

Output of "findCandidateSpots" function

Data: Frame 11 of 0320_01_Fab_EGFR_473

Parameters:

1. signal to noise ratio (SNR) = 2
2. r^2 of the gaussian fit = 0.2
3. minimum sigma of the fit = - inner circle mask radius
4. maximum sigma of the fit = inner circle mask radius

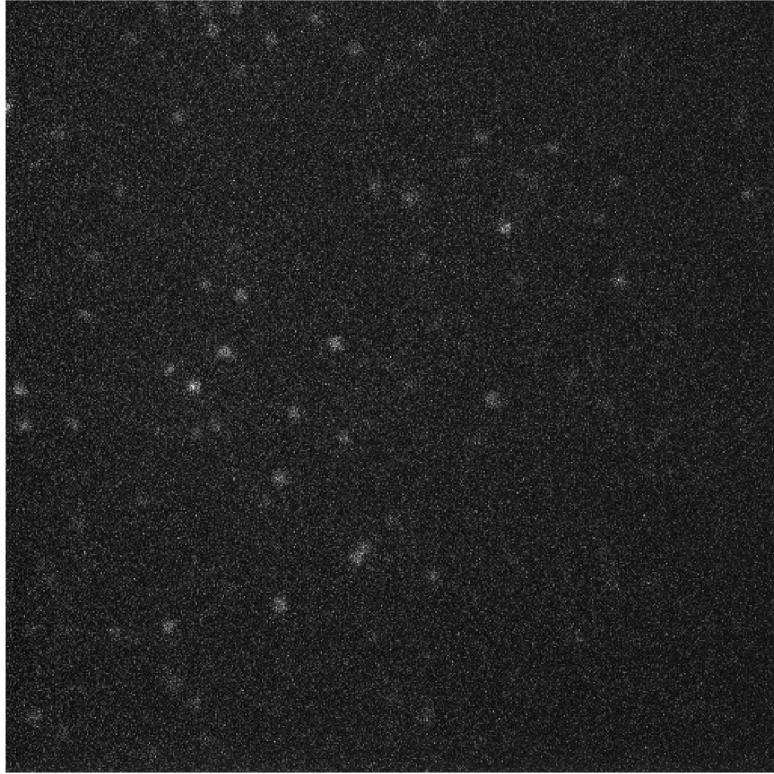


Figure 1: The raw data - a single frame from a video of GFP immobilised on a glass slide.

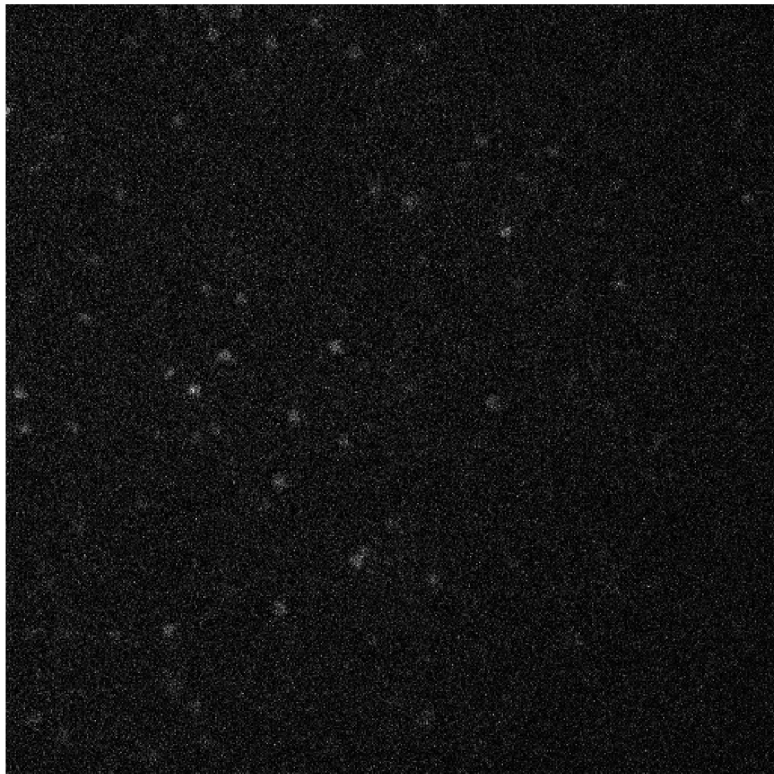


Figure 2: Evening out the background by applying a top hat transformation to the raw data (Figure ??).

