Review Article

Clinical Applications of Mesenchymal Stem Cells in Chronic Diseases

Andrea Farini, Clementina Sitzia, Silvia Erratico, Mirella Meregalli, and Yvan Torrente

Laboratorio di Cellule Staminali, Dipartimento di Fisiopatologia Medico-Chirurgica e dei Trapianti, Università degli Studi di Milano, Fondazione IRCCS Ca’ Granda Ospedale Maggiore Policlinico, Centro Dino Ferrari, Via F. Sforza 35, 20122 Milano, Italy

Correspondence should be addressed to Yvan Torrente; yvan.torrente@unimi.it

Received 4 December 2013; Revised 14 April 2014; Accepted 15 April 2014; Published 30 April 2014

Academic Editor: Katherine Athayde Teixeira de Carvalho

Copyright © 2014 Andrea Farini et al. This is an open access article distributed under the Creative Commons Attribution License, which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

Extraordinary progress in understanding several key features of stem cells has been made in the last ten years, including definition of the niche, and identification of signals regulating mobilization and homing as well as partial understanding of the mechanisms controlling self-renewal, commitment, and differentiation. This progress produced invaluable tools for the development of rational cell therapy protocols that have yielded positive results in preclinical models of genetic and acquired diseases and, in several cases, have entered clinical experimentation with positive outcome. Adult mesenchymal stem cells (MSCs) are nonhematopoietic cells with multilineage potential to differentiate into various tissues of mesodermal origin. They can be isolated from bone marrow and other tissues and have the capacity to extensively proliferate in vitro. Moreover, MSCs have also been shown to produce anti-inflammatory molecules which can modulate humoral and cellular immune responses. Considering their regenerative potential and immunoregulatory effect, MSC therapy is a promising tool in the treatment of degenerative, inflammatory, and autoimmune diseases. It is obvious that much work remains to be done to increase our knowledge of the mechanisms regulating development, homeostasis, and tissue repair and thus to provide new tools to implement the efficacy of cell therapy trials.

1. Introduction

Since the work of Friedenstein, that firstly described bone marrow- (BM-) derived stromal cells with the capacity of differentiation into bone [1], it was thought that nonhaematopoietic stem cell resided in the bone marrow, the so-called mesenchymal stem cells (MSCs) [2, 3]. The group of Caplan identified the MSCs from BM for the expression of the specific antigen markers CD105 and CD73 [4]. Pittenger defined the MSCs as multipotent stem cell with the ability to differentiate into adipose tissue, bone, and cartilage [5]. According to this multilineage differentiation potential, it was believed that MSCs mediated tissue and organ repair [6, 7]. However, further studies assessed that, following specific molecular cues, MSCs reached the site of injury and allowed the repair of tissues by means of the expression of different trophic factors [8–10]. In the last 20 years, MSCs were isolated from a wide range of tissues [11–14] and organs [15, 16]. Furthermore, it was demonstrated that under specific stimuli MSCs possessed an incredible capacity of transdifferentiation, developing in mesodermal (myocyte, osteocyte, endothelium, adipocyte, and cardiomyocyte), ectodermal (neuronal), and endodermal (hepatic, pancreatic, respiratory epithelium) lineages. In the presence of β-glycerol-phosphate, ascorbic acid-2-phosphate, dexamethasone, and fetal bovine serum, MSCs proliferated as osteoblasts. On the other side, when they were grown with a serum-free nutrient medium added with TFG-β or family-related molecules, MSCs proliferated as chondrocytes, expressing cartilage-specific extracellular matrix components [17]. Similarly, it could be possible to induce the formation of adipocytes by means of peroxisome proliferator-activated receptor-γ (PPAR-γ), fatty acid synthetase, and isobutylmethylxanthine while, on the contrary, IL-1 and TNF-α blocked MSCs-adipogenetic differentiation. As published by Barry and Murphy, MSCs differentiation into myoblasts was driven by
5-azacytidine and amphotericin B [17]. Recently, different works suggested that MSCs were strictly associated with vessels and possibly with pericytes, the perivascular cells that surround microvessels [18]. It was demonstrated that pericytes retained the ability to differentiate not only into osteoblasts, adipocytes, and fibroblasts but also into neural lineages if cultivated with bFGF [19] and into smooth muscle cells if stimulated with low concentration of oxygen [20]. It is well known that MSCs are able to express integrins, adhesion molecules, and chemokine receptors that regulate their capacity of migration and homing: CCR1, CCR4, CCR7, CCR10, and CXCR5 [4, 21]. Thanks to the expression of these molecules, MSCs can reach damaged tissues through endothelial cell layers and participate not only in tissue regeneration but also in BM microenvironment replenishment [22]. Stromal derived factor (SDF)-1 is associated with mobilization of stem cells into the periphery and homing to the site of injury [23, 24]: it was showed that in diverse tissue injuries SDF-1 functions as a MSCs chemotacticant [25–28]. According to these evidences, MSCs were evaluated in several studies for their safety and efficacy of transplantation. Studies published by Gao and Herrera confirmed the ability of MSCs to engraft into various organs following transplantation (liver, bone, and lung) [29, 30], while the groups of Jackson and Orlic successfully used them in the preparation of infarcted myocardium [31–33]. Furthermore, MSCs were noted to enhance angiogenesis in the myocardium [34] and also to allow the reduction of myocardial fibrotic area, probably due to their capacity of increasing the capillary density [35]. Hofstetter and colleagues demonstrated that MSCs exert their role also indirectly, enhancing the expression of growth factors that allowed the regeneration of damaged tissues [36]. However, further studies are necessary to better identify (i) all the molecules other than chemokines and adhesion molecules that drive MSCs to the site of injury; (ii) growth media to obtain reproducible culture techniques and to enhance safety of expanded MSCs; (iii) host responses to allogenic MSC therapy.

