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## Conidial variability in rice blast caused by *Pyricularia grisea* (Cooke.) Sacc. in Konkan region of Maharashtra

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DOI: <https://doi.org/10.22271/chemi.2020.v8.i4ag.10072>**Abstract**

Conidiophores of all the isolates were simple, hyaline with single conidium borne at the tip except that of isolate BLS-9 which had three conidia born at tip and one at the middle of the conidiophore. The conidia were two celled in isolates BLS-7 and BLS-10 and all the conidia of remaining isolates were three celled and pyriform in shape with broad/narrow base and pointed/blunt apex. The average size (LXB) of conidia irrespective of isolates ranged between 22.20 – 8.35 X 7.28 – 2.65  $\mu\text{m}$ . Among all the isolates BLS-11 formed larger conidia with average dimensions of 22.20 X 7.00  $\mu\text{m}$  followed by BLS-9 (21.40 X 6.56  $\mu\text{m}$ ). Comparatively, small sized conidia were produced in isolate BLS-1 (08.35 X 2.65  $\mu\text{m}$ ).

**Keywords:** Rice blast, conidia, conidiophore, dimensions, variability**Introduction**

Rice (*Oryza sativa* L.) is the most important staple food grain crop of the world which constitutes the principle food for about 60 per cent of the world's population. It contributes 43 per cent of total food grain production and 46 per cent of total cereal production in India. In Maharashtra state, rice is cultivated on an area of 15.13 lakh hectares in four regions viz., Vidharbha (7.95 L ha.), Konkan (3.83 L ha.), Western Maharashtra (3.23 L ha.) and Marathwada (0.12 L ha.) with annual production of 41.71 L tonnes unmilled (brown rice) and 28.78 L tonnes milled rice. Highest productivity was recorded in Konkan region i.e. 2.75 tonnes ha milled rice and 3.83 tonnes ha unmilled (brown rice) with total production of 15.26 L tonnes unmilled (brown rice) and 10.53 L tonnes milled rice from 3.83 L ha area (Anonymous, 2015). Rice is a crop of Asian origin. It belongs to family Poaceae with two species, *Oryza sativa* and *O. glaberrima*. *O. sativa* is a native of tropical and subtropical southern Asia while *O. glaberrima* is a native of West Africa. Studies have suggested that there are three races of *Oryza sativa* cultivars viz. Indica, Javonica and Japonica. The blast pathogen *Pyricularia grisea* (Cooke) Sacc. is the anamorph of *Magnaporthe grisea* Barr which is an Ascomycetes. It has potential to attack the plant in all stages of growth. The infected host initially manifests the infection in form of typical elliptical to spindle shaped spots with dark reddish brown margin and ashy grey centre. Lesions may enlarge and coalesce, growing together, to kill the entire leaves. In advanced stages, infection to the neck region culminates into neck blast. The fungus forms pyriform (pear shaped), light brown, three celled, discrete macro-conidia on light brown conidiophores. The conidiophores and conidia are pigmented due to melanin (Kato, 2001) [1]. Amongst all the fungal diseases, rice blast caused by *Pyricularia grisea* (Cooke) Sacc. is one of the most devastating diseases of rice worldwide, causing yield losses to the extent of 70-80% in various rice ecosystems (Ratna Madhavi *et al.*, 2011) [2].

**Materials and Methods**

**Isolation:** The diseased leaf samples were collected from different rice growing pockets of Konkan region viz., Palghar, Raigad, Ratnagiri and Sindhudurg. Rice leaves showing typical symptoms of the blast were selected and washed first with tap water and then rinsed with distilled sterile water. Small pieces of diseased tissue along with some healthy tissue were cut into pieces of desired size with the help of a sterile scalpel and surface sterilized with 1% sodium hypochlorite solution for one minute, rinsed thrice in sterile water and dried with

sterilized blotter paper. The surface sterilized and dried leaf pieces were later transferred aseptically into the sterilized Petri dishes containing solidified Potato dextrose agar (PDA) medium and plates were incubated at 28°C until sporulation. Well grown pure culture was maintained on PDA slants by periodic sub culturing.

**Conidial measurements:** The conidial and conidiophore variability among the isolates was explored by studying the morphological characters such as colour, width, growth, size, septation of conidiophores and conidia. The slide culture technique was used to study the morphological characters of the test fungus. 5 mm discs of pure culture from Petri plates was transferred with the help of incinerated cork borer to the center of each sterilized micro slide and covered with sterilized coverslip.

