




Version 2 ▼

Nov 02, 2022

S1: Step-by-step-guide using image J in the work flow. V.2

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ABSTRACT

How to use ImageJ in our 3D-workflow from data manipulation, import, alignment, segmentation and export as OBJ-file.

DOI

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PROTOCOL CITATION

Paul Kalke, Conrad Helm 2022. S1: Step-by-step-guide using image J in the work flow.. **protocols.io**
<https://dx.doi.org/10.17504/protocols.io.81wgb61xolpk/v2>
Version created by Paul Kalke



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CREATED

Nov 02, 2022

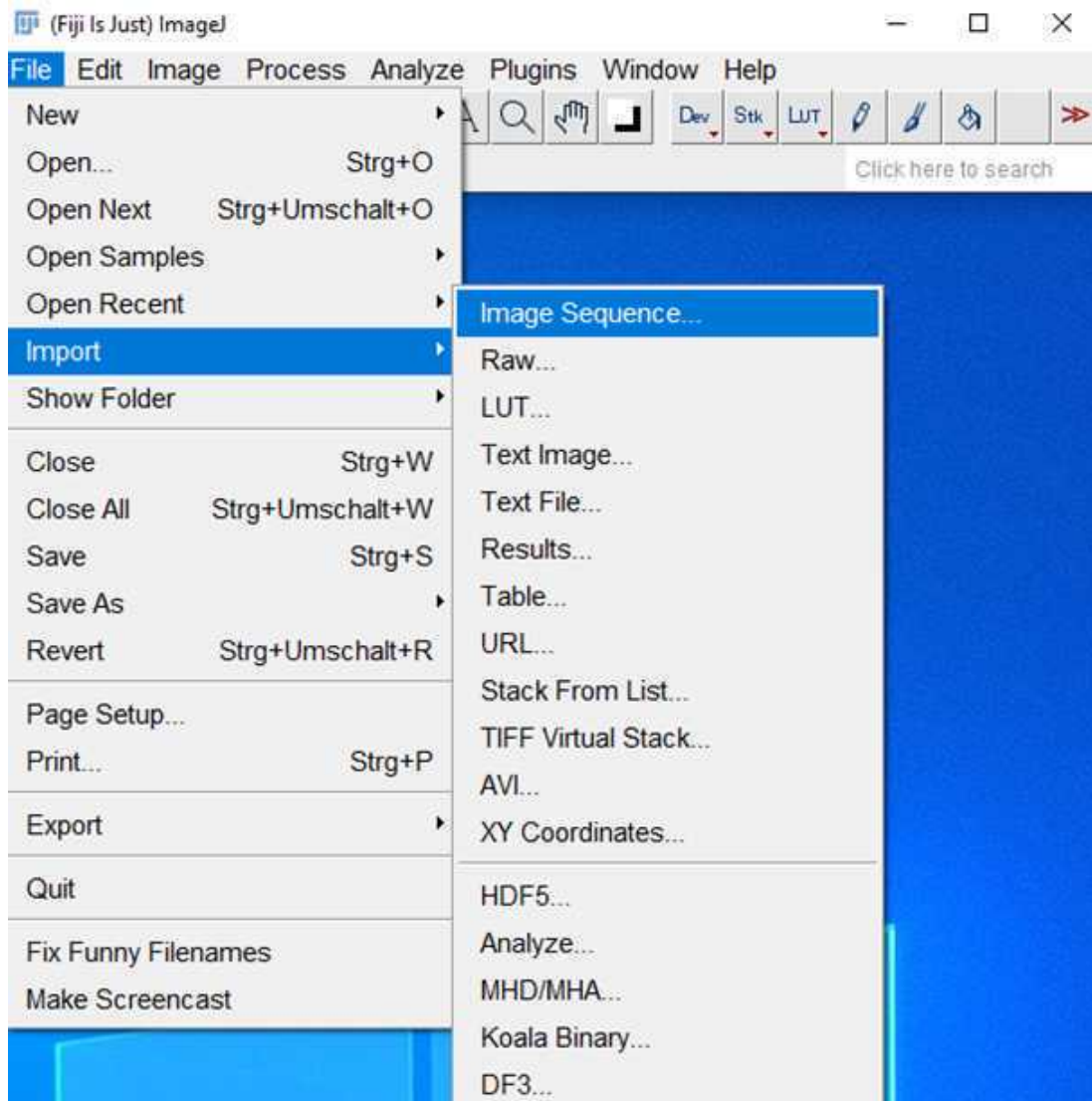
LAST MODIFIED

