



## PREVALENCE OF RICE BROWN SPOT DISEASE INCIDENCE IN NORTHERN DISTRICTS OF TAMILNADU, INDIA AND OBSERVATIONS ON MORPHO PATHOGENIC VARIABILITY AMONG ISOLATES OF *BIPOLARIS ORYZAE*

S. Sudhasha\*, P. Balabaskar, K. Sanjeev Kumar and T. Sivakumar

Department of Plant Pathology, Faculty of Agriculture, Annamalai University, Annamalai Nagar-608002, Tamilnadu, India

E mail id: sudhashapat@gmail.com

### Abstract

Rice productivity is severely constrained by many fungal pathogens amongst, Rice brown spot pathogen (*Bipolaris oryzae*) is causing severe economic losses by infecting the crop from seed to grain. This investigation was carried out to assess the prevalence of rice brown spot disease incidence in seven districts of Northern Tamilnadu and to report the disease severity in terms of PDI (Percent disease index) along with AUDPC (Area under disease progress curve) values. Amongst the seven districts on survey, the lowest PDI was recorded from Kancheepuram district (PDI - 15.22) with an AUDPC value of 647.98. The maximum PDI (34.31%) was recorded in Cuddalore district with 1444.18 as AUDPC value followed by Vellore district and Thiruvannamalai district which were on par. Based on these AUDPC values, the seven districts were divided in to four divisions and Cuddalore district came under more severe disease progress status as it recorded an AUDPC value of above 1200. Further, the fifteen isolates collected from Cuddalore district tested for their pathogenicity under pot culture on Kharif and Samba seasons showed significant variation in respect to morphology and pathogenicity. The brown spot incidence under potculture was observed more on samba season (Mean PDI- 46.071 ) than kharif season (Mean PDI- 26.20). The Mean AUDPC value directly correlated with PDI value was higher in Samba season (Mean AUDPC- 1046.90 ) than Kharif season (Mean AUDPC- 918.65) The isolates produced thin to fluffy and Pale grey to brownish black mycelium and took 3-5days to cover the Petri plate. Significant variations were observed with regard to sporulation ( $3.9-9.8 \times 10^4$  spores/ml) and size of conidia among the tested isolates.

**Keywords:** Survey, Rice brown spot, *Bipolaris oryzae*, PDI (Percent disease index), AUDPC (Area under disease progress curve).

### Introduction

Rice (*Oryza sativa* L.) is the staple food for half of the world population. It is susceptible to many of the fungal pathogens of which, the rice brown spot disease caused by *Bipolaris oryzae* (Breda de Haan) Shoemaker, resulted in two great famines, viz., in Krishna-Godavari delta during 1918-1919 and Bengal famine during 1942 which led to death of 2 million people (Padmanaban, 1973; Chakrabarthi, 2001). Since then, brown spot is widely reported in India (Reddy *et al.*, 2011) as it affects the crop from seed to grain. Besides, the pathogen affects millions of hectares worldwide every year and yield losses in relative terms vary widely from 4 to 52% (Barnwal *et al.*, 2013). Rice brown spot disease caused symptoms like leaf spot, decrease in number of tillers, reduced root and shoot elongation, chaffiness in grains, stalk rot symptom and grain discolouration (Vidyasekharan and Ramadoss, 1973; Ramakrishnan and Subramanian, 1977; Ou, 1985; Sunder *et al.* 2005). A light reddish- brown or lesion with a grey centre surrounded by a dark to reddish-brown margin with a bright yellow halo is a distinctive nature of the symptom produced by the pathogen.

Considering the importance of this disease, this investigation was carried out in seven districts of northern Tamilnadu, India to assess the extent of occurrence of this disease and express the damage in terms of per cent disease index (PDI) and AUDPC. Also it was proposed to isolate the strains of the pathogen from the district where the maximum incidence was observed to assess the cultural, morphological and pathogenic variability among the isolates of *B. oryzae*.

### Materials and Methods

#### Assessment of disease prevalence

Observations on incidence of rice brown spot disease were recorded from seven districts (northern districts) viz., Thiruvallur (13°09'N, 79°57'E), Kancheepuram (12°50'N,

79°45'E), Chengalpattu (12° 41' N, 79° 58'E), Villupuram (11°57'N, 79°32'E), Cuddalore (11°43'N, 79°49'E), Vellore (12°93'N, 79°19'E) and Thiruvannamalai (12°30'N, 79°04'E) of Tamilnadu following a standard roving survey during December 2018 to February 2019, to discern the status and distribution of rice brown spot disease incidence. Five locations were selected in each district and in each random plot selected per village, 10 plants were tagged for disease scoring. Disease scoring for foliar infection was visually evaluated at fifteen days interval. The PDI (Percent disease index) was computed using the standard disease rating scale of IRRI (2002).

#### Disease rating scale (IRRI, 2002)

SCALE (Affected leaf area)	
1	No incidence
2	Less than 1%
3	1-3%
4	4-5%
5	11-15%
6	16-25%
7	26-50%
8	51-75%
9	76-100%

#### Computation of Percent disease index (PDI) on survey

The percent disease index (PDI) was calculated by adopting the formula by Mckenny (1927).

$$\text{PDI} = \frac{\text{Sum of numerical rating}}{\text{Total numbers of plants observed}} \times 100$$

maximum disease scale

The PDI was computed at the time of harvesting on survey.

