ACTIVE RUPTURE IN EPITHELIA GOVERNED BY NANOSCALE REGULATION OF CELL-ECM INTERACTION

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INTRODUCTION

Epithelial monolayers rely on coordinated mechanical forces to maintain homeostasis and tissue integrity. These forces generate tissue-scale patterns that regulate cell division, migration, extrusion, and intercalation. Alongside cell–cell junctions, cell–substrate interactions play a central role. Integrin-mediated adhesions, particularly focal adhesions and hemidesmosomes, connect epithelial cells to the extracellular matrix (ECM). By clustering and binding to ECM proteins such as fibronectin, integrins strengthen adhesion and transmit mechanical cues. Among them, αvβ3 integrin is especially relevant for studying how epithelial cells respond to nanopatterned surfaces.

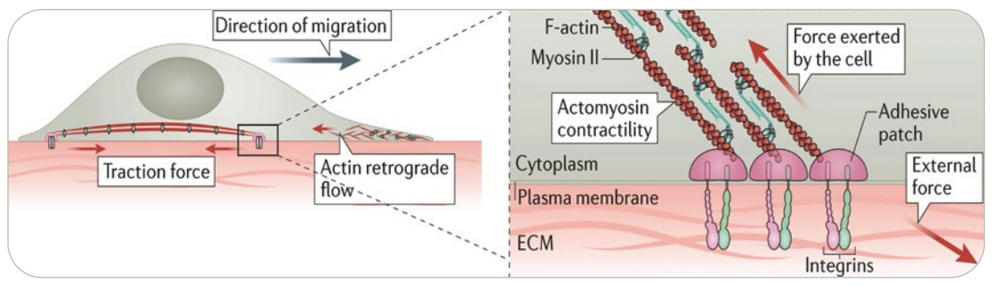


Figure 01: Integrin-mediated cell-matrix adhesions build traction forces inside the cell.

OBJECTIVES

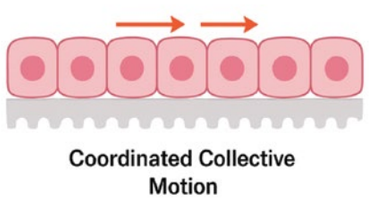


Figure 02: Establishing Monolayers on Nanopatterned Surfaces.

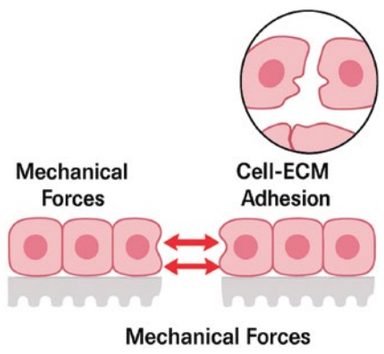


Figure 03: Understanding forces and events in Epithelial Fracture.

METHODS

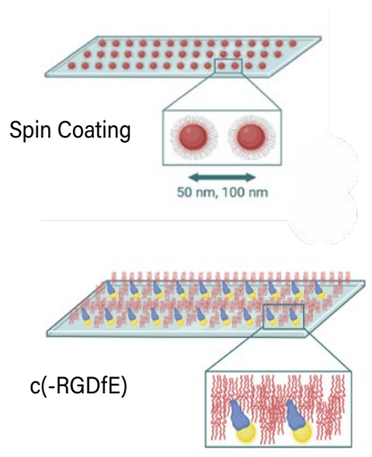


Figure 04: Illustration of the main steps to obtain Nanopatterned surfaces.

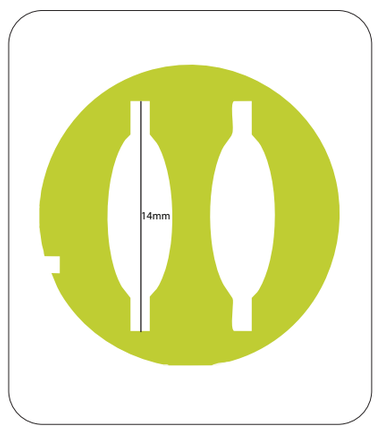


Figure 05: PDMS stamps inserted on Nanopatterned surfases to provide micro confimenment for cells.

