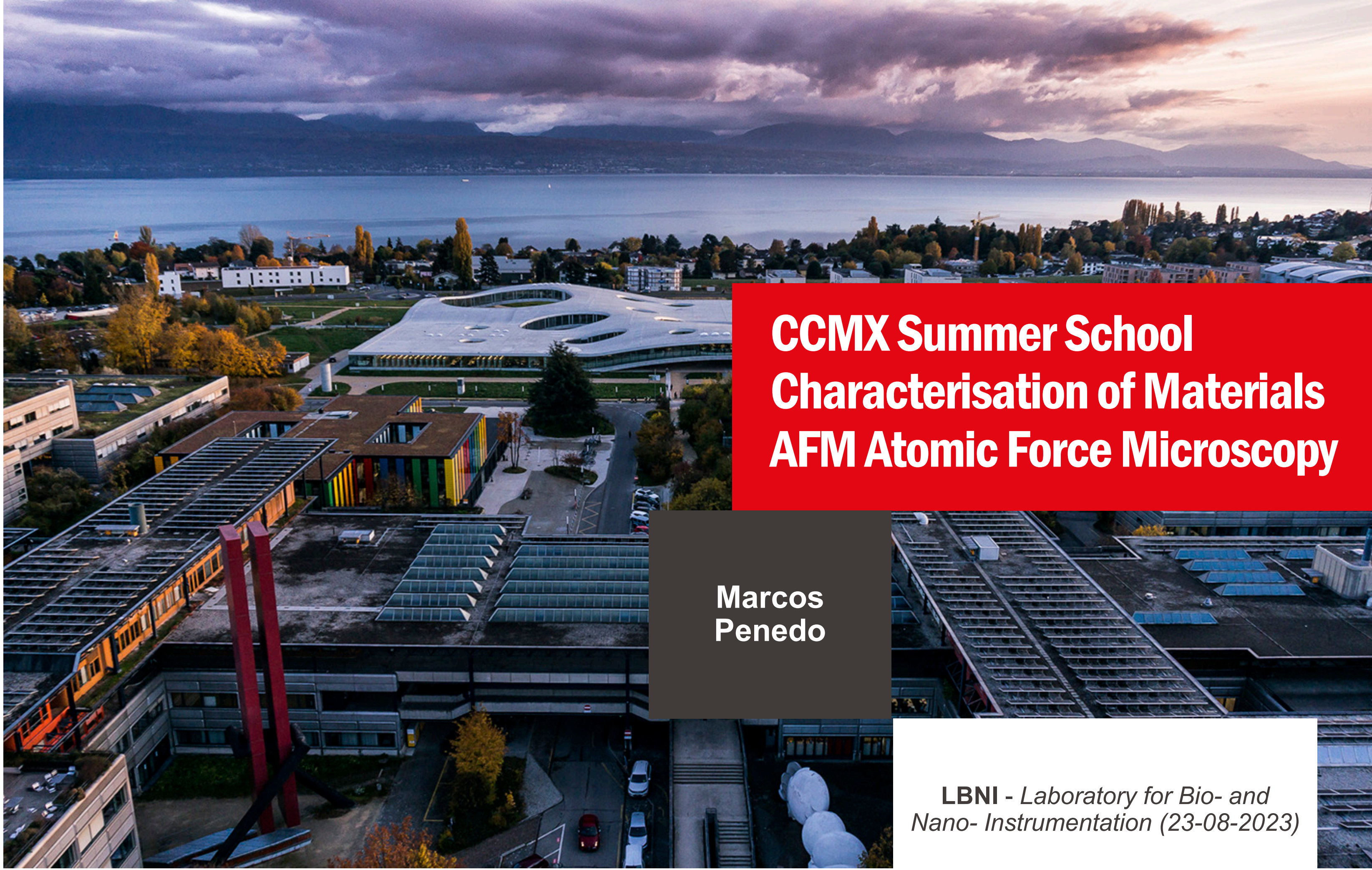
EPFL



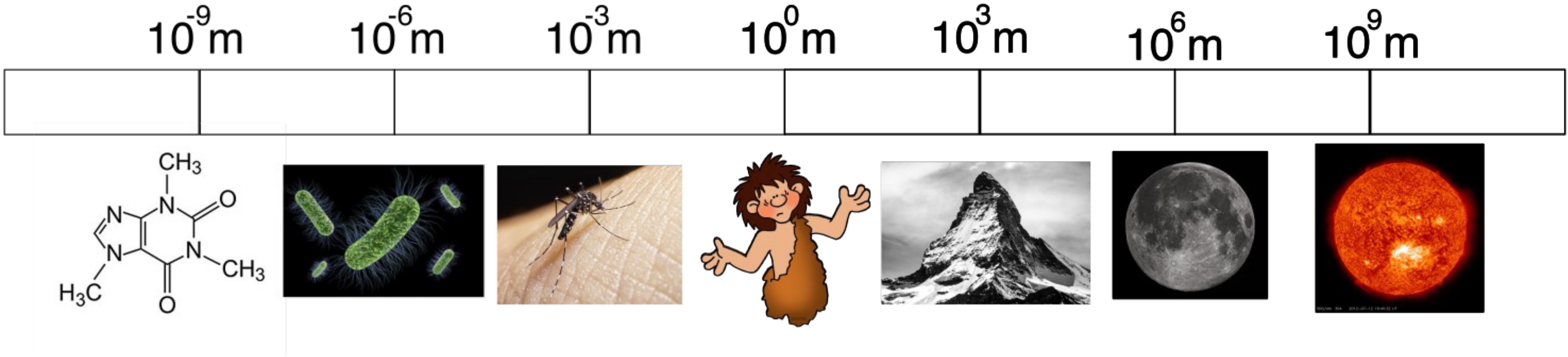
CCMX Summer School
Characterisation of Materials
AFM Atomic Force Microscopy

Marcos
Penedo

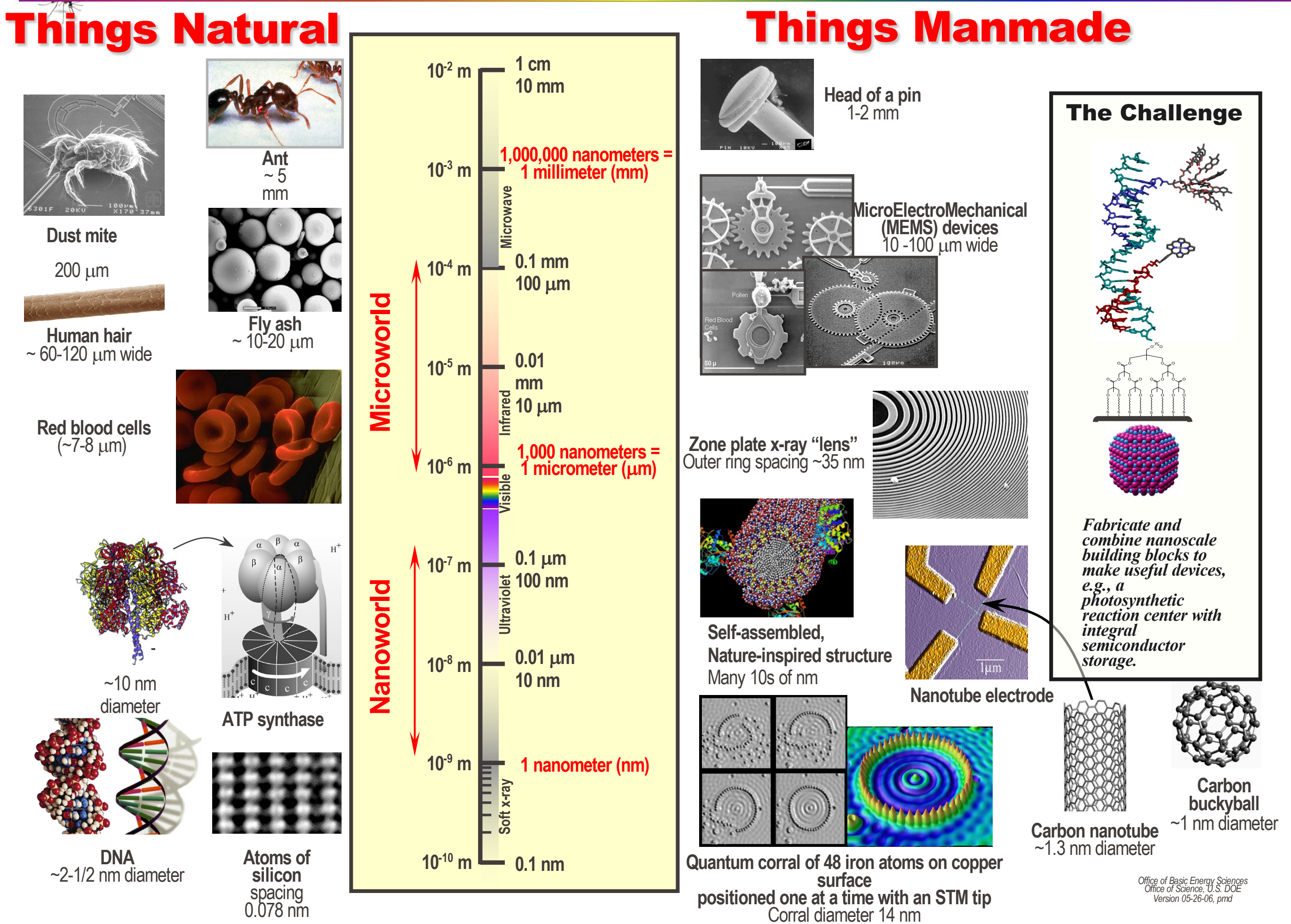
LBNI - *Laboratory for Bio- and Nano- Instrumentation (23-08-2023)*

École
polytechnique
fédérale
de Lausanne

Why we need nanoscale microscopy



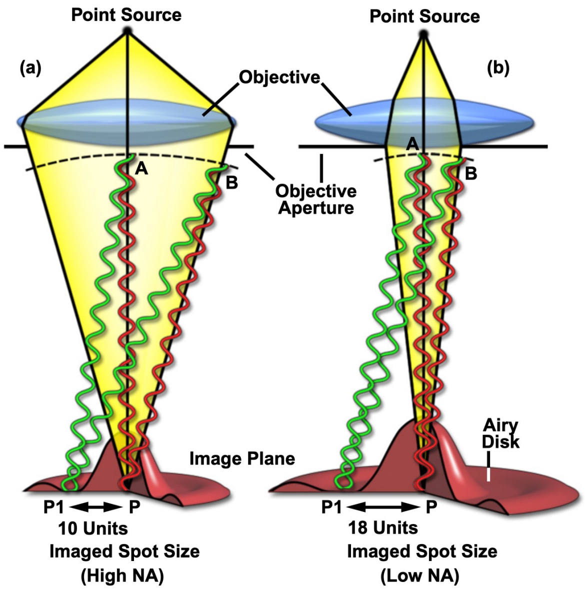
The Scale of Things – Nanometers and More



Why is it difficult to measure small things?
The diffraction limit

In any (far field) microscopy system where we create a magnified image of an object via an image projection using diffractive elements (such as lenses) we run into the *diffraction limit:*

Point sources (with zero size) are projected to an Airy disk with a certain size. Two-point sources that are close together will result in two Airy disks close together. If the disks are too close together, they can no longer be separated based on their intensity. That is then the resolution limit of the microscope.



What determines the achievable resolution

$$\textit{Abbe Resolution}_{x,y} = \frac{\lambda}{2NA}$$

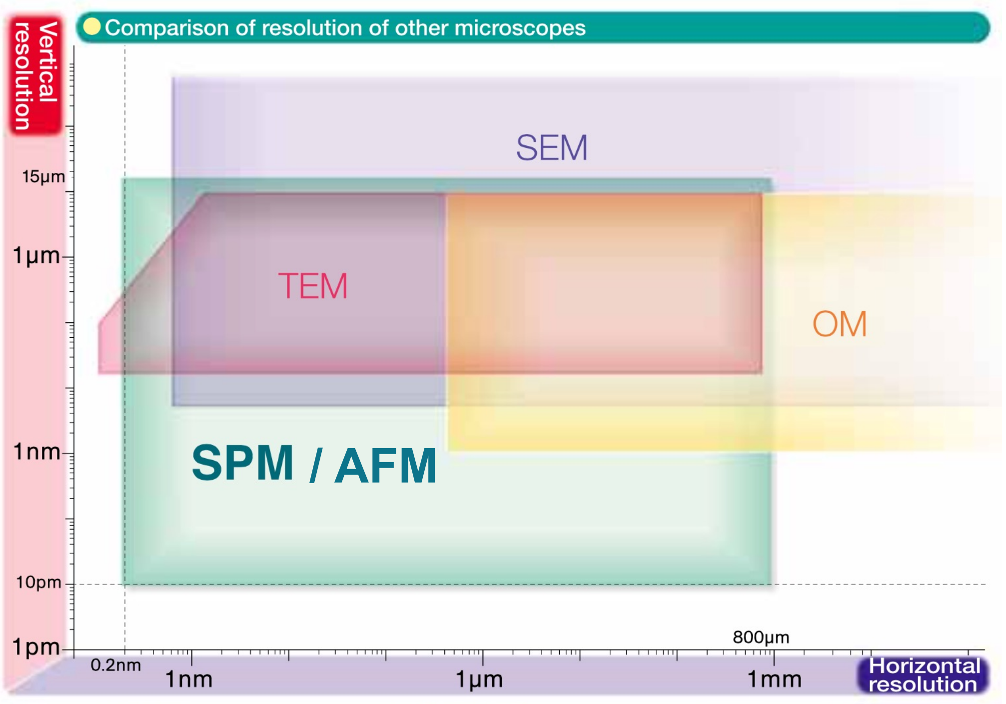
- λ... wavelength
- NA ... numerical aperture

What can we do to get around this?

