Original Article

Access this article online
Quick Response Code:
Website: www.ajts.org
DOI: 10.4103/ajts.AJTS_44_18

Effectiveness of bone marrow-derived mononuclear stem cells for neurological recovery in participants with spinal cord injury: A randomized controlled trial

Rajeshwar Nath Srivastava, Ashok Kumar Agrahari#, Alka Singh#, Tulika Chandra[1], Saloni Raj

Abstract:

BACKGROUND: Complete lesion after spinal cord injury (SCI) remains irreversible with little hope of neurological recovery. Newer interventions such as re-stimulation of damaged neurons using artificial agents and the use of stem cells for neuronal regeneration have shown promising results.

AIM: This study was undertaken for evaluating the neurological status of acute SCI participants after stem cell augmentation and comparing them with other treatment methods.

SETTING AND DESIGN: Randomized controlled trial in the northern Indian population.

MATERIALS AND METHODS: A total 193 SCI participants of complete paraplegia with unstable T4–L2 injury having thoracolumbar injury severity score ≥4 were enrolled in this study. Participants were randomly allocated for three different treatment modalities, namely, conventional with stem cell augmentation (Group-1), conventional (Group-2), and conservative (Group-3). Neurological recovery after 1 year was evaluated through the ASIA Impairment Scale (AIS)-grading, sensory, and motor scores.

STATISTICAL ANALYSIS: *T*-test for sensory-motor score analysis of each group and analysis of variance for comparison of same variables between the groups.

RESULTS: After 1-year significant difference was observed in the AIS-grade, sensory and motor scores in-Group 1 ($P < 0.001$). In Group-1 versus 2, the mean difference at 1 year for AIS grade, sensory and motor scores were 0.40 ($P = 0.010$, 95% confidence interval [CI] 0.075–0.727), 8.52 ($P = 0.030$, 95% CI 0.619–16.419), and 4.55($P = 0.003$, 95% CI 1.282–7.815), respectively. In Group-1 versus 3, 1.03, 19.02 and 7.22 ($P < 0.001$ for each of the parameters) and in Group-2 versus 3, 0.63 ($P < 0.001$), 10.49 ($P = 0.009$), and 2.68 ($P = 0.019$), respectively.

CONCLUSIONS: Significant motor neurological recovery and AIS-grade promotion was observed in Group-1 as compared to Group-2 and 3.

Keywords:

Acute spinal cord injury, autologous bone marrow mononuclear stem cells, conventional treatment, neurological recovery, thoracolumbar injury severity score

Departments of Orthopedic Surgery and [1]Transfusion Medicine, King George's Medical University, Lucknow, Uttar Pradesh, India

#Both the authors have contributed equally.

Address for correspondence:
Dr. Rajeshwar Nath Srivastava, Department of Orthopedic Surgery, King George's Medical University, Nabiullah Road, Daliganj, Lucknow - 226 018, Uttar Pradesh, India.
E-mail: drrnsrivastava@yahoo.com

Submission: 03-04-2018
Accepted: 02-12-2018

Introduction

Traumatic spinal cord injury (SCI) is a seriously debilitating disease with high mortality, and among survivors, a high degree of morbidity due to both motor and sensory deficit. Unfortunately, in spite of best efforts, little success has been achieved by any therapeutic modality in terms of neurological recovery.[1] The available

How to cite this article: Srivastava RN, Agrahari AK, Singh A, Chandra T, Raj S. Effectiveness of bone marrow-derived mononuclear stem cells for neurological recovery in participants with spinal cord injury: A randomized controlled trial. Asian J Transfus Sci 2018;XX:XX-XX.

modalities for neurological recovery produce only minor improvements.

Despite a great deal of advancement in therapeutics, the life expectancy, prognosis, functionality, and quality of life remain poor in SCI patients with impaired neurological status. The recovery rate in SCI patients remains poor with any type of treatment because the neuronal cells, which are already in the highly differentiated stage, have negligible regenerative power. Several researchers have attempted the induction of controlled differentiation of the fully-modified brain cells to turn back as undifferentiated progenitors, either by using a differentiation-inducing agent[2] or by stem cells application (trans-differentiation).[3] We have already conducted a study in support of this for neurogenesis enhancement and axonal re-myelination using olfactory ensheathing cells under defined media.[4] Further, our group had worked on ways to stimulate stem cells to regenerate neurons for functional recovery with promising results.

In SCI, the clinical application of embryonic, umbilical cord, adipose tissue, and bone marrow-derived mono-nuclear stem cells (BM-MNSCs) has been studied.[5] The advantages of using BM-MNSCs are: first, one can minimize all problems associated with the immunological rejection which are frequently caused in allogenic cell transplantation,[6] second, autologous cell infusion is considered safe by not being associated with carcinogenesis.[7]

Earlier, we conducted a Phase I trial to determine the safety and efficacy of noncultured autologous BM-MNSCs in the management of acute SCI (ASCI), and their role in neurological recovery.[8] There was a significant difference in percent mean recovery with stem cell application at the 12th month of follow-up.

The current study was performed to evaluate the effect of BM-MNSCs as an adjuvant to conventional management of traumatic SCI for neurological recovery.

Materials and Methods

The current study, as Phase 2 trial, was conducted in the SCI Unit, Department of Orthopedic Surgery in collaboration with the Department of Transfusion Medicine, King George's Medical University (KGMU), Lucknow (UP), India from February 2013 to June 2016. This study was designed using the results of the Phase 1 randomized clinical trial on 110 ASCI participants to evaluate the safety and feasibility of application of BM-MNSCs and the role of surgery, omentoplasty and BM-MNSCs on clinical outcomes. Before this, the algorithm of aspiration, collection, isolation, and infusion of BM-MNSCs in SCI were standardized by us.[9]

Study population

The target population participating in this study was from Northern India and Nepal.

Study design

This is an open-label, individually randomized controlled, Phase II trial based on computer-generated random table (randomized controlled trial). As per random table, participants were randomized into three parallel groups [Supplementary Table 1].

Study groups

- Group 1 (Stem cell augmentation): ASCI participants managed by posterior instrumentation (titanium pedicle screw and rod devices) followed by infusion of autologous BM-derived stem cells as an adjuvant
- Group 2 (Conventional): ASCI participants managed by posterior instrumentation (titanium pedicle screw and rod devices)
- Group 3 (Conservative): ASCI participants managed nonoperatively.

Sample size calculation

The sample size was calculated by using mean difference and standard deviation (SD) of recovery based on ASIA scores of Group 1 and Group 3 participants from Phase 1 trial. The mean difference and SD of stem cell augmentation group versus conservative group were 8.118 and 14.9, respectively. The level of significance was set at 5% ($Z_{\alpha/2} = 1.96$) and the power of the study was 80% ($Z_{\beta} = 0.84$). On adding 10% loss in follow-up, each group had 60 participants with a total of 180 participants for all the three groups.

Selection criteria of the participants

Inclusion criteria

- Participants of ASCI having complete lesion (ASIA Impairment Scale [AIS]-A grade) with thoracolumbar injury severity score (≥ 4) (unstable injury requiring stabilization by surgery)
- Thoracolumbar spine injury level between T4 and L2 vertebra
- Age between 18 and 65 years of either gender
- Duration of injury <6 weeks
- Sagittal continuity of the spinal cord and the presence of cord hemorrhage on MRI.

In MRI assessment of SCI, the axial and sagittal T2W images, and T2*W GRE images are particularly useful. The most common injuries following cord trauma are edema, hemorrhage, and a mixture of both.[10] Cord transaction patterns having worst prognosis are best predicted by the sagittal discontinuity of the spinal cord, whereas sagittal continuity provides a possibility of spinal cord containment within its sheath.[11,12]

Exclusion criteria

- Polytrauma patients with injury spine and associated thoraco-abdominal injuries and/or head injury
- Medically unfit patients not suitable for surgery
- Patients with other comorbid conditions such as osteoporosis, pressure sores (Grade III–IV), psychiatric illness and those on steroids or other immune suppressants
- Patients who did not give their consent for participating in the study.

