

Supplementary Information for:

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Particle/fluid interface replication as a means of producing topographically patterned polydimethylsiloxane (PDMS) surfaces for deposition of lipid bilayers

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Methods:

Particle silanization: Particles in chloroform at a concentration of 1 mg/ml were allowed to react with chlorotrimethylsilane or chloro(dodecyl)dimethylsilane for 30 minutes. Excess silane was removed from the particle suspension through 10 successive centrifugation-resuspension cycles with pure chloroform.

Bacteria: *Bacillus subtilis* strain PY79 was grown overnight on Luria-Bertani Broth agar plates at 37°C. Cells were then scraped off of the plate, resuspended in tryptone broth (TB) and grown under constant agitation at 37°C for 2 hours. The TB supernatant was discarded and replaced with ultrapure water (Milipore). 3 centrifugation resuspension cycles were carried out with ultrapure water, and the final resuspension was done with ethanol.

Preparation of small unilamellar vesicles (SUV): An appropriate amount of lipid/chloroform solution was placed in a glass scintillation vial (typically to obtain 3 mg of lipids per vial). A thin film of dry lipid was obtained by using a stream of nitrogen to evaporate the chloroform. The vial was left under vacuum overnight to remove all traces of chloroform. The dried lipid films were rehydrated with about 3 ml Tris-HCl buffer (10 mM Tris, 150 mM NaCl, 2 mM CaCl₂, adjusted with 12 M HCl to pH 7.5) (lipid concentration 1 mg/ml). The turbid

suspension, which contained multilamellar liposomes, was then sonicated using a probe sonicator (Branson) for 10 minutes at 30 percent power until the solution became clear. The vial was submerged in an ice bath during sonication to prevent excessive heating of the lipid suspension. The SUV suspension was then centrifuged for 10 minutes at 10,000 rpm to pellet platinum particles that were released from the sonicator tip.

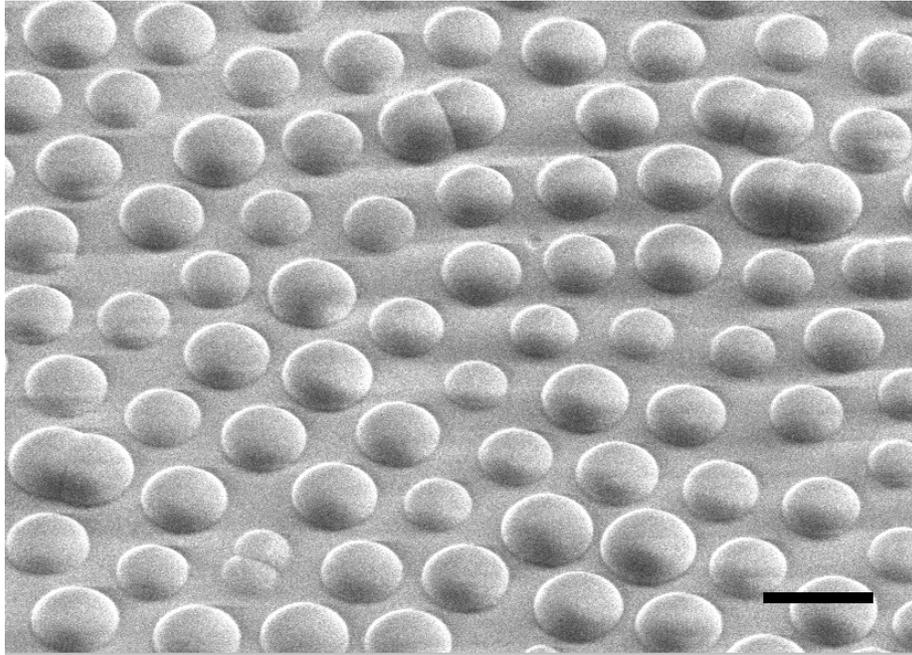
SUV fusion to produce supported lipid bilayers: PDMS substrates were cleaned by sonicating for 10 minutes sequentially in acetone, ethanol and ultrapure water. Substrates were then allowed to dry overnight in a 65°C oven. Tris-HCl buffer and the SUV suspension at a 20:1 volume ratio were introduced over the clean freshly plasma-oxidized PDMS substrate (10 seconds in a Harrick Plasma Cleaner/Sterilizer PDC-32G at maximum RF power). The substrate was allowed to incubate for 1 hour and then carefully washed with ultrapure water to remove excess unfused vesicles. Once prepared, the bilayer was kept immersed in an aqueous environment at all times.

