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Nanomechanics of respiratory cells probed with atomic force microscopy

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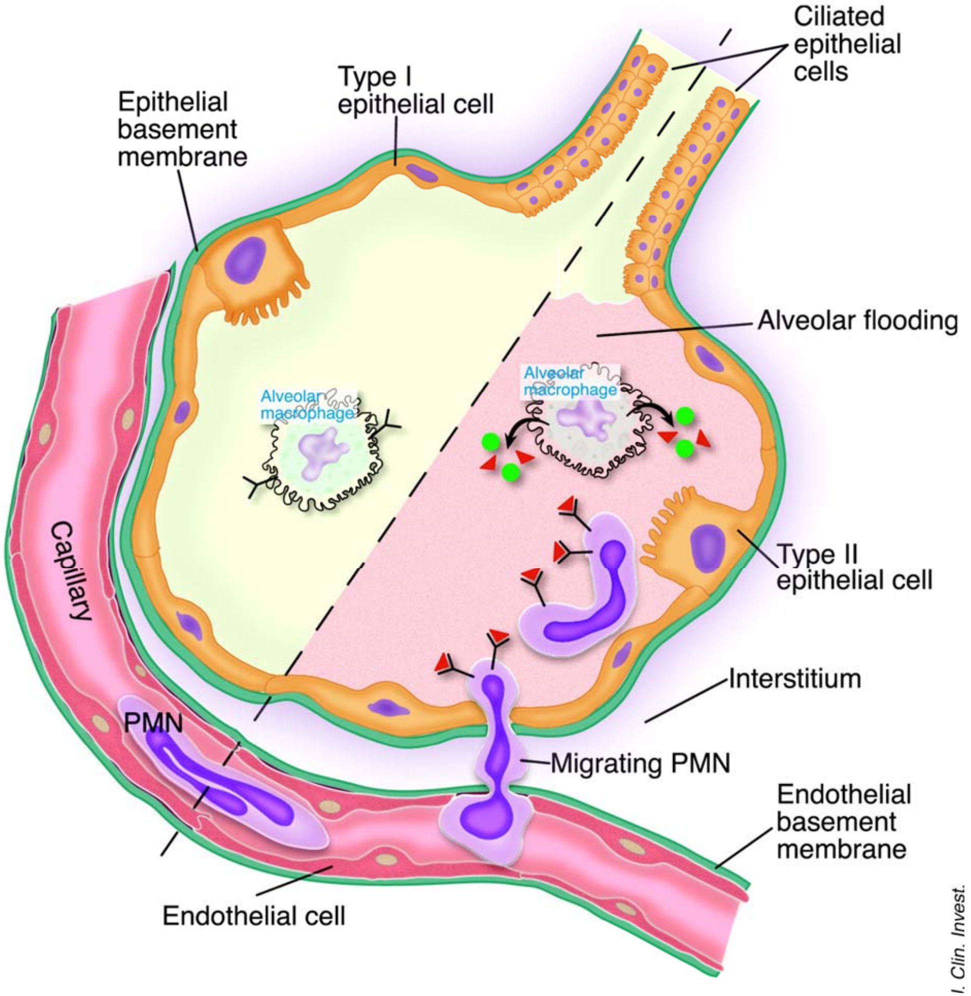
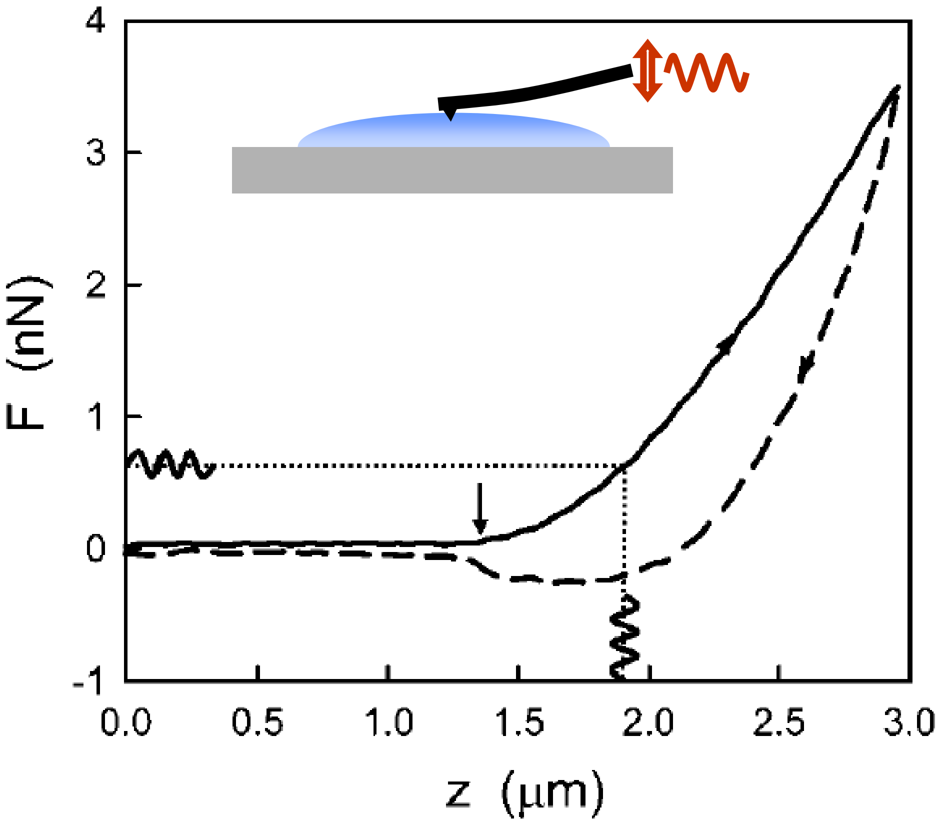
Mechanical properties of the cell play a key role in critical cellular functions