All enrolled participants were provided the standard surgical, medical as well as rehabilitation facilities. The assessment tool for recording the neurological status was the one proposed by the American Spinal Injury Association (ASIA). AIS grade, sensory, and motor scores were recorded at fixed time points after injury for neurological recovery. AIS Grade A, B, C, and D being nonparametric data were given numeric values such as 0, 1, 2, and 3, respectively, and then analyzed. In this study, we have documented neurological assessment at baseline and neurological recovery after 1 year [Supplementary Table 2].

Intervention

Participants in all the groups were managed for pressure offloading by turning and repositioning them every 2 h. The normal curvatures of the spine were maintained with the help of pillows, and care of bladder and bowel was ensured in participants suffering from incontinence. Routine investigations of fitness for surgery and anesthesia were done for surgical Groups 1 and 2. Standard operating procedures were used for instrumentation of pedicle screw and rods under fluoroscopic control. Distraction or compression was applied if required for reduction and stabilization of the fracture. Adequacy of the reduction was confirmed in anterior-posterior and lateral fluoroscopic views.

In addition, for Group 1 participants, aspiration, isolation, and purification of autologous BM-MNSCs were done as per algorithm described in our earlier study.[9] BM aspiration was done for the preparation of stem cell concentrate required for autologous infusion. Approximately, 80–90 ml of autologous BM was aspirated from the posterior iliac crest and collected in primary CPDA bag of Quadruple CPDA-1 blood bag set. The crude BM was differentially centrifuged at 1200 rpm for 10 min at 10°C. The supernatant (plasma with nucleated cells) was transferred in one of the empty satellite bags of the quadruple set by using plasma expresser, making it rich in mononuclear cells along with leukocyte. It was then centrifuged at 2500 rpm for 10 min at 10°C and separated into a second empty satellite bag, leaving a precipitate (about 15 ml) of MNSCs in the first bag as a buffy coat. Approximately, 10 ml of precipitate was used for infusion and rest of the sample was sent for MNCs and CD34+ count which was found to have a mean count of $2.41 \pm 1.198 \times 10^6$ live cells.

After fixation of the spine, spinous process of the fractured vertebra was removed along with portion of the lamina to visualize ligamentum flavum, which was excised for exposing the spinal cord. The dura mater of the spinal cord was held carefully with plain forceps at two different points separated by 1 cm distance. A stab incision with a #15 blade on a #7 knife handle was made in the dura mater up to the depth of arachnoid matter. Epidural catheter was placed in the subarachnoid space at the site of the injured cord. The arachnoid was closed over the catheter with silk 3–0. Fascia and skin closure was done leaving the other end of the tip of the catheter outside the stitched wound. The catheter was connected to the infusion pump, through which autologous stem cells were infused at the rate of 1 ml/h. The infiltration of infusion sample was done under aseptic condition in the postoperative room. After complete infusion of the BM-MNSCs, the catheter was pulled out carefully ensuring the presence of the blue tip at the end of the catheter, which confirms complete removal of the catheter from the spinal cord.

After 48 h of surgery, in both the surgery groups, the SCI participants were mobilized on a wheelchair with anterior hyper-extension braces. Following removal of stitches, preferably on the 10th day, the participants were transferred to the Department of Physical Medicine and Rehabilitation for structured rehabilitation protocol. The SCI participants of Group 3 were provided the same rehabilitation facilities as provided to both surgical groups. The facilities included early mobilization depending on neurological status, physiotherapy to facilitate recovery and prevention of known complications along with vocational training. The most common complications seen following ASCI were pressure sores, contractures, spasticity, bladder, bowel, and sexual dysfunction. SCI participants were followed-up in ortho outpatient department at 3-month interval for 1 year.

Statistical analysis

Data of all the participants were collected at the time of admission (baseline) and after 1 year (as follow-up). Data were analyzed using statistical analysis software packages, namely, Statistical Package for Social Sciences (SPSS) version 16 (South Wacker Drive, Chicago, IL, USA). 0 and GraphPad. Paired *t*-test was used to compare AIS grades, motor, and sensory scores at baseline and follow-up in terms of mean and (mean ± SD) with 95% confidence interval (CI) at 5% level of significance. Groups were compared with each other using one-way analysis of variance with

Bonferroni Correction *Post hoc* analysis to evaluate the mean difference, 95% CI and related *P* values of AIS grade, sensory, and motor scores after 1 year.

Desirable outcomes

BM-MNSCs augmentation improves neurological recovery in ASCI.

Statement of ethics

This study was approved by the Ethics Committee (IEC 60th ECM II-B/P14) and stem cell ethics committee (02/ISCES-12) of KGMU. The trial is registered under the Clinical Trial Registry of India (acknowledgment no. is REF/2017/08/015121). The study procedure was explained to all the participants in their native language. Written informed consent was also obtained from all the participants.

Results

Participants were recruited as per inclusion criteria and divided into three groups according to the treatment plan. A total of 220 participants were enrolled for the study. Twenty-seven out of 220 participants could not complete the study either due to drop out in between the study or lost to follow-up. Finally, 193 participants who could be followed for a year were included. Group 1 had 70 participants who underwent conventional surgery along with BM-MNSCs as an adjuvant. In Group 2, there were 68 participants who were treated by conventional surgery and Group 3 included 55 participants who underwent nonoperative conservative therapy. The maximum drop out was seen in the conservative group because of the nonoperative treatment mode, in which participants were not willing for follow-up. As pretreatment record, the complete neurological assessment was done at baseline using AIS grading, sensory, and motor scores. After 1 year, the neurological assessment was again performed and compared with baseline values.

Demographic information

Majority of the participants were men, 166 individuals (86.01%) and 28 were female (13.99%). All the groups had male dominance (Groups 1, 2, and 3 had 90%, 85.29%, and 80% of male participants, respectively) [Table 1]. The age group of 18–30 years was most prone to SCI followed by other age groups. The mean age of Groups 1, 2, and 3 were 30.84 ± 10.56, 31.30 ± 9.74, and 33.42 ± 12.07 years, respectively [Table 2]. Fall from height (70.98%) and road traffic accident (16.58%) were the two most common modes of injury in our study. The group-wise distribution of age and mode of injury were almost the same in all the three groups [Table 1]. The mean ± SD of admission within 2 weeks for Groups 1, 2, and 3 were 3.61 ± 2.84, 4.7 ± 4.12, and 5.14 ± 3.84 with CI of 2.78–4.45, 3.26–6.14, and 3.62–6.66, respectively. Almost the similar trend was seen in groups whose participants were admitted between 2–4 and 5–6 weeks. The mean ± SD of management within 2 weeks of injury for Groups 1 and 2 were 6.07 ± 1.90 and 6.75 ± 2.63, respectively. Again, a similar trend was seen in participants of both the groups who were managed between 2–4 and 5–6 weeks of injury [Table 2].

