

# Decoding Cellular Mechanical State in Stiffness-Driven Contact Guidance

Mathis Grelier<sup>1</sup>, Maja Milas<sup>2</sup>, Carlos Ureña Martín<sup>3</sup>, Mark Schwartzman<sup>3</sup>, Ana-Sunčana Smith<sup>12</sup>

<sup>1</sup> PULS Group, Friedrich-Alexander-Universität Erlangen-Nürnberg

<sup>2</sup> Division of Physical Chemistry, Institute Ruđer Bošković Zagreb

<sup>3</sup> Department of Materials Engineering and Ilse Katz Institute for Nanoscale Science and Technology, Ben-Gurion University of the Negev, Beer-Sheva

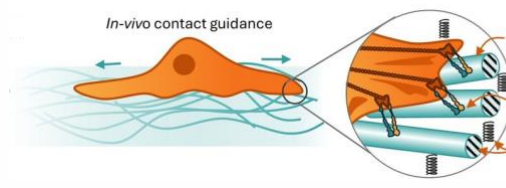
PULS Group

Physics Underlying Life Sciences

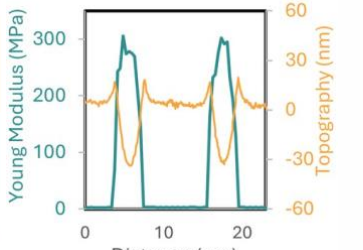
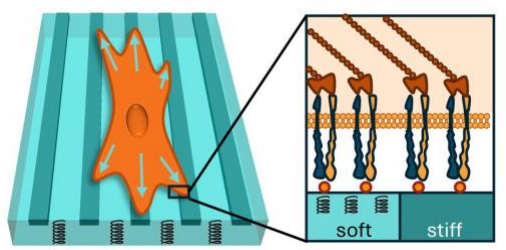
## Finding the Hidden Mechanical State of Cells

- **The Challenge:** A cell's internal mechanical state drives biological processes like contact guidance, yet directly measuring these properties typically requires invasive force microscopy.
- **Our Approach:** Instead of using physical probes to deform the cell, we use fluctuations in cell shapes as a non-invasive physical "excitation".
- **The Question:** While contact guidance is a striking phenomenon, we use it here as an experimental probe to answer a fundamental question: does an asymmetric elastic landscape actively alter the cell mechanical state, or does that state remain entirely invariant?

## Contact Guidance by Elastic Heterogeneity Alone

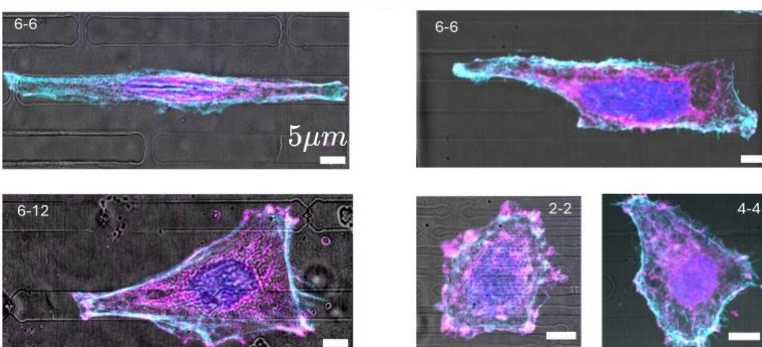


- **Subcellular Micro-Patterns:** We engineered flat, elastic PDMS substrates embedded with micro-strips of highly rigid silica with **Topographic Uniformity**.



- **Isolating the Cue:** We apply our approach to cells in a heterogeneous elastic environment, completely eliminating confounding environmental signals like topography or adhesive chemical patterns.

