"Biologie intégrative et physiologie" (BIP) Master Project – 2019 –

Team: "Signalling networks for stemness and tumorigenesis"

Project title: **Signalling cooperativity during tumour initiation and evolution: combining mouse genetics with -omics, biochemistry, and imaging.**

Supervisors: **Flavio Maina, PhD – Fabienne Lamballe, PhD**
IBDM Host Lab: **Maina Team**
email address: flavio.maina@univ-amu.fr - fabienne.lamballe@univ-amu.fr

Concept and Objectives. Large scale cancer (epi)genomic studies have significantly contributed to our understanding of how molecular alterations impact biological functions in cells, attributing tumorigenic properties, aggressiveness features, and resistance to therapies. However, several unmet needs limit our understanding of key biological principles associated with cancer, challenging the identification of triggers and impacting the discovery of effective therapeutic interventions.

Using mouse genetics, we demonstrated that specific tissues (liver and breast) are highly vulnerable to subtle increase of wild-type RTK levels, as illustrated by tumour initiation and evolution. Combining mouse genetics with -omics and integration of human cancer databases, the student will explore new functionally relevant combinatorial actions of genes at the origin of tumour initiation and responsible for sustained tumorigenicity. Studies will explore the cooperativity between signals during tumour initiation and evolution, as well as those ensuring a crosstalk of cancer cells with immune cells within the tumour microenvironment. The student will address these questions using either shRNA targeting plasmids or specific inhibitors. The molecular relevance will be analysed by RT-qPCRs and western blots. Functionality of these modulations will be assessed in cell culture using specific biological assays. Imaging will be applied to follow cell-cell interactions in vitro and tumour evolution/regression in vivo.

References

1) Arechederra M. et al. *Hypermethylation of gene body CpG islands predicts high dosage of functional oncogenes in liver cancer.* Revised version submitted to **Nature Communications**.

2) Fan Y. et al. *A Phosphokinome-based screen uncovers new drug synergies for cancer driven by liver-specific gain of non-oncogenic RTKs.* **Hepatology**, 66(5):1644-1661 (2017). PMID: 28586114

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"Biologie intégrative et physiologie" (BIP) Master Project – 2019 –

Team: "Signalling networks for stemness and tumorigenesis"

Project title: **Decipher signalling networks for human iPSC fate choices by acting on modulators of cell signalling perception**

Supervisors: **Rosanna Dono, PhD**
IBDM Host Lab: **Maina Team**
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Concept and Objectives. To date much attention is given to the potential clinical application of human induced pluripotent stem cells (hiPSCs). Their use for therapy is conditioned by strategies enabling an effective and efficient control of their self-renewal and differentiation properties. We have shown that down-regulation of the morphogen regulator Glypican4 (Gpc4) in hiPSCs poses them in a new biological state wherein hiPSCs are fully competent to propagate as undifferentiated cells, although become prone to undergo efficient lineage entry when exposed to differentiation signals. To enable an unbiased search for molecular and signalling changes underlying this unique biological state, we are combining global gene expression profiling of Gpc4-mutant and control hiPSCs at undifferentiated state with bioinformatics. This project will involve molecular and functional validation of selected candidates through biochemical and biological studies. Major methodologies include: 1) hiPSCs cell culture, expansion, and differentiation; 2) RT-qPCR; 3) immunostaining; 4) gain- and loss-of function studies.

References

1) Dono R. **Neural Regen Res.** 10:1576-7 (2015).

2) Fico et al. **The Journal of Neuroscience** 34(24):8318–8323 (2014).

3) Fico et al. **Stem Cells** 30(9):1863-74 (2012).

