

Diseases Caused by Fungi and Fungus-Like Organisms

First Report of *Fusarium proliferatum* Causing Sheath Rot Disease of Rice in Eastern India

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Sheath rot is one of the most devastating diseases of rice because of its ability to reduce the yield significantly in all rice cultivating areas of the world (Bigirimana et al. 2015). Sheath rot disease is associated with various pathogens such as *Sarocladium oryzae*, *Fusarium fujikuroi* complex, and *Pseudomonas fuscovaginae* (Bigirimana et al. 2015). Hence, this disease has become more complex in nature and added more seriousness. From September to December 2018, plants were observed with typical sheath rot symptoms in a research farm of ICAR – National Rice Research Institute and 10 farmer's fields of Cuttack district, Odisha, Eastern India. About 25 to 37% of sheath rot disease severity was recorded in the infected field. Diseased plants were observed with symptoms such as brownish or reddish-brown irregular lesions, which later became enlarged with grayish centers. Further, rotting of the topmost leaf sheaths that surround the young panicle was observed. At the severe stages, the young panicle was partially emerged from the sheath or completely rotted within the sheath. The white to pinkish powdery growth observed inside the infected sheath led to chaffy and discolored grains. The sheath rot symptomatic plants were collected from the infected fields. To isolate the causal pathogen, infected sheath tissues were surface sterilized in 1% sodium hypochlorite for 2 min, rinsed three times in sterile distilled water, and placed on potato dextrose agar medium (PDA) (HiMedia). Plates were incubated at 27 ± 1°C for 3 days. Further, fungal pathogen colonies were subcultured and purified to perform the pathogenicity test. On PDA, the colonies produced abundant white

aerial mycelium with violet to pink pigmentation, and hyphae were hyaline with septation. Abundant single-celled, oval-shaped microconidia (5.5 to 9 × 1.5 to 2 µm) were produced, whereas macroconidia were not produced, and the fungal pathogen was tentatively identified as *Fusarium* sp. In order to characterize the pathogen at a molecular level, ITS, alpha elongation factor gene (EF1-α), RNA polymerase II largest-subunit gene (RPB2), and calmodulin gene (cld) were amplified using the primer pair ITS1/ITS4, EF1/EF2, 5F/7CR, and CLPRO1/CLPRO2, respectively, and PCR amplicons were subjected to sequencing (Chang et al. 2015; O'Donnell et al. 1998; White et al. 1990). Furthermore, a species-specific primer, Fp3-F/Fp4-R, was used to identify the pathogen (Jurado et al. 2006). The resulting sequences were confirmed by BLAST analysis and the *FUSARIUM*-ID database (<http://isolate.fusariumdb.org/blast.php>). BLASTn search showed 100% similarity between the query sequence and ITS, EF1-α, RPB2, and calmodulin gene sequences of *Fusarium proliferatum* available in GenBank. The following GenBank accession numbers were obtained: MT394055 for ITS, MT439867 for EF1-α, MT790774 for calmodulin, MT940224 for RPB2, and MT801050 for species-specific to *F. proliferatum*. To confirm the pathogenicity under glass house conditions, fungus grown on sterilized chaffy grains was placed in between boot leaf sheath and panicle and covered with moist cotton (Saravanakumar et al. 2009). After 15 days postinoculation, rotting symptoms were observed, and these were similar to field symptoms. Pathogen was constantly reisolated from symptomatic tissue, satisfying Koch's postulates. Disease symptoms were not observed on uninoculated plants. Morphological characters, pathogenicity testing, and molecular characterization have identified the pathogen as *F. proliferatum*. To the best of our knowledge, this is the first confirmed report of *F. proliferatum* causing sheath rot disease on rice from Eastern India.

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