ASIA Impairment Scale grade comparison

After 1 year of follow-up, improvements in sensory and motor abilities were observed in participants of all the groups, wherein the stem cell group showed the best results. In AIS grading, significant improvement was observed in Group 1 as compared to that of other groups. In Group 1, 21.43% of participants remained in AIS A, whereas the percentage improvement to AIS B, C, and D was 30%, 41.43%, and 7.14%, respectively, whereas

Table 1: General characteristics of subjects

	Number of subjects			
	Total (*n*=193), *n* (%)	Group 1 (*n*=70), *n* (%)	Group 2 (*n*=68), *n* (%)	Group 3 (*n*=55), *n* (%)
Gender				
Male	166 (86.01)	63 (90)	58 (85.29)	44 (80)
Female	27 (13.99)	7 (10)	10 (14.7)	11 (20)
Age group (years)				
18-30	105 (54.4)	41 (58.57)	36 (52.94)	28 (50.9)
31-45	68 (35.23)	24 (34.29)	28 (41.18)	16 (29.09)
46-60	20 (10.36)	5 (7.14)	4 (5.88)	11 (20)
Mode of injury				
Fall from height	137 (70.98)	52 (74.28)	46 (67.65)	39 (70.9)
Road traffic accident	32 (16.58)	12 (17.14)	11 (16.18)	9 (16.36)
Weight over back	16 (8.29)	4 (5.71)	7 (10.29)	5 (9.09)
Others	8 (4.14)	2 (2.85)	4 (5.88)	2 (3.64)
Level of injury				
Level T4-T9	61 (31.61)	27 (38.57)	20 (29.41)	14 (25.45)
Level T10-L2	132 (68.39)	43 (61.43)	48 (70.59)	41 (74.55)

Values are represented as frequency and percentage (%)

in Group 2, the AIS A, B, C, and D values were 32.82%, 38.24%, 27.94%, and 0.0% and in Group 3, these were 78.18%, 12.73%, 9.09%, and 0.0%, respectively [Table 3].

Baseline mean values for AIS grade, sensory and motor scores were 0 ± 0, 144.03 ± 14.91, and 50 ± 0, respectively, in Group 1. After 1 year, the mean values increased to 1.34 ± 0.9 (95% CI: 1.56–1.13), 169.34 ± 18.62 (95% CI: 30.08-20.55), and 58.83 ± 10.70 (95% CI: −11.38–−6.28), respectively, with $P < 0.001$ for each of the above parameters. In "Group 2," baseline mean values for the above three parameters were 0 ± 0, 147.24 ± 16.56, 50 ± 0, and after 1 year were increased to 0.94 ± 0.79 (95% CI: 1.13–0.75), 160.82 ± 21.44 (95% CI:-16.44 to-10.73), and 54.28 ± 6.31 (95% CI:-5.81 to-2.75), respectively, with $P < 0.001$ for each parameter. These data indicate that in all the three parameters, the AIS grades, sensory, and motor scores highly significant improvement was observed in both the surgery groups (stem cell augmentation group and conventional group). In "Group 3" (conservative, nonoperative group) baseline mean values were 0 ± 0, 148.33 ± 15.37, 50 ± 0, respectively, whereas, after 1 year values increased to 0.31 ± 0.63 ($P = 0.0007$, % CI: 0.48–0.14), 150.33 ± 16.87 ($P = 0.19$, % CI: 5.03–1.03), 51.60 ± 5.13 ($P = 0.0245$, 95% CI: −2.99–−0.21), respectively, and were highly significant in AIS grading only [Table 4].

A *Post_hoc* analysis was performed for inter-group analysis to compare AIS grades, sensory, and motor scores in all the three groups (Group 1 vs. 2, 1 vs. 3, and 2 vs. 3) at baseline and after 1 year. The mean difference values for the AIS grade assessment were 0.40 ($P = 0.010$, 95% CI 0.075–0.727), 1.03 ($P < 0.001$, 95% CI 0.688–1.378), and 0.63 ($P < 0.001$, 95% CI 0.285–0.979) in Group 1 versus 2, 1 versus 3, and 2 versus 3, respectively. The mean difference values of sensory scores for treatment Group 1 versus 2, 1 versus 3, and 2 versus 3 were 8.52 ($P = 0.030$, 95% CI 0.619–16.419), 19.02 ($P < 0.001$, 95% CI 10.285–27.375), and 10.49 ($P = 0.009$, 95% CI 2.082–18.910), respectively. Similarly, the mean difference values of motor scores were 4.55($P = 0.003$, 95% CI 1.282–7.815), 7.22 ($P < 0.001$, 95% CI −3.77–10.685), and 2.68 ($P = 0.019$, 95% CI −0.799–6.158), respectively [Table 5]. These results indicate Group 1 (stem cell augmentation) had statistically significant difference from Group 2 and 3 in all the three specified parameters.

Table 2: Duration between injury to hospitalization and injury to management

Groups	Times	Mean±SD	P[a]	95% CI
Duration between injury to hospitalization				
Group 1	<2 weeks	3.61±2.84	<0.001*	2.78-4.45
	2-4 weeks	16.36±3.78		14.54-18.19
	5-6 weeks	30.00±1.41		27.74-32.25
Group 2	<2 weeks	4.70±4.12	<0.001*	3.26-6.14
	2-4 weeks	17.48±4.14		15.96-19.00
	5-6 weeks	29.33±0.57		27.89-30.76
Group 3	<2 weeks	5.14±3.84	<0.001*	3.62-6.66
	2-4 weeks	18.50±4.29		16.59-20.40
	5-6 weeks	31.00±2.00		28.90-33.09
Duration between injury to surgery				
Group 1	<2 weeks	6.07±1.90	<0.001*	5.47-6.67
	2-4 weeks	18.31±3.72		16.66-19.96
	5-6 weeks	31.28±1.70		29.70-32.86
Group 2	<2 weeks	6.75±2.63	<0.001*	5.79-7.70
	2-4 weeks	18.29±3.36		17.05-19.52
	5-6 weeks	30.40±1.51		28.51-32.28

Values are represented as mean±SD. [a]ANOVA, *Significant. 95% CI = Confidence interval, SD =Standard deviation

Table 3: American Spinal Injury Association impairment scale grades in different groups at baseline and 1 year

Groups	ASIA grades							
	At admission				After 1 year			
	A, *n* (%)	B	C	D	A, *n* (%)	B, *n* (%)	C, *n* (%)	D, *n* (%)
Group 1	70 (100)	0	0	0	15 (21.43)	21 (30)	29 (41.43)	5 (7.14)
Group 2	68 (100)	0	0	0	23 (33.82)	26 (38.24)	19 (27.94)	0
Group 3	55 (100)	0	0	0	43 (78.18)	7 (12.73)	5 (9.09)	0

Values are represented as frequency and percentage (%).AIS-A = Complete (no sensory or motor function), AIS-B = Incomplete (sensory present but no motor function), AIS-C=Incomplete (motor function is also present), AIS = ASIA impairment scale, ASIA = American Spinal Injury Association

Table 4: Intra-group comparison of American Spinal Injury Association impairment scale grades, sensory, and motor scores at baseline and 1 year

Groups	AIS (mean±SD)		P[a]	Sensory (mean±SD)		P[a]	Motor (mean±SD)		P[a]
	Baseline	After 1 year		Baseline	After 1 year		Baseline	After 1 year	
Group 1	00±00	1.34±0.90	0.0001*	144.03±14.91	169.34±18.62	0.0001*	50.00±00	58.83±10.70	0.0001*
	95% CI - 1.56–−1.13			95% CI - 30.08–−20.55			95% CI - 11.38–−6.28		
Group 2	00±00	0.94±0.79	0.0001*	147.24±16.56	160.82±21.44	0.0001*	50.00±00	54.28±6.31	0.0001*
	95% CI - 1.13–−0.75			95% CI - 16.44–−10.73			95% CI - 5.81–−2.75		
Group 3	00±00	0.31±0.63	0.0007*	148.33±15.38	150.33±16.87	0.19	50.00±00	51.60±5.13	0.0245*
	95% CI - 0.48–−0.14			95% CI - 5.03-1.03			95% CI - 2.99–−0.21		

Values are represented as mean±SD. [a]Paired *t*-test, *Significant. 95% CI = Confidence interval, SD = Standard deviation, AIS = ASIA impairment scale, ASIA = American Spinal Injury Association

Table 5: Inter-group analysis of the American Spinal Injury Association impairment scale grading, sensory, and motor scores after 1 year

