LOCALIZE_FIBERS MATLAB App README

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Fiber location table

The purpose of this sequence of steps is to find the locations of your fibers, and match them to the grid. An output struct with crucial information will be generated, and has the following fields.

- info: the size, and location of your CT scan
- fiber_tops: intermediate information from the steps for localization of the fiber tops
- fiber_bottoms: intermediate information from the steps for localization of the fiber bottoms
- fiber_table_orig: same as below, with the information before manual editing
- **fiber_table**: this is the main output table. This will have a row for each fiber, and contains information about the coordinates for each fiber top and fiber bottom, as well as atlas labels

Before you begin

If you have a grid arrangement, and you want to map your recording locations to locations on the grid, you will want a matlab (.mat) file, which, when loaded, contains one field which provides the grid mapping of your fibers. There are 2 ways to do this:

1. One is a coordinate system. This is a matrix, where each row represents a fiber, and the columns represent row coordinates, column coordinates, and (optional) ID#. In this example to my right, column 1 contains the column# for each fiber. Column 2 contains the row#. Column 3 contains an ID# that I assigned ahead of time.

```
ans =

     5    10     1
     5    12     2
     6    11     3
     6    13     4
     4     9     5
     8    11     6
     6     9     7
     8     9     8
     7     8     9
     3     6    10
     4     3    11
     1     4    12
     4     7    13
     8    13    14
     7    10    15
     9    14    16
    10    13    17
     7    14    18
    11    14    19
    11    16    20
    13    18    21
    12    17    22
    10    15    23
    13    16    24
    14    19    25
    12    15    26
    11    12    27
    12    13    28
    10     9    29
    14    17    30
    14    13    31
    13    10    32
     7    12    33
    10    11    34
```

Before you begin (cont'd)

2. The other options is a matrix that represents the layout of the grid. The values in the matrix are the ID# (you can use 1s if you don't have ID#s), and the locations are the actual row/column location in the grid arrangement. In my example below, fiber#64 is in row 1, column 2. Fiber#10 is in row 7, column 3.

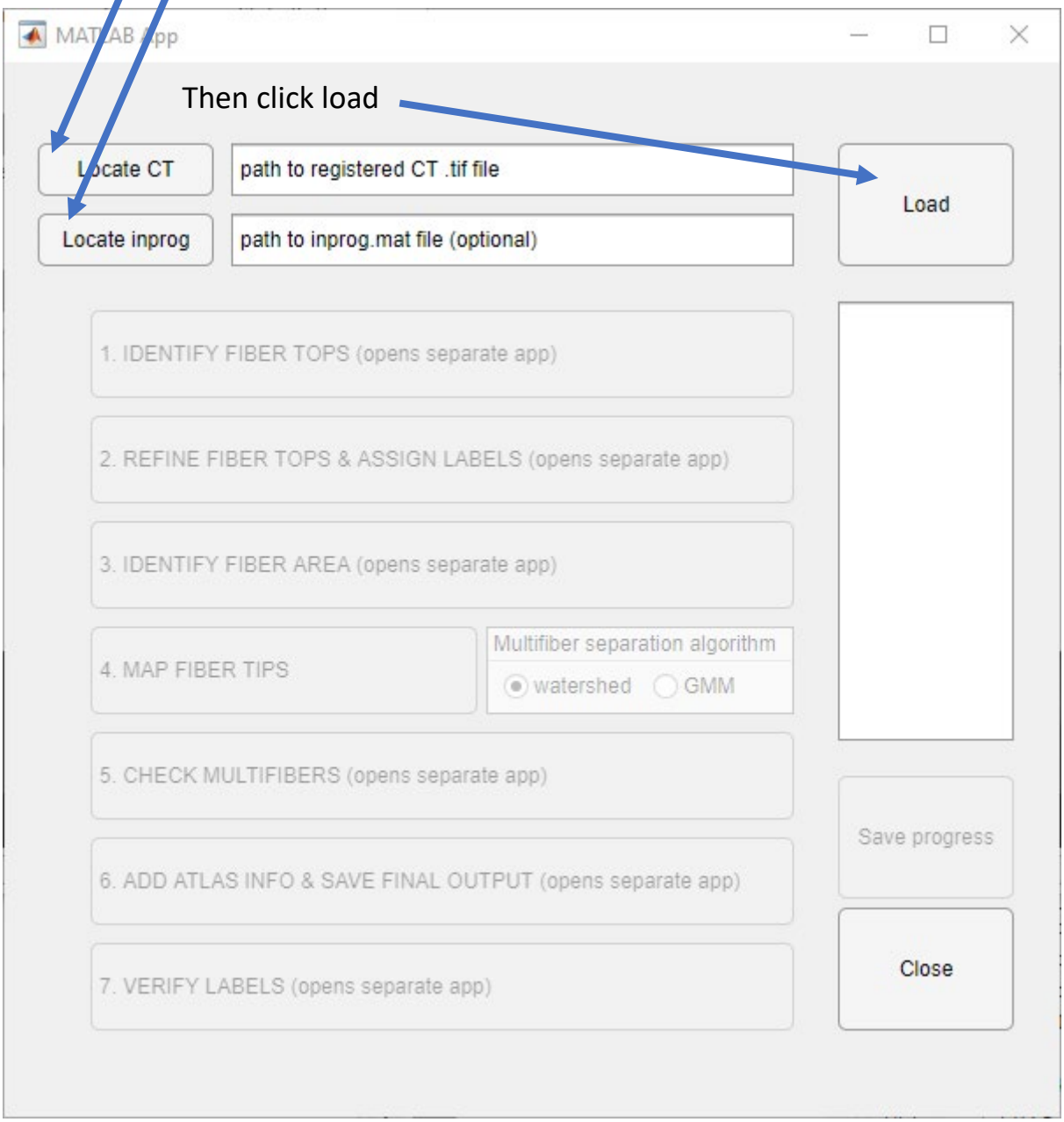
```
ans =

     0    64     0    69     0     0     0     0     0     0     0     0     0     0
    73     0    70     0    59     0     0     0     0     0     0     0     0     0
     0    38     0    11     0    65     0     0     0     0     0     0     0     0
    12     0    36     0    67     0    66     0    58     0     0     0     0     0
     0    39     0    37     0    68     0    63     0    60     0     0     0     0
     0     0    10     0    40     0    62     0    61     0    54     0     0     0
     0     0    42    13     0    57     0    56     0    48     0    55     0     0
     0     0    41     0    35     0     9     0    46     0    71     0    53     0
     0     0     0     5     0     7     0     8     0    29     0    51     0     0
     0     0     0     0     1     0    15     0    47     0    52     0    32     0
     0     0     0     0     0     3     0     6     0    34     0    50     0    49
     0     0     0     0     2     0    33     0    72     0    27     0    45     0
     0     0     0     0     0     4     0    14     0    17     0    28     0    31
     0     0     0     0     0     0    18     0    16     0    19     0    43     0
     0     0     0     0     0     0     0     0     0    23     0    26     0    44
     0     0     0     0     0     0     0     0     0     0    20     0    24     0
     0     0     0     0     0     0     0     0     0     0     0    22     0    30
     0     0     0     0     0     0     0     0     0     0     0     0    21     0
     0     0     0     0     0     0     0     0     0     0     0     0     0    25
```

Before you begin (cont’d)

Note: If you don’t have a grid arrangement, but you have some reference of what the grid tops should look like (a coordinate file like #1) or just an image you want to use, you can use that too.

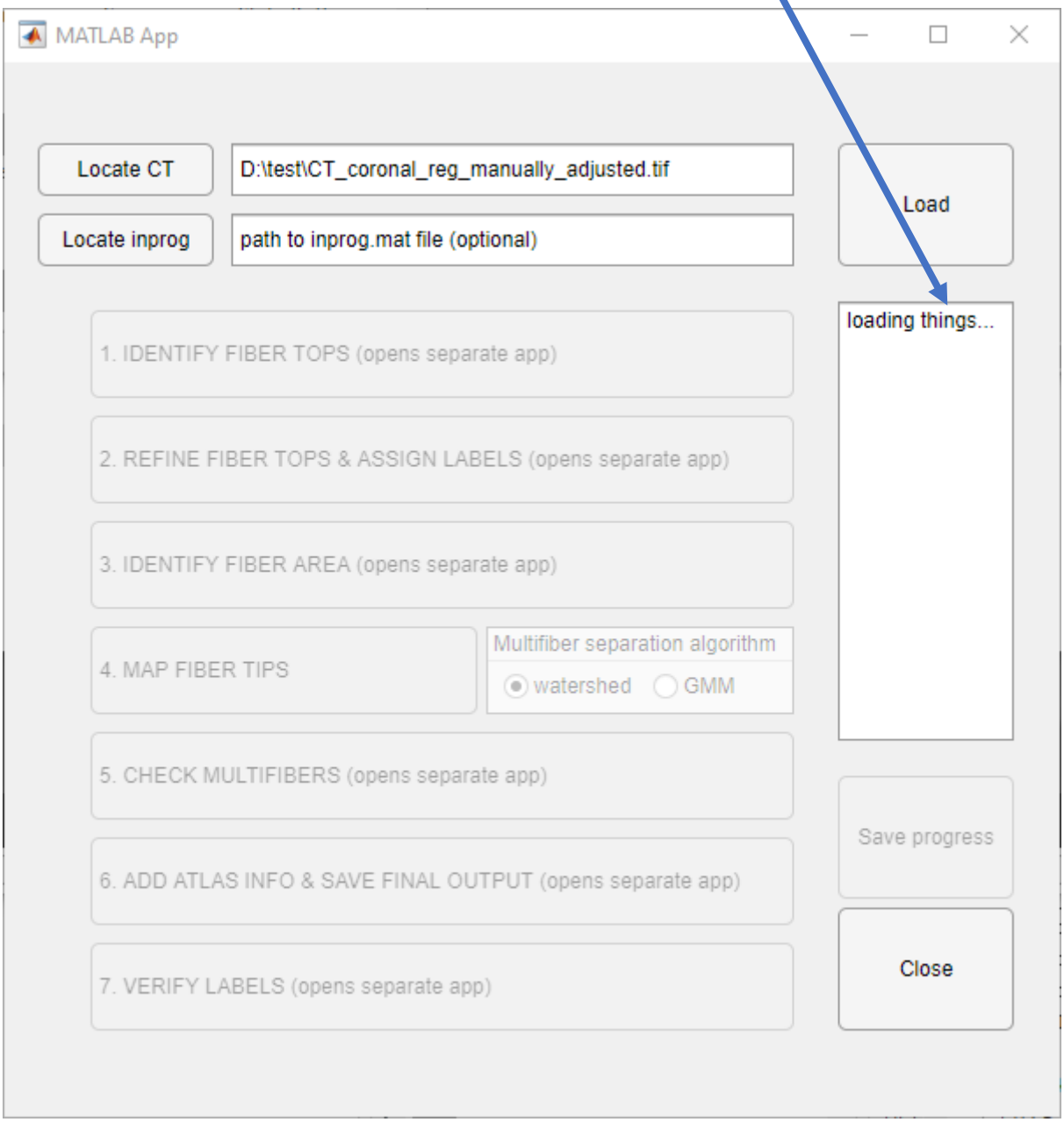
LOCALIZE_FIBERS

Click to locate your CT.
This button would be for to locate the in-progress localization file if you want to resume an unfinished localization.



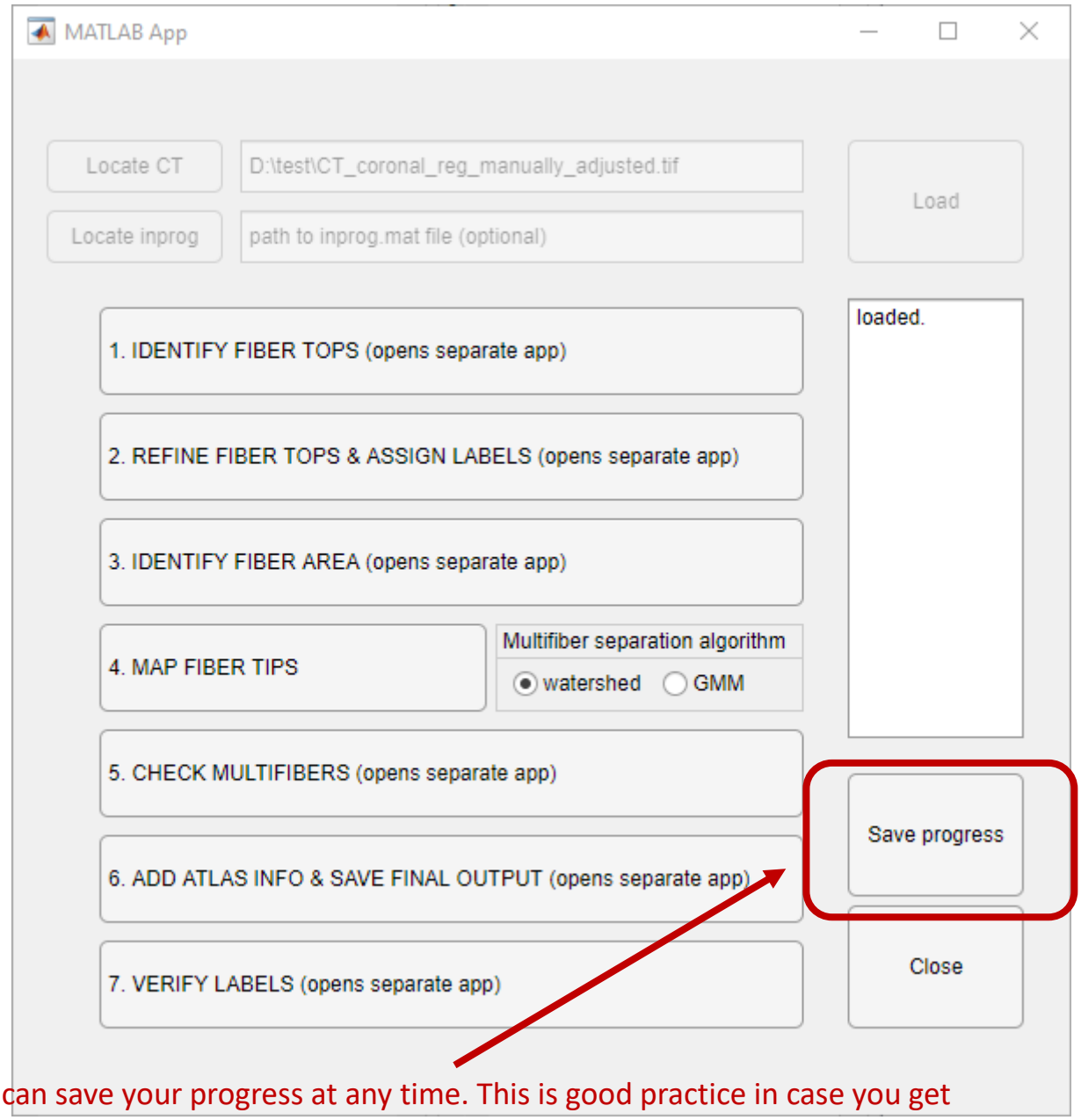
LOCALIZE_FIBERS

This text window will keep you updated.



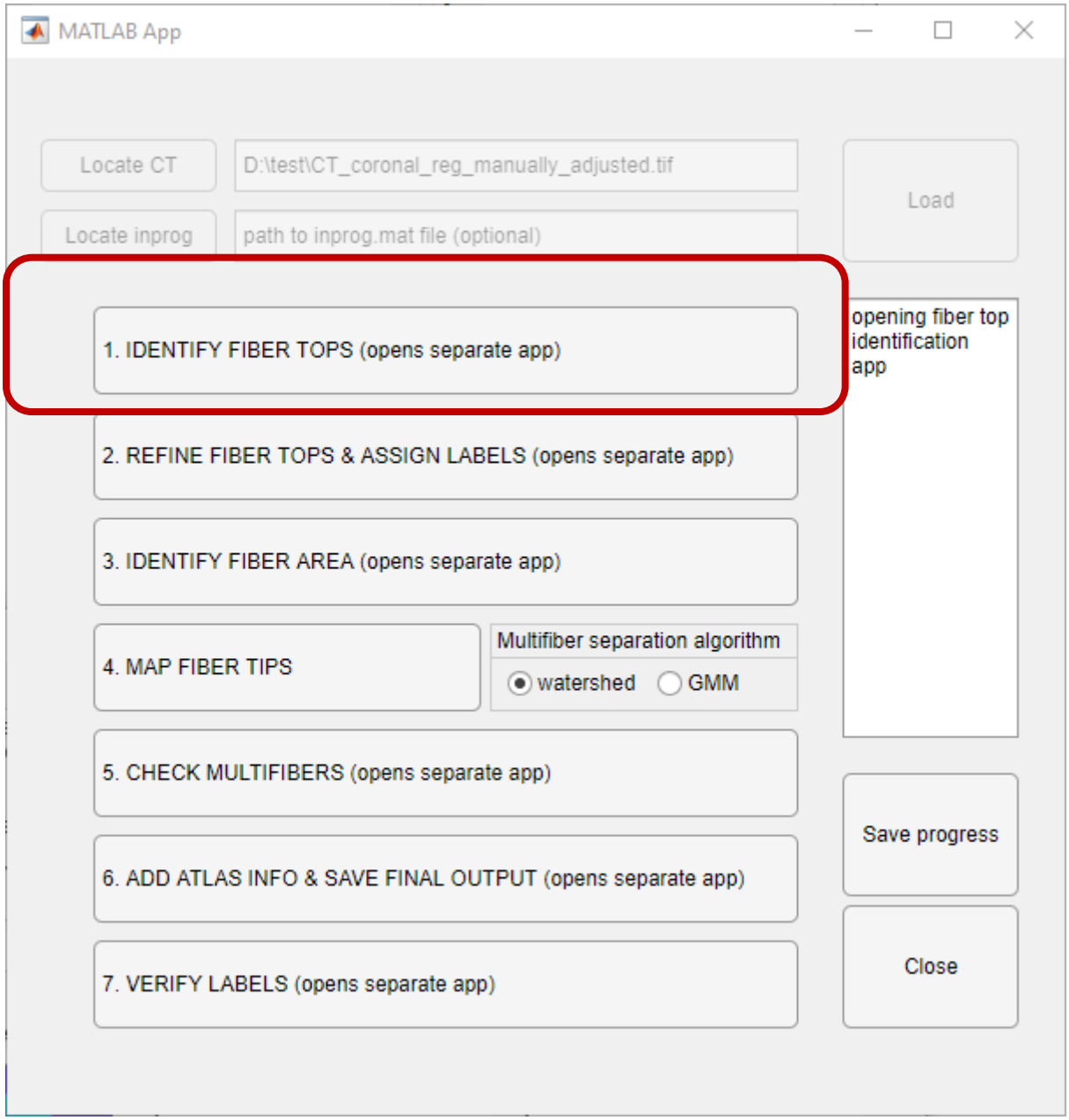
LOCALIZE_FIBERS

Now you'll go through the steps in order.



