STEM CELLS®

REGENERATIVE MEDICINE

Concise Review: Mesenchymal Stem Cells in Neurodegenerative Diseases

ROTEM VOLKMAN AND DANIEL OFFEN

Key Words. Mesenchymal stem cells • Neurodegenerative diseases • Neurotrophic factors • Immunomodulation • Neurogenesis

Tel Aviv University, Tel Aviv-Yafo, Israel

Correspondence: Daniel Offen, Ph.D., Felsenstein Medical Research Center, Rabin Medical Center, Jabotinsky Street 39 Petah Tikva, Israel 4963211. Telephone: +972-3-9376130; Fax: +972-3-9376181; e-mail: danioffen@gmail.com

Received February 2, 2017; accepted for publication May 6, 2017; first published online in STEM CELLS *EXPRESS* June 7, 2017.

1066-5099/2017/$30.00/0

http://dx.doi.org/10.1002/stem.2651

Available online without subscription through the open access option.

ABSTRACT

Stem cell-based therapies for neurodegenerative diseases aim at halting clinical deterioration by regeneration and by providing local support for damaged tissue. Mesenchymal stem cells (MSCs) hold great potential for cell therapy as they can be efficiently derived from adult tissue, ex vivo expanded in culture and safely transplanted autologously. MSCs were also shown to be able to differentiate toward neural fates and to secrete a broad range of factors able to promote nervous tissue maintenance and repair. Moreover, upon transplantation, MSCs were shown capable of homing toward lesioned areas, implying their potential use as vehicles for therapeutic agents administration. Indeed, various advantageous effects were reported following human MSCs transplantation into rodent models of neurodegenerative diseases, such as neurotrophic factor-mediated protection, enhanced neurogenesis, modulation of inflammation, and abnormal protein aggregate clearance. Recent studies have also used ex vivo manipulation for enhanced expression of potentially favorable factors, by so exploiting the homing capacity of MSCs for effective expression at the lesion site. Here, we will summarize current advancements in MSCs-based therapies for neurodegenerative diseases. We will examine the roles of central mechanisms suggested to mediate the beneficial effects of MSCs-based therapy and consider the augmentation of these mechanisms for superior clinical outcomes in rodent models of neurodegeneration as well as in clinical trials. STEM CELLS *2017;35:1867–1880*

SIGNIFICANCE STATEMENT

Mesenchymal stem cells (MSCs) hold great potential as source for cell-based therapy for neurodegenerative diseases. In this review, we summarize recent progress using human MSC transplantation into rodent models of neurodegeneration. We examine the molecular mechanisms mediating disease amelioration through this approach and emphasize the potential of ex vivo manipulation for these cells before transplantation.

INTRODUCTION

Neurodegenerative diseases involve progressive decline in neuronal function, brain atrophy, and often involve abnormal deposition of proteins. Although various neurodegenerative diseases, such as Alzheimer's disease (AD), Parkinson's disease (PD), Huntington's disease (HD), amyotrophic lateral sclerosis (ALS), and multiple system atrophy (MSA) occur in different brain regions and display different etiology, cumulative data suggest common cellular and molecular mechanisms.

Although there are immense efforts for the development of therapies for neurodegenerative diseases throughout the last several decades, effective therapeutic agents are still of need. This lack may result from several challenges: First, although multiple cellular and molecular mechanisms were shown to be involved in the etiology of these diseases, the cause of neuronal death still remains obscure, and no single molecular pathway was shown able to modulate disease progression. Second, for most of these diseases, early diagnosis is impeded due to the absence of efficient biomarkers. Third, progressed neurodegeneration often involves secondary effects such as chronic inflammation, requiring adjustment of treatment. Finally, drugs administered into the central nervous system (CNS) should be able to cross the brain–blood barrier (BBB), as well as to target specific cell types within different CNS regions, requiring efficient vectors able to carry therapeutic agents toward their target sites.

Mesenchymal stem cells (MSCs) are adult multipotent progenitors derived from various adult tissues and are capable of self-renewal in vitro [1]. MSCs are defined by their spindle-shaped

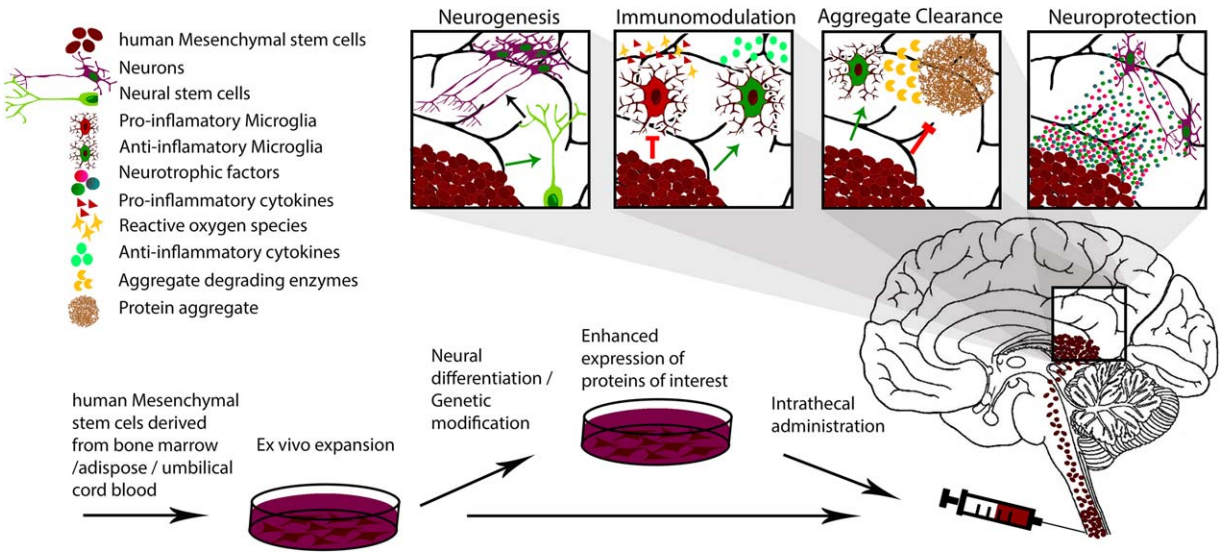

Figure 1. Human mesenchymal stem cell (hMSC)-based therapy for neurodegeneration. Shown is a scheme describing major stages in hMSCs-based therapy: derivation from adult tissue, ex vivo expansion and manipulation, administration into the spinal canal, and migration toward lesioned area. Enlarged areas show the molecular mechanisms shown to contribute to amelioration of neurodegenerative diseases upon MSCs transplantation.

morphology, their ability to adhere to tissue culture plastic, and their unique expression of cluster of differentiation cell surface molecules [2]. Upon growth in culture, MSCs are able to greatly expand while retaining their multipotent potential. This allows the generation of high cell quantities, setting these cells as highly efficient source for cell-based therapy. Also, MSCs were safely used for autologous transplantation, and exhibited no toxicity or tumorigenicity following transplantation into rodents or human patients [3, 4]. Moreover, upon transplantation, MSCs possess the capability to migrate toward neural lesions, presumably due to attraction by chemokines [5, 6]. Finally, paracrine secretion from these cells offer broad clinical potential by regulating immunomodulation, angiogenesis, apoptosis, oxidative stress, cell-differentiation, extra-cellular matrix composition, and more (reviewed in ref. 7).

Multiple reports over the last decade showed improvement in various models of neurodegenerative diseases or acute brain insults following MSCs transplantation in multiple rodent models. MSCs transplantation often improved survival rates, declined pathology, and rescued cognitive function decline. However, the exact mechanism by which MSCs exert their function remains debatable, as several mechanisms have been offered, such as neuroprotection by secretion of neurotrophic factors (NTFs), induction of neurogenesis, modulation of inflammation, and prevention of misfolded protein aggregation (illustrated in Fig. 1). As the majority of these diseases display complex etiology, it seems that multiple beneficial roles of MSCs are able to target different aspects of diseases. However, understanding the molecular mechanisms by which these cells exert their function could facilitate the generation of advanced MSCs-based therapies for neurodegeneration.

In this review, we will present the advances in human MSCs (hMSCs)-based therapies in rodent models of neurodegenerative diseases such as AD, PD, ALS, and MSA as well as in acute models of stroke. We will also review outcomes from human clinical trials using this therapeutic approach and will discuss the limitations that hamper the therapeutic potential of these cells. We will focus on the different mechanisms enhanced following treatment with these cells (as summarized in Table 1) and will describe the methods used to improve therapeutic efficacy of the hMSCs (as summarized in Table 2).

Induction of Neuronal Regeneration

Neurodegeneration is mainly characterized by progressive neuronal loss. However, various neurodegenerative diseases exhibit unique neuronal pathologies. PD involves the loss of dopaminergic neurons in the Substantia Nigra (SN). ALS involves the degeneration of motor neurons (MNs) in the brainstem and spinal cord. AD entails global neuronal loss in the cerebral cortex and hippocampus, and HD is characterized by degeneration of projection neurons in the dorsal striatum [8]. As the molecular mechanisms driving neuronal pathology in these diseases remains elusive, functional recovery by regeneration of damaged tissue is regarded a major therapy strategy.

