"Biologie intégrative et physiologie" (BIP) Master Project – 2019 – IBDM, Kelly Team

Kelly Team: "Genetic control of Heart Development"

Project title: **Dissecting the mechanisms regulating cell fate choice in the cardiac lineage**
Supervisors: **Robert Kelly, PhD – Lucile Miquerol, PhD**
IBDM Host Lab: **Kelly Team**
email address: Robert.Kelly@univ-amu.fr - Lucile.Miquerol@univ-amu.fr

Concept and Objectives. Mouse heart development provides a biomedically relevant model for major developmental process including progenitor cell specification, patterning, differentiation and organogenesis. Formation of the mammalian heart involves successive waves of mesodermal progenitor cell differentiation and epithelial morphogenesis during heart tube formation and extension to give rise to different cardiac compartments and specialised cell types. Defects in this complex process underlie a spectrum of congenital heart defects in human patienrts. Moreover, an understanding of myocardial development is essential for regenerative strategies to heart repair.

Our group studies how successive cell fate choices are made during cardiogenesis, focusing on early progenitor cell decisions to give rise to myocardium versus other mesodermal fates, including skeletal muscles of the head, cardiomyocytes of the arterial and venous poles of the heart and specialised cardiomyocytes of the conduction system that orchestrate the heartbeat. Our technological approaches involve mouse genetics, embryo culture, lineage analysis and animal models of congenital heart defects including 22q11.2 deletion syndrome, Holt-Oram syndrome and ventricular non-compaction models. The project will address the roles of progenitor cell patterning and cell biology in the acquisition of divergent fate choices in the embryonic and fetal heart. The student will have the opportunity to learn a variety of techniques to study gene expression and function in the developing mouse embryo.

References

1) Francou A, De Bono C, Kelly RG. 2017. Epithelial tension in the second heart field promotes mouse heart tube elongation. **Nature Communications** 8:14770.
2) Diogo R, Kelly RG et al. 2015. A new heart for a new head in vertebrate cardiopharyngeal evolution. **Nature** 520:466-73
3) Rana MS et al. 2014. Tbx1 coordinates addition of posterior second heart field progenitor cells to the arterial and venous poles of the heart. **Circulation Research** 2014 Oct 10;115(9):790-9.
4) Miquerol L, Kelly RG. 2013. Organogenesis of the vertebrate heart. **Wiley Interdiscip Rev Dev Biol**. 2:17-29.

"Biologie intégrative et physiologie" (BIP) Master Project – 2019 – IBDM, Kelly Team

Pagès Team: "DNA damage and genome instability"
CRCM (Cancer Research Center of Marseille)

Project title: **Genetic of error-free and mutagenic lesion bypass.**
Supervisor: **Vincent Pagès, PhD**
CRCM Host Lab: **Pagès Team**
email address: vincent.pages@inserm.fr

Concept and Objectives:

The encounter of a replication fork with a blocking DNA lesion is a common event that cells need to address properly to preserve genome integrity. Cells possess two main DNA damage tolerance pathways: Translesion synthesis (TLS) and Damage Avoidance (DA) pathways. While TLS pathways are error-prone and are the major source of point mutations, DA pathways are error-free as they rely on mechanisms related to homologous recombination with the sister chromatid. When dealing with accidental DNA lesions, mutagenic DNA damage tolerance mechanisms can lead to unwanted mutations, the initiating cause of cancer. On the other hand, when DNA damaging agents are used as therapeutics during chemotherapy, error-free tolerance mechanisms can lead to resistance to treatments.

Our team "DNA damage and genome instability" aims at understanding how the balance between error-free (DA) and mutagenic (TLS) pathways is controlled *in vivo*. We have developed a unique tool that allow to follow the fate of a single lesion introduced in the chromosome of a living cell, using bacteria and yeast as model organisms.

The student will use our system to understand the genetics that regulates the use of both pathways (TLS and DA). He will inactivate candidate genes potentially involved in the regulation of Homologous Recombination and Translesion Synthesis. He will then assess the role of these genes by measuring in the strains he built the output in term of mutagenic and error-free lesion bypass. A wide variety of molecular biology techniques will be used (DNA cloning, PCR, qPCR, ChIP, NGS…).