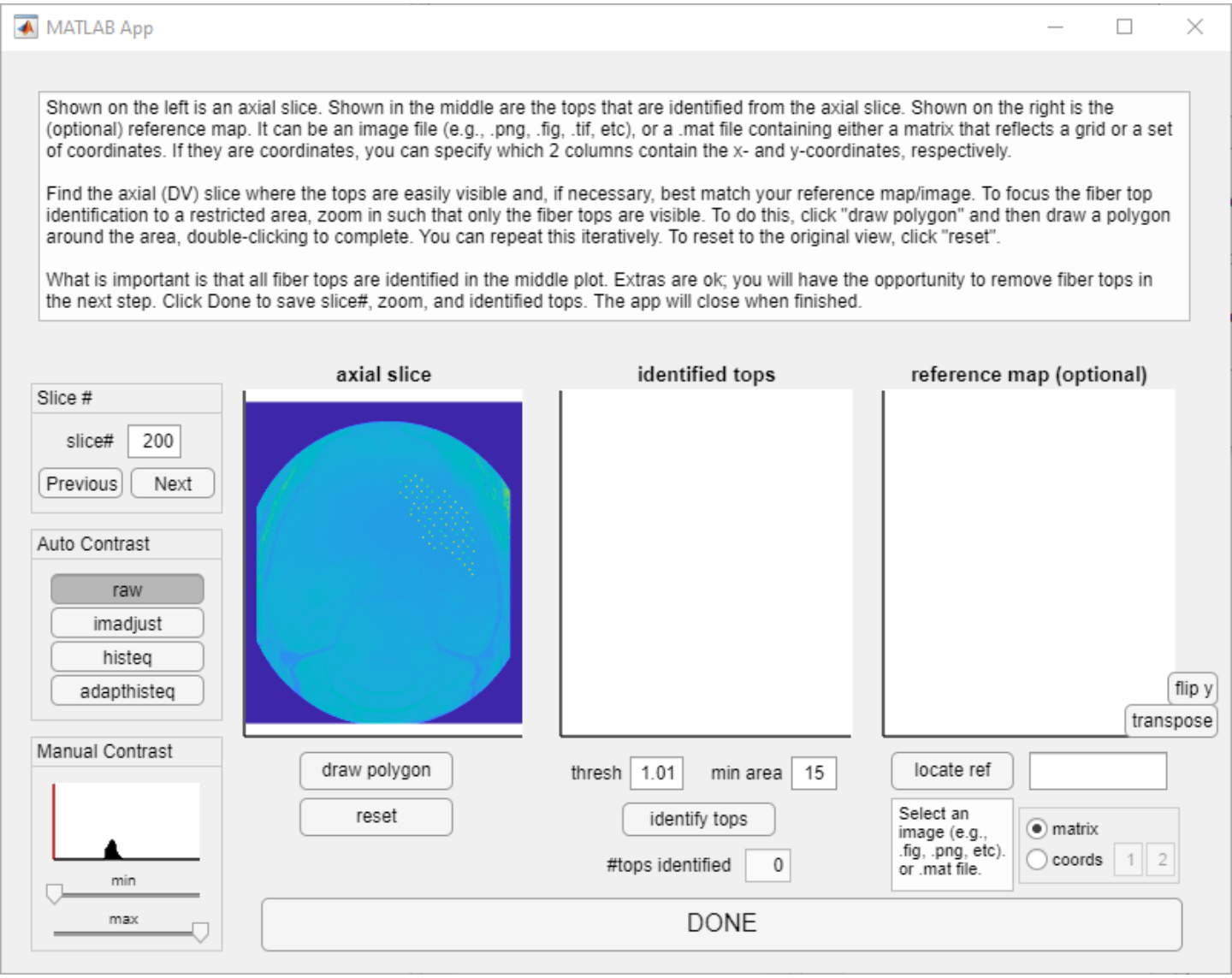
You can save your progress at any time. This is good practice in case you get interrupted, or you run into issues at a step. You'll be able to pick up where you left off. This will be name something like ___in_prog.mat, and after step 6, it will be named _____localized_fibers.mat

IDENTIFY FIBER TOPS

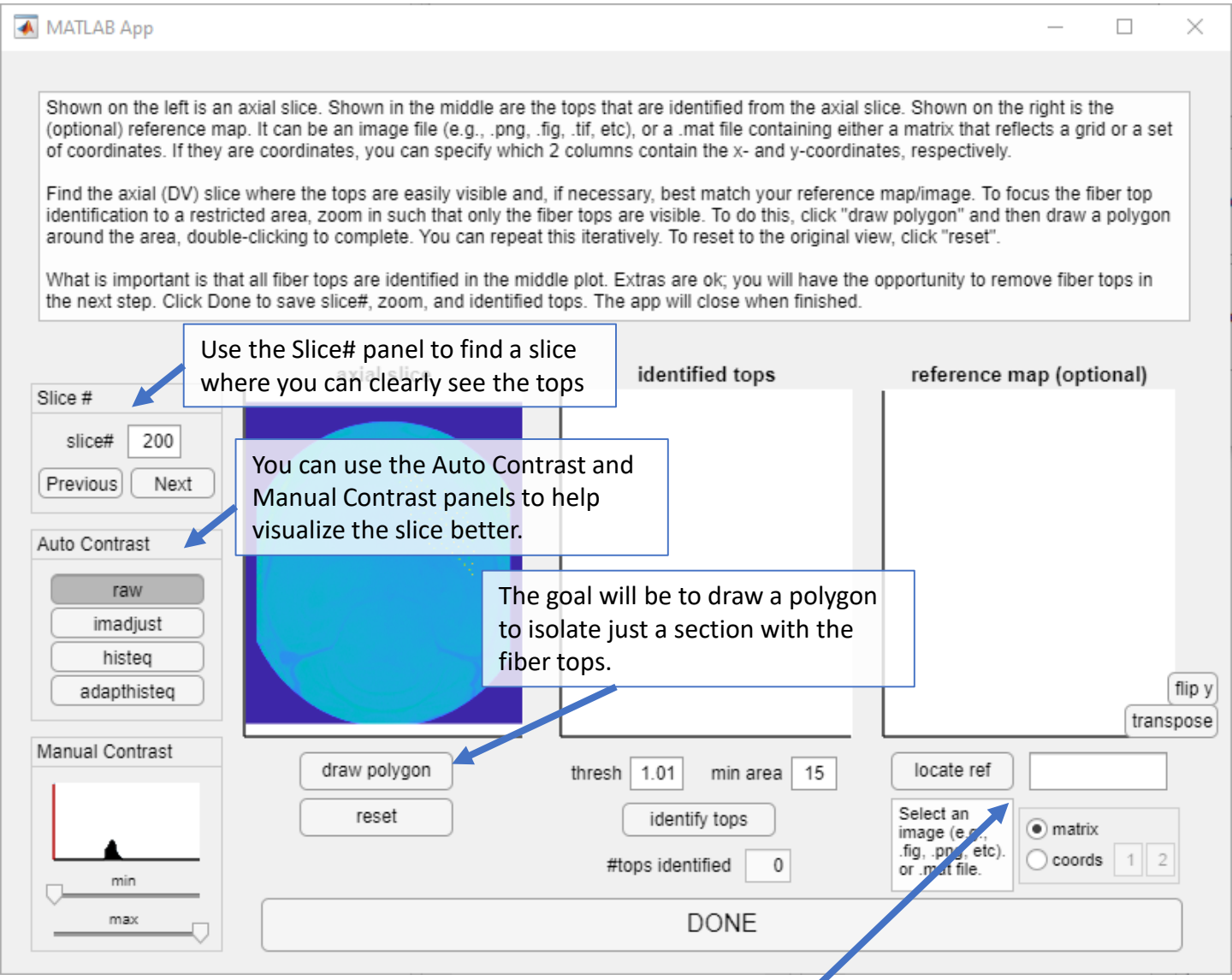


IDENTIFY FIBER TOPS

Eventually, we'll map each fiber top to each fiber bottom. The objective of this app is to locate your fiber tops.

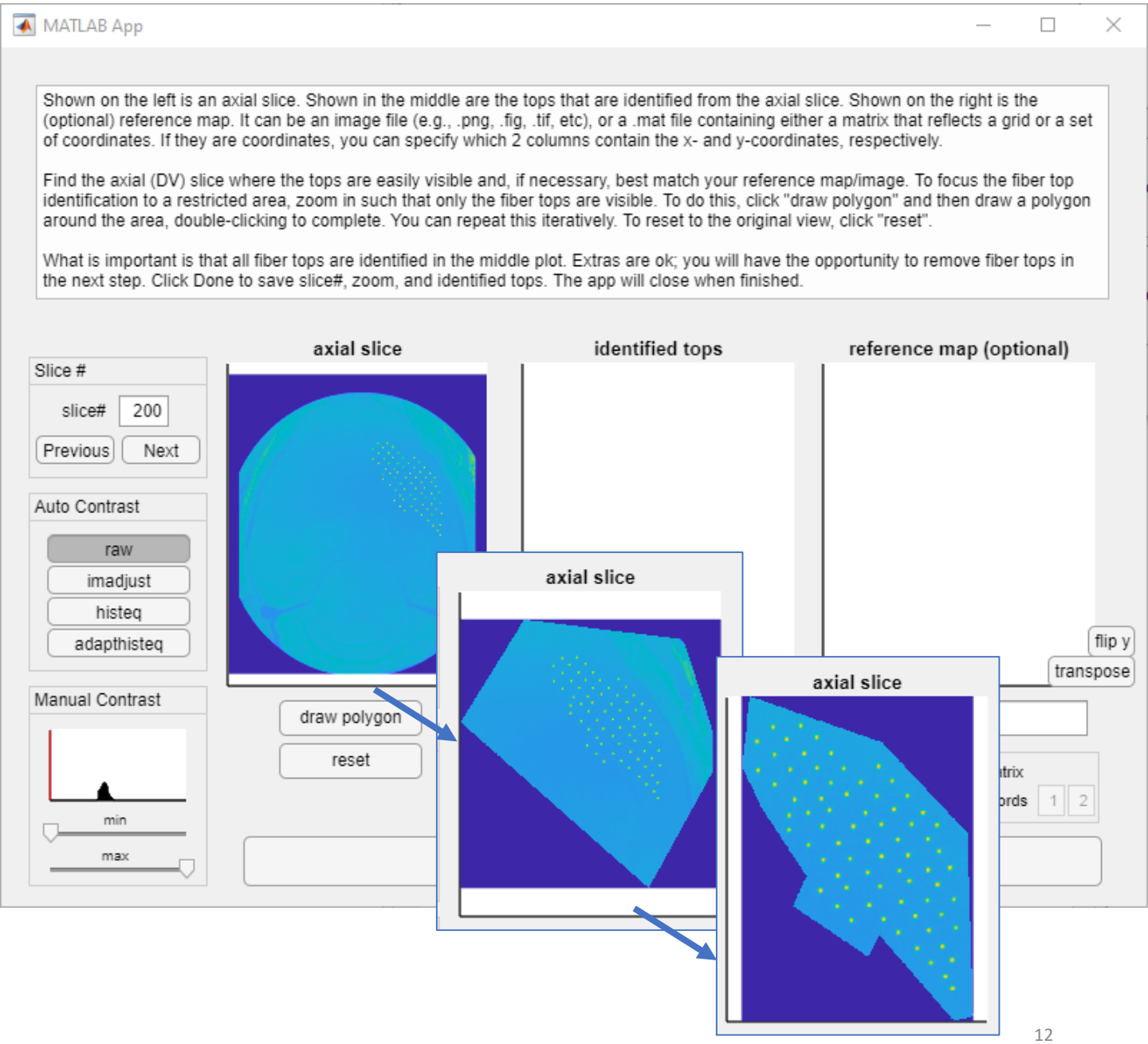


IDENTIFY FIBER TOPS

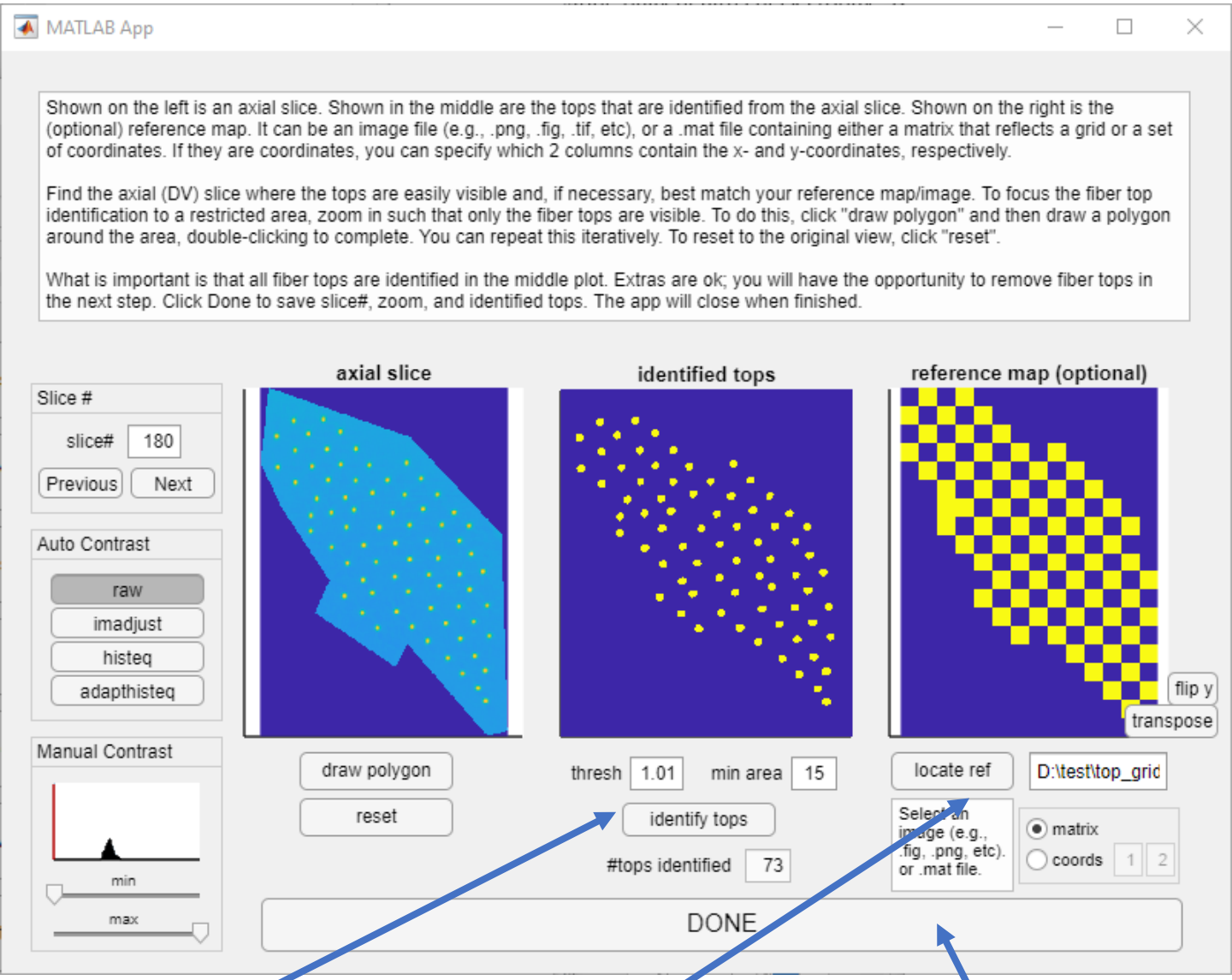


This is where you can load the reference mentioned in the “Before you begin” section. If it’s an image file, it’ll automatically display. If it’s a .mat file, specify if it’s a matrix arrangement or a coordinate file. You can specify which columns of the coordinate file contain the x-, and y-coordinates, respectively.

IDENTIFY FIBER TOPS



IDENTIFY FIBER TOPS

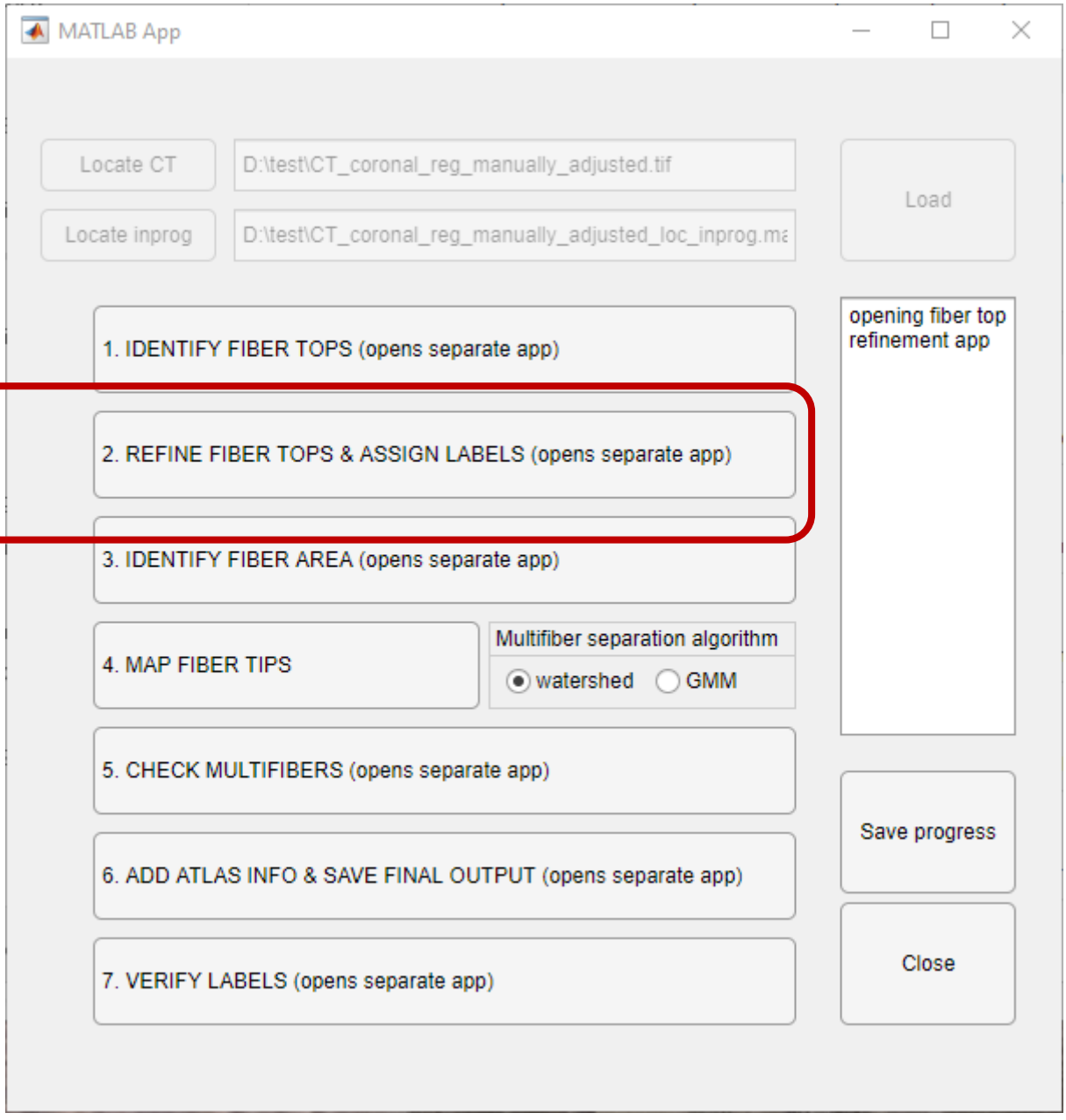


Now click identify tops. The #tops identified will be shown. You can adjust the threshold or minimum area to get better results. You may also need to pick a different slice, draw a different polygon, and identify tops again.

I con compare this to the reference I loaded that has my design. They match.

