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Evaluation of the PhD thesis of MSc. Emmanuel Arriaga-Varela

The PhD thesis of Emmanuel Arriaga-Varela was carried out at the Department of Zoology of the Charles University of Prague, under the supervision of Dr. Martin Fikáček and in the framework of the BIG4 project funded by the EU.

The first section of the introduction is informative and the aims of the PhD thesis are clearly outlined at the end. I really enjoyed reading the well-written second section. This indicates that the candidate is able to contemplate on issues beyond the challenging field of beetle taxonomy. It becomes also very clear that he made himself familiar with the relevant literature, very obviously with studies linked with the phylogeny and taxonomy of Hydrophilidae, but also with important older and recent works dealing with issues of evolutionary biology on a theoretical and philosophical level.

Spelling errors should be avoided or reduced to a minimum in a thesis. A misspelled important taxon in the first lines of the introduction (Sphae*rd*iinae) is nothing to be proud of. There are also different spellings of the name of the supervisor with varying use of diacritic marks. Generally, parts of the thesis (e.g. abstract: "*specific* …diversity", introduction: ". when did they originate*d*", "is *a* example of a lineage nested in Coleoptera *wich* evolutionary history…") would have benefitted from a linguistic check by a native speaker of English. However, these flaws are only marginal issues.

The general chapter "Material and method" (sic!) is short, but this is no problem as a more detailed account of the applied techniques is given in the individual studies included in the cumulative thesis. A very positive point is the transparency and availability of data accumulated in the thesis project, achieved by storage in suitable data bases.

The main part of the thesis is formed by seven studies, all of them with MSc Arriaga-Varela as 1st author, 4 of them published or in press, one submitted, and two as manuscripts not submitted yet. In all cases serious journals with peer review were chosen.

Chapter 1 is a thorough revision of species of the genus *Cercyon* from the Greater Antilles. It combines a classical taxonomic approach with modern molecular techniques, i.e. DNA barcoding for larval identification. The study is accurate, includes a useful key, descriptions of five new species, a key, and also information on larvae. As a whole it is a very positive contribution.

Chapter 2 is the description of a new genus of African coprophagous Hydrophilidae, named *Evanesternum*. Like the previous study, this contribution was thoroughly prepared and is very well illustrated. The monophyletic origin of the species assigned to the new taxon is convincingly shown. However, the possibility that they might be nested within an existing genus of Megasternini was not evaluated in this study (see below, Chapter 7).

Chapter 3 deals with exotic and previously very obscure flower-visiting species of *Cycreon*. Several new species of this fascinating group are thoroughly described. Field observations and analyses of gut contents provided important information on the biology.

Chapter 4 is an accepted study on hydrophilid species living in very unusual microhabitats, rotten cacti (in Mexico). *Agna* is another taxon with a very unusual specialization, very interesting from an ecological and evolutionary perspective. Thorough taxonomic work is combined with modern DNA barcoding methods. A discussion is missing in this study, but characters were carefully evaluated in differential diagnoses and comments.

Chapter 5 deals with fossil hydrophilid beetles embedded in amber and their reconstruction using µ-computed tomography (µ-CT). The great potential of this technique for reconstructing the morphology of extinct (and extant) taxa is convincingly demonstrated, even with rather poorly preserved material. Extinct species of several genera are carefully described and well-funded hypotheses concerning the phylogenetic placement are presented. One of the studied species, originally considered as a member of Hydrophilidae, is assigned to the cucujoid family Phalacridae with good arguments. The unusual composition of the Baltic amber fauna of Hydrophilidae (and other groups of beetles) is discussed.

The topic of the unpublished study forming chapter 6 is the phylogeny of terrestrial Hydrophilidae and diversification patterns. With multigene analyses and time estimates the candidate demonstrates that he is able to apply advanced phylogenetic methods. Patterns of shifts to terrestrial environments are convincingly reconstructed. Primary shifts to leaf litter were identified as a common evolutionary trend, followed by secondary shifts to excrements,

carrion etc. in different lineages. The entire study is very interesting and on a high scientific level. I have no doubts that it will be published in a suitable per-reviewed journal in the near future.

Chapter 7, also not published yet, contains a molecular phylogeny of Megasternini. Maximum likelihood and Bayesian inference are used, and I agree with the preference for the topology obtained with the latter approach. The pattern suggests that repeated and bidirectional ("bidirected") continental interchanges must have taken place in the Eocene, a very interesting and somehow surprising result. The results of the analyses also show that *Evanesternum*, the genus described in chapter 2, is in fact deeply nested within *Cercyon*, and therefore the generic status not justified. As stated by the authors, the phylogeny underlines that far-reaching changes in the classification of the group are required.

Generally, the thesis is very well illustrated. The quality of some of the SEM images could be better, but they are certainly sufficient to document the characters in question. The line drawings are excellent.

The relevant literature is fully covered. The layout of the list of references is a little bit unorthodox and the formatting not 100% consistent, but these are only marginal issues.

A very positive asset of this thesis (and the candidate) is that a very broad spectrum of techniques was used. This ranges from the taxonomic basics, line drawings, SEM, µ-CT, analyses of molecular data, and the dating of phylogenetic events.

What I really like and appreciate is that this thesis extends from the essential painstaking taxonomic work over innovative morphology, modern molecular systematics, and then finally the level of evolutionary biology. This is exactly how modern biodiversity investigations should be organized and carried out, honouring the "basic" but essential taxonomy, but also applying innovative techniques and approaches, and also keeping the larger evolutionary picture in mind.

Considering everything, despite of some minor flaws, I rate this thesis with the mark 1.3 (1.0 would be the best mark in our German system) (magna cum laude).

Prof. Dr. Rolf G. Beutel

**Questions:**

- Does the presence of several autapomorphies (or synapomorphies of several species) justify the erection of a new supraspecific taxon?
- How do the larvae cope with the switch to terrestrial environments? What are the physiological challenges?
- Do changes in the diet play a role?
- Where are eggs deposited?
- What could be the food substrate of hydrophilid larvae living below the bark of banana plants?
- How did these terrestrial beetles switch between continents?
- Can you outline a standardized procedure for standardized taxonomic descriptions providing maximum efficiency and information content, and full accessibility of data?
- Is maximum parsimony or Bayesian inference more reliable for reconstructing phylogenetic relationships based on morphological data?