Neuronal Differentiation of MSCs

The discovery that MSCs derived from both mouse and human origins can be manipulated to differentiate into functional neurons [9–11] encouraged the use of MSCs-derived neuronal cell types for the replacement of damaged neural tissue. Emerging protocols have used specific culture conditions for ex vivo differentiation of human MSCs into dopamine-secreting [12, 13] and acetylcholine (Ach)-secreting [14, 15] neuronal-like cells. Alternatively, superior similarity to original neuronal subtypes was reported through the genetic modification-mediated transdifferentiation of human bone marrow-derived MSCs (hBM-MSCs) using ectopic expression of neuronal subtype-specific transcription factors [16, 17]. Although few studies have shown functional recovery in brain injury upon transplantation of MSCs-derived neuronal cells into murine brains [17–20], it is not clear whether the observed recovery was indeed due to functional integration

Table 1. Naïve human mesenchymal stem cells in murine models of neurodegeneration

	Authors	Cell source	Model animal	Suggested mechanism	Clinical improvement
Alzheimer's	[61]	Umbilical cord blood	Aβ-inoculated mice	Reduced glia activation, oxidative stress, and apoptosis	Improved learning/memory performance
Alzheimer's	[36]	Umbilical cord blood	APP/PS1 mice	Reduced Aβ and p-tau deposition Modulation of microglia activation	Improved spatial learning and memory decline
Alzheimer's	[89]	Umbilical cord blood	APP/PS1 mice	sICAM-1 mediated upregulation of neprilysin Induced Aβ clearance	Increased neuronal survival in vitro
Alzheimer's	[93]	Not mentioned	Aβ-inoculated mice	Induction of autophagy Induced Aβ clearance	Enhanced hippocampal neurons survival
Alzheimer's	[37]	Not mentioned	Aβ-inoculated mice	Enhanced WNT signaling Enhanced endogenous neurogenesis	Improved working memory
Parkinson's	[34]	Bone marrow	6-OHDA mice	NTF secretion Enhanced endogenous neurogenesis	Enhanced dopaminergic neurons survival
Parkinson's	[36]	Bone marrow	MPTP-induced mice	Enhance EGFR expression Enhanced endogenous neurogenesis	Enhanced dopaminergic neurons survival
Parkinson's	[94]	Bone marrow	MPTP-induced mice	Induction of autophagy Induced α-synuclein clearance	Enhanced dopaminergic neurons survival
Parkinson's	[63]	Bone marrow	α-Synuclein-inoculated mice	Modulation of microglia activation MSCs IL-4 secretion Induced α-synuclein clearance	Increased neuronal survival
Stroke	[35]	Bone marrow	Acute ischemia rats	NTF secretion Enhanced endogenous neurogenesis Reduced apoptosis	Reduced infarct volume Functional recovery in NSS
Stroke	[65]	stable bone marrow derived B10 line	Acute ischemia rats	Reduced microglia activation Reduced NFκB signaling	Reduced infarct volume
Stroke	[71]	Adipose	Acute ischemia rats	BBB integrity maintenance Reduced neutrophil infiltration Reduced endothelial vasculature damage	Enhanced neuronal survival Improved functional behavior
Stroke	[70]	Bone marrow	LPS induced rats	BBB integrity maintenance Reduced neutrophil infiltration modulation of microglia activation	Enhanced neuronal survival
MS	[41]	Bone marrow	EAE mice	NGF mediated axonal protection	Reduced mortality Reduced disease severity Reduced neuronal loss
MS	[73]	Bone marrow	EAE mice	Reduced immune cell infiltration Induced Th2 immune response Enhanced oligodendrogenesis	Deceleration in disease progression
MS	[74]	Adipose	EAE mice	Decreased spinal cord inflammation Decreased demyelination Induced Th2 immune response Neurotrophic factor secretion Enhanced oligodendrogenesis	Reduced disease severity Reduced axonal loss
MS	[75]	Bone marrow	EAE mice	Reduced demyelination	Decreased white matter lesions Reduced disease severity
MS	[78]	Placenta	EAE mice	Neuroprotection Immunomodulation mediated by reduced TSG6 expression	Reduced disease severity
MS	[77]	Decidua	EAE mice	Decreased inflammatory infiltration Immunomodulation	Reduced disease severity

Table 1. *Continued*

	Authors	Cell source	Model animal	Suggested mechanism	Clinical improvement
MS	[76]	Adipose	EAE rats	Decreased immune cell infiltration HLA-G mediated Immunomodulation	Reduced disease severity Amelioration of axonal loss Enhanced vascular congestion
Inflammation	[70]	Bone marrow	LPS induced rats	BBB integrity maintenance Reduced neutrophil infiltration modulation of microglia activation	Enhanced neuronal survival
ALS	[60]	Bone marrow	SOD1 female mice	Reduced astrogliosis and microglia activation	Sustained survival of motor neurons Improved motor functions
ASD	[33]	Bone marrow	BLBP mice	NTF secretion Enhanced endogenous neurogenesis	Reduced stereotipical behavior Reduced cognitive rigidity Improved social behavior

Abbreviations: BBB, brain-blood barrier; EAE, experimental autoimmune encephalomyelitis; HLA-G, human leukocyte antigen G; 6-OHDA, 6-hydroxydopamine; IL-4, interleukin 4; LPS, lipopolysaccharide; MPTP, 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine; MSC, mesenchymal stem cells; NFκB, nuclear factor kappa-light-chain-enhancer of activated B cells; NGF, nerve growth factor; NSS, neurological Severity Score; NTF, neurotrophic factors; sICAM-1, soluble intracellular adhesion molecule-1; TSG6, tumor necrosis factor-inducible gene 6.

of MSCs-derived neuronal cells into excising neuronal networks. Moreover, the ability of MSCs to differentiate into fully functional neurons was not proven, as their transdifferentiation capacity and electrophysiological functionality remains under debate (reviewed in refs. 21, 22). These challenges, as well as the difficulty in establishing sufficient amount of cells for transplantation, discouraged further advancement of this strategy.

Induction of Endogenous Neurogenesis

An alternative approach for functional regeneration after neuronal loss is to encourage endogenous neural stem cells in the brain to generate the appropriate neurons. In mice, subventricular zone (SVZ) cell proliferation and neuronal differentiation are enhanced following stroke, providing evidence that the adult brain has the capacity to replace damaged cells using endogenous precursors [23]. Impaired neurogenesis, on the other hand, was associated with several neurodegenerative diseases [24], suggesting that inducing neurogenesis can be used as a therapy. Identification of factors that enhance endogenous neurogenesis in acute insult or chronic neurodegeneration was suggested as an effective therapeutic approach.

NTFs are secreted proteins that regulate multiple aspects of neural cell functions and are widely known to play central roles during brain development, homeostasis, and neurodegeneration [25]. In particular, multiple neurotrophic factors (NTFs) have been implicated in induction of neurogenesis in the adult SVZ. Brain-derived NTF (BDNF) and vascular endothelial growth factor (VEGF) administration into the lateral ventricles of adult rats was shown to increase the generation of new neurons [26]. The enhancement of neurogenesis by enriched environment was shown to be dependent on BDNF [27] and VEGF was shown to mediate exercise-induced neurogenesis [28]. Also glial cell line-derived NTF (GDNF), fibroblast growth factor 2 (FGF2), and neurotrophin-3 (NT-3) were shown to have roles in enhancing adult neurogenesis [29, 30].

Munoz and colleagues has shown that (hBM-MSCs) injected into the dentate gyrus of healthy mice were able to promote proliferation and differentiation of neural stem cells [31]. This effect was attributed to the elevated secretion of NTFs such as nerve growth factor (NGF), VEGF, cilliary NTF (CNTF) and FGF2 from transplanted cells. We showed that hBM-MSCs injected into the SVZ and sub-granular zone (SGZ) of healthy mice housed in enriched cages, promoted neurogenesis in the SVZ but not the SGZ associated with elevated BDNF secretion [32]. Notably, human nuclei staining analysis confirmed that neural progeny was derived from endogenous progenitors rather than from the transplanted cells.

The induction of neurogenesis by hMSCs administration was shown beneficial in several models of neurological diseases. We have recently shown a beneficial effect of hBM-MSCs in a BTBR mouse model of autism spectrum disorder. hBM-MSCs transplantation into these mice, who typically show reduced hippocampal neurogenesis and BDNF secretion, resulted in elevation of these properties along with improved cognitive functions [33]. Dramatic improvement in cognitive functions and social behavior were demonstrated 3 weeks following transplantation, suggesting the role of NTFs secretion-mediated induced neurogenesis in this model. Induced neurogenesis following hBM-MSCs transplantation was also reported in a 6-hydroxydopamine (6-OHDA) mouse model of PD, where grafted cells were shown to secrete

BDNF 23 days after transplantation [34]. In a rat model of cerebral ischemia, we also observed enhanced neurogenesis that was associated with induced levels of BDNF, NT-3, and VEGF were reported following transplantation, suggesting that neurogenesis was mediated by NTFs secretion [35].

Park et al. showed that hBM-MSCs administration enhanced the expression of epidermal growth factor receptor (EGFR) and enhanced neurogenesis in the SVZ and the SN in an 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP) model of PD. As EGFR is known to enhance neural progenitor cell (NPC) proliferation, the authors suggested that the neurogenic activity of hBM-MSCs was indeed mediated through EGFR signaling [36].

WNT signaling is known to be involved in proliferation capacity of NPCs. Beta-amyloid (Aβ) exposure of murine NPCs was shown to decrease their proliferation capacity as well as the expression level of the WNT components β-catenin and NGN1. Coculture of NPCs with hMSCs significantly rescued the expression of these genes and enhanced proliferation and neuronal differentiation. Importantly, hMSCs-mediated enhancement of neurogenesis was dependant on WNT signaling activation. hMSCs transplantation into Aβ-treated mice showed enhanced differentiation of NPCs in the hippocampus into mature neurons [37].

Together, these findings show that paracrine secretion from hMSCs results with enhanced neurogenic capacity and subsequent functional improvement. However, we cannot exclude the possibility of direct effect of NTF and other factors on various cellular processes such as homeostasis, reduction in oxidative stress, modulation of inflammation, and neuronal survival. As the modulation of neurogenesis is widely accepted as a key mechanism regulating neurodegeneration, the induction of neurogenesis by cell therapy using hMSCs holds great potential for future therapy (reviewed in ref. 25).

Enhanced NTFs Secretion

In addition to their role in promoting neurogenesis, ectopic expression of NTFs is known to improve neuronal survival following acute and chronic brain damage, providing a potential strategy for the treatment of various neurodegenerative diseases [38–40]. Functional improvement in murine models of neurodegeneration following hMSCs transplantation is frequently attributed to enhanced levels of NTFs in the brain, providing neuroprotection, reduction in oxidative stress, induced neurogenesis, and modulation of the inflammatory response [32–36, 41–45].hBM-MSCs were shown to express neural genes and were suggested to be predisposed to differentiate into neural cell fates [46]. We sought to harness this potential to generate hBM-MSCs-derived, NTFs secreting, astrocytic-like cells. We developed a novel protocol for ex vivo differentiation to astrocytic-like cells termed NTFs-secreting MSCs (MSC-NTFs). MSC-NTFs show elevated secretion of BDNF, GDNF, and insulin-like growth factor 1 as well as elevated expression of astrocytic markers such as glial fibrillary acidic protein and glutamine synthetase.

