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Efficacy of Rhizospheric Fungal Bioagent Against Rot of Ginger

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Abstract

The present study deals with the isolation of fungi associated with rhizome rot of ginger and its management with rhizospheric fungal bioagents. The rhizospheric fungal bioagents included 3 isolates each of *Aspergillus flavus* and *Aspergillus niger*, 2 isolates each of *Trichoderma koningii* and *Trichoderma pseudokoningii*, 3 isolates each of *Trichoderma viride* and *Trichoderma harzianum*. On the basis of the characters in culture and morphology, the isolates of these fungi were named separately as strains T₁ to T₁₆. These isolates were evaluated against causal pathogen of ginger rot i.e., *Fusarium* sp., *F. solani* and *F. oxysporum* f. sp. *zingiberis* by dual culture. *T. harzianum* (T₁₆), *T. viride* (T₁₃), *T. pseudokoningii* (T₁₀), *T. koningii* (T₈), *A. niger* (T₆) and *A. flavus* (T₃) significantly proved to be most effective in inhibition of test pathogens.

Key words:- 1. Antagonists, 2. Biocontrol agent (BCA), 3. Dual culture, 4. Rhizospheric soil mycoflora.

Introduction:

Ginger (*Zingiber officinale* Rosc.) is an important commercial crop valued for spice and medicinal value, despite of its high importance the production of ginger is lesser than requirement as it suffers from several diseases (Dohroo and Edison, 1989). Among the diseases of ginger, rot caused by *Pythium* sp. / *Fusarium oxysporum* f. sp. *zingiberis* is more prevalent throughout the world including India and very common in Bundelkhand region (Kaushal, 2001). It is basic need to develop high yielding varieties with better quality to increase the production and productivity of ginger (Ravishanker et al., 2014). Eco friendly management of the disease is necessary, because the inappropriate use of agrochemicals especially fungicides pose more carcinogenic risks than insecticides and herbicides together. (Osman and Abdulrahman, 1999). Biological control is another way to avoid environment pollution to minimize the extensive use of pesticides (Ibrahim, 1997), among the BCAs fungal antagonists are very useful. In Bundelkhand available information on control of rhizome rot disease of ginger by different biocontrol is scanty and the work so far is preliminary and non-conclusive. Under the circumstances present study was conducted to explore an ecofriendly and economic management strategy.

Materials and Methods:

The present investigation was carried out from 2016-2017 in the Department of Botany, Bundelkhand University, Jhansi (U.P).

Survey and Sample Collection:

Diseased sample of rhizome rot of ginger and rhizospheric soil were collected from different villages belonging to six districts of Bundelkhand.

Pathogen:

Six species belonging to two genera *Pythium* and *Fusarium* were found associated with rhizome rot of ginger. Among these *Fusarium oxysporum* f. sp. *zingiberi* was most frequently encountered. Pure culture of 5 days old test pathogen was taken for further studies.

Pathogenicity Test:

Healthy ginger rhizomes were inoculated with the causal pathogen, to confirm pathogenicity. Inoculated rhizomes were incubated for 12, 24, 36, 48 and 72 hours. These rhizomes were sown in pots containing sterilized soil. For each treatment there were three pots, with five plants in each pot, experiment was replicated thrice.

Biocontrol Agents(BCA):

According to soil dilution technique (Johnson et. al.,1995) rhizospheric soil mycoflora was obtained. Sixteen isolates belonging to six species of two genera *Trichoderma* and *Aspergillus* were selected for in-vitro studies.

Dual Culture Technique:

With the help of sterilized cork borer fungal bio agents and test fungus were kept on to petri plates having PDA. Radial growth inhibition was recorded at an interval of 24 hrs. for five days to record the stages of antagonism. The percent inhibition was calculated by the following formula –

$$\text{Percent inhibition} = \frac{\text{Radial diameter (mm) in check} - \text{Radial diameter (mm) in treatment}}{\text{Radial diameter (mm) in check}} \times 100$$

Results:

Six species belonging to two genera were found associated with ginger rhizome rot. Among these *Fusarium oxysporum* f. sp. *Zingiberi* was most frequently encountered. While *Pythium aphanidermatum*, and *Fusarium* sp. were occasionally present (Table 1).

In the pathogenicity test, it was found that rotting and emergence of seedling increased with increase of incubation period. The seedlings obtained after 72 hrs. of inoculation were most severely rotted (+++++) whereas with 12 hours of inoculation rotting was mild (++) , and absent in control (Table 2).

Sixteen fungal isolates were obtained from rhizospheric soil. On the basis of the characters in culture and morphology, the isolates of these fungi were named separately as strains, they included 3 isolates each of *Aspergillus flavus* and *Aspergillus niger*, 2 isolates each of *Trichoderma koningii* and *Trichoderma pseudokoningii*, 3 isolates each of *Trichoderma viride* and *Trichoderma harzianum*. Screening of these isolates for their antagonistic potential against rot pathogens by dual culture revealed that all fungal isolates significantly inhibited the radial growth of test pathogens in comparison to control. Among the rhizospheric bioagents, isolate of *T. harzianum* (T₁₆), *T. viride* (T₁₃), *T. pseudokoningii* (T₁₀), *T. koningii* (T₈), *A. niger* (T₆) and of *A. flavus* (T₃) significantly proved to be most effective in reducing the radial growth of *Fusarium oxysporum* f. sp. *zingiberi*, *F. solani* and *Fusarium* sp. there by resulting in maximum percent inhibition over control (Table 3, Fig.1). The most effective bio-agent against the pathogen was *T. harzianum* followed by *T. viride*. *A. flavus* was least effective. An increasing rate of percent inhibition in growth of *Fusarium oxysporum* f. sp. *zingiberi*, *F. solani* and *Fusarium* sp. in subsequent hour inoculation was visualized. This trend lead to infer that fungal antagonists primarily require a period of time to establish themselves in substantiating their effect on targeted pathogens there by getting involve themselves in one or more of the mechanisms of parasitism to suppress the pathogens.