Click DONE when satisfied

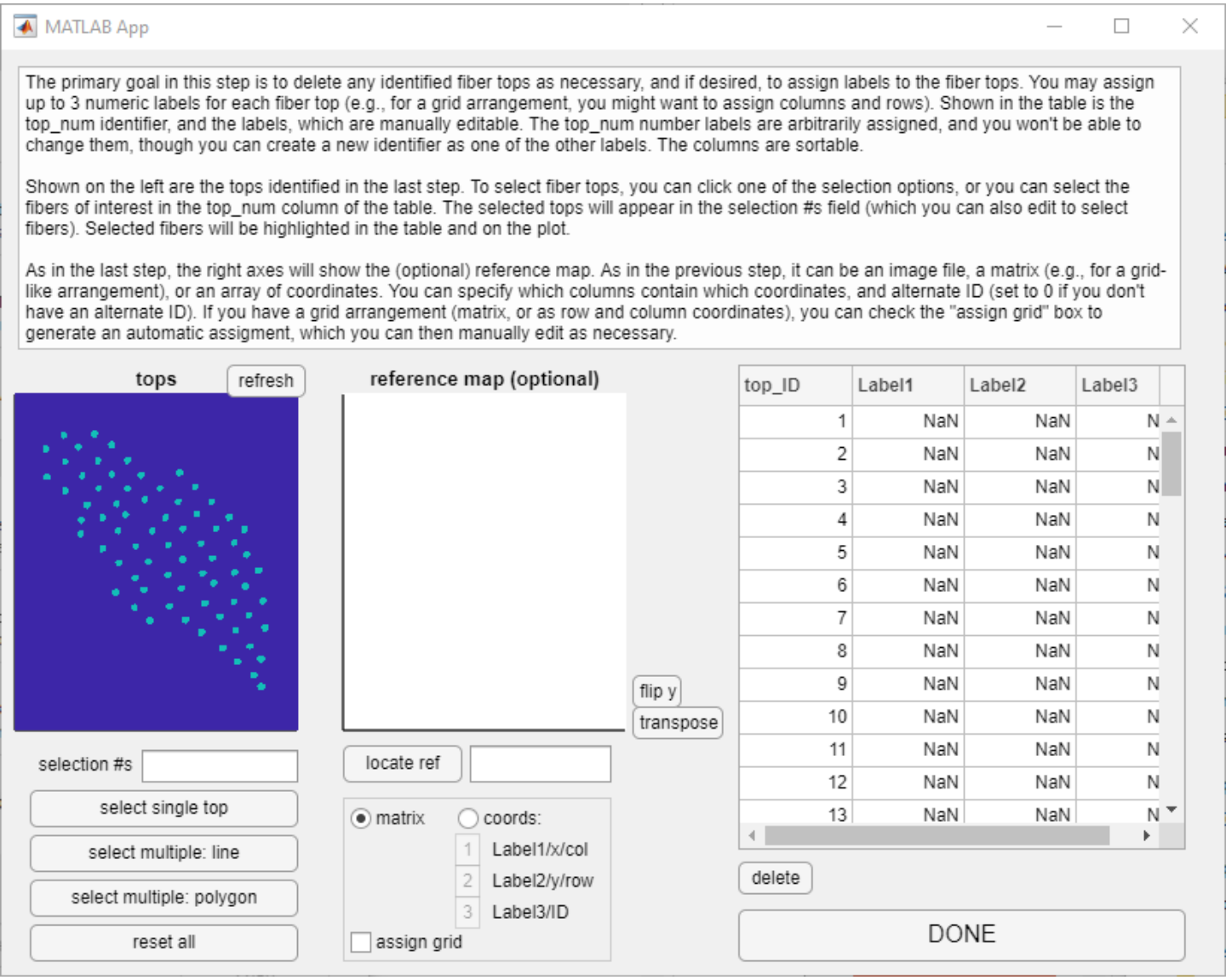
REFINE FIBER TOPS & ASSIGN LABELS



REFINE FIBER TOPS & ASSIGN LABELS

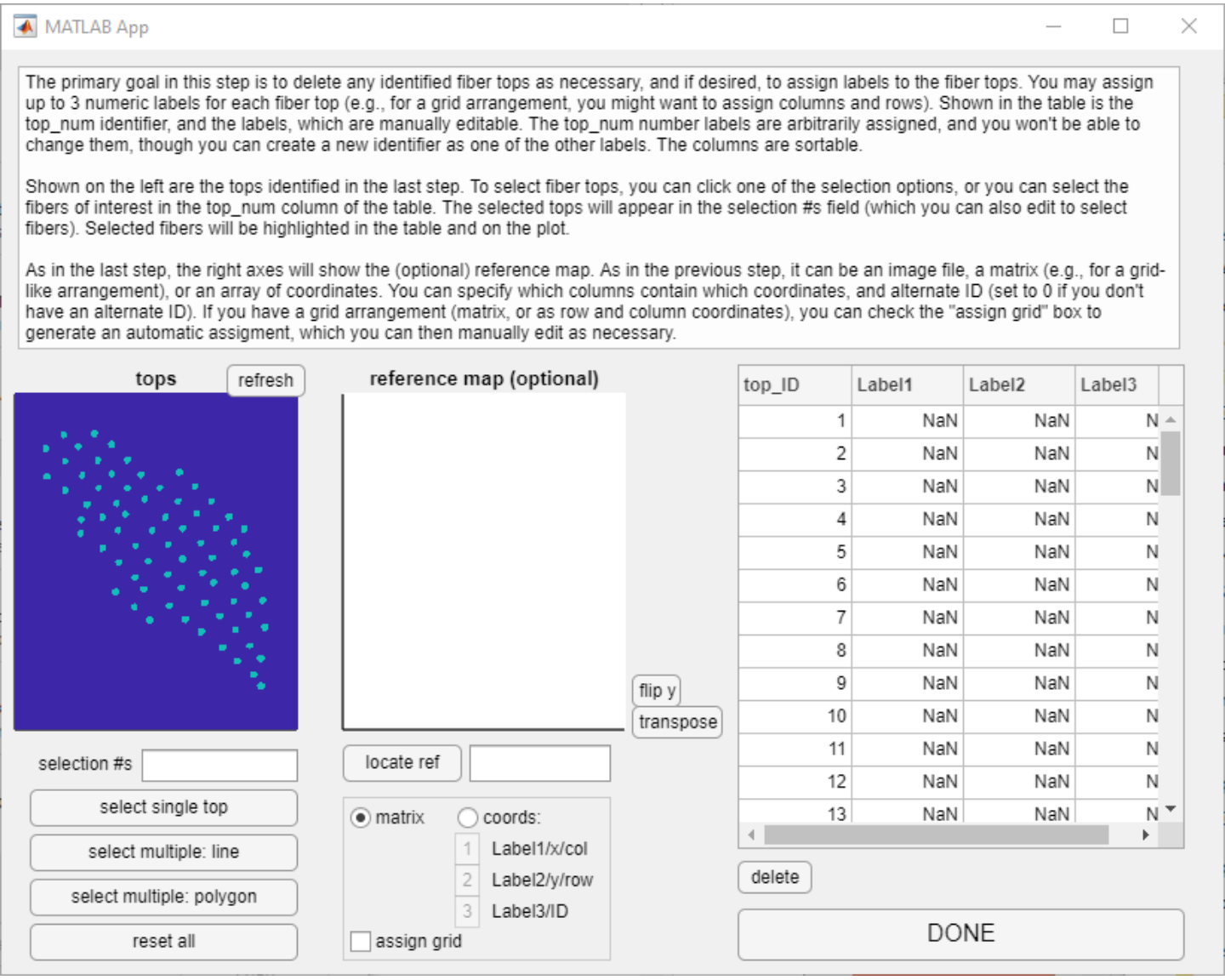
Here you can refine the tops. You cannot add tops, but you can delete tops.

If you have your fibers arranged in a grid, and want to assign each fiber top to a grid location, you can do that too.



REFINE FIBER TOPS & ASSIGN LABELS

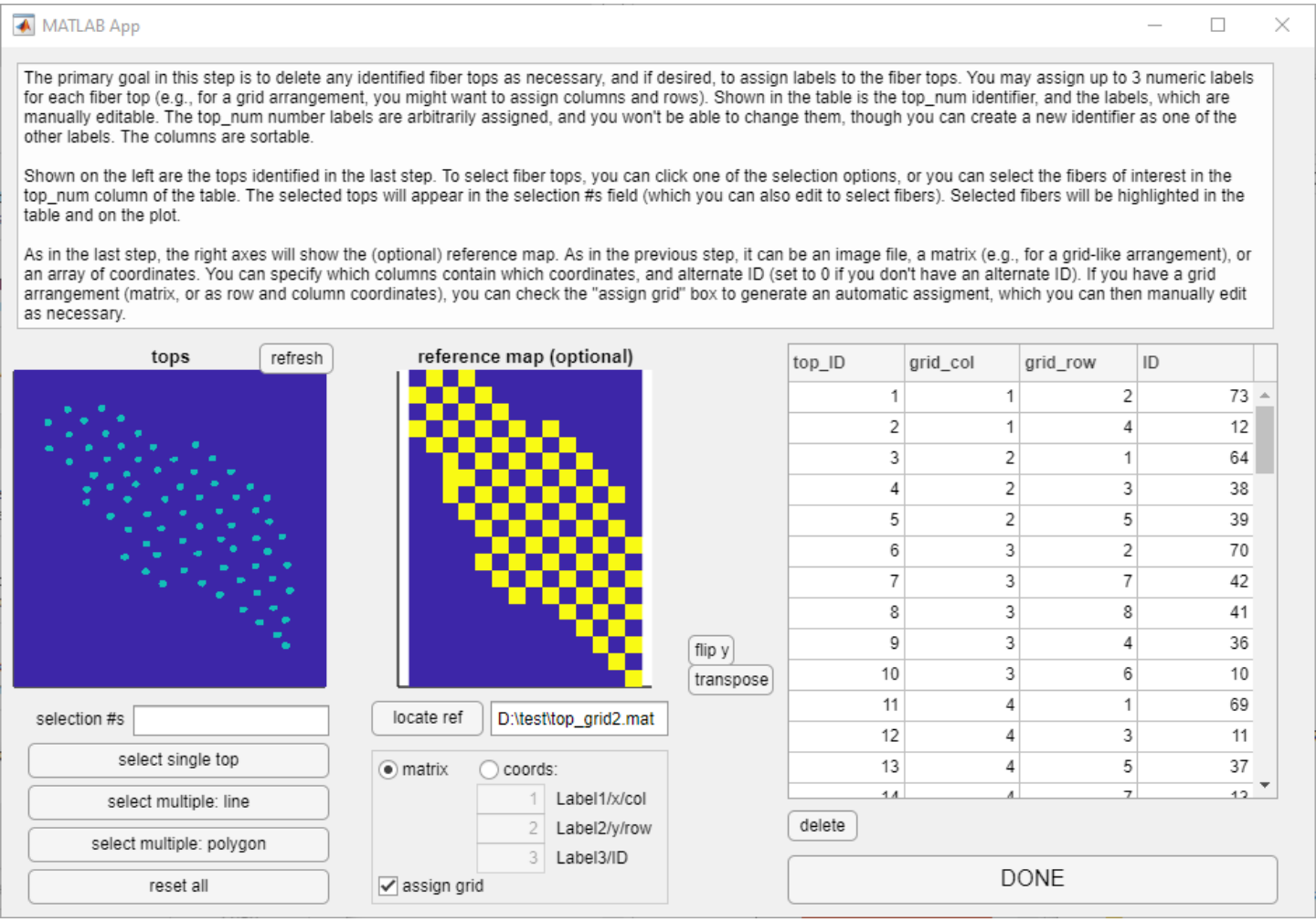
There are 3 ways to select fiber(s). You can enter numbers in the selection #s window. You can click a button to select a single top, multiple tops by drawing a line through them, or multiple tops by drawing a polygon around them. You can also select tops by clicking the numbers in the top_ID column. The whole row will be selected



REFINE FIBER TOPS & ASSIGN LABELS

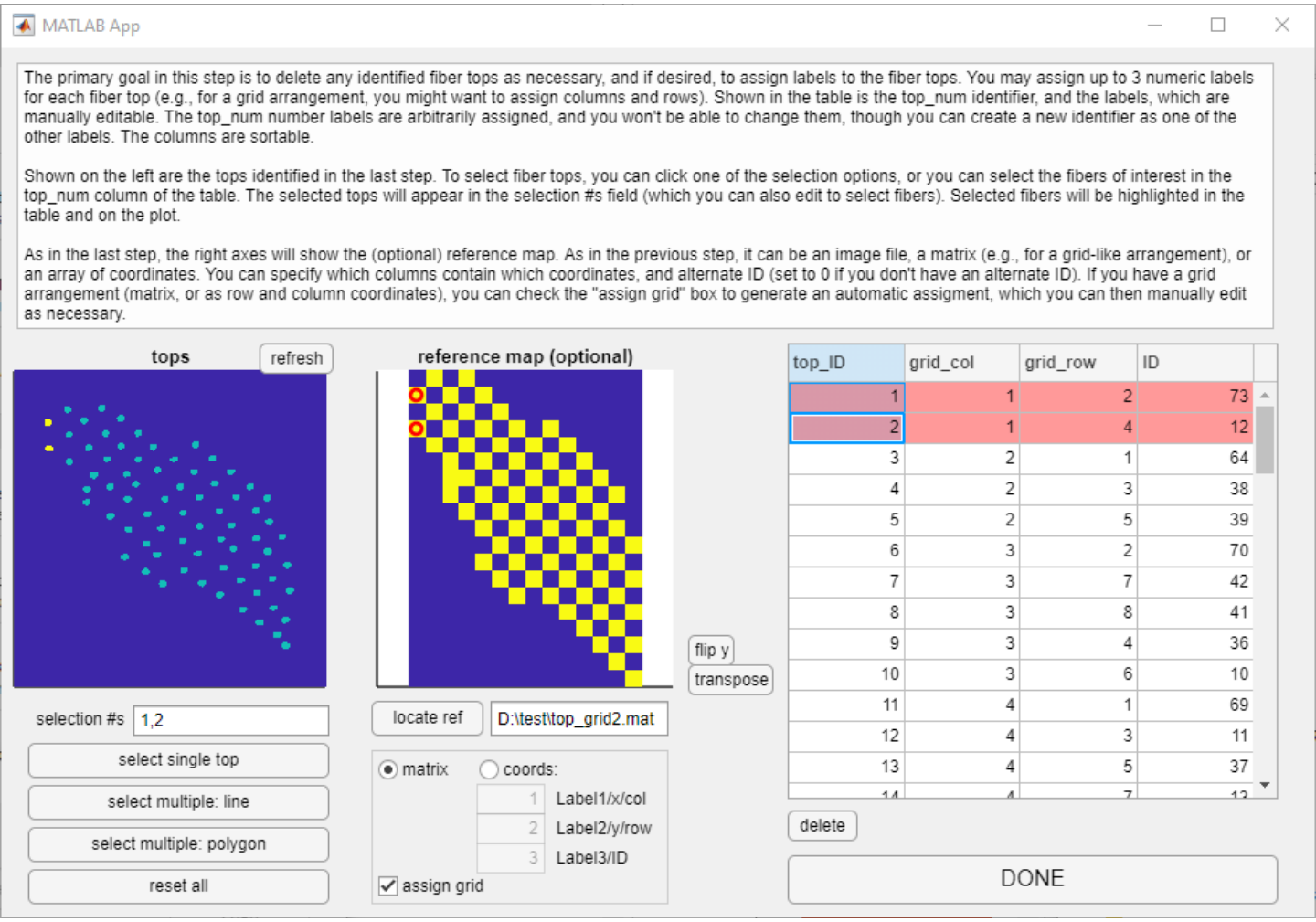
Here in this example, I've loaded my reference, and specified that it is a matrix (see slide 4). Note that "flip y" and "transpose" just apply to the display, and don't change the grid assignment.

To generate first-pass automatic grid location assignments, check the "assign grid" box. You'll see that the columns for grid_col, grid_row, and ID automatically populate.



REFINE FIBER TOPS & ASSIGN LABELS

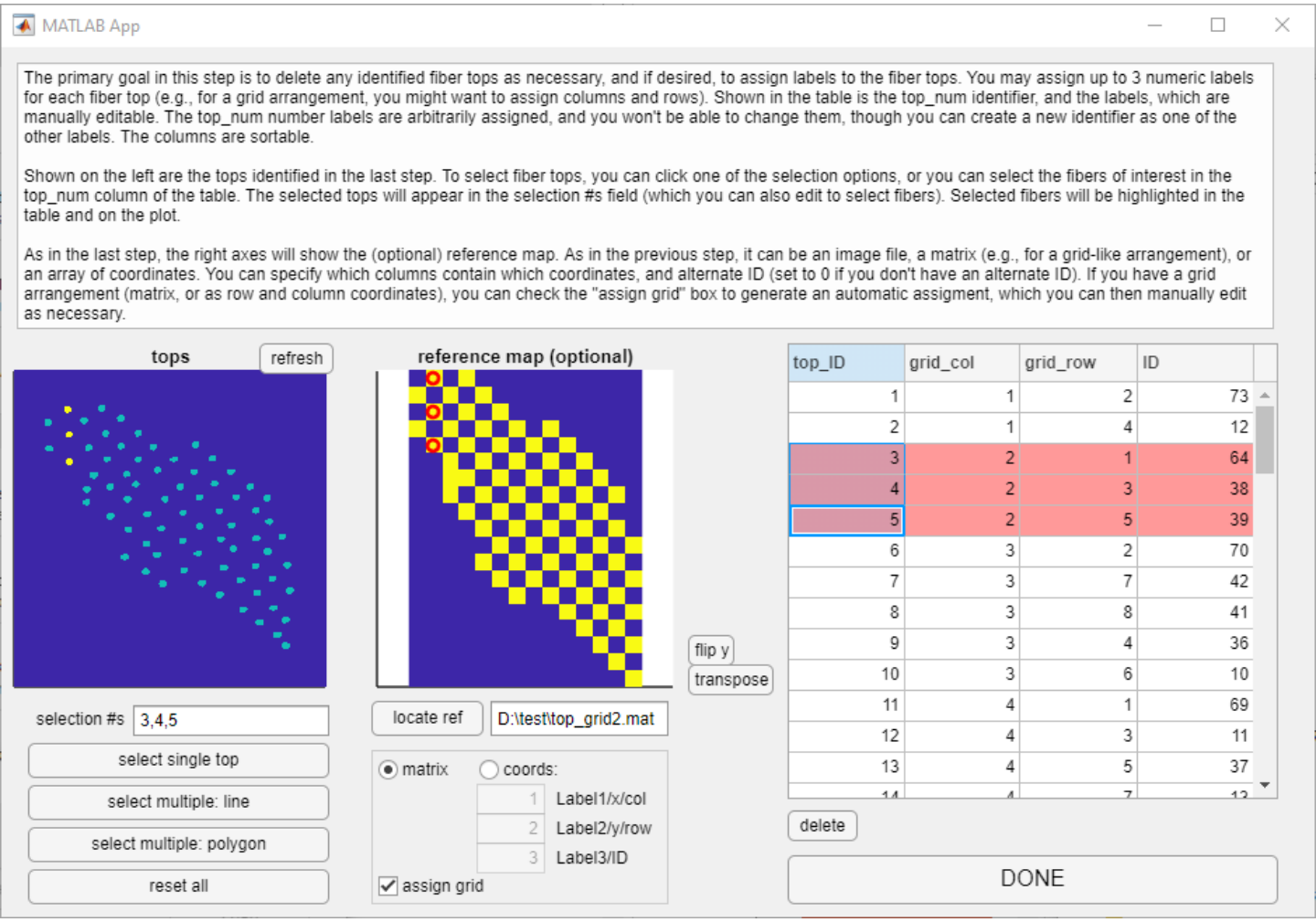
Now you need to manually verify that these are correct. In this example, I start with the grid columns but sorting the grid_col column (click the heading). Now I select the rows (remember: click in the top_ID column to make a selection) that correspond with column 1. The fibers will be highlighted in the identified tops (first image), and if applicable, the reference map will display the columns/rows highlighted.



If you want to change the column or row assignment of any of the fibers, click in the grid_col or grid_row box you want to change, and edit the number. Clicking delete will delete the tops corresponding to the highlighted rows.

