Research Paper :

Effect of *Pseudomonas fluorescens* Pf1 formulation application on rhizosphere and phyllosphere population in groundnut

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SUMMARY

Late leaf spot caused by *Cercosporidium personatum* is the destructive foliar disease in groundnut. The effect of seed treatment and foliar application of *Pseudomonas fluorescens* Pf1 formulation on groundnut rhizosphere and phyllosphere population was studied. The results revealed that when Pf1 formulation treated seeds were sown, the rhizosphere population of *P. fluorescens* was increased. The rhizosphere population of *P. fluorescens* increased with increase in age of the crop. *P. fluorescens* population was also found to be increased in phyllosphere due to foliar application of Pf1 formulation.

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The late leaf spot disease of groundnut, caused by the fungus, *Cercosporidium personatum* (Berk. and Curt.) Deighton is almost co-existent with the crop and contributes to significant loss in yield throughout the world (Wells *et al.,* 1994). Leaf spots damage the plant by reducing available photosynthetic area by lesion formation and by stimulating leaflet abscission. Smith *et al.* (1992) reported pod loss of 10 to 50 per cent by late leaf spot disease. Fluorescent Pseudomonads have emerged as the largest and potentially most promising group of plant growth promoting rhizobacteria (PGPR) for biocontrol of plant diseases (Kloepper and Schroth, 1978). Among the various biocontrol agents, fluorescent Pseudomonads are known to survive both in rhizosphere (Parks *et al.,* 1991) and phyllosphere (Wilson *et al.,* 1992). The present study was undertaken to find out the effect of seed treatment and foliar application of *P. fluorescens* Pf1 formulation on rhizosphere and phyllosphere population in groundnut.

MATERIALS AND METHODS

Groundnut seeds cv. (TMV 7) were treated with Pf1 formulation at different dosages *viz*., 10, 20, 50,100 and 125 g kg^{-1} of seed and sown in pots containing field soil at the rate of 5 seeds per pot and 5 pots for each replication. As a check, seeds were treated with carbendazim at the rate of 2 g kg^{-1} of seeds. At 45 days after sowing, the plants were inoculated with the pathogen. The trial was laid out in a randomized block design with six replications. The rhizosphere population of *P. fluorescens* was enumerated at 15 days interval by the method of Papavizas and Davey (1961).

Pf1 talc-based formulation at different concentrations *viz*., 0.5, 1.0, 1.25, 1.5 and 2.0 kg ha^{-1} was dissolved in water, allowed to settle for 2 h, filtered through muslin cloth and the filtrate was sprayed on to groundnut plants on 30, 45, 60, 75 and 90 DAS. As a check, the plants were sprayed with carbendazim at the rate of 500 g ha^{-1} and mancozeb 1 kg ha^{-1}. The plants were inoculated with pathogen at 45 DAS. The population of *P. fluorescens* was assessed from 1 g of leaf samples. The leaf samples without surface sterilization were transferred to a test tube containing 10 ml of sterile water and shaken well for 30 minutes. Then serial dilutions were made and the population of *P. fluorescens* was estimated using KB medium.

Table 1 : Effect of seed treatment with Pf1 formulation in promoting colonization of *P. fluorescens* in groundnut rhizosphere

