Author

Ryan Anderson's enthusiasm for sports and resulting interest in orthopedics led him to his joining Dr. Gupta's Peripheral Nerve Lab at UCI. Working on creating a spinal cord injury model, he employed a novel surgical procedure to test the behavioral and molecular outcomes following a contusive-type spinal cord injury. For his senior year, he moved on to a new project, which included taking care of a breeding colony, conducting electrophysiology, immunohistochemistry, electromicroscopy, and counting G-ratios. Ryan considers his research and volunteer work in an orthopedic clinic to be integral elements of his preparation for an eventual career as a practicing physician.

Key Terms

- Cervical Spine
- Duraplasty
- Spinal Cord Injury (SCI)

The Role of Early Surgical Decompression of the Intra-Dural Space Following Cervical Spinal Cord Injury in an Animal Model

Ryan L. Anderson
Biomedical Engineering

Abstract

The role of decompressing the intra-dural space through a durotomy as a treatment option for acute traumatic spinal cord injury (SCI) in the cervical spinal cord has not been explored in an animal model. We sought to determine the role of durotomy and duraplasty in acute cervical SCI and its effects on inflammation, scar formation and functional recovery. Seventy-two adult female Sprague-Dawley rats received: contusion injury alone, contusion injury with durotomy and decompression, or contusion injury with durotomy followed by placement of a dural allograft. Those animals receiving a dural allograft had significantly improved scores in the recovery period relative to other groups. Additionally, immunohistochemical analysis revealed decreased scar formation, cavitation, and inflammatory response in those animals receiving a dural allograft relative to other groups. Lesional volume measurements showed significantly increased cavitation size at four weeks in both the contusion only and durotomy only groups relative to those animals that received a dural allograft following durotomy. Surgical decompression of the intra-dural space after an acute traumatic cervical SCI may be an important new approach to reducing the deficits resulting from the secondary injury and warrants further investigation.

Faculty Mentor

Our lab focuses on therapeutic neuroscience for clinical conditions from carpal tunnel syndrome to brachial plexus injuries to spinal cord injury. From direct clinical observations in the operating room we develop working hypotheses to improve our understanding and treatment of traumatic neurologic conditions. In this project, we explored the idea of adding a surgical intervention to improve functional outcomes after an acute spinal cord injury. Ryan played an integral role in the project as he facilitated the experiments from animal model creation to functional outcomes assessment to imaging. Through a team approach with surgeons, scientists, and students, we are able to contribute in a meaningful way.

Ranjan Gupta
School of Medicine

Introduction

Clinical outcomes following spinal cord injury (SCI) in the cervical spinal cord have historically been poor. Although the initial mechanical trauma inevitably results in some degree of necrosis, this primary injury is not the major contributor to cell death in acute SCI. The secondary injury that follows results from a complex series of pathophysiological changes that ultimately lead to scar formation and glial-encapsulated cystic cavitation (Blight, 1994; Wagner, et al., 1977). While it remains poorly understood, a number of processes have been implicated in the development of the secondary injury including edema, ischemia, alterations in cerebrospinal fluid (CSF) flow hydrodynamics, and macrophage accumulation (Balentine, 1998; Blight, 1991; Blight, 1994; Ducker et al., 1971; Fitch, 1999; Fitch and Silver, 1997; Wallace et al., 1987; Williams et al., 1981). Multiple novel treatment strategies to reduce effects of the secondary injury have been developed in recent years and continue to remain at the forefront of investigation (Dinh et al., 2007; Rasouli et al., 2006; Rasouli et al., 2008). Some pharmacological therapies have shown modest therapeutic benefits in clinical trial while a number of other strategies, including stem cell transplantation, show considerable potential in animal models (Baptiste and Fehlings, 2006).

Although there have been multiple basic science and clinical advances, there remains no single efficacious treatment regimen to prevent the devastating paralysis of traumatic SCI. Over the past twenty years, the survival rate and long-term outcome of SCI patients have improved with advances in both the medical and surgical management of these patients. Decompression of the spinal cord and maintenance of adequate vascular perfusion to ensure physiologic spinal cord blood flow remain two very important strategies that are believed to have significant benefit on neurologic outcome (Tator and Fehlings, 1991; Fehlings and Perrin, 2006).

While most studies have focused on the decompression of extradural elements, few have examined the potential deleterious secondary events that occur beneath the dura as a result of primary contusive trauma. Following contusive SCI initiated by displaced bony and soft tissue elements, the ensuing edema and hemorrhage within the spinal cord and nerve roots may result in expanding volume and increased intradural pressures against a relatively noncompliant dura (Maikos et al., 2008). It has been proposed that increased swelling alters normal cerebrospinal fluid pressure gradients favoring extravasation of fluid into the extracellular parenchyma of the spinal cord. The end result is decreased spinal cord perfusion pressure and ischemia. This ischemia may augment further secondary injury.

