

# In Vitro Antimicrobial Activity and Phytochemical Analysis of Leaf Extracts of *Murraya Koenigii* L.

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**Abstract:** Plants have served as a vital source of medicine since the dawn of human civilization. The demand for plant-derived medicines, health products, pharmaceuticals, dietary supplements, and cosmetics is on the rise. *Murraya koenigii*, commonly known as the curry leaf tree, is a versatile tree that provides one of the medicinal products. Various parts of *M. koenigii* are utilized in traditional medicine for treating a range of ailments. It has been demonstrated to have significant wound healing properties and exhibits antioxidant activity with a high level of radical-scavenging capabilities. This article aims to present an overview of the chemical constituents found in the crude leaf extracts of *M. koenigii*, with a particular focus on their pharmacological effects. Qualitative phytochemical screening was conducted using crude leaf extracts in three different solvents: water, alcohol, and chloroform. The phytochemical analysis of the extracts indicated the presence of glycosides, alkaloids, oils, saponins, and flavonoids.

**Keywords:** *Murraya koenigii*, phytochemical analysis, antimicrobial

## I. INTRODUCTION

*Murraya koenigii*, commonly known as curry leaf, is a widely utilized spice and condiment in tropical regions, appreciated for its capacity to maintain flavor and other essential characteristics even post-drying. The key components that contribute to its unique flavor and aroma are pinene, sabinene, caryophyllene, cardinol, and cardinene.[1] Beyond its culinary uses, this plant has a long-standing history of being employed for its tonic and stomachic benefits. It is recognized for containing a variety of bioactive phytochemicals, such as lutein,  $\beta$ -carotene, phenolics, essential oils, minerals, proteins, and terpenoids, all of which enhance its medicinal and nutritional value.[2] Since ancient times, individuals have been investigating the natural world, particularly plants, in pursuit of new medicinal compounds. This exploration has led to the utilization of a vast array of medicinal plants known for their healing properties to address various ailments. [3] A survey conducted by the WHO indicates that 80% of the population in developing nations depend solely on traditional medicine for their primary healthcare needs, with a significant portion involving the application of plant extracts.[4]. Research on plants persists primarily to uncover novel secondary metabolites or phytochemicals, which are non-essential nutrients derived from plants that exhibit numerous protective functions for human consumers. *Murraya koenigii*, a small evergreen tree belonging to the Rutaceae family, is indigenous to India and is also found in Sri Lanka and other South Asian countries. [5] Various parts of *M. koenigii* are utilized in traditional medicine for treating a range of diseases. It has been demonstrated to possess considerable wound healing capabilities. Commonly referred to as the curry leaf tree, *Murraya koenigii* exhibits antioxidant activity with a significant capacity for radical-scavenging. [6]

Phytochemical screening is a technique that uncovers specific components or properties readily available in plants for bioactivity or ethnomedicinal applications. Plant-based antimicrobials hold immense therapeutic potential, as they can fulfil their purpose with fewer side effects typically associated with synthetic antimicrobials. Therefore, it is expected that phytochemicals with sufficient antibacterial efficacy can be employed in the treatment of bacterial infections. [7] The antioxidant and antimicrobial properties of various extracts from numerous plants have recently garnered significant interest in both research and the food industry due to their potential application as natural additives to replace synthetic antioxidants and antimicrobials with their natural counterparts. Therefore, medicinal plants are significant in the advancement of new pharmaceuticals due to their efficacy, reduced side effects, and comparatively lower cost in relation to synthetic medications.[8, 9] This study seeks to investigate the phytochemical components, as well as the antibacterial and antifungal characteristics of the crude leaf extracts from *Murraya koenigii*.

