Faculty of Science

Vorticity patterns in Tissues induced by Cell divisions

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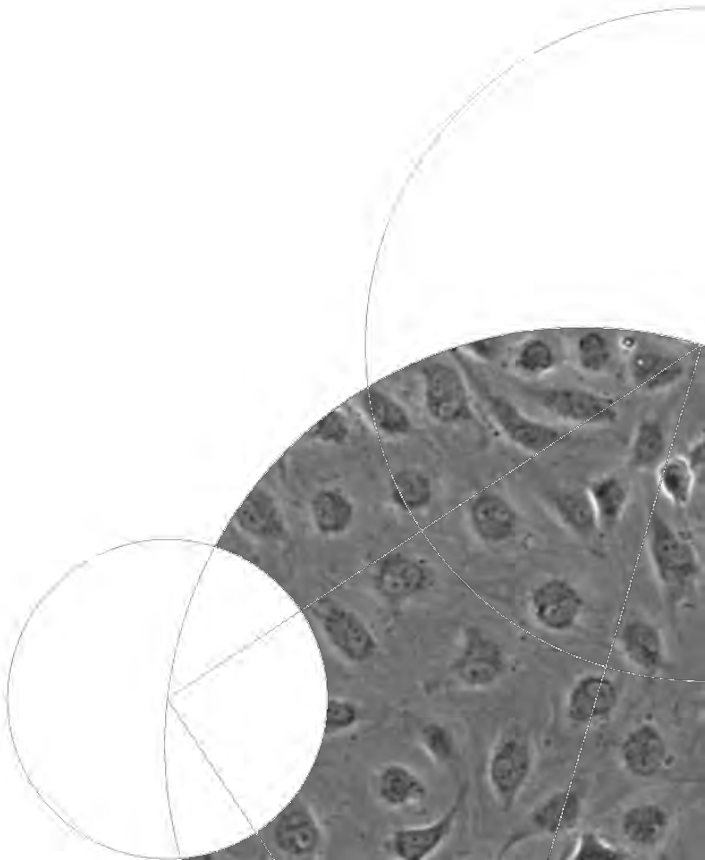
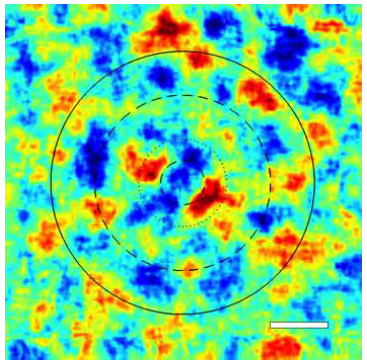
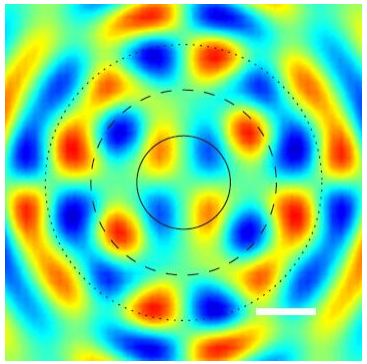


1. Confluent layer of endothelial cells:
   cell divisions cause 'motion'.
2. Track motion by PIV analysis:
   Achive velocity fields.
3. From velocity → vortiticy field:
   Primary, secondary, tertiary vortices.



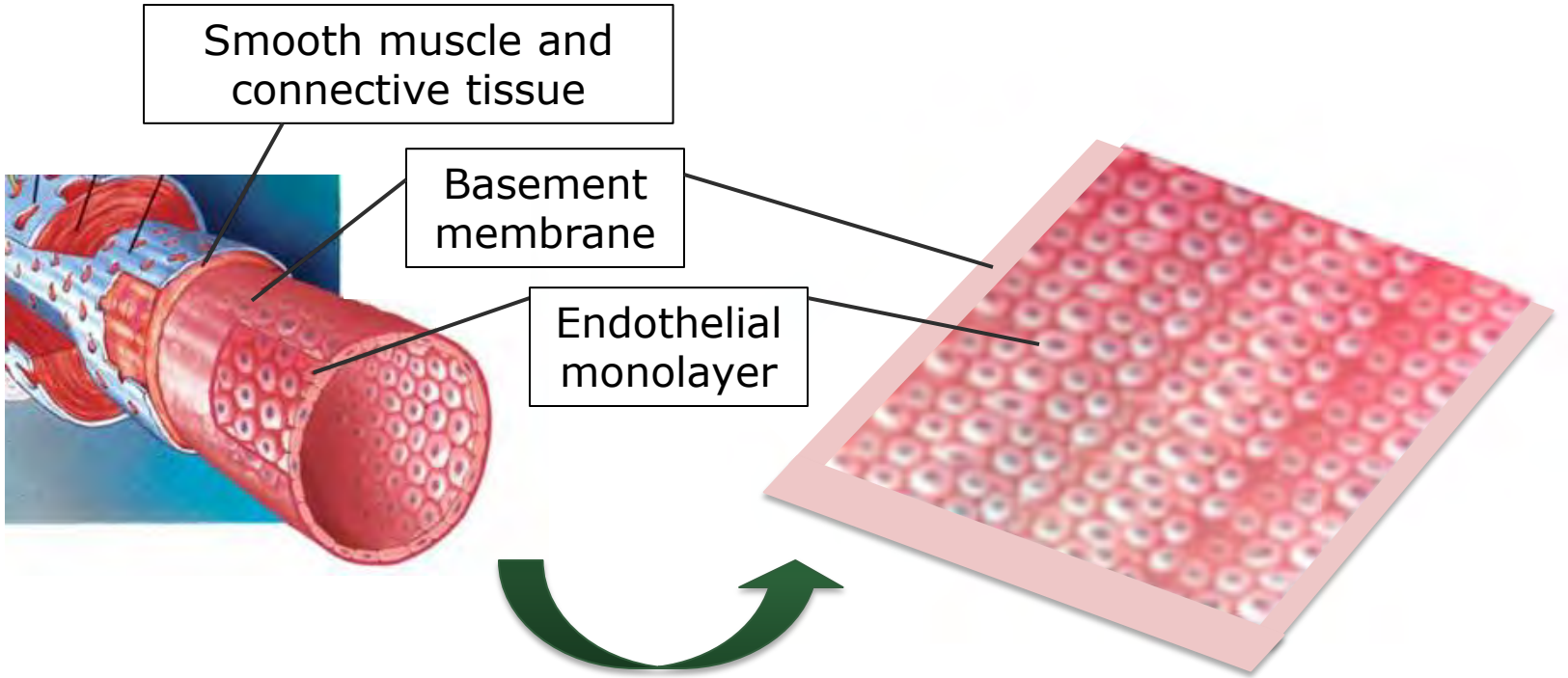
4. Model with Swift-Hohenberg model in velocity:
   Cells motion inject energy locally
5. Good agreement
   between theory and experiment.



"Long-range ordered vorticity patterns in living tissue induced by cell division", Nature Physics, in process (2013)

Our *in vitro* biological system

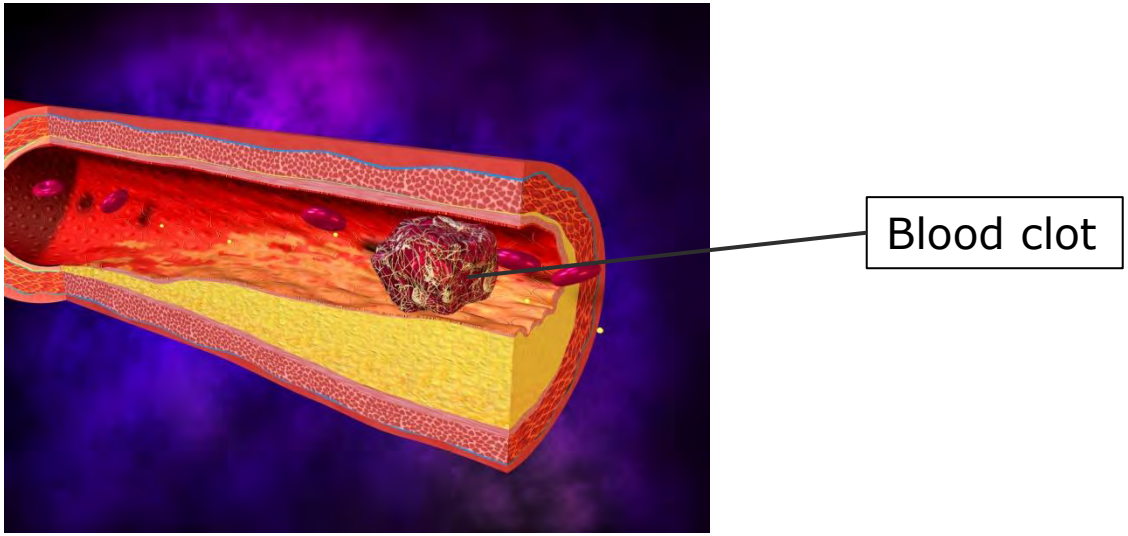
We monitored individual cell divisions in an *in vitro* confluent monolayer of endothelial cells.



No flow conditions

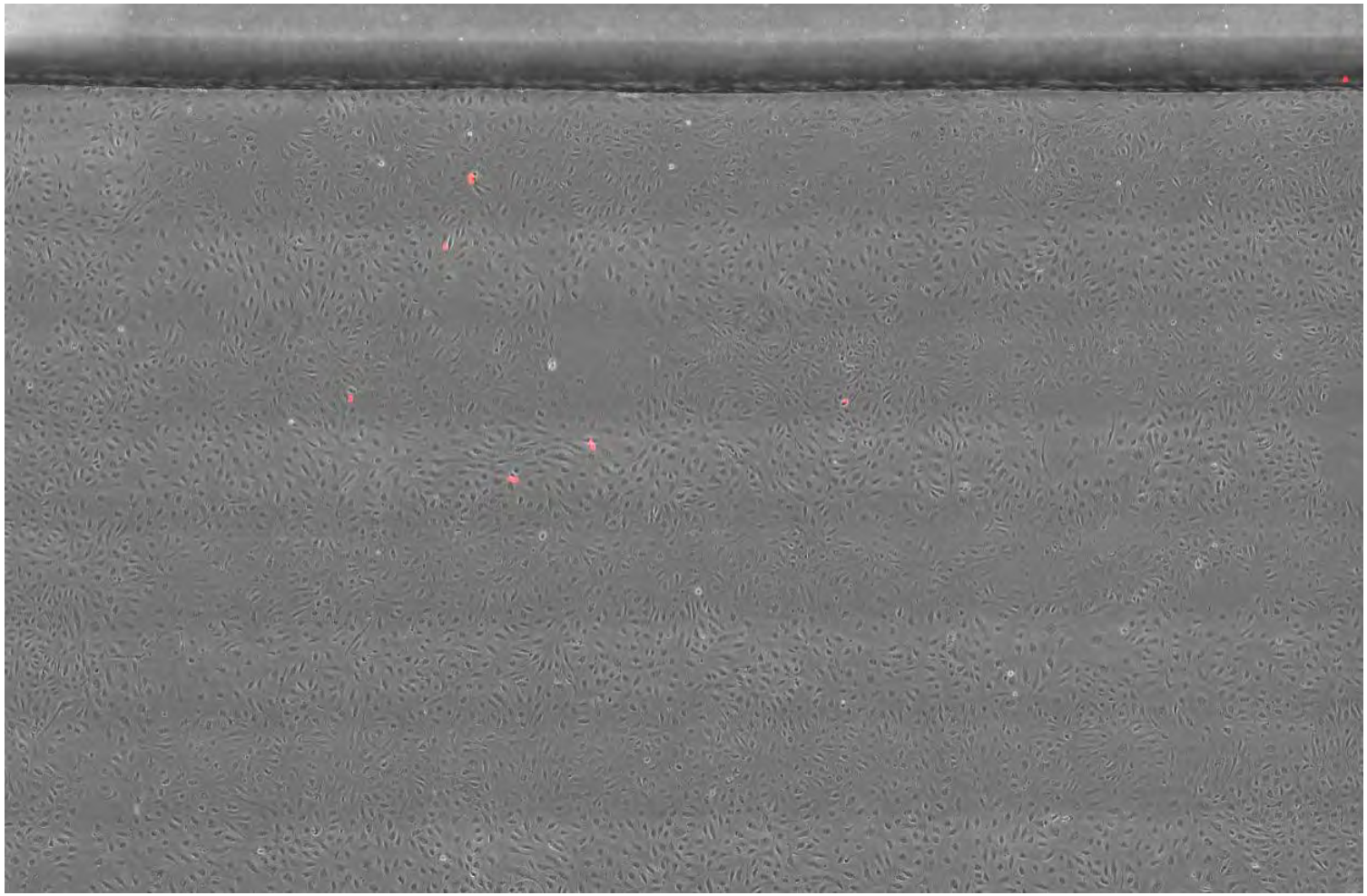
Cell division rate: significantly increased around tumors and around halted blood flows (blood clots).

