

Magnetic Resonance Microscopy of Human α -Motor Neurons and Neural Processes

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INTRODUCTION

Shortly after the development of MRI, it was theorized that microscopic resolutions would be attainable, even in the face of potential (and ultimately not apparent) microsusceptibility-based line broadening at high magnetic fields [1]. In 1986, the first imaging of a *bona fide* single cell was achieved, albeit on relatively large cells (frog ova) [2]. Until recently, SNR limitations restricted MR microscopy (MRM) of animal cells to the largest examples available: those from aquatic organisms (*Xenopus laevis* and *Aphysia californica* [3]).

In a recent study, our group was able to resolve, for the first time, cellular structure in mammalian (rat and pig) tissue using MRM [4]. Such experiments demonstrate MRM's ability to resolve the microstructural organization of mammalian tissue; however, because they were conducted in animal models, the relevance of this work with respect to clinical imaging remains an issue. Consequently, we report the first instance of cellular-level MRM on human spinal cord samples. We have also conducted higher-resolution MRM of porcine and human neurons in an attempt to resolve finer details of mammalian cellular structure. In this instance, animal tissue was employed for comparative purposes because sample preparation can be more closely regulated resulting in shorter post-mortem intervals and greater preservation of microstructure.

METHODS

MR imaging was conducted on a 600MHz Oxford spectrometer interfaced to a Bruker console. MR images of human α -motor neuron cell bodies were attained with a diffusion-weighted imaging protocol (TR = 2000ms, TE = 23.5ms, res = 7.8 μ m in-plane; b = 2000s/mm², Avg = 40, scan time = 5h40min) using a 500 μ m diameter micro surface-coil (Bruker Biospin, B6370). For MR data containing cellular processes, three dimensional spin-echo images (TR = 2000ms, TE = 12.75ms, res = 6.25 μ m isotropic, Avg = 14, scan time = 63h43min) were collected using a 100 μ m diameter micro surface-coil (Bruker Biospin, B6372). A linear smoothing step (Bruker Biospin, ParaVision 3.0.2) was used to aid in the visualization of cells and processes. Correlative histology was obtained subsequent to MRM and generated using a light microscope (Zeiss, Axioplan 2) to photograph Nissl-stained tissue sections.

RESULTS

Figure 1 shows representative (n = 3) Nissl-stained histology (A) and diffusion-weighted MR (B) images depicting cell bodies of α -motor neurons located in the ventral horn of a human spinal cord enlargement. Spatial characteristics of the cell bodies are reproduced faithfully in both imaging datasets providing a strong correlative match. Figure 2 offers higher resolution 3D spin-echo MRM of α -motor neurons taken in porcine (ABC) and human (DEF) spinal cord. These T2-weighted datasets collected at isotropic resolution (6.25 μ m) contain offshoots with similar MR contrast which are seen to extend from perikarya and which possess morphological traits consistent with cellular processes. 3D reconstructions were prepared from segmentation data (not pictured) and can be shown as videos.

DISCUSSION and CONCLUSIONS

We have reported what we believe to be the first instance of direct cellular imaging in human tissue and the visualization of cellular processes using MRM techniques. Such imaging is a necessary step towards multiple goals including an understanding of the cellular origins of MR signals in tissues, an understanding of disease-specific MR signal changes in tissues, and an ability to address cellular connectivity issues noninvasively. Future studies will involve live tissues undergoing physiological perturbation for developing mathematical models. These techniques could be used to observe microscopic changes in tissues associated with the earliest stages of disease progression and then used to interpret changes in clinical scans collected at macroscopic resolution.

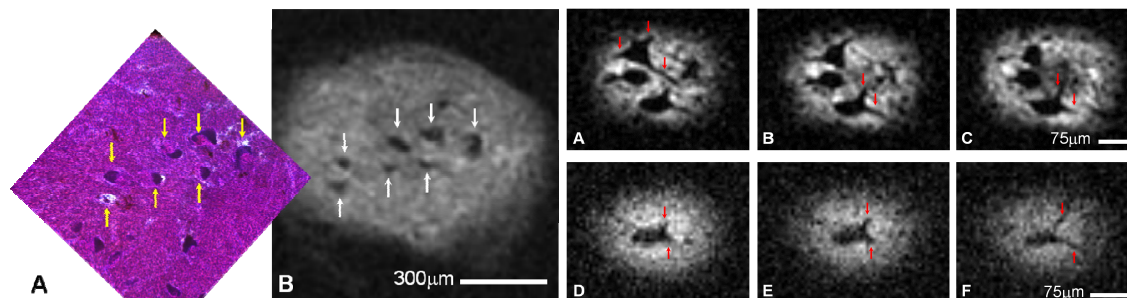


Fig 1 (A) Nissl-stained histology of a human spinal cord section (50 μ m) taken from the lumbar enlargement. Perikarya of α -motor neurons (yellow arrows) are visible and correlate spatially with hypointense structures (white arrows) in MRM (B) Diffusion-weighted MRM (res = 7.8 μ m in-plane, b = 2000s/mm²) of the tissue in (A) taken prior to Nissl staining using a 500 μ m diameter micro surface-coil.

Fig 2 Sequential images from 3D spin-echo MRM datasets (res = 6.25 μ m isotropic) showing porcine (ABC) and human (DEF) α -motor neurons. Neural processes (red arrows) are clearly visible in both datasets and manifest as hypointense structures continuous with the cell bodies.

REFERENCES and ACKNOWLEDGEMENTS

[1] Mansfield and Morris 1982 in Waugh (Ed.) NMR Imaging in Biomedicine. Advances in Magnetic Resonance. [2] Aguayo et al. Nature 1986 322:190-19. [3] Aiken, Hsu, and Blackband. J Magn Reson Anal. 1995 1:41-48 [4] Flint et al. Neuroimage 2009 46(4):1037-1040. Project funded by the NIH (1R01EB012874), the NSF through the National High Magnetic Field Laboratory, and the Danish National Research Foundation (95093538- 2458, project 100297).