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Efficacy of seed treatment with fungicides on the viability of sclerotia of *Rhizoctonia solani* admixtured with the rice seed

Y Sandhya, CPD Rajan, M Reddi Kumar and A Ranga Rani

Abstract
The efficacy of seed treatment with different fungicides viz., Carbendazim, Propiconazole, Hexaconazole, Validamycin, Tebuconazole, Thifluzamide, Azoxysrtrobin, Difenoconazole, Tebuconazole 50% +Trifloxystrobin 25% (Nativo) on the viability of sclerotia admixtured with the seed was tested, in which all the tested fungicides showed 100 per cent inhibition of sclerotial germination in both wet and dry seed treatments. The seed admixtured with sclerotia was treated with different fungicides given above, and the effect of sclerotia on germination and seedling growth was tested by paper towel and pot culture methods. All the seed was germinated in both paper towel and pot culture methods. The efficacy of fungicides on seedling growth varied in both wet and dry seed treatments. All the tested fungicides showed increased seedling growth (root length and shoot length) except Carbendazim and Tebuconazole 50% + Trifloxystrobin 25% (Nativo) when compared to control in both paper towel and pot culture methods.

Keywords: fungicides, sclerotia, *Rhizoctonia solani*, sheath blight

Introduction
Rice is affected by several fungal, bacterial and viral diseases. Of these, rice sheath blight, caused by *Rhizoctonia solani* Kuhn is second only to, and often rivals rice blast in importance. It is a prominent disease in irrigated rice ecosystem. Currently, this disease is distributed in almost all the rice growing states. A modest estimation of losses due to sheath blight disease alone in India has been upto 54.3%. The fungus produces brown sclerotia depending upon the environmental conditions (Ou, 1985) [8]. Sclerotia are superficial, more or less globose but flattened, white when young and becomes brown. Individual sclerotium measures upto 5mm but may unite to form large mass in culture (Ou, 1985) [8]. The fungus survives in the soil for years as hard, resistant structures. The pathogen is known to cause the damage at different stages *viz.*, seed germination, seedling establishment and vegetative growth phase. As a result, productivity and quality of grains and seeds are reduced considerably. Besides the disease management, practice through cultural methods, chemical control, is the net promising method. Hence, in the present study fungicides were used for seed treatment to evaluate their efficacy in controlling sheathblight. Taking into consideration the above facts, the present research studies were initiated with a view of finding out the effects of different fungicides, and biogenic nano silver on the growth of the fungus, *R. solani* and survival of sclerotia.

Materials & Methods
The present experiments were carried out in the Department of Plant Pathology, S.V. Agricultural College, Tirupati, and Agricultural Research Station, Nellore, of Acharya N.G. Ranga Agricultural University, Guntur, Andhra Pradesh. Sheath blight susceptible variety of rice NLR-34449 (Nellore Mahsuri) was used in present studies. The test pathogen *R. solani* was isolated from sclerotial bodies attached to the diseased portion of rice plants.

Effect of seed treatment with different fungicides on the viability of sclerotia admixtured with the seed
Sclerotia of *R. solani* which were grown in culture were mixed with 100 g per lot of paddy seed @ 10 sclerotia per seed lot. The fungicides mentioned below were used for wet and dry seed dressing of the seeds in seed lots. Three replicates were maintained. The seed admixtured sclerotia were retrieved 24 h after wet seed dressing, whereas the sclerotia were retrieved 48 h after the dry seed dressing. The viability of the sclerotia was tested on PDA medium. After retrieval of sclerotia, the seed in the particular lot were kept for germination by following rolled paper towel method (Agarwal, 1994) [1] and pot culture method.

Experimental design used was CRD and three replications were maintained per treatment. The growth parameters of the seedlings were recorded for all the treatments. The list of fungicides with their concentrations used in the study are presented in the Table.1

Table 1: List of fungicides

S. No.	Fungicide	Dry seed dressing /Kg of seed	Wet seed dressing/ lt of water
1	Carbendazim 50WP	3 g	1g
2	Propiconazole 25EC	3 ml	1 ml
3	Hexaconazole 5EC	6 ml	2 ml
4	Validamycin 3%L	6 ml	2 ml
5	Tebuconazole 25EC	4.5 ml	1.5 ml
6	Thifluzamide 24 SC	3 ml	1 ml
7	Azoxystrobin 23SC	2 g	1 g
8	Difenoconazole 25EC	2 g	1 g
9	Tebuconazole 50% + Trifloxystrobin 25% WG (Nativo)	2 g	0.8 g

Results and Discussion

Effect of seed treatment with different fungicides on the viability of sclerotia admixtured with the seed.

