Electronic Supplementary Material (ESI) for Soft Matter.

Electronic Supplementary Information

Disentangling collective motion and local rearrangements in 2D and 3D cell assemblies

Roberto Cerbino,[a] Stefano Villa,[a] Andrea Palamidessi,[b] Emanuela Frittoli,[b] Giorgio Scita,[bc] and Fabio Giavazzi[a]

a *Università degli Studi di Milano, Dipartimento di Biotecnologie Mediche e Medicina Traslazionale, 20090 Segrate, Italy.*
b *IFOM-FIRC Institute of Molecular Oncology, Milano, Italy*
c *Università degli Studi di Milano, Dipartimento di Oncologia e Emato-Oncologia, Milano, Italy*

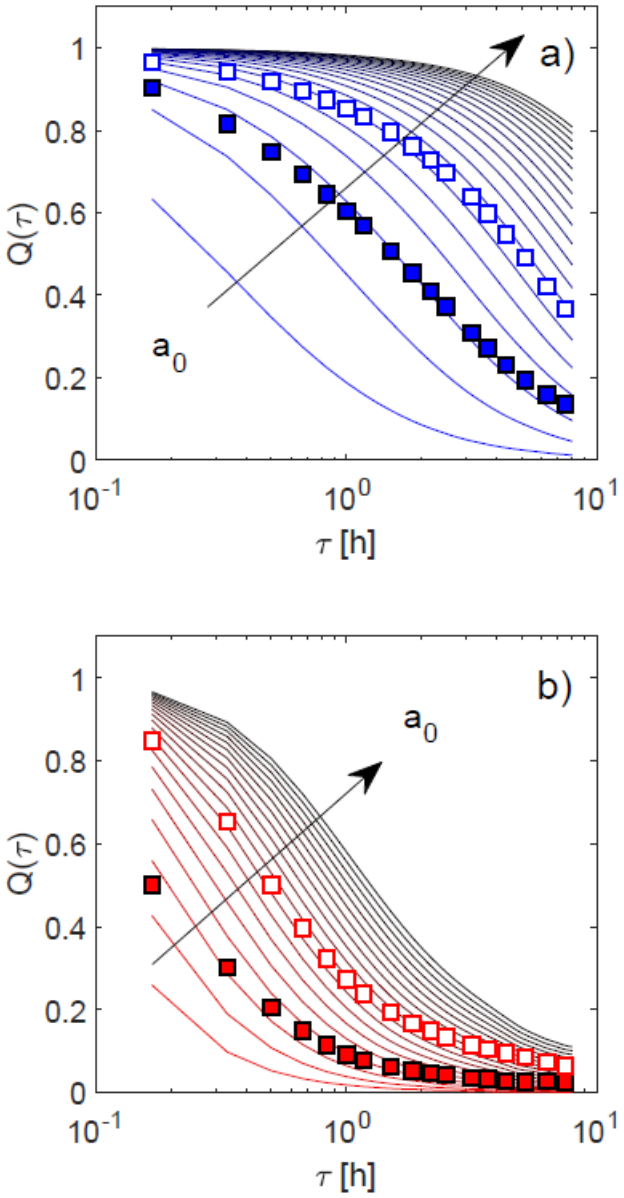

Figure S1: Overlap parameter $Q_{PT}(\tau)$ obtained from particle tracking analysis of fluorescence images acquired in the 2D cellular unjamming experiment on CTRL (panel a) and RAB5A-overpressing (panel b) MCF10A monolayers, respectively. In each pane, the continuous curves are obtained for different values of the overlap lengthscale $a_0 = 0.1R, 0.2R, \dots, 1.9R, 2R$, where $R = 4.7\ \mu m$ is the average nuclear radius. For CTRL (RAB5A-overexpressing) monolayers about 30% (3%) of the nuclei are tracked for the whole duration of the experiments and thus contribute to the calculation of $Q_{PT}(\tau)$. Filled and empty symbols in each panel corresponds to the overlap parameter obtained for the same experiment from DVA, performed on phase contrast and fluorescence images, respectively.

