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DISEASE NOTES

First Report of the Soybean Cyst Nematode, Heterodera glycines, in New York

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The soybean cyst nematode (SCN), Heterodera glycines Ichinohe, is the most damaging pathogen of soybean (Glycine max [L.] Merr.), causing more than \$1 billion in yield losses annually in the United States (Allen et al. 2017). The SCN distribution map updated in 2014 showed that SCN were detected in all major soybean-producing states in the U.S. except West Virginia and New York (Tylka and Marett 2014). Soybean shows great economic promise in NY and its production area in the region has been expanding rapidly. In coordination with a statewide soybean disease survey, soil samples have been collected from 17 counties throughout NY since 2013 to search for the presence of SCN. A postharvest soil sample collected in fall 2016 from a soybean field in Cayuga County, NY, was processed using the sugar centrifugal-flotation method to extract nematodes and a few brown and lemon-shaped cysts, similar to those of SCN, were isolated. The lemon-

shaped cysts were light to dark brown and had a zigzag pattern and ambifenestrate vulval cone. Morphometrics of cysts (n = 5) included body length (L) including neck (520 to 866 μ m, mean = 696.0 μ m); body width (W) (320 to 495 μ m, 399.8 μ m); L/W (1.4 to 2.1, 1.7 μ m); neck length (60 to 100 μ m, 74.0 μ m) and width (45 to 55 μ m, 50.0 μ m); fenestra length (52 to 65 μ m, 58.4 μ m) and width (32 to 40 μ m, 37.4 μ m); well-developed underbridge (70 to 110 μ m, 80.0 μ m); vulval slit (42 to 52 μ m, 48.2 μ m); and many

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bullae varying from round to finger-like. Second-stage juveniles (J2s) were typical for SCN with lateral field having four equally spaced incisures, a robust stylet with anteriorly protruding knobs, and a tail tapering uniformly to a finely rounded terminus. The key morphometrics of J2s (n = 15) included length of body (range = 381 to 510 µm, mean = 438.5 μ m); stylet (22.5 to 25.0 μ m, 23.8 μ m); tail (42 to 50 μ m, 47.3 μ m), and hyaline tail terminus (22.5 to 27.5 µm, 25. 1 µm). Morphology of the cysts and J2s was in agreement with those of H. glycines (Ichinohe 1952; Subbotin et al. 2010). Genomic DNA was extracted from hatched juveniles (n = 3), and ribosomal DNA of the ITS and 28S

regions were amplified using PCR primers TW81 and AB28, and D2A and D3B (Skantar et al. 2012), respectively. PCR products were cleaned with the Monarch DNA Gel Extraction Kit (NEB, Ipswitch, MA) and cloned into vector pSC-A-amp/kan using the StrataClone PCR Cloning Kit (Agilent, Santa Clara, CA). Plasmid DNA was prepared with the Monarch Plasmid Miniprep Kit (NEB) and sequenced by Macrogen, Inc. All nucleotide sequences (KY795943-45; KY794755-65) were submitted to GenBank. A BLASTN search was performed to verify the identity of the sequences. The three 28S sequences were identical to one another and 100% identical to those of H. glycines from China (GU595446) and 99% identical to several other accessions (e.g., JN684906 and HM560850). The ITS sequences varied between 2 and 6 bp; the consensus sequence from all clones matched a Chinese population sequence (HM560783) at 99% identity. BLASTN of individual ITS sequences matched several other H. glycines sequences in GenBank (e.g., KF745929 and EU106169) at 99% identity. To verify nematode viability, the rest of the nematode sample that contains ~750 eggs and juveniles was inoculated on a soybean plant (cv. Williams 82) and more than 4,800 nematode females and cysts were recovered from the inoculated plant after 65 days of growth in a growth chamber maintained at 25°C with 16-h light/day. These morphological and molecular analyses as well as the bioassay test confirmed the identity of the nematode species as H. glycines. To our knowledge, this is the first report of



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