Neurorestoration in Amyotrophic Lateral Sclerosis - A case report

Hemangi Sane[1], Alok Sharma[2], Nandini Gokulchandran[2],Sarita Kalburgi[1,3], Amruta Paranjape[1,3], Prerna Badhe[4]

1 Department of Research & Development, NeuroGen Brain and Spine Institute, India

2 Department of regenerative medicine and medical Services, NeuroGen Brain and Spine Institute, India

3 Department of NeuroRehabilitation, NeuroGen Brain and Spine Institute, India

4 Department of regenerative laboratory services, NeuroGen Brain and Spine Institute, India

Corresponding author: Sarita Kalburgi NeuroGen Brain and Spine Institute, StemAsia Hospital & Research Centre, Sector 40, Plot No 19, Palm Beach Road, Seawood (W), New Mumbai - 400706. Email: sarita.neurogen@gmail.com

ABSTRACT

Amyotrophic Lateral Sclerosis (ALS) is a rapidly progressive neurodegenerative disorder. The primary pathology includes progressive loss of motor neurons in the brain and spinal cord. Cellular therapy has recently emerged as a promising treatment modality for patients with ALS. Autologous bone marrow mononuclear cell (BMMNCs) transplantation has been shown to be safe and feasible treatment option in various neurological disorders. Herein we present a detailed case report of a 40 year old female suffering from ALS since 3 years. She was given intrathecal autologous BMMNCs transplantation along with riluzole, lithium and intensive rehabilitation. We monitored the patient over a period of 17 months. Post therapy we observed that there was a slowing of the disease progression, and improvements in neurological symptoms. The outcome measures used were Amyotrophic Lateral Sclerosis Functional Rating (ALSFRS-r) Scale and Functional Independence Measure (FIM) scale. Research shows that ALSFRS-r score deteriorates about 17% every 6 months, but in this patient the ALSFRS-r score dropped only by 8% over 17 months after cell transplantation. This demonstrates that cellular transplantation is safe and has beneficial effects for slowing down the progression of ALS.

KEYWORDS: *Amyotrophic Lateral Sclerosis (ALS), Stem Cell therapy, Autologous Bone marrow derived mononuclear cells (BMMNCs), ALSFRS-r scale, FIM scale.*

Introduction

Amyotrophic lateral sclerosis (ALS) is muscle atrophy (amyotrophy) due to selective injury to peripheral motor neurons of the anterior horns of the spinal cord, lateral columns of the spinal cord, brainstem motor nucleus and cortical motor neurons. (1) This progressive disease results in functional consequences of widespread muscle atrophy and profound weakness. This leads to severe motor disability that affects speech, swallowing and respiratory insufficiency which ultimately causes death. (2,3) The incidence of ALS in different population ranges from one-two to four-six cases per 100,000 people per year. In Europe, the incidence rate was 2.08 and prevalence rate was 5.40. (4) Currently, about 25,000 patients with a mean age of 55 years are listed in the U.S. for ALS. (1) Median survival from onset to death in ALS is reported to vary from 2 to 4 years. (5)

The cellular processes that occurs after disease onset includes mitochondrial dysfunction, protein aggregation, generation of free radicals, excitotoxicity, inflammation and apoptosis, but the underlying cause remains unknown. (6) There is only one medication, riluzole, which has shown prolongation of survival for about 3 months in ALS patients. (7) Recent studies in stem cell biology have revealed new therapeutic options for ALS. It helps either by replacing diseased cell populations directly or by introducing a cell population that can be supportive to the motor

neurons affected by the disease process. (6, 8) Animal studies have shown beneficial effects of cell therapy in ALS. (9, 10, 11) Numerous studies with autologous bone marrow mononuclear cell (BMMNCs) transplantation have demonstrated the safety and efficacy in patients with other neurological disorders. (12, 13, 14) Due to its self-renewal and differentiation ability into different types of mature cells, stem cells may aid in modifying or arresting the deterioration of the disease process. (15-18)

We present a case of ALS which was treated with intrathecal administration of autologous BMMNC's. The rehabilitation therapy was combined with cell transplantation along with standard medical treatment of riluzole and lithium. Cell transplantation was done in this patient as her condition deteriorated despite the treatment with conventional medical therapies.