Applying nanotechnology to probe mechanics of respiratory cells

Furthering our understanding of the mechanical behavior of the respiratory system to improve the diagnosis and treatment of respiratory diseases

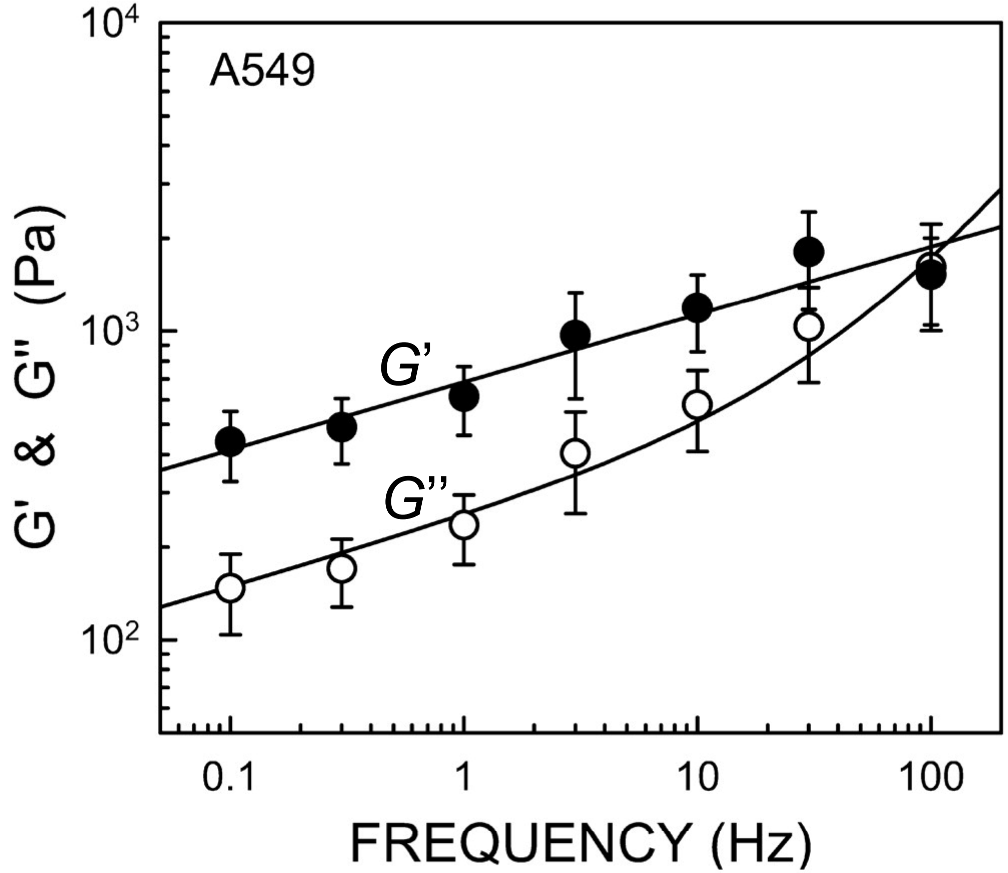
Cell rheology probed by AFM



$$G^*(\omega) = \frac{1-\upsilon}{3\delta_o \tan\theta} \frac{F(\omega)}{\delta(\omega)}$$

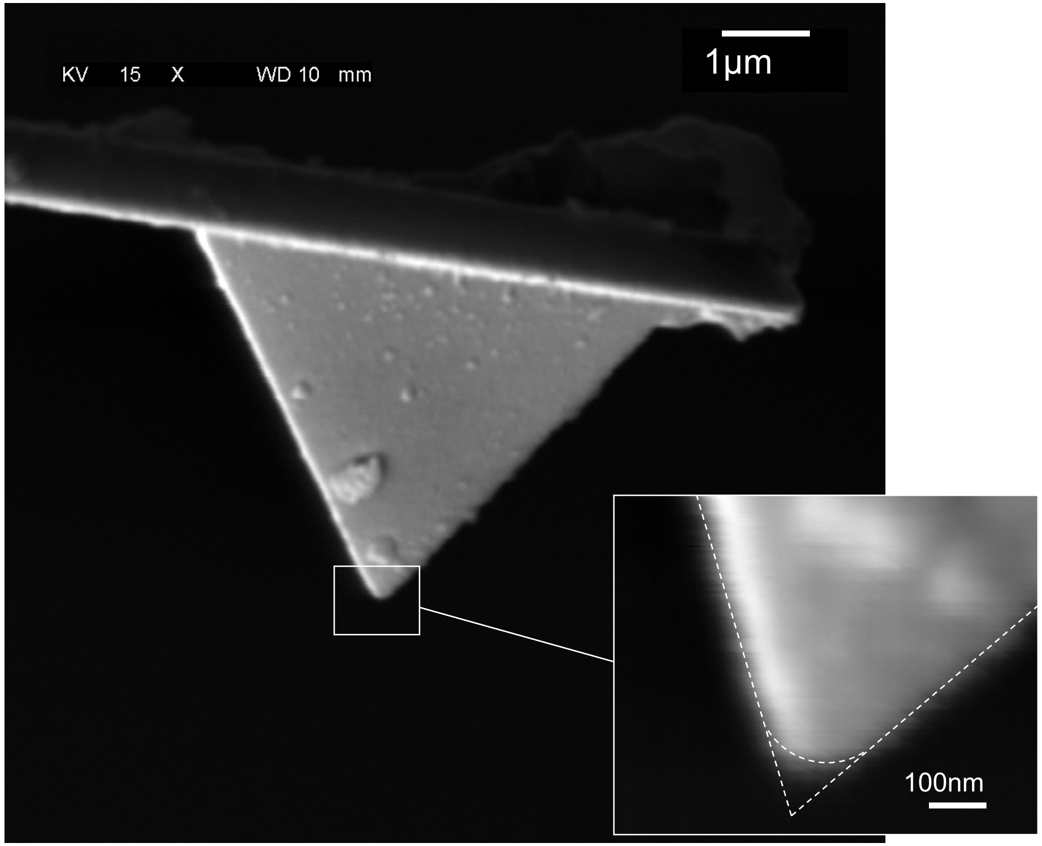
$$G^*(\omega) = G'(\omega) + jG''(\omega)$$

