Diffusion Tensor Imaging Reliably Detects Experimental Traumatic Axonal Injury and Indicates Approximate Time of Injury

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Introduction: There are an estimated incidence of 1.5 million new Traumatic Brain Injury (TBI) cases / year in the US, 50,000 are fatal. A total of 5.3 million Americans—2% of the U.S. population—currently live with disabilities resulting from TBI costing an estimated at $56.3 billion per year (Thurman, J of Head Trauma Rehabilitation 14 602-15 1999) and these numbers are set to rise due to military conflicts in Iraq and Afghanistan. Diffuse axonal injury (DAI) is thought to be a major contributor to cognitive dysfunction in patients following TBI. Diffusion Tensor Imaging (DTI) has been proposed as a technique to detect and differentiate axon and myelin degeneration, We have previously shown that DTI appears to be more sensitive to axonal injury than conventional MR imaging in a mouse model of TBI (Mac Donald et al, Exp Neurol 2007). DTI has also been proposed as potentially a new tool for earlier detection of injury and a prognostic measure of subsequent brain damage. To address this, we quantitatively compared DTI signal abnormalities with histological and electron microscopic characteristics of severe axonal injury in a well-characterized mouse model across multiple time points and varying degrees of injury severity at a single acute time point.

Methods: Adult, wild type mice were subjected to either a mild, moderate, or severe controlled cortical impact TBI. Animals were allowed to fully recover and then imaged, following the methods of Song et al, Neuroimage 2003, in a 4.7T scanner (Oxford Instrument 200/330) with an actively shielded gradient coil (180 mT/m, 400 µs rise time). Mice were imaged in vivo with DTI either at 4-6 hours, 24 hours, 4 days, 1 week, or 1 month post-injury (3s TR, 43ms TE, 25ms time between gradient pulse application, 10ms diffusion gradient duration, 0.5mm slice thickness, 2cm field-of-view). Mice were sacrificed and stereological techniques were employed to systematically quantify axonal injury as evidenced by amyloid-beta precursor protein and light-chain neurofilament immunostaining, as well as immunoreactive astrocytes noted with an antibody to GFAP on tissue slices following injury. Regions of interest for the quantitative analysis of DT images were chosen based on the spatial distribution of axonal injury, using anatomical landmarks visible on both MR images and histology. A subset of tissue was processed for electron microscopy.

Results: We found that at 4-6 hours and 24 hours there was a marked increase in axonal injury accompanied by statistically significant decreases in relative anisotropy and axial diffusivity but no change in radial diffusivity. At 1 week and 1 month, axonal injury was clearly present, though quantitatively reduced relative to the more acute time points. At these sub-acute time points, axial diffusivity had normalized whereas radial diffusivity was significantly increased and relative anisotropy was decreased relative to control. At all time points, DTI was more sensitive to axonal injury than conventional MRI and relative anisotropy distinguished injured from control mice with no overlap between groups (Figure 1). Axonal injury also correlated well across injury severities at 24 hours post-injury (Figure 2). Interestingly, with milder impact, there were DTI signal abnormalities detected, but no light microscopic immunohistochemical evidence of axonal injury.

Discussion: These results indicate that DTI may be capable of detecting axonal injury following TBI in an experimental animal model at multiple time points and injury severities. Remarkably, DTI changes strongly predicted the approximate time since injury. The DTI signal characteristics of white matter injury appear to evolve over time and the signature of abnormalities (axial diffusivity vs. radial diffusivity) may indicate the time interval that has elapsed between injury and imaging. If true in humans, this could have strong clinical potential as well as forensic implications. The underlying anatomical basis of the signal changes seen in milder injuries remain to be elucidated.

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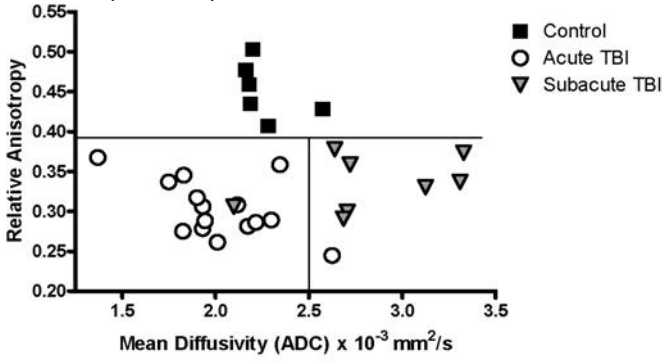

Figure 1 Discriminative Value of DTI. (A) Scatterplot of relative anisotropy vs. mean diffusivity for control, acute (0-4 days), and subacute (1 week – 1 month) time points. There was no overlap between the control and injured and there was little overlap between the acute and subacute injury groups. (Mac Donald et al J Neurosci 2007)

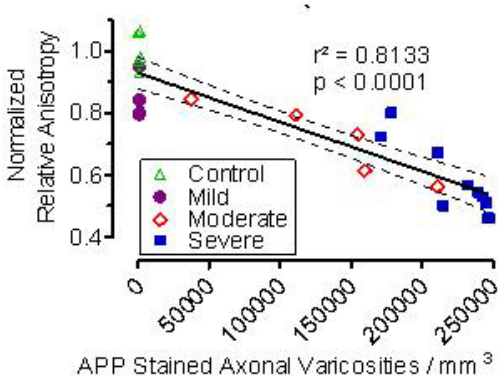

Figure 2 Quantitative Correlations Between Relative Anisotropy and Axonal Injury. Severity of axonal injury at 24 hrs correlates well with changes in relative anisotropy for moderate and severe injuries. *This correlation was lost in the mild injuries, as RA was abnormal, but no APP-stained axons were detected.* Dashed lines indicate 95% confidence interval.

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