The nervous systems of animals consist of interconnected neurons, forming a network structure. Such network structures can be naturally modeled with graph theory, which defines a graph with nodes and connecting edges. Graph theory provides tools to quantitatively measure the organization and the efficiency of the network, and to identify functional units and the structures that are crucial in regulating the network state1. Therefore, graph theory offers powerful ways to study both structure and function of the neural networks.

In the graphical models for structural analysis of the brain, graphs are constructed with nodes representing the functional units of the brain such as neurons or brain regions2, and edges representing their connections. Such models have been used to reveal the organizing principles of neural networks, and to identify critical regions or neurons in the networks3–6.

With the advances in large-scale recording techniques such as functional MRI (fMRI), electroencephalography (EEG), multielectrode array (MEA) and calcium imaging, there has been an increasing interest in studying the functional connectivity of the neural network in the brain. In recent years, many studies have applied graph theory in analyzing data from fMRI, EEG and MEA, taking brain regions7–9, voxels10–12 or electrode position13 as nodes, and the functional correlations as edges. Most of these studies constructed either directed or undirected graphs based on the activity association measurements such as cross-correlation, mutual information and Granger causality, and further measured and compared the network properties in resting state and cognitive processes as well as during development and pathological conditions1,7,14–16. However, few studies have applied graph theory with single cell resolution, where neurons can be taken as nodes, and edges as their inferred relationship. One of the first efforts generated graphs using microelectrode array recording from cultured neurons17, whereas several other studies analyzed calcium imaging data with graph models, either in slices or in vivo18–20. While most of these studies relied on the cross-correlation of neural activity to estimate edges, there is still a lack of systematic method to construct graphs from multi-neuronal data.

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