MIC-MAQ

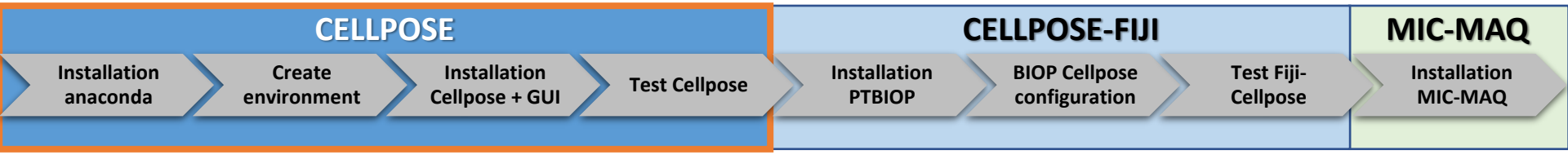
Microscopy Images of Cells-Multi Analysis and Quantifications

Installation

For MIC-MAQ

- Installation of Cellpose
  - Configuration of Fiji to run Cellpose from Fiji
- Installation of the plugin MIC-MAQ

Installation of Cellpose



• Download a python distribution

You have the possibility to download Miniconda (small version) or Anaconda (full version) :

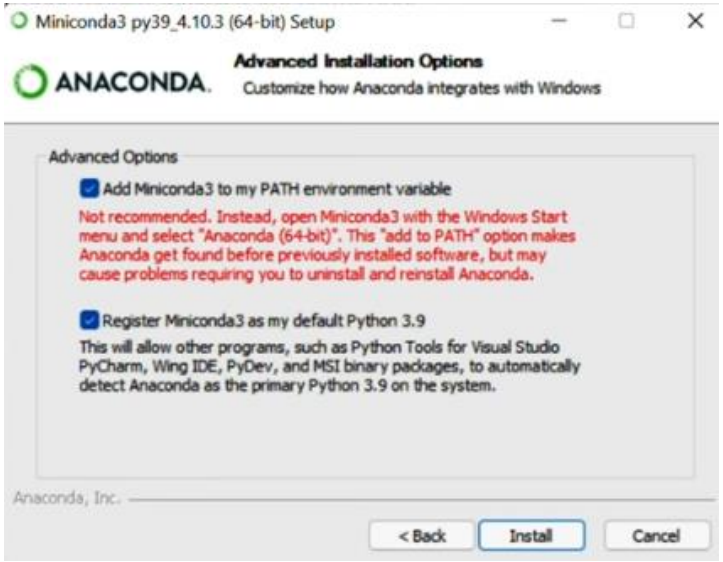
- Miniconda : https://docs.conda.io/en/main/miniconda.html
- Anaconda : https://www.anaconda.com/download

• • Installation of Miniconda/Anaconda

Follow installer instructions

Install the software in **C:\Users\YourSessionName\anaconda3**

During the installation, please tick the box **"add anaconda/miniconda to my PATH environment variable »** .

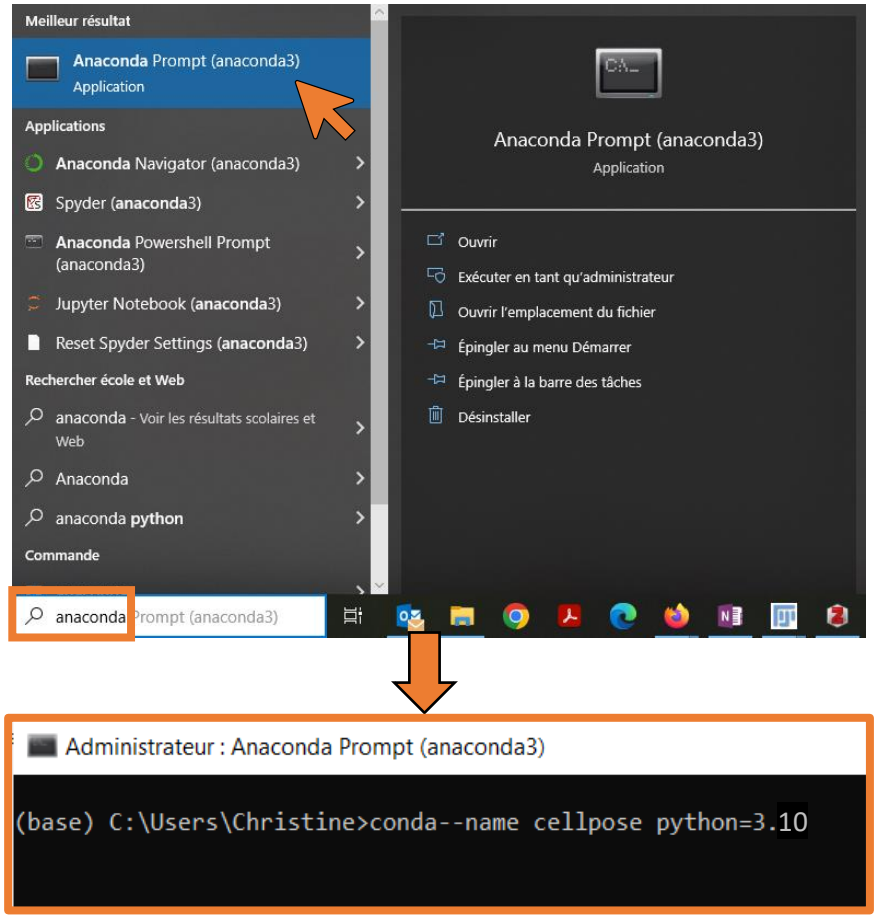


• Open the Anaconda Prompt application

In the Windows search toolbar, enter **anaconda** and open the **« Anaconda Prompt application »**.

Use the **MacOs terminal** directly.

Use the **terminal** directly.



• • Create a new anaconda environment

In the Anaconda Prompt window, write this command line and validate by pressing Enter key:

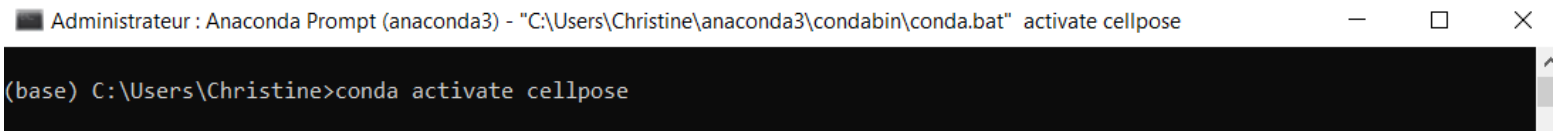
conda create --name cellpose python=3.10

!! Lot of packages will be downloaded and you need to press the « y » key to launch the installation !!

If you are behind a proxy, configure it using following instructions for your system:
https://docs.anaconda.com/free/anaconda/configurations/proxy/

• Activate the cellpose anaconda environment

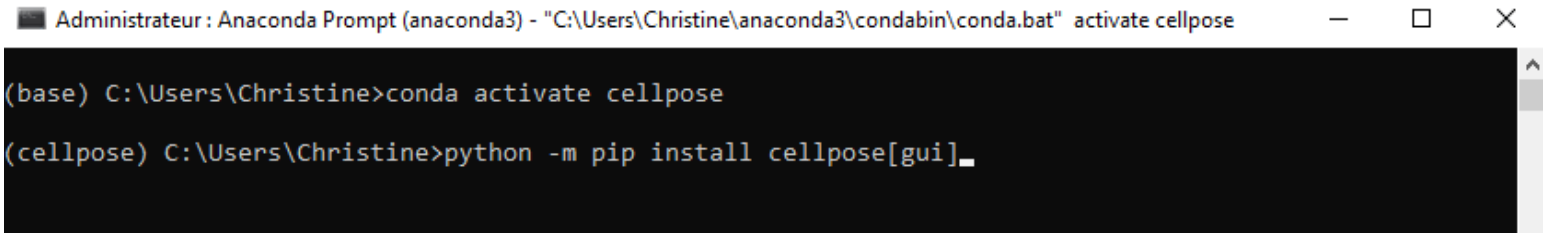
In the Anaconda Prompt window, write this command line : *conda activate cellpose*



• • Cellpose installation in anaconda (CPU installation)

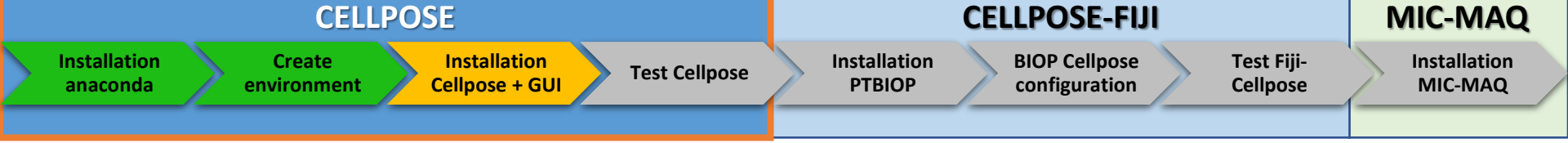
In the Anaconda prompt window, write this command line : *python -m pip install cellpose[gui]*

write this command line : *python -m pip install 'cellpose[gui]'*



This will install the last version of Cellpose (currently 4.0) which is based on CellposeSAM and is incompatible with previous versions.

To install a previous version use the command: *python -m pip install cellpose[gui]==3.1.1.2*



• Optional: GPU activation for Cellpose

We recommend to use the GPU installation version to speed up the segmentation process.
At least 8GB of RAM is required to run the Graphical User Interface (GUI) of Cellpose.