Parameters	Groups comparision	Mean diffrence	*P*[a]	95% CI
AIS scoring	1 versus 2	0.40	0.010*	0.075-0.727
	1 versus 3	1.03	0.000*	0.688-1.378
	2 versus 3	0.63	0.000*	0.285-0.979
Sensory	1 versus 2	8.52	0.030*	0.619-16.419
	1 versus 3	19.02	0.000*	10.655-27.375
	2 versus 3	10.49	0.009*	2.082-18.910
Motor	1 versus 2	4.55	0.003*	1.282-7.815
	1 versus 3	7.22	0.000*	3.771-10.685
	2 versus 3	2.68	0.193	−0.799-6.158

[a]One-way ANOVA with Bonferroni correction, *Significant. 95% CI = Confidence interval, AIS = ASIA impairment scale, ASIA = American Spinal Injury Association

Discussion

Traumatic injury to the spinal cord immediately causes primary insult to the neural tissue, which remains irreversible and resistant to any intervention.[13-15] After the primary injury, the inflammatory process gets activated and leads to secondary injury phase.[16] The main hindrance in the process of neuronal regeneration is growth inhibitors present at the site of injury.[17] Earlier studies were mostly focused on preventing and reducing the extent of secondary injury which may further damage the spinal cord.[18,19] An initial surgery is usually performed to provide support to damaged tissues and reduce the compression impact.[20,21] Surgery helps in spinal stabilization, preventing spinal deformity, and facilitating patient mobility but not in neurological recovery.[22]

The available modalities for neurological recovery include the use of steroids and GM-1 ganglioside,[19] functional electrical stimulation,[23] retraining neural circuits to restore body functions, use of adaptive devices for communication, physical and occupational therapy, rehabilitation, and self-grooming techniques.[24]

The repair of already damaged neurons may be initiated by stimulation of factors responsible for neuronal repair and regeneration. Studies have shown that stem cells can be stimulated to form new neurons, but their contribution to the healing process has not been supported by sufficient evidence.[25] It has been observed by the studies that intrinsic adult stem cells and progenitor cells accumulate at the site of cord injury and help in neural tissue repair.[26] Neural progenitor cells secrete neurotropic factors, triggering the growth of injured neurons.[27] Stem cells make axons incapable of recognizing growth inhibitory molecules leading to axonal growth.[13] Activities such as walking and sensory perception have shown marked improvement over time with stem cell therapy.[14] Hematopoietic stem cells indirectly improve muscle strength and nerve regeneration,[28,29] whereas other studies suggested the use of granulocyte macrophage colony stimulating factor for the same.[30]

This study was performed to evaluate the effect of BM-MNSCs, used as an adjuvant, to conventional management[31] of traumatic SCI for neurological recovery. The results show that neurological recovery is better with stem cell augmentation in SCI surgery. According to the WHO, the global incidence of SCI is between 40 and 80 per million of population per year.[32] The increasing incidences and persisting poor prognosis in neurological recovery cause despair in SCI participants and helplessness among physicians involved in their management.[33] Unfortunately, no progress has been possible to reverse the primary injury caused to the spinal cord. Earlier studies were mostly focused on preventing and reducing the extent of the secondary injury and empowering people with SCI to return to an active and productive life. Researchers have been continually working on new treatments, including prostheses and medications that may promote nerve cell regeneration or improve the function of the nerves after ASCI.[34]

In this study, the mean age of the participants was 31.74 years. The results indicate that youngsters are more prone to SCI. It may be because they are more active, courageous, and live an aggressive lifestyle. In contrast to participants from Western countries, the most common types of injuries in our study were fall from height (70.98%) followed by road traffic accidents (16.58%). This may be because in developing countries in rural areas, from where most of these participants arrive, fall from trees and uncovered roofs are common.[35] This study was restricted to traumatic paraplegia participants only. Traumatic paraplegia is the result of damage to the cord at T2 level and below. To overcome the possibility of ascending edema following ASCI, in our study, participants of traumatic paraplegia having a complete lesion with the level of injury between T4 and L2 were included, to avoid any chance of SCI participants having quadriplegia. The most common vertebral level involved in our participants was between T10 and L2. Several studies have correlated "fall from height" with the injury of thoracolumbar junction (T10–L2).[36]

Many studies have reported no significant improvement in SCI, if surgery is delayed by more than 3 weeks.[37] In this study, the majority of participants were operated within 2 weeks of admission, but due to the late arrival of some participants, the duration between the injury to surgery reached up to 6 weeks. The participants reach late because in developing countries; besides lack of awareness, illiteracy, and poverty, there are

misconceptions and superstitions, which hinder early-specialized health care services.[38] Despite the late presentation of participants after injury, we have found significant improvement in the neurological recovery in all the groups. Participants operated within 2 weeks and those operated later had similar outcomes. Delay in surgery up to 6 weeks of injury did not affect recovery. In this study, we have used AIS grades to determine the severity of ASCI in participants. In comparison to the baseline information, there is an improvement in follow-up after 1 year. Group 1 shows maximum recovery, wherein 30% AIS Grade A achieved AIS Grade B, 41.43% Grade C, and 7.14% Grade D, which is quite remarkable. Group 2 shows recovery with 38.24% in AIS Grade B and 27.94% in Grade C. The reason for better recovery in Group 1 as compared to that of Group 2 could probably be attributed to stem cell augmentation, as this was the only factor that was different in the two groups. Group 3 also shows some recovery, but 78.18% of participants remained in the AIS grade A.

The recovery seen in the conservative group is due to spontaneous regeneration of neurons and is dependent on the physiology of participants. Various studies have shown that recovery in the conventional group is better than that in the conservative group because surgery provides stability to the spine and helps in the recovery of the injured spinal column. Surgery also reduces the possibility of further damage and arrests the secondary injury phase after ASCI.[39] Decompressing the spinal cord further relieves the pressure on the cord and improves its vascularity. The recovery of participants in the conventional group was better as compared to those of conservative group, but was low as compared to that of the stem cell augmentation group. The plausible explanation for this is that the BM-MNSCs can repair the damaged area and may induce recovery by signals provided by the activators and inhibitors present at a particular niche of the damaged area. After infusion, BM-MNSCs also get converted into various types of cells (neurons, astrocytes, and oligodendrocytes) as per the requirement of the damaged site. Application of stem cells as an adjuvant at the site of injury provided the favorable environment for neurological recovery by attracting biochemicals required for the accelerated initiation, cell division, and differentiation of progenitor cells of peripheral blood into required cell types. At the same time, infused stem cells differentiate into neurons under the influence of existing biochemical environment caused by trauma.[40]

AIS evaluation has been performed at two specific stages, first, at admission (pre-treatment) and the other after one year of follow-up. Mean values for AIS grade, sensory, and motor scores for Group 1 and Group 2 were 1.34, 169.34, 58.83 and 0.94, 160.82, and 54.28, respectively. A significant difference was found between both the groups with stem cell group showing better results in all the three parameters. Group 3 scores were almost similar to the baseline, which suggests a very small recovery. The results suggest that stem cells have a role in neurological recovery. Surgical methods are better than conservative treatment was also established by the scores and the AIS grades achieved after 1 year in all the groups.

Comparison among the groups was made to obtain mean difference values and *P* values of significance for AIS grade, sensory, and motor scores. The significant mean difference values of sensory scores were 8.52, 19.02, and 10.49, respectively, for all comparison groups (Group 1 vs. 2, 1 vs. 3, and 2 vs. 3). Group 1 had a statistically significant difference from Group 2 to 3 in all the three specified parameters. Group 2 also showed a statistically significant difference from Group 3 in some of parameters, but the result shown by participants of Group 1 versus 3 is not so robust.

