

COMPATABILITY OF PHOSPHATE SOLUBILIZING MICROORGANISMS WITH DIFFERENT AGROCHEMICALS

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Abstract

Present investigation is subjected to Study the Compatability Studies of Isolated Phosphate Solubilising Microorganisims with different Agrochemicals. Total twenty one isolates of Phoshphate Solubilising Microorganisms were isolated from different medicinal and aromatic plants from Nagarjuna Medicinal Garden, Dr. P. D. K. V, Akola, Maharastra, India. These isolates were tested for compatability with different agro chemicals like fungicides *i.e.* thiram (0.3%), carbendazim (0.1%), mancozeb 0.25% and propiconazole (0.05%) showed 100 per cent growth inhibition of *A. niger* strain *i.e.* (*Aspergillus niger*-5 and *Aspergillus niger*-7), whereas COC+ streptocycline inhibited 38.64 and 41.76 per cent growth inhibition of *A. niger* strains. Among herbicides, quizalfop-ethyl (0.2%) and fenoxaprop-ethyl (0.2%) showed 100 per cent growth inhibition, while glyphosate (0.6%) showed 52.60 and 61.04 per cent growth inhibition, respectively. Insecticides did not show any detrimental effect on growth of *Aspergillus niger*.

Key words : Phoshphate solubilising microorganisms, Aspergillus niger, agrochemicals.

Introduction

Pesticide application is still the most effective and accepted means for the protection of plants from pest (Bolognesi, 2003). The application of pesticides starts from the pre sowing stage. Different treatments include soil application, seed treatment, foliar spray, etc. Repeated applications of pesticides contaminate the soil. Soil is the most important site of biological interactions. But the extensive use of pesticides over the past four decades has resulted in tribulations caused by the interaction with natural biological system (Ayansina and Oso, 2006). Fungicide seed treatment is frequently used to improve early plant emergence and to control the early attack by the pests.

The present study was designed to evaluate the adverse effect of Agro chemicals, on growth, survival of Phosphate solubilising micro organisms under laboratory conditions using various toxicity experiments. This leads to reducing PSM count in soil rhizosphere The main aim of this study was to screen out pesticides tolerant strains of Phosphate Solubilizing Microorganisms for the improvement of biofertilizer for sustainable agriculture. In conventional agricultural practices, the increased use of agrochemicals including pesticides has however, led to the frequent and deliberate contamination of cultivated soils. These chemicals in turn may adversely affect the rhizospheric organisms including PGPR efficient strain of PSM which are governed by the rate of application, the activity spectrum of the pesticides and the persistence and availability of chemicals.

Materials and Methods

In present study, attempts were made to isolate various Phosphate Solubilizing Microorganisms from rhizosphere of Seven Medicinal plants Kalmegh, Sadabahar, Sarpagandha, Safed musli, Shatavari, Muskdana, Ashwagandha and four Aromatic Plants-Citronella, Lemon grass, Rosha grass, Senna. The soil samples were collected from Nagarjuna medicinal plant garden Dr. P.D.K.V., Akola for studying their 'P' solubilizing efficiency.

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S no	Fungicide & antibiotic	Concentration	A.nige	r-5	A.niger-7		
5.10			Colony diameter in (mm)	Percent growth inhibition %	Colony diameter in (mm)	Percent growth inhibition %	
T ₁	Streptocycline	100ppm	90	0	90	0	
T ₂	Coc+Streptocycline	0.25%+100ppm	55.33	38.64	41.76	53.6	
T ₃	Propiconazole	0.05%	0.00	100	0.00	100	
T ₄	Dithane M-45	0.25%	0.00	100	0.00	100	
T ₅	Carbendazim 50WP	0.1%	0.00	100	0.00	100	
T ₆	Thiram 75 WP	0.3%	0.00	100	0.00	100	
T ₇	Control		90		90		
	F-test		SIG		SIG		
	S.E. (m)±		0.08		0.27		
	C.D. (p = 0.01)	-	0.34		1.16		

Table 1 : Compatibility of A. niger with fungicide and antibiotics.