- **Pattern-Driven Cell Diversity**



## A Probabilistic Energy Framework

**The Predictive Framework:** We study the effect of contact guidance on the cell mechanical state. The minimum energy functional that capture the mechanical state is :

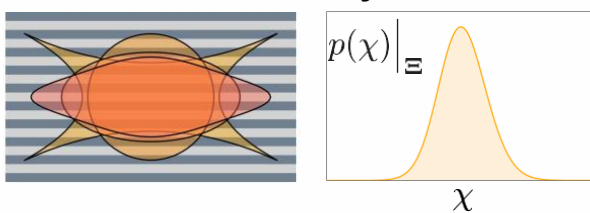
$$E(\chi) = \underbrace{-\alpha_h A_h(\chi) - \alpha_s A_s(\chi)}_{\text{Contact energy}} + \underbrace{K_A (A_0 - A_{\text{tot}}(\chi))^2}_{\text{Area elastic energy}} + \underbrace{\lambda_p P(\chi)}_{\text{Tension}}$$

**The State Parameter Set:** This minimal formulation fully captures the cellular mechanical state via the vector:  $\Xi = (\alpha_h, \alpha_s, K_A, A_0, \lambda_p)$

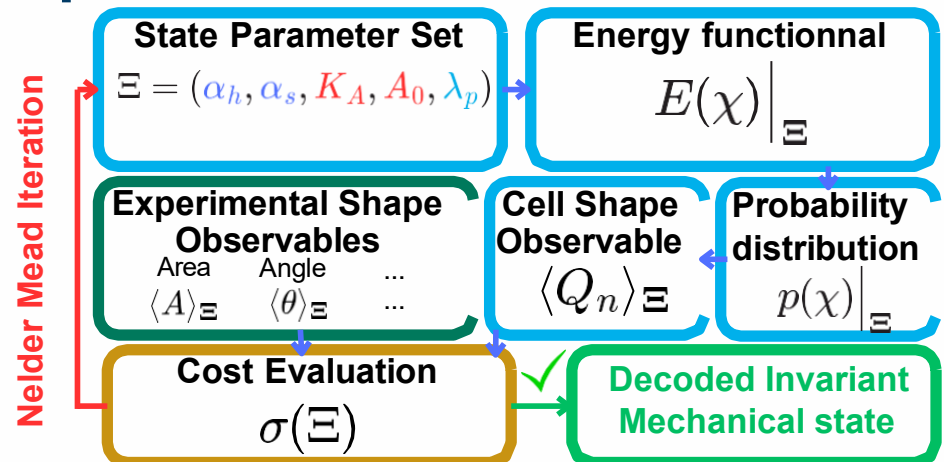
**The Shape Parameter Set:**  $\chi$  fully encompasses the shape ensemble parameterized by a superellipse basis.

$\alpha_h, \alpha_s$  Surface tensions  
 $K_A$  Area elasticity modulus  
 $A_0$  Preferred area  
 $\lambda_p$  Cortical tension

