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Evaluation of Bioactive Phytochemicals of *Lepidagathis keralensis*

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ABSTRACT

Lepidagathis keralensis, a plant native to the Kerala region of India, belongs to the family Acanthaceae, and has gained attention for its potential medicinal properties. There are many species in the genus, some of which are primarily found in tropical and subtropical regions. *Lepidagathis keralensis* species found in India. With a woody root stalk, it is a stiff prostrate shrub. It is usually found in exposed lateritic rocks in lateritic hills. It's commonly referred to as Paramullu. Prior research has demonstrated that plants are a rich source of numerous bioactive components and have therapeutic qualities. This review involves the evaluation of phytochemical screening of *Lepidagathis keralensis* methanolic extract and also analyzing the importance of metabolite present in *Lepidagathis keralensis*. This review aims to highlight *Lepidagathis keralensis* bioactive phytochemicals. A comprehensive phytochemical screening of *L. keralensis* reveals the presence of various bioactive compounds. Alkaloids, known for their pharmacological effects, have been identified in plant extracts. Glycosides, which possess medicinal properties and are often used in traditional medicine, are also present. Additionally, amino acids, essential for various physiological functions, sterols with potential therapeutic benefits, and carbohydrates contributing to the plant's nutritional value have been detected. This article also aiming to evaluate various phytochemicals obtained through GC MS analysis of *Lepidagathis keralensis.* The Important phytochemicals present in the plant extract are n-Hexadecanoic acid, 2-Decanynoic acid, 1,6-Octadiene, 3,7-dimethyl, & Cyclopentaneundecanoic acid. This review also containing an evaluation of Anti-microbial and Anti-oxidant properties of *Lepidagathis keralensis*. Antimicrobial studies were done by agar diffusion method. For antibacterial evaluation gram negative (Pseudomonas aeroginosa & Klebsiella pneumonia) and gram-positive (Streptococcus mutans & Staphylococcus aureus) bacterial strains were used. The antifungal efficacy was tested against Candida albicans. The analysis revealed the presence of several bioactive phytocomponents which could be responsible for the antimicrobial activity and Anti-oxidant activity of the *Lepidagathis keralensis.*

INTRODUCTION

Nearly all societies have long employed medicinal herbs. For thousands of years, people have employed medicinal herbs to treat illnesses and stave off sickness. A flowering plant native to Kerala, India's western ghats is called *Lepidagathis keralensis*. There are 100 species in the *Lepidagathis* genus, which are extensively distributed throughout tropical and subtropical regions of Asia and Africa. It is a perennial herbaceous plant that is a member of the Acanthaceae family. Because of their many medical properties, plant species in the Acanthaceae family are prized for their cultural and economic importance in horticulture and traditional medicine across the globe. The large genus *Lepidagathis* contains more than 110 species, 33 of which are found in India. There is minimal scientific information available on bioactivity. Plants from the *Lepidagathis* class have traditionally been used to treat polyuria, fever, mouth ulcers, diarrhea, uterine problems, and polyuria. The family plants are known to contain cytotoxic, anti-fungal, Anti-inflammatory, anti-oxidant, anti-viral, anti-pyretic, hepatoprotective, immunomodulatory and antiplatelet aggregation properties. The major isolated chemical constituents of the genus Lepidagathis include essential oils, sesquiterpenes, flavonoids, phenolics, alkaloids, inorganic minerals and saponins. [1,2]

Lepidagathis keralensis unique to Kerala. There are many species in the genus, some of which are primarily found in tropical and subtropical regions. *Lepidagathis keralensis* species found in India. With a woody root stalk, it is a stiff prostrate shrub. It is usually found in exposed lateritic rocks in lateritic hills close to the coast. It's commonly referred to as Paramullu. Prior research has demonstrated that plants are a rich source of numerous bioactive components and have therapeutic qualities. This review aims to highlight *Lepidagathis keralensis* bioactive phytochemicals. *Lepidagathis keralensis* (Acanthaceae) is a less explored plant for research studies. The plant is known to contain likes cytotoxic, anti-fungal, anti-inflammatory, anti-oxidant and insecticidal properties. The plant had many medicinal properties. The spines of the plant used by Paniya tribes for digestive disorders. The plant is also used for kidney stone, asthma, chest pain, blood purifier etc. The plant has many medicinal properties like anti-inflammatory, anti-cancer, antibacterial, antifungal, and diuretic properties. [3,4]

TAXONOMICAL CLASSIFICATION[5]

Kingdom: Plantae
Phylum: Tracheophyte
Class: Magnolisidae
Order: Laminale
Family: Acanthaceae
Genus: *Lepidagathis*
Species*: Lepidagathis Keralensis*

COMMON NAME

Nonganampullu
Paramullu
Venappacha

Figure1: *Lepidagathis keralensis*

BOTANICAL DESCRIPTION [6,7]

