# Skoltech
Skolkovo Institute of Science and Technology

## Jury Member Report – Doctor of Philosophy thesis.

**Name of Candidate:** Alexander Tyshkovskiy

**PhD Program:** Life Sciences

**Title of Thesis:** Molecular Signatures and Mechanisms behind Lifespan Extensions

**Supervisor:** Prof. Philipp Khaitovich

**Co-Supervisor:** Prof. Vadim Gladyshev

**Chair of PhD defense Jury:** Prof. Mikhail Gelfand          *Email*: *m.gelfand@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<br><br>(Alternatively, Reviewer can formulate a possible conflict) | Signature:<br><br>*[signature]*<br><br>**Date: 21-09-2018** |
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*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

<u>General summary</u>:

The Thesis work conducted by Mr. Alexander Tyshkovskiy is a multi-system investigation of molecular and genetic mechanisms underlying differences in lifespan across different rodents and among different lifespan-enhancing interventions. This work falls within the field of comparative biology of aging, providing novel important advancements that will be discussed below.

This work is composed of three result sections, the first focused on comparison of responses to gamma-irradiation in mouse and naked-mole rat cultured fibroblasts; the second focused on the analysis of changes in DNA methylation in blood cells during aging and caloric restriction; the third comparing gene expression profiles of liver tissue among several robust lifespan enhancing interventions.

The thesis is very well written, and I enjoyed reading its entirety, including the well-curated and informative introduction. The thesis' structure, language and figures are scientifically sound, clear and convincing. The discussion of the results is throughout succinct and balanced. The results are consistently of high quality and scientific relevance. Some of the presented results, especially the part relative to cross-species responses to fibroblast irradiation and the comparison of lifespan enhancing interventions will likely have broad reach, proposing new exciting avenues for future investigations of lifespan and aging-modulation strategies. Overall, the PhD candidate shows confidence and mature skills in bioinformatics and statistical methods to investigate expression data.

<u>Scientific significance and relevance of results</u>:

Gene expression comparison in mouse and naked-mole rat cultured fibroblasts shows very interesting species-specific responses to DNA damage. Naked mole rats are extremely interesting rodents, as they have a similar body mass than lab mice, but live ten times longer than mice, showing virtually no signs of aging. The combination of these unique features makes naked more rat a unique species to investigate the molecular and physiological mechanisms underlying lifespan regulation in mammals. According to Mr. Tyshkovskiy's results, naked-mole rats seem to be more protected from DNA damage, possibly improving DNA-repair machinery. Intriguingly, it is shown that NMR do induce senescence in response to high levels of irradiation, and levels of apoptosis are relatively low. This is very interesting and somehow counterintuitive, as several recent anti-aging interventions aim at eliminating senescent cells, due to their potentially cancerogenic secretory phenotype. This result will definitely provide ideal discussion material during the oral defense.

The analysis of the methylome in normal C57Bl/6 mice during aging and its comparison in the context of caloric restriction, allow to address which changes in DNA methylation are significantly associated with aging. Mr. Tyshkovskiy shows that methylation entropy increases with age, with genomic sites more methylated in early life becoming de-methylated during aging and vice-versa sites being hypo-methylated in young mice becoming more methylated during aging. Comparison of methylation changes during acute and chronic caloric restriction supports that indeed this intervention is able to also "rejuvenate" the methylome. It would be interesting to discuss – during the oral defense – what is the expected role of somatic mutations (e.g. following deamination of methylated cytosines) on the shift in methylation profile observed during aging. What is the amount of C to T transition expected during normal aging in a mouse? Would this bias the results of age-dependent shift in methylation profiles? If yes, how much?

The last part of the thesis does a superb job in comparing different lifespan enhancing interventions, focusing on the liver transcriptome. Most of the analyses are aimed at detecting common signatures across interventions. It would be also very important to know what unique signatures are typical of each

intervention and design an ideal "additive" intervention that would lead to further lifespan extension. In other words, which combination of interventions would lead to the maximal lifespan extension? Figure 20C is extremely interesting and is almost worth a publication in a of itself. The last part of this result section uses a mixed effect regression model to single out individual genes associated in the various interventions with median and maximum lifespan extension. This analysis is very interesting and confirms previous results, suggesting possible target for interventions, still within known aging pathways.

Quality of publications:

The publication record is solid, as Mr. Tyshkovskiy has obtained two important publications and is first author in a publication recently submitted to Cell Metabolism.

**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*