Case Report

A 40 year old female was diagnosed with ALS 3 years ago. The symptoms of pain and weakness began in both lower extremities which then progressed to upper extremities. She was unable to walk without support since 2 years. Her speech was affected since 1 year. She had complaints of dysarthria, easy fatigability, slowness in activities and difficulty in getting up from bed. Neurologically brisk deep tendon reflexes were present. She had fasciculations in bilateral lower extremities. There was hypertonia with grade 1 spasticity in bilateral upper and lower extremities. Her voluntary control of both upper and lower limbs was fair. She was walking with the help of walker and had foot drop. Functionally she was independent for all her activities of daily living. The Electromyography test showed evidence of an early neurogenic process in scattered muscles in multiple regions, suggestive of an early disorder of motor neurons or their axons. In the lower extremities there were additional features of upper motor neuron involvement. Her FIM score was 113/126 and ALSFRS-r scale score was 36/48.

Material and methodology

Therapy Selection of this patient for the treatment was based on World Medical Associations Revised Declaration of Helsinki. The ethical approval was obtained from Institutional committee for Stem Cell Research and (IC-SCRT) The patient was informed about the procedure and a duly filled informed consent form was obtained. Pre-procedure routine blood tests, urinalysis and chest x-ray were carried out to rule out active infection and assess fitness for anesthesia. (14) 300 mcg of Granulocyte colony-stimulating factor (G-CSF) injections were administrated 72 hours and 24 hours prior to BMMNC transplantation, to stimulate CD34+ cells and increase their survival and multiplication. A 100 ml volume of bone marrow was aspirated from the iliac bone. The density gradient separation method was used to obtain Mononuclear Cells (MNCs) The MNCs were then evaluated for CD34+ by FACS analysis and viable count was calculated and found to be about 98%. Approximately 4.6×108 MNCs were administered intrathecally immediately post separation at L4-L5 level using a lumbar puncture needle. 1 gm methyl prednisolone in 500 ml Ringer's Lactate (RL) was simultaneously injected intravenously to reduce local inflammation. (14)

To improve the effectiveness of the stem cell therapy patient was given exercise home program which included bed mobility exercises, active upper and lower extremity and trunk strengthening exercises, balance exercises and gait training. She was also given oromotor and facial exercises. Riluzole 50 mg once a day was continued. Lithium 300 mg once a day was started and the serum lithium levels were maintained between 0.5 to 0.8 mEq/L for six weeks. On the basis of improvements seen after five months a repeat dose of cell transplantation was given.

Result: (Table 1)

One week after first cell therapy her or motor functions was improved. She was experiencing less fatigue while speaking. There was improvement in tongue strength and speech clarity. Also there was spasticity reduction in all the extremities.

Five months after first cell therapy there was further improvement in her or motor functions. Her duration of speech was increased from 3 min to 15 min. She was able to vary voice pitch from soft to medium loud. Maximum Phonation Duration (MPD) was increased from 9 sec to 17

sec. The reaction time for chewing and pushing bolus towards pharynx was reduced. Tongue flexibility was better and she was able to say tongue twister clearly. There was improvement in lip control, blowing and sucking. The hand grip and forearm movements were better and stronger than before. Strength of interpose and extensor digitorum muscles had improved. There was reduction in clawing of bilateral hands. Improvements were noted in bed mobility, transfer and ambulation. She required less support to get up from sit to stand compared to before. There was improvement in sitting, standing static and dynamic balance. Walking balance had also improved. She maintained the muscle strength. Voluntary control of hip flexors improved from poor to fair. Her overall stamina had improved and she could work for 4-5 hrs/ 4-5 days. Frequency of falls was reduced from 2-3 per month to 1 per month.

