

Supporting Information for

Stiffness-Driven Modulation of Bactericidal Behavior in Nanostructured Polymer Thin Films

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Table S1. Geometric parameters of nanostructures fabricated on Silicon and PEGDMAs. (n = number of PEG repeating units)

	Si	n = 4.5	n = 9	n = 13.5
Height (nm)	551±7	499±18	495±16	496±15
Interpillar distance (nm)	366±16	355±18	353±11	352±19
Diameter at top (nm)	104±5	113±11	112±15	109±9
Diameter at bottom (nm)	246±22	235±7	234±24	233±26

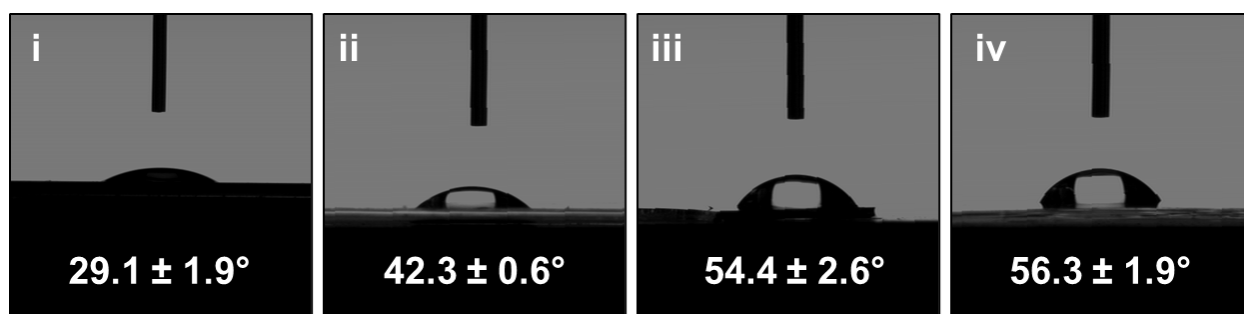


Figure S1. Contact angle measurement on flat i) Si, and PEGDMA with ii) 4.5 iii) 9 and iv) 13.5 PEG repeating units. 10 μ L water was dropped on each surface for the contact angle measurement.

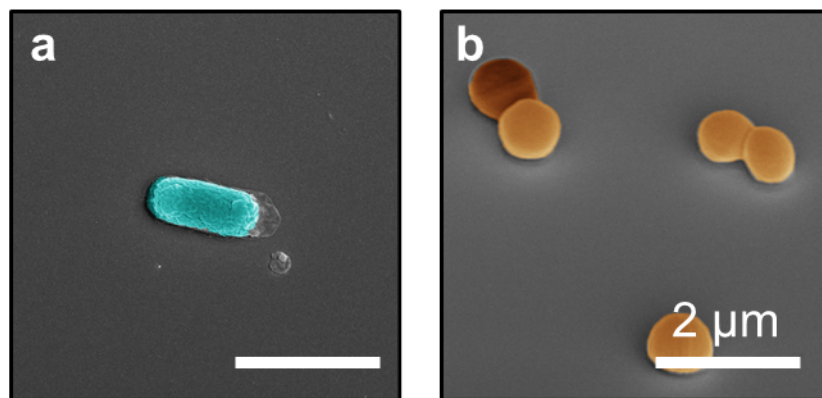


Figure S2. a) Top-view SEM images of *E. coli* incubated on flat Si in a static incubator at 37 $^{\circ}$ C for 4 h. b) Tilted-view SEM images of *S. aureus* incubated on flat Si in a static incubator at 37 $^{\circ}$ C for 4 h. All scale bars are 2 μ m.

