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Research Article

COMPATIBILITY OF FLUORESCENT PSEUDOMONADS ISOLATES WITH INSECTICIDES

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ABSTRACT
Fusarium wilt caused by *Fusarium oxysporum* f. sp. *ricini* Nanda and Prasad is a very serious disease in castor (*Ricinus communis* L.) growing areas of the state but particularly in north Gujarat. The pathogen is soil-borne and application of fungicides is very expensive and also polluting the ecosystem. Several strains of fluorescent pseudomonads isolates have been reported to suppress soil-borne diseases caused by fungal pathogens. Fifteen fluorescent pseudomonads isolates were obtained on King's B medium from the rhizosphere and rhizoplane of plant roots. A compatibility of isolates tested with insecticides at 250, 500 and 750 ppm concentrations except isolate FP-X. The growth of isolate FP-X was found to be checked at 750 ppm concentration of endosulphan, monocrotophos, triazophos and imidacloprid and also at 500 ppm concentration of endosulphan.

Key Words: *Isolation, Compatibility, Insecticides and Fluorescent Pseudomonads Isolates*

INTRODUCTION
This has lead to the search for alternate strategies for the management of plant pathogens. Biological control of plant pathogens is gaining importance these days. Its usefulness has been demonstrated for the management of soil borne diseases where other strategies have not met with much success. The microorganism isolated from the root or rhizosphere of a specific crop may be better adapted to that crop and may provide better control of diseases than organisms originally isolated from other plant species. Such plant associated microorganisms may make better Bio-control agents because they are already closely associated with and adapted to the plant or plant part as well as the particular environmental conditions in which they must function. The screening of such locally adapted strains has yielded improved Bio-control in some cases (Cook, 1993). The sensitivity of the bio-control active bacterial isolates to insecticides has important implication to integrate the strategies for management of a disease caused by soil borne plant pathogen as compared to fungicides and insecticides. Nicholson and Hirsch (1998) reported the effect of pesticides on the diversity of culturable soil bacteria. Results revealed the population from the pesticides treated plot showed a higher inoculum of heterotrophic bacteria. Shanthi *et al.,* (2003) reported that combined application of carbofuran 1 g and neem cake 250 g and Pseudomonas fluorescens 4 g/tree was effective in reducing nematode population in papaya.

MATERIALS AND METHODS
Collection of Soil and Plant Samples
Fifty soil and roots samples were collected from established castor field plots of different locations of Patan and Banaskantha districts where the castor is commonly grown. Healthy plants of castor (*Ricinus communis* L.) of 60-75 days growth were carefully uprooted along with adhering soil and was carried to the laboratory in polythene bags. The soil particles loosely adhering to the roots were gently teased out and used for isolation of rhizosphere bacteria. Soil particles adhering tightly to the roots were allowed to go with the roots for isolation of rhizoplane bacteria.

Isolation of Fluorescent Pseudomonads Isolates
Excess of soil adhering with roots was removed by gentle shaking. From each sample 10 g of closely associated rhizosphere was added to 250 ml flask containing 90 ml sterilized distilled water. For isolation of rhizoplane bacteria, roots were cut into approximately 2-3 cm long pieces and 10 g of root bits were

then transferred to 90 ml sterilized distilled water. The flasks were placed on a rotary shaker for 1 hr to allow root associated bacteria to diffuse. Three replications were kept for each location and serial dilution of rhizosphere and rhizoplane samples were made up to 10^6. An aliquot of 0.1 ml from 10^6 dilution of each sample was spread plated over solidified King's medium B (Protease peptone No. 3 20.00 g, Dipotassium hydrogen phosphate 1.50 g, Magnesium sulphate $7H_2O$ 1.50 g, Agar 20.00 g, Glycerol 15.00 ml and Distilled water 1 lit.), selective medium on which preferentially fluorescent pseudomonads recovered under aseptic conditions. The plates were incubated at $30^o \pm 1^oC$ for 24-48 hrs. Colonies of different morphology were examined for their fluorescence under ultraviolet light (240-340 nm). The colony showing fluorescence was picked-up and was further purified by streaking on same medium plates. The purified cultures were finally transferred onto solid King's B medium and preserved at low temperature (4^oC) in refrigerator in the Department of Plant Pathology, C. P. College of Agriculture, Sardarkrushinagar Dantiwada Agricultural University, Sardarkrushinagar, for further activities.

