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Evaluation of antifungal activity of Zingiber officinale against Fusarium oxysporum f.sp. lycopersici

Pratibha Rawal* and Rajendra Singh Adhikari

Botany Department, L. S. M. Govt. P.G. College Pithoragarh, Uttarakhand, India

ABSTRACT

Ginger (Zingiber officinale) has long been used as naturopathy due to its potential antimicrobial activity against different pathogens. This study was conducted to determine the antimicrobial activity of dried ginger powder, using paper disc diffusion assay, by using chloroform, ethanol, acetone and petroleum ether solvents, against Fusarium oxysporum f. sp. lycopersici. The present study showed the potent antimicrobial activity of the ginger extract against the pathogen by all tested solvents. Chloroform extract of ginger at its 750 mg/ml concentration showed highest zone of inhibition as 25.75 mm against tested pathogen. Whereas, other solvents showed moderate to minimum antifungal activity. These findings suggests that some plant extracts tested possess antifungal activities against Fusarium oxysporum f.sp. lycopersici.

Keywords: Fusarium oxysporum f.sp. lycopersici, Zingiber officinale, paper disc diffusion assay

INTRODUCTION

The Fusarium wilt of tomato (*Lycopersicon esculantum* Mill) caused by *Fusarium oxysporum* f. sp. *lycopersici*. It (Fol) is recognized as a devastating disease in tomato growing areas all over the world (2) (3), also in different regions of India from severe to moderate (50-60%) percentage (10) (7).

All over the world, the use of medicinal plants has significantly helpful for primary health care.

Plants possess antimicrobial properties because of the presence a wide variety of secondary metabolites, such as tannins, terpenoids, alkaloids, flavonoids etc. Microbiologists have strongly two reasons to be interested in the antimicrobial properties of plant extracts and their scopes in medical science. First, it is very likely discovered that these phyto chemicals will find their way into the arsenal of antimicrobial drugs prescribed by physicians. It is reported that, approximately two or three antibiotics at an average rate derived from microorganisms are launched each year (4).Second reason is that, the public is becoming aware of problems with the over prescription and overdose with antibiotics and at the same time because of the alter effects of allopathic medicinal system. It is estimated that there are 250,000 to 500,000 species of plants on Earth, A relatively very small percentage (1 to 10%) of these are used as foods by both humans and other animal species. It is possible to use them more for medicinal purposes (9).

Plant secondary metabolites are of low-molecular weight compounds that are not important for sustaining life, but are essential for the survival of the producing organism (6). Hydrophilic compounds (such as alkaloids, flavonoids, tannins, and saponins) are stored in the vacuole while the lipophilic SMs (such as terpenoids) are sequestered in

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resin ducts, laticifers, oil cells, trichomes, or in the cuticle. Bioactive compounds affect the tissues and cells of fungi via interference. The major targets portions includesBiomembrane, proteins and nucleic acids. These bioactive compounds are still regarded as a valuable pool for discovering novel mode of action (6).

Medicinal plants extracts have been used and studied extensively for their antimicrobial activity and have been demonstrated as good plant disease control agents (11).

Now a days, public pressure to reduce the use of synthetic fungicides in agriculture has increased. Concerns have been raised for both the environmental impact and the potential health risks. Hence, there is a great demand for safer antifungals belonging to a wide range of structural classes, which are selective on new targets with less side effects (1).

Ginger (*Zingiber officinale*) is a medicinal plant that has been widely used all over the world for various purposes. *Zingiber officinale* commonly known as Ginger, belongs to Zingiberaceae family (8). Rhizome or root part of ginger is extensively used in medicine for the management of different diseased conditions like nausea, vomiting, motion sickness, gastrointestinal ulcers, diabetes, fever, arterial tension, rheumatoid arthritis, dry mouth/xerostomia, cancer, migraine headache, sore throat and other minor respiratory ailments.

Ginger has been valued for its antibacterial properties for thousands of years in Asian cultures (12). The aim of this study is to investigate the effect of plant extract (ginger) on tomato wilt pathogen.

MATERIALS AND METHODS

The fresh ginger rhizomes were collected, cleaned, peeled, sliced and dried at room temperature. After drying, pieces of *Zingiber officinale* were grinded to fine particles in isolated manner utilizing a suitable grinder.

2.0 gram of organic extracts were prepared by extracting sample successively with chloroform, ethanol, acetone and petroleum ether in a ratio of 1:5. The resultant extract was weighed and stored in airtight sample bottles. All the extracts were oven evaporated till complete dryness for 5 -7 days at 30 ± 2 °C. Plant extract was second time extracted with DMSO. 200 mg of each extract was weighed into a sterilized sample bottle and dissolved in DMSO (Sigma) to make a concentration of 200 mg/ml. For the study 250, 500, 750 mg/ml concentrations were prepared.