Surgical decompression of the dura in patients with acute SCI has been explored clinically in the past with impressive but guarded results (Perkins and Deane, 1988). To our knowledge, however, no animal studies have examined the histologic and functional response to decompression of intradural elements following contusion injury. Currently, there is no standard treatment algorithm to decompress the subarachnoid space following spinal cord injury. We sought to determine the role of surgical decompression with either durotomy or duraplasty in acute cervical SCI and to evaluate its effects on inflammation, scar formation, and functional recovery.

Methods

Animals

Adult female Sprague-Dawley rats (Charles River Laboratories, Wilmington, Massachusetts), weighing 220–240 g were used for all surgical procedures involving production of a spinal cord injury. The Institutional Review Board of the University of California, Irvine, approved all surgical protocols. Seventy-two adult female Sprague-Dawley rats were randomly assigned to three groups: contusion injury alone (n=24), contusion injury with durotomy and decompression alone (n=24), and contusion injury with durotomy and decompression followed by placement of a dural allograft (n=24). For surgery, the animals were anesthetized with an intraperitoneal injection of a mixture of 50 mg/kg of Ketamine and 2.6 mg/kg of Xylazine. Corneal reflex and withdrawal from painful stimulus of the hindlimbs were used to ensure adequate anesthesia. Heating pads were used to maintain appropriate body temperature at 37 °C throughout the perioperative period. Animals were housed in warmed cages and provided easy access to food and water. They were monitored at 6–8 hour intervals for the first four days postoperatively. During this period they required frequent hand feeds and manual bladder expression. Postoperative prophylactic antibiotic therapy with enrofloxacin (2.5 mg/kg subcutaneously), fluid resuscitation with lactated Ringer solution (5 mg/100 kg subcutaneously), and analgesia with buprenorphine (0.01 mg/kg subcutaneously) were administered for the first three days postoperatively and then as needed thereafter.

Production of Spinal cord Injury

A moderate contusive injury was delivered to the cervical spinal cord in 72 adult female Sprague-Dawley rats. Prior to surgery the animals were prepared and anesthetized as

above. A dorsal midline incision was made and the paravertebral musculature was separated from the vertebra at the C4–C6 level. A laminectomy was performed under microscope magnification at C5 using fine scissors and rongeur with careful attention not to disrupt the underlying dura. Once the spinal cord was exposed, the spinal column was rigidly immobilized to a stereotactic frame both cranially and caudally to the exposed segment. A 200-kilodyne bilateral injury was delivered to the center of the exposed spinal cord using a commercially available force based impactor (Infinite Horizon Impactor; Precision Systems and Instrumentation, Lexington, Kentucky). (Figure 1) This device is equipped with a 2.5-mm stainless-steel impactor tip attached to a calibrated load cell and is designed to deliver consistent force (Scheff et al., 2003). Following the injury, animals were carefully released from the stereotactic frame and a layered closure of the muscle and connective tissue was performed using absorbable sutures (4-0 chromic gut). Staples were used to close the skin.

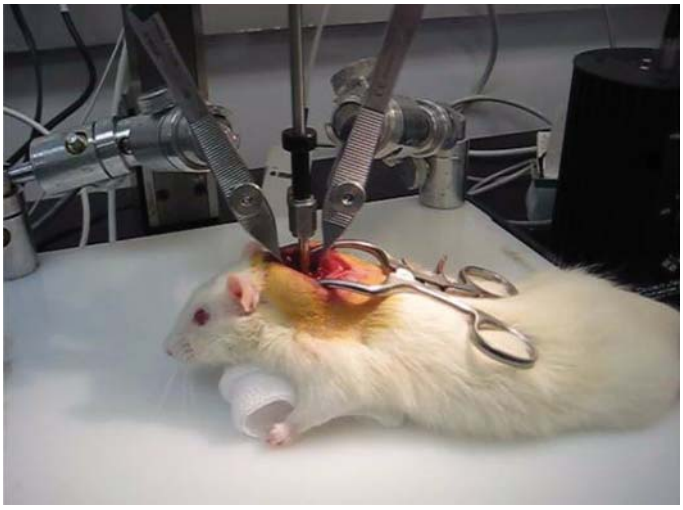

Figure 1

Animals were immobilized in a stereotactic frame with the C5 spinal cord segment exposed. A contusion injury was delivered using the Infinite Horizon Impactor.

Durotomy and Decompression

Animals receiving durotomy with decompression (n=48) were reanesthetized four hours following the initial injury using the techniques described above. After aseptic preparation, the spinal cord segment was re-exposed using the previous incisional wound. With the aid of an operating microscope, the segment was carefully cleaned of any soft tissue debris. A sterile 30-gauge needle was used to puncture a small hole in the dura/arachnoid matter at the center of lesion. A durotomy with a diameter of approximately 4 mm was created using microsurgical instruments. The defect extended circumferentially approximately 1 mm beyond the visibly injured segment. The exposed intradural spinal cord was examined and any residual hemorrhage and hematoma was carefully evacuated. Animals randomized to receiving only a durotomy (n=24) were closed using techniques described above. Animals receiving dural allograft were prepared for transplantation.

