Multi-scale imaging of epithelial tissues

Philippe P. Girard1,2, Olivier Renaud3, Olivier Leroy3, Maïté Coppey1 and Yohanns Bellaïche4

1: Institut Jacques Monod, Mechanotransduction: from Cell Surface to Nucleus, UMR 7592, CNRS, Paris Diderot University, Sorbonne Paris Cité, F-75013 Paris, France.
2: Biomedical and fundamental Science Faculty, Paris Descartes University, Sorbonne Paris Cité, F-75006 Paris, France.
3: Plateforme Imagerie Cellulaire et Tissulaire-Infrastructure en Biologie Santé et Agronomie, UMR3215 CNRS/U934 INSERM, Institut Curie, PSL Research University, F-75005 Paris, France.
4: Genetics and Developmental Biology, Team ‘Polarity, division and morphogenesis’, Institut Curie, UMR3215 CNRS, U934 Inserm, 26 rue d’Ulm, F-75248 Paris Cedex 05, France.

Author e-mail address: philippe.girard@ijm.fr

The mechanisms by which cells acquire positional identities as in a coordinate system have been well documented in the literature. However, our understanding of this phenomenon in relation to tissue shape and function remains less explored. For example, central problems are how positional information is set up, how it is recorded and then how it is integrated to regulate proliferation, planar polarization, and morphogenesis. A number of models have been proposed for the setting up of positional gradients but additional quantitative measurements would be needed. One of the reason is that the study of cell and tissue morphogenesis requires the intensive use of multi-scale optical imaging method since it requires both to have a large field of view (of the whole tissue) and sub-cellular resolution (e.g. membranes, myosin filaments, etc.). Confocal microscopy provides such a resolution, however, the field of view remains restricted to a limited acquisition area, and especially in the axial direction, and the acquisition speed is too low to reliably detect all the cell movement.

To overcome these limitations, we have developed a new methodology for imaging the 3D morphogenesis of mono-layered epithelial tissues from the cell to the tissue-scale with high spatiotemporal resolution and along different directions. This has been achieved in the context of the epithelium of Drosophila pupa which was fixed at the end of a metal rod mounted on a 4D motor system (3 motorized linear and 1 rotational stages) similar to a light sheet microscopy setup. Using confocal spinning disk microscopy with one dry objective, this setup has allowed us to image multi-view stacks of a large region of the mono-layered epithelium of the pupa at high speed and over time. This system will give us the opportunity to link gene expression, cytoskeleton organization, and global tissue scale movements to advance the understanding of the link between pattern formation and tissue dynamics in Drosophila. We plan to also present result where the sample will be imaged by spinning disk microscopy using two dry lenses in a symmetric configuration. The system that we developed should find several applications in the context of epithelial tissue dynamics and multiscale imaging.