

**Screening and evaluation of maize genotypes against banded leaf and sheath blight disease
under artificial epiphytotic conditions**

ABSTRACT

Maize is important cereal crop and provides nutrition and basic raw material for various industries. Its production is hampered by various biotic and abiotic factors. Among various pathogens attacking maize, banded leaf and sheath blight (BLSB) caused by *Rhizoctonia solani* f.sp. *sasakii* is a very serious impediment for quality maize production. There is very less availability of resistance sources against this disease. There is a need to develop resistant varieties to overcome the health hazards associated with the excessive use of chemical pesticides. Keeping in view the importance of the disease, the present investigation was carried out at Regional Research Station Uchani, Karnal of CCS Haryana Agricultural University, Hisar and twenty seven hybrids and sixty inbred lines of maize were screened and scored for their disease reaction against banded leaf and sheath blight disease to find out new sources of resistance so that it can boost future maize breeding programmes.

Key words: Banded leaf and sheath blight, Disease management, Maize, *Rhizoctonia solani* f. sp. *sasakii*, Screening

1. INTRODUCTION

Maize (*Zea mays* L.) is one of the most admired, adaptable crop grown in different agro-climatic conditions. Due to increasing population, maize has an immense role to food security worldwide and key cereal crop which is grown over 150 countries. Maize is considered as a miracle C₄ plant

having highest yielding potential among cereals. It is also known as “Queen of cereals”. A diet rich of cereals contribute in controlling cholesterol levels and blood pressure hence playing important role in disease prevention. In comparison to wheat and rice, maize contains about 72 per cent starch, 10 per cent protein and 4 per cent fat (Nuss and Tanumihardjo, 2010). It contributes approximately 9 per cent to food basket of India and 5 per cent to world’s dietary energy supply. Maize is also considered as one of the largest consumable cereal of the world (Kumar *et al.*, 2018). Major maize growing countries include USA, China, Brazil, France, Mexico and India. In India, it is being used as major source of feed (63%), food (23%), industries (12%) and only 2 per cent is utilized as seed (Malik *et al.*, 2018). In India, maize is grown throughout the year but during *Kharif* season it is grown over 85 per cent of the cultivated area in India (USDA, 2021). Despite of various pathogens that attacking maize crop, the banded leaf and sheath blight (BLSB) disease caused by *Rhizoctonia solani* f.sp. *sasakii* is of vital importance and has been reported from many maize growing states including Haryana, Punjab, Rajasthan, Himachal Pardesh and Uttar Pardesh and pose threat to its production and major constraint for quality maize production (Devi and Thakur, 2016). This disease was first time described and reported this disease from Sri Lanka as sclerotial disease of maize caused by *Rhizoctonia solani* Kuhn (Bertus, 1927). In India, BLSB disease of maize was first time reported from Terai region of Uttar Pardesh (Payak and Renfro, 1966). It is becoming a major disease of concern in maize growing areas as its damage ranges from 11 per cent to 40 per cent and even 100 per cent in areas where environmental conditions are highly favorable to the *R. solani* pathogen (Kumar and Singh, 2004; Madhavi *et al.*, 2011; Izhar and Chakraborty 2013; Gao *et al.*, 2014; Hooda *et al.*, 2017). Symptoms of the disease begin to appear on the leaves and sheath in 40-50 days older plants and later stages when the infection increases, the ear can also be

infected. The pre-flowering stage is most affected by the disease and the rings formed by lesions can be noticed on the lower leaves and sheath also. In the early stages of infection, the plant produces globular to elongated bands (1-3 mm thick in diameter) that appear as water-soaked lesions. Under favorable weather conditions, the symptoms may extend to silk, glumes and grains (Ahuja and Payak, 1982; Saxena, 2002). Fungicides are used as primary means for managing plant diseases but these are costly and pose negative impacts on environment and human health. Development of resistance among plant pathogens due to indiscriminate use of fungicides is also observed and to there is a need to find out environment friendly, non-toxic and cost effective methods to mitigate the health related risks and to overcome the negative impacts of plant diseases. There is a limited source of resistance against BLSB disease and it is considered as main bottleneck to accomplish any breeding programme effectively. Keeping in view the importance of the disease, the present investigation was carried out and twenty seven hybrids and sixty inbred lines of maize were screened and scored for their disease reaction against banded leaf and sheath blight disease to find out new sources of resistance so that it can boost future maize breeding programmes.