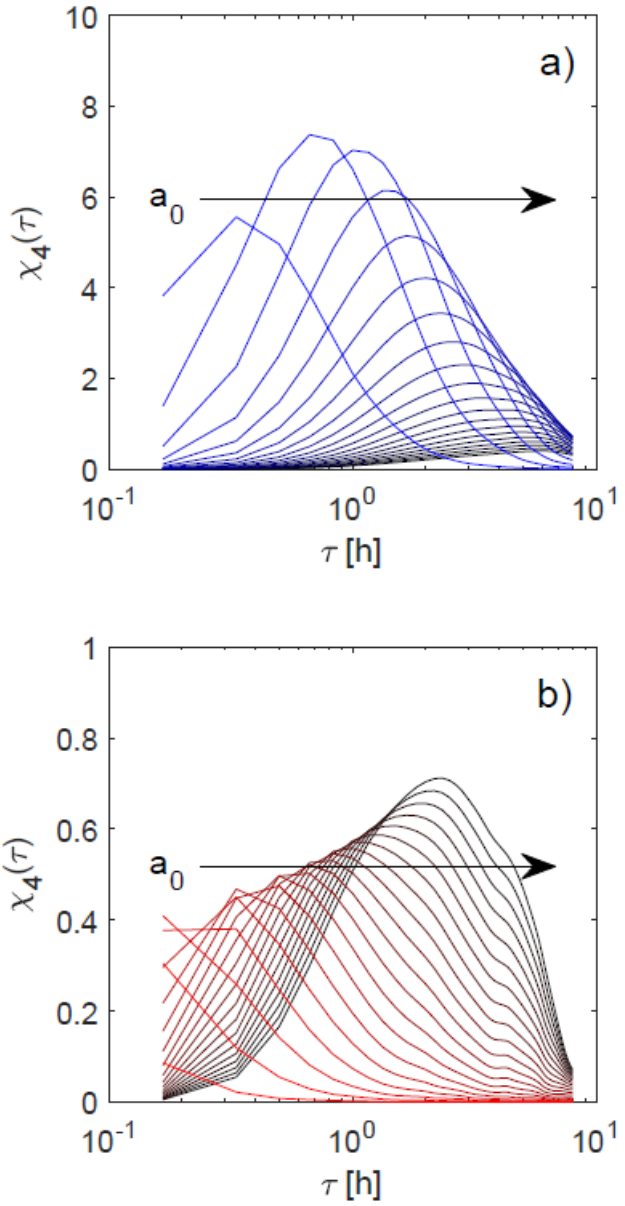

Figure S2: Dynamical susceptibility $\chi_{4,PT}(\tau)$ obtained from particle tracking analysis of the 2D cellular unjamming experiment on CTRL (panel a) and RAB5A-overexpressing (panel b) MCF10A monolayers, respectively. In each panel we show curves obtained for different values of the overlap lengthscale $a_0 = 0.1R, 0.2R, \dots, 1.9R, 2R$, where $R = 4.7\ \mu m$ is the average nuclear radius. For CTRL (RAB5A-overexpressing) monolayers about 30% (3%) of the nuclei are tracked for the whole duration of the experiments and thus contribute to the calculation of $\chi_{4,PT}(\tau)$.

