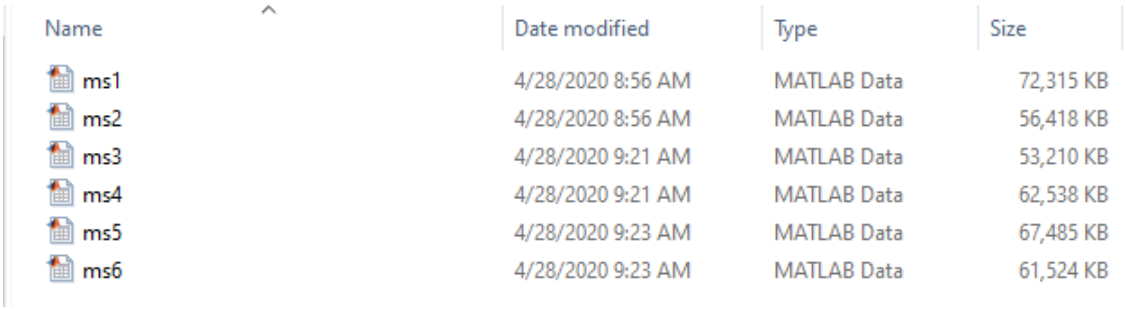
Custom CellReg Manual

How to run:

1. Set up your data in a separate folder and number them in chronological order.



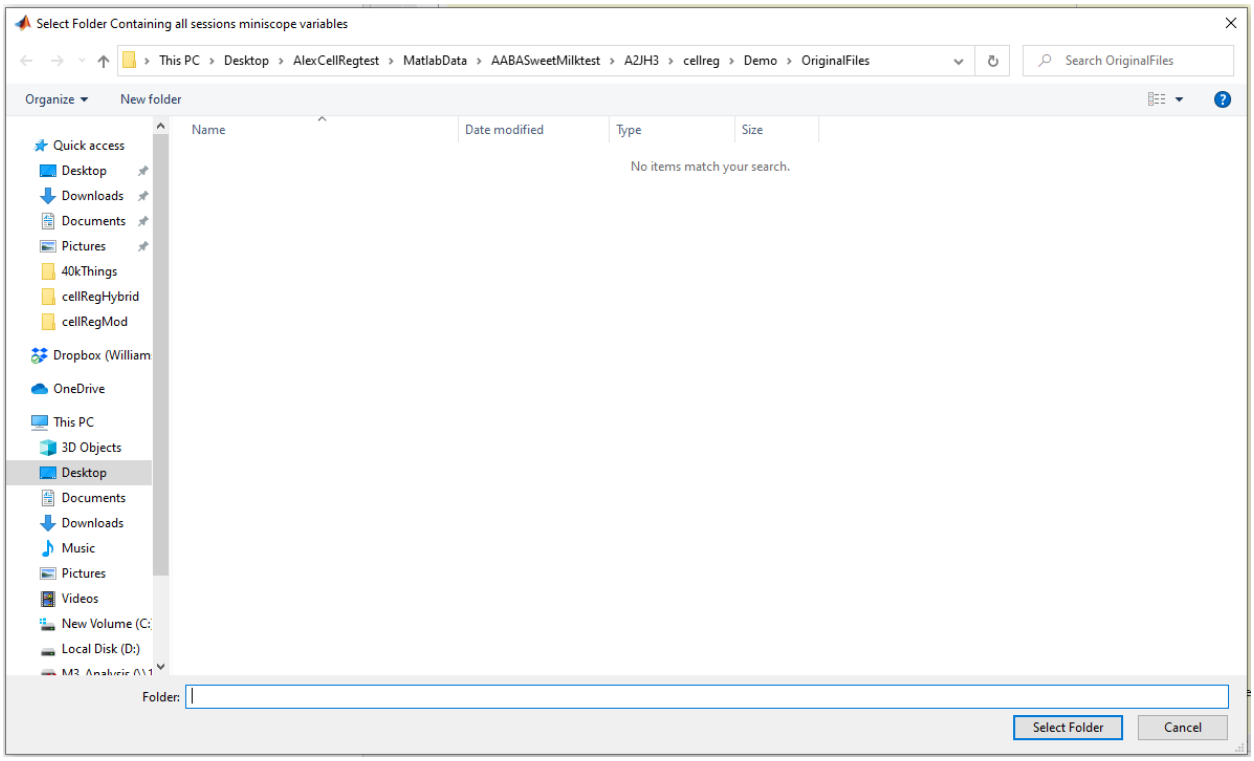
Name	Date modified	Type	Size
ms1	4/28/2020 8:56 AM	MATLAB Data	72,315 KB
ms2	4/28/2020 8:56 AM	MATLAB Data	56,418 KB
ms3	4/28/2020 9:21 AM	MATLAB Data	53,210 KB
ms4	4/28/2020 9:21 AM	MATLAB Data	62,538 KB
ms5	4/28/2020 9:23 AM	MATLAB Data	67,485 KB
ms6	4/28/2020 9:23 AM	MATLAB Data	61,524 KB

