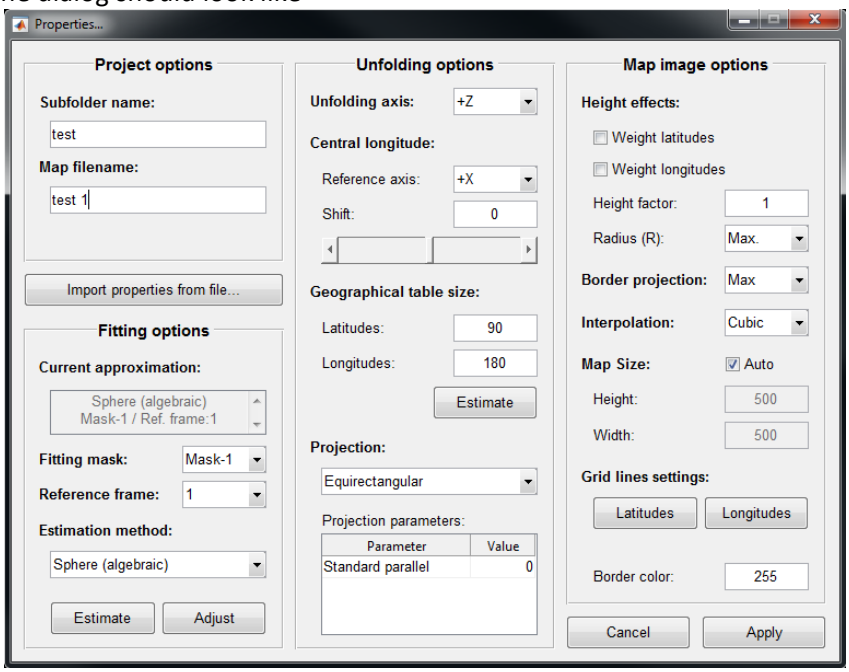
Map3-2D tutorial

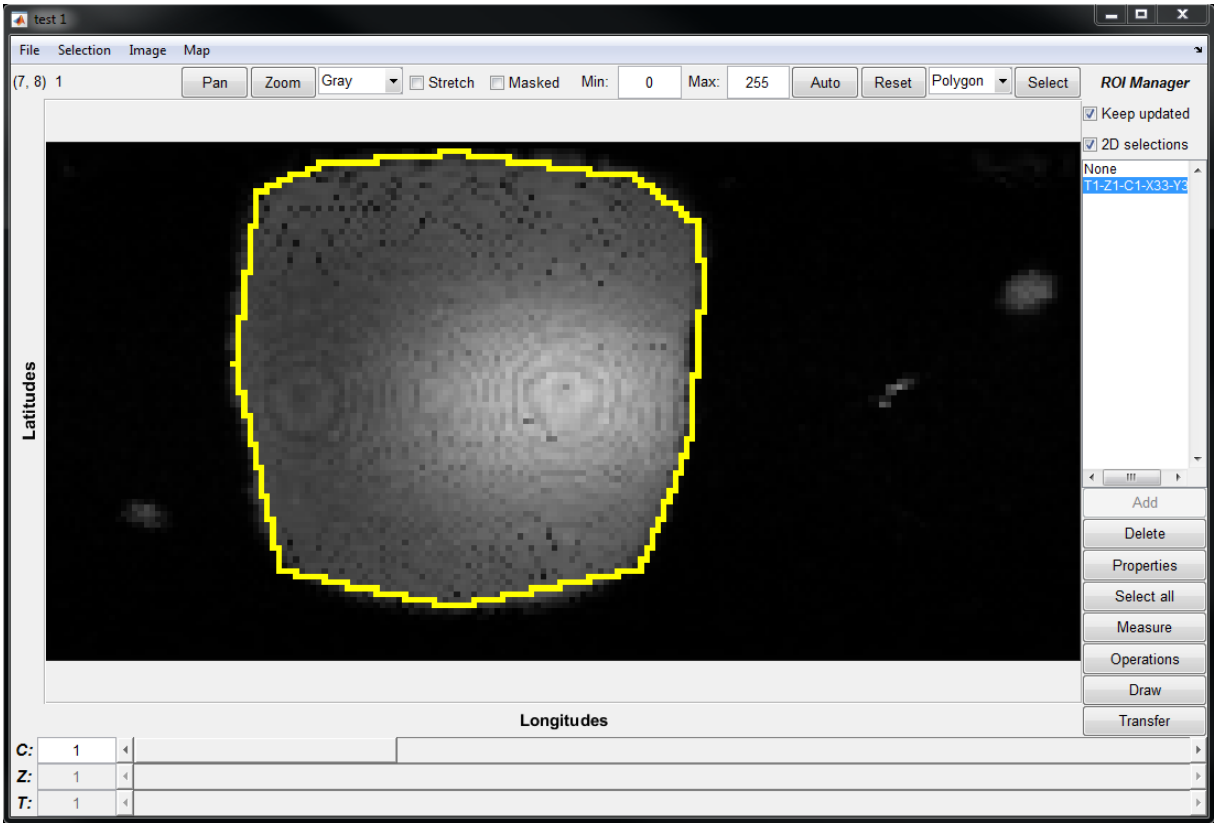
Version 1.0

GUV surface projection

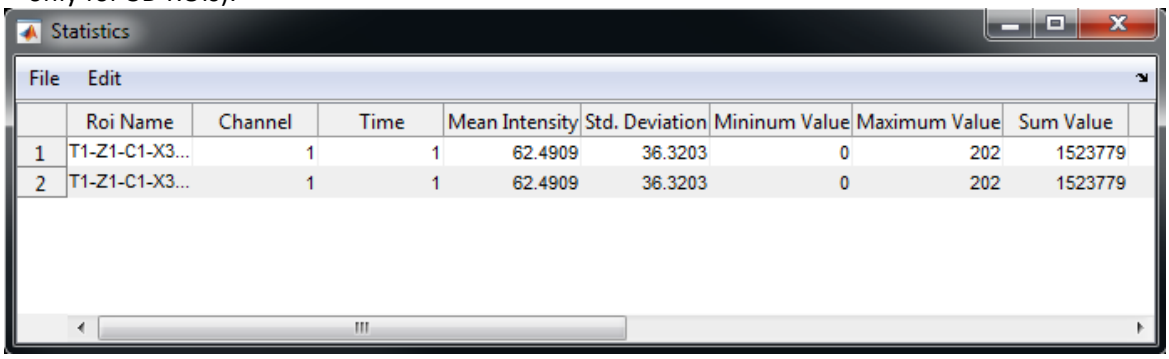
1. Uncompress the file “GUV.zip”. A folder will be created with the files “GUV.tif”, “GUV_Mask_thin.tif” and “GUV_Mask_thick.tif”
2. Run Map3-2D and open the file “GUV.tif”. This is a 2-channel 3D stack with 100 slices (check the sliders at the bottom of the window).
3. If a dialog appears to set this folder as the default folder, then click on *Yes*.
4. A voxel dialog will pop up to check if the detected voxel size is correct. Click on *Apply*.
5. In the menu bar go to *Image -> Add Mask 1*. A dialog will pop up.
6. Select the option *Load from an image stack file* and click on *Continue*
7. Open the file “GUV_Mask_thin.tif”. Everything that is not in the mask will be shown as background. The background color can be modified
8. In the menu bar go to *Process -> 2D projection*. A new window will be created and a *Properties* dialog will pop up on top of it.
9. In the Subfolder name write “test”
10. In the Map filename write “test 1”
11. As the GUV has a spherical shape, you can choose *Sphere* as the *Estimation method* and click on *Estimate.* The dialog should look like



12. Press *Apply* to get the corresponding 2D projection.
13. Use the sliders to check the two additional channels (grid and height). As the GUV is very spherical without any protrusion/indentation, the information on the height channel is almost zero. The display range can be enhanced with the *Auto* button in the toolbar.