## II. MATERIAL AND METHODS

The fully matured fresh leaves of *M. koenigii* were gathered from the college campus at Dr. L.K.V.D College, Tajpur, Samastipur, India. The leaves underwent thorough washing, were shade dried, and then finely powdered. The dried powdered leaves were subjected to extraction using three different solvents: water, acetone, and chloroform. For the aqueous extraction, 10 g of the powdered leaves were combined with 100 ml of distilled water, boiled for approximately two hours, and subsequently filtered. In contrast, the acetone and chloroform extracts were prepared by separately mixing ten grams of the powdered leaf samples with 100 ml of each solvent in a mechanical shaker for 48 hours at room temperature. The extracts were then filtered, concentrated, dried, and stored in the refrigerator at 4°C for future use.

**Phytochemical analysis:** The plant extracts that were prepared were examined for the presence of alkaloids, glycosides, saponins, proteins, amino acids, fixed oils, phenolic compounds, tannins, flavonoids, gum, and mucilage, among others. [10]

**Preparation of plant extract for antimicrobial screening:** For the purpose of antimicrobial screening, the concentrated, dried, and powdered ethanol leaf extract was dissolved in 10% dimethyl sulfoxide (DMSO) and stored at 40°C for future use.

**Test Organisms:** Antibacterial activity was assessed against two chosen gram-negative pathogens, specifically *Escherichia coli* and *Pseudomonas aeruginosa*, while antifungal activity was evaluated against two clinical fungal isolates, namely *Candida albicans* and *Aspergillus niger*. The strains utilized in this study were sourced from the Dr. Rajendra Prasad Central Agricultural University Pusa, Samastipur, India. To evaluate the biological significance and efficacy of the plant part, the minimal inhibitory concentration was determined using the Agar cup method.

**Antibacterial activity:** Petri dishes containing 20 ml of Muller Hinton medium were inoculated with a 24-hour-old culture of bacterial strains, including *E. coli* and *P. aeruginosa*. Wells, approximately 10 mm in diameter, were created using a well cutter, and 25 µl, 50 µl, and 100 µl of the extracts were introduced into the wells from a stock solution of 0.1 g/1 ml. The plates were subsequently incubated at 37°C for 24 hours. The antibacterial activity was evaluated by measuring the diameter of the inhibition zone in millimeters that formed around the wells. [11] Gentamicin (a standard antibacterial agent, concentration: 20 mg/ml) was utilized as a positive control.

**Antifungal activity:** Petri dishes containing 20 ml of Muller Hinton medium were inoculated with a 24-hour-old culture of bacterial strains, including *E. coli* and *P. aeruginosa*. Wells, approximately 10 mm in diameter, were created using a well cutter, and 25 µl, 50 µl, and 100 µl of the extracts were introduced into the wells from a stock solution of 0.1 g/1 ml. The plates were subsequently incubated at 37°C for 24 hours. The antibacterial activity was evaluated by measuring the diameter of the inhibition zone in millimetres that formed around the wells. [12] Gentamicin (a standard antibacterial agent, concentration: 20 mg/ml) was utilized as a positive control.

## III. RESULT AND DISCUSSIONS

### Phytochemical analysis

Table 1 illustrates the different phytochemical constituents found in the leaf extracts of *M. koenigii*. The phytochemical analysis of all three extracts indicates that the acetone and water extracts of the leaf samples yielded more favorable results for glycosides, oils, saponins, and flavonoids.

TABLE 1. Phytochemical Analysis of *Murraya koenigii* Leaf Extracts

Phytochemicals	Glycosides	Phytosterols	Alkaloids	Oils	Saponins	Phenols	Flavanoids
Water	-	-	+	+	+	+	+
Acetone	+	-	-	+	+	+	+
Chloroform	+	-	-	+	+	+	-

+ : Present, - : Absent

The preliminary phytochemical analysis indicated the existence of six compounds (Table 1), namely flavonoids, glycosides, oils, saponins, phenolic, gum, and mucilage. Flavonoids, glycosides, oils, and saponins were identified in the acetone and chloroform extracts. Traditionally, saponins have been widely utilized as detergents, pesticides, and molluscicides, in addition to their industrial uses as foaming and surface-active agents, and they are also recognized for their beneficial health effects. [13] Flavonoids extracted from the aqueous solution of *M. koenigii* demonstrate antimicrobial properties. Previous studies have reported that the plant contains glycosides, alkaloids, saponins, flavonoids, tannins, carbohydrates, phenolic compounds, and phytosterols.[14, 15]

