

# Researchers use X-ray diffraction microscope to reveal 3-D internal structure of whole cell

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(PhysOrg.com) -- Using the new technique, researchers were able to identify the 3-D morphology and structure of cellular organelles, including the cell wall, vacuole, endoplasmic reticulum, mitochondria, granules and nucleolus.

Three-dimensional imaging is dramatically expanding the ability of researchers to examine biological specimens, enabling a peek into their internal structures. And recent advances in X-ray diffraction methods have helped extend the limit of this approach.

While significant progress has been made in [optical microscopy](#) to break the diffraction barrier, such techniques rely on fluorescent labeling technologies, which prohibit the quantitative 3-D imaging of the entire contents of cells. Cryo-electron microscopy can image structures at a resolution of 3 to 5 nanometers, but this only works with thin or sectioned specimens.

And although X-ray protein crystallography is currently the primary method used for determining the 3-D structure of [protein molecules](#), many biological specimens — such as whole cells, cellular organelles, some viruses and many important protein molecules — are difficult or impossible to crystallize, making their structures inaccessible. Overcoming these limitations requires the employment of different techniques.

Now, in a paper published today in [Proceedings of the National](#)

[Academy of Sciences](#), UCLA researchers and their collaborators demonstrate the use of a unique X-ray diffraction microscope that enabled them to reveal the internal structure of yeast spores. The team reports the quantitative 3-D imaging of a whole, unstained cell at a resolution of 50 to 60 nanometers using X-ray diffraction microscopy, also known as lensless imaging.

Researchers identified the 3-D morphology and structure of cellular organelles, including the cell wall, vacuole, endoplasmic reticulum, mitochondria, granules and nucleolus. The work may open a door to identifying the individual protein molecules inside whole cells using labeling technologies.

The lead authors on the paper are Huaidong Jiang, a UCLA assistant researcher in physics and astronomy, and John Miao, a UCLA professor of physics and astronomy. The work is a culmination of a collaboration started three years ago with Fuyu Tamanoi, UCLA professor of microbiology, immunology and molecular genetics. Miao and Tamanoi are both researchers at UCLA's California NanoSystems Institute. Other collaborators include teams at Riken Spring 8 in Japan and the Institute of Physics, Academia Sinica, in Taiwan.

"This is the first time that people have been able to peek into the 3-D internal structure of a biological specimen, without cutting it into sections, using X-ray diffraction microscopy," Miao said.

"By avoiding use of X-ray lenses, the resolution of X-ray diffraction microscopy is ultimately limited by radiation damage to biological specimens. Using cryogenic technologies, 3-D imaging of whole biological cells at a resolution of 5 to 10 [nanometers](#) should be achievable," Miao said. "Our work hence paves a way for quantitative 3-D imaging of a wide range of biological specimens at nanometer-scale resolutions that are too thick for [electron microscopy](#)."

Tamanoi prepared the yeast spore samples analyzed in this study. Spores are specialized cells that are formed when they are placed under nutrient-starved conditions. Cells use this survival strategy to cope with harsh conditions.

"Biologists wanted to examine internal structures of the spore, but previous microscopic studies provided information on only the surface features. We are very excited to be able to view the spore in 3-D", Tamanoi said. "We can now look into the structure of other spores, such as Anthrax spores and many other fungal spores. It is also important to point out that yeast spores are of similar size to many intracellular organelles in human cells. These can be examined in the future."

Since its first experimental demonstration by Miao and collaborators in 1999, coherent diffraction microscopy has been applied to imaging a wide range of materials science and biological specimens, such as nanoparticles, nanocrystals, biomaterials, cells, cellular organelles, viruses and carbon nanotubes using X-ray, electron and laser facilities worldwide. Until now, however, the radiation-damage problem and the difficulty of acquiring high-quality 3-D diffraction patterns from individual whole cells have prevented the successful high-resolution 3-D imaging of biological [cells](#) by X-ray diffraction.

**More information:** Research paper: [www.pnas.org/content/early/2010/06/07/1000156107.abstract](http://www.pnas.org/content/early/2010/06/07/1000156107.abstract)

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