References

Chrabaszcz, E., Laureti, L., Pagès, V. (2018) DNA lesions proximity modulates damage tolerance pathways in *Escherichia coli*. ***Nucleic Acids Res***. https://doi.org/10.1093/nar/gky135

Laureti, L., Lee, L., Philippin, G., & Pagès, V. (2017) A non-catalytic role of RecBCD in homology directed gap repair and translesion synthesis. ***Nucleic Acids Res***. 45: 5877–5886 http://doi.org/10.1093/nar/gkx217

Naiman, K., Pagès, V., and Fuchs, R. P. (2016) A defect in homologous recombination leads to increased translesion synthesis in E. coli. ***Nucleic Acids Res***. 44: 7691–7699 http://nar.oxfordjournals.org/content/early/2016/06/01/nar.gkw488.full

Pagès, V. (2016) Single-strand gap repair involves both RecF and RecBCD pathways. ***Curr. Genet.*** 62: 519–521 http://link.springer.com/article/10.1007/s00294-016-0575-5

Laureti, L., Demol, J., Fuchs, R. P., and Pagès, V. (2015) Bacterial Proliferation: Keep Dividing and Don't Mind the Gap. ***PLoS Genet*** 11: e1005757 http://journals.plos.org/plosgenetics/article?id=10.1371/journal.pgen.1005757

Naiman, K., Philippin, G., Fuchs, R.P., and Pagès, V. (2014). Chronology in lesion tolerance gives priority to genetic variability. **Proc Natl Acad Sci USA** *111*, 5526–5531. http://www.pnas.org/content/111/15/5526.full

NAME of the Team: "Structural Plasticity in the post-traumatic and Developing Brain"

Project title: **Prenatal anoxia, does it means developmental deficits ?**
Supervisors: **Pellegrino Christophe**
INMED Host Lab: **Rivera's Team**
email address: **christophe.pellegrino@inserm.fr**

Concept and Objectives.

Moderate to severe hypoxic encephalopathy of the term neonate and brain hypothermia.

The goal of this rotation is to set up a new method to develop and study pre-natal anoxia in the mouse brain. The outcome of term newborns with birth asphyxia and moderate to severe hypoxic ischemic encephalopathy remains very poor. After the primary phase of energy failure during asphyxia, neuronal cell metabolism may deteriorate in a secondary phase of brain injury. The window between these two phases opens the way to potential neuroprotective treatments such as brain cooling. Promising experimental data on controlled hypothermia need to be examined with clinical trials. In the group we propose that these hypoxic encephalopathies may contribute to first increase neuronal cell death then leading to changes in neuronal network wiring finally giving rise to behavioural changes.

The student will monitore and quantify neuronal death, both for principal cells and interneurons which are known to regulate early on the neuronal dynamic. In a second phase a behavioural study will quantify the deficits of the new born animals.

References

GABAergic Hub Neurons Orchestrate Synchrony in Developing Hippocampal Networks, P. Bonifazi et al. Science 326, 1419 (2009);

Pioneer GABA Cells Comprise a Subpopulation of Hub Neurons in the Developing Hippocampus Picardo M et al, Neuron 71, 695–709, August 25, 2011;

In vivo Calcium Imaging of Evoked Calcium Waves in the Embryonic Cortex, Yuryev M et al, Frontiers in cellular neuroscience, January 2016 | Volume 9 | Article 500;

GABA function may be related to the impairment of learning and memory caused by systemic prenatal hypoxia-ischemia, Marta Cristina Cunha-Rodrigues et al, Neurobiology of Learning and Memory, 2018

"Biologie intégrative et physiologie" (BIP) Master Project – 2019 – IBDM, Kelly Team

Angiogenesis, microenvironment and cancer

Project title: **Endothelial-mesenchymal transition in cancer**
Supervisor: **Roselyne Tournaire – Head of the Group: Angiogenesis, microenvironment and cancer**
CRCM Host Lab: **Iovanna Team – Pancreatic cancer - INSERM 1068 - Cancer Research Center of Marseille (CRCM). Luminy Science Park**
email address: roselyne.tournaire@inserm.fr ; rtournaire@hotmail.com

Concept and Objectives. Stromal cells provide a microenvironment that is critical for cancer cell growth, invasion, and metastatic progression. Fibroblasts are at the center of these changes. They are referred to as "activated" fibroblasts or myofibroblasts or CAFs (Cancer-Associated Fibroblasts) and are located near tumor cells. The interaction between myofibroblasts and cancer cells is essential for tumor growth and metastasis and results in poor clinical prognosis. The endothelial-mesenchymal transition (EndMT) is characterized by the loss of endothelial cell markers and the expression of mesenchymal cell markers and has been described during embryogenesis and during tissue fibrosis. We have previously shown, on the one hand, that the absence of the Tie1 tyrosine kinase receptor in endothelial cells induces EndMT and, on the other hand, that EndMT is present in human tumors and in particular pancreatic tumors (J. Garcia et al., EMBO Reports, 13, 2012). The EndMT can participate in the pool of fibroblasts associated with cancer.
Our project will address two approaches in parallel:

i) to study which are precisely the pathways (TGF-β, Notch, Wnt ...) and the endothelial receptors (Tie1, Tie2, VEGFR1, VEGFR2, NRP1 ...) involved in the induction of the EndMT

ii) to study the different EndMT-specific markers in immunohistochemistry and immunofluorescence (confocal microscopy) on human tumor sections and tumor models in the mouse (orthotopic grafts of pancreatic cancer cells, and mice either Pdx1-cre / Kras G12D, or / INK4a - / - or INK4a - / +).