Laboratory cultured sclerotia of *R. solani* were mixed with 100 g lots of paddy seed at 10 sclerotia per seed lot. The fungicides mentioned below were used for wet and dry seed dressing using the above seed lots. Three replicates were maintained. The growth parameters of the seedlings were recorded for all the treatments.

The fungicides used for seed treatment are Carbendazim, Propiconazole, Hexaconazole, Validamycin, Tebuconazole, Thifluzamide, Azoxysrtrobin, Difenoconazole, Tebuconazole 50% + Trifloxystrobin 25% (Nativo).

Seed admixtured sclerotia were retrieved after wet and dry seed dressing and transferred to PDA for testing their viability. The results were presented in the Table 2

Table 2: Effect of seed treatment with fungicides on the sclerotial viability of *Rhizoctonia solani*

S. No	Fungicide	Dry seed treatment, dosage/kg of seed	Per cent germination of sclerotia in dry seed treatment	Wet seed dressing, dosage/l of water	Per cent germination of sclerotia in wet seed treatment
1	Carbendazim 50 WP	3.0 g	0.00	1.0 g	0.00
2	Propiconazole 25 EC	3.0 ml	0.00	1.0 ml	0.00
3	Hexaconazole 5 EC	6.0 ml	0.00	2.0 ml	0.00
4	Validamycin 3 %L	6.0 ml	0.00	2.0 ml	0.00
5	Tebuconazole 25 EC	4.5 ml	0.00	1.5 ml	0.00
6	Thifluzamide 24 SC	3.0 ml	0.00	1.0 ml	0.00
7	Azoxystrobin 23SC	2.0 g	0.00	1.0 g	0.00
8	Difinoconazole 25EC	2.0 g	0.00	1.0 g	0.00
9	Tebuconazole 50% + Trifloxystrobin 25% WG	2.0 g	0.00	0.8 g	0.00
10	Untreated control	--	100.00	--	100.00

****** Figures in parentheses are angular transformed values.

All the sclerotial bodies germinated in the control. All the fungicide treatments both in wet and dry seed treatments showed 100 % inhibition of sclerotial germination. Fig1 and 2.

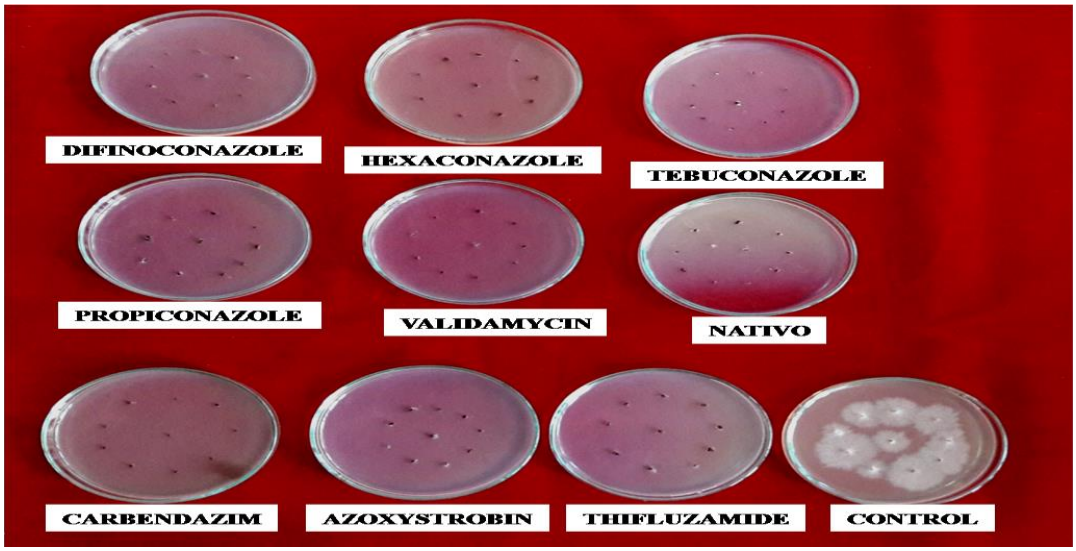

Fig 1: Effect of dry seed treatment with fungicides on viability of sclerotia admixtured with seed