Lepidagathis keralensis is a perennial**,** prostrate, woody herb, which is attached to hard lateritic soil. Rootstock is woody, stem always glabrous, quadrangular, more or less winged, much branched, and form a carpet of up to 1sq m, rooting at the nodes. Leaves are narrowly oblong, lanceolate, acute acuminate at apex, 10 mm, leaves are dark green with purple margins, rigid, plicate, glabrous and nerved. Spikes 1-3, terminal, procumbent, 2cm long, flower sessile, 1cm long, many sterile bracts (5-8), more or less uniform. Calyx villous,

deeply 5-lobed, erect or reflexed, pink, lower tip 3-lobed, pink with white to yellow palate, stamens 4, didynamous, up to 6mm long, hairy, 2celled, purple to deep violet color. Ovary compressed ovoid, 2mm long, 2 celled with one ovule in each, style slender, 7-8mm long, hairy at lower ventral region with glands. Fruits are conoid, compressed capsule, 6 mm long, glabrous. Seeds are 2, flat, soft, hairy with white aril. Flowering and fruiting, December to April. By May all plant parts, except the rootstock, become dry and dead. And onset of the monsoon June new shoots is arisen from rootstock. Spiny bracts and calyx help in seed dispersal. [6]

L*epidagathis keratosis* resembles *Lepidagathis prostrata* in its prostrate habit, glandular-pubescent, terminal inflorescence but differs by having much smaller, acute to blunt acuminate leaves, short (2 cm) inflorescence, and non-striated corolla lobes. One of the significant characteristics in diagnosis is 'shorter spikes' which grow up to 2 cm. Its occurrence in Goa is an error as the spike in those plants reaches up to 9 cm. The geographic range of *Lepidagathis keralensis* is restricted to Karnataka and Kerala.

HABIT	HERB
HABITAT	LATERITE HILLS
ROOTSTOCK	WOODY
STEM	GLABROUS, QUADRANGULAR,
LEAVES	SIMPLE, OPPOSITE

COLLECTION, DRYING AND EXTRACTION[8]

The plant specimen was collected from Dist. Kannur, Kerala, India. The plant specimen *Lepidagathis keralensis* was identified and authenticated by the Department of botany. The plant material was washed under running tap water, air dried in shade for two weeks and then it was homogenized to fine powder and stored in sterile air-tight bottles for the experimental use. The Powdered material was extracted with methanol at room temperature for six days using Soxhlet extraction method. The liquid extract was taken from round bottom flask and allow to evaporate the solvent. The extract was concentrated to get crude extract. Crude extract was stored in a air tight container for further phytochemical screening.

Figure 2; dried L. keralensis Figure 3; powdered L. keralensis Figure 4: Crude extract

PHYTOCHEMICAL SCREENING [9,10,11]

The phytochemical screening of *Lepidagathis keralensis* extracts was performed to establish the type of constituents present in Methanol extract and its fractions as per standard methods.

Chemical test for alkaloids

A small portion of dried alcoholic extract was shaken with dilute hydrochloric acid and filtered the filtrate was tested with the following reagent, to detect the presence of alkaloids.

a) Mayer's test

The acidified extract (two ml) was treated with 1 ml of Mayer's reagent (potassium mercuric iodide), shaken and noted for the presence of a creamy precipitate. The extract of lepidagathis keratosis shows white precipitate, that indicates the presence of an alkaloid.

b) Wagner's test

The acidified extract (two ml) was treated with a few ml of Wagner's reagent (solution of Iodine in potassium iodide) and observed for the presence of reddish-brown precipitate. The L. Keralensis crude extract shows brown precipitate, means the presence of alkaloids.

c) Hager's Test

The acidified extract (two ml) was treated with 1 ml of Hager's reagent (saturated picric acid solution) and observed for the presence of yellow precipitate. when reacting with the methanolic extract of *L. keralensis* showed a yellow precipitate.

d) Dragendorffs test

The acidified extract (two ml) was treated with a few ml of Dragendorffs reagent (Potassium bismuth iodide) and observed for the presence of orange red precipitate. The methanolic extract of *L. keralesis* reacts with Dragendorffs reagent doesn't shows an orange red precipitate.

Chemical tests for glycosides

A small portion of the extract was hydrolyzed with dilute hydrochloric acid for few hours on a water bath and the hydrolysate was later subjected to the following tests to detect the presence of glycosides.

a) Legal's Test

The residue (dry extract) left after evaporation was dissolved in a few milliliters of pyridine. Two milliliters of freshly prepared sodium nitroprusside solution were added to it and then made alkaline with sodium hydroxide solution. It was observed for the formation of pink-red color. The observation is red color formation with *L, keralensis* extract.

b) Baljet's test

The few ml of the extract was treated with 1ml sodium picrate solution and a yellow to orange color reveals the presence of cardiac glycosides. *L.keralesis* extract shows the presence of orange color confirming the presence of glycosides.

Chemical tests for phenolic compounds and tannins

a) Ferric chloride test

A small quantity of the extract diluted with water was treated with dilute ferric chloride solution (5%) and observed for the presence of blue color. *L.keralesis* extract shows blue color indicate the presence of phenolic compounds and tannins.

b) Gelatin test

the extract dissolved in water was filtered. To the filtrate, 2% solution of gelatin containing 10% sodium chloride was added. Noted for the presence of milky white precipitate. *L.keralesis* showed the presence of phenolic compounds and tannins.

c) Lead acetate test

the extract dissolved in water was treated with 10% lead acetate solution. Noted for the presence of bulky white precipitate. When lepidagathis keralensis treating with lead acetate solution shows the white precipitate indicates presence of phenolic compounds.

