*Phytopathological note*

# FIRST REPORT OF *FUSARIUM OXYSPORUM* CAUSING LEAF BLIGHT AND DIE BACK DISEASE OF LILY IN BANGLADESH

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ABSTRACT

Alam, K. M., Rahman, M. S., Alam, M. M., Arifunnahar, Islam, M., Momotaz, R., Islam, R., and Apu, M. M. R. B. 2022 First report of *Fusarium oxysporum* causing leaf blight and die back disease of lily in Bangladesh. Bangladesh J. Plant Pathol. 38(1&2): 65-68.

Lily (*Lilium* spp.) is a commercial flower crop newly introduced in Bangladesh. During May to June 2018 severe leaf blight and dieback symptoms about 10 % of lily plants were found in the field of Horticulture Research Centre, Gazipur, Bangladesh. Disease symptoms included brown to black water soaked lesion on leaves, stems and flower buds, appear die back, leaves chlorosis, defoliation, brown spots on bulbs, resulting blighting and death of lily plants. The causal organism was isolated and identified as *Fusarium oxysporum* through morphological, pathogenicity and molecular study. The ITS region sequence was documented under the Genebank accession number MH879787. Blast analysis showed that 98% identical to *F. oxysporum*.

Key words: *Lilium*, leaf blight, die back, *Fusarium oxysporum*

Lily (*Lilium* spp.) is a newly introduced horticultural crop in Bangladesh, which has gained great interests from farmers and users due to its high commercial value. The crop is severely affected by leaf blight disease incurring up to 10 % death of lily plants during May and June 2018, in the field of Horticulture Research Centre, Gazipur, Bangladesh. Disease symptoms included brown to black water soaked lesion on leaves, stems and flower buds, appear die back, leaves chlorosis, defoliation, brown spots on bulbs, resulting dwarfing and death of lily plants (Fig. 1).

Samples were collected from diseased plants from the infected fields. Small pieces of symptomatic leaves, stems and bulbs from 15 different plants were surface sterilized with 1% sodium hypochlorite for 5 min and then washed three times in sterilize distilled water. The tissues were placed on half strength potato dextrose agar (PDA) and incubated at 25°C for 3 days. Fungal growth observed on PDA, conidia were spread over 4% water agar, incubated at 25°C for 5 days and hyphal tips transferred to PDA. Seven isolates were purified following hyphal tips technique. Micro conidia (n= 100) were found on PDA in a loose connection with monophialitic conidiophore which look like false heads, oval or cylindrical, 5.5 to 12.5 µm long x 1.5 to 2.5 µm wide. For induction of macro conidial sporulation, weak media (mung bean agar) was prepared by mixing 20 g agar in 1 liter boiled water of 40 g mung bean seed boiled water up to first seed coat open. Macro conidia (n=50) were straight to slightly curved, 3-5 septate and 20 to 32 µm long x 3 to 4 µm wide (Fig. 2). Morphological characteristics were consistent with *Fusarium oxysporum* (Leslie and Summerell 2006).

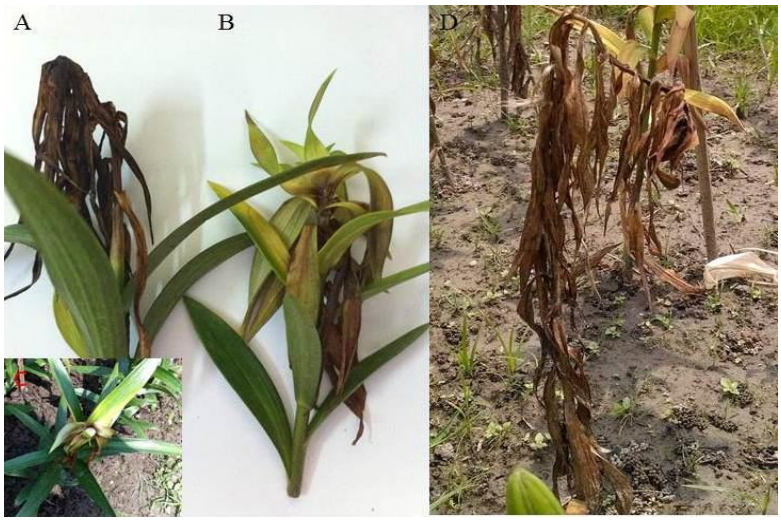


Figure 1: Symptom of leaf blight and die back disease of lily observed in the naturally infected field, the initial symptoms (A, B, C) and the whole plant died in the later stage (D)

