REVIEW ARTICLE

The promise of stem cells in amyotrophic lateral sclerosis: a review of clinical trials

Muhammad Usman Khalid, Taleaa Masroor

Abstract

Amyotrophic Lateral Sclerosis (ALS) is a neurodegenerative condition with high cost of care, poor treatment outcomes, and a significant decrease in quality of life, eventually culminating in high mortality rates. Stem cells present an attractive alternative to conventional therapies as they can regenerate tissue and introduce growth factors to slow down the progression of disease. We conducted a comprehensive review of literature available in the MEDLINE (PUBMED), Scopus, and Cochrane Library databases, of current usage of stem cells and stem cell-based biomaterials for ALS treatment. Clinical trials, less than 10 years old, on human subjects were included in the study. Overall, stem cells, whether mesenchymal, non-lineage, or neural stem cells all seem safe for use in therapy for ALS. However, due to the chronic nature of the disease the efficacy of the treatment is not proven and warrants further investigation.

Keywords: Amyotrophic Lateral Sclerosis. Biocompatible Materials, Neural, Stem Cells.

DOI: 10.47391/JPMA.AKUS-22

Introduction

Neurodegenerative diseases such as Amyotrophic Lateral Sclerosis (ALS), Alzheimer's, Parkinson's disease are debilitating conditions characterized by function and structural loss in neuronal tissues[1]. While treatment exists to slow down the rate of degeneration, it is often costly, with poor efficacy and significant side effects[2]. A major barrier to arresting the cell death and pathophysiological processes is the lack of regenerative capacity in the nervous system [3]. In the central nervous system (CNS) the blood brain barrier (BBB) is a major obstacle in the delivery of drugs that could potentially help slow down the disease process or potentially cure it[4,5]. Overall, neurodegenerative diseases lead to a loss of quality of life, increase the cost of care, and can spell a painful decline, particularly in resource limited areas where they are traditionally given lower priority [6].

Department of Surgery, Aga Khan University Hospital, Karachi, Pakistan.
Correspondence: Muhammad Usman Khalid . Email: uklive@hotmail.com

Amyotrophic lateral sclerosis (ALS) is an adult-onset, neurodegenerative condition that is characterized by progressive loss of upper and lower motor neurons[7]. Once the disease spreads to the diaphragm, it can cause severe respiratory distress, inevitably leading to death. Ninety percent of ALS patients are sporadically affected. Hereditary ALS cases have been linked to mutations in several different genes, with SOD1, FUS, and TARDBP mutations being the most frequent[8]. However, apart from a mean earlier onset, there is no difference between familial and sporadic ALS, with respect to disease severity and symptoms. Several theories have been proposed for the peculiar progression of the disease. It is speculated that like cancer, amyotrophic lateral sclerosis may involve a multi-process complex interaction between affected motor cells and the neuronal microenvironment[9]. The pathogenesis of ALS has also been linked to glutamate neurotoxicity. The only effective disease-modifying treatment for ALS that successfully slows disease progression and increases median patient survival by 3-6 months is the anti-glutamatergic drug Riluzole. However, even with its use symptoms persist and patients may develop non-cognitive impairments[10].

Due to the elusiveness of the cause of neuronal death and the difficulty in making an early diagnosis in ALS, there are currently few effective therapeutic agents available. The neuroprotective and differentiation potential of stem cells makes them a possible therapeutic choice for interrupting the progression of disease[11]. We reviewed 19 clinical trials investigating approaches to stem cell therapy, type of stem cells used, safety and efficacy of the routes of administration and safety of multiple dosing.

