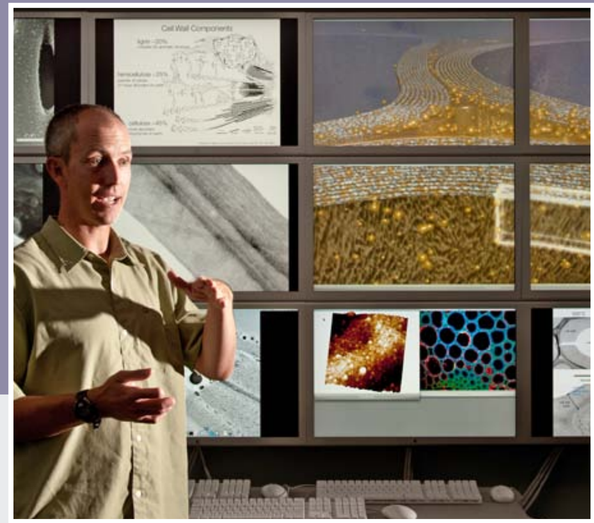
Biomass Surface Characterization Laboratory

Multi-scale, multi-mode imaging tools to understand the recalcitrant nature of biomass feedstocks and the performance of techniques to deconstruct biomass



In the cellular visualization room of the BSCL, NREL scientists can look at different views of the ultra-structures of pretreated biomass materials. *Photo by Dennis Schroeder, NREL/PIX 17880*

The Biomass Surface Characterization Laboratory (BSCL) is an integrated 2,800 square foot facility dedicated to advanced characterization of biomass substrates.

NREL's BSCL provides:

- Multi-scale imaging tools including light, electron, and scanning probe microscopes
- Micron- to nanometer-scale structural analysis of plant, algal, and microbial cells
- Spatial, chemical, and time domain information.

All of these features contribute to understanding the recalcitrant nature of biomass feedstocks.

BSCL imaging capabilities include:

- Confocal microscopy and Raman microscopy
- Transmission electron microscopy and tomography
- Scanning electron microscopy and energy dispersive spectroscopy
- Atomic force microscopy.

Biomass specific preparation and microtomy techniques to optimize imaging include:

- High-resolution carbon coating and sputter coating
- Critical point drying and freeze drying
- Cryo-preservation and embedding
- Thin and ultra-thin microtomy
- Chemical and immunological staining.

Equipment Specifications

System	Capabilities
Confocal microscopes Nikon E800 C1 Nikon Eclipse C1si Spectral Confocal	Laser wavelengths of 407 nm, 488 nm, 543nm Modular photomultiplier detector unit (PMT)
Raman microscope Horiba Scientific LabRAM HR	Fast Raman imaging modules with acquisition times down to <5 ms/point
Scanning electron microscope FEI Quanta FEG 400	High-vacuum, low-vacuum and ESEM modes In-situ imaging at temperatures up to 1,200°C 60 mm motorized z-range of the stage Energy dispersive X-ray analysis
Transmission electron microscope FEI G20 Tecnai 200kV	200 kV LaB6 filament Gatan Ultrascan 2K x 2K CCD digital camera with 4 port output High-tilt 360° rotation holder to acquire ± 70° tilt series Energy dispersive X-ray analysis
Atomic force microscopes Veeco MultiMode PicoForce and Veeco BioScope	Force-measurement features and proven SPM technology NanoScope software
Image processing and analysis	EZ C1, AutoQuant, Nikon Elements, Image Pro Plus, ImageJ, IMOD, Chimera, MatLab

NREL is a national laboratory of the U.S. Department of Energy, Office of Energy Efficiency and Renewable Energy, operated by the Alliance for Sustainable Energy, LLC.

Imaging Applications

Confocal scanning laser microscopy (CSLM)

- Eliminates image degrading out-of-focus information
- Performs serial, optical sectioning through thick specimens
- Monitors tissue-scale cellular dislocation.

Confocal Raman microscopy

- Non-destructive technique generating chemical images using the sample's Raman spectrum
- Allows direct overlay of chemical and spatial data
- Maps cell wall chemistry without stains or labels.

Transmission electron microscopy (TEM)

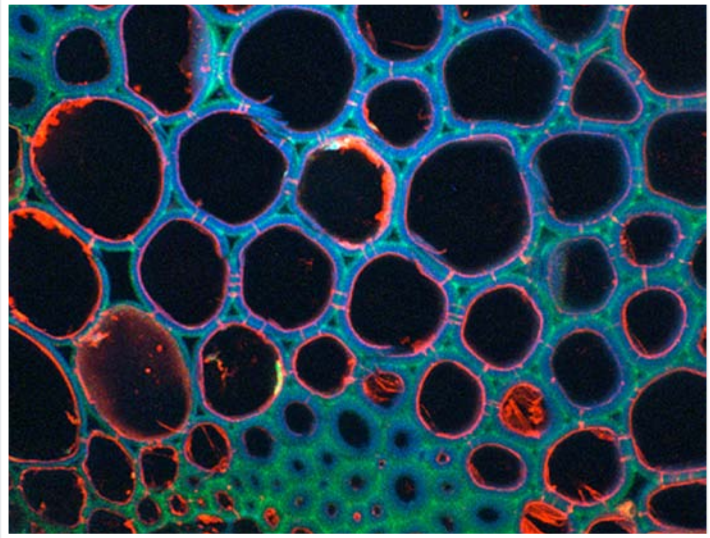
- Leading imaging technique for nano-scale biological research
- Allows the user to visualize the internal structure of materials of biological or non-biological origin
- Detects cell wall decontruction.

Scanning electron microscopy (SEM)

- The most versatile and widely used tool for surface characterization
- Allows the study of the surface morphology of both biological and non-biological materials
- Analyzes changes in surface area.

Atomic force microscopy (AFM)

- Images the topography of the sample surface at nanometer resolution
- Measures the attractive and repulsive forces between the scanning probe tip and the sample surface
- Images surface exposed cellulose microfibrils.



BSCL scientists use antibody and carbohydrate binding module probes to map the distribution of specific cell wall components within biomass samples, as shown in this confocal scanning laser microscope image of a cross section of a corn stover stem. *Image by BSCL, NREL/PIX 20331*

Associated publications

Donohoe, B.S., et al. (2011) "Surface and Ultrastructural Characterization of Raw and Pretreated Switchgrass." *Bioresour. Technol.* (102); pp. 11097-11104.

Haas, T.J., et al. (2009). "Real-Time and Post-Reaction Microscopic Structural Analysis of Biomass Undergoing Pyrolysis." *Energy Fuels* (23); pp. 3810-3817.

Donohoe, B.S., et al. (2009). "Detecting Cellulase Penetration into Corn Stover Cell Walls by Immuno-Electron Microscopy." *Biotechnol. Bioeng.* (103); pp. 480-489.

Brunecky, R., et al. (2009). "Redistribution of Xylan in Maize Cell Walls during Dilute Acid Pretreatment." *Biotechnol. Bioeng.* (102); pp. 1537-1543.

Donohoe, B.S., et al. (2008). "Visualizing Lignin Coalescence and Migration through Maize Cell Walls Following Thermochemical Pretreatment." *Biotechnol. Bioeng.* (101); pp. 913-925.

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NREL is a national laboratory of the U.S. Department of Energy
Office of Energy Efficiency and Renewable Energy
Operated by the Alliance for Sustainable Energy, LLC

NREL/FS-5100-54589 • April 2012