**Antibacterial activity**

The antibacterial properties of *M. koenigii* (leaf ethanol extract with DMSO) were evaluated in vitro using the agar cup method against clinical isolates of *E. coli* and *P. aeruginosa*. The table provided illustrates the microbial growth inhibition caused by the ethanol leaf extracts of *Murraya koenigii*. Among the different concentrations of leaf extracts tested, the higher concentrations demonstrated the greatest antibacterial activity against both isolates. Table 2 presents the zones of inhibition created by the extracts against the bacterial strains on Muller Hinton agar. The antibacterial activity sequence of the leaf extract against *E. coli* showed no effect at 25µl, but it resulted in inhibition zones of 13 mm and 19 mm at 50µl and 100µl concentrations, respectively. In the case of *P. aeruginosa*, the plant extract demonstrated no activity at both 25µl and 50µl concentrations, yet it produced a 17 mm inhibition zone at 100µl concentration.

TABLE 2. Zone Diameter of Inhibition of Ethanol Leaf Extract of *Murraya koenigii*

Organisms	Concentration of leaf Extracts ( in mm)			Control (positive)
	25	50	100	
<i>Escherichia coli</i>	Nil	13	19	20
<i>Pseudomonas aeruginosa</i>	Nil	Nil	17	20

Therefore, the antibacterial activity was observed to vary with different concentrations. A higher concentration of the leaf extract exhibited the most significant antibacterial effect. The findings may be deemed adequate for further investigations aimed at isolating and identifying the active principles, as well as assessing the potential antimicrobial properties of extracts from other parts of *Murraya koenigii*. Previous research also indicates that extracts from various parts of *M. koenigii* are utilized against microbial infections due to the presence of secondary metabolites, including phenols, essential oils, terpenoids, alkaloids, and flavonoids demonstrate strong antibacterial activity.[16]

**Antifungal activity**

To evaluate the biological importance and efficacy of the plant extract, the antifungal activity of *M. koenigii* (leaf ethanol extract with DMSO) was tested in vitro using the agar cup method against two clinical fungal isolates, namely *C. albicans* and *A.niger*. The table provided illustrates the antifungal activity of the plant species.

TABLE 3. Zone Inhibition of Ethanol Leaf Extract of *Murraya koenigii*

Organisms	Concentration of leaf Extracts ( in mm)			Control (positive)
	25	50	100	
<i>Candida albicans</i>	Nil	Nil	13	25
<i>Aspergillus niger</i>	Nil	Nil	13	25

The antifungal activity sequence of leaf extract against *C. albicans* and *A. niger* showed no activity at both 25µl and 50µl concentrations, but resulted in a 13 mm zone of inhibition at 100µl concentrations respectively (Table

3). This study indicates that the ethanol leaf extracts of *M. koenigii* demonstrated greater efficacy against clinical bacterial pathogens such as *E. coli* and *P. aeruginosa*. The antifungal activity was found to be minimal in comparison to the antibacterial activity. Previous literature has suggested that the antibacterial properties are attributed to various chemical agents present in the leaf extract, including essential oils, flavonoids, terpenoids, and other components classified as active antimicrobial compounds. [17] The findings of this study partially support the use of traditional medicinal plants in the treatment of human and animal diseases and strengthen the concept of an ethnobotanical approach in identifying plants as potential sources of bioactive substances. [18] The aqueous extract typically displays a significant level of antibacterial activity, which appears to validate the traditional therapeutic claims associated with this plant.[19]

#### IV. CONCLUSION

Medicinal plants are vital for human health due to their active phytochemical compounds with pharmacological effects. This study finds that *M. koenigii* leaves are rich in phytochemicals, though their composition varies among samples, with most components present in aqueous extracts. Secondary metabolites like glycosides, phytosterols, alkaloids, oils, saponins, phenols, and flavonoids contribute to the leaves' antibiotic properties and confirm their antimicrobial effectiveness against certain pathogens.

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