Enumerating the strength of this study, it is based on our pilot study in which participants of ASCI were treated with stem cells. This study had standard outcome measures with clear documentation and data management to ensure reproducibility. A properly matched baseline characteristics in all the participants minimized confounders. The maximum participation of participants throughout the follow-up period was ensured by staying in touch with them and their attendants, either by phone or through home visits. Finally, participants participating in this study represented a large geographical area to minimize the regional variation bias, and to ensure that the results will be representative of the population studied. The limitations of this study were the exclusion of SCI cases having tetraplegia (cervical and cervicothoracic lesions), SCI >6-week-old, the inclusion of AIS A participants only (complete lesions) and the concern of spontaneous recovery seen in SCI which may lead to clinician's bias in interpretation of results. Then, although we have infused $2.41 \pm 1.198 \times 10^6$ (mean count) of live stem cells, but did not correlate them individually with the recovery. It was because of ethical reasons that we could not take the BM from conventional and conservative group participants for stem cell count. Furthermore, at the end of follow-up, the number of participants in each group was unequal and the conservative group had the least number of participants. This group had a general thought that, as they were not operated, there was not much need of regular follow-up. Nonetheless, fortunately, the numbers of participants in both the surgery groups were more than the required sample size of 60 in each group and only five short (55) in the conservative group.

Conclusions

This study concluded that the conventional treatment by surgery to stabilize the unstable spine gave better results over conservative treatment. The results of delayed surgery by up to 6 weeks were comparable with those of early surgery. The synergistic use of stem cells as an adjuvant with conventional treatment showed the best recovery results among all the specified groups and ultimately stresses on the significant role of BM-MNSCs in SCI. Application of stem cells as an adjuvant at the site of injury provided the favorable environment and the precursors for neurological recovery.

Acknowledgments

There was no external source of funding. Our institute is a state government organization and the required facilities for research are already available. All the procedure of research and intervention provided to participants were totally free of cost.

The authors acknowledge the contribution of the following personnel for their guidance and assistance during the initiation, conduction, and completion of this study-Prof. Saroj Chooramani Gopal, Department of Paediatric Surgery, IMS-BHU, Varanasi, India. Prof. G. K. Singh, Prof. Ajai Singh, Dr. Abhishek Agrawal, and Dr. Tribhav Goel, Department of Orthopaedic Surgery, KGMU, Lucknow, India. Prof. R. K. Garg, Department of Neurology, Prof. A. A. Mahdi, Department of Biochemistry, Dr. Chhitij Srivastava, Department of Neurosurgery, Prof. Abhijit Chandra, Department of Surgical Gastroenterology and Dr. Anit Parihar, Department of Radiology, KGMU, Lucknow, India. Prof. Raja Roy, Centre of Biomedical Research, SGPGIMS, Lucknow, India and Dr. Rajneesh K. Chaturvedi, Senior Scientist, Indian Institute of Toxicology Research, Lucknow, India.

Financial support and sponsorship

Nil.

Conflicts of interest

There are no conflicts of interest.

References

1. Wirasinghe V, Grover S, Ma D, Vizcaychipi M. Anaesthetic management of patients with acute spinal injury. Internet J Anesthesiol 2010;30:1-10.
2. Zhang L, Yin JC, Yeh H, Ma NX, Lee G, Chen XA, *et al.* Small molecules efficiently reprogram human astroglial cells into functional neurons. Cell Stem Cell 2015;17:735-47.
3. Lukovic D, Stojkovic M, Moreno-Manzano V, Jendelova P, Sykova E, Bhattacharya SS, *et al.* Concise review: Reactive astrocytes and stem cells in spinal cord injury: Good guys or bad guys? Stem Cells 2015;33:1036-41.
4. Singh N, Gopal SC, Srivastava RN, Chandra T, Agarwal SP, Singh SK. *In vitro* maintenance of olfactory mucosa with enriched olfactory ensheathing cells. J Stem Cell Res Ther 2013;2:132.
5. Orbay H, Tobita M, Mizuno H. Mesenchymal stem cells isolated from adipose and other tissues: Basic biological properties and clinical applications. Stem Cells Int 2012;2012:461718.
6. Beachy PA, Karhadkar SS, Berman DM. Tissue repair and stem cell renewal in carcinogenesis. Nature 2004;432:324-31.
7. Ra JC, Shin IS, Kim SH, Kang SK, Kang BC, Lee HY, *et al.* Safety of intravenous infusion of human adipose tissue-derived mesenchymal stem cells in animals and humans. Stem Cells Dev 2011;20:1297-308.
8. Srivastava RN, Gopal SC, Chandra T, Chandra A, Singh A, Agrahari AK, *et al.* Role of surgery, omentoplasty and autologous bone marrow derived mononuclear cells infusion on clinical outcomes after spinal cord injury – A randomized controlled trial. Int J Health Sci Res 2015;5:82-91.
9. Srivastava RN, Agrahari AK, Chandra T, Raj S. A closed, safe and cost effective alternative algorithm of isolation of bone marrow derived mononuclear cells. J Int Acad Res Multidiscip 2015;3:32-42.
10. Kumar Y, Hayashi D. Role of magnetic resonance imaging in acute spinal trauma: A pictorial review. BMC musculoskeletal disorders 2016;17:310.
11. Bozzo A, Marcoux J, Radhakrishna M, Pelletier J, Goulet B. The role of magnetic resonance imaging in the management of acute spinal cord injury. J Neurotrauma 2011;28:1401-11.
12. Kim SJ, Moon GJ, Chang WH, Kim YH, Bang OY; STARTING-2 (STem cell Application Researches and Trials In NeuroloGy-2) collaborators. Intravenous transplantation of mesenchymal stem cells preconditioned with early phase stroke serum: Current evidence and study protocol for a randomized trial. Trials 2013;14:317.
13. Murry CE, Keller G. Differentiation of embryonic stem cells to clinically relevant populations: Lessons from embryonic development. Cell 2008;132:661-80.
14. Pérez López S, Otero Hernández J. Advances in stem cell therapy. Adv Exp Med Biol 2012;741:290-313.
15. Zhang N, Yin Y, Xu SJ, Wu YP, Chen WS. Inflammation & apoptosis in spinal cord injury. Indian J Med Res 2012;135:287-96.
16. Hu J, Zhang G, Rodemer W, Jin LQ, Shifman M, Selzer ME, *et al.* The role of RhoA in retrograde neuronal death and axon regeneration after spinal cord injury. Neurobiol Dis 2017;98:25-35.
17. Ohtake Y, Li S. Molecular mechanisms of scar-sourced axon growth inhibitors. Brain Res 2015;1619:22-35.
18. Jain KK. Regenerative therapy for central nervous system trauma. In: Steinhoff G, editor. Regenerative Medicine – From Protocol to Patient. 3rd ed. Switzerland: Springer International Publishing; 2016. p. 115-45.
19. Carvalho MO, Barros Filho TE, Tebet MA. Effects of methylprednisolone and ganglioside GM-1 on a spinal lesion: A functional analysis. Clinics (Sao Paulo) 2008;63:375-80.
20. Ryken TC, Hurlbert RJ, Hadley MN, Aarabi B, Dhall SS, Gelb DE, *et al.* The acute cardiopulmonary management of patients with cervical spinal cord injuries. Neurosurgery 2013;72 Suppl 2:84-92.
21. Walters BC, Hadley MN, Hurlbert RJ, Aarabi B, Dhall SS, Gelb DE, *et al.* Guidelines for the management of acute cervical spine and spinal cord injuries: 2013 update. Neurosurgery 2013;60 Suppl 1:82-91.
22. Jug M, Kejžar N, Vesel M, Al Mawed S, Dobravec M, Herman S, *et al.* Neurological recovery after traumatic cervical spinal cord injury is superior if surgical decompression and instrumented fusion are performed within 8 hours versus 8 to 24 hours after injury: A single center experience. J Neurotrauma 2015;32:1385-92.
23. Evans RW, Wilberger JE, Bhatia S. Traumatic disorders. In: Goetz CG, editor. *Textbook of Clinical Neurology*. 3rd ed., Ch. 51. Philadelphia, PA: Saunders Elsevier; 2007.

