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Two New Species of *Plistophora* (Microsporidea) from North American Fish with a Synopsis of Microsporidea of Freshwater and Euryhaline Fishes

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SYNOPSIS. Two new species of Microsporidea, *Plistophora salmonae* from steelhead and rainbow trout (*Salmo gairdneri*) and *Plistophora cepedianae* from gizzard shad (*Dorosoma cepedianum*) are described. Schizonts to spores of *P. cepedianae* were found at one time within the same cyst, while only sporonts and spores of *P. salmonae* were found within the cyst.

An illustrated synopsis of the known Microsporidea of freshwater and euryhaline fishes is given.

DURING 1962-63, occurrences in the states of California and Ohio of microsporidians belonging to the genus *Plistophora* infecting freshwater fish were brought to our attention.

In California the microsporidian was found infecting salmonid fish. Elmo B. Barney, hatchery biologist, Coleman National Fish Hatchery, Anderson, California, sent material for diagnosis and study.

In Ohio the microsporidian was found killing gizzard shad (*Dorosoma cepedianum*) of Charles Mill Reservoir in Richland County, but not affecting any other species of fish. Loren Mosely, State District fish management agent, sent material for diagnosis to Lyle Pettijohn of this laboratory, who made the material available to us.

At the present time *Plistophora* of salmonids has been reported only from California and the *Plistophora* of gizzard shad only from Ohio.

MATERIALS AND METHODS

Parasite cysts were preserved in formalin, then punctured and smears made of the contained material. After drying the slides were variously stained; best results were obtained with crystal violet, Giemsa's and fast green. Sections were made

and stained with Giemsa's, Harris' hematoxylin and eosin, and Heidenhain's iron-hematoxylin and eosin. All measurements are based on 10 or more specimens.

OBSERVATIONS

Plistophora salmonae n. sp.
(Figs. 3, 12 and 26.)

Diagnosis

Synonomy: Plistophora sp. Wales and Wolf, 1955 (15).

Host: Salmo gairdneri Richardson, 1836.

Site of infection: Gill tissue.

Locality reported: U.S.A. (California).

Type slide: U.S. National Museum Helm. Coll. No. 60027.

Spores: Piriform, refractive in unstained state and circular in cross section. Preserved, averaged 4.5 μ (4.2-5.3 μ) long by 2.2 μ (1.7-2.8 μ) wide. Anterior and posterior vacuole present. Polar capsule not visible. When focusing up and down on stained spores, lines running obliquely to the long axis of the spore can be seen giving the appearance of a coiled polar filament (Fig. 26). With fast green stain, a small structure *ca* 0.7 μ in diameter is visible in the posterior vacuole.

Vegetative stages: Sporonts 6-8 μ in diameter containing about 16 spores; several sporonts in one small cyst 23 μ in diameter.

Cysts: From 50 to 212 μ in diameter containing individual spore with no limiting membrane and found throughout the gill tissue.

Distinguishing characters: Spores larger than *P. acerinae, P. dalli, P. typicalis* and *Plistophora* sp. Bond, 1937, but smaller than *P. ehrenbaumi, P. macrospore* and *P. ovariae.* The spores are similar in size to *P. gadi* and *P. hyphessobryconis*, but both are muscle parasites of European fish. The spores of *P. ehrenbaumi. P. longifilis* and *P. mirandellae* are dimorphic and those of *P. oolytica* are trimorphic.

Relation to host: Cysts which were isolated on gill lamellae were surrounded by a thin layer of epithelium and no inflammation (Figs. 3 and 12). However, the tips of the gill filaments, where the cysts were more numerous, were badly clubbed (epithelial hyperplasia and fusion of lamellae). It is possible that other factors also contributed to the clubbing. Often the lamella blood vessel could be seen running through the spores mass (Fig. 12).

DISCUSSION

P. salmonae n. sp. is placed in the genus *Plistophora* because there are probably 16 or more spores produced in each sporont. We cannot count the spores accurately in our preserved material, but since we can clearly count 11-12 spores in each sporont which contained more, we presume that there are 16 or more.

Wales and Wolf (1955) reported a *Plistophora* sp. which is probably *P. salmonae.* They found it widespread in the gills of hatchery-reared and wild *Salmo gairdneri, Oncorhynchus nerka* and *Cottus* sp. in northern California. The fresh spores were 7.5 μ by 2.4 μ (our preserved spores were smaller). Young schizonts were 5 μ in diameter and developing sporonts 15 to 40 μ. Cysts were 120-180 μ in diameter. They reported hatchery epizootics wherein the gills showed moderate to great epithelial proliferation, and there was a good correlation between anemia and abundance of the organisms. They indicated that in more acute hatchery epizootics there was considerable mortality.

Plistophora cepedianae n. sp.
(Figs. 1, 2, 4-11, and 23.)

Diagnosis

Synonomy: Microsporidia sp. Bangham, 1941(1), 1962(2).

Host: Dorosoma cepedianum (Le Suer, 1818).

Site of infection: Visceral cavity.

Locality reported: U.S.A. (Ohio).