MSC-NTFs were shown to evoke improved clinical outcomes compared with naïve hBM-MSCs in several rodent models of neurodegeneration. Upon MSC-NTFs transplantation into the striatum of 6-OHDA model of PD, rats show improved behavior in the amphetamine-induced rotations test, higher preservation of the tyrosine-hydroxylase-positive area in the striatum and reduced dopamine depletion when compared with naive hBM-MSCs [47]. Notably, conditioned media from MSC-NTFs was shown to protect neuroblastoma cell line against 6-OHDA neurotoxicity. This NTF-induced protection was also shown in a quinolinic acid model of neurotoxicity [48].

Conditioned media from MSC-NTFs was also shown to promote neuronal protection against oxidative stress and to inhibit proliferation of immune cells in response to multiple sclerosis (MS) related antigens [49]. Additionally, MSC-NTFs were shown to promote survival of retinal ganglion cells after optic nerve injury [50], to improve motor functions and to inhibit neuromuscular junction degeneration in a rat model of sciatic nerve injury [51], suggesting an enhanced neuroprotective effect.

The neuroprotective and neurogenic capacity of MSC-NTFs in these models, has encouraged the use of MSC-NTFs for autologous transplantation in patients with ALS. To date, a phase I/II and a phase 2a clinical trials for the treatment of ALS patients with MSC-NTFs was conducted by Brainstorm® Cell Therapeutics, and succeeded in meeting safety and efficacy criteria (See MSCs Therapy in Human Clinical Trials section).

Alternative ex vivo approach uses genetic manipulation of isolated hBM-MSCs to enhance the expression of NTFs. Using this strategy, hBM-MSCs over-expressing GDNF were shown to promote recovery in a 6-OHDA rat model of PD, resulting in reduction of amphetamine-induced rotations and rejuvenation of dopamine fibers [52]. Transplantation of hBM-MSCs over-expressing GDNF and VEGF were also used in a mutant Superoxide dismutase 1 (*SOD1*) rat model of ALS and showed improved neuro-muscular junction innervation and improved MN survival [53]. Similarly, transplantation of hBM-MSCs over-expressing BDNF promoted neurogenesis, improved behavioral scores, and increased lifespan in mice models of HD [54].

In summary, these lines of research validate the important role of NTFs in restoring homeostasis and halting degeneration, but also highlight the capability of hMSCs to serve as vectors for ectopic expression of beneficial factors through paracrine secretion as a therapeutic approach for nervous system disorders.

Immunomodulation and Neuroinflammation

Neuroinflammation refers to a variety of chronic, proinflammatory, immune system-mediated processes, mainly associated with neurodegenerative diseases. Cumulative evidence suggests that inflammation plays a major role in the progression of several neurodegenerative diseases. Post-mortem AD brains exhibit activated microglia and astrocytes as well as positive staining for multiple anti-inflammatory chemokines and cytokines [55]. PD entails enhanced microglia activation, astrogliosis, and lymphocyte infiltration [55] as well as increased levels of proinflammatory cytokines in the blood and cerebrospinal fluid (CSF) [56]. Also in ALS, accumulation of activated microglia and macrophages was shown next to degenerating areas, along with multiple proinflammatory compounds and upregulation of the proinflammatory cytochrome-C oxidases 1 and 2 (COX-1 and COX-2) [57]. Moreover, many inflammation-related compounds have detrimental roles on neurogenesis, consequently hampering endogenous tissue repair mechanisms [58]. Therefore, the modulation of the immune response toward an anti-inflammatory state emerges

Table 2. Ex vivo manipulated human mesenchymal stem cells in murine models of neurodegeneration

	Authors	Source	Ex vivo manipulation	Model animal	Suggested mechanism	Clinical improvement
Parkinson's	[18]	Bone marrow	NICD transduction and NTF induction	6-OHDA rats	GDNF secretion Dopamine secretion	Decreased apomorphine induced rotations
Parkinson's	[5]	Bone marrow	MSC-NTF	6-OHDA rats	NTF secretion	Decreased amphetamine induced rotations Reduced dopamine depletion Enhanced striatal regeneration
Parkinson's	[52]	Bone marrow	NICD and GDNF transduction	6-OHDA rats	GDNF secretion	Decreased amphetamine induced rotations Dopamine-fibers rejuvenation
Stroke	[17]	Bone marrow	Neuronal differentiation by NGN1 overexpression	Acute Ischemia rats	Enahnced endogenous neurogenesis MSC neuronal differentiation Anti inflammation	Improved motor recovery Reduced ischemic core
MS	[51]	Bone marrow	MSC-NTF	EAE mice	NTF secretion Immunomodulation	Increased survival Delayed symptom onset
MS	[79]	Adipose	IL-4 transduction	EAE mice	Induced Th2 immune response	Reduced disease severity
MS	[80]	Adipose	IL-10 transduction	EAE mice	Induced Th2 immune response	Reduced disease severity
ALS	[53]	Bone marrow	GDNF and VEGF transduction	SOD1 rats	NTF secretion	Increased survival Slower motor deterioration Improved NMJ innervation Enhanced motor neuron survival
Huntington	[48]	Bone marrow	MSC-NTF	QA-induced rats	NTF secretion	Reduced striatal atrophy Decreased apomorphine induced rotations
Huntington	[54]	Bone marrow	BDNF transduction	YAC128 and R6/2 mice	BDNF secretion	Increased survival Reduced anxiety behavior Decreased striatal atrophy Enhanced neurogenesis
Schiatic nerve injury	[51]	Bone marrow	MSC-NTF	Schiatic nerve crushed rats	Neurotrophic factor secretion	Improved motor functions Lateral reflex recovery Axonal regeneration Reduced NMJ degeneration
Optic nerve injury	[50]	Bone marrow	MSC-NTF	Optic nerve transectioned rats	NTF secretion	Increased survival of retinal ganglion cells

Abbreviations: EAE, experimental autoimmune encephalomyelitis; GDNF, glial cell line-derived neurotrophic factor; 6-OHDA, 6-hydroxydopamine; MSC-NTFs, neurotrophic factor-secreting mesenchymal stem cells; NICD, notch intracellular domain; NGN1, neurogenin 1; NMJ, neuromuscular junction; NTF, neurotrophic factors; QA, quinolinic acid; VEGF, vascular endothelial growth factor.

as a potential disease-modifying therapeutic strategy for neurodegeneration [59].

The inflammatory response in the CNS, unlike the rest of the body, is primarily mediated by the activation of microglia cells, specialized macrophages of the nervous system. Under physiological conditions, deactivated microglia support tissue homeostasis through production of neurotrophic and anti-inflammatory factors [55]. Upon activation, following exposure to pathogen or brain injury, activated microglia migrate along a chemotactic gradient, recruit circulating immune cells that infiltrate into the CNS through the BBB. Microglia cells are also able to perform phagocytosis and mediate neuroinflammation through secretion of proinflammatory cytokines, chemokines, and reactive oxygen species [58].

Ultimately, enhanced proinflammatory activation of microglia cells can result in chronic inflammation and accelerated neuronal death through oxidative stress and apoptosis. Several studies suggested that hMSCs hold various immunomodulatory roles, mainly through the manipulation of microglia-mediated neuroinflammation. hBM-MSCs transplantation into SOD1 female mice decreased microglial activation and astrogliosis which was associated with improved behavioral score [60]. In an acute model of AD, human umbilical cord blood-derived MSCs (hUCB-MSCs) also reduced the levels of microglial and astrocytic activation as well as apoptosis [61]. Following work showed that familial AD model APP/PS1 mice exhibit substantially increased levels of the proinflammatory cytokines tumor necrosis factor (TNF)-α and interleukin (IL)-1β when compared with wild-type mice and that the expression of these factors is significantly reduced following hUCB-MSCs transplantation. Alternatively, the expression of the anti-inflammatory markers IL-4, AMCase, YM-1, and Arg-1 was elevated following transplantation indicating a switch in microglial activation from a proinflammatory state to an anti-inflammatory [62].

It was demonstrated that hBM-MSCs promote secretion of IL-4 from microglia cells and stimulated α-synuclein clearance in a PD mouse model [63]. A 1-year follow-up clinical trial using these cell for the treatment of MSA revealed a higher IL-4 expression and a reduction in the α-synuclein levels in the CSF of hBM-MSCs-treated patients when compared with placebo group [63, 64] (See MSCs Therapy in Human Clinical Trials section). Importantly, hBM-MSCs-treated patients showed a lesser decrease in cerebral glucose metabolism and grey matter density as well as decreased deterioration of cognition when compared with placebo group. Finally, recent in vitro and in vivo analyses have indicated the roles of other molecular pathways, such as nuclear factor kappa-light-chain-enhancer of activated B cells signaling [65] and sphingosine-1-phosphate signaling [66] in hMSCs mediated of modulation of inflammation.

hMSCs were also shown to maintain BBB integrity. Following neuroinflammation, the BBB is disrupted in a process involving morphogenic and paracrine changes in astrocytes and endothelial cells, allowing increased penetrability and neutrophil infiltration [67–69]. Park et al. showed that hBM-MSCs were able to restore BBB integrity in lipopolysaccharide-induced rats, resulting in reduced neutrophil infiltration and enhanced neuronal survival. The authors suggested that improved BBB integrity was due to morphogenic changes in astrocytes and endothelial cells as well as anti-inflammatory modulation of microglia [70]. Similarly, Chung et al. reported that human adipose derived MSCs (hA-MSCs) transplantation following transient global cerebral ischemia in rats, minimized BBB disruption and neutrophil infiltration induced by ischemia and improved endothelial vasculture [71]. Together, these reports demonstrate another therapeutic advance for hMSCs following acute injury or chronic inflammation.

