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DRESDEN LEIPZIG

CENTER FOR SCALABLE DATA ANALYTICS AND ARTIFICIAL INTELLIGENCE

Python: File handling & Image Visualization

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SACHSEN

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Quiz: Recap
What would this Python code result in?

```
files = ['a.tif', 'b.btif', 'c.tif', 'd.tif’]

files[1]
```

'a.tif'

'b.tif'

'c.tif'

'd.tif'

Quiz: Recap
What would this Python code result in?

```
files = ['a.tif', 'b.btif', 'c.tif', 'd.tif']

files[-1]
```

'a.tif'

'b.tif'

'c.tif'

'd.tif'

Working with files in folders

Key-skill when it comes to automating data analysis tasks

[2]:
```
# define the location of the folder to go through
directory = 'data_banana/'

# get a list of files in that folder
file_list = os.listdir(directory)

file_list
```

[2]:
```
['banana0002.tif',
 'banana0003.tif',
 'banana0004.tif',
 'banana0005.tif',
 'banana0006.tif',
 'banana0007.tif',

 'banana0026.tif',
 'image_source.txt']
```

It's just a Python list!

There are not just images inside

Filtering file lists

To focus on image data, we need to filter the file list

```
[3]: image_file_list = [file for file in file_list if file.endswith(".tif")]

     image_file_list
```

```
[3]: ['banana0002.tif',
      'banana0003.tif',
      'banana0004.tif',
      'banana0005.tif',
      'banana0006.tif',
      'banana0007.tif',

      'banana0026.tif']
```

A for-loop in a single line

Filtering file lists

To focus on image data, we need to filter the file list

[4]:
```
# go through all files in the folder
for file in file_list:
    # if the filename is of a tif-image, print it out
    if file.endswith(".tif"):
        print(file)
```

```
banana0002.tif
banana0003.tif
banana0004.tif
banana0005.tif
banana0006.tif
banana0007.tif

banana0026.tif
```

Filtering file lists

To focus on image data, we need to filter the file list

[5]:
```
# go through all files in the folder
for file in file_list:
    # if the filename is of a tif-image, print it out
    if file.endswith(".tif"):
        print(file)

        # store the image
        image = imread(directory + file)

        # show the image
        stackview.imshow(image)
```

Side note: comments

Your code will become longer…
Consider structuring it and putting comments and empty lines in between.

```
for file in file_list:
    if file.endswith(".tif"):
        print(file)
        image = imread(directory + file)
        stackview.imshow(image)
```

```
# go through all files in the folder
for file in file_list:
    # if the filename is of a tif-image, print it out
    if file.endswith(".tif"):
        print(file)

        # store the image
        image = imread(directory + file)

        # show the image
        stackview.imshow(image)
```

Image visualization

In Python there are many ways to visualize image data.
We focus on Jupyter Notebook compatible ways for now.

Before visualizing image data, get an idea about the dataset.

```
[2]: image = imread("../day2.1_image_segmentation/data/BMP4blastocystC3-cropped_resampled_8bit.tif")
     image.shape

[2]: (86, 396, 393)
```

Image.shape tells you the dimensions of an image

Not all image viewers support 3D data

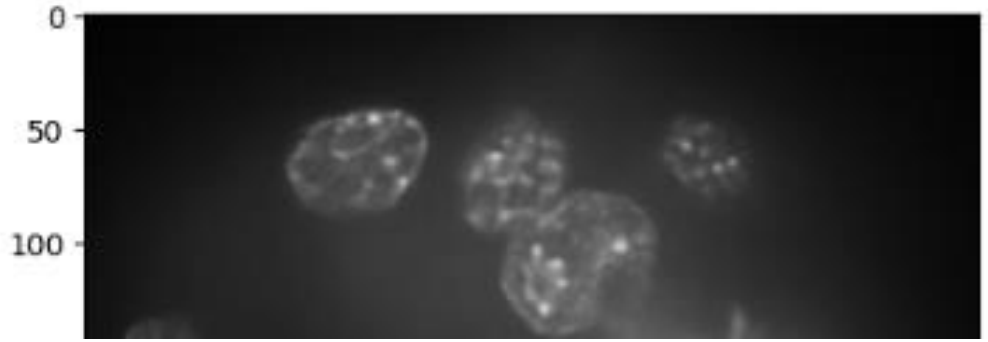
Image visualization

scikit-image's `imshow()` (originating from matlab) can only visualize 2 dimensions.

```
from skimage.io import imread, imshow
```

```
[4]: center_slice = int(image.shape[0] / 2)

     imshow(image[center_slice])
```

```
[4]: <matplotlib.image.AxesImage at 0x2c9b9164820>
```



The first shape-dimension – 2 is the center plane in Z

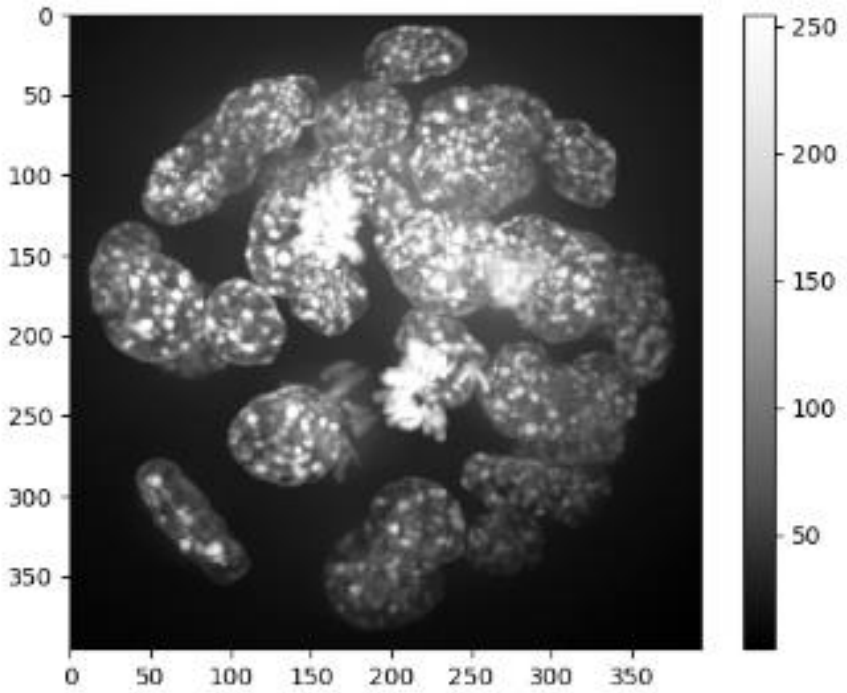
Image data in Python is commonly organized in Z-Y-X dimension order.

Image visualization

Stackview can show 3D images (by applying a projection along Z)

```
import stackview
```

```
stackview.insight(image)
```



shape	(86, 396, 393)
dtype	uint8
size	12.8 MB
min	0
max	255

dimensions

Basic statistics

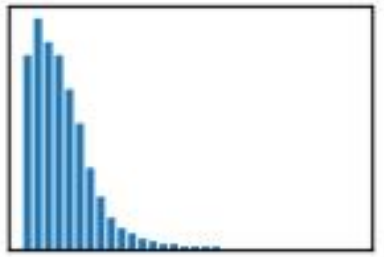


Image visualization

Stackview also has some interactive tools

```
binary_image = image > 80
```

```
stackview.slice(image)
```


```
stackview.picker(image)
```


```
stackview.curtain(image, binary_image)
```