Understanding how a small fluctuation arising form cellular division may affect the entire tissue can be crucial for understanding the growth of malignant tissues and for the healing process of blood clots.



Divisions in a monolayer without flow (18-24 hours)

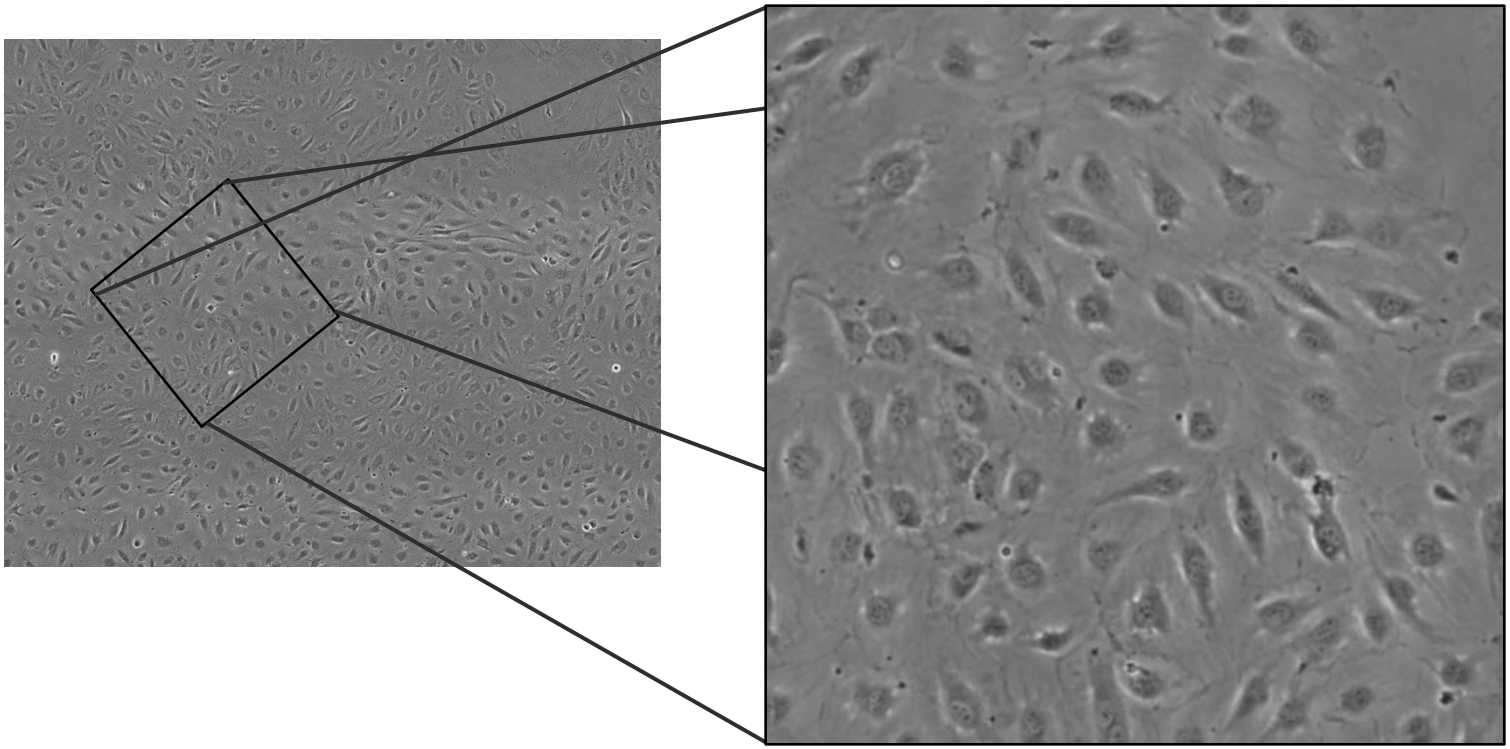
Dividing cell and daughter cells are highlighted in red



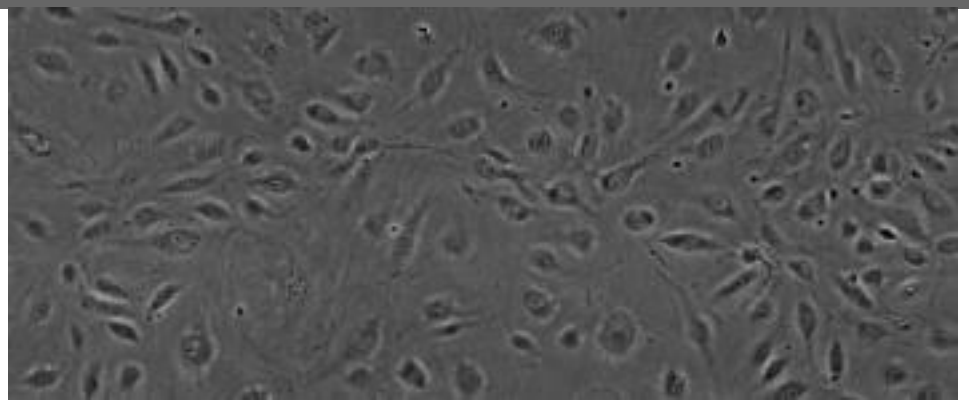
Zoom-in on cell division

Each cell division was followed for 90 minutes before and after cell division.

A 300x300 μm^2 movie was made, with the frame adjusted so that the cell division occured horizontally and in the middle.



Analyzing the velocity field using Particle Image Velocimetry (PIV)

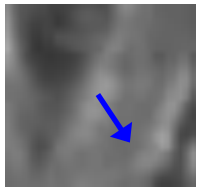


t_0 $t_1=t_0+10$

Interrogation area image input


Cross-correlation $$C(s) = \int \int_{IA} I_1(X) \cdot I_2(X - s) dX$$

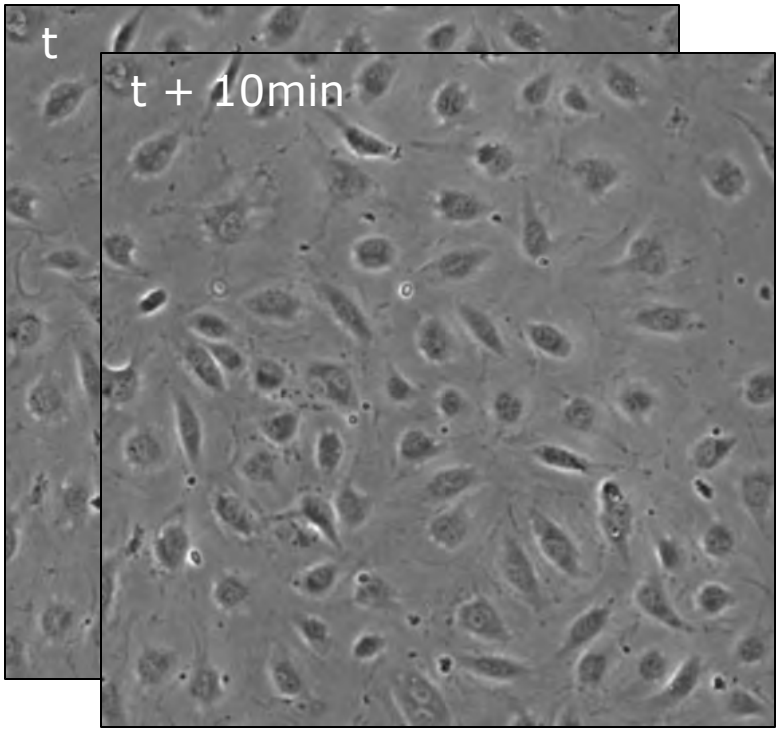
Max C(s) gives vector output



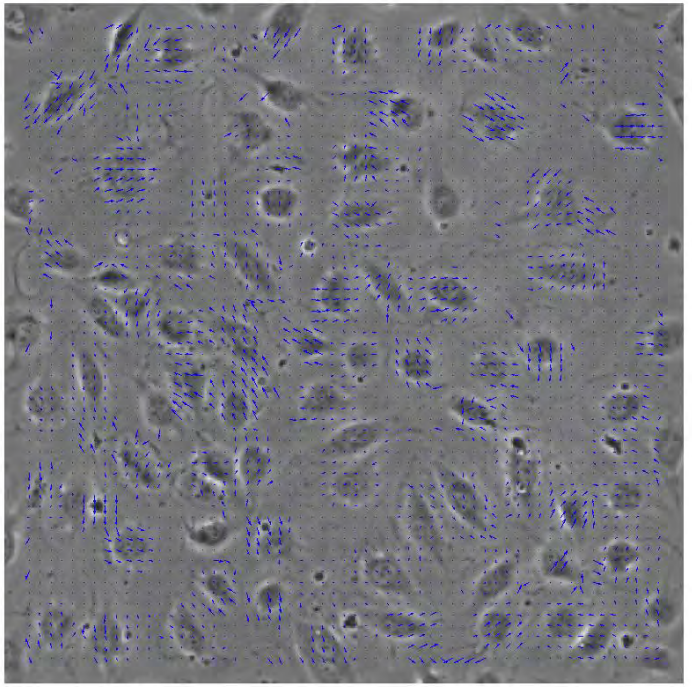
PIV quantifies the collective cell motion

PIV analyzes phase contrast images if the cells do not move too much.

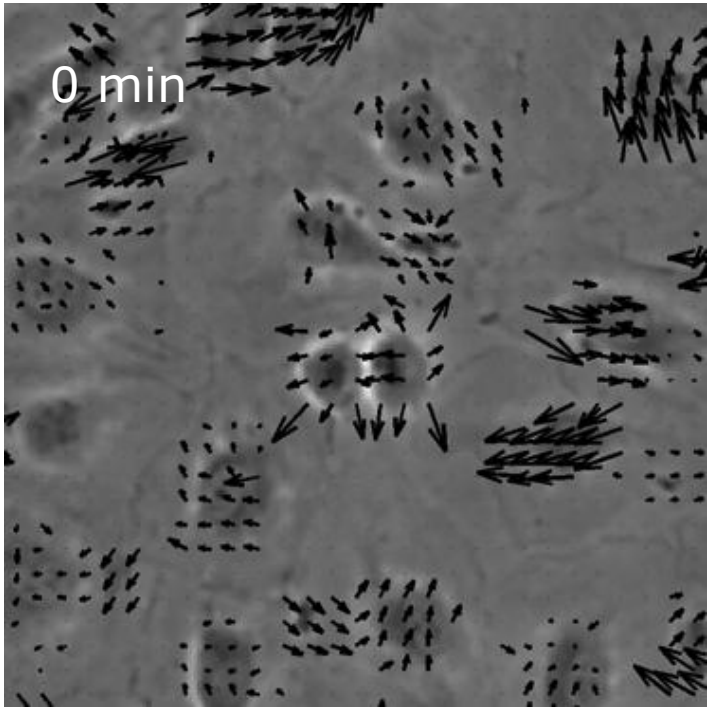
For endothelial cells this is possible by images taken at 10 minute intervals.

