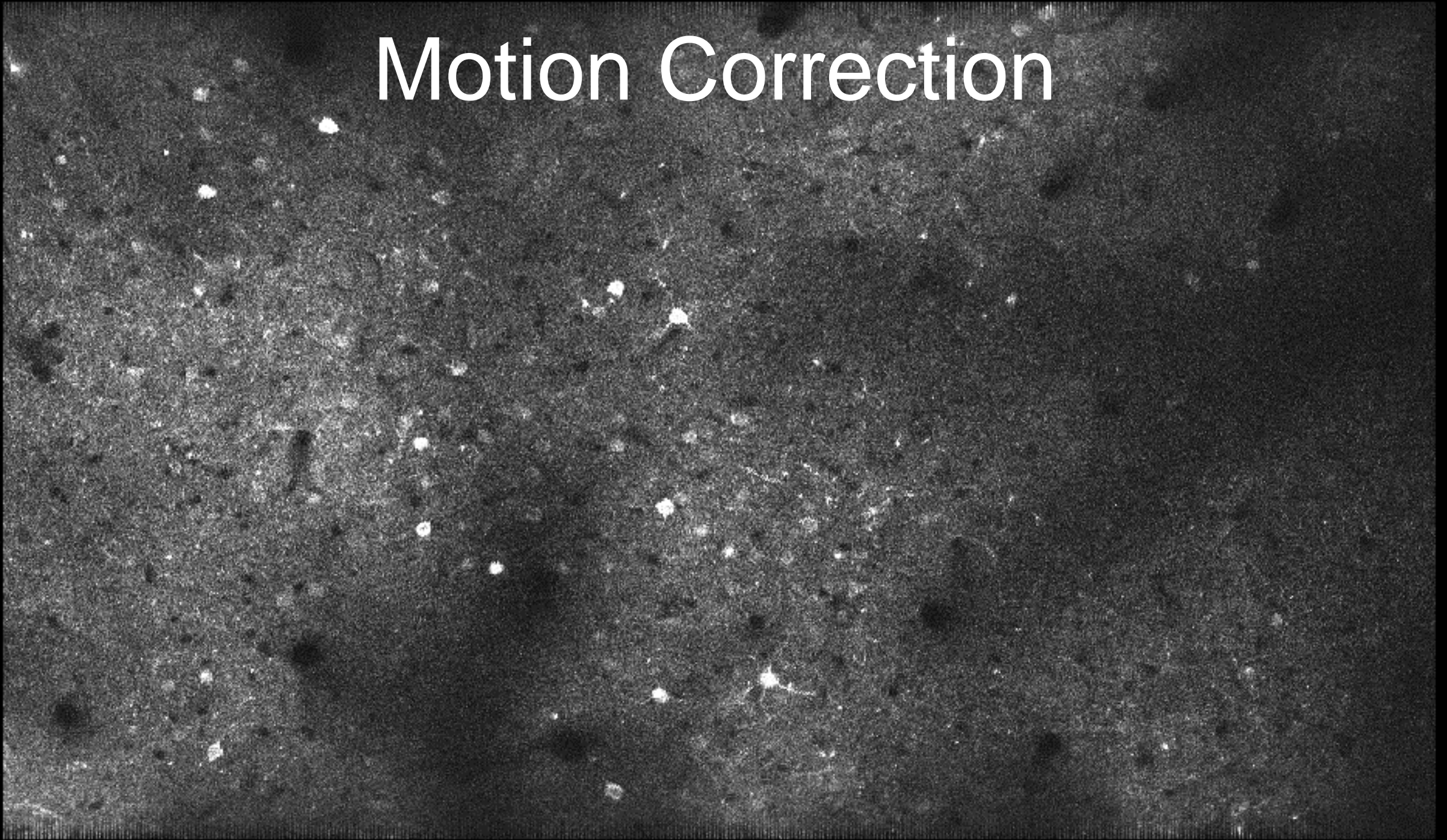


Motion Correction



Why should we care?

- Behaviors → brain movements!
- Movements can be in XY (as in video)
- Also movements in Z (harder to spot)
- First let's correct XY movement

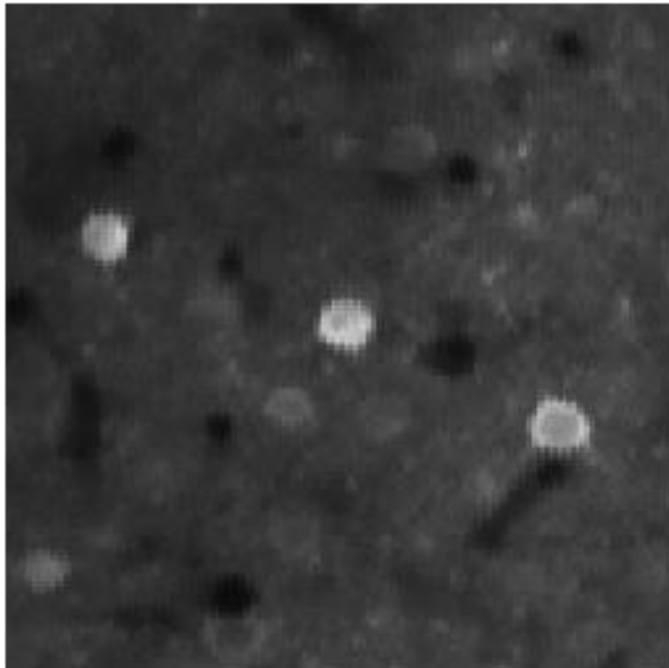
XY registration

- 1: compute reference image to align all frames to
- 2: compute rigid shifts relative to reference image
- 3: compute non-rigid shifts relative to reference image
- 4: benchmarking results

1: compute reference image

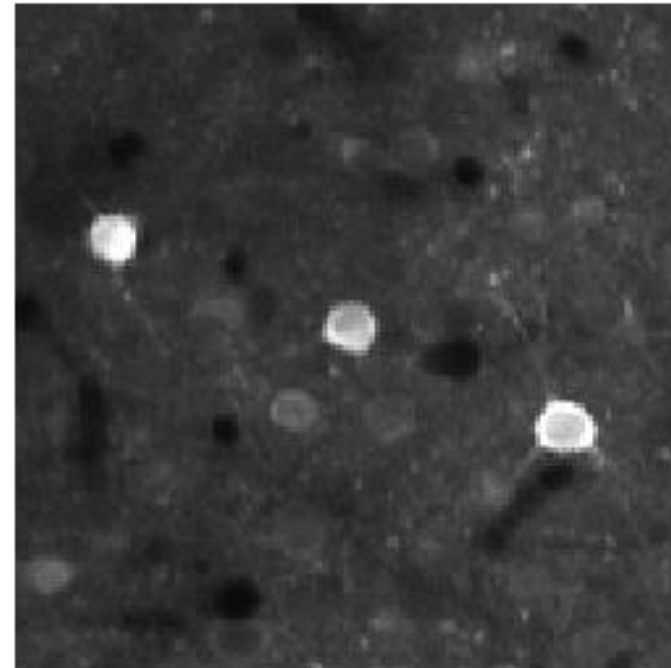
- a) Take 250 random frames and compute correlation matrix
- b) Take as the seed the frame with the most correlated neighbors
- c) Take the mean of the seed and the top 20 most correlated frames

