Ropewalker cells: architecture and migration of an epithelium on a cylindrical wire

H.G. Yevick, G. Duclos, I. Bonnet, P. Silberzan

Supplementary Figures

Supplementary Figure 1 : Scanning Electron Micrographs of the glass wires. Wires were imaged after gold metallization and had a very smooth surface (defect size is typically less than 0.1µm). A/ manually pulled wire. B/ Commercial wire. C/ wire obtained using a pipet puller.

Supplementary Figure 2: For small radii (R < 5 µm), there is room for one cell only around the wire. A) The leading cells can take very elongated shapes (xy projection). B) These cells fully wrap up thin wires (xz projection)

Supplementary Figure 3: A) Cross-sections through the glass wire of a monolayer wrapped around its circumference demonstrating that the apical side of the cells points outward, towards the medium Red: Ezrin, Blue: Nucleus, Green: F-Actin.

Supplementary Figure 4: Actin organizes in fibers at the basal plane, in contact with the wire. Images were acquired in confocal microscopy. Images at the right are separated by 2 µm. Fixed cells, actin green, nuclei blue.

Supplementary Figure 5: Rose plots showing the orientation according to the fiber direction of the basal actin fibers on fibers of radius 25 µm (left) and 85 µm (right). Plots are symmetrized (N > 2500 per radius)

Supplementary Figure 6 : A) MDCK plated on uncoated glass wires exhibit the same circumferential actin organization as on fibronectin-coated wires. B) RPE1 epithelial cells show the same circumferential actin organization as MDCK cells (fibronectin-coated wire). C) NIH-3T3 fibroblasts show a longitudinal orientation of their actin cytoskeleton (fibronectin-coated wire). Bars are 10 µm.

Supplementary Figure 7 : On flat surfaces, stress fibers are anchored to the surface by two well-developed focal adhesions at their extremities. Substrate: Fibronectin–coated glass. Actin (red), vinculin (green).

Supplementary Figure 8: Actomyosin cable at the front edge of the migrating monolayer on a wire (R = 28 µm). Main panel is F-actin. Inset : F-actin (red) colocalises with myosin (green) at the cable.

Supplementary Figure 9: Kymographs illustrating the different behaviors according to the wire radius. A) R = 20 µm: the movement is regular and defines a velocity of the order of 25 µm h^{-1} in this particular case. B) For smaller radii, (R = 4 µm in this particular case), the movements of the cells are much more erratic (going forward and backward) and exhibit strong fluctuations.

Supplementary Figure 10: A) When plated on tapered wires, the cells reach a point where the radius of the wire is too small for them to progress. They stop their migration (arrow) but continue to send out protrusions (inset). B) Sequence of the collective movement of the cells (time interval = 14h) eventually leading to the formation of an empty cyst (triangle).

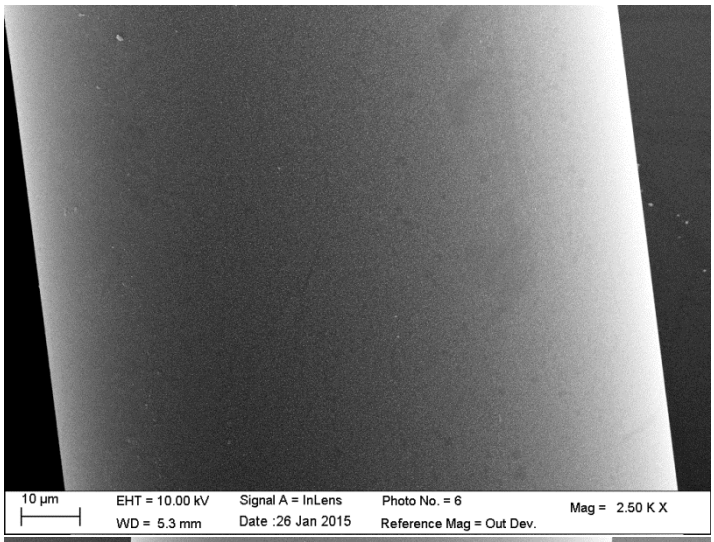
Supplementary Figure 11 Front edge velocity in presence of blebbistatin (myosin II inhibitor) or NSC23766 (Rac1 inhibitor) for two different radii. Note the different impacts of the two drugs depending on the wire radius, demonstrating the switch in migration modes. $13 < N < 75$ per box. Error bars are SDs.

