Biomechanical Characterisation of Chemoresistant Breast Cancer Cell Lines

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State of the Art

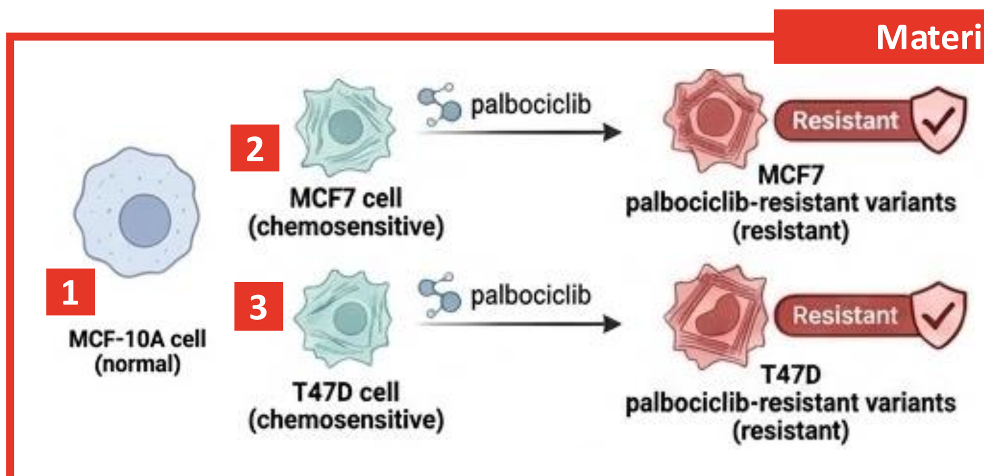
Breast cancer is one of the leading causes of cancer-related mortality worldwide, characterized by molecularly heterogeneous subtypes with distinct biological profiles and clinical outcomes. **CDK4/6 inhibitors** such as **palbociclib** have significantly improved progression-free survival in **hormone receptor-positive (HR+) breast cancer.** However, acquired resistance remains a major clinical challenge that limits long-term therapeutic efficacy. The **mechanical properties** of cancer cells, including stiffness and viscoelastic behavior, are increasingly recognized **as functional hallmarks of tumor biology**, with cytoskeletal alterations linked to tumor aggressiveness, metastatic potential and treatment response.

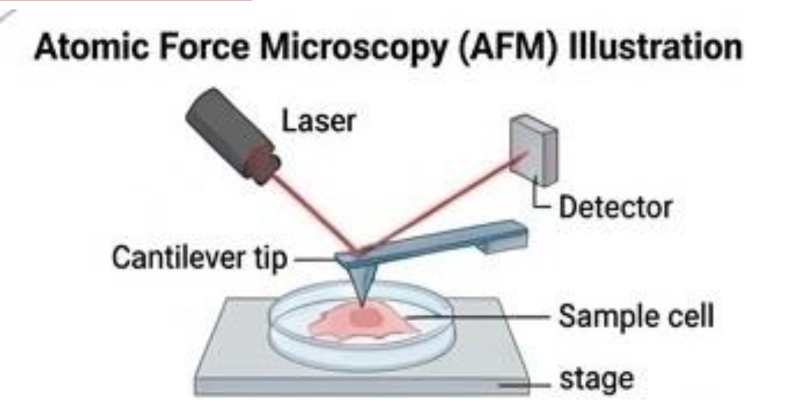
Aims

Could cell mechanics become a label-free marker of therapy resistance?