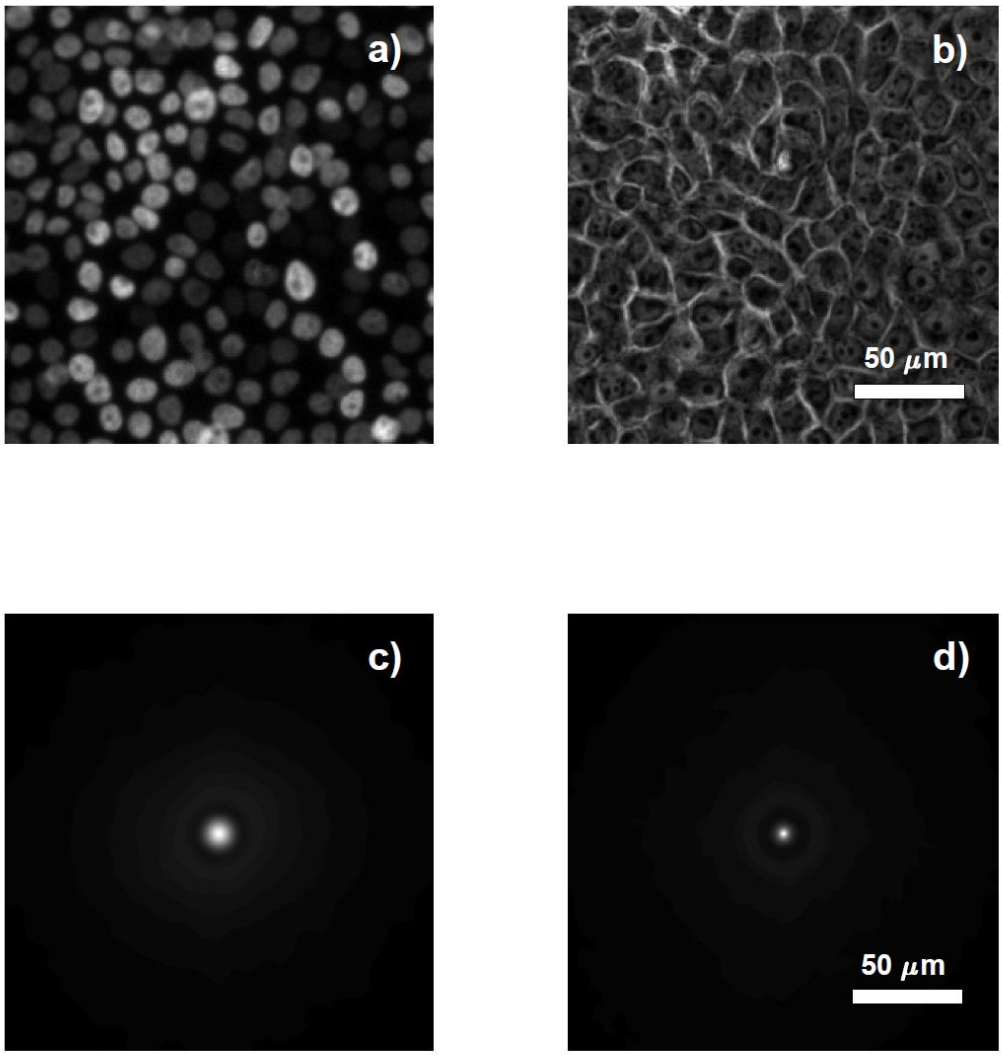

Figure S3: Representative ROIs of a confluent MCF10A monolayer imaged in fluorescence microscopy (a) and phase contrast microscopy (b), respectively. Two-dimensional self-correlation functions $C_I(r) = \langle I(\boldsymbol{x}_0 + \boldsymbol{r}, t) I(\boldsymbol{x}_0, t) \rangle_{\boldsymbol{x}_0, t}$ (averaged 5 independent fields of view) of the image intensity for fluorescence microscopy (a) and phase contrast microscopy (b) image acquisitions, respectively.

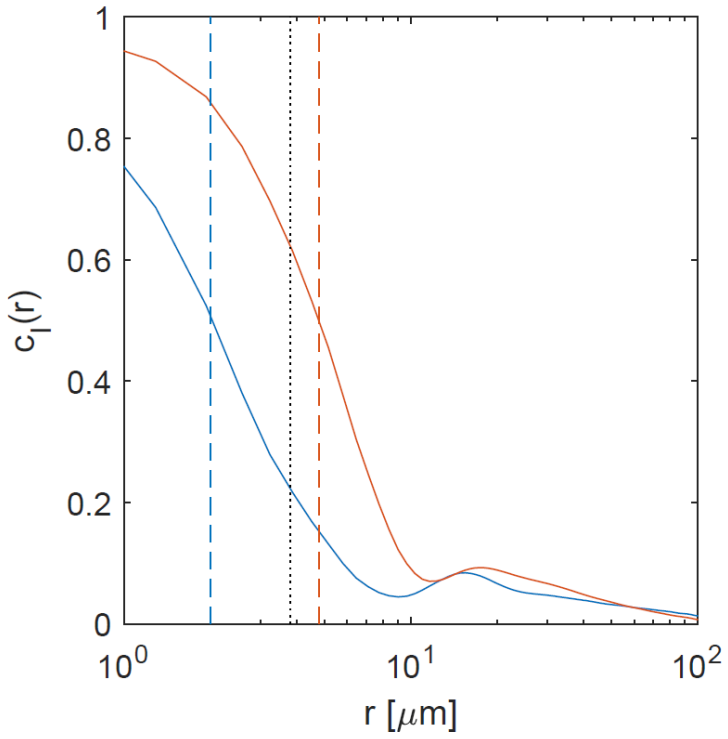

Figure S4: Azimuthally averaged normalized self-correlation functions $c_I(r) = C_I(r)/C_I(0)$ of the image intensity distribution for fluorescence (orange continuous line) and phase contrast (blue continuous line) images, respectively. The orange (blue) vertical dashed line corresponds to the half width at half maximum $HWHM = 4.8\ \mu m$ ($HWHM = 2.0\ \mu m$) of the corresponding self-correlation function. The black dotted line corresponds to the calculated HWHM of the self-correlation functions for a uniform disk of radius $4.7\ \mu m$, corresponding to the mean nuclear radius.

Supplementary Movies:

SM01: Time-lapse phase contrast microscopy acquisition of a confluent MCF-10A monolayer approaching kinetic arrest. Image size: 380 µm x 380 µm, duration: 82 h.

SM01_reg: Same as in SM01, after rigid registration. Image size: 290 µm x 140 µm, duration: 82 h.

SM02_CTRL: Time-lapse acquisition of CTRL confluent H2B-GFP MCF-10A epithelial cells. Upper panel: fluorescence microscopy, lower panel: phase contrast microscopy. Image size (each panel): 644 µm x 644 µm, duration: 16.6 h.

SM02_CTRL_reg: Same as in SM02_CTRL, after rigid registration. Image size (each panel): 360 µm x 260 µm, duration: 16.6 h.

SM02_RAB5: Time-lapse acquisition of RAB5A-overexpressing confluent H2B-GFP MCF-10A epithelial cells. Upper panel: fluorescence microscopy, lower panel: phase contrast microscopy. Image size (each panel): 644 µm x 644 µm, duration: 16.6 h.

SM02_RAB5_reg: Same as in SM02_RAB5, after rigid registration. Image size (each panel): 360 µm x 260 µm, duration: 16.6 h.

SM03_CTRL: Time-lapse confocal microscopy acquisition of a CTRL spheroid of mCherry-H2B MCF10DCIS.com cells. A single plane orthogonal to the optical axis and passing through the center of the spheroid is shown. Image size: 290 µm x 290 µm, duration: 24 h.

SM03_CTRL: Time-lapse confocal microscopy acquisition of a RAB5A-overexpressing spheroid of mCherry-H2B MCF10DCIS.com cells. A single plane orthogonal to the optical axis and passing through the center of the spheroid is shown. Image size: 290 µm x 290 µm, duration: 24 h.