Supplementary Figure 12: Inhibiting cell division with mitomycin C drastically slowed down the migration in the bulk of the monolayer while hardly affecting the front velocity. The white dashed line represents the addition of mitomycin. Note the change of slope corresponding to a decrease in velocity in the monolayer (upper left part of the kymograph) while the front keeps the same velocity. R = 40 µm.

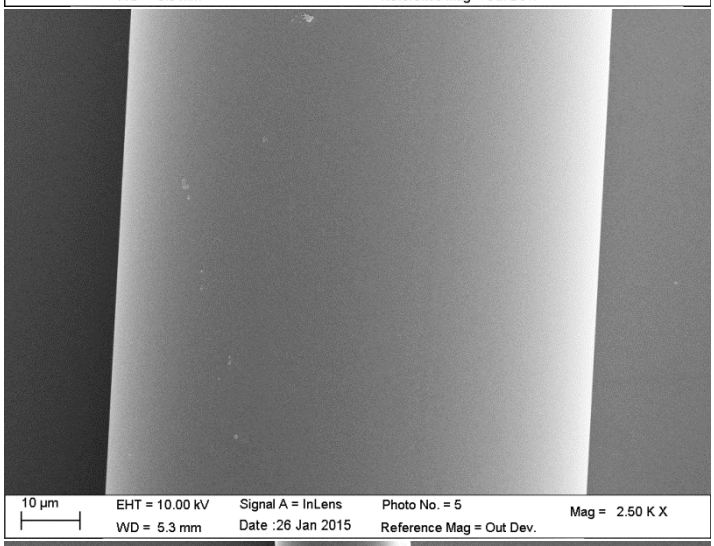
Supplementary Figure 13: Promoting a more individual behavior with HGF at sub-scattering concentration sets an upper limit for the collective migration that does not depend on the wires' radius.

Supplementary Figure 1

A/



B/



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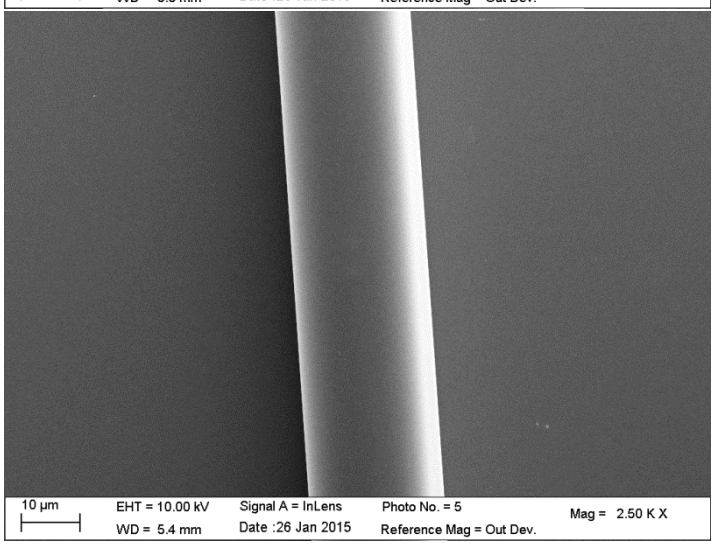
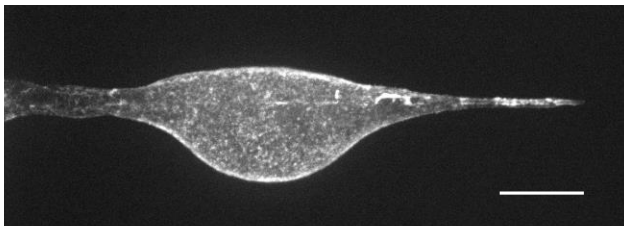


