

Materials List for

# Atomic Force Microscopy Cantilever-Based Nanoindentation: Mechanical Property Measurements at the Nanoscale in Air and Fluid

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## Materials

Name	Company	Catalog Number	Comments
Atomic force microscope	Bruker	Dimension Icon	Uses Nanoscope control software, including PeakForce Quantitative Nanomechanical Mapping (PF-QNM), FastForce Volume (FFV), and Point-and-Shoot Ramping experimental workspaces
AtomicJ	American Institute of Physics	<a href="https://doi.org/10.1063/1.4881683">https://doi.org/10.1063/1.4881683</a>	Flexible, powerful, free open source Java-based force curve analysis software package. Supports numerous contact mechanic models, such as Hertz, Sneddon DMT, JKR, Maugis, and cone or pyramid (including blunt and truncated). Also includes a variety of initial contact point estimation methods to choose from. Supports batch processing of data and subsequent statistical analysis (e.g., averages, standard deviations, histograms, goodness of fit, etc.). Literature citation is: P. Hermanowicz, M. Sarna, K. Burda, and H. Gabryś, "AtomicJ: An open source software for analysis of force curves" <i>Rev. Sci. Instrum.</i> <b>85</b> : 063703 (2014), <a href="https://doi.org/10.1063/1.4881683">https://doi.org/10.1063/1.4881683</a>
Buffer solution (PBS)	Fisher Chemical (NaCl), Sigma Aldrich (KCl), Fisher BioReagents (Na <sub>2</sub> HPO <sub>4</sub> and KH <sub>2</sub> PO <sub>4</sub> )	S271 (>99% purity NaCl), P9541 (>99% purity KCl), BP332 (>99% purity Na <sub>2</sub> HPO <sub>4</sub> ), BP362 (>99% purity KH <sub>2</sub> PO <sub>4</sub> )	Phosphate buffered saline (PBS) was prepared in the laboratory as an aqueous solution consisting of 137 mM NaCl, 2.7 mM KCl, 10 mM Na <sub>2</sub> HPO <sub>4</sub> , and 1.8 mM

			KH <sub>2</sub> PO <sub>4</sub> dissolved in ultrapure water. Reagents were measured out using an analytical balance, and glassware was cleaned with soap and water followed by autoclaving immediately prior to use.
Chloroform			
Diamond tip AFM probe	Bruker	PDNISP	Pre-mounted factory-calibrated cube corner diamond ( $E = 1140$ GPa) tip AFM probe (nominal $R = 40$ nm) with a stainless steel cantilever (nominal $k = 225$ N/m, $f_0 = 50$ kHz). Spring constant is measured at the factory ( $k = 256$ N/m for the probe, Serial #13435414, used here) and calibration data (including AFM images of indents showing probe geometry) is provided with the probe.
Diamond ultramicrotome blade	Diatome	Ultra 35°	2.1 mm width. Also used a standard glass blade for initial rough cut of sample surface before transitioning to diamond blade for final surface preparation
Epoxy	Gorilla Glue	26853-31-6	Epoxy resin and hardener were mixed in a 1:1 ratio, a small drop was placed on a stainless steel sample puck (Ted Pella), and V1 grade muscovite mica (Ted Pella) was attached to create an atomically flat surface for preparation of phospholipid membranes.
Ethanol			
LR white resin, medium grade (catalyzed)	Electron Microscopy Sciences	14381	500 mL bottle, Lot #150629
Mesenchymal stem cells (MSCs)	N/A	N/A	MSCs for nanomechanical studies were primary cells harvested from 8-10 week old male C57BL/6 mice as described in Goelzer, M. et al. "Lamin A/C Is Dispensable to Mechanical Repression of Adipogenesis" <i>Int J Mol Sci</i> <b>22</b> : 6580 (2021) doi:10.3390/ijms22126580 and Peister, A. et al. "Adult stem cells from bone marrow (MSCs) isolated from different strains of inbred mice vary in surface epitopes, rates of proliferation, and differentiation potential" <i>Blood</i> <b>103</b> : 1662-1668 (2004), doi:10.1182/blood-2003-09-3070.
Modulus standards	Bruker	PFQNM-SMPKIT-12M	Used HOPG ( $E = 18$ GPa) and PS ( $E = 2.7$ GPa). Also contains 2x PDMS (Tack 0, $E = 2.5$ MPa; Tack 4, $E = 3.5$ MPa), PS-LDPE ( $E = 2.0/0.2$ GPa), fused silica ( $E = 72.9$ GPa), sapphire ( $E = 345$ GPa), and tip characterization (titanium roughness) sample. All samples come pre-mounted on a 12 mm diameter steel disc (sample puck).
Muscovite mica	Ted Pella	50-12	12 mm diameter, V1 grade muscovite mica
Nanscope Analysis	Bruker	Version 2.0	Free AFM image processing and analysis software package, but designed for, and proprietary/limited to Bruker AFMs; similar functionality is available from free, platform-independent AFM image processing

