Study on the Effectiveness of Murraya Koenigii as an Antimicrobial Agent

Harshita Sisodia¹, Pravina Rathore²

¹Bhupal Nobles’ University, Udaipur, Rajasthan, India
Email: harshitasisodia97[at]gmail.com

²Bhupal Nobles’ University, Udaipur, Rajasthan, India
Email: drpravin[at]bnuniversity.ac.in

Abstract: The Murraya Koenigii plant, also known as the Curry leaf plant, is highly valued for its medicinal properties and bioactive compounds. Its various parts, such as the roots, leaves, stems, bark, fruits, and seeds, have been traditionally used to combat infections and strengthen the human system. The leaves of the plant are particularly popular for their aroma and are used as a flavoring agent in curries. This study assessed the in-vitro antimicrobial efficacy of leaf extracts of Murraya Koenigii against bacterial pathogen Staphylococcus aureus and fungal pathogen Alternaria solani using the Agar well diffusion method. The results showed that the acetone extract of the plant's leaves was more effective than aqueous extract. The acetone leaf extracts exhibited the highest inhibitory activity, with a minimum inhibitory concentration value of 0.2mg/ml for both fungal and bacterial pathogens, while the aqueous leaf extracts showed a MIC value of 0.8mg/ml for S. aureus and 0.2mg/ml for A. solani. The study suggests that the Murraya Koenigii plant extracts are a natural source of bioactive compounds that can be used as potent antimicrobial drugs to combat the emerging problem of pathogenic microorganisms' antibiotic resistance.

Keywords: Antimicrobial activity, Pathogen, Staphylococcus aureus, Alternaria solani.

1. Introduction

Folk medicine has been found to provide effective remedies for various illnesses with minimal risk of side effects (Zhang et al., 2016). After the emergence of modern medicine, the utilization of herbal medicine experienced a decline. However, in the past two or three decades, there have been significant advancements in the field of phytochemistry and the identification of plant compounds that exhibit efficacy against certain chronic diseases and multidrug resistant bacteria. This resurgence in interest for herbal medicine has been prompted by the global demand for alternative therapeutic options, especially in developing countries where infectious diseases are a major cause of mortality. The indiscriminate use of commercial antimicrobial drugs has contributed to the development of multidrug resistance, making it more difficult to treat infectious diseases effectively. In addition to the aforementioned concerns, antibiotic administration can have adverse effects on the host, such as immune suppression, hypersensitivity, and allergic reactions (Werner et al., 1999). Furthermore, environmental safety is a critical issue in many countries, and the use of synthetic agrochemicals carries the potential for causing harmful ecological impacts, including risks to human health, toxicity to non-targeted beneficial organisms, and environmental pollution. The utilization of natural products presents a promising approach to mitigate risk and address the need for new antimicrobial agents. Plant-based compound screening has yielded a wealth of active substances, leading to the discovery of novel therapeutic drugs that exhibit effective preventive and curative properties against a range of diseases, including cancer (Kumar et al.2004; Sheeja & Kuttam 2007). Phytochemical constituents found in medicinal plants possess bioactive properties, including antioxidant, insecticidal, and antibacterial activities. The various components of folk medicine, including extracts from different parts of plants, have been shown to possess significant antimicrobial activity without causing any serious adverse effects to the host.

Plants that are classified under the Rutaceae family can be herbs, shrubs, or trees with glandular punctuate. They have a distinct aroma and are comprised of approximately 150 genera and 1500 species. These plants are typically characterized by the presence of spines and winged pétioles. For ages, the leaves of Murraya Koenigii (also called “Meetha neem”) have been a popular ingredient in Indian curries, valued for their distinctive flavor. In addition to the leaves, the bark and roots of this plant can also serve as a tonic and stomachic.

The macroscopic investigation has shown that the leaves of are obliquely ovate or somewhat rhomboid in shape, with an apex that can be pointed, blunt, or sharp. These leaves are bipinnately compound and arranged alternately, lacking stipules. In addition, microscopic examination has revealed the distribution of stomata on the abaxial surface, whereas the adaxial surface lacks stomata. The type of stomata identified was anomocytic.