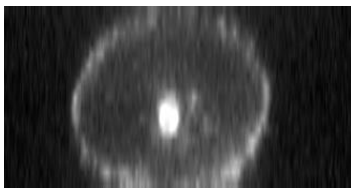
Supplementary Figure 2

A



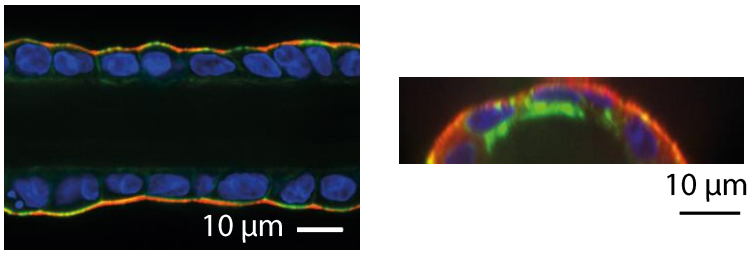
20 µm

B



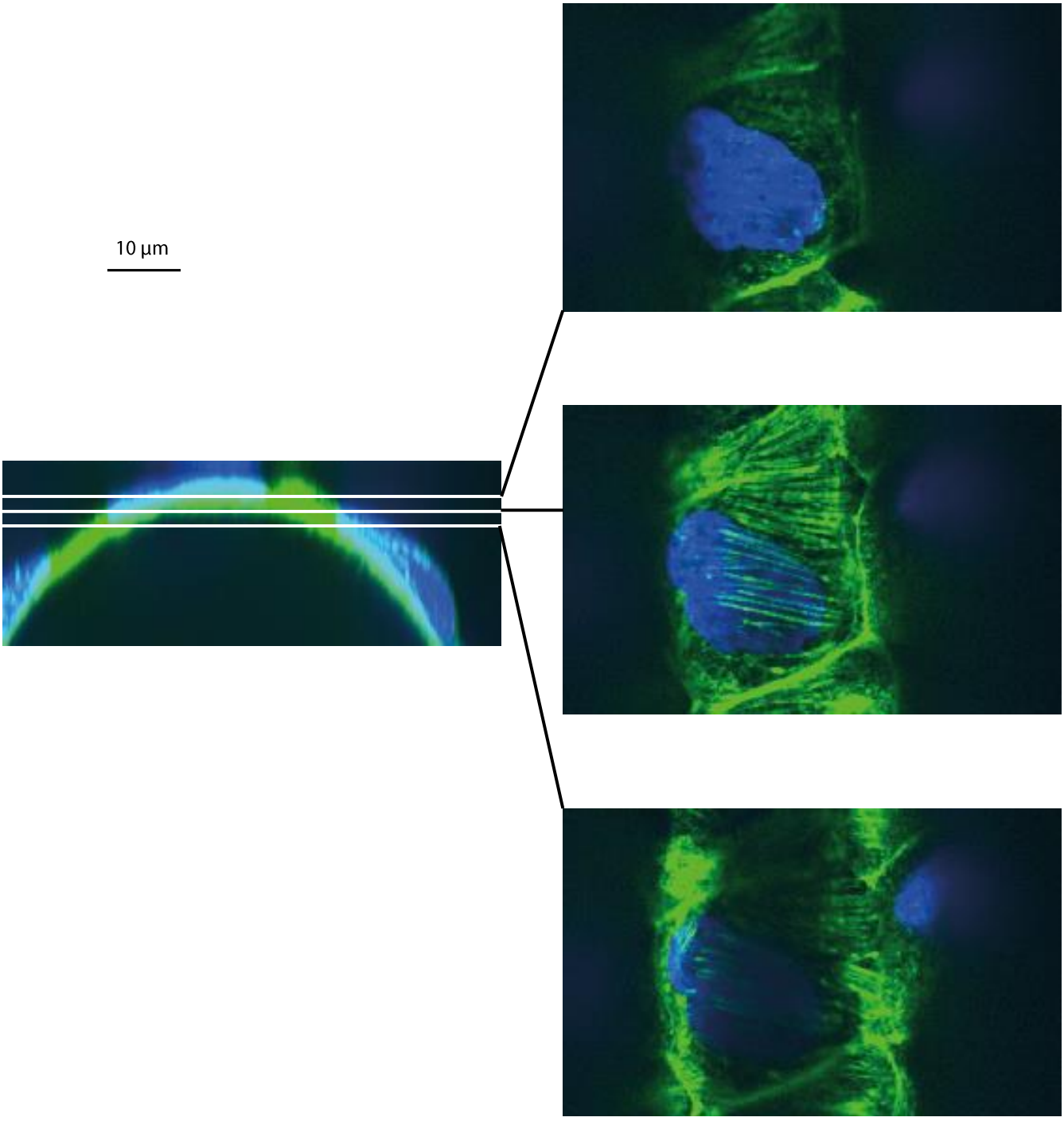
10 µm

