First Report of the cereal cyst nematode Heterodera filipjevi on winter wheat in Montana.

A. T. Dyer, R. Al-Khafaji and T. Lane, Department of Plant Sciences and Plant Pathology, Montana State University, 119 Plant BioScience Building. Bozeman, MT 59717-3150; T.
Paulitz, USDA-ARS, Pullman, WA 99164 and Z. A. Handoo, A. M. Skantar and D. J.
Chitwood, Nematology Laboratory, USDA, ARS, Beltsville, MD 20705

Among the cereal cyst nematode complex, three species *Heterodera avenae*, *H. filipjevi*, and *H. latipons* are the most destructive to wheat (Smiley and Yan 2015). While *H. avenae* occurs in several U.S. states, H. filipjevi has only been reported from Washington in 2008 (Smiley and Yan 2015). *Heterodera latipons* has not been reported in the U.S. In Chouteau County, Montana, a winter wheat field reported to contain *H. avenae* was observed with areas of heavily stunted winter wheat 'Yellowstone'. Roots from affected plants displayed moderate numbers of nematode cysts. Given symptom severity, H. filipievi was suspected (Golinowski et al. 1997). Living nematode cysts from wheat roots collected from the affected field were examined morphologically and by PCR for species identification. Observations of morphological characters critical for identification indicated that the specimens were H. filipjevi (Akinbade et al. 2010, Subbotin et al. 2010). Measurements of second-stage juveniles (J_2 ; n = 17) included length of body (range = 490 to 615 μ m, mean = 565.1 μ m), stylet (24.0 to 25.0 μ m, 24.3 μ m) with anchor-shaped basal knobs, tail (50.0 to 70.0 µm, 61.4 µm), and hyaline tail terminus (35 to 43 µm, 38.5 µm). The lateral field had four lines, inner two more distinct. Shapes of the tail, tail terminus, and stylet knobs were also consistent with *H. filipjevi*. The lemon-shaped cysts (n = 10) were light brown to yellow and had a zigzag pattern and bifenestrate vulval cone; morphometrics included body length (L) excluding neck (475 to 718 µm, 643.0 µm), body width (W) (336 to 545 μ m, 460.3 μ m), L/W (1.3 to 1.6 μ m, 1.4 μ m), neck length (60 to 100 μ m, 83.5

μm) and width (50 to 75 μm, 59.3 μm), fenestra length (40 to 55 μm, 48.0 μm) and μm width (20 to 30 μm, 24.8 μm), heavy underbridge (60 to 82.5 μm, 72.8 μm), vulval slit (7.5 to 12.5 μm, 9.4 μm), and many bullae. Molecular analysis of internal transcribed spacer (ITS1 and 2) and 28S rRNA confirmed identity as *H. filipjevi* (Akinbade, et al. 2010). Amplicons generated from three J2 (two hatched from one cyst; one from another) were cloned and sequenced (Akinbade et al. 2010). Sequence from the 28S region (GenBank accession KP878489) was >99.9% identical to those of *H. filipjevi* from China (accessions GU083594, GU083592, GU083597) and, distinct from several populations of *H. avenae* (99.0 to 99.2%). The ITS rDNA from this population (KP878490) matched with 99.9% identity to several *H. filipjevi* sequences available from GenBank, including ones from Russia (AF274399), Tajikistan (AY148402), UK (AY148403), USA (GU079654), and China (GU083595). The next closest species match was *H. avenae*, with 96 to 97% identity to the Montana population.

This is the first report of *H. filipjevi* from Montana. Eggs of *H. filipjevi* are noted for hatching in the fall during winter wheat planting and therefore have the potential to affect winter wheat more than *H. avenae*, whose eggs hatch only in the spring after a cold period. Moreover, resistance in wheat against these two nematode species is different and will require additional considerations.

References:

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