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In vitro Sensitivity of Trichoderma Isolates to Selected Herbicides

Key words : Herbicides, Sensitivity, *Trichoderma* species

Soil borne plant pathogenic fungi such as *Fusarium, Phytophthora, Pythium, Rhizoctonia, Sclerotium etc.* cause diseases in most of the economically important crop plants. Biological control with antagonist *Trichoderma* offers a novel approach when applied either alone or in combination with other management practices without the demerits of chemical control (Papavizas, 1985 and Mukhopadhyay, 1987). However, modern agriculture involves application of several agrochemicals. Some of them are found toxic to *Trichoderma* and thereby affecting its growth and in turn the biocontrol potential. In order to derive maximum antagonistic potential, it is necessary to know the sensitivity of *Trichoderma* isolate(s) to these agrochemicals and make necessary corrections before incorporating as a component in Integrated Disease Management (Singh *et al*., 1995). In recent times, application of herbicides is increasing to manage weeds. These herbicides, sprayed aerially, reach the soil (by means of air currents or are washed off the plant surface due to rain) and affect the growth and efficacy of *Trichoderma*. Differential tolerance of *Trichoderma* isolates to herbicides was reported earlier by Jayaraj and Radhakrishnan, (2000) and Gupta (2004). The present investigation was conducted to evaluate the sensitivity of two isolates of *Trichoderma* spp. *viz., T. harzianum* and *T. virens* to selected herbicides at field concentration.

Four herbicides, *viz.,* pendimethalin, glyphosate, alachlor and 2,4-D sodium salt were used in the present investigation to assess the *in vitro* sensitivity of *Trichoderma* isolates by using the poisoned food technique (Nene and Thapliyal, 1993) and slide germination technique (Montgomery and Moore, 1938). Isolates of *Trichoderma* available in the Department of Plant pathology, Agricultural College, Bapatla (*T. harzianum* isolated from cotton cropping system of Guntur dt. and *T. virens* isolated from citrus orchards of Guntur dt.) were utilized in the present study. Radial growth or spore germination of the test *Trichoderma* isolates were recorded after 48 h of incubation based on which per cent inhibition of growth or spore germination over control (unamended medium) was calculated.

In herbicide un-amended medium (control plates), *T. harzianum* recorded 5.1 cm diameter 24 h after inoculation and covered the 9 cm. Petri plate in two days. In case of *T. virens*, the growth was 4.6 cm up to 24 h and then covered the entire plate in two days (Table 1). Similarly, spore germination was on par in both the isolates with 100 per cent germination by 48^{th} h of incubation in herbicide un-amended potato dextrose broth.

In herbicide amended medium, all the herbicide showed inhibitory effect either on radial growth or spore germination or both. Variation existed between *Trichoderma* isolates in their sensitivity to different herbicides, between the growth stages of the same test fungus, *i. e.,* assimilative phase (radial growth) and spore phase (spore germination) and among different herbicides in their toxicity to *Trichoderma* isolates.

Variation between isolates of *Trichoderma*: Significant variation was observed in the sensitivity of *Trichoderma* isolates to herbicides. Except in glyphosate (76.9% inhibition) and 2, 4 - D sodium salt (49.8% inhibition) amended plates, in all other treatments the growth was completely inhibited (100%) on both the days of incubation (Table 1). All the herbicides showed significant reduction in the growth of *Trichoderma* isolates on 1^{st} and 2^{nd} day of observation. When mean inhibitory per cent in the radial growth of *Trichoderma* isolates was analyzed, cotton isolate *T. harzianum* was found less sensitive (79.1% inhibition) compared to citrus isolate *T. virens* (84.1%) (Table 2). However, both the isolates were found equally sensitive to herbicides in spore germination without any significant difference between them (Table 3). Relatively high sensitivity of *T. virens* is attributed to minimal use of herbicides in citrus orchards where by *T. virens* lacked the required adoptaion to the xenobiotic. In other words, *T. harzianum* was less sensitive as it was isolated from cotton where in the herbicidal usage is comparatively more.

Table 1. Effect of herbicides on *Trichoderma* growth (diameter in cm).