Supplementary Figure 3

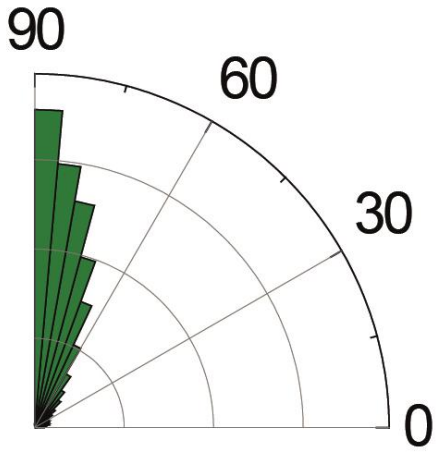


Ezrin F-Actin DAPI

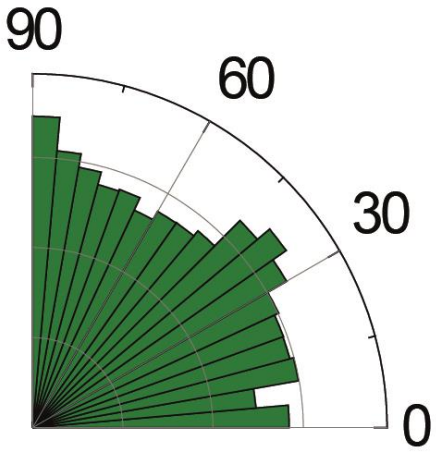
Supplementary Figure 4



Supplementary Figure 5

