Electronic Supplementary Material (ESI) for Soft Matter.

Supporting Information

Paper: Local and Transient Permeation Events are Associated with Local Melting of Giant Liposomes

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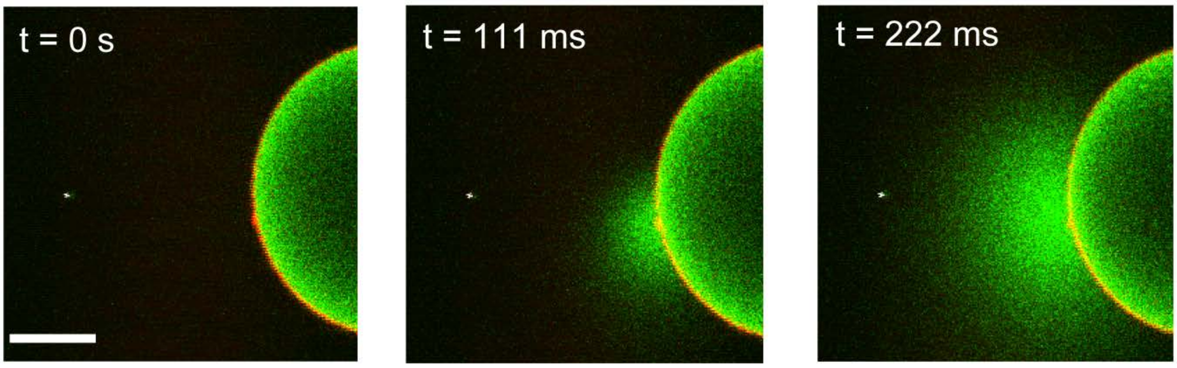

Figure S1 Example of a burst shown by 3 consecutive images. Membrane is labeled red using Texas red DHPE and calcein is green. The trapped 80 nm AuNP (white dot) is imaged by reflection microscopy. Upon leakage the calcein dilutes and de-quenches and results in a burst of intensity. Scale bar 10 μm.

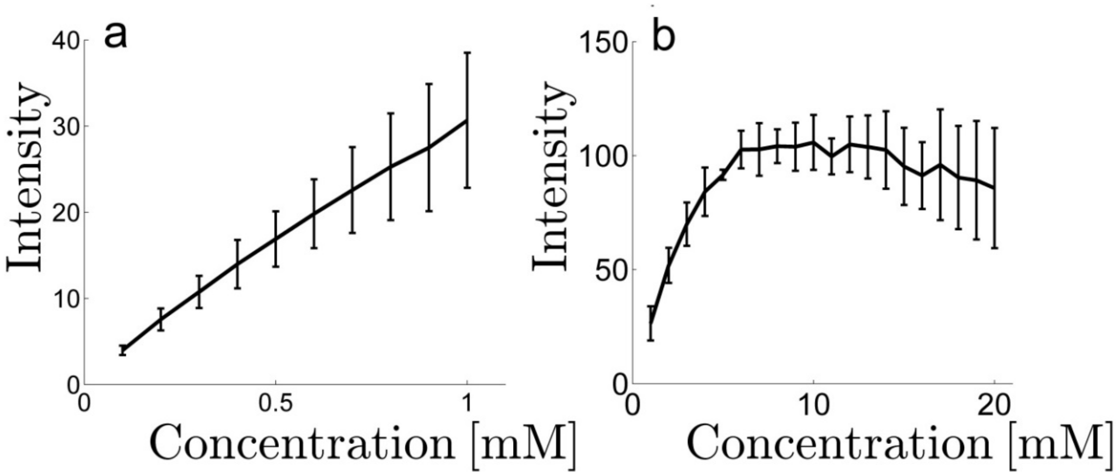

Figure S2 Self quenching of calcein versus concentration measured in a microscope chamber using confocal detection. (a) The intensity increases approximately linearly up to 1 mM. (b) To avoid saturation in the image we changed the imaging settings for calcein concentrations higher than 1 mM and continued to measure intensities for concentrations up to 20 mM. The intensity versus concentration curve starts to become increasingly nonlinear as the concentration exceeds 1 mM. The data represent 20

independent experiments. All intensities are collected at the same height relative to the bottom glass surface corresponding to the z-position where the intensity was highest.