Methods

We conducted a review of the available literature on clinical trials in stem cell therapy for the neurodegenerative disease. A bibliographic search was conducted through three major electronic databases. The MEDLINE (PUBMED), the Cochrane Library, and Scopus were the databases used with Medical Subject Headings (MeSH) terms (((neurodegenerative disease) OR (ALS) OR (Amyotrophic Lateral Sclerosis)) AND ((stem cells) OR (mesenchymal cells) OR (pluripotent stem cells) OR

Table-1: Summary of literature with pertinent findings

Date	Authors	Country	Salient findings
2019	Berry JD et al.[16]	United States	In a prespecified rapid progressor subgroup, rate of disease progression was improved at early time points ($p < 0.05$). Rapid progressors also showed significant improvement at 4 and 12 weeks ($p = 0.004$ and 0.046, respectively). CSF neutrophic factors and inflammatory biomarkers significantly decreased in treat participants ($p<0.05$).
2016	Glass JD et al.[31]	United States	Minimal adverse events and similar outcomes show stem cells to be safe for use in human populations for neurodegenerative diseases.
2016	Petrou P et al.[15]	United States	Treatment was safe and well tolerated in all participants with only mild transient side-effects. The rate of progression of forced vital capacity and increase in ALSFSRS-R scale were both significant in treated patients.
2022	Cudkowicz ME et al.[18]	United States	A pre-specified analysis of participants with baseline ALSFRS-R ≥ 35 ($n = 58$) showed a clinical response rate at 28 wk of 35% MSC-NTF and 16% placebo (OR = 2.6, P = .29). Significant improvements in cerebrospinal biomarkers of neuroinflammation, neurodegeneration, and neurotrophic factor support were observed with MSC-NTF, with placebo unchanged.
2020	Barczewska M et al. [17]	Poland	The female sex and a good therapeutic response to the first administration are significant predictors of efficacy following wharton's jelly-derived mesenchymal stem cells therapy.
2020	Siwek T et al. [11]	Poland	In patients who had ALS with an inherently rapid course, slowing of the disease was noted following treatment with MSCs. However a small sample size precluded statistical significance.
2018	Sobus A et al.[27]	Poland	C3 inflammatory proteins were significacntly lower in the treatment group, and miRNA expression levels were also different. miR-206 increased while miR-378 decreased.
2021	Petrou P et al. [15]	Singapore	Repeated intrathecal injections of autologous MSC was safe in patients with ALS and provide indications of medium-term clinical benefits that were related to the intervals between the administrations of the cells.
2014	Kim HY et al.[34]	England	VEGF, ANG, and TGF-β levels in MSCs could be used as potential biological markers to predict the effectiveness of autologous MSC therapy and to identify those patients who could optimally benefit from MSC treatment.
2014	Feldman EL et al.[14]	United States	The cervical injection procedure was well tolerated and disease progression did not accelerate in any subject, verifying the safety and feasibility of cervical and dual-targeting approaches.
2019	Mazzini L et al.[32]	Italy	Results show that transplantation of hNSC is a safe procedure that causes no major deleterious effects over the short or long term.
2015	Mazzini L et al. [33]	Italy	Two patients showed a transitory improvement of the subscore ambulation on the ALS-FRS-R scale (from 1 to 2). A third patient showed improvement of the MRC score for tibialis anterior, which persisted for as long as 7 months.
2017	Syková E et al.[21]	Czech Republic	Intrathecal application of BM-MSCs in ALS patients is a safe procedure and it can slow down progression of the disease.
2016	Staff NP et al.[24]	United States	The clinical findings were associated with elevated CSF protein and nucleated cells with MRI of thickened lumbosacral nerve roots. Longitudinal ALSFRS-R questionnaires confirmed continued progression of disease in all treated patients.
2022	Mohseni R et al.[23]	Italy	All five patients in the intervention group showed significant improvement in the motor amplitude response of the tibial nerve (0.56mV; p: 0.029).
2015	Rushkevich YN et al.[10]	United States	Evaluation of the results of cell therapy after 12-month follow-up revealed slowing down of the disease progression in 10 patients in comparison with the control group consisting of 15 patients.
2014	Riley J et al.[28]	United States	Delivery of a cellular payload to the cervical or thoracolumbar spinal cord was well tolerated by the spinal cord in this vulnerable population.
2015	Oh KW et al. [34]	Republic of Korea	No serious adverse events were observed during the 12-month follow-up period.Decline in the ALSFRS-R score was not accelerated during the 6-month follow-up period.
2016	Ruiz-Lopez FJ et al.[19]	Netherlands	The spinal injection of of autologous bone marrow mononuclear cells in patients with amyotrophic lateral sclerosis is safe and it is possible in ALS patients without worsening the disease.

