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Tissue Mapper

Forked from [Single-Cell ICN Neuron Mapping and 3D Heart Reconstruction with Tissue Mapper](#)Maci Heal¹¹MBF Bioscience**1** *Works for me* dx.doi.org/10.17504/protocols.io.bjykpse

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SUBMIT TO PLOS ONE

ABSTRACT

This protocol describes the process of using the Tissue Mapper software to map single neurons of the intrinsic cardiac nervous system, annotate or "trace" key cardiac anatomy on select histological sections of an image volume, and to visualize the mapped neurons and traced anatomy in a 3D reconstructed heart.

DOI

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FORK NOTE

FORK FROM

[Forked from Single-Cell ICN Neuron Mapping and 3D Heart Reconstruction with Tissue Mapper, Clara Leung](#)

KEYWORDS

3D heart reconstruction, Intrinsic cardiac neurons, Intrinsic Cardiac Nervous System, Mapping, Image volume, Image stack, Tissue Mapper, Heart, Rat, KESM

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MATERIALS TEXT

MATERIALS

[Tissue Mapper](#) MBF Bioscience[Image stack \(.jpx\)](#) Contributed by users

DISCLAIMER:

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Software installation setup

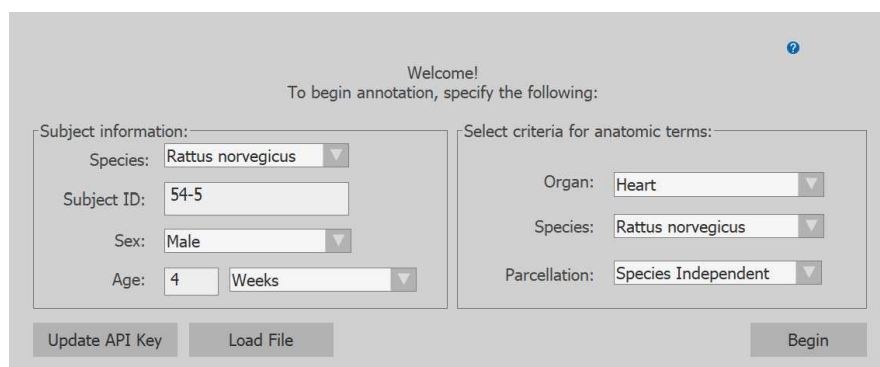
- 1 Have Tissue Mapper installed and licensed on your workstation of choice through an MBF Bioscience representative.

Tissue Mapper 2020.1.1 [↗](#)
by MBF Bioscience

- 2 After launching Tissue Mapper, set an API key to your profile to pull the ontology list from SciCrunch to annotate your organ of choice with curated SPARC anatomy terms.

Segmentation: Markers and Contours

- 3 **Metadata setup.** To begin, create an empty document by filling in the subject information of your sample with the species, sample ID, sex, and age on the left side of the dialog. Then specify the species and organ in which you are annotating on the right side of the dialog to access the curated SPARC anatomy terms and press "Begin" when you are done.

