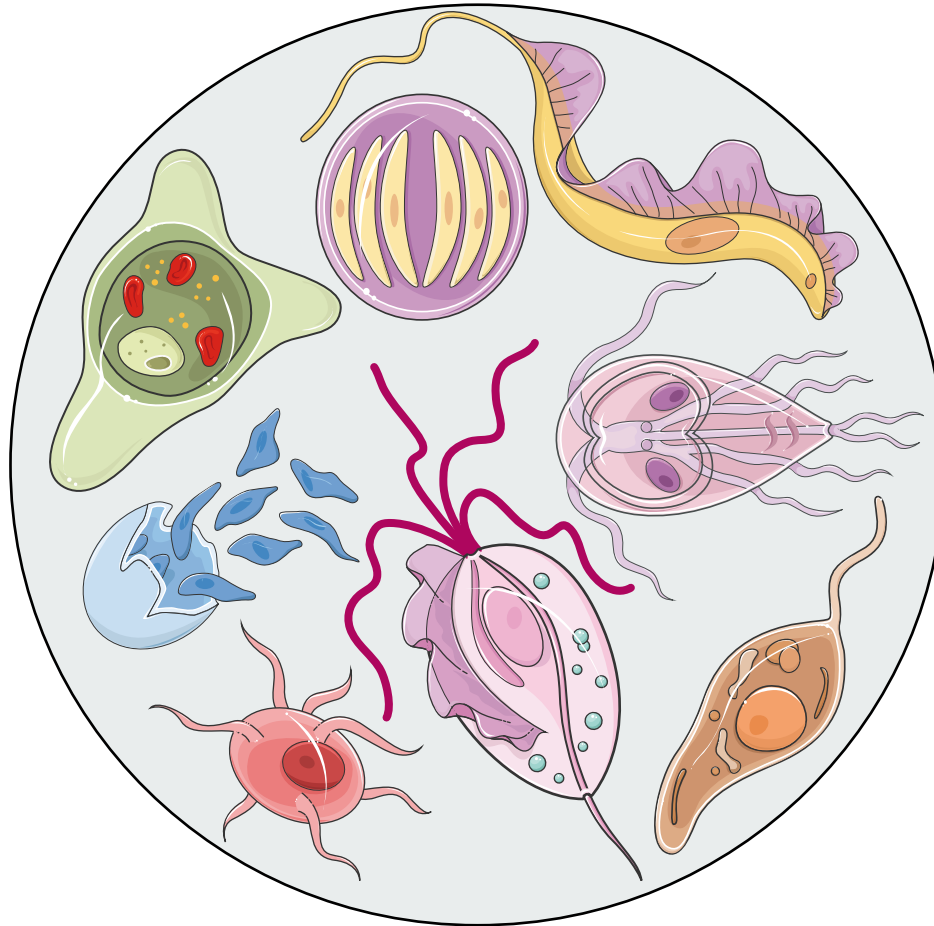


# Biology of parasitic protozoa

## IX. Microsporidia (Opisthokonta, Fungi)



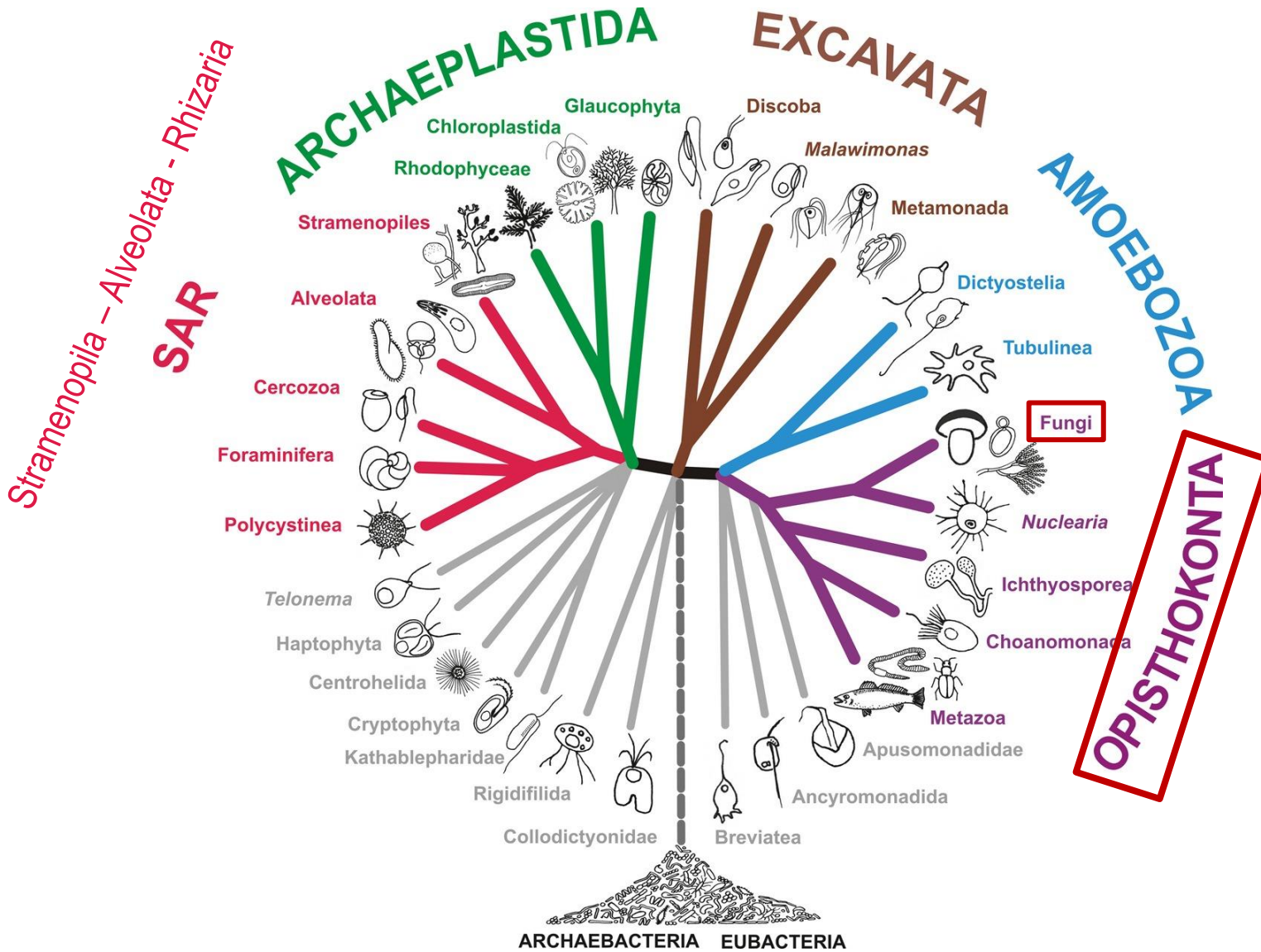
Andrea Bardůnek Valigurová

[andreav@sci.muni.cz](mailto:andreav@sci.muni.cz)

# Notice

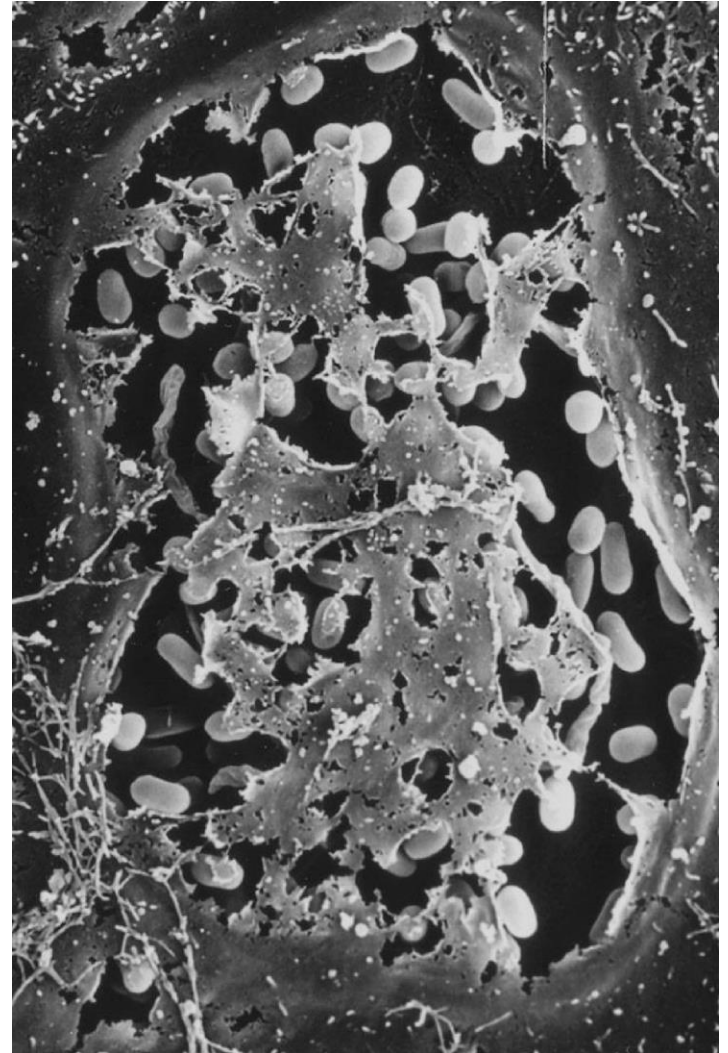
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# 5 supergroups = megagroups



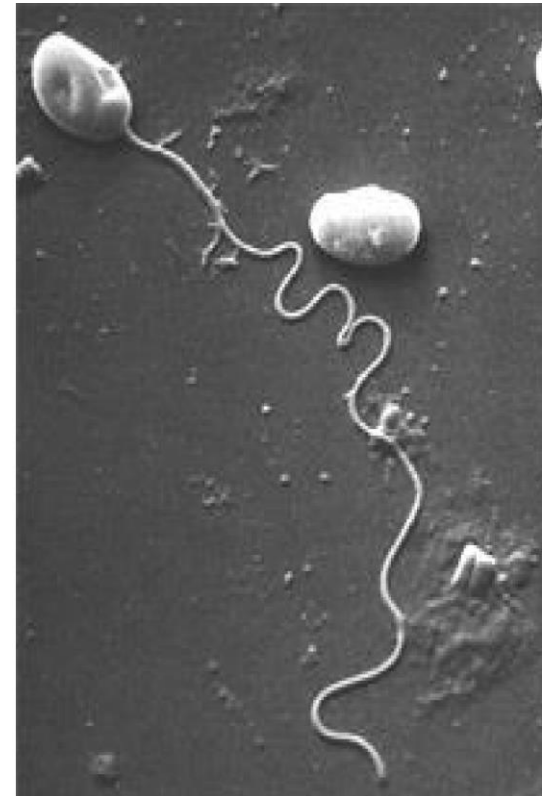
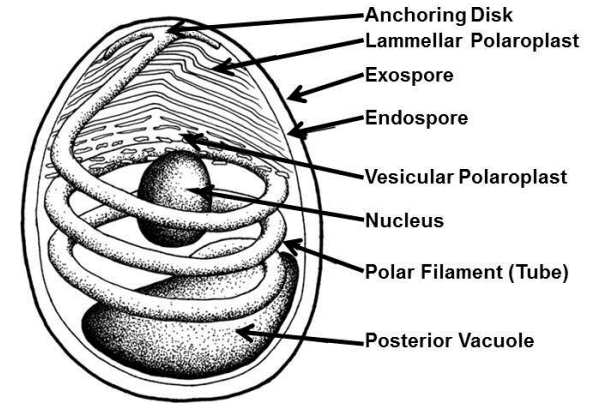
# Microsporidia

- monophyletic group
- about 1,500 species in 187 genera
- traditionally thought to be a unique phylum of spore-forming protozoa
- smallest of eukaryotes = size of spores vary in range 1-40  $\mu\text{m}$  (in medically important species usually 1-4  $\mu\text{m}$ )
- smallest eukaryotic genomes; 2.5 to 11.6 Mb in size
- mitochondria highly reduced to mitosomes
- obligate intracellular parasites
- most infect insects, but they are also responsible for common diseases of crustaceans and fish
- several species also infect humans



# Microsporidia

- spores with inner chitin wall and outer proteinaceous wall are highly resistant in the environment  $\Rightarrow$  surviving for months to years
- spore germination occurs when microenvironmental stimuli result in extrusion of the polar filament (tube)
- polar filament responsible for injecting the sporoplasm into the host cell (epithelial cells, macrophages, endothelial cells)
- replication by binary division within a parasitophorous vacuole
- development of proliferative forms (meronts) that undergo binary division and differentiate into sporoblasts and sporonts