The script should be able to take any naming format so long as they are sorted numerically like above. The analysis is designed to work with CNMFe output, specifically in Alex's or Guillaume's data format.

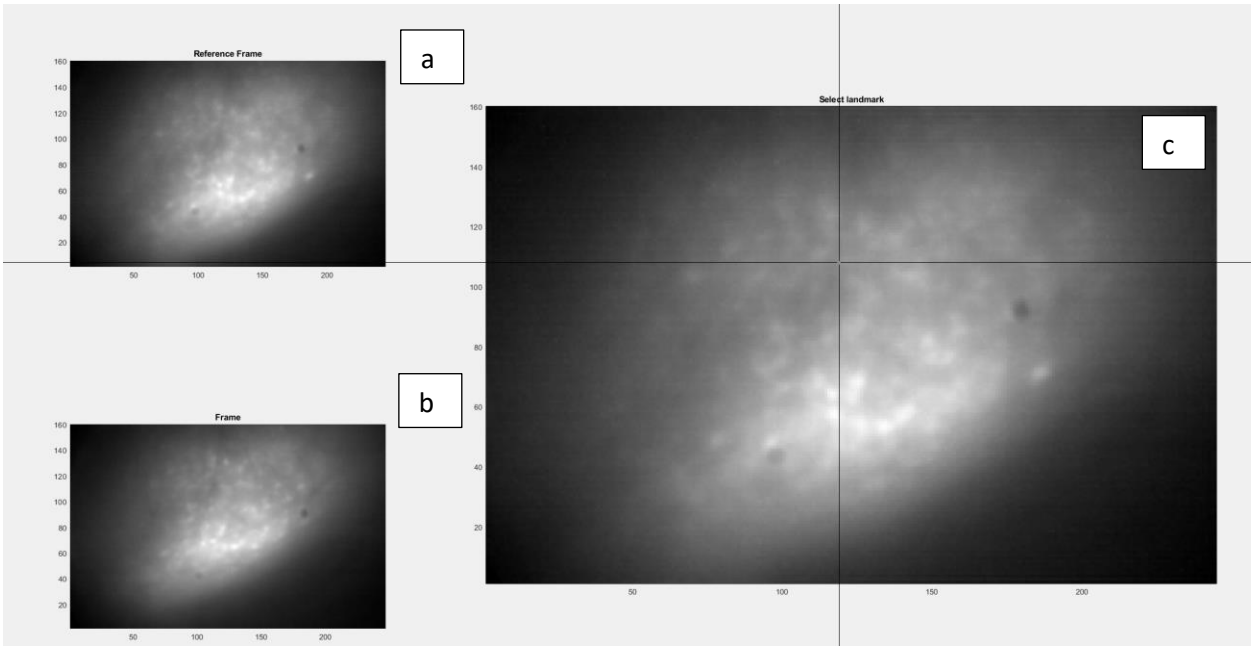
2. After adding the scripts to your path, run analysis script: CellReg_2020Execute.m