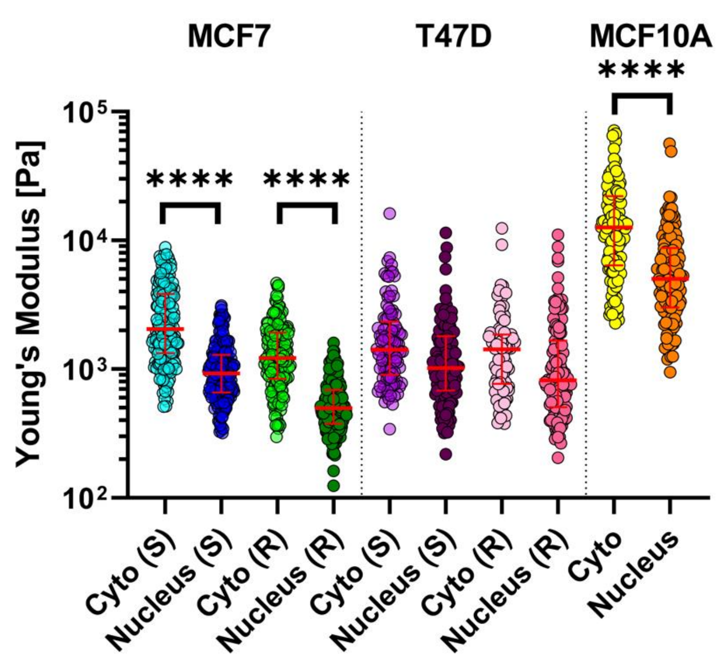
This project investigates whether palbociclib **resistance** in HR+ breast cancer is associated with **biophysical reprogramming**, employing Atomic Force Microscopy (**AFM**) for high-resolution single-cell **nano-mechanical characterization** alongside complementary molecular analyses to define resistance at both the cellular and biophysical level.

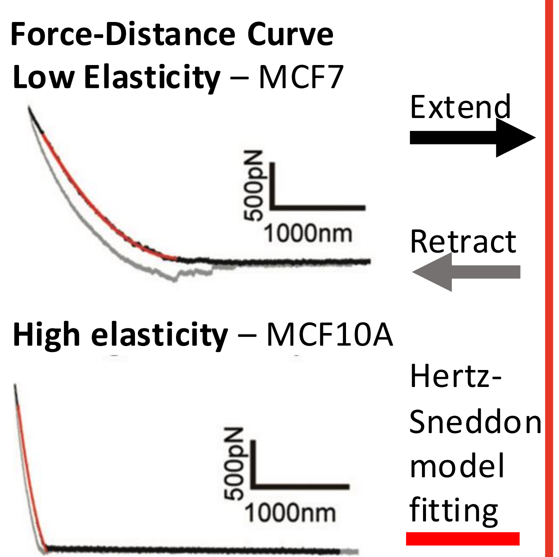
Material & Methods





Young's Modulus E [Pa]



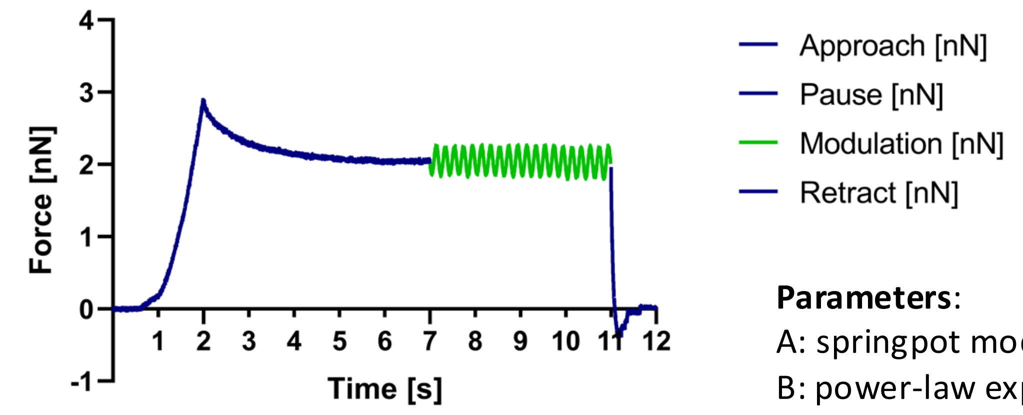


Hertz-Sneddon model, adapted to the **pyramidal tip** geometry, extraction of the **Young's Modulus:**

$$F = \frac{E}{1-\nu^2}\frac{\tan\alpha}{\sqrt{2}}\delta^2 \qquad a = \frac{\tan\alpha}{\sqrt{2}}\delta$$

- δ: indentation depth;
- a: radius of the contact circle;
- $\nu = 0.5$: poisson's ratio;

