Image Segmentation

By
Yashwanth Sai AND
Sandipta Subir Khare

Contents discussing
Segmentation, Type of Segmentation, Particle Analysis, Counting Cells, Thresholding, Multi-Thresholding, Otsu Segmentation, Cell Segmentation, Measuring Intensity

WHAT IS SEGMENTATION?

Image segmentation is a technique for partitioning an image into various portions or areas, frequently based on the characteristics of the pixels in the image. Separating the foreground from the background or clustering pixels based on color or shape similarity are examples of image segmentation.

TYPES OF SEGMENTATION DISCUSSING

- Thresholding
- Multi-Thresholding
- OTSU Thresholding
- Cell segmentation

Particle analysis

Analysing each particle information in image

The particle analysis can be automated via plugins or macros once the correct threshold value and particle size range has been determined for your objects of interest.

ImageJ File Edit Image Process Analyze Plugins Window Help

Wed Dec 8 0:50

Results

	Area	Mean	Min	Max	Circ.	AR	Round	Solidity
107	83	105.663	79	141	0.387	1.531	0.653	0.476
108	74	116.230	85	141	0.702	2.091	0.478	0.860
109	67	86.194	79	97	0.712	2.230	0.448	0.865
110	150	102.700	78	141	0.797	1.809	0.553	0.915
111	183	87.503	78	103	0.488	2.193	0.456	0.728
112	201	98.667	78	126	0.609	1.043	0.959	0.776
113	155	111.471	80	141	0.690	1.497	0.668	0.749
114	177	98.119	78	126	0.843	1.748	0.572	0.915
115	239	105.071	78	141	0.600	1.160	0.862	0.630
116	358	111.014	78	141	0.825	1.308	0.764	0.856
117	357	113.423	78	141	0.470	1.489	0.672	0.703
118	82	84.476	78	96	0.567	1.378	0.726	0.726
119	162	96.457	79	137	0.772	1.855	0.539	0.866
120	691	113.918	78	141	0.216	2.133	0.469	0.406
121	1403	107.520	78	141	0.070	1.511	0.662	0.347
122	223	102.516	78	139	0.835	1.293	0.774	0.910
123	73	114.096	85	141	0.875	1.384	0.722	0.901
124	520	102.063	78	141	0.238	3.077	0.325	0.470
125	22	82.045	78	85	1.000	1.101	0.908	0.936
126	92	93.750	78	118	0.850	1.407	0.711	0.872
127	285	99.091	78	134	0.504	1.684	0.594	0.723
128	198	107.657	78	141	0.781	1.473	0.679	0.834
129	42	110.071	85	140	1.000	1.128	0.886	0.894
130	202	102.045	78	138	0.778	1.777	0.563	0.904
131	177	108.605	78	139	0.741	1.304	0.767	0.760
132	311	107.447	78	141	0.272	3.200	0.312	0.487
133	35	82.000	78	85	0.569	2.928	0.342	0.722
134	134	98.754	79	141	0.815	1.848	0.541	0.912
135	97	90.062	78	114	0.489	1.496	0.668	0.716
136	38	110.474	85	141	1.000	1.078	0.927	0.905
137	585	111.699	78	141	0.199	2.191	0.456	0.382
138	285	103.884	78	141	0.792	1.760	0.568	0.903
139	144	102.799	78	139	0.671	1.753	0.570	0.778
140	852	112.445	78	141	0.110	2.606	0.384	0.267
141	745	108.917	78	141	0.308	1.898	0.527	0.526
142	44	108.432	85	138	0.996	1.438	0.696	0.926
143	122	92.705	78	125	0.615	1.370	0.730	0.797
144	180	99.850	78	125	0.886	1.537	0.651	0.918
145	47	109.830	85	140	0.993	1.032	0.969	0.887
146	197	101.208	78	138	0.870	1.298	0.771	0.914
147	43	110.047	85	139	0.974	1.169	0.856	0.896

