Diseases Caused by Fungi and Fungus-Like Organisms

First Report of *Neovaginatispora fuckelii* Causing Leaf Spot on *Phoenix canariensis*

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Phoenix canariensis Chabaud is an ornamental widely planted in urban China (Lan et al. 2020). In December 2019, six of seven *P. canariensis* plants exhibited symptoms of leaf spot in Chengdu campus of Sichuan Agricultural University (SAU) in Sichuan, China, with ~80% of leaves per plant affected. Symptomatic leaves initially showed dark brown spots with a yellow halo at the periphery. Spots gradually expanded and eventually formed necrotic spots, with the whole leaf dying when severely infected. Ten diseased leaves from different plants were collected and deposited in the SAU Herbarium. A single-spore isolation was performed (Chomnunti et al. 2014) and transferred to PDA. Five fungal isolates from five different infected leaves were deposited in the SAU Culture Collection. Fungal colonies were incipiently greyish-white, becoming pale brown at the margins and dark brown in the center. The reverse was the same as the obverse with radial cracking at the center. Morphological characteristics on the host were examined by light microscopy. Ascomata were semi-immersed to immersed, black, pyriform to subglobose, and 180 to 310 × 140 to 260 µm. The peridium comprised several layers of hyaline cells of textura angularis and was 9 to 27 µm wide. Pseudoparaphyses were numerous, septate, hyaline, usually longer than asci. Asci were 8-spored, bitunicate, cylindric-clavate, and 50 to 95 × 8 to 11.5 µm ($n = 35$). Ascospores were 1-septate, straight or slightly curved, hyaline, usually with globose appendages at both ends, and 14 to 18.5 × 4 to 6.5 µm ($n = 50$). Conidiogenesis was not observed on PDA incubated 4 months at 25°C and 95% RH on a 12-h fluorescent light/dark incubation. Morphology was

consistent with the sexual stage of *Neovaginatispora fuckelii* (Sacc.) A. Hashim. et al. described by Hashimoto et al. (2018), Tennakoon et al. (2018), and Hyde et al. (2020). The rDNA ITS, 28S LSU, 18S SSU, TEF 1- α , and RPB2 gene regions were amplified by using the primers ITS5/ITS4, LROR/LR5, NS1/NS4, EF1-983F/EF1-2218R, and fRPB2-5F/fRPB2-7cR, respectively. The DNA amplification products of representative isolate SICAUCC 20-0008 were sequenced (GenBank MT427731, MT427734, MT427738, MT441923, and MT441920, respectively). Nucleotide BLAST showed these five sequences were greatly identical with *N. fuckelii* (GenBank: ITS, LC001732, 522/524, 99.62%; LSU, AB619009, 839/840, 99.88%; SSU, AB618690, 976/976, 100%; TEF 1- α , LC001750, 895/912, 98.14%; RPB2, MN482130, 935/935, 100%). Using phylogenetic analysis in which GenBank reference sequences were included, SICAUCC 20-0008 clustered within *N. fuckelii* with high bootstrap support. To conduct Koch's postulates, 15 healthy *P. canariensis* 2- to 3-year-old plants were inoculated by placing a mycelium plug from the growing margin of 15-day-old colonies upside down directly onto fresh wounds punctured with a fine needle. Five healthy plants were inoculated with a sterile agar plug as the control. Plants were incubated in a growth chamber at 25 ± 1°C and 95% RH on a 12-h fluorescent light/dark regimen. After 10 days, the leaves of inoculated plants turned brown and gradually expanded and became necrotic spots, similar to symptoms observed in the field. Control plants were symptomless. The pathogen was reisolated from leaf lesions and identified by morphological characteristics; no fungus was recovered from control treatments. *N. fuckelii* has often been reported as a saprobe on numerous hosts including species of *Acer*, *Carpinus*, *Carya*, *Epilobium*, *Prunus*, *Quercus*, *Rhus*, *Salix*, and *Vitis*, as well as other genera (Hashimoto et al. 2018; Hyde et al. 2020; Thambugala et al. 2015). This is the first report of *N. fuckelii* causing leaf spot on *P. canariensis*. Some proteaceous plants and other palms are known to be infected by this fungus (Hyde et al. 2000; Taylor et al. 2000), which may result in a threat to ornamental horticulture. Fungicide treatments should be considered to prevent damage to plants and spread of this fungus.

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