The BioScope™ Catalyst™

Accessing All Biological Size Scales with High-Resolution AFM Imaging

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High Resolution Imaging
From Single Molecules to Cells & Tissues

180 nm image of Human endothelial cells captured at 2k x 2k pixel resolution.
(Contact mode in fluid.)

500 nm phase image of E. coli S-layer membranes exhibiting the characteristic 14nm lattice periodicity.
(TappingMode™ in fluid.)

600 nm height image of Lambda DNA adsorbed onto a mica surface.
(TappingMode™ in fluid.)

180 µm image of Human endothelial cells captured at 2k x 2k pixel resolution.
(Contact mode in fluid)
The BioScope™ Catalyst™

Accelerating Discovery in Life Science Research

Key Benefits of the BioScope Catalyst:

- XY-scan range ≥ 150 μm
- Z-range ≥ 20 μm
- Functional Integration with a variety of light microscopy techniques.
- MIRO Software
  - Optically guided navigation of the AFM probe.
  - Image Overlay
  - Correlated datasets
- Environmental Control
  - Perfusion Stage Incubator (PSI)
  - Stage Heater
  - Small Volume Flow Cell (60 μL)
The BioScope™ Catalyst™: MIRO

Microscopy Image Registration & Overlay

- MIRO Software= true functional integration
  - Registration of optical field of view (transmitted light / fluorescence data) to AFM scan area.
  - Optical images captured directly into NanoScope AFM Software.
  - Use registered optical images to navigate AFM probe to region of interest for high resolution imaging / mechanical property measurements.
  - Direct correlation of structure and functional sample properties.

Combined AFM / CLSM

Fibroblast Cells labeled with Alexa Fluo 546 Phalloidin (red) & DAPI (blue).

Confocal fluorescence images obtained using a 40x oil immersion objective.

AFM images obtained in contact mode in buffer solution at 37°C.
Perfusion Stage Incubator (PSI)

- Typical use
  - Extending viability of cell cultures for longer duration studies

- Features
  - Uses standard 50mm glass bottom petri dishes (from WillCo Wells)
  - Unique sealed design minimizes evaporation without interfering with normal AFM operation
  - Allows continuous or intermittent perfusion of cell media or buffer
  - Allows perfusion of gas, e.g. oxygen / carbon dioxide blend
  - Integrates with heater stage
The Catalyst Small Volume Flow Cell allows fluid exchange in a 60μL total volume.

It is ideal for buffer exchange experiments, especially those using limited or expensive reagents or when more rapid exchange of fluids is desired.
The BioScope Catalyst facilitates high resolution imaging of single molecules and small biomolecular complexes.

- Highest Quality Engineering and Mechanical Stability (even when operated on an inverted optical microscope)
- Adaptive Scan
- PeakForce Tapping
- AFM Probes

C60H122 alkane is spin cast onto an HOPG substrate. The resulting ultra-thin alkane layer exhibits a lamellar structure ~7.5nm in width and 0.4nm in height. BioScope Catalyst TappingMode™ Phase Image, 1μm scan. Image was acquired using FESP probes (k ~3N/m).
Adaptive Scan

Closed Loop Accuracy With Open Loop Noise Performance

- High resolution imaging at small scan sizes can often be affected by XY-sensor noise (‘jitter’ in image).
- XY-Adaptive Scan:
  - Removes sensor noise (jitter)
  - Facilitates faster scan rates (eliminates ringing)
  - Maintains linearity and offset accuracy of XY-closed loop operation

![Closed Loop XY](image1.png)
![Adaptive XY](image2.png)

SBS Copolymer in Air
PeakForce Tapping
Controls and measures force as feedback signal

How PeakForce Tapping Works:
- The Z position is modulated at a small amplitude, we measure a series of very fast force curves, at least once per pixel.
- The peak force of each of these curves is used as the feedback signal.
- This force can easily be <100 pN, about 10x lower than typical with TappingMode.