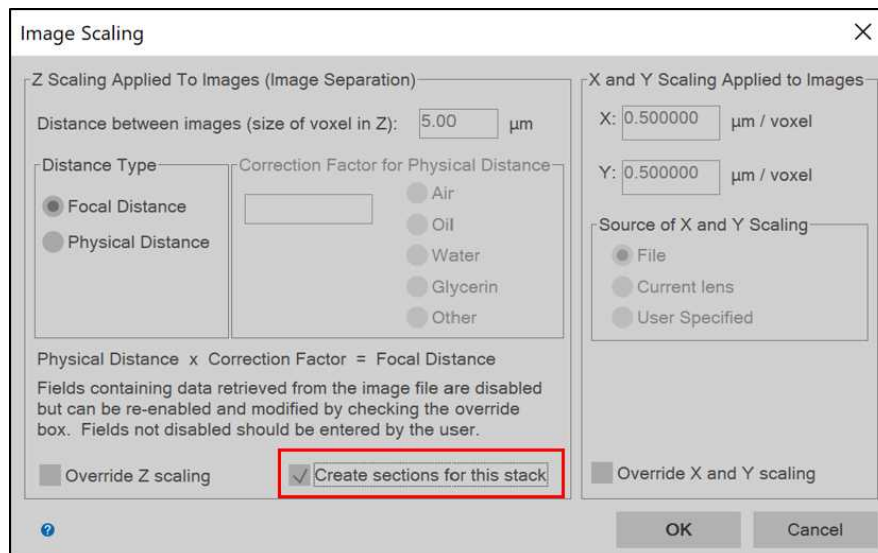


More information on SPARC vocabulary services can be found here:

<https://www.mbfbioscience.com/help/tissuemapper/Default.htm#cshid=8044>

- 3.1 Manually inputting information about your sample only occurs once when you first create an empty document to annotate your sample. After you save your work as an .xml file and load it into the program, the metadata dialog will autopopulate and you can resume your work.

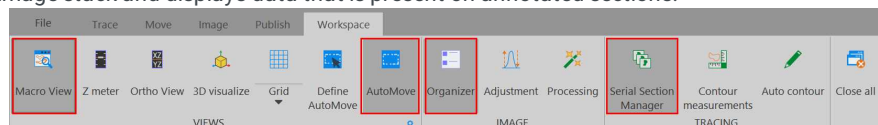
- 4 **Configuring and opening the image file.** Open the image stack (.jpx) through the "File" ribbon and under the "Open" menu, select "Image stack," and select the desired .jpx file. A dialog will appear to verify or adjust the XYZ scaling properties of the image stack. If the scaling properties are correct and do not need adjustment, check the "Create sections for this stack" to annotate individual sections and click "OK" to load the image stack.



Instructions for adjusting the image scaling can be found here:

<https://www.mbfbioscience.com/help/tissuemapper/Default.htm#cshid=1185>

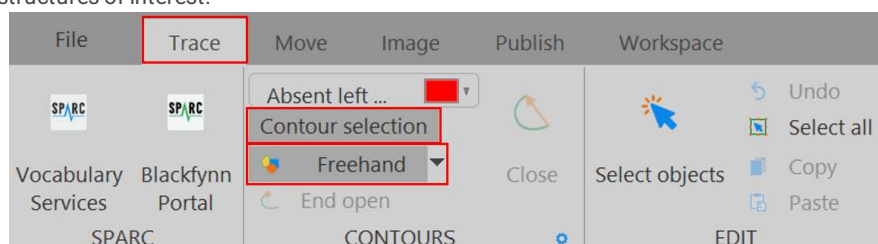
- 5 Workspace setup.** For ease of annotation, it is recommended to set up your workspace with dockable windows. Under the "Workspace" ribbon, enable the "AutoMove" function and dock the "Macro view," "Image organizer," and the "Serial Section Manager." The AutoMove feature will define a working area and automatically reposition your image to the next adjacent region when your contour moves beyond the bounds of the working area. The "Macro view" acts as a mini-map in a separate window to show your location on the image section and to also take you to any region of interest. The "Image organizer" allows you to show, hide, save, or discard the image data and is especially helpful for visualizing the contours in 3D. Lastly, the "Serial Section Manager" enables movement through individual sections of the image stack and displays data that is present on annotated sections.



More information on setting up dockable windows can be found here:

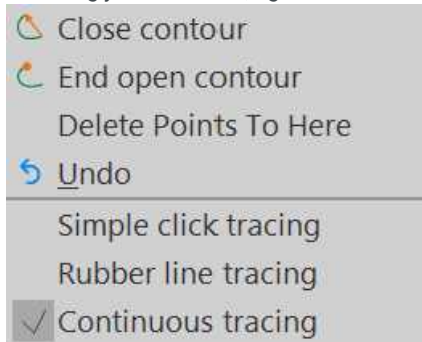
https://www.mbfbioscience.com/help/tissuemapper/Default.htm#About/WindowsSetup.htm%3FTocPath%3DWorkspace%2520overview%7C___4

- 6 Contour preparation.** To begin contouring or tracing anatomical features, go to the "Trace" ribbon and in the "CONTOURS" submenu, click "Contour selection" to display the list of curated SPARC anatomy terms. Under the "Contour selection" button, you will want to set tracing options to "Freehand" to manually contour the anatomical structures of interest.