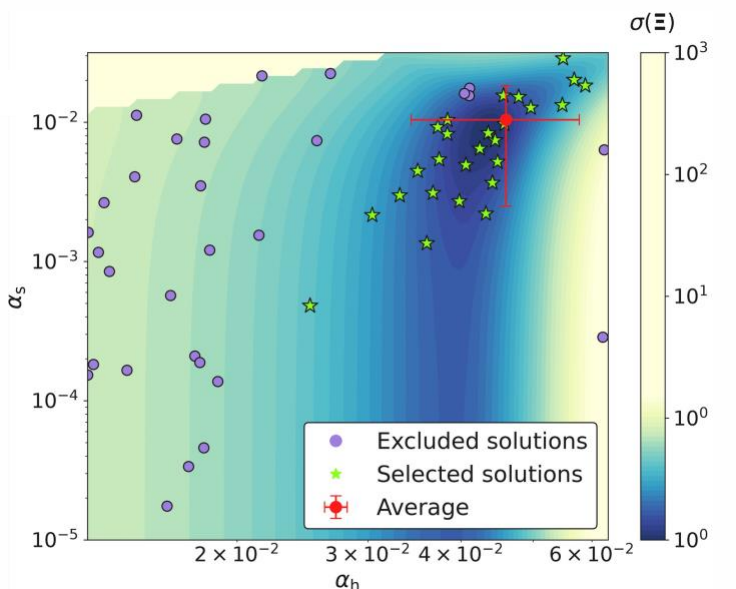
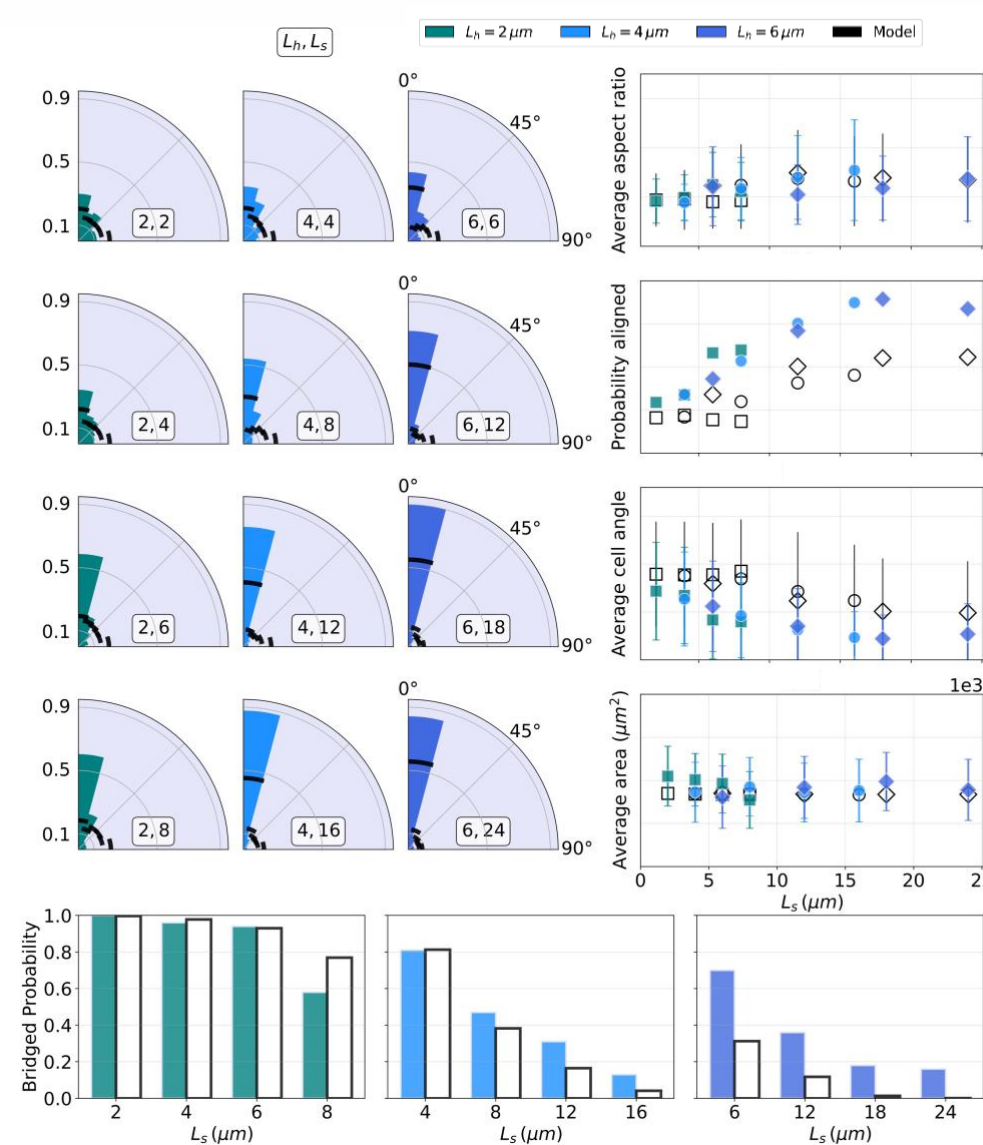
**The Probability Distribution:**



