Disease Note

Diseases Caused by Bacteria and Phytoplasmas

First Report of Rhizome Rot of Banana Caused by *Klebsiella variicola* in India

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Rhizome rot or soft rot disease is one of the major problems in banana (*Musa* spp.) cultivation, because it causes germination failure and death of early-stage plants. A roving survey conducted during 2017 to 2019 in the major banana growing states of India indicated a 5 to 30% incidence of rhizome rot in commercial cultivars. The symptoms observed were yellowing of leaves, necrotic drying with or without heart rot, and yellow or brown water-soaked spots with dark brown margins in the rhizomes. Decay of tissues, cavity formation, brown ooze with foul smell, and toppling were also observed. To isolate bacteria, dissected diseased tissues were surface sterilized and plated on crystal violet pectate (CVP) medium. Of 60 samples plated on CVP medium, three samples collected from cultivars NeyPoovan-AB (Karur, Tamil Nadu, 10°56′36.8′′N; 78°24′12.5′′E), Grand Naine-AAA (Tiruchirappalli, Tamil Nadu, 10°47′26.1′′N; 78°34′14.8′′E), and Thellachakkarakeli-AAA (East-Godavari, Andhra Pradesh, 16°51′32.1′′N; 81°46′08.4′′E) did not yield any bacteria; however, when plated on nutrient agar, they produced whitish to dull white, mucoid, raised, round, and translucent colonies, and three isolates were named as NPK-3-48, GTC-5, and 1-1B-3, respectively. Because these colonies were distinct from colonies obtained on CVP medium (which were analyzed and confirmed separately as *Pectobacterium* sp.) (Gokul et al. 2019), they were further characterized. Amplification of 16S rDNA genes of NPK-3-48, GTC-5, and 1-1B-3 isolates using universal primers (27F, 5′-AGAGTTTGATCCTGGCTCAG-3′; 1492 R, 5′-GGTTACCTTGTTACGACTT-3′) and *rpoB* gene (Rosenblueth et al. 2004) was carried out; the amplicons were sequenced and deposited in NCBI (accessions MW036529 to MW036531; MW497572 to MW497574). Phylogenetic analysis of *rpoB* clearly showed that the isolates NPK-3-48, GTC-5, and 1-1B-3 are *Klebsiella variicola* (Rosenblueth et al. 2004). Additionally, biochemical tests also indicated that all three isolates were gram negative, catalase positive, oxidase negative, and able to utilize glucose, maltose, and citrate (Ajayasree and Borkar 2018). Therefore, the above morphological, molecular, and biochemical analyses carried out indicated that NPK-3-48, GTC-5, and 1-1B-3 are *K. variicola.* Earlier, *K. variicola* causing soft rot has been reported on banana in China (Fan et al. 2016), plantain soft rot in Haiti (Fulton et al. 2020), and carrot soft rot in India (Chandrashekar et al. 2018). For pathogenicity tests, these three isolates were grown in nutrient broth for 48 h at 37 ± 1°C, and the cells were harvested by centrifugation. Five milliliters of the culture suspension (2×10^8 CFU/ml) taken in a syringe was injected into rhizomes of 3-month-old tissue-cultured Grand Naine plants. Each bacterial isolate was injected into eight banana plants at soil level. Appropriate controls were maintained. Inoculated plants were maintained in a glasshouse at 32 ± 2°C, and after 30 to 35 days rhizome rot symptoms appeared in all three bacterial isolates inoculated plants but in none of the control plants. Koch's postulates were proved by reisolation and identification. To the best of our knowledge, this is the first report of *K. variicola* causing rhizome rot disease of banana in India.

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