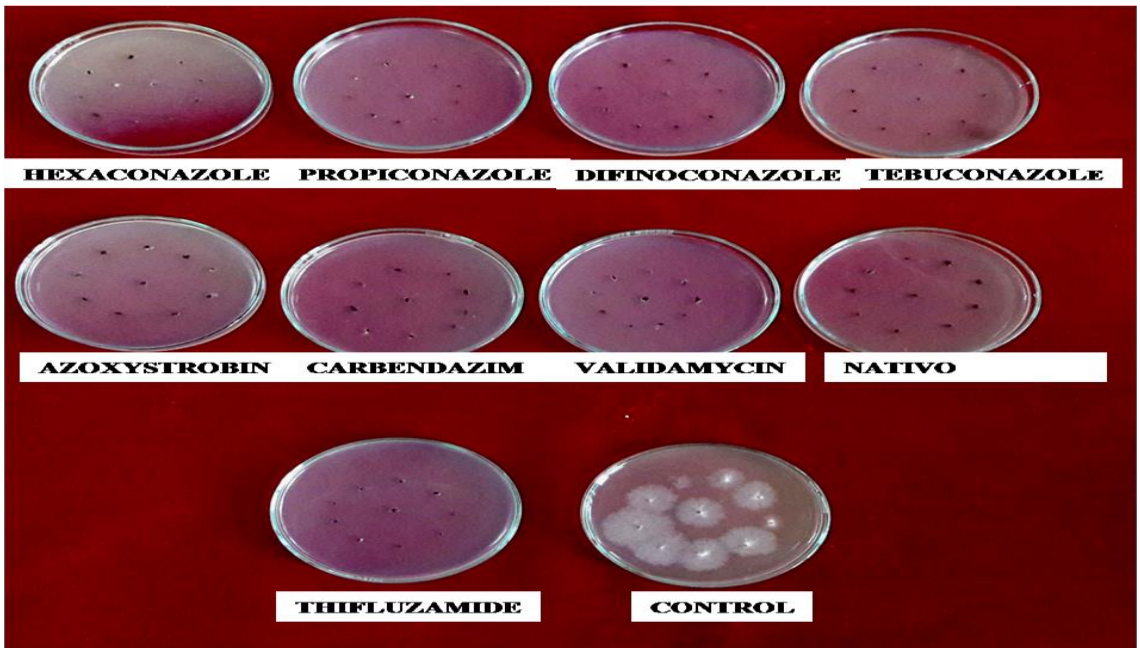

Fig 2: Effect of wet seed treatment with fungicides on viability of sclerotia admixtured with seed

Effect of dry seed treatment with fungicides on the seedling growth in rolled paper towel method.

After retrieval of sclerotia, the seed in the particular lot were kept for germination by following paper towel method. After 10 d the seedling growth was measured. The data was presented in the Table 3 and Fig 3. Seed germination was 100 per cent.

Table 3: Effect of dry seed treatment with fungicides on the seedling growth in paper towel method

S. No.	Fungicide	Dry seed dressing dosage/kg seed	Root length of seedling	Shoot length of seedling
			(mean of 10 seedlings)	
1	Carbendazim 50WP	3.0 g	5.80 (13.93)**	4.23 (11.86)
2	Propiconazole 25EC	3.0 ml	8.23 (16.67)	4.17 (11.77)
3	Hexaconazole 5EC	6.0 ml	8.56 (17.00)	4.80 (12.65)
4	Validamycin 3%L	6.0 ml	8.10 (16.53)	5.43 (13.47)
5	Tebuconazole 25EC	4.5 ml	7.00 (15.33)	5.73 (13.85)
6	Thifluzamide 24 SC	3.0 ml	6.26 (14.47)	5.57 (3.64)
7	Azoxystrobin 23SC	2.0 g	7.33 (15.70)	5.13 (13.09)
8	Difinoconazole 25EC	2.0 g	9.00 (17.45)	7.97 (16.39)
9	Tebuconazole 50 % + Trifloxystrobin 25% WG	2.0 g	6.66 (14.95)	4.03 (11.58)
10	Untreated control	--	6.40 (14.64)	4.33 (12.00)
	CD (P=0.01)		0.59	0.55
	SEm±		0.19	0.18
	SEd±		0.28	0.26
	CV (%)		2.19	2.44

