Crystal Structures of the Synaptotagmin-SNARE Complex that is Essential for Synchronous Synaptic Neurotransmitter Release

Scientists at Stanford University School of Medicine have determined the 3-D structure of a complex of synaptic proteins that controls the release of signaling chemicals (called neurotransmitters, such as glutamate, dopamine or serotonin) from brain cells in less than one-thousandth of a second, which ultimately could help unlock a new realm of drug research targeting brain disorders [1]. They used the Linac Coherent Light Source (LCLS) X-ray Free Electron Laser (XFEL) at in collaboration with the Structural Molecular Biology (SMB) program at the Stanford Synchrotron Radiation Lightsource (SSRL).

This work suggests that the synaptic complex (Figure 1) is important for calcium-triggered neurotransmitter release. One part of the complex (the "SNAREs") provides the energy for the neurotransmitter release and the other ("synaptotagmin-1") is acting as the calcium sensor. It has been known that both parts essential, but until now it was unclear how those two pieces fit and work together. The two protein components central to the study had been extensively studied before, although mysteries remained about how they bind and work together. Earlier x-ray studies, including experiments at SSRL nearly two decades ago, determined the structure of the SNARE complex, a helical protein bundle found in yeasts and mammals. On the other hand, synaptotagmin-1 plays an important role as a calcium sensor and a calcium-dependent trigger for neurotransmitter release. The new structure has identified unanticipated interfaces between synaptotagmin-1 and the neuronal SNARE complex that reveal, among others, a large calcium-independent and specific binding interface.

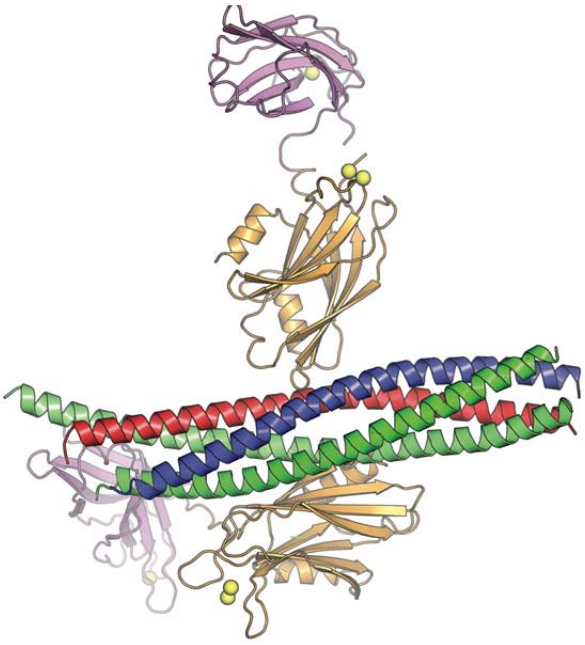

Figure 1. Crystal structure of the complex between synaptotagmin-1 (orange and magenta) and the neuronal SNARE complex (green, red, and blue). Yellow spheres indicate bound calcium. Shown is one of the two complexes in the asymmetric unit of the crystal form that produced a nearly complete data set at LCLS.

The new crystal structures, along with functional studies, suggest that the SNAREs and synaptotagmin-1 join up, they act as an amplifier for a small increase in calcium concentration, triggering a gunshot-like release of neurotransmitters from one neuron to another. The work also for the first time suggests that the proteins join together before they arrive at a neuron's membrane, which helps to explain how they trigger brain signaling so rapidly. The researchers speculate that several of the joined protein complexes may group together and simultaneously interact with the same synaptic vesicle, triggering neurotransmitter release efficiently. Upon calcium binding, the plasma membrane is deformed, triggering fusion.

To collect diffraction data for this synaptic protein complex, the researchers used a robotic system and a goniometer-based system, both developed at SSRL to study radiation-sensitive macromolecular complexes, at the XPP endstation of LCLS [2]. The researchers combined and analyzed hundreds of x-ray images from about 150 protein crystals to reveal the atomic-scale details of the joined structure. A new experimental station (MFX) under development at LCLS is intended to increase the access for these types of biologically relevant experiments.

Primary Citation

Q. Zhou, Y. Lai, T. Bacaj, M. Zhao, A. Y. Lyubimov, M. Uervirojnangkoorn, O. B. Zeldin, A. S. Brewster, N. K. Sauter, A. E. Cohen, S. Michael. Soltis, R. Alonso-Mori, M. Chollet, H**.** T. Lemke, R. A. Pfuetzner, U. B. Choi, W. I. Weis, J. Diao, T. C. Sudhof and A. T. Brunger, "Architecture of the Synaptotagmin - Snare Machinery for Neuronal Exocytosis", *Nature* **525**, 62 (2015). DOI: 10.1038/nature14975.

References

1. Q. Zhou, *et al*., *Nature* **525,** 62 (2015).

2. A. E. Cohen, *et al., Proc. Natl. Acad. Sci.* **111**, 17122 (2014).

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