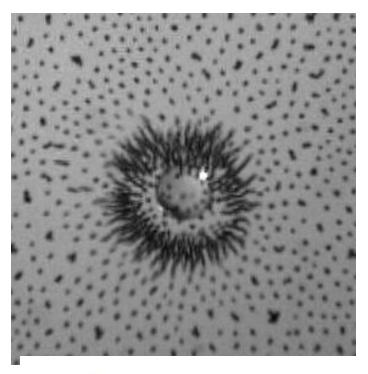
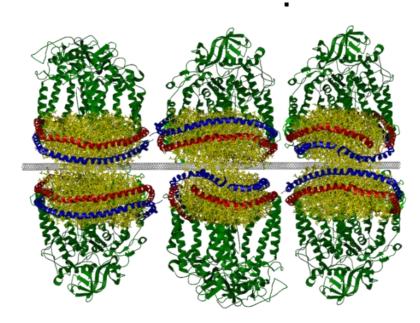
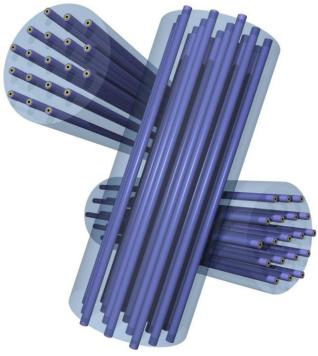
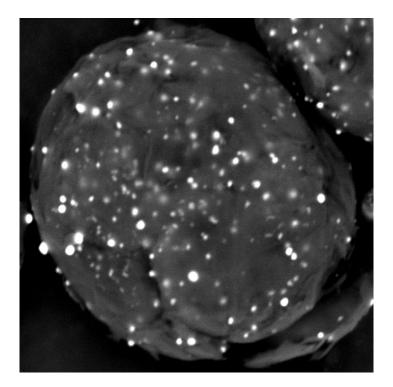
# **Biomolecular Materials Principal Investigators' Meeting–2011** October 23–26, 2011 Annapolis Westin Hotel, Annapolis, MD











**Office of Basic Energy Sciences** Materials Sciences and Engineering Division

## **Toward Capturing Soft Molecular Material Dynamics**

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### **Biological Atomic Force Microscopy**

### **Scientific Challenge**

Many soft materials readily deform under the minimum force required to perform an AFM measurement precluding imaging at high temporal and spatial resolution. Methods to reduce significantly the minimum detectable force and increase imaging rate are required.

### **Research Achievement**

Although quite fashionable, attempts to use feedback methods such as Q-control and frequency modulation have failed to improve the image quality when in solution. Qcontrol amplifies weak tip-sample interactions and the thermal noise equally providing no overall advantage.<sup>1</sup> Frequency modulation also amplifies weak tip-sample interactions but controls the amplitude noise. However, the feedback shifts the amplitude noise to the time domain precluding a precise measurement of frequency providing no overall advantage either.<sup>2</sup> Instead, the thermal force-noise of the cantilever is the principal limitation to reducing sample Minimizing a cantilever's deformation. cross- section reduces its noise significantly and the minimum size of the cantilever is currently limited by a conventional deflection detection scheme, which requires

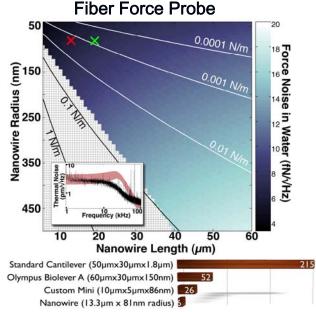


Figure 1 - The calculated force noises of nanowires as a function of their radii and lengths, in water. Contour lines denote cantilever stiffnesses, and the hashed region corresponds to thermal fluctuations that are too small to be detected with our system, with 5 mW of laser power. Inset figure is the measured thermal noise spectrum of the 2±1 mN/m stiff nanowire marked with a red  $\times$  (force noise of 6±3 fN/ $\!\sqrt{\rm Hz}$ ), as well as a theoretical prediction based on its dimensions and material properties. The green  $\times$  is a measured 19:4  $\mu m$  by 82.5 nm nanowire (force noise of 7±4 fN/ $\!\sqrt{\rm Hz}$ ). The bar chart shows Fiber Force Probe force noise relative to other AFM cantilevers. The Fiber Force Probe has record low force noise in solution.