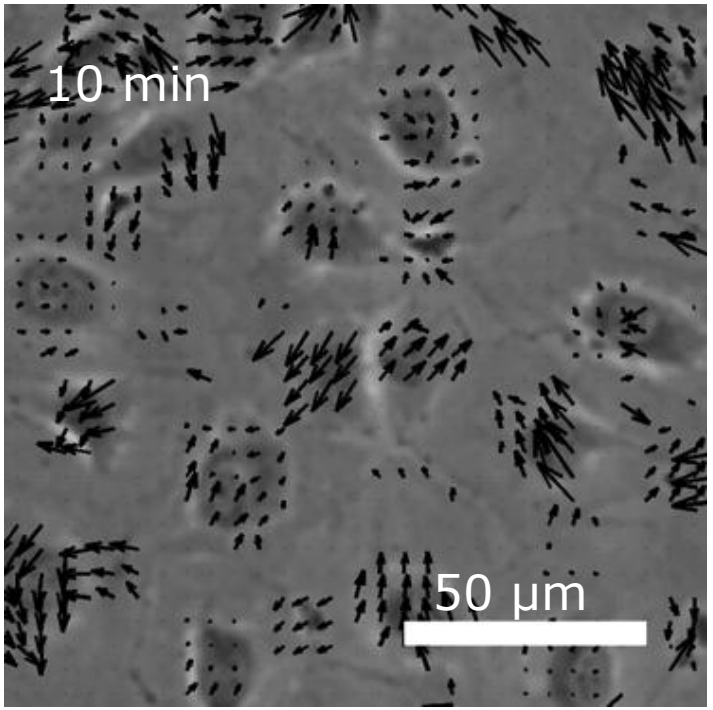


PIV



Divergence and Vorticity from the Velocity-field



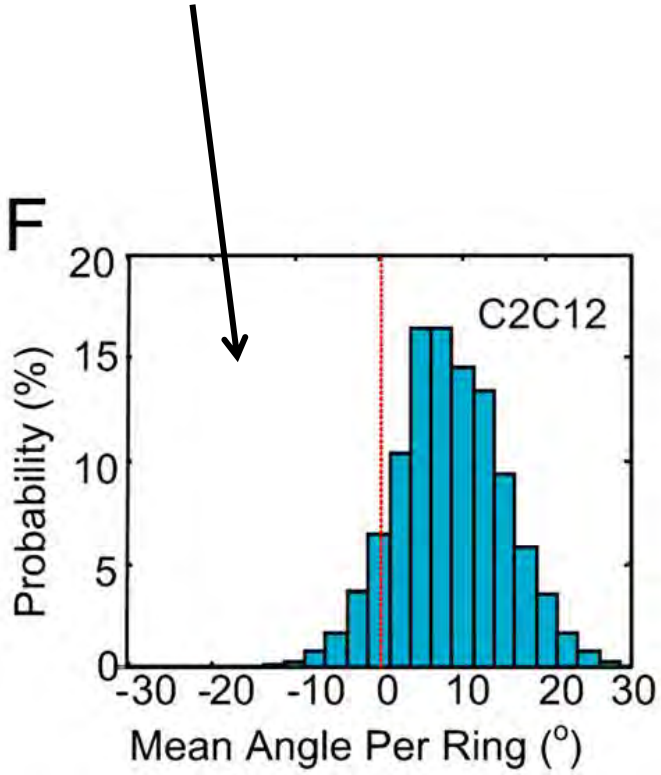


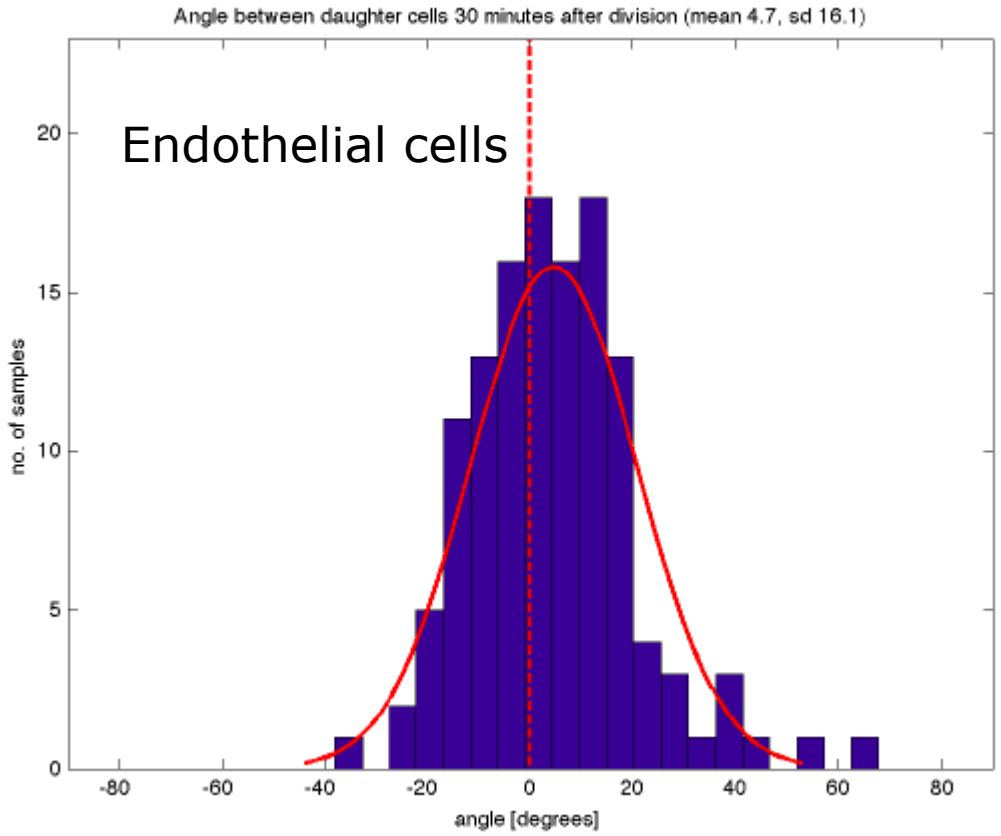
Divergence: $$d = \frac{\partial}{\partial x} v_x + \frac{\partial}{\partial y} v_y = \sum_{r \in O} \frac{v_x(r) r_x + v_y(r) r_y}{A}$$

Vorticity: $$\omega = \frac{\Gamma}{A} = \frac{\oint \vec{v}(r) \cdot \vec{dr}}{A} = \sum_{r \in O} \frac{v_x(r) r_y - v_y(r) r_x}{A}$$

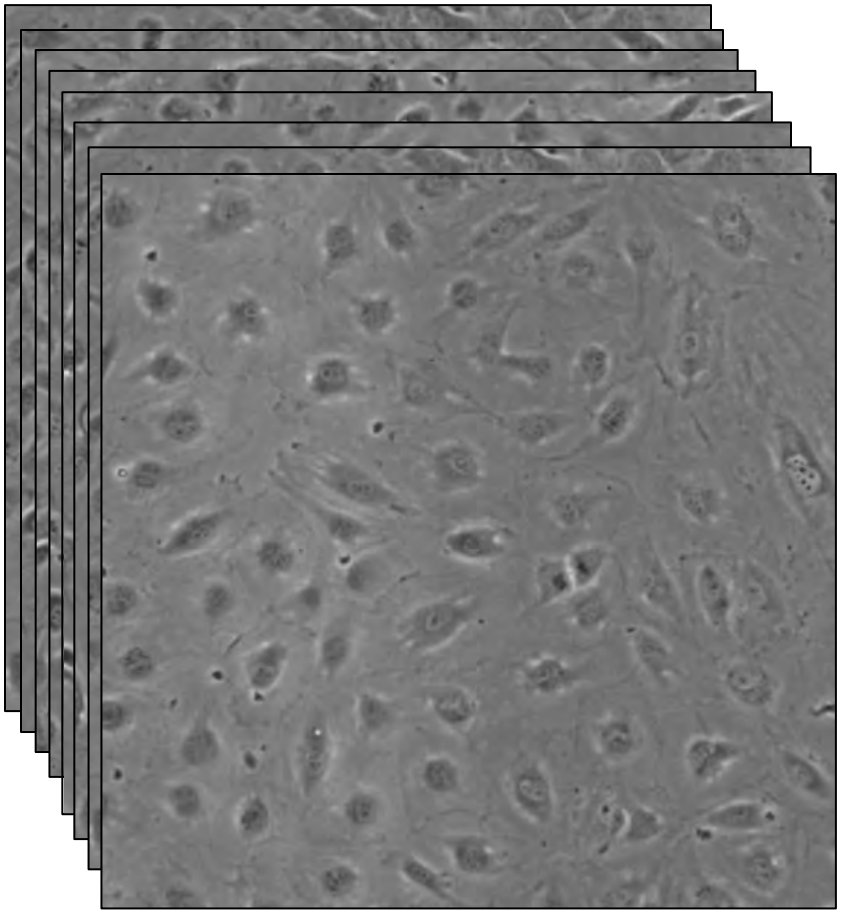
Symmetry of cell division plane

On average, there is no preferred rotational direction for the endothelial daughter cells. This is in contrast to the behavior of, e.g., HeLa cells (Nature Cell Biology, vol.7 p.948 2005) and mouse myoblasts (PNAS, vol 108 p.12295 2011)