Threshold

8.48 %

78

141

Default | Red

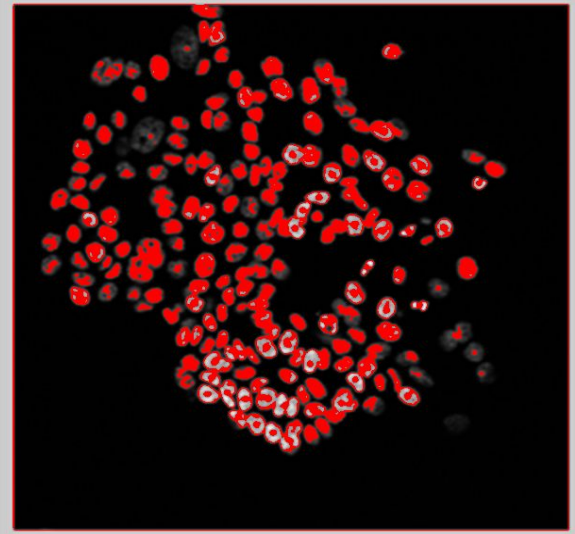
Dark background | Stack histogram

Don't reset range

Auto | Apply | Reset | Set

68647_wl_astellas_fig4_wl.jpeg

663x668 pixels; 8-bit; 433K



YouTube · Adam Wentworth

ImageJ

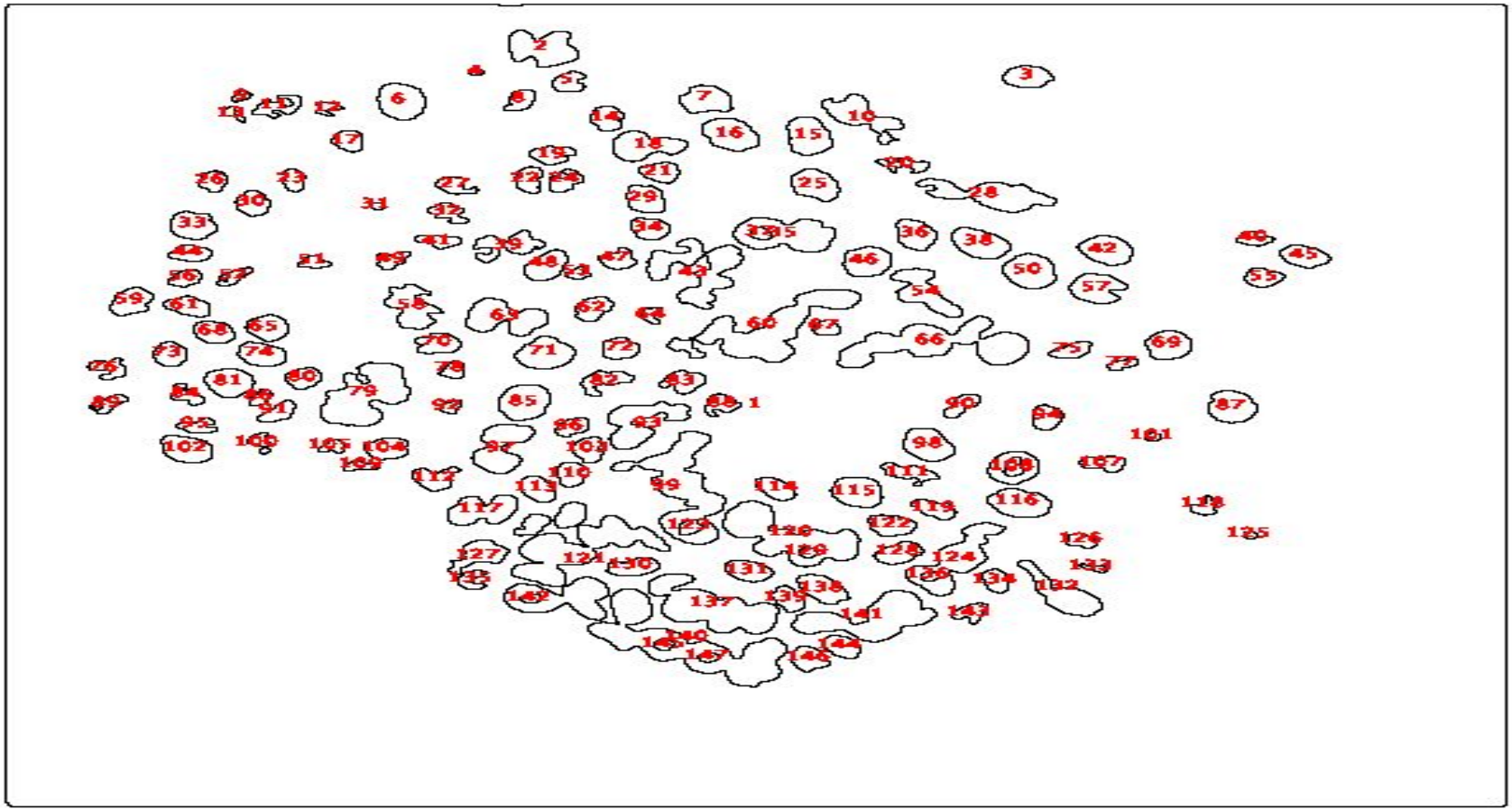
Call_pics 146 items

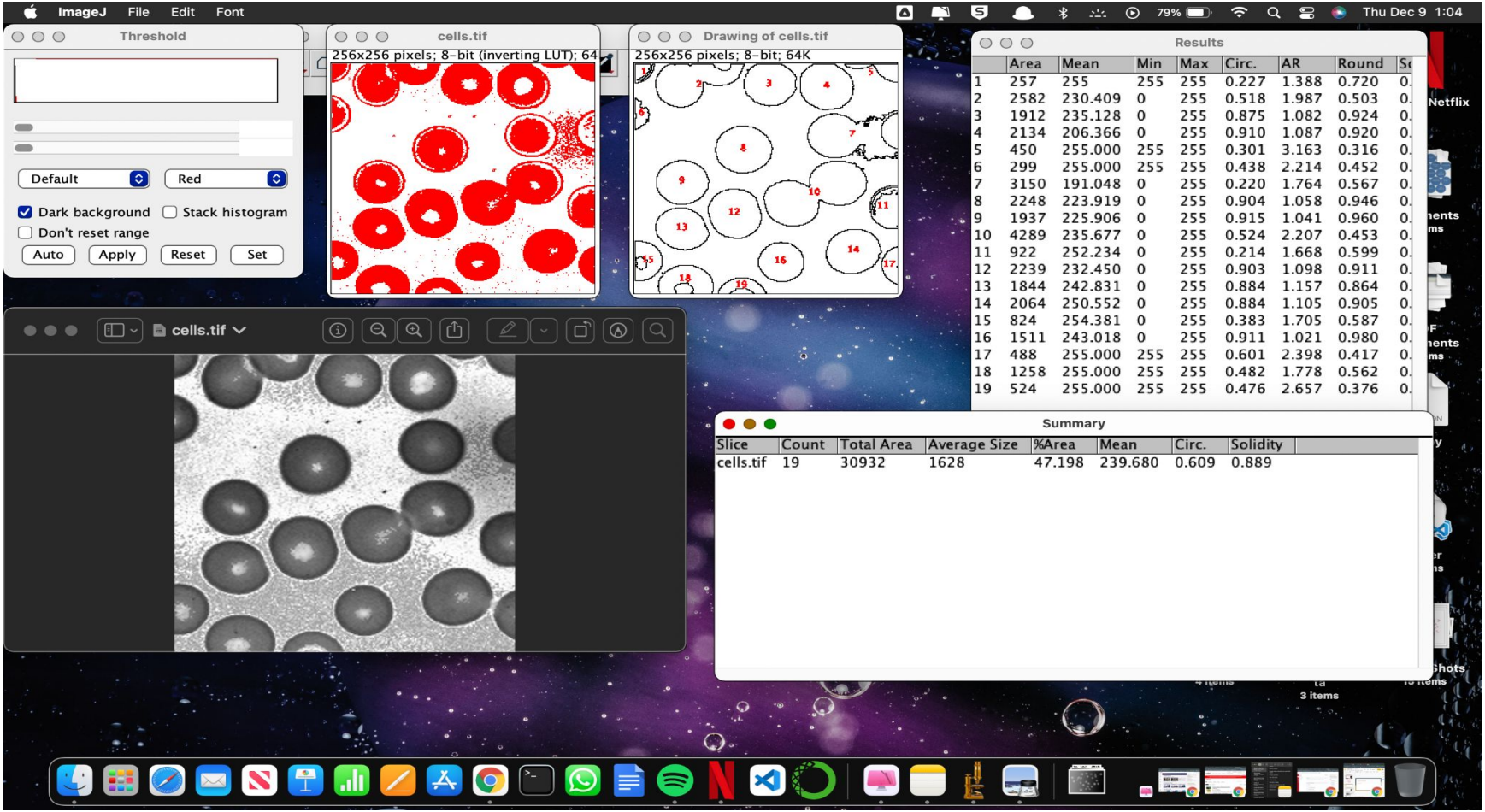
sports_data 7 items

1 item

Drawing of 68647_wl_astellas_fig4_wl.jpeg

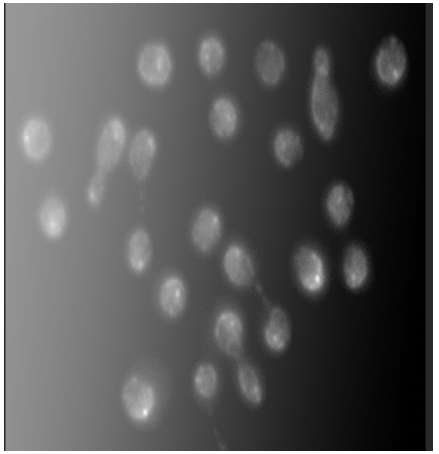
663x668 pixels; 8-bit; 433K



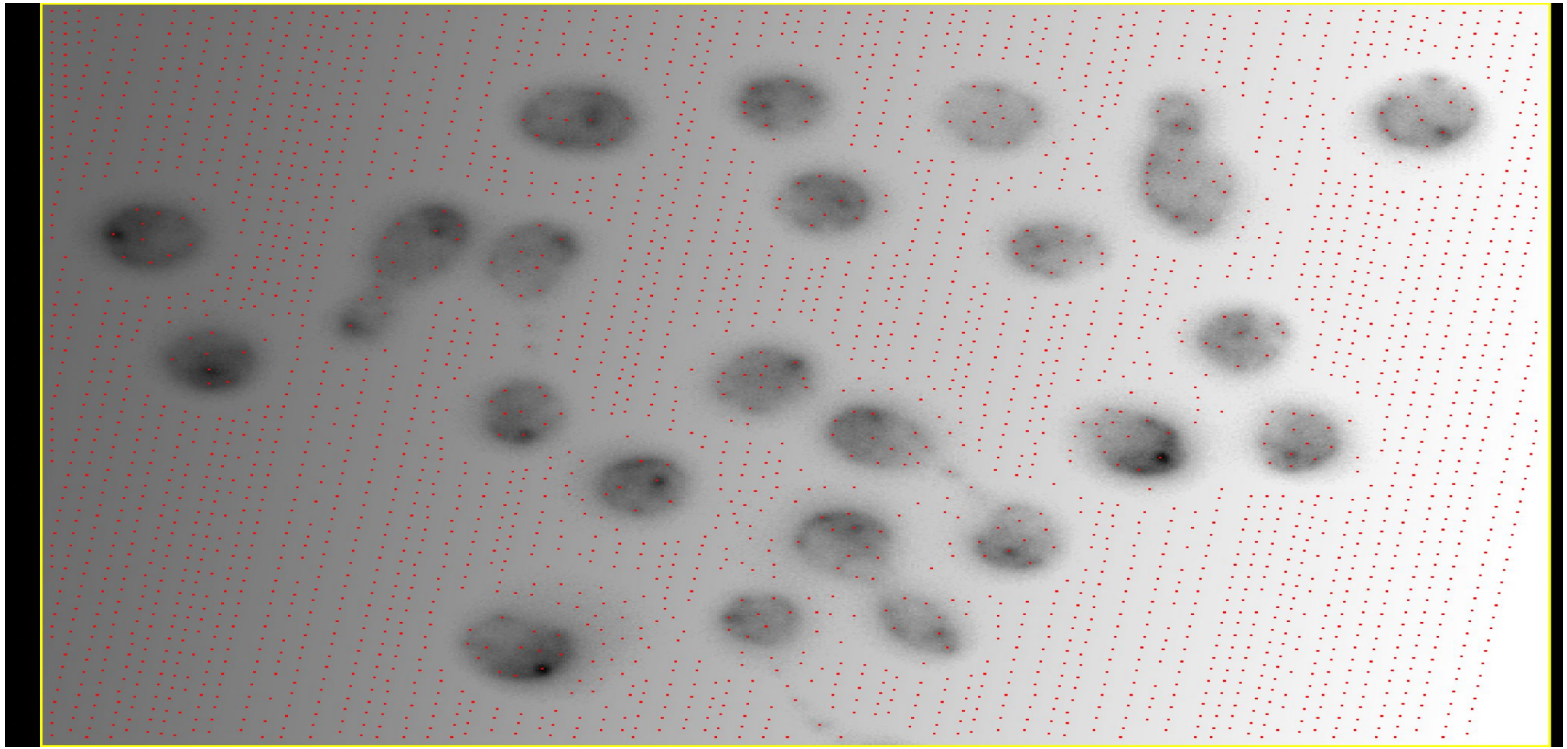

ImageJ File Edit Font
Thu Dec 9 1:04
Threshold
Default
Red
Dark background
Stack histogram
Don't reset range
Auto
Apply
Reset
Set
cells.tif
256x256 pixels; 8-bit (inverting LUT); 64
Drawing of cells.tif
256x256 pixels; 8-bit; 64K
cells.tif
Results
Area Mean Min Max Circ. AR Round
1 257 255 255 255 0.227 1.388 0.720
2 2582 230.409 0 255 0.518 1.987 0.503
3 1912 235.128 0 255 0.875 1.082 0.924
4 2134 206.366 0 255 0.910 1.087 0.920
5 450 255.000 255 255 0.301 3.163 0.316
6 299 255.000 255 255 0.438 2.214 0.452
7 3150 191.048 0 255 0.220 1.764 0.567
8 2248 223.919 0 255 0.904 1.058 0.946
9 1937 225.906 0 255 0.915 1.041 0.960
10 4289 235.677 0 255 0.524 2.207 0.453
11 922 252.234 0 255 0.214 1.668 0.599
12 2239 232.450 0 255 0.903 1.098 0.911
