Determining the Mechanics of Living Cells Using AFM

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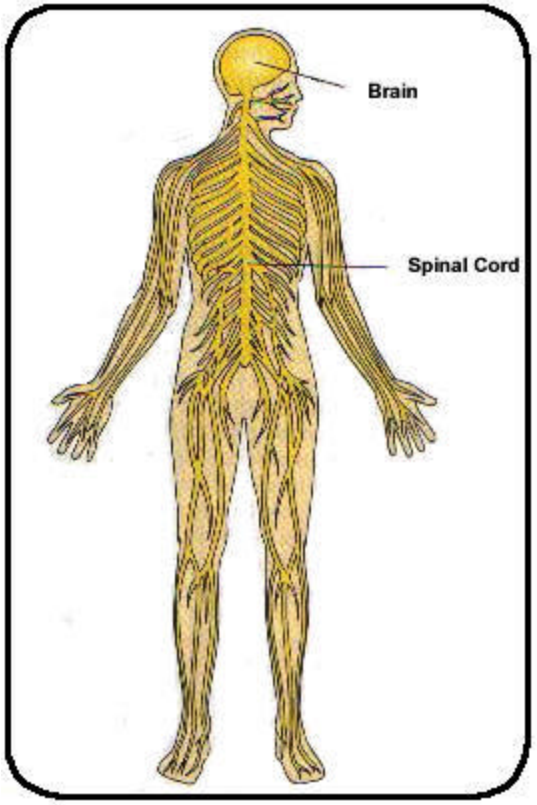
Outline

- Purpose/Applications
- AFM Basics
- Introduction to Asylum AFM
- Elasticity of PDMS
- Nanoindentation of Yeast Cells
- ***Aplysia californica*** nerve cell

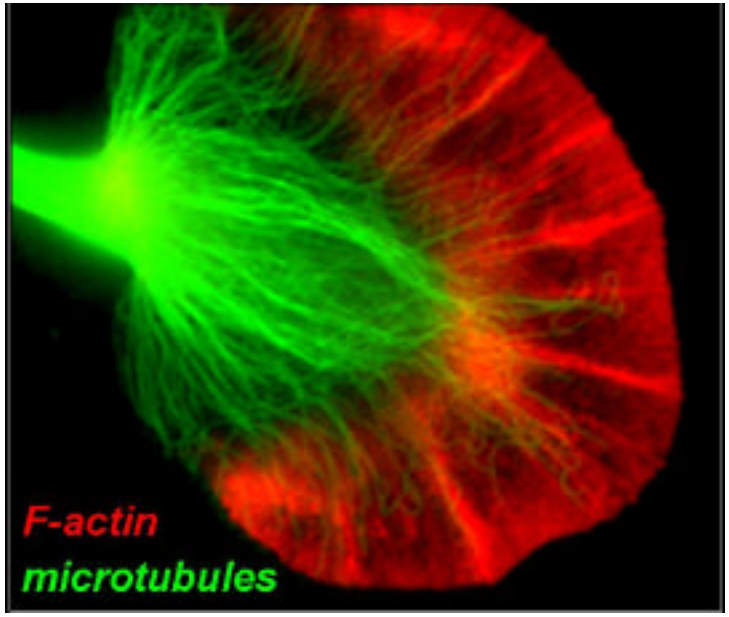
Purpose/Applications



- Understand mechanics of nerve cells
- Develop method of applying forces to cells

