02740 Project #2: Particle Detection

Alan Shteyman, Luigi Leung, Yiming Xin, Yuxian Ruan

B.1 Image data

The image sequence was downloaded

B. 2. Part I: Particle feature detection

B.2.1 Calibration of dark noise

The mean and standard deviation of the background was found to be ~ 307 and ~24, respectively.

B.2.2 Detection of local maxima and local minima

The goal in this section is to filter the image into points (coordinates) of local maxima and local minima with a mask.
Using the equation $3\sigma = 0.61\lambda/NA$. Since one pixel is 65nm wide, then σ is about 1.18 pixels. 1.18 was used to calculate the Gaussian Mask which was a 3X3 matrix.We then compared the performance of using a 3X3 mask vs. a 5X5 mask for finding local maximums.
It is observed that there are more local maxima and local minima found in 3x3 than in 5x5. This makes sense due to smaller masks result in more positions getting picked.

Figure 1a. Local maxima using 3x3 Mask

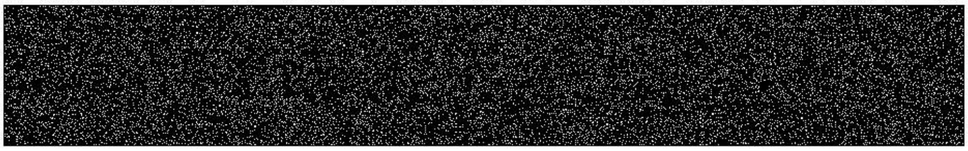


Figure 1b. Local maxima using 5x5 Mask

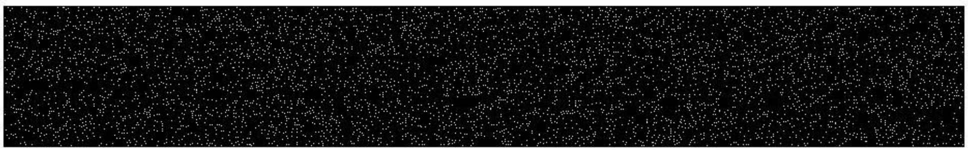


Figure 2a. Local minima using 3x3 Mask

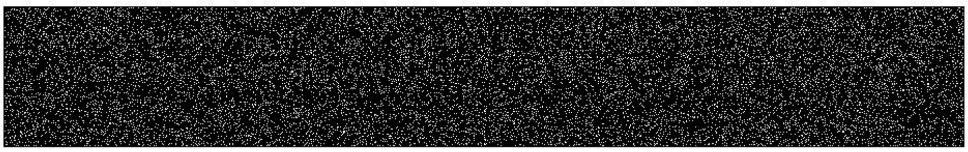
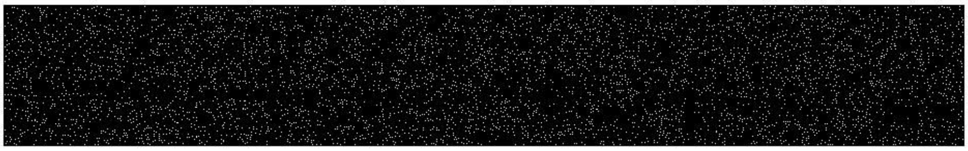
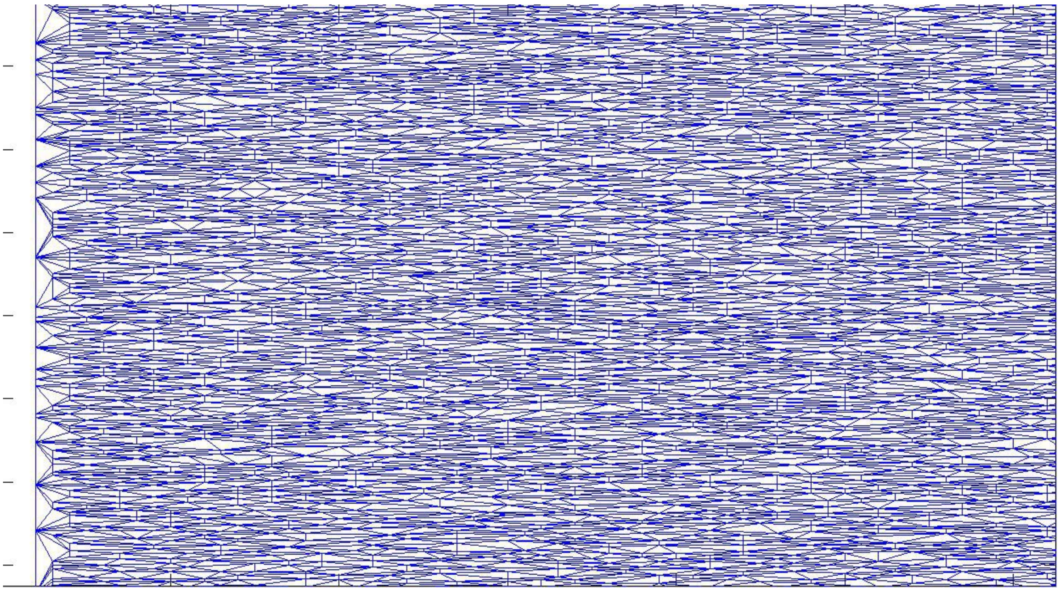