- Work with smaller wavelengths: instead of photons use particles with much smaller wavelength (such as electrons: de Broglie wavelength of an electron with acceleration voltage of 10kV = $1{,}22\cdot10^{-11}$m, which is 40’000 times smaller than that of a photon). That is what we use in electron microscopy
- Try to use non far field microscopy techniques (near field techniques or scanning probe techniques). This is what we do in atomic force microscopy (AFM), scanning tunneling microscopy (STM) or scanning near field optical microscopy (SNOM)

Resolution is NOT everything…

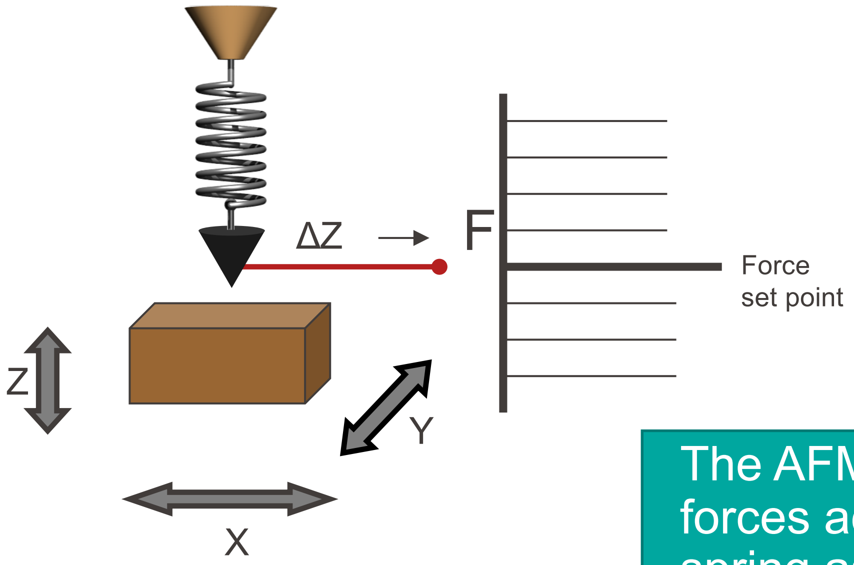
…but it's sure nice to have a good one



What is an AFM?

(don’t be fooled by the word *atomic*)

“Scanning force microscopy“ SFM

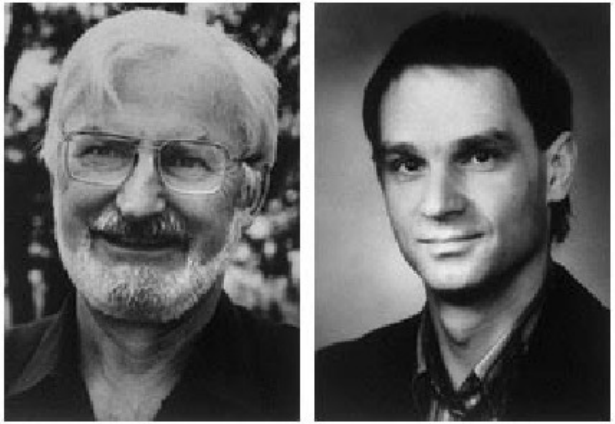


The AFM measures the effect of forces acting on the sharp tip on a spring as a function of the position on the surface. – ***sometimes*** these forces are due to topography

It all started with *Tunneling*…

- Binnig, Gerber, Rohrer, Wiebel *Tunneling through a controller vacuum gap.*(Applied Physics Letters **40**, 178 (1982)

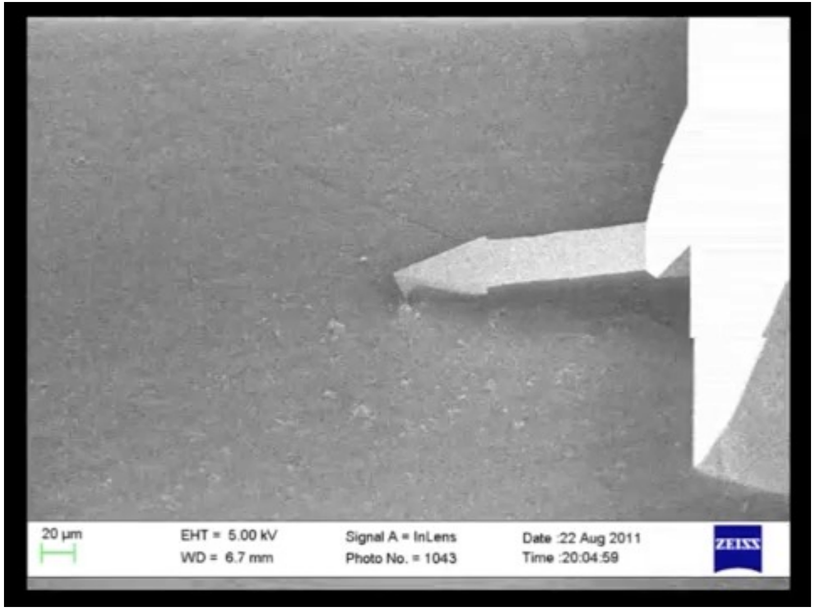
- *“This investigation is the first step towards the development of scanning tunneling microscopy, where the surface is scanned by a tunnel current and should open the door to a new area of surface studies.”*



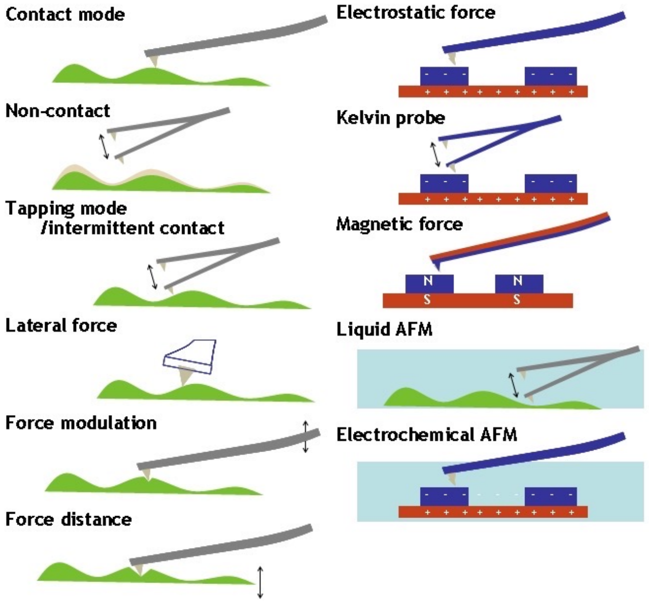
Scanning tunnelling microscopy was invented by Gerd Binnig (right) and Heinrich Rohrer (left) in 1981. They were awarded the Nobel Prize in 1986.

…but only for conducting samples!

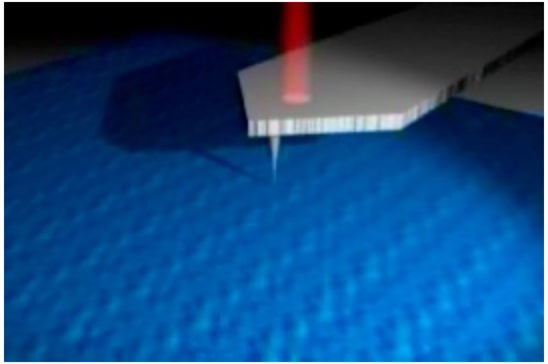
Atomic force microscopy (AFM)



From www.zeiss.com

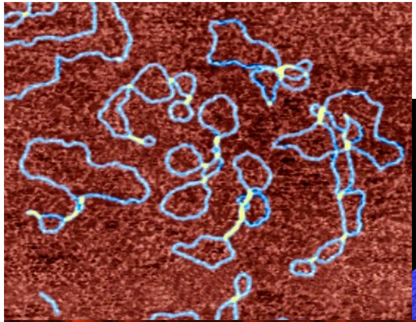


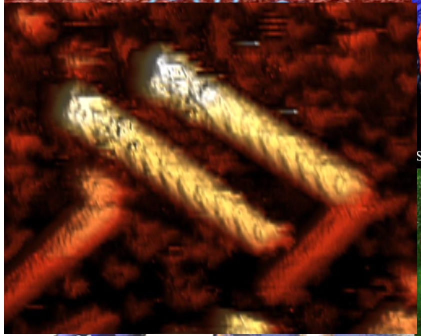
Atomic force microscopy

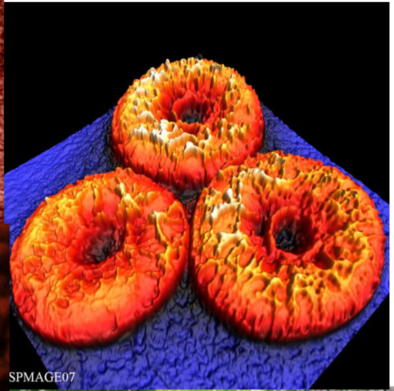


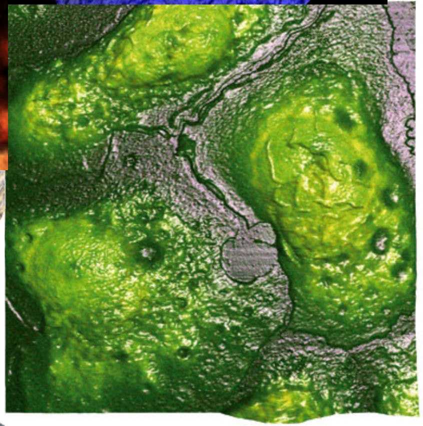
A versatile tool for nanoscale measurements…

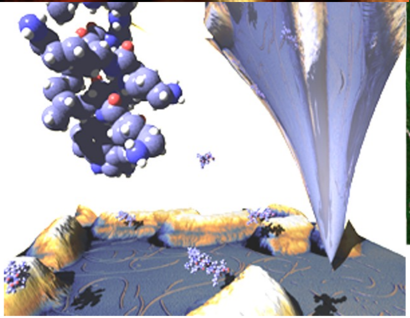
- Single molecule resolution
- High resolution imaging in aqueous solution
- Nanomanipulation
- Single molecule mechanics
- Imaging of living cells





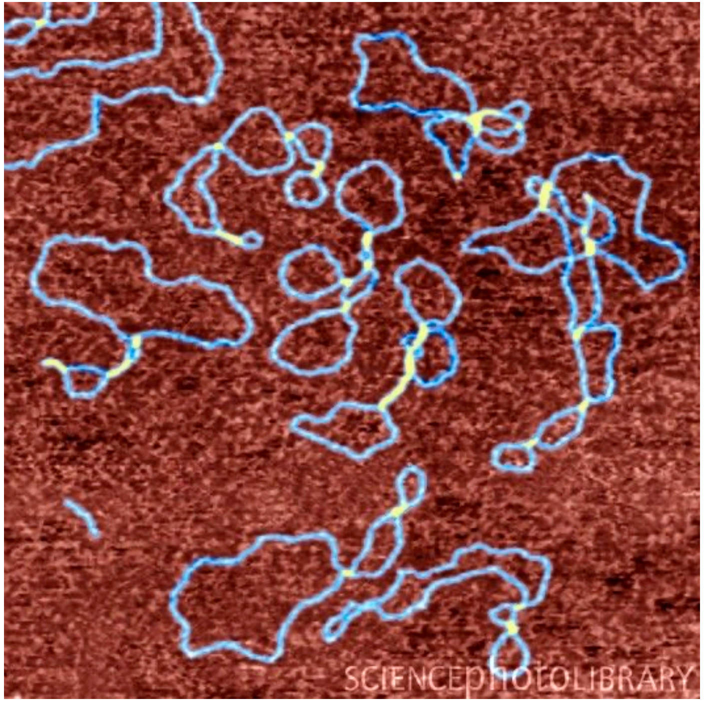






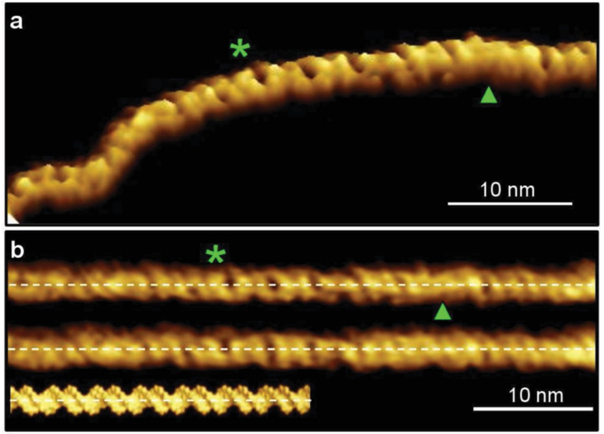
Single molecule resolution

Plasmid DNA on mica



Source: SciencePhotoLibrary

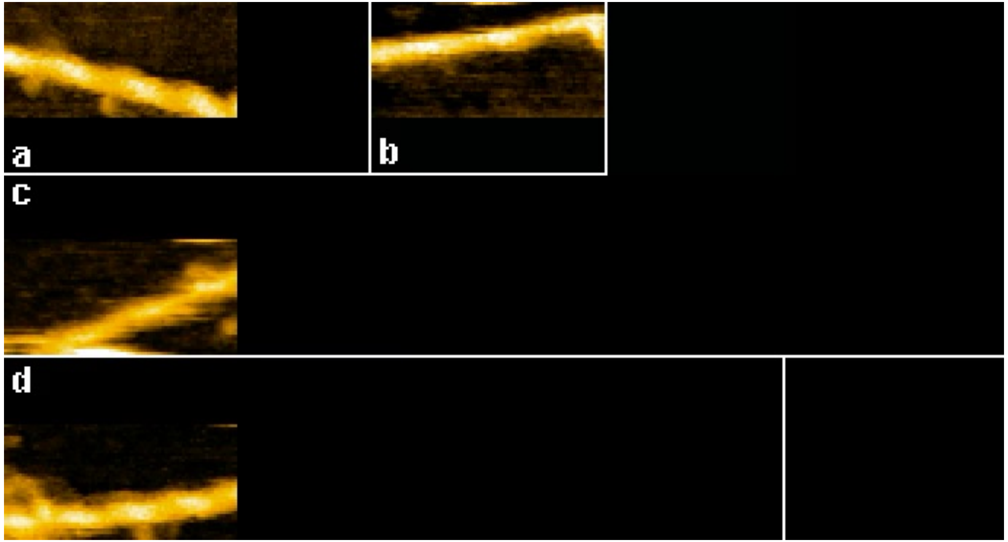
- Single molecules can be easily resolved
- Even the double helix!



