MICROSPORA IN FISH

Glugea, Pleistophora, Loma, Heterosporis, Kabatana, Microgemma, Tetramicra, Ichthyosporidium et al.

Overview

Microsporidia are obligate intracellular parasites which lack mitochondria and form small unicellular spores. They were long considered to be a primitive basal group of Protista, but molecular phylogenetic studies have revealed many similarities in biochemical pathways and structural components to fungi, where they are now classified. Numerous species have been described in invertebrates (especially insects) and lower (rarely higher) vertebrates. The parasites proliferate in host tissues by merogony (asexual division) followed by sporogony (often involving plasmotomy prior to sporoblastogenesis). Developmental stages may be monokaryotic (single nucleus) or diplokaryotic (paired nuclei) and sporonts may be surrounded by a membranous sporophorous vesicle (pansporoblast) or lie free in the host cell cytoplasm. All spores contain a unique coiled polar tube which can be extruded to inject the infective sporoplasm into host cells. Infections may be disseminated throughout host tissues or they may cause focal lesions and inflammation involving cysts, granulomas or tumour-like xenomas (enlarged host cells). Various species are found in insects (some causing dysentery in honey bees), crustaceans (some denaturing muscles), fish (some forming lesions or deformities) and even humans (some causing diarrhoea, myositis, encephalitis or corneal lesions).

Classification

Domain:	Eukaryota (membrane-bound nucleus)
Supergroup:	Amorphea (unikonts with single flagellum, or nonflagellated amoebae)
Kingdom:	Fungi (with chitinous walls, includes microsporidia)
Division:	Microsporidia (form unicellular spores, with coiled polar tubes, amitochondriate, all parasitic)
Class:	Microsporea (polar filament well-formed, oval spores)
Order:	Microsporida (polaroplast present)
Suborder:	Pansporoblastina (sporophorous vesicle present)
Family:	Glugeidae (all stages monokaryotic, numerous sporoblasts formed in sporophorous vesicles)
Genera:	*Glugea/Pleistophora/Loma/Heterosporis* (parasitic in tissues of fish)
Suborder:	Apansporoblastina (sporophorous vesicle absent)
Family:	Unikaryonidae (all stages monokaryotic, in cell cytoplasm or in parasitophorous vacuole)
Genera:	*Kabatana/Microgemma/Tetramicra/Ichthyosporidium* (parasitic in tissues of fish)
Species:	various species cause lesions, cysts or xenomas in freshwater and marine fish

Parasite biodiversity and host range: Microsporidia possess a remarkable apomorphic adaptation to life as intracellular parasites, their unicellular spores have polar tubes coiled up inside which can be forcibly everted to penetrate host cells and inject their infective germs (sporoplasm). Microsporidia have reduced cellular complexity, they have small genomes, they are amitochondriate and they have metabolomes partway between 'prokaryotes' (archae- and eu-bacteria) and eukaryotes (nucleated cells). They were long considered to be a primitive basal group of Protista but molecular phylogenetic studies have revealed many similarities in nuclear genes, biochemical pathways and structural components to fungi, where they are now classified.

The systematics of microsporidia has progressed over decades from phenotypic classifications based on spore morphology, developmental cycles and host range to genotypic classifications based on comparative gene sequences. Classical studies divided the microsporidia into those with or without an envelope (sporophorous vesicle) around sporoblasts (Pansporoblastina and Apansporoblastina respectively) and recognized families on the basis of vegetative growth, genera on the process of spore formation and species on the basis of spore morphology. The subsequent inclusion of ultrastructural features led to the recognition of three major assemblages: 'primitive' groups with rudimentary polar tubes and no polaroplasts; 'intermediate' groups with short polar tubes and rudimentary polaroplasts; and 'higher' groups with well-developed polar tubes and polaroplasts. Families continued to be identified on the basis of type of reproduction (merogony and sporogony) and nuclear condition (mono- or diplo-karyotic). More recently, chromosome cycles were used to separate microsporidia into Dihaplophasea (diplokaryon in some phase of life-cycle) and Haplophasea (unpaired nuclei in all life-cycle stages). It is thought that the developmental cycle of Dihaplophasea involves a pairing of gametes which proliferate and undergo haplosis either by meiosis (order Meiodihaplophasida) or by nuclear dissociation (order Dissociodihaplophasida), whereas the development of Haplophasea is entirely haplophasic. Molecular phylogenetic studies, however, have not provided good support for conventional classifications but have separated representative species into three major clades mostly correlated with host habitat: including the Aquasporidia (in freshwater insects, crustaceans and bryozoans), Marinosporidia (in marine fish and crustaceans) and Terresporidia (in terrestrial insects and vertebrates). Although many taxa have yet to be analysed and classified, most clades are polyphyletic with conventional genera split within and between clades. It is not known whether this reflects an evolutionary history of parasites switching hosts or habitats, or even hosts switching habitats: events quite likely to have occurred considering that many invertebrate hosts have aquatic origins and many have retained aquatic stages in their life-cycles. Further studies are required to reconcile phenotypic and genotypic classifications.

Numerous microsporidian species have been described in invertebrates (especially insects and crustaceans) and lower (rarely higher) vertebrates. Over 1,400 species belonging to ~200 genera have been described; with some 800 species in 109 genera infecting insects, 400 species in 62 genera infecting crustaceans, 120 species in 26 genera infecting fish, 17 species in 11 genera infecting mammals, 5 species in 3 genera infecting birds, reptiles and amphibians, 40 species in 27 genera infecting invertebrates (8 genera in annelids, 7 in arachnids, 4 in molluscs, 4 in bryozoans, 2 in ciliates, one in helminths and one in kinorhynchs) and another 20 species in 14 genera being hyperparasitic in other parasites (8 genera in gregarines, 3 in trematodes, 2 in myxozoans and one in mesozoa). The key characteristics of the microsporidian genera are tabulated below:

Microsporidian genera	**Hosts**	**No. nuclei (meronts, spores)** [1= monokaryotic, 2 = diplokaryotic, 1-2 = both]	**Spore types** [1 = monomorphic, 2 = dimorphic, 3 = polymorphic]	**Intracellular location** [CY = cytoplasm, NU = nucleoplasm, PV = parasitophorous vacuole, SP = sporophorocyst, SV = sporophorous vesicle]	**Spores per SV** [n = numerous, na = not applicable]	**Xenoma formation**
Abelspora	decapoda	1,1	1	PV	2	xenoma?
Aedispora	diptera	1-2,1-2	2	PV, SV	2-8	absent
Agglomerata	branchiopoda	1,1	1	SV	8-32	absent
Agmasoma	decapoda	2,1	?	SV	8	absent
Alfvenia	copepoda, maxillopoda	2,1	1	SV	1-2	absent
Alloglugea	anura	1,1	1	PV	na	xenoma
Amazonspora	fish	1,1	1	CY	na	xenoma
Amblyospora	copepoda, amphipoda, maxillopoda, diptera	2,1-2	3	SV	8	absent
Ameson	decapoda, diptera	2,1	1	CY	na	absent
Amphiacantha	hyperparasitic in gregarines in polychaetes	1,1	1	SV	n	absent
Amphiamblys	hyperparasitic in gregarines in polychaetes	1,1	1	SV	32-50	absent
Andreanna	diptera	2,2	1	PV, SV	8	absent
Anisofilariata	diptera	1,1	1	SV	2-16	absent
Anncaliia	diptera, coleoptera, humans	2,2	1	CY	na	absent
Antonospora	hymenoptera, psocoptera	?,2	1	CY	na	absent
Areospora	decapoda	1,1	1	SV	8	xenoma
Auraspora	collembola	2,1-2	2	CY, SV	n	absent
Bacillidium	oligochaeta, thysanura, diptera	2,2	1	CY	na	absent
Baculea	branchiopoda	1,1	1	PV, SV	n	absent
Becnelia	heteroptera	1,1	2	SV	8	absent
Berwaldia	branchiopoda	1,1	1	SV	1	absent
Binucleata	branchiopoda	1-2,1	1	SV	8	absent
Binucleospora	ostracoda	2,2	1	PV	na	absent
Bohuslavia	diptera	2,1	1	SV	8-16	absent
Brachiola	diptera, humans	2,2	1	CY	na	absent
Bryonosema	bryozoa	2,2	1	CY	na	absent
Burkea	oligochaete	1,1	1	PV	na	absent
Burunella	hymenoptera	1-2,1-2	2	PV, SV	8	absent
Buxtehudea	thysanura	1,1	1	PV	na	absent
Campanulospora	diptera	2,2	1	PV	na	absent
Canningia	coleoptera	1,1	1	CY	na	absent
Caudospora	diptera	2,2	1	CY	na	absent
Chapmanium	decapoda, diptera	2,1		SV	8	absent
Chytridiopsis	coleoptera	1,1	1	PV, SV	n	absent
Ciliatosporidium	ciliophora	1,1	1	CY	na	absent
Coccospora	diptera	2,1	1	SV	8	absent
Cougourdella	copepoda, maxillopoda, diptera	1,1	1	SV	4	absent

