DynamicST Assist Operation Manual

Software Documentation

DynamicST Assist includes two functions: DynaBlot image and microscope image Alignment, fiducial alignment and tissue detection.

DynaBlot image and microscope image Alignment is used to align microscope image with DynaBlot image.

In this function,the following files are required：

1. DynaBlot image
2. Microscope image

Fiducial alignment and tissue detection is used to adjust DynaBlot image and detect tissue area to obtain the location information of each spot and whether it is covered by tissue area.

In this function, DynaBlot image is required.

Operation Manual

The operation of DynamicST Assist consists of three parts, start software, DynaBlot image and microscope image alignment and fiducial alignment and issue detection.

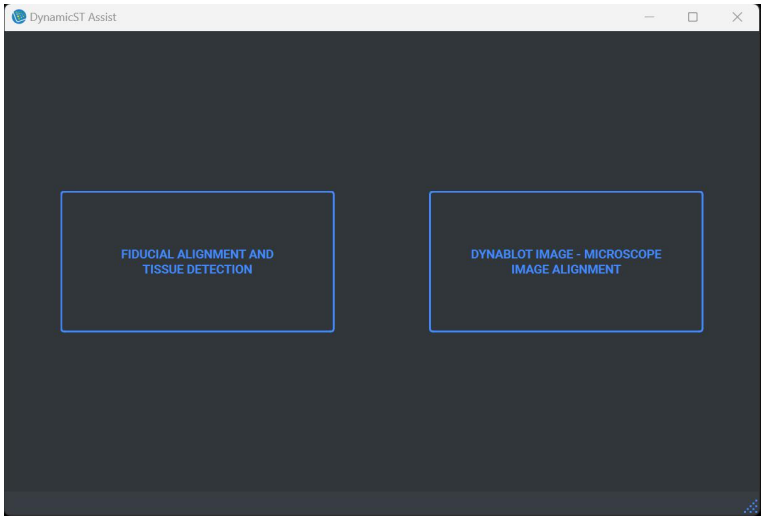
During the operation, you can zoom the image by scroll the mouse wheel to, or drag the image by pressing the right mouse button.

1. Start software

   In this part, you have two choices.The operation process is as follows:

   (1) If you only have DynaBlot image, you need to click button “DYNABLOT IMAGE - MICROSCOPE IMAGE ALIGNMENT” and software will skip DynaBlot image and microscope image alignment. In this part, you can adjust DynaBlot image and detect tissue area.

   (2) If you have DynaBlot image and microscope image of the same tissue and need to align the two image, you need to click button“FIDUCIAL ALIGNMENT AND TISSUE DETECTION” and software will skip DynaBlot image and microscope image alignment. In this part, you can align DynaBlot image and microscope image. After alignment, you need to continue to fiducial alignment and issue detection.



2. DynaBlot image-microscope image alignment

In this part, you can align DynaBlot image and microscope image. The operation process is as follows:

(1) Load images

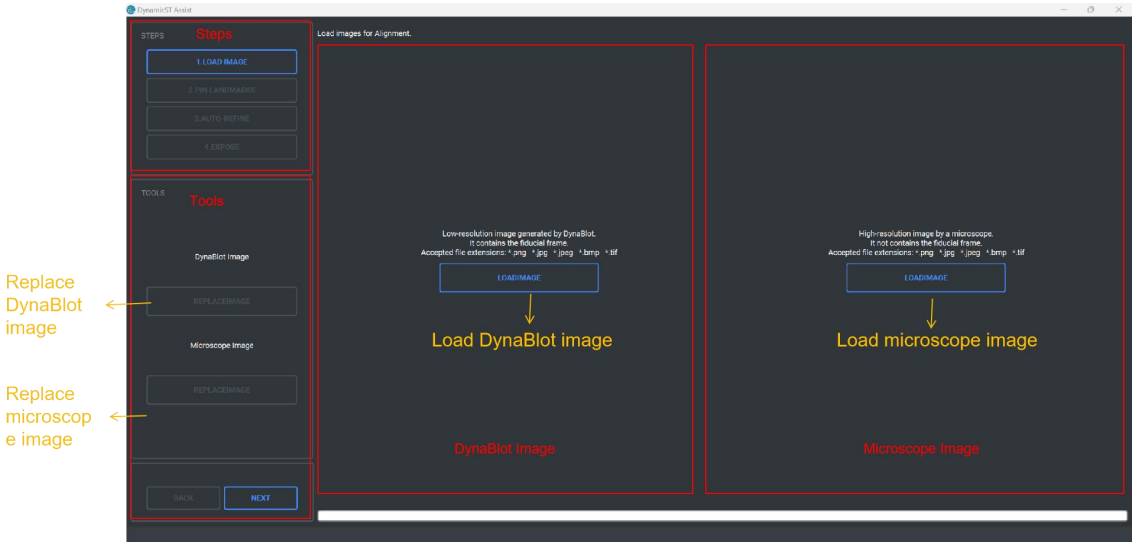
A. Click button “LOADIMAGE” on the left to load DynaBlot image, the DynaBlot image will be flipped horizontally.

