Research Paper Botany

In Vitro Antimicrobial Activities on Different Plant Extracts

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ABSTRACT

Fungal and Bacterial infections are among the ailments treated by traditional healers. The World Health Organization has expressed high interest in traditional medicine, and it is important to demonstrate scientifically that remedies employed in folk medicine are indeed therapeutically active. The aim of the study was to scientifically test if plants used in traditional medicine for the treatment of infections showed indeed antifungal and antibacterial activity. The plants Enicostemma littorale, Citrus lemon, (Seeds), Aristolochia clamatitis (Leaves and seeds), Phyllanthus erecta. Simple laboratory conditions can be applied to validate the antifungal and antibacterial properties of plants used in traditional medicine. Different extracts of the selected plant parts was evaluated against four different fugal and bacterial cultures which are plant and human pathogenic.

KEYWORDS Enicostema, Aristolochia, Antifungal and Antibacterial.

INTRODUCTION : - In India many plants are mostly used as traditional medicines for the treatments of various diseases.Medicinal plants have been an integral parts of the life in various regional communities for food and drugs both .Everyone is concerned with high & growing no. of diseases associated with many microorganisms especially bacteria and Fungi. Bacteria also have become far more resist to many antibacterial agents.The current research focuses on the extraction and assay of the antimicrobial components from the different plant leaves and seeds .Like Enicostemma littorale, Citrus limon ,(Seeds), Aristolochia clamatitis (Leaves and seeds), Phyllanthus erecta. These are easily available at zero cost and found everywhere.Now a day, the use of phytochemicals for pharmaceutical purpose has gradually increased in many countries. According to "World Health Organization" (WHO). (Selva Mohan T. et. al.)Plant produces a wide variety of secondary metabolites which are used either directly as precursors or as lead compounds in the pharmaceutical industry.

1) Enicostemma littorale- E. littorale bloom called chota chirayita in (Marathi), has been used traditionally for many diseases. According to Ayurvedic literature survey. The fresh juice of leaves has been use stomachic and laxative, blood purifier in dropsy, rheumatism, abdominal ulcers hernia, swelling, itches and insect poisoning. The leaves paste is applied on boils. The leaves are feed to cattle to increase appetite.Plant extract were reported for the biological activities such as anti-diabetic, anti-inflammatary and useful in skin diseases. It also act as ethnomedicine for snakebite. The plant is used to cure leucorrhoea

2) Phyllanthus erecta - In India, many plants are mostly used to as traditional medicines for the treatment various infective diseases. The active ingredients present in those plants are highly used for curing the diseases.Phyllanthus erecta is an herbaceous plant. Its fruits are so tiny, found below the branches. Belonging to family Euphorbiaceae. It is found all over the tropical regions. It is widely used for the treatment of Jaundice, Syphilis against constipation gonorrhea and Kidney disorders.

3) Citrus lemon - Everyone is concerned with the high and growing number of diseases associated with microorganism especially Bacteria and Fungi. Bacteria also have become for more resistant to many anti bacterial agents. Fruit also have been studied researchers for the presence bioactive compounds choose related with herbs. The current research also focuses on the extraction and assay of the antimicrobial components from the peels of citrus fruit which are easily available at zero cost thus decreasing the cost for production. The following fruits have been targeted in the present study. The Citrus lemon is used for culinary and non culinary purposes throughout the world primarily for its juice; through the pulp &rind is also used, mainly in cooking &baking.

4) Aristolochia clamatitis (seed & leaves) - Plants based antimicrobial represent a vest untapped source for medicines,& further exploration of plant antimicrobial needs to occurs. Aristolochia clamatitis commonly known as Iswari, nakuli & Gandhanakuli have enormous therapeutic, potential & it was found to be effective in the treatment of intermittent fever ,malaria, parasitic in festations, various skin disease , as an aphrodisiae an anthelmintic & it is also used oeclema, intestinal disorders fungal and bacterial infection

MATERIALS & METHODS

COLLECTION OF PLANTS

The aerial plants parts of Enicostemma littorale, Phyllanthus erecta, Aristolochia, Citrus Lemon were found to be growing in the campus of the Institute of Science, Aurangabad in Marathwada regions in the month of August-September 2014. The plant species were identified and confirmed by using the standard literatures. The collected plant samples were deposited in the form of herbarium in the Dept. of Botany, Govt. Institute of science Aurangabad. The bacterial and fungal strains for the study were isolated in the Dept. of Botany, Govt. Institute of Science, Aurangabad from different infected plant parts .

