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FEMS Microbiology Letters 219 (2003) 253–259

FEMS MICROBIOLOGY Letters

www.fems-microbiology.org

# *Saprolegnia oliviae* sp. nov. isolated from an Argentine river (Tierra del Fuego Province, Argentina)

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Received 13 December 2002; received in revised form 28 December 2002; accepted 30 December 2002

First published online 5 February 2003

**Abstract**

*Saprolegnia oliviae* sp. nov. is described from litter (floating dead twigs, leaves and roots) in the Olivia River, Ushuaia Department, Tierra del Fuego Province (Argentina). The new species is illustrated and compared with other species of the genus. Distinguishing characteristics of *S. oliviae* are the production of smooth oogonia (with some lateral or terminal projections) and the absence of antheridial branches on the majority of the oogonia, but when present, they are mostly diclinous, at times oogonia are supplied with androgynous and monoclinous antheridial branches. The oogonial stalks are predominantly short and straight or long and bent, curved or many times coiled; oospores are distinctive subcentric, (1–) 15–50 (–70) per oogonium. Morphological details of the new species and its comparison with other described species are discussed here.


*Keywords:* Straminipiles; Oomycota; *Saprolegnia*; Argentina; Systematics; New species

## 1. Introduction

During the course of study of zoosporic organisms occurring in water and floating organic matter in the Olivia River, Ushuaia Department, Tierra del Fuego Province (Argentina), a fungus was isolated belonging to the genus *Saprolegnia*. This isolate has its own distinctive features which separates it from other species within the genus. Hence it is being described as a new species, *S. oliviae* Steciow, belonging to the Kingdom Straminipila, Phylum Heterokonta, Class Peronosporomycetes [1,2].

The genus *Saprolegnia* Nees includes 19 species of widespread distribution occurring in fresh water and in soil as saprotrophs, playing an important ecological role in the decomposition and recycling of materials in aquatic ecosystems, but some of them can be parasite of fishes or their eggs, amphibians and aquatic invertebrates [3,4]. This genus is easily recognized by its profusely branched, coenocytic mycelium which give rise to long, cylindrical, and usually terminal zoosporangia that proliferate internally and by producing primary and secondary zoospores (dimorphic and diplanetic species). This type of zoospore release is different in most of *Achlya* species, which could be considered as monomorphic because the primary spore (lack flagella) produce cysts at the tip of the zoosporangium and germinate after a few hours, each releasing a secondary zoospore that swims away [1].

This is the fourth in a series of contributions to the knowledge of zoosporic organisms from Tierra del Fuego Province, the southernmost province of Argentina and of South America [5–7].

## 2. Materials and methods

The methods described by Coker [8], Sparrow [9], and Seymour [4] were used to isolate zoosporic organisms. Samples of brown decaying twigs, leaves and wood were collected from the Olivia River and brought to the laboratory in separate sterile polythylene bags. These samples were distributed in water in sterile Petri dishes containing several hemp seed halves (*Cannabis sativa*) and incubated at room temperature (15–20°C). Once the seeds were colonized by the fungal hyphae, a small bit of this was aseptically transferred to a cornmeal agar (CMA) medium. After 3–4 days, a block of agar from the edge of the fungal

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0378-1097/03/$22.00 
doi:10.1016/S0378-1097(03)00024-7