average of 250 frames

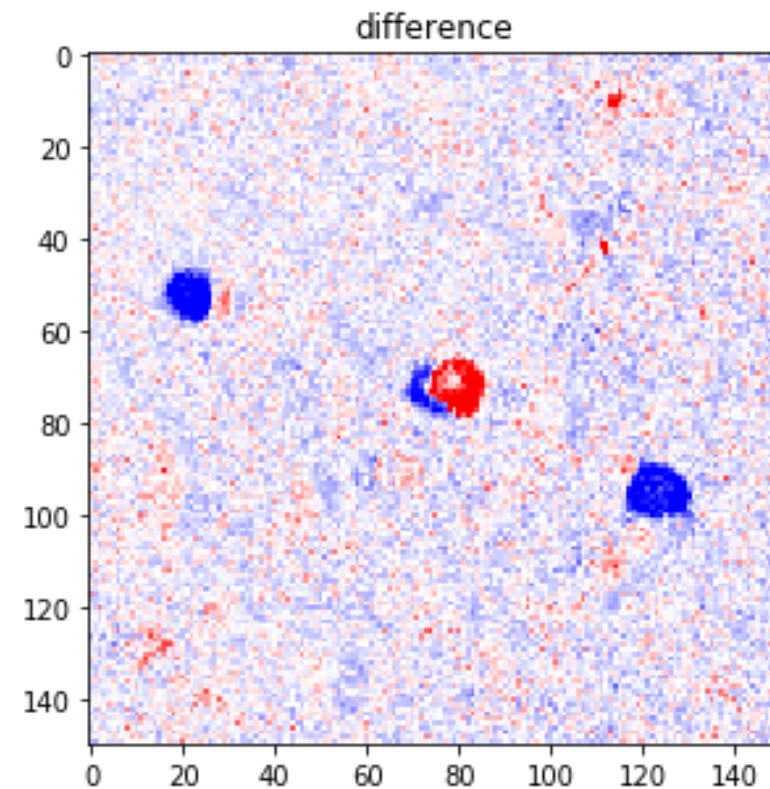
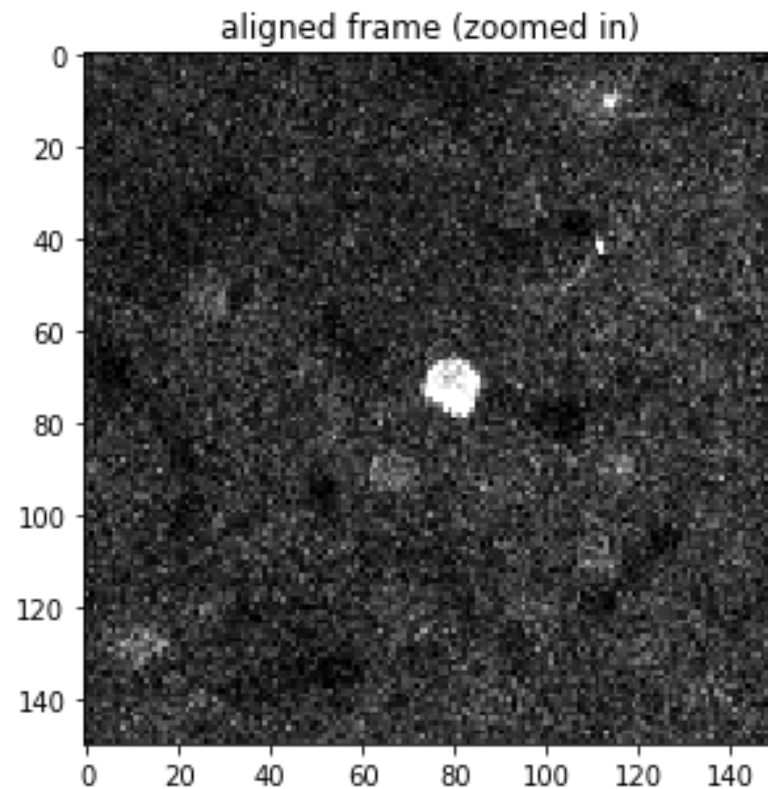
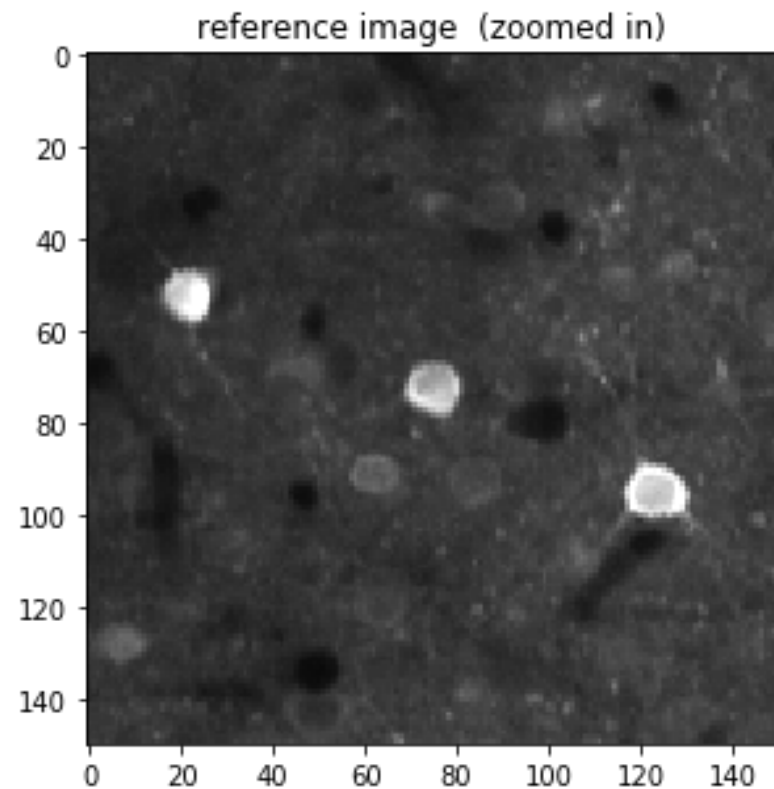


VS

iteratively aligned average of 250 frames

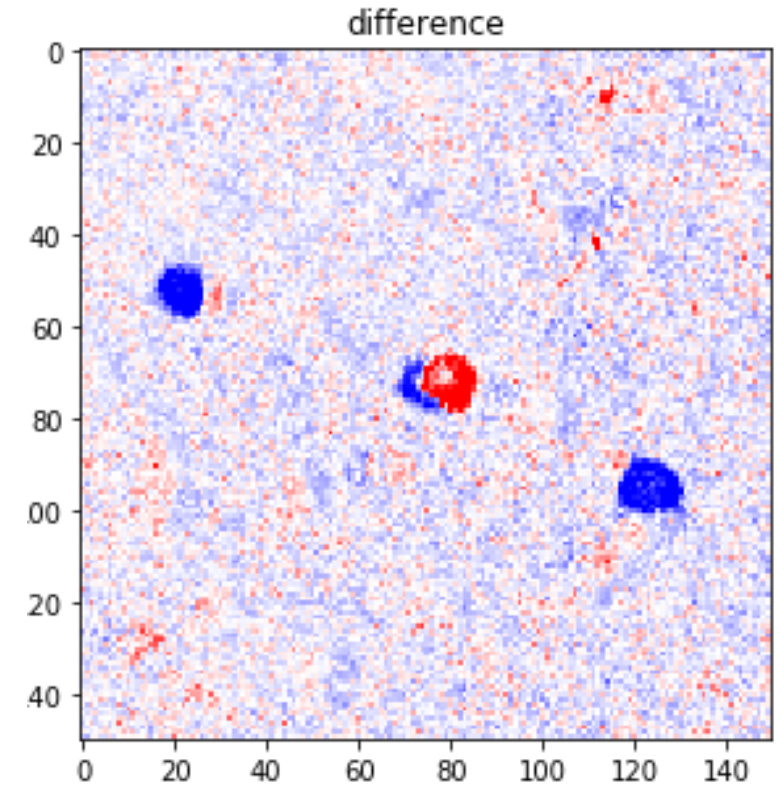
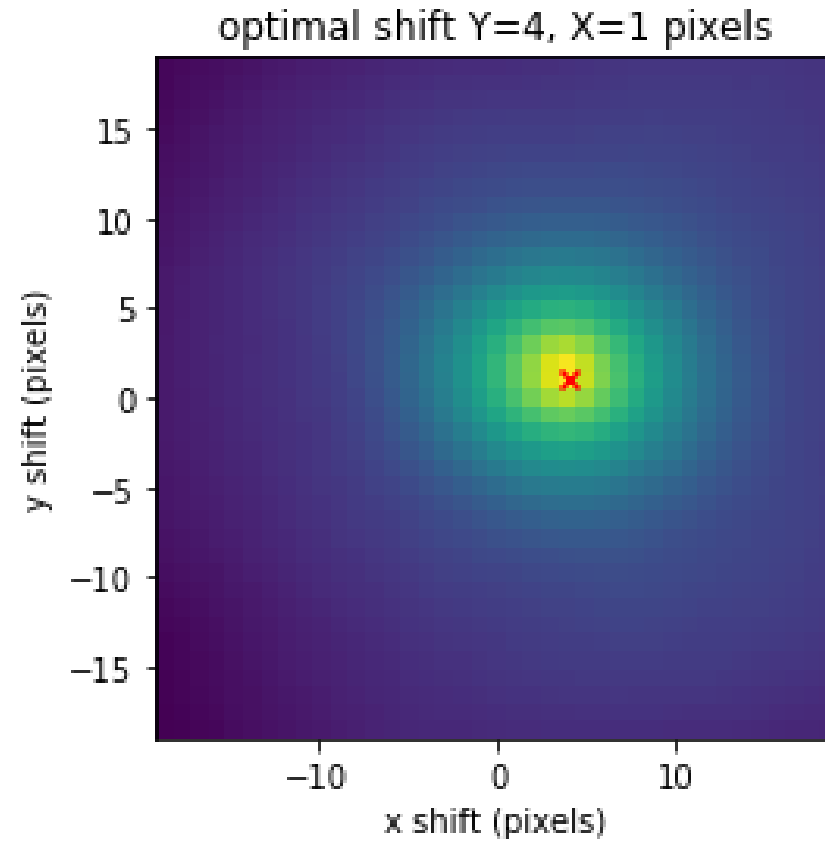
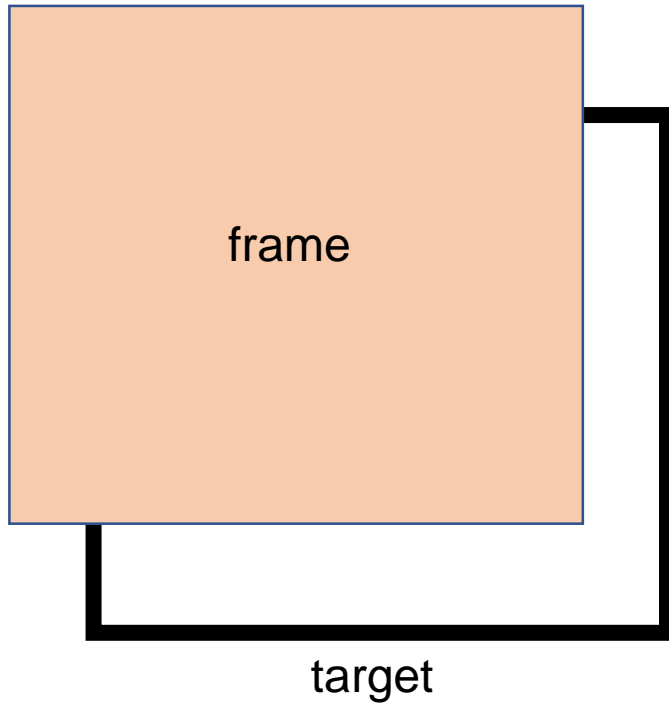


2: rigid registration



2: rigid registration

Cross-correlation
between frame and
reference:



2: rigid registration

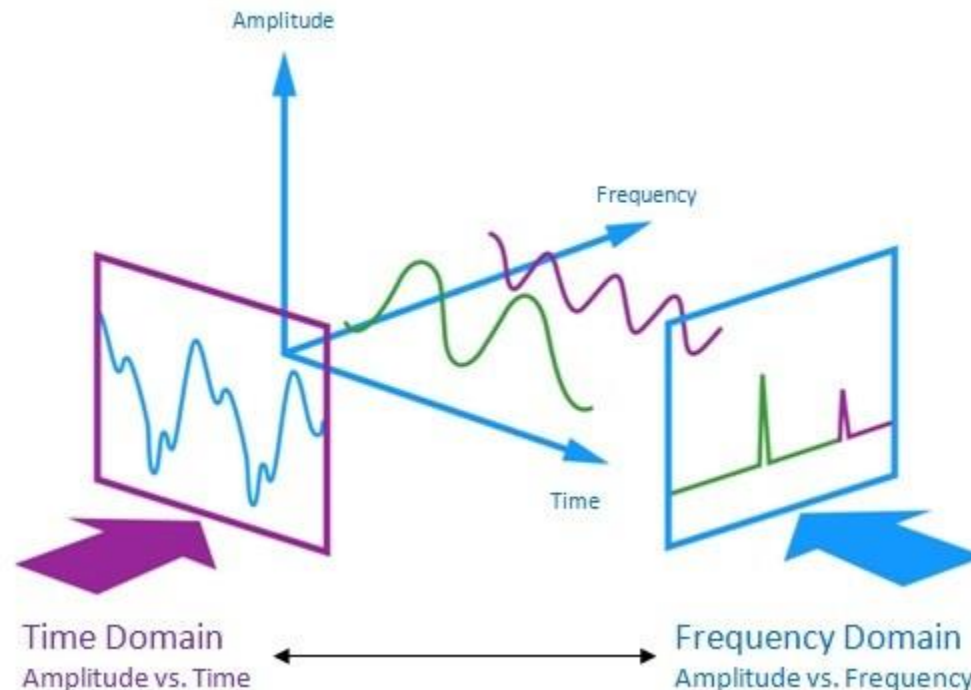
Cross-correlation is slooowww, but...

