

## Spatial organization and dynamics of cortical actin network in living cells revealed with the instantaneous FluoPolScope

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We developed a polarized light imaging system, the *Instantaneous FluoPolScope*, that reports the position and the dipole orientation of single fluorophores in live cells [1]. Our imaging is useful for studying assembly/disassembly dynamics and the spatial organization of biological molecules (nucleotides, lipids, polysaccharides and proteins) in living cells. We have used this imaging system to explore the spatial organization of actin filaments at the cell cortex of fertilized eggs. Spatial reorganization of cytoplasm in zygotic cells is critically important for establishing the body plans of many animal species. In ascidian zygotes, maternal determinants (mRNAs) are first transported to the vegetal pole a few minutes after the fertilization, and then to the future posterior side of the zygotes in later phase of the cytoplasmic reorganization, before the first cell division [2]. By using the instantaneous FluoPolScope, we mapped the local alignments and the time-dependent changes of cortical actin networks in ascidian, *Ciona* eggs. The initial cytoplasmic reorganization started with the contraction of vegetal hemisphere ~20s after the fertilization induced  $[Ca^{2+}]$  increase. Timing of the vegetal contraction was consistent with the emergence of highly aligned actin filaments at the cell cortex of vegetal hemisphere which ran perpendicular to the animal-vegetal axis. We propose that the first ooplasmic segregation is initiated by the local contraction of laterally aligned cortical actomyosin in the vegetal hemisphere, which in turn generates the convective flow of cytoplasm within whole eggs.

### References

- [1] Mehta, S. B., McQuilken, M., Riviere, P. J. L., Occhipinti, P., Verma, A., Oldenbourg, R., Gladfelter, A. S. and Tani, T. (2016). Dissection of molecular assembly dynamics by tracking orientation and position of single molecules in live cells. *PNAS* **113**, E6352–E6361.
- [2] Sardet, C., Paix, A., Prodon, F., Dru, P. and Chenevert, J. (2007). From oocyte to 16-cell stage: Cytoplasmic and cortical reorganizations that pattern the ascidian embryo. *Developmental Dynamics* **236**, 1716–1731.