New optogenetic tool moves proteins within cells to study biological changes

April 18 2016, by Mark Derewicz

Scientists at the University of North Carolina School of Medicine have developed a way to embed light-responsive switches into proteins so that researchers can use lasers to manipulate protein movement and activity within living cells and animals.

Using this technique, the UNC team of scientists forced proteins out of the cell nucleus and into the cytoplasm, where they could no longer do their jobs. The researchers then watched in real time as the cell responded to its personnel shortfall; the team discovered that the resulting cellular processes were more dynamic than previously expected.

The findings, published today in the journal *Nature Chemical Biology*, demonstrate the value of new research approaches that can rapidly probe the function of genes and proteins.

"By the time you get your hands on a knockout mouse for a particular gene or protein, the cells that had that protein have already adapted to their new circumstances of having one of its genes taken away; everything has changed," said senior author Brian Kuhlman, PhD, professor of biochemistry and biophysics. "By using light, we can inactivate a protein instantaneously. We can do it in a specific type of cell, at a specific moment in development. This can give us the resolution we need to truly understand the function of a particular protein."

Typically, when scientists want to learn about a biological system (a cell, organ, or animal), they make a change and then observe what happens. In biology, this is often accomplished by "knocking out" or deleting a specific gene. For instance, a researcher interested in whether a protein is important in cancer might remove the gene for that protein, and look to see how it affects tumor formation. One of the problems with this method is that it creates a permanent change, and therefore the biological system has a chance to compensate before anyone can study it.

Kuhlman and his colleagues wanted to develop a strategy that would allow scientists to rapidly activate (or inactivate) a protein with the pinpoint precision of lasers. The approach is part of a growing discipline called optogenetics, where beams of light can act like the strings of a puppeteer to direct activities within cells. In this study, the researchers decided to use optogenetics to control the activity of proteins by controlling their location.

They started with a plant protein called AsLOV2 that changes its shape in response to light. The researchers attached a short amino acid sequence to the protein AsLOV2; this sequence postmarked the protein for the cytoplasm. In the dark, this nuclear export signal remained locked tightly in its "photocage." But when it was bathed in blue light, it was released and sent proteins out of the nucleus.

Lead study author Hayretin Yumerefendi, PhD, a postdoctoral fellow in the Kuhlman lab, fused this construct to a fluorescent protein and then expressed these protein chimeras in mouse cells. When he first looked at the cells under the microscope, he could see tiny red fluorescent orbs crowded inside the nucleus. After he exposed the cells to a certain wavelength of light, he found the red dots had traveled into the cytoplasm.

Yumerefendi then embedded these light switches into two proteins

called LexA and Bre1 that act on DNA and thus normally reside in the nucleus. In both cases, he found that the proteins traveled into the cytoplasm after photoactivation. What's more, he showed that this move was accompanied by a loss in protein activity. Yumerefendi and his colleagues were surprised to learn that the cells adapted quickly to their new normal. For example, they found that the chemical tags that Bre1 sticks onto DNA disappeared in a matter of minutes when Bre1 was removed with light.

"One of the key discoveries we made was that these cellular processes, which were thought to be relatively slow, are actually quite dynamic," said Yumerefendi. "They happen on timescales that are 30 times faster than previously thought. Our finding emphasizes how important it is that we develop new ways to watch biological events in real time."

Proteins can play different roles at different stages of development, in different parts of an organism, and during various disease states. Therefore, the researchers are planning to apply their new optogenetic tool to study the function of different proteins and examine how the "behavior" of these proteins changes depending on both time and space.

More information: Light-induced nuclear export reveals rapid dynamics of epigenetic modifications, *Nature Chemical Biology*, DOI: 10.1038/nchembio.2068

Provided by University of North Carolina Health Care

Citation: New optogenetic tool moves proteins within cells to study biological changes (2016, April 18) retrieved 19 July 2024 from https://phys.org/news/2016-04-optogenetic-tool-proteins-cells-biological.html

This document is subject to copyright. Apart from any fair dealing for the purpose of private study or research, no part may be reproduced without the written permission. The content is provided for information purposes only.