Run >> CellReg_2020Execute

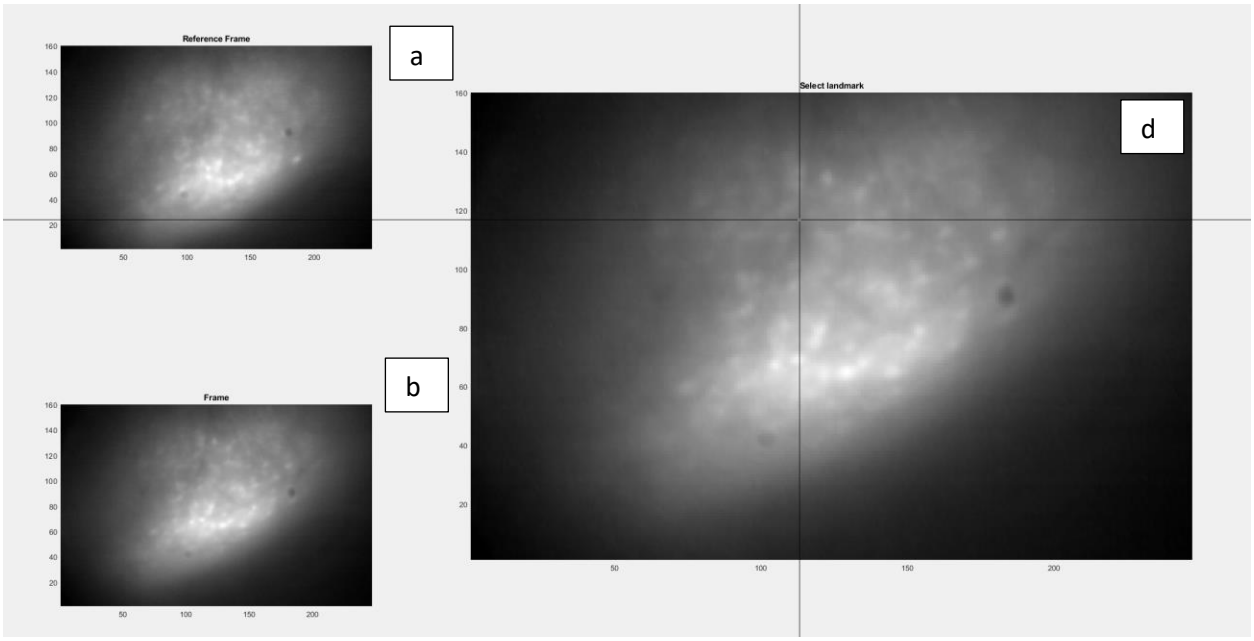
3. A prompt will come up asking for the location of the folder in question containing the files you want registered.