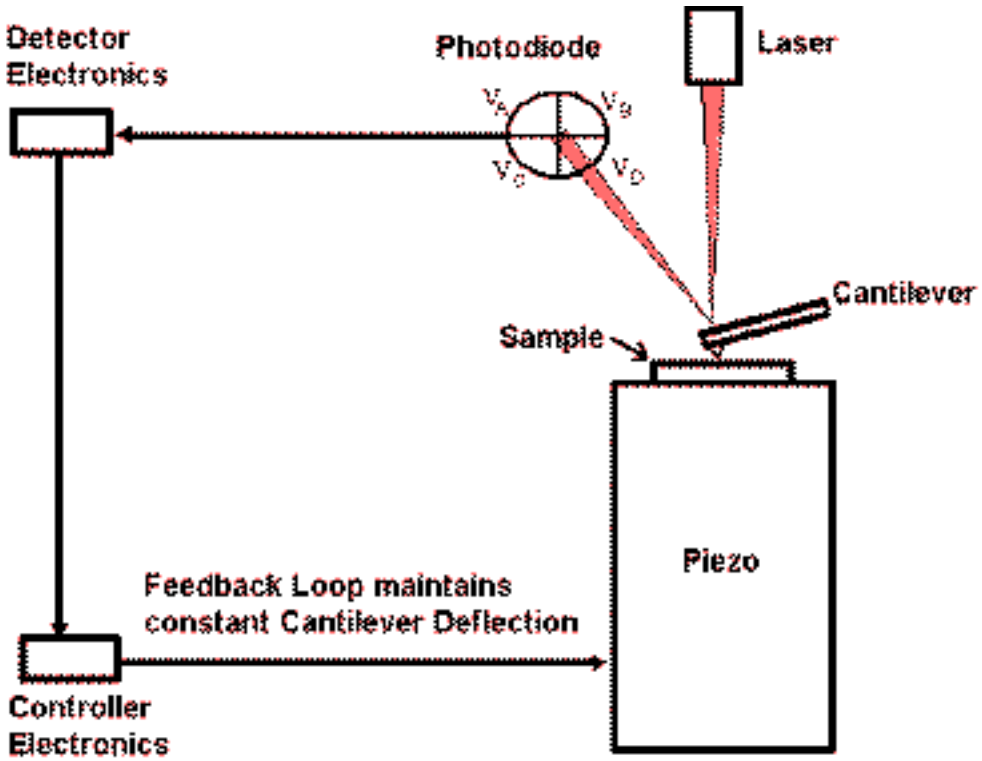
Cell Mechanics of Growth Cone of a Nerve Cell

- Cytoskeleton provides support structure for eukaryotic cells
- Little known of mechanics of the cell.



Prof. Daniel Suter. Purdue University

Atomic Force Microscopy



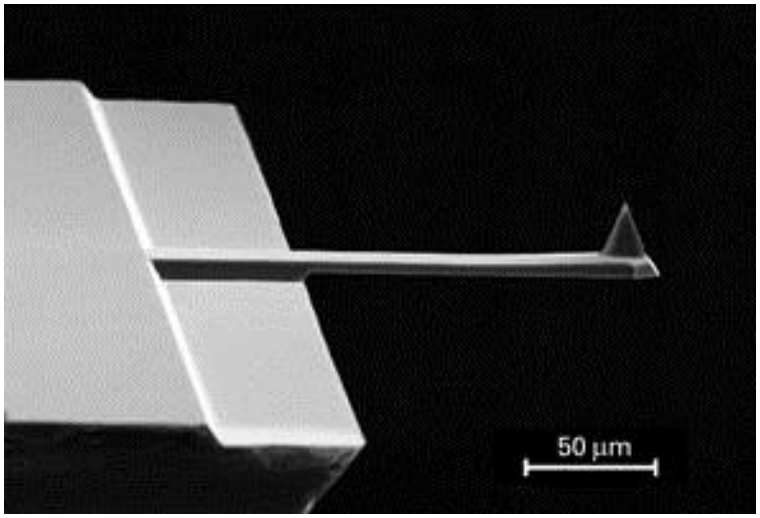
Courtesy of Jin-Won Park

- Sharp tip on cantilever scans surface
- Deflection measured by laser and photodetector
- Uses:
  - Imaging
  - Surface Force Analysis