** Figures in parentheses are angular transformed values

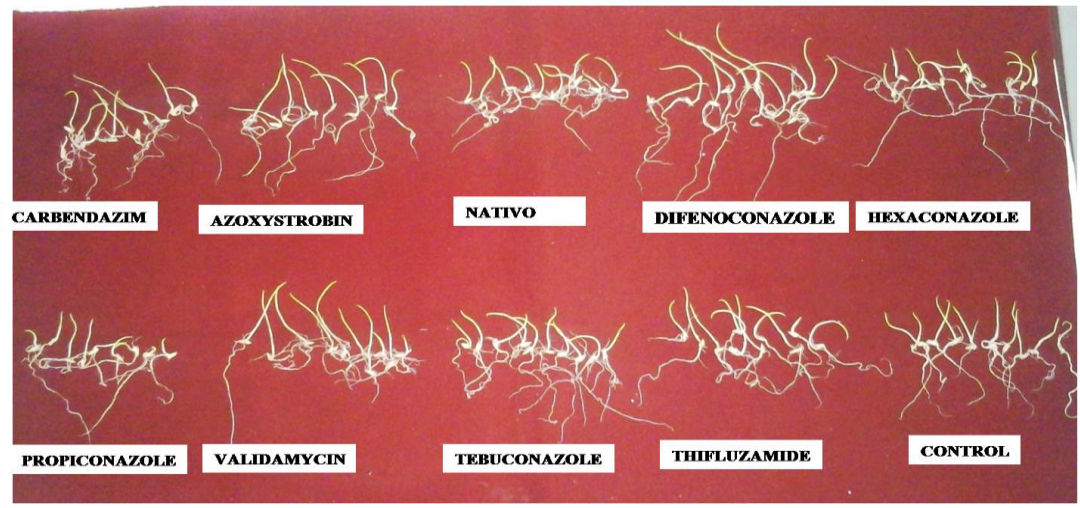

Fig 3: Efficacy of dry seed treatment with different fungicides on seedling growth in paper towel method

Root length (cm): At 10 DAS, significant differences were observed in the root length among the treatments. All the treatments are superior over control (6.4 cm) except Carbendazim (5.8 cm). The root length among the treatments varied from 5.8 cm (Carbendazim) to 9.0 cm (Difenoconazole).
Among the treatments Difenoconazole (9.0 cm) and Hexaconazole (8.56 cm) are on par but significantly superior over other treatments in increasing root length. Propiconazole (8.23 cm) and Validamycin (8.10 cm) are on par but significantly higher than other treatments Azoxystrobin (7.33 cm), Tebuconazole (7.00 cm), Tebuconazole 50% + Trifloxystrobin 25% (6.66 cm), Thifluzamide (6.26 cm) and Carbendazim (5.8 cm).

Shoot length (cm): All the treatments are superior over control (4.33 cm) except Carbendazim (4.23 cm), Propiconazole (4.17 cm) and Tebuconazole 50% + Trifloxystrobin 25% (4.03 cm). Difenoconazole (7.97 cm) was significantly superior to all other treatments followed by Tebuconazole (5.73 cm), Thifluzamide (5.57cm) and Validamycin (5.43 cm) which are statistically on par. Effect of Azoxystrobin (5.13 cm) is significantly higher than other treatments Carbendazim (4.23 cm), Propiconazole (4.17 cm) and Tebuconazole 50% + trifloxystrobin 25% (4.03 cm).

Effect of wet seed treatment with fungicides on the seedling growth in rolled paper towel method.
After retrieval of sclerotia, the seed in the particular lot were kept for germination by following paper towel method. After 10 days the seedling growth was measured. The data was presented in the Table 4 and Fig 4.

