

After working through this demo you should be able to use the hyper_stitch GUI to stitch many contiguous high dimensional microscopy datasets together. All source code can be found on [Github](#).

INSTALLATION

1. Install the relevant python packages. Hyper_stitch is largely built upon [OpenCV](#) and [Kornia](#), pip installing kornia should provide both of these and all other requirements.

2. Download the code and data from [Github](#) and then unzip the files.

3. In terminal navigate to the newly downloaded directory.

4. There should be a copy of the following files located there:

```
pip install kornia
```

[Download](#)

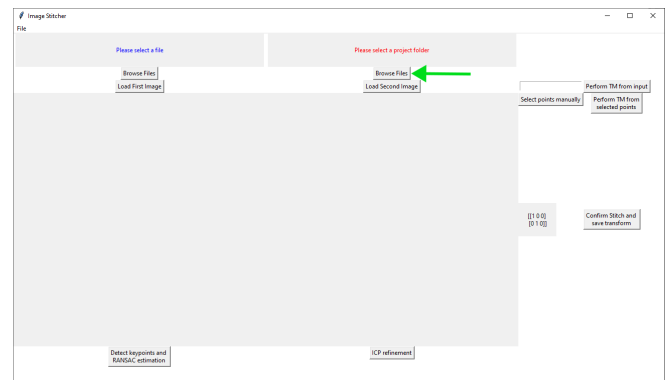
```
cd ../Downloads/hyper_stitch-main
```

```
gui.py
korina_env.yml
LICENSE
README.md
stitching.py
transform_list
utils.py
python gui.py
```

5. Run gui.py with python

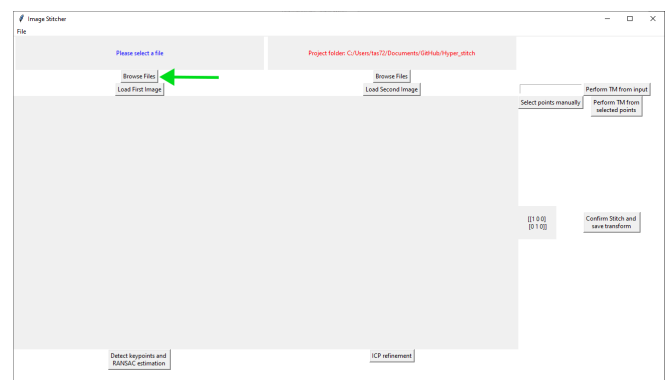
DEMO

1. First select a project folder, this is where the 'transform_list' will be updated and saved as you continually stitch images together. Select, you should see the path appear in red in the top right.



2. Load the images, we use virtual bright field images as a proxy for the 4D-STEM dataset which are saved as two .npy files in the /data directory.

2(i) Select 'Browse Files' on the left. This will open a popup, from here navigate and select one of the images located in /data. You should see the path appear in blue in the top left.