Dural Allograft Harvest

Adult female Sprague-Dawley rats (Charles River Laboratories, Wilmington, Massachusetts) weighing 280–300g were euthanized via intraperitoneal injection with pentobarbital (Nembutal; 100mg/kg) and exanguinated by transcardial perfusion with 100 mL of physiological saline (0.9%). Using sterile technique, the entire spinal cord (C1 to S5) was dissected and placed into a petri dish containing Hank's Balanced Salt Solution (HBSS). Large sections of dura mater were carefully dissected from the spinal cord and placed in the dish with careful attention to maintaining proper surface orientation prior to grafting. The cadaveric dura mater was stored in HBSS at 37 °C overnight prior to grafting.

Transplantation of Dural Allograft

Following decompressive durotomy as described above, twenty-four animals received dural allograft transplantation. The remaining intact dura surrounding the durotomy site was freed from any residual soft and bony tissue to ensure adequate area to overlay and fix the graft (~1–2 mm). With careful attention to maintaining the surface orientation of the cadavaric dura, the allograft was overlaid onto the edges of the durotomy ensuring contact between the graft

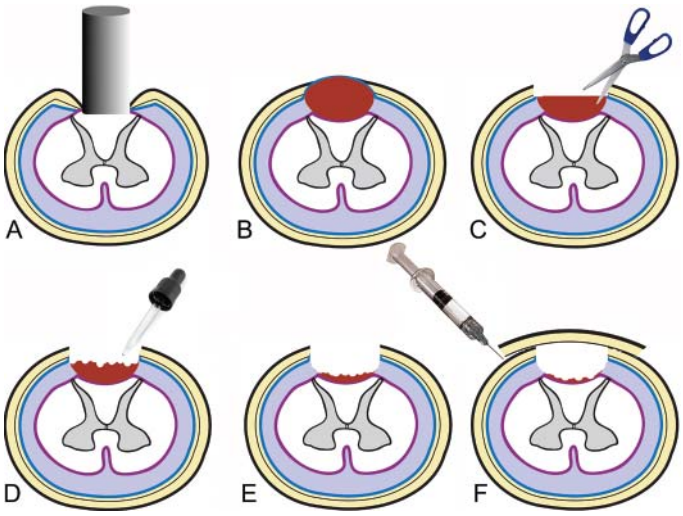

Figure 2

Schematic representation of surgical technique: A) Spinal cord impaction, B) Subdural edema and hematoma formation, C) Durotomy, D) Evacuation of hematoma and expansion of intradural space, E) Decompressed subarachnoid space, and F) Duraplasty

and host dura. Some redundancy was maintained at the center of the graft to allow for expansion of the intradural structures. Fibrin sealant (Tisseel; Baxter Healthcare Corp., Hyland Immuno, Glendale, CA) containing concentrated human fibrinogen (75–115 mg/mL), thrombin, aprotinin (a fibrinolysis inhibitor), and calcium chloride was used to affix the graft. The wounds were closed according to the technique described previously and postoperative care was initiated. (Figure 2)

Behavioral Assessment: Grip Strength Meter

The Grip Strength Meter (GSM—designed by TSE-Systems and distributed by SciPro, Inc.) was used to assess forelimb function according to previously described protocol (Anderson et al., 2007). (Figure 3) Each forepaw grip strength was measured five times per session. GSM testing was performed for a total of five sessions prior to SCI and was carried out every other day following surgery until sacrifice. In order to enhance reliability, a single experimenter obtained measurements from all animals. Prior to obtaining forelimb strength measurements, animals were handled for a minimum of five days to acclimate them to the experimenter. Following acclimation, the animals underwent seven training sessions. Animals were held at their midsection, facing the handle bar of the GSM. With one forearm manually restrained by the experimenter, the unrestrained forepaw was brought into contact with the handle bar, at which time the animals grasped the bar. The animal was then gently pulled away from the device. The GSM measured the maximum force before the animal released the bar.

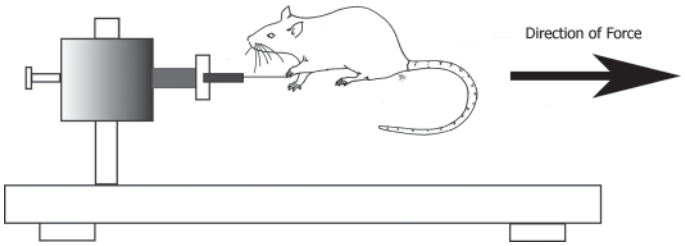

Figure 3

Representative schematic of Grip Strength Meter used to assess the functional recovery of animals post injury.

