Antifungal Activities of Leaf and Stem Extract of the Medicinal Plants

Dr. Prem Singh Meena, Associate Professor
Department of Botany, Government College,
Tonk

Abstract

The goal of this study was to assess the antifungal properties of two different medicinal plants, including *Cassia fistula* and *Acacia nilotica*, by identifying the zone where the leaf extract inhibited certain fungal strains. After examining the extract's antifungal capabilities at various concentrations (25, 50, 75, and 100 μg/ml). The Ethanolic, Petroleum ether and aqueous extract of leaf was tested against *F.moniliforme* ,*F.oxysporum* and *F.graminearum.*
Keywords: Zone of inhibition, Antifungal Activities , Fungal strains, *Cassia fistula, Acacia nilotica*.

1. INTRODUCTION

The primary infectious agents in plants, even post-harvest, are pathogenic fungi, which influence plant development at all stages. Numerous fungus genera exist in fruit and vegetables, and they can cause issues with aspect, nutritional value, organoleptic qualities, and shelf life (1). Additionally, because they produce mycotoxins or allergens, fungi are occasionally indirectly to blame for allergic or toxic illnesses in consumers. Synthetic fungicides are typically used to control phytopathogenic fungus, but their usage is becoming increasingly limited as a result of the negative impacts pesticides have on both human health and the environment (2). The search for novel active molecules and new control methods is justified by the rising demand for production, the restrictions on the use of agrochemicals,and the advent of infections resistant to the products used. Since ancient times, the plant kingdom has produced several substances with well-known therapeutic qualities, including analgesics, anti-inflammatories, asthma medications, and others. Plant extracts have been found to have antibacterial activities more frequently in recent years from all over the world (3).

Cassia fistula known as Golden Shower in English, is a Leguminosae family plant, used as Hepatoprotective, anti-inflammatory, coμgh-suppressant, anticancer, antioxidant, and antibacterial activities (4).

Acacia nilotica It is a multifunctional herbal plant with important medicinal characteristics that has long been used to treat a variety of diseases (5).

2. MATERIALS AND METHODS

2.1. Plant sample collection

The healthy leaves of *Cassia fistula* and *Acacia nilotica*, were collected, and brought to the laboratory. The leaves were rinsed under running water and allowed to air dry at room temperature before being ground up in a blender and placed in airtight plastic bags for further examination. According to APG IV classification, every plant underwent botanical authentication.

2.2. Preparation of plant extract

The plants were disinfected and shade-dried for buffer stock. The plant pieces were appropriately ground into powder using grinder before being extracted using a Soxhlet Extraction Unit. For each plant part, a 1:10 mixture of Ethanol, Petroleum ether and water was used to make the extracts. For additional research, freshly obtained extracts were divided into concentration groups of 25, 50, 75, and 100 μg/ml.

2.3 Fungal strain

The following harmful fungi strains were gathered from Chandigarh, (IMTECH) gene bank and Microbial Type Culture Collection.

a. F.moniliforme
b. F.oxysporum
c. F.graminearum

2.4 Determination of antifungal activity

Agar well diffusion method is widely used to evaluate an antifungal drμg's effectiveness. Potato dextrin Agar was added to the petri dish, and after 20 minutes, the agar plates solidified. Spread Fungus liquid culture (1ml) is then dispersed using a spreader onto the hardened agar plate.

Each plate has four 5mm-diameter wells that were excavated with the use of a sterile corn borer. Please indicate 25, 50, 75, and 100 µg/ml plates with wells in them. The several plant samples were made by combining different plant extracts with ethanol, petroleum ether, and water at concentrations of 25, 50, 75, and 100 µg/ml. A decontaminated syringe was used to inject 80–100 ul of solution in different dilutions from different extracts into the well, which was maintained at 4°C for 10 minutes. Following that, 28°C fungal pathogen culture was performed on each plate for 46–48 hours. At each and every well, the diameter of the inhibitory zone was measured to record the results (mm). In a replication of three, the components of all plants were tested against each fungal strain taken into account for the tests for each extract (all solvents).

2.5 Statistical Analysis

Statistical software was used to calculate the mean values for the plant extracts' zones of growth inhibition from the data. Antibacterial activity against the bacterium was deemed to be active at values below 9 mm, and antifungal activity was deemed to be active at values above 15 mm (6).