Chemical tests for flavanones and flavonoids

a) Aqueous sodium hydroxide test

Aqueous sodium hydroxide solution was added to the few ml of the extract and the presence of yellow coloration of the solution was noted. The extract of *L. keralesis* shows yellow coloration due to the presence of flavonoids.

Chemical tests for carbohydrates

A small quantity of ethanolic extract was mixed with water or alcohol and filtered. The filtrate was subjected to the following tests to detect the presence of carbohydrates.

a) Molisch's Test

The filtrate (two ml) was treated with a few drops of Molisch's reagent and two ml of concentrated sulphuric acid was added through the sides of the test tube without shaking. Observed for the presence of violet ring at the junction of two solutions. A violet ring appears to indicate the presence of sugar in *Lepidagathis keralensis*.

b) Fehling's Test

The filtrate (one ml) treated with 1 ml each of Fehling's solution A and B and boiled on a water bath for half an hour, then observed for the presence of red residue at the bottom of test tube. Red residue formed with the extract, indicate the carbohydrate in *L. keralesis*.

c) Benedict's Test

The filtrate (a few drops) was treated with two ml of Benedict's reagent. Then the mixture was heated on a boiling water bath for two min and the presence of red precipitate was noted. Red precipitate identifies the presence of *lepidagathis keralensis*.

Chemical tests for proteins and amino acids

a) Million's Test

The extract (two ml) was treated with few drops of Million's reagent (1gm of mercury+ 9ml of fuming nitric acid) and observed for the presence of white precipitate, which on warming turned into a red colored solution. Millions reagent react with *lepidagathis keralensis* extract forms a white precipitate indicate the presence of amino acids.

b) Biuret Test

The extract (two ml) was treated with one drop of 2% copper sulphate solution. To this 1ml of 95% ethanol was added followed by excess of potassium hydroxide solution and Observed for the presence of violet colored solution. violet color was formed by the crude extract indicate the presence of amino acids.

c) Ninhydrin Test

The extract (few ml) was treated with two drops of ninhydrin solution and heated on a water bath and then the presence of violet color was noted. violet color was formed by the crude extract of *L. keratosis* indicate presence of amino acids.

Chemical test for terpenoids

a) Salkowski's Test

The extract (few ml) was dissolved in chloroform. An equal volume of concentrated sulphuric acid was added to it and noted for the appearance of red color in the chloroform layer and greenish-yellow fluorescence in the acid layer. The absence of color change indicates the absence of terpenoids.

Chemical tests for sterols

A little amount of the alcoholic extract was refluxed with solution of alcoholic potassium hydroxide until saponification was observed. The mixture was diluted and extracted with solvent ether. The ethereal extract was evaporated, and the residue was subjected to Liebermann-Burchard's and Salkowski's tests.

a) Liebermann – Burchard's test

concentrated sulphuric acid through the sides of the test tube and observed for the development of a deep red color in the lower portion and green color in the upper portion which changes to blue and violet. Color changes with the extract may indicate the presence of sterols.

b) Salkowski's Test

The residue was dissolved in chloroform and equal volume of concentrated sulphuric acid was added to it and observed for the red color in the lower layer. Red color formation as a result of Salkowski test indicate the presence of sterol in *Lepidagathis keralensis*.

Chemical tests for saponins

a) Foam or Froth Test

A small quantity of extract was mixed with 20 ml of distilled water in a graduated cylinder. The suspension was shaken for fifteen minutes and allowed to find any froth was formed. The suspension of *L. keratosis* shows froth indicate that presence of saponin.

IMPORTANCE OF BIOACTIVE PHYTOCHEMICALS

ALKALOIDS

An alkaloid is a class of naturally occurring organic compounds that mostly contain basic nitrogen atoms. They are produced by a variety of organisms, including plants, animals, bacteria, and fungi. Alkaloids often have potent pharmacological effects on humans and other animals, making them significant in medicine, agriculture, and even recreational drug use. The importance of alkaloids lies in their diverse range of biological activities. Many alkaloids have medicinal properties and are used as drugs to treat various diseases Some alkaloids also play roles in plant growth and development. *Lepidagathis keratosis* is a plant species found in the Western Ghats of India. It belongs to the family Acanthaceae. These alkaloids may contribute to the *L. keralesis* medicinal properties or serve as a defense mechanism against herbivores. Alkaloids are commonly found to have antimicrobial properties against both Gram-positive and Gram-negative bacteria. The results for the phytochemical screening of *lepidagathis keralensis* revealed the alkaloids presence in the methanolic extract

of *lepidagathis keralensis.* Further research would be needed to identify and characterize the alkaloids present in *Lepidagathis keralensis* and understand their significance. [12,13,14]

GLYCOSIDES

A broad class of substances called glycosides is present in microorganisms, animals, and plants. They are made up of a sugar molecule called glycone that is joined to a non-sugar molecule called aglycone by a glycosidic bond. Numerous non-sugar molecules can exist, giving rise to a large variety of glycosides with unique characteristics and roles. Numerous phytochemicals, including glycosides, are known to be present in it. *Lepidagathis keralensis* contains a variety of glycosides, some of which may have pharmacological properties or have other biological purposes for the plant. To completely comprehend the significance of the glycosides in *Lepidagathis keratosis* and to identify and define them, more research is required. [13,14,15]