Rheology Experiment



Fractional Kelvin-Voigt Model

$G'(\omega) = A\cdot\cos(\pi B/2)\cdot\omega^B$

$G''(\omega) = A\cdot\sin(\pi B/2)\cdot\omega^B + u\cdot\omega$

$\eta(\omega) = \frac{G''(\omega)}{G'(\omega)}$

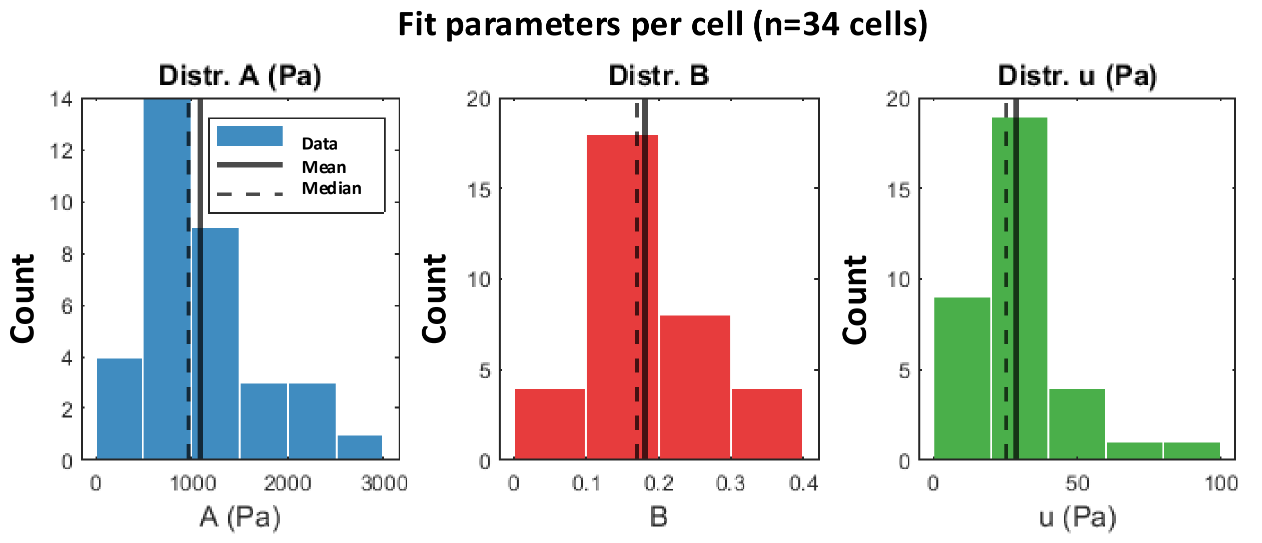
$G'(\omega)$: shear modulus
$G''(\omega)$: loss modulus
$\eta(\omega)$: loss tangent

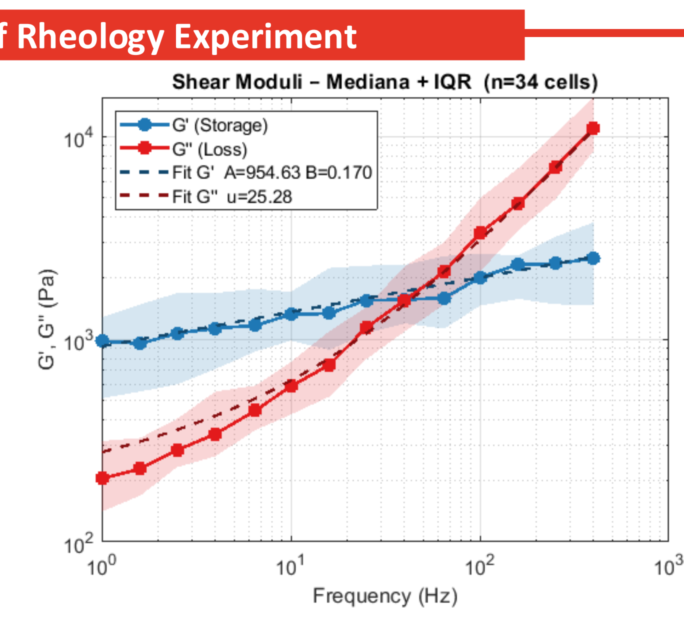
Parameters:
A: springpot modulus [Pa]
B: power-law exponent [–]
u: newtonian term [Pa·s]
f_{cross}: crossover frequency [Hz]

