Blob Analysis

Activity 10 Short Report

Lou Josef S. Tan

Tasks

- Integration of image processing skills (image segmentation, morphological operations, labelling, etc.)
- Measure several properties of cells (e.g., areas, centroids, eccentricities) using blob analysis

Segmentation of Cells

Given the image in Fig. 1, we were to segment the cells perfectly (i.e., no overlaps, no fused blobs), so that we could subject them to blob analysis and measure several properties.

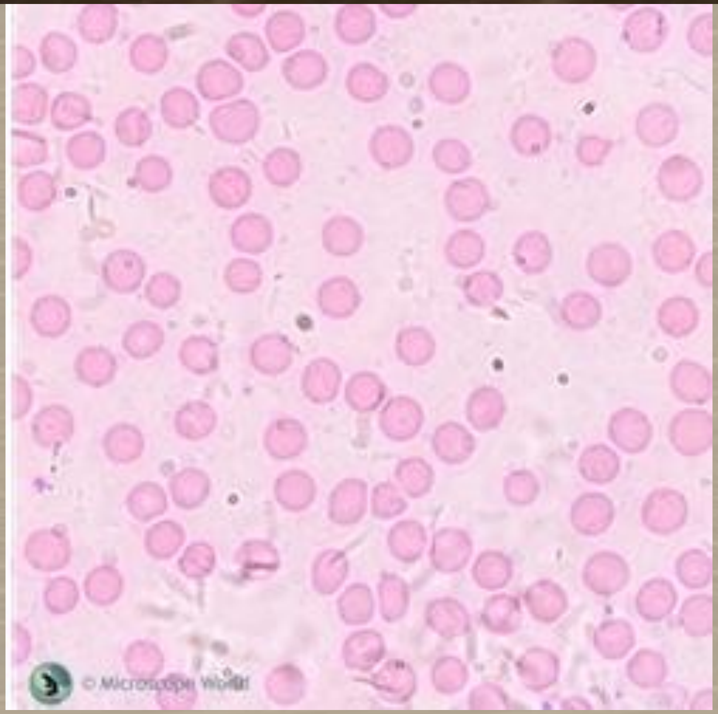

Figure 1. Image of cells

Segmentation of Cells

Upon segmentation, we get the resulting image displayed in Fig. 2. At first glance, the initial result might seem good enough. However, some of the blobs are connected, when in reality, they should be completely separated. Blob analysis here would be erroneous.

We then attempt to separate these connected blobs.

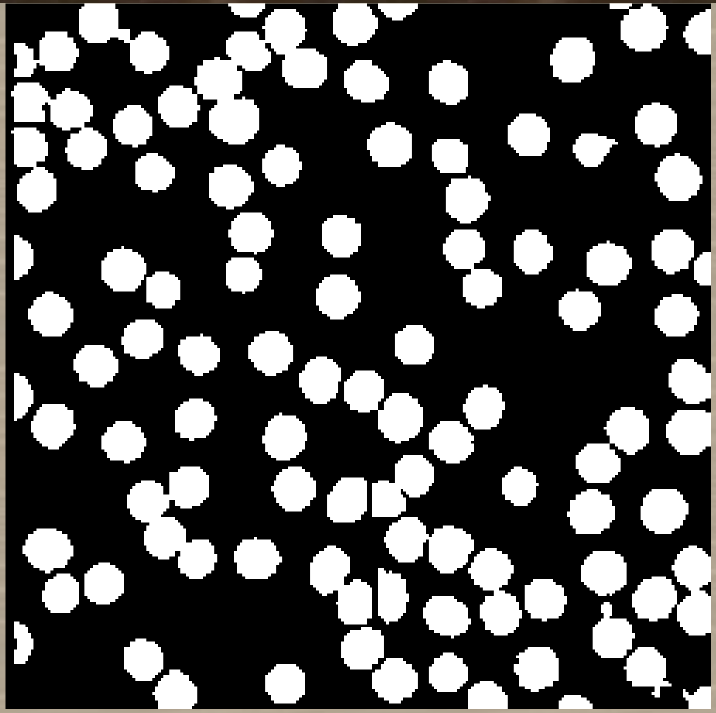

Figure 2. Segmented image of cells

Segmentation of Cells

To do this, we take only the connected blobs from the segmented image as in Fig. 3. To separate the blobs, we use a segmentation technique known as *watershed segmentation*.

