

Original Research Article

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Management of Fusarium Wilt of Castor (*Ricinus communis* L.) Caused by *Fusarium oxysporum* f. sp. *ricini* with Antagonist, Botanical Extract and Pot Experiment

B. Vahunia^{1*}, P. Singh², N.Y. Patel¹ and A. Rathava¹

¹Department of Plant Pathology, N. M. College of Agriculture, Navsari Agricultural University, Navsari, Gujarat, India

²Department of Plant Pathology, Aspee Shakilam Agri Biotechnology Institute, Nau, Navsari, Gujarat, India

*Corresponding author

ABSTRACT

Keywords

Castor wilt, *Fusarium oxysporum* f. sp. *ricini*, Antagonist, Botanical extract, *in vitro*.

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The bioefficacy of antagonist and botanical extract were tested *in vitro* against *Fusarium oxysporum* f. sp. *ricini* causing wilt of castor. *Trichoderma harzianum* (72.22%), *T. viride* (70.37%) and *Pseudomonas fluorescens* appeared potent antagonist against *Fusarium oxysporum* f. sp. *ricini* followed by *T. faciculatum*, *T. longibrachyatum* and *Bacillus subtilis*. Among the botanical, the extract of turmeric rhizome was effective in inhibiting mycelial growth 42.22 Per cent followed by 36.67 per cent inhibition by leaves extract of marigold. Pot culture experiment conducted at Department of Plant Pathology, College of Agriculture, Navsari. Antagonists and botanical extract which were found promising in *in vitro* study were tested in pot culture experiment. Result revealed that seed treatment with *Trichoderma harzianum* found superior in control of *Fusarium oxysporum* f. sp. *ricini* causing wilt of castor.

Introduction

Castor (*Ricinus communis* L.) is one of the most important non-edible oilseed crops of arid and semi-arid regions of India. It belongs to genus *Ricinus* of *Euphorbiaceae* family and its common name is castor bean, other common names include castor-oil plant and palmachristi. In Gujarat (India), castor seed is grown over an area of 7.34 lakh hectares with the production of 13.95 lakh tones (Anonymous, 2015). It is attacked by several pathogens, viz., fungi, bacteria, viruses, nematodes and phytoplasmas. The important

fungal diseases are Phytophthora seedling blight, Rust, Alternaria leaf spot, Cercospora leaf spot, Anthracnose, Powdery Mildew, Stem rot, and Fusarium wilt. Important bacterial diseases are Bacterial leaf spot, Bacterial wilt or Slime disease and viral diseases are like Tobacco ring spot, Tobacco necrosis and cucumber mosaic virus. Among these, the wilt is one of the major constraints and known to cause heavy losses due to occurrence of frequent epidemic in intensive castor growing area of Gujarat. Therefore,

effort were made to find out the cheap, effective, eco-friendly, botanical and bioagents by testing *in vitro* against *Fusarium oxysporum* f. sp. *ricini* from these effective can be later on tested in pot condition.

Materials and Methods

Experimental location

This work was conducted in Department of Plant Pathology, N. M. College of Agriculture, Navsari agricultural University, Navsari during 2015-16. Determine the antifungal activity of *Trichoderma viride*, *T. harzianum*, *T. longibrachyatum*, *T. koningii*, *T. fasciculatum*, *Pseudomonas fluorescens* and *Bacillus subtilis* against *Fusarium oxysporum* f. sp. *ricini* by dual culture technique (Skidmore and Dickinson, 1976).

Evaluation of antagonistic potentiality of bioagent *in vitro*

The test organisms and pathogen were grown separately on PDA. From seven day old culture, 4 mm discs of the test organisms and *Fusarium oxysporum* f. sp. *ricini* were cut aseptically from the periphery of the colony and placed opposite to each other approximately 60 mm apart on to PDA contained in the Petri plates as adopted by Dennis and Webster (1971). To determine the antagonistic action of isolates of bacterial species *viz.*, *Bacillus subtilis* and *Pseudomonas fluorescence* against *Fusarium oxysporum* f. sp. *Ricini in vitro*. The bacterial antagonists were then streaked on one end of the Petri plate onto PDA medium 24 hrs. Prior to *Fusarium oxysporum* f. sp. *ricini* inoculation and just opposite to bacterial streak a 4 mm diameter disc of *Fusarium oxysporum* f. sp. *ricini* from seven days old culture was placed. Three repetitions of each treatment were kept and the Petri plates with 4 mm diameter discs of 7 days old culture of the pathogen on PDA served as control. The

