Review of the PhD thesis

Local adaptation of the rare herb *Aster amellus* in fragmented landscape

by *Jana Raabová*

submitted to Charles University, Prague

Reviewer: Jan Lepš

The thesis consists of three "chapters", in fact, three independent papers, one of them published, and two prepared for publication (as a matter of fact, I believe that both of them will be finally published in proper scientific journal). In all the three papers, the candidate is the first author. All the three papers deal (from various points of view) with local adaptation of rare herb *Aster amellus*. In fact, the motivation of all the papers is applied – within active nature conservation measures, the possibility to enforce small local populations of endangered species with individuals of the same species from other (but larger) local populations is often considered. This measure is seen as controversial by many, and there are arguments both in support, but also against this measure. Probably the most important of them is the local adaptation of small populations, which might be disrupted by introduction of non-adapted individuals. From this point of view, the presented thesis provides useful information for decision making in nature conservation. Beside this, the results are interesting by themselves, contributing to the population biology theory.

I like the selection of the model species – in particular, the fact that the species is found in two ploidy level (two cytotypes) is itself interesting, so that one can study both, the local adaptation within cytotype, and also the ecological differentiation of the two compared sytotypes. The results are, in my view, also an important contribution to the theory of evolutionary ecology.

The author uses wide range of methods, including "molecular" analyses characterising the genetic differentiation of populations, pollination experiments, transplant experiments, and finally, the classical phytosociological relevés, which might undoubtedly improve the communication with nature conservation authorities.

The candidate demonstrated her competence, ability of independent work, and finally, ability to finish the work and publish the results. I sincerely believe that the final result of the thesis will be three papers published in good peer reviewed journals. **Consequently, I definitely consider the thesis as a sufficient basis for awarding the PhD to the candidate.**

For discussion, I would like the candidate to answer the following questions (or to express her opinion):

All the results are carefully analysed with modern statistical methods. I must admit that for me, the analysis of such complicated models with mixture of fixed and random effects (some of which are nested), is a nightmare. Could you explain the way, how you have formulated the statistical models to be used (e.g. how you decided which of the interactions to drop), and how you have decided what should be the "error tern", i.e. denominator in the F-tests and quasi-F tests? Further, it seems to me, that in Chapter 1 you have used different approach

from Chapter 2, where, as I have understood, you have used the approach of contrasts. If so, why? (Related question regarding the Figures: How were calculated the standard errors of mean? I believe that even their calculation should reflect the hierarchical structure of the design. (and then they might become considerably larger (?)).

Second question is rather practical. You have demonstrated that the most sensitive phase of the life cycle is the establishment of an individual. Moreover, whereas the development of an individual in the field is a long process, in a garden, the individuals can be multiplied quite fast. If we would decide for an "active" measure to increase the population size, would not be the safest way to multiply individuals in the garden and use them as "autochtonous" transplants to enforce the local population. Other possibility is to use the produced seeds (which would make sure that only adapted individual would establish). What would be your recommendation to practitioners?

Third question: With decreasing area of calcareous grasslands, the habitat is more and more fragmented (particularly in comparison with situation 100 – 50 years ago). Nevertheless, the species is clearly confined to the open habitats. Do you think that the habitats are more fragmented now than used to be, say, in the Holocene Climatic Optimum? Or, at least in some more distant past?

In addition, I have some further (minor, or "editorial") comments on individual papers:

Chapter 1

I highly appreciate the amount of work in the reciprocal transplants, and I consider the nine localities as maximum what was probably feasible to do. Nevertheless, for comparison of vegetation and soil characteristics of localities of different cytotypes, it would be better to have more localities with more phytosociological relevés and soil characteristics. This is fairly less laborious than the transplant experiments, and would provide better support for the habitat differentiation. The power of the tests (Table 1) is very low (otherwise, I would be surprised that the intentionally selected high and low productivity localities would not differ in soil properties). Perhaps, such a comparison was done in other paper(s) of the supervisor's group?

I am afraid that seedling survival as used here is not what I would consider seedling survival. Did I understood correctly that it is number of seedling alive at the time of census divided by the number of seeds sown? I consider this characteristic ecologically meaningful, but I would stress somewhere that it is in fact seedling survival multiplied by germination (or seedling establishment) rate.

p. 36 What is *M. annua*?

Chapter 2

Methods – Why did you used the two-tailed Mantel test? I think that the question in hand calls for the one-sided hypothesis and one-sided test (I can not imagine mechanism causing, e.g. negative correlation between geographic and genetic distance); the one sided test would result in significant (instead of marginally significant) correlations (making some journal reviewers happy).

p. 49 …data transformed according to van der Maarel (1979). This paper suggests several transformations (in fact, the flexible transformation is also mentioned there), so the reference is not unequivocal, I expect you have used the "ordinal" transformation. Moreover, I think that this paper introduces Eddy's own extension of Br.Bl. scale (with 2 split into three categories [a, b, m]), which is finally subjected to various transformations; you have in Appendix degree 2 split into two categories, + and -. Which scale and which transformation was actually used? [I expect the ordinal transformation.]

p. 52 … Most of isozyme variation was within populations (83.7%; $F_{ST} = 0.16$; p<0.001). – Which null hypothesis is tested here?

Chapter 3

I know that there is a strong pressure to publish the PhD results. Nevertheless, would not be better to wait one year and include the results from 2008 (e.g., your plants did not flower in 2007, and this might be very useful parameter to characterise your plants – do you plan to follow the fate of individual plants also in 2008?)

As already expressed above, I recommend the thesis for the defence.

České Budějovice, 27 April 2008   Jan Lepš