2. MATERIALS AND METHODS

The performance of maize hybrids and inbred lines was evaluated at Regional Research Station Uchani, Karnal of CCS Haryana Agricultural University situated at 29°43' North latitude and 76°58' East longitude at an altitude of 245 m from mean sea level. Crop was raised during *Kharif* season in 2018 by following package of practice of Haryana state (Anonymous, 2017). In this study, twenty seven hybrids and sixty inbred lines of maize were screened.

2.1 Isolation of the Pathogen

Diseased leaves and sclerotia collected from previous season maize crop were used for isolation of pathogen. The diseased bits of leaves and sclerotia were washed under running tap water and then dipped into sodium hypochlorite (0.1 %) solution for surface sterilization for 30-50 seconds. Following this step, two times washing was done to remove the traces of sodium hypochlorite. These bits were placed in PDA slants and Petri plates under aseptic conditions. For providing proper growth conditions, there were placed in biological oxygen demand (BOD) incubator at $28\pm 1^{\circ}\text{C}$. So, culture of fungus was obtained and purified by hyphal tip method and regular sub culturing was done after every 15 days and stored at $5\pm 1^{\circ}\text{C}$ in refrigerator.

2.2 Artificial inoculation under field conditions

Maize plants were subjected to pathogen by following the artificial inoculations in the field done twice at 30 days after sowing (DAS) and 45 DAS by incorporating a grain culture (using 4-5 barley grains) with the help of a needle directly from the Petri plate on the stem and the second or third internode on the plants. Diseased leaves and sclerotia collected from previous year's crop were also used for artificial inoculation. The infected leaves were soaked in water for two days and the solution was sprayed on the maize plants to facilitate infection. Sclerotia of *R. solani* pathogen were crushed and healthy plants were dusted with this sclerotial powder. To provide moist conditions and to assist infection, the crop was sprayed with water daily for a week. The crop exhibited characteristic symptoms of the infection.

2.3 Observations

The observations on per cent disease incidence and severity were recorded. Disease reaction of maize hybrids and inbred lines was determined by using BLSB rating scale (1-9) given in Table 1 (Anonymous, 2016).

Disease incidence was recorded and calculated by using the formula of Goswami *et al.* (2002).

$$\text{Disease incidence} = \frac{\text{No. of infected plants}}{\text{Total no. of plant assessed}} \times 100$$

Disease severity was calculated by the formula given by AICRP on maize (Anonymous, 2016).

$$\text{Per cent Disease severity} = \frac{\text{Sum of all disease rating}}{\text{Total no. of rating} \times \text{maximum disease grade}} \times 100$$

3. RESULTS

In this investigation, twenty seven hybrids and sixty inbred lines of maize were screened for their relative resistance against *R. solani* under artificial inoculation conditions in field during *Kharif* 2018. The results in table 2 and 3 revealed that seven maize hybrids *viz.*, HM-8, HKI 295×L287, HKI 327T×L287, HKI 1041-2×L287, HKI 1663×193-2, PC-8ER-2×PC SONA and PCMYD-7×PC SONA showed resistant reaction (≤ 33.33 % disease severity) against *R. solani* under artificial inoculation conditions. However, twenty maize hybrids *viz.*, HM-11, HM-13, HKI 193-1×L287, HKI 323×L287, HKI 4887×L287, HKI 1042×193-1, HKI 1670×193-1, HKI 1041-2×193-1, HKI 1042×193-2, HKI 1659×193-2, HKI 1662×193-2, HKI 1664×193-2, HKI 1670×193-2, HKI 295×323, HKI 327T×488, HKI 1040-7×1105, PC-3ER-3×PC SONA, PC-7ER-5×PC SONA, PCMYD-8×PC SONA and PALL×PC SONA showed moderately resistant reaction (33.34-55.55 % disease severity) to *R. solani*. None of the maize hybrids showed moderately susceptible to susceptible reaction even under artificial inoculation conditions.

