Spotlight on the active zone

Nava Segev* and Simon Alford

Department of Biological Sciences; University of Illinois at Chicago; Chicago, IL USA

A chemical synapse is a junction between a pre-synaptic neuron and a post-synaptic cell, neuron or non-neuron, through which signals are transmitted by neurotransmitters.

The active zone is the site in pre-synaptic neurons through which neurotransmitters are secreted to the synaptic cleft, the gap between the pre- and post-synaptic cells. The release of neurotransmitters from synaptic vesicles in the active zone requires a rapid and large calcium pulse to initiate release, which is extremely fast, but is also sensitive to low and re-occurring transient increases in calcium concentration. Active zones are defined morphologically as electron-dense regions and multiple proteins are known that act at this area to contribute to the release of neurotransmitters. One of these components is the Rab3 GTPase, which is found on synaptic vesicles and recruits Rab3-interacting molecules, termed RIMs.

In this issue, Kaeser summarizes three exciting papers from the Sudof lab, which together dissect the role of the multi-domain RIM proteins in the organization of active zones in synapses.¹⁻³ It was known that RIMs interact with synaptic vesicles via Rab3 GTPase and tether these vesicles to calcium-channels in active zones. However, the exact mechanism that underlies synaptic vesicle tethering, docking, priming and fusion remains subject to intense debate. I asked Pascal to explain the new findings regarding the “RIMs of synaptic vesicles” to “non-synaptic” readers.⁴ And here is my take on this:

There are many RIMs; however, the first major result is comforting because, in spite of this multiplicity of the RIMs, one RIM variant can restore the function of all RIM variants. This simplified the quest and allowed the analysis of individual RIM domains. This analysis yielded an extremely convenient result, namely, two RIM functions can be uncoupled by splitting RIM to two fragments: the N- and C-terminal halves: The C-terminal half interacts with calcium-channels to tether them to active zones. The N-terminal half interacts with Rab3 on synaptic vesicles and with Munc13, and activates Munc13 to allow SNARE-dependent vesicle fusion of the synaptic vesicle with the cell membrane. The third interesting result is that RIM affects synaptic vesicle fusion by interacting with Munc13 and activating it by causing its monomerization. Monomeric Munc13 may then interact with the SNARE complex, which is necessary for vesicle fusion to occur. Together, these findings show that by interacting with multiple active-zone players, RIMs coordinate

synaptic vesicle priming and fusion, allowing the amazing speed and accuracy by which this process occurs.

In our opinion, the most unexpected finding in these studies is that Ca^{2+} channels are tethered, or localized, to the active zone by their interaction with RIM. This suggests that the calcium channels can be oriented accurately towards the calcium-sensitive components on the vesicle, such as synaptotagmin. Such a mechanism would ensure fast and accurate release of neurotransmitters from the docked vesicle with a minimal membrane calcium flux. For cellular mechanics this is a very satisfactory explanation.

However, from the cellular logistical point of view, this finding raises a new basic questions. Synaptic vesicles are free to move in the cytoplasm. If they are not tethered by RIM to synaptic vesicles, it seems that Ca channels are also free to move in the membrane. If both vesicles and Ca channels are free to move, what anchors them to the active zone? Which membrane protein in the active zone keeps all these otherwise floating elements in place?

It is important to consider that the constituents of the active zone, particularly including Rab3A are dynamic with respect to the proteins with which they interact. RIM is an attractive candidate to tether and enable docking. Other proteins, however, play a role in tethering vesicles distal to the active zone, such as the Synapsins, which interact with each other as well as SH3 and BAR domain containing scaffolds and Dynamin 1.⁵ Perhaps the structure and identity of the active zone is maintained dynamically. Vesicles are tethered first by Synapsins 1 and 2, but then prior to and during docking Rab3A may coordinate RIM as a short-term anchor while ternary SNAREs form at the primed vesicle aided by Munc13. In this way, we may envisage a dynamic recruitment of vesicles, v-SNAREs and synaptotagmin to the active zone membrane components of fusion—tSNAREs, Ca^{2+} channels and complexins. The identity of the site of fusion exists fleetingly, but as a functional structure. This view might contrast to a more static view of anchoring that one is tempted to conclude from ultrastructural images which are themselves mere snapshots in time (Fig. 1).

But this is, of course, conjecture. If anyone knows what might be an active-zone anchor or wants to speculate on it, or if you want to comment on other aspects of RIM and the active zone, please, use the CellLogBlog:

www.landesbioscience.com/journals/cellularlogistics/blog

*Correspondence to: Nava Segev; Email: nava@uic.edu
Submitted: 08/02/11; Accepted: 08/02/11
DOI: 10.4161/cl.1.3.17634

Figure 1. RIM-dependent transition of vesicles from Synapsin tethered in the releasable pool, to primed ternary SNARE containing vesicles attached to the active zone membrane. RIM coordinates both the action of Munc13 and the tethering of calcium channels, to the active zone. RIM interacts with synaptic vesicles (SV) through Rab3 GTPase. Interaction between RIM and calcium channels tethers the channels to the active zone (discussed in ref. 4). This provides orientation of the calcium channel towards calcium sensors on SV and may target the vesicles and their attendant fusion apparatus to the active zone (dark gray membrane region). In the final step of priming ternary SNAREs are formed, which upon calcium flux and its binding to synaptotagmin, enable a sequence of events that leads to the fusion of SV with the active-zone membrane. This model stresses a constant dynamic interaction, but perhaps there exists specific tether of SVs and Ca^{2+} channels to the active zone.

References

1. Deng L, Kaeser PS, Xu W, Sudhof TC. RIM proteins activate vesicle priming by reversing autoinhibitory homodimerization of Munc13. *Neuron* 2011; 69:317-31.
2. Han Y, Kaeser PS, Sudhof TC, Schneggenburger R. RIM determines Ca^{2+} channel density and vesicle docking at the presynaptic active zone. *Neuron* 2011; 69:304-16.
3. Kaeser PS, Deng L, Wang Y, Dulubova I, Liu X, Rizo J, et al. RIM proteins tether Ca^{2+} channels to presynaptic active zones via a direct PDZ-domain interaction. *Cell* 2011; 144:282-95.
4. Kaeser PS. Pushing synaptic vesicles over the RIM. *Cell* 2011; In press.
5. Shupliakov O, Haucke V, Pechstein A. How synapsin I may cluster synaptic vesicles. *Semin Cell Dev Biol*, 2011; In press.