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Screening of Antimicrobial Potential of *Annona squamosa* and *Momordica charantia* Against Selected Bacterial Pathogens

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Abstract

The increasing resistance of pathogens to existing antibiotics necessitates the search for new antimicrobial agents. Medicinal plants, known for their bioactive compounds, present a promising alternative. This study aims to evaluate the antimicrobial potential of *Annona squamosa* (custard apple) and *Momordica charantia* (bitter gourd) using aqueous, methanolic, and ethanolic extracts against selected bacterial pathogens. The agar well diffusion and disc diffusion methods were employed to determine zones of inhibition. The results indicate that methanolic and ethanolic extracts of *Annona squamosa* exhibit strong antimicrobial activity, while the ethanolic extract of *Momordica charantia* also shows significant inhibition. These findings suggest potential applications of these plant extracts as natural antimicrobial agents.

Keywords:

Annona squamosa, *Momordica charantia*, Antimicrobial Activity, Agar Well Diffusion, Disc Diffusion, Medicinal Plants

1. Introduction

Nature has been a source of medicinal agents for thousands of years and an impressive number of modern drugs have been isolated from natural sources, many based on their use in traditional medicine (Nair *et al.*, 2004).

Various medicinal plants have been used for years in daily life to treat diseases all over the world. They have been used as a source of medicine. Plants are the basic source of knowledge of modern medicine.

The basic molecular and active structures for synthetic fields are provided by rich natural sources. Higher plants, as sources of medicinal compounds, have continued to play a dominant role in the

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maintenance of human health since ancient times (Farombi, 2003). Over 50% of all modern clinical drugs are of natural product origin (Stuffness, 1982), and natural products play an important role in drug development programs in the pharmaceutical industry (Baker, 1995). There has been a revival of interest in herbal medicines.

Plants with possible antimicrobial activity should be tested against an appropriate microbial model to confirm the activity and to ascertain the parameters associated with it. The effects of plant extracts on bacteria have been studied by a very large number of researchers in different parts of the world (Ates, 2003; Reddy, 2001).

It has been suggested that aqueous and ethanolic extracts from plants used in allopathic medicine are potential sources of antiviral, antitumoral and antimicrobial agents (Chung, 1995; Vlietinck, 1995).

2. Aims and Objectives

To screen and evaluate the antimicrobial activity of *Annona squamosa* and *Momordica charantia* leaf extracts against selected bacterial pathogens using standardized microbiological techniques.

3. Materials and Methods

3.1 Materials

- **Plant Material:**
  - *Annona squamosa* (Leaves)
  - *Momordica charantia* (Leaves)
- **Bacterial Cultures:**
  - [Specify Bacterial Strains Used]
- **Culture Media:**
  - Hi-sensitivity test agar and broth
- **Solvents:**
  - Distilled water, Methanol, Ethanol
- **Miscellaneous:**
  - Petri plates, antibiotic zone reader scale, sterile discs, micropipettes

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3.2 Preparation of Plant Extracts

Leaves were shade-dried, powdered, and extracted using distilled water, methanol, and ethanol. Extracts were filtered and concentrated using rotary evaporation.

3.3 Antimicrobial Assay

a) Agar Well Diffusion Method

A loopful of test organism was inoculated in 5 ml of Hi-sensitivity test broth and incubated at 37°C for 8 hours. A lawn culture was prepared on agar plates. Wells (6 mm diameter) were bored and 0.1 ml of plant extract was added. Plates were kept at 4°C for 1 hour to allow diffusion, then incubated at 37°C for 24 hours. Zones of inhibition were measured in millimeters.

b) Disc Diffusion Method

Sterile filter paper discs were impregnated with plant extracts and placed on inoculated agar plates. Plates were incubated at 37°C for 24 hours, and zones of inhibition were measured using an antibiotic zone reader scale.

4. Results and Discussion

Herbal medicine represent one of the most important field of traditional medicine all over the world. To promote the proper use of herbal medicine and to determine their potential as sources for new drugs, it is essential to study medicinal plant.

During the past decade traditional system of medicine have become increasingly important in view of their safety. A current estimate suggest that in many developing countries a large proportion of population realise heavily on traditional practices and medicinal plant to meet primary health care needs (Tambekar *et al.*, 2009).

The present study was conducted to investigate antimicrobial properties of two selected plants *Annona squamosa* and *Momordica charantia*. Herbal remedies play a fundamental role in traditional medicine in rural areas of India. Here the therapeutic treatment of choice as antiseptic, anti-inflammatory.

In treatment of infectious diseases including diarrhea in present study attempt was made to co-relate traditional herbal medicinal

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Knowledge held by the Indian native people with modern scientific laboratory-based assay (Dabur et al., 2007).

The leaves of two medicinal plant were tested for antimicrobial activity. The plant used were Annona squamosa and Momordica charantia. The six different types of extracts including aqueous (distilled water), solvents – methanol, ethanol, acetone petroleum ether, and chloroform were prepared.