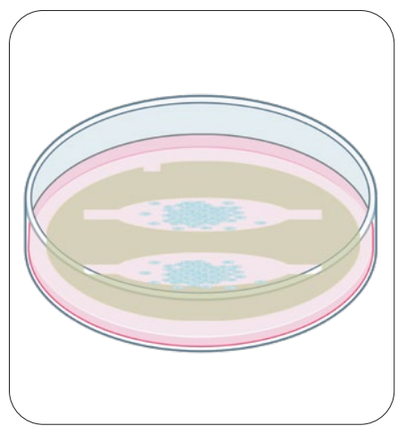


Figure 06: Cells seeded on Nanopatterned surfaces with PDMS confimenment.

RESULTS

FULL FIELD OF VIEW - 80NM SPACING

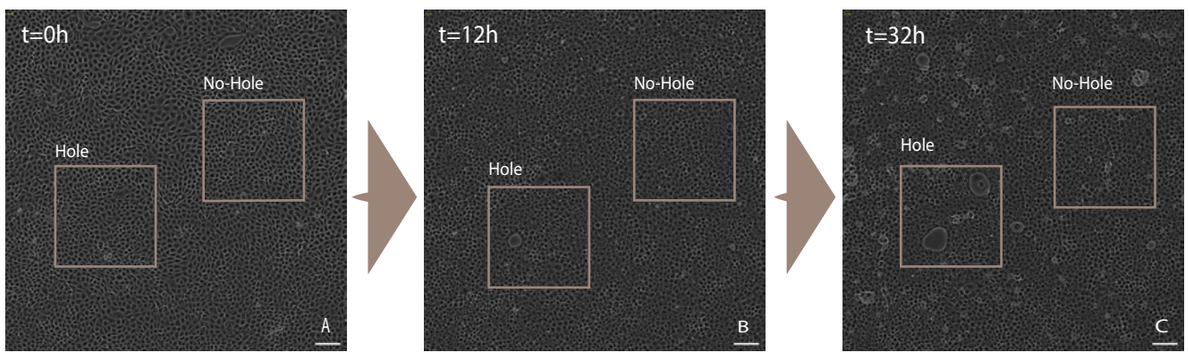


Figure 07: A B & C : MDCK Monolayer seeded on confined Nanopattrened substrates to understand their collective behaviour and dynamics of tear in epithelia.

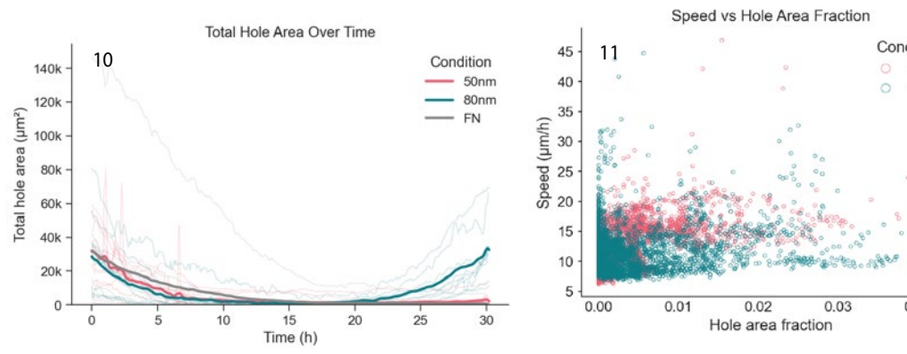


Figure 10 : Dynamics of holes appearing over time . Figure 11: Appearance of holes when related with speed of the monolayer.

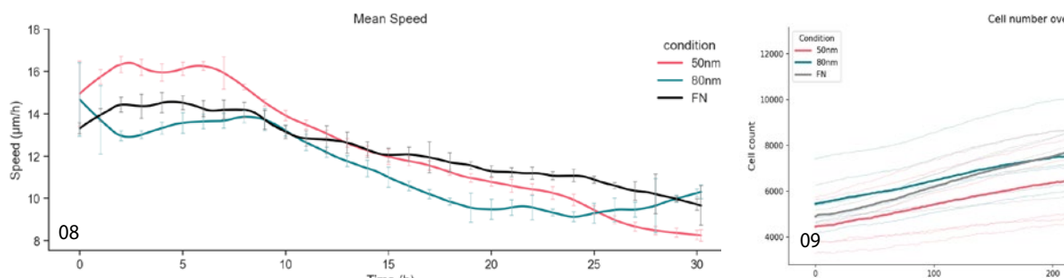


Figure 08: Average speed of the monolayer across 30h .We observe increased speed in 80nm which is uncharecteristic to jammed monolayer.
Figure 09: Decrease in cell density over time implying improper jamming in the monolayer.

