CellSeg2

Main Algorithmic Steps

- Identify Background
- Identify Points within Cells
- Segment Cells

Identify Background

Why?

- Needed for robustness against artefacts / etc.

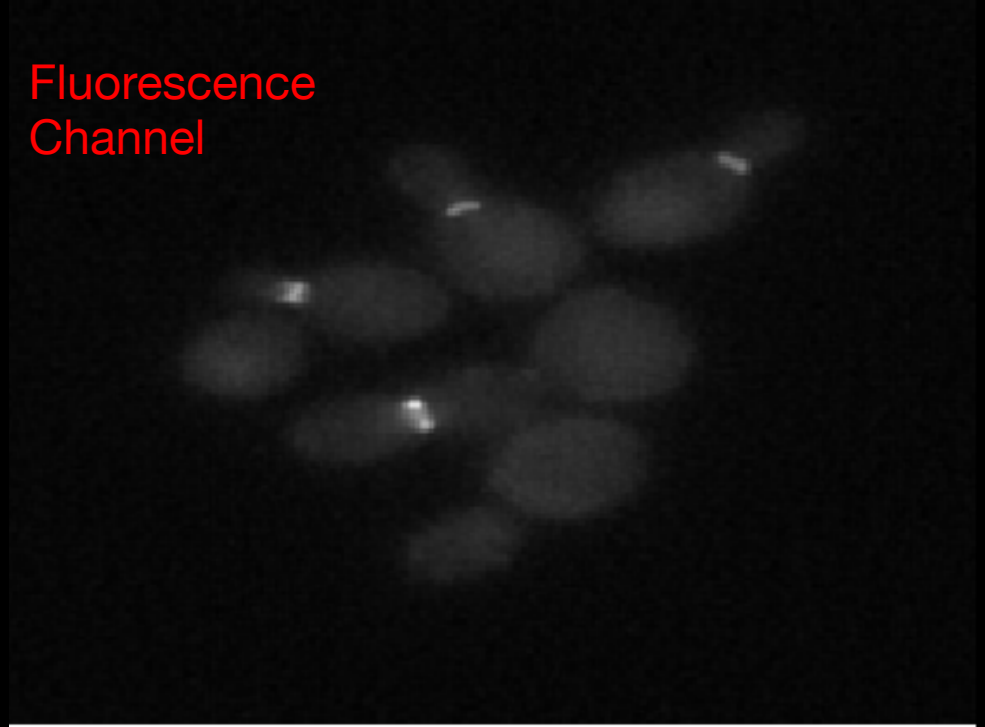
How?

- Based on fluorescence channel with high background fluorescence of cells
- Blur image with high radius
- Otsu thresholding
- Dilate thresholded image
- Invert

Otsu Thresholding

- Divide Pixels into foreground and background
- Create histogram of Pixel intensities, which is assumed to be bimodal
- Finds threshold in “valley” between two modes, by minimizing the variance within the two clusters

Fluorescence Channel



Otsu thresholding

Dilated, inverted

Point in Cell Identification

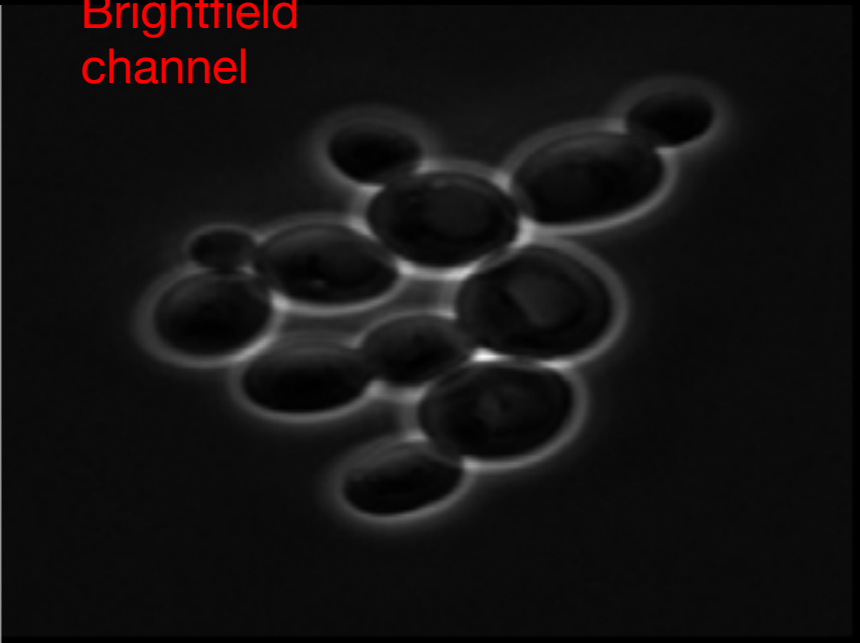
Why?

