

# Surface reconstruction

Robert Haase

Using materials from Alba Villaronga Luque and Jesse Veenliet (MPI CBG Dresden), Marcelo Leomil Zoccoler, Johannes Soltwedel and Mara Lampert, PoL, TU Dresden

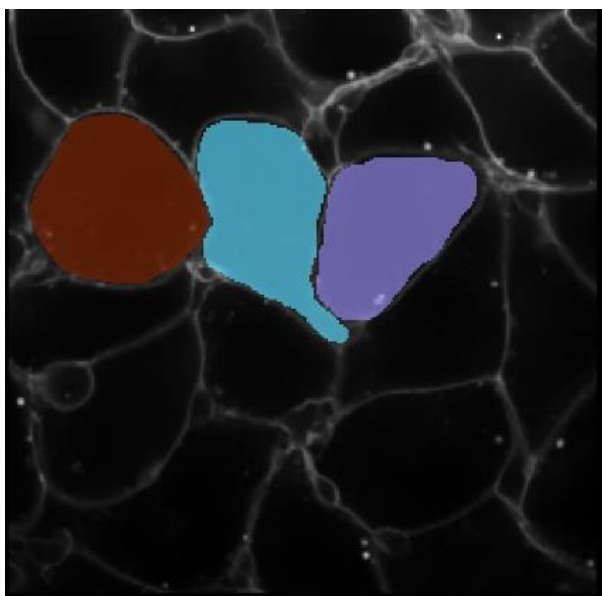
GEFÖRDERT VOM



Bundesministerium  
für Bildung  
und Forschung

Diese Maßnahme wird gefördert durch die Bundesregierung aufgrund eines Beschlusses des Deutschen Bundestages. Diese Maßnahme wird mitfinanziert durch Steuermittel auf der Grundlage des von den Abgeordneten des Sächsischen Landtags beschlossenen Haushaltes.

# Sparse Jaccard Index



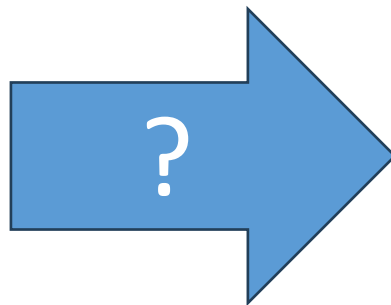
This is a ...

Sparse  
instance  
segmentation

Sparse  
semantic  
segmentation

# Quiz: Recap

- How is this operation called?



Dilation



Erosion



Opening

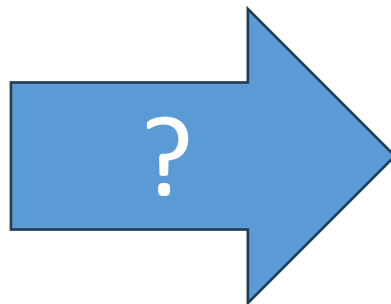


Closing



# Quiz: Recap

- How is this operation called?



Dilation



Erosion



Opening

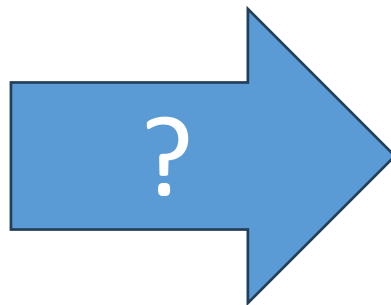


Closing



# Quiz: Recap

- How is this operation called?



Dilation



Erosion



Opening

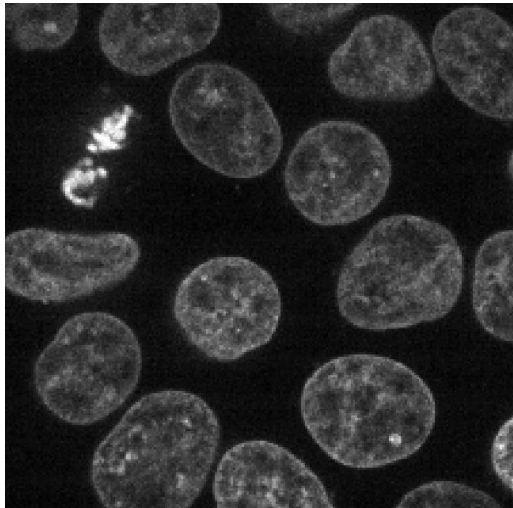


Closing

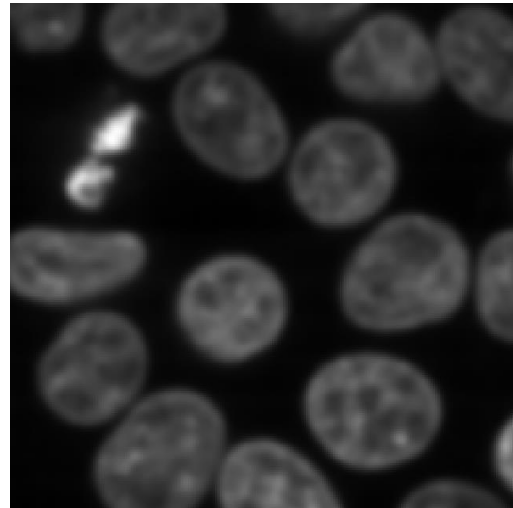


# Motivation: Surface reconstruction

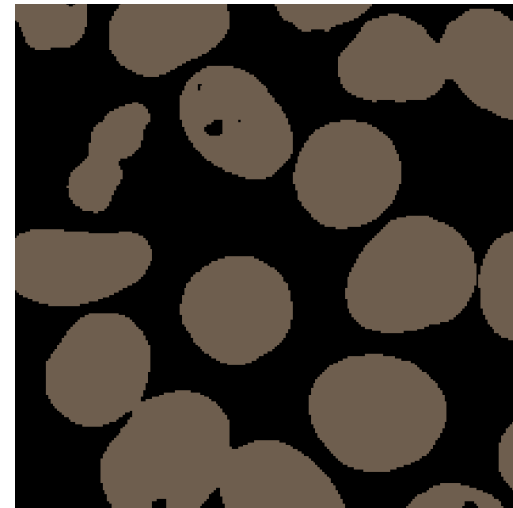
- Pixel and voxel arrays can be huge in memory
- Processing 3D arrays is time-consuming



1024 x 1024 x 100  
16-bit image



1024 x 1024 x 100  
16-bit image



1024 x 1024 x 100  
8-bit image



1024 x 1024 x 100  
16-bit image

How much memory does  
this workflow cost?

700 MB

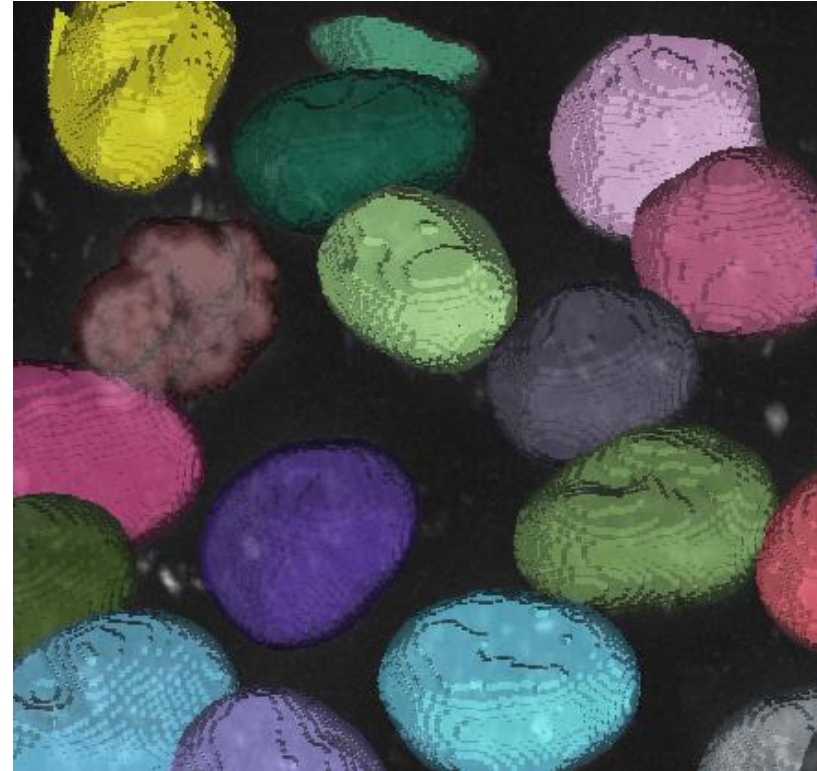
400 MB

4 GB

7 GB

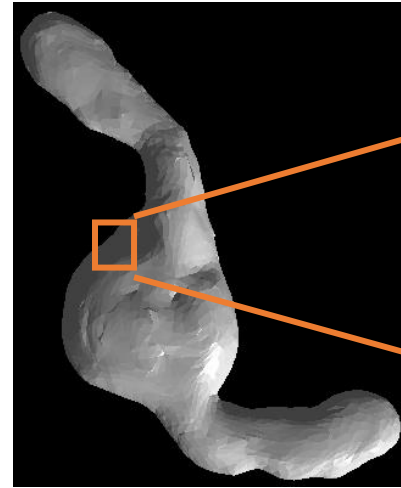
# Motivation: Surface reconstruction

- Pixel and voxel borders introduce artifacts, potentially problematic for measurements, e.g. surface area



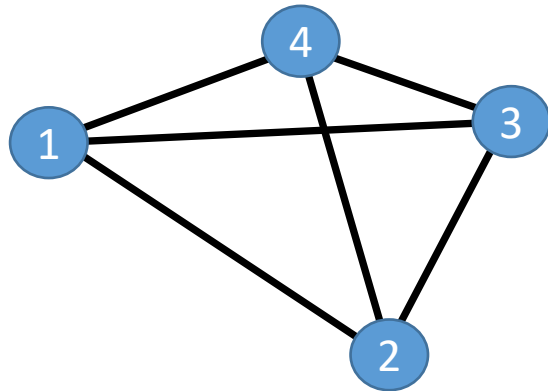
# Surface meshes

- Points on a surfaces connected by triangles forma a surface mesh



“Vertices” / points

“Faces” / Triangles



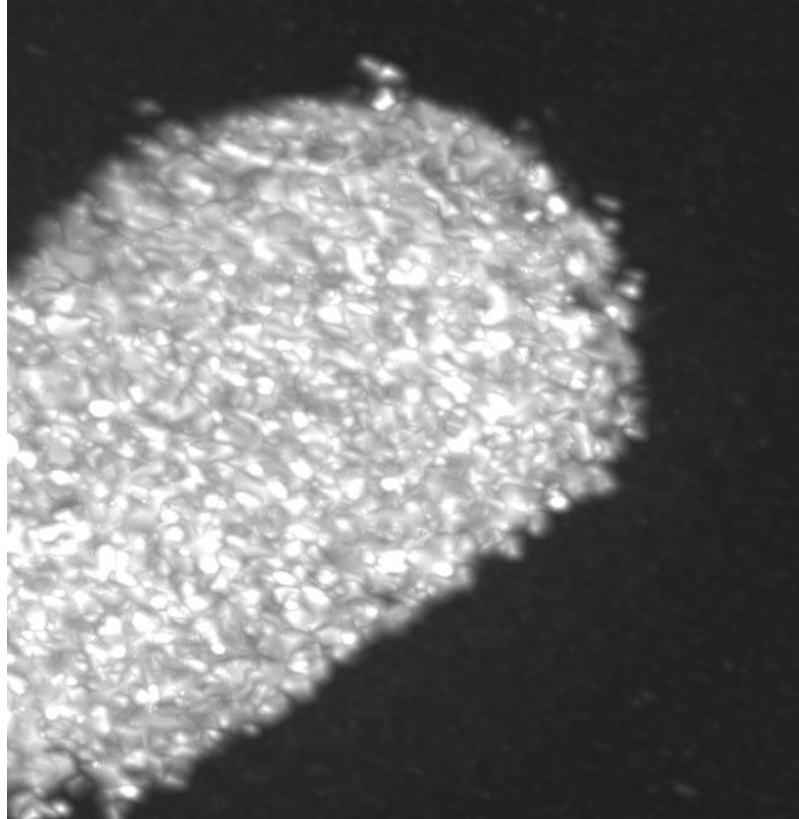
Point x	Point y	Point z
$x_1$	$y_1$	$z_1$
$x_2$	$y_2$	$z_2$
$x_3$	$y_3$	$z_3$
$x_4$	$y_4$	$z_4$
...	...	...

+

Point 1	Point 2	Point 3
1	2	3
1	2	4
2	3	4
1	3	4



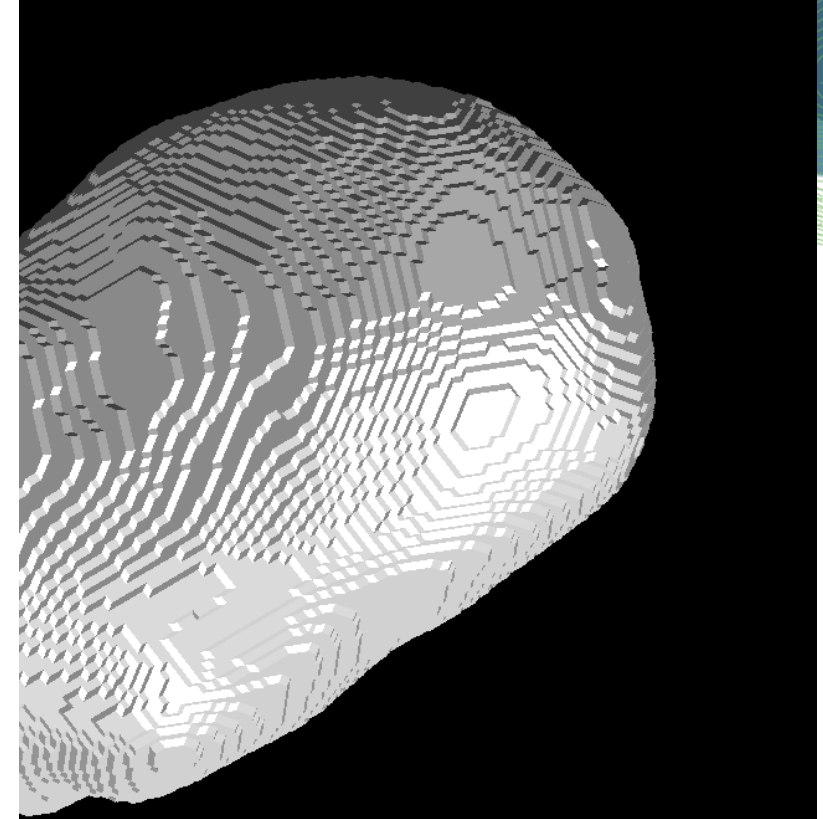
# Surface reconstruction



3D image of nuclei



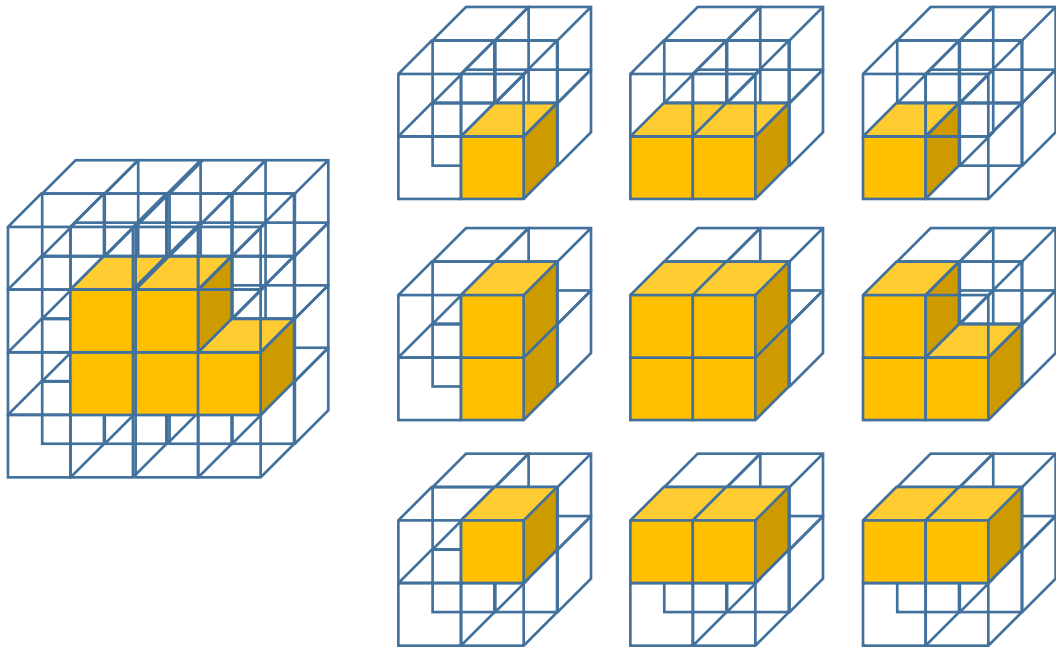
Gaussian filtered



Binary 3D image  
(visualized as surface mesh)

# Marching cubes algorithm

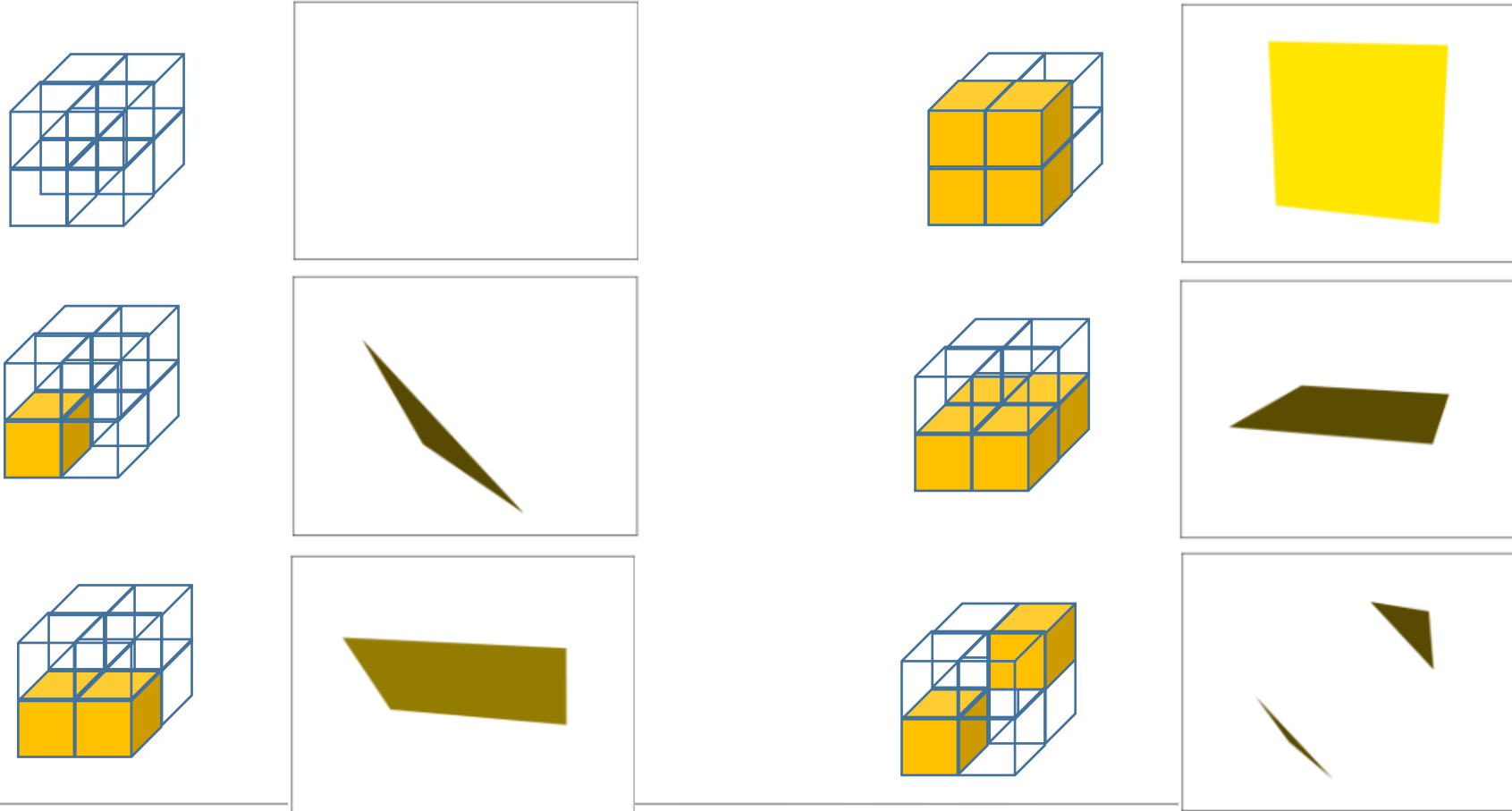
- Starting point: 3D binary image
- Cuts the image in small cubes and iterates over them



Split into cubes

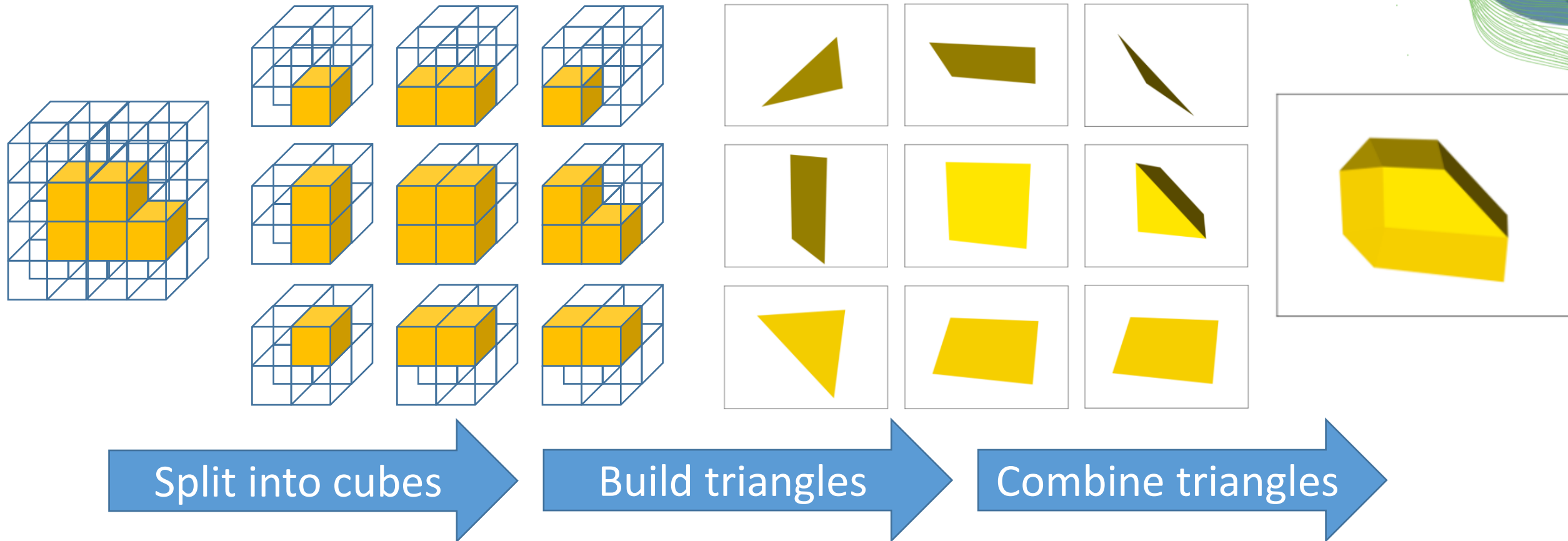
# Marching cubes algorithm

- Starting point: 3D binary image
- Cuts the image in small cubes and iterates over them



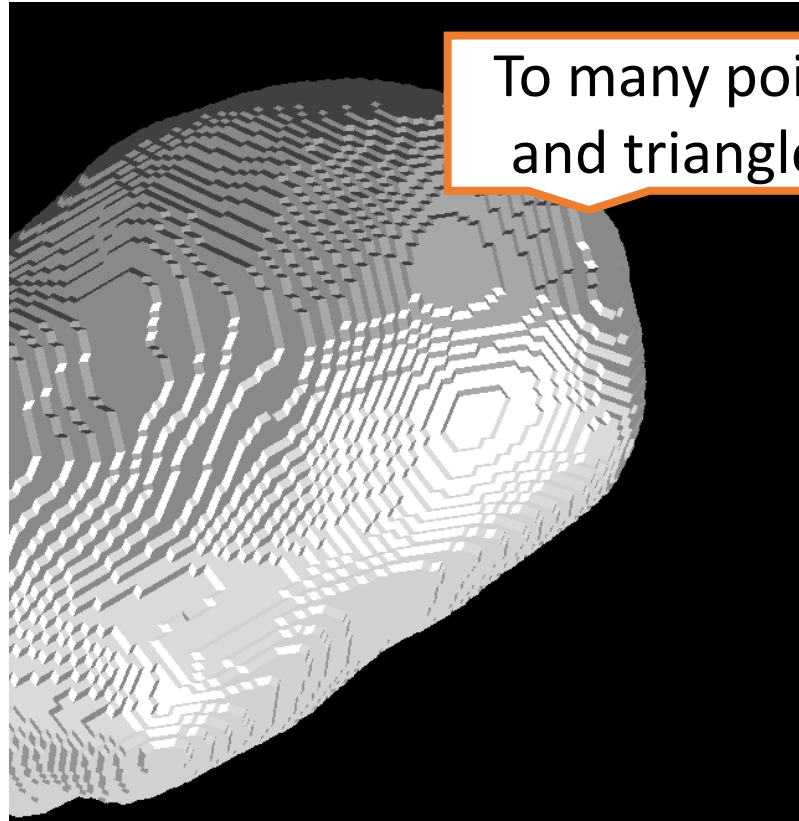
# Marching cubes algorithm

- Starting point: 3D binary image
- Cuts the image in small cubes and iterates over them



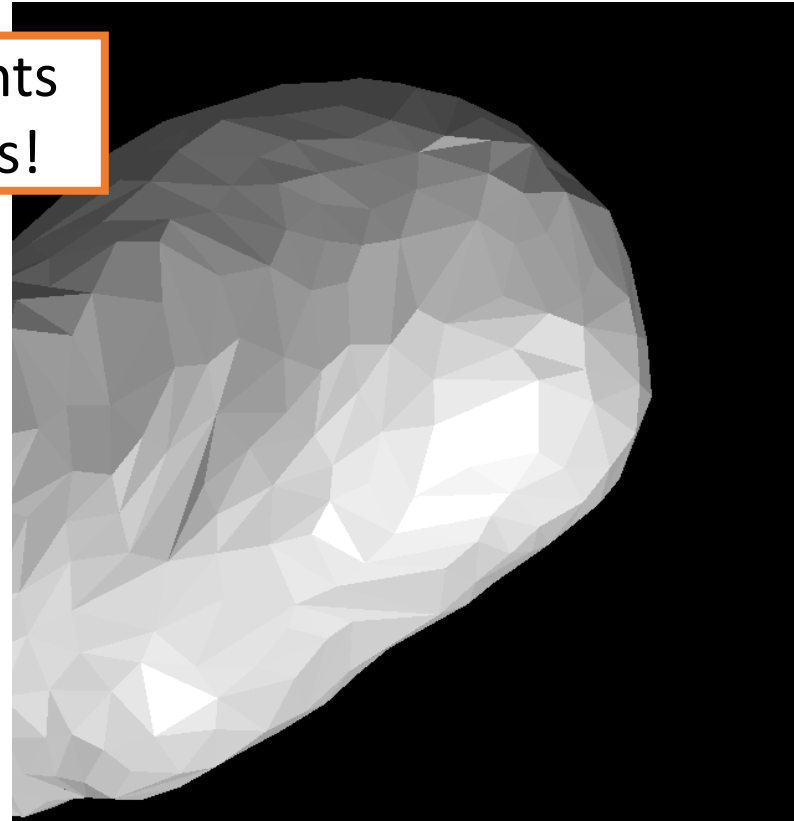
# Surface post-processing

- Necessary to better match biological reality.

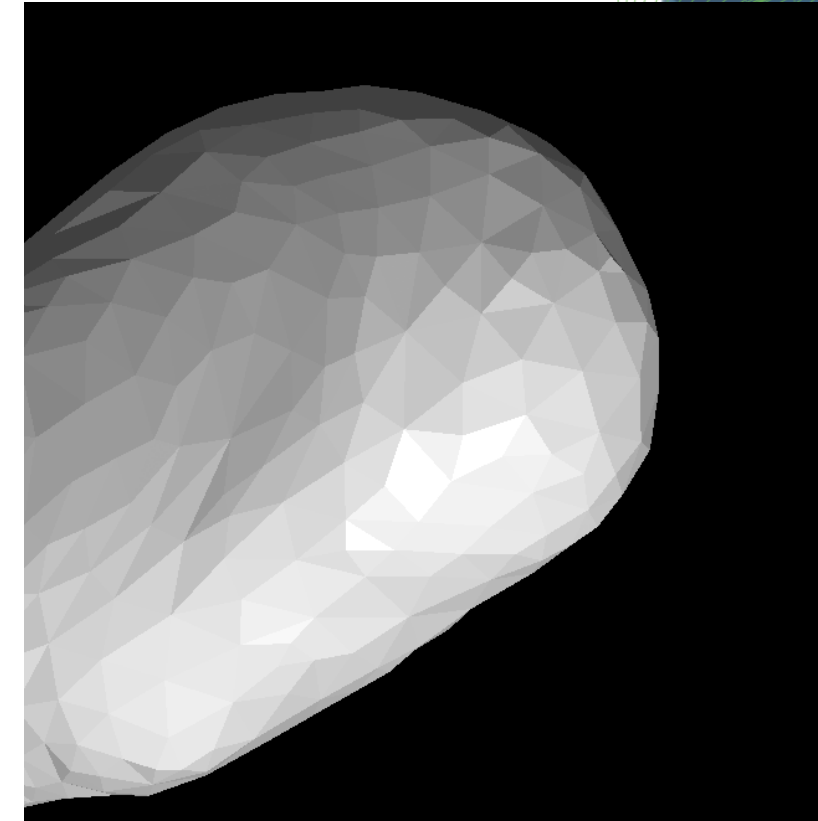


To many points  
and triangles!