Type slide: U.S. National Museum Helm. Coll. No. 60026.

Spores: Piriform, refractive in unstained state, circular in cross section. Preserved unstained, average 4.9 μ (4.2-5.6 μ) long by 2.3 μ (1.7-2.8 μ) wide. Preserved stained, average 4.1 μ (3.5-4.4 μ) long by 2.5 μ (2.1-2.8 μ) wide. Anterior and posterior vacuole present. Between these vacuoles is a darker area (sporoplasm) which is 1.2 μ wide at its narrowest part (Fig. 23). The polar filament was not visible.

In spores stained with crystal violet, the sporoplasm takes on a deep purple, with the vacuoles remaining clear. With fast green, a small structure *ca* 1.4 μ in size and varying from triangular to round appears in the center of the posterior vacuole. Also with fast green or iodine solution, a small granule (sometimes two), appears within the anterior vacuole.

Vegetative stages: Stages between the sporoplasm of the spore and the schizont were not definitely recognized. In this species the schizonts are cylindrical and vary from straight to "C"-shape with the most common being the "C"-shape. They are an average of 35 μ long by 3.3 μ wide, and contain various numbers of nuclei which are *ca* 1.4 μ in diameter. At the end of some schizonts, a cup-like structure resembling a scopula is seen (Fig. 4).

The schizont probably gives rise to small, spherical, binucleate bodies which are *ca* 5.2 μ in size (Fig. 5). The nuclei are *ca* 1.0 μ in diameter and stain very dark with crystal violet. These binucleated bodies appear to give rise to irregular structures 9.0 μ in size with a nucleus of 3.2 μ, representing the beginning sporont (Fig. 6).

