Action number: CA16119

SHORT TERM SCIENTIFIC MISSION (STSM) – SUMMARY of SCIENTIFIC REPORTS

1. Grantee name: BART LEEMANS
   STSM title: "Development of an equine 3D oviduct model"
   STSM start and end date: 01/02/2018 to 05/04/2018

The conventional in vitro fertilization could not be induced using the equine oviduct explant model, which indicates that a more sophisticated model is required to resemble the in vivo fertilization supporting environment of the oviduct. The grantee hypothesized that an equine 3D culture system of oviduct epithelial cells can re-establish the in vivo-like differentiation status and function. As a main achievement, grantee in collaboration with host researchers, designed an equine 3D oviduct-on-a-chip system in a microfluidic chamber. As a final step, the survival of oviduct cells and ability to self-assemble into a ciliated secretory epithelium was assessed.

2. Grantee name: ANJA TOLIĆ
   STSM title: "Interplay of TETs and PARPs in DNA (de)methylation"
   STSM start and end date: 04/02/2018 to 04/04/2018

Major goal of proposed research was to investigate interplay between TET enzymes and PARP enzymes in vitro and in cellulo as well as to try to establish a 3D cell culture of wt and PARP-1 knock out (KO) mouse embryonic fibroblasts in order to evaluate levels of modified cytosine in cells grown in more physiologically natural environment, being mainly exposed to cell-to-cell and not cell-to plastic interactions. Kinetics assay was performed for all TET/PARP combinations but only TET1 PARyled with mPARP-1 showed conclusive results. The obtained results clearly point to inhibitory effect of PARylation on TET1 activity. In parallel with in vitro experiments wt and PARP-1 KO cells were cultured as spheroids by forming liquid marbles using polytetrafluoroethylene coating. DNA isolated from these cells as well as DNA from kinetics assay was prepared and all the samples will be sent for LC/MS analysis.

3. Grantee name: ALESSIO GIZZI
   STSM title: "Micromechanically motivated constitutive modelling of the visco-elastic active behavior of the cytoskeleton"
   STSM start and end date: 11/02/2018 to 25/02/2018

The aim of the STSM research stay was related to mathematical and computational modelling of visco-elastic active behavior of cell microstructures. Such a goal was conducted within the context of theoretical and computational mechanics, multiphysics mathematical modeling, nonlinear dynamics, computational biology and evolutionism. The main result of the STSM project is a theoretically formulated, mathematically and thermodynamically consistent continuum model of the active force generation in the cytoskeleton that accounts for the structural polymorphism of the cytoskeleton. Such a model will serve as robust and reliable basis for computational studies.

4. Grantee name: MARTINA ŠTAMPAR
   STSM title: “The application of 21-days old spheroids developed from HepG2/C3A cells for the assessment of genotoxic activity of xenobiotic compounds”
   STSM start and end date: 23/02/2018 to 30/03/2018

The aim of the proposed STSM was to use 3D HepG2/C3A cell model for the assessment of DNA damage induced by genotoxic compounds by applying the methodology that is commonly used on traditional 2D cell models in vitro and to optimize it for 3D models. Within the STSM the methodology for development of viable spheroids from hepatic HepG2/C3A cells in the rotating bioreactors was used. The 21-days old spheroids (dispersed in single cell format) were used for the assessment of genotoxic activity of xenobiotic substances via Comet assay. It was also determined if HepG2/C3A cells grown in 3D structure showed higher sensitivity towards genotoxic compounds compared to cells grown in 2D monolayes and could therefore represent more sensitive model for the detection of genotoxic compounds.

5. Grantee name: GABRIELLA PÁSTI
   STSM title: “Genome Engineering training”
   STSM start and end date: 02/03/2018 to 11/03/2018

This STSM was focused on CRISPR/Cas9 strategies and design. The purpose was to introduce participants with the state-of-the-art knowledge on genome engineering using precision DNA nucleases, with a special focus on the RNA-guided DNA endonuclease CRISPR/Cas9. The applicant also had a special interest in hiPSC-related CRISPR/Cas9 applications, knock in and fluorescent protein tagging. These topics have also been covered by the training. By the end of the training, the grantee gained an in-depth knowledge on genome engineering (mainly focusing on the CRISPR/Cas9 system and its various applications) and also a hands-on experience on CRISPR-related preparatory cloning steps.

6. Grantee name: MATTEO GHIRINGHELLI
   STSM title: “Organ perfusion and organ decellularization as novelty tools for 3-D in Vitro Study”
   STSM start and end date: 15/03/2018 to 18/04/2018

The aim of this study was to create a whole decellularized heart with a medium/long perfusion time and the low flow/pressure circuit using a pig heart collected from the local slaughterhouse. The main goal of the decellularization protocol is to create a well obtain ECM, in particular, from the Mitral, Tricuspid valves, left and right atrium. The protocol was a modification to a previous published report, including longer exposure to flow which provided more repeatable results. The main protocol’s characteristic allows the use of a heart from a slaughter pig avoiding a further use of an animal euthanized for decellularization purposes. This protocol is to be considered to obtain a well atria and major vessels decellularization instead of all heart decellularization.