Crepidulospora	diptera	1,1	1	CY	na	absent
Crispospora	diptera	?,1-2	2	PV	na	absent
Cristulospora	diptera	2,1-2	2	CY, SV	8	absent
Cryptosporina	acari	1,1-2	1	SV	8	absent
Cucumispora	amphipoda	2,2	1	CY	na	absent
Culicospora	diptera	1-2,1-2	2	PV, SV	2-8	absent
Culicosporella	diptera	1-2,1-2	3	PV, SV	2-8	absent
Cylindrospora	diptera	2,1	1	PV, SV	8	absent
Cystosporogenes	lepidoptera	1,1	1	SV?	<60	absent
Dasyatispora	elasmobranch	1,1	1	SV	n	absent
Desmozoon	maxillopoda, copepoda, fish	1-2,1	1	CY	na	absent
Desportesia	hyperparasitic in gregarine in echiurid	1,1	1	CY, SV	32	absent
Dictyocoela	amphipod	2,1-2	1	SV	8	absent
Dimeiospora	diptera	1,1	2	SV	8	absent
Duboscqia	branchiopoda, copepoda, isoptera, diptera	1,1	1	SV	16	absent
Edhazardia	diptera	2,1-2	3	PV, SV	1-8	absent
Encephalitozoon	maxillopoda, orthoptera, acari, birds, mammals (incl. humans)	1,1	1	PV	na	absent
Endoreticulatus	lepidoptera, coleoptera, decapoda	1,1	1	PV	na	absent
Enterocytozoon	mammals (inc. humans), fish	1,1	1	CY	na	absent
Enterospora	decapoda, fish	2,2	1	CY, NU	na	absent
Episeptum	trichoptera	1,1	1	SV	4	absent
Euplotespora	ciliophora	1,1-2	1	SV	1	absent
Evlachovaia	diptera	2,1-2	2	PV, SV	2	absent
Facilispora	maxillopoda, copepoda	1,1	1	CY	na	absent
Fibrillanosema	amphipoda, branchiopoda	1,1	1	CY	na	absent
Flabelliforma	copepoda, ostracoda, cladocera, diptera	1,1	1	CY, SV	n	absent
Geussia	hyperparasitic in gregarine of ephemeroptera	?		SV	6-8	absent
Glugea	amphipoda, fish	1,1	1	SV	n	xenoma
Glugoides	branchiopoda	1,1	1	PV, SV	16	absent
Golbergia	diptera	2,1-2	2	CY	na	absent
Gurleya	branchiopoda, copepoda, decapoda, cladocera, diptera, ephemeroptera, isoptera, lepidoptera, odonata, trichoptera	1,1	1	SV	4	absent
Gurleyides	branchiopoda	?	2	SV	1,4	absent
Hamiltosporidium	branchiopoda	1,2	2	SV	8	absent
Hazardia	diptera	1-2,1-2	2	CY	na	absent
Helmichia	diptera	2,1-2	1	SV?	8	absent
Hepatospora	decapoda	1,1	1	PV	na	absent
Hessea	diptera	2,1-2	1	SV	n	absent
Heterosporis	fish, seasnakes	1,1	3	SP, SV	n	absent
Heterovesicula	orthoptera	2,1-2	2	SV	8-n	absent
Hirsutosporos	diptera	2,2	1	CY	na	absent
Holobispora	copepoda, maxillopoda	?,1	1	CY	na	absent
Hrabyeia	oligochaeta	2,2	1	CY	na	absent
Hyalinocysta	diptera, copepoda	2,1	1	SV	8	absent
Ichthyosporidium	fish	2,2	1	PV	na	xenoma
Inodosporus	decapoda	2,1	1	SV	8	absent
Intexta	acari	1,1	2	PV	na	absent

Intrapredatorus	diptera	2,1-2	3	SV	8	absent
Issia	trichoptera, diptera	2,2	2	SV?	2	absent
Janacekia	diptera, coleoptera	2,1-2	1	SV	1	absent
Jirovecia	fish, oligochaete	2,2	1	CY, PV?	na	xenoma
Jiroveciana	oligochaete	1,1	1	PV	na	absent
Johenrea	orthoptera	1,1	1	SV	8,16	xenoma
Kabatana	fish	1,1	1	CY	na	absent
Kneallhazia	hymenoptera	1-2,1	1	SV	8	absent
Kinorhynchospora	kinorhyncha	1,1	2	SV	n	absent
Krishtalia	diptera	2,1-2	2	CY	na	absent
Lanatospora	branchiopoda, maxillopoda, copepoda	1,1	1	SV	6-16	absent
Larssonia	branchiopoda	1,1	1	SV	4-32	absent
Larssoniella	lepidoptera	1,1	1	CY	na	absent
Liebermannia	orthoptera	2,2	1	PV	na	absent
Loma	fish	1,1	1	SV	4	xenoma
Marssoniella	maxillopoda	1,1	2	SV	4-8	absent
Merocinta	diptera	2,1-2	2	PV	na	absent
Metchnikovella	hyperparasitic in gregarine in polychaete	1,1	1	PV, SV	8-32	absent
Microfilum	fish	1,1	1	CY	na	xenoma
Microgemma	fish	1,1	1	PV	na	xenoma
Microsporidium (often used for *species inquirenda, incertae sedis*)	branchiopoda, copepoda, cirripedia, isopoda, amphipoda, mollusca, insecta, fish, mammals (incl. humans)	?	?	?	?	?
Mitoplistophora	ephemeroptera	1,1	1	PV	na	absent
Mockfordia	psocoptera	?,1	1	PV	na	absent
Mrazeckia	isopoda	2,2	1	CY	na	absent
Multilamina	isoptera, diptera	1,1	1	SV	1	absent
Myospora	decapoda	2,2	1	CY	na	absent
Nadelspora	decapoda	1,1	1	CY	na	absent
Napamichum	diptera, acari	2,1	1	SV	8	absent
Nelliemelba	copepoda, maxillopoda	1,1	1	SV	1	absent
Nematocida	nematode	1,1	1	PV	na	absent
Neoflabelliforma	oligochaete, hyperparasitic in myxozoa in oligochaete	1,1	1	SV?	?	absent
Neonosemoides	fish	2,1	1	CY	na	xenoma
Neoperezia	diptera	2,1	2	SV	2	absent
Nolleria	siphonoptera	1,1	1	PV, SV	n	absent
Norlevinea	branchiopoda	1,1	1	SV	4	absent
Nosema	branchiopoda, copepoda, decapoda, amphipoda, mollusca, hymenoptera, lepidoptera, acari, fish, mammals (incl. humans); hyperparasitic in myxozoa in fish, trematodes in snails, oysters, fish	2,2	2	CY	na	absent
Nosemoides	branchiopoda, fish, hyperparasitic in gregarine in nemertean	1,1	1	CY	na	xenoma
Novothelohania	diptera	1,1	1	SV	8	absent
Nucleospora	fish	1,1	1	NU	na	absent
Nudispora	odonata	2,1	1	CY	na	absent
Obruspora	fish	1,1	1	CY	na	xenoma
Octosporea	branchiopoda, isopoda, amphipoda, cladocera, diptera, ephemeroptera,	2,2	1	SV	8	absent

