

Survey and evaluation of efficacy of some fungicides against virulence of *Sarocladium oryzae* inciting sheath rot disease of rice

R Naveenkumar^{1*} and R Mohanpriya²

¹Banaras Hindu University, Varanasi-221005, UP, India

²S Thangapazham Agriculture College, Vasudevanallur, Tirunelveli-627758, TN, India

*Corresponding author e-mail: pathonaveen92@yahoo.com

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ABSTRACT

A field survey was conducted to assess the extent of sheath rot occurrence of rice in selected rice growing areas from different districts of Tamil Nadu during the year of 2014-2015. Among the different locations surveyed for rice sheath rot incidence, highest disease incidence were found in Orathur (30.5%) village followed by Mutharasanallur (27.4%), Thennamanadu (24.5%) and the least incidence was found in Vennankuzhi (12.1%). All the isolates were inoculated into tillers of rice plants (IR36) to assess the pathogenecity. Among the different isolates of *S. oryzae* disease incidence were produced which were varied between 36.33 and 72.36%. Efficacy of seven different chemical fungicides such as Antracol, Aytar, ICF-310, Indofil M-45, Indofil Z-78, Merger and Saaf were screened against *S. oryzae* pathogen by using poisoned food technique under in vitro condition. All the fungicides were tested at different concentration such as 100, 200 and 300ppm level. Result of the present study revealed that, fungicides antracol, aytar, ICF-310 and merger were completely inhibited the mycelial growth of the test pathogen even at 300ppm concentration whereas rest of the fungicides were not resulted in complete mycelial growth at 300ppm concentration.

Key words: Disease, fungicides, pathogenecity, rice, *Sarocladium oryzae*, sheath rot

Rice, an important crop worldwide, is serving as the staple food for half of the population and additionally being used in industry and for animal feed. Rice is grown in various agro-ecological zones in tropical and subtropical areas, especially in Asia. Asian continent alone accounting 90% of the world rice production (IRRI 2015a). Irrigated rice is prone to attack by more than 100 species of insects and diseases. Together, pest and disease accounts for 30 to 40 per cent crop losses (Kindo *et al.* 2015). The crop suffers with a number of diseases caused by fungi, bacteria, viruses, nematodes and mycoplasma like organism (Karmakar *et al.* 2016). The crop has been affected by as many as fungal diseases, among them sheath rot has gained the status as a major disease of rice (Reddy and Ghosh 1985) and yield losses varies from 9.6 to 85% depending on the weather conditions during the crop growth period (Phookan and Hazarika 1992). In general, *S. oryzae* is present in all

rice-growing countries worldwide, being very common in rainy seasons (Mew and Gonzales 2002; Naeimi *et al.* 2003).

The major symptoms describing rice sheath rot caused by *S. oryzae* are as following, the rot occurs on the upper most leaf sheaths enclosing the young panicles; the lesions start as oblong or somewhat irregular spots, 0.5-1.5 cm long, with brown margins and grey centers, or they may be greyish brown throughout; they enlarge and often coalesce and may cover most of the leaf sheath; the young panicles remain within the sheath or only partially emerge; an abundant whitish powdery growth may be found inside affected sheaths and young panicles are rotted (Ou 1985). *S. oryzae* infection results in chaffy, discoloured grains, and affects the viability and nutritional value of seeds (Sakthivel 2001; Gopalakrishnan *et al.* 2010).