Marching cubes result



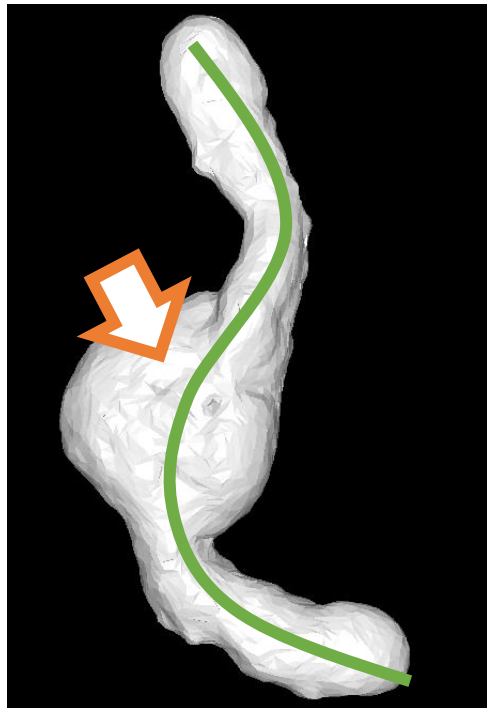
Simplified mesh  
(less points, locally averaged)



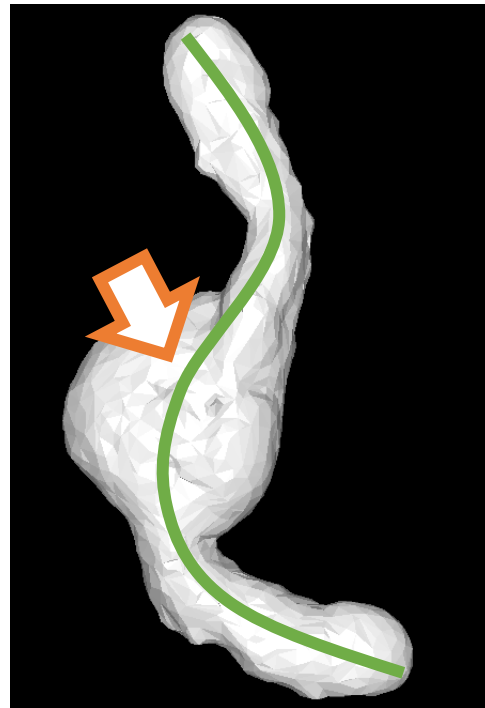
Smoothed mesh  
(position locally planarized)

# Surface post-processing

- Every processing step has consequences errors of later measurements
- Depends on desired measurement



Surface mesh

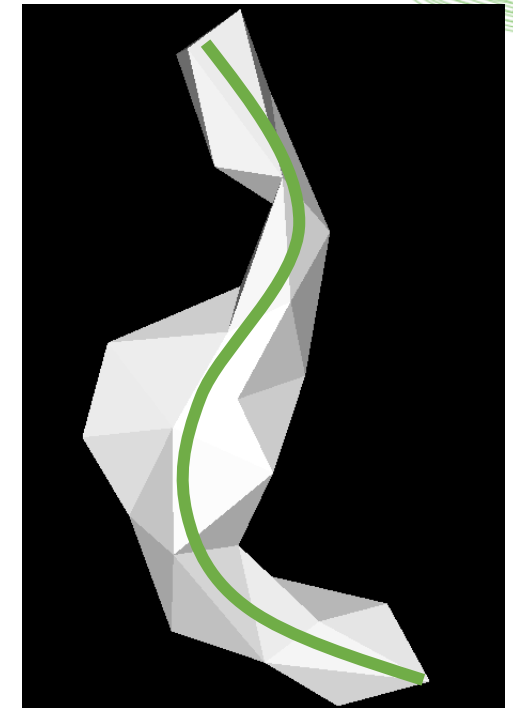


Simplified by factor 0.5



Simplified by factor 0.05

Number of small concave regions

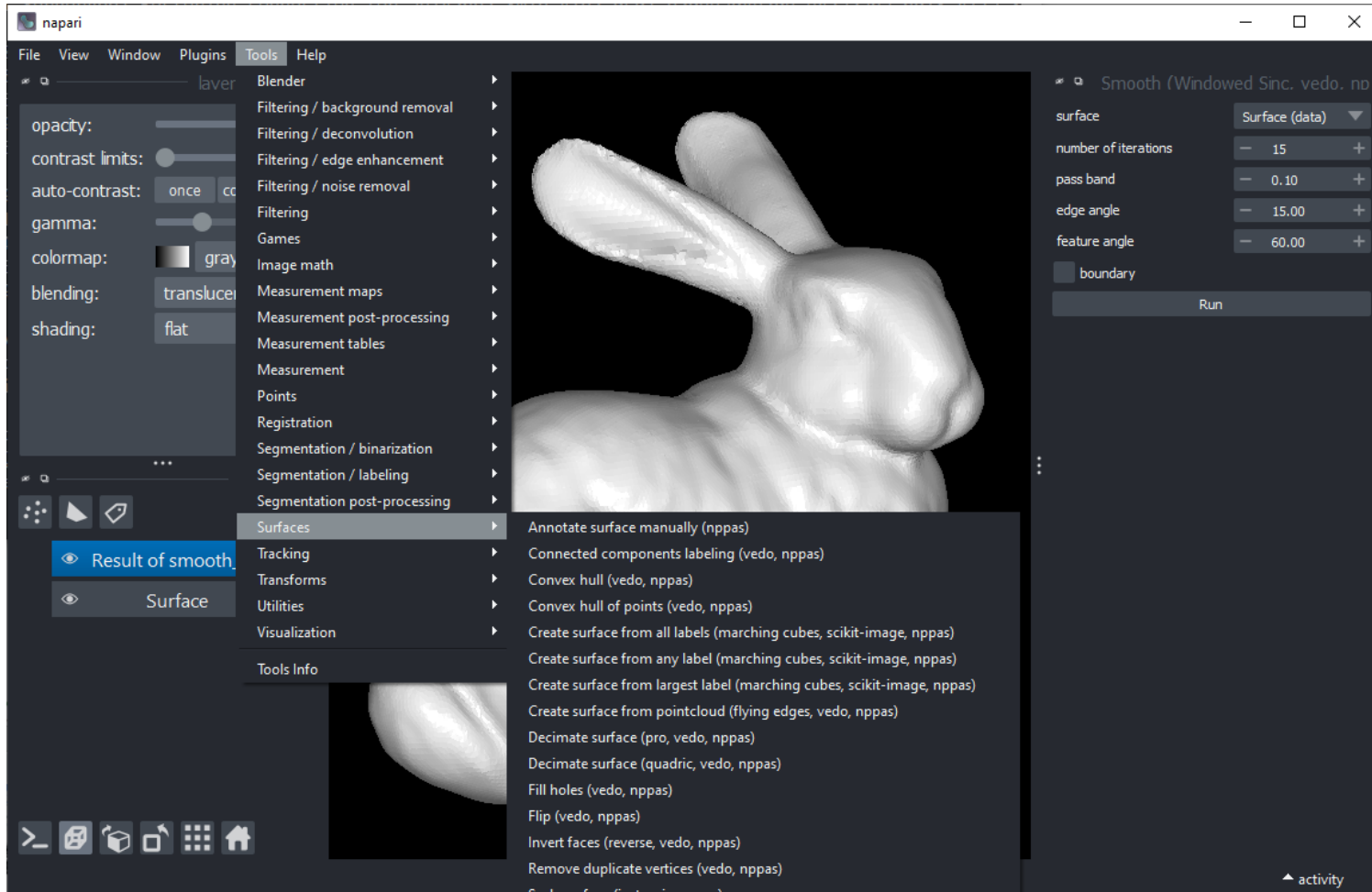


Simplified by factor 0.01

Total length

# Surface reconstruction / Processing

- Tools > Surfaces > Create surface ...



You need to install an extra napari-plugin:  
<https://github.com/haesleinhuepf/napari-process-points-and-surfaces>

# Surface reconstruction

- Turn binary and/or label images into surface meshes

```
surface = nppas.all_labels_to_surface(binary_filled)
```

```
surface
```

## nppas.SurfaceTuple

origin (z/y/x) [0. 0. 0.]

center of mass(z/y/x) 57.710,309.963,440.042

scale(z/y/x) 1.000,1.000,1.000

bounds (z/y/x) 12.500...113.500  
111.500...461.500  
169.500...807.500

average size 170.769

number of vertices 330776

number of faces 661548

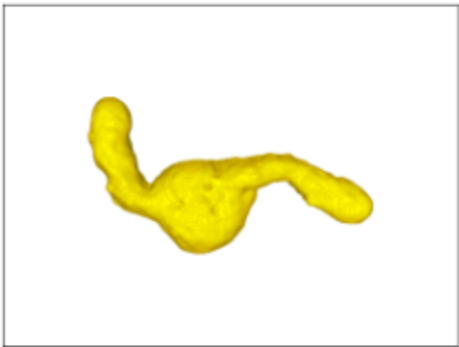




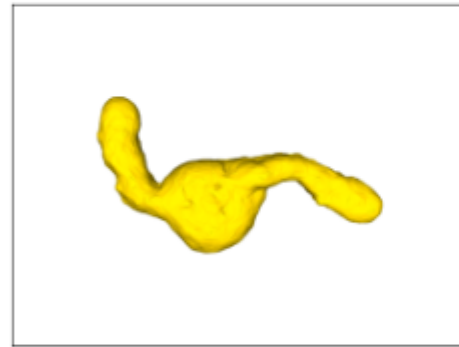
# Surface mesh processing

- Surface mesh simplification
- To prevent the computer freezing

```
simplified_surface = nppas.decimate_quadric(surface, fraction=0.01)  
simplified_surface
```



nppas.SurfaceTuple	
origin (z/y/x)	[0. 0. 0.]
center of mass(z/y/x)	57.710,309.963,440.042
scale(z/y/x)	1.000,1.000,1.000
bounds (z/y/x)	12.500...113.500 111.500...461.500 169.500...807.500
average size	170.769
number of vertices	330776
number of faces	661548



nppas.SurfaceTuple	
origin (z/y/x)	[0. 0. 0.]
center of mass(z/y/x)	57.928,308.938,440.985
scale(z/y/x)	1.000,1.000,1.000
bounds (z/y/x)	13.231...113.510 111.642...461.602 170.022...806.468
average size	170.083
number of vertices	3310
number of faces	6615

# Surface mesh processing

- Surface mesh smoothing

```
smoothed_surface = nppas.smooth_surface(simplified_surface)
smoothed_surface
```

## nppas.SurfaceTuple

origin (z/y/x) [0. 0. 0.]

center of mass(z/y/x) 57.928,308.938,440.985

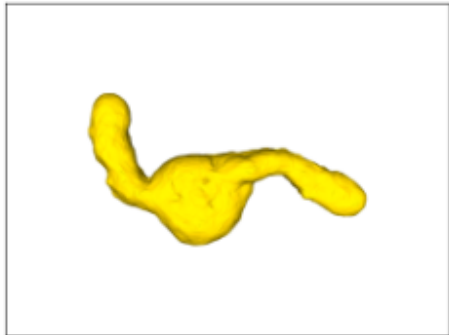
scale(z/y/x) 1.000,1.000,1.000

bounds (z/y/x) 13.231...113.510  
111.642...461.602  
170.022...806.468

average size 170.083

number of vertices 3310

number of faces 6615



## nppas.SurfaceTuple

origin (z/y/x) [0. 0. 0.]

center of mass(z/y/x) 57.913,308.988,440.878

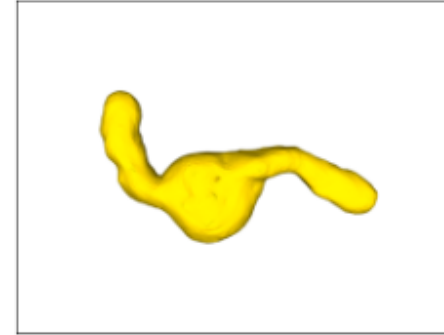
scale(z/y/x) 1.000,1.000,1.000

bounds (z/y/x) 13.901...113.627  
110.982...461.191  
169.711...807.193

average size 170.378

number of vertices 3310

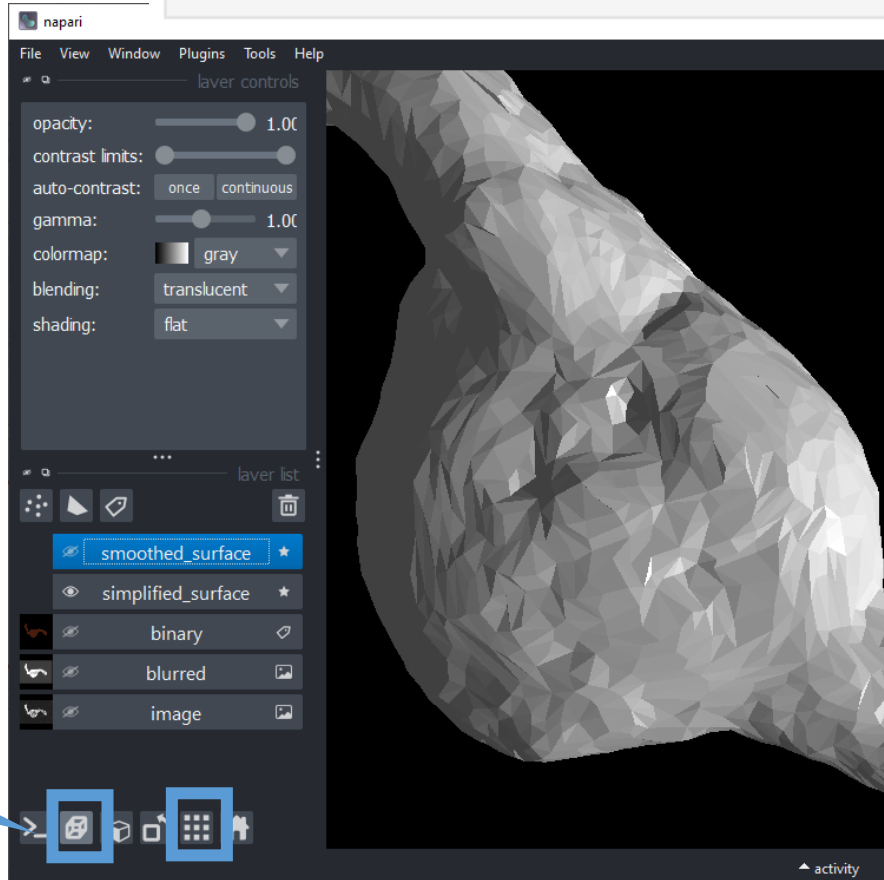
number of faces 6615



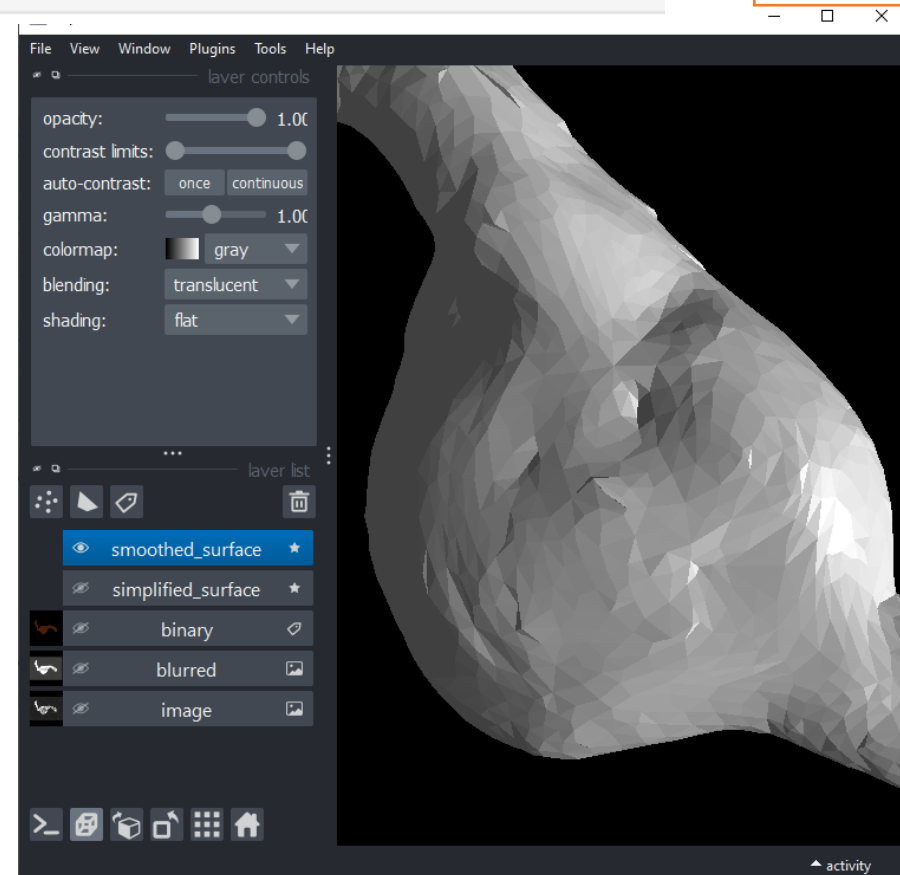
# View surface meshes in Napari

```
viewer.add_surface(surface, scale=[zoom, zoom, zoom])  
viewer.add_surface(simplified_surface, scale=[zoom, zoom, zoom])  
viewer.add_surface(smoothed_surface, scale=[zoom, zoom, zoom])
```

In case your computer freezes, comment out this line



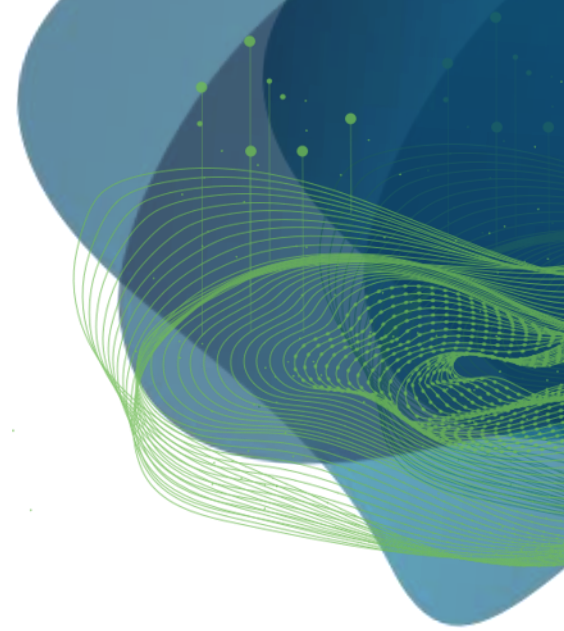
Toggle 3D view and grid mode



# Segmentation quality estimation

Robert Haase

Reusing materials from Lena Maier-Hein, Annika Reinke (DKFZ) et al. and Martin Schätz (Charles Uni Prague)



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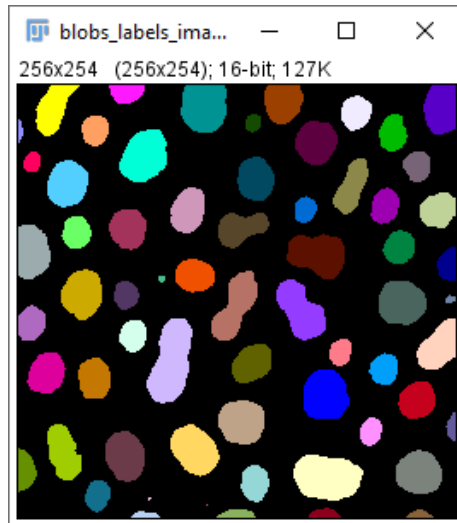


Diese Maßnahme wird gefördert durch die Bundesregierung aufgrund eines Beschlusses des Deutschen Bundestages. Diese Maßnahme wird mitfinanziert durch Steuermittel auf der Grundlage des von den Abgeordneten des Sächsischen Landtags beschlossenen Haushaltes.

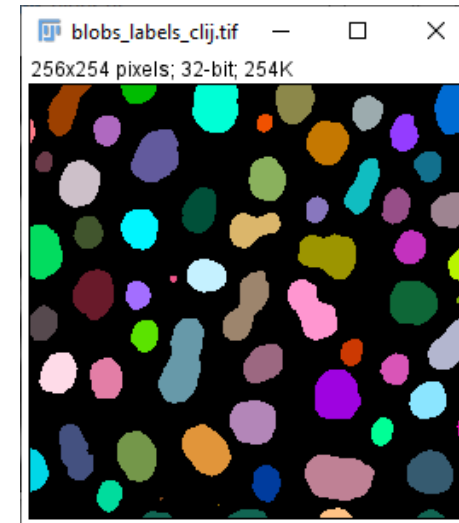
# Goal

- Compare label images quantitatively, to know
  - how “good” a segmentation algorithm is and/or
  - how “variable” segmentations (from humans or computers are)

How can we know if these results are the same?



Human  
annotation  
("ground truth")



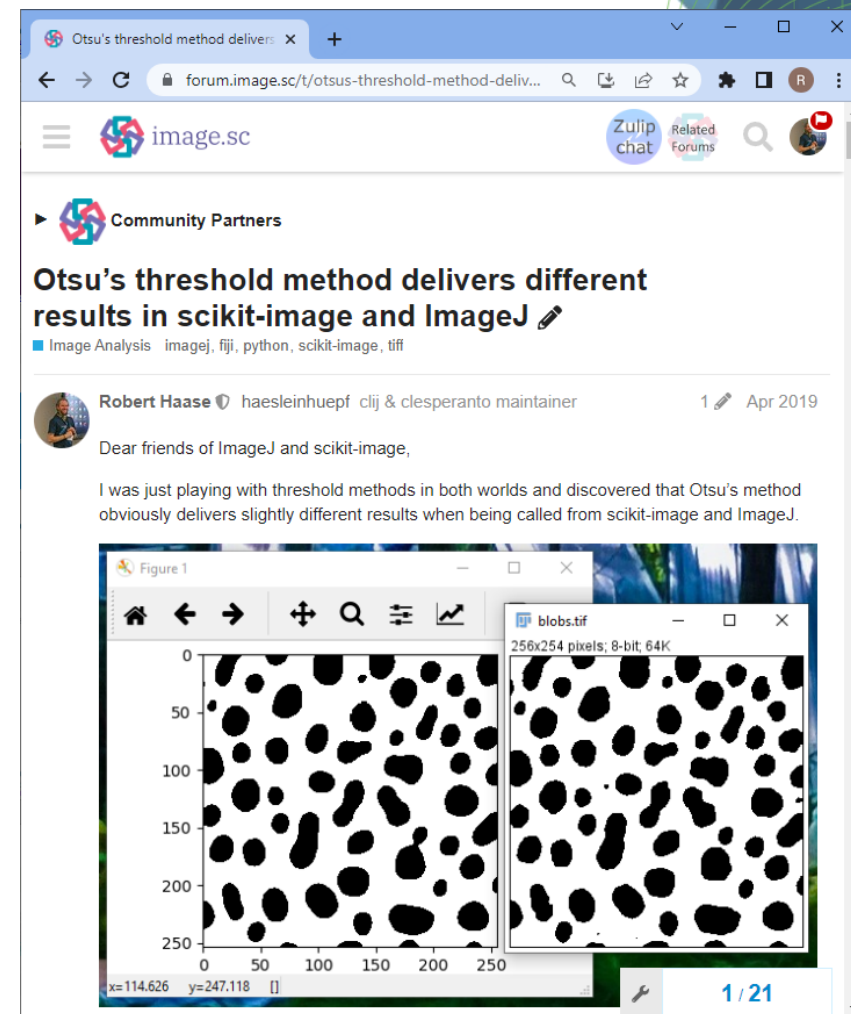
Algorithm result

# Why do results vary?

Potential reasons of same workflows delivering different results:

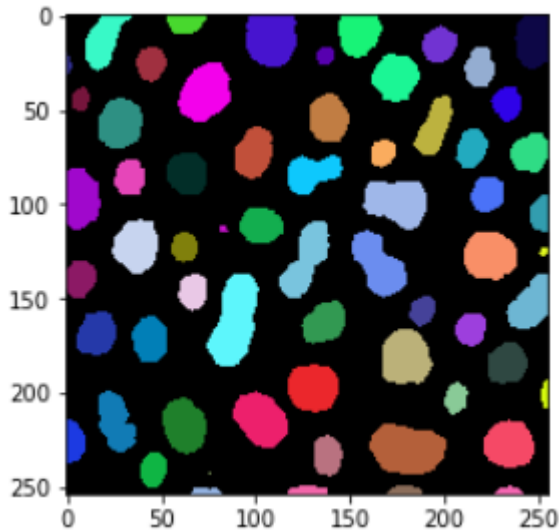
- Image data type (8/16/32-bit float/int)
- Workflow implementation
- How histograms are determined
- How the threshold is determined from the histogram
- Compute architecture (CPU, GPU, TPU, ...)
- Hardware vendor
- Software / driver versions

Lecture in 2 weeks

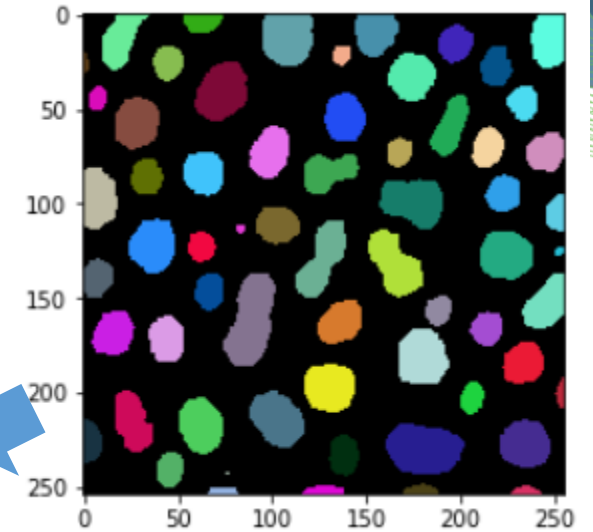
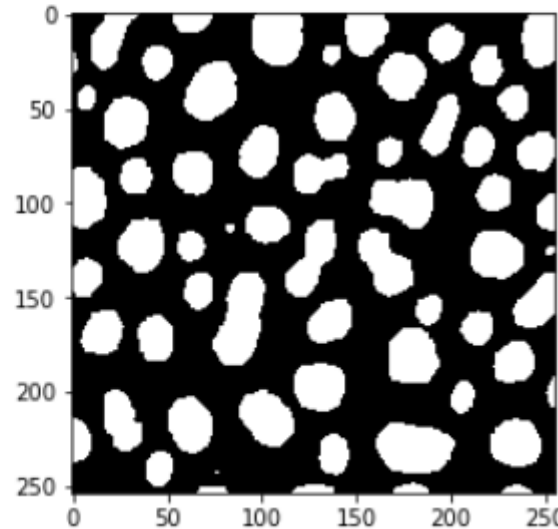
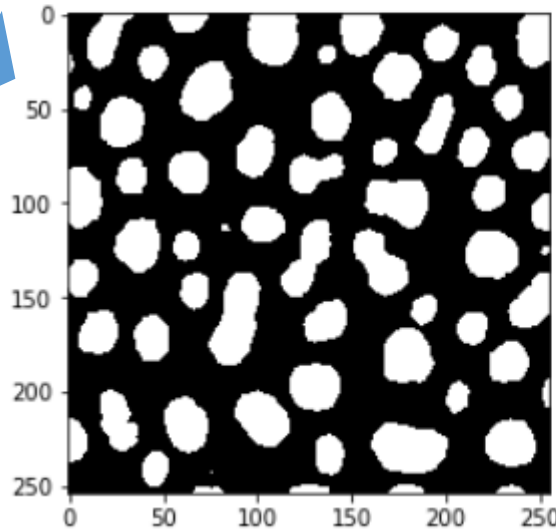


# Visual comparison

- The order in label images may be different. To compare them visually, we need to turn them into binary images first.

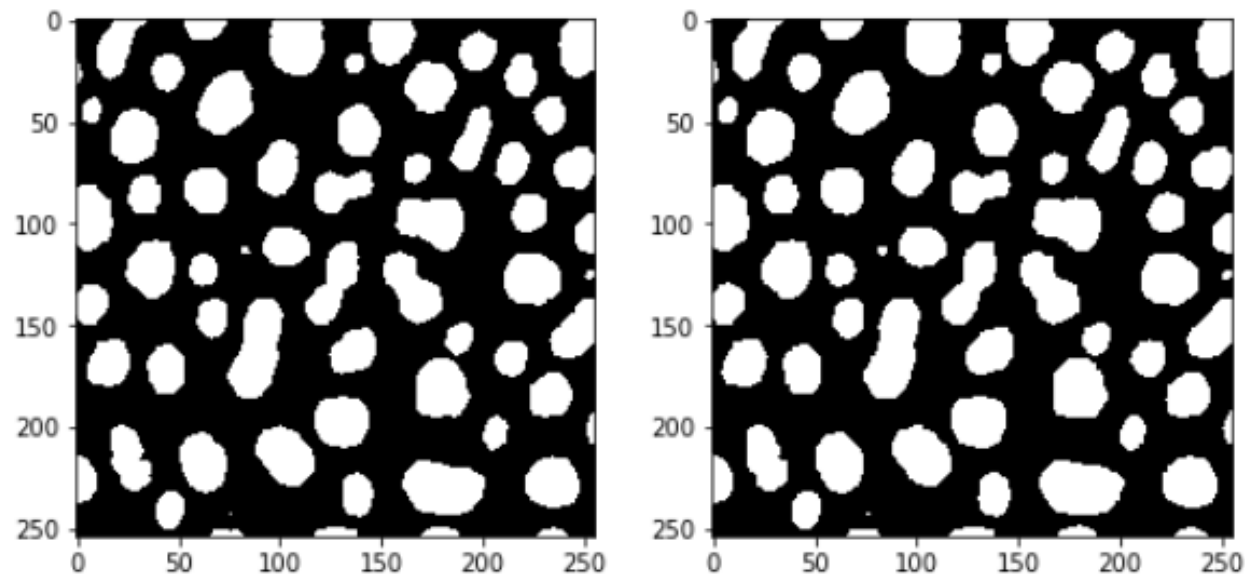


```
binary_blobs_imagej = imread(filenamees[0]) > 0  
binary_blobs_skimage = imread(filenamees[1]) > 0  
imshow(binary_blobs_imagej)  
imshow(binary_blobs_skimage)
```



# Quiz

- How many pixels  $n$  in these two images are different?