### Computation of Area under disease progress curve (AUDPC) on survey

To determine the measure of disease development throughout the period, Area under disease progress curve (AUDPC) (Reynolds and Neher, 1997) was calculated using the formula given by Campbell and Madden (1990).

$$\text{AUDPC} = \sum_{i=1}^{n-1} (Y_{i+1} + Y_i) 0.5 (T_{i+1} - T_i)$$

Where,

$Y_i$  = brown leaf spot disease severity on the  $i$ th date

$T_i$  = date on which the disease was scored

$n$  = numbers of dates on which disease was scored

AUDPC values were recorded at the interval period of 95 – 110DAT, 110 – 125 DAT and 125 – 140 DAT. Based on these AUDPC values, the seven districts were divided in to four divisions.

AUDPC Values	Division	Disease Progress status	Districts
≤800	1	Low	Kancheepuram
800 – 1000	2	Moderate	Thiruvallur, Chengalpattu
1000– 1200	3	Severe	Vellore, Thiruvannamalai
≥1200	4	More Severe	Cuddalore

On survey, since the Cuddalore district was coming under more severe category (Division 4) this district was investigated further in detail. In Cuddalore district, the pathogenic isolates were collected from 15 places and the pathogenic variability was assessed during Kharif (May) and Samba (September) seasons under pot culture condition and they were assessed for the cultural and morphological variability also.

### Isolation of pathogenic isolates (*Bipolaris oryzae*)

The leaves showing typical symptoms collected from fifteen places of Cuddalore district, were washed with sterile water and the necrotic patches of diseased leaves were cut into small pieces. The cut pieces were surface sterilized by dipping in 1% Sodium hypo chloride solution for one minute. Then they were washed by sterilized water repeatedly to remove the remnants if any and inoculated in sterilized Petri dish amended with potato dextrose agar medium (PDA) and incubated at room temperature ( $28 \pm 2^\circ$ ). After 5 to 7 days, the fungal growth was examined and aseptically transferred to PDA slants. The pure culture of the pathogen isolates were obtained using single hyphal tip technique. Mycelia and asexual spores (conidia) were examined under the microscope for identification of the pathogen (Subramanian, 1972). Confirmation of the pathogenicity was done by following Koch's Postulates.

### Pathogenicity test

These fifteen isolates collected were tested for their pathogenic variability under pot culture condition during Kharif (April-May) and Samba (August-September) seasons. The rice variety ASD 16 was used during Kharif season and the rice variety CR1009 was used during Samba season as these varieties were susceptible to *B. oryzae*.

### Artificial Inoculation of pathogen

The pathogen was mass multiplied on sterilized paddy chaffy grains and allowed to grow for 10 days. Conidia of this pathogen were harvested by mixing the colonized paddy chaffy grains with sterile water. Spore suspension ( $5 \times 10^5$  spore  $\text{ml}^{-1}$ ) of the fifteen isolates of *B. oryzae* separately were then sprayed on the seedlings of Kharif season crop on 35 DAT and on 70DAT for Samba season crop using hand atomizer. The symptoms appeared 7-10 days after artificial inoculation under pot culture. Three replications were maintained for each of the test isolate.

### Computation of PDI and AUDPC in Kharif and Samba seasons

The PDI was computed at the time of harvesting during Kharif season (ASD16) and Samba season (CR1009).

AUDPC values were recorded at the 15 days interval period of 45 – 60DAT, 60 – 75DAT and 75 – 90 DAT for Kharif crop and the same was recorded at the interval period of 95 – 110 DAT, 110 – 125 DAT and 125 – 140 DAT for Samba season crop following the above said formulae.

### Morphological observation of pathogenic isolates

The pathogenic isolates were multiplied on Potato Dextrose Agar medium and the colony characteristics (growth pattern, colony colour) were observed (Sobanbabu *et al.*, 2018). For assessing the sporulation, the isolates of *B. oryzae* were multiplied in Rice Extract Agar and incubated at  $26 \pm 1^\circ\text{C}$  for 15 days. After incubation, spores were scraped off into 10 ml of sterile distilled water per plate, and the number of spores was determined with a hemocytometer (Hau and Rush, 1980). The sporulation, conidiophore size, conidia size and conidial septation were also observed for all the fifteen isolates and recorded.

### Statistical analysis

Data entry and processing were carried out in MS Excel-10. Randomized Block Design was used for analysis. Analysis was done at 5% level of significance (Gomez and Gomez, 1984).

### Results and Discussion

Amongst the isolates collected from seven districts, the lowest PDI was recorded from Kancheepuram district (PDI - 15.22) along with AUDPC value of 647.98. The maximum PDI (34.31%) was recorded in Cuddalore district along with 1444.18 as AUDPC value followed by Vellore district (PDI value -28.25, AUDPC value - 1150.90) and Thiruvannamalai district (PDI value -27.65, AUDPC value - 1158.48) which were on par (Table 1).