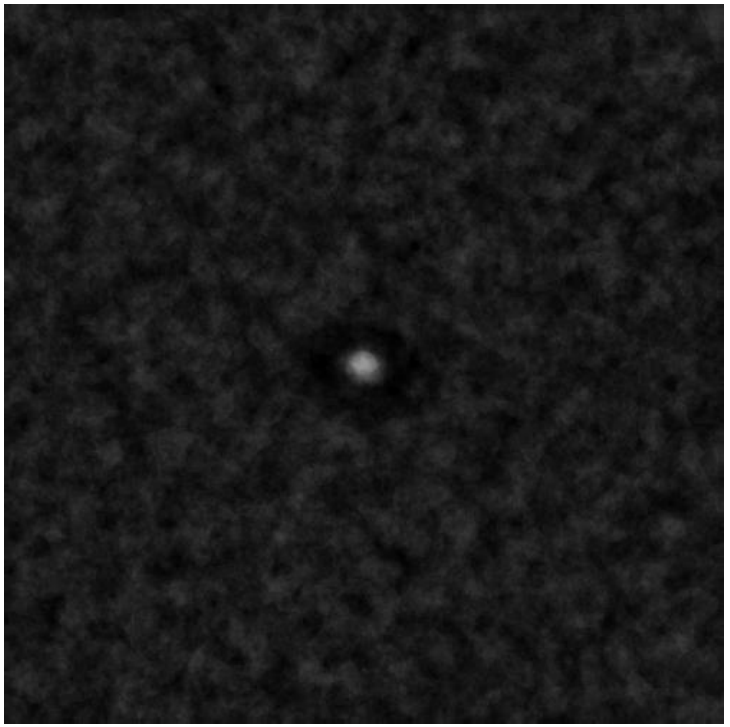




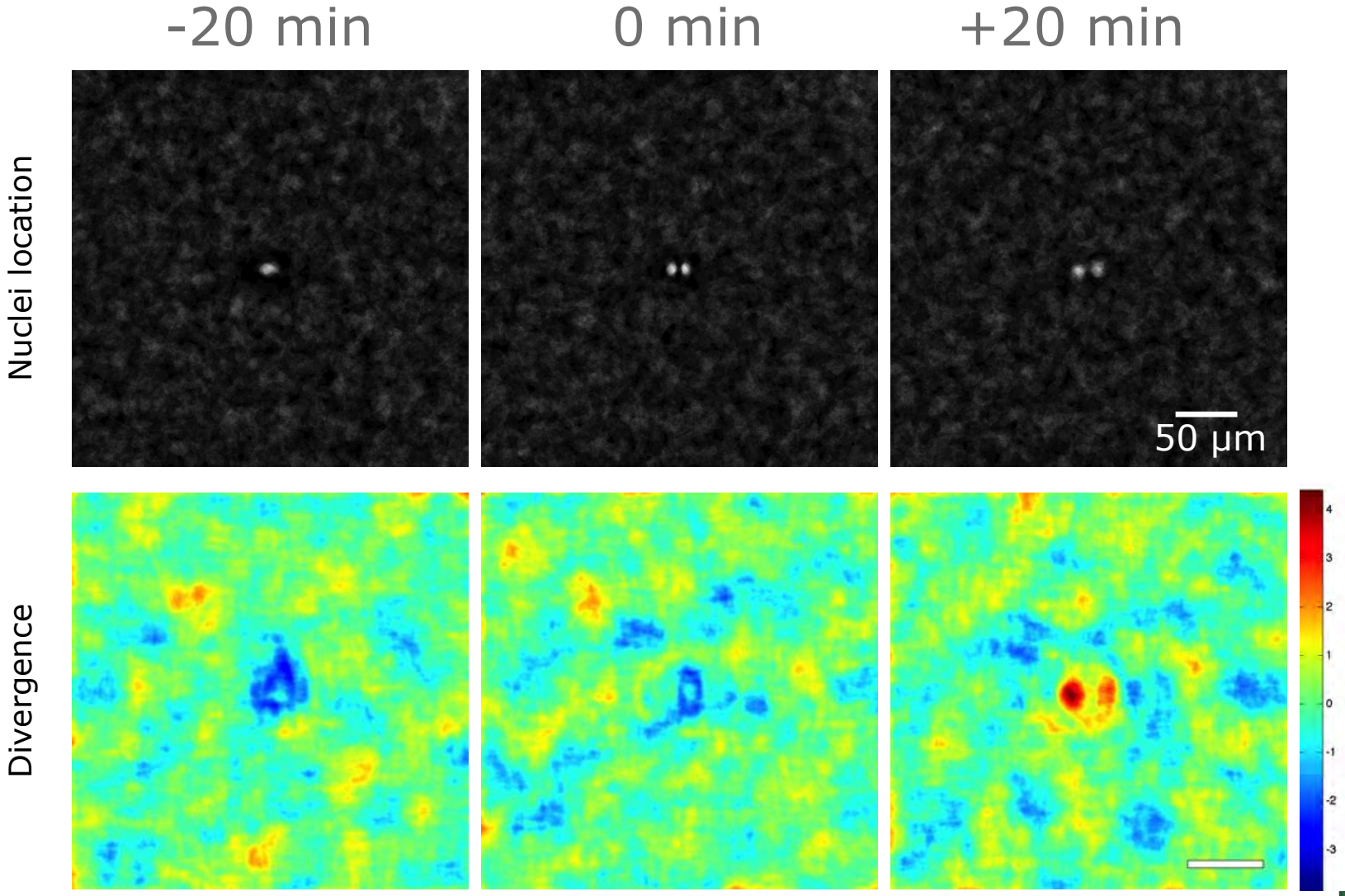
The motility parameters averaged over 30 cell divisions



The positions of cell nuclei (white) averaged over 100 cell divisions

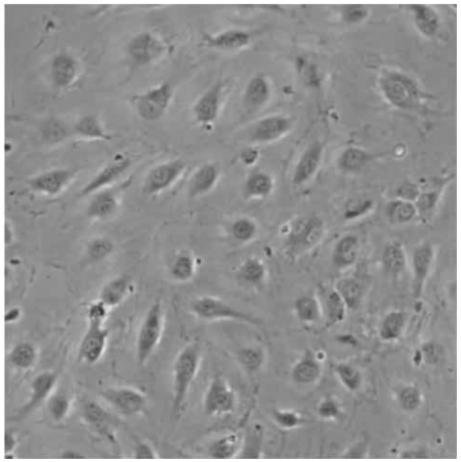


Divergence changes significantly during cell division

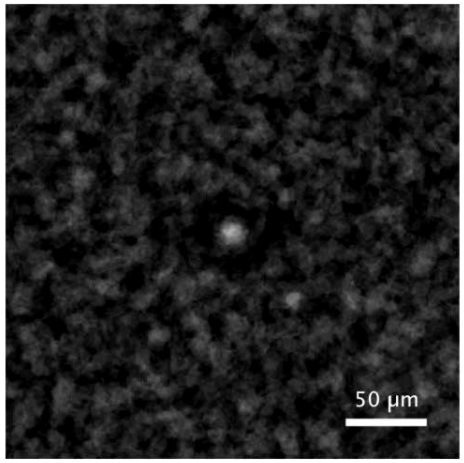


Single cell, averages, divergence, vorticity

Single "Isolated" Cell Division



Average Nuclei Density



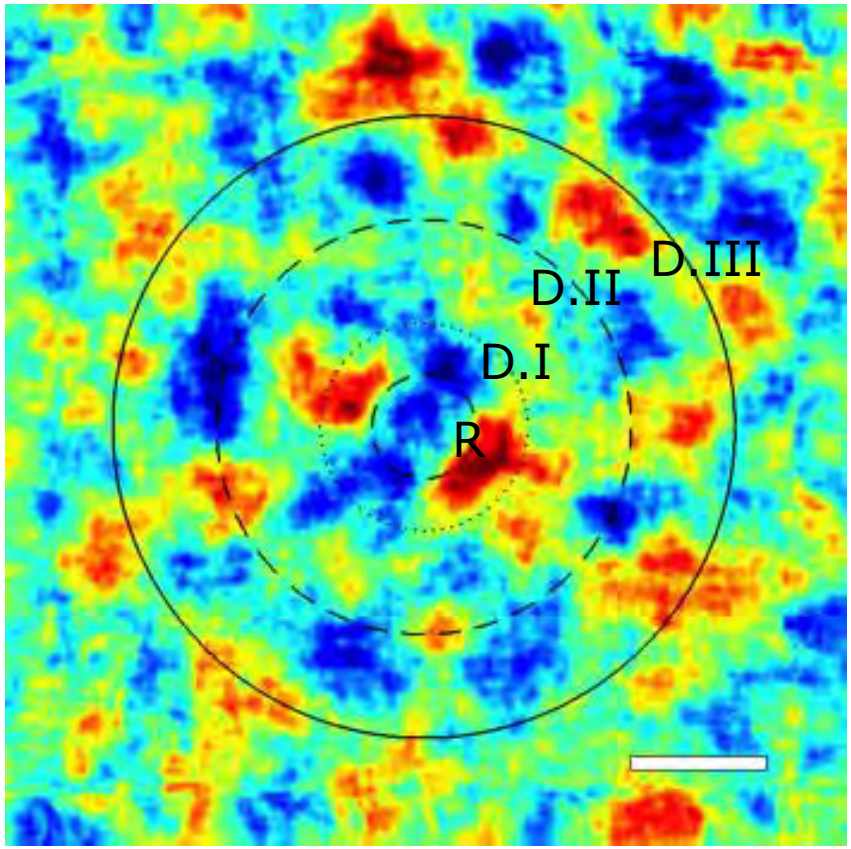
Average Divergence [min^{-1}]

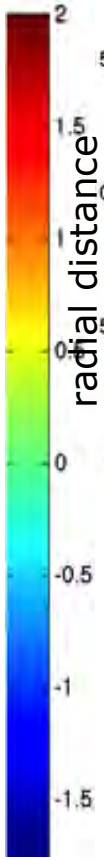
Average Vorticity [min^{-1}]

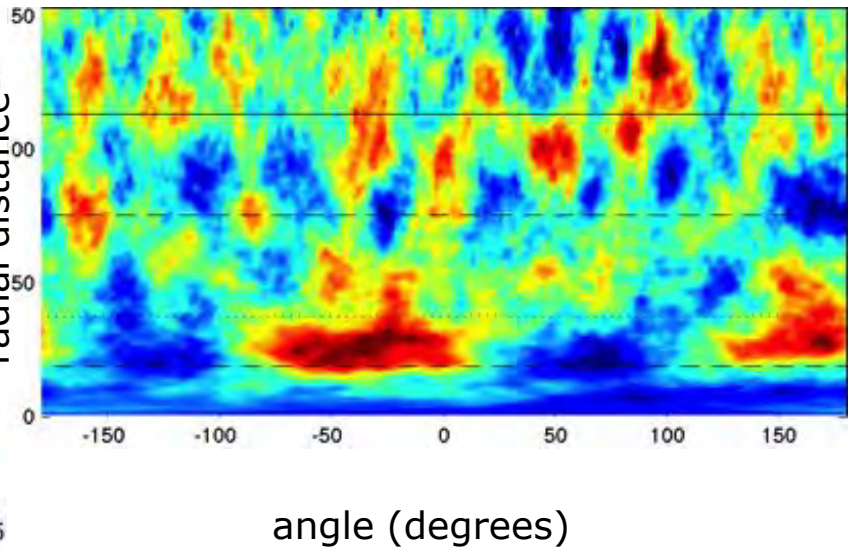
Time: −75 min

(Ninna Rossen)

Primary and secondary vortices arise around a dividing cell



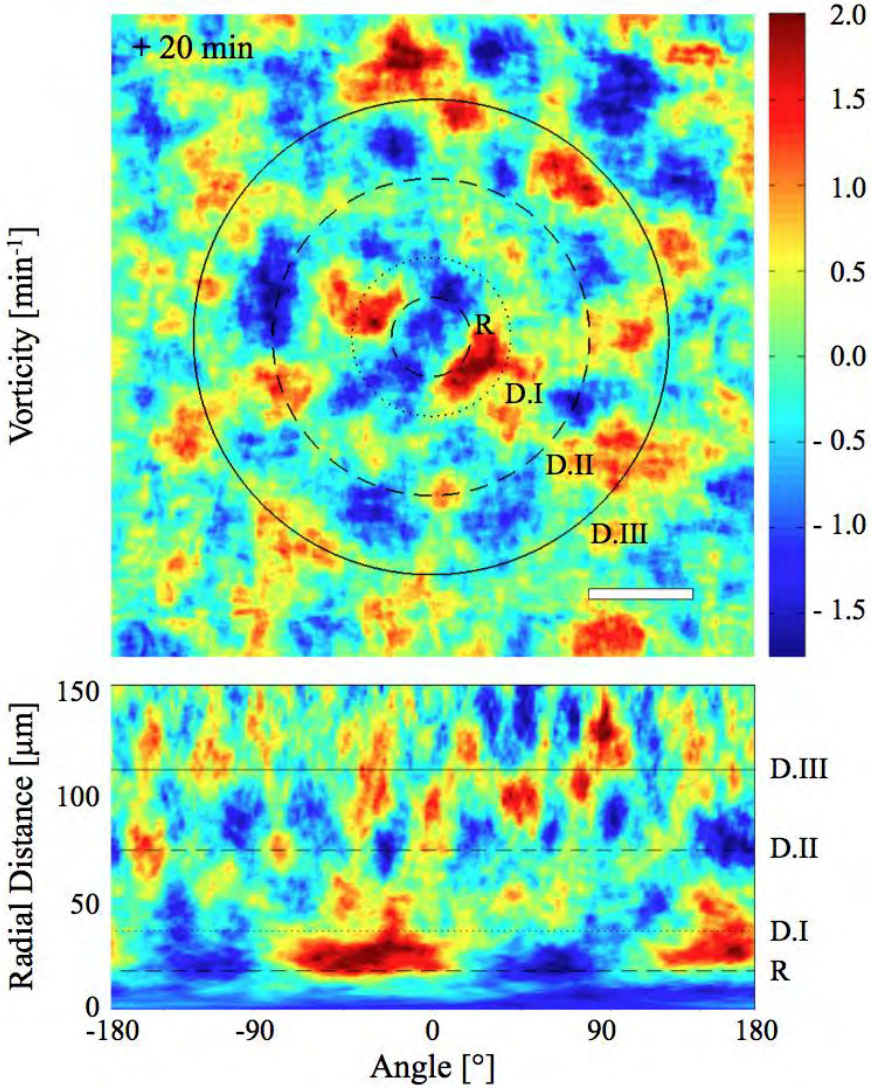




Reynolds number ~10^{-3}

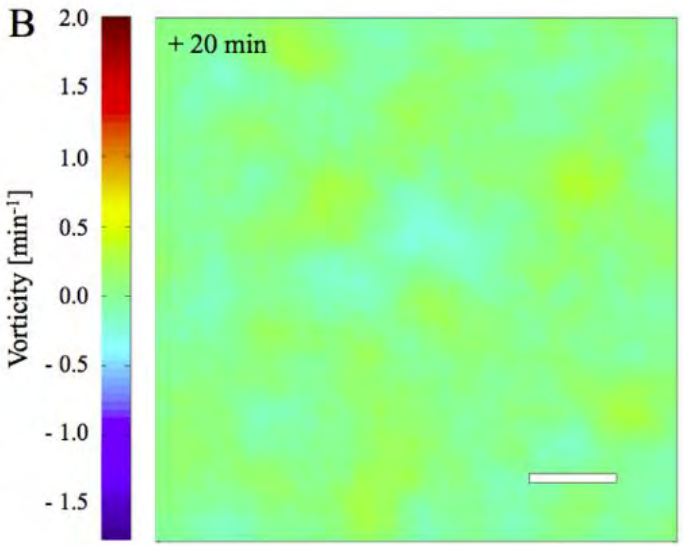
R: one cell radius
D.I: one cell diameter
D.II: two cell diameters
D.III: three cell diameters