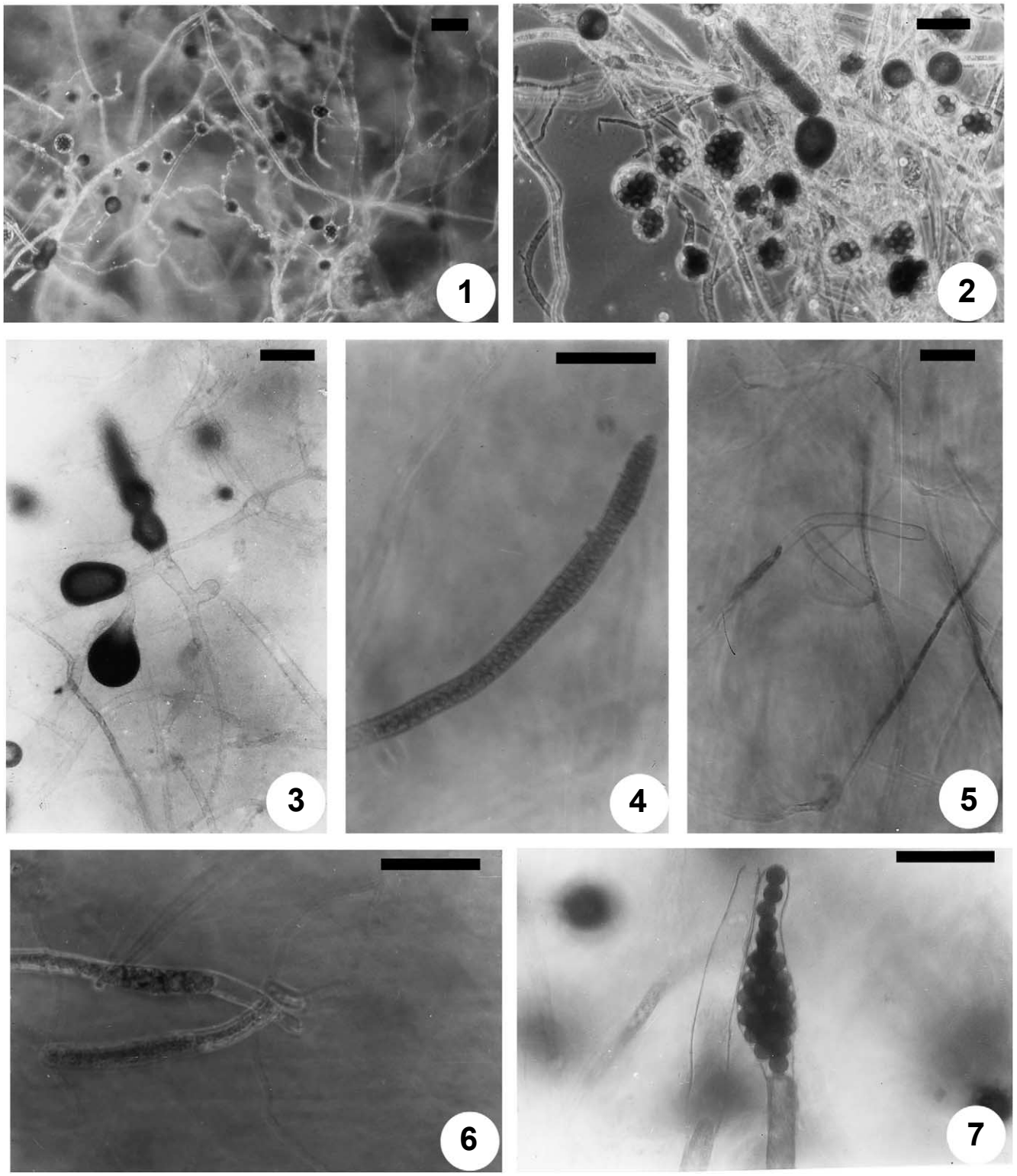


Fig. 1. 1–7: *S. oliviae*. 1–2: Aspect of mycelium with oogonia and gemmae in water culture. 3: Gemmae. 4: Zoosporangium containing zoospores. 5: Discharged zoosporangium. 6: Zoosporangium renewal by internal proliferation. 7: Naviculate discharged zoosporangium. Scale bars: 1 = 50 μm; 2,3,5 = 100 μm; 4,6,7 = 50 μm.

