MAP_ROIS MATLAB App README

Mai-Anh Vu

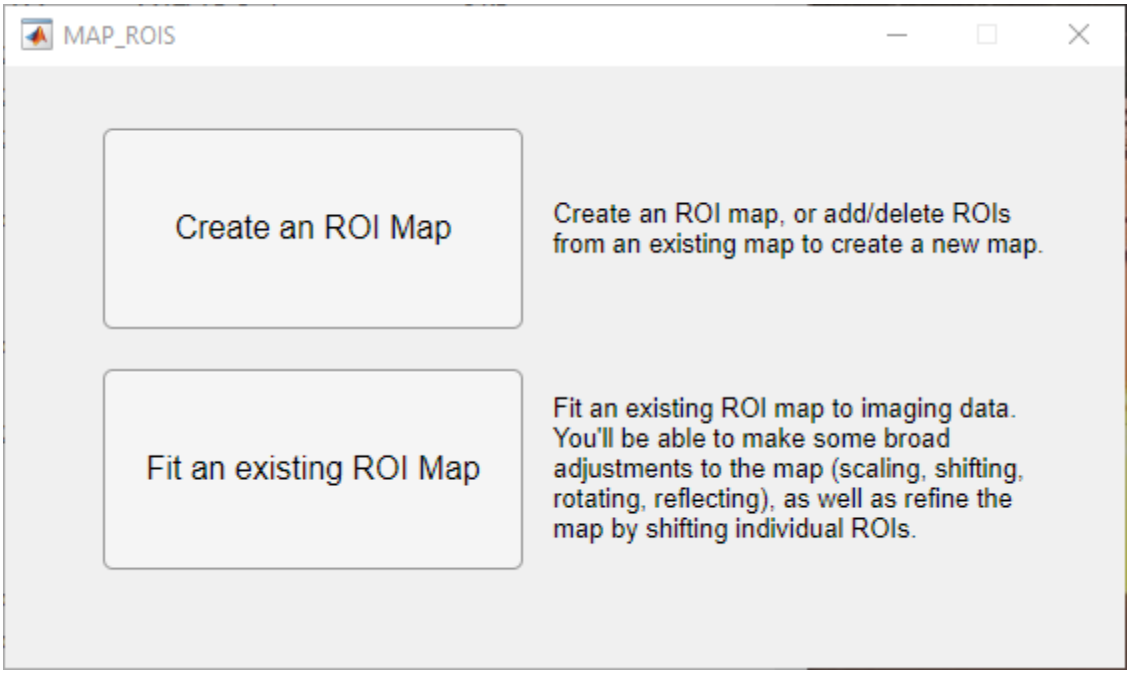
3/25/2024

ROIs struct

All of these functions generate a .mat file that contains the following fields:

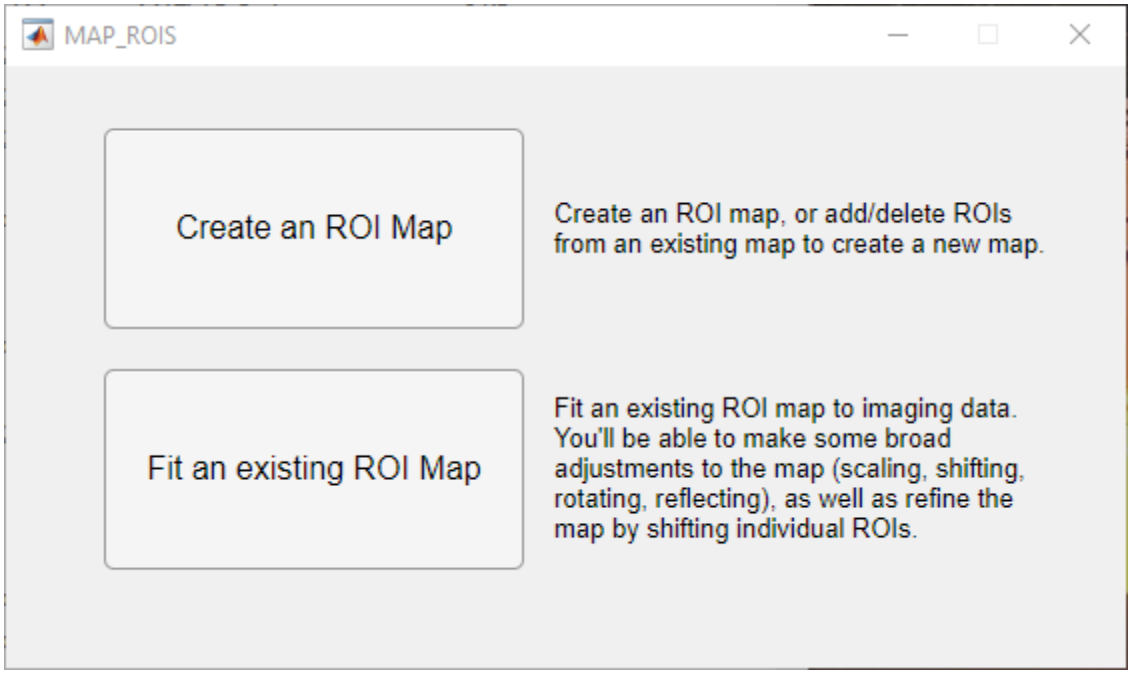
- ROIs – the centers of the ROIs
- datapath – the path to the associated .tif file
- snapshot – a snapshot of a frame from the .tif movie
- radius – the radius of the ROIs
- ROImasks – an m x n x p matrix of p binary ROI masks
- FtoFcWindow – the window used to calculate baseline
- F – the extracted raw fluorescence
- Fc – the calculated ΔF/F
- Fc_baseline – the calculated baseline
- Fc_center – the calculated center, which becomes 0

MAP_ROIs



These let you assign the ROIs on the imaging data, and extract the fluorescence timeseries (i.e., averaging the pixels within each ROI per frame).

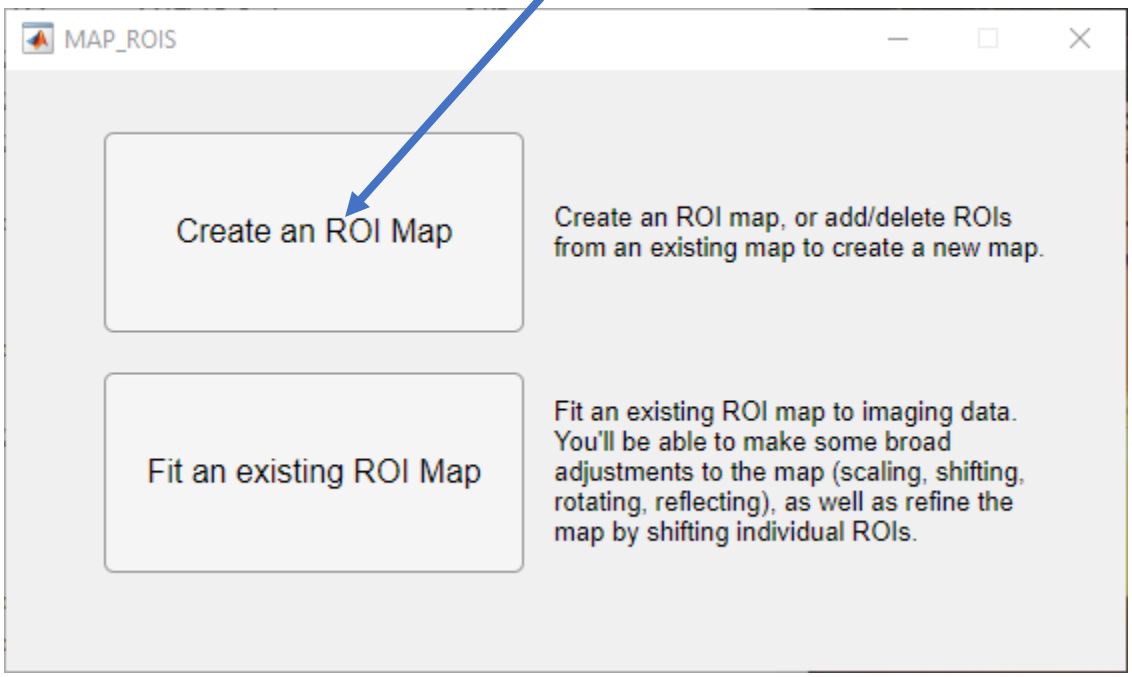
Running the standalone functions outside of this GUI



These 2 buttons call the functions **create_ROI_map.m**, and **fit_ROI_map.m**, respectively. You can run either of these functions from the command line without going through this GUI.