Cross-correlation is the same as the **convolution** of the frame with the reference.

convolution theorem: convolution = multiplication in the frequency domain

➔ Take FFT of reference and frame and multiply

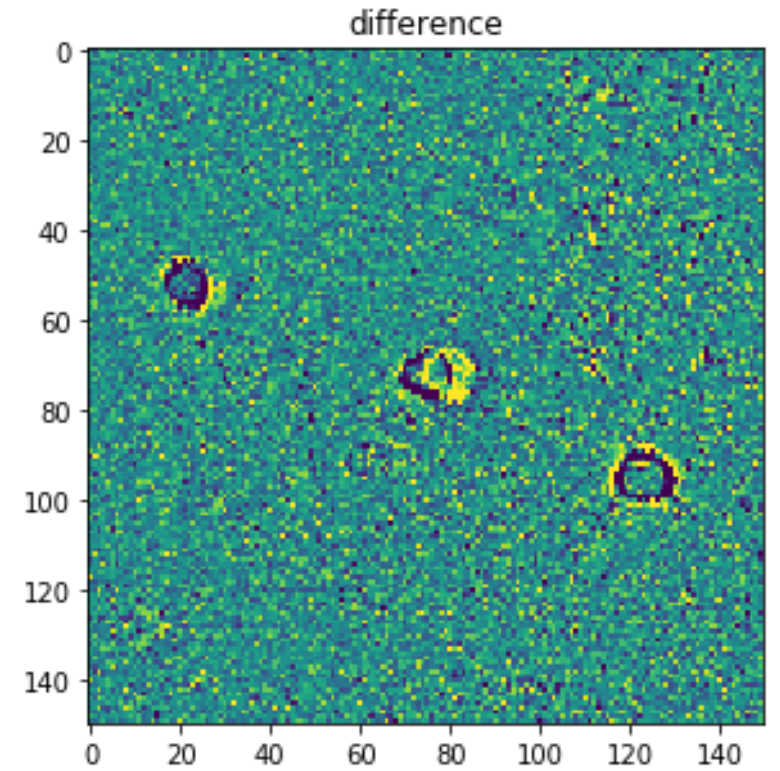
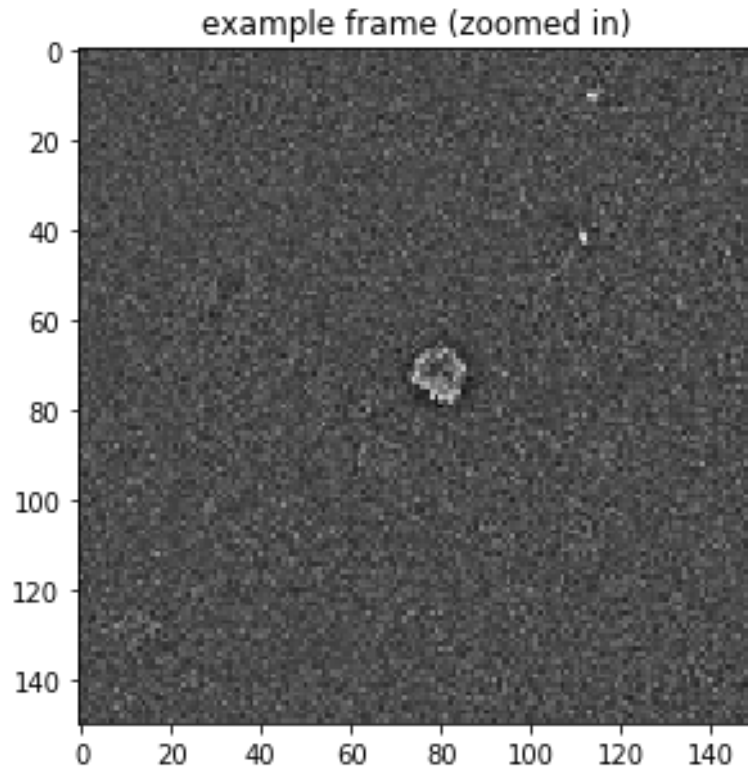
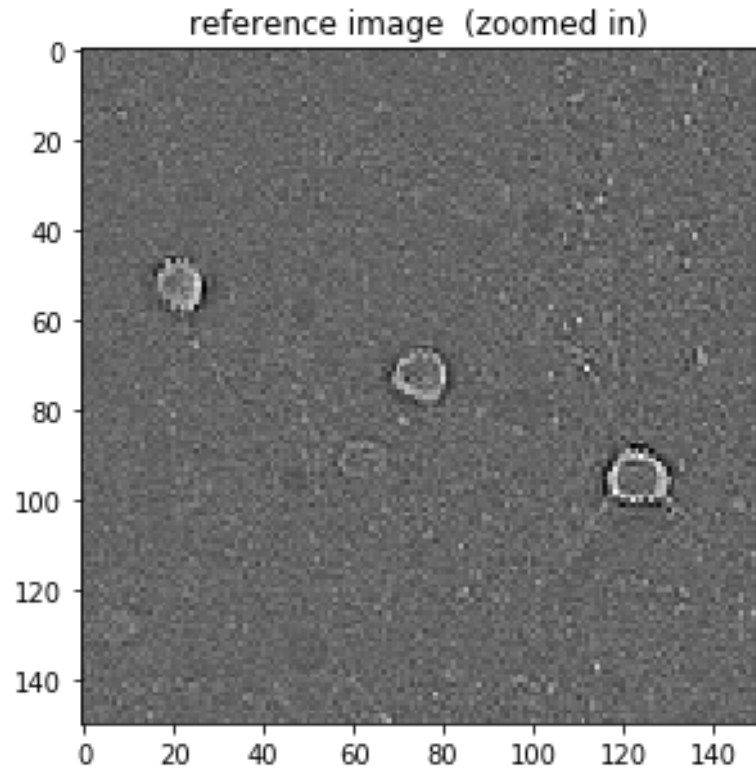
100x faster



