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IN VITRO EVALUATION OF FUNGICIDAL RESPONSES ON THE GROWTH OF PATHOGENIC *RHIZOCTONIA SOLANI* KUHN, ANTAGONISTIC BINUCLEATE *RHIZOCTONIA* AND *TRICHODERMA HARZIANUM* RIFAI

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Abstract

Two pathogenic isolates of *Rhizoctonia solani* Kuhn causing stem canker/black scurf disease of potato plants and four antagonist isolates, two of binucleate *Rhizoctonia* and two of *Trichoderma harzianum* Rifai were isolated from crop fields and evaluated *in vitro* for their fungicidal responses against eight fungicides. Vitavax was effective in inhibiting the growth of *R. solani* and binucleate *Rhizoctonia* but it did not inhibit the growth of *T. harzianum* at 100 ppm concentration. Terraclor Super X, Dithane M 45 and Boric acid are the fungicides which at 100 ppm concentration did not inhibit the growth of antagonist isolates of *T. harzianum* and binucleate *Rhizoctonia* but inhibited the growth of isolates of *R. solani* to some extent. The *in vitro* findings suggest that any one of these three fungicides along with antagonist isolates of binucleate *Rhizoctonia* and *T. harzianum* can be used as biocontrol agents to reduce soil borne inocula of *R. solani*.

Rhizoctonia solani Kuhn (perfect stage: *Thanatephorus cucumeris* (Frank) Donk) occurs world-wide and causes various diseases of agricultural crops. It is the causal organism of stem canker and black scurf disease of potato. Black sclerotia on the surface of the tuber and sunken necrotic lesions on roots, stems and underground parts of potato plants are common symptoms of Rhizoctonia disease (Bandy *et al*. 1984, Carling and Leiner 1986). Some of the isolates of *Rhizoctonia* closely resemble *R. solani* in mycelial characteristics but possess predominantly binucleate hyphal cells. Binucleate *Rhizoctonia* has potential as biocontrol agent for protection of potato from Rhizoctonia canker (Escande and Echandi 1991). *T. harzianum* was found to be an effective biological control agent for protecting a number of crop plants from damage induced by *R. solani* and *Sclerotium rolfsii* Sacc. under both greenhouse and field conditions (Elad *et al.* 1980). *R. solani* is an unspecialized parasite, survive in soil in the absence of host plant. Reduction or elimination of soil borne inocula of a plant pathogen can be achieved by using its antagonist/s as biocontrol agent/s with a suitable fungicide/s. To achieve the above objective the present work was aimed to find out a fungicide/s following *in vitro* evaluation which will have at least some inhibiting effect on the growth of the plant pathogen *R. solani* but having no such effects on its antagonist isolates of binucleate *Rhizoctonia* and *T. harzianum*.

The isolates DK64 and BTB115 of *R. solani* were isolated from potato plant parts having stem canker and black scurf symptoms. Among the isolates, these two isolates of *R. solani* were found to be most virulent against potato. Binucleate *Rhizoctonia* isolates GK206 and CD169 and *T. harzianum* isolates CU1 and W9 were isolated from soils of different crop fields. These isolates of binucleate *Rhizoctonia* and *T. harzianum* have potential antagonistic effects against the virulent *R. solani* isolates DK64 and BTB115 (Khandaker *et al*. 2005). Eight fungicides, namely Bavistin (carbendazim), Topsin M (thiophanate methyl and thiram), Homai (thiophanate methyl), Terraclor

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Super X, Vitavax (carboxin), Dithane M 45 (zinc and meneb), Rovral (iprodione) and Boric acid were used to evaluate the growth inhibition of *R. solani* and above mentioned antagonistic fungi *in vitro*. The fungicides were collected from Bangladesh Agricultural Research Institute, Gazipur.

Four concentrations, *viz.* 0, 1, 10 and 100 ppm in term of active ingredient were used for all fungicides. Requisite amount of fungicide was mixed with PDA medium. Autoclaved medium was poured into 90 mm diameter Petri dishes @ 20 ml per plate. After solidification of the medium, the plates were inoculated with 5 mm diameter mycelial blocks cut from 3-day-old colony of *R. solani* isolates (BTB115 and DK64). Mycelial blocks from cultures of binucleate *Rhizoctonia* (GK206 and CD169) and *T. harzianum* (CU1 and W9) isolates were also transferred to fungicides amended PDA plates. There were four replications for each treatment. The plates were incubated at 25 ± 1ºC for ten days. Data on radial mycelial growth of the colonies was recorded after five days of inoculation. Per cent inhibition of growth was calculated with the following formula (Sundar *et al*. 1995):

$$\% \text{ inhibition of growth} = \frac{(X - Y)}{X} 100$$

where, X = growth of pathogen/antagonist without fungicide and

Y = mycelial growth of the pathogen with fungicide in the medium.