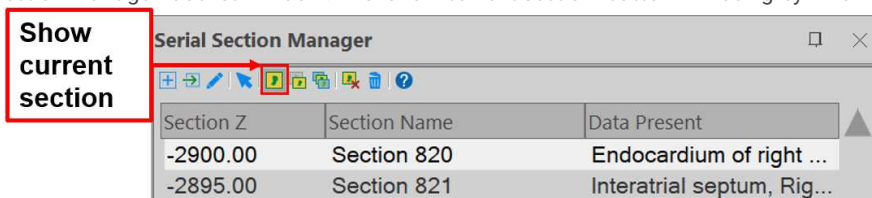
- 7 Section annotation.** Select the desired anatomical structure that you wish you annotate under the "Contour Selection" list. Navigate to a section of image data with the "Serial Section Manager," zoom in on the image as needed, and contour your desired structure by clicking along the structure to place points and draw lines. Alternatively, you can also continuously draw a contour by right clicking and selecting the "Continuous tracing" option. This method of tracing works best when paired with a Wacom tablet or other digital tablet drawing device. When you are finished with

annotating your structure, right click to select "Close contour" or "End open contour."



Right-click menu for contouring and tracing options

- 8 **Additional section annotation.** Move to a different section in the image stack with the Page up (PgUp) or Page down (PgDn) keys and repeat step 9. As the image sections become more populated with contours and tracing data, it will be best to only keep the contours on each section visible by clicking the "Show current section" button in the "Serial Section Manager" docked window. The "Show current section" button will look grey when it is selected.



More information on the workspace setup and annotation instructions can be found here:

[https://www.mbfbioscience.com/help/tissuemapper/Default.htm#About/TMgetStarted.htm%3FTocPath%3DCreate%2520annotations%2520\(contours\)%7C_____1](https://www.mbfbioscience.com/help/tissuemapper/Default.htm#About/TMgetStarted.htm%3FTocPath%3DCreate%2520annotations%2520(contours)%7C_____1)

- 9 **Marker placement and cell mapping.** In the Markers toolbar, select a marker that you like to use to best represent the cells (neurons) or region of interest to be pinpointed. To rename it, right click and select "Rename" to specify what the selected marker represents. To change the color of a marker, right click and select "Change Marker Color" and you can choose from preset colors or a user-defined color, click "OK" when you are finished. Next, select your desired marker and go to a section that has the cell or area of interest and click on the image to place a marker. Repeat this step until all your cells of interest are mapped.