Pyne et al. *Small, 10, Nr16, 2014*

Single molecule resolution

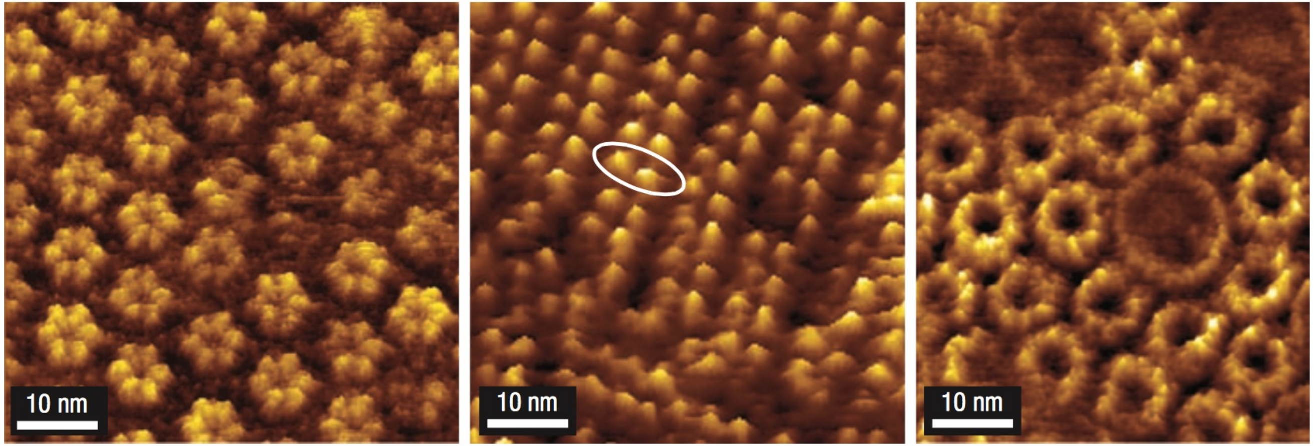
Walking myosin V on actin fibers



*Nature 468, 72 (**2010**)*

High resolution images in fluid of proteins

- Imaging of membranes and membrane bound proteins
- Imaging of live cells



From Review Nature Nanotechnology 2008,D. Müller and I. Dufren,

AFM can be used for nanomanipulation

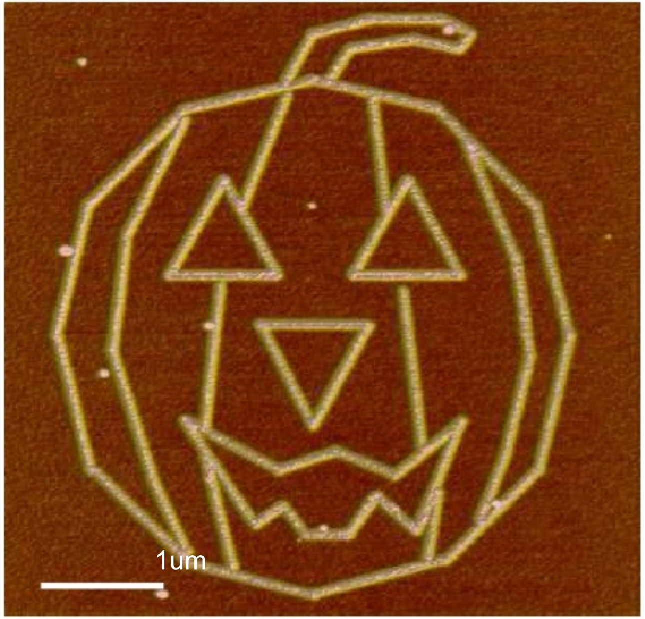


Image from:
http://www.veeco.com/library/nanotheater

- AFM patterning of a silicon surface using anodic oxidation
- Other approaches have been developed such as
  - dip-pen nanolithography and
  - Thermal scanning probe lithography (tSPL)

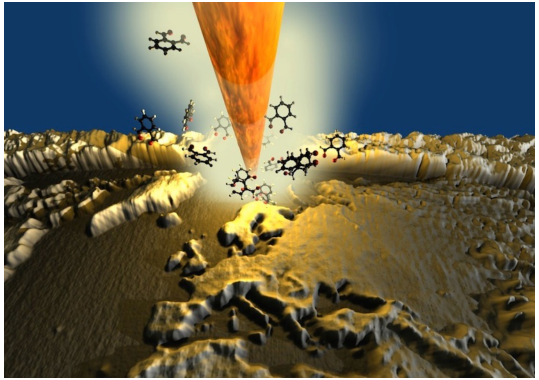


Image from: https://www.swisslitho.com

Different types of scanning probe microscopes

- SPM = scanning probe microscopy
- AFM= Atomic force microscopy (AFM), also known as
- SFM =scanning force microscopy
- STM scanning tunneling microscopy
- …
- SSETM = scanning single-Electron transistor microscopy

Wikipedia lists 41 different SPM modes!

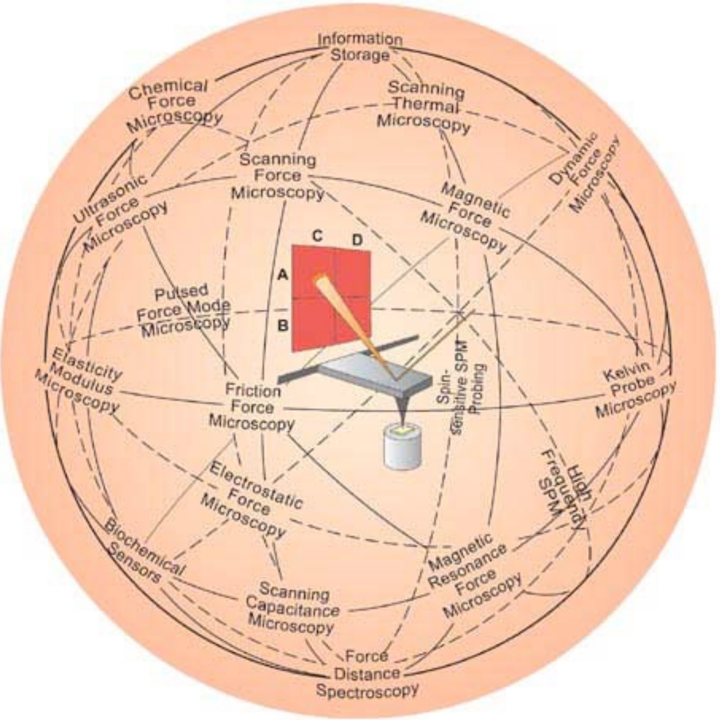
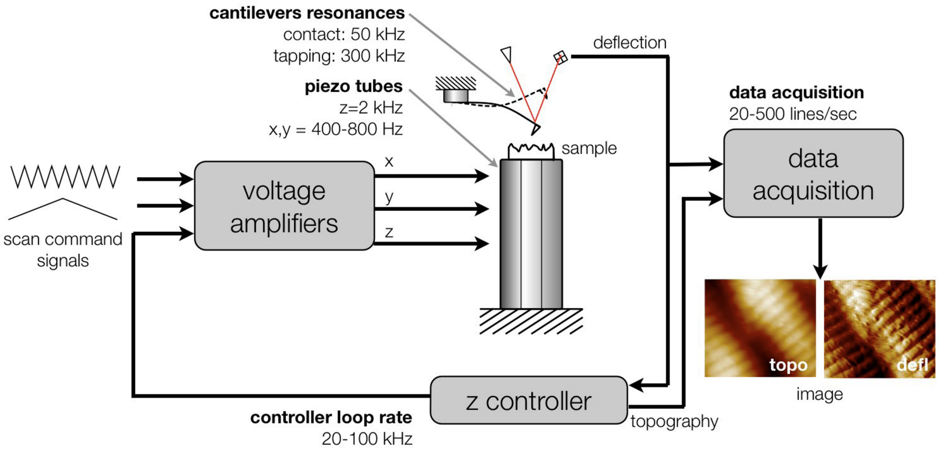


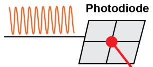
image: Christoph Gerber; copyright Nature Publishing Group

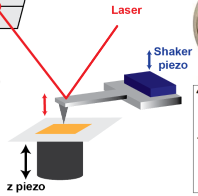
What's in an AFM?

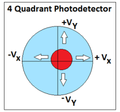


Photodiode (PSD)

- Converts light into an electrical current
- It will detect the cantilever deflection





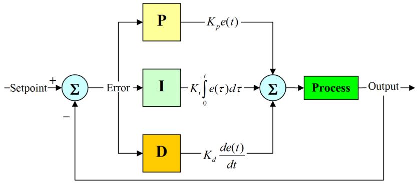


Piezo

Element that moves the sample in XYZ by applying a voltage

Feedback loop

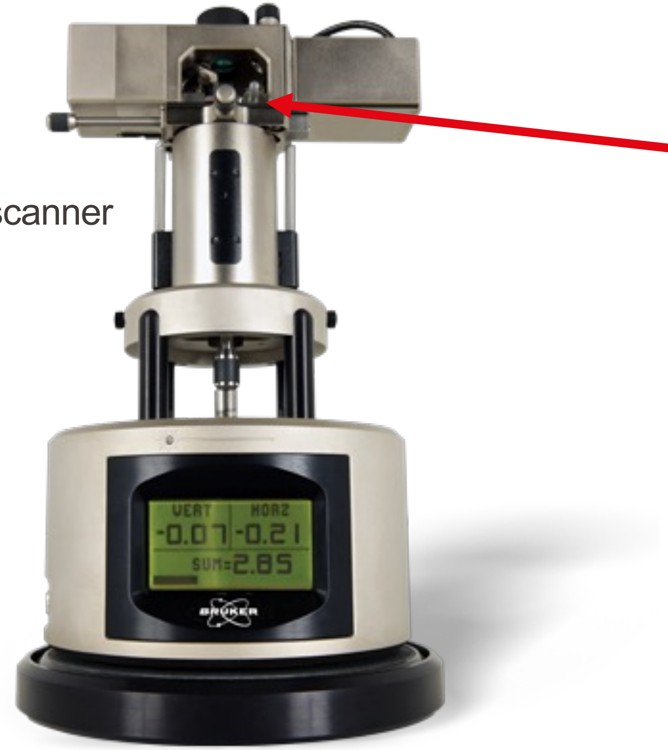
A feedback system is one in which the output signal is sampled and then fed back to the input to form an error signal that drives the system



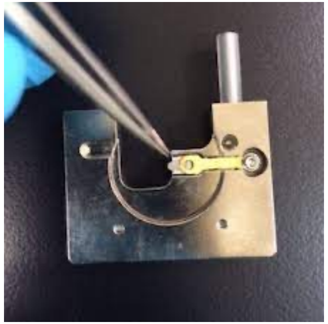
What's in an AFM?

cantilever head

scanner



cantilever holder



controller and HV amplifier

A few principles we should understand

- Optical lever detection
- Piezo-scanners
- Feedback/setpoint
- Force curves
- Imaging modes

Optical lever detection

Transduces cantilever deflection into a voltage

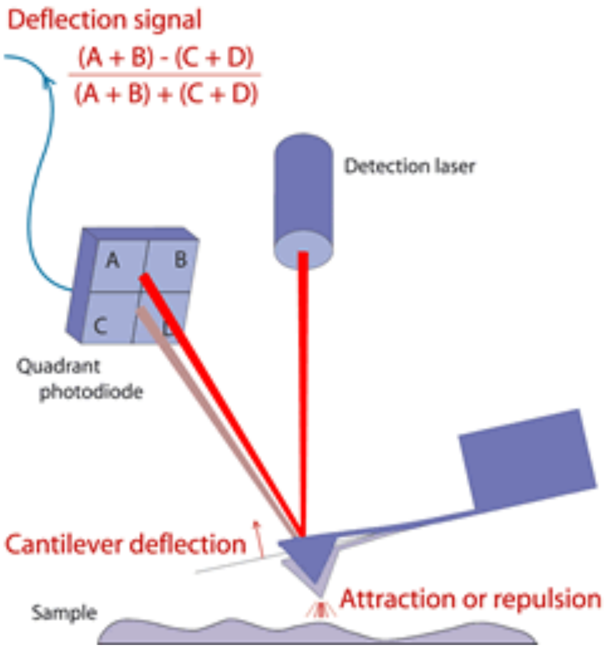
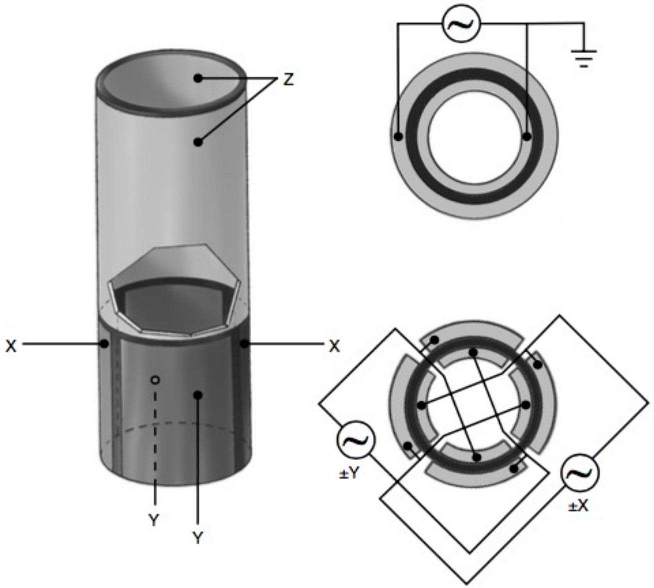


