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"Assessment of Antimicrobial Potency of A.indica Against Target Bacteria and Formulation of Neem Based Floor Cleaner as a Disinfectant"

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Abstract

The current study was carried out to check the efficacy of A.indica against microflora. Total 52 samples from floor, wash besins of college department and sewage samples were also collected during the study and further processed for isolation and identification. The predominant organisms were *S.aureus, E.coli, P.aeruginosa, S.typhi.* The activity of A.indica Neem was checked in different organic solvents such as acetone, ethanol, aqueous similarly during the study neem based floor cleaner was also formulated. The physical parameter as well as antimicrobial activity of formulated floor cleaner was also checked. Excellent result in terms of zone of inhibition was observed, indicating neem plant have strong antibacterial activity in controlling microflora.

Keywords- A.indica, *S.aureus, S.typhi, P.aeruginosa, E.coli*, floor cleaner.

INTRODUCTION

Plants have been a source of herbal remedies through-out the history of mankind. Various medicinal plants have been used for years in daily life to treat diseases all over the world (Nimri et al., 1999; Saxena, 1997). Azadirachta indica A. Juss (syn. Meliaazadirachta) is well known in India and its neighboring countries. It is popularly known as Indian neem (margosa tree) or Indian lilac. It is an evergreen tree, cultivated in various parts of the Indian subcontinent. Every part of the tree has been used as traditional medicine for household remedy against various human ailments. Azadirachta Indica (A. Indica) belongs to the family Meliaceae, commonly known as neem. It is used in traditional medicine as a source of many therapeutic agents. A. Neem is fast-growing tree, generally 15–20 m tall (sometimes up to 40 m tall), with a crown diameter up to 20 m. Neem is evergreen but can shed most of its leaves under dry conditions.

MATERIALS AND METHODS

Selection of plant

The plant neem (Azadirachta indica) was selected for study. Its leaves were collected from local area.

Leaf extract

The completely shade dried material was coarsely powdered. The powder is then aseptically kept in air tight container at the moisture free place.

Ethanol, Acetone, Aqueous extract.

5gm of dried leaf powder were taken in a separate container.

50ml of ethanol in one container and 50 ml of acetone in another container was added and kept for 24 hours with periodic shaking then filter the suspension using Whatmann filter paper no1.

The filtrate was then evaporated on a water bath. After evaporation add 2ml of DMSO solution (dimethyl sulphoxide) and the filtrate was collected in air tight container.

Phytochemical analysis (As described as odebiyiet al.,1978)

The coarse powder was soaked in 10 ml of distilled water after 1hourhe suspension were filtered using Whatmann filter paper no 1 then filtered was used for the phytochemical analysis**.**

Test for carbohydrates

1ml of Molisch's reagent was added to 2 ml of neem extract after which a few drops of concentrated sulphuric acid was added. A purple colouration depicts the presence of carbohydrates.

Test for Tannins

2 ml of 5% ferric chloride was added 1 ml of neem extract. A greenish black colouration depict that tannins are present.

Test for Saponins

2 ml distilled water was added 2ml neem extract and shaken for 15 minutes. Foam formation indicates that saponins are present.

Test for Flavonoids

5ml of dilute NH3 solution was added into 1ml of neem extract prior to the addition of concentrate sulphuric acid. A yellow colouration depicts that flavonoid are present**.**

Test for Terpenoids

2 ml chloroform and concentrated H2SO4 was added to 0.5 ml of neem extract. A red brown ring formation at the interface depict that terpenoids are present.

Test for Phenol

2 ml distilled water and a few drops of 10% ferric chloride was added to 1ml of neem extract. A green colouration depicts that phenol are present.

For Acids

Sodium bicarbonate solution was added to 1ml on neem extract. Formation of effervescence depict the presence of acids.

Test for glycosidase

3 ml of chloroform and 10% NH3 solution was added to 2 ml neem extract. A pink colouration depicts that glycosidase are present

Test for Alkaloids

2 ml concentration HCL was added 2ml neem extract before a few drops of Mayer's reagent were added. A greenish colouration depict that alkaloids are present.

Collection of samples

Sewage sample was collected from slum area nearby to Shri Shivaji College of Arts Commerce and Science, Akola. Few samples were collected from the wash basins of labs and floor of different washrooms, as well as infected urine samples from different laboratory.

Isolation and identification from different sources

The collected samples inoculated on nutrient agar by using swabbing technique and then incubate for 24hours in incubator at 37℃. Identification of the isolates was done on the basis of cultural, morphological and biochemical characters on the basis of Bergey's manual of determinant bacteriology**.**

Antimicrobial activity

The organisms were inoculated in Nutrient Broth and incubated at 37℃ for 3-4 hours and these was used for inoculum. Antimicrobial activity was checked by agar well diffusion method on Muller Hinton Agar. A sterile cotton swab was inserted into the bacterial suspension and then rotated and compressed against the wall of the test tube so as to express the excess fluid. Evenly spread the swab so the uniform layer is formed on the agar.Now with the help of cork borer four well of 6mm are bored and the wells are filled with the plant extract i.e (Ethanol, Acetone and Water).And the fourth well is poured with distilled water as a control. By using micropipette the plants extracts are put in wells. Then the petri plates were incubated in incubator for 24 hours at 37℃. Zone of inhibition is form. Zone of inhibition is measured in mm.

