

# Autologous intravenous stem cells infusion in chronic stroke : a pilot study in Indian patients

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**Introduction:** Stroke is the leading cause of disability in India. Stem cell transplantation has claimed to restore function in experimental models as well in clinical trials of ischemic stroke<sup>1, 2</sup>. Mesenchymal stem cells have an advantage of easy feasibility, multi-potentiality and homing in characteristic to the infarcted core. This study evaluates the safety, feasibility and efficacy of autologous mesenchymal stem cell transplantation in chronic ischemic stroke. Functional MR imaging is used to monitor stem cell engraftment and functional recovery using BOLD contrasts, diffusion tensor imaging and spectroscopy.

**Methods:** Four ischemic stroke patients (mean age =44.5 , SD = 18.1) were recruited from the neurology clinics. The inclusion criteria were 3 months after the onset and within one year, MRC (medical research council) grade of hand muscles of at least >2; NIHSS (NIH stroke scale) of between 4 and 24, conscious and able to comprehend. The exclusion criteria were bleeding disorders, immunocompromised subjects, pregnancy, any other brain lesion (tumours, infectious diseases), severe spasticity, comatose/ mechanically ventilated patients and contraindication to MRI. The study was approved by the Institutional Ethics (stem cell) Committee. Informed consent was taken from all the subjects prior to bone marrow aspiration and during stem cell transplantation. Fugel meyer, brunnsstrom, barthel index and MRC grade of strength of hand muscles was used to assess the patients pre and post treatment. FMRI was performed to assess the effect of physiotherapy using block design with alternate baseline and activation task with a total of 80 whole brain EPI measurements (TR =4520ms, TE =44 ms, slices =31, slice thickness =4 mm). The subjects were asked to perform the motor task with paretic hand at self paced (minimum 0.5Hz) fist clenching of the hands.

**Intervention:** Subjects were grouped equally into experimental (stem cells and physiotherapy) and control group (physiotherapy only in patients 3,4). 50-60 ml of bone marrow aspiration was done under aseptic condition. The ex vivo culture of cells took around 21 ± 5 days in the stem cells facility which was good manufacturing practice (GMP) certified. Two patients (pat. id 1,2) were administered with 5x 10<sup>8</sup> mesenchymal stem cells in the median cubital vein for 4-6 hours followed by 6 weeks of physiotherapy. Safety tests at day 1, 3, 7, and at 6 weeks were performed. Intense exercise regimen was administered to all the patients for 5 days a week for 6 weeks. The aims of physiotherapy treatment were facilitation of voluntary control, strength training, tone reduction and functional activity training.

**Results:** All the four patients showed improvements on fugel meyer, barthel index and MRC grade (p<0.05). The experimental group patients did not report any adverse events and the routine blood tests were within normal limits during the four monitoring periods, which is indicative of its safety and feasibility. There was a considerable increase in the number of voxels in Brodmann area (BA) 4, 6 post therapy in both the groups, which indicates treatment induced plasticity in the lesioned cortex (figure 1). One patient (id 2) did not show any significant improvement and radiological markers post therapy (table1), however there was an increased activation of ipsilateral cerebellum post therapy suggesting its role for reorganization of corticospinal tracts. Fractional anisotropy asymmetry index of the affected hemisphere was calculated using the DTI software (M/s Siemens) with an index of 0.25 or less demonstrating a greater recovery in patients. Patient (id 2 and 4) showed an increased FA asymmetry index indicating a lowered potential as compared to the patients id (1 and 3). To evaluate efficacy, the inter group analysis could not be done as the data was less. The laterality index (LI) index of contralateral premotor area (BA 6) showed a marginal increase post therapy as compared to primary motor cortex (BA 4), suggesting the dominance of premotor area in facilitating neural plasticity. The in vivo MR spectroscopic data (single voxel and CSI) showed presence of lipid peaks and NAA in stem cell group patients showing increased neurogenesis, however detailed analysis could not be done due to poor quality of the spectra.

Table1. BOLD activation pattern and FA asymmetry index in pre, post-treatment in stroke patients

Pat. ID & Loc of MCA infarcts	Task	PRE TREATMENT										POST TREATMENT									
		BA 4		LI of Contr. BA 4	BA6		LI of Contr BA 6	Cerebellum		Ips CB ratio	FA asym m	BA4		LI of Contr. BA 4	BA6		LI of Contra. BA 6	Cerebellum		Ips CB ratio	FA asym m
		L	R		L	R		L	R			L	R		L	R		L	R		
1. RP	LWE	-	59	1	86	150	0.27	176	-	1	0.38	-	390	1	-	250	0.79	176	-	1	0.23
2. RFT	LWE	786	-	-1	1044	-	-1	-	-	-	0.79	-	-	-	230	-0.6	946	179	0.84	0.75	
3. RFP	LWE	60	112	0.30	150	343	0.39	-	-	-	0.22	-	1552	1	1685	0.49	1711	575	0.74	0.20	
4. RIC	LWE	-	970	1	20	229	0.83	844	605	0.47	0.45	1712	-	1	60	1712	0.93	-	-	-	0.36

RP: Right parietal; RFT: Right fronto temporal; RFP: Right frontoparietal, RIC: Right internal capsule, LI : Laterality index : C-I/C+I, FA asym m index =FA<sub>unaffected</sub> - FA<sub>affected</sub> / FA<sub>unaffected</sub>+ FA<sub>affected</sub>, Ips CB = Ips cerebellum , Ips CB ratio= ips CB/ Ips CB + Contr CB

**Discussion:** Intravenous ex vivo cultured mesenchymal stem cell transplantation is safe and feasible in chronic stroke patients. Intensive physiotherapy regime aids in stroke recovery and augments the principles of plasticity<sup>3-5</sup>. Benefit of stem therapies in stroke augmenting clinical and behavioral recovery has been demonstrated in this study. However, validation of efficacy of stem cell transplantation can be proven with more number of trials. The study is in progress with further recruitments.

## References:

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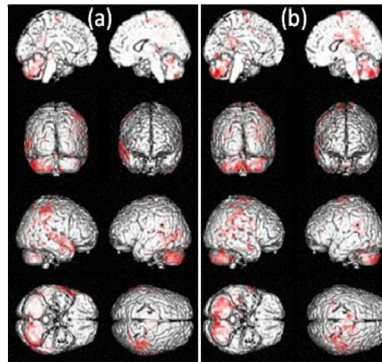


Figure 1. Brain activation pattern of a patient (id. 1) superimposed on rendered image during (a) pre stem cell therapy and (b) post stem cell therapy.