Evaluation of Antimicrobial Potential of Local Plant Extracts Against Common Pathogenic Microorganisms

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***Abstract*—This study evaluates the antimicrobial potential of crude extracts from selected medicinal plants locally available in the Shirur Kasar region of Maharashtra, India. In the wake of rising multidrug resistance among pathogenic bacteria, exploring natural phytochemical compounds has become crucial. In this research, aqueous and ethanolic extracts of Azadirachta indica (Neem) and Oscimum sanctum (Tulsi) were screened against common pathogens including Escherichia coli, Staphylococcus aureus, and Pseudomonas aeruginosa. The antimicrobial activity was assessed using the agar well diffusion method. The ethanolic extracts demonstrated a significantly higher zone of inhibition compared to aqueous extracts. Staphylococcus aureus was found to be the most susceptible organism, showing a maximum zone of inhibition of 18 mm with ethanolic Neem extract. The results validate the traditional use of these plants and suggest that local flora contains potent bio-active molecules that can be exploited for future drug development.**

***Index Terms*—Microbiology, Antimicrobial Activity, Plant Extracts, Shirur Kasar, Pathogens, Agar Well Diffusion.**

I. INTRODUCTION

The rapid emergence of multiple drug-resistant (MDR) bacterial strains poses a severe threat to global public health. Overuse and misuse of conventional antibiotics have accelerated the evolutionary pressure on pathogens, rendering many standard treatments ineffective. Consequently, there is an urgent need to identify novel, effective, and safe antimicrobial agents.

Plants have been a rich source of medicine for thousands of years. The Indian traditional system of medicine, Ayurveda, heavily relies on plant-based formulations. Phytochemicals such as alkaloids, flavonoids, tannins, and terpenoids naturally synthesized by plants serve as a defense mechanism against microbial infections.

The geographical region of Shirur Kasar in the Beed district of Maharashtra possesses a unique semi-arid climate that influences the secondary metabolite profile of local flora. While plants like Azadirachta indica (Neem) and Ocimum sanctum (Tulsi) are widely known for their therapeutic properties, a systematic microbiological evaluation of samples specific to the microclimate of Shirur Kasar remains limited. This paper aims to isolate, extract, and quantitatively analyze the efficacy of these local plant extracts against standard Gram-positive and Gram-negative bacterial pathogens.

II. MATERIALS AND METHODS

A. Collection and Identification of Plant Material

Healthy, disease-free leaves of Azadirachta indica and Ocimum sanctum were collected from various eco-zones within Shirur Kasar. The plant materials were thoroughly washed with tap water followed by distilled water to remove surface dust and debris. The leaves were then shade-dried at room temperature (25°C to 28°C) for 10-12 days until they became brittle. The dried leaves were pulverized into a fine powder using a sterile mechanical grinder and stored in airtight containers.

B. Preparation of Extracts

Two types of solvents were used for extraction: organic (Ethanol) and aqueous (Distilled Water).

1. Ethanolic Extract: 20g of leaf powder was soaked in 100 mL of 95% ethanol in a conical flask, kept

on a rotary shaker for 48 hours. The mixture was filtered through Whatman No. 1 filter paper.

2. Aqueous Extract: 20g of leaf powder was boiled in 100 mL of sterile distilled water for 2 hours, allowed to cool, and filtered using the same procedure.

Both extracts were concentrated using a vacuum evaporator at reduced pressure, and the final crude yield was preserved at 4°C for further microbiological assays.

C. Test Microorganisms

The bacterial strains utilized in this study included:

- Staphylococcus aureus (Gram-positive)
- Escherichia coli (Gram-negative)
- Pseudomonas aeruginosa (Gram-negative)

The cultures were maintained on Nutrient Agar (NA) slants and sub-cultured periodically.

D. Antimicrobial Screening via Agar Well Diffusion

The antimicrobial activity was determined using the standard agar well diffusion method.