Nov 02, 2022

PROTOCOL INTEGER ID

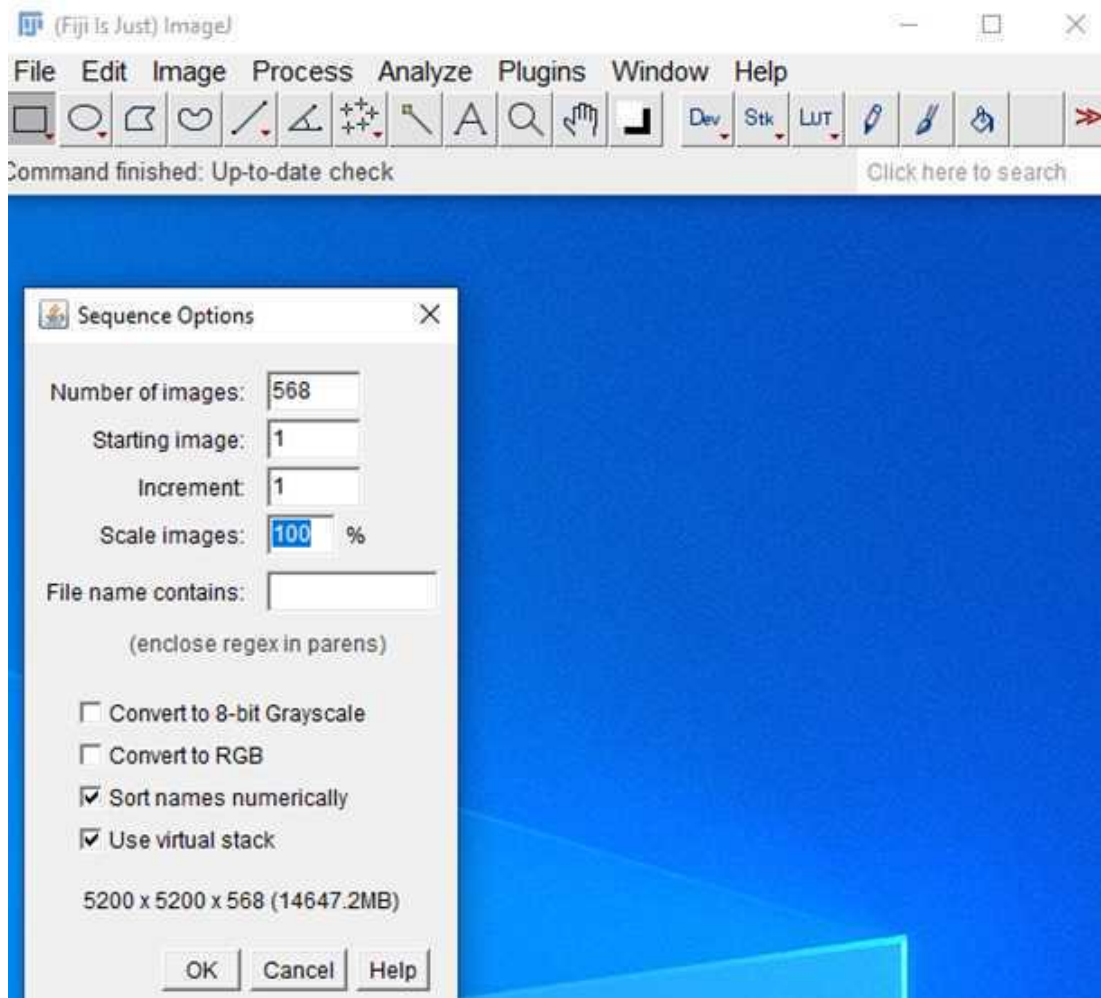
72198

1



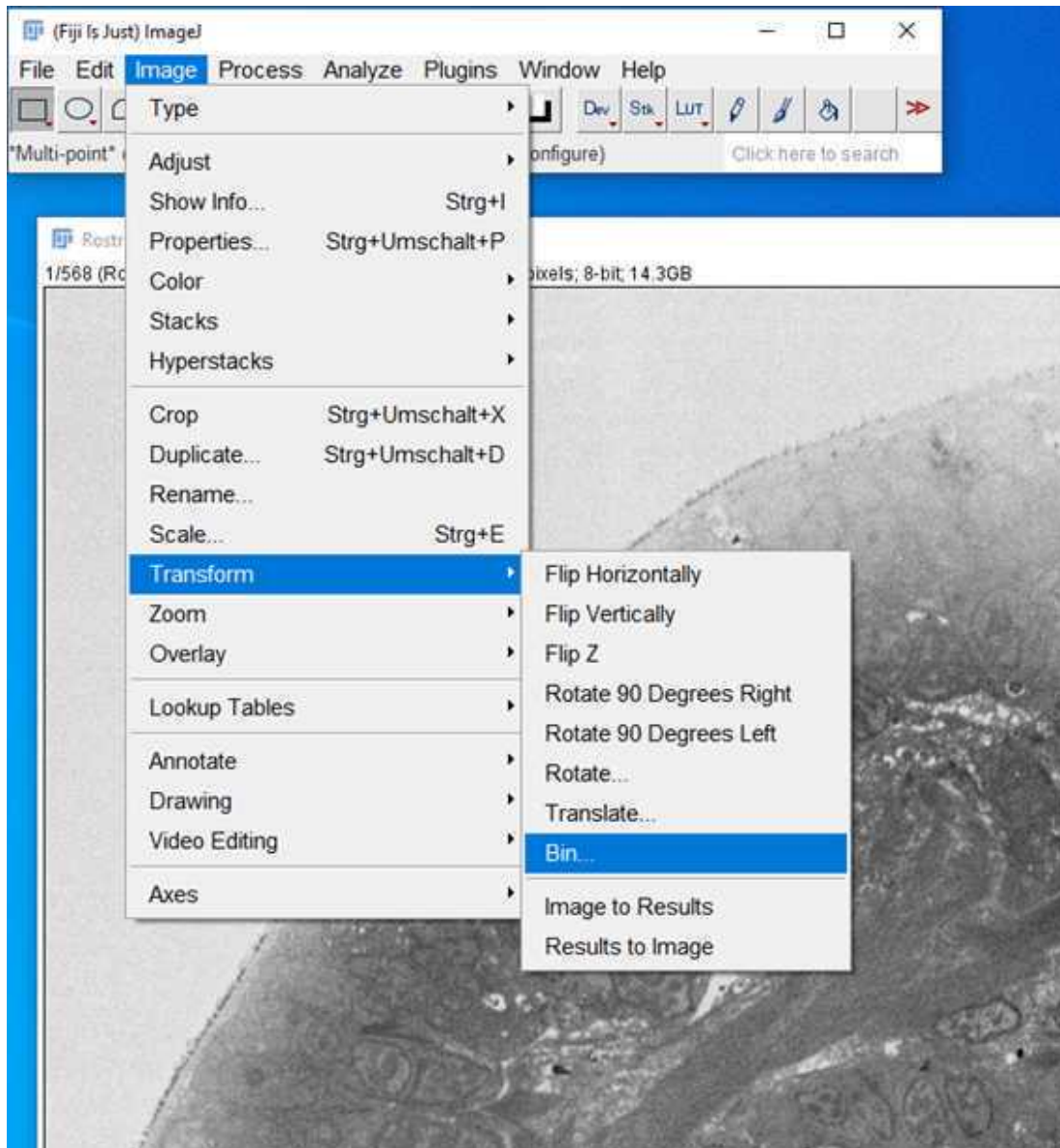
- Open Image J and import your *Image Sequence*
- It is possible in Image J to transform stacks into Images Sequences and vice versa

2



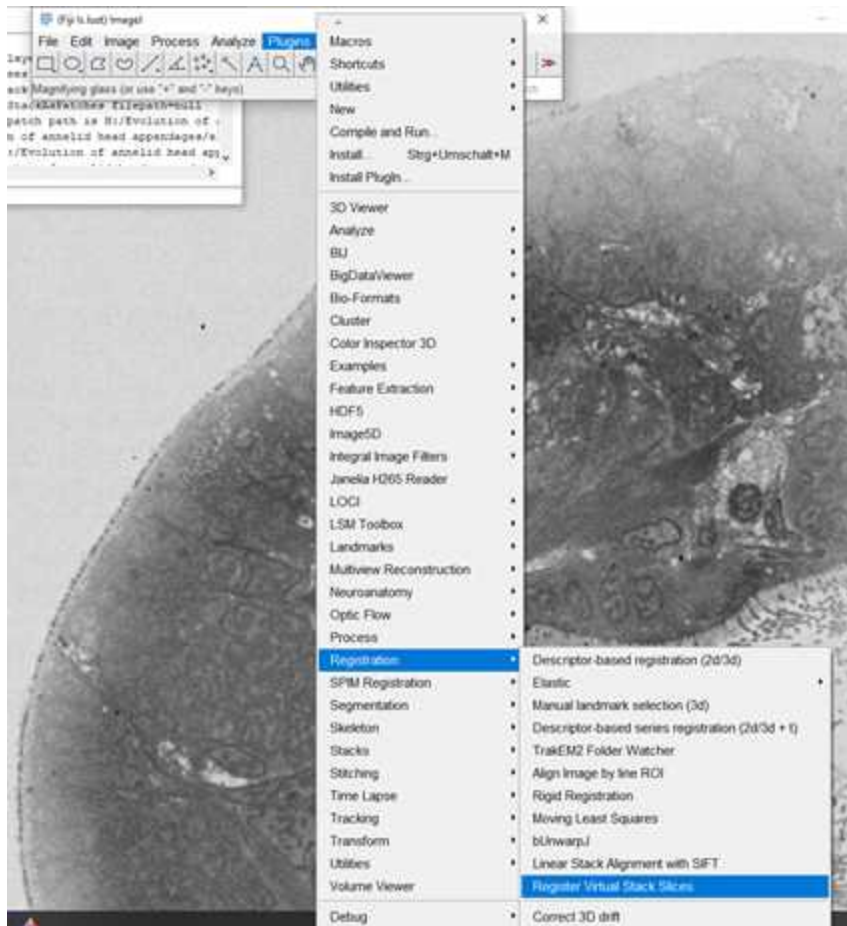
- Make sure your *Image Sequence* starts with 1 and choose *Use virtual stack* to save RAM
- You can scale your data set down too

3



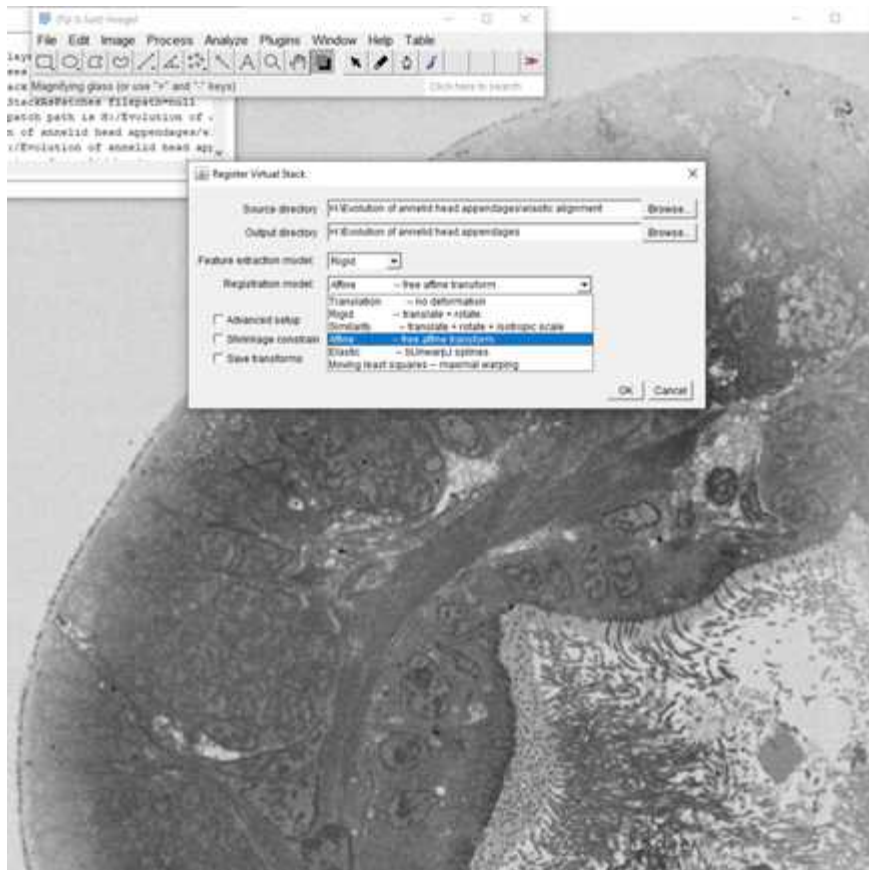
- Make sure your data set is processable for your computer
- Binning is easy and suitable for reducing data size, here *bin2* from 14.3 to 3.4 GB
- Only bin x and y not z
- Save binned data set

