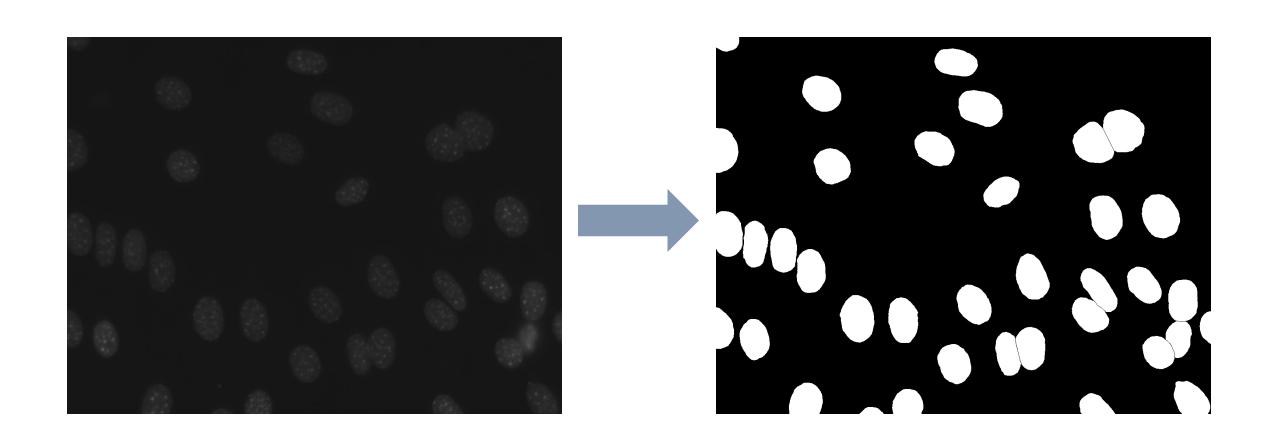
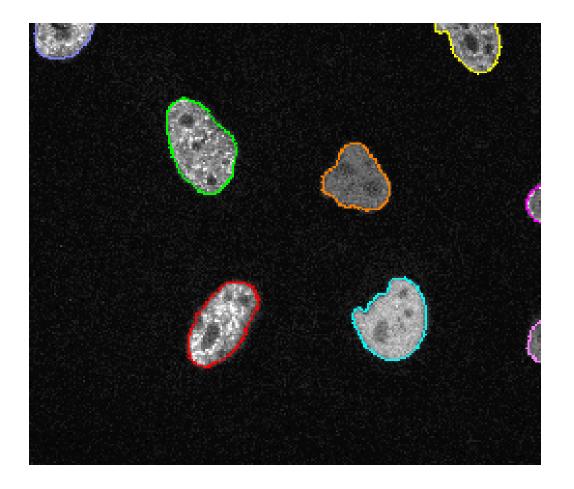
Image segmentation



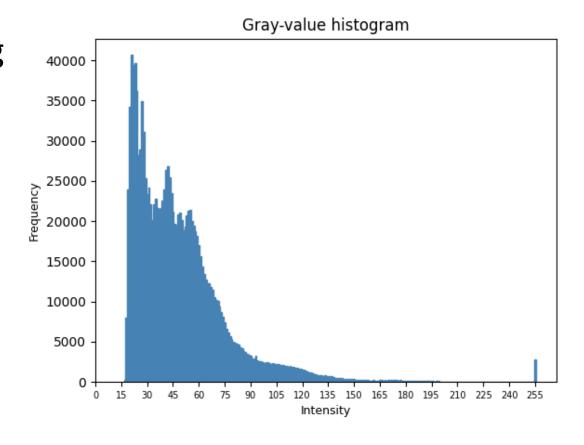
Applications

- High-throughput cytometry
 - Cell size
 - Cell counting
 - Cell-cycle determination
- Cell tracking



