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Development and Pathology of Two Undescribed Species of Microsporidia Infecting the Predatory Mite, *Phytoseiulus persimilis* Athias-Henriot

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Two undescribed species of microsporidia were found in mass-reared *Phytoseiulus persimilis* Athias-Henriot from two commercial sources during a routine examination of these predators for pathogens. Both microsporidian species were described from specimens that had been prepared for transmission electron microscopy; live specimens were unavailable for examination. One microsporidium, identified as Species A, was described from two specimens obtained from a commercial insectary in North America. All observed stages of this microsporidium were uninucleate. Rounded-to-ovoid schizonts appeared to develop in direct contact with the cytoplasm of lyrate organ cells (ovarian tissue). Mature spores of Species A were elongate-ovoid and measured 2.88 × 1.21 μm. A polar filament coiled 7 to 10 times in the posterior half of the spore. Sporoblasts and spores were observed in the cytoplasm of cells of numerous tissues and in developing eggs within gravid females. A second species, identified as Species B, was described from five specimens obtained from a commercial source in Israel. All observed stages of this microsporidium were uninucleate. Schizonts of Species B were observed within the cytoplasm of cecal wall cells and within the nuclei of lyrate organ cells. Mature spores were ovoid and measured 2.65 × 1.21 μm. A polar filament coiled 3 to 4 times in the posterior half of the spore. Densely packed ribosomes often concealed the polar filament and other internal spore characteristics. Spores were observed in the cytoplasm of cells of numerous tissues and occasionally within the nuclei of lyrate organ cells. Numerous spores and presporal stages were observed within the ovary and developing eggs. The development and pathology of Species A and B were compared to those of *Microsporidium phytoseiuli* Bjørnson, Steiner and Keddie, a microsporidium previously described from *P. persimilis* obtained from a commercial source in Europe. The occurrence of three species of microsporidia within *P. persimilis* from three sources raises questions regarding the origin of these pathogens. Because microsporidia may have profound impact on the performance of *P. persimilis,* consideration must be given to the identification and exclusion of microsporidia from field-collected specimens or from predators that may be shared among commercial sources.

Key Words: **Acari; *Phytoseiulus persimilis;* biological control; microsporidia; quality control.**

INTRODUCTION

Microsporidia are considered to be the most important protozoan pathogens of insects (Tanada and Kaya, 1993). The majority of these spore-forming protozoa have been described from arthropod hosts that are agricultural pests; however, microsporidia are also known to infect beneficial arthropods, including natural enemies that are mass-produced and used for biological control (Beerling and van der Geest, 1991; Bjørnson *et al.*, 1996). Because microsporidia are known to produce subtle, yet often profound effects on host performance, the presence of microsporidia in mass-produced natural enemies is of considerable interest.

The majority of microsporidia reported from mass-produced natural enemies have been detected in predatory mites. In one study, microsporidian spores of three sizes were reported in *Amblyseius cucumeris* Oudemans and *A. barkeri* Hughes, predatory mites used for control of western flower thrips (*Frankliniella occidentalis* Pergande) and onion thrips (*Thrips tabaci* Lindeman), respectively (Beerling and van der Geest, 1991). It is unclear whether these spores represent three different species of microsporidia or one microsporidium with three spore types. In a separate study,

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microsporidia were reported in *A. cucumeris* and *Phytoseiulus persimilis* Athias-Henriot, a predatory mite used for spider mite control in commercial greenhouses and on field crops (Steiner, 1993). The microsporidium *Microsporidium phytoseiuli* Bjørnson, Steiner and Keddie was described from *P. persimilis* in a subsequent study (Bjørnson *et al.*, 1996). Furthermore, microsporidia have been detected in *Acarus siro* L. and *Tyrophagus putrescentiae* Schrank (Beerling *et al.*, 1993; Larsson *et al.*, 1997), two forage mites used as food sources in commercial mass-rearings of *A. cucumeris* and *A. barkeri*. Although *A. siro* and *T. putrescentiae* are not themselves considered to be beneficial arthropods, it is not known whether microsporidia in these mites contribute toward a decline in the productivity of mass-reared predatory mites.

