Molecular taxonomy of Spirochona gemmipara (Ciliophora, Chonotrichia), an epibiont living on the gills of Gammarus fossarum. First results.

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In memory of Denis Lynn (1947-2018)

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ABSTRACT. The chonotrichs (Chonotrichia, Ciliophora) live settled on diverses appendices (antennas, peropods, gills, etc.) of several kinds of marine and freshwater crustaceans (Decapods, Nebaliaceas, Isopods, Amphipods, ...) ¹. The chonotrichs reproduce principally by budding, and exhibit complex life cycles driven by the mouthing of their hosts. The chonotrich taxonomy was established in the milestone book of Jankowski [1973] ¹ which described more than 40 genera and 100 species living all around the world. Ultrastructural data are known for some species ¹, ² and molecular data are actually restricted to 2 species only ⁵, ⁶. Here, we present the SSU seq of Spirochona gemmipara sampled in several localities between Geneva and Lausanne.

MATERIAL AND METHODS

The material was sampled in different watercourses between Geneva and Lausanne (Switzerland) in autumn 2018. Water flow was roughly estimated, air and water temperatures were measured. Stains and wood branches were displaced with a metallic hook; moving gammadis were collected in a metallic strainer (Ø 20 cm) and immediately preserved in a small glass jar. In laboratory, gammadis were kept living in an air-conditioned cabinet, at 10 °C, with a 12/24 h illumination period. Gammarids were examined under a Wild M5 binocular and gills were dissected with a pair of fine tweezers; the edge of colonized gills were cut off with indestructive scissors. Gills fragments were examined and framed under an Olympos BH2 microscope, then processed for DNA extraction.

DNA extractions were done with a Qiagen® Blood and Tissue Extraction kit, and/or in guanidin, after Chomczynsky and Sacchi (1987). SSU gene fragment were amplified with primers CL+F [TGG TAG TAG ATT GGA CWA CCA] and CL+R [TCT RAT YGT YTT TGA TCC CYT] (Stoeck, 2014); products were purified with a Roche® High Pure PCR Cleanup Micro Kit and then directly sequenced (with the amplification primers).

Sequences were edited in CodonCode Aligner and BioEdit.

Phylogenetic tree was constructed with Seaweed.

RESULTS AND DISCUSSION

The six new Swiss spirochones sequences form a solid group, facing 2 other solid groups, the isochonines and the chilodochonines. The "uncultured" sequence, coming from organisms not cytologically identified, could be an isochonine or a lobochonine (two close species) or one other chonotrich. The dysterids are considered to be chonotrich ancestors. The sequence clearly "roots" the three chonotrich groups.

Gammarids were collected in 3 rivers, and 6 stations. Rivers from station A (Gillon) and B (Gland) belong to the Rhone watershed, while rivers from station D, G and H belong to the Rhone watershed. The resolution between the new sequences is poor. This is probably due to the short SSU fragment (ca 600 bp) analyzed. So, it is actually not possible to know if the spirochones of the Rhone and Rhine watersheds are different.

LITTERATURE


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