

## How Do Cells Die? Biophotonic Tools Reveal Real-Time Dynamics in Living Color

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Paras Prasad and a team of researchers have developed a biophotonic imaging approach to monitor programmed cell death. Credit: Douglas Levere University at Buffalo

Apoptosis, programmed cell death, is essential to normal development, healthy immune system function, and cancer prevention. The process dramatically transforms cellular structures but the limitations of conventional microscopy methods have kept much about this structural reorganization a mystery.

Now, in research featured on the cover of the current issue of



<u>Proceedings of the National Academy of Sciences</u>, University at Buffalo scientists have developed a biophotonic imaging approach capable of monitoring in real-time the transformations that cellular macromolecules undergo during <u>programmed cell death</u>.

The work could help realize the potential of customized molecular medicine, in which chemotherapy, for example, can be precisely targeted to cellular changes exhibited by individual patients. It can also be a valuable drug development tool for screening new compounds.

"This new ability provides us with a dynamic mapping of the transformations occurring in the cell at the molecular level," says study co-author Paras N. Prasad, PhD, executive director of the UB Institute for Lasers, Photonics and Biophotonics (ILPB) and SUNY Distinguished Professor in the departments of Chemistry, Physics, Electrical Engineering and Medicine. "It provides us with a very clear visual picture of the dynamics of proteins, DNA, RNA and lipids during the cell's disintegration."

Prasad notes that <u>molecular medicine</u>, in which treatments or preventive measures can be tailored to cellular properties exhibited by individual patients, depends on much better methods of visualizing what's happening during critical cellular processes.

"This research helps improve our understanding of cellular events at the molecular level," he says. "If we know that specific molecular changes constitute an early signature of a disease, or what changes may predispose a patient to that disease, then we can take steps to target treatment or even prevent the disease from developing in the first place."

To capture the cellular images, the interdisciplinary UB team of biologists, chemists and physicists, led by Prasad, utilized an advanced biophotonic approach that combines three techniques: a nonlinear,



optical imaging system (CARS or Coherent anti-Stokes Raman scattering), TPEF (two-photon excited fluorescence), which images living tissue and cells at deep penetration and Fluorescence Recovery after Photobleaching to measure dynamics of proteins.

"For the first time, this approach allows us to monitor in a single scan, four different types of images, characterizing the distribution of proteins, DNA, RNA and lipids in the cell," says Aliaksandr V. Kachynski, PhD, research associate professor at the ILPB and co-author.

The resulting composite image integrates in one picture the information on all four types of biomolecules, with each type of molecule represented by a different color: proteins in red, RNA in green, DNA in blue and lipids in grey, as shown on the PNAS cover.

Multiplex imaging provided new information on the rate at which proteins diffuse through the cell nucleus, the UB scientists say.

Before apoptosis was induced, the distribution of proteins was relatively uniform, but once apoptosis develops, nuclear structures disintegrate, the proteins become irregularly distributed and their diffusion rate slows down, says Artem Pliss, PhD, research assistant professor at the ILPB and co-author on the paper.

"This research gives us the unique ability to study and improve our understanding of individual subcellular structures and the transformations they go through," says Pliss.

Such precise information will be especially useful for monitoring how specific cancer drugs affect individual cells.

"For example, say drug therapy is being administered to a cancer patient; this system will allow for the monitoring of cellular changes throughout



the treatment process," notes Kachynski. "Clinicians will be able to determine the optimal conditions to kill a cancer cell for the particular type of disease. An improved understanding of the drug-biomolecule interactions will help discover the optimal treatment doses so as to minimize side effects."

Andrey Kuzmin, PhD, research assistant professor at the ILPB and coauthor, adds that a new paper from the UB team, forthcoming in Biophysical Journal, further extends this work.

"The benefits of the UB multiplex imaging system and its molecular selectivity have been further extended into a new fundamental cellular study, structural reorganization throughout the mitotic cell cycle," he says.

## Provided by University at Buffalo

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