Out of sixty inbred lines of maize one inbred line *i.e.* HKI 1663 LM-6 was not germinated. However, only two inbred lines *viz.*, HKI 1128 and HKI 1345 W of maize showed resistant reaction (≤ 33.33 % disease severity), eight inbred lines namely HKI 164-7-4, HKI 193-1, HKI 193-2, HKI 288-2, HKI 488, MBR-139, HKI 1378 W and PC-1ER-4 showed moderately resistant reaction (33.34-55.55 % disease severity) against this pathogen under artificial inoculation conditions (Table 4 & 5). Rest of the inbred lines of maize showed moderately susceptible to susceptible reaction against this necrotrophic pathogen under field conditions.

4. Discussion

Host plant resistance is one of the most promising approaches for managing plant diseases. Controlling plant diseases with resistance breeding programme is a most efficient, eco-friendly and economical method. To date, complete resistance to the pathogen has not been identified. Twenty seven hybrids and sixty inbred lines of maize were screened for their relative resistance against *R. solani* under artificial inoculation conditions in field. Out of twenty seven maize hybrids, seven hybrids *viz.*, HM-8, HKI 295×L287, HKI 327T×L287, HKI 1041-2×L287, HKI 1663×193-2, PC-8ER-2×PC SONA and PCMYD-7×PC SONA showed resistant reaction (≤ 33.33 % disease severity) against *R. solani* under artificial inoculation conditions. However, twenty maize hybrids *viz.*, HM-11, HM-13, HKI 193-1×L287, HKI 323×L287, HKI 4887×L287, HKI 1042×193-1, HKI 1670×193-1, HKI 1041-2×193-1, HKI 1042×193-2, HKI 1659×193-2, HKI 1662×193-2, HKI 1664×193-2, HKI 1670×193-2, HKI 295×323, HKI 327T×488, HKI 1040-7×1105, PC-3ER-3×PC SONA, PC-7ER-5×PC SONA, PCMYD-8×PC SONA and PALL×PC SONA showed moderately resistant reaction (33.34-55.55 % disease severity) to *R. solani*. None of the maize hybrids showed moderately susceptible to susceptible reaction even under artificial inoculation conditions. Out of sixty inbred lines of maize one inbred line *i.e.* HKI

1663 LM-6 was not germinated. However, only two inbred lines *viz.*, HKI 1128 and HKI 1345 W of maize showed resistant reaction (≤ 33.33 % disease severity), eight inbred lines namely HKI 164-7-4, HKI 193-1, HKI 193-2, HKI 288-2, HKI 488, MBR-139, HKI 1378 W and PC-1ER-4 showed moderately resistant reaction (33.34-55.55 % disease severity) against this pathogen under artificial inoculation conditions. Rest of the inbred lines of maize showed moderately susceptible to susceptible reaction. Several workers had also screened many inbred lines/hybrids for their relative resistance to banded leaf and sheath blight of maize (Vimala *et al.*, 1998; Sharma *et al.*, 2003; Meena, 2004; Biswas *et al.*, 2007; Madhavi *et al.*, 2012; and Malik *et al.*, 2015). Among various inbred lines evaluated CM-104, P-217407, CM-103, CM-105, CM-300, and hybrid VL-43 were reported resistant under *in vivo* and *in vitro* conditions (Ahuja and Payak, 1984). In Korea, Lee *et al.*, (1989) revealed from his screening experiment that P 3055, Suweon 89, Jinjuok, P 3160, Suweon 83, DK 689, Suweon 87 and XCG were highly tolerant to this pathogen. Malik *et al.*, (2015) revealed that four inbred lines namely HKI - 488WG, HKI- C-141, HKI - 1040-5 and HKI - 1347-4LT were found resistant against banded leaf and sheath blight disease incited by *R. solani*. There is need to test the promising germplasm against different isolates of *R. solani* pathogen. There is unavailability of resistant source against this pathogen, so to overcome this problem the hybrids and inbred lines identified as source of resistance can be utilized for multilocational trials for screening to find out stable source of resistance.

