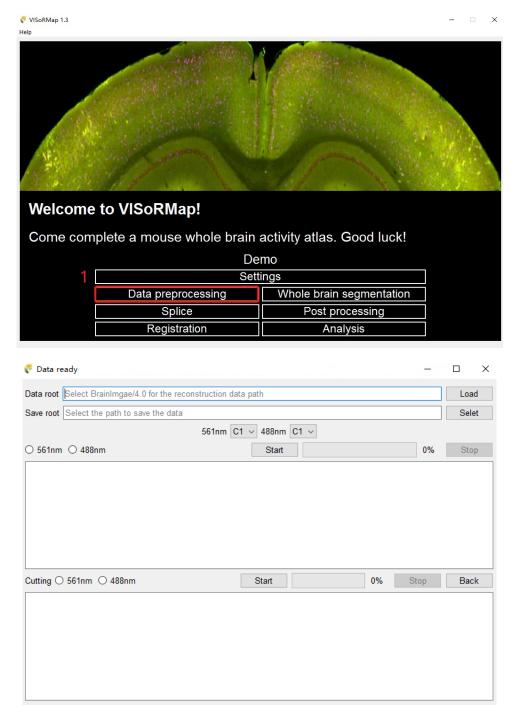
# VISoRMap Software User Guide

### 1. Data preparation



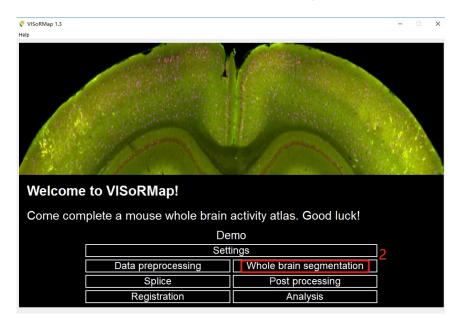
(1). Data preparation for 3D brain slices: The data root is BrainImage/4.0 after reconstruction. Select channels corresponding

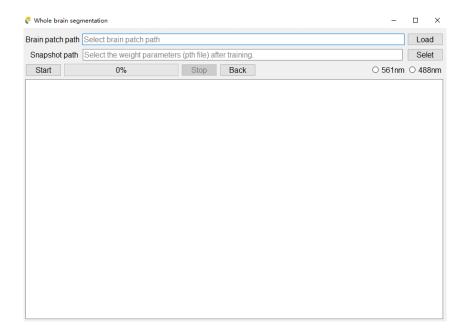
to 561nm and 488nm. By default, the 3D data size is (3500, 2500, 64) for (x, y, z) dimensions. Click **Start** to begin the preparation process for either 561nm or 488nm channels. **Stop** will change to **Finished** when a single data preparation is complete.

(2). After the 3D data preparation is complete, start cropping the data. Crop one data into 70 subvolume brain images with dimensions (256, 256, 64) for (x, y, z). Perform the operation as before, and click **Back** to return to the main page after completion.

#### 2. Whole-brain immunolabeled signal segmentation

After cropping, start the whole-brain automated segmentation of c-Fos+ and spontaneous fluorescence signals in the whole brain.



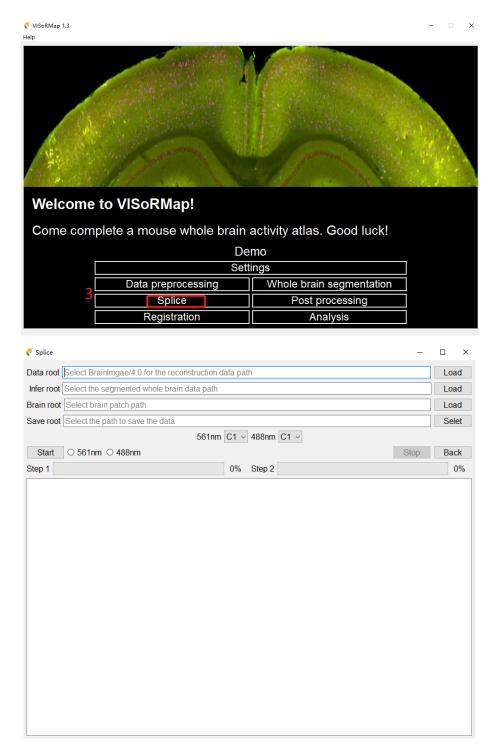


Brain patch path refers to the parent path of the Subvolume brain images after cropping. Snapshot path is the path for loading the trained parameters of the neural network. The default path for the trained parameters is:

pages\Whole\_brain\_seg\swin\_T\_checkpoint\_22\_01\_09.