Table 1: Results over the period of 17 months

	One week after stem cell therapy	**Five months after stem cell therapy**	**Seventeen months after stem cell therapy**
Oromotor functions	Less fatigue while speaking, improvement in tongue strength and speech clarity	Increased duration of speech from 3 min to 15 min, increased Maximum Phonation Duration (MPD) from 9 sec to 17 sec, reduced reaction time for chewing and pushing bolus towards pharynx, increased tongue flexibility and was able to say tongue twister, improved lip control, blowing and sucking.	Improved speech clarity and slurring of the speech was only at the end of the day.
Spasticity	Reduced in all four extremities	Same	Same
Hand Functions		Better hand grip and forearm movements, improved strength of interossei and extensor digitorum and reduction in bilateral clawing of hands.	Same
Bed Mobility, Transfers and Ambulation		Required less support to get up from sit to stand compare to before, improved sitting, standing and walking balance.	Same
Muscle strength and voluntary control		Maintained muscle strength. Improved voluntary control of hip flexors from poor to fair	Voluntary control maintained in shoulder, elbow, wrist, fingers, ankle and neck. Was able to perform activities much easily compare to before.
Stamina		Stamina improved and could work for 4-5 hrs/4-5 days.	Same
Frequency of falls		Reduced from 2-3 per month to 1 per month.	Before stem cell therapy it was 2-3 per month then 1per month after 5 months and later there was no fall after second dose of cellular transplantation

Seventeen months after first cell therapy i.e. 12 months after second dose of cell therapy. Her speech clarity was improved. There was slurring of the speech only at the end of the day. There was marked reduction in frequency of falls. Before cell therapy it was 2-3 per month then 1per month after 5 months and later there was no fall after second dose of cellular transplantation. Her voluntary control was maintained in shoulder, elbow, wrist, fingers, ankle and neck. She was able to perform activities much easily compared to before. FIM score was maintained throughout 17 months i. e. 113. ALSFRS-r score was maintained for 15 months after first dose of cellular transplantation then it decreased from 36 to 33. (Table 2)

Table 2: Changes in outcome measures over the period of 17 months

Outcome measures	At assessment before 1st transplantation	At 5 months after 1st transplantation (Just before 2nd dose)	At 17 months after 1st transplantation (12 months after 2nd dose)
ALSFRS-r	36	36	33
FIM	113	113	113

Discussion

Amyotrophic Lateral Sclerosis (ALS) is a neurodegenerative disorder, characterized by progressive axonal degeneration of motor neurons in the spinal cord and motor cortex with the sparing of sensory system. Recent advances have suggested involvement of non neuronal cells like glial cells. (19) Up regulation of astrocytic glutamate, superoxide dismutase leads to unchecked intracellular peroxidation reactions and subsequent oxidative stress. Excitotoxicity, autoimmunity and widespread neuroinflammatory response are also implicated. Recently various genes have been found to be involved. (20) Pathophysiology of ALS is heterogeneous and therefore makes it more complex disease to manage. The current management strategies have demonstrated very limited effect on the survival of patients. Disease-modifying options for patients with ALS are limited to treatment with riluzole, noninvasive positive pressure ventilation, and nutritional support, each having only a modest effect on disease progression and survival. There is growing interest in the use of cell therapy in ALS. (6) which replace the lost cells, provide neurotrophic support, and improve the diseased microenvironment.(6,8,21)

Clinical trials have revealed feasibility and safety of the bone marrow stem cells in various medical conditions. (3) The mechanism of action of BMMNCs is twofold; to protect the existing motor neurons and to replace the degenerated motor neurons in ALS. Autologous BMMNCs have neurogenic potential but the functionality of regenerated neurons is debatable in the highly toxic cellular microenvironment observed in patients with ALS.(22) Use of adult stem cells for therapy avoids ethical problems and can be isolated from the patients. This overcomes the problem of immunological rejection and reduces the risk of tumor formation compared to the use of embryonic stem cells. (23) Intrathecal delivery was aimed to increase the number of neuromuscular connections, decrease pro-inflammatory cytokines in the brain and spinal cord, decrease microglia and astroglia density and promote migration and differentiation of endogenous precursors.(24, 25)

In our patient there was no significant decline in function or acceleration of disease progression following cellular transplantation and neurorehabilitation. In fact the intensive and repeated monitoring of patient showed consistent positive changes. Improvements were noted in oromotor functions, voluntary control, ambulation, hand functions and stamina. The foremost importance of neurorehabilitation is to assist in movement restoration, to increase muscle strength, endurance, co-ordination and balance, to reduce spasticity and increase joint range of motion and prevent contractures. Various studies have shown that exercises enhance the effect of local stem cells by helping their mobilization and angiogenesis.(27) Hence the concept of combination of neuroregenration and rehabilitation have better therapy outcome.