4

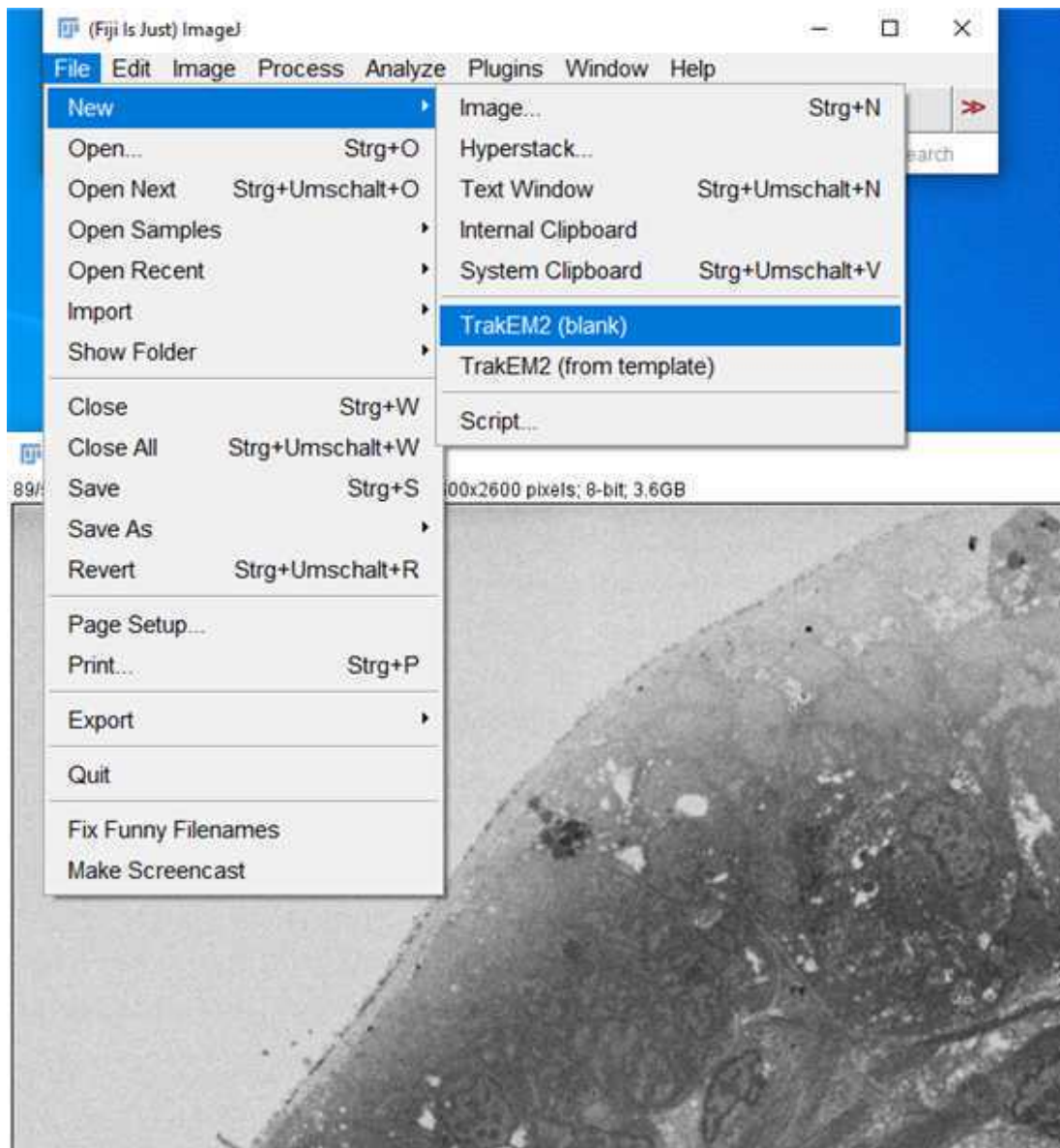


- For the alignment or registration of your virtual stack
 - Go to Plugins – Registration – Register Virtual Stack Slices
- Also see:
- <https://imagej.net/plugins/elastic-alignment-and-montage>

5

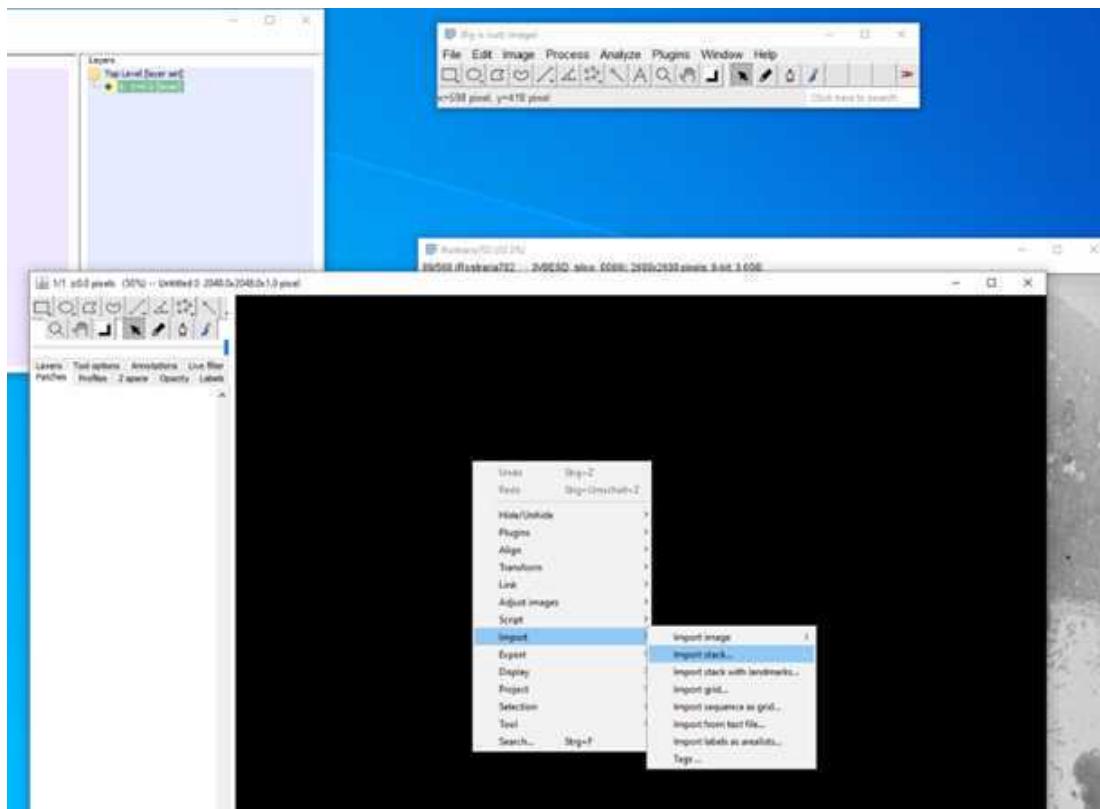


- Type in your Source directory and choose an Output directory
- You can choose a Feature extraction model and a Registration model
--> Our best results were with shown models
- After clicking on OK, click on first image of your stack and its done