13 1844 242.831 0 255 0.884 1.157 0.864
14 2064 250.552 0 255 0.884 1.105 0.905
15 824 254.381 0 255 0.383 1.705 0.587
16 1511 243.018 0 255 0.911 1.021 0.980
17 488 255.000 255 255 0.601 2.398 0.417
18 1258 255.000 255 255 0.482 1.778 0.562
19 524 255.000 255 255 0.476 2.657 0.376
Summary
Slice Count Total Area Average Size %Area Mean Circ. Solidity
cells.tif 19 30932 1628 47.198 239.680 0.609 0.889
Netflix
3 items

Cell counting

Inverted the image then converted to 8 bit binary before applying ITCN.



ITCN is an ImageJ plugin for automatically counting the number cells within an image. The inputs are: (1) an estimation of the diameter of a cell, (2) an estimation of the minimum distance between cells



Thresholding

Threshold technique is one of the important techniques in image segmentation. This technique can be expressed as:

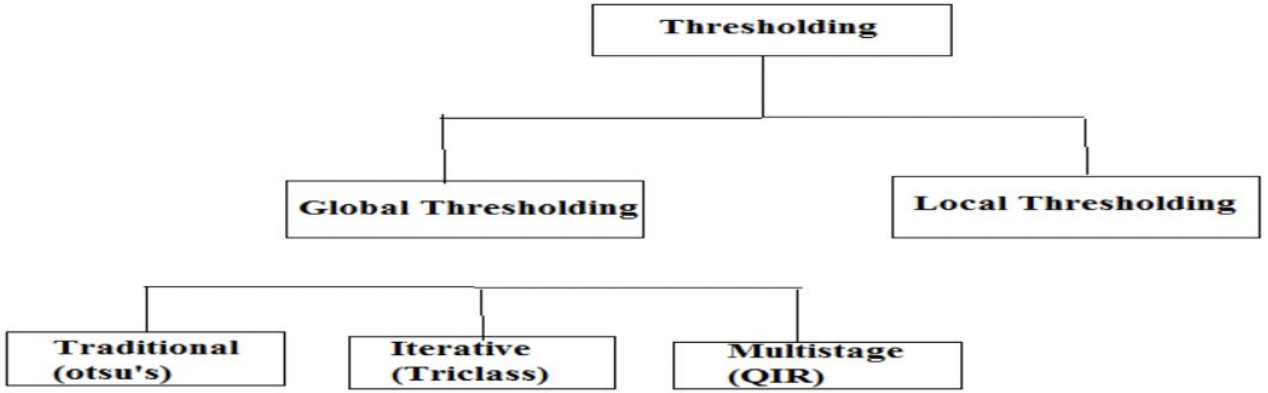
$$T=T[x, y, p(x, y), f(x, y]$$

Where T is the threshold value. x, y are the coordinates of the threshold value point.p(x,y) ,f(x,y) are points the gray level image pixels [9].Threshold image g(x,y) can be define:

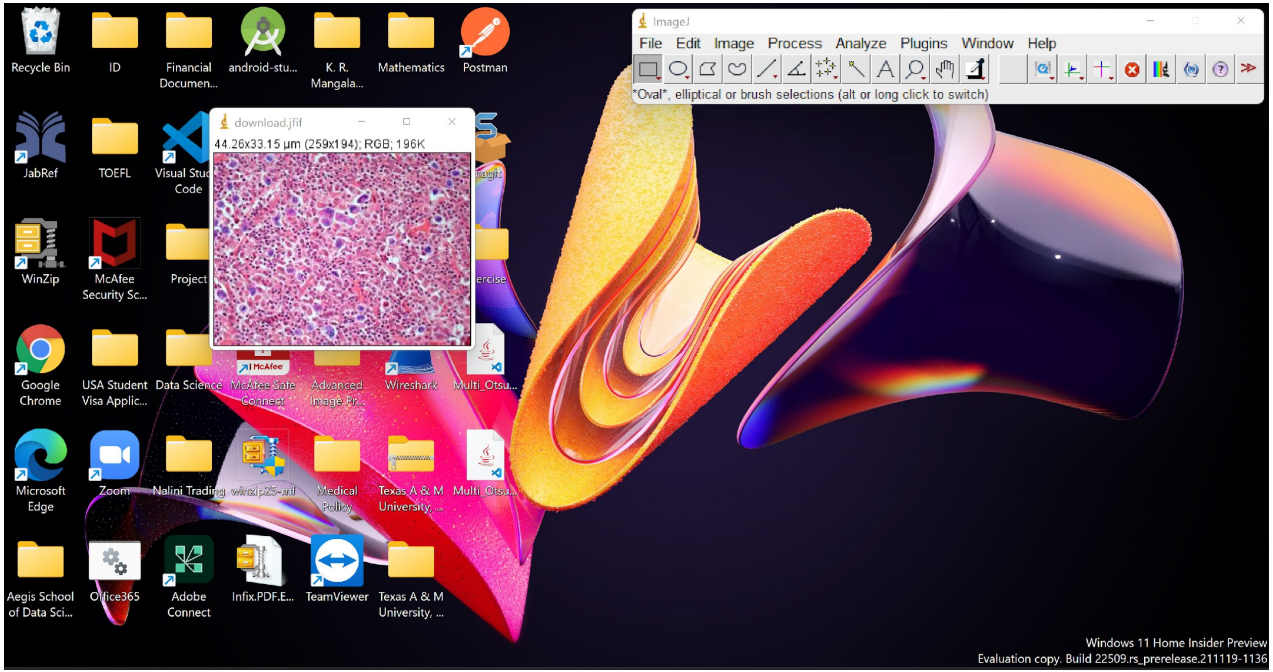
$$g(x,y)=\begin{cases} 1 & if\ f(x,y) > 1 \\ 0 & if\ f(x,y) \leq 0 \end{cases}$$

TYPES OF THRESHOLDING

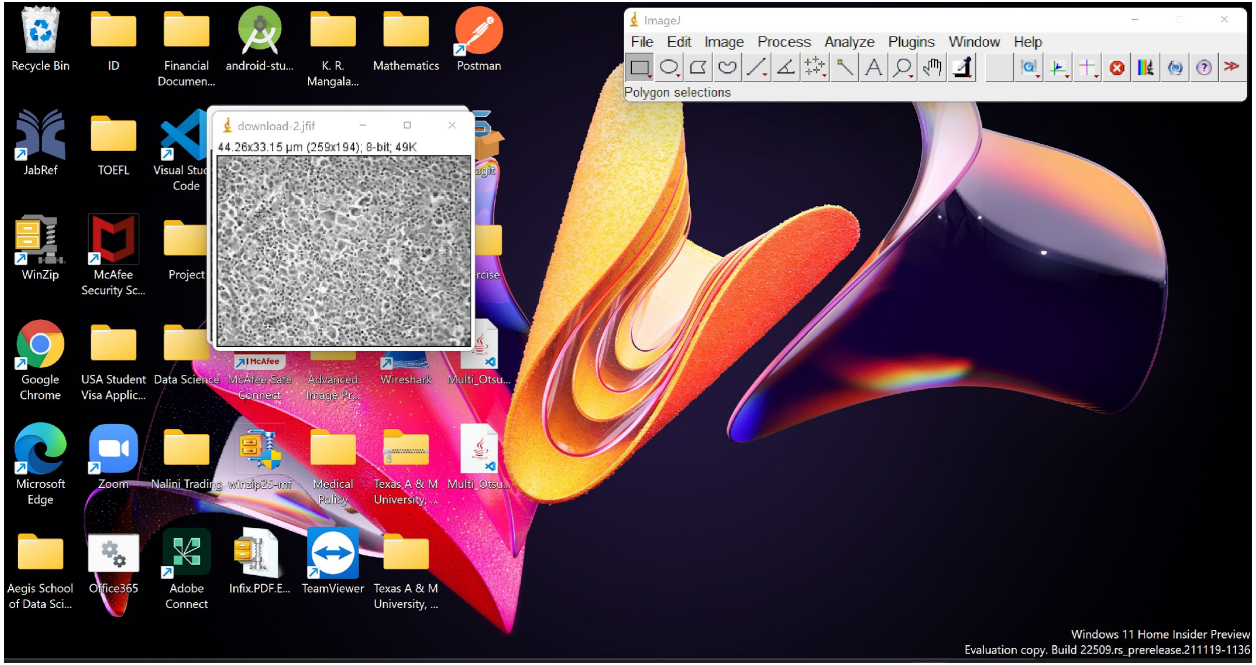
Thresholding techniques are classified as bellow