	hemiptera, lepidoptera, collembola					
Octotetraspora	diptera	2,1	1	SV	4,8	absent
Oligosporidium	acari, opiliones	1,1	2	CY	na	absent
Ordospora	branchiopoda	1,1	1	PV	na	absent
Ormieresia	decapoda	2,1	1	CY, SV	8	absent
Orthosomella	lepidoptera, coleoptera	1,1	1	CY	na	absent
Ovavesicula	coleoptera	2,1	1	CY, SV	32	absent
Ovipleistophora	fish, hyperparasitic in trematode in fish	1,1	2	CY, SV	na	absent
Pankovaia	ephemeroptera	1,1	11	CY, SV	1	absent
Paradoxium	decapoda	1-2,1	1	CY	na	absent
Paraepiseptum	trichoptera	1,1	1	SV	4	absent
Parahepatospora	decapod	1,1	1	PV	na	absent
Paranosema	coleoptera, orthoptera	1-2,2	1	CY	na	absent
Paranucleospora	maxillopoda, fish	1-2,1-2	2	CY, NU	na	absent
Parapleistophora	diptera	1,1	1	SV	48-64	absent
Parastempellia	diptera	2,1-2	2	SV	4,8,16	absent
Parathelohania	maxillopoda, diptera	2,1-2	2	SV	8	absent
Pegmatheca	diptera, tricoptera	2,1	1	SV	8	absent
Perezia	branchiopoda, decapoda, lepidoptera, coleoptera, hymenoptera, orthoptera, plus hyperparasitic in gregarine of tunicate	2,1	1	CY	na	absent
Pernicivesicula	diptera	2,1	1	SV	24-64	absent
Pilosporella	diptera	2,1-2	2	SV	8	absent
Pleistophora (Plistophora)	branchiopoda, copepoda, decapoda, blattaria, coleoptera, diptera, lepidoptera, orthoptera, mollusca, fish, mammals (incl. humans	1,1	2	SV	n	absent
Pleistophoridium	hyperparasitic in gregarine in ephemeroptera	1,1	1	CY, PV	na	absent
Polydispyrenia	diptera	2,1	2	PV, SV	n	absent
Potaspora	fish, decapod	1,1	1	CY	na	xenoma
Pseudoloma	fish	1,1	1	SV	16	atypical xenoma
Pseudonosema	bryozoa	2,2	1	CY	na	absent
Pseudopleistophora	lepidoptera, polychaete	2,2	1	PV, SV?	n	absent
Pulicispora	siphonoptera	1,1-2	1	SV	8,16,32	absent
Pyrotheca	copepoda, maxillopoda	1,1	1	CY, SV	4	absent
Rectispora	oligochaeta	2,2	1	CY	na	absent
Resiomeria	odonata	2,1	1	SV	8	absent
Ringueletium	diptera	2,2	1	CY	na	absent
Schroedera	bryozoa	1-2,2	1	CY	na	xenoma
Scipionospora	diptera	2,2	1	SV	4	absent
Semenovaia	diptera	?,1-2	2	CY	na	absent
Senoma	diptera	2,2	1	PV	na	absent
Septata	human	1,1	1	PV	na	absent
Simuliospora	diptera	2,1	2	SV	6,32	absent
Spherospora	diptera	2,1-2	2	SV	8-32	absent
Spiroglugea	diptera	?	1	SV?	8	absent
Spraguea	fish	1-2,1-2	2	CY	na	xenoma
Steinhausia	bivalve, gastropod	1,1	1	PV, NU	na	absent
Stempellia	copepoda, amphipoda, opiliones, ephemeroptera, diptera, coleoptera, isoptera	1,1	2	PV, SV	4	absent

Striatospora	diptera	2,1	1	SV	8	absent
Systenostrema	diptera	2,1-2	1?	SV	8	absent
Tabanispora	diptera	1-2,2	2	SV	1-10	absent
Takaokaspora	diptera	1-2,1-2	2	CY, SV	?	absent
Tardivesicula	trichoptera	1,1	1	SV	16-32	absent
Telomyxa	ephemeroptera, diptera, coleoptera	1,1	1	SV	2	absent
Tetramicra	fish	1,1(2?	1	PV	na	xenoma
Thelohania	branchiopoda, copepoda, decapoda, amphipoda, diptera, collembola, ephemeroptera, hemiptera, lepidoptera, hymenoptera, odonata, trichoptera, fish	1-2,1	1	SV	8	absent
Toxoglugea	branchiopoda, diptera, plecoptera, odonata, hemiptera, homoptera	2,1	1	SV	8	absent
Toxospora	diptera	?,1-2	1	SV	8	absent
Trachipleistophora	mammals (incl. human)	1,1	1	PV, SV	2-n	absent
Trichoctosporea	diptera	2,1	2	SV	8	absent
Trichodubosquia	ephemeroptera	2,1	1	SV	16-32	absent
Trichonosema	bryozoa	2,2	1	CY	na	absent
Trichotuzetia	copepoda, maxillopoda	1,1	1	SV	1	absent
Tricornia	diptera	2,1-2	1	SV	8	absent
Triwangia	decapoda	1,1	1	SV	n	xenoma
Tubulinosema	diptera, orthoptera, hymenoptera, coleoptera, mammals (humans)	2,2	1	CY	na	absent
Tuzetia	branchiopoda, copepoda, maxillopoda, ephemeroptera	1,1	1	SV	1	absent
Unikaryon	coleoptera, acari, hyperparasitic in trematode in bivalve	1,1	1	SV	2	absent
Vairimorpha	decapod, lepidoptera, hymenoptera, diptera	2,1-2	2	CY, SV	8	absent
Vavraia	ostracoda, decapoda, diptera, coleoptera, lepidoptera	1,1	1	SV	16-64	absent
Vittaforma	mammals (humans)	2,2	1	PV	na	absent
Weiseria	diptera	2,2	1	CY	na	absent
Wittmania	hyperparasitic in mesozoan in cephalopod	1-2,2	1	CY	na	absent
Zelenkaia	trichoptera	1,1	1	SV	2	absent

Although many microsporidian species have been described on the basis of presumed host specificity, recent molecular characterization studies have demonstrated that the host ranges for some species can be very broad, encompassing not only hosts from disparate taxa (e.g. insects and mammals) but also hosts from disparate environments (e.g. marine and terrestrial). While considerable work remains to determine the host ranges and phylogenetic affinities of most microsporidia, the following text considers microsporidia from one of three perspectives associated primarily, but not exclusively, with different host groups; namely, arthropods (mainly insects and crustaceans), fish (bony and cartilaginous), and tetrapods (mammals, birds, reptiles and amphibians). This section considers the microsporidia of fish, notably those kept and managed for economic benefit; including ornamental species (aquarium trade), farmed species (freshwater and marine aquaculture) as well as wild fisheries.

Microsporidian infections have been found in both wild and farmed fish from freshwater, estuarine and marine habitats around the world. Most species appear to be oioxenous (host specific) or stenoxenous (infecting closely-related hosts), although a few have been found to be euryxenous (infecting a broad range of hosts). Many infections are disseminated throughout various host tissues, while others may cause focal space-occupying lesions which may become visible (macroscopic) as cysts or tumour-like xenomas (hypertrophic host cells). Infections have been associated with significant morbidity and mortality, especially in salmonid culture. Microsporidia of fish may be divided into two groups, comprising pansporoblastic and apansporoblastic species whose spores do or do not develop within sporophorous vesicles, respectively. Most pansporoblastic genera are monokaryotic and monomorphic (e.g. *Glugea, Loma, Pseudoloma*), although *Thelohania* forms diplokaryotic meronts and some *Pleistophora* spp. are dimorphic. A notable exception is the genus *Heterosporis* which may undergo three sporulation sequences, all enveloped within a unique sporophorocyst (parasite-derived envelope). The pansporoblastic genera *Glugea, Loma* and *Pseudoloma* form xenomas, while the remainder cause disseminated infections or tissue-bound cysts. Most apansporoblastic genera are monokaryotic and monomorphic (e.g. *Microgemma, Kabatana, Tetramicra, Potaspora, Microfilum*), although several are diplokaryotic and monomorphic (*Ichthyosporidium, Jirovecia*) or diplokaryotic and dimorphic (*Nosema, Spraguea*). Xenomas are formed by most apansporoblastic genera, except *Kabatana* and *Nosema* which form tissue cysts. Meronts of most genera develop either directly within the host cell cytoplasm or within parasitophorous vacuoles, but two notable exceptions are the genera *Nucleospora* and *Enterospora* which develop within the nucleoplasm of host cells.

