



Review of the PhD thesis

PhD candidate: **Martin Kostka**

reviewer: **Miroslav Oborník**

The PhD thesis called "Molecular Phylogeny of Selected Protists" written by Martin Kostka is composed of two parts: General review covering very briefly introduction to the "protist" phylogeny, taxonomic status of slopalinids and *Blastocystis*, and also introduction to eopharyngians and amoebozoans, supplemented by the list of relevant author publications and brief conclusion. The introductory part also contain chapter (1.5.) called "data filtering" describing the way how the data are treated by using slow-fast analysis and SlowFaster program in particular. The review takes 21 pages, including 4 pages of references.

The second part of the thesis is composed of attached reprints and manuscripts published (4) or submitted (2) by PhD candidate. There are four papers that are published in respected journals *Molecular Phylogenetics and Evolution* (three papers, IF 3,99) and *BMC Bioinformatics* (IF 3,49). Two manuscripts cover the taxonomy and molecular phylogeny of amoebozoans and were submitted to *Acta protozoologica* (IF 1,22). Martin Kostka is the first author of three published papers (two *Mol Phyl Evol*, *BMC Bioinformatics*), second author on two other (*Mol Phyl Evol* and one of the papers submitted to *Acta Protozoologica*), and the third author of one of the amoebae papers (submitted to *Acta Protozoologica*).

The general introduction covers all appropriate aspects of the field. However, since organisms studied in this thesis belong to three different eukaryotic groups (Chromalveolata, Excavata, and Amoebozoa), the use of term "protists" looks like a making a virtue of necessity through the connection of non-related organisms by the traditional name with no taxonomic and phylogenetic relevance. I personally think that at least the addition of papers submitted to *Acta Protozoologica* was a kind of shot in the eye, because they definitely do not increase the relevance of the thesis and at the same time, the thesis is becoming even more fragmented. I have to avow that I am a bit disappointed by the absence of detailed view on eukaryotic phylogeny.



Since the candidate has worked with organisms belonging to three different supergroups, these groups should be characterized at full length.

On the other side it has to be noted that PhD candidate showed excellent publication record. I would personally highlight two papers: the one that solve the phylogenetic position of *Protoopalina intestinalis*, and the paper describing new automated program SlowFaster to perform slow-fast method in a much more user friendly way. The paper that demonstrated phylogenetic position of *P. intestinalis* within stramenopiles was actually the first trustable attempt to solve this question. This paper was published in the time where sequences wrongly annotated as opalinids were already in the public databases. It is always more difficult to submit paper that contradict previously published results. SlowFaster paper has a very high citation potential, because of quite limited operability of manually performed Slow-Fast analysis or the computer analysis computed in less user friendly environment. This program will help to solve some phylogenetic problems caused by LBA and other artifacts. Since so far it was very difficult and time consuming to perform slow-fast analysis (manually), it will become much more frequently used from now. I really appreciate this phylogenetic tool designed to deal with the most usual phylogenetic artifacts.

I have a single formal remark: I would appreciate if the citations in the introductory text would be in the form of written names, instead of numbers referring to the list of cited literature. For readers, it is not very pleasant to search each citation in the list.

Although most of attached papers went already through the peer-review process, I have two questions covering both, the general introduction and attached publications:

- 1) The title of the thesis sounds, with the current knowledge of the eukaryotic phylogeny, confusing. First, no such group as "protists" formally exists, because protists are definitely not monophyletic in their nature and these single celled eukaryotes (formerly called "Protista") actually belong to all



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eukaryotic supergroups. This is not only a taxonomic question. This incomprehension has led in the past to the factual separation of protistology (including parasitology) and algology and establishment of two separated scientific fields, which both work with closely related organisms, however, they very rarely communicate to each other. Mainly in the case of PhD candidate I would expect let say progressive view of the taxonomy and phylogeny of these organisms. So, what are your opinions on the phylogeny of eukaryotes and the composition of eukaryotic supergroups? How should we change the way of teaching protistology and algology to bring them closer each other?

- 2) Can you please explain, on what ground you have excluded the ciliate sequences you have obtained from your samples when amplifying *Protoopalina* SSU sequences?? There is just mentioned that ciliate sequences were excluded from further analysis. Since slopalinids had been also thought to belong to or to be related to ciliates, this should be explained comprehensively.

In a summary I can conclude that Martin Kostka PhD thesis definitely fulfills all matters required by the Charles University to cap the doctor degree and I fully recommend this thesis for the defense.

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