Diseases Caused by Bacteria and Phytoplasmas

First Report of *Pectobacterium aroidearum* Causing Soft Rot of Pepper (*Capsicum annuum*) Fruits in Brazil

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During field inspections carried out in August 2016, pepper (*Capsicum annuum*) plantations located in municipalities of the state of Pernambuco, Brazil, presented an incidence of 10 to 18% of pepper fruits still stuck to plants showing symptoms of soft rot. At least one pepper fruit with soft rot symptoms was collected at every municipality for identification of the bacterium. The strains were selectively isolated as described by Barroso et al. (2019). Fourteen strains were isolated, and each strain was obtained from a pepper fruit with soft rot symptoms. The strains CCRMPA106, CCRMPA109, CCRMPA131, and CCRMPA145 were obtained from Chã Grande; CCRMPA429 was obtained from Bezerros; and CCRMPA621, CCRMPA631, CCRMPA637, CCRMPA639, CCRMPA646, CCRMPA647, CCRMPA655, CCRMPA658, and CCRMPA670 were obtained from Petrolina. These strains were tested for pathogenicity in pepper fruits (cv. All Big). Ten microliters of bacterial suspension (10^7 CFU/ml) were deposited on wounds created with an entomological pin on the surface of pepper fruits. Six replicates were used for each isolate, with each repetition consisting of one pepper fruit. For negative and positive controls, pepper fruits were treated similarly with sterile distilled water and inoculated with one strain (CCRMPA35) of *Pectobacterium aroidearum* (Barroso et al. 2019). After 24 h of incubation at 28°C in a moist chamber, the soft rot symptoms were

observed, and the strains were reisolated to complete Koch's postulates. The negative control remained symptomless, whereas the positive control showed soft rot symptoms. The strains were identified using the Biolog Gen III System as *Pectobacterium* with a similarity level of 70%. Molecular identification was performed by sequencing a fragment of 1,400 bp of the 16S rRNA region (Hauben et al. 1998) and fragments of 450, 520, and 630 bp of the genes *gapA*, *icaA*, and *proA* (Ma et al. 2007). Using BLASTn analysis, the 16S rRNA region showed that the 14 strains shared 99 to 100% similarity with several species including *P. aroidearum* (SCRI109^T). The Bayesian inference analysis (BIA) carried out using the sequences of the 16S rRNA region of the 14 strains (GenBank MK450555, MN883867, MK450557, MK450559, MK450563, MK450568, MK450572, MN883869, MK450573, MK450576, MK450577, MK450580, MK450582, and MN883868) and sequences of all species and subspecies of *Pectobacterium*, obtained from GenBank, grouped these strains with the strain SCRI109^T, with a posterior probability above 90%. The strains CCRMPA109, CCRMPA637, and CCRMPA670 were selected for sequencing of the genes *gapA* (GenBank MK752922 to MK752924), *icaA* (GenBank MK752925 to MK752927), and *proA* (GenBank MK752928 to MK752930), and BLASTn analysis showed similarity values above 98% between these strains and SCRI109^T. The BIA performed using the concatenated sequences of genes *gapA*, *icaA*, and *proA* of three strains together with the sequences of all species and subspecies of *Pectobacterium*, obtained from GenBank, grouped the three isolates with SCRI109^T, with a posterior probability of 100%. *P. aroidearum* has been described as a plant pathogen with a preference for monocotyledonous plants (Nabhan et al. 2013). However, in recent years it has been observed causing soft rot in different dicotyledonous plants, such as zucchini (Moraes et al. 2017), lettuce, Chinese cabbage (Barroso et al. 2019) and, in the present study, pepper fruits. Thus, to our knowledge, this is the first report of *P. aroidearum* causing soft rot in pepper fruits in Brazil.

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