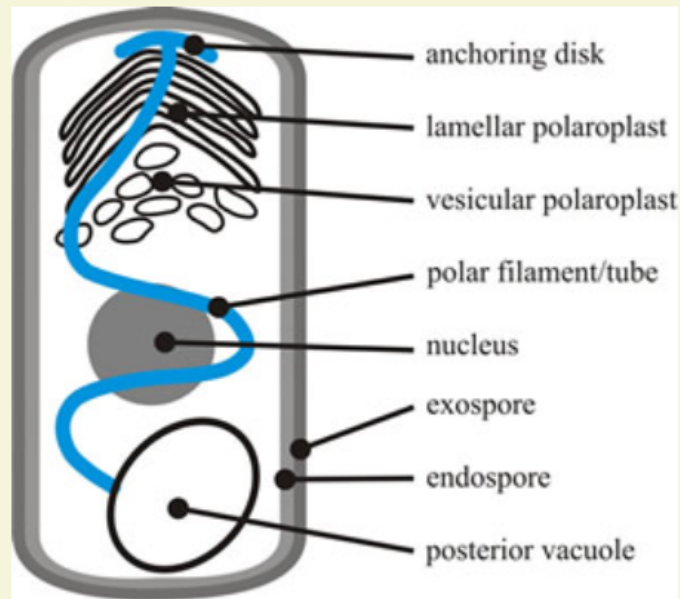


Fig. 1. The major structures of a typical microsporidian spore. The spore cytoplasm is enclosed by a normal plasma membrane with two rigid extracellular walls. The exospore wall has a dense, proteinaceous matrix; the endospore wall, which thins at the apex, is composed of chitin and proteins. The cytoplasm of the spore (the sporoplasm) is the infectious material of microsporidia. It contains either one nucleus or two closely appressed nuclei (= a diplokaryon). The cytoplasm is rich in 70S ribosomes, and is dominated by infection structures: the polaroplast, the polar filament or polar tube, and the posterior vacuole. The membranous polaroplast occupies the anterior part of the spore and is differentiated into the closely stacked membranes of the lamellar polaroplast, and the loosely organised posterior vesicular polaroplast. The polar filament is composed of membrane and glycoprotein layers and ranges from 0.1 to 0.2  $\mu\text{m}$  in diameter and 50 to 500  $\mu\text{m}$  in length, being attached at the apex of the spore via an umbrella-shaped anchoring disk. The polar filament is straight for one third to one half the length of the spore, and the rest is helically coiled in the sporoplasm. The number of coils, their relative arrangement, and angle of helical tilt are conserved and diagnostic for a particular species. The polar filament terminates at the posterior vacuole, though the point of contact has never been observed. Based on illustrations in Keeling & Fast, 2002.

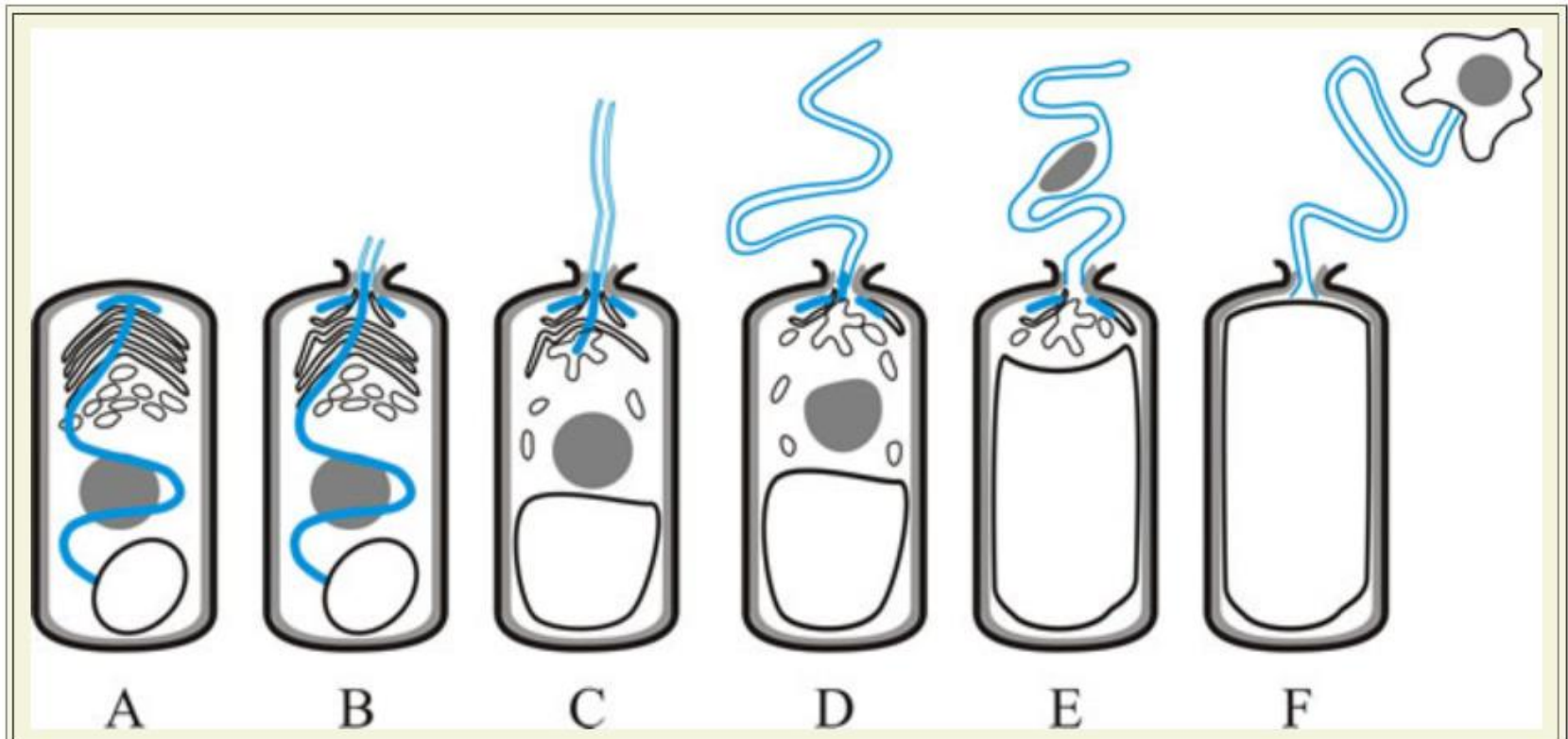
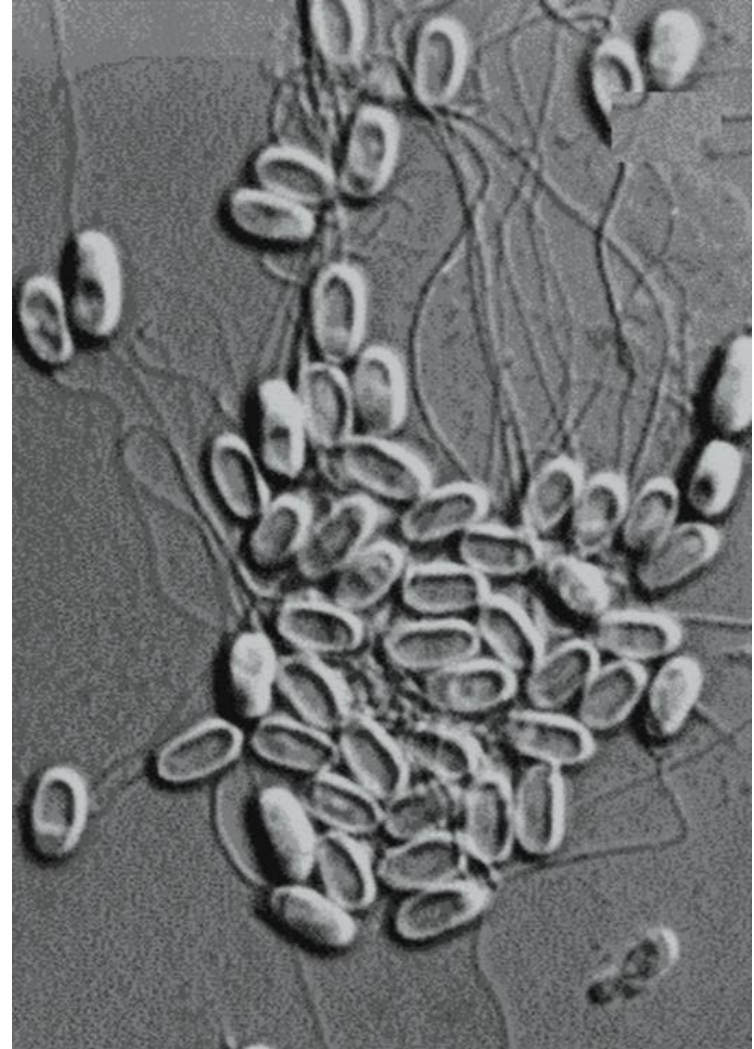


Fig. 2. Germination of a microsporidian spore depends on eversion of the polar filament or tube, and is illustrated in this cartoon strip. **A**, dormant spore, showing polar filament (blue), nucleus (grey), polaroplast and posterior vacuole (as in Fig. 1). **B**, polaroplast and posterior vacuole begin to swell and anchoring disk ruptures; polar filament begins to emerge, everting as it does so. **C**, polar filament continues to evert. **D**, with the polar tube fully everted, the sporoplasm is forced into and then (**E**) through the polar tube. **F**, sporoplasm emerges from the polar tube bound by new membrane. Based on illustrations in Keeling & Fast, 2002.

# Extrusion of polar tube





# Microsporidia - perfect parasites



- ✓ low pathogenicity
- ✓ metabolically completely dependent on the host cell
- ✓ intracellular development during which they "behave" more like an organelle of the host cell
- ✓ host cells react to their presence by muscle hypertrophies (xenomas in fish)
- ✓ host cell dies only after spore formation
- ✓ spores are resistant to environmental conditions
- ✓ spread by water and contaminated food

# Microsporidia in fish

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## Phylogeny and morphology of *Glugea hertwigi* from rainbow smelt *Osmerus mordax* found in Prince Edward Island, Canada

J. Lovy<sup>1,\*</sup>, M. Kostka<sup>2</sup>, I. Dyková<sup>2</sup>, G. Arsenault<sup>3</sup>, H. Pecková<sup>2</sup>, G. M. Wright<sup>1</sup>,  
D. J. Speare<sup>3</sup>

<sup>1</sup>Department of Biomedical Sciences, Atlantic Veterinary College, University of Prince Edward Island, 550 University Ave., Charlottetown, Prince Edward Island C1A 4P3, Canada

<sup>2</sup>Institute of Parasitology, Biology Centre, ASCR, Branišovská 31, 370 05 České Budějovice, Czech Republic

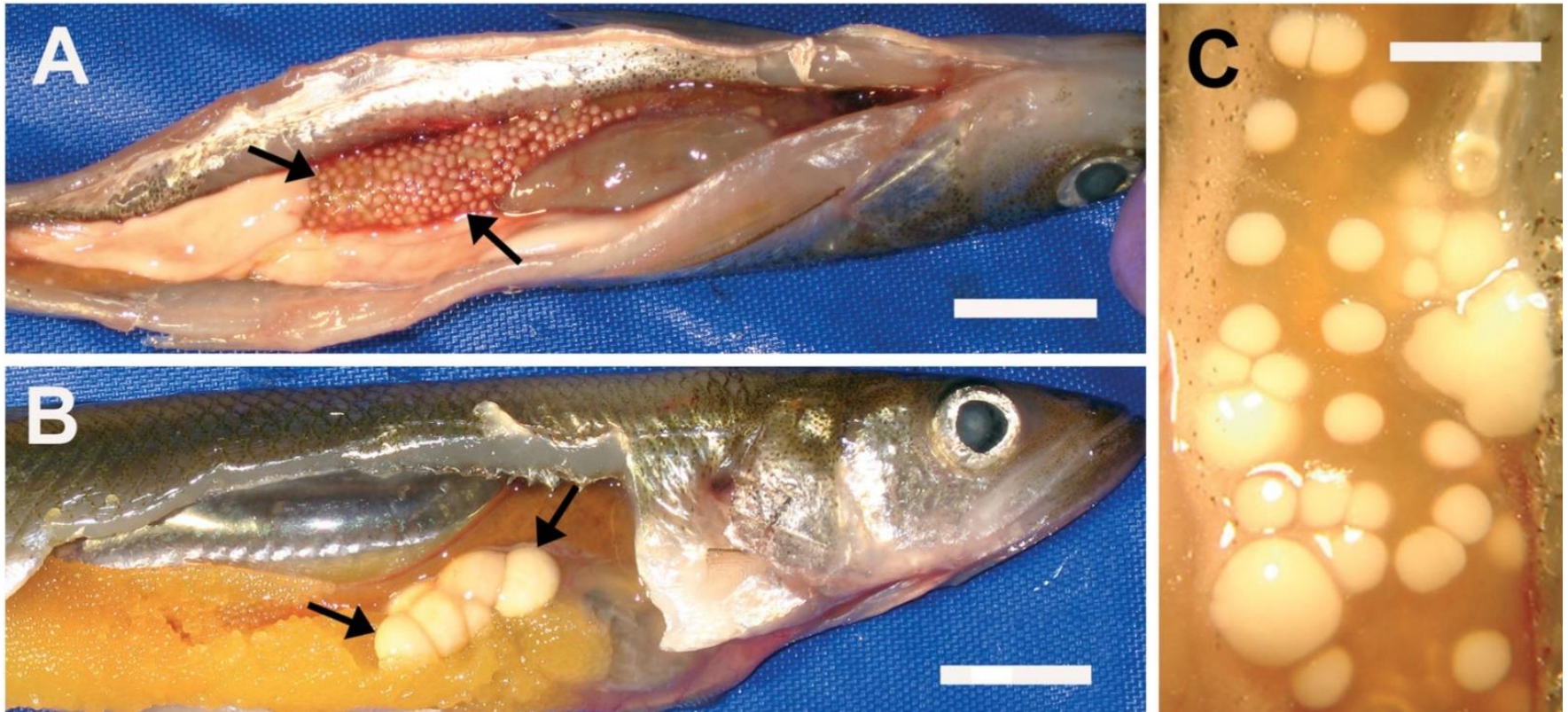
<sup>3</sup>Department of Pathology and Microbiology, Atlantic Veterinary College, University of Prince Edward Island, 550 University Ave., Charlottetown, Prince Edward Island C1A 4P3, Canada