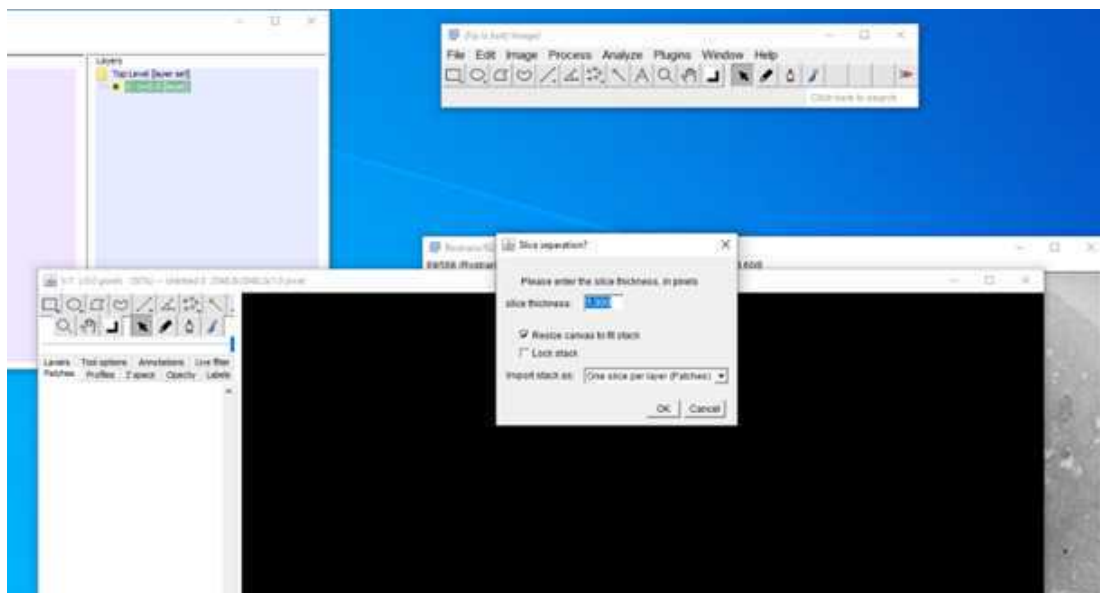
- For reconstruction, use *TrakEM2*
- Open *blank*
- Also see, the Trek EM2 User guide:
<https://www.youtube.com/watch?v=kUyXQRcKWOk>

7



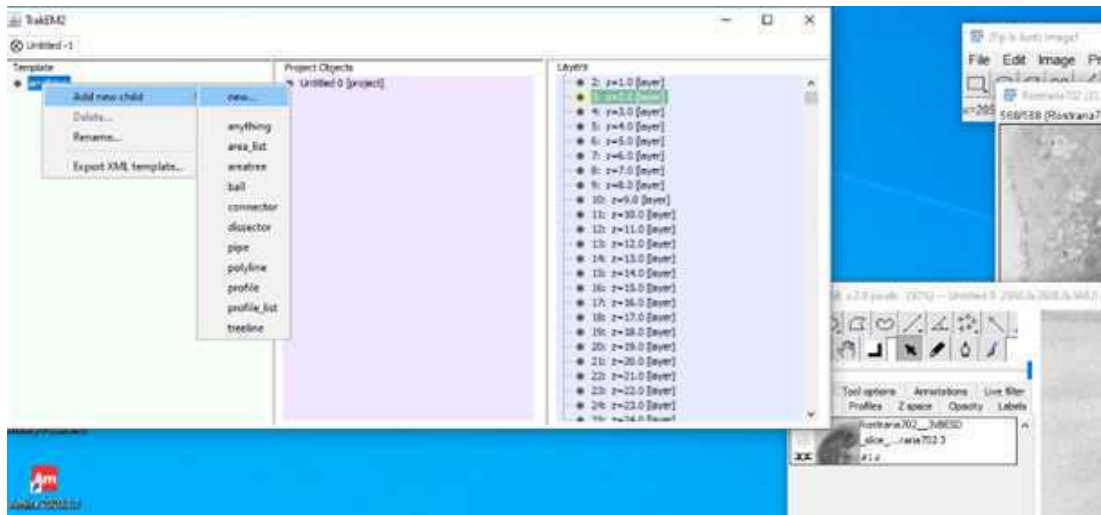
- Import your binned data set by right click in blank (black window) and select folder of binned data set

8



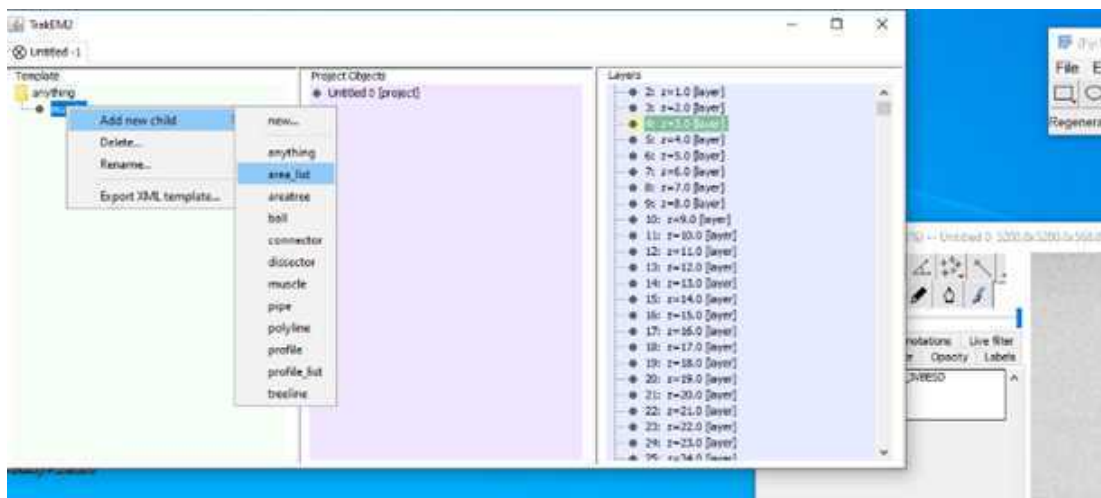
- Choose *slice thickness* depending on your data set, adjustments are possible in all programs

9



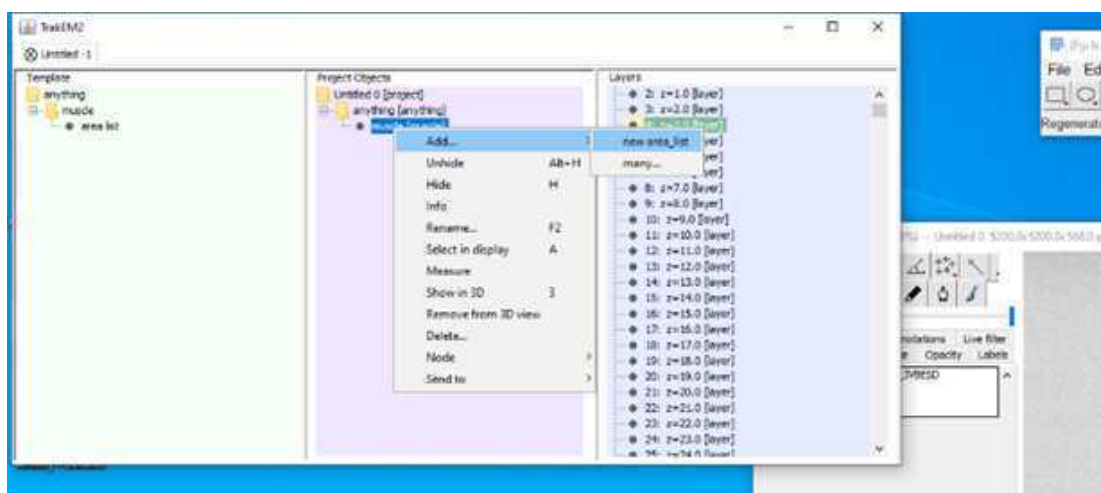
- To create different labels, each for a structure you want to reconstruct, you need a *new child* and give it the name of your label

10



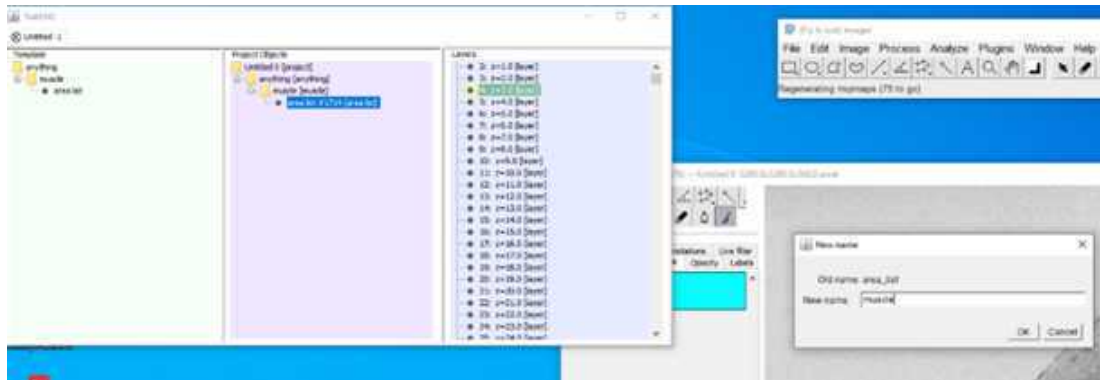
- Then right click on the *new child* and create an *area_list*