Since, the rice brown spot disease is air borne and seed borne, this variation in the extent of damage may be due to epidemiological conditions such as air current, moisture level in air, temperature condition prevailing there, amount of agricultural inputs used and the variety chosen for cultivation.

Jones *et al.* (1993) have also reported that incidence and severity of brown spot under low land irrigated conditions in Cameroon region was higher as compared to the uplands. In a similar line Baranwal *et al.* (2013) also opined that rice fields under water scarcity and imbalance supplement of nitrogenous fertilizers exaggerated the efficacy of the pathogen on rice yield and grain quality. Further, it was also

reported that the tolerant varieties showed less PDI and disease progress values than susceptible varieties (PremBahadur Magar, 2015; Aryal, 2016). All these earlier reports lend support to the present findings.

Based on the results, the isolates collected from Cuddalore district were assessed for their pathogenic, culture and morphological variability. The symptoms appeared 7-10 days after artificial inoculation under pot culture and disease progression was higher in Samba season than Kharif season.

During Kharif season, the isolates RBS1 (AUDPC-1185.24), BS7 (AUDPC-1033.73), RBS14 (AUDPC-1209.25) and RBS15 (AUDPC-1165.50) showed more disease progress values  $\geq 1000$  with along with PDI values of 37.84, 31.87, 40.58 and 38.15 correspondingly regarded as severe virulence. The isolates RBS2, RBS6, RBS8, RBS9, RBS11, RBS12 and RBS13 showed more than 800 - 1000 AUDPC values with 20- 30% PDI values considered as moderate virulence. The remaining isolates RBS 3, RBS 4, RBS 5 and RBS 10 have recorded PDI values of below 20% and the AUDPC values of below 800 (low virulence) (Table 2)

During Samba season, the isolates RBS1, BS7, BS13, BS14 and RBS15 showed more severe disease progress values ranging from 1185.0 – 1712.85 with PDI values of 49.00 – 58.24 %. The isolates RBS2, RBS6, RBS8, RBS9 and RBS10 showed more AUDPC values ranged from 810.91 – 990.45 along with the PDI values of 40.94 – 44.65%. The isolates RBS3, RBS4, RBS5, RBS9 and RBS12 recorded PDI values in the range of 37.6 – 44.56% (Table 3).

The mean PDI for all the 15 isolates collected from Cuddalore district showed higher values in Samba season (46.07) than Kharif season. The disease progress (AUDPC) was also comparatively faster in Samba season (Mean AUDPC-1046.90) than Kharif season (Mean AUDPC-918.65). This variation may be due to the difference in the virulence of the pathogen and better weather parameters (Annexure-I) in Samba season rather than Kharif season. These results were in accordance with the results of André (2015) who found that the rice brown spot disease causing pathogen was dependent on seasonal weather conditions. Vishal Gupta *et al.* (2013) also reported that, the variation in PDI and AUDPC at different locations may be due to the variations of environmental factors prevailing in these areas coupled with cultivation of susceptible varieties.

Besides, it was observed that the PDI and AUDPC values were directly proportionate to each other. AUDPC values increased with time of observation in both the seasons. This is in accordance with Aryal *et al.* (2016) also proved that the PDI and AUDPC values were increased with time of observation.

The mean PDI and mean AUDPC values in Cuddalore district recorded on survey on samba season were 34.31 and 1444.18. The same isolate when inoculated under pot culture in samba season recorded the mean PDI as 46.07 and mean AUDPC as 1046.90. The computed mean value of PDI on survey was lesser than that of pot culture condition or an artificial condition. This may be due to the surveyed main field areas were exposed to various biotic and abiotic factors

than greenhouse condition. The virulence nature of the pathogen was very severe in artificially inoculated condition than open field condition where it might be restricted by various external factors. Hence the PDI report of the same isolates threaten the crop more in pot culture than mainfield. In contrast, the mean AUDPC values were more in main field than green house condition comparatively. This may be due to the secondary spread through air which, may be limiting factor under green house condition in comparison with field condition and this may be the cause for the variability in AUDPC values for the isolates under field and pot culture condition. This result was in accordance with Anna Maria picco, 2002 who estimated the bipolaris oryzae count was started at the end of June and peaked at July in atmospheric air.

### Morphological Variability

The colonies grown on Petri plates varied from dull white, grey to dark brownish colonies, fluffy mycelium or thin mycelium with profuse growth covering the entire plate from 3- 5 days. The results revealed that the isolates RBS 3, RBS 10, RBS12, RBS13, RBS 14, and RBS15 were covering the 90mm Petri plate in 3 days with profuse and dark coloured mycelium which also showed more sporulation than other isolates.