A.niger-5-Aspergillus niger -5

- 1. Compatibility of PSM with different agrochemicals *viz.*, fungicides, herbicide, antibiotics and insecticide (Fathia *et al.*, 2006)
- (a) Compatibility of bacteria by Paper Disc method

PSB isolates were studied for their resistance towards different agrochemicals (fungicides, herbicides, antibiotics, insecticides) through disc diffusion method. Stock solutions of test fungicide, herbicides, antibiotics, insecticides were prepared by adding various concentrations of commercial formulation of fungicides, herbicides, antibiotics and insecticides in distilled water and filter sterilized using 0.45 µm filter papers. Filter paper discs (5 mm diameter) were prepared by adding three different concentrations of these stock solutions to evaluate the concentrations equivalent to, above and below the recommended dose of test fungicides, herbicides, antibiotics and insecticides. 1 ml of individual bacterial culture $(10^7 \text{ cells/ml harvested at early})$ logarithmic growth) was spread on Pikovskaya's agar plates with sterile glass spreader aseptically. Autoclave sterilized 5mm disc of Whatman's filter paper were soaked in each concentration of 5 minutes. These discs were then kept on sterilized blotter paper to drain out the excess water from the disc. These discs were placed at equidistance on the agar surface in triplicate. The plates were incubated for 2-3 days at $28 \pm 2^{\circ}$ C and the diameter of the inhibition zones was measured in mm.

(b) Compatibility of phosphate solubilising fungi by poisoned food technique (Rathod *et al.*, 2010)

Potato dextrose agar medium was prepared, equally distributed measuring 100ml in 250 ml conical flask and

A.niger-7-Aspergillus niger-7

sterilized in autoclave. Requisite quantity of each of the fungicide was added in sterilized melted (45°C) PDA separately so as to obtain desired concentration. Flask containing poisoned medium was shake well to have even and uniform distribution of fungicide. About 20ml of melted poisoned PDA was poured in each sterilized Petri plate and allowed to solidify. These Petri plates were inoculated by test fungus separately. Six mm disc of one week old fungus culture was cut with a sterilized cork borer, lifted and transferred aseptically in the centre of a Petri plate containing the medium poisoned with test fungicide. The control plates were kept where the culture disc were grown in same condition on PDA without fungicide. Treated plates were incubated at room temperature $(27\pm2^{\circ}C)$ for a period of seven days. Colony diameter was recorded in mm and per cent of mycelial inhibition was calculated as per formula given below based on the average of colony diameter. The data of mycelial growth was also subjected to statistical analysis and conclusions were drawn.

Results and Discussion

- 1. Compatibility of phosphate solubilising microorganism with agrochemicals (fungicide, antibiotics, herbicide and insecticide)
- (a) Compatibility of phosphate solubilising fungi with different agro chemicals

Compatibility of phosphate solubilizing fungi were tested *in vitro* by commonly used fungicides *viz.*, thiram, carbendazim, mancozeb and propiconazole and antibiotic *viz.*, streptocycline and in combination with COC+ streptocycline by adopting "Poisoned Food Technique"

S no	Fungicide & antibiotic	Concentrations	A.ni	ger-5	A.niger- 7		
5.10.	rungelie & antibiote	Concentrations	Colony diameter in (mm)	Percent growth inhibition %	Colony diameter in (mm)	Percent growth inhibition %	
T ₁	Quizalfop- p-ethyle 5 EC	0.2%	0.00	100	0	100	
T ₂	Emamectin benzoate 5 SG	0.04%	90	0	90	0	
T ₃	Cypermethrin	0.03%	90	0	90	0	
T ₄	Fenoxaprop-p-ethyle 9 EC	0.2%	0.00	100	0	100	
T ₅	Imidacloprid 17.8 S	0.3%	0.00	100	0	100	
T ₆	Glyphosate	0.6%	42.66	52.60	35.06	61.04	
T ₇	Control		90		90		
	F-test		SIG		SIG		
	SE(M)		0.12		0.48		
	CD		0.49		2.01		

Table 2 : Compatibility of PSF Aspergillus niger with herbicides and insecticides.

A.niger-5-Aspergillus niger -5

A.niger-7-Aspergillus niger-7

Table 3: Compatibility of PSB isolates with different concentration of fungicides by paper disc method (m