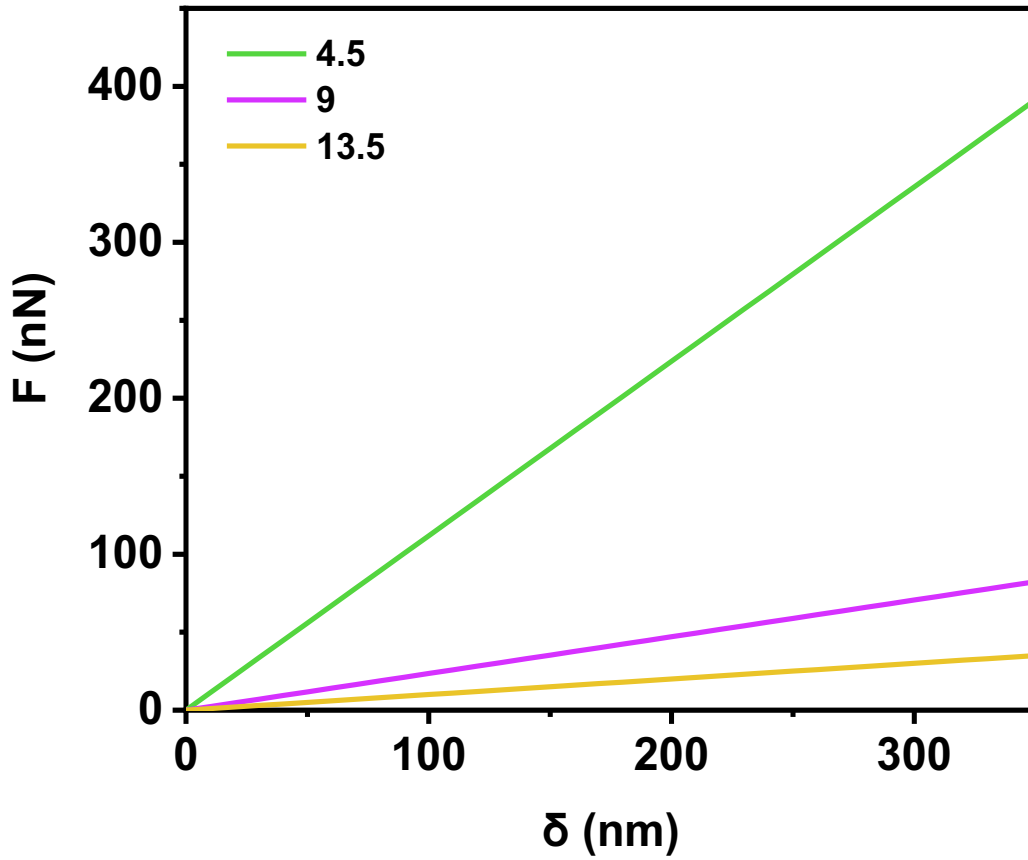


Figure S3. Force (nN) required to bend nanopillars of different stiffnesses (PEGDMA with PEG repeating units of 4.5, 9, and 13.5) as a function of displacement δ in nm on the nanopillar tip.

Table S2. Table of symbols and values used in the calculation of the surface-tension-induced force and pressure on the bacterial membrane as a function of relative liquid level during the water evaporation.

Symbol	Value	Description
d	110 nm	Diameter of nanostructures
p	350 nm	Pitch of nanostructures
f	0.0160	Solid area fraction of cylindrical posts
r_b	250 nm	Radius of bacterium
L	2.00 μm	Length of bacterium
D	500 nm	Diameter of bacterium
γ_{LG}	72 mN m^{-1}	Interfacial tension of water-gas ¹
θ_{CL}	28°	Contact angle between cell and liquid droplet ²
α	0° < α < 180°	Angle of three-phase interface at hemispherical edge
H	0 nm < H < 500 nm	Liquid level

For systems exposed to air, Valiei *et. al.*³ showed the considerable force exerted on the bacterial membrane upon the drying process, known as the capillary force. During the evaporation process, this capillary force arises as the liquid level decreases to a value smaller than the bacterial cell height. Thus, to quantify the pressure applied on the bacterial membrane following substrate adhesion, we quantified the force due to the surface tension applied over the projected area of the bacterial membrane by the nanoarray, taking into account the solid area fraction. The surface tension is induced upon exposure to air in the direction toward the interior of the nanopillars vertically along the z-axis. This surface tension in our system acts around the periphery of the meniscus of a water droplet at the water-air interface and is thus derived to obtain the following³:

$$F_{ST} = \int \gamma_{LG} dl \quad (1)$$

where γ_{LG} is in interfacial tension of the water-air interface and is integrated over the entire meniscus perimeter. Assuming a constant cell-liquid contact angle, θ_{CL} , and idealized bacterium with hemispherical caps of radius r_b and body length L , Eq. 1 can be simplified as follows to describe the force at any point along the evaporation process defined by α , or the angle of the three-phase contact line at the bacterium hemispherical edge:

$$F_{ST} = 2\gamma_{LG} \sin(\alpha + \theta_{CL}) (\pi r_b \sin \alpha + L) \quad 0 \leq \alpha \leq \pi \quad (2)$$

where θ_{CL} is the bacterium-LB media contact angle, and all remaining variables have been previously defined.

The correlation between relative liquid level (H/D) and α can be further described in Eq 3.

$$\frac{H}{D} = \frac{r_b(1-\cos(\alpha))}{D} \quad (3)$$

where H is the liquid level and D is the diameter of the bacterium.

Finally, to compare quantities among cells of varying areas, it is useful to utilize the calculated force applied normalized by its respective projected area in contact with a cylindrical post to describe the pressure induced on the bacterial membrane by the nanoarray:

$$P_{ST} = \frac{F_{ST}}{(\pi r_b^2 + 2r_b L)f} \quad (4)$$

where f is the solid area fraction that describes the bacterium's projected surface area in contact with a nanocylinder.