Table 4: Effect of wet seed treatment with fungicides on the seedling growth in paper towel method

S. No	Fungicide	Wet seed dressing dosage/ lit of water	Root length of seedling	Shoot length of seedling
			(mean of 10 seedlings)	
1	Carbendazim 50 WP	1.0 g	5.43 (13.47)**	5.87 (14.01)
2	Propiconazole 25 EC	1.0 ml	6.55 (14.82)	5.50 (13.55)
3	Hexaconazole 5 EC	2.0 ml	7.60 (15.99)	6.17 (14.37)
4	Validamycin 3 % L	2.0 ml	5.68 (13.78)	5.93 (14.09)
5	Tebuconazole 25 EC	1.5 ml	5.23 (13.21)	5.63 (13.71)
6	Thifluzamide 24 SC	1.0 ml	6.83 (15.14)	7.20 (15.56)
7	Azoxystrobin 23 SC	1.0 g	6.88 (15.19)	5.30 (13.29)
8	Difinoconazole 2 5EC	1.0 g	6.59 (14.87)	7.80 (16.21)
9	Tebuconazole 50% + Trifloxystrobin 25% WG	0.8 g	5.45 (13.49)	5.40 (13.42)
10	Untreated control	--	6.40 (14.62)	5.50 (13.53)
	CD (P=0.01)		0.86	0.97
	SEm±		0.29	0.32
	SEd±		0.40	0.46
	CV (%)		3.46	3.98

****** Figures in parentheses are angular transformed values

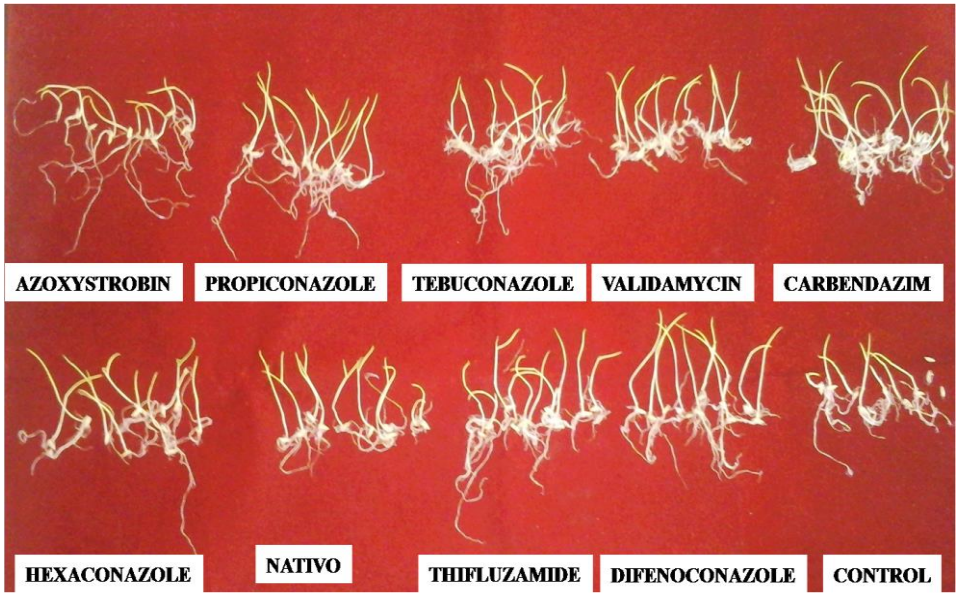

Fig 4: Efficacy of wet seed treatment with different fungicides on seedling growth in paper towel method

Root length (cm): In wet seed treatment Hexaconazole (7.6 cm) was significantly higher in promoting root length than all treatments followed by Azoxystrobin (6.88 cm), Thifluzamide (6.83 cm), difinoconazole (6.59 cm), Propiconazole (6.55 cm) and control (6.4 cm) which are on par but significantly higher than Validamycin (5.68 cm), Tebuconazole 50% + Trifloxystrobin 25% (5.45 cm), Carbendazim (5.43 cm) and Tebuconazole (5.23 cm).

Shoot length (cm): With regard to shoot length, all the treatments are superior over control (5.50 cm) except Tebuconazole 50% + Trifloxystrobin 25% (5.4 cm),

Azoxystrobin (5.3 cm). Difenoconazole (7.8 cm) and Thifluzamide (7.2 cm) are on par and significantly higher than other treatments, followed by Hexaconazole (6.17 cm), Validamycin (5.93 cm), Carbendazim (5.87 cm), Tebuconazole (5.63 cm), Propiconazole (5.55 cm) and control (5.55 cm) which are on par but significantly higher than Tebuconazole 50% + Trifloxystrobin 25% (5.4 cm) and Azoxystrobin (5.3 cm).

Effect of dry seed treatment with fungicides on the seedling growth in pot culture method.

After retrieval of sclerotia, the seed in the particular lot were kept for germination in pots. After 10 days the seedling growth was measured. The data was presented in Table 5 and Fig 5.