Details of the results on the growth responses of two isolates of plant pathogenic *R. solani* and four antagonist isolates, two of binucleate *Rhizoctonia* and two *T. harzianum* are shown in Tables 1 and 2. The results show that Bavistin effectively restricted the growth of *R. solani*, binucleate *Rhizoctonia* and *T. harzianum* isolates at 10 ppm concentration. The growth of *R. solani* and *T. hazianum* isolates was checked by Homai and Topsin M at 10 ppm concentration but growth of binucleate *Rhizoctonia* isolates was inhibited at 100 ppm concentration. Rovral totally restricted the growth of both pathogen and antagonist fungi isolates at 100 ppm concentration except binucleate *Rhizoctonia* isolate GK206. Vitavax inhibited the growth of *R. solani* and binucleate

Table 1. Growth responses of *R. solani* isolates at different concentrations of fungicides.

Fungicides	Dose (ppm *a.i.*)	% inhibition		Fungicides	Dose (ppm *a.i.*)	% inhibition	
		Isolate DK64	Isolate BTB115			Isolate DK64	Isolate BTB115
Bavistin	0	0	0	Boric acid	0	0	0
	1	92.37	95		1	0	0
	10	100	100		10	0	0
	100	100	100		100	18.45	10.23
Vitavax	0	0	0	Rovral	0	0	0
	1	29.62	29.60		1	55.00	62.90
	10	59.62	58.88		10	82.59	83.33
	100	100	100		100	100	100
Homai	0	0	0	Dithane M-45	0	0	0
	1	40	46.66		1	0	0
	10	100	100		10	0	0
	100	100	100		100	37.09	25.09
Terraclor Super X	0	0	0	Topsin M	0	0	0
	1	0	0		1	17.4	6.29
	10	0	0		10	100	100
	100	10.33	13.33		100	100	100

Table 2. Growth responses of binucleate *Rhizoctonia* and *T. harzianum* at different concentrations of fungicides.

Fungicides	Dose (ppm *a.i.*)	% inhibition				Fungicides	Dose (ppm *a.i.*)	% inhibition			
		Binucleate *Rhizoctonia*		T. harzianum				Binucleate *Rhizoctonia*		*T. harzianum*	
		GK206	CD169	CU1	W9			GK206	CD169	CU1	W9
Bavistin	0	0	0	0	0	Boric acid	0	0	0	0	0
	1	95	93	95	96		1	0	0	0	0
	10	100	100	100	100		10	0	0	0	0
	100	100	100	100	100		100	0	0	0	0
Vitavax	0	0	0	0	0	Rovral	0	0	0	0	0
	1	31.74	30.95	0	0		1	33.33	79.62	35.18	40.73
	10	62.00	62.53	0	0		10	46.29	100	70.33	69.62
	100	100	100	0	0		100	70.00	100	100	100
Homai	0	0	0	0	0	Dithane M-45	0	0	0	0	0
	1	38.18	21.11	90.37	83.33		1	0	0	0	0
	10	100	80.74	100	100		10	0	0	0	0
	100	100	100	100	100		100	0	0	0	0
Terraclor Super X	0	0	0	0	0	Topsin M	0	0	0	0	0
	1	0	0	0	0		1	64.81	4.81	80.73	81.48
	10	0	0	0	0		10	100	61.11	100	100
	100	0	0	0	0		100	100	100	100	100

Rhizoctonia isolates but did not inhibit the growth of *T. harzianum* isolates at 100 ppm concentration. Although Terraclor super X, Boric acid and Dithane M 45 inhibited the growth of *R. solani* to some extent at 100 ppm concentration but these fungicides did not inhibit the growth of binucleate *Rhizoctonia* and *T. harzianum* isolates. The findings suggest that Terraclor super X, Boric acid and Dithane M 45 along with antagonist isolates of binucleate *Rhizoctonia* and *T. harzianum* can be used as biocontrol agents to reduce soil borne inocula of *R. solani*.

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