Create an ROI map

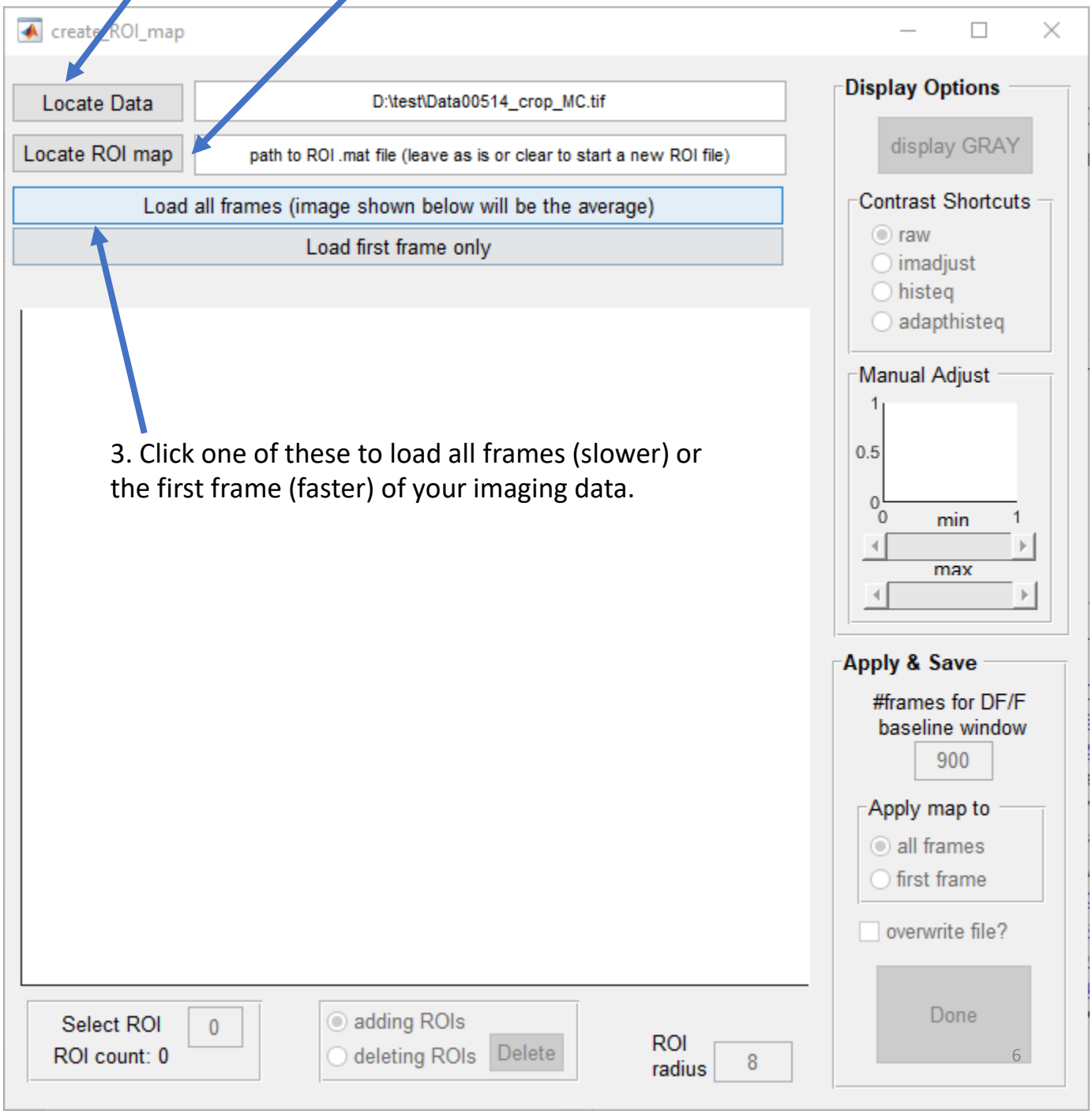
Use this one if you're fitting an ROI map to some imaging data *for the first time* or if you want to edit an existing map to create a *new* map.



Loading data

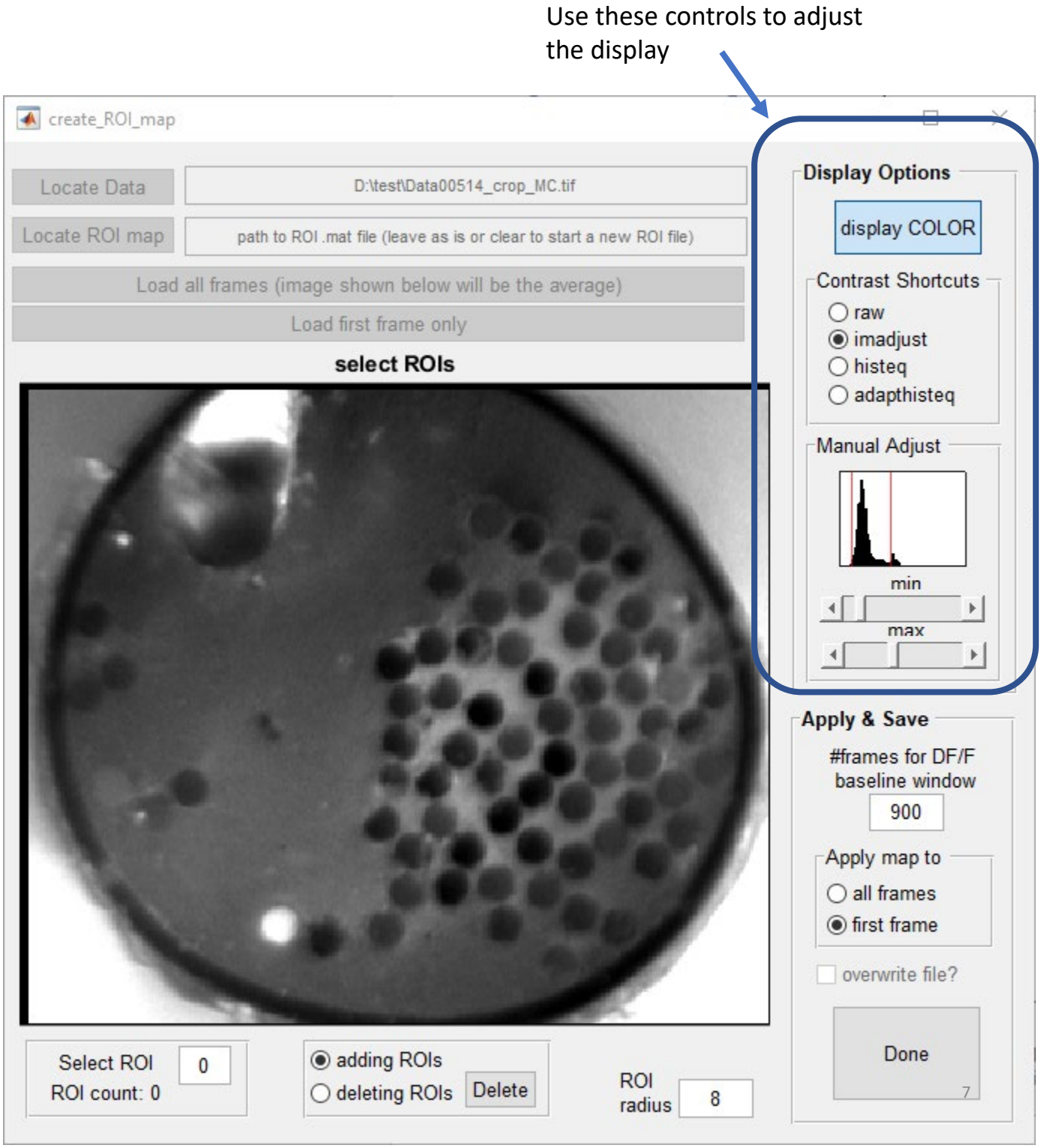
1. Click here to locate your .tif data, or enter the path into the text field