Herbicides	*T. harzianum*		*T. virens*	
	Day 1	Day 2	Day 1	Day 2
Pendimethalin @ 0.7%	0.0 (1.0)[d]	0.0 (1.0)[d]	0.0 (1.0)[d]	0.0 (1.0)[d]
Glyphosate @ 0.6%	1.1 (1.5)[c]	2.3 (1.8)[c]	1.0 (1.4)[c]	1.8 (1.7)[c]
Alachlor @ 0.6%	0.0 (1.0)[d]	0.0 (1.0)[d]	0.0 (1.0)[d]	0.0 (1.0)[d]
2,4-D sodium salt @ 0.4%	2.5 (1.9)[b]	5.2 (2.5)[b]	2.0 (1.7)[b]	3.9 (2.2)[b]
Check	5.1 (2.5)[a]	9.0 (3.2)[a]	4.6 (2.4)[a]	9.0 (3.2)[a]
CV (%)	2.3	2.3	3.0	1.4
CD (P=0.01)	0.07	0.08	0.08	0.05

*Each treatment replicated five times
*Figures in parenthesis are square root transformed values
* Figures with similar alphabets do not differ significantly

Table 2. Effect of herbicides on *Trichoderma* growth - per cent inhibition

Herbicides	*T.harzianum*	*T.virens*	Mean
Pendimethalin @ 0.7%	100.0 (90.0)	100.0 (90.0)	100.0 (90.0)[a]
Glyphosate @ 0.6%	74.1(59.4)	79.7 (63.2)	76.9 (61.3)[b]
Alachlor @ 0.6%	100.0 (90.0)	100.0 (90.0)	100.0 (90.0)[a]
2,4-D sodium salt @ 0.4%	42.5 (40.7)	57.0 (48.9)	49.8 (44.8)[c]
Mean	79.1 (62.8)[a]	84.1 (66.5)[b]	
CV (%)		1.0	
CD (P=0.01)	Herbicides	Isolate	Interaction
	1.1	0.8	1.6

*Each treatment replicated five times
*Figures in parenthesis are square root transformed values
* Figures with similar alphabets do not differ significantly

Variation between growth stages: Assimilative phase (radial growth) was more sensitive than spore phase. Variation in the effect of herbicides on different growth stages of *Trichoderma* was maximum with penidimethalin, glyphosate and 2,4-D sodium salt which were found to be more toxic to assimilative phase but less toxic to spore phase.

Variation in fungicide toxicity:

Alachlor was toxic to both isolates on both the growth phases, while penidimethalin (100 and 65%), glyphosate (76.9 and 100%) and 2,4D sodium salt (49.8 and 17.6%) showed variation in their toxicity to assimilative and spore phases.

Table 3. Effect of herbicides on *Trichoderma* - per cent inhibition in spore germination.

Herbicides	*T.harzianum*	*T.virens*	Mean
Pendimethalin @ 0.7%	75.3 (60.3)	54.7 (47.7)	65.0 (54.0)[b]
Glyphosate @ 0.6%	100.0 (90.0)	100.0 (90.0)	100.0 (90.0)[a]
Alachlor @ 0.6%	100.0 (90.0)	100.0 (90.0)	100.0 (90.0)[a]
2,4-D sodium salt @ 0.4%	12.9 (21.0)	22.3 (28.2)	17.6 (24.6)[c]
Mean	72.0 (65.3)	69.3 (64.0)	
CV (%)		2.84	
CD (P=0.01)	Herbicides	Isolate	Interaction
	2.1	NS	3.1

*Each treatment replicated five times
*Figures in parenthesis are square root transformed values
* Figures with similar alphabets do not differ significantly

Table 4. Grouping of herbicides based on per cent inhibition in the radial growth and spore germination of *Trichoderma*.

Parameter	Group	Herbicide
100 % inhibition either in growth or in spore germination	Dangerous	Alachlor, pendimethalin, glyphosate
50 to 99% inhibition either in growth or in spore germination	Cautious	2, 4 D sodium salt*
< 50 % inhibition either in growth or in spore germination	Safe	2, 4 D sodium salt*

*found place in cautious with respect to *T. virens* and safe with respect to *T. harzianum*

Similar results of 2,4-D toxicity at 1000 ppm and above to *Trichoderma* was reported by Gupta (2004). Jayaraj and Radhakrishnan (2000) who reported compatibility of 2,4 -D with Trichoderma.

Based on the results obtained, all the test herbicides were grouped in to ‘dangerous’, ‘cautious’ and ‘safe’ and presented in Table 4. Alachlor, pendimethalin and glyphosate were dangerous to *Trichoderma* spp with 100 per cent inhibition of either radial growth or spore germination or both. While only 2, 4-D was found safe for the *T. harzianum* where as with *T. virens* it was in the cautious group.

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