# FiberO Manual

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### Requirements

### Operating system

• Windows 10 or later (recommended).

When running the software on MacOS, an error related to saving results may occur. Therefore, it is recommended to use Windows systems for optimal performance.

#### MATLAB version

MATLAB 2020 or later (recommended).

#### Additional MATLAB toolbox

MATLAB image processing toolbox is required.

# Installation guide

First, download the FiberO installation file from the original source on GitHub. Once downloaded, it will be saved in your device as "FiberO.mlappinstall" by default. To install it, simply double-click on the file. This will open a MATLAB installation window, as shown in Figure 1. Click on "Install" and the app should be added to the "My Apps" section in the "Apps" tab.



Figure 1. Installation window.

### Features and functionality

When the application is launched, from the "Apps" tab, the window depicted in Figure 2a will appear, showing basic information about the software. Selecting the "2D analysis" button will open the analysis window.

The analysis window is initially divided into inputs and outputs, both displayed in the same window for intuitive and efficient use of the software. Additionally, to assist the user in navigating the analysis process, the status of the analysis is shown in the top left corner of the window (see green square in Fig. 2.B). This provides real-time feedback and guidance to the user as they interact with the interface and perform the analysis.

#### Selection of image

To begin the analysis, the user needs to select the desired image by clicking on the blue icon in the top left part of the window, see red arrow in Figure 2b. When the image is uploaded, provisional results for the first two steps are shown. The first result shows the area of the detected tissue while the second result shows the detected collagen fiber area.

**Note:** the dark areas that are not initially considered because of their low brightness, will be considered for the calculation of the area of the tissue if they are surrounded by detected tissue and the image will be updated after during the analysis of the image. Therefore, canals or pores in the tissue will also be considered.

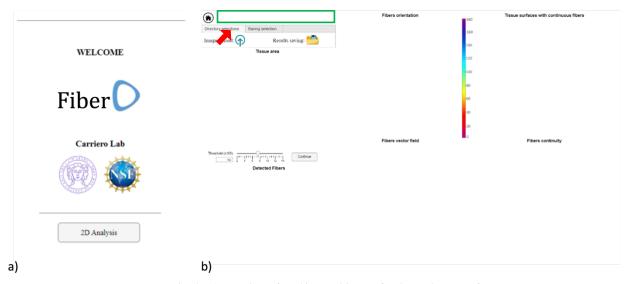


Figure 2. a) FiberO initial window. b) Initial layout for the analysis window.

# Defining a threshold

For the second image, if the user wants to adjust the threshold that defines the collagen fibers, a slider and label are available for quick parameter modification (Fig. 3). This value is saved to maintain consistency for subsequent analysis.

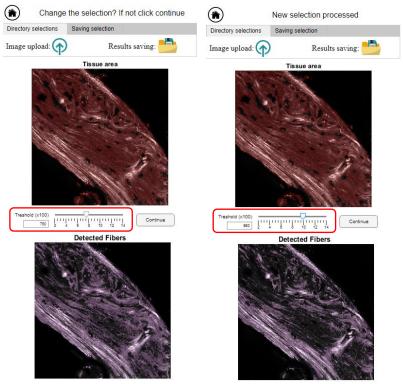


Figure 3. Differences of collagen fiber detection when the threshold is changed.

### Manual removal of regions

After adapting the threshold, the user confirms the value by clicking the "continue" button, which will open the next window for artifacts removal. If any region of the image is identified by the user as artifact and this is selected for removal, the previous results will be updated accordingly. The removal of the artifacts will be done by a manual selection of different points that will define the region of interest to be erased from the tissue. These points will be selected with the left-click of the mouse and to finally close the region a double-click or selecting again the first point can be used (Fig. 4). When closed, the artifact area from the image will be immediately removed by turning black. If the selection of more regions is required, the user is able to left click on the image and start the selection of the new region. To end this process and run the analysis of the image the "enter" key should be pressed.

**Note:** all the points defined by the user can be modified when grabbing them with a right-click. To remove the pixels that are close to the edge of an image, is recommended to define a point close to the edge and grab it to the outside of the image.

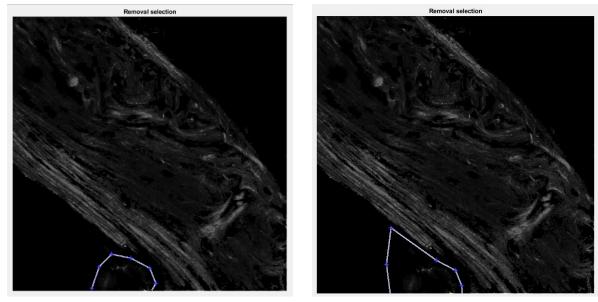


Figure 4. Removal of artifacts in the image by editing the position of the previously defined points.

#### Plotting the results

The next four plots in Fig. 5 show the results for fiber orientation, the fibers vector field, the surfaces with continuous collagen fibers, and finally, the collagen fiber continuity that contributes to the overall alignment of the tissue. These visual representations provide valuable insights into the orientation and organization of collagen fibers in the image.

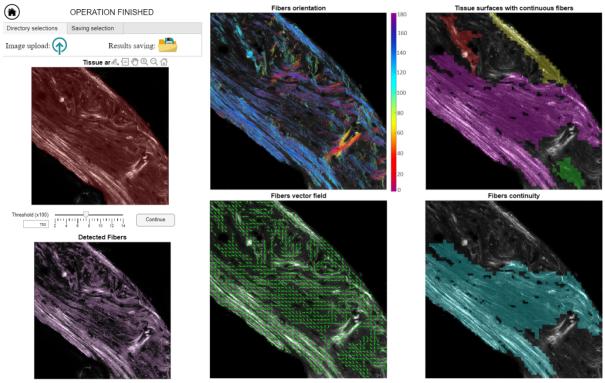


Figure 5. Final layout of the analysis window after the processing operations are done.

# Saving the results

Once all the results are displayed, the user has the option to save them (Fig. 6) in the selected directory on their computer together with the original image and the ".txt" file for the numerical results by clicking on the folder image at the top left region of the window. Additionally, if the user wishes to save storage space, they can choose which specific images to be saved by deselecting the checkboxes at the "Saving selection" tab. This allows for flexibility in saving only the necessary information for the user.



# Troubleshooting

If an error occurs, ensure that MATLAB is running so that error messages can be displayed in the command window, indicating the issue. Review the error message to identify the cause of the problem.