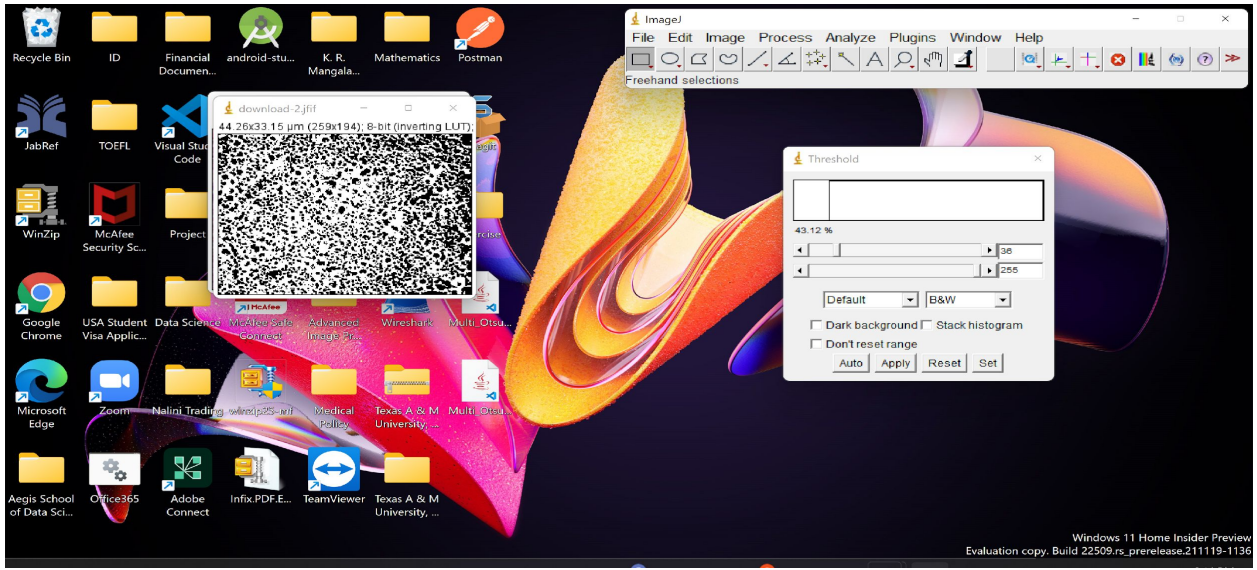
EXAMPLE FOR THRESHOLDING



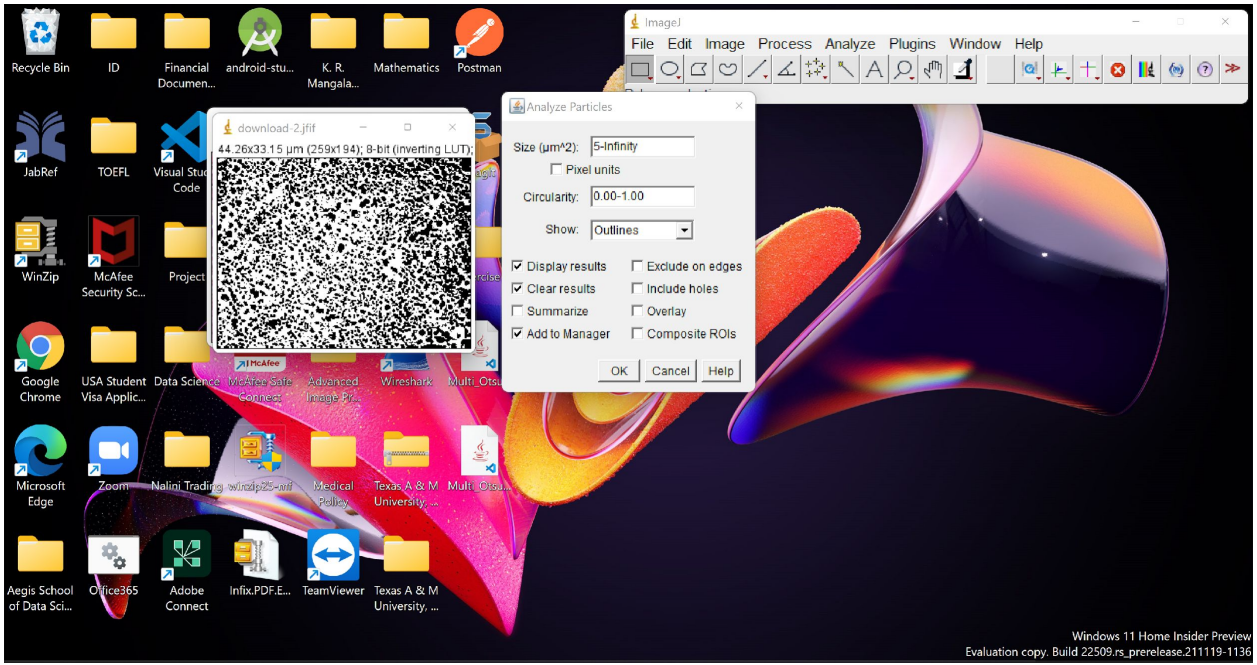
8-BIT IMAGE


Recycle Bin
ID
Financial Documen...
android-stu...
K. R. Mangala...
Mathematics
Postman
JabRef
TOEFL
Visual Stud... Code
WinZip
McAfee Security Sc...
Project
Google Chrome
USA Student Visa Applic...
Data Science
McAfee Safe Connect
Advanced Image Pr...
Wireshark
Multi_Otsu...
Microsoft Edge
Zoom
Nalini Trading
Medical Policy
Texas A & M University, ...
Multi_Otsu...
Aegis School of Data Sci...
Office365
Adobe Connect
Infix PDF E...
TeamViewer
Texas A & M University, ...
download-2.jfif
44.26x33.15 µm (259x194); 8-bit; 49K
ImageJ
File Edit Image Process Analyze Plugins Window Help
Polygon selections
Windows 11 Home Insider Preview
Evaluation copy. Build 22509.rs_prerelease.211119-1136