## Optimization Procedure



## Soft Stripes as Energetic Barriers



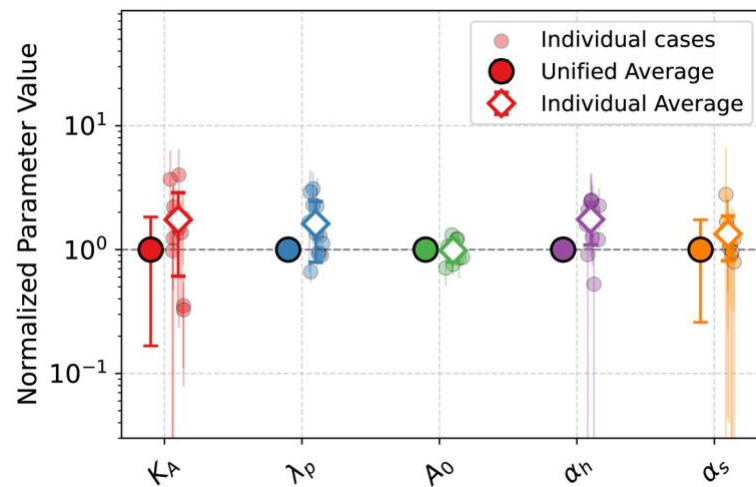
| Parameter   | Unit             | Fit Results ± SD                 | Expected            |
|-------------|------------------|----------------------------------|---------------------|
| $K_A$       | N/m <sup>3</sup> | $(5 \pm 4) \times 10^8$          | $10^7 - 10^{11}$    |
| $\lambda_p$ | N                | $(1.11 \pm 0.05) \times 10^{-8}$ | $10^{-9} - 10^{-7}$ |
| $A_0$       | μm <sup>2</sup>  | $(3.9 \pm 0.4) \times 10^2$      | $10^2 - 10^3$       |
| $\alpha_h$  | N/m              | $(6.4 \pm 0.7) \times 10^{-2}$   | $10^{-3} - 10^{-1}$ |
| $\alpha_s$  | N/m              | $(7 \pm 5) \times 10^{-3}$       | $10^{-3} - 10^{-1}$ |

**Signal Averaging:** On narrow soft stripes, contact guidance is negligible.

**Confinement and Alignment:** As the soft stripe width increases, it acts as a progressive energetic barrier.

## Patterning Geometry Does not Affect the Cell State

**Global vs. Individual Optimization:** We compared a unified global fit across all 12 experimental geometries simultaneously against independent optimizations performed pattern-by-pattern.

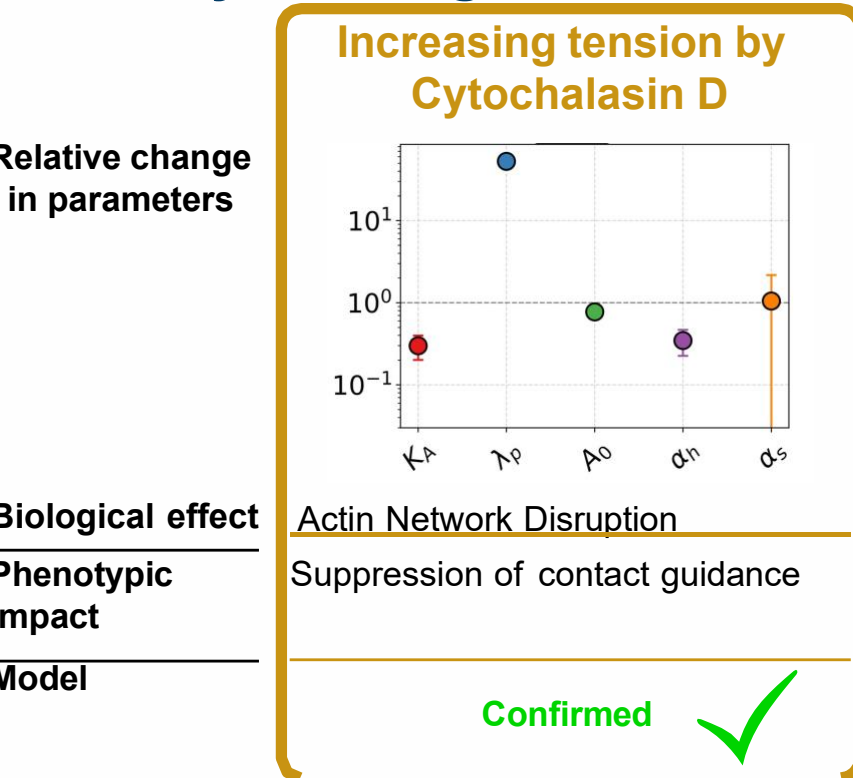


**The Invariance Proof:** The mechanical parameters calculated from individual pattern fits tightly cluster around the unified global average.