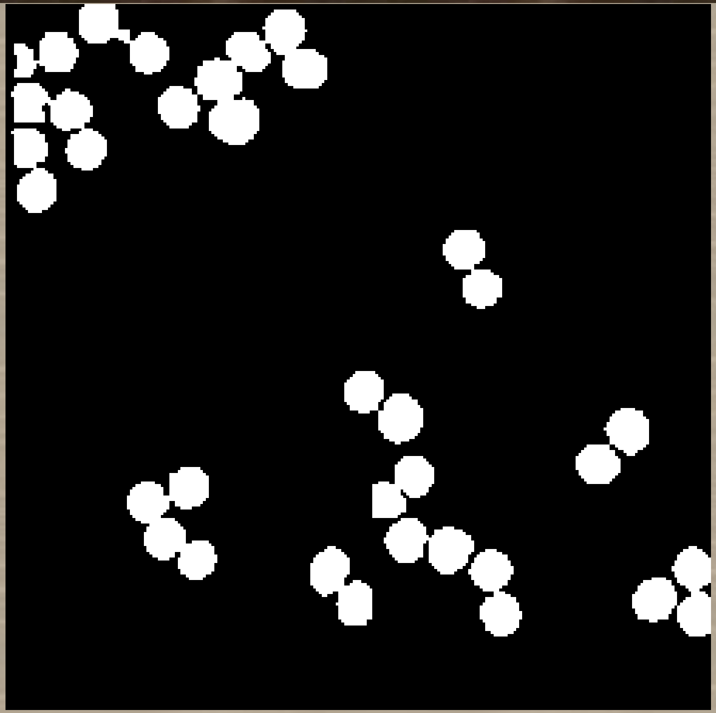

Figure 3. Connected blobs from segmented image.

Segmentation of Cells

Watershed Segmentation[1]

Watershed segmentation makes use of the idea of watersheds and catchment basins. In its literal definition, watersheds are ridges that divide different catchment basins, and catchment basins are areas that drain into rivers or reservoirs.

[1] – For more info about watershed segmentation, visit: https://www.mathworks.com/company/newsletters/articles/the-watershed-transform-strategies-for-image-segmentation.html

Segmentation of Cells

Watershed Segmentation[1]

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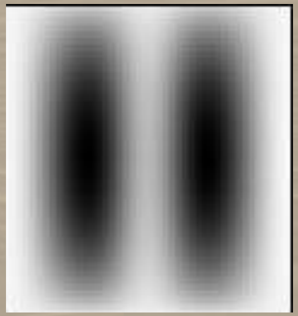
Relating this to image processing, say, we have two dark blobs, its corresponding watershed and catchment basin is shown in Fig. 4.



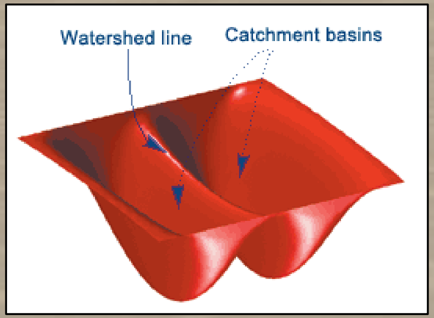


Figure 4. L: Two dark blobs R: Corresponding watershed and catchment basin.

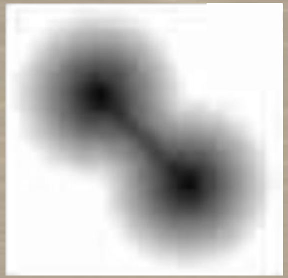
[1] – For more info about watershed segmentation, visit: https://www.mathworks.com/company/newsletters/articles/the-watershed-transform-strategies-for-image-segmentation.html

Segmentation of Cells

Watershed Segmentation[1]

The **`watershed()`** function is then able to find the watershed lines and catchment basins for grayscale images. The general procedure for watershed segmentation is presented below:

Initially, there are two touching objects to be separated via watershed segmentation



The distance transform of the image is taken. This now resembles the two dark blobs in the previous slide: we have two catchment basins

Watershed Transform

The watershed transform is taken, which outputs a label matrix containing (+) integers for the locations of catchment basins, and watersheds (zero-valued).

