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plant disease

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[Home](#) > [Plant Disease](#) > [Table of Contents](#) > [Full Text HTML](#)

[Previous Article](#) | [Next Article](#)

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Page 2662

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DISEASE NOTES

First Report of *Aphelenchoides besseyi* Infecting the Aerial Part of Cotton Plants in Brazil

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Cotton plants (*Gossypium hirsutum* L.) exhibiting stunting, loss of floral buds, foliage distortion, and thickened nodes were observed in May 2017 in the county of Sapezal, Mato Grosso State, Brazil (13°32'33"S; 58°48'51"W). Samples of leaves and stems in 10-g aliquots were processed using the blender-centrifugal flotation method. Nematodes of the genus *Aphelenchoides* were extracted at fresh tissue densities of 41 specimens/g. Specimens were collected individually, narcotized in distilled water with light heat, and mounted in temporary slides. Males and females were examined and measured using a compound microscope (Olympus BX 53) coupled with a video image system. Morphological analysis of females indicated a slender body, a labial region slightly wider than the first body annulus, a hexa-radiated labial structure, a stylet 11 µm long, an oval metacarpus with a distinct valve, the excretory pore located at the anterior edge of the nerve ring, the lateral field marked by four incisures, an oval and long spermatheca filled with spermatozoa, a narrow postuterine sac without spermatozoa and shorter than 1/3 of the vulva-anus distance, and a conoid tail with a terminus bearing a mucro with four pointed processes. These morphological characters and those of the males matched those reported in the original description of *Aphelenchoides besseyi* by Christie (1942) and a revision by Fortuner (1970) and Franklin and Siddiqi (1972). Additional individual *A. besseyi* specimens from cotton were used for phylogenetic analysis to verify genetic congruence between the sequences of this population of *A. besseyi* from cotton and those published for other populations of this nematode in Brazil. The genomic DNA extraction and the sequences were obtained for the near full length 225 of the ribosomal small subunit (SSU) and the expansion segment 226 of the large subunit (D2-D3 LSU) according to the protocol of de Jesus et al. (2016). The newly obtained sequences (MH187564 and MH187565) were assembled using BioEdit version 7.2.5 (<http://www.mbio.ncsu.edu>

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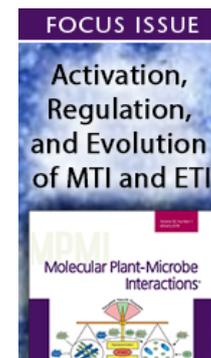
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[/bioedit/page2.html](#)). Guide sequences to phylogenetic analysis were from [Meyer et al. \(2017\)](#). Sequence data were aligned using Mega 7.0 (ClustalW algorithm). The ambiguously aligned regions from SSU and LSU alignments were removed to optimize the phylogenetic analysis. For phylogenetic reconstruction, the best fitting model of sequence evolution was determined for each data set separately using the Akaike information criterion in Mega 7.0. The model GTR+G+I (general time reversible + gamma distributed with invariant sites) was selected for both SSU and LSU. Therefore, phylogenetic trees were constructed applying the maximum likelihood method with 100 bootstrap replications. Phylogenetic analysis indicated that the sequence of the cotton population of *A. besseyi* grouped with those of a population infesting soybean ([Favoreto and Meyer 2017](#); [Meyer et al. 2017](#)) and other populations included in the group rice by [de Jesus et al. \(2016\)](#). The ability of *A. besseyi* to infect cotton was confirmed in two greenhouse experiments. For these experiments, specimens of *A. besseyi* obtained from field-infected cotton plants were reared on fungal cultures of *Fusarium* sp. growing in potato dextrose agar in Petri dishes. The reared nematodes were injected at the stem base of individual healthy cotton cultivar TMG 47B2RF seedlings growing in pots at a density of 6,000 specimens per seedling. The same field symptoms were observed in the inoculated seedlings, and a high number of nematodes were recovered. These results fulfill the modified Koch's postulates. The final nematode densities in the two experiments were 970 and 2,244 specimens per gram of fresh tissue. Field observations in Brazil also indicated that soybeans grown after or before cotton in the same nematode-infested area were damaged by the nematode as well (similar symptoms on soybean aerial tissues with population levels of 70 specimens/g). *A. besseyi* therefore poses a considerable threat to cotton-soybean cropping systems in Mato Grosso State in Brazil.

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Section:

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