Journal of Neurology & Neurophysiology

Saeed S et al., J Neurol Neurophysiol 2014, 5:4
DOI: 10.4172/2155-9562-5-1000219

Review Article Open Access

The Use of Mesenchymal Stem Cells in the Treatment of Multiple Sclerosis: An Overview of Open Labels and Ongoing Studies

Shahbeigi Saeed[1*], **Sepehry Amir Ali**[1,2] **and Oger Joe**[1,3]

[1]*Neuroimmunology fellow from UBC Division of Neurology, Department of Medicine, Vancouver, Canada*

[2]*University of British Columbia (UBC), College for Interdisciplinary Studies, Graduate 3program in Neuroscience, Vancouver, Canada*

[3]*UBC Division of Neurology, Department of Medicine, Vancouver, Canada*

[*]**Corresponding author:** Dr Saeed Shahbeigi, UBC Hospital, 2211 Wesbrook Mall Room S-159, Vancouver BC Canada V6T 2B5, Canada, Tel: 604-971-3002; Fax: 604-822-0758; E-mail: s_shahbeigi@yahoo.com

Received date: May 08, 2014, **Accepted date:** Aug 26, 2014, **Published date:** Sep 01, 2014

Abstract

Multiple sclerosis (MS) is a demyelinating disease of unknown etiology that affects the Central Nervous System (CNS) where autoimmune-mediated mechanisms are thought to be at work. There are two possible options for treating MS, to prevent damage, and to repair the already impaired CNS. Stem Cells (SC) therapy emerges as a potential new hope for MS patients as it could accomplish both functions. There is a growing body of literature that supports the potential of the SC for immunomodulation and re-myelination. Here we focus on examining the registered published and on-going clinical trials using the Mesenchymal Stem Cell (MSC) therapy in MS. We have found that a total of 85 patients were enrolled in 9 cell-base studies with encouraging results. These studies were not statistically analyzed; however, they showed safety of the MSC therapy. Based on the results emerging from these patients, who failed to respond to even immunosuppressive drugs, clinical improvement was observed in 62%, a stable course in 22%; and 16% remaining in a progressive course. Given the evidence, we support that cell-based therapies are safe and reasonable to initiate a double blind, randomized controlled trials. This would represent a new and unique therapeutic approach for the progressive forms of MS.

Keywords: Mesenchymal stem cells; Multiple sclerosis; Neural progenitor MSC; Open-label and RCT clinical trials

Introduction

Multiple sclerosis (MS) is a demyelinating inflammatory disease, partly immune mediated of an unknown etiology, which roughly affects 400,000 individuals in the United States, and 560,000 in the European Union [1]. There are two generally accepted options for treating MS. First, preventing the damage (e.g., immunomodulatory therapies), and second, to repair the existing damage in Central Nervous System (CNS) (e.g., by re-myelination) [2]. Immunomodulatory treatments are also called disease-modifying drugs in MS (DMD). They improve the relapsing phase of the disease by reducing the frequency of relapses and new T2 lesions, but have very limited effect on the progressive neuro-degeneration phase of the disease. In addition, these DMDs, especially the newest ones (Tysabri®, or Gileniya® and Alemtuzumab) have serious side effects that limit their usage.

Endogenous adult human stem cells can re-myelinate the CNS; however, these stem cells are not very potent and re-myelination often fails or is inhibited, resulting in chronic demyelination and progressive axonal death [3]. Consequently, the exogenous stem cells (SC) emerge as a potential new hope for treatment of MS. There is a growing body of literature that supports the immunomodulation and re-myelination of the SC [2,4].

The biological source of the SC comes from embryonic (ESC), amniotic fluid (AFS) and adult type cells (ASC). Blastocysts are the first stage of developmental differentiation of embryonic cells, with an outer trophoblastic layer and an inner cell mass [5,6]. The Embryonic stem cells (ESC) are derived from the inner cell mass and are pluripotent cells capable of differentiating into three primary germ layers [7]. The FDA approved ESC for clinical trials in spinal cord injuries in 2009 in the United States [5,8].

The Amniotic Fluid Stem Cells (AFS) are multi-potent and expand substantially without feeders. These are non-tumorogenic because they retain long telomeres and a normal karyotype even after over 250 doublings. Additionally, they can differentiate into different cells including neuronal lines [9]. The AFS are preferred over the ESC because they can be prepared easily without needing intact human embryos [9].

The ASC are divided into many cells types including mesenchymal stem cells (MSC), neural progenitor cells (NPC), and hematopoietic stem cells (HSC). Recently a new type of pluripotent SC called induced pluripotent stem cell (iPSC) has been artificially derived from an adult somatic cell by inducing specific programmed genes into the cells [10,11]. This review will focus solemnly on the MSC.

MSC: General Properties

The MSC was first described in a population of adult stromal progenitors of the mesodermal lineage, in 1974 [12]. Within the bone marrow (BM), MSC is in close proximity with the hematopoietic stem cells (HSC). The MSC are important components of the HSC niche, which potentially support hematopoiesis [13]. The HSC are the progenitor source of myeloid and lymphoid cells, which produce various blood cells. Besides the close proximity of MSC to HSC in BM niche, it has been thought they have controlling and modulating

effects on hematopoiesis. Bartholomew et al. [14] showed that the MSC strongly suppress lymphocyte proliferation and demonstrated that MSC have an immunosuppressive activity. Administration of the MSC to six patients with metachromatic leukodystrophy (MLD) was the first transplantation approach to treat neurological diseases. The MSC infusion was followed by some improvements in neurological conditions in 4 out of 6 subjects with MLD [15].