3. RESULTS

In terms of antifungal efficacy of *Cassia fistula*, leaf extracts were recorded highest antifungal activity than stem extracts against all fungus . Significant antifungal activity was observed by the Leaf ethanol extract against *F.moniliforme* at 100 µg/ml (24mm). A minimum antifungal activity test was performed on a 100 µg/ml stem water extract against *F.oxysporum*(15mm).

In terms of antifungal efficacy of *Acacia nilotica* Compared to leaf extract, stem extract has higher antifungal efficacy against all fungi. Stem ethanol extract at 100 µg/ml (25 mm) significantly reduced the growth of the fungus *F. moniliforme*. The minimum antifungal activity (15mm) of 100 µg/ml stem ethanol extract was tested against *F. oxysporum.*

Table :1 Zones of inhibition of extracts against *F.moniliforme* at different concentrations

Plant Name	**INHIBITION AT DIFFERENT CONC.(IN mm)**											
	25 µg/ml			**50** µg/ml			**75** µg/ml			**100** µg/ml		
	W	**E**	**P.E.**	**W**	**E**	**P.E.**	**W**	**E**	**P.E.**	**W**	**E**	**P.E.**
Cassia fistula(Leaf)	11	9	10	14	15	12	20	21	18	22	24	20
Cassia fistula(Stem)	11	12	9	13	15	12	17	20	16	21	22	19
Acacia nilotica(Leaf)	8	10	7	13	15	11	19	20	18	20	21	19
Acacia nilotica(Stem)	10	11	9	14	16	12	20	21	25	21	25	20

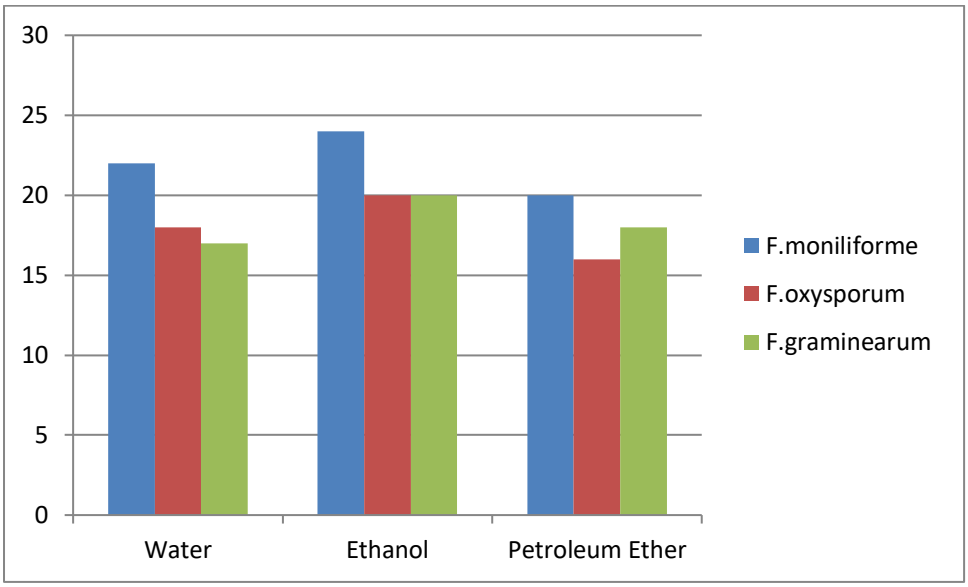
W=Water,
E=Ethanol
P.E.= Petroleum Ether

Table :2 Zones of inhibition of extracts against *F.oxysporum* at different concentrations