Asylum AFM

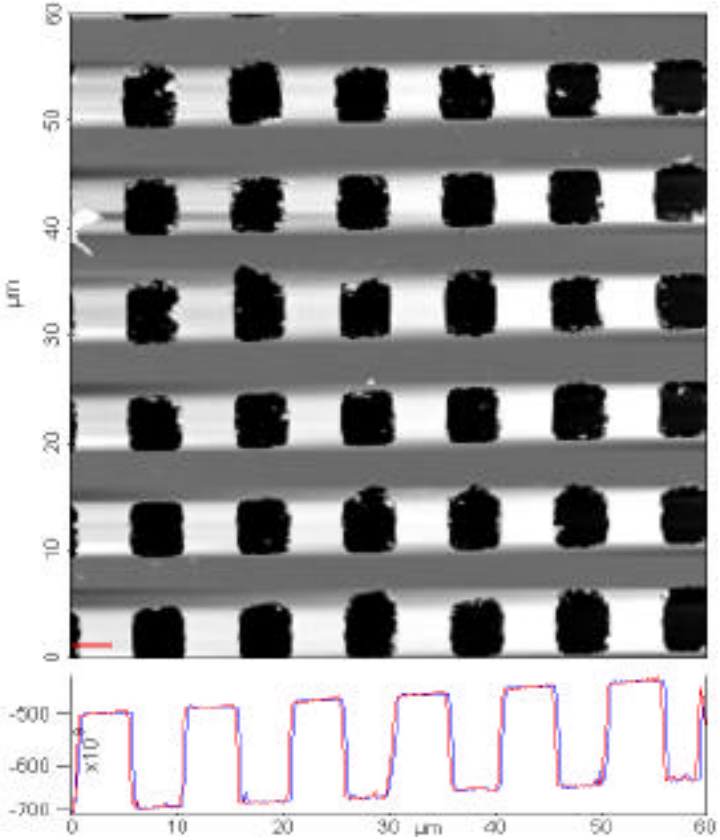
- Shown with Inverted Optical microscope
- x-y piezoelectric scanner in base
- Nanopositioning system corrects for piezo hystersis

Cantilevers



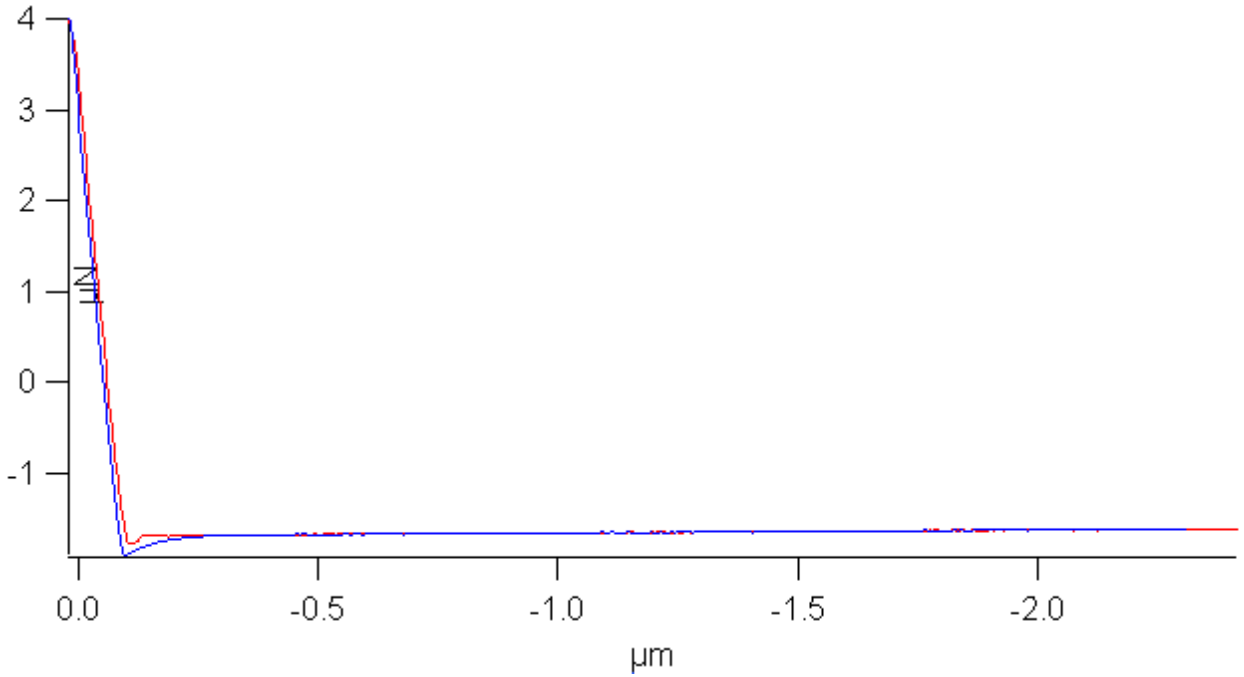
- Made of Si or SiN
- Tip radius ≈ 15 nm
- Calibrate
  - spring constant
  - optical lever sensitivity
  - radius of curvature