$n=0$



$0 < n < 100$



$100 < n < 1000$



$n > 1000$



# Visual comparison

- Binary image comparison: difference or XOR

```
difference = np.logical_xor(binary_blobs_imagej, binary_blobs_skimage)

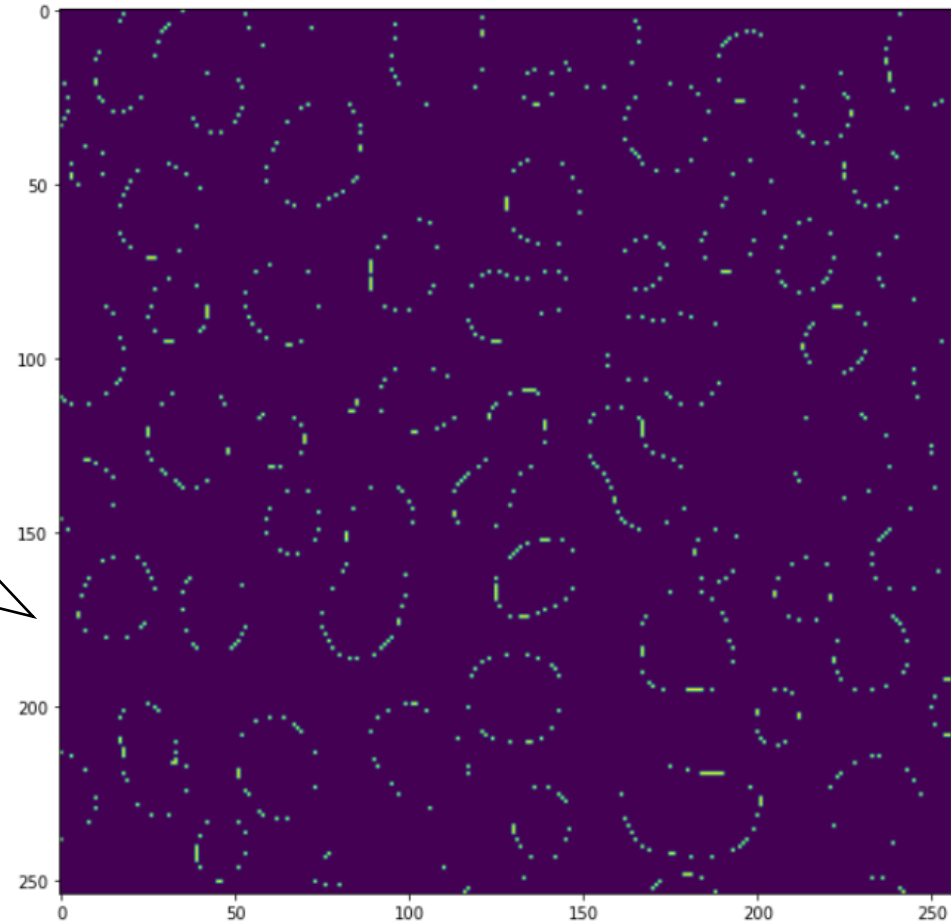
fig, axs = plt.subplots(figsize=(10,10))
axs.imshow(difference)
```

Number of different pixels:

```
[5]: np.sum(difference)
```

```
[5]: 830
```

- Does not work well if labels are close-by



# Quantitative comparison

- Voxel-wise Youden-Index

$$YI = p_{TP} + p_{TN} - 1$$

- Volume error

$$\Delta_V = V_A - V_B$$

$$\delta_V = \frac{\Delta_V}{V_B}$$

- Dice Index

$$DI(A, B) = \frac{2|A \cap B|}{|A| + |B|}$$

- Jaccard Index

$$JI(A, B) = \frac{|A \cap B|}{|A \cup B|} = \frac{DI}{2 - DI}$$

- Contour distance

$$d_{e,min}(a, B) = \min(d_e(a, b) | b \in B)$$

$$\bar{d}_c(A, B) = \frac{\sum_{\forall a \in C(A)} d_{e,min}(a, C(B))}{|C(A)|}$$

$$\bar{d}_{bil,c}(A, B) = \frac{\bar{d}_c(A, B) + \bar{d}_c(B, A)}{2}$$

- Hausdorff distance

$$d_H(A, B) = \max(d_{e,min}(a, B) | a \in A)$$

$$d_{bil,H}(A, B) = \max(d_H(A, B), d_H(B, A))$$

- Simplified Hausdorff distance

$$d_H(A, B) = \max(d_{e,min}(a, C(B)) | a \in C(A))$$

- Volume standard deviation

$$\delta_{\bar{V}} = 2 \frac{|V_A - V_B|}{|V_A + V_B|}$$

- Classification error

$$e_{Class} = \frac{H}{|TP| + |FN|}$$

- Hamming distance

$$\begin{aligned} d_h &= |A \cup B| - |A \cap B| \\ &= |FP| + |FN| \end{aligned}$$

# Choosing the right metric is key

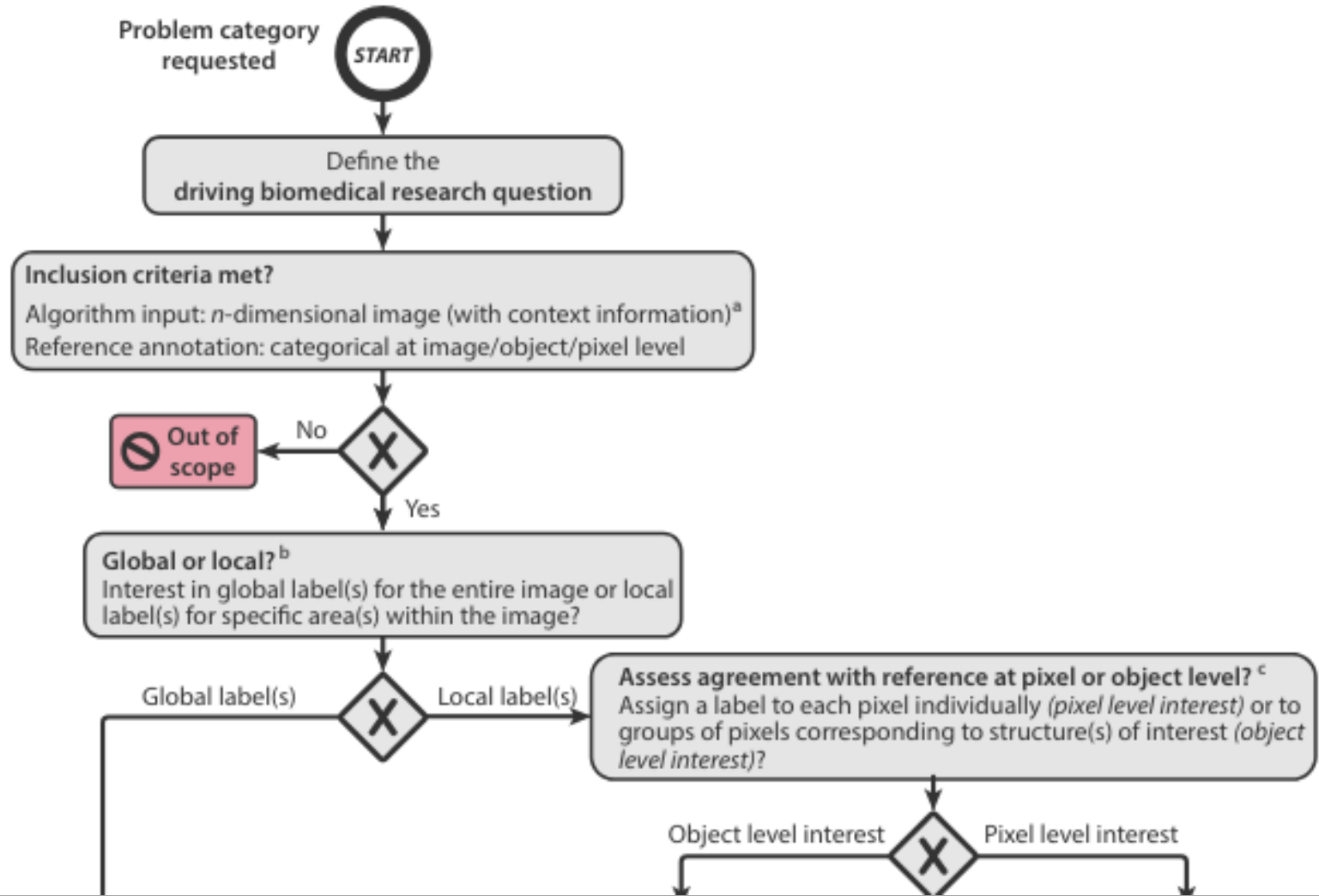
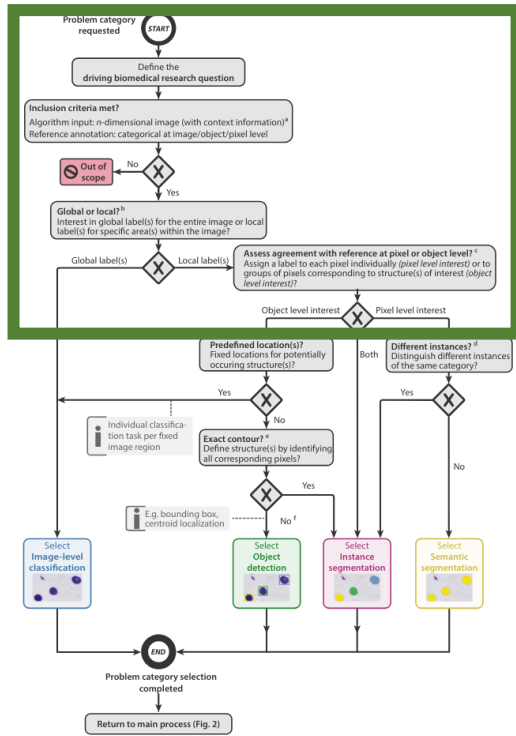
- Systematic overview by Maier-Hein, Reinke at al.

The screenshot shows the arXiv preprint page for the paper "Metrics reloaded: Recommendations for image analysis validation". The page is from the Cornell University arXiv repository, specifically in the Computer Science > Computer Vision and Pattern Recognition section. The title is "Metrics reloaded: Recommendations for image analysis validation". The authors listed are Lena Maier-Hein, Annika Reinke, Patrick Godau, Minu D. Tizabi, Florian Buettner, Evangelia Christodoulou, Ben Glocker, Fabian Isensee, Jens Kleesiek, Michal Kozubek, Mauricio Reyes, Michael A. Riegler, Manuel Wiesenfarth, A. Emre Kavur, Carole H. Sudre, Michael Baumgartner, Matthias Eisenmann, Doreen Heckmann-Nötzel, Tim Rädtsch, Laura Acion, Michela Antonelli, Tal Arbel, Spyridon Bakas, Arriel Benis, Matthew Blaschko, M. Jorge Cardoso, Veronika Cheplygina, Beth A. Cimini, Gary S. Collins, Keyvan Farahani, Luciana Ferrer, Adrian Galdran, Bram van Ginneken, Robert Haase, Daniel A. Hashimoto, Michael M. Hoffman, Merel Huisman, Pierre Jannin, Charles E. Kahn, Dagmar Kainmueller, Bernhard Kainz, Alexandros Karargyris, Alan Karthikesalingam, Hannes Kennigott, Florian Köfler, Annette Kopp-Schneider, Anna Kreshuk, Tahsin Kurc, Bennett A. Landman, Geert Litjens, Amin Madani, Klaus Maier-Hein, Anne L. Martel, Peter Mattson, Erik Meijering, Bjoern Menze, Karel G.M. Moons, Henning Müller, Brennan Niciporuk, Felix Nickel, Jens Petersen, Nasir Rajpoot, Nicola Rieke, Julio Saez-Rodriguez, Clara I. Sánchez, Shravya Shetty, Maarten van Smeden, Ronald M. Summers, Abdel A. Taha, Aleksei Tulpin, Sotirios A. Tsaftaris, Ben Van Calster, Gaël Varoquaux, Paul F. Jäger. The submission date is 3 Jun 2022 (v1), and the last revised date is 23 Feb 2024 (this version, v8). The abstract states: "Increasing evidence shows that flaws in machine learning (ML) algorithm validation are an underestimated global problem. Particularly in automatic biomedical image analysis, chosen performance metrics often do not reflect the domain interest." The right sidebar contains links for "Access Paper" (View PDF, HTML (experimental), TeX Source, Other Formats), "Current browse context" (cs.CV), "Change to browse by" (cs), "References & Citations" (NASA ADS, Google Scholar, Semantic Scholar), "Export BibTeX Citation", and "Bookmark".

The screenshot shows the Nature Methods journal article page for the paper "Metrics reloaded: recommendations for image analysis validation". The journal is Nature Methods, published on 12 February 2024. The authors listed are Lena Maier-Hein, Annika Reinke, Patrick Godau, Minu D. Tizabi, Florian Buettner, Evangelia Christodoulou, Ben Glocker, Fabian Isensee, Jens Kleesiek, Michal Kozubek, Mauricio Reyes, Michael A. Riegler, Manuel Wiesenfarth, A. Emre Kavur, Carole H. Sudre, Michael Baumgartner, Matthias Eisenmann, Doreen Heckmann-Nötzel, Tim Rädtsch, Laura Acion, Michela Antonelli, Tal Arbel, Spyridon Bakas, Arriel Benis, ... Paul F. Jäger. The article is categorized as a Perspective. The abstract states: "Increasing evidence shows that flaws in machine learning (ML) algorithm validation are an underestimated global problem. Particularly in automatic biomedical image analysis, chosen performance metrics often do not reflect the domain interest." The right sidebar contains a "Download PDF" button and "Associated content" section with the title "The future of bioimage analysis" and the subtitle "Understanding metric-related pitfalls in image analysis validation". The bottom of the page shows "5619 Accesses | 12 Citations | 228 Altmetric | Metrics".

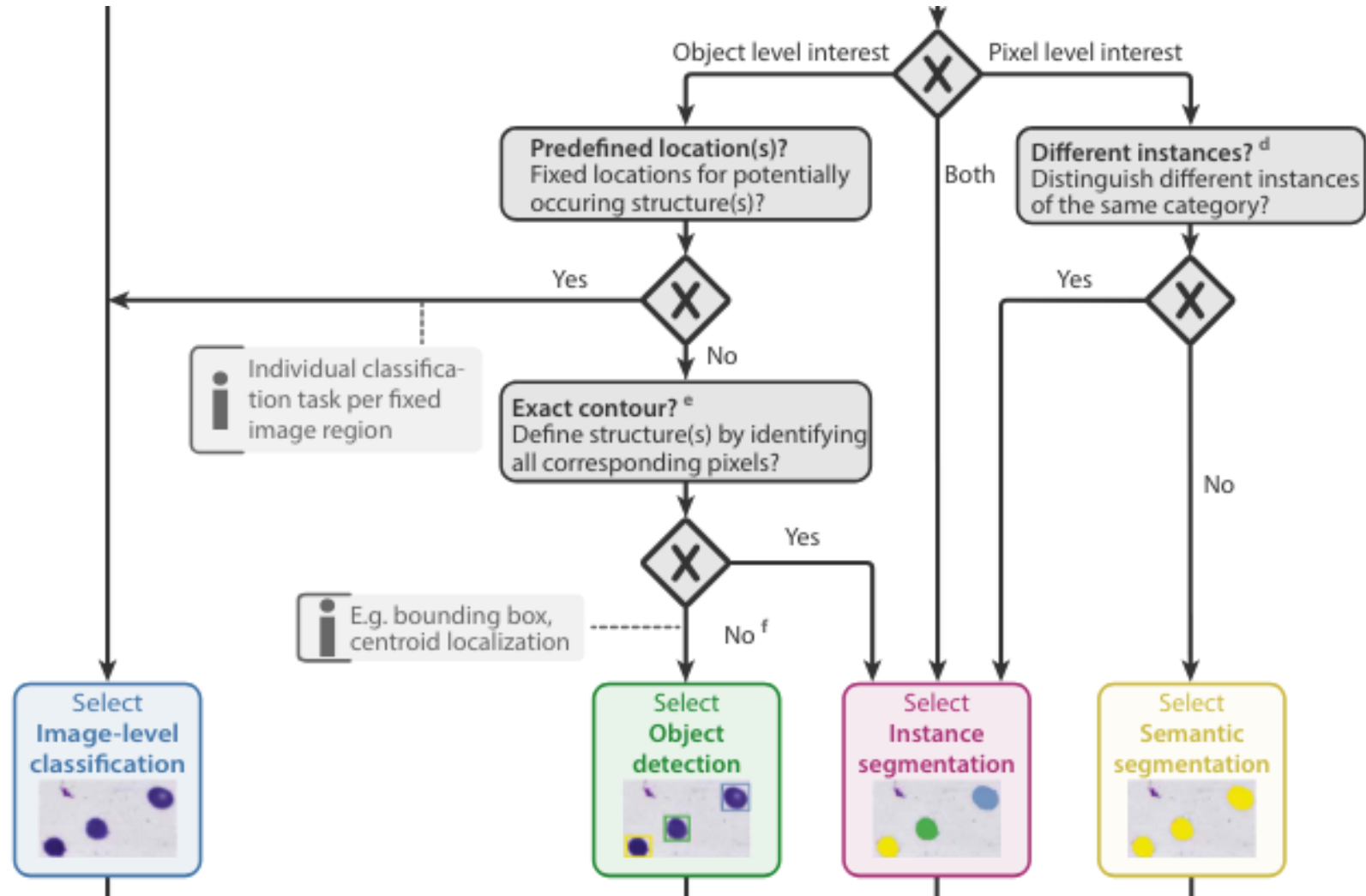
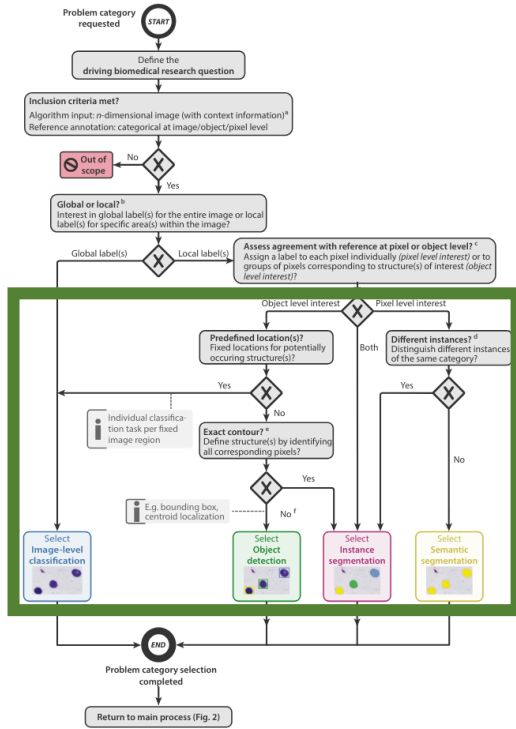
# Choosing the right metric is key

+ S1



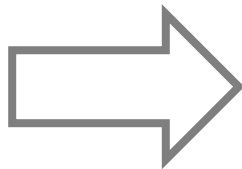
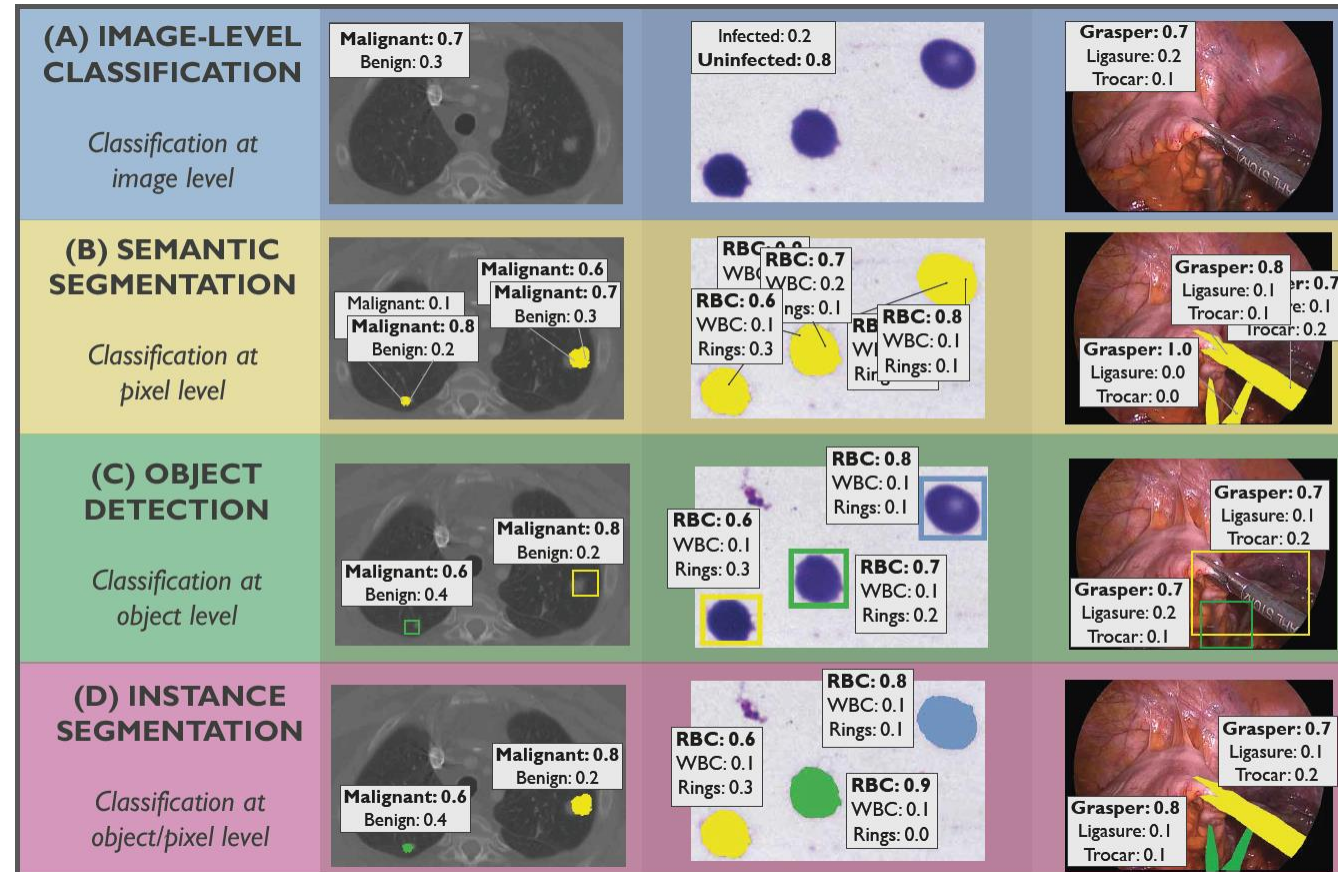
# Choosing the right metric is key

+ S1



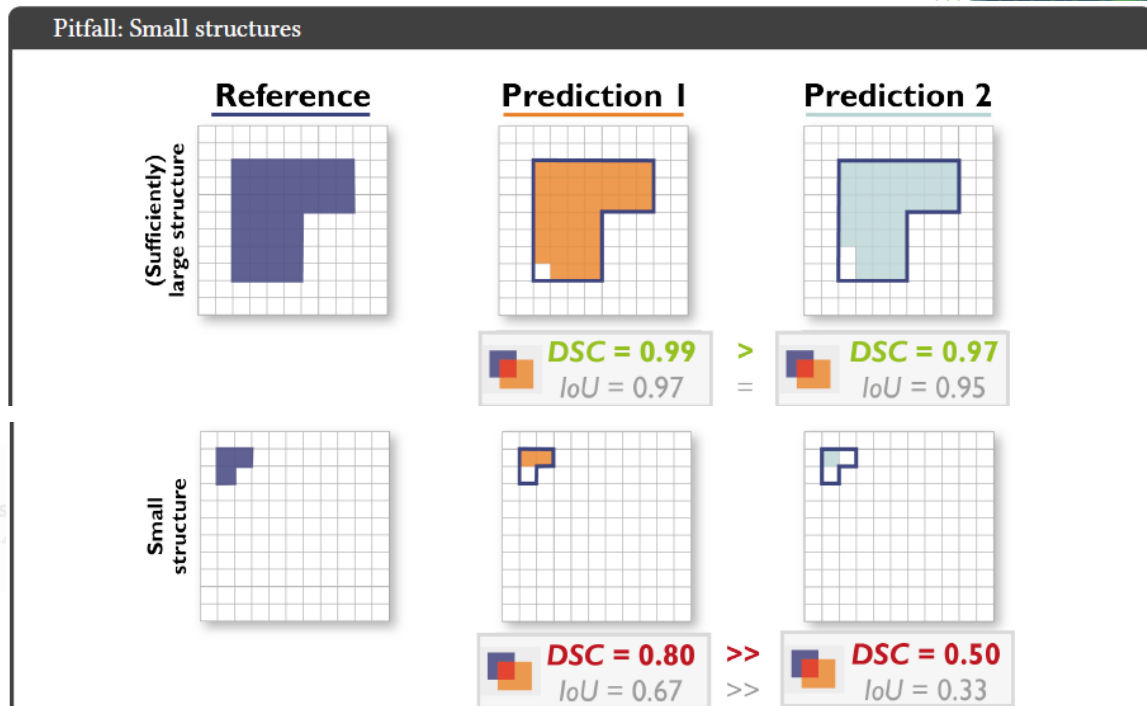
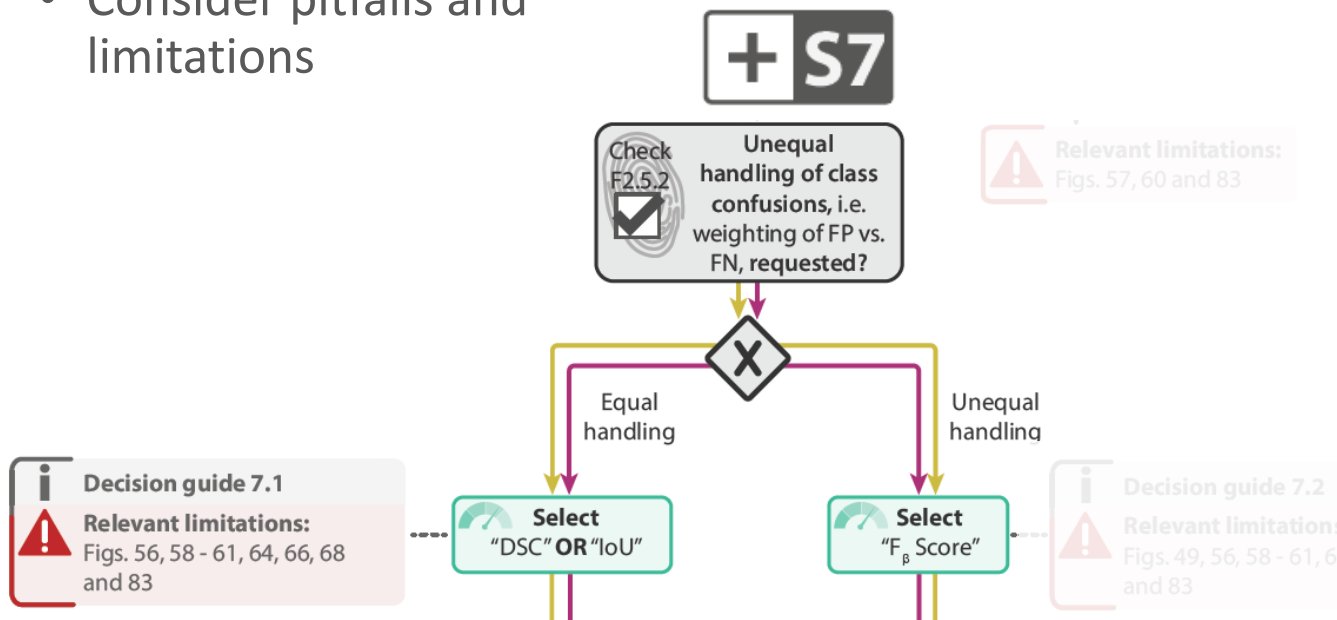
# Choosing the right metric is key

- Define your question



# Overlap metrics

- Consider pitfalls and limitations



(From Figure 56)

## E.6 Decision guide S7.

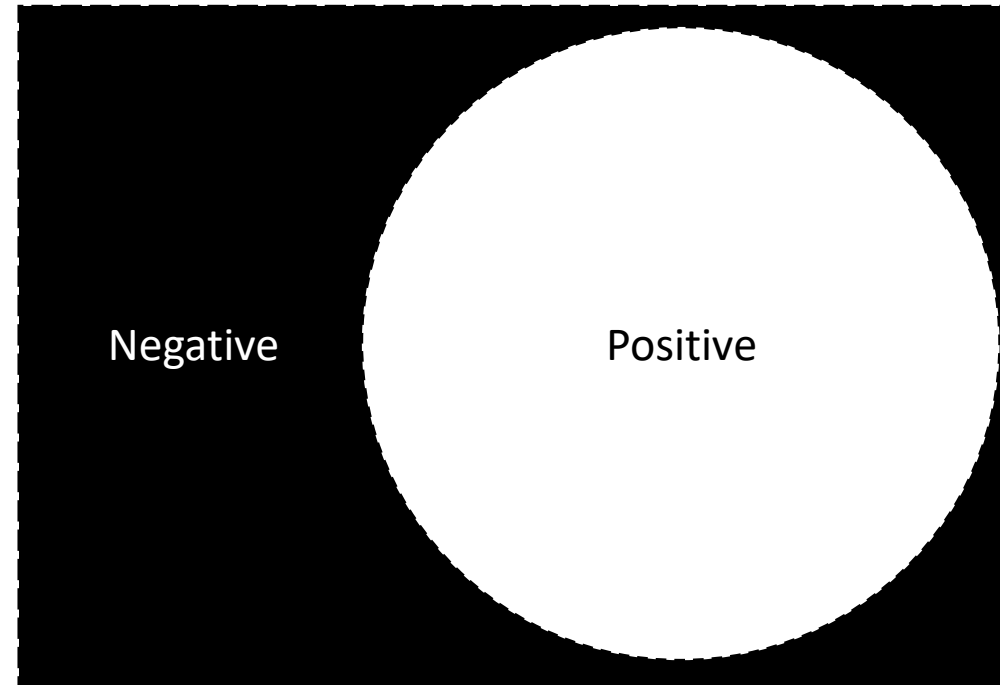
### D7.1: Dice vs IoU

The DSC is identical to the  $F_1$  Score on pixel level and closely related to the IoU, which, in turn, is identical to the Jaccard Index (see equations 5 and 6). The two metrics will yield the same ranking (of aggregated metric values) in most applications (theoretically, deviations are possible), such that there is no value in combining

them. Commonly, the computer vision community prefers the IoU, while the medical image community favors the DSC.