Definition criteria of MSC

The International Society for Cellular Therapy (ISCT) defined the MSC by A) plastic-adherence in vitro, B) the absence of hematopoietic surface markers CD14, CD11b, CD19, CD34, CD45 and HLA-DR, C) the presence of surface markers CD73, CD90 and CD105, and D) in-vitro differentiation into adipocytes, chondroblasts and osteoblasts [16,17]. It is noteworthy of mentioning that there are multiple concerns about MSC.

First, whether the MSC from MS patient differ from those of normal individuals is an important question. Indeed, studies have shown that more than half of MS patients had a decrease in BM cellularity and a lower CD45+ cells content compared to healthy individuals. This may have been associated with previous immunosuppressive therapies. Nonetheless, the structure of the BM microenvironment and the ratio of the major lymphoid subsets appear to be unchanged [17-20].

Second, which type of stem cell, autologous or allogeneic MSC could be transplanted. Allogeneic MSC transplantation appears feasible because MSC are minimally immunogenic. To date, both animal and human studies support the use of allogeneic MSC without any rejection [21-24]. This property allows the use of universal donors [16].

Finally, it is not certain that the BM or adipose tissue provide the optimal source of MSC. The evidence points to adipose tissue as being a superior source for MSC because they are easy to harvest, have higher availability, and expand better ex-vivo than the BM cells [25]. They also provide a better recovery for blood flow, becoming an ideal source for therapeutic angiogenesis in ischemic and immunological diseases [26,27].

The immune effects of MSC

Studies completed on experimental allergic encephalomyelitis (EAE), revealed that the MSCs have beneficial effects. These experiments have revealed that the MSC, can improve EAE. Interestingly, T cells from the lymph nodes of the MSC-treated mice develop immune tolerance [28,29]. In addition, Darlington has demonstrated that Th-17 responses are increased in the experimental studies . However, “Human model” [30] fitting with recent studies showed that the MSC could modulate the peripheral immune system through suppression of Th17 responses [31]. The MSC potentially interact with cells of both the innate and adaptive immune systems. This has been speculated to inhibit the release of pro-inflammatory cytokines and promote the survival of damaged cells [32,33].

There are evidence showing that the MSC can alter the phenotype of NK cells and suppress proliferation, cytokine secretion, and cytotoxicity against HLA-class I- expressing targets. Some of these effects require cell-to-cell contact, whereas others are mediated by soluble factors, including transforming growth factor-beta1 (TGF-beta1) and prostaglandin E2, pointing to the existence of diverse mechanisms for the MSC-mediated NK-cell suppression [34]. The MSC have been reported to block the differentiation of monocytes into dendritic cells (DC) and impair antigen presentation [35] as well as IL-12 production. Also the human MSC (hMSC) alter cytokine secretion and induce more anti-inflammatory responses. Specifically, the hMSC by induction of mature dendritic cells (DC) decrease tumor necrosis factor alpha (TNF-alpha) secretion and increase IL-10 secretion [36]. The hMSC inhibit Th1 cells, decrease interferon gamma, and affect Th2 cells by increasing secretion of IL-4. This causes an increase in the proportion of T- Regulatory cell switches the CD4+ T cell responses from a Th1 to a Th2 polarized phenotype resulting in a decrease secretion of IFN-gamma from NK cells. The hMSC produce elevated prostaglandin E2 [31,36-38]. The B-cell proliferation and differentiation is inhibited by the hMSC, and secretion of IgM, IgG, and IgA is significantly impaired [37].

Gordon et al. [39] used intra-peritoneal infusion of the hMSC to evaluate whether the CNS engraftment is a significant concern for treatment. They demonstrated an extensive improvement of clinical symptoms in EAE mice with little CNS infiltration. These results also suggest that the MSC exert their therapeutic effect on inflamed CNS through a combination of the peripheral control of inflammation and in-situ neuro-immune effect cells as well as a neuro-protective effect [40].

A 24 hour longitudinal immunological assay in a MSC clinical trial with MS patients revealed an increase in the proportion of CD4(+) CD25(+) regulatory T cells, and a decrease in the proliferative responses of lymphocytes, after transplantation [41]. Nevertheless, according to Bonab and colleagues, gene expression and cytokine variations of IL-4, IL-10, IFN-gamma and TGF-b did not have any changes in the patients who received MSC by intratechal infusion. They have shown that interleukins and cytokines (e.g., IL-4, IL-10, IFN-G, and TGF-b) fail to change after intratechal infusion of MSC in MS patients, with exception of the IL.6. Interestingly in those patients without clinical improvement after MSC infusion, the IL6 was increased [42].

