Inheritance of foliar blast resistance in pearl millet (*Pennisetum glaucum*)

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*Vol 131, Issue 1, Pages 217-219, Year February 2012*


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**Institution:**


**Title:**

Inheritance of foliar blast resistance in pearl millet (*Pennisetum glaucum* L. (R.) Br.)

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Abstract

Foliar blast, caused by *Pyricularia grisea* (Cooke) Sacc, has recently emerged as a serious disease of pearl millet in India. To study the inheritance of resistance to this disease, two resistant restorer lines (ICMR 06222 and ICMR 07555) and two susceptible maintainer lines (ICMB 95444 and ICMB 89111) were selected on the basis of foliar blast reaction in tests conducted under field and greenhouse conditions. Each of the two resistant parents was crossed with two susceptible parents to generate 4 sets of $F_1$s, $F_2$s and their backcrosses with both resistant and susceptible parental lines. These were evaluated for disease reaction with artificial inoculation under both field and greenhouse conditions. The disease reaction of the $F_1$s, and the segregation patterns of resistance in the $F_2$s and backcross generations, showed that resistance to foliar blast in pearl millet is controlled by a single dominant gene.

**Key words:** *Pennisetum glaucum* - foliar blast - leaf spot - inheritance
Pearl millet foliar blast, also known as leaf spot caused by *Pyricularia grisea* (Cooke) Sacc. [teleomorph: *Magnaporthe grisea* (Herbert) Barr] was first reported in 1942 from Kanpur, Uttar Pradesh in India (Mehta *et al.* 1953). However, until recently, it had not been a disease of any economic significance in this country, which annually cultivates it on about 9.5 million ha, and hence has the largest pearl millet area in the world. Leaf blast has been considered a serious disease in southern coastal plains of the USA where infection from this disease has been found to have significant adverse effects on green forage yield and digestible dry matter (Wilson and Gates, 1993). It is known that host plant resistance is the most cost-effective strategy to effectively manage this disease. Thus, sources of blast resistance were identified, and efforts were made to incorporate resistance into improved hybrid parents and elite breeding lines in the USA (Hanna *et al.* 1988). Recently, leaf blast has emerged as a serious disease in pearl millet in India (Lukose *et al.* 2007; Anonymous, 2009), which becomes more severe during humid weather conditions, especially in dense plant stands. Breeding for blast resistance is yet to begin in India, though field and greenhouse screening techniques have been developed and resistance sources have been identified (Thakur *et al.* 2009). Knowledge of the inheritance of resistance will have a direct bearing on the breeding efficiency for genetic management of this disease. We report on the results of a study of the inheritance of blast resistance to the pathogen population prevalent at ICRISAT, Patancheru research center.

**Materials and Methods**
Based on the results of a previous study (Thakur et al. 2009), ICMR 06222 and ICMR 07555 were selected as resistant parents, and ICMB 89111 and ICMB 95444 as susceptible parents for foliar blast disease. These selected parental lines were re-confirmed for their foliar blast reaction in the greenhouse at ICRISAT, Patancheru. Four F₁s were generated by crossing both resistant lines (P₂) on each of the two susceptible lines (P₁) in the cool post-rainy season during November-February 2008-09. During the subsequent hot dry season, March-June 2009, in each F₁, 8-10 panicles were selfed using parchment paper bags to generate a F₂ population, and bulk pollen from 8-10 F₁ panicles was used to pollinate the corresponding susceptible and resistant parents to develop BCP₁ (susceptible parent × F₁) and BCP₂ (resistant parent × F₁) populations, respectively.

All the parents, four F₁s, four F₂s, four BCP₁s and four BCP₂s were screened against P. grisea Patancheru isolate in the greenhouse in July-August 2009 in three replications. In each replication, 3 pots of the parents and F₁, 10 pots each of both BCP₁ and BCP₂, and 20 pots of F₂ were planted for each cross. Seeds were sown in 15-cm diameter pots (10 seeds/pot) filled with sterilized soil-sand-FYM mix (2:1:1) and placed in a greenhouse bay maintained at 30±1°C. The seedlings (12 day-old) were spray-inoculated with an aqueous conidial suspension (ca. 1×10⁵ spores ml⁻¹) of P. grisea (Patancheru isolate) and exposed to high humidity (>90% RH) under misting for 10 days. Blast severity was recorded 10 days after inoculation using a 1-9 progressive scale (Thakur et al. 2009). Following
this, the plants having score of $\leq 3$ were rated as resistant and with score of $>5$ as susceptible.

The above parents and populations were also evaluated under field conditions during rainy season of 2009. The experiment was conducted in a randomized complete block design with 3 replications with 1 row of 4m long for each F$_1$ and parents, 4 rows of each BCP$_1$ and BCP$_2$, and 8 rows of each F$_2$ planted in each replication. Systematic susceptible checks (ICMB 95444, -99666 and -89111) were grown every 5$^{th}$ row, alternately. Plants were thinned to 20 plants/row 15 days after planting and standard agronomic practices were followed for crop management. Plants were spray-inoculated twice, first at pre-tillering stage and second at flowering stage with an aqueous conidial suspension (ca. $1 \times 10^5$ spores mL$^{-1}$) of $P.$ grisea (Patancheru isolate) High humidity was provided by perfo-irrigation twice a day on rain-free days, 30 min each between 11-12 h and 16-17 h, to promote disease development. Disease severity was recorded using same 1-9 progressive scale as mentioned for greenhouse screening.