2. MSCs Isolation

Citofluorimetric analysis performed on MSCs showed that they express CD44, CD73, CD90, and CD105 receptors while lacking hematopoietic stem cell markers such as CD14, CD31, CD33, CD34, and CD45. Due to the absence of specific mesenchymal cell markers and the heterogeneity of the MSC populations, the Mesenchymal and Tissue Stem Cell Committee of the International Society for Cellular Therapy (ISCT) established three minimal criteria that MSCs isolated from human bone marrow and other mesenchymal tissues must have in vitro: (i) adherence to plastic in standard culture conditions, (ii) display of a specific surface antigen expression pattern (CD73+ CD90+ CD105+ CD34− CD45− CD11b− CD14− CD19− CD79a− HLA-DR−), and (iii) multipotency, that is, differentiation potential along the osteogenic, chondrogenic, and adipogenic lineages [37]. The heterogeneity of the MSC population is demonstrated by in vitro differentiation assays, where most of the population showed a differentiation potential towards the classical three cell types.

3. Immunomodulatory Effects of MSCs

Several studies have demonstrated that MSCs can inhibit cytotoxic T cells and natural killer (NK) cells [38, 39] by means of different pathways. MSCs can exert their immunomodulatory functions by secreting suppressors of T-cell development (TGFβ and hepatocyte growth factor (HGF)) [40] and proliferation such as leukemia inhibitory factor (LIF) [41] and IFN-γ [42]. Furthermore, MSCs can induce the expression of TNF-α and IL-1 leading to unbalanced secretion of chemokines and inducible nitric oxide (iNOS) [43]. More interestingly, the works of Spaggiari et al. [44] and Poggi et al. [45] showed that MSCs isolated from BM are not recognized by NKs as they express human leukocyte antigen (HLA) class I molecules. This way, MSCs were seen as the most feasible population of stem cells for cell transplantation experiments. Otherwise, recent studies demonstrated that MSCs were efficiently lysed by the cytotoxic immune effectors [39, 46]. The work of Jewett et al. showed that IL-2 treated NKs recognized and destroyed MSCs while IFN-γ had the opposite effect [47]. As the IFN-γ is secreted by monocytes, the authors postulated that these cells not only served as protector of MSCs but also allowed the differentiation of stem cells by NFκB dependent and independent pathways [47]. Giuliani et al. showed that MSCs expressed functional Toll-like receptors (TLR) that promote their proliferation and cytokine secretion [48]. They also identified a molecule, called MICA, that formed a complex with other immunoregulatory proteins and together with TLRs ligands protected MSCs against NKs aggression [48].

