Not all that looks like a Tubulinean is a Tubulinean: expectations and reality for the parasitic amoeba *Janickina pigmentifera* (Grassi, 1881)

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A parasitic amoeba living in the testes that occupy the caudal part of chaetognaths was firstly reported by Grassi in 1881. Later, two species Janickina pigmentifera and Janickina chaetognathi were re-isolated four times from different places in the Mediterranean Sea: from the strait of Messine, Naples, near Algeria, and near Villefranche-sur-mer. Janickina has a monopodial, or limax locomotive form similar to Hartmannella, Saccamoeba or Glaesseria (Amoebozoa, Tubulinea). Also, it was reported the presence of an "ocellus" in the cell similar to intracellular symbiont Perkinsela-like organism (PLO) of Paramoeba eilhardi (Paramoebidae). Only in 1980, Hollande presented the ultrastructural data of J. pigmentifera showing the kinetoplastid nature of the "ocellus" later also confirmed in Paramoeba. He also showed that the cell coat of Janickina was a stratified glycocalyx. It was not similar to the microscales of Paramoeba or glycocalyx of Neoparamoeba, but usual for Thecamoeba instead. These morphological characters were too contradictory, so the genus Janickina was placed incertae sedis. The studies of these amoebae were always illustrated with drawings and not light microscopic micrographs. Molecular phylogenetic data are not yet available for Janickina. Therefore, the genus remains incertae sedis today. We re-isolated Janickina pigmentifera from planktonic chaetognaths of the Bay of Villefranche. The first microphotographs of locomotive forms for J. pigmentifera were obtained. These amoebae showed a monopodial locomotive form typical for Tubulinea. For the first time, we obtained the 18S rRNA gene sequences of Janickina pigmentifera and its PLO. Contrary to our expectations based on morphology, the preliminary molecular phylogenetic analysis based on 18S rRNA gene showed that in spite of its morphological characters, Janickina pigmentifera grouped within the clade of Neoparamoeba as a sister to Neoparamoeba branchiphila. A similar result was obtained for molecular phylogenetic analysis of 18S rRNA gene of PLO from J. pigmentifera. It grouped as a separate long branch – sister to the PLO of Neoparamoeba branchiphila. This result undermines the morphological concept of the Tubulinea.