Immunomodulation and MS

MS is a chronic, autoimmune, and neurodegenerative disease of the CNS, in which immune cells, predominantly autoreactive CD4+ T-helper cells, infiltrate into the CNS and promote an inflammatory response, resulting in myelin injury and axonal loss [72]. Similarly to microglia directed therapy, current treatment strategies for MS commonly aim at modulating the immune response, through a shift from a proinflammatory response, mediated by cytokines secreted by T-helper 1 (Th1) cells, to an anti-inflammatory response, mediated by cytokines secreted by T-helper 2 (Th2) cells [72].

hMSCs were widely evaluated for the treatment of MS, through transplantation into the experimental autoimmune encephalomyelitis (EAE) mice model of MS. Following intravenous transplantation into rodents, hMSCs migrate from the blood stream into the CNS and localize to white matter demyelination sites, and to peripheral lymph organs and consequently induce EAE amelioration [41, 73–75]. Repeated reports ascribed this amelioration to a modulation of the immune response. Using hBM-MSCs, Bai et al. showed a decrease in leukocyte infiltration into the CNS and enhanced a Th2 cytokine profile, but also enhanced oligodendrogenesis, suggesting that hBM-MSCs affect immunomodulation but also enhanced neuronal repair [73]. Similar results showing a Th2 cytokine shift and enhanced oligodendrogenesis were reported using hA-MSCs [74, 76] and using human decidua-derived MSCs (hD-MSCs) [77]. hA-MSCs were also shown to express the immunosuppressive gene *human leukocyte antigen G (HLA-G)* [76] and to induce the secretion of the anti-inflammatory protein TNF-α-stimulated gene/protein 6 following culture with inflammatory cytokines [78]. Payne et al. tested the effect of hA-MSCs ex vivo modified to over-express the anti-inflammatory ILs-4 and −10 in EAE mice. In both cases, they observed disease attenuation and enhanced Th2 anti-inflammatory response, confirming the significance of immunomodulation in this model [79, 80].

HBM-MSCs-mediated improvement in MS was also attributed to NTFs secretion. Zhang et al. showed that EAE clinical score is correlated with decreased NGF expression, both rescued upon hBM-MSCs transplantation [41]. We previously showed that MSC-NTFs (discussed earlier) injected intracerebroventricularly (ICV) into EAE mice were able to ameliorate motor functions and suppress EAE in mice to a greater extent than naïve hBM-MSCs, suggesting neurotrophic-mediated disease amelioration [49].

Finally, hBM-MSCs differentiated into neural progenitor fate shown both enhanced immune-regulatory properties and NTFs secretion [81]. Multiple intrathecal injections of these cells into EAE mice reduced immune cell infiltration into the CNS and enhanced endogenous neural progenitor proliferation [81] and clinical trials using these cells are ongoing (ClinicalTrials.gov identifier: NCT01933802).

Protein Aggregate Clearance

The abnormal aggregation of proteins and formation of inclusion bodies is a major hallmark of neurodegenerative diseases [82], and protein aggregation is known to be a major factor in neurodegeneration onset. PD involves intraneuronal formation of inclusions in the SN termed Lewy bodies, mostly composed of the α-synuclein protein and triplication of the *SNCA* gene encoding α-synuclein was found to cause PD [83]. HD is characterized by expansion of CAG repeats in the N-terminus of the huntingtin gene, resulting in protein polyglutamination, leading to formation of fibers with β-sheet structure and aggregation. It was shown that the number of CAG repeats within the Huntingtin gene linearly correlates with early HD onset [84]. AD is associated with both extracellular amyloid plaques mostly containing the Aβ peptide, and intracellular neurofibrillary tangles containing the phosphorylated tau protein. According to the amyloid cascade hypothesis [85], the formation of amyloid plaques is essential for AD pathology. ALS involves the intraneuronal formation of inclusions, mostly in spinal MNs, containing TAR DNA-binding protein 43 (TDP-43), FUS and SOD1 proteins. Moreover, various prion diseases such as Creutzfeldt-Jakob disease and Gerstmann–Sträussler–Scheinker disease involve both intra- and extracellular deposition of abnormally folded prion proteins. Therefore, reducing abnormal protein aggregates by preventing aggregation as well as by imposed clearance of aggregating proteins is a main strategy for neurodegenerative disease therapy.

Microglia and Proteolytic Enzyme Secretion

As the main constituent of extracellular plaques in AD, Aβ levels are thought to dictate AD progression. Enhanced expression of Aβ-degrading enzymes such as insulin-degrading enzyme or neprilysin in AD mice models resulted in reduced Aβ burden and premature lethality rescue [86]. Moreover, the secretion of Aβ-degrading enzymes from microglia was shown to be affected by age [87] and regulated by ApoE [88], the two major risk factors for AD. Recent coculture experiments shown that soluble intracellular adhesion molecule-1 secreted by hUCB-MSCs induced the secretion of neprilysin from microglia [89]. Transplantation of these cells into APP/PS1 mice also shown induced neprilysin expression, alongside with reduced Aβ plaques in the hippocampus, demonstrating a role for hMSCs in enhancing the cells endogenous proteolytic machinery [89]. A phase I/II clinical trial based on these results is currently recruiting patients (ClinicalTrials.gov identifier: NCT02054208).

Autophagy

Autophagy is a cellular pathway involved in protein and organelle degradation through formation of autophagic vacuoles that fuse with lysosomes [90]. Expectedly, autophagy was shown to play a key role in aggregate clearance in several neurodegenerative disease models [91]. Knock-out of the essential autophagy gene Atg7 in mice was reported to cause accumulation of poly-ubiquitinated proteins, substantial neuronal loss and behavioral defects [92]. Shin et al. reported that ICV administration of hBM-MSCs into Aβ-inoculated mice increased the survival of hippocampal neurons and reduced the levels of Aβ. Aβ clearance was attributed to the recovery of the cells inherent clearance machinery through the autophagy-lysosomal pathway [93], as seen by formation of autophagic vacuoles and induction of the expression of the autophagy initiator BECN1. This group has also shown that hBM-MSCs-mediated activation of autophagy improved viability and reduced α-synuclein accumulation in the midbrains of MPTP-treated mouse model of PD [94]. Together, these findings indicate that hBM-MSCs are able to induce protein aggregate clearance through an increase in autophagy.

MSCs Therapy in Human Clinical Trials

Along with the advancements in murine models of neurodegeneration, hMSCs are widely examined as therapeutic agents in human clinical trials aiming to ameliorate neurodegeneration and acute brain injury as well as various other pathologies. To date, over 700 clinical trials using hMSCs are listed in clinicaltrials.gov, out of which more than 40 have reached phase III. hMSCs were reported safe for administration to the CNS through intravenous and intrathecal transplantation and were studied in the context of multiple neurodegenerative diseases and acute brain injuries. In this section, we will review current results using hMSCs-based therapy in human clinical trials (summarized in Table 3).

Substantial progress have been done using hMSCs for the treatment of MS. Li et al. demonstrated that hUCB-MSCs derived from healthy donors and administered into MS patients were able to ameliorate disease symptoms and reduce relapse occurrence. The immunomodulatory beneficial outcome was evident as a shift in peripheral blood cytokine expression toward a Th2 response [95]. Immunomodulation was also recorded following hBM-MSCs transplantation in MS and ALS patients [96]. Peripheral blood monocyte analysis 24 hours after transplantation revealed an increase in the proportion of immunosuppressive CD4+ CD25+ regulatory T cells and a decrease in the proportion of proinflammatory myeloid dendritic cells. Expanded disability status scale scores of hBM-MSCs-treated MS patients showed functional improvement 6 months following transplantation, while ALS functional rating scale (ALS-FRS) scores in ALS patients remained stable throughout this period, indicating immunomodulatory-mediated clinical potential for hBM-MSCs. A work by Mohajeri et al. has also reported a similar immunomodulatory effect following autologous transplantation of hBM-MSCs in six MS patients [97]. Six months following transplantation, all patients demonstrated clinical stability. They have also demonstrated a significant upregulation in the expression of the transcription factor FoxP3 in blood mononuclear cells. As Foxp3 is known as a marker of immunosuppressive CD4+ CD25+ regulatory T cells, this study further indicate the anti-inflammatory role of hBM-MSCs [98]. In another clinical study, Connick et al. administered autologous hBM-MSCs intravenously to 10 patients with secondary progressive MS with clinical evidence of optic nerve involvement. After hBM-MSCs treatments, patients showed improved visual acuity and an increase in optic nerve area, providing evidence that hBM-MSCs treatment can affect disease score in progressive MS [99].

hBM-MSCs were also beneficial following autologous injection into ALS patients. Mazzini et al. autologously administered hMSCs into the spinal cord in seven ALS patients and

Table 3. Human mesenchymal stem cells in human clinical trials

	Authors	Cell source	Administration	Number of patients enrolled	Clinical outcome
MS	[95]	Allogenic, Umbilical cord blood	Intravenous injection	13 in treatment group, 10 in control group	Improved functional performance Reduced relapse occurrence Enhanced Th2 response
MS and ALS	[98]	Autologous, Bone marrow	Intathecal and Intravenous injections.	15 in MS treatment group, 19 in ALS treatment group	Improved functional performance in MS patients Slower disease progression rate in ALS patients Enhanced anti-inflammatory response
MS	[97]	Autologous, Bone marrow	Intrathecal injection	7 patients in the treatment group	Clinical stability Enhanced peripheral blood FoxP3 expression
MS	[99]	Autologous, Bone marrow	Intravenous injection	10 patients in the treatment group	Improved visual acuity Increased optic nerve area
MSA	[36]	Autologous, Bone marrow	Intra-arterial and Intravenous injections	11 in treatment group, 16 in placebo group	Smaller increase in clinical severity. Reduced size of glucose hypo-metabolism areas.
Parkinson's	[106]	Autologous, Bone marrow	Stereotactic injection into the lateral ventricles	7 patients in the treatment group	Steady improvement in clinical score in 3/7 patients
Stroke	[3]	Autologous, Bone marrow	Intravenous injection	5 in treatment group, 25 in control group	Decreased brain atrophy Improved functional performance
Stroke	[61]	Autologous, Bone marrow	Intravenous injection	16 in treatment group, 36 in control group	Reduced mortality Improved functional performance
Stroke	[105]	Autologous, Bone marrow	Intravenous injection	12 patients in the treatment group	Improved neurological scores Reduced infarct size
ALS	[101]	Autologous, Bone marrow	Intrathecal injection	8 patients in the treatment group	Slower disease progression rate Elevated anti-inflammatory markers
ALS	[100]	Autologous, Bone marrow	Intrathecal injection	7 patients in the treatment group	Slower disease progression rate
ALS	[103]	Autologous, Bone marrow, ex vivo differentiated	Intramuscular and intrathecal injections.	26 patients in the treatment group	Slower disease progression rate
ALS	Brainstorm Cell therapeutics	Autologous, Bone marrow, ex vivo differentiated	Intramuscular and intrathecal injections.	36 in treatment group, 12 in control group	Decreased inflammatory markers Slower disease progression rate

reported a significant slowing down in the force vital capacity (FVC) scale in four of the patients [100]. Similar improvement in clinical score was also reported by Oh et al. following autologous hBM-MSCs transplantation [101]. A parallel work by this group has also analyzed the relationship between the expression of factors induced by hBM-MSCs and the patient's responsiveness to therapy [102]. They have shown that enhanced secretion of trophic factors such as VEGF, angiogenin, and transforming growth factor β were associated with responsiveness to treatment, providing evidence to the contribution of these factors.