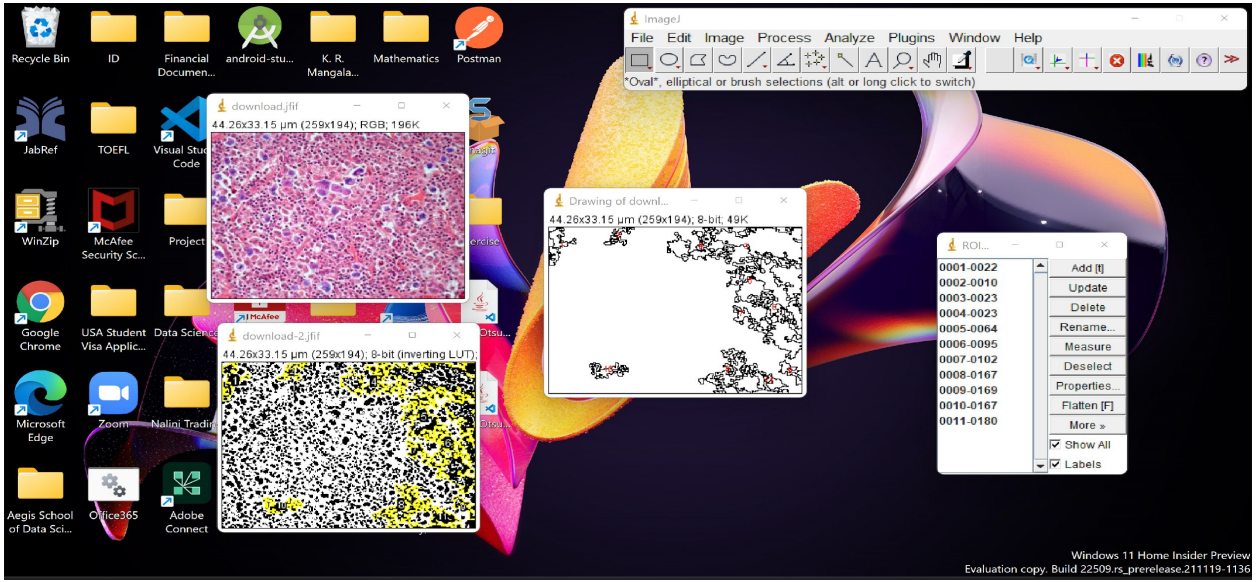
APPLYING THRESHOLDING



APPLYING ANALYZE PRATICLES



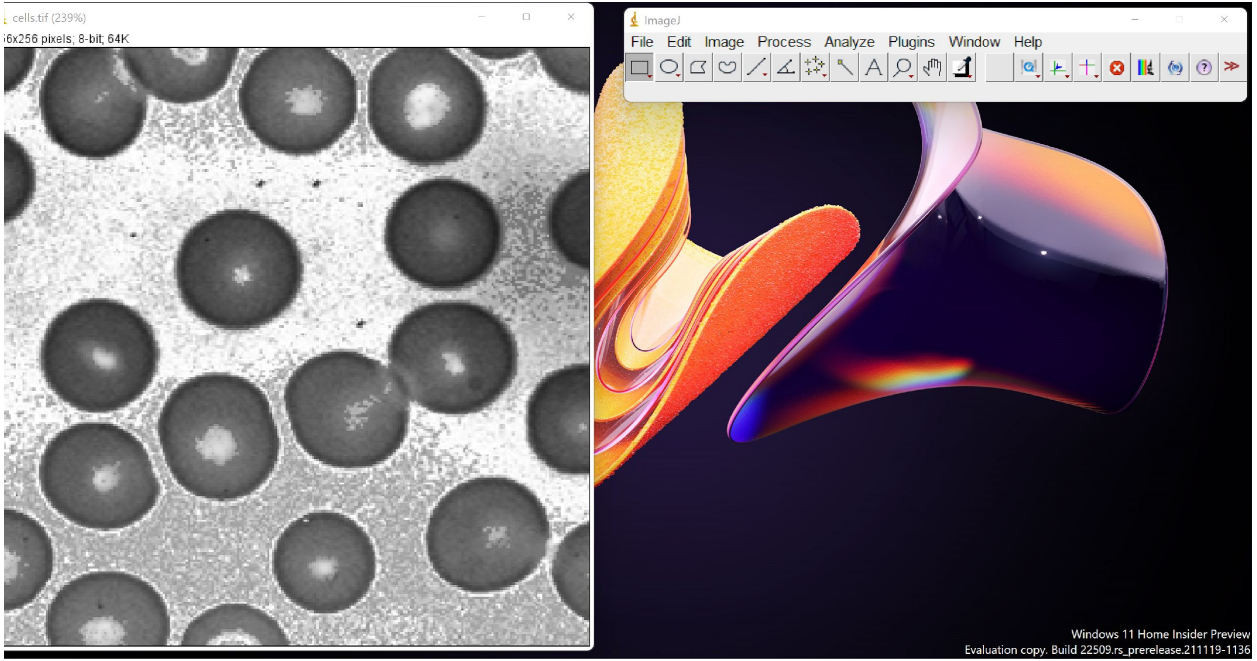
OUTPUT FOR THRESHOLDING



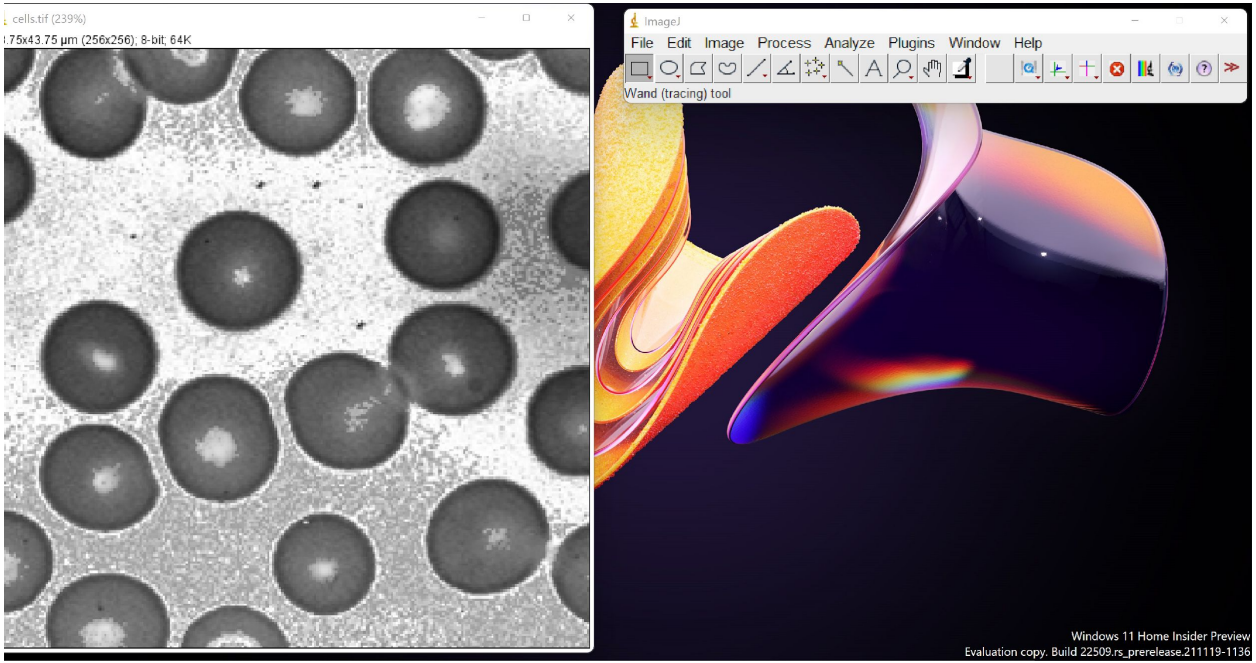
MULTI-THRESHOLDING

Multi-thresholding approach generalizes the image thresholding by finding multiple thresholds which aim to separate multiple objects.

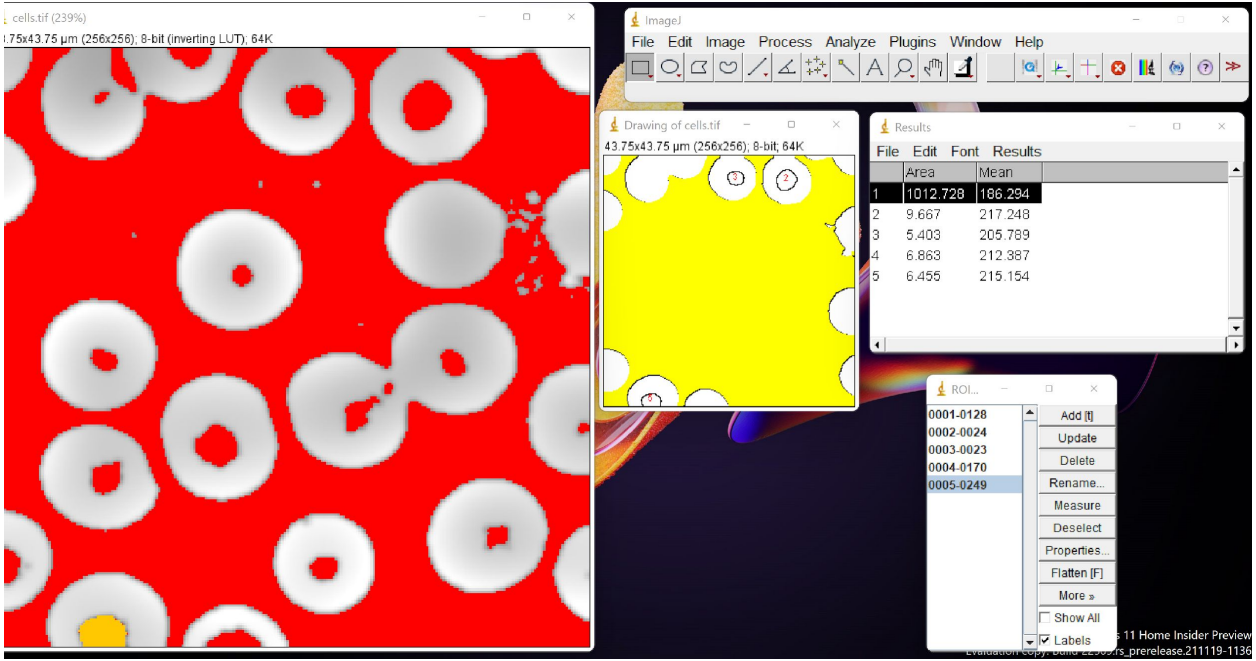
EXAMPLE OF MULTI-THRESHOLDING



APPLYING SET SCALE



APPLYING MULTI-THRESHOLDING

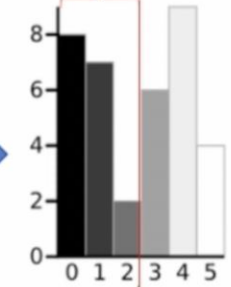


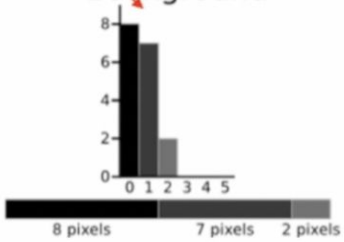
Otsu Approach

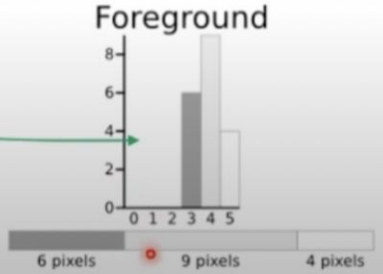
Otsu’s method: means further extension of mean based method.