11



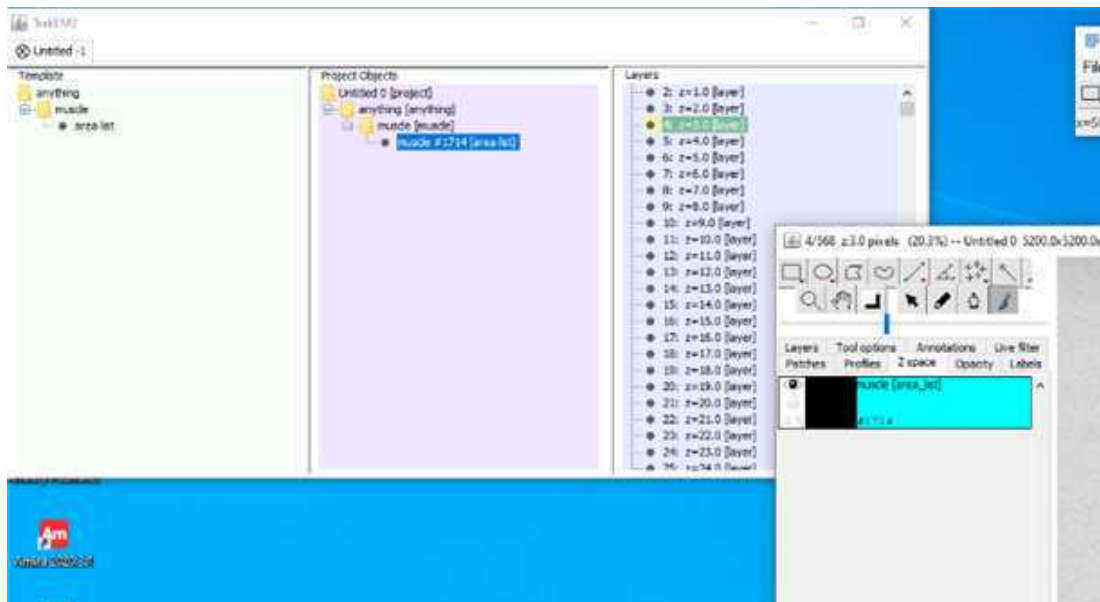
- Drag `n` drop the named folder (child) together with the *area_list*
- Right click on the *area_list* and add a *new area_list*

12

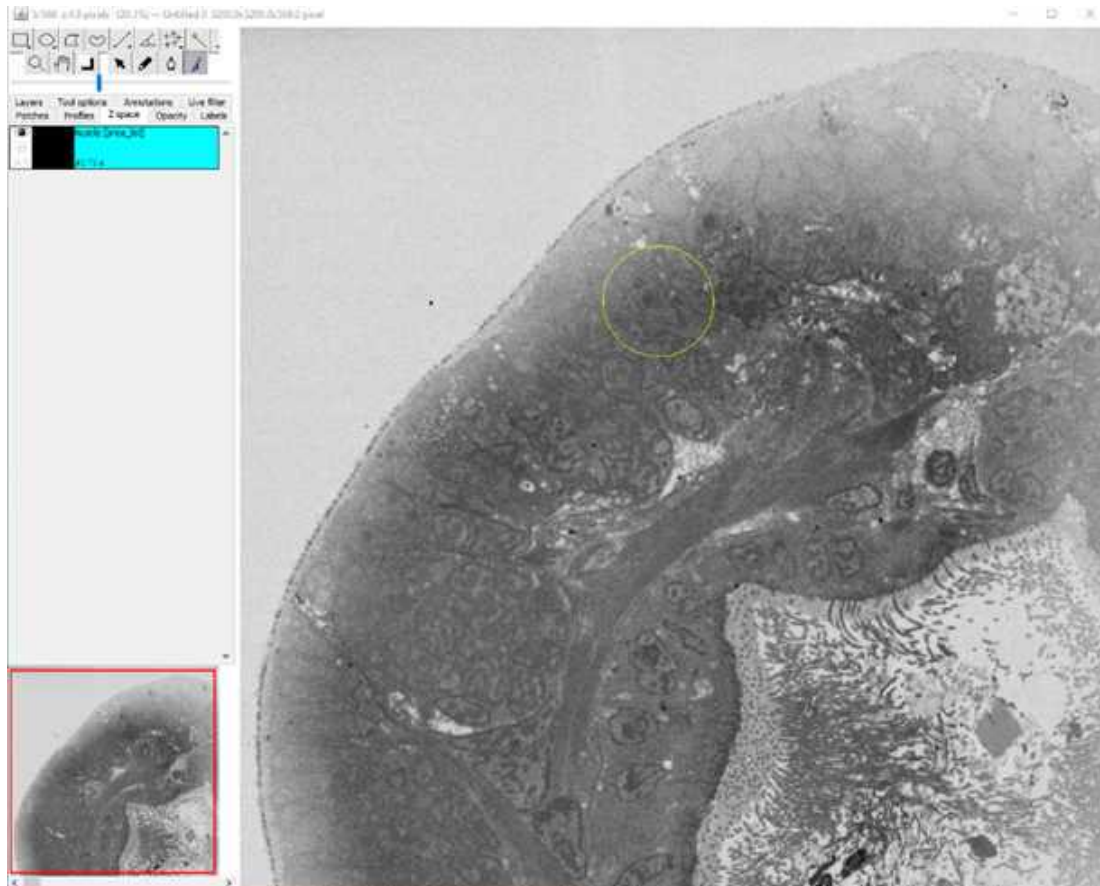


- Name the *new area_list* like the child in the beginning
- save your Trak EM2 file by press save in main tool bar (.xml-file)