B cells	Inhibit B cell proliferation, differentiation
DC cells	Impaired antigen presentation Production of anti-inflammatory cytokines (IL-10)[31] Decrease TNFa, Decrease the expression of HLA-DR on myeloid dendritic cells[41]
NK cells	Inhibiting the proliferation of NK cells and cytokine production
T cells	Inhibit the proliferation of T lymphocytes, Promote the generation of CD4+ T regulatory cells (Treg)[23] Switch CD4+ T cell responses from a Th1 to a Th2 polarized phenotype Induction of Th1 drop INF Gamma Induction of Th2 increase IL.4

Table 1: The immune effects of MSC

Drugs’ interaction with MSCs

The hMSC have different patterns of response to chemotherapy commonly used in transplantation. Following BMT, the hMSC were shown to be relatively sensitive to a panel of cytotoxic agents, (e.g., paclitaxel, vincristine, etoposide and cytarabine). Furthermore,

different recovery patterns were noted. The evidence supports sustained suppression in the hMSC following 3-day exposure to paclitaxel, cytarabine and etoposide [43]. In contrast, significant recovery was observed in the hMSC treated with dexamethasone and vincristine, but the latter drug is known to transiently suppress the MSC proliferation [17].

Previous exposure to methotrexate, corticosteroids, anti-cytokine and biological agents or other disease-modifying anti-inflammatory drugs as well as drugs such as cyclosporine-A, D-penicillamine, hydroxychloroquine, leflunamide, and sulphasalazine-A fail to proliferate the BM MSCs [44]. Similarly, compounds such as azathioprine, cyclophosphamide, interferon, and mitoxantrone have been shown not to influence MSC [17].

Pretreatment of MSCs

When MSC are sitting in high IFN- γ, an inflammatory microenvironment, they appear to act as an anti-inflammatory agent [42]. In the presence of elevated amounts of IFN- γ (≥ 500 units/ml), the MSC express lower amounts of MHC- II, lose their antigen presenting function, and acquire immunosuppressive properties, which prevents dendritic cells maturation [16]. In addition, exposure to 17B-estradiol enhances the efficacy of adipose-derived MSCs [45]. Also evidence shows that pretreating the hMSC with the pro-inflammatory cytokine IL-1β accentuates the effect of MSC, and causes decrease in the Th1/17 subset [31].

Clinical Trials

MSC

The first publication of a clinical trial using MSC was in 2007. Bonab and colleagues assessed safety and efficacy of the MSC in 10 patients with the progressive type of MS. They were unresponsive to the DMD as well as Mitoxantrone. They did not report any major adverse events (AE) after MSC therapy. Their Expanded Disability Status Scale (EDSS) score ranged from 3.5 to 6 (Table 2). The Patients had bone marrow derived autologous MSC injection. During 13 to 26 months of follow up (mean: 19 months), the EDSS of one patient improved from 5 to 2.5 score, 4 showed no change, and 5 increased from 0.5 to 2.5. The MRI after 12 months showed 7 patients without any change, 2 had one more T2 lesion, and 1 had one less. This preliminary report emphasized the feasibility of autologous MSC for treatment of progressive type MS [46].

The subsequent study by Yamout et al. [47] explored the safety and therapeutic benefit of autologous BM derived MSC (BM-MSC) in 10 progressive MS patients (EDSS: 4.0 to 7.5). They infused the MSC into the subarachnoid space at C1–C2 and L2–L3 disc space levels under fluoroscopic guidance. The only major AE was a transient encephalopathy with seizures in one patient who received more than 100 million cells. In the next 6 months, the EDSS improved by 0.5–1.0 in 5 of 7 patients, unchanged in 1 patient, and worsened by 0.5 in 1 patient. The MRI results at 3 months revealed new or enlarging lesions in 5 of 7 patients and Gadolinium (Gd+) enhancing lesions in 3 of 7 patients. Vision and low contrast sensitivity testing at 3 months showed improvement in 5 of 6 and worsening in 1 of 6 patients. The overall attrition was 30%. They concluded that autologous MSC could be a treatment in progressive MS patients [47].

Karussis et al. published the other clinical trial in 2010 with 15 MS patients and 19 ALS patients [41]. All the patients were unresponsive to currently available agents for MS. Patients with MS received a mean intratechal autologous BM-MSC and 5 MS patients also received intravenous MSC. Meningeal irritation and aseptic self-limited meningitis was observed only in 1 patient. No major AE were reported in any of the patients during 25 months after infusion. After the 6 months, the EDSS score remained unchanged in 4 patients and was reduced by 0.5 point in 5. It improved by 1.0 point in 1 patient, by 1.5 points in 3, by 2 points in 1, and by 2.5 points in 1 patient. The EDSS score did not deteriorate in any of the patients. In all MS patients, the brain MRI showed no new or gadolinium-enhancing lesions.

Odinak et al. [48] in 2011 examined the autologoug MSC in 8 progressive MS patients. Patients received MSC via intravenous (IV) infusion every month for 4 to 8 months, and the efficacy of treatment was assessed for 12 months. No major AE was reported even after several infusions. The improvement of 0.5 point on EDSS was seen in 5 of 8 patients after 4 months. After 12 months, the improvement of 0.5-1 point on EDSS was seen in 6 of 8, stabilization in 1 of 8 and progression in 1 of 8 [48].

A UK study was conducted in 2012 with 10 patients with secondary progressive MS involving the visual pathways. Participants received a single IV infusion autologous BM-MSC. They did not encounter any severe AE. They had some improvement after treatment in visual acuity and VEP latency with an increase in optic nerve area. Pre-treatment rate of change for EDSS was +0.026 and post-treatment it was at -0.001. This variable showed a tendency towards significance ($p< 0.028$), which was lost after statistical correction. T1 hypointense lesion volume decreased after treatment and magnetization transfer ratio increased, but these changes were no longer statistically significant. These findings could provide indirect support for this idea that MSC can promote re-myelination and can have a neuroprotective effect [49].