The observations implied that conidiophores of the fungi were single, unbranched, brown, thick walled and geniculated. Conidia were brown, multiseptate, elongated oval or spindle, straight, sometimes bent to one side, broad at middle, narrowed at tips. These also showed variation in size where, the isolates collected from Cuddalore district showed the range of size of conidiophores (69.8-170.0  $\mu\text{m}$  x 44.2 – 104.2  $\mu\text{m}$ ) and conidia (44.2 – 104.2  $\mu\text{m}$  X 6.4-16.4  $\mu\text{m}$ ) (Table 4). These results were in accordance with the observations of Ou (1985) who reported that the size of conidiophores and conidia of *Bipolaris oryzae* isolates has been observed to vary from 70-175 x 5.6-7  $\mu\text{m}$  and 45-106 x 14-17  $\mu\text{m}$  in India. Besides, considerable variation among the pathogen isolates with respect to size, colour, number of cells per conidium and nature of infection have also been reported earlier (Misra, 1985; Harish *et al.*, 2007).

### Conclusion

The findings of this study revealed that the seed cum air borne rice brown spot disease is endemic in the areas surveyed with the maximum incidence of the disease recorded in Cuddalore district of Tamilnadu. The isolates collected from Cuddalore district showed significant variation in respect of pathogenicity and morphological characters. Also the study clearly revealed the preference of samba season climate for its perpetuation. Such understanding about the morphological variations and virulence coupled with the climatic preferences among the isolates of rice brown spot pathogen is the primary step in devising disease management practices. Also, it is shown that PDI and AUDPC values are very important parameters to be computed in rice cultivation as these two values represent the entire data about one location which can lead to effective forecasting to farmers.

