USING RIBOSOMAL RNA TECHNOLOGY FOR CLASSIFYING MICROSPORIDIA

C. R. Vossbrinck, M. Baker, J. Maddox, M. R. Jeffords, and B. A. Debrunner-Vossbrinck
Illinois Natural History Survey, Champaign, Illinois 61820

ABSTRACT

The microsporidia are an obligately parasitic group of protists with a number of unusual cytological and molecular characteristics. They have no mitochondria, their nuclear division is primitive, their ribosomes and ribosomal RNA's are reportedly of prokaryotic size, and their large ribosomal subunit contains no 5.8S rRNA. There are about 87 microsporidian genera that are based primarily on spore shape, numbers of nuclei in the spore, nature of the sporophorous vesicle, and several ultrastructural characteristics of the spores and vegetative stages. Because they have few other morphological characters it is difficult to develop phylogenetic relationships and identify specific isolates of microsporidia.

Molecular sequencing methods have been useful in establishing the phylogenetic relationships of higher taxa, including Microsporida. These same sequencing methods are currently being used to develop a phylogeny of microsporidian genera and to identify specific microsporidian species. Portions of the 23S and 16S rRNA have been sequenced for all five species of microsporidia isolated from the gypsy moth.

The sequences of the gypsy moth microsporidia were compared and a phylogenic tree developed by the use of parsimony analysis. Based on these results, we can distinguish all of the five species of gypsy moth microsporidia and we believe that rRNA sequences can be used for identifying most other species of microsporidia. The use of rRNA has great promise in the development of probes for species specific identifications because of the high copy number of ribosomes in cells.

We currently need relatively large quantities of pure microsporidian spores in order to obtain rRNA sequences, but the use of newly developed techniques (PCR) to amplify specific rRNA genes or pieces of DNA should greatly reduce the quantify of spores needed for restriction enzyme analysis.