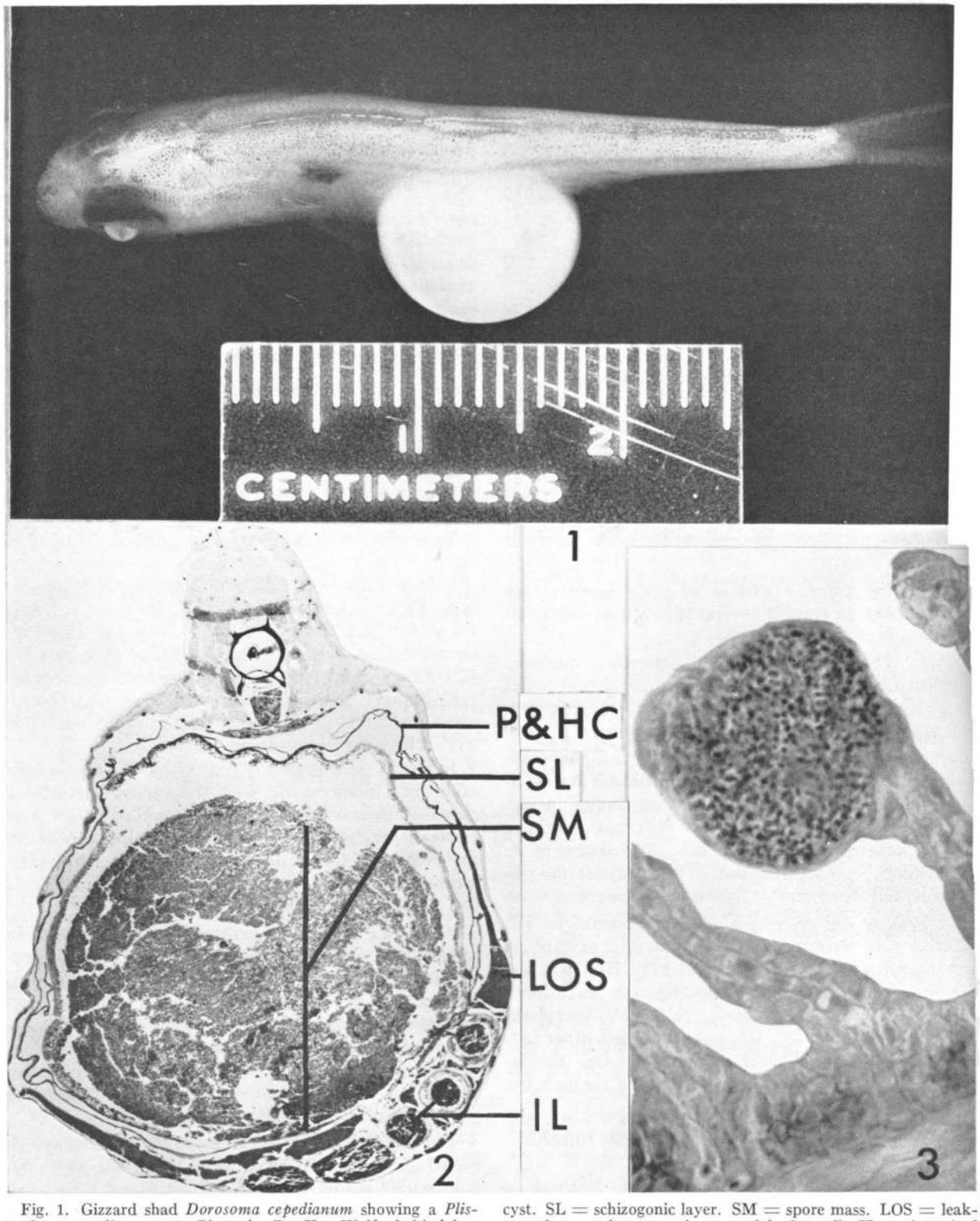


Fig. 1. Gizzard shad *Dorosoma cepedianum* showing a *Plistophora cepedianae* cyst. Photo by Dr. Ken Wolf of this laboratory. Fig. 2. Photomicrograph of cross section of gizzard shad showing relation of *P. cepedianae* cyst to host. Stained with Harris' hematoxylin. P & HC = Peritoneum and host cyst. SL = schizogonic layer. SM = spore mass. LOS = leakage of spores between viscera and body wall. IL = intestinal loops. Magnification 17.6 ×. Fig. 3. Photomicrograph of a sectioned salmonid gill stained with Heidenhain's iron-hematoxylin showing a *P. salmonae* cyst. Magnification 800 ×.

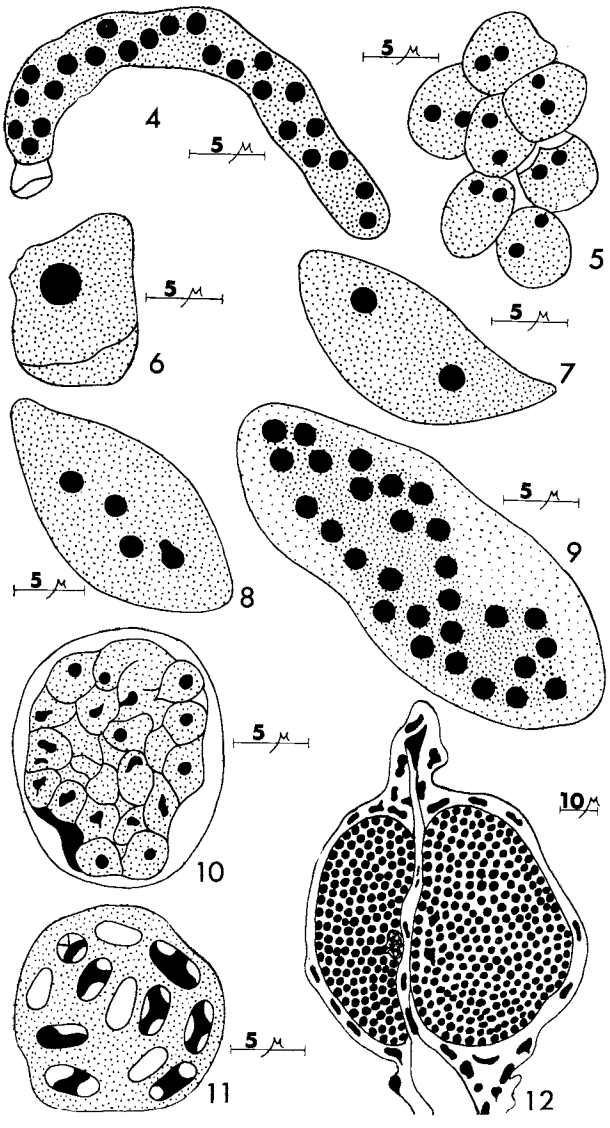


Fig. 4. Schizont of *Plistophora cepedianae* showing scopula-like structure. Fig. 5. Binucleated bodies of *P. cepedianae*, post schizont and pre-sporont stage. Fig. 6. Young sporont of *P. cepedianae*. Fig. 7. Binucleate sporont of *P. cepedianae*. Fig. 8. Quadrinucleate sporont of *P. cepedianae*. Fig. 9. Polynucleate sporont of *P. cepedianae*. Fig. 10. Sporont of *P. cepedianae* with young sporoblasts. Fig. 11. Sporont of *P. cepedianae* containing nearly mature spores. Fig. 12. Drawing of a sectioned salmonid gill lamella stained with Heidenhain's iron-hematoxylin containing a *P. salmonae* cyst.

The sporont grows with cytoplasmic and nuclear division taking place until oblong structures are formed (Figs. 7, 8 and 9) with nuclei *ca* 1.5 μ in diameter.

These oblong sporonts begin to round up, and become ovoid, varying in size from 15-30 μ in diameter. Cytoplasmic division within these sporonts takes place until small spheres, *ca* 3 μ in diameter, with a light staining cytoplasmic area around a darker irregularly shaped nucleus are seen (Fig. 10). These represent the beginning sporoblasts which vary in number from 6 to 20 within the sporonts.

The changes from the young uninucleate sporoblast to the mature spore were not observed.

Cysts: From 0.5-1.0 cm in diameter. Vegetative stages from schizonts to spores found within the same cyst. Only one cyst per fish has been found.

