

STUDIES ON WOUND HEALING PROPERTY OF COASTAL MEDICINAL PLANTS

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Abstract

In the modern days multiple drug resistance has been developed in many microbes due to random use of existing antimicrobial drugs in the treatment of infectious diseases. This paves the way for reconsidering traditional medicine; hence a study was carried out to explore the antimicrobial activities of the water and acetone extract of *Azadirachta indica* and *Acalypha indica* against *Staphylococcus aureus* and *Pseudomonas aeruginosa*. The results revealed that water extract of both the plants were more effective than Acetone extract particularly on *pseudomonas* sp. Hence both the plants can be vitally used in treating various diseases caused by those pathogens.

Key words:

Azadirachta indica,
Acalypha indica,
Antibacterial activity.

1. INTRODUCTION:

Most of the plants used today were known to the people of ancient cultures throughout the world and were highly considered their preservative and medicinal powers. Scientific experiments on the antimicrobial properties of plants and their components have been documented in the late 19th century [14]. India has a rich flora that is widely distributed throughout the country. Herbal medicines have been the basis of treatment and cure for various diseases and physiological conditions in traditional methods practiced such as Ayurveda, Unani and Siddha.

Medicinal components from plants play an important role in conventional as well as western medicine. 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 [8, 9] Much work has been done on ethno medicinal plants in India [10, 11, 12].

Plant derived drugs have been a part of the evolution of human, healthcare for

thousands of years. Plant based drugs were commonly used in India and China [15]. At the same time, indigenous people of the rest of the planet were also utilizing plants for curing diseases. Today, nearly 88% of the global populations turn to plant derived medicines as their first line of defense for maintaining health and combating diseases. One hundred and nineteen secondary plant metabolites derived from plants are used globally as drugs; 15% of all angiosperms have been investigated chemically and of that 74% of pharmacologically active plant derived components were discovered. Currently, people of Asia and India are utilizing plants as part of their routine health management [16].

Acalypha indica L. (family: *Euphorbiaceae*) is a weed widely distributed throughout the plains of India. It has been reported to be useful in treating Pneumoniae, asthma, rheumatism and several other ailments[17]. The dried leaves of *Acalypha indica* was made into a poultice to treat bedsores and wounds and the juice of *Acalypha indica* is added to oil or lime and used to treat a variety of skin disorders. The leaves of *Acalypha grandis* have also been reported to possess contraceptive activity[18]. Several

chemical[19] and biological[20] investigations have been carried out on this plant. In the present study, an attempt has been made to enrich the knowledge of antibacterial activity of *Acalypha indica* plant extract against Gram positive and Gram-negative bacteria.

Azadirachta is a genus of two species of trees in the flowering plant family Meliaceae (Mahogany family). Numerous species have been described in the genus but only two are currently recognized, *A. excelsa* (Jack) Jacobs, and the economically important Neem tree, *A. indica* A. Juss [21, 22]. Both species are native to the Indomalaysian region, and *A. indica* is also widely cultivated and naturalized outside its native range. The resins from the trees have been attributed with medical benefits.

2. MATERIALS AND METHODS

2.1. COLLECTION OF SAMPLE:

Pus sample were collected from the Government Hospital at Mayiladuthurai from patients of Diabetes, Filarial wound by using sterile swabs. The swabs were immediately immersed into saline and transported to the laboratory.

2.2. COLLECTION OF PLANTS FOR EXTRACTION:

The plants such as *Azadirachta indica* and *Acalypha indica* were collected from Nagapattinam district, Tamilnadu, South – east coast of India (Plate – 1, fig.a&b). The leaves were washed thoroughly with distilled water to remove extraneous matter and dried under shade. The dried leaves were powdered using a pestle and mortar.

2.3. PREPARATION OF PLANT EXTRACTS:

The powder of plant leaves were extracted in acetone and water by following cold

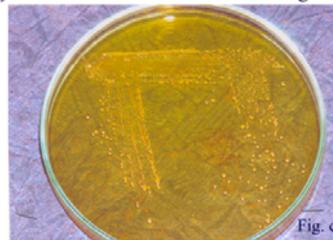


percolation method and evaporated at room temperature of 26°C. This resulted in a brown residue and it was used for antibacterial assay.

2.4. BACTERIAL STRAINS FOR ANTIBACTERIAL ASSAY:

Antibacterial activities of the extracts were determined against two predominant bacterial strains one gram positive and one gram negative strain. (*Staphylococcus aureus* and *Pseudomonas aeruginosa*) This strain was screened from samples of Filarial, Diabetes wound (fig.1).

Staphylococcus aureus in Mannitol Salt agar medium



pseudomonas aeruginosa in agar medium



2.5. DETAIL OF STRAIN:

The strain was tested with following to confirm.

