Connectome reconstructions in the Drosophila brain

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The serial-section electron microscopy (Serial-EM) technique is an established method for reconstructing a map of the entire synaptic connections, or connectome. However, the technique has a resolution in z-axis that is limited by section thickness, and thus poorer than x-y resolution, causing many ambiguities in tracing neurites. A new imaging technique with better z resolution should therefore make reconstruction easier and its product more accurate. Focused-ion beam milling scanning electron microscopy (FIBSEM) offers improved z resolution and is the only technique enabling the collection of isotropic image datasets at such higher resolution. Here, using FIBSEM technique, we reconstructed connectomes in two brain regions in *Drosophila*, the second optic neuropil, or medulla, and the mushroom body alpha lobe. We find that direction-selective T4 cells receive synaptic inputs from the following neurons: columnar Mi1, Mi4 and Mi9, a centrifugal C3, a wide-field CT1, and two types of relay neurons Tm3 and TmY15. Significantly, the placement of these inputs differs: Mi9 clusters at the tips of T4 dendrites while Mi1 or Tm3 distribute along the shafts, and input from Mi4, C3 and CT1 localizes to their bases. Combined with the diversity of neurotransmitters employed by the inputs (excitatory onto T4 dendrite shafts and inhibitory (GABA) onto the dendrite bases), our findings strongly suggest that motion detection in *Drosophila* utilizes two types of motion detector models proposed by Hassenstein and Reichardt (H-R model, 1956) and Barlow and Levick (B-L model, 1965). Our connectome results also provide reliable foundations for analyzing and understanding neural circuit functions of the olfactory system in *Drosophila*.

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