Methods

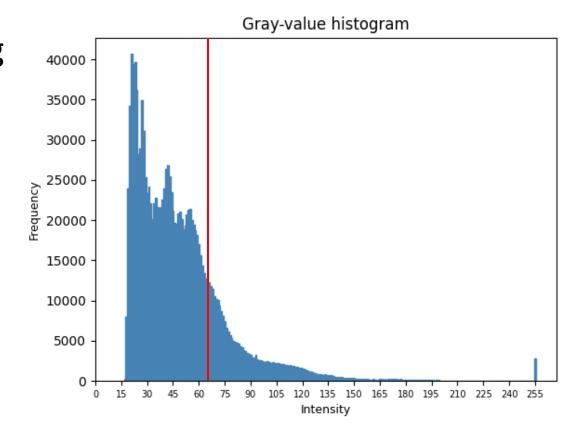
- Thresholding → Intensity clipping
- Region growing
- Machine learning
- etc.



Methods

Thresholding → Intensity clipping

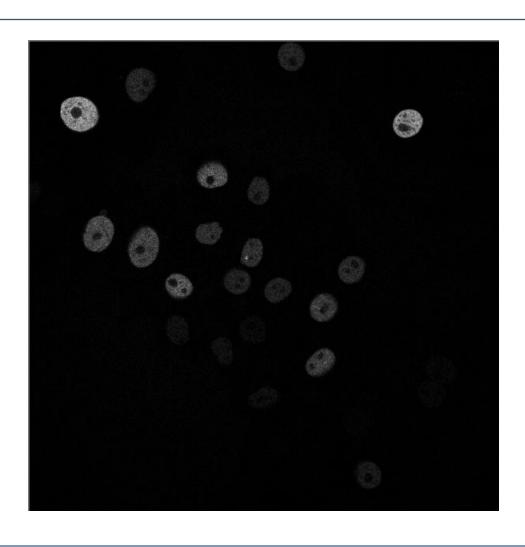
Otsu thresholding



Procedure

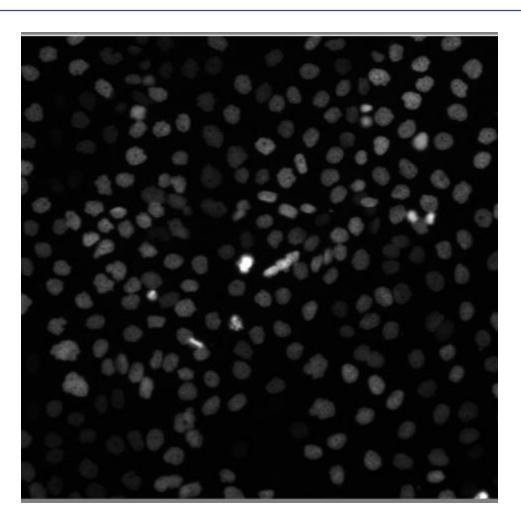


Input 1: N2DH-GOWT1 cells



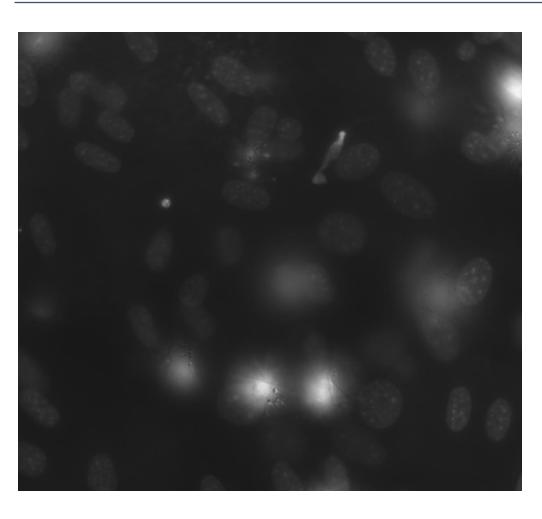
- GFP-Gowt1 mouse embryonic stem cells
- Time-laps confocal microscopy and GFP-staining
- Investigate genomic integrity of the cells
- Challenges:
 - Brightness of cells varies
 - Some cells hardly visible

