Diseases Caused by Fungi and Fungus-Like Organisms

First Report of Leaf Spot Disease Caused by *Alternaria brassicicola* on Broccoli in Papua New Guinea

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During 2017 and 2018, severe symptoms of dark spots on leaves were observed on broccoli plants (*Brassica oleracea* [L.] var. *italica*) cultivar 'Shogun' in two farmers' fields, with 45 to 37% disease incidence and 70% yield loss, in Massy and Kabiufa villages of Goroka District of Eastern Highlands Province, Papua New Guinea. The symptoms on 6-week-old plants consisted of darkened spots with concentric rings and sunken brown necrotic lesions on the leaves measuring 3 × 6 mm. The lesions on severely infected leaves showed a shot-hole appearance. Tiny brown-colored warts on the stem and stalks of the flower heads were also observed. Ten infected leaf and stem samples from the margin of the necrotic area were collected, surface sterilized with 0.2% NaOCl, washed with sterile distilled water, and plated on potato dextrose agar (PDA) for 7 days at 23°C. The incubated samples all produced similar cultural morphology. The mycelium of the 7-day-old single-spore pure culture grew a greyish white to brown color fungal mat. Hyphae were branched, septate, brownish with simple olive brown septate conidiophores. The conidia were 18 to 54 µm long and 7 to 11 µm wide, solitary or in short chains, mostly ovoid with short conical or

cylindrical apical beaks or no beaks, and smooth walled. The morphological characters of the isolate were consistent with the previous report of Lawrence et al. (2013). The genomic DNA was isolated from the isolate and amplified using fungal-specific primer pair ITS1/ITS4 (White et al. 1990), and the amplicon was sequenced. BLAST analysis revealed 97% identity to *Alternaria brassicicola*, a pathogen that causes leaf spot of Brassicaceae members (Woudenberg et al. 2013). Sequence data of accession number MN700129 showed close resemblance with accession number JN108901 in the NCBI database. A pathogenicity test was conducted on five 6-week-old broccoli plants (cv. Shogun) under greenhouse conditions. The plants were sprayed with a 7-day-old-culture suspension of 9×10^5 conidia/ml in sterilized distilled water until run-off; plants without inoculation served as the control, and the experiment was repeated twice. Development of similar symptoms observed in the farmers' fields appeared after 7 to 11 days on all the inoculated plants, whereas the control plants remained symptomless. The fungus was reisolated on PDA plates from the inoculated plants, producing similar morphological characters with the isolate, which confirmed Koch's postulates. This is the first report of *A. brassicicola* causing broccoli leaf spot disease in Papua New Guinea. Because *A. brassicicola* inoculum can survive in the soil for a long period of time, the disease could be a threat to broccoli production. Hence, there is a need to develop effective disease management strategies to reduce damage caused by the disease in the affected areas.

References:

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