# Segmentation quality estimation

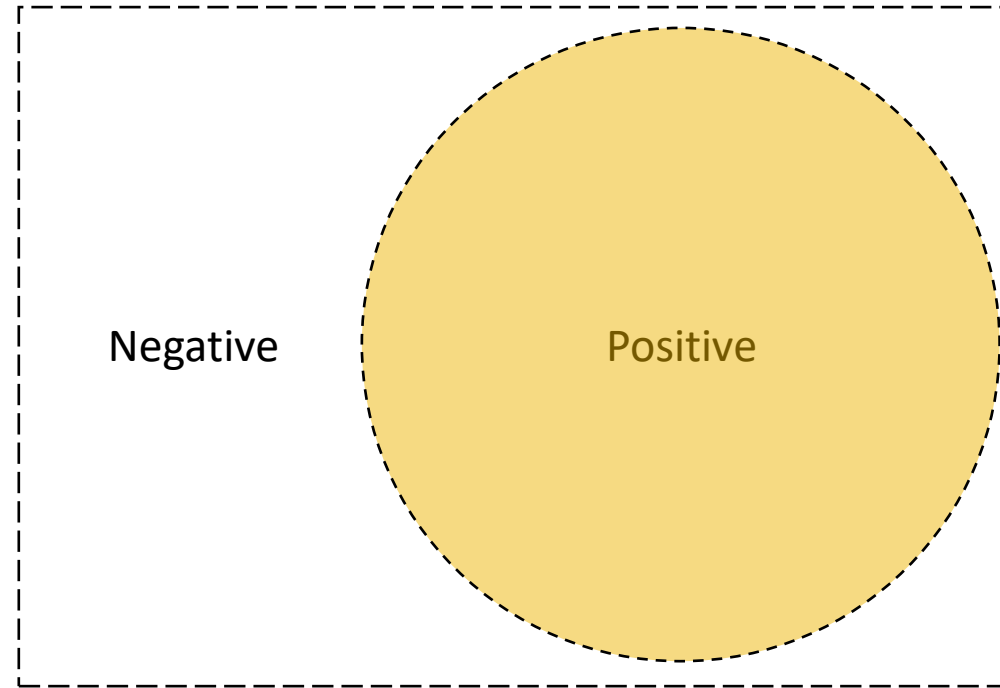
- In general
  - Define what's positive and what's negative.





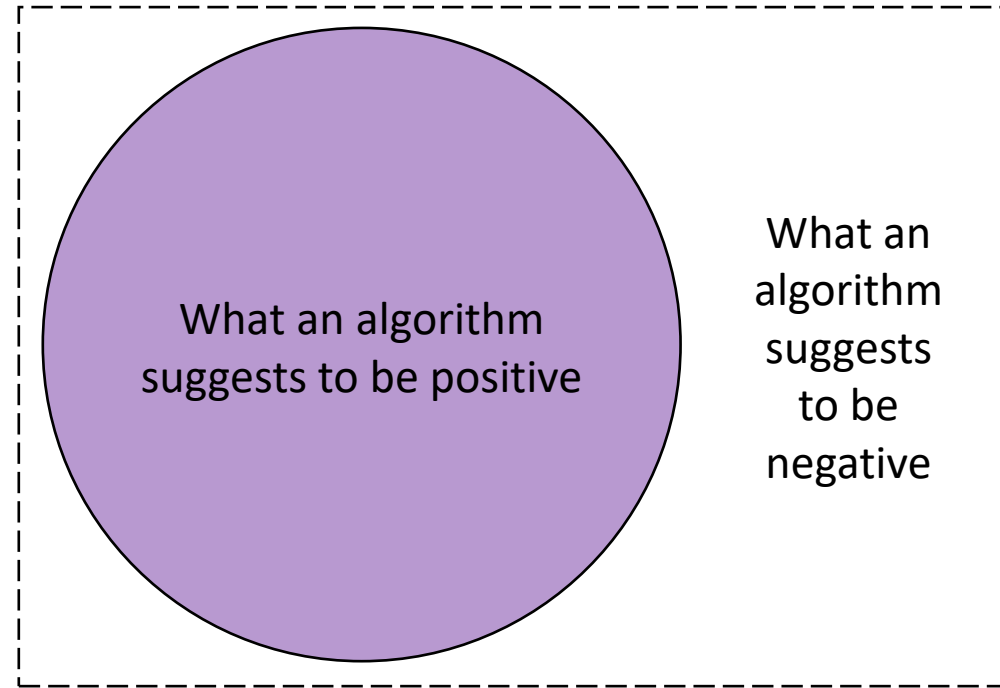
# Segmentation quality estimation

- In general
  - Define what's positive and what's negative.



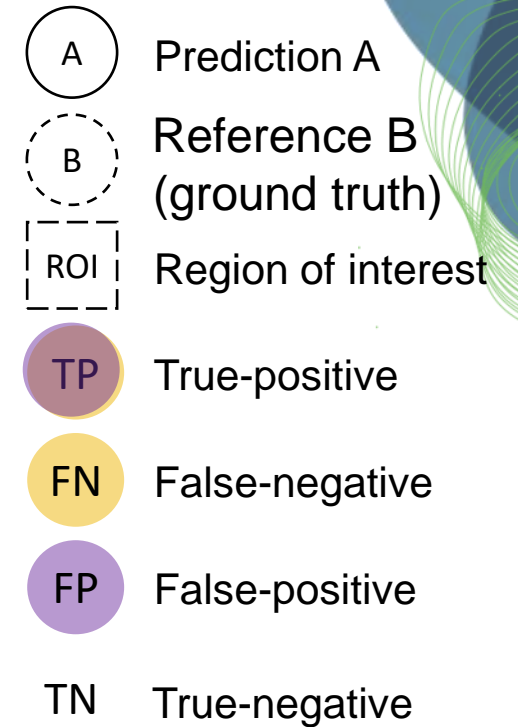
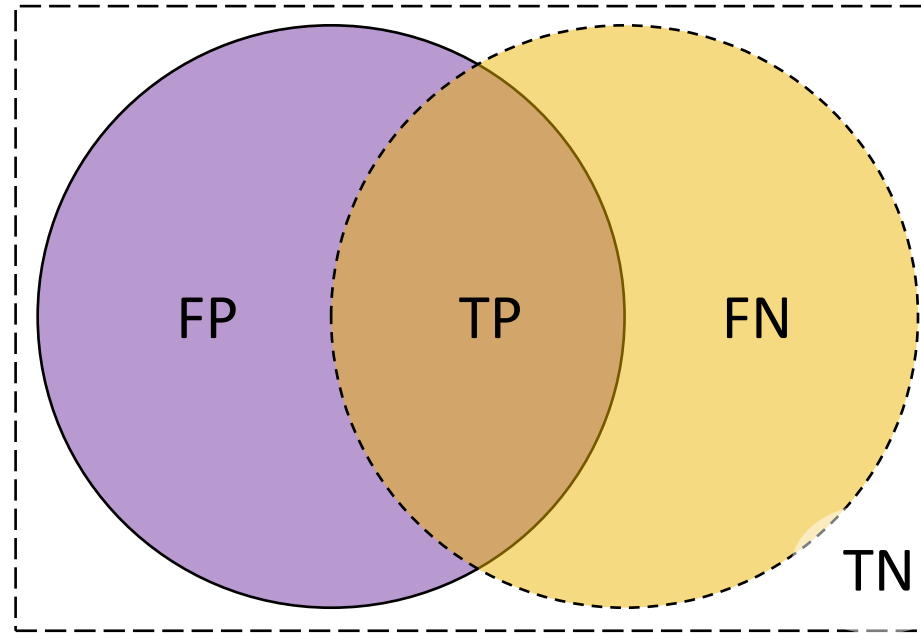
# Segmentation quality estimation

- In general
  - Define what's positive and what's negative.



# Segmentation quality estimation

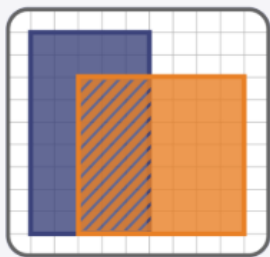
- In general
  - Define what's positive and what's negative.
  - Compare with a reference to figure out what was true and false
- Welcome to the Theory of Sets



# Dice versus Jaccard Index (IoU)

## DICE SIMILARITY COEFFICIENT (DSC)

Synonyms: Dice, Dice Coefficient, Sørensen–Dice Coefficient,  $F_1$  Score, Balanced F Score



$$\begin{aligned} \text{DSC}(A,B) &= \frac{2 |A \cap B|}{|A| + |B|} \\ &= \frac{2 \text{PPV} \cdot \text{Sensitivity}}{\text{PPV} + \text{Sensitivity}} \end{aligned}$$

■ A ■ B ■ A ∩ B

VALUE RANGE: [0, 1] ↑

### DESCRIPTION

DSC measures the overlap between two structures.

### DEFINITION

[Dice, 1945]

### RECOMMENDED FOR

ImLC  SemS  ObD  InS

### RECOMMENDATIONS

- An overlap-based metric (by default the DSC or IoU) should be used in most cases of segmentation assessment. An exception is the case of consistently tiny structures along with a noisy reference.
- DSC should generally be used in combination with a boundary-based metric if boundaries are of interest.
- DSC should generally not be considered if...
  - ... there is a high variability of structure sizes within an image or across images.
  - ... inter-rater variability is requested to be compensated.
  - ... over- and undersegmentation should be treated similarly.
- DSC should be considered as a metric in the medical community rather than in the computer vision and biology communities (where the almost identical IoU is preferred).

### IMPORTANT RELATIONS

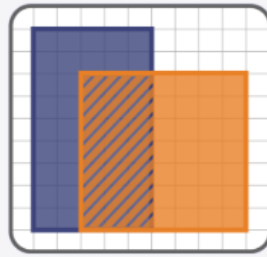
DSC is closely related to the IoU = Jaccard index:

$$\text{DSC} = \frac{2 \text{IoU}}{1 + \text{IoU}}$$

DSC is equal to the  $F_1$  Score ( $\beta = 1$  in  $F_\beta$  Score) at pixel level.

## INTERSECTION OVER UNION (IoU)

Synonyms: Jaccard Index, Tanimoto Coefficient



$$\begin{aligned} \text{IoU}(A,B) &= \frac{|A \cap B|}{|A| + |B| - |A \cap B|} \\ &= \frac{|A \cap B|}{|A \cup B|} \\ &= \frac{\text{PPV} \cdot \text{Sensitivity}}{\text{PPV} + \text{Sensitivity} - \text{PPV} \cdot \text{Sensitivity}} \end{aligned}$$

■ A ■ B ■ A ∩ B

VALUE RANGE: [0, 1] ↑

### DESCRIPTION

IoU measures the overlap between two structures. It is often referred to as **Box IoU** when comparing bounding boxes, **Mask IoU** when comparing segmentation masks, or **Approx IoU** when comparing approximations of objects beyond bounding boxes.

### DEFINITION

[Jaccard, 1912]

### RECOMMENDATIONS

- An overlap-based metric (by default DSC or IoU) should be used in most cases for segmentation assessment. An exception is the case of consistently tiny structures along with a noisy reference.
- IoU should generally be used in combination with a boundary-based metric if boundaries are of interest.
- IoU should generally not be considered if...
  - ... there is a high variability of structure sizes within an image or across images.
  - ... inter-rater variability is requested to be compensated.
  - ... over- and undersegmentation should be treated similarly.
- IoU should be considered as a metric in the computer vision and biology communities rather than in the medical community (which prefers the almost identical DSC).

### RECOMMENDED FOR

ImLC  SemS  ObD  InS

### IMPORTANT RELATIONS

$$\begin{aligned} \text{IoU} &= \frac{\text{DSC}}{2 - \text{DSC}} & \text{IoU} &= \frac{F_\beta}{2 - F_\beta} \\ & & & \text{for } \beta = 1 \end{aligned}$$

# Dice versus Jaccard Index (IoU)

## 2.7.5 Decision guide S6.

### DG6.1: Dice Similarity Coefficient (DSC) versus Intersection over Union (IoU)

#### Summary of DG6.1: DSC versus IoU

##### DSC

- ➔ Identical to  $F_1$  Score
- ➔ Close relation to IoU (see Eq. 5)
- ➔ Preference in medical community

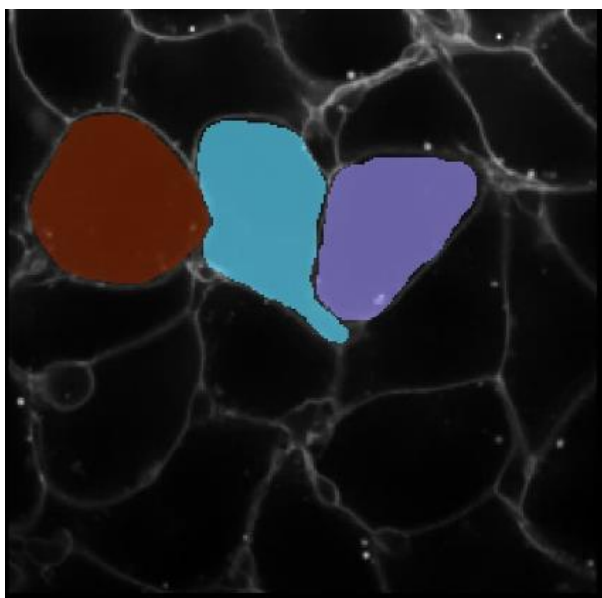
##### IoU

- ➔ Identical to Jaccard Index
- ➔ Close relation to DSC (see Eq. 4)
- ➔ Preference in computer vision community

Extended Data Tab. SN 2.18. Comparison of Dice Similarity Coefficient (DSC) and Intersection over Union (IoU) in the context of the decision guide DG6.1 for Subprocess S6. Context: no exclusive interest in the center line of structures (FP2.3 = FALSE, FP3.3 = FALSE) and equal severity of class confusions (FP2.5.2 = FALSE).

# Sparse Jaccard Index

- For every annotated object, we compute the maximum IoU with any segmented object.
- We average this value over all annotated objects



Sparse  
instance annotation

IoU = 0.35

IoU = 0.66

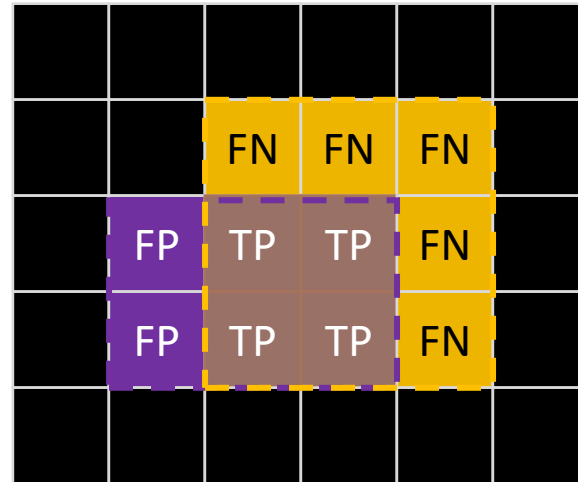
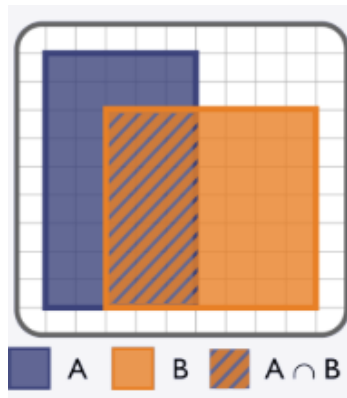
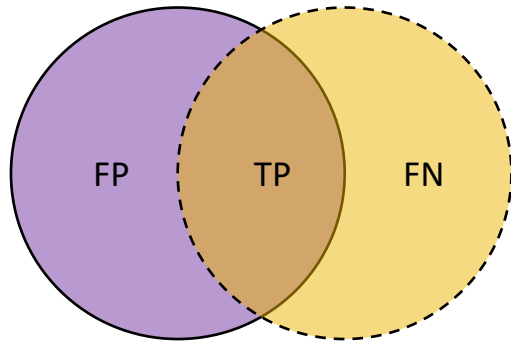
IoU = 0.69

Instance segmentation candidates

# Pixel-wise versus Object-wise evaluation

- Pixel wise: Segmentation quality

Prediction      Ground truth



True-positive: 4

False-negative: 5

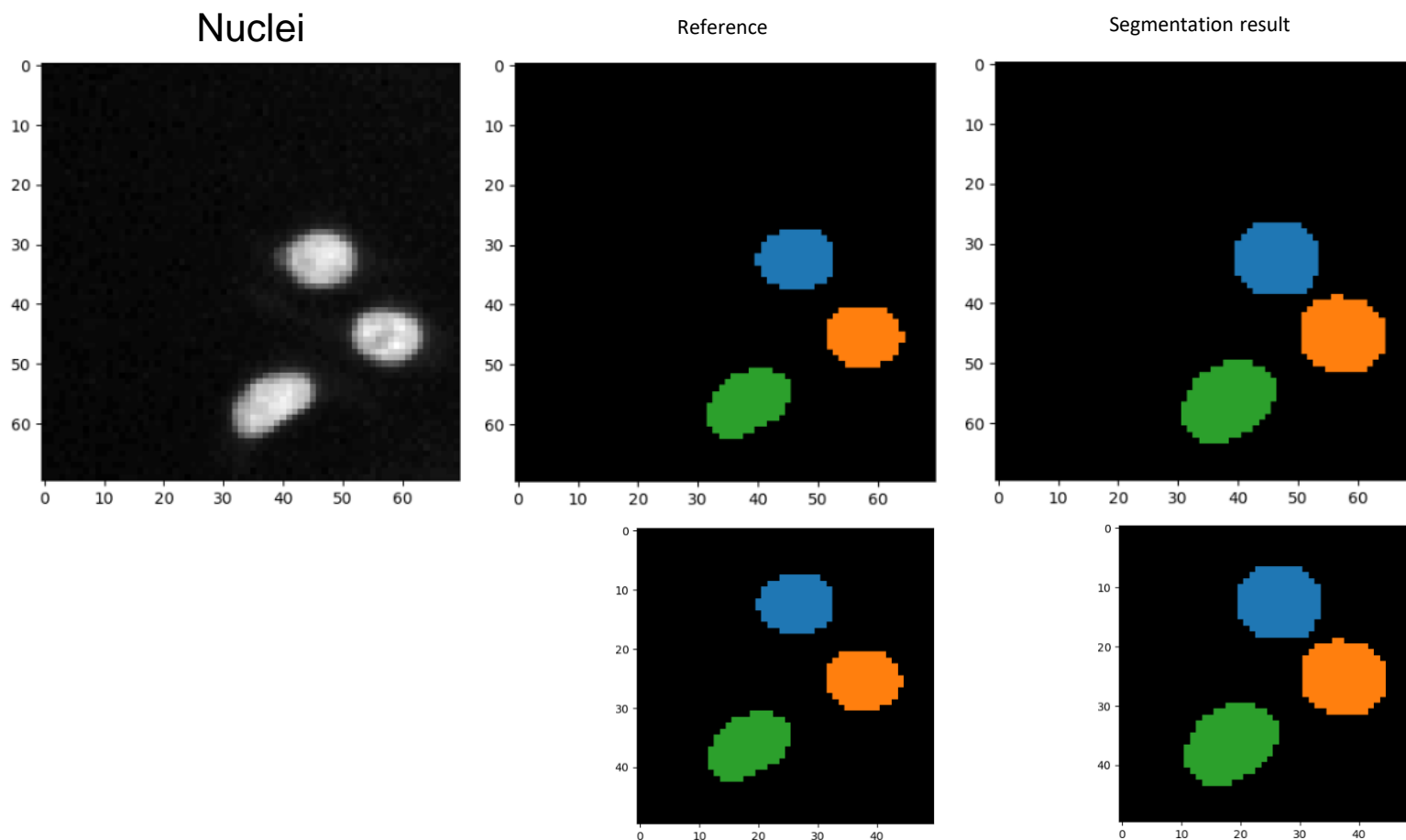
False-positive: 2

$$IoU(A,B) = \frac{|A \cap B|}{|A| + |B| - |A \cap B|} = \frac{|A \cap B|}{|A \cup B|}$$

$$IoU = 4 / 11$$

# Accuracy versus Jaccard Index (IoU)

- Side-effects of image size and number of nuclei



$$A = \frac{TP + TN}{FN + FP + TP + TN}$$

$$J = \frac{TP}{FN + FP + TP}$$

Accuracy: 0.97

Jaccard Index: 0.73

Accuracy decreases because there are less correct black pixels (TN)

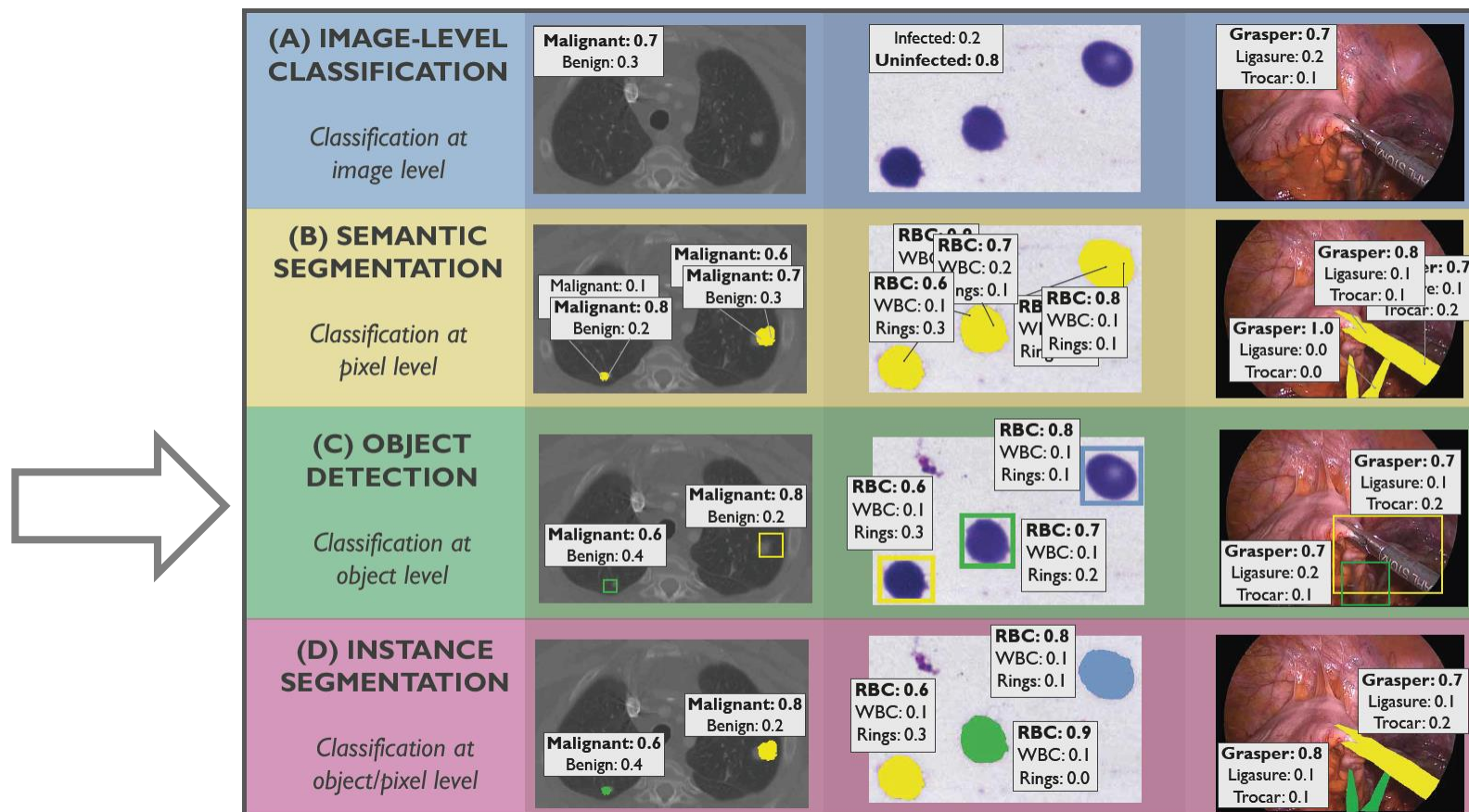
Accuracy: 0.95

Jaccard Index: 0.73



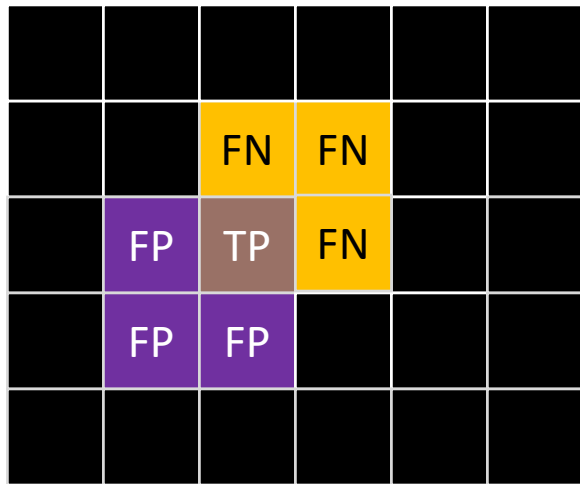
# Choosing the right metric is key

- Define your question

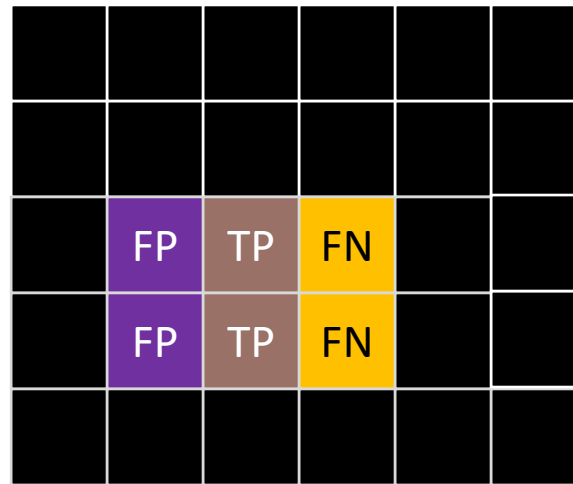


# Pixel-wise versus Object-wise evaluation

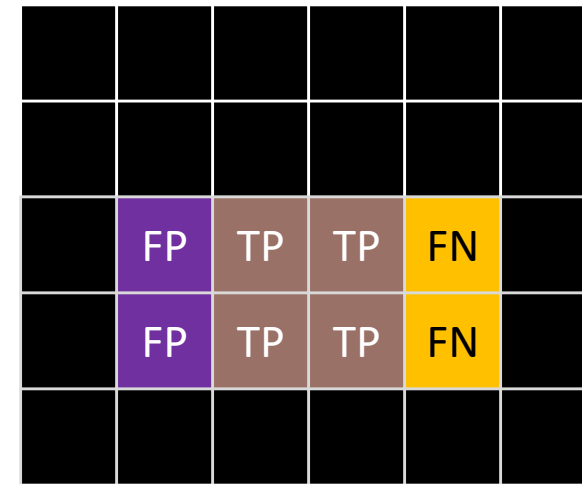
- Are these objects overlapping?



$$\text{IoU} = 1 / 7$$



$$\text{IoU} = 1 / 3$$

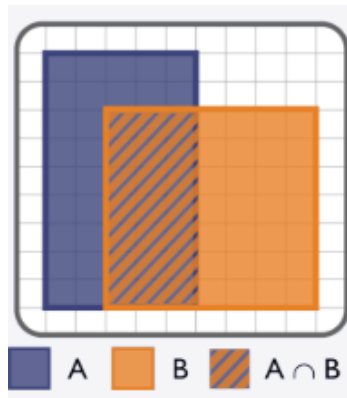
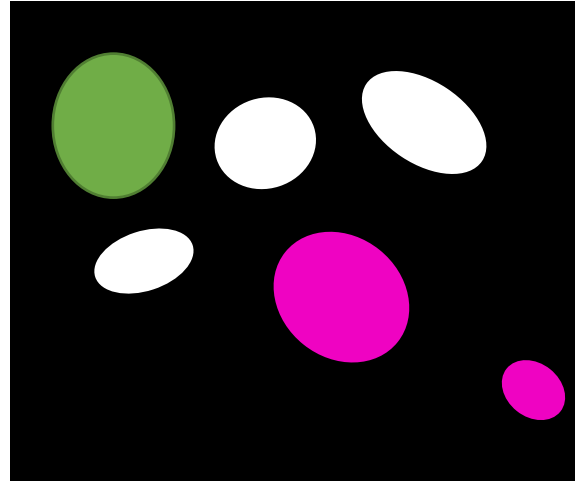
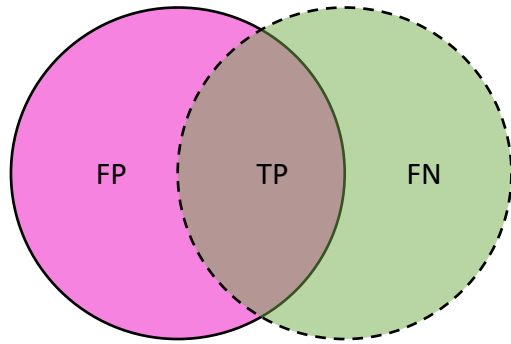


$$\text{IoU} = 1 / 2$$

# Pixel-wise versus Object-wise evaluation

- Object wise: Detection quality

Prediction Ground truth



True-positive: 3

False-negative: 1

False-positive: 2

$$\text{IoU}(A,B) = \frac{\text{hatched square}}{\text{blue square} + \text{orange square} - \text{hatched square}}$$
$$= \frac{|A \cap B|}{|A| + |B| - |A \cap B|} = \frac{|A \cap B|}{|A \cup B|}$$

$$\text{IoU} = 1 / 2$$

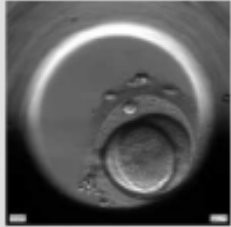

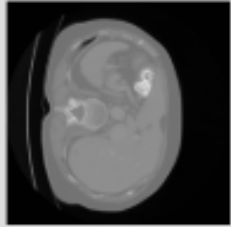

# Pixel-wise versus Object-wise evaluation

- Play with metrics to gain understanding



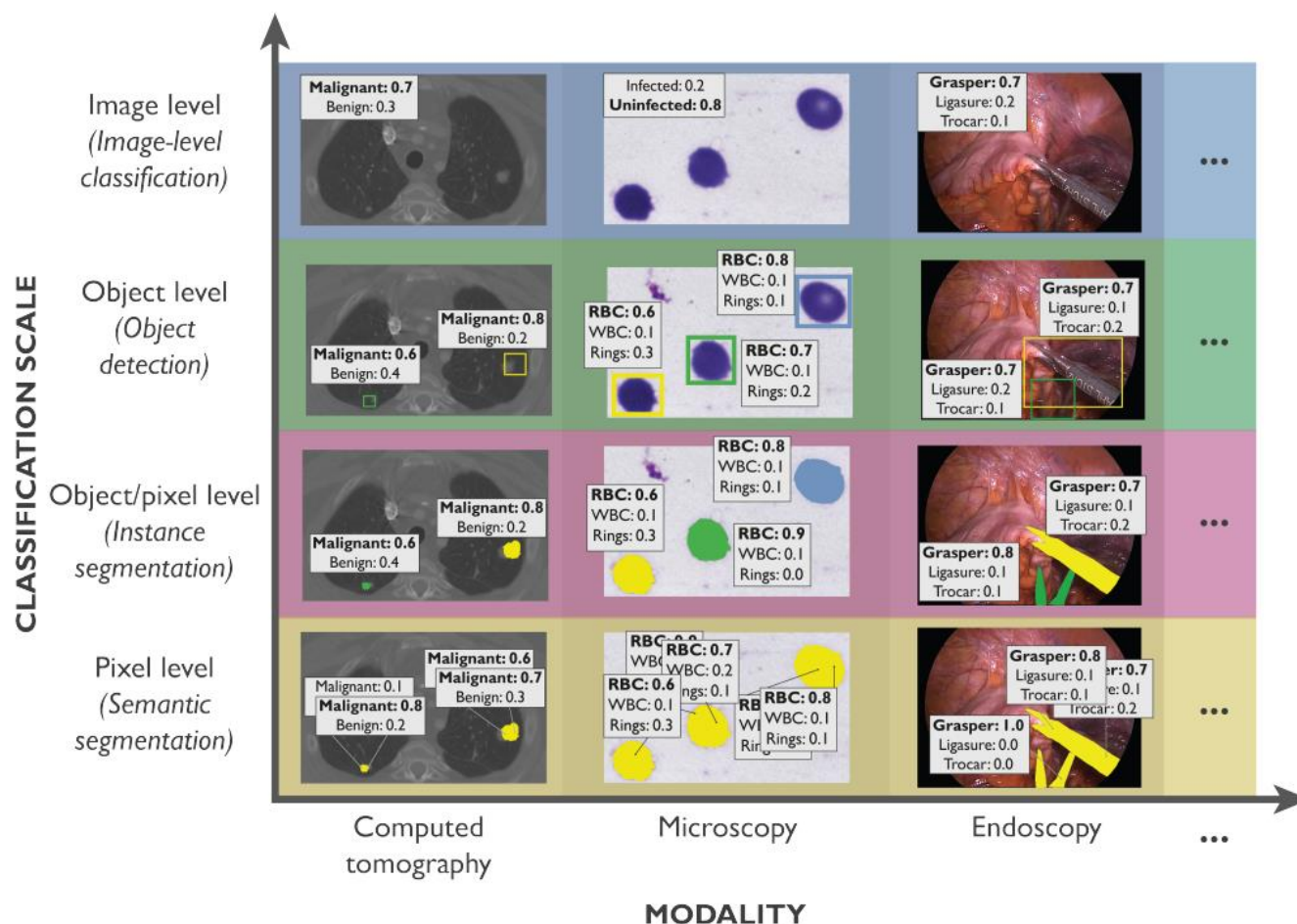
With Martin Schätz (Charles Uni, Prague)  
@schatzcz

