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Management of anthracnose of black pepper with biocontrol agents

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Anthracnose (fungal pollu) disease in black pepper (*Piper nigrum* L) caused by *Colletotrichum gloeosporioides* is wide spread damaging several black pepper gardens in the high ranges of Idukki district, Kerala and is becoming a major problem. Crop loss due to pests and diseases has been identified as a major constraint in the production of black pepper. *C. gloeosporioides* is characterized by straight conidia, with rounded or at times pointed ends, ranging 12-19 mm long (Arx, 1957) and 5-35 mm long (Davies *et al.,* 1992). The fungus infects the leaves as well as the spikes of black pepper. Anthracnose caused by *C. gloeosporioides,* earlier restricted to nurseries, also causes spike shedding in adult vine. The disease is noticeable on affected berries by the presence of characteristic brown colour and sunken patches during the early stages. Early infection prevents the development of the berries. Spike infection during early stages of growth results in premature shedding. The presence of characteristic cracks on the infected berries is a distinguishing feature of the disease. The fungus also causes angular to irregular characteristic brownish lesions with chlorotic halo on the leaves (Shanmugavelu *et al*., 2002). In recent years, fungal pathogens have been successfully managed by using antagonists (Elad *et al*., 1980, Vijayan *et al*., 1994, Vijayan *et al*., 2009), but no work has been reported on the management of anthracnose disease of black pepper in the field using bioagents. Being export oriented spice crop, it should be free from pesticide residue. Keeping these in view, a study was conducted using promising native isolates of antagonists *viz., Trichoderma harzianum*, *Pseudomonas fluorescens* and *Bacillus subtilis* for management of anthracnose disease in black pepper in the field.

The field experiment for management of the anthracnose disease was conducted in a disease prone planters' field in Idukki district, Kerala for three consecutive years from 2005 to 2007. The trial was laid out with six treatments in randomized block design and each treatment was replicated four times with 15 plants per plot. The plots were maintained followed by standard agronomical practices. The treatments were i) *Bacillus subtilis* as foliar spray and basal drenching (10^9 cfu ml $^{-1}$); ii) *Pseudomonas fluorescens* as foliar spray and basal drenching (10^9 cfu ml $^{-1}$); iii) *Trichoderma harzianum* (10^9 cfu ml $^{-1}$) as foliar spray and basal drenching; iv) Consortium of *B. subtilis*, *P. fluorescens* and *T. harzianum* as foliar spray and soil drenching (10^9 cfu ml $^{-1}$); v) Bordeaux mixture (1%) as foliar spray and basal drenching and vi) control without any biocontrol agents or fungicides. Three rounds of treatments were given during July, August and September months at 30 days intervals. The bioagents were mass multiplied in liquid media (Kings' B Media for *P. fluorescens,* Nutrient broth *for B. subtilis* and PDA broth for *T. harzianum*) and used for spraying and drenching in the field. All the plants in the trial plots were given vermicompost @ 5 kg per plant and neem cake @ 500 gm per plant. In all cases, spraying with respective treatments (biocontrol agents and Bordeaux mixture) was given after the phytosanitation. Observations on percentage disease

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Table 1. Effect of bioagents and Bordeaux mixture on leaf infections in black pepper

Treatments	Concentration (%)	Mean leaf infection (%)			Pooled mean	Disease control (%)
		I year	II year	III year		
T1- *B. subtilis*	10^9cfu ml^{-1}	39.74(38.89)	10.31(18.71)	19.17(25.85)	23.07(28.59)	58.80
T2- *P. fluorescens*	10^9cfu ml^{-1}	38.94(38.22)	9.74(18.05)	12.50(20.55)	20.39(26.73)	63.58
T3- *T. harzianum*	10^9cfu ml^{-1}	61.55(51.73)	29.49(32.51)	27.83(31.81)	39.62(39.01)	29.24
T4- *T. harzianum*+ *P. fluorescens*+ *B. subtilis*	10^9cfu ml^{-1}	31.08 (33.43)	13.10 (21.09)	11.33(19.63)	18.50 (25.24)	66.96
T5- Bordeaux mixture	1%	26.23 (30.57)	13.01 (20.92)	12.67(20.83)	17.10 (24.40)	69.45
T6- Control	-	65.94 (54.30)	46.79 (43.13)	53.0(46.73)	55.99 (48.43)	-
CD (5%)		11.55	6.49	4.18	4.58	

Figures in parenthesis are arc sine transformed values

infections on spikes and leaves were recorded periodically. The data were statistically analyzed.