Such micro slides were placed on a pair of glass rods in a sterilized Petri plate lined with moist blotting paper. Plates were kept for incubation at room temperature ( $27 \pm 1^\circ\text{C}$ ). After 48 hrs. cover slips from micro slides were removed gently. Fungal growth on the micro slide was stained immediately with lactophenol cotton blue, covered with cover slip and was observed under high power objective (100X) using Labovision binocular compound microscope with image analyzer. The average size (length and width) and shape of conidia was recorded.

## Results and Discussion

### Conidial Morphology

The morphological characters of hyphae, conidiophores and conidia of *P. grisea* were recorded. In all the isolates, the mycelium was septate, hyaline and branched. The conidia were single septate with two celled in BLS-7 and BLS-10. In all the remaining isolates, conidia were bi-septate, three celled and pyriform in shape. The base of the conidium was broad with pointed apex in isolates BLS-3, BLS-5, BLS-8, BLS-11, BLS-12 and narrow base with pointed apex in isolates BLS-2, BLS-4, BLS-6 and BLS-9. The isolates BLS-7 and BLS-10 had narrow base with blunt apex. In all the isolates the conidiophore was simple, hyaline with single conidium borne at the tip except isolate BLS-9 wherein three conidia were borne at the tip and one at the middle of the conidiophore (Table 1). As per the description mentioned by Shirai (1896)<sup>[3]</sup>, Hori (1898), Sawada (1917)<sup>[4]</sup> and Nishikado (1917)<sup>[5]</sup> the conidiophores are one to many, fasciculate, simple or rarely branched, 2 to 4 septate, not or slightly constricted at septa, olivaceous to fuliginous, base swollen, dark coloured and becoming lighter in colour towards the apex. Conidia are variable in size and shape, terminal, pyriform to obclavate, base rounded, apex narrowed and generally 2 septate, rarely 1-3 septate with or without constriction at septa.

Gashaw *et al* (2014)<sup>[6]</sup> reported that, in all the six isolates, the shape of the conidia was typically pyriform with base rounded, apex narrowed, 2-3 septate, 2-4 celled. The conidial morphology of the isolates in the present study also exhibited the variation in terms of number of cells. All the isolates formed pyriform conidia with narrow or broad base. These observations are in concurrence with earlier descriptions. The

findings of present study also confirm that spore morphology varies with geographical situation.

### Conidial Measurements

Data presented in Table 2 revealed that, size of the conidia varied among the isolates. Some of them were very long and narrow, while some were fairly broad. The average length of conidia of *P. grisea* ranged between 22.20-8.35  $\mu\text{m}$  and this difference was statistically significant. The highest average conidial length (22.20  $\mu\text{m}$ ) was exhibited by isolate BLS-11 and it was on par with BLS-9 (21.40  $\mu\text{m}$ ) and statistically significant over rest of all the isolates. Isolate BLS-9 (21.40  $\mu\text{m}$ ) was on par with isolate BLS-2, while shortest length of conidia (8.35  $\mu\text{m}$ ) was observed in BLS-1 and it was on par with BLS-8. Gashaw *et al* (2014)<sup>[6]</sup> reported that the two isolates out of six had the longest conidia admeasuring between 24.36 - 35.43  $\mu\text{m}$ . The lowest conidial length was 15.66 - 24.37  $\mu\text{m}$  and highest conidial width was observed for isolates Pg.40 and Pg.22 i.e. 8.35 - 11.92  $\mu\text{m}$  and 7.70-12.90  $\mu\text{m}$ , respectively.

Significant variation was also observed in the breadth of conidia. The average width of conidia ranged between 7.28 - 2.65  $\mu\text{m}$ . Maximum average width (7.28  $\mu\text{m}$ ) was recorded in isolate BLS-4 and it was on par with BLS-3, BLS-6 and BLS-11 and statistically significant over rest of all the isolates. The lowest average width (2.65  $\mu\text{m}$ ) was observed in BLS-1 and it was statistically on par with BLS-8, BLS-7 and BLS-10. Shirai (1896)<sup>[3]</sup>, Hori (1898), Sawada (1917)<sup>[4]</sup> and Nishikado (1917)<sup>[5]</sup> reported that conidia were variable in size from 14-40 X 6-13  $\mu\text{m}$  in size, usually 19-23 X 7-9  $\mu\text{m}$ , with small basal appendage. As described by Pordel *et al.* (2015)<sup>[7]</sup> the conidia of *P. oryzae* were 16-25 X 7-10  $\mu\text{m}$ . The results of present study are in agreement with their findings.

