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Drosophila Dscam - function in dendrites and localization

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The *Drosophila melanogaster* gene Dscam is alternately spliced to generate over 38,000 different isoforms that interact in *trans* in a homophilic, isoform-specific manner. Since its discovery first published in 2000, Dscam has been shown to be essential for proper axonal circuitry in a number of different systems. Preliminary studies have also suggested a role for Dscam in dendrite morphogenesis as well as synaptogenesis. However, there have been no reported studies examining the mechanism of Dscam's function in dendrites.

The purpose of this work was to investigate the role of Dscam in dendrite morphogenesis. Based on the hypothesis that Dscam mediates repulsive signaling between dendritic sister branches resulting in self-avoidance, a series of genetic manipulations in dendritic arborization neurons were performed. Dscam loss-of-function led to abnormal dendrite morphogenesis with defects such as self-crossing between sister branches. Dscam overexpression, on the other hand, caused phenotypes that indicated increased and abnormal repulsion, both between sister dendrites and between dendrites of cells expressing the same isoform. Transgenic fly lines containing FLAG-tagged Dscam isoforms confirmed Dscam localization to dendrites with differential localization levels dependent on the transmembrane domain. Overall, the results lead to the model that dendrite self-avoidance is controlled by Dscam through homophilic interactions triggering repulsion. Therefore, Dscam diversity allows for recognition of self versus non-self.

The mechanism through which Dscam mediates repulsion remains to be elucidated. The experiments examining Dscam downstream signaling showed that it likely bypasses the kinase Pak in dendrites as opposed to axons.

With established roles for Dscam both in axons and dendrites, as well as Dscam's implication in synaptic partner matching in a previous study, a second series of experiments was performed to examine whether Dscam might be found at synapses in the *Drosophila* visual system. Data obtained by immunohistochemistry in larval stages as well as in pupal stages, however, showed Dscam to be close to but not directly at synaptic markers. Higher resolution experiments seem to be required for conclusive answers on ultrastructural details. For these future endeavours, a number of monoclonal antibodies were succesfully raised against variable extracellular domains of Dscam.

Many questions about the temporal and spatial course of isoform expression and its significance remain. These issues could be tackled by methods of selectively labeling individual endogenously expressed isoforms. The new antibodies were tested for their binding specificity with regard to the variable extracellular domains of Dscam but were not found to be isoform-specific.

Research on Dscam continues to unravel mechanisms through which molecular diversity plays a role in establishing neuronal connectivity and specificity. The work presented here has contributed in the understanding of neurite morphogenesis in vivo and the quest for understanding more general physiological and pathological developmental processes in the brain.