Tissue Preparation

Animals were euthanized at 14 and 28 days following SCI with pentobarbital (Nembutal; 100mg/kg) and perfused transcarially with cold saline (0.9%) followed by cold 4% paraformaldehyde (PFA). The spinal cords were carefully dissected and post-fixed in 4% PFA at 4 °C overnight. The tissues were cryoprotected in sucrose gradient of 10% for one hour, 20% for one hour, and then 30% overnight at 4 °C. The spinal cords were then submerged in TissueTek (VWR International, West Chester, PA) and frozen over dry ice. The cryostat was used to cut 20 μm sections spanning beyond visibly injured level. Sections were mounted onto polylysine-treated slides and stored at -80 °C until ready for staining.

Immunohistochemistry

In preparation for staining, the sections were thawed at room temperature for 20 minutes to promote section adherence to the slides. Frozen sections were first fixed in 4% PFA for 20 minutes. Next, sections were rinsed with 1x PBS for 5 minutes, followed by immersion in 0.25% Triton X-100 in 1x phosphate buffered solution (PBS) for 20 minutes. Nonspecific binding was blocked with 5% milk in 1x PBS for one hour. Sections were incubated in monoclonal mouse anti-rat ED-1 antibody (1:300; Serotec, Inc., Raleigh, NC), monoclonal rat anti-mouse OX-42 antibody (1:400; Serotec, Inc., Raleigh, NC), monoclonal mouse anti-glial fibrillary acidic protein (GFAP; 1:400; Sigma, St. Louis, MO), and monoclonal mouse anti-chondroitin sulfate proteoglycan (CSPG; 1:300; Sigma, St. Louis, MO) overnight at 4 °C. On the following day, the sections were washed three times in 1x PBS for 5 minutes and subsequently reacted with FITC-conjugated goat anti-mouse IgG secondary antibodies (1:200) for one hour. Next, sections were washed three times in 1x PBS for 5 minutes, mounted with medium containing DAPI (4',6-diamidino-2-phenylindole), visualized under an Olympus DP7x fluorescence microscope and photographed with a Hamamatsu DCAM camera. The primary antibodies used to assess scar formation and inflammatory infiltration stained for the following: ED-1—systemic macrophages, OX-42—activated macrophages and microglia, GFAP—astrocytes, and CSPG—potent inhibitors of CNS regeneration.

Lesional Volume

Tissue samples intended for lesion volume analysis were prepared using the hematoxylin and eosin stain (H & E). Sections were stained serially. Lesion volumes were then calculated using ImageJ 1.41, an image processing and analysis program developed by the National Institutes of Health (NIH). The experimenter was blind to the treatment received by each sample. The lesion areas were outlined in three separate trials using the freehand technique. Lesion areas for each section were averaged over the three trials and multiplied by the section length to obtain the lesion volume.

Statistical Analysis

GSM: For each time point, the average force exerted by the left and right forepaw was determined from the mea-

surements gathered. For each group, a repeated measures two-way analysis of variance (ANOVA) was conducted to identify differences between groups and time points in the post-injury period. The Bonferroni test was used for post-hoc analysis to correct for multiple comparisons.

Lesional Volume: The significance of the differences in lesional volume size between treatment groups was initially determined using one-way ANOVA. The unpaired t-test was used to determine the significance between treatment groups at both time-points. Differences were considered significant if $p<0.05$. Analysis and histograms were created using GraphPad Prism (version 4.0; GraphPad Software, Inc., San Diego, CA).

Results

Six of 72 animals died within the first day of injury giving an overall survival rate of 91.7%. Five other animals were removed from the study due to forepaw injuries they sustained in the post-injury period.

Gross Examination

Gross examination of the spinal cords following re-exposure four hours following impaction showed increased swelling and hematoma formation beneath the dura. (Figure 4) In addition, animals showed evidence of congestion within the dorsal vasculature. With subsequent durotomy and decompression, these vessels appeared to become re-perfused. In addition, the spinal cord appeared to herniate from the underlying intradural space as if swelling had overcome the space available for the cord contained within the dura mater.

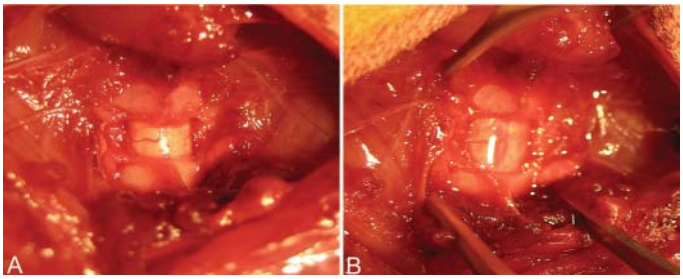

Figure 4

Exposed spinal cord at C5: (A) Preinjury, and (B) Four hours following injury.

Gross examination of the spinal cord following harvest showed more significant scarring and cavitation in the animals receiving durotomy only relative to those animals that received durotomy followed by placement of dural allograft. (Figure 5) Animals receiving contusion injury alone displayed similar gross characteristics to those animals that received dural transplantation.