- Used for segmentation later on

How?

- Based on (blurred) brightfield image
- Otsu thresholding, series of binary image transformation to give good approximation of where the cell borders are
- Calculate distance transform
- Find maxima of smoothed distance transform that aren’t within background

Brightfield channel



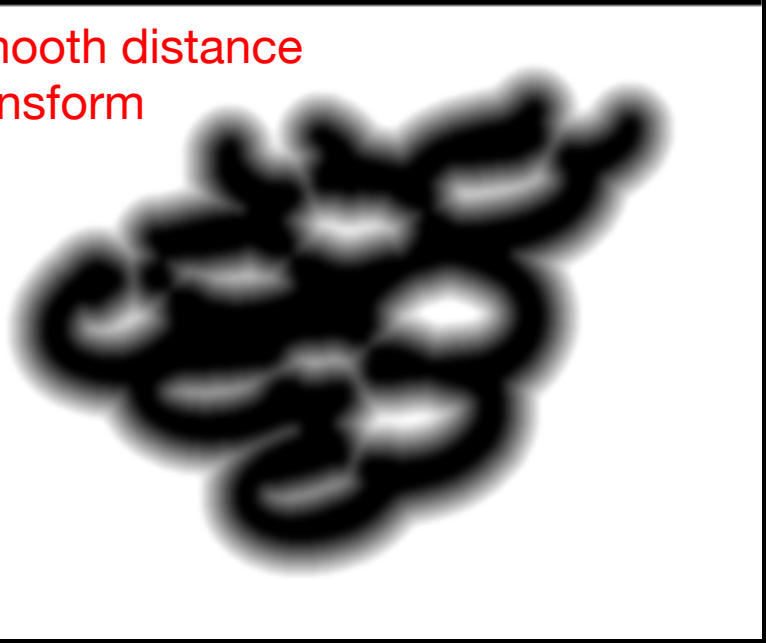
Otsu thresholding

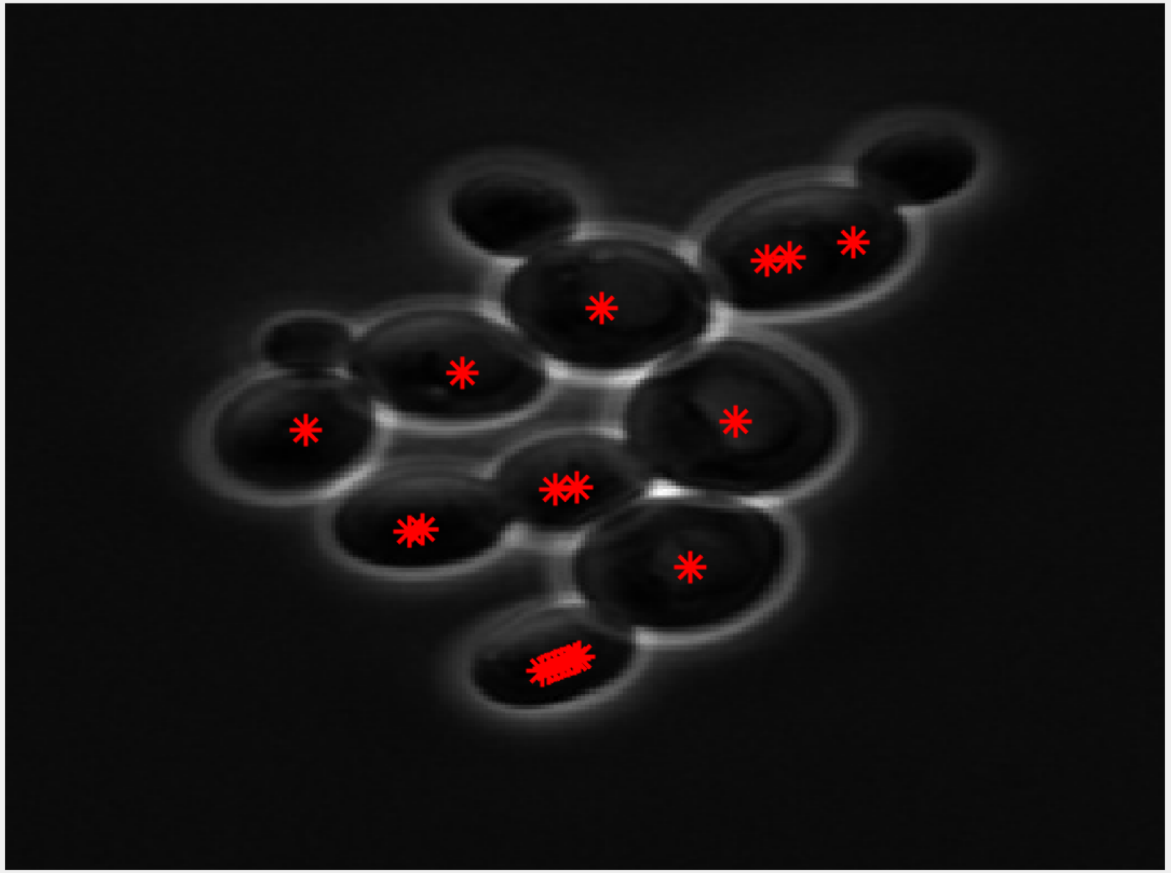
Erode image, remove noise