Team: "A developmental scaffold for the organization of cortical networks"

Project title: **The role of inhibitory neurons in the orchestration of hippocampal assemblies**
Supervisors: **Rosa Cossart, Marco Bocchio**
INMED Host Lab: **Cossart team**
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Concept and Objectives. The last decades of research have revealed that the brain computes information not only through the activity of single neurons, but also through complex patterns of recruitment of multiple neurons. In cortical areas, these patterns involve the synchronous activation of several excitatory cells that have been termed 'neuronal assemblies'. Current evidence suggests that assemblies are fundamental units of brain computation and cognition that might go awry in certain neuropsychiatric disorders. Despite the significance of assemblies, it remains unclear how these cells are orchestrated by inhibition. This project aims at unravelling this question using the mouse hippocampus as model. Hippocampal cell assemblies and inhibitory neurons will be recorded using *in vivo* 2-photon calcium imaging and electrophysiology in awake mice. Immunohistochemistry will be applied on perfusion-fixed brains to understand which populations of inhibitory cells were previously recorded. The student will team up with a post-doc to perform imaging and immunohistochemistry experiments. Additionally, the student will contribute to the analysis of imaging/electrophysiology data.

References

1) Malvache, A., Reichinnek, S., Villette, V., Haimerl, C., and Cossart, R. (2016). Awake hippocampal reactivations project onto orthogonal neuronal assemblies. **Science** (80-). *353*, 1280–1283.

2) Villette, V., Malvache, A., Tressard, T., Dupuy, N., and Cossart, R. (2015). Internally Recurring Hippocampal Sequences as a Population Template of Spatiotemporal Information. **Neuron** *88*, 357–366.

3) *Reviews about neuronal assemblies*

Holtmaat, A., and Caroni, P. (2016). Functional and structural underpinnings of neuronal assembly formation in learning. **Nat. Neurosci**. *19*, 1553–1562.

Yuste, R. (2015). From the neuron doctrine to neural networks. **Nat. Rev. Neurosci.** 16, 487–497.

"Biologie intégrative et physiologie" (BIP) Master Project – 2019 –

Team: "Stem cells and Brain repair"

Project title: **Unraveling the molecular and cellular bases of Myelin regeneration**
Supervisors: **Pascale Durbec**
IBDM Host Lab: **Durbec team**
email address: pascale.durbec@univ-amu.fr

Concept and Objectives. In some patients affected by multiple sclerosis, spontaneous remyelination can occur. Although insufficient to counter the damages caused by the repetitive attacks, this spontaneous regenerative process represents great therapeutic hopes. Our objectives are to uncover the cellular and molecular mechanisms controlling this process in order to increase our fundamental knowledge on brain regeneration and to promote myelin repair (For review see El Waly et al. 2014). Using mouse models of demyelination and genetic tracing, we have shown that adult neural stem cells from the sub-ventricular zone (SVZ) can efficiently participate to this repair process (Brousse et al. 2015) and that forced reprograming of SVZ derived neuronal progenitors into oligodendrocytes can promote myelin formation after lesion (El Waly et al. 2018). More recently, we have compared the gene expression profile of SVZ cells in control and demyelinated conditions using Single cell RNA sequencing. We aim to use this set of data to understand 1) if SVZ derived neural progenitors spontaneously recruited to injured areas can exert local immunomodulatory effects and 2) how SVZ derived neuronal progenitors can trans-differentiate to generate oligodendrocytes in vivo after lesion.

References

1) El Waly B, Cayre M, Durbec P. Promoting Myelin Repair through In Vivo Neuroblast Reprogramming. **Stem Cell Reports**. 2018 Mar 27. S2213-6711(18)30106-1. In press.

2) Brousse B., Magalon K, Durbec P and M Cayre (2015) Region and dynamic specificities of adult neural stem cells and oligodendrocyte precursors in myelin regeneration in the mouse brain. **Biol Open.** 2015 Jul 3;4(8):980-92.

3) El Waly B, Macchi M, Cayre M, Durbec P. Oligodendrogenesis in the normal and pathological central nervous system. **Front Neurosci**. 2014 Jun 12;8:145. Review.

