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TITLE: Effects of Early Acute Care on Autonomic Outcomes in SCI: Bedside to Bench and Back

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14. ABSTRACT

Early management of blood pressure (BP) may be critical to outcome after spinal cord injury (SCI), but evidence-based protocols are needed. Optimal early treatment and management of SCI has not been established in clinical practice, nor in animal models. Guidelines for management of BP in acute SCI have been influenced by the rather clear evidence of a relationship between hypotension and poor outcomes in TBI, and the aim of maintaining cerebral blood flow in the face of increased intracranial pressure (ICP), but doubt remains about what is best for SCI. This grant focuses on the following two hypotheses: *1) Episodes of low BP (measured by mean arterial pressure (MAP) and systolic BP) in the early management of clinical SCI predict worse long-term functional outcomes, and 2)spontaneous hypotensive episodes in the perioperative period of experimental SCI in rats will result in worse outcomes.* Both clinical data and experimental modeling studies address these specific hypotheses.

15. SUBJECT TERMS

Nothing listed

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Progress Report – Year 3
Award Number: W81XWH-13-1-0297
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Project Title: Effects of Early Acute Care on Autonomic Outcomes in SCI: Bedside to Bench and Back
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Grants Officer's Representative : Patricia A. Henry, PhD

Accomplishments

1. UCSF Animal Protocol and ACURO Protocol approvals were received.
2. Human Subjects Protocol approval letters for the retrospective study were received from SFGH, Santa Clara Valley Medical Center and Palo Alto VA Health Sciences Center were approved.
3. Human subjects protocol approvals for the prospective study at UCSF have been obtained. During this year we have enrolled 35 patients.
4. Established 1) surgical methods for implanting Data Sciences blood pressure transducers in rats, and 2) the drug delivery techniques using phenylephrine, dopamine, and norepinephrine to determine infusion rate, method of delivery, concentration, etc. for holding blood pressure (BP) at specified levels for 4 hours after SCI. We have obtained data on rats sustaining 250 kilodyne impact at T3. BP, heart rate (HR), bladder function and locomotor function was assessed for 4-6 weeks after injury. This data is currently being analyzed and prepared for publication.
5. Held meetings with SFGH clinicians to work on analyzing data from SFGH patient records for the retrospective study. We were able to access the large existing database containing q 1min BP data from SFGH SCI patients from 2007-2013. This analysis showed that patients with more epochs of hypotension had poorer outcome. A manuscript describing this was published in Journal of Neurotrauma (Hawlryluk GWH, Whetstone W, Saigal R, Ferguson AR, Talbott JF, Bresnahan JC, Dhall SS, Pan J, Beattie MS, Manley GT (2015) Mean arterial blood pressures and duration of hypotension correlate with neurological recovery following human spinal cord injury: Analysis of high frequency physiologic data. J. Neurotrauma, 2015 Feb 10. PMID: 25669633. PMCID: PMC4677564.
6. Once the access to the SCI patient data was established, we were able to evaluate MRI records as well. In collaboration with Dr. Jason Talbott of

the Department of Radiology, a new MRI scoring system was established to use this information for prediction of injury severity in cervical injury. Talbott JF, Whetstone W, Ready W, Ferguson AR, Bresnahan JC, Saigal R, Hawlryluk GWH, Beattie MS, Mabray M, Pan J, Manley GT, Dhall SS. (2015) The Brain and Spinal Injury Center (BASIC) spinal cord injury (SCI) score: A novel, simple, and reproducible method for assessing severity of acute cervical SCI using axial T2 MRI. J.Neurosurgery (Spine), 2015, 23:495-504. PMID: 26161519