Confirmation test	<i>S. aureus</i>
Gram staining	Positive
Motility	non-motile
Catalase	Positive
Mannitol Salt agar	Positive

Confirmation test	<i>P.aeruginosa</i>
Gram staining	Negative
Motility	motile
Methyl red	positive
Voges-proskauer	positive
Oxidase	positive

2.6. IMPORTANCE OF STRAIN:

Staphylococcus aureus is a Gram positive which cause wound infection, food poisoning, pyogenic infection, pneumonia and toxic shock. *Pseudomonas aeruginosa* is a common bacterium which can cause disease in animals and humans. It is found in soil, water, skin flora and most man-made environments throughout the world.

2.7. BACTERIAL CULTURE:

Nutrient broth medium was prepared by adding the nutrient broth in the distilled water and sterilized in an autoclave at a pressure of 15lb and temperature of 120° C. The bacterial strains were inoculated in the nutrient broth medium and the cultures were maintained at 28°±2°C. The culture was swabbed over the Muller Hinton agar plates.

2.8. ANTIBACTERIAL ASSAY (Kirby Bauer, 1960)

Antibacterial assay was carried out by disc diffusion technique. The plant extracts were tested against *Staphylococcus* and *Pseudomonas* bacterial strain using the Mueller Hinton agar. Medium was

prepared, pH was adjusted and poured 20 ml per plate. After solidification, the culture was swabbed on the plates. Then sterile 5mm diameter paper discs were used for impregnated with acetone, water extract of plant and its initial weight and final weight were measured. The sterile discs impregnated with methanol were used as control. Test discs were the aseptically applied to the surface of the agar medium and the plates were incubated for 24 hrs. After incubation period the zone of inhibition was measured.

3. RESULTS AND DISCUSSION

Among ten samples collected from the Diabetes patients, eight samples had *Staphylococcus aureus*, and six had *pseudomonas aeruginosa*. The organism was isolated by using gram staining, Mannitol salt agar and pseudomonas isolation agar (selective media) medium, identified based on their biochemical properties. Then the bacterial culture was maintained in nutrient agar medium, used for further studies.

Antibacterial assay was carried out by 4 crude corresponding to 2 plants. Plant extracts of two plants showed antibacterial activity against the bacterial strain. Table 1 shows the antibacterial activities of water and acetone extracts of *A. indica* and *Azadirachta indica* against tested organisms. In this present investigation compare to acetone extract, water extract of the plant recorded significant zone of inhibition activities against these two tested bacterial strains. The water extract of *Acalypha indica* showed the maximum zone of inhibition for *Staphylococcus aureus* (9 mm) and *Pseudomonas aeruginosa* (10 mm), and The acetone extract of *Acalypha indica* showed the minimum zone of inhibition

for *Staphylococcus aureus* (1 mm) and *Pseudomonas aeruginosa* (3 mm)

The water extract of *Azadiracta indica* exhibited significant result against *S. aureus* and *Pseudomonas aeruginosa* ranging from 7 and 9 mm respectively.

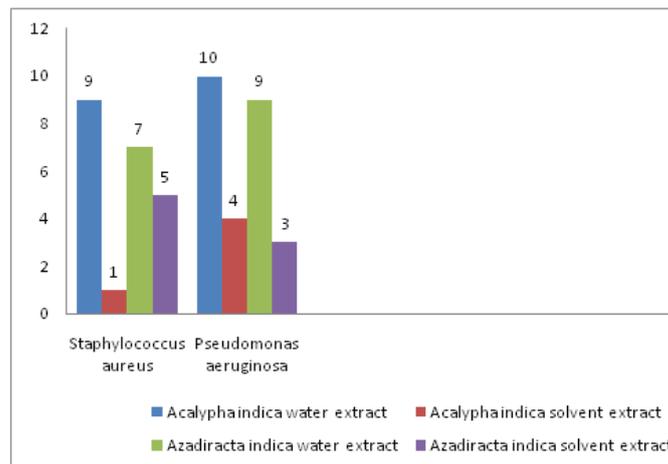
And the acetone extract of *Azadiracta indica* exhibited significant result against *S. aureus* and *P. aeruginosa* ranging from 5 and 4 mm respectively. Both dilution screening for antibacterial activity showed a promising effect. When compare to

Table -1 Inhibitory Effect of Medicinal plants against gram-positive and negative bacteria

Bacterial strain	<i>Acalypha indica</i>		<i>Azadiracta indica</i>	
	A	B	A	B
	Diameter in mm	Diameter in mm	Diameter in mm	Diameter in mm
<i>Staphylococcus aureus</i>	9	1	7	5
<i>Pseudomonas aeruginosa</i>	10	3	9	4

A: Water extract; B: Acetone extract

FIGURE – 1



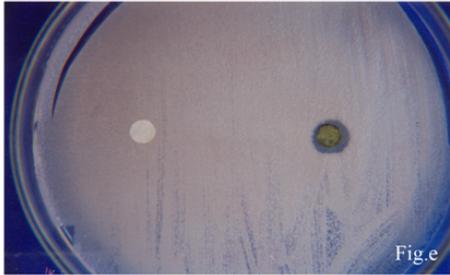
Azadiracta indica, *Acalypha indica* is very effective against *S. aureus* and *P. aeruginosa*. Whereas, *A. indica* was found to be active against *S. aureus* and *P. aeruginosa*.

