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Supporting Material

Substrate topography induces a cross-over from 2D to 3D behavior in fibroblast migration

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MOVIE CAPTIONS

Movie S1: Migration of 3T3 cells in micropillar substrates. The height of the pillar is 6 µm and their diameter and spacing are 5 µm. Duration of the movie = 4 hours.

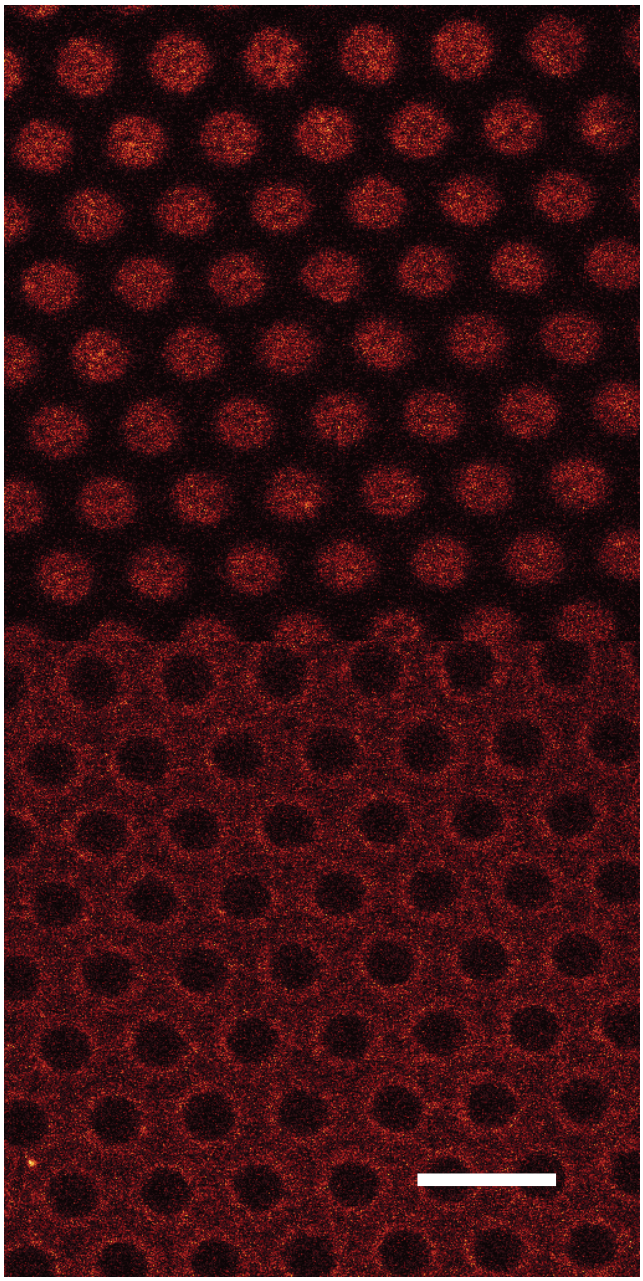
Movie S2: Image stacks of confocal microscopy of fixed 3T3 cells on (7-10-10) micropillar substrates. Cells are immunolabelled for actin in red and vinculin in green. The first image corresponds to the top of the micropillars and the last one to the bottom of the pillars.

Movie S3: Image stacks of confocal microscopy of fixed 3T3 cells on (6-5-5) micropillar substrates. Cells are immunolabelled for actin in red and vinculin in green. The first image corresponds to the top of the micropillars and the last one to the bottom of the pillars.