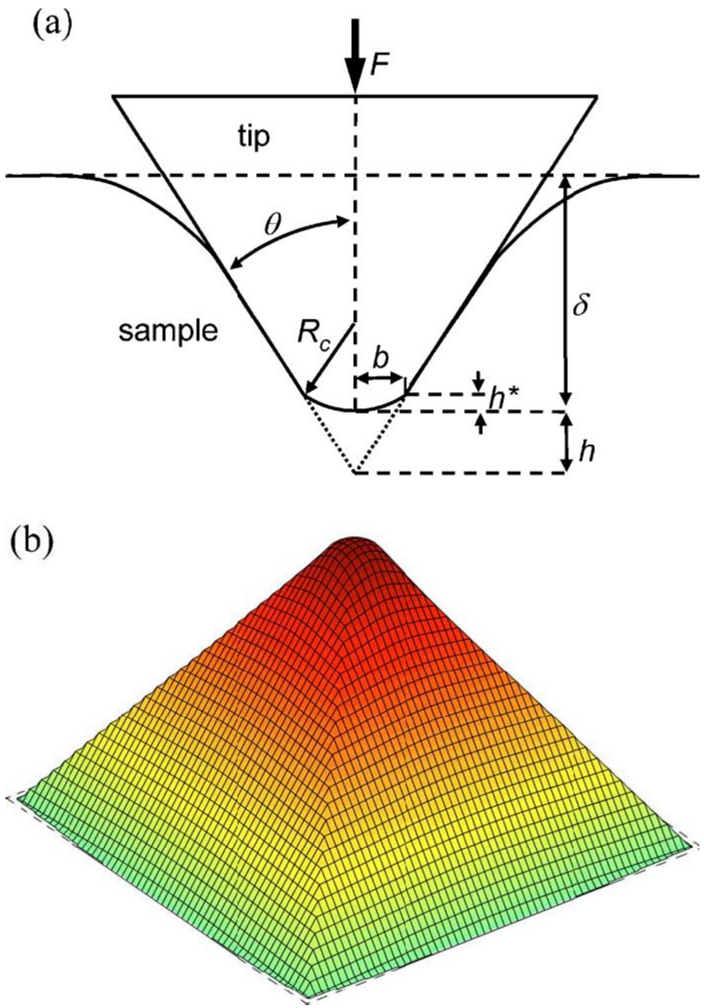
$$\eta(\omega) = G''(\omega) \;/\; G'(\omega)$$