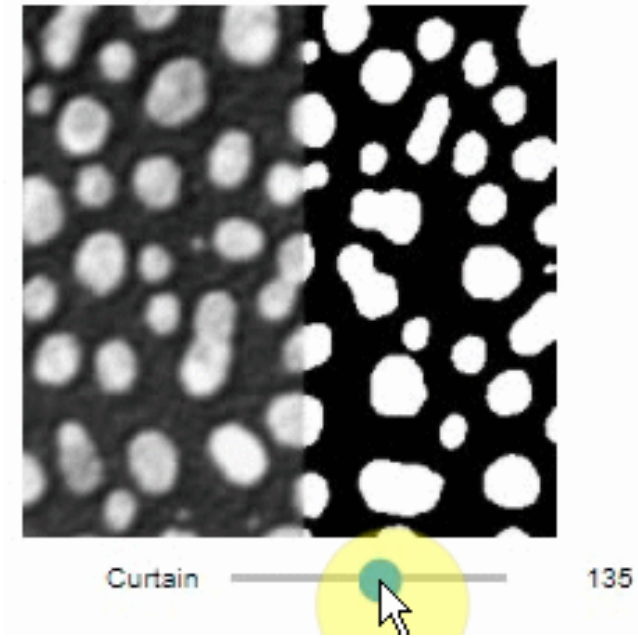


Image visualization

Stackview also has some interactive tools

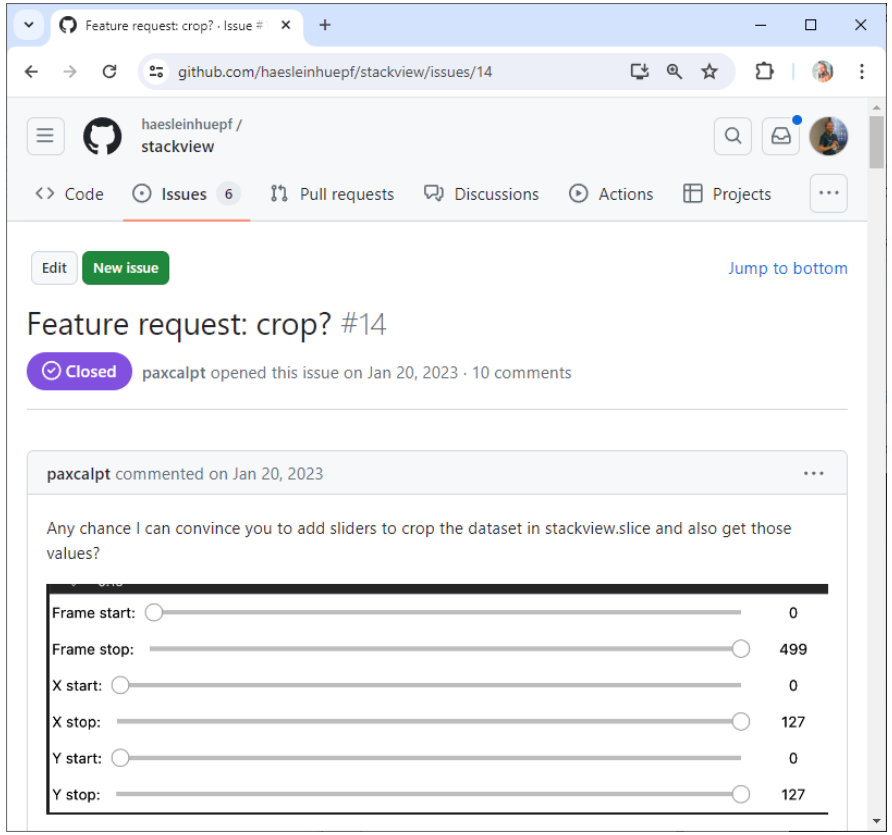
```
stackview.switch(
    {
        "Lysosomes":    lysosomes_channel,
        "Mitochondria":mitochondria_channel,
        "Nuclei":       nuclei_channel
    },
    colormap=[
        "pure_magenta",
        "pure_green",
        "pure_blue"
    ],
    toggleable=True
)
```

Identical but less readable:

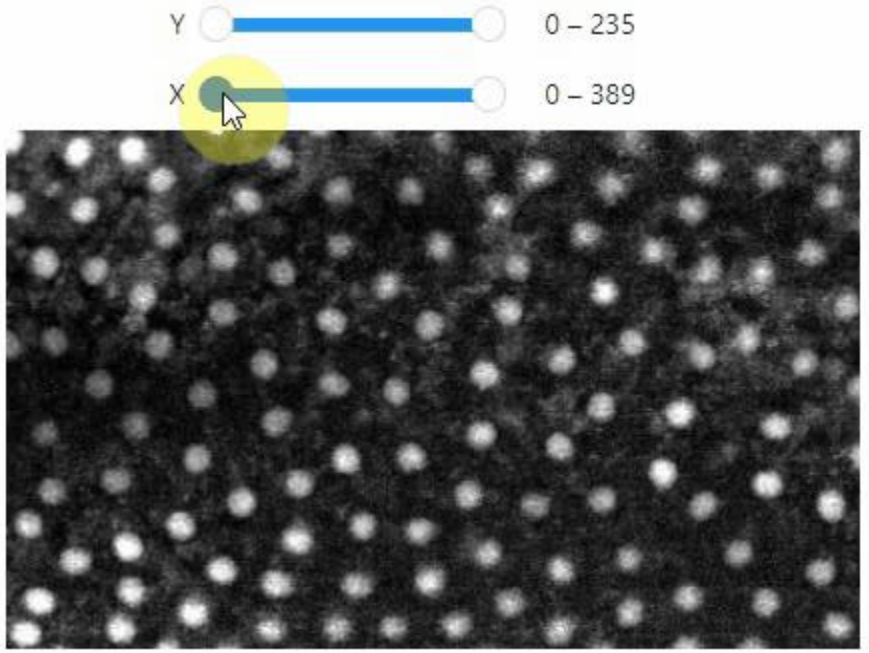
```
stackview.switch({"Lysosomes":lysosomes_channel,
"Mitochondria":mitochondria_channel,"Nuclei":nuclei_channel},
colormap=["pure_magenta", "pure_green", "pure_blue"], toggleable=True)
```

Image Visualization

Stackview is developed for microscopists. If something doesn‘t work as you expect, communicate it :-)




```
crop_widget = stackview.crop(image_stack, continuous_update=True)
crop_widget
```



Loading domain-specific file formats

Common Python libraries cannot open file formats such as OME-TIF, CZI, LIF, ND2… Solution: aicsimageio

From the Allen Institute for Cell Science

```
from aicsimageio import AICSImage
```

```
aics_image = AICSImage("data/EM_C_6_c0.ome.tif")
aics_image
```

```
<AICSImage [Reader: OmeTiffReader, Image-is-in-Memory: False]>
```

Loading domain-specific file formats

Aicsimageio supports dimension names and physical voxel sizes.

```
aics_image.shape
```

What's width, height, depth and time?

```
(1, 1, 256, 256, 256)
```

```
aics_image.dims
```

```
<Dimensions [T: 1, C: 1, Z: 256, Y: 256, X: 256]>
```

Key feature when working with big data!