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ABSTRACT: Infection of rainbow smelt *Osmerus mordax* with the microsporidian *Glugea hertwigi* was diagnosed for the first time in Prince Edward Island, Canada. The prevalence of infection was 24%, 45 infected out of 187 examined fish captured in February and March 2009. Both large and small xenomas of *G. hertwigi* observed within the submucosa of the gastrointestinal tract and along the mesentery of the host contained only mature spores. Advanced and degraded xenomas associated with host reaction were described using light and transmission electron microscopy. The first rDNA sequence of *G. hertwigi* prepared in the present study completed the set of sequences of *Glugea* spp. available for comparison. The high level of rDNA sequence identity between *Glugea* spp. suggests that these may be variants of a single species.

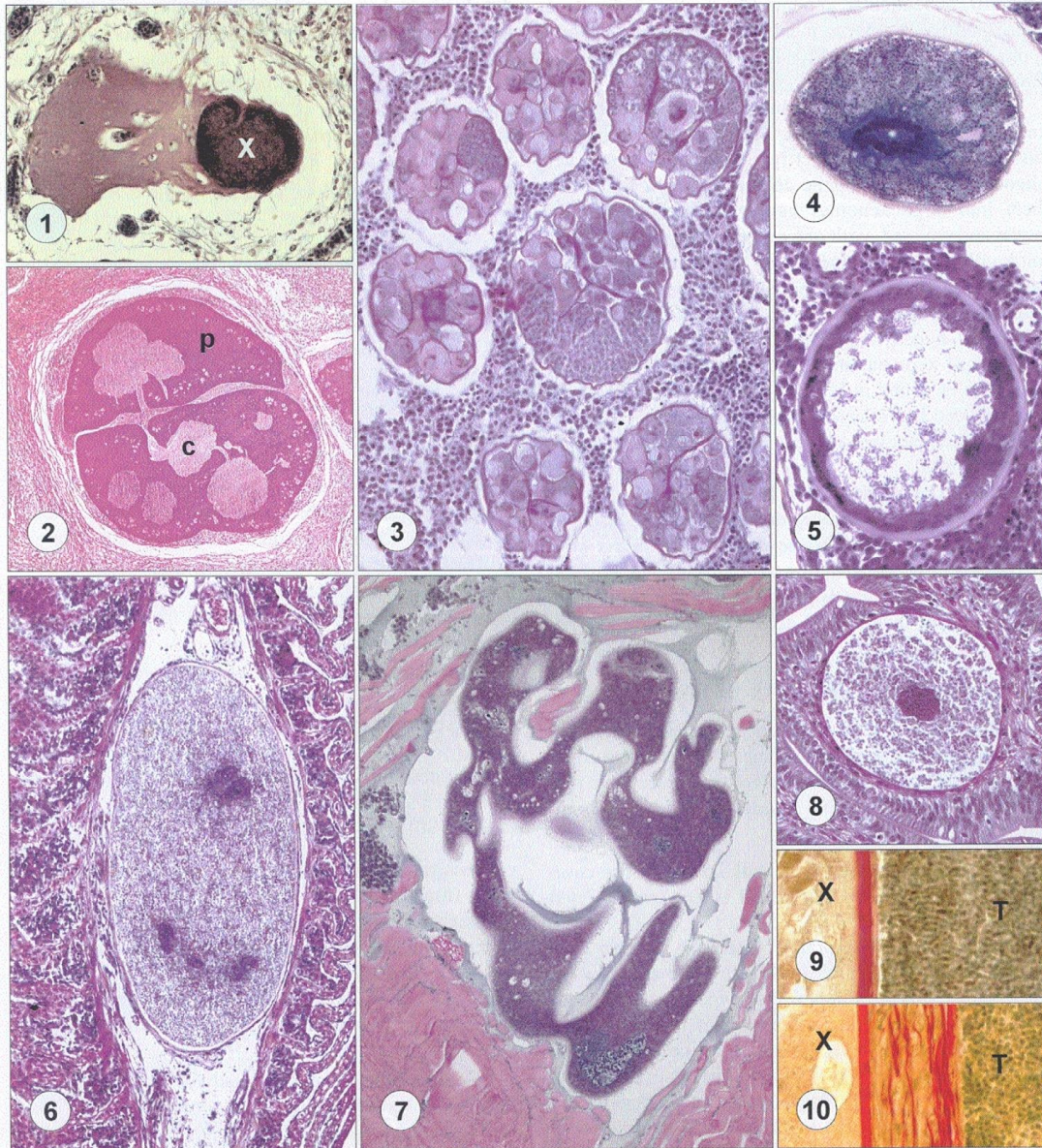
# Microsporidia in fish - xenomas

Xenoma is a growth caused by various protists and fungi, most notably microsporidia. The host cell undergoes hypertrophy and has many, mostly polyploid, nuclei. This outcome is due to the microsporidian parasite proliferating inside the host cell. Symbiotic co-existence develops between host cell and microsporidian and both partners turn into a well-organised xenoparasitic complex.



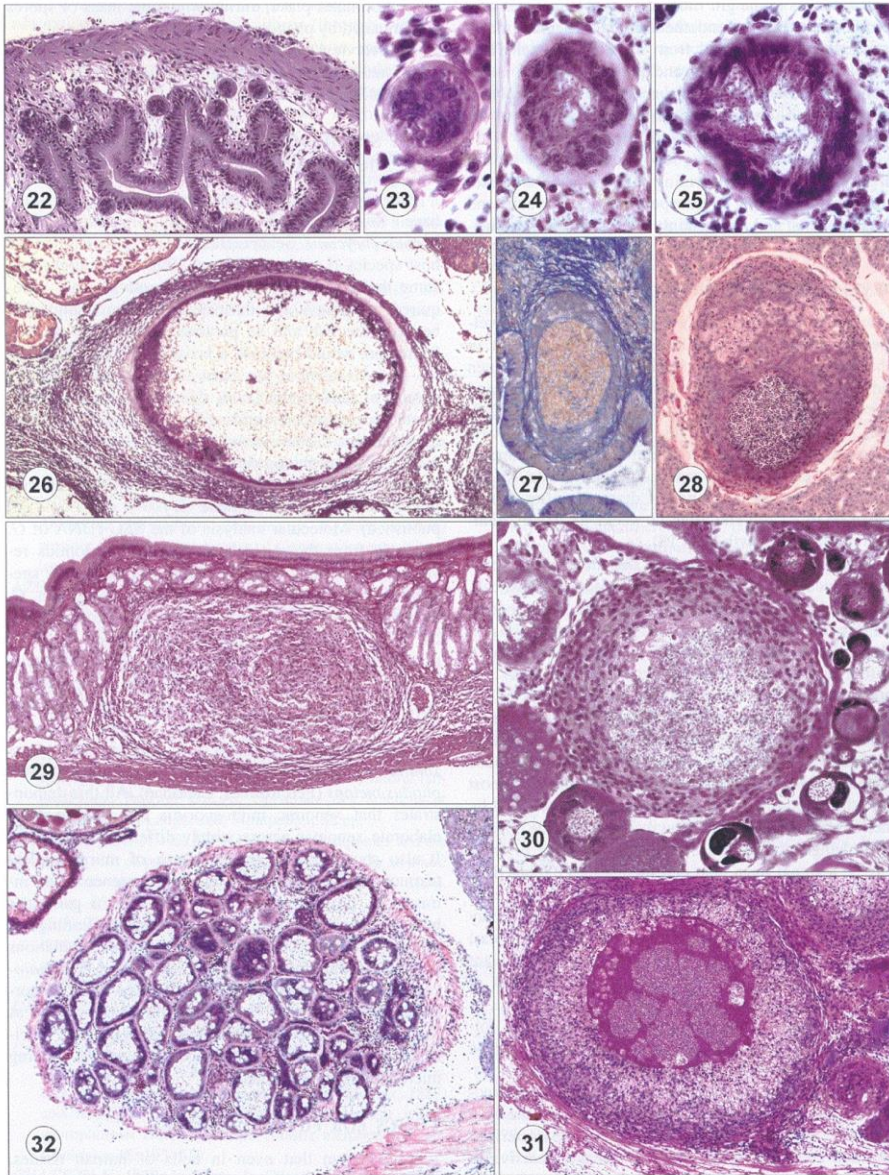
*Glugea hertwigi* infecting *Osmerus mordax*. Variably sized xenomas in the gastrointestinal tract of infected rainbow smelt. **A**) Heavily infected individual with uniformly small xenomas (scale bar = 1 cm). **B**) Moderate infection with uniformly large xenomas (scale bar = 1 cm). **C**) Individual with a mixed infection of small and large xenomas (scale bar = 4 mm).

# Different types of xenomas of fish microsporidia



1) Early stage of *Spraguea lophii* xenoma; parasite mass (X) occupies only part of the host ganglion cell. Bodian. 2) Advanced stage of *S. lophii* xenoma in ganglion. Note the different staining of parasite mass at the periphery (p) with *Nosemoides*-type spores and in the centre (c) with *Nosema*-type spores. 3) "Cystic" stages preceding formation of huge xenomas of *Ichthyosporidium giganteum*. Compartments contain different stages of merogonial proliferation. 4) Xenoma of *Tetramicra brevilum*, in a liquid-filled cavity in host liver parenchyma. 5) Mature xenoma of *Glugea anomala* in the body cavity. 6) Xenoma of *Loma branchialis* in fish gills. 7) Xenoma of *T. brevilum* in folded-over shape in the host muscle tissue. 8) *Loma acerinae* xenoma with a centrally located host cell nucleus in the subepithelial connective tissue of intestine. 1-8 = HE. 9-10) Parts of the wall of similar, mature *G. plecoglossi* xenomas (X), localised in host testes (T). Xenoma wall and mature encircling connective tissue (present in 10) are stained red. Van Gieson.

# Early stages of xenoma development

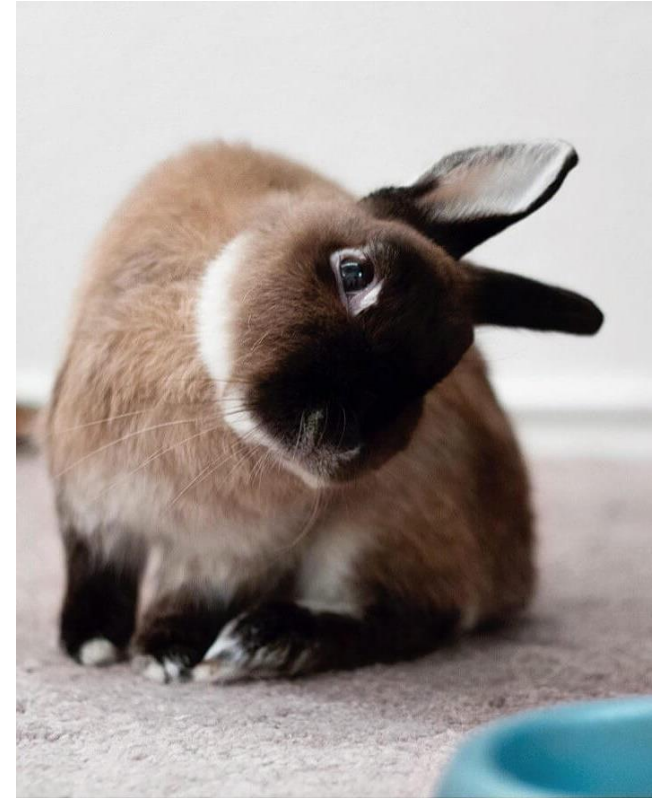


**Xenomas of *Glugea anomala* in early stages of development.** 22) Spontaneous infection with *G. anomala*. 23-25) Early xenomas with hypertrophic branched nuclei and cylindrical meronts, which predominate in 24 and 25. 26-31) Examples of xenoma transformation due to the onset of proliferative inflammation of the host. 26) *Glugea plecoglossi* infection in ovaries. HE. 27) Proliferation of granulation tissue in *Loma acerinae* visualised by Masson's trichrome staining. 28) Xenoma of *Tetramicra brevifilum* transformed into granuloma in the liver. 29) Granulomatous lesion at the site of *Glugea anomala* xenoma in the glandular part of host stomach wall. 30) Granuloma in fish ovary replacing *G. anomala* xenoma. 31) *Spraguea lophii* xenoma partly transformed into a granuloma. 32) Overview of a massive spontaneous infection of *G. anomala* in fish intestine. HE.

