ctFIRE V1.0 Users Manual

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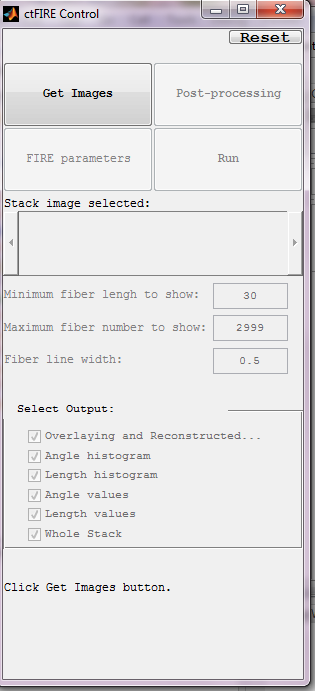
# Introduction

The purpose of this standalone MATLAB package is to allow users to automatically extract and quantify the alignment of collagen fibers in an image. The program reads in image files, extract the individual collagen fibers via ctFIRE ( curvelet transform plus FIRE algorithm) , which is an approach combining the advantage of the fast discrete curvelet transform (<http://curvelet.org/>) for denoising the image and enhancing the fiber edge features and the advantage of FIRE algorithm ([1]) for extracting individual fiber, and returns the segmented fibers along with descriptive statistics, such as fiber angle and length histograms as well as other optional outputs. The output may be displayed on the screen and/or written to .xlsx files.

In the ctFIRE, the user can choose to run FIRE, ctFIRE or both of them. Parameters to run FIRE and ctFIRE have default values and are also adjustable via GUI . Optional outputs include: overlaid image of the segmented fibers on the original image and the CT reconstruction image, figure of fiber angle histogram, figure of fiber length histogram, fiber angle value and fiber length value. The detailed information about the parameters and output of ctFIRE is automatically saved for a later post-processing.

# GUI control panel

The GUI in ctFIRE V1.0 is modular, so that the main user interface is in a separate window from the outputs. This allows for the users to resize the output windows to their preferred size. The main user interface window is shown below.



*where:*

**Reset**: rerun the GUI

**Get files**: open an image, a stack or a folder. Currently the processing of a stack and folder is in development.

**FIRE parameters**: set FIRE parameters.

**Run**: select the output folder, run mode (FIRE, ctFIRE or both) and additional ctFIRE parameters if ctFIRE is chosen to run.

**Post-processing**: after Run, change the output setting to get the desirable output

**Stack image selected**: when open a stack, move the slide bar to select the slices to run and show the current slice. Currently stack image can be shown but not to be analyzed.

Output of the extracted fibers

**Minimum fiber length to show**: only extract fibers which are longer than this value, unit is pixel, default value is 30.

**Maximum fiber number to show**: maximum fiber number, if there are too many extracted fibers, probably need to reset the running parameters, default value is 2999

**Fiber line width**: control the line width of the extracted fibers on the overlaid image, default value is 0.5

Select Output: select the desirable output, default is output all

**Overlaying and reconstructed**: overlaid image of the segmented fibers on the original image and the CT reconstruction image,

**Angle histogram**: figure of fiber angle histogram

**Length histogram**: figure of fiber length histogram

**Angle values**: save fiber angle value into an Excel file

**Length values**: save fiber length value into an Excel file

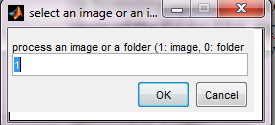
**Whole Stack**: analyze all the slices in a stack, this function is in development