24. Spinal Cord Injury: Hope through Research. NIH Publication No. 13-160. NINDS; July 2013. Available from: https://www.ninds.nih.gov/disorders/patient-caregiver-education/hope-through-research/spinal-cord-injury-hope-through-research. [Last modified on 2018 Aug 08; 12:14].
25. Mothe AJ, Tator CH. Advances in stem cell therapy for spinal cord injury. J Clin Invest 2012;122:3824-34.
26. Jones KS, Connor B. Intrinsic regulation of adult subventricular zone neural progenitor cells and the effect of brain injury. Am J Stem Cells 2012;1:48-58.
27. Vrana KE, Hipp JD, Goss AM, McCool BA, Riddle DR, Walker SJ, *et al*. Nonhuman primate parthenogenetic stem cells. Proc Natl Acad Sci U S A 2003;100 Suppl 1:11911-6.
28. Shen WJ, Shen YS. Nonsurgical treatment of three-column thoracolumbar junction burst fractures without neurologic deficit. Spine (Phila Pa 1976) 1999;24:412-5.
29. Baker M. Bone marrow stem cells, surgery assessed for spinal cord injury. Nature Reports Stem Cells 2009. Available from: https://www.nature.com/stemcells/2009/0904/090409/full/stemcells.2009.57.html. [Last accessed on 2018 Apr 01]. Doi:10.1038/stemcells.2009.57.
30. Yoon SH, Shim YS, Park YH, Chung JK, Nam JH, Kim MO, *et al*. Complete spinal cord injury treatment using autologous bone marrow cell transplantation and bone marrow stimulation with granulocyte macrophage-colony stimulating factor: Phase I/II clinical trial. Stem Cells 2007;25:2066-73.
31. Misra S, Sen S, Das S, Chatterjee A, Sengupta A, Saha S. Evaluation the results of surgical management of traumatic paraplegia in traumatic thoracolumbar fractures. Int J Res Med Sci 2016;4:2262-70.
32. Srivastava RN, Singh A, Garg RK, Agarwal A, Raj S. Epidemiology of traumatic spinal cord injury: A SAARC perspective. Int J Mol Biol Biochem 2015;3:9-22.
33. Yang NP, Deng CY, Lee YH, Lin CH, Kao CH, Chou P. The incidence and characterisation of hospitalised acute spinal trauma in Taiwan – A population-based study. Injury 2008;39:443-50.
34. Chacko V, Joseph B, Mohanty SP, Jacob T. Management of spinal cord injury in a general hospital in rural India. Paraplegia 1986;24:330-5.
35. Razzak AT. Early care following traumatic spinal cord injury (TSCI) in a rehabilitation centre in Bangladesh – An analysis. Disabil CBR Inclusive Dev 2013;24:64-78.
36. Price C, Makintubee S, Herndon W, Istre GR. Epidemiology of traumatic spinal cord injury and acute hospitalization and rehabilitation charges for spinal cord injuries in Oklahoma, 1988-1990. Am J Epidemiol 1994;139:37-47.
37. Yousefifard M, Rahimi-Movaghar V, Baikpour M, Ghelichkhani P, Hosseini M, Jafari A, *et al*. Early versus late spinal decompression surgery in treatment of traumatic spinal cord injuries; a systematic review and meta-analysis. Emerg (Tehran) 2017;5:e37.
38. Aswani KK, Subrahmanyam BV, Phanidra SV, Satish KS, Harish PN, Ramamohan P, *et al*. Demographic pattern, clinical profile and outcome of traumatic spinal cord injuries at a tertiary care hospital. Rom Neurosurg 2015;29:312-7.
39. Rajasekaran S, Kanna RM, Shetty AP. Management of thoracolumbar spine trauma: An overview. Indian J Orthop 2015;49:72-82.
40. Li M, Ikehara S. Bone-marrow-derived mesenchymal stem cells for organ repair. Stem Cells Int 2013;2013:132642.

Supplementary Table 1: Computer generated random number table

Subject number	Groups	Random numbers
1	2	0.379691597
2	3	0.826511184
3	2	0.626390294
4	1	0.52476002
5	2	0.38423567
6	1	0.596431231
7	2	0.363551065
8	1	0.038267156
9	2	0.243976593
10	3	0.337591993
11	3	0.922991656
12	1	0.981388731
13	3	0.880309098
14	3	0.455295657
15	2	0.650959411
16	2	0.521302357
17	3	0.856419611
18	1	0.072280925
19	2	0.116210275
20	2	0.310114162
21	2	0.715737802
22	1	0.339025221
23	3	0.22235138
24	3	0.724534926
25	3	0.442970448
26	2	0.147874383
27	3	0.033903279
28	3	0.979011285
29	3	0.16217519
30	1	0.981780414
31	3	0.775088438
32	3	0.610992297
33	1	0.637599785
34	1	0.564163613
35	1	0.74016944
36	3	0.292297306
37	1	0.120094748
38	2	0.781976938
39	1	0.387951285
40	1	0.572985361
41	2	0.347058089
42	1	0.277429124
43	2	0.518900366
44	1	0.286803152
45	2	0.520115372
46	1	0.525508453
47	1	0.59348895
48	1	0.843203105
49	3	0.20808
50	1	0.904458204
51	1	0.78062498
52	3	0.831248384
53	3	0.861647999
54	2	0.344417904

Contd...

Supplementary Table 1: Contd...

Subject number	Groups	Random numbers
55	2	0.674902365
56	3	0.994801921
57	1	0.824338099
58	2	0.75967438
59	3	0.220128963
60	1	0.218574498
61	3	0.491022986
62	3	0.658040752
63	1	0.719478884
64	1	0.137761463
65	3	0.0490272
66	3	0.396562856
67	3	0.512734026
68	3	0.113149317
69	2	0.633816836
70	2	0.463564631
71	2	0.895262476
72	1	0.602966628
73	2	0.811485079
74	3	0.944729554
75	1	0.729150839
76	2	0.022129039
77	2	0.800427832
78	3	0.020484784
79	1	0.347439363
80	2	0.013584913
81	2	0.946513751
82	3	0.144395366
83	2	0.897122424
84	2	0.29992265
85	3	0.768663499
86	1	0.448666161
87	3	0.968265434
88	3	0.562397966
89	1	0.479438355
90	2	0.560432281
91	2	0.610297489
92	1	0.486000972
93	1	0.711688911
94	2	0.889402243
95	3	0.452224664
96	1	0.740168457
97	1	0.765864377
98	3	0.675975322
99	1	0.231078238
100	2	0.860856493
101	1	0.627745947
102	2	0.784194508
103	3	0.454721028
104	3	0.615127956
105	1	0.763861445
106	3	0.531415144
107	1	0.640770206
108	1	0.522753706
109	2	0.974808892

Contd...

Supplementary Table 1: Contd...

Subject number	Groups	Random numbers
110	1	0.519330807
111	3	0.785856001
112	1	0.892269363
113	1	0.160209714
114	2	0.883511449
115	1	0.302554145
116	2	0.620697894
117	3	0.254555353
118	1	0.348993761
119	2	0.601861355
120	2	0.033719411
121	3	0.25806827
122	2	0.881962391
123	2	0.997635486
124	2	0.074555961
125	1	0.504423549
126	1	0.936731586
127	1	0.832966973
128	2	0.045613097
129	2	0.582627046
130	3	0.413905064
131	3	0.670844049
132	3	0.110731028
133	2	0.143868011
134	1	0.541707503
135	2	0.264096873
136	3	0.384344029
137	3	0.652926606
138	2	0.727197548
139	1	0.420559078
140	1	0.456590187
141	3	0.822688315
142	1	0.49628237
143	3	0.041932486
144	2	0.445526725
145	1	0.104513056
146	1	0.486760096
147	2	0.644425124
148	2	0.41466249
149	3	0.162011219
150	3	0.141040177
151	3	0.219040713
152	2	0.147631247
153	3	0.008752019
154	3	0.591987509
155	2	0.93616581
156	1	0.102459597
157	1	0.283961581
158	3	0.809201759
159	2	0.059533695
160	3	0.851372108
161	1	0.933543767
162	3	0.224078755
163	3	0.204600738
164	2	0.278396763

Contd...