Later in 2012, Bonab et al. enrolled 25 progressive MS patients; those were unresponsive to conventional treatments. Patients received a single intrathecal injection of autologous BM-MSC. After 12 months no major AE was reported. The EDSS improved in 4, deteriorated in 6 and had no change in 12 patients. This study had 12% attrition due to personal reasons. In MRI evaluation, 15 patients showed no change, 6 showed new T2 or gadolinium-enhanced lesions, and 1 was lost to follow-up [50].

Stromal vascular fraction cellular transplantation

In 2009, the first trial of Stromal Vascular Fraction (SVF) isolated from adipose tissue was conducted with 3 progressive male MS patients. The SVF contains endothelial progenitor cell (EPC), MSC, T regulatory cells, endothelial precursor cells, preadipocytes, as well as anti-inflammatory M2 macrophages. The SVF cells were infused by IV and intratechal. Infusions were very well tolerated without any AE or side effects. A couple of months after stem cell infusion their clinical condition were improving, and their brain MRI had no other new lesions [25].

Autologous BM cellular therapy

The Study of Intravenous Autologous Marrow in Multiple Sclerosis (SIAMMS) is the first reported trial of autologous BM stem cell therapy without myelo- or lympho-suppressive preconditioning. This study showed the safety and feasibility of BM harvesting and reinfusion. To exploit the possible therapeutic effects of all potentially beneficial BM stem cell subpopulations, the authors did not select any

single subpopulation of cells. Instead, this early clinical trial infused the whole BM. They treated 6 progressive MS patients with infusion of autologous BM cells. No major AE was reported. Clinical and paraclinical parameters including Global EP scores improved during the first 3 months post transplantation. The improvement was sustained, and was statistically significant at 1 year after transplantation. The EDSS scores remained stable over a period of 12 months after the therapy, and the scores on the MS Functional Composite scale (MSFC) were stable or even improved. On MRI, 3 months after transplantation, no new gadolinium-enhancing lesions were found [1].

Allogeneic Umbilical Cord MSC (UCMSC)

A 55-year-old woman with refractory progressive MS received transplanted umbilical cord mesenchymal stem cells (UCMSC) intrathecal (IT) and IV. The baseline EDSS score for this patient was 8.5, and 5 months post transplantation changed to 5.5. The MRI was dramatically improved. No major adverse effects were reported [24].

All these studies had in common the absence of blinding and controlled untreated randomized group, small number of subjects, and variable study durations and type of cells transplanted. They had similar conclusions, intrathecal, intraspinal and IV infusion of cell-based therapy had no major AE, and the patients tolerated the infusion very well. In addition, by pooling the evidence, shown in Table 2 and 3, one can see that a total of 85 patients were enrolled in 9 cell-base studies with encouraging results. Fifty-one patients (62%) had clinical improvement, 18 had a stable course (22%), and 13 (16%) had a worsening course. This means that 62% of the patients with progressive and aggressive type of MS that did not have any response to even immunosuppressive drugs, had an improvement in their disease course. However, these were open-label or case studies, which warrant future double blind RCTs.

No.	Study, year	Country	N (Total=85)	Female (n)	Age [MEAN (SD)]	EDSS [RANGE (MEAN)]	SC type	Infusion method	Cell count dosage injected	Times
1	Bonab 2012	Iran	25	19	34.7	4-6.5	MSC	IT	29.5x10(6)	Single
2	Connick, 2012	UK	10	3	48.8	5.5-6.5	MSC	IV	1.6x10(6)/KG	Single
3	Odianc, 2011	Russia	8	3	37.5	3.5-6.5 (5.6)	MSC	IV	x10(6)/kg	4-8 times
4	Karussis, 2010	Israel	15	8	35.3	4-8 (6.7)	MSC	IT(10), IT +IV(5)	63.2-+10(6)	Single
5	Yamout, 2010	Lebanon	7	4	39.3	4.5-7.5(6.5)	MSC	IT+IS	3-5x10(7)	Single
6	Bonab, 2007	Iran	10	0	33	3.5-6	MSC	IT	8.3x10(6)	Single
7	Riordan, 2009	USA	3	-	32 & 50*	-	SVF	IV.IT	5x10 (6)	Several
8	Rice, 2011	UK	6	4	45.85	4.5-6.5(5.9)	BM	IV	1.4X10(8) 1.1X10(6)	Single
9	Linag, 2009	China	1	1	55	8.5	Al-UC-MSC	IV, IT	1x107 2x107	Single

Table 2: Baseline demographic of the open-label clinical trials of Stem Cell-Based Therapy in MS; Note: EDSS: Expanded Disability Scoring System; IT: Intra-Techal; IS: Intra-Spinal; IV: Intra-Venous; *age for 2 of the 3 patients was reported by the authors.

