

High-resolution adult zebrafish brain model

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Introduction: The Zebrafish is an important model used to study a wide variety of neurological diseases such as epilepsy and Alzheimer's disease. Embryonic zebrafish are widely used to study development as they are transparent and easily imaged using bright field and fluorescence microscopy. Adult zebrafish, however, are opaque and require other imaging techniques such as MR microimaging to measure disease-related changes of the brain microstructures.

In this project, we developed a high-resolution probabilistic wildtype adult zebrafish brain model using T_1/T_2^* -weighted structural imaging and super-resolution short track tract density imaging (stTDI) at 16.4T. This model will provide a template to enable quantitative measurements of the effects of environmental or genetic changes that affect brain morphology.

The adult zebrafish brain has a relatively small size brain ($4 \times 2 \times 1 \text{ mm}$), or ~64 times smaller than an adult mouse brain. Therefore, zebrafish brain atlases need to be reconstructed at high-resolution to enable accurate segmentation and interpretation of the brain microstructures. Previously, we completed an anatomical brain map segmentation of a single wildtype adult zebrafish using 3D gradient-echo MRI data acquired at 10-micron isotropic resolution¹. In this study, we present a 7-micron resolution brain model from T_1/T_2^* -weighted structural imaging and super-resolution short-track track density imaging (stTDI) data.

Method: *Sample preparation:* After anaesthesia, the dorsal cranium of an adult zebrafish was removed and the brain was incubated for 12h in 4% PFA and 0.5% Magnevist. Subsequently, brain samples were set in Fomblin for MRI using a 16.4T Bruker vertical wide-bore NMR spectrometer. *Imaging method:* (1) 3D T_1/T_2^* GE images were acquired using a 5 mm high-resolution NMR coil in a Micro5 gradient with a special holder to allow tandem brain imaging. TR/TE = 8.3/50 ms, FA=30°, NEX=16, 10-micron isotropic resolution with the acquisition time of 17h. (2) 3D spin-echo HARDI data was acquired using a 5mm solenoid coil (single brain imaging). TR/TE = 450/22 ms, 4 averages, matrix = $144 \times 54 \times 54$, 48-micron isotropic resolution, 30 diffusion gradient directions, B=5000 s/mm², two B=0 with diffusion times $\delta/\Delta = 2.5/14 \text{ ms}$, with the acquisition time of 47 h.

The final minimum deformation model AB brain is generated from 23 datasets using a modified iterative recursive registration protocol². stTDI maps (n=5) were created at 5-micron resolution using the program MRTrx 0.2.9, employing constrained spherical deconvolution and probabilistic tracking 60 million tracks³.

Results: The zebrafish AB brain model has enhanced signal-to-noise, improved contrast and resolution, which enable visualization of small tracks and brain nuclei, which were otherwise invisible in the single data. The stTDI maps provided also provide valuable means for the visualisation of colour-coded directional tracks projections in the model. Examples of the data presented are shown in Figure 1.

Conclusion: A population-based atlas of structures and white matter fibre bundles in wild-type zebrafish brains will provide an important platform to perform voxel-based comparisons between model and wild type animals. This will allow a more comprehensive account of the effects of genetic and/or environmental manipulation on brain structure.

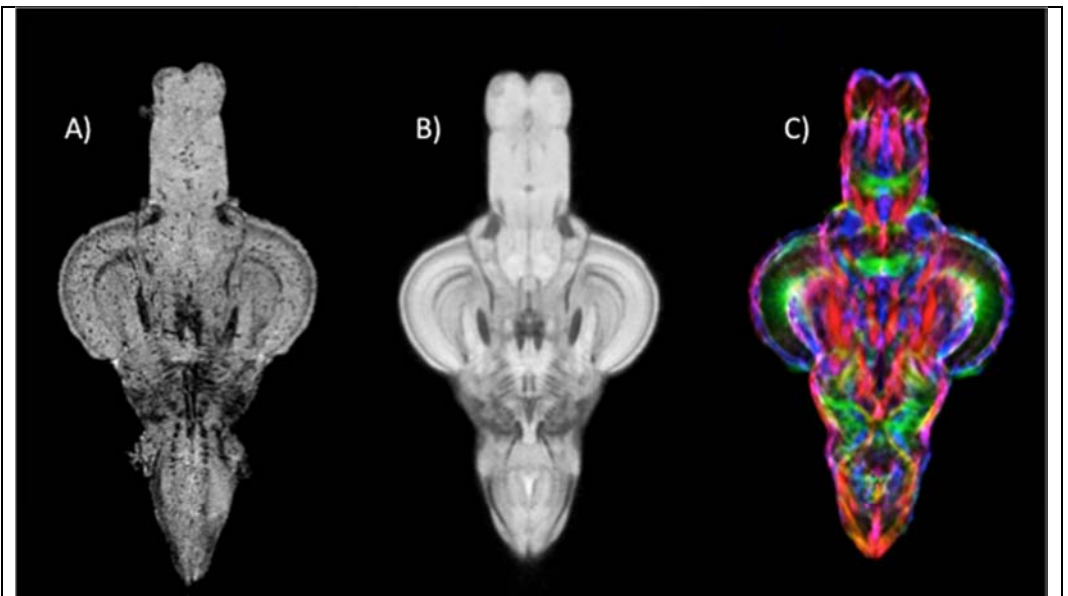


Figure 1: (A) 3D Gradient echo image of tandem zebrafish AB brain at 10-micron isotropic resolution. (B) The AB model at 7-micron isotropic resolution. (C) stTDI map. All images were shown of the same plane.

References: (1) Ullmann JFP *et al.*, A three-dimensional digital atlas of the zebrafish brain. *Neuroimage*. 2010, 51(1):76-82. (2) Ullmann JFP *et al.*, Segmentation of the C57BL/6J mouse cerebellum in magnetic resonance images. *NeuroImage* 62 (2012) 1408–1414. (3). Ullmann JFP *et al.*, Enhanced characterization of the zebrafish brain as revealed by super resolution track- density imaging. *Brain Struc Func* (Accepted 27 October 2013).