Distinguishing characters: One large cyst which fills and protrudes from the visceral cavity. Spores most resemble *P. dalli* in size, shape and presence of two vacuoles. No dimorphism noted. Apparently specific for *Dorosoma cepedianum*.

Relation to host: The parasite cyst is located in the visceral cavity and becomes so large that the visceral organs are compressed against the body wall. Occasionally the cyst protrudes from the body of the fish (Figs. 1, 2). In certain cases some of the spores have leaked out of the cyst and accumulated in the body cavity of the fish (Fig. 2). In some, the thin, hyaline cyst membrane (2-3 μ thick) is surrounded by a layer of loose host connective tissue 9-29 μ thick which is moderately vascular. There is no evidence of adjacent cellular reaction in most, although there is a rather extensive reaction involving tissue eosinophils in some infections. In others, which have not produced as much pressure on the fish organs, the host cyst is composed of very loose connective tissue up to 300 μ thick.

DISCUSSION

P. cepedianae n. sp. has been placed in the genus *Plistophora* since its sporogony is characteristic of that genus. i.e., the sporont develops into variable numbers of sporoblasts, often more than 16.

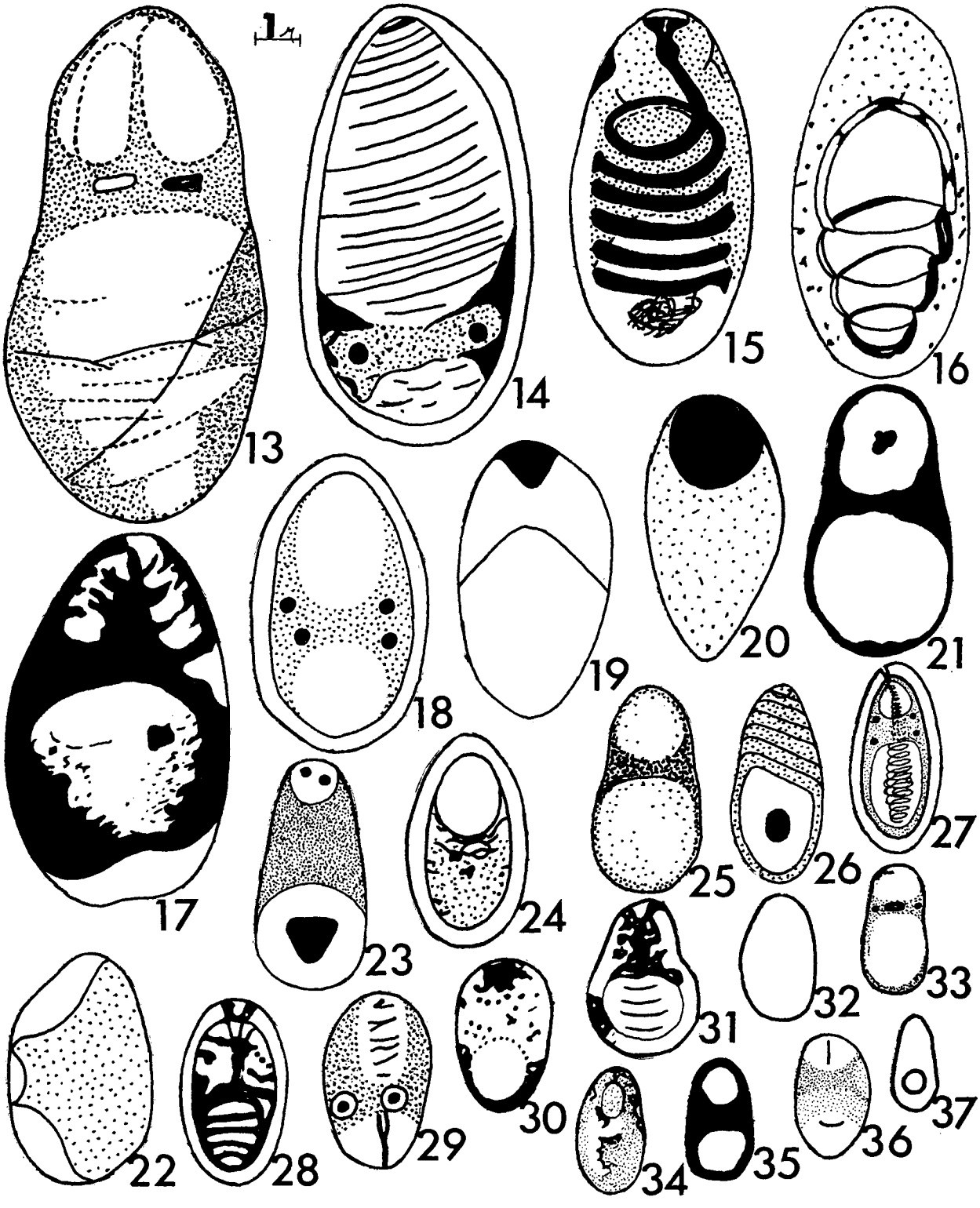
The size of *P. cepedianae* cysts, and the fact that vegetative stages as well as spores were present within the same cyst, might be attributed to repeated schizogony or to the spores being able to germinate in the same host body with the number of infected cells increasing.