## 3. Whole brain splicing of segmentation results

After completing the whole-brain signal segmentation for the dual channels, it is necessary to restore the segmentation results of the Subvolume brain images to the original size of the brain slices.



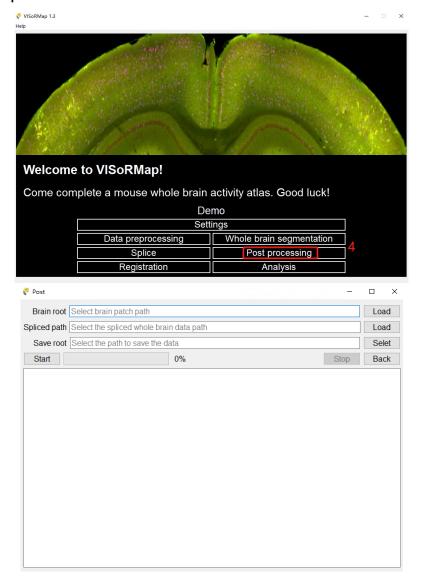
The data root path is as described above. The Infer root refers to the path of the whole-brain dual-channel segmentation results, and the Brain root refers to the parent path of the Subvolume brain images.

The stitching process consists of a total of 2 steps. The first step

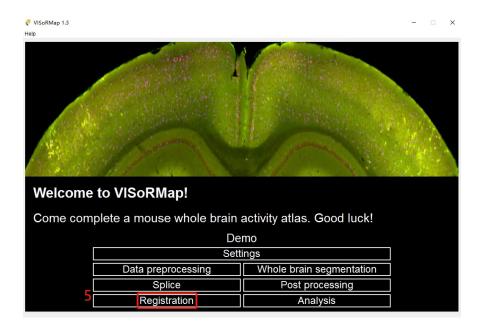
is to convert the 3D segmentation results to 2D and stitch them in a 2D layer-wise manner for the whole brain (3500, 2500). The second step is to combine all stitched 2D segmentation results to generate a 3D volume (3500, 2500, 64).

### 4. Post processing

After completing the whole-brain signal segmentation stitching, post-processing operations are performed. The splice path refers to the parent path of the stitched results.

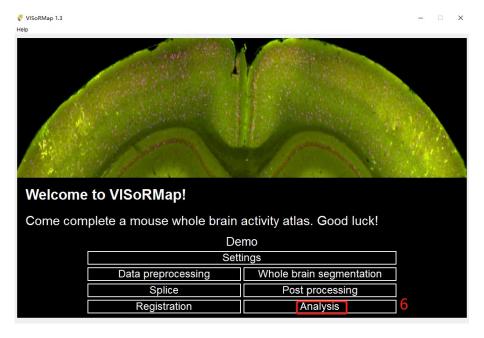


#### 5. Registration



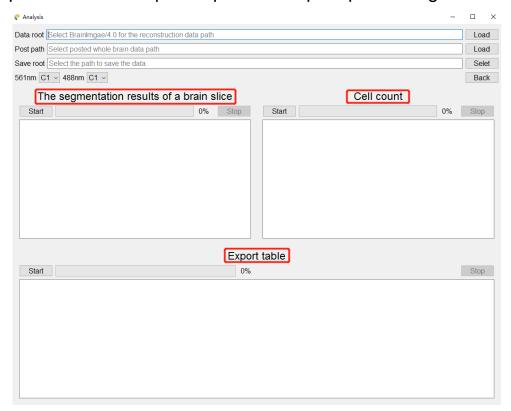
This module can register the whole brain to the Allen Common Coordinate Framework atlas. The operation is the same as above, and when finished, the **Stop** button displays **Finished**.

#### 6. Cell counts



After registration and post-processing are completed, the

segmentation results need to be mapped to the Allen CCFv3. The Post path refers to the parent path of the post-processing results.



- (1) The module is used to restore the size of the segmentation results from (3500, 2500, 64) to (3500, 2500, 75), which corresponds to a thickness of 300  $\mu$ m (75 x 4  $\mu$ m).
- (2) Cell count. This module is used to count cells in the whole brain signal segmentation results.
- (3) Export table. This module is used to generate a csv file that can be imported into Freesia, which records the centroid coordinates of each segmented c-Fos signal.

## Exported file

2022/11/13 22:37
2022/11/13 22:19
2022/11/20 16:09
2022/11/19 18:11
2022/11/22 17:14
2022/11/23 22:26
2022/11/20 20:36
2022/11/19 20:47
2022/11/19 17:16
2022/11/20 10:00
2022/11/19 18:49
2022/11/18 19:41