Evidence shows that the Neural stem cells (NSC) exert a potent immunomodulation, neuroprotective and engineering effect on the nervous system [2]. The NSC-types are already 'neuralized' and there is no need for specific condition that lead to commitment of the nervous system. The NSC have neuronal specific markers called nestin, MAP-2, and tyrosine hydroxylase (TH) [51]. The prolonged culturing of the NSC lead to an ever increasing glial differentiation pattern at the expense of neuronal differentiation, which significantly reduces the therapeutic potential of the NSC [6]. Furthermore, neural progenitor cells (NPC) and oligodendrocyte progenitor cells (OPC) are embedded in the adult CNS requiring invasive techniques to acquire [2,52]. Therefore, it seems that these kinds of stem cells are not the optimal candidates for stem cell-based therapy. Nevertheless, there are studies that have shown hMSC emerge phenotypes and can differentiate into NSC-like cells in vitro [53]. Neurotrophic cytokines, such as human epidermal growth factor (hEGF) and bovine fibroblast growth factor (bFGF) can induce mesenchymal stem cells to differentiate into NSC. When BM-MSC are cultured with hEGF and bFGF, as a result the RNA expression of neuronal specific markers Nestin, MAP-2, and tyrosine hydroxylase (TH) are observed [51].

Interestingly, experts showed that neural stem cell derived from autologous bone MSC are safe and effective for treatment of motor deficits related to cerebral palsy [53]. As a result, one new area in treatment of CNS disease is the application of the NSC. An ongoing trial about MSC-NPC in treatment of progressive MS has been recently recruiting in the USA. Their results are expected to be ready for publication by the year 2015 (Table 3).

Study year		Follow-up	Adverse events	Clinical outcome			Imaging outcome		
		Months	Major	Improvement	Stable	Worsened	Improvement	Stable	Worsened
Bonab, 2012		12	None	4/22	12/22	6/22	0/22	15/22	6/22
Connick, 2012		10	None	10/10	0/10	0/10	Not significant		
Odianc, 2011		12	None	6/8	1/8	1/8			
Karussis, 2010		6	Transient aseptic meningitis	15/15	0/15	0/15	15/15	0/15	0/15
Yamout, 2010		12	Transient encephalopathy and seizure	5/7	1/7	1/7	0/7	2//7	5/7
Bonab, 2007		19	None	1/10	4/10	5/10	1/10	7/10	2/10
Riordan, 2009		12	None	3/3	0/3	0/3	0/3	3/3	0/3
Rice, 2011		12	None	6/6	0/6	0/6	6/6	0/6	0/6
Liang, 2009		12	None	1/1	0/1	0/1	1/1	0/1	0/1
Total:		6-19		51/82 (62%)	18/82 (22%)	13/82 (16%)	23/64 (36%)	27/64 (42%)	13/64 (20%)

Table 3: Outcome measures of the Stem Cell-Based Therapy clinical trials with MS patients

NSC

Ongoing Clinical Trials with MSC

Both United Kingdom and the US National MS Societies, with the support of national MS Societies from Italy, France, Canada and Australia, organized a meeting in London, UK on the 19th May of 2009 with the sole aim of producing a consensus statement on the use of stem cell therapies in MS [2]. Henceforth, the experts prepared a proposal implementation of an international research team on MSC in MS. Based on this proposal, they were willing to run a double blind study in a group of patients with MS. However, many scientists fail to commit to that proposal and seems that the proposed plan is not a complete pathway for cell-based therapy. Here we have reviewed all clinical trials that were registered in the US National Library of Medicine, PubMed ("clinicaltrial.gov") by searching both published and unpublished clinical trials with the following key terms: "MS"AND "STEM CELL", we have found 17 ongoing clinical trials about stem cells and MS. It should be taken into account that we did not consider hematopoietic stem cells (HPSC) because that needs substantial detail for its explanation, which is beyond the scope of this paper, and it needs to be discussed in future papers (Table 4).

NCT	Age	Completion date	Country	Intervention	Patients	Phase
01377870	18-55	Dec.2013	Iran	BM.MSC	30	I-II
01895439	18-65	Dec.2014	Jordan	BM.MSC	30	I-II
01883661	18-65	Dec.2015	India	BM, UC. MSC	15	-
01844957	18-50	Sep.2014	Italy	BM.MSC	20	-
01364246	16-65	Dec.2014	China	UC.MSC	20	I, II
01730547	18-50	Dec.2015	Sweden	BM.MSC	15	I, II
01056471	>=18	June.2012	Spain	AD.MSC	30	I, II
01228266	18-50	Dec.2013	Spain	BM.MSC	16	II
00813969	18-55	Dec.2014	USA	BM.MSC	24	-
01745783	18-50	Nov.2014	Spain	BM.MSC	30	I, II
01933802	18-70	Oct.2016	USA	NP-MSC	20	I
01606215	18-50	July.2015	England	BM.MSC	13	-
----------	18-50	-	Canada	BM.MSC	30	I, II
01453764	18-80	Dec.2014	Mexico	AD.MSC	10	I, II
01932593	25-80	Sep.2015	UK	BM.MSC	6	I
00927108	>=20	Dec.2011	Thailand	ODP.SC	10	-
01815632	18-65	Oct.2018	UK	BM	80	II

Table 4: Ongoing registered clinical trails on Stem Cell-Based Therapy in MS around the world

