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Evaluation of fungicides for the control of Sclerotinia stem rot of Indian mustard caused by *Sclerotinia sclerotiorum* (Lib.) de Bary

Rakesh*, A.S. Rathi, Anil Kumar and Hawa Singh

Department of Plant Pathology, CCS Haryana Agricultural University, Hisar-125 004 (Haryana), INDIA
*Corresponding author. E-mail: punia.rakesh98@gmail.com

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Abstract: The current investigation was carried out under -*in vitro* and under sick plot conditions of the Department of Plant Pathology, CCS Haryana Agricultural University, Hisar to test the efficacy of different fungicides against *Sclerotinia sclerotiorum*. The experiment was carried out through poison food technique under *in vitro* conditions and through foliar sprays under sick plot conditions. Efficacy of fungicides *in* vitro against *S. sclerotiorum* showed that carbendazim and hexaconazole completely inhibited mycelial growth up to 100 per cent at 50 ppm concentration. Propiconazole inhibited 96.39 per cent mycelial growth at 50 ppm and 100 per cent at 1000 ppm, while tricyclazole failed to inhibit mycelial growth up to 100 ppm, however, at 1000 ppm concentration, there was 100 per cent mycelial inhibition. Mancozeb and captan completely inhibited up to 100 per cent mycelial growth at 500 ppm concentration. Copper oxychloride and pencycuron were not effective at lower concentrations but had inhibited 45.28 and 22.50 per cent mycelial growth at 1000 ppm. Field experiment was conducted to test the efficacy of those fungicides which were found best under *in vitro* conditions. All the four fungicides tested significantly controlled Sclerotinia stem rot disease and increased the seed yield as compared to untreated control. Two Foliar sprays with hexaconazole @ 0.05% at 45 and 60 days after sowing was found most effective in controlling disease incidence up to 78.3 per cent and increasing seed yield up to 37.6 per cent as compared to untreated control.

Keywords: Fungicides, Mycelia growth, Sclerotinia stem rot, Sclerotinia sclerotiorum

INTRODUCTION

Indian mustard [*Brassica juncea* (L.) Czern & Coss.] is one of the major oilseed crops cultivated in India and around the world. Among rapeseed-mustard growing countries in the world, India ranks third in area and production after China and Canada (Anonymous, 2013). In India, rapeseed-mustard crops are cultivated on an area of 6.51 million ha with a production of 7.67 million tones and with an average yield of 1179 kg/ha (Anonymous, 2013). Despite considerable increase in productivity and production, a wide gap exists between yield potential and yield realized at farmer's field, which is largely due to biotic and abiotic stresses. The destructive diseases of rapeseed-mustard include those caused by fungi, bacteria, viruses and phytoplasma. Among them, Sclerotinia stem rot is the most serious fungal disease that causes maximum damage in Indian mustard (Rakesh *et al*., 2015). Once the pathogen is established, it is difficult to manage due to its soil borne nature and wide host range. Control of this disease by the use of different fungicides with varying degree of success has been reported in the literature (Mehta *et al*., 2005) but no economical and practical solution through the use of fungicides has been made so far. Moreover, chemical sprays in mustard is not feasible and economical as this disease appears late at pod formation stage to maturity in Haryana conditions (Rathi and Singh, 2009). However, significant control of disease through prophylactic sprays with systemic fungicides has been reported under field conditions (Rathi *et al*., 2012). The explosive pathogenicity of *S. sclerotiorum* under favourable conditions and the ability of its sclerotia to withstand adverse conditions allow it to be a successful pathogen. Although efficacy of various fungicides against *Sclerotinia* species has been well demonstrated (Singh *et al.*, 2014), it has not been controlled consistently and economically due to prolonged viability and unpredictable nature of fungal propagules (Singh and Kapoor, 1993). *In vitro* effect of two systemic fungicides *viz.*, carbendazim and metalaxyl at different concentrations (22, 50 and 100 mg/ml) and three non-sytemic *viz.*, captaf, mancozeb and copper oxychloride at same concentration were evaluated against Sclerotinia stem rot pathogen. Among all the five fungicides carbendazim was found to compeletly (100%) inhibited the mycelia growth of the pathogen (Chand *et al*., 2009). Shivpuri *et al*., (2001) observed that fungicides, carbendazim, thiophenate methyl and phenylpyrrole had completely inhibited the growth of the pathogen at all the concentrations tested *in vitro*. Mancozeb was found effective at higher concentration while, copper oxychloride was least effective as it did not cause substantial reduction