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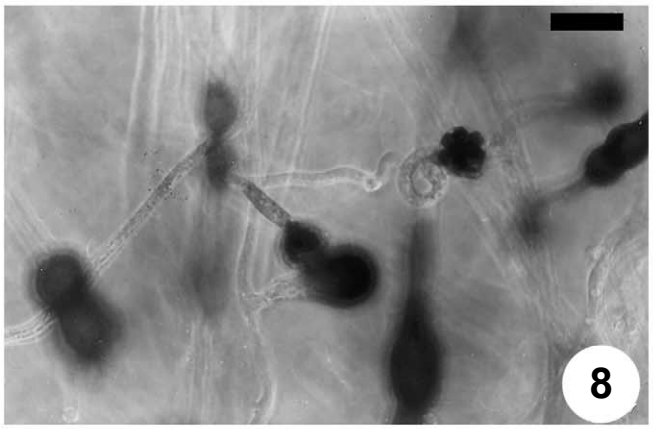
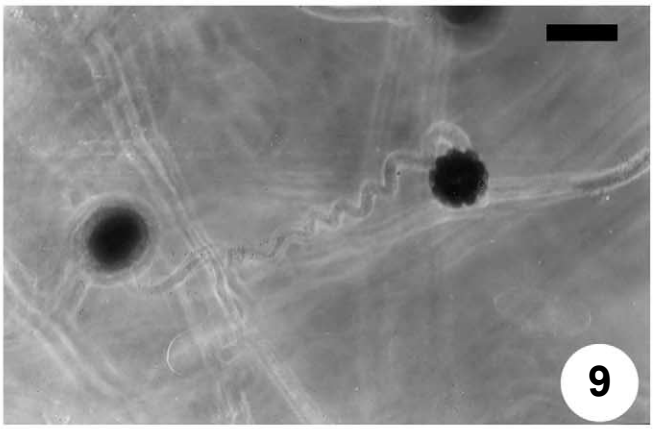
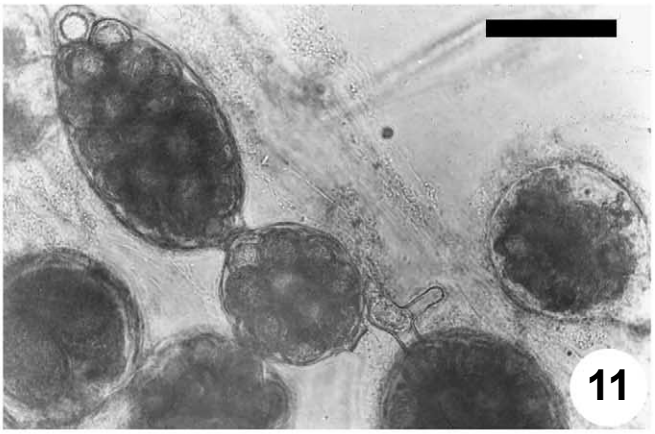
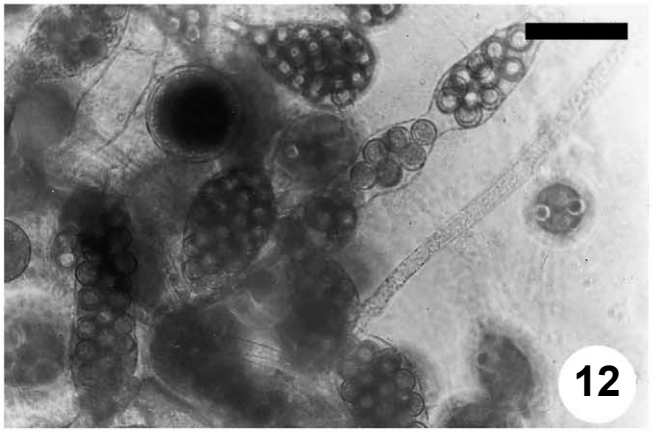
Fig. 2. 8–15: *S. oliviae*. 8,9: Coiled oogonial stalk. 10: Intercalary oogonium. 11,12: Single and catenulate oogonia of variable forms lacking antheridial branches but having many oospores, one of them with lateral projection. 13: Papillate oogonium with apical projection. 14: Long oogonial stalk. 15: Oogonium with lateral projection. Scale bars: 8,9 = 100 μm; 10–15 = 50 μm.