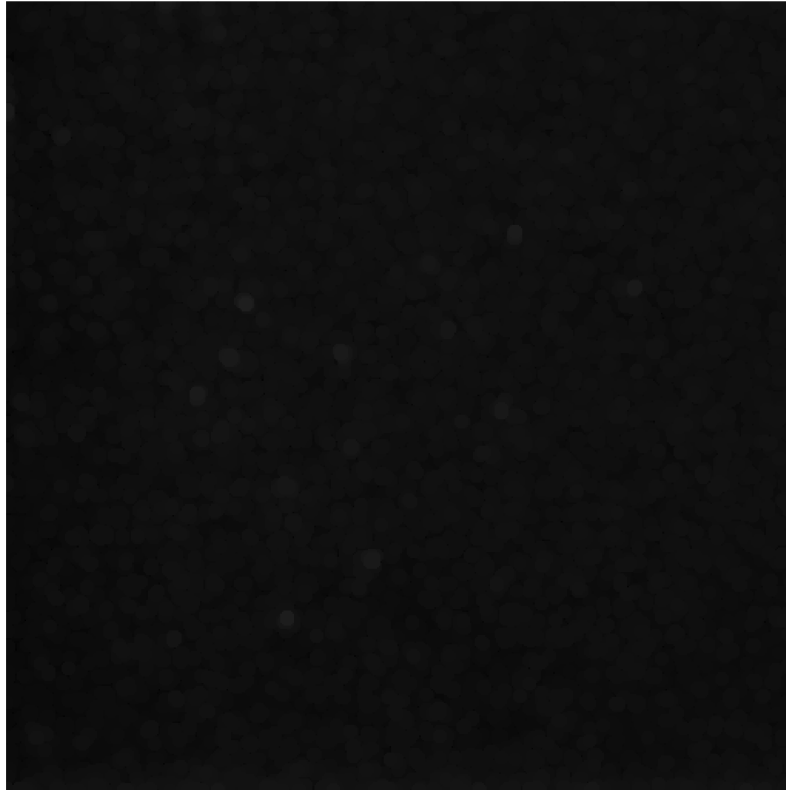


Figure 3: Morphological opening of Figure ??, with a disk shaped structural element of radius of 5 pixels. This is to estimate the background.

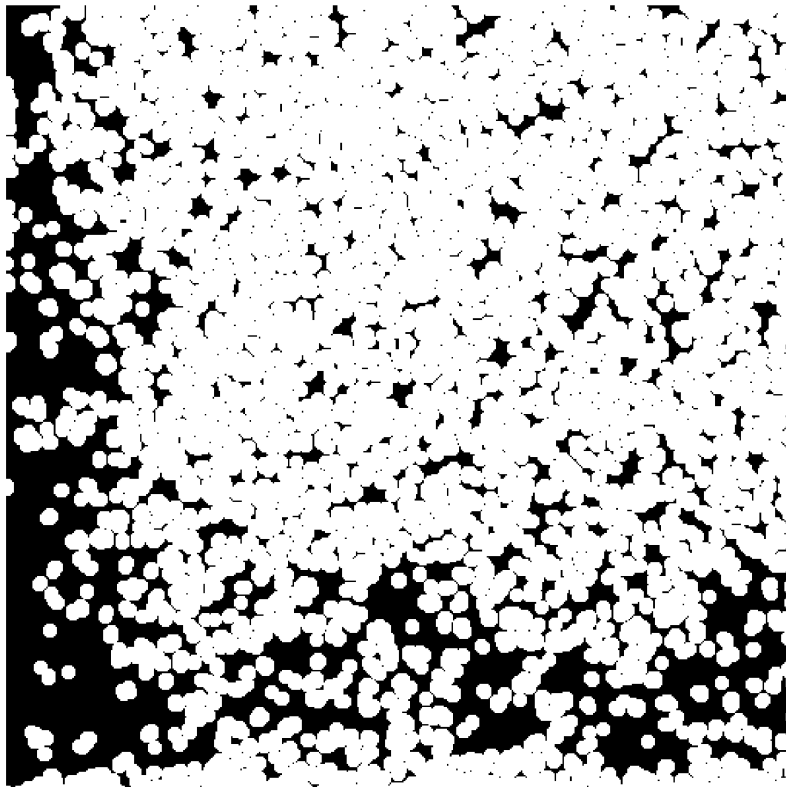


Figure 4: Thresholding (using Otsu's method) of Figure ??, followed by conversion to a binary image, resulting in a Signal Mask

