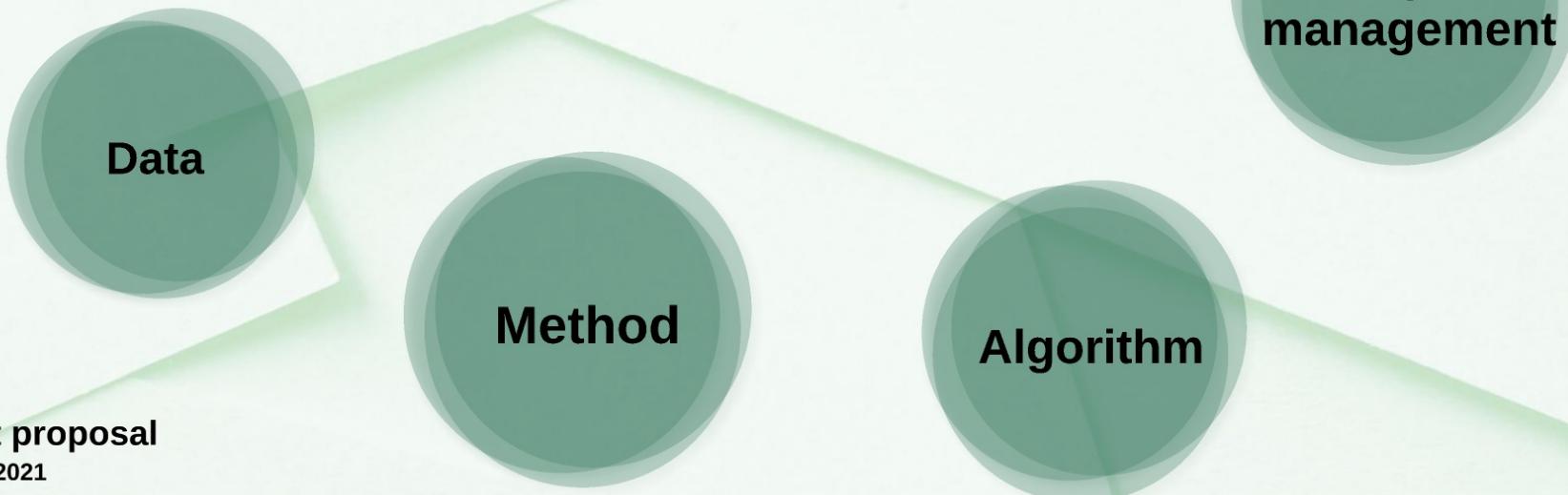


# **Cell nuclei segmentation**

via implementation and evaluation of region growing



## **Project proposal**

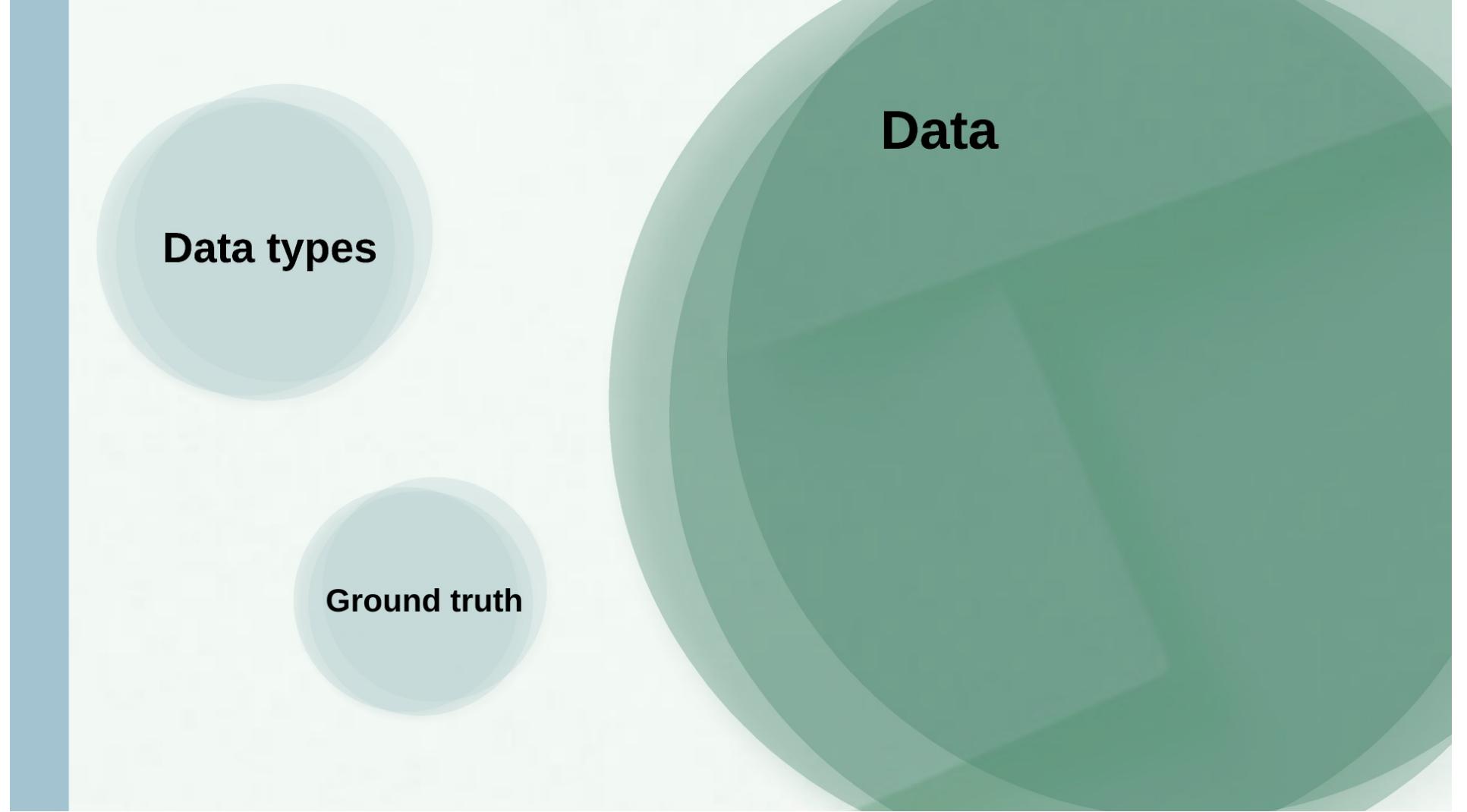
12th May 2021

Data Analysis MoBi SS2021

Supervisor: Dr. Karl Rohr, Christian Ritter; Tutor: Nicholas Peschke

Topic 04: Biomedical image analysis

Group 04: Marie Becker, Ina Jung, Laura Kaschnitz, Johanna Möller



# Data

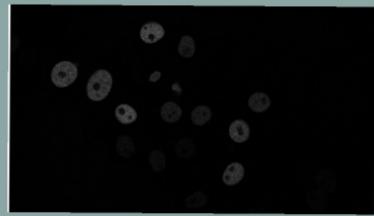
Data types

Ground truth

# Data types

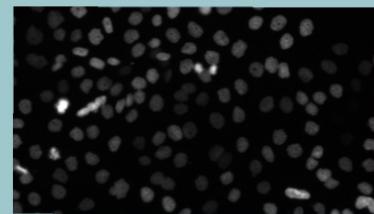
## N2DH-GOWT1

- Mouse (*mus musculus*)
- Embryonic Stem Cells
- Established from the epitheloid cervix carcinoma
- Fluorescent chromosomes, core histone 2B tagged with green GFP