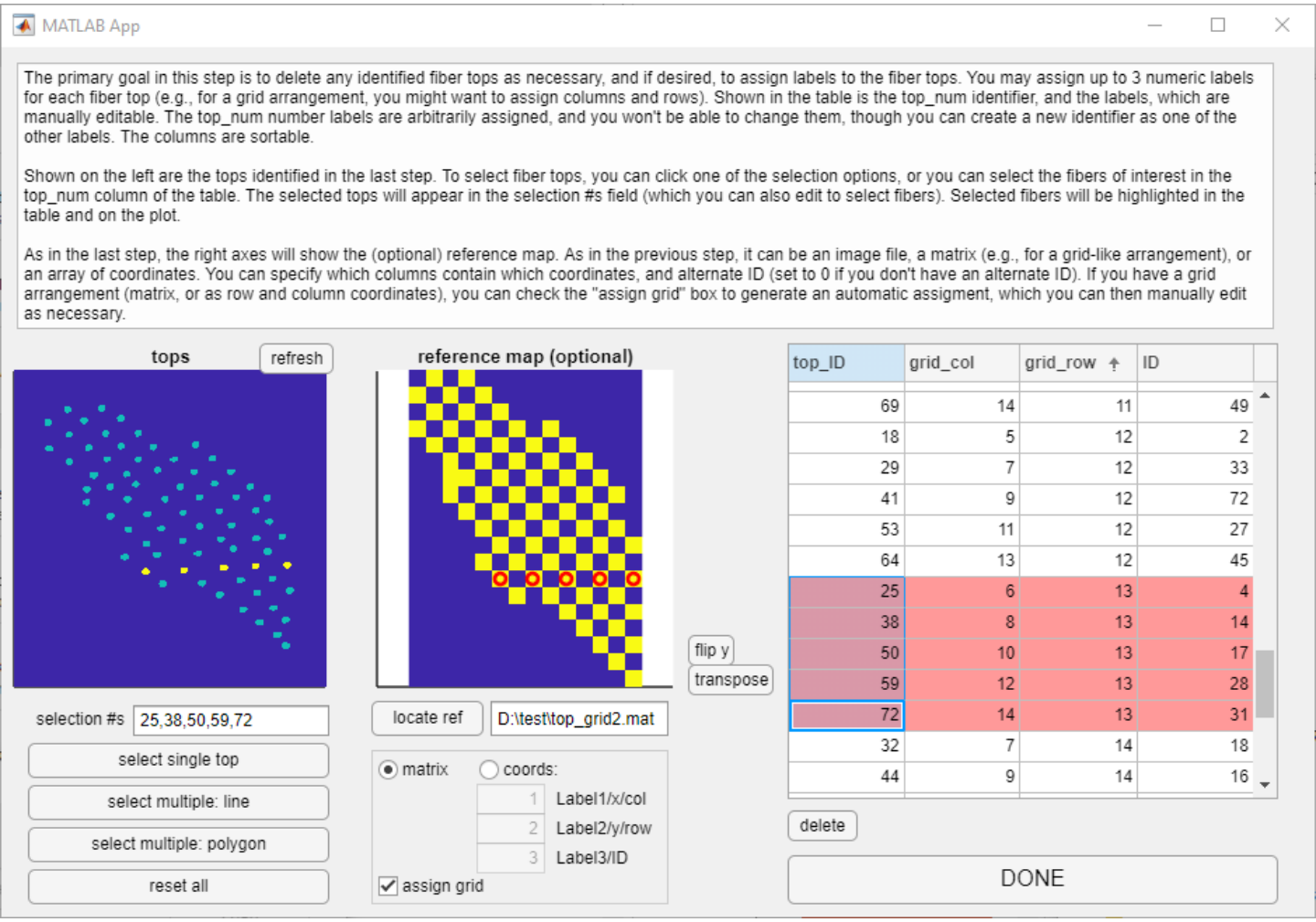
REFINE FIBER TOPS & ASSIGN LABELS

Go through all the columns



REFINE FIBER TOPS & ASSIGN LABELS

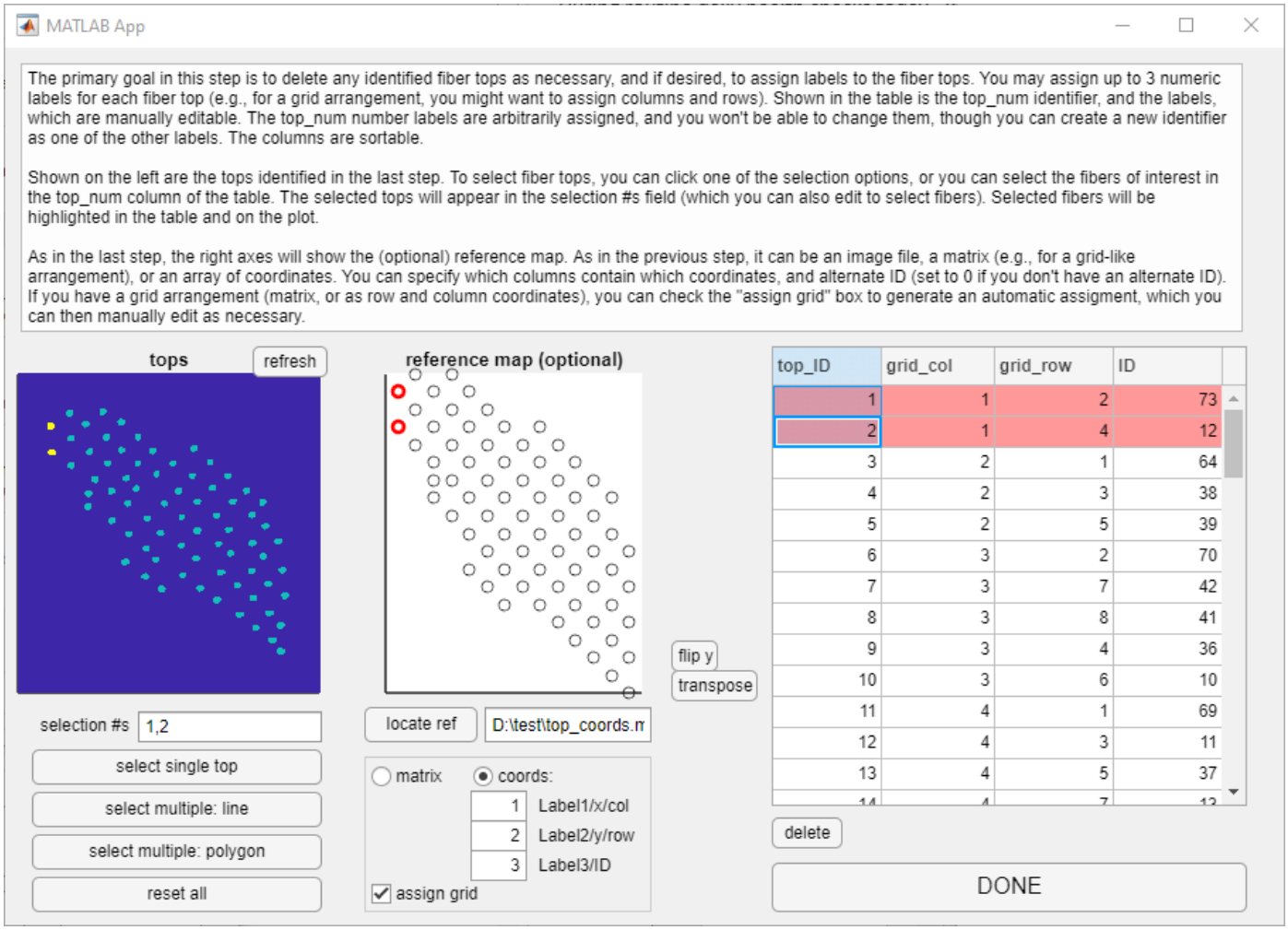
And also the rows. To easily look at rows, sort the grid_row column.



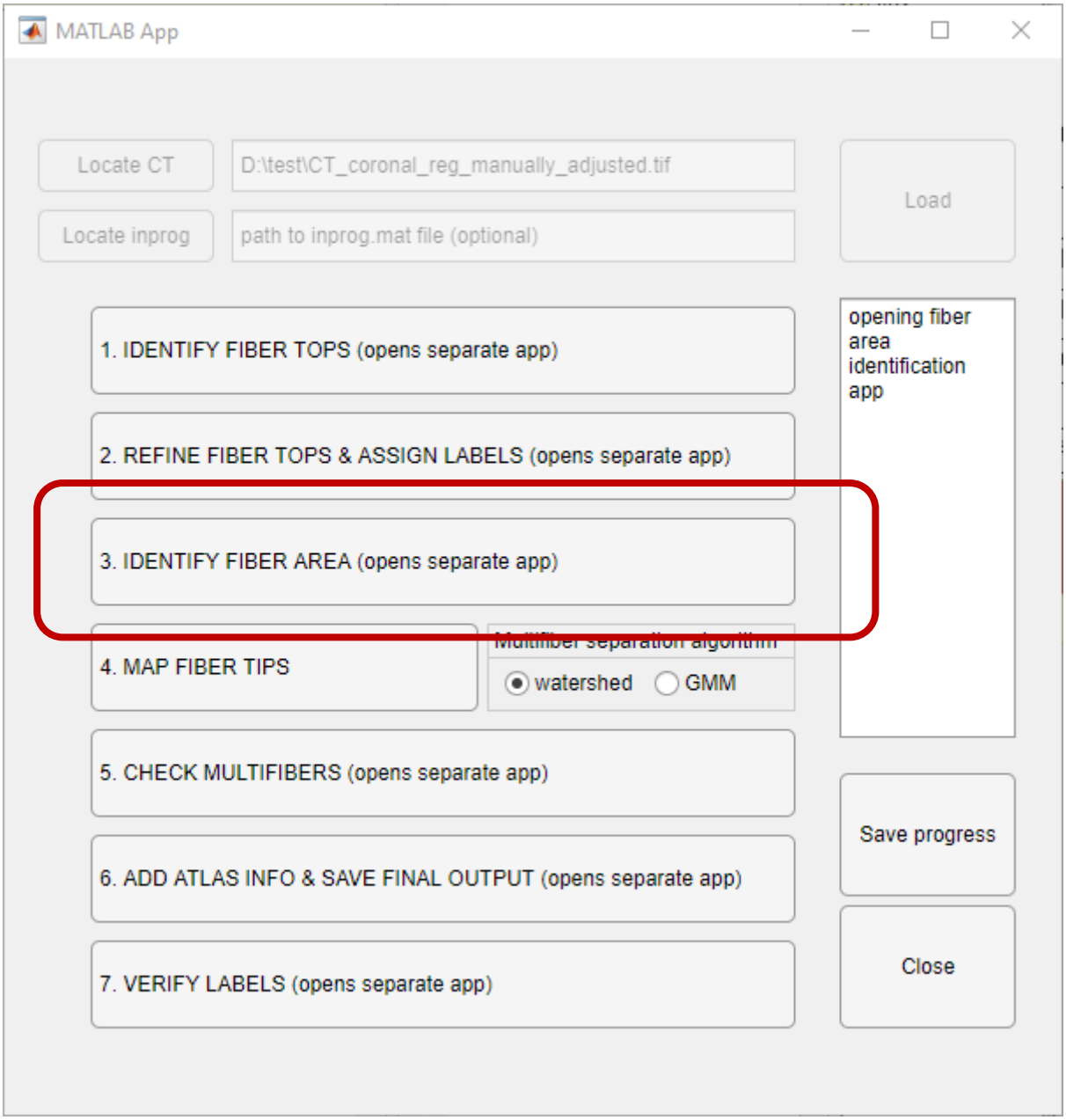
Click done when you are satisfied.

REFINE FIBER TOPS & ASSIGN LABELS

Here's an example of what the reference map would look like if you loaded a set of coordinates instead.

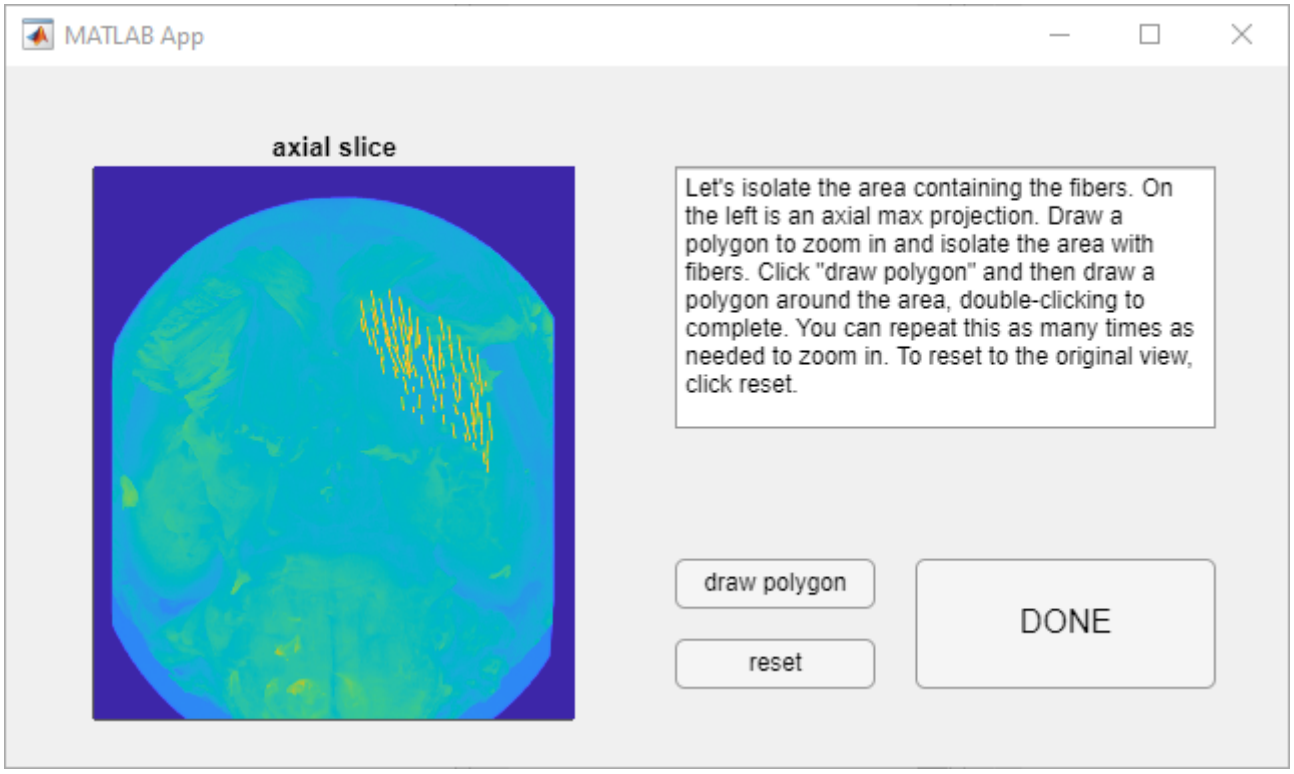


IDENTIFY FIBER AREA



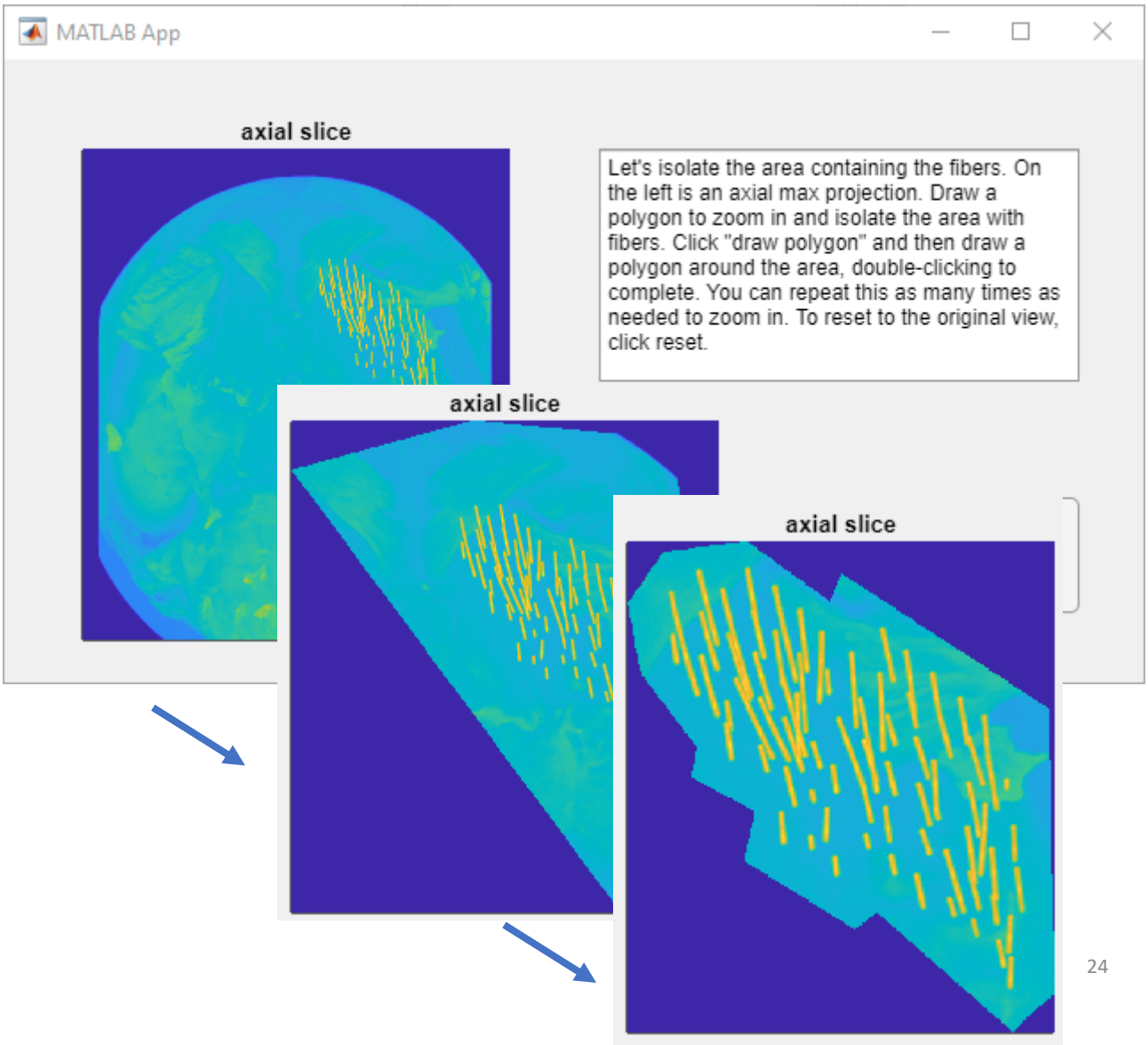
IDENTIFY FIBER AREA

We will eventually be automatically detecting fibers based on brightness. To make that search easier, we will restrict the detection area axially based on where we know fibers are, based on the maximum projection. Use the draw polygon button to isolate this area. Repeat as necessary. You can reset. Click done when you are satisfied



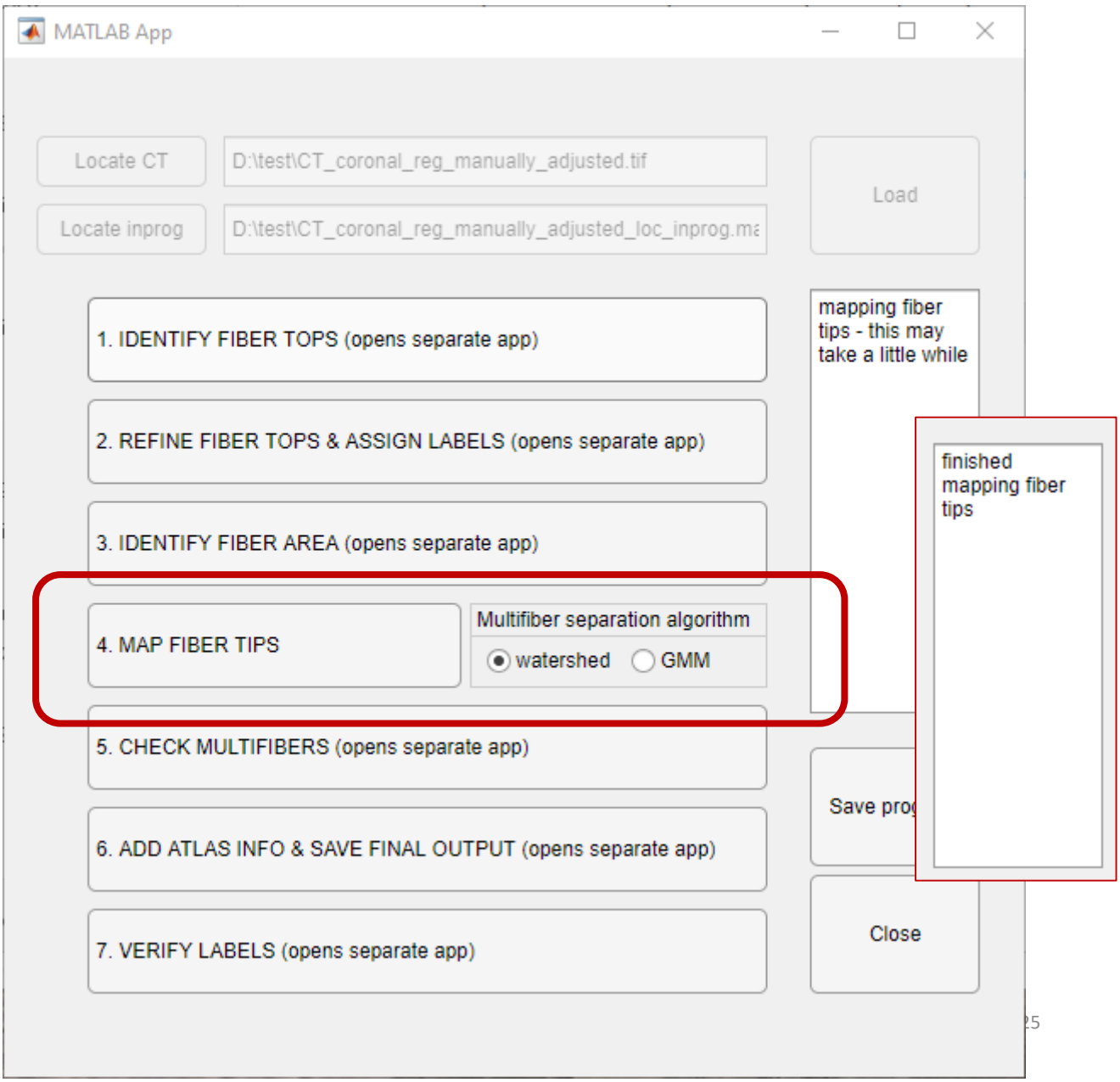
IDENTIFY FIBER AREA

We will eventually be automatically detecting fibers based on brightness. To make that search easier, we will restrict the detection area axially based on where we know fibers are, based on the maximum projection. Use the draw polygon button to isolate this area. Repeat as necessary. You can reset. Click done when you are satisfied