Using the zero-valued elements (watershed lines) of the watershed transform, we separate the blobs.

[1] – For more info about watershed segmentation, visit: https://www.mathworks.com/company/newsletters/articles/the-watershed-transform-strategies-for-image-segmentation.html

Segmentation of Cells

Going back to our image of connected blobs. We now take its distance transform.

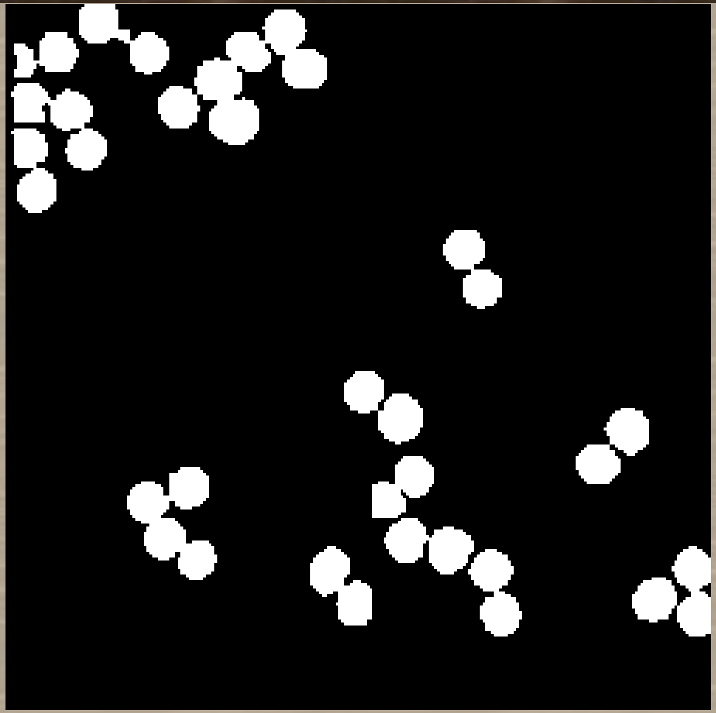


Figure 3. Connected blobs from segmented image.

Segmentation of Cells

Going back to our image of connected blobs. We now take its distance transform. From Fig. 5, the catchment basins are very apparent.

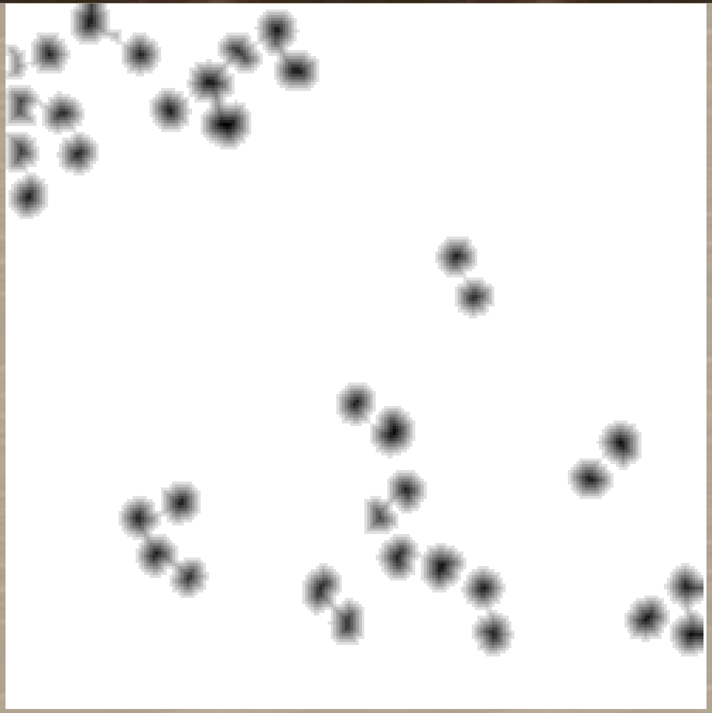


Figure 5. Distance transform of connected blobs

Segmentation of Cells

Going back to our image of connected blobs. We now take its distance transform. From Fig. 5, the catchment basins are very apparent. We could now take the watershed transform, and then completely separate the connected blobs.