A systematic review of literature including a total of 1100 patients treated with Lithium showed no statistically significant differences in the rates of functional decline, deterioration of respiratory function or survival time since onset of the disease as compared to patients treated with Riluzole or placebo.(28) The rationale for prescribing Lithium to our patient was to enhance the survival and potency of transplanted cells. (29) The survival probability declines steadily over a year in patients with ALS (30) but (as shown in graph 1), we observed that this patient have maintained her condition for 15 months. Even at 17 moths follow up there was minimal functional decline in her condition.

Her FIM score was 113 for 17 months and ALSFRS-r score was 36 for 15 months after the first cellular transplantation. After 17 months ALSFRS-r score was decreased from 36 to 33. The components in which there was reduction were handwriting, Cutting food and handling, and adjusting bed cloths (Table 3). Previous studies have reported decline of ALS-FRS r score to be a prognostic indicator of the disease. Various epidemiological studies have reported different rates of decline of ALS-FRS score. De Carvahelo et al 2005 (29) reported 17% decline of ALS-FRS scores every 6 months but in this study our patient have dropped by only 8 % (3 points) and maintained her condition over the period of 17 months (table 4 and graph) In another study by Prabhakar S, Marwaha N et al 10 ALS patients were treated with autologous BMMNCs intrathecally and on follow up of one year ALSFRS-r score was reduced by four points. (30) This indicates slowing down of progression of ALS after autologous BMMNC's transplantation.

Table 3:- ALSFRS-r scale score changes over 17 months

ALS-FRSr category		Score at the time of admission	Score after 5 months of transplan-tation	Score after 17 months of transplan-tation
1. Speech	4 - Normal Speech processes 3 - Detectable speech with disturbances 2 - Intelligible with repeating 1 - Speech combined with nonvocal communication 0 - Loss of useful speech	3	3	3
2. Salivation	4 - Normal 3 - Slight but definite excess of saliva in mouth; may have nighttime drooling 2 - Moderately excessive saliva; may have minimal drooling 1 - Marked excess of saliva with some drooling 0 - Marked drooling; requires constant tissue or handkerchief	4	4	4
3.Swallowing	4 - Normal eating habits 3 - Early eating problems - occasional choking 2 - Dietary consistency changes 1 - Needs supplemental tube feeding 0 - NPO (exclusively parenteral or enteral feeding)	4	4	4