in growth of the pathogen and Antracol was mildly effective exhibiting mean growth of 40.6 mm as against 88.3 mm in check. Sharma *et al*. (2006) observed that captan completely inhibited mycelia growth with EC_{50} value less than 1 µg a.i. ml^{-1}. Seed treatment with carbendazim and foliar spray of the same at 65 days after sowing (DAS) proved most effective in reducing disease incidence (91%), intensity (98%) and increasing seed yield (91%) over untreated check. Use of carbendazim and captan as foliar sprays has been reported to be very effective against Sclerotinia rot of pea (Sharma, 1987), rape (Shen, 1993). Roy and Saikia (1976) found that mancozeb @ 0.15% also reduced white mould in mustard. Singh *et al*., (1994) reported that the benomyl, carbendazim and mancozeb @ 0.2% controlled *S. sclerotiorum* on mustard and reduced the disease by 91.3, 85.7 and 54.7 per cent, respectively. Sasirekhamani *et al*., (2013) reported that the hexaconazole exerted an excellent fungistatic effect at 100 µg/mL concentration on *S. sclerotiorum*. Therefore, this fungicide could be effectively used for the control of the notorious pathogen, *S. sclerotiorum* at a time when many pathogens are acquiring resistance to different classes of fungicides. Rathi *et al*., (2012) also reported that prophylactic foliar spray with carbendazim @ 0.1% twice at 45 and 60 DAS was most effective in controlling Sclerotinia stem rot disease in Indian mustard. Since the pathogen is very difficult to manage due to its soil borne nature and wide host range, therefore the present study was carried out with the objective to evaluation of fungicides for the control of Sclerotinia stem rot of Indian mustard caused by *Sclerotinia sclerotiorum* under *in vitro* and sick plot conditions.

MATERIALS AND METHODS

Efficacy of different fungicides on inhibition of *S. sclerotiorum in vitro* and under sick plot conditions

Evaluation of fungicides under *in vitro* conditions: The Efficacy of eight fungicides viz., mancozeb, carbendazim, copper oxychloride, tricyclazole, propiconazole, hexaconazole, captan and pencycuron on the growth of *S. sclerotiorum* were tested under *in vitro* conditions using the standard procedure of poison food technique as given by Mayer (1962). Stock solution of each fungicide was prepared in double strength *i.e.* 50, 100, 200, 500 and 1000 ppm by dissolving weighed or measured quantity of fungicide in a measured volume of sterilized water. The double strength potato dextrose agar (PDA) medium was also prepared and sterilized at 15 lb pressure for 20 minutes. An equal volume of chemical solution and PDA was mixed in a sterilized conical flask and poured aseptically in the Petri plates. After solidification of medium, each Petri plate was centrally inoculated with 5 mm disc of fungus taken from 8 days old culture of *S. sclerotiorum* with the help of sterilized cork borer and incubated at 21±1°C. Suitable controls were maintained for each chemical. Four replications of each fungicide were maintained and CRD was followed. Colony diameter of the fungus of each treatment along with control was measured (mm) and recorded after every 24 hours, till the test fungus occupied the full Petri plate in the controlled treatment. The per cent inhibition of mycelial growth over control was calculated by following formula given by Vincent (1947).

$$\text{Growth inhibition (\%)} = \frac{(C\text{-}T)}{C} \times 100$$

Where,

C= Radial growth of *S. sclerotiorum* mycelium in control.