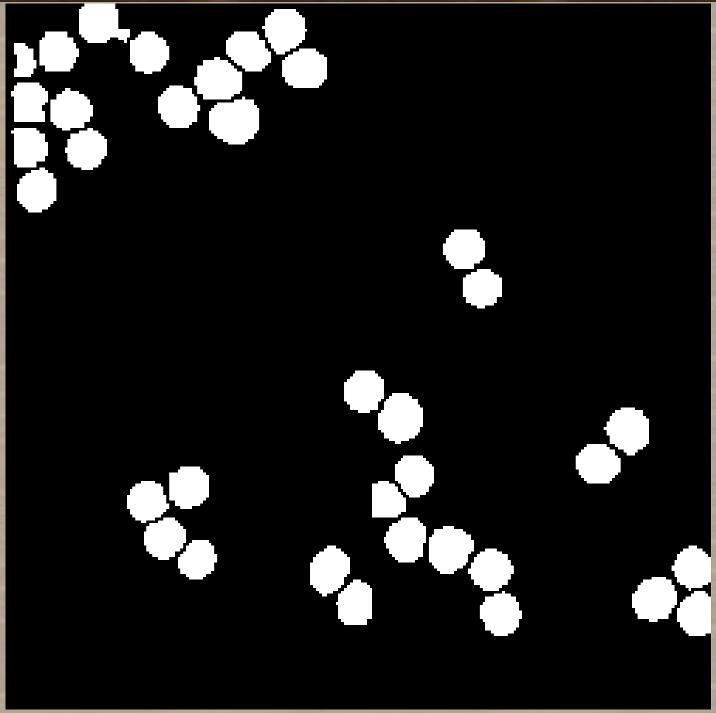


Figure 6. Separation of connected blobs via watershed segmentation.

Segmentation of Cells

Going back to our image of connected blobs. We now take its distance transform. From Fig. 5, the catchment basins are very apparent. We could now take the watershed transform, and then completely separate the connected blobs.

Finally, we combine the newly-separated blobs with the already-separated blobs from before. Our segmented image is now ready for blob analysis.

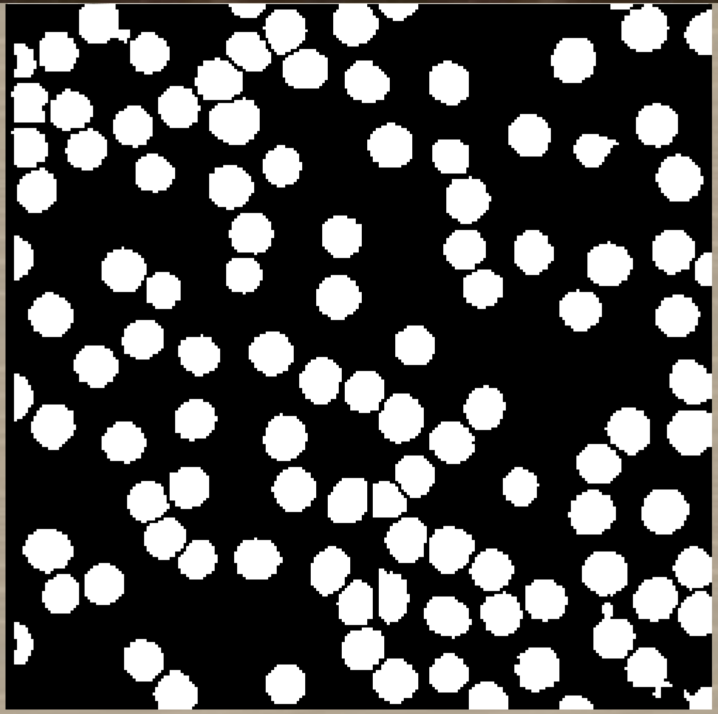


Figure 7. Segmented image with completely separate blobs

Blob Analysis

Centroid and Bounding boxes

First off, we use `regionprops()` to take the centroids and bounding boxes of each blobs in our image. In total, there are 111 cells in our image.

