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Variability in Isolates of Rice Brown Spot Pathogen, *Bipolaris oryzae* in Andhra Pradesh

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ABSTRACT

Bipolaris oryzae isolates obtained from six geographically distant rice growing locations of Andhra Pradesh differed marginally in colour and type of colony growth in culture. All the isolates exhibited a cottony growth and appeared whitish initially and turned dark brown with age on Czapek dox agar, potato dextrose agar and rice leaf extract agar. Isolates significantly differed in initiation and intensity of sporulation as also in spore dimensions. Longer and wider spores were generally observed when the isolates were cultured on potato dextrose agar and rice leaf extract agar than on Czapek dox agar. Isolates exhibited varied pathogenic ability on rice genotypes with both culture filtrate and spore suspension inoculations in terms of incubation period and spot size.

Key words : Brown spot, *Bipolaris oryzae*, Rice, Variability.

Bipolaris oryzae (Breda de Haan) Shoemaker is one of the important pathogens causing brown spot disease in all rice producing areas of the world. The pathogen attacks rice starting from germination causing leaf blight to grain discolouration at later stages. The disease occurs regularly every year in mild to severe form occasionally taking epiphytotic proportions. Prevalence of brown spot disease of rice is on the increase in recent years causing loss in yield, and quality in terms of grain discolouration. The disease caused yield loss of even 100% and was the cause of the historically significant Bengal famine.

Variability in *B. oryzae* is reported from several parts of the world (Misra and Chatterjee, 1963; Estrada *et al*., 1982 and Weikert-Oliveira *et al*., 2002). Variable rice varietal reaction to brown spot reported by several workers (Padmanabhan *et al*., 1966 and Kanjanasoon and Sitthichai, 1967) may have relevance to the variability in the pathogen populations. In view of the lack of any knowledge about the possible variations in *B. oryzae* in Andhra Pradesh, which is an important rice growing state of India, the present investigation was envisaged to assess cultural, morphological and pathogenic variability of isolates.

MATERIAL AND METHODS

Cultures of *B. oryzae* were isolated from brown spot affected rice leaves collected from Bapatla (Guntur district), Maruteru (West Godavri district), Nellore (Potti Sriramulu Nellore district), Ragolu (Srikakulam district), Palem (Mahaboobnagar district) and Khammam (Khammam district) and maintained in pure culture on potato dextrose agar (PDA). The isolates were designated as Bpt, Mtu, Nlr, Sklm, Plm and Khm, respectively. Variation in radial growth of isolates on three different nutrient media, Czapek dox agar (CDA), PDA and rice leaf extract agar (RLE) was assessed. A 3-mm disc cut from a 7-day old culture of each isolate and inoculated on to the medium at the centre of each plate and incubated. Radial growth of each isolate on the three media was measured daily on two axes and the mean diameter of the colony was calculated.

The duration from inoculation to start of sporulation of each isolate on each medium was recorded. Spores from colony were harvested by gently scraping the colony with a sterilized inoculation needle in 10 ml of water and the spore suspension was collected in a test tube. After thorough stirring of the spore suspension, spore concentration ml^{-1} was determined five times and the average number of spores ml^{-1} was calculated for each isolate on the three nutrient media.

Spore dimensions were determined by observing under a microscope. Length and width of 100 spores were measured using a Labomed Lx 400 microscope with ProgRes CapturePro 2.5 version software (Labo America Inc., Fremont, California, USA). The range and average for length and width of 100 spores for each isolate on all the nutrient media were arrived.

Rice genotypes from the Rice Research Unit, Bapatla *viz.,* BPT 1768, BPT 3291, BPT 2444, BPT 2448, BPT 2406, BPT 2425, BPT 2270, NLR 3041, NLR 40058, NLR 20017, NLR 20023 and NLR 145 were selected for studying the pathogenic variability of the *B. oryzae* isolates. Pathogenic variability among isolates was determined by inoculating with propagule free culture filtrate and also with spore suspension of the isolates.

