CELL BIOLOGY

How kinases attack signaling networks

Two computational tools find residues that determine the specificity of any kinase and the effects of mutations in these residues on the phenotype of cancer cells.

It is hard to overstate the importance of kinases for the proper functioning of a cell. By phosphorylating their substrates, these enzymes pass information along signaling networks and thereby regulate many cellular functions. Rune Linding from the University of Copenhagen in Denmark has spent most of his career deciphering kinases. Describing himself as a 'signaling guy', he says, "I am very motivated to find out how networks adapt and change in response to specific genetic lesions."

He was bothered by the discrepancy between the number of new mutations being discovered for diseases such as cancer and the number of these mutations with a known phenotypic effect, which is comparatively small. "There is an information gap," he says, "and we wanted to attack it head on."

Five years ago, his team, in particular former PhD student Pau Creixell, started to develop computational tools that aimed to predict the effects of mutations on signaling networks.

The first step was an algorithm to find the residues that are determinants of specificity (DoS) for kinases. Previous studies had characterized the DoS of certain kinases, but Linding wanted a more general tool. His team developed KINspect, trained on large amounts of experimental data about kinases' ability to phosphorylate certain peptides as well as on kinase structures. KINspect starts with a random set of residues for each kinase, then iteratively determines those residues that are crucial for a kinase's specificity.

Specificity determinants often do not correspond to conserved residues and can span regions distant to the substrate-binding sites. Although this finding is not intuitive, it can be explained by the fact that for diversity to arise between kinase families, DoS have to diverge from each other during evolution.

The team validated KINspect on a set of kinases with known DoS and also found a large number of determinants not reported in the literature. "It is always particularly hard to validate tools that can do something you could not do before," says Linding. He hopes that many more researchers will now use KINspect to make and test new predictions.

The predictions of KINspect formed the basis for ReKINect, a tool used to find mutations in DoS that are responsible for changing kinase specificity—so-called network-attacking mutations (NAMs). These can affect networks in several ways: they can disrupt a network by activating or inactivating a kinase, they can rewire a network by creating specificities for new substrates, and they can destroy or create phosphorylation sites. The team used ReKINect to systematically classify known cancer variants for their ability to perturb signaling networks. They validated some of the NAMs and also showed that they could predict cell-proliferation rates using knowledge about how mutations affect entire networks.

Linding hopes that the information gleaned from ReKINect will shed more light on disease mechanisms and translate into better treatments down the line. In collaboration with several research groups in the United States, he is applying ReKINect to decipher network changes in patient tumors grafted into mice with different genetic backgrounds. The goal is to understand how these changes affect drug responses.

Linding also stresses that the new algorithms are not limited to cancer data, or even kinases. "You can expand this framework to other post-translational modifications or the epigenetic code," he says. "We hope that other communities get inspired to study specificity more systematically and thereby better understand mutations."

Interaction with users will be important to hone KINspect and ReKINect. The team plans to set up a feedback forum to encourage people to report software glitches, as well as to discuss new predictions. Although improvements to the code will enhance the performance of the tool, Linding cautions users to not discount their intuition about a system and to ask about any new model that is being developed, "Does it make sense?"

Nicole Rusk

RESEARCH PAPERS

Creixell, P. *et al.* Unmasking determinants of specificity in the human kinome. *Cell* **163**, 187–201 (2015).

Creixell, P. *et al.* Kinome-wide decoding of network-attacking mutations rewiring cancer signaling. *Cell* **163**, 202–217 (2015).