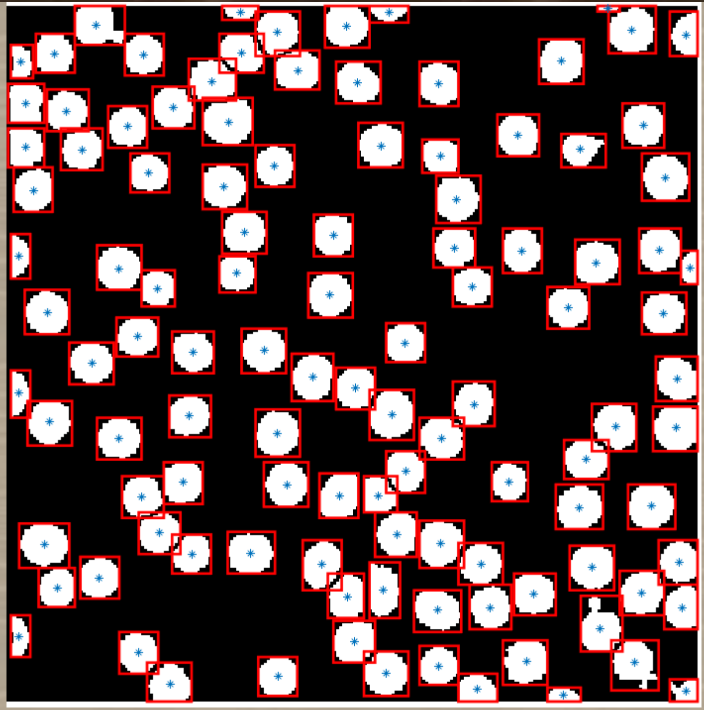


Figure 8. Blobs (cells) marked by the centroids and enclosed in bounding boxes.

Blob Analysis

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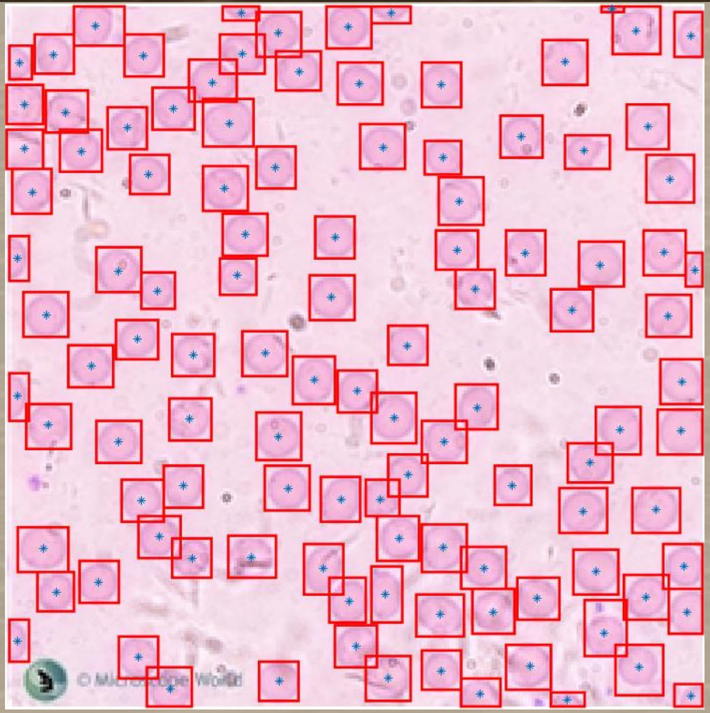

Figure 8. Blobs (cells) marked by the centroids and enclosed in bounding boxes.

Blob Analysis

Area

Shown to the right is the histogram of the pixel area of the cells. The left region of the histogram (Area < 120) comprises the half-cells at the borders of the image. These half-cells were excluded in computing the average cell size. The best estimate for the average cell size is 179.7 ± 21.4 sq. pixels.