Table 5: Effect of dry seed treatment with fungicides on the seedling growth in pot culture method

S. No	Fungicide	Dry seed dressing dosage/kg seed	Root length of seedling	Shoot length of seedling
			(mean of 10 seedlings)	
1	Carbendazim 50WP	3.0 g	5.10 (13.04)**	6.33 (14.56)
2	Propiconazole 25EC	3.0 ml	6.50 (14.76)	8.83 (17.28)
3	Hexaconazole 5EC	6.0 ml	9.33 (17.77)	7.50 (15.88)
4	Validamycin 3%L	6.0 ml	9.66 (18.10)	11.66 (19.96)
5	Tebuconazole 25EC	4.5 ml	7.16 (15.52)	8.16 (16.59)
6	Thifluzamide 24 SC	3.0 ml	7.66 (16.06)	9.33 (17.77)
7	Azoxystrobin 23SC	2.0 g	4.60 (12.36)	7.93 (16.35)
8	Difinoconazole 25EC	2.0 g	8.83 (17.28)	7.83 (16.24)
9	Tebuconazole 50 % + Trifloxystrobin 25% WG	2.0 g	4.46 (12.18)	5.66 (13.75)
10	Untreated control	--	9.5 (17.94)	6.96 (15.29)
	CD (P=0.01)		0.93	0.79
	SEm±		0.31	0.26
	SEd±		0.44	0.37
	CV (%)		3.50	2.83

****** Figures in parentheses are angular transformed values.

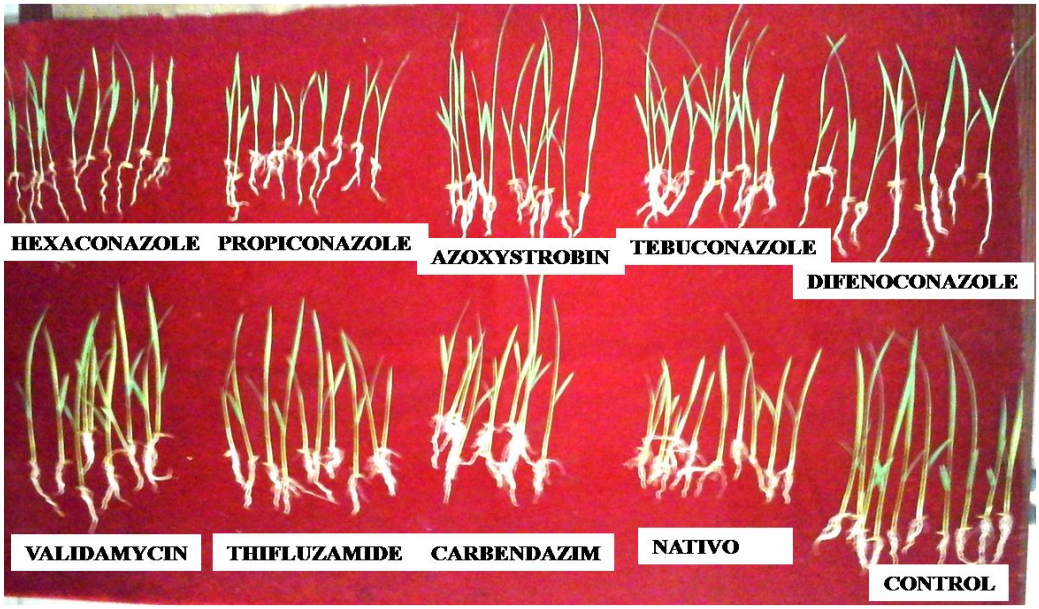

Fig 5: Efficacy of dry seed treatment with different fungicides on seedling growth in pot culture method

Seed Germination: 100 per cent seed germination was recorded.

Root length (cm): There is no significant difference with regard to root length among the treatments, Validamycin (9.66 cm), control (9.5 cm), Hexaconazole (9.33 cm), difinoconazole (8.83 cm) which are followed by Thifluzamide (7.66 cm), Tebuconazole (7.16 cm), Propiconazole (6.5 cm), Carbendazim (5.1 cm), Azoxystrobin(4.6 cm) and Tebuconazole 50% + Trifloxystrobin 25% (4.46 cm).