Figure 2a. Local minima using 5x5 Mask



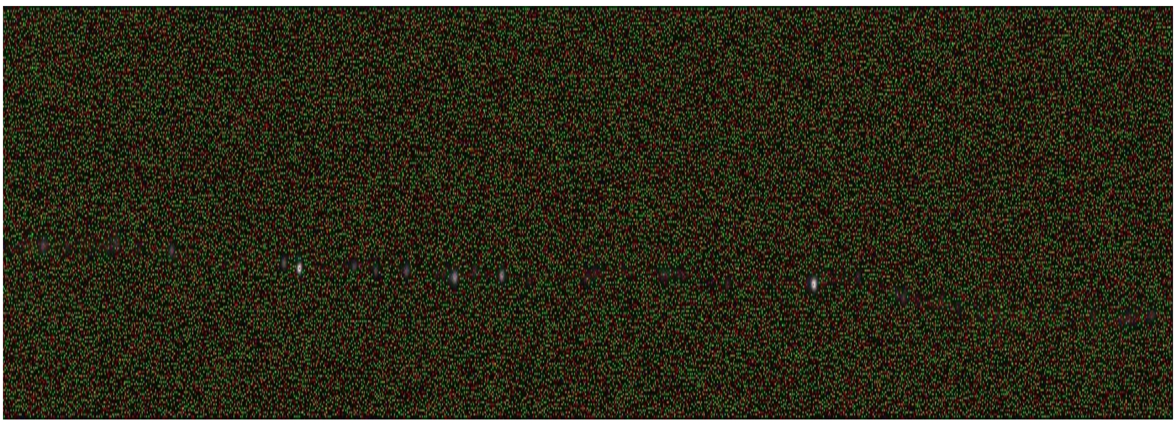
B.2.3 Establishing the local association of maxima and minima

We use Delaunay triangulation to establish corresponding between local maxima and local minima. By checking the position of local minima (labeled in the previous process), we get the vectors for its x coordinate and y coordinate, then using *delaunayTriangulation()* function. Since the image itself is very large and consequently the number of local minima is quite big before t test, we would see that the triangulation appears very compact in the space.



B.2.4 Statistical selection of local maxima

Before we conduct the filtering step, the local maximum and minimum is shown as following:

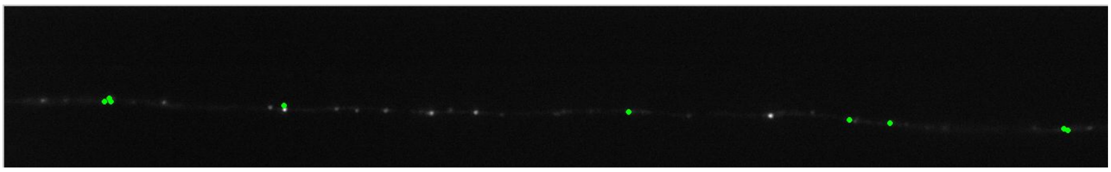


localMax: green ; localMin: red

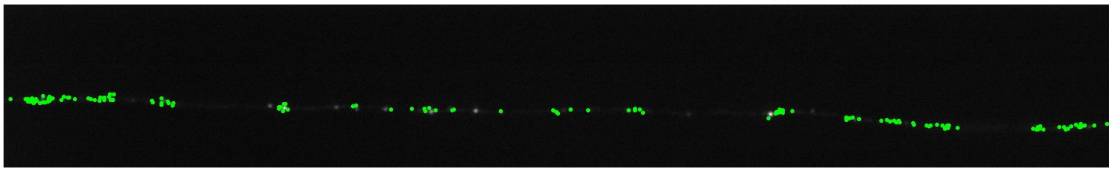
We perform the t-test for all the local maximum using the formular mentioned in reference 1.

We calculated the local background of a specific local maximum using the mean of nearest triangle’s local minimums.

```
if (localmax(i,j) == 1)
    mean=trianglemean(j,i,DT,x,y,image);
    T = image(i,j) - mean;
    tSet = [tSet T];
    if(T < Q*sigma)
        localmax(i,j) = 0;
    end
```



Q = 10.0, Sigma = 13.2

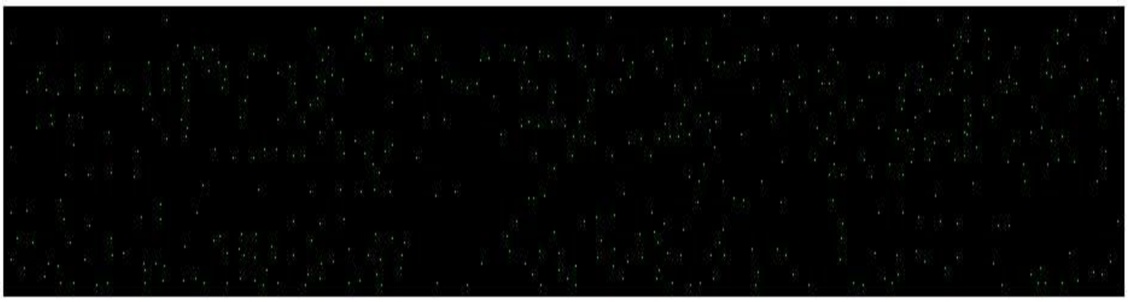


Q = 4.0 Sigma = 13.7

B.3 Part II: Sub-pixel resolution particle detection

B.3.1 synthetic image generation

The goal of this section is to visualize ground truth of the original image using previously found local maxima.



B.3.2 sub-pixel resolution detection using pixel oversampling

We perform the oversampling on each local maxima remained after the t-test. First we get the vectors and matrix needed as the input of *interp2* function, taking the nearest 8 neighbors of the target local maxima as the first three arguments of *interp2* function。 Then we divide the pixel into 5X5 parts and set the index of each part. Further, we use these 25 new image intensity values and

the corresponding indexes to perform the Gaussian fitting (using *lsqcurvefit* for least-square fitting) for the local maxima and we can get the sub-pixel top point coordinates of the Gaussian.
The result contain the relative position of the (x_0,y_0) in the 25 sub-pixels, such like (1,5) is denominated as the top right corner of the local maximum pixel.

B.3.3 Benchmarking subpixel resolution particle detection
Run the code testpart3.m to check it.

References

[1] A. Ponti, P. Vallotton, W. C. Salmon, C. M. Waterman-Storer, and G. Danuser, Computational analysis of F-actin turnover in cortical actin meshworks using fluorescent speckle microscopy, Biophysical Journal, 84:3336-3352, 2003.
[2] M. K. Cheezum, W. F. Walker, and W. H Guilford, Quantitative comparison of algorithms for tracking single fluororescent particles, Biophysical Journal, 81:2378-2388, 2001.