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Antifungal activity of different leaf extracts against *Alternaria alternata* (fr.) Keissler.

Arak Sanghamitra V and Sawant RJ

Research centre in Botany Shri Muktanand College, Gangapur -431109
*Corresponding author Email: sanghamitraarak@gmail.com

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Abstract

The present investigation was carried out to isolate the fungal pathogen of *Alternaria alternata* (Fr.) Keissler from some ornamental plants.The antifungal activity was studied by using different selected medicinal plant leaf extracts. The fungal pathogen was isolated on potato dextrose agar medium. The crude extracts were prepared from *Adhatoda vasica* L., *Aegle marmelos* L., *Annona squamosa* L., *Azadirachta indica* L., *capsicum annum* L., *Datura inoxia* L., *ocimum sanctum* L., *Nerium indicum* L. *etc.* Out of these extracts *Azadirachta indica* L. has given promising results in cup plate method. The study of antifungal activity is useful for bio-control of fungal diseases

Keywords: Fungal leaf spot, anti-fungal activity.

Introduction

Ornamental plants are grown in world for beautification and commercial purpose. They are mainly cultivated for attractive flowers and foliage but some of them beneficial for medicinal and industrial value [1]. These ornamental plants are susceptible to the different diseases. Mostly the Fungal diseases found on plants [2]. They usually affected on leaves, buds, flowers etc. For the management of fungal diseases used biological and chemical methods. Medicinal values of different leaf extract have been reported by many workers [3-5].

The present investigation is to control of one of fungal pathogen *Alternaria alternata* (Fr.) Keissler by doing *in vitro* experiment.

The crude leaf extract of different medicinal plants namely *Adhatoda vasica,*L. *Aegle marmelos* L., *Annona squamosa* L., *Azadirachta indica* L., *capsicum annum* L., *Datura inoxia* L., *ocimum sanctum* L., *Nerium indicum* L. *etc* were used for antifungal activity.

Methodology

Collection and isolation of disease samples:
Diseased plant materials were collected from different regions like gardens, fields, orchards of Aurangabad district. The diseased samples were kept in pre-sterilised polythene bags and brought to the laboratory for further investigations. The fresh samples were used for isolation of the pathogen.

b) Isolation & Identification of fungal pathogen:
The infected leaves were washed with 0.1% $HgCl_2$ solution for about 30 to 60 seconds and then washed with sterilised distilled water. A piece of infected tissue from the infected plant part was used for isolation of fungi. The infected tissue segment were cut aseptically and transferred to medium known as Potato Dextrose Agar (PDA). The isolation was carried out at 24 ± 2 ^{o}C and the growth of the pathogen was observed after 7 days. The isolated fungus was purified and multiplied on PDA slants. These slants were used for further study.

Preparation of leaf extracts:
The leaves of medicinal plants were collected from different areas. Leaves were thoroughly washed under tap water and then rinsed with sterile distilled water. 5gms of leaves were crushed in mortar and pastel. The paste was made by adding 10 ml sterile distilled water. Then a paste was centrifuged to ultracentrifuge for 20 min at -4℃ at the 11000 rpm.

Cup plate method:
It is a method of studying antifungal activity. For this the antifungal suspension was prepared by adding 10 ml sterile distilled water to 2 days old fungus culture. Five drops of antifungal cell suspension were poured in sterilized petriplates (9 cm diameter) on to which The 20 ml of Potato Dextrose Agar was pour on petri plates thoroughly mixed and allowed to solidify. In the centre of the medium a cup cavity of 8mm diameter was made with sterilized no. 4 cork borer. This cup was filled with 0.1 ml of the leaf extract by using of micropipette. Petridishes were incubated for 5 to 6 days at 25±2℃. And the observations were recorded as diameter of inhibitory zone measured in 3-4 angles and mean was considered for accuracy. Cup cavity filled with sterile distilled water was used as control in experiments

Results and Discussions

As per the observations on cultured nutrient agar plates, antifungal activity of Leaf extract evaluated against *Alternaria alternata* (Fr.) Keissler The highest zone of inhibition observed in *Azadirachta indica* L. (Mean =16.5mm), *Datura inoxia* L.(mean = 11.5) *Capsicum annum L.* (Mean = 10.25) as compare to other leaf extracts.

Table 1. Antibacterial activity of leaf extract. Showing Zone of inhibition. (in mm)

Sr no	Name of plants	Exp A	Exp B	Exp C	Exp D	Mean
1	*Adhatoda vasica* L.	-	-	-	-	-
2	*Aegle marmelos* L.	-	-	-	-	-
3	*Annona squamosa* L.	-	-	-	-	-
4	*Azadirachta indica* L.	18	15	16	17	16.5
5	*capsicum annum* L.	10	11	10	10	10.25
6	*Datura inoxia* L.	14	11	11	10	11.5
7	*ocimum sanctum* L.	-	-	-	-	-
8	*Nerium indicum* L.	-	-	-	-	-

The Antifungal activity against *Alternaria alternata* a studied by Zakir and Mosallanejad in 2010[6]. Singh *et al* were also recorded antifungal activity against *Alternaria alternata* in 2014[7].The antimicrobial acitivity of ethnolic leaf extract were studied by Khalil 2012 in Sudan also Antibacterial acitivity of *A.indica* was dony by Mohmmad and Omer 2015[8]. Sheema and Durai also studied antifungal activity against *Alternaria brassicae* by using aqueous leaf extract in 2014[9]. Chudhary *et.al* were also recorded antifungal activity of Ethanolic extract against diseased rice plant[10]. Satpute and Vanmare [11] were studied antifungal activity of *Tamarindus indica* L. Against pathogenic Fungi in Aurangabad Maharashtra.

Conflicts of interest: The authors stated that no conflicts of interest.

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