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Research article

STUDY OF ANTIFUNGAL POTENTIAL OF *AEGLE MARMELLOS*: A MEDICINAL PLANT

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ABSTRACT: In the present research work methanolic, ethanolic and aqueous extract of leaf, fruit and stem of *Aegle marmelos* were screened for its potential against four fungal strains : *Candida albicans*, *Penicillium chrysogenum*, *A. niger* and *Fusarium solani* using agar well diffusion assay. The length of inhibition zone was measured in millimeters. The results were referenced against Glucanazole antifungal agent. Methanolic fruit extract showed strong antifungal activity against most of the strains where as moderate antifungal potential was shown by leaf extract in aqueous solution.

Keywords: Antifungal potential, Glucanazole, Plant extract, Zone of inhibition

INTRODUCTION

Medicinal plants represent rich source of antimicrobial agents [9]. Medicinal plants have curative properties due to presence of various complex chemical substances found as plant secondary metabolites in one or more parts of them. Plant extracts have been developed and proposed for use as antimicrobial substances [3]. Interest in large number of traditional natural products has increased [17]. Chemical principles from natural resources have contributed significantly for development of new drugs from medicinal plants [2].

Aegle marmelos : family Rutaceae, is one of the most important medicinal tree of India, Burma and Ceylon [16]. It prefers dry, sunny and warm parts of the hill slopes with well drained loamy soil [5]. Leaves, fruits, stem and roots of this tree at all stages of maturity are used as ethnomedicines against various human ailments. Bael fruits are used in gastric troubles, as brain and heart tonic and in gonorrhoea [14,12]. Leaves are also widely used to treat diarrhoea, skin and eye diseases [7,8,10]. Objective of this study was to identify antifungal potential of different plant extracts of *Aegle marmelos* (leaf, fruit and stem) against fungal strains.

Material and Methodology

Plant parts (leaf, fruit and stem) were washed, air dried and grinded into powder form for preparation of extract. Aqueous plant extract was prepared by macerating powdered plant sample with 50 ml sterile distilled water. The macerate was filtered and filtrate was centrifuged at 8000 rpm for 15 minutes. Supernatant obtained after centrifugation was heat sterilized at 120^o C for 30 minutes. Extract obtained was preserved aseptically. Solvent extracts of plant parts were prepared in 70% ethanol/ methanol using Soxhlet extraction [6] for 72 hours and extract was preserved at 4^o C in air tight bottles. 1mg of each solvent residue was re dissolved in 1ml of respective solvent and were used as test extract for antifungal activity.

Test Fungal Strains

The test fungal strains namely *Aspergillus niger* MTCC 282, *Penicillium chrysogenum* MTCC 161, *Candida albicans* MTCC 183, *Fusarium solani* MTCC 9667 were used to study antifungal potential. They were collected from Institute of Microbial Technology, Chandigarh, India.

Antifungal activity assessment

In vitro antimicrobial activity was screened by using Potato Dextrose agar (PDA) using agar well diffusion method [1]. Fungal strains were activated in Potato Dextrose broth (PDB) and incubated for 24 hours. 0.05ml of inoculum was uniformly spread on agar plates. Ethanolic, methanolic and aqueous extracts were introduced in agar wells in concentration of 25PPM, 50PPM, 75PPM and 100PPM. Control experiment was carried out with Glucanazole. Antifungal potential was then determined on the basis of diameter of zone of inhibition.

Table 1: Zone of Inhibition of Fruit, leaf and Stem of Methanol, Ethanol and Aqueous Extract with test fungal cultures and control drug at 25 PPM

Fungal Strains	Zone of Inhibition (mm)									Control Glucanazole
	Methanolic Extract			Ethanol Extract			Aqueous Extract			
	Fruit	Leaf	Stem	Fruit	Leaf	Stem	Fruit	Leaf	Stem	
<i>Penicillium chrysogenum</i>	17.33±0.47	9.66±0.47	10±0.00	18±1.63	14±1.63	7.33±0.47	12.±1.41	9.66±0.94	6±1.63	9.66±0.47
<i>Fusarium solani</i>	13±0.00	12±1.63	10±0.81	3.66±0.47	13±0.81	15±0.81	10±1.63	14±1.41	13.66±0.94	7±0.00
<i>Aspergillus niger</i>	15±1.41	10±0.00	7.66±0.47	17±0.81	14.33±0.47	12±1.41	11±0.81	14±0.00	7.33±0.94	17±0.81
<i>Candida albicans</i>	21.33±0.94	10±0.81	12±0.81	15.66±0.47	8±1.41	2±0.00	14±1.41	16.66±0.47	3±1.00	10.66±0.47

All values are mean inhibition zone (mm) ± S.D of three replicates

Table 2: Zone of Inhibition of Fruit, leaf and Stem of Methanol, Ethanol and Aqueous with test fungal cultures and control drug at 50 PPM

Fungal Strains	Zone of Inhibition (mm)									Control Glucanazole
	Methanolic Extract			Ethanol Extract			Aqueous Extract			
	Fruit	Leaf	Stem	Fruit	Leaf	Stem	Fruit	Leaf	Stem	
<i>Penicillium chrysogenum</i>	20±1.41	13±0.81	14±0.81	20±1.41	16±1.41	11±0.81	14.66±0.47	14±0.81	8±0.81	13±0.00
<i>Fusarium solani</i>	16.66±0.94	14±0.81	12±0.94	7±1.41	15±0.81	15±0.81	12±0.81	16.33±0.94	17±1.63	10.66±0.47
<i>Aspergillus niger</i>	18±1.41	13.33±0.47	11±1.41	20±1.63	18±0.00	14.66±0.47	13.66±0.47	21.66±0.47	10.66±0.47	20±1.63
<i>Candida albicans</i>	22.66±0.47	14±0.00	14±0.81	18±0.81	12±1.41	4.33±0.47	18±2.15	20.66±0.47	5±1.63	13±0.00