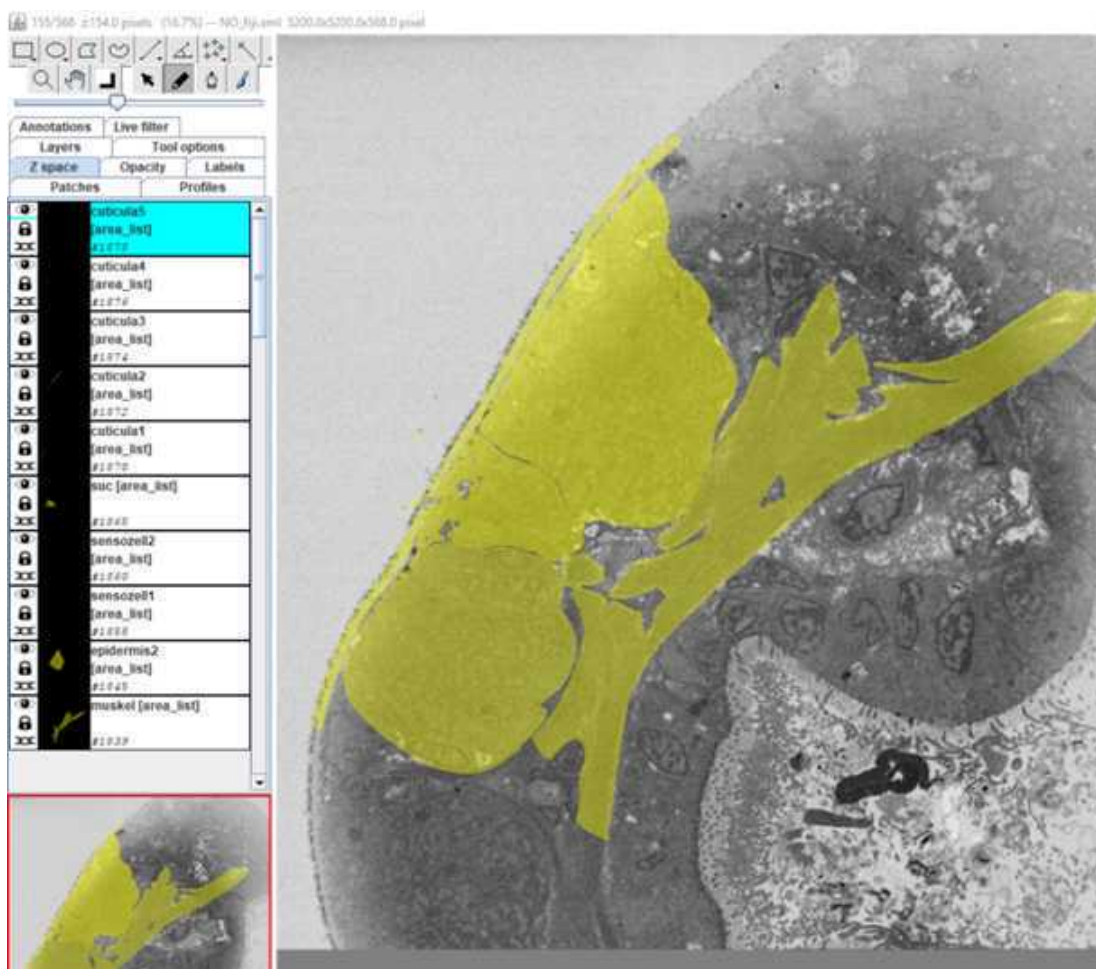
13



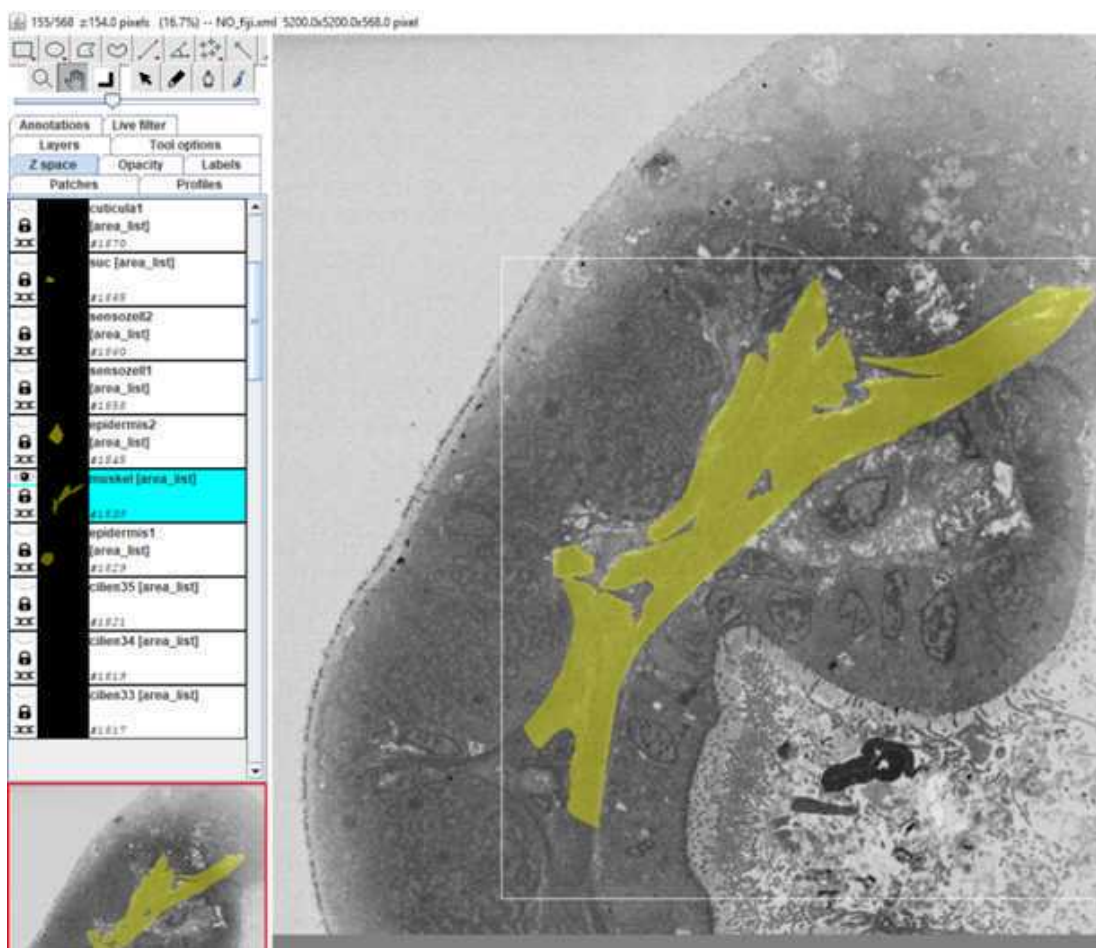
- Now your named label is available for segmentation under *z space*



- Now start to segment your structure
- Make sure the brush tool in the main tool bar is checked
- Hot keys and how to reconstruct:
 - Mouse wheel (mw); , and . : scrolling through data set
 - Ctrl + mw; + and - : zoom in and out
 - Shift + mw: size of pen
 - Left mouse button (l mb): mark
 - Shift + l mb: fill
 - Alt + l mb: erase
 - Alt + shift + l mb: delete all on slice
 - Right mouse button (r mb) – *Area*: for all kind of interpolations

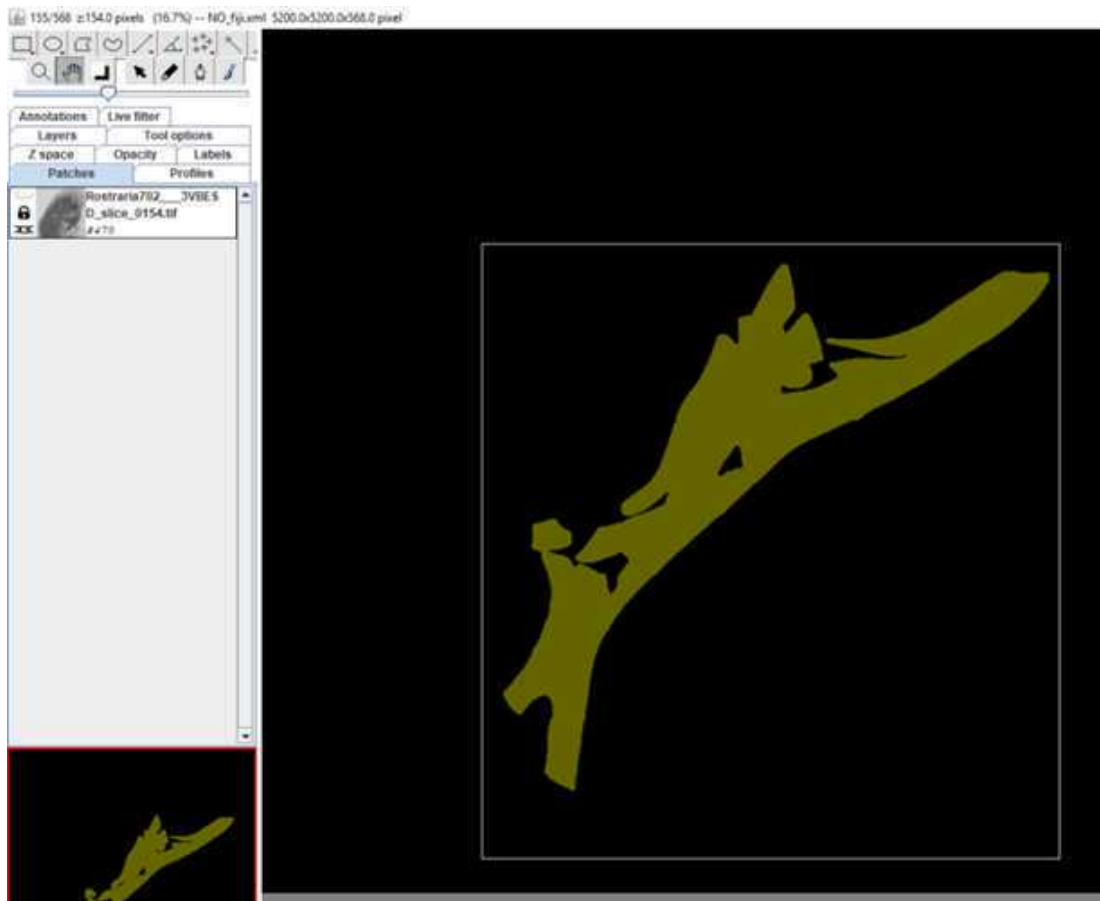


- after finishing your segmentation it is time to export your labels
- it is possible to export different labels solely, or together, but then they work like one
- e.g. in this example it is smart to export all parts of the cuticle as one label



