Sticky transmission

Nerve cells restrict neurotransmitter release to their synapses. That restriction, say Markus Missler (Georg-August Universität, Göttingen, Germany), Thomas Sudhof (University of Texas Southwestern, Dallas, TX), and colleagues, is provided in part by the synaptic proteins α-neurexins. These cell adhesion proteins promote calcium channel activity, and thus fusion of neurotransmitter vesicles, at the synaptic junctions.

The polymorphism and adhesion properties of neurexins led researchers to suspect a synapse-forming function. But Missler and coworkers found that mice lacking all three α-neurexins had ultrastructurally normal synapses. The mice breathed with difficulty and died on the first day after birth. In the brain stem, where breathing rhythms are generated, synapses showed reduced frequency of spontaneous transmission— a possible sign of presynaptic problems. Reduced amplitudes of synaptic responses were also evident after stimulation of neurons in the neocortex.

The reduced transmission was not further reduced by drugs blocking N-type calcium channels, suggesting that neurexins normally help the channels to function. Channels were still made and transported to the cell membrane. Thus, channels may no longer be localized to the synapse or, as Missler suggests, no longer get activated. Consistent with the latter idea, whole cell calcium currents from the cell bodies were also reduced in mutants.

According to the activation hypothesis, “in neurons, unlike in other tissues, a negative clamp may exist on the function of calcium channels, and neurexin is needed to remove this clamp,” says Missler. “It may sound a bit out of the blue. But neurons have to control the number of active calcium channels very tightly because of the negative consequences of excessive calcium influx. So neurexin may provide a localized activation of calcium channels at synapses.”

Reference: Missler, M., et al. 2003. Nature. 423:939–948.

Activation by reduction

A plant signaling protein is turned on when its intermolecular disulfide bonds are reduced and it splits into a monomeric form, say Zhonglin Mou, Weihua Fan, and Xinnian Dong (Duke University, Durham, NC). Only the monomeric form of this NPR1 protein can enter the nucleus and activate transcription.

NPR1 is made constitutively, but activated by salicylic acid (SA) as part of a general plant defense against infection. Dong initially set out to purify the NPR1 complex by gel filtration. She was puzzled that an NPR1 peak was found only with SA-treated samples, but then discovered that in the uninduced samples DTT mimicked SA: it liberated monomeric NPR1 from a complex that was too big to enter the column.

SA is produced when plants first blast away at infecting microbes with oxidants, with SA prompting various enzymes to boost oxidant production. The cells then overcompensate with antioxidants.

It is this later reducing environment that unhinges NPR1 from an oligomeric complex, say the researchers. Cysteine-substituted NPR1 mutants were constitutively monomeric and constitutively active in inducing downstream PR genes, which provide longer-term antimicrobial defenses.

The intermolecularly bonded storage form appears to be novel for signaling networks, although intramolecular disulfide formation is known to drive conformational changes that either activate transcription (by bacterial OxyR) or conceal a nuclear export signal (in yeast yAP1).

Reference: Mou, Z., et al. 2003. Cell. 113:935–944.

Doing the CDC20 shuffle

For perhaps a decade, Suc1 was the hardly little protein that featured in almost every cell cycle paper. But it was used only as a reagent—its binding to cyclin-dependent kinases (such as Cdc2 or Cdc28) yielded nearly pure MPF.

Then the budding yeast version, called Cks1, was found to be essential for growth. Now, May Morris, Steven Reed, and colleagues (The Scripps Research Institute, La Jolla, CA) have found that Cks1 helps shuffle proteins, including Cdc28, at the critical CDC20 cell cycle promoter, thus helping push cells through mitosis.

Cdc20 activates the ubiquitination machinery, which then destroys mitotic cyclins and sister chromatid glue to initiate anaphase. Morris stumbled on CDC20 because its overexpression suppressed a cks1 mutant. She then found that Cks1 helped turn on CDC20 expression during mitosis, and that Cks1 and Cdc28 both localized to the CDC20 promoter. But a version of Cdc28 that interacts poorly with Cks1 actually stuck to the promoter more avidly, suggesting that Cdc28 binds the promoter first, then brings in Cks1, and finally is kicked off via a Cks1-related mechanism.

Indeed, at the peak of both CDC20 expression and Cks1 binding to the promoter, Cdc28 was released. The release may be triggered by the proteasome, which cofractionates with Cks1 and can also bind transiently to the CDC20 promoter. Proteasome protease function is not necessary for CDC20 expression, so perhaps the proteasome is recycling a promoter-bound protein, such as Cdc28, after it has fulfilled some essential phosphorylation function, or remodeling either chromatin or a stalled transcription complex.

Reference: Morris, M.C., et al. 2003. Nature. 423:1009–1013.