CARBOHYDRATES

Carbohydrate content in L. keralesis species can vary widely depending on factors such as species, plant part, maturity, and growing conditions. Carbohydrates are essential for plants as they serve as a source of energy and as structural components. carbohydrates in *lepidagathis keralensis* are primarily composed of sugars, starches, and fiber. Sugars such as glucose, fructose, and sucrose are soluble carbohydrates that provide readily available energy to the plant. Starches, which consist of long chains of glucose molecules, serve as a storage form of energy in plant tissues such as seeds, tubers, and roots. Fiber, including cellulose, hemicellulose, and pectin, provides structural support to the plant cell walls and aids in digestion for herbivores. [14,15,16]

AMINO ACIDS

Similar to animal amino acids, plant amino acids are essential for many biological activities such as cell signaling, enzyme activity, and protein synthesis. Through photosynthesis and other metabolic processes, plants are able to manufacture all the amino acids they require. *Lepidagathis keralensis* is a plant species that contains different varieties of amino acids. L.keralesis can have different amino acid compositions based on species, types of tissues, developmental stages, and environmental influences**.** [16,17,18]

STEROLS

Sterols are a type of lipid and are essential components of cell membranes, where they help maintain membrane fluidity and integrity. The most well-known sterol in animals is cholesterol, while in plants, the primary sterol is called phytosterol. Phytosterols present in *Lepidagathis keralensis*, its concentration and significance vary according to its biology. Phytosterols have potential health benefits, including their role in lowering LDL cholesterol levels and reducing the risk of heart disease. [19, 20 ,21]

PHENOLIC COMPOUNDS

Phenolic compounds are a diverse group of secondary metabolites found in plants, characterized by the presence of one or more phenol groups. Lepidagathis keralensis contains phenolic compounds that help in plant growth, development, and defense. Some common types of phenolic compounds found in plants include flavonoids, phenolic acids, lignans, and tannins.[22] The importance of phenolic compounds in Lepidagathis keralensis helps plants to combat oxidative stress caused by factors such as UV radiation, pollution, and pathogens. They scavenge free radicals and reactive oxygen species, thereby protecting plant cells from damage. Many phenolic compounds have been identified for their potential medicinal properties in humans, including antioxidant, anti-inflammatory, and anticancer effects. [,23,24,]

Table 1; preliminary screening of Lepidagathis keralensis

SL NO.	PHYTOCHEMICALS	METHANOLIC EXTRACT
1.	ALKALOIDS	PRESENT
2.	CARBOHYDRATES	PRESENT
3.	GLYCOSIDES	PRESENT
4.	AMINOACIDS	PRESENT
5.	SAPONINS	PRESENT
6.	FLAVANOIDS	PRESENT
7.	PHENOLIC COMPOUNDS	PRESENT
8.	SAPONINS	PRESENT
9.	TERPENOIDS	ABSENT

EVALUATION OF GC MS ANALYSIS [25]

The plant extract of *Lepidagathis keralensis* was undergone GC-MS analysis using a Thermo Scientific Trace 1300 Gas chromatograph equipped with an ISQ-QD Mass Spectrometer. Gas chromatography-mass spectrometry (GC-MS) analysis plays a crucial role in the study of Lepidagathis keralensis extracts. GC-MS allows for the separation and identification of individual compounds present in complex *Lepidagathis keralensis* extracts. This is essential for understanding the chemical composition of *L. keralesis.* GC-MS helps in identifying bioactive compounds with potential pharmacological or therapeutic properties in *Lepidagathis keralensis*.[26,27]

GC MS analysis results of *Lepidagathis keralensis* extract shows 38 phytocomponents in *Lepidagathis keralensis* leaves extract and 20 phytocomponents in stem extract. Total of 58 phytochemicals are present in *Lepidagathis keralensis* extract. N-Hexadecanoic acid (14.53%), 2-Decanynoic acid (16.71%), 1,6-Octadiene, 3,7-dimethyl- (20.34%) & 1,5-Heptadiene, 3,3-dimethyl-, (E)-(12.77%), Cyclopentaneundecanoic acid (25.06), Oxalic acid, allyl hexyl ester (10.41) ,1- Iodo-2-methyl nonane (9.91) & n-Hexadecanoic acid (9.96%) were the major components. From the above major components n-Hexadecanoic acid (14.53%), 2-Decanynoic acid (16.71%), 1,6-Octadiene, 3,7-dimethyl- (20.34%) & 1,5-Heptadiene, 3,3-dimethyl-, (E)-(12.77%) present in leaves extract and Oxalic acid, allyl hexyl ester (10.41) ,1- Iodo-2-methylnonane (9.91) & n-Hexadecanoic acid (9.96%) are the components present in stem extract.[28,29]

For analyzing the GC MS study, a thermoscientific Trace 1300 Gas Chromatograph with an ISQ-QD Mass Spectrometer and a TG-5MS non-polar column was used to conduct the analysis. Ionization of electrons for GC-MS detection, a device with an ionizing energy of 70 eV was employed. A split ratio of 1:8 was utilized, with helium gas (99.99%) serving as the carrier gas at a steady flow rate of 1 ml/minute and an injection volume of 1 µl. The ion source temperature was set at 200ºC, while the injection port temperature was set to 280ºC. For analyzing different extract, the oven temperature was adjusted differently, for leaf extract 70 ºC (isothermal for 3 minutes) with an increase of 5 ºC / minute to 180 ºC with a hold time of 3 minutes. Then temperature was increased at a rate of 5 ºC /min till 240 ºC with a hold time of 5 minutes. Total GC running time was 45 minutes. For stem extract the oven temperature was programmed from 80 ºC (isothermal for 3 minutes) with an increase of 15 ºC / minutes to 180 ºC with a hold time of 2 minutes. Then temperature was increased at a

rate of 5 ºC /minutes till 240 ºC with a hold time of 5 minutes. Total GC running time was 25 minutes.[30]

