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## **Supervisor's opinion regarding PhD thesis Mitochondrial gene expression in trypanosomatids by Michaela Procházková**

Míša was a member of our lab between the years of 2009 – 2015. She joined the lab as a master student since her former supervisor left for a different job offer. At that time she was working on a putative methyltransferase MT420 from *Trypanosome brucei*. This enzyme was supposed to methylate spliced leader RNA and thus be involved in RNA transcription. Míša clearly showed that this is not the function of the enzyme, and hypothesized based on detailed bioinformatic analysis that MT420 might be responsible for methylation of a release factor, protein that is responsible for translation termination. We will hear a lot about the mitochondrial release factor during Misa's presentation since this protein became the heart of her research. Funnily enough I had no intention to work on mitochondrial translation and its termination, but we followed the results one by one slowly, but surely. Once Misa joined our lab as a PhD student, she was given a different project. She was supposed to implement a strategy to inducibly control gene expression in *Leishmania* parasites by using so called destabilization domain. I don't have to remind the audience that at the time functional genomics was not a straightforward method in *Leishmania* as some of these parasites lack RNAi pathway and they possess plastic genomes hindering studies of essential genes by classic conditional double knock-out strategies. So our plan was to tag essential mitochondrial proteins with destabilization domain, which stabilize or destabilized the protein based on the presence or absence of a molecule called Shield-1. Long story short, this did not work, at all. Míša and others spend valuable time on strategy that was not viable. However in a meantime, she continued to work on her favourite proteins, MT420 and release factors. Since this story was actually developing quite nicely, at the end we decided that this is going to be her PhD topic.

I think we will both agree that we went through too many ups and downs during her PhD. I was still getting used to be a PI and Míša was struggling with reproducibility of her experiments and with various experimental approaches that were not giving us expected results. Míša's independence and overconfidence got her sometimes into the troubles as she would follow wrong lead and misinterpreted data. However during the time we learned how to work together and this huge achievement is culminating today.

I would like to also highlight Míša's biggest assets for the work in the lab. She is very well organized, smart as a monkey (as we would say in Czech), and she is extremely efficient. When she is in the lab she wants to work. That was especially handy at the time when we did not have technician and Misa in her free time helped with regular and common tasks of the research lab. I thanked her for that.



Míša left the lab at the beginning of 2016 and joined the lab of dr. Pavel Plevka at CEITEC. When recommending her for this position I felt this is going to be a good fit for her. She will be solving structures of various viruses by state-of art methods of cryo-EM. Projects that require tedious analysis, computer skills and dedicated long hours of work. Here she can use her strengths; effectiveness, organization skills and drive for the results.

Unfortunately it took us long two years to write and publish her first-author paper. The process was slowed down by my second maternity-leave and her absence. It was painful and nerve-racking time. Especially the 5-months long re-submission to Molecular Microbiology followed by rejection. Anyway, now I can just say: what does not kill you, it makes you stronger.

In my opinion, Míša experienced everything what PhD programme offers: new methodologies, dead-end projects, excitements, falls followed by rises, conferences, international stay, intellectual stimulation, boss who is sometimes not happy, team work, friendships.

I strongly believe that her PhD thesis fulfils all the requirements to gain a title of Doctor of Philosophy at the University of South Bohemia.

I wish her bright future and a lot of solved structures.

In České Budějovice, 18.6.2018

A handwritten signature in blue ink, appearing to be 'P. Plevka'.