RACking Up Tools to Move Cell Biology Forward

October 2008--What has a brain, needs to be fed, and moves rapidly when you hold out a “treat”? Why, a migrating cell, of course.

Some cells can “walk” toward chemoattractant “treats” using footlike projections called pseudopods. While it’s known that activating the small GTPase Rac can lead to pseudopod formation, the rest of the molecular events that occur to get a cell to move are not so clear.

Now, the arrival of Takanari Inoue to IBBS’s Center for Cell Dynamics gives Hopkins researchers access to a new technique that can rapidly activate Rac and other proteins to study signals that lead to cell movement in real time.

Moving cells—and the whole field—forward starts with a first step, and Inoue took that step a few years ago by developing a tool to decipher a piece of the biochemical puzzle of cell movement. He figured out what causes a particular lipid to collect at the front edge of the moving cell— the pseudopod.

“Knowing that Rac was involved in getting cells to grow feet and move was a useful starting point, but at the same time confounding. Cells have tricks to adjust themselves after you increase the level of one of their proteins,” says cell biologist, Peter Devreotes, one of Inoue’s collaborators.

“They end up altering the levels of other proteins as well.” According to Inoue, within tens of seconds Rac can activate many different molecules, some of which loop back to Rac itself in a very intricate manner.

These rapid and nonlinear molecular interactions create a complicated molecular web that Inoue lacked the tools to disentangle. Inoue reasoned that if he were to study Rac in isolation, he needed be able to activate Rac so quickly that the side effects wouldn’t have time to occur. After racking his brain, he came up with a method that he thought just might do the trick.

By forcing a Rac activator to the membrane where Rac normally sits, Inoue was able to activate Rac in a record-breaking 10 seconds in single living cells. The technique relies on a controllable switch that can be manipulated to study virtually any signaling proteins at the membrane.

In studying neutrophils—the speediest cells in the bloodstream—Inoue discovered that rapid Rac activation triggers actin polymerization but unexpectedly did not trigger the feedback loop at the leading edge of the cell. By modifying the recruited module in his inducible technique, Inoue was able to directly elevate the lipid at the membrane rather than activating Rac. This caused cells to generate pseudopods, trigger the feedback loop, and subsequently “walk.” “Not only is it fast, it’s also very specific,” says Inoue. “It’s really exciting.”

The importance of the lipid has been supported by another line of evidence. Devreotes has previously suggested that PTEN, which lowers the level of the lipid, is required for suppressing extra pseudopods. Without PTEN, “the cells are sort of confused about where they’re going.”

Unlike Devreotes’ experimental cells, Inoue himself definitely is moving forward.

He plans to continue improving his technique to study cell migration and trying out other proteins. Additionally, he’d like to take his technique one step further and figure out how to rapidly recruit proteins to specific regions of the membrane—the leading edge, for instance.

The technique isn’t limited to studying cell migration, says Devreotes. It can be applied to “just about any field involving signal transduction—cell division, for example—or whatever anyone wants to look at.”Denise Montell, director of the Center for Cell Dynamics, agrees.

“Takanari’s tools will be quite generally useful to everyone who studies cell biology. I think there’ll be lots of opportunities for collaborations to blossom.”

--Michelle Jones