Petri plates were incubated at $27 \pm 2^{\circ}\text{C}$ temperature and after seven days of incubation, radial growth of the test organism and the *Fusarium oxysporum* f. sp. *ricini* was measured. Per cent growth inhibition of the fungus in each treatment in comparison to control was calculated by the following equation (Bliss, 1934):

$$\text{PGI} = \frac{\text{C} - \text{T}}{\text{C}} \times 100$$

Where,

PGI = Per cent growth inhibition

C = Colony diameter in control (mm)

T = Colony diameter in treatment (mm)

Efficacy of botanicals against *Fusarium oxysporum* f. sp. *ricini*

Extract of plants tested against castor wilt pathogen *Fusarium oxysporum* f. sp. *ricini* were, leaves of *Bougainvillea spectabilis* L., *Datura stramonium* L., *Ocimum sanctum* L., *Hibiscus bombycideron*, *Tegateserecta.*, rhizome of *Zingiberofficinale* Rosc., *Curcuma longa* L. (Table 2). Solvents i.e. distilled water was employed for extraction from plant parts and results were derived based on comparative toxicity value. Different parts of plants including leaf and rhizome tested were first washed with sterile distilled water, then sterilized with 90 per cent methanol and then air-dried. Weighed 100 g plant material was crushed in electrically operated mixer and grinder using 1:1 w/v amount of distilled water (Singh and Majumdar, 2001).

The material was homogenized for 5 minutes and filtered through double layered muslin cloth and filtrate was centrifuged at 5000 rpm for 15 minutes and clear supernatant was collected. This was considered as 100 per cent concentration and used for experiment at various percentages.

For evaluation of antifungal activities of the extract, desired concentrations (5%) were obtained by adding appropriate amount of standard solution of plant extracts to 100 ml potato dextrose agar medium in conical flasks. Then about 20 ml extract mixed PDA was poured in sterilized Petri plates. After, solidification of PDA, plates was inoculated with 4 mm diameter discs of seven days old culture of *Fusarium oxysporum* f. sp. *ricini* raised on PDA medium. Each treatment was repeated three times. PDA inoculated with 4 mm fungal discs without plant extract served as control. The inoculated plates were incubated at $27 \pm 2^\circ\text{C}$ for 7 days and diameter of colony was measured.

The per cent growth inhibition of the fungus in each treatment in comparison with control was calculated by the equation given by Bliss, 1934 as mentioned earlier.

Management of wilt under pot condition

The antagonists and botanicals which found promising *in vitro* study were tested for the control of castor wilt in pots as seed treatment.

The inoculum of *Fusarium oxysporum* f. sp. *ricini* was multiplied on sand maize meal medium for 20 days at $27 \pm 2^\circ\text{C}$ and thoroughly mixed with the sterilized soil in proportion of 1:10 w/w, and then filled in the sterilized pots. The pots were watered and kept for a week for uniform spread of the pathogen. The formulation of each two promising bioagent@ 6g/kg seed and botanicals was prepared. The seeds of castor variety were treated with the botanicals and bioagent@ 6g/kg seed formulation. Seeds sown without treatment in the inoculated pots served as control.

Treated seeds of castor hybrid GCH-4 were sown in each pot. Untreated seeds were sown

in inoculated pots served as control. Three repetitions were maintained for each treatment. They were uniformly irrigated soon after seed sowing. Observation on wilt incidence was recorded periodically up to 30 days after sowing and data were statistically analysed.

Result and Discussion

In vitro* testing of antagonists against *Fusarium oxysporum* f. sp. *ricini

The result presented in table 1 and figure 1 revealed that, *Trichoderma harzianum* (72.22%), *Trichoderma viride* (70.37%), *Pseudomonas fluroscens* (67.78%), *Trichoderma longibrachytum* (64.44%) and *Trichoderma koningi* (62.96%) significantly inhibited the pathogen.

Whereas, *Trichoderma faciculatum* (55.19%) and *Bacillus subtilis* (45.19%) were comparatively least effective.

Our result are in harmony with earlier workers Meghwal *et al.*, (2014) found that *T. harzianum* and *T. viride* significantly inhibited the mycelial growth of *Fusarium oxysporum* f. sp. *ricini* (68-72%) by Dual culture technique. Patil *et al.*, (2015) carried out interaction study of known antagonist by dual culture method and found strong antagonistic effect on *Fusarium oxysporum* f. sp. *ciceri* with *T. viride*, *T. koningii*, *G. virens*, *T. pseudokoningii* and *Pseudomonas fluroscens* *in vitro*.