SYNOPSIS OF THE KNOWN MICROSPORIDEA OF FRESHWATER AND EURYHALINE FISHES

The object of this review is to bring together spore drawings and salient information necessary for the identification of the known Microsporidea of freshwater and euryhaline fishes. Microsporidea belonging to the genera *Glugea, Nosema, Plistophora*, and *Thelohania* have been reported from fish.

Genus *Glugea* Thélohan, 1891 emend. Weissenberg, 1913.
Each sporont develops into two spores. Host cells become enormously hypertrophied, forming the so-called *Glugea*-cysts.

G. anomala (Moniez, 1887) (Gurley, 1893).
(Fig. 27.)



Figs. 13-37. Spores of *Glugea, Nosema, Plistophora* and *Thelohania* of freshwater and euryhaline fish; all drawn to same scale and arranged from largest to smallest. Fig. 13. *P. longifilis* macrospore, after Kudo(12). Fig. 14. *P. elegans*, after Kudo(12). Fig. 15. *P. macrospora*, after Kudo(12). Fig. 16. *P. ovariae*, after Summerfelt(14). Fig. 17. *P. oolytica* macrospore, after Weiser(16). Fig. 18. *T. ovicola*, after Kudo(12). Fig. 19. *N. branchiale*, after Kudo(12). Fig. 20. *P. gadi*, after Bychowskaya-Pavlovskaya *et al.*(4). Fig. 21. *P. oolytica*, medium size spore, after Weiser(16). Fig. 22. *P. hyphessorbry-*

Synonyms: Nosema anomala Moniez, 1887, *Glugea microspora,* Thélohan, 1892. Cope(6) reported this as *Nosema anomela.*

Information from Kudo(12).

Fish host: Gasterosteus aculeatus, G. pungitius and *Gobius minutus.*

Site of infection: Subcutaneous connective tissue, cornea, ovary, peritonium, and alimentary canal.

Locality reported: France, England, Russia, Germany, Belgium, and U.S.A. (Alaska); freshwater and marine.

Spores: 3-6 μ long by 1.5-2 μ wide. Clear space in spore (vacuole). Ovoidal, one end attenuated. Spore membrane composed of two valves. Filament length 150 μ.

Vegetative stages: Schizonts uninucleated, undergo division. Nuclear divisions continue without cytoplasmic constriction. Elongated plasmodia with 2-30 nuclei placed lengthwise are produced. Plasmodia round up and break into individuals possessing two nuclei. From these zygotes are formed. Sporonts in vacuole previously occupied by plasmodia. Each sporont divides into two sporoblasts by mitosis.

Cysts: Lodged in epidermis between bundles of fibrous connective tissue cells, 3-4 mm in diameter. Continuous membrane around; 5-10 μ thick.

G. hertwigi Weissenberg, 1911.

(Fig. 24.)

Synonyms: G. stephani, Mavor, 1915, *Glugea* sp. Schrader, 1921.

Information from Fantham *et al.*(8), Haley(9-11) and Kudo (12).

Fish host: Osmerus eperlanus and *O. mordax.*

Site of infection: Subcutaneous connective tissue, cornea, gut wall, serous membrane of hindgut, stomach, liver and connective tissue of gonads.

Locality reported: Germany, United States, Canada, and Russia.

Spores: Elongated and piriform, 4.6-5 μ long by 2.3 μ wide. A large vacuole at the posterior end. Filament length 100 μ.

Vegetative stages: Schizonts 7 μ long, in large groups in peripheral layer and central portion of cyst. Binucleate schizonts up to 9.4 μ long. Schizonts grow with nuclear division forming 32-42 nuclei. Fluid vacuole formed around the schizont, and it breaks into many cells 3-3.5 μ in diameter with a nucleus of 1.4 μ in diameter.

Cysts: Lie closely together, have distinct membrane 2 mm in diameter. Cyst wall 2 μ thick. Sporonts, sporoblasts, and ripe spores may all be found in a single cyst with earliest stages near the periphery.

Remarks: Found in both freshwater and marine forms of *O. mordax.* The European and American forms were placed in one species by Kudo(12). Fantham *et al.*(8) classify the *Glugea* of Canadian *O. mordax* with the one of the European and United States *Osmerus,* but because of different situation and disposition of the polar filament and a smaller length and breadth of spores suggest the name, *G. hertwigi canadensis.*

G. intestinalis Chen, 1956.

(Fig. not given here.)

Information from Chen(5).

Fish host: Mylopharyngodon piceus.

Site of infection: Mucous tissue of small intestine.

Locality reported: China.

Spores: Ovate, circular in cross section 6.2 μ by 3.6 μ. Oval or ellipsoidal polar capsule occupies the anterior half to two-thirds of the spore. Within the capsule in some specimens, coils of the polar filament could be observed. Rest of spore filled with cytoplasm containing a rounded nucleus and a large oval vacuole at the posterior end.

Vegetative stages: Information not given.

Cysts: Information not given.