**Bottom information label**: indicate what to do or the program status

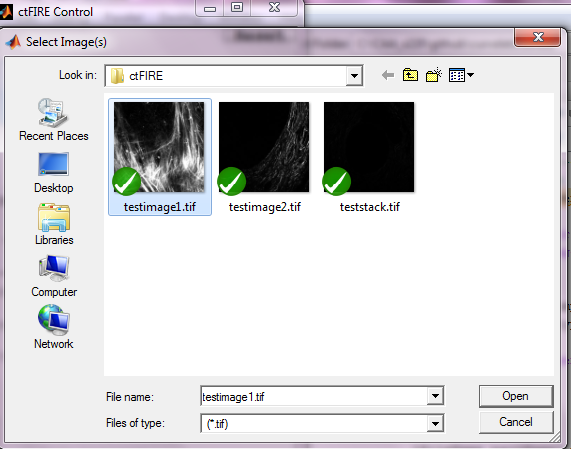
# An example

## step 1: open images

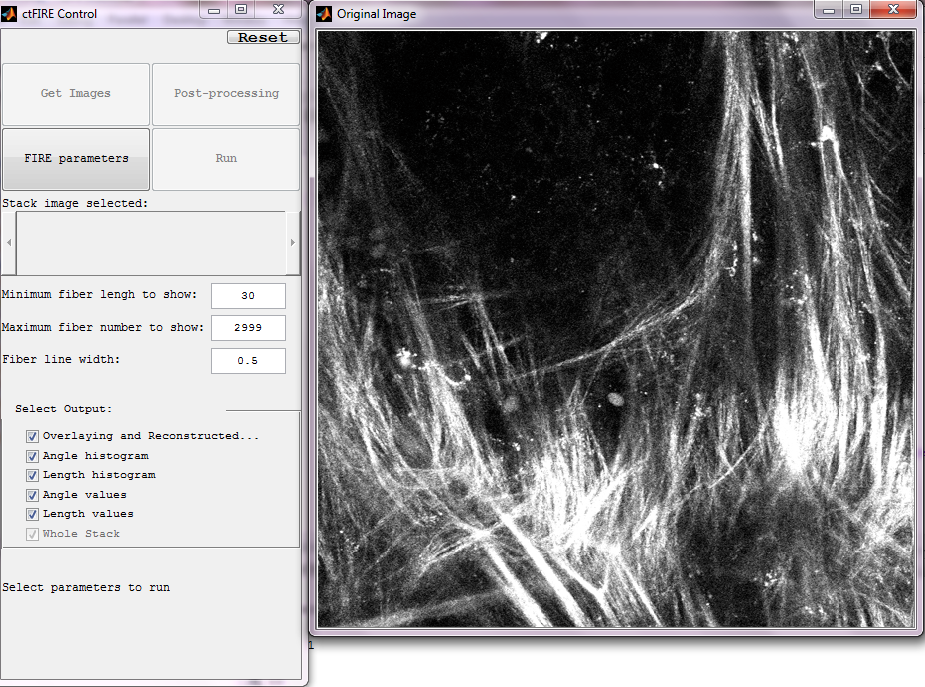
click Get images , then get the



If analyze an image or image stack , input 1, if analyze the images in a specified folder, input 0. Click OK to choose the "testimage1.tif" as in the follows



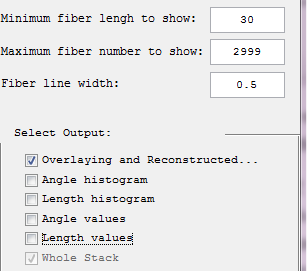
click open, then goes to



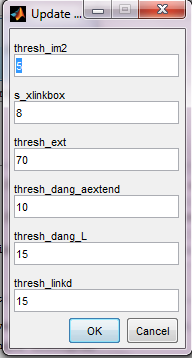
The selected image is shown and the FIRE parameters button as well as other input and output settings are enabled.