(IPSCs))). Most studies in the search originated from high income countries (HICs). The inclusion criteria were that studies needed to be less than 10 years old, clinical trials or outcome assessments, and on human populations. A total of 157 studies were initially screened from the databases on August 21st 2022 and imported into Rayyan software[12]. Duplicates were removed, studies were sorted by type, and 56 clinical trials were selected. From these

title and abstract screening for relevance was done and 20 articles were selected for the final full text review which was completed by August 30 2022. These are summated in Table 1.

Discussion

Stem cells can either be derived from an embryonic source (pluripotent stem cells), or from adult cell-lineage precursors i.e., haematopoietic stem cells, neural stem cells and olfactory ensheathing stem cells. Apart from the ability for neurodifferentiation, stem cells can secrete neurotrophic factors (NTFs) encased within exosomes that carry them past the blood brain barrier (BBB)[13]. Mesenchymal stem cells (MSCs) are multipotent progenitor cells that can differentiate into neural cells and neuroglia[14]. They have shown great therapeutic promise in the treatment of neurodegenerative diseases. The safety and efficacy of MSCs for the treatment of ALS has been established through several phase 1/ 2 trials conducted in the past decade[15,16].

Safety and Efficacy of Bone-marrow MSC transplantation

Berry et al. conducted a phase 2 clinical trial where 48 ALS patients were randomized to receive either placebo or intramuscular and intrathecal doses of MSC-NTF cells[17]. No serious adverse events attributable to the intervention were noted. However, there was no difference in ALS progression within the two groups, except in a subgroup of rapid progressors, who showed significant improvement after 4 and 12 weeks of treatment ($p< 0.05$). To investigate the impact on survival time and ALS progression Barczewska et al. developed an approach comparing the impact of administering three doses of 30 x 10^6 MSCs derived from Wharton's jelly on a two-monthly basis to a cohort of 134 patients[18]. There were 67 patients in the control and in the unexposed group. It showed a two-fold increase in survival time but no difference in ALS progression on the revised Amyotrophic Lateral Sclerosis Functional Rating Scale (ALSFRS-R)[19]. The intervention was deemed safe as no serious drug reactions were noted. The study also found that female gender and good therapeutic response to first dose of treatment were predictors of treatment efficacy. Similar safety profile and improvements were recorded in a phase 3 trial conducted by Cudkowicz et al, in which clinical response rates at 28 weeks were 35% MSC-NTF and 16% placebo[20]. They had pre-specified analysis of participants with baseline ALSFRS-R 35 ($n = 58$) ($OR = 2.6$, $P = .29$). Concerns regarding the viability and safety of infusing autologous bone-marrow mononuclear cells (ABMNC) via an intraspinal intramedullary injection, were investigated by Ruiz-Lopez et al. in an open phase 1 clinical trial[21]. The implementation was successful with no serious adverse events or changes in forced vital capacity compared to pre-intervention levels.

To compare efficacy of an intrathecal versus intramuscular approach to administration of bone marrow harvested MSCs, a phase 2a trial was conducted by Petrou et al.[22]. MSC-NTF cells were injected into the biceps and triceps at 24 different sites (1x 10^6 cells/site) or administered intravenously (1x 10^6/kg) during phase 1/2. In the second stage (phase 2a) of the study, three dosing cohorts of patients received both IT and IM treatment. Only the groups treated with IT showed systemic benefit with an improvement in the mean monthly rate of progression of the ALS-FRS-R score and FVC (from 1.56 to 0.28 and from 3.5% to 2.3%, respectively in phase 1/2; and from 1.4 to 0.6 and 2.6% to 0.86%, respectively in phase 2a). It is possible that a combined approach may exert a synergistic effect and improve outcome[23].