2. If you're editing an existing ROI map to *create a new map*, click here to locate the ROI map, or enter the path into the text field. Ignore this if you're creating a new ROI map de novo

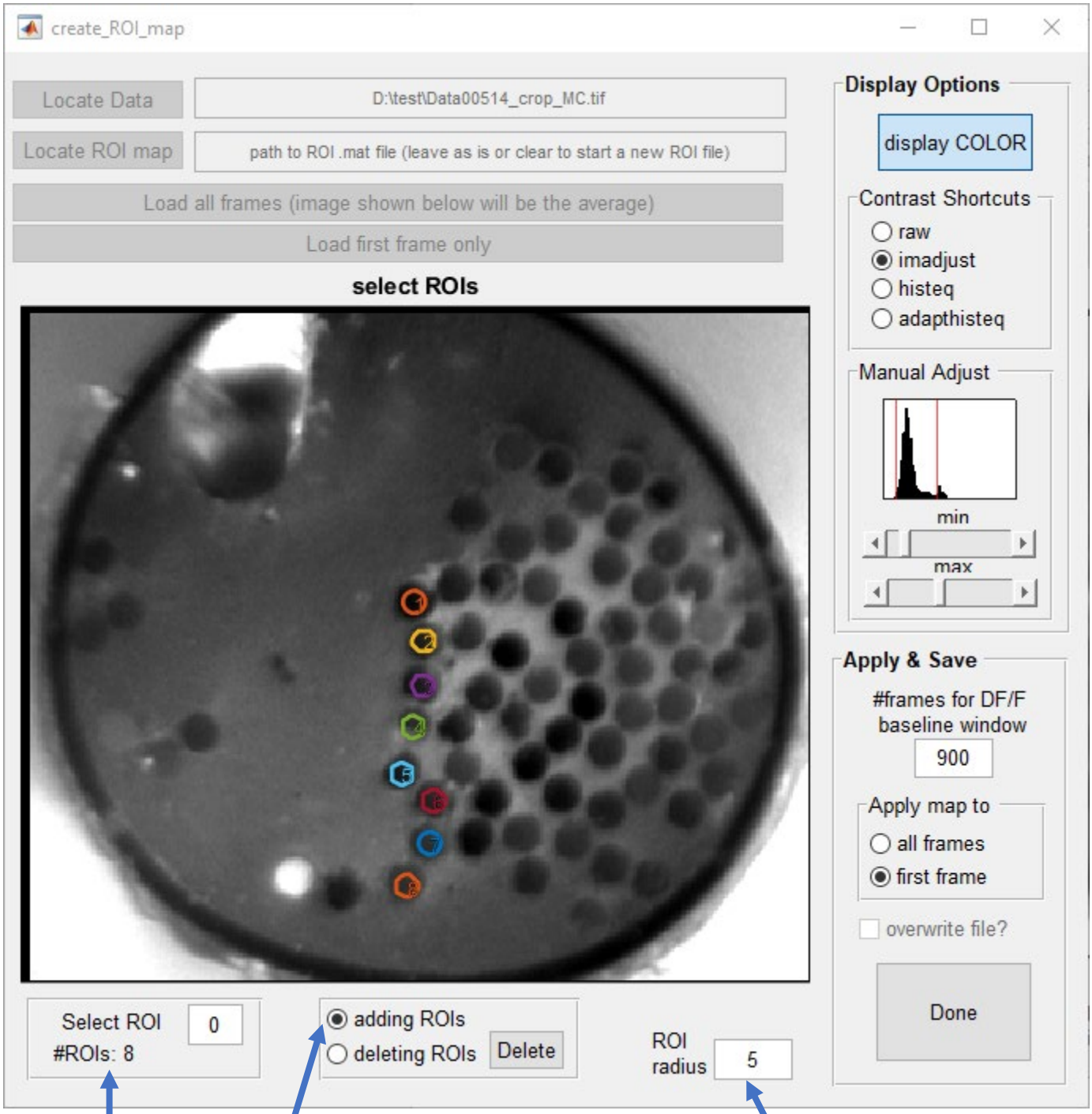


3. Click one of these to load all frames (slower) or the first frame (faster) of your imaging data.

Adjusting display



Adding ROIs

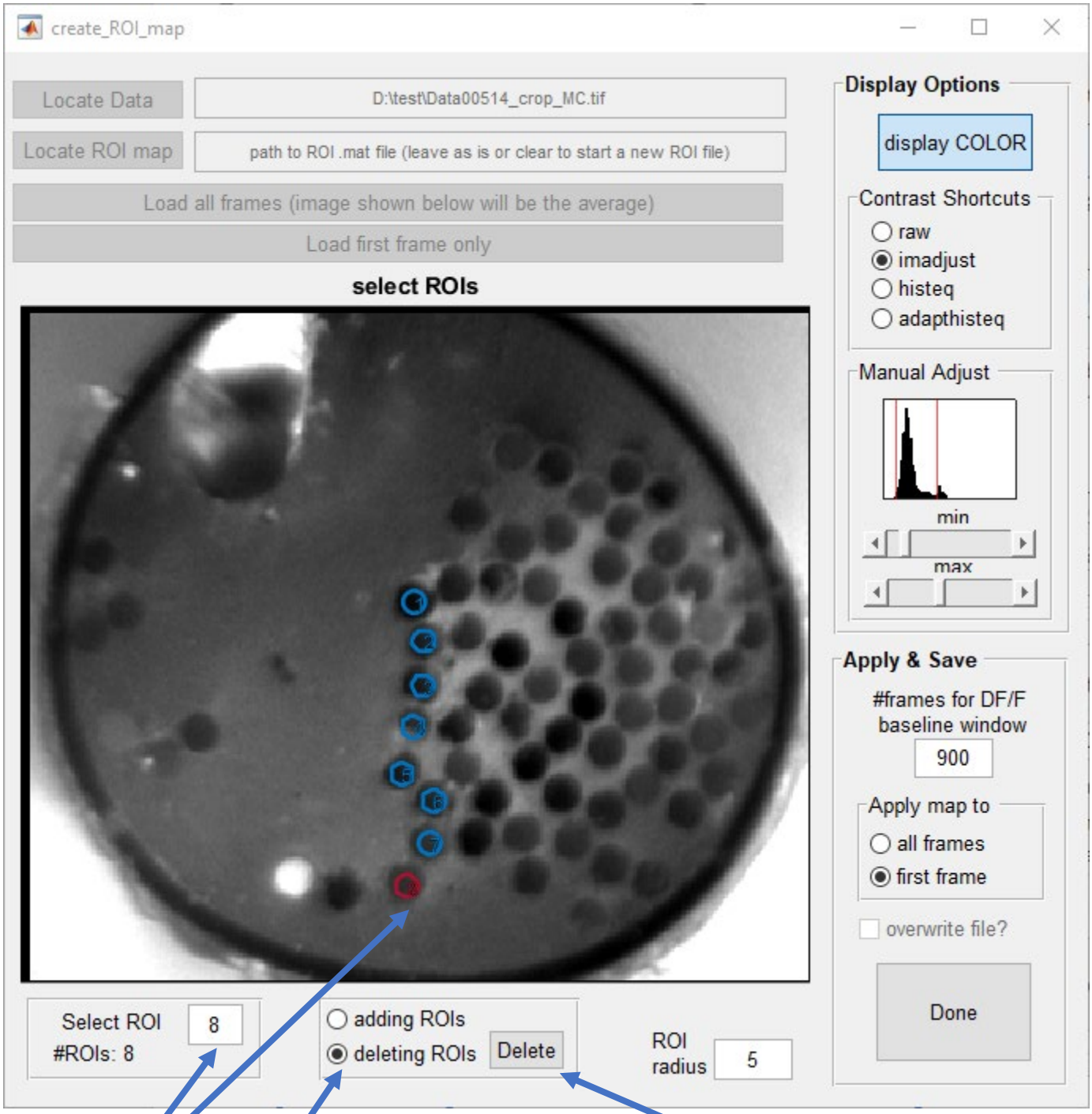


