Journal of Biological Control, 29(3): 162-166, 2015

Research Article

Efficacy of bioagents alone and in combination microbial population against the wilt incidence of cumin

J. R. TALAVIYA* and K. B. JADEJA
Polytechnic in Agriculture, Junagadh Agricultural University, 365640, Dhari, Gujarat, India.
**Corresponding author E-mail*: jrtalaviya@gmail.com

ABSTRACT: Cumin (*Cuminum cyminum L.*) is an annual herb of the family Apiaceae (Umbelliferae) and an important spice crop and its production is limited due to wilt disease. So, for management of cumin wilt and to gain higher seed yield broadcasting application of *Trichoderma harzianum* or *Trichoderma viride* or *Pseudomonas fluorescens* either alone or in combination @ 5kg talc based product/ ha (carrier FYM 500 kg/ha) should be given at the time of sowing and one month after sowing (carrier soil 100 kg/ha).As the population of *Trichoderma* spp. and *Pseudomonas* spp. increases the population of *Fusarium* spp. decreases and the wilt incidence also decreases.

KEY WORDS: Cumin, *Fusarium oxysporum*, *Trichoderma* spp., *Pseudomonas fluorescens*

(Article chronicle: Received: 21-09-2015; Revised: 26-09-2015; Accepted: 26-09-2015)

INTRODUCTION

Cumin (*Cuminum cyminum L.*) is an annual herb of the family Apiaceae (Umbelliferae) and an important spice crop. India is the leading producer (70% of world production), exporter and consumer of cumin in the world. Gujarat state shares 59% of Indian production and Surendranagar is the leading district (Anon, 2012). Because of its low water requirements, farmers are interested in the cultivation of cumin in drought affected areas, where most of the other crop plants cannot be grown economically. However, production of cumin is limited due to several biotic stresses, of which, wilt disease is the most serious (Valizadeh *et al.,* 2007; Agrawal, 1996). The pathogen is soil borne and difficult to eradicate, as fungal chlamydospores survive in soil up to 6 years even in the absence of the host plant (Haware *et al*, 1996). This disease has been reported as a limiting factor in cumin production worldwide including Argentina (Gaetan and Madia, 1993), Egypt (Arafa, 1985), Greece (Pappas and Elena, 1997) and India (Champawat and Pathak, 1990; Jadeja and Nandoliya, 2008). Hence, present investigation was carried out to testing bioagents and their consortium for the wilt management under field condition.

MATERIALS AND METHODS

A field experiment was laid out to study the efficacy of consortium of bioagents against cumin wilt. The experiment was carried out during *Rabi* 2012-13 and 2013-14 at the Plant Pathology Department Farm, JAU, Junagadh in Randomized Block Design with eight treatments (Table 1) along with three replications. The gross and net plot sizes were 5 x 2.5 m and 4 x 2.0 m, respectively. Cumin variety Gujarat cumin-4 was used in the experiment. All the experimental plots were artificially inoculated with 10 day old culture of *Fusarium oxysporum*f. sp. *cumini* prepared on sorghum grain two week prior to sowing @ 250g/plot.

The observation on wilted plants were recorded weekly onward from initiation. After each observation wilted plants were uprooted and destroyed. The disease incidence was calculated using following formula.

$$\text{Per cent wilt incidence} = \frac{\text{Total no. wilted plants recorded through}}{?} \times 100$$

Quantification of microbial population

To quantify the density of total fungi, total bacteria, *Fusarium* spp., *Trichoderma* spp. and *Pseudomonas* spp. from different treatments the soil samples were assessed. Soil samples (1g/plot) were collected from each replication at three times during crop season: initial, 45 DAS and at harvest. Similarly at 45 day crop age soil samples were also collected from rhizoplane of wilted plant for each replication.

All soil samples were stored in refrigerator. The population of microbes in each soil sample was determined on

selective media by dilution technique as mentioned by Benson (2002).The selective media *viz.* Potato Dextrose Agar (PDA) for total fungi (Atlas, 2010), Nutrient Agar (NA) for total bacteria (Atlas, 2010). Komada for *Fusarium* (Komada, 1975), Rose Bengal Agar for *Trichoderma* (Elad and Chet, 1983), and King's B medium for *Pseudomonas* (Atlas, 2010) were used.

