**Manual for 3D registration of neuron morphologies**

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Installation:

The main program, called *NeuroRegistration*, has been written using the following libraries:

* C++ STL (usually ships with the compiler)
* ITK v3.20
* VTK v5.6
* GSL v1.13

*NeuroRegistration* comes with a CMakeLists.txt file. Build it using CMake from the *NeuroRegistration* folder. If you have questions about the installation, please contact Robert.

Introduction:

*NeuroRegistration* is used for automated registration of reconstructed *in vivo* neuron morphologies to the standardized barrel field (SBF). It is part of a 3D neuron reconstruction and registration pipeline. The results are only meaningful if this tool is used together with *NeuroConv* and *NeuroControl*.

For further information, see Egger et al. PloS Comp. Biol. 2012.

Naming Conventions:

Anatomical structures and landmarks of the reconstruction should be named already in *NeuroLucida* and *NeuroConv*:

Neuronal structures:

Soma: soma

Axon: axon

Basal dendrite: dend

Apical dendrite: apical

Anatomical landmarks:

Pia: alpha01-alpha10

White matter: WM01-WM10

Barrels:

Alpha, Beta, Gamma, Delta, A1, A2, A3, A4, B1, B2, B3, B4, C1, C2, C3, C4, C5, C6, D1, D2, D3, D4, D5, D6, E1, E2, E3, E4, E5, E6

Important: Spelling is important – e.g., note the difference between alpha (Pia) and Alpha (Barrel)!

Also, barrel names C5/6, D5/6 and E5/6 are recognized, but not part of the SBF.

Usage:

Case 1: registration of neuron reconstruction to home column in standard barrel field

Input:

.hoc file created by *NeuroConv* and checked for consistency of Pia and WM with *NeuroControl*

Output:

.hoc file containing registered neuron morphology

.hoc file containing registered landmarks (for visualization only)

.am file containing registered neuron morphology, reconstructed 3D barrels and neuron orientation (for visualization only)

.log text file containing information about transformations used during registration, and registration results. In particular, it contains the automatically determined registered home column, Pia-soma distance, soma-column axis distance, laminar position and columnar/septal position.

Use:

[/path/to/executable/]NeuroRegistration [input-filename.hoc] [output-filename-prefix] [neuron orientation structure] [laminar position] [apical dendrite flag] [pia correction flag (optional)] [axon only mode (optional)]

The parameters can take the following values:

[neuron orientation structure]:

0: none – do not determine neuron orientation (e.g. spiny stellate cell without reconstructed axon).

1: use apical dendrite to determine neuron orientation (usually good for all infragranular cells, L4Py, and sometimes L4sp).

2: use main axon leaving soma towards WM to determine neuron orientation (works pretty reliably, suggested for use when no clear apical dendrite orientation is present, e.g. L2 or L4ss).

[laminar position]:

1: supragranular

2: granular

3: infragranular

This parameter is only used when also using the apical dendrite to determine the neuron orientation, but must be set at all times.

[apical dendrite flag]:

0: not clear – use heuristic method to determine orientation

1: clear – compute principal axes to determine orientation (very robust when clear apical dendrite is visible)

2: bi-tufted dendrite present. Only the part of the dendrite labeled as “Apical dendrite” is used to determine the neuron orientation.

[pia correction flag (optional)]:

0: set to 0 when neuron IS NOT located below top pia contour

1: set to 1 when neuron IS located below top pia contour – corrects systematic error due to pia extrapolation

[axon only mode (optional)]:

0: Off

1: On. Ignores all neuronal structures except for the axon and does not determine a neuron orientation. Important: in this mode, a home barrel has to be set manually.

Note: when using optional parameters, all parameters before the last used parameters have to be set, even if they also are optional.

Example 1: L5tt neuron

Neuron orientation is determined by computing the principal axes of the dendrites; no pia correction is necessary.

Input:

regger@marcel3:/disk1/regger/NeuroMap/tutorial/L5tt> ~/project\_src/BarrelField3D/NeuroRegistration/NeuroRegistration 87\_CDK20050131nr4L5B.hoc 87\_CDK20050131nr4L5B\_home\_column 1 3 1

Output:

Reading hoc file 87\_CDK20050131nr4L5B.hoc

Soma found in input hoc file!

Dendrite found in input hoc file!

Apical dendrite found in input hoc file!

Axon found in input hoc file!

Pia found in input hoc file!

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>>> Starting registration!

>>> Manual home barrel: D2

>>> Computing landmark reconstruction...

>>> Computing landmark registration...

>>> Found common landmark: A1

>>> Found common landmark: A2

>>> Found common landmark: A3

>>> Found common landmark: A4

>>> Found common landmark: B1

>>> Found common landmark: B2

>>> Found common landmark: B3

>>> Found common landmark: B4

>>> Found common landmark: C1

>>> Found common landmark: C2

>>> Found common landmark: C3

>>> Found common landmark: C4

>>> Found common landmark: D1

>>> Found common landmark: D2

>>> Found common landmark: D3

>>> Found common landmark: D4

>>> Found common landmark: E2

>>> Found common landmark: E3

>>> Found common landmark: E4

>>> Found common landmark: Alpha

>>> Neuron axis unit vector: [-0.00250828,-0.013052,0.999912]

>>> Computing neuron morphology transformations...