Total # ROIs

In adding mode, clicking on the image will add an ROI centered on your click

Set the radius (pixels) here. All ROIs will have the same radius. The fit ROI or refine ROI steps enable 2 different radii.

Deleting ROIs

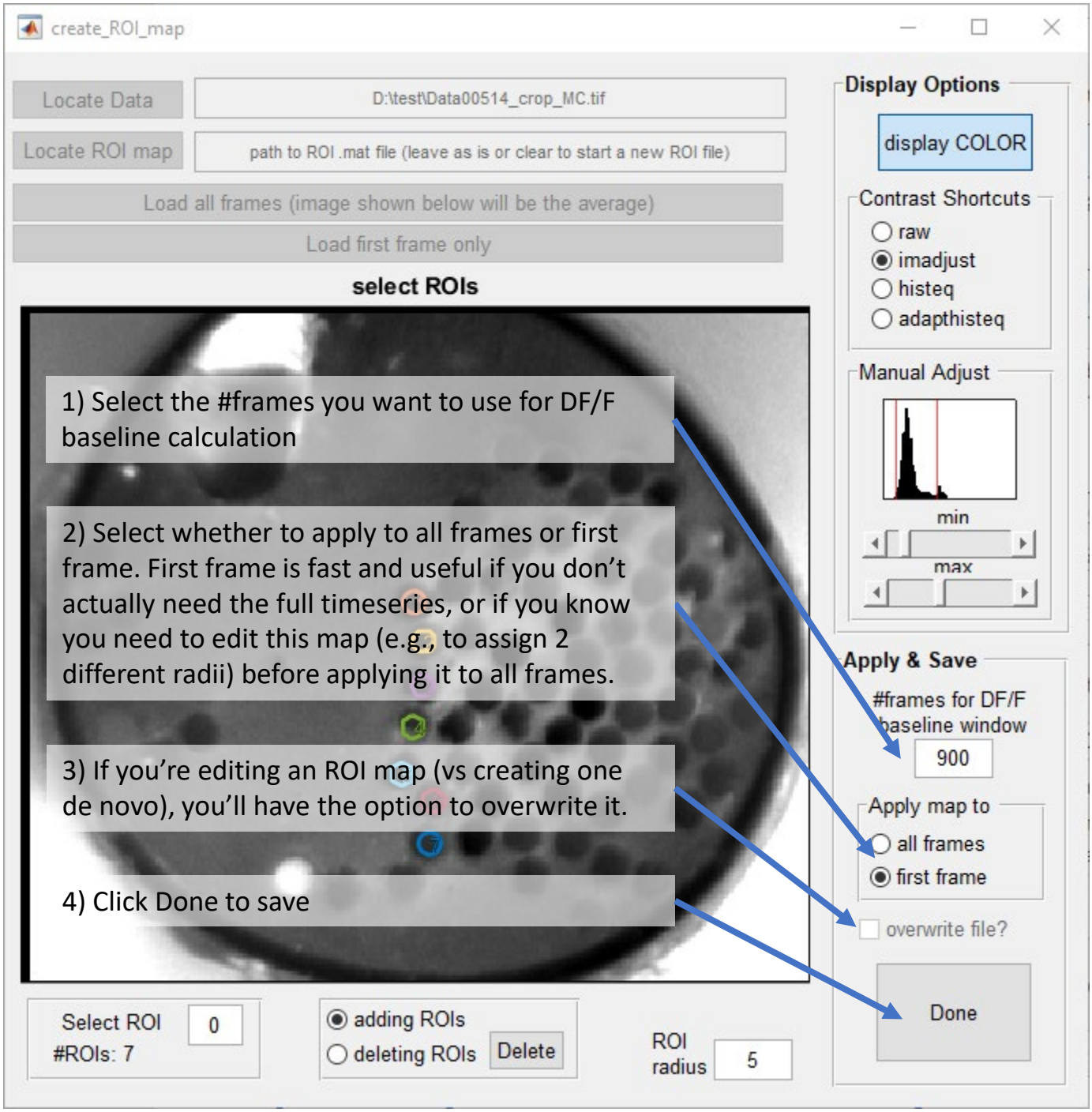


1) Change to deleting mode

2) Click the ROI you want to delete or enter its # in the "Select ROI" window. It will turn red.