B. Click button “LOADIMAGE” on the right to load microscope image.

C. Click button “REPLACEIMAGE” on the top of “TOOLS” to replace DynaBlot image, the DynaBlot image will be flipped horizontally.

D. Click button “REPLACEIMAGE” on the bottom of “TOOLS” to replace microscope image.

E. Click button ”NEXT” to Alignment image step.

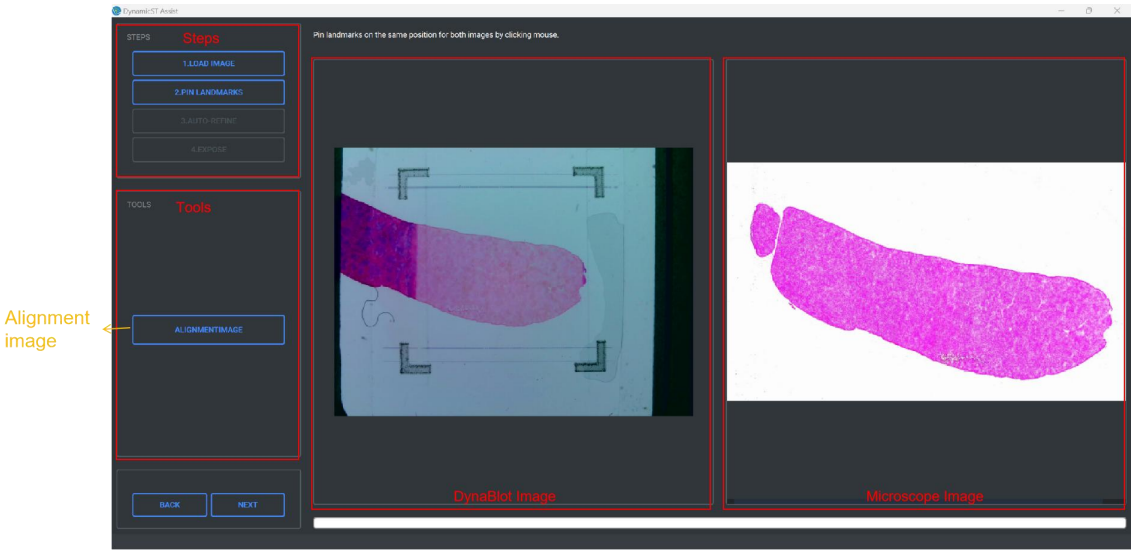


(2) Alignment image

A. Pin landmarks on the images by clicking the mouse.

B. Click button “ALIGNMENTIMAGE” to alignment microscope according to landmarks.

C. Click button ”NEXT” to Auto refine step.



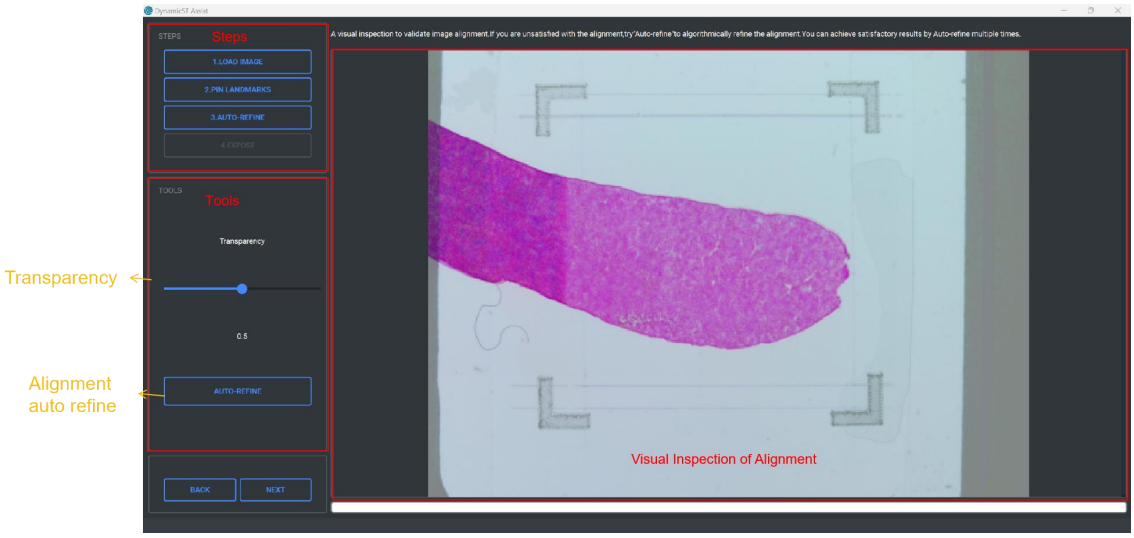
(3) Auto refine

A. Adjust the Slider “Traqnsparency” to visual inspect the result of alignment.

B. Click button “AUTO-REFINE” to automatically fine tune the result of alignment.