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Table 1: BLSB Rating Scale (1-9)

Rating scale	Degree of infection (per cent DLA)	PDI	Disease reaction
1.0	Disease on one leaf sheath only; few small non-coalescent lesions present ($\leq 10\%$).	\leq 11.11	Resistant (R) (Score: ≤ 3.0) (PDI: ≤ 33.33)
2.0	Disease on two sheaths; lesions large and coalescent (10.1-20%).	22.22	
3.0	Disease up to four sheaths; lesions many and always coalescent (20.1-30%).	33.33	
4.0	As in disease rating symptoms of 3.0, + rind discoloured with small lesions (30.1-40%).	44.44	Moderately resistant (MR) (Score: 3.1-5.0) (PDI: 33.34-
5.0	Disease on all sheaths except two internodes below	55.55	

	the ear (40.1-50%).		55.55)
6.0	Disease upto one internode below ear shoot; rind discoloration on many internodes with large depressed lesions (50.1-60%).	66.66	Moderately susceptible (MS) (Score: 5.1-7.0)
7.0	Disease up to the internodes bearing the ear shoot but shank not affected (60.1-70%)	77.77	(PDI: 55.56-77.77)
8.0	Disease on the ear; husk leaves show bleaching, bands and cracking among themselves as also silk fibers; abundant fungal growth between and on kernels; kernels formation normal except being luster less; ear size less than normal; some plants prematurely dead (70.1-80%).	88.88	Susceptible (S) (Score: > 7.0) (PDI:
9.0	In addition to disease rating symptoms of 8.0, shrinkage of stalk; reduced ear dimensions; wet rot and disorganization of ear; kernel formation absent or rudimentary; prematurely dead plants common; abundant sclerotia production on husk leaves, kernels ear tips and silk fibers.	99.99	>77.77)

Table 2: Evaluation of hybrids for disease incidence (%) and disease severity (%)

Sr. No.	Hybrid	Disease incidence (%)	Disease severity (%)
		Mean	Mean
1.	HM-8	61.11 (51.47)	25.56 (30.15)
2.	HM-11	66.67 (55.43)	50.90 (45.49)
3.	HM-13	90.00 (76.71)	42.05 (40.36)
4.	HKI 193-1×L287	83.33 (72.36)	33.55 (35.19)
5	HKI 295×L287	8.33 (12.04)	4.50 (8.72)
6.	HKI 327T×L287	74.99 (60.29)	20.32 (26.76)
7.	HKI 323×L287	89.44 (71.01)	35.52 (36.24)
8.	HKI 4887×L287	62.72 (52.89)	34.57 (35.79)
9.	HKI 1041-2×L287	80.00	30.27

		(70.37)	(33.15)
10.	HKI 1042×193-1	100.00 (90.00)	35.60 (36.52)
11.	HKI 1670×193-1	91.67 (77.94)	31.38 (34.05)
12.	HKI 1041-2×193-1	100.00 (90.00)	38.60 (38.09)
13.	HKI 1042×193-2	79.16 (62.93)	34.97 (35.76)
14.	HKI 1659×193-2	77.78 (69.08)	48.52 (44.13)
15.	HKI 1662×193-2	70.13 (57.67)	37.00 (37.13)
16.	HKI 1663×193-2	100.00 (90.00)	25.52 (30.12)
17.	HKI 1664×193-2	78.57 (69.54)	27.65 (31.62)
18.	HKI 1670×193-2	70.91	31.15

		(57.74)	(33.67)
19.	HKI 295× 323	79.16 (62.93)	39.70 (38.64)
20.	HKI 327T×488	75.00 (67.49)	35.47 (35.92)
21.	HKI 1040-7×1105	74.02 (60.75)	38.25 (37.97)
22.	PC-3ER-3×PC SONA	80.35 (63.87)	50.32 (45.17)
23.	PC-7ER-5×PC SONA	80.35 (63.87)	51.29 (45.72)
24.	PC-8ER-2×PC SONA	83.33 (72.36)	29.65 (32.43)
25.	PCMYD-7×PC SONA	44.31 (41.44)	31.00 (33.45)
26.	PCMYD-8×PC SONA	66.47 (55.82)	40.52 (38.85)
27.	PALL×PC SONA	91.67	53.65