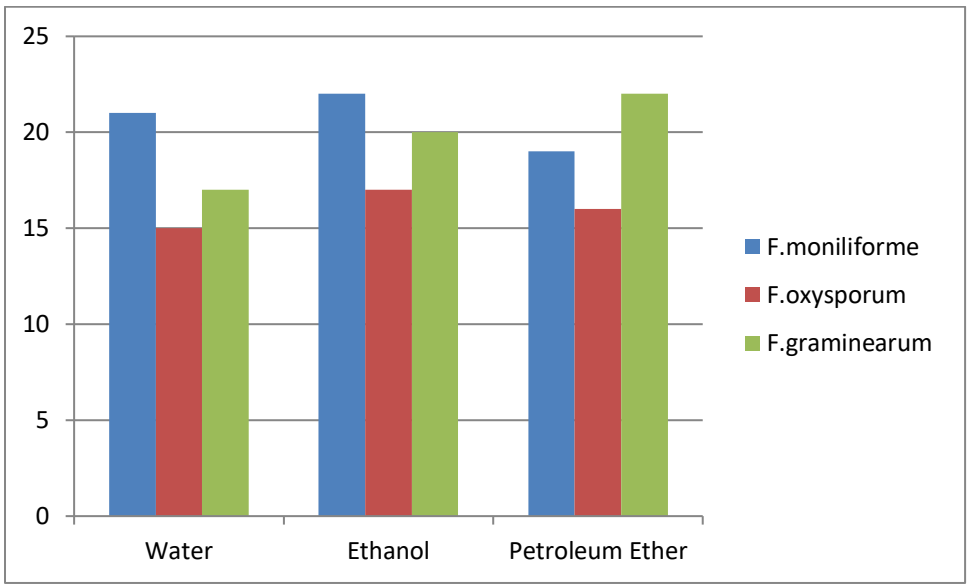
Plant Name	**INHIBITION AT DIFFERENT CONC.(IN mm)**											
	25 µg/ml			**50** µg/ml			**75** µg/ml			**100** µg/ml		
	W	**E**	**P.E.**	**W**	**E**	**P.E.**	**W**	**E**	**P.E.**	**W**	**E**	**P.E.**
Cassia fistula(Leaf)	8	7	8	13	14	12	16	17	14	18	20	16
Cassia fistula(Stem)	10	11	8	11	13	11	12	14	13	15	17	16
Acacia nilotica(Leaf)	8	8	7	9	11	10	12	14	13	15	17	16
Acacia nilotica(Stem)	9	8	9	11	10	11	16	14	15	17	15	16

Table :3 Zones of inhibition of extracts against *F.graminearum* at different concentrations

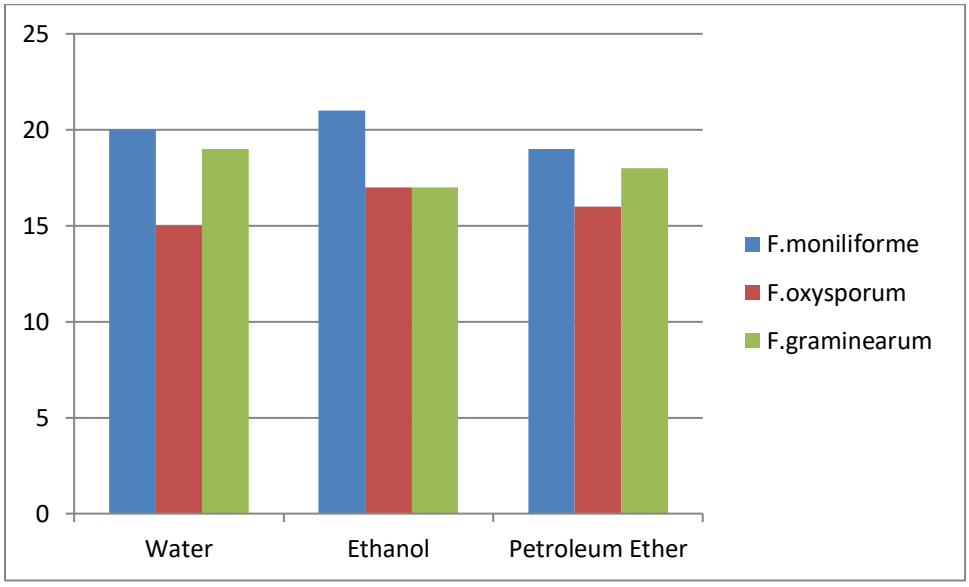
Plant Name	**INHIBITION AT DIFFERENT CONC.(IN mm)**											
	25 µg/ml			**50** µg/ml			**75** µg/ml			**100** µg/ml		
	W	**E**	**P.E.**	**W**	**E**	**P.E.**	**W**	**E**	**P.E.**	**W**	**E**	**P.E.**
Cassia fistula(Leaf)	10	12	12	12	17	15	14	18	17	17	20	18
Cassia fistula(Stem)	12	9	16	14	15	18	16	20	21	17	20	22
Acacia nilotica(Leaf)	13	11	12	15	14	14	18	16	17	19	17	18
Acacia nilotica(Stem)	12	13	12	13	16	12	15	18	14	17	20	16