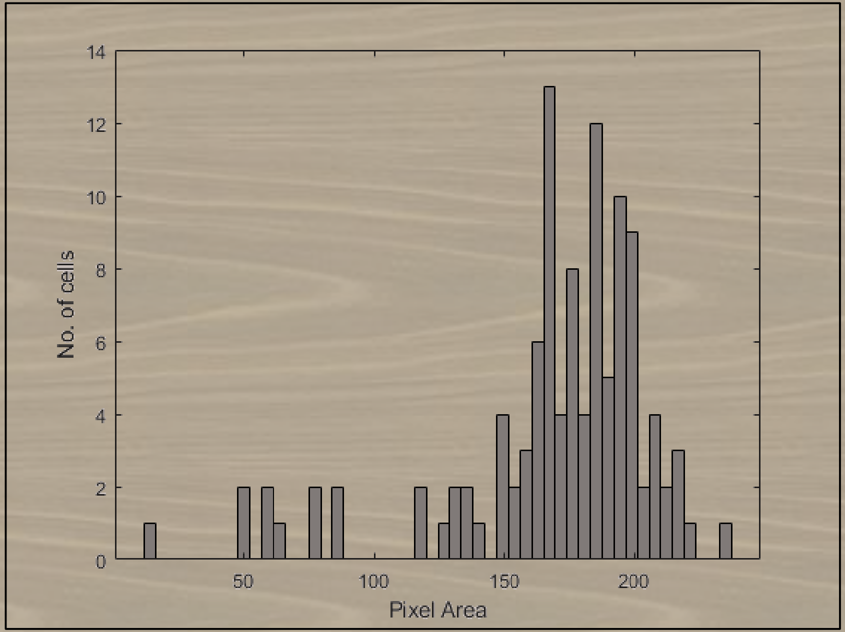

Figure 9. Histogram of the pixel area of cells.

Blob Analysis

Perimeter

On the other hand, the best estimate for the average perimeter of the cells is 45.9 ± 3.4 pixels.

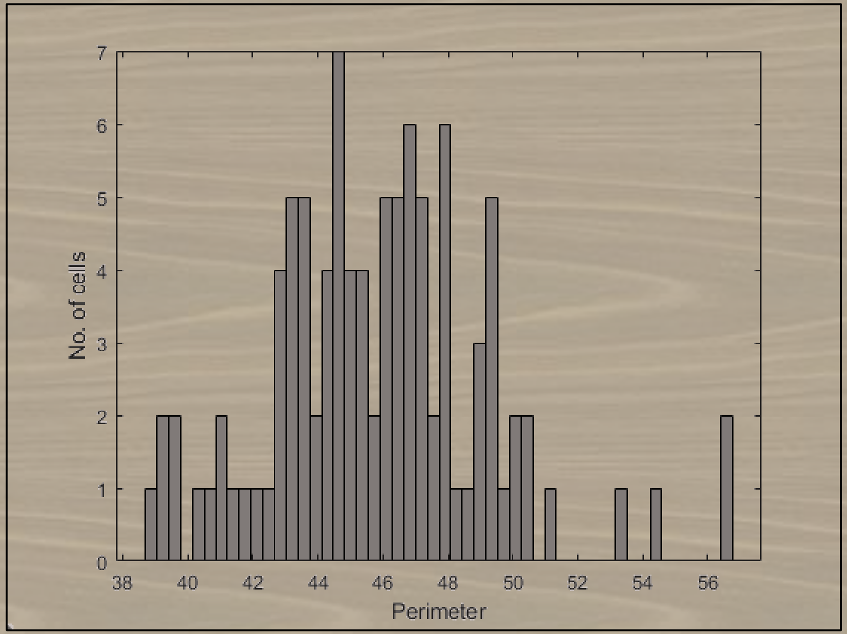

Figure 10. Histogram of the perimeter of cells.

Blob Analysis

Eccentricity

Here we have the histogram of the eccentricity of the cells. We can see that majority of the cells have eccentricities below 0.5, leaning towards being nearly circular. Computing for the average eccentricity we have 0.39 ± 0.13.