Lepidagathis keralensis extracts are a rich source of natural compounds that can serve as leads for drug discovery. GC-MS analysis aids in the identification of novel compounds with pharmacological activities in *L. keralensis* extract that provide valuable insights for the development of new drugs or therapeutic agents.[31]

N-HEXADECANOIC ACID

n-Hexadecanoic acid is an important phytochemical present in lepidagathis leaf extract. The peak area percentage was found to be 14.53%. n-Hexadecanoic acid, also known as palmitic acid, is a saturated fatty acid commonly found in plants. Palmitic acid is a major component of plant cell membranes, where it contributes to the structural integrity and fluidity of the membrane. During periods of high energy demand, such as seed germination or seedling establishment, stored palmitic acid can be mobilized and metabolized to provide energy. Palmitic acid serves as a precursor for the biosynthesis of longer-chain fatty acids, complex lipids (e.g., phospholipids, glycolipids, and sphingolipids), and various lipid-derived compounds such as waxes, sterols, and signaling molecules. Palmitic acid can also act as a signaling molecule involved in the regulation of gene expression.[32]

When n-hexadecanoic acid is identified through gas chromatography-mass spectrometry (GC-MS) analysis of plant extracts, it provides valuable insights into the chemical composition and metabolism of the plant material. When n-hexadecanoic acid is identified, it contributes to the chemical profile of the plant extract, providing information about the presence and abundance of this fatty acid. Identification of n-hexadecanoic acid reveals information about the fatty acid composition of the plant. Fatty acids are important constituents of lipids, which serve various roles in plants, including energy storage, membrane structure, and signaling. the identification of n-hexadecanoic acid through GC-MS analysis of plant extracts offers valuable information about the modulation of plant responses to abiotic stresses such as cold, drought, and salinity.[33]

2-DECANYNOIC ACID

2-Decanynoic acid is an important phytochemical present in lepidagathis leaf extract. The peak area percentage was found to be 16.71%.2-Decanynoic acid, also known as 2-decyne-1-oic acid, is a fatty acid with a chain length of ten carbons and a terminal triple bond. 2-

decanoic acid, can act as allelochemicals, which are compounds produced by plants that influence the growth, development, or behavior of other organisms, including other plants. These compounds can have allelopathic effects, inhibiting the growth of neighboring plants, pests, or pathogens. Fatty acids are integral components of cell membranes in plants.

2-Decanynoic acid has been identified in a *Lepidagathis keralensis* extract through GC-MS analysis, exploring its potential functions, such as allelopathic activity, defense mechanisms, or metabolic regulation, as mentioned earlier. They also contribute to membrane structure, fluidity, and function of *L. keralensis.* Fatty acids are also involved in metabolic regulation in *L. keralesis.* They serve as energy storage molecules and precursors for the biosynthesis of other lipid compounds, such as phospholipids, glycolipids, and plant hormones. 2-Decanynoic acid in a plant extract through GC-MS analysis opens up avenues for further research and exploration into its biological significance, ecological roles, and potential applications.[34]

1,6-OCTADIENE, 3,7-DIMETHYL

1,6-Octadiene, 3,7-dimethyl-, also known as myrcene, is a naturally occurring organic compound found in various plants. It is a monoterpene, which belongs to the class of terpenes, a large and diverse group of natural compounds synthesized by plants. Myrcene is believed to have various medicinal properties, including analgesic (pain-relieving), anti-inflammatory, and sedative effects. It is often present in essential oils derived from aromatic plants and has been used in traditional medicine for its therapeutic benefits. Myrcene exhibits various biological activities, including antioxidant, antimicrobial, and insecticidal properties.[35]

The identification of 1,6-Octadiene, 3,7-dimethyl in *Lepidagathis keralensis* through GC-MS study with a percentage of 20.34 gives valuable insights into the chemical diversity, ecological interactions, and potential applications of natural compounds in plants. In *Lepidagathis keralensis*, it may serve as a defense mechanism against microbial pathogens and herbivorous insects.