C. If the fine-tuning result is unsatisfactory, you can click it several times until the result is satisfactory

D. Click button ”NEXT” to Expose alignment result step.



(4) Expose alignment result

A. Click button ”CONTINUE TO MANUAL FIDUCIAL ALIGNMENT” to skip manual alignment of DynaBlot image with fiducial frame.

B. In this step, microscope image after alignment will be saved to the path of the microscope image and named as “microscope image name_tissue_image.tif”.

3. Manual alignment of DynaBlot image with fiducial frame

In this part, you can adjust DynaBlot image and detect tissue area.The operation process is as follows:

(1) Load image

A. Click button “LOADIMAGE” to load DynaBlot image.

B. Click button “REPLACEIMAGE” on the “TOOLS” to replace DynaBlot image.

C. Choose chip type in combobox.

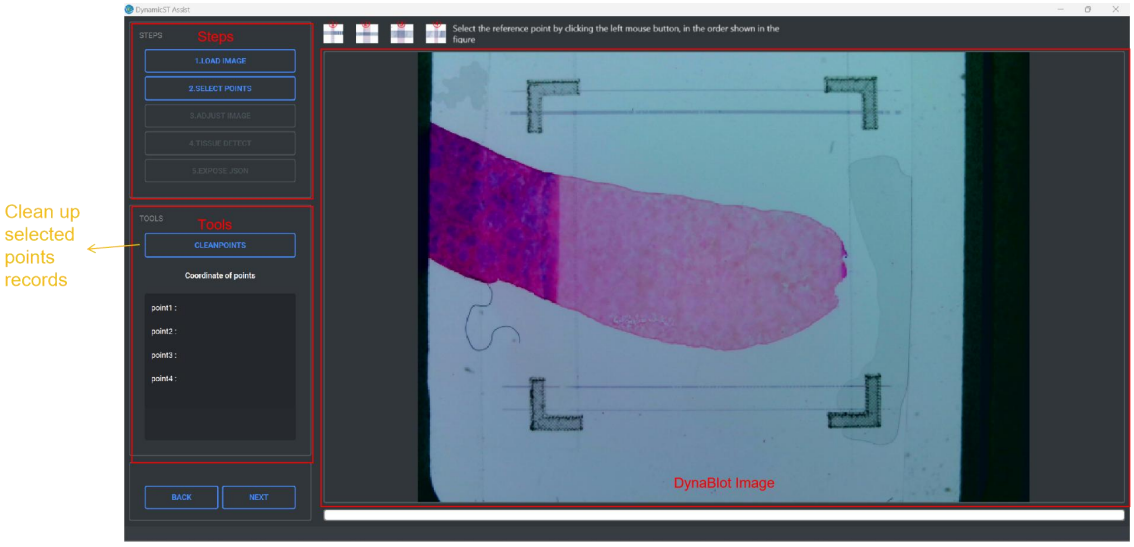
D. Click button ”NEXT” to Select points step.