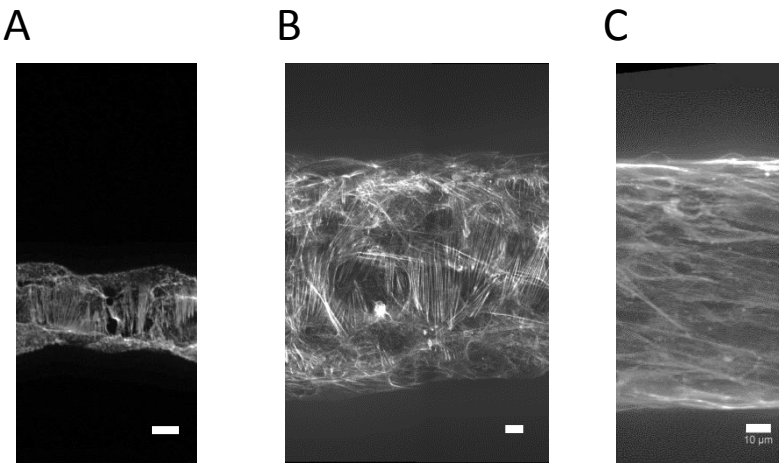


R = 25 µm



R = 85 µm

Supplementary Figure 6



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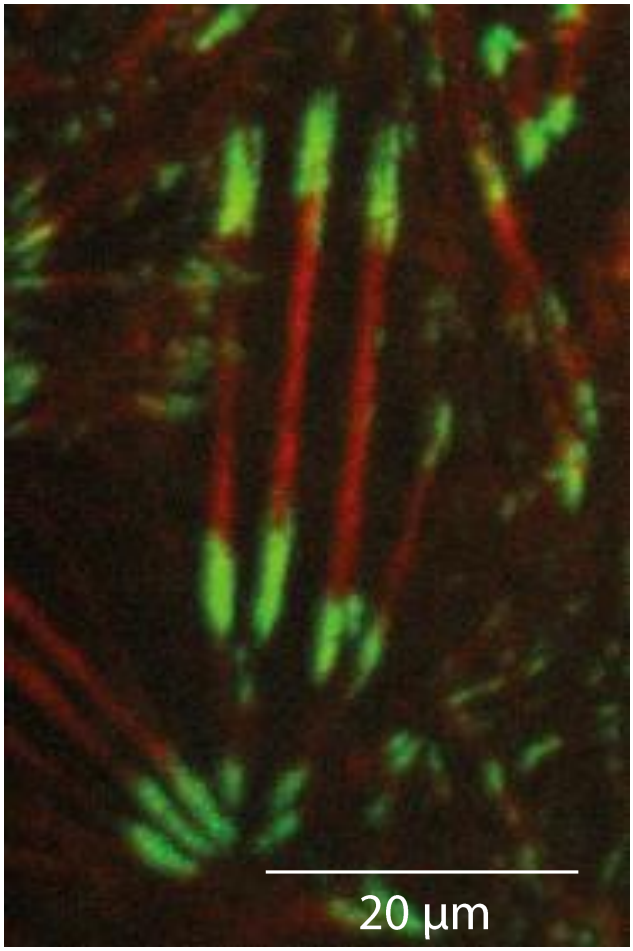

