Institute of Animal Physiology and Genetics CAS

Rumburská 89, 277 21 Liběchov, Czech Republic

tel.: +420 315 639 532; fax: +420 315 639 510

E-mail: uzfg@iapg.cas.cz; URL http://www.iapg.cas.cz

# Report evaluation

**Dr. Josef Matousek Award**

Competition for Matoušek Award 2020

**Nomination number**

DRMA-2020-0001

**Nominee**

RNDr. Dávid Drutovič, Ph.D.

**Workplace**

81 / 810005 ( 810005)

**Name of Project**

The development and optimization of live-cell imaging of mouse zygotes and human

**Period**

1.3.2020 - 31.3.2021

**Short review**

The evaluated project was focused on the optimization of the method for the live cell visualization of mouse oocytes and human vitrified/thawed oocytes.using unique Viventis LS1 Live inverted light-sheet microscope system. The main scientific goal was the time lapse analysis of chromosomal segregation errors occurring during meiosis. The protocol was firstly tested on the mouse zygotes and preimplantation embryos employing the microinjection of H2B-mCherry mRNA and SiR-tubulin for the visualization of chromosomes and microtubules respectively. Two mRNA concentrations were tested (35 and 50 ng/ul) altogether with standard (20%) or low (5%) oxygen environments. To further improve the development up to blastocyst stage, the author compares the microinjection and microscopic observation from the one-cell and two-cell stage embryos. Surprisingly, better embryonic development was observed in the human rather than mouse cultivation media. On the other hand, not surprisingly, higher numbers of blastocysts were encountered using transgenic H2B-GFP mouse oocytes without need of the mRNA microinjection. The optimized protocol was then successfully tested on the human vitrified/thawed oocytes. Here the author encountered the problem with lower permeability of SiR-tubulin compared to mouse oocytes. The effective solution was the microinjection of microtubule staining Map4-Egfp cRNA and following incubation in lower concentrated SiR - tubulin dye. This improve the the first polar body extrusion of human oocytes up to 85%. The costs were spent in accordance with the project proposal.

**The results of the project are**

A - Excellent

**Final comment**

The evaluated project accomplished all aims. Author optimized the live-cell visualization protocol for the study of spindle formation and chromosomal segregation of mouse and human oocytes and preimplantation embryos employing unique Viventis LS1 Live inverted light-sheet microscope system.