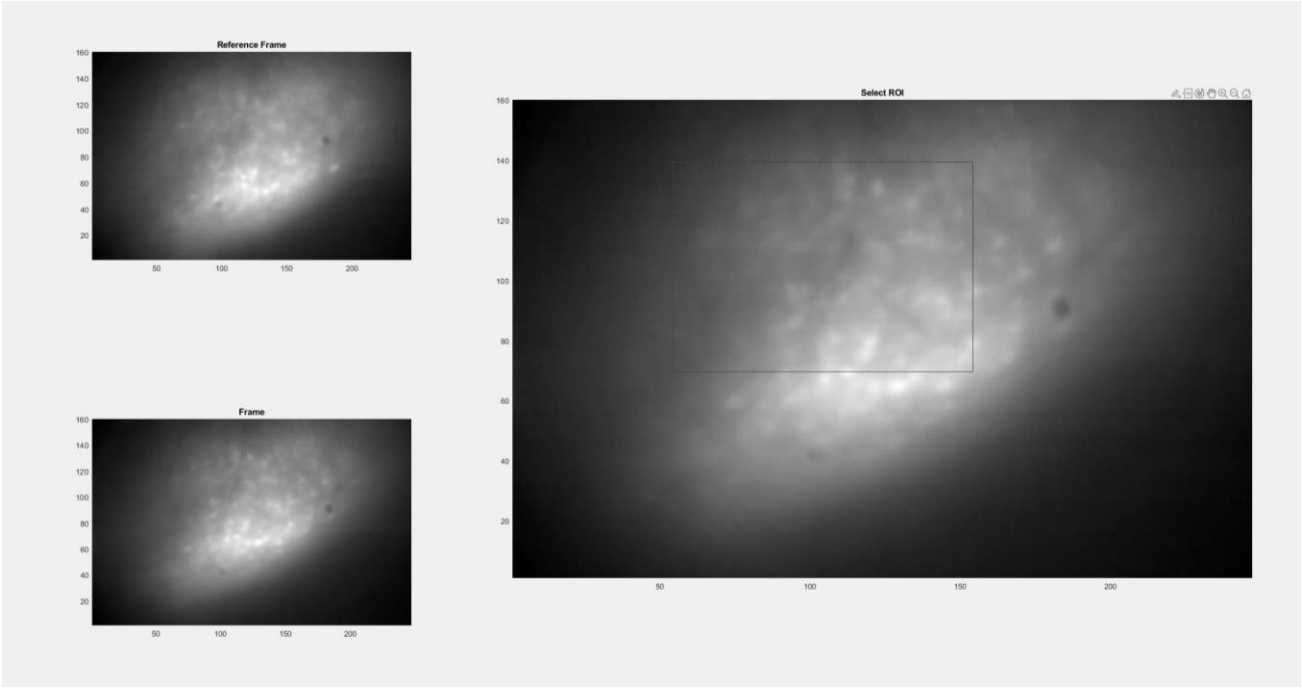
4. After selecting you analysis folder, it will duplicate your data for safe keeping under "OriginalFiles" before applying shifts in the FOV across all sessions. Once it loads the sessions, you will then be asked to select a landmark.



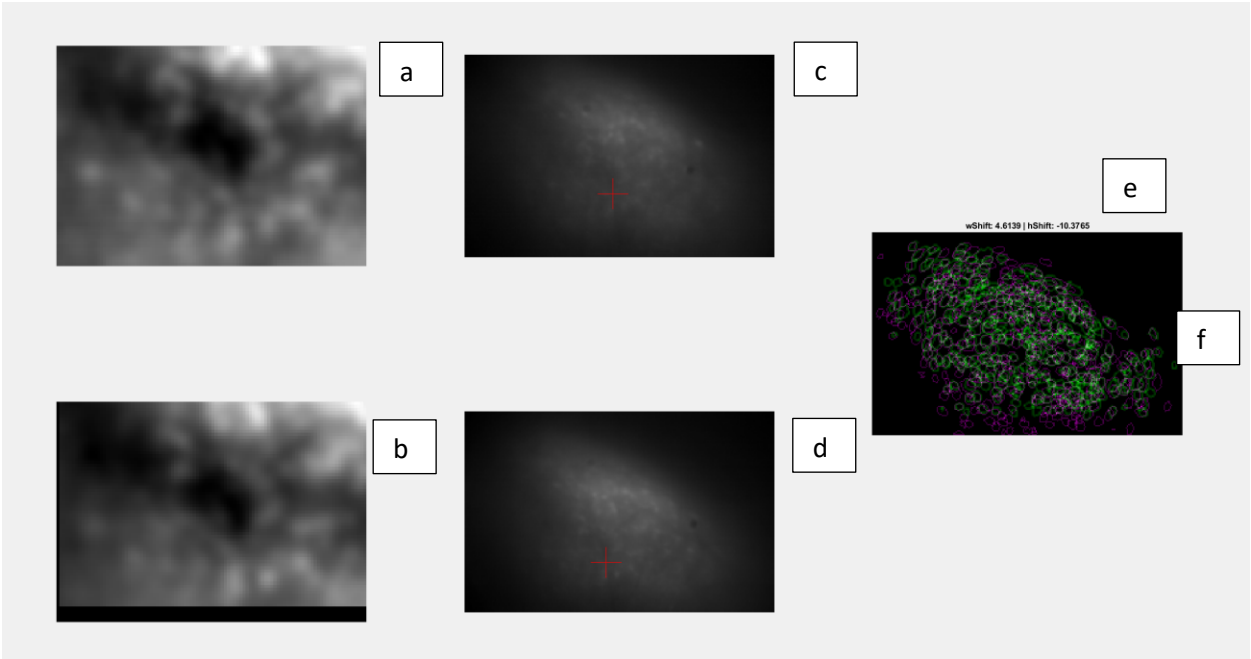
Where **a** represents the FOV of the first session, **b** the second session and **c** an enlarged view of frame **a**. The crosshair represents the location of a common landmark which is selected by the user. After selecting the landmark in the first session in window **c** the process will be repeated for the following session as seen in **d** below, which is an enlarged view of **b**.



