THE AUTHOR FILE

Robert E. Campbell

Developing biosensors takes patience and running through the snow.

To stay fit and healthy, protein engineer Robert Campbell runs the 5 kilometers to and from work every day. By combining commute and exercise, he saves precious time so he can spend more on his research in the chemistry department at the University of Alberta and on his family. He is still exploring how to best deal with the buildup of thick icicles on his eyelashes that threaten to freeze his eyes shut. After all, he runs in the snow and at –20 degrees Celsius.

Yi Shen

Robert E. Campbell in front of an ice sculpture at the University of Alberta

Campbell develops tools for the fluorescent protein (FP) community. He did his PhD work in chemistry at the University of British Columbia, was a postdoctoral fellow in Roger Tsien's lab at the University of California at San Diego, and is now on the faculty at the University of Alberta.

To help biologists, fluorescent sensors must produce a detectable change in fluorescence when an event of interest happens in a cell. Traditionally, fluorescence resonance energy transfer between two FPs has been the only method that enabled researchers to image dynamic protein-protein interactions. Now Campbell and his team have developed another approach to do so: fluorescent protein exchange (FPX). "My hope is that the community will embrace FPX technology and do things with it that we never imagined," he says.

Usually, he says, "we would need to endlessly fiddle with the biosensor to get it to normally work well." The FPX biosensors work without additional optimization. FPX converts changes in protein interaction or protein localization into dramatic and reversible shifts between green and red fluorescence.

For most research applications, such as imaging a labeled cytoskeleton, it is better when the FPs are monomers. Dimeric and tetrameric FPs, however, are brighter and more stable than their monomeric equivalents, which suggests that FPs 'prefer' to be oligomers, he says. FPX technology, more specifically the dimerization-dependent FPs, exploits this increased brightness of dimeric FPs.

Dimerization-dependent FPs are fluorescent only when two different proteins, 'A' and 'B', come together. The researchers put two different versions of A into the cell, one that turns green and one that turns red, along with a single version of B.

The biosensor is designed such that the B partner is swapped between the two different A partners when a biochemical event of interest occurs. When B interacts with the first A copy, the cell is green; when it interacts with second, the cell is red. "It's a molecular ménage à trois, if you will," says Campbell. In view of their commercial potential, Campbell has filed a patent for FPX sensors and says he looks forward to working with industry partners to develop robust, new live-cell assays that can be used, for example, in drug discovery.

As a boy, Campbell dreamed of being an inventor not unlike Professor Brainard, the chemist in the Disney movie *The Absent-Minded Professor* who invented Flubber, a rubbery substance with intriguing properties.

Campbell's love of invention-oriented science has guided his career. "However, as I approach my mid-career stage, I'm also finding that focusing on invention alone can be kind of scary," he says. Hypothesis-driven research will always lead to an answer, even if it is not the right answer. "In contrast, invention-driven research is often binary," he says. "It either works or it doesn't work, and the failures are rarely publishable."

> "It's a molecular ménage à trois, if you will."

Campbell says his graduate students offer a steady flow of creativity and imagination and help him sustain this invention-focused model to keep developing FP-based tools.

As with most science, this work takes patience, which comes naturally to him. "Many of my ideas have taken years to come to fruition, so patience is critical," says Campbell. One of his lab's earliest projects was the development of a red fluorescent calcium sensor, work that he began in 2003. But it wasn't until 2011 that he and his team reported a first successful example.

In breakout sessions at scientific conferences, Vincent Pieribone, a neurobiologist at Yale School of Medicine, sees that Campbell is increasingly regarded as a sage in the world of fluorescence indicator science.

Pieribone has a joint grant with Campbell as part of the US National Institutes of Health's Brain Research through Advancing Innovative Neurotechnologies (BRAIN) Initiative. "Robert applies a steely determination as he faces down high-risk problems that have often taken years for him to solve," says Pieribone. "I think he embodies the single-minded resolve and intellectual fortitude that we all strive to achieve as scientists."

Vivien Marx

Ding, Y. *et al.* Ratiometric biosensors based on dimerization-dependent fluorescent protein exchange. *Nat. Methods* **12**, 195–198 (2015).