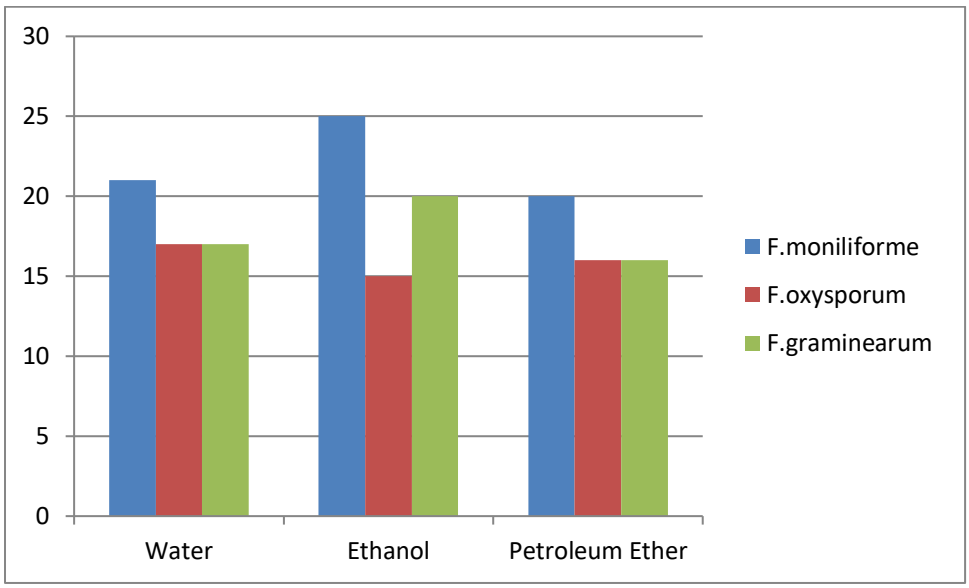
Graph1 . *Cassia fistula* (Leaf) extracts 100 µg/ml and zone of inhibition of Fungus



Graph2: *Cassia fistula* (Stem) extracts 100 µg/ml and zone of inhibition of Fungus



Graph 3. *Acacia nilotica*(Leaf) extracts 100 µg/ml and zone of inhibition of Fungus



Graph4 *Acacia nilotica* Stem extracts 100 µg/ml and zone of inhibition of Fungus

DISCUSSION:

Numerous researchers found that *Cassia fistula* contains a wide range of phytochemicals and is efficient against numerous harmful fungus strains.

Phytochemicals such alkaloids, flavonoids, carbohydrates, glycosides, protein and aminoacids, saponins, and triterpenoids indicated that polar extracts of Cassia fistula (ethanol, methanol, and water) contained the bulk of these components when compared to nonpolar extracts (petroleum ether and chloroform)(7).

Cassia fistula extracts were tested against three different fungi at concentrations of 5, 25, 50, 100, and 250 g/ml to determine their antibacterial and antifungal properties (Aspergillus niger, Aspergillus clavatus, and Candida albicans)(8) Different tree extracts were used to assess the spore germination, mycelial growth, and mycelial weights of two fungi, Penicillium italicum and Aspergillus niger. Using the inhibitory zone and radial growth approaches, mycelial development on PDA medium was encouraged. After extraction using different solvents, it was shown that methanol, diethyl ether, acetone, and the aqueous extracts were all extremely successful at preventing the growth of both fungi. The results also showed that different tree parts had different effects, with bark and pod extracts being the most effective (9).

4. Conclusion

The findings of this study demonstrated that the screened medicinal herbs have antifungal effects on fungus species more specifically. Plant extracts provide efficient bioactive ingredients for fungus growth inhibition. These species demonstrated antifungal activity even at low doses that was almost on par with the commercial fungicide employed as a positive control. To identify the chemical composition of the bioactive substances responsible for the reported antifungal action, more research is required. Natural fungicides generated from plants may provide novel, alternative active substances, particularly those with antifungal activity. The assayed species' high percentage of active extracts, which were chosen based on existing ethnobotanical data.

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