G. luciopercae (Dogiel, 1939).

(Fig. 28.)

Information from Bychowskaya-Pavlovskaya *et al.*(4).

Fish host: Lucioperca lucioperca.

Site of infection: Intestine, mesentery.

Locality reported: Russia (Caspian Sea).

Spores: 3.8-5 μ long by 1.8-2.5 μ wide.

Vegetative stages: Russian text not translated yet.

Cysts: 91-106 μ in diameter.

G. stephani (Hagenmuller, 1899) Woodcock, 1904.

(Fig. 30.)

Synonym: Nosema stephani, Hagenmuller, 1899, Sporozoan, Johnstone, 1901, Protozoan, Linton, 1901.

Information from Bychowskaya-Pavlovskaya *et al.*(4) and Kudo(12).

Fish host: Pleuronectes flesus (Flesus passer), P. platessa, and *Pseudopleuronectes americanus.*

Site of infection: Intestinal walls, liver and mesentery.

Locality reported: France, England, United States and Russia.

Spores: Oblong-ovate. 3 μ long by 1.5 μ wide. Anterior and posterior vacuole.

Vegetative stages: Sporonts and spores in endoplasm layer within cyst. Sporoblasts surrounded by a fibrous envelope.

Cysts: Small, white and ovoidal or spherical. 0.5-1.0 mm in diameter.

Remarks: This may be a marine species, but it was included in the freshwater key of Bychowskaya-Pavlovskaya *et al.*(4).

Genus *Nosema* Naegeli, 1857 emend. Pérez, 1905.

Each sporont develops into a single spore.

N. branchiale Nemeczek, 1911.

(Fig. 19.)

Information from Bychowskaya-Pavlovskaya *et al.*(4), Dogiel *et al.*(7), Fantham *et al.*(8), and Kudo(12).

Fish host: Gadus aeglifinis and *G. callarias.*

Site of infection: Branchial lamellae.

Locality reported: Austria, Canada, and Russia.

Spores: Oval, distinct vacuole at one end. Sporoplasm granular. Fresh spores 6.3 μ long by 3.5 μ wide. Filament 90 μ long.

Vegetative stages: Youngest forms rounded to triangular with granular cytoplasm. Each schizont develops into one sporont and one sporoblast forming one spore.

conis, after Schäperclaus(13). Fig. 23. *P. cepedianae* n. sp. Fig. 24. *G. hertwigi,* after Kudo(12). Fig. 25. *P. dalli,* after Bychowskaya-Pavlovskaya *et al.*(4). Fig. 26. *P. salmonae* n. sp. Fig. 27. *G. anomala,* after Kudo(12). Fig. 28. *G. luciopercae* after Bychowskaya-Pavlovskaya *et al.*(4). Fig. 29. *N. pimephales,* after Fantham *et al.*(8). Fig. 30. *G. stephani,* after Bychowskaya-Pavlovskaya *et al.*(4). Fig. 31. *P. peponoides,* after Bychowskaya-Pavlovskaya *et al.*(4). Fig. 32. *P. acerinae,* after Kudo(12). Fig. 33. *P. longifilis* microspore, after Kudo(12). Fig. 34. *P. typicalis,* after Kudo(12). Fig. 35. *P. oolytica* microspore, after Weiser(16). Fig. 36. *P. species,* after Bond(3). Fig. 37. *N. girardini,* after Kudo(12).

Cysts: Spherical, 0.2-0.5 mm in diameter and embedded in lamella.

Remarks: Kudo(12) lists *N. branchiale* provisionally since Nemeczek's observations on the vegetative form are inadequate to assign it definitely to the genus *Nosema*.

N. girardini Lutz and Splendore, 1903.

(Fig. 37.)

Information from Kudo(12).

Fish host: Girardinus caudimaculatus (Cyprinodontidae).
Site of infection: Muscles, serosa, and mucosa of intestine.
Locality reported: Brazil.
Spores: Piriform, 2-2.5 μ long by 1-1.5 μ wide.
Vegetative stages: No available information.
Cysts: No available information.

N. pimephales Fantham, Porter, and Richardson(8), 1941.

(Fig. 29.)

Information from Fantham *et al.*(8).

Fish host: Pimephales promelas.
Site of infection: Abdomen.
Locality reported: Canada (Quebec).
Spores: Vacuole at one pole, nucleus at the other 3.8-4.4 μ long by 1.9-3.3 μ wide. Cytoplasm girdle-like. Filament 70-90 μ long.
Vegetative stages: Amoebula 2 μ in diameter, and uninucleate or binucleate. Smallest trophozoites or meronts 1.5-2.2 μ in diameter with many about 3 by 2.6 μ. Clusters of schizonts found in cyst. One sporont and subsequent sporoblast formed by each uninucleate schizont giving rise to one spore.
Cysts: Large cyst distending abdomen and particularly obliterating coelomic cavity.

Genus *Plistophora* Gurley, 1893.

Each sporont develops into many (more than 16) spores.
Synonym: Pleistophora Gurley, 1893.