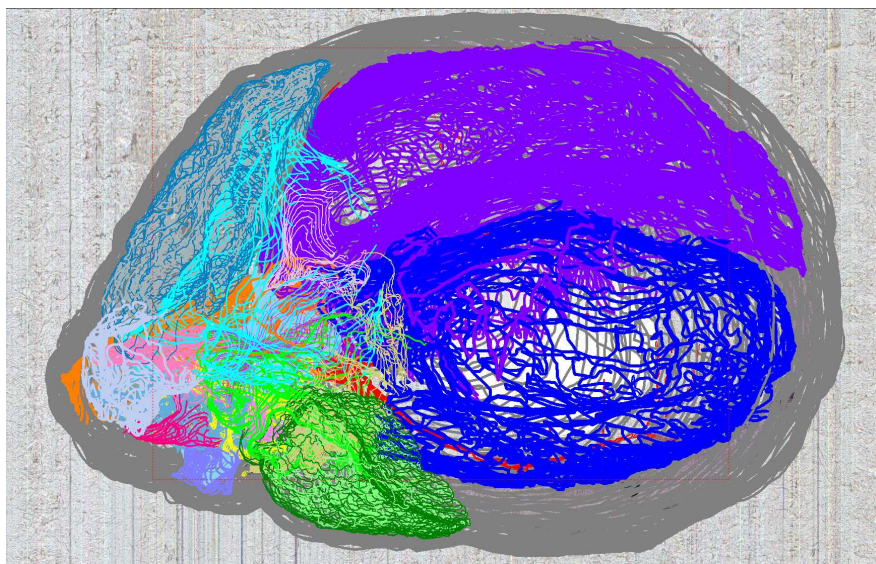
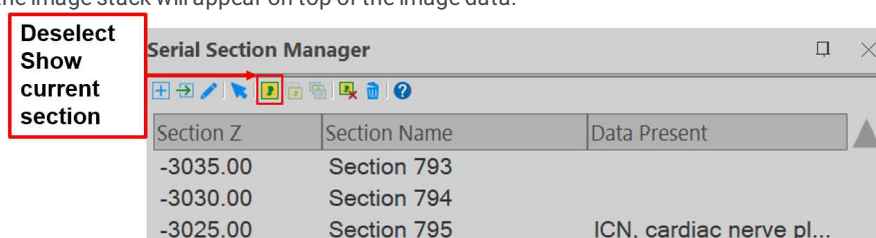
Right-click menu for Markers toolbar options

- 10 **Saving your work.** To preserve your progress in mapping and annotation, go to the "File" ribbon and under the tab "Save as," select "Data File" and save your work as an XML document file (.xml). The associated metadata from the initial dialog box will autopopulate when you load the file to resume your work.

3D visualization

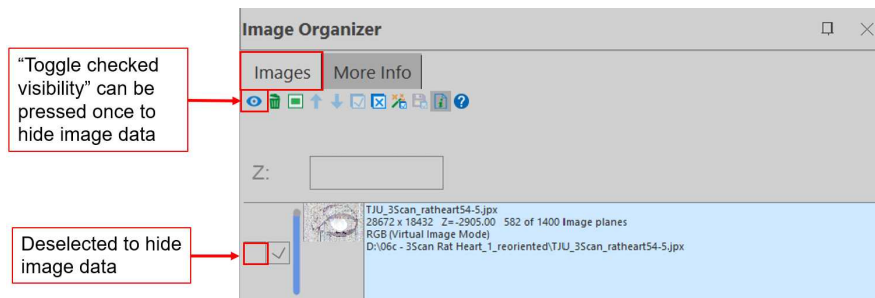
- 11 **3D visualization of mapped neurons and contours.** Visualization of the contours and mapped neurons of the entire annotated image stack in the 3D viewer are achieved through three steps and will be described in sub-points below.

- 11.1 **First,** deselect the "Show current section" button in the docked "Serial Section Manager window." The "Show current section" button will not look grey and a maximum projection of the contours throughout the image stack will appear on top of the image data.