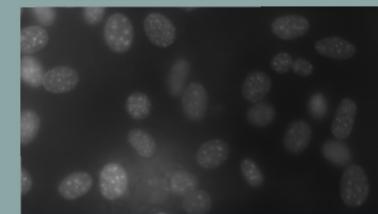
## N2DL-HeLa

- Human (*Homo sapiens*)
- Cervix carcinoma
- Established from the epitheloid cervix carcinoma
- Fluorescent chromosomes, core histone 2B tagged with green GFP

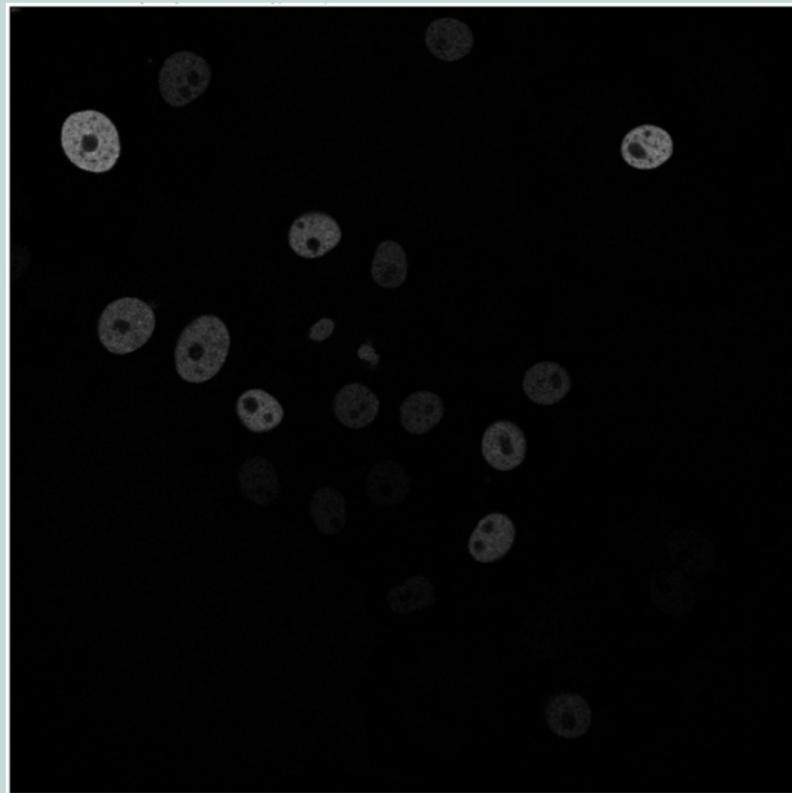


## NIH3T3

- Mouse (*mus musculus*)
- Embryo fibroblast
- Contact-inhibited NIH Swiss mouse embryo
- CD-tagged protein, used EGFP