Compatibility of Fluorescent Pseudomonads isolates with Insecticides

Commonly and widely applicable insecticides for the seed coating, foliar spray and soil drenching to manage the various plant diseases were examined. Compatibility of fluorescent pseudomonads isolates evaluated by using poison food technique (Grover and Moore, 1962). Insecticides was tested at the concentration of 250, 500, 750 and 1000, 2000, 3000 ppm, respectively (Table 1). Required quantity of the fungicides, insecticides and herbicides solution was added aseptically in to 100 ml flask containing sterilized 20 ml of King's B medium, just before pouring the plates. A loopful of 24 hours old culture of bacterial isolates inoculated centrally after solidification of these King's medium B plates. Three replications were kept for each treatment and plates were incubated at $30^o \pm 1^oC$ for 48 hours and finally growth of bacterial isolates recorded.

RESULTS AND DISCUSSION

Isolation of Fluorescent Pseudomonads Isolates from Rhizosphere and Rhizoplane

Fifteen fluorescent bacterial isolates were obtained on selective medium viz., King's B medium from the rhizosphere and rhizoplane of castor by dilution plating method (106 cfu ml–1) after incubation period of 24-48 hours at 30o ± 1oC and examined the fluorescence under ultraviolet light (200-340 nm). These isolates were designated as FP-I, FP-II, FP-III, FP-IV, FP-V, FP-VI, FP-VII, FP-VIII, FP-IX, FP-X, FP-XI, FP-XII, FP-XIII, FP-XIV and FP-XV. Out of 20 samples collected from ten villages of Patan district, nine fluorescent pseudomonads isolates (FP-I to FP-IX) were obtained, whereas six isolates (FP-X to FP-XV) were gained from 30 samples from seven villages of Banaskantha district. These results are in accordance with the methodology adopted by Vidhyasekaran and Muthamilan (1995), Gupta *et al.*, (2000), Yeole and Dube (2001), Gholve and Kurundkar (2004), Samanta and Dutta (2004) and Sen *et al.*, (2006).

Table 1: Insecticides tested for compatibility with fluorescent pseudomonads isolates *in vitro*

Sr. No.	Trade name	Common name	Chemical name
1.	Endosulphan (35 EC)	endocel	6, 7, 8, 9, 10, 11-Hexachloro-1,1,5,5a,6,9,9a-Hexahydro-8,9-methano-2,4,3-benzodixathiepin-3-exide
2.	Monocrotophos (36 EC)	monocrotophos	Dimethyl 1-methyl-3(methyl amino)-3-oxo-1-propenyl phosphate
3.	Hostathion (40 EC)	triazophos	O,o-diethyl O-Cl-Phenyl-1,H-1,2,4-triazole 3YL Phosphorothioate
4.	Tafban (20 EC)	chlorpyriphos	O,o,-dimethyl o-(3,5,6-trichloro-2-pyridyl phosphorothiate
5.	Tatamida (72 WS)	imidacloprid	1-[C6-chloro-3-pyridinyl-methyl]-N-nitro-2-imidazolidinimine

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Compatibility of Fluorescent Pseudomonads Isolates with Insecticides

Insecticides endosulphan and chlorpyriphos are recommended as seed treatment and soil application for the management of termite under North Gujarat conditions. Whereas, imidacloprid as seed treatment is advocated for sucking pest management. Therefore, these insecticides along with common insecticides monocrotophos and triazophos were evaluated at 250, 500 and 750 ppm concentrations by poison food technique.