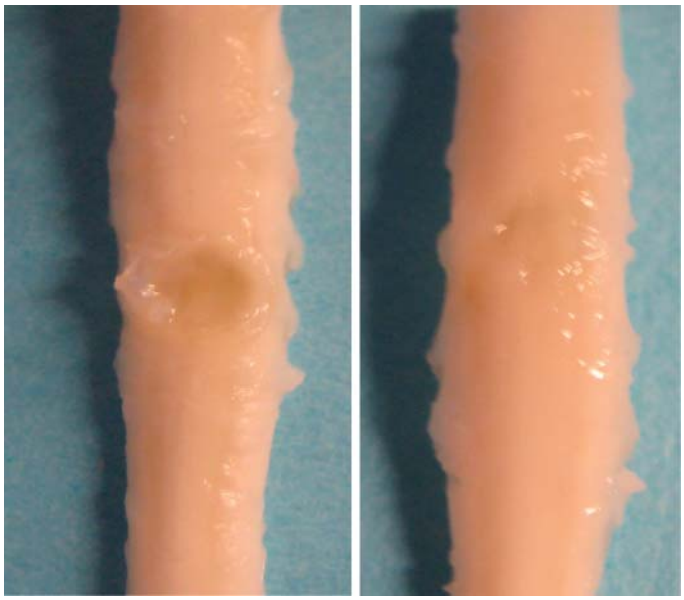

Figure 5

Gross spinal cord specimens four weeks after sacrifice. Animals receiving durotomy alone (A) displayed more gross scarring and dorsal cavitation relative to animals receiving dural allograft (B).

Histology

Immunohistochemical analysis was performed to assess inflammatory cell infiltration along with scar formation. (Figures 6 and 7) Antibodies to assess inflammation included ED-1 (marker for macrophages) and OX-42 (marker for macrophages and activated microglia). Glial scar formation was assessed with antibodies to GFAP (marker for astrocytes) and to chondroitin sulfate proteoglycan (CSPG). Morphometric analysis revealed consistently increased cellular infiltration around the lesion site in animals receiving durotomy without allograft transplantation on both ED-1 and OX-42 stained sections. (Figure 8) This was consistent between the 2- and 4-week time points. In groups receiving contusion only and in those receiving durotomy followed by placement of a dural allograft, there was increased cellular density in the areas surrounding the lesion although it was not as marked as that seen in animals receiving durotomy alone. These differences were not quantitatively verified between the two groups. Antibodies staining for glial scar formation (CSPG, GFAP) showed similar increases in astrocyte proliferation and fibrosis surrounding the cavitation and extending more diffusely throughout the peri-lesional areas in those animals receiving only a durotomy. This scarring became more pronounced at the 4-week relative to the 2-week interval timepoint. Again, slight differences in immunoreactivity for glial cell markers were observed

between the contusion alone and dural allograft groups with slightly less scar formation observed in the latter. Histologic examination of the transplanted dura showed a well-bridged continuity between host and donor dura with minimal amount of fibrotic scar tissue.

Lesional volume measurements showed significantly decreased cavitation size at four weeks in the group that received decompression followed by placement of a dural allograft (6.8±1.4mm3) relative to both the contusion only (12.6±0.5mm3) and durotomy only groups (15.1±1mm3) ($p<0.05$). There was no significant difference in cavitation size between the contusion only (9.9±0.5mm3) and dural allograft transplantation (6.8±1.5mm3) groups at 2 weeks. At both the 2- and 4-week intervals, animals receiving durotomy alone showed significantly increased cavitation size (12.39±0.8mm3) relative to other groups ($p<0.05$). (Figure 9)

Functional Recovery

The GSM has been shown to provide sensitive, quantifiable and reproducible measurements of forepaw strength following cervical SCI in the rat (Anderson et al., 2004; Anderson et al., 2005; Anderson et al., 2007). Pathways involved in the grip strength task and their role in recovery are currently being elucidated (Anderson et al., 2005).

Pre-injury GSM measurements were taken at five time-points at least 10 days prior to surgery. No significant grip strength differences were found between sides or groups in the pre-injury period. Following SCI, animals receiving dural allograft transplantation showed improved GSM scores at timepoints after post injury day 10 relative to animals receiving contusion injury alone. These values were significant at post injury day 14 (92.4g vs. 63.4g), 16 (102.2g vs. 75.4g), 18 (115.2g vs. 83.2g), and 22 (129.0g vs. 77.2g) ($p<0.05$, 95% CI). Of note, the grip strength values measured at the final timepoint (day 24) displayed no significant difference ($p>0.05$) with an apparent downward trend in functional recovery in the group