CYCLOPENTANEUNDECANOIC ACID

The identification of Cyclopentaneundecanoic acid in the GC-MS analysis of Lepidagathis keralensis, suggests its presence as a natural compound within this plant. Cyclopentaneundecanoic acid is a compound belonging to the class of fatty acids. It consists

of a cyclopentane ring fused with an undecanoic acid moiety, which is a fatty acid with an 11-carbon chain. The peak percentage of Cyclopentaneundecanoic acid was found to be 25.06%. This compound might possess biological activities with potential pharmacological or therapeutic applications. It could be investigated for its antimicrobial, antioxidant, anti-inflammatory, or other bioactive properties. [36]

Lepidagathis keratosis GC-MS study revealed the presence of Cyclopentaneundecanoic acid, which offers up new research opportunities on its ecological roles, biological importance, and its uses in a variety of sectors.[37]

PHARMACOLOGICAL EVALUATION [38]

ANTI MICROBIAL ACTIVITY

Anti-microbial assays are crucial in evaluating the potential medicinal properties of *Lepidagathis keralensis* extracts. Anti-microbial assays help in screening plant extracts for compounds that inhibit the growth of bacteria, fungi, viruses, or other microorganisms. This can lead to the discovery of new antibiotics or antimicrobial agents. The anti-microbial assay for *Lepidagathis keralensis* was evaluated by doing an Anti-bacterial assay and Anti-fungal assay.[39]

ANTI BACTERIAL ASSAY

The Agar well diffusion method was used to test the components of *Lepidagathis keralensis* for antibacterial activity. Two-gram negative strains of Klebsiella pneumonia and Pseudomonas aeruginosa and two-gram positive strains of Streptococcus aureus and Streptococcus mutants are employed. While the Petri plate was still hot, 20 milliliters of the prepared Muller-Hinton Agar medium was added. As needed, nutrient agar was distributed uniformly. After that, it is autoclaved for 15 minutes at 121˚c. A bacterial culture was then added to the plates as an inoculant. Plates with wells were made, and various concentrations of material (25 µg, 50 µg, and 100 µg) were added. The zone of inhibition (ZOI) surrounding the well was evaluated after a 24-hour incubation period at 37˚C for petri plates. The methanol extract of the leaf and the acetone extract of the stem both suppress the growth of the gram-negative bacteria Klebsiella pneumonia. The study showed that, the importance of *Lepidagathis keralensis* plant species as an anti-bacterial agent.[40,41]

ANTI FUNGAL ASSAY

The agar well diffusion method is used to measure the antifungal activity of *Lepidagathis keralensis*. *Candida albicans* is used for fungal strains. The fungal strain was cultivated and inoculated on potato dextran agar plates. are ready on the plates, then Samples were introduced at various amounts (25 µg, 50 µg, and 100 µg). After incubating the plates, the zone of inhibition was calculated. Leaf and stem methanol extracts of *Lepidagathis keralensis* work well against fungi. The measured values of zone inhibition were 15 and 16 mm, respectively. This study indicated the importance *L. keralesis* as an anti-fungal agent.[42,43]

ANTIOXIDANT ASSAY

The anti-oxidant activity of *Lepidagathis keralensis* was done by different methods. The study made use of the DPPH radical scavenging assay, the reducing power assay, and the total antioxidant activity methods. Antioxidants lower DPPH in the DPPH scavenging experiment by giving up a proton, which lowers the solution's absorbance in methanol at 517 nm. The staining of DPPH in methanol was used to gauge the extracts' capacity for scavenging. The leaf methanolic extracts had the highest inhibitory activity of 94.78% among the various extracts, which was similar to normal ascorbic acid's 96.38%. The IC50 value of the methanolic extract is 122.46±0.85. [44]

The extract of *L. keralensis* was examined in the reducing power experiment, and the results indicated that absorbance increased as concentration increased, indicating that the plant had strong antioxidant qualities. At 700 nm, the absorbance was measured. The leaf's methanol extract displayed the maximum absorbance (1.226), which was nearly identical to the absorbance of ascorbic acid standard (1.462). The acetone extract from the stem had the best potential for antioxidants among the extracts. The phosphomolybdenum technique was used to measure the total antioxidant activities of the extracts. The results showed that the methanolic extract of the leaf had the highest total antioxidant activity (656.89±1.68), followed by the acetone extract of the stem (532.0±2.0).[45]

Phytochemicals in *Lepidagathis keralensis*, such as polyphenols and flavonoids, have been linked to its Anti-oxidant activity, they are the reason to numerous health benefits, including reducing the risk of reducing the risk of chronic diseases like cancer, cardiovascular diseases, and neurodegenerative disorders. Evaluating antioxidant activity helps determine the potential health-promoting effects of *L. keralensis* extracts.

CONCLUSION

Lepidagathis keralensis is a valuable medicinal plant rich in bioactive chemicals with diverse pharmacological activities. The evaluation of phytochemical screening, evaluation of GC-MS analysis, and biological activities underscore the therapeutic potential of *L. keralensis*, in managing various health conditions. Several studies have documented the phytochemical profile of *L. keralensis*, revealing the presence of diverse bioactive compounds. Alkaloids, glycosides, amino acids, sterols, and carbohydrates are among the major constituents identified in various parts of the plant. Gas chromatography-mass spectrometry (GC-MS) analysis has been employed to elucidate the chemical composition of *L. keralensis* extracts. This analytical technique has identified numerous compounds present in this plant, The GC-MS analysis provides valuable insights into the chemical complexity of *L. keralensis* and aids in understanding its medicinal properties. *L. keralensis* extracts have demonstrated significant antimicrobial and antioxidant activities in various studies. These biological properties are attributed to the presence of bioactive compounds such as alkaloids, glycosides, and phenolic compounds.

The collected phytochemical and pharmacological findings indicate the strong potential of *Lepidagathis keralensis*. The review reveals that *L. keralesis* is a potential source of antimicrobials and antioxidants. Overall, this plant can be considered as a candidate for the introduction of a new drug compound with numerous applications in medical field. Further studies are still required to evaluate more secondary metabolites of this plant, also to determine biological activities and the mechanism behind its activities.