In the Anaconda prompt window, write this first command line :

conda uninstall pytorch

In the Anaconda prompt window, write this second command line :

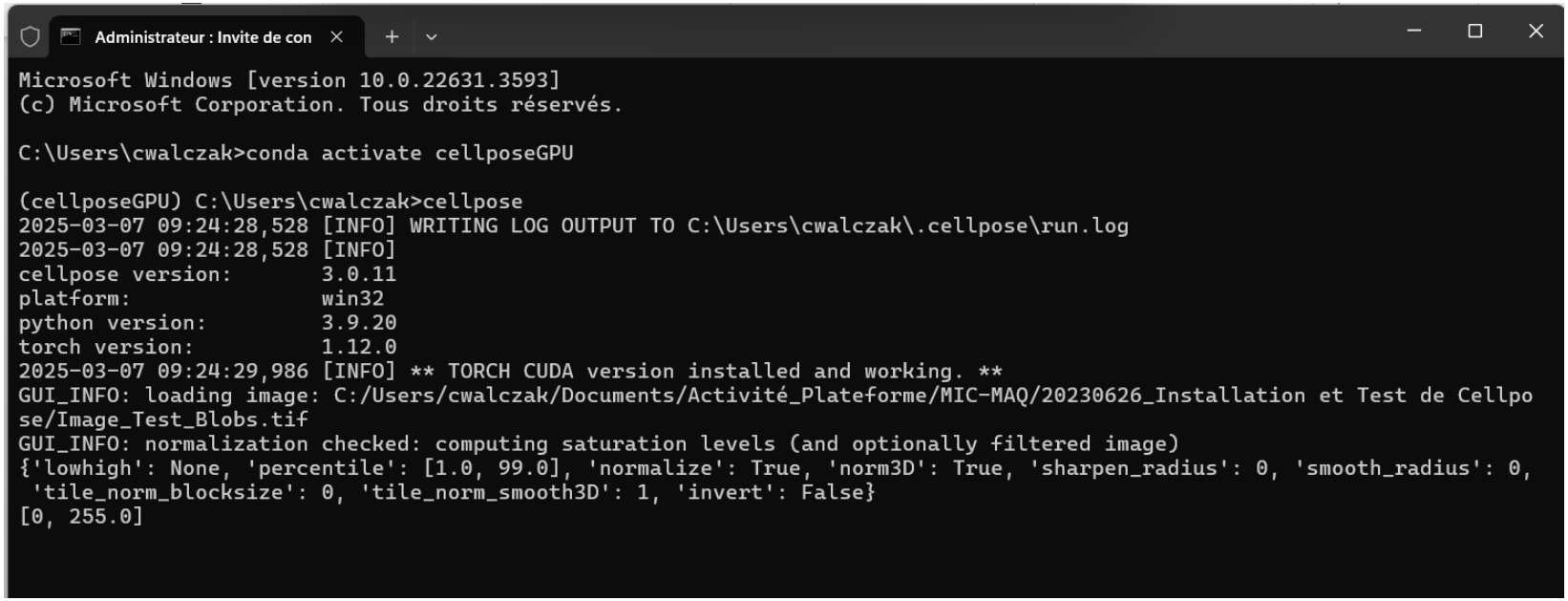
conda install pytorch pytorch-cuda=11.6 -c pytorch -c nvidia

Complete installation instructions at these websites:

- https://github.com/MouseLand/cellpose#gpu-version-cuda-on-windows-or-linux
- https://github.com/BIOP/ijl-utilities-wrappers#conda-cellpose-gpu

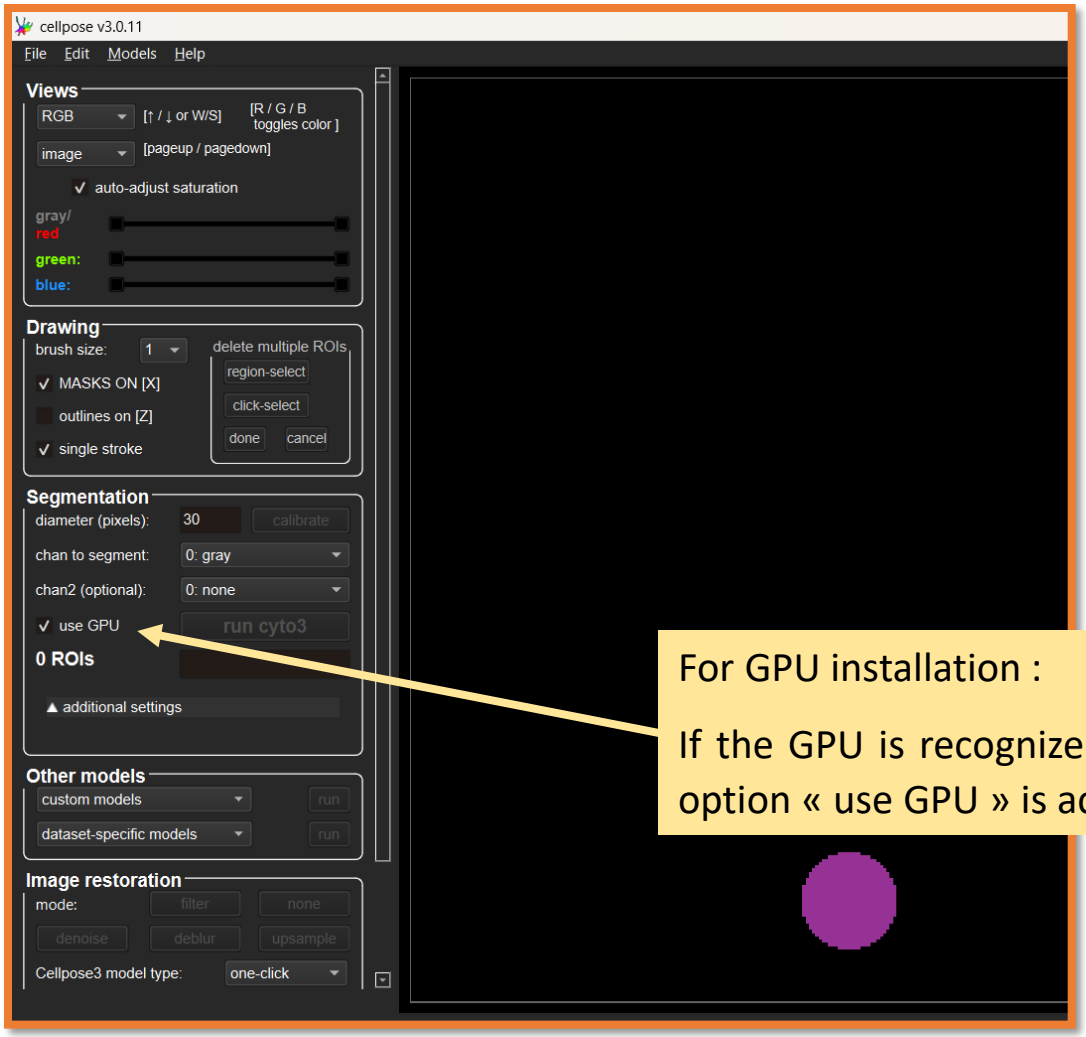
• Start Cellpose in Anaconda

In the Anaconda Prompt window, write this command line : *python –m cellpose*



• Start Cellpose in Anaconda

In the Anaconda Prompt window, write this command line : *python –m cellpose*



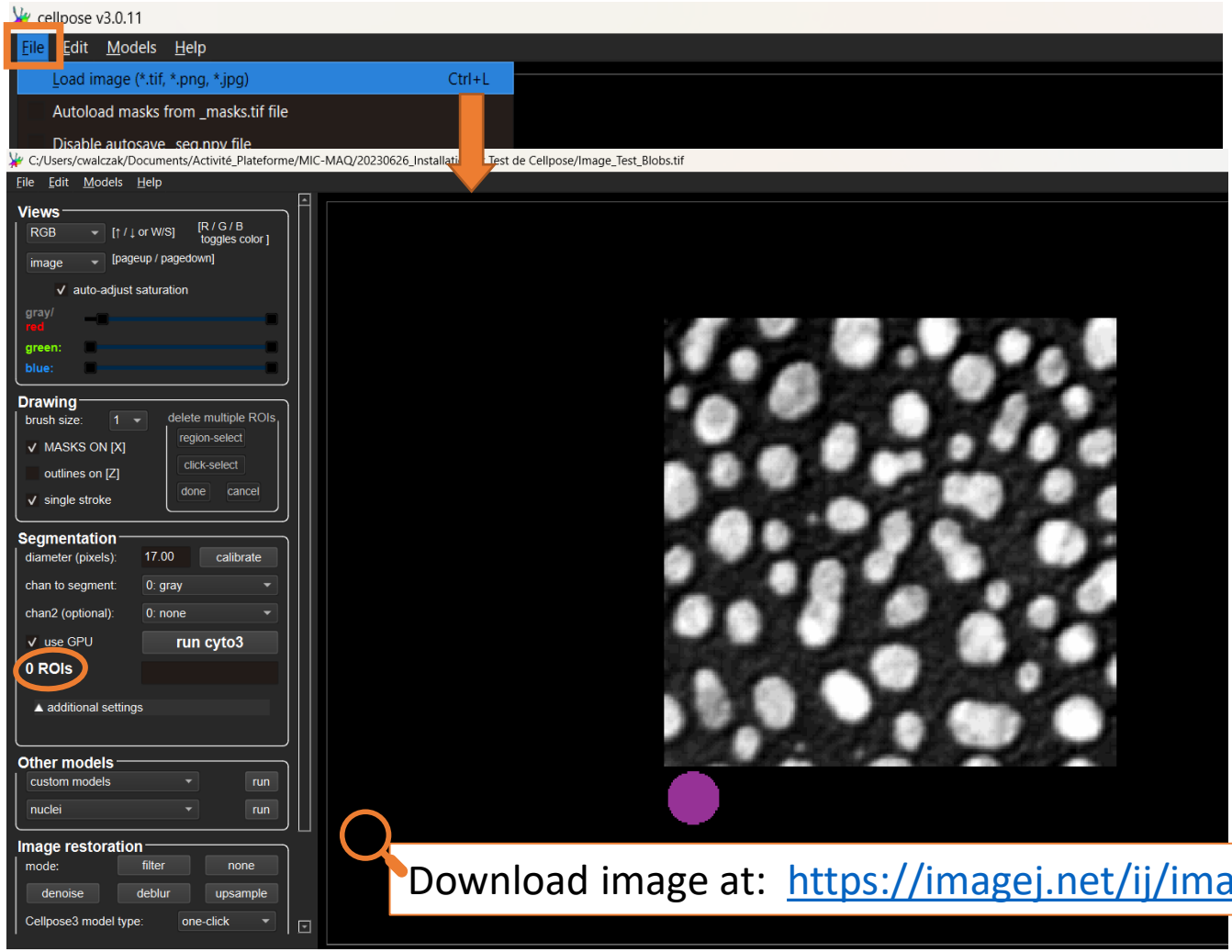
This is the GUI for Cellpose 3

For GPU installation :

If the GPU is recognized by Cellpose the option « use GPU » is activated.