As mentioned earlier, MSC-NTFs developed in our laboratory have also shown beneficial evidence in ALS patients. In a phase I/II study performed by Brainstorm Cell Therapeutics, 87% of the patients treated intrathecally with MSC-NTFs (NurOwn®) showed at least 25% improvement in the slope of disease progression at 6 months after treatment, as indicated by the ALS-FRS and the FVC measurements [103].

Recently, Brainstorm Cell Therapeutics announced findings from a randomized, double blind, placebo-controlled phase II study showing that autologous MSC-NTFs treatment in 36 patients indeed led to clinically meaningful benefit (http://ir.brainstorm-cell.com/phoenix.zhtml?c=142287&p=irol-newsArticle&ID=2186054). Patients treated with NurOwn® showed enhanced levels of NTFs in the CSF and decreased inflammatory markers 2 weeks after intrathecal transplantation. Importantly, these patients exhibited slower disease progression when compared with the placebo group, as measured by the ALS-FRS scale. As announced by the company, a phase III clinical trial is expected to begin in 2017.

Several clinical trials have also demonstrated the potential of hMSCs for the treatment of patients following ischemic stroke. Patients subjected to autologous hBM-MSCs transplantation 6 weeks following infarction within the middle cerebral artery territory, showed decreased brain atrophy and improved daily performance, as measured by the Barthel scale [3]. A follow-up study by this group has reported that hBM-MSCs treated group had lower mortality rate and tended toward improved clinical outcome 5 years after transplantation [104]. In a different trial, autologous hBM-MSCs transplantation following stroke was reported to reduce infarct size by 20% a week following transplantation [105].

Finally, autologous transplantation of hBM-MSCs was also performed in patients with synucleopathy. Venkataramana et al. reported that three of the seven PD patients treated by stereotactic injection of these cells into the sublateral ventricular zone (VZ) have shown improvement in their Unified Parkinson's Disease Rating Scale score [106]. Lee et al. have shown that intra-arterial and intravenous administration of hBM-MSCs to MSA patients resulted in a lesser increase in the severity of their neurological deficits when compared with placebo group, throughout a period of more than 3 years [64].

Although future research will have to determine the efficacy of these treatments in larger cohorts, these clinical results collectively demonstrate the beneficial potential of hMSCs-based therapy in patients with neurodegeneration.

Efficacy and Limitations of hMSCs-Based Therapy

hMSCs are regarded as efficient cell source for therapy, as they can be safely harvested from and transplanted into donors or patients, have no major ethical concerns, have low immunogenicity and possess a wide therapeutic potential. The results from both preclinical and clinical trials reviewed here indicate the potential of hMSC-based treatment in meeting several key aspects of neurodegeneration, such as neuroprotection, immunomodulation, and protein aggregate clearance. However, meaningful improvement of neurodegeneration would probably require a highly efficient and specific treatment throughout a long period of time.

As hMSCs can be harvested from various tissues, one measure for enhancing the efficiency of hMSCs-based therapy is by revealing the clinical relevance of hMSCs derived from various tissues. hMSCs derived from bone marrow and adipose show similar differentiation potential and expression of common hMSCs surface markers such as CD34, CD44, CD45, CD105, CD29, and CD90 [107–109]. However, recent comparative analyses have indicated variable protein secretion patterns [110, 111] and identified additional markers that differentially express in hMSCs derived from various origins (reviewed in ref. 112). Notably, hA-MSCs were reported to have a higher proliferative capacity over hBM-MSCs and to express a wider range of chemokine receptors, which are considered important for their homing capacity [107–119, 113]. hA-MSCs were also previously reported to possess a higher immunomodulatory potential [108], while a recent report using hA-MSCs and hBM-MSCs from the same donor has reported no major differences in their aspect [114].

Notably, recent publications comparing the administration of hMSCs derived from bone marrow or adipose tissue in mice models of hind limb ischemia have shown mixed results in terms of which source is superior [109, 115]. In a murine model of Crohn's disease, hA-MSCs were reported superior over hBM-MSCs in promoting an anti-inflammatory response [116]. However, there is lack of direct evidence for superiority of specific tissue-derived hMSCs for the treatment of neurodegenerative diseases. As the variation between hMSCs derived from different tissues may result in heterogeneous clinical outcomes, future studies should be performed to determine the relevant hMSCs sources.

Enhancing clinical efficacy for hMSCs-based treatment might also be achieved by extending hMSCs survival following transplantation. hMSCs survival in the CNS following transplantation was repeatedly reported to be limited up to several months in rodent models [18, 19, 33, 44, 74] with one report indicating survival after 45 weeks [41] and some reporting survival of only several days following transplantation [31, 43]. hMSCs survival following transplantation was shown to be affected by several factors including an immune response against the grafted cells [117], hypoxic/ischemic stress [118] ,or by an oxidative environment in site of damage [119]. Several methods, such as growth factor preconditioning, or pretreatment exposure to oxidative stress, were suggested for enhancing hMSCs survival in vivo following transplantation into the ischemic myocardium (reviewed in ref. 120). However, the factors associated with hMSCs survival in the CNS were not identified yet.

As described earlier, one promising mean of enhancing the beneficial outcome of hMSCs-based therapy is by enhancing the mechanisms throughout which hMSCs function was exerted. This was demonstrated through ex vivo differentiation or by genetically engineering of hMSCs to enhance NTFs

or immunomodulatory molecules expression. Similarly, hMSCs were suggested as a system to deliver shRNA into lesion area. Olson et al. have shown that hMSCs transduced to express shRNA against the huntingtin gene were able to reduce its expression in coculture neurons [121].

Finally, a potential clinical benefit is expected from the combination of hMSCs-based therapy with other novel therapeutic agents. To date, several compounds have shown a synergistic effect upon combined treatment with hMSCs in models of stroke and dementia [122–125] and combined transplantation of multiple cell-type is clinically examined in patients with stroke [126, 127].

Conclusion

Upon transplantation into rodent models as well as patients with neurodegenerative diseases, hMSCs are repeatedly reported to home toward lesion sites and secrete a broad range of molecules that modulate various aspects of diseases. Initially, hMSCs broad differentiation capacity was intended to be used for the derivation of neurons for cell regeneration of damaged tissue. However, limited efficacy for the derivation of neuronal cell types using this approach has motivated the use of hMSCs differentiation capacity for the establishment of supportive cells providing neuroprotection for damaged tissue. This concept is well demonstrated by our group's reports on the beneficial effect of NTF-secreting hMSCs in various models of neurodegeneration as well as in ALS patients. As different neurodegenerative diseases entail abnormalities in various molecular pathways and different cell types, it is expected that the trophic array of molecules essential to support neuronal and glial function may vary as well. While many favorable factors affecting substantial properties of disease have been identified, the potential of this approach is only beginning to unravel, as clinical trials using these cells meet safety criteria. We envisage that the use of hMSCs as vehicles for delivery of therapeutics into lesion area will provide an efficient platform for targeted therapy in various neurodegenerative diseases. Therefore, it seems promising that hMSCs will serve as efficient tools for neurodegenerative disease therapy in the future.

Author Contributions

R.V.: manuscript writing; D.O.: manuscript writing, final approval of manuscript.

Disclosure of Potential Conflicts of Interest

The authors indicate no potential conflicts of interest.

References

1 Pittenger MF, Mackay AM, Beck SC et al. Multilineage potential of adult human mesenchymal stem cells. Science 1999;284:143–147. Available at: http://www.ncbi.nlm.nih.gov/pubmed/10102814. Accessed January 15, 2017.

2 Salem HK, Thiemermann C. Mesenchymal stromal cells: Current understanding and clinical status. Stem Cells 2010;28:585–596. doi:10.1002/stem.269.

3 Bang OY, Lee JS, Lee PH et al. Autologous mesenchymal stem cell transplantation in stroke patients. Ann Neurol 2005;57:874–882. doi:10.1002/ana.20501.

4 Ra JC, Shin IS, Kim SH et al. Safety of intravenous infusion of human adipose tissue-derived mesenchymal stem cells in animals and humans. Stem Cells Dev 2011;20:1297–1308. doi:10.1089/scd.2010.0466.

5 Sadan O, Melamed E, Offen D. Bone-marrow-derived mesenchymal stem cell therapy for neurodegenerative diseases. Expert Opin Biol Ther 2009;9:1487–1497. doi:10.1517/14712590903321439.

6 Hellmann MA, Panet H, Barhum Y et al. Increased survival and migration of engrafted mesenchymal bone marrow stem cells in 6-hydroxydopamine-lesioned rodents. Neurosci Lett 2006;395:124–128. doi:10.1016/j.neulet.2005.10.097.

7 Liang X, Ding Y, Zhang Y et al. Paracrine mechanisms of mesenchymal stem cell-based therapy: Current status and perspectives. Cell Transpl 2014;23:1045–1059. doi:10.3727/096368913X667709.

8 Gorman AM. Neuronal cell death in neurodegenerative diseases: Recurring themes around protein handling. J Cell Mol Med 2008;12:2263–2280. doi:10.1111/j.1582-4934.2008.00402.x.

9 Woodbury D, Schwarz EJ, Prockop DJ et al. Adult rat and human bone marrow stromal cells differentiate into neurons. J Neurosci Res 2000;61:364–370. doi:10.1002/1097-4547(20000815)61:4<364::AID-JNR2>3.0.CO;2-C.