Shoot length (cm): Among all the treatments, shoot lengths in the treatments of Validamycin (11.6 cm), Thifluzamide (9.33 cm), Propiconazole (8.83 cm), Tebuconazole (8.16 cm), Azoxystrobin (7.9 cm), Hexaconazole (7.5 cm) were significantly higher than control (6.97 cm). Carbendazim (6.33 cm) is on par with control and Tebuconazole 50% + Trifloxystrobin 25% (5.66 cm) is significantly lower than control (6.9 cm).

Effect of wet seed treatment with fungicides on the seedling growth in pot culture method.

After retrieval of sclerotia, the seed in the particular lot were kept for germination in pots. After 10 d the seedling growth was measured. The data was presented in the Table 6 and Fig 6. 100 per cent seed germination was recorded.

Table 6: Effect of wet seed treatment with fungicides on the seedling growth in pot culture method

S. No.	Fungicide	Wet seed dressing dosage/ lit of water	Root length of seedling	Shoot length of seedling
			(mean of 10 seedlings)	
1	Carbendazim 50 WP	1.0 g	4.66 (12.47)**	11.83 (20.11)
2	Propiconazole 25 EC	1.0 ml	5.66 (13.75)	12.83 (20.97)
3	Hexaconazole 5 EC	2.0 ml	6.16 (14.37)	13.5 (21.54)
4	Validamycin 3 % L	2.0 ml	6.16 (14.37)	13.33 (21.40)
5	Tebuconazole 25 EC	1.5 ml	5.83 (13.97)	13.16 (21.26)
6	Thifluzamide 24 SC	1.0 ml	4.66 (12.45)	12.67 (20.80)
7	Azoxystrobin 23 SC	1.0 g	4.83 (12.67)	12.00 (20.24)
8	Difinoconazole 25 EC	1.0 g	6.00 (14.16)	12.50 (20.11)
9	Tebuconazole 50 % + Trifloxystrobin 25 % WG	0.8 g	5.23 (13.22)	10.16 (18.57)
10	Untreated control	--	4.50 (12.23)	14.83 (22.64)
	CD (P=0.01)		1.04	1.37
	SEm±		0.35	0.46
	SEd±		0.49	0.65
	CV (%)		4.54	3.83