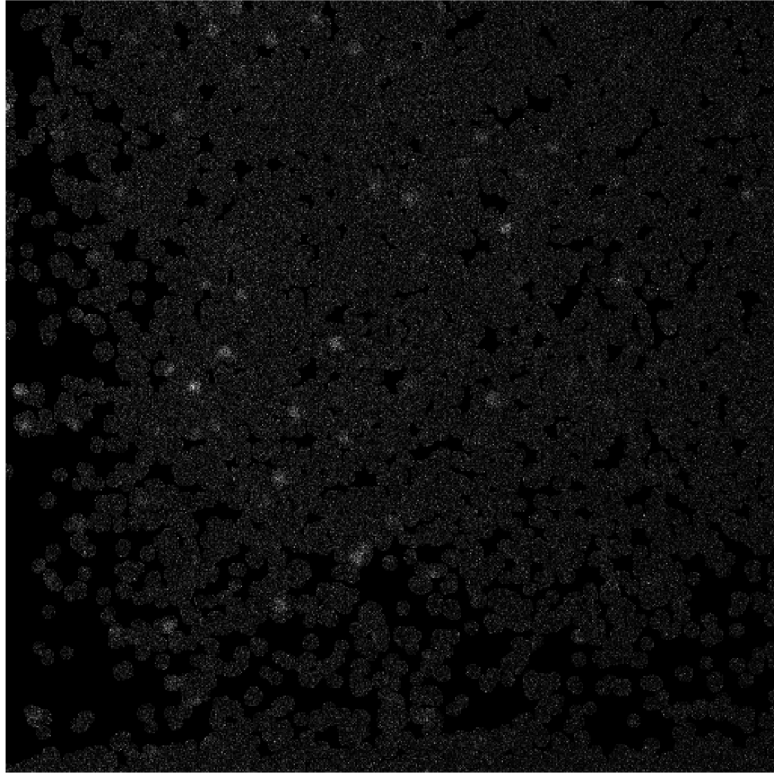


Figure 5: The top hat transformation of the raw data (Figure ??) is pushed through the Signal Mask (Figure ??) to remove the background signal.

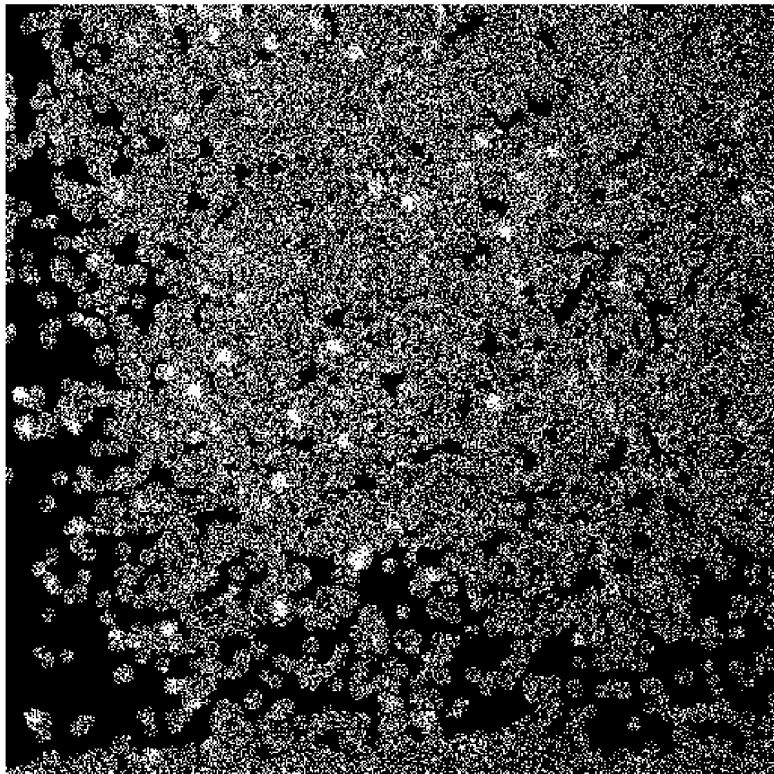


Figure 6: Thresholding (using Otsu's method) of Figure ??, followed by conversion to a binary image.