**Table 1 :** Prevalence of rice brown spot disease incidence in northern districts of Tamilnadu during 2019

S. No.	District	Location	Variety	PDI (Percent Disease Index) (At the time of harvest)	AUDPC values for the given interval period			
					95-110 DAT	110-125 DAT	125-140 DAT	Total AUDPC
1	Thiruvallur	Pakkam	TKM 9	25.13	264.15	339.23	405.98	1009.35
2		Veppampattu	CO 49	27.76	306.08	381.30	441.38	1128.76
3		Periyapalayam	ADT 44	23.76	242.55	339.00	384.98	946.58
4		Pattabiram	ADT49	25.00	264.75	339.00	404.03	1007.78
5		Paruthipattu	CO49	20.98	218.10	278.10	337.43	833.63
<b>Mean±S.E</b>				<b>24.52±1.1</b>	<b>259.13±14.52</b>	<b>335.35±16.48</b>	<b>394.76±16.98</b>	<b>985.22±48.05</b>
<b>Range</b>				<b>20.98-27.76</b>	<b>218.10-306.08</b>	<b>278.10-381.30</b>	<b>337.43-441.38</b>	<b>833.63-1128.76</b>
<b>CV</b>				<b>2.46</b>	<b>12.53</b>	<b>10.99</b>	<b>9.62</b>	<b>10.90</b>
6	Kancheepuram	Parandur	ADT44	15.09	158.93	203.93	248.93	611.74
7		Govindavadi	ADT49	18.90	188.40	254.10	309.75	752.25
8		Naduveerapattu	CR1009	12.90	125.25	170.25	420.30	715.80
9		Padappai	ADT44	13.09	129.30	178.73	226.43	534.46
10		Arasankalani	ADT49	16.12	149.25	207.15	269.25	625.65
<b>Mean±S.E</b>				<b>15.22±1.10</b>	<b>150.23±11.39</b>	<b>202.83±14.65</b>	<b>294.93±34.21</b>	<b>647.98±38.84</b>
<b>Range</b>				<b>12.90-18.90</b>	<b>129.30-188.40</b>	<b>170.25-254.10</b>	<b>226.43-420.30</b>	<b>534.46-752.25</b>
<b>CV</b>				<b>16.19</b>	<b>16.96</b>	<b>16.15</b>	<b>25.94</b>	<b>13.40</b>
11	Chengalpattu	Karumbakkam	ADT49	25.73	269.48	208.95	423.45	901.88
12		Edayankuppam	CR1009	27.50	304.65	379.65	441.75	1126.05
13		Salavakkam	ADT49	23.66	239.70	313.95	380.70	934.35
14		Osivakkam	CR1009	21.80	205.05	279.30	357.00	841.35
15		Sennari	ADT44	25.86	263.18	344.03	419.40	1026.69
<b>Mean±S.E</b>				<b>24.91±0.99</b>	<b>256.12±16.52</b>	<b>305.18±29.21</b>	<b>404.46±15.48</b>	<b>966.06±49.98</b>
<b>Range</b>				<b>21.80-27.50</b>	<b>205.05-304.65</b>	<b>208.95-379.65</b>	<b>357.00-441.75</b>	<b>841.35-1126.05</b>
<b>CV</b>				<b>8.87</b>	<b>14.41</b>	<b>21.40</b>	<b>8.56</b>	<b>11.57</b>
16	Villupuram	Arasamangalam	CR1009	28.10	313.50	388.50	463.80	1165.80
17		Chittalampattu	ADT44	24.36	257.25	329.48	403.80	990.53
18		Esalam	BPT5204	29.13	329.85	405.83	474.38	1210.05
19		Kottamangalam	CR1009	24.76	226.40	341.40	405.45	1013.25
20		Melpadi	BPT5204	29.00	330.00	405.00	465.00	1200.00
<b>Mean±S.E</b>				<b>27.07±1.04</b>	<b>291.40±21.03</b>	<b>374.04±16.17</b>	<b>442.49±15.57</b>	<b>1115.93±47.27</b>
<b>Range</b>				<b>24.36-29.13</b>	<b>226.40-330.00</b>	<b>329.48-405.83</b>	<b>403.80-474.38</b>	<b>990.53-1210.05</b>
<b>CV</b>				<b>8.60</b>	<b>16.14</b>	<b>9.66</b>	<b>7.87</b>	<b>9.47</b>
21	Cuddalore	Seeyapadi	CR1009	40.27	516.60	573.38	634.13	1724.11
22		Manjakuzhi	BPT5204	36.15	429.00	505.13	571.73	1505.86
23		Sathiyamangalam	CR1009	38.25	479.93	541.95	589.13	1611.01
24		Sivapuri	BPT5204	29.30	342.00	417.00	475.50	1234.50
25		Annamalai Nagar	BPT5204	27.56	321.00	391.95	432.45	1145.40
<b>Mean±S.E</b>				<b>34.31±2.50</b>	<b>417.71±37.99</b>	<b>485.88±35.17</b>	<b>540.59±37.42</b>	<b>1444.18±110.28</b>
<b>Range</b>				<b>27.56-29.30</b>	<b>321.00-516.60</b>	<b>391.95-573.38</b>	<b>432.45-634.13</b>	<b>1145.40-1611.01</b>
<b>CV</b>				<b>16.30</b>	<b>20.34</b>	<b>16.19</b>	<b>15.48</b>	<b>17.08</b>
26	Vellore	Devakapuram	ADT44	33.65	385.28	471.38	523.13	1379.79
27		Anaipakkam	ADT44	28.87	323.10	406.28	449.78	1179.16
28		Athithangal	CR1009	30.21	348.90	424.58	479.63	1253.11
29		Chinnakallupalli	ADT44	26.56	306.00	364.95	425.48	1096.43
30		Erukkampattu	CR1009	21.98	208.20	283.20	354.60	846.00
<b>Mean±S.E</b>				<b>28.25±1.94</b>	<b>314.30±29.70</b>	<b>390.08±31.72</b>	<b>446.52±28.17</b>	<b>1150.90±89.32</b>
<b>Range</b>				<b>21.98-33.65</b>	<b>208.20-385.28</b>	<b>283.20-471.38</b>	<b>354.60-523.13</b>	<b>846.00-1379.79</b>
<b>CV</b>				<b>15.38</b>	<b>21.13</b>	<b>18.18</b>	<b>14.11</b>	<b>17.35</b>
31	Thiruvannamalai	Athipadi	ADT49	25.87	272.03	355.05	415.28	1042.36
32		Chellankuppam	ADT44	29.08	331.95	399.45	474.45	1205.85
33		Devanur	CR1009	30.08	343.35	413.10	481.50	1237.95
34		Kadambai	ADT44	25.08	273.68	344.85	406.50	1157.85
35		Keeranur	CR1009	28.16	309.75	384.45	454.20	1148.40
<b>Mean±S.E</b>				<b>27.65±0.95</b>	<b>306.15±14.63</b>	<b>379.38±12.94</b>	<b>446.9±15.23</b>	<b>1158.48±33.28</b>
<b>Range</b>				<b>25.08-30.08</b>	<b>272.03-343.35</b>	<b>344.85-413.10</b>	<b>406.50-481.50</b>	<b>1042.36-1237.95</b>
<b>CV</b>				<b>7.67</b>	<b>10.69</b>	<b>7.63</b>	<b>7.63</b>	<b>6.42</b>

PDI :Percent Disease Index; AUDPC: Area Under Disease Progress Curve; CV:Coefficient of variance; SEm (±) : standard error of mean

**Table 2 :** Estimation of disease severity of Rice brown spot incidence by different isolates collected from Cuddalore district under pot culture (Kharif)