Input 2: N2DL-HeLa cells



- human epithelial cells of cervical cancer
- live imaging of fluorescently labelled chromosomes
- Phenotypic profiling of the human genome
- Challenges:
 - Some cells hardly can be seen
 - Not easy to distinguish background and cells
 - Not much contrast

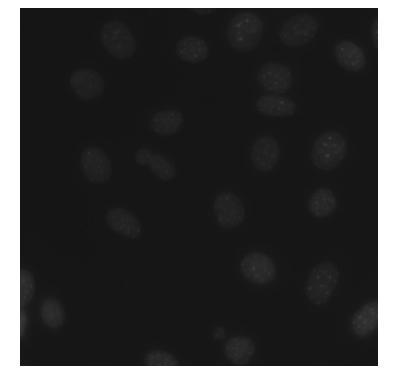
Input 3: NIH3T3 cells



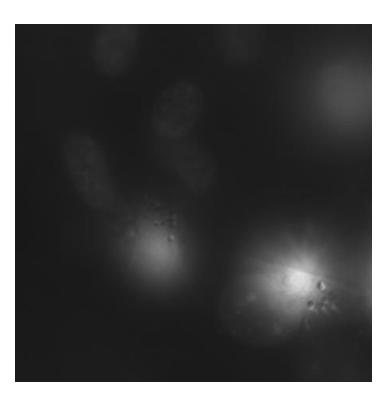
- Several mouse embryonic fibroblast cells
- Fluorescence microscopy images
- Evaluation of image analysis pipelines
- Challenges:
 - Visible debris: light spots
 - Nuclei vary in brightness

Problems

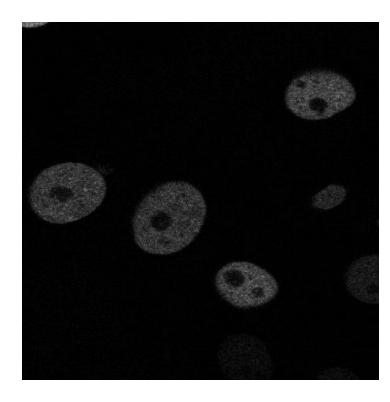
Low contrast



Reflections



Random noise

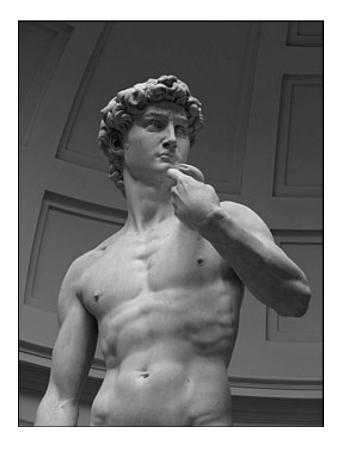


Preprocessing

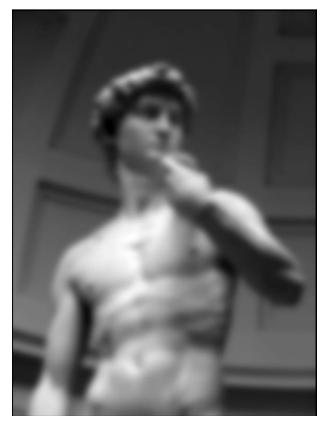
Solutions:

- Random noise
- → Gauss filter, median filter
- Reflections

Low contrast



Original

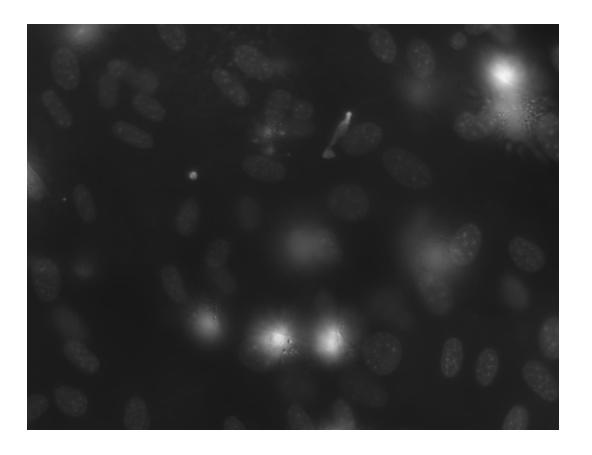


Gauss filter ($\sigma = 3$)

