Skoltech
Skolkovo Institute of Science and Technology

Jury Member Report – Doctor of Philosophy thesis.

Name of Candidate: Aleksei Mikhalchenko

PhD Program: Life Sciences

Title of Thesis:

COMPARATIVE BIOLOGY OF AGING THROUGH THE LENS OF INDUCED PLURIPOTENT STEM CELLS

Supervisor: Prof. Philipp Khaitovich

Co-Supervisor: Prof. Vadim Gladyshev

Chair of PhD defense Jury: Prof. Olga Dontsova ***Email**: o.dontsova@skoltech.ru*

Date of Thesis Defense: 23 October 2018

Name of the Reviewer: Dario Riccardo Valenzano

I confirm the absence of any conflict of interest (Alternatively, Reviewer can formulate a possible conflict)	**Signature:**  **Date: 19-09-2018**

The purpose of this report is to obtain an independent review from the members of PhD defense Jury before the thesis defense. The members of PhD defense Jury are asked to submit signed copy of the report at least 30 days prior the thesis defense. The Reviewers are asked to bring a copy of the completed report to the thesis defense and to discuss the contents of each report with each other before the thesis defense.

If the reviewers have any queries about the thesis which they wish to raise in advance, please contact the Chair of the Jury.

Reviewer's Report

Reviewers report should contain the following items:

- Brief evaluation of the thesis quality and overall structure of the dissertation.
- The relevance of the topic of dissertation work to its actual content
- The relevance of the methods used in the dissertation
- The scientific significance of the results obtained and their compliance with the international level and current state of the art
- The relevance of the obtained results to applications (if applicable)
- The quality of publications

The summary of issues to be addressed before/during the thesis defense

The PhD Thesis submitted by Mr. Aleksei Mikhalchenko reports the research done on comparative biology of aging, focusing on tool development, biotechnological advancement and investigation of basic biological mechanisms in the exceptionally long-lived rodent naked mole rats (*Heterocephalus glaber*).

This thesis work includes three main results parts, namely i) the development of a successful protocol for iPSCs derivation and characterization by transcriptome analysis in naked mole rat and. Harnessing this methodology, Mr. Mikhalchenko could proceed to ii) the generation of early naked-mole rat/ mouse chimaeras, as well as rat-mouse chimaeras. Finally, this thesis work tackles iii) an in-depth study of comparative thermogenesis in naked-mole rats and mice.

This thesis is very well written, clear in its language, structure and overall exposition. The aims and methods are well presented and the results are in general solid and convincing. Mr. Mikhalchenko shows a very mature style in presenting his work and all the methods used are state-of-the-art.

Scientific significance and relevance of results:

The method development to derive naked-mole rat iPSC is different from standard methods used in mice iPSCs, making it pioneering. iPSC derivation in naked-mole rat will pave the way for studies aimed at mechanistically dissect the cellular and organ-level features that separate naked-mole rats from other mammals. This work includes important characterizations of specific features of naked-mole rat iPSCs, such as the tendency to become polyploid and the decreased tendency to generate tumors compared to mice iPSCs.

The Chimera generation between naked-mole rat and mice, as well as between rats and mice is again a bold, innovative and pioneering scientific enterprise, which promises to address several fundamental questions in developmental biology and in biology of aging. While the naked-mole rat / mouse chimera is still in its early stage of development, rat-mouse chimeras are at a more advanced stage and provide important insights into relative contribution of integrated vs host cell types during development and aging of the chimeras. The result shown are interpreted as conclusive evidences that the relative contribution of rat cells in adult chimeras becomes progressively smaller. This opens interesting scenarios of mouse vs. rat cell competition via selective immune elimination, senescence, apoptosis or other mechanisms.

The third and final scientific part studies the molecular basis of thermogenesis in naked-mole-rat. This organism is a great model to study the evolution of thermogenesis in mammals, as it is considered to be the only mammalian poikilotherm. This thesis work supports that no major changes in UCP1 function may be responsible for the limited capacity to maintain a constant core body temperature in this species. This chapter is very engaging and ranges from sequence comparison to cell-biology based assays of protein function, physiological test of energy consumption and surface temperature, whole-body temperature adaptation to experimental shifts and physiological responses to beta3-adrenergic agonist stimulation. Overall, this part supports that naked-mole rats have limited capacity to compensate external temperature compared to mice and seem to add significant new results to the current body of literature on naked-mole rat specific physiological traits.

Minor points of discussion: Some of the figures would have benefited from higher resolution and larger font.

The thesis work is a bit disconnected and the three parts could be independent projects. Especially the part related to thermogenesis feels a bit too disconnected from the two first parts. However, this does not take anything away from the merit of the work done.

The coat color changes in rat-mouse chimeras are interpreted as a change in relative rat-lineage contribution in older chimeras. However, an alternative and untested interpretation is that rat cells undergo stem cell depletion and or lose pigment granules. Loss of pigment granules – as opposed to loss of rat cells lineages – in aged chimeras would still not affect the relative contribution of rat to the chimera during aging. This could be easily tested by genotyping.

The claim that BAT is used up to compensate for lower temperatures (pg 123-125), measuring BAT mass before and after cold exposure for 24 hours, is not necessarily controlled by a similar measure in WAT mass. It could be that BAT consumption is not different from WAT usage and could be due to energy metabolism. Higher BAT than WAT consumption upon cold exposure would indeed support intact BAT function in naked-mole rat thermogenesis.

Quality of publications

The publications are of good quality and the body of work presented likely will result in additional important publications.

Provisional Recommendation

☒ *I recommend that the candidate should defend the thesis by means of a formal thesis defense*

☐ *I recommend that the candidate should defend the thesis by means of a formal thesis defense only after appropriate changes would be introduced in candidate's thesis according to the recommendations of the present report*

☐ *The thesis is not acceptable and I recommend that the candidate be exempt from the formal thesis defense*