7. A similar analysis was performed evaluation thoracic injury. Mabray MC, Talbott JF, Whetstone WD, Dhall SS, Phillips DB, Pan JZ, Manley GT, Bresnahan JC, Beattie MS, Haefeli J, Ferguson AR. Multidimensional analysis of MRI predicts outcome in thoracic and thoracolumbar spinal cord injury. J Neurotrauma, 2015 Sep 28. PMID: 26414451. PMCID: PMC4876497.
8. Retrospective data analysis at SFGH has identified complications with vasopressor usage in central cord injuries. Readdy WJ, Whetstone W, , Ferguson AR, Talbott JF, Inoue T, Saigal R, Bresnahan JC, Beattie MS, Pan J, Manley GT, Dhall SS (2015) Complications and outcomes of vasopressor usage in acute traumatic central cord syndrome. J. Neurosurgery (Spine), 23: 574-580.
9. Work on parallel blood pressure evaluations and correlation with outcome using novel methods for data evaluation (topological data analysis) has been performed. Nielson, J, Paquette J, Liu AW, Guandique CF, Inoue T, Irvine KA, Gensel JG, Petrossian TC, Lum PY, Carlsson GE, Manley GT, Beattie MS, Bresnahan JC, Ferguson AR. Big-data visualization for translational neurotrauma. Topological data analysis for discovery in preclinical spinal cord injury and traumatic brain injury. Nature Communications, 2015, 6:8581.
10. Retrospective data analysis also showed that penetrating injuries have a different profile. Readdy WJ, Saigal R, Whetstone W, Ferguson AR, Talbott JF, Inoue T, Bresnahan JC, Beattie MS, Pan JZ, Manley GT, Dhall SS (2016) Failure of mean arterial pressure goals to improve outcomes following penetrating spinal cord injury. Neurosurgery, (05.03.2016).doi: 10.1227/NEU.0000000000001249.
11. A Redcap data acquisition system was developed for the prospective data collection at SFGH and is currently being used. This system has now also been transferred Drs. Creasey and McKenna to identify procedures for accessing data from Santa Clara Valley Medical Center and Palo Alto VA Health Sciences Center. Intraoperative record data from SFGH and SCVMC has undergone preliminary analysis and shows that even for the time that patients were undergoing spinal cord surgery, there is an effect of low blood pressure.

Major Task 1: Regulatory set up for animal and human studies (Specific aims 1-3)
Subtask 1:UCSF Retrospective study IRB approval

UCSF IRB and HRPO approvals for retrospective chart review for this project have been obtained.

Subtask 2: UCSF IACUC approval

The UCSF IACUC approval was obtained and then submitted to ACURO and approval was received Jan. 15, 2014.

Subtask 3: PAVAHCS and SCVMC retrospective study IRB approvals

The VA approval and the SCVMC approval were completed, the subcontracts for these organizations have been finalized and approved by the DoD.

Subtask 4: SFGH Prospective study: Consent forms, IRB approval

Protocol A-17989.2a, "Transforming Research and Clinical Knowledge in Spinal Cord Injury", is currently pending approval. The initial research protocol application and supportive documents were submitted and received by the DoD on August 4, 2015. The UCSF team received a request for revisions September 22, 2015 which included a request for local IRB application modifications. Completed DoD revisions were approved after IRB applications were approved by the UCSF Committee of Human Research (CHR). Biosketches for Principal Investigator/Associate Investigators and human subjects protection training certifications were also sent over at that time. An additional DoD request was sent to the UCSF team on October 21, 2015 regarding medical record release language in the Informed Consent Form. The UCSF team received CHR approval for the consent modification and DoD approval.

The continuing review for the retrospective study A-17989.1a, "Effects of Early Acute Care on Autonomic Outcomes in SCI: Bedside to Bench and Back" was submitted and approved.

Milestone(s) Achieved: Animal and retrospective studies approved

Specific Aim 1: Examine the available evidence for a correlation between early BP (and bladder/bowel) management, vasopressor use, and later outcomes, including outcomes on autonomic, bladder and bowel function. (year 1-2)

Major Task 2:

Retrospective review of paper and electronic medical records of SCI patients

We have held meetings with the SFGH spinal cord injury clinicians and have established methods for accessing a large dataset collected over the past several years from the ICU using the Aristein monitoring system which contains q 1min blood pressure data for all SCI patients during their ICU stay. Data have been loaded into the HIPA compliant database 'RedCap' for querying. Dr. Whetstone organized other data from most of these SCI patients treated at SFGH; this data has been matched to the data in the Aristein monitoring system by Dr. Gregory Hawryluk who was able to access the BP data using Matlab programs that he wrote. We have analyzed the q 1 min data on physiological monitoring of all SCI patients seen between 2005 and 2011 and compared it to the ASIA grade status over the ICU stay. The data show that patients with more epochs of low BP show worsening AIS motor scores (Fig. 1, from Hawlyruk et al.,2015). The advantage of these data is that it doesn't only show average MAPs but shows every instance of low blood was a significant relationship between the number of epochs of MAP below 80 and