A second example

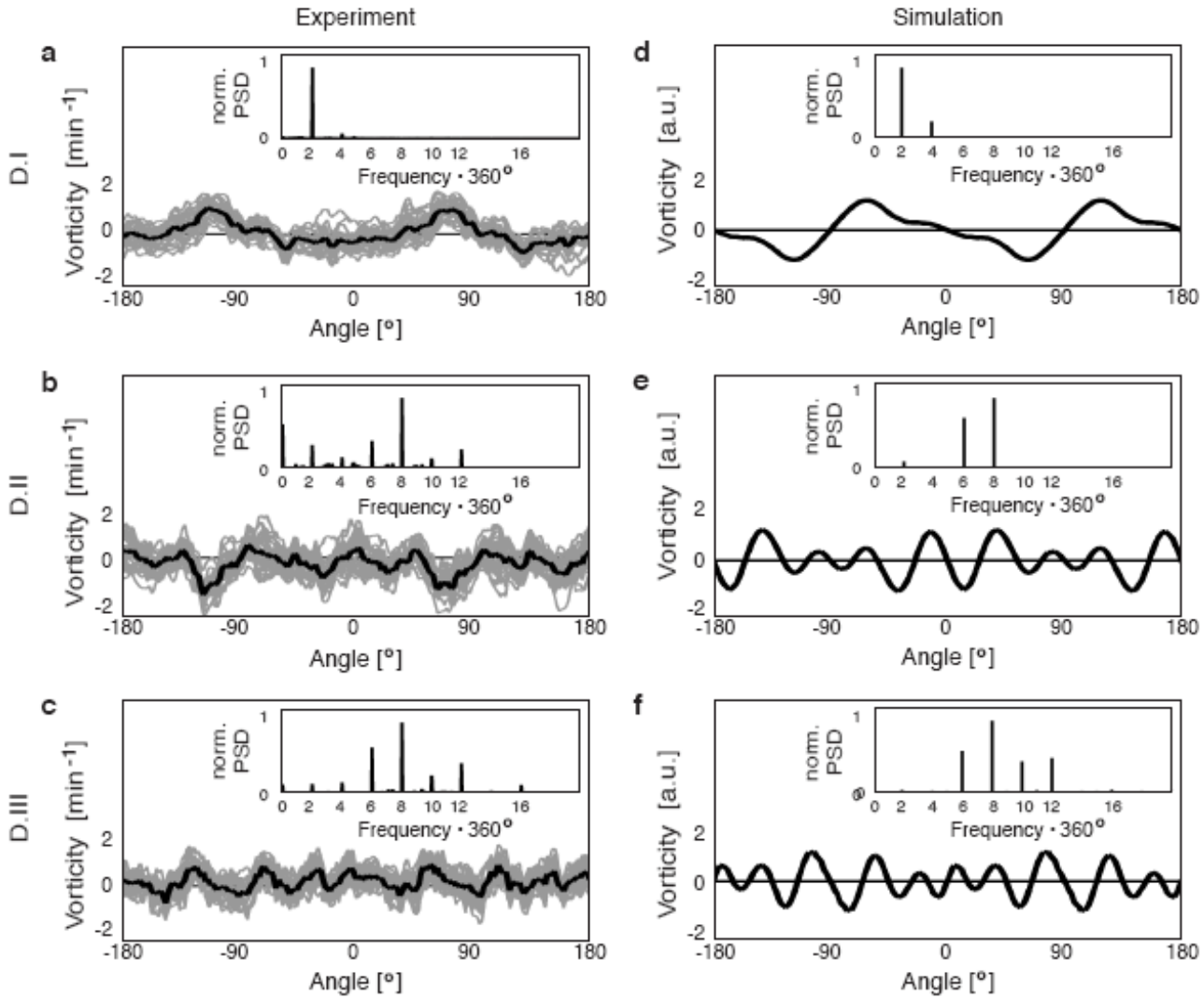


R: one cell radius
D.I: one cell diameter
D.II: two cell diameters
D.III: three cell diameters

Control without divisions

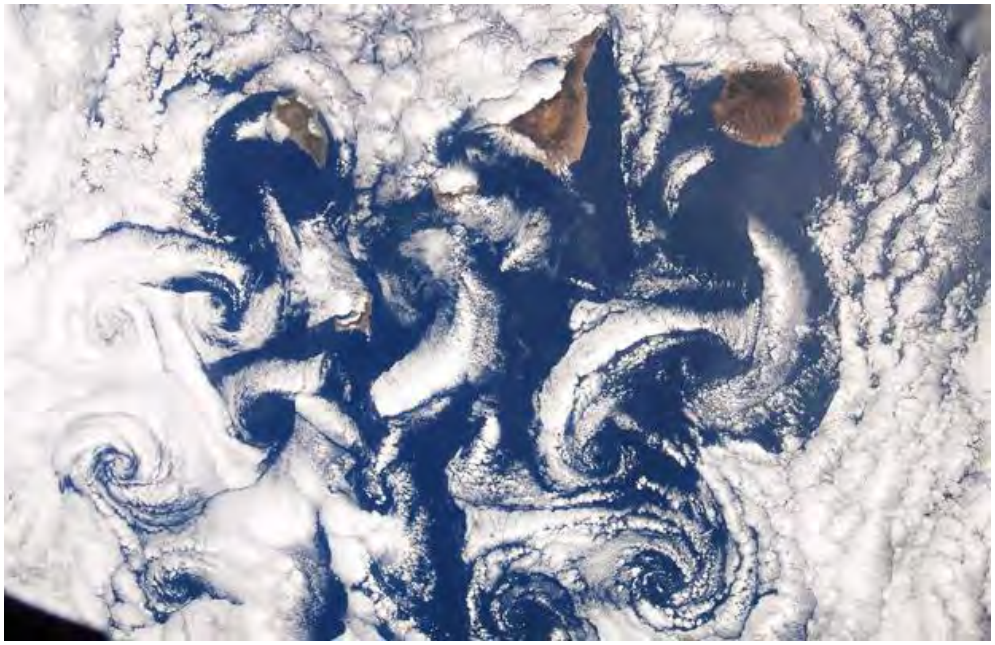


Quantification of vorticity by Fourier analysis



Emergence of secondary vortices in other systems

Airflows around winglets



Clouds

Visco-elastic simulation

Theoretical model

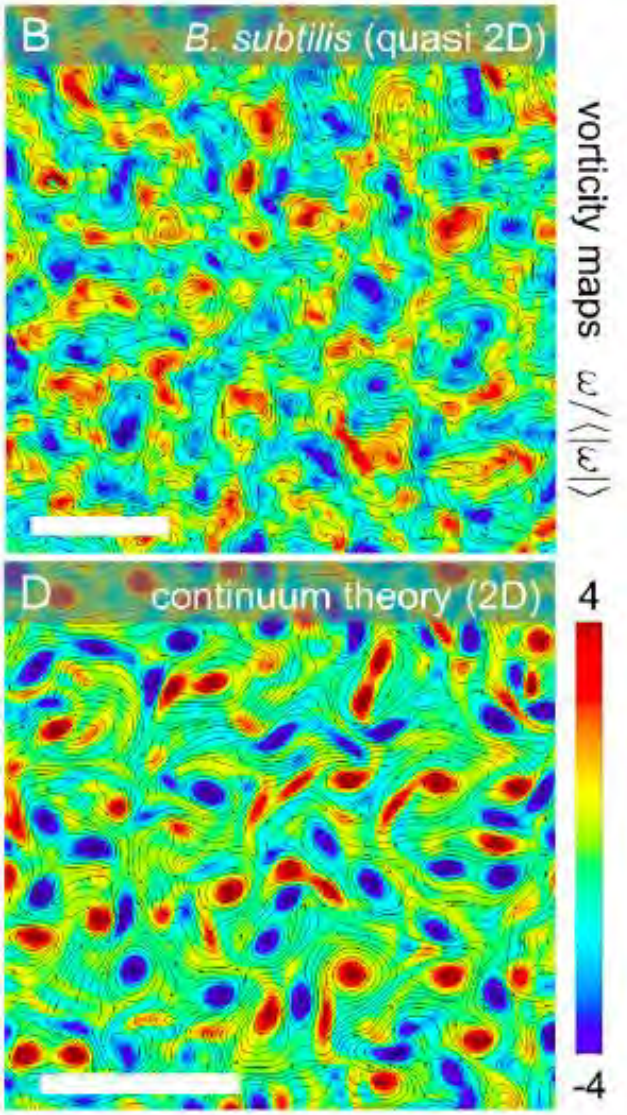
Reynolds number=10^{-3} - 10^{-5}, system not classically turbulent.

Inspiration from '**Mesoscale turbolence** in living fluids' by Goldstein, Yeomans (PNAS vol. 109 p. 14308, 2012)

Mathematical characterization of turbulence phenomena in active non-equilibrium fluids. Apply continuum theory to describe motion within quasi 2D *Bacilus subtilis* suspensions.

$$(\partial_t + \lambda_0 \boldsymbol{v}\cdot\nabla)\boldsymbol{v} = -\nabla p + \lambda_1 \nabla \boldsymbol{v}^2 - (\alpha + \beta|\boldsymbol{v}|^2)\boldsymbol{v} + \Gamma_0 \nabla^2 \boldsymbol{v} - \Gamma_2 (\nabla^2)^2 \boldsymbol{v},$$

For $\Gamma_0<0$ and $\Gamma_2>0$ the model exhibits a range of unstable modes, resulting in turbulent states.