Table 1: Fungi associated with ginger rhizome, isolated by PDA method.

S. No.	Present in different Area	Phythiumapha nidermatum	Phythium myriotylu m	Phythium sp.	F. oxysporum f. sp. Zingiberi	F. solani	Fusarium sp.
1	Baruasagar	-	-	-	++	+	+
2	Majra	++	-	-	+++	++	++
3	Jugbai	++	+	-	+++	++	+
4	Rasoi	-	-	-	++	+	+
5	Maharajpura	+	-	+	++	+	++
6	Mutra	+	-	-	+++	+	+
7	Panhari	+	-	+	+	+	+
8	Papauni	-	-	-	++	-	++
9	Ber	-	+	+	+++	-	++
10	Simra	-	-	-	+	+	+
11	Januli	+	-	-	+	++	-
12	KareelaBatta	-	-	+	+	+	-

Table 2: Appearance of rot symptoms on plants incubated with test pathogen for different days.

Inoculated and incubated for hours	Typical symptoms appearance	Length of seedling	rotting
12 hrs	13 th days	6.56cm	++
24 hrs	14 th days	6.13cm	++
36 hrs	15 th day	5.80cm	+++
48 hrs	15 th day	5.36cm	++++
72 hrs	15 th day	5.00cm	+++++
Control	12 th day	8.73cm	--
CD (P=0.05)	0.57	0.61	

Table 3: In-vitro antagonistic efficacy of rhizospheric fungi against rot pathogens of ginger by dual culture technique.

Antagonist	Isolate No.	Fusarium oxysporumf. sp. Zingiberi		Fusarium solani		Fusarium sp.	
		Radial Growth *(mm)	Inhibiti on (%)	Radial Growth *(mm)	Inhibiti on (%)	Radial Growth *(mm)	Inhibiti on (%)
A.flavus	T1	32.3	43.8	33.7	42.4	20.9	36.1
	T2	29.3	47.4	30.0	46.8	18.9	39.5
	T3	18.7	60.1	18.7	60.3	18.2	51.8
A.niger	T4	17.7	61.5	18.0	60.9	17.9	52.2
	T5	15.0	64.6	14.0	65.8	14.2	56.5
	T6	13.7	66.2	13.7	66.2	13.9	56.8
T. koningii	T7	13.0	65.0	13.3	66.6	13.5	57.2

	T8	12.7	67.4	12.3	67.8	12.9	58.0
T. pseudokoningii	T9	12.0	68.2	12.0	68.2	12.2	58.8
	T10	11.0	69.4	10.7	69.8	11.2	59.9
T. viride	T11	10.3	70.2	10.0	70.6	10.5	60.9
	T12	09.3	71.4	09.0	71.8	09.5	61.9
	T13	08.3	72.6	08.3	72.4	08.2	63.6
T. harzianum	T14	06.3	74.9	06.3	74.9	06.5	65.5
	T15	04.0	77.7	04.0	77.7	04.2	68.2
	T16	0.3	80.8	02.0	79.9	03.2	69.4
Control		85.0	00	75.0	00	77.9	00
CD at 5%		0.11		2.1		2.2	

Discussion:

Biological significance of rhizosphere fungi is very important and has been used by many workers. It is well documented that when two or more organisms are grown in close proximity, the interaction could be stimulating, inhibiting or antagonistic. In our study the antagonistic effect of rhizospheric fungal isolates on test pathogen has been observed more or less similar findings have been reported by (Eladet al., 1980; Alfansoet al., 1987; Khara and Hadwan, 1989; Ushamalini et al., 1997; Lucicowa, 1990; Melo and Faull, 2000; Tamuli and Boruah, 2002; Prasad et al., 2001). According to Lorito, et. al., 1996; Sanz, et. al., 2004; Seidl, et. al., 2005; Liu and Yang, 2007 and Viterbo, et. al., 2007) *Trichoderma* species are well known for the production of lytic enzymes involved in general antibiosis or specific mycoparasitism. It is known that *Trichoderma* is a hostile mycoparasite, which can control already established pathogens as well as newly entered pathogens (Sharma 2011). May be due to this reason in present investigation *Trichoderma* species were most dominant in inhibition of test pathogen. In support to our results many workers have suggested that the ability of isolates of *Trichoderma p.* and *Aspergillus sp.* to control root rot pathogens (Sivan et al., 1984; Shanmugan and Verma, 1999; Hazarika et al., 2000).

Conclusion:

The fungicidal potential of all the isolates of the bioagents has been in the descending order as *Trichoderma harzianum* > *Trichoderma viride* > *Trichoderma pseudokoningii* > *Trichoderma koningii* > *Aspergillus niger* > *Aspergillus flavus*. The genus *Trichoderma* is responsible for the inhibition of pathogens and can be utilized for the development of rot resistant lines of ginger by applying biotechnological approach.

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