

Morphological and Molecular Characterization of a Myxosporean Parasite Infecting the Skeletal Musculature of an Ornamental Fish Species from the Amazon River

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The freshwater teleost fish *Symphysodon aequifasciatus* Pellegrin, 1904, commonly known as blue discus, is endemic of the Amazonic basin in Brazil, Peru and Colombia. Endowed by a beautiful shape and coloration, this is a popular ornamental species that presents high commercial value worldwide. Its overexploitation has, however, been leading to the decline of wild populations, but other factors may be involved, such as parasitic diseases. Parasites of the class Myxosporea Bütschli, 1881, and more specifically of the genus *Kudoa* Meglitsch, 1947, are important pathogens of fish, both at the economic and at the ecological level [1]. The study here presented relies on ultrastructural and molecular data to characterize a species of *Kudoa* that was found infecting the muscle of several specimens of blue discus captured from the Amazon basin in Brazil.

Infected tissue was observed and photographed using the light microscope (LM) for measurement of fresh cysts and myxospores, and then prepared for transmission electron microscopy (TEM) and molecular analysis. The SSU rRNA gene was sequenced using both eukaryotic and kudoid-specific primers. For inferring phylogenetic relationships, neighbour-joining (NJ), maximum parsimony (MP) and maximum likelihood (ML) methodologies were performed in MEGA 5.05.

Prevalence of infection was estimated at 80%. Numerous whitish fusiform pseudocysts were observed among the skeletal myofibers (Fig. 1(1)). Myxospores quadrangular with rounded ends in apical view, measuring $5.3 \pm 0.7 \mu\text{m}$ in length and 7.3 ± 0.5 (6.8–7.8) μm in width. Myxospores wall composed by four symmetric valves, smooth and united along a curved suture line, and surrounding four pyriform polar capsules, equal-sized and measuring $3.8 \pm 0.5 \mu\text{m}$ in length and $2.5 \pm 0.5 \mu\text{m}$ in width. The polar capsules were located at the same level, two by two in opposite sides of the myxospores longitudinal axis, and each containing a polar filament coiled in 2 turns (Fig. 1(2) and Fig. 1(3)).

The morphological and molecular data obtained identify this parasite as a possible new member of the genus *Kudoa* that clusters together with *Kudoa orbicularis*, the only other freshwater SSU rRNA sequence of this genus in GenBank. The phylogenetic analysis further reveals tissue tropism as an important evolutionary signal for *Kudoa* (Fig. 1(4)) [2].

References :

- [1] J. Lom and I. Dyková, *Folia Parasitologica* **53** (2006) p. 1.
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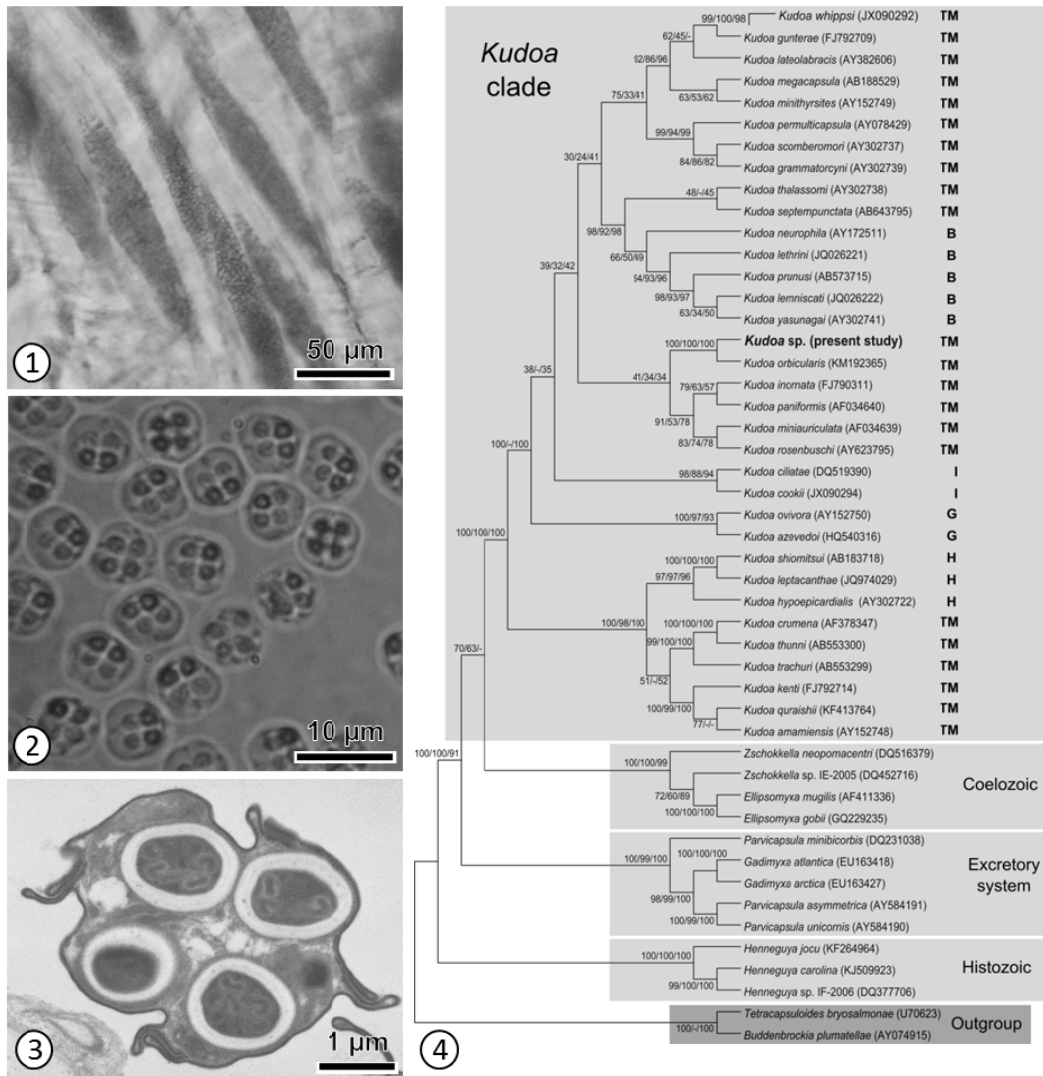


Figure 1. (1) LM micrograph showing numerous pseudocysts in the muscle; (2) LM micrograph showing numerous free fresh mature myxospores, after rupture of a pseudocyst; (3) TEM micrograph revealing the overall ultrastructure of a myxospore in transverse section; (4) NJ, MP and ML tree for the SSU rRNA sequences of *Kudoa* sp. and other selected marine myxosporeans. GenBank accession numbers in parentheses after the species name; TM, trunk muscle; B, brain; I, intestine; G, gonad; H, heart.