S. no.	Isolates	Thiram		Mancozeb		Carbendazim			Propiconazole				
1	PSB-1	0.00	6.00	9.33	0.00	7.00	8.00	0.00	0.00	0.00	0.00	0.00	0.00
2	PSB-2	0.00	6.00	8.00	0.00	6.33	7.33	0.00	0.00	0.00	0.00	0.00	0.00
3	PSB-3	0.00	6.66	8.00	0.00	6.33	7.66	0.00	0.00	0.00	0.00	0.00	0.00
4	PSB-4	0.00	6.67	9.33	0.00	7.33	8.33	0.00	0.00	0.00	0.00	0.00	0.00
5	PSB-5	0.00	6.33	8.66	0.00	7.00	8.00	0.00	0.00	0.00	0.00	0.00	0.00
6	PSB-6	0.00	6.67	9.00	0.00	7.66	8.33	0.00	0.00	0.00	0.00	0.00	0.00
7	PSB-7	0.00	6.63	8.66	0.00	7.33	8.66	0.00	0.00	0.00	0.00	0.00	0.00
8	PSB-8	0.00	6.00	9.00	0.00	7.66	8.66	0.00	0.00	0.00	0.00	0.00	0.00
9	PSB-9	0.00	6.00	8.33	0.00	7.00	8.00	0.00	0.00	0.00	0.00	0.00	0.00
10	PSB-10	0.00	6.66	8.00	0.00	6.33	7.33	0.00	0.00	0.00	0.00	0.00	0.00
11	Standard culture	0.00	6.67	9.33	0.00	7.33	7.66	0.00	0.00	0.00	0.00	0.00	0.00
	Average SD	-	avg 6.99 ±29	avg 8.66 ±0.45	-	avg 7.02 ±0.49	avg 8.0 ±0.45	-	-	-	-	-	-

given by Rathod et al. (2010).

The data revealed that 100 per cent growth inhibition of *Aspergillus niger*-5 and *A. niger*-7 was recorded in thiram (0.3%), carbendazim (0.1%), mancozeb (0.25%) and propiconazole (0.05%). All tested fungicides with different concentration showed negative effect on growth of *A. niger*-5 and *A. niger*-7. In case of COC (0.25%) + streptocycline (100 ppm) inhibited growth of *A. niger*-5 and *A. niger*-7 *i.e.* 38.64 per cent and 53.6 per cent respectively, whereas streptocycline (100 ppm) alone did not show any detrimental effect on growth of both *A.* *niger* isolates. From the result, it is concluded that all the fungicides were not compatible with tested organisms, however COC + streptocycline and streptocycline were compatible with *A. niger* isolates.

(b) Compatibility of PSF *Aspergillus niger* with herbicides and insecticides

In vitro effect of herbicides and insecticides on growth of *Aspergillus niger* isolates was tested. The herbicides *viz.*, Glyphosate (0.6%), quizalfop-ethyl (0.2%) and fenoxaprop-ethyl (0.2%) and insecticides *viz.*,

cypermethrin (0.03%), emamectin benzoate (0.04%) and Imidacloprid 0.03% were used to test their compatibility against *A. niger*-20 and *A. niger*-5 stains.

The data showed quizalfop-ethyl (0.2%), fenoxapropethyl (0.2%) and imidacloprid (0.3%) showed 100 per cent growth inhibition of *A. niger*-5 and *A. niger*-7 followed by glyphosate *i.e.* 52.6 and 61.04 per cent growth of *A. niger*-5 and *A. niger*-7 isolates respectively, It concluded that quizalfop-ethyl, fenoxaprop-ethyl and imidacloprid with tested concentration were not compatible with both the strains of *A. niger*. However glyphosate and tested insecticides were compatible with both the isolates of *A. niger*.

2. Compatibility of phosphate solubilizing bacteria with agrochemicals (fungicides, antibiotics, herbicides and insecticides) by paper disc method (mm)

Compatibility of fungicides with phosphate solubilising bacteria

All the isolates tolerant to 0.15% and 0.12% concentration of thiram and mancozeb, respectively. At 0.3% concentration of thiram, maximum growth showed by PSB-4, PSB-6 (6.67 mm inhibition zone) followed by PSB-10 and PSB-3 (6.66 mm), whereas minimum growth was recorded in PSB-1, PSB-2, PSB-8 and PSB-9 (6.00 mm). At one and half than recommended dose (0.45%)

maximum growth was observed in PSB-11(Standard culture) and PSB-9 (9.33 mm inhibition zone) followed by PSB-4 (9.33 mm) PSB-2 ,PSB-5,7,showed least growth (8.66 mm), whereas minimum growth showed in PSB-2, and PSB-3 (8.00 mm).

Regarding mancozeb, at 0.25% concentration, PSB-6 and PSB-8 showed maximum growth (7.66 mm), whereas PSB-2 and PSB-3 showed minimum growth (6.33 mm). At 0.38% maximum growth showed by PSB-7 (8.66 mm inhibition zone) followed by PSB-8 (8.66 mm), least growth recorded in PSB-10, PSB-2, (7.33 mm).

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