Table 1. Efficacy of consortium of bioagents against cumin wilt

Treatment	Treatment Details
T1	Soil application of *Trichoderma harzianum* @ 5 kg in 500 kg of FYM/ha at the time of sowing and soil application of *T. harzianum* @ 5 kg/ha in 100 kg soil one month after germination
T2	Soil application of *Trichoderma viride* @ 5 kg in 500 kg of FYM/ha at the time of sowing and soil application of *T. viride* @ 5 kg/ha in 100 kg soil one month after germination
T3	Soil application of *Pseudomonas fluorescens* @ 5 kg in 500 kg of FYM/ha at the time of sowing and soil application of *P. fluorescens* @ 5 kg/ha in 100 kg soil one month after germination
T4	Soil application of *Trichoderma harzianum* @ 2.5 kg + *T. viride* @ 2.5 kg in 500 kg of FYM/ ha at the time of sowing and soil application of *T. harzianum* @ 2.5 kg + *T. viride* @ 2.5 kg /ha in 100 kg soil one month after germination
T5	Soil application of *Trichoderma harzianum* @ 2.5 kg + *P. fluorescens* @ 2.5 kg in 500 kg of FYM/ ha at the time of sowing and soil application of *T. harzianum* @ 2.5 kg + *P. fluorescens* @ 2.5 kg/ha in 100 kg soil one month after germination
T6	Soil application of *T. viride* @ 2.5 kg + *P. fluorescens* @ 2.5 kg in 500 kg of FYM/ha at the time of sowing and soil application of *T. viride* @ 2.5 kg + *P. fluorescens* @ 2.5 kg ha in 100 kg soil one month after germination
T7	Soil application of consortium *T. harzianum* + *T. viride* + *P. fluorescens* @ 5 kg in 500 kg of FYM/ ha at the time of sowing and soil application of *T. harzianum* + *T. viride* + *P. fluorescens* @ 5 kg/ha in 100 kg soil one month after germination
T8	Control

Note: Talc based bioagents with 107/g CFU strength.

Yield

The seed yield was recorded in kg at the time of harvest. The per cent increase in yield in each treatment over control was worked out by the following formula:

$$\text{Yield increase (\%)} = \frac{\text{Yield in treatment (kg) - Yield in control (kg)}}{\text{Yield in control (kg)}} \times 100$$

RESULT S AND DISCUSSION

Fusarial population was reduced up to harvest in the treatments of combined application of bioagents *viz*. *T. harzianum* + *T. viride*, *T. harzianum* + *P. fluorescens, T. viride* + *P. fluorescens* and *T. harzianum* + *T. viride* + *P. fluorescens*. Among these highest reduction of fusarial population was observed in combined application of *T. harzianum*+*T. viride* + *P. fluorescens*. In this treatment the rhizosphere reduction after 45 days of sowing and at harvest was 61.02 per cent and 66.95 per cent respectively, while reduction in fusarial population in rhizoplane was 45.34 per cent after 45 days of sowing. Highest increase in population of *Fusarium* was recorded in control at both the observation times (Table 2 and 3).

Trichoderma population increased as against previous counts in all treatments in both observation time. Higher population was observed in treatments of combined application of bio agents i.e.*T. harzianum* + *T. viride* (61.0 x 10^3 g^{-1}soil), and *T. harzianum* + *T. viride* + *P. fluorescens* (70.8 x 10^3 g^{-1}soil) (Table 2 and 3).

In case of *Pseudomonas* population it was observed that higher increase in *Pseudomonas* population was recorded in *Pseudomons* amended four treatments. i.e. *Pseudomonas* alone (*P. fluorescens*), *T. harzianum* + *P. fluorescens,T. viride* + *P. fluorescens* and *T. harzianum* + *T. viride* + *P. fluorescens.* The population increased up to the harvest in these treatments. Although there was little increase in remaining treatments, the rate of increase without and with *Pseudomonas* was in the range of 2.04 per cent to 24.58 per cent in rhizosphere after 45 days of sowing, 5.44 per cent to 29.24 per cent in rhizoplane after 45 days of sowing and 11.68 per cent to 41.18 per cent at harvest, respectively (Table 2 and 3).

