Identifying Synapses using Deep and Wide Multiscale Recursive Networks

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Abstract
In this work, we propose a learning framework for identifying synapses using a deep and wide multi-scale recursive (DAWMR) network, previously considered in image segmentation applications. We apply this approach on electron microscopy data from invertebrate fly brain tissue. By learning features directly from the data, we are able to achieve considerable improvements over existing techniques that rely on a small set of hand-designed features. We show that this system can reduce the amount of manual annotation required, in both acquisition of training data as well as verification of inferred detections.

1 Introduction
The emerging field of connectomics involves determining the connectivity between neurons in a brain. Recent advances in electron microscopic (EM) imaging offer unprecedented access to the neuronal intricacies of a brain. However, the process of extracting consumable information (i.e., reconstruction) from a series of highly-detailed EM images is time-consuming, typically requiring comprehensive manual annotation. While efforts to automatically extract neurons using machine image segmentation has simplified annotation, other parts of the process, like identifying physical connections between neurons via chemical synapses, has only recently been considered by the machine-learning community.

Discovering the connectome of an organism is a time-intensive process and has only been done to completion in C. elegans [16]. To derive the connectome for larger organisms, significant advances must be made in automatic image analysis. In particular, Plaza et al. [11] note that with advances to image segmentation, synapse identification is becoming a significant bottleneck. Synapse identification is essential to accurately determine whether two neurons are connected. Relatively small brains, such as in Drosophila, contain tens of millions of synapses. If each synapse could be identified and annotated in approximately one minute, this would take a single annotator a staggering 5,000 work days to complete.

To help mitigate the bottlenecks of synapse annotations, prior work has defined a methodology whereby pre-synaptic sites are automatically predicted using a classifier [12]. Due to the inaccuracy of the predictor, subsequent manual verification was needed. To truly scale synapse annotation to larger volumes, better synapse prediction is required.

In this paper, we introduce a system for automatic synapse detection using deep learning. Our approach is able to achieve a large improvement in accuracy compared to existing methods [8, 12], while also requiring much less training data. Therefore, the immediate benefit is a dramatic reduction in the amount of manual effort required, both in obtaining training data and verifying putative detections. However, we also note that the quality of our system makes a significant step toward achieving human-level performance, and makes it possible to consider fully-automated annotation.
without any manual verification. We provide extensive validation against a large image dataset in the Drosophila [12].

We next give some background by discussing related literature in automatic synapse detection and deep learning. In Section 3, we describe our system for detection using DAWMR networks, and in Section 4, we compare the results of our method against existing work. We conclude with a discussion on the potential for fully-automated detection.

2 Background

In this section, we review related work in automated synapse detection and deep learning.

2.1 Automated Synapse Detection

Most of the existing work on automated synapse detection has used the ilastik toolkit [13], which allows for interactive training of Random Forest (RF) classifiers using hand-designed features. ilastik has been applied to detecting synapses in vertebrate (mammalian) tissue, both with isotropic resolution [8] and anisotropic resolution [7]. Our work focuses on detecting specific pre-synaptic structures in invertebrate (Drosophila) tissue. This problem has also been addressed using ilastik [12], and we compare with this existing work in Section 4.

Becker et al. [1] take a slightly different approach to detecting synapses, by estimating the location and orientation of the synaptic cleft, and then computing (hand-designed) features at specific locations relative to the synaptic cleft. These features are used as input to AdaBoost for classification. It is uncertain how well this method would translate to synapse detection in Drosophila, due to the dependency on being able to consistently and reliably determine the synaptic cleft, and the polyadic nature of many synapses in Drosophila (having multiple post-synaptic partners per pre-synaptic site).