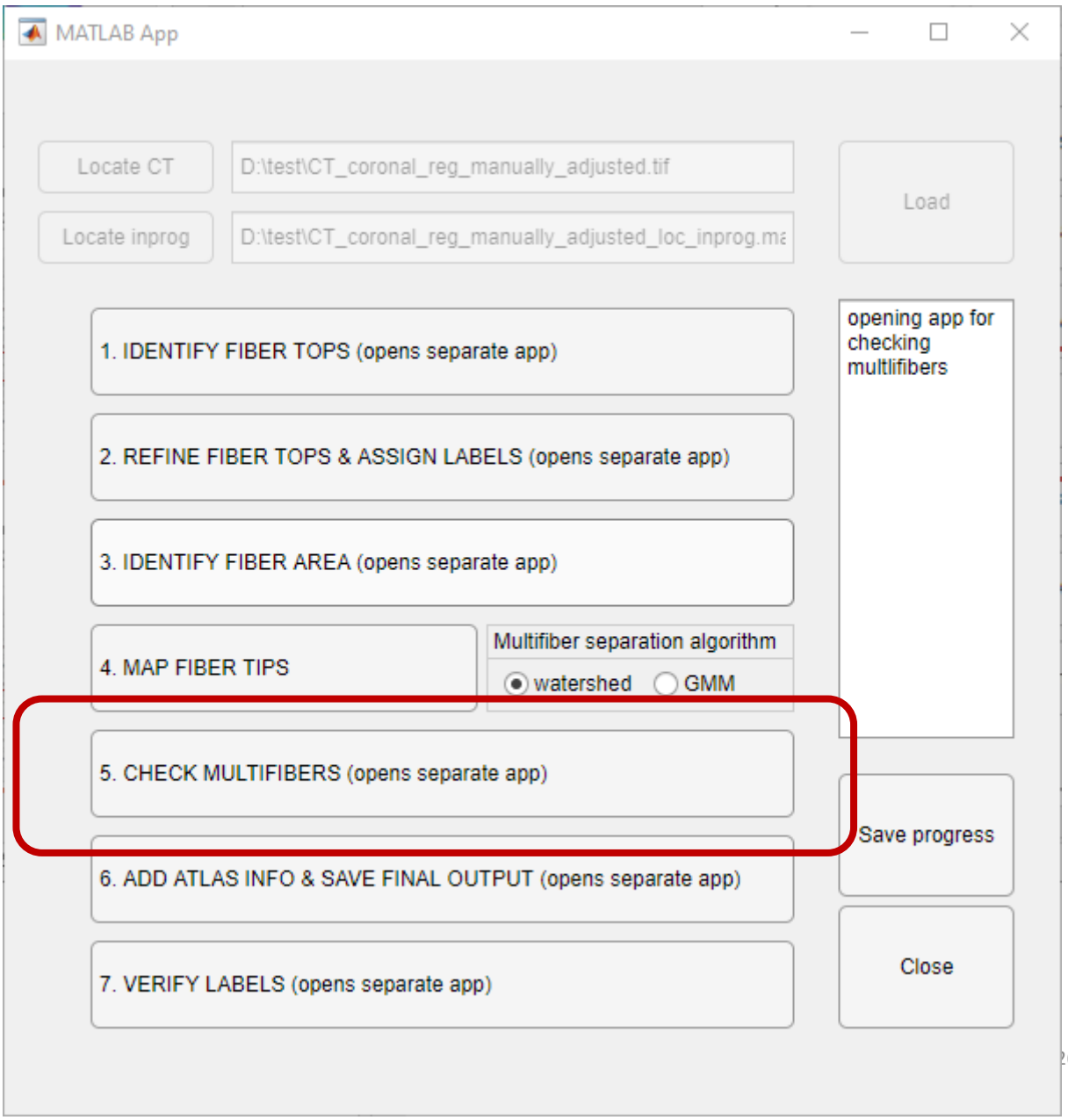
MAP FIBER TIPS

Now it's time to automatically detect the fibers and map them to your identified tops. For separating any fibers that are touching ("multifibers"), you can choose between the watershed (see Vu et al., 2024) and gaussian mixture models (this might entail some trial and error). This does not open a separate app. You will be notified when it is finished.

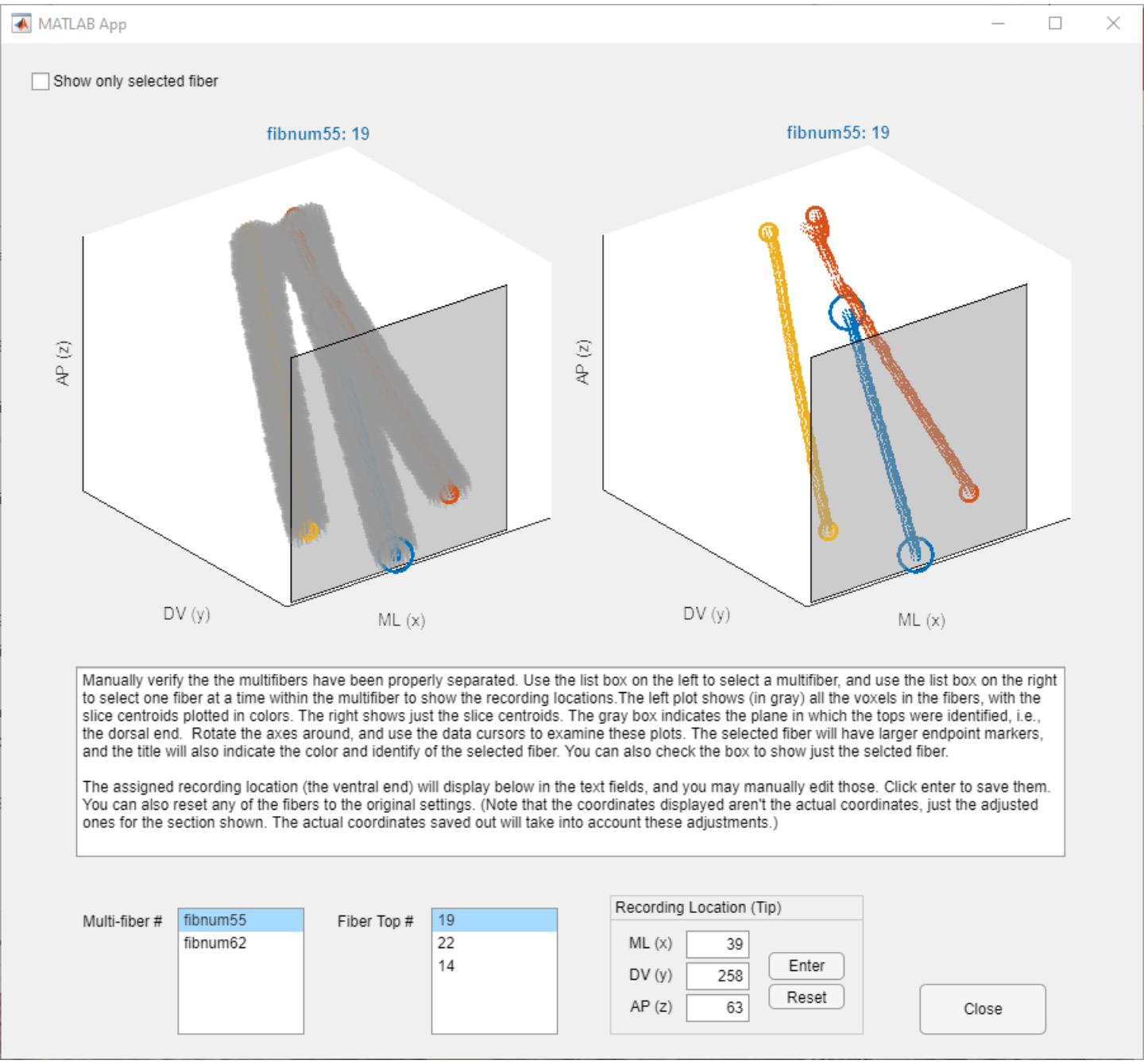


CHECK MULTIFIBERS

Fibers that are touching get initially lumped together as a single fiber. The mapping step includes an automatic separation of them, but it's not perfect and you'll need to manually check the separation, and refine as necessary.

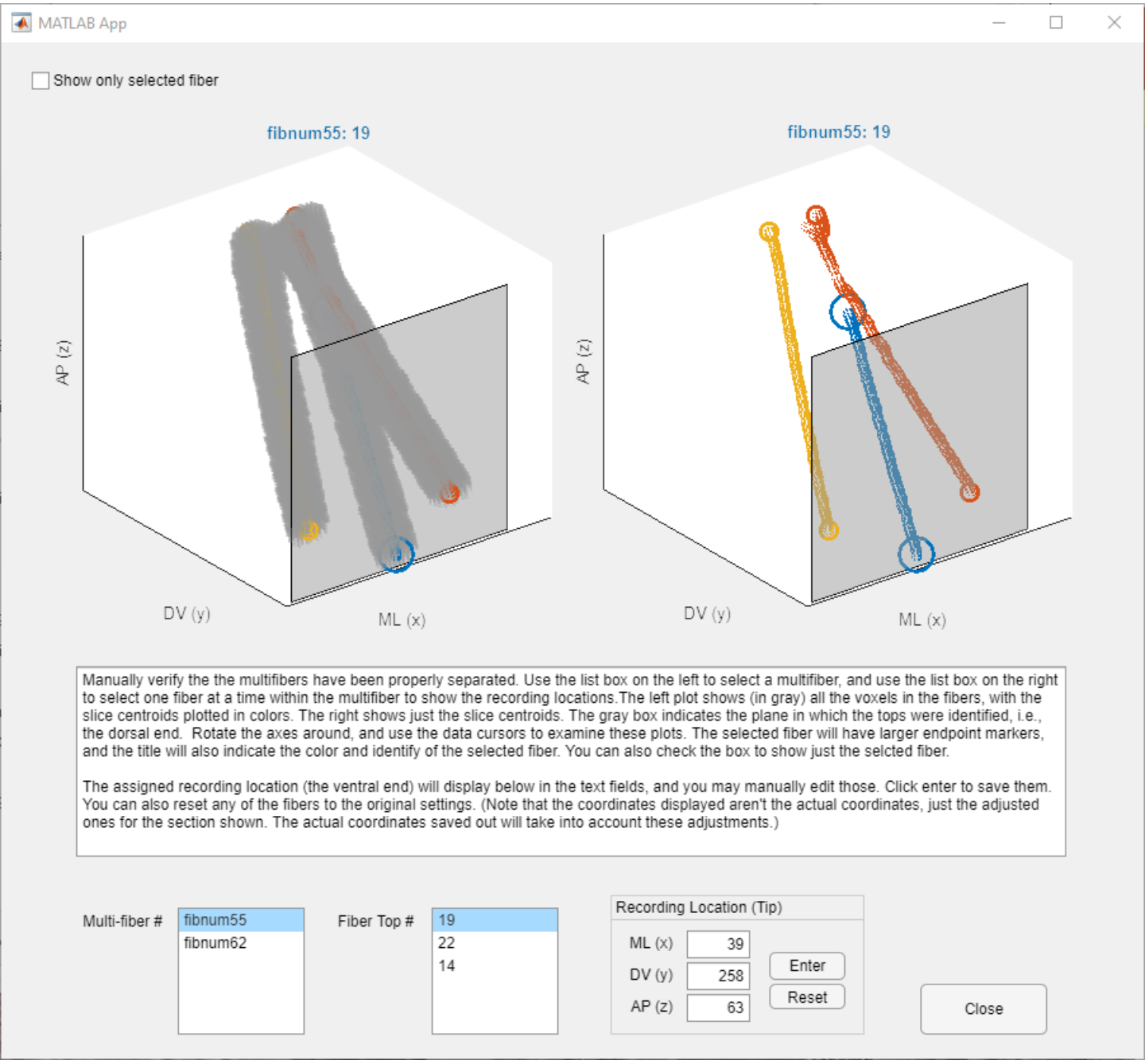


CHECK MULTIFIBERS



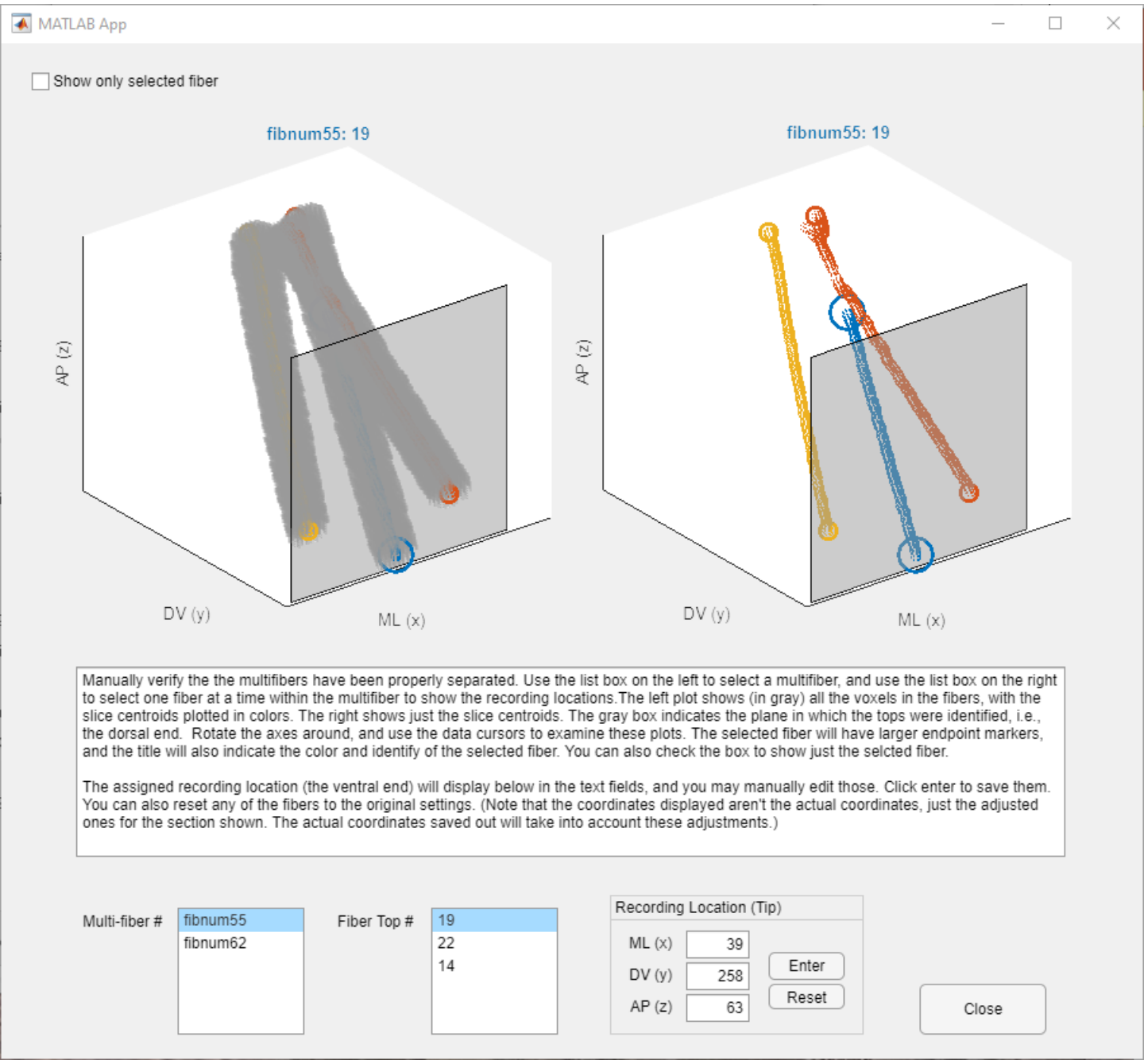
The left menu contains the multifibers (fiber bottom #), and the right menu contains the fiber top #s that belong in this multifiber (these identification numbers themselves are automatically assigned in previous steps).

CHECK MULTIFIBERS



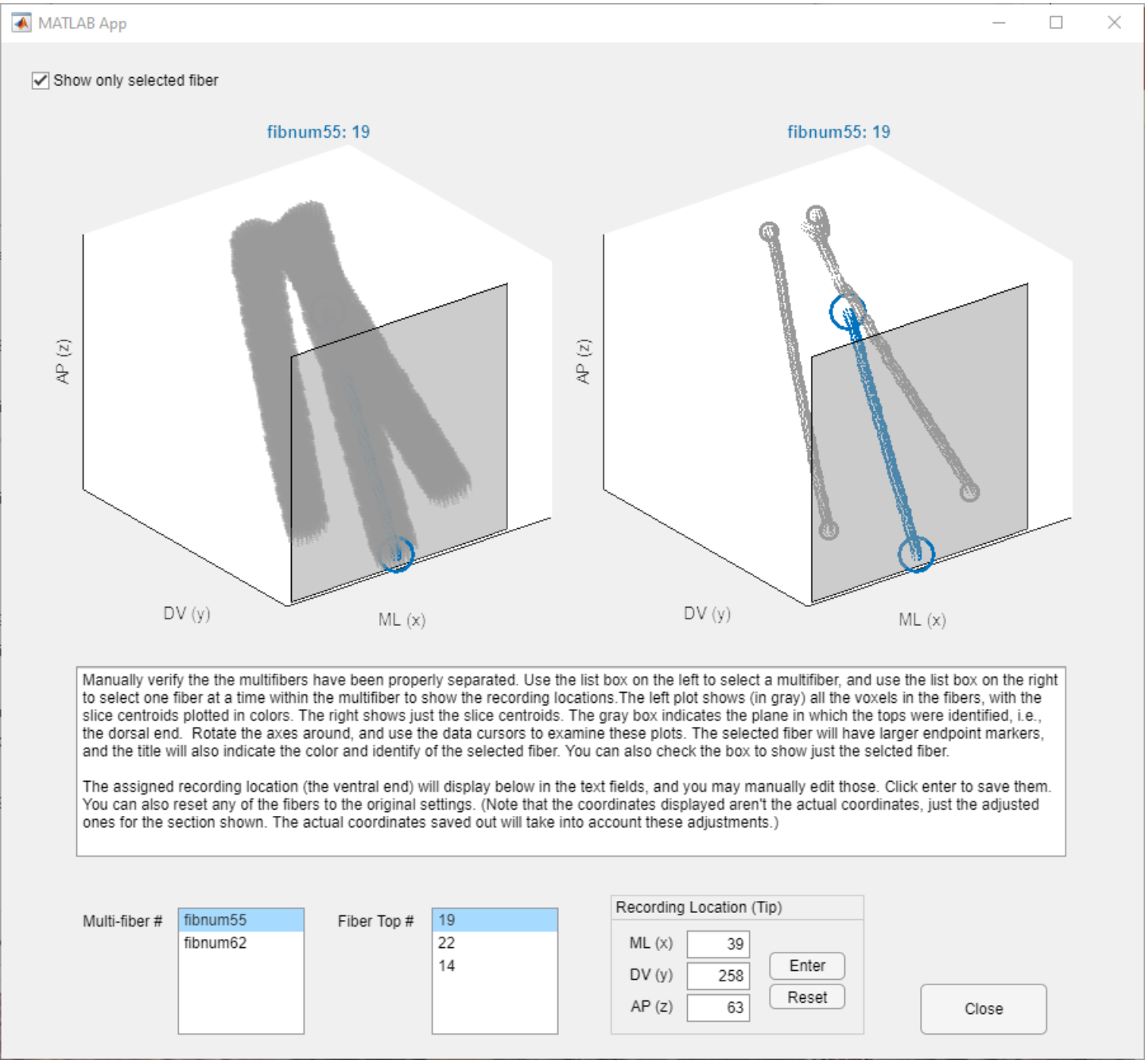
The left hand figure shows (in gray) all the voxels in this multifiber. Plotted in color (both plots) are what was automatically identified as the centroids of those cross sections.