The examination revealed the presence of uniseriate multicellular trichomes on both surfaces of the leaf, with a higher frequency on the upper surface of the midrib region. Furthermore, upon analyzing the transverse section of the leaf, it was observed that the outermost layer of the epidermis was composed of rectangular cells on both the upper and lower surfaces. The upper epidermis was covered with a deposition of cuticle.

Phytochemical investigation has revealed the presence of both alkaloids and volatile oil in the leaves of the plant. The plant has been traditionally utilized as a stimulant, stomachic, febrifuge, and analgesic, as well as for treating...
diarrhea, dysentery, and insect bites. According to reports, the leaves possess significant medicinal properties, including antibacterial, anti-inflammatory, and anti-feedant effects. Chemical tests were used to carry out an initial investigation of the phytochemical constituents, aiming to detect a variety of secondary metabolites, such as phytosterols, coumarins, alkaloids, flavonoids, and carbohydrates.

In order to conduct an initial screening of phytochemicals, chemical tests were employed to identify various secondary metabolites, including alkaloids, phytosterols, flavonoids, coumarins, and carbohydrates. The essential oil obtained from the leaves of Murraya Koenigii demonstrated significant antimicrobial activity against Bacillus subtilis, Streptococcus aureus, Mycobacterium tuberculosis, and Corynbacterium pyogenes.

Murrayacine, an alkaloid, has been identified in Murraya Koenigii, an endangered medicinal plant that has been traditionally valued for its digestive, carminative, anthelmintic, and laxative properties. This species has a long history of traditional medicinal use and possesses therapeutic properties that may be beneficial for managing diabetes, heart disease, neurological disorders, cancerous tumors, and liver disorders. Additionally, its seeds have been used for their wound-healing, antioxidant, anti-inflammatory, analgesic, and contraceptive activities.

In recent years, a multitude of scientific and technological fields have dedicated themselves to conducting experiments aimed at advancing human growth and welfare of mankind. Within the medical field, numerous systems are being practiced in India, including Ayurveda, Unani, Siddha, Amchi, and local health traditions. One rapidly evolving area of medicine is nanomedicine, which utilizes nanotechnology for the diagnosis, treatment, and prevention of diseases at the molecular and cellular level. This approach holds great promise for achieving improved therapeutic efficacy and reduced toxicity, as well as enhanced specificity and targeting capabilities.

The initial efficacy of antibiotics in the treatment of microbial infections was highly encouraging, leading to their widespread use in clinical practice.

Rapid development of antibiotic resistance followed shortly after the introduction of these agents, posing a persistent and menacing threat to the efficacy of antimicrobial treatments. Finding a solution to the issue of antimicrobial resistance is a critical imperative and requires urgent action. Natural products are considered a highly advantageous class of compounds that have undergone evolutionary processes in order to selectively engage with a diverse array of protein targets for specialized functions.

In this research, various extractions of Murraya Koenigii leaves were generated and subjected to assessments in order to evaluate the effectiveness of the leaves against microorganisms.

2. Material and Methods

Plant Material
During the summer season, fresh leaves of the Murraya Koenigii plant were collected from the Udaipur area. Taxonomic identification of the plant was confirmed, and the leaves were subsequently prepared for further investigation. The collected leaves were thoroughly washed using running water for 2-3 cycles, followed by a rinse with distilled water. The washed leaves were dried under shaded conditions for a period of 25-30 days. The dried leaves were pulverized into a fine powder and stored in a sterilized container at room temperature for subsequent use in scientific investigation.

Extraction of Plant Material
The plant material extraction was performed using a Soxhlet apparatus with aqueous and acetone solvent (fig. a). Different concentrations of the extracted sample were prepared, including 0.2 mg/ml, 0.4 mg/ml, 0.6 mg/ml, 0.8 mg/ml, and 1.0 mg/ml.

Figure (a): Soxhlet apparatus

Microorganisms
Staphylococcus aureus and Alternaria solani are the two microorganisms that were used in the experiment.

