

Speed and power of X-ray laser helps unlock molecular mysteries

New nanocrystallography technique shines light on biomolecules in action

By outrunning a laser's path of destruction, an international research team has created 3D images of fragile but biologically important molecules inside protein nanocrystals. Using the Linac Coherence Light Source (LCLS), a powerful X-ray laser at the SLAC National Accelerator Laboratory in Menlo Park, Calif., the scientists fired femtosecond (one quadrillionth of a second) bursts of light at a stream of tumbling molecules, obliterating them as they pass, but not before capturing otherwise illusive images of their crystalline structures.

An overview and early results of this new imaging technique will be presented at the 2012 meeting of the American Crystallographic Association (ACA), which takes place July 28 – Aug. 1 in Boston, Mass.

"These laser pulses are so brief that we are able to outrun the radiation's damaging effects," said John C.H. Spence of Arizona State University, one of more than 70 international researchers from institutions including SLAC; DESY, the German Electron Synchrotron; and the Max-Planck Institute in Heidelberg, Germany.

"Using this so-called 'diffract-then-destroy' approach, our research team recorded about a hundred scattering patterns per second from protein nanocrystals," said Spence. "This is an important step toward the making of movies of biomolecules at work."

In traditional crystallography, a beam of X-rays first interacts with a crystal and then appears on a photo-detector as diffraction spots of greater and lesser intensity. These patterns encode the density of electrons in the crystal, enabling scientists to determine the three-dimensional position of atoms, chemical bonds, and other information. To obtain this information, the crystal is frozen, to reduce radiation damage, and placed on a rotating mount and bombarded with X-rays as its orientation is changed. A scattering pattern is slowly built up and the 3D structure can eventually be deduced.

This traditional method of using frozen molecules, however, prevents observation of the molecules at work in their native liquid environment at room temperature.

To obtain images of these molecules in the more natural state, the researchers sent the protein nanocrystals streaming in a single-file micron-sized droplet beam (rather like an ink-jet printer) in vacuum across the X-ray beam, in a method developed at Arizona State University.

Next they fired incredibly brief bursts of X-ray laser light, about 100 times each

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second, at the molecules in the droplet beam, and detected the scattered X-ray patterns from each particle before the intensity of the beam blasted them apart. The researchers were able to combine these millions of snapshots to build up 3D models of the molecules with atomic-scale resolution.

One particular molecule that was studied this way was Photosystem 1-ferredoxin, which is the chemical powerhouse that drives photosynthesis. The molecules for this experiment were made in the laboratory of Arizona State University researcher Petra Fromme.

Photosystem 1 harnesses sunlight to split water to make the oxygen we breathe, absorb carbon dioxide, and produce sugars, which maintains our biosphere. These molecules were studied "in action" by exciting them with a pulse of green laser light (to mimic the effect of sunlight falling on a leaf) a few microseconds before taking their X-ray snapshot. Each snapshot then became one frame of a movie. By changing the delay between green pulse and X-ray pulse, the researchers could create a 3D movie of a biomolecule in action.

"Many other groups we are supporting now are applying the method to other proteins, such as enzymes, drug molecule targets, and imaging chemical reactions as they develop along the liquid jet," said Spence. "The important thing was to get atomic-resolution snapshot images from nanocrystals at room temperature without radiation damage."

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A complete listing of the collaborating research institutions follows:

- Center for Free-Electron Laser Science, DESY, Hamburg, Germany
- Photon Science, DESY, Hamburg, Germany
- Department of Chemistry and Biochemistry, Arizona State University, Tempe
- Department of Physics, Arizona State University
- Laboratory of Molecular Biophysics, Department of Cell and Molecular Biology, Uppsala University, Sweden
- Department of Chemistry, Biochemistry, and Biophysics, Göteborg University, Sweden
- Max Planck Advanced Study Group, Center for Free Electron Laser Science (CFEL), Hamburg, Germany
- Max-Planck-Institut für medizinische Forschung, Heidelberg, Germany
- PULSE Institute and SLAC National Accelerator Laboratory, Menlo Park, Calif.
- LCLS, SLAC National Accelerator Laboratory
- CEA, Institut de Biologie et de Technologies de Saclay, France
- European XFEL GmbH, Hamburg, Germany
- Department of Physics, Cornell University, Ithaca, New York
- Max-Planck-Institut für Kernphysik
- Lawrence Livermore National Laboratory, Livermore, Calif.
- PNSensor GmbH, München, Germany
- Max-Planck-Institut Halbleiterlabor, München, Germany
- Advanced Light Source, Lawrence Berkeley National Laboratory

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- University of Hamburg,
- Max-Planck-Institut für extraterrestrische Physik, Garching, Germany

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MORE INFORMATION ABOUT THE 2012 ACA MEETING

The ACA is the largest professional society for crystallography in the United States, and this is its main meeting. All scientific sessions, workshops, poster sessions, and events will be held at the Westin Waterfront Hotel in Boston, Mass.

USEFUL LINKS:

Main meeting website: <http://www.amerystalassn.org/2012-meeting-homepage> [1]

Meeting program: <http://www.amerystalassn.org/2012-tentative-program> [2]

Meeting abstracts: <http://www.amerystalassn.org/app/sessions> [3] Exhibits: <http://www.amerystalassn.org/2012-exhibits> [4]

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