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Supervisor' review of dissertation thesis by Binnypreet Kaur

Binny came directly from India and joined us when we were still in the old building. Since her knowledge of molecular and cell biology was rather rudimentary, in the first year she was involved in numerous small projects in the lab, in order to familiarize herself with laboratory work and science in general. In that part of her PhD she worked with *Trypanosoma brucei*, in particular was helping with dissecting the function of enigmatic catalase. The resulting story is now in press, with Binny among the coauthors.

When it came to the point that we needed to pick a serious topic, I gave her diplonemids, which at that time were newcomers to the lab more or less as she was. One thing was clear with Binny right from the start – she had the ambition to succeed. Hence, the choice of the topic did not make her happy at all, because she saw herself as a bystander of most what was going on in the lab, a marginal subject that unfortunate fell upon her to deal with.

I was trying to dissuade her from this opinion and hope I eventually managed. A key moment was that Dr. Drahomíra Faktorová was willing to become a co-supervisor of Binny, as my directorial duties did not allow me to spend as much time with her as I deemed necessary. Drahuška also soon took diplonemids close to her heart and along with first funding that we have subsequently received for studies of these strange critters, things started to pick up.

Still, with the dearth of knowledge, we all had hard time in figuring out how to proceed towards our main aim, namely to make *Diplonema papillatum* amenable to functional genomics. We were struggling with generating decent transcriptomes, defining proper conditions (diplonema grows under a wide range of them but changes its morphology, behavior and almost certainly also its metabolism), with obtaining contigs long enough for genetic manipulations etc. However, it soon became a genuine team work and Binny always could find someone to help her.

I will not describe Binny's long path, also not to steal her thunder. It suffices to say that it was not an easy one indeed. We had no clue whether we will be able to get any DNA into *D. papillatum*, not mentioning more sophisticated steps. It was here when Binny's stubbornness and Drahuška's guidance proved a pretty good combination, which brought first breakthroughs. While we still do not have the CRISPR/Cas9 method functional in diplonemids,

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thanks to both of these ladies we can now integrate DNA either randomly or in the target position, we can tag genes, pull down their interacting partners and do other tricks, which promise to help shed light on what these hyperdiverse and pretty ubiquitous protists are doing in the vast ocean. Moreover, they seem to have departed, in many aspects, from what is considered the "standard" eukaryotic cell, so it will be exciting to explore their mitoribosome, respiratory complexes, "missing" kinetochore proteins and so on.

Although it took a while to teach Binny the rigorousness of science both at the bench and behind the computer, she picked it all. In the process, she matured into a young ambitious scientist who can ask questions, design and perform experiments, interpret the data and write first draft. Besides fulfilling standard PhD duties, Binny presented at international meetings, and attended a methodological workshop in the US. Later on, she spent 3 months pipetting her fingers to the bone in the lab of our collaborators Gertraud Burger and Matus Valach in Montreal, in what turned out to be a productive stay, which resulted in a nice Nucleic Acids Research paper that is just coming out.

In my opinion Binny did very well and matured into a promising young researcher. It would strengthen her position if she adds a bit more of a team work, always attempts to get a broader picture, and will try more of the "out-of-the-box" thinking.

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 Binny for your dedication and good work.

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