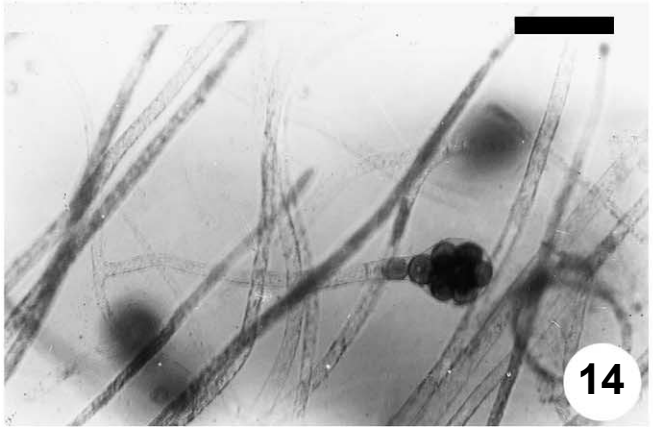



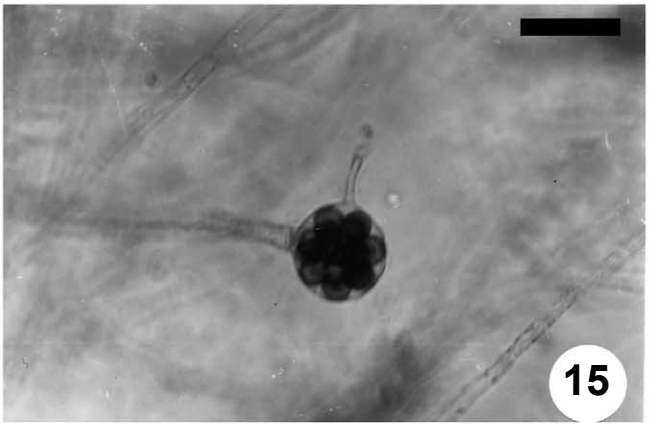








colony on CMA was cut off and placed in sterile Petri plates containing distilled water and hemp seed halves in order to obtain new bacteria free colonies. Morphological studies were done on these colonies. The water cultures on hemp seeds and the CMA plates were incubated at 5, 10, and 25°C to observe the possible effect of temperature on the sexual structures. The measurements like the diameters of fungal colonies, diameters of oogonia, number of oospores per oogonia, and diameters of oospores were calculated from 50 counts of each of three replicates. These were done by using an Olympus BX 40 microscope (Olympus Optical Co., Ltd., Tokyo, Japan) equipped with phase contrast optics. The total percentage of type of antheridial branches was calculated from all of these replicates.

The type specimen is deposited in the mycological herbarium of the Spegazzini Institute (LPS) culture collection.

## 3. Results

### 3.1. Saprolegnia oliviae *Steciow, sp. nov.*

Mycelium densum, cultura in seminibus *C. sativa* 1–4.5 cm diam. Hyphae ramosa, pleraque 15–73 μm late diam. ad basim. Sporangia in culturis juvenilibus, fusiformia, naviculata, clavata vel filiform, 194–600 μm larga et 24–50 μm lata, renovata per proliferationem internam. Ejecto sporarum pro genus typica, zoospori incystatis globosi 8–10 μm. Gemmae parcus. Oogonia copiosa, sphaerica vel pyriformia, (29–) 60–140 (–160) μm diam. Paries oogonia laevis, ramulus lateralibus provenientia, (24–) 190–400 μm. Oospori (1–) 15–50 (–70) per oogonium, subcentrici, (12) 19–30 μm diam. Ramulus antheridiales parcus, ramosus, plerumque origine diclina (35%) et androgyna (14%) sed interdum monoclina (7%).

Mycelium extensive, denser near substratum, 2-week-old colony on hempseed, 1–4.5 cm diam.; principal hyphae stout, sparingly branched, 15–73 μm diam. at the base, with sinuous, coiled and contorted extremes at the periphery of the mycelium. Gemmae spherical, pyriform, clavate or irregular, simple or catenulate. Zoosporangia often fusiform, naviculate, rarely clavate or filiform; 194–600×24–50 μm; straight or bent, usually terminal, renewal usually by internal proliferation. Zoospore discharge saprolegnoid. Encysted spores globose, 8–10 μm diam. Oogonia very abundant, terminal, lateral or frequently intercalary, often catenulate; mainly spherical, pyriform, often naviculate or rarely obovate, dolioform or filiform when intercalary; (29–) 60–140 (–160) μm. Oogonial wall smooth, very rarely with one papilla or with lateral projections or with apical outgrowth; pitted. Oogonial stalks frequently stout and straight when short or bent, curved or many times coiled, slender, tapering toward the end, often branched; (24–) 190–400 μm long. Oospheres maturing. Oospores subcentric type I, filling the oogonium; spherical or ellipsoid; (1–) 15–50 (–70) in number; (12–) 19–30 μm diam. Antheridia absent, or very rarely present. Antheridial branches when present principally diclinous (35%) and androgynous (14%), occasionally monoclinous (7%), branched. Antheridial cells simple or branched; apically or laterally appressed. Fertilization tube not observed (Figs. 1–4).