Based on differences in development and spore morphology, two undescribed species of microsporidia were identified in *P. persimilis* from two commercial sources. Species A was found in *P. persimilis* obtained from a commercial source in North America. Species B was found in predators from Israel. The aim of this study was to document the occurrence of these two undescribed species of microsporidia and to describe the differences among life stages, spore morphology, and pathology. The development and pathology of these two microsporidia are compared to those of *M. phytoseiuli*, a microsporidium described from *P. persimilis* obtained from a commercial source in Europe (Bjørnson *et al.*, 1996).

MATERIALS AND METHODS

P. persimilis were obtained from commercial suppliers in North America and Israel in November 1990 and July 1991, respectively. Smear preparations were made from 25 specimens from each source. These were stained in 15% buffered Giemsa and examined by light microscopy. Whole mites (n = 25 from each source) were embedded for transmission electron microscopy (TEM). Four specimens from North America and 19 from Israel were embedded. Of these, 2 specimens from North America and 6 specimens from Israel were examined.

Adult female *P. persimilis* were placed in a fixative consisting of 1% paraformaldehyde and 1.5% glutaraldehyde in cacodylate buffer (pH 7.4) for 24 to 48 h. The fixative was replaced with 0.12 M cacodylate buffer (pH 7.2) with three changes in 60 min. Tissue was postfixed for 2 h in 1% osmium tetroxide in 0.12 M cacodylate buffer.

Following fixation, mites were placed in distilled water for 10 min and dehydrated in the following ethanol series: 50% (30 min), 70% (30 min), 90% (30 min), 100% (60 min), followed by propylene oxide:absolute ethanol (1:1) (30 min), propylene oxide (60 min), propylene oxide:low-viscosity Spurr resin (16 h), and low-viscosity Spurr resin (24 h). Each solution was changed three times. Each mite was placed in Spurr resin within a flat mold and cured for a minimum of 16 h in a 60°C oven.

Ultrathin gold sections, approximately 100 nm thick, were cut with a diamond knife using an LKB Nova Ultramicrotome. Sections were placed on Formvar-coated grids and stained with prefiltered, 4% uranyl acetate for 20 min in a 60°C oven and then with lead citrate for 6 min at room temperature. Sections were examined with an Hitachi H-600 transmission electron microscope (acceleration voltage, 75 kV).

