Diseases Caused by Bacteria and Phytoplasmas

Confirmation Report of Bacterial Leaf Streak Disease of Rice Caused by *Xanthomonas oryzae* pv. *oryzicola* in Senegal

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Xanthomonas oryzae pv. oryzicola (Xoc), the causal agent of bacterial leaf streak (BLS), is considered one of the most important emerging pathogens of rice in Africa. This disease is estimated to be responsible for 20 to 30% yield loss (Sileshi and Gebeyehu 2021) and has been characterized in several west African countries, including Mali and Burkina Faso since 2003 and more recently in the Ivory Coast (Diallo et al. 2021; Wonni et al. 2014). The presence of BLS symptoms in Senegal was reported by Trinh (1980) but, to our knowledge, BLS occurrence has never been validated further and no strain of Xoc has ever been isolated from Senegalese rice fields. Xoc is transmitted by seeds, which contributes to its spread through the rice trade (Sileshi and Gebeyehu 2021). To confirm Trinh's observations, we surveyed rice fields between 2014 and 2016 in eight different regions where rice is produced in Senegal. Typical disease symptoms characterized by yellow-brown to black translucent leaf streaks, sometimes along with exudates, were detected in fields of several regions and collected. Leaf pieces were successively sanitized in 1% sodium hypochlorite and 70% ethanol, rinsed in sterile water, and symptomatic fragments were ground using the Qiagen Tissue Lyser System (QIAGEN, Courtaboeuf, France). The leaf powder was diluted in 1.5 ml of sterile water and incubated for 30 min at room temperature. Ten microliters of the suspension was streaked on semiselective PSA medium (peptone 10 g, sucrose 10 g, glutamic acid 1 g, and agar 16 g per liter) and incubated at 28°C for 3 to 7 days. Characteristic round, convex, mucous, straw-yellow Xoc candidate colonies were purified from six individual leaf samples from two distinct sites in Ndiaye and one in Fanaye in the region of Saint Louis. To confirm their identity, isolated strains were tested for pathogenicity and molecular characterization. All isolates were subjected to the multiplex PCR developed for the identification of X. oryzae pathovars (Lang et al. 2010) and revealed the same PCR profile (two amplicons of 324 and 691 base pairs) similar to that of the Xoc reference strain BLS256. Leaves of 5-week-old plants of Oryza sativa cv. Kitaake were infiltrated with a needleless syringe containing a bacterial suspension adjusted to 10⁸ CFU/ml. After 7 days of incubation under greenhouse conditions (27 \pm 1°C with a 12-h photoperiod), all infiltrated spots (two spots on three plants per isolate) developed water-soaked lesions similar to those caused by control strain BLS256, except when leaves were infiltrated with water. Symptomatic leaf tissues were ground and plated on PSA medium, producing colonies with typical Xanthomonas morphology that were diagnosed as Xoc by multiplex PCR typing, thus fulfilling Koch's postulates. Finally, four of the isolates were subjected to gyrB sequencing upon PCR amplification using the universal primers XgyrB1F and XgyrB1R (Young et al. 2008). Analysis of 780 bp partial gyrB sequences of strains S18-3-4, S23-1-12, S52-1-4, and S52-1-10 highlighted 100% identity with the gyrB sequence of strain BLS256 (acc. no. CP003057). To our knowledge, this is the first report of BLS in Senegal that is supported by molecular characterization methods. This study validates the presence of BLS in Senegal and will serve as a basis for future rice breeding efforts for locally adapted resistance. More studies are needed to clarify the spatial distribution and prevalence of BLS in Senegal as rice cultivation is expanding rapidly in the country.

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