# Microsporidia in mammals

## *Encephalitozoon cuniculi*

- the most common and important animal pathogen
- natural infections with *E. cuniculi* in a wide range of hosts (e.g. rabbits, mice, cats, dogs, foxes, humans)
- important pathogenic agents of pet rabbits with 37-68% seroprevalence
- transmission - transplacental, ingestion or inhalation of spores passed in the urine
- most cases of infection are asymptomatic
- infection of brain and kidneys
- clinical signs: liver and kidney failure and calcification, limb weakness and pressure on the inner ear can lead to a loss of balance and hopping in circles, ataxia, head tilt, hind limb paralysis
- end phase signs: more frequent and stronger seizure attack, coma and death



<https://www.youtube.com/watch?v=MER1YAwaO70&t=8s>

## *Nosema apis* / *Nosema ceranae*

- causative agent of bee dysentery
- intestinal epithelium of bees

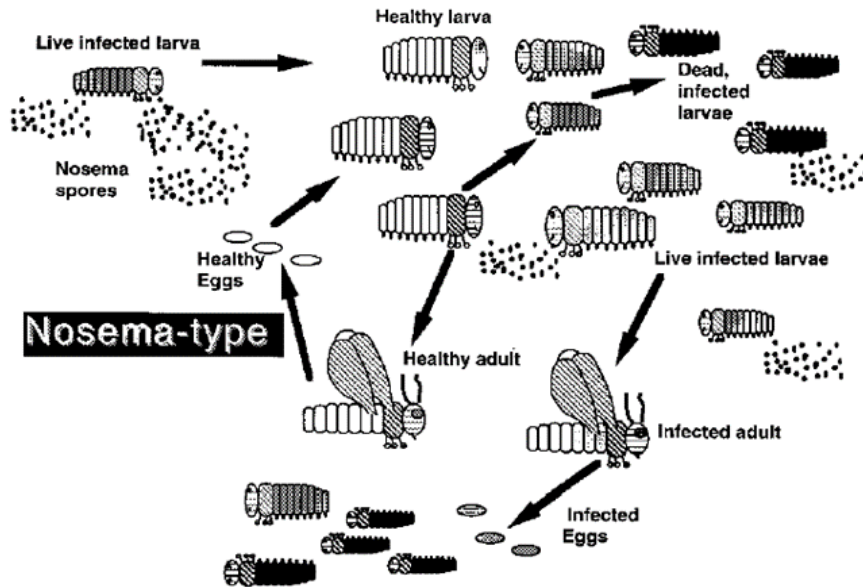
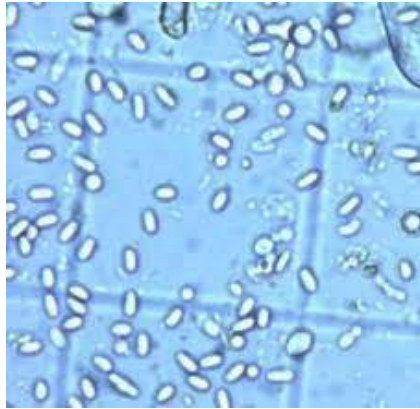


Figure 1. Diagrammatic representation of the interactions between *Nosema*-type microsporidia and their hosts. Healthy larvae become infected by ingesting microsporidian spores which are present in the feces and/or silk of infected individuals. Larvae infected by ingesting spores may die from the infection if they consume many spores at an early larval stage, but many infected larvae can develop into infected adults. Mortality may occur during pupation and emergence as adults. Much of the mortality caused by *Nosema*-type microsporidia occurs in the in the transovarially infected offspring of infected females. Transovarially-infected larvae may be heavily infected and die in early larval stadia.



NOSEMOSIS OF HONEY BEES

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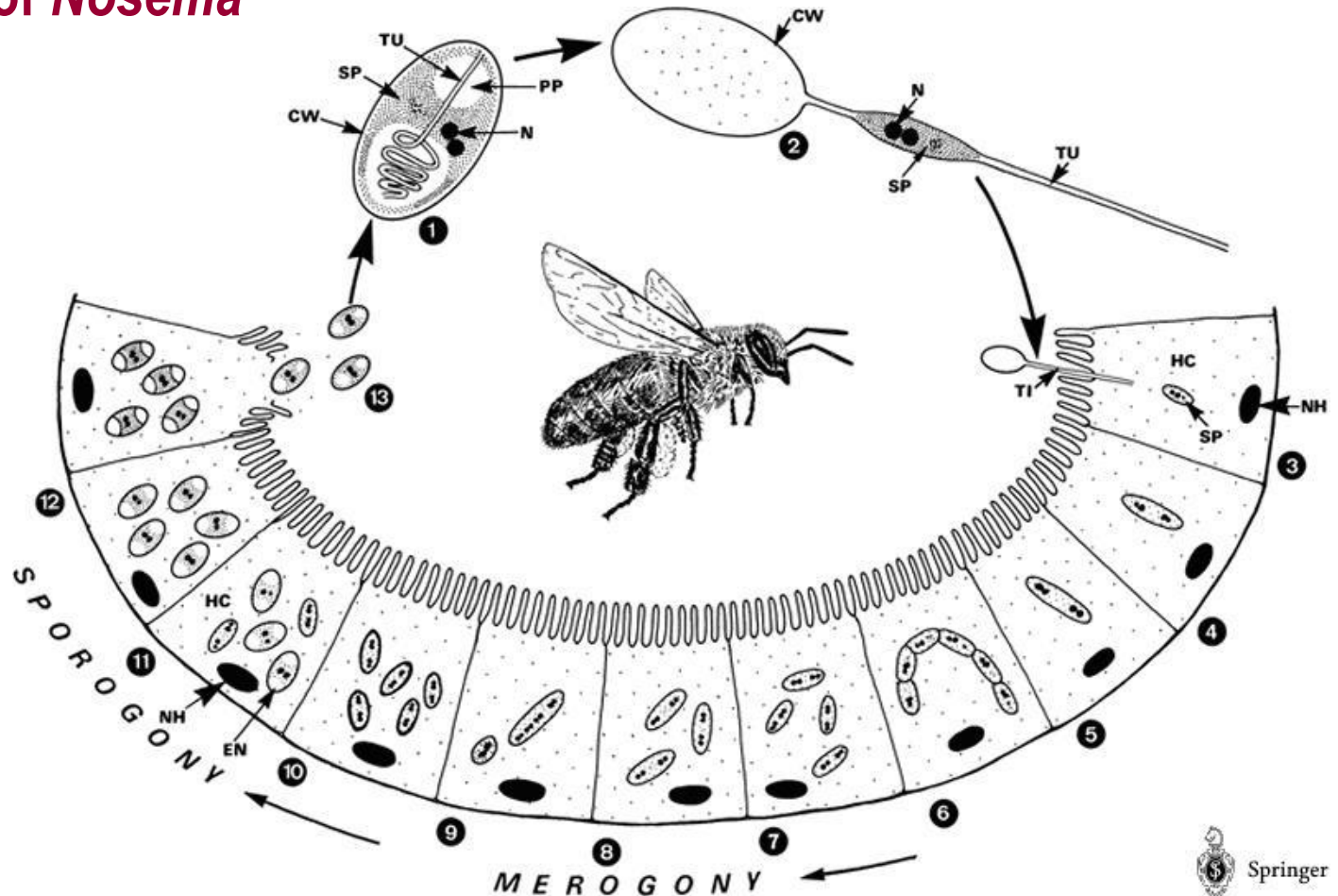
## SUMMARY

*To date, two microsporidian parasites have been described from honey bees: Nosema apis (Zander) and Nosema ceranae (Fries). Nosema apis is a parasite of the European honey bee (Apis mellifera) and Nosema ceranae of the Asian honey bee (Apis cerana) (11) and the European honey bees). The latter has recently been detected in several geographically separated populations of European honey bees in Europe (12), South and North America (14) and Asia (13). The pathological consequences of Nosema ceranae in Apis mellifera are not well known. In the following chapter, only Nosema apis is described. Both types are presumably very similar. Nosema apis is a parasite that invades the epithelial cells of the ventriculus of the adult honey bee. Infections are acquired by the uptake of spores during feeding or grooming. The disease occurs throughout the world, but treatment of bees can help to prevent the spread of infection to unaffected bee colonies.*

*The parasite invades the posterior region of the ventriculus, giving rise to large numbers of spores within a short period of time. The parasite is ubiquitous. Nosema levels generally increase when bees are confined, such as in the autumn and winter in colder climates when the amount of brood is decreasing and perhaps in the early spring when there is an increase in the brood. The disease is transmitted among bees via the ingestion of contaminated comb material and water, and by trophallaxis; honey stores and crushed infected bees may also play a role in disease transmission. Spores are expelled with the faeces where they may retain their viability for more than 1 year. Spores may also remain infective after immersion in honey and in the cadavers of infected bees; however they may lose viability after 3 days when submerged in honey at hive temperature. The relative importance of faeces, honey and cadavers as reservoirs of infective spores is not fully understood. However, it seems likely that faecal contamination of wax, especially in combs used for brood rearing, or other hive interior surfaces, provides sufficient inoculum for N. apis to be successfully transmitted to the next generation of bees. The spores are inactivated by acetic acid or by heating to 60°C for 15 minutes. To be effective, these treatments, which inactivate spores on hive surfaces and combs, can be combined with feeding colonies with the antibiotic fumagillin to suppress infections in live bees. The EU prohibits the use of antibiotic fumigation (EU 3/01/081).*



# Life cycle of *Nosema*



Infection begins when a bee ingests *Nosema* spores, which then germinate inside the midgut of the bee. Parasite enters the host enterocytes and begins to absorb nutrients ⇒ damage to the cell and increased susceptibility to secondary infections. *Nosema* grows and multiplies infesting more of the midgut cells and produces spores. Several million spores can be produced in a single bee worker. Spores either germinate within the bee's midgut, infecting new cells, or pass through the bee's digestive system. Faecal material containing spores can contaminate food and water sources, where they can then be ingested by other bees. Spores can also be spread to non-infected bees when they clean contaminated combs, or rob contaminated hives and ingest spores in the process.

## Effect of *Nosema* on honey bees

- ✓ hypopharyngeal (brood food) glands of infected nurse bees lose the ability to produce royal jelly which is fed to honey bee brood
- ✓ high proportion of eggs laid by the queen of an infected colony may fail to produce mature larvae
- ✓ infected queens cease egg-laying and die within a few weeks
- ✓ young infected nurse bees cease brood rearing turning to guarding and foraging duties - usually undertaken by older bees
- ✓ reduction of life expectancy of infected bees - in spring and summer, infected bees live half as long as non-infected bees
- ✓ increase of dysentery in adult bees although *Nosema* is not the prime cause of dysentery

# Other significant species of microsporidia

## *Nosema bombycis*

- first described microsporidia
- parasite of silkworm caterpillars, transovarian transmission
- Pasteur recommended rearing caterpillars from clutches of microscopically inspected females

## *Nosema locustae*

- biocontrol

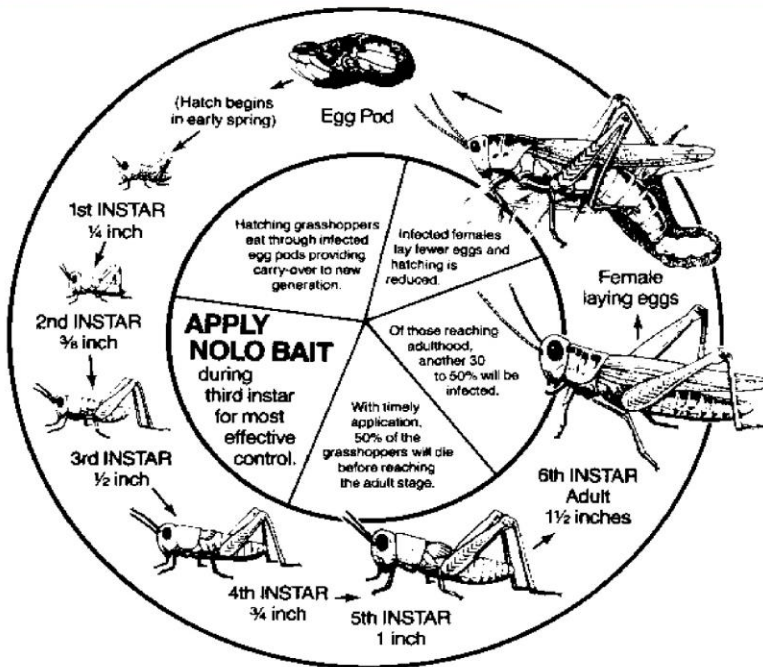
## *Vairimorpha necatrix*

- butterfly caterpillars

## *Pleistophora hypohessobryconis*

- muscles of neon tetra

# Grasshopper control with *Nosema locustae* baits



# Grasshopper control with *Nosema locustae* baits

## STORAGE AND DISPOSAL

Do not contaminate water, food, or feed by storage and disposal.