RESULTS

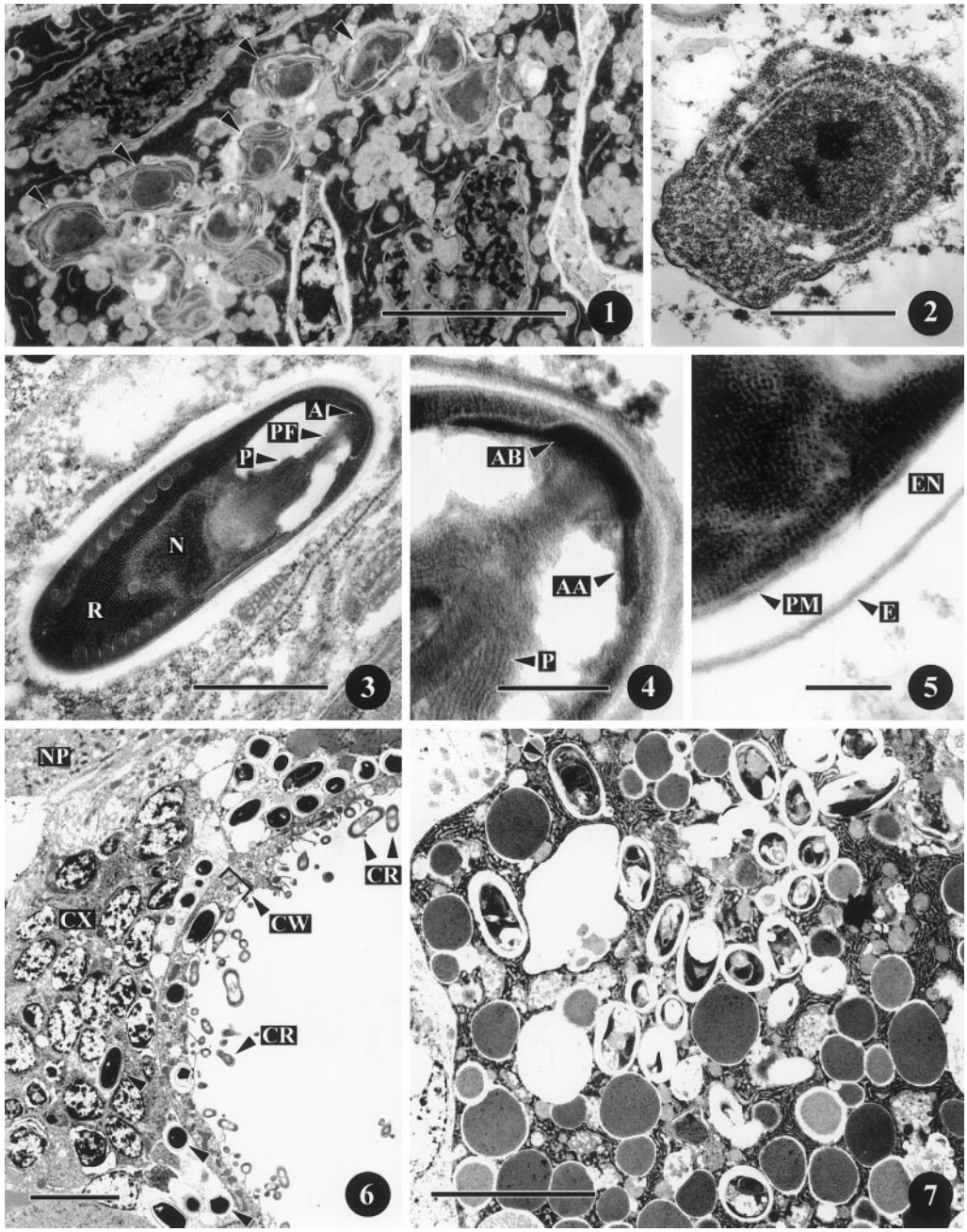
Two unidentified species of microsporidia were found within *P. persimilis* from a North American and an Israeli source and are designated as Species A and Species B, respectively. Spores were detected in smear preparations; however, developmental stages were not observed by light microscopy. All measurements and life cycle observations were made exclusively from transmission electron micrographs on a small series of observations. It is difficult to make accurate evaluations of spore sizes based on ultrathin sections since the image, and consequently the size, are influenced by the plane of sectioning.

Species A (North American Source)

Presporal stages. Both specimens examined by TEM were infected with microsporidia. All observed developmental stages were uninucleate. Rounded-to-ovoid schizonts measured 2.44 ± 0.19 × 1.64 ± 0.06 μm (n = 13). Schizonts appeared to undergo cellular division to become groups of five, six, or more cells. Schizonts were observed exclusively within cells of the lyrate organ (ovarian tissue) where they appeared to develop in direct contact with host cell cytoplasm (Fig. 1). Sporonts could not be distinguished from schizonts.

Individual cells described here as early sporoblasts measured 2.73 ± 0.48 × 1.49 ± 0.15 μm (n = 4). These cells often contained darkened nucleoli and were distinguishable by their electron-dense cell membrane and loosely packed ribosomes (Fig. 2). Sporoblasts were proximal to mature spores. One sporoblast showing polar filament development was observed within the nucleus of an unidentified cell type.

The mature spore. Mature spores were elongate-ovoid (Fig. 3) and measured 2.88 ± 0.14 × 1.21 ± 0.03 μm (n = 11). A polar filament coiled 7 to 10 times in the posterior half of the spore. In some cases, the final one to two coils appeared attenuated. The anchoring disc base was broad and flat and measured 277 nm, with stout arms each measuring 188 to 222 nm in length (Fig. 4). A lamellar polaroplast occupied approximately