Imaging with AFM



- Calibration grid
- 5 _m wells evenly spaced
- 200 nm deep

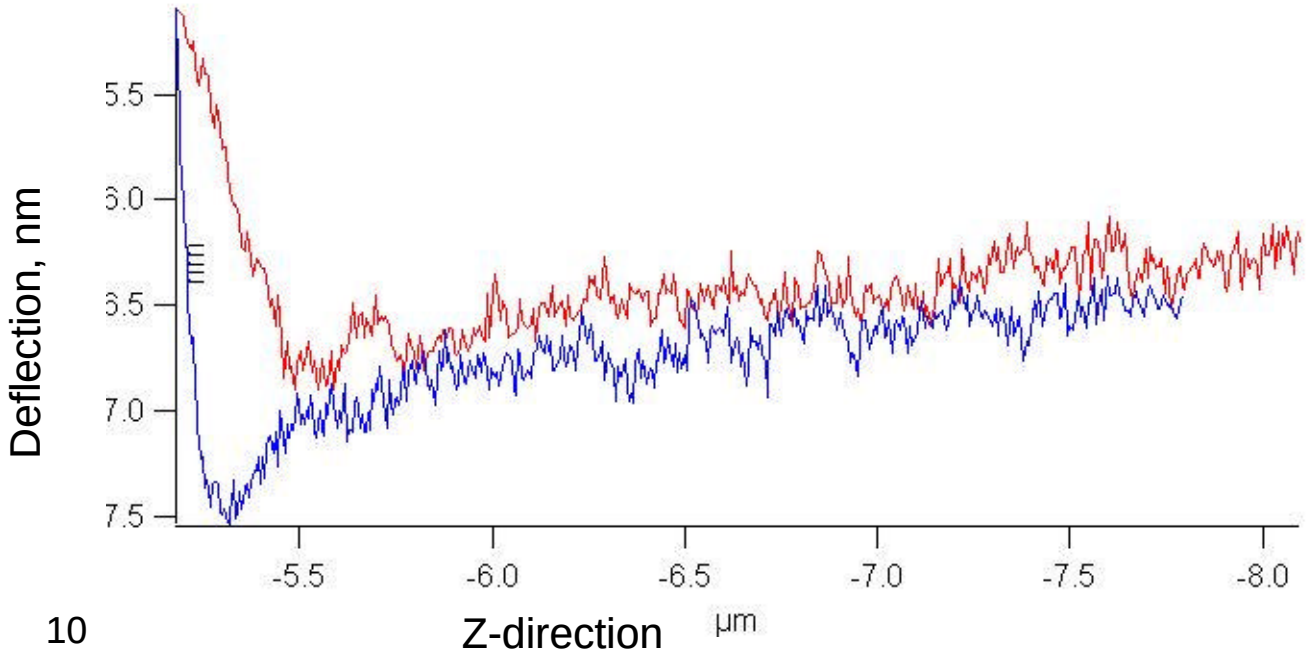
Force Curves



- Deflection vs. LVDT (z-position)
- Typical hard surface force curve
- Used to calculate optical lever sensitivity

Elasticity of PDMS

- Polydimethylsiloxane – thin film on Si wafer
- Calculate Young's modulus by nanoindentation of polymer