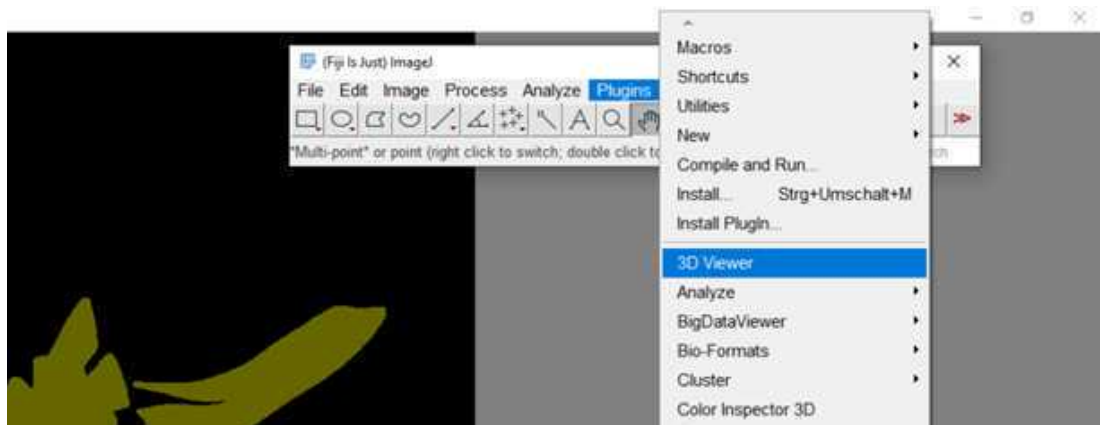
- hide all labels you do not want to export in this turn by unchecking the eye button in the Z space

17

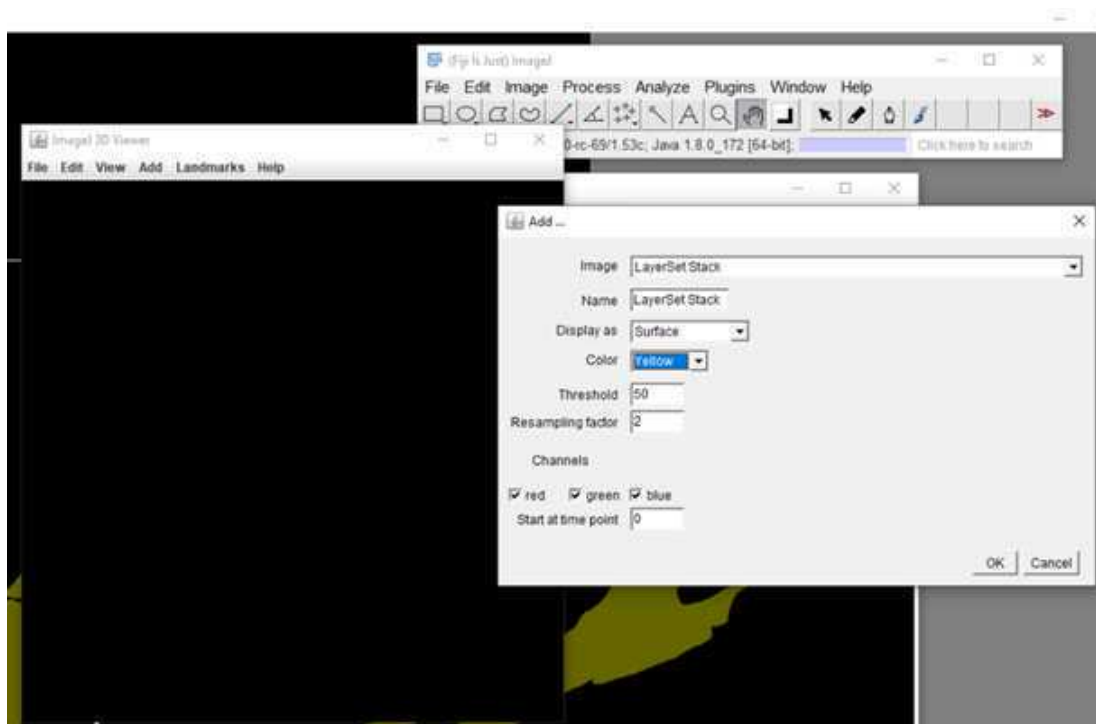


- now turn the "background" black, go to patches and push the fader on top to the left
- now you should only see your marked label, check the whole stack