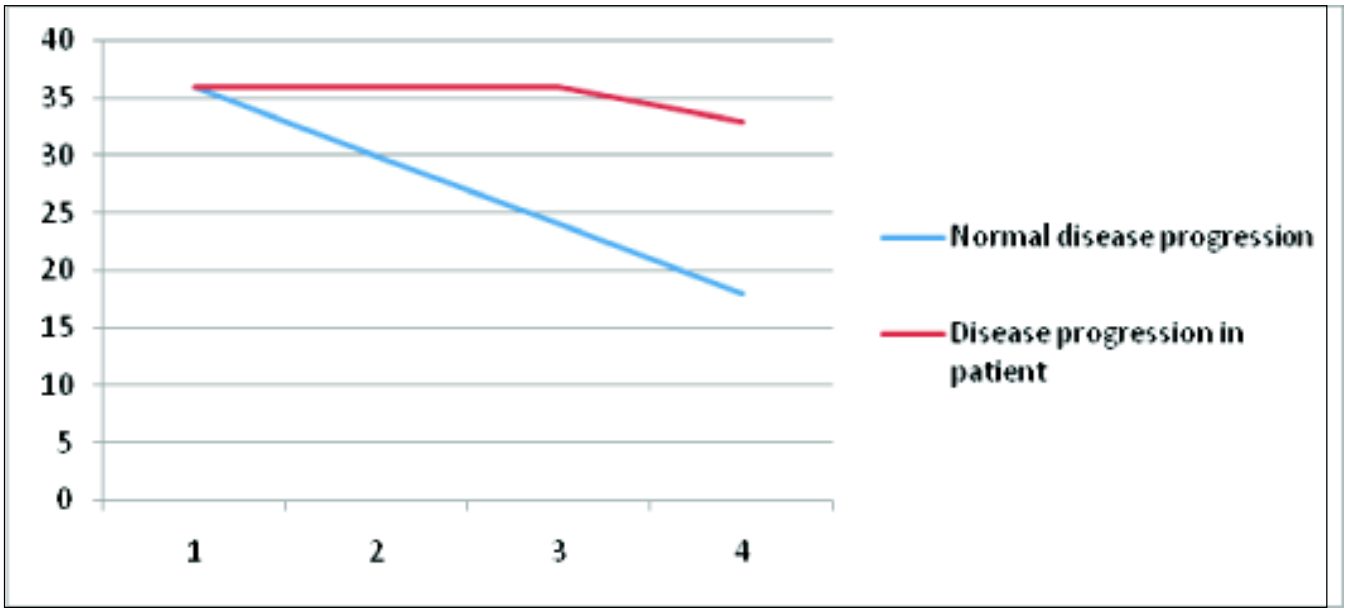
ALS-FRSr category		Score at the time of admission	Score after 5 months of transplan-tation	Score after 17 months of transplan-tation
4. Hand-writing	4 - Normal 3 - Slow or sloppy; all words are legible 2 - Not all words are legible 1 - Able to grip pen but unable to write 0 - Unable to grip pen	1	1	0
5. Does subject have gastrostomy? No - Answer 5a Yes - Answer 5b a. Cutting Food and Handling Utensils (patients without gastrostomy)	4 - Normal 3 - Somewhat slow and clumsy, but no help needed 2 - Can cut most foods, although clumsy and slow; some help needed 1 - Food must be cut by someone, but can still feed slowly 0 - Needs to be fed	4	4	3
b Cutting Food and Handling Utensils (alternate scale for patients with gastrostomy)	4 - Normal 3 - Clumsy but able to perform all manipulations independently 2 - Some help needed with closures and fasteners 1 - Provides minimal assistance to caregivers 0 - Unable to perform any aspect of task	NA	NA	NA
6. Dressing and Hygiene	4 - Normal function 3 - Independent and complete self-care with effort or decreased efficiency 2 - Intermittent assistance or substitute methods 1 - Needs attendant for self-care 0 - Total dependence	2	2	2
7.Turning in bed and adjusting bed clothes	4 - Normal 3 - Somewhat slow and clumsy, but no help needed 2 - Can turn alone or adjust sheets, but with great difficulty 1 - Can initiate, but not turn or adjust sheets alone 0 - Helpless	3	3	2

ALS-FRSr category		Score at the time of admission	Score after 5 months of transplantation	Score after 17 months of transplantation
8. Walking	4 - Normal 3 - Early ambulation difficulties 2 - Walks with assistance 1 - Nonambulatory functional movement only 0 - No purposeful leg movement	2	2	2
9. Climbing Stairs	4 - Normal 3 - Slow 2 - Mild unsteadiness or fatigue 1 - Needs assistance 0 - Cannot do	1	1	1
R-1- Dyspnea	4 - None 3 - Occurs when walking 2 - Occurs with one or more of the following: eating, bathing, dressing 1 - Occurs at rest, difficulty breathing when either sitting or lying 0 - Significant difficulty, considering using mechanical respiratory support	4	4	4
R-2 Orthopnea	4 - None 3 - Some difficulty sleeping at night due to shortness of breath, does not routinely use more than two pillows 2 - Needs extra pillow in order to sleep (more than two) 1 - Can only sleep sitting up 0 - Unable to sleep	4	4	4
R-3 Respiratory Insufficiency	4 - None 3 - Intermittent use of NIPPV 2 - Continuous use of NIPPV during the night 1 - Continuous use of NIPPV during the night and day 0 - Invasive mechanical ventilation by intubation or tracheostomy	4	4	4