Control of fungal diseases through chemotherapy is receiving much attention because of its efficacy and success rate, and not having any phytotoxic effect on the plant vigour and other components of the ecosystem (Ibiam *et al.* 2008). In the past, several fungicides have been employed in the control of fungal diseases of rice and other crops. Agrochemicals like Benlate, Bavistin, Apron Plus 50 DS, Fernasan-D and Dithane M-45 were used to control a number of diseases caused by fungal pathogens of rice (Ibiam *et al.* 2008). Lakshmanan and Mohan (1988) reported that mancozeb (Dithane M-45) has been used solely, and in combination with other fungicides for the control of fungal diseases of rice. Few workers reported that to control the sheath rot disease by using the fungicides like Benomyl 50% WP or Mancozeb 80%, WP (Mishra *et al.* 1994); Dithane M-45, Ridomil, Carbendazim (Arshad *et al.* 2009); Carbendazim 50% WP and Carbendazim 64% +Mancozeb 8% WP (Bag *et al.* 2010; Bag and Saha 2009). However, more investigations are required for this disease, otherwise it will be a serious problem in future.

Hence, the objectives of present investigation are as follows, (1) To assess the prevalence and incidence of sheath rot diseases of rice from selected rice growing tracts of Tamil Nadu, India during 2014-2015, (2) To assess the cultural characters and pathogenic variability among the isolates of *Sarocladium oryzae* (SR) and (3) To test the *in vitro* efficacy of new fungicides against the test pathogen.

A field survey was conducted to assess the extent of sheath rot occurrence of rice in selected rice growing areas of Tamil Nadu (Ariyalur, Cuddalore, Karur, Nagapattinam, Tanjavore, Thiruvarur and Trichy) during the year of 2014-2015. The village where rice is traditionally grown was selected for assessing the prevalence of sheath rot disease caused by *S. oryzae*. Diseased plants showing sheath rot symptom were collected from the field and the samples were kept in clean polythene bag and each sample was marked clearly to show details of the location and variety. The samples were brought to the laboratory for microscopic examination, isolation, purification and pathogenicity test. For disease scoring, the typical assessment system for rice developed by the IRRI 1996 was followed.

The pathogen was isolated from the infected sheaths rice plants showing the typical rot symptoms by tissue segment method (Rangaswami 1972) on potato dextrose agar (PDA) medium. The axenic cultures of the pathogen were obtained by single hyphal tip method (Rangaswami 1972) and these were maintained on PDA slants. The morphological and cultural characterizations of the isolates grown on PDA were studied and compared with those mentioned by Booth (1971). Totally 15 isolates were obtained and maintained on PDA slants.

To test pathogenicity, rice cultivar IR36 plants were grown in the green house. At booting stage and the panicle-emerging stage, tillers were inoculated with *S. oryzae* following the standard grain inoculation technique (Sakthivel and Gnanamanickam 1987). A total of 45 tillers from 15 plants were inoculated with each isolate of *S. oryzae*. The incidence of sheath rot disease was recorded after 14 days using the standard evaluation system for rice (IRRI 1996).

Sterile PDA media was poured into sterile petri dishes and allowed to solidify. A nine mm culture disc of *S. oryzae* obtained from actively growing region was aseptically placed at the centre of the dish and incubated. The radial growth and colour of the mycelium (in mm) was measured seven days after inoculation.

Efficacy of commonly available seven different new fungicides were assessed against *S. oryzae* by radial growth assay following poisoned food technique (Table 3) (Nehal and EI-Mougy 2009). The fungicides were tested in the concentration range of 100-300ppm (w/v). 50 ml of sterilized PDA, different concentration of fungicides were mixed separately and dispensed to sterile Petri dish. All the plates were gently rotated for even dispersal of fungicides. Plates without fungicides were served as control. Eight mm discs of the test fungi taken from the advancing edge of test pathogens were placed in fungicide containing PDA plates and incubated at $28 \pm 2^\circ$ C. Three replicates for each treatment were maintained. The observations for mycelial growth were recorded at every 5 days interval up to 15 days of the inoculation. The percent inhibition was calculated as per the following formula described by Vincent (1947).

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

Whereas, C = Diameter of the fungus colony

(mm) in control plate. T = Diameter of the fungus colony (mm) in treated plate.