Parasite species	**Spore dimensions** (mi = microspores; ma = macrospores; me = meiospores)	**Hosts**	**Location**	**Distribution**
Suborder PANSPOROBLASTINA (sporophorous vesicle (SV) present)				
Family GLUGEIDAE (monokaryotic, variable but large number spores produced)				
Genus ***Glugea*** (sporogony by multiple then binary fission, spores in SV with membrane-like wall, xenomas)				
forming large xenomas with numerous branched peripheral nuclei				
G. anomala	3.0-6.0 x 1.9-2.7 µm	freshwater fish (sticklebacks, killifish)	xenomas in connective tissues	North America, Eurasia
G. atherinae	3.5-5.7 x 2.2-2.9 µm	estuarine fish (smelt, silversides, smallmouth hardyhead, short-snout hardyhead, pikehead hardyhead, silver fish)	xenomas in connective tissues of gut, body cavity and mesenteries	Mediterranean, Australia
G. capverdensis	3.6-4.8 x 1.8-2.6 µm	marine fish (lanternfish)	xenomas in intestine, mesentery, ovary	Atlantic
G. cepedianae	4.9 x 2.3 µm	freshwater fish (shad)	xenomas in body cavity	North America
G. fennica	7.4 x 2.8 µm	freshwater fish (lingcod)	xenomas in subcutaneous tissues	Northern Europe
G. heraldi	3.6-4.5 x 1.8-2.3 µm	marine fish (seahorses)	xenomas in connective tissues	Atlantic
G. hertwigi	4.6-5.4 x 2.2-2.3 µm	marine and freshwater fish (smelts, whitefish)	xenomas in intestines, organs	Holarctic
G. pimephales	4.5-6.0 x 2.0-3.0 µm	freshwater fish (minnows)	xenomas in body cavity	North America
G. plecoglossi	5.8 x 2.1 µm	freshwater fish (ayu sweetfish, rainbow trout)	xenomas in visceral organs	Japan
G. sardinellensis	5-5.5 x 2.5-3 µm	marine fish (round sardinella)	xenomas in connective tissue	Tunisia
G. stephani	3.0-3.9 x 1.5-1.7 µm	marine fish (plaice, flounder, sole, turbot)	xenomas in intestines, liver	Europe, North Atlantic
G. vincentiae	ma 7.5-12.0 x 2.0-4.0 µm (12-14 coils) mi 4.5-6.0 x 2.0-2.7 µm (12-14 coils)	marine fish (cardinalfish)	xenomas in subcutaneous tissues	Australia
forming small xenomas with a centrally located nucleus				
G. berglax	6.4 x 2.7 µm	marine fish (rattails)	xenomas in intestines, gall bladder	Canada
G. destruens	3.6 x 2 µm	marine fish (dragonets)	xenomas in muscles	France
G. luciopercae (syn. *G. dogieli*)	4.5 x 2.1 µm	freshwater fish (perch, pike)	xenomas in intestines	Eurasia
G. machari (syn. *Octosporea*)	3.7 x 1.1 µm	marine fish (sea bream)	xenomas in liver	Adriatic
G. tisae	4.5 x 2.4 µm	freshwater fish (catfish)	xenomas in intestines	Europe

G. truttae	5 x 1.5 µm	freshwater fish (brown trout)	yolk sac	Europe
Genus ***Pseudoloma*** (atypical xenomas)				
P. neurophila	4.8-5.9 x 2.3-3.1 µm (13-16 coils)	freshwater fish (zebrafish)	muscles, nervous tissue	North America
Genus ***Pleistophora*** (polysporoblastic sporogony, SV, macro-, medium-, micro-spores (ma, me, mi)				
species infecting skeletal muscle				
P. atretti	4.8-5.4 x 1.8-2.5 µm	freshwater snake	muscles	India
P. duodecimae	ma 6.2 x 3.3 µm mi 4.3 x 2.7 µm	marine fish (rattails)	cysts in muscles	Southern Atlantic
P. ehrenbaumi	ma 7.5 x 3.5 µm mi 3.0 x 1.5 µm	marine fish (wolf fish)	cysts in muscles	North Sea
P. finisterensis	4.0 x 2.0 µm	marine fish (blue whiting)	cysts in muscles	Europe
P. hippoglossoides	4.7 x 2.7 µm	marine fish (flatfish, sole)	cysts in muscles	North Sea
P. hyphessobryconis	ma 6.5-7.0 x 4.0 µm (34 coils) mi 4.0-6.0 x 2.0-3.3 µm (up to 34 coils)	freshwater fish (tetras, danios, barb, goldfish)	cysts in muscles, internal organs, testes, subcutaneous	worldwide (aquarium trade)
P. littoralis	ma 7.7 x 3.8 µm (39 coils) mi 3.9 x 2.3 µm (17 coils)	marine fish (blennies)	cysts in muscles	England
P. macrozoarcidis	ma 8 µm mi 5.5 x 3.5 µm	marine fish (ocean pout)	cysts in muscles	North Atlantic
P. mulleri		marine amphipod (scuds)	muscles	Atlantic
P. typicalis	ma 6.3-8.3 x 3.0-3.3 µm (33-39 coils) mi 3.0-5.6 x 1.5-3.0 µm (10-22 coils)	freshwater fish (sculpins, blennies, sticklebacks)	cysts in muscles	Europe
low organ specificity				
P. aegyptiaca	1.5-2.7 x 1.2-1.8 µm (26-32 coils)	marine fish (lizardfish)	cysts in peritoneum	Red Sea
P. dammami	ma 6.0 x 3.0 µm (20-24 coils) mi 2.5 x 2.0 µm (6 coils)	marine fish (lizardfish)	gi tract	Arabian Gulf
P. pagri	1.5-2.7 x 1.2-1.8 µm (3-5 coils)	marine fish (bream)	viscera	Egypt
P. peponoides	3.6 x 2.0-2.3 µm	freshwater fish (sleeper)	connective tissue	Russia
P. priacanthicola	ma 8.2 x 3.8 µm me 5.4 x 3.1 µm mi 2.9 x 1.9 µm	marine fish (bigeyes)	cysts in gi tract, heart, gonads, visceral organs	South China Sea
species infecting ovaries transferred to new genus *Ovipleistophora*				
O. longifilis (syn. *P. longifilis*)	ma 12 x 6 µm mi 3 x 2 µm	freshwater fish (barbels, roaches)	spore masses in ovaries, ova	Europe, Asia
O. mirandellae (syn. *P. mirandellae, P. oolytica, P. elegans*)	ma 7.3-12.0 x 3.5-6.4 µm me 4.0-6.5 x 2-3.5 µm mi 3.0-7.5 x 1.5-4.0 µm	freshwater fish (cyprinids, esp. barbels, bream, dace, roach, pike, huchen, bleak)	spore masses in ovaries, ova	Europe, Asia
O. ovariae (syn. *P. ovariae*)	8.4 x 4.2 µm	freshwater fish (shiners, minnows)	spore masses in ovaries, ova, liver, kidneys	North America
Genus ***Dasyatispora*** (species in rays)				
D. levantinae	3.8-4.3 x 2.6-2.8 µm (9-12 coils)	marine elasmobranchs (stingrays)	muscle	Turkey
Genus ***Loma*** (polysporoblastic sporogony, up to 8 spores in SV, xenomas up to 1mm with central nucleus)				
L. acerinae (syn. *Glugea*)	2.3-4.5 x 1.6-3.4 µm (10-14 coils)	freshwater fish (perch, gobies)	xenomas in connective tissues, intestines	Central Europe
L. branchialis (syn. *L. morhua, Nosema, Glugea*)	4.8 x 2.3 µm	marine fish (haddock, cod)	xenomas in gills, viscera	North Atlantic
L. camerouensis		freshwater (Nile tilapia)	xenomas in gut	Africa