(2) Select points

A. Select the points by clicking the mouse, in the order shown in the figure.

B. Clean up the records of the selected points by clicking button “CLEANPOINTS”.

C. Click button ”NEXT” to Adjust image step.



(3) Adjust image

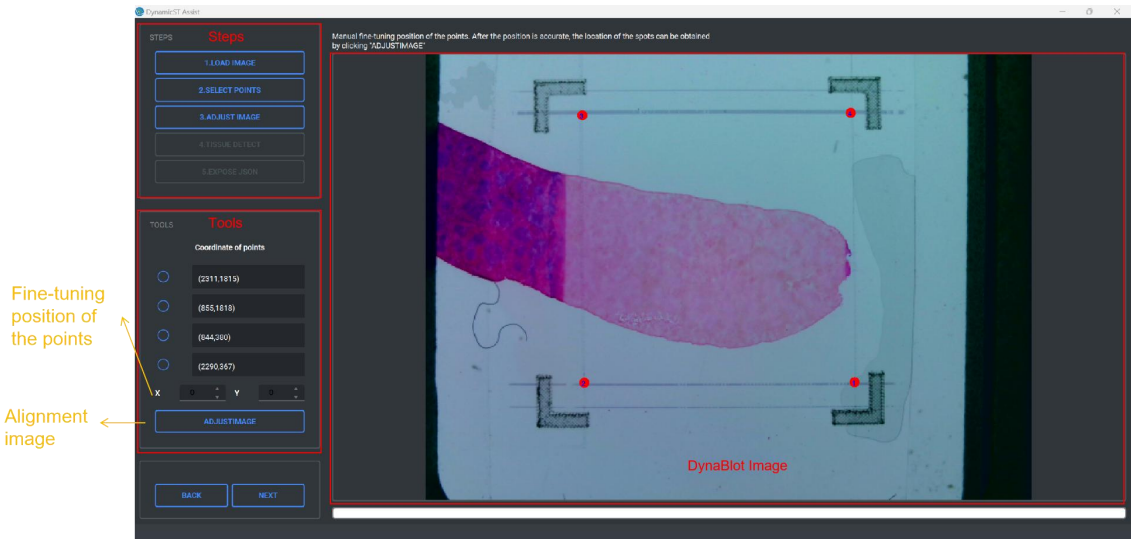
A. If the coordinates of the points are not accurate, you can select the point to be tuned, and then fine tune the coordinate of this point in the x and y directions in the spinbox.

B. Click button “ADJUSTIMAGE” to adjust DynaBlot image.

C. The spots corresponding to the DynaBlot image will be displayed on the image.

D. In tihs step, if you have aligned DynaBlot image and microscope image, the microscope image will be adjusted in the same way as DynaBlot image and replace “microscope image name_tissue_image.tif”.

E. Click button ”NEXT” to Detect tissue area step.



(4) Detect tissue area

A. If you have aligned DynaBlot image and microscope image, select image for tissue area detection, microscope image or DynaBlot image can be selected.

B. Modify Sensitive slider to set parameter of tissue area detection algorithm.

C. Choose image channel of trissue area detection algorithm.

D. Click button “TISSUEDETECTION” to automatically detect tissue area.

E. Modify Transparency slider can modify the transparency of spots.

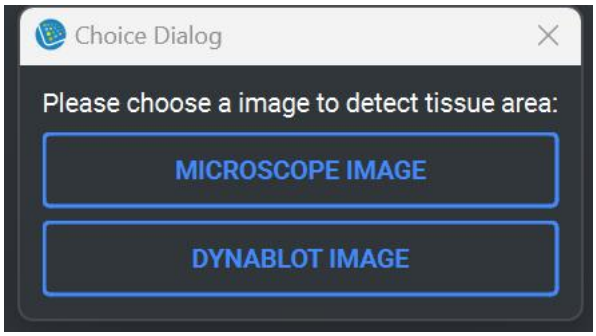
F. If the result of automatic detection is not accurate, you can fine tune the tissue area through the brush function.

