



OPPONENT'S REVIEW ON MASTER THESIS

Name of the student: Bc. Kateřina Jaklová

Thesis title: Characterization of nuclear and cytoplasmic interactome of tick-borne encephalitis virus capsid protein

Supervisor: RNDr. Martin Selinger, Ph.D.

Referee: Doc. RNDr. Jindřich Chmelař, Ph.D.

Referee's affiliation: Department of medical biology, Faculty of Science, USB

	Point scale ¹	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	2,5
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		20,5
(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
Completeness of the description of the used techniques	0-3	3

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

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	2
Practical requirements – points in total		26
POINTS IN TOTAL (MAX/AWARDED)	48	46,5

Comments of the reviewer on the student and the thesis:

In her thesis, Kateřina Jaklová focused on disclosing an interactome of TBEV C protein while fractionating to nuclear, cytosolic and membrane proteins. She used proteomics for the analysis of fractions that she separated in first, optimizing part of the work.

The work is written in nice English, sometimes I would write it differently, but it reads well. I have no objection or criticism to the work as I think it is excellent. The introduction goes from general description of flaviviruses to more details about TBEV and the protein of interest itself. The methodology part is detailed and carefully written, accompanied adequately by figures and tables. Results are described chronologically, showing the progress of the work. Discussion is written well, comparing obtained results with the literature, trying to speculate on possible meaning of found interacting partners.

Some minor comments and suggestions: In some places, author jumps from passive voice to ichform, this should be unified. In methods, there is a formula for counting cells in Bürker chamber, using multiplication factor of 2, which is not explained. Did author dilute the cells in trypan blue? Since there is low number of interacting proteins, would it be possible to use some positive control to prove that the fractionation method works properly?

Questions:

1. The results suggest that there is nothing much going on in the interaction between TBEV C and cell proteins despite high number of proteins identified in TBEV C interactome by Martin Sellinger in his JBC paper. How do you explain this discrepancy?
2. I saw a lot of figures showing the distribution of TBEV in Europe and Asia, and I was always wondering, why the distribution in Asia forms relatively sharp borders in the south, where I don't see any mountain range or something. Is there some reason for that?
3. Of what I know, subcellular fractionation usually needs ultracentrifuge. You were using several methods with highest speed of 16000g, for which I suppose ultracentrifuge was not used. Are the techniques, based on the kits and table-top centrifuges, as reliable as ultracentrifuge based techniques?
4. You tried to eliminate DNA mediated binding. Shouldn't this binding be also of the interest? Is something known about it?
5. One of the binding partners is a histone and you speculate that by interaction with histone, the cell replication is impaired. Why the virus would do that? How is it beneficial for it?

Overall the thesis is very good and I believe that Kateřina has the potential to continue in science, if she decided to.

Conclusion:

In conclusion, I

r e c o m m e n d

the thesis for the defense and I suggest the grade excellent (1) .²

In **České Budějovice** date 16.5.2023

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signature

² You can suggest a grade, which can be modified during the defense based on the presentation. However, if the reviewer is not present at the defense, the grade will not be counted. Grades: excellent (1). Very good (2), Good (3), Unsatisfactory/failed (4).