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Bc. Thesis Review

Author: Kamila Machová

Title: RNA biology of symbiotic bacteria in insects

This thesis is focused on non-coding RNAs and RNA modification enzymes in endosymbiotic bacteria of insects, which is truly neglected topic in this field of research. Kamila's thesis raises numerous questions about molecular mechanisms that lead to working translation in highly reduced symbiotic genomes. I'm convinced that finding the correct answers will result in really good scientific publications.

The thesis is written in English and supposed to be 46 pages long, however, it looks like this number should be 48. The thesis is well structured and was easy to follow. Though I think that Kamila made a good piece of work and her thesis is excellent, I have to do my job and be critical.

Language of Kamila's thesis is not flawless. I spotted misspellings. For example, in the section Annotation: there is "Anotation" instead of "Annotation". There are also numerous missing commas in sentences. Sometimes a sentence is not correctly structured and it is not easy to understand. Since Kamila is a bachelor student, I consider the level of her English really good.

Introduction is 6 pages long and it is divided into two logical blocks: Symbionts and ncRNAs. I would personally include one short paragraph describing differences between obligate (e.g. *Buchnera aphidicola*) and facultative endosymbionts (e.g. *Sodalis glossinidius*). In the ncRNA part, I'm missing a brief description of all ncRNA that occur in bacteria. I'm aware that Kamila focused on rRNA and tRNA, but there are definitely more RNAs in a bacterial cell that can be called ncRNA. The text is easy to read, but I think that paragraphs should be a little bit more linked together.

The Material and methods section is 5 pages long. It describes methods and experimental design that Kamila used. Whole Kamila's work was performed *in silico* and there were no experiments. I have to pinpoint that Kamila not only used existing software, but also wrote her own scripts, which is great. I'm satisfied with this section except of missing reference for the Infernal software.

The Results section is 9 pages long and it describes ncRNA, ncRNA modification sites and ncRNA modification enzymes in studied endosymbiotic lineages. There is really a lot of information in this text. The only thing I did not like there was table 2, in which Kamila uses a

term S symbiont for obligate co-symbionts. If terminology is not changed and I remember it correctly, the term S symbiont is used for facultative endosymbionts (e.g. *Sodalis glossinidius*). I think that using the term S symbiont in the way that Kamila did, might be misleading for some readers.

The section Discussion is 9 pages long, which is a very nice surprise, since students usually struggle with discussing their results in detail. I enjoyed this part of the thesis very much. The only thing I did not like and consider erroneous is Kamila's preference for one explanation over the other ones without any experimental data. She prefers idea of transport of missing tRNAs from a co-symbiont or from a host against other possible hypotheses that she suggests.

The section Conclusions and future prospects is 1 page long. Kamila clearly summarizes main results and suggests additional *in silico* analyses of endosymbiotic genomes that should be performed. She is also aware of the necessity of experimental results, however, she does not suggest anything specific.

The section Bibliography is 12 pages long and it contains 113 references. I think this is more than adequate number for a Bachelor thesis.

The thesis further contains Supplementary material section that is 4 pages long and it comprises description of scripts and data used in Kamila's study.

QUESTIONS:

Can you explain existence of obligate mutualists (P symbionts) with high GC content?

Do you know any other ncRNAs (other than tRNA and rRNA) that occur in bacterial cell?

Why there were no facultative endosymbionts included in your analyses? They represent a transition state between free living bacteria and obligate symbionts.

Why do you prefer a hypothesis about tRNA import over other explanations?

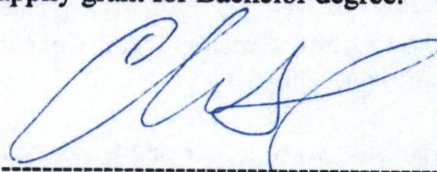
What (do you think) is a difference between obligate endosymbiont and an organelle? Can be symbionts that lost their ability to maintain their independent translation called organelles?

You are aware of the fact that experimental data are needed. I want to ask: What experiments are needed and how would you perform them?

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In conclusion, I think that Kamila Machová fulfilled the requirements of a Bachelor thesis. I would like to evaluate the thesis as "excellent" and happily grant for Bachelor degree.

18.5.2016 České Budějovice



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