Lazy loading (virtual stacks)

Allows processing files larger than computer memory.

```
np_image = aics_image.get_image_data("ZYX", T=0)
np_image.shape
```

```
(256, 256, 256)
```

Loading domain-specific file formats

Aicsimageio supports dimension names and physical voxel sizes.

```
aics_image.physical_pixel_sizes
```

```
PhysicalPixelSizes(Z=0.1678467289719626, Y=0.16776018346253663, X=0.16776018346253663)
```

```
def get_voxel_size_from_aics_image(aics_image):
    return (aics_image.physical_pixel_sizes.Z,
            aics_image.physical_pixel_sizes.Y,
            aics_image.physical_pixel_sizes.X)
```

Such a helper-function may be different from project to project

```
get_voxel_size_from_aics_image(aics_image)
```

```
(0.1678467289719626, 0.16776018346253663, 0.16776018346253663)
```

Loading domain-specific file formats

Common Python libraries cannot open file formats such as OME-TIF, CZI, LIF, ND2… Solution: aicsimageio

In case your file format is not supported, it gives hints what to install.

```
czi_image = AICSImage("data/PupalWing.czi")
czi_image.shape
```

```
Attempted file (C:/structure/code/BIDS-training-2024/day1.2_file_handling/data/PupalWing.czi) load with reader: aicsimageio.readers.czi_reader.CziReader failed with error: aicspylibczi is required for this reader. Install with `pip install 'aicspylibczi>=3.1.1' 'fsspec>=2022.7.1'`
Attempted file (C:/structure/code/BIDS-training-2024/day1.2_file_handling/data/PupalWing.czi) load with reader: aicsimageio.readers.bioformats_reader.BioformatsReader failed with error: bioformats_jar is required for this reader. Install with `pip install bioformats_jar` or `conda install bioformats_jar`
```

```
czi_image = AICSImage("data/PupalWing.czi")
czi_image.shape
```

```
(1, 1, 80, 520, 692)
```

```
np_czi_image = czi_image.get_image_data("ZYX", T=0)
np_czi_image.shape
```

```
(80, 520, 692)
```

```
get_voxel_size_from_aics_image(czi_image)
```

```
(1.0, 0.20476190476190476, 0.20476190476190476)
```

Working with files in the cloud

Example nextcloud / owncloud

Install another Python library into your environment

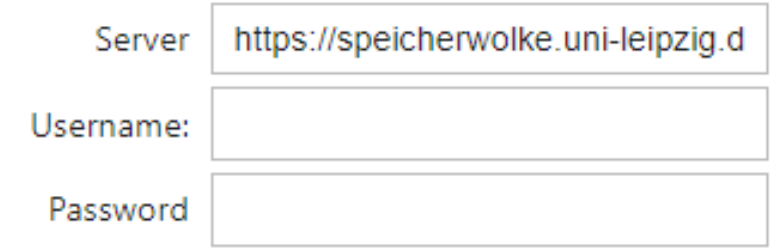
```
conda activate devbio-napari-env
pip install pyncclient
```

```
[2]: import ipywidgets as widgets
     import nextcloud_client
```

Login-form

```
[3]: server_widget = widgets.Text(value='https://speicherwolke.uni-leipzig.de', description='Server')
     username_widget = widgets.Text(description='Username:')
     password_widget = widgets.Password(description='Password')

     widgets.VBox([server_widget, username_widget, password_widget])
```

[3]:



Actually logging in

```
[6]: ncc = nextcloud_client.Client(server_widget.value)
     ncc.login(username_widget.value, password_widget.value)
```

Working with files in the cloud

Listing files in a remote folder

[7]:
```
# enter a folder on the owncloud
remote_folder = "/data/"

for f in ncc.list(remote_folder):
    print (f.path)
```

```
/data/blobs.tif
```

Uploading a file

[13]:
```
ncc.put_file(remote_folder, local_file_to_upload)
```

[13]:
```
True
```

Downloading a file

[8]:
```
# enter the source file here
remote_source_file = '/data/blobs.tif'
# enter the destination
local_file = 'blobs.tif'

ncc.get_file(remote_path=remote_source_file,
             local_file=local_file)
```

[8]:
```
True
```

Listing files again

[14]:
```
for f in ncc.list(remote_folder):
    print (f.path)
```

```
/data/blobs.tif
/data/blobs_labels.tif
```

Quiz: Large Language Models

How often do you use ChatGPT or similar AI-based tools?

monthly

weekly

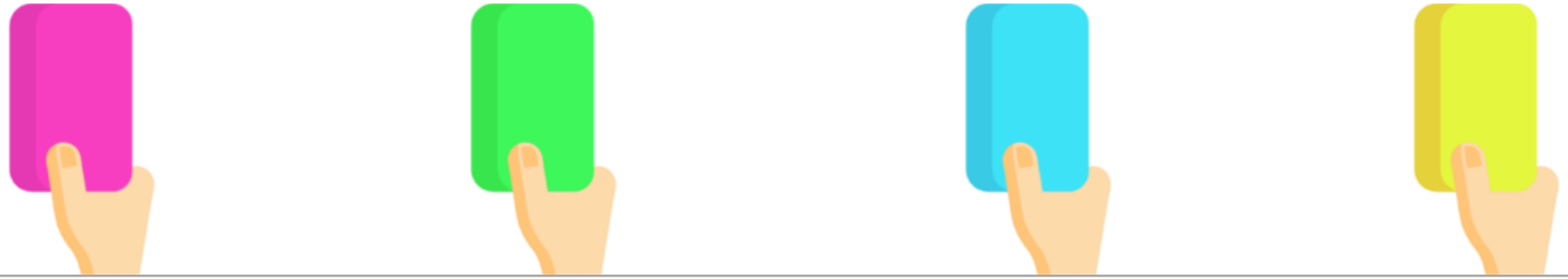
daily

hourly

Quiz: Large Language Models

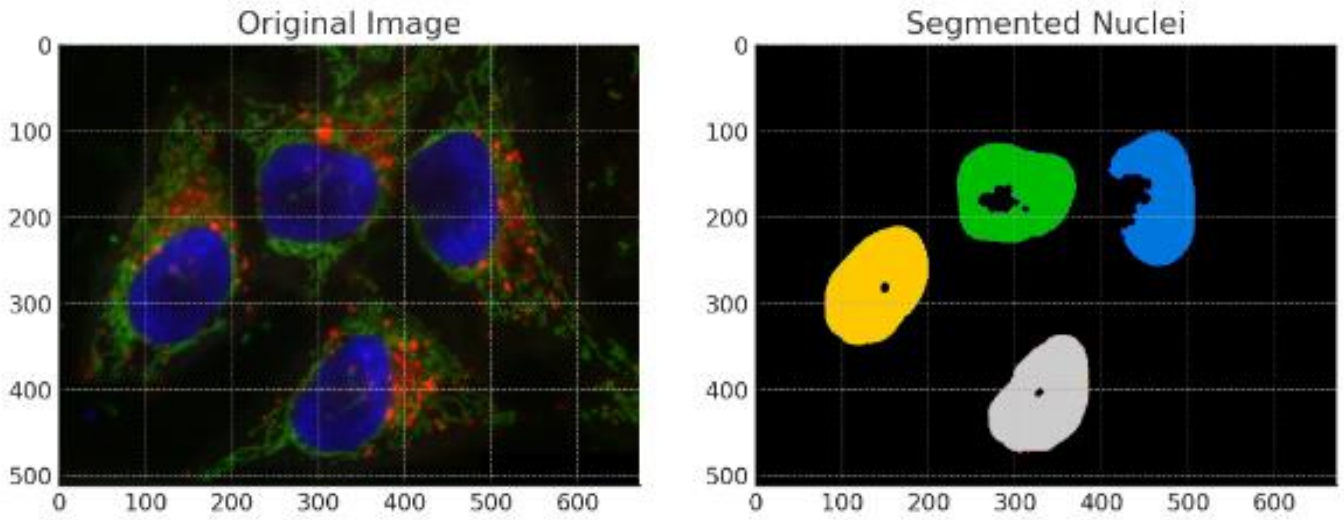
What do you use ChatGPT and similar AI-based tools for?