Team "Host-pathogen interaction in the Drosophila model"

Project title: **Studying the Gut-Brain axis in the Drosophila model**
Supervisors: **Bernard Charroux, Leo Kurz, Olivier Zugasti**
IBDM Host Lab: **Julien Royet**
email address: Julien.royet@univ-amu.fr

Concept and Objectives. A universal feature of organisms with open digestive tracts is colonization of the gastrointestinal tract by a characteristic commensal microbiota. This microbiota, which thrives on the nutrients produced by host's diet, is shaped by host-specific selective pressures such as the intestinal environment and food preference. In turn, the microbiota manipulates host metabolism by generating essential nutrients and excreting metabolites that serve as a form of interspecies communication. Intestinal bacteria are therefore implicated in setting basal metabolic tone, educating the immune system or even in impacting central functions, such as appetite control and mood. Studies in mice and humans have allowed considerable progress in defining how intestinal bacteria influence human physiology and metabolism. However, the mechanisms underlying microbiota-host communication are extremely complex and remain largely to be elucidated. Because of its less-complex and more-tractable microbiota in comparison to mammals, *Drosophila* provides a useful model in which to study the governing principles of the host's metabolic interaction with its microbiota. Our previous results have shown that the universal bacterial cell wall component, peptidoglycan, is one of the key mediator of the dialog between *Drosophila* and its gut microbiota. Sensing of microbiota-derived PGN by pattern recognition receptor belonging to the PGRP (peptidoglycan recognition receptors) family activates the NF-kB pathway locally in the enterocytes. Our latest genetic data demonstrate that the gut-derived PGN is able to reach and to signal to the central nervous system demonstrating that PGN is a major regulator of the Gut-Brain communication in eukaryotes.

We propose here a rotation for a student that will be interested to use the power of *Drosophila* genetics and latest imaging techniques to understand how gut derived metabolites dialogs with the central nervous system of the host.

References

1) Kurz CL, Charroux B, Chaduli D, Viallat-Lieutaud A, Royet J. (**2017**) Peptidoglycan sensing by octopaminergic neurons modulates Drosophila oviposition. ***Elife.*** March 7;6. pii: e2193

2) Charroux B, Capo F, Kurz CL, Peslier S, Chaduli D, Viallat-lieutaud A, Royet J (**2018**). Cytosolic and secreted peptidoglycan-degradingenzymes in *Drosophila* respectively control local and systemic immune responses to microbiota. ***Cell Host and Microbe***. Cell Host Microbe. 2018 Jan 30. pii: S1931-3128(17)30543-7

3) Masuzzo A, Royet J .(2018) Lipid Catabolism Fuels Drosophila Gut Immunity. **Cell Host Microbe.** 2018 Mar 14;23(3):288-290

"Biologie intégrative et physiologie" (BIP) Master Project – 2019 –

Team : "Polarization and binary cell fate decisions in the nervous system

Project title: **Live imaging of transcription**
Supervisors: **Antoine Barrière**
IBDM Host Lab: **Vincent Bertrand**
email address: antoine.barrière@univ-amu.fr

Concept and Objectives. Our team is interested in the dynamics of transcription, as it pertains to cell fate determination and maintenance in *C. elegans*. To that end, we developped a method to image transcription in whole *C. elegans* as well as in cultured cells, allowing us to view and follow active transcription sites.

The project will require quantitative live microscopy, smFISH imaging, standard microbiology and CRISPR-Cas9-based transgenesis. The student will optimize the methods to image transcription, measure transcription of multiple genes in several tissues, in particular neurons, and modify small RNA pathways.

References

1) Bordet G, Bertrand V. Zic Genes in Nematodes: A Role in Nervous System Development and Wnt Signaling. **Adv Exp Med Biol**. 2018; 1046:59-68.