L. dimorpha	4.5 x 1.9 µm	marine fish (gobies)	xenomas in intestines	Mediterranean
L. embiotocia	4.0-5.0 x 2.0-3.0 µm (14-18 coils)	marine fish (perch)	xenomas in gills	Canada
L. kenti	4.0-5.0 x 1.9-4.4 µm (14-16 coils)	marine fish (tomcod)	xenomas in gills, viscera	Canada
L. myrophis		freshwater fish (worm eel)	xenomas in midgut	South America
L. psittaca	4.2 x 2.8 µm (10-12 coils)	freshwater fish (pufferfish)	xenomas in gi tract	Amazon River
L. salmonae	3.0-7.5 x 1.6-2.8 µm	freshwater fish (trout)	xenomas in gills	North America, Japan, Europe
Genus ***Heterosporis*** (stages encased within sporophorocyst wall, polysporoblastic sporogony in SV)				
H. anguillarum	ma 6.7-9.0 x 3.3-5.3 µm (33-46 coils) mi 2.8-5.0 x 2.0-2.9 µm (33-36 coils)	marine fish (eels) [also recorded in garter snakes?]	muscles, skin	Japan, Taiwan, Korea
H. finki	ma 7.0-9.0 x 2.0-5.0 µm (30-36 coils) mi 3.0 x 1.5 µm (7 coils)	freshwater fish (angelfish)	muscles, connective tissues	Germany, France (aquaria)
H. saurida	ma 5.0-6.0 x 3.0-3.8 µm (20-21 coils) mi 3.0-3.8 x 1.5-2.5 µm (5-6 coils)	marine fish (lizardfish)	muscles	Arabian Gulf
H. schuberti	ma 5.4-8.8 x 2.9-4.9 µm mi 3.4-4.9 x 2.4-3.4 µm	freshwater fish (mouthbrooder, Jumbie tetra)	muscles	worldwide (aquaria)
H. sutherlanda	4.8-6.3 x 3.2-3.6 µm	freshwater fish (walleye, yellow perch, northern pike)	muscles	North America
Family THELOHANIIDAE (meronts usually diplokaryotic, spores monokaryotic, 8 spores in SV)				
Genus ***Thelohania*** (uninucleate spores with isofilar polar tube)				
T. baurei	5.4 x 2.7 µm	marine fish (sticklebacks)	spore masses in ovaries	Baltic Sea
T. ovicola	7.0 x 5.0 µm	freshwater fish (whitefishes)	spore masses in ovaries	Switzerland
Suborder APANSPOROBLASTINA (sporophorous vesicle (SV) absent)				
Family UNIKARYONIDAE (monokaryotic, diplo- to poly-sporoblastic sporogony)				
Genus ***Nosemoides*** (polysporoblastic sporogony, sporoblasts in rosette formation)				
N. syacii	2.9-4.9 x 1.8-2.7 µm (4-5 coils)	marine fish (turbot)	xenomas in gut, liver	Senegal
N. tilapiae	2.5-3.0 x 1.5-2.0 µm (4-5 coils)	freshwater fish (cichlids)	xenomas in gills, mesentery	Africa
Genus ***Microgemma*** (sporogonial plasmodia produce sporoblasts by exogenous budding)				
M. carolinus	3.8 x 2.4 µm (8-9 coils)	marine fish (carangids)	xenomas in liver	Brazil
M. caulleryi (syn. *Glugea*)	2.6 x 1.2 µm (7-9 coils)	marine fish (greater sand eel)	xenomas in liver	Spain
M. hepaticus	4.2 x 2.4 µm (7-10 coils)	marine fish (mullet)	xenomas in liver	England
M. ovoidea (syn. *Glugea*)	2.5 x 1.5 µm	marine fish (rockling, bandfish, mullet)	xenomas in liver	Europe
M. tincae	3.6 x 1.2 µm (9 coils)	marine fish (wrasse)	xenomas in liver	Tunisia
Genus ***Kabatana*** (infects muscles, no xenoma formation)				
K. arthuri (syn. *Microsporidium*)	3.1 x 2.1 µm	freshwater fish (catfish)	muscles	Thailand
K. newberryi	2.8 x 1.9 µm (9-10 coils)	marine fish (gobies)	muscles	North America
K. rondoni	4.2 x 2.4 µm (8-10 coils)	freshwater fish (knifefish)	muscles	Amazon River
K. takedai	2.8-4.9 x 1.7-2.3 µm	marine fish (salmonids)	muscles, heart	Japan

(syn. *Glugea, Nosema, Microsporidium*)				
Genus ***Tetramicra*** (tetrasporoblastic sporogony, xenomas with single central nuclei)				
T. brevifilum	3.7-4.8 x 2.0-2.7 µm (3-4 coils)	marine fish (turbot, anglerfish)	xenomas in connective tissues, muscles	England, Spain
Genus ***Unikaryon*** (disporoblastic sporogony)				
U. nomimoscolexi	3.4 x 1.5 µm (6-8 coils)	hyperparasitic in cestode (*Nomimoscolex,* but probably *Proteocephalus*) in gut of freshwater fish (whitehead catfish)	parenchymal cells	Africa
Genus ***Potaspora*** (tetrasporoblastic sporogony, spore with manubrium, xenoma formation)				
P. aequidens	3.4 x 1.9 µm (8-9 coils)	freshwater fish (cichlid)	xenomas in muscles	Brazil
P. morhaphis	2.8 x 1.5 µm (9-11 coils)	freshwater fish (needlefish)	xenomas in body cavity	Amazon River
Genus ***Microfilum*** (tetrasporoblastic sporogony, spore with manubrium, xenoma formation)				
M. lutjani	4.5 x 2.6 µm	marine fish (snapper)	xenomas in gills	Africa
Family SPRAGUEIDAE (dimorphic species)				
Genus ***Spraguea*** (two developmental sequences, monokaryotic forming oval spores, diplokaryotic forming curved spores)				
S. gastrophysus	3.4 x 1.7 µm (5-6 coils)	marine fish (anglerfish)	xenomas in ganglia, kidneys	Brazil
S. lophii (syn. *Nosema, Glugea, G. americanus*)	oval 4.2 x 2.5 µm (5-6 coils) curved 3.7 x 1.4 µm (3-4 coils)	marine fish (anglerfish)	xenomas in ganglion cells	Mediterranean, Atlantic
Family NOSEMATIDAE (diplokaryotic, diplosporoblastic, monomorphic)				
Genus ***Nosema*** (no xenomas, species in fish include hyperparasites of myxozoans)				
N. branchiale	6.3 x 3.5 µm	marine fish (cod)	gills, viscera	Eurasia, North America
N. ceratomyxae	ns	in myxozoan (*Ceratomyxa*) in gall bladder of marine fish (rabbitfish)	in myxozoan plasmodia	Red Sea
N. girardini	2.0-2.5 x 1.0-1.5 µm	marine fish (poeciliids)	gi tract	Brazil
N. marioni	8.0 x 3.0 µm	in myxozoan (*Ceratomyxa*) in gall bladder of marine fish (wrasses)	in myxozoan plasmodia	Mediterranean
N. notabilis	3.3 x 2.0 µm	in myxozoan (*Ortholinea*) in urinary bladder of marine fish (toadfish)	in myxozoan plasmodia	Atlantic
N. pimephales	3.8-4.4 x 1.9-3.3 µm	freshwater fish (minnows)	abdomen	Canada
N. podocotyloidis	3.1-4.0 x 1.8-3.3 µm	in trematode (Podocotyloides) in marine fish (African striped grunt)	in trematode parenchyma	Atlantic
Genus ***Ichthyosporidium*** (forming lobose xenomas)				
I. giganteum (syn. *Pleistophora gigantea*)	6.0-7.3 x 4.0-5.2 µm (32-46 coils)	marine fish (wrasses, spot croaker)	xenomas in subcutaneous tissues, liver	Atlantic, Black Sea
I. hertwigi	6.0 x 4.5 µm	marine fish (wrasses)	xenomas in gills	Black Sea
I. weissii	ma 7.1-10.1 x 4.8-6.2 µm (36-50 coils) mi 4.9-7.0 x 2.9-5.3 µm (36-50 coils?))	marine fish (gobies)	xenomas in gonads	North America
Family BACILLIDIIDAE (diplokaryotic, disporoblastic, cylindrical spores)				
Genus ***Jirovecia*** (basal polar tube thickened into rod)				
J. piscicola	20.0 x 6.0 µm	marine fish (whiting)	skin	France, India
Family ENTEROCYTOZOONIDAE (monokaryotic, sporogony with formation of electron-dense discs)				
Genus ***Nucleospora*** (multinucleate merogony and sporogony, stages in host cell nuclei)				

N. salmonis (syn. *Enterocytozoon salmonis*)	2.0 x 1.0 µm (8-12 coils)	marine fish (salmonids)	haematopoietic cells, kidney (intranuclear)	Atlantic
N. secunda	1.6 x 0.8 µm (4-5 coils)	freshwater fish (redfin notho)	enterocytes (intranuclear)	Europe
Genus ***Enterospora*** (develop in nucleoplasm)				
E. nucleophila	1.7 x 1.1 µm (5-6 coils)	marine fish (bream)	enterocytes	Spain
Microspora *incertae sedis*				
Genus ***Amazonspora*** (xenoma formation)				
A. hassar	2.7 x 1.8 µm	freshwater fish (thorny catfish)	xenomas in gills	South America
Genus ***Neonosemoides*** (two merogonial phases in host cell cytoplasm, one diplokaryotic, one monokaryotic)				
N. tilapiae	2.5-3.2 x 1.5-2.2 µm (4-5 coils)	freshwater fish (cichlids)	xenomas in gut, gills	Africa

Various reports have also not been able to assign microsporidian species to particular genera, so they have been reported as belonging to a collective group called *Microsporidium s.l.* (*sensu latu* = in the broadest sense).