FIGS. 1–7. The microsporidium found infecting *Phytoseiulus persimilis* from North America (Species A). Fig. 1. Several schizonts (arrowheads) within cytoplasm of a single cell (lyrate organ). Fig. 2. Early sporoblast. Fig. 3. Mature spore. Fig. 4. Anchoring disc detail. Fig. 5. Spore wall. Fig. 6. Sporoblast and spores within cells lining the cecal lumen and adjacent cortex of supraesophageal ganglion (arrowheads). Fig. 7. Sporoblast (arrowhead) and spores in eggs within gravid females. Scale bars: Figs. 1, 6, and 7, 5 μm; Figs. 2 and 3, 1 μm; Figs. 4 and 5, 0.25 μm. AA, arms of anchoring disc; AB, base of anchoring disc; A, anchoring disc; CR, crystal; CW, cecal wall; CX, cortex (synganglion); DE, developing egg; E, exospore; EN, endospore, MT, Malpighian tubule; N, monokaryon; NP, neuropile (synganglion); OV, ovarian tissue; P, polaroplast; PF, polar filament; PM, plasma membrane; PV, posterior vacuole; R, ribosomes.

one third of the anterior region of the mature spore. Large cavities in the polaroplast are artifacts probably resulting from incomplete fixation. Spores were densely packed with ribosomes and although posterior vacuoles were not commonly observed, when present they occupied one third of the spore.

Spore wall thickness was relatively uniform except for a noticeable thinning at the anterior end. Measurements from micrographs showed them to be 78 to 200 nm thick and to consist of three layers: an innermost plasma membrane, a translucent endospore layer measuring 44 to 167 nm, and an electron-dense exospore

measuring 16 to 33 nm (Fig. 5). Spores did not appear to be confined by an interfacial envelope.

Pathology. Sporoblasts were proximal to mature spores; neither were restricted to any given tissue. Infected cells lining the cecal lumen (Fig. 6) were often hypertrophied. Both sporoblasts and spores were observed within the cortex of the supra- and suboesophageal ganglia but only rarely in the neuropile. Spores were also observed within the cytoplasm of cells underlying the cuticle, within muscle fibers, and in unidentified leg tissues. Sporoblasts and spores were not observed within lyrate organ cells but were observed in developing eggs within gravid females (Fig. 7).

Species B (Israeli Source)

Five of the six specimens examined were infected with microsporidia. Two of these specimens were also infected with rickettsia (within unidentified tissues) (not illustrated; see Bjørnson *et al.*, 1997).

Presporal stages. All observed stages of this microsporidium were uninucleate. Schizonts of Species B were generally more elongate than those of Species A and measured 2.53 ± 0.23 μm × 1.58 ± 0.10 μm (n = 14). Variable numbers of schizonts were located within the nuclei of lyrate organ cells and within the cytoplasm of cells lining the cecal lumen. Schizonts were observed within lightened areas of the host cell nucleus, suggesting that they may be grouped together, although there was no evidence of interfacial envelopes (Fig. 8). Sporonts could not be distinguished from schizonts.

Stages herein described as early sporoblasts contained a darkened nucleolus and were distinguished from schizonts by a more electron-dense membrane, a thickened plasmalemma, and loosely packed ribosomes (Fig. 9). Early sporoblasts measured 2.06 ± 0.19 × 1.41 ± 0.09 μm (n = 11). These were observed in unidentified cells adjacent to cells of the lyrate organ, within cecal wall cells, and within groups of spores in unidentified tissue. Developing sporoblasts measured 2.88 ± 0.25 × 1.12 ± 0.03 μm (n = 5). Each contained an immature polar filament and a prominent Golgi apparatus that was less conspicuous in older sporoblasts. Developing sporoblasts were observed exclusively within the cytoplasm of cells lining the cecal lumen, proximal to mature spores.

The mature spore. Mature spores were ovoid (Fig. 10) and measured 2.65 ± 0.23 × 1.21 ± 0.07 μm (n = 5). Although an isofilar polar filament coiled three to four times in the posterior half of the spore, densely packed ribosomes often concealed the polar filament and other internal spore characteristics. Features were more conspicuous in sporoblasts. The base of the anchoring disc was broad and measured 266 nm, with thin, short arms each measuring 177 to 222 nm in length (Fig. 11). A lamellar polaroplast occupied one third of the anterior region of the mature spore. Large cavities in the polaroplast are artifacts, probably resulting from inadequate fixation. Posterior vacuoles occupied one third of the spore.

Spore walls were relatively uniform in thickness but thinner at the anterior end. Measurements from micrographs showed them to be 55–64 nm thick and to consist of three layers: the innermost plasma membrane, a translucent endospore layer measuring 36–45 nm, and an electron-dense exospore measuring 9–18 nm (Fig. 12).