Theoretical Model:

Momentum eq (**v** order parameter):

$$\partial_t \mathbf{v} + (\mathbf{v}\cdot\nabla)\mathbf{v} = \frac{1}{\rho}\nabla\cdot\sigma - (\alpha + \beta|\mathbf{v}|^2)\mathbf{v}$$

Charac. speed

$$v_c = \sqrt{\frac{|\alpha|}{\beta}}$$

Generalized stress tensor:

$$\sigma_{ij} = -p\delta_{ij} + \eta_0(\partial_i v_j + \partial_j v_i) - \eta_2\nabla^2(\partial_i v_j + \partial_j v_i)$$

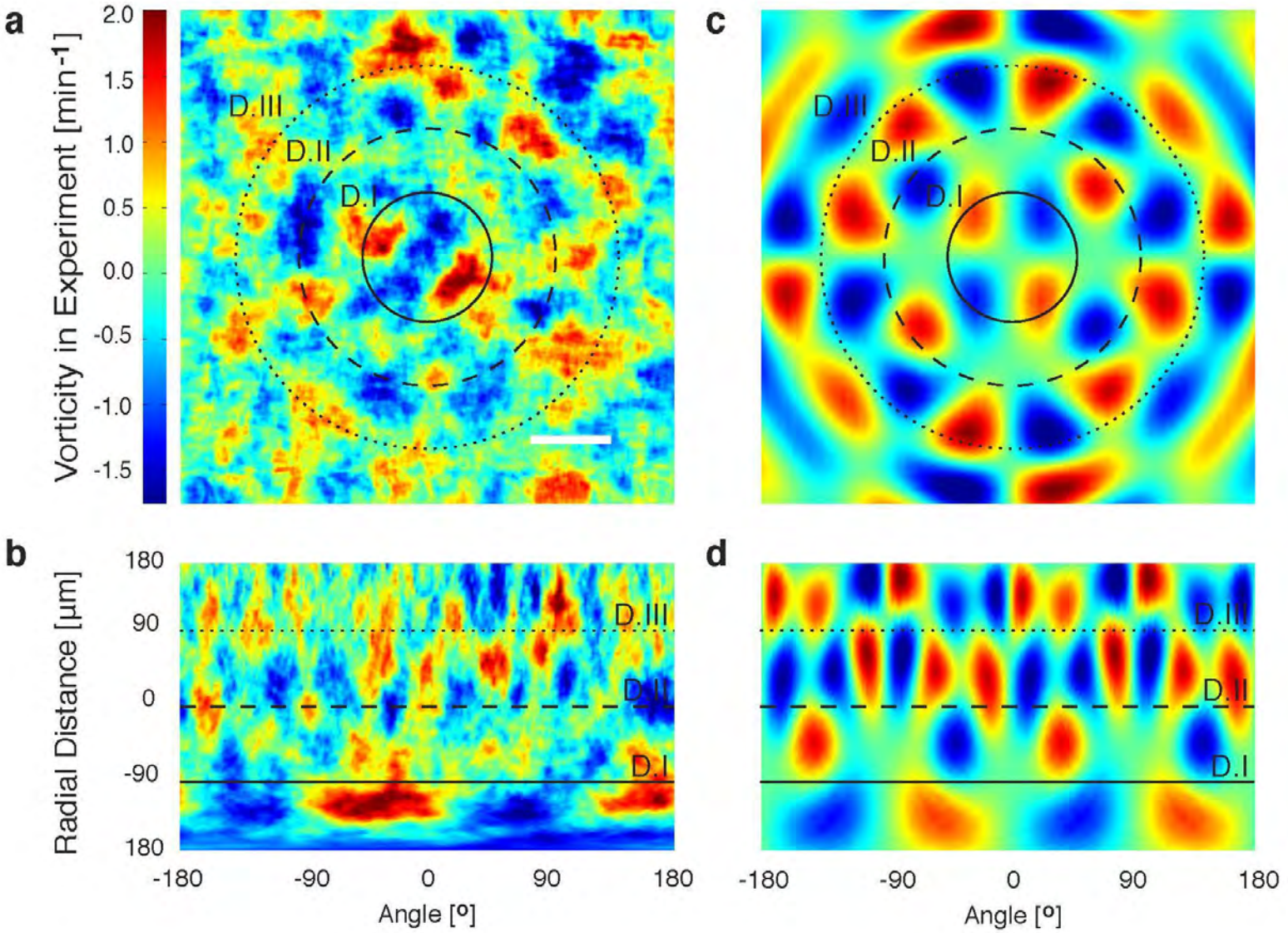
$$\eta_0 < 0 \qquad \eta_2 > 0$$

Swift-Hohenberg model in velocity field:

$$\partial_t \mathbf{v} + (\mathbf{v}\cdot\nabla)\mathbf{v} = -\frac{1}{\rho}\nabla p + \nu_0\nabla^2\mathbf{v} - \nu_2\nabla^4\mathbf{v} - (\alpha + \beta|\mathbf{v}|^2)\mathbf{v}$$

$$\nu_0 = \eta_0/\rho \text{ and } \nu_2 = \eta_2/\rho$$

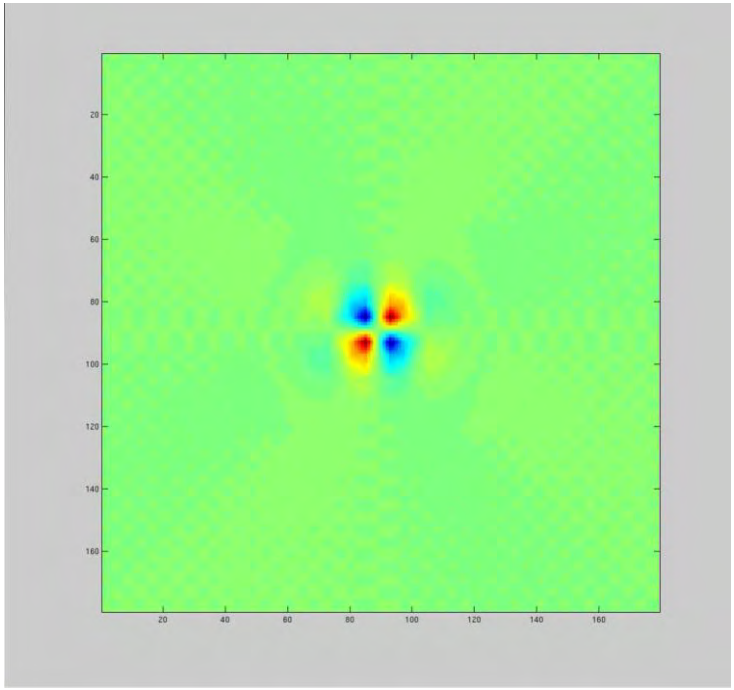
Comparison between experimental results and simulation: vorticity

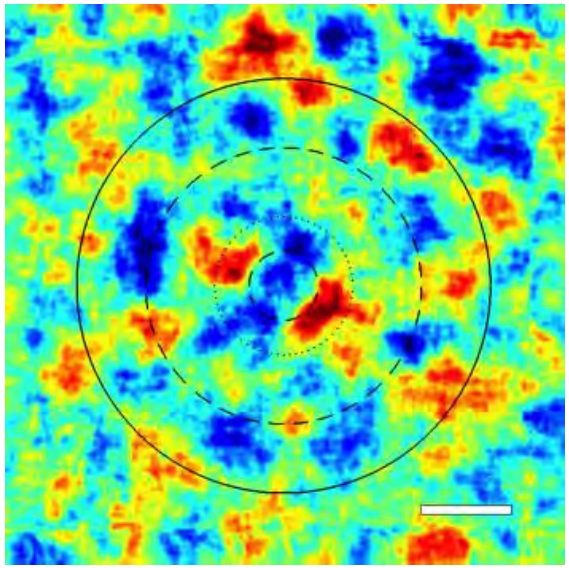


Application of Continuum model to 'meso-scale' turbulence in cell division

Γ_0<0, a negative viscosity makes the entire sheet unstable

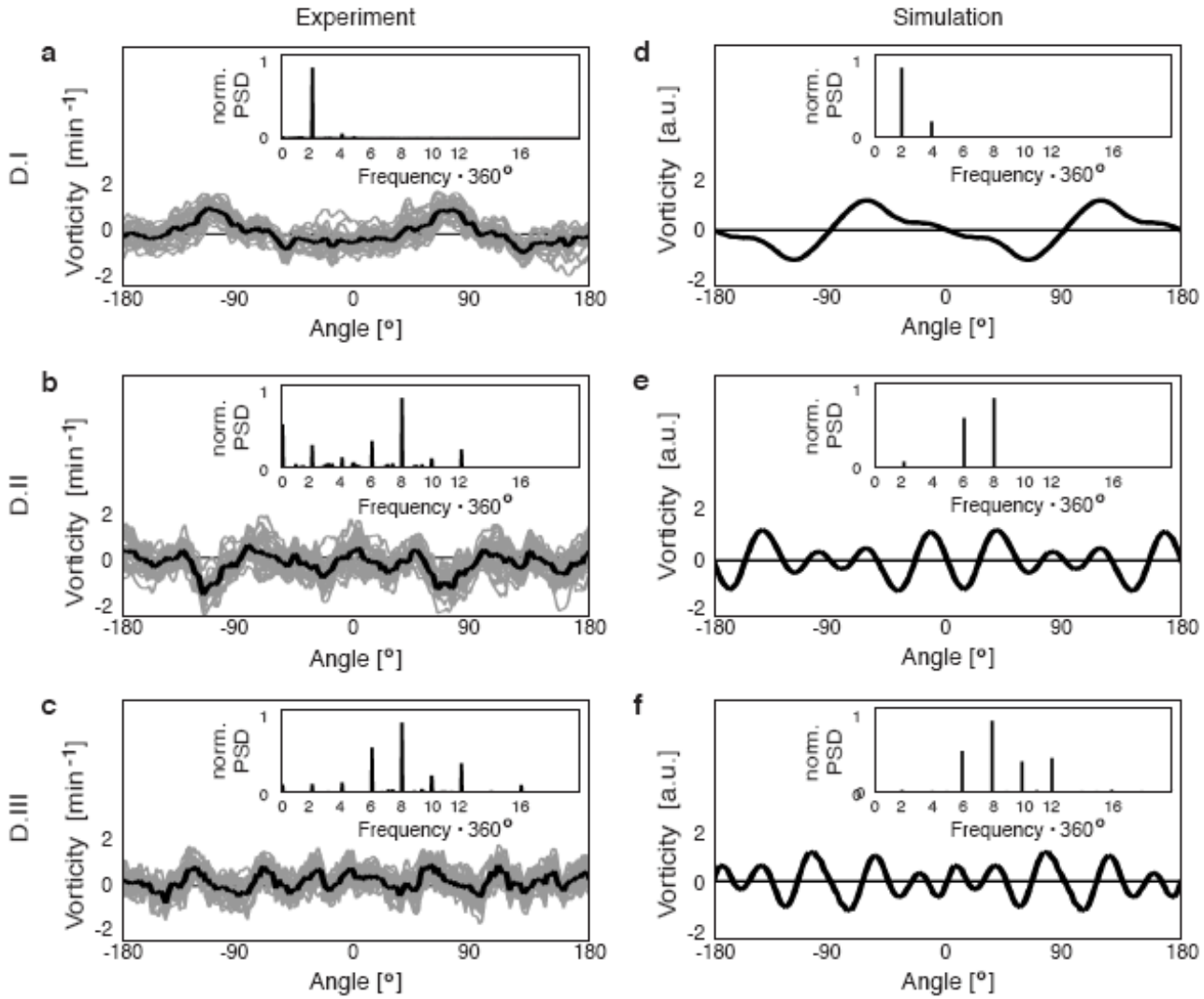
A local pressure increase in the center of sheet mimicks the disturbance (or local injection of energy) caused by the cell division.





(Jens Terp)

Comparison between experiments and simulations: power spectral analysis



Conclusions:

Long range ordering of endothelial cell tissue around a division site: not random.

Re~10^{-3}: several rings of vortices appear, even several cell diameters away from the dividing cell.

Hydrodynamical continuum model: Velocity as order parameter.

Simulating cell division with a local pressure increase.

Understanding of hydrodynamic properties of bio-material such as blood vessels: healing of endothelial tissue and for successful creation of artificial blood vessels.