All the experiments were of completely randomized design (CRD) and repeated twice. Data were subjected to analyses of variance and treatment means were compared by an appropriate Duncan's multiple range test ($P < 0.05$). The IRRISTAT package version 92-1, developed by the International Rice Research Institute Biometrics Unit, Philippines, was used for analysis (Gomez and Gomez 1984).

Totally 15 isolates were obtained from different rice grown regions of Tamil Nadu and they were subjected to identification (Table 1). The results revealed that all the isolates belong to *Sarocladium oryzae*. The isolate number was designated as SR1-SR15.

In irrigated rice, the severity of sheath rot disease was found to vary significantly with respect to location from selected districts of Tamil Nadu. Among the different locations surveyed for rice sheath rot incidence, highest disease incidence were found in Orathur (30.5%) village followed by Mutharasanallur (27.4%), Thennamanadu (24.5%) and the least incidence was found in Vennankuzhi (12.1%). Whereas rest of the villages recorded moderate disease incidence (Table 1). In India, Reddy *et al.* (2001) reported a higher incidence and severity of sheath rot disease in rain-fed rice than in irrigated rice. However, little information is available regarding seasonal variations and the

occurrence of this disease. Chowdhury *et al.* (2015) conducted the survey to assess the disease severity of sheath rot disease of rice both irrigated and rain-fed conditions from Bangladesh, and concluded that the severity of sheath rot incidence was higher in rainfed rice than the irrigated rice. The above findings were in conformity of the present studies.

Mycelial growth of the *Sarocladium oryzae* on PDA medium was varied location to location. All the fifteen isolates of *S. oryzae* were identified based on morphological characteristics (white cottony aerial mycelium, irregularly branched conidiophores with one or two whorls of appressed branches and fusiform, curved, hyaline, single-celled conidia). Chowdhury *et al.* (2015) reported mycelial growth of 29 representative isolates was found to vary on PDA and the isolates were divided into 6 groups. The range of the overall size of conidia of the selected isolates was 2.40-7.20 x 1.20-2.40 μm .

Fourteen days after inoculation, *S. oryzae* inoculated tillers of rice plants (IR36) were produced typical sheath rot lesions on the uppermost flag leaf enclosing the panicle. Variable disease incidence (36.33-68.63%) produced by different test isolates of *S. oryzae* were observed (Table 2). Results of the present study are in accordance with Ayyadurai *et al.* (2005). Ayyadurai *et al.* (2005) assessed the virulence of 32 isolates of *S. oryzae* pathogen from different locations of India and reported 45-98% disease incidence

Table 1. Survey on the incidence of sheath rot of rice in selected district of Tamil Nadu

Sl. No.	District	Village	Crop stage	Soil type	Sheath rot Incidence (%)
1	Ariyalur	Meensuruty	Panicle initiation	Clay loam	18.5 ^c
2		Vennankuzhi	Panicle initiation	Alluvial	12.1 ⁿ
3	Cuddalore	Sivapuri	Panicle initiation	Clay loam	17.1 ^f
4		Perampattu	Panicle initiation	Clay	14.0 ^k
5		Orathur	Panicle initiation	Clay	30.5 ^a
6	Karur	Puliyur	Panicle initiation	Clay loam	13.2 ^l
7	Nagapattinam	Kollidam	Grain filling	Sandy loam	16.5 ^g
8		Thaikkal	Grain filling	Clay loam	15.0 ⁱ
9		Thandavankulam	Panicle initiation	Sandy loam	19.4 ^d
10	Tanjavur	Thennamanadu	Panicle initiation	Clay	24.5 ^c
11		Valamarkottai	Panicle initiation	Clay loam	8.5 ^o
12	Thiruvarur	Palliyamangalam	Panicle initiation	Clay loam	12.5 ^m
13		Karumbiyur	Grain filling	Clay	14.6 ^j
14	Trichy	Vayalur	Grain filling	Clay loam	16.2 ^h
15		Mutharasanallur	Grain filling	Clay	27.4 ^b

**Mean of three replications

*Value within a column with same letter do not differ significantly according to the DMRT method ($P = 0.05$)

variability among the different isolates of *S. oryzae*.

