Combining high resolution MRI and DTI with dense whole-brain histology for mouse

Cold Spring Harbor Laboratory
Cold Spring Harbor, NY

V. Pinskiy
A. Mukherjee
A. Tolpygo
A. Badea
A.G. Johnson
P. Mitra

Mouse Brain Architecture

Overview

Objective

With advances in digital imaging and computing technologies, there is increasing demand to cross-reference and freely share whole-brain image data sets of different modality. The Waxholm Space (WHS) digital atlas, now in its 3rd year and widely adopted, was developed to meet this demand by providing a common framework for whole mouse brain digital neuroanatomy.

The work we present is aimed at updating the histological component of the current WHS atlas with improved spatial resolution and image quality. We co-register the high-resolution whole-brain Nissl with simultaneously scanned MRI/DTI that represent significant advance in the quality of histological imaging for mouse brains. It will be made available online through the Mouse Brain Architecture Project website at http://mouse.brainarchitecture.org.

Waxholm Space (WHS)

Ultra-high resolution MR atlas of the C57BL/6 mouse brain, co-registered with a whole-brain, segmented and annotated, Nissl-histology map of the same brain.

Motivation

The existing Nissl map (obtained as horizontal stack) of the WHS can be improved upon by

- better preservation of tissue integrity
- better image quality
- higher digital spatial resolution – (current: 21 µm section thickness; 9.9 µm pxls)
- more accurate 3d-to-3d registration to MR atlas

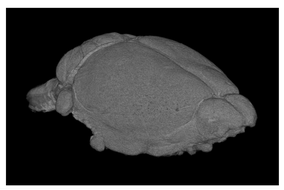
MR Methods & Data Sets

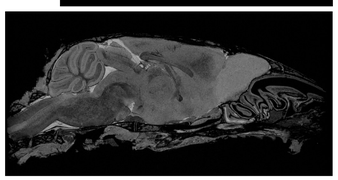
Experiment

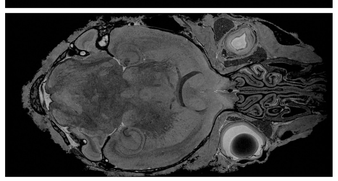
species	C57BL/6 mice
age / sex	adult (P56) male
brain preparation	post-fixed intracranial
fixative	10% buffered formalin (+ contrast agent)

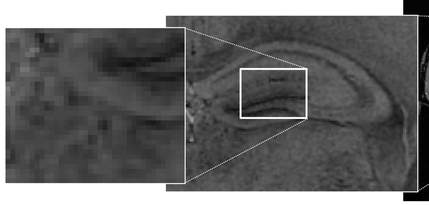
MR data acquisition

protocol	T2* -weighted
sequence	gradient-recalled echo
TR	50.0 ms
TE	4.3 ms
field strength	9.4 T
coil	12 mm
gradient	860 mT/m
contrast enhancement	high-proton density active stain (gadopentetate dimeglumine)
acquisition matrix	768 x 512 x 512
display matrix	1024 x 512 x 512
image volume	3mm isotropic cube
voxel resolution (MRI)	21.4 µm isotropic
voxel resolution (DTI)	43 µm isotropic









MBA Pipeline

After the MR imaging, the perfusion-fixed brains of the same animals were histologically processed using the automated Mouse Brain Architecture Project (MBA) histological pipeline. The histological data set consists of a thin-sectioned, Nissl-stained, whole-brain, coronal stacks.

- brain inside the lightly decalcified skull cryo-sectioned (10 µm thickness)
- sections (skull+brain) transferred to slide using the tape-transfer system
- sections were Nissl stained on slide.
- whole slides imaged using the NanoZoomer 2.0HT (0.46 µm/pixel in plane)

Nissl (in Skull) Data

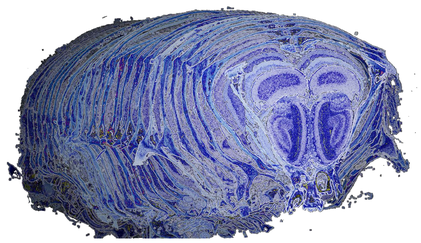
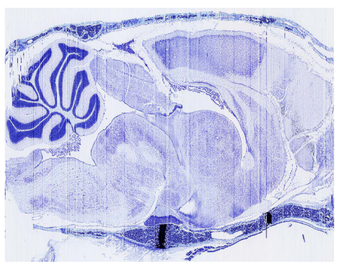
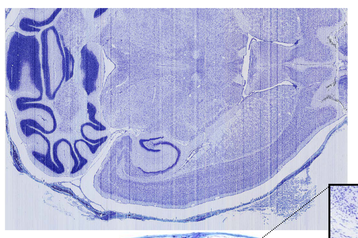
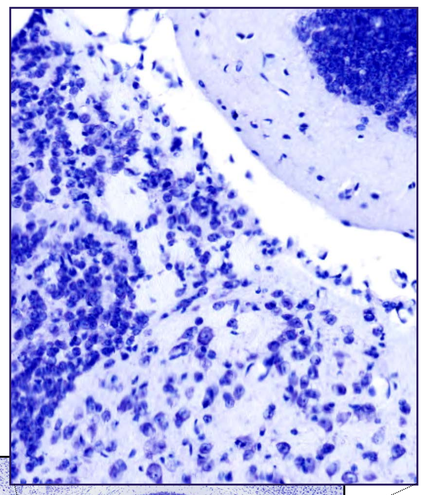


