Characterization of bendless and interacting partners in Drosophila central synapse formation

Smitha Babu Uthaman, *University of Massachusetts Amherst*

Abstract

Synapses are the functional units of neuronal circuits and the sites of integration for multiple signaling pathways. Understanding the molecular basis of synaptic function is critical to understanding the bigger picture of how we think, learn, remember and how neurological diseases and disorders disrupt these faculties. Here we characterize the role of *bendless* (*ben*) in central synapse formation by utilizing the giant fiber system (GFS), a well established neuronal network in *Drosophila melanogaster.* Ben is an E2 conjugase and a key member of the enzyme cascade involved in ubiquitin dependent regulation. Ben was originally identified more than two decades ago and was believed to be involved in axon guidance as mutants lacked a synaptic connection between the giant fiber (GF) and its target, the jump motor neuron (TTMn). We have been able to redefine Ben function by demonstrating that an incipient GF-TTMn synaptic connection is present in *ben* mutants. We have also analyzed the synapse with the help of synaptic markers as well as studied its features at an ultrastructural level. By conducting cell autonomous rescue experiments we have spatially determined that *ben* has a presynaptic function in the GFS. We then used the TARGET system to temporally characterize the gene and have isolated a critical period for Ben function during development. We further assayed protein localization by generating GFP-tagged Ben constructs and have found the protein to be nuclear as well as cytoplasmic. Subsequent studies have identified two multifunctional proteins—Semaphorin1a and Distracted—to be putative targets of Ben action. We have also carried out a preliminary characterization of the synaptic roles other components of the ubiquitin system have in the GFS, such as the ubiquitin ligase *highwire* and the deubiquitinating proteases, *fat facets* and UBP2. In summary, we have found the ubiquitin conjugase Ben to have a novel and distinct role as a developmental switch in the establishment of a central synapse. The identification of likely downstream targets of Ben and comparison with related ubiquitin associated proteins suggest that a delicate regulatory balance has to be maintained in order for a synapse that is functionally and morphologically normal to be sculpted.^

Subject Area

Molecular biology|Neurosciences|Cellular biology

Recommended Citation