

ANTIMICROBIAL ACTIVITY OF HYDROGELS OF MALABAR SPINACH ON HUMAN SKIN MICROBIAL ISOLATE

Vidya J , Josvitha Dsouza, Aysha Shamla, Kathija Shafa, Krishna Prasad

Nooralabettu,* Shabeeba V , and Krishna Prasad Nooralabettu

Department of Biotechnology, P. A. College of Engineering, Karnataka, Mangaluru, India

E-mail: nooralabettu@gmail.com

Abstract

This study explores the antimicrobial potential of hydrogels derived from Malabar spinach (*Basella alba*) against *Staphylococcus aureus*, a common human skin microbial isolate. Known for its medicinal and nutritional properties, *Basella alba* was investigated for its ability to combat skin infections. Bioactive compounds were extracted from powdered leaves and stems using ethanol and aqueous solvents via Soxhlet extraction to enhance yield. The antimicrobial efficacy of these extracts was assessed through the disc diffusion method. Results demonstrated significant inhibitory effects, highlighting the potential of *Basella alba* hydrogels as a natural antimicrobial agent for managing skin-related microbial infections.

1 Introduction

Malabar spinach (*Basella alba*), a tropical vine belonging to the Basellaceae family, is known for its climbing growth habit and significant nutritional value. Native to South Asia and

Africa, it thrives in hot, humid climates, making it suitable for cultivation in tropical and subtropical regions [1, 2]. Distinct from traditional spinach (*Spinacia oleracea*), Malabar spinach is often favored for its heat tolerance and rapid growth, which contribute to its popularity among home gardeners and commercial cultivators [3, 4].

The plant's tender leaves and stems are widely consumed as a leafy vegetable, prized for their unique mucilaginous texture when cooked [3]. These parts are not only rich in vitamins A and C but also contain iron, calcium, and antioxidants, making them a vital dietary component in many cultures [5]. Beyond its nutritional benefits, Malabar spinach has garnered attention for its medicinal properties. Traditional medicine systems have employed this plant to treat a variety of ailments, including digestive issues and skin conditions [6, 7].

Recent scientific studies have further highlighted the potential of *Basella alba* in the pharmaceutical and biomedical fields. Research has demonstrated its antimicrobial [7, 8], anti-inflammatory [9], and antioxidant activities [10], suggesting its utility in developing natural therapeutic agents. In particular, the antibacterial properties of *Basella alba* extracts have been investigated against various pathogens, underscoring its relevance in addressing microbial resistance [8, 11, 12].

Phytochemical analyses reveal that the plant is a rich source of bioactive compounds, including saponins, flavonoids, and phenolic acids, which are responsible for its diverse biological activities [13, 14]. These findings underscore the importance of extracting and characterizing these compounds to understand their pharmacological potential better. Employing advanced techniques such as Soxhlet extraction and spectroscopy has enabled researchers to isolate and identify these constituents effectively [14, 15].

This study aims to evaluate the antimicrobial efficacy of *Basella alba* leaf extracts using established laboratory methodologies. By leveraging techniques such as the Kirby-Bauer disk diffusion assay [16] and in vitro antimicrobial activity testing [17], the research seeks to elucidate the plant's potential as a source of natural antimicrobial agents. Through these efforts, the study contributes to the growing body of knowledge on *Basella alba*, bridging

traditional uses with modern scientific exploration.

2 Experimental Procedure

2.1 Materials

2.1.1 Reagents and Chemicals

All chemicals and reagents used were of analytical grade, sourced from Gennext Chemicals, Mulki, Karnataka. The use of analytical-grade reagents ensured the purity and reliability of the experimental results

2.1.2 Plant Material: Malabar Spinach

Fresh Malabar spinach (*Basella alba*) leaves and stems were collected from local villages in the undivided Dakshina Kannada district, Karnataka. The plant material was washed thoroughly under running tap water to remove soil and dirt, as per Harborne's protocol [18]. After washing, the material was dried in a hot air oven at 65°C to preserve bioactive compounds.