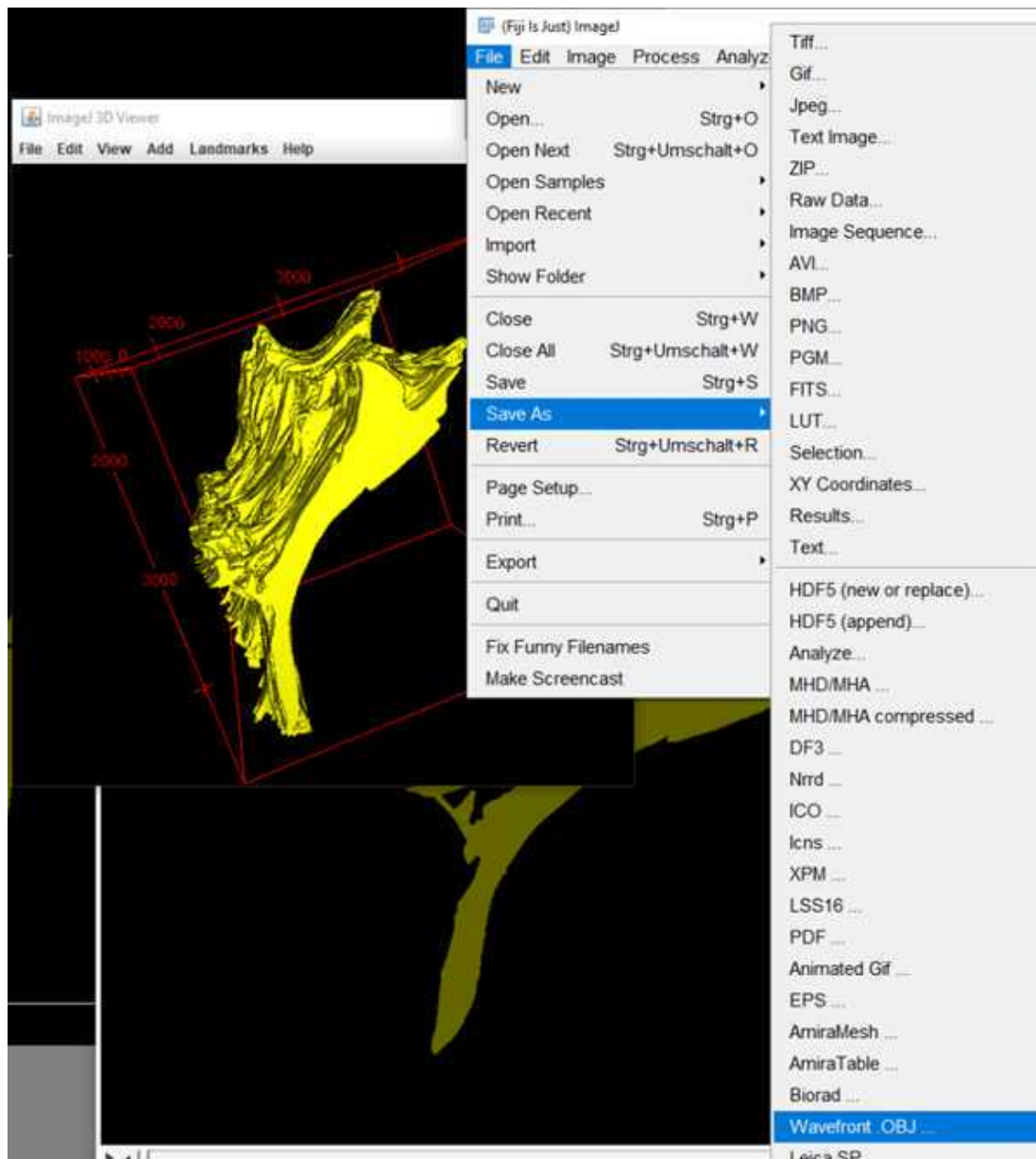
18



- open the 3D viewer

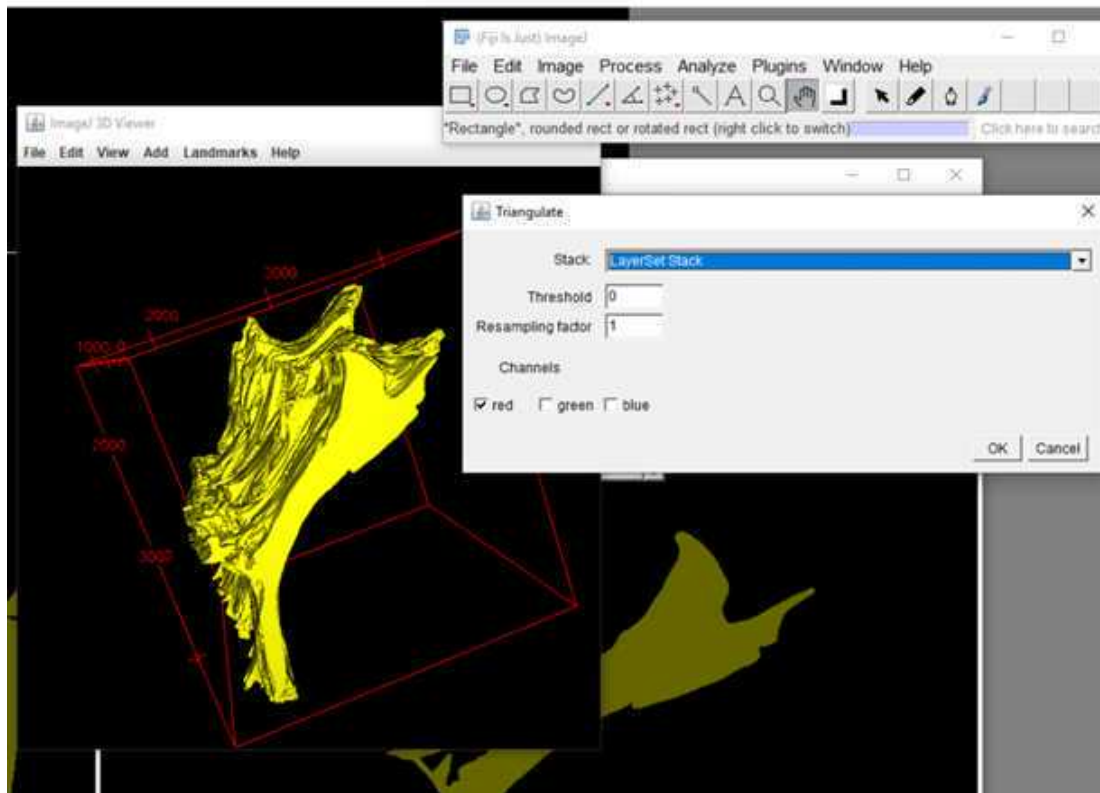


- you can choose in *Display as* between *surface or volume*
- for later work with MeshLab and Blender it doesn't matter
- if you are not familiar with ImageJ keep the other parameters as in the shown image



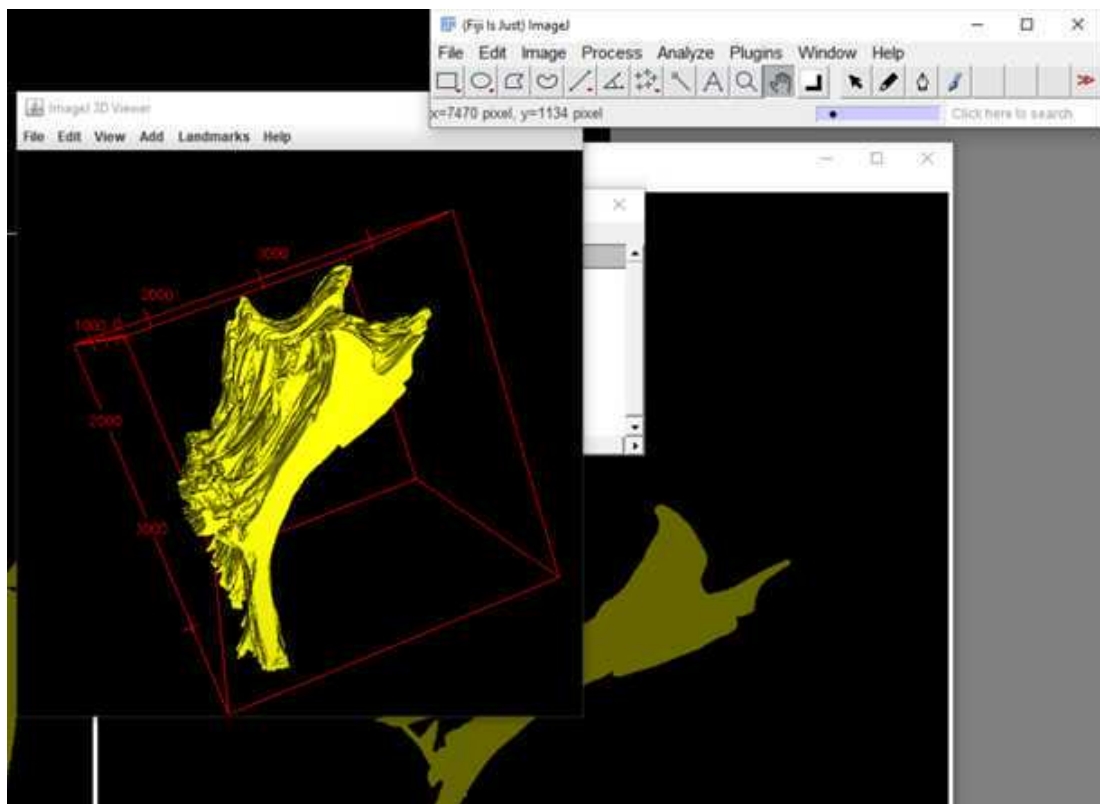
- press ok and wait for the result
- if it is stretched or clinched don't bother, you can change later or go back to the *TrakEM2* window (see last step)
- save label as *wavefront*-file (.OBJ)

21

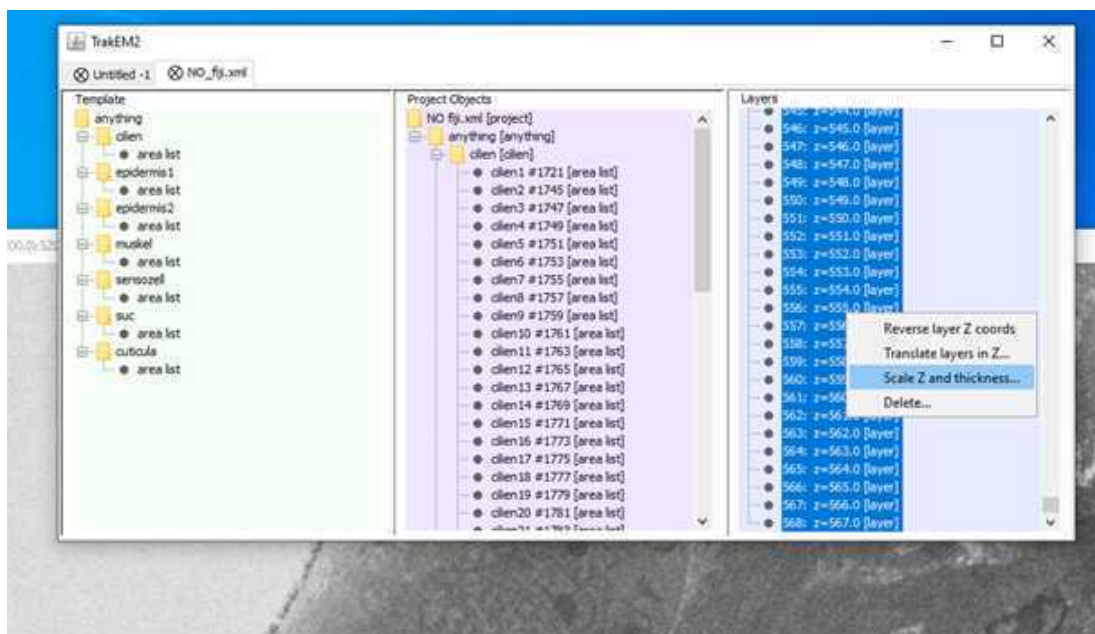


•use parameters as shown in image, press ok

22



•wait for the little black dot in the main tool bar before you export other labels



- if you want to change the slice thickness go to *Layers* of the *TrackEM2* window, select all slices and right click
- Choose a factor for all slices