Ultrastructural Morphology and Phylogeny of *Henneguya* sp. (Myxozoa) Infecting the Gills of the Teleostean Fish *Cyphocharax Gilbert* (Curimatidae) in Brazil

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The phylum Myxozoa Grassé, 1970 constitutes a heterogeneous group of microscopic parasites that occur in fish, and that in many cases are lethal for their hosts [1]. Among the numerous species comprising this group, those of the genus *Henneguya* are highlighted for their occurrance in different geographic regions, and the large economic impact that they have on fish populations. Presently, this taxon envelops nearly 200 species, of which about 25% infect Brazilian freshwater fish (rarely marine) [2, 3]. Here a *Henneguya* spp. found infecting the gill filaments of a Brazilian freshwater fish is described, with basis on ultrastructural and phylogenetic data.

Specimens of *Cyphocharax gilbert* Quoy and Gaimard, 1824 (Fam: Curimatidae) (common name: Sairú) were collected in the estuarine region of the Guandú River, city of Seropédica, State of Rio de Janeiro, Brazil. Dissection revealed parasitic cysts containing myxospores located in the gill filaments of some of the analyzed specimens. Isolated myxospores were observed by optical microscopy with differential interference contrast (Nomarski), whereas the infected fragments were processed for transmission electron microscopy (TEM). Parasitized tissue samples were also prepared for molecular studies. Extraction of genomic DNA was performed using a GenEluteTM mammalian genomic DNA kit (Sigma), and the SSU rRNA gene was amplified with the primer pairs 18e/ACT3r and ACT3f/18r. The alignment of obtained sequences and the molecular analysis for the Kimura 2-parameter model were performed in MEGA 5.05 software. A phylogenetic tree was constructed for Maximum Likelihood.

The parasite form cysts in which developmental stages appear located in the outermost region, while mature myxospores occupy the central region. The latter have a fusiform body with two tails, for which they were identified as belonging to the genus *Henneguya*. The body of the myxospores is about ~12.5 μ m long, ~5.0 μ m wide and ~3.8 μ m thick; tail length is ~14.7 μ m (Figs 1a and b). The two polar capsules are symmetric, measuring ~ 6.2 μ m in length and ~ 1.6 μ m in width. Each polar capsule contains an isofilar polar filament coiled in 9-10 turns. The binucleate sporoplasm is located in the basal portion of the myxospore and contains numerous sporoplasmosomes. (Fig. 1c). The fragment of genomic DNA amplified and sequenced gave rise to a consensus DNA sequence of the SSU rRNA gene composed of 2002 bp.

A comparative molecular analysis of the obtained SSU rRNA sequence with related species, namely from South America, allowed confirmation of the information resulting from the ultrastructural observations, as it placed the parasite within the genus *Henneguya* (Fig. 1d) [4].

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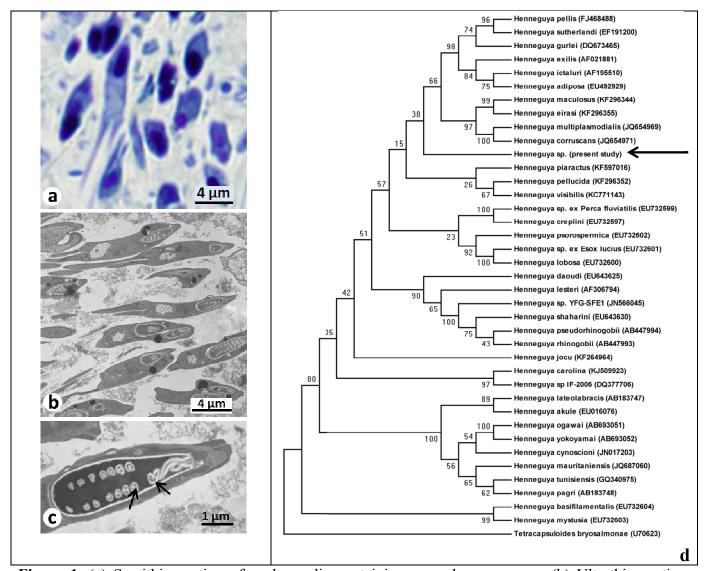


Figure 1. (a) Semithin section of a plasmodia containing several myxospores. (b) Ultrathin section showing several myxospores sectioned at different levels and presenting two polar capsules. (c) Ultrastructural detail of a polar capsule showing the organization of the polar filament (arrows). (d) Maximum Likelihood tree of the SSU rDNA sequence of *Henneguya* sp. and other Myxozoa species. The numbers on the branches are bootstrap confidence levels on 100 replicates.