Total fungal population increased in all the treatments up to the harvest and as the crop growth stage advanced. The total fungal population at harvest ranged between 67.8 x 10^3 g^{-1} soil and 77.4 x 10^3 g^{-1} soil as against 43.2 x 10^3 g^{-1} soil to 60.1 x 10^3 g^{-1} soil in initial stage, respectively. Population was higher in rhizoplane in comparison to rhizosphere. Total bacteria population increased in all the treatments and highest population observed at harvest. Total bacterial population was increased up to the harvest. Population was little higher in rhizoplane as compared to rhizosphere (Table 2 and 3).

Looking to the pooled data of two years lowest disease incidence was recorded in the treatment of *T. harzianum*

Table 2. Assessment of soil microbial population in cumin grown field under bioagent consortium treatments (mean of two years)

Sr No.	Initial (After inculation at time of sowing)					After 45 DAS										At Harvest (Rhizosphere)				
						Rhizosphere					Rhizoplane									
	*Fusarium**	*Trichoderma**	*Pseudomonas***	Total fungi*	Total Bacteria**	*Fusarium**	*Trichoderma**	*Pseudomonas***	Total fungi*	Total Bacteria**	*Fusarium**	*Trichoderma**	*Pseudomonas***	Total fungi*	Total Bacteria**	*Fusarium**	*Trichoderma**	*Pseudomonas***	Total fungi*	Total Bacteria**
1	23.3	23.4	13.7	47.7	24.7	27.3	31.7	14.3	65.3	28.3	29.3	33.6	14.8	74.0	30.6	33.8	36.0	15.3	78.8	31.6
2	26.6	25.6	11.6	53.1	24.3	31.2	32.4	12.3	65.1	24.6	34.1	33.1	12.9	67.3	25.6	36.4	37.5	14.2	69.8	27.5
3	29.4	12.6	32.3	44.1	37.7	35.6	13.9	38.6	58.7	47.9	37.4	14.6	41.5	66.4	49.3	41.4	16.3	45.6	67.8	50.0
4	25.7	30.3	14.7	60.1	32.9	15.5	41.6	15.0	73.2	34.4	17.9	48.5	15.5	76.0	34.9	18.4	61.0	16.8	77.5	37.3
5	28.7	19.3	23.4	47.9	31.6	24.4	28.4	28.6	60.3	33.4	29.0	29.4	29.6	69.0	35.2	21.0	53.0	31.7	78.5	36.0
6	30.0	21.5	31.6	49.8	39.4	29.5	30.9	36.9	62.4	41.7	34.4	33.6	38.6	65.8	42.8	22.7	45.0	40.4	72.8	49.0
7	23.6	24.6	30.1	51.2	40.0	9.20	39.4	37.5	62.3	41.9	12.9	40.1	38.9	65.9	42.5	7.8	70.8	41.5	77.4	44.8
8	26.4	14.4	13.4	43.2	28.1	40.0	15.9	14.4	56.9	30.1	44.4	17.3	15.2	61.1	33.8	50.6	17.3	16.4	70.6	39.6

* CFU (103 g-1 soil)
** CFU (108 /g-1 soil)

Table 3. Percent increase or decrease in microbial population as compare to initial stage in cumin grown field under bioagent consortium treatments (mean of two years)

Sr No.	After 45 DAS*										At Harvest* (Rhizosphere)				
	Rhizosphere					Rhizoplane									
	Fusarium	*Trichoderma*	*Pseudomonas*	Total fungi	Total Bacteria	*Fusarium*	*Trichoderma*	*Pseudomonas*	Total fungi	Total Bacteria	*Fusarium*	*Trichoderma*	*Pseudomonas*	Total fungi	Total Bacteria
1	17.17	35.47	04.38	36.90	14.57	25.75	43.59	08.03	55.14	23.89	45.06	053.85	11.68	65.20	27.94
2	17.29	26.56	06.03	22.60	01.23	28.20	29.30	11.21	26.74	05.35	36.84	046.48	22.41	31.45	13.17
3	21.09	10.32	19.50	33.11	27.06	27.21	15.87	28.48	50.57	30.77	40.82	029.37	41.18	53.74	32.63
4	-39.69	37.29	02.04	21.80	04.56	-30.35	60.07	05.44	26.46	06.08	-28.40	101.32	14.29	28.95	13.37
5	-14.98	47.15	22.22	25.89	05.70	01.05	52.33	26.50	44.05	11.39	-26.83	174.61	35.47	63.88	13.92
6	-01.67	43.72	16.77	25.30	05.84	14.67	56.28	22.15	32.13	08.63	-24.33	109.30	27.85	46.18	24.37
7	-61.02	60.16	24.58	21.68	04.75	-45.34	63.01	29.24	28.71	06.25	-66.95	187.80	37.87	51.17	12.00
8	51.52	10.42	07.46	31.71	07.12	68.18	20.14	13.43	41.44	20.28	91.67	020.14	22.39	63.43	40.93

