

Review of the PhD thesis “*Patterns and processes in desmid communities: Insights from functional and phylogenetic approaches*” presented by Mgr. Helena Bestová

Reviewer: Francesco de Bello (CSIC-Spain, University of South Bohemia-Czech Republic)

The PhD thesis by Helena Bestová is possibly among of the best theses I had the pleasure to review in the last few years. It is generally well written, nicely synthesized, and relatively easy to follow, demonstrating some notable pedagogical attitude from the PhD candidate, a good knowledge of the field, and good communication skills. The main aim of the thesis is to assess the structure of freshwater phytobenthic green algae desmids communities, and to do so from a functional and phylogenetic point of view. The candidate was able to move beyond more traditional taxonomical approaches in these communities with the help of the supervisors and excellent international collaborations. By using functional and phylogenetic approaches it was thus possible to assess external and internal drivers of these communities' structure, which allowed answering type of questions linked to the ecology of these specific organism types and to community ecology in general. The doctoral candidate nicely combined large-scale sampling (Ch 2 and 3), of communities under markedly different climatic and local conditions (e.g. water pH), with experimental work (Ch 4), cultivating monocultures of species from a given genus (*Micrasterias*). Chapter 1 provides a general introduction together with some overall summary of the main findings. One of the chapters (Ch 2) has been already published in a good general ecology journal (*Oecologia*) and another is likely going to be accepted sometimes soon also in a very good general ecology journal (possibly *Proceedings of the Royal Society B*). The final manuscript (Ch 3) is almost ready for submission in very good specialized journal in freshwater biology. As such, the thesis is composed, mainly, by 3 manuscripts which will be likely all get published well. This is a very good result for a PhD thesis, although I must admit that most theses I have reviewed so far included 4 published/publishable manuscripts, or more. Otherwise the data analysis in the different chapters is generally robust and the main findings well supported by the results, which is greatly appreciated for a PhD thesis. Therefore, I formally confirm the thesis presented by Mgr. Bestová has more than sufficient merits to allow its public defense.

The structure of the thesis is coherent and understandable. At the same time the choice to add the section “Key results and conclusions” within Chapter 1, i.e. “General introduction”, was somehow questionable. From a traditional approach, and for some aspects even logical, I would have preferred to have a general closing chapter, say Chapter 5, including a more elaborated discussion of the different results from the different chapter. I missed a bit tighter connections between findings, with a more elaborated text, together with the already existing summary of the key results and conclusions. In this sense, it is a bit strange to see the main results and conclusions before having seen the chapters (I would say the Abstract has this function). It is difficult to judge if it is a good summary or not, before reading the individual chapters. Anyway, the lack of a nice closing chapter seems to be a common ‘trait’ of many of the current PhD theses, so I

think it is quite generalized limitation. This often includes an over-exceeding attention to different publications/manuscript without fully demonstrating (at least in some thesis) the ability to connect findings across chapters. Since this synthesizing ability of the candidate is already partially apparent in the Chapter 1, I expect that the thesis defense will better stress the connections between chapters.

Chapter 2 and 3 use roughly the same data set, from either a functional or phylogenetic point of view respectively. Both studies are interesting and, while I might understand the multiple pragmatical reasons behind such a splitting, I am not completely sure this is very clear/logical for readers of the thesis. Even the indices chosen and the approaches are sometimes not consistent, and the reasons behind their choice remain a bit unclear. Of course the readers of the two manuscripts, when published, might not need to make tighter connections, but as a reader of the thesis I missed a more convincing justification of the splitting of the analyses, the different analytical approaches, and a final connections of the findings. At the beginning of chapter 3, the third sentence actually refers to Cadotte et al. 2013, which interestingly advocates for combining functional and phylogenetic information. This sentence is, instead, used to support the solely focus on phylogenetic diversity and excluding functional diversity. This reads as being a bit strange. In the same introduction it is said that phylogeny is a good proxy of traits (second paragraph in the introduction Ch 3) when in other sections the authors cite Gerhold et al. 2015, or many others, showing that phylogeny alone can be sometimes of little information about traits. Of course these potential problems are possibly in the less developed (but still almost ready for submission) manuscript. In this sense I would be happy to see results for Table 3.1 and Figure 3.2, also including functional diversity. It would be actually interesting to have a look at these results during the thesis defense, while using the same index. The problem here, in fact, is that I expected that the same index should be used for taxonomic, phylogenetic and functional turnover (see also de Bello et al. 2010 JVS) which I do not think it is the case so far. In a way, as nicely summarized by the candidate, the significance of phylogenetic diversity, in response to several gradients (Ch 3), as opposed to the effect of the same gradients on the considered traits (Ch 2), indeed shows that phylogenetic diversity brings information of potentially unmeasured functional traits (rather than being, in general a good proxy of given traits). This further advocates for combining functional and phylogenetic diversity. In this sense, approaches to combine functional and phylogenetic diversity has been proposed (beside Cadotte et al. 2013, see various works by Sandrine Pavoine with trait dissimilarity at different evolutionary periods; see also de Bello et al. 2017 *Methods in Ecology and Evolution* with various Czech colleagues). I would like the candidate to elaborate more about these issues, including the potential need to sqrt-transform phylogenetic distances, as suggested by Letten & Cornwell (2015; *Methods in Ecology and Evolution*).