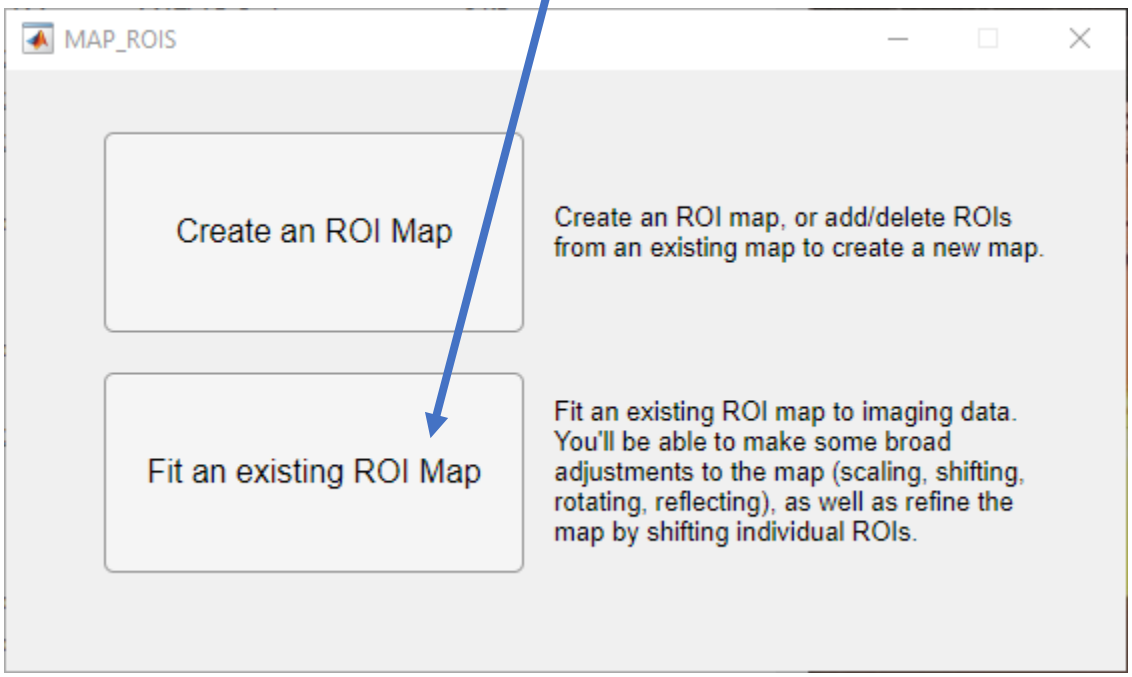
3) Click delete

Saving


create_ROI_map
Locate Data
D:\test\Data00514_crop_MC.tif
Locate ROI map
path to ROI .mat file (leave as is or clear to start a new ROI file)
Load all frames (image shown below will be the average)
Load first frame only
select ROIs
1) Select the #frames you want to use for DF/F baseline calculation
2) Select whether to apply to all frames or first frame. First frame is fast and useful if you don't actually need the full timeseries, or if you know you need to edit this map (e.g., to assign 2 different radii) before applying it to all frames.
3) If you're editing an ROI map (vs creating one de novo), you'll have the option to overwrite it.
4) Click Done to save
Display Options
display COLOR
Contrast Shortcuts
raw
imadjust
histeq
adapthisteq
Manual Adjust
min
max
Apply & Save
#frames for DF/F baseline window
900
Apply map to
all frames
first frame
overwrite file?
Done
Select ROI
0
#ROIs: 7
adding ROIs
deleting ROIs
Delete
ROI radius
5

Fit an existing ROI map to data

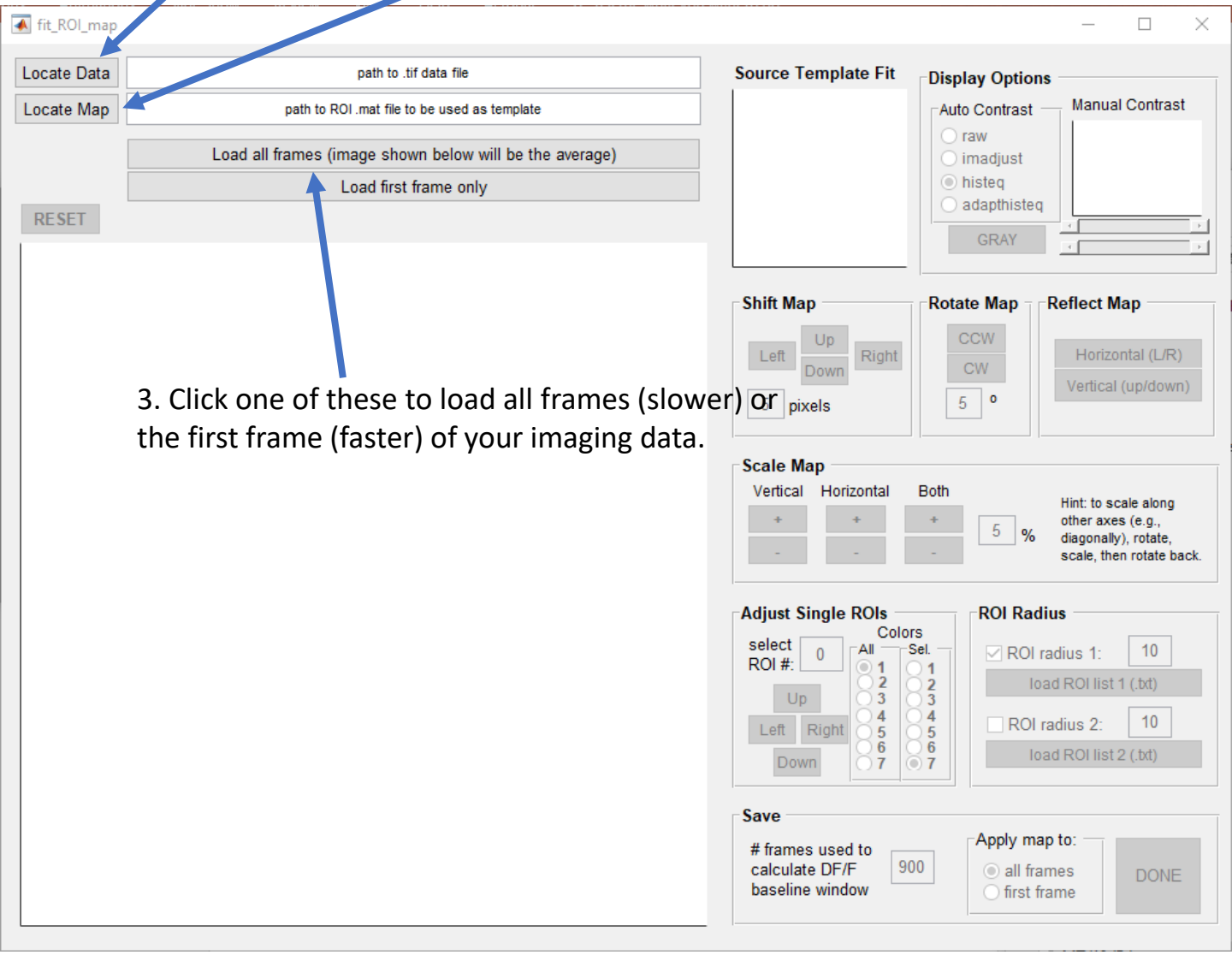
Because you want your ROI maps to be consistent from each recording to the next, use this one to fit an existing ROI map to data



Loading data

1. Click here to locate your .tif data, or enter it into the text field

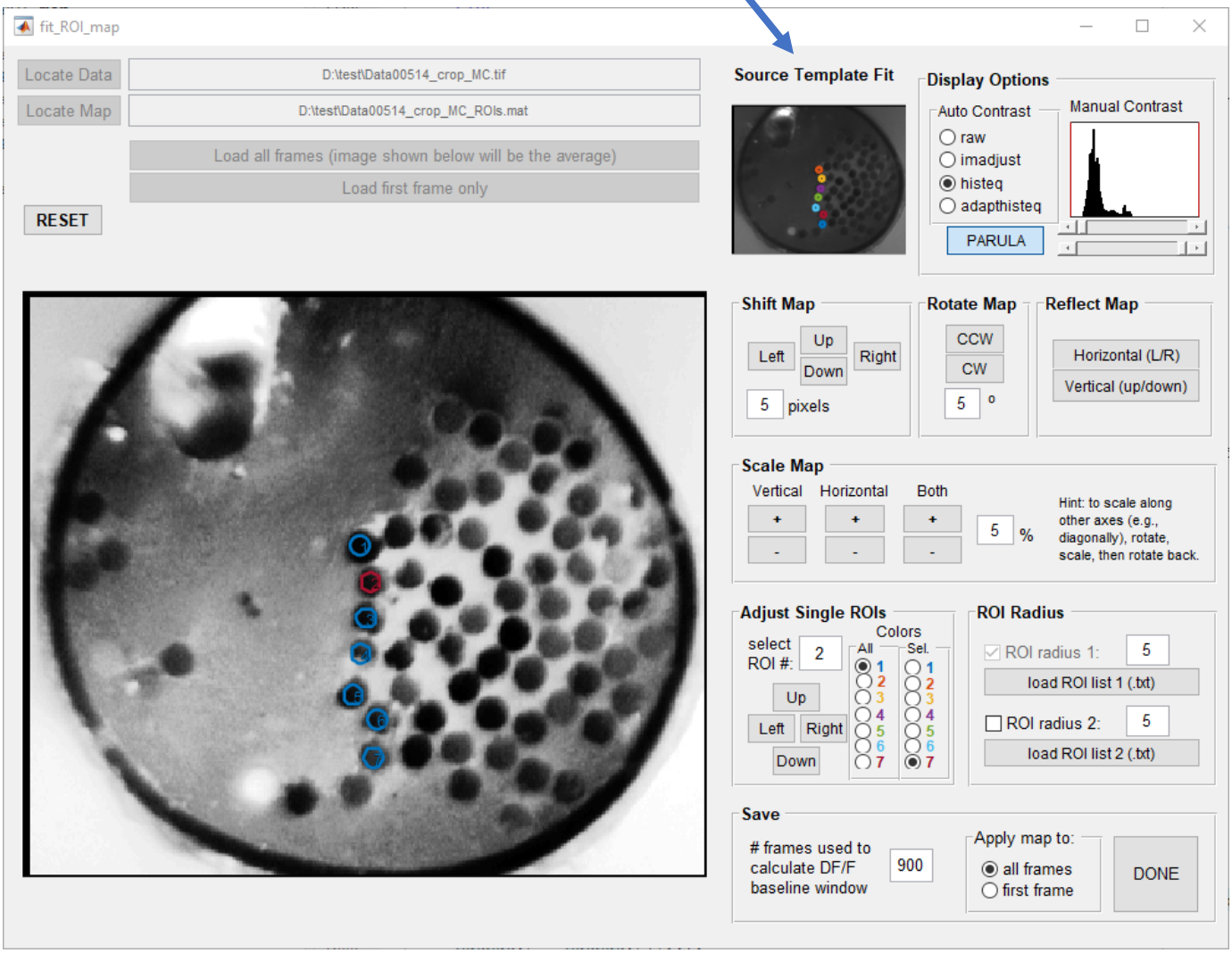
2. Click here to locate the ROI reference map, or enter the path into the text field.



