



OPPONENT'S REVIEW ON BACHELOR/DIPLOMA^{1*} THESIS

Name of the student: Mirjam Odermatt

Thesis title: Investigation of stability and dynamics of photosynthetic pigment - protein complexes using optical spectroscopy

Supervisor: RNDr. David Bína, Ph.D.

Referee: Mgr. Tomáš Fessler, Ph.D.

Referee's affiliation:

	Point scale ²	Points
(1) FORMAL REQUIREMENTS		
Extent of the thesis (for bachelor theses min. 18 pages, for masters theses min. 25 pages), balanced length of the thesis parts (recommended length of the theoretical part is max. 1/3 of the total length), logical structure of the thesis	0-3	3
Quality of the theoretical part (review) (number and relevancy of the references, recency of the references)	0-3	3
Accuracy in citing of the references (presence of uncited sources, uniform style of the references, use of correct journal titles and abbreviations)	0-3	3
Graphic layout of the text and of the figures/tables	0-3	3
Quality of the annotation	0-3	3
Language and stylistics, complying with the valid terminology	0-3	3
Accuracy and completeness of figures/tables legends (clarity without reading the rest of the text, explanation of the symbols and labeling, indication of the units)	0-3	3
Formal requirements – points in total		21
(2) PRACTICAL REQUIREMENTS		
Clarity and 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 the results and their discussion with the literature (absence of discussion with the literature is not acceptable)	0-3	3
Logic in the course of the experimental work	0-3	3

^{1*} Choose one

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

Completeness of the description of the used techniques	0-3	3
Experimental difficulty of the thesis, independence in experimental work	0-3	3
Quality of experimental data presentation	0-3	3
The use of up-to-date techniques	0-3	3
Contribution of the thesis to the knowledge in the field and possibility to publish the results (after eventual supplementary experiments)	0-3	3
Practical requirements – points in total		27
POINTS IN TOTAL (MAX/AWARDED)	48	(0-48) 3

Comments of the reviewer on the student and the thesis:

In this bachelor thesis Mirjam Odermatt studied isolated violaxanthin-chlorophyll-a-binding protein (VCP) of *Nannochloropsis oceanica* using optical spectroscopy with a specific focus on time-resolved fluorescence spectroscopy. Utilizing pigment and intrinsic tryptophan fluorescence, Mirjam studied dynamism of the pigment-protein interactions in VCP with respect to non-photochemical quenching.

The extent of 43 pages (including the list of references and appendix) and quality of this work already fulfils the criteria set for the master thesis. The thesis is structured into eight chapters: Preface, Introduction, Aims of the thesis, Materials and Methods, Results, Discussion, Conclusions and Publication Bibliography. I will comment on each of the chapters separately.

Preface is very well written. It summarizes key processes in photosynthesis and slowly guides the reader towards the main topic of the thesis, protective mechanisms that prevent damage to the light-converting complexes.

Introduction is well structured and comprehensive, easy to follow and well referenced. This chapter covers 6 pages with 4 figures. It covers (i) photoprotective regulatory mechanisms in photosynthesis, (ii) summary of available knowledge about VCP in *Nannochloropsis oceanica* and (iii) intrinsic protein fluorescence with a focus on tryptophan fluorescence.

Aims of the thesis are clearly stated.

Materials and Methods

This chapter covers 4 pages and is generally well written. It is divided into 7 subchapters covering sample preparation, and steady-state absorption and fluorescence spectroscopy as well as excitation spectroscopy, time-resolved fluorescence spectroscopy and circular dichroism. I highly appreciate the depth of description of the time resolved spectroscopy of tryptophans.

Comment: I would personally prefer if Figure 6 would be in section Results. Nevertheless I get the point that the author wants to demonstrate the technical point of view here (IRF etc).

Results. This chapter is a key part of the thesis. It is rich in unique data of high quality. The data

³ Enter the number of points awarded.

are well presented in 9 subchapters and documented. Data shown here are publishable in a reputable journal.

Comment: before publishing this data I would recommend acquiring additional biological replicas and subsequently more statistics. For example, Figures 7, 10, 12 would benefit from having error bars. I would also add statistics and an error estimate to Tables 1, 4, 5. However, I do fully appreciate the formidable amount of work behind this thesis, especially when taking into account the available time to finish the bachelor project in a double-degree study programme.

Discussion is well written. It is divided into 2 logical subchapters and it covers 4 pages. The comparison of the results with already published data truly helps to put new findings into the context of what is already known. It is also well referenced and easy to follow.

Comment: I would appreciate a longer reflection about the future plans.

Conclusions are of appropriate length and well written.

Suggestions and questions, to which the student has to answer during the defense.

Mistakes, which the students should avoid in the future:

Suggestions and mistakes, which the students should avoid in the future:

- (1) I would recommend using more positive controls in the future. For example proving that you can reproduce tryptophan lifetimes of already published proteins would be a good start.
- (2) I would recommend to refrain from using words like “immensely blue-shifted” (as seen on page 31) in scientific texts.

Questions:

- (1) How many tryptophans there are in the VCP, how many of them are exposed vs buried inside?
- (2) Theoretically, you could attribute the change in fluorescence lifetimes of tryptophans you observe to either VCP aggregation or conformational rearrangement within individual complexes. How would you distinguish between those two scenarios?

Conclusion:

**In conclusion, I
r e c o m m e n d**

the thesis for the defense and I suggest the grade Excellent.⁴

In date

České Budějovice, 22 January, 2021

⁴ You can suggest a grade, which can be modified during the defense. If the reviewer is not present at the defense, the grade will be (1), Satisfactory (2), Unsatisfactory/failed (3), Unsatisfactory/failed (4).