Occasionally, host cells became packed with sporoblasts and spores. Groups of spores were observed within a dark matrix of unknown origin surrounded by a membrane. Schizonts and spores occur together (Fig. 8), which indicates that the interfacial envelope probably is a sporophorous vesicle. Individual spores and groups of spores were observed within many tissues, including the nuclei of lyrate organ cells (Fig. 8, arrow), in both the nuclei and the cytoplasm of cells lining the cecal lumen, in the supra- and suboesophageal ganglia, in cells underlying the cuticle, and within cells lining the Malpighian tubules. Spores were observed within the ovary and eggs of gravid females.

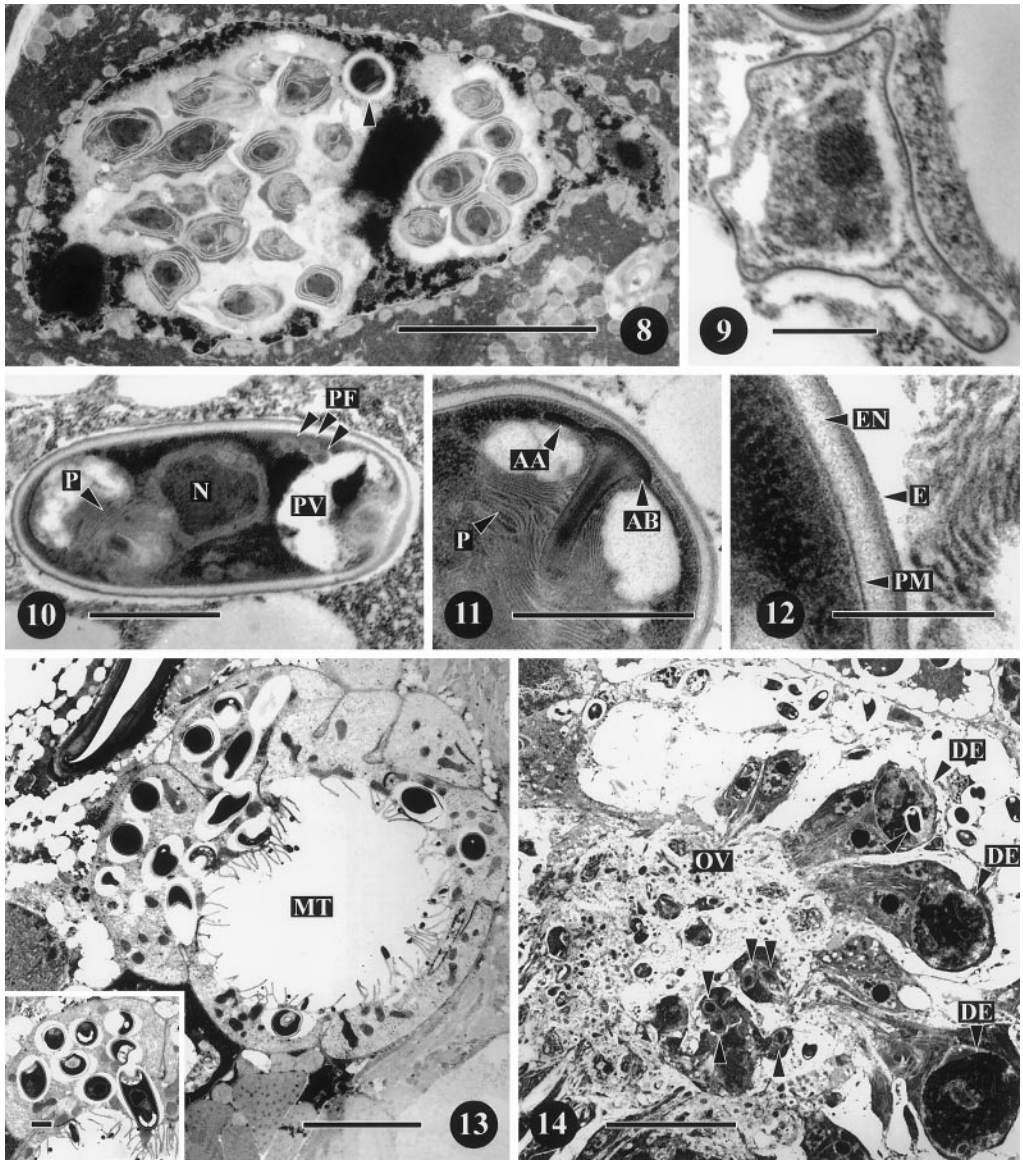
Pathology. Nuclei of lyrate organ cells containing few to numerous schizonts were greatly hypertrophied (Fig. 8). Mature spores were observed in the cecal lumen. Schizonts, sporoblasts, and spores were observed within the cytoplasm of cells underlying the cecal lumen and in cells lining the Malpighian tubules (Fig. 13). These infected cells were often hypertrophied. Numerous spores and presporal stages were observed within developing eggs and ovarian tissue (Fig. 14, arrows).