Image source: http://usa.jpk.com

- A very sensitive way to measure cantilever ***angle change***
- The change of angle is amplified by the distance from the cantilever tip to the 4-quadrant photodiode
- Each quadrant creates a current which is turned into a voltage using a transimpedance amplifier (I/V converter)
- The cantilever deflection is the ***normalized*** difference of the top quadrants minus the bottom quadrants

Piezo scanners

Piezo materials expand when a voltage is applied



Piezo scanners can be:

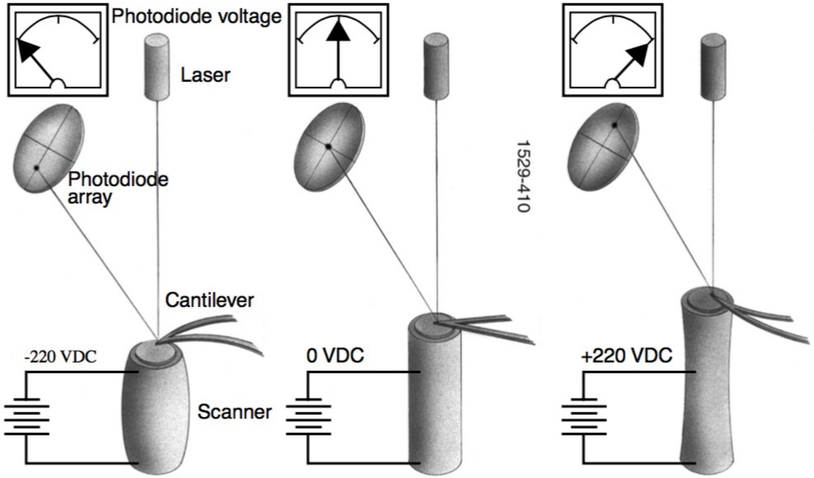
- Tubes
- Stacks
- Plates
- Monolythic piezo blocks

Or other types of actuation can be used:

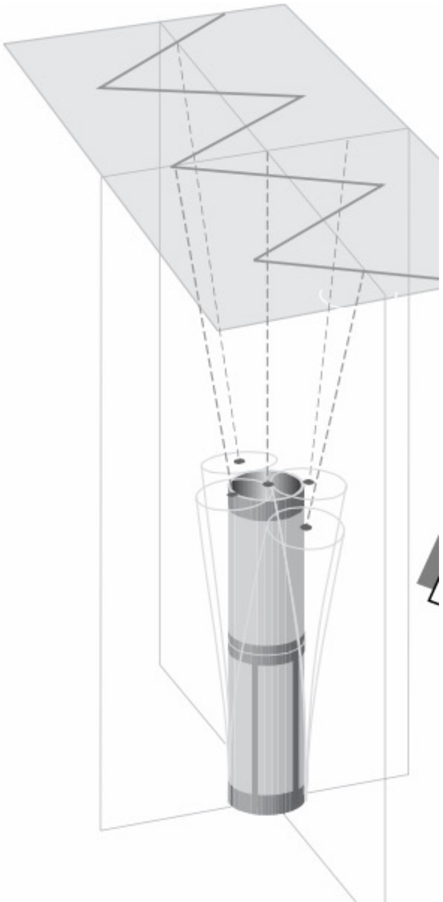
- Voice coil actuation
- Electrostatic combs
- Linear magnetic motors

Piezo scanners

Piezo materials expand when a voltage is applied



Z actuation

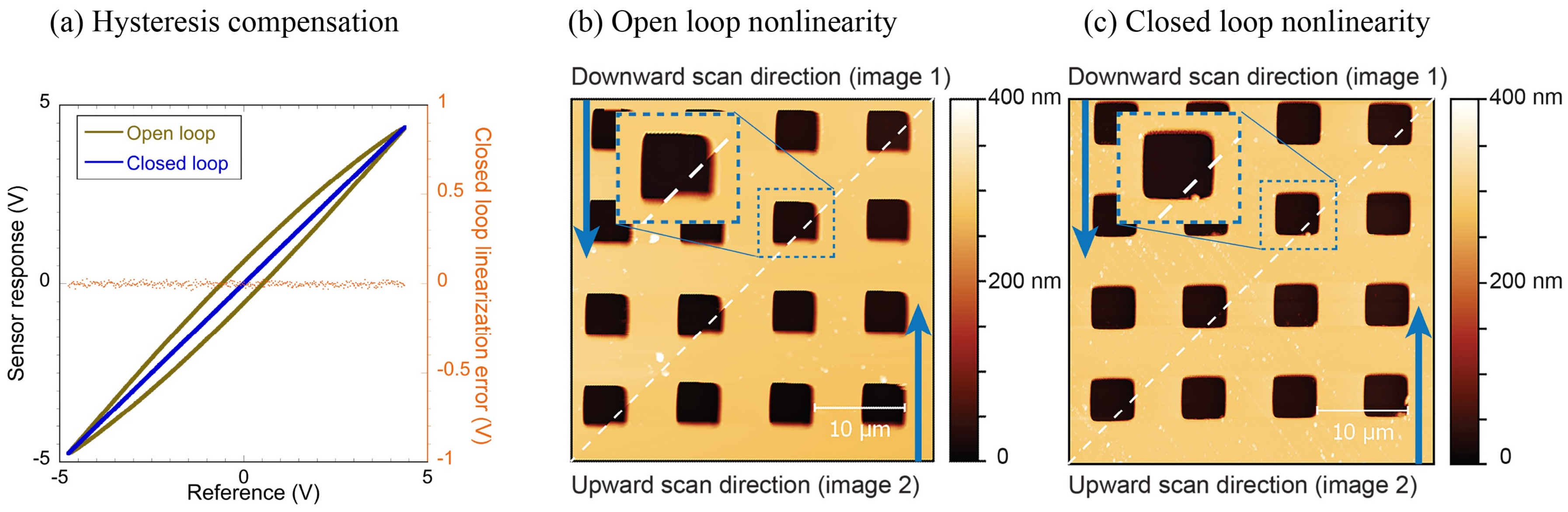


X,Y actuation

Images: Bruker Multimode Manual

Problems with piezo actuators

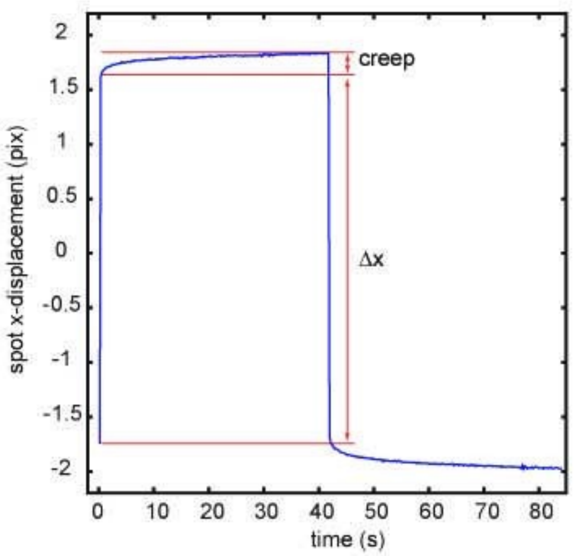
Hysteresis

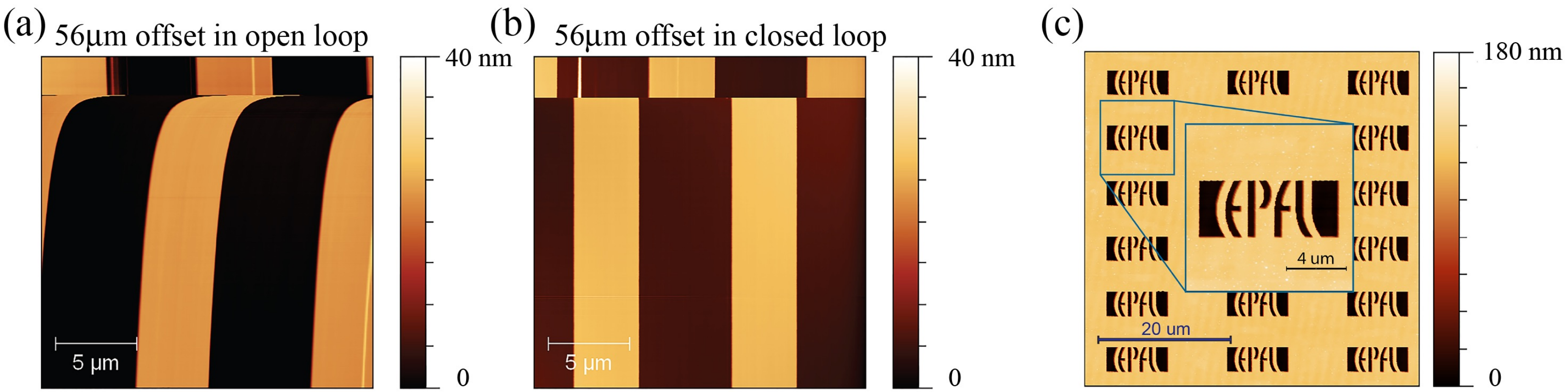


Problems with piezo actuators

Creep

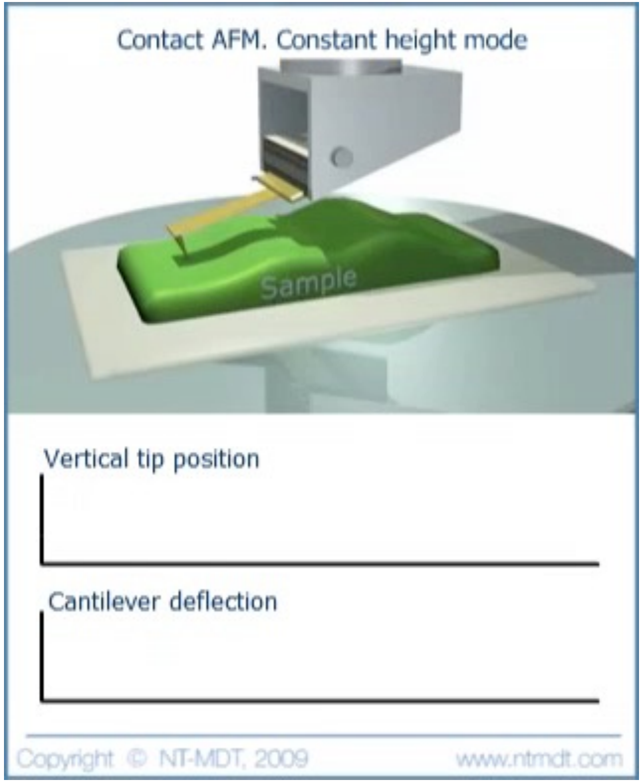
- The piezo only moves ca 90% of the requested distance right away. The rest of the way it creeps very slowly!
- This causes image artefacts like distortion





Why do we need feedback?

Constant height mode

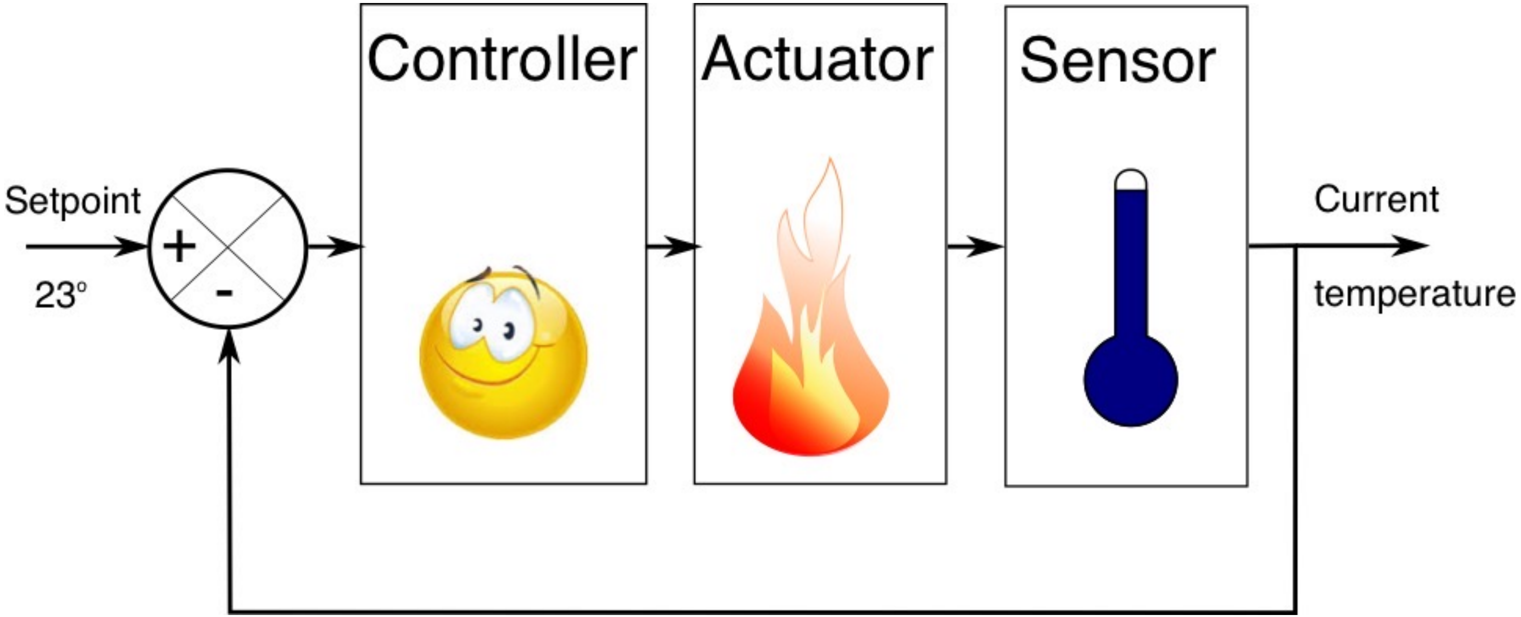


Why don't we just drag the cantilever over the surface?