Otsu divides into classes it maximize inter class variance and minimize with in class variance

Otsu's approach (driven by statistical-decision theory) is optimum in the sense that it minimizes the in-class variance or maximizes the between class-variance, where a class means a set of pixels belonging to a region.







$$\text{Weight} \quad W_b = \frac{8+7+2}{36} = 0.4722$$

$$\text{Mean} \quad \mu_b = \frac{(0\times 8)+(1\times 7)+(2\times 2)}{17} = 0.6471$$

$$\text{Variance} \quad \sigma_b^2 = \frac{((0-0.6471)^2\times 8)+((1-0.6471)^2\times 7)+((2-0.6471)^2\times 2)}{17}$$

$$= \frac{(0.4187\times 8)+(0.1246\times 7)+(1.8304\times 2)}{17}$$

$$= 0.4637$$

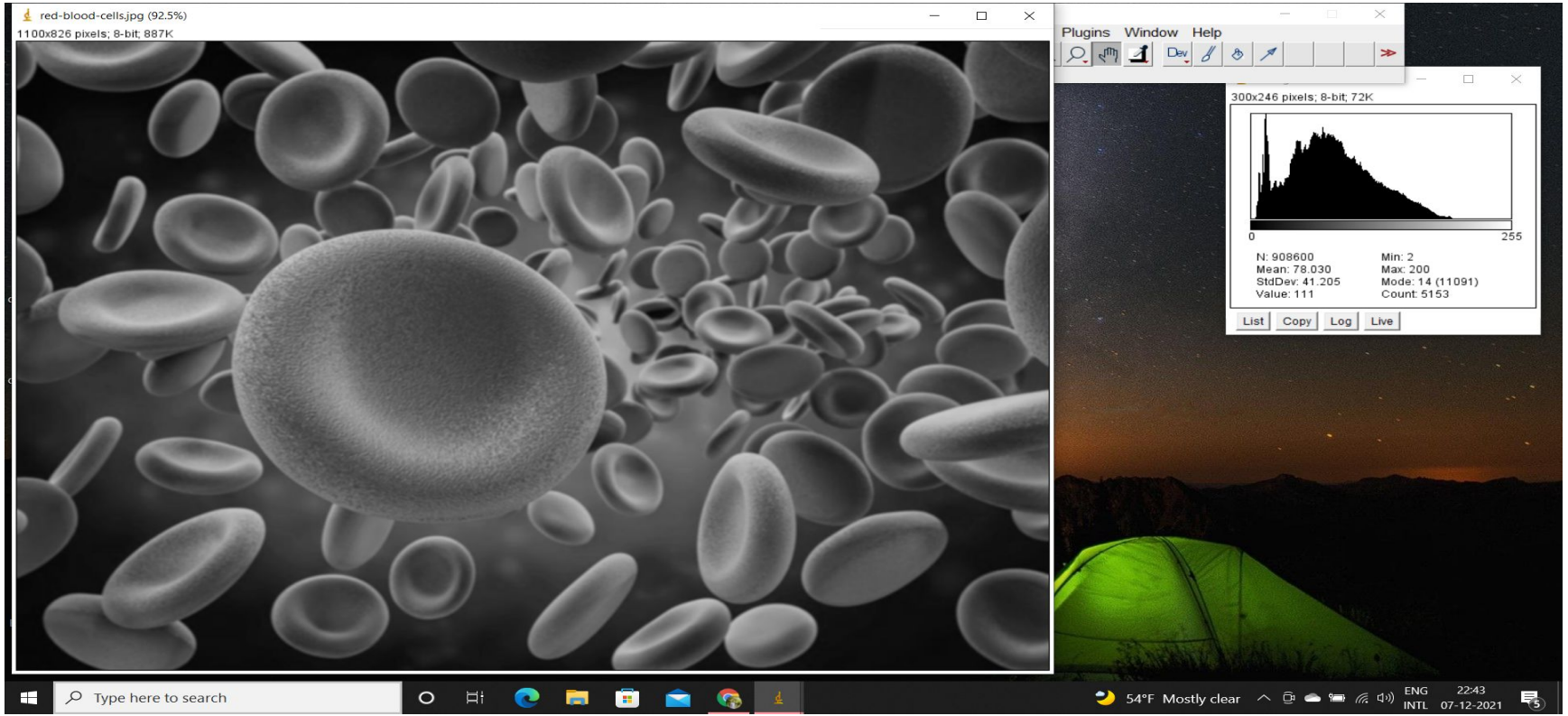
$$\text{Weight} \quad W_f = \frac{6+9+4}{36} = 0.5278$$

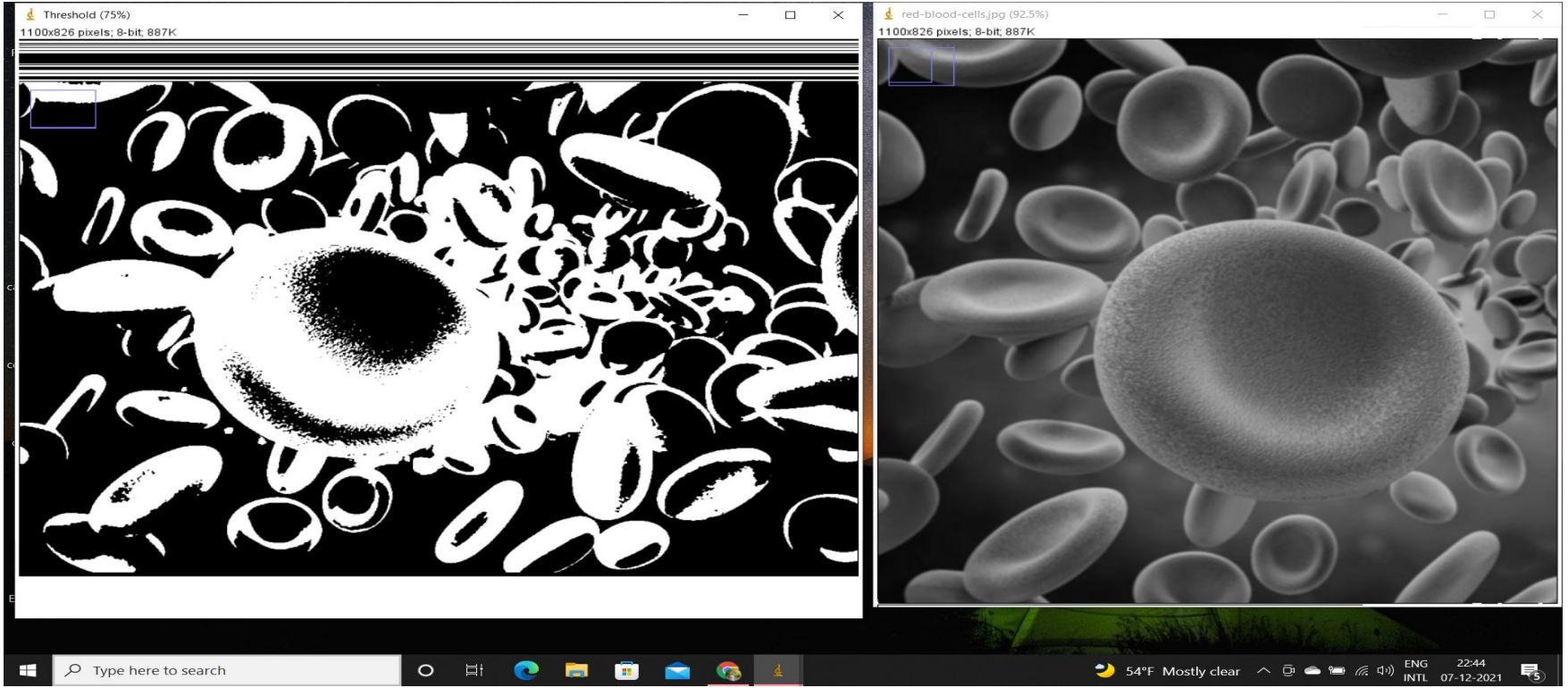
$$\text{Mean} \quad \mu_f = \frac{(3\times 6)+(4\times 9)+(5\times 4)}{19} = 3.8947$$

$$\text{Variance} \quad \sigma_f^2 = \frac{((3-3.8947)^2\times 6)+((4-3.8947)^2\times 9)+((5-3.8947)^2\times 4)}{19}$$