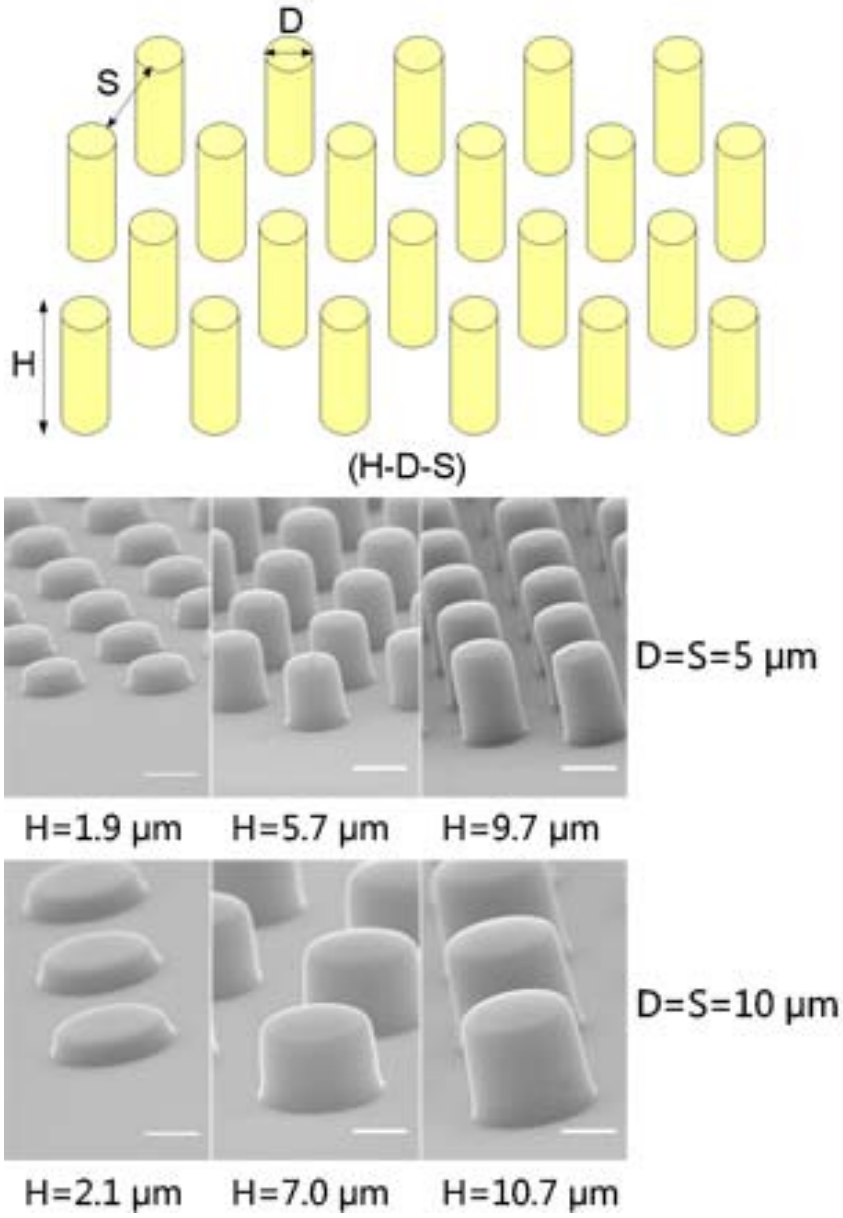
Movie S4: A cell approaches the boundary between the micropillar substrate and the flat surface from the flat side, sends some protrusions in between the pillars but stays on the flat part ((6-5-5) substrate).

Name	Height (µm)	Diameter (µm)	Spacing (µm)
(2-5-5)	1.9	5	5
(2-10-10)	2.1	10	10
(6-5-5)	5.7	5	5
(7-10-10)	7	10	10
(10-5-5)	9.7	5	5
(10-10-10)	10.7	10	10

