum tissue nitric oxide synthase (NOS) activity. In short-term restricted toxicity assessments conducted on male mice, the AC did not exhibit any noticeable toxicity. According to the study, *V. tessellata* has a significant activator (AC) of endothelial NOS and neuronal NOS that can activate these two proteins and provide an aphrodisiac effect through cGMP-mediated pathways. This finding may result in the creation of an effective treatment for erectile dysfunction or sexual dysfunction.^[74]

The plant material studied for aphrodisiac properties in various experimental paradigms (*in vitro*, *in vivo* on animal models, or in human clinical trials) is summarized by Patel *et al*. Medicinal plants can be employed for a variety of therapeutic uses or as building blocks to create effective medications with a range of phytochemicals. Many people utilize plants to

treat sexual dysfunction. It was discovered that an alcoholic extract of *V. tessellata* flowers when taken orally at levels of 50 and 200 mg/kg, enhanced the effectiveness of mating and, in general, raised the progeny's male-to-female ratio. There was no general toxicity in this extract.^[75]

Singh *et al.* when used male mice to screen orchids for aphrodisiac properties, *V. tessellata* was found to have exceptionally strong aphrodisiac properties. It was discovered that male mice's mounting behavior was stimulated by the flower and, to a lesser extent, the root (but not the leaf). The flower's alcoholic extract contained this activity. In mice, the flower extract (50 or 200 mg/kg) improved mating efficiency. The male/female ratio of the pups fathered by the extract-treated mice showed a rising tendency, and the pups were confirmed to be normal. There has been no obvious harm in mice according to preliminary toxicology testing. There were no reputable clinical trials available for this herb that demonstrated its ability to treat male sexual dysfunction. [76]

In male mice, Kumaret al. evaluated that the extract of *V. tessellata* (Roxb.) exhibited aphrodisiac properties. Male mice were given an aqueous suspension (2 g/kg) or extract (water or alcohol, 200 mg/kg) of the roots, flowers, or leaves of *V. tessellata*, and their mounting behavior was assessed 1 h later. While leaf extracts had little effect, floral and, to a lesser amount, root alcohol extracts promoted mounting behavior. Mice were given the most active extract (an alcohol extract of flowers) at dosages of 50 and 200 mg/kg, po, and the results of mating and reproduction were recorded. The male-to-female ratio of the progeny tended to rise as a result of this extract's greater ability to facilitate mating. There was no overall toxicity in this extract.^[77]

ANTI-PYRETIC ACTIVITY

Shekhawat and Sisodia evaluated for the antipyretic activity of unrefined water extract of *V. roxburghii* plant. To determine the pharmacological activity, dried, ground, and extracted leaves of V. roxburghii were used. For future research, the extract was kept in an amber-colored container. Four experimental groups—control, standard, and V. roxburghii were randomly assigned to the rats. Next, the animals were given a 15% suspension of Brewer's yeast in 0.9% saline. The rats were injected, and the rectal temperatures were measured after 18 h. Oral administration of both V. roxburghii and the conventional medication was used. According to the study, when compared to the common medication paracetamol, both extracts exhibited strong antipyretic effectiveness. To achieve therapeutic results, these extracts must be used in repeated dosage treatments. Significant antipyretic action against paracetamol was demonstrated by Extract from V. roxburghii (100 mg/kg body weight), indicating that repeated dosage administration is required for therapeutic results. The study emphasizes how V. roxburghii extract may be used to control the antipyretic and analgesic effects on animals.^[70]

CONCLUSION

Since the very beginning of humankind, medicinal plants have been one of the most reliable and trustworthy sources of safe therapy. In addition to being widely utilized in traditional medicine and perhaps having a wide variety of medicinal characteristics, The Vanda genus is renowned for its beautiful blossoms. More studies should also be done on the sustainable usage, farming, and conservation of these species. It has been discovered that V. roxburghii contains a wide range of chemical compounds. This herb possesses antioxidant, antifungal, antiulcer, anticonvulsant, and aphrodisiac properties. To find novel lead compounds, these functions in the orchid should be further examined pharmacologically and molecularly. Enhancing human health requires more study into the ethnomedicinal qualities of exotic and native orchids. This plant has properties that include anti-inflammatory, antibacterial, antimicrobial, analgesic, antimitotic, aphrodisiac, antifungal, antiulcer, anticonvulsant, and antioxidant properties.

To find novel lead compounds, these functions in the orchid should be further examined pharmacologically and molecularly. To improve human well-being, further research is needed into the ethnomedicinal properties of both native and exotic orchids.

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