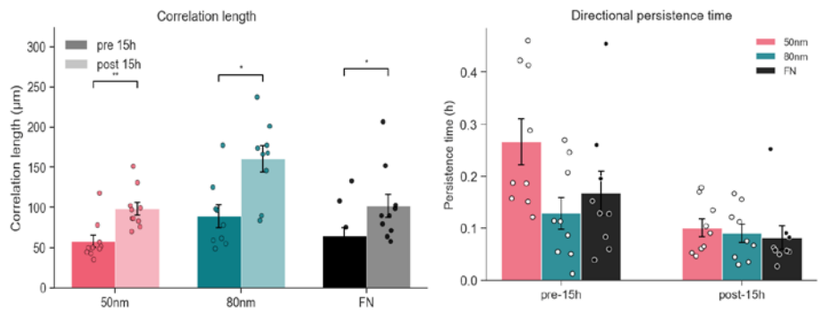
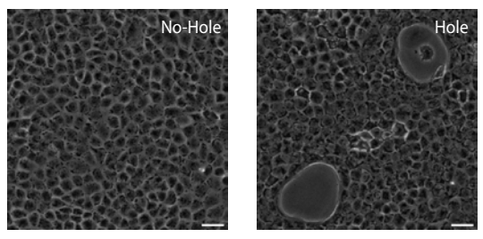
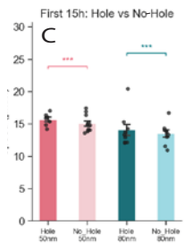
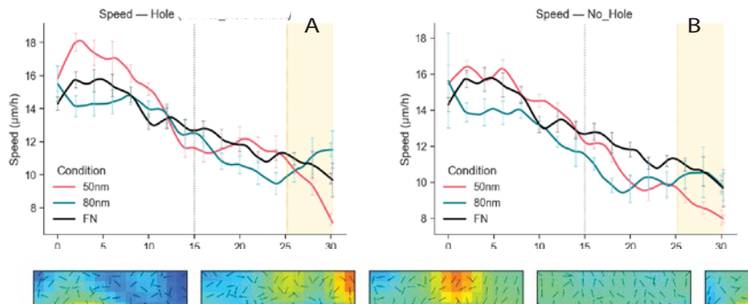


Figure 12 & 13: Temporal heterogeneity in the monolayer due to improper unjamming.

CROPPED FOV : HOLE AND No-HOLE REGIONS







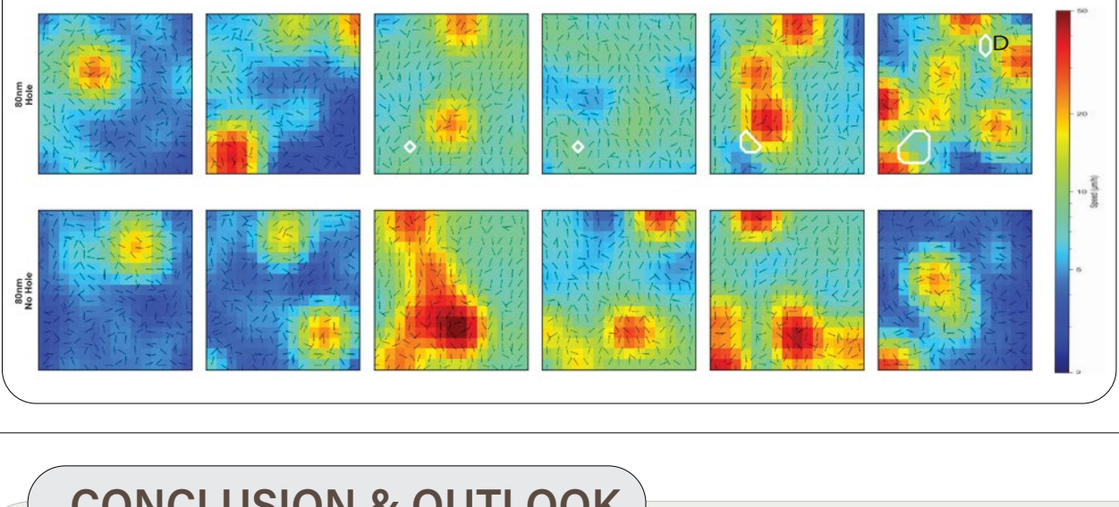
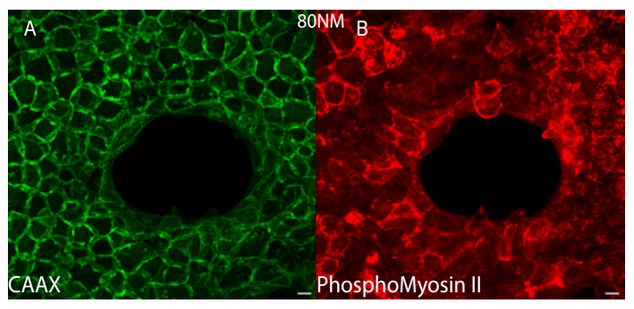
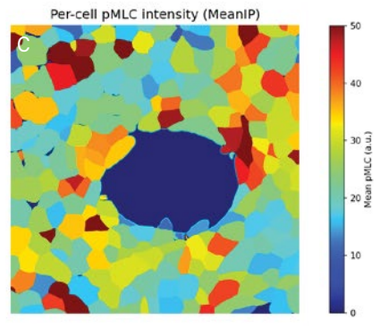
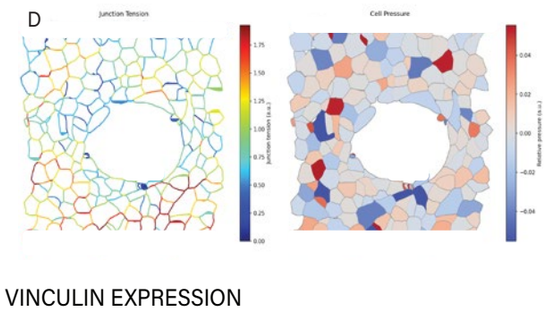