2) Tiveron MC, Beclin C, Murgan S, Wild S, Angelova A, Marc J, Coré N, de Chevigny A, Herrera E, Bosio A, Bertrand V, Cremer H. Zic-Proteins Are Repressors of Dopaminergic Forebrain Fate in Mice and C. elegans. **J Neurosci**. 2017 Nov 1;37(44):10611-10623

"Biologie intégrative et physiologie" (BIP) Master Project – 2019 –

Team : "Biology of Ciliated Epithelia"

Project title: **Mechanisms of multiciliated cell construction**
Supervisors: **Laurent Kodjabachian**
IBDM Host Lab: **Biology of Ciliated Epithelia**
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Concept and Objectives. Vertebrate animals possess specialized epithelia containing multiciliated cells (MCCs), whose hundreds of motile cilia beat coordinately to generate directional fluid flow [1]. In humans, MCCs help airway cleansing, ovum implantation and cerebrospinal fluid circulation. Understanding the biology of MCCs is a fundamental issue with high biomedical relevance, as their dysfunction can cause severe respiratory syndromes and infertility [2, 3]. Our team studies MCCs in three experimental paradigms: the multiciliated skin of the amphibian *Xenopus laevis* embryo [4-6]; the mouse post-natal brain [7]; a newly developed model of *in vitro* MCC culture (unpublished). One of the mystery in this field of research is how the right number of cilia per MCC is produced. As each cilium rests on a single centriole, the question becomes how centriole multiplication is operated and controlled. Our team currently develops an integrated cell and developmental biology program to address this question.

The selected candidate may receive training in some of the following areas: molecular biology, biochemistry, *Xenopus* micro-injection, mouse brain electroporation, cell culture, fluorescent confocal and super-resolution imaging. Command of english is compulsory. The selected candidate must have received a solid background in one or more of the following subjects: cell biology, developmental biology, molecular biology, biochemistry.

References

1) Brooks, E.R. and J.B. Wallingford, Multiciliated cells. Curr Biol, 2014. 24(19): p. R973-82.
2) Boon, M., et al., MCIDAS mutations result in a mucociliary clearance disorder with reduced generation of multiple motile cilia. Nat Commun, 2014. 5: p. 4418.
3) Wallmeier, J., et al., Mutations in CCNO result in congenital mucociliary clearance disorder with reduced generation of multiple motile cilia. Nat Genet, 2014. 46(6): p. 646-51.
4) Cibois, M., et al., BMP signalling controls the construction of vertebrate mucociliary epithelia. Development, 2015. 142(13): p. 2352-63.
5) Marcet, B., et al., Control of vertebrate multiciliogenesis by miR-449 through direct repression of the Delta/Notch pathway. Nat Cell Biol, 2011. 13(6): p. 693-9.
6) Chevalier, B., et al., miR-34/449 control apical actin network formation during multiciliogenesis through small GTPase pathways. Nat Commun, 2015. 6: p. 8386.
7) Boutin, C., et al., A dual role for planar cell polarity genes in ciliated cells. Proc Natl Acad Sci U S A, 2014. 111(30): p. E3129-38.

"Biologie intégrative et physiologie" (BIP) Master Project – 2019 –

Team: "Developmental plasticity of GABAergic synapses"

Project title: **Emergence of functional cortical GABAergic inhibition in health and disease.**
Supervisors: **Jean-Luc Gaiarsa, PhD – Igor Medina, PhD – Christophe Porcher, PhD.**
INMED Host Lab: **Gaiarsa Team**
email address: jean-luc.gaiarsa@inserm.fr

Concept and Objectives. Most brain computations rely on a proper balance between excitation and inhibition, which progressively emerges during development. The γ-aminobutyric acid (GABA) is the main inhibitory transmitter in the adult brain. However, in rodents, at fetal and postnatal stages, GABA induces a membrane depolarization due to elevated intracellular chloride concentration ($[Cl^-]_i$). During the second postnatal week of life, the functional expression of the chloride extruder, K^+-Cl^- type 2 co-transporter (KCC2), causes a decrease of $[Cl^-]_i$ and consequently shifts the chloride-dependent GABAergic responses towards a more hyperpolarized value (*1, 2*). This developmental sequence is likely shifted toward fetal life in humans.