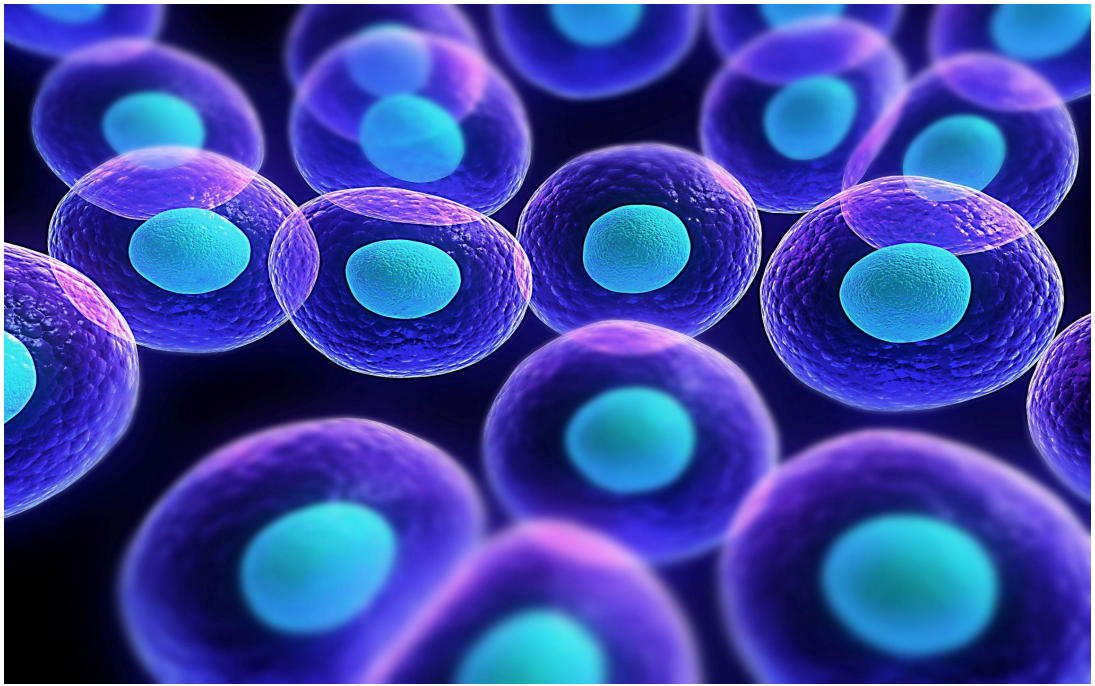
$$= \frac{(4.8033\times 6)+(0.0997\times 9)+(4.8864\times 4)}{19}$$

$$= 0.5152$$


red-blood-cells.jpg (92.5%)
1100x826 pixels; 8-bit; 887K
Plugins
Window
Help
300x246 pixels; 8-bit; 72K
0
255
N: 908600
Mean: 78.030
StdDev: 41.205
Value: 111
Min: 2
Max: 200
Mode: 14 (11091)
Count: 5153
List
Copy
Log
Live
Type here to search
54°F Mostly clear
ENG
INTL
22:43
07-12-2021

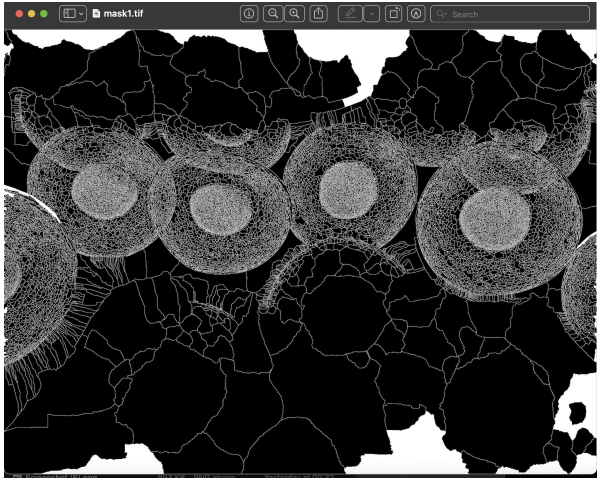

Threshold (75%)
1100x826 pixels; 8-bit; 887K
red-blood-cells.jpg (92.5%)
1100x826 pixels; 8-bit; 887K
Type here to search
54°F Mostly clear
ENG
INTL
22:44
07-12-2021

Image



Cell segmentation

Cell Segmentation is a task of splitting a microscopic image domain into segments, which represent individual instances of cells.



First duplicated the image

Applied find maxima output as segmented particles(Which uses watershed concept) (mask1)

Duplicated image then applied thresholding

Then smoothing

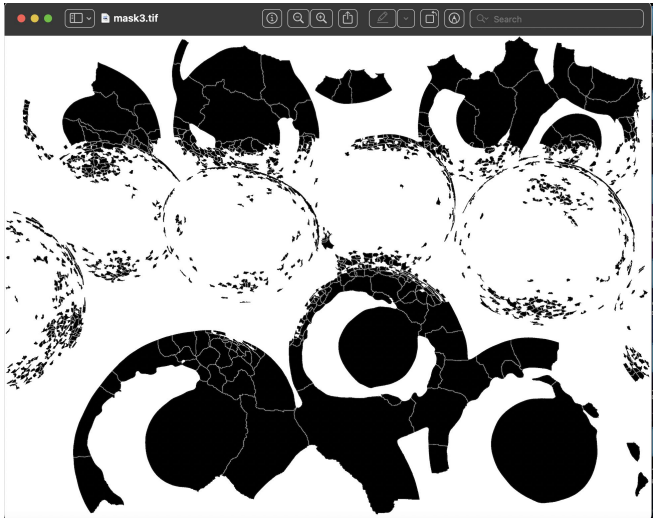
Saved as mask2

Image calculator

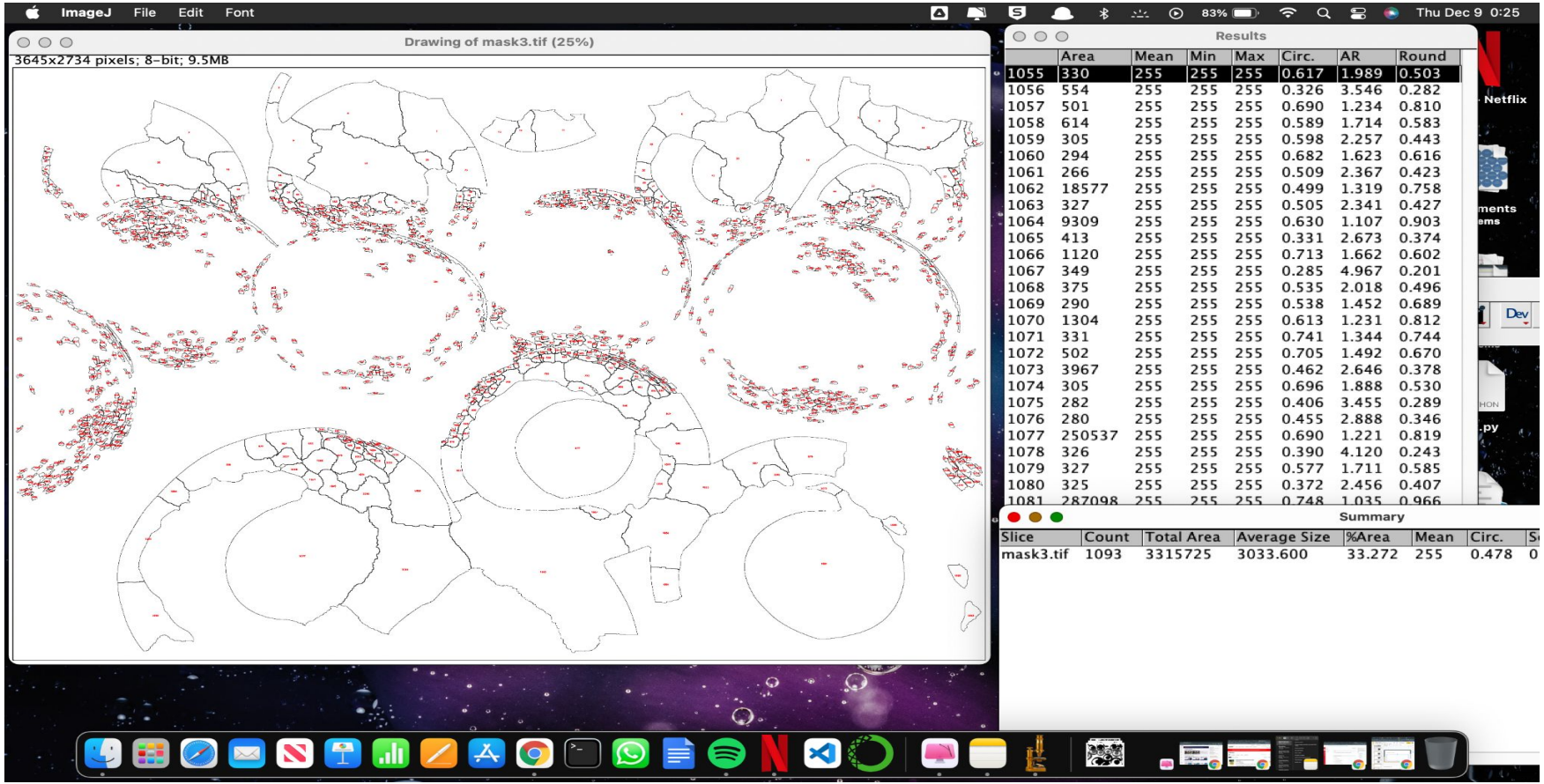
Now i used image calculator using mask1 AND mask2

Then Particle analysis .

