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Disease Notes

First Report of *Calonectria hongkongensis* Causing Fruit Rot of Rambutan (*Nephelium lappaceum*)

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e-Xtra

Fruit rot of rambutan is a pre- and post-harvest disease problem of rambutan orchards. In 2011, fruit rot was observed at USDA-ARS orchards in Mayaguez, Puerto Rico. Infected fruit were collected and 1 mm^2 tissue sections were surface disinfested with 70% ethanol followed by 0.5% sodium hypochlorite. Infected fruit were rinsed with sterile, deionized, double-distilled water and transferred to acidified potato dextrose agar (APDA). Plates were incubated at 25 ± 1°C for 6 days. Three isolates of *Calonectria hongkongensis* (*Cah*), CBS134083, CBS134084, and CBS134085, were identified morphologically using taxonomic keys (2,3). In APDA, colonies of *Cah* produced raw sienna to rust-colored aerial mycelial growth. Conidiophores of *Cah* had a penicillate arrangement of primary to quaternary branches of 2 to 6 phialides. Conidia (*n* = 50) were cylindrical, hyaline, 1-septate, rounded at both ends, and 44 to 52 µm × 3.5 to 4.5 µm. Conidiophores produced terminal and lateral stipe extensions with terminal sphaeropedunculate vesicles that were 8 to 12 µm wide. Subglobose to ovoid perithecia, 300 to 500 µm × 200 to 350 µm and orange to red-brown, were produced in groups of 3. Asci were clavate and contained 8 ascospores aggregated at the top of the ascus. Ascospores (*n* = 50) were hyaline, guttulate, fusoid with rounded ends, straight to curved, 1-septate with constriction at the septum, and 28 to 36 µm × 4 to 7 µm. For molecular identification, the ITS rDNA, fragments of β-tubulin (BT), histone H3 (HIS3), and elongation factor (EF1-α) genes were amplified by PCR, sequenced, and compared using BLASTn with *Calonectria* spp. submitted to the NCBI GenBank. The sequences of *Cah* submitted to GenBank include accessions KC342208, KC342206, and KC342207 for ITS; KC342217, KC342215, and KC342216 for BT; KC342211, KC342209, and KC342210 for HIS3; and KC342214, KC342212, and KC342213 for EF1α.

The sequences were >99% or identical with the ex-type specimen of *Cah* CBS 114828 for all genes used. Pathogenicity tests were conducted on 5 healthy superficially sterilized fruits per isolate. Both scalpel-wounded and unwounded fruit tissues were inoculated with 5-mm mycelial disks from 8-day-old pure cultures grown in APDA. Untreated controls were inoculated with APDA disks only. Fruits were kept in a humid chamber for 8 days at 25°C under 12 h of fluorescent light. The test was repeated once. Three days after inoculation (DAI), white mycelial growth was observed on the fruit. Five DAI, the fruit changed color from red to brown and yellowish mycelia colonized 50 to 62% of the fruit surface. Eight DAI, all the fruit turned brown, the mycelium growth covered the entire fruit, and conidiophores were produced on spinterns (hairlike appendages). Fruit rot of spinterns, exocarp (skin), endocarp (aril), and light brown discoloration were observed inside the fruit. Untreated controls showed no symptoms of fruit rot and no fungi were reisolated from tissue. *Cah* was reisolated from diseased tissue, fulfilling Koch's postulates. *Calonectria* spp. (or their *Cylindrocladium* asexual states) have been associated with lychee decline syndrome in North Vietnam (1). Both fruits belong to the Sapindaceae family. To our knowledge, this is the first report of *Cah* causing fruit rot of rambutan.

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