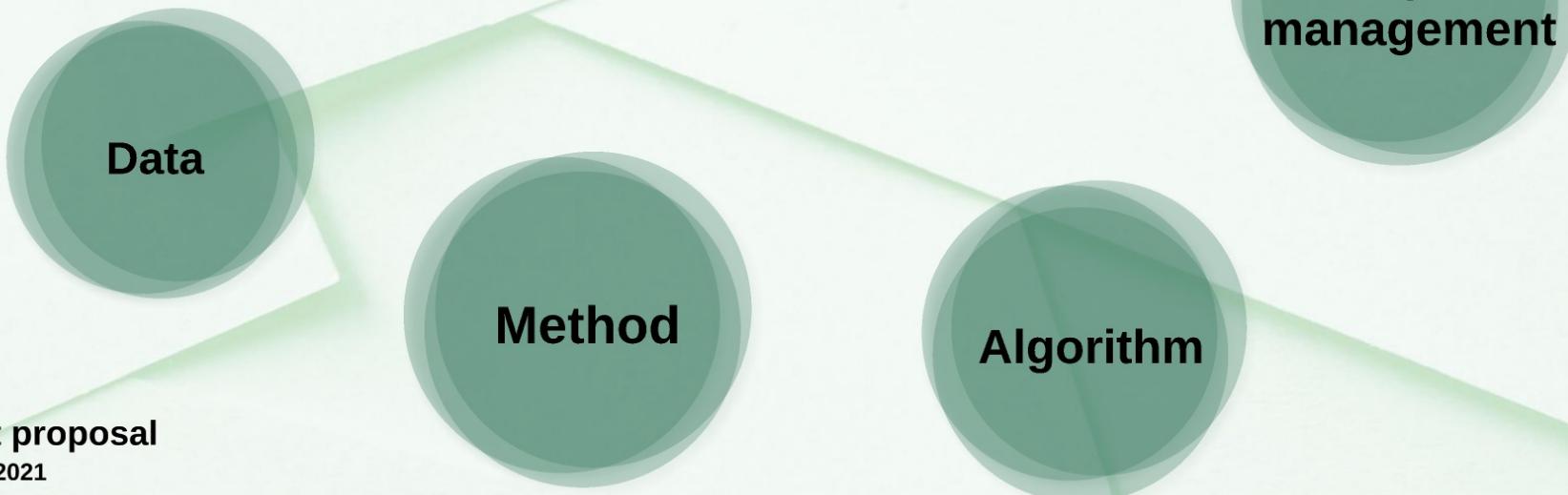


# Normal image vs. ground truth image



# **Cell nuclei segmentation**

via implementation and evaluation of region growing



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# Method

**Seeded Region  
Growing**

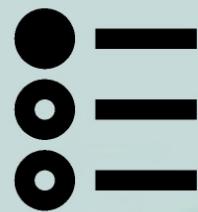
**Unseeded  
Region  
Growing**

# Seeded Region Growing

Basic procedure



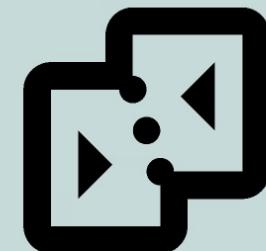
Seed  
selection



Region  
growing



Region  
merging



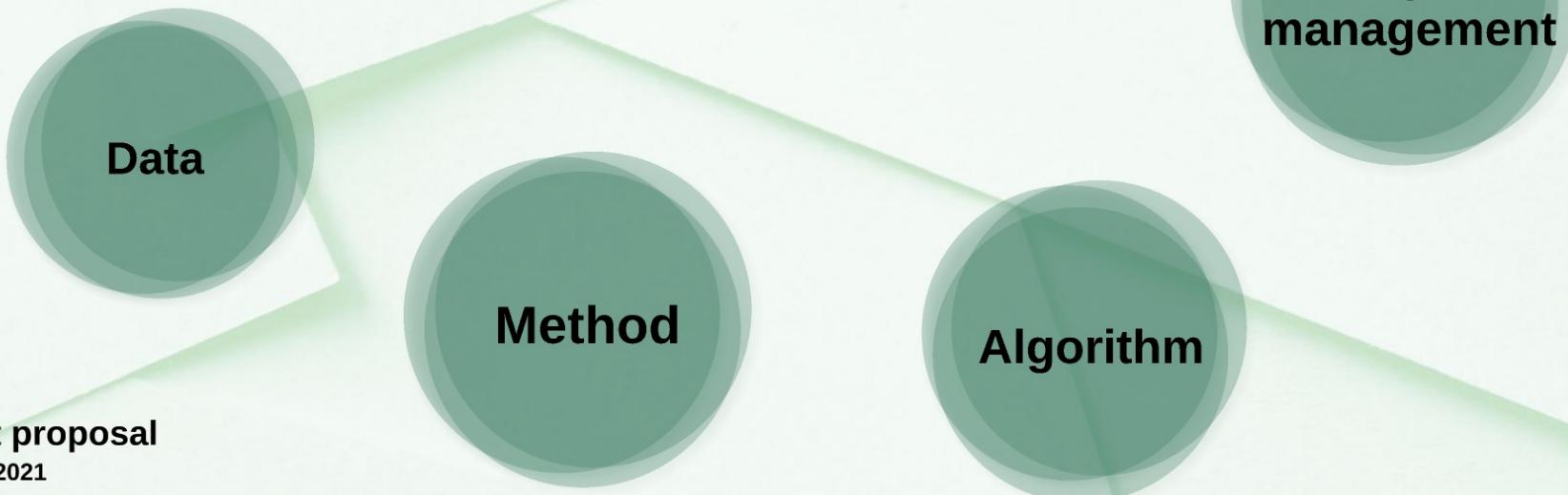
# Method

**Seeded Region  
Growing**

**Unseeded  
Region  
Growing**

# **Cell nuclei segmentation**

via implementation and evaluation of region growing



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Euclidean  
distance

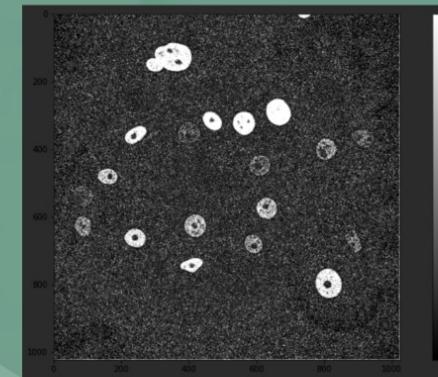
Standard  
deviation

Relative  
euclidean  
distance

# Automatic seed selection algorithm

```
def seeds(img, T1, T2): # automatic seed selection algorithm
    result = numpy.zeros(img.shape)
    sd_seeds = standarddeviation(img, 3) # standard deviation
    sd_flat = sd_seeds.flatten() # standard deviation as 1D-array
    similarity_seeds = 1 - sd_seeds/max(sd_flat) # calculates similarity of every pixel to its neighbors
    eurel_seeds = euclidean_relative(img, 3) # relative euclidean distance of every pixel to its neighbors
    for p in ndindex(img.shape): # border pixel value is zero
        if similarity_seeds[p] > T1 and eurel_seeds[p] < T2: # compares pixel with threshold
            result[p] = 1 # assigns value 1 to seeds
    return result

seed_final = seeds(img, 0.4, 20) img: {ndarray: (1024, 1024)}
show_image(seed_final, 15, 8) seed_final: {ndarray: (1024, 1024)}
```



# Standard deviation

```
def standarddeviation(img, size): # calculates standard deviation of every pixel (image, size of filter mask)
    result = numpy.zeros(img.shape) # create empty array (zeros)
    n = (size - 1) // 2
    for p in ndindex(img.shape): # iterates over every pixel
        neighborhood_sum = 0
        mean = 0
        deviation = 0
        if p[0]-n >= 0 and p[1]-n >= 0 and p[0]+n <= img.shape[0] - 1 and p[1]+n <= img.shape[1] - 1: # no calculation of border pixels
            for q in ndindex(size, size): # iterates over filter mask (nxn neighborhood)
                i = p[0]-n + q[0]
                j = p[1]-n + q[1]
                neighborhood_sum += img[i,j]
            mean = neighborhood_sum/(size**2) # calculate mean in nxn neighborhood
            for q in ndindex(size, size): # iterates over filter mask (nxn neighborhood)
                i = p[0]-n + q[0]
                j = p[1]-n + q[1]
                deviation += (img[i,j] - mean)**2 # calculate deviation from mean
        result[p] = sqrt(1/(size**2) * deviation) # calculate standard deviation
    return result
sd_test = standarddeviation(img,3) img: {ndarray: (1024, 1024)}
```

```
sd_2 = standarddeviation(img,3) # apply method for testing
sd_2 = sd_2/max(sd_2.flatten()) # normes standard deviation
similarity_2 = 1-sd_2 # calculates similarity
show_image(similarity_2, 15, 8)
```