		(77.94)	(47.55)
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*Figures in parenthesis indicate angular transformed values

Table 3: Screening of maize hybrids against banded leaf and sheath blight

Disease scale	Reaction	No. of genotypes	Entries
≤ 3.0	Resistant (R)	7	HM-8, HKI 295×L287, HKI 327T×L287, HKI 1041-2×L287, HKI 1663×193-2, PC-8ER-2×PC SONA and PCMYD-7×PC SONA
3.1-5.0	Moderately Resistant (MR)	20	HM-11, HM-13, HKI 193-1×L287, HKI 323×L287, HKI 4887×L287, HKI 1042×193-1, HKI 1670×193-1, HKI 1041-2×193-1, HKI 1042×193-2, HKI 1659×193-2, HKI 1662×193-2, HKI 1664×193-2, HKI 1670×193-2, HKI 295×323, HKI 327T×488, HKI 1040-7×1105, PC-3ER-3×PC SONA, PC-7ER-5×PC SONA, PCMYD-8×PC SONA and PALL×PC SONA
5.1-7.0	Moderately susceptible (MS)	0	0
>7.0	Susceptible	0	0

	(S)		
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Table 4: Evaluation of inbred lines for disease incidence (%) and disease severity (%)

Sr. No.	Inbred lines	Disease incidence (%)	Disease severity (%)
		Mean	Mean
1.	HKI 194-6	87.50 (74.99)	59.35 (50.94)
2.	HKI 139	100.00 (90.00)	84.65 (67.29)
3.	HKI 3-4-7	100.00 (90.00)	93.25 (74.93)
4.	HKI GD-16	100.00 (90.00)	95.40 (77.73)
5	HKI 164-7-2	80.00 (70.37)	68.80 (57.36)
6.	HKI 164 D-3-3	95.45	57.00

		(81.21)	(49.46)
7.	HKI 164 3-3-2	85.00 (73.38)	72.05 (59.34)
8.	HKI 164-7-4	53.02 (46.86)	34.15 (35.65)
9.	HKI 190	100.00 (90.00)	91.52 (73.07)
10.	HKI 191-2-6	100.00 (90.00)	77.52 (63.16)
11.	HKI 193-1	87.50 (74.98)	45.45 (42.36)
12.	HKI 193-2	83.33 (72.35)	50.00 (44.98)
13.	HKI 194-7	80.35 (63.87)	72.00 (58.03)
14.	HKI 288-2	84.52 (66.82)	40.50 (39.09)
15.	HKI 323	95.45	66.45

		(81.21)	(55.22)
16.	HKI 327T	87.50 (74.98)	93.45 (75.18)
17.	HKI 327D	100.00 (90.00)	74.50 (60.94)
18.	HKI 488	100.00 (90.00)	41.57 (40.13)
19.	HKI 488T	100.00 (90.00)	85.52 (67.72)
20.	HKI 577	87.50 (74.98)	66.68 (56.45)
21.	HKI 536C	87.50 (74.98)	93.57 (75.32)
22.	HKI 766RG	100.00 (90.00)	94.65 (76.64)
23.	HKI 788	68.33 (55.74)	83.27 (65.84)
24.	HKI 1011	100.00	75.03

		(90.00)	(60.02)
25.	HKI 1015 WG-8	88.46 (75.63)	56.55 (48.77)
26.	HKI 1015-6	87.50 (74.98)	86.68 (69.51)
27.	HKI 1035 RG	100.00 (90.00)	57.50 (49.51)
28.	HKI 1035-11	100.00 (90.00)	81.85 (65.37)
29.	HKI 1040-4	100.00 (90.00)	93.37 (75.10)
30.	HKI 1105	100.00 (90.00)	93.50 (75.24)
31.	HKI 1105-6	100.00 (90.00)	59.00 (52.19)
32.	HKI 1105-2ML(O)	68.75 (63.87)	74.72 (62.09)
33.	HKI 1155-1(1+2)	100.00	67.69