Table 4 Normal disease progression and in this patient

Months	Normal disease progression	Disease progression in patient
0	36	36
5	30	36
15	24	36
17	18	33

Graph: Normal Disease progression in ALS and in patient



Also a retrospective controlled study has shown statistically significant increased survival duration by 30.38 months in ALS patients who underwent autologous BMMNCs transplantation as compared to the control group. The survival duration in this controlled study was also higher than previous epidemiological studies. (20)

There were no adverse events noted in our patient. The findings of this case point towards the safety and beneficial effects of cell transplantation for slowing down the progression of ALS. Further larger clinical studies are required for conclusive results.

Conclusion

This case report demonstrates that autologous BMMNC intrathecal transplantation has slowdown the progression of disease and maintained functional status for a period of 17 months. So it has great potential as a novel therapeutic modality. It may have a positive effect on the duration of survival in ALS patients, when used in conjunction with riluzole and neurorehabilitation.

Conflict of interest statement

The author declares that there is no conflict of interest regarding the publication of this article.

Reference:

1. Valetdinova K R, Medvedev S P et al. Systems of Motor Neuron Diseases As a platform for Studying Pathogenic Mechanisms and Searching for Therapeutic Agents. Acta Naturae, 2015; Vol. 7 No. 1:24.
2. Edward J K, Marta S M et al. Estimating daily energy expenditure in individuals with amyotrophic lateral sclerosis. Am J Clin Nutr, 2014; 99:792-803.
3. Vincent M, Pierre-François P et al. Safety, Pharmacokinetic, and Functional Effects of the Nogo-A Monoclonal Antibody in Amyotrophic Lateral Sclerosis: A Randomized, First-In-Human Clinical Trial. Plos One, 2014; 9:5.
4. Chiòa A, Logroscinob G et al. Global Epidemiology of Amyotrophic Lateral Sclerosis: a Systematic Review of the Published Literature. Neuroepidemiolog,. 2013; 41(2): 118-130.
5. Gordon P H, Amyotrophic lateral sclerosis: pathophysiology, diagnosis and management. CNS Drugs, Jan 2011; 25(1):1-15.
6. Jonathan D. Glass, Nicholas M. Boulis et al. Lumbar Intraspinal Injection of Neural Stem Cells in Patients with Amyotrophic Lateral Sclerosis: Results of a Phase I Trial in 12 Patients. STEM CELLS, 2012; 30:1144-1151.
7. Traynor BJ, Alexander M et al. An outcome study of riluzole in amyotrophic lateral sclerosis-a population based study in Ireland, 1996-2000. J Neurol, 2003;250(4):473-9.
8. Amyotrophic lateral sclerosis: pathophysiology, diagnosis and management. CNS Drugs, 2011; 25(1):1-15.
9. Rizvanov AA, Guseva DS et al. Genetically modified human umbilical cord blood cells expressing vascular endothelial growth factor and fibroblast growth factor two differentiate into glial cells after

transplantation into amyotrophic lateral sclerosis transgenic mice. Exp Biol Med, 2011; 236: 91-8.

