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Efficacy of botanicals, bio-agents and fungicides against *Fusarium Oxysporum F. Sp. Ciceri*, in chickpea wilt sick plot

[1]D.R. Kamdi, *[1]M.K. Mondhe, [2]G. Jadesha, [1]D. N. Kshirsagar, [1]K. D. Thakur

[1]Department of Plant Pathology, College of Agriculture, Nagpur-440001
[2]Department of Plant Pathology, College of Agriculture, Rajendranagar, Hyderabad-500030

ABSTRACT

Chickpea (Cicer arietinum L.) is the most important pulse crop grown all over India. Fusarium oxysporum f. sp. ciceri cause chickpea wilt is one of the major diseases on Chickpea in Vidharbha region, which is soil and seed borne. Heavy inoculum in soil and favorable environment condition results in the death of infected plant and therefore total yield loss. In this study, two antagonists, two fungicides and two botanical extract were studied against Fusarium oxysporum f. sp. ciceri causing Chickpea wilt. Field studies found that Carbendazim seed treatment (@ 2g/ kg seed) gave minimum wilt incidence (26.38%) and maximum yield (13.47 qt/ha) followed by T. viride + Carbendanzim, T. viride + Thiram and Trichoderma viride alone. Bio-agents Bacillus subtilis and aqueous plant extracts of Azadirachta indica and Lantana camara seed treatments were also significant in reducing wilt incidence and more yield as compared to control.

Key words: Chickpea wilt, *Fusarium oxysporum* f. sp. *ciceri*, Fungicides, Bio-agents, Plant extracts

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INTRODUCTION

Fusarium oxysporum f. sp. *ciceri* is a soil and seed borne pathogen colonizing the xylem vessels and blocking them completely to cause wilting. Wilt (*Fusarium oxysporum* f. sp. *ciceri* (Pad.) Snyd and Hans is one of the serious disease of Chickpea, causing heavy loss upto 10-100% depending on fungal inoculum and environmental condition [7] [8]. Soil drenching with fungicides are generally used to control of this disease, however, frequent and indiscriminant use of it leads to ill effects on environment causing soil and water pollution and development of new strain with more virulence, hence Bio-control and Botanicals has been advocated as one of promising alternative strategy to overcome these problems. The present study was conducted to find out effective Bio-agents, Fungicides and Plant extracts for eco-friendly and economical management of Chickpea wilt.

MATERIALS AND METHODS

Isolation and purification of *Fusarium oxysporum* f. sp. *ciceri.*

Chickpea plants showing yellowing, drooping of leaves was collected from Fusarium wilt sick plot, Plant Pathology Research Field, College of Agriculture, Nagpur. The roots were washed wash under tap water to remove soil particles. Disease plant were cut at collar region into small pieces of 5 – 8 cm long and surface sterilized by dipping in 0.1% $HgCl_2$ solution for 1 minutes followed by 3 time wash with sterilized distilled water. Those bit were transfer on PDA media under ascetic condition and inoculated plates were kept at 28^{o}C in the incubator with 12 hr of light and 12 hr darkness for the emergence of pathogenic fungi. The mycelium growth emerging form cut trisection was transferred by hyphal tip method on PDA media for sporulation. Again, these plates were kept in the incubator with earlier mention control condition for next seven days. The morphological character of isolated pathogen for shape and size of macroconidia, microconidia and chlymadospore were examined under calibrated compound microscope.

Test pathogen was identified as *Fusarium oxysporum* f.sp.*ciceri* on basis its microscopic observation. Pathogenicity test was carried on JG-62 in sick pot and Koch's postulates were proved.

Preparation of mass inoculum of *Fusarium oxysporum* f. sp. *ciceri.*

The sorghum grains were soaked partially for one hour in warm water (40 to 45^{0}C) and then spread on the clean blotting paper for air drying. About 300 g moistened grains were filled in each 1000 ml flask with 10 ml water and autoclaved for 30 minute at 15 lbs psi pressure. The mycelium bit of pure culture of *Fusarium oxysporum* f. sp. *ciceri* were inoculated under aseptic condition in those flask containing grains and incubated at 28± 2^{0}C for 10 days. Meanwhile flasks were shaken to avoid clumping of grains and to facilitate early growth of the fungus. The grains turn whitish due to mycelial growth of the test fungus. These mass inoculums were spread in the experimental sick plot before two weeks of sowing.

Preparation of Aqueous Plant Extracts

Freshly leaves of selected botanicals viz. *Azadirachta indica* and *Lantana camara* were cleaned with tap water to remove the dust particle. Aqueous plant extracts was prepared by crushing the leaves in the mortar and pestle with sterile distilled water (1:1, w/v). The macerated extract was filtered through three-folded muslin cloth to remove fibrous and suspended material, this extracts was used as crude aqueous extract for experimental works [1].

Seed treatments

The bio-agents, *Trichoderma viride* and *Bacillus subtilis* were obtained from Department of Plant Pathology, College of Agriculture, Nagpur were used for seeds treatments. Fungicides treatment was given @ 2 gm /Kg of seeds. For plant extracts treatment, seeds were soaked for half an hour in the desired concentration of aqueous plant extracts and untreated seed served as control. The experiment was carried out in three replication and periodic observations for wilting incidence were recorded. The Bio-agent, Fungicides and Botanicals were evaluated *in vivo* by Randomized Block Design and data were statistically analyzed.

