SHORT COMMUNICATION

EFFECT OF CULTURE FILTRATES OF PHYLLOPLANE FUNGI ON CONIDIAL GERMINATION OF *ALTERNARIA ALTERNATA* AND *COLLETOTRICHUM CAPSICI*

Gunjan Joshi, D.K. Jain, P.N. Singh and Reena Bansal

Department of Botany, Meerut College, Meerut (U.P.)

Abstract: *In vitro* antagonistic activity of culture filtrates of phylloplane fungi was tested against pathogens causing diseases of *Impatiens balsamina*. The conidia germination of *Alternaria alternata* was greatly inhibited by culture filtrate of *Trichoderma viride* followed by *Aspergillus flavus, A. niger, Chaetomium globosum* and *Curvularia lunata.* Germ tube growth was inhibited by all test fungi, maximum by *Trichoderma viride* followed by *Aspergillus niger, A. flavus* and *Chaetomium globosum.* Maximum inhibition of conidia germination of *Colletotrichum capsici* was caused by *Trichoderma viride* followed by *Aspergillus niger, Fusarium oxysporum, Aspergillus flavus* and *Chaetomium globosum.* Maximum inhibition of germ tube growth was also caused by *Trichoderma viride* followed by *Aspergillus niger, Fusarium oxysporum* and *Aspergillus flavus.*

Keywords: Fungi, Germination, *Alternaria alternata,* Colletotricum capsici

INTRODUCTION

Impatiens balsamina L. (a member of family Balsaminaceae) are tender, succulent herbs commonly grown as bedding and house plants. It has both ornamental and medicinal values. A lotion of fresh leaves is used to treat eczema, itches and insect bites. A decoction of flowers is taken for infections, vomiting, urine retention and as a tonic. In India, flowers are regarded as cooling, tonic and useful when applied to burns and scalds. The flowers are also used for lumbago and intercostals neuralgia, snakebite, improves circulation and relieve stasis. *Alternaria alternata* (Fr) keissler and *Colletotrichum capsici* (Syd.) Butler and Bisby are two main pathogen on *Impatiens balsamina* causing leaf spot diseases.

Several effective pesticides havebeen recommended against the pathogen, but they are not considered to be long term solutions due to concerns of expense, exposure to health risk, fungicide residue and other enviornmental hazards. Bioagents are considered as new ray of hope, because they are ecofriendly, biodegradable, and can be used as an effective alternative measure to control plant disease.The aim of present investigation is to study the antagonistic effect of leaf surface saprophytic fungi on pathogen causing diseases of *Impatiens balsamina* L., because a little work has been done on the antagonism between the pathogen and leaf surface fungi over this plant.

MATERIAL AND METHODS:

Isolation of leaf pathogens

Leaves of *Impatiens balsamina* infected with *Alternaria alternata* and *Colletotrichum capsici* were collected separately.The leaf discs were surface sterilized with 0.01 % mercuric chloride for 60 seconds, washed in three changes of sterilized distilled water, blotted dry and five such leaf discs were placed in each Petri dish containing molten, cool and sterilized CDYA medium. The developing fungi were identified. Both fungi were tested for their pathogenicity to *Impatiens balsamina* using Koch's postulates. The pure culture of pathogenS were maintained on CDYA medium.

Biological control agent and their culture

Aspergillus flavus, A. niger, Chaetomium globosum, Cladosporium cladosporioides, Curvularia lunata, Drechslera australiensis, Fusarium oxysporum, Myrothecium roridum, Phoma humicola and *Trichoderma viride* were isolated from *Impatiens balsamina* leaf. Above leaf surface fungi were maintained in pure culture on sterilized CDYA slants for study.

Effect of culture filtrates of phylloplane fungi on conidial germination of *Alternaria alternata* and *Colletotrichum capsici*

This aspect was studied by growing the above fungi in liquid Czapek's medium. 50 ml of sterilized medium was taken in 150 ml sterilized Borosil conical flasks. It was inoculated with 5 mm disc of the test fungus cut by sterilized cork borer obtained from 6 days old cultures grown on Czapek's dox yeast agar medium. The flasks were incubated at 25±1°C for 30 days. Three replicates were maintained for each fungus. The contents of each flask were filtered through Whatman filter paper No. 44. The filtrate was centrifuged at 1,000 rpm for 15-20 minutes to free it from spores and finally collected in sterilized test tubes. Germination of conidia and germ tube growth of both pathogens were noted in the respective culture filtrates of each fungus separately, using hanging drop method. Thirty days old sterilized Czapek's medium served as control.

Table 1. Effect of culture filtrate of test fungi on conidia germination percentage and germ tube length (mm) of *Alternaria alternata* (incubated for 12 h at 25 ± 1°C).

Test fungi	Conidia germination %	Value of t	Inhibition over Control %	Mean germ tube length (mm)
Aspergillus flavus Link ex Fries	30.66±1.21	40.79**	68.23	49
A. niger Van Tieghem	38.19±0.42	39.53**	60.43	45
Chaetomium globosum Kunze ex Fries	40.59±1.16	34.90**	57.94	56
Cladosporium cladosporioides (Fresen.) de Vries	74.55±2.40	10.92**	22.75	102
Curvularia lunata (Walker) Boedijn	45.68±1.96	27.57**	52.67	81
Drechslera australiensis (Bugnicourt) Subram. and Jain ex Ellis	78.56±4.35	6.18**	18.60	97
Fusarium oxysporum Schlechtendahl	57.17±3.11	17.01**	40.76	88
Myrothecium roridum Tode ex Fries	61.79±2.23	17.87**	35.97	92
Phoma humicola Gilman and Abbott	94.29±3.05	0.97	2.30	145
Trichoderma viride Pers.	15.44±1.06	51.36**	84.00	39
Control (Czapek's medium)	96.5±2.52			157

** Significant at 1% level.

Table 2. Effect of culture filtrate of test fungi on conidia germination percentage and germ tube length (mm) of *Colletotrichum capsici* (incubated for 12 h at 25 ± 1°C).