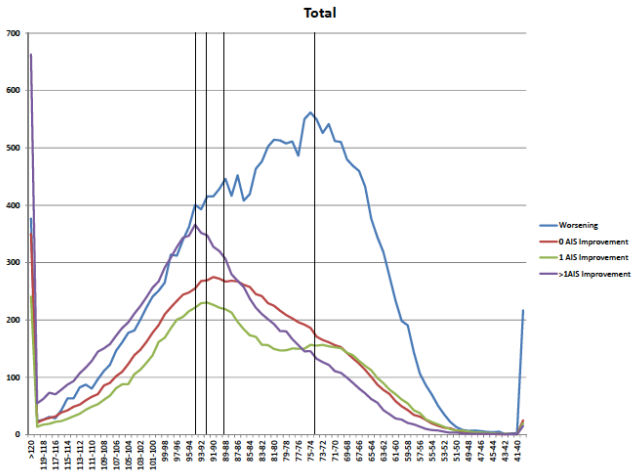


Fig. 1. Mean frequency of total MAP episodes (ordinate) for each BP range (abscissa) for groups of patients with worsening AIS scores (blue), no change in AIS scores (red), 1 AIS score improvement (green), or 2 AIS score improvement (purple). (from Hawlryluk et al, 2015).

poorer outcome pressure during the entire recording period from admission to discharge in the ICU. There at discharge from the ICU providing initial support for the hypothesis driving this grant.

Specific Aim 2: Provide detailed reports and physiological monitoring in the pre-hospital, ED and ICU to identify cardiovascular parameters and (events) during early management of SCI that are associated with poor outcome, including bowel and bladder function.

Major Task 4: Perform detailed physiological monitoring in the ED and ICU for 1st 7 days after SCI.

Subtask 1: Use prior SCI+TBI and early results of consortium BP record evaluations to finalize data collection strategy for prospective study.

We have currently set up a RedCap database and are using the data entry forms for the prospective study based on our retrospective analyses.

Subtask 2: Train and coordinate investigators and clinical staff in ED, ICU, and rehab. New members have been added to the SFGH/UCSF group to participate as clinical investigators including Sanjay Dhall, MD, Neurosurgery, Jason Talbott MD,PhD Radiology, Jonathan Pan MD,PhD, Anesthesiology, will join co-investigators William Whetstone MD from Emergency Medicine and Geoffrey Manley MD, PhD who are already on the project. This team will also be conducting a related but separate prospective study entitled "Canadian Multicentre CSF Monitoring and Biomarker Study" CAMPER. The Rick Hansen Institute has sent trainers to train and certify our clinical coordinators and nurse-practitioners to perform ASIA sensory and motor scoring. We are sharing clinical coordinators with the large prospective observational study TRACK-TBI which allows us, with limited funds, to cover 24/7 enrollment of SCI patients. The human subjects protocols have been approved by our IRB and the DoD.

Subtask 3: PAVAHCS and SCVMC retrospective study IRB approvals.

Subtask 4: SFGH Prospective study: Consent forms, IRB approval; Local IRB approval and DoD approval has been obtained. We currently have enrolled 36 patients. The patient accrual update is shown below in Table 1.