Dilate, make border area larger

Close, better handling at cell connections

Smooth distance transform





Pan | Zoom

Frame Selection

Region 1
Z 1
Timeframe 1
Channel 3
Position Nbr 1

Point Detection

Add Point
Remove Point
☑ Show Points

Plot Fluorescence

Plot Selected

Segmentation

Segment
Add to Region
Merge Regions
☐ Show Cell Numbers

Labeling

Redo Labels
Change Label
New Cell in Area

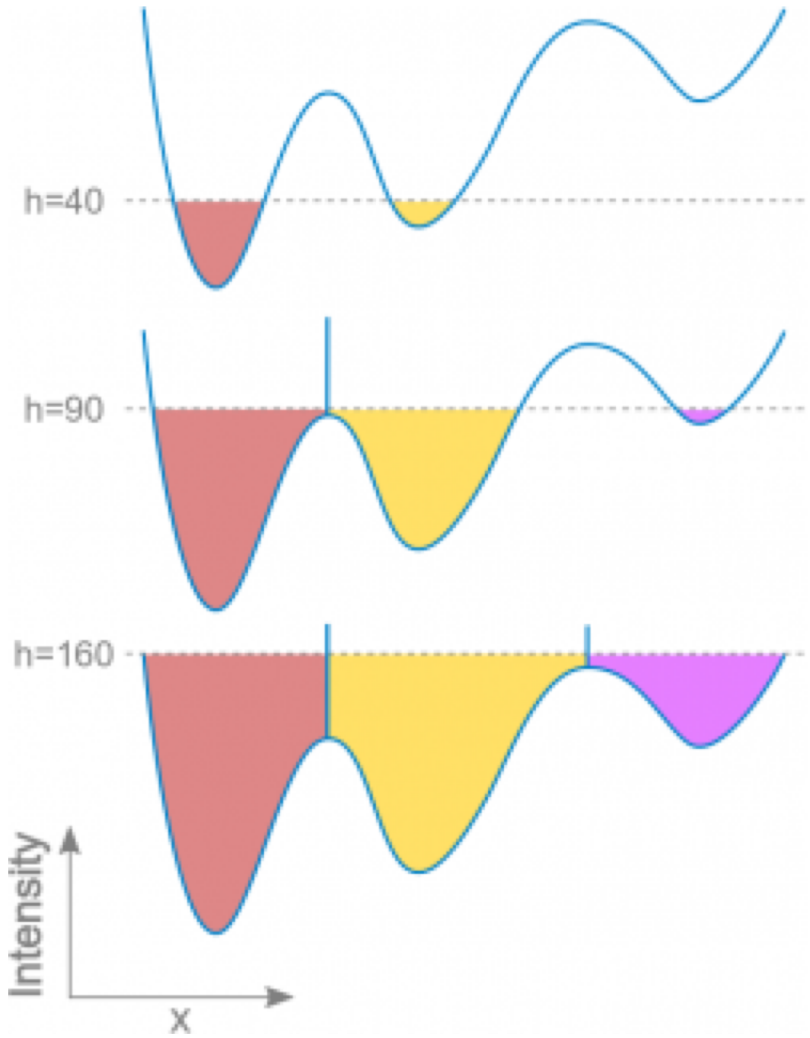
Segment Cells

Why?

