**Setting up the required packages**

1. Designate a root folder. In this example, it is *E:\EMImages*.
2. Download and copy the *Tools* folder into the parent folder, as in *E:\EMImages\Tools*.

Alternatively, download and copy the files from the websites listed below into the *Tools* folder.

* 1. The ba\_interpolation package, available here -   
     <http://www.mathworks.com/matlabcentral/fileexchange/20342-image-interpolation-bainterp2>
  2. MR8 filter bank, available here -  
     <http://www.robots.ox.ac.uk/~vgg/research/texclass/filters.html>
  3. HoG descriptors, available here -   
     <http://www.mathworks.com/matlabcentral/fileexchange/33863-histograms-of-oriented-gradients>
  4. LibSVM, available here -  <http://www.csie.ntu.edu.tw/~cjlin/libsvm/>
  5. the RFS filter - <http://www.robots.ox.ac.uk/~vgg/research/texclass/>

1. Run the script *load\_packages.m*. This will compile all the required MEX files.

(Type ‘load\_packages’ in the command window and hit ‘Enter’).

**Analysis of images for synapse detection**

1. **Folder structure –**

All images from a single sample is stored in a specific folder. The folders of different samples are in one parent directory which resides in the root folder (here – E:\EMImages) where all the packages have been installed.

1. **Labeling the synapses**

This step lets the user manually label synapses across all samples and save the labeled images in a single folder

Program to be used – *build\_training\_data\_from\_segments.m*

While calling the function,

1. Specify the parent folder containing all samples.
2. Name a new folder where the program will store all the labeled objects, e.g. ‘trainingset\_P1839’.

It should be called like this –

build\_training\_data\_from\_segments(‘P-1839’,'trainingset\_P1839');

1. **Analyzing the images**

Program to be used *run\_synapse\_detection.m*

It should be called like this –

run\_synapse\_detection(‘P-1839’,'trainingset\_P1839');

* 1. Parent folder is *P-1839*
  2. The second argument would be the folder you created to store your labeled images (step 2.ii). Here, it would be *trainingset\_P1839*. This is used by *build\_features\_samplewise* to build the feature set for the labeled objects.
  3. The confidence threshold will be determined that will proved a recall of 50%. This is done by generating the ROC curves in the function *determine\_threshold* and then using *get\_threshold* to choose the appropriate threshold.
  4. Then, the function *test\_directory* will go through all samples individually that are part of the parent folder and analyze all the images in each sample folder to detect synapses based on the threshold.