Diseases Caused by Fungi and Fungus-Like Organisms

First Report of Leaf Spot on *Scaevola taccada* Caused by *Alternaria longipes* in China

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As a bushy shrub, *Scaevola taccada* (Goodeniaceae) is widely distributed in the coastal islands of the Pacific and Indian oceans (Goldstein et al. 1996). It is a salt- and drought-tolerant species, exhibiting high photosynthetic capacity and water-use efficiency. In May 2018, a leaf spot disease was observed on *S. taccada* during a disease survey of the native plants on the Xisha Islands, China. Initial symptoms were small, round or elliptical brown lesions on the leaves, which subsequently enlarged and coalesced, resulting in large, circular necrotic areas in about 10 days. Ultimately, the leaves turned yellow, died, and fell off in a month. In all cases, a few isolates were studied, and the same fungus was consistently isolated from symptomatic leaves. A single-spore isolate of the fungus (isolate No. 35) was selected at random, for pathogenicity test, and characterization was carried out based on morphology as well as the ribosomal DNA internal transcribed spacer (ITS) regions and glyceraldehyde-3-phosphate dehydrogenase (GAPDH) gene sequences. The healthy leaves were artificially inoculated with 5-day-old mycelium-agar disks (5 mm in diameter) or with 20 µl of a conidial suspension (10⁶ conidia/ml) in water. The controls were inoculated with sterile potato dextrose agar (PDA) medium and distilled water, respectively. The inoculated leaves were maintained at 25°C

with a relative humidity of 90 to 95% in a growth chamber. The tests were repeated five times. At 15 days postinoculation, leaves inoculated with mycelial plugs and conidia showed unambiguous brown spots, which were similar to the symptoms observed on the original leaves. However, control leaves remained symptomless. The same fungus was successfully recovered from symptomatic plants and confirmed according to morphological characteristics, which completed Koch's postulates. The fungal colony was round with a smooth edge on PDA medium. The colony was initially white or white-gray, and then it turned dark-green in the center with a white-gray edge with age. Conidia produced on potato carrot agar medium were in short, straight chains usually or rarely single. Conidia were ellipsoid or obclavate, olive to black brown, 20.0 to 85.0 × 7.8 to 16.5 µm (*n* = 100), with three to seven transverse septa and zero to two longitudinal septa. Some conidia produced long beaks. The boundary between conidia and beak was obvious. This pathogenic fungus was identified as *Alternaria* sp. based on morphological characteristics (Simmons 2007). DNA was extracted from the mycelium of a 10-day-old culture. The ITS and GAPDH gene regions were amplified using ITS1/ITS4 (Long et al. 2009) and gpd1/gpd2 primers (Berbee et al. 1999), respectively. The obtained ITS sequences (GenBank accession no. MN178643) of isolate No. 35 showed a sequence identity of 100% to the type specimen of *Alternaria longipes* (GenBank accession no. AY278835). GAPDH regions of 589 bp (GenBank accession no. MN256460) showed an identity of 99% to the type specimen of *A. longipes* (GenBank accession no. AY278811) (Pryor and Bigelow 2003). The fungus was identified as *A. longipes* using ITS regions and GAPDH gene sequences. To the best of our knowledge, we, for the first time, report leaf spot of *S. taccada* caused by *A. longipes* in China. To protect the native plants and the ecological environment of the Xisha Islands, China, it is necessary to develop an integrated management strategy to control the disease in *S. taccada*.

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