Test fungi	Conidia germination %	Value of t	Inhibition over Control %	Mean germ tube length (mm)
Aspergillus flavus Link ex Fries	39.78±2.27	24.61**	56.29	82
A. niger Van Tieghem	28.82±1.08	35.89**	68.33	56
Chaetomium globosum Kunze ex Fries	45.11±2.42	21.48**	50.43	84
Cladosporium cladosporioides (Fresen.) de Vries	70.98±3.12	8.27**	22.00	124
Curvularia lunata (Walker) Boedijn	51.59±3.07	16.42**	43.30	106
Drechslera australiensis (Bugnicourt) Subram. and Jain ex Ellis	75.55±4.13	5.36**	16.98	118
Fusarium oxysporum Schlechtendahl	32.76±1.22	33.03**	64.00	75

Myrothecium roridum Tode ex Fries	60.99±2.16	14.69**	32.98	98
Phoma humicola Gilman and Abbott	86.27±2.50	2.18	5.20	152
Trichoderma viride Pers.	18.17±1.04	42.23**	80.03	43
Control	91.00±2.80			176

** Significant at 1% level

RESULT AND DISCUSSION

The filtrates from 30 day old cultures of all test fungi caused inhibition of *Alternaria* and *Colletotrichum* conidia germination to varying extents. Maximum inhibition of conidia germination of *Alternaria alternata* was caused by *Trichoderma viride* followed by *Aspergillus flavus, A. niger, Chaetomium globosum* and *Curvularia lunata*. Maximum germ tube growth inhibition was caused by *Trichoderma viride* followed by *Aspergillus niger, A. flavus* and *Chaetomium glabosum* (Table 1).

In case of conidial germination of *Colletotrichum capsici* maximum reduction was caused by *Trichoderma viride* followed by *Aspergillus niger, Fusarium oxysporum, Aspergillus flavus* and *Chaetomium globosum*. Maximum inhibition of germ tube growth was caused by *Trichoderma viride* followed by *Aspergillus niger, Fusarium oxysporum* and *Aspergillus flavus* (Table 2).

The inhibition of conidial germination by antagonistic fungi may be attributed to antibiotic action. The antibiotic produced by the antagonistic fungi might inhibit the conidial germination which led to the disintegration of the viable fungal spore. A similar possibility of involvement of some inhibitory substances in suppressing the germination of spores of some other pathogens has been expressed earlier by Mishra and Tewari (1976) and Rai and Singh (1980). Bhatt and Vaughan (1963) have shown that the sterile filtrates of *Cladosporium herbarum, Dendrophoma* and *Pullularia pullulans*, associated with different stages of fruit development of strawberry, though did not suppress spore germination of fruit rot pathogen *Botrytis cinerea*, but restricted its mycelial growth. Norse (1972) studied that although none of the cell-free culture filtrates of leaf surface fungi of tobacco had significant effect on germination of conidia of *Alternaria longipes* (brown spot disease of tobacco) some of them inhibited germ tube growth of its conidia. While studying phylloplane fungi of sunflower, Gupta and Gupta (1978) observed that culture filtrate of *Aspergillus niger* totally checked the spore germination of the leaf blight pathogen, *Alternaria alternata*. Germination of conidia of *Phyllactinia dalbergiae* was decreased *in vitro* by culture

filtrates of *Cladosporium spongiosum*, a hyperparasite of powdery mildew fungus (Mathur and Mukerji, 1981). Kumar and Singh (1981) also studied that culture filtrates of *Fusarium equiseti, Mucor hiemalis* and *Penicillium aurantiogriseum* were most effective in reducing the spore germination and germ tube length of *Phytophthora infestans* followed by *Actinomucor* sp. Gupta *et al.* (1981) reported that culture filtrates of leaf surface fungi on sunflower, namely *Aspergillus niger, A. luchuensis, A. terreus, Trichothecium roseum, Penicillium simplicissimum* and *Fusarium semitectum* caused 100% inhibition of germination of conidia of the blight pathogen *Alternaria alternata*, followed by *Fusarium moniliforme* and *Cochibolus lunatus* which caused more than 70% inhibition. Bastos *et al.* (1981) demonstrated that the culture filtrate of *Cladobotryum amazonense* completely inhibited spore germination of *Phytophthora palmivora* and *Crinipellia perniciosa*. They also reported that culture filtrates (extra cellular metabolites) of *Cladobotryum amazonense*, a hyperparasite of some agaries including *Crinpellis perniciosa* (cocoa witches broom fungus) completely inhibited the radial growth of *Phytophthora palmivora, Ceratocystis fimbriata* and *Botryodiplodia theobromae* markedly reduced the growth of *Colletotrichum gloeosporioides, Corticum salmonicolor* and *Phellinus noxius in vitro*. Singh (1981) also observed that culture filtrate of *Epicoccum purpurascens* possessed some antagonistic compounds which inhibited the infection of chilli fruits by *Colletotrichum capsici* and C. *piperatum* and of cucurbit fruits by *C. lagenarium, C. capsici* and *Macrophomina phaseolina*.

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