2.2 Deep Learning

The term deep learning refers to a class of algorithms in machine learning whose models contain multiple non-linear transformations, in distinction to traditional “shallow” algorithms such as support vector machines. A notable example of a deep learning algorithm is the deep belief network [4]. Often, such algorithms can be trained directly on unlabeled data, to perform unsupervised feature learning, such as the convolutional deep belief network [10]. Deep learning algorithms have recently been applied to many tasks in computer vision and achieved state-of-the-art results, for instance, in object recognition [9] and face verification [14].

Deep learning techniques have also been successfully applied to problems of segmentation and boundary prediction in EM images [6, 3]. In this paper, we make use of Deep and Wide Multiscale Recursive (DAWMR) networks, introduced by Huang and Jain [5] for EM boundary prediction, and additionally applied to 3d agglomeration of neural fragments [2]. DAWMR networks consist of a fast unsupervised feature learning component, extracting features at multiple scales and pooling features spatially for larger context, followed by supervised classification with multilayer perceptron. These networks were demonstrated to achieve higher accuracy than a supervised feedforward multilayer convolutional network, as well as requiring less time to train.

Our work differs from previous applications of deep learning to problems in EM reconstruction, as the problem of synapse detection is at the object-level, requiring an aggregation of evidence over multiple voxels, rather than a binary decision applied to every voxel or every pair of neighboring fragments. We next discuss how we adapt DAWMR networks to the general problem of object detection.

3 Methodology

DAWMR networks were designed for the problem of image labeling, in which a label (e.g., boundary/not-boundary) is associated with every pixel of an image or voxel of an image volume. In contrast, the problem of synapse detection is one of object detection: given an image volume, one is to identify every synapse in the volume, such as by placing a bounding box around the synapse or identifying every voxel belonging to the synapse. For the data used in this paper, described below
in Section 4.1, the synaptic structures to be detected are all of approximately the same size, and therefore we identify each structure with a single $x, y, z$ coordinate defining its center, and implicitly a fixed radius defining its extent.

In order to use DAWMR networks for automated synapse detection, then, there are two main issues that must be addressed: how to use object-level ground-truth to train a voxel-wise classifier, and how to produce object-level predictions from voxel-wise output.

### 3.1 Synapse Voxel-wise Training

At training time, we are given an image volume and a set of coordinates $\{s_1, \ldots, s_N\}$ indicating the center of every synapse within the volume. We use a simple strategy for forming voxel-wise training data given this object-level supervision: a voxel at spatial position $i$ has an associated label $l_i = 1$ if its distance to any synapse at position $s_j$ is less than a predefined threshold $r_l$, e.g. $\exists j : d(i, s_j) \leq r_l$, and $l_i = 0$ otherwise.

Given the typical density of synapses within a volume, and reasonable values for $r_l$, this training strategy leads to a large class imbalance between positive and negative examples. DAWMR networks are typically trained using balanced sampling of class examples, which in this case amounts to using all of the examples with a positive label, and subsampling from the examples with a negative example.

### 3.2 Object Prediction

After training a DAWMR network or another voxel-wise classifier, we can perform inference on a new image volume, producing a real-valued prediction $p_i$ at each spatial position $i$. For automated object detection, we next need to transform these voxel-level predictions to a set of coordinates indicating the set of predicted synapse centers.

To do so, we propose a straightforward method of averaging followed by simple non-maximum suppression (NMS). We first convolve the set of predictions $\{p_i\}$ with an averaging filter of radius $r_a$ to produce averaged predictions $\{a_i\}$. We then perform an iterative procedure where we select $j = \arg \max_i a_i$, predict a synapse at location $j$ with confidence $a_j$, remove from consideration all $i$ within some set distance of $j$ (i.e., $a_i$ set to 0 if $d(i, j) \leq r_n$), and then repeat. A final set of predicted synapses can be obtained by thresholding at a particular confidence value.

### 4 Experiments

We compare our approach against a commonly-used baseline for automated synapse detection. In this section, we first give details on the data set used for comparison, then show results in terms of precision-recall.