Preprocessing

Solutions:

- Random noise
- → Gauss filter, median filter
- Reflections
- **→** Thresholding
- Low contrast

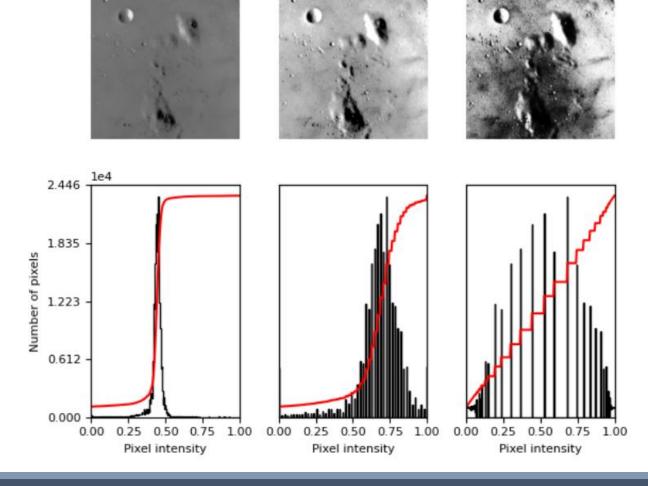


Preprocessing

Low contrast image

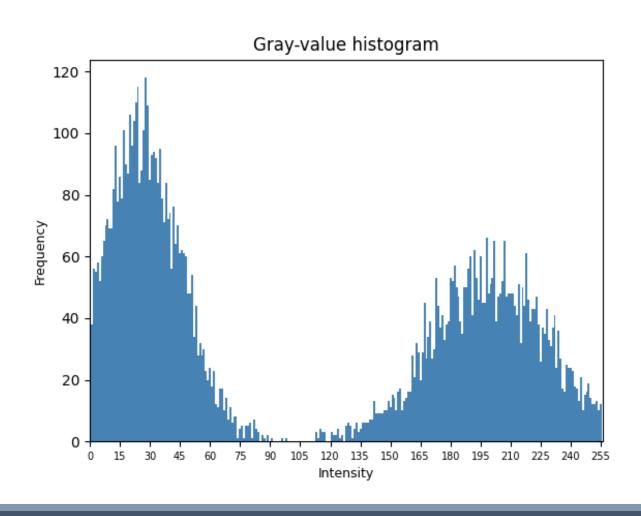
Solutions:

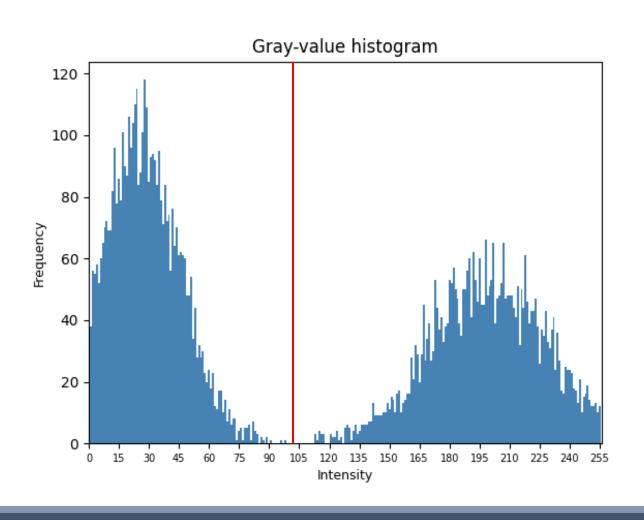
- Random noise
- → Gauss filter, median filter
- Reflections
- **→** Thresholding
- Low contrast
- Histogram equalization

