Myelin visualization using q-space MRI

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Introduction

Diffusion weighted imaging (DWI) enables us to evaluate directly biological structures compared to that by other MR methodologies. In particular, diffusion tensor imaging (DTI) is a powerful tool to evaluate fibers of the white matter in the central nervous system. We previously established a reproducible spinal cord injury (SCI) model in adult common marmosets [1] and have reported that diffusion tensor tractography (DTT) enables *in vivo* tracing of the neural tracts in the intact and injured spinal cord of marmosets [2]. DTT provides information regarding the fiber orientation and connectivity ,but gives us little imformation about demyelination and remyelination after SCI. QSI[3] is one of the most recently developed DWI methodologies and is able to detect the microstructural size of biological architecture. Here we demonstrated that myelin visualization using QSI in dysmyelination mutant mice as well as demyelination and remyelination model of common marmoset. Further, we confirmed the accuracy of QSI of myelin visualization through histology.

Materials and Methods

MRI experiments: MRI was performed on a 7T PharmaScan 70/16 System (Bruker BioSpin). A 22mm (*ex vivo*) and a 60mm (*in vivo*) i.d. volume coil tuned 300.5MHz for proton resonance was used. We performed QSI with two diffusion time (15 and 200msec) and up to two q-values (400 and 2,800 cm^{-1}). MPG direction was perpendicular to the long axis in the spinal cord. After measurements were obtained, QSI analysis was performed using custom written codes in IDL@ (ITT.inc). The normalized diffusion curve $E_\Delta(b)$ was rearrangement to q-space E_Δ (q). The displacement profile P(R) was calculated from the FFT of the $E_\Delta(q)$. P(R) = FFT ($E_\Delta(q)$). We visualized myelin map based on the kurtosis of P(R).

Dysmyelination mutant mice: *Shiverer* mice (n=4), which have a spontaneous mutation of myelin basic protein (MBP) resulting in lack of compact CNS myelin [4], [5]. The mice were perfused intracardially with 4% paraformaldehyde under general anesthesia maintained by isoflurane and then the spinal cord tissues were removed. After acquisition *ex vivo* QSI, tissue preparations were stained with Luxiol fast blue (LFB) for evaluation of the myelinated area. Then immunostaining for MBP and electron microscopy were performed.

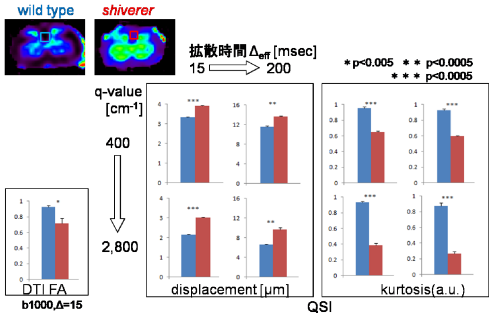

Figure 1. FA(DTI), displacement and kurtosis (QSI) in dorsal funiculus of wild type and *shiverer* mouse.

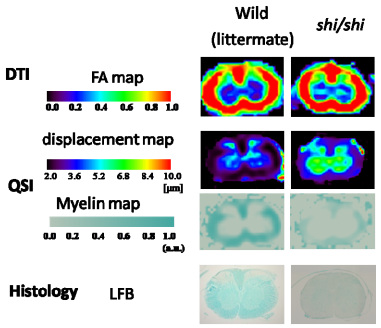

Figure 2. FA, displacement, and myelin maps and LFB stained images of spinal cords in wild type and *shiverer* mice.

Demyelination monkey model: Four adult female marmosets were used. All surgeries were performed under general anesthesia induced by intramuscular injection of ketamine and maintained by isoflurane. After laminectomy at the C5 level, 3 μ l of 1% lysophatidylcholine (LPC) was injected 1.0 mm deep into the dorsal funiculus of the spinal cord using a glass needle attached to a Hamilton Syringe[6]. QSI was performed pre- and post-injury 1, 2, and 6 weeks. After QSI, LFB staining and electron microscopy was performed. All procedures were approved by the Animal Ethics Committee of Central Institute for Experimental Animals.

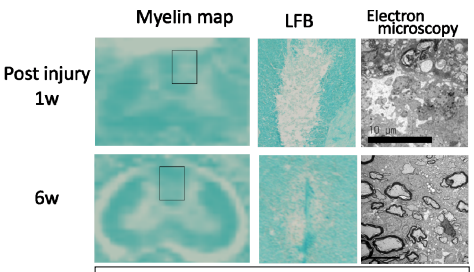

Figure 3. Myelin map, LFB stained and electron micrograph in spinal cord injury model of marmoset.

Results and Discussions

In figure 1, Kurtosis showed the difference of myelinated area between wild type and *shiverer* more sensitively than other parameters. And using higher q-value, the dufference of tow gtoupes was more increased. However, there was no signifivant difference between wild type and shiverer in the length of diffusion time. Based on these results, it is thought that there are optimal parameters in QSI which can be detected myelin existence. In figure 2, myelin maps, obtained from optimized QSI, are remarkably similar to the LFB positive myelinated area reflecting the differences between wild and *shiverer* mice.

One week after injury, low signal area of the dorsal funiculi in myelin map correspond to LFB negative demyelinated area and we confirmed demyelination with EMG. Further, 6w after injury, small foci were observed on LFB staining, and we could also observe remyelination shown by the existence of a thin myelin sheath on EMG. *In vivo* myelin mapping in chemical spinal cord injury model depicted both demyelination and remyelination clearly with time.

Conclusion

We succeeded in depicting *in vivo* myelin visualization by focusing on the strong restriction of water diffusion in myelin architecture. Applying myelin mapping to mice with dysmyelination and marmosets' chemical spinal cord injury model with demyelination and remyelination, we demonstrated that myelin mapping depicts the existence of myelin. Myelin mapping using q-space MRI will be a powerful tool with tremendous potential for investigating the pathological conditions underlying various nerve diseases.

References [1] *Iwanam and Yamane et al: J Neuro. Res. 2005.* [2] *Fujiyoshi and Yamada et al: J Neurosci. 2007.* [3] *Assaf, et al: NMR Biomed. 2002.* [4] *Bird et al: J Neurochem. 1978.* [5] *Privat et al: Neurosci Lett. 1979.* [6] *Watabnabe et al: J neurosci Res. 2002.*