

Molecular phylogeny of metchnikovellids (Microsporidia: Opisthosporidia).

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Metchnikovellids are hyperparasites of gregarines inhabiting the gut of polychaetes and some other marine invertebrates. Recent phylogenomic analyses demonstrated that metchnikovellids branch at the root of microsporidian tree and thus represent one of the key taxa for reconstructing the phylogeny of Opisthosporidia (ARM clade, which includes aphelids, rozellosporidia and microsporidia). Thick-walled cysts containing spores is a characteristic feature of metchnikovellids. The general morphology of cysts is a keystone in their classification developed 100 years ago and not yet evaluated by molecular means. Since 2010, six isolates of metchnikovellids have been found and investigated during our extensive screenings in the populations of polychaetes in the White Sea, North-West Russia. Using micromanipulations, single-cell PCR and whole genome amplification, we were able to amplify and sequence rRNA genes from genomic DNA extracted from the cysts of metchnikovellids and/or from the individually isolated gregarines infected with these hyperparasites. The sequences of metchnikovellids are fast-evolving and significantly divergent within the group. They share many In/Dels typical of higher microsporidia, but also show some unique and opisthokont-specific signatures. In contrast to *Amphiamblys* spp. and *Amphiacantha* spp. the studied species of *Metchnikovella* do not form a monophyletic clade. Apparently, this most numerous and species-rich genus is an artificial assemblage of genetically heterogeneous species. Poor morphological characters used to unify its members should be reconsidered following molecular findings. Supported by RFBR grant 18-04-01359.