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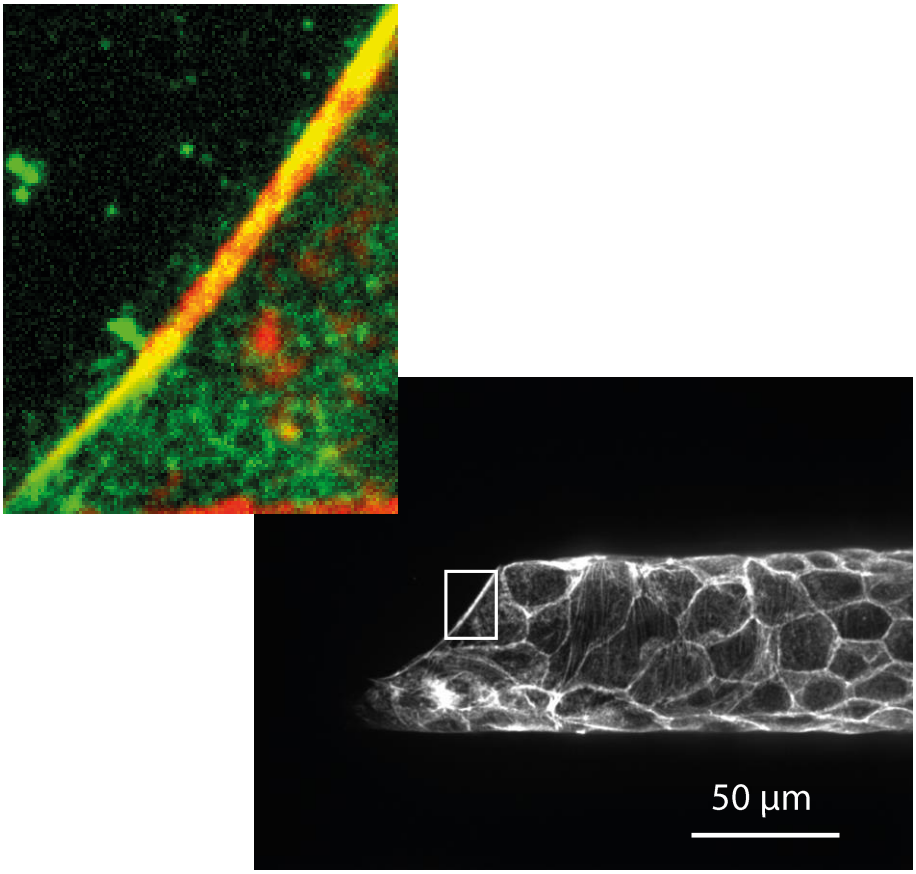