10 Wislet-Gendebien S, Hans G, Leprince P et al. Plasticity of cultured mesenchymal stem cells: Switch from nestin-positive to excitable neuron-like phenotype. Stem Cells 2005;23:392–402. doi:10.1634/stemcells.2004-0149.

11 Cho KJ, Trzaska KA, Greco SJ et al. Neurons derived from human mesenchymal stem cells show synaptic transmission and can be induced to produce the neurotransmitter substance P by interleukin-1α. Stem Cells 2005;23:383–391. doi:10.1634/stemcells.2004-0251.

12 Trzaska KA, Kuzhikandathil EV, Rameshwar P. Specification of a dopaminergic phenotype from adult human mesenchymal stem cells. Stem Cells 2007;25:2797–2808. doi:10.1634/stemcells.2007-0212.

13 Barzilay R, Kan I, Ben-Zur T et al. Induction of human mesenchymal stem cells into dopamine-producing cells with different differentiation protocols. Stem Cells Dev 2008;17:547–554. doi:10.1089/scd.2007.0172.

14 Liang J, Wu S, Zhao H et al. Human umbilical cord mesenchymal stem cells derived from Wharton's jelly differentiate into cholinergic-like neurons in vitro. Neurosci Lett 2013;532:59–63. doi:10.1016/j.neulet.2012.11.014.

15 Liu X, Li D, Jiang D et al. Acetylcholine secretion by motor neuron-like cells from umbilical cord mesenchymal stem cells. Neural Regen Res 2013;8:2086–2092. doi:10.3969/j.issn.1673-5374.2013.22.008.

16 Barzilay R, Ben-Zur T, Bulvik S et al. Lentiviral delivery of *LMX1a* enhances dopaminergic phenotype in differentiated human bone marrow mesenchymal stem cells. Stem Cells Dev 2009;18:591–602. doi:10.1089/scd.2008.0138.

17 Kim S-S, Yoo S-W, Park T-S et al. Neural induction with neurogenin1 increases the therapeutic effects of mesenchymal stem cells in the ischemic brain. Stem Cells 2008;26:2217–2228. doi:10.1634/stemcells.2008-0108.

18 Dezawa M, Kanno H, Hoshino M et al. Specific induction of neuronal cells from bone marrow stromal cells and application for autologous transplantation. J Clin Invest 2004;113:1701–1710. doi:10.1172/JCI20935.

19 Lu J, Moochhala S, Moore X-L et al. Adult bone marrow cells differentiate into neural phenotypes and improve functional recovery in rats following traumatic brain injury. Neurosci Lett 2006;398:12–17. doi:10.1016/j.neulet.2005.12.053.

20 Yang H, Xie Z, Wei L et al. Human umbilical cord mesenchymal stem cell-derived neuron-like cells rescue memory deficits and reduce amyloid-beta deposition in an AβPP/PS1 transgenic mouse model. Stem Cell Res Ther 2013;4:76. doi:10.1186/scrt227.

21 Maltman DJ, Hardy SA, Przyborski SA. Role of mesenchymal stem cells in neurogenesis and nervous system repair. Neurochem Int 2011;59:347–356. doi:10.1016/j.neuint.2011.06.008.

22 Neirinckx V, Coste C, Rogister B et al. Concise review: Adult mesenchymal stem cells, adult neural crest stem cells, and therapy of neurological pathologies: A state of play. Stem Cells Transl Med 2013;2:284–296. doi:10.5966/sctm.2012-0147.

23 Arvidsson A, Collin T, Kirik D et al. Neuronal replacement from endogenous

precursors in the adult brain after stroke. Nat Med 2002;8:963–970. doi:10.1038/nm747.
24 Winner B, Kohl Z, Gage FH. Neurodegenerative disease and adult neurogenesis. Eur J Neurosci 2011;33:1139–1151. doi:10.1111/j.1460-9568.2011.07613.x.
25 Siegel GJ, Chauhan NB. Neurotrophic factors in Alzheimer's and Parkinson's disease brain. Brain Res Rev 2000;33:199–227. doi:10.1016/S0165-0173(00)00030-8.
26 Zigova T, Pencea V, Wiegand SJ et al. Intraventricular administration of BDNF increases the number of newly generated neurons in the adult olfactory bulb. Mol Cell Neurosci 1998;11:234–245. doi:10.1006/mcne.1998.0684.
27 Rossi C, Angelucci A, Costantin L et al. Brain-derived neurotrophic factor (BDNF) is required for the enhancement of hippocampal neurogenesis following environmental enrichment. Eur J Neurosci 2006;24:1850–1856. doi:10.1111/j.1460-9568.2006.05059.x.
28 Fabel K, Fabel K, Tam B et al. VEGF is necessary for exercise-induced adult hippocampal neurogenesis. Eur J Neurosci 2003;18:2803–2812. doi:10.1111/j.1460-9568.2003.03041.x.
29 Kobayashi T, Ahlenius H, Thored P et al. Intracerebral infusion of glial cell line-derived neurotrophic factor promotes striatal neurogenesis after stroke in adult rats. Stroke 2006;37:2361–2367. doi:10.1161/01.STR.0000236025.44089.e1.
30 Ghosh A, Greenberg ME. Distinct roles for bFGF and NT-3 in the regulation of cortical neurogenesis. Neuron 1995;15:89–103. doi:10.1016/0896-6273(95)90067-5.
31 Munoz JR, Stoutenger BR, Robinson AP et al. Human stem/progenitor cells from bone marrow promote neurogenesis of endogenous neural stem cells in the hippocampus of mice. Proc Natl Acad Sci USA 2005;102:18171–18176. doi:10.1073/pnas.0508945102.
32 Kan I, Barhum Y, Melamed E et al. Mesenchymal stem cells stimulate endogenous neurogenesis in the subventricular zone of adult mice. Stem Cell Rev Reports 2011;7:404–412. doi:10.1007/s12015-010-9190-x.
33 Segal-Gavish H, Karvat G, Barak N et al. Mesenchymal stem cell transplantation promotes neurogenesis and ameliorates autism related behaviors in BTBR mice. Autism Res 2016;9:17–32. doi:10.1002/aur.1530.
34 Cova L, Armentero M-T, Zennaro E et al. Multiple neurogenic and neurorescue effects of human mesenchymal stem cell after transplantation in an experimental model of Parkinson's disease. Brain Res 2010;1311:12–27. doi:10.1016/j.brainres.2009.11.041.
35 Bao X, Wei J, Feng M et al. Transplantation of human bone marrow-derived mesenchymal stem cells promotes behavioral recovery and endogenous neurogenesis after cerebral ischemia in rats. Brain Res 2011;1367:103–113. doi:10.1016/j.brainres.2010.10.063.
36 Park H-J, Shin JY, Lee BR et al. Mesenchymal stem cells augment neurogenesis in the subventricular zone and enhance differentiation of neural precursor cells into dopaminergic neurons in the Substantia Nigra of a Parkinsonian model. Cell Transplant 2012;21:1629–1640. doi:10.3727/096368912X640556.
37 Oh SH, Kim HN, Park H-J et al. Mesenchymal stem cells increase hippocampal neurogenesis and neuronal differentiation by enhancing the Wnt signaling pathway in an Alzheimer's disease model. Cell Transpl 2015;24:1097–1109. doi:10.3727/096368914X679237.
38 Acsadi G, Anguelov RA, Yang H et al. Increased survival and function of SOD1 mice after glial cell-derived neurotrophic factor gene therapy. Hum Gene Ther 2002;13:1047–1059. doi:10.1089/104303402753812458.
39 Kromer LF. Nerve growth factor treatment after brain injury prevents neuronal death. Science 1987;235:214–216. Available at: http://www.ncbi.nlm.nih.gov/pubmed/3798108. Accessed January 11, 2017.
40 Fischer W, Wictorin K, Björklund A et al. Amelioration of cholinergic neuron atrophy and spatial memory impairment in aged rats by nerve growth factor. Nature 1987;329:65–68. doi:10.1038/329065a0.
41 Zhang J, Li Y, Lu M et al. Bone marrow stromal cells reduce axonal loss in experimental autoimmune encephalomyelitis mice. J Neurosci Res 2006;84:587–595. doi:10.1002/jnr.20962.
42 Pan H-C, Cheng F-C, Chen C-J et al. Post-injury regeneration in rat sciatic nerve facilitated by neurotrophic factors secreted by amniotic fluid mesenchymal stem cells. J Clin Neurosci 2007;14:1089–1098. doi:10.1016/j.jocn.2006.08.008.
43 Wakabayashi K, Nagai A, Sheikh AM et al. Transplantation of human mesenchymal stem cells promotes functional improvement and increased expression of neurotrophic factors in a rat focal cerebral ischemia model. J Neurosci Res 2009;88:NA–NA. doi:10.1002/jnr.22279.
44 Zhang M-J, Sun J-J, Qian L et al. Human umbilical mesenchymal stem cells enhance the expression of neurotrophic factors and protect ataxic mice. Brain Res 2011;1402:122–131. doi:10.1016/j.brainres.2011.05.055.
45 Cui Y, Ma S, Zhang C et al. Human umbilical cord mesenchymal stem cells transplantation improves cognitive function in Alzheimer's disease mice by decreasing oxidative stress and promoting hippocampal neurogenesis. Behav Brain Res 2017;320:291–301. doi:10.1016/j.bbr.2016.12.021.
46 Blondheim NR, Levy YS, Ben-Zur T et al. Human mesenchymal stem cells express neural genes, suggesting a neural predisposition. Stem Cells Dev 2006;15:141–164. doi:10.1089/scd.2006.15.141.
47 Sadan O, Bahat-Stromza M, Barhum Y et al. Protective effects of neurotrophic factor-secreting cells in a 6-OHDA rat model of Parkinson disease. Stem Cells Dev 2009;18:1179–1190. doi:10.1089/scd.2008.0411.
48 Sadan O, Shemesh N, Barzilay R et al. Mesenchymal stem cells induced to secrete neurotrophic factors attenuate quinolinic acid toxicity: A potential therapy for Huntington's disease. Exp Neurol 2012;234:417–427. doi:10.1016/j.expneurol.2011.12.045.
49 Barhum Y, Gai-Castro S, Bahat-Stromza M et al. Intracerebroventricular transplantation of human mesenchymal stem cells induced to secrete neurotrophic factors attenuates clinical symptoms in a mouse model of multiple sclerosis. J Mol Neurosci 2010;41:129–137. doi:10.1007/s12031-009-9302-8.
50 Levkovitch-Verbin H, Sadan O, Vander S et al. Intravitreal injections of neurotrophic factors secreting mesenchymal stem cells are neuroprotective in rat eyes following optic nerve transaction. Investig Ophthalmol Vis Sci 2010;51:6394–6400. doi:10.1167/iovs.09-4310.
51 Dadon-Nachum M, Sadan O, Srugo I et al. Differentiated mesenchymal stem cells for sciatic nerve injury. Stem Cell Rev 2011;7:664–671. doi:10.1007/s12015-010-9227-1.
52 Glavaski-Joksimovic A, Virag T, Mangatu TA et al. Glial cell line-derived neurotrophic factor-secreting genetically modified human bone marrow-derived mesenchymal stem cells promote recovery in a rat model of Parkinson's disease. J Neurosci Res 2010;88:2669–2681. doi:10.1002/jnr.22435.
53 Krakora D, Mulcrone P, Meyer M et al. Synergistic effects of GDNF and VEGF on lifespan and disease progression in a familial ALS rat model. Mol Ther 2013;21:1602–1610. doi:10.1038/mt.2013.108.
54 Pollock K, Dahlenburg H, Nelson H et al. Human mesenchymal stem cells genetically engineered to overexpress brain-derived neurotrophic factor improve outcomes in Huntington's disease mouse models. Mol Ther 2016;24:965–977. doi:10.1038/mt.2016.12.
55 Glass CK, Saijo K, Winner B et al. Mechanisms underlying inflammation in neurodegeneration. Cell 2010;140:918–934. doi:10.1016/j.cell.2010.02.016.
56 Nagatsu T, Sawada M. Inflammatory process in Parkinson's disease: Role for cytokines. Curr Pharm Des 2005;11:999–1016. Available at: http://www.ncbi.nlm.nih.gov/pubmed/15777250. Accessed January 9, 2017.
57 McGeer PL, McGeer EG. Inflammatory processes in amyotrophic lateral sclerosis. Muscle Nerve 2002;26:459–470. doi:10.1002/mus.10191.
58 Russo I, Barlati S, Bosetti F. Effects of neuroinflammation on the regenerative capacity of brain stem cells. J Neurochem 2011;116:947–956. doi:10.1111/j.1471-4159.2010.07168.x.
59 Gao H-M, Hong J-S. Why neurodegenerative diseases are progressive: Uncontrolled inflammation drives disease progression. Trends Immunol 2008;29:357–365. doi:10.1016/j.it.2008.05.002.
60 Vercelli A, Mereuta OM, Garbossa D et al. Human mesenchymal stem cell transplantation extends survival, improves motor performance and decreases neuroinflammation in mouse model of amyotrophic lateral sclerosis. Neurobiol Dis 2008;31:395–405. doi:10.1016/j.nbd.2008.05.016.
61 Lee HJ, Lee JK, Lee H et al. The therapeutic potential of human umbilical cord blood-derived mesenchymal stem cells in Alzheimer's disease. Neurosci Lett. 2010;481:30–35. doi:10.1016/j.neulet.2010.06.045. Epub 2010 Jun 19.
62 Lee HJ, Lee JK, Lee H et al. Human umbilical cord blood-derived mesenchymal stem cells improve neuropathology and cognitive impairment in an Alzheimer's disease mouse model through modulation of neuroinflammation. Neurobiol Aging 2012;33:588–602. doi:10.1016/j.neurobiolaging.2010.03.024.
63 Park HJ, Oh SH, Kim HN et al. Mesenchymal stem cells enhance α-synuclein