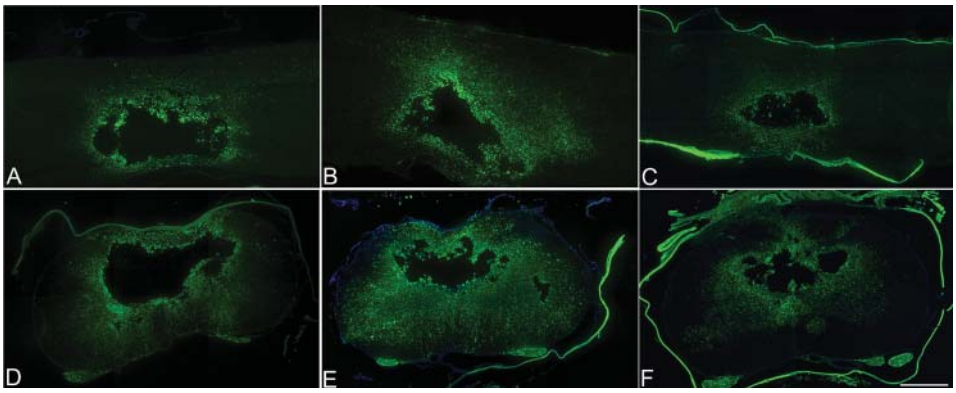

Figure 6

ED-1 immunohistochemistry 4x. At four weeks, animals receiving dural allograft following decompression (C) and (F) show decreased inflammatory response relative to the durotomy only (B) and (E) and contusion only groups (A) and (D). Animals receiving durotomy alone displayed the greatest inflammatory response. Scale bar = 500μm.

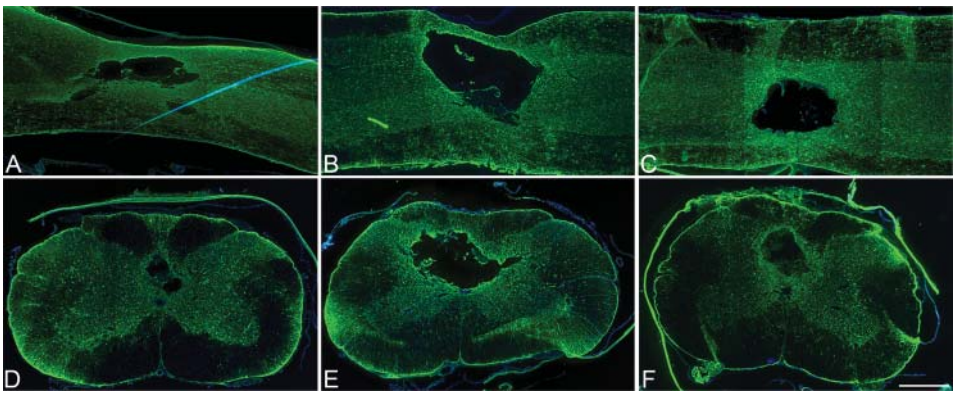

Figure 7

GFAP immunohistochemistry 4x. At four weeks, animals receiving decompressive durotomy alone (B) and (E) display the most significant astrocyte proliferation indicating greater scarring relative to animals receiving contusion only (A) and (D) and animals receiving dural allograft (C) and (F). Scale bar = 500μm.

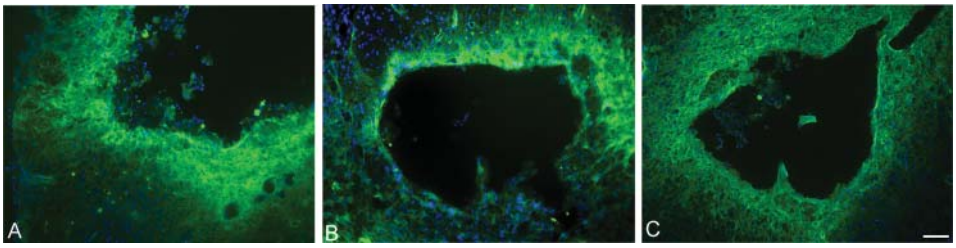

Figure 8

OX-42 immunohistochemistry 10X. Animals receiving durotomy followed by placement of a dural allograft (C) show the least amount of peri-lesional infiltration of macrophages and activated microglia relative to animals receiving contusion (A) or durotomy (B) alone. Scale bar = 50μm.

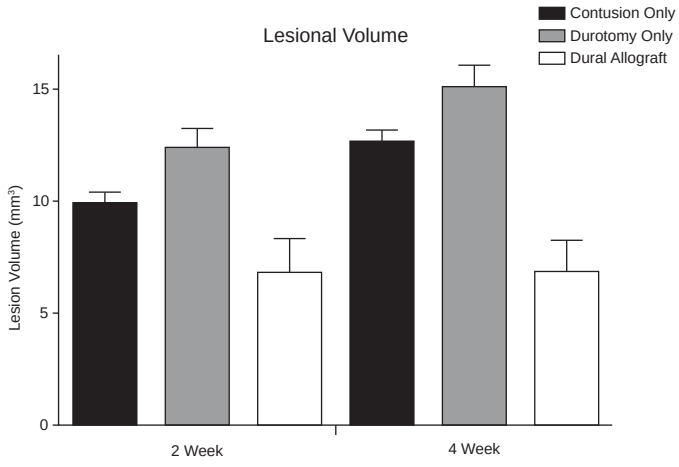

Figure 9

Lesional volume measurements show significantly increased ($p<0.05$) cavitation size at four weeks in both the contusion only and durotomy only groups relative to those animals that received decompression followed by placement of a dural allograft. At the 2-week interval, animals receiving durotomy alone show significantly increased cavitation size relative to the other treatment groups.