CHECK MULTIFIBERS



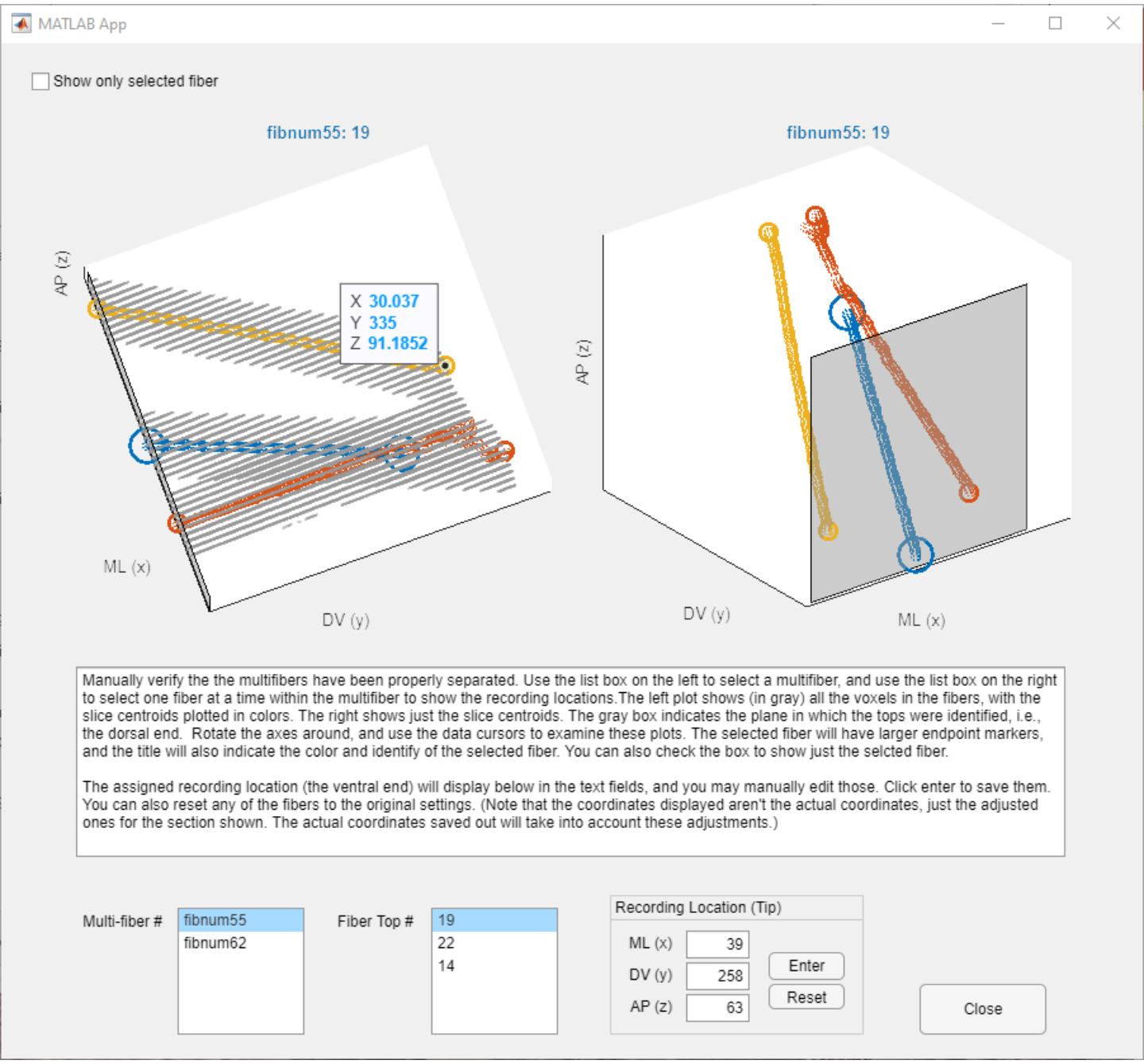
The selected fiber will have larger circles at the endpoints, and the coordinates of the location are displayed. The title of the plots show the selection and its corresponding color.

CHECK MULTIFIBERS



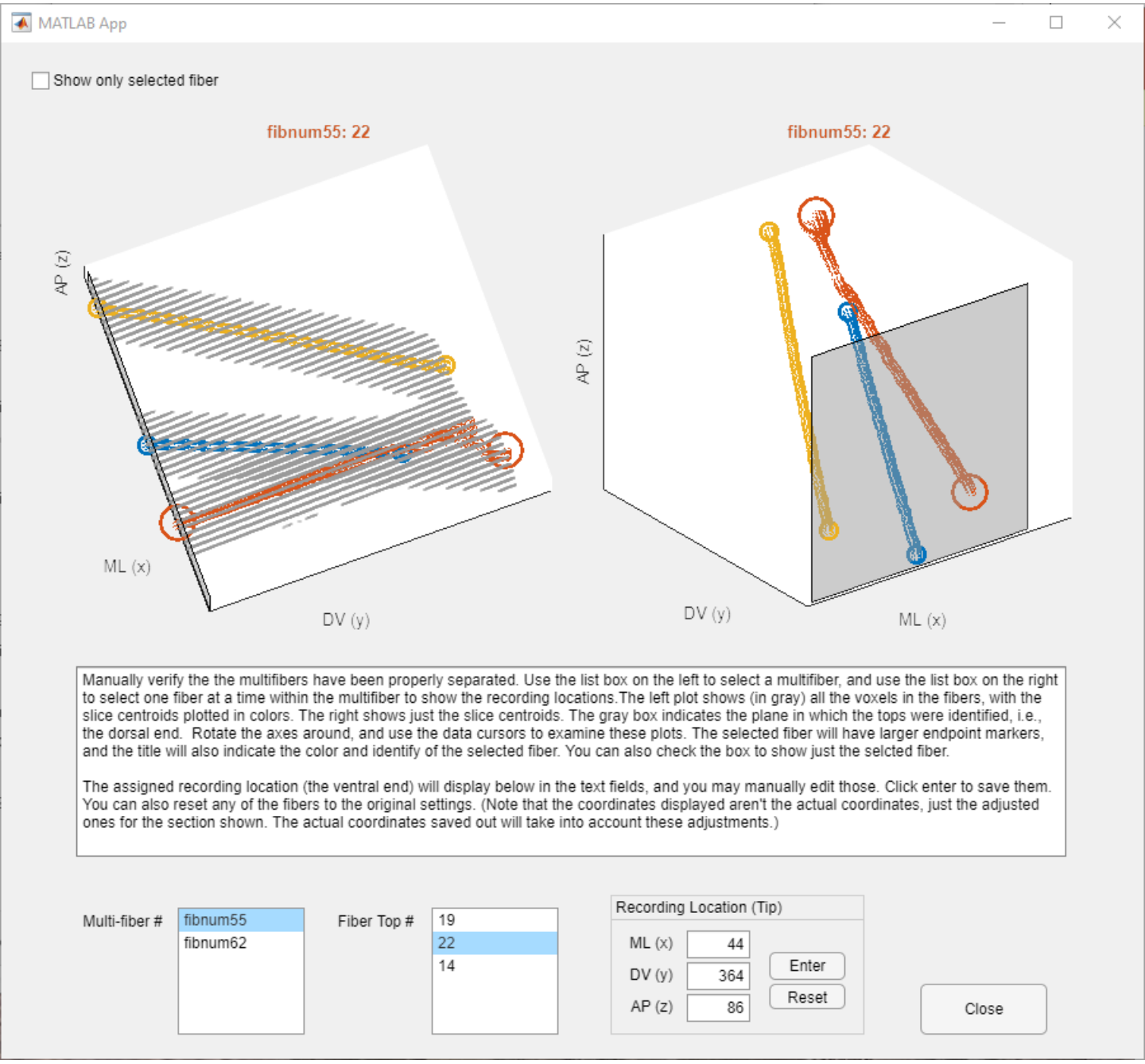
Checking the box allows you to just display the selected fiber

CHECK MULTIFIBERS



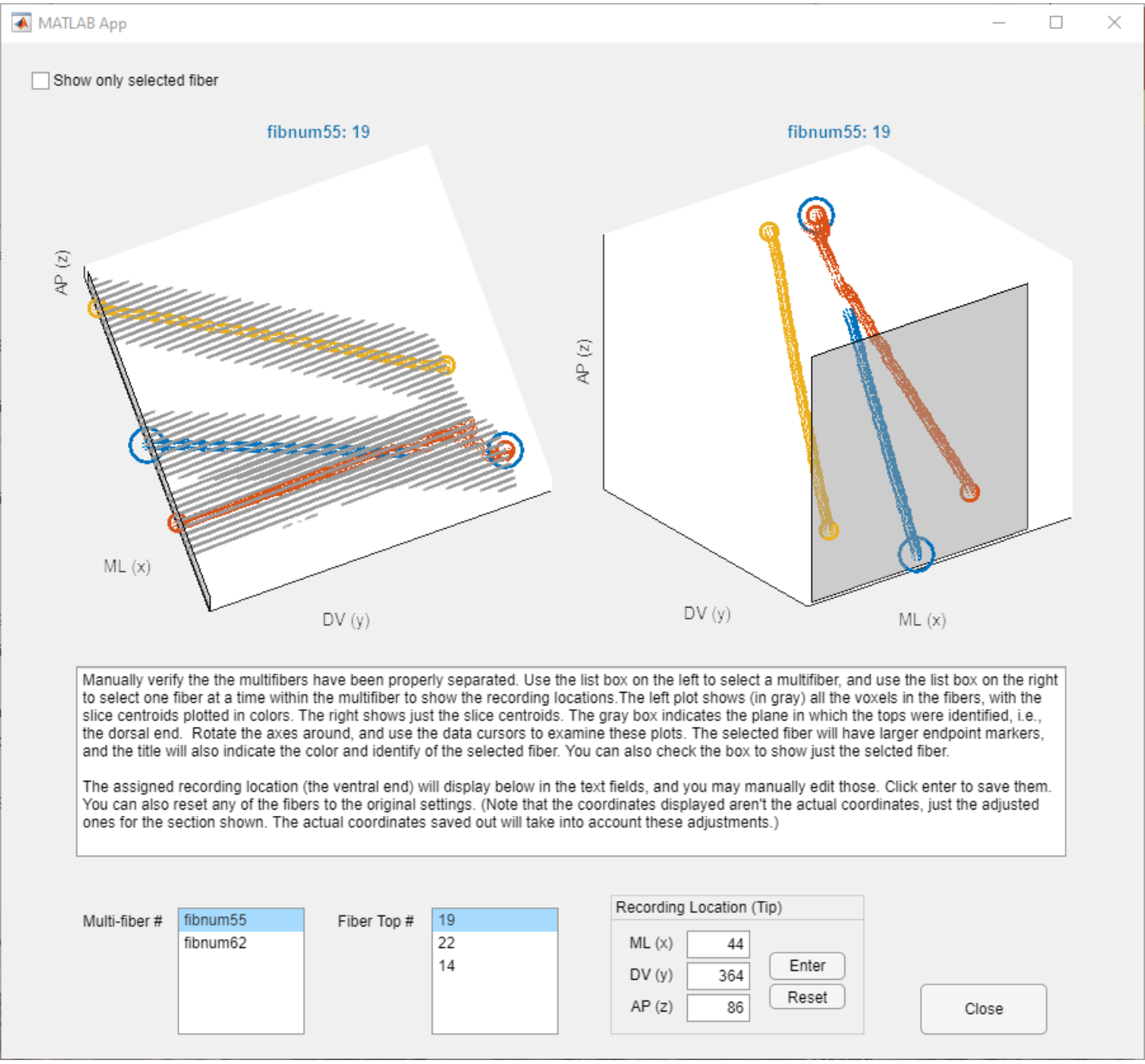
Use the plot tools (however near the title to see tools to rotate, pan, zoom, or reset the view. Swivel the plots around to see whether you agree. Clicking will display coordinates of your clicked point. In this case, we see that the blue and orange endpoints where incorrectly identified.

CHECK MULTIFIBERS



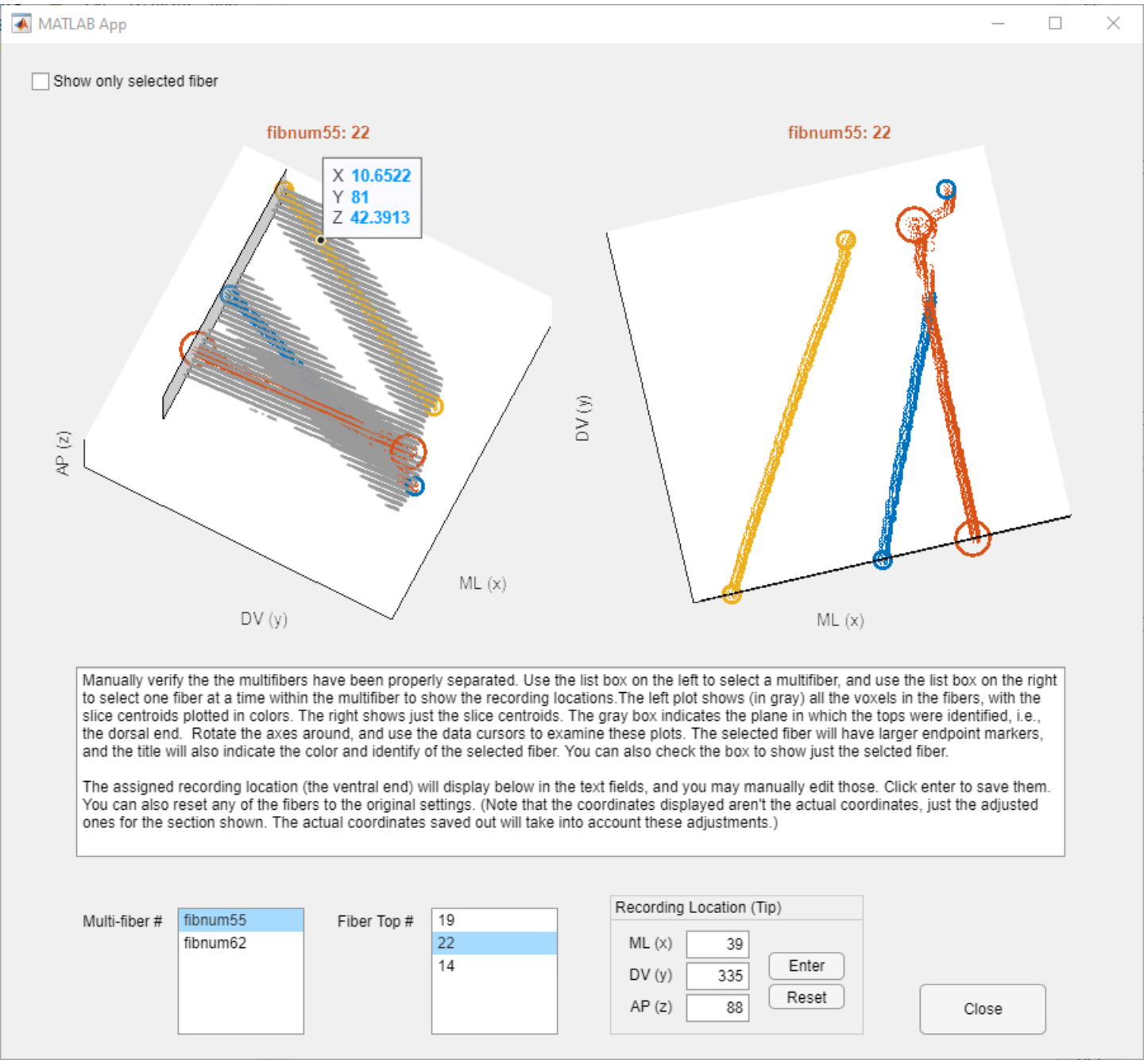
The orange endpoint should be the blue endpoint, so we click the orange fiber to note those coordinates.

CHECK MULTIFIBERS



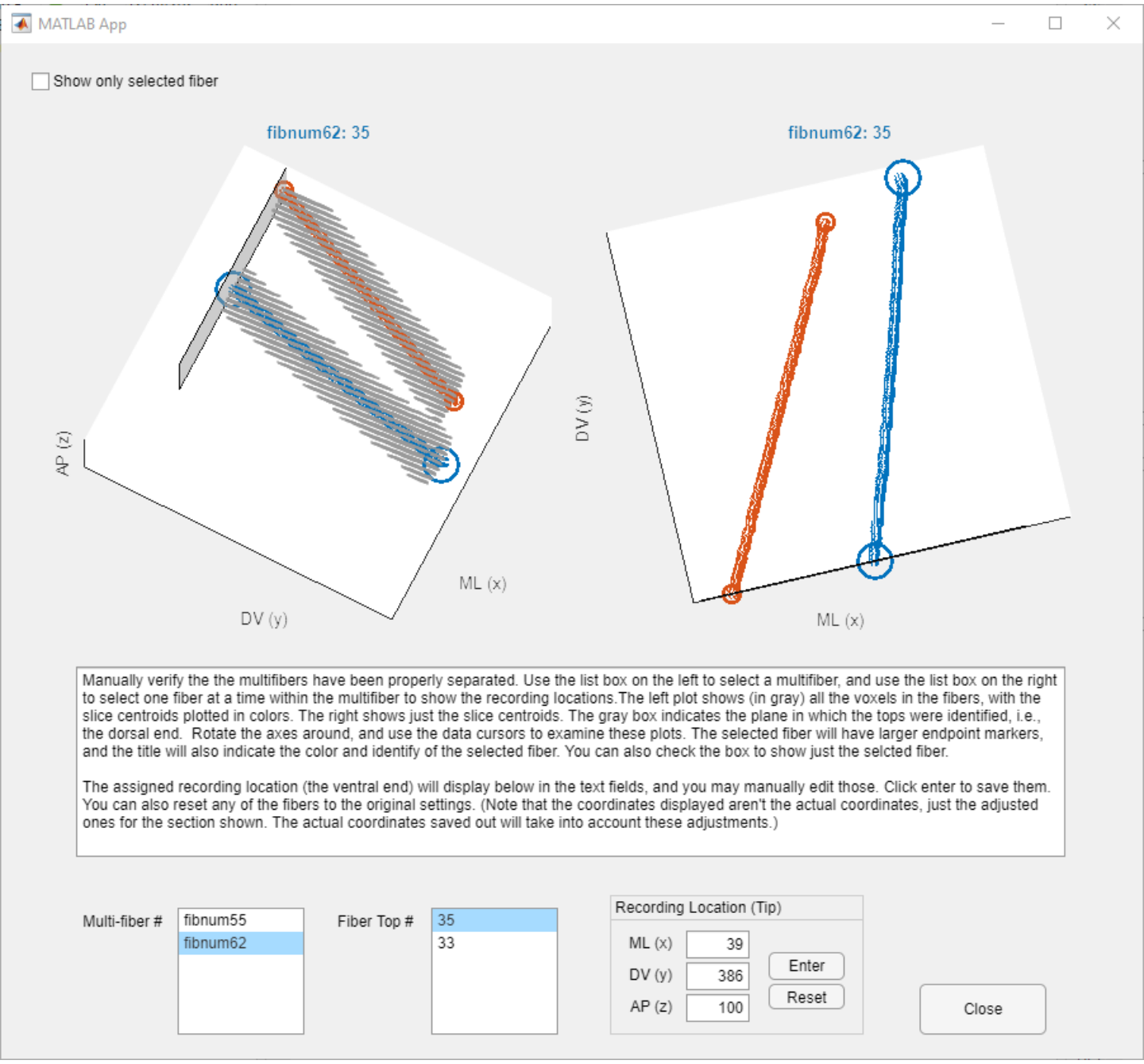
Now we go back to the blue fiber, enter those coordinates, and click “Enter”. The plot is updated.