Preparation of giant unilamellar vesicles (GUV) and double bilayers: For the double bilayer system upper bilayers were formed by rupturing giant unilamellar vesicles (size range 5-50 μm in diameter) onto a preexisting bilayer produced through the SUV fusion technique. The giant vesicles were formed using the electroformation technique[1]. 6 μl of the desired lipid mixture at a concentration of 1 mg/ml were spread onto a clean indium tin oxide (ITO) coated glass slide in a thin layer using a glass syringe (Hamilton). The slide was placed in a vacuum dessicator overnight to remove all traces of chloroform. A PDMS gasket was used to construct a barrier around the lipid film, and the slide was placed onto a 65°C hotplate (to ensure the lipids were above the phase transition temperature). About 600 μl of 500 mM sucrose solution was used to hydrate the lipid film, and a second ITO slide was used to close the chamber. The slides were connected to a function generator (Agilent) with conductive

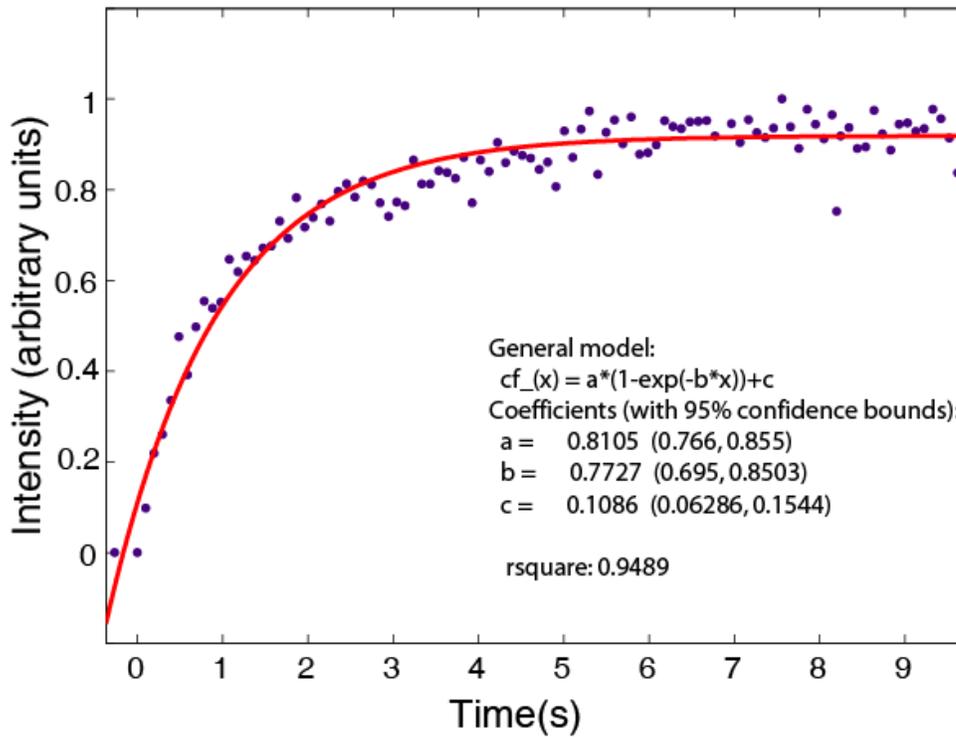
copper tape. A sinusoidal AC field was applied at a field strength of 0.5 V/mm and frequency of 10 Hz for at least 2 hours. The giant vesicles were carefully removed from the chamber and suspended in 500 mM glucose. 100 μ l of the GUV suspension was introduced to the chamber containing the lower bilayer in Tris-HCl buffer (at room temperature). The vesicles sedimented onto the lower bilayer, where many of them ruptured and formed well-defined bilayer patches in close apposition to the lower membrane[2].

[1] M. I. Angelova, D. S. Dimitrov, *Faraday Discuss.* **1986**, *81*, 303.

[2] R. Parthasarathy, C.-h. Yu, J. T. Groves, *Langmuir* **2006**, *22*, 5095.



Supporting Figure 1: SEM image of 2.37 μm radius silica particles trapped in PDMS, Several doublet particles are visible. Here the surface is chemically heterogeneous, the curved features are composed of silica while the flat regions are PDMS. Scale bar 5 μm .



Supporting Figure 2: Typical FRAP recovery curve. Blue dots are experimental data while the red line is a nonlinear least squares fit of the function $f(x) = a(1 - e^{-bx}) + c$ to the data.

Half time of recovery is calculated by $t_{half} = \frac{\ln(2)}{b}$. Diffusion coefficients were calculated

using $D = \frac{0.88(r)^2}{4t_{half}}$, where r is the radius of the bleach spot (1.98 μm for all our

experiments).