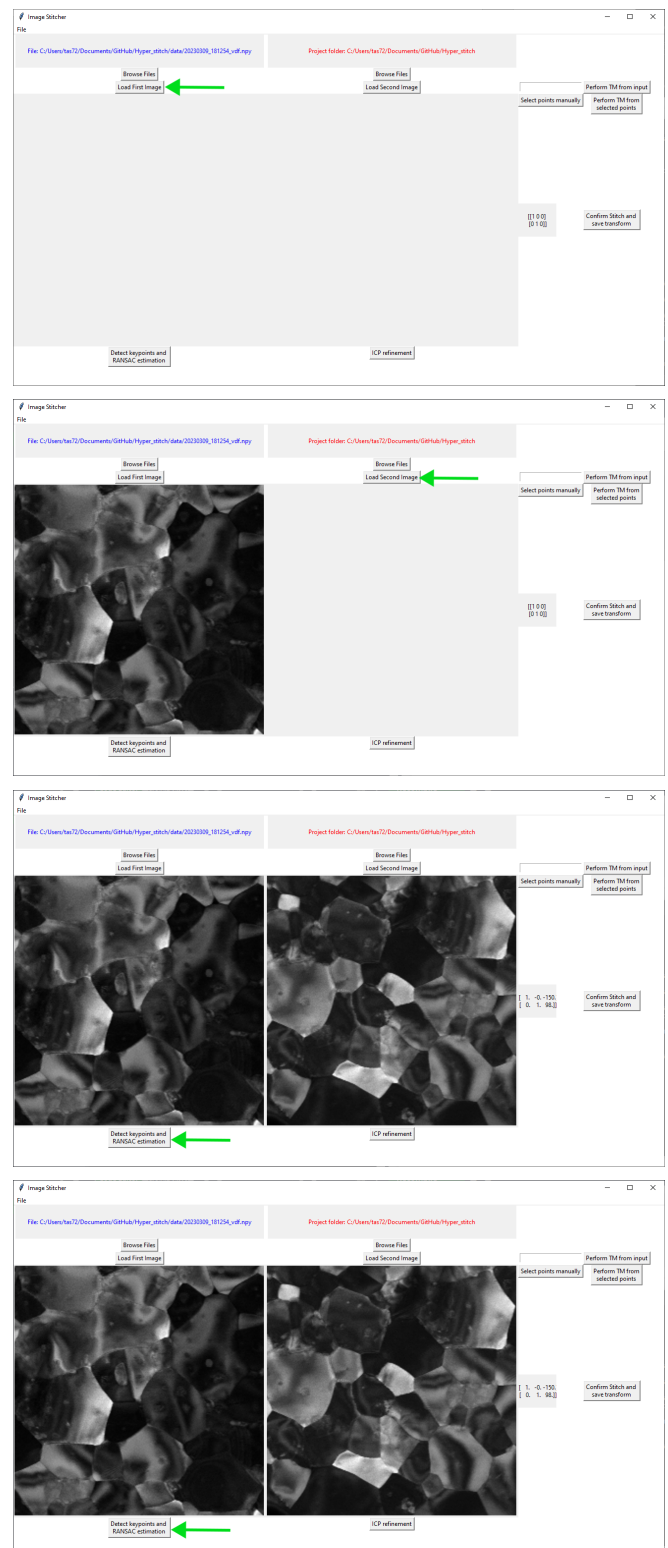


2(ii) Select 'Load First Image' to load the image at the selected path.

2(iii) Repeat this process but now loading the second image with the 'Load Second Image' button.

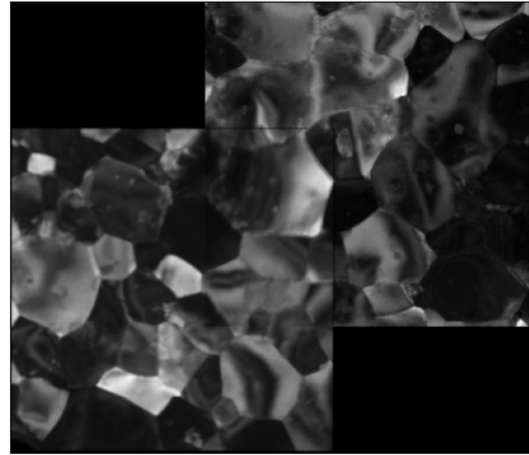
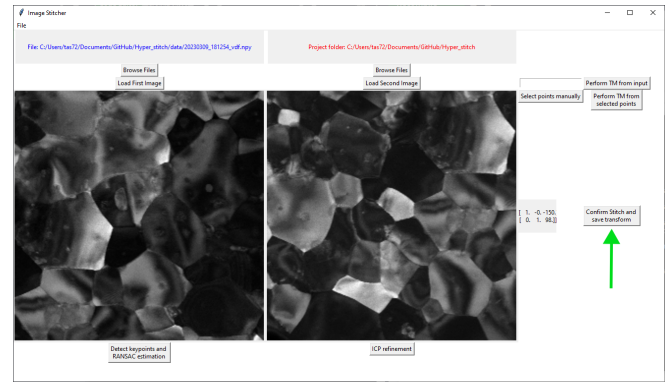
3. Now use the 'Detect keypoints and RANSAC estimation' to map one image onto the other. Once this has been performed two windows will appear sequentially where you can inspect the stitching. An affine transform will now be displayed to the right of the images.

4. If the stitching needs to be improved one way to do this is to use an iterative closest point (ICP) refinement. To do this press the 'ICP refinement' button in the bottom right. Note with the current implementation this can be very computationally costly.



5. Once you are happy with the stitching confirm this with the ‘Confirm Stitch and save transform’ button. **This will delete the original images and save the stitched version in their place. We therefore recommend to make a copy of all unstitched raw images before confirming the stitch.** The transform_list will also be saved in the project folder as a ‘transform_list’ file. This is a record of all affine transforms and which images they are applied to. **Again this will overwrite any old ‘transform_list’ file in the same directory. We therefore recommend loading in any previous ‘transform_list’ file by navigating to File -> Load transform file when the GUI is first started.**

6. The stitch is now complete and you can inspect the results. To load the ‘transform_list’ file in python use `pandas, pd.read_pickle(path_to_file)`. The transform_list can then be used to calculate where each pixel from each image ends up in the stitched dataset. This can then be used to calculate the position of pixels once spatially correlated to another microscopy technique.