P. acerinae Vaney and Conte, 1901.

(Fig. 32.)

Information from Kudo(12).

Fish host: Acerina cernua.
Site of infection: Mesentery.
Locality reported: France (Lyon).
Spores: Ovoid, 3 μ long by 2 μ wide. Filament 15 μ long.
Vegetative stages: Round sporonts, young ones with thin membrane, mature ones with thick membrane.
Cysts: Whitish elongated mass *ca* 3 mm long.

P. cepedianae n. sp.
(see Observations, this paper)

P. dalli Zhukow, 1963.

(Fig. 25.)

Information from Bychowskaya-Pavlovskaya *et al.*(4).

Fish host: Dallia sp.
Site of infection: Subcutaneous connective tissue at base of fins.
Locality reported: Russia.
Spores: Oval, slightly narrower anterior end. 3.9-5.5 μ long by 2.2 μ wide.
Vegetative stages: Sporont gives rise to more than 16 spores.
Cyst: Visible to naked eye. Apparently formed by growth of single cell.

P. ehrenbaumi Reichenow.

(No. fig.)

Information from Schäperclaus(13).

Fish host: Catfish.
Site of infection: Musculature.
Locality reported: Germany.
Spores: Dimorphic 7.5 × 3.5 μ and 3 × 1.5 μ.
Vegetative stages: Sporonts 10-24 μ.
Cysts: No information.
Remarks: Listed here provisionally as we are not sure whether the catfish Schäperclaus notes is a freshwater or marine form.

P. elegans Auerbach, 1910.

(Fig. 14.)

Information from Kudo(12).

Fish host: Abramis brama × *Leuciscus rutilis.*
Site of infection: ovary.
Locality reported: Germany (Karlsruhe).
Spores: Highly elongated and narrow 10 μ long by 4 μ wide. Large polar capsule with coiled filament, and sporoplasm around.
Vegetative stages: Schizonts multinucleated at a certain stage and abundant in connective tissue. Sporulation only in host ova.
Cyst: No available information.

P. gadi (Poljansky, 1955).

(Fig. 20.)

Information from Bychowskaya-Pavlovskaya *et al.*(4).

Fish host: Atlantic cod.
Site of infection: Musculature.
Locality reported: U.S.S.R. (Barents Sea).
Spores: Ovoid, narrow at anterior end, 5.4-7.2 μ long by 2.7-3.6 μ wide. Large vacuole at posterior end.
Vegetative stages: Pansporoblasts 18-23 μ diam. containing large number of spores.
Cysts: Large, 0.5-0.8 cm in diam.
Remarks: This species was included in the Russian freshwater key of Bychowskaya-Pavlovskaya *et al.*(4).

P. hyphessobryconis Schäperclaus, 1941.

(Fig. 22.)

Information from Schäperclaus(13) and Lom & Vavra(17).

Fish host: Hyphessobrycon innesi and *H. gracilis.*
Site of infection: Musculature.
Locality reported: Germany.
Spores: 5-6 μ long × 3.3 μ wide.
Vegetative stages: Pansporoblasts 25-36 μ in diam.
Cysts: No available information.

P. longifilis Schuberg, 1910.

(Figs. 13 and 33.)

Information from Kudo(12).

Fish host: Barbus fluviatilis.
Site of infection: Testis.
Locality reported: Germany (Heidelberg).
Spores: Dimorphic. Piriform. Macrospores up to 12 μ long by 6 μ wide. Microspores 3 μ long by 2 μ wide. Filament up to 510 μ long.
Vegetative stages: Mature sporonts spherical 8-45 μ in diam. Number of spores per sporont inconstant and large. Maximum diam. of sporonts with microspores, 30 μ, with macrospores up

to 12 μ long by 6 μ wide. Microspores 3 μ long by 2 μ wide. Filament up to 510 μ long.

Cysts: Clear, whitish, and rounded spots of various size distending the surface of testis.

P. macrospora Cépède, 1906.
(Fig. 15.)
Information from Kudo(12).

Fish host: Cobitis barbatula.
Site of infection: Muscles of abdomen.
Locality reported: France (Grenoble).
Spores: Ovoid, some with vacuole at one end, others at both ends. 8.5 μ long by 4.25 μ wide. Filament is 225 μ long.
Vegetative stages: Sporont spherical or subspherical, 25-30 μ in diameter. Contain mature spores in a large but variable number.
Cysts: Yellowish white and transparent. Ellipsoidal, *ca* 3 mm in diameter.

P. mirandellae Vaney and Conte, 1901.
(No fig.)
Information from Kudo(12).

Fish host: Alburnus mirandellae.
Site of infection: Ovary and ovum.
Locality reported: France.
Spores: Dimorphic, ovoid macrospores scattered in host connective tissue, 12 μ long by 6 μ wide. Vacuole at one end. Microspores 7.5 μ by 4 μ.
Vegetative stages: Sporonts develop in ovaries and ova.
Cysts: Small cysts contain a rounded mass of microspores. Larger cysts contain scattered macrospores.

