

## Diseases Caused by Fungi and Fungus-Like Organisms

### First Report of Head Blight of Wheat Caused by *Fusarium vorosii* in Serbia

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The cosmopolitan species *Fusarium graminearum* Schwabe directly reduces yield, as well as grain quality of cereals, due to its ability to synthesize mycotoxins. Previously it was considered to be one species occurring on all continents. However, phylogenetic analysis employing the GCPSR method (genealogical concordance phylogenetic species recognition) revealed the existence of 15 phylogenetic species within what is now recognized as the *Fusarium graminearum* species complex (FGSC) (Sarver et al. 2011). During 1996–2008, a collection of FGSC isolates was established at the Maize Research Institute, Zemun Polje, and isolates originating from wheat (5), maize (3), and barley (2) were selected for further study. Morphological features including the appearance of colonies and macroconidia (average size 38.5 to 53.1 × 4.6 to 5.4 μm,  $n = 50$ ) of all 10 isolates on potato dextrose agar (PDA) were consistent with descriptions of *F. graminearum* (Leslie and Summerell 2006; O'Donnell et al. 2004). Total DNA was isolated from mycelium removed from 7-day-old colonies of single-spore isolates grown on PDA using the DNeasy Plant Mini Kit (Qiagen, Hilden, Germany). Further identification was based on amplification and sequencing of elongation factor *TEF-1α*, histone *H3*, and  $\beta$ -tubulin in both directions, with primers ef1/ef2, H3-1a/H3-1b, and T1/T22, respectively (Jacobs et al. 2010). The sequences were deposited in GenBank under accession numbers MF974399–MF974408 (*TEF-1α*), MG063783–MG063792 ( $\beta$ -tubulin), and MF999139–MF999148 (histone *H3*). Sequence analysis was performed using BLAST while genetic similarity was calculated using MEGA 6.0 software. Isolate 1339 originating from wheat (collected at the

locality of Kikinda in 2006) shared 100% nucleotide identity with *TEF-1α* (DQ459745), histone *H3* (DQ459728), and  $\beta$ -tubulin (DQ459643) of *F. vorosii* isolate NRRL37605 (Starkey et al. 2007). The remaining nine isolates were identified as *F. graminearum* as they shared 99 to 100% nucleotide similarity with *F. graminearum* NRRL 28439 (O'Donnell et al. 2004). Pathogenicity was tested using artificial inoculations of spikes during wheat flowering (Mesterházy et al. 1999). Thirty classes were inoculated with each isolate, in three replicates. Inoculum was prepared from 7-day-old colonies on PDA, and 30 ml of a conidial suspension ( $1 \times 10^5$  conidia/ml) was used. Control plants were inoculated with sterile water. Three weeks after inoculation, typical Fusarium head blight symptoms were visible on inoculated plants, from which all 10 isolates were successfully reisolated. Control spikes remained symptomless. Disease severity was estimated on the 1 to 7 scale (Blandino et al. 2012). Average pathogenicity of the *F. vorosii* isolate 1339 was 1.9, and 2.4–5.1 for *F. graminearum* isolates. Toxin production was determined using gas chromatography-tandem mass spectrometry. Kernels inoculated with the 10 isolates were ground and tested for the presence of deoxynivalenol (DON) and its acetyl derivatives 3ADON, 15ADON, and NIV. *F. vorosii* isolate 1339 possessed the 15ADON chemotype, as well as eight *F. graminearum* isolates, while only one *F. graminearum* isolate was 3ADON chemotype. To date, *F. vorosii* has only been detected in Hungary on wheat (Tóth et al. 2005) and in Korea on barley, corn, and rice (Lee et al. 2016). This is the first report of *F. vorosii* in Serbia, which is of great importance, because it indicates the spread of this toxigenic species. Further studies should be focused on determining the distribution, aggressiveness, and toxicological profile of *F. vorosii*.

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