

## **Webinar**

**Buffered EGFR signalling regulated by Spitz to Argos expression ratio is critical for patterning the Drosophila eye**

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Generation and reproducibility of tissue pattern in an organism depends on tightly controlled cell division and fate determination. The Drosophila EGF Receptor (DER) signalling regulates both these cellular processes and achieves specificity through multiple ligands and feedback loops, which fine-tune signalling spatiotemporally. DER signalling is known to be an important player in the development of the fruit-fly eye, which is made up of precisely arranged units called ommatidia. The reproducibility of the ommatidial pattern depends on highly regulated division and fate determination choices, both controlled by DER signalling. The principle EGF in the fly, cleaved Spitz, and the negative feedback molecule, Argos are diffusible and can act both in a cell autonomous and non-autonomous manner. The relative expression level of Spitz and Argos has been shown to be critical in patterning the Drosophila eye. Quantitative information on the expression levels and the identity of the cells expressing Spitz and Argos in the larval eye disc has been elusive with extant methods. Following diffusible protein products might be problematic for determining cell-based expression, which can be circumvented by detecting mRNA in situ. In this talk, I first describe the adaptation and standardisation of single molecule RNA fluorescent in-situ hybridisation (smFISH) in a variety of wholemount Drosophila tissues. Further, using smFISH against Spitz and Argos mRNA, I will highlight the signatures of directionality in DER signalling in the larval eye disc, by the exclusive cell non-autonomous activity of Spitz. By genetically tuning DER signalling, I show that rather than absolute levels of expression, the ratio of expression of Spitz and Argos to be critical for determining the adult eye phenotype. I will also show that proper ommatidial patterning is robust to thresholds around a tightly maintained wildtype ratio, and breaks down beyond, which provides a powerful instance of developmental buffering.

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***2:00 PM***