Acknowledgements

Collaborators:

Ninna Struck Rossen (graduate student, NBI, exp)

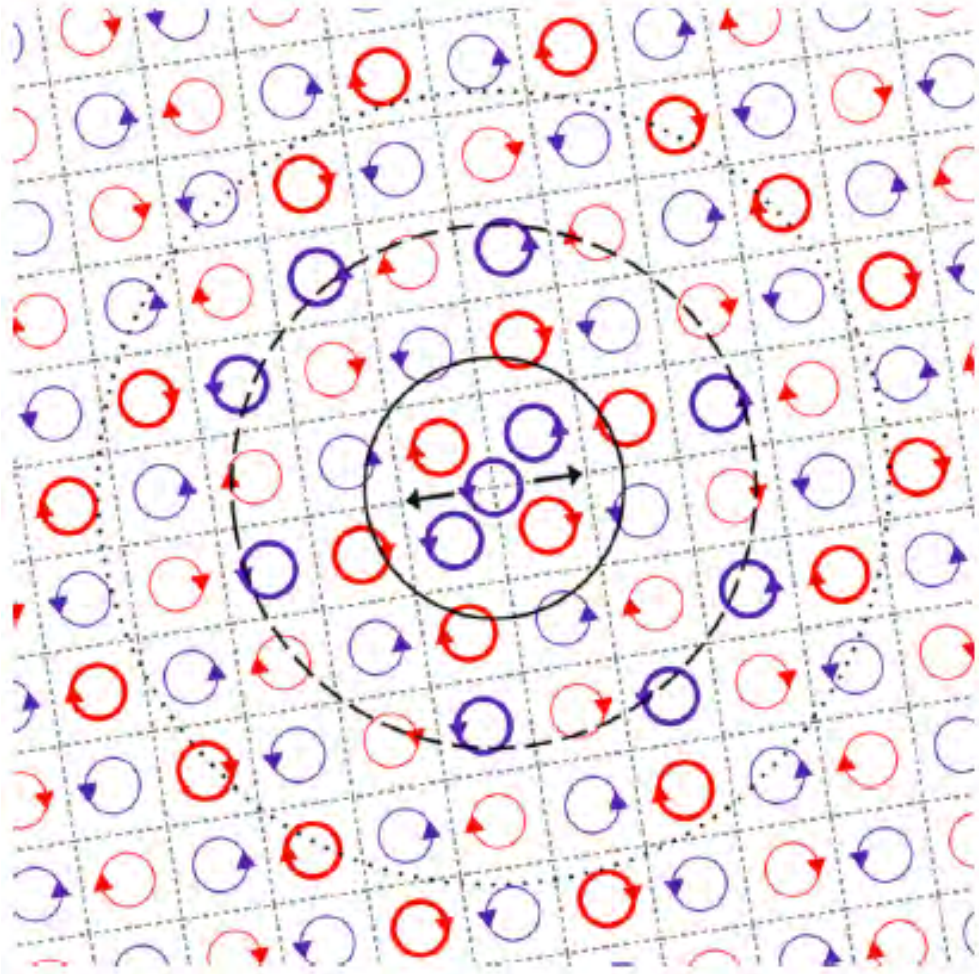
Lene B. Oddershede (NBI, exp)

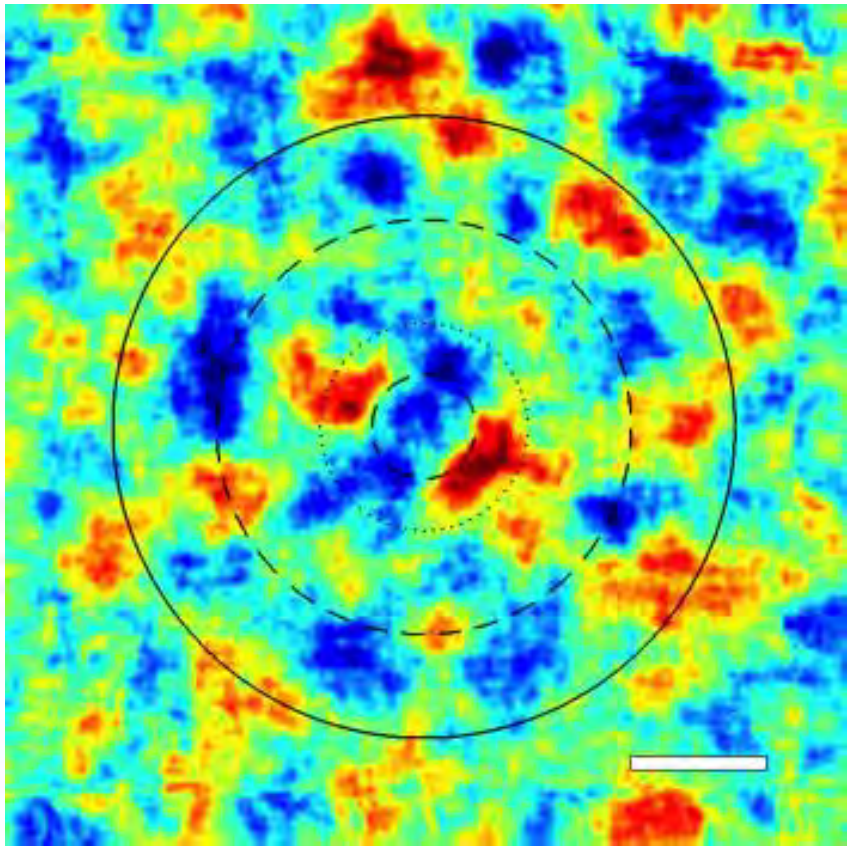
Joachim Mathiesen (NBI, theory)

Jens Tarp (graduate student, NBI, theory)

Good advice from:
Julia M. Yeomans, University of Oxford

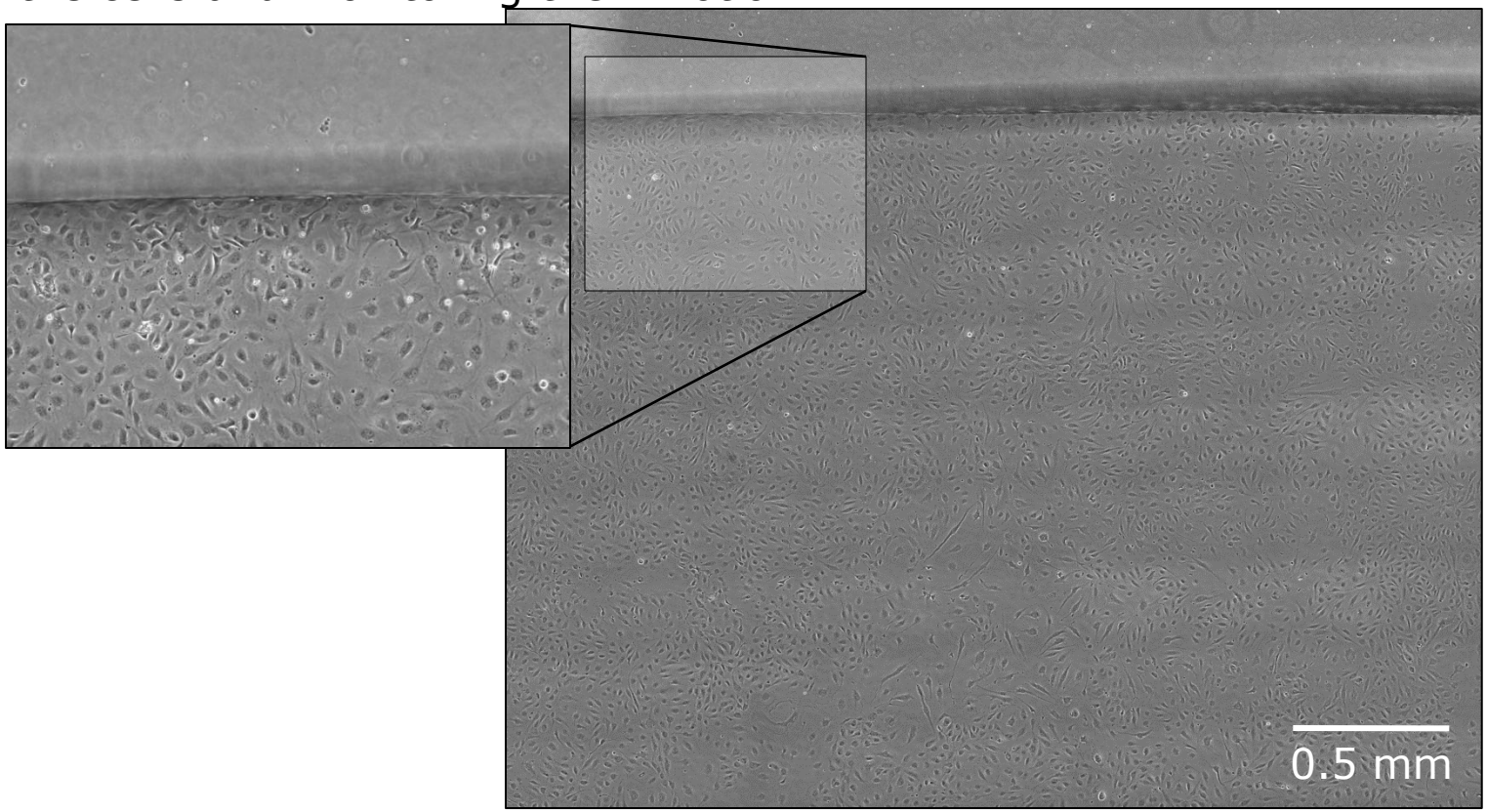
Schematic of the vorticity pattern





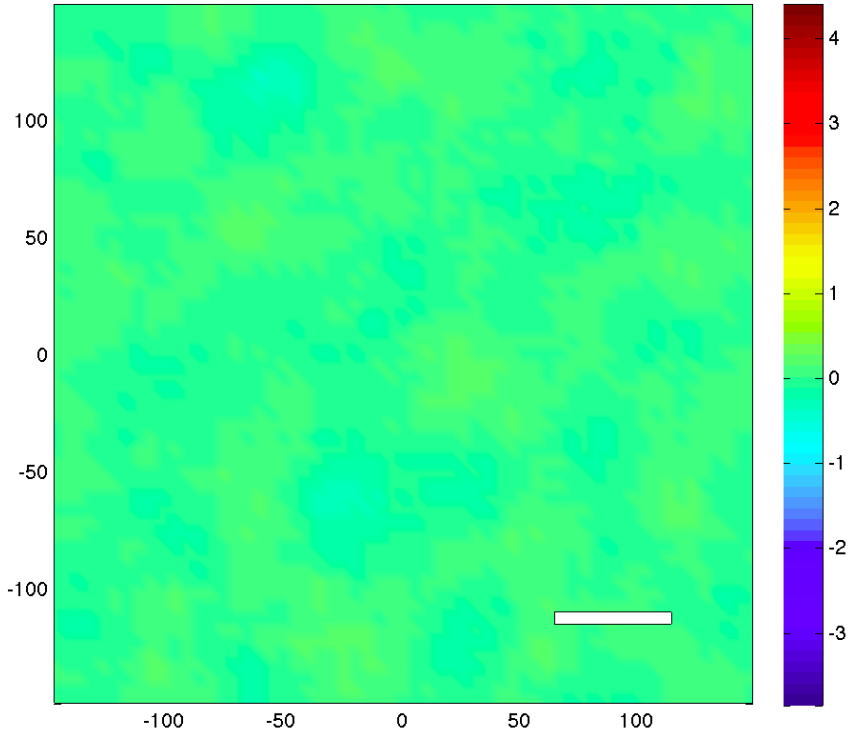
2-D Model of Angiogenesis

Endothelial cells migrating into the surrounding tissue (creating new blood vessels) are modelled by removing a barrier that constrains the cells and monitoring their motion.