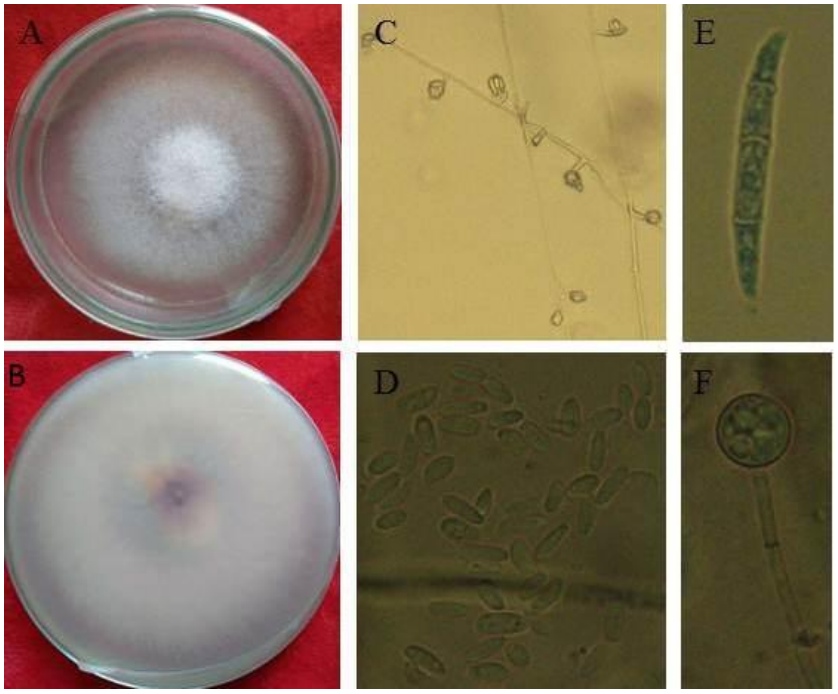


Figure 2: Cultural and morphological characters of F. oxysporum. A; Upper side of the pure culture on PDA, B; lower side of culture plate on PDA. C; false heads of microconidia with monophialidic chonidiophore. D; microconidia. E; Macroconidia.F; Chlamydospore.

To identify the fungus *Fusarium* to species level, universal primers ITS1/ITS4 (White *et al*. 1990) were used for PCR-based molecular identification. The amplified region of FOL (*Fusarium oxysporum* of Lily) isolate was sequenced and submitted to Gene bank (Accession No. MH879787). Blast analysis showed that almost 98% identical to *F. oxysporum* (Gene bank Accession Nos. MN 533762, MN 633364).

Evolutionary analysis of the FOL isolates (Accession No. MH879787) were performed using MEGA11 [2] software. From the phylogenetic analysis of ITS region, it was observed that FOL isolates in the clade of *Fusarium oxysporum* nucleotides (Fig. 3). The FOL isolates was marked with red arrow in the tree. Other *Fusarium* species nucleotides formed three different clades. Two out group nucleotides, *Botrytis cinerea* and *Alternaria alternata* were formed individual group.

For performing pathogenicity test, detached leaves were inoculated by three isolates (FOL, FOL 3 and FOL 6) in the laboratory. Mycelia and conidial plugs (2 mm diameter) were taken from a five-day-old culture and placed on upper surface of detached leaves of lily. The inoculated leaves were placed on sterilized distilled water soaked blotter paper for ensuring sufficient moisture in petri dish and incubated in 12 hours light and 12 hours dark at 25°C. Water soaked brown to black lesion symptom was developed on leaves after 3 days of artificial inoculation (Fig. 4) and the fungus was re-isolated and showed the same characteristics as described above.

