Diseases Caused by Fungi and Fungus-Like Organisms

First Report of Stem Rot Disease of Miracle Plant (*Bryophyllum pinnatum*) Caused by *Athelia rolfsii* in Tripura, India

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Plant Dis. 105:708, 2021; published online as <https://doi.org/10.1094/PDIS-05-20-1086-PDN>. Accepted for publication 1 October 2020.

Miracle plant (*Bryophyllum pinnatum* [Lam.] Oken) is a succulent plant native to Madagascar. It is also known as Paṣaṇabheda in Ayurveda, India, and noted for its effective use against an array of human diseases, and it has become cultivated in tropical and subtropical areas (Yadav et al. 2016). Miracle plants exhibiting symptoms of an unknown stem rot disease having characteristic white mycelial mats and sclerotia at the crowns were observed in the middle of July 2018 at the medicinal garden of the College of Agriculture, Tripura University, in Lembucherra, Tripura (India). Out of 150 plants examined, 20 plants were found to be symptomatic. Several plant samples were collected and surface disinfected with 0.25% sodium hypochlorite for 1 min, triple washed with sterilized distilled water, placed in Petri plates containing laboratory-made potato dextrose agar (PDA), and incubated at 25°C. After 1 week of incubation, mycelia and sclerotia were present, and the hyphal tips were transferred onto fresh PDA plates to obtain pure cultures. Pure cultures with abundant white aerial mycelia with typical clamp connections were observed under a microscope. Initial growth of sclerotia required 7 days on PDA, but after 14 days sclerotia were spherical and dark black in color with an average size of 0.8 to 1.2 mm in diameter. The fungal cultures were tentatively identified as *Athelia* sp. based on morphology (Aycok 1966). Identification was confirmed by molecular characterization. Pure fungal sclerotia were transferred into a

flask containing potato dextrose broth medium for mycelial mat preparation and kept at 25°C. The genomic DNA was extracted from mycelial mats using a Zymo Research fungal/bacterial DNA miniprep kit (Irvine, CA), and the internal transcribed spacer region of rDNA was amplified using universal primers ITS1 and ITS4 (White et al. 1990). A PCR product of 650 bp was amplified, sequenced, and deposited in GenBank (MN121532). The resulting sequence showed 99% similarity with *Athelia rolfsii* strain CBS: 132553, GenBank accession JX566993. Based on morphological and molecular characterization, the fungus was identified as *A. rolfsii*. A greenhouse study was conducted in triplicate to confirm Koch's postulates. Three 3-month-old miracle plants were inoculated with 10-day-old mycelia grown in potato dextrose broth. The stem of each miracle plant was artificially injured and then wrapped with moistened sterilized cotton containing mycelia of *A. rolfsii*. Three separate miracle plants were artificially injured and inoculated with sterile distilled water and served as a negative control. After 15 days, symptoms of stem rot on the inoculated plants developed and were similar to those observed in the field. The fungus was reisolated from the inoculated plants using the procedure described above and identified based on cultural and morphological characterization. No symptoms were observed on the negative control plants. A culture of isolated fungus was deposited in the Indian Type Culture Collection, Division of Plant Pathology, ICAR-IARI, New Delhi, India (ITCC 8432). To the best of our knowledge, this is the first report of stem rot disease of miracle plant caused by *A. rolfsii*. Due to extensive medicinal application, miracle plants have significant economic value, and therefore this disease report is of great significance.

References:

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The author(s) declare no conflict of interest.

Keywords: *Athelia rolfsii*, stem rot, miracle plant, sclerotia

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