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PP Salvi
Ph.D. Scholar, Department of Plant Pathology, College of Agriculture, Dr. BSKKV, Dapoli, Dist. Ratnagiri, (M.S.), India

VS Pande
Professor and Head Department of Plant Pathology, College of Agriculture, Dr. BSKKV, Dapoli, Ratnagiri, (M.S.), India

SV Pawar
M.Sc. Student, Department of Plant Pathology, College of Agriculture, Dr. BSKKV, Dapoli, Ratnagiri, (M.S.), India

PV Joshi
M.Sc. Student, Department of Plant Pathology, College of Agriculture, Dr. BSKKV, Dapoli, Ratnagiri, (M.S.), India

Correspondence
SV Pawar
M.Sc. Student, Department of Plant Pathology, College of Agriculture, Dr. BSKKV, Dapoli, Ratnagiri, (M.S.), India

Effect of different fungicides and bio control agents against *Sclerotium rolfsii* sacc. Causing collar rot and root rot of pigeon pea under *in vitro* condotion

PP Salvi, VS Pande, SV Pawar and PV Joshi

Abstract
Fungicides and bio control agents have long been used for the management of plant diseases. The objectives of this study were to investigate the efficacy of different fungicides and bio-agents against pathogen. For effective management of collar rot and root rot disease, eight fungicides *viz*., Difenoconazole, Hexaconazole, Propiconazole, Thiophenate methyl at 0.1 per cent, Hexaconazole and Propiconazole at 0.05 per cent and Mancozeb at 0.25 per cent concentration and three bio-agents namely *Trichoderma viride*, *T. harzianum* and *Pseudomonas flurescens* were screened *in vitro*. All fungicides were shows inhibitory effects against growth of mycelium and sclerotia formation of *Sclerotium rolfsii*. *Trichoderma viride* shows maximum growth inhibition (83.33%) against pathogen.

Keywords: *Sclerotium rolfsii*, Pigeon pea, Fungicides and Bio control agents,

Introduction

Pulses are the major source of dietary protein in vegetarian diet in our country. Besides being a rich source of protein, they maintain soil fertility through biological nitrogen fixation in soil and thus play a vital role in furthering sustainable agriculture. At present, globally 60 million tonnes of pulses are produced annually from 70 million hectares. The contribution of developing countries like India, China, Brazil, Turkey and Mexico accounts for nearly two third production. India is largest producer with 33 per cent of global area contributing 22 per cent of the world's production. Normally the area under pulses in the country is around 23.63 million hectares with production of 14.76 million tonnes. The average productivity of the country is about 760.33 kg/ha against the average global productivity of 857 kg/ha. Pigeon pea is an important pulse crop in semi-arid tropics of Andhra Pradesh and Maharashtra States. It is a versatile crop and ideally suited for drought-prone areas. Being a drought tolerant crop, pigeon pea is being raised as a sole main crop in Andhra Pradesh State, while it is grown as an inter-crop and subsidiary crop in Maharashtra State with cotton, sorghum and/or green gram as the main crops.

In konkan region of Maharashtra pigeon pea is grown mainly on bunds of rice fields. This crop is sown during kharif in month of June-July after transplanting of rice. This crop matures in the month of November-December. This crop is also grown in rice fields after harvest of rice on residual moisture during the month of October and matures in the month of February-March. The farmers are using the seed material of any pigeon pea variety and therefore yield is less. It is necessary to identify the pigeon pea variety for growing on rice bunds. In konkan region of Maharashtra rice is grown on about 4.2 lakh hectare areas. This crop is grown on bunds of rice fields as the rice bunds have more residual moisture than the field, the growing of pigeon pea on rice bund increase the total cropped area and this crop grows very well and produces a good yield. The diseases on pigeon pea is one of the major constrain for lower yield of this crop. The severe incidence of collar rot and root rot disease caused by *Sclerotium rolfsii* was recorded in Tamnath village in Karjat tahasil of Raigad district. As this fungus was observed for first time in the Konkan region. Therefore it was felt necessary to carry out the basic studies against the pathogen

Materials and Methods

Efficacy of fungicides against pathogen

Efficacy of eight fungicides along with control were studied *in vitro* by following Poisoned Food Technique (Nene and Thapliyal, 1993) [7] against collar and root rot of pigeon pea with different concentrations (0.05, 0.1 and 0.25). Required quantity of each fungicide under study was mixed thoroughly in sterilized 100 ml PDA media filled in 250 ml flask separately under aseptic condition. The medium was supplemented with streptomycin sulphate (50 ppm) to prevent bacterial contamination. The poisoned medium (20 ml) was then poured in sterilized Petri plates and allowed it to solidify. The plates were then inoculated with five mm diameter disc of seven days old culture of test pathogen by placing in the centre of the plate. Control was maintained for each set where fungal disc were placed on PDA medium without fungicide. Each treatment was replicated three times. The inoculated plates were then incubated at 27 ± 1º C in BOD incubator. The observations on mycelia growth, number of sclerotia and per cent growth inhibition of test pathogen were recorded after 7 days of incubation. The per cent growth inhibition (PGI) of pathogen in each treatment was calculated by the method suggested by Horshfall (1956) [4].

$$X = \frac{Y - Z}{Y} \times 100$$

Where,
X = Per cent inhibition
Y = Growth of fungus in control (mm)
Z = Growth of fungus in treatment (mm)

Efficacy of bio-agents against pathogen

The effectiveness of fungal bio agents *viz*, *Trichoderma harzianum*, *T. viride* and *Pseudomonas fluorescens* against causal organism *in vitro*, a trial was laid out in Petri plates. For this experiment, *T. harzianum*, *T. viride*, and *Pseudomonas fluorescens* and causal organism were grown on PDA for seven days until the Petri plates were fully covered and the fungal disc of 5 mm in diameter were cut with sterile cork borer for transferring them aseptically on PDA in Petri plates. The fungal discs were placed in such a manner in plate, so that the fungi get equal opportunity for growth. In all seven treatments were replicated three times. Treatment number seven served as pure check. The observations were recorded for the colony diameter, colony growth on which sclerotial formation was inhibited due to antagonistic effect of these bio-agents. Per cent inhibition of growth was calculated as described earlier.