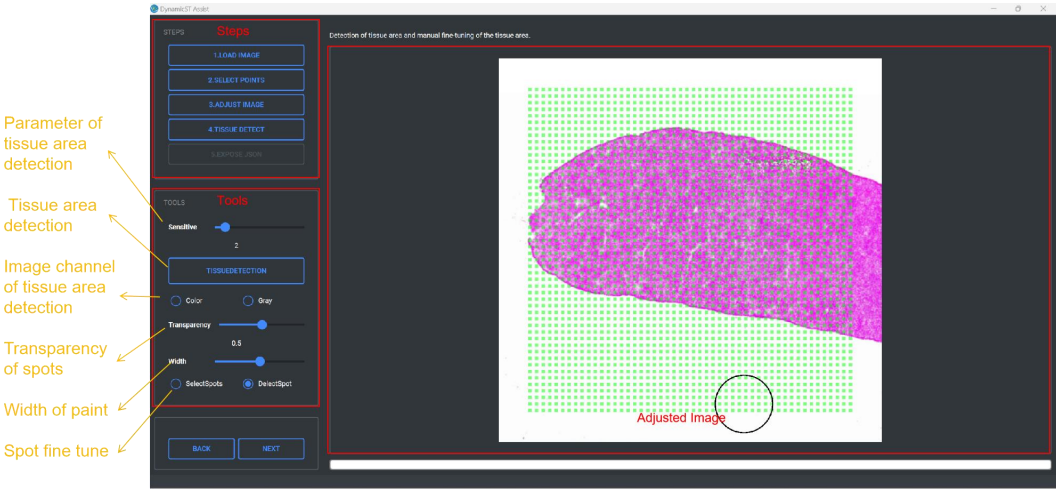
   a. Select “SelectSpot” to add spots covered by tissue to the image.

b. Select “DeleteSpot” to delete spots covered by tissue to the image.

c. Modify Width slider to modify width of brush.

G. Click button ”NEXT” to Expose json step.





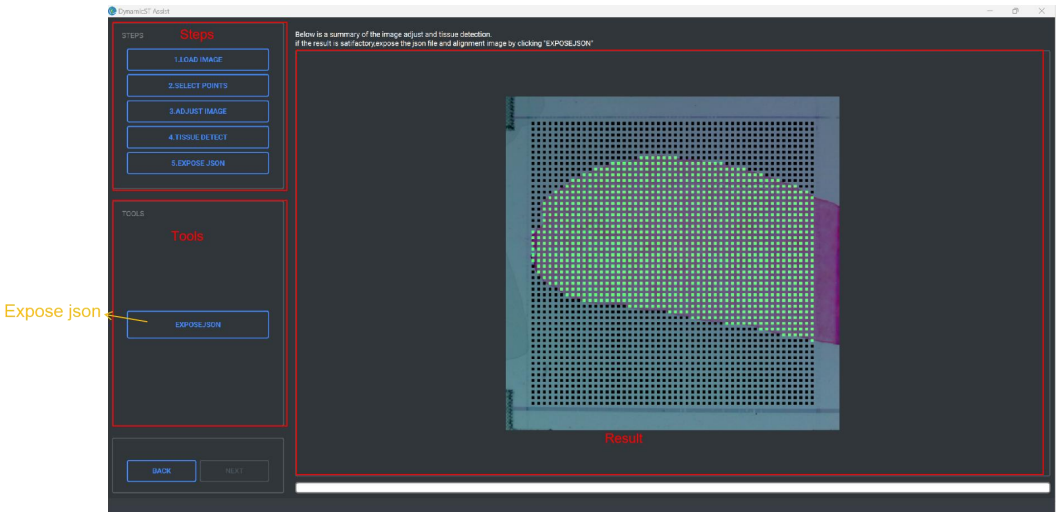
(5) Expose json

A. Click button “EXPOSEJSON” to expose adjusted image and json file.

B. The json file willed be named “DynaBlot image name_alignment.json”

C. The adjusted DynaBlot image will be saved in the same path of json file.If you have aligned DynaBlot image and microscope image, the adjusted DynaBlot image will be

named “DynaBlot image name_dynaimage.tif”, else the adjusted DynaBlot image will be named “DynaBlot image name_tissue_image.tif”.



Output Description

The output of DynamicST Assist include json file and adjusted images.

The json file displays the information of each spot as shown in the figure, including the coordinate of the spot on the adjust image, the spot diameter, the row and column coordinate of the spot, whether the spot is covered by tissue, and the barcode of the spot. The coordinate of spots correspond to adjusted image.

If you have aligned DynaBlot image and microscope image, the adjusted image include “microscope image name_tissue_image.tif” and

“DynaBlot image name_dynaimage.tif”. These two images have the same size, and the same tissue area.

Else, the adjusted image only include “DynaBlot image name _tissue_image.tif”.