# Rapid and high-resolved Diffusion Tensor Imaging of mouse lower brain and cervical spinal cord

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## Introduction

Mouse diffusion tensor imaging (DTI) of both the brain and the spinal cord (SC) may reveal useful information on local or general tissue damage consequent to inflammatory or degenerative diseases. The alteration of the corticospinal tract (CST) along the course of the tract, following stroke or amyotrophic lateral sclerosis (ALS), is a good illustration of such damage.

To rapidly assess the geometrical and functional extent of the pathology, a large coverage of the central nervous system, with sufficient spatial resolution, is required. However, in mouse SC models, standard pulse sequences preclude obtaining such requirements as they do not allow achieving sufficient signal to noise ratio within an acceptable time frame.

In order to cover both the brain and the cervical SC in a minimal scan time, we propose to use a spin-echo (SE) EPI-based sequence [1]. Our goal was to obtain spatially resolved images of both the cerebellum and cervical SC within 30 minutes. The high temporal resolution of EPI has also been exploited to acquire nearly isotropic voxel and obtain 3-dimensional DTI reconstructions that may help to better characterize specific area such as the region of the pyramidal decussation (fig.1a and b).

## Materials and Methods

C57BL/6J mice (25-30 g) were anaesthetized with an isoflurane+air mixture and placed in a transmitting/receiving birdcage coil (diameter 2 cm, homogeneous length 3 cm). All experiments were performed on an 11.75T vertical Bruker Avance 500 WB system, in agreement with the guide for care and use of laboratory animals. The DTI acquisitions were performed using a 4-shots spin-echo EPI sequence [1] and the following parameters: TE/TR 14.25/500 ms, acquisition matrix 128/128, field of view 1.8*1.8 mm², $\Delta/\delta$ 6.82/2.3 ms, 6 diffusion encoding directions, b={0, 700} s/mm². In order to cover the region from the cerebellum to the C7 segments (fig. 2a), 15 axial slices were acquired (0.75-mm thickness, no inter-slice gap, 8 signal averaging (NEX), DTI acquisition time 1.9 minutes/slice, voxel resolution 140x140x750 $\mu m^3$). The benefit of acquiring nearly isotropic voxel size images and obtaining 3-dimensional DTI views was also investigated by collecting 0.15-mm thickness axial images (19 slices, total extent 2.85 mm (fig. 2b), voxel size 140x140x150 $\mu m^3$, 40 NEX, DTI acquisition time 9 minutes/slice). DTI metrics (diffusivity, fractional anisotropy, eigenvector maps) and fiber tracking were processed using Brainvisa software.

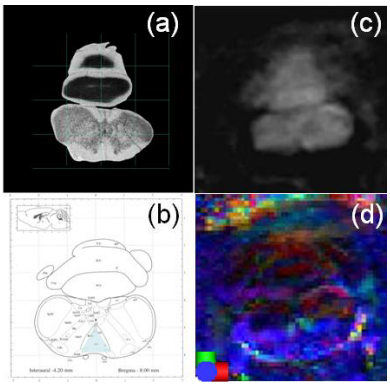


***Fig. 1 – Axial section of the mouse brain at the pyramidal decussation level (pyx): (a) histological slice from [2], (b) drawing from [3], (c) DWI and (d) eigenvector map collected with our SE-EPI DTI protocol.***

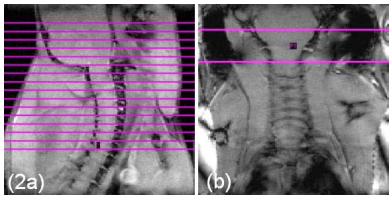


***Fig. 2. – (a) Location of the 15 0.75-mm thickness EPI axial slices (sagittal scout). (b) Pack-extent of the 19 0.15-mm thickness EPI axial slices (coronal scout).***

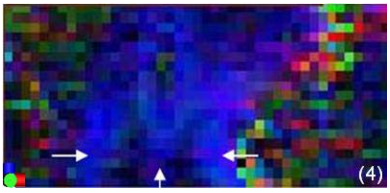


***Fig. 4 – Coronal eigenvector map reconstruction. (voxel size 140x140x150 $\mu m^3$) corresponding to the pack extent indicated in fig 2b. Horizontal and vertical arrows respectively indicate lateral and ventral tracts.***

