



First report of *Curvularia* leaf spot on gladiolus from Kashmir

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Gladiolus (Iridaceae), a bulbous plant renowned for its vibrant colors, size, and elegant flower structure, holds global popularity as an ornamental garden flower and a favored choice for cut flower arrangements. In the spring, 2023, white pin-spots, which later turned brown, emerged on *Gladiolus grandiflorus* leaves within a commercial cut flower field situated at Shalimar, Srinagar, Jammu and Kashmir. The spots transformed into oval to circular spots as they increased in size and were encircled by a conspicuous yellow halo (Fig.1a).

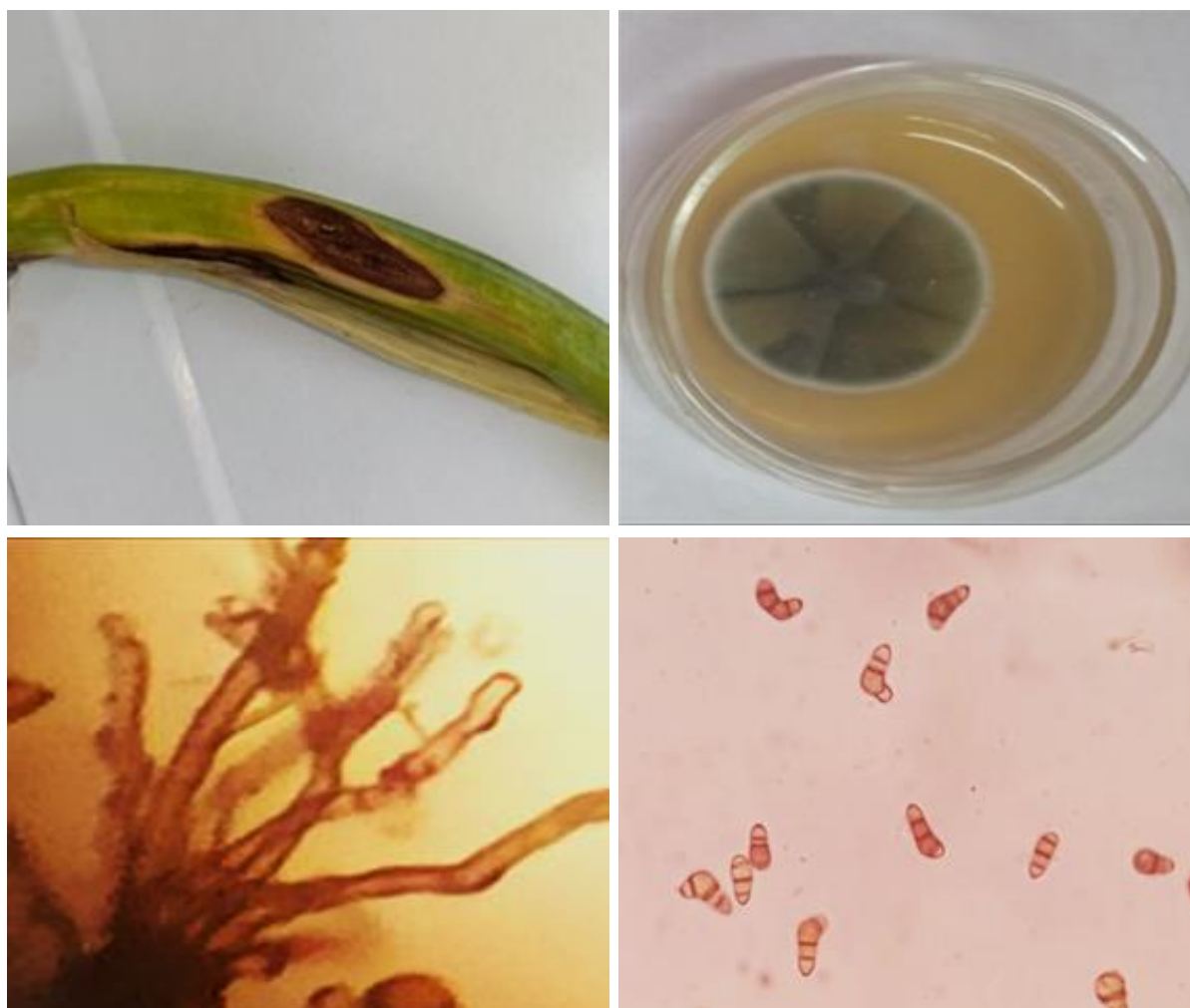


Fig. 1: Morphological characters of *Curvularia gladioli* (a) Dark brown oval spots with yellow halo on leaf surface, (b) Colony of *C. gladioli* on PDA media (c) Conidiophore (d) Distoseptate conidia having curvature at the third cell from the base