Emails Paperwork Code other



Bio-image Analysis

My job …

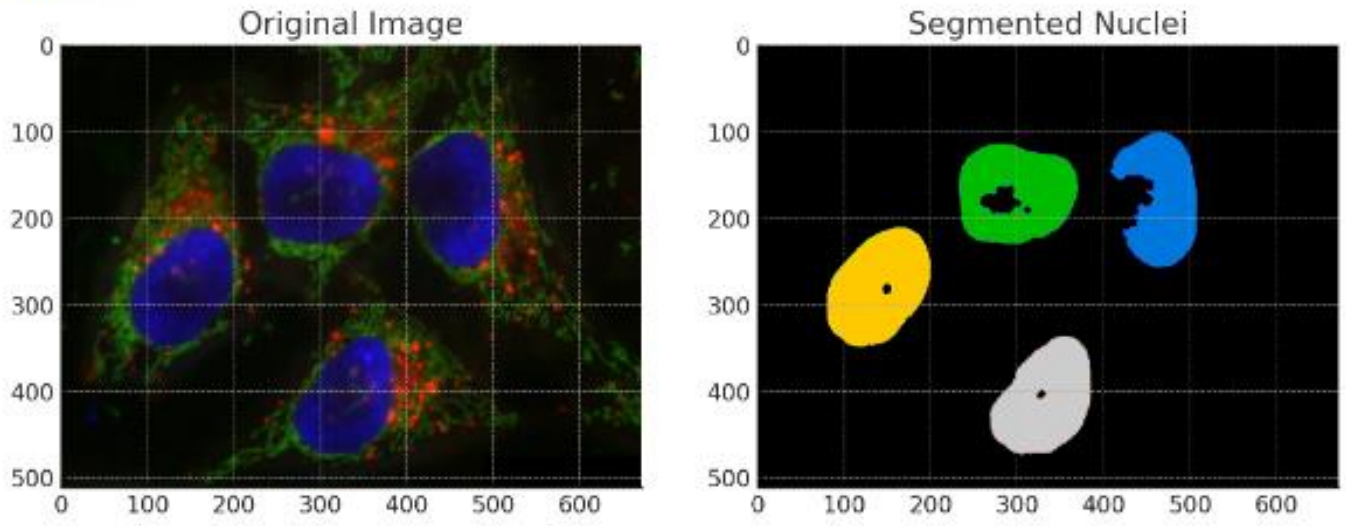


Bio-image Analysis using Large-Language Models

My job is changing, since we have ChatGPT

Prompt:
“Please segment the nuclei in the blue channel of the image”

ChatGPT



The segmentation of the nuclei has been performed on the blue channel of the image. The right side of the figure shows the segmented nuclei in different colors, each corresponding to a different nucleus in the field of view. Each color represents a unique label assigned to the individual nuclei detected in the image.

Analysis

```
from skimage import io, filters, measure, morphology
import matplotlib.pyplot as plt
import numpy as np

# Load the image
image = io.imread('/mnt/data/hela-cells-8bit.png')

# Since it's a color image, we'll just take the blue chann
blue_channel = image[:, :, 2]

# Threshold the image to get the binary image for nuclei
thresh = filters.threshold_otsu(blue_channel)
binary = blue_channel > thresh

# Remove small objects and perform morphological closing t
cleaned = morphology.remove_small_objects(binary, 50)
closed = morphology.closing(cleaned, morphology.disk(3))

# Label the image
label_image = measure.label(closed)

# Plot the original image and the binary image
fig, ax = plt.subplots(1, 2, figsize=(12, 6))
ax[0].imshow(image)
ax[0].set_title('Original Image')
ax[1].imshow(label_image, cmap='nipy_spectral')
ax[1].set_title('Segmented Nuclei')
plt.show()
```

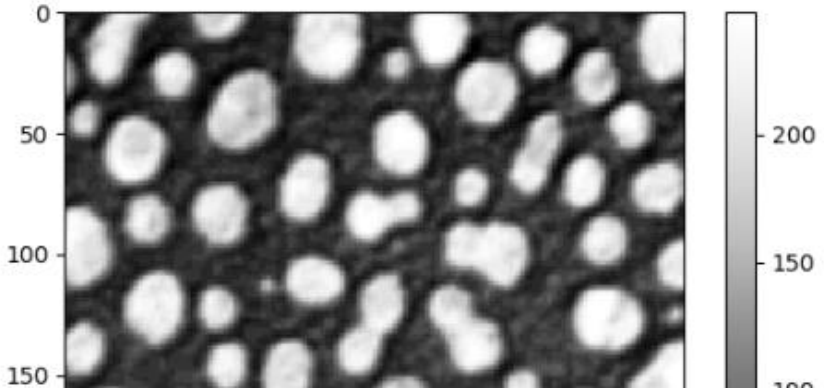
Coding assistance: BiA-Bob

Use `%bob` ... to ask for small things.

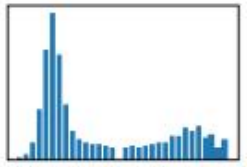
In [2]:
```
%bob Load the blobs.tif image file and show it.
```

In [3]:
```
from skimage.io import imread
import stackview

image = imread('blobs.tif')
stackview.insight(image)
```

Out[3]:



shape	(254, 256)
dtype	uint8
size	63.5 kB
min	8
max	248



Coding assistance: BiA-Bob

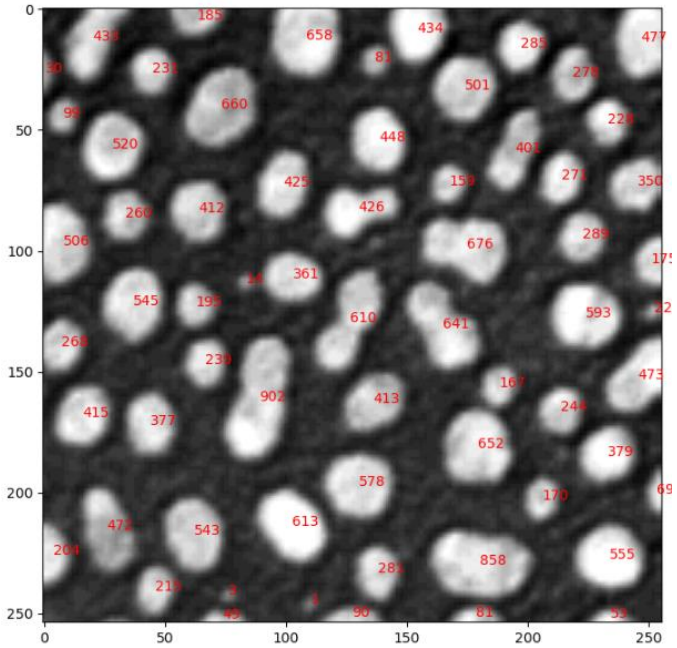
Use `%%bob ...` to ask for complex analysis tasks.

In [4]:

```
%%bob
Load the blobs.tif grayscale image from disc.
Segment the nuclei-like objects in it using
Otsu's method and connected component labeling.
Measure area of the segmented labels.
Print the area on top of the labels.
```

In [5]:

```
from skimage.io import imread
from skimage.filters import threshold_otsu
from skimage.measure import label, regionprops
import matplotlib.pyplot as plt
import stackview

# Load the image
image = imread('blobs.tif')

# Apply Otsu's thresholding
threshold = threshold_otsu(image)
binary_image = image > threshold

# Perform connected component labeling
labels = label(binary_image)

# Measure area of the segmented labels
props = regionprops(labels)
areas = [prop.area for prop in props]

# Create an image with labels and area values on top
fig, ax = plt.subplots(figsize=(8, 8))
ax.imshow(image, cmap='gray')

for prop in props:
    y, x = prop.centroid
    area = prop.area
    ax.text(x, y, f"{area}", color='red')

stackview.curtain(image, labels)
```



Coding assistance: BiA-Bob

- Bob adds a new cell to your code. If you ask it 3 times, it will make 3 cells.