## step 2: set input and output parameters:

let's uncheck the angle and length output as follows and only output the overlaid and reconstructed image, while keep the minimum fiber length, maximum fiber number and fiber line width as default value as in follows:



Then set FIRE parameters by click the button of "FIRE parameters", going to the default parameters:



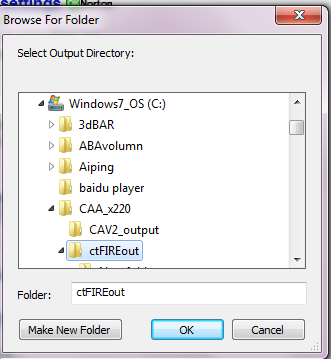
The description of the six parameters is listed below

|  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| thresh\_im2 | thresh\_im2 is for a hard value, main adjustable parameters | | |  | | | |  | |  | |  | |  | |
| s\_xlinkbox | radius of box in which to check to make sure xlink is a local max of the distance function | | | | | | | | |  | |  | |  | |
| thresh\_ext | angle similarity required for a fiber to extend to the next point | | | | | | |  | |  | |  | |  | |
| thresh\_dang\_aextend | maximum dangle angler difference at cross-link | | | | |  | |  | |  | |  | |  | |
| thresh\_dang\_L | dangler length threshold |  | | |  | |  | |  | |  | |  | |
| thresh\_linkd | distance for linknig same-oriented fibers | |  |  | |  | |  | |  | |  | |  | |

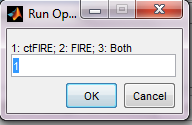
Change the thresh\_im2 to 30 and s\_xlinkbox to 5 , click ok. After setting the FIRE parameters , the Run button is enabled.

## step 3: Run

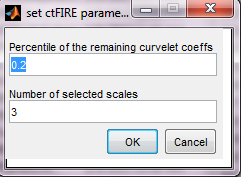
Clicking Run button shows "Select Output Directory"



Let's choose ctFIREout in this case. Then shows Run Options:



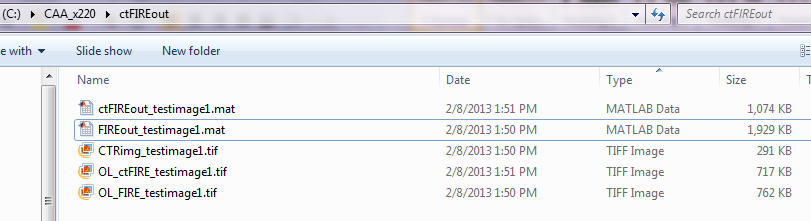
Input 1 to run ctFIRE, 2 to run FIRE, 3 to run both FIRE and ctFIRE. Default is run ctFIRE. Here Let's choose run option 3 to run both FIRE and ctFIRE. Then shows to set two ctFIRE parameters:



Here, the percentile of the remaining curvelet coeffs is a hard threshold applied to the curvelet coefficients in all of the scales. Number of the selected scales is to set the scales to reconstruct the image. Specifically, if the whole scales is N, 3 is the number of selected scales, then the actual scales used for reconstruction is scale N-1, N-2 and N-3. Let's use the default values and click OK to continue. Then the progress of the fiber extracting will be listing in the Command Window. In this example, the overlaid and reconstructed images will also show up.

## step 4: check the results:

In the output folder : C:\CAA\_x220\ctFIREout, 5 output files are shown:



*FIRE output:*

FIREout\_ image name.mat: the .mat file for the output by FIRE

OL\_FIRE\_image name.tif : the .tif overlaid image of FIRE

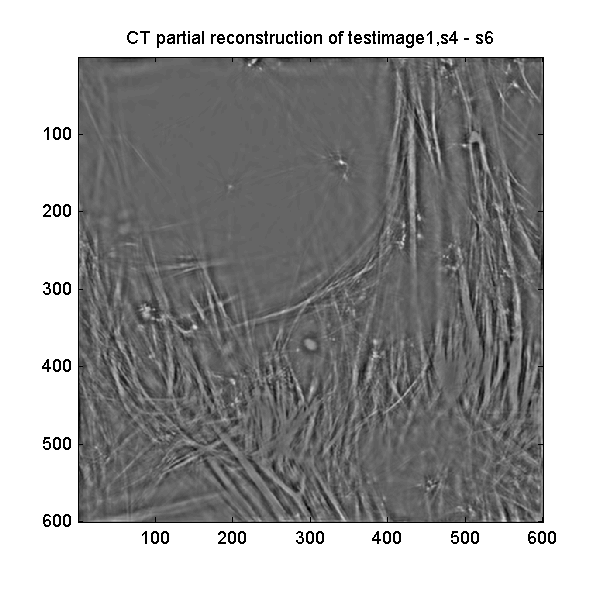
*ctFIRE output:*

ctFIREout\_ image name.mat: the .mat file for the output by ctFIRE

OL\_ctFIRE\_image name.tif : the .tif overlaid image of ctFIRE

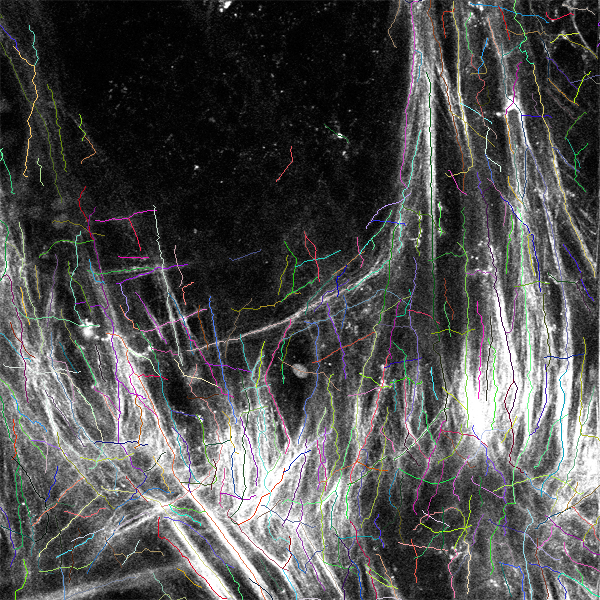
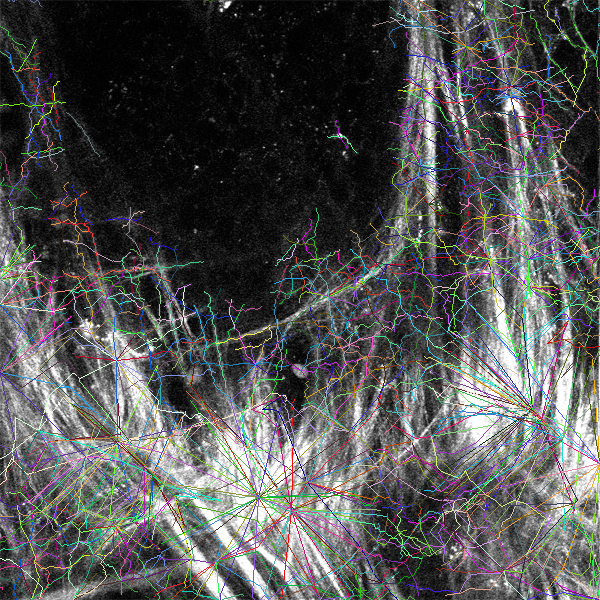
CTRimg\_image name.tif: the .tif CT reconstruction image

The three images are shown below:



CTRimg\_testimage1.tif

OL\_ctFIRE\_testimage1.tif

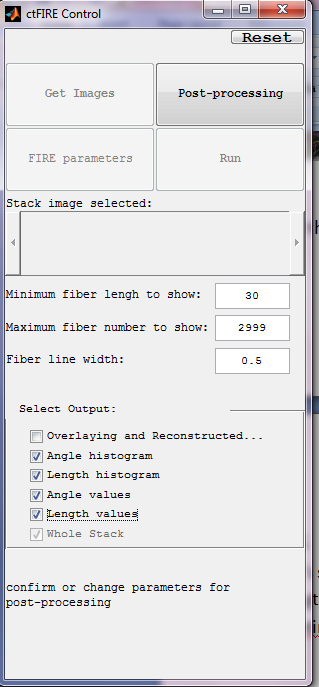


OL\_FIRE\_testimage1.tif

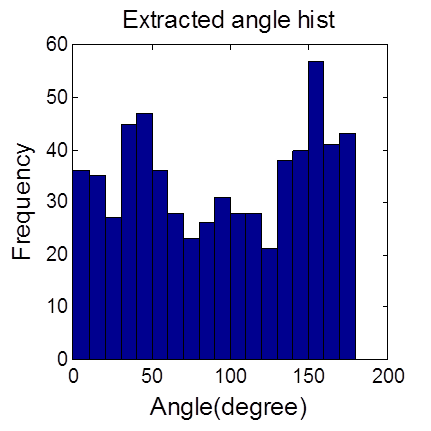
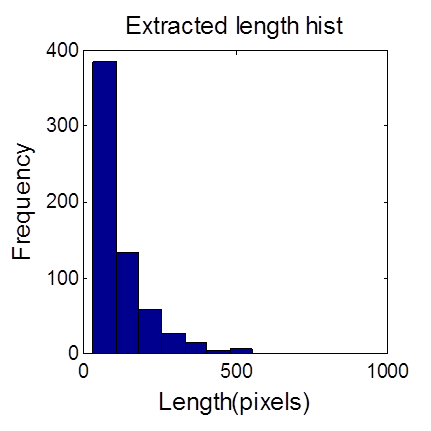
For this example, the results of ctFIRE looks much better than those of the FIRE.

## step 5: post-processing

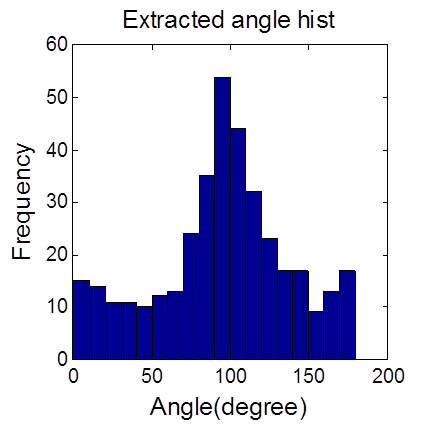
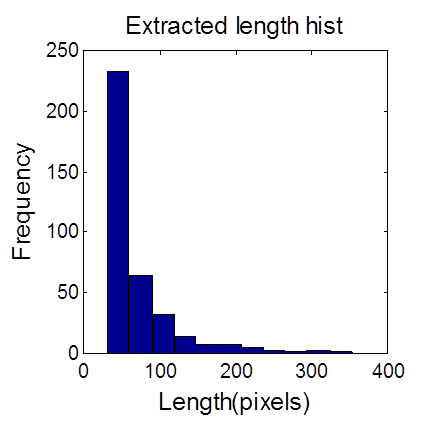
Let's suppose we are satisfied with the overlaid image, now we want to see the angle and length histograms and save the correspond values. In the control panel, the corresponding settings are:



Click Post-processing, then shows:

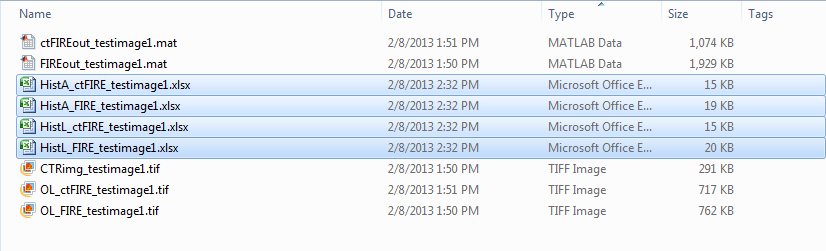


FIEE output



ctFIEE output

The output folder : C:\CAA\_x220\ctFIREout, shows 4 additional output files highlighted below which are the angle and length values of both FIRE and ctFIRE:



## step 6: Reset

Reset to process a new image or re-process the same image with different parameters.