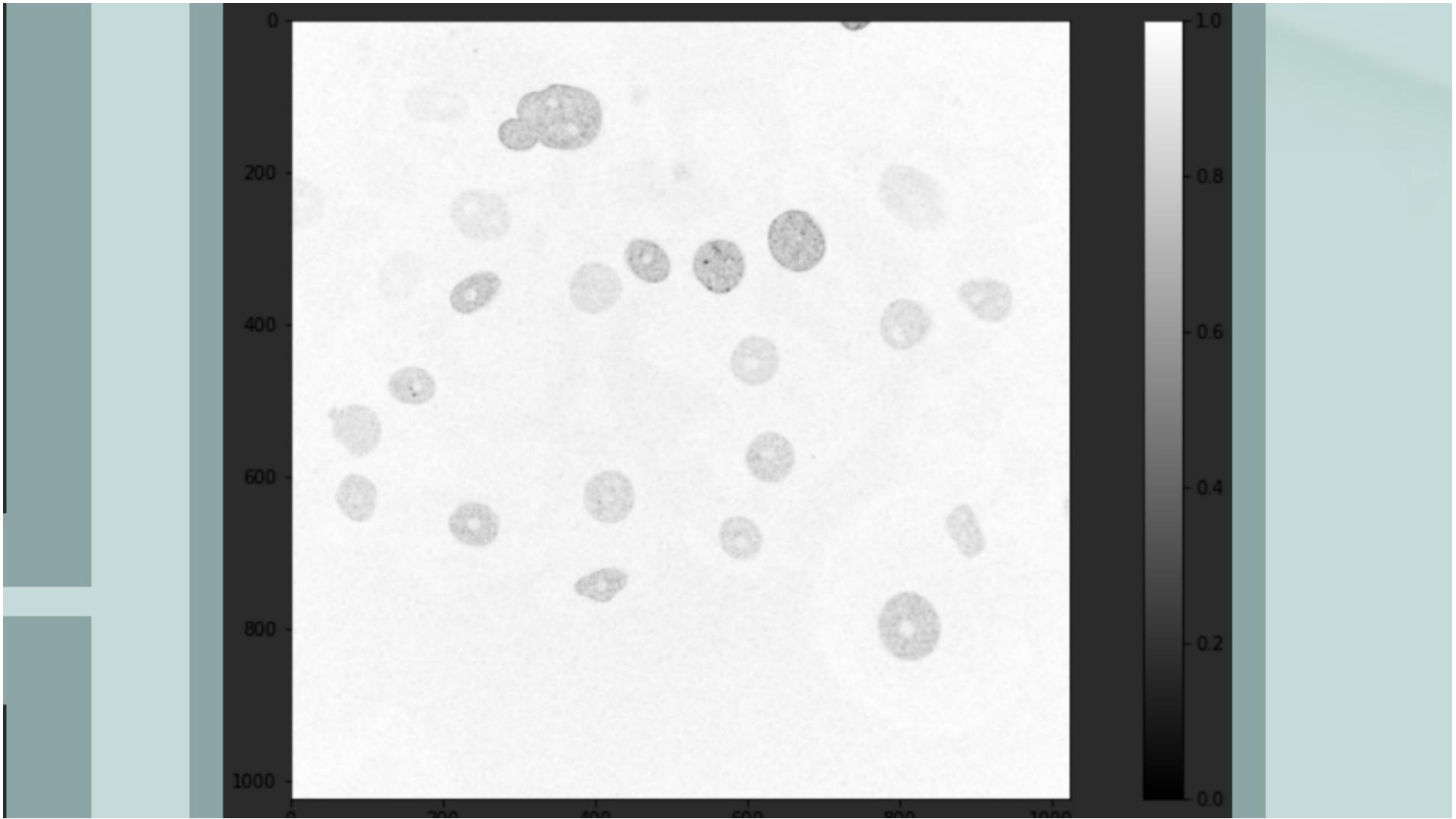
# Standard deviation

```
def standarddeviation(img, size): # calculates standard deviation of every pixel (image, size of filter mask)
    result = numpy.zeros(img.shape) # create empty array (zeros)
    n = (size - 1) // 2
    for p in ndindex(img.shape): # iterates over every pixel
        neighborhood_sum = 0
        mean = 0
        deviation = 0
        if p[0]-n >= 0 and p[1]-n >= 0 and p[0]+n <= img.shape[0] -1 and p[1]+n <= img.shape[1] -1: # no calculation of border pixels
            for q in ndindex(size, size): # iterates over filter mask (nxn neighborhood)
                i = p[0]-n + q[0]
                j = p[1]-n + q[1]
                neighborhood_sum += img[i,j]
            mean = neighborhood_sum/(size**2) # calculate mean in nxn neighborhood
            for q in ndindex(size, size): # iterates over filter mask (nxn neighborhood)
                i = p[0]-n + q[0]
                j = p[1]-n + q[1]
                deviation += (img[i,j] - mean)**2 # calculate deviation from mean

        result[p] = sqrt(1/(size**2) * deviation) # calculate standard deviation
    return result
sd_test = standarddeviation(img,3)  img: {ndarray: (1024, 1024)}
```

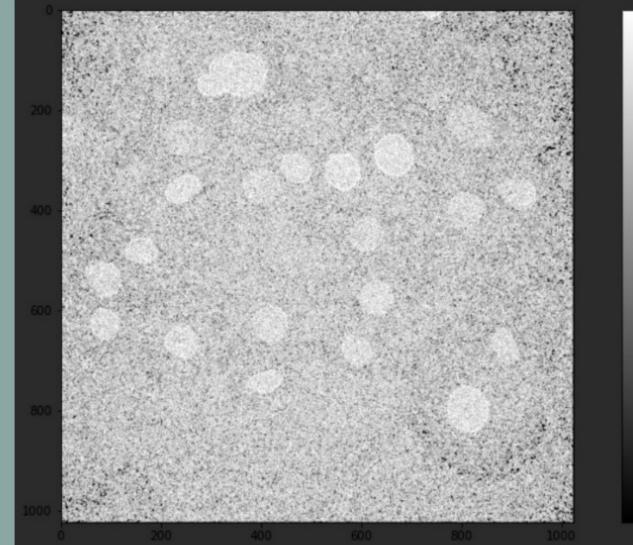
```
        deviation += (img[i,j] - mean)**2 # calculate deviation from mean
    result[p] = sqrt(1/(size**2) * deviation) # calculate standard deviation
return result
sd_test = standarddeviation(img,3) img: {ndarray: (1024, 1024)}
```

```
sd_2 = standarddeviation(img,3) # apply method for testing
sd_2 = sd_2/max(sd_2.flatten()) # normes standard deviation
similarity_2 = 1-sd_2 # calculates similarity
show_image(similarity_2, 15, 8)
```