CHECK MULTIFIBERS



Now we want to change the orange endpoint. Click around in the left plot until you find what is the endpoint (I moved the data cursor here so as not to occlude the end). Enter those coordinates, and click Enter.

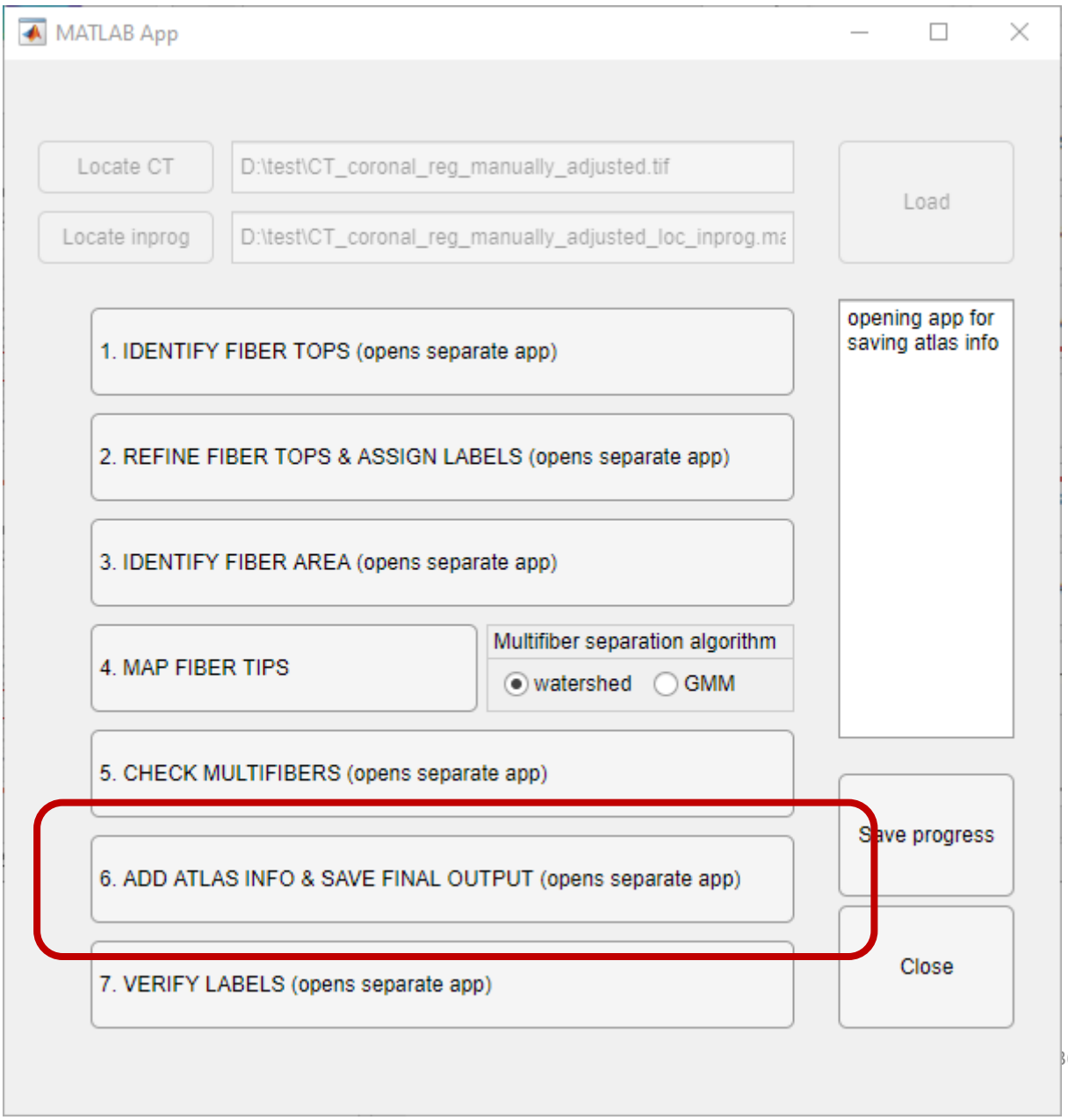
CHECK MULTIFIBERS



We can see here the second multifiber was properly separated. Once you're finished, click Close to return to the main fiber localization app.

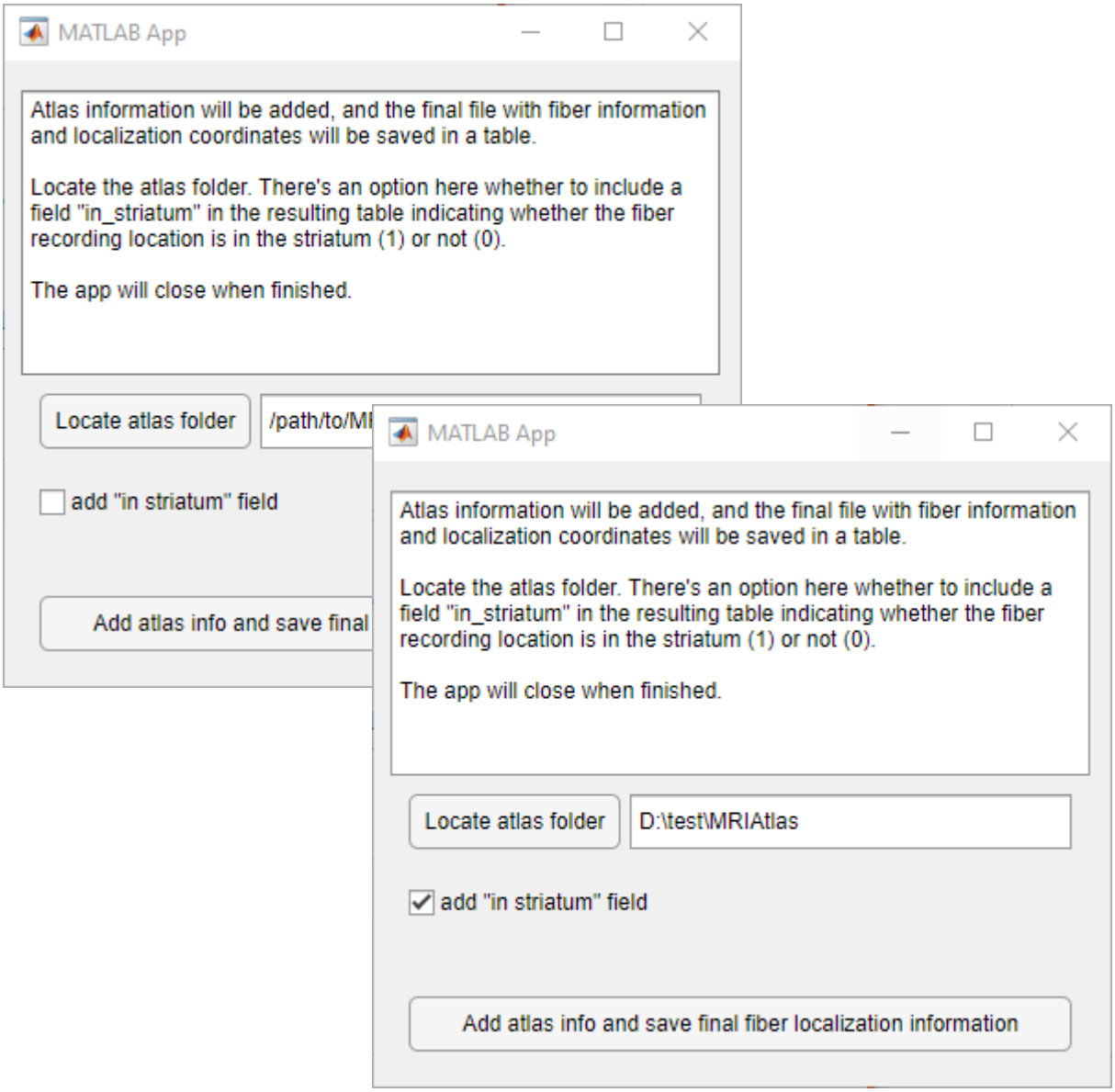
ADD ATLAS INFO & SAVE

Now that you're satisfied with the identification and mapping of fiber tops and bottoms, it's time to assign them to the atlas (see Vu et al., 2024 for atlas references).



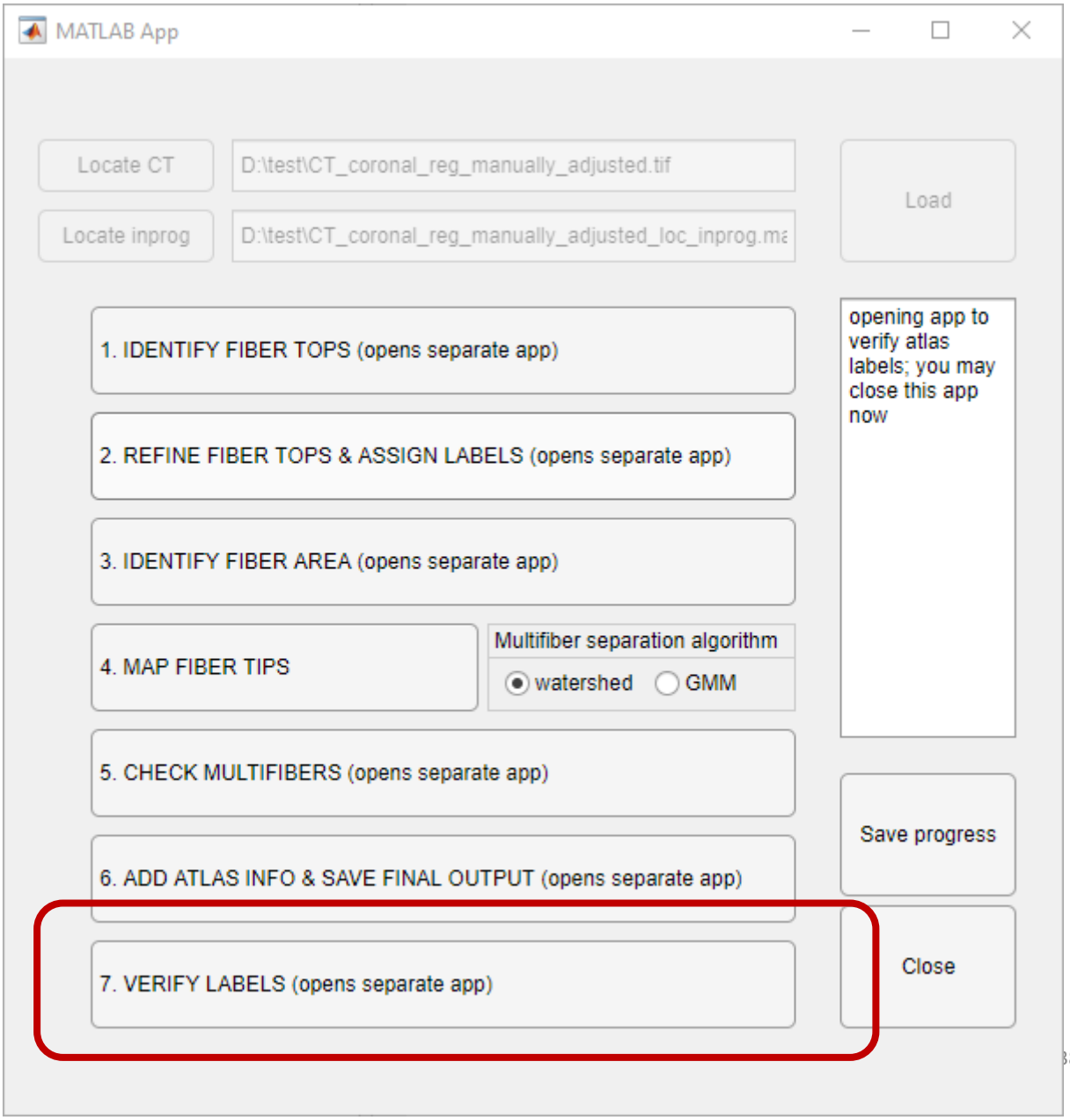
ADD ATLAS INFO & SAVE

Locate your atlas folder. In Vu et al., 2024, we were only looking in striatum, so built in is a filter for that, which will output a column in the resulting table with 1 and 0 indicating whether the recording location is in the striatum. You may opt into that as well. This app saves and closes automatically when finished. Note that it may take a while.



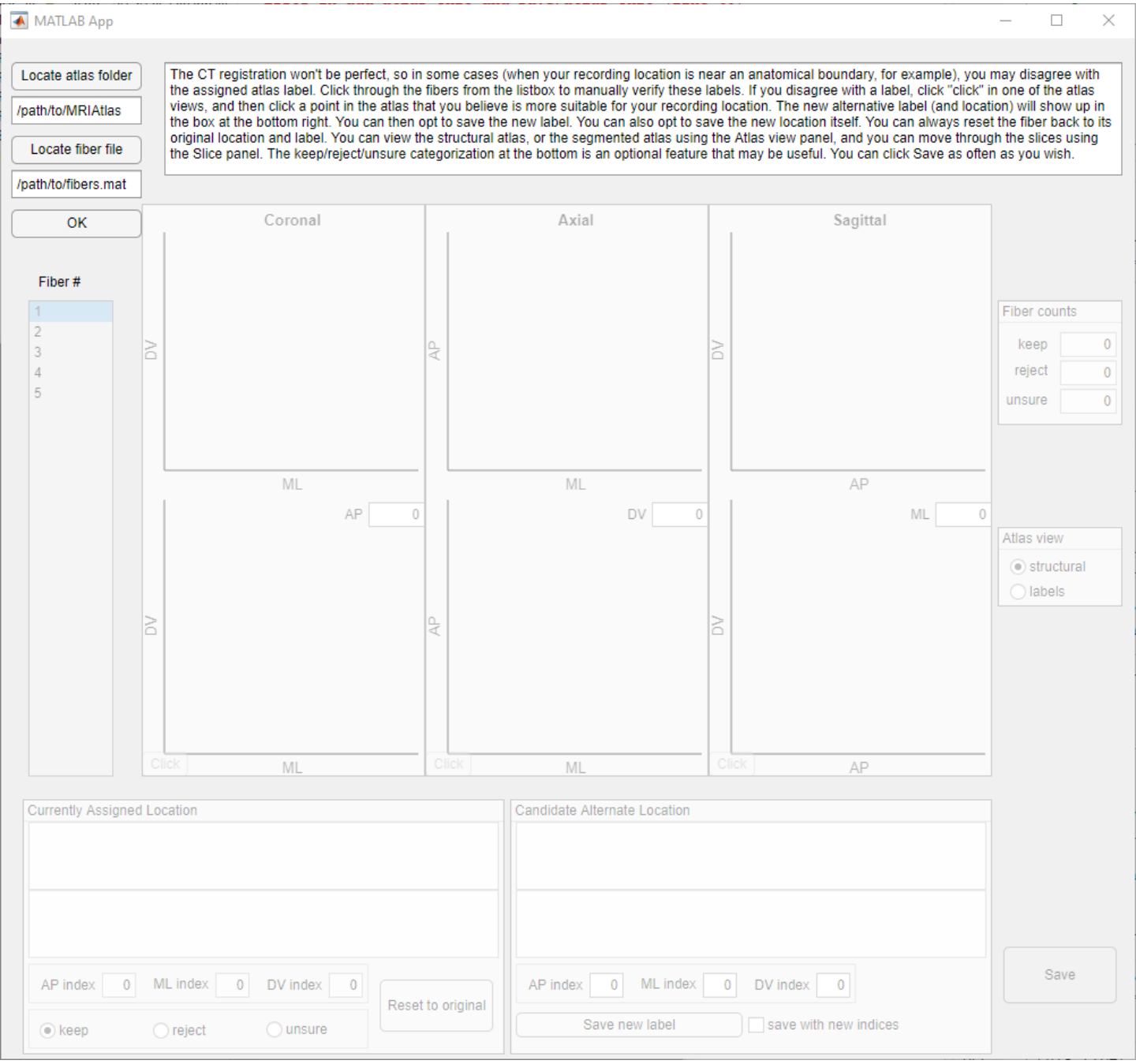
VERIFY LABELS

Now it's time to manually verify that assigned labels. This will open a separate app **verify_atlas_labels.mlapp**. You can run this externally as well and don't need to go through this LOCALIZE_FIBERS app. You can close this app now.



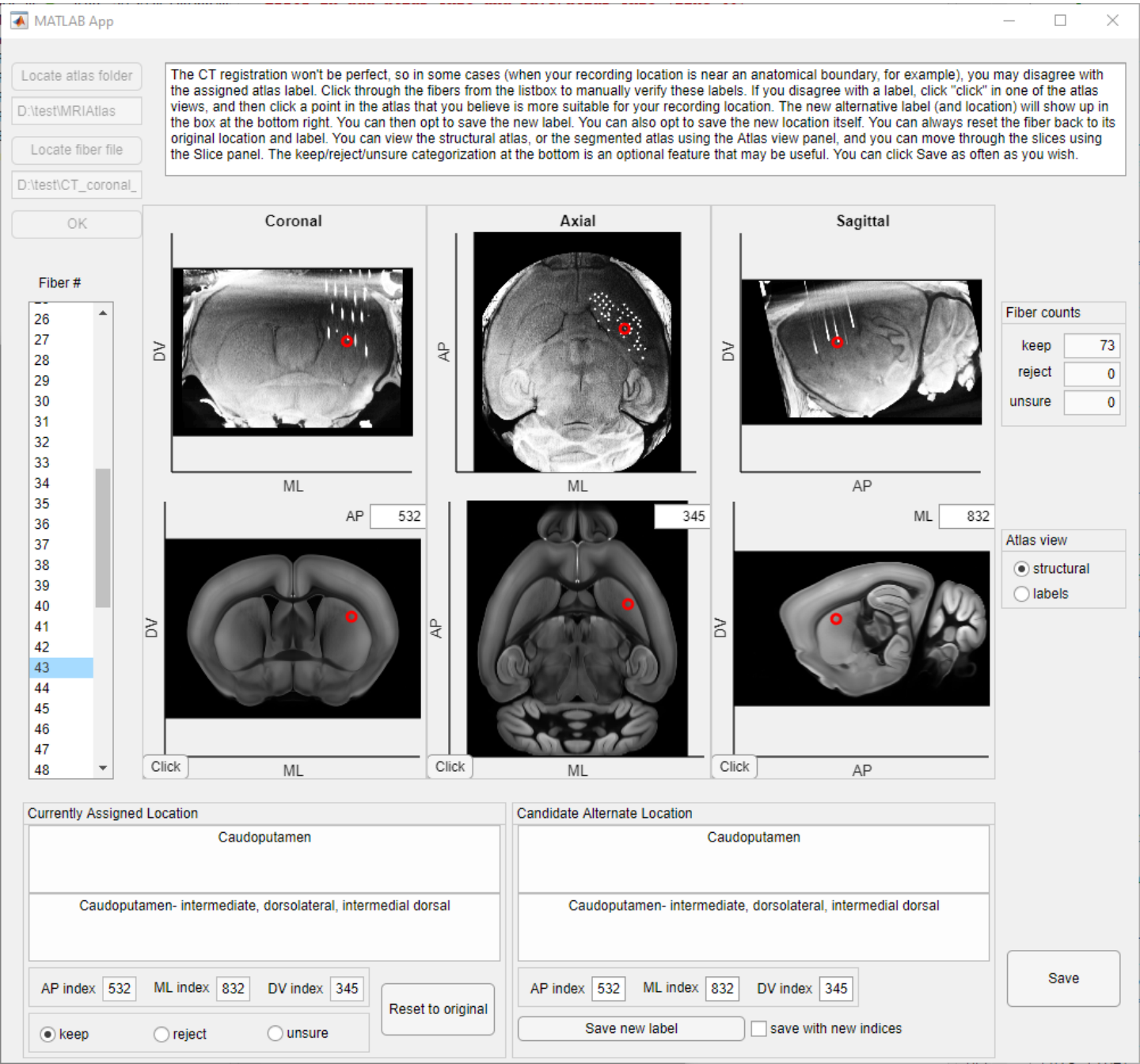
VERIFY LABELS

Locate the atlas folder, as well as the fiber file that was saved in the previous step. The loading may take a little while