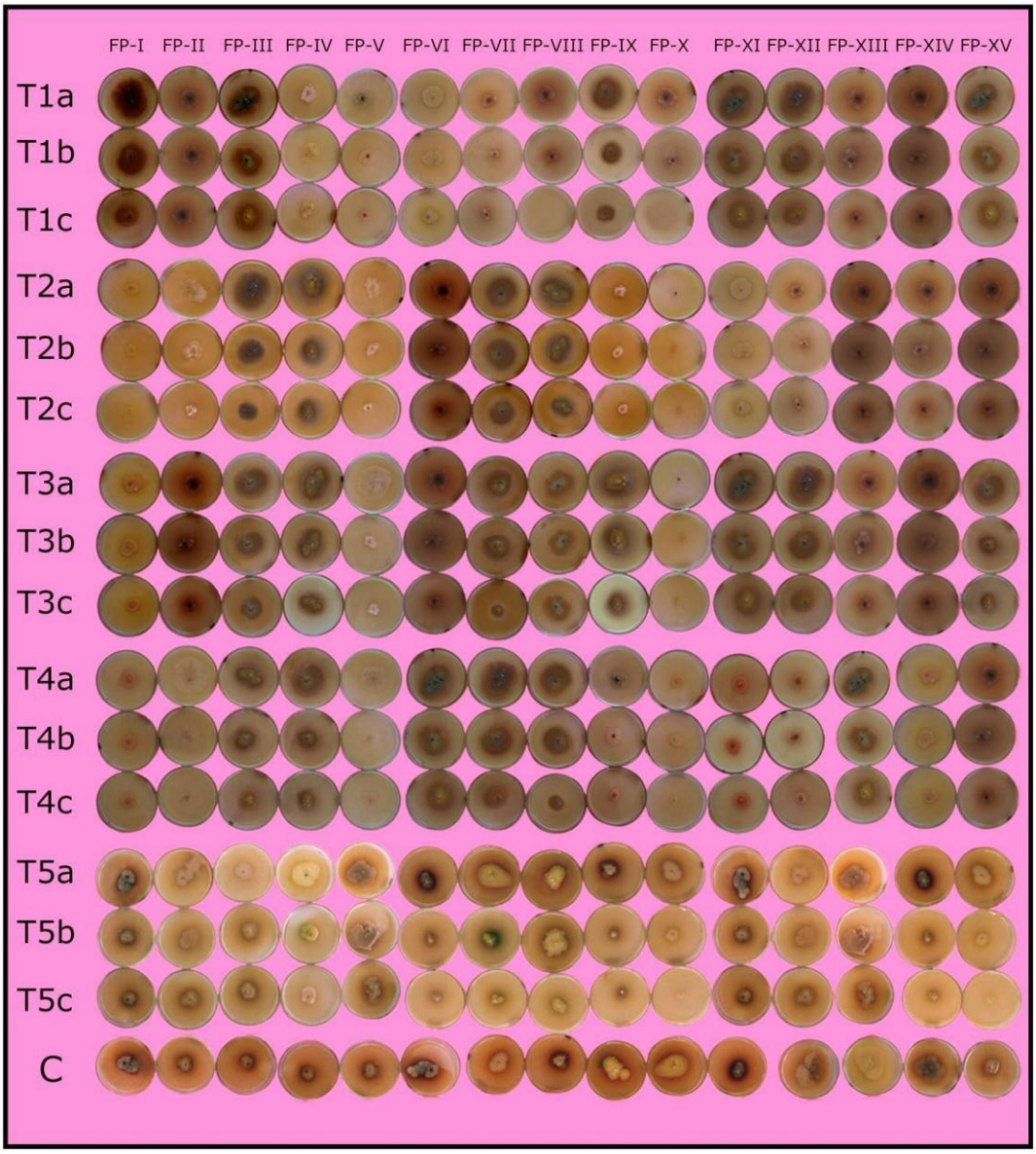

Figure 1: Compatibility of Fluorescent *Pseudomonads* isolates with various insecticides

T1: Endosulphan **a: 250 ppm**
T2:Monocrotophos **b: 500 ppm**
T3: Triazophos **c: 750 ppm**
T4: Chlorpyriphos
T5: Imidaclorprid
C: Control

Table 2: Compatibility of fluorescent pseudomonads isolates with various insecticides

Sr. No.	Character	ISOLATES														
		FP-I	FP-II	FP-III	FP-IV	FP-V	FP-VI	FP-VII	FP-VIII	FP-IX	FP-X	FP-XI	FP-XII	FP-XIII	FP-XIV	FP-XV
1.	**Endosulphan**															
	250 ppm	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
	500 ppm	+	+	+	+	+	+	+	+	+	-	+	+	+	+	+
	750 ppm	+	+	+	+	+	+	+	+	+	-	+	+	+	+	+
2.	**Monocrotophos**															
	250 ppm	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
	500 ppm	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
	750 ppm	+	+	+	+	+	+	+	+	+	-	+	+	+	+	+
3.	**Triazophos**															
	250 ppm	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
	500 ppm	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
	750 ppm	+	+	+	+	+	+	+	+	+	-	+	+	+	+	+
4.	**Chlorpyriphos**															
	250 ppm	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
	500 ppm	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
	750 ppm	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
5.	**Imidacloprid**															
	250 ppm	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
	500 ppm	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+
	750 ppm	+	+	+	+	+	+	+	+	+	-	+	+	+	+	+
6.	Control	+	+	+	+	+	+	+	+	+	+	+	+	+	+	+

+ = Growth ; – = No Growth

The results (Table 2; Plate 1) revealed that growth of all the isolates was observed in all the tested insecticide at 250, 500 and 750 ppm concentrations except isolate FP-X. The growth of isolate FP-X was found to be checked at 750 ppm concentration of endosulphan, monocrotophos, triazophos and imidacloprid and also at 500 ppm concentration of endosulphan.

Conclusion

Insecticides endosulphan, monocrotophos, chlorpyriphos, triazophos and imidacloprid were evaluated for compatibility. Growth of all the isolates was observed in all the tested insecticide at 250, 500 and 750 ppm concentrations except isolate FP-X. The growth of isolate FP-X was found to be checked at 750 ppm concentration of endosulphan, monocrotophos, triazophos and imidacloprid and also at 500 ppm concentration of endosulphan.

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