The pathogen isolates were grown in Czapek dox broth (CDB) for 10 days. The mycelial mat was removed and the filtrate was passed through two layers of sterilized Whatman No. 41 filter paper twice to ensure freedom from spore or mycelial fragments. Detached rice leaf pieces (5-7 cm long) from four weeks old seedlings were cut and floated on benzimidazole (25 ppm) solution in sterile Petri plates. The leaf pieces were inoculated by placing 20 µl of the culture filtrate on the floating leaf bit using a micropipette. Petri dishes with inoculated leaf bits were placed at room temperature (~ 30^0 C) and observed for symptoms of the effect of the culture filtrates. Four replications were maintained for each genotype. Leaf bits treated with sterilized CDB served as controls. Pathogenic variability was assessed by the lesion size and lesion description in inoculated rice genotypes. Earthen pots (25 cm × 20 cm size) were used for raising the rice plants in which potting medium (sand, loamy soil and farm yard in 1:2:1 ratio) was added. Twelve rice genotypes were planted in separate pots at three seedlings per pot. Four weeks old seedlings were then artificially inoculated with conidial suspension (1×10^4 spores/ml) of *B. oryzae* isolates using an atomizer. The plants sprayed with conidial suspension were covered for 16 h with polythene bags moistened inside to maintain high relative humidity. Plants sprayed with sterile distilled water without conidia served as control.

Incubation period (IP) was recorded in each genotype for each isolate by recording the time taken for appearance of first symptom after inoculation.

Latent period (LP) was determined by removing the leaf bits when the first symptom was observed from the inoculated plants in pots. The leaf bits were immediately stuck to the inside of the lid of Petri dish. The bottom plate of the Petri dish was lined with sterilized moist blotter papers over which a sterilized microscope slide was kept for collecting spores discharged from the spots. The slides were observed under a microscope daily for the presence of release of spores. The period between inoculation and first observation of spore release from the spots was recorded as LP.

RESULTS AND DISCUSSION

Bipolaris oryzae isolates varied for cultural, morphological characters and pathogenic ability. Isolates differed marginally in colour and type of colony growth in culture. All the isolates exhibited a cottony growth and appeared whitish initially and turned dark brown with age on CDA, PDA and RLE. Nlr and Sklm isolates showed slight variation with change in culture medium with prolonged incubation and with change in culture medium. On CDA or PDA, the colony colour of Nlr isolate was deep black where as on RLE it was dark brown. Sklm isolate was deep black on PDA or RLE whereas on CDA it was greenish black that gradually turned grayish to blackish in colour

Isolates exhibited wide variation in radial growth on three culture media CDA, PDA and RLE. Bpt isolate was the fastest while Mtu isolate was the slowest in radial growth. PDA supported better radial growth than CDA and RLE for most isolates. *Helminthosporium oryzae* (syn: *B. oryzae*) was reported to grow better on PDA than on several culture media including CDA and host leaf extract agar (Misra and Chatterjee, 1963). However, in this study Mtu isolate had shown a preference for RLE (host leaf extract) than PDA and CDA indicating variability among *B. oryzae* isolates with regards to culture medium preference. The results of this study corroborate the findings of Kumar *et al.* (2011) who reported colony and morphological variability of isolates of *B. oryzae* on PDA (Table 1).

Bipolaris oryzae isolates significantly varied in time taken for sporulation after inoculation. Bpt was the earliest to initiate sporulation (13.7 days) on culture media while Nlr isolate was the last (40.7 days). Initiation of sporulation varied between 16.7 and 30.3 days for other isolates. Mean time taken by the isolates for sporulation significantly varied on the three culture media. Isolates sporulated significantly earlier on CDA (23.8 days) than on RLE (28.5 days) and PDA (26.5 days) (Table 2). Significant variation was observed among isolates for sporulation on the three culture media. Mtu isolate produced significantly highest number of spores (33×10^4 spores/ml) while Nlr isolate produced the least number of spores (13.8×10^4 spores/ml). Sporulation in other isolates varied between 15×10^4 spores/ml and 29.2×10^4 spores/ml. Variation among *B. oryzae* isolates for sporulation was reported by several workers (Mukherjee, 1960; Matsmuhra, 1927; Nisikado, 1927; Misra and Chatterjee, 1963; Stevens, 1922). Stevens (1922) and Misra and Chatterjee (1963) reported an inverse relationship between linear

Table 1. Radial growth (cm) of *B. oryzae* isolates on different culture media.

