



First report of *Xylaria* sp. causing tuber rot on glory lily in India

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Glory lily (*Gloriosa superba*, family *Colchicaceae*) is a medicinal plant grown commercially in India. The tubers and seeds are rich in colchicines, isoperlopyrine and tropolane alkaloids which have anti-gout, anti-inflammatory and anti-tumour activity (Pandey *et al.*, 2008).

In July 2015, a survey was conducted in the major growing areas of Tamil Nadu state. A tuber rot disease was observed at the flowering stage (90 days after planting) in all surveyed areas, that is the Dindigul, Erode, Karur, Salem and Tiruppur districts of the state. Initial disease symptoms were yellowing and drying of leaves (Fig. 1) and white mycelial growth on the collar region of stem and roots (Fig. 2), which led to rotting of tubers (Fig. 3) and a severe loss in seed yield. Infected tubers were surface sterilised using 0.1% mercuric chloride solution. A fungus was consistently isolated from diseased tubers plated on potato dextrose agar medium and incubated at 25 ±2°C (Fig. 4). The fungal colony was initially creamy white, later turning greyish black in colour. Single-celled cylindrical hyaline conidia measuring 3-4 µm × 1-2 µm were produced at the tips of conidiophores. Based on the morphological characters, the fungus was identified as *Xylaria* sp. and the identification was confirmed by the Indian Type Culture Collection, Indian Agricultural Research Institute, New Delhi (reference no. 868/88).

Molecular identification was done using fungal-specific primers (unpublished commercial primers) for amplification of ITS 1 and ITS 2 regions of rDNA (forward, 5'-TCMGTAGGTGAPCCWBCGS-3' and reverse, 5'-TCCTNCGYTKATKGUTADGH-3'), using an initial denaturation step at 94°C for 5 minutes, 35 cycles of 30 seconds at 94°C, 30 seconds at 52°C and 45 seconds at 72°C, and final extension at 72°C for 7 minutes (Lee *et al.*, 2000). The 650 bp PCR product was sequenced (GenBank Accession No. KX419439) and BLAST analysis showed 99% identity with a *Xylaria* sp. isolated from mushroom in India (KR155088). A phylogenetic tree was constructed and the fungus was shown to be an undescribed *Xylaria* sp., closely related to isolate R006 from India (KC405623.1).

To confirm pathogenicity, sand and ground maize seeds were mixed (19:1, v/v, respectively), moistened with water and sterilised by autoclaving. The

fungus was inoculated into the medium and incubated for 28 days at 28 ±2°C (Riker & Riker, 1936). Earthen pots (25 cm diameter) were filled with 5 kg potting medium (equal volumes of red soil, sand and farmyard manure). The medium was sterilised by autoclaving and inoculated with 5 g *Xylaria* inoculum. Healthy tubers of *G. superba* were planted in pots, maintained in a glasshouse with uniform watering, and assessed after 90 days. The inoculated plants developed symptoms similar to those observed in the field, including yellowing of leaves and rotting of tubers. Koch's postulates were fulfilled by re-isolation of the fungus from the infected tubers and molecular identification of *Xylaria* sp. by sequencing. Control plants did not show any symptoms.

A tuber rot disease of glory lily caused by *Macrophomina phaseolina* was observed by Alice & Sundravadhana (2012). However, to the best of our knowledge, this is the first report of *Xylaria* sp. infecting glory lily and causing tuber rot disease, either in India or worldwide.

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Figure 1



Figure 2



Figure 3



Figure 4

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