2: rigid registration

But this still isn't the best approach

Get better results if you whiten: phase-correlation



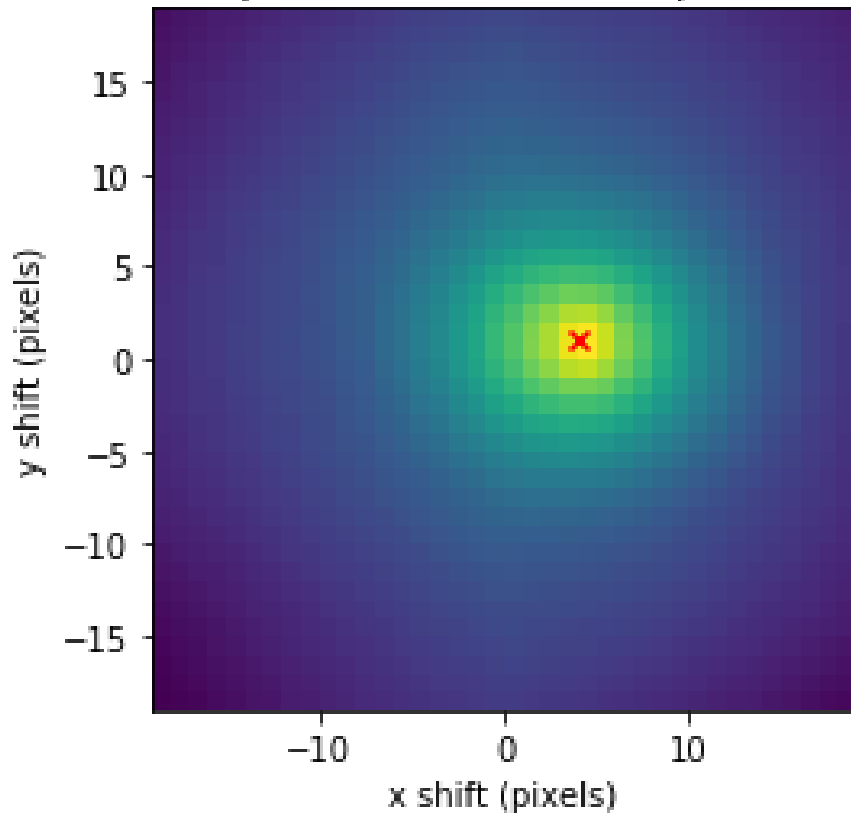
2: rigid registration

But this still isn't the best approach

Get better results if you whiten: phase-correlation

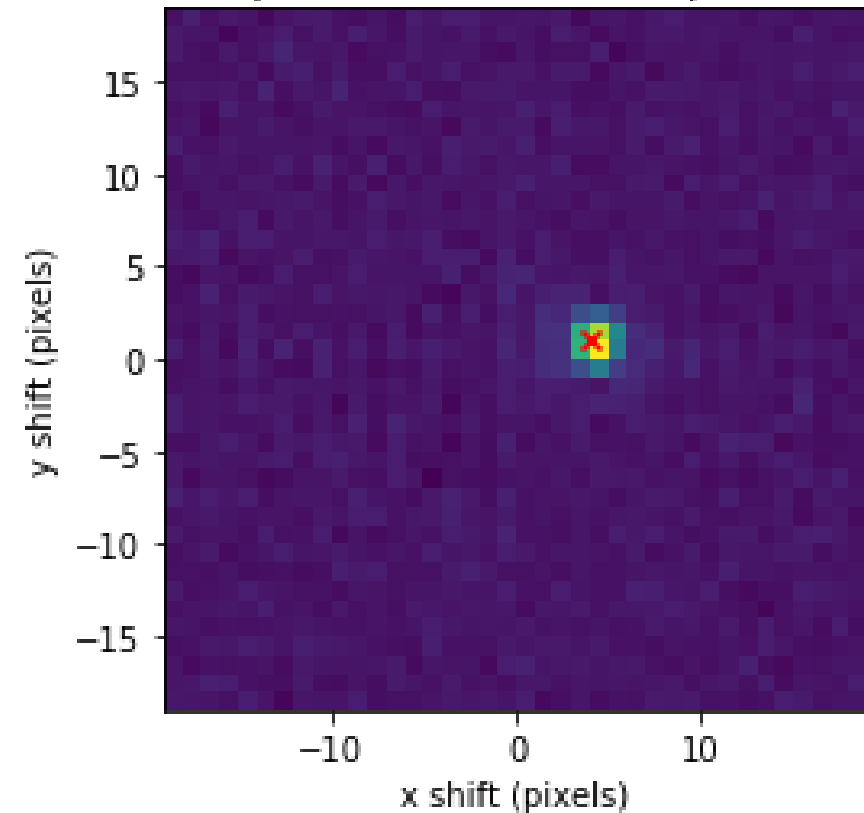
cross-correlation

optimal shift Y=4, X=1 pixels



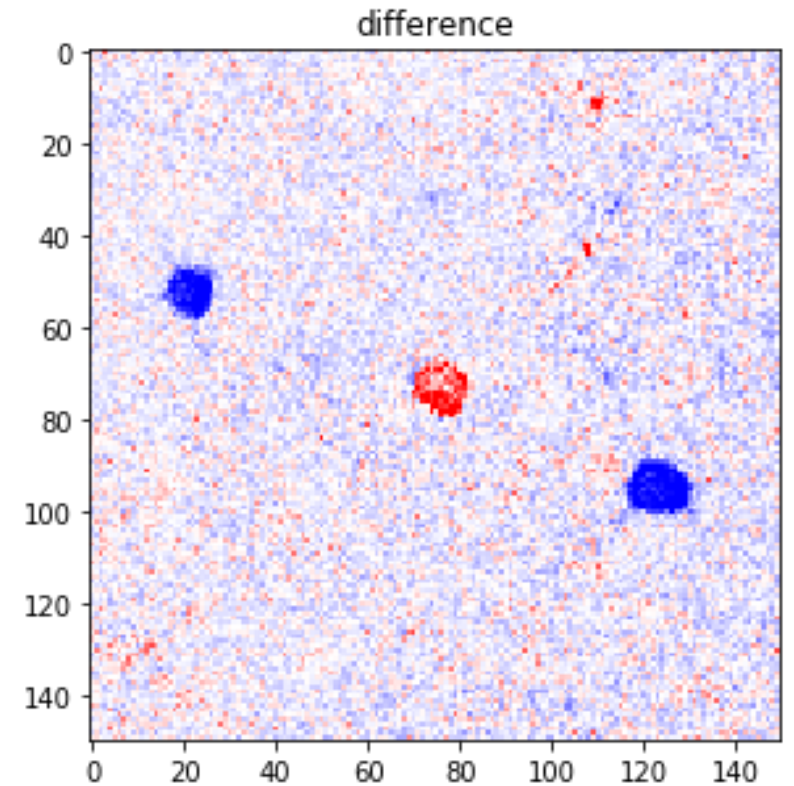
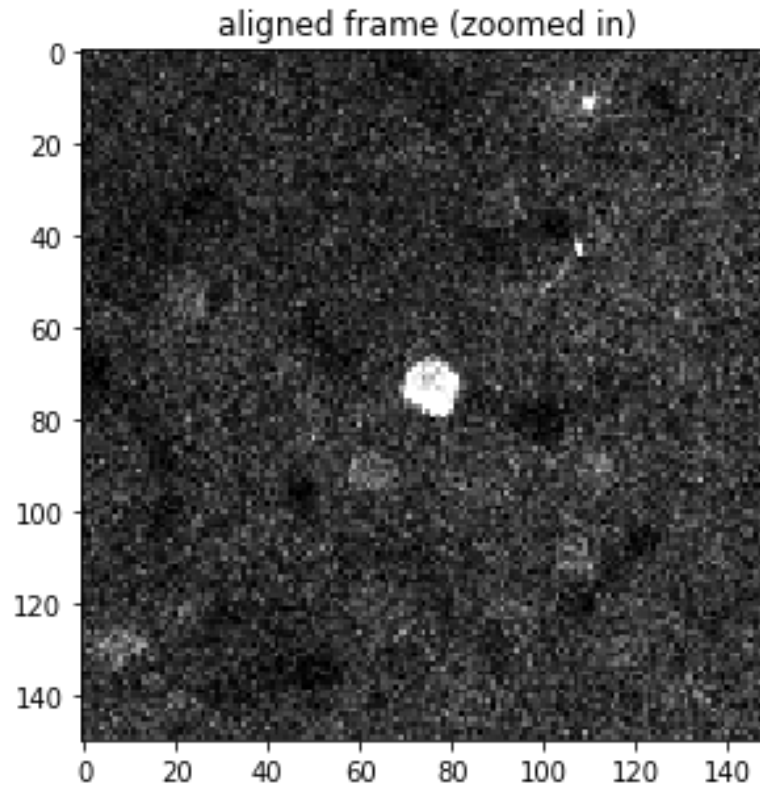
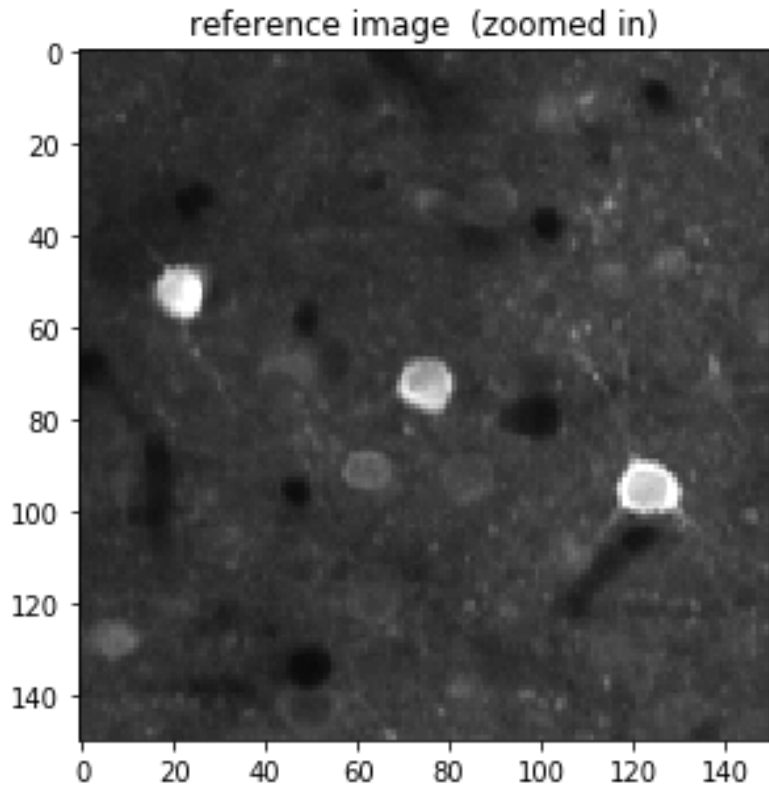
phase-correlation

optimal shift Y=4, X=1 pixels



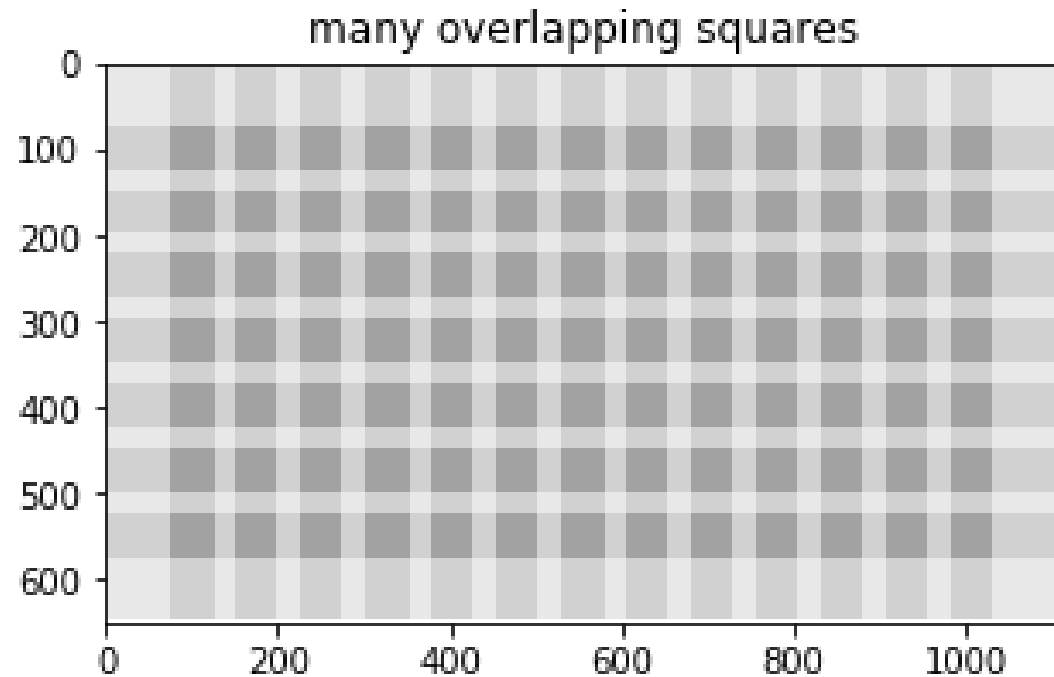
2: rigid registration

Move frame by calculated integer shifts (4,-1)