S. No.	Locations	Isolate Number	PDI	AUDPC values for the given interval period			Total AUDPC
			Kharif (April-May)	45-60 DAT	60-75 DAT	75-90 DAT	
1	Manalur	RBS 1	37.84 <sup>bcd</sup>	356.58 <sup>abc</sup>	392.28 <sup>abcd</sup>	436.38 <sup>abc</sup>	1185.24 <sup>abcd</sup>
2	Kakkangudi	RBS 2	23.68 <sup>g</sup>	254.80 <sup>f</sup>	286.30 <sup>f</sup>	317.63 <sup>e</sup>	858.73 <sup>f</sup>
3	Ayepettai	RBS 3	18.76 <sup>defg</sup>	219.98 <sup>def</sup>	252.28 <sup>def</sup>	272.93 <sup>cde</sup>	745.19 <sup>def</sup>
4	Annamalai Nagar	RBS 4	17.56 <sup>cdef</sup>	214.55 <sup>bcd</sup>	247.80 <sup>abcde</sup>	273.35 <sup>abcd</sup>	735.70 <sup>abcde</sup>
5	Sivapuri	RBS 5	18.78 <sup>efg</sup>	225.23 <sup>def</sup>	257.43 <sup>def</sup>	273.35 <sup>cde</sup>	756.01 <sup>def</sup>
6	Avalur	RBS 6	24.56 <sup>defg</sup>	261.63 <sup>cdef</sup>	300.86 <sup>def</sup>	329.00 <sup>cde</sup>	891.49 <sup>def</sup>
7	Sathiyamangalam	RBS 7	31.87 <sup>abc</sup>	314.58 <sup>ab</sup>	345.56 <sup>abc</sup>	373.59 <sup>ab</sup>	1033.73 <sup>abc</sup>
8	Vanrampattu	RBS 8	24.45 <sup>ab</sup>	273.00 <sup>a</sup>	301.00 <sup>ab</sup>	339.50 <sup>a</sup>	913.50 <sup>ab</sup>
9	Sethiyathopu	RBS 9	19.65 <sup>defg</sup>	245.21 <sup>def</sup>	270.55 <sup>def</sup>	298.55 <sup>de</sup>	814.31 <sup>ef</sup>
10	Adivaraganatham	RBS 10	20.98 <sup>fg</sup>	229.39 <sup>ef</sup>	264.60 <sup>ef</sup>	295.75 <sup>de</sup>	789.74 <sup>ef</sup>
11	Rettaikulam	RBS 11	22.46 <sup>bcd</sup>	247.42 <sup>bcd</sup>	273.98 <sup>bcde</sup>	305.73 <sup>abcde</sup>	827.13 <sup>bcde</sup>
12	Manjakuzhi	RBS 12	24.65 <sup>defg</sup>	259.00 <sup>cdef</sup>	291.55 <sup>def</sup>	318.64 <sup>bcde</sup>	869.19 <sup>def</sup>
13	Pravallur	RBS 13	29.00 <sup>defg</sup>	301.49 <sup>def</sup>	329.49 <sup>def</sup>	353.99 <sup>cde</sup>	984.97 <sup>def</sup>
14	Seeyapadi	RBS 14	40.58 <sup>cdef</sup>	368.55 <sup>bcde</sup>	404.25 <sup>cdef</sup>	436.45 <sup>bcde</sup>	1209.25 <sup>cdef</sup>
15	Odaiyur	RBS 15	38.15 <sup>a</sup>	353.50 <sup>a</sup>	390.25 <sup>a</sup>	421.75 <sup>a</sup>	1165.50 <sup>a</sup>
Mean ± S.E			26.20± 1.96	147.56± 11.80	307.21±13.73	336.44±14.75	918.65±41.80
CV			17.01	10.92	11.04	11.08	11.29
CD (0.05)			9.53	62.24	70.20	77.22	214.75

PDI : Percent Disease Index; AUDPC: Area Under Disease Progress Curve CV: Coefficient of variance and CD(0.05): Critical Difference at 5 % level of significance, SEM (±) : standard error of mean

**Table 3 :** Estimation of disease severity of Rice brown spot incidence by different isolates collected from Cuddalore district under pot culture (Samba season)

S. No.	Locations	Isolate Number	PDI	AUDPC values			Total AUDPC
			Samba (August-Sept)	95-110 DAT	110-125 DAT	125-140 DAT	
1	Manalur	RBS 1	57.60 <sup>bcd</sup>	444.15 <sup>bcd</sup>	525.15 <sup>bcd</sup>	595.35 <sup>abc</sup>	1564.65 <sup>bcd</sup>
2	Kakkangudi	RBS 2	43.86 <sup>g</sup>	240.15 <sup>f</sup>	316.20 <sup>g</sup>	394.35 <sup>cd</sup>	950.70 <sup>f</sup>
3	Ayepettai	RBS 3	38.90 <sup>efg</sup>	174.30 <sup>def</sup>	246.75 <sup>defg</sup>	306.45 <sup>bcd</sup>	727.50 <sup>def</sup>
4	Annamalai Nagar	RBS 4	37.60 <sup>cdef</sup>	173.25 <sup>cdef</sup>	234.75 <sup>cdef</sup>	306.45 <sup>abc</sup>	714.45 <sup>cdef</sup>
5	Sivapuri	RBS 5	38.90 <sup>efg</sup>	185.70 <sup>ef</sup>	247.20 <sup>efg</sup>	308.85 <sup>cd</sup>	741.75 <sup>def</sup>
6	Avalur	RBS 6	44.65 <sup>defg</sup>	256.95 <sup>def</sup>	331.95 <sup>defg</sup>	401.55 <sup>d</sup>	990.45 <sup>ef</sup>
7	Sathiyamangalam	RBS 7	51.65 <sup>abc</sup>	365.85 <sup>abc</sup>	439.88 <sup>abc</sup>	504.9 <sup>ab</sup>	1310.63 <sup>abc</sup>
8	Vanrampattu	RBS 8	44.35 <sup>ab</sup>	254.93 <sup>ab</sup>	329.93 <sup>ab</sup>	404.93 <sup>a</sup>	989.79 <sup>ab</sup>
9	Sethiyathopu	RBS 9	39.85 <sup>defg</sup>	192.53 <sup>def</sup>	264.53 <sup>defg</sup>	332.85 <sup>bcd</sup>	789.91 <sup>def</sup>
10	Adivaraganatham	RBS 10	40.94 <sup>fg</sup>	196.88 <sup>f</sup>	271.13 <sup>fg</sup>	342.90 <sup>cd</sup>	810.91 <sup>f</sup>
11	Rettaikulam	RBS 11	42.56 <sup>bcd</sup>	230.10 <sup>bcde</sup>	302.70 <sup>bcde</sup>	355.65 <sup>abc</sup>	888.45 <sup>bcde</sup>
12	Manjakuzhi	RBS 12	44.56 <sup>defg</sup>	256.50 <sup>def</sup>	333.75 <sup>defg</sup>	133.58 <sup>bcd</sup>	723.83 <sup>def</sup>
13	Pravallur	RBS 13	49.00 <sup>defg</sup>	322.50 <sup>def</sup>	397.50 <sup>defg</sup>	465.00 <sup>bcd</sup>	1185.00 <sup>def</sup>
14	Seeyapadi	RBS 14	60.40 <sup>bcde</sup>	497.70 <sup>cdef</sup>	572.70 <sup>cdef</sup>	642.45 <sup>cd</sup>	1712.85 <sup>cdef</sup>
15	Odaiyur	RBS 15	58.24 <sup>a</sup>	464.26 <sup>a</sup>	539.25 <sup>a</sup>	599.18 <sup>a</sup>	1602.68 <sup>a</sup>
Mean ± S.E			46.071± 1.932	283.72± 28.37	356.89± 29.12	406.30±35.04	1046.90±89.49
CV			9.66	22.49	18.32	24.11	20.09
CD (0.05)			9.55	136.46	139.83	209.52	449.80