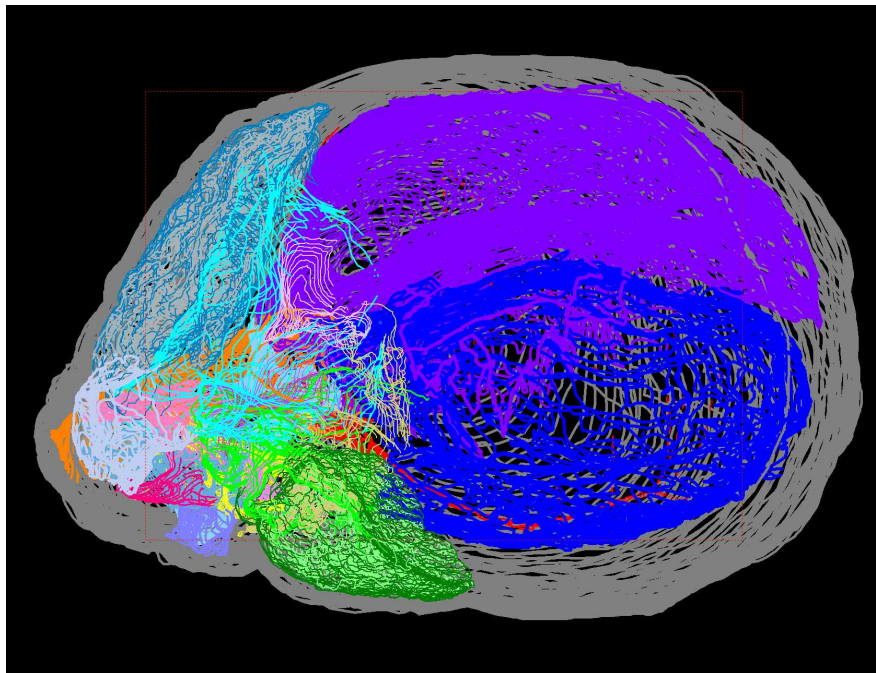


Maximum projection of all the contours overlaying the image data in the tracing workspace after deselecting the "Show current section" button.

- 11.2 **Second,** in the docked "Image Organizer" window under the "Images" ribbon, deselect the eye or press the "Toggle Checked Visibility" button to hide the image data. In the main tracing workspace, only the contours and markers should be present with a black background.

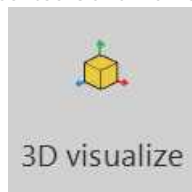


Two options for hiding the image data.

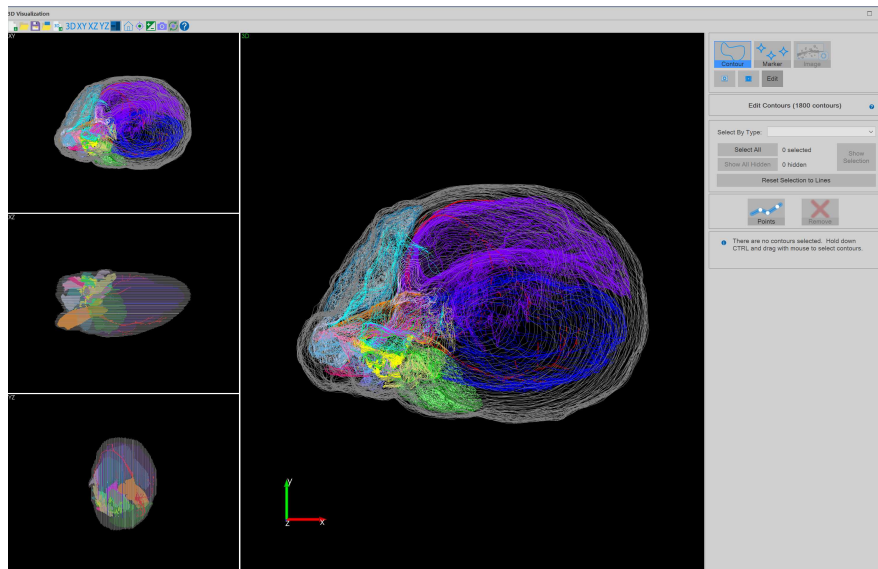


How the main workspace appears after hiding the image data to show only the contours and markers.

- 11.3 **Third**, to visualize all the contours and markers in the 3D viewer, press the "3D Visualize" button under either the "Trace" or "Workspace" ribbon. A separate window will open to allow interaction with the 3D reconstructed heart model. In the 3D viewer, the user can selectively show, hide, and edit specific contours and markers, create video clips of the data, and export the 3D representations.



3D visualize button can be found in either the Trace or Workspace ribbon.



3D Visualization window for manipulation and interaction of contours and markers in the 3D space.

More information on 3D Visualization capabilities are described here:

https://www.mbfbioscience.com/help/tissuemapper/Default.htm#3Dvisualization/3D_Tour.htm%3FTocPath%3DWorkspace%2520overview%7C____2

- 11.4 3D visualization of mapped neurons, contours, and image data.** It is also possible to load the 3D image data along with the markers and contours. To have the image stack load with the contours, go to the docked "Image Organizer" and under the "Images" ribbon, make sure to have the eye checked to show the image data.