>>> Checking morphology for z-scale correction

>>> Z-scale factors: [0.843882,1.18508,1]

>>> Total column scale: 0.979501

>>> Applying inhomogeneous z-scaling

>>> Final home barrel: D2

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Writing hoc file 87\_CDK20050131nr4L5B\_home\_column\_neuron\_registered\_D2.hoc

Example 2: L2 neuron

Neuron orientation is determined by determining the direction of the main axon leaving the soma; pia correction is necessary.

Input:

regger@marcel3:/disk1/regger/NeuroMap/tutorial/L2> ~/project\_src/BarrelField3D/NeuroRegistration/NeuroRegistration 04\_RMB20040916\_nr21.hoc 04\_RMB20040916\_nr21\_home\_column 2 1 0 1

Output:

Reading hoc file 04\_RMB20040916\_nr21.hoc

Soma found in input hoc file!

Dendrite found in input hoc file!

Apical dendrite found in input hoc file!

Axon found in input hoc file!

Pia found in input hoc file!

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>>> Starting registration!

>>> Manual home barrel: D2

>>> Computing landmark reconstruction...

>>> Computing landmark registration...

>>> Found common landmark: A1

>>> Found common landmark: A2

>>> Found common landmark: A3

>>> Found common landmark: A4

>>> Found common landmark: B1

>>> Found common landmark: B2

>>> Found common landmark: B3

>>> Found common landmark: B4

>>> Found common landmark: C1

>>> Found common landmark: C2

>>> Found common landmark: C3

>>> Found common landmark: C4

>>> Found common landmark: D1

>>> Found common landmark: D2

>>> Found common landmark: D3

>>> Found common landmark: D4

>>> Found common landmark: E1

>>> Found common landmark: E2

>>> Found common landmark: E3

>>> Found common landmark: E4

>>> Found common landmark: Alpha

>>> Found common landmark: Beta

>>> Found common landmark: Gamma

>>> Found common landmark: Delta

>>> Neuron axis unit vector: [-0.0241148,-0.0618248,0.997796]

>>> Computing neuron morphology transformations...

>>> Checking morphology for z-scale correction

>>> Z-scale factors: [1.09911,0.769701,1]

>>> Total column scale: 0.969995

>>> Applying inhomogeneous z-scaling

>>> Final home barrel: D2

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Writing hoc file 04\_RMB20040916\_nr21\_home\_column\_neuron\_registered\_D2.hoc

Example 3: VPM axon

The neuron orientation is not determined and all neuronal structures except for the axon are ignored (if present). Therefore, the home barrel has to be set manually. Pia correction is not necessary.

Input:

regger@marcel3:/disk1/regger/NeuroMap/tutorial/VPM> ~/project\_src/BarrelField3D/NeuroRegistration/NeuroRegistration 10\_TCP\_RMB20070525b2\_CR\_3x3\_scaled.hoc 10\_TCP\_RMB20070525b2\_CR\_3x3\_scaled\_home\_column 0 0 0 0 1

Output:

Reading hoc file 10\_TCP\_RMB20070525b2\_CR\_3x3\_scaled.hoc

Soma found in input hoc file!

Dendrite found in input hoc file!

Apical dendrite found in input hoc file!

Axon found in input hoc file!

Pia not found in Input SpatialGraph. Could not determine z-direction.

Setting zReversed = 0

Using axon registration mode; ignoring any other cell structures!

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>>> Starting registration!

>>> Manual home barrel: D2

>>> Computing landmark reconstruction...

>>> Computing landmark registration...

>>> Found common landmark: C1

>>> Found common landmark: C2

>>> Found common landmark: C3

>>> Found common landmark: D1

>>> Found common landmark: D2

>>> Found common landmark: D3

>>> Found common landmark: E1

>>> Found common landmark: E2

>>> Found common landmark: Delta

Neuron axis invalid!

>>> Computing neuron morphology transformations...

>>> Checking morphology for z-scale correction

>>> Z-scale factors: [1,0.805167,1]

>>> Total column scale: 0.957288

>>> Applying inhomogeneous z-scaling

>>> Final home barrel: D2

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Writing hoc file 10\_TCP\_RMB20070525b2\_CR\_3x3\_scaled\_home\_column\_neuron\_registered\_D2.hoc

Case 2: transformation of registered neuron from home column to different column

Input:

.hoc file of registered neuron morphology created by *NeuroRegistration*

Output:

.hoc file containing neuron morphology transformed to different column

.log text file containing information about transformations used during registration, and registration results. In particular, it contains the automatically determined Pia-soma distance in the newly registered column.

Use:

[/path/to/executable/]NeuroRegistration [input-filename.hoc] [output-filename-prefix] [new column name]

Example: Transform L5tt neuron from D2 to C2

Input:

regger@marcel3:/disk1/regger/NeuroMap/tutorial/L5tt> ~/project\_src/BarrelField3D/NeuroRegistration/NeuroRegistration 87\_CDK20050131nr4L5B\_home\_column\_neuron\_registered\_D2.hoc 87\_CDK20050131nr4L5B\_different\_column C2

Output:

Reading hoc file 87\_CDK20050131nr4L5B\_home\_column\_neuron\_registered\_D2.hoc

Soma found in input hoc file!

Dendrite found in input hoc file!

Apical dendrite found in input hoc file!

Axon found in input hoc file!

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>>> Starting registration to column C2

>>> Checking morphology for z-scale correction

>>> Z scale factors: [0.935411,1.01384,0.958692]

>>> Total column scale: 0.962435

>>> Applying inhomogeneous z-scaling

>>> Final home barrel: C2

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Writing hoc file 87\_CDK20050131nr4L5B\_different\_column\_registered\_C2.hoc

Questions? Suggestions? Bugs?

Please let me know…