*per cent values

+ *T. viride* + *P. fluorescens* (9.13 %) with 63.39 per cent disease controlled over check and remained significantly superior than all treatments. This was followed by application of *T. harzianum* + *T. viride* (15.02 %), *T. harzianum* + *P. fluorescens* (16.60 %), *T. viride* + *P. fluorescens* (17.93 %), *T. harzianum* (17.16 %), *T. viride* (18.04 %) and *P. fluorescens* (19.84 %) and were at par. Highest disease incidence was recorded in control (24.94 %). Highest seed yield of 893 kg/ha was also recorded in *T. harzianum* + *T. viride* + *P. fluorescens* which was 52.38 per cent higher over control and remained significantly superior to all treatments. This was followed by *T. viride* + *P. fluorescens* (736 kg/ha), *T. harzianum* (687 kg/ha), *T. harzianum*+*P. fluorescens* (685 kg/ha), *T. viride* (677 kg/ha), *T. harzianum* + *T. viride* (666 kg/ha) and *P. fluorescens* (629 kg/ha)and were at par. Lowest seed yield was recorded in control (425 kg/ha) (Table 4).

It is concluded from the microbial population study from field trial that as the population of *Trichoderma* spp. and *Pseudomonas* spp. increase the population of *Fusarium* spp. decrease and the wilt incidence also decreases. Similar observations were recorded by Vyas and Mathur (2002) regarding distribution of *Trichoderma* spp. in cumin rhizo-

Table 4. Efficacy of bioagents and their consortium against cumin wilt

Treatments	2012-13		2013-14		Pooled				Percent Disease control	Yield increased over control (%)
	Disease Incidence (%)	Seed yield (kg/ha)	Disease Incidence (%)	Seed yield (kg/ha)	Disease Incidence (%)		Seed yield (kg/ha)			
T 1	26.97 (20.29)	712	22.16 (14.23)	662	24.47 (17.16)		687		31.19	38.04
T 2	27.74 (21.67)	704	22.52 (14.68)	650	25.13 (18.04)		677		27.67	37.14
T 3	28.55 (22.83)	658	24.35 (17.00)	600	26.44 (19.84)		629		20.45	32.35
T 4	24.44 (17.12)	708	21.17 (13.04)	625	22.80 (15.02)		666		39.78	36.16
T 5	25.05 (17.92)	716	23.04 (15.31)	654	24.04 (16.60)		685		33.44	37.90
T 6	26.65 (20.12)	765	23.46 (15.84)	708	25.05 (17.93)		736		28.11	42.23
T 7	16.86 (08.42)	925	18.32 (09.88)	862	17.58 (09.13)		893		63.39	52.38
T 8	31.83 (27.81)	454	28.09 (22.16)	397	29.95 (24.94)		425		-	-
					T	Y x T	T	Y x T		
SEm ±	01.63	048.37	1.21	55.87	01.02	1.43	036.96	52.26		
C.D. at P = 5%	04.95	146.00	3.67	169.00	02.94	NS	107.00	NS		
C.V. %	10.88	011.86	9.15	15.02	10.18		013.41			

*The figures in the parentheses are retransformed values

sphere and their effect on wilt in farmer's field after 60 days of sowing. They found that higher soil population of *Trichoderma* spp. reduced fusarial count and wilt incidence.

