Research Roundup

How transmembranes tell of their approach

Nascent transmembrane sequences (TMSs) can, while still well inside the ribosome, fold into an $\alpha$-helix, according to Cheryl Woolhead, Arthur Johnson, and colleagues (Texas A&M, College Station, TX). That $\alpha$-helix appears to send a message through the ribosome to the bound protein translocation apparatus so that it knows when to open and close its various gates.

Making a secreted protein is easy: a ribosome docks to the ER translocon, forming a tight seal, and the protein threads through the translocon tunnel. But with transmembrane proteins, the ribosome and translocon must make space for the release of both the TMS into the membrane and any following cytoplasmic loop into the cytoplasm. Previous work suggested that an ion-tight seal is maintained by the binding of BiP to the luminal surface of the translocon.

But how does BiP know it is needed? Johnson had earlier established that BiP binding is induced if synthesis is halted just four amino acids after the completion of the TMS, with the TMS still well within the ribosome. “This threw us for a loop,” says Johnson. Perhaps the nascent chain tunnel was not a passive tunnel, but a transducer of signals about TMS arrival.

Johnson now suggests that the ribosome detects a TMS as different based on the TMS’s propensity to form an $\alpha$-helix. His evidence for $\alpha$-helix formation consists of fluorescence resonance energy transfer (FRET) between dyes placed at either end of a newly synthesized TMS. Other, presumably extended, sequences showed no such energy transfer.

The TMS makes two unique ribosomal contacts. The first, with L17, coincides with and may induce BiP binding. L17 is a plausible signaler, as it has one end near the site of protein synthesis and the other near the site of contact with the translocon. Further down the tunnel, the TMS contacts L39. This coincides with the later release of the ribosome–translocon seal, which may allow cytoplasmic domains to escape.

Crystallographers have suggested a very different model in which the translocon pore forms an adjustable seal. Johnson’s model of sequential (ribosome then BiP) seals to the translocon is based on the changing accessibility to fluorescence-quenching ions. Resolution of the issue may have to await the tricky crystallization of a translocon with a bound nascent protein.

Reference: Woolhead, C.A., et al. 2004. Cell. 116:725–736.

All in the (network) family

Networks from very different systems fall into just a few superfamilies, say Ron Milo, Uri Alon, and colleagues (Weizmann Institute of Science, Rehovot, Israel). The superfamilies are based on the relative frequencies of certain local network motifs or recurring circuit elements that have defined information processing tasks.

Alon says he wants to “break the network down into elementary circuits and building blocks. We’re very much inspired by engineering. If you want to understand the device…you break the problem up.”

He has previously found that networks have higher than expected frequencies of certain circuit elements; such elements have functions such as detecting persistence or imposing temporal order. The team now quantifies the relative occurrence of such elements in a wide variety of networks and finds that just a few superfamilies emerge.

Reference: Milo, R., et al. 2004. Science. 303:1538–1542.
Surviving to remember

Not all memories live forever. Some memories may, at least in birds, die with the death of the neurons that encode them. Now, Benjamin Alvarez-Borda, Bhagwattie Haripal, and Fernando Nottebohm (Rockefeller University, New York, NY) suggest that the life expectancy of certain canary neurons, and perhaps of their associated memories, depends on one factor.

Canaries start singing and learning songs in the fall as the mating season approaches. Neurons entering the high vocal center (HVC) during this time stick around so that song memories persist. But by spring most of the birds have found a mate and ignore singing in favor of other pursuits. Neurons born in the spring come and go more rapidly.

The Rockefeller team turned the short-lived spring neurons into long-lived neurons by a short burst of brain-derived neurotrophic factor (BDNF) given 14–20 days after the neurons were born, which is when differentiation is occurring. Adding BDNF one week earlier or later did not enhance survival.

BDNF may increase survival time by strengthening neural connections. But why is this regulation needed? The memory capacity of adult bird brains may be limited by the number of available nerve cells. If cell number can’t be increased, survival time may need to be limited to make space for new memories. BDNF is the first handle on how canary brains assign lifetimes to neurons and perhaps memories. Similar processes may even apply to our own memory-processing hippocampus.

Reference: Alvarez-Borda, B., et al. 2004. Proc. Natl. Acad. Sci. USA. 10.1073/pnas.0308118101.

Actin as staismaster

Simultaneously holding onto and building something is no mean feat. Yingwu Xu, Michael Eck (Dana-Farber Cancer Institute and Harvard Medical School, Boston, MA), and colleagues find that the formin homology-2 (FH2) domain is ideally suited to the task. The apparently flexible structure of an FH2 dimer is consistent with a stair-stepping model of actin filament growth in which each half of the dimer alternately dissociates to make room for an incoming actin monomer even as the other half of the dimer maintains its grip on the actin filament.

The model did not come to Eck immediately. “We stared at the structure for about a year,” he says. But then the group tried to reconcile the perfect twofold symmetry of the FH2 crystal structure with the helical alignment of actin monomers in a filament. The only way the two would come together, they realized, was if the FH2 was flexible.

A prime suspect for the source of flexibility was a linker region of FH2. It was relatively unstructured in the crystal, could be clipped by proteases, and was the only significant tie between two parts of the dimer. Sure enough, a crystal of a mutant FH2 that was still functional had a completely different orientation of the linker and thus of the two parts of FH2.

Eck believes the two structures prove flexibility rather than demonstrating the two most important orientations of the FH2 regions. Eck hopes that eventually he will see oscillations in fluorescence resonance energy transfer (FRET) as the FH2 steps alternately close to and far away from the monomers of a growing actin filament.

Reference: Xu, Y., et al. 2004. Cell. 116:711–723.

Snaring bugs in NETs

Neutrophils spill out their DNA to ensnare and kill extracellular bacteria, according to Volker Brinkmann, Arturo Zychlinsky (Max Planck, Berlin, Germany), and colleagues.

The team figured that degradative enzymes from neutrophils, if they were to be effective, should somehow be directed to meet bacteria. When they looked at elastase localization, they discovered what they call neutrophil extracellular traps (NETs). The fibrous NETs contained degradative enzymes that could destroy bacterial virulence factors, but the overall structures break down with DNase not proteases.

NET formation does not appear to be part of an apoptotic program, and cells with NETs are still alive: they do not release cytoplasmic proteins and continue to exclude vital dyes. (Neutrophils are, however, programmed to die within hours after entering the circulation.) Whether the cells with NETs can later phagocytose bacteria remains unclear.

NETs were apparently seen before by others, but their importance was not realized. Part of their antibacterial action derives from their histones. The antibacterial action of histones was noted back in 1958 by James Hirsch. He was reportedly discouraged by others who questioned the relevance of the result, based on the idea that bacteria would never see histones anyway. Zychlinsky is now focusing on how NETs are formed, and whether they are directed at bacteria or erected as barriers in which motile bacteria will eventually be snared.

Reference: Brinkmann, V., et al. 2004. Science. 303:1532–1535.

Neutrophils trap bacteria in DNA-rich webs.