	Day 1			Day 2			Day 3			Day 4			Day 5			Day 6			Day 7			Mean
Isolate	CDA	PDA	RLE	CDA	PDA	RLE	CDA	PDA	RLE	CDA	PDA	RLE	CDA	PDA	RLE	CDA	PDA	RLE	CDA	PDA	RLE	
Mtu	0.3	0.5	0.6	1.6	2.0	2.0	2.3	2.4	2.6	2.5	2.6	3.5	3.7	3.4	4.8	4.5	3.8	5.2	4.8	4.0	5.3	3.0
Sklm	1.2	1.1	1.2	2.5	2.9	2.6	3.9	4.6	4.1	5.9	7.4	6.0	8.4	8.2	7.5	8.7	9.0	8.8	9.0	9.0	9.0	5.8
Plm	0.9	1.1	1.1	2.1	2.5	2.3	3.4	3.5	3.2	4.4	3.6	3.5	5.2	6.5	4.9	6.4	7.3	5.7	8.8	9.0	7.5	4.4
Nlr	0.7	1.1	1.1	1.8	2.4	2.4	3.3	4.3	3.5	4.8	6.8	5.0	6.1	8.6	6.6	7.3	9.0	7.9	8.9	9.0	9.0	5.2
Bpt	2.1	3.3	3.1	6.7	8.7	8.8	9.0	9.0	9.0	9.0	9.0	9.0	9.0	9.0	9.0	9.0	9.0	9.0	9.0	9.0	9.0	8.0
Khm	0.7	0.7	0.9	2.6	3.0	2.5	4.1	4.5	3.6	4.8	6.3	4.6	6.4	8.6	6.0	8.1	9.0	6.3	9.0	9.0	7.2	5.1
Mean	0.9	1.3	1.3	2.9	3.6	3.4	4.3	4.7	4.3	5.2	6.0	5.3	6.5	7.4	6.5	7.3	7.9	7.2	8.3	8.2	7.8	
CD ($P \leq 0.05$) for isolates													0.3									
CD ($P \leq 0.05$) for media													0.2									
CD ($P \leq 0.05$) for days													0.3									
CD ($P \leq 0.05$) for isolates× media													0.5									
CD ($P \leq 0.05$) for isolates × days													0.7									
CD ($P \leq 0.05$) for media × days													N.S									
CV (%)													8.8									

Table 2. Days required for the sporulation of *B. oryzae* isolates on culture media.

Media	Isolates						Mean
	Mtu	Sklm	Plm	Nlr	Bpt	Khm	
CDA	15.0	29.0	26.0	38.0	13.0	21.0	23.8
PDA	18.0	34.0	28.0	42.0	15.0	23.0	26.5
RLE	17.0	29.0	26.0	42.0	13.0	22.0	24.8
Mean	16.7	30.3	26.7	40.7	13.7	22.3	
CD ($P \leq 0.05$)) for isolates			0.7				
CD ($P \leq 0.05$) for media			1.0				
CD ($P \leq 0.05$) for isolates × media			1.8				
CV (%)			5.0				

Table 2 contd… Sporulation of *B. oryzae* isolates on different culture.

Media	Isolates						Mean
	Mtu	Sklm	Plm	Nlr	Bpt	Khm	
CDA	34.10	16.25	21.40	15.60	29.55	22.00	23.15
PDA	31.05	12.90	19.00	12.80	27.40	16.70	19.98
RLE	33.75	15.95	15.35	13.10	30.73	25.00	22.31
Mean	32.97	15.03	18.58	13.83	29.23	21.23	
CD ($P \leq 0.05$) for isolates			0.33				
CD ($P \leq 0.05$) for media			0.47				
CD ($P \leq 0.05$) for isolates × media			0.82				
CV (%)			2.64				

growth and conidial production. In this study also the slow growing Mtu isolate produced the highest number of conidia compared to other fast growing isolates. This observation however does not hold true for Bpt isolate which produced significantly higher number of conidia than the slow growing Sklm, Nlr, Khm and Plm isolates. Producers of high number of conidia were also significantly the earliest in initiation of sporulation. Observations on the effect of culture medium on the growth and sporulation revealed that the slow linear growth supporting CDA and RLE promoted significantly higher number and early production of spores than PDA (Table 2). Media rich in host parts were proved to support more sporulation by Chattopadhyay and DasGupta (1965) and Sharma and Singh (1975). Other media that promoted good sporulation of *B. oryzae* isolates were rabbit food agar (Hau and Rush, 1980) and V-8 agar (Ojeda and Subero, 2006). Isolates were found to vary in spore production on PDA (Kumar *et al.*, 2011).

Spore dimensions of *B. oryzae* varied among isolates. The longest and widest spores were produced by Khm isolate (101.2 × 15.6 µm) while the shortest and narrowest spores were in Plm isolate (60.5 × 12.5 µm). Longer and wider spores were generally observed when the isolates were cultured on RLE and PDA than on CDA. Spore length and width of all the *B. oryzae* isolates on the three media (32.3 to 153.2 µ × 7.7 to 20.3 µ) were within the ranges reported from different countries like China (Wei, 1957), USA (Drechsler, 1923), India (Sundararaman, 1922), Japan (Nisikado and Miyake, 1922) and Java (Breda de Haan, 1900) (Table 3).

