Diseases Caused by Fungi and Fungus-Like Organisms

Nigrospora oryzae Causing Leaf Spot on Asiatic Dayflower in Chongqing, China

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Asiatic dayflower (*Commelina communis* L.) is an annual herbaceous weed that is distributed throughout China. A foliar disease on Asiatic dayflowers was discovered in one farm field in Dianjiang County, Chongqing, China (30°3'22" N, 107°18'5" E) in summer 2019. The disease incidence was observed on about 10% (13/127) of the plants. Symptoms appeared as round, tan lesions 2 to 5 mm in diameter that occurred randomly and irregularly on the whole leaves. The centers of lesions become grayish white with reddish borders as the disease progressed. The leaves with typical symptoms were detached and wiped with 70% ethanol for surface disinfestation before isolating the causal agent. Subsequently, three pieces (3 to 4 mm²) of tissue were taken from the margin of the leaf lesion, disinfested in 1.5% NaClO for 1 min, rinsed three times in sterilized distilled water, and placed onto potato dextrose agar (PDA) medium containing 50 µg/ml each of kanamycin and ampicillin. A fungus was exclusively and consistently isolated from the disinfested leaf lesion sections. The colonies on PDA grew rapidly and covered the entire Petri dish within 5 days at 28°C. Colonies were at first grayish white, cotton wool-like, round, with abundant aerial mycelium, and later turned black as conidia were produced. The abundant conidia formed on PDA were initially yellow brown and gradually became black, oblate to ellipsoidal, smooth, single-celled, and ranged

in size from 4 to 10 × 3.5 to 9 µm. They were borne on a colorless, hyaline, and inverted flask-shaped cell at the tip of each conidiophore. The morphology characteristics were consistent with those of *Nigrospora* spp. (Wang et al. 2017). Genomic DNA was extracted from one representative isolate NDJ0819. The amplification and sequencing of the gene fragments including the internal transcribed space (ITS) region of ribosomal DNA and beta-tubulin were performed using the primers ITS1/ITS4 (White et al. 1990) and Bt2a/Bt2b (Glass and Donaldson 1995), respectively. Fragments of 536 bp for ITS and 408 bp for beta-tubulin were obtained. A phylogram of the combined ITS and beta-tubulin sequences reconstructed using the maximum likelihood bootstrapping method implemented in the software MEGA version 7.0 (Kumar et al. 2016) indicated that isolate NDJ0819 clustered with *Nigrospora oryzae*. Both ITS and beta-tubulin sequences were deposited into GenBank (accession nos. MT140353 and MT157509, respectively). Pathogenicity test was performed by rub-inoculating needle-wounded leaves of three 4-week-old Asiatic dayflowers with a spore suspension (2.6 × 10⁶ conidia/ml) of NDJ0819 prepared in water containing 0.05% Tween-20, and holding plants at 28°C in the growth chamber. The pathogenicity test was repeated twice. Brown, round lesions developed on leaves inoculated with spores at 15 days postinoculation. However, the centers of the lesion did not become grayish white, compared with those of lesions seen in naturally infected leaves. No symptoms developed on leaves inoculated with sterilized distilled water. *N. oryzae* was reisolated from the lesions. All results described above indicated that *N. oryzae* was responsible for the leaf spot of Asiatic dayflower. To our knowledge, this is the first report of *N. oryzae* causing leaf spot on Asiatic dayflower in China. Research into the potential use of *N. oryzae* as a candidate biological agent against the weed is worth being initiated.

References:

- Glass, N. L., and Donaldson, G. C. 1995. Appl. Environ. Microbiol. 61:1323.
Kumar, S., et al. 2016. Mol. Biol. Evol. 33:1870.
Wang, M., et al. 2017. Persoonia 39:118.
White, T. J., et al. 1990. Page 315 in: PCR Protocols: A Guide to Methods and Applications. Academic Press, San Diego, CA.

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