Formulation of floor cleaner

The floor cleaner was prepared by using different component such Sodium Bicarbonate, Sodium Hydroxide, EDTA etc. Add all these ingredients in a beaker one by one in beaker and place it at room temperature. Lastly the development of colour was checked. Prepared floor cleaner was spread on lab platforms to check its cleaning activity. Similarly, floor cleaner was also inoculate on nutrient agar to check the presence of any contaminant in it. Same Procedure was repeated 4 to 5 times. Visual examination of floor

cleaner was done by checking colour odour and consistency. Antimicrobial activity of floor cleaner was checked against isolates obtained.

Results and discussion.

As per the observation the antimicrobial activity of different extract of neem leaf showed more zone of inhibition against *S.aureus*, *E.coli* and while *S.typhi* and *P.aureginosa* are less susceptible to neem extract.(table1; fig1)

Table 1 : Antimicrobial Activity of Different Extract of Neem (in mm)

Sr no	Name of the organism	Ethanol	Acetone	Aqueous
1	*S.aureus*	20.0	21.2	20.1
2	*P.*aureginosa	17.6	19.4	17.5
3	*E.coli*	18.2	17.2	12.5
4	*S.typhi*	18.0	20.0	15.5

Table 2: Formulation of Floor Cleaner in Laboratory

Sr no	Ingredients	Weight %
1	Water	100ml
	EDTA	0.5
3	Sodium bicarbonate	3
4	Sodium hydroxy	1
5	Acid slurry	6
6	Perfume	0.5
7	Neem oil	0.3
8	Sodium dodecyl sulphate	0.3
9	Colour	2
10	Benzalkonium chloride	0.2
11	SLES	10
12	Sodium hypochlorite	0.5
13	pH strip	1-2

14	Neem extract	2
15	Salt /NaCl	1-2

Table 3: Antimicrobial activity of prepared floor cleaner. (in mm)

Name of organisms	**Distill water**	**Floor cleaner1**	**Floor cleaner2**
S aureus	R	19.1	20.2
P aeruginosa	R	17.2	19.3
E coli	R	18.0	22.1
S typhi	R	24.0	15.1

In this study leaf extract of A. Indica, exhibits antimicrobial effects against the target pathogens, *Staphylococcus aureus, Pseudomonas aeruginosa, and E.coli ,S.typhi*. It is a simple and inexpensive method for the better sanitization with no toxic effects.

Conclusion

The current investigation concluded that:

The aqueous extract of neem plant leaves in acetone extract have shown strong antimicrobial properties and has higher inhibition rate. The prepared floor cleaner shows strong antibacterial activity in controlling the lab floor and platform microflora. Further studies are needed including toxicity evaluation and purification of active antibacterial constituents from A.indica extracts looking towards a pharmaceutical use. The neem plant contains different phytochemicals and had beneficial effect in controlling the pathogenic microflora and thus can be used in therapeutic formulation in near future.

References

Ali,A. (1993). Textbook of Pharmacognosy, Publication and Information Directorate, New Delhi, India.

Almas K (1999). The antimicrobial effects of extracts Azadirachta indica (Neem) and Salvadora persica (Arak) chewing sticks. Indian J Dent Res 10:23-26.

Bandyopadhyay U, Biswas K, Chatterjee R, Bandyopadhyay D, Chattopadhyay I, Ganguly CK, Chakraborty T, Bhattacharya K, Banerjee RK (2002). Gastroprotective effect of neem (Azadirachta indica) bark extract: possible involvement of H+-K+-ATPase inhibition and scavenging of hydroxyl radical. Life Sci 71:2845-2865

Baswa M, Rath CC, Dash SK, Mishra RK (2001). Antibacterial activity of Karanj (Pngamia pinnata) and neem (Azadirachta indica) seed oil: a preliminary report. Microbios 105:183-189

Bhomick BN, Choudhary BK (1982). Antifungal activity of leaf extracts of medicine plants on Alternaria alternata. Indian Botanical Reporter 1:164-165

Chopra RN, Nayar SL, Chopra IC (1956). Glossary of Indian Medicinal plants. Council of Scientific and Industrial Research, New Delhi

Fujiwara T, Takeka TO, Gihara Y, Shimizu M, Nomura T, Tomuta Y (1982). Studies on the structure of Polysaccharidesfrom the bark of Medica Azardichta. Chemical Pharmaceu Bull 30:4025-4030.