



Přírodovědecká fakulta
Faculty of Science

Jihočeská univerzita
v Českých Budějovicích
University of South Bohemia
in České Budějovice

STATEMENT OF THE BACHELOR THESIS OPONENT

Name of the student: Paraskevi Tziortzouda
Study program: Biological chemistry
Department/Institute: Department of molecular biology, University of South Bohemia
Thesis title: Creation of *Drosophila melanogaster* mutants for multiple sirtuin genes (Sirt2, Sirt6 and Sirt7)

Supervisor: RNDr. Alena Krejci, Ph.D.
Supervisor's affiliation: University of South Bohemia, Faculty of Science

Point scale¹ Points

(1) FORMAL REQUIREMENTS

Formal and graphical quality of the thesis	0-3	2
Ability to work with literature	0-3	2
Language and stylistics	0-3	2
Formal requirements – points in total		6

(2) PRACTICAL REQUIREMENTS

Fulfillment of the aims	0-3	3
Ability to understand the results, their interpretation, and clarity of the results, discussion, and conclusions	0-3	3
Discussion quality – interpretation of results and their discussion with the literature	0-3	2
Experimental difficulty of the thesis, independence in experimental work	0-3	3
Contribution of the thesis to the knowledge in the field and the possibility to publish the results (after eventual supplementary experiments)	0-3	3
Practical requirements – points in total		14

POINTS IN TOTAL (MAX AWARDED)

MAX 24 20

¹ Mark as: 0-unsatisfactory, 1-satisfactory, 2-average, 3-excellent.

Comments of the opponent on the thesis:

Paraskevi Tziortzouda's thesis aimed to create multiple mutants for sirtuin genes in the fruitfly, *Drosophila melanogaster*. It is really very ambitious project resulting in double/triple mutant fly. In essence, the student had to absorb basics of *Drosophila* genetics, learn logic of CRISPR/CAS9 methodology and of course obtain training in molecular biology techniques. The amount of work might be even sufficient for master thesis, but the quality of the written part is the weak point of the whole thesis.

Paraskevi's thesis has classical structure, detailed introduction, result section and discussion. Sometimes I have impression, that the author did not control for typos, and did not remove automatic formatting done by word processing program. This is slightly sad, because too many typos are degrading impression from otherwise amazing work. I am not trying to criticize and score cheap points as a reviewer. I just would like to emphasize that very solid work can be degraded and you should avoid it next time - when writing another technical report, master thesis or, job application or CV.

The important problem is the literature. Some references are numbered, sometimes authors are listed, and in one case (page 18) everything is present including DOI. It is not formalism from the reviewer.... It is simply important to provide reader with somewhat unified text structure - and that did not happen here. Specifically, it is often very difficult to identify original work. Probably the first reference (or one of the first) is number 9 on page 12.... Legend of Table 1 is rich for references, which are completely missing in reference list.

Technical aspects of the work. Given the number of work done, I would expect slightly better gel picture for the Figure 14, where contamination with marker was loaded....

Minor points and mistakes (individually would not be a problem, but all together are not negligible).

- Latin names are usually written in *italics* in scientific literature. Indeed, the author follows this consensus, but only sparingly.
- The genus name starts with capital, the species name does not... Please, avoid these mistakes next time (page 9: *melanogaster* should not start with capital, similarly on page 20 *Streptococcus thermophilus*, where *thermophilus* should not start with capital).
- if you talk about temperature, make sure that you include degree sign, not just a square (page 6, page 23, page 24)

Specific questions that Paraskevi can address during her defense, based on her knowledge and information from the literature:

- On page 21 you say that off-target effect is a problem of CRISPR/CAS9 approach, but more with complex genomes, not in *Drosophila*. Please, comment on possibilities how to avoid off target effect in *Drosophila*.
- Could you, please, show Fig. 16 and explain what is loaded and what it means? I am afraid I do not understand why the PCR product was digested, if the mutant was obtained by recombination.

As I wrote before the thesis is of excellent experimental quality, huge amount of work was done and Paraskevi certainly achieved the aims she set. My only concern is that the written part is also important aspect of training and learning and should not be underestimated.

Conclusion:

In conclusion, I recommend the thesis to be defended with the grade excellent or very good, depending on quality of the presentation.

In Ceske Budejovice date 12th June 2017

David Doležel



signature