Isolates varied in their pathogenic ability in both culture filtrate and spore suspension inoculations. Culture filtrate of isolates could cause necrotic spots with a water soaked halo on leaves of rice genotypes indicating elaboration of a toxic principle in their culture filtrates. Largest spots were

Table 3. Variations in spore measurements of *B. oryzae* isolates on different culture media.

Isolates	Spore Length (µm)				Spore Width (µm)			
	Range				Range			
	CDA	PDA	RLE	Mean	CDA	PDA	RLE	Mean
Mtu	32.3 – 103.1 (76.3)*	41.6 – 105.2 (76.0)	51.6 – 89.7 (70.9)	74.4	7.7 – 20.1 (14.2)	9.7 – 24.7 (14.9)	11.1 – 20.8 (14.7)	14.6
Sklm	50.6 – 99.1 (76.6)	59.9 – 95.7 (82.1)	48.5 – 102.4 (80.7)	79.8	9.0 – 17.5 (12.8)	11.2 – 20.6 (16.2)	10.3 – 17.5 (13.9)	14.3
Plm	45.5 – 83.8 (63.9)	38.1 – 88.9 (58.7)	45.7 – 104.8 (58.8)	60.5	9.0 – 16.0 (12.2)	8.6 – 16.2 (12.3)	9.2 – 20.6 (13.0)	12.5
Nlr	61.2 – 104.4 (82.0)	39.7 – 88.1 (67.8)	42.7 – 106.2 (67.8)	72.3	10.8 – 15.8 (13.0)	8.4 – 16.1 (13.0)	12.8 – 20.2 (16.5)	14.2
Bpt	60.1 – 90.1 (73.5)	53.2 – 97.2 (72.3)	72.2 – 96.7 (82.2)	76.0	11.1 – 15.6 (13.5)	10.0 – 16.9 (13.9)	12.2 – 17.7 (14.8)	14.1
Khm	48.2 – 134.8 (87.3)	77.5 – 140.7 (107.3)	61.5 – 153.2 (108.9)	101.2	8.9 – 22.1 (15.9)	11.1 – 21.6 (15.9)	10.7 – 17.8 (15.0)	15.6
Mean	76.6	77.4	78.2		13.6	14.4	14.6	
CD ($P \leq 0.05$) for isolates			1.9			0.5		
CD ($P \leq 0.05$) for media			NS			0.3		
CD ($P \leq 0.05$) for isolates × media			3.3			0.8		
CV (%)			3.0			4.1		

- Figures in parentheses are mean values

produced by culture filtrate of Plm isolate (2.7 mm) and the smallest spots were observed in inoculations with culture filtrate of Nlr isolate (2.1 mm) (Table 4). Rice genotypes showed differential sensitivity to culture filtrates of isolates indicating variation in genotype – isolate interaction. Longest mean spot size (3 mm) was observed in Nlr 3041 genotype and the smallest was in Bpt 2425 (1.9 mm). Observations reveal that some pathogenic principle is produced by the isolates when grown on CDB. Vidhyasekaran *et al.* (1986) characterized the toxin, ophiobolin produced by *H. oryzae* which caused characteristic brown spot symptoms on rice leaves. Elaboration of toxins in culture filtrates of *H. oryzae* was detected by Simhachalam (1971). Variability between the effects of culture filtrates of two *H. oryzae* isolates on rice cultivars was observed by Narain and Simhachalam (1976).

Incubation period varied with pathogen isolate and host genotype when inoculated with spore suspension in pot culture. Incubation period was the longest for Khm isolate (21.86 h) while the shortest was with Bpt isolate (20.36 h). Rice genotypes too showed significant variation in IP ranging from 16.72 h (Nlr 145) to 23.06 h (Bpt 2425) (Table 5). All the isolates produced symptoms within 24 h in all the genotypes which was in agreement with the observations of several workers (Baruah *et al.*, 1980; Sherf *et al.*, 1947; Ocfemia, 1924). Latent period in rice genotypes although varied did not differ significantly. Bpt genotypes in general recorded longer LP (94 – 99.17 h) than Nlr genotypes (85.82 – 94 h) which trend was similar to that observed for IP (Table 6). The LP taken by isolates in some genotypes (96 h) was in agreement with the observations of Baruah *et al.* (1980), while in other genotypes it was either slightly shorter or longer. IP and LP are good indicators of the onset and spread of an epidemic (Vanderplank, 1963). In general reddish brown spots measuring 1.1 – 3.17 mm dia were observed in different rice genotypes inoculated with the six *B. oryzae* isolates. However, Bpt isolate produced a dark reddish brown, close to black spot on BPT 1768 genotype. Mean spot size for isolates differed significantly. Mean spot size for Plm isolate (2.14 mm) was significantly the largest followed by that of Bpt isolate (2.02 mm). The

Table 4. Variability in effect of culture filtrates (mm) of *B. oryzae* isolates on detached leaves of rice genotypes.