4. MSCs and Chronic Diseases

4.1. MSCs and Musculoskeletal Diseases. As for the other tissues previously described, MSCs were isolated from human adult skeletal muscle [49, 50]. In addition, Gonçalves described the ability of MSCs to complement dystrophin deficiency [51], and Németh et al. showed that MSCs increased the survival rate of model animals by the modulation of macrophage activity [52]. For these reasons, these cells became feasible to therapeutic application for Duchenne muscular dystrophy (DMD). Different groups demonstrated that transplantation of MSCs into murine model of DMD replenished the host satellite cell compartment—allowing the expression of dystrophin and ameliorating the dystrophic phenotype—and also remained as a pool of quiescent satellite cells [53, 54]. Similarly, de Bari and coworkers isolated MSCs from human synovial membrane and injected them into mdx mice, showing MSCs persistence into host muscles up to six months [55]. Gang et al. demonstrated that MSCs derived from human umbilical cord blood (UCB-MSCs) differentiated into skeletal muscle and expressed MyoD and myogenin, muscle-specific transcription factors: transplanted into dystrophic mice, they allowed highly detectable expression of myosin [56].

The role that MSCs play in regulating inflammation is now clear. It is a “multistep” event as MSCs could exert their role as a negative controller/suppressor, by expressing SDF-1 and CCL2 [57], by inhibiting macrophage activation [58]
or by Th1, NK, and cytotoxic T-cell generation [39]. Alternatively, MSCs could act as positive controller/activator by enhancing the proliferation of Th2 cells and regulatory T cells (Treg) [59] and by the expression of immune suppressive cytokines and enzymes [60, 61]. Given these evidences, together with the fact that the only functional treatment for DMD is the corticosteroid therapy that regulates the inflammatory reactions, MSCs were widely used in dystrophic animal models. Firstly, MSCs were injected into the uterus of mdx mice at different days of pregnancy: the cells were observed to engraft in different muscles but their functionality was not altered [62, 63]. Adipose-derived MSCs (AD-MSCs) were transplanted into dystrophic mice and they homed to necrotic fibers. Moreover, AD-MSCs allowed the re-expression of dystrophin and muscular remodeling, even if at lower rate [64]. Furthermore, injection of MSCs was seen to inhibit the expression of creatine kinases whereas increasing the number of centrally nucleated myofibers [65]. According to these evidences, Kong et al. transplanted UCB-MSCs into animal model of limb girdle muscular dystrophies (LGMDs), characterized by predominant weakness and wasting of proximal muscles, but they did not obtain promising results [66]. Although the bones naturally restore without significant scarring, infections, trauma, and cancer could impair their functional restoration, causing several bone defects [67, 68]. Cell-based therapies need to isolate MSCs from the BM of the patient, to expand and enrich the cells and to seed them into the most suitable three-dimensional scaffold and/or matrix [69]. As an example, osteonecrosis is caused by femoral death due to poor blood supply [70]: three patients were treated with MSCs infusion with TCP-treated matrix and good results were obtained [71]. Similarly, Nöth et al. injected a preparation of MSCs into three patients and obtained encouraging results, as shown by radiographic and magnetic resonance imaging examination [72]. MSCs were also successfully used for spinal fusion disease [73], so that phase I clinical trials arose [74, 75]. Patients affected by severe osteogenesis imperfecta were injected systemically with purified allogenic MSCs: these cells were able to engraft into host bones, where they proliferated into osteoblasts and allowed an amelioration in the total bone mineral density [76, 77]. Although these encouraging studies were performed, the amount of MSCs recruited into the bones was too small in a clinical point of view. Alternatively, the group of Le Blanc treated a female fetus with multiple intrauterine fractures with allogenic fetal MSC [13].