## PESTICIDE STORAGE

Store in a cool, dry shaded area in original container or refrigerate. If refrigerated continuously at 40° F. add 30 days to expiration date. If frozen continuously at 15° F. add 60 days to expiration date.

## PESTICIDE DISPOSAL

To avoid wastes, use all material in this container by application according to label directions. If wastes cannot be avoided, offer remaining product to a waste disposal facility or pesticide disposal program (often such programs are run by state or local governments or by industry).

## CONTAINER DISPOSAL

Nonrefillable container. Do not reuse or refill this container. Triple rinse container (or equivalent) promptly after emptying. Triple rinse as follows: Empty the remaining contents into application equipment or mix tank. Fill the container 1/4 full with water and recap. Shake for 10 seconds. Pour rinsate into application equipment or mix tank or store rinsate for later use or disposal. Drain for 10 seconds after the flow begins to drip. Repeat this procedure two more times. Then offer for recycling if available or puncture and dispose of in a sanitary landfill, or by incineration. Do not burn, unless allowed by state and local ordinances.

Batch code \_\_\_\_\_

## Warranty

No warranty is expressed or implied concerning this product except for that on the label.

## Use Before Expiration Date

(Stamped On Container Lid)

# A BIOLOGICAL INSECTICIDE FOR GRASSHOPPERS

# SEMASPORE

# BAIT



Active Ingredient: *Nosema locustae*\*..... 0.05%  
Other Ingredients: ..... 99.95%  
Total: ..... 100.00%

\* Contains  $1.0 \times 10^9$  viable *Nosema* spores per lb (454 grams)

**KEEP OUT OF REACH OF CHILDREN**

(See back panel for additional precautionary statements.)

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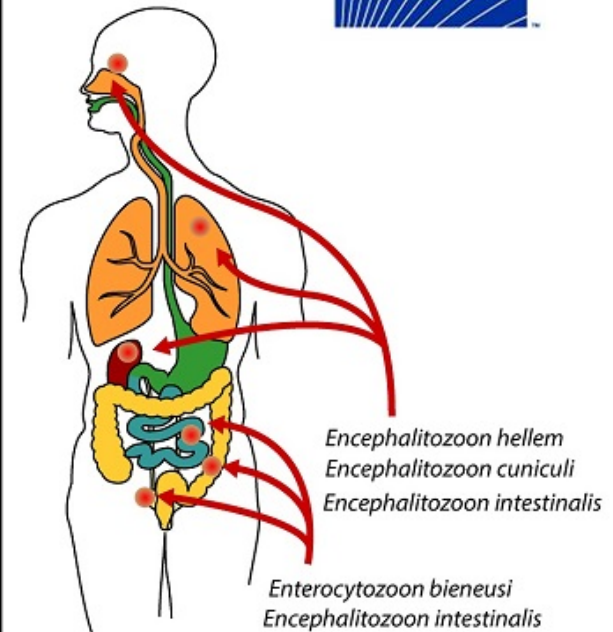
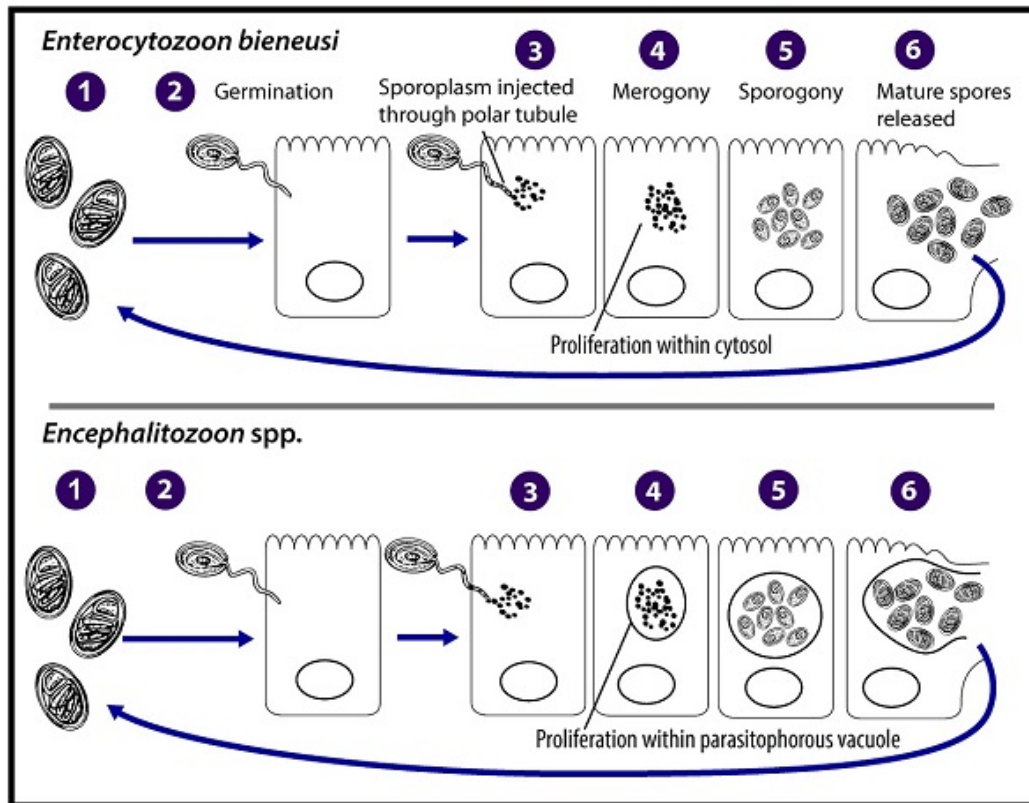
Organic Materials Review Institute

# Human microsporidiosis

- at least 15 microsporidian species that have been identified as human pathogens
  - Enterocytozoon bieneusi*
  - Encephalitozoon cuniculi*
  - Encephalitozoon intestinalis*
  - Encephalitozoon hellem*
- other genera and species - isolated infections
- opportunistic parasites with worldwide distribution
- mostly in severely immunocompromised patients with AIDS, but also in immunocompromised not infected with HIV and immunocompetent persons
- diverse clinical manifestations, depending on species and route of infection, while *E. bieneusi*-associated diarrhoea is the most common one
- some of them are thermotolerant (causing systemic infections), while some are only capable of causing eye infections
- disseminated infection can be fatal
- many domestic and wild animals may be naturally infected with various medically-important microsporidia

# Human microsporidiosis

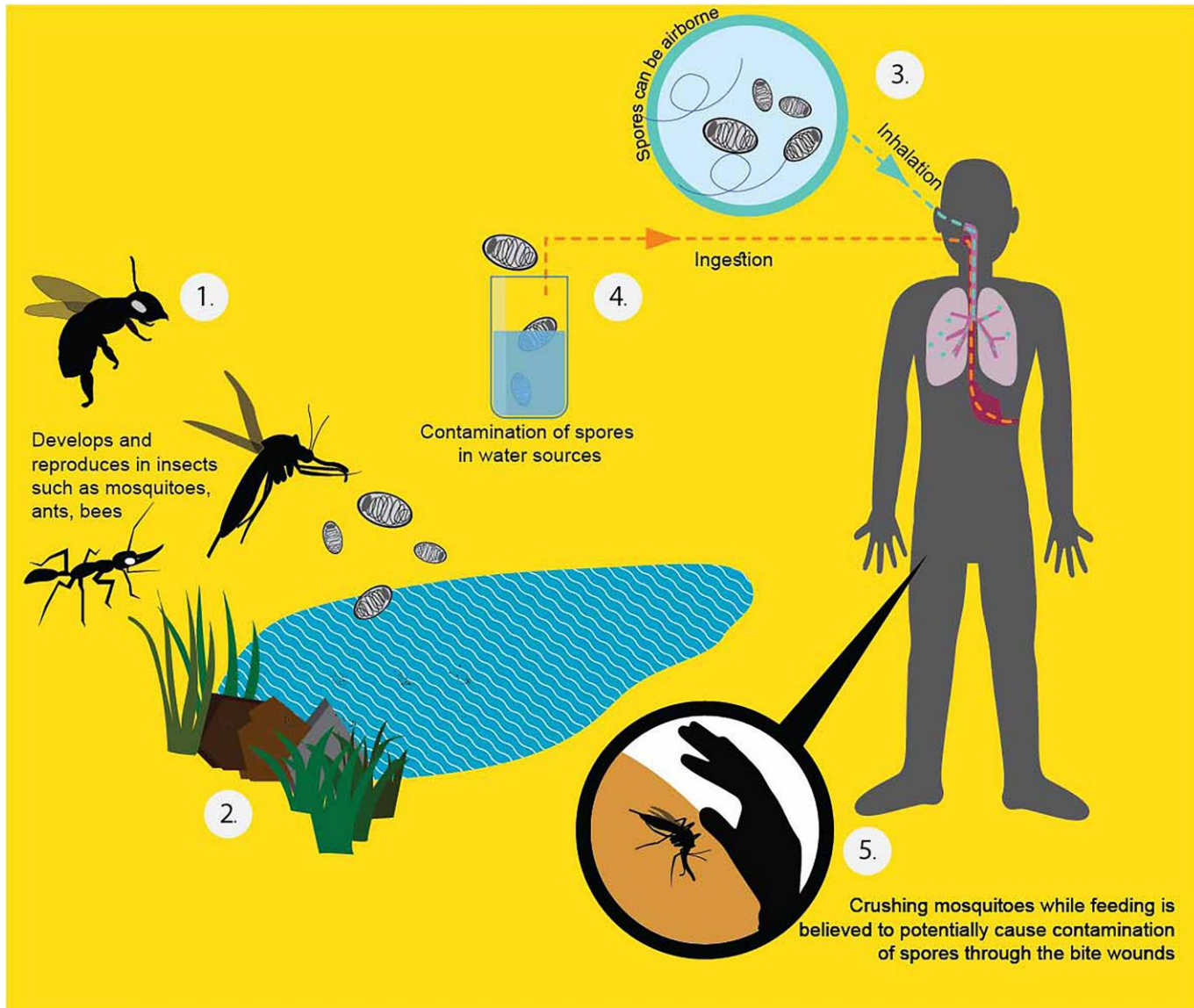
## Intracellular development



Infective stage is the resistant spore (1) which germinates, rapidly everting its polar tubule which contacts eukaryotic host cell membrane (2). Spore injects the infective sporoplasm into host cell through the polar tubule (3). Inside the cell, the sporoplasm enters proliferative phase marked by extensive multiplication via merogony (binary fission or multiple fission), creating meronts (4). Location of this stage within host cell varies by genus; it can occur either in direct contact with host cell cytosol (*Enterocytozoon*, *Nosema*), inside a parasitophorous vacuole of unknown origin (*Encephalitozoon*), in a parasite-secreted envelope (*Pleistophora*, *Trachipleistophora*), or surrounded by host cell endoplasmic reticulum (*Endoreticulatus*, *Vittaforma*) (5). Following proliferative phase, meronts undergo sporogony in which the thick spore wall and invasion apparatus develop, creating sporonts and eventually mature spores when all organelles are polarised. When the spores increase in number, completely filling host cell cytoplasm, cell membrane is disrupted, and spores are released to the surroundings (6). Free mature spores can infect new cells to continue the cycle.

Mature spores of intestinal species may be shed in feces, but the route of transmission is uncertain for many species. Exposure to spores in water or in soil appears to be a potentially major route. Cases of donor-derived microsporidiosis (*Encephalitozoon cuniculi*) following bone marrow, kidney, liver, and heart transplantation have been confirmed.