Table 1. Clinical Summary **N=36 unless otherwise noted**	**Mean/Count (Range)**
Demographics	
Total Patients Enrolled	36
Male	26
Age	49.6 (18-79)
Trauma Characteristics	
Level of Injury	
Cervical	20
Cervical-Thoracic	3
Cervical-Lumbar	1
Thoracic	6
Thoracic-Lumbar	3
Lumbar	2
Cervical-Thoracic-Lumbar-Sacrum	1
AIS at ED Admission	
A	8
B	5
C	2
D	6
Unable to Assess (sedated/altered mental state/etc)	16
Concurrent TBI Injury	6
Mechanism of Injury	
Fall	30
Transport	9
Assault	3
Crush Injury	1
Other	2
Blunt Injury	26
ISS on Arrival (N=19)	29.6 (10-75)
Central Cord	13
History of Hypertension	10
Hospital Stay	
Transport Time (N=25)	16.7 min (2-49)
Time in ED (N = 35)	229.74 min (48-782)
Time to OR (N=30)	10.9 hr (1.7-23.15)
ICU Length of Stay (N= 23)	9.6 Days (1.88-37.6)
Hospital Length of Stay (N= 23)	17.9 days (3.67-93)
Discharge (N= 33)	
Acute Rehab (N= 33)	21
Deceased (N= 33)	3
Nursing Home (N= 33)	2
Home/Private Residence (N= 33)	4
Group Living (N= 33)	1
Other Hospital (N= 33)	2
Follow Up	
# of 3Mo Phone Calls Completed	15
# of 6Mo In Person Visits Completed	14
# of 12Mo In Person Visits Completed	10

We are now analyzing the human retrospective data to determine if high blood

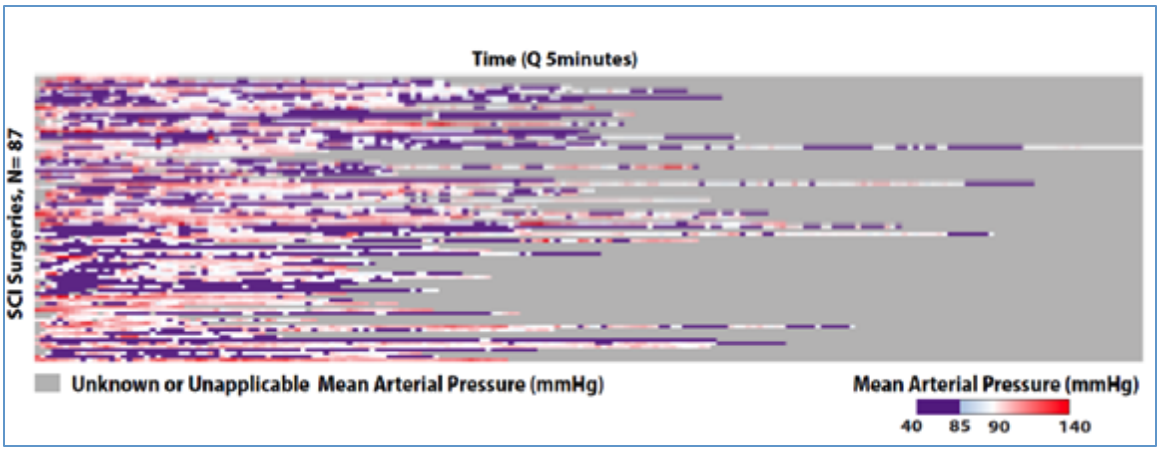


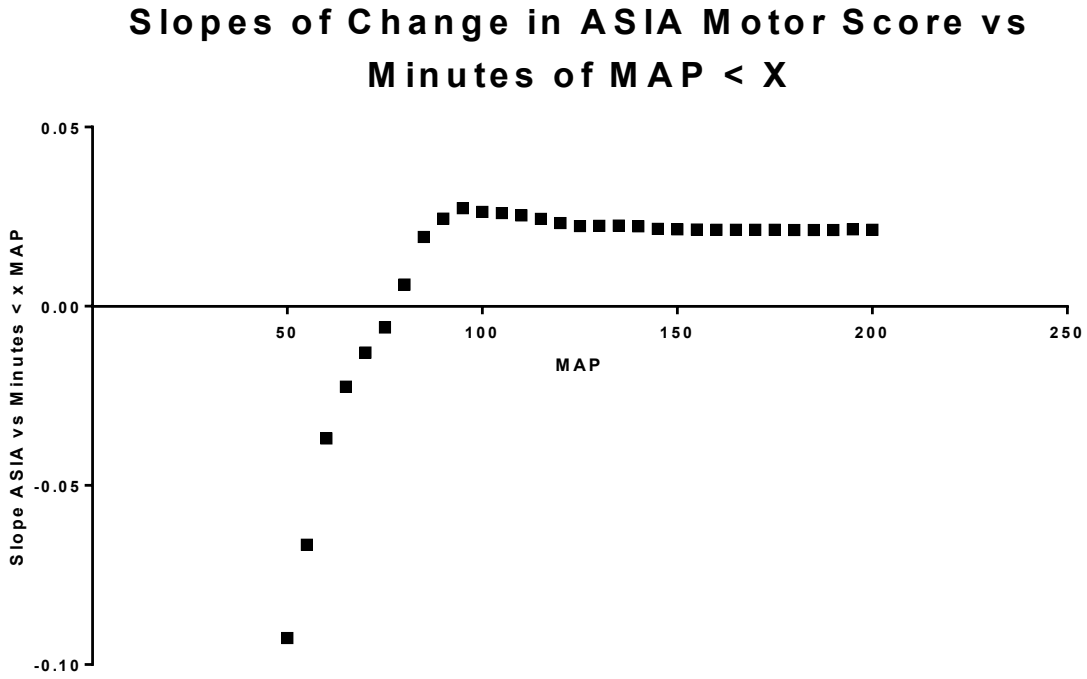
Figure 2. Plot of MAP over time in the OR during surgery. Each line represents an individual patient (n=87) with the colored area of each line representing the MAP (as shown in the indicator, low MAP=purple; high MAP=red), and the length of the line represents the surgery duration. . Both high and low MAPs are observed.