All values are mean inhibition zone (mm) ± S.D of three replicates

Table 3: Zone of Inhibition of Fruit, leaf and Stem of Methanol, Ethanol and Aqueous with test fungal cultures and control drug at 75 PPM

Fungal Strains	Zone of Inhibition (mm)									Control Glucanazole
	Methanolic Extract			Ethanol Extract			Aqueous Extract			
	Fruit	Leaf	Stem	Fruit	Leaf	Stem	Fruit	Leaf	Stem	
<i>Penicillium chrysogenum</i>	23±0.81	15±1.41	19±1.41	22±1.63	20.33±0.47	13±1.41	18±0.81	18±0.00	10.66±0.47	16±0.81
<i>Fusarium solani</i>	21.33±0.47	16±0.81	15±0.81	8.66±1.69	17±1.41	19±0.00	14±0.00	18±0.81	19.33±1.24	12.66±0.47
<i>Aspergillus niger</i>	21±0.81	15±0.81	14±1.41	22.33±0.94	22±0.81	16±0.81	18±1.41	24.33±0.94	13±0.00	24±0.81
<i>Candida albicans</i>	25±0.00	19.66±0.47	16.33±0.47	21±1.41	15.33±0.47	8±1.41	22±1.41	23±0.00	7±1.41	17±0.81

All values are mean inhibition zone (mm) ± S.D of three replicates

Table 4: Zone of Inhibition of Fruit, leaf and Stem of Methanol, Ethanol and Aqueous with test fungal cultures and control drug at 100 PPM

Fungal Strains	Zone of Inhibition (mm)									Control Glucanazole
	Methanolic Extract			Ethanollic Extract			Aqueous Extract			
	Fruit	Leaf	Stem	Fruit	Leaf	Stem	Fruit	Leaf	Stem	
<i>Penicillium chrysogenum</i>	26±0.81	19±1.41	22.66±0.47	25±0.81	23±0.00	14.66±0.47	19.66±0.47	22±0.81	14±0.00	20±0.81
<i>Fusarium solani</i>	24±0.00	18±1.41	19±1.41	11±1.41	20.66±0.47	23.33±0.47	17.33±0.47	22±1.41	21±0.81	17±1.41
<i>Aspergillus niger</i>	23.33±0.47	19±0.81	17±0.00	25±1.47	25±1.47	18±1.47	22±1.41	26±0.81	15±0.81	27±0.00
<i>Candida albicans</i>	27±2.15	20±0.81	18±0.00	24.33±0.94	18.33±0.47	12±1.41	25±1.41	26±0.81	9.66±0.94	20±1.63

All values are mean inhibition zone (mm) ± S.D of three replicates

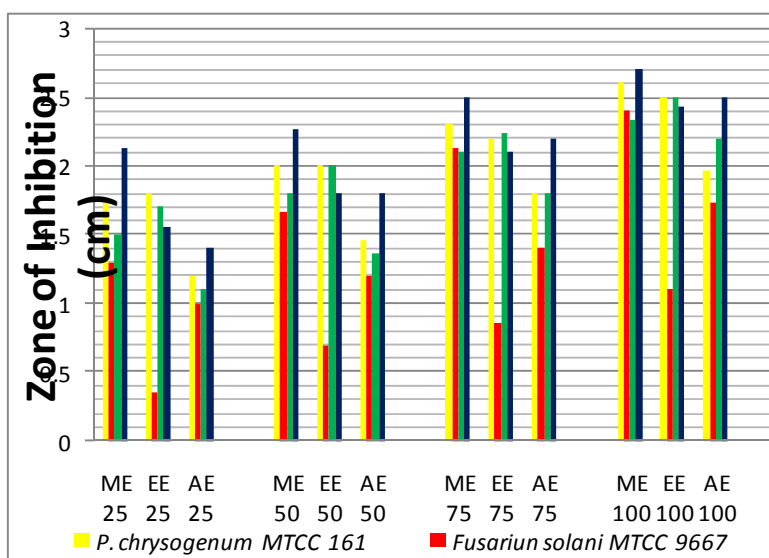


Fig-1: Comparative Study of Methanolic, Ethanollic & Aqueous Extracts of fruit of *Aegle marmelos* at different Concentration (25 PPM, 50 PPM, 75 PPM, 100 PPM) against fungal test strains.

RESULT AND DISCUSSION

All the concentrations of plant extract had shown activity against test fungal organisms. The results showed that increase in concentration of extract increased zone of inhibition. *Penicillium chrysogenum* and *Candida albicans* were most susceptible to methanolic fruit extract by forming inhibition zone of 17.33 to 26mm and 21.33 to 27mm respectively (graph). Methanolic leaf extract was found to be less effective for *Penicillium chrysogenum* and *Fusarium solani*. Petroleum ether leaf extract of *Aegle marmelos* has shown no zone of inhibition for *Penicillium chrysogenum* [15].

Maximum activity against *Aspergillus niger* was shown by leaf extract in aqueous solution by forming inhibition zone of 14mm, 21.66, 24.33mm and 26mm at 25PPM, 50PPM, 75PPM and 100PPM respectively. Researchers have suggested antifungal activity of medicinal plants against *A. niger* and *Candida albicans* [4,11]. Unsaponifiable matter of Bael seeds has also shown *invitro* activity against various fungi namely *A. fumigatus*, *A. niger* and *A. flavous* [13]. Ethanollic stem extract showed notable antifungal potential for *Fusarium solani* at 100PPM (ZI=23.33mm) (table 4) and ethanollic fruit extract showed very less activity (ZI= 11mm) for the same strain at same concentration.

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