Transient Hypoxia in a Model of Distraction Spinal Cord Injury Results in a Reduction of Ventral Motor Neuron Size

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Introduction: Scoliosis corrective surgery can impart significant distraction forces to the spinal cord which, in extreme cases, can cause a primary mechanical and/or vascular injury to the cord resulting in new neurological deficits. The complex nature of the injury includes not only the primary insult but subsequent secondary signaling cascades that can remain undetected until after completion of the surgery. Distraction spinal cord injury remains an understudied injury paradigm, as the majority of animal models utilize transection or contusion injuries. Here we present the development of a relevant animal model that serves to separate the vascular component of the injury from the mechanical and aids in the understanding of the underlying mechanisms of this type of insult. This model will serve as a platform on which to screen potential neuroprotective therapeutics.

Materials and Methods: Previously, an animal model of distraction was established using the UTA Spinal Distractor in order to replicate the stretching force experienced during corrective surgery. Electrophysiological monitoring and behavioral and histological analysis showed a graded acute injury response due to 3-mm, 5-mm, and 7-mm distraction. Recent modifications to the 5-mm distraction injury have led to a more clinically relevant model. In the current study a 5-mm distraction injury was induced in female Long Evans rats at a rate of 0.5 mm/sec and then stretch was maintained for 15 minutes (n=60). The slower distraction rate and prolonged stretch decreased the initial observed variability. The current characterization focuses on evaluating the sub-acute secondary mechanism injuries. To determine if distraction induces a hypoxic event, the partial pressure of oxygen (pO_2) was measured using a Licox CC1.R(Integra-LS) oxygen electrode. Protein carbonyl content, an indicator of protein oxidation, was measured biochemically and immunohistochemistry was used to visualize tissue loss, cellular injury response (ED1 and GFAP), and to determine changes in ventral motor neuron (VMN) perikaryal size.

Results and Discussion: The induced distraction resulted in a mild injury as indicated by intact electrocompetency of the spinal cord, no observable loss of tissue and mild functional deficits with a BBB score of 18 in all animals. An induced hypoxic event was clearly noted after injury. Specifically, there was an average percent pO_2 decrease of 47.08± 5.79% following prolonged distraction whereas a value of only 1.61 ± 0.54 % was seen following a sham injury (Fig 1A). Furthermore, the hypoxic event was shown to induce mitochondrial dysfunction with an increase of 216% over the control in protein oxidation 30 minutes post-injury. Additionally, although there was no reduction in the number of neurons in the dorsal horns at the epicenter, there was a significant decrease (37%) in VNM perikaryal size.

Conclusions: The presented distraction model has provided a clinically relevant injury which indicates hypoxic insult leading to mitochondrial dysfunction as a possible mechanism in distraction SCI. This insult was sufficient to cause changes in the VMN soma area, leading to an observed functional deficit.

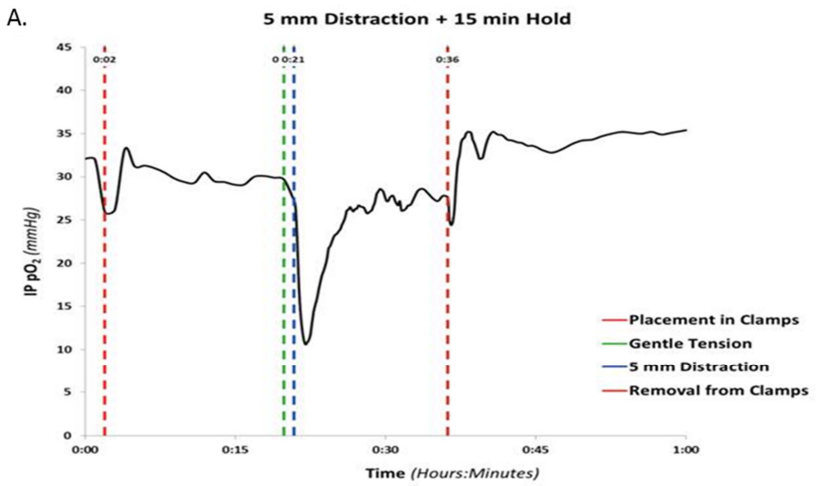

Figure 1: Oxygen tension in the spinal cord drops immediately after distraction and partially recovers within minutes.