PDI :Percent Disease Index; AUDPC: Area Under Disease Progress Curve CV: Coefficient of variance and CD(0.05):Critical Difference at 5 % level of significance, SEM (±) : standard error of mean

### Annexure -1

Samba season which starts with August – September, the last month of the rainy (monsoon) season, in Chidambaram, with average temperature fluctuating between 31.8°C – 24.3°C (89.2°F-75.7°F), 113mm-230mm of precipitation and the average length of the day was 12.2h-11.9h (Online resource- Weather Atlas). Whereas, the Kharif season starts with April-May is a hot summer month in Chidambaram, with average temperature fluctuating between 33.9°C-36.4°C (93°F-97.5°F), 23-47mm of precipitation the average length of the day in April-May is 12.2- 12.6h (Online resource- Weather Atlas).

**Table 4 :** Morphological Variability of *B. oryzae* isolates collected from different places of Cuddalore district

S. No	Isolate	Isolate Number	Colony morphology	Mycelial coverage on entire plate(days)	Spores per ml x 10 <sup>4</sup>	Conidiophore size (µm)	Conidia size (µm)
1	Manalur	RBS 1	Fluffy, grey	4	6.0	98 × 6.4	54 × 16.2
2	Kakkangudi	RBS 2	Fluffy, grey	4	6.5	88.4 × 5.8	49 × 15.1
3	Ayepettai	RBS 3	Fluffy, olive brown	3	7.4	69.8 × 5.4	44.2 × 14.8
4	Annamalai Nagar	RBS 4	Fluffy, dull white	5	3.9	94.3 × 4.9	51.8 × 14.5
5	Sivapuri	RBS 5	Fluffy, pale grey	4	5.1	106.1 × 6.4	68 × 16.4
6	Avalur	RBS 6	Thin mycelium, pale grey	5	4.7	100.5 × 2.4	64.6 × 16.4
7	Sathiyamangalam	RBS 7	Thin, dull grey	4	5.6	78.8 × 4.4	49.2 × 6.4
8	Vanrampattu	RBS 8	Fluffy, dull grey	5	4.5	96.6 × 3.4	56.6 × 6.4
9	Sethiyathopu	RBS 9	Fluffy, dull brownish grey	5	5.0	112 × 5.4	89.4 × 11.2
10	Adivaraganatham	RBS 10	Fluffy, olive brown	3	7.1	118 × 4.3	94.2 × 10.4
11	Rettaikulam	RBS 11	Fluffy, olive brown	4	6.4	108.8 × 3.4	92.4 × 13.2
12	Manjakuzhi	RBS 12	Fluffy, olive grey	3	7.2	126.6 × 4.9	97.6 × 14.6
13	Pravallur	RBS 13	Fluffy, darkish grey	3	9.8	143.3 × 5.8	99.1 × 14.4
14	Seeyapadi	RBS 14	Fluffy, brownish black	3	8.8	155.5 × 5.9	104.2 × 16.4
15	Odaiyur	RBS 15	Fluffy, darkish grey	3	9.4	170 × 6.2	102.6 × 15.4
Range			Thin – Fluffy, Pale grey- brownish black	3-5	3.9-9.8	69.8-170.0 × 2.4 -6.4	44.2 – 104.2 × 6.4-16.4

