WEB WATCH

From signal to sequence

No scientist is an island and. as the Human Genome Project testifies, we can achieve remarkable things when we work as a team. Biocarta has adopted a community approach to mapping cellular pathways: it provides the tools; the scientific community provides and constantly updates —

The Biocarta website contains several types of information, but its pathway diagrams, which cover all areas of cellular regulation from cell division to apoptosis, immunology to neuroscience, are the main attraction. Clicking on a pathway category gives you a list of pathways: for example, clicking on cell-cycle regulation links to a menu containing the ATM pathway, regulation by cyclins, the p53 pathway and more. Clicking on any single pathway provides a stylized, information-packed diagram that evolves as users provide new information. In the future, pathways will also have 'aurus' responsible for assessing submitted information on their pathway. If you don't agree with what you see, you can send comments to a discussion group and help the pathway to evolve.

Each component of the pathway is a gateway to a wealth of information: clicking on any protein takes you to a table containing links to just about any public-domain database you can think of. Whether you want sequences, structures, information on genetic diseases, or just some relevant abstracts, you can link to them from here.

If your favourite pathway isn't in the list, you can submit it. Biocarta even provides a template with which to draw your pathway diagram. If the template doesn't have the right components, you can send diagrams in any format, "even a paper napkin". So don't sit back and watch this site evolve: join in!

Cath Brooksbank

CELL MOTILITY

The sting in WASP's tail

How are the many stimuli that tell cells to move Inactive WASP translated into changes in actin (Autoinhibited) polymerization? Evidence points to Polyproline members of the Wiskott-Aldrich CDC42-binding Acidic syndrome protein (WASP) family as the interpreters but, for scientists, the language of cell movement has proved difficult to learn. Two papers in The Journal of Cell Biology provide some clues as to how a lipid and a protein GTP-bound GDP-bound collaborate to activate two WASP-family CDC42 CDC42 members. The details seem protein specific but the general message is the same - activation of WASPs Active Polyproline CDC42-binding involves stopping them from biting their own tails. Actin nucleation is stimulated by the Arp2/3 complex, which is activated by WASPs. A Actin polymerization small GTPase, Cdc42, and a phospholipid, phosphatidylinositol-4,5-bisphosphate (PtdIns(4,5)P₂), activate WASPs, but how they do so is controversial: recombinant WASPs often have some constitutive activity, and inactive, GDP-bound

Henry Higgs and Tom Pollard sought to end this controversy — at least for the haematopoieticcell-specific member of the family, WASP — by purifying native WASP from bovine thymus. They find that purified WASP alone has no effect on actin polymerization rates, but that micelles containing PtdIns(4,5)P, activate polymerization through WASP. In the presence of Cdc42, PtdIns(4,5)P, micelles, or vesicles containing either PtdIns(4,5)P, or another acidic phospholipid, phosphatidylserine, produce halos of polymerized actin surrounding the phospholipid (see picture). This effect requires PtdIns(4,5)P, or phosphatidylserine, and Cdc42 must be both GTPbound and prenylated, indicating that it needs to be membrane associated to do its job.

Cdc42 sometimes seems capable of activating them.

Rohatgi and colleagues, working with a recombinant form of the widely expressed N-WASP, have a different story: they find that Cdc42, but not PtdIns(4,5)P2, can partly activate N-WASP, and that the two molecules synergize to activate N-WASP fully. They identify a basic region, close to the Cdc42-binding domain, that seems to bind PtdIns(4,5)P₂. In actin nucleation assays, a mutant N-WASP lacking this domain remains sensitive to Cdc42 but is insensitive to the additive effects of Cdc42 and PtdIns(4,5)P₂. These researchers previously showed that PtdIns(4,5)P, stimulates actin polymerization in Xenopus egg extracts. They now show that this effect depends on N-WASP but, curiously, the deletion mutant can also translate a PtdIns(4,5)P, signal into limited actin polymerization, albeit more slowly.

WASP's carboxyl terminus is constitutively active, suggesting an autoinhibitory mechanism. Both groups show that a separate Cdc42-binding domain can curb the How Cdc42 and acidic phospholipids might activate WASP. Inset shows an actin-filament halo (red) surrounding a vesicle (green). Photo courtesy of Henry Higgs and Tom Pollard, Salk Institute, La Jolla, California, USA.

activity, including the C motif and an acidic region (C and A in the figure), of the carboxyl terminus in trans. Full-length N-WASP, however, can't inhibit N-WASP's carboxyl terminus, presumably because the full-length protein is folded into its autoinhibited conformation. Higgs and Pollard find that inhibition of WASP's carboxyl terminus is relieved by GTP-Cdc42 but not by PtdIns(4,5)P2, whereas Rohatgi and co-workers find that their intermolecular inhibitory complex is regulated in exactly the same way as wild-type N-WASP.

We're still straining to understand what WASPs, Cdc42 and PtdIns(4,5)P, are saying to each other. Perhaps WASP and N-WASP respond to their two activators slightly differently, or maybe the discrepancies are due to variations between purified and recombinant proteins. Is WASP's PtdIns(4,5)P₂-binding domain equivalent to N-WASP's? And can N-WASP be activated by phosphatidylserine? Further studies should clarify how Cdc42 and acidic phospholipids unleash WASP's sting.

References and links

ORIGINAL RESEARCH PAPERS Higgs, H. N. & Pollard, T. D. Activation by Cdc42 and PIP, of Wiskott-Aldrich syndrome protein (WASP) stimulates actin nucleation by Arp2/3 complex. J. Cell Biol. 150, 1311–1320 (2000) | Rohatgi, R. et al. Mechanism of N-WASP activation by CDC42 and Phosphatidylinositol 4,5-bisphosphate. J. Cell Biol. 150, 1299-1309 (2000)

FURTHER READING Cameron, L. A. et al. Secrets of actin-based motility revealed by a bacterial pathogen, Nature Rev. Mol. Cell Biol. 1, 110-119 (2000)