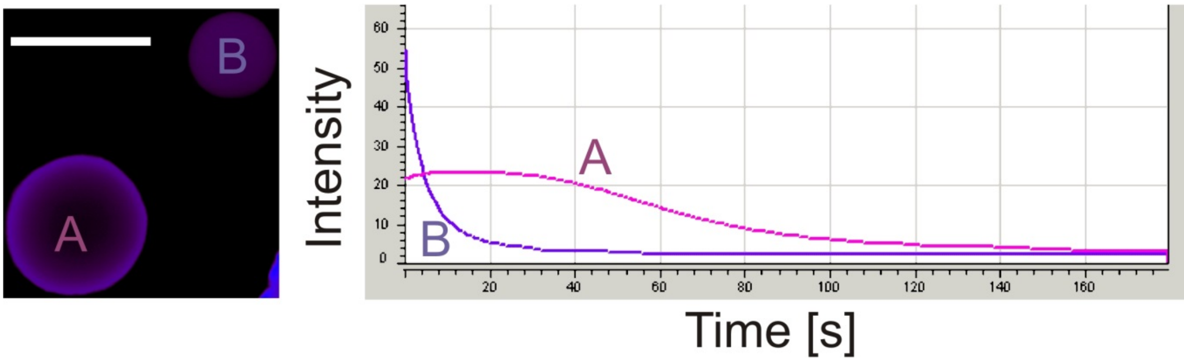

Figure S3 Calcein quenching control. Two GUVs, denoted by A and B respectively, containing different concentrations of calcein clearly indicated by the different appearance of the interior signal. GUVs containing highly quenched calcein have a darker region near the center (GUV A) whereas GUVs containing un-quenched calcein have uniform lumen intensity (GUV B). Graphs show resulting bleaching curves of the two GUVs in the image. The bleaching of the GUV containing quenched calcein results in an increase followed by a decrease in the measured intensity, red curve. Bleaching of GUV B, results in a typical exponentially decaying bleaching, blue curve. Scale bar is 35 µm.

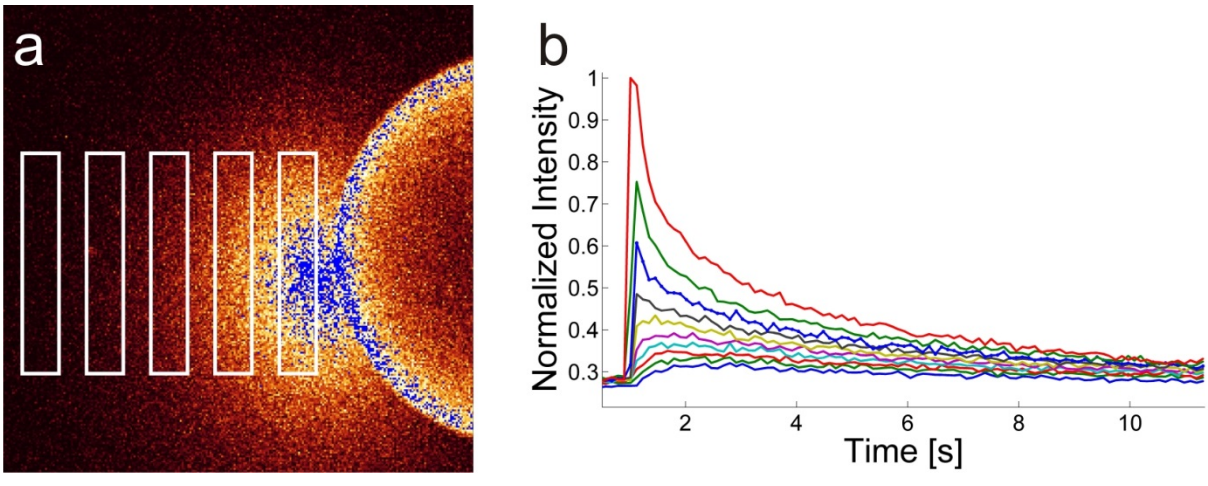

Figure S4 Quantification of intensity inside ROIs placed near the GUV membrane. (a) The ROI was placed a distance of r_{ROI} = 5 µm away from the nearest point on the membrane where the release of calcein could be observed. (b) The effect of placing a ROI at different distances from the GUV (as depicted in (a)), results in Intensity curves having different localization of the intensity peak. The successive curves represent the integrated intensity in ROIs placed 10 pixels apart as depicted in (a).