In 1995 Meckas et al .,[1] tested 135 plant species of which 6 showed highest activity against Gram positive bacteria *Staphylococcus aureus* and *Sansevieria roxburghiana* exhibited moderate activity against the same bacterial strain. Garlic contain antimicrobial component Allicin. It can able to inhibit the staphylococcus

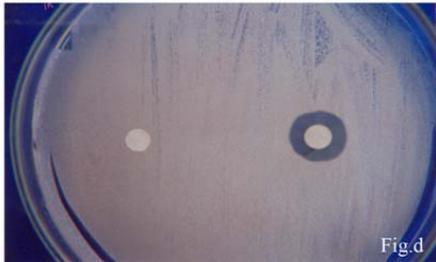
aureus. [2] Fifty to hundred mg of freshly crushed onion in 1ml of nutrient solution completely inhibits the growth of *Staphylococcus aureus*. [22]

Dilution method was employed to determine the effect of petroleum ether extract (40-60 Degree) chloroform and methanolic extract of dried leaves of *Acalypha indica* Linn (Euphorbiaceae) against fungi (*Candida albicans*) and bacteria (*Escherichia coli*, *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Salmonella typhosa*, *Bacillus substilis*,

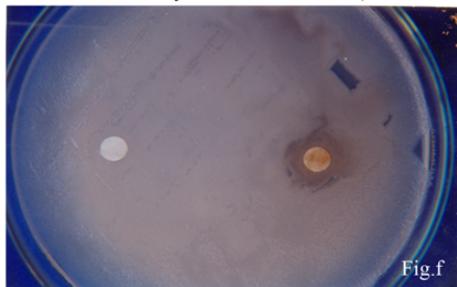
Zone of inhibition by *Azadirachta indica* (Acetone extract)



Zone of inhibition by *Acalypha indica* (water extract)



Zone of inhibition by *Azadirachta indica* (water extract)



Klebsiella pneumoniae). Except the petroleum ether extract, all the extract exhibited a prominent antimicrobial activity. The methanolic extract was further fractionated into acetone soluble and insoluble parts. Both the parts exhibited prominent antimicrobial activity. The acetone insoluble part exhibited MIC of .0040 mg/ml against *Staphylococcus aureus* and both acetone soluble and insoluble parts exhibited MIC of .05 mg/ml against *Salmonella typhosa*. [3]

The ethanolic extracts of *Heliotropium indicum*, *Plumbago zeylanicum* and *Acalypha indica* were evaluated for their wound healing activity in rats. Wound healing activity was studied using excision

and incision wound models in rats following topical application. Animals were divided into four groups with six in each group. Ten percent w/v extract of each plant was prepared in saline for topical application. *H. indicum* possesses better wound healing activity than *P. zeylanicum* and *Acalypha indica*. Tensile strength results indicate better activity of *H. indicum* on remodeling phase of wound healing [4].

Even though much work has been done on ethnomedicinal plants in India, interest in a large number of traditional natural products has increased of late. Several medicinal plants have been reported to possess antimicrobial, antifungal and other activity has been elucidated by various workers [5]. Phytochemical extracts from Neem plant are potential sources of antiviral, antitumor and antimicrobial agents [6]. Several workers have evaluated antibacterial, antisecretory, antihemorrhagic, insecticidal activity of *A. indica* based drugs to meet the health care needs [7].

In the present work *Staphylococcus aureus* shows minimum inhibition in all plant extracts. In the present study, higher activity was found in water extracts *Acalypha indica* showed high activity against the same strain.

4. CONCLUSION

Healing power of the plant is an ancient idea and the traditional medicine is an integral part of rural health care. In many areas, local plants are effectively used to treat ailment. In this study *Acalypha indica* and *Azadirachta indica* were chosen because they are easily available, economical and have high medicinal values. The study revealed that the water extract of *Acalypha indica* and *Azadirachta indica* were more effective

against the pathogens when compared with the acetone extract.

The screenings of antibacterial activity from coastal populations of medicinal plants has been proved highly effective to control the wound infective bacteria. There is a possibility for extracting bioactive substances from the effective medicinal plants. This deserves more research. It can be hoped that the coastal medicinal plants will provide that the most valuable drugs against wound infection.

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