



## ANTIBACTERIAL ACTIVITY OF *AZADIRACHTAINDICA* FRUIT AND LEAF EXTRACT AGAINST *STAPHYLOCOCCUAUREUS* BACTERIA

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### ABSTRACT

*Azadirachta indica* fruit pulp and leaf extract were used to test antimicrobial activity against disease causing bacteria *Staphylococcus aureus*. Extracts of varying concentrations of *Azadirachta indica* fruit pulp and leaf extract were prepared using Phosphate Buffer and tested against test organisms using agar diffusion method. Oxfloracin of same varying concentrations were used to compare the effect of antimicrobial activity of fruit pulp and leaf extract.

Keywords: *Azadirachta indica*, Antimicrobial activity, *Staphylococcus aureus*.

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### I. INTRODUCTION

*Azadirachta indica*, also known as Neem, is a tree in the mahogany family Meliaceae. It is one of two species in the genus *Azadirachta*, and is native to India and the Indian subcontinent including Nepal, Pakistan, Bangladesh and Sri Lanka. It is typically grown in tropical and semi-tropical regions. Its fruits and seeds are the source of neem oil. Neem is a fast-growing tree that can reach a height of 15–20 metres (49–66 ft), and rarely 35–40 metres (115–131 ft). It is evergreen, but in severe drought it may shed most or nearly all of its leaves. The branches are wide and spreading. The fairly dense crown is roundish and may reach a diameter of 15–20 metres (49–66 ft) in old, free-standing specimens. The fruit

of *Azadirachta indica* has a smooth glabrous rind with single embedded seed. Each fruit measures up to 1.4 – 2.8 centimetres. It has a very thin exocarp and fibrous mesocarp which appears yellowish white in color. The density of the mesocarp measures to 0.3-0.5 cm thick. The fruit possesses a very unique bitter- sweet taste that is most fondly consumed by birds. Neem oil which is commonly used as a nature bactericide is made extracted from this fruit.

The dried neem leaves are placed in cupboards and other grain storage areas to prevent infestation of insects. When those dried leaves are burnt the produced smoke helps to keep away mosquitoes. It is used as a traditional medicine for

curing diseases in almost all parts of the Asian countries. The biological activities are attributed to the presence of many bioactive compounds in its different parts. Aqueous extract of Neem leaf extract has a good therapeutic potential as an antihyperglycaemic agent in insulin-dependent and non-insulin-dependent diabetes mellitus[1]. It is used in traditional medicine as a source of many therapeutic agents. *A. indica* (leaf, bark and seeds) are known to contain antibacterial and antifungal activities against different pathogenic microorganisms; in addition to antiviral activity against vaccinia, chikungunya, measles, and Coxsackie B viruses[2]. It is regarded as an ayurvedic herb and so used in baths. .Neem leaf is used for leprosy, eye disorders, bloody nose, intestinal worms, stomach upset, loss of appetite, skin ulcers, diseases of the heart and blood vessels (cardiovascular disease), fever, diabetes, gum disease (gingivitis), and liver problems.

The purpose of present study of is to investigate the antimicrobial activity of fruit pulp and leaf extract of of *Azadirachtaindica* against disease causing bacteria; *Staphylococcuareus* .*Staphylococcus aureus* is a gram-positive coccal bacterium that is a member of the Firmicutes, it is a common cause of skin infections such as abscesses, respiratory infections such as sinusitis, and food poisoning.

*S. aureus* appears as grape-like clusters when viewed through a microscope, and has large, round, golden-yellow colonies, often with hemolysis, when grown on blood agar plates[3].

## II. MATERIAL AND METHOD

*Plant materials*-*Azadirachtaindica*(neem) fruit and leaves were collected from KailashVihar, City Center, Gwalior ,India .

*Fruit Pulp and Leaf extract*-The fruit pulp and leaves were washed thoroughly with methanol. Thereafter , crushed and grinded with 5ml of Phosphate Buffer[4] of pH=7.4 and then centrifuged at 10,000RPM for 5minutes under aspectic conditions.

*Test microorganisms*-The disease causing strains of *S.aureus* were used.

*Generic Drug* –Amoxicillingeneric antibiotic of same varying concentration were used for the test.