			and analysis software packages such as Gwyddion, WSxM, and others. Has built-in capabilities for force curve analysis, but AtomicJ is more flexible/full featured (e.g., more built-in contact mechanics models to choose from, statistical analysis of force curve fitting results, etc.) for force curve analysis and handles batch processing of force curves.
Phospholipids: POPC, Cholesterol (ovine)	Avanti Polar Lipids	POPC: CAS # 26853-31-6, Cholesterol: CAS # 57-88-5	POPC lipid dissolved in chloroform (25 mg/mL) was obtained from vendor and used without further purification. Cholesterol powder from the same vendor was dissolved in chloroform (20 mg/mL).
Probe holder (fluid, lipid bilayers)	Bruker	MTFML-V2	Specific to the particular AFM used; MTFML-V2 is a glass probe holder for scanning in fluid on a MultiMode AFM.
Probe holder (fluid, MSCs)	Bruker	FastScan Bio Z-scanner	Used with Dimension FastScan head (XY flexure scanners). Serial number MXPOM5-1B154.
Probe holder (standard, ambient)	Bruker	DAFMCH	Specific to the particular AFM used; DAFMCH is the standard contact and tapping mode probe holder for the Dimension Icon AFM, suitable for nanoindentation (PF-QNM, FFV, and point-and-shoot ramping)
Sample Puck	Ted Pella	16218	Product number is for 15 mm diameter stainless steel sample puck. Also available in 6 mm, 10 mm, 12 mm, and 20 mm diameters at <a href="https://www.tedpella.com/AFM_html/AFM.aspx#anchor842459">https://www.tedpella.com/AFM_html/AFM.aspx#anchor842459</a>
Sapphire substrate	Bruker	PFQNM-SMPKIT-12M	Extremely hard surface ( $E = 345$ GPa) for measuring deflection sensitivity of probes (want all of the deflection to come from the probe, not the substrate). Part of the PF-QNM/modulus standards kit.
Scanning electron microscope	Hitachi	S-3400N-II	Located at Boise State. Used to perform co-localized SEM/EDS on all samples except additively manufactured (AM) Ti-6Al-4V.
Silicon AFM probes (standard)	NuNano	Scout 350	Standard tapping mode silicon probe with reflective aluminum backside coating; $k = 42$ N/m (nominal), $f_0 = 350$ kHz. Nominal $R = 5$ nm. Also available uncoated or with reflective gold backside coating. Probes with similar specifications are available from other manufacturers (e.g., Bruker TESPA-V2).
Silicon AFM probes (stiff)	Bruker	RTESPA-525, RTESPA-525-30	Rotated tip etched silicon probes with reflective aluminum backside coating; $k = 200$ N/m (nominal), $f_0 = 525$ kHz. Nominal $R = 8$ nm for RTESPA-525, $R = 30$ nm for RTESPA-525-30. Spring constant of each RTESPA-525-30 is measured individually at the factory via laser Doppler vibrometry and supplied with the probe.
Silicon carbide grit paper (abrasive discs)	Allied	50-10005	120 grit

Silicon nitride AFM probes (soft, large radius hemispherical tip)	Bruker	MLCT-SPH-5UM, MLCT-SPH-5UM-DC	Also MLCT-SPH-1UM-DC. New product line of factory-calibrated (probe radius and spring constants of all cantilevers) large radius ( $R = 1$ or $5$ mm) hemispherical tip (at the end of a $23$ mm long cylindrical shaft) probes. DC = drift compensation coating. 6 cantilevers/probe (A-F). Nominal spring constants: A, $k = 0.07$ N/m; B, $k = 0.02$ N/m; C, $k = 0.01$ N/m; D, $k = 0.03$ N/m; E, $k = 0.1$ N/m; F, $k = 0.6$ N/m.
Silicon nitride AFM probes (soft, medium sharp tip)	Bruker	DNP	4 cantilevers/probe (A-d). Nominal spring constants: A, $k = 0.35$ N/m; B, $k = 0.12$ N/m; C, $k = 0.24$ N/m; D, $k = 0.06$ N/m. Nominal radii of curvature, $R = 10$ nm.
Silicon nitride AFM probes (soft, sharp tip)	Bruker	ScanAsyst-Air	Nominal values: resonance frequency, $f_0 = 70$ kHz; spring constant, $k = 0.4$ N/m; radius of curvature, $R = 2$ nm. Designed for force curve based AFM imaging.
Superglue	Henkel	Loctite 495	Cyanoacrylate based instant adhesive. Lots of roughly equivalent products are readily available.
Syringe pump	New Era Pump Systems	NE1000US	One channel syringe pump system with infusion and withdrawal capacity
Tip characterization standard	Bruker	PFQNM-SMPKIT-12M	Titanium (Ti) roughness standard. Part of the PF-QNM/modulus standards kit.
Ultrahigh purity nitrogen (UHP $N_2$ ), 99.999%	Norco	SPG TUHPNI - T	T size compressed gas cylinder of ultrahigh purity (99.999%) nitrogen for drying samples
Ultramicrotome	Leica	EM UC6	Equipped with a glass blade (standard, for initial sample preparation) and a diamond blade (for final preparation)
Ultrapure water	Thermo Fisher	Barnstead Nanopure Model 7146	Model has been discontinued, but equivalent products are available. Produces $\geq 18.2$ M $\Omega$ *cm ultrapure water with 1-5 ppb TOC (total organic content), per inline UV monitoring. Includes $0.2$ $\mu$ m particulate filter, ion exchange columns, and UV oxidation chamber.
Variable Speed Grinder	Buehler	EcoMet 3000	Used with silicon carbide grit papers during hand polishing.
Vibration isolation table (active)	Herzan	TS-140	Used with Bruker MultiMode AFM. Sits on a TMC 65-531 vibration isolation table. Bruker Dimension Icon AFM utilizes strictly passive vibration isolation (comes from manufacturer with custom acoustic hood, air table, and granite slab).
Vibration isolation table (passive)	TMC	65-531	35" x 30" vibration isolation table with optional air damping (disabled). Used with Bruker MultiMode AFM. Herzan TS-140 "Table Stable" active vibration control table is located on top.