clearance via M2 microglia polarization in experimental and human parkinsonian disorder. Acta Neuropathol 2016;132:685–701. doi:10.1007/s00401-016-1605-6.
64 Lee PH, Lee JE, Kim H-S et al. A randomized trial of mesenchymal stem cells in multiple system atrophy. Ann Neurol 2012;72:32–40. doi:10.1002/ana.23612.
65 Wang H, Nagai A, Sheikh AM et al. Human mesenchymal stem cell transplantation changes proinflammatory gene expression through a nuclear factor-κB-dependent pathway in a rat focal cerebral ischemic model. J Neurosci Res 2013;91:1440–1449. doi:10.1002/jnr.23267.
66 Marfia G, Navone SE, Hadi LA et al. The adipose mesenchymal stem cell secretome inhibits inflammatory responses of microglia: Evidence for an involvement of sphingosine-1-phosphate signalling. Stem Cells Dev 2016; 25:1095–1107. doi:10.1089/scd.2015.0268.
67 Willis CL, Nolan CC, Reith SN et al. Focal astrocyte loss is followed by microvascular damage, with subsequent repair of the blood-brain barrier in the apparent absence of direct astrocytic contact. Glia 2004;45: 325–337. doi:10.1002/glia.10333.
68 Gurney KJ, Estrada EY, Rosenberg GA. Blood-brain barrier disruption by stromelysin-1 facilitates neutrophil infiltration in neuroinflammation. Neurobiol Dis 2006;23:87–96. doi:10.1016/j.nbd.2006.02.006.
69 Hawkins BT, Davis TP. The blood-brain barrier/neurovascular unit in health and disease. Pharmacol Rev 2005;57:
70 Park HJ, Shin JY, Kim HN et al. Mesenchymal stem cells stabilize the blood–brain barrier through regulation of astrocytes. Stem Cell Res Ther 2015;6:187. doi:10.1186/s13287-015-0180-4.
71 Chung TN, Kim JH, Choi BY et al. Adipose-derived mesenchymal stem cells reduce neuronal death after transient global cerebral ischemia through prevention of blood-brain barrier disruption and endothelial damage. Stem Cells Transl Med 2015;4:178–185. doi: 10.5966/sctm.2014-0103.
72 Neuhaus O, Archelos JJ, Hartung H-P. Immunomodulation in multiple sclerosis: From immunosuppression to neuroprotection. Trends Pharmacol Sci 2003;24:131–138. doi:10.1016/S0165-6147(03)00028-2.
73 Bai L, Lennon DP, Eaton V et al. Human bone marrow-derived mesenchymal stem cells induce Th2-polarized immune response and promote endogenous repair in animal models of multiple sclerosis. Glia 2009;57: 1192–1203. doi:10.1002/glia.20841.
74 Constantin G, Marconi S, Rossi B et al. Adipose-derived mesenchymal stem cells ameliorate chronic experimental autoimmune encephalomyelitis. Stem Cells 2009;27:2624–2635. doi:10.1002/stem.194.
75 Gordon D, Pavlovska G, Uney JB et al. Human mesenchymal stem cells infiltrate the spinal cord, reduce demyelination, and localize to white matter lesions in experimental autoimmune encephalomyelitis. J Neuropathol Exp Neurol 2010;69:1087–1095. doi: 10.1097/NEN.0b013e3181f97392.
76 Shalaby SM, Sabbah NA, Saber T et al. Adipose-derived mesenchymal stem cells modulate the immune response in chronic experimental autoimmune encephalomyelitis model. IUBMB Life 2016;68:106–115. doi: 10.1002/iub.1469.
77 Bravo B, Gallego MI, Flores AI et al. Restrained Th17 response and myeloid cell infiltration into the central nervous system by human decidua-derived mesenchymal stem cells during experimental autoimmune encephalomyelitis. Stem Cell Res Ther 2016; 7:43. doi:10.1186/s13287-016-0304-5.
78 Fisher-Shoval Y, Barhum Y, Sadan O et al. Transplantation of placenta-derived mesenchymal stem cells in the EAE mouse model of MS. J Mol Neurosci 2012;48:176–184. doi:10.1007/s12031-012-9805-6.
79 Payne NL, Dantanarayana A, Sun G et al. Early intervention with gene-modified mesenchymal stem cells overexpressing interleukin-4 enhances anti-inflammatory responses and functional recovery in experimental autoimmune demyelination. Cell Adh Migr 2012;6:179–189. doi:10.4161/cam.20341.
80 Payne NL, Sun G, McDonald C et al. Human adipose-derived mesenchymal stem cells engineered to secrete IL-10 inhibit APC function and limit CNS autoimmunity. Brain Behav Immun 2013;30:103–114. doi:10.1016/j.bbi.2013.01.079.
81 Harris VK, Faroqui R, Vyshkina T et al. Characterization of autologous mesenchymal stem cell-derived neural progenitors as a feasible source of stem cells for central nervous system applications in multiple sclerosis. Stem Cells Transl Med 2012;1:536–547. doi: 10.5966/sctm.2012-0015.
82 Ross CA Poirier MA. Opinion: What is the role of protein aggregation in neurodegeneration? Nat Rev Mol Cell Biol 2005;6: 891–898. doi:10.1038/nrm1742.
83 Singleton AB, Farrer M, Johnson J et al. Synuclein locus triplication causes Parkinson's disease. Science 2003;302:841–841. doi: 10.1126/science.1090278.
84 Arrasate M, Finkbeiner S. Protein aggregates in Huntington's disease. Exp Neurol 2012;238:1–11. doi:10.1016/j.expneurol.2011.12.013.
85 Hardy JA, Higgins GA. Alzheimer's disease: The amyloid cascade hypothesis. Science 1992;256:184–185. Available at: http://www.ncbi.nlm.nih.gov/pubmed/1566067. Accessed January 15, 2017.
86 Leissring MA, Farris W, Chang AY et al. Enhanced proteolysis of beta-amyloid in APP transgenic mice prevents plaque formation, secondary pathology, and premature death. Neuron 2003;40:1087–1093. Available at: http://www.ncbi.nlm.nih.gov/pubmed/14687544. Accessed January 15, 2017.
87 Hickman SE, Allison EK, El Khoury J. Microglial dysfunction and defective -amyloid clearance pathways in aging Alzheimer's disease mice. J Neurosci 2008;28:8354–8360. doi:10.1523/JNEUROSCI.0616-08.2008.
88 Jiang Q, Lee CYD, Mandrekar S et al. ApoE promotes the proteolytic degradation of Aβ. Neuron 2008;58:681–693. doi:10.1016/j.neuron.2008.04.010.
89 Kim J-Y, Kim DH, Kim JH et al. Soluble intracellular adhesion molecule-1 secreted by human umbilical cord blood-derived mesenchymal stem cell reduces amyloid-β plaques. Cell Death Differ 2012;19:680–691. doi: 10.1038/cdd.2011.140.
90 Mizushima N, Levine B, Cuervo AM et al. Autophagy fights disease through cellular self-digestion. Nature 2008;451:1069–1075. doi:10.1038/nature06639.
91 Rubinsztein DC. The roles of intracellular protein-degradation pathways in neurodegeneration. Nature 2006;443:780–786. doi: 10.1038/nature05291.
92 Komatsu M, Wang QJ, Holstein GR et al. Essential role for autophagy protein Atg7 in the maintenance of axonal homeostasis and the prevention of axonal degeneration. Proc Natl Acad Sci USA 2007;104:14489–14494. doi:10.1073/pnas.0701311104.
93 Shin JY, Park HJ, Kim HN et al. Mesenchymal stem cells enhance autophagy and increase β-amyloid clearance in Alzheimer disease models. Autophagy 2014;10:32–44. doi:10.4161/auto.26508.
94 Park HJ, Shin JY, Kim HN et al. Neuroprotective effects of mesenchymal stem cells through autophagy modulation in a parkinsonian model. Neurobiol Aging 2014;35:1920–1928. doi: 10.1016/j.neurobiolaging.2014.01.028.
95 Li J-F, Zhang D-J, Geng T et al. The potential of human umbilical cord-derived mesenchymal stem cells as a novel cellular therapy for multiple sclerosis. Cell Transpl 2014;23:113–122. doi: 10.3727/096368914X685005.