3. Click one of these to load all frames (slower) or the first frame (faster) of your imaging data.

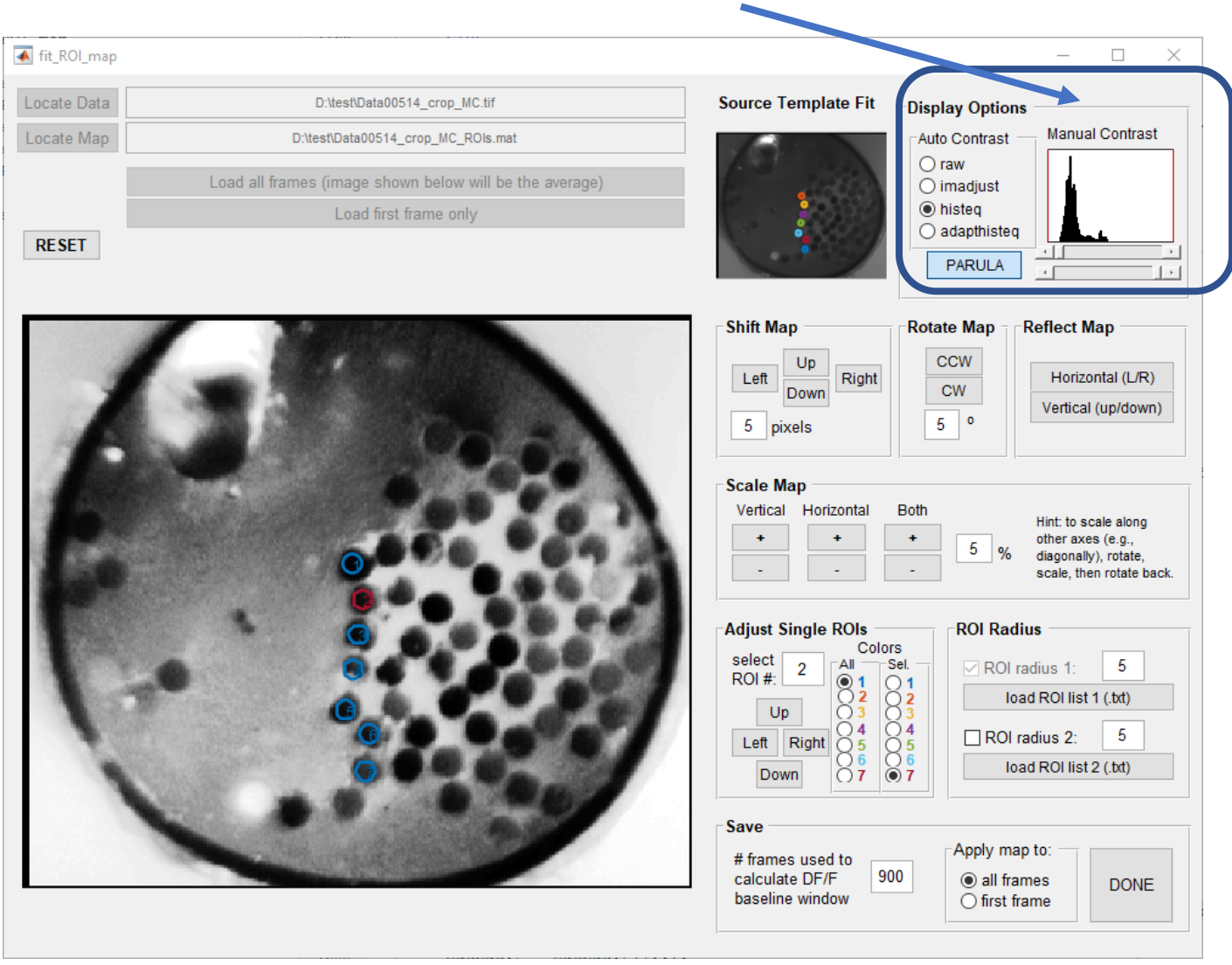
Reference

This image will show you the original map over the imaging data it was created for.