DISCUSSION

This study presents some fundamental but limited information on the morphological, developmental, and pathological characteristics of two previously undescribed microsporidia in *P. persimilis.* Because the information provided is currently insufficient for the description of two new species, these microsporidia were not assigned to a particular genus or given a specific name.

Presporal Stages

All stages of Species A and Species B were uninucleate. A summary of presporal stages and mature spore characteristics for microsporidia found in *P. persimilis* is provided in Table 1. Schizonts of all three microsporidian species were observed within cells of the lyrate organ and few to several shizonts were observed within an individual host cell. Round to ovoid schizonts of



FIGS. 8–14. The microsporidium found infecting *Phytoseiulus persimilis* from Israel (Species B). Fig. 8. Numerous schizonts and mature spore (arrowhead) within cell nucleus (lyrate organ). Fig. 9. Early sporoblast. Fig. 10. Mature spore. Fig. 11. Anchoring disc and polaroplast detail. Fig. 12. Spore wall. Fig. 13. Infected cells of Malpighian tubules causes cell hypertrophy. Inset: Spores within infected cells appear to be sloughed into the Malpighian tubules. Fig. 14. Sporoblasts (lower center) and numerous spores within ovarian tissue and young, developing egg (arrowhead, right). Scale bars: Figs. 8 and 13, 5 μm; Fig. 9, 0.5 μm; Figs. 10, 11, and 13 inset, 1 μm; Fig. 12, 0.25 μm; Fig. 14, 10 μm. See legend to Figs. 8–14 for abbreviations.

Species A (North America) developed within plasmodia-like groups. These were observed almost exclusively within the cell cytoplasm. Schizonts of Species B (Israel) appeared independent of one another and were more elongate than those of Species A. Schizonts of species B were observed most frequently in the nuclei of lyrate organ cells. In contrast, round to slightly ovoid schizonts of *M. phytoseiuli* (Europe) were observed within the nuclei of lyrate organ cells, typically within chromatin-rich areas adjacent to the nuclear membrane (Bjørnson *et al.*, 1996). These cells were previously misidentified as digestive cells by the authors.

Only one sporoblast was observed for Species A. Those of species B and *M. phytoseiuli* were not restricted to specific tissues. Sporoblasts of *M. phytoseiuli* had a distinctive caudal prolongation extending from the posterior end (Bjørnson *et al.*, 1996); this structure was not observed in sporoblasts of Species A or B.

Mature Spores

Spores of Species A were slightly larger than those of Species B. Similarities in spore size may pose difficul-

TABLE 1
Summary of Presporal Stages and Mature Spore Characteristics of Microsporidia Found in *Phytoseiulus persimilis* from Three Commercial Sources