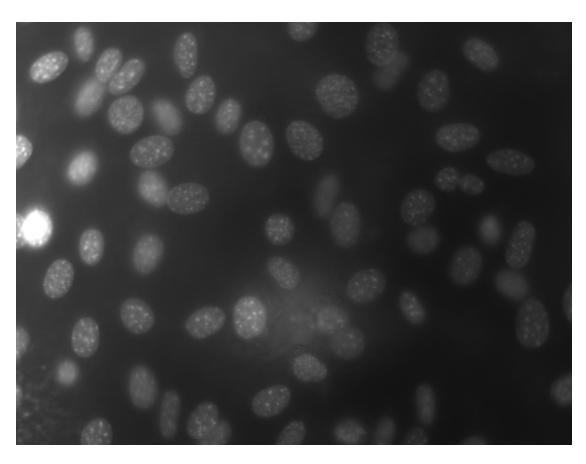


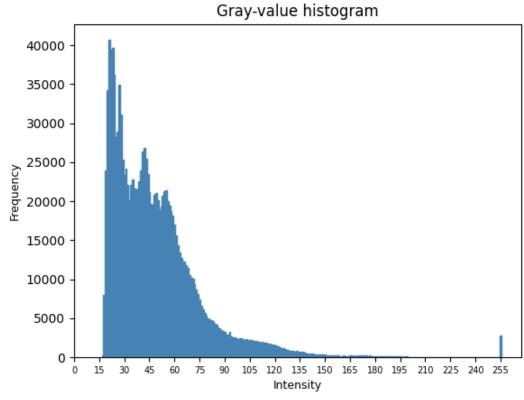
Contrast stretching

Histogram equalization







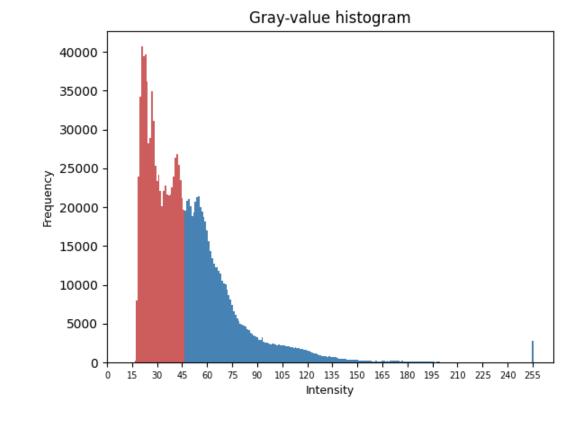


Threshold value $k \in [0,255]$

Between-class variance

$$\sigma_{\rm B} = \omega_0 \omega_1 (\mu_1 - \mu_0)^2$$

 $\omega_{0,1}=$ probability of class occurrence $\mu_{0,1}=$ mean intensity values



Threshold value $k \in [0,255]$

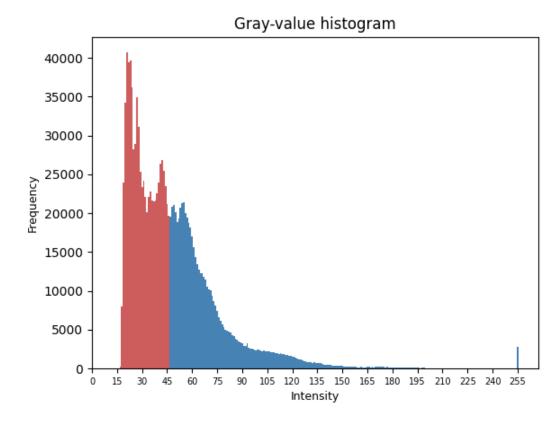
Criterion measure

$$\eta(k) = \frac{\sigma_B^2(k)}{\sigma_T^2}$$

 σ_B = between-class variance

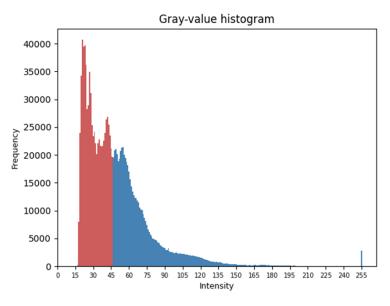
 $\sigma_T = \text{total variance}$

 $\eta(k) \in [0,1]$

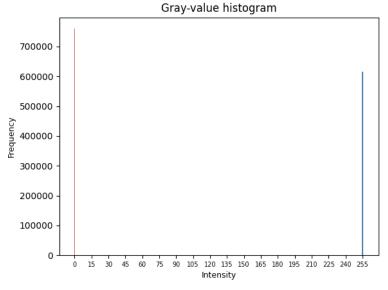


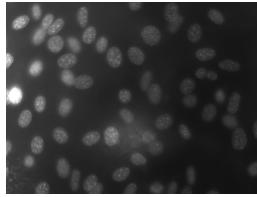
Threshold value $k \in [0,255]$

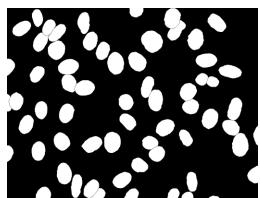
Image Clipping

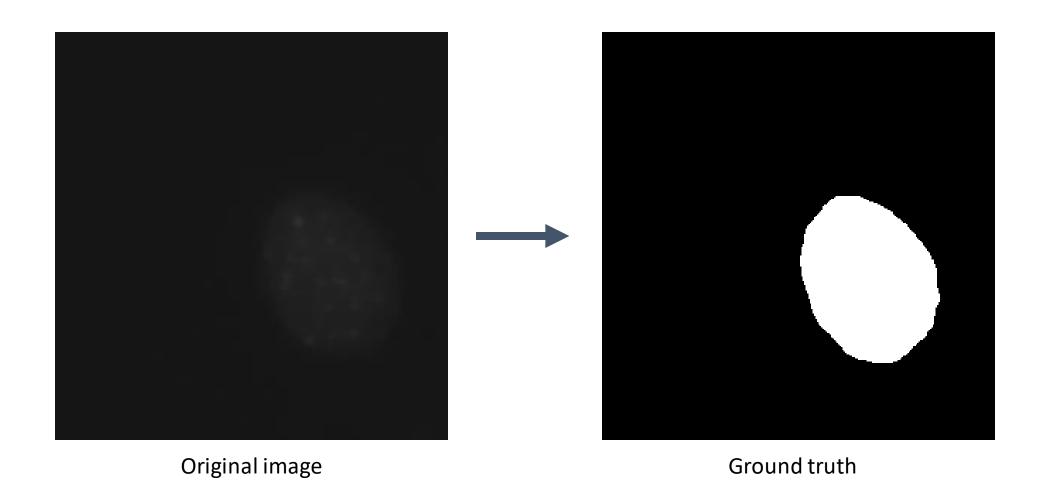


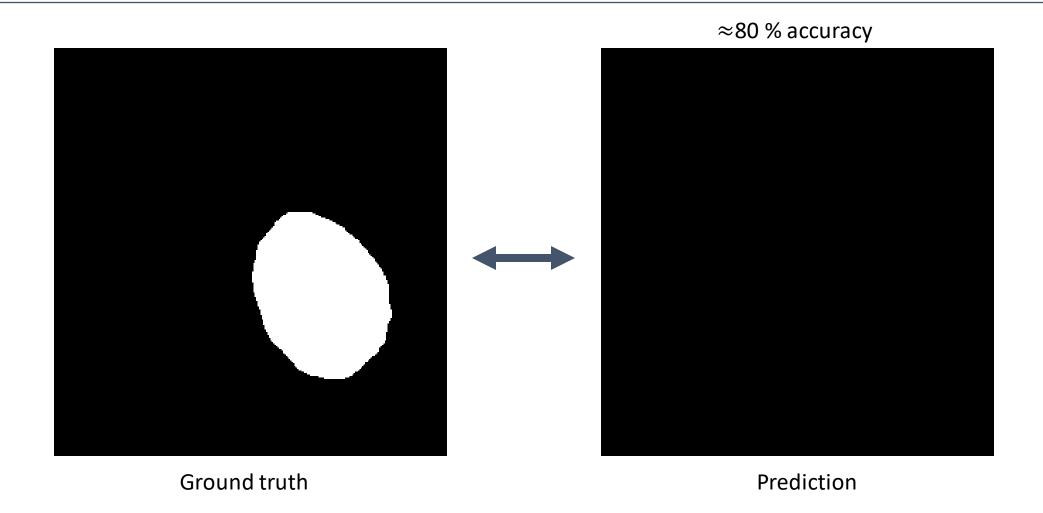
$$g_{clip}(x,y) = \begin{cases} 0 & \text{if } g(x,y) \le k \\ 255 & \text{if } g(x,y) > k \end{cases}$$