Similar mechanisms are also reported in chickpea. Khan *et al.* (2004) reported low population of *Fusarium* spp. with lower chick pea wilt incidence under higher population of *Trichoderama* spp./*Pseudomonas fluorescens*. Efficacy of *Trichodema* spp. and fluorscent *Pseudomonas* has been reported against fusarial wilt of other crops also. Field application of *Trichoderma* spp. and *P. fluorescens* has been proved effective for the control of pegion pea wilt (Somasekhara *et al.* 1996) and banana wilt (Saravanan *et al.* 2003).

Combined application of *Trichoderma harzianum* + *T. viride* + *Pseudomonas fluorescens* was found most effective in controlling disease and also had highest seed yield. It is recommended from this field trial that for the management of cumin wilt and to gain higher seed yield broadcasting application of *T. harzianum* or *T. viride* or *P. fluorescens* either alone or in combination @ 5 kg talc based product/ha should be given at the time of sowing (carrier FYM 500 kg/ha) and one month after sowing (carrier soil 100 kg/ha).

REFERENCES

Agrawal S. 1996. Volatile oil constituents and wilt resistance in Cumin (*Cuminum cyminum* L.). *Curr Sci.* **71** (3): 177–178.

Anonymous. 2012. District wise estimates of area, production and productivity of cumin. Directorate of Agriculture, Gandhinagar.

Arafa MKM. 1985. Studies on Fusarium wilt of cumin. M.Sc. Thesis, (Unpublished) Assiut University, Egypt.

Atlas RM. 2010. *Handbook of Microbilogical media*. 4th Edition. CRS Press. 2043pp.

Benoson HJ. 2002. *Microbial Application.* 8th Edition. McGraw Hill. 87pp.

Champawat RS, Pathak VN. 1989. Cultural, morphological and pathogenic variations in *Fusarium oxysporum* f. sp. *cumini*. *Indian J Mycol Pl Pathol.* **19**: 178–183.

Elad Y, Chet I. 1983. Improved selective media for isolation of *Trichoderma* spp. and *Fusarium* spp. *Phytoparasitica* **11** (1): 55–58.

Gaetan S, Madia M. (1993). The presence of cumin (*Cuminum cyminum*) wilt caused by *Fusariumoxysporum* f. sp. *cumini* Patel, Prasad, Mathur, Mathur in Argentina. Boletin-de-Sanidad-Vegetal. *Plagas* **19** (3): 503–507

Haware MP, Nene YL, Natarajan M. 1996. Survival of *Fusarium oxysporum* f. sp. *ciceri*in soil in the absence of chickpea. *Phytopath Mediter.* **35**: 9–12.

Jadeja KB, Nandoliya DM. 2008. Integrated management of wilt of cumin (*Cuminum cyminum* L.). *J Spices Arom Crops* **17** (3): 223–229.

Khan MR, Khan SM, Mohiddin FA. 2004. Biological control of Fusarium wilt of chickpea through seed treatment with the commercial formulation of *Trichoderma harzianum* and/or *Pseudomonas fluorescens. Phytopathol Mediterr*. **43**: 20–25.

Komada H. 1975. Development of a selective medium for quantitativeisolation of *Fusarium oxysporum* from natural soil. *Pl Prot Res.* **8**: 115–125.

Pappas AC, Elena K. 1997. Occurrence of *Fusarium oxysporum* f, sp. c*umini* in the island of Chos, Greece. *J Phytopathol.* **145**: 271–272.

Saravanan T, Muthusamy M, Marimuthu T. 2003. Development of integrated approach to manage the fusarial wilt of banana. *Crop Prot.* **22**: 1117–1123.

Somasekhara YM, Anilkumar TB, Siddarad AH. 1996. Biocontrol of pigeonpea wilt *Fusarium udum*. *Mysore J Agric.* **30**: 159–163.

Valizadeh M, Kazemi SKT, Nematzadeh GA. 2007. Effect of Plant Growth Regulators on Callus Induction and Regeneration of Cumin (*Cuminum cyminum*). *Asian J Agri Res.* **1**: 17–22.

Vyas RK, Mathur K. 2002. Distribution of *Trichoderma* species in cumin rhizosphere and their potential in supression of wilt. *Indian Phytopathol.* **55** (4): 451–457.