- Special case: We elaborate segmentation quality of one / large object:

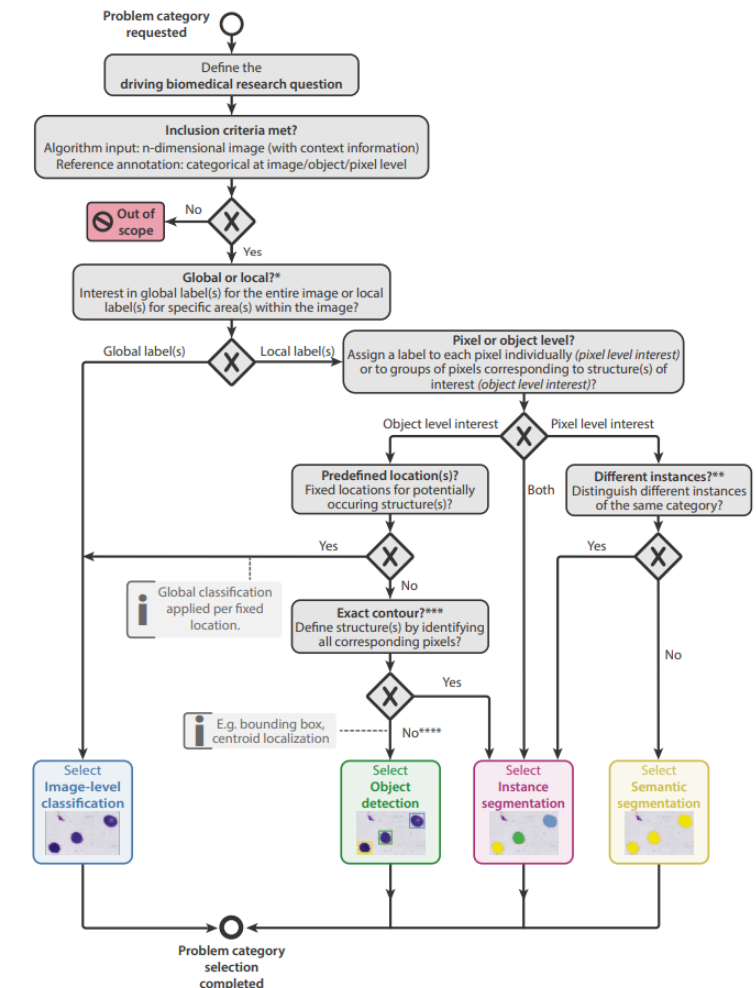
PROBLEM DESCRIPTION	ID	SCENARIO	SAMPLE INPUT IMAGE	RECOMMENDED OUPUT	RECOMMENDATION
Segmentation of large objects	SemS-1	Embryo segmentation from microscopy images			<b>Problem category:</b> <b>Semantic segmentation</b>  <b>Overlap-based metric (S6):</b> Dice Similarity Coefficient (DSC)  <b>Boundary-based metric (S7):</b> Normalized Surface Distance (NSD)  <b>Specific property-related metric:</b> Liver segmentation: Absolute Volume Difference
	SemS-2	Liver segmentation in computed tomography (CT) images			

# What metric to use when?

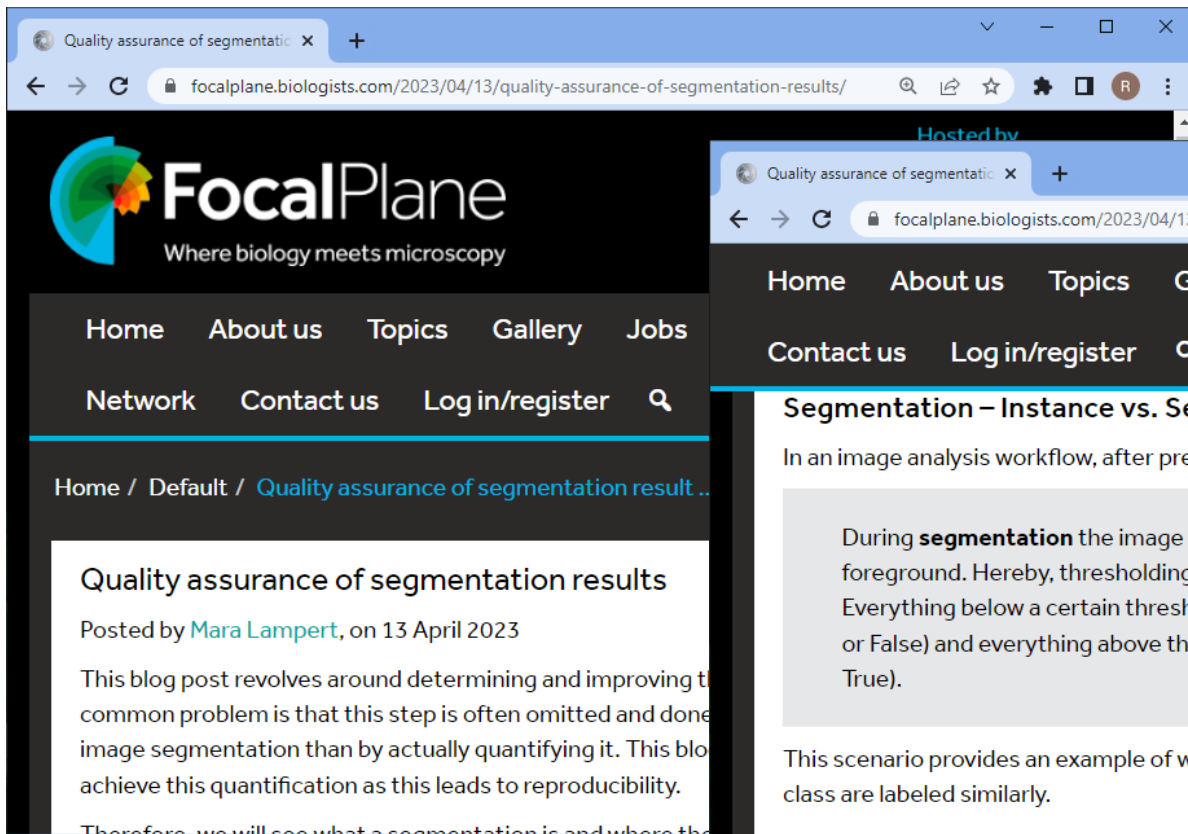
- “Metrics reloaded: Pitfalls and recommendations for image analysis validation”  
Maier-Hein, Reinke et al. <https://arxiv.org/abs/2206.01653>



**+ S1**



# Further reading



Quality assurance of segmentatic x +

focalplane.biologists.com/2023/04/13/quality-assurance-of-segmentation-results/

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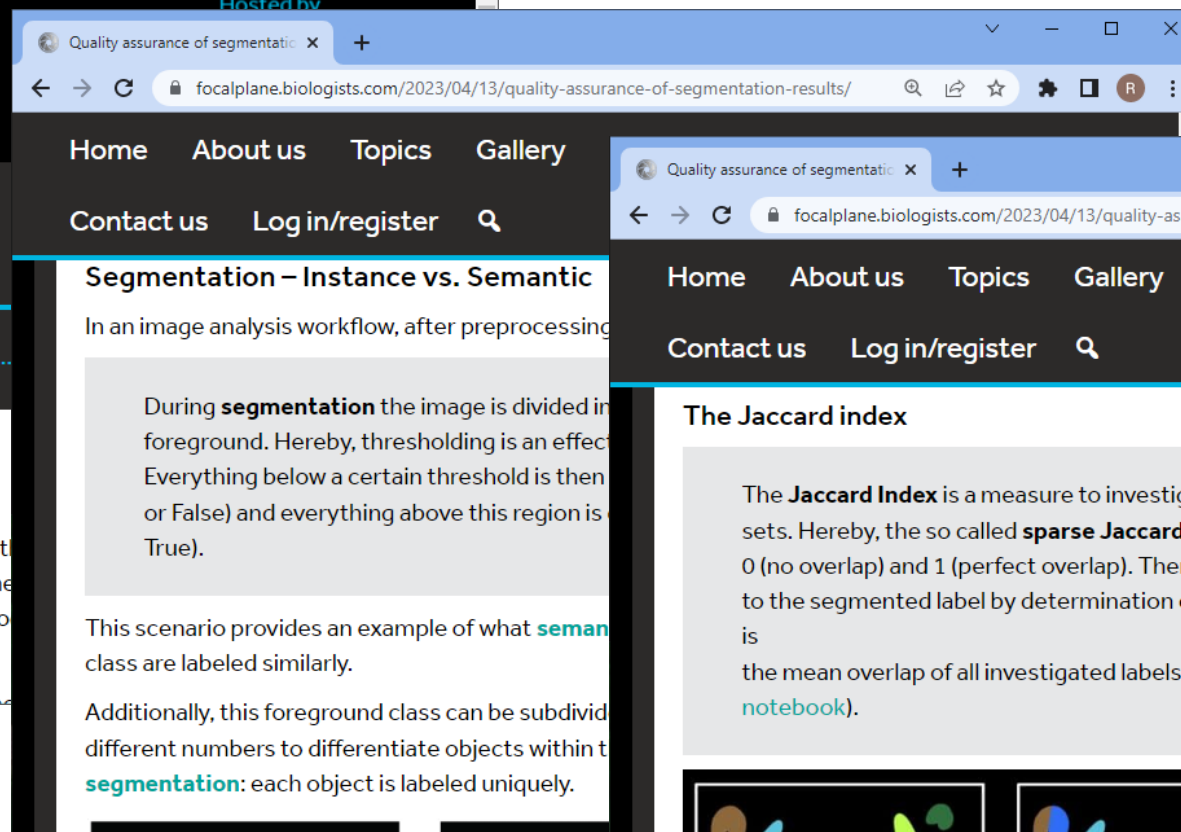
Home / Default / [Quality assurance of segmentation result...](#)

## Quality assurance of segmentation results

Posted by [Mara Lampert](#), on 13 April 2023

This blog post revolves around determining and improving the quality of image segmentation. A common problem is that this step is often omitted and done visually rather than by actually quantifying it. This blog aims to achieve this quantification as this leads to reproducibility.

Therefore, we will see what a segmentation is and where the



Quality assurance of segmentatic x +

focalplane.biologists.com/2023/04/13/quality-assurance-of-segmentation-results/

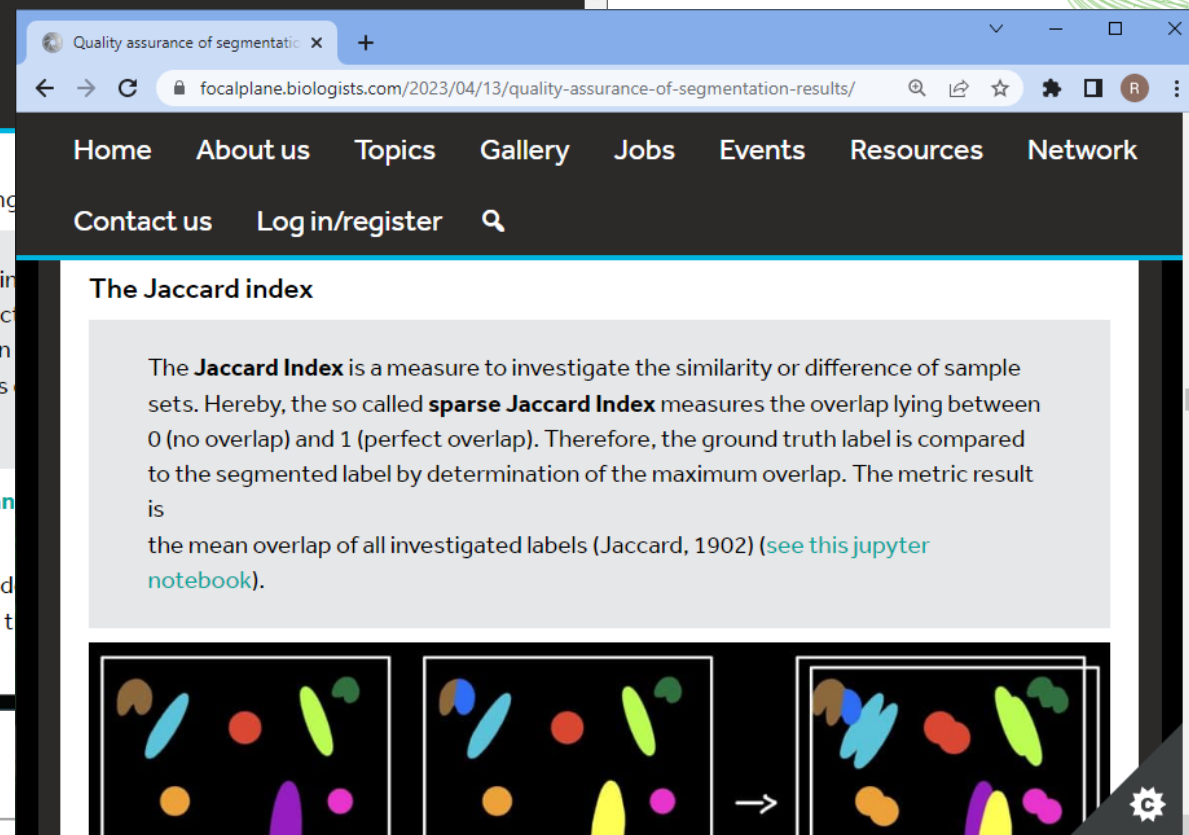
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## Segmentation – Instance vs. Semantic

In an image analysis workflow, after preprocessing the image, the next step is segmentation. During **segmentation** the image is divided into foreground and background. Hereby, thresholding is an effective method. Everything below a certain threshold is then considered as background (or False) and everything above this region is considered as foreground (or True).

This scenario provides an example of what **semantic segmentation** class are labeled similarly.

Additionally, this foreground class can be subdivided into different numbers to differentiate objects within the image. This is called **instance segmentation**: each object is labeled uniquely.



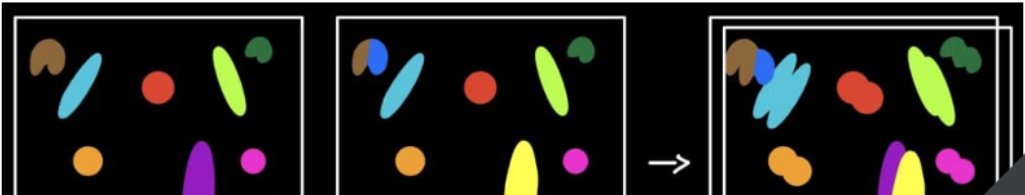
Quality assurance of segmentatic x +

focalplane.biologists.com/2023/04/13/quality-assurance-of-segmentation-results/

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## The Jaccard index

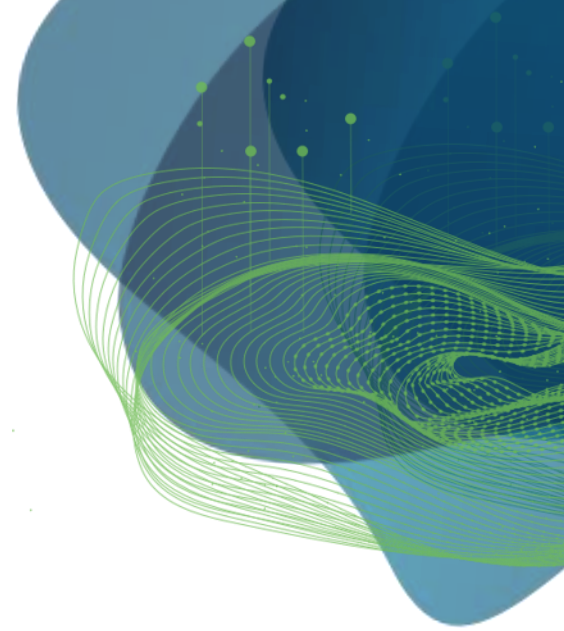
The **Jaccard Index** is a measure to investigate the similarity or difference of sample sets. Hereby, the so called **sparse Jaccard Index** measures the overlap lying between 0 (no overlap) and 1 (perfect overlap). Therefore, the ground truth label is compared to the segmented label by determination of the maximum overlap. The metric result is the mean overlap of all investigated labels (Jaccard, 1902) (see [this jupyter notebook](#)).



# Feature extraction

Robert Haase

Using materials from Johannes Soltwedel, PoL, TU Dresden



GEFÖRDERT VOM



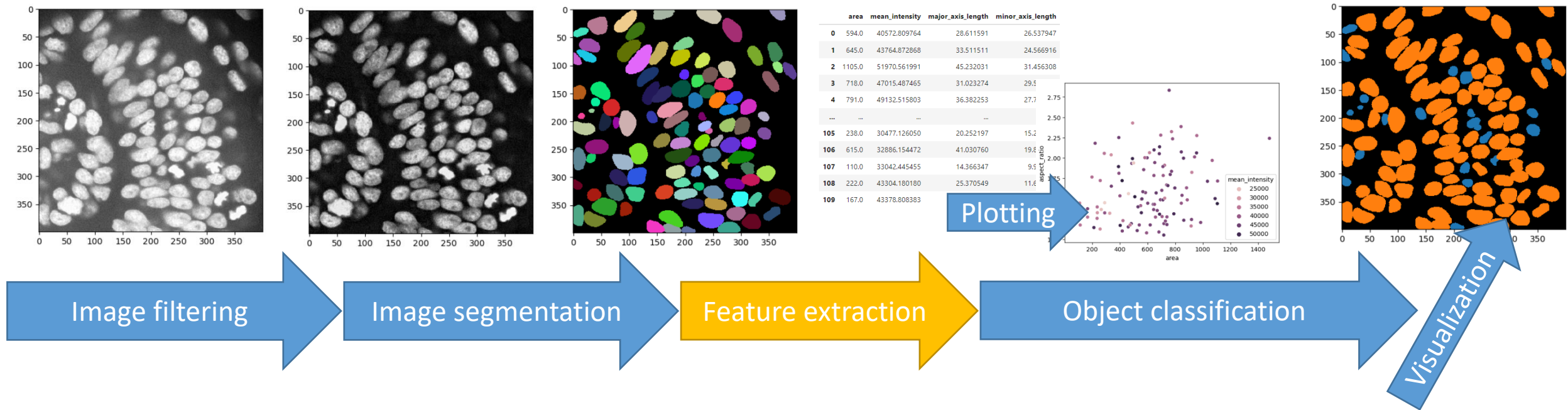
Bundesministerium  
für Bildung  
und Forschung

Diese Maßnahme wird gefördert durch die Bundesregierung aufgrund eines Beschlusses des Deutschen Bundestages. Diese Maßnahme wird mitfinanziert durch Steuermittel auf der Grundlage des von den Abgeordneten des Sächsischen Landtags beschlossenen Haushaltes.



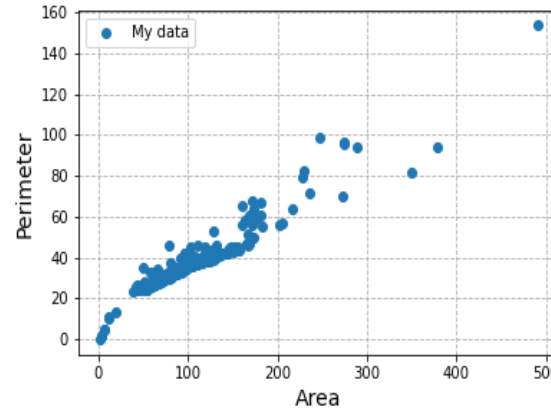
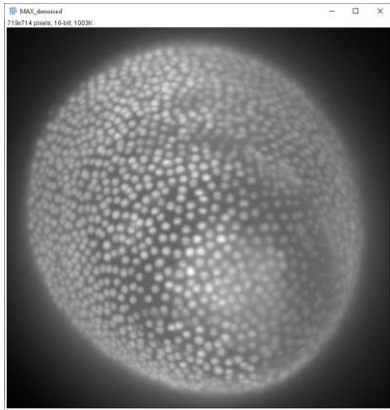
# Lecture overview: Bio-image Analysis

- Image Data Analysis workflows
- Goal: Quantify observations, substantiate conclusions with numbers

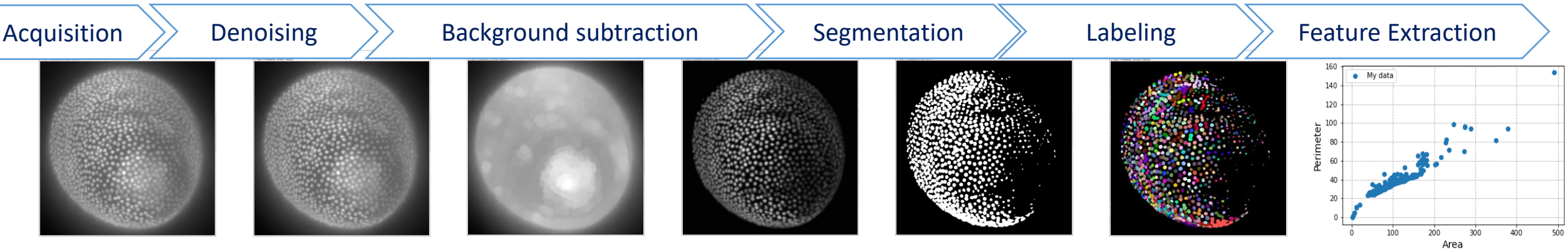


# Feature extraction

- Feature extraction is a *late* processing step in image analysis.
- It can be used for images or

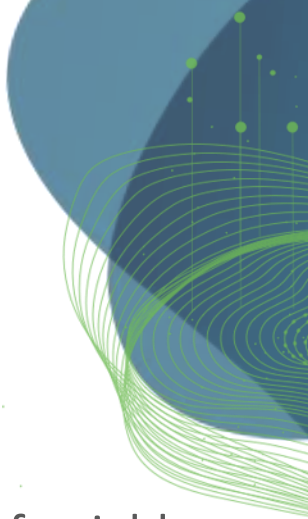


- or segmented/labelled images



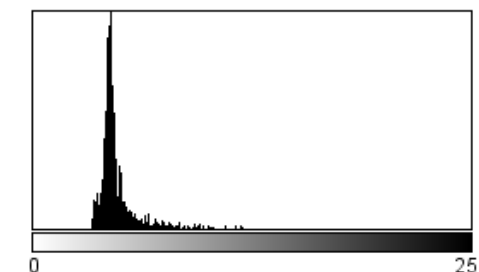
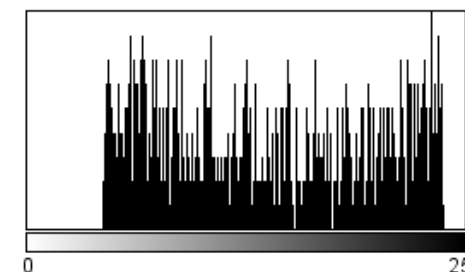
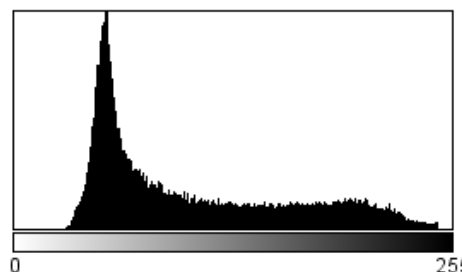
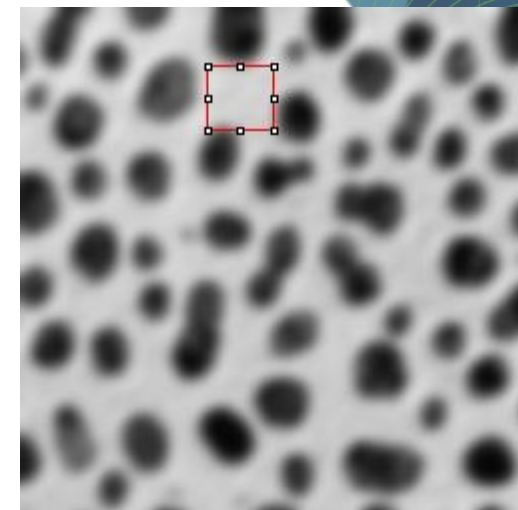
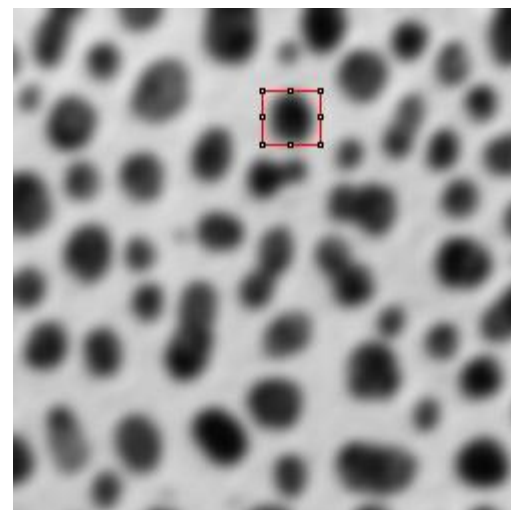
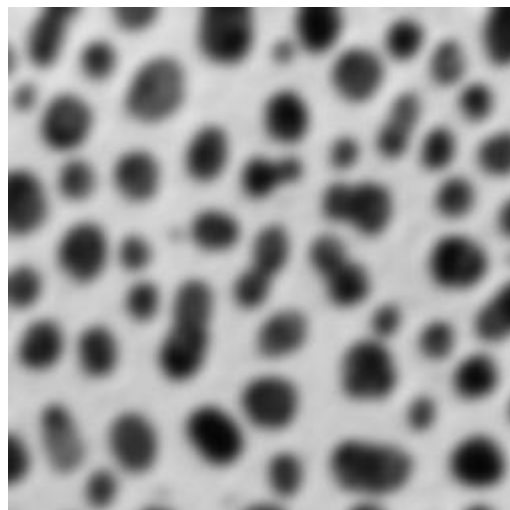
# Feature extraction

- A *feature* is a countable or measurable property of an image or object.
- Goal of feature extraction is finding a minimal set of features to describe an object well enough to differentiate it from other objects.
- **Intensity based**
  - Mean intensity
  - Standard deviation
  - Total intensity
  - Textures
- **Shape based /spatial**
  - Area / Volume
  - Roundness
  - Solidity
  - Circularity / Sphericity
  - Elongation
  - Centroid
  - Bounding box
- **Spatio-temporal**
  - Displacement,
  - Speed,
  - Acceleration
- **Topological**
  - Number of neighbors
- **Others**
  - Overlap
  - Colocalization
- **Mixed features**
  - Center of mass
  - Local minima / maxima
  - Distance to neighbors
  - Average intensity in neighborhood