### References

- Aluko, M.O. (1970). The measurement of brown spot of rice. PANS, 16: 76- 81.
- Anna, M.P. and Marinella, R. (2002). *Pyricularia grisea* and *Bipolaris oryzae* : A preliminary study on the occurrence of airborne spores in rice field. Aerobiologica. 18(2): 163-167.
- André, A.; Schwanck & Priscila, R.; Meneses & Cândida R.J.; Farias & Gustavo R.D.; Funck & Aline H.N.; Maia & Emerson, M. and Del Ponte (2015). *Bipolaris oryzae* seed borne inoculum and brown spot epidemics in the subtropical lowland rice-growing region of Brazil. Eur J Plant Pathol. 142: 875–885.
- Aryal, L.; Bhattarai, G.; Subedi, A.; Subedi, M.; Subedi, B. and Sah, G.K. (2016). Global Journal of Biology Agriculture and Health Sciences, 5(2): 50-54.
- Baranwal, M.K.; Kotasthane, A.; Magculia, N.; Mukherjee, P.K.; Savary, S.; Sharma, A.K.; Singh, H.B.; Singh, U.S.; Sparks, A.H.; Variar, M. and Zaidi, N. (2013). A review on crop losses, epidemiology and disease management of rice brown spot to identify research priorities and knowledge gaps. Eur. J. Plant Pathol. 136: 443-457.
- Dhaliwal, G.S.; Arora, R. and Dhawan, A.K. (2004). Crop losses due to insect pests in Indian Agriculture: an update. Indian J. Ecol. 31(1): 1-7.
- Jones, M.P.; Jeutong, F. and Tchatchoua, J. (1993). A survey of rice diseases in Cameroon. Plant Dis. 77:133-136.
- Gomez, K.A. and Gomez, A.A. (1984). Statistical procedures for agricultural research (2nd ed). A Wiley Inter science Publication, New York, USA. 655 p.
- Harish, S.; Saravanakumar, D.; Kamalakannan, A.; Vivekananthan, R.; Ebenezar, E.G. and Seetharaman, K. (2007). Phylloplane microorganisms as a potential biocontrol agent against *Helminthosporium oryzae* BredadeHaan, the incitant of rice brown spot. Arch. Phytopathol. Pl. Prot. 40: 148-15
- Hau, F.C. and Rush, M.C. (1980). A System for Inducing Sporulation of *Bipolaris oryzae* Plant Disease, 64(8): 788-789.
- Kohls, C.L.; Perich, J.A. and Hout, C.M. (1987). Wild rice yield losses associated with growth stage specific fungal brown spot epidemics. Plant Dis., 71: 419-422.
- Misra, A.K. (1985). Variability in *Drechslera oryzae*- the causal organism of brown spot disease of rice. Indian Phytopath. 38: 168-169.
- Ou, S.H. (1985). Rice diseases. Commonwealth Mycological Institute, Kew.
- Padmanabhan, S.Y. (1973). The Great Bengal famine. Ann Rev. Plant Pathol. 11:11-24
- Pandey, H.N.; Menon, T.C.M. and Rao, M.V. (1989). A single formula for calculating Area Under Brown Spot Progress Curve. Rachis, 8: 38-39.
- Prembahadur, M. (2015). Screening of rice varieties against brown leaf spot disease at Jyotinagar, chitwan, nepal. International journal of applied sciences and biotechnology, 3(1): 56-60.
- Ramakrishnan, G. and Subramanian, C.L. (1977). Studies on helminthosporiose of rice: Involvement of different species in disease development. Madras Agric. J. 64: 60-61.
- Reddy, C.S.; Laha, G.S.; Prasad, M.S.; Krishnaveni, D.; Castilla, N.P.; Nelson, A. and Savary, S. (2011). Characterizing multiple linkages between individual diseases, crop health syndromes, germplasm deployment and rice production situations in India. Field Crops Res., 120: 241–253.
- Sobanbabu, G.; Sabarinathan, K.G.; Parthiban, V.K. and Ramamoorthy, V. (2018). Isolation, Screening and Identification of virulent Isolates of *Bipolaris oryzae*

- causing Rice Brown Spot and *Sarocladiumoryzae* causing Sheath Rot Disease. *International Journal of Current Microbiology and Applied Sciences*, 7(09): 2319-7706.
- Sreenivasan, P.S. (1980). Meteorological aspects of rice production in India. In: Proceedings of a symposium on the Agrometeorology of the Rice crop. International Rice Research Institute, Manila, Philippines. 19-31.
- Vidhyasekharan, P. and Ramadoss, N. (1973). Quantitative and qualitative losses in paddy due to *Helminthosporium* epidemic. *Indian Phytopathology* 16: 479-84.
- Vishal, G.; Shamas, N.; Razdan, V.K.; Sharma, B.C.; Sharma, R.; Kaur, K.; Singh, I.; John, D. and Kumar, A. (2013). Foliar application of fungicides for the management of brown spot disease in rice (*Oryza sativa* L.) caused by *Bipolaris oryzae*. *African journal of agricultural research* 8(25): 3303-3309.