P. oolytica Weiser, 1949.
(Figs. 17, 21 and 35.)
Information from Weiser(16).

Fish host: Leuciscus cephalus and *Esox lucius.*
Site of infection: Ova.
Locality reported: Czechoslovakia (Svitava River, near Brno and vicinity of Vranovice).
Spores: Two vacuoles, and ring-shaped cytoplasmic body between. *Leuciscus cephalus*—three types: large, 8.4 μ by 4.2 μ, medium size, 5.5-6.5 μ by 3.5 μ (found most frequently) and small, 3 by 1.5 μ. *Esox lucius* spores, large, 7.3 by 3.5 μ, medium 5 by 3 μ and small 3 by 1.5 μ.
Vegetative stages: Schizonts oval 23 μ in size with one or two nuclei. Chains of sporonts form. Spherical sporoblasts develop, 2.3-3.4 μ in size.
Cysts: No information given except that infected ova are white.

P. ovariae Summerfelt, 1964.
(Fig. 16.)
Information from Summerfelt(14).

Fish host: Notemigonus crysoleucas.
Site of infection: Ovaries, liver and kidney.
Locality reported: U.S.A. (Arkansas, Illinois, Kentucky and Missouri).
Spores: Ovoid or ellipsoidal. Arithmetic means and standard errors of 50 fresh spores, 8.42 μ $\pm$ 0.17 by length and 4.24 μ $\pm$ 0.15 width; 50 Schaudinn-fixed spores 6.44 μ $\pm$ 0.11 by 3.43 μ $\pm$ 0.05; 50 Carnoy-fixed spores 6.60 μ $\pm$ 0.08 by 3.31 μ $\pm$ 0.05.
Vegetative stages: Sporonts 17.8-22.8 μ in length and 15-17.8 μ in width. Eight, 12 or 16 spores produced with 12 the most frequent number.
Cysts: In aggregates 1.0-1.75 mm; individual cyst range from 345-550 μ.

P. peponoides Schulman, 1964.
(Fig. 31.)
Information from Bychowskaya-Pavlovskaya *et al.*(4).

Fish host: Percottus glehi.
Site of infection: Subcutaneous connective tissue of fins.
Locality reported: Russia (Amur River Basin).
Spores: Uniform in size, narrow anterior end, resembles a crookneck squash or flask, 3.6 μ long by 2-2.3 μ wide.
Vegetative stages: Large number of sporonts in which up to 8 spores are found.
Cysts: Spherical, white, and up to 1 mm in diameter.
Remarks: This may be *Thelohania* because it has 8 spores per sporont.

P. salmonae n. sp.
(see Observations, this paper)

P. typicalis Gurley, 1893.
(Fig. 34.)
Information from Kudo(12).

Fish hosts: Cottus bubalis, C. scorpius, Blennius pholis and *Gasterosteus pungitius.*
Site of infection: Musculature.
Locality reported: France (Concarneau, Roscoff, and Rennes).
Spores: Ovoid, polar capsule present. Spore 3 μ long by 1.5-2 μ wide.
Vegetative stages: Sporonts spherical, 15-18 μ in diameter. Contain large and inconstant number of spores.
Cysts: Small masses of protoplasm each with a distinct membrane and nuclei between fibrillae, 4 μ long by 2.5-3 μ wide.

Plistophora sp. Bond, 1937.
(Fig. 36.)
Information from Bond(3).

Fish host: Fundulus heteroclitus.
Site of infection: Muscle tissue.
Locality reported: U.S.A. (Chesapeake Bay, Maryland).
Spores: Oval, 2.5-3 μ wide by 1.5-2 μ long. Vacuole at each end, sporoplasm in middle.
Vegetative stages: Sporonts 18-30 μ in diameter. Sporont nuclei in a number of cases were similar to the structure of the surrounding tissue nuclear forms.
Cysts: Swelling about 1 mm above the surface on the left side of body anterior to the dorsal fin above the lateral line, and posterior to the pectoral fin.
Remarks: Bond's sporont may be a host cell containing *Nosema* spores as suggested by his own statement, "pansporoblastic nuclei (sporont) in a number of cases similar to structure of surrounding tissue nuclear forms."

Genus *Thelohania* Henneguy, 1892.

Each sporont develops into eight sporoblasts and ultimately into eight spores. The sporont membrane may degenerate at different times of development.

Thelohania ovoicola (Auerbach, 1910) Kudo, 1924.
(Fig. 18.)
Information from Kudo(12).

Fish host: Coregonus exiguus bondella.
Site of infection: Eggs.
Locality reported: Switzerland (Lake of Neuchatel).

Spores: Oval, middle part often constricted, spore membrane thick, size variable. Anterior and posterior vacuoles, 6-8 μ long by 4-6 μ wide. Filament 25-30 μ long.

Vegetative stages: Young sporonts oval, 6 μ in length with two dividing nuclei. Older sporonts spherical, 6-10 μ in diameter. Sporonts with sporoblasts or mature spores, either spherical or somewhat irregular and 10-12 μ in diameter.

Cysts: No mention of cysts; infected eggs are milky white.

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