4.2. MSCs and Cardiovascular Diseases. Starting from the evidences that MSCs not only secreted molecules that exerted important effects on cellular microenvironment [36] but also differentiated in vitro into cardiomyocytes [78, 79], these cells were extensively used for cardiovascular repair. Shake and Nagaya demonstrated that, following systemically injection into rodent models of these diseases, MSCs engrafed and partially repaired the infarcted myocardium [80, 81]. In particular, Nagaya and collaborators showed that transplanted MSCs increased capillary density and decreased the collagen volume fraction and the fibrosis in the myocardium of a rat suffering from dilated cardiomiopathy [82]. Furthermore, they also noted a significant ventricular functional recovery as previously demonstrated [83]. According to these promising evidences, Katritsis et al. treated 11 infarcted patients with autologous MSCs, together with endothelial progenitor’s cells, and showed partial improvement of myocardial contractility. Unfortunately, they were not able to decipher the mechanisms responsible for these phenomena [84]. Similarly, several infarcted patients that were subjected to coronary intervention were transplanted with autologous MSCs that improved left ventricular function [85]. Takahashi and collaborators assessed that the molecules secreted by MSCs were able to protect the myocardium by preserving its contractile capacity; in particular, MSCs-derived cytokines inhibited the apoptosis of cardiomyocytes, allowing the formation of new vessels in damaged tissues [86].

4.3. MSCs and Liver Disease. Fulminant hepatic failure (FHF) is a severe disease characterized by massive hepatocellular death: the only treatment is liver transplantation that requires lifelong immunosuppression and high costs. Different works demonstrated that MSCs-secreted molecules not only allowed tissue repair of infarcted tissue [82] but furthermore prevented parenchymal cell loss [87]. This way, van Poll and colleagues reported that, following systemic injection of MSCs in a rat model for FHF; there was an amelioration of the pathological phenotype—so that liver injury biomarkers were not released—and, more interestingly, hepatocellular death was drastically reduced, while hepatocytes proliferation increased [88]. Concerning cirrhosis, four patients were injected with autologous MSCs in a phase I trial; they did not suffer from any side effects, thus improving their quality of life [89]. Similarly, 8 patients with end-stage liver disease were treated with MSCs and their condition ameliorated demonstrating that this treatment could be feasible and efficient for this kind of pathologies [90].

4.4. MSCs and Autoimmune Diseases. Since Riordan et al. suggested that in the bone marrow one of the most important functions of MSCs could be the protection of haematopoietic precursor from inflammatory damage [91], the use of MSCs as inhibitors of inflammation became conceptually appealing. This way, MSCs were used to block the development of chronic inflammatory processes that are typical of DMD (as described in detail in Section 4.1), autoimmune arthritis, diabetes, and lupus.

4.4.1. Rheumatoid Arthritis. Rheumatoid arthritis (RA) is characterized by chronic joint inflammation due to loss of immunologic self-tolerance. Gonzalez and colleagues injected DBA/1 mice that suffered from collagen-induced arthritis, with MSCs derived from human adipose tissue, and evaluated the inflammatory response of treated animals [92]. They showed that following the injection of AD-MSCs the levels of inflammatory cytokines and chemokines were decreased as the expansion of antigen-specific Th1/Th17 cell. In contrast this treatment increased the production of IL-10. Together with its well-known function as an anti-inflammatory factor [93], recent findings demonstrated that
IL-10 is fundamental for the development of Tregs that control self-antigen–reactive T cells and induce peripheral tolerance in vivo [94]. Interestingly, they found that treated mice had an increase in the percentage of CD4+ CD25+ Foxp3+ Tregs and suggested that these cells could migrate to the joints, regulating the suppression of the self-reactive response [92]. It is known that type-II collagen (CII), one of the components of hyaline cartilage, acts as an autoantigen in RA. When CII and the other antigenic peptides are recognized by T cells, they cause the uncontrolled activation of immune system cells, leading to destruction of the joints typical of RA patients. