# Scientists track chemical changes in cells as they endure extreme conditions

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One of nature's most gripping feats of survival is now better understood. For the first time, scientists from the U.S. Department of Energy's Lawrence Berkeley National Laboratory observed the chemical changes in individual cells that enable them to survive conditions that should kill them.

The team tracked the chemical changes in Desulfovibrio vulgaris, which is a single-cell bacterium that normally can only exist in an oxygen-free environment. They exposed the cells to the most hostile of conditions — air — and watched as some cells temporarily survived by initiating a well-orchestrated sequence of chemical events.

Until now, scientists have not been able to monitor, at a molecular level, the chemical changes in individual cells as they survive extreme conditions. The ability to watch this Herculean adaptation to stress, from such an up-close and real-time vantage, gives scientists an improved way to study adaptive responses in a range of microbes, such as disease-causing pathogens and microbes that play a role in photosynthesis, energy production, and geochemical phenomena. Their work was recently published online in the journal Proceedings of the National Academy of Sciences.

"We can now follow chemical changes in living bacteria as they respond to extreme environments. This opens up a new window into how bacteria adapt and carry out some of life's most important processes," says Hoi-Ying Holman, a staff scientist in Berkeley Lab's Earth Sciences Division.

To achieve this milestone, the team used the Advanced Light Source, a synchrotron and national user facility located at Berkeley Lab that generates bright light for scientific research. In a pioneering approach, they used a beamline equipped with a high-resolution infrared microscope to detect the molecular signatures of a cell's biochemical and metabolic activity, such as spikes in levels of free radicals and organic acids.

Specifically, the infrared microscope tracks the instantaneous response of hydrogen bond structures in cellular water as their immediate surroundings change. The spectral measurements indicate changes in hydrogen bonds, which in turn indicate changes in the presence of ions and various molecules such as radicals and metabolites.

First, the team exposed D. vulgaris to their much-preferred oxygen-free environment. They then exposed the same group of cells to air and monitored them for several hours.

The foreign environment proved too much for many cells; they died due to a toxic accumulation of free radicals. But some cells with adequate stores of energy survived.

And for the first time, thanks to the high-sensitivity synchrotron infrared beamline, the scientists watched as the surviving cells unleashed a series of metabolic changes that enabled them to endure in the presence of oxygen, like a fish out of water.

"We monitored several molecules simultaneously in the same bacteria, and watched their metabolic response to stress and extreme conditions," says Holman. "We found that multiple chemical processes allow them to adapt."

The scientists studied D. vulgaris because the bacterium, which is among a class of bacteria that reduce sulfate, plays a critical role in many important geochemical processes such as element and nutrient cycling in soils. It also assists in bioremediation and may someday be used to aid energy production and carbon sequestration efforts.

D. vulgaris also intrigues scientists because it is an obligate anaerobe — meaning it can't survive in the presence of oxygen — yet it participates in many geochemical processes in which oxygen levels fluctuate. For example, it thrives in algae mats, which produce very high concentrations of oxygen during the day.

Scientists have puzzled over this riddle for years. They've studied the bacterium's gene expression, which provides valuable clues to how it adapts. But, as Holman explains, scientists must also understand the ever-changing chemistry inside a cell in order to fully understand its gene expression and adaptive-response pathways.

Scientists have also teased out the chemical changes in the bacterium by studying different cells at various stages in a population of cells. This is not the same as studying the same cell over time, however.

"We are studying the same individuals as opposed to studying the population. We want to watch the same cells over time and not rely on the assumption that all cells within a population behave the same," says Holman. "This work, which represents a new way to study adaptive response in individual cells, is made possible by the great progress we've made in synchrotron infrared cellular imaging."

More information: "Real-time molecular monitoring of chemical environment in obligate anaerobes during oxygen adaptive response" by Hoi-Ying Holman, Eleanor Wozei, Zhang Lin, Luis Comolli, David Ball, Sharon Borglin, Matthew Fields, Terry Hazen, and Kenneth Downing was published June 16, 2009 in the online early edition of the

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