Diseases Caused by Bacteria and Phytoplasmas

First Report of Bacterial Wilt Disease of Tomato, Pepper, and Gboma Caused by *Ralstonia solanacearum* Species Complex in Togo

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Tomato (*Solanum lycopersicum*), pepper (*Capsicum annum*), and gboma (*Solanum macrocarpon*) are major vegetables in Togo, with many people depending on these crops for their livelihood. In December 2018, during the dry season with temperatures between 21 and 35°C, tomato ('Petomech'), pepper ('Gboyebesse'), and gboma (local landrace) showing wilt symptoms without foliar yellowing were collected from two locations, Tchouloum and CECO-AGRO sites in the Sotouboua Prefecture of Togo, ~300 km from the capital city of Lomé. Disease incidence ranged between 10 and 50% in multiple fields. Cut stems of most wilting tomato, pepper, and gboma plants produced bacterial ooze in water, and vascular discoloration was visible in longitudinal stem sections. Ground cut stem tissue tested positive with *Rs* ImmunoStrips specific to the *Ralstonia solanacearum* species complex (RSSC) (Agdia, Elkhart, IN). Collected samples were stored at ambient temperature and cultured within 36 h. Culturing sap from cut stems plated on modified SMSA medium (Engelbrecht 1994) yielded colonies with typical RSSC morphology: slow-growing, irregular, mucoid, and white with red centers. Genomic DNA was extracted from 13 isolates: two from gboma, five from tomato, and six from pepper. The expected 280-bp band was amplified from all 13

genomic DNAs following polymerase chain reaction (PCR) using the 759/760 RSSC-specific primer pair (Opina et al. 1997). PCR with the 630/631 primers, which identify the race 3 biovar 2 RSSC subgroup, did not yield a product from any Togo isolate (Opina et al. 1997). The phylotype multiplex PCR identified all Togo isolates as belonging to the phylotype I subgroup, also called *R. pseudosolanacearum* (Fegan and Prior 2005; Prior et al. 2016). Phylotype control DNAs were from strains GMI1000 (phylotype I, Asia), K60 (phylotype II, Americas), CMR15 (phylotype III, Africa), and PSI07 (phylotype IV, Indonesia). Comparative genomic analysis of the partial endoglucanase (*egl*) gene, amplified with the Endo primer pairs (Poussier et al. 2000), revealed all Togo strains belonged to sequevar 17, a group known to cause bacterial wilt of peanut in China (Xu et al. 2009). The *egl* sequences are in NCBI GenBank as accessions MT572393 to MT572405. Koch's postulates were completed by inoculating 28-day-old bacterial wilt-susceptible 'Bonny Best' tomato plants by soil soak (Khokhani et al. 2018). Briefly, soil around each unwounded plant was drenched with 50 ml of a 10⁸ CFU/ml suspension of bacteria grown from a single colony. Five plants were inoculated with each of four randomly selected Togo strains. RSSC phylotype I strain GMI1000 served as a positive control and water-treated plants as negative controls. Plants were kept in a 28°C growth chamber with a 12-h photoperiod. All RSSC-inoculated plants were fully wilted within 1 week; symptoms resembled those observed in the field. Water-treated control plants did not wilt. Culturing sap from all inoculated plants on SMSA medium yielded colonies with typical RSSC morphology that tested positive with the *Rs* ImmunoStrips. This is the first identification of RSSC in Togo. These results will guide development of disease management strategies and regionally appropriate breeding of vegetable lines with resistance to the phylotype I RSSC strains present in Togo.

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