Optogenetic approaches in development and cancer

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***Short Abstract* — The development of novel optogenetic tools presents an opportunity to affect physiological networks and quantitatively study their response to local perturbations. We have used the local uncaging of caged-cyclofen to activate many proteins (transcription factors, recombinases, etc.). Here we will present our results using optogenetics to quantitatively study the somitogenetic wavefront in zebrafish and to estimate the probability of tumorigenesis upon turning on a single oncogene in a single cell of zebrafish.**

***Keywords* — Optogenetics, Development, Cancer**

I. Introduction

Living organisms are made of cells that are capable of responding to external signals by modifying their internal state (gene expression or protein phosphorylation patterns) and subsequently their external environment by the release of signaling molecules. In multicellular organisms in particular, cellular differentiation and signaling is essential for the development of the organism. While many of the key actors of these processes are known (morphogens in development, oncogenes in cancer) much less is known of the quantitative rules that govern their interaction with one another and with other cellular players. New optogenetic tools can now be used to probe these interactions by local spatio-temporal perturbations of the key actors in these processes.

II. Results

In this talk I will present our results regarding the development of means to optically control the expression and activity of proteins at the single cell level in a live organism and its use as a tool to study quantitatively somitogenesis and tumorigenesis in zebrafish. Our basic idea is to use light to control the activity of proteins through the uncaging of a small ligand (cyclofen). The photo-activated ligand releases the targeted protein fused with the ligand receptor from its complex with cytoplasmic chaperones[1]. The protein (transcription factor, recombinase, etc.) can then turn on the expression of a desired gene (encoding for an oncogene or a morphogen).

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We have used that approach to perturb the morphogen gradients involved in the propagating wavefront of somitogenesis. Using time-lapse microscopy and In Situ Hubridization we have quantified the response to that perturbation and compared it to the predictions of a wavefront model of somitogenesis[2]. We have also used local optogenetic activation to turn on a single oncogene in single cells of a zebrafish[3] and measured the probability of tumorigenesis which is extremely small (about 0.5%).

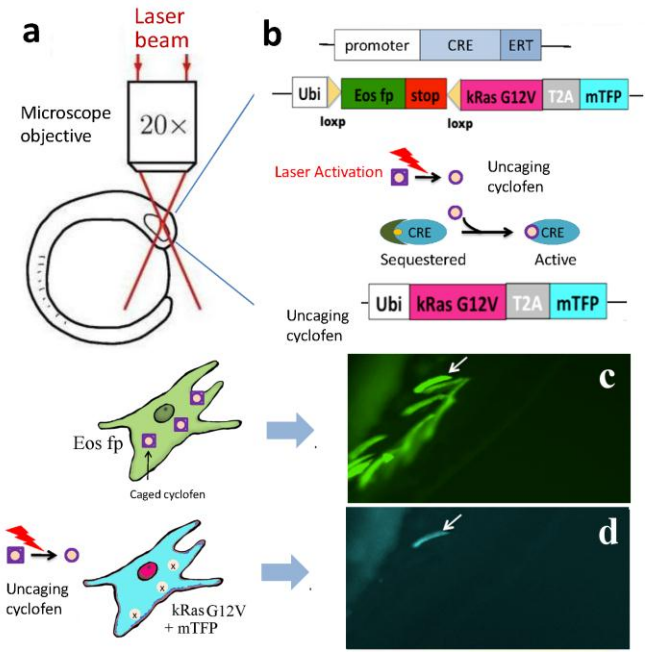

Fig.1: permanent activation of an oncogene: kRasG12V. Local photo-activation by a 2-photon laser (a) of caged cyclofen, results in activation of a Cre-ERT recombinase (b) which floxes a EosFP insertion marker (c) and turns on the expression (d) of the oncogene and its marker (mTFP).

III. Conclusion

Optogenetics opens a new vista on the quantitative study of physiological networks via their response to local spatio-temporal perturbations.

References

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