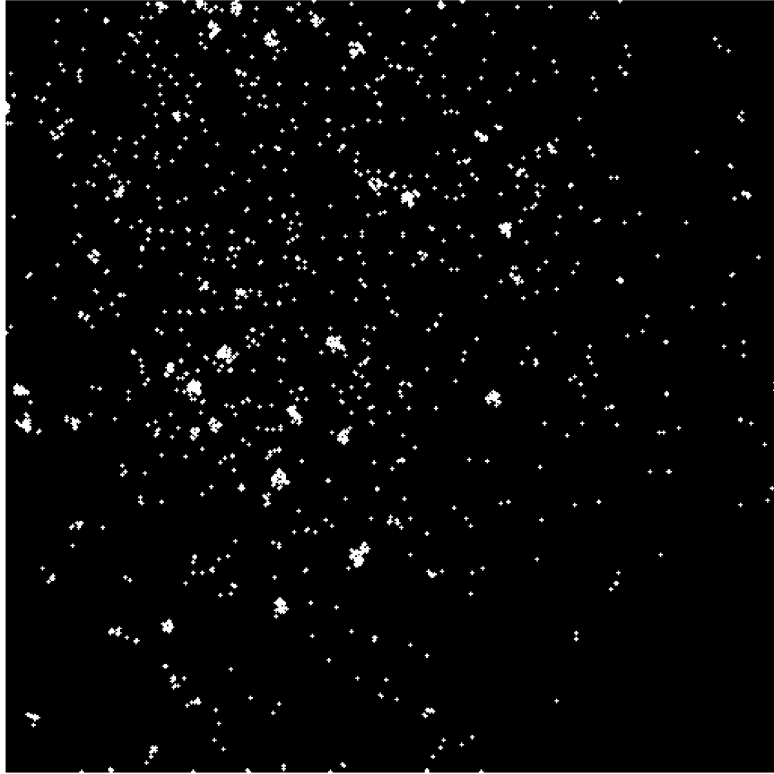


Figure 7: Removal of single foreground pixels by application of morphological opening with a disk shaped structural element, of radius 1 pixel, to Figure ??.

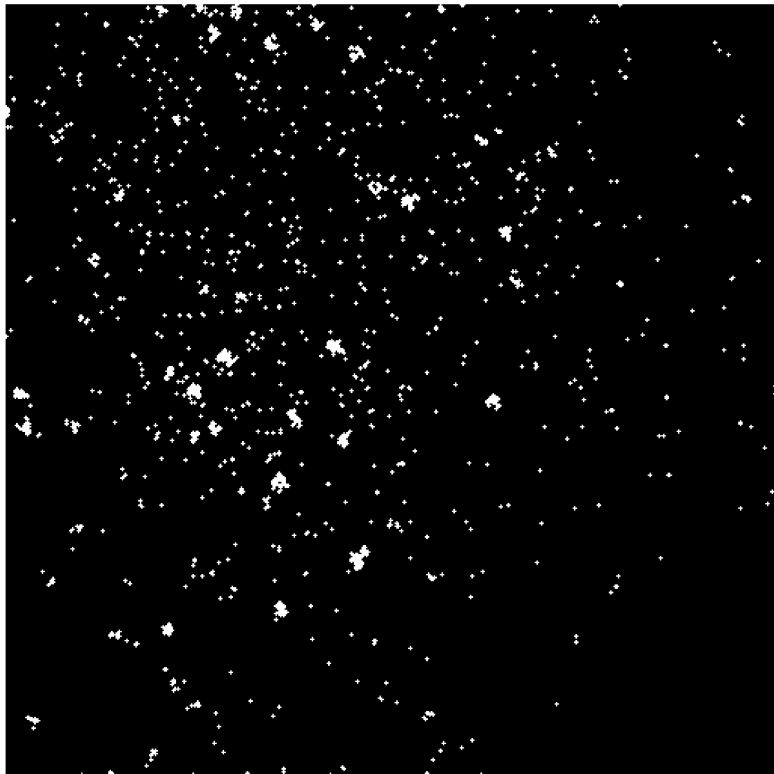


Figure 8: Single background pixels in Figure?? are allocated to foreground signal.



Figure 9: Morphological opening with a square structural element, of size 3×3 pixels, of Figure ??.



Figure 10: Ultimate erosion of Figure ??.

(For each initial group of foreground pixels, repeated erosions are performed until any further erosion would eliminate the group of foreground pixels completely.)