Rodent studies showed that defective chloride homeostasis contributes to the pathogenesis of an array of neurological disorders including autism spectrum disorders (ASD), Rett syndrome, Down syndrome and Huntington's disease. Conversely, pharmacological manipulations aimed at restoring low $[Cl^-]_i$ improve neurological symptoms in rodents and humans (*3*). Therefore, unveiling mechanisms controlling the schedule of the GABA developmental sequence is decisive to identify molecular targets to correct abnormal developmental trajectories.

The present project aims at (1) identifying the key cellular mechanisms and players controlling the emergence of functional GABAergic inhibition in health and (2) question whether and how altered expression or activity of the identified players contributes to pathogenesis of neurological disorders (*4-6*). To address these questions we combine different technical approaches (molecular and cellular biology, live fluorescence imaging, morphology, electrophysiology and behavior) and experimental models (neuronal cultures, acute slices, *in vivo*).

References

1) Ben Ari Y. *et al.* GABA: a pioneer transmitter that excites immature neurons and generates primitive oscillations. ***Physiol Rev.*** 87, 1215 (2007). PMID: 17928584

2) Medina I. *et al*. Current view on the functional regulation of the neuronal K(+)-Cl(-) cotransporter KCC2. ***Front Cell Neurosci.*** 8, 27 (2014). PMID: 24567703

3) Lemonnier *E. et al*. Effects of bumetanide on neurobehavioral function in children and adolescents with autism spectrum disorders. ***Transl. Psychiatry*** 7, (2017). PMID: 28485727

4) Gaiarsa J.L. *et al*. Contribution of metabotropic GABA(B) receptors to neuronal network construction. ***Pharmacology & Therapeutics*** 132, 170 (2011). PMID: 21718720

5) Riffault *B. et al.* Pro-Brain-Derived Neurotrophic Factor (proBDNF)-Mediated p75NTR Activation Promotes Depolarizing Actions of GABA and Increases Susceptibility to Epileptic Seizures. ***Cerebral Cortex*** 28, 510 (2018) PMID: 27913431

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"Biologie intégrative et physiologie" (BIP) Master Project – 2019 –

Team "Physical and Molecular Principles Governing Cytoskeletal Organization"

Project title: **Impact of Disease-Causing Actin Mutations on the Organization of the Cytoskeleton.**

Supervisors: **Alphée Michelot, PhD**

IBDM Host Lab: **Michelot Team**

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Concept and Objectives. Actin filaments are key components of the cytoskeleton, and their assembly into various complex tridimensional networks serves as an **internal architecture for eukaryotic cells**. The fast dynamics of these polymeric networks enable cells to exert forces in order to change their shape, move, and divide themselves. The different properties of actin filament networks are constantly **remodeled by dozens of families of actin binding proteins (ABPs)** which are interacting with the actin filaments, and which are responsible for the nucleation, crosslinking, rearrangement and/or disassembly of actin networks. Importantly, **these families of proteins are usually highly conserved**, which enables us to question their activities and functions in various organisms ranging from yeast to higher eukaryotes.

Many examples in the literature report how tightly regulated actin network properties must be to perform cellular functions optimally. A slight imbalance in this complex equilibrium can lead to severe cellular defects, and **several point mutations on the human actin genes are directly linked with a number of diseases** [1]. However, our community lacks experimental systems enabling us to understand globally how each of these mutations affect interactions between the actin cytoskeleton and families of ABPs**.**

We have recently developed in the lab a system for rapid actin gene replacement in yeast. **The objective of this specific project** is to use this system to study systematically how deleterious mutations of the actin gene affects the organization of the actin cytoskeleton in this organism, and how the recruitment of various ABPs is affected. These results will be essential to hypothesize which important interactions may be affected in mutant human cells, and these hypotheses will be verified with various techniques ranging from biochemistry to cell biology.