Alcaraz *et al*. *Biophys J* 84:2071-2079, 2003

Contact model of blunted pyramidal cantilever tips



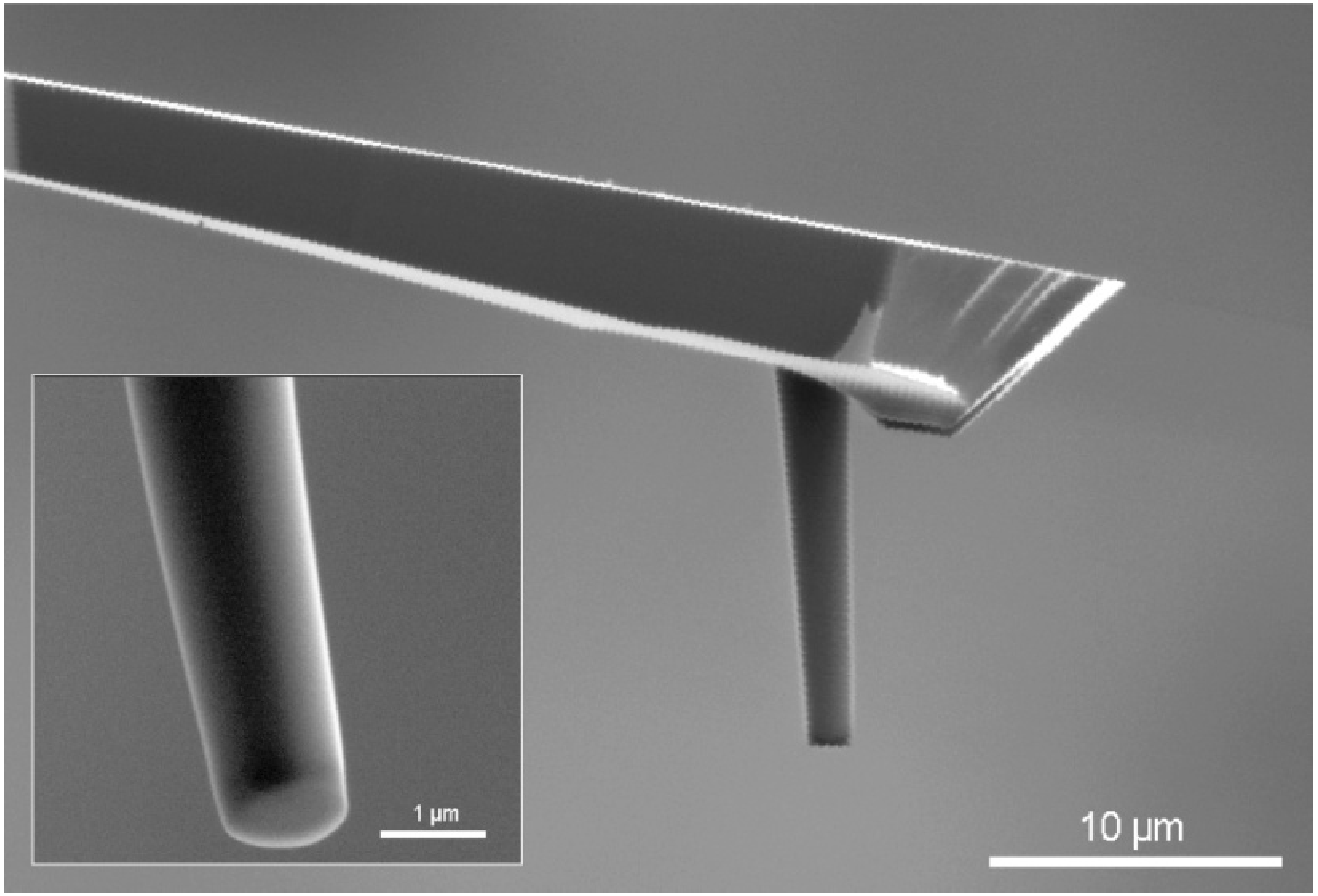


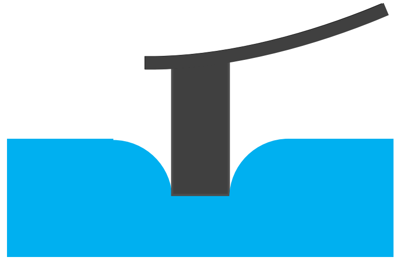
$$F = \frac{2E}{1-\nu^2}\left[\delta a - \frac{2^{1/2}}{\pi}\frac{a^2}{\tan\theta}\left(\frac{\pi}{2} - \arcsin\frac{b}{a}\right) - \frac{a^3}{3R_c} + (a^2 - b^2)^{1/2}\left(\frac{2^{1/2}}{\pi}\frac{b}{\tan\theta} + \frac{a^2 - b^2}{3R_c}\right)\right]$$

$$\delta - \frac{a}{\tan\theta}\frac{2^{3/2}}{\pi}\left(\frac{\pi}{2} - \arcsin\frac{b}{a}\right) + \frac{a}{R_c}\left((a^2 - b^2)^{1/2} - a\right) = 0$$