5. The following step requires the selection of an ROI, which will be used to correlate landmarks. The ROI must contain the point of interest selected in step 4 and should contain no more than half the FOV. The code will break if the ROI is too large may may miss align if the ROI is too small. Generally you will want he ROI to contain as many distinct landmarks as possible while avoiding artifacts (like dust particles on the lens).



6. This step you verify the shift across the sessions.



Where **a** and **b** are the ROI's of both sessions that were matched, **c** and **d** the location of the ROI's in their respective FOV's, **e** the shift in both width and height in pixels and finally **f** the overlapping footprint segments.
In your command window you will have a prompt which you will answer (y,n or s) based on how you like your results:

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Do you want to keep these results? Y/N/S:
```

Y: Yes, you keep the results and move on to the next pairwise comparison.

N: No, repeat the process on the same sessions.

S: Skip, will proceed to the next session pair without applying any shift. Avoid this option if this possible since it will affect all sessions following it.

After selecting your response, press ENTER and repeat these steps until you have done all neighbouring session pairs.

7. Select Registration method: Do you want Rigid or Non-Rigid registration?

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Non-Rigid Registration?:Y/N
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By selecting Y it will run the Non-Rigid alignment and dynamically adjust each cell in order to find the best fit. Selecting N will run the Rigid analysis based on Alex's previous registration method.

After choosing which is your preferred method of alignment press ENTER and the analysis will start.

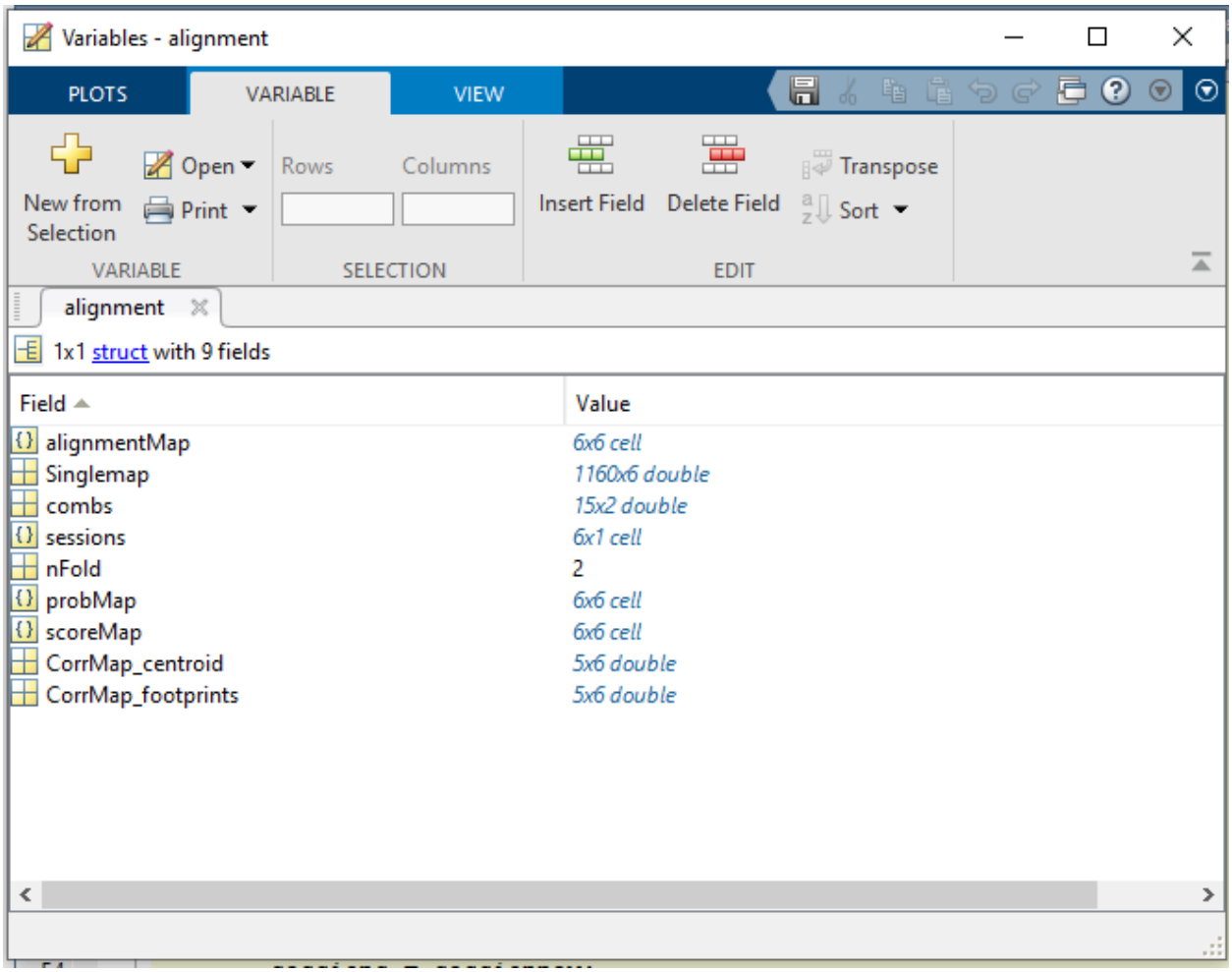
Results: What do they mean?

The output will be found within the first datafile, which in our case is ms1, under the name of alignment. Figures will also be outputted in a "Results" folder in the same directory as the data in question. Under the Results folder you will find subfolders numbered in order of pairwise comparisons across sessions.

