MR MICROSCOPY OF ZEBRAFISH

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Background

Zebrafish are an invaluable vertebrate system for the study of genomics, development, human disease processes, and for drug discovery. The study of adult zebrafish is currently of great interest in the field of stem cell and regenerative biology as zebrafish are able to regenerate damaged tissues such as cardiac muscle and spinal cord. Understanding how this occurs in the adult fish provides insight into new therapies for human tissue repair in conditions such as in spinal cord injury and myocardial infarction. Zebrafish are small, cheap to maintain, and as with mice, animal models of human disease can be created by genetic and environmental manipulation. Being a relatively new animal model there is no comprehensive 3D anatomical atlas onto which temporal or spatial data can be projected. Methods for in-vivo imaging of adult fish would allow functional MR imaging techniques such as MRS, MEMRI, BOLD and ASL that are used in mouse models. The major challenges are the very small size of the fish, and imaging the live fish in water. We present methods for in-vivo MRI of zebrafish, and a 3D atlas of zebrafish anatomy.

Methods and Results

For the atlas: Specifically designed volume TR coils were made, each optimized for the size of specimen being imaged (0.8mm egg to 25mm adult). Fish are euthanized, fixed and placed in perflurocarbon for imaging to maximize SNR. Gadolinium is used as required to boost SNR. Data are acquired using a 7T Bruker scanner. Isotropic voxels ranging from 15u to 30u are used for all acquisitions except for the embryonic stages (fig A). Data are segmented and 3D rendered (B). Segmented MRI data from an adult female zebrafish (20mm). Individual color-coded organs can be viewed from any angle and organ volumes calculated.

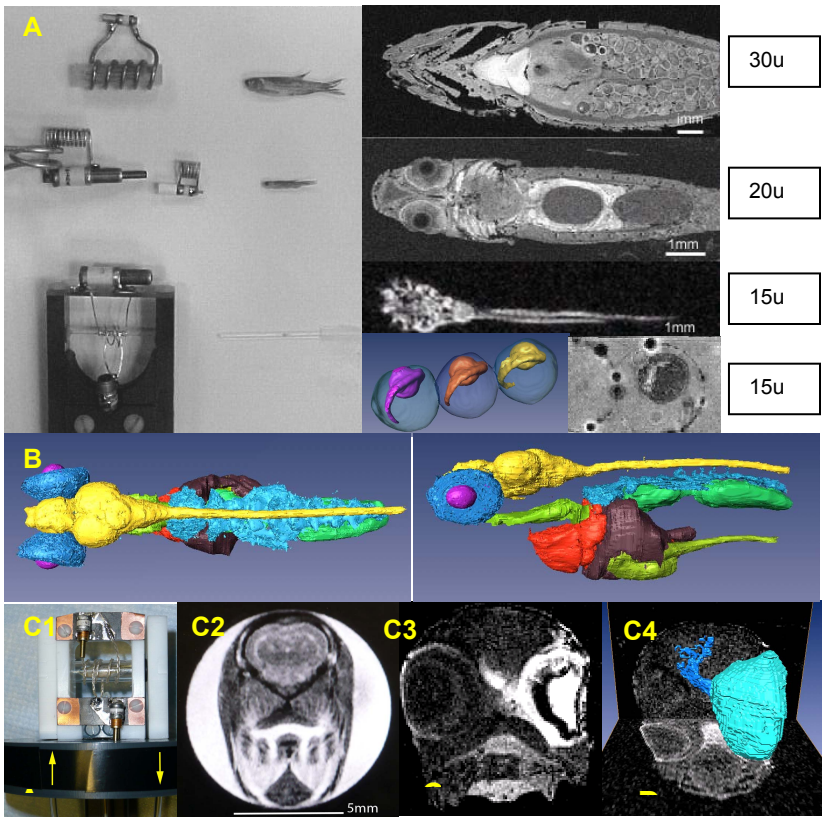

Table of volumetric data (µl) comparing 10mm and 20mm (length) adult female fish

Organ:	(10mm)Volume	(20mm) Volume
Eye (left)	0.72	3.4
Eye (right)	0.74	3.3
Lens (left)	0.05	0.29
Lens (right)	0.05	0.25
Brain	2.2	5.8
Heart	2.3	4.1
Liver	2.7	8.8

Table1: Data from segmented anatomical structures: Comparison of tissue volumes can be made between fish of varying size. There is agreement between symmetrical structures with only minimal calculated volume differences.

For in-vivo imaging We have built a specially designed system for passing water over the anesthetized fish (C1) during imaging (C2), and tested the system by successfully imaging white matter tracts using MnCl (C3,4).

Discussion and Conclusion

MRI is an ideal modality for the development of the atlas as it can provide 3D quantitative data, which cannot be attained by any other imaging modality. The ability to perform in-vivo imaging will allow researchers to acquire physiological and functional data such as MRS and manganese tract tracing. This will further accelerate the use of zebrafish for biomedical studies that customarily use rodent as animal models.