Parasite species	Spore dimensions	Hosts	Location	Distribution
Genus ***Microsporidium*** (collective group, depository for unidentified microspora)				
forming xenomas				
M. aurata	1.5-2.5 x 1.0-2.0 µm (5-11 coils)	marine fish (bream)	xenomas in muscles, viscera	Egypt
M. cotti	9 x 3 µm	marine fish (sculpins)	xenomas in testes	France
M. hepaticum	4.2-5.0 x 2.1-2.9 µm (6-10 coils)	marine fish (greenback flounder, bridled leatherjacket, sixspine leatherjacket, brown-striped leatherjacket)	xenomas in liver	Australia
M. merluccius	2.5-3.3 x 1.8-2.1 µm (11-12 coils)	marine fish (hake)	xenomas in muscles	Namibia
not forming xenomas				
M. ovoideum	2.5 µm	marine fish (cod, bandfish, mullet)	liver	France
M. prosopium	5.0-7.0 x 3.0-4.0 µm (13-16 coils)	freshwater fish (whitefish)	muscle	Canada
M. rhabdophilia (possibly *Enterocytozoon salmonis*)	2.3-3.5 x 0.8-1.2 µm	marine fish (salmonids)	skin, gills, gi tract (intranuclear)	North America
M. seriolae (possibly *Kabatana*)	3.3 x 2.2 µm	marine fish (yellowtails)	muscles (Beko = myoliquefaction)	Japan
M. sulci (syn. *Cocconema, Pleistoiphora*)	2.5 x 2.5 µm	freshwater fish (sturgeon, sterlet)	ovaries	Volga and Danube Rivers

Parasite morphology: Microsporidia form three sequential developmental stages: meronts, sporonts and spores. The unique unicellular spores are spherical, ovoid or cylindrical, most ranging in length from 2-8 µm. They are encased within tough chitinous walls comprising a thin electron-dense exospore and a thicker electron-lucent endospore. Mature spores possess an elongate polar tube coiled up inside; most tubes being isofilar (of uniform diameter) although some are anisofilar (tapered, showing a reduction in diameter over length). The polar tubes are attached to an anterior anchoring disc enveloped by a membranous polar sac. For most microsporidian genera, the wall of the polar tube and its central canal are inserted into the polar sac, but for chytridiopsid genera (*Buxtehudea, Chytridiopsis* and *Nolleria*), only the central canal surrounded by a honeycomb layer is inserted into the polar sac. The anterior section of the polar tube is straight and surrounded by the polaroplast, which may be lamellar, tubular or both. Mature spores (especially in piscine microsporans) contain a prominent posterior vacuole (often visible by light microscopy) and an amoeboid nucleated sporoplasm. Spores may be monokaryotic (uninucleate) or diplokaryotic (with two closely-appressed nuclei). Many microsporidia form only one type of spore (monomorphic), while others are heterosporous (dimorphic or polymorphic) forming several different types (usually micro-spores and macro-spores, sometimes meio-spores). Following host cell invasion, parasites undergo asexual merogony (schizogony) and then 1-3 sporulation (sporogony) sequences forming sporoblasts (sporoblastogenesis) which mature to form infective spores (sometimes referred to as germination). Meronts are located either directly within the host cell cytoplasm (often surrounded by host endoplasmic reticulum and sometime host mitochondria) or bound within parasitophorous vacuoles (membranous envelopes of host origin). They appear as clusters of small nucleated intracellular parasites that have divided by binary or multiple fission, although several species form multinucleated plasmodial stages. Sporonts also appear as small clusters of parasitic cells but are characterized by thickened plasmalemmas due to the deposition of parasite secretions on their surface membranes. Pansporoblastic species also form an isolating envelope (thick- or thin-walled membranous sporophorous vesicle, special thick-walled sporophorocyst in the case of *Heterosporis*) whereas apansporoblastic species lie direct in the host cell cytoplasm or within parasitophorous vacuoles. Sporonts divide internally one or more times by binary or multiple fission or plasmotomy to form sporoblasts which then mature into spores. Sporonts of a few species (mostly in insects) are also thought to divide by meiosis to form uninucleate meiospores.

Site of infection: All microsporidian species are histozoic parasites with obligative intracellular development within host cells; either being in direct contact with the host cell cytoplasm (a few even occurring in the nucleoplasm) or being enclosed within parasitophorous vacuoles (membrane of host origin), sporophorocysts or sporophorous vesicles (envelopes of parasite origin). Many species exhibit tissue tropism with development occurring within particular tissues (including gut, gills, subcutis, muscles, connective tissues, visceral organs or gonads) or they may be disseminated throughout multiple tissues, some being systemic.

Pathogenesis: During development, microsporidia are metabolically dependent on host cells and are able to mobilize host cell organelles to meet their demands. Infected cells become swollen and the host cell nucleus becomes enlarged or fragmented. The parasites ultimately cause lysis of infected host cells, thereby resulting in both structural and functional deficits. Infections in tissues may be diffuse, spreading from cell to cell, or localized within cysts. Some microsporidia cause infected host cells to undergo massive hypertrophy forming tumour-like xenomas (xenoparasitic complexes, xenoparasitomes) occupying part or all of the cell volume, surrounded by a simple plasmalemma or a thick wall with surface modifications (microvilli, invagination), and containing a single central hypertrophic host cell nucleus or multiple peripheral (syncytial) nuclei. Disseminated infections, cysts and xenomas are responsible for tissue displacement/replacement, inflammation, space-occupying lesions and pressure atrophy as well as causing structural/growth deformities and disfigurement. Microsporidial spores are highly refractile due to their chitinous walls so heavy infections may cause tissue opacities and unsightly lesions. Host cell lysis often leads to cellular infiltration of the tissues (slow to develop) and sometimes granuloma formation. Host reactions may eventually lead to the destruction of spores, with both humoral and cellular immune responses implicated in parasite clearance. Surviving hosts are left with space-occupying lesions filled with connective tissue fibres, host cellular infiltrates and parasite fragments. Some infections have also been linked to post-mortem (and putatively ante-mortem) liquefaction of muscles. Infected fish have been shown to have reduced fitness, longevity and fecundity, as well as being more susceptible to predation by piscivores. Microsporidial infections therefore not only adversely affect fish health and survival but also cause significant economic losses in aquaculture due to production-limiting diseases in aquaculture as well as reducing the commercial value of wild and farmed fish due to poor quality product.