#### 4.1 Data and Evaluation

We evaluate synapse detection algorithms on a subset of the data used in Plaza et al. [12]. This electron microscopy (EM) data was imaged from seven medulla columns of the Drosophila optic lobe. The image volume, produced with FIB-SEM imaging, is of nearly isotropic, $10^3 \text{ nm per voxel}$ resolution.

The specific task we consider is to identify pre-synaptic sites in the Drosophila, often called T-bars due to a pedestal and platform structure forming a T-like shape. 20 image volumes of size $520^3$ voxels were used for testing, containing just over 5,000 T-bars total. A single $520^3$ voxel image volume was used for training.

We compute detection performance in terms of precision and recall. In order for a predicted synapse detection to be counted as a true positive, we require that it be within a certain distance of a ground-truth synapse location. For our experiments, we set this distance to be 30 voxels (300 nm), approximately equal to the width of a T-bar structure.
4.2 Results

We compare results against the automatic T-bar detection algorithm presented by Plaza et al. [12], which uses the Random Forest (RF) voxel-wise classifier from the ilastik toolkit [13]. This work used a $K$-means based post-processing algorithm to produce object predictions from voxel-wise output. We first compare this method with the averaging and non-maximum suppression approach given in Section 3.2.

The ilastik classifier is trained interactively, in an iterative procedure where manual, voxel-level annotations are made, the classifier trained, and the classifier output examined for whether additional manual annotations are necessary. As the labels are at the voxel level, this type of supervision is much more costly in terms of time to obtain than object-level annotations. Therefore, we also compare against an ilastik classifier trained statically, using the same approach used for the DAWMR networks, presented in Section 3.1.

Experimental parameters were set as follows: for forming training data, $r_l = 7$, for averaging voxel-wise predictions, $r_a = 7$, and for NMS, $r_n = 21$.

Results for these methods, compared with the proposed DAWMR network approach, are given in Figure 1. First, we can see that the averaging and NMS method for producing object predictions performs comparably to the $K$-means approach by Plaza et al., except at high recall where averaging and NMS performs significantly better. We hypothesize that this is due to the hard binarization that occurs in order to produce connected components, prior to the $K$-means step.

Next, we see that training the Random Forest classifier statically, using the simple transformation from object-level labels to voxel-wise labels, gives comparable performance despite requiring much less manual supervision and time than the typically used interactive approach. Lastly, the DAWMR network outperforms the Random Forest classifiers throughout the range of recall values.

![Figure 1: Precision-recall curve for automated T-bar detection.](image)

Figure 1: Precision-recall curve for automated T-bar detection. For Random Forest (RF) classifiers, using averaging and non-maximum suppression (-ave-nms) yields higher precision at high recall than $K$-means for forming object predictions, and training statically (-static) gives comparable results to interactive training, despite requiring much less supervision. The DAWMR network outperforms the RF methods throughout the range of recall values.
5 Discussion

Identifying synapses is a time-consuming and necessary component in determining a connectome from EM image data. In this work, we introduced techniques for automated synapse detection based on deep learning that dramatically improve precision at high recall, while decreasing the amount of supervision required for training.

In Plaza et al. [12], automated synapse detection, thresholded to produce a recall of approximately 0.9, is used as input to a manual verification step. For the data set they considered, this manual verification required approximately 350 hours. Our proposed DAWMR-based method doubles the precision at a 0.9 recall, therefore by substituting this method, the time required can be reduced by half.

Unlike the problem of automatically segmenting neuronal shapes, we believe automatic synapse detection is more tolerant of mis-predictions, as many connections are formed from a high number of synaptic contacts, and thus the underlying connectivity graph can still be recovered even if some fraction of synapses are not detected [15]. Additionally, Plaza et al. also estimated human-to-human manual agreement to be close to 0.9 precision at 0.9 recall. For these reasons, our work represents a significant step toward achieving human-level accuracy and fully-automated synapse detection, which will considerably enhance downstream algorithms for focusing annotation efforts and improving segmentation [12].

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