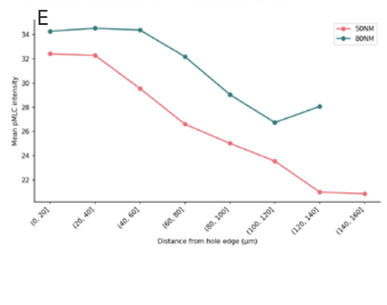
Figure 14 : A,B &C Combination of spatial and temeproral heterogeneity in speed leading to the formation of tears in monolayers.
D: Heat map of speed in the Hole & No Hole depicting the evolution of regions around the tear

MYOSIN DISTRIBUTION NEAR TEARS









VINCULIN EXPRESSION

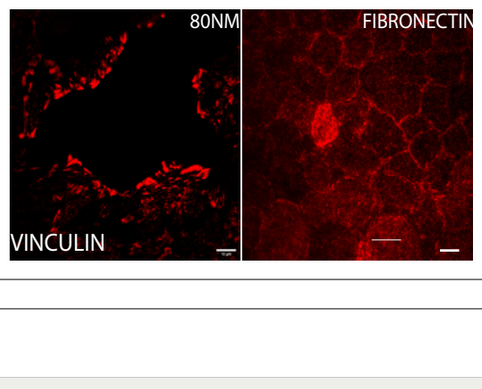


Figure 15: A&B CAAX and pMLC dislaying the recruitment of Myosin around the holes in a monolayer
C : Myosin intensity disctribution in cells around the hole
D : Force inference and pressure inference relating to myosin expression.
E: Sapatial distribution of Myosin expression.

Figure 16: Increased Vinculin exprssion in Focal Adhesion on Nano patterned surface

CONCLUSION & OUTLOOK

- Spatiotemporal heterogeneity in collective motion underlies hole formation and epithelial fracture across nanopatterned surfaces with varied integrin spacing.
- Increased Vinculin expression at Focal Adhesion sites suggests enhanced mechanosensing on nanopatterned substrates.
- Elevated pMLC around holes reflects actomyosin-driven contractility as cells attempt to recover monolayer integrity after tear formation.
- Further understanding of Actin organization, E-cadherin junction remodeling, and cryptic lamellipodia to understand how coordination breaks down at the nanoscale.

REFERENCES

- Ladoux & Mège, Nat Rev Mol Cell Biol, 2017
- Sonam et al., Nat Phys, 2023
- Levario-Diaz V., et al. (2022) Front. Cell Dev. Biol.,10.
- Cavalcanti-Adam et al., Biophys J, 2007