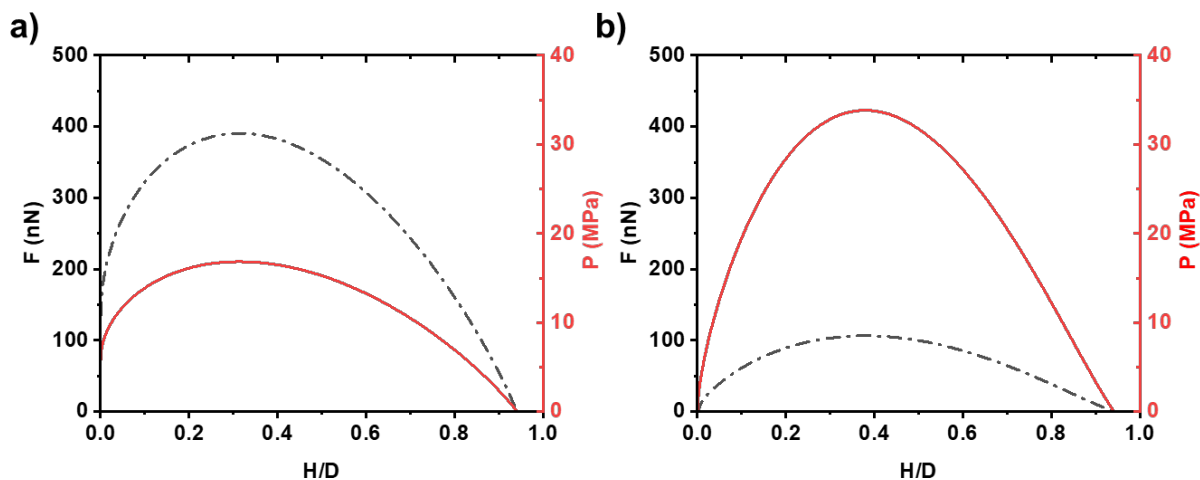


Figure S4 Surface-tension-induced force (nN) and the resulted pressure (MPa) applied on the membrane of a) *E. coli* and b) *S. aureus* as a function of the relative liquid level (H/D) during the water evaporation.

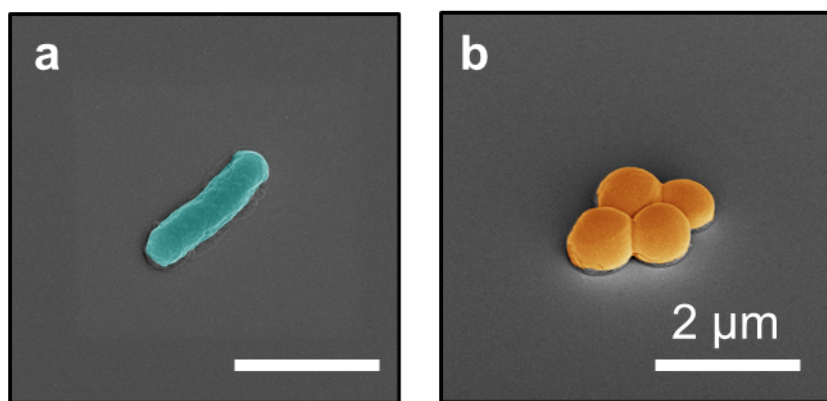


Figure S5. a) Top-view SEM images of *E. coli* incubated on flat Si in a static incubator at 37 °C for 4 h followed by 60 s drying in air. b) Tilted-view SEM images of *S. aureus* incubated on flat Si in a static incubator at 37 °C for 4 h followed by 60 s drying in air. All scale bars are 2 μm.

References:

1. Vargaftik, N.; Volkov, B.; Voljak, L., International tables of the surface tension of water. *Journal of Physical and Chemical Reference Data* **1983**, *12* (3), 817-820.
2. Hamadi, F.; Latrache, H.; Zahir, H.; Elghmari, A.; Timinouni, M.; Ellouali, M., The relation between *Escherichia coli* surface functional groups' composition and their physicochemical properties. *Brazilian Journal of Microbiology* **2008**, *39* (1), 10-15.
3. Valiei, A.; Lin, N.; Bryche, J.-F.; McKay, G.; Canva, M.; Charette, P. G.; Nguyen, D.; Moraes, C.; Tufenkji, N., Hydrophilic mechano-bactericidal nanopillars require external forces to rapidly kill bacteria. *Nano letters* **2020**, *20* (8), 5720-5727.