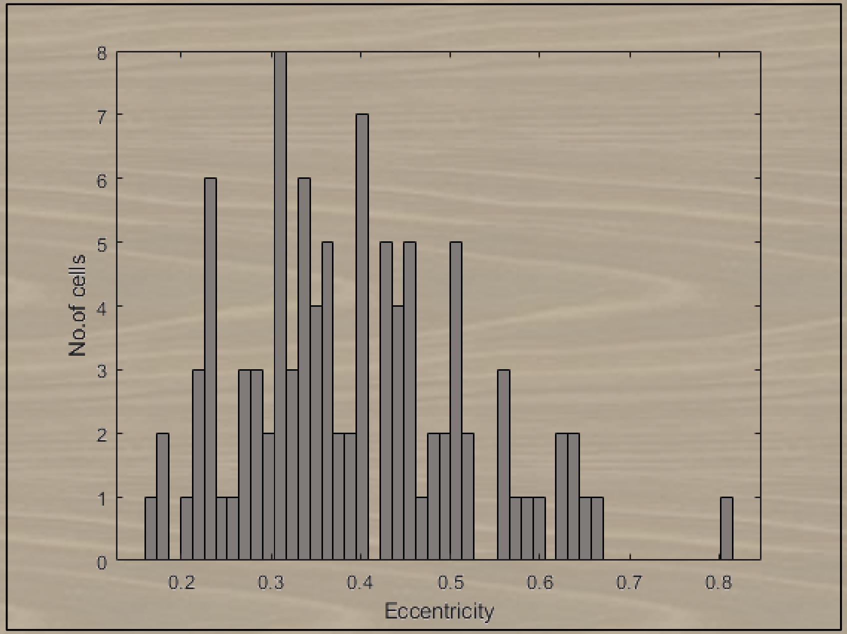

Figure 11. Histogram of the eccentricity of cells.

Blob Analysis

Major and Minor Axis Length

The average major axis length of the cells is 16.0 ± 1.1 pixels, while the average minor axis length is 14.5 ± 1.0 pixels. The values for both averages are near, indicating that the cells may be nearly circular. This agrees with what we previously observed for the average eccentricity.

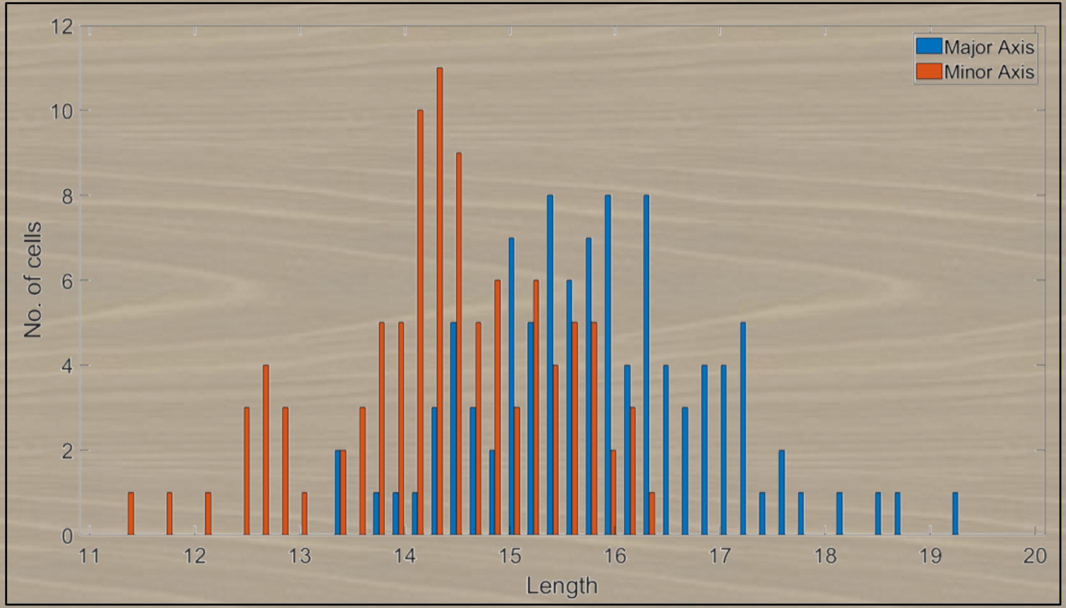

Figure 12. Histogram of major and minor axis lengths of cells.

Evaluation

I give myself a score of 11/10 for producing all required output, and applying advanced techniques for image segmentation (watershed segmentation).

References:

[1] – The Watershed Transform: Strategies for Image Segmentation. https://www.mathworks.com/company/newsletters/articles/the-watershed-transform-strategies-for-image-segmentation.html

[2] – Watershed transform question from tech support. https://blogs.mathworks.com/steve/2013/11/19/watershed-transform-question-from-tech-support/

[3] – Color-Based Segmentation Using K-means Clustering. https://www.mathworks.com/help/images/color-based-segmentation-using-k-means-clustering.html;jsessionid=ffc56fe3a1ed3900594bba3fbde3