Mouse models of ALS show that systemic or intraspinal dose of adipose-derived mesenchymal stem cell (MSC) transplantation slow motor neuron degeneration and enhance motor function[24]. Furthermore, harvesting MSCs via open biopsy of adipose tissue is simpler to execute compared to harvesting from bone marrow, however literature is unclear on efficacy of adipose derived MSCs[25]. A phase 1 trial aimed at determining impact of escalating doses of intrathecal autologous adipose-derived mesenchymal stromal cells showed that it had a good safety profile[26]. Participants were able to tolerate an intrathecal dose of 1 x 10^8 cells. However, there was no impact on ALS progression on ALSFRS-R scale and dose-dependent association with pain was noted. The most frequently reported side effects were transient low back and radicular leg pain at the highest dose level. These findings were linked to possible neuroinflammation, as there was a documented rise in nucleated cells and levels of CSF protein, with thickening of lumbosacral nerve roots on MRI.

CSF biomarkers and MSC transplantation

CSF biomarkers can help track treatment response at the level of the neural microenvironment. Vascular endothelial growth factor has been linked to neuroprotection in motor neurons, and reduced levels have been correlated with neurodegeneration[27]. Trials investigating VEGF levels show that patients receiving MSC-NTF reported significantly higher levels of VEGF expression compared to placebo[20]. They also saw significant reduction in cerebrospinal biomarkers of

neuroinflammation, neurodegeneration, and support for neurotrophic factor. Berry et al. also noted similar findings with a significant increase in neurotrophic factors such as Leukaemia inhibitory factor and Basal hepatocyte growth factor post MSC transplantation[17].

Lin-Cells

Lin cells or lineage-negative cells are a heterogenous group comprising of precursor, progenitor, and stem cells that lack mature morphotic blood components. They have demonstrated ability for self-renewal and paracrine release of different angiopoietic and neurotrophic factors. Paczkowska et al. showed that NTFs secreted by Lin cells increased expression of Ki-67 (antigen marker of cells in the active phases of the cell cycle) and anti-apoptotic Bcl-2 gene in a culture of serum-free SH-SY5Y line of human neuronal cells[28]. The safety of their use in ALS patients was established by a clinical trial conducted in Poland on 12 ALS patients that showed no transient or chronic adverse events related to intrathecal administration[29]. The trial simultaneously assessed the levels of neurotrophic factors and CSF inflammatory biomarkers after a single intrathecal dose of bone marrow derived Lin-cells and showed no sustained change in the CSF levels of BDNF and NGF at one-month follow-up. Reasons for the recorded findings could be a low baseline expression of BDNF in ALS patients with an undetectable rise post-transplantation. Furthermore, the study underlines the challenges of transplanting and monitoring the therapeutic potential of NTFs. Short half-life and inability to cross the blood-brain barrier (BBB), combined with an incompletely understood mechanism whereby they may exert effects by increasing expression of NTFs locally, makes them a challenging entity to assess[30].

