A Research paper on

STUDIES ON ANTAGONISTIC EFFECT OF ENDOPHYTIC BACTERIA AGAINST HUMAN REISSTANT PATHOGENIC BACTERIA

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DR. RAVI RANJAN KUMAR

Submitted by

MS. HARMI D. BAKRANIYA – 200601003

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Department of Biotechnology, Faculty of Science

Atmiya University - Rajkot

Studies on Antagonistic Effect of Endophytic Bacteria against Human Resistant Pathogenic Bacteria

Harmi D. Bakraniya, Guide: Dr.Ravi Ranjan Kumar

Department of Biotechnology, Atmiya University

ABSTARCT

The study aimed to isolate and screen Endophytic bacteria from various medicinal plants for their potential antimicrobial activity against human pathogenic bacteria. This provides insight into the potential of endophytic bacteria as a source of novel antibiotics to overcome the growing problem of antimicrobial resistance. The plant samples were collected from Atmiya University's medicinal garden and processed them using surface sterilization procedures. The collected endophytic bacteria were then subjected to primary and secondary screenings to determine their antimicrobial activity. 10 bacterial colonies were isolated, and only one colony showed a zone of inhibition (ZOI) against one human pathogenic bacteria, Enterobacteriaceae (M3).

Introduction

Endophytic bacteria are a group of bacteria that reside in plant tissues. They are not harmful to host plant. Natural products which contain antimicrobial, antioxidant, antidiabetic, antiviral, anticancer activity & immunosuppressant can be obtained from endophytic microbes. Endophytes produce antibiotics that directly inhibit growth of pathogen. Therefore, to deal with increasing number of drug-resistant pathogen Endophytic Bacteria could serve as a potential source of novel antibiotics. Bacteria is supposed to die when supplemented with antibiotic. But due to multiple time exposure with the same antibiotic they become resistant to the same.

Antimicrobial resistance (AMR) happens when germs like bacteria and fungi develop the ability to defeat the drugs designed to kill them. Resistant infections can be difficult, and sometimes impossible, to treat. So we have 3 solution

So, Increase dose/concentration of antibiotics, Modification in structure of antibiotics, **Find new antibiotics that inhibit drug resistant bacteria.**

AMR is a growing in public health concern worldwide, it can result in increased mortality and morbidity, long hospital stays, increased healthcare costs and the potential for the spread of infections to others.

Materials and methods

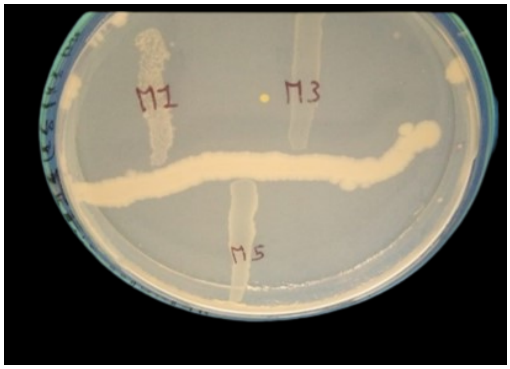
Sample collection: 8 plant samples, including stalk and leaves were collected from medicinal garden of Atmiya University. The plant samples taken are Gugal (*Commiphora wightii*), Jambu (*Syzygium cumini*), Papaya (*Carica papaya*), Rayan (*Manilkara hexandra*), Machi patram (*Artemisia indica*), Thuja (*Thuja occidentalis*), Panfuti (*Kalanchoe pinnata*).

Pathogens were collected from Microbiology Lab. Rajkot. *Salmonella typhi* (M1), Enterobacteriaceae (M3), *Acinetobacter baumannii* (M5).

Isolation of endophytes: 1 sample from each plant was aseptically crushed into 0.85% N-saline, filtered with muslin cloth and incubated 0.1 ml on nutrient medium. These plates were incubated at 37˚C for 1 to 2 days.

Surface sterilization: The collected samples were washed by tap water, and processed by a 3 step surface sterilization procedure. Which included 5 min. washing with liquid detergent such as tween20/tween80 or teepol, wash with tap water, 5 min. wash with 3% NaOCl, followed by washing with D/W. Then further procedure is done under Laminar Airflow. Wash with autoclaved D/W, 1o min. washing with 0.1% $HgCl_2$ & give last wash with autoclaved D/W. To check sterilization take last D/W (0.1 mL) as control plate on nutrient agar, incubated at 37˚C for 1 day and examined for microbial growth.