BACTERIAL/ FUNGAL STRAINS & CULTURE PREPARATION

The bacterial strain used in this study were, Pseudomonas aerogeonasa, Basillus stabtilis, E.coli, Bacillus thorogenesis,

the fungal strain used in this study were Fusarium oxisporium, Fusarium monilifomii, Helmanthosporium sativum, bacterial stains were maintained on nutrient Agar plates. They were sub cultured weekly & subsequently stored at 4˚C .The strains were inoculated in the nutrient broth (PH .7.0) the Agar plates, they were sub cultured weekly & subsequently stored at 4˚C and all fungal species were confirmed in the Dept. of Botany, Govt. Institute science Aurangabad.

PREPRATION OF PLANT CHEMICAL EXTRACTS:-
The methanolic, chloroform & aqueous extracts of all 5 medicinal plants were prepared by. Following method.

PREPARATION OF (CHEMICALS)
1) 80% Chloroform
2) 80% Methanol
3) 100 ml D/W (Aqueous extract) –

PREPARATION:-
10 gm of fresh healthy leaves were collected, washed with D/W and filter with whatmann filter paper No.-1.

Table 1) :- Preparation of dilution – For each plant extract.

Sr.No.	Plant Extract	D/W	Extract Conc.	Control
1.	2 ml	2 ml	2:2	80% Ethanol
2.	4 ml	2 ml	2:4	D/W and 80%
3.	8 ml	2 ml	2:8	Chloroform
4.	16 ml	2 ml	2:16	

Preparation of sterile disc:-
The filter paper was punched into 5 mm disc and sterile in the oven at 1210C for 30 min.

Assay Antibacterial activity wing disc diffusion methods.
In the1 litre. Sterile Nutrient Agar we added bacterial culture which are E.coli, Pseudomonas aroginosa, B.T., Basillus sabtilis in different- different flask then nutrient Agar was poured into sterile petri plates after solidification then mark on plates as 2:2, 2:4, 2:8, 2:16, concentration, then in sterile condition at laminar between two burners with the help of wire loop take a disc. Which have different concentration of plants extract and put it into labeled petri dish and remove wire loop safely for all bacteria using this same procedure for all concentration and all chemicals and make one plate for control.

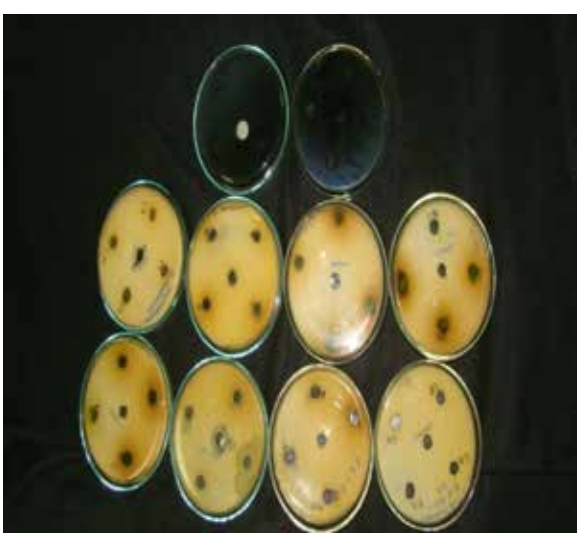
Assay of anti-fungal activity using well diffusion methods:-
PDA medium was poured into the sterile petriplates aseptically and that were also to solidify then spread the fungal culture on that plates Fussarium oxisporium, Fussarium moniliformi, Helmenthosporium and incubated it for three days after the completed growth of fungal mycelium in sterile condition at the laminar make a well with the help of cork borer make on the plate 2:2,2:4,2:8,2:16.Then with the sterile pipette add aqueous solution as a control and aqueous different plant extract dilution same procedure followed for other to chemical methanol and chloroform.