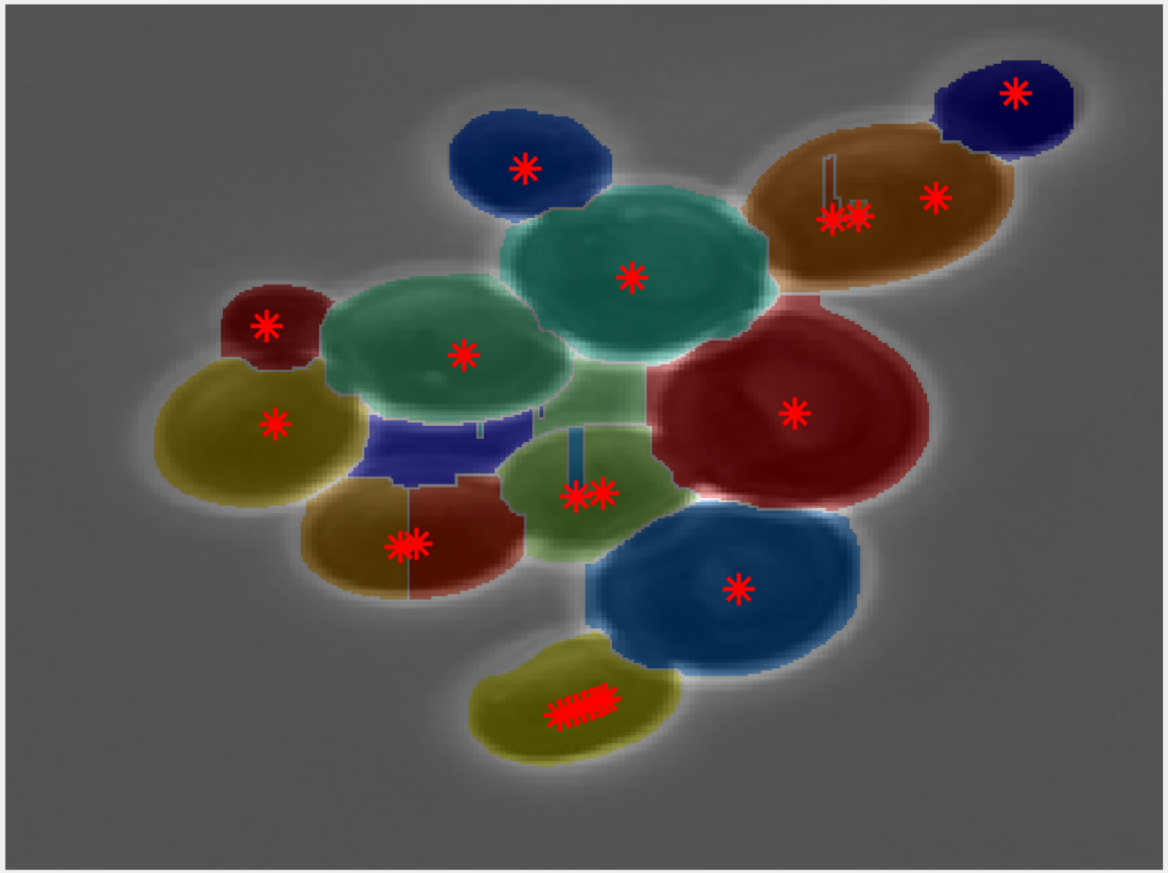
- For every pixel, assign it to a cell

How?

- Watershed algorithm
- Start at identified cell points and identified background
- Look at the image intensity as a landscape. Start filling this landscape up with water at starting points. If two lakes meet, identify this as the border of the cell.