## Results

Examples of 140x140x750 $\mu m^3$ mouse diffusion-weighted (DW) images and eigenvector maps (cerebellum, medulla, cervical spinal cord) are given on figure 3. The total acquisition time required to cover the lower brain and SC region (fig. 2a) equals 28 minutes. Diffusivity and fractional anisotropy measured in the SC agreed with the literature values (data not shown). Standard deviations of the mean DTI values do not exceed 15%.

The 140x140x150 $\mu m^3$ axial acquisitions allowed to reconstruct good quality sagittal and coronal DTI images. An illustration of a coronal eigenvector map where lateral and ventral tracts can be easily seen is given on figure 4.

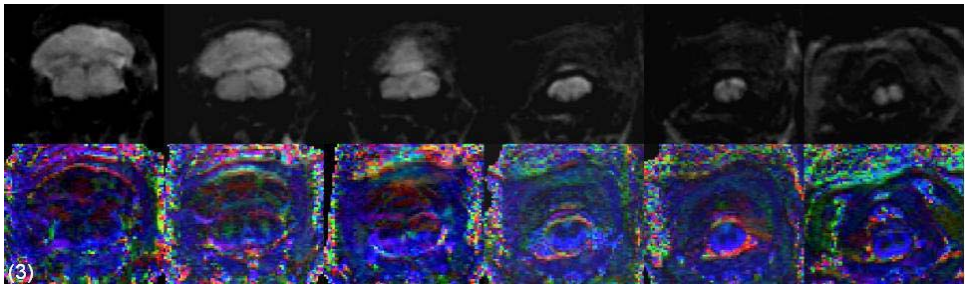


***Fig. 3 – DW images (top) and eigenvector maps (bottom) at different levels (cerebellum, medulla, spinal cord). Fibers running in the cephalocaudal direction were shown in blue, those in the anteroposterior direction in green, and those in the left-right direction in red.***

## Discussion

DTI metrics are expected to be significantly different in injured cords relative to uninjured controls. In this study, the standard deviation observed on FA and diffusivity measurements (<15%) is compatible with the detection of reported white matter (WM) alteration (~30% [4,5]). Moreover, the large coverage offered by this protocol is well suited to follow the extension of degenerative or inflammatory diseases that propagate through brain and SC.

Increasing the spatial resolution may improve the angular sensitivity in the fiber tracking process and enables the visualization of smaller and subtler fibers. The isotropic protocol is therefore quite appealing to test the possibility of visualizing CST in mouse, although the acquisition currently requires long scan time (almost 3 hours). Such experiments may eventually complete manganese-enhanced (ME)-MRI [6].

## Conclusion

Spatially resolved DTI acquisitions (140x140x750 $\mu m^3$) were performed in a region ranging from the mid-cerebellum to the C7 segment in less than 30 minutes. This protocol may be used to rapidly investigate the integrity or extent of WM damage. A protocol that allows nearly isotropic reconstruction (140x140x150 $\mu m^3$) was also performed to attempt a visualization of the CST decussation and improve the fiber tracking.

With this DTI-EPI tool, one can non-invasively and longitudinally monitor the progression of numerous intrinsic inflammatory and degenerative disorders that affect spinal cord tract integrity (multiple or amyotrophic lateral sclerosis, spinal cord injury…) within a scan time that is compatible with animal anesthesia procedure. Further experiments, under pathological conditions, are under investigation.

## References

[1] Callot *et al*., Magn. Reson. Mater. Phy, Magma (2007);[2] http://www.mbl.org; [3] The Mouse Brain in Stereotaxic Coordinates, Elsevier Academic Press, 2004, p.98; [4] Kim *et al*., Neurobiol. Dis. (2006); [5] Gaviria *et al*., Neurobiol. Dis. (2006); [6] Bilgen *et al*., J. Neurosci Meth. (2006).