Rico *et al. Phys Rev E* 72: 021914, 2005

Nanofabrication of flat-ended cylindrical tips using FIB technology

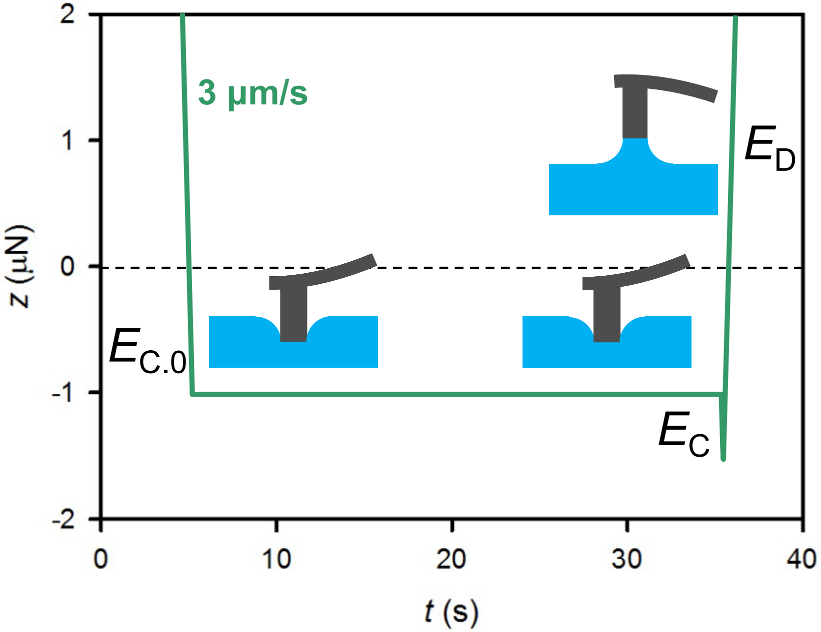


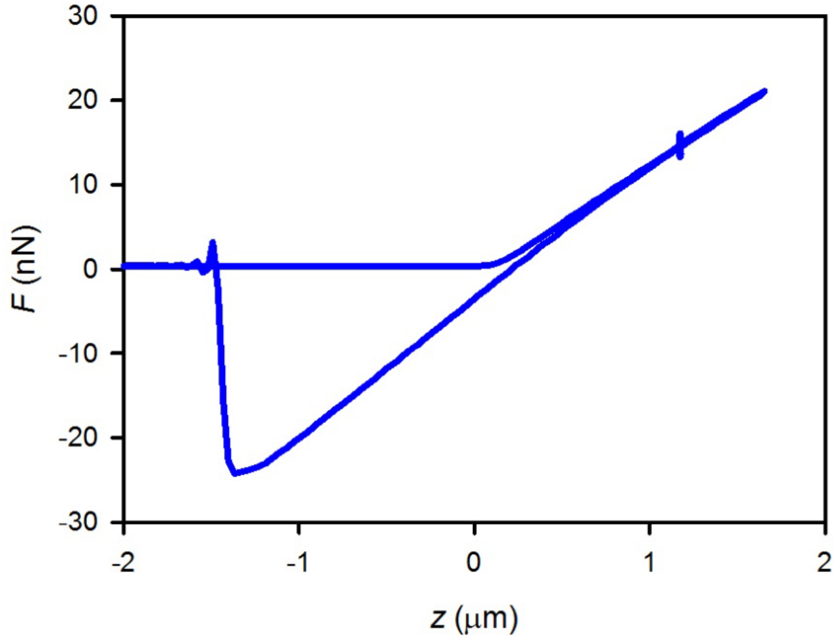


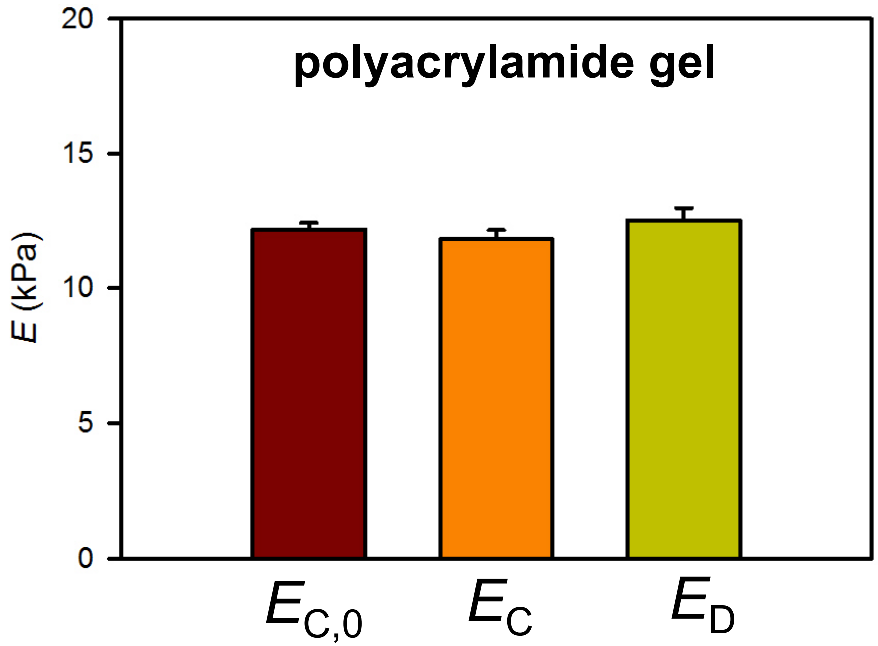
$$F = \frac{2E}{1 - \nu^2} a\delta$$

Rico *et al. J Mol Recognit*, 2007

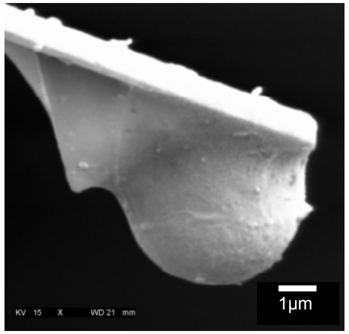
Probing mechanics of gels in compression and distension

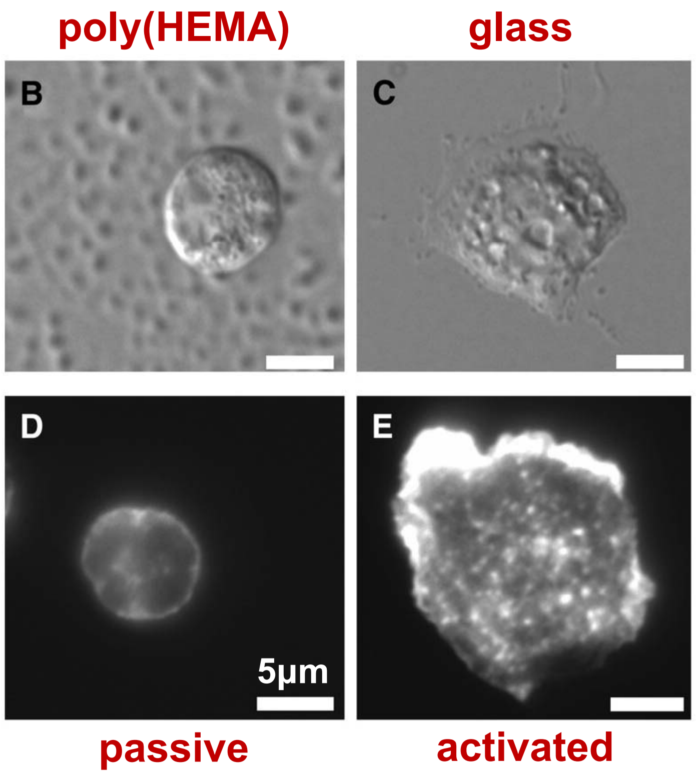






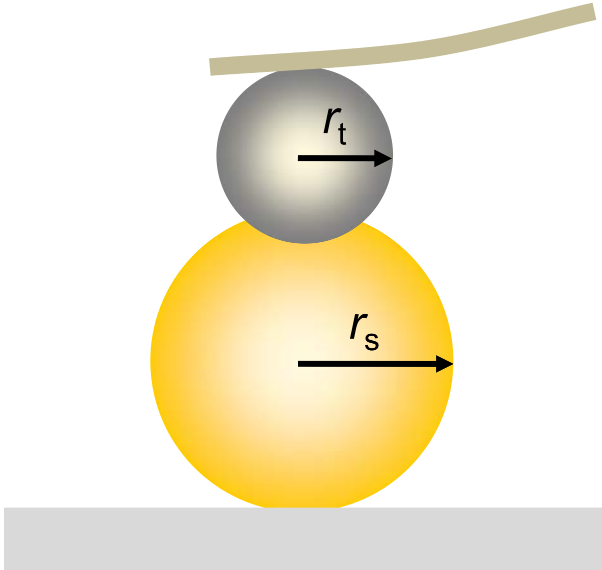
Neutrophil nanomechanics probed with AFM





