Phylogenetic analysis of Longidorus and Xiphinema species (Nematoda: Longidoridae) using ITS1 sequences of nuclear ribosomal DNA

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Abstract

Members of Longidoridae in the genera Longidorus and Xiphinema are important plant parasites as well as vectors for plant nematode species. Discrimination based entirely on morphology is often questionable and controversial because of the morphometric overlap and the lack of a phylogenetic framework for this group. Phylogenetic analysis using the DNA sequences of nuclear ribosomal DNA ITS1 was conducted to construct an evolutionary tree. Extant interspecies sequence variation and minor intraspecies sequence variation were observed. The aligned sequences of the ITS1 region including the 5' end of the 5.8S DNA gene, 5' end of the 18S DNA gene from each species ranged from 870 bp to 1354 bp for all longidorids. This fragment is highly variable; only 15.6% sequence was constant within the longidorids examined. The analyses enabled two groups within Arkansas Longidoridae species: (1) Xiphinema group including X. americanum Cobb, 1913, X. auditor Williams, 1984, X. elongatus Thorne, 1939, and Longidorus diadecturus Erichson & Allen, 1882. (2) Longidorus group including L. brevicaudatus Notton & Hoffman, 1975, L. antennatus Thorne, 1974, L. fragilis Thorne, 1974, and others. Described species and one non-Arkanas species L. elongatus (De Man, 1876) Thorne & A. Strong, 1976 from Scotland. The phylogenetic analysis largely corresponded to the genera and species defined by morphology. Results demonstrated the potential for the use of DNA ITS1 to infer phylogenetic relationships and PCR-RFLP-based identification among longidorids.

Materials

Table 1. Longidorus and Xiphinema populations and species used in ITS1 DNA sequencing.

Table 2. Statistics of nucleotides of ITS1 DNA sequences compared among species of Longidorus and Xiphinema.

Results


Fig. 2. Blue-white screening on ITS1 clones hosted in competent E.coli JM109 on selective and differentiative agar (L.magnus/Gal3-gal gene plasmid to identify nucleotide sequence).

Fig. 3. Best maximum likelihood tree for ITS1 of Longidorus and Xiphinema

Fig. 4. Consensus tree from the maximum parsimony bootstrapping analysis for ITS1 of Longidorus and Xiphinema populations. Bootstrapping and bootstrap values supporting the clades are indicated at the branch points.

Fig. 5. Alignment of nucleotide bases of ITS1 DNA nucleotide sequence for Longidorus and Xiphinema (primers in bold, 5' - 3' 25.6 kb sequence unedited).