Candidates should be creative while being highly rigorous. The capacity to provide a comprehensive and synthetic description of ongoing work will be required. The host team is international, so speaking French is not mandatory.

References

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2) MICHELOT A, DRUBIN DG. Building Distinct Actin Filament Networks in a Common Cytoplasm. ***Curr Biol***. 2011 Jul 26;21(14):R560-9

"Biologie intégrative et physiologie" (BIP) Master Project – 2019 –

Team "Mechanisms of Gene Regulation by Transcription Factors"

Project title: **Uncovering principles and molecular modalities of genome regulation**
Supervisors: **Y Graba, A Saurin**
IBDM Host Lab: **Graba/Saurin**
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Concept and Objectives. Genome regulation is central to development, evolution and disease. Our team aims at uncovering and understanding fundamental rules of genome regulation by studying the molecular modalities of Hox proteins, representatives of one of the largest class of transcription factors [1]. Our approach combines genomics and proteomics to set the molecular landscape, structural biology and molecular modeling to grasp the atomic details of the mechanisms, and classical molecular genetics and imaging to probe in vivo function. Our recent work uncovered plasticity in protein interaction as a key feature of Hox proteins [2, 3], and established a direct link towards proteins of the basal transcription machinery and epigenetic regulatory complexes [4, 5]. These two findings are central to our current research topics, and define the context of the M2 internship proposed. The precise focus of the work, and accordingly the supervision will depend upon the exact project defined in discussion with the M2 student.

References

1) Rezsohazy, R., et al. (2015) Cellular and molecular insights into Hox protein action. Development 142, 1212-1227
2) Foos, N., Maurel-Zaffran, C., Jesús Maté, M., Vincentelli, R., Hainaut, M., Berenger, H., Pradel, J., Saurin, A. J., Ortiz-Lombardía, M., and Graba, Y. (2015) A flexible extension of the Drosophila Ultrabithorax homeodomain defines a novel Hox/PBC interaction mode Structure 23, 270–279
3) Ortiz-Lombardia, M., et al. (2017) Hox functional diversity: Novel insights from flexible motif folding and plastic protein interaction. BioEssays 39
4) Boube, M., et al. (2014) Drosophila melanogaster Hox Transcription Factors Access the RNA Polymerase II Machinery through Direct Homeodomain Binding to a Conserved Motif of Mediator Subunit Med19. PLoS genetics 10, e1004303
5) Zouaz, A., et al. (2017) The Hox proteins Ubx and AbdA collaborate with the transcription pausing factor M1BP to regulate gene transcription. EMBO J 36, 2887-2906

"Biologie intégrative et physiologie" (BIP) Master Project – 2019 –

Cremer Team: "Molecular Control of Neurogenesis""

Project title: **Identification of transcriptional control mechanisms underlying neuronal phenotype and integration**

Supervisors: **Nathalie Core**
IBDM Host Lab: **Cremer Team**
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Concept and Objectives. In the subventricular zone (SVZ) of the forebrain lateral ventricles (LV) in the mammalian brain pre-determined neuronal stem cells generate large amounts of neuronal progenitors that migrate into the olfactory bulb (OB). Here they differentiate into interneurons that use mainly GABA, dopamine and also glutamate as their neurotransmitters. This ability of the postnatal brain to generate and integrate new neurons raised hope for the use of adult neurogenesis in cell therapeutic approaches to neurodegenerative diseases.
Over the past years we identified several factors regulating the determination of neurons towards specific neurotransmitter fates. For example, we demonstrated that a molecular interaction between Pax6 mRNA and the microRNA mir-7a, that fine tunes Pax6 protein concentration, is crucial for the control of dopaminergic neurotransmitter phenotype in the olfactory bulb (de Chevigny A.et al., Nature Neuroscience, 2012). More recently we showed that Zic-transcription factors induce GABAergic fate by suppressing Pax6 (Tiveron et al., J. Neurosci 2017).