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REFERENCES

1. Arun Nivrutti Chandore, Devidas Bhausaheb Borude, Nilesh Appaso Madhav & Shrirang Ramachandra Yadav., *Lepidagathis sabui* (Acanthaceae), a new species from the lateritic plateaus of Konkan region of Maharashtra, India. Phyto taxa. 2020; 464 (2);159-166
2. Ali Sobhanizadeh, HoshangYadegari, BahmanFazeli-nsaab, and BarataliFakheri. Introduction on application of herbal medicine. The 1st annual Iranian agriculture research conference.2015;20(1) 1- 11.
3. RefazAhmd Dar, MohmdShahnawaz, Parvaiz Hassan Qazi. General overview of medicinal plants. A review the journal of phytopharmacology.2017;6(6):349-351.

4. PalakkalLeena, N.H. ZeinulHukuman, A.R. Biju, MullapallyJisha. Studies on methanolic extract of Lepidagathiskeralensis as a green corrosion inhibitor for mild steel in 1M HCl. Journal of electrochemical science and technology. 2019 ;10(2): 231-243.
5. Poornima M, Jacob A, AjithBabu T. K, Malavika T. M, *Lepidagathis Keralensis*: An overview. Int J Res and Sci Innov. S 2021;8 (7): 25-7.
6. Madhusoodanan, P, V, Singh, N, P, A new species of Lepidagathiskeralensis (Acanthaceae) from south India. New bulletin.1992;27(2):301-303
7. BeebiRazeena, P,M,MiniMBioefficiency & phytochemical analysis of Lepidagathiskeralensis of Acanthaceae. World journal of pharmaceutical and sciences.2017; 3(2):124-126.
8. Leena. P, ZeinulHukman N H, Jisha. M. Evaluation of antimicrobial activity of crude extract of Lepidagathiskeralensis. IJRP. 2017;8(3):321-326
9. Amita Pandey, Shalini Tripathi, Concept of standardization, extraction and pre phytochemical screening strategies for herbal drug. Journal of Pharmacognosy and Phytochemistry 2014; 2 (5): 115-119.
10. Shalini S and *Prema Sampathkumar Phytochemical screening and antimicrobial activity of plant extracts for disease management INT J CURR SCI 2012, 209-218 RESEARCH ARTICLE ISSN 215.
11. A. Doss, Preliminary phytochemical screening of some Indian Medicinal Plants Ancient Science of Life, Vol. 29, No.2 (2009) Pages 12 -16.
12. LeenaPalakkal, ZeinulHukuman. N.H, Jisha. M. Antioxidant activities and chemical composition of various extracts of Lepidagathiskeralensis. Journal of Applied Pharmaceutical Science. 2017 ;7(06): 182-189
13. Chashoo IA, Kumar D, Bhat ZA, Khan NA, and Kumar V. Antimicrobial studies of Sambucus wightiana Wall. Wight & Arn JPR.2012; 5: 2467.
14. Bader GN, Antimicrobial studies of rhizome of Swertia petiolata. J App Pharm Sci 4: 2013;128-130.
15. Dahanukar SA, Kulkarni RA, Rege NN. Pharmacology of medicinal plants and natural products. Indian J Pharmacol 32:2000; S18-S118.
16. Lacey J , The microbiology of cereal grains from areas of Iran with a high incidence of oesophageal cancer. J Stored Prod Res.1998; 24: 39-50.
17. Desjardins AE, Manandhar G, Plattner RD, Maragos CM, Shrestha K, et al. Occurrence of Fusarium species and mycotoxins in Nepalese maize and wheat and the effect traditional processing methods on mycotoxin levels. J Agricult Food Chem 48:2000; 1377-1383.
18. Awan AJ, Aslam MS . Family Acantheceae and Genus Aphelandra: Ethnopharmacological and phytochemical review. Int J Pharma Pharm Sci 6:2014; 44-45.
19. Kumar D, Bhat ZA, Kumar V, Chashoo IA, Khan NA, et al. Pharmacognostical and phytochemical evaluation of Angelica Archangelica Linn. Int J Drug Dev & Res 3.2011: 173-188 16.
20. Kumar D, Bhat ZA, Shah MY. Effect of successive extracts of Stachys tibetica Vatke (Lamiaceae) in anxiety. Opem.2012; 12: 247-253
21. Mahdavi R, Nikniaz Z, Rafraf M, Jouyban A. Determination and comparison of total polyphenols and vitamin C content of natural fresh and commercial fruit juices. Pak J of Nutri.2010; 9: 968-972.
22. Zhishen J, Mengcheng T, Jianming W. The determination of flavonoid contents in mulberry and their scavenging effects on superoxide radicals. Food Chem.1999 64: 555-559.
23. Chaudhary A, Sharma U, Vig AP, Singh B, Arora S. Free radical scavenging, antiproliferative activities and profiling of variations in the level of phytochemicals in different parts of broccoli (Brassica oleracea italica). Food Chem.2014; 148: 373-380.
24. Marin S, Homedes V, Sanchis V, Ramos AJ, Magan N. Impact of Fusarium moniliforme and F proliferatum colonisation of maize on calorific losses and fumonisin production under different environmental conditions. J Stored Prod Res 1999;35: 15-26.
25. Janardhana GR, Raveesha KA, Shetty HS. Mycotoxin contamination of maize grains grown in Karnataka (India). FCT .1999;37: 863-868.
26. Jessica A., Rao M. R. K., Anthony J., Prabhu K, Johnson W.M.S., Balasubramanian B.S., Sundaram L., Dinakar, S. The GC-MS Study of One Ayurvedic Preparation Katakakhadiradi Kashayam. Int. J. Pharm. Sci. Rev. Res.,2016; 39(2): 216-224.