The observed ratios of resistant to susceptible plants in the segregating populations in greenhouse and field experiments were compared to theoretical ratios using chi-square test after pooling of plants from all the replications.

**Results and Discussion**

All the plants of the susceptible parents were susceptible (score of $>5$) both under greenhouse and field conditions. In the F$_2$ and BCP$_1$ there was clear cut segregation either for resistant plants (score of $\leq 3$) or susceptible plants
(score of >5), and no plant had score of 4 and 5 for blast reaction both under greenhouse and field conditions. All the plants of the two resistant parents were resistant both under greenhouse and field conditions. All plants in all the four F₁s and their corresponding four BC₂₁s were also resistant to blast under greenhouse and field conditions (Table 1). The F₂ population from cross ICMB 89111 x ICMR 06222 had a good fit to the segregation ratio of 3R:1S in both the greenhouse and field screens, indicating dominant monogenic control of blast resistance. The BC₁ of this cross had good fit to the 1R:1S ratio expected for monogenic inheritance in both greenhouse and field screens. The F₂ of cross ICMB 95444 x ICMR 06222 also gave good fit to the segregation ratio of 3R:1S in both greenhouse and field screens, and BC₁ segregation of this cross had good fit to 1R:1S segregation ratio in field screen but not in the greenhouse where excess of susceptible plants were observed. The resistant parent ICMR 07555 when crossed to the susceptible parents ICMB 89111 and ICMB 95444 gave a good fit to segregation ratio of 3R:1S in the F₂ both in greenhouse and field screens, again indicating monogenic control of blast resistance. The BC₁ ratio of these crosses had significant deviations from the expected 1R:1S segregation ratio due to excess of susceptible plants in both the greenhouse and field experiments. Thus, in all the five cases of BC₁ where segregation ratio had significant deviation from the expected 1R:1S ratio, it was due to excess of susceptible plants, which most likely could have resulted from some selfing in the susceptible parents that were used as female parents in deriving the BC₁ generation. Such
deviation from expected ratio could also result from segregation distortion caused by segregation distortion loci identified in pearl millet (Busso et al. 1995), although segregation distortion appears less likely cause of the deviation from expected ratios which almost all were found in BCP1 and not in the F2 generation of all crosses.

The goodness of fit to 3R:1S segregation ratio in all the four F2s, and 1R:1S ratio in 3 out of the 8 BCP1 populations under both greenhouse and field conditions leads us to conclude that foliar blast resistance in the pearl millet lines used for this study is controlled by a single dominant gene. In an earlier study, three independent dominant genes were reported to control blast resistance in which Tifton PS34, a weedy relative of pearl millet Pennisetum glaucum ssp monodii, was used as resistant source and evaluated against a pathogen population from Georgia, USA (Hanna and Wells, 1989). In yet other study involving Tift 85DB, a blast resistant inbred line derived by backcrossing Tifton PS34 to cultivated pearl millet, resistance to blast was reported to be under dominant monogenic control (Wilson et al. 1989). Thus only one of the three resistant genes from Tifton PS34 got introgressed into Tift 85DB during backcrossing program, and it was as effective for resistance as the three genes. However, Tift 85DB was found to be susceptible to the Patancheru isolate used in our study, indicating that the pathotype used in our study is different from the one used in the above study. We also observed that all the 150 plants of a F2 population derived from cross ICMR 06222 x ICMR 0755 when tested for blast reaction in the greenhouse were resistant to the disease, indicating that
both parents carried the same common gene for resistance. It is significant
to note that the resistant parents used in this study are of very diverse
origin: ICMR 06222 (SDMV 90031-S1-3-3-2-1-3-2-2-1-1-B) is derived
from an *iniari* landrace-based open-pollinated variety developed by
ICRISAT in southern Africa, and ICMR 07555 (ICMS 8511 S1-17-2-1-1-
4-1-B-3-3-2-2-B) is derived from a non-*iniari*-based synthetic developed at
ICRISAT in India. A Blast Resistant Seed Parent Composite has been
constituted from the intercrosses of 8 blast resistant seed parental lines of
diverse origin developed at ICRISAT. About 500 plants of this composite
were evaluated during the 2009 rainy season under field condition using
artificial inoculation. Interestingly, all the plants were found resistant to
moderately resistant with no segregation for susceptible plants, indicating
that all lines involved in this composite carried a common resistance gene.
Considering the severity and wider occurrence of this disease in India,
extensive efforts should be made to identify additional sources of resistance
to the pathotype used in our study as well as to other more virulent
pathotypes recently identified and being studied for virulence diversity
(Rajan Sharma unpublished)