Given the current available data, 2 studies should have been completed by Dec. 2013, 6 studies by 2014, 5 by 2015, one by 2016 and finally one study in 2018. One study should have been done in 2011; however, we did not have any evidence to support the fate of this study. Spain with 3 ongoing studies has the most research about the MSC on MS. In terms of cell-based therapy, the most common type is the BM-MSC (10 ongoing studies), and the other sources are adipose tissue MSC, umbilical stem cell, neural progenitor derived MSC, oligodendrocytes progenitor stem cell and finally the BM stem cell. The route of infusion in all of these studies is intravenous (IV), except in one Mexican study where the route of infusion is IV and IT. Nine of

the 17 studies have determined the dosage for administration of the MSC. All studies but 3 are going to have only one single infusion. Nine studies will be done as double blind; the rest are open labels. In total, since early registration of these studies in 2007, with expected completion by 2018, after about 11 years, 416 patients will be enrolled. Interestingly, the enrollment criteria were clearly defined in only 11 studies and among those only 9 will be double blind, and sadly 6 of them will have similar initial EDSS and final outcome measures. In other words, at the end of these studies, we will have only an estimated 124 patients that will be enrolled in similar manners. This means that the worldwide trend for use of the MSC in MS has a slow progressing way; despite it is the only hope to treat progressive unresponsive MS.

NCT	Route	Dose	Times	Masking	EDSS	Selection criteria	Out-comes
01377870	IV	?	Single	DB	3-6.5	D	AE, EDSS, MSFC, RAO, B-C. MRI, QOL
01895439	IV	?	Single	OL	<=6	ND	AE, B.MRI, OPHTHALMIC TESTS
01883661	IV	?	6 in one month	OL	?	ND	AE, EDSS, QOL, B.MRI, CSF, VEP
01844957	IV	1-2x10(6)/kg	Single	DB	3-6.5	D	B.MRI, Relapse, EDSS
01364246	IV	?	?	OL	?	ND	AE, EDSS, BAER, SSEP, VEP, B.MRI
01730547	IV	?	?	DB	3-6.5	D	AE, B.MRI, Relapse, Disability progression
01056471	IV	10(6), 4x10(6)/kg	Single	DB	>=5.5, <=9	D	AE, B.MRI, IMMUNOLOGY, NEUROPHYSIOLOGY, NEUROPSYCHOLOGY
01228266	IV	2x10(6)/kg	Single	DB	3-6.5	D	AE, Clinical, B.MRI, OCT, IMMUNOLOGY, GOL
00813969	IV	2x10(6)/kg	Single	OL	3-6.5	D	AE, B.MRI, Disease activity
01745783	IV	2x10(6)/kg	Single	DB	?	D	AE, B.MRI, EDSS, MSFC, QOL, Disease free patients
01933802	IV	2-10x10(6)/kg	3 each three months	OL	>=3	D	AE, Evoked potentials, QOL, EDSS, MSFC, B.MRI
01606215	IV	2x10(6)/kg	Single	DB	3-6.5	D	AE, B.MRI, MSFC, EDSS, IMMUNOLOGY
----------	IV	1-2x10(6)/kg	Single	DB	3-6.5	D	?
01453764	IV, IT	?	Single	OL	?	ND	AE, EDSS, Relapse
01932593	IV	?	Single	OL	?	ND	AE, Evoked potentials, B.MRI, EDSS, MSFC, MSIS-29
00927108	?	?	?	OL	?	ND	?
01815632	IV	1.5X10x8/kg	Twice	DB	4.0-6.0	D	Global EP, Safety, EDSS, MSIS, MSFC, B-C. MRI, OCT

Table 5: International ongoing clinical trials of the Stem Cell-Based Therapy in MS: Study designs;

Conclusions

Based on the presented data, it is clear that the cell-based therapies are safe, reasonable, and represents a new and unique therapy for progressive MS.

To be accepted as a feasible treatment, cell-based therapy should be, a) Evaluated in double blind randomized clinical trials; b) Applied not only in the late degenerative phase of MS, but also in the early relapsing phase of MS where the immune system is much more active than in the progressive phases; c) Possibly in association with disease modifying drug (DMD); d) Done at high dose of stem cells for example 4-5 million cells per kg in divided doses.

References

1. Rice CM, Mallam EA, Whone AL, Walsh P, Brooks DJ, et al. (2010) Safety and feasibility of autologous bone marrow cellular therapy in relapsing-progressive multiple sclerosis. Clin Pharmacol Ther 87: 679-685.
2. Martino G, Franklin RJ, Baron Van Evercooren A, Kerr DA; Stem Cells in Multiple Sclerosis (STEMS) Consensus Group (2010) Stem cell transplantation in multiple sclerosis: current status and future prospects. Nat Rev Neurol 6: 247-255.