• Load image in Cellpose GUI

In the Cellpose GUI, load the image called « *Image_Test_Blobs* »: **File > Load image**



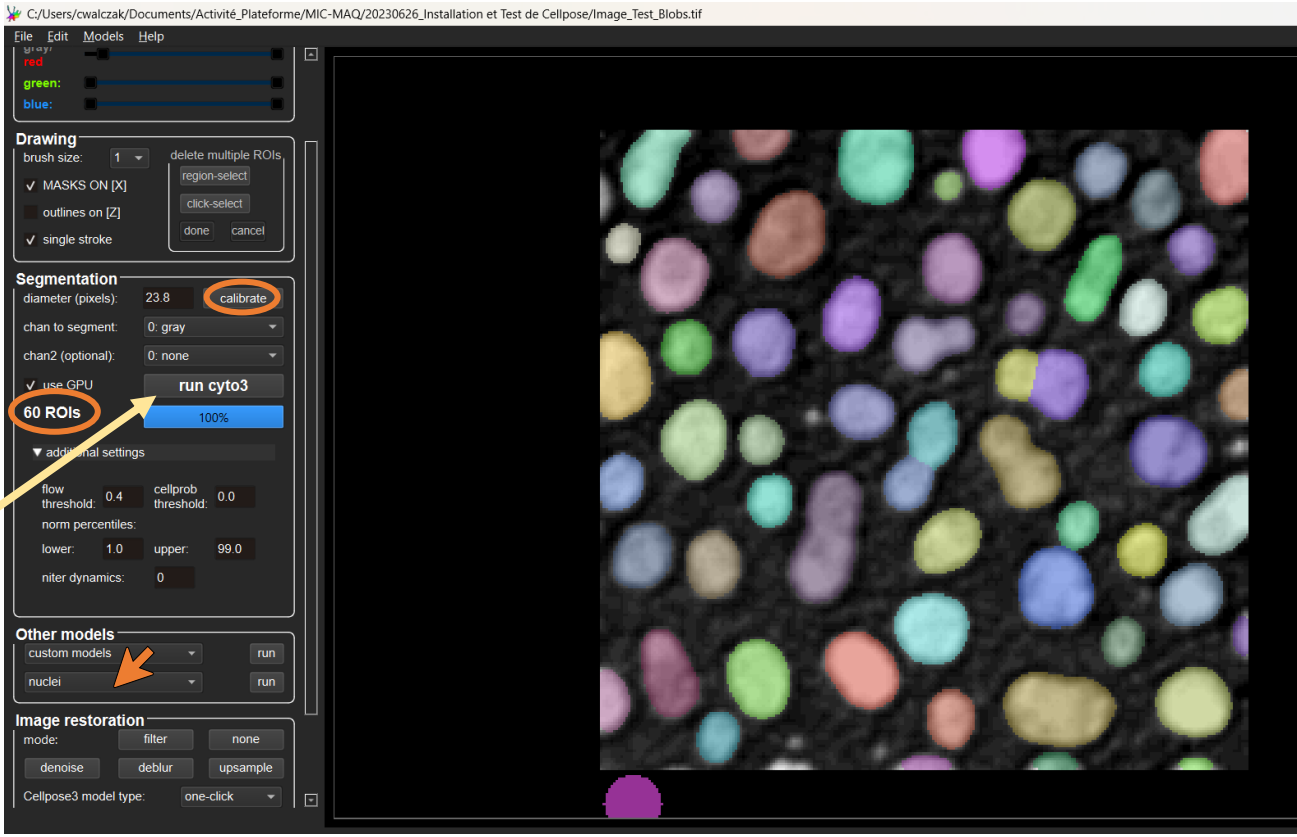
Download image at: https://imagej.net/ij/images/blobs.gif

• Run Cellpose

In the Cellpose GUI:

- Click on « **calibrate** » to define automatically the **object's diameter** (the value should be at 23.8 pixels)

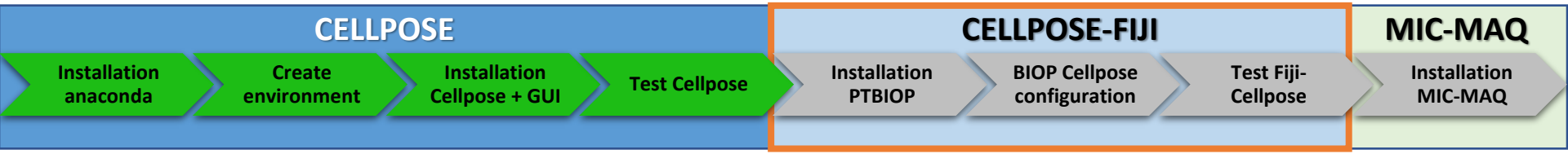
- Click run on **« cyto 3 » cellpose model zoo**



As results, the number of segmentation regions will appear on the GUI (60 ROIs) and a mask of segmented objects will be created with random color rendering.

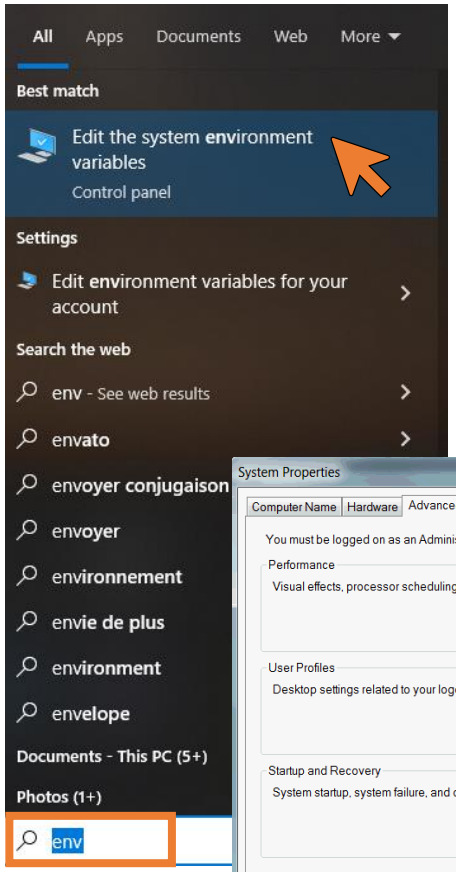
Installation of Cellpose

Configuration to run from Fiji

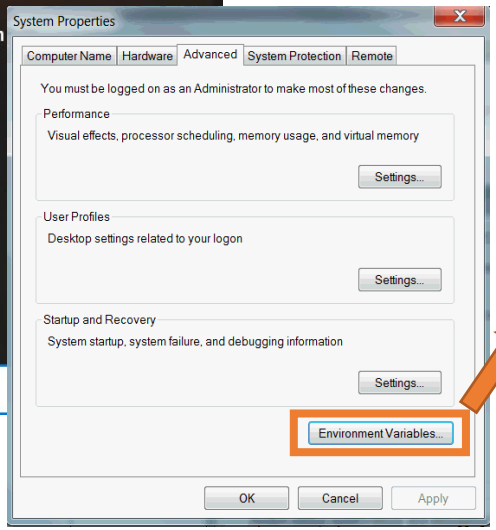


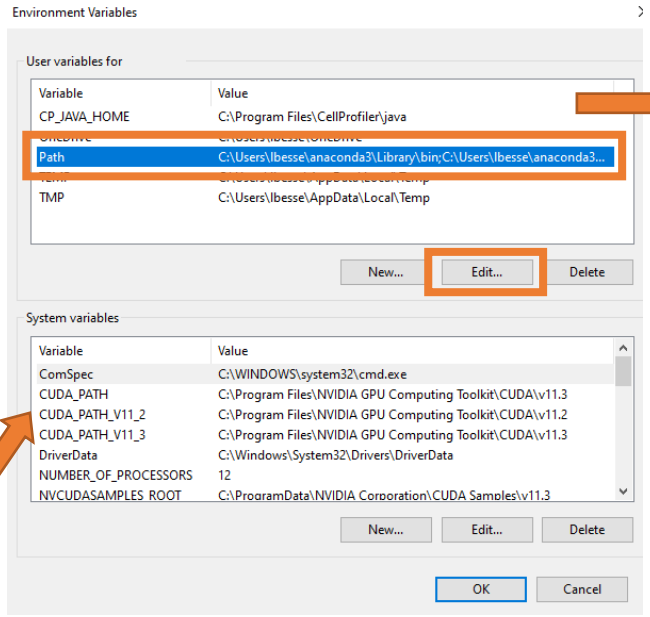
• Modify the Environment Variables

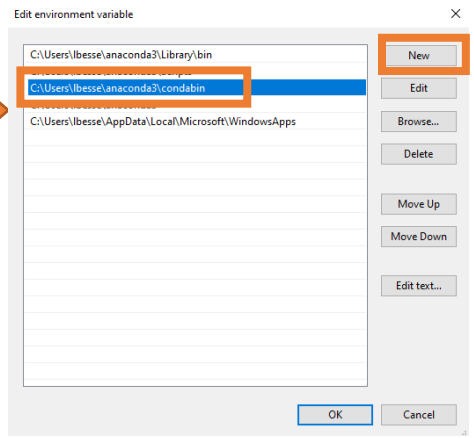
In the Windows search toolbar, enter « **env** » and open **« edit the system environment variables »** pannel.



