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# Image Registration of MALDI IMS to Microscopy

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**Protocol status:** Working

**We use this protocol and it's working**

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## Abstract

Scope:

To process to register MALDI IMS images to different types of microscopy images.



- 1 Generate a MALDI imaging mass spectrometry (IMS) pixel map using regToolboxMSRC  
<https://github.com/nhpatterson/regtoolboxmsrc>
- 2 Using FIJI ImageJ, select corresponding laser ablation marks and IMS pixels in the post-acquisition autofluorescence (post-AF) and IMS pixel map, respectively.  
<https://fiji.sc>
- 3 Use a "Landmark Correspondences" FIJI plugin to find an affine transformation between the two images, resampling the post-AF image to the IMS pixel map by selecting post-AF image as source image, and IMS pixel map as the template image within the plugin.
- 4 Save the post-AF image.
- 5 Using regToolboxMSRC again, align other microscopy modalities (pre-IMS acquisition autofluorescence, brightfield PAS, multiplex immunofluorescence) to the newly saved post-AF image.
- 6 When complete, all images will be sampled in the same coordinates as the IMS pixel map.