Neural Stem Cells

Neural stem cells or human spinal cord-derived stem cells (HSSCs) are progenitor cells that are isolated from an 8-week-old foetus's cervicothoracic spinal cord[31]. Animal studies have shown that injection of HSSCs in rats with ischaemic spinal cord injury caused survival of endogenous motor neurons and partial motor neuron recovery, prompting further inquiry into the novel approach[32]. Glass et al conducted a phase 1 clinical trial using HSSCs in 12 ALS patients, which showed promising outcomes[33]. The surgical intervention did not exacerbate disease presentation or cause treatment-associated morbidity. Mazzini et al. evaluated the feasibility of micro-transplanting hNSCs into the gray matter tracts of the lumbar or cervical spinal cord of 18 patients with spinal onset ALS[34]. The study utilized clinical grade hNSC lines that were isolated from miscarried foetus brain biopsies and reproducibly and steadily expanded ex vivo. The ALS FRS R, Ashworth Spasticity Scale, Medical Research Council (MRC) scale of 34 muscle groups in the upper and lower limbs, and FVC were used to assess clinical assessment and rate of disease progression[19]. Pre- and post-transplantation assessment was done based on clinical, psychological, neuroradiological, and neurophysiological data. None of the patients experienced severe side effects or accelerated disease progression for up to 60 months following surgery. The utility of Mazzini et al.'s findings lies in the description of a safe and uniform brain cell drug product that is readily available, can be used for larger and more uniform trials, and may even make it possible to implement global, multicentric clinical studies where patients receive the same care at every location[37]. Although the data for safety and efficacy looks promising, the small sample sizes make it difficult for any conclusions with regards to impact on disease progression to be drawn[38].

Conclusion

Stem cells are a safe option for treatment of ALS and has shown a favourable side effect profile. It has shown improvement in the overall survival, although no significant improvement in rate of degeneration has been reported in these early phase trials. Late phase trials and longitudinal outcome studies are needed to investigate the efficacy of stem cell treatment for ALS.

Disclaimer: None to declare.

Conflict of Interest: None to declare.

Funding Disclosure: None to declare.

References

1. Sivandzade F, Cucullo L. Regenerative Stem Cell Therapy for Neurodegenerative Diseases: An Overview. Int J Mol Sci 2021;22:2153. doi: 10.3390/ijms22042153.
2. De Gioia R, Biella F, Citterio G, Rizzo F, Abati E, Nizzardo M, et al. Neural Stem Cell Transplantation for Neurodegenerative Diseases. Int J Mol Sci 2020;21:3103. doi: 10.3390/ijms21093103.
3. Chang CY, Ting HC, Liu CA, Su HL, Chiou TW, Lin SZ, et al. Induced Pluripotent Stem Cell (iPSC)-Based Neurodegenerative Disease Models for Phenotype Recapitulation and Drug Screening. Molecules 2020;25:2000. doi: 10.3390/molecules25082000.
4. Tam VH, Sosa C, Liu R, Yao N, Priestley RD. Nanomedicine as a non-invasive strategy for drug delivery across the blood brain barrier. Int J Pharm 2016;515:331-42. doi: 10.1016/j.ijpharm.2016.10.031.
5. Wen MM, El-Salamouni NS, El-Refaie WM, Hazzah HA, Ali MM, Tosi G, et al. Nanotechnology-based drug delivery systems for Alzheimer's disease management: Technical, industrial, and clinical challenges. J Control Release 2017;245:95-107. doi: 10.1016/j.jconrel.2016.11.025.
6. Lunn JS, Sakowski SA, Hur J, Feldman EL. Stem cell technology for neurodegenerative diseases. Ann Neurol 2011;70:353-61. doi:

10.1002/ana.22487.
7. Brotman RG, Moreno-Escobar MC, Joseph J, Pawar G. Amyotrophic Lateral Sclerosis. Treasure Island, FL: StatPearls Publishing; 2022.
8. Andersen PM, Al-Chalabi A. Clinical genetics of amyotrophic lateral sclerosis: what do we really know? Nat Rev Neurol 2011;7:603-15. doi: 10.1038/nrneurol.2011.150.
9. Al-Chalabi A, Calvo A, Chio A, Colville S, Ellis CM, Hardiman O, et al. Analysis of amyotrophic lateral sclerosis as a multistep process: a population-based modelling study. Lancet Neurol 2014;13:1108-13. doi: 10.1016/S1474-4422(14)70219-4.
10. Rushkevich YN, Kosmacheva SM, Zabrodets GV, Ignatenko SI, Goncharova NV, Severin IN, et al. The Use of Autologous Mesenchymal Stem Cells for Cell Therapy of Patients with Amyotrophic Lateral Sclerosis in Belarus. Bull Exp Biol Med 2015;159:576-81. doi: 10.1007/s10517-015-3017-3.
11. Siwek T, Jezierska-Woźniak K, Maksymowicz S, Barczewska M, Sowa M, Badowska W, et al. Repeat Administration of Bone Marrow-Derived Mesenchymal Stem Cells for Treatment of Amyotrophic Lateral Sclerosis. Med Sci Monit 2020;26:e927484. doi: 10.12659/MSM.927484.
12. Wang J, Hu WW, Jiang Z, Feng MJ. Advances in treatment of neurodegenerative diseases: Perspectives for combination of stem cells with neurotrophic factors. World J Stem Cells 2020;12:323-38. doi: 10.4252/wjsc.v12.i5.323.
13. George S, Hamblin MR, Abrahamse H. Differentiation of Mesenchymal Stem Cells to Neuroglia: in the Context of Cell Signalling. Stem Cell Rev Rep 2019;15:814-26. doi: 10.1007/s12015-019-09917-z.
14. Feldman EL, Boulis NM, Hur J, Johe K, Rutkove SB, Federici T, et al. Intraspinal neural stem cell transplantation in amyotrophic lateral sclerosis: phase 1 trial outcomes. Ann Neurol 2014;75:363-73. doi: 10.1002/ana.24113.
15. Petrou P, Kassis I, Yaghmour NE, Ginzberg A, Karussis D. A phase II clinical trial with repeated intrathecal injections of autologous mesenchymal stem cells in patients with amyotrophic lateral sclerosis. Front Biosci 2021;26:693-706. doi: 10.52586/4980.
16. Berry JD, Cudkowicz ME, Windebank AJ, Staff NP, Owegi M, Nicholson K, et al. NurOwn, phase 2, randomized, clinical trial in patients with ALS: Safety, clinical, and biomarker results. Neurology 2019;93:e2294-305. doi: 10.1212/WNL.0000000000008620.
17. Barczewska M, Maksymowicz S, Zdolińska-Malinowska I, Siwek T, Grudniak M. Umbilical Cord Mesenchymal Stem Cells in Amyotrophic Lateral Sclerosis: an Original Study. Stem Cell Rev Rep 2020;16:922-32. doi: 10.1007/s12015-020-10016-7.
18. Cudkowicz ME, Lindborg SR, Goyal NA, Miller RG, Burford MJ, Berry JD, et al. A randomized placebo-controlled phase 3 study of mesenchymal stem cells induced to secrete high levels of neurotrophic factors in amyotrophic lateral sclerosis. Muscle Nerve 2022;65:291-302. doi: 10.1002/mus.27472.
19. Ruiz-López FJ, Guardiola J, Izura V, Gómez-Espuch J, Iniesta F, Blanquer M, et al. Breathing pattern in a phase I clinical trial of intraspinal injection of autologous bone marrow mononuclear cells in patients with amyotrophic lateral sclerosis. Respir Physiol Neurobiol 2016;221:54-8. doi: 10.1016/j.resp.2015.11.007.
20. Petrou P, Gothelf Y, Argov Z, Gotkine M, Levy YS, Kassis I, et al. Safety and Clinical Effects of Mesenchymal Stem Cells Secreting Neurotrophic Factor Transplantation in Patients With Amyotrophic Lateral Sclerosis: Results of Phase 1/2 and 2a Clinical Trials. JAMA Neurol 2016;73:337-44. doi: 10.1001/jamaneurol.2015.4321.