****** Figures in parentheses are angular transformed values.

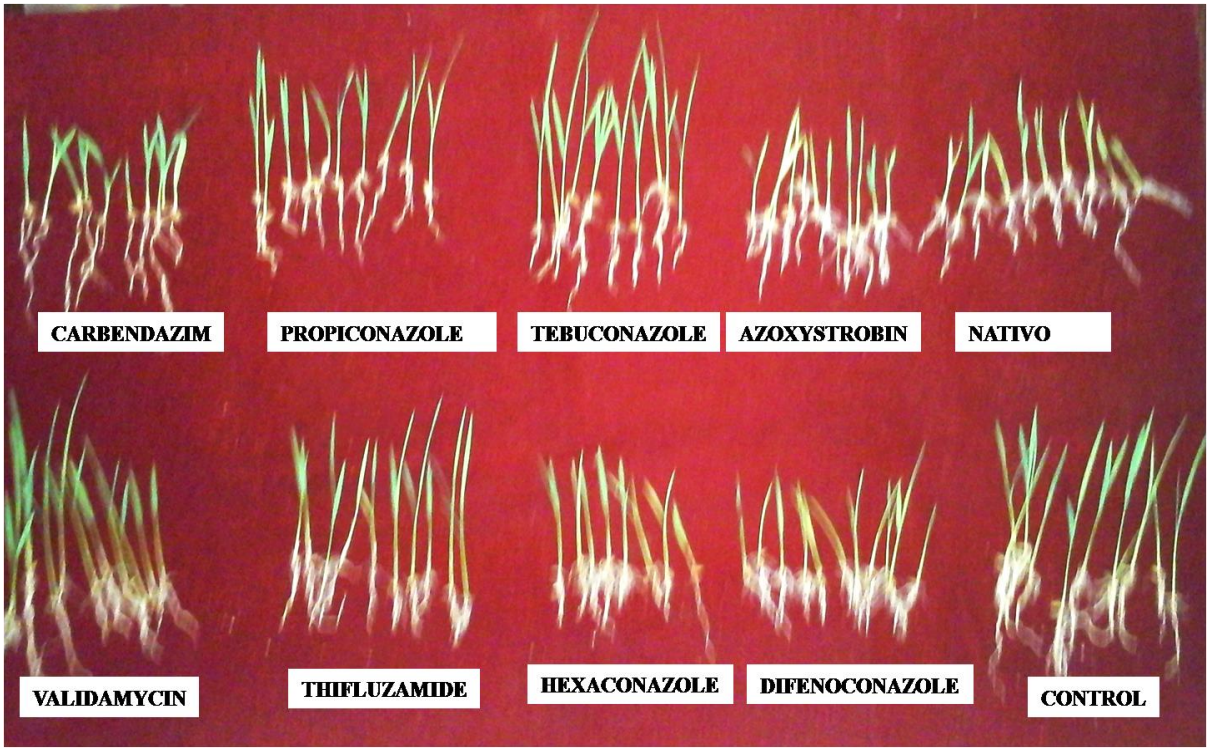

Fig 6: Efficacy of wet seed treatment with different fungicides on seed ling growth in pot culture method

Root length (cm): Among all the treatments root lengths in the treatments of Validamycin (6.16 cm), Hexaconazole (6.16 cm), difinoconazole (6.00 cm), Tebuconazole (5.83 cm), Propiconazole (5.66 cm) are significantly higher than control (4.5 cm) and there is no significant difference among Tebuconazole 50% + Trifloxystrobin 25% (5.2 cm), Azoxystrobin (4.8 cm), Carbendazim (4.66 cm), Thifluzamide (4..66 cm) and control (4.5 cm).

Shoot length (cm): Among all the treatments there is no significant difference in shoot lengths between control (14.88 cm)**,** Hexaconazole (13.5 cm), Validamycin (13.33 cm), Tebuconazole (13.16 cm). The shoot length in the treatments of Propiconazole (12.83 cm), Thifluzamide (12.66 cm), difinoconazole (12.50 cm), Azoxystrobin (12.00 cm), Carbendazim (11.83 cm), Tebuconazole 50% +trifloxystrobin 25% (10.16 cm) are significantly lower than control (14.88 cm).

Ali *et al*. (2002) [2] studied the effect of seed treatment on the management of sheath blight. On transplanted crop, three sprays of bavistin 0.1% and mancozeb 0.1% cum seed treatments proved to be more effective.

Efficacy of fungicides Flutalonil, Pencycuron and Validamycin alone and in combination with biocontrol agent filtrate against *R. solani* were tested. Results showed that growth of the pathogen was almost completely inhibited by the tested fungicides at 50µg/ml respectively. The sclerotial germination after 72 h of treatment with the fungicides at 25 ^{0}C was reduced at 200µg/ml (Nobutaka someya *et al*., 2005) [7].

Bag, 2007 evaluated the efficacy of a new fungicide – a combination of two systemic fungicides viz., Trifloxystrobin 25% (Strobilurin compound) and Tebuconazole 50% (Triazole compound) along with two other commercially available fungicides Hexaconazole and Validamycin under challenge inoculation condition. The new fungicide was most

effective in decreasing disease severity (37.61% lower over control).

Various fungicides, insecticides, nematicides and herbicides were tested against *R. solani* (Kumhar and Tripathi, 2007) [5] in which Thiophanate – methyl, Carbendazim, MEMC and Carboxin were found to be effective in inhibiting the mycelial growth by more than 95 per cent at 200 ppm and herbicides also affected the mycelial growth drastically.

Sawai Boukaew *et al.,* (2013) [10] observed that sclerotial germination of *R. solani* was 100 per cent inhibited by the chemical fungicides viz., Carbendazim, Validamycin, Propicanozole, Mancozeb after soaking for more than 12 h.

The above results are in agreement with the present study and fungicides used in this study are found to be effective in inhibiting sclerotial germination of *R. solani.*

Conclusions

When the seed admixtured with sclerotia was treated with different fungicides, effect on sclerotial viability was tested, all the fungicides inhibited the germination of sclerotia when compared to control. The effect of sclerotia on germination and seedling growth was tested by paper towel and pot culture methods. All the seeds germinated in both paper towel and pot culture methods. The efficacy of fungicides on seedling growth varied in both wet and dry seed treatments. All the tested fungicides showed increased seedling growth (root length and shoot length) except carbendazim and tebuconazole 50% + trifloxystrobin 25% (Nativo) when compared to control in both paper towel and pot culture methods.

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