3: non-rigid registration

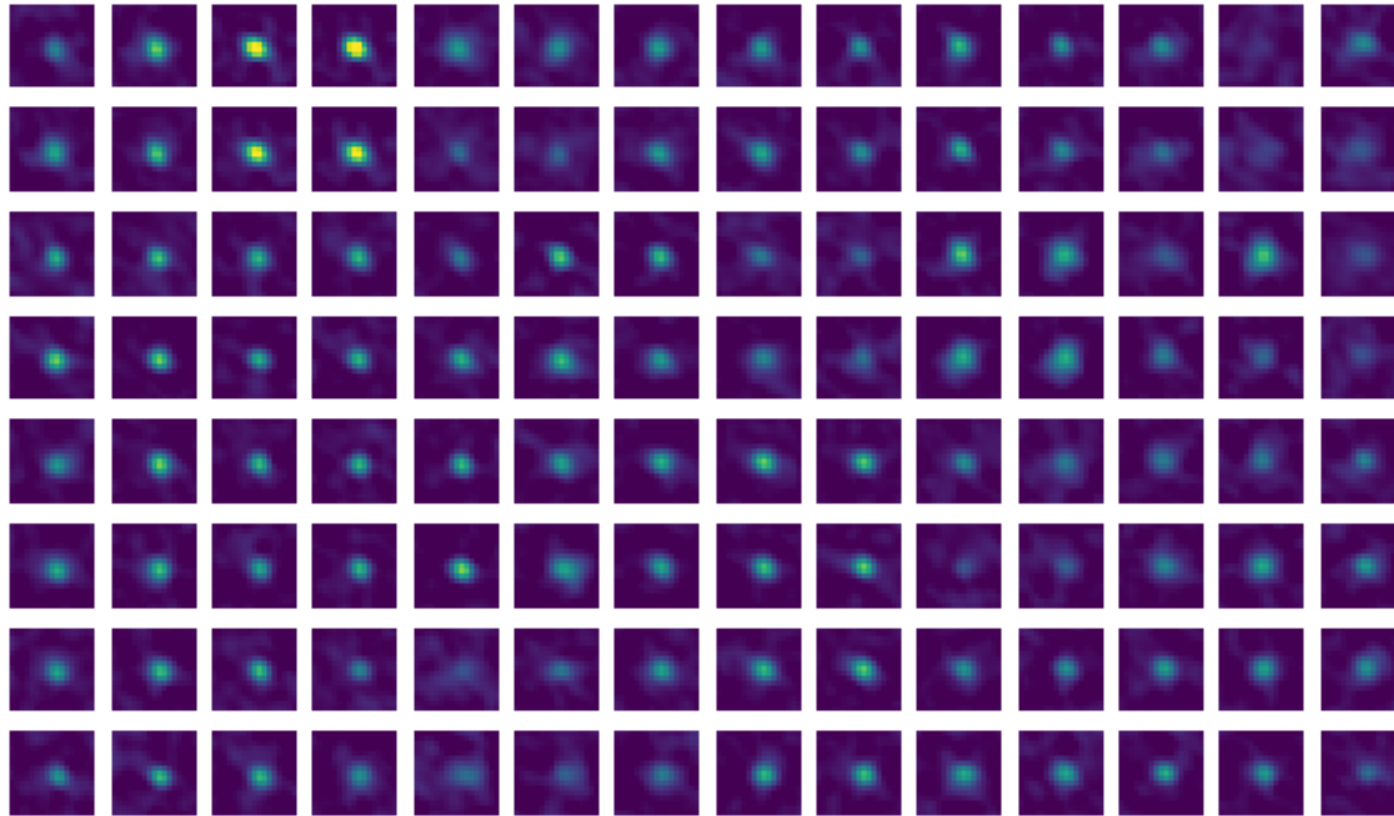
Divide field of view (FOV) into over-lapping blocks



Now we will compute the phase-correlation between each block and the reference image