In vitro* testing of botanicals against *Fusarium oxysporum* f. sp. *ricini

The result presented in table 2 revealed that, turmeric rhizome extract (*Curcuma longa* L.) 42.22 per cent showed maximum growth inhibition of the pathogen followed by marigold leaves (*Tegetes erecta* L.) 36.67 per

cent, Tulsi leaves (*Ocimum sanctum* L.) 34.44 per cent, Bougainvillea leaves (*Bougainvillea spectabilis* L.) 31.85 per cent, Jasud leaves (*Hibiscus bombycideron* L.) 30.74 per cent,

Ginger (*Zingiber officinalis* Rosa.) 25.93 per cent and Datura (*Datura stramonium* L.) 18.89 per cent in sterilized plant extract (Fig. 2).

Table.1 Efficacy of native bio-agent against *Fusarium oxysporum* f. sp. *ricini* in vitro

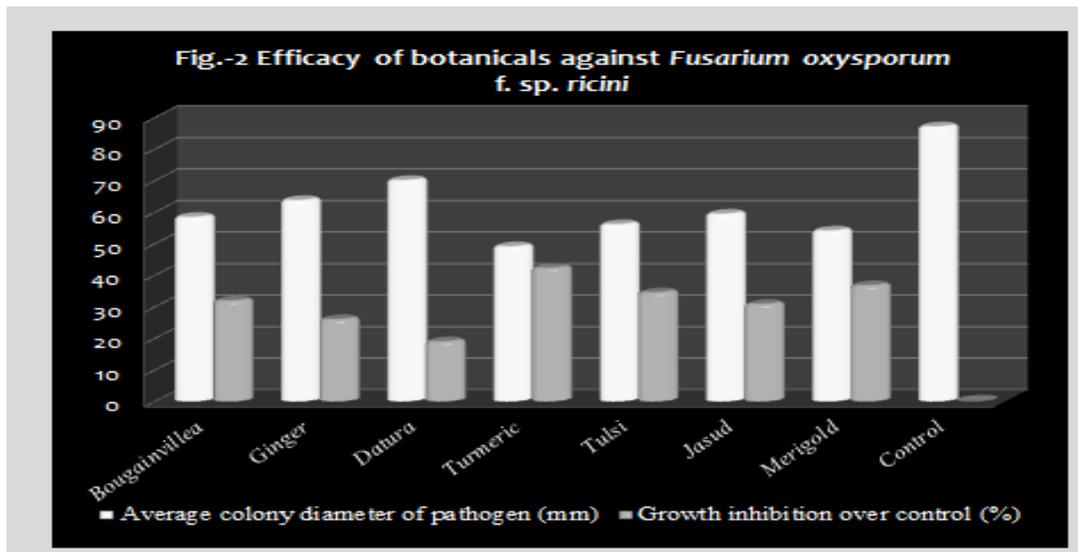
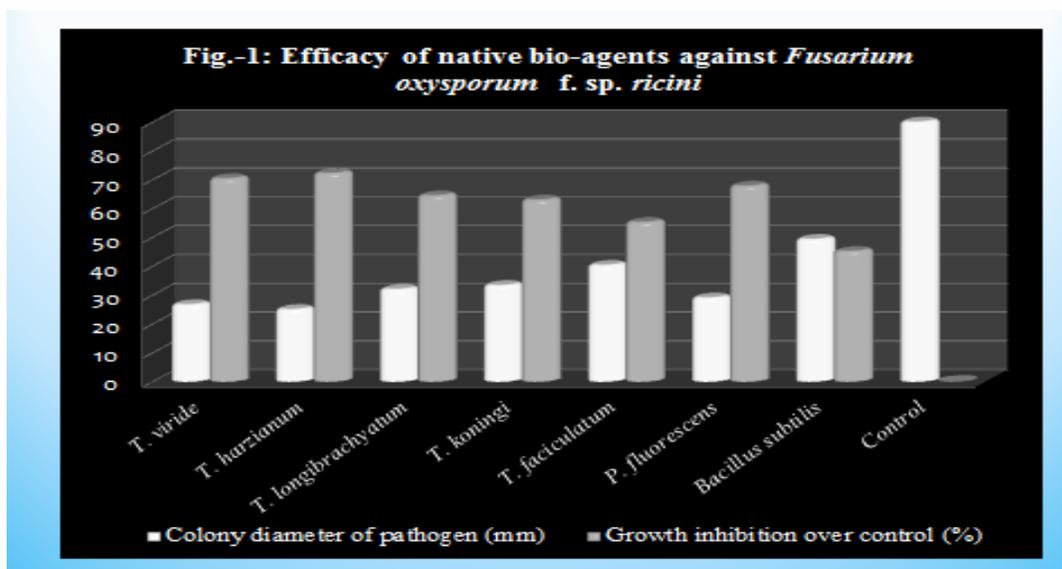
Sr. No.	Test organism	Colony diameter of pathogen (mm)	Growth inhibition over control (%)
1	<i>Trichoderma viride</i> Navsari isolate	26.67	70.37
2	<i>Trichoderma harzianum</i> Navsari isolate	25.00	72.22
3	<i>Trichoderma longibrachyatum</i> Navsari isolate	32.00	64.44
4	<i>Trichoderma koningi</i> Navsari isolate	33.33	62.96
5	<i>Trichoderma faciculatum</i> Navsari isolate	40.33	55.19
6	<i>Pseudomonas fluorescens</i> Navsari isolate	29.00	67.78
7	<i>Bacillus subtilis</i> Navsari isolate	49.33	45.19
8	Control	90.00	0.00
	S.Em.±	0.99	
	C.D. at 5%	2.96	
	C.V.%	4.20	