The pathogens selected in the present study were Staphylococcus aureus, Bacillus subtilis, Escherichia coli, Klebsiella pneumoniae, Proteus vulgaris, Salmonella typhi and Pseudomonas aeruginosa.

Thus a total of six different extract of each medicinal plant were tested for antimicrobial activity against these selected bacterial pathogens.

Table 1: Antimicrobial activity of *Annona squamosa* by Agar well diffusion method

[3]

Microorganism	Methanol	Ethanol	Acetone	Petroleum Ether	Distilled Water	Chloroform
Staphylococcus aureus	R	R	R	R	R	16 mm
Bacillus subtilis	R	R	R	R	R	14 mm
Escherichia coli	R	R	R	R	R	18 mm
Klebsiella pneumoniae	R	R	R	R	R	20 mm
Proteus vulgaris	R	R	R	R	R	14 mm
Salmonella typhi	R	R	R	R	R	16 mm
Pseudomonas aeruginosa	R	R	R	R	R	18 mm

Table 2: Antimicrobial activity of *Annona squamosa* by Disc diffusion method

Microorganism	Methanol	Ethanol	Acetone	Petroleum Ether	Distilled Water	Chloroform
Staphylococcus aureus	R	R	R	R	R	10 mm

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Bacillus subtilis	R	R	R	R	R	10 mm
Escherichia coli	R	R	R	R	R	10 mm
Klebsiella pneumoniae	R	R	12 mm	R	R	10 mm
Proteus vulgaris	R	R	R	R	R	10 mm
Salmonella typhi	R	R	R	R	R	R
Pseudomonas aeruginosa	R	R	R	R	R	10 mm

Note:

- "R" = Resistant (no antimicrobial activity observed)
- The values in mm indicate the zone of inhibition (antimicrobial activity)

After performing agar well diffusion method for Annona squamosa of these methanolic & Ethanolic extracts were with antimicrobial potential against all the tested pathogen.

Except **Staphylococcus aureus** (Table 1) these results showed similarity with that of (Jha et al.*(2006);Kokate et al.*(2001)).

Staphylococcus aureus was the only pathogen which showed resistance to all the extract except chloroform extract.

Petroleum ether & Distilled water of **Annona squamosa** does not have any antimicrobial potential because all the tested organisms were resistance to it.

Disc diffusion method for **Annona squamosa** showed that ethanol extract have antimicrobial potential in which **Staphylococcus aureus**, **Escherichia coli** and **Klebsiella pneumoniae** were found to be sensitive to Ethanol extract (Table 2).

Methanol, petroleum ether, and distilled water does not showed any antimicrobial activity against the selected pathogen (Dwivedi et al., 2006).

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Table 3: Antimicrobial activity of Momordica charantia by Agar well diffusion method

Leaves Extract	Methanol	Ethanol	Acetone	Petroleum ether	Distilled water	Chloroform
Staphylococcus aureus	14 mm	14 mm	14 mm	R	20 mm	12 mm
Bacillus subtilis	14 mm	R	11 mm	R	14 mm	R
Escherichia coli	R	R	R	R	R	R
Klebsiella pneumoniae	14 mm	R	14 mm	R	10 mm	R
Proteus vulgaris	10 mm	10 mm	14 mm	R	R	18 mm
Salmonella typhi	R	R	R	R	14 mm	10 mm
Pseudomonas aeruginosa	R	R	R	R	R	R

Table 4: Antimicrobial activity of Momordica charantia by Disc diffusion method

Leaves Extract	Methanol	Ethanol	Acetone	Petroleum ether	Distilled water	Chloroform
Staphylococcus aureus	12 mm	R	R	R	10 mm	10 mm
Bacillus subtilis	10 mm	R	R	R	R	R
Escherichia coli	R	R	R	R	R	14 mm
Klebsiella pneumoniae	R	R	R	R	R	16 mm
Proteus vulgaris	10 mm	R	R	R	10 mm	10 mm
Salmonella typhi	10 mm	R	R	R	14 mm	R
Pseudomonas aeruginosa	R	R	R	R	R	10 mm

Note: Where **R = Resistant**

Out of all the six extract of *Momordica charantia* tested by agar well diffusion method all the organisms were sensitive to ethanol extract except *Escherichia coli* (Table 3).

Staphylococcus aureus was highly sensitive to acetone extract (26 mm). Petroleum ether & Distilled water does not show any antimicrobial activity against the selected pathogen.

By performing Disc diffusion method for *Momordica charantia*, *Staphylococcus aureus*, *Escherichia coli*, *Klebsiella pneumoniae*, *Proteus vulgaris* and *Pseudomonas aeruginosa* were

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sensitive to chloroform extract (Table 4). These result were similar with the finding of (Jagessar *et al.*, 2008).

5. Conclusion

The result of the antimicrobial activity of *Annona squamosa* and *Momordica charantia* showed that Methanol and Ethanol extract of *Annona squamosa* have great potential as antimicrobial activity against selected pathogens. While Ethanol extract of *Momordica charantia* also showed antimicrobial activity against selected pathogens.

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