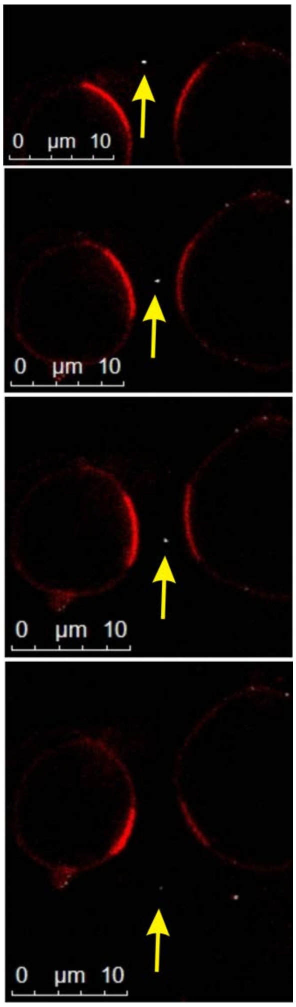

Figure S5 Local melting of two GUVs using an optically trapped gold nanoparticle as a nanoscopic heat generator. The AuNP is trapped in the space between two GUVs and the proximal membrane regions facing the nanoparticle undergo a gel to fluid phase transition as shown in Move 1. The fluid regions exhibit a stronger fluorescent intensity due to increased intercalation of the lipophilic fluorophore, di-4-ANEPPDHQ, into the fluid part of the membrane.[24] The trapped AuNP (indicated by the yellow arrow) is translated through the gap between the GUVs to demonstrate how the location of the locally melted regions depends on the position of the trapped particle.

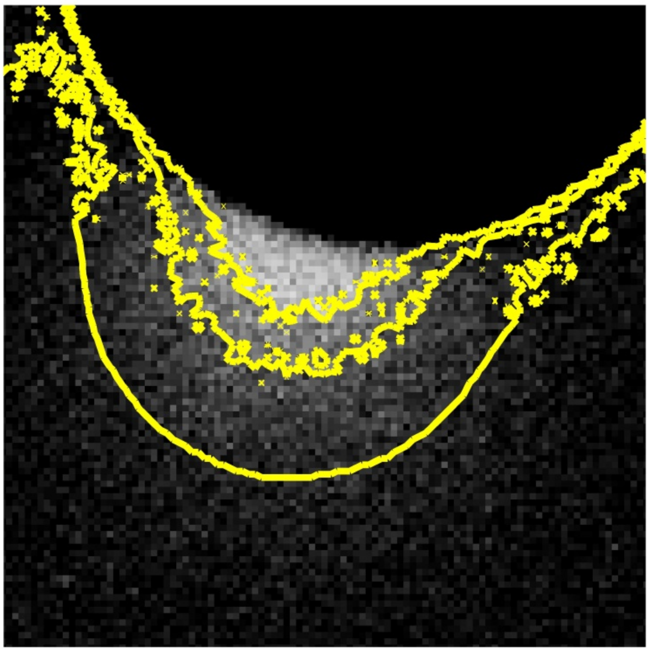

Figure S6 Contour plot of a typical burst showing concentric half-circles of constant intensity which are centered near the membrane. The intensity from the interior of the GUV has been removed by subtracting with the image just prior to the release.