PeakForce Tapping Standard Data Types:
- Height, Peak Force Error, and Phase.
Self-Optimizing AFM Imaging Mode

- Automatic image optimization results in faster, more consistent results, regardless of user skill level.
- Direct force control at ultra-low forces helps protect delicate samples and tips from damage.
- ScanAsyst makes it dramatically easier to image in liquids.
  - No need to tune cantilever resonance.
  - The imaging setpoint does not drift.
  - The imaging force can be precisely controlled at ultra-low setpoints.

3D topography image of pUC plasmid DNA adsorbed onto a mica substrate. The individual DNA strands are clearly visible against the mica background. Images were acquired on a BioScope Catalyst AFM operated in PeakForce Tapping in buffer solution using ScanAsyst Fluid+ AFM probes (k ~0.7N/m). Image XY-Scale = 2mm.
Simultaneously obtain quantitative modulus, adhesion, dissipation, and deformation data while imaging topography at high resolution.

Direct force control keeps the imaging force low, which limits indentation depths to deliver non-destructive, high-resolution imaging.

Material properties can be characterized over a very wide range to address samples in many different research areas (kPA – GPa).

\[
F_{\text{tip}} = \frac{4}{3} E^* \sqrt{R \delta^{3/2}} + F_{\text{adhesion}}.
\]
Amyloid fibrils were adsorbed onto a freshly cleaved mica surface. Images were obtained on a BioScope Catalyst operated in PeakForce QNM mode using ScanAsyst AFM Probes (K~0.4 N/m). Sample courtesy: Dr. Xingfei Zhou, Ningbo University, China.
AFM images were obtained on the BioScope Catalyst operated in PeakForce Tapping mode in buffer conditions and using ScanAsyst Fluid+ AFM probes (k ~0.7N/m). Sample courtesy of Dr. Wouter Roos & Prof. Gijs Wuite, Vrije Universiteit, Amsterdam, Netherlands.
Images were obtained on a BioScope Catalyst operated in TappingMode in buffer conditions using SNL AFM probes (k ~0.32N/m). Sample courtesy of Prof. Hans Oberleithner, Institute for Physiology II, University of Muenster, Germany.
AFM Probes are critical to high resolution imaging studies.

- Choice of probe for sample-type and imaging mode.
- Quality of probes (reproducibility of data and time-to-results)

Advantages to Bruker having its own Probes Facility.

- Wide-range of probes available for all operating modes and samples (biological polymers data storage).
- Closely tied to technology and applications development.

*ScanAsyst Fluid+ SNL MSCT*
Summary

- AFM has many advantages for high-resolution studies of single biomolecules and biomolecular complexes.
  - No staining / coating (native surface remain unaltered)
  - No crystallization of sample
  - Ability to operate under fluid
  - Environmental Control

- The BioScope Catalyst provides researchers with a wide-range of unique high-resolution imaging opportunities.
  - Mechanical Stability when integrated with light microscopy
  - MIRO Software
  - PeakForce Tapping (ScanAsyst & PeakForce QNM)
  - Small Volume Flow Cell (60 μL)
  - Perfusion Stage Incubator (PSI) & Stage Heater
New Application Note: “Common Approaches to Tip Functionalization for AFM Based Molecular Recognition” by Ben Ohler & Alexandre Berquand

- This App Note reviews the major strategies that can be used to functionalize AFM tips with molecules of interest.
- With this guide, any AFMist, even beginner, can start with tip functionalization.
What is the interest?

- In biology, using functionalized probes to track molecules of interest present at the surface of living cells is a possible way to get better insight in what’s happening underneath the plasma membrane.

- This article reports the major ways to attach ligands at the end of AFM tips and lists the main drawbacks and benefits of using each of them.

- Using this approach together with quantitative investigation techniques like Peak Force QNM can become a real breakthrough combination in the world of force measurements.
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Conference Chair: Paul Hansma, University of California, Santa Barbara, USA

Preliminary Agenda:

July 19 - AFM Tutorial, Bruker Nano Surfaces Business, Santa Barbara, Ca., USA

July 20-22 - Scientific Sessions, University of California, Santa Barbara, USA

For more information, please visit:

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The conference will be held at Institut Curie
Hosted by: Institut Curie, Inserm and CEA

Featured sessions and confirmed chairs:
- High-resolution imaging / Zhifeng Shao
- Force and mechanical measurements / Carlos Bustamante
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