# Proposed transmission of *Trachipleistophora hominis* in humans





# Human microsporidiosis

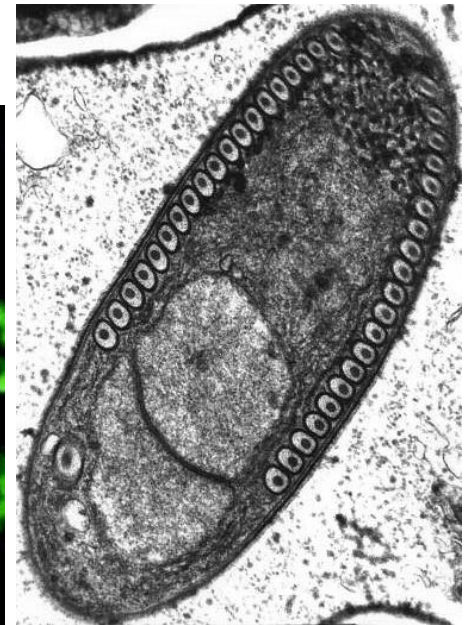
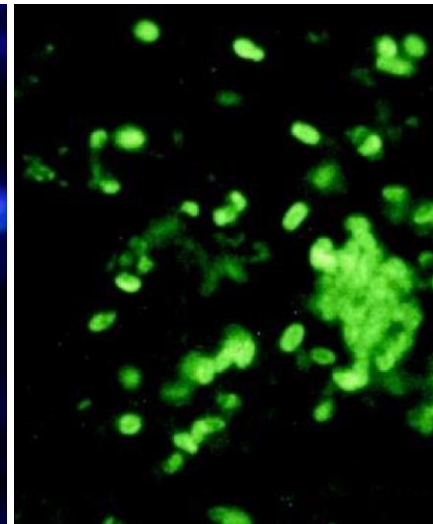
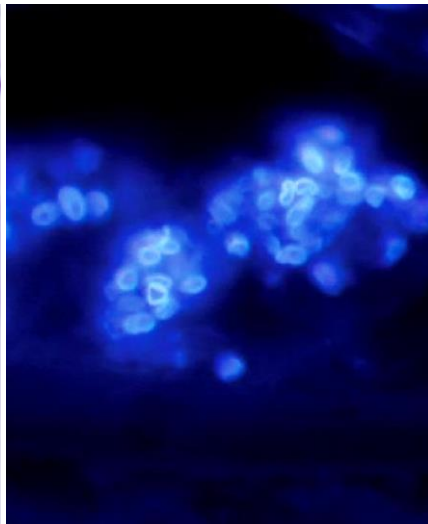
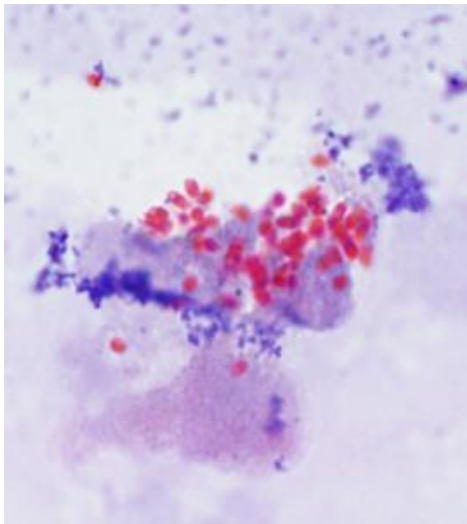
**TABLE 1** Species of microsporidia infecting humans

Species	Synonym(s)	Common sites of infection in humans	Other mammalian hosts	Reported nonmammalian hosts	Disease manifestations reported
<i>Anncaliia algerae</i>	<i>Brachiola algerae</i> <i>Nosema algerae</i>	Eye Muscle		Mosquitoes	Myositis, keratoconjunctivitis, cellulitis
<i>Anncaliia connori</i>	<i>Brachiola connori</i> <i>Nosema connori</i>	Systemic			Disseminated disease
<i>Anncaliia vesicularum</i>	<i>Brachiola vesicularum</i>	Muscle			Myositis
<i>Encephalitozoon cuniculi</i>	<i>Nosema cuniculi</i>	Systemic	Wide host range	Birds	Hepatitis, encephalitis, peritonitis, urethritis, prostatitis, nephritis, sinusitis, keratoconjunctivitis, cystitis, diarrhea, disseminated infection
<i>Encephalitozoon hellem</i>		Eyes	Bats	Birds	Superficial keratoconjunctivitis, sinusitis, pneumonitis, nephritis, prostatitis, urethritis, cystitis, diarrhea, disseminated infection
<i>Encephalitozoon intestinalis</i>	<i>Septata intestinalis</i>	Small intestine	Wide host range	Geese	Diarrhea, intestinal perforation, cholangitis, nephritis, superficial keratoconjunctivitis, disseminated infection
<i>Enterocytozoon bieneusi</i>		Small intestine Biliary tract	Wide host range	Birds	Diarrhea, malabsorption with wasting syndrome, cholangitis, rhinitis, bronchitis
<i>Microsporidium africanum</i>		Eyes			Stromal keratitis
<i>Microsporidium ceylonensis</i>		Eyes			Stromal keratitis
<i>Microsporium</i> CU (Endoreticulatus-like)		Muscle			Myositis
<i>Nosema ocularum</i>		Eyes			Stromal keratitis
<i>Pleistophora ronneafiei</i>		Muscle		Fish <sup>a</sup>	Myositis
<i>Trachipleistophora anthropoptera</i>		Eyes Systemic		Insects <sup>a</sup>	Encephalitis, keratitis, disseminated infection
<i>Trachipleistophora hominis</i>		Eyes Muscle		Mosquitoes <sup>a</sup>	Myositis, keratoconjunctivitis, sinusitis, encephalitis
<i>Tubulinosema acridophagus</i>		Muscle Systemic		Fruit fly <sup>a</sup>	Myositis, disseminated infection
<i>Vittaforma corneae</i>	<i>Nosema corneum</i>	Eyes Urinary tract			Keratoconjunctivitis, urinary tract infection

<sup>a</sup>Putative host(s) based on phylogeny or host relationships of other species within the genus.

# Diagnosis of microsporidiosis

- examined material: stool, urine, BAL, biopsy material, autopsy material
- detection of spores by light microscopy:
  - ✓ chromotrope based staining - Gram, Ziehl-Nielsen
  - ✓ chemofluorescent staining - Calcofluor White M2R, Uvitex
- serological test for antibody detection (IFAT, ELISA)
- transmission electron microscopy
- PCR



# Cultivation of microsporidia

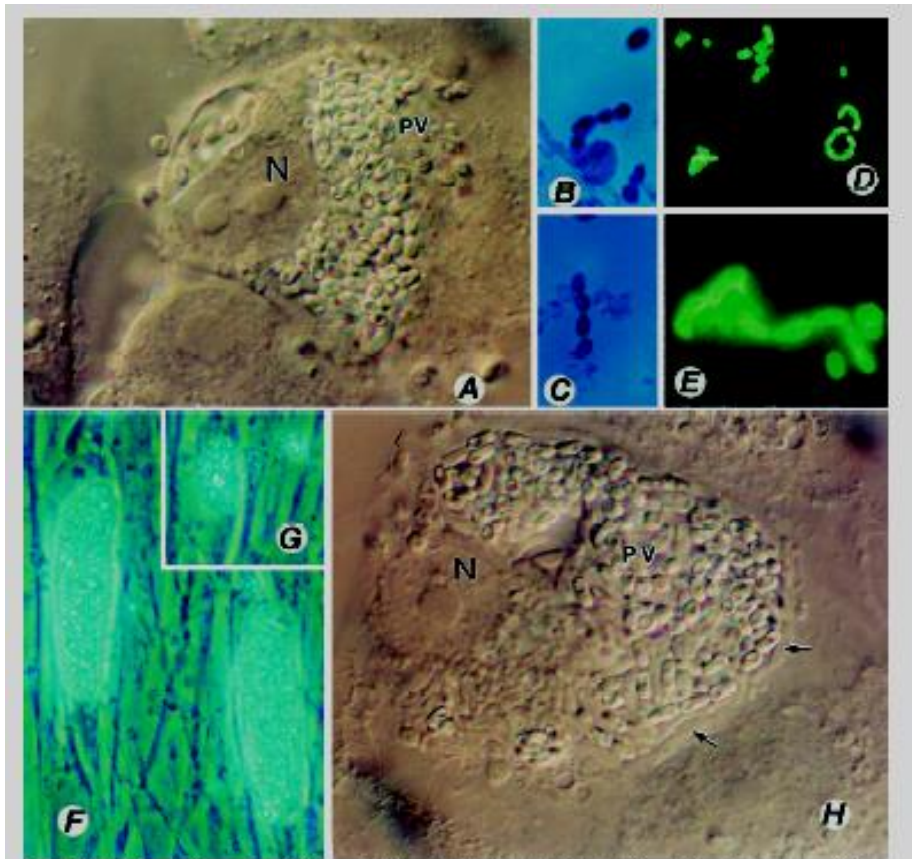


FIG. 3. (A to E) *Eosporoblastores coenocytii*. (A) A monkey kidney (HE) cell distended with spores of *E. coenocytii* growing inside a parasitophorous vacuole (PV). Magnification,  $\times 1,200$ . N, host cell nucleus. (B and C) Gram-chromotrope-stained preparations exhibiting chains of spores with four spores. Magnification,  $\times 1,200$ . (D and E) Immunofluorescence patterns of spores. A centrifuged pellet obtained from an infected flask reacted first with a 1:1,000 dilution of a rabbit anti-*E. coenocytii* serum and subsequently with a 1:100 dilution of a fluorescein isothiocyanate-conjugated goat anti-rabbit immunoglobulin G. Note spore chains of two, three, and five spores (D). Magnification,  $\times 250$ . Note chains of two and four spores (E). Magnification,  $\times 1,500$ . Such chains of spores are also seen in cultures of *E. isolae* and *E. intestinalis*, illustrating poly-spore sporogony, which is commonly seen in cultures infected with *Eosporoblastores* species. (F to H) Cell cultures infected with *Eosporoblastores isolae*. Note two HLF cells distended with spores and developing stages of the parasite, giving the appearance of corn on the cob (F). Magnification,  $\times 600$ . (G) Early stages depicting the process of invasion and the formation of cots on the cob architecture. Magnification,  $\times 600$ . (H) An HE cell distended with spores and developing stages (small arrows) of *E. isolae*. Magnification,  $\times 1,200$ .

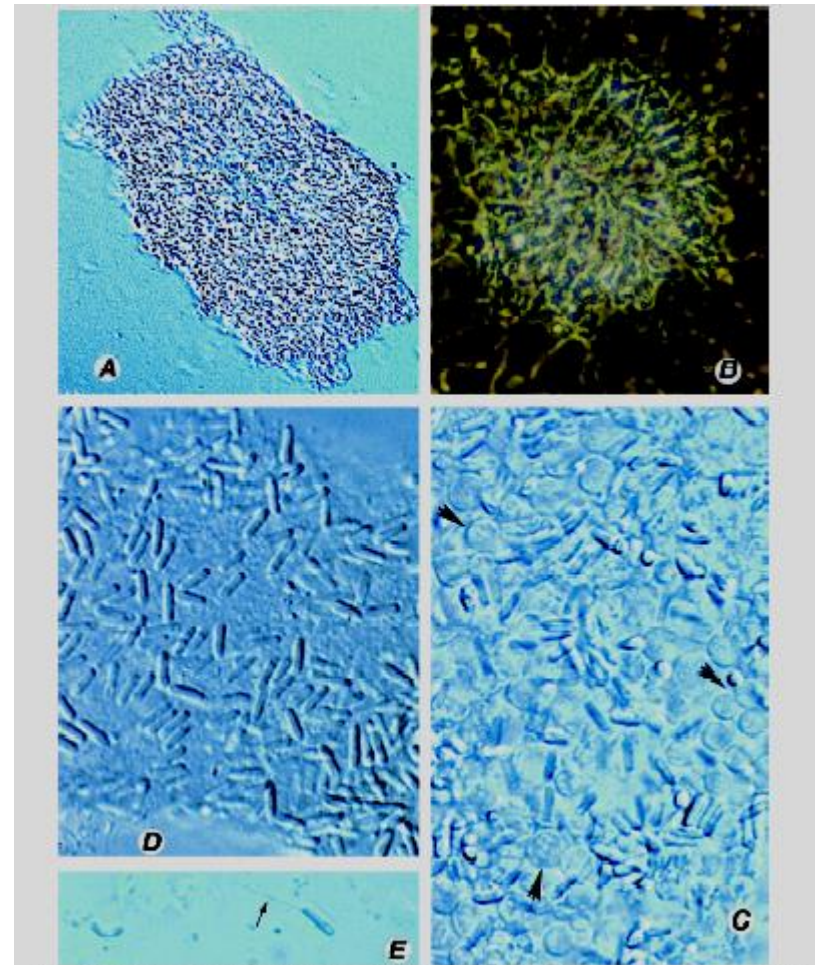
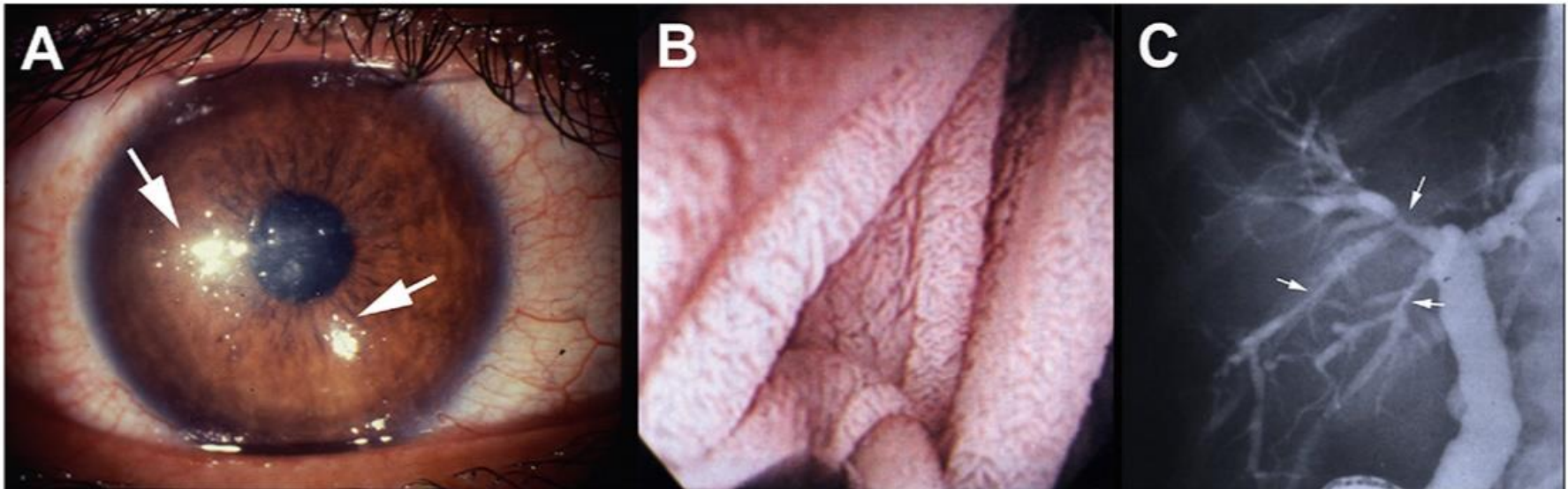