Dosage of Pf1 formulation (g kg^{-1} seed)	Rhizosphere population of *P. fluorescens*					
	0	15	30	45	60	75
10	3.0^{a} (7.4)	3.7^{b} (7.5)	4.3^{c} (7.6)	5.0^{d} (7.6)	5.7^{c} (7.7)	7.0^{c} (7.8)
20	3.0^{a} (7.4)	3.7^{b} (7.5)	4.7^{c} (7.6)	6.0^{cd} (7.7)	7.3^{bc} (7.8)	9.0^{bc} (7.9)
50	3.3^{a} (7.5)	4.3^{b} (7.6)	6.3^{b} (7.7)	7.0^{bc} (7.8)	8.3^{b} (7.9)	9.7^{b} (7.9)
100	3.3^{a} (7.5)	5.1^{a} (7.7)	7.0^{ab} (7.8)	8.7^{ab} (7.9)	10.3^{a} (8.0)	11.0^{ab} (8.0)
125	3.0^{a} (7.4)	5.1^{a} (7.7)	7.3^{a} (7.8)	9.0^{a} (7.9)	10.3^{a} (8.0)	12.3^{a} (8.0)
Carbendazim @ 2 g kg^{-1} seed	1.3^{b} (7.1)	1.7^{c} (7.2)	1.7^{e} (7.2)	2.3^{e} (7.3)	3.0^{d} (7.4)	3.3^{d} (7.5)
Control – Pathogen inoculated	1.3^{b} (7.1)	1.7^{c} (7.2)	2.0^{d} (7.3)	2.3^{e} (7.3)	2.3^{d} (7.3)	2.7^{d} (7.4)
Control – Pathogen uninoculated	1.3^{b} (7.1)	2.3^{c} (7.3)	2.7^{d} (7.4)	2.7^{e} (7.4)	3.0^{d} (7.4)	3.7^{d} (7.5)

Data represent mean of three replications
Data followed by the same letter in a column are not significantly different (P=0.05) by DMRT
Figures in the parentheses are log transformed values

Table 2 : Effect of foliar spray with Pf1 formulation on phyllosphere population of *P. fluorescens*

Dosage of Pf1 formulation (kg ha^{-1})	Phyllosphere population of *P. fluorescens* (cfu x 10^{6} g^{-1} leaf)				
	Days after foliar spray				
	0	5	10	15	30
0.5	2.3^{b}	3.0^{a}	3.7^{b}	5.0^{b}	2.0^{ab}
1.0	3.0^{a}	3.7^{a}	6.0^{a}	8.0^{a}	2.7^{a}
1.25	3.0^{a}	3.7^{a}	6.0^{a}	8.3^{a}	2.7^{a}
1.5	3.3^{a}	4.0^{a}	6.0^{a}	8.3^{a}	3.0^{a}
2.0	3.3^{a}	4.0^{a}	6.3^{a}	8.3^{a}	3.0^{a}
Carbendazim 500 g ha^{-1} + Mancozeb 1 kg ha^{-1}	1.0^{c}	1.3^{b}	1.3^{c}	1.0^{c}	1.0^{b}
Control – Pathogen inoculated	1.0^{c}	1.3^{b}	1.0^{c}	1.0^{c}	1.0^{b}
Control – Pathogen uninoculated	1.0^{c}	1.7^{b}	1.3^{c}	1.3^{c}	1.0^{b}

Data represent mean of three replications
Data followed by the same letter in a column are not significantly different (p=0.05) by DMRT

RESULTS AND DISCUSSION

When Pf1 formulation treated seeds were sown in pots under greenhouse condition, the bacterium multiplied in the rhizosphere well. The rhizosphere population of *P. fluorescens* increased with increase in the age of the crop. The increasing dosage of seed treatment with Pf1 formulation increased rhizosphere population of *P. fluorescens* (Table 1). When the *P. fluorescens* treated seeds were sown, the bacterium established well in the rhizosphere of groundnut. The good establishment of *P. fluorescens* in the rhizosphere of plants was due to the capacity of the antagonist to compete for root exudates and the disease suppression by the introduced bacteria depend on their ability to colonize roots.

In the present study, *P. fluorescens* population was found to be increased in phyllosphere at 15 days after foliar application of Pf1 formulation and thereafter declined (Table 2). The results clearly indicated that *P. fluorescens* was able to multiply utilizing the nutrients available on and in the leaves. *P. fluorescens* when sprayed on rice foliage, the intensity of sheath blight disease was reduced (Vidhyasekaran and Muthamilan, 1999). The bacterium was found to survive on the foliage when sprayed on the plants. Phyllosphere population of the bacterium was 1.1, 1.4, 1.5 and 2.3 x 10^{4} cfu g^{-1} leaves when 0.2, 0.3, 0.4 and 0.5 per cent of the peat-based formulation was sprayed, respectively (Rabindran and Vidhyasekaran, 1996).

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