1_2	5/7/2020 9:43 AM	File folder
1_3	5/7/2020 9:45 AM	File folder
1_4	5/7/2020 9:46 AM	File folder
1_5	5/7/2020 9:48 AM	File folder
1_6	5/7/2020 9:50 AM	File folder
2_3	5/7/2020 9:51 AM	File folder
2_4	5/7/2020 9:53 AM	File folder
2_5	5/7/2020 9:54 AM	File folder
2_6	5/7/2020 9:56 AM	File folder
3_4	5/7/2020 9:57 AM	File folder
3_5	5/7/2020 9:59 AM	File folder
3_6	5/7/2020 10:01 AM	File folder
4_5	5/7/2020 10:02 AM	File folder
4_6	5/7/2020 10:04 AM	File folder
5_6	5/7/2020 10:05 AM	File folder

Where each number corresponds to its respective sessions. Within each folder you will find the figure outputs for CellReg. This way you can go over the registration to make sure it was done correctly.

Next we will take a look at the output variable "alignment" which you will find as a subvariable in your first data file (ms1). This is what you will find:



Within alignment you can find the following variables:

alignmentMap:

	1	2	3	4	5	6
1	[]	533x2 double	477x2 double	498x2 double	679x2 double	458x2 double
2	[]	[]	538x2 double	557x2 double	799x2 double	698x2 double
3	[]	[]	[]	531x2 double	505x2 double	460x2 double
4	[]	[]	[]	[]	665x2 double	506x2 double
5	[]	[]	[]	[]	[]	530x2 double
6	[]	[]	[]	[]	[]	[]

This variable is a cell matrix containing the pairwise registration across each day. Where the row number is the reference session and column number the comparison session. Ie. Session 4 registered across session 6 can be found in alignmentMap{4,6}.

Singlemap:

	1	2	3	4	5	6
1	364	0	0	0	0	0
2	363	0	0	0	0	0
3	362	0	383	0	0	0
4	361	447	0	449	0	0
5	360	0	0	0	0	0
6	359	0	0	378	0	0
7	358	0	0	340	0	346
8	357	0	0	0	0	0
9	355	0	0	0	0	0
10	353	0	0	0	0	0
11	352	281	279	212	0	0
12	351	133	215	249	287	234
13	350	452	0	451	0	0
14	349	427	0	0	0	0
15	347	0	358	380	380	0
16	346	0	0	294	304	187
17	345	396	367	175	0	0
18	344	414	0	462	0	0
19	343	230	314	251	0	235
20	341	302	359	288	0	319
21	340	0	319	120	0	120

Singlemap is an amalgamation of the pairwise registration across all days. Each column represents every session in chronological order (session 1 is in the first column, etc). All numbers found across rows are registered across days, 0's represents no registration for that day. This variable will string together cells which have the highest average probability of registration and will not include uncertain combination of cells.

