New sources identified for resistance to brown spot disease in rice

Brown spot, caused by Bipolaris oryzae (Shoem.), is a widespread disease in all rice-growing areas of the world. However, only limited information is available on resistance sources and their genetics. The fungus can infect rice plant at all stages of growth—from germination to maturity. It causes seedling blight, reduces photosynthetic rate at the vegetative phase, poor grain filling, and grain abortion at the reproductive phase, and ultimately brings about considerable yield loss both in quantity and quality. This disease is more prevalent in rained lowlands and uplands or in rice areas with poor soil condition. Rice plants get infected by externally and internally seedborne infection, spore inoculum on crop residues and soils, as well as airborne spore inoculum. In the study, we developed a screening protocol for brown spot development and assessment at the seedling stage, identified sources of resistance, and produced mapping populations for detecting linkage with molecular markers.

Pathogen

More than 40 isolates of the fungus from leaves collected from farmers’ fields in Laguna, Batangas, and Cagayan in the Philippines were used to study the colony and conidial characteristics and some differences were observed. Eighteen isolates from 22 isolates were used to identify the sources of resistance.

Host

A diverse germplasm of rice—comprising 22 modern high-yielding varieties, 6 traditional wild species representing 9 genomes, 11 introduction lines, 10 hybrids, 5 CMS lines, and 10 restorers—was evaluated with a newly developed screening protocol at seedling stage. Significant differences were obtained between cultivars/lines for individual isolates based on lesion type and lesion density after seedling inoculation.

Host-pathogen interaction

The 22 isolates were tested on six cultivars identified as resistant, susceptible, and intermediate. They showed significant cultural-isolate interaction, and susceptibility of isolates differed significantly on highly susceptible cultivar IR64 and medium-resistant cultivar Dinorado. Traditional cultivar Dinorado was found to be highly resistant (lesion type ratio = 0.25) to all 22 isolates. Besides Dinorado, wild species (Oryza minuta) was also found to be resistant to all 22 isolates tested.

Protocol for screening

- Multiplication of mononastic isolates on freshly prepared PDA with pH around 8 in culture growth room under NUV and fluorescent light with temperature 28 ± 2 °C in the daytime and 12 h photosperiod.
- Inoculation of 14 to 15-d-old seedlings (from sowing) with spore concentration around 10⁴ at 0.1 ml seedling⁻¹ prepared from 10-d-old culture plate. Spores were washed two times by centrifugation with sterile water before estimation of final concentration. Incubation of control was with sterile water.
- Incubation of inoculated seedlings in humid chamber in darkness for 40 h at room temperature 18±2 °C.
- Incubation of seedlings for disease development in seedling growth room for 7 d at temperature 28±4 °C, humidity above 80%, and 12-h photosperiod.
- Disease assessment on 2nd leaf from the top as number and size of lesion cm² (small = pin head, medium = up to 1 mm linear length, and large = above 1 mm linear length), which were converted into lesion type ratio (LTR) as large lesion cm² / (small lesion cm² + medium lesion cm²). Resistance = LTR < 0.25; Intermediate = LTR 0.25-0.50; Susceptible = LTR > 0.50.

Genetic analysis

F₂ progenies were produced from crosses between Dinorado (resistant) and IR36 (susceptible). Two hundred F₂ lines were screened using aggressive isolate SM2 to determine the inheritance of resistance. Of the 150 SSR markers tested, 10 revealed polymorphism between resistant and susceptible bulk comparable with that of the parents. Further analysis is in progress to tag the gene governing resistance to brown spot.

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S.P. Banu, B. Miah,* A. Ali,* D. Brar,* and C.M. Vera Cruz*

Bangladesh Agricultural Research Institute, Gazipur, Bangladesh

International Rice Research Institute, Philippines