Table.2 Efficacy of botanicals against *Fusarium oxysporum* f. sp. *ricini* in vitro

Sr. No.	Name of plant	Botanicals name (5%)	Plant parts used for preparation of extract	Average colony diameter of pathogen (mm)	Growth inhibition over control (%)
1	Bougainvillea	<i>Bougainvillea spectabilis</i> L.	Leaves	58.33	31.85
2	Ginger	<i>Zingiber officinalis</i> L.	Rhizome	63.67	25.92
3	Datura	<i>Datura stramonium</i> L.	Leaves	70.00	18.89
4	Turmeric	<i>Curcuma longa</i> L.	Rhizome	49.00	42.22
5	Tulsi	<i>Ocimum sanctum</i> L.	Leavs	56.00	34.44
6	Jasud	<i>Hibiscus bombycideron</i>	Leaves	59.33	30.74
7	Marigold	<i>Tegetes erecta</i>	Leaves	54.00	36.67
8	Control	87.00	0.00
	S.Em.±			1.32	
	C.D. at 5%			3.97	
	C.V.%			3.69	

Table.3 Management of wilt under pot condition

Sr. No.	Treatment	Average no. of plant infected	Percent disease incidence	Percent disease reduction over control
1	<i>Trichoderma viride</i> Navsari isolate	2.67	26.67	62.92
2	<i>Trichoderma harzianum</i> Navsari isolate	2.33	23.33	70.83
3	Turmeric	3.33	33.33	58.33
4	Marigold	3.67	36.67	54.17
5	Control	8.00	80.00	0.00
	S.Em.±	0.21		
	C.D. at 5%	0.69		
	C.V.%	9.13		



The present studies are in confirmation with those described by earlier worker Chohan and Perveen (2015) studied the effect of botanicals on *Fusarium oxysporum* f. sp. *lycopersici* and reported that out of rhizomes extracts tested, Turmeric (*Curcuma longa* L.) extract was found most inhibitory to *Fusarium oxysporum* f. sp. *lycopersici*.

Singh *et al.*, (2014) reported fungitoxic properties of Turmeric, Datura, Ginger, Tulsi and marigold and against growth and of *Fusarium oxysporum* f. sp. *lini*. Shalini *et al.*, (2015) tested botanicals, among them turmeric

extracts (5%) gave 68.05 per cent and ginger extract (5%) gave 45.92 per cent inhibition of *Fusarium oxysporum* f. sp. *ricini*.

Pot experiment

The data presented in table 3 revealed that the effects of all the treatments were found significantly superior over control in managing the wilt disease of castor.

Considering disease incident in seed treatment (*Trichoderma harzianum* @ 6g/kg seed) has lowest (23.33%) disease incidence followed by

(*Trichoderma viride* @ 6g/kg seed) (26.67%), turmeric extract (33.33%) and marigold extract (36.67%) disease incidence.

The similar results to our present investigation were achieved by Magar *et al.*, (2014) reported that coconut cake, groundnut cake, *Trichoderma viride* and *Trichoderma harzianum* were reduced wilt of chick pea by (43.41%), (31.67%), (65.77%) and (65.77%) respectively.

Meghwal *et al.*, (2014) conducted to assess soil amendments and bioagent. *Trichoderma harzianum* and *T. viride* found most effective in reducing the disease of castor wilt caused by *Fusarium oxysporum* f. sp. *ricini*.

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