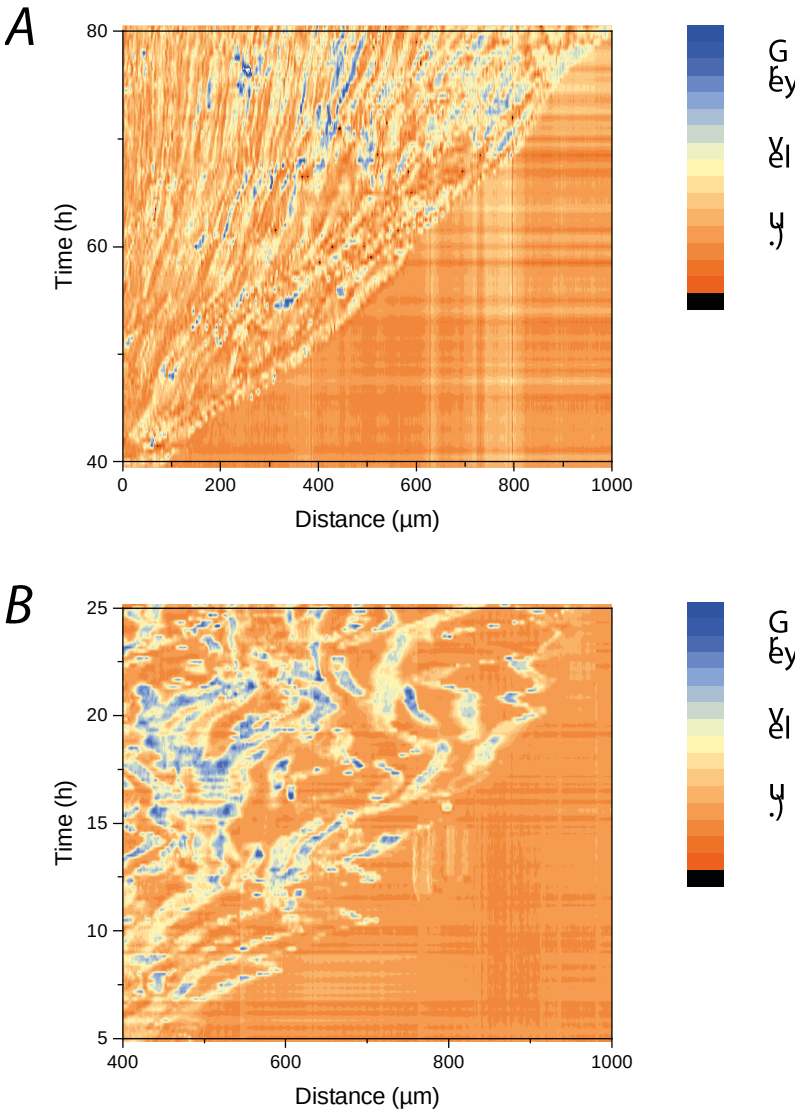
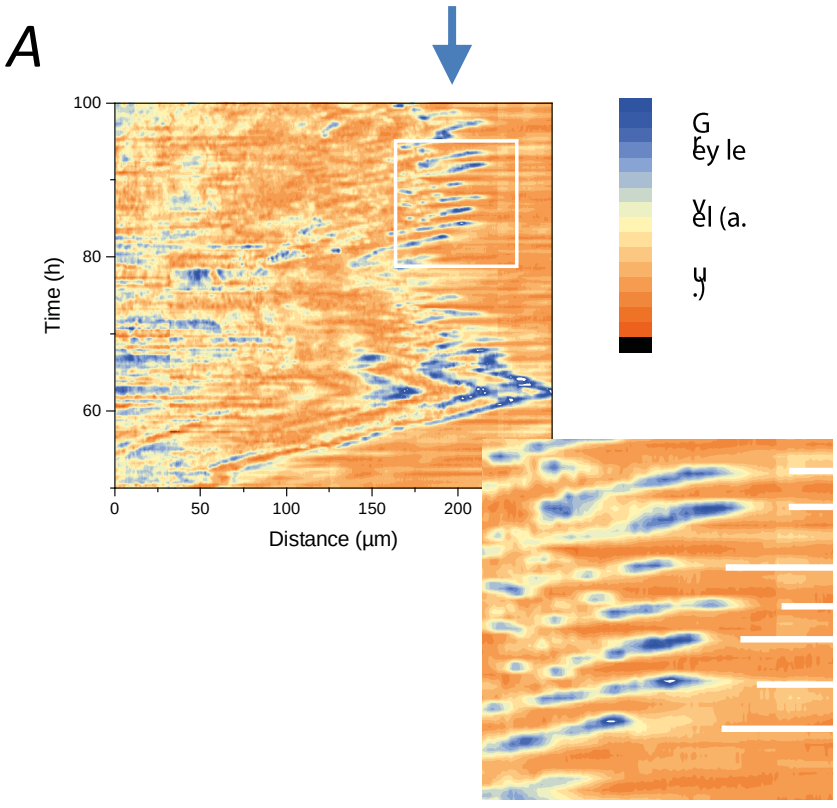
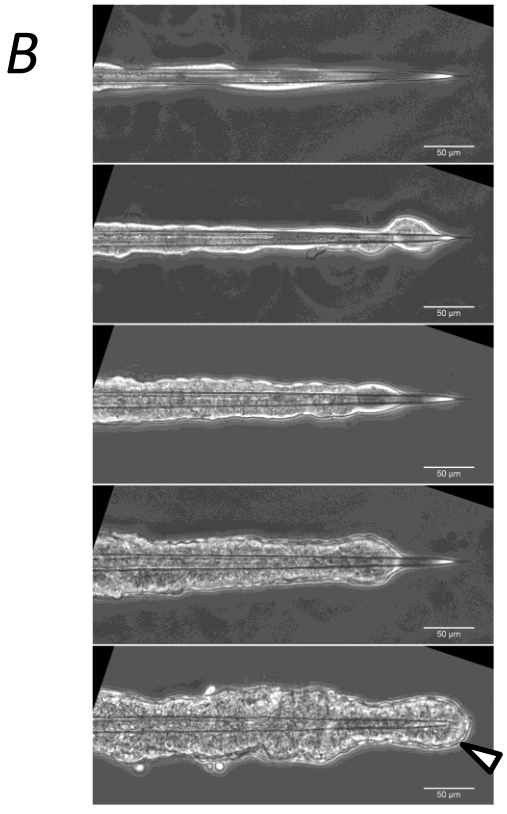