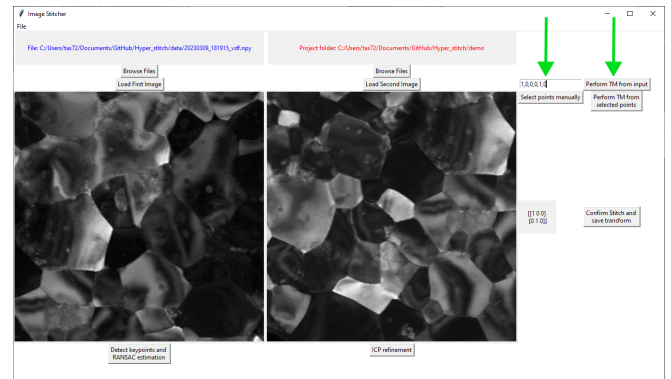
ADDITIONAL FEATURES AND SETTINGS

0.1 Entering an affine transform manually

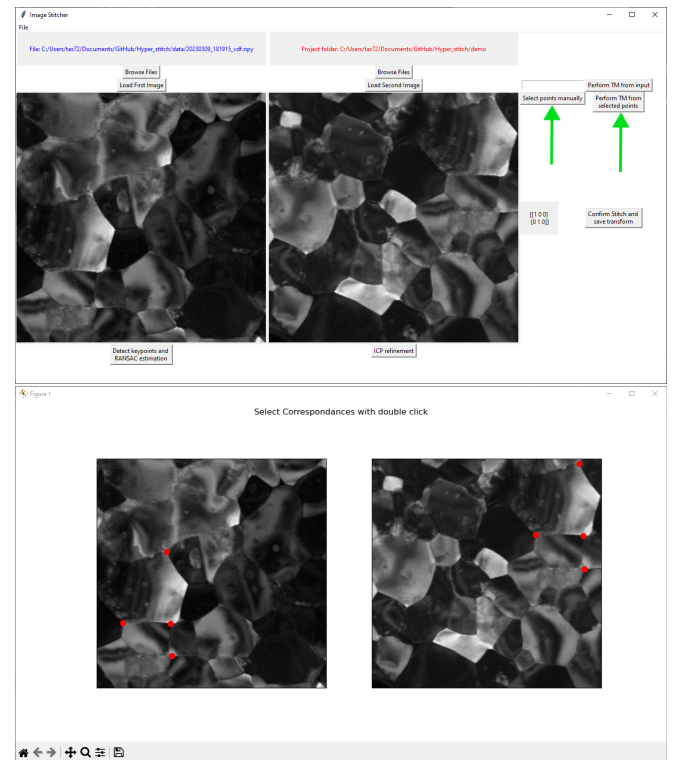
If you want to manually enter a known affine transform this can be done via the entry box in the top right. The entered values should be of the form a, b, t_x, c, d, t_y and separated by commas giving the

$$\text{affine transform: } M = \begin{bmatrix} a & b \\ c & d \end{bmatrix} + \begin{bmatrix} t_x \\ t_y \end{bmatrix}$$

To inspect the results of the transform simply press ‘perform TM from input’ to the right of the inputted values. The stitching can be completed and the transform_list updated via the ‘Confirm stitch and save transform’ button.



Manually chosen key-points between the two images can also be inputted. To do this select the 'Select points manually' button. This will open a window with the two images. Double a point on one image then the corresponding point on the other to link them as correspondences. Once all corresponding points have been entered (3 minimum) close the window and inspect the stitch with the 'Perform TM from selected points'. To confirm the stitch select 'Confirm Stitch and save transform'.



0.2 Changing key-point detection hyper-parameters

To alter quality of the key-point detection and affine transform some hyper-parameters can be altered.

(i) Firstly considering the key-point and RANSAC algorithm, navigate to 'File -> Alter SIFT and RANSAC params'. This will then allow an input via terminal where variable corresponding to the key-point detection and RANSAC algorithms can be adjusted.

(ii) The key-point detection algorithm can also be changed by pressing the 'File -> Change detection mode'. This cycles between KeyNet detector + AffNet + HardNet (kornia); the scale invariant feature transform (sift); binary robust invariant scalable keypoint (brisk); and a combination of all algorithms (combo). It has been found empirically that the kornia option gives the most reliable results.

