Three-dimensional distribution patterns of newborn neurons in the adult olfactory bulb

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Abstract:

We present a new method to study the three-dimensional (3D) spatial distribution patterns of newborn neurons in the mouse olfactory bulb (OB). Newborn neurons were transduced, in vivo, using lentiviruses to express green fluorescent protein (GFP). Two-photon (2P) microscopy was used to image thick (OB) slices (approximately 250 microm) at single cell resolution. Image-stacks were captured semi-automatically, and concatenated offline, to create larger image-stacks containing the positional information of all the labeled neurons. Serial reconstruction of the large image-stacks resulted in a three-dimensional virtual model, containing the exact position of all the labeled newborn neurons within large volumes of the OB. The feasibility of this method was demonstrated by analyzing the cell distributions of thousands of GFP labeled newborn neurons. This analysis identified 3D clusters in which the newborn cells' density is significantly higher than the mean density. We show that our method reveals information that is overlooked when sampling only a small fraction of the tissue in 2D. This method may serve as a valuable tool, not only for analyzing newborn neurons in the OB, but also for other neuronal types as well as for other brain regions.

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