Evaluating Secondary Degeneration After Spinal Cord Injury

Faith H. Reece[1], Nyoman D. Kurniawan[2], Gary J. Cowin[2], and Marc J. Ruitenberg[1,3]

[1]*School of Biomedical Sciences, The University of Queensland, Brisbane, Queensland, Australia,* [2]*Centre for Advanced Imaging, The University of Queensland, Brisbane, Queensland, Australia,* [3]*The Queensland Brain Institute, The University of Queensland, Brisbane, Queensland, Australia*

Background

Traumatic spinal cord injury (SCI) is characterised by immediate and severe loss of neural tissue integrity at the site of impact and consequential functional impairments.[1] A major complication following SCI is secondary degeneration, a phenomenon produced by cellular and molecular events (e.g. ischemia, excitotoxicity and inflammation) invoked by the primary trauma which worsens the primary damage by compromising nearby neurons that were originally spared.[1] As secondary degeneration is amenable to pharmacological intervention, this study utilised diffusion tensor imaging (DTI) to evaluate the chronology of secondary injury development in a mouse model of SCI to establish the therapeutic window for treatment.

Methods

3 C57Bl6/J mice underwent T10 laminectomy and moderate (70kdyn) contusive SCI using the Infinite Horizons impactor device.[2] Live imaging was performed prior to injury, then at 2 hours, 1 day, 3 days, 7 days and 30 days post-injury (DPI). Mice were imaged using a 16.4T Bruker NMR scanner with an 89 mm vertical bore magnet, 25 mT/m/A gradient coil set and Paravision 5.0 software. Animals were anaesthetised using an isoflurane/oxygen mixture (maintained at 0.5-1.5 %) and the MRI data acquired using a transmit/receive linear surface coil (1.5 x 3 cm). The core temperature of the mouse was maintained at 30°C and the respiration rate was monitored using Biotrig. To minimise movement artefacts, the data was acquired with respiratory gating.

DTI data was acquired axially using a DTI spin-echo sequence, in an interleaved fashion using TR/TE = 2400/21 ms, and acquisition matrix = 128 x 170 over the field of view 9 x 12 mm^2 (final resolution 70 µm x 70 µm). Slice thickness was 0.5 mm and the number of excitations = 1. Diffusion sensitising gradients were applied in 12 non-colinear, uniformly distributed directions with the gradient strength b = 1500 s/mm^2, 2 ms diffusion encoding and 20 ms diffusion separation. The DTI acquisition time was 3 hours, with a zero fill acceleration factor = 2. Ten axial images covering thoracic and lumbar segments were acquired as described previously.[3] The diffusion tensor parametric images were computed using the Paravision 5.0 diffusion tensor calculation routine; and were used for subsequent analyses of spared white matter (SWM), ventral funiculi (VF), lateral funiculi (LF) and dorsal columns (DC).

Results

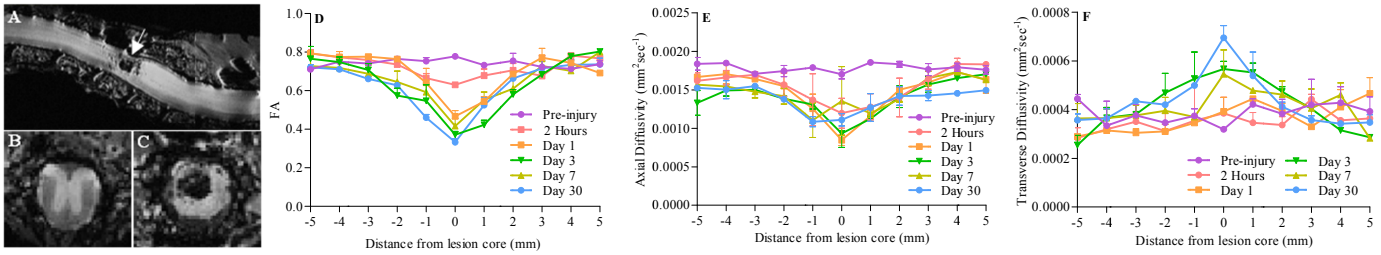

Figure 1: A-C: Representative DTI images of injured mouse spinal cord at 30 DPI in fractional anisotropy (FA) view. A: sagittal view showing disruption of tissue anatomy at the lesion site (arrow). B-C: coronal view of uninjured (B) and injured (C) segments of the cord. Analysis of the SWM determined that the FA (D) and axial diffusivity (E) decrease whereas transverse diffusivity (F) increases with proximity to the lesion core.

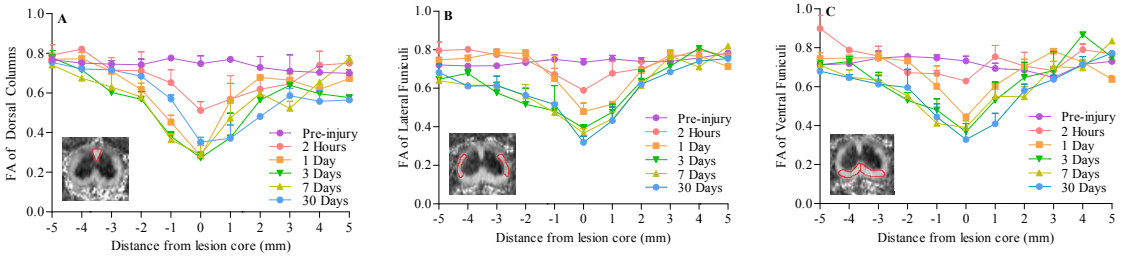

Figure 2: FA values in the DC (A), LF (B) and VF (C) with time. Tissue integrity is quickly lost in the DC, whereas the VF and LF show progressive de-generation with time (regions of interest shown in red) (Mean+SEM).

Conclusions

Analysis of SWM revealed that most of the tissue integrity is lost between 2hrs-1DPI. Regional analysis revealed that tissue damage progressively spreads to the LF and VF over time, whereas at the site of direct impact (DC), the neuronal architecture is rapidly destroyed. The initial impact produced damage 2mm along the cord, which had spread to 6mm by 30DPI. Therapeutic intervention designed to counteract secondary degeneration would be most advantageous if given 2 hours-1DPI and preserve spinal tracts involved in locomotion and coordination.

References

1.Thuret S, Moon LD, Gage FH (2006) Therapeutic interventions after spinal cord injury *Nat Rev Neurosci* **7** 628-43.

2.Scheff SW, Rabchevsky AG, Fugaccia I, Main JA, Lumpp Jr JE (2003) Experimental modelling of spinal cord injury: characterisation of a force-defined injury device *J Neurotrauma* **20**:179-193.

3.Clare K. Underwood CK, Kurniawan ND, Butler TJ, Cowin GJ, Wallace RH (2011) Non-invasive diffusion tensor imaging detects white matter degeneration in the spinal cord of a mouse model of amyotrophic lateral sclerosis *NeuroImage* **55**:455-461.