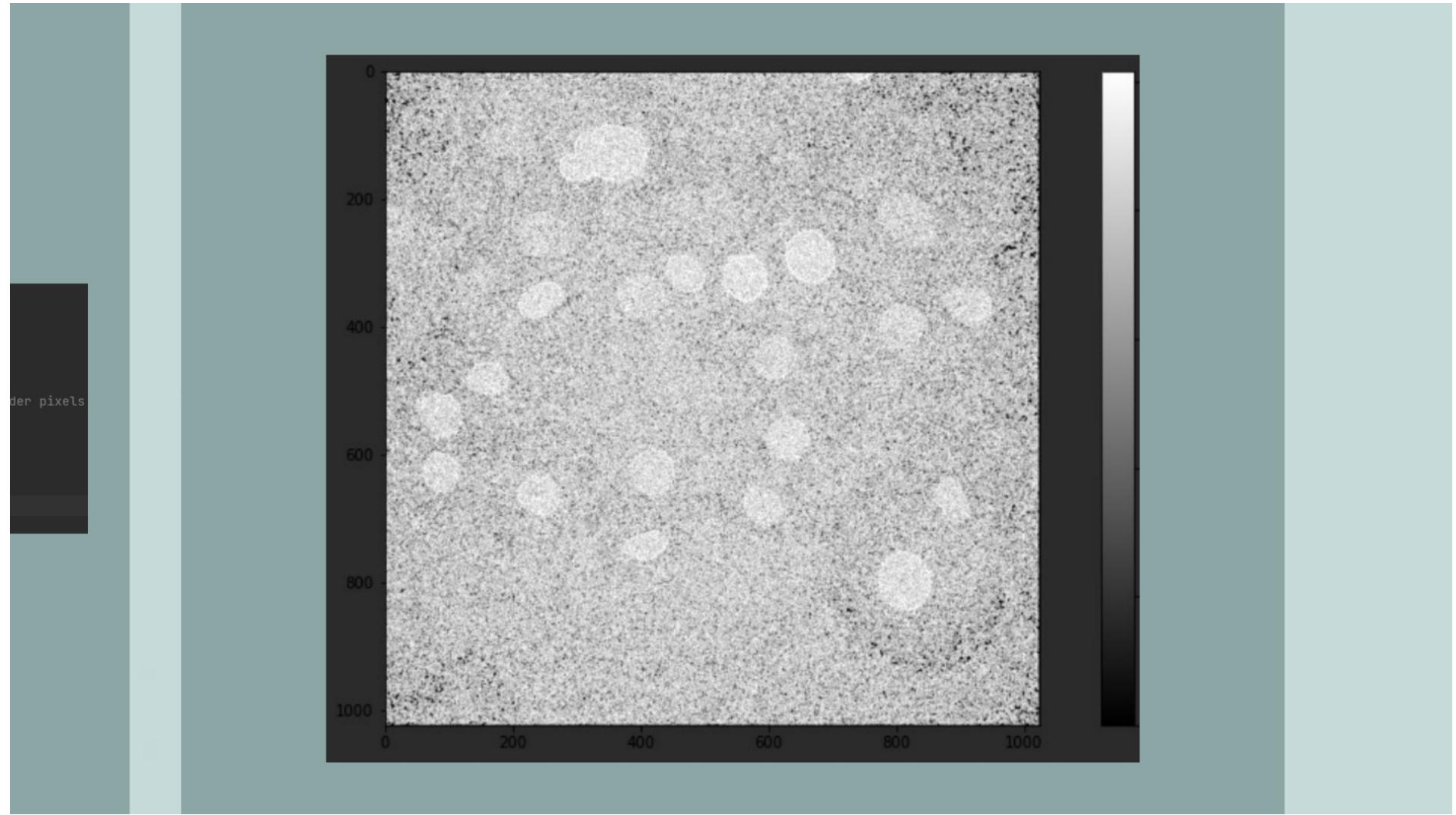


# Euclidean distance

```
def euclidean_n(img, size): # calculates maximum euclidean distance for every pixel in nxn neighborhood
    result = numpy.zeros(img.shape) # create empty array (zeros)
    n = (size - 1) // 2
    for p in ndindex(img.shape): # iterates over every pixel using a tuple
        neighborhood_distance = [] # creates a list for distances
        if p[0]-n >= 0 and p[1]-n >= 0 and p[0]+n <= img.shape[0] -1 and p[1]+n <= img.shape[1] -1: # no calculation of border pixels
            for q in ndindex(size, size): # iterates over nxn neighborhood
                i = p[0]-n + q[0]
                j = p[1]-n + q[1]
                neighborhood_distance.append((img[p]-img[i,j])) # adds euclidean distance to list
            result[p] = max(neighborhood_distance) # chooses maximum distance
    return result
```



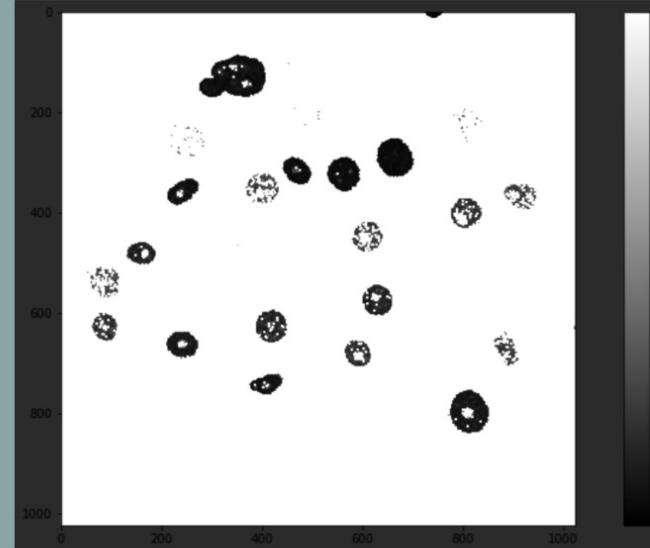
```
def euclidean_n(img, size): # calculates maximum euclidean distance for every pixel in nxn neighborhood
    result = numpy.zeros(img.shape) # create empty array (zeros)
    n = (size - 1) // 2
    for p in ndindex(img.shape): # iterates over every pixel using a tuple
        neighborhood_distance = [] # creates a list for distances
        if p[0]-n >= 0 and p[1]-n >= 0 and p[0]+n <= img.shape[0] -1 and p[1]+n <= img.shape[1] -1: # no calculation of border pixels
            for q in ndindex(size, size): # iterates over nxn neighborhood
                i = p[0]-n + q[0]
                j = p[1]-n + q[1]
                neighborhood_distance.append((img[p]-img[i,j])) # adds euclidean distance to list
        result[p] = max(neighborhood_distance) # chooses maximum distance
    return result
```