- Click on « Environment Variables » to open a new window.
- Select Path variables and click on Edit
- Select New and paste the folder directory of your condabin for anaconda
  As example: *C:\Users\YourSessionName\anaconda3\condabin*
- Validate by clicking on OK





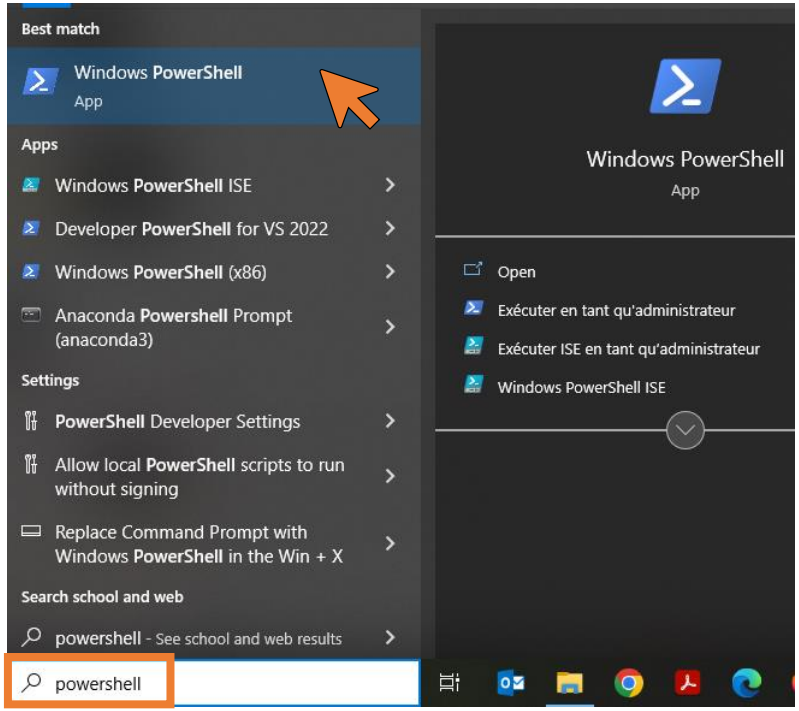


- **Initialize Conda for Windows**

In the Windows search toolbar, enter **powershell** and open the **« Windows Powershell »** application.

In the **Powershell** window, write this command line :

conda init



Check the initialization:

In the Windows search toolbar, enter **[cmd]** and open the **« Command Prompt »** application.

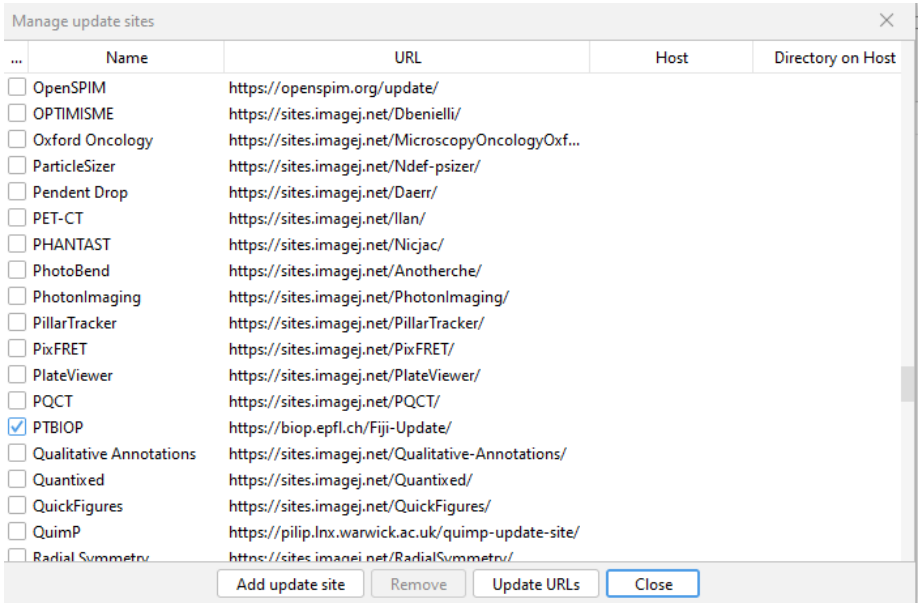
In the **Command Prompt** window, write this command line : *conda env list*

As results, the list of the conda environment is displayed.

• Installation of Wrapper BIOP plugin under Fiji

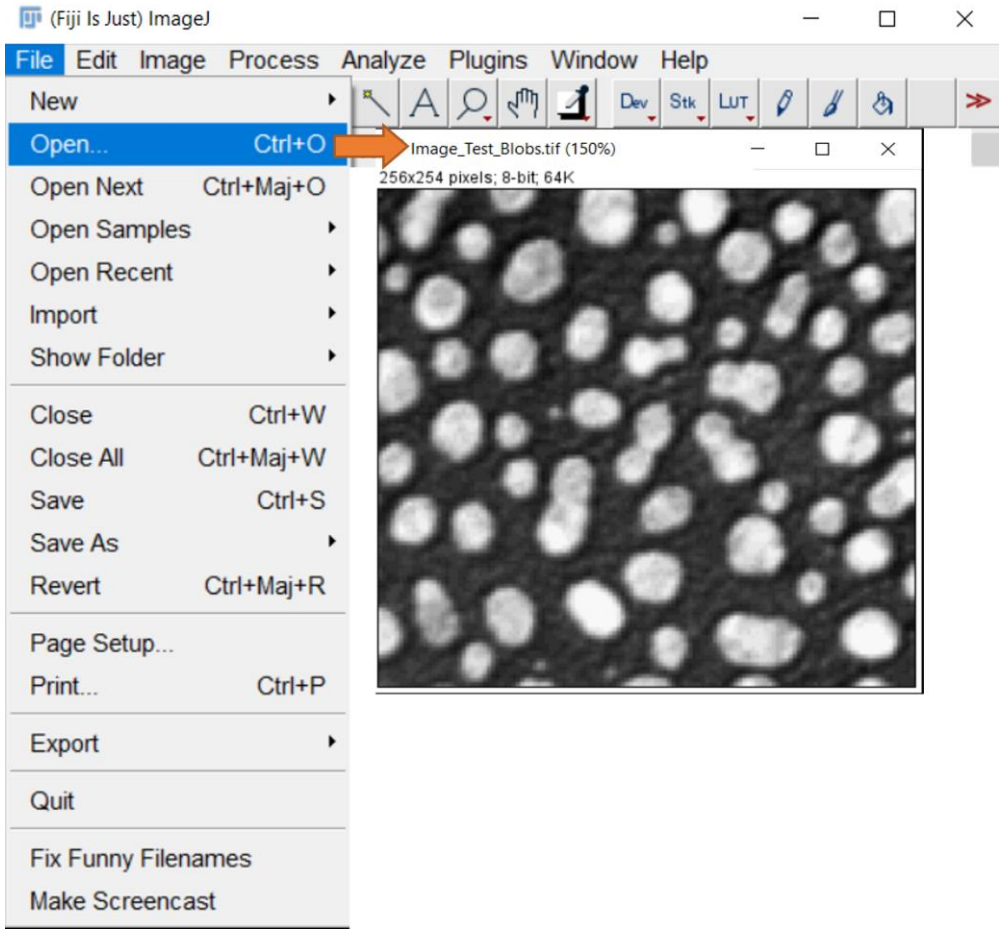
Use the built-in update manager : **Help > Update...**

- Click on the button “Manage update sites”
- Install PTBIOP
  - ❑ Scroll down the list and tick the checkbox for update site “PTBIOP”
  - ❑ Click the “Close” button
- Click the “Apply changes” button
- Restart Fiji