*Agar diffusion method*-The method is suitable for organisms that grows rapidly overnight at 35-37o C The well is made in medium after inoculation with microorganisms. When well is loaded with antibiotics, it diffuses in the medium and inhibits the growth of organism. There is logarithmic reduction in antibiotic concentration. The zone of inhibition of bacterial growth around each well is measured and the susceptibility is determined. Medium-Muller Hinton Agar (3.8gm/100ml of distilled water) was prepared, autoclaved at 121° C for 15minutes at 15lbs and poured in sterile petri plates up to a uniform thickness of approximately 5-6mm and the agar was allowed to set at ambient temperature and used. Inoculums-The microorganisms were inoculated in Nutrient broth and incubated at 37° C and were used as inoculums. 25 µl of inoculum was spread over the MHA medium, using sterile spreader.

After few minute, four wells were made in each Petri plate and loaded with 4%, 8% .12% ,16% and 20% concentration of fruit pulp and then leaf extract which were made earlier by Phosphate Buffer respectively. Similarly 4 %,8% .12% ,16% and 20% concentration Amoxicillin solution was added in another plate. Plates were incubated at 37°C for 24hrs. Antimicrobial activity was evaluated by measuring zone of inhibition by using Hi-media zone scale.

## III. RESULT AND DISCUSSION

The fruit pulp and leaf extract of *A.indica* against *S.aureus* bacteria showed variated zone of inhibition.

Table.1. Antibacterial activity of *A.indica* fruit against *S.aureus*

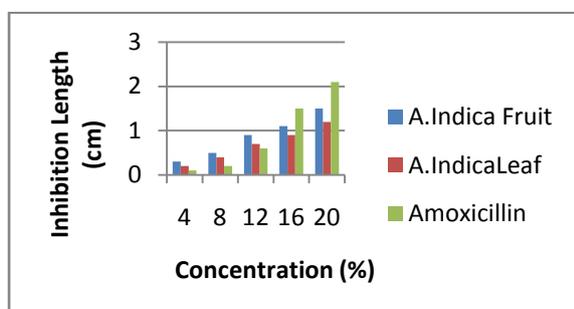
Concentration %	Well Diameter (cm)	Zone of Inhibition (cm)	Inhibition Length (cm)
4	0.8	1.1	0.3
8	0.8	1.3	0.5
12	0.8	1.7	0.9
16	0.8	1.9	1.1
20	0.8	2.3	1.5

Table.2. Antibacterial activity of *A.indica* leaf against *S.aureus*

Concentration %	Well Diameter (cm)	Zone of Inhibition (cm)	Inhibition Length (cm)
4	0.8	1.0	0.2
8	0.8	1.2	0.4
12	0.8	1.5	0.7
16	0.8	1.7	0.9
20	0.8	2.0	1.2

Table.3. Antibacterial activity of Amoxicillin against *S.aureus*

Concentration %	Well Diameter (cm)	Zone of Inhibition (cm)	Inhibition Length (cm)
4	0.8	0.9	0.1
8	0.8	1.0	0.2
12	0.8	1.4	0.6
16	0.8	2.3	1.5
20	0.8	3.2	2.4



Graph 1: Comparison of Antibacterial activity of *A.indica* fruit, *A.indicaleaf* and *Amoxicillin* against *S.aureus*

#### IV. CONCLUSION

*Azadirachta indica* (Neem) is a multipurpose tree with multiple health benefits. Different parts of the tree like fruit pulp and leaves were shown to exhibit antimicrobial effects against *S.aureus*. Screening of this medicinal plant for bioactive compounds may lead to development of less expensive new antimicrobial agents with improved safety and efficacy along with comparatively no side effects.

In this study, we have shown that extracts of *A. indica* (Neem) fruit pulp and leaf to exhibit high antibacterial activity against *S.aureus* at all concentrations used.

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#### REFERENCES

- [1]. Sonia Bajaj, Srinivasan B.P. Investigation into the anti-diabetic activity of *Azadirachta indica*. Indian journal of Pharmacology, 1999;31:138-141
- [2]. Biswas K, Ishita C, Ranajit K B, Uday B. Biological activities and medicinal properties of Neem (*Azadirachta indica*). Current Science, 2002;82:1336-1345.
- [3]. Ryan, KJ; Ray, CG, eds. (2004). Sherris Medical Microbiology (4th ed.). McGraw Hill. ISBN 0-8385-8529-9.
- [4]. K.R. Aneja (2003) Experiments in Microbiology Plant Pathology and Biotechnology (4<sup>th</sup> Edition) ISBN 81-224-1494-X