Contact model of a spherical tip indenting a spherical sample

$$F = \frac{4\,R^{1/2}E}{3\,(1-\upsilon^2)}\,\delta^{3/2} \qquad 1/R = 1/r_t + 1/r_s$$



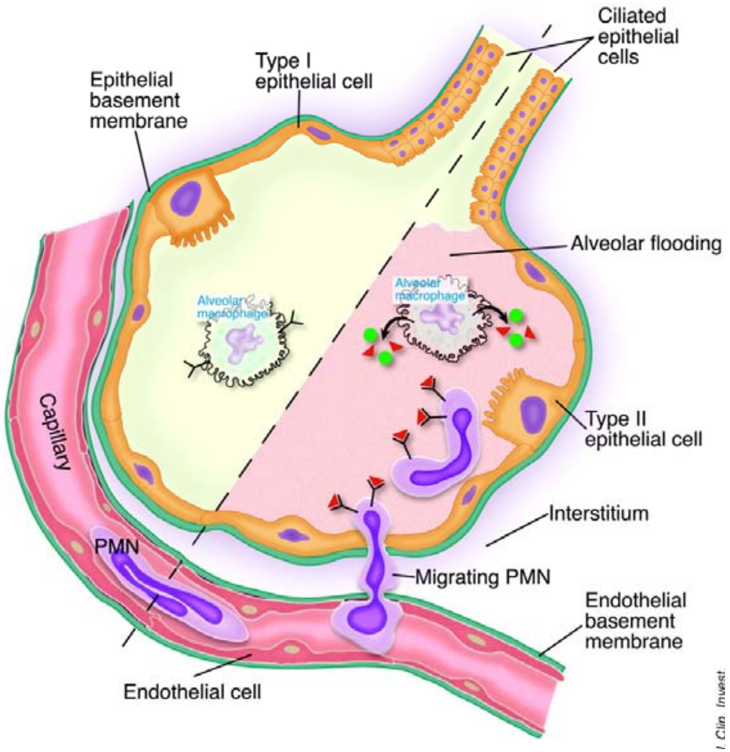
Spherical polystyrene bead $r_t = 2.25$ µm

Neutrophil $r_s \sim 4.5$ µm.

Cell thickness: *F*-*z* in a bare region

Roca-Cusachs *et al*. *Biophys J* 91:3508-3518, 2006

Neutrophil nanomechanics in COPD patients assessed with AFM



COPD is the 4th leading cause of death (5.1%)

In the pulmonary microcapillary circulation, neutrophils (Ø = 6 - 8 μm) must cross a network of narrow 40–100 capillary segments (Ø = 2 - 15 μm).

Neutrophil deformability regulates their passage through the pulmonary capillary bed.

Hypothesis.

The abnormal inflammatory response associated with COPD induces neutrophil stiffening. Cell stiffening slows down the capillary passage, promoting neutrophil pulmonary sequestration and subsequent transendothelial infiltration.

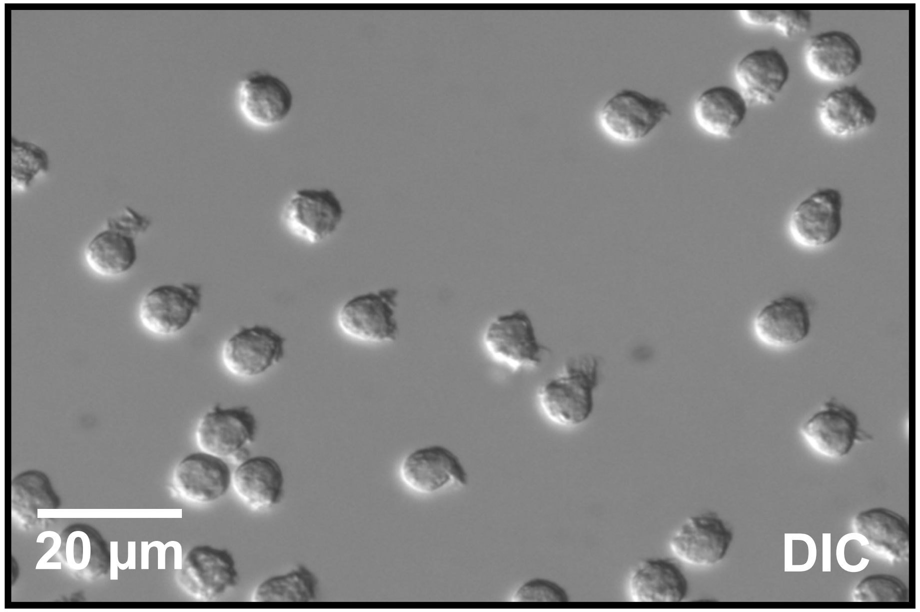
Objective

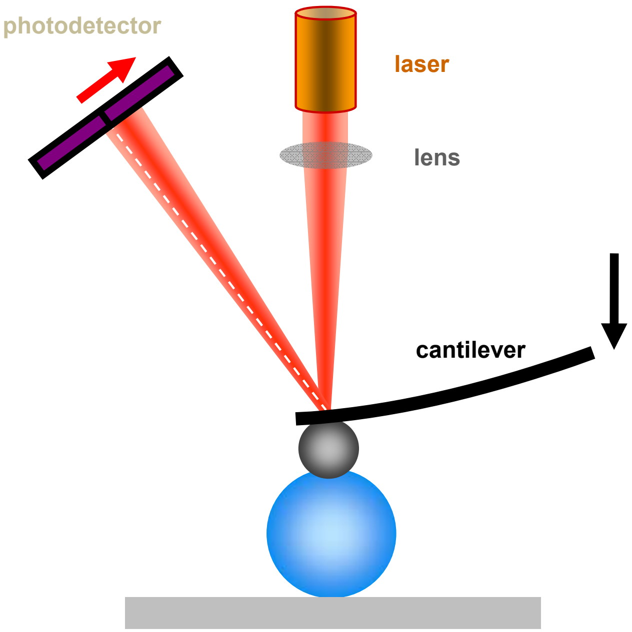
To assess neutrophil mechanics by AFM in patients with COPD.