3. Karussis D, Petrou P, Kassis I (2013) Clinical experience with stem cells and other cell therapies in neurological diseases. J Neurol Sci 324: 1-9.
4. Laroni A, Novi G, Kerlero de Rosbo N, Uccelli A (2013) Towards clinical application of mesenchymal stem cells for treatment of neurological diseases of the central nervous system. J Neuroimmune Pharmacol 8: 1062-1076.
5. Embryonic Stem Cell Therapy At Risk? Geron Ends Clinical Trial. Science Debate, 2011.
6. Gögel S, Gubernator M, Minger SL (2011) Progress and prospects: stem cells and neurological diseases. Gene Ther 18: 1-6.
7. Keirstead HS, Nistor G, Bernal G, Totoiu M, Cloutier F, et al. (2005) Human embryonic stem cell-derived oligodendrocyte progenitor cell transplants remyelinate and restore locomotion after spinal cord injury. J Neurosci 25: 4694-4705.
8. Kaiser J (2011) Embryonic stem cells. Researchers mull impact of Geron's sudden exit from field. Science 334: 1043.
9. De Coppi P1, Bartsch G Jr, Siddiqui MM, Xu T, Santos CC, et al. (2007) Isolation of amniotic stem cell lines with potential for therapy. Nat Biotechnol 25: 100-106.
10. Baker M (2007) Adult cells reprogrammed to pluripotency, without tumors.
11. Takahashi K, Yamanaka S (2006) Induction of pluripotent stem cells from mouse embryonic and adult fibroblast cultures by defined factors. Cell 126: 663-676.
12. Friedenstein A J, Chailakhyan R K, Latsinik N V, Panasyuk A F and Keiliss-Borok I V (1974) Stromal cells responsible for transferring the microenvironment of the hemopoietic tissues. Cloning in vitro and retransplantation in vivo. Transplantation. 17(4): p. 331-40.
13. Méndez-Ferrer S, Michurina TV, Ferraro F, Mazloom AR, Macarthur BD, et al. (2010) Mesenchymal and haematopoietic stem cells form a unique bone marrow niche. Nature 466: 829-834.
14. Devine SM, Cobbs C, Jennings M, Bartholomew A, Hoffman R (2003) Mesenchymal stem cells distribute to a wide range of tissues following systemic infusion into nonhuman primates. Blood 101: 2999-3001.
15. Koç ON, Day J, Nieder M, Gerson SL, Lazarus HM, et al. (2002) Allogeneic mesenchymal stem cell infusion for treatment of metachromatic leukodystrophy (MLD) and Hurler syndrome (MPS-IH). Bone Marrow Transplant 30: 215-222.
16. Auletta JJ, Bartholomew AM, Maziarz RT, Deans RJ, Miller RH, et al. (2012) The potential of mesenchymal stromal cells as a novel cellular therapy for multiple sclerosis. Immunotherapy 4: 529-547.
17. Mallam E, Kemp K, Wilkins A, Rice C, Scolding N (2010) Characterization of in vitro expanded bone marrow-derived mesenchymal stem cells from patients with multiple sclerosis. Mult Scler 16: 909-918.
18. Carrai V, Donnini I, Mazzanti B, Alterini R, Amato M P, et al. (2013) Immunohistochemistry analysis of bone marrow biopsies in multiple sclerosis patients undergoing autologous haematopoietic stem cells transplantation. Clin Neurol Neurosurg 115: 1044-1048.
19. Mazzanti B, Aldinucci A, Biagioli T, Barilaro A, Urbani S, et al. (2008) Differences in mesenchymal stem cell cytokine profiles between MS patients and healthy donors: implication for assessment of disease activity and treatment. J Neuroimmunol 199: 142-150.
20. Rice CM, Scolding NJ (2010) Adult human mesenchymal cells proliferate and migrate in response to chemokines expressed in demyelination. Cell Adh Migr 4: 235-240.
21. Djouad F, Plence P, Bony C, Tropel P, Apparailly F, et al. (2003) Immunosuppressive effect of mesenchymal stem cells favors tumor growth in allogeneic animals. Blood 102: 3837-3844.
22. Hare J M, Traverse J H, Henry T D, Dib N, Strumpf R K, et al. (2009) A randomized, double-blind, placebo-controlled, dose-escalation study of intravenous adult human mesenchymal stem cells (prochymal) after acute myocardial infarction. J Am Coll Cardiol 54: 2277-2286.
23. Carrion FA, Figueroa FE (2011) Mesenchymal stem cells for the treatment of systemic lupus erythematosus: is the cure for connective tissue diseases within connective tissue? Stem Cell Res Ther 2: 23.
24. Liang J, Zhang H, Hua B, Wang H, Wang J, et al. (2009) Allogeneic mesenchymal stem cells transplantation in treatment of multiple sclerosis. Mult Scler 15: 644-646.
25. Riordan NH, Ichim TE, Min WP, Wang H, Solano F, et al. (2009) Non-expanded adipose stromal vascular fraction cell therapy for multiple sclerosis. J Transl Med 7: 29.
26. Kim Y, Kim H, Cho H, Bae Y, Suh K, et al. (2007) Direct comparison of human mesenchymal stem cells derived from adipose tissues and bone marrow in mediating neovascularization in response to vascular ischemia. Cell Physiol Biochem 20: 867-876.
27. Keyser KA, Beagles KE, Kiem HP (2007) Comparison of mesenchymal stem cells from different tissues to suppress T-cell activation. Cell Transplant 16: 555-562.
28. Zappia E, Casazza S, Pedemonte E, Benvenuto F, Bonanni I, et al. (2005) Mesenchymal stem cells ameliorate experimental autoimmune encephalomyelitis inducing T-cell anergy. Blood 106: 1755-1761.
29. Zhang J, Li Y, Lu M, Cui Y, Chen J, et al. (2006) Bone marrow stromal cells reduce axonal loss in experimental autoimmune encephalomyelitis mice. J Neurosci Res 84: 587-595.
30. Rafei M, Campeau P M, Aguilar-Mahecha A, Buchanan M, Williams P, et al. (2009) Mesenchymal stromal cells ameliorate experimental autoimmune encephalomyelitis by inhibiting CD4 Th17 T cells in a CC chemokine ligand 2-dependent manner. J Immunol 182: 5994-6002.
31. Darlington PJ, Boivin MN, Renoux C, François M, Galipeau J, et al. (2010) Reciprocal Th1 and Th17 regulation by mesenchymal stem cells: Implication for multiple sclerosis. Ann Neurol 68: 540-545.
32. Uccelli A, Moretta L, Pistoia V (2008) Mesenchymal stem cells in health and disease. Nat Rev Immunol 8: 726-736.
33. Uccelli A, Laroni A, Freedman MS (2013) Mesenchymal stem cells as treatment for MS - progress to date. Mult Scler 19: 515-519.
34. Sotiropoulou PA, Perez SA, Gritzapis AD, Baxevanis CN, Papamichail M (2006) Interactions between human mesenchymal stem cells and natural killer cells. Stem Cells 24: 74-85.
35. Ramasamy R, Fazekasova H, Lam EW, Soeiro I, Lombardi G, et al. (2007) Mesenchymal stem cells inhibit dendritic cell differentiation and function by preventing entry into the cell cycle. Transplantation 83: 71-76.
36. Aggarwal S, Pittenger MF (2005) Human mesenchymal stem cells modulate allogeneic immune cell responses. Blood 105: 1815-1822.
37. Corcione A, Benvenuto F, Ferretti E, Giunti D, Cappiello V, et al. (2006) Human mesenchymal stem cells modulate B-cell functions. Blood 107: 367-372.
38. Prevosto C, Zancolli M, Canevali P, Zocchi MR, Poggi A (2007) Generation of CD4+ or CD8+ regulatory T cells upon mesenchymal stem cell-lymphocyte interaction. Haematologica 92: 881-888.
39. Gordon D, Pavlovska G, Glover C P, Uney J B, Wraith D, et al. (2008) Human mesenchymal stem cells abrogate experimental allergic encephalomyelitis after intraperitoneal injection, and with sparse CNS infiltration. Neurosci Lett 448: 71-73.
40. Uccelli A, Laroni A, Freedman MS (2011) Mesenchymal stem cells for the treatment of multiple sclerosis and other neurological diseases. Lancet Neurol 10: 649-656.
41. Karussis D, Karageorgiou C, Vaknin-Dembinsky A, Gowda-Kurkalli B, Gomori JM, et al. (2010) Safety and immunological effects of mesenchymal stem cell transplantation in patients with multiple sclerosis and amyotrophic lateral sclerosis. Arch Neurol 67: 1187-1194.
42. Mohyeddin Bonab M, Mohajeri M, Sahraian MA, Yazdanifar M, Aghsaie A, et al. (2013) Evaluation of cytokines in multiple sclerosis patients treated with mesenchymal stem cells. Arch Med Res 44: 266-272.
43. Li J, Law HK, Lau YL, Chan GC (2004) Differential damage and recovery of human mesenchymal stem cells after exposure to chemotherapeutic agents. Br J Haematol 127: 326-334.