On the basis of this study it is concluded that, the mean average size of conidia of *P. grisea* irrespective of isolates was 14.26 X 4.84  $\mu\text{m}$  with a range of 22.20 – 8.35 X 7.28 – 2.65  $\mu\text{m}$ . Conidia of isolate BLS-11 were found to be larger with average dimensions of 22.20 X 7.00  $\mu\text{m}$  followed by BLS-9 (21.40 X 6.56  $\mu\text{m}$ ). Comparatively, small sized conidia were observed in BLS-1 (08.35 X 2.65  $\mu\text{m}$ ). Three distinct groups have been formed on the basis of conidial size as depicted in Table 3.

### Summary and Conclusion

In all the isolates the conidiophore was simple, hyaline with single conidium borne at the tip except isolate BLS-9 which had three conidia born at the tip and one at the middle of the conidiophore. The conidia were two celled in isolates BLS-7 and BLS-10, while in the remaining 10 isolates conidia were three celled. Though there was difference in number of cells in a conidium, the conidia of all the collected isolates were typically pyriform.

The mean average size (LXB) of conidia irrespective of isolate ranged between 22.20 – 8.35 X 7.28 – 2.65  $\mu\text{m}$ . Among all the isolates, BLS-11 was found to have larger average dimensions of 22.20 X 7.00  $\mu\text{m}$  followed by BLS-9 (21.40 X 6.56  $\mu\text{m}$ ). Comparatively, small sized conidia were produced by isolate BLS-1 (08.35 X 2.65  $\mu\text{m}$ ).

**Table 1:** Conidial and conidiophore morphology of different isolates of *P. grisea*

Isolates	Characteristic features	
	Conidiophore	Conidia
BLS-1	Simple, hyaline with single conidium born at the tip of the conidiophore	2 septate, 3 celled, pyriform with broad base and narrow apex
BLS-2	Simple, hyaline with single conidium born at the tip of the conidiophore	2 septate, 3 celled, pyriform with narrow base and pointed apex
BLS-3	Simple, hyaline with single conidium born at the tip of the conidiophore	2 septate, 3 celled, pyriform with broad base and pointed apex
BLS-4	Simple, hyaline with single conidium born at the tip of the conidiophore	2 septate, 3 celled, pyriform with narrow base and pointed apex
BLS-5	Simple, hyaline with single conidia born at the tip of the conidiophore.	2 septate, 3 celled, pyriform with broad base and pointed apex
BLS-6	Simple, hyaline with single conidium born at the tip of the conidiophore.	2 septate, 3 celled, pyriform with narrow base and pointed apex
BLS-7	Simple, hyaline with single conidium born at the tip of the conidiophore.	septate, 2 celled, pyriform with narrow base and blunt apex
BLS-8	Simple, hyaline with single conidium born at the tip of the conidiophore.	2 septate, 3 celled, pyriform with broad base and pointed apex
BLS-9	Simple, hyaline with 3 conidium born at the tip and 1 middle of the conidiophore.	2 septate, 3 celled, pyriform with narrow base and pointed apex
BLS-10	Simple, hyaline with single conidium born at the tip of the conidiophore.	septate, 2 celled, pyriform with narrow base and blunt apex
BLS-11	Simple, hyaline with single conidium born at the tip of the conidiophore.	2 septate, 3 celled, pyriform with broad base and pointed apex
BLS-12	Simple, hyaline with single conidium born at the tip of the conidiophore.	2 septate, 3 celled, pyriform with broad base and pointed apex