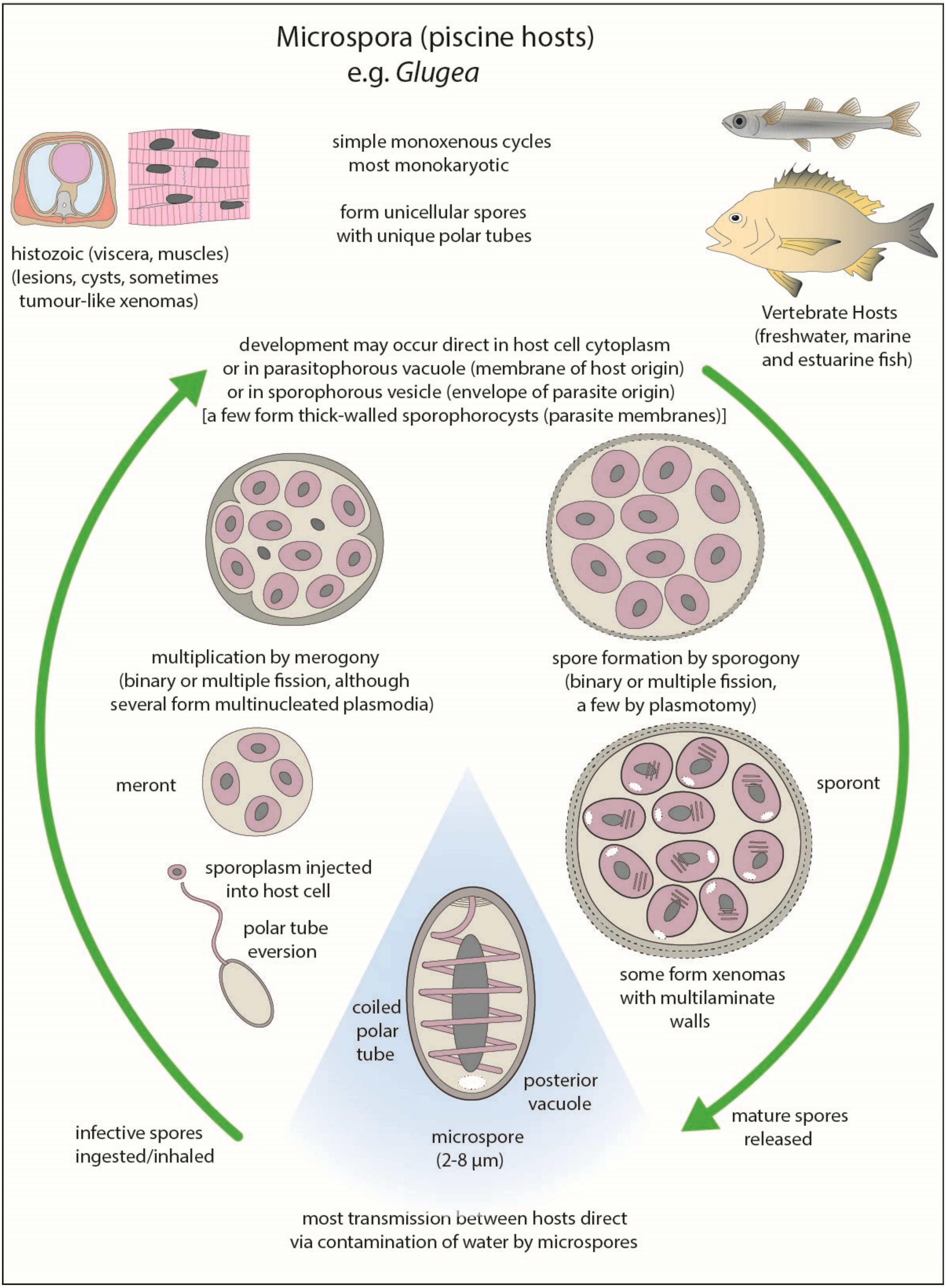
Xenoma formation has been documented in a variety of fish tissues: often progressing through 3 phases from ‘weakly-reactive’ stages (local proliferation of connective tissue leading to collagenous envelope, usually multilaminate in appearance) to ‘ripe and productive’ stages (prominent inflammation with degenerative changes in xenoma wall) to ‘granuloma involution’ stages following xenoma rupture (involving phagocytes and hyalinization of fibrous connective tissue). Several *Glugea* spp. form synchronous xenomas (enclosed parasites at similar stages of development) with multilaminate walls in submucosal intestinal cells; *Loma* spp. may form asynchronous xenomas (enclosed parasite at different stages of development) in endothelial cells throughout vascularized organs (esp. gills); *Tetramicra* spp. may form xenomas without multilaminate walls throughout the skeletal musculature; *Ichthyosporidium* spp. may form massive multilobate (syncytial-type) xenomas in connective tissues throughout the abdominal cavity and gills, and *Spraguea* spp. may form massive thin-walled xenomas in the hindbrains of fish. Non-xenoma-forming genera may also cause extensive tissue damage as they disseminate throughout host tissues. *Pleistophora* spp. often cause diffuse infections in skeletal muscles with musculature destruction or liquefaction, deformity and the production of tumour-like masses (but not granulomas). *Kabatana* spp. may cause massive infections of the trunk musculature (Beko disease) with cyst formation, fibrosis, deformities and liquefaction. *Heterosporis* spp. mainly infect the muscles of eels forming unique sporophorocysts (thick dense envelope of parasite origin). *Ovipleistophora* spp. infect oocytes and mature eggs causing macroscopic lesions with proliferative granulomatous inflammatory responses that reduce host fecundity. *Nucleospora* spp. infect the nuclei of haemoblasts (particularly lymphoblasts or plasmablasts) causing massive proliferation of immature cells (suggestive of neoplasia), often associated with plasmacytoid leukaemia in salmon. A *Paranucleospora* sp. has recently been found in copepods on salmon in association with ‘proliferative gill disease’.

Infections in fish by different microsporidial species have been associated with reduced growth rates, weight loss, inappetance, anorexia to the point of starvation, and emaciation (*Pseudoloma neurophilia* causing ‘skinny disease’). Infected fish have demonstrated unusual behaviours associated with respiratory distress and impaired swimming ability, occasionally associated with spinal curvature (scoliosis and kyphosis). Infections of the skin and subcutis can result in focal swellings, loss of body coloration, development of pale patches, bristled scales and ragged fins. Systemic infections have been associated with anaemia and myeloencephalitis, while infections of the gonads have been associated with reduced reproductive capacity (reduced fertility and fecundity) and occasionally parasitic castration (mostly involving ova, rarely testes). Infections are also more prevalent and severe in cultured fish, particularly those confined within small aquaria or ponds at higher stocking densities. Clinical infections usually involve younger fishes, as older fish either develop resistance or do not become infected because of differing feeding habits or physical environments (note that infected fish probably do not survive to older age groups). Various host defense mechanisms have been observed in experimentally and naturally infected fish; including phagocytosis of spores by macrophages, humoral responses (esp. agglutinating antibodies), mononuclear cellular responses, and even premunitive immunity (persistence of some parasites contributing to resistance to super-infection, rather than clearance of infection as occurs in sterile immunity). Microsporidia have also been shown to evade host immune responses by preventing phagosome-lysosome fusion, the production of immunomodulatory serum factors, and the suppression of host inflammatory responses (paradoxically, host stress leading to elevated cortisol levels and immunosuppression has also been associated with more severe infections).

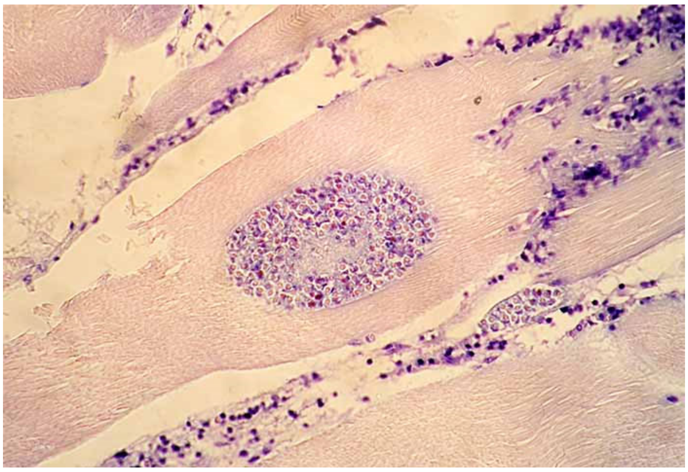
Developmental cycle and mode of transmission: The life-cycles of microsporidia vary considerably. Most piscine microsporidia have simple asexual monoxenous life-cycles, while some insect microsporidia have complex heteroxenous life-cycles with both asexual and sexual reproduction involving successive host generations or crustacean intermediate hosts. Mature spores have tough resistant walls which allow them to survive moderate ranges of various physical and chemical factors encountered in host environments (temperature, moisture, UV radiation, salinity, pH), but they are still susceptible to extremes of heat, cold, desiccation, sunlight, hypersalinity, acidity or alkalinity. Each spore contains an infective sporoplasm which is injected into a host cell through the polar tube when it is forcibly everted by swelling of the posterior vacuole and polaroplast (triggered by changes in calcium influx, osmotic pressure, pH, mechanical compression, etc.). The parasites then undergo vegetative reproduction by merogony (schizogony) by binary or multiple fission, although cytokinesis is sometimes delayed thus forming multinucleate plasmodia which divide by plasmotomy to form more plasmodia or segment into uninucleate meronts again. Meronts may lie direct in the host cell cytoplasm or be contained within membranous parasitophorous vacuoles (the exception being *Heterosporis* which forms a unique sporophorocyst envelope). The parasites then form thickened cell membranes and/or enveloping sporophorous vesicles and undergo further division called sporogony. Sporonts have electron-dense coats and undergo sporoblastogenesis by dividing through binary fission (disporoblastic sporogony) to form sporoblasts, or first forming multinucleated sporogonial plasmodia that divide by multiple fission, by gradual fragmentation, or sometimes producing intermediary mother cells (e.g. genus *Glugea*), to form sporoblasts. The sporoblasts subsequently mature to form spores (completing sporulation). The developmental stages of many microspora have single isolated nuclei throughout their development, ultimately giving rise to uninucleate sporoblasts and spores. Others have paired nuclei which divide synchronously and remain closely appressed throughout their development into diplokaryotic sporoblasts. Some diplokaryotic sporonts also appear to undergo meiotic reduction forming monokaryotic spores. The number of spores formed by each sporont (di-, tetra-, octo-, poly-sporous) can be a defining characteristic for many genera, particularly those developing within sporophorous vesicles. Merogony and sporogony may take place within the same or different tissues of individual hosts. Many microsporidian species are monomorphic (forming only one type of spore) while other species may produce different types of spores (macro-, medi-, micro-, meio-spores) at different stages of development, each type probably having a different function, such as autoinfection (facilitating dissemination within the same host), horizontal transmission (from host to host) and vertical transmission (from mother to offspring). Most infections in fish appear to be transmitted directly between hosts by contact with mature infective spores (skin or gill epithelia) or following their ingestion (gut epithelia). Microsporidian spores have been shown to be liberated from infected hosts from eruptive lesions in the gills and skin, from the digestive and urinary tracts and from degenerating tissues in decomposing carcases. Direct transmission by spore ingestion has been recorded for many species, while some apparently require spores to be primed or concentrated in invertebrate paratenic hosts (water fleas, brine shrimp, amphipods). Fish-to-fish transmission has been achieved experimentally via cohabitation, predation, scavenging, cannibalism and feeding on crustaceans. Vertical transmission has also been demonstrated for microsporidia infecting fish gonads, involving both intra-ovum (transovarial) transmission within eggs and extra-ovum (transovum) transmission on eggs (the latter occurring much more frequently in insect microsporidia during oviposition).