*M. grisea* infecting rice had shown large pathogenic variability. Thus, a
preliminary assessment of the pathogenic variability for virulence was
conducted in pearl millet using 20 isolates from different locations in India.
The most resistant line ICMR 06222 used in this study was found
susceptible to four isolates, indicating pathogenic variability in the
pathogen, and suggesting the use of different pathotypes for the
identification of resistance sources. In rice, about 50 blast resistance genes have been identified and several of them have been incorporated into rice cultivars. However, most of these resistance genes have broken down to blast disease because of their race specificity and also due to the rapid changes in pathogenicity of the blast fungus (Suh et al. 2009). Various potential mechanisms, including sexual recombination, heterokaryosis, parasexual recombination and aneuoploidy have been proposed to explain frequent race changes in the rice blast fungus (Kang and Lee 2000). Therefore, efforts should be made to study pathogenic variability in P. grisea isolates from different pearl millet growing areas in India and identify resistant sources to different pathotypes for utilizing them in breeding program to manage this disease through host plant resistance.

References

Anonymous 2009: Annual Report, All India Coordinated Pearl Millet Improvement, Indian Council of Agricultural Research. Project Coordinating Unit, Agricultural research Unit, Mandore, Jodhpur-342304, India.


Hanna, W.W., H.D. Wells, G.W. Burton, and W.G. Monson, 1988:
Kang, S., and Y.H. Lee, 2000: Population structure and race variation of
Lukose, C.M., D.L. Kadvani, and C.J. Dangaria, 2007: Efficacy of
fungicides in controlling blast disease of pearl millet. Indian
Phytopathology 60, 68-71.
Mehta, P.R., B. Singh, and S.C. Mathur, 1953: A new leaf spot disease of
bajra (Pennisetum typhoides Staph and Hubbard) caused by a species of
Piricularia. Indian Phytopathology 5, 140-143.
Suh, J.P., J.H. Roh, Y.C. Cho, S.S. Han, Y.G. Kim, and K.K. Jena, 2009:
The Pi40 gene for durable resistance to rice blast and molecular
analysis of Pi40-advanced backcross breeding lines. Phytopathology
99, 243-250.
Thakur, R.P., Rajan Sharma, K.N. Rai, S.K. Gupta, and V.P. Rao, 2009:
Screening techniques and resistance sources for foliar blast in pearl
Wilson, J.P. and R.N. Gates, 1993: Forage Yield losses in hybrid pearl
millet due to leaf blight caused primarily by Pyricularia grisea.
Phytopathology 83, 739-743.
to Pyricularia grisea in pearl millet accessions from Burkino Faso and
inbred Tift 85DB. Journal of Heredity 80, 499-501.
Table 1. Segregation for blast resistant (R) and susceptible (S) plants in F1, F2, BCP1, and BCP2 generations and test of goodness of fit for hypothetical Mendelian ratios in four crosses of two susceptible parents with the two resistant parents in pearl millet, in greenhouse and field experiment, rainy season 2009, ICRISAT–Patancheru.

<table>
<thead>
<tr>
<th>Cross</th>
<th>Environment</th>
<th>Generation</th>
<th>No. of plants observed</th>
<th>Expected Ratio</th>
<th>No. of expected plants</th>
<th>χ²</th>
<th>P</th>
</tr>
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<td>ICMB 89111 x ICMR 06222</td>
<td>Greenhouse</td>
<td>F1</td>
<td>145 0</td>
<td>- -</td>
<td>R S</td>
<td>-</td>
<td>-</td>
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<tr>
<td></td>
<td></td>
<td>F2</td>
<td>338 107</td>
<td>3 1</td>
<td>334 111</td>
<td>0.21</td>
<td>0.64</td>
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<td></td>
<td></td>
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<tr>
<td></td>
<td>Field</td>
<td>F1</td>
<td>52 0</td>
<td>- -</td>
<td>- -</td>
<td>-</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td></td>
<td>F2</td>
<td>494 149</td>
<td>3 1</td>
<td>482 161</td>
<td>1.14</td>
<td>0.28</td>
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<td></td>
<td></td>
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<td>206 201</td>
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<td>203.5 203.5</td>
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<td></td>
<td></td>
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<td>F1</td>
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<td>-</td>
<td>-</td>
</tr>
<tr>
<td></td>
<td></td>
<td>F2</td>
<td>561 189</td>
<td>3 1</td>
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<td></td>
<td>BCP1</td>
<td>109 156</td>
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<td>132.5 132.5</td>
<td>8.33</td>
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</tr>
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<td>-</td>
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<td>Field</td>
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<td>- -</td>
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<tr>
<td></td>
<td></td>
<td>F2</td>
<td>544 164</td>
<td>3 1</td>
<td>531 177</td>
<td>1.27</td>
<td>0.26</td>
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<td></td>
<td>BCP1</td>
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<td>1 1</td>
<td>98 98</td>
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<td>0.25</td>
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<tr>
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<td>BCP2</td>
<td>180</td>
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<tr>
<td>ICMB 89111 x ICMR 07555</td>
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<td>F1</td>
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<td>-</td>
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<td></td>
<td></td>
<td>F2</td>
<td>396 161</td>
<td>3 1</td>
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<td></td>
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<td>F2</td>
<td>570 165</td>
<td>3 1</td>
<td>551 184</td>
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<td>F1</td>
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