Primary screening: After incubating all the plates for 1 day, select colonies from each plate and streak on another nutrient agar plate. Incubate for 1 day. After 1 day incubation of selected colonies, cross streak pathogen on each plate and incubate for 1 day. Next day observe zone of incubation.



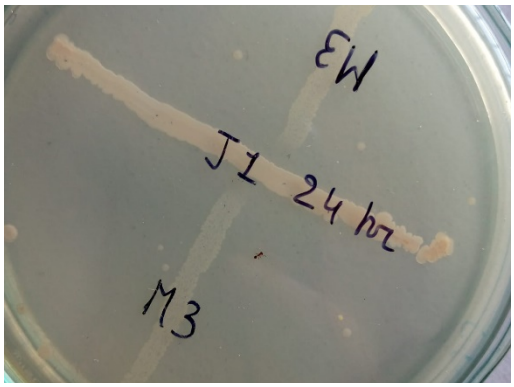


Figure: positive result of jambu plant leaves against M3.

Secondary screening: now for secondary screening, select those culture which showed positive result in primary screening. Now incubate those culture in nutrient broth for 1 to 2 days. Centrifuge 3 mL culture from each flask, Take supernatant and Inoculate 100 µL culture into molten agar plate.

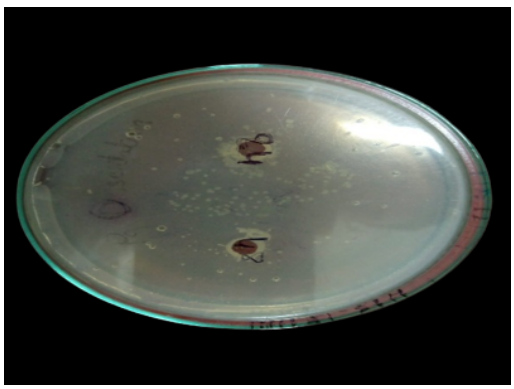
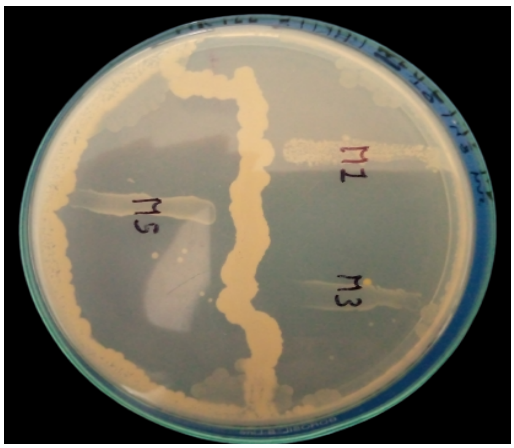


Figure: negative result of secondary screening.

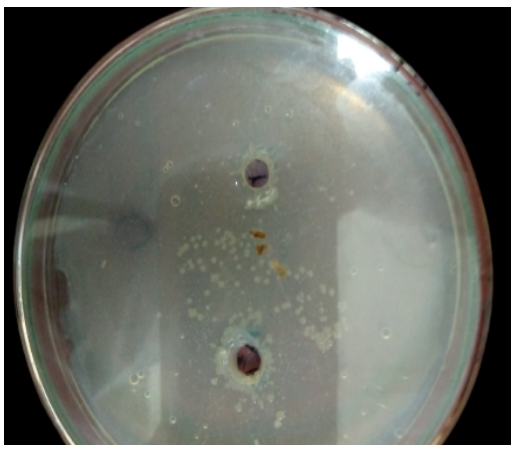
Morphological analysis:

	Size	Shape	Texture	Colour	Elevation	Optical property
Gugal	Large	Round	Smooth	White	Convex	Transparent
Jambu	Large	Irregular	Smooth	White	Raised	Translucent
Papaya	Small	Round	Dry	White	Convex	Opaque
Rayan	Medium	Round	Smooth	White	Convex	Transparent
Machi Patram	Small	Round	Smooth	Orange	Flat	Opaque
Panfuti	Small	Round	Smooth	Yellow	Convex	Transparent
Thuja	Small	Round	Smooth	Orange	Convex	Transparent

Results:



Primary screening: Positive result of jambu plant leaves' colony against M3. Negative result against M1 and M5.



Secondary screening: Negative result observed.

Discussion:

10 different bacterial colonies were isolated and were subjected to primary screening. Among them only 1 bacterial colony showed ZOI against one human pathogenic bacteria Enterobacteriaceae (M3). That selected bacterial colony was subjected to secondary screening. Negative results were found to be observed in secondary screening due to presence to contamination while transferring the culture.

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