pressure could be identified as an issue in the SCI patients. At the current time, we have identified very high MAP in a subset of patients during surgery and a preliminary analysis performed by Dolores Torres, MD candidate, of data from 87 patients is shown above. Certainly, in the TBI literature, both very high and very low BP predict poorer outcome and likely the same will hold true for SCI.

A preliminary analysis of comparable OR data from Santa Clara Valley Medical Center shows a dose response effect of exposure to hypotension during surgery on expected neurological recovery in individuals with traumatic SCI. Thirty two subjects with traumatic SCI that received surgery and acute rehabilitation at Santa Clara Valley Medical Center and had autonomic data collected at 1 second intervals through surgery, were studied. American Spinal Injury Association Impairment Scale and Functional Independence Measure data were collected pre-operatively, post-operatively, at admission to acute rehabilitation, and at discharge from acute rehabilitation. The surgical information system (SIS) data capture system was queried for mean arterial pressures (MAPs) for all subjects in the study, MAP values were obtained on a second by second basis and this data was condensed and analyzed on a minute by minute basis. Exposure to MAPs was quantified at sequential thresholds from 50 mmHg to 150 mmHg. Change in American Spinal Injury Association Impairment Scale and Functional Independence Measure Score was calculated based on the earliest post-operative PM&R evaluation vs. PM&R evaluation at discharge from acute rehabilitation. Linear regression models were used to estimate the rate of recovery based on time exposure to hypotension. The rate of recovery of AIS motor scores was found to have a positive slope for exposure to hypotension until a threshold MAP of 70 mmHg (Fig. 3). Exposure to MAPs less than 70 mmHg was found to have a progressive deleterious effect on predictive recovery rate. A similar analysis of FIM motor scores was inconclusive on functional

outcomes based on exposure to hypotension. The inflection point in AISA motor score recovery for MAP exposure is 75-80 . There was no further change in ASIA motor score for MAP exposure > 95 in this sample.

Figure 3. Slope of change in ASIA Impairment Scale scores vs minutes less than a MAP of x (MAP is shown on the x axis) during surgery. These data suggest that low MAP during surgery also predicts poorer outcome.



These preliminary results suggest a dose response effect of exposure to hypotension on expected neurological recovery. These data provide clinical evidence that MAP deviations during the operative period in patients with acute spinal cord injury may have long term effects in their trajectory of recovery. This cohort has excellent longer-term outcome measures and will be an important comparison data set for comparing to the ZSFGH OR data. This work is ongoing.

Specific Aim 3: Determine the effects of episodes of hypotension and hypertension on the recovery of locomotor and bladder and bowel function in our rat model of high thoracic contusion SCI. We will examine the effects of commonly used vasopressors on outcome.