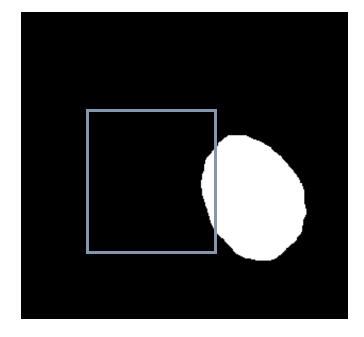




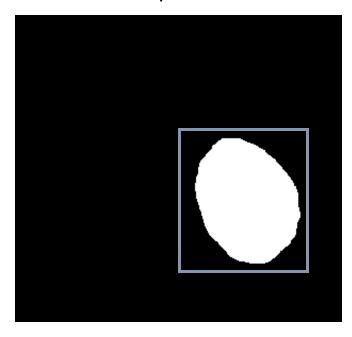




Bad prediction

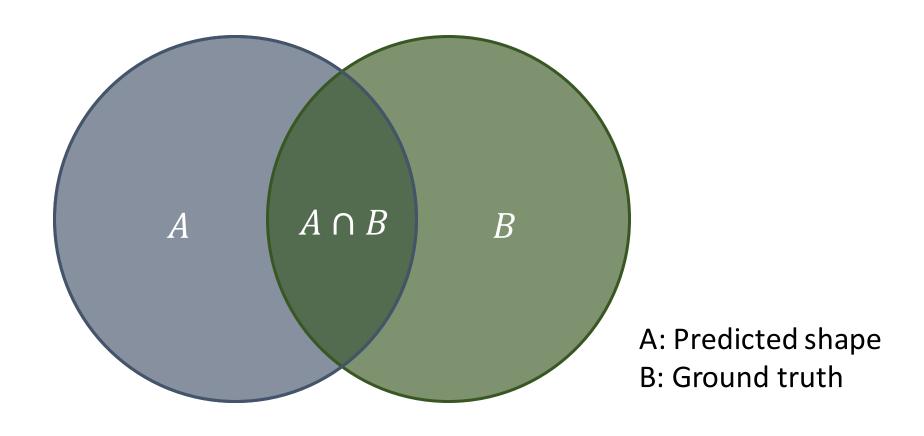


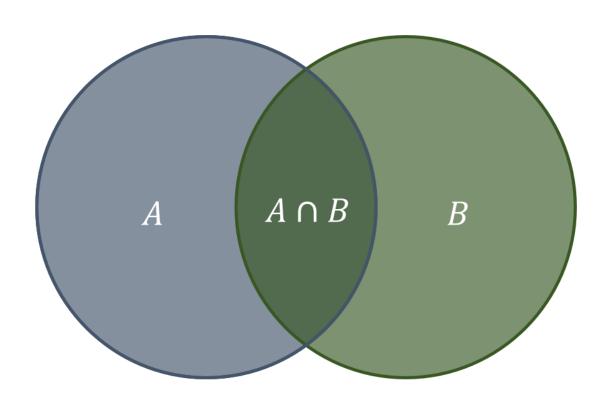
Good prediction



Very good prediction



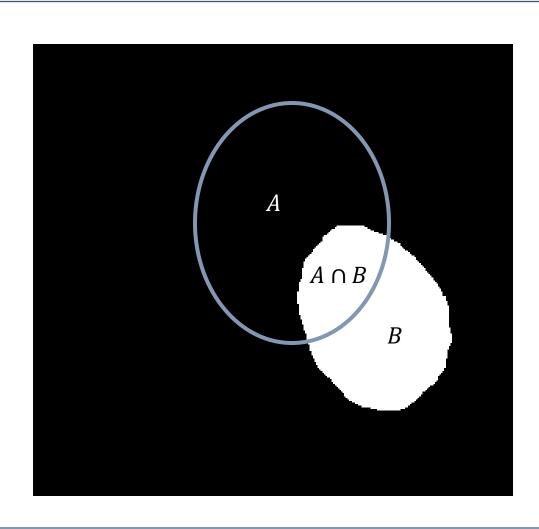




$$DSC = \frac{2 \times A \cap B}{A + B}$$

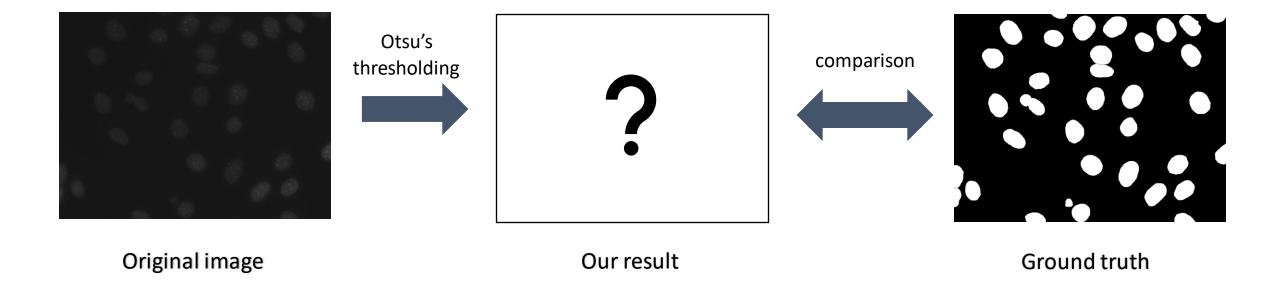
A: Predicted shape

B: Ground truth



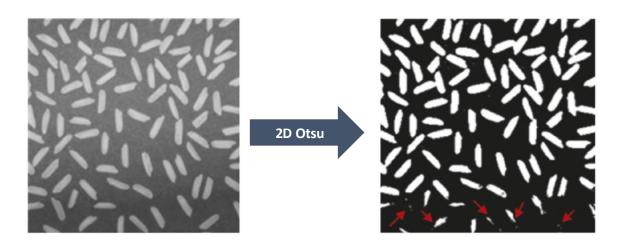
$$DSC = \frac{2 \times A \cap B}{A + B}$$

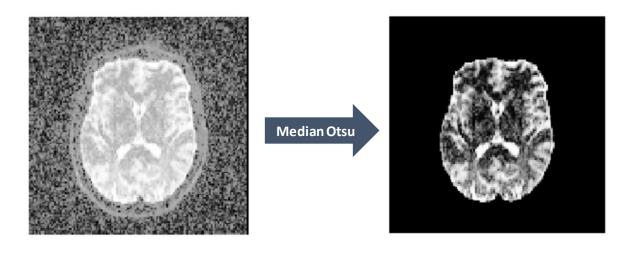
Our goal: compare ground truth images with our results



Further ideas

- 2D Otsu
- Median Otsu
- Algorithm for counting cells
- Algorithm for drawing cell trajectories





Timeline

Date	Milestone
Already done:	Researched Otsu thresholding and Dice scoring; Prepared project presentation
19.05.	Explore data with histograms and similar (All)
26.05.	Code algorithms for Otsu thresholding (H, E) and Dice scoring (L, V)
02.06.	Assemble the whole pipeline, test different preprocessing options (All)
09.06.	Research alternative evaluation methods
16.06.	 Test our pipeline on data from the BBBC Implement alternative evaluation methods: IoU, pixel accuracy (All) Hausdorff metric (H, E) NSD (L, V)
23.06.	Compare results with group 4.4 and 4.5
30.06.	Complete report in Jupyter Notebook (All)
07.07.	Complete final presentation (All)

Thank you for your attention!