Table 1: Efficacy of fungicides against *Sclerotium rolfsii*

Tr. No	Treatments	Conc. %	Mean colony Diameter (cm)*	Per cent Inhibition Over control	No. of Sclerotia formed
T_1	Carbendazim	0.1	8.8	2.22	320
T_2	Hexaconazole	0.05	0.0	100	0
T_3	Validamycin	0.1	8.5	5.55	185
T_4	Hexaconazole	0.1	0.0	100	0
T_5	Propiconazole	0.05	0.0	100	0
T_6	Copper Oxychloride	0.2	8.6	4.44	210
T_7	Difenconazole	0.1	0.0	100	0
T_8	Propiconazole	0.1	0.0	100	0
T_9	Thiophenate methyl	0.1	0.0	100	0
T_{10}	Mancozeb	0.25	0.0	100	0
T_{11}	Control	--	9.0	--	370
S. E. m ±			0.39		
C.D at 1%			1.58		

* Mean of three replications

Table 2: Efficacy of bio-agents against *Sclerotium rolfsii*

Tr. No	Placement detail	Mean colony diameter (cm)*	Per cent inhibition over control	No. of sclerotia formed
T_1	*Th Sr*	1.50	83.33	10
T_2	*Th Sr Th*	2.25	75.00	25
T_3	*Pf Sr*	6.50	27.77	115
T_4	*Pf Sr Pf*	7.00	22.22	112
T_5	*Tv Sr*	3.50	61.11	50
T_6	*Tv Sr Tv*	2.25	75.00	42
T_7	*Sr*	9.00	--	180
S.Em ±		1.68		
C.D at 1%		7.10		

* Mean of three replications

Results and Discussion

The data on the efficacy of different fungicides and their effect on mycelial growth and sclerotia formation of *Sclerotium rolfsii* are presented in Table 1. The data revealed that all the fungicides inhibited the mycelial growth and sclerotia formation. Hexaconazole (0.05%), Hexaconazole (0.1%), Propiconazole (0.05%), Propiconazole (0.1%), Difenoconazole (0.1%), Thiophenate methyl (0.1%) and Mancozeb (0.25%) completely inhibited (100%) the growth and sclerotia formation of *S. rolfsii*. Validamycin (0.1%), Copper Oxychloride (0.2%) and Carbendazim (0.1%) were totally ineffective against the pathogen as they were statistically at par with control with respect to mycelial growth. The number sclerotia formed in these three treatments was also high indicating their inefficacy in checking sclerotia formation. These findings are in concurrence with those reported by Prabhu and Hiremath (2003) [11], Tiwari and singh (2004) [13], Mundhe (2005) [6], Patil (2007) [10], Haralpatil and

Raut (2008) [3], Patel *et al.* (2008) [9], Sawant *et al.* (2013) [12] and Pandav *et al.* (2013) [8]. The results of present study revealed that the fungicides in Trizole group are very effective against the soil borne pathogen.

The data presented in table 2 showed that, treatment T_1 was significantly superior to rest of the treatments. However, treatments T_2, T_3, T_4, T_5 and T_6 were statistically at par with control (T_7). In Treatment T_3 the pathogen grew profusely but the growth was restricted when it reached closer to the growth of antagonist. In treatment T_5 also initially the growth of the pathogen was restricted by antagonist but some mycelium of the pathogen grew over to that of the antagonist after few days. In case of the treatments T_2, T_4 and T_6 wherein a single bit of the pathogen was placed in centre and two bits of antagonist were placed on both sides of the pathogen, only the fungal antagonists (*T. harzianum* -T_2) and *T. viride* - T_6) were successful in inhibiting the mycelial growth of the pathogen, but the growth of bacterial antagonist (P. *fluorescens* –T_4)was covered by the mycelial growth of the pathogen indicating that the pathogen was successful in overcoming the antagonism of *P. fluorescens*. The above findings are in conformity with the results of Bell *et al*. (1982) [1], who reported that *T. harzianum* was the most effective bio-control against *S. rolfsii*, both *in vitro* and *in vivo*. Dutta and Das (2002) [2] found that out of three *Trichoderma* spp. T. *harzianum* was inhibitory to *Sclerotium rolfsii* Sacc. as it caused 61.5 per cent inhibition of mycelial growth followed by *T. viride* (59.1%) and *T. koningii* (57.2%) as compared to control. Mundhe *et al*. (2009) [5] found that maximum inhibition of mycelial growth of *Sclerotium rolfsii* causing Nagli foot rot, occurred due to *T. harzidnum* (73.77%) followed by *T. viride* (72.66%) and *P. fluorescens* (71.55%). The results of present study are in accordance with those of Mundhe *et al*. (2009) [5], in respect of fungal bio-agents but not in case of *P. fluorescens*. It was interesting to note that, in the present study, the mycelium of the pathogen overgrew on the growth of *P. fluorescens*.

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