

RNDr. Magdaléna Krulová, Ph.D.
The Faculty of Science
Viničná 7
128 43 Prague 2

Review of the Ph.D. thesis by Igor Grekov: Experimental murine leishmaniasis and its application for drug discovery and study of host-pathogen interactions.

The thesis is devoted to three subtopics: 1) refinement of experimental procedures relevant to experimental murine leishmaniasis, 2) using mouse model for mapping loci of susceptibility to leishmaniasis and 3) characterization of leishmanicidal effect of calcium ionophore calcimycin.

The introduction represents an extensive and well written literature review. It covers general information about *Leishmania*, clinical significance of leishmaniasis, methods of diagnostics and treatment, cultivation of *Leishmania* and, finally, mouse models for genetic studies of leishmaniasis.

The aims of the study are clearly formulated. All of them have been accomplished.

Material and methods describe all the experimental procedures used in the research including statistical analysis.

The results include four chapters dedicated to the publications representing the outcome of the research. Two of the papers, where Igor Grekov is a co-author, have been published in highly impacted international journals: Nature Protocols (IF = 8.632) and Immunogenetics (IF = 2.942). Of two other papers, where Igor Grekov is the first author, one has been accepted to The Journal of Microbiological Methods (IF = 2.018) and another has been submitted to PloS One (IF = 4.411). Each chapter of the results is structured as a brief introduction, aims of the research, summary of results and the contribution of the Ph.D. student, followed by the text of the corresponding article. The substantial contribution of Igor Grekov to the first author publications, including the design and the interpretation of the experiments should be noted.

The discussion summarizes the author's results and considers them in the context of current knowledge in the field.

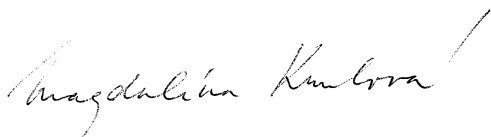
The conclusion clearly and concisely summarizes the main results of the research.

I would like to address the author the following questions:

- 1) Have *L. Major* promastigotes been grown in chemically defined media?
- 2) The manufacturer states that LDS-751 is a vital dye staining chromatin of the nucleus. The materials and methods include a chapter "Colocalization of laser dye styryl 751 (LDS-751) staining with *Leishmania* kinetoplast by light microscopy", however the results do not show the data obtained in these experiments. Could you provide the evidences for the mitochondrial localization of LDS-751?
- 3) What could be the base of the selective toxicity of calcimycin to *Leishmania* parasites?
- 4) Are you going to study in vivo treatment of experimental leishmaniasis with calcimycin?

Conclusion: Te thesis meets requirements for Ph.D. thesis and is strongly recommended as the basis for obtaining the degree Ph.D.

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