Efficacy of seven different chemical fungicides were evaluated against *S. oryzae* pathogen by using poisoned food technique under *in vitro* condition. All the fungicides were tested at different concentration viz., 100, 200 and 300 ppm. Among the fungicides, antracol, avtar, ICF-310 and merger completely inhibited the mycelial growth of the test pathogen at 300ppm concentration (Table 4). However the fungicides such as Indofil M-45, Indofil Z-78 and Saaf did not inhibit complete mycelial growth even at 300ppm concentration (Fig. 1). Similar observation was made by Karmakar *et al.* (2016) and reported Trifloxystrobin + Tebuconazole, Carbendazim+Mancozeb,

Table 2. Pathogenicity of *Sarocladium oryzae* isolates

Sl.No	Isolate number	Sheath rot incidence (%) ** 15 DAI
1	SR1	58.93d
2	SR2	38.69h
3	SR 3	56.63d
4	SR 4	41.33gh
5	SR 5	72.36a
6	SR 6	39.33h
7	SR 7	52.66e
8	SR 8	47.36f
9	SR 9	62.39c
10	SR 10	63.63c
11	SR 11	36.33i
12	SR 12	39.66h
13	SR 13	43.63g
14	SR 14	49.39f
15	SR 15	68.63b

DAI- Days after Inoculation

**Mean of three replications

*Value within a column with same letter do not differ significantly according to the DMRT method (P=0.05)

Table 4. Efficacy of fungicides against *S. oryzae* under *in vitro* condition

Tr. No.	Fungicides	Radial growth (mm)					
		100 ppm	Per cent reduction over control	200 ppm	Per cent reduction over control	300 ppm	Per cent reduction over control
1	Antracol	22.2 ^c	62.17	10.6 ^b	81.33	00 ^a	100
2	Avtar	18.6 ^b	68.26	9.4 ^b	83.45	00 ^a	100
3	Indofil- M 45	30.6 ^{de}	47.80	23.2 ^c	59.15	17.4 ^c	70.10
4	Indofil- Z 78	32.4 ^e	44.70	25.8 ^d	54.57	18.2 ^c	68.72
5	ICF 310	19.4 ^b	66.90	10.6 ^b	82.04	00 ^a	100
6	Merger	14.6 ^a	75.08	6.8 ^a	88.02	00 ^a	100
7	Saaf	28.2 ^d	51.88	22.6 ^c	60.21	15.6 ^b	73.20
8	Control	58.6 ^f	-	56.8 ^e	-	58.2 ^d	-

**Mean of three replications

*Value within a column with same letter do not differ significantly according to the DMRT method (P=0.05)

Table 3. List of fungicides used for the present study

Sl. No.	Trade Name	Active Ingredient
1	Antracol	Propineb 70 WP (70% w/w)
2	Avtar	Zineb 68% + Hexaconazol 4% WP
3	ICF -310	Mancozeb 68% + Hexaconazole 4%
4	Indofil M-45	Mancozeb 75% WP
5	Indofil Z-78	Zineb 75% WP
6	Merger	Tricyclazole 18 % + Mancozeb 62 % WP
7	Saaf	Carbendazim 12% + Mancozeb 63% WP

Tricyclazole+Mancozeb, Tricyclazole, Propiconazole+Difeconazole had shown complete inhibition of growth of *S. oryzae* both in vitro and in vivo conditions. Among the four highly effective test fungicides Merger performed best and it is in combination of tricyclazole and mancozeb. Tricyclazole is highly effective fungicide against other important foliar rice disease particularly blast of rice caused by *P. grisea* (Dutta *et al.* 2012). Other two effective fungicides, viz., Avatar and ICF-310 have hexaconazole which is triazole group fungicide and dithiocarbamate group fungicides, efficiently control many other phytopathogenic fungi causing rice diseases. Dodan *et al.* (1996) also reported the efficacy of carbendazim, propiconazole, mancozeb, edifenphos and tricyclazole in controlling sheath rot caused by *S. oryzae*. The above findings are agreement with the results of the present studies.