2.1.3 Soxhlet Extractor

A Soxhlet extractor was used for solvent and aqueous extractions, ensuring efficient extraction of bioactive compounds Kumoro et al [19].

2.1.4 Ceramic Materials

Crushed ceramic pieces were added during aqueous extraction to minimize foaming, ensuring smooth operation

2.2 Methodology

2.2.1 Extraction Processes

Pre-treatment of Plant Material: The collected plant material was washed, dried in a hot air oven at 65°C for 24 hours, and ground into a fine powder using a mechanical blender. A measured 30 g of powdered material was used for extraction processes.

Solvent Extraction: The Soxhlet apparatus was set up using 300 mL of ethanol as the solvent [19]. Powdered leaves and stems were placed in a cellulose thimble and subjected to ethanol extraction. Ethanol was heated to its boiling point (78.37°C), and the vapor condensed to percolate through the plant material. The process was repeated for 16 cycles (40–45 minutes per cycle) until the solvent in the thimble became clear. The resulting extract was concentrated to 10 mL using a water bath.

Aqueous Extraction: For aqueous extraction, 30 g of powdered plant material was mixed with 300 mL of distilled water in a 500 mL round-bottom flask. Crushed ceramic pieces were added to reduce foaming during boiling. The Soxhlet apparatus was used continuously for 16 hours. The aqueous extract was filtered and concentrated to 10 mL using a water bath as described by Harborne[18].

Antimicrobial Activity Testing: The antimicrobial activity of the extracts was determined using the disc diffusion method, following the standardized protocol of Bauer et al [20]. Nutrient agar was prepared by dissolving 7 g of agar in 250 mL of distilled water. The solution was sterilized in an autoclave at 121°C for 15–20 minutes, poured into sterile Petri plates, and allowed to solidify.

Preparation of Test Samples: Whatman filter paper discs (6 mm diameter) were impregnated with 10 µL of each extract (aqueous and solvent extracts of leaves and stems) and dried under aseptic conditions.

Inoculation and Disc Placement: Isolated colonies of *Staphylococcus aureus* were streaked onto nutrient agar plates using an L-shaped spreader. Antibiotic discs (Kanamycin

1000 μg , Ampicillin 25 μg) served as positive controls. Extract-impregnated discs were placed on the agar plates at designated positions.

Incubation and Observation: The inoculated plates were incubated at 37°C for 24 hours. Zones of inhibition (in mm) around the discs were measured using a Vernier caliper to evaluate antimicrobial activity

Here, efficient extraction of antimicrobial compounds from *Basella alba* using Soxhlet techniques. Testing the extracts' efficacy was performed against *Staphylococcus aureus*, a key skin-associated pathogen, using robust standardized techniques.

3 Results and Discussions

3.1 Antimicrobial Activity

The antimicrobial activity of Malabar spinach (*Basella alba*) extracts was assessed against *Staphylococcus aureus* using the widely accepted disc diffusion method. This method measures zones of inhibition to evaluate the efficacy of antimicrobial agents. The findings underscore the potential of *Basella alba* extracts as natural antimicrobial agents, offering a complementary alternative to standard antibiotics such as Kanamycin and Ampicillin.

3.1.1 Observations from Standard Antibiotics

To benchmark the performance of *Basella alba* extracts, standard antibiotics were included, where Kanamycin (1000 μg), A robust zone of inhibition measuring 3.5 cm was observed, confirming its potent antimicrobial activity. Kanamycin's efficacy serves as a gold standard for comparison, and ampicillin (25 μg), The zone of inhibition was 1.5 cm, reflecting moderate activity against *S. aureus*. This result highlights Ampicillin's reduced potency compared to Kanamycin.

3.1.2 Antimicrobial Activity of Basella alba Extracts

The antimicrobial activity of *Basella alba* extracts was assessed for aqueous and ethanol solvent systems, with results standardized at a concentration of 100 mg/mL.

Ethanol Extracts

The ethanol extract of the stem exhibited a zone of inhibition of 2.0 cm, indicating moderate antimicrobial activity. Like the stem, the ethanol extract of the leaf showed a 2.0 cm zone of inhibition, suggesting a comparable distribution of ethanol-soluble compounds across the plant parts.