# Intensity based features

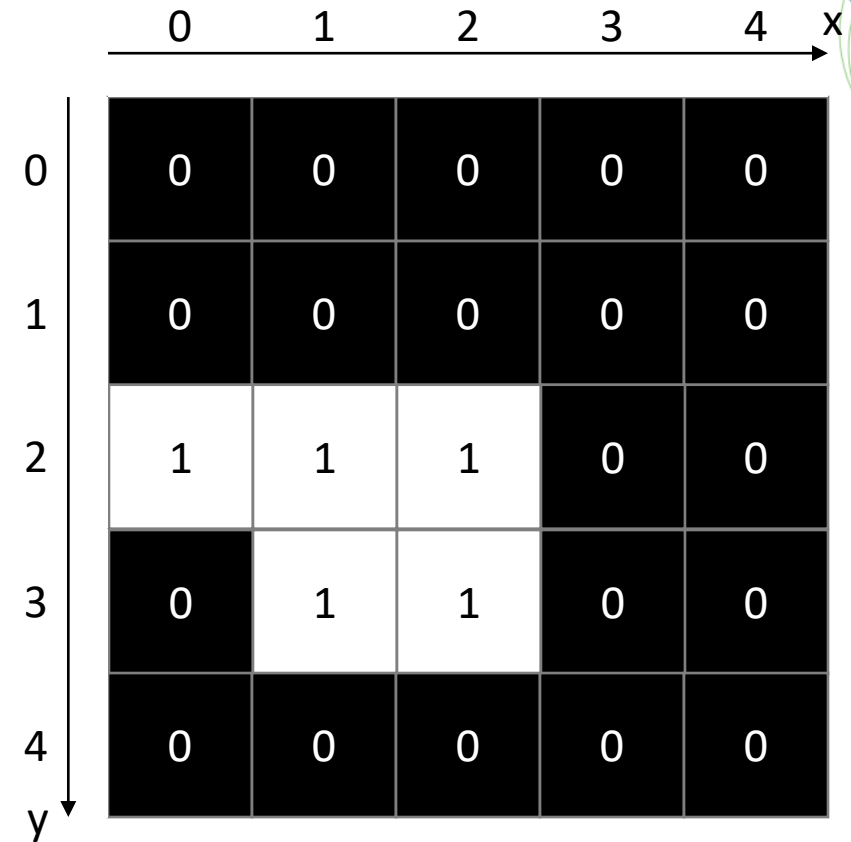
- Min / max
  - Median
  - Mean
  - Mode
  - Variance
  - Standard deviation
- 
- Can be derived from pixel values
  - Don't take spatial relationship of pixels into account
- 
- See also:
    - descriptive statistics
    - histogram



# Bounding rectangle / bounding box

- Position and size of the smallest rectangle containing all pixels of an object
  - $x_b, y_b$  ... position of the bounding box
  - $w_b$  ... width of the bounding box
  - $h_b$  ... height of the bounding box

variable	value
$x_b$	0
$y_b$	2
$w_b$	3
$h_b$	2



# Center of mass

- Relative position in an image weighted by pixel intensities

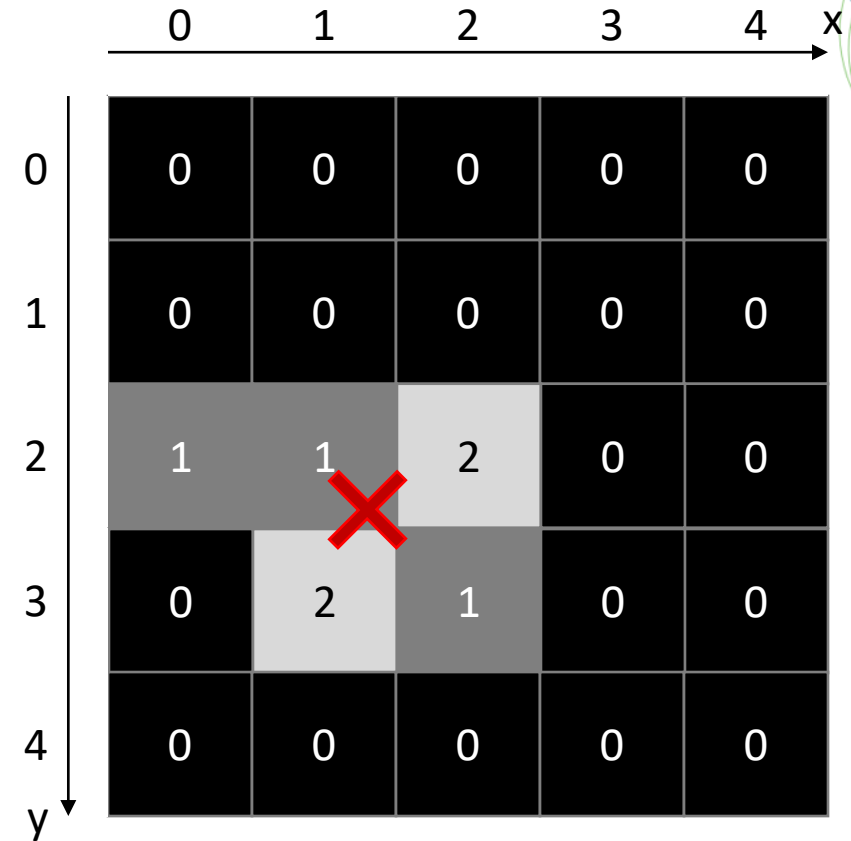
- x, y ... pixel coordinates
- w ... image width
- h ... image height
- $\mu$  ... mean intensity
- $g_{x,y}$  ... pixel grey value
- $x_m, y_m$  ... center of mass coordinates

$$\mu = \frac{1}{wh} \sum_{y=0}^{h-1} \sum_{x=0}^{w-1} g_{x,y}$$

$$x_m = \frac{1}{wh\mu} \sum_{y=0}^{h-1} \sum_{x=0}^{w-1} x g_{x,y}$$

$$y_m = \frac{1}{wh\mu} \sum_{y=0}^{h-1} \sum_{x=0}^{w-1} y g_{x,y}$$

“sum intensity”  
“total intensity”



$$x_m = 1/7 (1 \cdot 0 + 1 \cdot 1 + 2 \cdot 2 + 2 \cdot 1 + 1 \cdot 2) = 1.3$$

$$y_m = 1/7 (1 \cdot 2 + 1 \cdot 2 + 2 \cdot 3 + 2 \cdot 2 + 1 \cdot 3) = 2.4$$

# Center of geometry / centroid

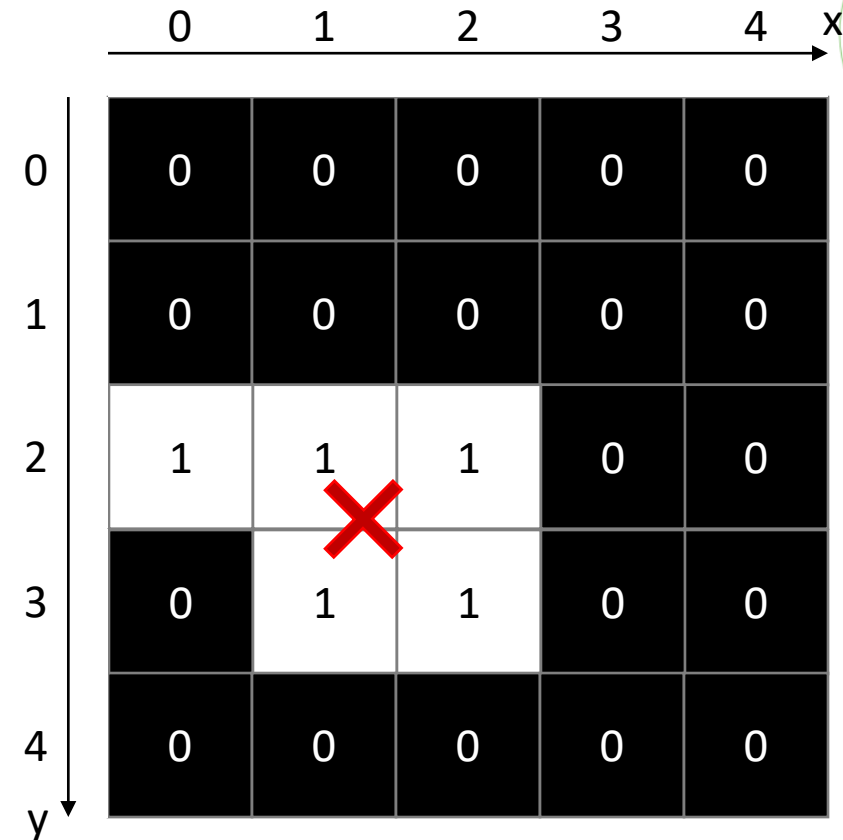
- Relative position in an image weighted by pixel intensities
- Special case of center of mass for binary images
  - $x, y$  ... pixel coordinates
  - $w$  ... image width
  - $h$  ... image height
  - $\mu$  ... mean intensity
  - $g_{x,y}$  ... pixel grey value, integer in range [0;1]
  - $x_m, y_m$  ... center of mass coordinates

$$\mu = \frac{1}{wh} \sum_{y=0}^{h-1} \sum_{x=0}^{w-1} g_{x,y}$$

$$x_m = \frac{1}{wh\mu} \sum_{y=0}^{h-1} \sum_{x=0}^{w-1} x g_{x,y}$$

$$y_m = \frac{1}{wh\mu} \sum_{y=0}^{h-1} \sum_{x=0}^{w-1} y g_{x,y}$$

Number of white pixels

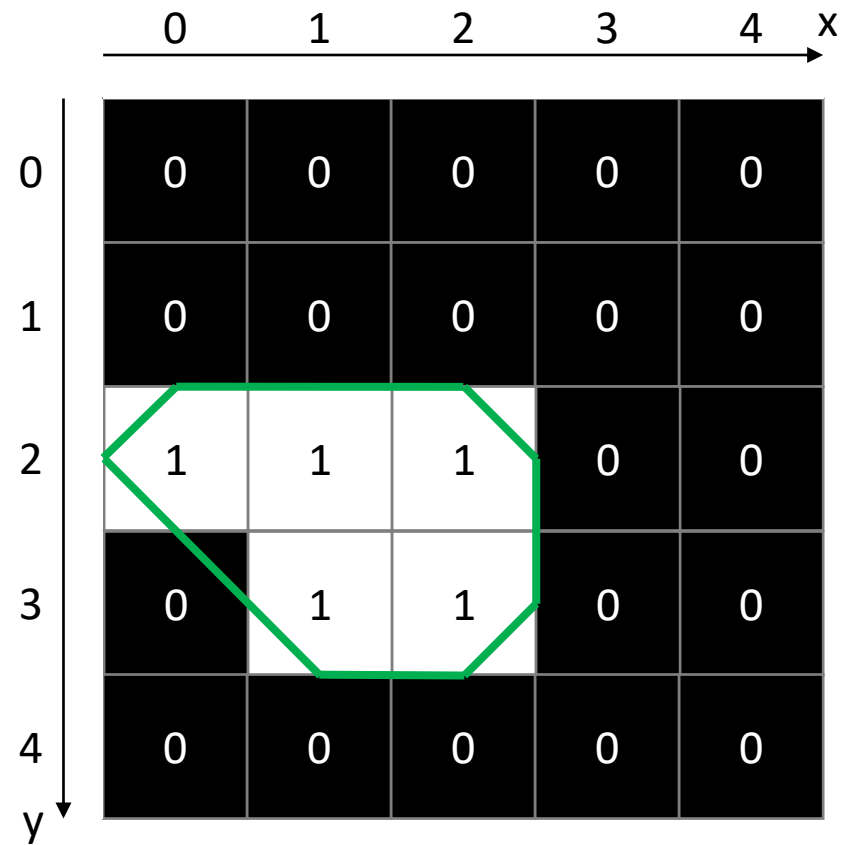
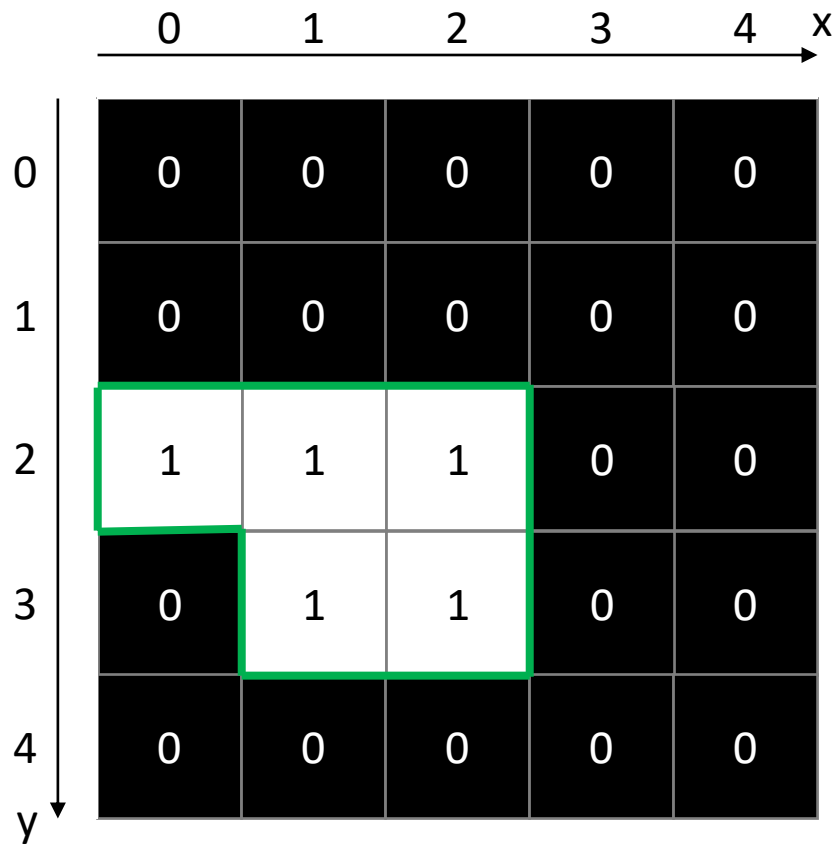


$$x_m = 1/5 (1 \cdot 0 + 1 \cdot 1 + 1 \cdot 2 + 1 \cdot 1 + 1 \cdot 2) = 1.2$$

$$y_m = 1/5 (1 \cdot 2 + 1 \cdot 2 + 1 \cdot 3 + 1 \cdot 2 + 1 \cdot 3) = 2.4$$

# Perimeter

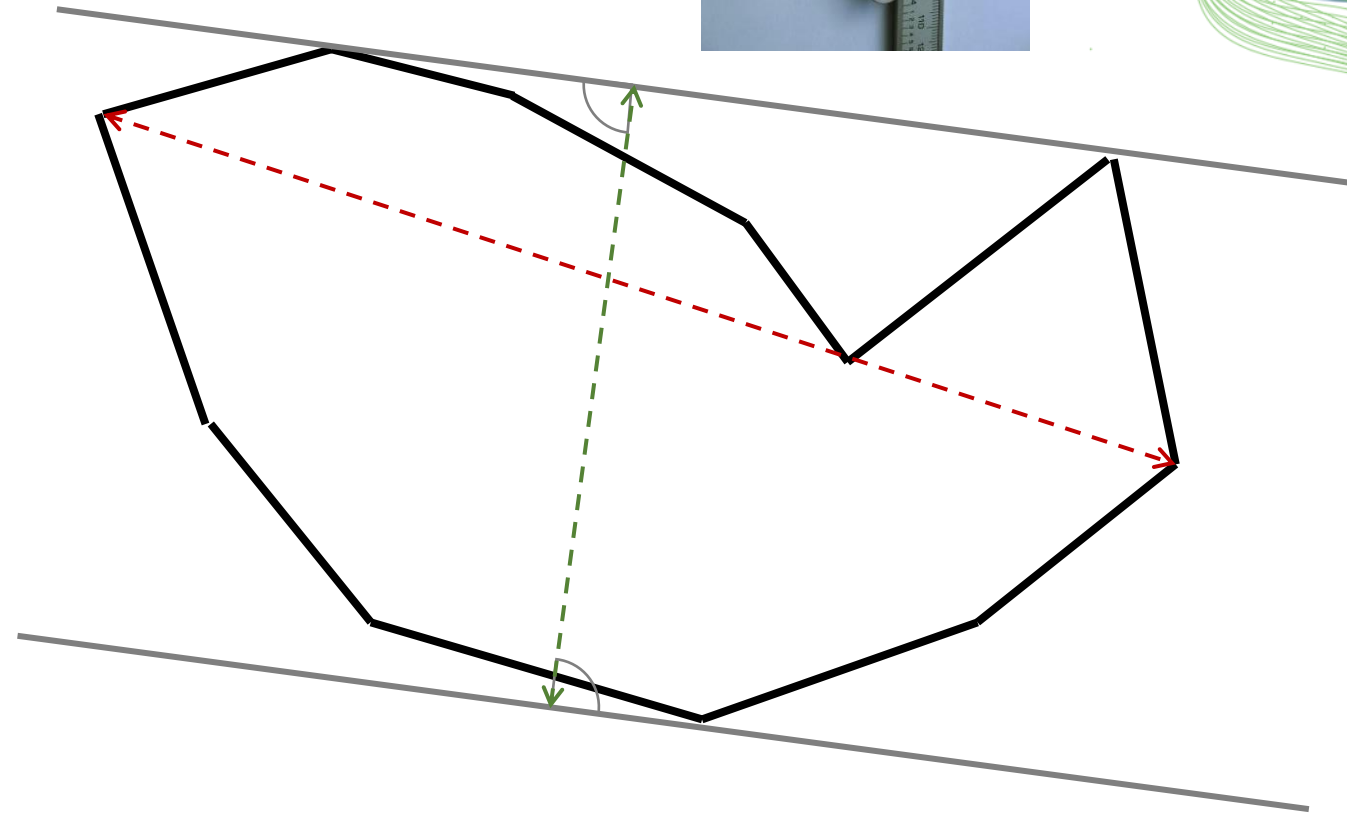
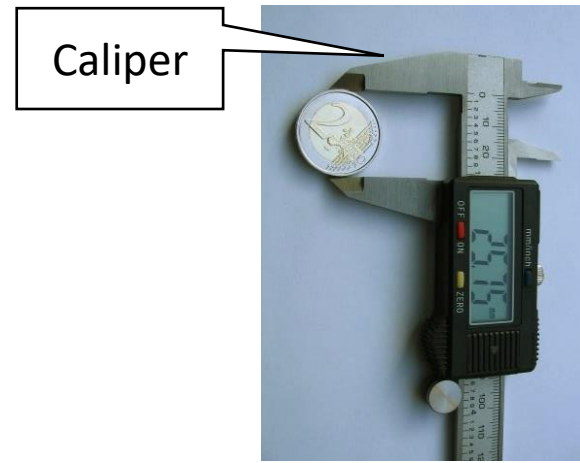
- Length of the outline around an object
- Depends on the actual implementation





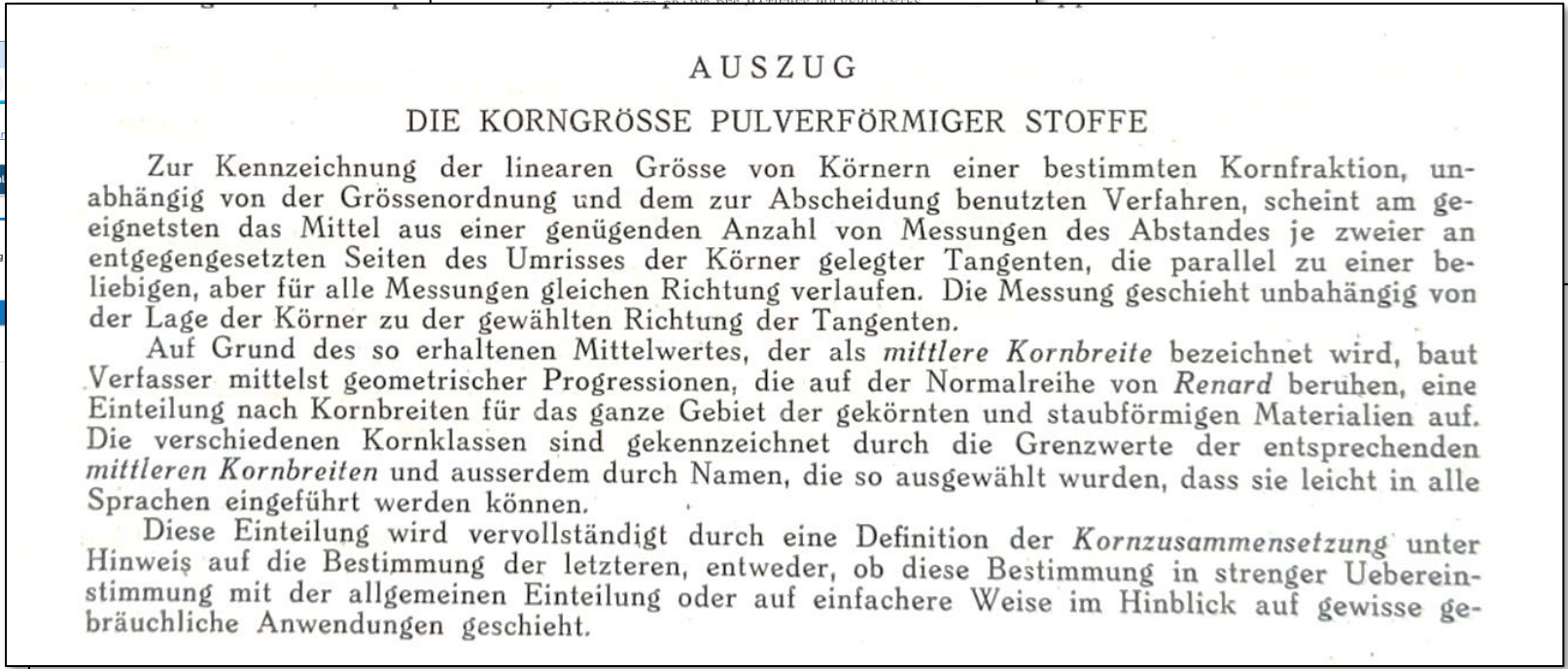
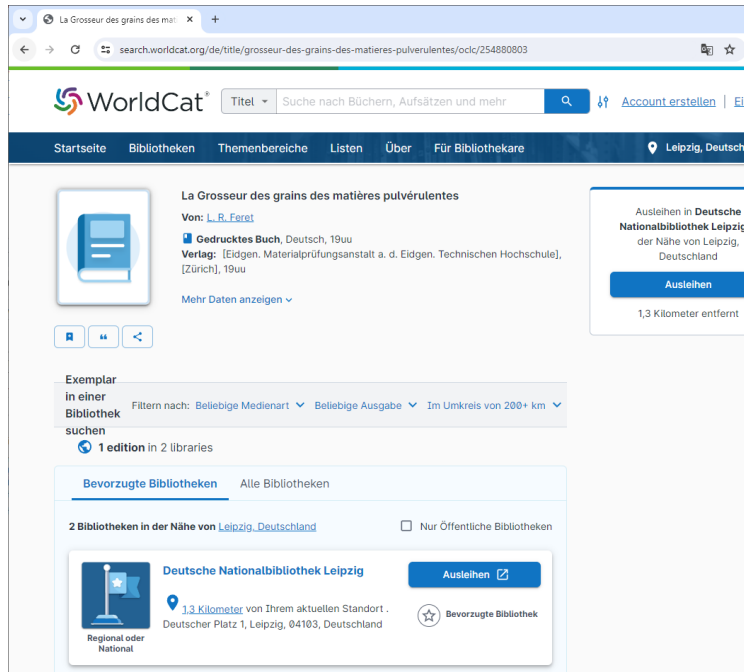
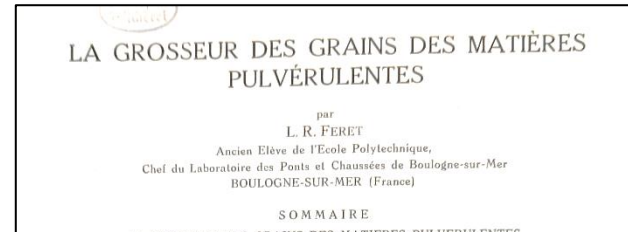
# Feret's diameter

- **Feret's diameter** describes the maximum distance between any two points of an outline.
- The **minimum caliper** ("Minimum Feret") describes the shortest distance, the object would fit through.
- Feret and Minimum Feret do not need to be perpendicular to each other!



# Feret's diameter

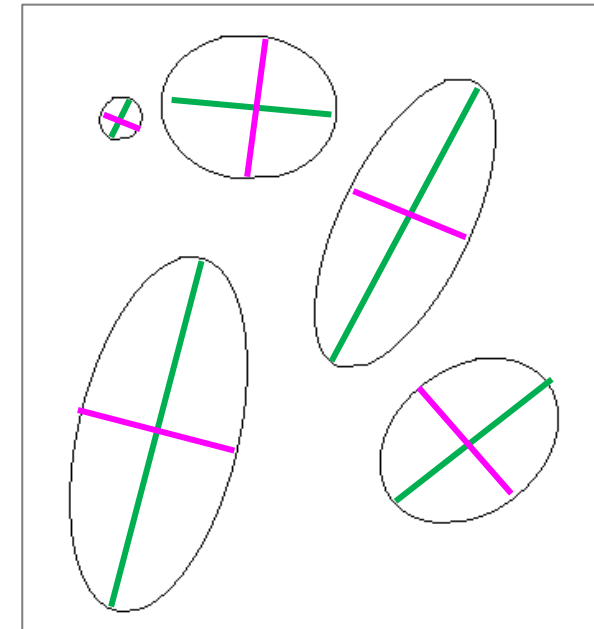
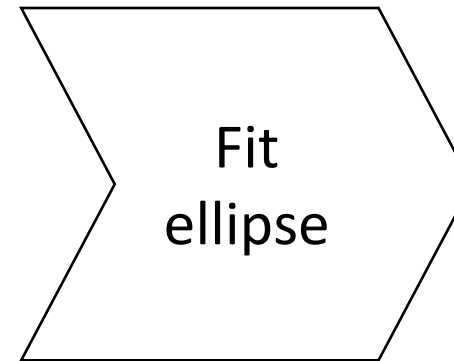
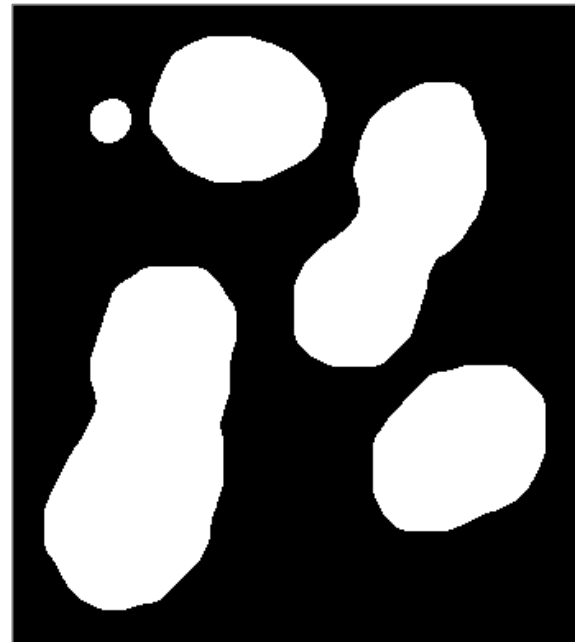
- Feret's diameter (L.R. Feret, 1931) is often cited, but impossible to read online ...
- The term "Feret's Diameter" was established in the 1970s



# Minor / major axis

- For every object, find the optimal ellipse simplifying the object.
- Major axis ... long diameter
- Minor axis ... short diameter

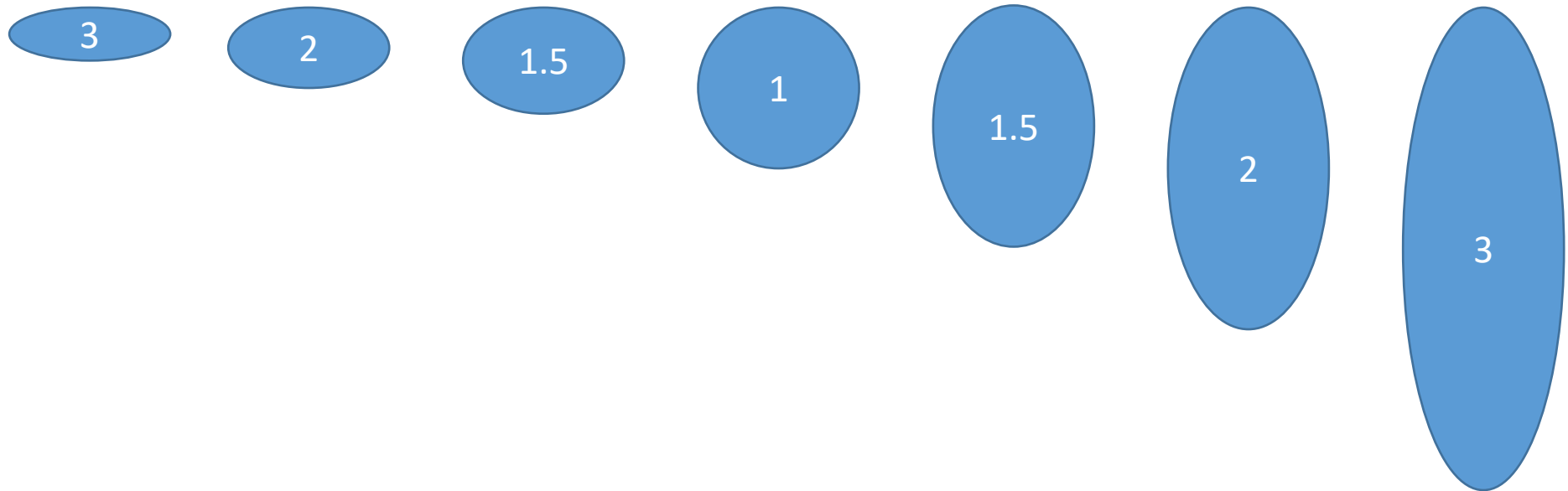
- Major and minor axis are perpendicular to each other



# Aspect ratio

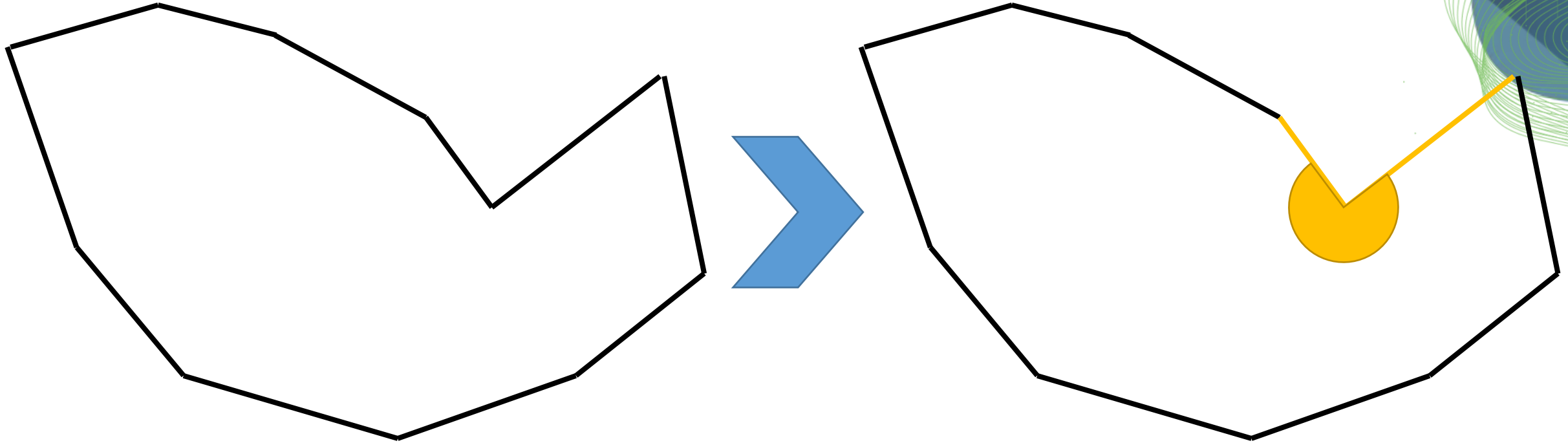
- The aspect ratio describes the elongation of an object.

$$AR = \text{major} / \text{minor}$$



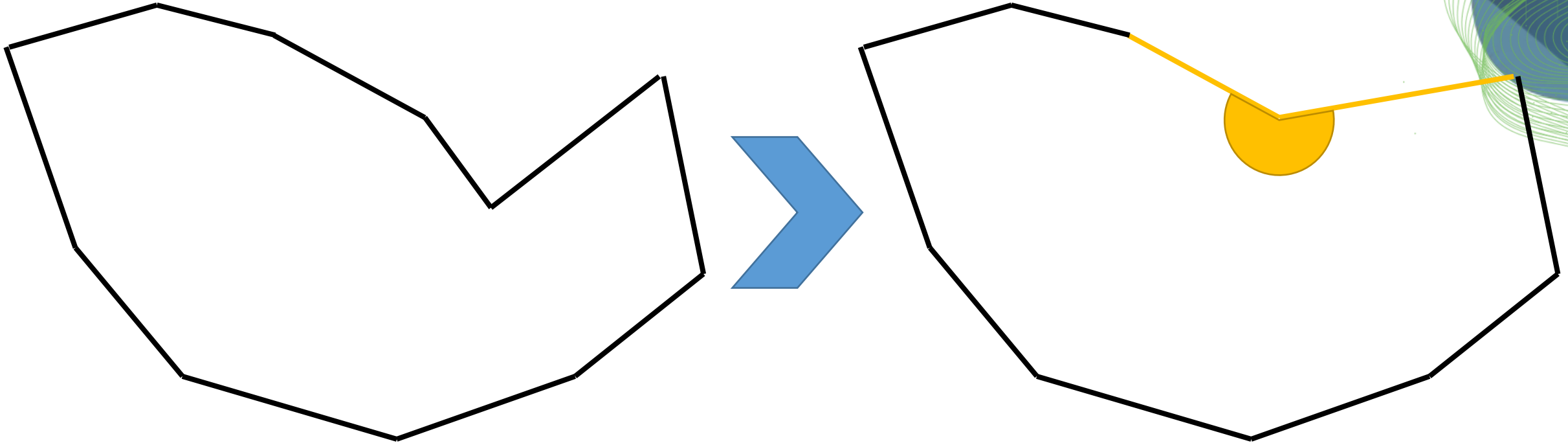
# Convex hull

- By removing all concave corners of an object, we retrieve its **convex hull**.