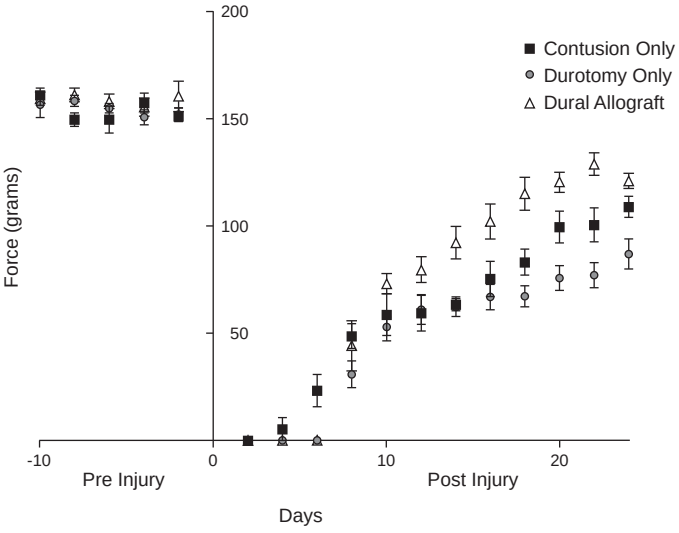

Figure 10

GSM: Animals receiving dural allograft transplantation show significantly improved GSM scores following injury relative to animals receiving contusion injury and durotomy alone ($p<0.05$). Animals receiving durotomy alone without dural allograft display significantly worse functional outcome relative to the other groups. No significant differences in grip strength were found in the preinjury period.

receiving dural allograft (121.2g) relative to contusion only animals (108.9g). Although grip strength measurements were not obtained at later time points, this may indicate a possible end to a trend in functional recovery.

Animals receiving a decompressive durotomy alone without dural allograft transplantation consistently displayed the worst functional outcome after 2 weeks post injury. Relative to the contusion only group, there was a trend of poorer grip strength recovery although this trend was statistically significant only at day 20 (99.7g vs. 75.8g) and day 22 (100.6g vs. 77.2g) ($p<0.05$, 95% CI). No animals achieved complete recovery to baseline levels. (Figure 10)

Discussion

Many strategies to reduce secondary injury in the face of acute SCI have been proposed that attempt to address the multiple pathways involved, including oxidative cell injury, glutamatergic excitotoxicity, apoptosis, ischemia, inflammation, and edema (Fehlings and Tighe, 2008). Historically, attempts to reduce these events have been approached primarily by pharmacologic means. Methylprednisolone, for example, has been used extensively although its efficacy is questioned because of the controversial interpretation of the NASCIS trials. Few surgical strategies have been proposed to address the intradural pathoanatomical changes—including edema, ischemia and alterations in the cerebral spinal flow—that contribute to the secondary injury process.

Decompression of the bony and soft tissue elements (extradural) in the setting of acute SCI has long been the focus of early surgical intervention. Acute versus delayed surgical decompression remains a topic of intense controversy due in part to equivocal outcomes in both treatment groups (Fehlings and Perrin, 2006; Levi et al., 1991). It is possible that traditional methods of decompression are not addressing intradural compressive changes that contribute to secondary spinal cord injury. In a report of six cases, Perkins et al. performed a durotomy decompression in the setting of acute spinal cord injury (Frankel A,B) (Perkins and Deane, 1998). In their series, they describe a distended and pulseless thecal sac following extradural decompression, which displayed evidence of vascular decongestion and reperfusion following durotomy. All patients showed neurologic improvement; three made complete neurological recovery.

Our data suggests that it is beneficial to decompress the intradural elements in order to reduce secondary injury. We found that decompression of the cervical spinal cord following a moderate contusion injury results in improved functional recovery and limits secondary inflammatory infiltration, glial scar formation, and cystic cavitation. This benefit, however, was only apparent in those animals that received transplanted dural allografts following durotomy. Based on the size limitations of a small animal model that we observed in our preliminary experiments, it was not pos-

sible to surgically reapproximate the dura consistently after animals received a decompressive durotomy alone. As such, this group displayed the worst functional and histologic outcomes. It should not be surprising that the benefit of functional and histologic recovery after a decompressive durotomy would require the continuity of an intact overlying dura with an expanded intradural space available for the spinal cord. To our knowledge no animal study exists that evaluates the role of decompressing the spinal cord by means of a durotomy.