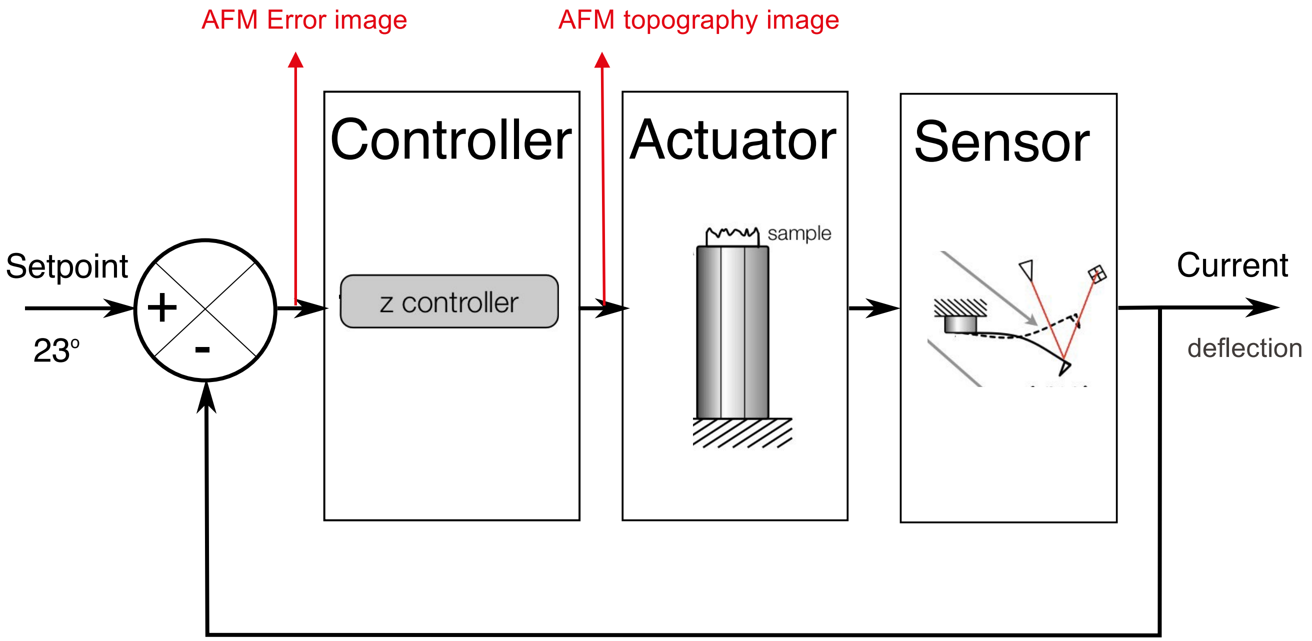
- Cantilever deflection is not linear → height measurement is distorted
- Force on cantilever is not constant → tip and sample can get damaged

What do you do if you are cold?

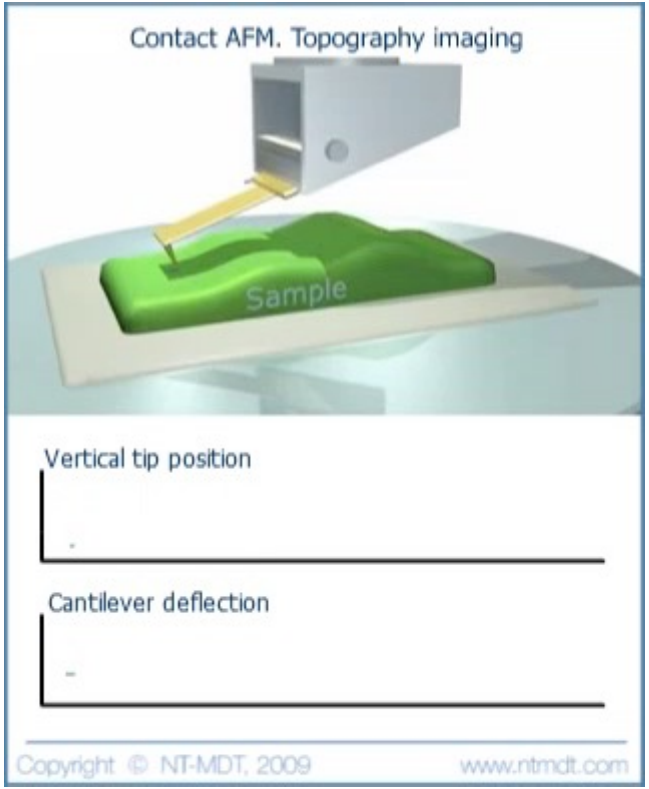
Rearranging into a feedback loop



Rearranging into a feedback loop



Feedback keeps the tip/sample interaction constant

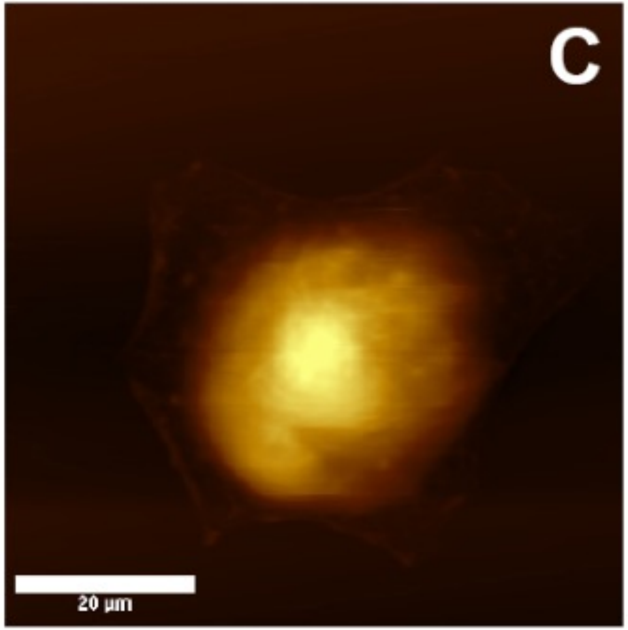


Benefits of operating in feedback:

- Cantilever deflection varies only slightly around setpoint
- The amount that the controller has to move the piezo up or down *approximates* the topography of the sample

Height image vs error image

Height



Error

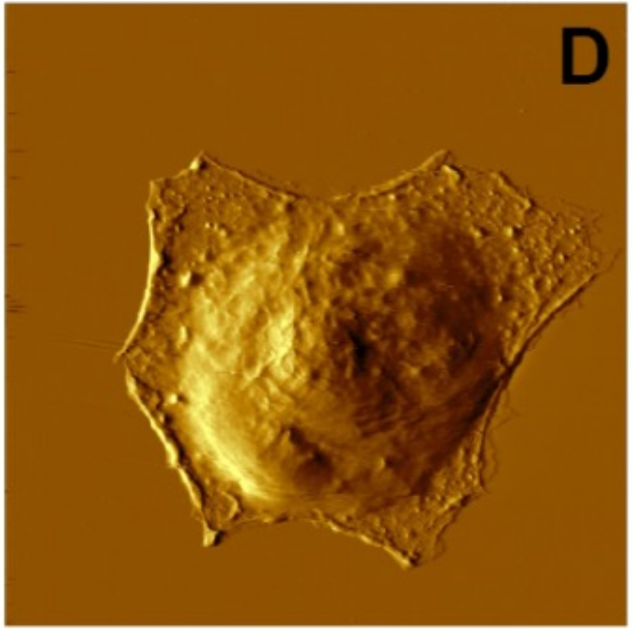


Image source: http://www.ntmdt.com

What is the meaning of the error signal?

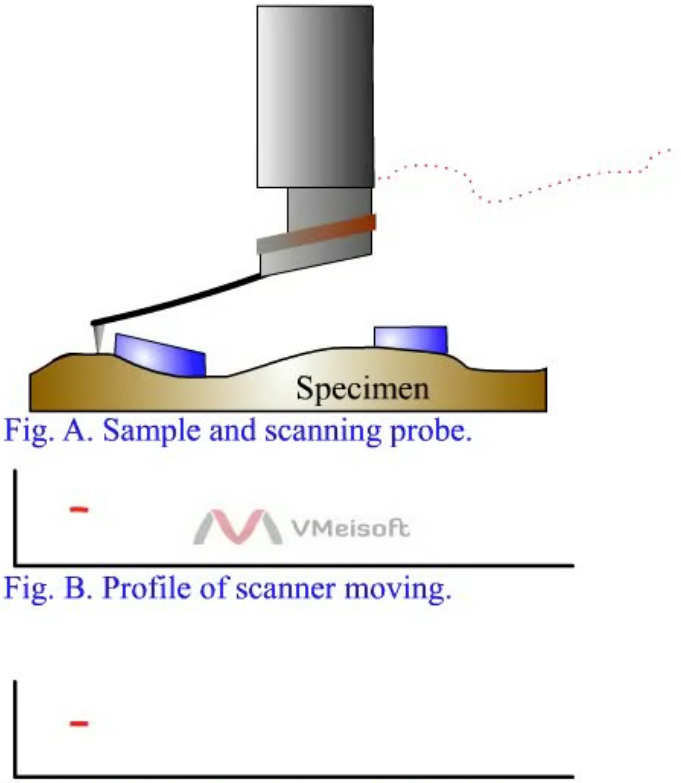


Fig. A. Sample and scanning probe.

Fig. B. Profile of scanner moving.

Fig. C. Profile of cantilever deflection changing.

- The deflection/error signal is as much part of the AFM image as the topography image (also called height image)!
- It accentuates edges and features with small spatial frequencies
- The height image combined with the error image represent the “true topography”

Tip sample interactions

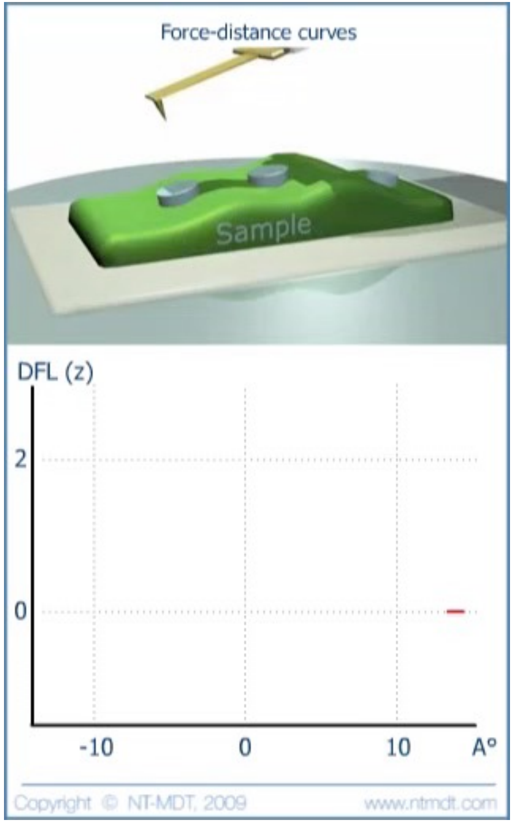
Force curves

There are many forces that can act between the tip and the sample

- Van Der Waals forces (attractive)
- Pauli repulsion (repulsive)
- Electrostatic forces (attractive or repulsive)
- Capillary forces (attractive)
- Magnetic forces (attractive or repulsive)
- …

We can measure what forces act on a cantilever as a function of distance from the surface by measuring a ***Force curve***

Force curves



Force curves can tell us a lot about the tip sample interaction:

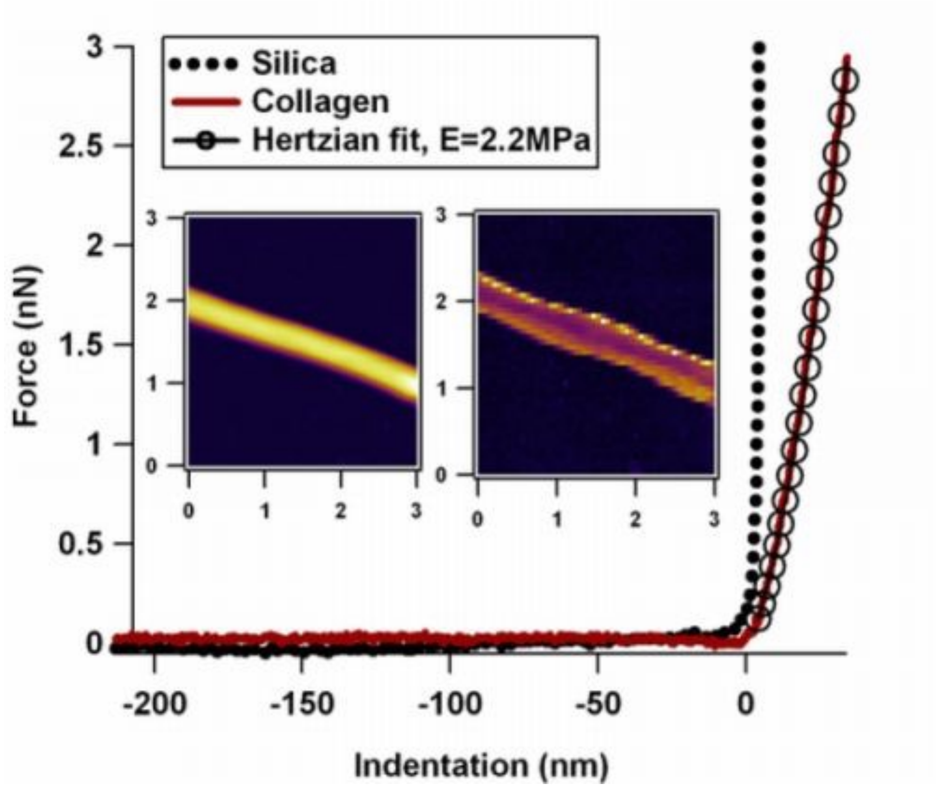
- What is the adhesion of tip to sample
- What is the hardness of the sample
- What is the energy dissipation per cycle

Or about our measurement setup

- What is the deflection sensitivity (how many nm do we have to deflect the cantilever to measure 1V shift in the 4-quadrant photodiode)

Force volume mode

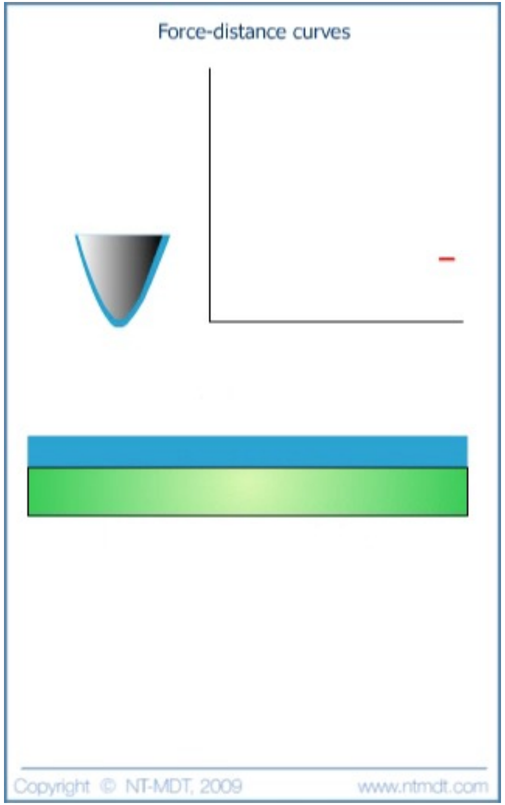
Creating mechanical properties maps



http://www.imaging-git.com

Surface adhesion

Capillary forces are always present when imaging in air!



All surfaces in ambient are covered with a thin water layer.

- Capillary forces act between tip and surface
- When imaging in air they create a “snap-in” as well as adhesion
- They can lead to many artefacts and instabilities

Single molecule force spectroscopy

Force curves as a tool for single molecule mechanics

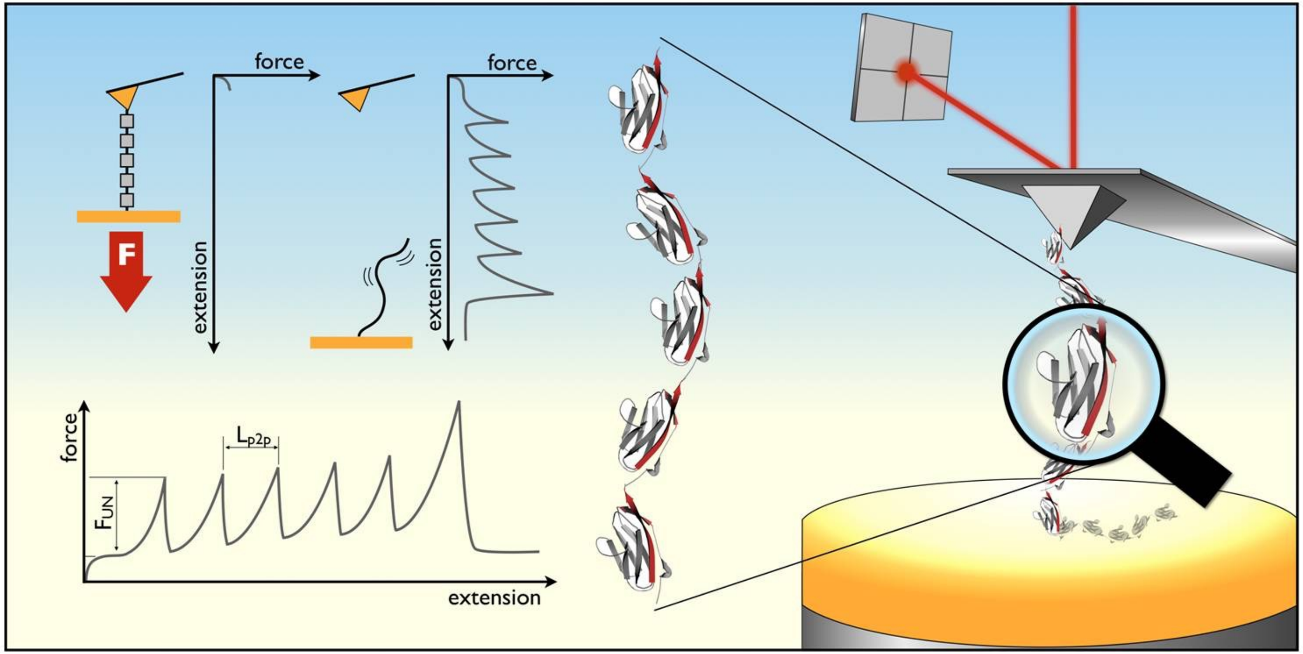
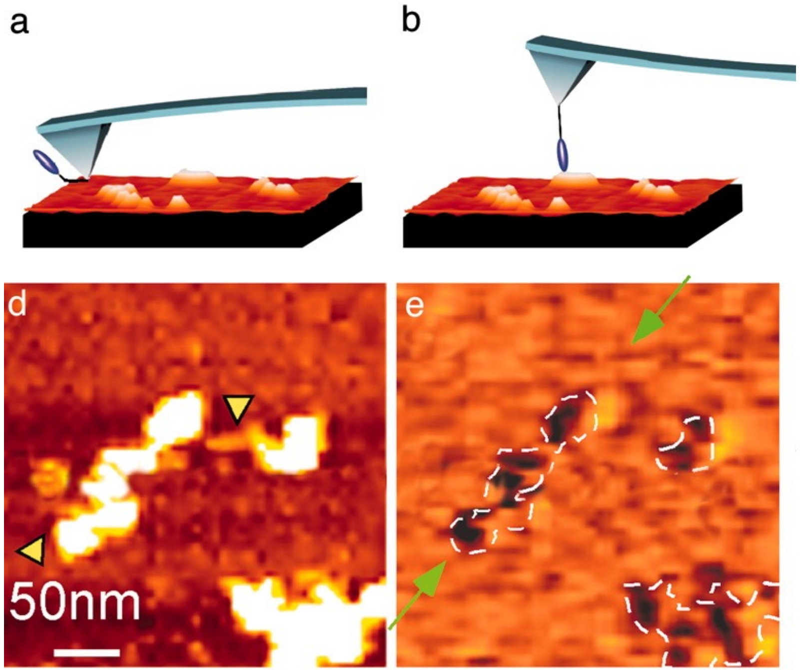


Image source: Dr Lorna Dougan - http://www1.mnp.leeds.ac.uk/ldougan/

Single molecule recognition imaging

Detecting specific antigens on a surface



Adapted from: C. Stroh et al. PNAS 2004;101:12503-12507

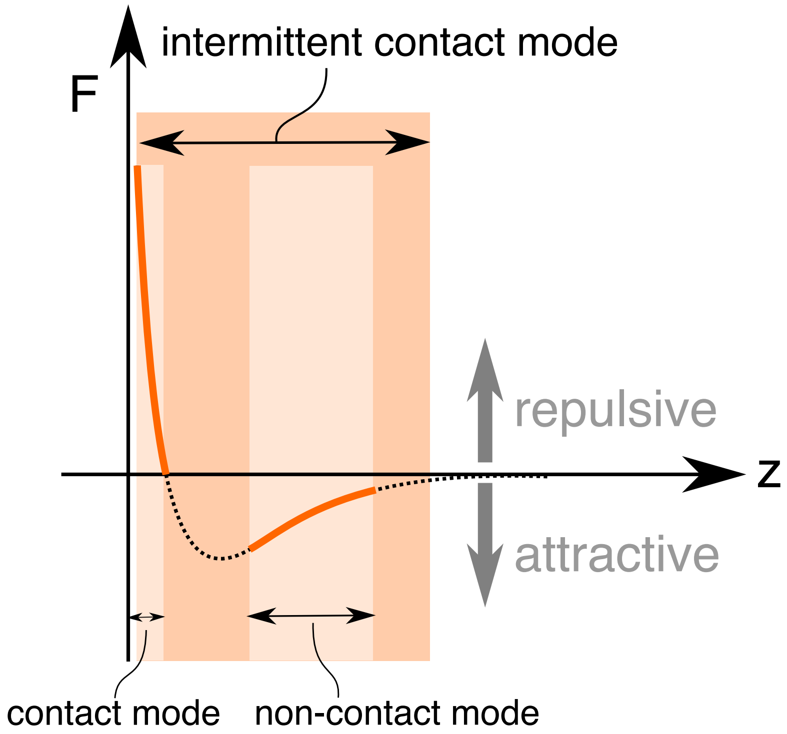
Dynamic modes

Reduces tip sample interactions

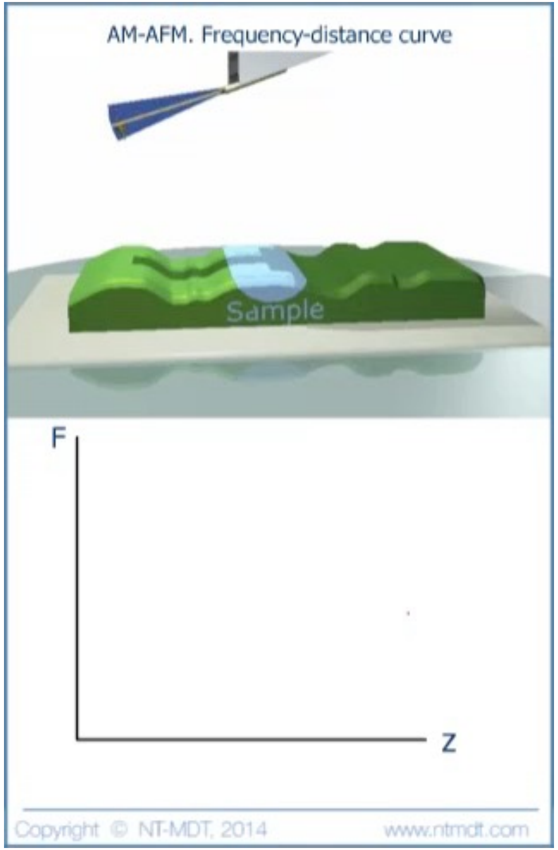
- Tapping mode™ (intermittent contact mode, amplitude modulation mode, dynamic mode,…)
- Non-contact mode
- Off resonance modes (Peak Force Tapping™, QI mode™, hopping mode™, HybriD mode™,…)

Lennard-Jones potential

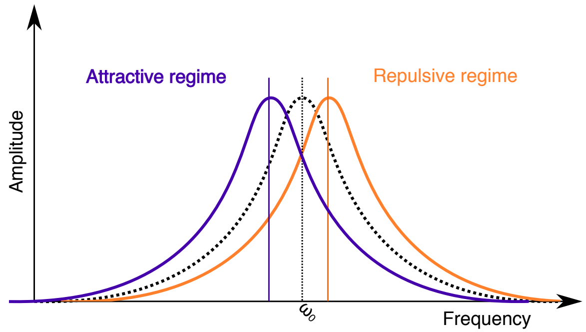
The cantilever feels different force regimes



