

Grr User Guide

Getting Started

Once you have the program open, the first step is to load data. To do this go to File>Import .tif files. This will open a file explorer. *Note: You need to have already extracted your data from its original file format to be a stack of .tif files in a folder.* Navigate to the folder containing the .tif files from the data you wish to analyze. Select any one of the .tif files in this folder and select 'Open'.

Alternatively, go to File>Import .ser files. This will open a file explorer. Navigate to a .ser file containing image data and select 'Open'

The program will now create an image stack, look at the shifts between the images, calculate the outliers of the shifts between the images, disregard those shifts, use the remaining shifts to calculate an averaged image, and display this image, along with its Fourier transformation. You will also be able to see the shifts between each image in the x and y direction and see which data points were deemed outliers and not used in the generation of the averaged image.

In the display, you see, the screen is divided into 4 sections. In the top left is the final output: the average image and the Fourier transformation. Directly under this plot is a toolbar that allows users to zoom in on either one of the images displayed and move around it to explore. You may also use the Home button to revert to the original review. In the top right, individual slices from data set are displayed. To go through them, use the buttons on either side of this pane. On the right side are the graphs of the shifts between images.

On the bottom, we have the graphs of the shifts in the X and Y direction. These are the shifts between each image that occur due to the stage moving. On the left is the shifts without a mask, and on the right is the shifts with the mask covering the outlying points. The image on the right may look different since it removes the extreme values from the data, so the colors may appear brighter in this image. To help compare the original and masked data set, you can hold down the mouse button on the left image without a mask and it will cause the mask to overlay on this image. If you hold down the mouse button on the right image that has a mask, you can see the version of the image without the extreme values removed.

Editing Output

If you are not satisfied with the output, there are some things you can control to fix it, most having to do with how the outliers in the image shifts are determined.

The first option is to change the range of images used in the stack of images. To change this, go to Edit > Change nz range. A popup will come up, allowing you to change the nz min value and max value. These values refer to the stack of images produced. If some of the images at the beginning or at the end of the stack are not good to include for whatever reason, they can be removed from the stack temporarily and the average image recalculated without using these images.

Another option is to change how the outliers are calculated. To access these controls, go to Edit > Change Outlier Method. There are two methods you can choose from. The first method the outlier's method uses is a polynomial fit. In this method, a multidimensional polynomial is fit to the outlier shifts since in most cases there appears to be a pattern in these shifts due to the moving stage. After this is found, the program then finds the z-value for each point in the data set compared to the expected value found from the polynomial. If the z-score is too high, the point is considered an outlier and rejected. If you wish to use this method, select the Use Polynomial Fit radio button at the top of the popup. This will then allow you to edit the order of the polynomial used to fit the data and the z-score that each data point must fall under. Note: It is not recommended to use large order polynomials (greater than 4) since it will take a large amount of time to calculate and may not be accurate.

The other method goes through each point and compares it to its direct neighbors. If the difference in the value between the given point and its neighbors, then the program considers it to be a jump. If there are multiple jumps between one point and its neighbors, it is considered an outlier and removed. If you wish to use this method, you may select the radio button 'Use Nearest Neighbors' at the top of the popup. This will then allow you to change the max jump – the difference between the point examined and its neighbor before it is considered an outlier.

Lastly, you can also create a custom mask. This can be found by going to Edit>Custom Mask. A popup will appear showing the shifts in the X and Y direction with the mask overlaid. You can click on any point in either of the two plots to toggle the mask at that point on or off. Note that since the mask on both plots is the same and is diagonally symmetrical, up to 4 points may change with one single click.

After doing any of these edits, make sure to hit the save button in the popup menu. After selecting the Save button, the popup menu will automatically close and the program will recalculate the outliers and then the new resulting image with the new parameters inputted.

Saving Output

After going through all the calculations for, you naturally will want to export your results to another format to be saved. You can save your averaged image as an image in .tif or another format, a PDF report of your results, or you can get the data from the shifts in a .txt file.

There are a couple different ways to save the averaged image. You can go to File > Save Menu and a file selector will come up for you to select the location of the file and name it. Another way of doing this is selecting the save icon at the very right side of the toolbar. Using this method will allow you to save as an image with a variety of file types.

If you wish to get a PDF report of your results, which includes a graph of the shifts, both with and without the mask and shows the averaged image along with the Fourier transformation. To do this, go to File > Generate Report. This will open a file selector for you to select the destination of the PDF to be saved.

If you wish to export the shifts between the image in the X and Y direction, you can go to File > Export Shift Data and select the location and file name for the shift data to be saved to. This will create a text file that could be imported into another data editing program, using spaces as the delimiting character.