REFERENCES

- Arshad HMI, Khan JA, Naz S, Khan SN and Akram M 2009. Grain discoloration disease complex: A new threat for rice crop and management. Pak. J. Phytopathology 21: 31-36
- Ayyadurai N, Kirubakaran SI, Srisha S and Sakthivel N 2005. Biological and molecular variability of *Sarocladium*

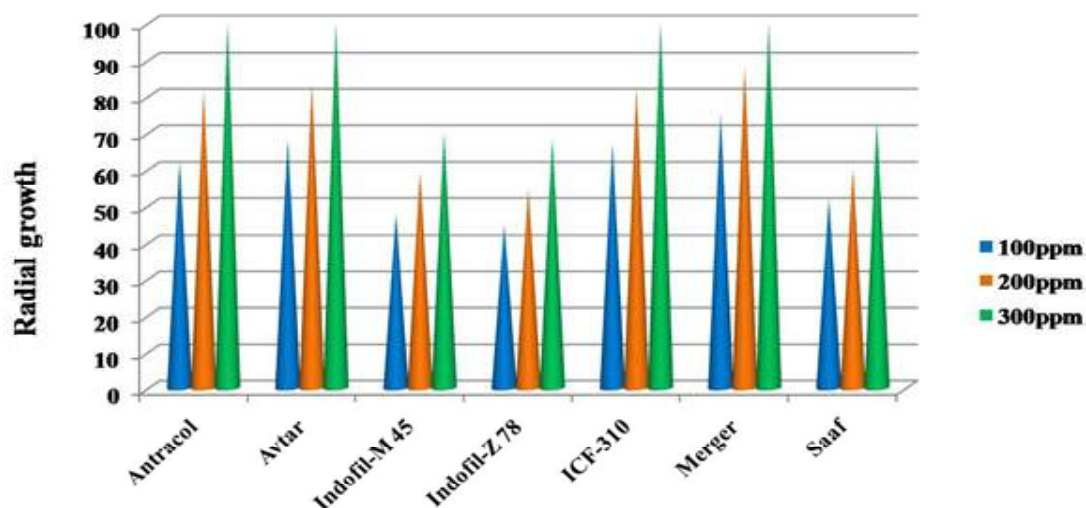


Fig. 1. Efficacy of percent inhibition of fungicides against *S. oryzae* under *in vitro* condition

