Internship Proposal

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Project Title:
Making tissues robust by self-recovery of epithelial organisation
Level:

Master Student

Project Summary:

Apical-basal organisation confers direction to the specialized functions of epithelial tissues (e.g. absorption and secretion) and ensures their ability to compartmentalize all animal organs. Work from a variety of groups, including our own (Morais-de-Sa et al., 2010; Moreira et al., 2019; Ventura et al., 2020), has established that epithelial cells use a conserved set of polarity factors to define apical and basolateral domains, while positioning intercellular junctions. Loss of apical-basal organization underlies debilitating disorders, including diseases of the gastrointestinal tract and cancer (Halaoui & McCaffrey, 2015; Schneeberger et al., 2018). Importantly, transient exposure of epithelial tissues to external challenges, such as smoke and bacterial infection, disrupts apical-basal organization and can drive precancerous lesions. However, how and if differentiated epithelial cells have intrinsic potential to restore apical-basal organization remains unknown. We developed unique optogenetic approaches that enable high-temporally controlled disruption a central regulator of apical-basal polarity, atypical Protein kinase C (aPKC) (Osswald et al., 2022). Taking advantage of the reversibility of optogenetic control, we obtained evidence for the ability of epithelial tissues to self-recover apical-basal organisation, and are now characterising this unexplored property of epithelial cells to uncover pathways that maintain epithelial function and prevent cancer development.

Work to be developed by the student:

The student will use Drosophila genetics and range of microscopy techniques (including spinning disk confocal microscopy and super-resolution approaches (STED and SIM) and atomic force microscopy in Drosophila ovaries cultured ex-vivo), optogenetic perturbation approches and molecular biology, contributing to the following taks:

GOAL 1) Characterize the dynamic recovery of apical-basal organization after transient polarity disruption

Combining optogenetic approaches with quantitative imaging in Drosophila tissues, the student will contribute to comprehensively characterization of the dynamic recovery of functional apical-basal organization. To this end, the student will cross-correlate the redistribution of markers of different subcellular domains and of the actomyosin network with unprecedent detail using live imaging approaches and super-resolution microscopy (STED (stimulated emission depletion microscopy), SIM (Structured illumination microscopy).

GOAL 2) Define molecular cues provided by the extracellular matrix that promote the recovery of epithelial polarity

The extracellular matrix (ECM) may provide a biomechanical cue for polarization. To investigate this, the student will use a) Atomic Force Microscopy to measure ECM stiffness after different periods of polarity disruption/recovery, and b) tissue-specific RNAi to pinpont components of the cell-ECM interface that contribute to polarity recovery.

References:

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Schneeberger, K., Roth, S., Nieuwenhuis, E. E. S., & Middendorp, S. (2018). Intestinal epithelial cell polarity defects in disease: lessons from microvillus inclusion disease. Dis Model Mech, 11(2). https://doi.org/10.1242/dmm.031088

Ventura, G., Moreira, S., Barros-Carvalho, A., Osswald, M., & Morais-de-Sa, E. (2020). Lgl cortical dynamics are independent of binding to the Scrib-Dlg complex but require Dlg-dependent restriction of aPKC. Development, 147(15). https://doi.org/10.1242/dev.186593

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