# Relative euclidean distance

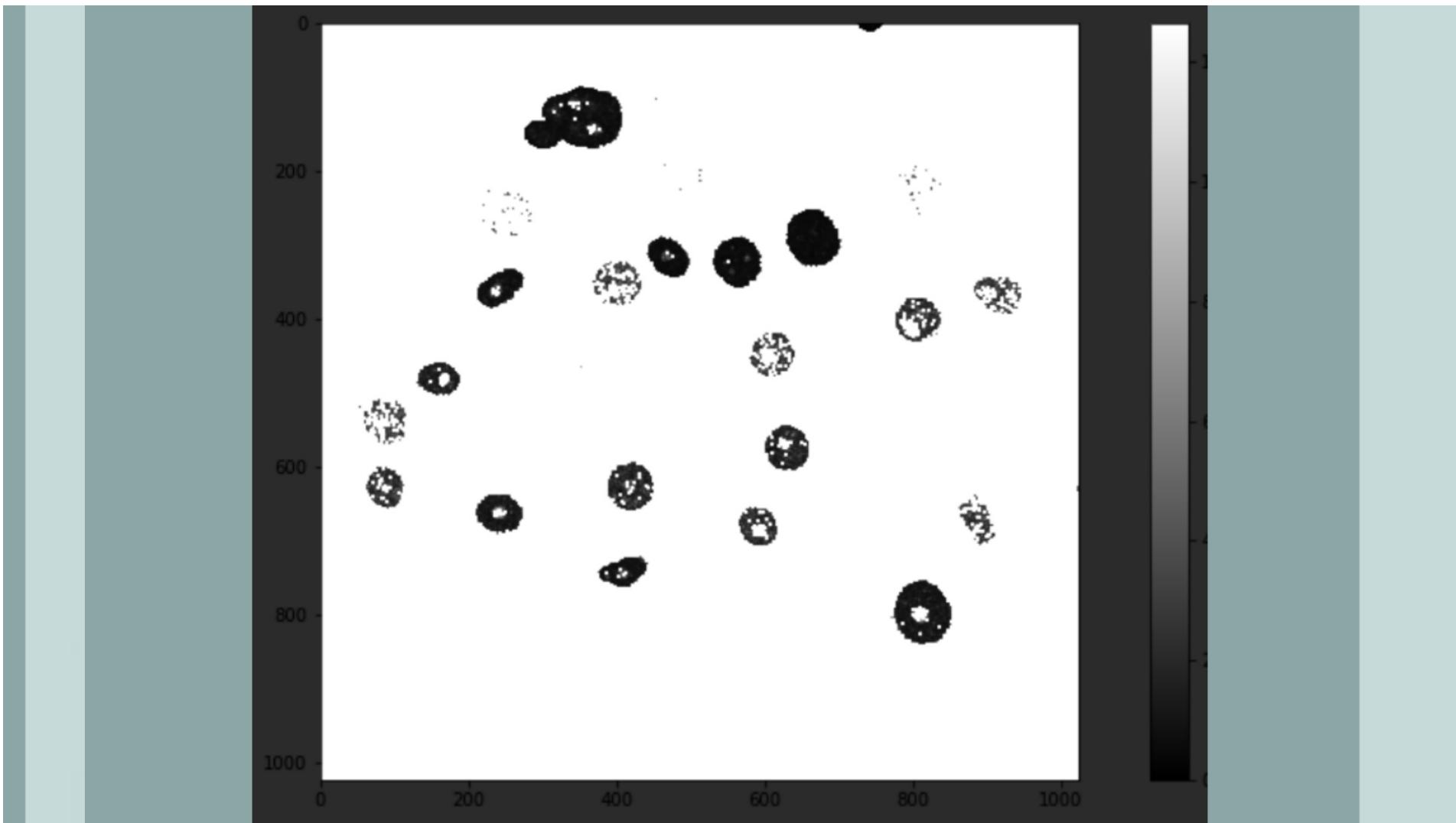
```
def euclidean_relative(img, size): # calculates maximum relative euclidean distance for every pixel in nxn neighborhood
    result = numpy.zeros(img.shape) # create empty array (zeros)
    n = (size - 1) // 2
    for p in ndindex(img.shape): # iterates over every pixel using a tuple
        neighborhood_distance = [] # creates a list for distances
        if p[0]-n >= 0 and p[1]-n >= 0 and p[0]+n <= img.shape[0] -1 and p[1]+n <= img.shape[1] -1: # no calculation of border pixels
            for q in ndindex(size, size): # iterates over nxn neighborhood
                i = p[0]-n + q[0]
                j = p[1]-n + q[1]
                neighborhood_distance.append((img[p]-img[i,j])/img[p]) # adds relative euclidean distance to list

        result[p] = max(neighborhood_distance) # chooses maximum distance
    return result
```



```
def euclidean_relative(img, size): # calculates maximum relative euclidean distance for every pixel in nxn neighborhood
    result = numpy.zeros(img.shape) # create empty array (zeros)
    n = (size - 1) // 2
    for p in ndindex(img.shape): # iterates over every pixel using a tuple
        neighborhood_distance = [] # creates a list for distances
        if p[0]-n >= 0 and p[1]-n >= 0 and p[0]+n <= img.shape[0] -1 and p[1]+n <= img.shape[1] -1: # no calculation of border pixels
            for q in ndindex(size, size): # iterates over nxn neighborhood
                i = p[0]-n + q[0]
                j = p[1]-n + q[1]
                neighborhood_distance.append((img[p]-img[i,j])/img[p]) # adds relative euclidean distance to list

        result[p] = max(neighborhood_distance) # chooses maximum distance
    return result
```