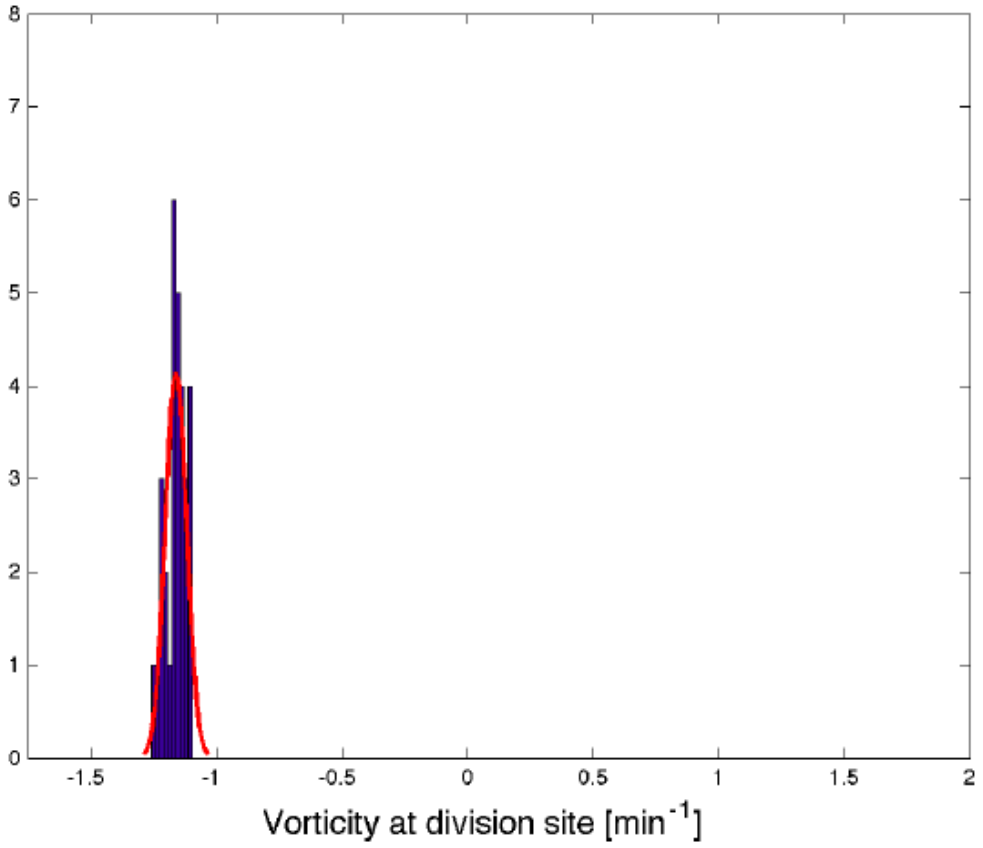


The cells can be monitored every 10 minutes for 48 hours

Vorticity, average of 30 non-dividing control samples

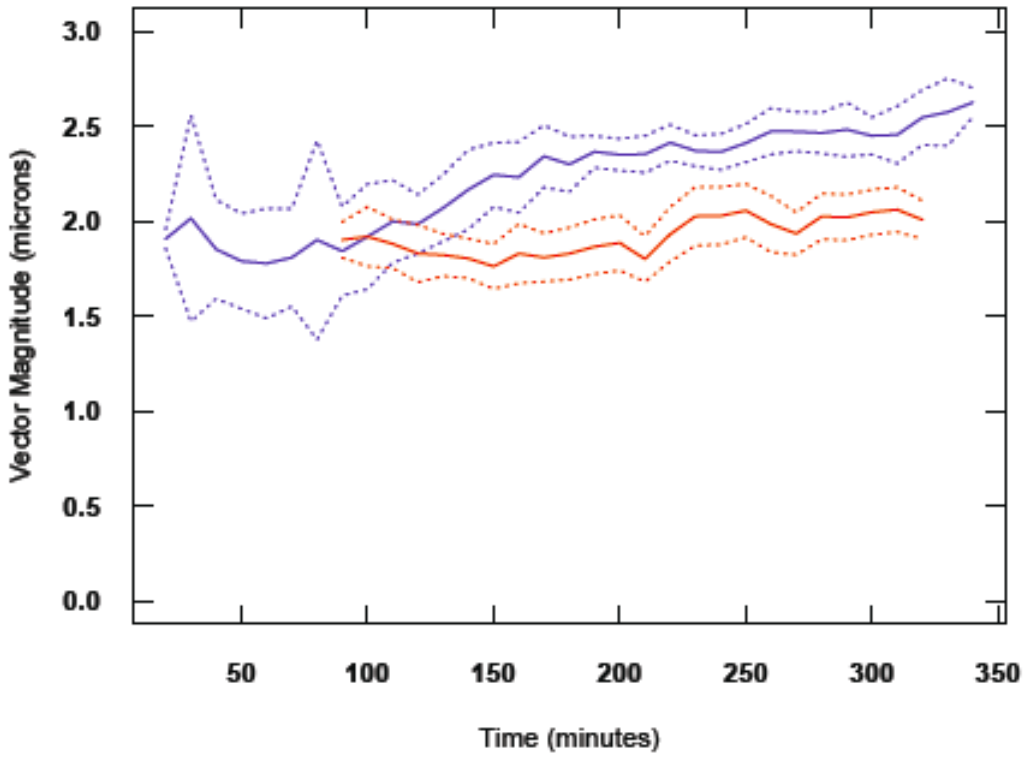


Vorticity at the center



Average vorticity: -1.16 ±0.04

Endothelial cells speed up after division

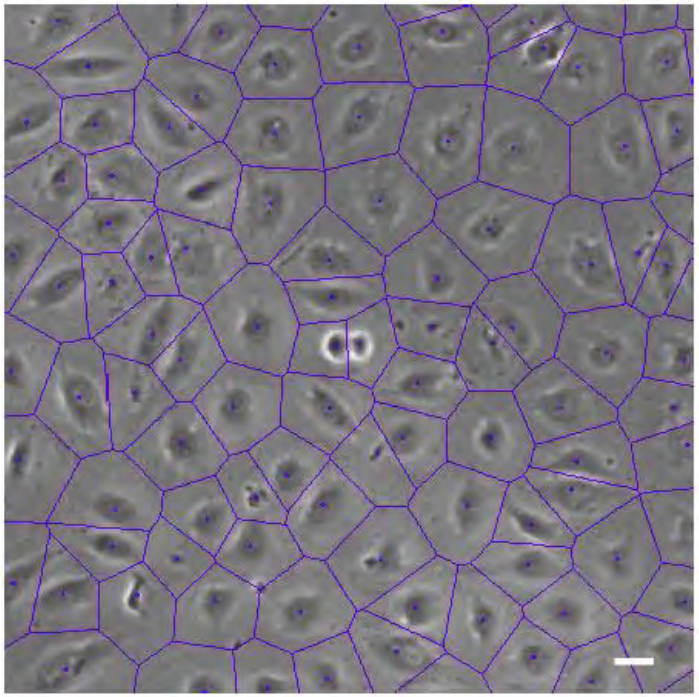


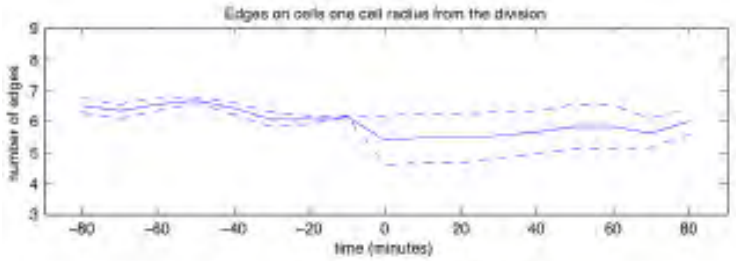
Blue: speed of dividing cells (division at 0 min)
Red: speed of non-dividing control cells

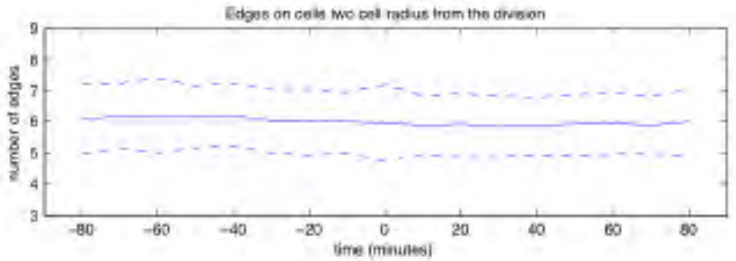
Voronoi analysis

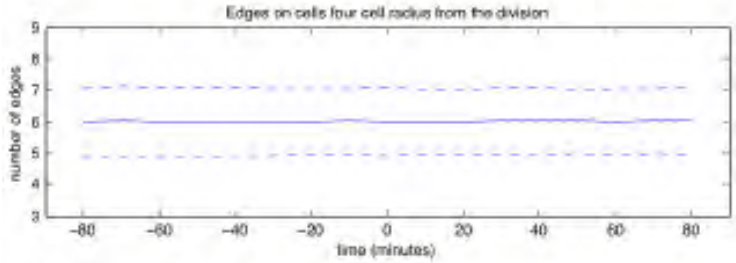
Average number of neighbours for non-dividing cells is 6
- hexagonal packaging.

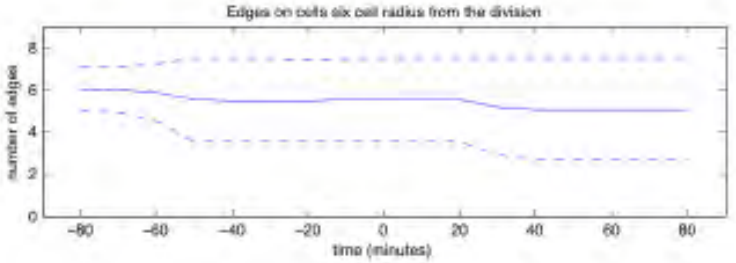
During division, the number of neighbours of the dividing cell decreases to ~5.





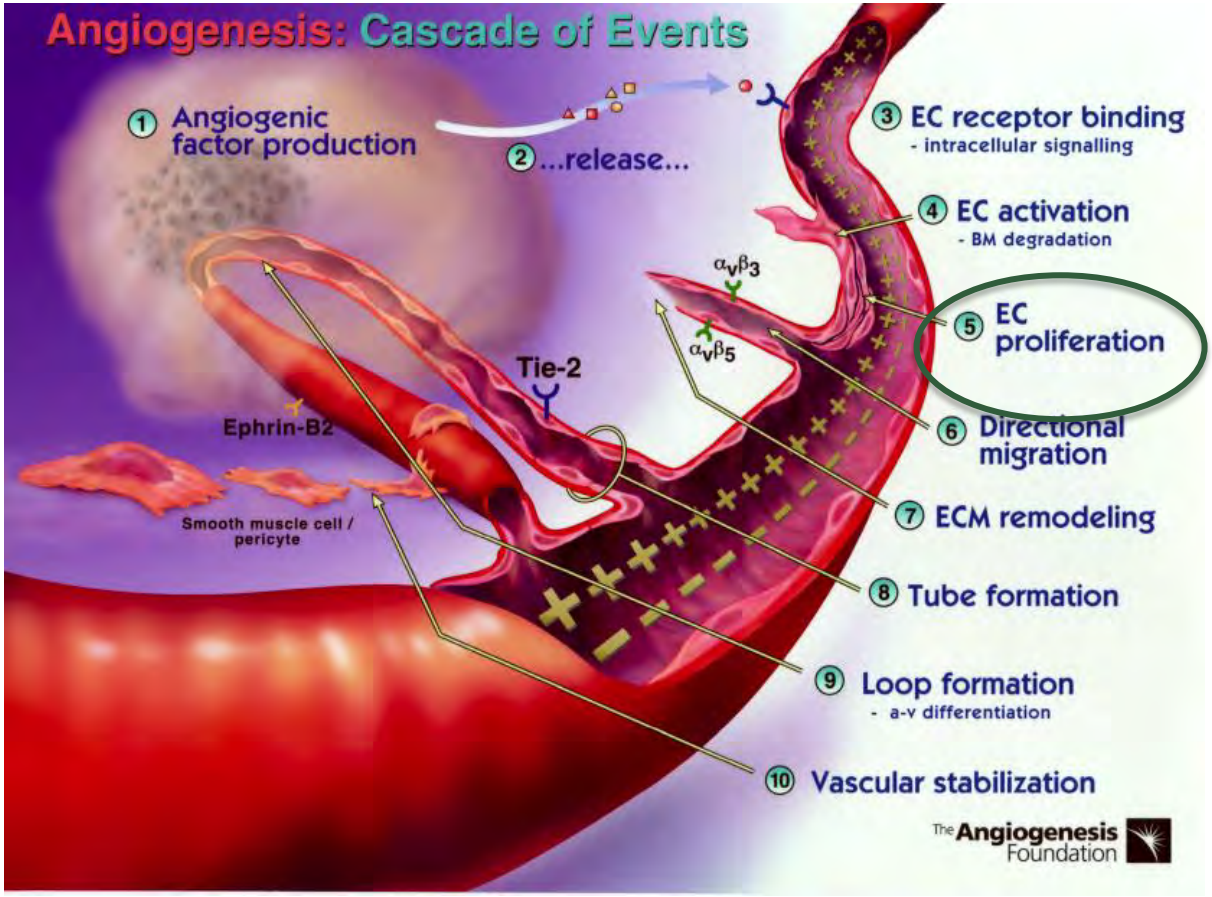






The Biological System

Endothelial cells line the vessels of the circulatory blood system. In healthy tissue, cell division rate is relatively low, only sufficient to replace apoptotic cells.



Divisions in the monolayer, divides every 18-24 hours -- in vivo under flows much longer, days.

The divisions appear uniformly distributed spatially.