		(90.00)	(57.49)
34.	HKI 1128	100.00	30.55
		(90.00)	(33.35)
35.	HKI 1332	100.00	94.25
		(90.00)	(76.25)
36.	HKI 1616-4	75.00	67.68
		(67.49)	(57.02)
37.	HKI 1042 NP-79 ER-1	100.00	84.37
		(90.00)	(66.72)
38.	HKI 1043 NP-84 ER-1	60.00	63.92
		(58.27)	(54.06)
39.	HKI 1651 14-3-1 Win K-11	88.31	59.51
		(70.09)	(50.52)
40.	HKI 1657 5ER-4 Win K-14	90.00	74.88
		(76.71)	(59.93)
41.	HKI 1654 15ER-1-2 Win K-12	100.00	63.50
		(90.00)	(55.57)
42.	HKI 1659 40126 ER-4	100.00	95.46

		(90.00)	(77.73)
43.	HKI 1663 LM-6	NG**	NG**
44.	MBR-139	100.00 (90.00)	37.97 (37.98)
45.	C-141	87.50 (74.98)	72.45 (60.21)
46.	HKI 1078-4	46.97 (43.09)	57.08 (50.72)
47.	HKI 1342 W	91.66 (77.94)	65.46 (56.73)
48.	HKI 1344 ER-4 W	65.00 (54.19)	79.12 (64.36)
49.	HKI 1345 W	83.33 (65.87)	32.65 (34.59)
50.	HKI 1348 W	100.00 (90.00)	93.57 (75.32)
51.	HKI 1348-8-2 W	96.15 (81.93)	91.15 (72.67)

52.	HKI 1352 W	91.66 (77.94)	65.57 (55.48)
53.	HKI 1378 W	90.91 (77.36)	37.50 (37.74)
54.	PC-1ER-4	85.00 (73.38)	39.19 (38.73)
55.	PC-3ER-2	100.00 (90.00)	60.80 (51.70)
56.	PC-4B	100.00 (90.00)	90.50 (72.02)
57.	PC-8	100.00 (90.00)	78.85 (63.27)
58.	PC-8-3	95.45 (81.21)	93.80 (75.59)
59.	PC-1473-5	94.45 (80.25)	86.52 (68.65)
60.	PC 1558-4	100.00 (90.00)	93.45 (75.20)

** Not Germinated

*Figures in parenthesis indicate angular transformed values

Table 5: Screening of maize inbred lines against banded leaf and sheath blight

Disease scale	Reaction	No. of genotypes	Entries
≤ 3.0	Resistant (R)	2	HKI 1128 and HKI 1345 W
3.1-5.0	Moderately Resistant (MR)	8	HKI 164-7-4, HKI 193-1, HKI 193-2, HKI 288-2, HKI 488, MBR-139, HKI 1378 W and PC-1ER-4
5.1-7.0	Moderately susceptible (MS)	25	HKI 194-6, HKI 164-7-2, HKI 164 D-3-3, HKI 164 3-3-2, HKI 191-2-6, HKI 194-7, HKI 323, HKI 327D, HKI 577, HKI 1099, HKI 1015 WG-8, HKI 1035 RG, HKI 1105-6, HKI 1105-2ML(O), HKI 1155-1(1+2), HKI 1616-4, HKI 1043 NP-84 ER-1, HKI 1651 14-3-1 Win K-11, HKI 1657 5ER-4 Win K-14, HKI 1654 15ER-1-2 Wink-12, C-141, HKI 1078-4, HKI 1342 W, HKI 1352 W and PC-3ER-2
>7.0	Susceptible (S)	24	HKI 139, HKI 3-4-7, GD-16, HKI 190, HKI 327T, HKI 488T, HKI 536C, HKI 766RG, HKI 788, HKI 1015-6, HKI 1035-11, HKI 1040-4, HKI 1105, HKI 1332, HKI 1042 NP-79 ER-1, HKI 1659 40126 ER-4, HKI 1344 ER-4 W, HKI 1348 W, HKI 1348-8-2 W,

			PC-4B, PC-8, PC-8-3, PC-1473-5 and PC 1558-4
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UNDER PEER REVIEW