• Load image in Fiji

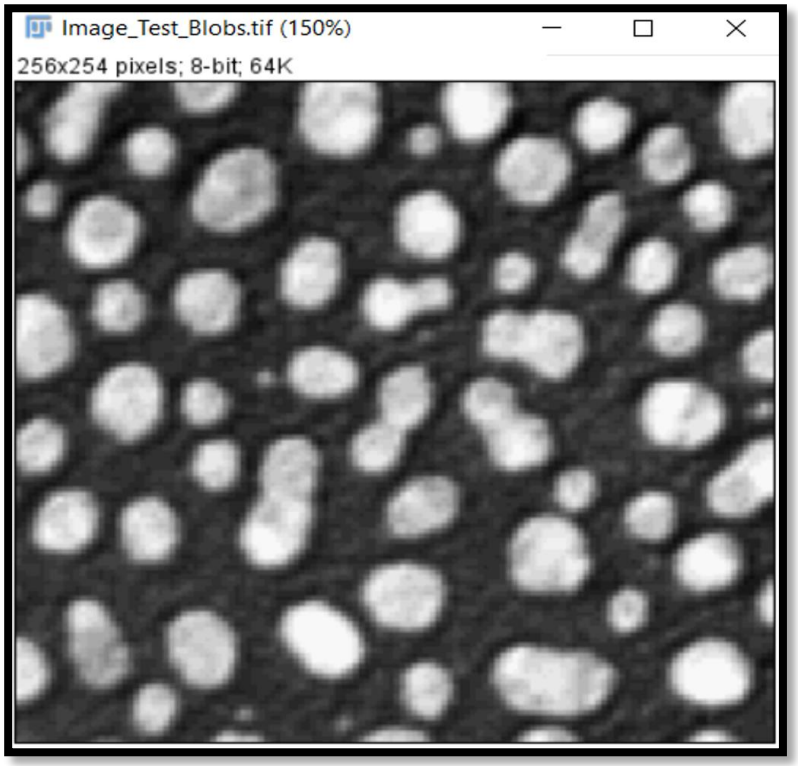
In Fiji, load the image called « *Image_Test_Blobs* »: **File > Open**

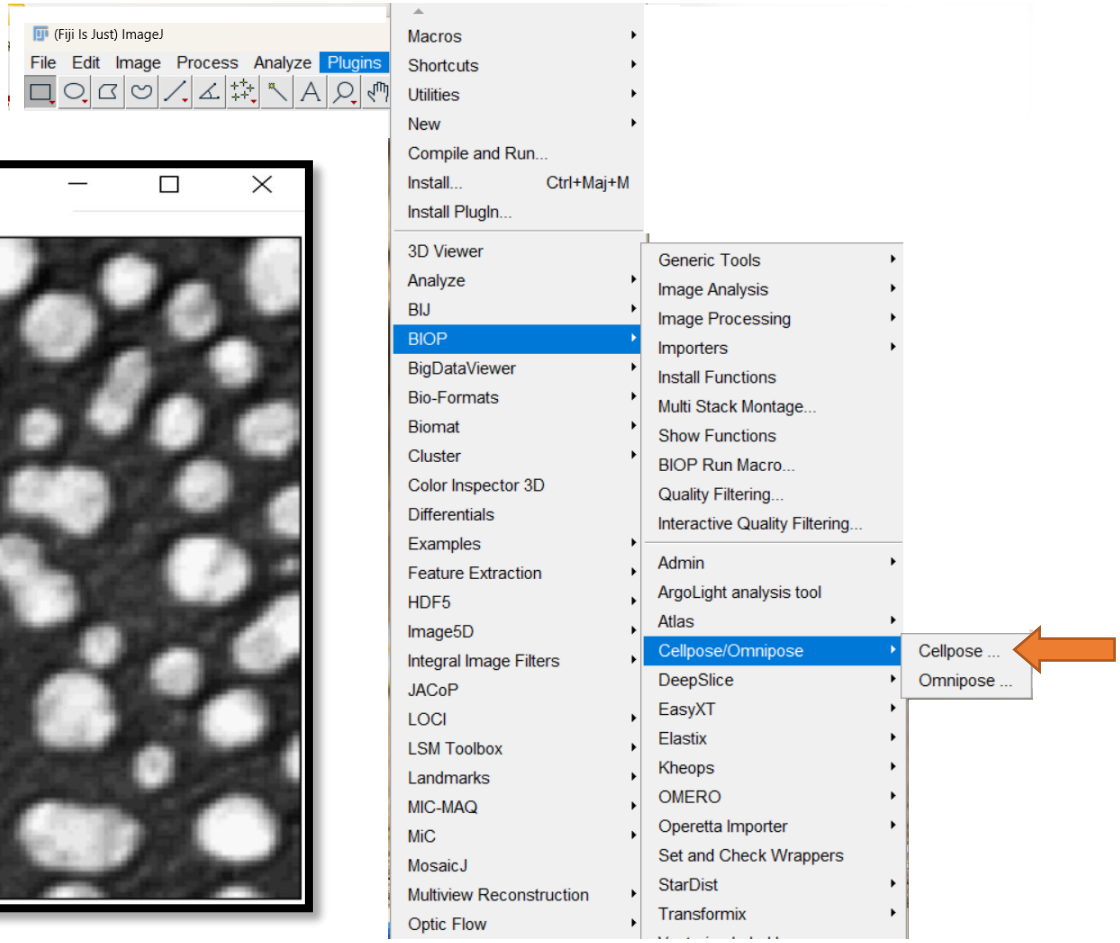


Download image at: https://imagej.net/ij/images/blobs.gif

• Run Cellpose under Fiji

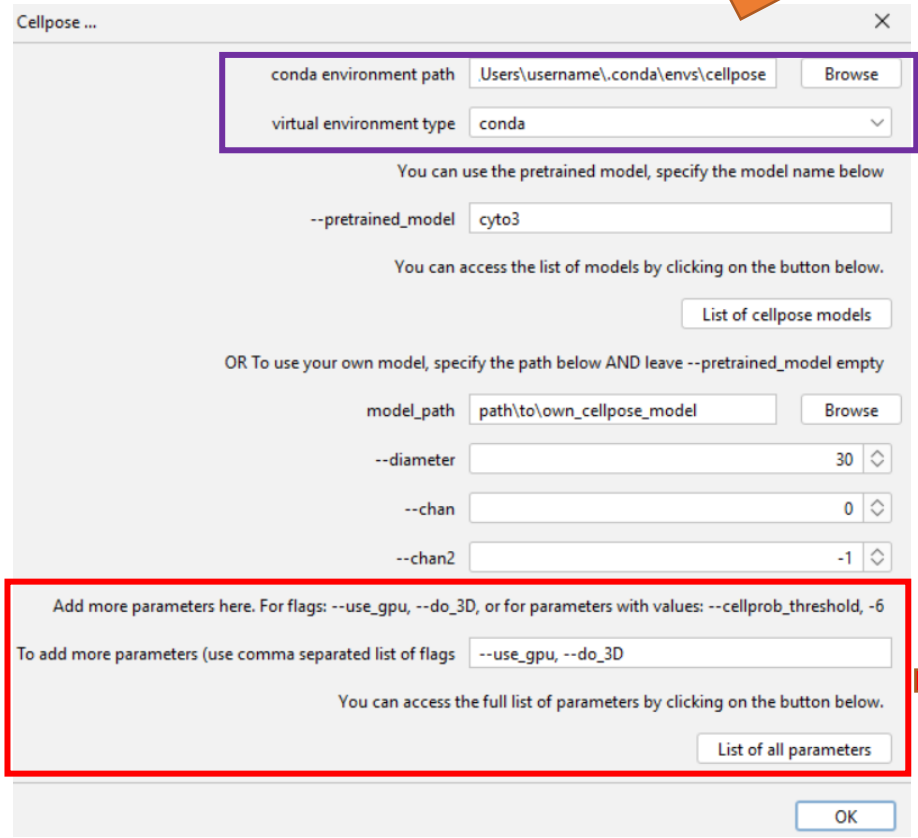
In Fiji, run cellpose with the wrapper BIOP on the test image: **Plugins > BIOP > Cellpose/Omnipose > Cellpose…**





● Run Cellpose under Fiji

Global configuration of Cellpose



❑ Select the path to your working Cellpose virtual environment:

example: *C:\Users\UsersFolder\anaconda3\envs\cellpose*

❑ Select Environment Type : **conda**

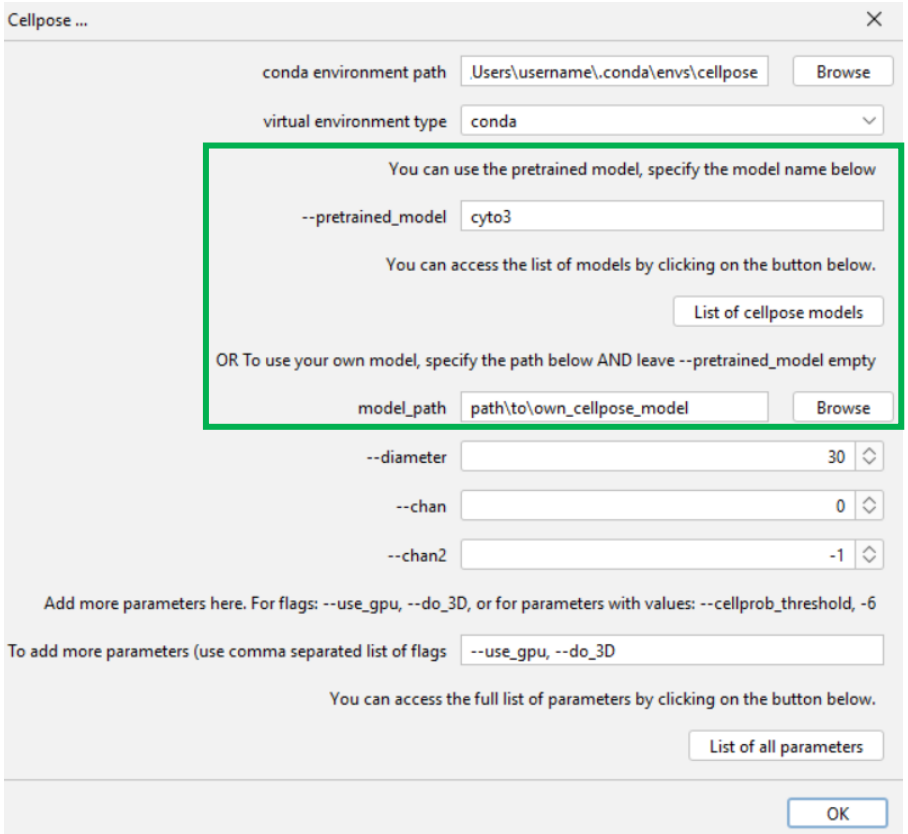
❑ Additionnal parameters if needed.
The most common one is :