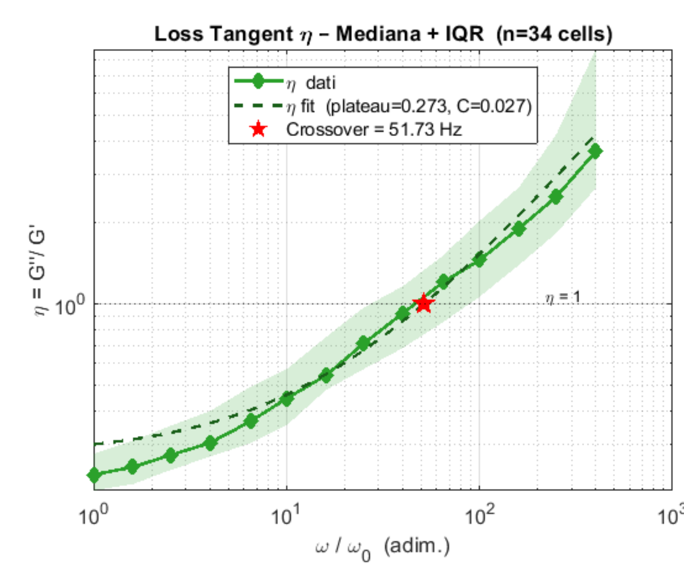
Statistics:
Wilcoxon–Mann–Whitney test, p-values $* \; p < 0.05$

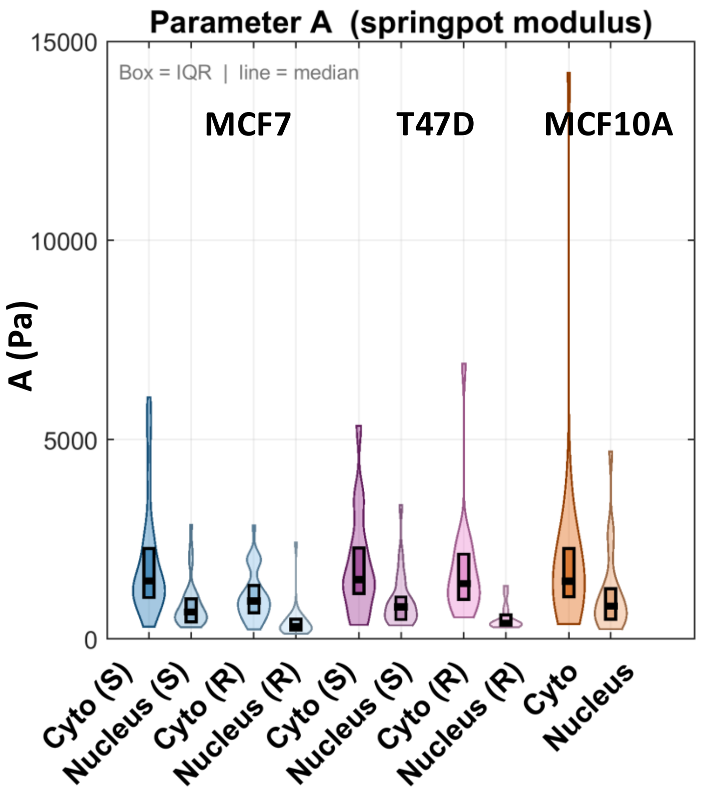
Resistance model: chronic palbociclib exposure (2 µM, 6 months) on MCF-7 and T47D