- Nutrient agar plates were prepared and inoculated with $100\ \mu\text{L}$ of 24-hour-old bacterial broth culture adjusted to 0.5 McFarland standard.
- Wells of 6mm diameter were bored into the agar medium using a sterile corn borer.
- $50\ \mu\text{L}$ of each plant extract (at concentrations of 50 mg/mL and 100 mg/mL) was introduced into respective wells.
- Standard antibiotic disks (Ciprofloxacin, $10\ \mu\text{g}$) were used as positive controls, while pure solvents served as negative controls.
- The plates were incubated at 37°C for 24 hours. The zone of inhibition (ZOI) was measured in millimeters (mm) using a digital caliper.

III. RESULTS AND ANALYSIS

The crude extracts exhibited varying degrees of antibacterial activity against the tested pathogens. Overall, ethanolic extracts demonstrated superior efficacy compared to their aqueous counterparts.

Plant Extract Type	Concentration (mg/mL)	ZOI against S. aureus (mm)	ZOI against E. coli (mm)	ZOI against P. aeruginosa (mm)
A. indica (Eth)	100	18.2	14.1	11.5
A. indica (Aq)	100	11.0	9.4	7.0
O. sanctum (Eth)	100	15.5	12.0	10.2
O. sanctum (Aq)	100	9.2	8.0	6.5
Ciprofloxacin (+)	$10\ \mu\text{g}$	24.5	22.1	19.8

As indicated in the data table, the ethanolic extract of Azadirachta indica at 100 mg/mL concentration displayed the most prominent zone of inhibition (18.2mm) against Staphylococcus aureus. Gram-negative bacteria (E. coli and P. aeruginosa) displayed higher resistance to the extracts, which can be chemically attributed to the presence of an outer lipopolysaccharide membrane that restricts the entry of hydrophobic compounds.

IV. DISCUSSION

The results of this study clearly indicate that the phytochemical profiles of plants grown in the Shirur Kasar region hold robust biological activity against pathogenic bacteria. The superior performance of ethanolic extracts over aqueous extractions implies that the active antimicrobial compounds (such as flavonoids, polyphenols, and azadirachtin) are highly soluble in organic solvents.

Staphylococcus aureus, being Gram-positive, lacks the outer protective lipid membrane found in Gram-negative bacteria like E. coli. This structural distinction allows the plant-derived secondary metabolites to easily disrupt the bacterial cell wall, leading to cell lysis (breaking down of the cell) and

cytoplasmic leakage. The moderate zone of inhibition against Pseudomonas aeruginosa is particularly interesting, given that this organism is notorious for its high intrinsic resistance to most commercial antibiotics.

V. CONCLUSION AND FUTURE SCOPE

This research conducted under the collaboration of Chhatrapati Sambhajiraje Management Science College and Shilpkar College, Shirur Kasar, establishes that local ecological populations of Neem and Tulsi are promising sources of natural antimicrobial compounds. These extracts could safely complement synthetic antibiotics to counter drug resistance.

Future studies will focus on:

1. Thin-Layer Chromatography (TLC) and Gas Chromatography-Mass Spectrometry (GC-MS) to precisely isolate and identify the active molecular compounds.
2. Evaluating the synergistic effects of combining these plant extracts with lower doses of standard commercial antibiotics.
3. In-vivo toxicity testing to check the biocompatibility of these extracts for potential pharmacological applications.

REFERENCES

[1] K. R. Aneja, *Experiments in Microbiology, Plant Pathology and Biotechnology*, 4th ed. New Delhi, India: New Age International Publishers, 2014.

[2] J. B. Harborne, *Phytochemical Methods: A Guide to Modern Techniques of Plant Analysis*. London, U.K.: Chapman and Hall, 1998.

[3] S. S. Patil and R. G. Tote, "Microbial flora of semi-arid regions of Marathwada," *Journal of Environmental Biology*, vol. 42, no. 3, pp. 211–217, 2021.

[4] R. H. Jadhav, "Screening of medicinal plants for phytochemical compounds," *International Journal of Phytomedicine*, vol. 14, no. 2, pp. 89–95, 2023.

[5] Clinical and Laboratory Standards Institute (CLSI), *Performance Standards for Antimicrobial Susceptibility Testing*, 30th ed., CLSI Supplement M100, 2020.