



Přírodovědecká
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Faculty
of Science

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

SUPERVISOR'S STATEMENT ON BACHELOR/DIPLOMA* THESIS

Name of the student: Lena Graf
Study program: Biological Chemistry
Department/Institute: Faculty of Science, University of South Bohemia, České Budějovice, Czech Republic
Thesis title: Transformation of marine protist *Diplonema papillatum* - characterization of selected cell lines
Supervisor: RNDr. Drahomíra Faktorová, PhD.
Supervisor's affiliation: Laboratory of Molecular Biology of Protists, Institute of Parasitology Biology Center, AS CR and Faculty of Science, University of South Bohemia, České Budějovice, Czech Republic

	Point scale ¹	Points
(1) FORMAL REQUIREMENTS		
Formal and graphical quality of the thesis	0-3	3
Ability to work with literature	0-3	3
Language and stylistics	0-3	3
Formal requirements – points in total		9
(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	3
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		15

* Choose one

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

Comments of the supervisor on the student and the thesis:

Lena first came to our laboratory (Molecular Biology of Protists – PI: prof. RNDr. Julius Lukeš, CSc.) in September 2018. I offered her that she could work on the project of transformation of the marine protist *Diplonema papillatum*.

Lena immediately agreed and started with verification of the cell lines that I obtained after the transformation of *Diplonema papillatum* with constructs that should lead to alpha-tubulin replacement with puromycin resistance marker (*Puro^R*) or its tagging with red fluorescent protein (mCherry).

Before Lena joined the lab, the reproducible transformation protocol of *D. papillatum* was already established in the laboratory, however we had problems with the integration of the constructs to the expected position of *D. papillatum* genome.

Goal of the Lena's project was mainly to verify whether at least some of the newly obtained cell lines finally have the construct targeted to the expected position of the genome, which would mean a breakthrough in this project. It worked in 2 cell lines that we decided to further characterise.

Lena continued with verification of the expression of the constructs by SL RT-PCR and Western blot analysis, measuring of the growth curves as well as checking the knock-out cell line using transmission electron microscopy.

With not much of previous experience from molecular biology, Lena oriented herself very quickly in the laboratory and soon she mastered various techniques of molecular and cellular biology.

Lena successfully finished all experiments, which were planned for her project and as a cherry on the cake during writing the thesis we submitted the manuscript entitled: "Targeted integration by homologous recombination enables in-situ tagging and replacement of genes in the marine microeukaryote *Diplonema papillatum*" to the *Environmental Microbiology* journal where Lena is a co-author. The results of the thesis are part of the manuscript that was recently accepted for publication.

Based on the plethora of experimental methods used, which led to obtain important results, Lena has certainly acquired the essential skills needed to continue in Natural Sciences and I wish her all the best in her future career.

In conclusion, I was very happy to supervise Lena and have her in our laboratory and in my opinion her thesis is of very high standard. My recommendation is therefore to grant Lena Graf the Bc degree.

Conclusion:

In conclusion, I **r e c o m m e n d** the thesis for the defense and I suggest the grade 1.

In České Budějovice

date July 27th, 2020

signature