Table1: Effect of Bio-agents, aqueous plant extracts and fungicides seed treatment on wilt incidence in sick soil.

Sr. No.	Treatment	Incubation period (days)	Wilt Incidence (%)				% Wilt reduction over control
			30 DAS	45 DAS	60 DAS	90DAS	
1	*Trichoderma viride*	21	7.05 (2.66)**	13.15 (3.63)**	22.11 (28.05)*	28.19 (32.07)*	53.02
2.	*Bacillus subtilis*	21	8.87 (2.94)	14.32 (3.78)	23.67 (29.11)	30.01 (33.22)	49.99
3	*Azadirachata indica*	19	9.68 (3.11)	14.80 (3.85)	24.74 (29.83)	32.66 (34.85)	45.58
4	*Lantana camara*	19	11.82 (3.44)	16.89 (4.11)	29.90 (33.15)	37.17 (37.57)	38.06
5	Carbendanzim	25	6.02 (2.45)	10.90 (3.30)	20.71 (27.07)	26.38 (30.90)	56.04
6	Thiram	23	6.73 (2.59)	13.75 (3.71)	23.07 (28.71)	28.73 (32.41)	52.12
7	*Trichoderma viride* + Carbendanzim	25	6.45 (2.54)	11.94 (3.46)	22.98 (28.64)	27.15 (31.40)	54.76
8	*Trichoderma viride* + Thiram	24	7.26 (2.69)	12.53 (3.54)	23.09 (28.72)	28.05 (31.98)	53.26
9	Control	15	20.40 (4.52)	28.20 (5.31)	41.88 (40.33)	60.01 (50.77)	---

*** Figures in parenthesis are square root transformed values.*
** Figures in parenthesis are arcsine-transformed values.*

'F' test	Sig.	Sig.	Sig.	Sig.
SE (m) ±	0.075	0.095	0.43	0.48
CD (P= 0.05)	0.22	0.29	1.27	1.45

RESULTS AND DISCUSSION

The isolated fungi from disease chickpea plant was identified on the basis of macroconidia which showed 3-4 septation with slight curved at apex and foot shape at based with average size 15.0-8.0x2.5-1.0 um. Microconidia were oval shaped, 0 spectation with average size 4.5-2.0x1.5-1.0 um.

In the current study, it was observed that all the seed treatments were significantly superior over control (untreated seeds). Wilt incidence was appeared in 17th days after sowing in control treatments while maximum incubation period of 25 days was observed in seed treatment with Carbendazin and combined treatment of Carbendenzin plus

Trichoderma viridae . Data presented in Table 1 , revealed Seed treatment with Carbendazim showed minimum wilt incidence (26.38%), followed by *T. viride* + Carbendazim (27.15%), *T. viride* + Thiram (28.05%) and *T. viride* (28.19%) on 90 DAS and showed reduced wilting percent over control. Similar results were reported by [8] [9].

Data presented in Table 2, it was reported that all the seed treatments gave significantly increased yield per plot over control. Seed treatment with Carbendazim gave more seed yield (13.47 q/ha) followed by *T. viride* + Carbendazim (13.19 q/ha), *T. viride* + Thiram (12.78 q/ha) and *T. viride* (12.50 q/ha), where as control gave (7.22 q/ha) yield. Similar results were found by [2] [3].

Table2: Effect of seed treatments (Bio-agents, Aqueous plant extracts and Fungicides) on Grain Yield (qt/ha)

Sr. No.	Treatment	Grain yield per plot (g)			Mean Grain Yield (g)	Grain Yield (qt/ha)	Increase Grain Yield over control (%)
		RI	RII	RIII			
1.	*Trichoderma viride*	485	440	425	450	12.50	73.13
2.	*Bacillus subtilis*	395	420	430	415	11.53	59.70
3.	*Azadirachata indica*	410	370	375	385	10.69	48.06
4.	*Lantana camara*	345	390	375	370	10.28	42.24
5.	Carbendanzin	450	505	500	485	13.47	86.57
6.	Thiram	445	450	425	440	12.22	69.25
7.	*Trichoderma viride* + Carbendanzim	490	465	470	475	13.19	82.69
8.	*Trichoderma viride* + Thiram	475	465	440	460	12.78	77.01
9.	Control	250	245	285	260	7.22	---
‘F’ test							Sig.
SE (m) ±							0.37
CD (P= 0.05)							1.10

In the present investigation, it was observed that seed treatment with Carbendazim was found significantly superior for increasing seed germination, reducing wilt incidence and increase yield. Thiram also showed significant effect. Among the Bio-agents, seed treatment with *Trichoderma viride* was found effective followed by *Bacillus subtilis*. Amongst aqueous leaf extracts *Azadirachta indica* was found effective followed by *Lantana camara* at 5% concentration.

CONCLUSION

Fusarium oxysporum f. sp. *ciceri* causal agent of chickpea wilt, is one of the major diseases of chickpea in India, results in huge economic losses. As the disease is seed borne and persist for long years in soil, management of disease by single approaches will be difficult and uneconomical. The results of present finding will help to tackle the disease by integration of bioagents, botanicals and fungicides which is efficient and ecofriendly.

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