44. Kastrinaki M C, Sidiropoulos P, Roche S, Ringe J, Lehmann S, et al. (2008) Functional, molecular and proteomic characterisation of bone marrow mesenchymal stem cells in rheumatoid arthritis. Ann Rheum Dis 67: 741-749.
45. Ragerdi Kashani I, Hedayatpour A, Pasbakhsh P, Kafami L, Atlasi N, et al. (2012) 17Î²-Estradiol enhances the efficacy of adipose-derived mesenchymal stem cells on remyelination in mouse model of multiple sclerosis. Acta Med Iran 50: 789-797.
46. Mohyeddin Bonab M, Yazdanbakhsh S, Lotfi J, Alimoghaddom K, Talebian F, et al. (2007) Does mesenchymal stem cell therapy help multiple sclerosis patients? Report of a pilot study. Iran J Immunol 4: 50-57.
47. Yamout B, Hourani R, Salti H, Barada W, El-Hajj T, et al. (2010) Bone marrow mesenchymal stem cell transplantation in patients with multiple sclerosis: a pilot study. J Neuroimmunol 227: 185-189.
48. Odinak MM, Bisaga GN, NovitskiÄ AV, Tyrenko VV, Fominykh MS, et al. (2011) [Transplantation of mesenchymal stem cells in multiple sclerosis]. Zh Nevrol Psikhiatr Im S S Korsakova 111: 72-76.
49. Connick P, Kolappan M, Crawley C, Webber DJ, Patani R, et al. (2012) Autologous mesenchymal stem cells for the treatment of secondary progressive multiple sclerosis: an open-label phase 2a proof-of-concept study. Lancet Neurol 11: 150-156.
50. Bonab MM, Sahraian MA, Aghsaie A, Karvigh SA, Hosseinian SM, et al. (2012) Autologous mesenchymal stem cell therapy in progressive multiple sclerosis: an open label study. Curr Stem Cell Res Ther 7: 407-414.
51. Long X, Olszewski M, Huang W, Kletzel M (2005) Neural cell differentiation in vitro from adult human bone marrow mesenchymal stem cells. Stem Cells Dev 14: 65-69.
52. Rivera FJ, Aigner L (2012) Adult mesenchymal stem cell therapy for myelin repair in multiple sclerosis. Biol Res 45: 257-268.
53. Chen G, Wang Y, Xu Z, Fang F, Xu R, et al. (2013) Neural stem cell-like cells derived from autologous bone mesenchymal stem cells for the treatment of patients with cerebral palsy. J Transl Med 11: 21.