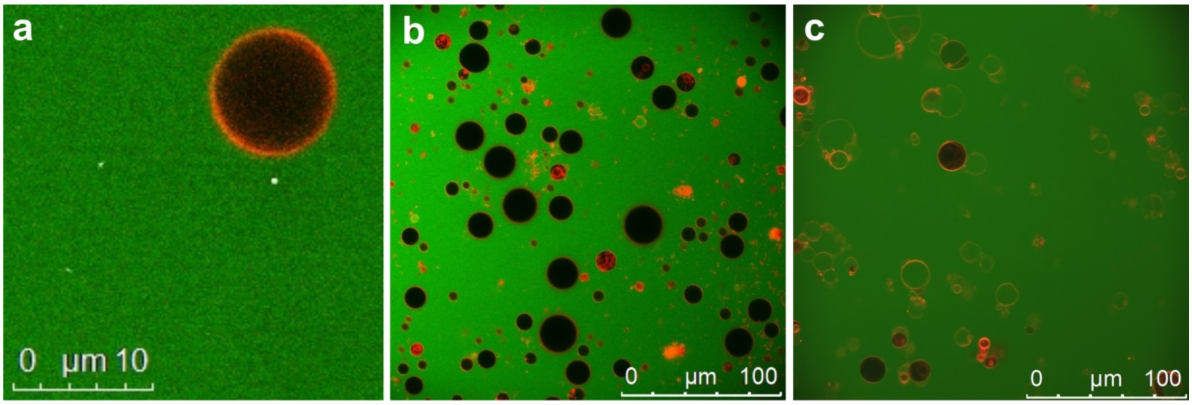

Figure S7 Permeability controls of GUVs. (a) Control with DOPC and optically trapped AuNP. An 80 nm AuNP is optically trapped and brought into vicinity of a GUV made of DOPC lipids (T_m = -17 °C) and 0.3% TR-DHPE. 1 mM (non-quenched) calcein is mixed with the solution surrounding the GUV. No calcein influx was measured after moving the AuNP close to the membrane (see Movie 2). The optical trapping power was 450 mW. (b) Control showing that fluid phase membranes exhibit minimal porosity. Calcein leakage through fluid phase DOPC vesicles after incubation for 30 min was minimal. The concentration in this experiment is 1 mM calcein outside the vesicles. (c) $DC_{15}PC$ GUVs after incubation at T_m for 1 min with 1 mM calcein.

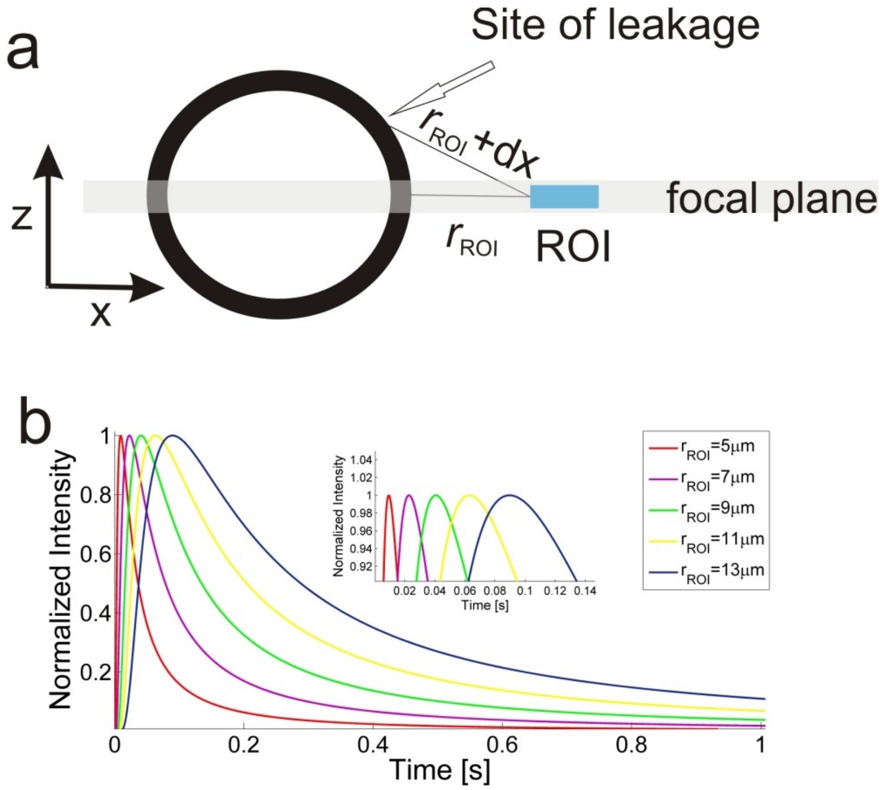

Figure S8 Effect of distance to a point source on the localization of the intensity peak as predicted by the solution to Fick's second law, eq. 1 in the paper. (a) Schematics showing the distance for a leakage site positioned out of focus and the distance corresponding to the shortest distance between the ROI and the GUV, r_{ROI}. (b) By changing the distance from 5 µm to 13 µm the position of the intensity peak only shifts ca. 80 ms showing that a large uncertainty in the distance translates into a small uncertainty in the location of the peak which is smaller than the experimental time resolution.

Movie Legends

Movie 1 A local phase transition induced by optically trapping a gold nanoparticle (*d* = 80 nm) near two GUVs made from $DC_{15}PC$. The phase transition occurs at T_m ~ 33°C and the fluid part of the membrane is visualized using the potentiometric dye di-4-ANEPPDHQ which partitions strongly into the fluid phase of the membrane.

Movie 2 Control showing that no leakage occurs when locally heating a GUV in fluid phase. An optically trapped *d* = 80 nm gold nanoparticle is used to locally heat the GUV which is made of DOPC lipids (phase transition T_m = -17°C).