Supplementary Table 1: Contd...

Subject number	Groups	Random numbers
165	3	0.440816528
166	2	0.752755481
167	3	0.732375328
168	2	0.852064425
169	2	0.949470832
170	1	0.159852727
171	2	0.357160501
172	3	0.5393509
173	1	0.704169159
174	3	0.062861198
175	1	0.867120013
176	3	0.018921388
177	2	0.564542985
178	2	0.310075636
179	1	0.777801383
180	2	0.814567661
181	2	0.772694835
182	1	0.352408732
183	2	0.638175748
184	2	0.856321592
185	3	0.585713091
186	2	0.87465694
187	2	0.794606356
188	1	0.230684358
189	3	0.020600605
190	1	0.645948846
191	1	0.617203562
192	3	0.155417885
193	3	0.874684865
194	2	0.475437254
195	2	0.407702187
196	2	0.577829604
197	3	0.515830601
198	3	0.146759892
199	3	0.171817855
200	1	0.606407918
201	3	0.946249798
202	2	0.27035361
203	3	0.092812486
204	1	0.998575296
205	2	0.238553067
206	1	0.026033118
207	1	0.99545938
208	1	0.534981311
209	3	0.644973775
210	3	0.989091339
211	1	0.982432692
212	1	0.63680991
213	3	0.087602035
214	1	0.644558885
215	2	0.541237674
216	1	0.489242512
217	3	0.923200653
218	2	0.398570752
219	1	0.562082058
220	2	0.240944539

Supplementary Table 2: Details of the participants

Subjects	Age	Gender	MOI	Baseline			After 1 year			LOI	TLISS
				AIS grade	Sensory	Motor	AIS grade	Sensory	Motor		
						Group 1					
1	20	1	1	0	128	50	1	160	50	T12	7
2	18	2	3	0	168	50	2	188	68	L1	8
3	40	1	1	0	136	50	0	136	50	T6	6
4	30	1	1	0	120	50	2	178	72	T8	6
5	36	1	3	0	152	50	1	152	50	T12	7
6	18	1	2	0	112	50	2	166	50	T8	6
7	28	1	1	0	104	50	2	172	68	T6	7
8	18	2	1	0	152	50	3	198	78	T12	7
9	50	1	2	0	152	50	2	182	66	T4	7
10	38	1	2	0	160	50	1	160	50	L1	6
11	43	1	2	0	160	50	0	160	50	T8	6
12	18	2	1	0	160	50	3	196	88	L1	8
13	35	1	1	0	168	50	1	168	50	T12	6
14	35	1	1	0	136	50	3	200	84	L1	6
15	18	1	1	0	160	50	2	184	58	L1	8
16	44	1	1	0	148	50	0	144	50	T9	6
17	30	2	3	0	136	50	2	188	68	T11	6
18	35	1	1	0	136	50	1	166	50	T12	6
19	22	1	2	0	152	50	0	144	50	T7	7
20	28	1	1	0	136	50	1	160	50	T8	9
21	45	1	1	0	140	50	0	140	50	T5	7
22	20	1	1	0	154	50	2	196	64	L1	6
23	26	1	1	0	136	50	2	168	62	T10	6
24	18	1	1	0	156	50	1	168	50	T7	6
25	55	1	1	0	152	50	0	152	50	T12	8
26	46	1	1	0	144	50	1	148	50	T9	6
27	24	1	1	0	144	50	2	188	64	T12	6
28	37	1	1	0	116	50	0	116	50	T6	8
29	28	1	2	0	88	50	0	120	50	T7	6
30	15	1	1	0	160	50	1	160	50	L1	6
31	18	1	2	0	144	50	1	162	50	T10	6
32	30	1	1	0	168	50	2	188	50	L2	7
33	20	1	2	0	152	50	0	152	50	T5	9
34	40	1	1	0	128	50	2	188	50	T10	7
35	24	1	2	0	140	50	1	160	50	T10	6
36	45	1	1	0	140	50	1	172	50	T9	6
37	25	1	1	0	136	50	1	160	50	T10	6
38	35	1	1	0	160	50	0	164	50	L1	8
39	18	1	1	0	148	50	2	166	50	T10	6
40	24	1	1	0	152	50	2	168	50	T11	6
41	27	1	1	0	144	50	0	144	50	T10	8
42	33	1	1	0	144	50	1	166	50	T11	6
43	30	1	1	0	152	50	0	152	50	T9	6
44	60	1	4	0	136	50	2	186	58	T10	6
45	30	2	4	0	152	50	2	188	66	T12	7
46	45	1	1	0	152	50	2	180	62	T12	9
47	40	1	2	0	160	50	1	160	50	L1	7
48	19	1	1	0	152	50	2	190	60	L1–2	6
49	25	1	1	0	152	50	2	184	58	L1	6
50	58	2	1	0	144	50	2	182	68	L1	8
51	45	1	1	0	152	50	2	178	72	T12	6
52	14	2	1	0	152	50	2	184	62	T12, L1	6
53	22	1	1	0	128	50	1	176	50	T8	7

Contd...

Supplementary Table 2: Contd...

Subjects	Age	Gender	MOI	Baseline			After 1 year			LOI	TLISS
				AIS grade	Sensory	Motor	AIS grade	Sensory	Motor		
Group 1											
54	25	1	1	0	128	50	0	144	50	T9	6
55	30	1	1	0	128	50	1	168	54	T7–8	7
56	35	1	2	0	144	50	0	144	50	T6	7
57	35	1	1	0	152	50	3	196	76	T12	7
58	32	1	1	0	144	50	0	144	50	T12	6
59	45	1	2	0	152	50	2	180	58	L1	6
60	25	1	1	0	124	50	2	182	62	T9	8
61	22	1	1	0	160	50	1	168	50	L1	6
62	22	1	1	0	144	50	1	182	64	T9–10	6
63	35	1	3	0	128	50	2	188	68	T9	8
64	22	1	1	0	144	50	2	186	58	L1	6
65	30	1	1	0	160	50	2	184	56	T12	6
66	35	1	1	0	144	50	2	190	50	T8	6
67	27	1	1	0	136	50	1	164	50	T9–10	7
68	30	1	1	0	144	50	1	160	50	T10–11	9
69	40	1	1	0	136	50	2	174	64	T7	7
70	24	1	1	0	160	50	3	192	66	L1	6
Group 2											
1	26	1	1	0	152	50	0	152	50	T4	6
2	30	1	1	0	136	50	2	154	62	T10	6
3	12	1	1	0	160	50	2	174	64	L1	7
4	34	1	3	0	128	50	0	128	50	T8	8
5	22	1	1	0	104	50	1	120	54	T4–5	7
6	15	2	1	0	160	50	2	174	68	L1	7
7	40	1	1	0	144	50	0	144	50	T8–9	6
8	35	1	3	0	148	50	2	168	62	T10–11	7
9	30	1	1	0	168	50	0	168	50	T5	6
10	29	1	2	0	108	50	0	108	50	T4–5	6
11	35	1	1	0	152	50	1	166	54	T12–L1	7
12	38	1	1	0	148	50	0	148	50	T10	6
13	60	1	1	0	148	50	2	176	66	T10	6
14	18	2	1	0	148	50	0	148	50	T6	6
15	18	2	1	0	160	50	2	188	68	L1	6
16	60	1	1	0	136	50	1	164	52	T10	6
17	18	1	1	0	168	50	1	188	56	L2	6
18	24	1	2	0	152	50	2	184	60	T12	5
19	25	1	1	0	152	50	1	166	52	T12	6
20	35	1	5	0	144	50	0	144	50	T9	5
21	42	1	3	0	160	50	1	184	52	L1	6
22	32	1	1	0	160	50	0	160	50	L1	7
23	22	1	3	0	166	50	2	178	68	L2	7
24	32	1	1	0	122	50	0	122	50	T4	6
25	40	1	1	0	152	50	1	188	52	T12	6
26	28	1	1	0	160	50	1	174	54	L1	6
27	27	1	1	0	160	50	2	184	65	L1	7
28	30	1	3	0	160	50	2	184	66	L2–3	8
29	55	1	1	0	144	50	1	168	52	T10–11	7
30	45	1	1	0	152	50	0	152	50	T9	6
31	40	2	1	0	144	50	1	168	50	T11–12	7
32	24	1	2	0	96	50	1	112	50	T4–5	6
33	35	1	1	0	144	50	0	144	50	T9	6
34	20	2	1	0	160	50	1	168	50	L2–3	7
35	50	1	1	0	128	50	0	128	50	T9	6