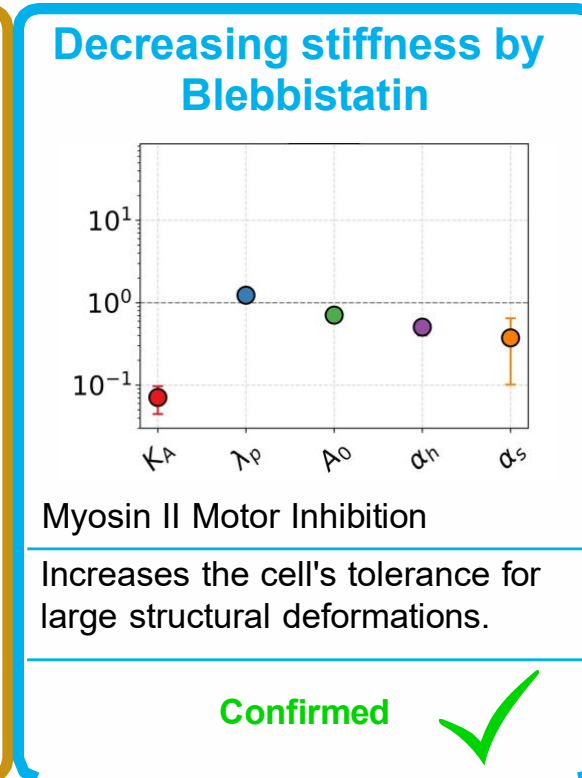
**Intrinsic State Characterization:** The cell's underlying mechanical state is conserved.

Confirmed ✓

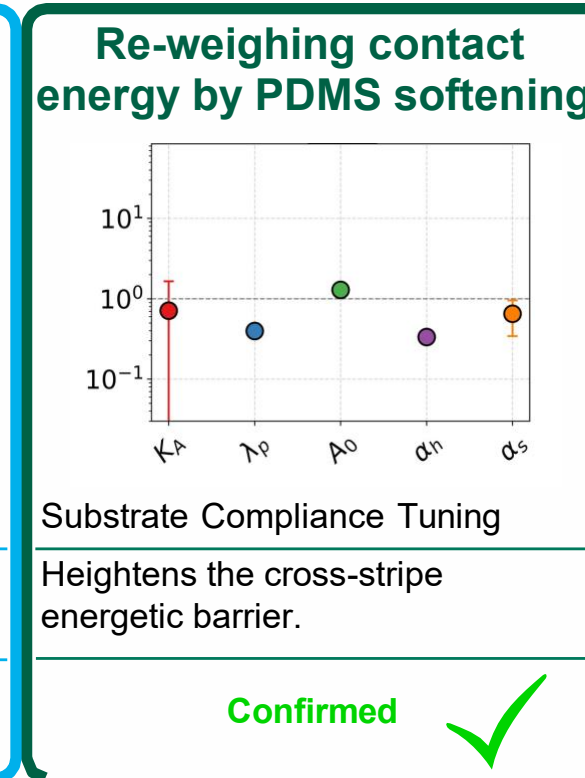
## Actively Shifting the Mechanical State... or not



**Biological effect** Actin Network Disruption  
**Phenotypic impact** Suppression of contact guidance  
**Model** Confirmed ✓



**Biological effect** Myosin II Motor Inhibition  
**Phenotypic impact** Increases the cell's tolerance for large structural deformations.  
**Model** Confirmed ✓



**Biological effect** Substrate Compliance Tuning  
**Phenotypic impact** Heightens the cross-stripe energetic barrier.  
**Model** Confirmed ✓

## Conclusions & Impact

