# Researchers develop new device to help image key proteins at room temperature

December 20 2013, by John Spence

A group of researchers from Arizona State University are part of a larger team reporting a major advance in the study of human proteins that could open up new avenues for more effective drugs of the future. The work is being reported in this week's *Science* magazine.

In the paper, "Serial femtosecond crystallography of G-protein-coupled receptors," the team reports that they have been successful in imaging at room temperature the structure of G protein-coupled receptors (GPCR) with the use of an x-ray free-electron laser.

GPCR's are a highly diverse group of membrane proteins that mediate cellular communication. Because of their involvement in key physiological and sensory processes in humans, they are thought to be prominent drug targets.

The method described in the paper was applied for the first time to this important class of proteins, for which the 2012 Nobel Prize was awarded to Brian Kobilka and Robert Lefkowitz, said John Spence, an Arizona State University professor of physics. Spence also is the director of science at National Science Foundation's BioXFEL Science and Technology Center and a team member on the *Science* paper.

"These GPCR's are the targets of a majority of drug molecules," Spence said, but they are notoriously difficult to work with. This is the first time structural observations of the GPCR's have been made at room temperature, allowing researchers to overcome several disadvantages of

previous imaging methods of the proteins.

"Normally, protein crystallography is performed on frozen samples, to reduce the effects of radiation damage," Spence said. "But this new work was based on an entirely new approach to protein crystallography, called SFX (Serial Femtosecond Crystallography) developed jointly by ASU, the Deutsches Elektronen-Synchrotron (DESY) and the SLAC National Accelerator Laboratory."

"This method uses brief pulses of x-rays instead of freezing the sample to avoid damage, and so it reveals the structure which actually occurs in a cell at room temperature, not the frozen structure," Spence added. "The 50 femtosecond pulses (120 per second) 'outrun' radiation damage, giving a clear picture of the structure before it is vaporized by the beam."

The femtosecond crystallography technique could enable researchers to view molecular dynamics at a time-scale never observed before. Spence said the method basically operates by collecting the scattering for the image so quickly that images are obtained before the sample is destroyed by the x-ray beam.

By 'outrunning' radiation-damage processes in this way, the researchers can record the time-evolution of molecular processes at room temperature, he said.

Spence said ASU played a crucial role in the project described in *Science*, through the invention by Uwe Weierstall (an ASU physics professor) of an entirely new device for sample delivery suited to this class of proteins.

The lipic cubic phase (LCP) injector that Weierstall developed replaces the continuous stream of liquid (which sends a continuously refreshed

stream of proteins across the pulsed x-ray beam) with a slowly moving viscous stream of 'lipid cubic phase solution,' which has the consistency of automobile grease.

"We call it our 'toothpaste jet,'" Spence said.

He added that the LCP solves three problems associated with previous SFX work, and made this new work possible:

- The viscosity slows the flow rate so the crystals emerge at about the same rate as the x-ray pulses come along, hence no protein is wasted. This is important for the study of human protein, which is more costly than diamond on a per gram basis.
- The "hit rate" is very high. Nearly all x-ray pulses hit protein particles.
- Most important, LCP is itself a growth medium for protein nanocrystals.

"A big problem with the SFX work we have been doing over the past four years is that people did not know how to make the required nanocrystals," Spence said. "Now it seems many can be grown in the LCP delivery medium itself."

**More information:**
www.sciencemag.org/content/342/6165/1521.abstract

Provided by Arizona State University