Another, to me, relatively small incongruence between Chapter 2 and 3 is the number of plots (and regions) considered. In Chapter 3, a total of 102 plots were considered, which I understand were a subset of the 148 plots considered in Chapter 2, which also included more regions. Again I might understand the

reasons behind of such sub-setting but, if I did not miss it, I feel that the reasoning and justification was not clear enough for the readers of the thesis (and potentially for informed reviewers of the manuscript resulting from Chapter 3). It now looks a bit strange, even a sort of cherry picking approach, which I am sure it is not the case of course. But it needs to be better discussed. This is not very convincing, at the moment, and the potential confounding effects of using different datasets (and indices), can limit the potential for synthesizing effects of FD (Ch2) and PD (Ch3).

The last manuscript (Ch 4) is a nice, concise and very well focused experiment aiming at demonstrating classic allometric relationships and trade-offs on size related traits linked to species functioning and demographic patterns. The results are very clear and demonstrate nicely, for the exemplary genus considered, the importance of the classic Kleiber's law on the relationship between size, metabolic rate, population growth (and possibly even mortality etc). Interestingly such "population growth rate and its scaling are independent of the geometry of the organism, beyond the effect of body mass or volume". The results, although considering "only" a given genus, with 24 strains, show that the Metabolic Scaling Theory is generally valid also at the microorganism scale. In this respect, and also following Chapter 3, I am not particularly surprised that phylogenetic "corrections" did not make a great importance on the Metabolic Scaling, given the relatively small phylogenetic extent of the study (one genus). In this sense I would be happy to discuss, more broadly, with the candidate the meaning of phylogenetic corrections in trait analyses. Otherwise I felt that the study possibly could have increased considerably its interest if more than 1 phylogenetic clustered group of organisms was considered. I am not completely sure that this potential limitation is fully discussed in the manuscript but the authors do claim that "Micrasterias possibly represents a key transitional group in the evolution of allometry, offering a unique opportunity to investigate geometrical constraints of cell morphology on growth rates". I am not sure what "possibly" means here and what "transitional group" implies, as opposed to other groups. This is possibly because I do not know these organisms very well, as likely other readers in Proceedings, or similar. More explanations, possibly during the thesis defense, will be surely welcome for non-experts like me.

While the experiment in Chapter 4 is clear and effective I wonder if there could be a limitation, or actually an opportunity for future works. The experiment used in Chapter 4 is, if I understand, basically run within one environmental condition (with constant temperature, light etc). This is actually quite fair for the questions being asked and for the resulting manuscript. At the same time, to my limited knowledge the Kleiber's law, the Allen's rule (which based on surface per mass/size used here) and the r/K trade-offs all indicate that these scaling rules should provide different adaptations to different environmental conditions (at least temperature, if not disturbance/density). Even they could provide different adaptations to fluctuating environments. Given that I understand these experiments are not overly large, and relatively short-term, maybe they could have been expanded over multiple temperature and soil pH, also following part of the results obtained in Ch 2 and 3. This is what I missed, but maybe it is already on-going in other projects. Anyway, I am not sure I understood well the

experimental design: was it 24 species/strains x 4 replicates? Not sure how the well-plates were organized, blocking factors in the data-analyses etc. I got a bit lost on the design to fully appreciate the experimental effort needed. In principle I would be happy to hear from the candidate if accounting for Metabolic Scaling Theory across environmental conditions is of some significance and, most important, how this could be tested. Similarly I found that it was a pity to speak about ecological niche of species, in Ch 3, and then losing the ability to test these niches, in controlled conditions, in relation to traits, their allometry, and population growth. In this sense I would be happy to hear more from the candidate about the potential connections between Kleiber's law, the Allen's rule, the r/K trade-offs, the CSR scheme etc. and how potential connections could be expanded on an experimental basis.

Finally, the thesis has several mentions to competition. I do not know much the model organisms but I wonder how competition works in these systems and with this type of organisms. I ask this from a complete ignorance point of view. At the same time, the sentence in the introduction "Evidencing the biotic interaction in microbial communities is difficult" is possibly not very satisfying in this sense. I know that in plants can be size symmetric or asymmetric, depending on ecosystems/productivity, traits etc. And in this thesis size related traits seem to be central. What is the relation expected between size and competition? If I am not completely wrong not much is said how competition is working in the study model of this thesis. There is even some rather indirect/vague mention in the thesis that higher relative growth rate increases competition, or at least some connections are briefly attempted. Hence I would be happy to hear how the competition is expected to work in this system, compared to others, and what the candidate expects as the relationship between relative growth rate and competition. Do different views on competition~relative growth rate connects with different ecological frameworks on how competition works, for example comparing predictions from Tillman and Grime, or the classic r/K trade-offs (on which largely the CSR scheme by Grime is built)? Again, following also the nice papers by Violle et al. 2011 and 2012 (cited in the thesis) with the already classic species pairwise experiments on bacterivorous protists, I expected (1) more clear predictions of competition on these systems and (2) that the experimental approach followed in Chapter 4 could be expanded to test hypotheses of competition raised in Chapter 2 and 3. I would be happy to hear how this could be done and the (dis)advantages of using experimental vs. field sampling based tests of community assembly mechanisms.