Pooled data for the three consecutive years are presented in table 1 and 2. The data indicate that application of Bordeaux mixture, consortium of bioagents and *P. fluorescens* were on par for the management of foliar infection caused *C. gloeosporioides* in black pepper in the field followed by spraying and basal drenching of *B. subtilis* (Table 1). The results of the field study showed that plants treated with consortium of bioagents (*B. subtilis*, *P. fluorescens* and *T. harzianum*) significantly reduced the incidence of spike infection followed by application of *P. fluorescens* and *B. subtilis* and Bordeaux mixture application as compared to control plots (Table 2). The data also revealed that maximum disease control (87.79%) was recorded with the application of consortium of bioagent followed by Bordeaux mixture spray and basal drenching of individual bacterial bioagent (*P. fluorescens* or *B. subtilis*). A maximum yield of 1378 kg ha^{-1} was recorded in the plot treated with consortium of bioagents followed by foliar spray and basal drenching with *P. fluorescens* (1008.8 kg ha^{-1}).

The results of the present findings revealed that application of consortium of bio-agents was most effective in reducing the anthracnose (fungal pollu disease) of black pepper in the field plantations. Three applications of consortium of bioagents as foliar spray and basal drenching at 30 days intervals from July month onwards were significantly effective in controlling the disease in the plantations. This offers an advantage for the organically managed black pepper plantations where the use of chemical fungicides is excluded. A prophylactic spraying of

Table 2. Effect of bioagents and Bordeaux mixture on spike infections in black pepper

Treatments	Concentration) (%)	Mean spike infection (%)			Pooled mean	Disease control (%)	Yield (kg ha^{-1})
		I year	II year	III year			
T1- *B. subtilis*	10^9cfu ml^{-1}	19.76(25.47)	5.52(13.02)	14.00(21.95)	13.09(21.17)	79.19	912.00
T2- *P. fluorescens*	10^9cfu ml^{-1}	20.96(27.17)	4.80(12.45)	10.67(18.94)	12.14(20.34)	80.70	1008.80
T3-*T. harzianum*	10^9cfu ml^{-1}	38.08(37.65)	20.03(26.25)	28.33(32.13)	28.81(32.41)	54.20	438.40
T4- *T. harzianum*+ *P. fluorescens*+ *B. subtilis*	10^9cfu ml^{-1}	10.08(17.73)	5.96(13.87)	7.00(15.18)	7.68(16.0)	87.79	1378.40
T5- Bordeaux mixture	1%	9.37(17.27)	11.89(20.06)	12.50(20.60)	11.24(19.51)	82.13	976.00
T6- Control	-	56.67(48.87)	72.97(57.95)	59.08(50.26)	62.91(52.52)	-	412.00
CD (5%)		11.33	5.79	4.05	3.16		

Figures in parenthesis are arc sine transformed values

biocontrol agents based on early or late infection of anthracnose in black pepper was stressed in the high range of Idukki District. It has been suggested that PGPR isolated from black pepper rhizosphere was more efficient for controlling black pepper disease in the nursery cuttings (Lisha *et al.,* 2002).

Strains of *P. fluorescens* and *Bacillus* sp. were found to increase the growth and vigour of in small cardamom seedlings apart from suppressing the soil borne diseases (Thomas and Vijayan, 2003). Biological control methods have been developed for managing the diseases in cardamom, ginger, vanilla and black pepper (Anandaraj and Sarma, 1994; Vijayan *et al*., 1994; Sarma and Anandaraj, 1998; Anandaraj, 2000; Vijayan *et al*., 2009). Native isolates of *P. fluorescens* were found effective in reducing the leaf rot caused by *C. gloeosporioides* and *Exerohilum rostratum* in coconut root wilt effected areas (Sarma *et al*., 2003). It has been suggested that fluorescent pseudomonads have the ability to synthesize hydrogen cyanide (HCN), which is known to inhibit the expression of pathogenic fungi (Voisard *et al*., 1989) and also possess the ability to hydrolyze the toxin produced by some pathogenic fungi (Mauch *et al*., 1988).

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