Antimicrobial activity test
Using the surface spread plate technique, the bacterial and fungal inoculum was evenly dispersed on the media surfaces of Nutrient Agar and Potato Dextrose Agar plates, which were employed for the inoculation of the aforementioned microorganisms. Agar well diffusion was the approach that Alade and Irobi utilised to figure out whether or not each crude extract has antimicrobial characteristics. After the process of autoclaving was finished, the medium is allowed to cool before being placed into petri dishes. Prior to use, the plates were pre-incubated at 35 degrees Celsius to guarantee sterility. A sterile spreader was employed to achieve the objective of uniformly dispersing the test inoculum throughout the solidified agar surface. Five wells of identical size were aseptically bored into the agar plate. To determine the total antimicrobial activity, the diameters of the inhibition zones were measured in millimeters.
3. Results

Determination of antimicrobial activity of *Murraya Koenigii*

The current investigation aimed to evaluate the antimicrobial potential of *Murraya Koenigii* extracts, which were prepared using various solvents, against two distinct types of pathogens: the bacterial pathogen *Staphylococcus aureus* and the fungal pathogen *Alternaria solani*. Based on the findings presented in Table 1, it was observed that of the two Murraya extracts tested, the acetone extract exhibited the most potent and extensive antimicrobial activity against the microorganisms studied. In order to quantitatively evaluate the antimicrobial activity of the Murraya Koenigii extracts (Aqueous and Acetone), the diameter of the Zone of inhibition was measured at five different concentrations of 0.2mg/ml, 0.4mg/ml, 0.6mg/ml, 0.8mg/ml and 1.0mg/ml.

**ZOI for *S. aureus* (Aqueous and Acetone extracts)**

According to the results, the aqueous extracts of *Murraya Koenigii* did not exhibit any ZOI (zone of inhibition) at concentrations of 0.2mg/ml, 0.4mg/ml, and 0.6mg/ml, whereas a ZOI with diameter of 8mm and 10mm was detected for the other tested concentrations. As shown in Table 1. The acetone leaf extract of Murraya was more potent against *Staphylococcus aureus* when compared to the aqueous leaf extract ranged from 12mm at 0.2mg/ml to 22mm at 1.0mg/ml, while the aqueous extract exhibited less significant antimicrobial activity.

**Table 1: Antimicrobial activity of *M. Koenigii* extracts against *S. aureus***

<table>
<thead>
<tr>
<th>Different concentrations (mg/ml)</th>
<th>Aqueous extract (ZOI in mm)</th>
<th>Acetone extract (ZOI in mm)</th>
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<tbody>
<tr>
<td>0.2</td>
<td>-</td>
<td>12</td>
</tr>
<tr>
<td>0.4</td>
<td>-</td>
<td>14</td>
</tr>
<tr>
<td>0.6</td>
<td>-</td>
<td>16</td>
</tr>
<tr>
<td>0.8</td>
<td>8</td>
<td>17</td>
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<tr>
<td>1.0</td>
<td>10</td>
<td>22</td>
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**ZOI for *A. solani* (Aqueous and Acetone extracts)**

Among the extracts assayed the acetone leaf extract of the plant demonstrated the highest ZOI of 16mm at a concentration of 1.0mg/ml against *Alternaria solani* compared to aqueous extract of the same plant as shown in Table 2. The acetone leaf extracts displayed the most potent inhibitory activity, with a minimum inhibitory concentration (MIC) value of 0.2mg/ml against both fungal and bacterial pathogen. In contrast, the aqueous leaf extract exhibited an MIC value of 0.8mg/ml against *S. aureus* and 0.2mg/ml against *A. solani*.

4. Conclusion

Assessment of the antimicrobial activity of medicinal plants that possess pharmacological properties is necessary to utilize them as a potential source of efficacious natural drugs. The present investigation aimed to assess the antimicrobial properties of *Murraya*, a medicinal plant, against fungal and bacterial pathogen in various solvents. The results revealed that the acetone extract of the plant species exhibited the most substantial inhibitory activity compared to the aqueous extract. Based on this finding, this study concludes that *Murraya Koenigii* possesses significant antimicrobial potential that can be further explored for medicinal drug development. As microbial resistance to antibiotics continues to be growing concern, the use of this plant as a natural source for the development of potent antimicrobial drugs in future could prove to be a valuable alternative.

**References**