Differential diagnosis: Infections may sometimes be suspected on clinical grounds, with visible cyst-like lesions (sometimes discoloured) on body surfaces and/or erratic fish behaviours (impaired swimming, feeding, respiration). Infections are best diagnosed by the direct detection of parasites within host tissues, using both macroscopic techniques to detect visible cysts/xenomas and microscopic techniques to detect spores. Visual examination of fish tissues during dissection may reveal the presence of distinctive white cysts, tumour-like xenomas or unnatural opaque tissues (pale porcelain discolourations due the presence of numerous spores). Microscopic examination of wet tissue mounts, squash preparations, impression smears or histological sections may reveal the presence of characteristic microsporidian spores. Unstained samples are best examined at medium to high power (400X magnification) by bright-field microscopy with a suboptimal illumination system (introduce diffraction/contrast through specimen by racking down condenser and/or partially closing diaphragm) or alternatively using phase-contrast or differential interference-contrast microscopy. Mature spores are highly refractile, phase-bright, Gram-positive, acid-fast, and have a PAS-positive polar granule. Treatment of spores with dilute hydrogen peroxide can induce mature spores to evert their polar tubes. Histological sections are best stained using Ziehl- Neelson acid-fast, Periodic acid Schiff or Giemsa stains to highlight merogonous and sporogonous stages. Given the small spore size and their homogenous appearance, transmission electron microscopy is often conducted to reveal the presence of the coiled polar tube within spores and/or the presence of a sporophorous vesicle. Some species have been successfully propagated *in vitro*, e.g. *Nucleospora salmonis* in chinook salmon leucocyte cultures. More recently, immunodiagnostic procedures have been developed to detect host antibodies or parasite antigens, including the use of polyclonal and monoclonal antibodies, fluorescent-labels and enzyme immunoassays. Considerable success has been achieved in detecting parasite DNA by polymerase chain reaction (PCR) amplification of internal transcribed spacer (ITS) regions and small subunit (SSU) ribosomal RNA (rRNA) gene sequences. Molecular characterization techniques are facilitating not only more sensitive and specific diagnoses but also allowing more comprehensive phylogenetic analyses of relationships between taxa.

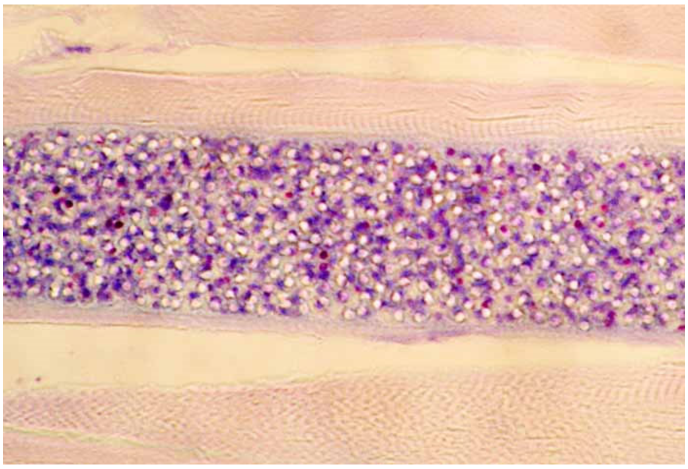
Treatment: Most drug treatments for microsporidian infections of fish have been applied in experimental situations or in intensive rearing facilities. The antimicrobial agent fumagillin was developed to treat infections in bees, and has been widely used to treat infections in fish although high concentrations or prolonged treatment has adverse effects on the hosts. Fumagillin is not heat stable so foodstuffs must be coated after milling. The anticoccidial triazinone, toltrazuril, has also proven effective as a systemic treatment for some infections. Limited success in treating fish has been reported using other anticoccidial drugs (amprolium, nitrofurazone) and anthelmintic benzimidazole derivatives (albendazole, mebendazole, fenbendazole). Quarantine and disinfection procedures may be used to control and prevent the spread of infections. Translocations of live fish (and source waters) between geographic zones should be avoided to prevent spreading infections. All new stock introduced into aquaculture facilities should be held in quarantine pending satisfactory screening. It is also important to isolate or cull infected fish as soon as possible after disease presentation to prevent water contamination by faeces, necrotic lesions or decaying carcases and spore transmission by contact or ingestion (including scavenging and cannibalism). At present, there are no practical ways of removing infections from wild fish populations without complete extirpation of all fishes in the water body. When processing fish, it is recommended not to dispose of offcuts, heads, entrails or skeletons into source waters, but to dispose of them by burning or burying in landfill. The prevention of infections is based on maintaining good water quality and minimising environmental stressors. Spores have been shown to remain infective under suitable conditions for up to one year, so if infections persist, aquaria or tanks need to be drained and disinfected (chemical treatment with monoaminoacridine or trypaflavine, or liming and drying over summer). Partial success has also been reported in killing spores in water by adjusting the pH to 7.5-8.0, treating with moderate levels of ozonation, or using ultraviolet (UV) sterilisation in water recirculation systems. Fish have been shown to develop some resistance to infection, so several studies developed and tested experimental vaccines in cultured fish. While several preparations (live low-virulence or inactivated whole spores) were found to provide partial protection against microsporidial gill disease in salmonids, as determined by better survival and reduced xenoma formation, no commercial vaccines are yet available. Some promising results have also been obtained when using small interfering RNA molecules (siRNAs) for *in vitro* silencing of microsporidial genes (ATP/ADP antiporter and methionine aminopeptidase) in cultured lizardfish.



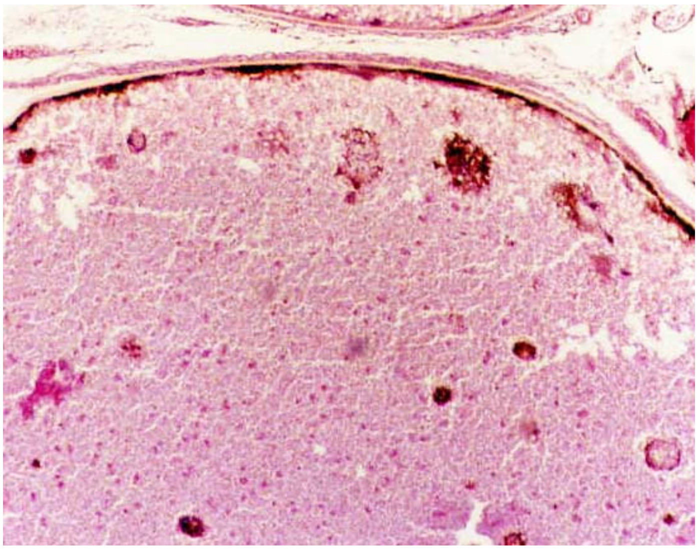
Microspora (piscine hosts)
e.g. *Glugea*
simple monoxenous cycles
most monokaryotic
form unicellular spores
with unique polar tubes
histozoic (viscera, muscles)
(lesions, cysts, sometimes
tumour-like xenomas)
Vertebrate Hosts
(freshwater, marine
and estuarine fish)
development may occur direct in host cell cytoplasm
or in parasitophorous vacuole (membrane of host origin)
or in sporophorous vesicle (envelope of parasite origin)
[a few form thick-walled sporophorocysts (parasite membranes)]
multiplication by merogony
(binary or multiple fission, although
several form multinucleated plasmodia)
spore formation by sporogony
(binary or multiple fission,
a few by plasmotomy)
meront
sporont
sporoplasm injected
into host cell
polar tube
eversion
coiled
polar
tube
posterior
vacuole
microspore
(2-8 µm)
some form xenomas
with multilaminate
walls
infective spores
ingested/inhaled
mature spores
released
most transmission between hosts direct
via contamination of water by microspores



Pleistophora cyst in fish muscle



Pleistophora cyst in fish muscle



Glugea cyst (xenoma) in fish viscera