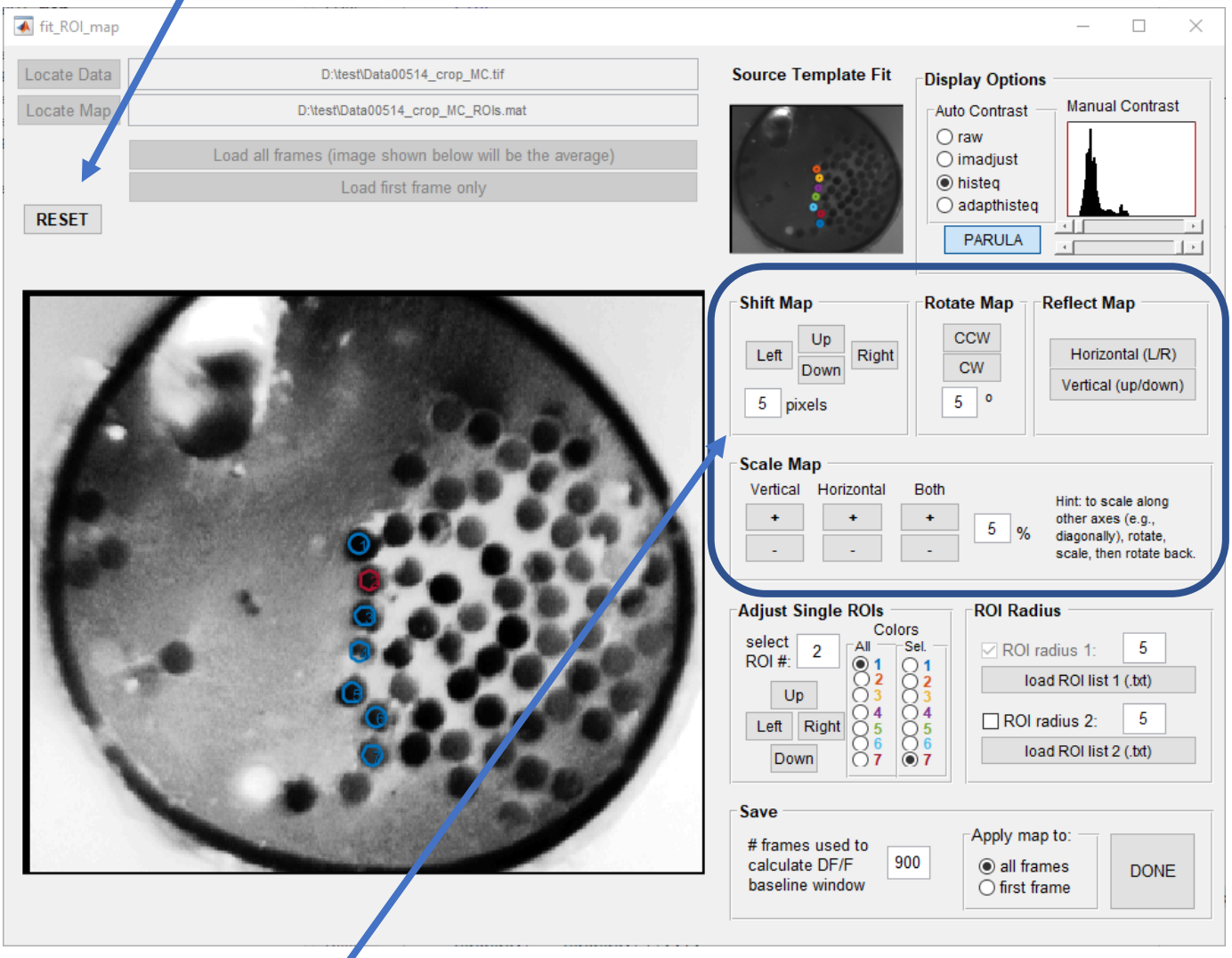
Adjusting the display

Use these controls to adjust the display. You can use one of the Auto Contrast options and/or set the contrast manually using the Manual Contrast sliders. You can also toggle between displaying the image in GRAY (grayscale) or PARULA (blue/yellow) colormap.



Adjusting the fit

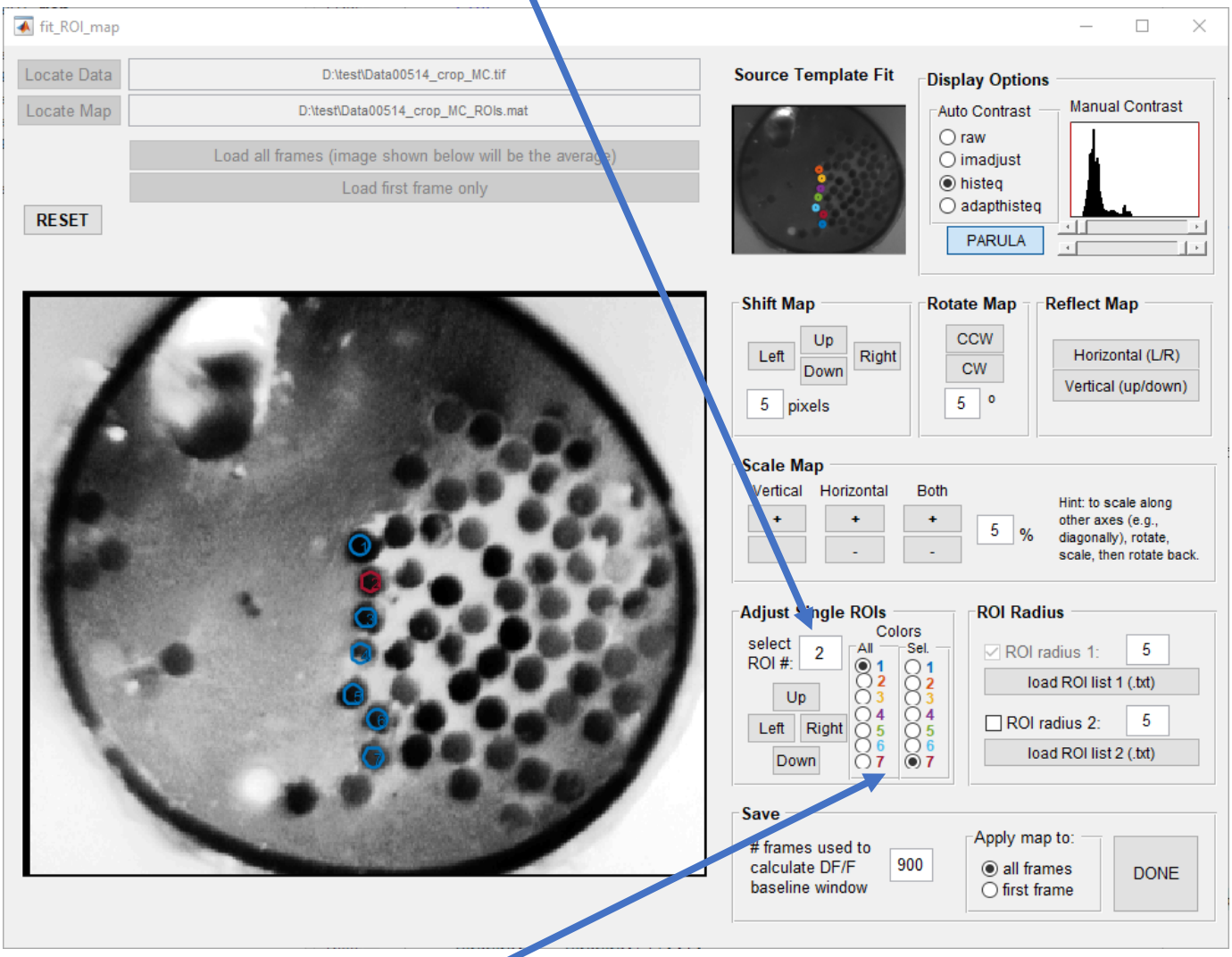
Clicking this will reset the map to its original position, orientation, etc



Use these controls to make large adjustments to the map. You can shift (translate) the map, rotate the map (CCW=counterclockwise, CW = clockwise), reflect the map, or scale the map. To scale (or reflect) along axes other than vertical or horizontal, rotate it first, scale/reflect, and then rotate back.