HOLOTYPE: Argentina Tierra del Fuego, Ushuaia, Olivia River, on floating litter; February 2000, leg. M.M. Steciow, LPS N° 45828; culture collection N° 746.

ETYMOLOGY: referring to the freshwater site of collection: the Olivia River where this new species was found.

## 4. Discussion

*S. oliviae* Steciow appears to be related to *Saprolegnia ferax* (Gruith.) Thuret. Both species have smooth oogonia and variable number of antheridial branches, often lacking. However, *S. oliviae* has only subcentric oospores which can be up to 70 per oogonium whereas *S. ferax* has fewer (up to 40) mainly centric oospores [4]. The form and the diameter of the oogonia as well as the nature of oogonial stalks are also different between the two species.

It is important to note that the type of oospore, the absence and type of antheridial branches when present, and the measurements of oogonia are very constant features in this new Argentine species. There was little variation in type and size of zoosporangia in different temperatures; they are fusiform, naviculate, rarely clavate or filiform and reached a mean range in length of 194–600 μm. The shape of oogonia remained constant, mainly spherical, pyriform or dolioform (smooth, rarely with one papilla or with lateral projections or with apical outgrowth; pitted).

The initial growth rate was faster at higher temperature (25°C), but after 20–30 days there was little difference in mean diameter of colonies from the other two temperature regimes.

Oogonial production was unaffected by the different temperatures; all colonies developed oogonia, but mature oospores were less abundant at lower temperatures and the number of oogonia and the oospores were proportionally lower as the temperature decreased.

*Saprolegnia hypogyna* (Pringsheim) de Bary appears to resemble the Argentine species because it also lacks an-

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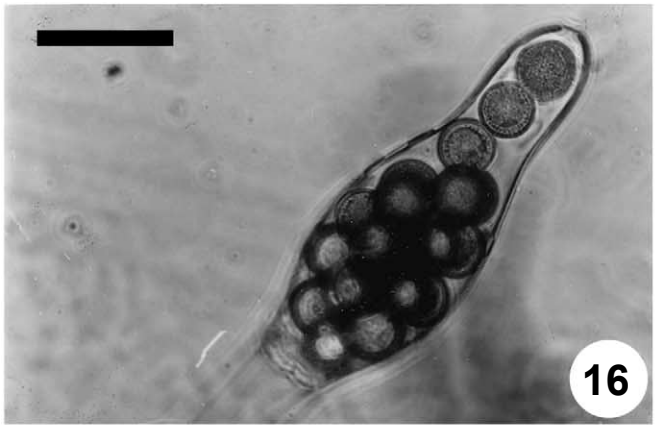
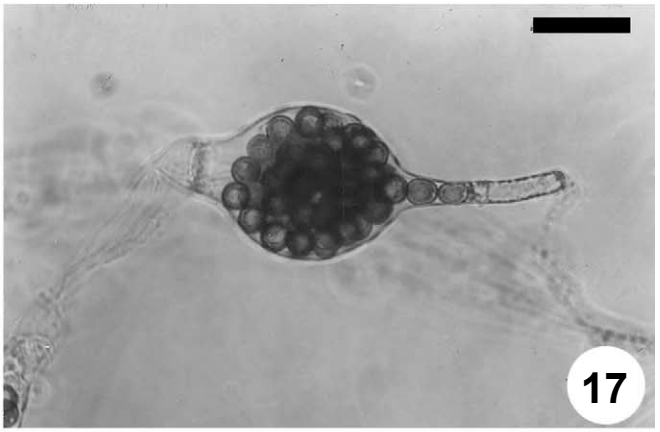
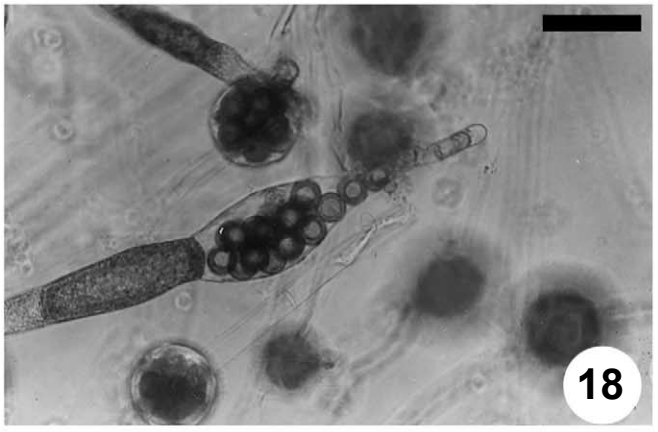
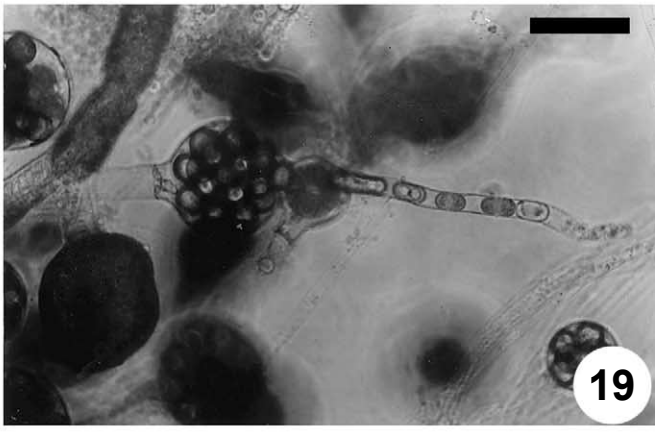
Fig. 3. 16–23: *S. oliviae*. 16: Naviculate pitted oogonium with subcentric oospores 17–19: Oogonia with apical projection containing oospores. 20: Branched oogonial stalk with spherical oogonia. 21,22: Pitted spherical oospores and pyriform oogonia with subcentric oospores. 23: Obovate oogonium. Scale bars: 50 μm.