« --use_gpu » to compute on the GPU

« --do_3D » for 3D computation

- **Run Cellpose under Fiji**

Parameters for the segmentation



Choose model between :

❑ **Pretrained**

Fill first text field (available model are listed here [1])

For this exemple:

Enter *cyto3* in the first text field

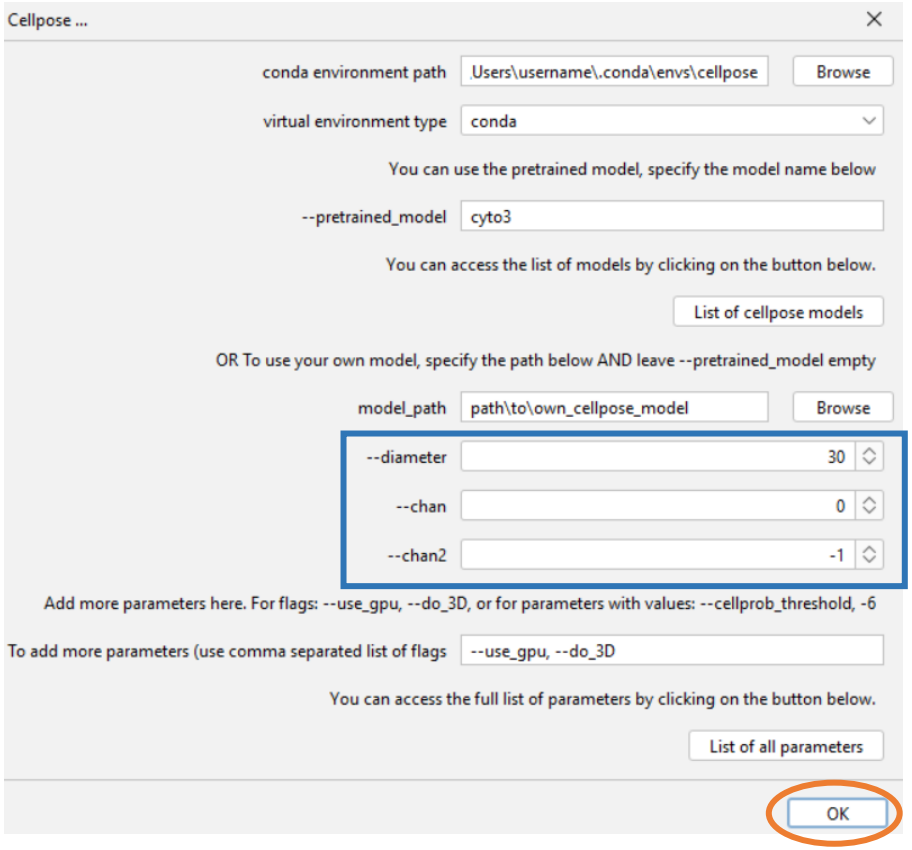
❑ **own model**

- Let first text field empty
- And select the path to the model

[1] : Models — cellpose 3.1.1.1-7-g549126d documentation

- **Run Cellpose under Fiji**

Parameters for the segmentation

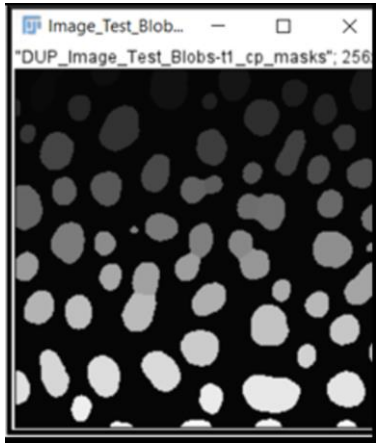


Cellpose parameters :

- ❑ Aproximative diameter of object
- ❑ **Chan :** Define the segmentation channel (default value 0 when you have only one channel)
- ❑ **Chan2 :** when you have additional nuclei channel, you need to define the order of the channel in the composite

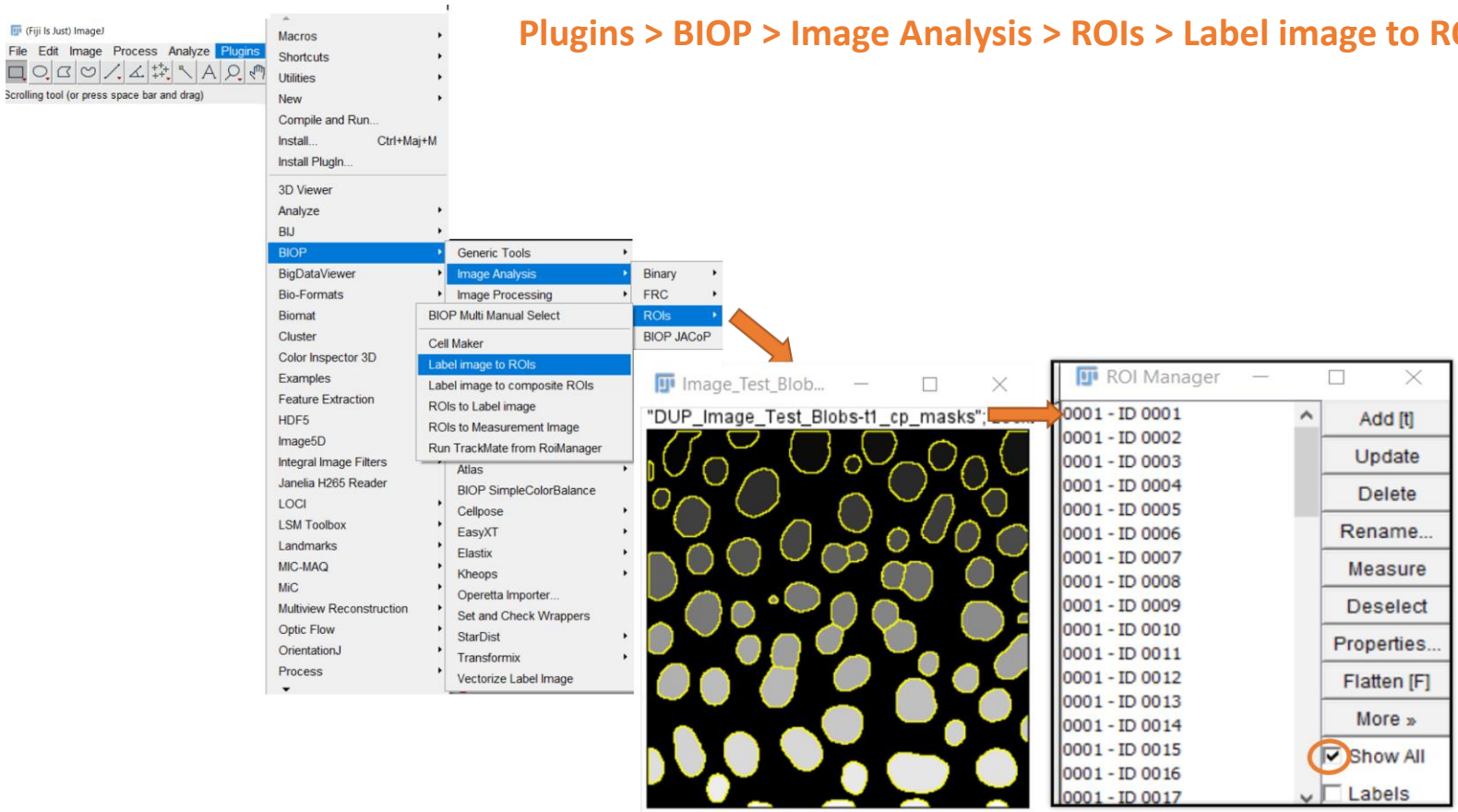
For this exemple:
Diameter: 30
Chan : 0
Chan2 : -1