The spots enlarged and ultimately coalesced which gave a manifestation of leaf blight. The symptomatic leaves were collected from the field and used for the isolation of the causal pathogen by using the tissue bit isolation technique. The purification of the isolated fungus was made by single spore method. The fungal colony appeared white initially and then olivaceous in colour with whitish margins (Fig. 1b). The conidiophores were flexuous to cylindrical, septate, light brown in colour, emerged singly or in clusters (Fig. 1c) and measured 52.0-83.5 x 3.5-8.0 μm in size. The conidia were 3-distoseptate, exhibited curvature at the third cell from the base (Fig. 1d) and measured 24.0-33.0 x 10.5 to 16.5 μm in size. The third cell displayed a distinct swelling on one side, rendering it larger than the other cells. These morphological features were consistent with the established description of *Curvularia gladioli* Boerema & Hamers (Boerema and Hamers, 1989; Torres *et al.*, 2013). For molecular taxonomic confirmation, DNA was extracted using CTAB method. PCR was performed using ITS3 and ITS 4 primers to amplify the internal transcribed spacer (ITS) region of rDNA. The retrieved sequence (accession number R497775) was subjected to BLAST in NCBI that showed 99.66 percent base pair matching with the sequences of *C. gladioli* isolates from Netherlands (accession number HG778987) and Switzerland (accession number LT631345) (Fig. 2).

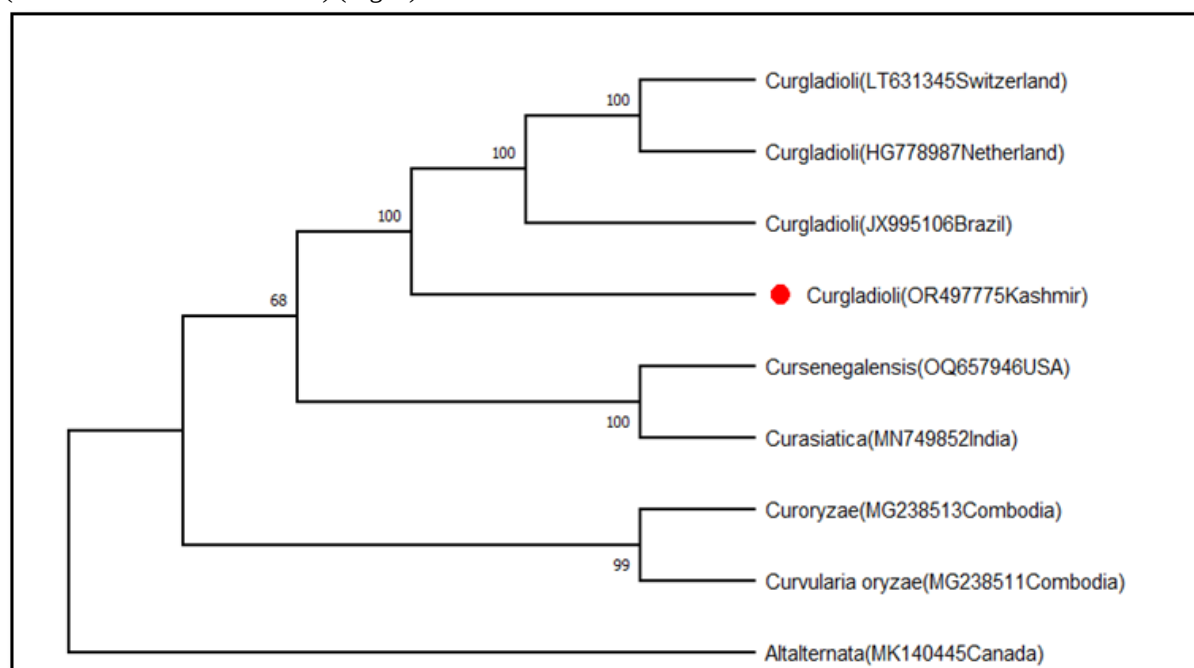


Fig. 2: Phylogenetic analysis of *Curvularia gladioli* based on neighbor joining analysis of Internal Transcribed Spacer (ITS) gene sequence

To confirm Koch's postulates, one month old healthy gladiolus plants were deliberately wounded by inoculation needle. The wounded leaves were sprayed with a conidial suspension (2×10^4 conidia ml^{-1}) of isolated pathogen (Zhang *et al.*, 2021). The control plants were also wounded and sprayed with distilled water. These experimental samples were placed within a greenhouse at a temperature of 25°C and 90% relative humidity. The inoculated leaves showed brown leaf spots within 4 days of inoculation. These symptoms were consistent with those initially observed on *C. gladioli*. However, the leaves of control plants remained symptomless. The pathogen was successfully re-isolated from the symptomatic tissue and confirmed by morphology and molecular identification as stated above. To our knowledge, this is the first report of *C. gladioli* causing leaf spots on gladiolus from UT of Jammu and Kashmir. Identification of this pathogen will help to develop integrated management strategies against this disease.

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