10. Garbuzova-Davis S, Rodrigues MC et al. Multiple intravenous administrations of human umbilical cord blood cells benefit in a mouse model of ALS. Plos One, 2012; 7.
11. Garbuzova-Davis S, Willing AE et al. Intravenous administration of human umbilical cord blood cells in a mouse model of amyotrophic lateral sclerosis: distribution, migration, and differentiation. J Hematother Stem Cell Res, 2003; 12: 255-70.
12. Sharma A, Sane H, Paranjape A, Gokulchandran N, Biju H et al. Cellular transplantation may modulate disease progression in spinocerebellar ataxia-A case report. Indian Journal of Medical Research and Pharmaceutical Sciences, 2014; 1(3)
13. Sharma A, Sane H, Paranjape A, Gokulchandran N, Nagrajan A et al. Autologous Bone Marrow Mononuclear Cells Intrathecal Transplantation in chronic stroke. Hidawi Publishing Corporation, Stroke research and treatment, 2014.
14. Yoon H, Shim S, Park YH, Chung JK, Nam JH et al. Complete Spinal Cord Injury Treatment Using Autologous Bone Marrow Transplantation and Bone Marrow Stimulation with Granulocyte Macrophage-Colony Stimulating Factor: Phase I/II Clinical Trial. Tissue Engineering, 2005; 11 (5-6):913-922.
15. Sharma A, Gokulchandran N et al. Administration of autologous bone marrow-derived mononuclear cells in children with incurable neurological disorders and injury is safe and improves their quality of life. Cell Transplant, 2012; 21 Suppl 1:S79-90.
16. Sharma A, Gokulchandran N et al. Autologous bone marrow mononuclear cell therapy for autism: an open label proof of concept study. Stem Cells Int, 2013.
17. Kim SU, de Vellis J. Stem cell-based cell therapy in neurological diseases: a review. J Neurosci Res, 2009; 87: 2183-200.
18. Sharma A, Gokulchandran N, Sane H, Nagrajan A, Paranjape A et al. Detailed analysis of the clinical effects of cell therapy for thoracolumbar spinal cord injury: an original study. Journal of Neurorestoratology, 2013; 1: 13-22.
19. Al-Chalabi A, Jones A et al. The genetics and neuropathology of amyotrophic lateral sclerosis. Acta Neuropathol, 2012; 124: 339-52.
20. Sharma A, Sane H, Paranjape A, Gokulchandran N, Nagrajan A et al. The effect of autologous bone marrow mononuclear cell transplantation on the survival duration in Amyotrophic Lateral Sclerosis - a retrospective controlled study. Am J Stem Cells, 2015;4(1):50-65
21. Feldman EL, Boulis NM, Hur J, Johe K, Rutkove SB et al. Intraspinal Neural Stem Cell Transplantation in Amyotrophic Lateral Sclerosis: Phase 1 Trial Outcomes. Ann Neurol, 2014;75:363-373
22. Thonhoff JR, Ojeda L, Wu P et al. Stem Cell-Derived Motor Neurons: Applications and Challenges in Amyotrophic Lateral Sclerosis. Curr Stem Cell Res The, 2009; 4(3):178-99.
23. Glover LE, Tajiri N, Weinbren NL, Ishikawa H, Shinozuka K et al. A Step-up Approach for Cell Therapy in Stroke: Translational Hurdles of Bone Marrow-Derived StemCells. Translational Stroke Research, 2012; 3: 90-98.
24. Mazzini L, Vercelli A, Ferrero I, Mareschi K, Boido M et al. Stem Cell in Amyotrophic Lateral Sclerosis: State of art. Informa Healthcare, 2009; 9(10):1245-1258.
25. Sharma A, Badhe P et al. Autologous Bone Marrow Derived Stem Cells For Motor Neuron Disease with Anterior Horn Cell Involvement. Bombay Hospital Journal, 2011; Vol.53:No.1.
26. Gamez J, Salvado M, Martínez de la Ossa A, Badia M. Lithium for treatment of amyotrophic lateral sclerosis: much ado about nothing. Neurologia, 2013; 13 :(21-2)
27. Richman CM, Kinnealey A, Hoffman PC. Granulopoietic effects of lithium on human bone marrow in vitro. Exp Hematol, 1981;9(4):449-55
28. Mandrioli J, Pietro Faglioni et al. Amyotrophic lateral sclerosis: Prognostic indicators of survival. Amyotrophic Lateral Sclerosis, 2006; 7: 217-226.
29. De Carvalho M, Costa J, Swash M. Clinical trials in ALS: a review of the role of clinical and neurophysiological measurements. Amyotroph Lateral Scler Other Motor Neuron Disord. 2005;6:202-12.
30. Prabhakar S, Marwaha N, Lal V, Sharma RR, Rajan R et al. Autologous bone marrow-derived stem cells in amyotrophic lateral sclerosis: a pilot study. Neurol India, 2012; 60: 465-9.