Aqueous Extracts

The aqueous extract of the stem resulted in a 2.5 cm zone of inhibition, reflecting greater activity than ethanol extracts, likely due to the solubility of hydrophilic compounds. The aqueous leaf extract displayed the highest activity with a 3.0 cm zone of inhibition, highlighting the dominance of water-soluble bioactive compounds in the leaves. Overall, aqueous extracts demonstrated higher antimicrobial activity than ethanol extracts, with leaf extracts outperforming stem extracts in both solvent systems.

3.1.3 Comparative Analysis

Figure 5.1 and Figure 5.2 summarize the zones of inhibition for all samples, showing that, Kanamycin exhibited the highest activity with a 3.5 cm zone of inhibition, the aqueous leaf extract followed, with a 3.0 cm zone of inhibition, and ethanol and aqueous stem extracts displayed moderate activity, while Ampicillin showed the least activity.

Fig 1.2: Zone of inhibition of microorganism

3.2 Antimicrobial activity of the extracts

3.2.1 Efficacy of Basella alba Extracts

The antimicrobial activity observed in *Basella alba* extracts can be attributed to bioactive compounds such as saponins, flavonoids, alkaloids, and phenolic compounds, consistent with findings from prior studies. These compounds disrupt microbial cell membranes, inhibit protein synthesis, and interfere with enzymatic pathways.

Solvent System Effect: Aqueous extracts outperformed ethanol extracts, suggesting that hydrophilic compounds are key contributors to the antimicrobial properties. Ethanol extracts, while less effective, indicate the presence of lipophilic compounds that contribute to moderate activity.

Plant Part Variation: Higher activity in leaf extracts supports the hypothesis that leaves, as sites of photosynthesis and metabolite accumulation, harbor more bioactive compounds.

3.2.2 Comparison with Standard Antibiotics

The results reveal that Kanamycin's superior activity affirms its efficacy against *S. aureus*. However, rising antibiotic resistance underscores the need for alternative antimicrobial agents. The aqueous extract of *Basella alba* leaves demonstrated greater activity than Ampicillin, suggesting its potential as a natural antimicrobial agent.

3.2.3 Implications for Future Research

This study provides a foundation for further exploration of *Basella alba*'s antimicrobial properties.

Phytochemical Profiling, where advanced techniques like HPLC and mass spectrometry can identify and quantify active compounds. **Mechanism of Action**, where elucidating the molecular mechanisms of antimicrobial activity. **Spectrum Testing**, where evaluating efficacy against a broader range of pathogens, including Gram-negative bacteria and fungi.

Synergistic Studies, where Assessing the potential for synergism between *Basella alba* extracts and standard antibiotics. Formulation Development, where creating topical or hydrogel formulations for treating infections caused by *S. aureus*.

While valuable, the study faced certain limitations, where testing was confined to *S. aureus*, limiting generalizability, dose-response studies were not conducted, and only two solvent systems were explored; additional solvents could yield broader insights

4 Conclusion

In conclusion, this study successfully isolated *Staphylococcus aureus* from pimple pus and evaluated the antimicrobial properties of Malabar Spinach (*Basella alba*) extracts from its stems and leaves. The findings revealed significant antimicrobial activity against *Staphylococcus aureus*, suggesting the plant's potential in treating acne and other skin infections. Future research should focus on optimizing extraction methods to enhance bioactive compound yield and potency. Additionally, expanding the investigation to include other microbial strains will provide a more comprehensive understanding of the plant's antimicrobial spectrum. The study's comparison of Malabar Spinach extracts with conventional antibiotics such as Kanamycin and Ampicillin highlights its natural efficacy. The promising results suggest that Malabar Spinach could serve as a viable alternative to synthetic antibiotics. Future applications, such as pimple patches infused with Malabar Spinach extracts, could offer a more effective, natural treatment for acne. Overall, Malabar Spinach shows significant promise in dermatology and skincare.

5 Acknowledgement

We are thankful to the Department of Biotechnology, P.A. College of Engineering, Mangalore for providing lab facilities to conduct our research work.