

EVALUATION OF ANTAGONISTIC BEHAVIOR OF SOIL MYCOFLORA OF BILASPUR DISTRICT

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ABSTRACT

The antagonistic potential of native fungal isolates (*Alternaria alternata*, *Cladosporium herbarum* and *Trichoderma viride*) were investigated *In vitro* for antagonistic activity against the most prevalent soil borne pathogens i.e. two fungi *Sclerotium rofsii*, *Botrytis cinerea*, and two bacteria i. e. *Xanthomonas sp.* and *Agrobacterium sp.* (Taken from phytopathology laboratory). Native Fungal isolates have shown effective inhibition on growth pattern of pathogens. Specifically two isolates (*Trichoderma viride* & *Alternaria alternata*) showed maximum inhibition. These antagonists have the properties of potential bio control agent so the isolates proved effective in controlling the pathogen *in-vitro* than the commercial formulation, indicating their superiority in the bio control of phytopathogens.

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KEY WORDS:- Antagonistic potential, Biocontrol, Commercial formulation, Growth inhibition, Phytopathogens

Introduction

The fungi has a large host range of 500 species in about 100 families including flowers, vegetables, cereals, plants and weeds⁷. Several chemical pesticides are used to manage plant pathogenic diseases^{1,10}. Soil born phytopathogenic fungi such as, *Rhizoctonia*, *Pithium*, cause diseases in most of the economically important plants. Sheath blight caused by *Rhizoctonia solani* is one of the most important destructive diseases of rice next to rice blast⁶. *Trichoderma sp.* which is a common saprophytic filamentous fungus in almost any soil and rhizosphere microflora, is well recognized as bio control agent against various plant pathogenic fungi. It is reported that these bioactive substance though chemically different

exhibit antagonistic property against varied bacterial and fungal pathogens³. More over fungicidal application as seed or soil treatment however has been found to be ineffective against these pathogens as the propagules are distributed in the soil and often beyond the reach of chemicals⁴. Obtaining bio-control measure through microorganisms that occur naturally in the area has been made a reality via antibiotics⁹. Biological control therefore holds a promise as a strategy, the objective for disease management and it is environment friendly too. Therefore, the aim of this paper was to found the antagonistic activities of soil inhabiting isolates against pathogenic microorganisms.

Material and Methods

Selection of antagonists for fungal and bacterial phytopathogens:

Three isolates of native soil fungi i.e. *Alternaria alternata*, *Cladosporium herbarum*, and *Trichoderma viride* and Commercial formulation were tested by dual culture technique. For this purpose 15 ml of autoclaved PDA was poured aseptically in to sterile Petri dishes. 2 mm mycelial disc of three fungal isolates and two laboratory strains i.e. *Sclerotium rofsii* and *Botrytis cinerea* (borrowed from phytopatholab) were placed opposite to each other near the periphery of the petri plate and incubated at 27 °C. *Sclerotium rofsii* and *Botrytis cinerea* alone inoculated plate served as control. Mycelial growth of the pathogen was measured and observations were recorded on formation of inhibition zone, over growth and lysis of pathogen mycelium. After the plates were incubated at 27°C for 2-6 days radial growth percentage of the test fungi was measured using following formula.

$$RI(\%) = \frac{Rc - Ri}{Rc} \times 100$$

Where, RI=Radial growth inhibition

Rc= Radial growth in control plates

Ri= Radial growth in incubated plates

For testing the antagonistic activity of fungal isolates against phytopathogenic bacteria, A total of 3 fungal isolates obtained from soil were screened for antagonism against *Xanthomonas sp.* and *Agrobacterium sp.s* (borrowed from phytopatholab). Plates of this medium were inoculated with a transverse streak of spores of the fungus to be examined, or by a mycelial transplant if spores were not sufficiently abundantly produced. The inoculated plates were incubated for 4 days at 25°C. Using a platinum loop, 24 hr. broth cultures of two bacteria (*Xanthomonas sp.* and *Agrobacterium sp.*) were then streaked at right angles to the advancing edge of the fungus colony. The plates were incubated for a further 24-48 hr. at 37°C after which they were examined and the degree of antagonism, as represented by the presence or absence, adjacent to the fungus colony, of a clear zone.

Result and Discussion

During present investigation variety of fungal sp. were collected and identified. On the basis of morphological characteristics nine fungal species were observed via direct microscopic examination. The antagonistic behavior of selected three fungal isolates against phytopathogenic fungi and bacteria have been observed and were mentioned in Table-1-2 and Figure-1-2 respectively.