T= Radial growth of *S. sclerotiorum* mycelium in treatment.

Evaluation of fungicides under sick plot conditions: The field experiment was conducted at oilseeds research area of Department of Genetics and Plant Breeding, CCS HAU, Hisar during *rabi* 2013-14 to test the efficacy of four fungicides which were found best under *in vitro* conditions. Cultivar Varuna was sown during first week of November, 2013 under sick plot conditions 5×4 m plots using RBD with three replications. These four fungicides (carbendazim @ 0.1%, captan @ 0.2%, hexaconazole @ 0.05% and propiconazole @ 0.05%) were used as two foliar sprays, first at 45 days and second at 60 days after sowing (DAS). Plots sprayed with plain water served as control. Observation on per cent disease incidence was recorded 15 days before harvest. The plant showing even a minute lesion of stem rot was considered as a diseased plant. Seed yields (q/ha) were also recorded per plot.

RESULTS AND DISCUSSION

Efficacy of fungicides was tested *in vitro* under laboratory conditions for the per cent mycelial growth inhibition of *S. sclerotiorum*. The results of the experiment in table 1 clearly show that carbendazim and hexaconazole completely inhibited mycelial growth up to 100 % at 50 ppm concentration. Propiconazole at 200 ppm concentration completely inhibited up to 100 % as compared to 96.39 % inhibition at 50 ppm concentration. Mancozeb and captan at 500 ppm concentration completely inhibited up to 100 %, while tricyclazole completely inhibited mycelial growth at 1000 ppm. Copper oxychloride and pencycuron were found least effective, as they inhibited 45.28 and 22.50 % of fungal mycelial growth even at 1000 ppm, respectively.

The disease has become a major threat to successful cultivation of Indian mustard in Haryana state; hence use of fungicides as prophylactic foliar sprays seems to be a solution to this problem. The fungicide carbendazim (0.1%) was found most effective as it completely inhibited (100%) the pathogen growth

Table 1. In vitro evaluation of different fungicides against *S. sclerotiorum.*

Fungicides	Per cent inhibition at different concentration (ppm)					Mean
	50*	100*	200*	500*	1000*	
Copper oxychloride**	0.00 (4.05)	0.00 (4.05)	0.00 (4.05)	19.44 (24.96)	45.28 (42.22)	12.94 (15.86)
Mancozeb	15.56 (23.21)	43.33 (41.13)	46.67 (43.08)	100.0 (90.00)	100.0 (90.00)	61.11 (57.48)
Captan	43.33 (40.48)	77.50 (61.89)	90.56 (72.26)	100.0 (90.00)	100.0 (90.00)	82.28 (70.93)
Tricyclazole**	0.00 (4.05)	0.00 (4.05)	15.56 (23.13)	91.94 (73.64)	100.00 (90.00)	41.50 (38.97)
Pencycuron**	0.00 (4.05)	0.00 (4.05)	6.67 (14.58)	10.00 (17.96)	22.50 (28.26)	7.83 (13.78)
Hexaconazole	100.0 (90.00)	100.0 (90.00)	100.0 (90.00)	100.0 (90.00)	100.0 (90.00)	100.00 (90.00)
Carbendazim	100.0 (90.00)	100.0 (90.00)	100.0 (90.00)	100.0 (90.00)	100.0 (90.00)	100.00 (90.00)
Propiconazole	96.39 (79.25)	98.61 (83.29)	100.0 (90.00)	100.0 (90.00)	100.0 (90.00)	99.00 (86.51)
Mean	44.41 (41.88)	52.43 (47.30)	57.43 (53.39)	77.67 (70.82)	83.47 (76.31)	
	Treatment F1		Concentration F2		Treatment × Concentration F1 × F2	
SEm±	0.90		0.71		2.02	
CD (p=0.05)	2.53		2.00		5.67	

*Mean of four replications; **+0.5 has been added in each observation of copper oxychloride, tricyclazole and pencycuron for statistical analysis; Figures in parenthesis indicate angular transformed values.