Applied Fill holes concept



This command fills holes (4--connected background elements) in objects by filling the background

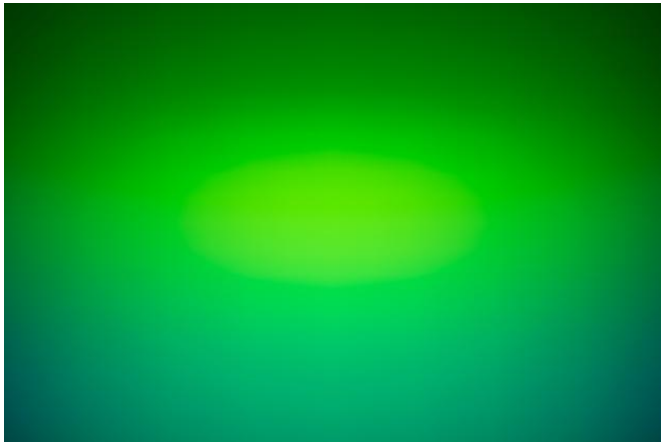


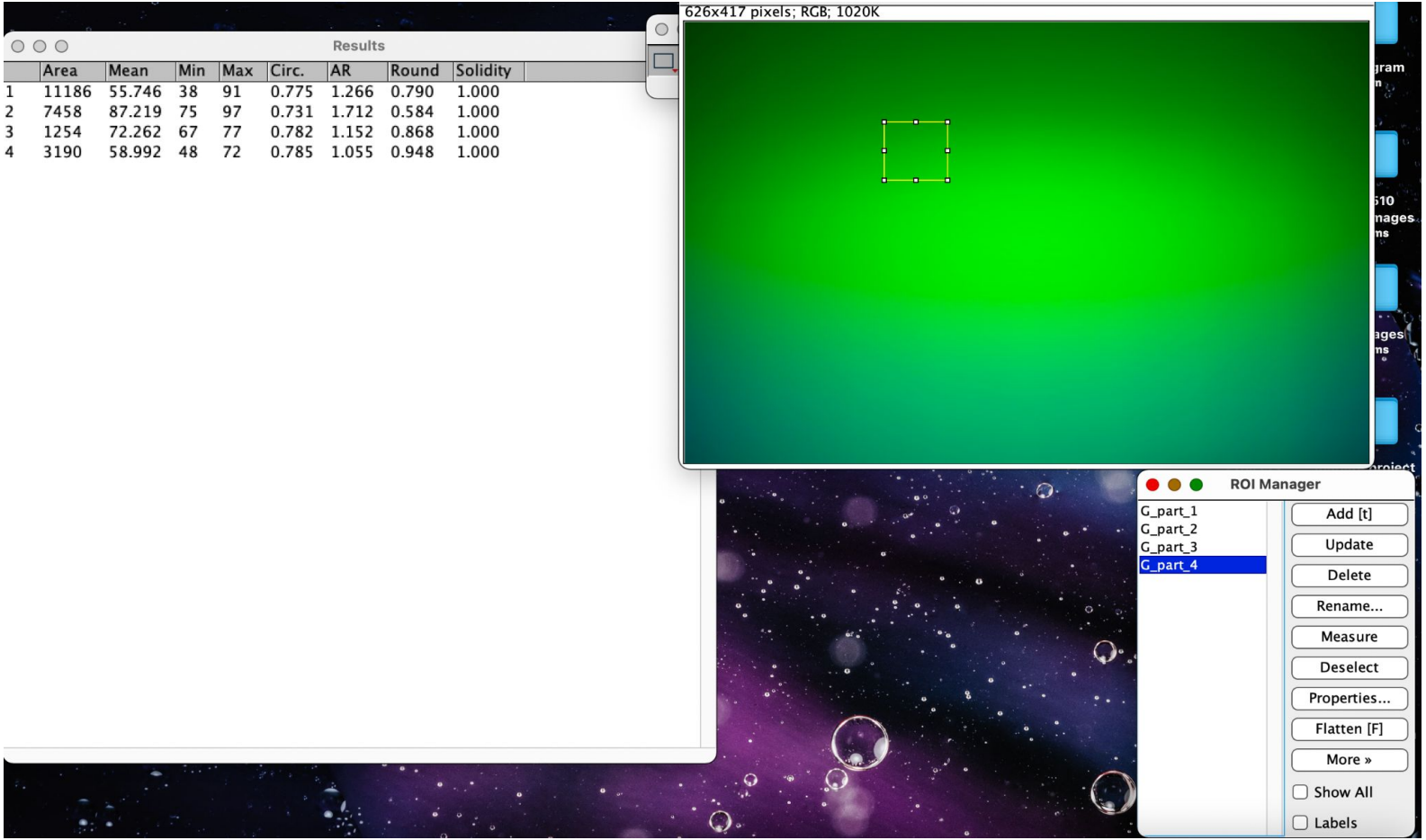
	Area	Mean	Min	Max	Circ.	AR	Round
1055	330	255	255	255	0.617	1.989	0.503
1056	554	255	255	255	0.326	3.546	0.282
1057	501	255	255	255	0.690	1.234	0.810
1058	614	255	255	255	0.589	1.714	0.583
1059	305	255	255	255	0.598	2.257	0.443
1060	294	255	255	255	0.682	1.623	0.616
1061	266	255	255	255	0.509	2.367	0.423
1062	18577	255	255	255	0.499	1.319	0.758
1063	327	255	255	255	0.505	2.341	0.427
1064	9309	255	255	255	0.630	1.107	0.903
1065	413	255	255	255	0.331	2.673	0.374
1066	1120	255	255	255	0.713	1.662	0.602
1067	349	255	255	255	0.285	4.967	0.201
1068	375	255	255	255	0.535	2.018	0.496
1069	290	255	255	255	0.538	1.452	0.689
1070	1304	255	255	255	0.613	1.231	0.812
1071	331	255	255	255	0.741	1.344	0.744
1072	502	255	255	255	0.705	1.492	0.670
1073	3967	255	255	255	0.462	2.646	0.378
1074	305	255	255	255	0.696	1.888	0.530
1075	282	255	255	255	0.406	3.455	0.289
1076	280	255	255	255	0.455	2.888	0.346
1077	250537	255	255	255	0.690	1.221	0.819
1078	326	255	255	255	0.390	4.120	0.243
1079	327	255	255	255	0.577	1.711	0.585
1080	325	255	255	255	0.372	2.456	0.407
1081	287098	255	255	255	0.748	1.035	0.966

Slice	Count	Total Area	Average Size	%Area	Mean	Circ.	S
mask3.tif	1093	3315725	3033.600	33.272	255	0.478	0

Measuring intensity

Using ROI option measured the mean intensity, Area, min, max and mode in below image in 4 different selections in image.




Results
Area Mean Min Max Circ. AR Round Solidity
1 11186 55.746 38 91 0.775 1.266 0.790 1.000
2 7458 87.219 75 97 0.731 1.712 0.584 1.000
3 1254 72.262 67 77 0.782 1.152 0.868 1.000
4 3190 58.992 48 72 0.785 1.055 0.948 1.000
626x417 pixels; RGB; 1020K
ROI Manager
G_part_1
G_part_2
G_part_3
G_part_4
Add [t]
Update
Delete
Rename...
Measure
Deselect
Properties...
Flatten [F]
More »
Show All
Labels

References

https://www.researchgate.net/publication/339738360_On_the_Use_of_ImageJ_Segmentation/link/5e61f1aa92851c7d6f25aedd/download

https://www.youtube.com/watch?v=PqHFsmS1_JY

https://www.youtube.com/watch?v=BrNtHKDbTk0

https://www.youtube.com/watch?v=82N-eIPqnwM

https://www.youtube.com/watch?v=5RlHyhWSqqI

https://imagej.net/imaging/particle-analysis

https://imagej.nih.gov/ij/plugins/itcn.html

https://www.youtube.com/watch?v=_Ku2yXd4hc

THANK YOU....!!!