On the basis of the morphological and molecular study, the tested fungus was identified as *F. oxysporum*. The fungus *F. oxysporum* was reported previously in lily crops of Netherlands, Italy, Poland, Spain and USA (Löffler *et al.* 1995, Prados-Ligero *et al.* 2008). It was seemed to be the first report of *F. oxysporum* causing leaf blight and die back disease of lily in Bangladesh.

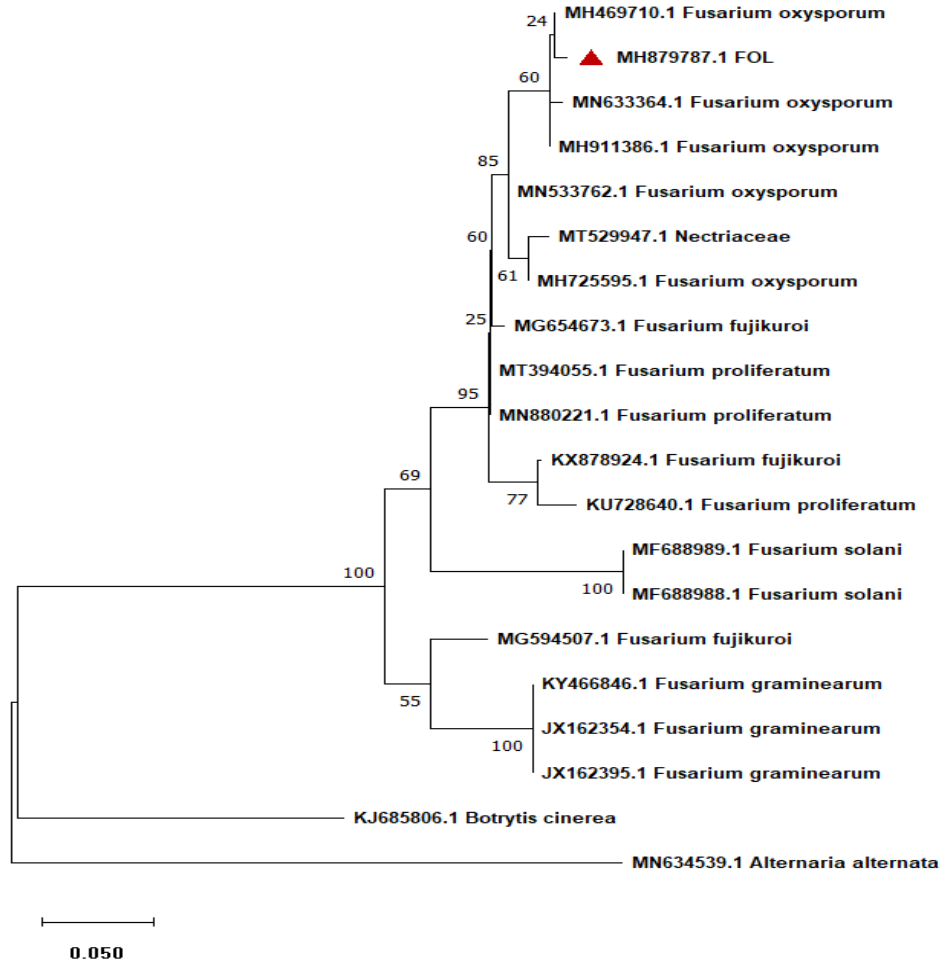


Figure 3. Phylogenetic tree by Maximum Likelihood method of internal transcribed spacer region of ribosomal DNA of FOL isolates of Lily


Figure 4: Symptom observed in the lily leaf after artificial inoculations. A; inoculated leaves, B; un-inoculated control leaves.

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