In this M2 project we will focus on additional factors regulating defined steps in the neurogenic sequence. In particular we will study the role of the transcriptional regulator Vax1 in determination and neuronal integration. At the experimental level the project is based on the use of Vax1 deficient mice in combination with in brain electroporation. The applicant will use state of the art imaging technology, including confocal, light sheet and 2-photon in vivo imaging.

References

1) Tiveron MC, Beclin C, Murgan S, Wild S, Angelova A, Marc J, Coré N, de Chevigny A, Herrera E, Bosio A, Bertrand V and Cremer H (2017) Zic-proteins are repressors of dopaminergic forebrain fate in mice and C. elegans, **J. Neuroscience** 29, 3888-16; DOI: https://doi.org/10.1523/JNEUROSCI.3888-16.2017
2) Bugeon,S., de Chevigny, A., Boutin, C., Coré, N., Wild, S., Bosio, A. Cremer, H. (co-senior, corresponding author), Beclin, C. (2017) Direct and efficient transfection of mouse neural stem cells and mature neurons by in vivo mRNA electroporation, **Development,** 144(21):3968-3977. doi: 10.1242/dev.151381
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5) de Chevigny A, Coré N (co-first authors), Follert P, Gaudin N, Barbry P, Béclin C and **Cremer H** (2012) miR-7a regulation of Pax6 in neural stem cells controls the spatial origin of forebrain dopaminergic neurons, **Nature Neuroscience** 15(8):1120-6,doi:10.1038/nn.3142

Prud'homme Team: "Evolution and development of morphology and behavior"

Project title: Functional characterization of gustatory receptor neurons in *Drosophila* oviposition behavior

Supervisors: **Benjamin Prud'homme, PhD – Matthieu Cavey, PhD**
IBDM Host Lab: **Prud'homme Team**
email address: **benjamin.prudhomme@univ-amu.fr**

Concept and Objectives. Phenotypic evolution proceeds via changes in gene expression and function. These changes modify molecular and cellular processes, producing novel morphological and behavioral traits. In the past decades, the field of evolutionary-development (evo-devo) has focused primarily on the mechanisms underlying morphological evolution (1) but much less is known on behavioral evolution (2). How are neuronal circuits reconfigured to modify behavior during evolution?

To address this question, we use a comparative approach with several *Drosophila* species and classical neurobiology experiments to manipulate their nervous systems. Specifically, we use a simple behavior which has diverged among different *Drosophila* species over a few million years: fruit flies use sensory information to choose where to lay their eggs (oviposition behavior). While most species - including *Drosophila melanogaster* - prefer to lay eggs on rotten fruits, the invasive pest *Drosophila suzukii* prefers to lay on fresh/ripe fruits (this actually poses a major threat to the fruit industry). Our goal is to understand the cellular, molecular and genetic changes in neuronal circuits that have led to the evolution of this behavior.

We have previously demonstrated that *D. suzukii*'s divergent oviposition behavior is linked to changes in its responses to olfactory and gustatory stimuli present in fruits (3). We are currently characterizing the sensory neurons responding to these stimuli to understand their contribution to oviposition behavior evolution. This project will focus on gustatory neurons responding to sugars (i.e. neurons expressing the gustatory receptor Gr64f), which appear to play a major role in driving *D. suzukii*'s preference for fresh (i.e. sweet) fruits. The student will use genetic tools to express specific transgenes in these gustatory neurons to study their function. Several transgenes will be used to manipulate the excitability of these neurons (e.g: UAS-Kir2.1 or UAS-TNT to silence them and UAS-dTrpA1 or UAS-NaChBac to activate/hypersensitize them) and determine how this affects oviposition choices in well-controlled behavioral assays. In addition, the student will use the calcium indicator GCaMP6 and live imaging techniques to characterize the electrical responses of these neurons to specific sensory stimuli and compare these neuronal responses across different *Drosophila* species. These experiments aim to identify critical functional differences in the nervous system of these different species. This project will involve genetic experiments (crosses), behavioral experiments, dissection/tissue preparation, confocal microscopy, image processing/analysis, quantifications and statistics.