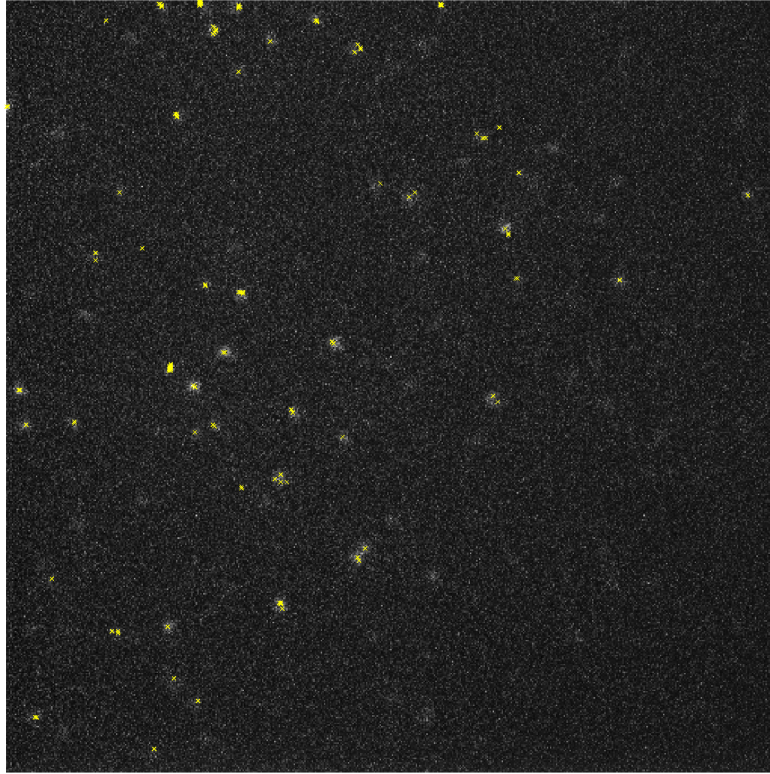


Figure 11: This figure shows the resulting "candidate spot centres" in yellow (same as Figure ??) plotted on top of the original data from (Figure ??).

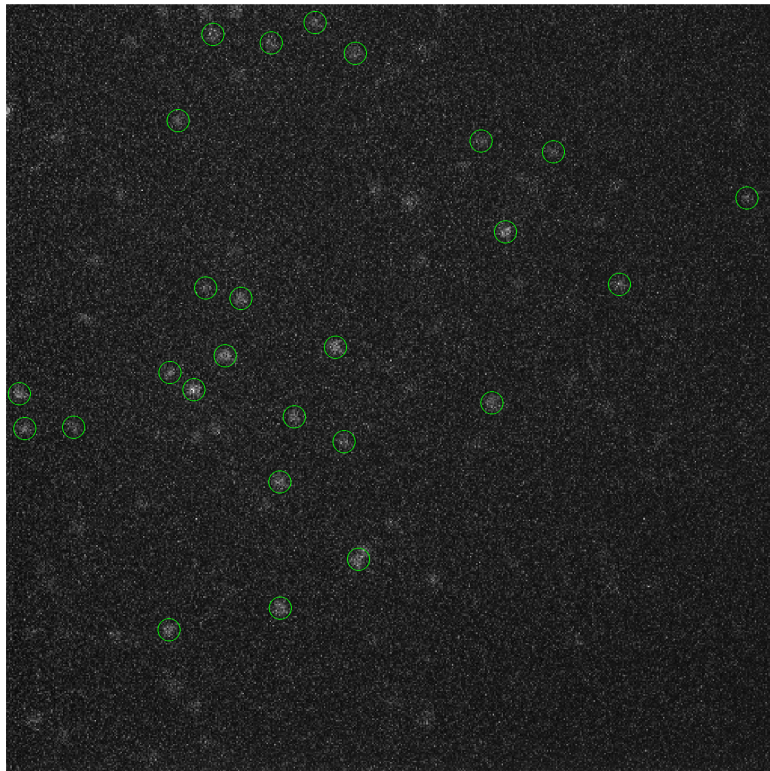


Figure 12: This figure demonstrates the output of the next part of the algorithm. The next part uses the "candidate spot centres" as a starting point for fitting a gaussian to the spot. The green circles indicate the spots where gaussians have been fitted and accepted by the algorithm.