	Unidentified Species A (North America) $n = 2$	Unidentified Species B (Israel) $n = 6$	*Microsporidium phytoseiuli* (Europe) $n = 10$
Presporal stages			
Schizonts	Single or groups of 5 or more	Variable numbers of schizonts	Single or groups of 2 or more
Schizont mean ± SE (μm)	2.44 ± 0.19 × 1.64 ± 0.06 ($n = 13$)	2.53 ± 0.23 × 1.58 ± 0.10 ($n = 14$)	1.9 ± 0.13 to 2.9 ± 0.23 ($n = 10$)
Sporoblasts	One observed in nucleus of unknown cell type	Numerous; in cytoplasm of cecal wall cells	Common; caudal prolongation; in nuclei of lyrate organ cells and cytoplasm of other infected cells
Sporoblast mean ± SE (μm)	None available	2.88 ± 0.25 × 1.12 ± 0.03 ($n = 5$)	Up to 5.6 μm long; measurement similar size as spores ($n = 8$)
Mature spores			
Measurement mean ± SE (μm)	2.88 ± 0.14 × 1.21 ± 0.03 ($n = 11$)	2.65 ± 0.23 × 1.21 ± 0.07 ($n = 5$)	4.33 ± 0.35 × 1.27 ± 0.15 ($n = 13$)
Polar filament	Isofilar; in some cases, the final 1 to 2 coils appeared attenuated	Isofilar; polar filament often concealed by densely packed ribosomes	Isofilar
No. of polar filament coils	7 to 10; coiled in posterior $\frac{1}{2}$ of spore	2 to 4; coiled in posterior $\frac{1}{2}$ of spore	12 to 15; coiled in posterior $\frac{2}{3}$ of spore
Polaroplast	Lamellar; occupied $\frac{1}{3}$ anterior region	Lamellar; occupied $\frac{1}{3}$ anterior region	Lamellar; occupied $\frac{1}{3}$ anterior region
Anchoring disc base/arms (nm)	277/188–222	266/177–222	150/250
Spore wall (nm)	78–200	55–64	89–101
Endospore/exospore (nm)	44–167/16–33	36–45/9–18	54–65/30–36
Posterior vacuole	Occupied $\frac{1}{3}$ of spore if present (uncommon)	Occupied $\frac{1}{3}$ of spore	Occupied $\frac{1}{4}$ of spore
Interfacial envelopes	None observed	Spores occasionally surrounded by membrane of unknown origin	Present; spores in groups of 4, 8, 16 or more

ties in distinguishing Species A and Species B by light microscopy. In contrast, spores of these two species were considerably smaller than those of *M. phytoseiuli* (Bjørnson *et al.*, 1996).

Pathology

A summary of pathology caused by microsporidia in *P. persimilis* is provided in Table 2. In all three cases of microsporidiosis in *P. persimilis,* damage to the cecal wall was extensive. Infected cecal wall cells hypertrophied and often lacked microvilli. This damage suggested that mature spores were released into the cecal lumen and may be subsequently excreted with feces onto foliage where they could provide an inoculum for horizontal transmission to occur. In the case of *M. phytoseiuli,* spores were observed within the cecal lumen of infected *P. persimilis* females (see Bjørnson *et al.*, 1996) and, although horizontal transmission of *M. phytoseiuli* was observed in the laboratory, the mechanism of transmission was not established (Bjørnson, 1998). Infection of cells lining the Malpighian tubules (Species B) and severe cell damage observed with mature spores also suggests that spores are excreted. Excretion of spores, however, does not ensure that horizontal transmission will readily occur, as illustrated by *M. phytoseiuli* in the laboratory (Bjørnson, 1998).

Spores and developing stages of all three microsporidia were found in most *P. persimilis* tissues. These spores are not available for horizontal transmission to new hosts unless they are liberated into the environment during decomposition of the host body following death, or when infected hosts are cannibalized.

In all three cases, the presence of numerous schizonts within cells of the lyrate organ suggests that microsporidian development begins in these cells. Early stages of all three microsporidia reported in *P. persimilis* appeared to occupy distinct sites within lyrate organ cells. Schizonts of Species A were restricted to the cell cytoplasm, whereas those of Species B and *M. phytoseiuli* were primarily located within cell nuclei (Bjørnson *et al.*, 1996).