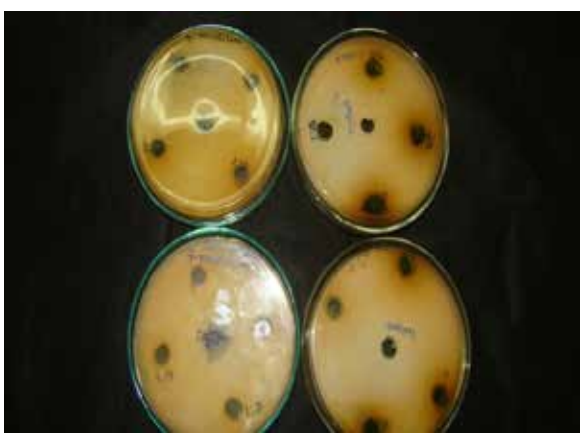
Table 2- Antimicrobial activity of Alcoholic and Aqueous extract of leaves

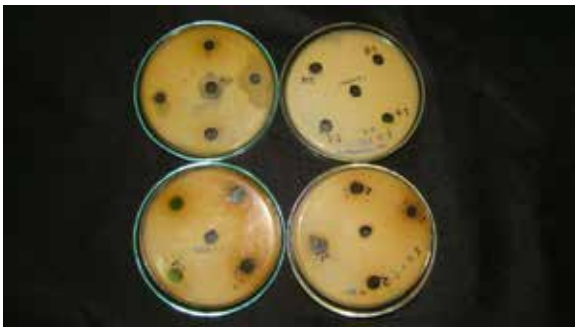
Sr.No.	Test Microorganism	Zone of inhibition (mm)			
		Conc. of leaf extract (Alcoholic and Aqeuos)			
		1:1	1:2	1:4	1:8
1	E.coli.	---	---	---	---
2	B.subtilis	--	--	--	--
3	B. thourangenesis		++	++	++
4	P.auroginosa	--	--	--	--

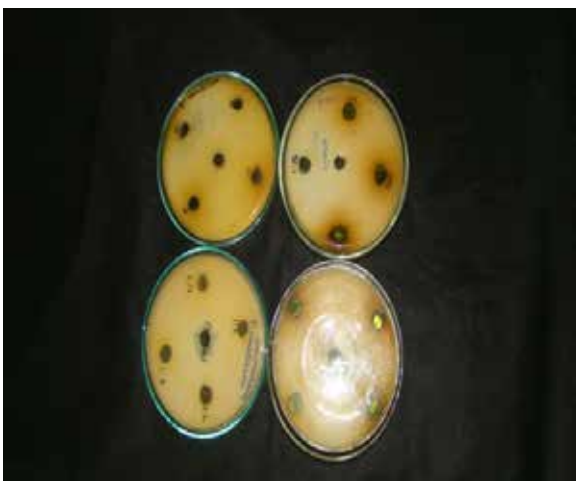
Table 3 - Antifungal activity of Alcoholic and Aqueous extract of leaves

Sr.No.	Test Microorganism	Zone of inhibition (mm)				Control
		Conc. of leaf extract (Alcoholic and Aqueous)				(Alcoholic and Aqueous) (80%)
		2:2	2:4	2:8	2:16	
1	F. monoliformi	+++	+++	+++	+++	++++++
2	F. oxysporum	+++	+++	+++	+++	++++++
3	H. sativum	---	---	----	---	+++++









Photoplate-1. In vitro antimicrobial activity by Well Diffusion Method.

CONCLUSSION:

Among the selected plant leaves extracts only Arostilocia leaf extracts at 10% and 60% concentration shows positive antifungal activity against the Fusarium oxysporum and F. moniliformi. While Aristolocia leaf extracts at 20% concentration shows positive activity against Bacillus thorengensis. The rest of the plant extracts in different solvents and concentrations are found to be negative against the test organisams.

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