Combs:

	1	2
1	1	2
2	1	3
3	1	4
4	1	5
5	1	6
6	2	3
7	2	4
8	2	5
9	2	6
10	3	4
11	3	5
12	3	6
13	4	5
14	4	6
15	5	6

This variable is for analysis, linearizing all of the unique session combinations.

Sessions:

	1
1	C:\Users\blarg\Desktop\AlexCellRegtest\MatlabData\AABASweetMilktest\A2JH3\cellreg/ms1.mat
2	C:\Users\blarg\Desktop\AlexCellRegtest\MatlabData\AABASweetMilktest\A2JH3\cellreg/ms2.mat
3	C:\Users\blarg\Desktop\AlexCellRegtest\MatlabData\AABASweetMilktest\A2JH3\cellreg/ms3.mat
4	C:\Users\blarg\Desktop\AlexCellRegtest\MatlabData\AABASweetMilktest\A2JH3\cellreg/ms4.mat
5	C:\Users\blarg\Desktop\AlexCellRegtest\MatlabData\AABASweetMilktest\A2JH3\cellreg/ms5.mat
6	C:\Users\blarg\Desktop\AlexCellRegtest\MatlabData\AABASweetMilktest\A2JH3\cellreg/ms6.mat

Sessions saves the path and variable names of all sessions used for this analysis.

nFold:

nFold	2

This is how many sessions will by analyzed simultaneously (this version will only work with an nFold of 2).

probMap:

	1	2
1	0.9999	0.9999
2	0.9963	0.9963
3	0.9984	0.9984
4	0.8071	0.8071
5	0.9993	0.9993
6	0.9851	0.9851
7	0.9963	0.9963
8	0.9924	0.9924
9	0.9993	0.9993
10	0.9999	0.9999
11	0.9963	0.9963
12	0.9993	0.9993
13	0.9998	0.9998
14	0.9725	0.9725
15	0.6293	0.6293
16	0.7249	0.7249
17	0.9993	0.9993
18	0.9999	0.9999

Here probMap and scoreMap are the same thing, scoreMap is kept for ease of use for Alex's code. These variables represent how likely/confident the CellReg algorithm is in the registration of the cells.

CorrMap_centroid

	1	2	3	4	5	6
1	0	0.4298	0.4024	0.2825	0.1767	0.2045
2	0	0	0.4021	0.2887	0.1772	0.1802
3	0	0	0	0.4083	0.2298	0.2773
4	0	0	0	0	0.2928	0.3474
5	0	0	0	0	0	0.3859

Outputs the correlation of centroid location of the segment bodies. Rows and column numbers represent their respective sessions, ie. Row 1 Column 3 is Session1 vs Session3.

CorrMap_footpints

	1	2	3	4	5	6
1	0	0.6196	0.6439	0.5505	0.4419	0.4681
2	0	0	0.6356	0.5462	0.4480	0.4150
3	0	0	0	0.6497	0.5234	0.5641
4	0	0	0	0	0.5917	0.5786
5	0	0	0	0	0	0.6423

Outputs the footprint correlation for the cell bodies across the sessions.