FIG. 4. *Palaemonis coenocytii*. (A) Low-power view of a heavily infected culture. Magnification,  $\times 120$ . (B) The same culture viewed with a high dry lens. Magnification,  $\times 600$ . (C) A heavily infected cell culture with spores and developing stages (arrowhead) of *P. coenocytii*. Magnification,  $\times 1,200$ . Note the absence of the normal architecture of the cell culture. (D) Fully formed spores are interspersed with the host cell debris. Magnification,  $\times 1,300$ . (E) A spore with everted polar tubule (arrow) and posterior vacuole. Magnification,  $\times 1,200$ .

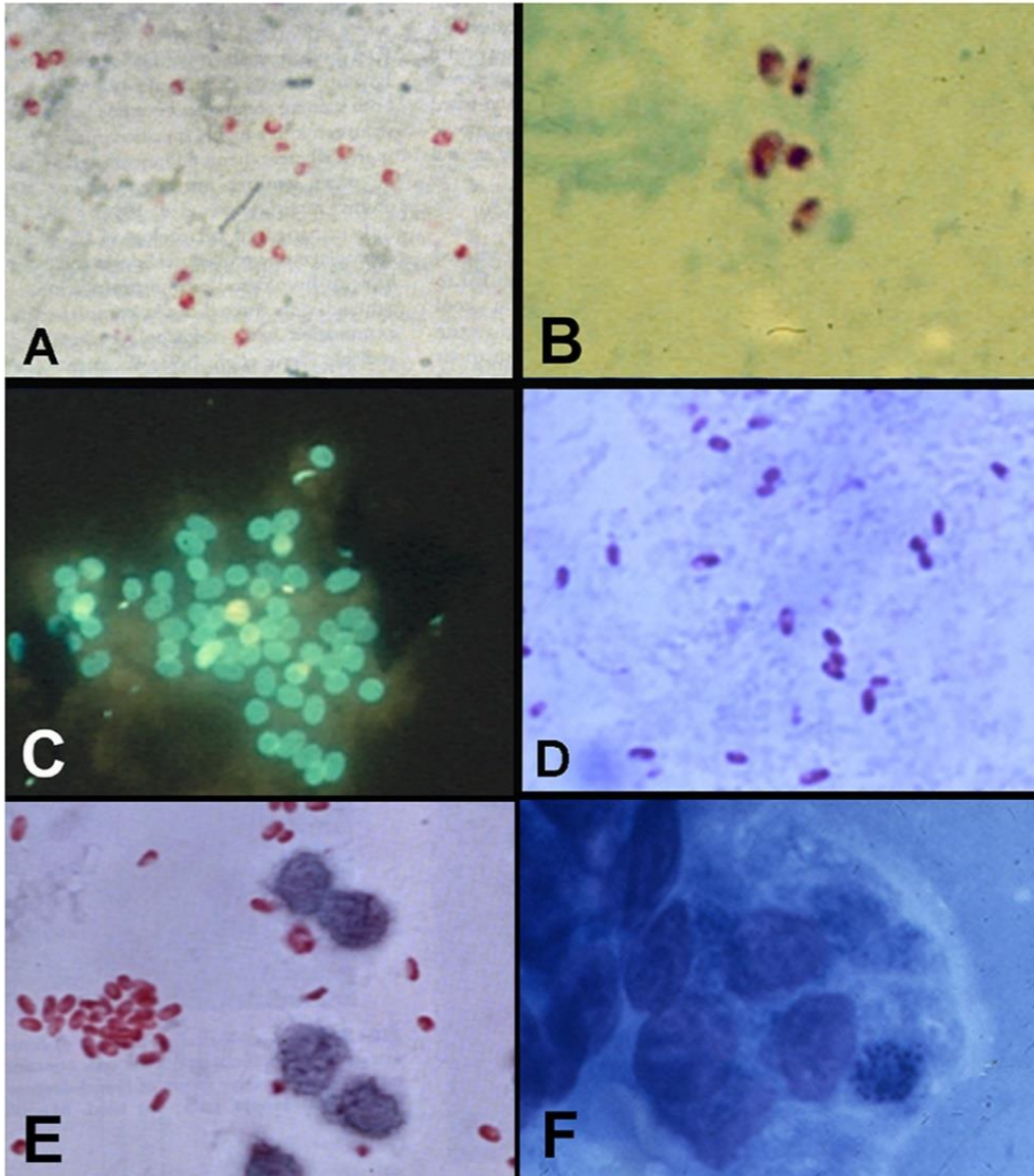
# Pathology and diagnosis of microsporidiosis



**Clinical images from patients with microsporidiosis.** A) *Encephalitozoon hellem* keratoconjunctivitis demonstrating punctate corneal lesions (white arrows). B) Endoscopy of jejunal mucosa of a patient with gastrointestinal microsporidiosis due to *E. bienersi* demonstrating fusion of the villi. C) Endoscopic retrograde cholangiogram (ERCP) of a patient with HIV infection (AIDS) and sclerosing cholangitis due to *E. bienersi* infection. ERCP demonstrates diffuse dilations of the common bile duct with irregular walls, plus areas of narrowing and dilation (arrows) of the intrahepatic bile ducts.