Oscillating approach curves

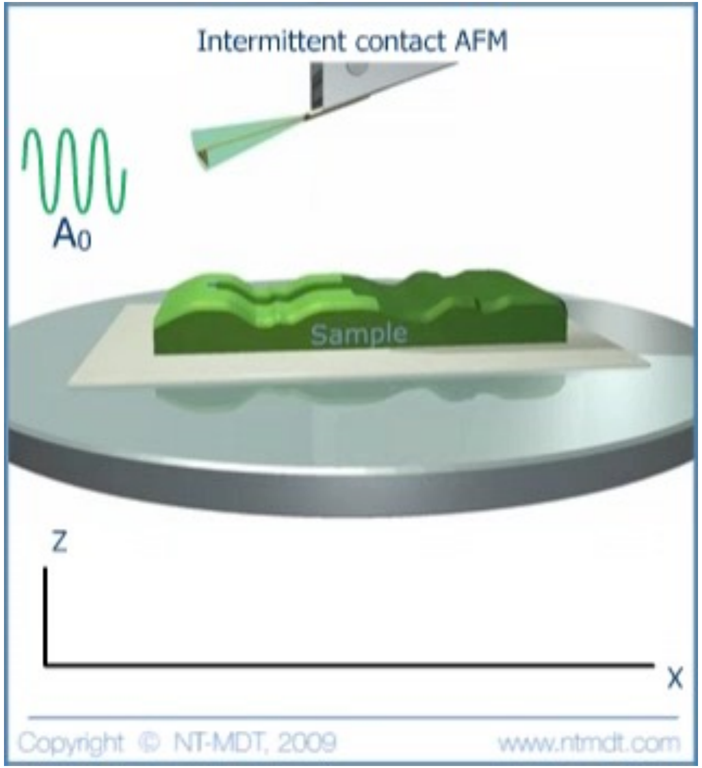


- As the cantilever approaches the surface it feels different forces (due to the Lennard-Jones potential)
- When the cantilever is in the attractive regime the resonance frequency decreases
- When the cantilever is in the repulsive regime the resonance frequency increases

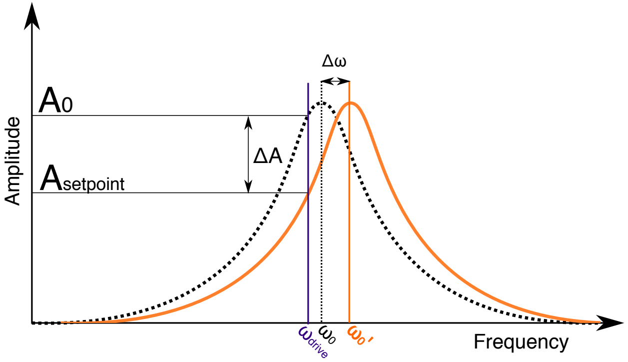


Amplitude modulation

(a.k.a. Tapping mode™)

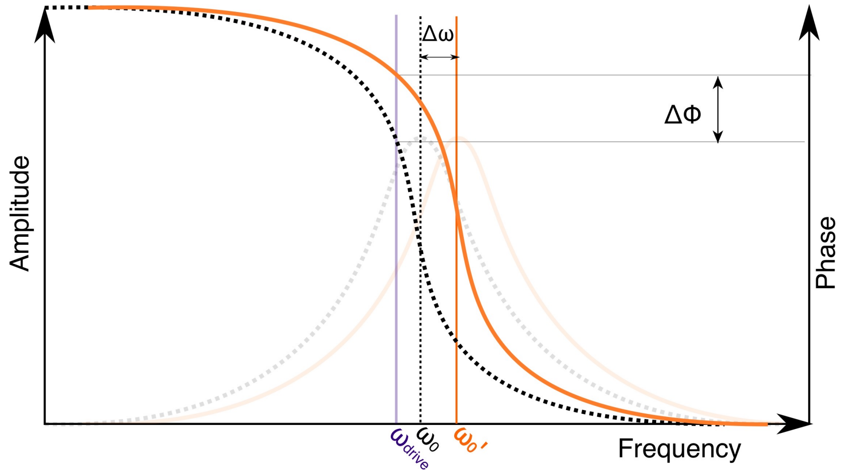


- In tapping mode we excite the cantilever at a fixed frequency ω_{drive} slightly below its resonance frequency
- As the cantilever approaches into the repulsive regime, the resonance frequency (of cantilever + sample force) increases.
- At the fixed frequency ω, the resulting amplitude will therefore drop as we enter the repulsive regime
- The amplitude error is used for the feedback parameter



Phase imaging

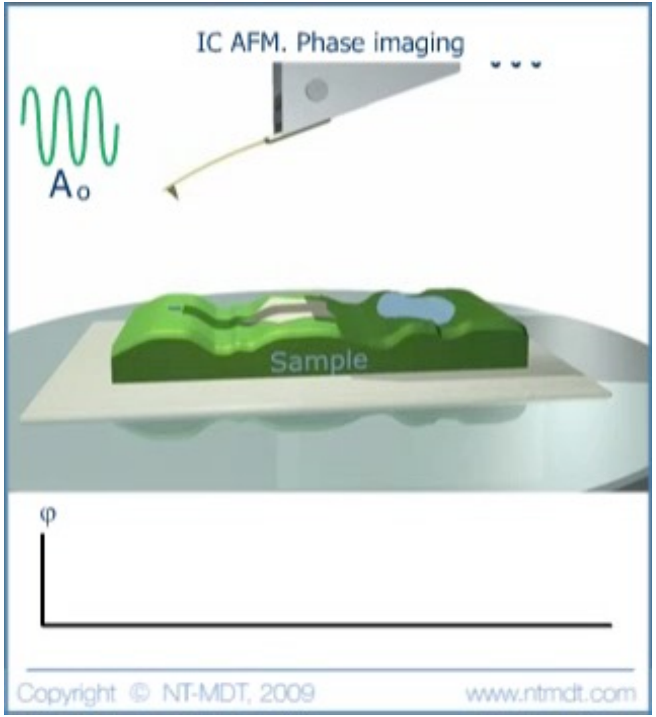
In tapping mode



- The phase difference between the cantilever drive signal and the cantilever oscillation is called the “phase signal”
- The resonance shift $\Delta\omega$ also introduces a phase shift $\Delta\varphi$ at the driving frequency ω_{drive}
- This shift could also be used for feedback, but…
- … other factors such as materials properties affect phase as well

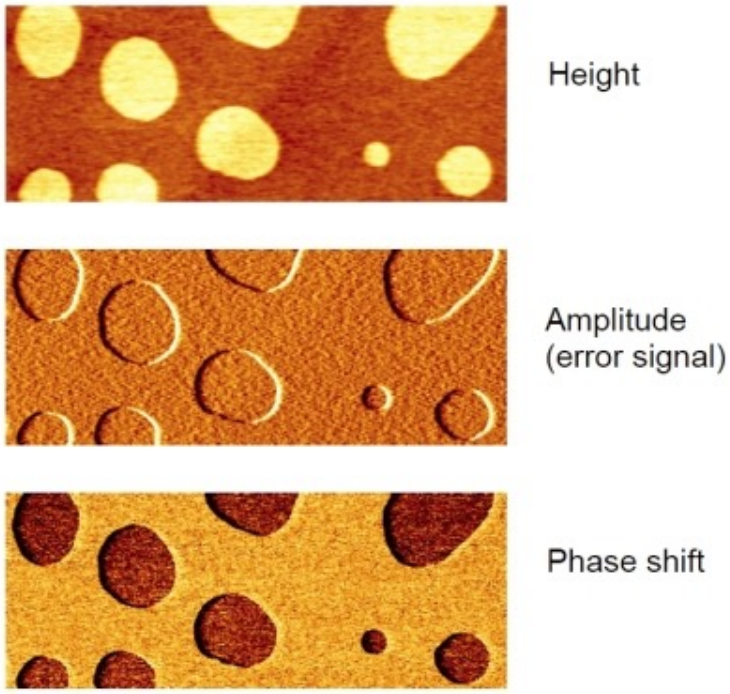
Phase imaging

In tapping mode can give materials contrast



- In tapping mode, the phase channel is an additional observable channel
- If no topography is present, the phase signal can be used to distinguish materials properties
- But beware of interpretation artefacts!

Height vs Amplitude vs Phase

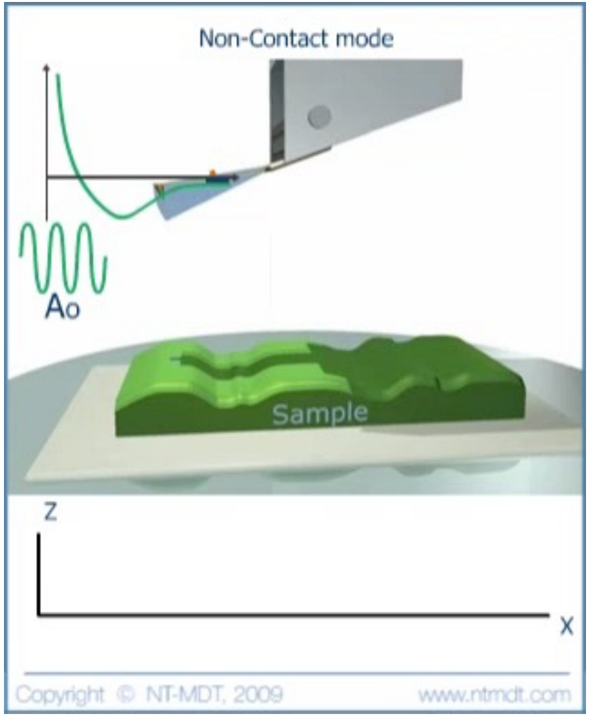


Lipid phase contrast from intermittent contact mode AFM in liquid, 18 x 7 micrometer area. The height (top) and phase (bottom) images show contrast between the lipid phases. The amplitude image (center) shows contrast at the edges of the patches.

http://www.microscopy-analysis.com

Non-contact mode

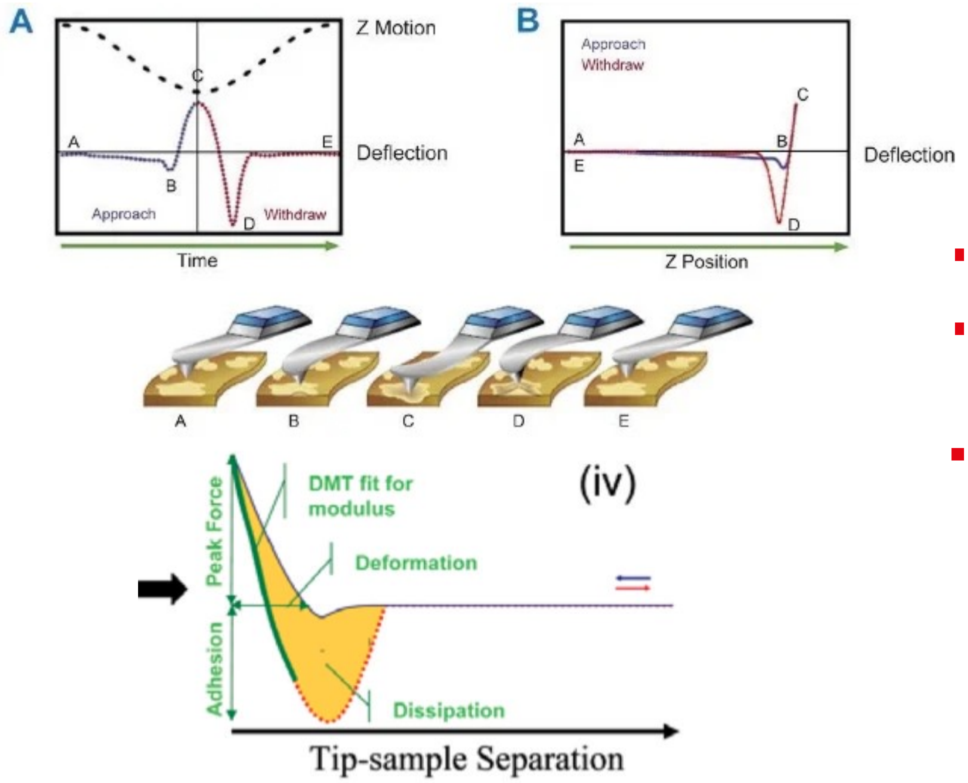
Imaging in the attractive regime



- In non-contact mode, the cantilever "never touches" the surface
- Non-contact mode is difficult to maintain due to the low forces and small force gradients
- Primarily used in vacuum

Off-resonance modes

Peak Force Tapping, QI mode, hopping mode, HybriD mode,…

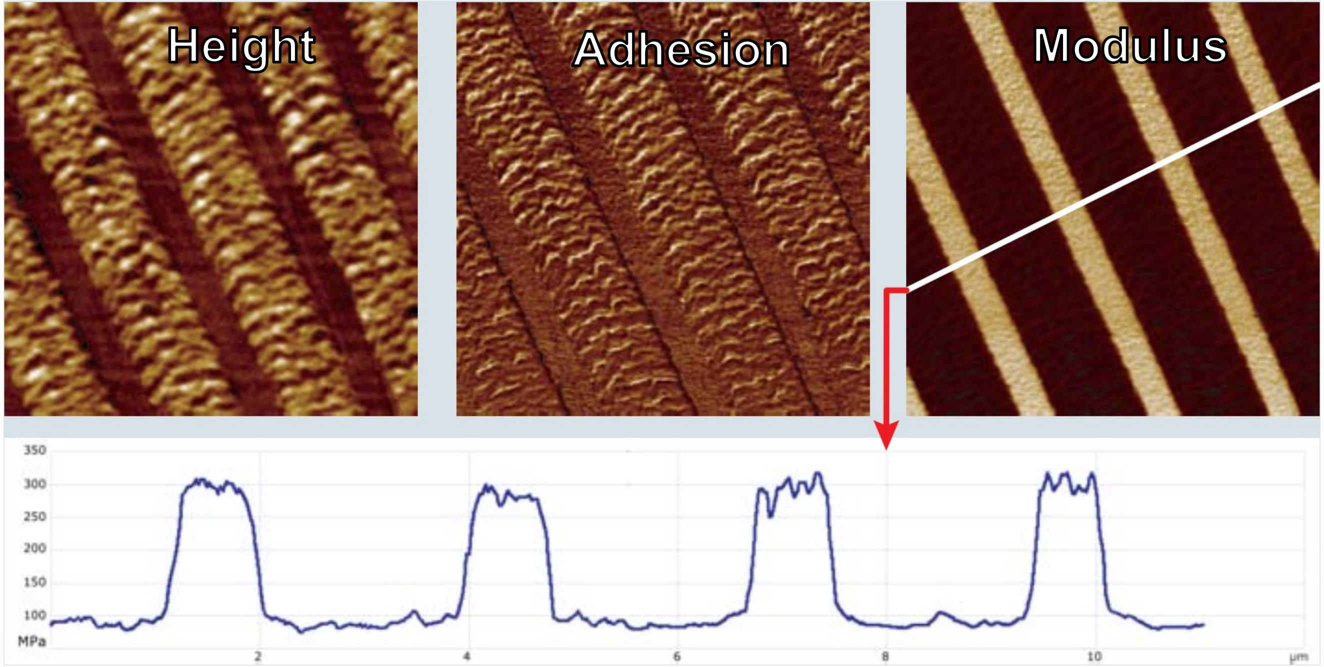


- Ramp often done sinusoidally
- After baseline correction a force curve for each pixel is extracted
- Data extracted from the force curve:
  - Elastic modulus
  - Adhesion
  - Indentation
  - Energy dissipation