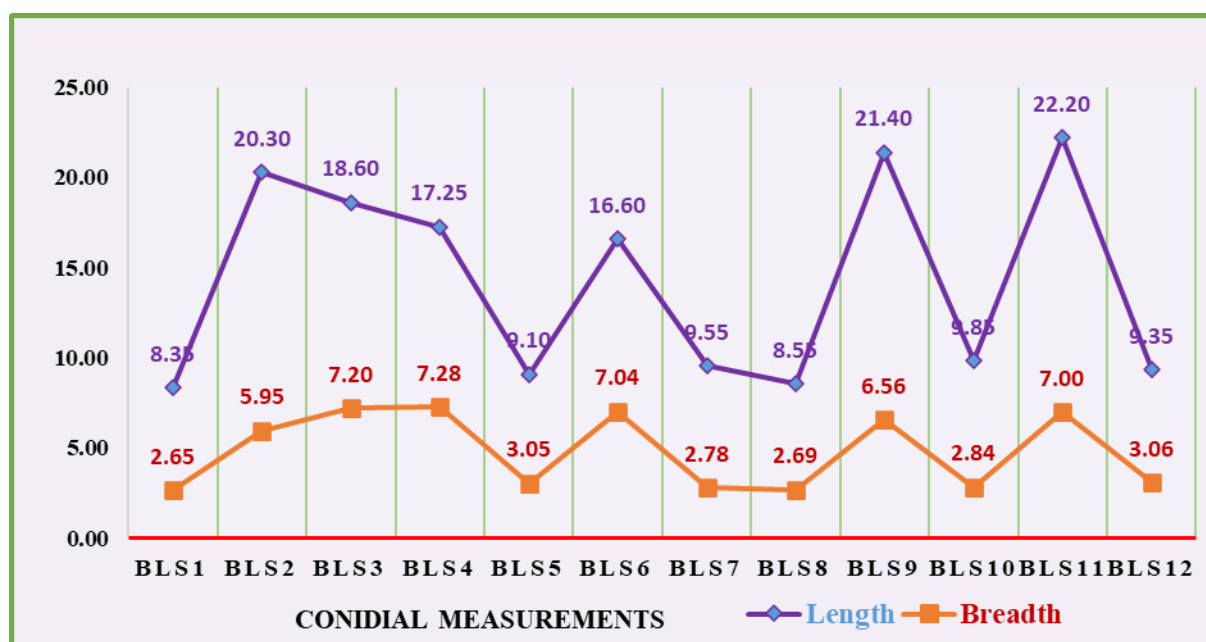
**Table 2:** Variation in conidial measurements of different isolates of *P. grisea* on PDA.

Sr. No.	Isolates	Length ( $\mu\text{m}$ )*		Breadth ( $\mu\text{m}$ )*		Mean LxB ( $\mu\text{m}$ )
		Range	Mean	Range	Mean	
1.	BLS-1	06.60 - 10.10	8.35	2.10 - 3.20	2.65	08.35 × 2.65
2.	BLS-2	18.60 - 22.00	20.30	4.50 - 7.40	5.95	20.30 × 5.95
3.	BLS-3	16.10 - 21.10	18.60	6.60 - 7.80	7.20	18.60 × 7.20
4.	BLS-4	15.20 - 19.30	17.25	5.40 - 9.16	7.28	17.25 × 7.28
5.	BLS-5	08.00 - 10.20	9.10	2.49 - 3.61	3.05	09.10 × 3.05
6.	BLS-6	12.20 - 21.00	16.60	6.16 - 7.91	7.04	16.60 × 7.04
7.	BLS-7	08.60 - 10.50	9.55	2.19 - 3.36	2.78	09.55 × 2.78
8.	BLS-8	06.90 - 10.20	8.55	2.14 - 3.24	2.69	08.55 × 2.69
9.	BLS-9	19.60 - 23.20	21.40	5.90 - 7.21	6.56	21.40 × 6.56
10.	BLS-10	08.80 - 10.90	9.85	2.39 - 3.28	2.84	09.85 × 2.84
11.	BLS-11	21.10 - 23.30	22.20	6.60 - 7.40	7.00	22.20 × 7.00
12.	BLS-12	08.10 - 10.60	9.35	2.63 - 3.49	3.06	09.35 × 3.06
SE (m) +		0.21		0.16		-
CD @ 1%		0.82		0.62		-

\* Mean of 50 observations

**Table 3:** *P. grisea* isolates grouped on the basis of conidial length.

Conidia with higher length	20.00 - 22.20 $\mu\text{m}$	BLS-2, BLS-9 and BLS-11
Conidia with moderate length	19.90 - 10.00 $\mu\text{m}$	BLS-6, BLS-4 and BLS-3
Conidia with shorter length	9.90 - 8.00 $\mu\text{m}$	BLS-1, BLS-5, BLS-7, BLS-8, BLS-10 and BLS-12.