Major Task 5: Establish methods for BP regulation using the proposed hypo- and hypertensive treatments in the high thoracic injury model.
Subtask 1: Consult with clinical investigators to appropriately model the cardiovascular manipulations in the animal study.
Dr. Jonathan Pan MD, PhD of the Anesthesia Department who has training in SCI research, has helped with this aspect of the project.

Subtask 2. Perform control study in rats with high thoracic SCI to determine appropriate drug and dosing for hypo- and hypertensive treatments. (see below)

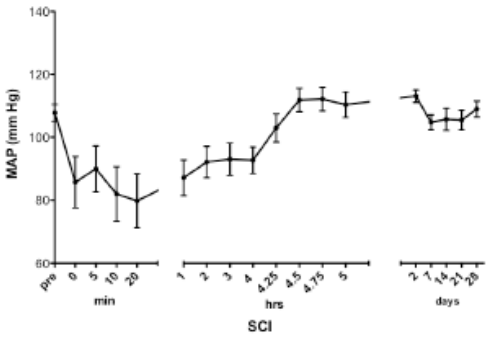
Major Task 6: Perform high thoracic, moderate-severe SCI in cohorts of rats and monitor BP, bladder and bowel functional measures, and locomotor function over 6 weeks. Groups include a) control group - no manipulation or treatments; b) group with MAP maintained at 75 with dopamine; c) group with BP maintained at 90 mm Hg using pressors; d) hypertensive group – BP maintained at 120 mm Hg induced with pressors.
Subtask 1: Implant telemetric pressure transducers for continuous monitoring of BP and activity in rats.
Subtask 2: Induce high thoracic injuries and monitor recovery of BP, bladder/bowel function, locomotor function.

We tested the effect of a 200 kilodyne impact using the Infinite Horizons contusion device at T2-3, on recovery of locomotor function in a group of pilot rats. This level of injury reliably produced an initial severe deficit followed by a fairly rapid recovery of weight supported stepping; but, the animals never recovered consistent forelimb-hindlimb coordination over the 6 weeks post-injury survival time (mean BBB final score of 12). Spontaneous urination was observed at a mean of 4.2 days. After this preliminary study, we decided to proceed with a more severe injury of 250kdynes using the IH device.

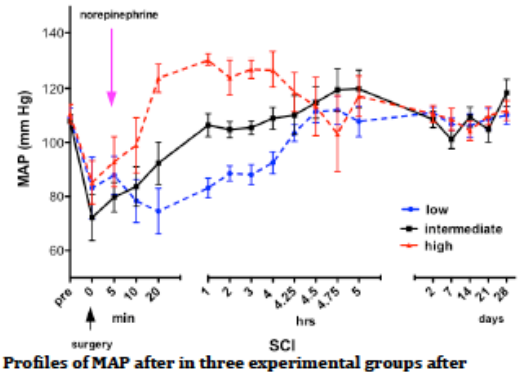
We established the surgical and procedural methods for continuous telemetric monitoring of blood pressure in the rats after spinal cord injury at the high thoracic level using the chronically implanted Data Science transducers. After piloting the surgical procedures for implanting the transducers and testing the data collection system, we proceeded to implant transducers in groups of animals with SCI and blood pressure, bladder function and locomotor recovery was monitored up to 6 weeks. Animals were followed for 6 weeks with BP monitoring, bladder function and behavioral testing.

A new approach to catheterizing the tail vein using a stylet and small diameter catheter was developed for this study. Isoflurane concentration was maintained at about 1.5% and norepinephrine was used to manipulate blood pressure. The results of this study are shown in the graphs in Figure 4 below, and indicate that both high and low blood pressure are predictive of poorer outcome; both locomotor and bladder function showed a significant effect. Interestingly, the amount of tissue sparing also reflected that the normotensive group had better outcome. These data are currently being prepared for publication.

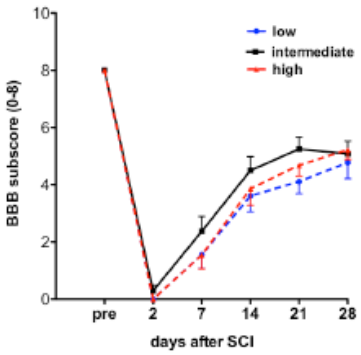
Figure 4. Outcomes of MAP modulation after high thoracic (T3) severe SCI in rats. Top two panels show MAP over time for all subjects (left), and for each group (right) of low (blue), intermediate (black) and high (red) MAP during the operative period. The middle two panels show locomotor (left) and bladder (right) recovery. The bottom panel shows MAP effects on tissue damage.



