First report of Neoscytalidium dimidiatum causing fruit rot on Guava (Psidium guajava L.) in Malaysia

ABSTRACT

Guava (Psidium guajava L.) is an economically important fruit crop in Malaysia with annual production of 67,087 tons in 2018 (FAO 2018). In February 2019, fruit rot symptoms were observed postharvest on approximately 30% of guava cultivar Lohan collected from a commercial orchard in the Rawang district (3°23′22.8″N, 101°26′55.7″E) of Selangor province, Malaysia. Symptoms on the fruits appeared as small, circular brown spots (ranging from 5 to 20 mm in diameter) that coalesced and rapidly expanded to cover the entire fruit. Severely infected fruits became soft and rotted. Ten diseased guava fruits were collected from the sampling location. Small pieces (5 × 5 × 5 mm) of symptomatic fruit tissues were excised from the lesion margin, surface sterilized with 0.5% NaOCl for 1 min, rinsed twice with sterile distilled water, plated on potato dextrose agar (PDA), and incubated at 25°C for 5 days. A Scytalidium-like fungus was consistently isolated from symptomatic tissues on PDA after 4 days. For morphological identification, single-spore cultures were grown on PDA at 25°C, and a representative isolate (LB1) was characterized further. The fungal colonies were initially white and powdery, and they later turned grayish-black with the onset of sporulation. The mycelia were branched with septa, pigmented, and brown in color. Fungal colonies produced dark-brown arthroconidia with thick-walled, had 0 to 1 septa, averaged 9 × 5 µm (n = 20), and were cylindrical to oblong in shape. For molecular identification, genomic DNA was extracted from fresh mycelium of isolate LB1 using a DNeasy Plant Mini kit (Qiagen, Germantown, MD). The internal transcribed spacer (ITS) region of rDNA and translation elongation factor 1-alpha (TEF1-α) gene were amplified using ITS5/ITS4 (White et al. 1990) and EF1-728F/EF1-986R primer sets (Carbone and Kohn 1999), respectively. Both ITS (954-bp) and TEF1-α (412-bp) sequences exhibited 100% identity to Neoscytalidium dimidiatum with GenBank accession numbers FM211432 and MK495414, respectively. The resulting sequences were deposited in GenBank (ITS, accession no. MT565490; TEF1-α, accession no. MT572846). Based on the morphological and molecular data, the pathogen was identified as N. dimidiatum (Penz.) Crous & Slippers (Crous et al. 2006). A pathogenicity test was conducted on five healthy detached mature guava fruits cultivar Lohan by wound inoculating using a sterile needle and pipetting 10 µl of a conidial suspension (1 × 106 conidia/ml) of isolate LB1 to the wound. Five additional fruits were wounded and pipetted with 10 µl of sterile distilled water to serve as controls. Inoculated fruits were placed in a sterilized plastic container and incubated at 25 ± 1°C, 90% relative humidity, with a photoperiod of 12 h, and the experiment was conducted twice. All inoculated fruits developed symptoms as described above 4 to 7 days postinoculation, whereas the control fruits remained asymptomatic. N. dimidiatum was reisolated from all symptomatic tissues, confirming Koch's postulates. N. dimidiatum has been reported causing brown spot disease on pitaya (Lan et al. 2012) and stem canker on dragon fruit in Malaysia and Florida (Mohd et al. 2013; Sanahuja et al. 2016), but this is the first report of N. dimidiatum causing postharvest fruit rot on guava in

Malaysia. This disease can cause significant postharvest losses to guava production, which could lower marketable yield, and proper control strategies should be implemented.

Keyword: Fruit rot; Neoscytalidium dimidiatum; Guava