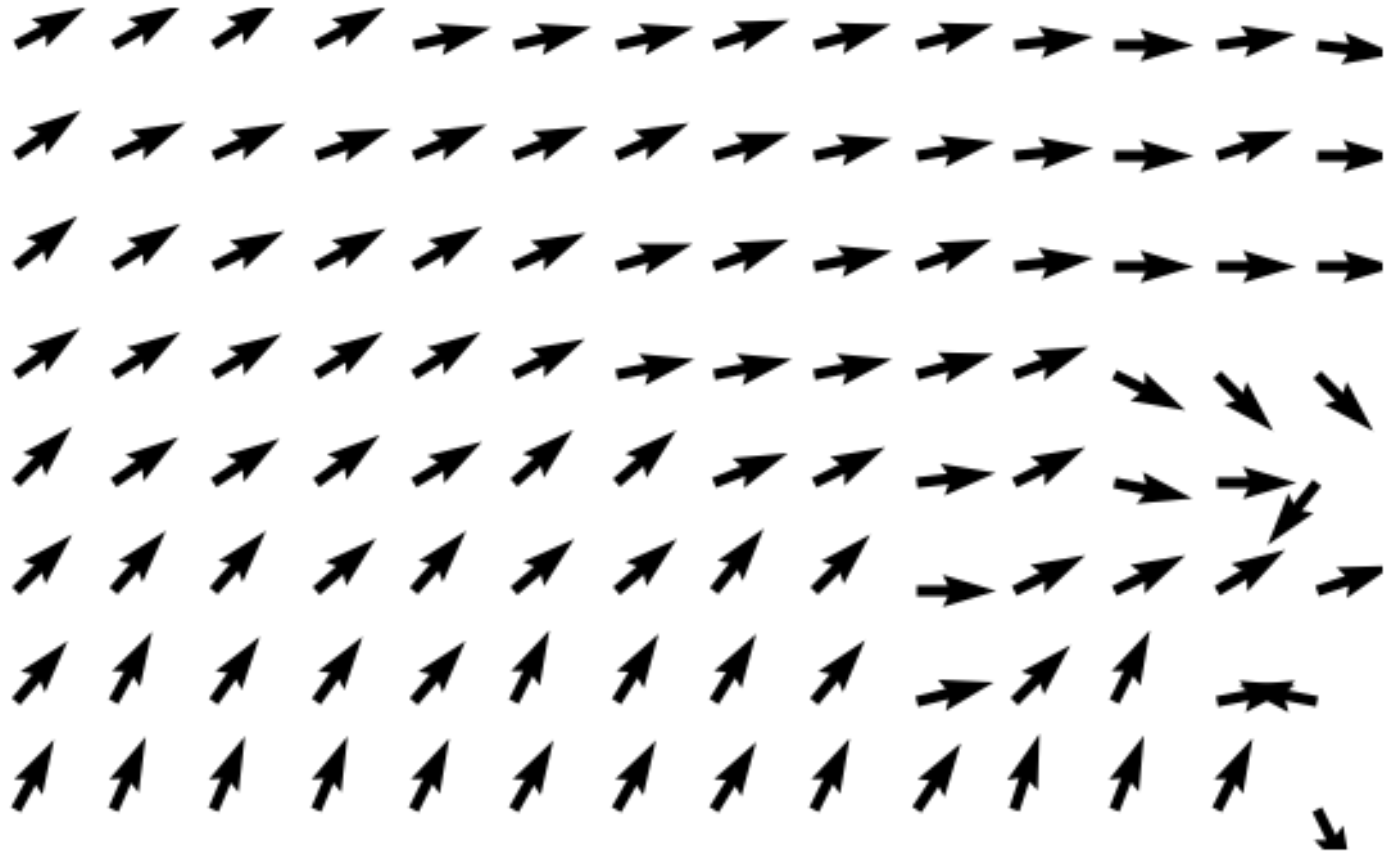
3: non-rigid registration

Phase correlation of each block



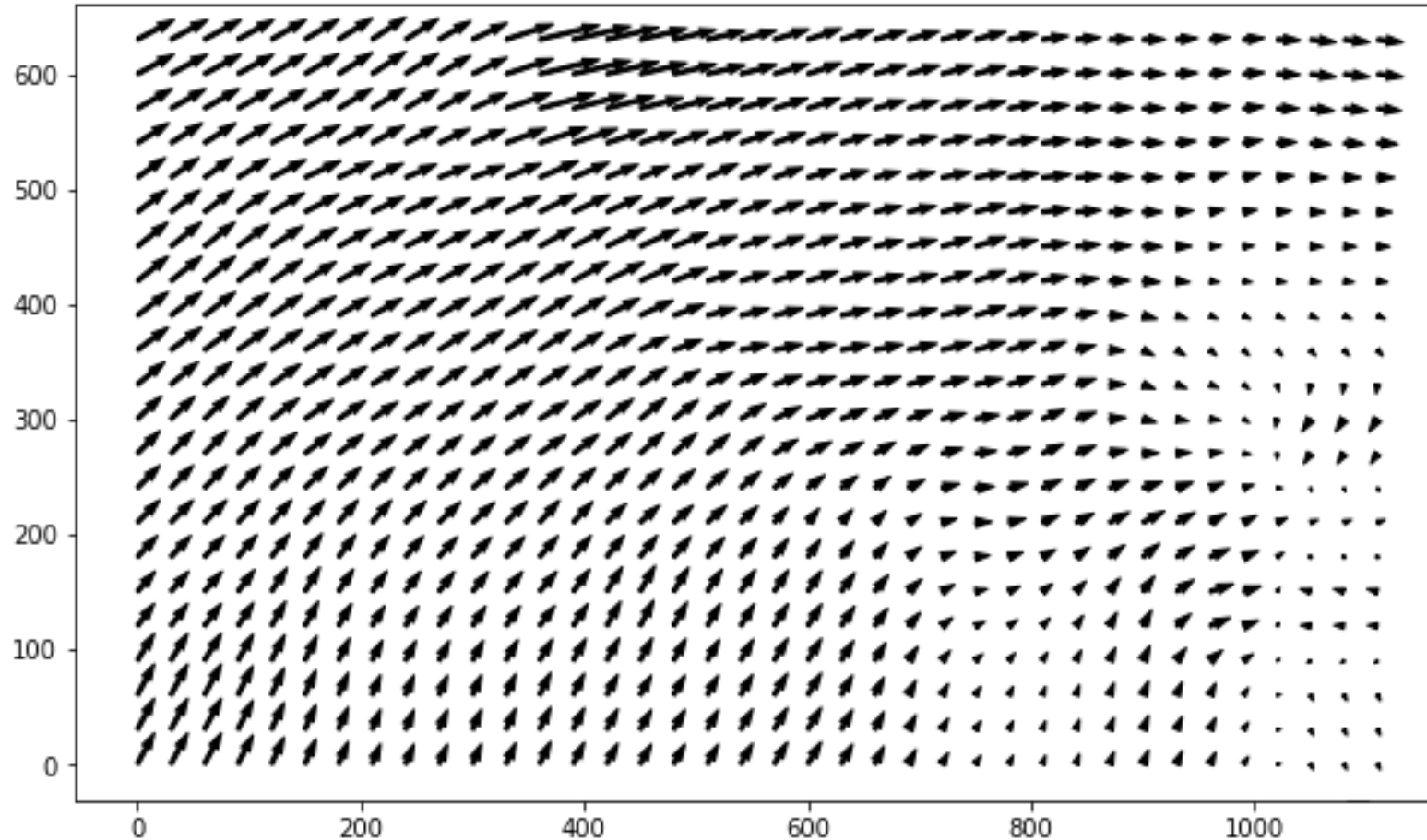
3: non-rigid registration

Phase correlation of each block



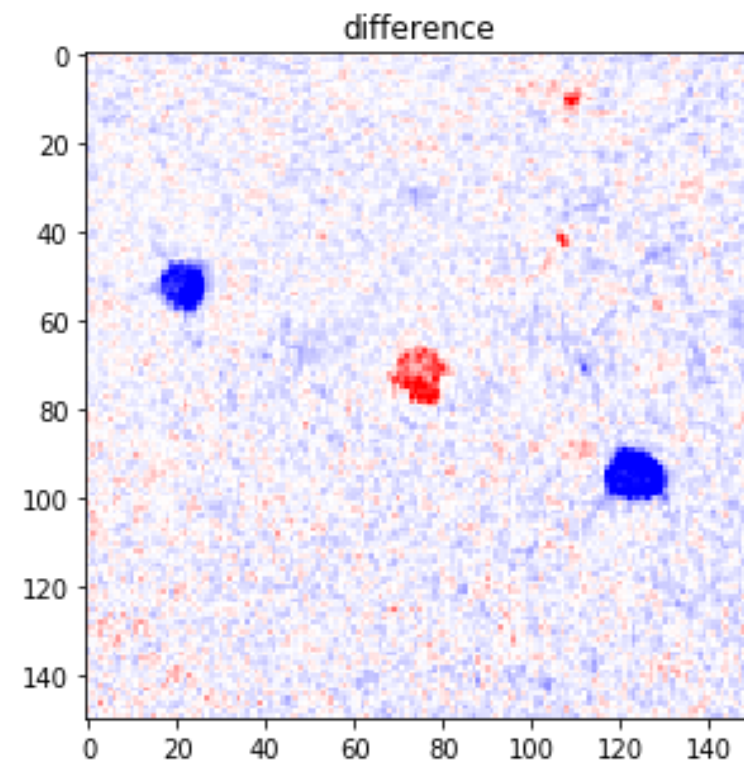
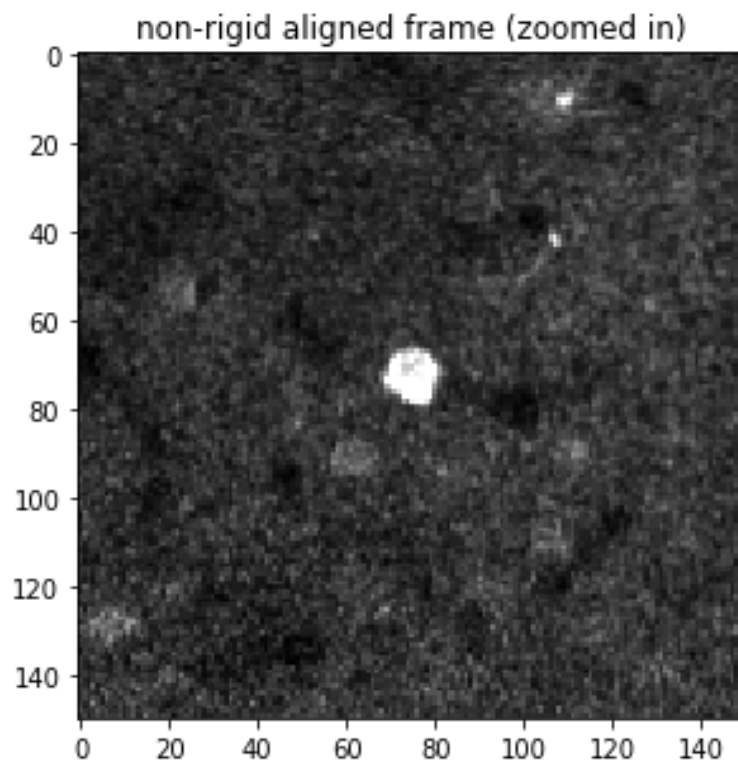
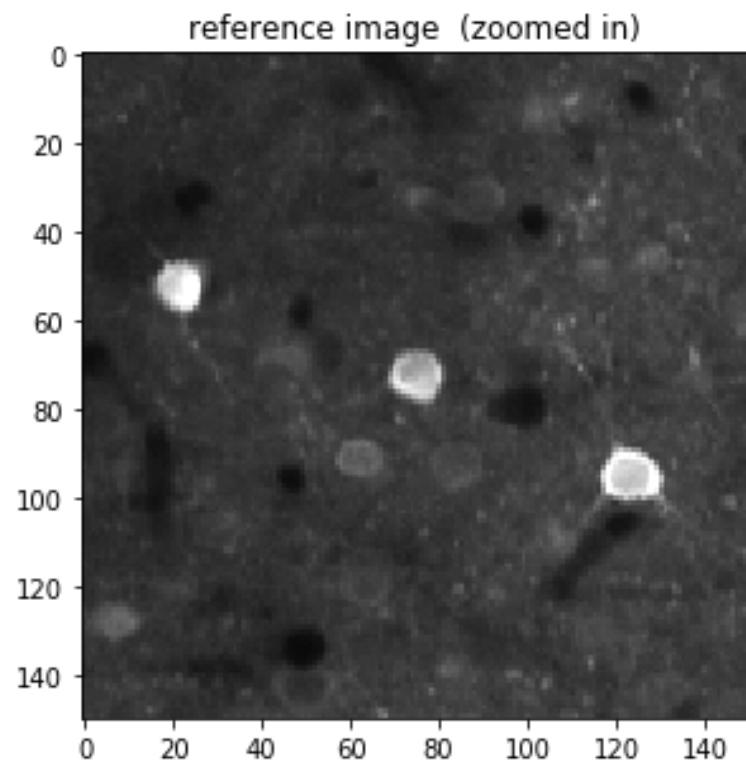
3: non-rigid registration

Phase correlation of each block



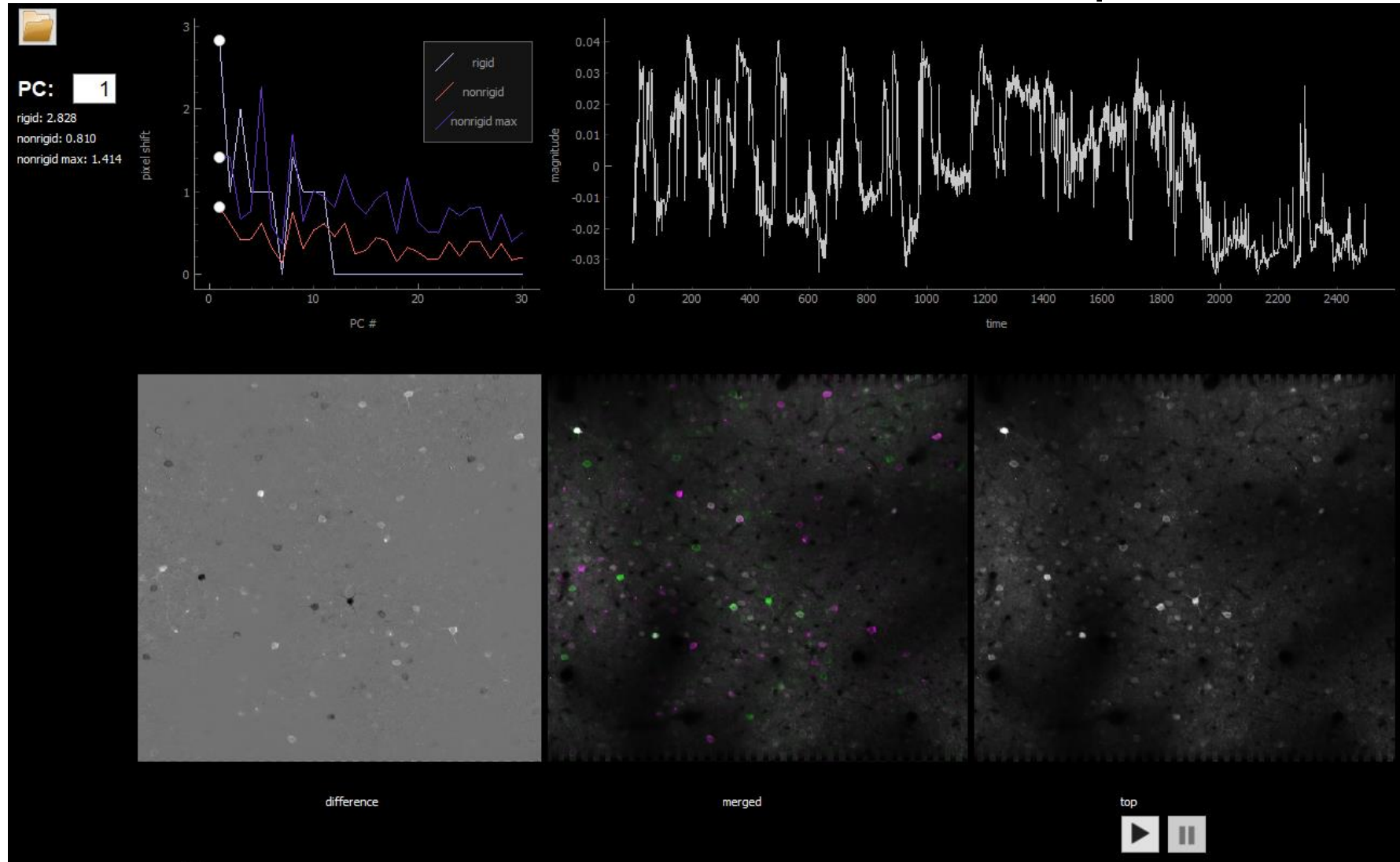
Upsample to get shift for each pixel using bilinear interpolation