- Clean up from time to time!

```
[3]: %bob load blobs.tif and show it
```

We need to load an image named `blobs.tif` from the disk and display it.

```
[ ]: from skimage.io import imread
     import stackview

     # Load the image from disk
     image = imread('blobs.tif')

     # Display the image
     stackview.insight(image)
```

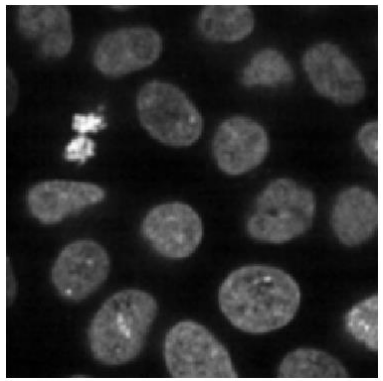
```
[ ]: from skimage.io import imread
     import stackview

     # Load the image file
     image = imread('blobs.tif')

     # Display the image
     stackview.insight(image)
```

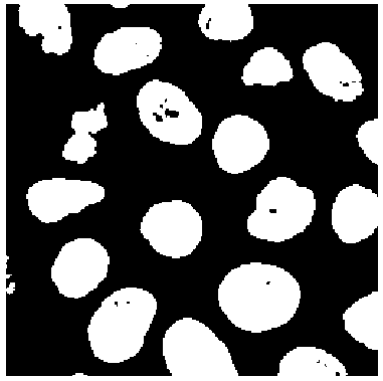
```
[ ]: from skimage.io import imread
     import stackview

     # Load image from disk
     image_blobs = imread('blobs.tif')

     # Display the image
     stackview.insight(image_blobs)
```

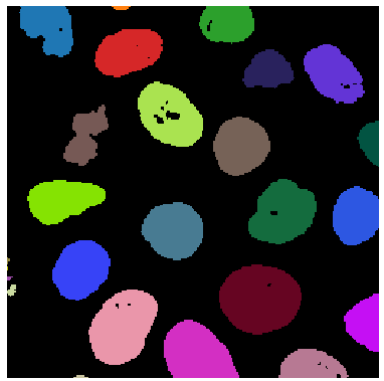
Click on the bar here and enter **dd**

Benchmarking LLMs for Bio-image Analysis

Use case: segment the image and measure the average area of objects.







	area
0	955.0
1	31.0
2	815.0
3	1166.0
4	1135.0
5	680.0
6	1374.0
7	712.0

858.04

Unit-test pass-rate (n=10):

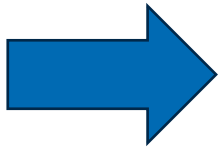
	reference	**gpt-4-turbo-2024-04-09**	Claude-3-opus-20240229	gpt-4-1106-preview	gpt-3.5-turbo-1106	gemini-pro	codellama
workflow_segmentation_measurement_summary	1.0	0.9	1.0	0.8	0.5	0.5	0.1

https://www.biorxiv.org/content/10.1101/2024.04.19.590278v1
https://github.com/haesleinhuepf/human-eval-bia

Benchmarking LLMs for Bio-image Analysis

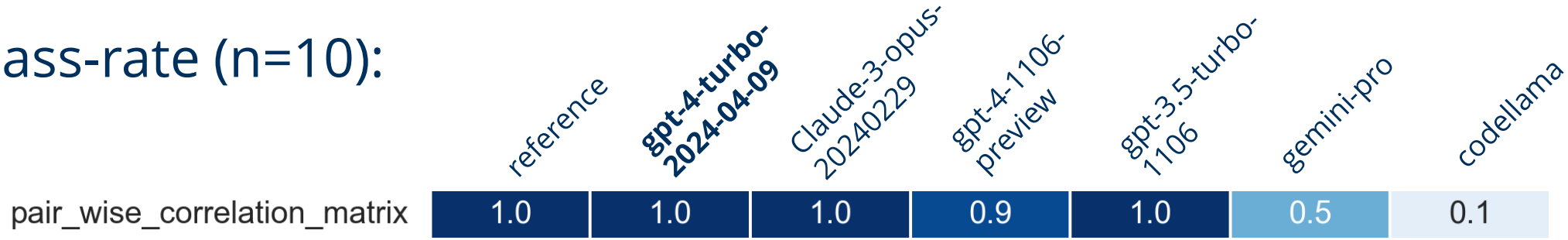
Use-case: correlation matrix

	a	b	c	d	e
0	1.600000	0.100000	1.600000	1.700000	1.700000
1	2.300000	0.200000	2.300000	2.400000	2.400000
2	2.600000	0.300000	2.600000	2.400000	2.400000
3	3.700000	0.300000	3.700000	3.600000	3.600000
4	3.400000	0.400000	3.400000	3.500000	3.500000
5	3.900000	0.400000	3.900000	3.900000	3.900000
6	4.300000	0.400000	4.300000	4.400000	4.400000
7	4.300000	0.500000	4.300000	4.200000	4.200000
8	4.000000	0.500000	4.000000	4.100000	4.100000
9	5.100000	0.500000	5.100000	5.000000	5.000000
10	5.200000	0.600000	5.200000	5.100000	5.100000
11	5.300000	0.600000	5.300000	5.400000	5.400000
12	5.500000	0.600000	5.400000	5.600000	5.600000



	a	b	c	d	e
a	1.000000	0.949504	0.999775	0.995800	0.995800
b	0.949504	1.000000	0.949594	0.946039	0.946039
c	0.999775	0.949594	1.000000	0.995001	0.995001
d	0.995800	0.946039	0.995001	1.000000	1.000000
e	0.995800	0.946039	0.995001	1.000000	1.000000

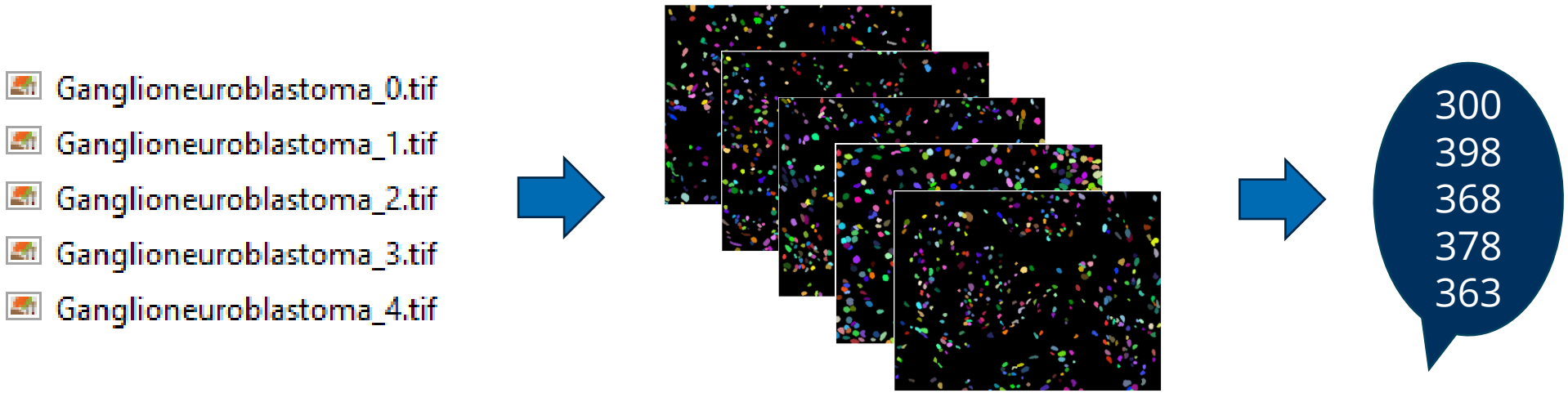
Unit-test pass-rate (n=10):



https://www.biorxiv.org/content/10.1101/2024.04.19.590278v1
https://github.com/haesleinhuepf/human-eval-bia

Benchmarking LLMs for Bio-image Analysis

Use case: Count segmented objects in a folder of segmentation results.