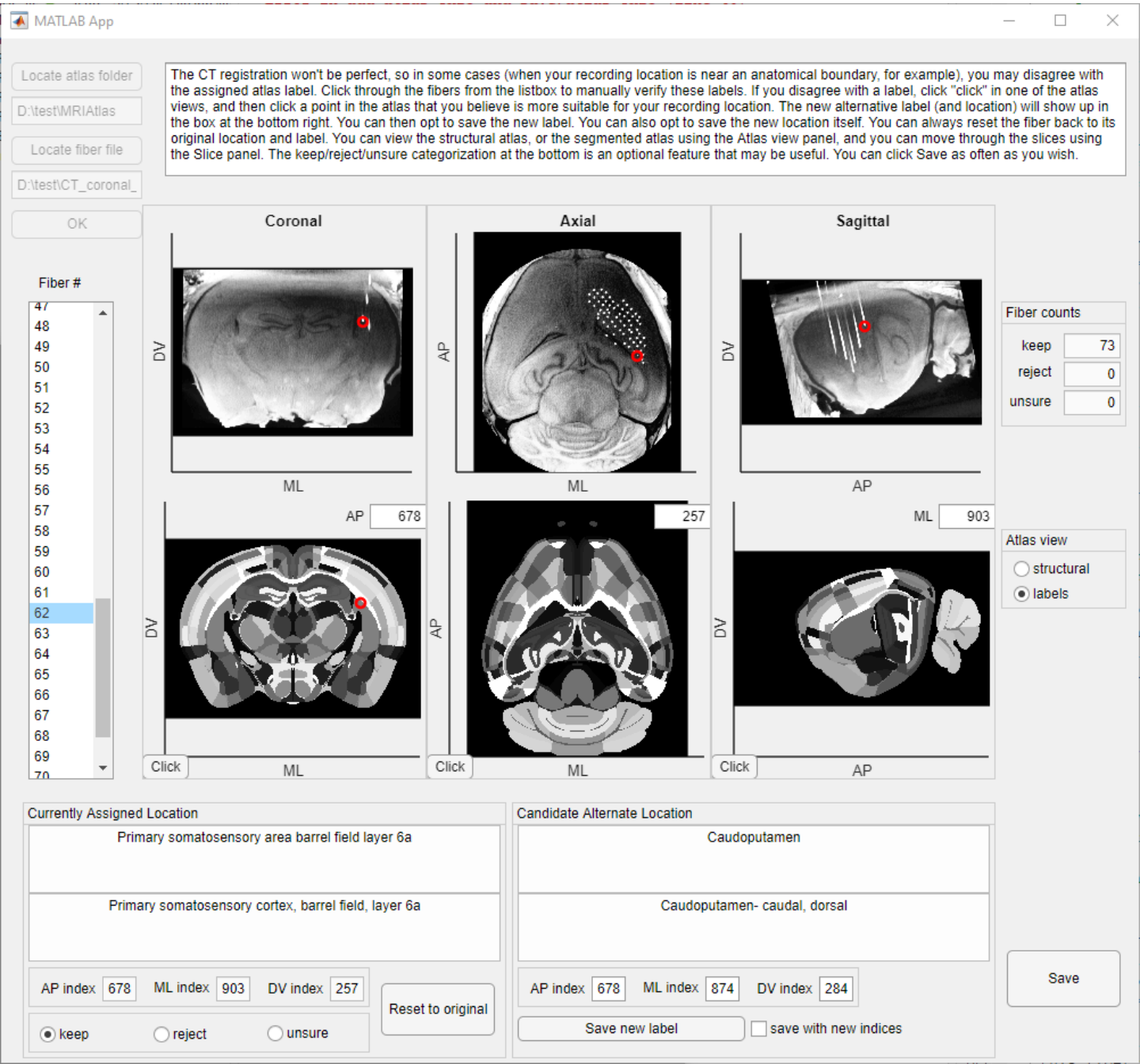
VERIFY LABELS

When the files are loaded, the Fiber # menu will populate (use this menu to select fibers), and some images will pop up. The top row shows the CT with your selected fiber tip highlighted. The bottom row shows the atlas, and you can toggle between the structural view (shown) and the label view (next slide)



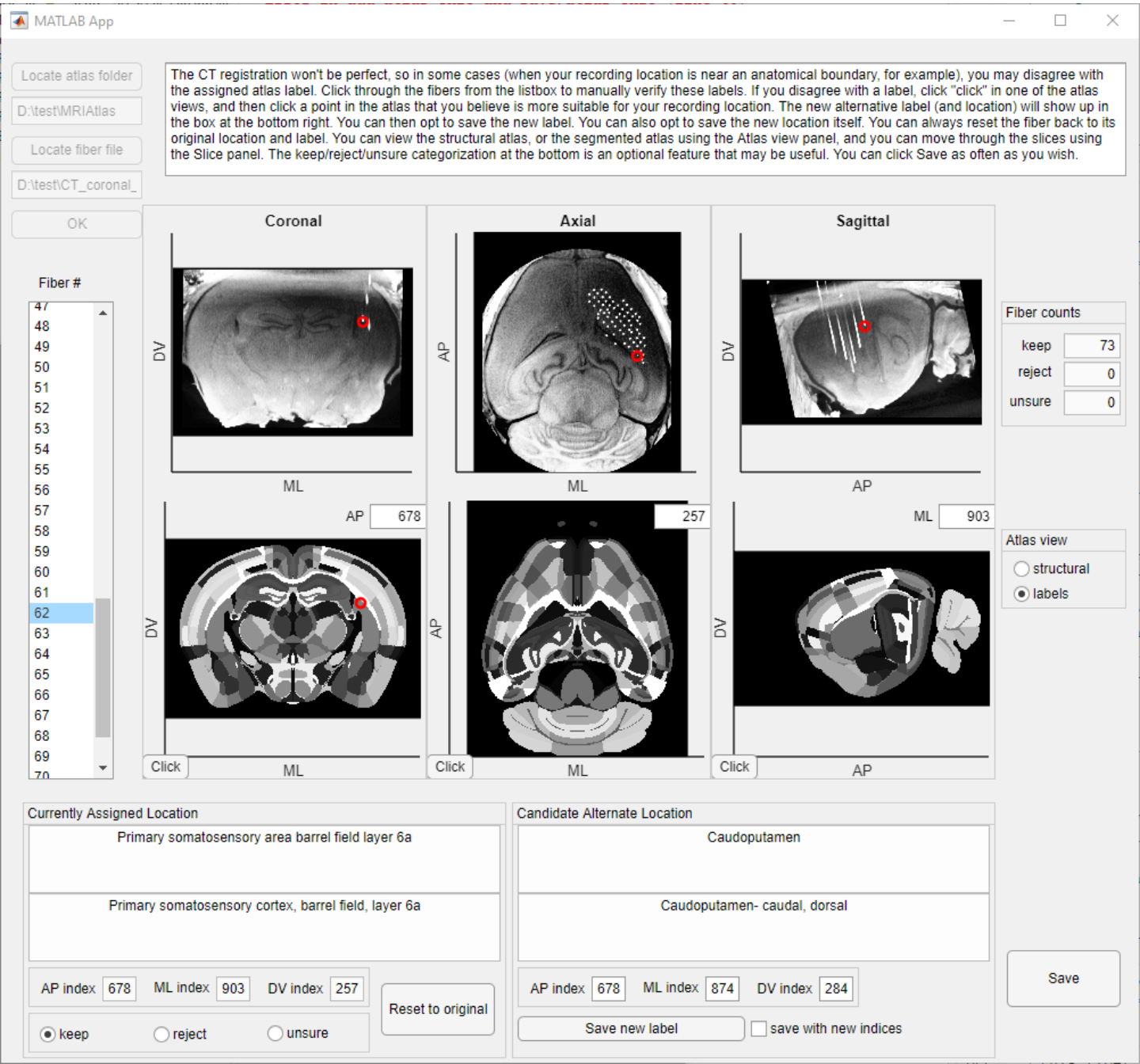
VERIFY LABELS

Atlas label view



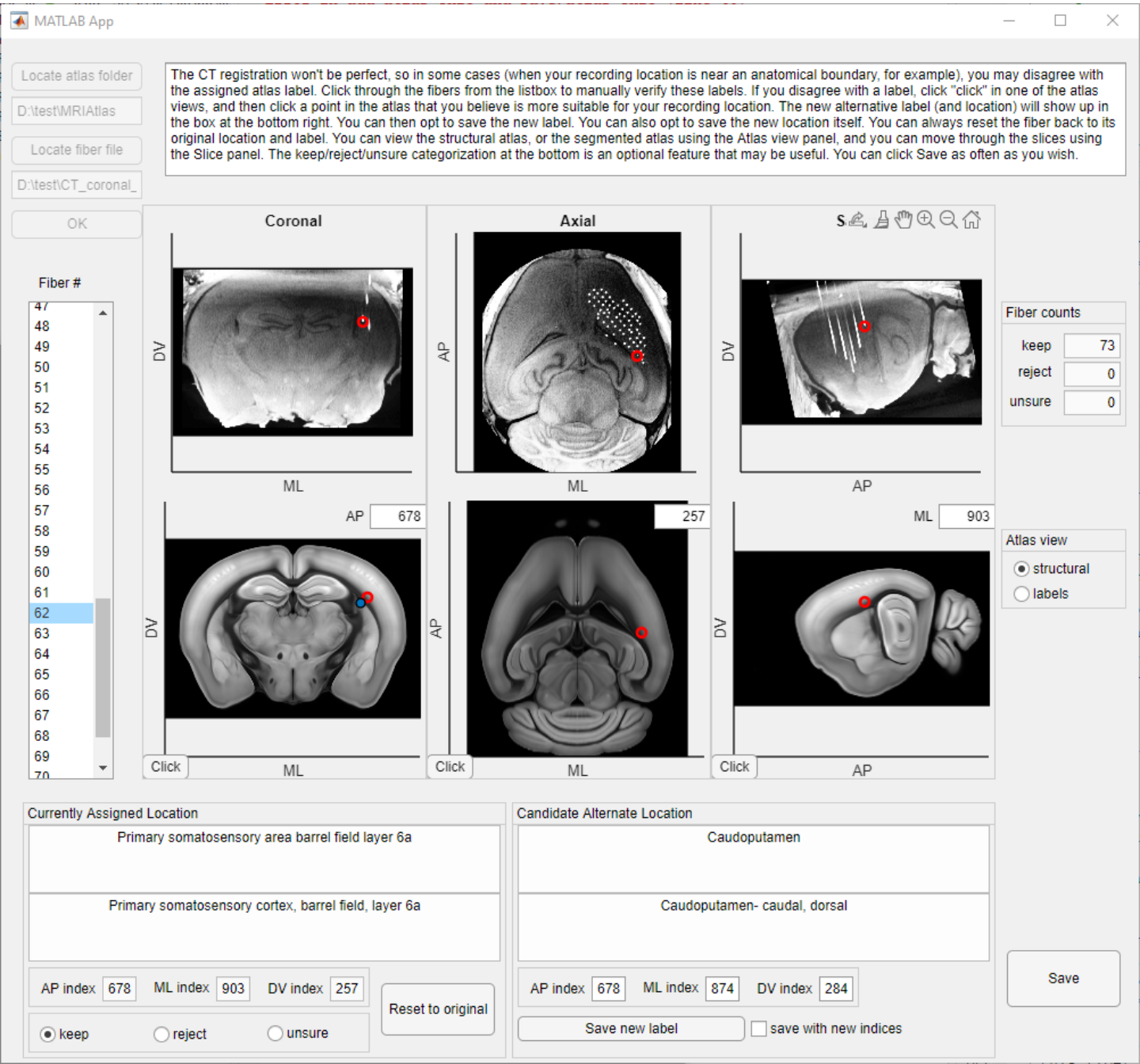
VERIFY LABELS

The bottom left panel shows the currently assigned location. You'll see atlas labels as well as the location. There is also an optional feature that lets you decide whether to keep or reject each fiber. This will add a column to the output table where 1 means keep, 0 means reject, and 0.5 means you're unsure. Running totals are shown in the Fiber counts panel.



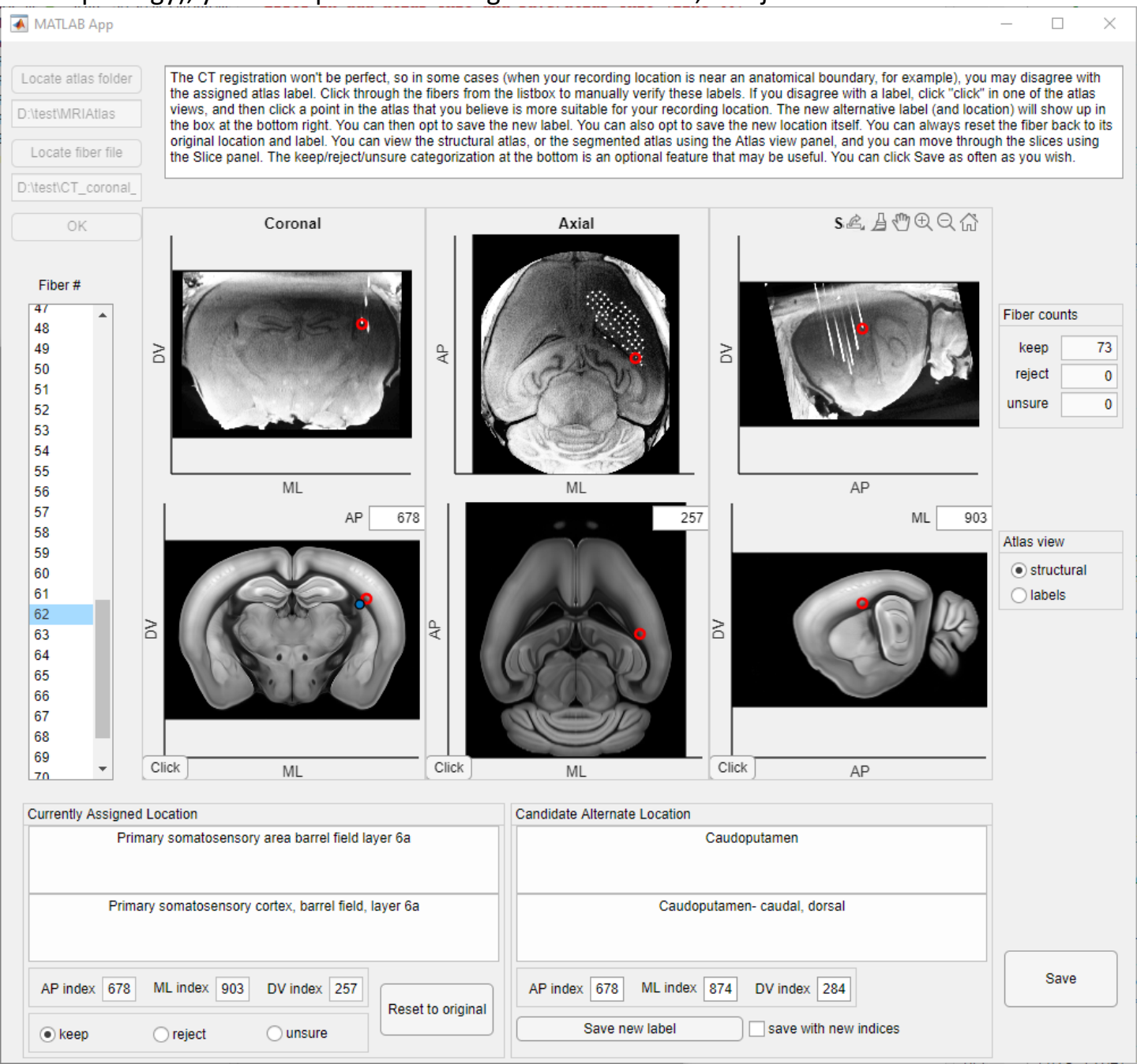
VERIFY LABELS

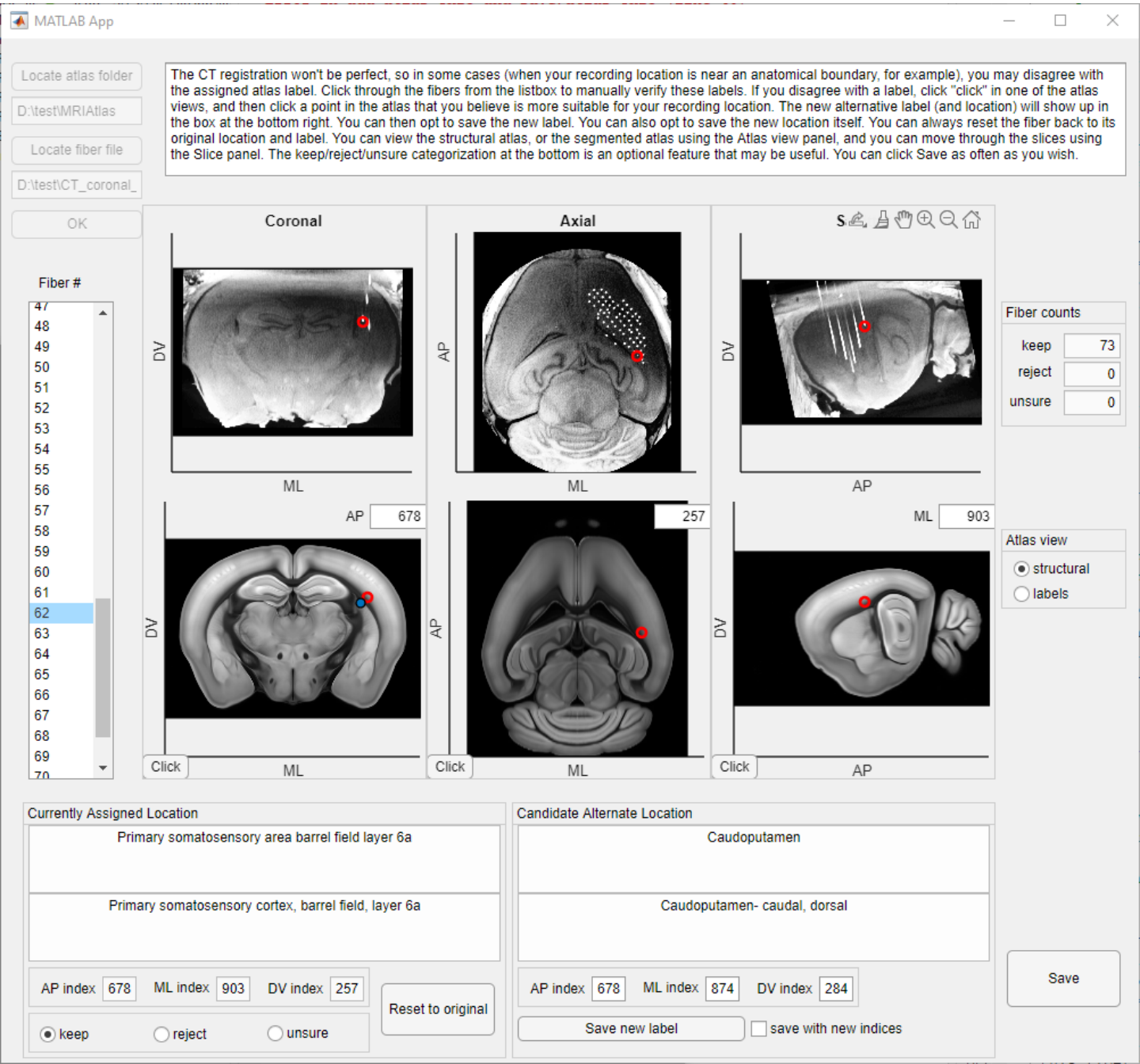
Let's say in this example, you disagree with the original label. Here I clicked "Click" in the Coronal view on the atlas slice, and clicked what I think more accurately captures my fiber location. You can see in the bottom right panel the new labels and coordinate.



VERIFY LABELS

If you agree with the new label, you can save the new label. You can also opt to save the new coordinates too. If you want to preserve the spatial relationships of these fiber locations (note: in this registered atlas space) but want to just edit the anatomical label (as may be appropriate just due to individual differences in brain morphology), you can opt out of saving the new indices, and just save the label itself.



VERIFY LABELS

Save as often as you wish. Close the app when finished. Note that the original fiber table generated in the prior step is saved in the output in a field called fiber_table_orig. Any changes you've done in this app are reflected in the fiber_table field.