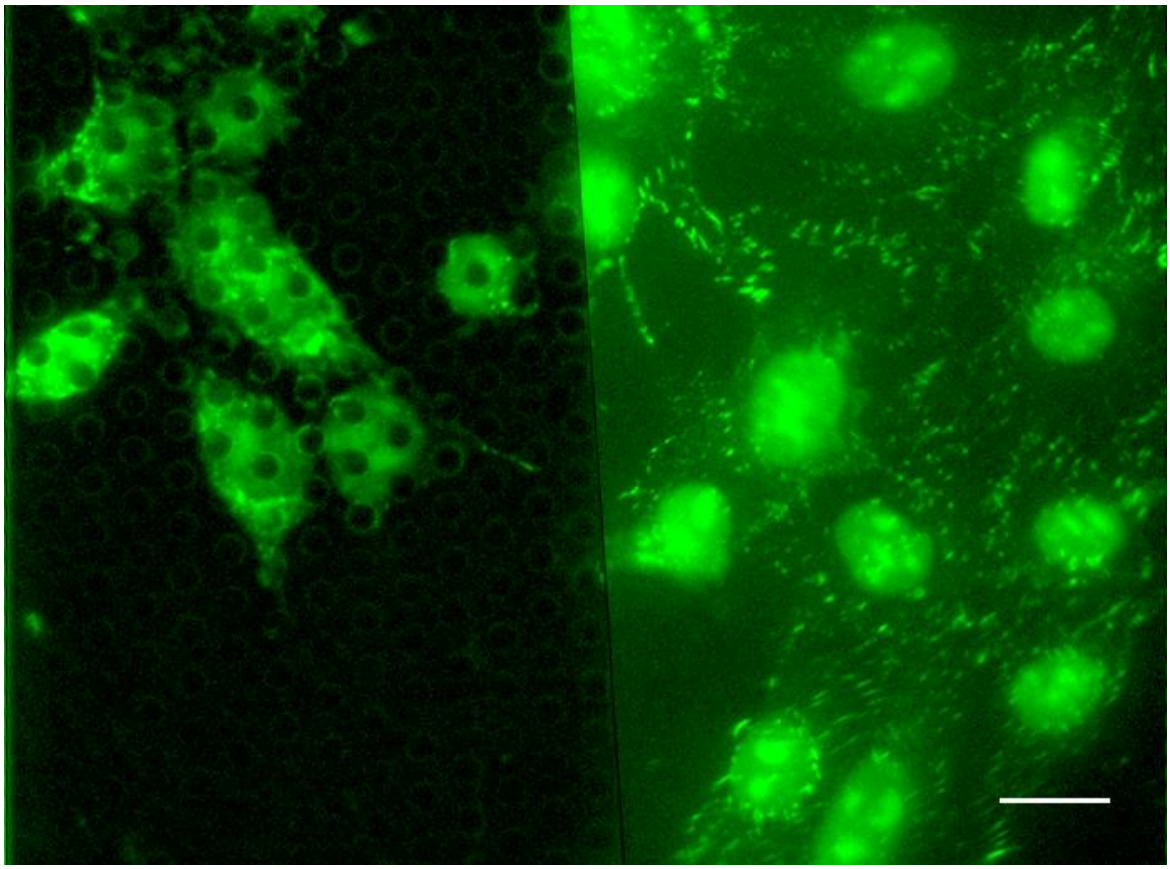
Supplemental Table S1: Dimensions of the micropillar substrates defined by three numbers (H-D-S), Height, Diameter and Spacing, respectively. The substrates consist of a hexagonal array of cylindrical pillars with three different heights, H, from 2 to 10 µm, diameters, D, 5 and 10 µm and spacing from edge to edge of the pillars, S, 5 and 10 µm.