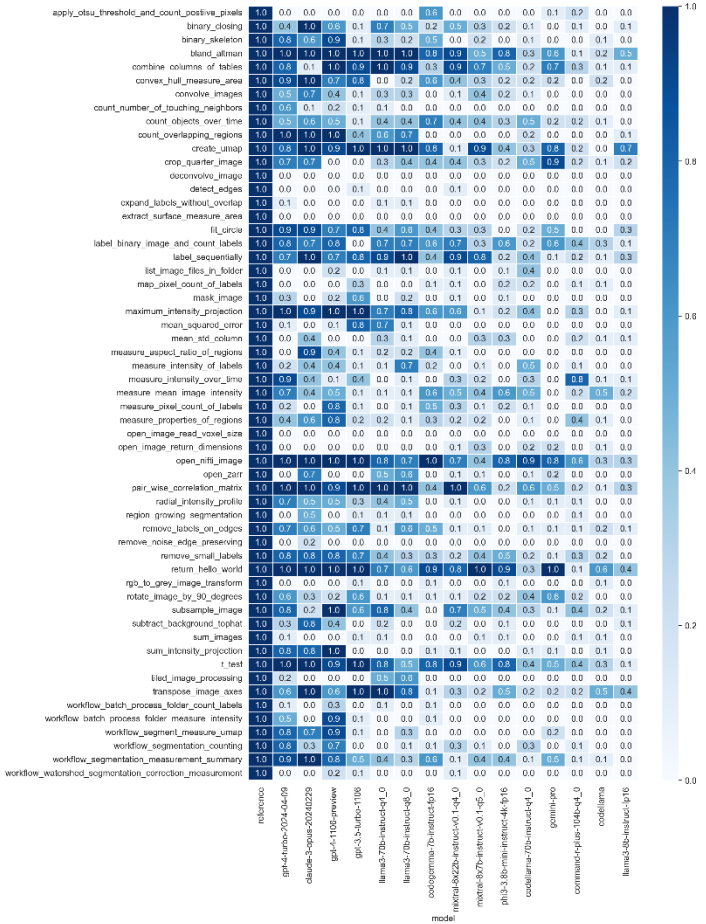
Unit-test pass-rate (n=10):

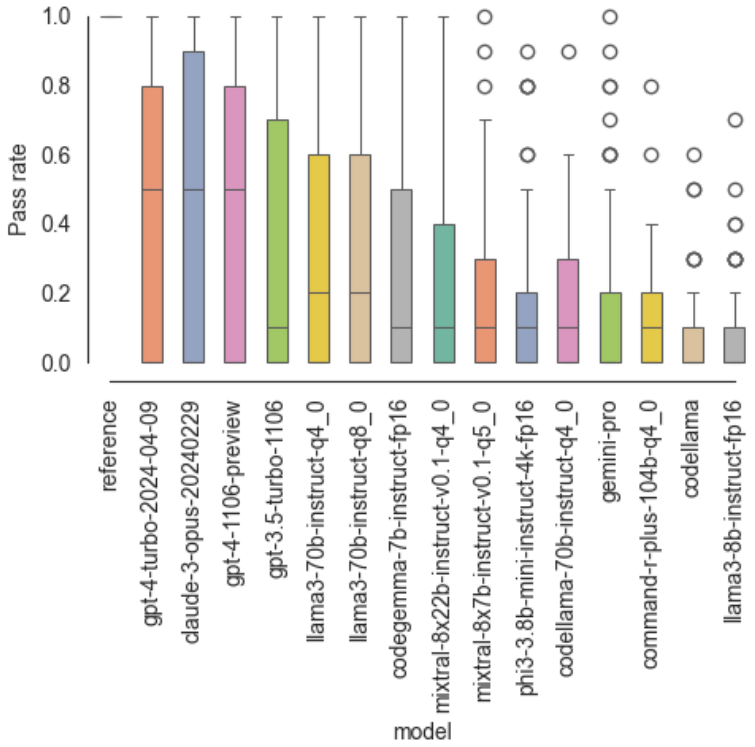
	reference	gpt-4-turbo-2024-04-09	Claude-3-opus-20240229	gpt-4-1106-preview	gpt-3.5-turbo-1106	gemini-pro	codellama
workflow_batch_process_folder_count_labels	1.0	0.1	0.0	0.3	0.0	0.0	0.0

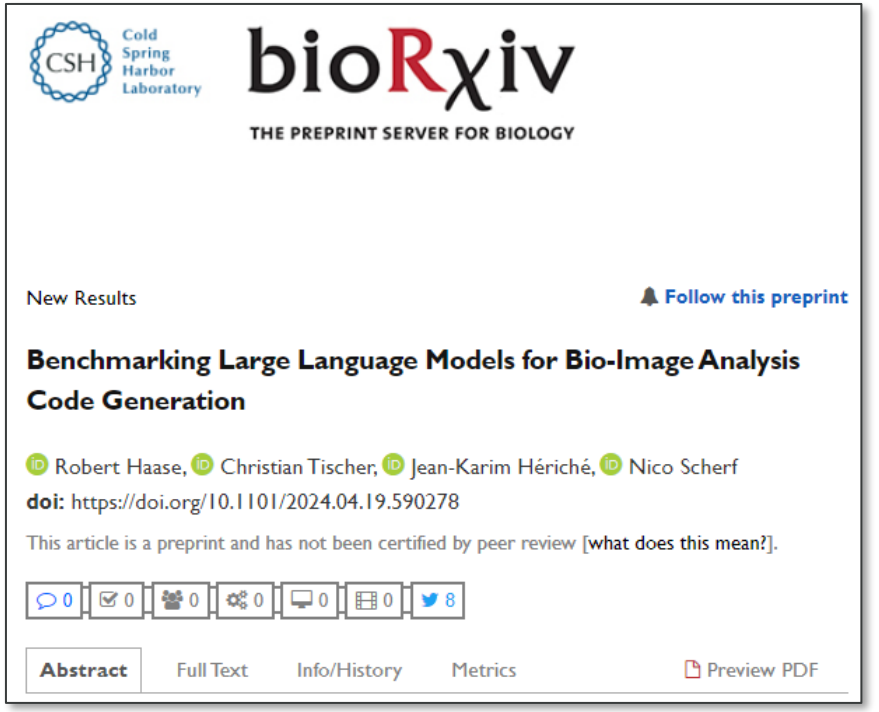
https://www.biorxiv.org/content/10.1101/2024.04.19.590278v1
https://github.com/haesleinhuepf/human-eval-bia
Data Source: https://www.ebi.ac.uk/bioimage-archive/galleries/S-BIAD634-ai.html

Benchmarking LLMs for Bio-image Analysis

Summary: 57 use-cases (yet), 15 LLMs (yet), n=10







Coding assistance: BiA-Bob

Keep your feed on the ground with *Bob*. Bob can do crazy things, but you are responsible for what it does with your data.

Do not enter personal / private information. What you enter will be sent to the server of an american company.

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CENTER FOR SCALABLE DATA ANALYTICS AND ARTIFICIAL INTELLIGENCE

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SACHSEN

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.

