First report of *Fusarium oxysporum* causing wilt and root rot in common borage (*Borago officinalis*) in Spain

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Common borage (*Borago officinalis* L.) is a horticultural species largely cultivated and consumed in the center of Ebro valley in the Zaragoza province, northeast of Spain. Over the last 10 to 15 years, greenhouse crops from different producers have been affected by disease outbreaks characterized by symptoms such as severe yellowing and wilting. Symptomatic plants' crown tissues and upper roots are covered with reddish-brown to blackish-brown necrosis, which sometimes progresses up to the lower third of the stem including leaf veins, and is followed by plant death. Severely infected plants are stunted and eventually die, resulting in up to 70% crop loss. Small pieces (3-4 mm) of symptomatic tissues from crown and roots were surface disinfected (0.5 min in 60% ethanol and 2 min in 3% NaOCl), washed four times with sterilized distilled water, plated onto potato dextrose agar (PDA) amended with streptomycin sulphate (0.5 g L^{-1}), and incubated at 25°C in the dark for 1-3 days. *Fusarium*-like colonies were consistently isolated and transferred to PDA and Spezieller Nährstoffarmer agar for morphological characterization. Macroconidia were slender, pointed at the apexes, fusoid-subulate or falcate, 3-5 septate in average, 48 (29-53) x 4.1 (3.5-4.5) µm. Aerial microconidia were abundant, borne on short, lateral, narrowly lageniform

monophialides, ovoid, oval-ellipsoid or cylindrical, sometimes allantoid, mostly aseptate 8 (5.2-17) x 3.1 (2.5-3.8) µm. Chlamydospores were globose, mostly intercalary and not in chains. Isolates were tentatively identified as belonging to the *Fusarium oxysporum* Schltdl. species complex (Booth, 1970). The internal transcribed spacer region (ITS) and translation elongation factor-1α (TEF-1α) gene of isolate MYC-1304 were PCR amplified using ITS1/ITS4 (White et al., 1990) and EF1-728F/EF1-986R (Carbone and Kohn, 1999) primers, respectively, sequenced and deposited in GenBank with accession numbers MK290853 (ITS) and MK332141 (TEF-1α). BLASTn analysis of the two sequences showed 99% and 100% homology with those of *F. oxysporum* KU527806 (ITS) and LS479649 (TEF-1α), respectively. Isolate MYC-1304 was grown in 250 ml flasks containing potato sucrose medium for 3 days at 25ºC in the dark with constant agitation. Ten 20-day-old borage seedlings (cv 'Movera') grown in trays with sterilized substrate, were removed, dipped into a suspension of 6 x 10^6 conidia/ml for 2 min, and transferred to plastic pots with sterilized substrate. Three non-inoculated plants dipped in sterile water were used as controls. Plants were incubated in a growth chamber (25ºC; 16/8 h photoperiod). Severe wilting and yellowing accompanied by dry necrosis of the central veins of some leaves, followed by plant death were observed 10 days post-inoculation. Non-inoculated controls remained asymptomatic. The fungus was re-isolated and identified using ITS and TEF-1α sequences from all the inoculated plants. Both *F. oxysporum* and *F. solani* have been cited as a cause of Viper's Bugloss wilt in Iran (Nasr Esfahani and Monazzah, 2011; Okhovvat et al. 2005), in spite that such common name refers to both *B. officinalis* and *Echium vulgare* L. in Europe. Other *Fusarium* species (i.e. *F. avenaceum*) have also been cited from common borage (Mulenko et al., 2008). To our knowledge, this is the first report of *F. oxysporum* causing wilt and root rot of common borage in Europe. The

disease represents a serious threat to this local vegetable and its epidemiology. Seeds and seedlings as well as their production substrate have been hypothesized as possible carriers of infection propagules.

References

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