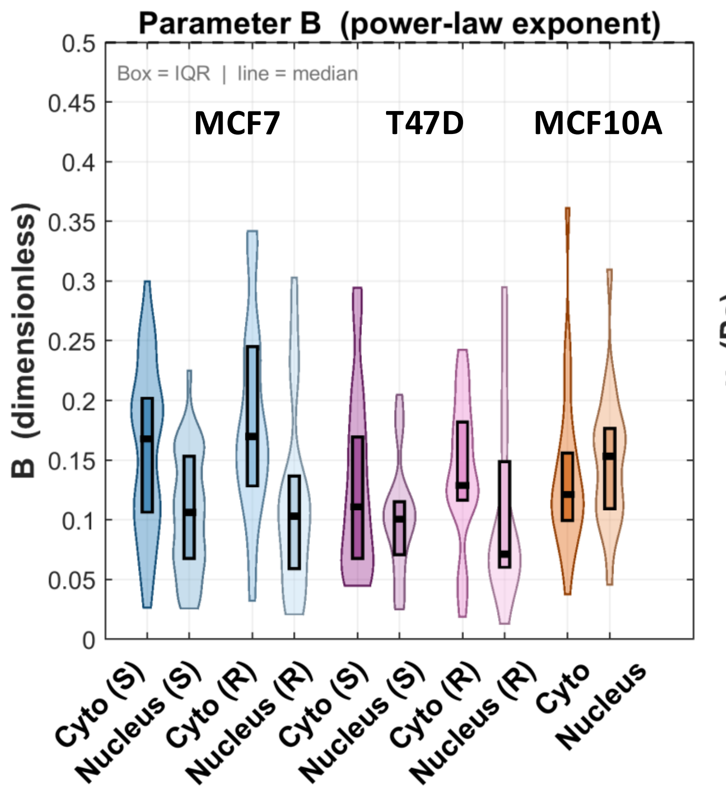
Results of Rheology Experiment

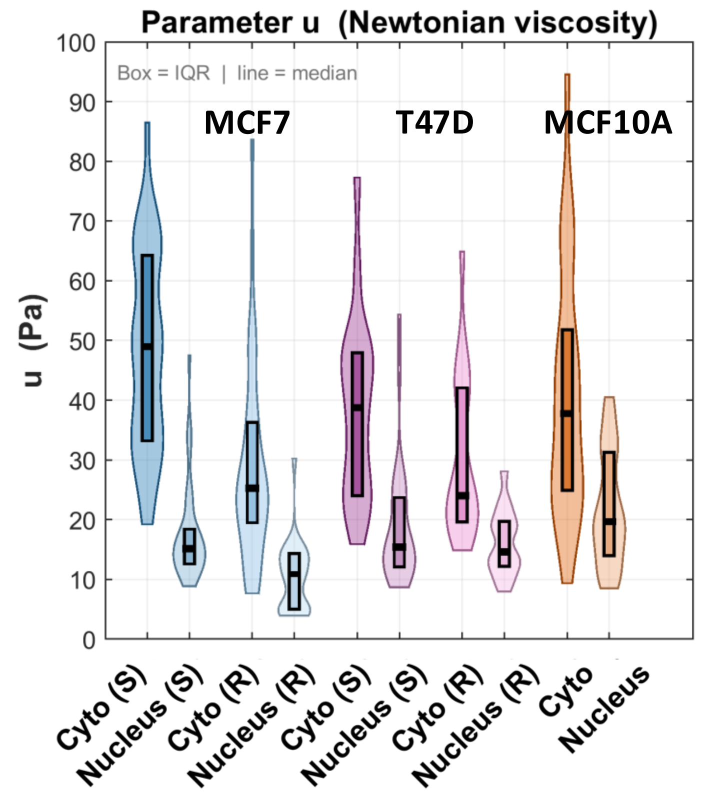


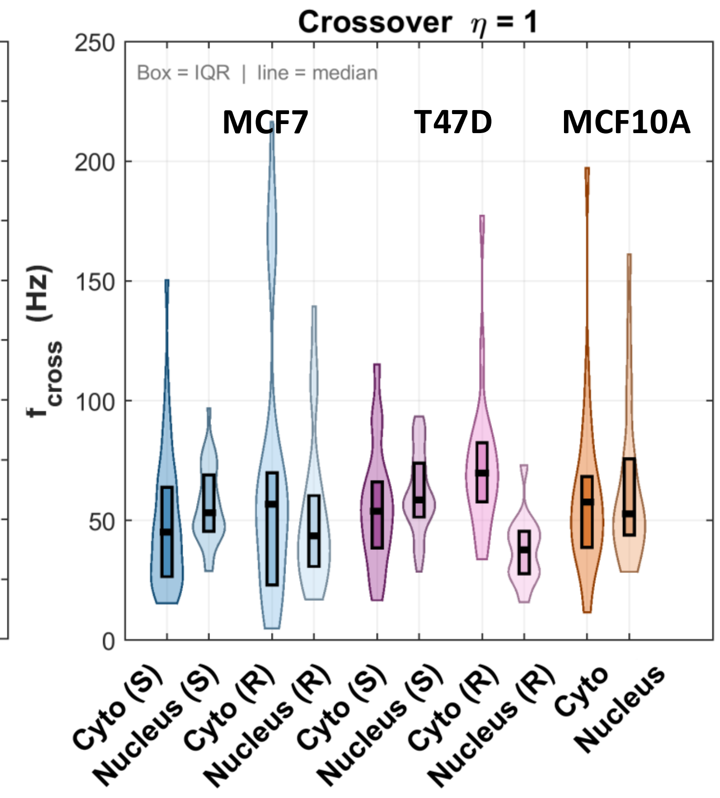




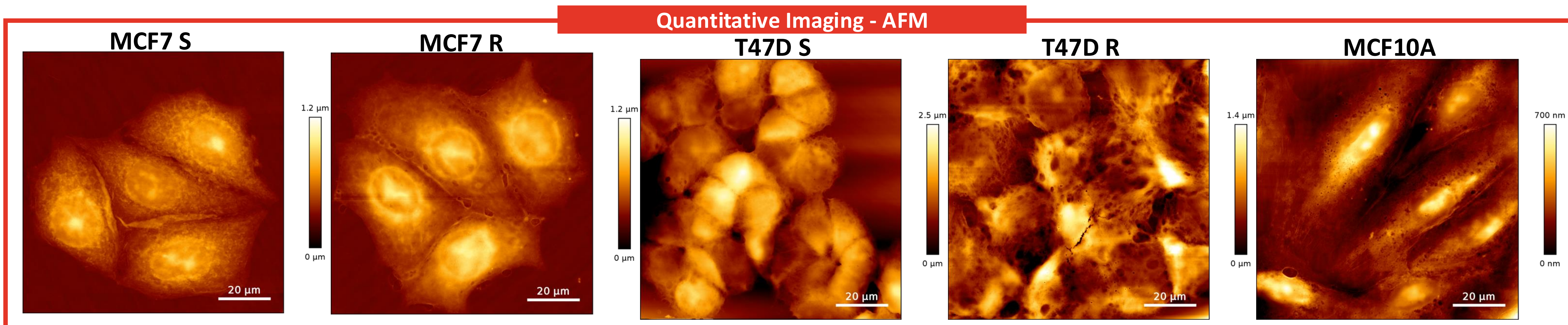








Quantitative Imaging - AFM



Representative **AFM topography images** acquired in **QI-AFM (Quantitative Imaging) mode** of **five breast cell lines**. Cells were seeded on Petri dishes and fixed with 4% paraformaldehyde (PFA) prior to imaging. Scale bar: 20 µm. Color scale represents surface height (dark = low, bright = high). **QI-imaging settings**: peak force setpoint 60 nN, scan speed 300 µm/s, z-length 600 nm, resolution 1024×1024 pixels, scan size 100×100 µm², acquired in air at room temperature using RTESP-300 cantilever (nominal k ≈ 40 N/m).

Conclusion

Take-home: *Palbociclib resistance leaves a measurable, cell-line-specific fingerprint on cell viscoelasticity, detectable by AFM microrheology.*

- HR+ cancer cells (MCF7, T47D) are **softer** than the MCF10A control.
- Resistant variants show **altered cytoplasm–nucleus coupling**, with cell-line-specific crossover shifts.
- The exponent **B** is **modulated upon resistance,** a *reorganization* of the mechanical hierarchy.

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