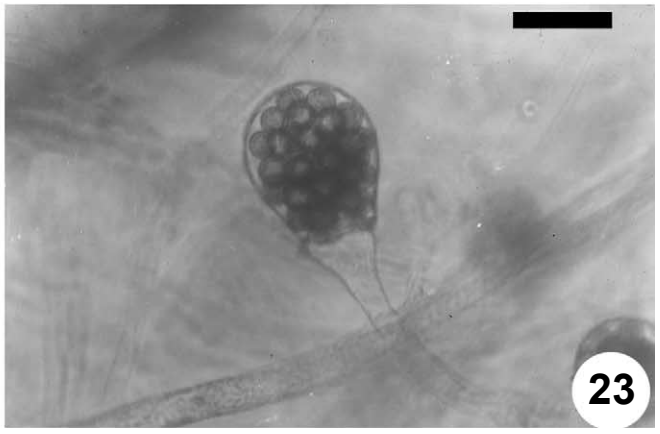









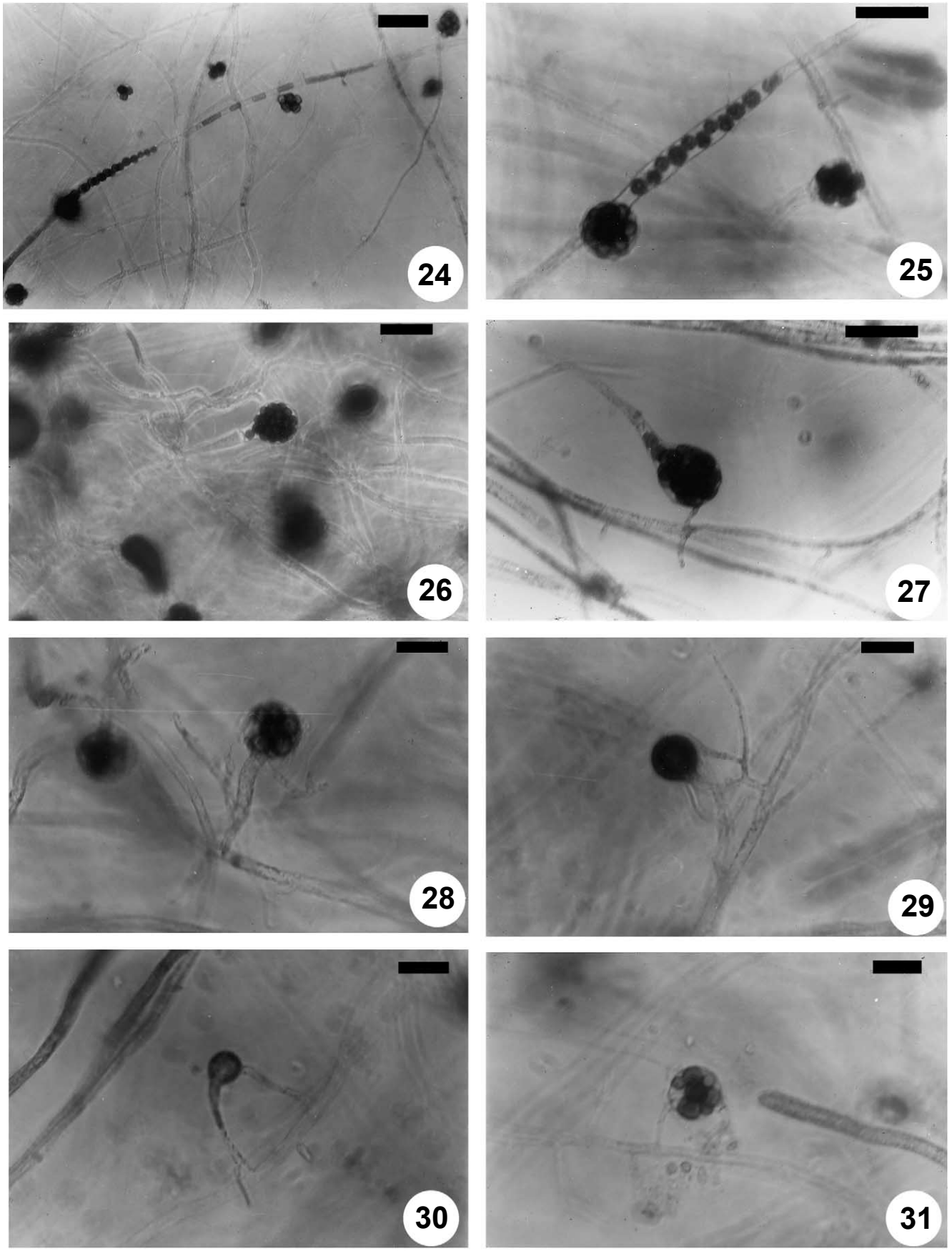



Fig. 4. 24–31: *S. oliviae*. 24,25: Characteristic intercalary and filiform oogonia 26,27: Diclinous antheridial branches. 28,29: Androgynous antheridial branches. 30,31: Monoclinous antheridial branches. Scale bars: 24,26,28–31 = 100 μm; 25,27 = 50 μm.

theridial branches. However *S. oliviae* is different from this species as it lacks the characteristic hypogynous antheridial cells. The number and type of oospores per oogonium is also very different.

*Saprolegnia diclina* Humphrey, with centric or rarely subcentric oospores, also differs in having antheridial branches almost always present, diclinous, rarely monoclinous, at times androgynous ones with also fewer oospores which are formed after prolonged period of time [4].

*Saprolegnia parasitica* Coker, with spherical subcentric oospores, is mainly parasitic on fish and their eggs, and differs from *S. oliviae* in having diclinous antheridial branches, very rarely androgynous, often wrapping about the oogonium and its hypha, persistent oogonia formed only after prolonged period of time, and lesser oospores per oogonium.

*Saprolegnia australis* [10] and *Saprolegnia longicaulis* [11], with subcentric oospores, are also different from *S. oliviae* in having predominantly diclinous antheridial branches and at times monoclinous ones, whereas the antheridial branches are lacking in *S. oliviae*, the proposed new species from Argentina.

## Acknowledgements

The author wishes to thank the Universidad Nacional de La Plata and the Argentine National Research Council (CONICET) for its financial support of the research through a Grant to Gernot Vobis (PMT-PICT 0382/97-99).

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