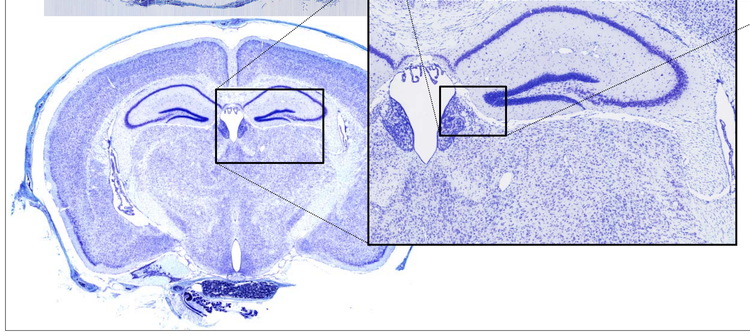
Image Stack

Sections: coronal (10 µm thickness)
Volume: ~1000 sections per brain
pxl resolution: 0.46 µm









MR-to-Nissl Co-Registration & Atlas Segmentation

Data preprocessing

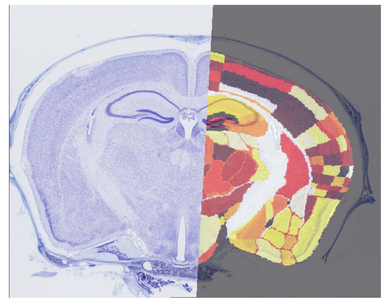
- Nissl image stack 2d-aligned (rigid transformation of adjacent sections)
- Nissl images subsampled to 20 µm (to approximate MRI voxels size of 21.4 µm)
- Nissl stack converted to inverted gray-scale (to compare with MR modality)
- MRI & Nissl stacks skull stripped by morphological operations

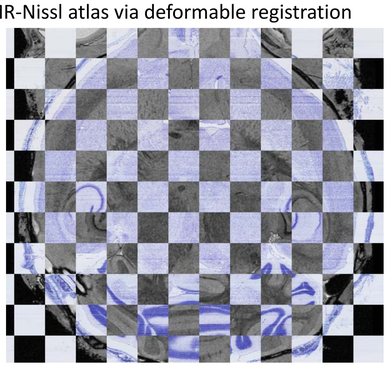
Multimodal deformable registration [1]

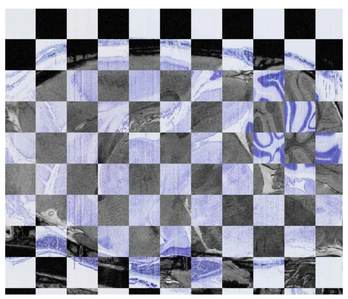
- Step 1: Affine transformation
- Step 2: Free-form deformation estimated on a 3D grid
- 3D Grid-size: 240 µm cube
- Resampling : cubic-splines
- Multi resolution : 5 levels
- Objective function: maximized normalized mutual information

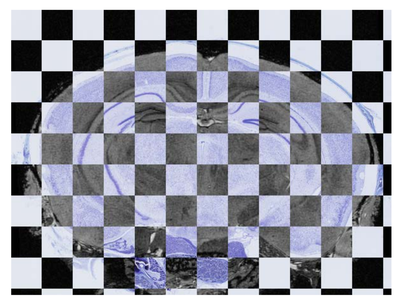
Atlas segmentation

- mouse brain reconstructed with high resolution from serial block-face tomography & annotated by co-registration with Allen Reference Atlas (P. Osten Lab; CSCHL)
- Annotated mouse brain morphed to MR-Nissl atlas via deformable registration









Reference
[1] S. Klein, M. Staring, K. Murphy, M.A. Viergever, J.P.W. Pluim, "elastix: a toolbox for intensity based medical image registration," IEEE Transactions on Medical Imaging, vol. 29, no. 1, pp. 196 - 205, January 2010.

Future Work

- Repeat procedure on multiple animals
- Deformable registration to be aided by salient anatomical landmarks
- Improve rostro-caudal resolution via optical re-slicing
- Assemble and publish high-resolution Nissl component of WHS

Acknowledgements

We gratefully acknowledge funding support from the

- NIH (Directors Transformative Award MH087988, DA036400)
- NSF (Inspire Award PHY-1344069)
- Mathers Foundation
- Crick-Clay Professorship to Dr. Mitra
- CSHL internal funding.