Mitochondria detection in electron microscopy images
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Objective
We present an approach for detecting mitochondria in electron microscopy images of brain tissue and demonstrate it on the rat neuropil. This would enable the study of statistics of mitochondria and their location relative to other structures such as cell boundaries, vesicles and synapses.

Data and challenges
Neural tissue from the Lateral part of the rat's brain was cut into 50nm slices and photographed with a Transmission Electron Microscope (TEM) to generate a 3D stack of grayscale images. Figure 1 shows an example of the images obtained.

Figure 1: Example of EM image of Rat neuropil.

Appearance of mitochondria is characterized by dark ribbed texture. The detection is challenging:
1. The density of the ribbing varies across mitochondria.
2. Mitochondria have approximately tubular shape with variable length and cross-section.
3. Other parts of the tissue such as vesicles and cell boundaries appear similar to mitochondria in electron micrographs.

**Approach**

Our approach is to detect image pixels likely to belong to mitochondria based on texture features. It consists of the following steps:

1. Histogram equalization and median filtering is performed to reduce variations in electron beam intensity and noise.
2. Histograms of grayscale and Gabor filter responses are computed in neighborhood windows around each pixel. A large set of window sizes and Gabor filter frequencies are applied simultaneously, enabling generalization to different tissue characteristics. This results in a large dimensional (in 100s) feature vector for each pixel.
3. Machine learning is used to recognize pixels located within mitochondria. Our approach used Gentle-Boost as it is well suited for handling large number of potentially irrelevant and redundant features. The classifier computes a *mitochondria confidence* map for each image plane. Pixels with high values are likely to belong to mitochondria.
4. Vesicle detection cues were used to reduce false detections within vesicle clusters.
5. Morphological filtering is employed for computing high confidence clusters in 3D. This consisted of connected component analysis and filling in holes within detected clusters.

**Experimental results**

A stack of 20 863x863 EM images was manually annotated with regions occupied by mitochondria. These were used to train and test the approach using a 2-fold protocol wherein 50% of the data was used for training and the remaining for testing.

![Figure 2: False acceptance and false rejection ratios for mitochondria detections](image)

The detections computed by approach were evaluated using false acceptance/rejection curves. Let $m$ be
the total number of voxels occupied by mitochondria. For our dataset \( m=631621 \). Let \( t \) denote a threshold applied on the mitochondria confidence maps. Let \( n( t ) \) be the number of voxels classified to belong to mitochondria at threshold \( t \). Let \( c( t ) \) be the number of voxels correctly classified as mitochondria.

The false acceptance ratio is defined as

\[
FA = \left( \frac{n( t ) - c( t )}{n( t )} \right).
\]

The false rejection ratio is defined as

\[
FR = \left( \frac{m - c(t)}{n(t)} \right).
\]

Figure 2 shows the FA-FR curves for mitochondria detection in the presence and absence of vesicle detection cues and morphological filtering. It indicates the efficacy of including vesicle detection cues and morphological filtering. For mitochondria confidence threshold at 0.5, the false acceptance ratio was \( \sim 25\% \) and false rejection ratio was \( \sim 20\% \).

Figure 3 shows an example of manually annotated and automatically detected mitochondria regions overlayed on images. Many of the false detections occurred in regions in which cell boundaries grazed the cutting plane producing a large gray textured area.

Summary and future work

We presented an approach for automatic detection of mitochondria in electron microscopy images and applied it rodent brain tissue. Future work includes

- Quantitative evaluation on a larger dataset.
- Investigating the approach's applicability to other tissue types and preparations.
- Employing the detections to analyze the location and density of mitochondria relative to neural structures.
Figure 4 shows mitochondria detection volumes on 90 slice stack.