Microorganism used in this study *Fusarium oxysporum* f.sp. *lycopersici* was obtained from Microbial Type Culture Collection MTCC, Institute of microbial technology, Chandigarh. Fungus culture (MTCC 1755) was maintained in Potato sucrose agar medium for an optimum pH of 6.8. The fungal spore suspension was prepared by the addition of a loopful of fungal spores in a 5 ml of sterile distilled water and 1 ml Tween 20. Then 0.1 ml fungal spore suspension was mixed well in aseptic conditions and spread evenly on the petridishes containing 20 ml of solidified potato sucrose agar.

ANTIMICROBIAL ACTIVITY

In Paper disc diffusion method some amount of Potato Sucrose Agar (PSA) was dispersed in petridishes and allowed to solidify. A micropipette will be used to introduce 0.1 ml. spores on agar medium and spread with glass rod spreader under sterile conditions. Sterilized discs (6 mm, Whatmann No. 1 filter paper) will be prepared by soaking in different concentrations of the extracts ie, 250, 500, 750 mg/ml for 6 hour. The discs will be then removed and allowed to dry. To assay for antifungal activity various discs impregnated with different concentrations of the extracts will be placed on the fungal spore or mycelium with the help of sterilized forceps. The petridishes incubated at 35 °C for 48 h. Antifungal activity will be determined by measurement of the zone of inhibition around the discs after the period of incubation.

Experiments were performed in the laboratory of L.S.M.G.P.G. College Pithoragarh.

DATA ANALYSIS

Data from antifungal activity screening were analyzed using simple statistics from Microsoft Excel and recorded in appropriate tables as mean \pm standard deviation of mean.

RESULTS AND DISCUSSION

Table -1- showed that the rhizome extract of *Zingiber officinale* was very effective against the tested pathogen *Fusarium oxysporum* f.sp. *lycopersici*. The chloroform extract showed 15.87 mm inhibition zone at 250 mg/ml concentration. 500 mg/ml concentration was moderately effective with 20.25 mm inhibition zone. At 750 mg/ml the zone of inhibition was observed to be as 25.75 mm (PLATE-1).

The ethanol extract showed 11.00 mm inhibition zone at 250 mg/ml concentration. 500 mg/ml concentration was effective with 16.12 mm inhibition zone. 16.27 mm inhibition zone was observed at 750 mg/ml (PLATE-2). The acetone extract showed 13.50 mm inhibition zone at 250 mg/ml concentration. 500 mg/ml concentration was moderately effective with 15.50 mm inhibition zone. At 750 mg/ml the inhibition zone was observed to be as 25.00 mm (PLATE-3)

While its petroleum ether extract showed 15.00 mm inhibition zone at 250 mg/ml concentration. 500 mg/ml concentration was effective with 16.87 mm inhibition zone. 20.50 mm inhibition zone was observed at 750 mg/ml concentration (PLATE-4).

Table.1-In -vitro antifungal activity of Zingiber officinale, dried rhizome extract at various concentrations against fungal pathogen Fusarium oxysporum f.sp. lycopersici

Plant Name	Concentrations (mg/ml)	Mean Inhibition Zone in various solvents (In mm)			
Zingiber officinale		Chloroform	Ethanol	Acetone	P.E.
	250	15.87 ± 0.8	11.00 ± 2.5	13.50 ± 1.9	15.00 ± 0.8
	500	20.25 ± 1.7	16.12 ± 1.6	15.50 ± 1.2	16.87 ± 1.4
	750	25.75 ± 2.7	16.27 ± 1.4	25.00 ± 1.7	20.50 ± 1.2



Results are the mean of four replications \pm S.D

Graph.1 Antifungal activity of Zingiber officinale



PLATE-1 Antifungal activity of chloroform extract of Zingiber officinale



PLATE-2- Antifungal activity of ethanol extract of *Zingiber officinale*



PLATE-3- Antifungal activity of acetone extract of *Zingiber officinale*



PLATE-4-Antifungal activity of petroleum ether extract of *Zingiber officinale*

The use of phytochemicals derived from plants, with known antimicrobial properties, are of great significance to medicinal treatments (3).

The results of this study showed that of all the extracts screened, zinger chloroform extract had ahigher inhibitory activity against the test organism. This could be as a result of better extraction with chloroform solvent. Other solvents were similarly effective against the tested pathogen.

Ginger extract containing Gingerol inhibits the growth of many bacteria and fungi *in-vitro* and the activity might be contributed to the preventive effects of its different agents (5).

CONCLUSION

The results of this study suggest a fairly good correlation between medicinal use and the *in vitro* antifungal activity. It has been concluded from present research that certain plant extracts are a source of cheap and effective fungicides of *Fusarium oxysporum* f.sp. *lycopersici*, also it doesn't have human and environment health implications.

Fusarium oxysporum f.sp.*lycopersici* can cause severe damage in tomatos. It has been concluded from the present research that certain plant extracts are the source of cheap and effective fungicide of *Fusarium oxysporum*, also they don't have human and environmental health implications.

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