Resulting image

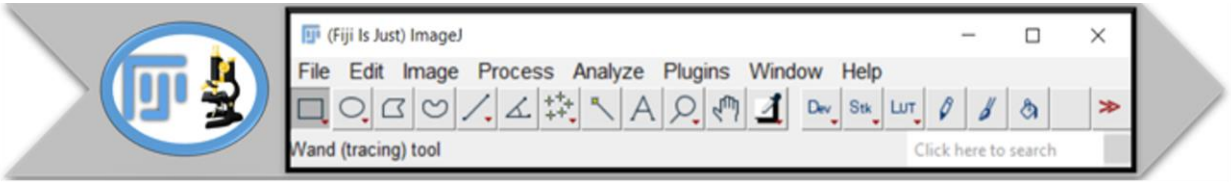


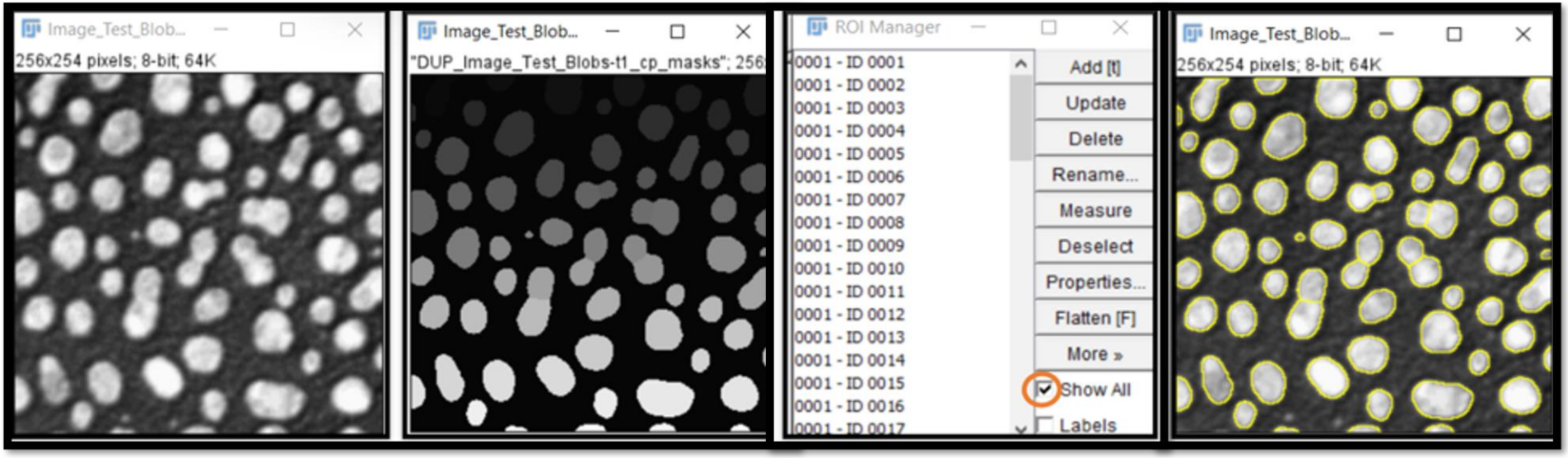
• Create ROIs from the cellpose mask

In Fiji, select the cellpose mask and create ROIs from the cellpose mask:



Summary of the Cellpose test under Fiji





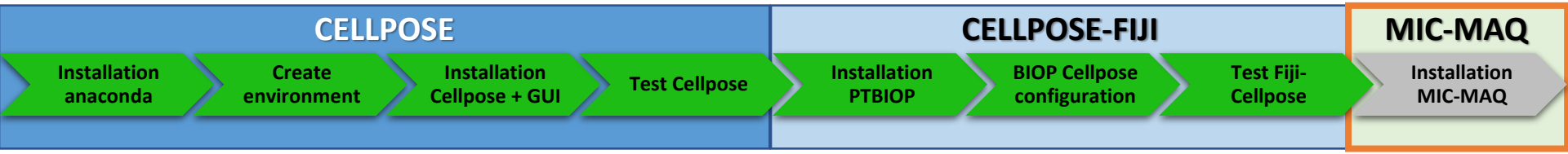
Raw image

Cellpose segmentation mask

List of ROIs obtained with Cellpose segmentation

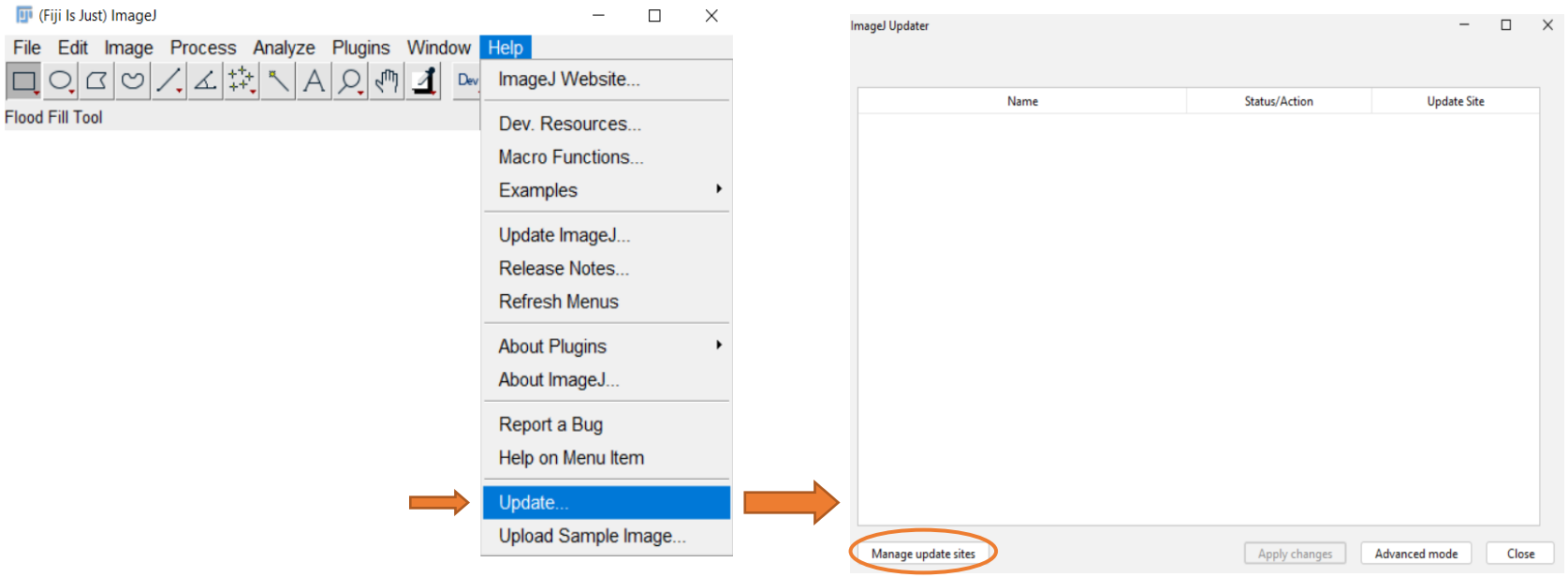
Raw image + ROIs obtained with Cellpose segmentation

Installation of MIC-MAQ



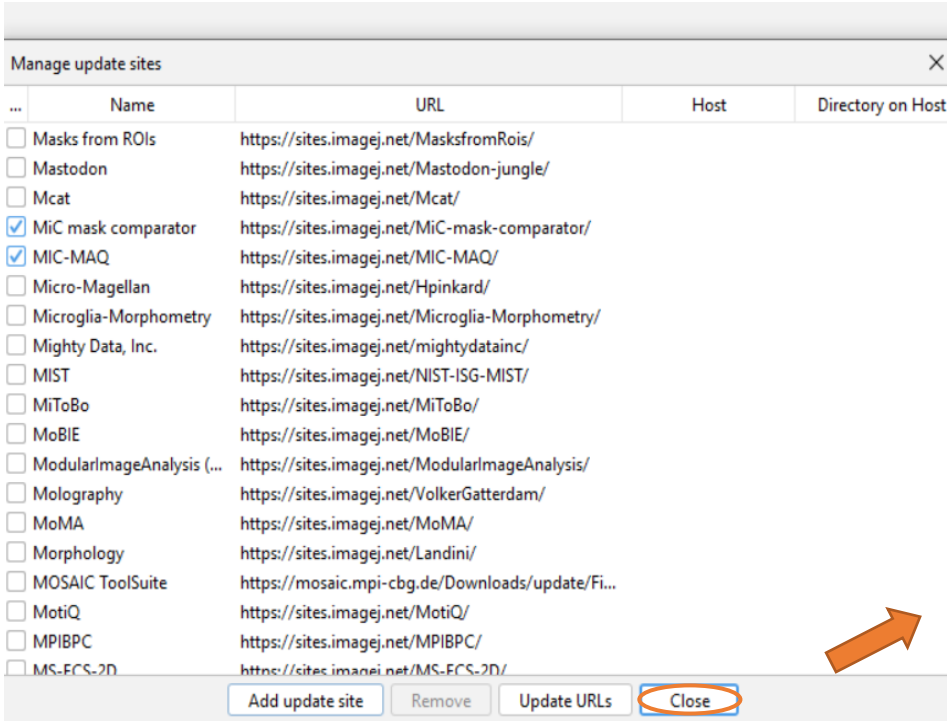
● Start the Fiji updater

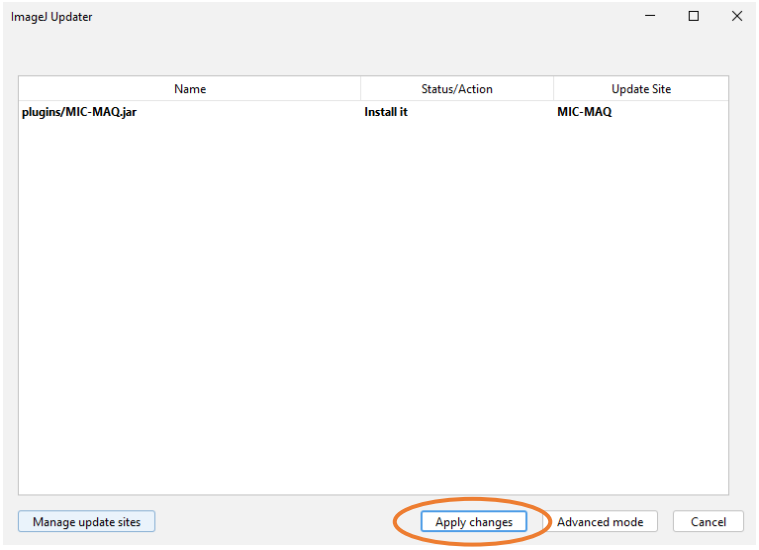
- Use the built-in update manager : **Help > Update...**
- Click on the button “Manage update sites”