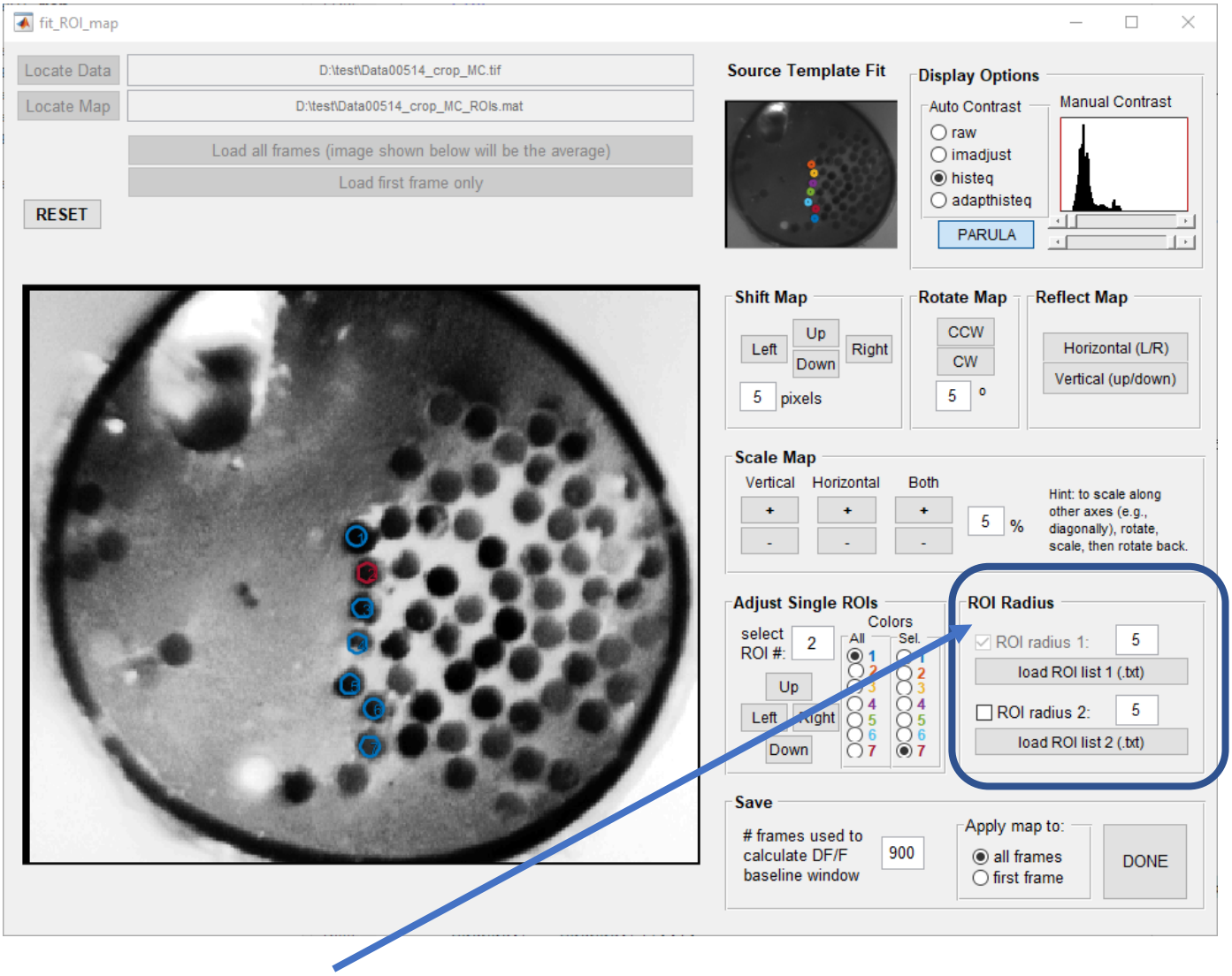
Shifting individual ROIs

You can select an ROI by clicking in the image or by entering it in the Select ROI# text field. The selected ROI will turn red. Use the buttons to shift the ROI up, down, left, or right. It will move 1 pixel at a time.



Right now the ROIs are displayed in blue (color #1), and the selected ROI is displayed in red (color #7). You can change that with these radio buttons.

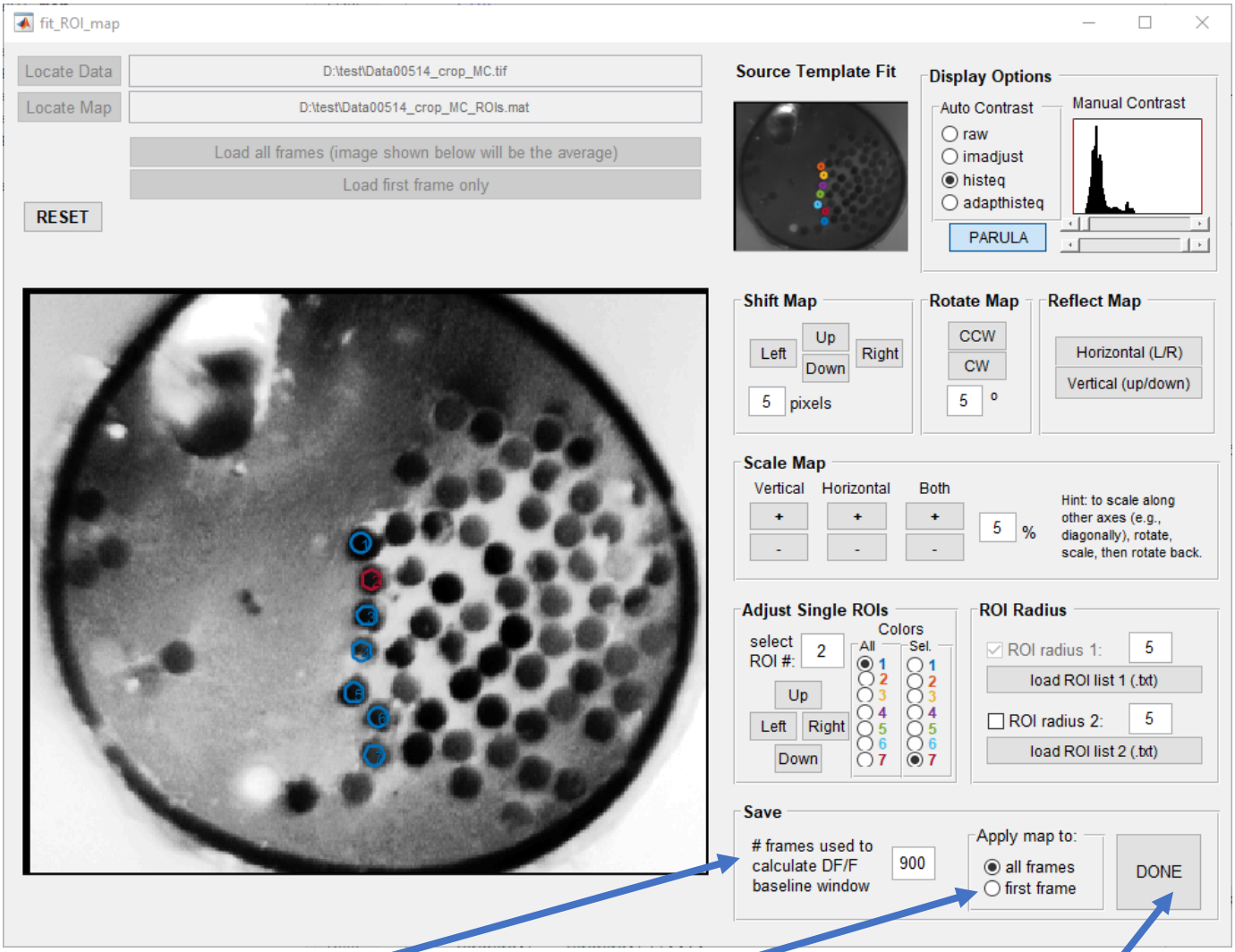
Assigning ROI radii



To assign the radius value, change the number in the textbox next to ROI radius 1 (currently it is set to 5). If you want to assign 2 different radii, check the checkbox next to ROI radius 2 and enter the second radius value in the box next to it. If you do need 2 different radii, you'll need a list of ROI #s (.txt file) for each radius value. Each text file should contains ROI #s, separated by new lines.

Saving

The datafile will be saved as *tif_file_name*_ROIs.mat – in this example, that would be Data00514_crop_MC_ROIs.mat. If that file already exists, it will append a timestamp to the filename so as not to overwrite the existing file.



1) Select the #frames you want to use for DF/F baseline calculation

2) Select whether to apply to all frames or first frame. First frame is fast and useful if you don't actually need the full timeseries, or if you know you need to edit this map again before applying it to all frames.

3) Click Done to save. If there's already an ROI file for this data, it'll save it with a date and time stamp.