3: non-rigid registration



4: benchmarking registration

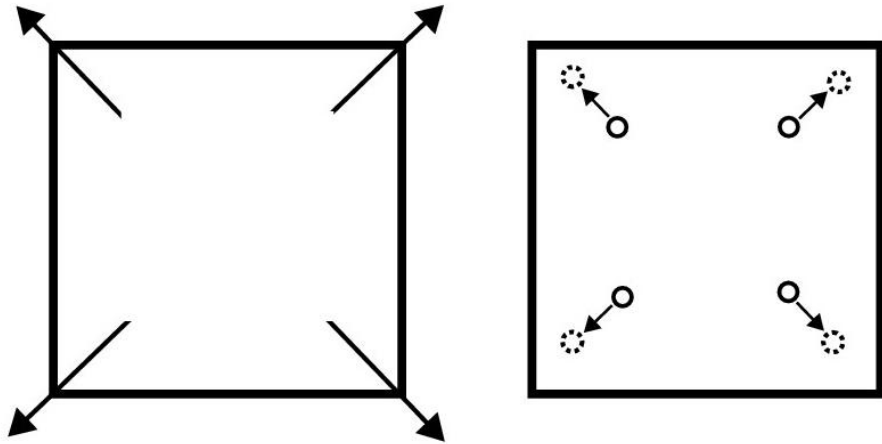
Take PCA of whole movie and see if PC's correspond to motion



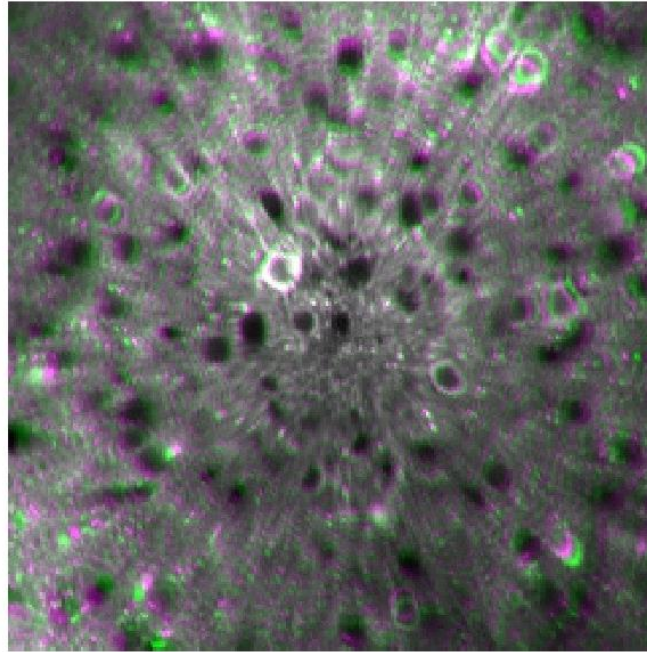
4: benchmarking registration

What do different brain movements look like?

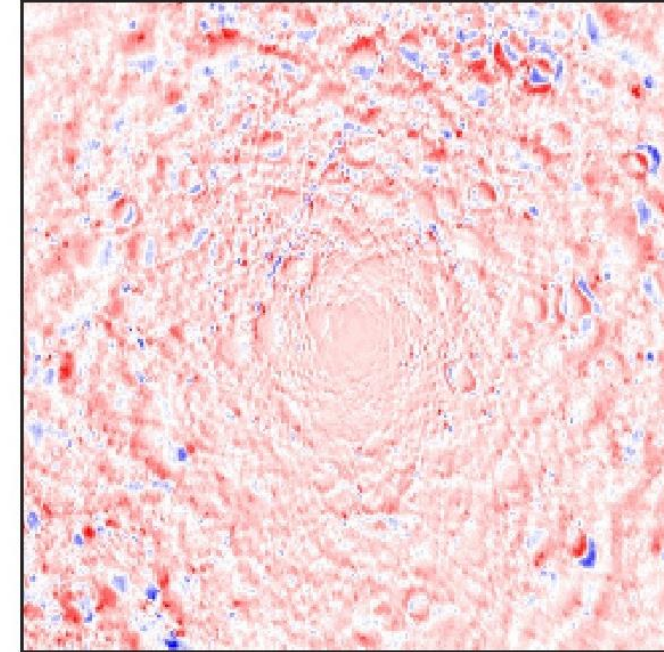
(d) Non-rigid tissue deformation



merged



difference



Let's run registration!

Don't run cell detection

Choose run options (hold mouse over parameters to see descriptions)

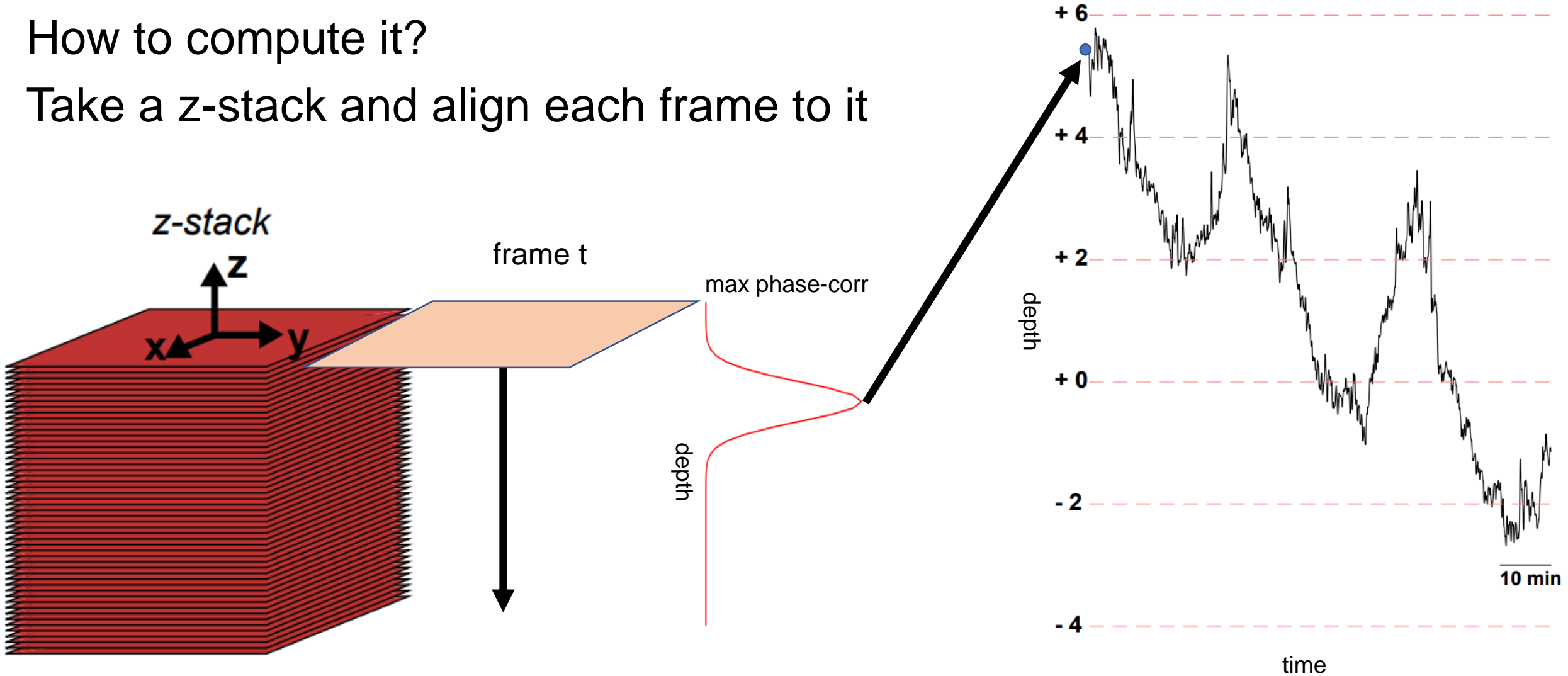
File paths look_one_level_down 0 Add directory to data_path data_path D:/DATA/GT1 OR add h5 file path Add save_path (default is 1st data_path) Add fast_disk (default is save_path) RUN SUITE2P STOP Add a clean-up *.py	Load ops file Save ops as default Save ops to file Load example ops 1P imaging dendrites/axons	Main settings nplanes 1 nchannels 1 functional_chan 1 tau 1.0 fs 10.0 delete_bin 0 do_bidiphase 0 bidiphase 0	Output settings preclassify 0.0 save_mat 0 combined 1 reg_tif 0 reg_tif_chan2 0 aspect 1.0	Registration do_registration 1 align_by_chan 1 nimg_init 300 batch_size 500 smooth_sigma 1.15 maxregshift 0.1 th_badframes 1.0 keep_movie_raw 1	Nonrigid nonrigid 1 block_size 128, 128 snr_thresh 1.2 maxregshiftNR 5 1P 1Preg 0 spatial_hp 50 pre_smooth 2 spatial_taper 50	ROI detection roidetect 0 sparse_mode 0 diameter 12 spatial_scale 0 connected 1 threshold_scaling 1.0 max_overlap 0.75 max_iterations 20 high_pass 100 save settings and add more (batch)	Extraction/Neuropil allow_overlap 0 inner_neuropil_radius 2 min_neuropil_pixels 350 Deconvolution win_baseline 60.0 sig_baseline 10.0 neucoeff 0.7 remove last added
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Keep unregistered movie

Drift in Z

How to compute it?

