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## IMAGE ANNOTATION FOR ELECTRON TOMOGRAPHIC DATA

Presenting Author: MARYANN E. MARTONE, Associate Adjunct Professor

L. Fong<sup>1</sup>, L. Chen<sup>2</sup>, R. West<sup>1</sup>, M. Wong<sup>1</sup>, S. Lamont<sup>1</sup>, A. Gupta<sup>2</sup>, M. E. Martone<sup>3</sup> <sup>1</sup>Dept Neuroscience, UCSD, San Diego, CA, <sup>2</sup>San Diego Supercomputer Center, UCSD, San Diego, CA, <sup>3</sup>Dept Neuroscience (M008), UCSD, San Diego, CA.

Segmentation tools for electron tomographic data have been available to researchers for a number of years, providing a feature for users to extract the structures of interest for their particular study. As an increasing number of objects are segmented, it becomes difficult to manage the data; all of the structures essentially become disjoint from one another. There is no way to record whether a segmented structure is located next to another structure, or whether a structure is part of a larger structure. In addition, there is no way to compare structures annotated by different researchers. To address this challenge, we have developed Ovoxtrace, a segmentation tool that allows microscopic image data to be annotated in a highly structured manner. The prototype for Ovoxtrace was developed as an extension of Xvoxtrace, a widely used tool developed at the National Center for Microscopy and Imaging Research (NCMIR) for segmenting EM tomograms. A web based version, Jinx, is currently under development. During segmentation in Ovoxtrace, as objects of interest are identified in each slice throughout a tomographic volume, rather than supplying their own object name identifiers, users select entities that have already been defined in the Ontology for the Subcellular Anatomy of the Nervous System (Fong et al., submitted). An ontology consists of a set of concepts linked by relationships such as 'is a' and 'has part,' e.g., 'neuron is a cell; cell has part plasma membrane.' It is an important means by which knowledge in a field is formalized for classifying and describing scientific observations. The Subcellular Anatomy Ontology (SAO) is an ontology for describing the subcellular anatomy of the nervous system. It includes neurons, glia, multicellular microdomains and their associated functional compartments, cellular components, and molecular constituents. Cell components are drawn from the Gene Ontology (<http://www.geneontology.org/>). The SAO includes general characteristics of neuronal and glial morphology and illustrates specialized views of the properties of individual neuronal classes, e.g., Purkinje cells and Pyramidal cells. During the segmentation process, users create instances of the structures contained in the tomographic volumes. A given instance can either be independent or defined as part of another entity, e.g., mitochondria\_0000 is part of dendrite\_0001, and additional relationships such as 'associated with' and 'synapses with' are also supported. . The system provides user feedback by showing users a complete description of their object relative to other existing elements within the SAO. We have also incorporated a feature to enable users to add new terms not currently present in the ontology during the segmentation process for immediate use in annotating their data. The output of Ovoxtrace includes a list of entities and their parts, along with other relationships. As objects are annotated and segmented based on the concepts and relationships in the SAO, they are stored as instances in OntoQuest, a knowledge base designed to capture structural and molecular properties of nerve cells, their parts, and supracellular domains (Chen et al., 2006). In addition to storing all of the annotated information, OntoQuest generates statistical and aggregate properties of cells, assembling information of associating compartments, components, and morphometrics from these instances. Many of the datasets contained in the CCDB have been annotated with the SAO and stored as instances in OntoQuest (Martone et al., 2003). Because the SAO is publicly available and is cross referenced to other public ontologies, data annotated with the SAO can be compared and aggregated with data accruing from other sites and at other scales. The advantage of Ovoxtrace is that annotation occurs concurrently with segmentation rather than after the fact. The image annotation function of Ovoxtrace thus becomes an important piece in streamlining the development and validation of novel hypotheses of neuroscience data based on these aggregate properties. We are implementing the ontology extensions with automatic segmentation routines to be offered through the web-based version, Jinx. In addition to the standard manual tracing tools available in Xvoxtrace, Jinx will implement fuzzy segmentation (Garduno and Wong, 2006) and level set segmentation (Sethian 1996, Whitaker et al) methods. In addition to the Nuages (Geiger, 1995) surface tiling method, we will include the Power Crust method (Amenta, Choi, Kolluri, 2001) method.