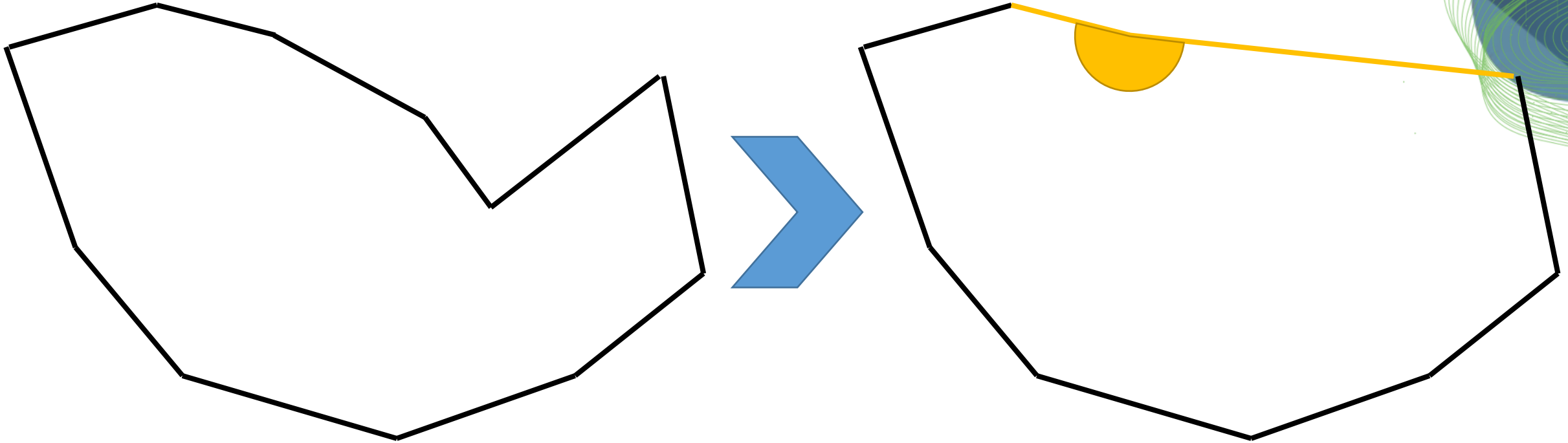
# Convex hull

- By removing all concave corners of an object, we retrieve its **convex hull**.



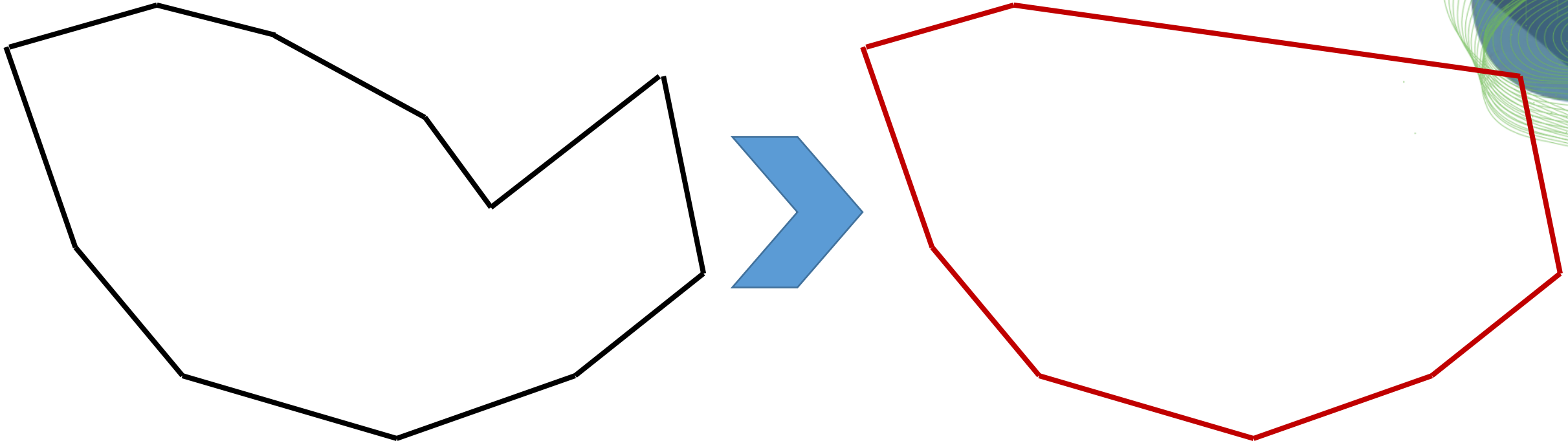
# Convex hull

- By removing all concave corners of an object, we retrieve its **convex hull**.



# Convex hull

- By removing all concave corners of an object, we retrieve its **convex hull**.



$$\text{solidity} = \frac{A}{A_{\text{convexHull}}}$$



# Roundness and circularity

- The definition of a circle leads us to measurements of circularity and roundness.
- In case you use these measures, define them correctly. They are not standardized!

Diameter

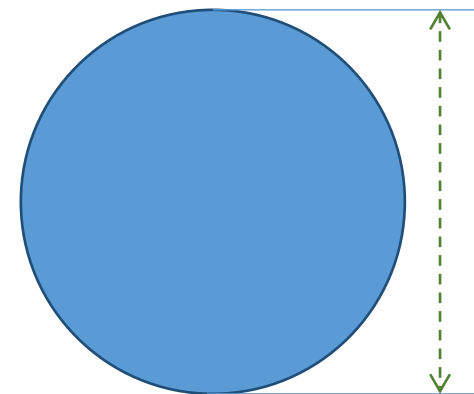
$d$

Circumference

$$C = \pi d$$

Area

$$A = \frac{\pi d^2}{4}$$



$$roundness = \frac{4 * A}{\pi major^2}$$

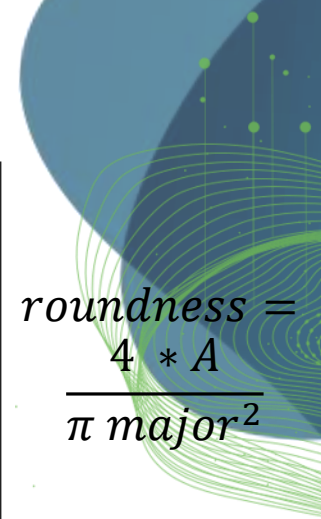
$$circularity = \frac{4\pi * A}{perimeter^2}$$

Roundness = 1  
Circularity = 1

Roundness  $\approx$  1  
Circularity  $\approx$  1

Roundness < 1  
Circularity < 1

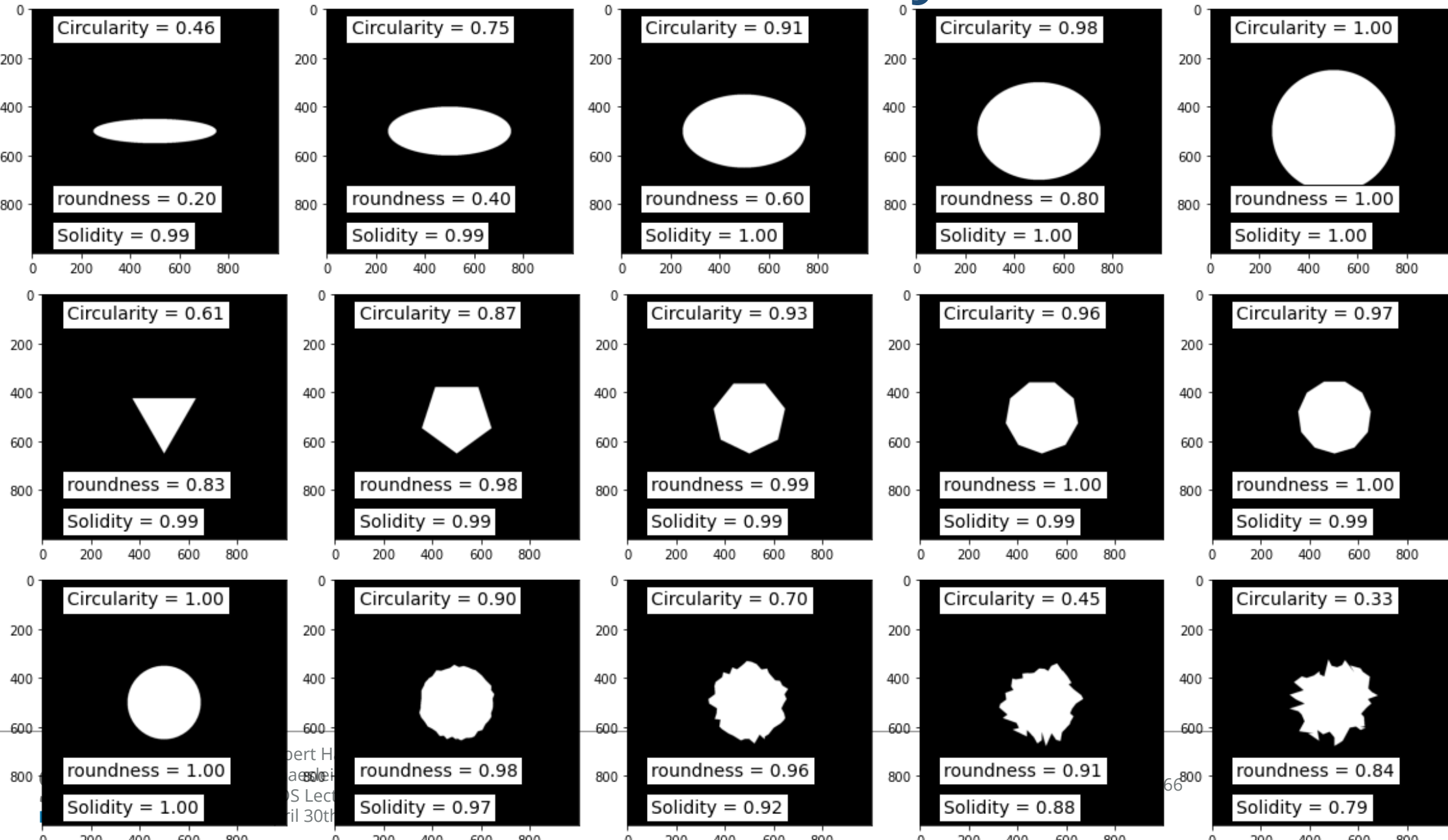
# Roundness and circularity



$$\text{roundness} = \frac{4 * A}{\pi \text{major}^2}$$

$$\text{circularity} = \frac{4\pi * A}{\text{perimeter}^2}$$

$$\text{solidity} = \frac{A}{A_{\text{convexHull}}}$$



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# Feature extraction in Python

- In Python: `from skimage import measure`

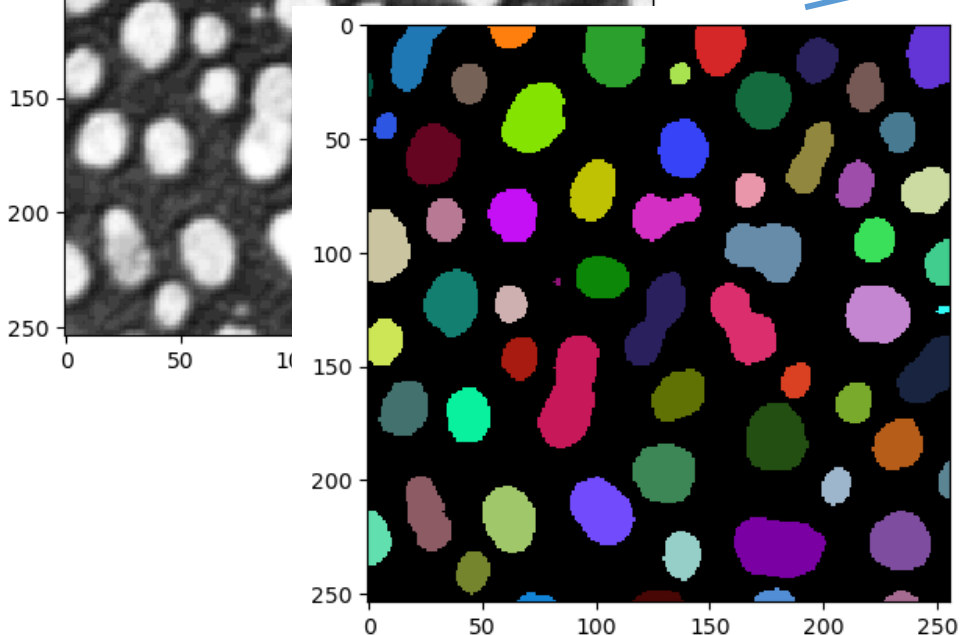
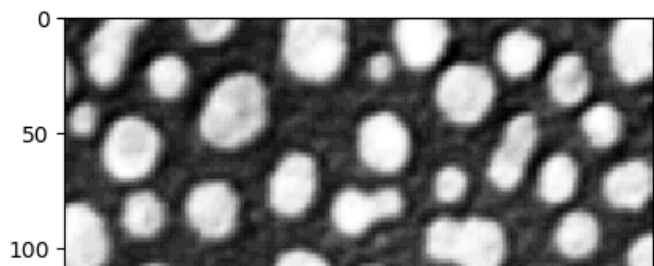
<https://scikit-image.org/docs/stable/api/skimage.measure.html>

```
skimage.measure.regionprops (label_image[, ...])    Measure properties of labeled image regions.  
  
skimage.measure.regionprops_table (label_image)    Compute image properties and return them as a pandas-compatible table.
```

```
area : int  
    Number of pixels of the region.  
  
area_bbox : int  
    Number of pixels of bounding box.  
  
area_convex : int  
    Number of pixels of convex hull image, which is the smallest convex polygon that encloses the region.  
  
area_filled : int  
    Number of pixels of the region with all the holes filled in. Describes the area of the image_filled.  
  
axis_major_length : float  
    The length of the major axis of the ellipse that has the same normalized second central moments as the region.  
  
axis_minor_length : float  
    The length of the minor axis of the ellipse that has the same normalized second central moments as the region.
```

# Feature extraction in Python

- The transition from image data to tabular data / pandas DataFrames



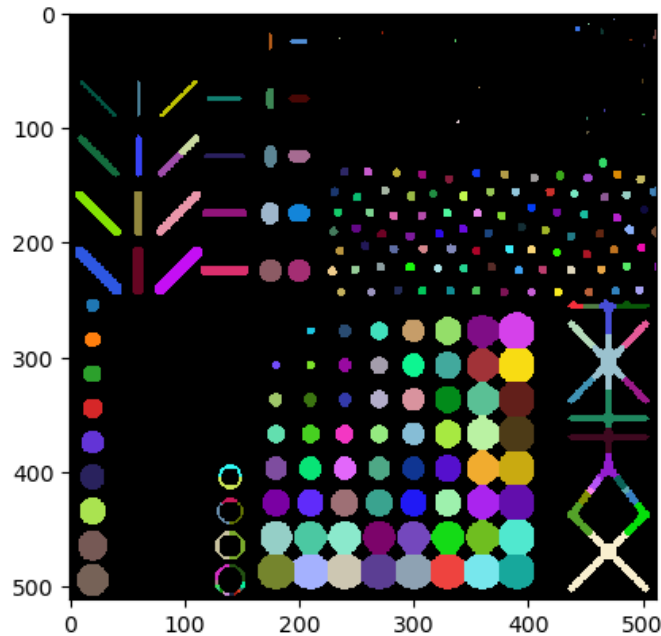
```
[4]: df = pd.DataFrame(regionprops_table(image, label_image,
perimeter = True,
shape = True,
position=True,
moments=True))
df
```

```
[4]:
```

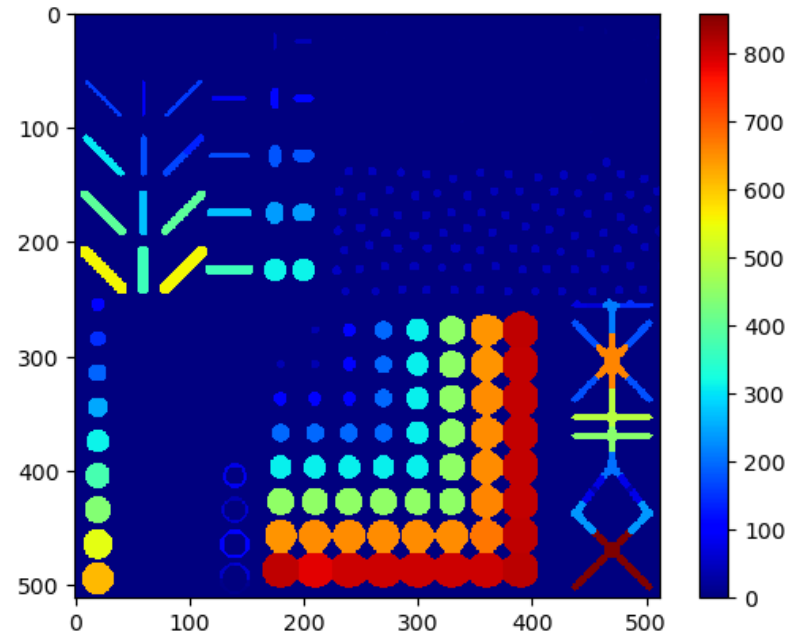
	label	area	bbox_area	equivalent_diameter	convex_area	max_intensity	mean_intensity	min_intensity	perimeter	perimete
0	1	429.0	750.0	23.371345	479.0	232.0	191.440559	128.0	89.012193	
1	2	183.0	231.0	15.264430	190.0	224.0	179.846995	128.0	53.556349	
2	3	658.0	756.0	28.944630	673.0	248.0	205.604863	120.0	95.698485	
3	4	433.0	529.0	23.480049	445.0	248.0	217.515012	120.0	77.455844	
4	5	472.0	551.0	24.514670	486.0	248.0	213.033898	128.0	83.798990	
...	...	...	...	...	...	...	...	...	...	...
57	58	213.0	285.0	16.468152	221.0	224.0	184.525822	120.0	52.284271	
58	59	79.0	108.0	10.029253	84.0	248.0	184.810127	128.0	39.313708	
59	60	88.0	110.0	10.585135	92.0	216.0	182.727273	128.0	45.692388	
60	61	52.0	75.0	8.136858	56.0	248.0	189.538462	128.0	30.692388	
61	62	48.0	68.0	7.817640	53.0	224.0	173.833333	128.0	33.071068	

# Parametric images

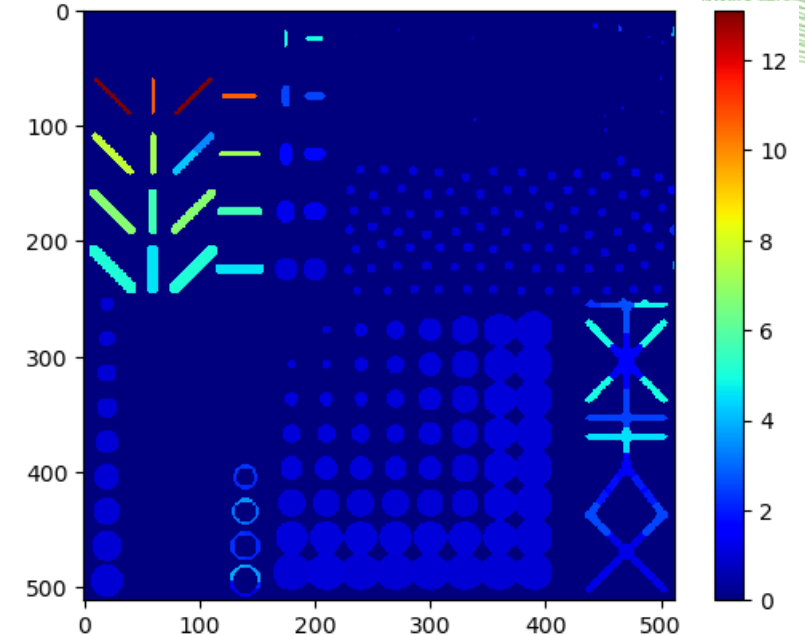
- The way back: Visualizing quantitative measurements



Label image



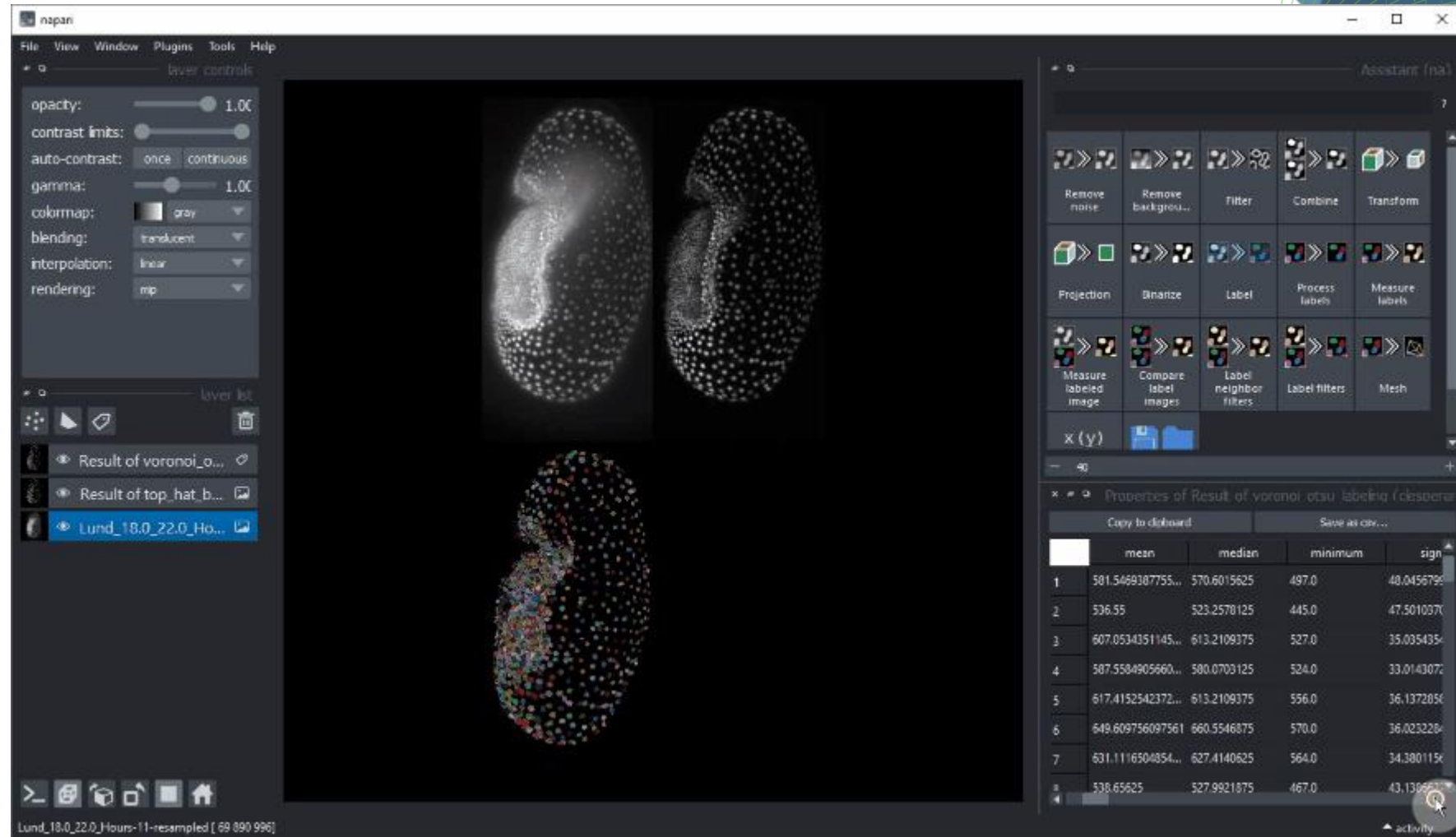
Pixel count image



Aspect ratio image

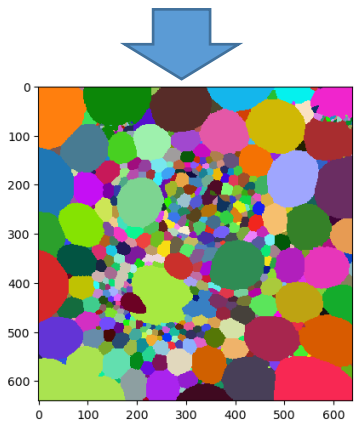
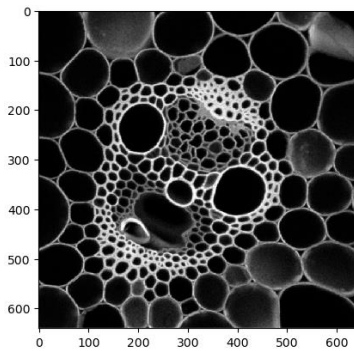
# Exploring features in Napari

- Double-click on table column to retrieve a parametric map image

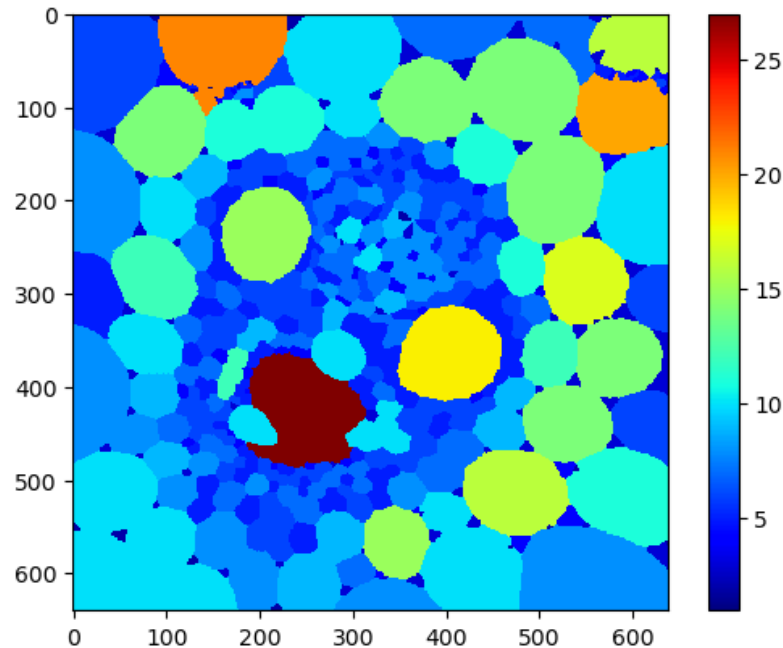


# Exploring neighborhood relationships between cells

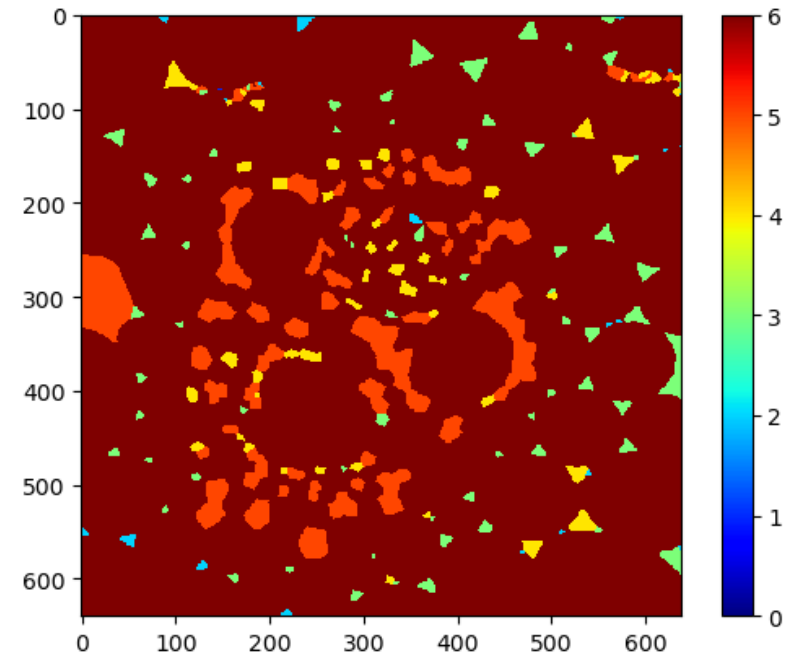
- Study how many neighbours objects have.
- How likely is it that an object with 3 neighbors is a cell?



```
num_neighbors_map = cle.touching_neighbor_count_map(objects)  
  
cle.imshow(num_neighbors_map,  
           color_map='jet',  
           colorbar=True)
```