In *P. persimilis,* the lyrate organ is one of the largest organs observed; the densely packed and clearly defined cells stain deeply, have well-defined nuclei, and lack any intercellular space or tissue. These cells are similar in appearance to the cells of the lyrate organ of

TABLE 2
Summary of Pathology Caused by Microsporidia in *Phytoseiulus persimilis* from Three Commercial Sources

	Unidentified Species A (North America)	Unidentified Species B (Israel)	*Microsporidium phytoseiuli* (Europe)
Tissue tropism			
Cecal wall cells	Sporoblasts, spores in cell cytoplasm	Schizonts, sporoblasts and spores within cytoplasm; spores in nuclei	Schizonts, sporoblasts and spores in cytoplasm
Supra- and suboesophageal ganglia	Spores rarely in neuropile; spores and sporoblasts in cortex	Spores in cortex only	Spores in cortex only
Cells underlying cuticle	Sporoblasts, spores in cytoplasm	Schizonts, spores in cytoplasm	Spores in cytoplasm
Muscle fibers	Spores in cytoplasm (within fibers)	Tissue not observed	Spores in cytoplasm; schizonts in cytoplasm of muscle underlying cecal wall
Cells lining Malpighian tubules	Tissue not observed	Spores in cytoplasm	Tissue not observed
Lyrate organ	Few to several schizonts within cell cytoplasm	Few to numerous schizonts primarily within lightened areas of cell nuclei and occasionally cell cytoplasm; spores in cell nuclei	Few to several schizonts within chromatin-rich area of cell nuclei (adjacent to nuclear membrane); schizonts in cell cytoplasm; spores and sporoblasts in cell cytoplasm; spores in cell nuclei
Ovarian tissue	Tissue not observed	Schizonts, sporoblasts and spores	Tissue not observed
Eggs[a]	Sporoblasts, spores	Schizonts, spores	Spores

[a] Eggs observed within gravid females.

other mesostigmatid mites (Michael, 1892; Jakeman, 1961) and are referred to, more specifically, as nutrimentary tissue (Alberti, 1989). Michael (1892) suggested that the lyrate organ could function as either a vitelligenic or, possibly, a germiniferous organ. Similarly, Alberti (1989) speculated that the likely role of nutrimentary tissue of the lyrate organ is to accelerate oogenesis and/or embryogenesis. In all three cases, presporal stages were observed within lyrate organ cells and spores were observed in developing eggs. Infection of the lyrate organ may be essential for vertical transmission of these pathogens.

In our study, spores of Species A and B were similar in size and shape, but ultrastructurally distinct. There was no evidence of ultrastructural differences among spores of the same species. Spores of Species A were found only within *P. persimilis* from North America, whereas spores of Species B were found exclusively within predators from Israel. Spores of *M. phytoseiuli* were much larger than those of Species A or B and were observed exclusively in predators from a European source.

Sporoblasts of *M. phytoseiuli* had a unique caudal prolongation not observed in sporoblasts of the other two species. Furthermore, schizonts of all three species appeared to have a site preference for development within cells of the lyrate organ (Table 2). Based on spore ultrastructure, pathological observations, and the exclusive origin of each microsporidium observed, these likely represent three distinct microsporidian species rather than the spore dimorphism of a single species.

P. persimilis and Microsporidia

The predatory mite *P. persimilis* was first discovered in 1960 when it was inadvertently imported from Chile to Germany in a shipment of orchids (Hussey, 1985). *P. persimilis* has since been mass-produced and distributed throughout the world for release on a variety of greenhouse and field crops. The discovery of three microsporidian species in *P. persimilis* almost 40 years following the initial discovery of this predator raises questions regarding the source of these pathogens. Since distinct microsporidian species were observed in *P. persimilis* obtained from three different locations, it seems unlikely that these pathogens had infected the original stock of *P. persimilis* and were then distributed to different areas of the world. Even if this were the case, subsequent predator collection from the field or restocking of predators from existing commercial or research sources would likely contribute toward the distribution or redistribution of these pathogens. Because microsporidia may have profound impact on the performance of *P. persimilis* (Bjørnson and Keddie, 1999), consideration must be given to the identification and exclusion of microsporidia from field-collected

specimens or from predators that may be shared among commercial sources.

Although the sources of the microsporidia in *P. persimilis* are unknown, it seems a reasonable hypothesis that these microsporidia are endemic to each region in which *P. persimilis* are mass-produced. *M. phytoseiuli* were not detected in laboratory-reared *Tetranychus urticae* that was used as food in a previous study (Bjørnson *et al.*, 1996); however, *T. urticae* from the commercial sources were not available for examination. It is possible that these microsporidia originated from infected *T. urticae* that were used as food for the mass-production of *P. persimilis*, or from another endemic acarine species.

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