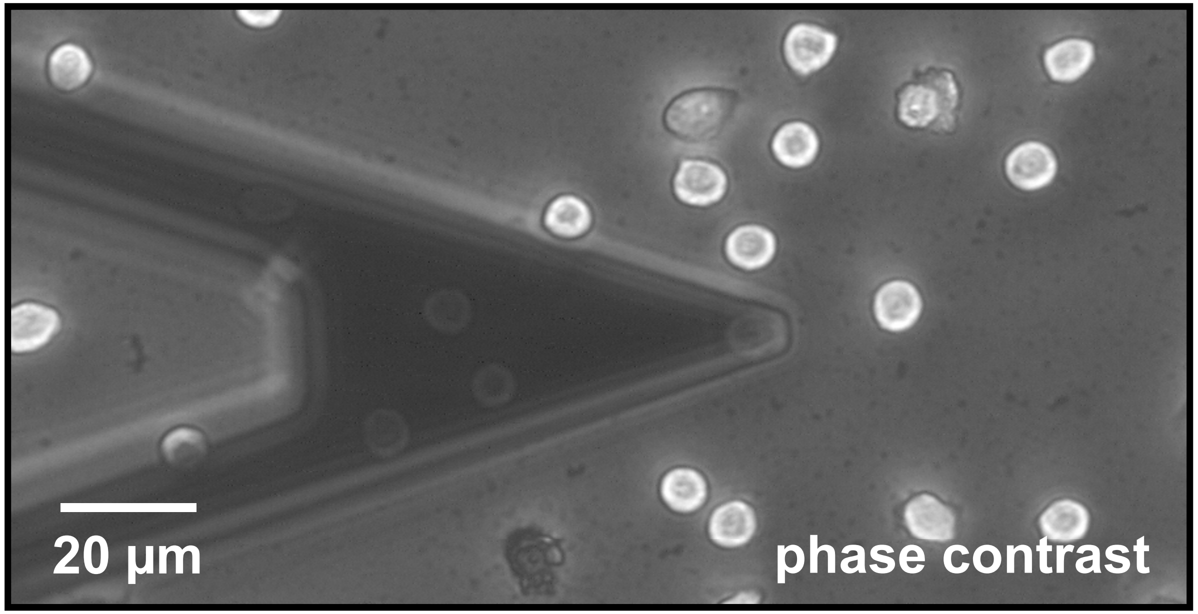
Neutrophil nanomechanics in COPD patients assessed with AFM

AFM measurements.

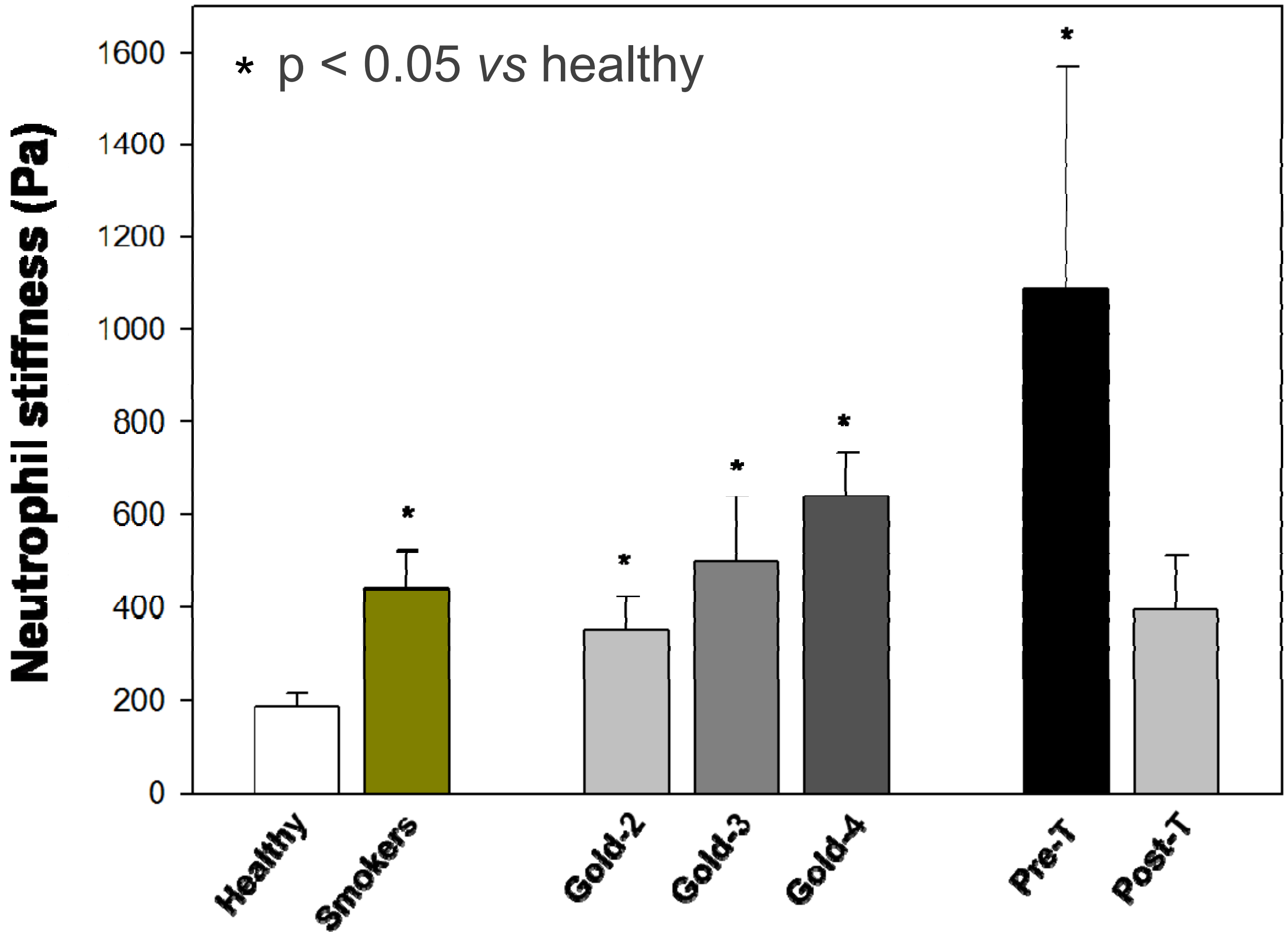
- Netrophils on poly-HEMA (0.5 mg/mL) coated cover-slips
- Spherical tip (Ø = 2.25 μm)
- E computed from F-z curves by fitting two-sphere contact model.