Image adapted from Bruker application note 128

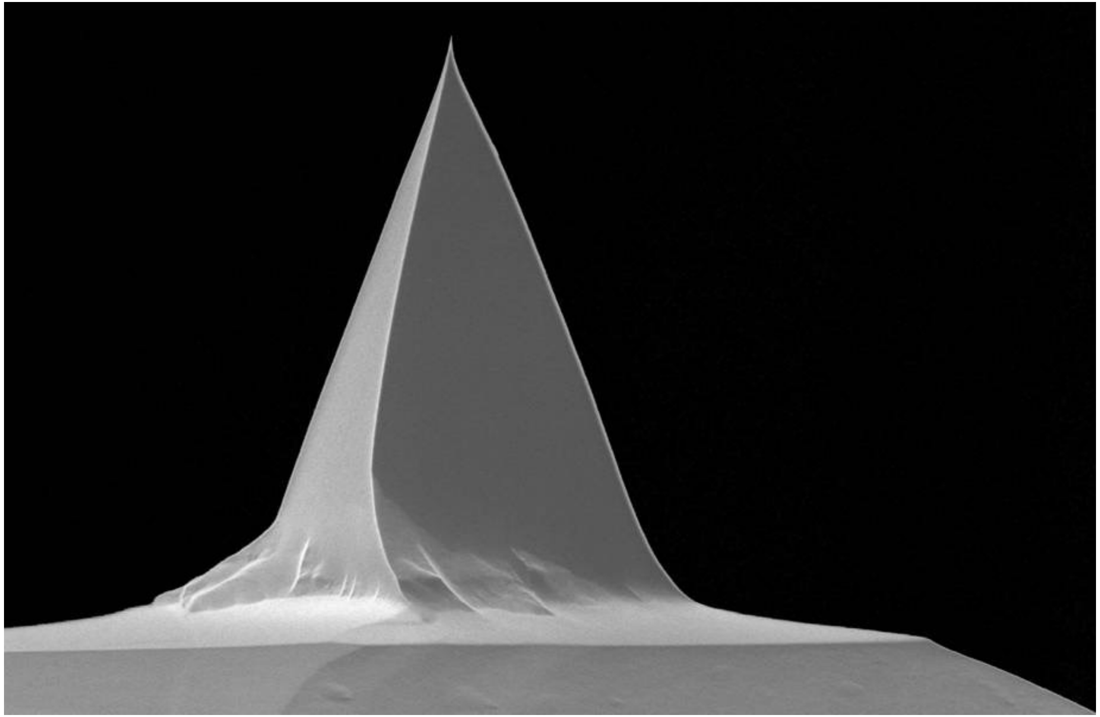
Measuring multiple sample properties

Multilayer polymer optical film



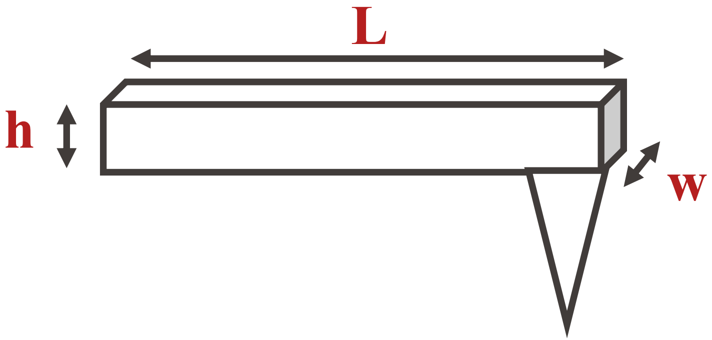
Adapted from Bruker application Note #128

The cantilever



Cantilevers

What considerations govern the probe dimensions?



Resonance frequency

$$F_R \cong 0.162\sqrt{\frac{E}{\rho}} \bullet \frac{h}{L^2}$$

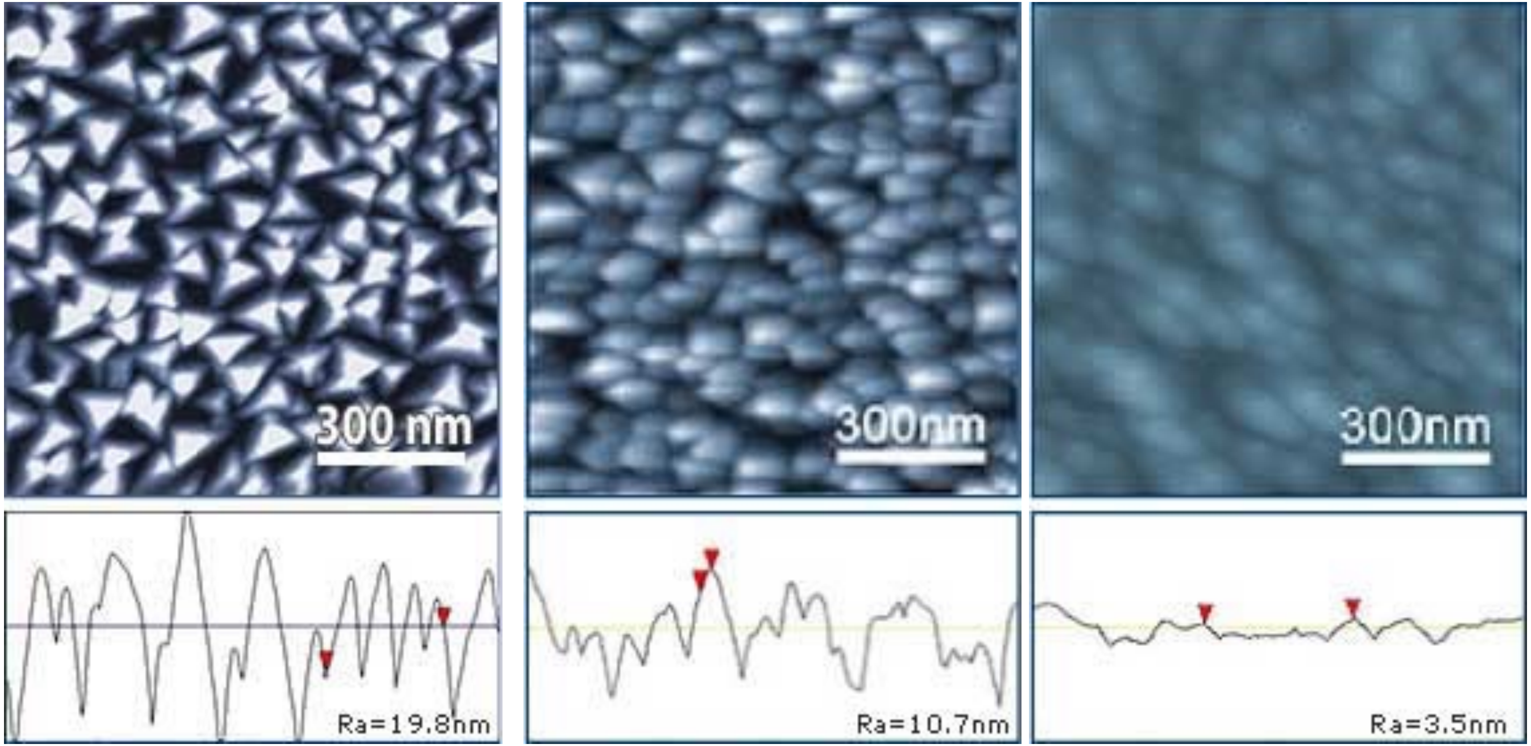
Normal spring constant

$$k_N = \frac{Ewh^3}{4L^3}$$

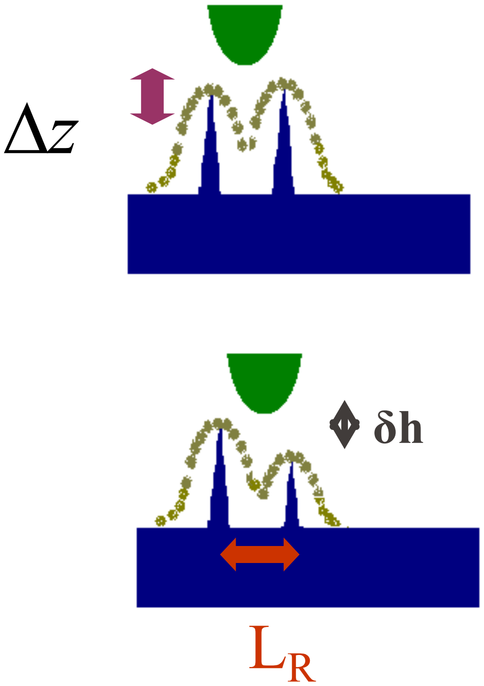
E= Young's modulus, ρ = density

Image resolution

Influence of tip sharpness



Resolution depends on…



- Resolution depends on:
  - Instrument noise floor (Δz)
  - Tip radius (R)
  - Sample topography (δh)

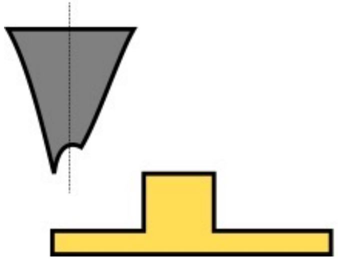
$$L_R = \sqrt{2R}\left(\sqrt{\Delta z} + \sqrt{\delta h + \Delta z}\right)$$

If the sample is compliant the resolution is even further reduced by the indentation (l):

$$l = 2\left[\frac{3RF}{4E^*}\right]^{1/3}$$

Tip artefacts

Draw the height profile the tip would measure



Tip artefacts

Draw the height profile the tip would measure

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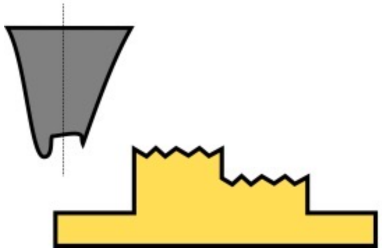
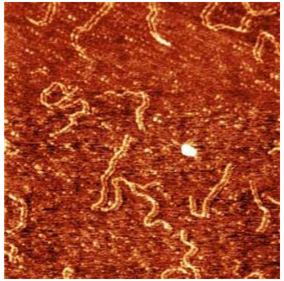
Draw the height profile the tip would measure



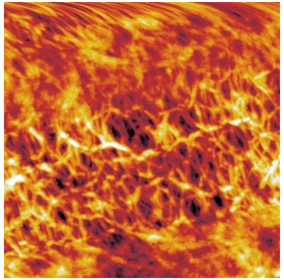
AFM image artefacts

Always be critical when interpreting your AFM images!

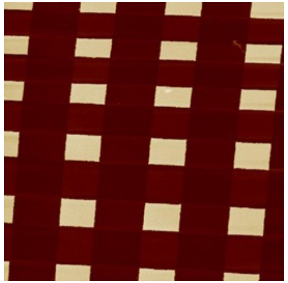
Double tip



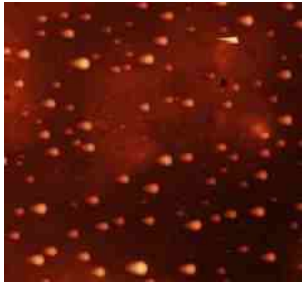
Piezo creep



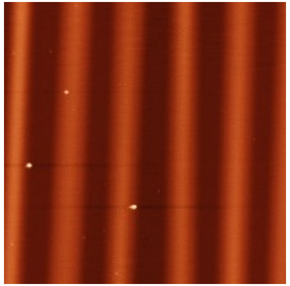
Piezo hysteresis



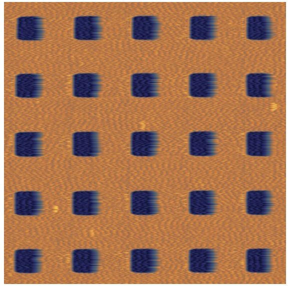
Parashooting



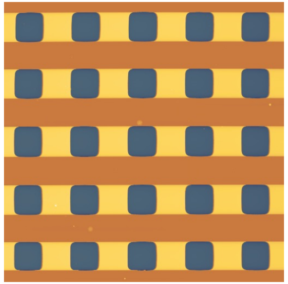
Laser interference



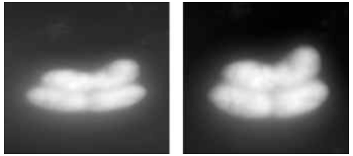
Gains too high



Bad image processing



Sample drift



Questions?