Edema is a well-recognized cause of secondary SCI in animals and humans (Boldin et al., 2006; Flanders et al., 1999; Fujii et al., 1993; Kwon et al., 1994; McDonald and Sadowsky, 2002; Miyanji et al., 2007; Saadoun et al., 2008). It has been suggested that following SCI, the swollen spinal cord compresses against the dural elements resulting in increased intraparenchymal cord pressure (Saadoun et al., 2008). This increase in pressure can have a tamponade effect on the spinal vasculature, initially limiting venous outflow from the spinal cord. The resulting spinal cord edema may further increase the intraparenchymal pressure, perpetuating an already hostile ischemic environment. Although objective pressure measurements were not obtained in our study, it is logical that this environment existed in our model. In the animals that received durotomy with decompression, there was obvious spinal cord edema visible upon re-exposure four hours following injury. In addition, the spinal cord appeared to partially herniate through the durotomy indicating significant parenchymal edema.

The role of CSF and its contribution to secondary SCI has been explored. Particularly, alterations in CSF flow and pressure hydrodynamics and its contribution to ischemia and parenchymal edema are of great interest. External forces from the traumatic event may initiate the edematous process. The resulting spinal cord swelling and any sustained external pressure may block normal CSF flow. CSF continues to be produced, resulting in variable intradural pressure gradients favoring spinal cord interstitial edema (Klekamp et al., 2001). This rise in intradural pressure may further perpetuate the spinal cord edema because of altered pressure gradients and limitations in regulating fluid balance secondary to altered vascular flow.

Decreasing the intrathecal pressure by CSF drainage in thoracoabdominal aneurysm repair surgery is routinely performed to reduce the incidence of ischemia induced paraplegia (Coselli et al., 2002; Safi et al., 1996). These results prompted investigators to define the role of reducing intrathecal pressures in the setting of acute spinal cord injury in an attempt to optimize spinal cord perfusion pressures. Animal studies show potential tissue protective qualities following use of this technique although the effects on spinal cord perfusion and functional recovery are not yet clearly defined (Horn et al., 2005). Recently, a prospective randomized clinical trial was conducted to evaluate the role of CSF drainage during the acute phase of SCI (Kwon et al., 2009). In this study, patients were noted to have increases in intrathecal pressures caudal to the injured level following spinal decompression. This increase in intrathecal pressure corresponds to decreases in spinal perfusion pressure. Our model of decompression encompasses similar principles of reducing intrathecal pressures and restoring normal gradients across the spinal cord in order to improve spinal cord perfusion and limit secondary ischemia. We hypothesize that subsequent return of CSF flow hydrodynamics may limit propagation of edema and subsequent ischemia.

The role of maintaining integrity of the dura in SCI has been explored previously. Iannotti et al. studied the histologic response to laceration type SCI in a rodent model (Iannotti et al., 2006). In their study, they report decreased lesional macrophage accumulation, cystic cavitation, scarring and improved CSF flow in animals receiving dural allografts relative to animals left with a large dural defect. They hypothesized that maintaining the continuity of the dura following laceration injury maintained a more physiologic CSF fluid flow pattern and prevented extradural factors from inhibiting neuroregeneration and promoting inflammation. In a rodent clip compression SCI model, Fernandez et al. also reported the importance of maintaining dural continuity to prevent epidural and spinal cord fibroblast proliferation and scar formation (Fernandez and Pallini, 1985). The results we obtained from those animals that received durotomy parallel these results. It is likely that similar alterations in cerebral fluid flow, increases in proinflamatory cytokine/growth factors, increased inflammatory cell accumulation, and meningeal fibroblastic proliferation are contributing to increased scar formation and cystic cavitation in this subgroup of animals. These results coincided with a worsened functional recovery relative to animals that maintained dural integrity.

There are limitations to the study design. The current spinal cord injury protocol requires a decompressive laminectomy prior to producing the contusive spinal cord injury. This does not mimic a true clinical spinal cord injury scenario where a displaced spinal column may cause the primary injury and result in residual cord compression. In our model, fixation of the transplanted dural allograft was provided by a fibrin sealant. Its effects on the spinal cord and

the potential neuroprotective and anti-inflamatory properties need to be further investigated. The functional benefits observed in animals receiving duraplasty began at 14 days and continued through post injury day 22. This functional recovery appeared to trend downward as the study period ended. Further investigation is required to determine if these effects were sustained beyond the 4-week time-point.

The data demonstrates that acutely decompressing the spinal cord with duraplasty may limit the degree of secondary injury in a small animal model. Both functional and histologic evidence of neuroprotection are evident in those animals that received dural decompression yet maintained the integrity of the overlying dura. Although these results are compelling, translating them into larger animal models or a potential clinical treatment strategy will require further investigation.

Cervical SCI continues to result in devastating long term disability. Regaining even modest control of upper extremity function can have a profound effect on quality of life. Surgical decompression with duraplasty after an acute traumatic cervical SCI may be an important new approach to attenuating secondary injury and warrants further investigation.

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