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Supervisor' review of dissertation thesis by Anna Nenarokova

Anna came to us from Russia. Soon after her formal acceptance, when the visa process was not going as fast as she hoped, she sent me an e-mail asking if I could speed it up, because Putin had some weird statements on TV about intelligent people leaving Russia etc., and that with such ominous signs, she wants to get out ASAP. It worked out after all and she arrived with her husband Serafim, who soon became a graduate student in bioinformatics in the lab of Martin Kolisko, so getting a PhD at our institute became a family affair. Anna had no problem in passing all the exams, as her background both in molecular and cell biology and informatics as well as her English were pretty strong.

One hurdle she had to deal with from the start was the fact that I as her supervisor have poor knowledge of the current field. In fact, I used to do first DNA analyses, phylogenies etc. in the institute, but that was decades ago, and I did not keep up at all with the pace in bioinformatics and totally rely on in-house experts and collaborators. For the ambitious and capable Anna finding this out at some stage was certain disappointment, but she had to live with that. Indeed, Anna came when our first dedicated bioinformatician Pavel Flegontov was leaving and the experienced Anzhelika Butenko was still in Ostrava, so most of her PhD she was on her own, drawing methodological advice mostly from her husband and Kika Zahonova, who later on became affiliated with us, but was always working in Bratislava. On the bright side, this made Anna a genuine PhD who after being given usually just a general outline of what and why should be studied, had to figure things out. Most of the time, Anna did and pretty fast on top of that. Consequently, she was right from the start a very good presenter, as she managed to grasp the complexities of the projects, and not only present them, but was almost eager to have the opportunity to answer tough questions addressed at her during our labmeetings, which may sometimes stress others. Another of her strengths was sharp thinking, as she frequently came with smart proposals for wet-lab experiments. Here the small disadvantage was that as someone who never worked at the bench, she could not distinguish which experiment takes days and which one several months.

While her main project throughout her PhD was the bizarre genetic code of *Blastocrithidia*, which she will primarily talk about today, as a capable bioinformatician Anna became involved in a number of other projects, which is

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reflected by her co-authorship on a set of solid papers. I am not going to describe them one by one, as they are all attached to the thesis, but will only mention the mBio paper, where Anna is rightly the first author. Being a careful observer, during her work with *Blastocrithidia* Anna made an interesting finding that parasitic lineages tend to lose the nonsense mediated end joining pathway. It came as no surprise that the paper became editor's pick and may have a substantial impact.

Not included in her thesis is a study of the most complex mitoproteome described so far, led by Michael Hammond, which was accepted to Mol. Biol. Evol. just a few days ago. And more papers will result from the on-going projects. However, her main *Blastocrithidia* project turned out to be a hard nut to crack, and we decided to combine Anna's in-depth bioinformatic analyses performed on the bugs genome, transcriptome and proteome with extensive experimental work, led by Ambar Kachale and others. We all hope that we will eventually reveal the secrets of this unprecedented switch of the genetic code.

Anna presented her work at meetings in Czechia, but also gave convincing talks at meetings abroad. She stayed for 3 months in Tom William's lab at the University of Bristol, where she was among die-hard bioinformaticians and hopefully learned a lot. There, she participated in the annotation of the *Diplonema papillatum* genome and specialized on putative vs. real horizontal gene transfers.

With all that praise being said, I have to add that the two of us had our share of intense debates or sometimes even quarrels. We are quite different personalities, both on the stronger side of the spectrum I guess, which was not always helping. I like to rely on deadlines, while Anna sees them as something binding her freedom and creativity. A problem was also that with my poor insight into the methodology of her projects, I did not know whether certain task took her 10 or 100 hours, which I can still much better estimate for most of the benchwork protocols.

But that's just to say that not everything was perfect as in a standard family (and of course I have my share in that). It is without any doubt that during all those years, Anna became a valuable member of our lab. I believe she has everything to succeed in the current science if she decides to go in this direction and if she accepts that in our over-organized society, pretty much no one has the privilege to do whatever and whenever he or she wants. Anna will be even better off, if she improves her communication skills with her collaborators. But she is smart enough to know all that.

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Finally, I would like to state that the presented dissertation thesis fulfils, in my opinion, all postulations and I recommend it to be accepted as a partial fulfillment of the requirements for the degree of Doctor of Philosophy at the Faculty of Biology of the University of South Bohemia.

Thanks Anna for your good work.

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