- oryzae*, the sheath rot pathogen of rice (*Oryza sativa* L.). Curr. Microbiol. 50: 319-323
- Bag MK, Adhikari B and Bhowmik MK 2010. Evaluation of fungitoxic effect of some commercially available agrochemicals against discoloration (Gd) disease of rice in West Bengal. The Journal of Plant Protection Science 2(1): 103-104
- Bag MK and Saha S 2009. Fungitoxic effect of Nativo 75 wg (trifloxystrobin 25% + tebuconazole 50%) on grain discoloration (gd) disease of rice in West Bengal. Pestology 33(7): 47 - 49
- Booth C 1971. The genus *Fusarium*. Common Wealth Mycological Institute, Kew, Surrey, England, pp. 237
- Chowdhury MTI, Mian MS, Taher Mia MA, Rafii MY and Latif MA 2015. Agro-ecological variations of sheath rot disease of rice caused by *Sarocladium oryzae* and DNA fingerprinting of the pathogen's population structure. Genetics and Mol. Res. 14 (4): 18140-18152
- Dodan DS, Singh R, Sunder S and Singh R 1996. Efficacy of fungi toxicants against sheath rot of rice. Indian J. Mycology and Pl. Pathol. 26 (3): 283-284
- Dutta D, Saha S, Ray DP and Bag MK 2012. Effect of different active fungicides molecules on the management of rice blast disease. International Journal of Agriculture, Environment and Biotechnology 5(3): 247-251
- Gomez KA and Gomez AA 1984. Statistical procedures for agricultural research. 2nd edn. John Wiley and Sons, New York
- Gopalakrishnan C, Kamalakannan A and Valluvaparidasan V 2010. Survey of seed-borne fungi associated with rice seeds in Tamil Nadu, India. Libyan Agriculture Res. Center J. Int. 1(5): 307-309
- Ibiam OFA, Umechuruba CI and Arinze AE 2008. A survey of seed borne fungi associated with seeds of rice (*Oryza sativa* L. FARO 12, 15 and 29) in storage and the field in Afikpo North local Government area of Ebonyi State. Scientia Africana 7(2): 1-4
- IRRI (1996). International Rice Research Institute (IRRI). Annual report of rice. pp. 25
- IRRI (2015a). World Rice Statistics in 2013. Available at: <http://ricestat.irri.org>
- Karmakar S, Mondal R, Dasgupta S, Guha P, and Mandal AK 2016. Evaluation of Some Newly Evolved Fungicides against *Helminthosporium oryzae*, *Alternaria padwickii*, *Fusarium moniliforme*, *Curvularia lunata* and *Sarocladium oryzae* Causing Grain Discoloration Disease of Rice Under In Vitro condition. Intern. J. Sci. Res. 5(6): 1108-1111
- Kindo D, Bhagat RK and Tiwari PK 2015. Efficacy of fungicides for the management of sheath rot disease in rice under in vitro and in vivo conditions. Oryza 52(3): 227-230
- Lakshmanan P and Mohan S 1988. Effect of seed treatments on brown spot disease and their influence on rice seedlings. Madras Agricultural Journal 75(1-2): 57-58
- Mew TW and Gonzales 2002. A Handbook of Rice Seed Borne Fungi. IRRI Science Publishers pp. 83

- Misra JK, Mersa SD and Mew TW 1994. Organisms causing grain discoloration and damage in : A manual of rice seed health testing. ed. by TW Mew and JK Misra IRRI. Los Banos, Laguna, Philippines, pp. 99-100
- Naeimi S, Okhovvat SM, Hedjaroude GA and Khosravi V 2003. Sheath rot of rice in Iran. Commun. Agric. Appl. Biol. Sci. 68: 681-684
- Nehal S and EI-Mougy 2009. Effect of some essential oils for limiting early blight (*Alternaria solani*) development in potato field. J. Plant Protect. Res. 49:57-61
- Ou SH 1985. Rice Diseases. CAB International Mycological Institute, Kew, Surrey, U.K.
- Phookan AK and Hazarika DK 1992. Distribution of sheath rot (ShR) in six agroclimatic zones of Assam, India. IRRN 17:16
- Rangaswami G 1972. Diseases of crop plants in India. Prentice Hall of India Pvt. Ltd., New Delhi pp. 520
- Reddy CS and Ghosh A 1985. Sheath rot incidence and yield losses in rice due to the joint infection of rice tungro virus and sheath rot fungus. Indian Phytopath. 35: 165-165
- Reddy MM, Reddy CS and Reddy AGR 2001. Influence of weather parameters and insect pest populations on incidence and development of sheath rot of rice. Indian Phytopathol 54: 179-184
- Sakthivel N and Gnanamanickam SS 1987. Evaluation of *Pseudomonas fluorescens* for suppression of sheath rot disease and for enhancement of grain yields in rice (*Oryza sativa* L). Appl. Environ. Microbiol. 53: 2056-2059
- Sakthivel N 2001. Sheath rot disease of rice: current status and control strategies in Major Fungal Diseases of Rice: Recent Advances, eds S. Sreenivasa Prasad and R Johnson (Dordrecht:Springer), 271-283
- Vincent JM 1947. Distortion of fungal hyphae in presence of certain inhibitors 159 : 850