(image from ImageJ)



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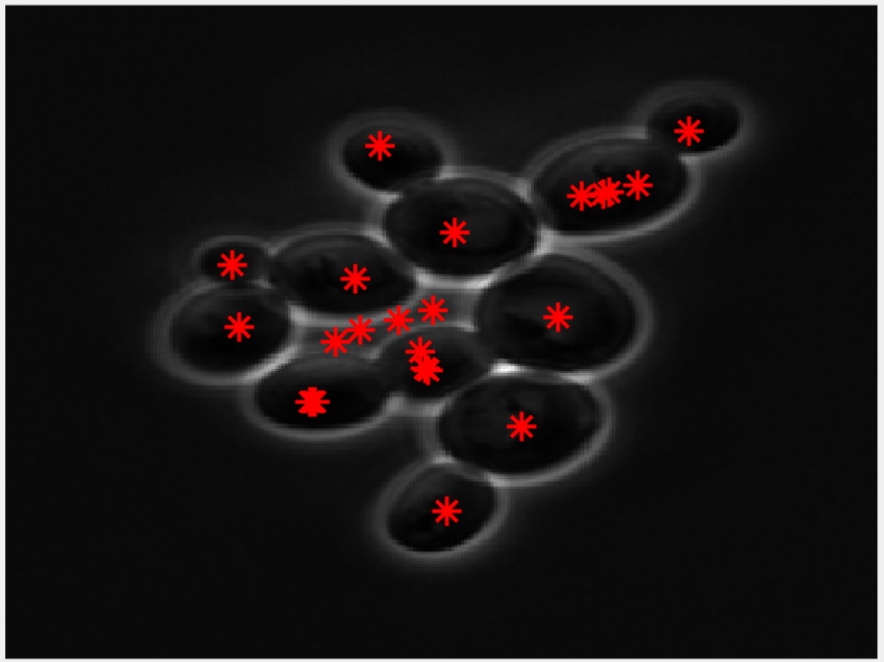
Change Label

New Cell in Area

Learn from Previous Frame

Two key improvements in the next frame:

- If a cell in the previous frame doesn’t have a point within it in the next cell, add the centroid as point
- After segmentation look at previous segmentation to identify cell correspondence.



Pan Zoom

Frame Selection

Region 1
Z 1
Timeframe 2
Channel 3
Position Nbr 1

Point Detection

Add Point
Remove Point
☑ Show Points

Plot Fluorescence

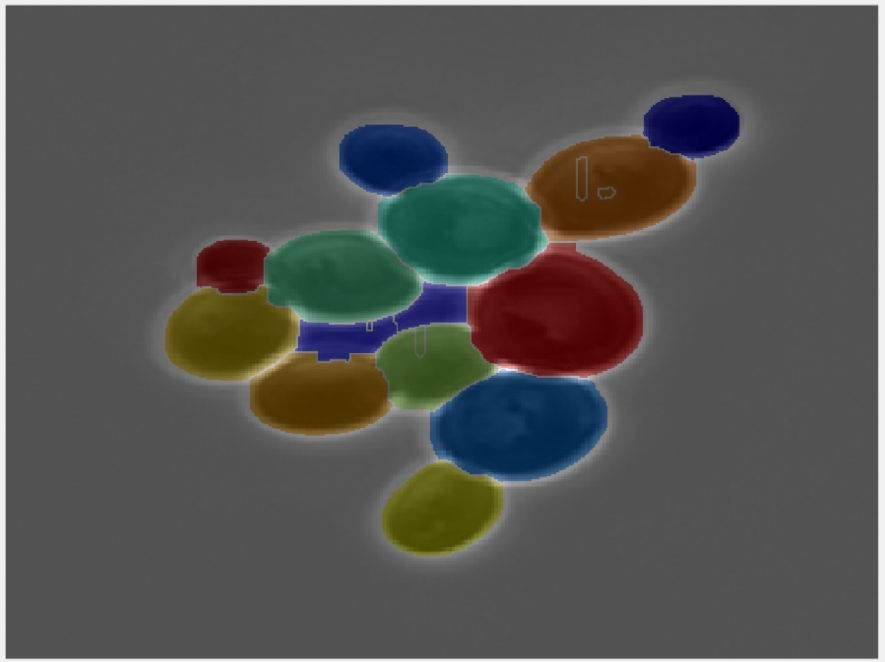
Plot Selected

Segmentation

Segment
Add to Region
Merge Regions
☐ Show Cell Numbers

Labeling

Redo Labels
Change Label
New Cell in Area



Pan Zoom

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