Table 2. Efficacy of different fungicides on stem rot incidence and seed yield of Indian mustard under field conditions.

Treatment*	Disease incidence (%)	Control (%)	Seed yield (q/ha)	Increase in Seed yield (%)
Carbendazim	12.3 (20.6)	76.3	14.3	31.1
Hexaconazole	11.3 (19.5)	78.3	15.0	37.6
Propiconazole	16.1 (23.6)	69.0	13.5	23.8
Captan	20.2 (26.7)	61.2	12.7	16.5
Control	52.1 (46.2)	-	10.9	-
CD (p=0.05)	4.0		1.2	
CV (%)	7.7		4.9	

Note: carbendazim (Bavistin @ 0.1%), hexaconazole (Contaf @ 0.05%), propiconazole (Tilt @ 0.05%), captan (Captaf @ 0.2%); *Two foliar sprays at 45 and 60 DAS; Figures in parentheses are angular transformed values.

(Prasad and Kumar, 2007; Chand *et al*., 2009; Singh *et al*., 2014). Sasirekhamani *et al*. (2013) also reported that the hexaconazole exerted an excellent fungistatic effect at 100 µg/mL concentration on *S. sclerotiorum.* Shivpuri *et al*. (2001) observed that the fungicides *viz*., carbendazim, thiophenate methyl and phenylpyrole had completely inhibited the growth of the pathogen at all the concentrations evaluated, while mancozeb was found effective at higher concentrations only, when tested under *in vitro* conditions. Similarly, Copper oxychloride, however, was found the least effective, as it did not cause substantial reduction in growth of the pathogen as compared to check. Effectiveness of mancozeb at higher concentrations only and least effectiveness of copper oxychloride was also observed even up to 1000 ppm in present studies.

All the four fungicides tested under field conditions significantly controlled Sclerotinia stem rot disease and increased the seed yield. Foliar spray with hexaconazole @ 0.05% twice at 45 and 60 DAS was found most effective in controlling disease incidence up to 78.3 % and increasing seed yield up to 37.6 % as compared to untreated control. Results with foliar spray with carbendazim @ 0.1% at same DAS were found at par with the above treatment, as it controlled the disease incidence up to 76.3 % and increased seed yield up to 31.1 % over untreated control. Propiconazole @ 0.05% followed by captan were also found effective (Table 2) in reducing the disease incidence and increasing the seed yield.

Large number of fungicides has been evaluated and carbedazim as seed treatment and foliar spray was reported to be the most effective in controlling disease up to some extent (Rathi *et al*., 2012). Use of carbedazim and captan as foliar sprays has also been reported to be very effective against Sclerotinia rot of pea and rape (Roy and Saikia, 1976; Sharma, 1987; Shen, 1993). Benomyl, carbendazim and mancozeb @ 0.2% as sprays were also reported to show their effectiveness in controlling stem rot in mustard by Singh *et al*., (1994). Rathi *et al*., (2012) were of the opinion that under Haryana conditions, the disease at farmer's field remained unnoticed till maturity and at that growth stage of plant there was not any solution to the problem as one could not physically enter in field to spray. They found good results with prophylactic sprays with carbendazim and recommended that in areas, where Sclerotinia rot was regularly appearing, spray the crop twice with carbendazim @ 0.1% at 45- 50 and 65-70 DAS.

Conclusion

The present study concluded that hexaconazole @ 0.05% was most effective under *in vitro* as it completely inhibited mycelia growth up to 100% at 50 ppm concentration. The foliar spray with same fungicide twice at 45 and 60 DAS was found most effective in controlling disease incidence up to 78.3 % and increas-

ing seed yield up to 37.6 % under field conditions. It may be tested as prophylactic sprays at farmer's fields in mustard, so that a recommendation can be made for control of this notorious pathogen with wide host range.

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