Imaging and Microscopy Center Biomedical Core Facility



הפקולטה לרפואה ע״ש רות וברוך רפפורט הטכניון - מכון טכנולוגי לישראל

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2013-06-17 how to insert scale bar post acquisition in FIJI.docx

Obtain an image with a defined calibration grid in the same conditions: microscope, pixel camera size, objective. Please ask staff for help.

Attach is our grid, image distance between 2 major gridlines is 100um.

Open the image in Fiji.

Analyze → Set Scale... Make sure Global is unchecked Click "Click to Remove Scale" OK





From the tool bar choose the line tool.

At the calibration grid draw a line of a known distance,

e.g. between major gridlines, precisely, using magnification.

At the status bar below the tool bar you see the number of pixels you have indicated as length, as long as you hold the line.



Analyze  $\rightarrow$  Set Scale Set Scale III Set Scale "Distance in pixels" should already show the number you saw Distance in pixels: 75 Distance in pixels: 75 on the status bar. 100 Known distance: 0.00 Known distance: "Known distance" enter the Pixel aspect ratio: 1.0 Pixel aspect ratio: 1.0 number you know this distance Unit of length: pixel Unit of length: μm represents, e.g., 100 between major gridlines. Click to Remove Scale Click to Remove Scale Global "Unit of length" enter µm (this will bbal be used literally in the scale bar) Scale: <no scale> Scale: 0.750 pixels/µm Check Global OK | Cancel Help OK | Cancel Help OK

In any picture you wish to apply a scale bar with this calibration (i.e., similarly acquired) you do: Analyze  $\rightarrow$  Tools  $\rightarrow$  Scale bar...

You can indicate the length and formatting parameters, and if you wish it not to be in one of the standard corners you can (beforehand) draw a line in the location you wish it to be placed.