

First Report of *Fusarium incarnatum-equiseti* species complex causing leaf spot on rockmelon (*Cucumis melo* L.) in Malaysia

ABSTRACT

Rockmelon, (*Cucumis melo* L.) is an economically important crop cultivated in Malaysia. In October 2019, severe leaf spot symptoms with a disease incidence of 40% were observed on the leaves of rockmelon cultivar Golden Champion at the Faculty of Agriculture, Universiti Putra Malaysia. Symptoms appeared as brown necrotic spots, 10 to 30 mm in diameter, with spots surrounded by chlorotic halos. Pieces (5 × 5 mm) of diseased tissue were sterilized with 0.5% NaOCl for 1 min, rinsed three times with sterile distilled water, plated onto potato dextrose agar, and incubated at 25°C for 7 days with a 12-h photoperiod. Nine morphologically similar isolates were obtained by using the single-spore isolation technique, and a representative isolate B was characterized further. Colonies were abundant, whitish aerial mycelium with orange pigmentation. The isolates produced macroconidia with five to six septa, tapered with pronounced dorsiventral curvature, and measured 25 to 30 µm long × 3 to 5 µm wide. Microconidia produced after 12 days of incubation were single-celled, hyaline, ovoid, nonseptate, and 1.0 to 3.0 × 4.0 to 10.0 µm. Morphological characteristics of the isolates were similar to the taxonomic description of *Fusarium equiseti* (Leslie and Summerell 2006). Genomic DNA was extracted from fresh mycelium using the DNeasy Plant Mini kit (Qiagen, U.S.A.). To confirm the identity of the fungus, two sets of primers, ITS4/ITS5 (White et al. 1990) and EF1-728F/EF1-986R (Carbone and Kohn 1999), were used to amplify complete internal transcribed spacer (ITS) and partial translation elongation factor 1-alpha (TEF1-α) genes, respectively. BLASTn search in the NCBI database using ITS and TEF1-α sequences revealed 99 to 100% similarities with species of both *F. incarnatum* and *F. equiseti*. BLAST analysis of these in the FUSARIUM-ID database showed 100 and 99% similarity with *Fusarium incarnatum*-*F. equiseti* species complex (FIESC) (NRRL34059 [EF-1α] and NRRL43619 [ITS]), respectively (Geiser et al. 2004). The ITS and TEF1-α sequences were deposited in GenBank (MT515832 and MT550682). The isolate was identified as *F. equiseti*, which belongs to the FIESC based on morphological and molecular characteristics. Pathogenicity testing was conducted on five healthy leaves of 1-month-old rockmelon (cv. Golden Champion) grown in five plastic pots filled with sterile peat moss. The leaves were surface sterilized with 70% ethanol and rinsed twice with sterile distilled water. Then, the leaves were wounded using a 34-mm-diameter florist pin frog and inoculated by pipetting 20 µl of conidial suspension (1 × 10⁶ conidia/ml) of 7-day-old culture of isolate B onto the wound sites. Control leaves were inoculated with sterile distilled water only. The inoculated plants were covered with plastic bags for 5 days and maintained in a greenhouse at 25°C, 90% relative humidity, with a photoperiod of 12 h. After 7 days, inoculated leaves developed necrotic lesions similar to the symptoms observed in the field, whereas the control treatment remained asymptomatic. The fungus was reisolated from the infected leaves and was morphologically identical to the original isolate. *F. equiseti* was previously reported causing fruit rot of watermelon in Georgia (Li and Ji 2015) and China (Li et al. 2018).

This pathogen could cause serious damage to established rockmelon, because it can spread rapidly in the field. To our knowledge, this is the first report of a member of the FIESC causing leaf spot on *C. melo* in Malaysia.

Keyword: Leaf spot; *Cucumis melo* L.; *Fusarium incarnatum equiesiti*