21. Syková E, Rychmach P, Drahorádová I, Konrádová Š, Růžičková K, Voříšek I, et al. Transplantation of Mesenchymal Stromal Cells in Patients With Amyotrophic Lateral Sclerosis: Results of Phase I/IIa Clinical Trial. Cell Transplant 2017;26:647-58. doi: 10.3727/096368916X693716.
22. Marconi S, Bonaconsa M, Scambi I, Squintani GM, Rui W, Turano E, et al. Systemic treatment with adipose-derived mesenchymal stem cells ameliorates clinical and pathological features in the amyotrophic lateral sclerosis murine model. Neuroscience 2013;248:333-43. doi: 10.1016/j.neuroscience.2013.05.034.
23. Mohseni R, Hamidieh AA, Shoae-Hassani A, Ghahvechi-Akbari M, Majma A, Mohammadi M, et al. An open-label phase 1 clinical trial of the allogeneic side population adipose-derived mesenchymal stem cells in SMA type 1 patients. Neurol Sci 2022;43:399-410. doi: 10.1007/s10072-021-05291-2.
24. Staff NP, Madigan NN, Morris J, Jentoft M, Sorenson EJ, Butler G, et al. Safety of intrathecal autologous adipose-derived mesenchymal stromal cells in patients with ALS. Neurology 2016;87:2230-34. doi: 10.1212/WNL.0000000000003359.
25. Guo J, Yang X, Gao L, Zang D. Evaluating the levels of CSF and serum factors in ALS. Brain Behav 2017;7:e00637. doi: 10.1002/brb3.637.
26. Paczkowska E, Kaczyńska K, Pius-Sadowska E, Rogińska D, Kawa M, Ustianowski P, et al. Humoral activity of cord blood-derived stem/progenitor cells: implications for stem cell-based adjuvant therapy of neurodegenerative disorders. PLoS One 2013;8:e83833. doi: 10.1371/journal.pone.0083833.
27. Sobuś A, Baumert B, Litwińska Z, Gołąb-Janowska M, Stępniewski J, Kotowski M, et al. Safety and Feasibility of Lin- Cells Administration to ALS Patients: A Novel View on Humoral Factors and miRNA Profiles. Int J Mol Sci 2018;19:1312. doi: 10.3390/ijms19051312.
28. Riley J, Glass J, Feldman EL, Polak M, Bordeau J, Federici T, et al. Intraspinal stem cell transplantation in amyotrophic lateral sclerosis: a phase I trial, cervical microinjection, and final surgical safety outcomes. Neurosurgery 2014;74:77-87. doi: 10.1227/NEU.0000000000000156.
29. Guo X, Johe K, Molnar P, Davis H, Hickman J. Characterization of a human fetal spinal cord stem cell line, NSI-566RSC, and its induction to functional motoneurons. J Tissue Eng Regen Med 2010;4:181-93. doi: 10.1002/term.223.
30. Cizkova D, Kakinohana O, Kucharova K, Marsala S, Johe K, Hazel T, et al. Functional recovery in rats with ischemic paraplegia after spinal grafting of human spinal stem cells. Neuroscience 2007;147:546-60. doi: 10.1016/j.neuroscience.2007.02.065.
31. Glass JD, Hertzberg VS, Boulis NM, Riley J, Federici T, Polak M, et al. Transplantation of spinal cord-derived neural stem cells for ALS: Analysis of phase 1 and 2 trials. Neurology 2016;87:392-400. doi: 10.1212/WNL.0000000000002889.
32. Mazzini L, Gelati M, Profico DC, Sorarù G, Ferrari D, Copetti M, et al.. Results from Phase I Clinical Trial with Intraspinal Injection of Neural Stem Cells in Amyotrophic Lateral Sclerosis: A Long-Term Outcome. Stem Cells Transl Med 2019;8:887-97. doi: 10.1002/sctm.18-0154.
33. Mazzini L, Gelati M, Profico DC, Sgaravizzi G, Projetti Pensi M, Muzi G, et al. Human neural stem cell transplantation in ALS: initial results from a phase I trial. J Transl Med 2015;13:17. doi: 10.1186/s12967-014-0371-2.
34. Oh KW, Moon C, Kim HY, Oh SI, Park J, Lee JH, et al. Phase I trial of repeated intrathecal autologous bone marrow-derived mesenchymal stromal cells in amyotrophic lateral sclerosis. Stem Cells Transl Med 2015;4:590-7. doi: 10.5966/sctm.2014-0212