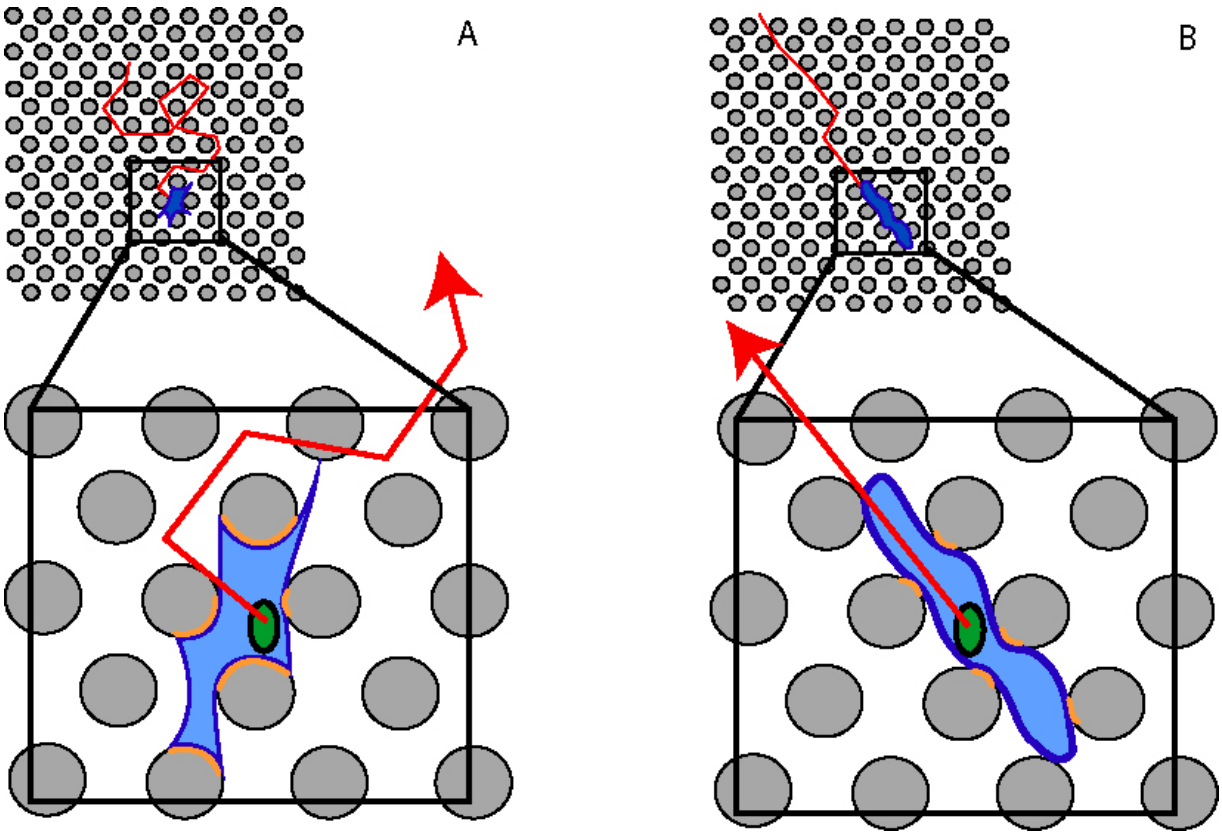
Supplemental Figure S1: Confocal images of single slices of micropillar substrates fluorescently labelled with fibronectin. (A) Bottom and (B) Top of the micropillar substrates. Scale bar= 15 µm.



Supplemental Figure S2: Schematic representation of the micropillar substrates indicating the important geometrical parameters (H, D and S correspond respectively to the height, the diameter and the spacing between the pillars). Scanning Electron Micrographs of the different types of pillars used in this study. Bar = 5 µm



Supplemental Figure S3: Immunofluorescent staining images of vinculin protein at the boundary of a micropillar (10-5-5) substrate (left side) and a flat surface (right side). The distribution of vinculin protein is different on both sides with a higher cytoplasmic signal and less FAs on the micropillar side than on the flat one. Scale bar = 20µm.



Supplemental Figure S4: Schematic description of the dynamics of cell movements on micropillar substrates. (A) Cells moving between the micropillars develop large FAs on the edge of the pillars and exhibit a slow down but more persistent movement than on a flat surface. Focal adhesions (labeled in orange) are stabilized along the micropillars and promote cell migration from pillar to pillar. (B) Cells treated with blebbistatin exhibit a directed motion between the pillars due to the geometrical constraints.