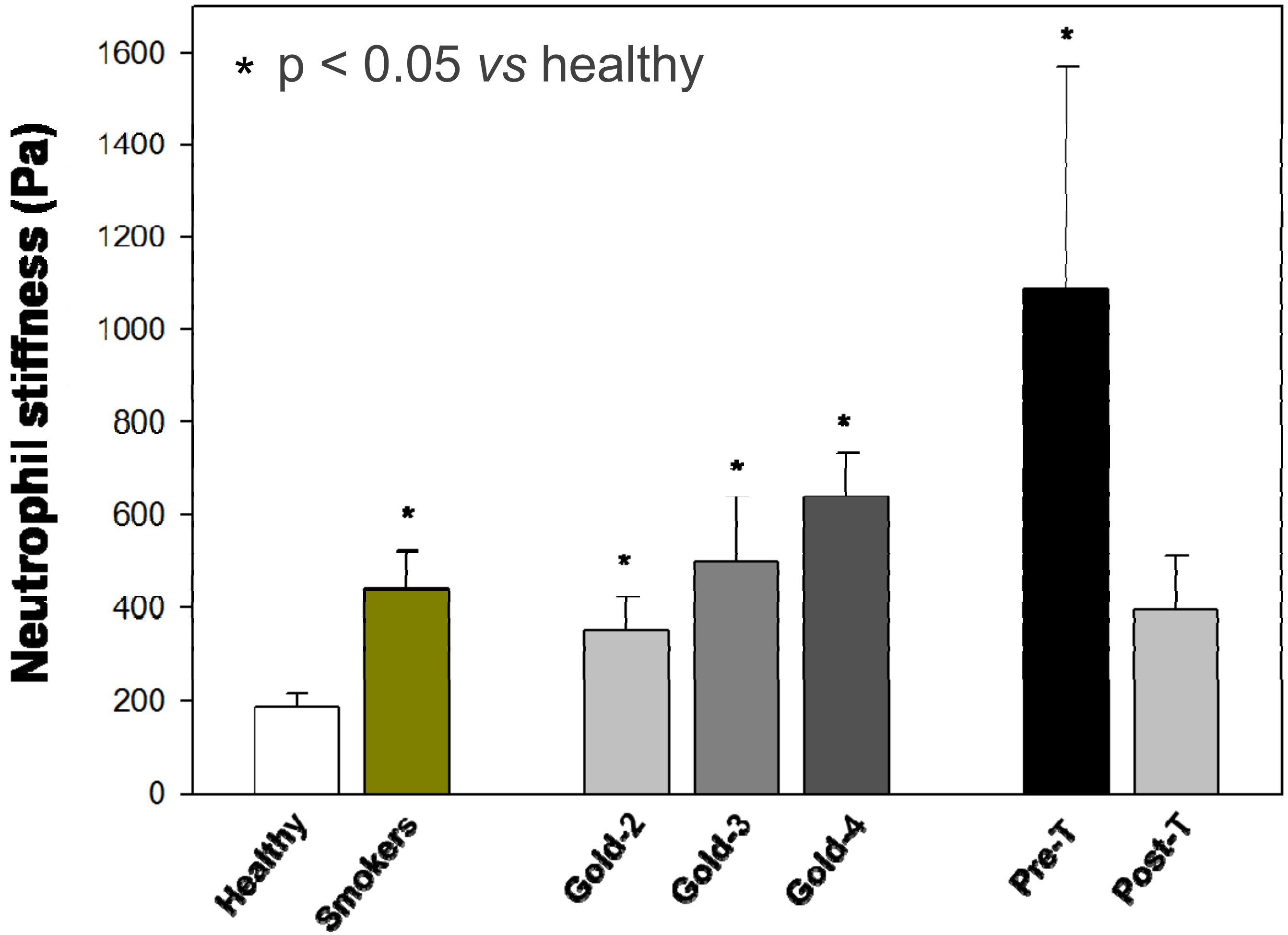




COPD (and smoking) is associated with neutrophil stiffening



Neutrophil stiffness decreases after lung transplantation



COPD induced neutrophil stiffening slows down the capillary passage, promoting neutrophil pulmonary sequestration and subsequent transendothelial infiltration.

Neutrophil nanomechanics in COPD patients

Changes in biomechanical properties of neutrophils reflect the progression of COPD

There is a need for the development of disease activity biomarkers for use in COPD. Neutrophil stiffness could be a useful COPD biomarker.

AFM is a useful technique for assessing neutrophil mechanical abnormalities induced by inflammatory diseases.

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