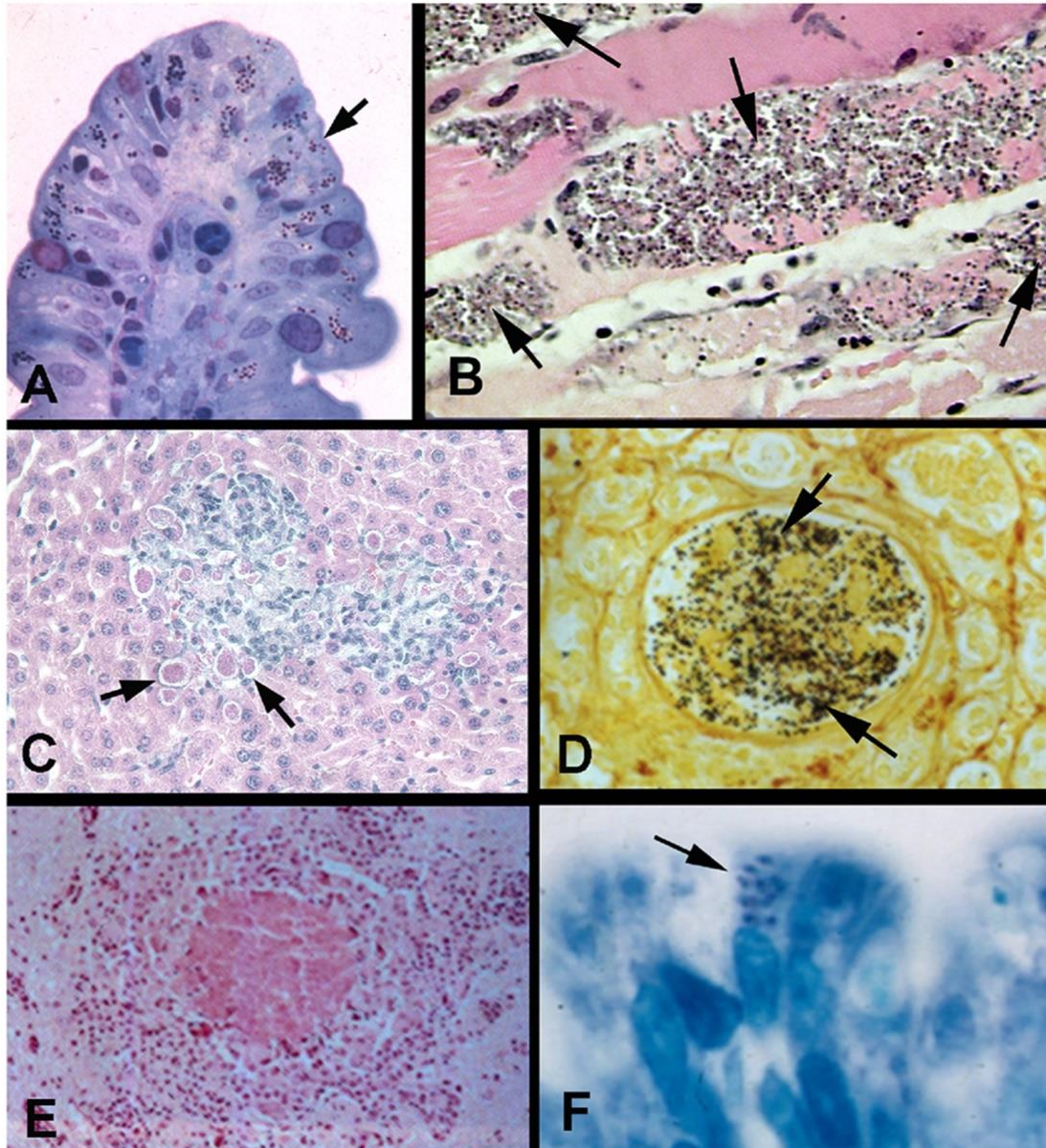
<https://doi.org/10.1128/CMR.00010-20>

# Pathology and diagnosis of microsporidiosis



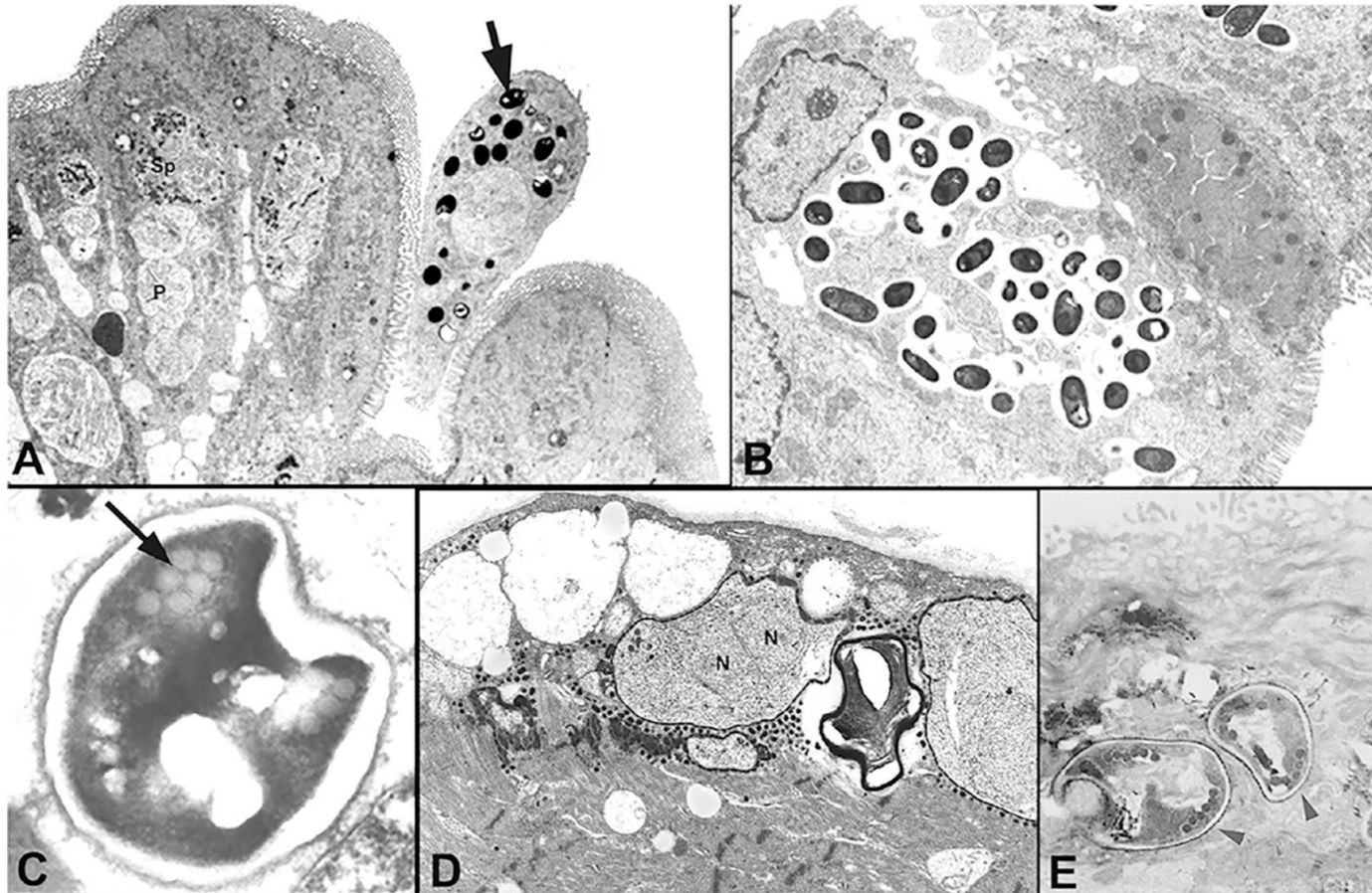
**Microsporidia in stool, urine, and other clinical specimens.** A-B) Chromotrope staining of stool specimens from patients with AIDS and diarrhoea demonstrating spores of *Enterocytozoon bienersi* measuring 0.7-1 x 1.1-1.6  $\mu\text{m}$ . Spores have a pink to reddish hue with this stain. Stained spores can have a safety pin appearance as well. C) Conjunctival scraping from a patient with *E. hellem* keratoconjunctivitis stained with chemifluorescent brightening agent (staining the chitin) demonstrating microsporidian spores. D) Stool specimen from a patient with AIDS and *E. intestinalis* infection stained with a quick hot Gramchromotrope staining demonstrating spores of 1-1.2 x 2-2.5  $\mu\text{m}$ . Spores have a violet hue with this stain. E) Chromotrope staining of urine sediment from a patient with HIV infection who had a disseminated *E. cuniculi* infection demonstrating pink-to red-coloured spores that are 1.0 to 1.5 by 2.0 to 3.0  $\mu\text{m}$ . F) Methylene blue-azure II-fuchsin staining of a touch preparation of an intestinal biopsy specimen (obtained by endoscopy) from a patient with intestinal *E. bienersi* infection demonstrating intracellular spores.

# Pathology and diagnosis of microsporidiosis



**Biopsy specimens from patients with microsporidiosis.** **A)** Intestinal biopsy specimen from patient with *Encephalitozoon intestinalis* (arrow) infection. Methylene blue-azure II-fuchsin stain. **B)** Muscle tissue from a patient with rheumatoid arthritis treated with antibody to TNF- $\alpha$  demonstrating *Anncalia algerae* (arrow) myositis. HE. **C)** Liver biopsy specimen from an immunodeficient mouse infected with *Encephalitozoon cuniculi* (arrows). Chromotrope stain. **D)** Renal biopsy revealing *Encephalitozoon hellem* (black spores) within lumen of renal tubule. Steiner stain. **E)** Brain tissue from a rabbit with torticollis, demonstrating a microgranuloma with central necrosis due to *Encephalitozoon cuniculi* infection. No spores are seen in this image. HE. **F)** Intestinal villus biopsy specimen from a patient with AIDS and *Enterocytozoon bieneusi* infection (arrow). Methylene blue-azure II-fuchsin.

# Pathology and diagnosis of microsporidiosis



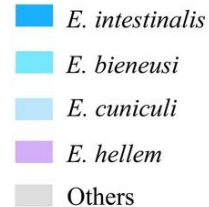
**TEM of biopsy specimens from patients with microsporidiosis.** **A)** Duodenal epithelium from a patient with AIDS and *Enterocytozoon bienewisi* infection demonstrating proliferating stages (P) and late sporogonial plasmodia (Sp). The arrow points to a sloughing enterocyte containing mature spores. **B)** Intestinal biopsy specimen from a patient with AIDS and *E. intestinalis* infection demonstrating spores within vacuoles containing a fibrillar matrix. **C)** *E. bienewisi* spore demonstrating the characteristic finding of two rows of three cross sections of the polar tube (arrow). **D)** Muscle biopsy specimen from a patient with rheumatoid arthritis on an anti-TNF- $\alpha$  antibody who presented with myositis. Proliferative forms are seen in the biopsy specimen with a characteristic diplokaryotic nucleus (N). **E)** Conjunctival biopsy specimen from a patient with keratoconjunctivitis due to *E. hellem* demonstrating characteristic spores (arrowheads) with a single row of six cross sections of polar tube.

# Global geographic prevalence of microsporidia

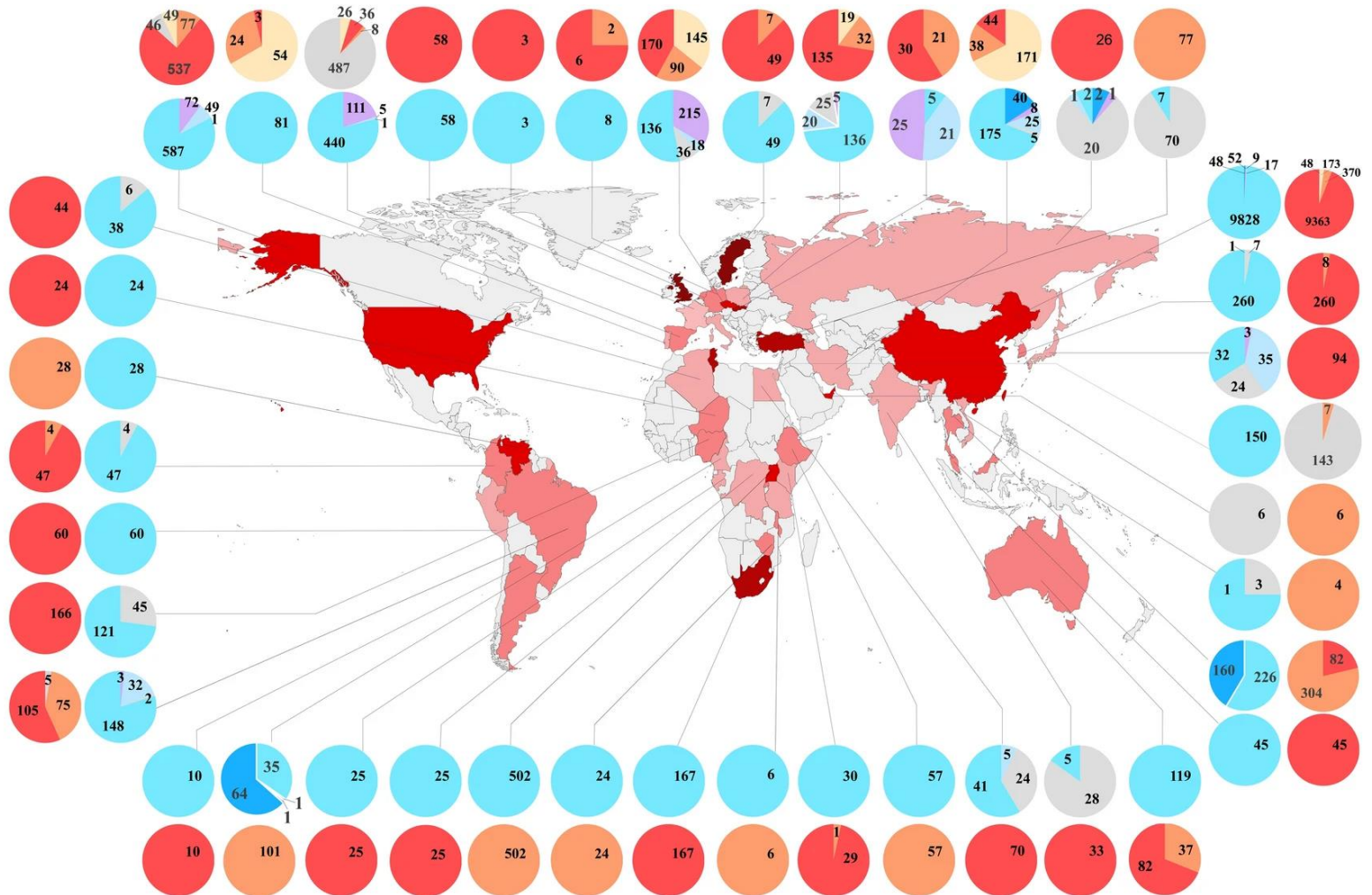
## Hosts & Water



## Pathogens

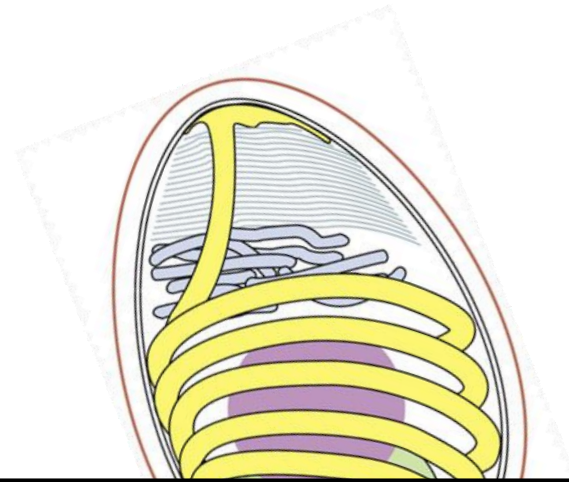
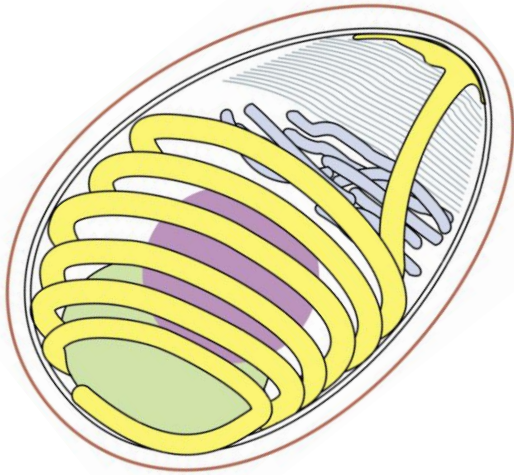


## Prevalence rate

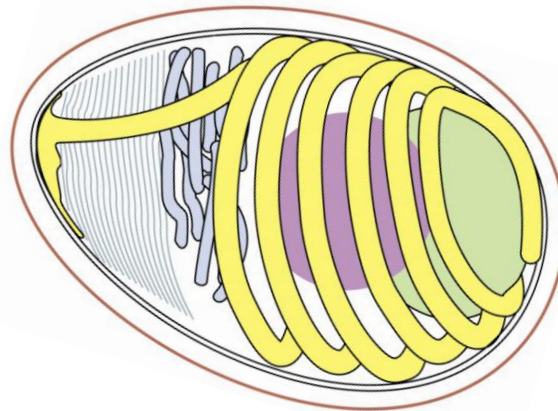


The overall prevalence of microsporidia in countries worldwide is indicated by colour depth on a world map. The outer and inner pie charts show the overall infected hosts and hosts infected by different microsporidia in each country, respectively. The number on the outer and inner pie charts represents the number of positive samples of microsporidia detected in different hosts and pathogens.





Thank you for your attention 😊



## Lectures

- ✓ Introduction: BPP 2022 I
- ✓ Euglenozoa (Excavata): BPP 2022 II
- ✓ Fornicata / Preaxostyla / Parabasala (Excavata): BPP 2022 III
- ✓ Apicomplexa I (SAR): BPP 2022 IV
- ✓ Apicomplexa II (SAR): BPP 2022 V
- ✓ Amoebae (Excavata, Amoebozoa): BPP 2022 VI
- ✓ Ciliophora, Opalinata (SAR): BPP 2022 VII

- ✓ Pneumocystis (Opisthokonta, Fungi): BPP 2022 VIII
- ✓ Microsporidia (Opisthokonta, Fungi): BPP 2022 IX
- ⇒ **Myxozoa (Opisthokonta, Animalia): BPP 2022 X**