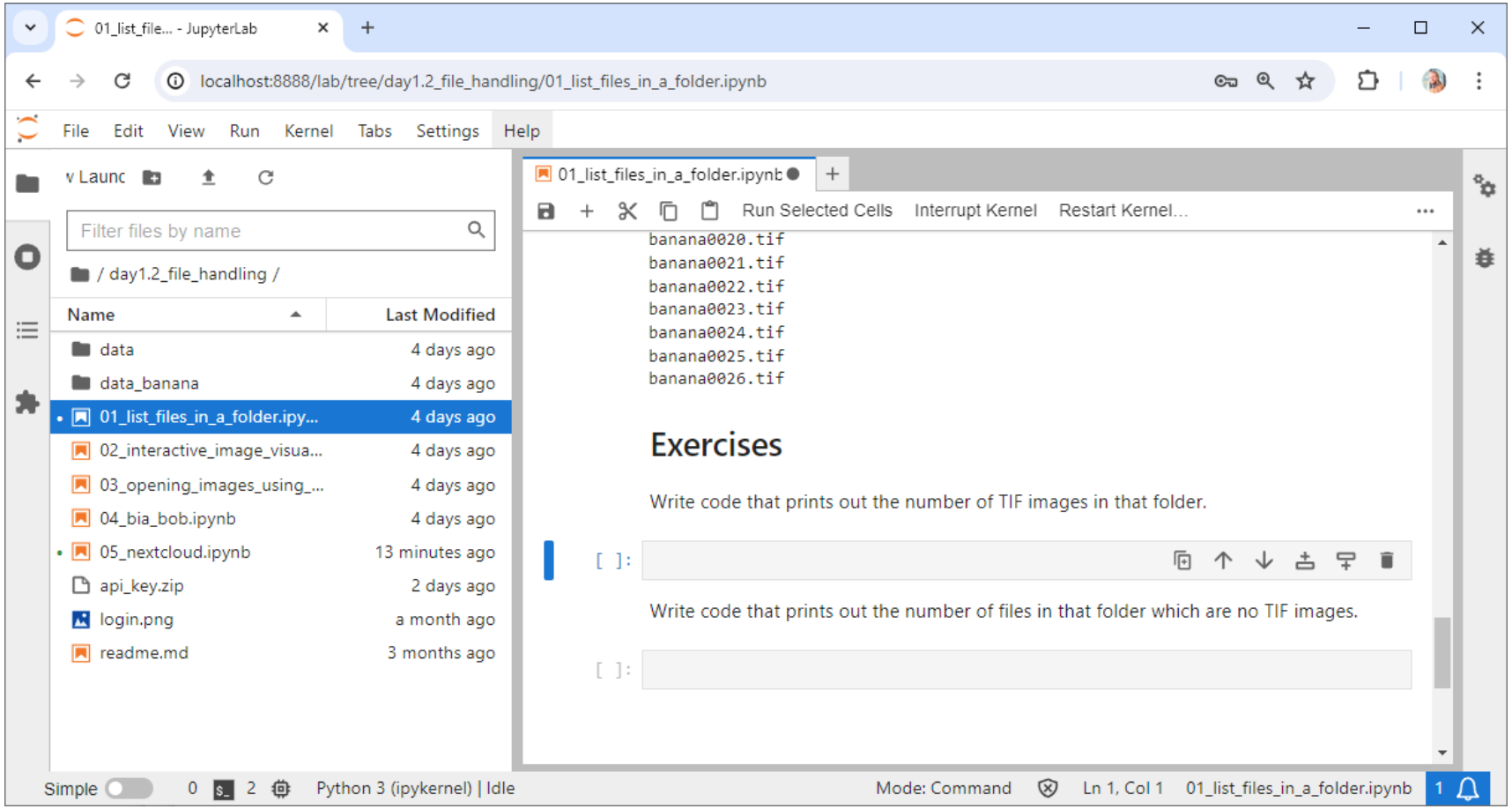
Exercises

Robert Haase

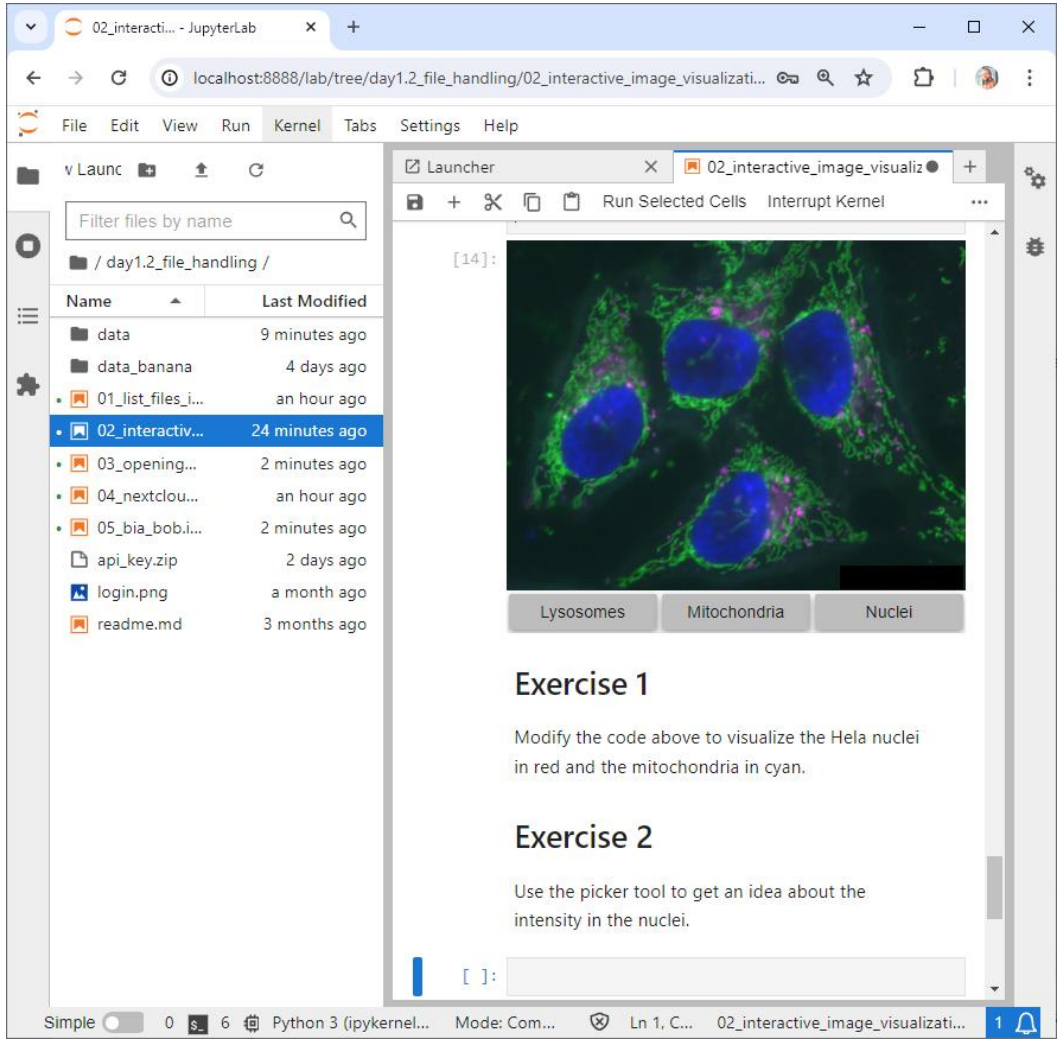
https://doi.org/10.5281/zenodo.10841765

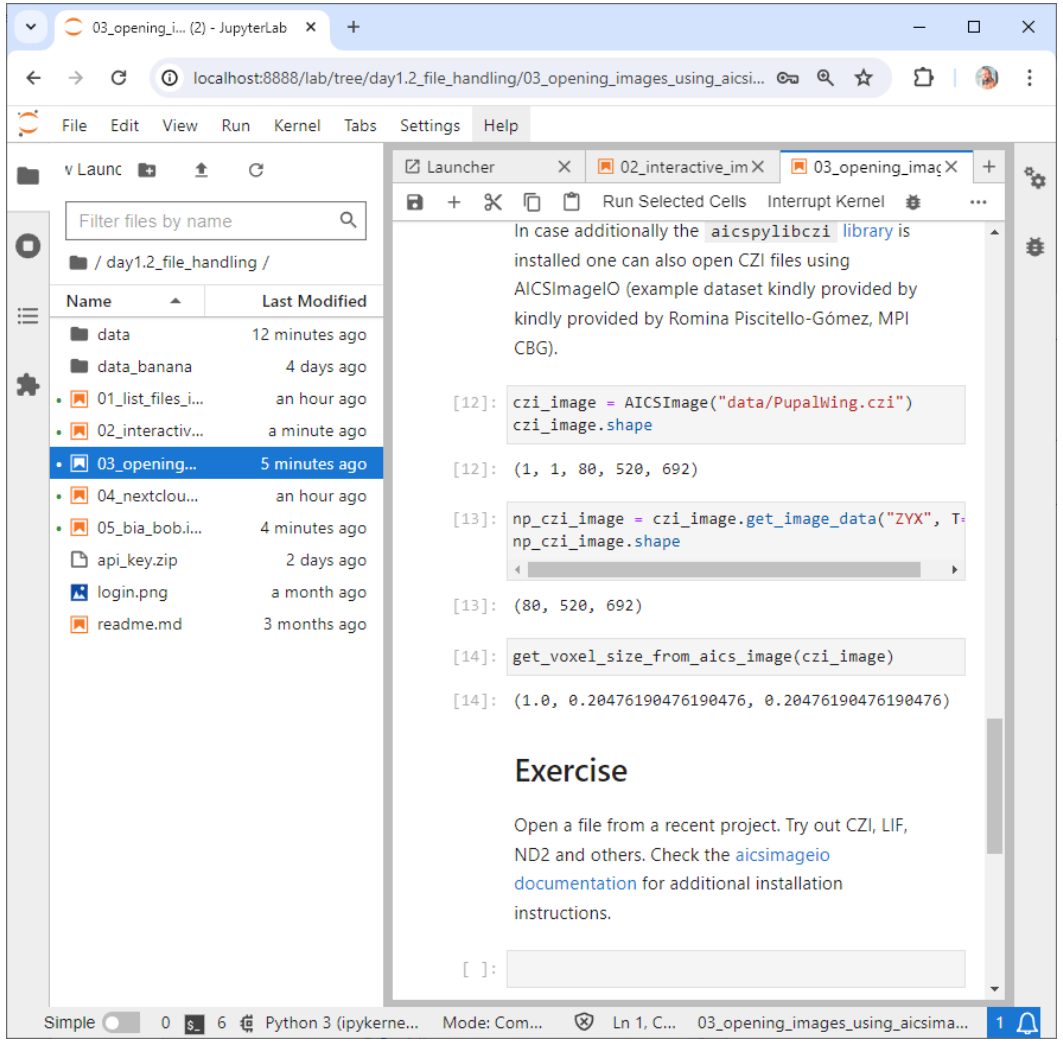
Exercise: File lists and folder

Apply your knowledge about Python lists to list of files.