96 Karussis D, Karageorgiou C, Vaknin-Dembinsky A et al. Safety and immunological effects of mesenchymal stem cell transplantation in patients with multiple sclerosis and amyotrophic lateral sclerosis. Arch Neurol 2010;67:1187–1194. doi:10.1001/archneurol.2010.248.
97 Mohajeri M, Farazmand A, Mohyeddin Bonab M et al. FOXP3 gene expression in multiple sclerosis patients pre- and post mesenchymal stem cell therapy. Iran J Allergy Asthma Immunol 2011;10:155. 61. doi: 010.03/ijaai.155161.
98 Buckner JH. Mechanisms of impaired regulation by CD4+CD25+FOXP3+ regulatory T cells in human autoimmune diseases. Nat Rev Immunol 2010;10:849–859. doi: 10.1038/nri2889.
99 Connick P, Kolappan M, Crawley C et al. Autologous mesenchymal stem cells for the treatment of secondary progressive multiple sclerosis: An open-label phase 2a proof-of-concept study. Lancet Neurol 2012;11:150–156. doi:10.1016/S1474-4422(11)70305-2.
100 Mazzini L, Mareschi K, Ferrero I et al. Autologous mesenchymal stem cells: Clinical applications in amyotrophic lateral sclerosis. Neurol Res 2006;28:523–526. doi:10.1179/016164106X116791.
101 Oh K-W, Moon C, Kim HY et al. Phase I trial of repeated intrathecal autologous bone marrow-derived mesenchymal stromal cells in amyotrophic lateral sclerosis. Stem Cells Transl Med 2015;4:590–597. doi:10.5966/sctm.2014-0212.
102 Kim HY, Kim H, Oh K-W et al. Biological markers of mesenchymal stromal cells as predictors of response to autologous stem cell transplantation in patients with amyotrophic lateral sclerosis: An investigator-initiated trial and in vivo study. Stem Cells 2014;32:2724–2731. doi:10.1002/stem.1770.
103 Petrou P, Gothelf Y, Argov Z 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;73:337–344. doi:10.1001/jamaneurol.2015.4321.
104 Lee JS, Hong JM, Moon GJ et al. A long-term follow-up study of intravenous autologous mesenchymal stem cell transplantation in patients with ischemic stroke. Stem Cells 2010;28: 1099–1106. doi:10.1002/stem.430.
105 Honmou O, Houkin K, Matsunaga T et al. Intravenous administration of auto serum-expanded autologous mesenchymal stem cells in stroke. Brain 2011;134:1790–1807. doi:10.1093/brain/awr063.
106 Venkataramana NK, Kumar SKV, Balaraju S et al. Open-labeled study of unilateral autologous bone-marrow-derived mesenchymal stem cell transplantation in Parkinson's disease. Transl Res 2010;155:62–70. doi:10.1016/j.trsl.2009.07.006.
107 Lee RH, Kim B, Choi I et al. Characterization and expression analysis of mesenchymal stem cells from human bone marrow and adipose tissue. Cell Physiol Biochem 2004;14:311–324. doi:10.1159/000080341.
108 Li C, Wu X, Tong J et al. Comparative analysis of human mesenchymal stem cells from bone marrow and adipose tissue under xeno-free conditions for cell therapy. Stem Cell Res Ther 2015;6:55. doi:10.1186/s13287-015-0066-5.
109 Bortolotti F, Ukovich L, Razban V et al. In vivo therapeutic potential of mesenchymal stromal cells depends on the source and the isolation procedure. Stem Cell Reports 2015; 4:332–339. doi:10.1016/j.stemcr.2015.01.001.
110 Jeon Y-J, Kim J, Cho JH et al. Comparative analysis of human mesenchymal stem cells derived from bone marrow, placenta, and adipose tissue as sources of cell therapy. J Cell Biochem 2016;117:1112–1125. doi: 10.1002/jcb.25395.
111 Pires AO, Mendes-Pinheiro B, Teixeira FG et al. Unveiling the differences of secretome of human bone marrow mesenchymal stem cells, adipose tissue-derived stem cells, and human umbilical cord perivascular cells: A proteomic analysis. Stem Cells Dev 2016; 25:1073–1083. doi:10.1089/scd.2016.0048.
112 Lv F-J, Tuan RS, Cheung KMC et al. Concise review: The surface markers and identity of human mesenchymal stem cells. Stem Cells 2014;32:1408–1419. doi:10.1002/stem.1681.
113 Ahmadian Kia N, Bahrami AR, Ebrahimi M et al. Comparative analysis of chemokine receptor's expression in mesenchymal stem cells derived from human bone marrow and adipose tissue. J Mol Neurosci 2011;44:178–185. doi:10.1007/s12031-010-9446-6.
114 Valencia J, Blanco B, Yáñez R et al. Comparative analysis of the immunomodulatory capacities of human bone marrow– and adipose tissue–derived mesenchymal stromal cells from the same donor. Cytotherapy 2016;18:1297–1311. doi:10.1016/j.jcyt.2016.07.006.
115 Kim Y, Kim H, Cho H et al. 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 2007; 20:867–876. doi:10.1159/000110447.
116 Xie M, Qin H, Luo Q et al. Comparison of adipose-derived and bone marrow mesenchymal stromal cells in a murine model of crohn's disease. Dig Dis Sci 2017;62:115–123. doi:10.1007/s10620-016-4166-6.
117 Ankrum J. A, Ong JF Karp JM. Mesenchymal stem cells: Immune evasive, not immune privileged. Nat Biotechnol 2014;32: 252–260. doi:10.1038/nbt.2816.
118 Deschepper M, Oudina K, David B et al. Survival and function of mesenchymal stem cells (MSCs) depend on glucose to overcome exposure to long-term, severe and continuous hypoxia. J Cell Mol Med 2011;15:1505–1514. doi: 10.1111/j.1582-4934.2010.01138.x.
119 Boopathy AV, Pendergrass KD, Che PL et al. Oxidative stress-induced Notch1 signaling promotes cardiogenic gene expression in mesenchymal stem cells. Stem Cell Res Ther 2013;4:43. doi:10.1186/scrt190.
120 Li Q, Wang Y Deng Z. Pre-conditioned mesenchymal stem cells: A better way for cell-based therapy. Stem Cell Res Ther 2013; 4:63. doi:10.1186/scrt213.
121 Olson SD, Kambal A, Pollock K et al. Examination of mesenchymal stem cell-mediated RNAi transfer to Huntington's disease affected neuronal cells for reduction of huntingtin. Mol Cell Neurosci 2012;49:271–281. doi:10.1016/j.mcn.2011.12.001.
122 Kong D, Liu Q, Xu G et al. Synergistic effect of tanshinone IIA and mesenchymal stem cells on preventing learning and memory deficits via anti-apoptosis, attenuating tau phosphorylation and enhancing the activity of central cholinergic system in vascular dementia. Neurosci Lett 2017;637:175–181. doi:10.1016/j.neulet.2016.11.024.
123 Zhang Q, Chen Z-W, Zhao Y-H et al. Bone marrow stromal cells combined with sodium ferulate and *n* -butylidenephthalide promote the effect of therapeutic angiogenesis via advancing astrocyte-derived trophic factors after ischemic stroke. Cell Transplant 2017;26:229–242. doi: 10.3727/096368916X693536.
124 Wang C, Li F, Guan Y et al. Bone marrow stromal cells combined with oxiracetam influences the expression of B-cell lymphoma 2 in rats with ischemic stroke. J Stroke Cerebrovasc Dis 2014;23:2591–2597. doi:10.1016/j.jstrokecerebrovasdis.2014.05.035.
125 Zhao Y, Lai W, Xu Y et al. Exogenous and endogenous therapeutic effects of combination sodium ferulate and bone marrow stromal cells (BMSCs) treatment enhance neurogenesis after rat focal cerebral ischemia. Metab Brain Dis 2013;28:655–666. doi: 10.1007/s11011-013-9425-z.
126 Chen L, Xi H, Huang H et al. Multiple cell transplantation based on an intraparenchymal approach for patients with chronic phase stroke. Cell Transplant 2013;22:83–91. doi:10.3727/096368913X672154.
127 Qiao L, Huang F, Zhao M et al. A two-year follow-up study of cotransplantation with neural stem/progenitor cells and mesenchymal stromal cells in ischemic stroke patients. Cell Transplant 2014;23:65–72. doi: 10.3727/096368914X684961.