Isolates	Spot sizes in different genotypes (mm)												
	NLR 3041	NLR 40058	NLR 145	NLR 20023	NLR 20017	BPT 2425	BPT 1768	BPT 2406	BPT 3291	BPT 2270	BPT 2448	BPT 2444	Mean
Mtu	2.75	1.75	3.00	2.25	2.75	2.25	1.63	1.75	2.75	1.75	2.13	2.13	2.24
Sklm	3.25	1.88	2.75	1.63	2.25	1.63	2.13	1.50	2.25	2.25	3.75	4.25	2.46
Plm	4.40	1.75	3.25	2.25	1.75	2.25	2.25	3.50	4.25	1.75	2.25	2.38	2.67
Nlr	1.75	3.88	3.50	1.50	1.75	1.75	2.25	1.50	2.25	1.50	2.25	1.75	2.14
Bpt	2.50	3.12	2.25	2.25	2.75	2.25	1.75	2.25	3.25	2.00	2.25	2.13	2.39
Khm	3.25	1.50	2.25	2.75	2.75	1.50	3.63	1.75	2.63	2.75	3.00	2.25	2.50
Mean	2.98	2.31	2.83	2.10	2.33	1.94	2.27	2.04	2.90	2.00	2.60	2.48	
CD ($P \leq 0.05$)) for isolates									0.10				
CD ($P \leq 0.05$) for genotypes									0.15				
CD ($P \leq 0.05$) for isolates × genotypes									0.36				
CV (%)									10.6				

Table 5. Variation in incubation period (h) in rice genotypes inoculated with spore suspension of *B. oryzae* isolates.

Isolates	IP in different genotypes (h)												
	NLR 3041	NLR 40058	NLR 145	NLR 20023	NLR 20017	BPT 2425	BPT 1768	BPT 2406	BPT 3291	BPT 2270	BPT 2448	BPT 2444	Mean
Mtu	20.33	21.00	16.00	19.33	21.67	23.67	23.67	20.33	23.33	23.33	21.00	23.00	21.39
Sklm	18.33	18.67	17.00	20.67	19.33	22.33	20.67	22.67	23.67	20.67	23.00	23.67	20.89
Plm	18.33	19.33	17.33	19.33	20.33	23.33	23.67	22.33	22.67	23.67	20.00	21.00	20.94
Nlr	18.67	19.67	17.33	19.00	20.67	23.67	22.33	20.00	21.67	22.67	19.33	21.33	20.53
Bpt	18.33	19.33	15.67	20.33	18.33	21.67	21.67	20.33	22.67	22.00	23.33	20.67	20.36
Khm	20.33	21.33	17.00	20.33	22.67	23.67	23.67	22.67	20.67	23.00	23.67	23.33	21.86
Mean	19.06	19.89	16.72	19.83	20.50	23.06	22.61	21.39	22.44	22.56	21.72	22.17	
CD ($P \leq 0.05$)) for isolates									0.40				
CD ($P \leq 0.05$) for genotypes									0.56				
CD ($P \leq 0.05$) for isolates × genotypes									1.38				
CV (%)									4.07				

Table 6. Variation in latent period (h) in rice genotypes inoculated with spore suspension of *B. oryzae* isolates