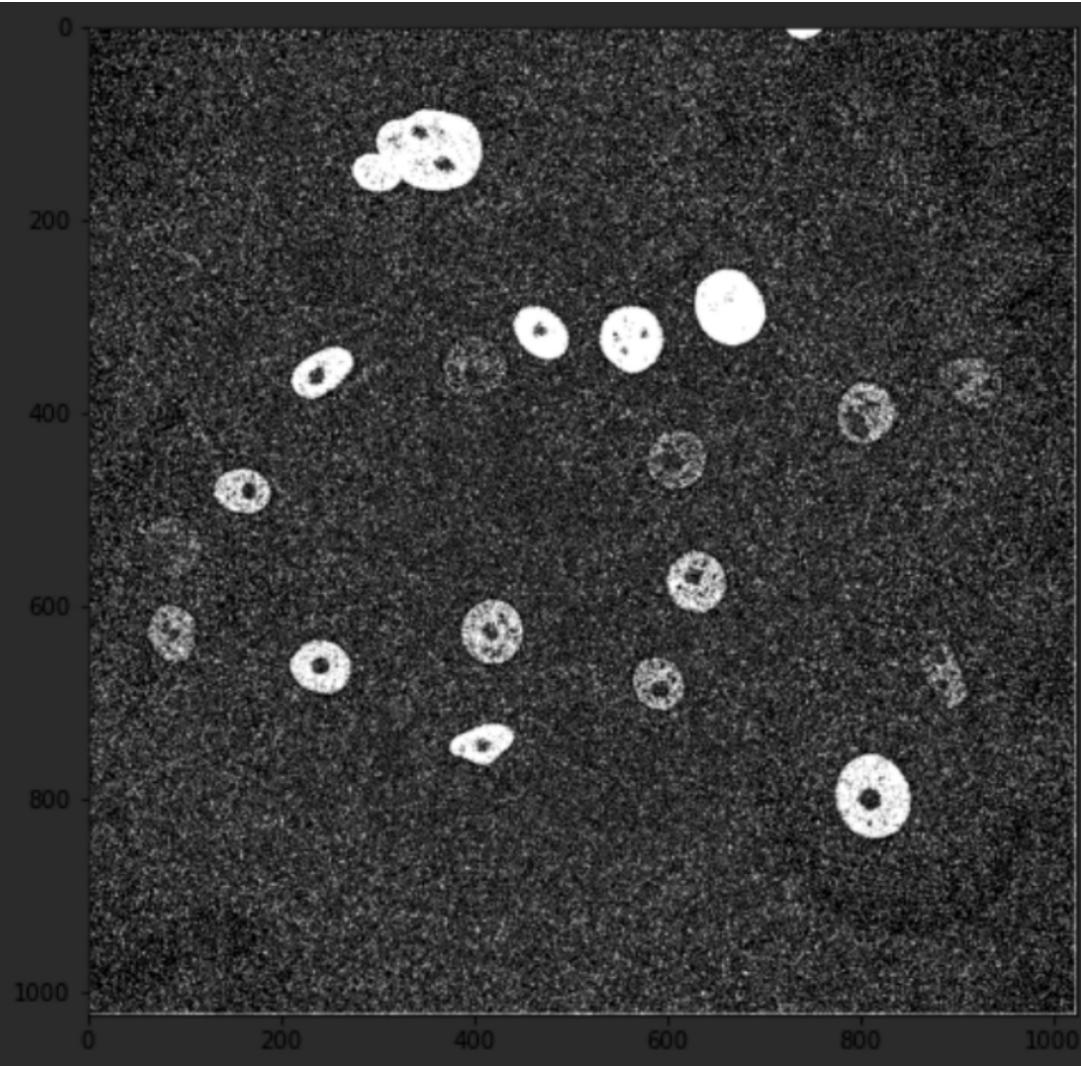


# selection algorithm

```
def seeds(img, T1, T2): # automatic seed selection algorithm
    result = numpy.zeros(img.shape)
    sd_seeds = standarddeviation(img, 3) # standard deviation
    sd_flat = sd_seeds.flatten() # standard deviation as 1D-array
    similarity_seeds = 1 - sd_seeds/max(sd_flat) # calculates similarity of every pixel to its neighbors
    eurel_seeds = euclidean_relative(img, 3) # relative euclidean distance of every pixel to its neighbors
    for p in ndindex(img.shape): # border pixel value is zero
        if similarity_seeds[p] > T1 and eurel_seeds[p] < T2: # compares pixel with threshold
            result[p] = 1 # assigns value 1 to seeds
    return result

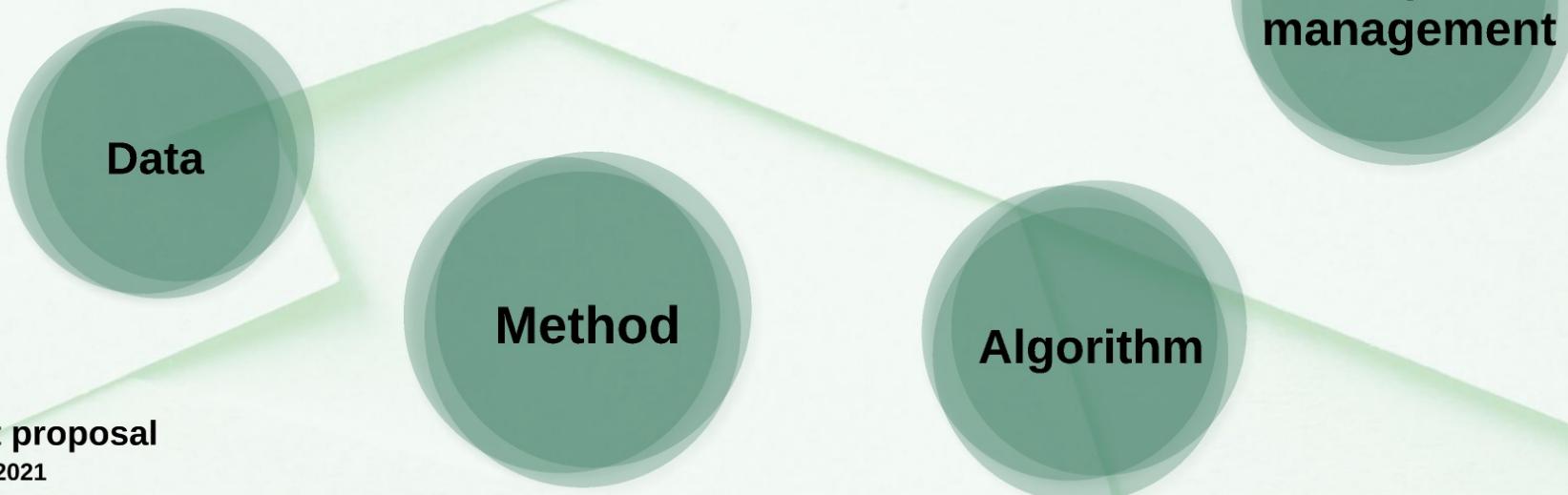
seed_final = seeds(img, 0.4, 20) img: {ndarray: (1024, 1024)}
show_image(seed_final, 15, 8) seed_final: {ndarray: (1024, 1024)}
```





# **Cell nuclei segmentation**

via implementation and evaluation of region growing



**Project proposal**

12th May 2021

Data Analysis MoBi SS2021

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Topic 04: Biomedical image analysis

Group 04: Marie Becker, Ina Jung, Laura Kaschnitz, Johanna Möller

# Project Management

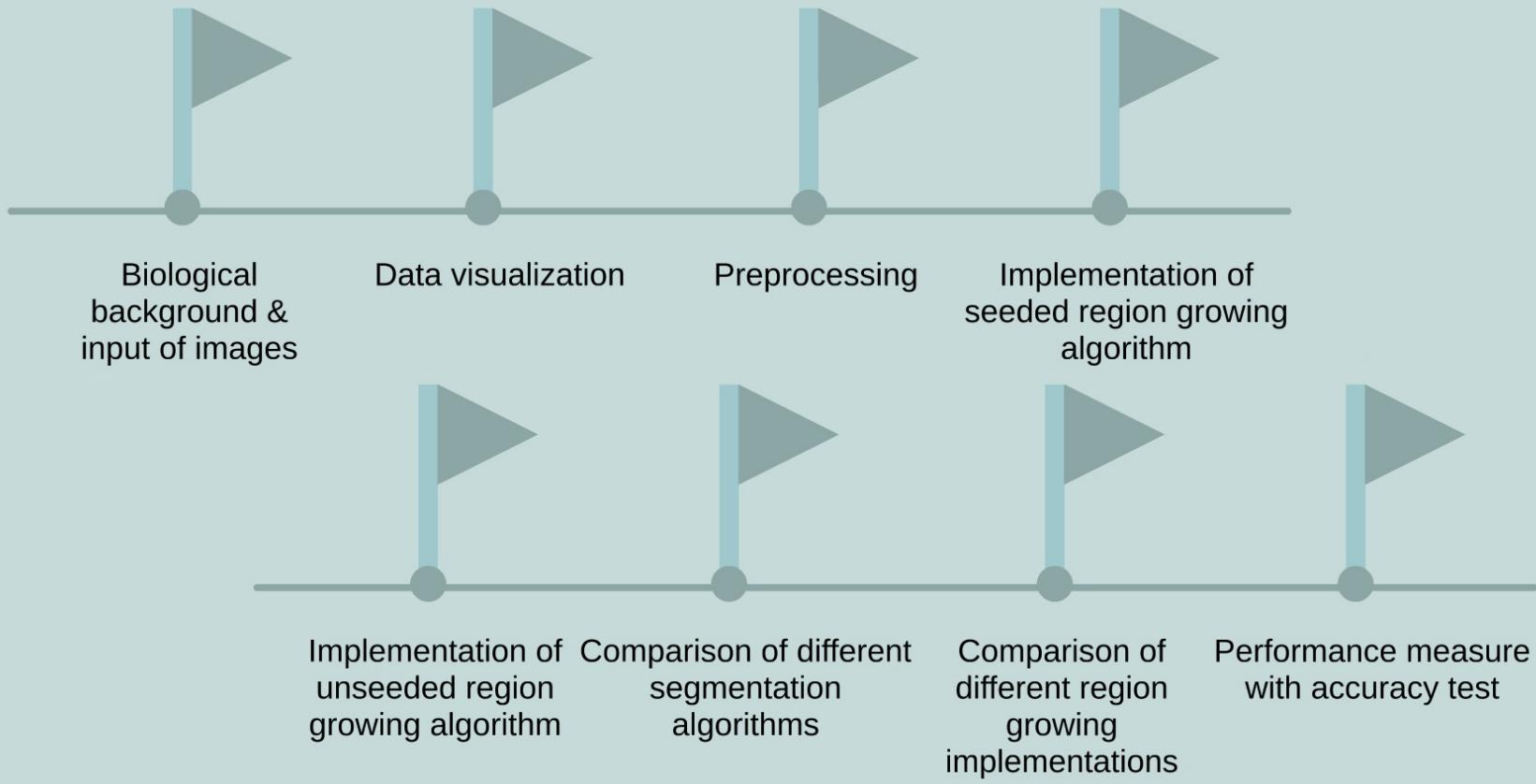
Agile project management

Implementation in GitHub

Timeline

Teamwork

# Milestones



# Calendar

Project timetable		
Sprint 1	Sprint 2	Sprint 3
<ul style="list-style-type: none"><li>• Biological background &amp; input of images</li><li>• Data visualization</li></ul>		

