Přírodovědecká fakulta
Faculty of Science
Jihočeská univerzita v Českých Budějovicích
University of South Bohemia in České Budějovice

STATEMENT OF THE BACHELOR/DIPLOMA* THESIS SUPERVISOR

Name of the student:	**Stefanie Pezelj**
Study program:	**Joint Biological Chemistry**
Department/Institute:	**Department of Molecular Biology & Genetics**
Thesis title:	***Generating a fluorescently tagged MARK2 fusion protein as a marker for the basolateral membrane of preimplantation stage mouse embryo blastomeres.***
Supervisor:	**doc. Alexander W. Bruce Ph.D.**
Co-supervisor (Consultant):	**Mgr. Lenka Gahurová Ph.D.**
Supervisor's affiliation:	**Faculty of Science, University of South Bohemia**

	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
Logic in the plan of the experimental work	0-3	3
Experimental difficulty of the thesis, independence in experimental work	0-3	2
Contribution of the thesis to the knowledge in the field and the possibility to publish the results (after eventual supplementary experiments)	0-3	2
Practical requirements – points in total		16
POINTS IN TOTAL (MAX/AWARDED)	27	25

Overall classification: 3-excellent

* Choose one

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

Eventual additional comments of the supervisor on the student and the thesis:

It was my pleasure to have Stefanie working in my laboratory. She was/ is well liked by myself and other members of the research group (particularly, my post-doc and her co-supervisor, Lenka Gahurová, with whom she worked closely on a daily basis). She proved to be a diligent and careful worker and met our collective anticipation of what is expected of a Bachelors' level research student (both during her practical time in the lab and perhaps more so in her written thesis).

As may be known, the main focus in our laboratory is the study of the acquisition of one of three cell-fates during mouse preimplantation embryonic development; such work is technically demanding and requires extensive training periods, particularly in terms of embryo handling and micro-manipulation, before meaningful experiments can be conducted. Moreover, the study of development pays no respect to the chronology of an individual student's teaching timetable. It is for these fundamental reasons that the projects we can offer are not always best suited to Bachelors level students; although it is also true to say this largely depends on the individual in question. Therefore, in the case of designing a project for Stefanie, we decided to 'play it safe' and offer her a relatively straight forward work plan that was almost exclusively molecular biology based. Consequently, Stefanie was tasked with the aim of cloning a specific gene cDNA (encoding the basolateral membrane localised polarity protein MARK2) into an existing plasmid vector habouring the cDNA for the Venus fluorescent protein. The aim was to derive a recombinant cDNA that would encode a fluorescent MARK2 fusion protein, that could then be *in vitro* transcribed to yield a mRNA that could then be microinjected into single blastomeres of the developing mouse embryo and as act as a reporter of the onset and maintenance of intra-cellular apical-basolateral polarity (specifically the basolateral component), from the 8-cell stage onwards. I am pleased to confirm that Stefanie was able to fully achieve this aim and confirmed her derived construct's germane expression and localisation within cells of the mouse preimplantation stage embryo; moreover, this construct is now available for use by other members of the group to facilitate their own research aims.

The one slight criticism I might make of Stefanie is that she did not write up her research project immediately after finishing in the lab. I would have preferred this, as I think it is better for both student and supervisor that the process is concluded whilst the memories are still relatively fresh! However, this I am sure is not a problem unique to Stefanie amongst her peers, and as a collective of supervisors on this programme we should be more forceful in suggesting students write-up more expediently. Notwithstanding this minor point, I must acknowledge the good job Stefanie made of writing her thesis and to pay particular credit to her theoretical understanding, especially evident in the Discussion section she wrote.

Overall, I am more than satisfied Stefanie has received a solid foundation in experimental design, execution, interpretation and presentation and would be able to successfully draw on this training in her future career (should she decided to stay in science). I have not the slightest hesitation in recommending her Bachelors project thesis is accepted as successfully defended (with the grade 3-excellent).

Conclusion:

In conclusion, I, **Alexander W. Bruce**

recommend/ ~~**do not recommend***~~**.**

In: České Budějovice

Date: 17th June 2019