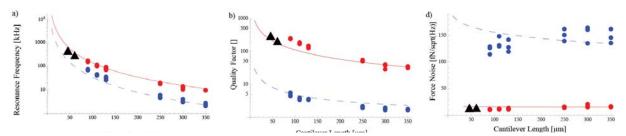


Figure 2 - Resonance frequency, quality factor, and force noise as a function of cantilever length for normal cantilevers in air ( $\bullet$ ) and in water ( $\bullet$ ). The encased cantilevers ( $\blacktriangle$ ) in water have the same high-performance as normal cantilevers in air.

Regular Si Cantilever

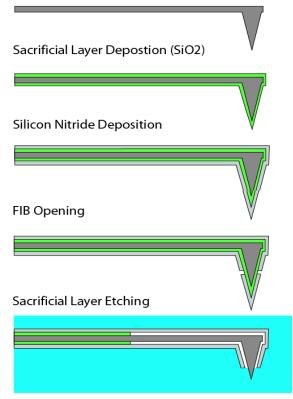


Figure 3 - Cartoon of encased cantilevers fabrication process. PE CVD is used to deposit a sacrificial layer and the encasement. The sacrificial layer is etched back from an opening at the probe apex. Surface tension prevents water from entering the encasement.

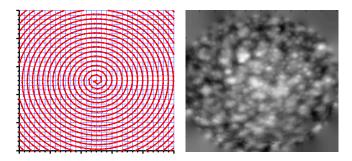


Figure 4 - spiral scans require lower acceleration and force them raster scanning enabling higher tip velocities and frame rates for the same scanner

a large surface area for laser specular reflection. A forward scattering optical deflection detection technique enables the use of nanowires as cantilevers. We achieved a force noise in water of 6 fN/ $\sqrt{Hz}$  that is orders of magnitude gentler than conventional AFM using the Fiber Force Probe AFM.<sup>3</sup> The Fiber Force Probe has a number of significant limitations such as slow scan speed, difficult sample geometry, and lack of robustness. To mitigate these challenges we reduced force noise by reducing the fluid viscosity with a protective encasement for the cantilever. The cantilever operates in air but the probe protrudes from the encasement through the solution to the Encased cantilevers sample. have exceptionally high resonance frequency, Q factor, and detection sensitivity and low force noise enabling gentle high speed imaging.<sup>4</sup> They also work in all commercial AFM systems without modification. These are significant milestones towards non-invasive scanning probe imaging of biological processes on the surfaces of vesicles and cell membranes.

Present raster scan techniques are poorly matched to the instrument limitations of Atomic Force Microscopy. Serial data

> collection from the local probe makes image collection slow and unable to match the timescales of many chemical and biological processes. One basic issue is the propensity of scientists to oversample data. We have used advanced image processing tools such as inpainting to recover high-resolution images from sparse quickly collected images to improve temporal resolution without applying more force or

High SampledLow SampledInpaintedDifferenceImage: DifferenceImage: Dimage: DifferenceImage: Differe

Figure 5 - inpainting diffuses data from known regions of an image to unknown regions by balancing fidelity to the original data and smooth connectedness of the resulting image. For simple images and edges quickly acquired low density data is all that is required enabling higher frame rates.

increasing bandwidth.<sup>5</sup> We are also

using non-raster scan algorithms such as spiral and cyclic scanning to increase temporal resolution. Spiral scanning better matches the mechanical limitations of the AFM scanner and allows higher tip velocities without distortion. Inpainting or interpolation is used to quickly create images from the nongrided data.<sup>6</sup>

The use of gentle high bandwidth probes and fast scanning algorithms will enable

Atomic Force Microscopy to probe soft materials with high resolution and capture dynamics of assembly and function.

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