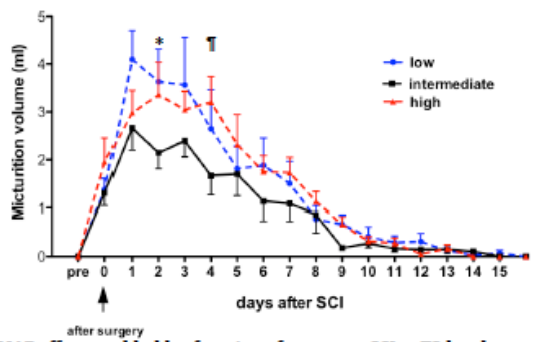
Profiles of MAP after spinal cord injury at T3 (250 Kdyne). Severe spinal cord injury at high thoracic spinal cord levels leads early hypotension. The decreased MAP is increased by 4 hr after injury. One Way ANOVA followed by Bonferroni's test, $p<0.0001$, $F_{17,216}=4.561$, N=13, *p<0.05, **p<0.001.



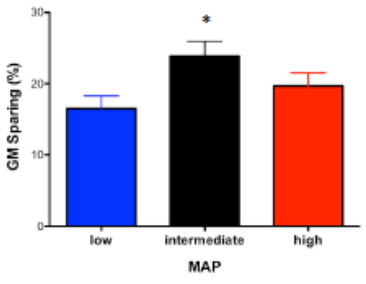
Profiles of MAP after in three experimental groups after severe SCI at T3 level. MAP was controlled by injecting norepinephrine after SCI for 4 hr after SCI. Three experimental groups were divided (low, MAP 70-90, n=9; intermediate, MAP 90-110, n=12; high, MAP 110-130, n=11). Two Way ANOVA followed by Bonferroni test, Interaction; $p<0.0001$, $F_{34,518}=2.56$, Treatment; $p<0.0001$. $F_{2,518}=23.05$. Time: $p<0.0001$. $F_{17,518}=8.08$.

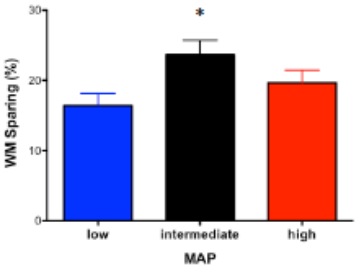


MAP effects on motor functional recovery after severe SCI at T3 level. Intermediate, but not low or high, MAP leads better motor functional recovery after SCI over 4 weeks (low, MAP 70-90, n=9; intermediate, MAP 90-110, n=12; high, MAP 110-130, n=11). Two Way ANOVA, followed by Bonferroni test, Interaction; $p=0.9232$, $F_{10,174}=0.4$, Treatment; $p=0.0395$, $F_{2,174}=3.29$, Time; $p<0.0001$. $F_{5,174}=144.47$.



MAP effects on bladder function after severe SCI at T3 level. Intermediate, but not low or high, MAP leads better recovery in bladder function after SCI over 4 weeks (low, MAP 70-90, n=9; intermediate, MAP 90-110, n=12; high, MAP 110-130, n=11). Two Way ANOVA followed by Bonferroni test, Interaction; $p=0.5880$, $F_{34,521}=0.93$, Treatment; $p=0.0003$, $F_{2,521}=8.23$, Time; $p<0.0001$, $F_{17,521}=34.41$. Posthoc test; *P<0.05 (between low and intermediate),¶p<0.05 (between intermediate and high)





MAP effects on tissue damages after severe SCI at T3 level. Intermediate MAP, but not low or high, results better tissue sparing in gray and white matter. One Way ANOVA followed by Tukey's test. **Left)** gray matter sparing, $P=0.0409$, $F_{2,29}=3.575$. *p<0.05 compared to low. **Right)** White matter sparing, $P=0.0423$. $F_{2,29}=3.535$. *p<0.05 compared to low.