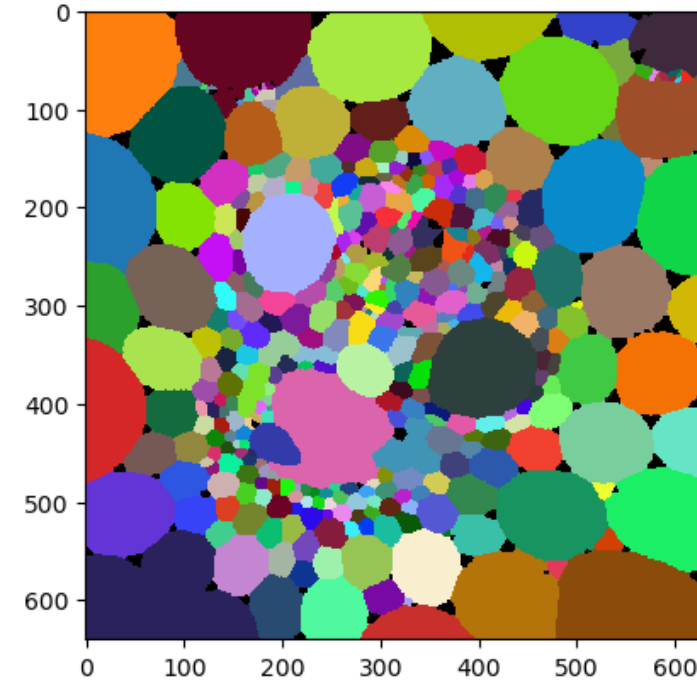
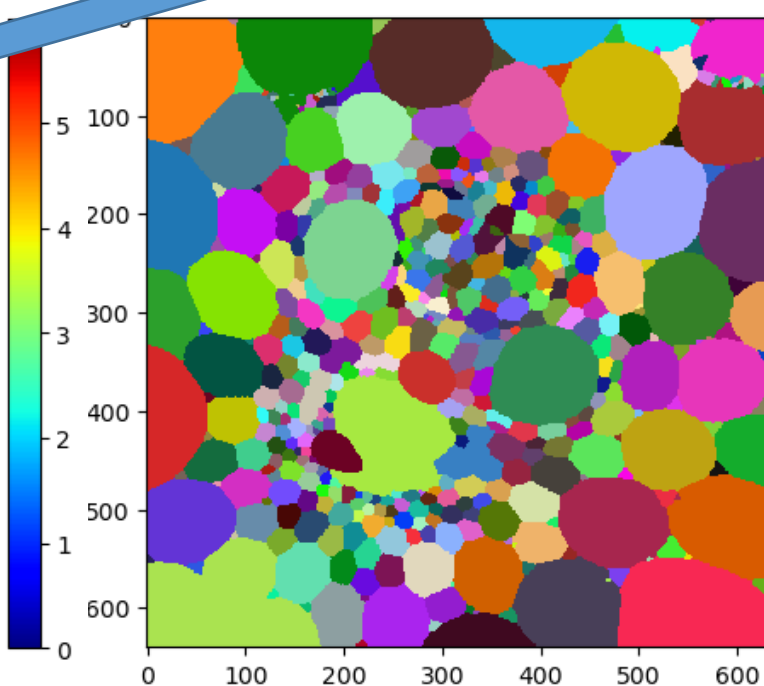
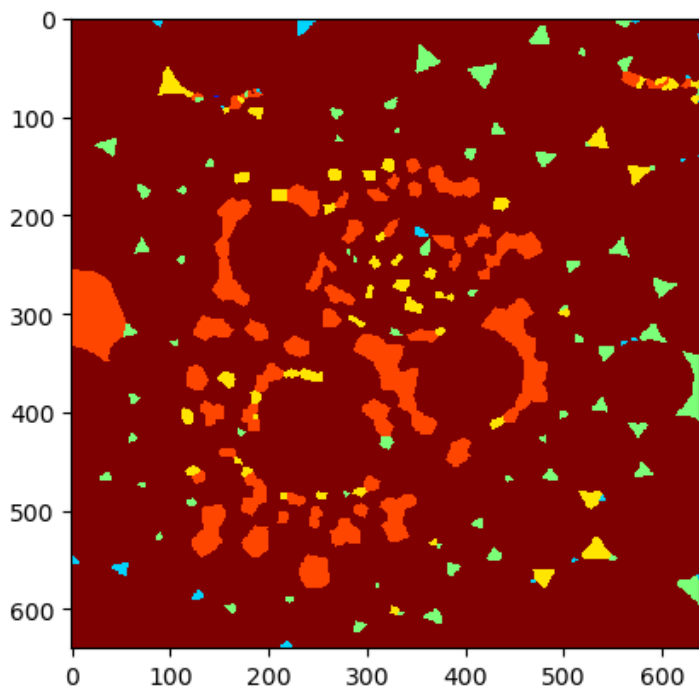
```
cle.imshow(num_neighbors_map,  
           min_display_intensity=0,  
           max_display_intensity=6,  
           color_map='jet',  
           colorbar=True)
```



# Neighborhood-based label filters

- Filter out objects which have an unreasonable number of neighbors

```
cells = cle.exclude_labels_with_map_values_out_of_range(num_neighbors_map, objects, minimum_value_range=4)  
cle.imshow(cells, labels=True)
```



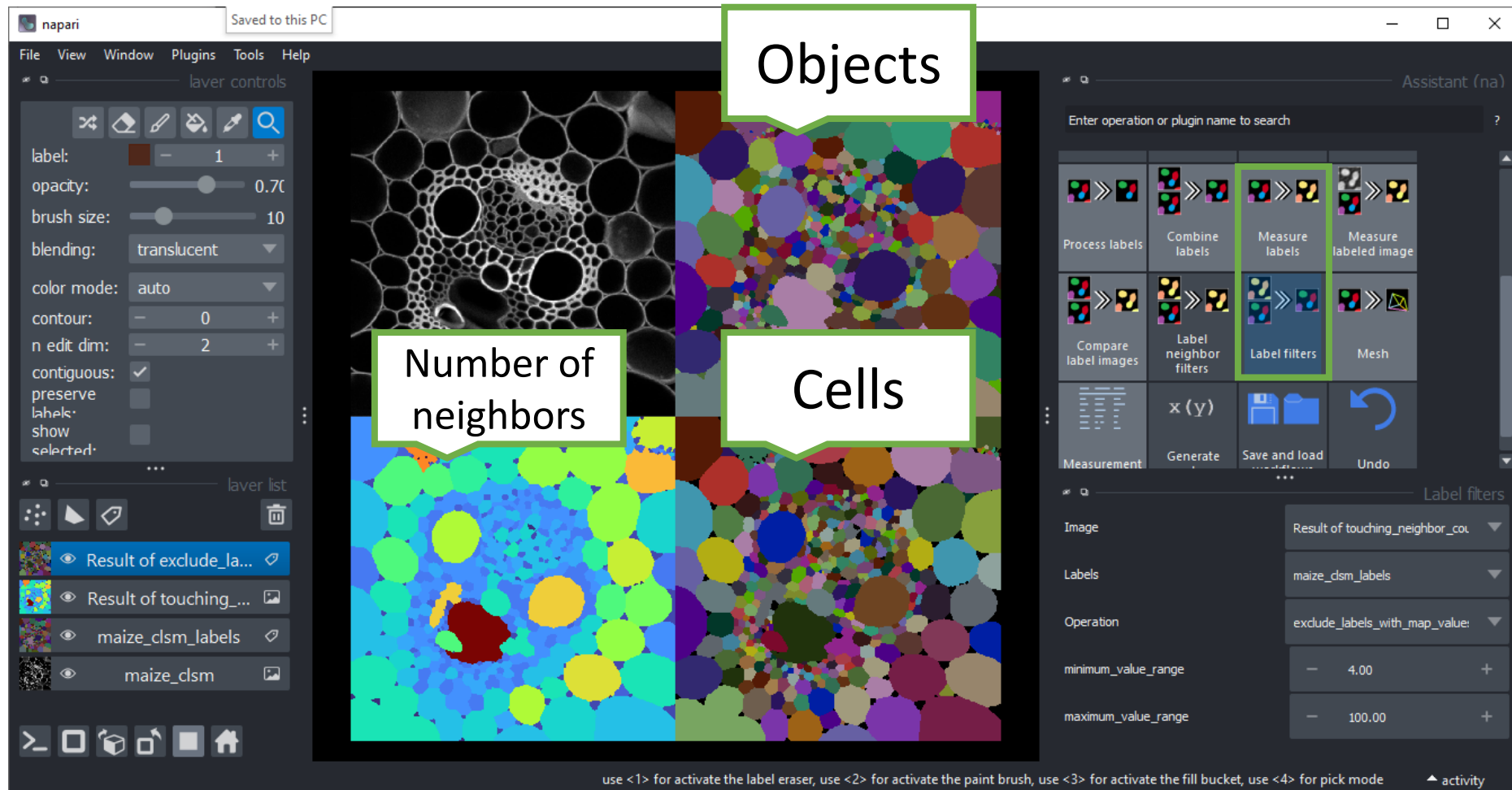
neighbor count

objects



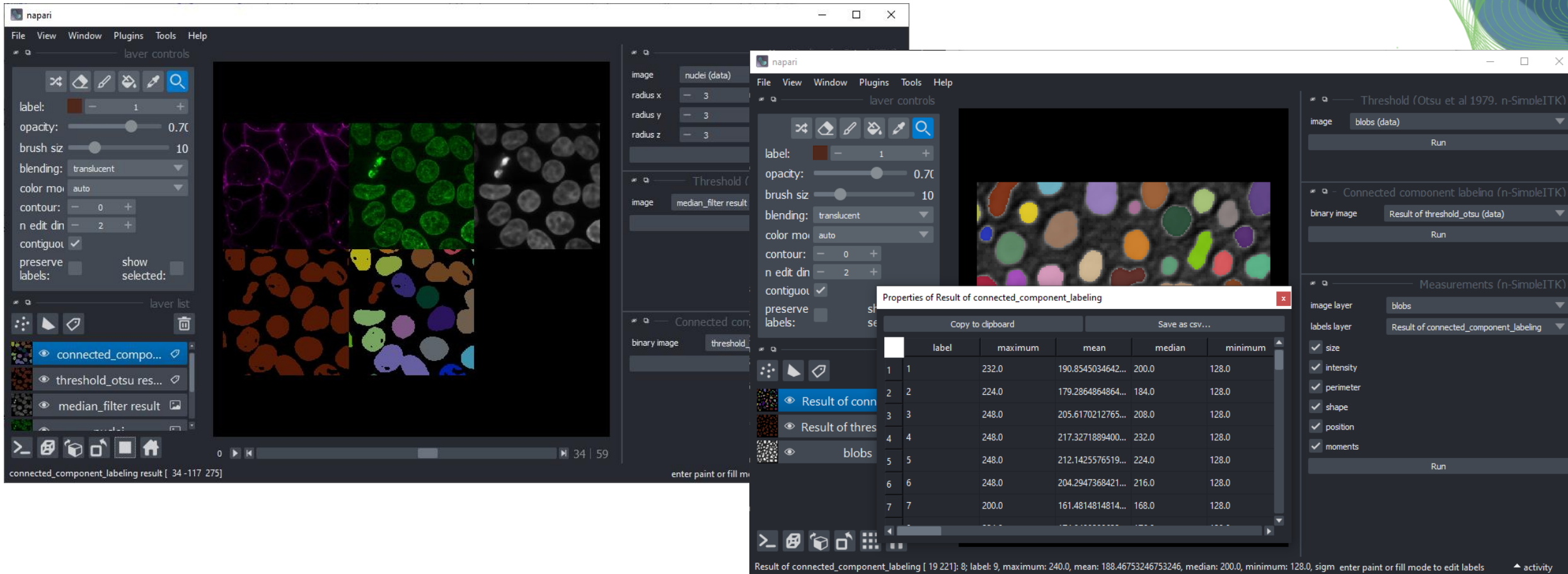
# Neighborhood-based label filters

- Filter labeled objects using Measure Labels and Label Filters in Napari.



# SimpleITK

- Recommended for 3D-measurements, based on the SimpleITK-project



# SimpleITK

- Many Napari plugins for feature extraction can also be called from Python

```
statistics = label_statistics(blobs, labels,  
                             intensity=True,  
                             size=True,  
                             shape=True,  
                             perimeter=True,  
                             position=True,  
                             moments=True)
```

```
df = pd.DataFrame(statistics)  
df
```

	label	maximum	mean	median	minimum	sigma	sum	variance	bbox_0	bbox_1
<b>0</b>	1	224.0	137.526132	136.0	112.0	13.360739	157880.0	178.509343	0	0
<b>1</b>	2	232.0	193.014354	200.0	128.0	28.559077	80680.0	815.620897	11	0
<b>2</b>	3	224.0	179.846995	184.0	128.0	21.328889	32912.0	454.921516	53	0
<b>3</b>	4	248.0	207.082171	216.0	120.0	27.772832	133568.0	771.330194	95	0
<b>4</b>	5	248.0	223.146402	232.0	128.0	30.246515	89928.0	914.851647	144	0
<b>5</b>	6	248.0	214.906725	224.0	128.0	26.386796	99072.0	696.263020	238	0
<b>6</b>	7	248.0	211.565891	224.0	136.0	30.197236	54584.0	911.873073	189	7
<b>7</b>	8	200.0	166.171429	168.0	136.0	16.466894	11632.0	271.158592	133	17

# Exploring features in Napari

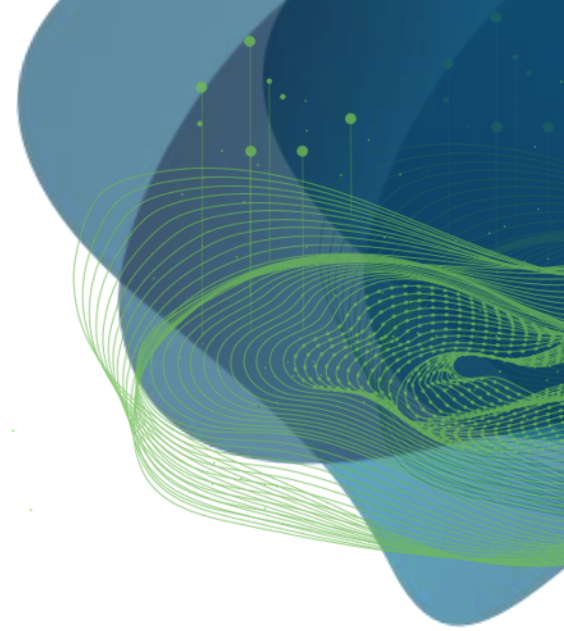
- Select table rows and view corresponding object in 2D/3D space

The screenshot displays the Napari application interface. On the left, the 'Layers' panel shows two layers: 'voronoi\_otsu\_labeli...' and 'nuclei'. The main view shows a 3D visualization of cell nuclei, with one nucleus highlighted in yellow. On the right, a table displays the features of the selected object. The table has columns for 'label', 'area', 'bbox\_area', and 'convex\_area'. The row for label 10 is highlighted, showing an area of 5917, a bounding box area of 1.327e+05, and a convex area of 67149.

label	area	bbox_area	convex_area
3	295	680	333
4	33660	73950	38125
5	37629	73950	40063
6	15077	34320	17000
7	2290	6498	2845
8	24546	45849	25769
9	5917	1.327e+05	67149
10	34268	64170	39030
11	35230	68634	37399
12	47803	93600	51680
13	44426	85833	46414
14	39179	77376	41735
15	3608	8640	3980
16	596	1292	661
17	39978	75264	42647
18	35121	59940	38226
19	35980	76500	38102
20	50092	1.091e+05	56547
21	4955	12025	5668
22	41369	76755	45651
23	38750	1.123e+05	50019

# Complex exercise

Robert Haase



GEFÖRDERT VOM



Bundesministerium  
für Bildung  
und Forschung



Diese Maßnahme wird gefördert durch die Bundesregierung aufgrund eines Beschlusses des Deutschen Bundestages. Diese Maßnahme wird mitfinanziert durch Steuermittel auf der Grundlage des von den Abgeordneten des Sächsischen Landtags beschlossenen Haushaltes.

# Complex exercise

- Scenario: Imagine a biologist sent you some data (images + corresponding label image). They ask you to write an image-analysis workflow for processing these images + more images of similar kind.
- You will receive a link to data in-person
  - You can return the link and exchange it with another link 2 times.
- Scientific tasks
  - Develop an image-segmentation workflow, which produces label images
  - Extract features from these images
  - Visualize relationships between these features
  - Find out which features are strongly correlated and which not.

# Complex exercise

- Engineering tasks
  - Setup a software environment
  - Setup an image processing workflow
  - Setup a data analysis / visualization workflow
  - Setup a quality assurance procedure
- Documentation tasks
  - Installation instructions
  - User guide
  - Documentation of used data
  - Explanation of the used algorithms

Act as if you would communicate with a biologist, with limited image-analysis, conda and programming skills.

# Complex exercise

- Submission

- Submit a password-protected ZIP file to [robert.haase@uni-leipzig.de](mailto:robert.haase@uni-leipzig.de) (Why password protected: The virus scanner cannot reject python files in encrypted zip-files)
- Allowed file formats: ipynb, py, docx, pdf, md, csv, yml, json, xml, txt
- Deadline: June 24<sup>th</sup> 2024

- Hint

- Send this ZIP file to a friend and ask them to run the analysis. If they can follow your instructions successfully, without communicating with you, proceed to final submission.



# Complex exercise

- Checklist
  - The software environment is reproducible
  - The example data is available in the right directory (note: you cannot submit a 500MB ZIP file via email) The image/data analysis code is executable
  - The code is well documented / commented
  - Segmentation results are visualized
  - Segmentation results are stored to disc as label images
  - The quality of the segmentation result is measured
  - Used algorithms are cited, and well explained
  - Extracted features / measurements are saved as CSV-file in a way that one can associate an entry in the CSV file with the corresponding segmented object
  - Resulting plots and visualizations have reasonable axis labels and are well explained
  - The copyright of re-used data and code are respected

# Next week:

- 15:15-16:30 Short lecture + short practicals (SG 312)
- Afterwards:



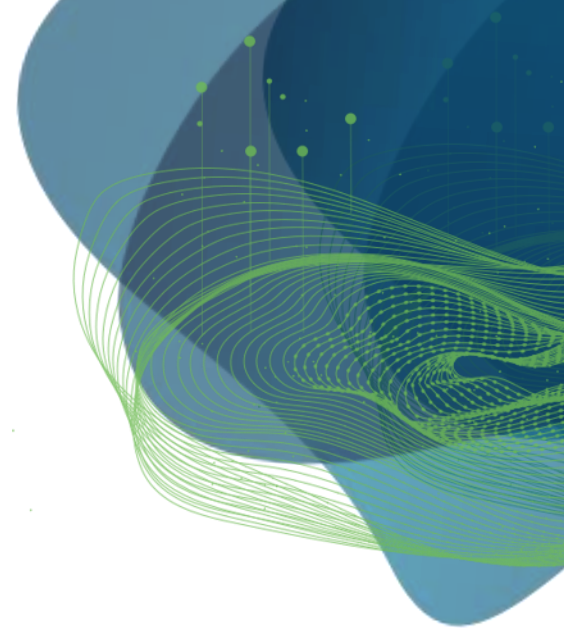
Meetup Relaunch  
**ChatBots  
und Generative KI**  
7. Mai 2024 | START 17:00

ScaDS.AI  
DRESDEN LEIPZIG

Living Lab  
ScaDS.AI Dresden/Leipzig  
Humboldtstraße 25, 04105 Leipzig

# Exercises

Robert Haase



GEFÖRDERT VOM



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und Forschung



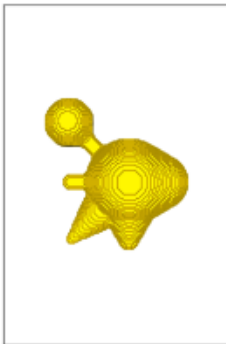
Diese Maßnahme wird gefördert durch die Bundesregierung aufgrund eines Beschlusses des Deutschen Bundestages. Diese Maßnahme wird mitfinanziert durch Steuermittel auf der Grundlage des von den Abgeordneten des Sächsischen Landtags beschlossenen Haushaltes.

# Exercise: Surface meshes

- Creating, storing, processing surface mesh data

```
[9]: new_surface = nppas.to_napari_surface_data(new_mesh)
new_surface
```

[9]:



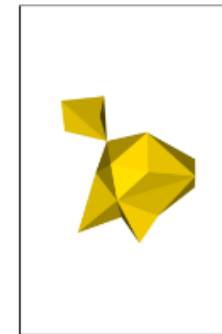
nppas.SurfaceTuple	
origin (z/y/x)	[0. 0. 0.]
center of mass(z/y/x)	50.000,46.575,42.589
scale(z/y/x)	1.000,1.000,1.000
bounds (z/y/x)	25.500...74.500 2.500...88.500 2.500...83.500
average size	31.277
number of vertices	19040
number of faces	38076

## Exercise

Load the `skimage.data.cells3d` dataset, extract the nuclei channel and create a surface mesh for every individual nucleus. Store all these surface meshes to disc.

```
[11]: simplified_surface2 = nppas.decimate_quadric(smoothed_surface, fraction=0.001)
simplified_surface2
```

[11]:



nppas.SurfaceTuple	
origin (z/y/x)	[0. 0. 0.]
center of mass(z/y/x)	49.959,46.174,40.689
scale(z/y/x)	1.000,1.000,1.000
bounds (z/y/x)	25.809...76.552 3.858...93.107 3.385...83.730
average size	35.058
number of vertices	22
number of faces	38

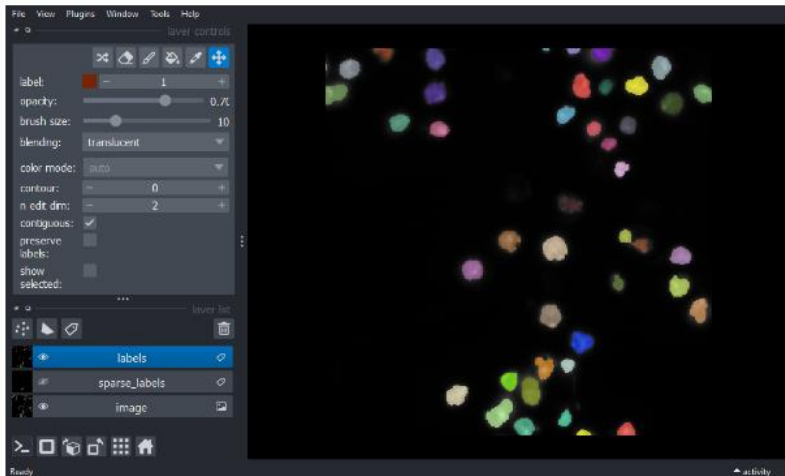
## Exercise

Store the branchoid as 8-bit integer image to disc. Compare the file size to the differently simplified meshes above.

# Segmentation quality

- Measure segmentation quality of a given algorithm applied to a folder of images.

```
[7]: def my_segmentation_algorithm(input_image):  
  
    # background subtraction  
    background_subtracted = nsbatwm.white_tophat(input_image, radius = 10)  
  
    # instance segmentation / labeling  
    labels_result = nsbatwm.voronoi_otsu_labeling(background_subtracted,  
  
    return labels_result
```



## Quality estimation: Sparse Jaccard Index

From the two label images loaded and produced above we can compute the sparse Jaccard Index.

```
[9]: metrics.jaccard_index_sparse(sparse_labels, labels)
```

```
[9]: 0.8357392602053431
```

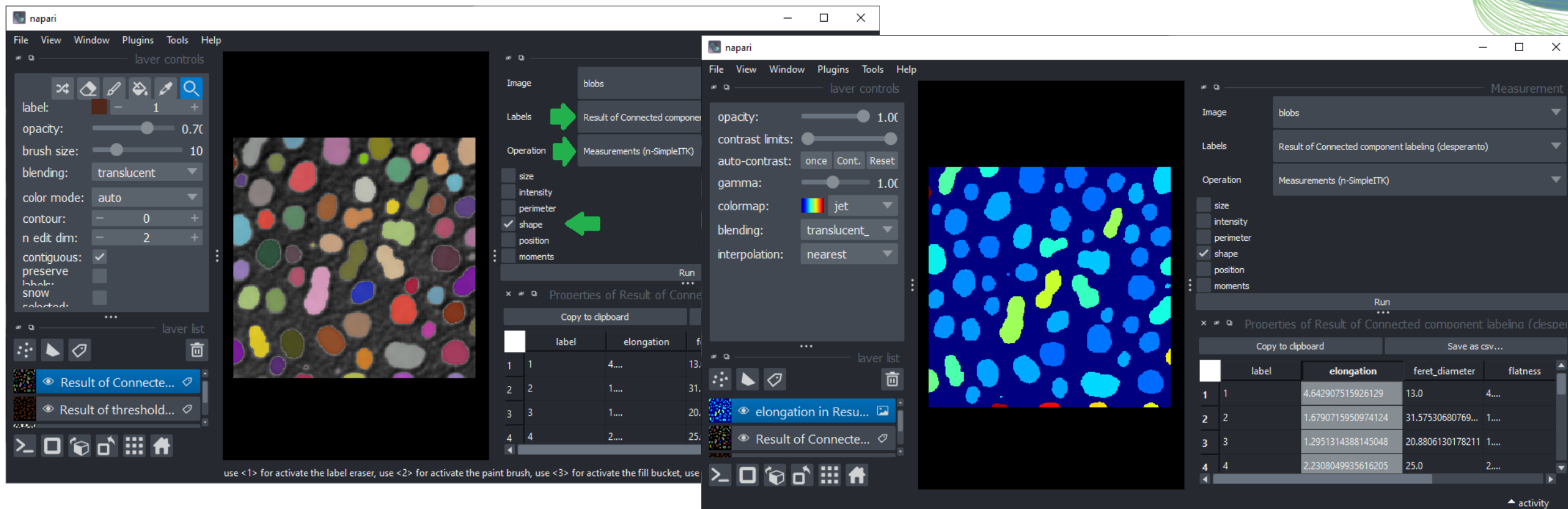
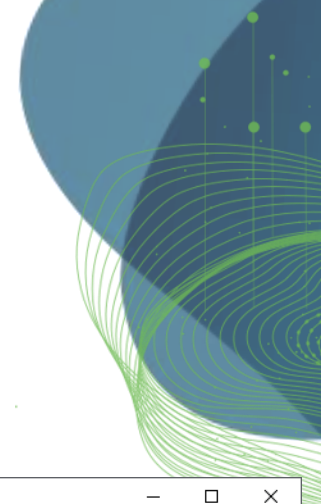
## Exercise

Use the following for-loop and code snippets from above to compute the segmentation quality of all images in the folder. Provide the average quality over all images.

```
[10]: for image_filename in os.listdir(image_folder):  
        print(image_folder + image_filename)  
  
data/BBBC007_batch/17P1_POS0013_D_1UL.tif  
data/BBBC007_batch/20P1_POS0005_D_1UL.tif  
data/BBBC007_batch/20P1_POS0007_D_1UL.tif  
data/BBBC007_batch/20P1_POS0010_D_1UL.tif  
data/BBBC007_batch/A9_p7d.tif  
data/BBBC007_batch/AS_09125_040701150004_A02f00d0.tif
```

# Exercise: Parametric maps

- Produce a parametric map representing 'elongation' in Napari.
- Reproduce the same map using Python



# Exercise: Quantitative measurements

- Use the given feature extraction notebook to apply some basic statistics to measurements

```
[5]: df = pd.DataFrame(regionprops_table(image , label_image,
                                     perimeter = True,
                                     shape = True,
                                     position=True,
                                     moments=True))
df
```

```
[5]:
```

	label	area	bbox_area	equivalent_diameter	convex_area	max_intensity	mean_intensity	min_intensity	perimeter	perimete
0	1	429.0	750.0	23.371345	479.0	232.0	191.440559	128.0	89.012193	
1	2	183.0	231.0	15.264430	190.0	224.0	179.846995	128.0	53.556349	
2	3	658.0	756.0	28.944630	673.0	248.0	205.604863	120.0	95.698485	
3	4	433.0	529.0	23.480049	445.0	248.0	217.515012	120.0	77.455844	
4	5	472.0	551.0	24.514670	486.0	248.0	213.033898	128.0	83.798990	
...	...	...	...	...	...	...	...	...	...	...
57	58	213.0	285.0	16.468152	221.0	224.0	184.525822	120.0	52.284271	
58	59	79.0	108.0	10.029253	84.0	248.0	184.810127	128.0	39.313708	
59	60	88.0	110.0	10.585135	92.0	216.0	182.727273	128.0	45.692388	
60	61	52.0	75.0	8.136858	56.0	248.0	189.538462	128.0	30.692388	
61	62	48.0	68.0	7.817640	53.0	224.0	173.833333	128.0	33.071068	

62 rows × 86 columns

## Exercises

Make a table with only `area`, `mean_intensity`, `standard_deviation_intensity` and `label`.

```
[ ]:
```

How many object are in the dataframe?

```
[ ]:
```

How large is the largest object?

```
[ ]:
```

What is the mean intensity of the brightest object?

```
[ ]:
```

What are mean and standard deviation intensity of the image?

```
[ ]:
```