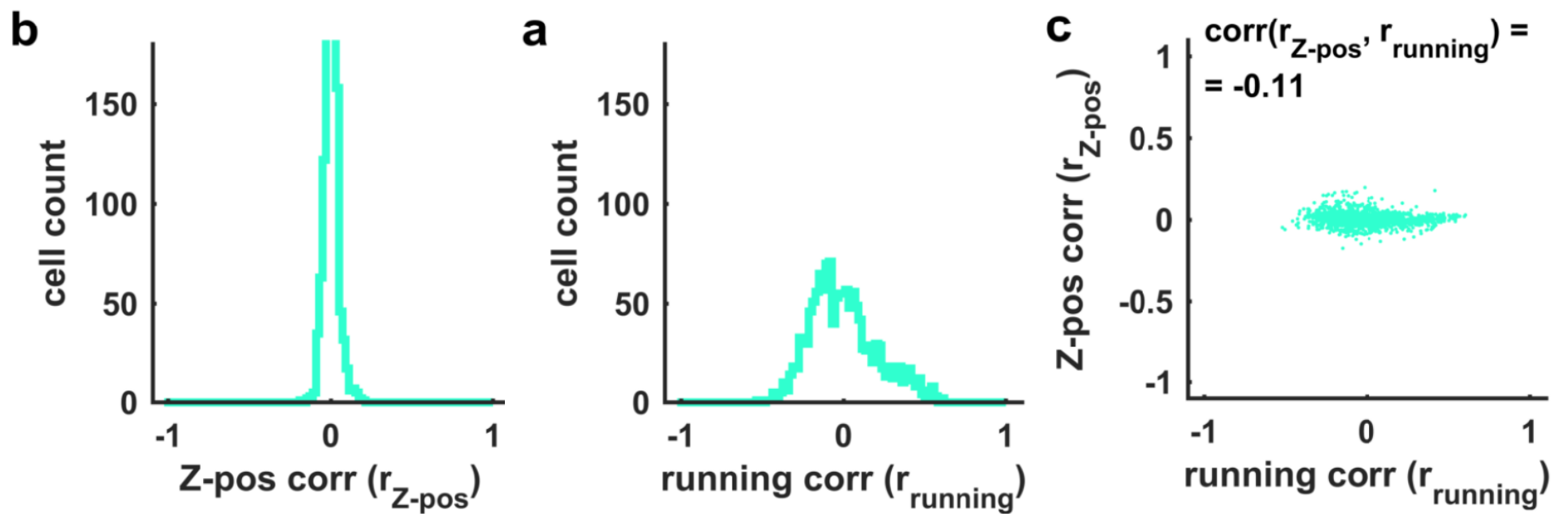
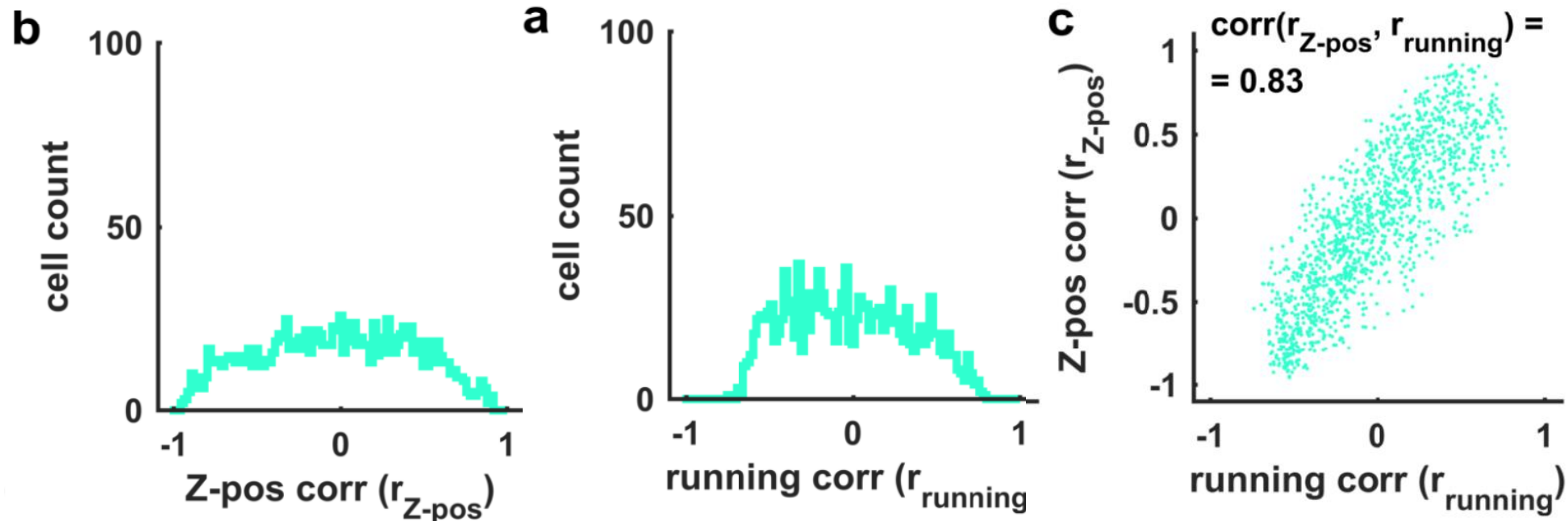
Take a z-stack and align each frame to it



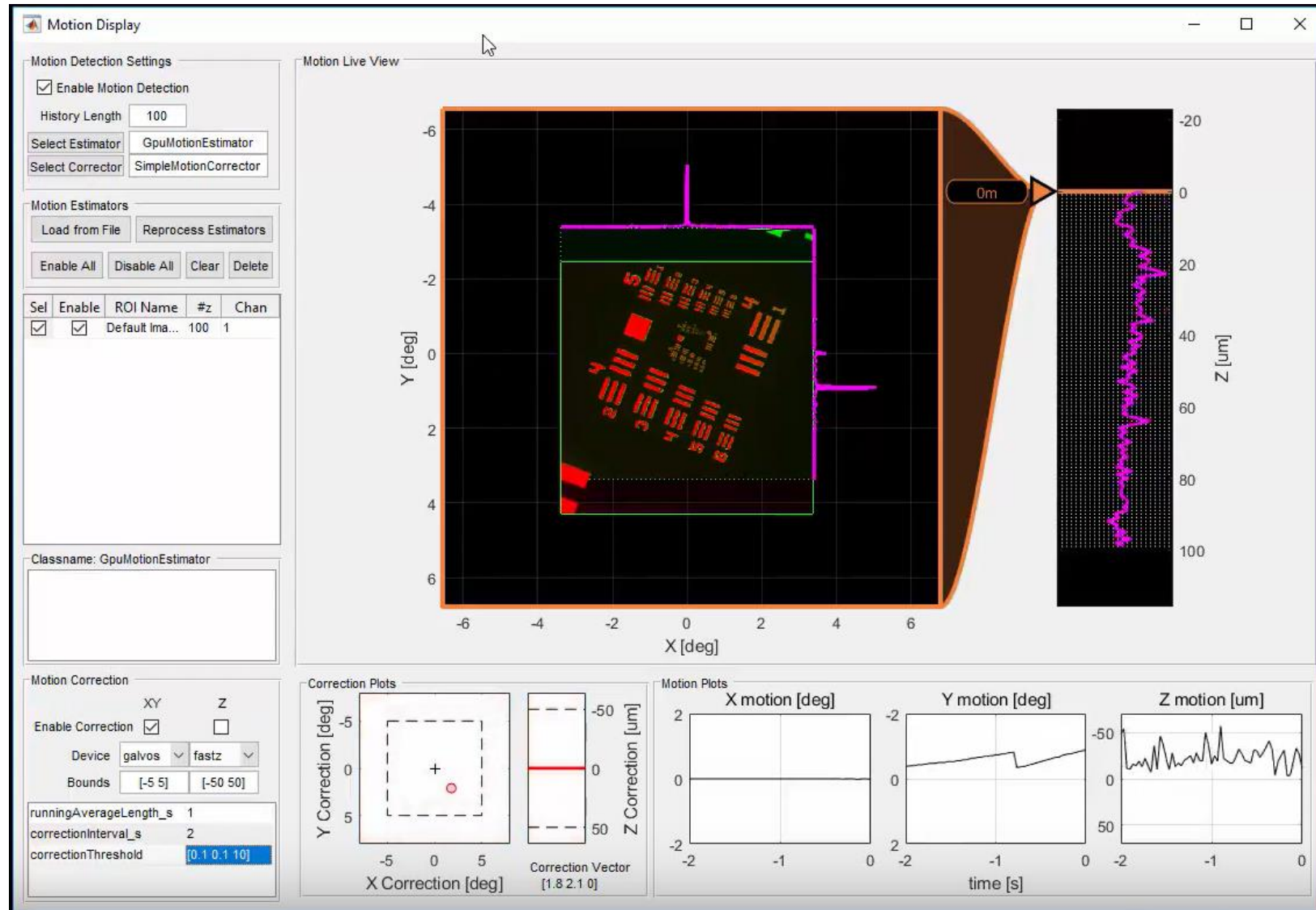
How does this drift affect neural activity?

Drift in Z

“z-corrected”



Scanimage 2018 (free version) has automated z-drift correction!



Motion correction summary

- Be aware of different types of brain movements and how they might affect neural activity
- Perform online Z correction (if possible)