During study of antagonistic activity against *Sclerotium rofsii* and *Botrytis cinerea* it was found that *Trichoderma viride* and *Alternaria alternata* were more effective against both *Sclerotium rofsii* and *Botrytis cinerea* while *Cladosporium herbarum* was less effective. In case of *Xanthomonas sp.* antagonistic activity were recorded under following manner: *Trichoderma viride* > *Cladosporium herbarum* > *Alternaria alternata*. While *Cladosporium* gave no effect on *Agrobacterium sp.*, the *Alternaria alternata* showed mordant effect and maximum inhibition was recorded by *Trichoderma viride*. Previous studies (Gopalakrishnan et al. 2003, Muhammad, A. et al. 2010) reported inhibition of *R. solani* by *Trichoderma* isolates in dual culture and rated them for strong antagonism. In the present investigation three native isolates have shown maximum growth inhibition of 68, 70, and 72% against phytopathogens in dual culture studies. Hence these native isolates are far more superior in their antagonistic effect against phytopathogens compared to the results obtained by earlier researchers.

Conclusion

Findings of present investigation reveals that significant changes occur in the diversity of important mycoflora. Results also revealed that Knowledge on phenotypic and functional traits of antagonistic fungi will help to determine their fitness for successful bio-fertilization and biological control. This study reveals for the first time the presence of fungi with antagonistic activity against *Sclerotium rofsii*, *Botrytis cinerea* and *Xanthomonas sp.* *Agrobacterium sp.* in Bilaspur district of Chhattisgarh. It also provides essential information to develop broad spectrum biocontrol agent.

TABLE-1: Antagonistic behavior of fungal isolates against phytopathogenic bacteria:

S. No.	Name of fungal isolates /Standard antibiotics	Zone of inhibition in mm	
		<i>Xanthomonas sp.</i>	<i>Agrobacterium sp.</i>
01.	<i>Alternaria alternata</i>	9.5 ± 0.1.1	7.2 ± 1.5
02.	<i>Cladosporium</i>	13.3 ± 6.8	0 ± 0.0
03.	<i>Trichoderma viride</i>	17.8 ± 2.5	9.1 ± 2.3
04.	Gentamycin	29.00 ± 0.55	22.00 ± 0.56
05.	Kenamycin	25.00 ± 0.86	21.00 ± 0.67



Fig.1: Antagonistic behavior of fungal isolates against phytopathogenic bacteria

TABLE-2: Antagonistic behavior of fungal isolates against phytopathogenic fungi

S. No.	Name of fungal isolates/Fungicides	Growth of <i>Sclerotium rofsii</i>				Growth of <i>Botrytis cinerea</i>			
		2 nd day	4 th day	6 th day	% mycelial inhibition	2 nd day	4 th day	6 th day	% mycelial inhibition
01.	<i>Alternaria alternata</i>	0.9	2.3	2.5	70.5 ± 5.3	1.0	2.6	3.2	60.8 ± 1.6
02.	<i>Cladosporium</i>	0.8	1.5	2.6	68.7 ± 3.3	1.1	2.7	3.8	52.5 ± 2.7
03.	<i>Trichoderma viride</i>	1.2	2.0	2.2	72.8 ± 2.8	0.9	2.4	2.4	70.3 ± 2.9
04.	Nystatin (1g/10ml)	-	-	-	100	-	-	-	100
05.	Endosulfan (1g/10ml)	-	-	-	100	-	-	-	100

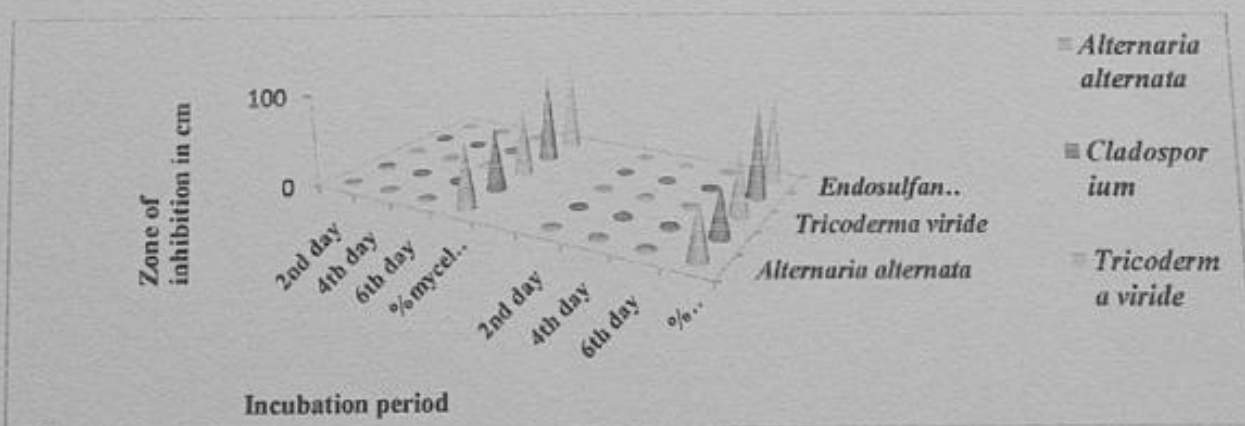


Fig. 2: Antagonistic behavior of fungal isolates against phytopathogenic fungi

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