Isolates	LP in different genotypes (h)												
	NLR 3041	NLR 40058	NLR 145	NLR 20023	NLR 20017	BPT 2425	BPT 1768	BPT 2406	BPT 3291	BPT 2270	BPT 2448	BPT 2444	Mean
Mtu	76.67	80.67	84.00	80.00	82.00	86.00	89.33	83.33	88.00	90.67	86.67	84.67	84.33
Sklm	82.67	92.00	86.00	77.33	89.33	120.00	90.00	119.33	90.67	84.00	94.00	90.67	93.00
Plm	92.33	93.00	89.33	85.00	98.67	102.33	103.00	96.00	117.33	112.67	108.33	112.67	100.89
Nlr	109.00	107.67	105.00	101.67	120.00	115.67	109.67	107.00	104.00	110.67	101.00	119.00	109.19
Bpt	88.67	95.33	74.00	76.00	85.33	75.33	76.67	80.00	87.00	81.33	78.00	93.33	82.58
Khm	94.00	95.33	89.33	94.67	89.33	87.33	88.33	90.00	93.67	91.33	96.00	94.67	92.00
Mean	90.56	94.00	87.94	85.78	94.11	97.78	92.83	95.94	96.78	95.11	94.00	99.17	
CD ($P \leq 0.05$)) for isolates									6.72				
CD ($P \leq 0.05$) for genotypes									NS				
CD ($P \leq 0.05$) for isolates × genotypes									NS				
CV (%)									15.40				

Table 7. Variation in spot size (mm) on rice genotypes inoculated with spore suspension of *B. oryzae* isolates

Isolates	Spot sizes in different genotypes (mm)												
	NLR 3041	NLR 40058	NLR 145	NLR 20023	NLR 20017	BPT 2425	BPT 1768	BPT 2406	BPT 3291	BPT 2270	BPT 2448	BPT 2444	Mean
Mtu	3.17	2.17	3.37	1.33	1.50	1.20	2.17	1.80	2.50	1.00	1.10	1.27	1.88
Sklm	2.50	1.43	1.17	1.17	1.10	2.20	1.10	1.57	1.10	1.57	1.43	1.77	1.51
Plm	2.90	1.17	1.90	2.50	1.43	2.83	2.43	1.50	2.43	3.10	1.83	1.67	2.14
Nlr	2.50	1.83	1.77	1.17	1.60	1.17	1.50	2.23	2.27	2.43	1.60	3.00	1.92
Bpt	2.50	2.67	1.90	1.20	2.10	1.17	1.70	2.50	2.27	2.73	1.70	1.77	2.02
Khm	1.60	1.80	1.67	2.37	1.70	3.27	2.33	1.67	1.83	1.17	1.57	1.00	1.83
Mean	2.53	1.84	1.96	1.62	1.57	1.97	1.87	1.88	2.07	2.00	1.54	1.74	
CD ($P \leq 0.05$)) for isolates									0.10				
CD ($P \leq 0.05$) for genotypes									0.14				
CD ($P \leq 0.05$) for isolates × genotypes									1.33				
CV (%)									10.90				

smallest mean spot size was observed for Sklm isolate (1.51 mm). The mean spot size for Khm (1.83 mm), Mtu (1.8 mm) and Nlr (1.92 mm) isolates was on a par (Table 7).

Rice genotypes also varied significantly for mean spot size caused by the six *B. oryzae* isolates. The largest mean spot size was observed in NLR 3041 (2.53 mm) which was significantly more than the spot sizes recorded in other rice genotypes. The smallest mean spot size was produced in BPT 2448 (1.54 mm) which was on a par with the spot size recorded in NLR 20017 (1.57 mm) and NLR 20023 (1.62 mm). Mean spot size in other rice genotypes varied between 1.74 mm in BPT 2444 and 2.07 mm in BPT 3291 genotype.

Genotype × isolate interaction was also found to be significant for spot size. Mtu isolate produced largest spots in NLR 145 (3.37 mm) and NLR 3041 (3.17 mm) and produced the smallest spots in BPT 2270 (1.00 mm). The largest spot produced by Sklm isolate was in NLR 3041 (2.5 mm) while the smallest was (1.10 mm) in BPT 3291 and NLR 20017. Plm isolate produced the largest spots in BPT 2270 genotype (3.10 mm) followed by NLR 3041 (2.90 mm) and produced the smallest in NLR 40058 (1.17 mm). Nlr isolate produced the largest spot in NLR 3041 (2.5 mm) while the smallest was BPT 2425 and NLR 20023 (1.17 mm). The largest spot for Bpt isolate was produced in BPT 2270 (2.73 mm) while smallest was in BPT 2425 (1.17 mm). Khm isolate caused the largest spot (3.27 mm) in BPT 2425 and the smallest spot in BPT 2444. Sato (1965) considered spot size as an important criterion for categorising rice genotypes. Rice genotypes were classified in to minute spot (< 0.5 mm), medium spot (0.6 – 1.0 mm) and large spot (1 x 2 mm or above) categories. Accordingly, rice genotypes in the present investigation belong to large spot category of Sato.

LITERATURE CITED

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