# Project Management

Agile project management

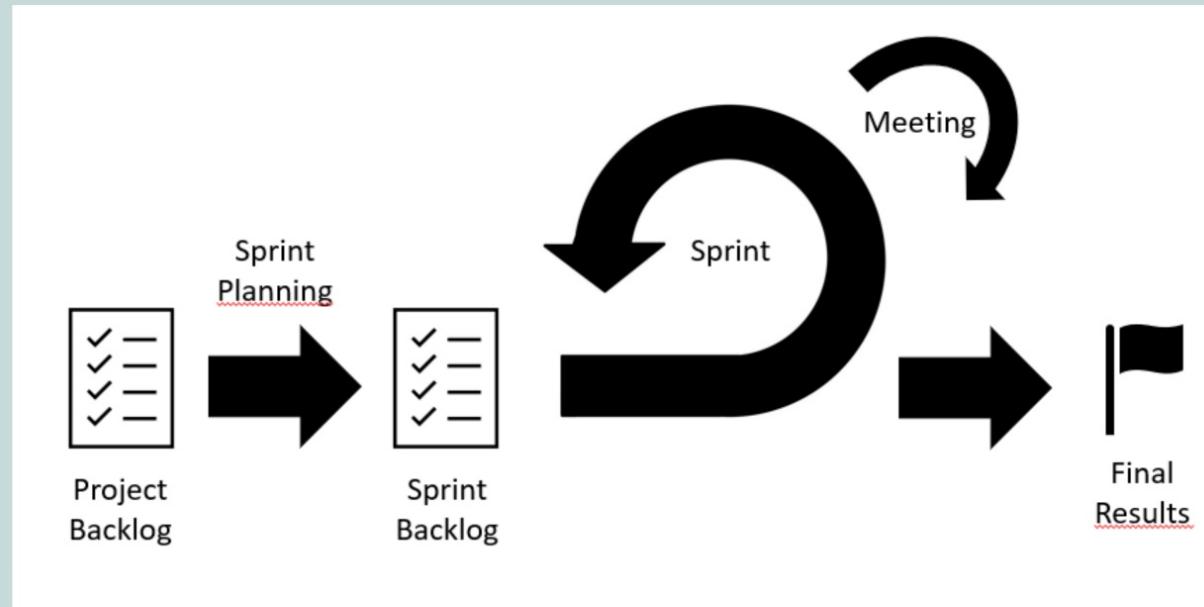
Implementation in GitHub

Timeline

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# Agile Project Management

## Scrum



# ToDo

14 To do

- Region growing algorithm #8 opened by marie3003  
↳ 4. Implement seeded region growing alg...
- Region merging algorithm #9 opened by marie3003  
↳ 4. Implement seeded region growing alg...
- Compare different image segmentation methods

Automated as To do

# Sprint 1

8 Sprint1

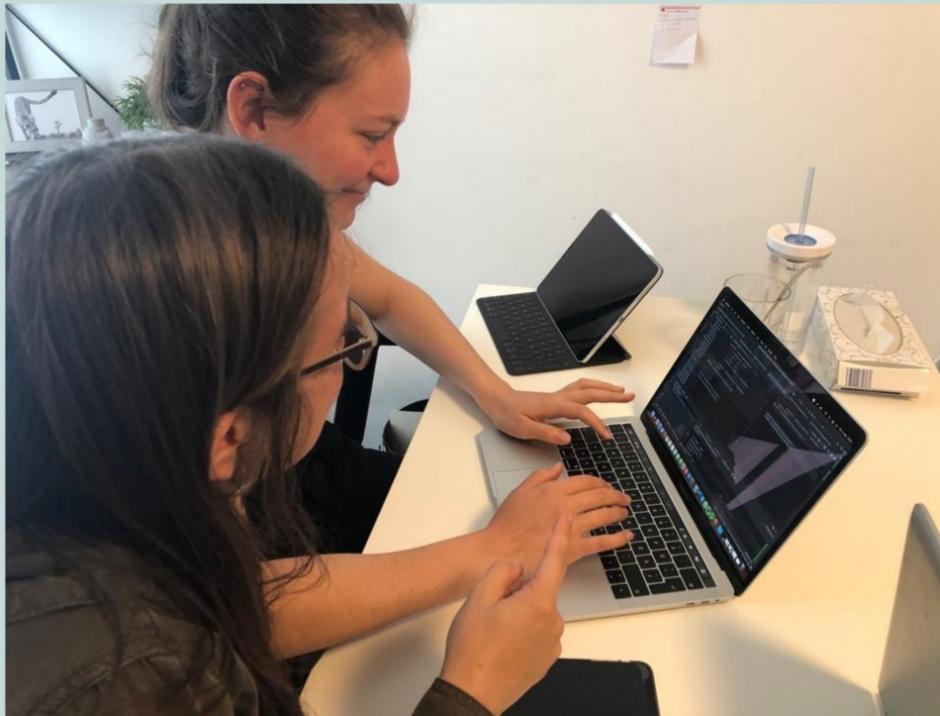
- 0. Presentation
- Introduction into the method #19 opened by LauraKaschnitz  
↳ 0. Presentation
- Presentation of current results #22 opened by marie3003  
↳ 0. Presentation
- Milestones, Sprints - Structure of the project

# Done

3 Done

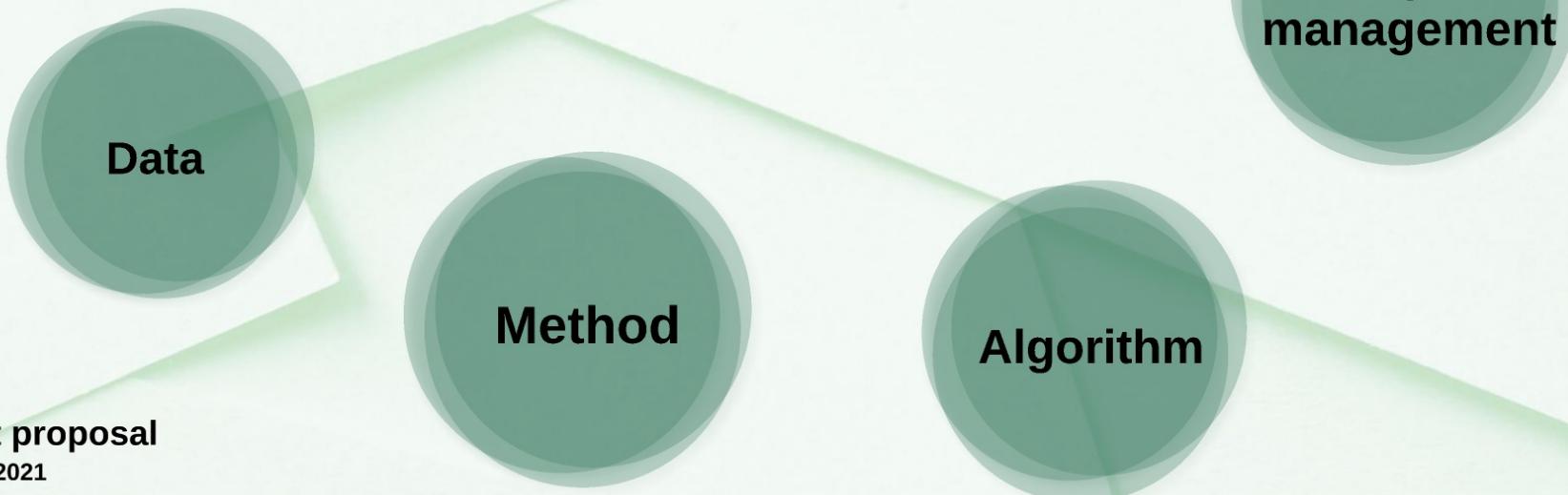
- Automatic seed detection algorithm #7 opened by marie3003  
↳ 4. Implement seeded region growing alg...
- Upload images #26 opened by johannamoeller  
↳ 1. Biological Background and Input of Im...
- Layout #25 opened by LauraKaschnitz  
↳ 0. Presentation

# Pair programming



# Cell nuclei segmentation

via implementation and evaluation of region growing



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