**Fig 1:** Variation in conidial measurements ( $\mu\text{m}$ ) of different isolates of *P. grisea* on PDA

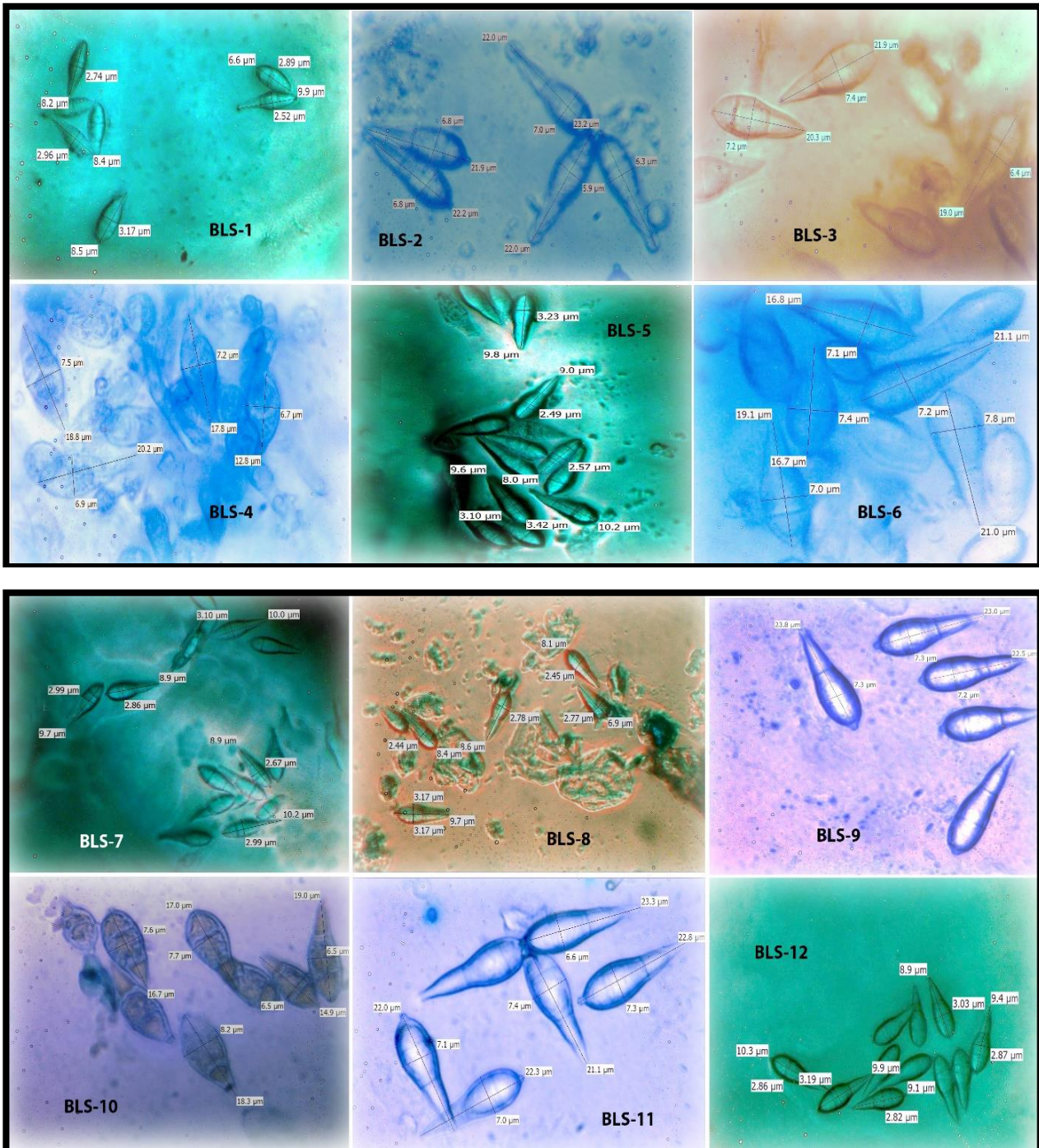


Plate 1: Variation in conidial measurements (µm) of different isolates of *P. grisea*.



Plate 2: Conidiophore and conidia of *P. grisea*

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