Supplementary Figure 9

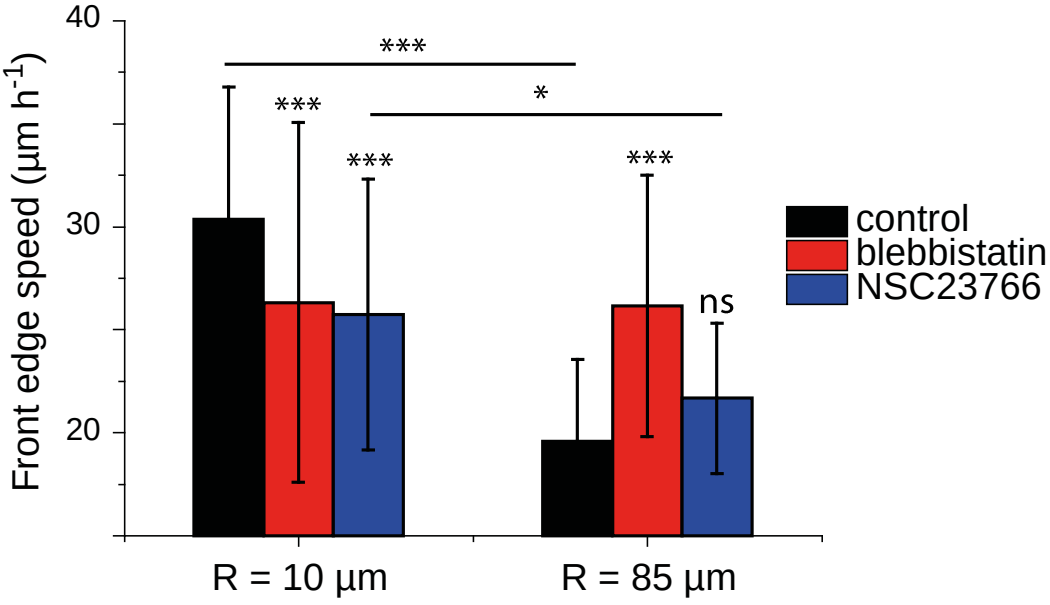


Supplementary Figure 10

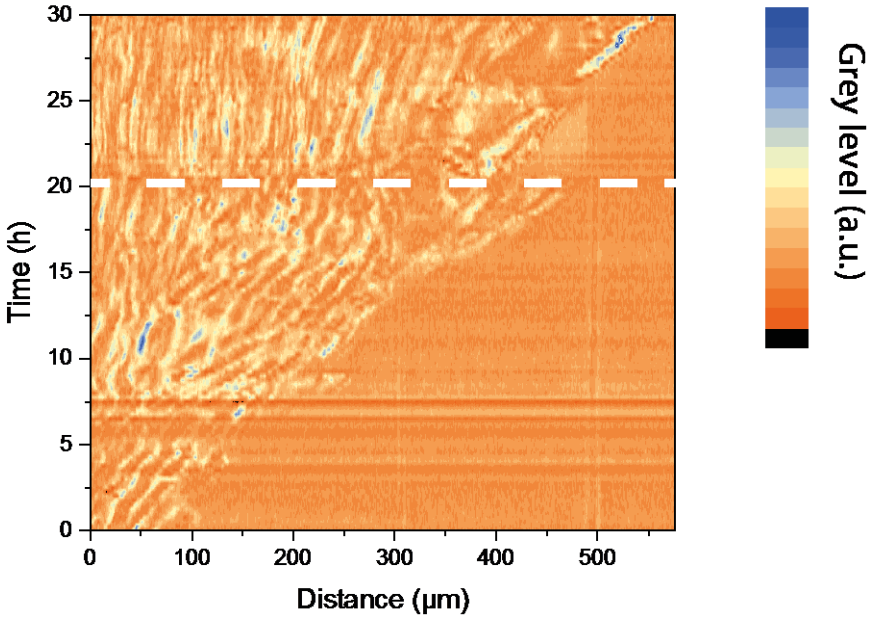




Supplementary Figure 11



Supplementary Figure 12



Supplementary Figure 13