Specific Aims

Background and rationale: Identifying neuronal cell types (or classes) is a logical first step toward a connectome-based investigation of brain function and dysfunction. A strong existing body of work exists in this area, but a brain-wide survey of neuronal cell type distribution based on high-resolution morphology data correlated to molecular properties is not available. Furthermore, biophysical properties of the cell such as elasticity that serve as important indicators for health/dysfunction have not been included in the classification scheme. This project will address these gaps in neuronal cell type classification.

Goal: The overall goal of this project is to conduct a global survey of neuronal cell types in the mouse brain, based on morphological, electrical, biophysical, molecular, and computational properties of neurons.

Aim 1: Morphological survey of neuronal cell types across major cortical and subcortical nuclei in the mouse brain. Whole mouse brain data sets from the Knife-Edge Scanning Microscope, a serial sectioning light microscope that achieves a resolution of 1 um^3, will be the main technical resource of this project. Golgi and fluorescence probes (also see aim 3) will be used to label subsets of neurons across the whole brain, and automated algorithms will be developed for 3D reconstruction of neuronal morphology. Morphometric properties will be assessed and their localization and distributions across the brain will be computed. Based on this, initial morphological cell types of neurons will be compiled.

Aim 2: Measurement of biophysical properties of different morphological cell types of neurons. High throughput microfluidics technologies will be developed to measure the biophysical properties such as elasticity at single-cell resolution with ultra-high throughput. When combined with microdissection, this will also provide information on localized distribution of the cell types. Cell classification data based on their biophysical properties will then be compared to the morphological (Aim 1) and molecular properties (Aim 3) of neurons.

Aim 3: Molecular characterization of different morphological cell types. Immunohistochemistry techniques will be used to validate the morphological cell types determined in Aim 1. Transgenic mice will be used to contrast broad cell types (such as excitatory vs. inhibitory neurons) and correlate them with morphological characteristics. Potential differences in molecular characteristics of the same neuronal cell types dependent on their localization in the brain will be investigated.

Aim 4: Computational characterization of different morphological cell types. Multicompartmental simulations will be conducted based on the reconstructed geometry of neurons and their known ion channel types (from the literature), electrical and biophysical properties (from Aim 2), and molecular characteristics (from Aim 3) to study variations in electrophysiological behavior due to variations in morphology.

Aim 5: Neuroinformatics platform for the exploration and analysis of data from Aims 1 through 4. A web-based mouse brain atlas enriched with data and statistics from Aims 1 through 4 will be developed to accelerate dissemination and discovery. Facilities to link the data to external resources such as the Brain Architecture Knowledge Management System (BAMS) and the Allen Brain Atlas (ABA) will be included, so that our detailed neuronal cell-type distribution information can be aligned with the existing knowledge base (BAMS) and massive gene- and connectivity data (ABA).

Expected outcomes and impact: *(1) From structure to function:* Once correlations between morphology and other functional properties (e.g., electrical, molecular, and biophysical properties) are known and sorted into different cell types, functional properties can be inferred from morphology-based classification. The proposed morphology-based classification of neurons and multi-modal validation of those neuronal cell types are expected to span the current knowledge gap between structure and function. *(2) Distribution of neuronal cell types across the mouse brain:* Information regarding the localization and distribution of such neuronal cell types across the whole brain will be mapped, providing a unique and invaluable resource for neuroscience research. Also, potential differences within the same neuronal cell types due to localization could be identified. The outcome of this project will serve as a logical first step in connectome-based investigation of brain structure and function.