Elasticity of PDMS

- Hertzian models

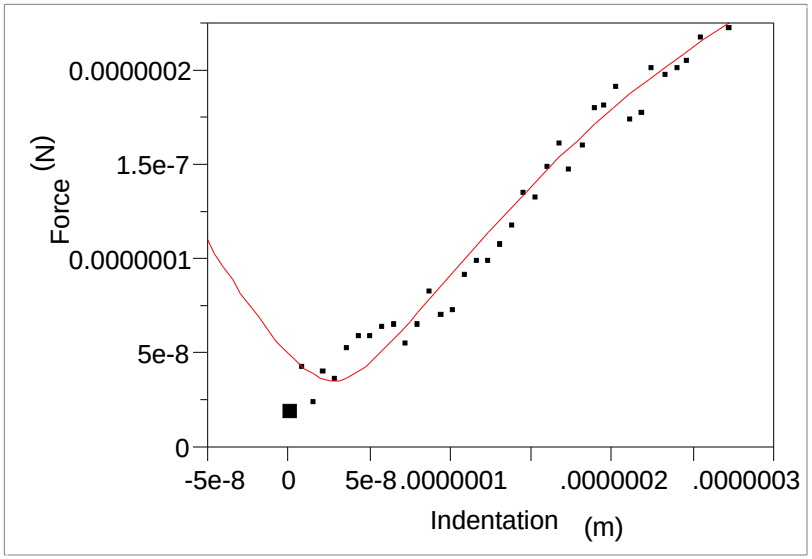
$$F_{cone} = \frac{2}{\pi}(\tan\alpha)E^{*}\delta^{2}$$

$$E^{*} = \frac{E}{1-\upsilon^{2}}$$

$$F_{paraboloid} = \frac{4}{3}E^{*}R^{1/2}\delta^{3/2}$$

Where; _ = half-opening angle, E* = surface elastic constant R = radius of curvature of tip, _ = indentation depth, υ = Poisson ratio, E = Young's modulus

Elasticity of PDMS



- Paraboloid fit (F vs. _$^{3/2}$)

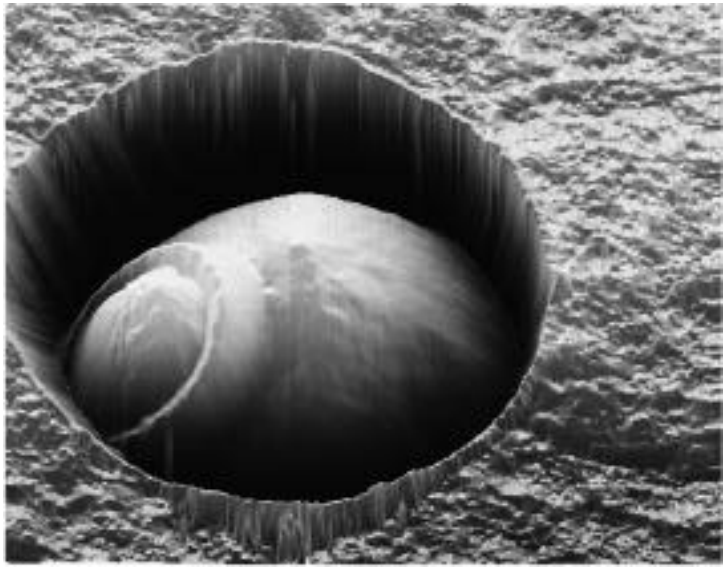
$$F_{paraboloid} = \frac{4}{3} E^{*} R^{1/2} \delta^{3/2}$$

- F = 2963039.8*_$^{3/2}$
- R = 20 nm; υ = 0.5

- E = 6.85 MPa

Nanoindentation of Yeast

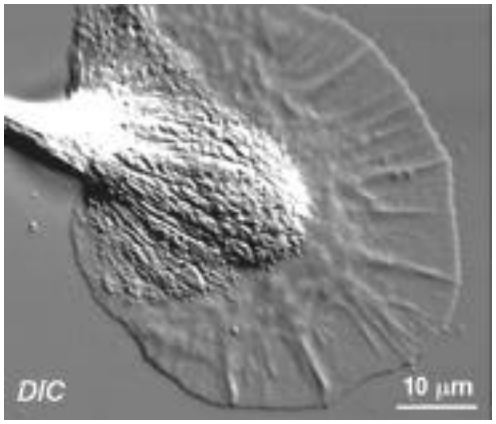
- ***Saccharomyces cerevisiae*** – Brewer's yeast
- Cell wall = stronger surface than animal cell membranes
- Trap yeast in nanoporous membrane



Touhami, et al. Langmuir 2003, 19:4539.

Aplysia californica

Growth cone of neuron



Sea slug

- Cells easily maintained at room temp
- Actin filaments in growth cone
- Land tip on growth cone and examine forces exerted by cell.

Acknowledgements

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