27. Keerthiga ,M Anand, S. P. Bioactive Compound Evaluation of Ethanol Extract from Geodorum densiflorum (Lam.) Schltr. by GC-MS analysis. International Journal of Pharmacological Research,2015; 5 (6): 139-144.
28. Kumar R.N., Vasantha K. and Mohan V.R. GC-MS Analysis of Bioactive Components of Tubers of Ruellia tuberosa L. (Acanthaceae). American Journal of Phytomedicine and Clinical Therapeutics,2014; 2(2): 209-216.
29. Lakshmi Ch. S., Uma A., Lakshminarasu M., Venkanna B. Evaluation of Antimicrobial Property of Thespesia Populnea Root Extracts against Genitourinary Tract Infectious Pathogens. International Journal for Pharmaceutical Research Scholars,2014; 3(1): 505-519.
30. Madhusoodanan P.V., Singh.N.P. Lepidagathis keralensis, Kew Bull.11992; 47(2): 301
31. Meenakshi V.K., Gomathy S., Senthamarai S., Paripooranaselvi M., Chamundeswari K.P. GC-MS determination of the bioactive components of Microcosmus exasperatus heller, 1878.J. Curr. Chem. Pharm. Sc,2012; 2(4): 271-276.
32. Muthulakshmi A., Margret J.R., & Mohan V.R. GC-MS Analysis of Bioactive Components of Feronia elephantum Correa (Rutaceae). Journal of Applied Pharmaceutical Science,2012; 02 (02: 69-74.
33. Villanova, PA., National Committee for Clinical Laboratory Standards. Performance Standards for Antimicrobial Disk Susceptibility., Approved Standard M2-A5.1993; 1-10
34. Prasad S. K. Exploration and elucidation of traditional medicinal plants of erstwhile Tulunadu and surrounding area of Kerala and Karnataka handle.net;2012;419-51.
35. Reddy P. A., Rao J.V. A review of Lepidagathis cristata.Int.Res.J.Pharm.2013; 4(11): 6-8.
36. Sasikala K., Mohan S.C. (2014). Total phenolic, flavonoid contents and GC-MS analysis of Canthium coromandelicum leaves extract. International Journal of Pharmacy and Pharmaceutical Sciences, 6(8): 379-381
37. Sharmila. S, Nalli. R, Surumbayee. M, Ramya. EK. GC-MS Analysis of bio-active components in petroleum ether extract of Lepidagathisscariosa (Nees.)-Acanthaceae. IJPR.2019 Feb;54(1):56-63.
38. Raviraj Anand Devkar, Shilpee Chaudhary, Sahithi Adepu, Seena Kanniparambil Xavier, Kodangala Subraya Chandrashekar, Maganahalli Manjunath Setty. Evaluation of antiurolithiatic and antioxidant potential of *Lepidagathis prostrata*: A Pashanbhed plant. Pharma Biol: 2016;54(7).1237-1245.
39. Rajeev Rattan, Amita Kumari, Veena Gautam, Bharat Inder Fozdar, Upendra Sharma, Dinesh Kumar. Preliminary phytochemical screening, antioxidant, and antifungal activity of *Lepidagathis cuspidata*. Int. J. Drug Dev & Res: 2016; 8(1). 01-03.
40. Malika FA, Rafi SM, Naqui B. Studies of antibacterial activity of ethanolic extract from Nericum indicum and Hibiscus rosasinensis. J Islamic Acad Sci. 1994;7: 167-168.
41. Alzoreky NS, Nakahara Antibacterial activity of extracts from some edible plants commonly consumed in Asia. IJFM .2003;80: 223-230.
42. Sawadogo Wamtinga Richard, Lompo Marius, Some Noya, Guissou Innocent Pierre, Nacoulma-Ouedraogo Odile Germaine. Anti-inflammatory, Analgesic, and Antipyretic effects of *Lepidagathis anobrya* Nees (Acanthaceae). Afr.J. Tradit Complement Altern Med: 2011; 8(4). 420-424.
43. Brand WW, Cuvelier ME, Berset C. Use of the free radical method to evaluate antioxidant activity. Lebensmittel-Wissenschaft und-Technol.1995; 28: 25-30.
44. Bursal E, Gulcin I. Polyphenol contents and in vitro antioxidant activities of lyophilized aqueous extract of kiwifruit (Actinidia deliciosa). Food Res Int.2011; 44: 1482-1489.
45. Leena P.*, Zeinul Hukuman N.H. and Jisha M. In Vitro Antimicrobial Efficacy and Gc- Ms Analysis of Bioactive Components from Lepidagathis Keralensis (Acanthaceae. World Journal of Pharmaceutical Research .2016;5(12), 937-948.