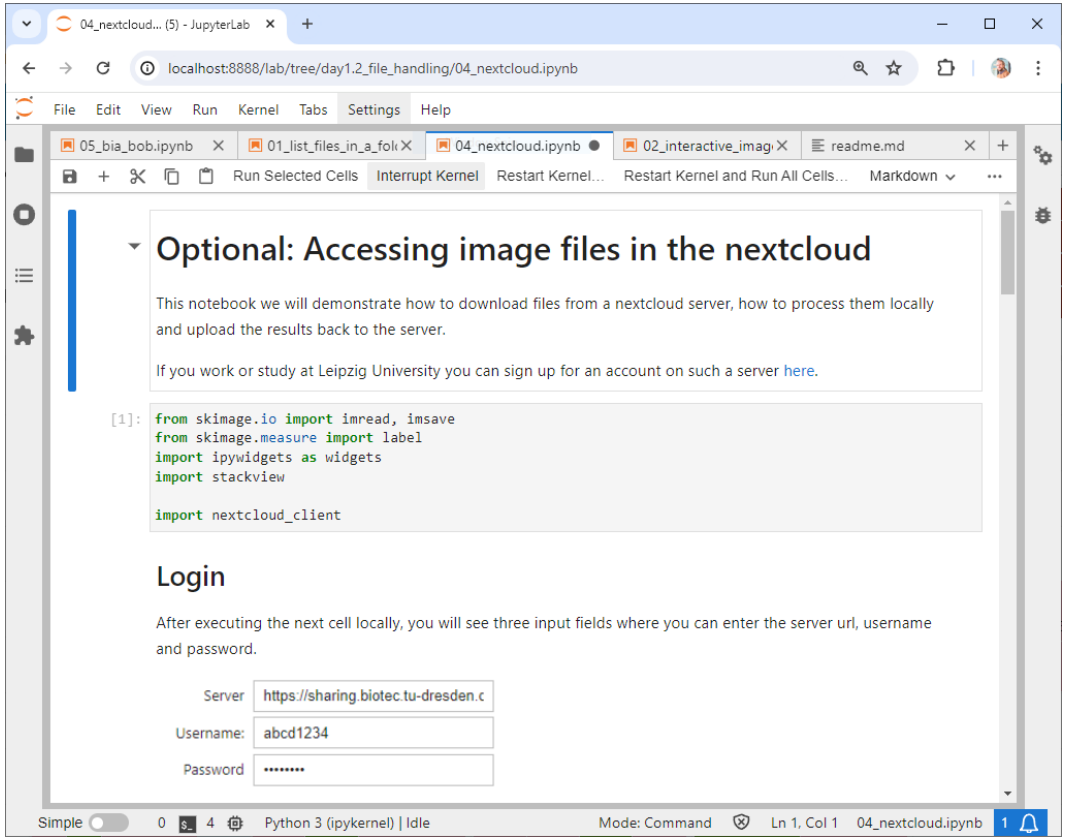
Exercise: Loading and visualizing image files

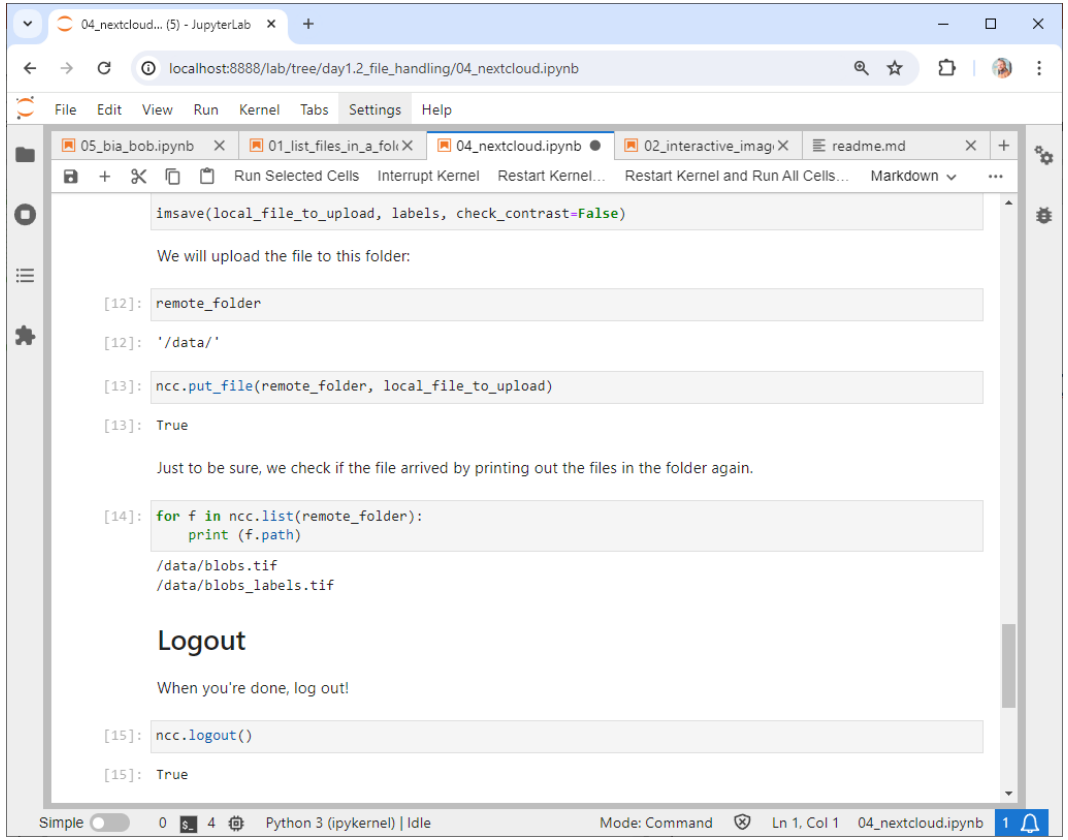




Optional Exercise: Nextcloud

Download a file from your nextcloud/owncloud, process it and upload it again.





Optional exercise: BiA-Bob