In This Issue

CAST into the synapse

For the nervous system to function properly, neurotransmitters must be released in a regulated fashion from an active zone in the presynaptic membrane, but relatively little is known about how synaptic vesicles are directed to this zone in the first place. On page 577, Ohtsuka et al. characterize a novel protein associated with the active zone.

The cytomatrix at the active zone (CAZ) is believed to be important in determining synaptic vesicle localization. In the new work, the authors began with traditional biochemistry, comparing crude membrane and postsynaptic density fractions from rat brain to identify proteins associated with the synapse. One novel protein was found and named CAZ-associated structural protein (CAST). Electron microscopy places CAST at the CAZ in conventional synapses. CAST associates with two known CAZ proteins, RIM1 and Munc13–1, in a ternary complex that associates with the CAZ protein bassoon, providing the first evidence that CAZ proteins form a network of protein–protein interactions in vivo.

CAST appears to help determine RIM1 localization in neurons, and the authors found that CAST is expressed in the early stages of synapse formation in primary cultured neurons. The data suggest that complexes of CAZ proteins associate with vesicles during the early stages of synapse formation, and that these protein–vesicle complexes are then transported to the newly forming synapses. Fusion of the vesicles with the plasma membrane could then determine the location of the new active zone.

A database search uncovered a putative orthologue of CAST in C. elegans, suggesting that the new protein is a conserved component required for CAZ formation in metazoans. If so, then targeted disruption of CAST in mice or worms should provide additional insight into the formation of this crucial structure.

GETTING TRAPPED IN A ROUGH NEIGHBORHOOD

The ER is differentiated into rough ER, where membrane-bound polysomes translate proteins for insertion into or translocation across the ER membrane, and smooth ER, which lacks polysomes. But how is this differentiation established? Nikonov et al. (page 497) analyzed the lateral diffusion of translocon complexes in the ER membrane. The results suggest that association of translocons with polysomes provides the basis for ER differentiation, and that translocons remain assembled even when they are not translocating nascent polypeptides.

The authors were able to measure the diffusion of translocon complexes in the ER membrane by transfecting a temperature-sensitive mutant cell line with a GFP-tagged version of the translocon-associated oligosaccharyltransferase component Dad1. In cells that are actively carrying out translation, the diffusion constant of GFP-Dad1 is about seven times less than the diffusion constant of a freely diffusing control protein. When translation initiation is inhibited or nascent polypeptide chains are terminated, the GFP-Dad1 diffusion constant increases significantly, but remains two- to threefold less than that of the control protein.

The results suggest that as a polysome associates with multiple translocon complexes, tethering them together, diffusion of the resulting array is severely restricted. The extension of multiple polypeptide chains from this array into the viscous lumen of the ER may further restrict diffusion. The diffusion rate of GFP-Dad1 in cells where translation is inhibited suggests that the translocon complex remains assembled even when it is not associated with a polysome. These free translocon complexes are able to diffuse in the ER membrane considerably faster than complexes associated with polysomes.

According to the authors’ model, polysome arrays, once formed, may remain relatively immobile and define the location of the rough ER. Translocon complexes released from these arrays after translation termination could diffuse relatively freely through the ER before reassociating with the polysomes to initiate a new round of translation.