References

1) Prud'homme et al. 2007. **Proc Natl Acad Sci U S A.** May 15;104

2) Cande et al. 2012. **Curr Opin Neurobiol.** Feb;23(1):152-8

3) Karageorgi et al. 2017. **Current Biology** Mar 20;27(6):847-853

"Biologie intégrative et physiologie" (BIP) Master Project – 2019 –

Prud'homme Team: "Evolution & Development of morphology and behaviour"

Project title: **How evolution of gene regulatory networks drives morphological pattern diversification ?**

Supervisors: **B. Prud'homme, PhD**
IBDM Host Lab: **Prud'homme Team**
email address: benjamin.prudhomme@univ-amu.fr

Concept and Objectives. The formation of animal morphological patterns is orchestrated by gene regulatory networks that operate during embryonic development. In turn, the evolution of morphological patterns between species must result from changes in the activity or the architecture of the underlying gene regulatory networks. Yet, how these networks are assembled during evolution and how functional changes in these networks drive morphological evolution is poorly understood.

We are addressing these general questions by studying the formation and evolution of wing pigmentation patterns in various Drosophila species. We focus in particular on the activity and evolution of the enhancers that connect the genetic components of the network and that are favoured evolutionary targets to modulate the activity of the network and, in turn, the morphological output. We use a vast repertoire of approaches, ranging from comparative functional genomics (RNAseq, atacseq, Chipseq), genetics, imaging, transgenesis and genome editing in various species to characterize the architecture and the activity of the gene network encoding wing pigmentation patterns. The student will be using a combination of these approaches to characterize enhancers of interest.

References

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Team "Neuronal coding and plasticity in epilepsy"

Project title: **Neuronal coding and plasticity in naïve and epileptic neuronal circuits**
Supervisors: **Valérie Crépel**
INMED Host Lab: **Neuronal coding and plasticity in epilepsy (http://www.inmed.fr/integrative-properties-of-plastic-neuronal-circuits-in-health-and-disease-fr)**
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Concept and Objectives. Our team is interested in the coding of information in the hippocampus, a structure involved in memory. We focus our works on the dentate gyrus, sitting between the entorhinal cortex and area CA3. This region is both anatomically well positioned and physiologically predisposed to play the role of a gate, blocking or filtering excitatory activity from the entorhinal cortex. We investigate the neuronal computation and plasticity of dentate granule cells in normal and pathological conditions. Our studies are conducted at multiscale levels i.e. from the individual spine to the microcircuit.

References

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Name of the Team: "Early life imprinting and neurodevelopmental disorders"

Project title: "Oxytocin in pathogenesis and for treatment of a mouse model of Prader-Willi syndrome and autism"

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Concept and Objectives. The *MAGEL2* gene is a gene involved in feeding and behavioral alterations observed in Prader-Willi and Schaaf-Yang syndromes (autistic syndromes). We showed that *Magel2*-deficient mice display a disturbance in early feeding and later on in adulthood showing alterations in social behavior and learning abilities. Importantly, an administration of oxytocin (a brain neuromodulator) in *Magel2*-deficient pups, in a critical period of post-natal development, restores a normal sucking activity and normalizes the social behavior and learning abilities in adulthood. However, we do not know how an administration of exogenous oxytocin acts during brain development to allow a long term effect in *Magel2-KO* mice. As we have shown recently, our results opened the door to a powerful pharmacological therapy for the PWS and might be considered for autism spectrum disorders. Now we are deciphering the key role of the oxytocinergic system in early postnatal development to shape the feeding behavior, social behavior and cognition in physiological and pathophysiological conditions using mouse models. We are doing *in vivo* and *ex-vivo* studies using a large panel of techniques allowing behavioral, physiological cellular and molecular studies. Several projects are proposed.

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Keywords: neurodevelopment, autism, genetics, epigenetics.