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Structure of Synaptic Connectors Solved

Researchers have a new picture of how neurons forge physical connections between one another in order to communicate. Establishing these connections is critical for proper brain function, and errors in the process are thought to be associated with autism and other disorders. The new understanding of the structure of the proteins that form this connection and mutations that can disrupt their interaction may help explain some forms of these disorders.

Howard Hughes Medical Institute investigators Axel Brunger and Thomas Südhof reported the structure of the complex of the proteins, called neuroligin-1 and neurexin-1 beta, in an article in the December 20, 2007, issue of the journal *Neuron*. Brunger is at Stanford University and Südhof is at The University of Texas Southwestern Medical Center.

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— Axel T. Brunger

Neurons communicate with each other at specialized intercellular junctions called synapses. A presynaptic neuron releases a chemical neurotransmitter into the synapse that is recognized by the postsynaptic neuron, triggering a nerve impulse. In the nineties, Südhof's lab identified two families of proteins produced by the neurons on either side of the synapse. These proteins extend outside of the cells where they are produced and contact one another to form a physical link across the synapse. In the presynaptic neuron, this connector protein is called neurexin; its partner on the postsynaptic neuron is known as neuroligin. Past studies had indicated that neuroligins and neurexins establish their connection by forming a complex in which two neuroligin molecules link to each other, and a neurexin molecule attaches to each side of the pair.

As neurons create new synapses during learning, they must form this neuroligin-neurexin connection for those synapses to become functionally mature. If a newly formed synapse does not mature, it will be eliminated.

Problems with this process of synapse development are thought to be a major cause of brain disorders such as autism and mental retardation. The evidence that these interactions are important is overwhelming, but exactly what neuroligin and neurexin do is very unclear, said Südhof.

To gain a better understanding of how the two proteins interact, Brunger and his colleagues investigated their molecular structure using x-ray crystallography, a technique in which scientists direct x-rays through protein crystals then deduce their structure by analyzing how the beams have diffracted. They crystallized and analyzed neuroligin-1 by itself, as well as the complex of neuroligin-1 and neurexin-1 beta.

According to Brunger, the structure of neuroligin-1 hinted that it might interact with partner molecules other than neurexin. The researchers identified a structural feature on neuroligin-1 that might act as an additional binding site, and the two laboratories are searching for molecules that might interact there.

The researchers learned even more from analyzing the structure of the neuroligin-1/neurexin-1 beta complex, said Brunger. That structure allowed them to visualize the interfaces between the two neuroligin and two neurexin molecules in the four-molecule complex, as well as binding sites for calcium, which is required for complex formation.

To determine whether the structure they had developed reflected the actual behavior of the proteins, the researchers mutated neuroligin-1 at locations they predicted would disrupt the interface between it and neurexin. These mutations greatly inhibited the complex from forming.

Not only did those mutations show that we could disrupt the interface without disrupting the folding of the neuroligin molecule, but they have given us a molecular tool to probe the details of these interactions, said Brunger.

Südhof and his colleagues are now investigating how these neuroligin-1 mutations alter synaptic function, both in cultures of neurons and in mice. With these mutations, we can now begin to explore how these central proteins in the synapse actually work, Südhof said. We can also begin to ask whether neuroligins might have other interactions beside with neurexin that influence synaptic function, he said.

Brunger and Südhof said the new structural information and an understanding of how mutations affect that structure could offer clues into how the complex might be disrupted in individuals with autism. While the causes of autism are unknown, HHMI investigator Huda Zoghbi, who is at Baylor College of Medicine, has proposed that subtle, inherited alterations in synaptic function might underlie some forms of the disorder. Since she advanced her theory in 2003, other studies have implicated mutations in neuroligins and neurexins as being associated with autism spectrum disorders.

The researchers say that further work is needed to establish whether these types of mutations disrupt brain function by interfering with neuroligin's interaction with other proteins. If this idea is validated, Brunger said, it is not inconceivable that pharmaceutical approaches to treating autism could be developed to stabilize such interactions and compensate for the effect of the mutations.