- **Install MIC-MAQ**

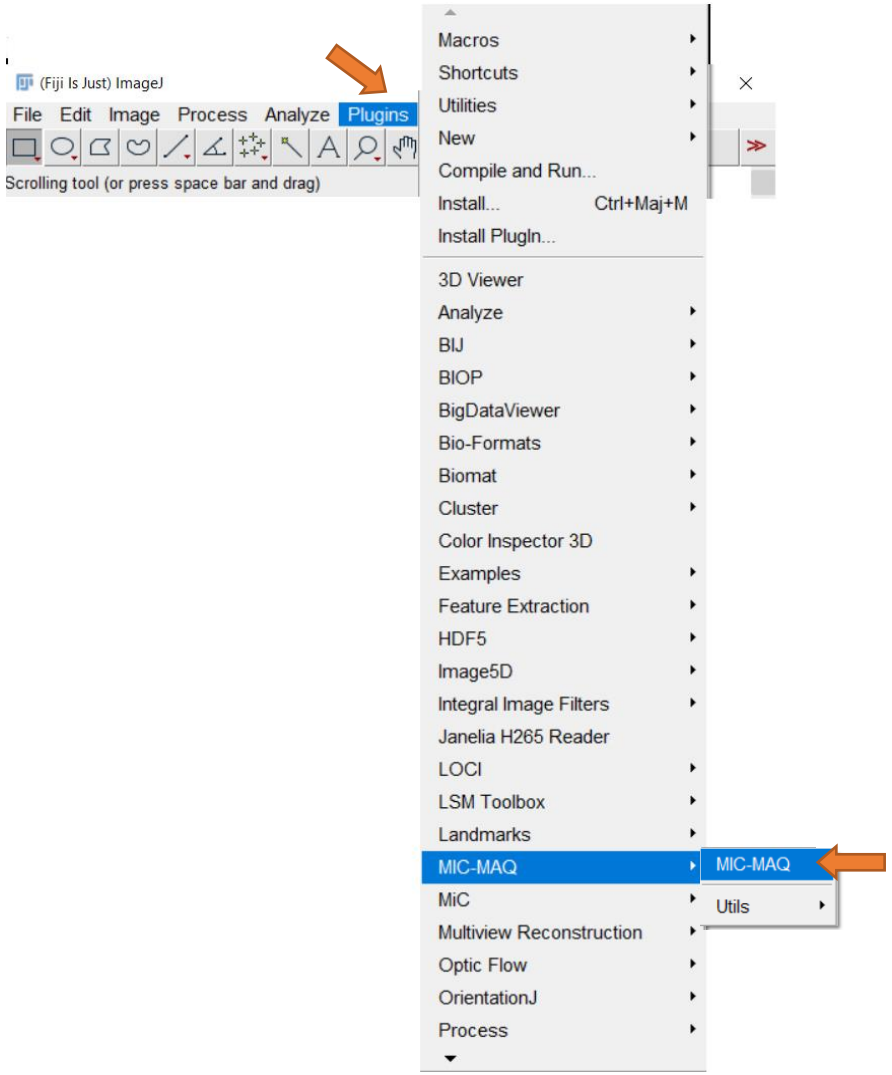
  - Scroll down the list and tick the checkbox for update sites “MIC-MAQ”
  - Click on “Close” button
  - Click on “Apply changes” button
  - Restart Fiji



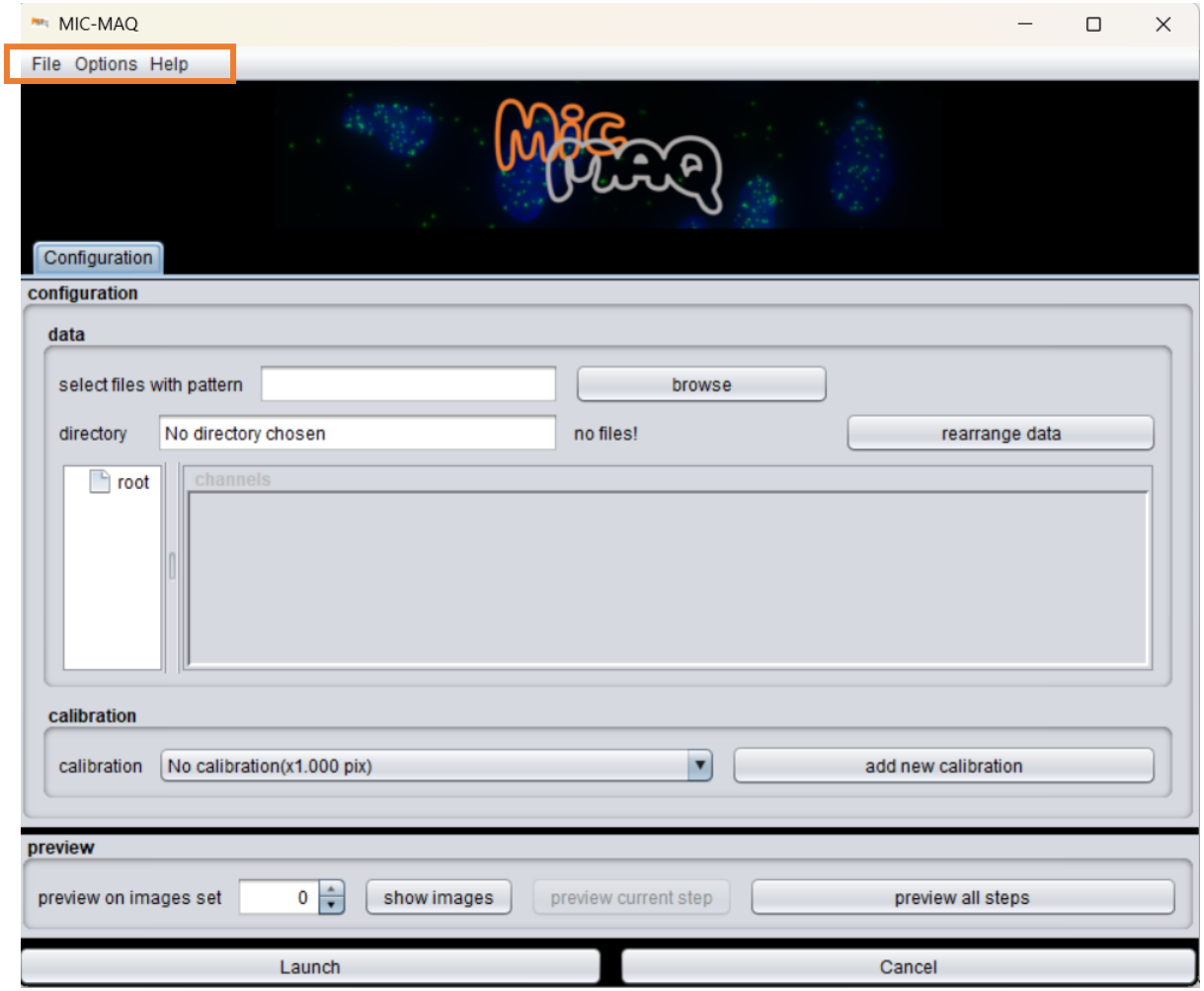


- **Open MIC-MAQ**

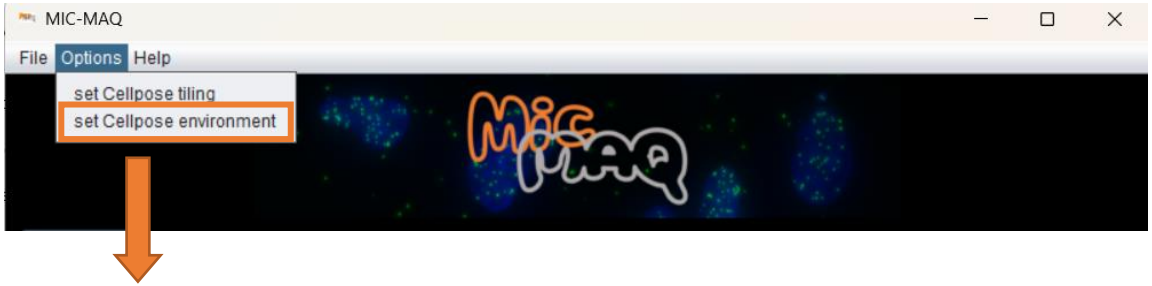
In the toolbar select: **Plugins > MIC-MAQ >**

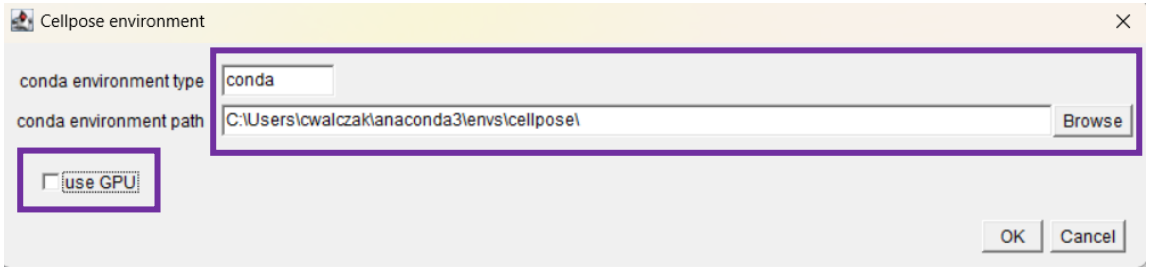


Configure Cellpose under MIC-MAQ



Configure Cellpose under MIC-MAQ





- ❑ Select Environment Type : **conda**
- ❑ Select the path to your working Cellpose virtual environment
- ❑ Check « use GPU » if cellpose is configured correctly

If the environment contains an install of Cellpose 4 (see slide 6), Cellpose will always run the model cpsam even if you choose another model.
If you need to use the cyto, cyto2, cyto3, nuclei or user-refined models, please install Cellpose3 as explained in slide 6

Now open the manual for explanations of MIC-MAQ workflow