14. Use the *Selection -> Make selection -> Polygon* tool (or use the shortcut on the toolbar) to make a selection on the first channel, and add it to the ROI Manager.



15. In the sidebar click on *Transfer* (and *Apply* in the dialog) to see this region in the main window.
16. Go to the main window and click on the new ROI that appeared in the ROI Manager. This is a 3D ROI which is different for every slice. Please move the *Z* slider to see the ROI extent.
17. In the sidebar click on *Measure*. A measurements table will pop up with information about this 3D ROI.
18. Repeat the same but pressing [Ctrl] + *Measure*. Only the current slice will be measured (valid only for 3D ROIs).



	Roi Name	Channel	Time	Mean Intensity	Std. Deviation	Mininum Value	Maximum Value	Sum Value
1	T1-Z1-C1-X3...	1	1	62.4909	36.3203	0	202	1523779
2	T1-Z1-C1-X3...	1	1	62.4909	36.3203	0	202	1523779

19. Add an extra channel in the main window image (*Image -> Add extra channel*).
20. Select this extra channel with the *C* slider and select the ROI in the ROI Manager (if not already selected).
21. In the sidebar click on *Draw* and press *Apply* in the subsequent dialog. The selection is now drawn in this additional channel.
22. Return to the 2D projection window and click on *Map -> Transfer Grid* at the menu bar.
23. Go back to the main window and click on *Menu -> Save*. The saved image(s) can contain the main channels and/or additional channels. Leave the default options and click on *Save*. The resulting image is a BigTiff file (BTF) that can be opened in ImageJ/FIJI.