Contd...

Supplementary Table 2: Contd...

Subjects	Age	Gender	MOI	Baseline			After 1 year			LOI	TLISS
				AIS grade	Sensory	Motor	AIS grade	Sensory	Motor		
						Group 2					
36	14	2	5	0	136	50	0	136	50	T4	6
37	35	1	3	0	160	50	2	190	62	L1	6
38	20	1	1	0	160	50	0	160	50	T5	6
39	35	1	2	0	144	50	1	154	50	T11	6
40	15	1	1	0	152	50	1	170	50	L1	6
41	35	1	1	0	144	50	0	144	50	T11	5
42	25	1	1	0	152	50	2	178	66	L1	6
43	40	1	1	0	144	50	1	154	50	T12	5
44	30	1	1	0	160	50	1	168	50	L1	6
45	22	2	2	0	152	50	2	176	62	T12	7
46	24	1	2	0	144	50	1	160	50	T11	7
47	30	1	1	0	128	50	1	144	50	T8	6
48	40	1	1	0	152	50	2	178	68	T12	6
49	45	1	2	0	112	50	1	124	50	T7	6
50	30	1	1	0	160	50	2	186	64	L1–2	7
51	36	1	5	0	144	50	0	144	50	T10	7
52	30	1	1	0	152	50	1	164	50	T12	6
53	18	1	2	0	160	50	1	196	50	L1	6
54	45	2	1	0	152	50	1	164	50	T12	6
55	28	1	3	0	152	50	1	164	50	L1–2	6
56	35	1	2	0	152	50	1	168	50	L1	6
57	25	1	1	0	160	50	2	174	54	L1	6
58	35	1	1	0	144	50	1	160	50	T11	6
59	40	1	1	0	136	50	2	184	54	T7–8	6
60	38	1	2	0	152	50	1	160	50	T12	7
61	20	1	1	0	160	50	0	160	50	T11	6
62	28	1	5	0	168	50	0	168	50	L2	7
63	17	1	1	0	168	50	0	168	50	L2	6
64	17	2	1	0	152	50	2	174	56	L1	6
65	30	1	2	0	144	50	0	168	50	T7	6
66	35	1	1	0	160	50	0	160	50	T12	5
67	45	1	1	0	88	50	0	88	50	T5	7
68	40	2	1	0	144	50	2	174	62	T12	7
						Group 3					
1	36	2	1	0	160	50	2	160	50	L1	6
2	35	2	1	0	136	50	0	136	50	T9–10	6
3	18	2	1	0	160	50	0	160	50	T6	7
4	18	1	1	0	136	50	0	136	50	T10	6
5	50	1	1	0	152	50	2	174	54	T12	7
6	35	1	1	0	152	50	1	152	50	L1	7
7	40	1	1	0	136	50	0	136	50	T10	7
8	19	1	1	0	174	50	0	160	50	T12	6
9	22	1	1	0	136	50	2	136	50	T5	6
10	25	1	1	0	160	50	0	160	50	T12	8
11	20	1	1	0	120	50	0	120	50	T10	6
12	22	1	2	0	144	50	1	160	50	L1	6
13	22	1	4	0	136	50	0	136	50	T10	8
14	45	1	1	0	112	50	0	112	50	T11	6
15	30	1	2	0	160	50	1	168	50	L1–2	6
16	18	2	1	0	126	50	0	162	50	T6–7	6
17	19	2	1	0	160	50	0	160	50	T10	7

Contd...

Supplementary Table 2: Contd...

Subjects	Age	Gender	MOI	Baseline			After 1 year			LOI	TLISS
				AIS grade	Sensory	Motor	AIS grade	Sensory	Motor		
						Group 3					
18	22	1	1	0	136	50	0	136	50	T12	9
19	36	1	1	0	160	50	0	160	50	T12	7
20	45	1	1	0	136	50	0	136	50	T10	6
21	28	1	1	0	152	50	2	174	56	T12	7
22	30	1	1	0	160	50	0	152	50	T8	7
23	32	1	1	0	160	50	0	136	50	L1	6
24	35	1	2	0	160	50	2	184	60	L2	6
25	55	1	1	0	160	50	0	136	50	T10–11	6
26	48	1	1	0	160	50	0	160	50	T6	6
27	36	2	2	0	120	50	0	120	50	T10	6
28	55	1	1	0	160	50	1	160	50	L1	6
29	56	1	1	0	160	50	0	136	50	T12	6
30	60	1	1	0	112	50	0	112	50	T7	6
31	46	1	3	0	160	50	1	168	50	L1–2	7
32	36	2	2	0	126	50	0	162	50	T6–7	6
33	24	1	1	0	160	50	0	160	50	T6	7
34	38	1	1	0	152	50	0	152	50	T12	6
35	25	2	1	0	160	50	1	176	50	T10	6
36	25	1	2	0	160	50	0	160	50	L1	6
37	55	1	1	0	146	50	0	146	50	T11	5
38	36	1	5	0	138	50	0	138	50	T10	7
39	30	1	1	0	160	50	0	160	50	L1	7
40	40	2	1	0	122	50	0	122	50	T8	7
41	48	1	2	0	160	50	0	160	50	T7	8
42	30	1	3	0	160	50	0	160	50	T11	6
43	30	2	1	0	152	50	0	152	50	T12	6
44	28	1	1	0	114	50	0	114	50	T7	7
45	24	1	1	0	146	50	0	146	50	T11	6
46	22	1	3	0	152	50	1	168	50	T11	7
47	22	1	1	0	152	50	0	152	50	T12	7
48	26	1	2	0	160	50	0	160	50	T7	7
49	59	1	1	0	160	50	0	160	50	L1	6
50	48	1	1	0	160	50	0	160	50	L1	6
51	35	1	3	0	160	50	0	160	50	L1–2	8
52	18	1	1	0	136	50	0	136	50	T8–9	6
53	19	2	1	0	146	50	0	146	50	T11	6
54	30	1	1	0	160	50	0	160	50	L1	8
55	32	1	3	0	160	50	0	160	50	L2	6

Gender=1: Male, 2: Female, MOI=1: Fall from height, 2: Road traffic accident, 3: Weight over back, 4+5: Others, AIS grade=A: 0, B: 1, C: 2, D: 3, LOI = Level of injury, MOI=Mode of injury, TLISS = Thoraco-lumbar injury severity score, AIS = ASIA impairment scale, ASIA = American Spinal Injury Association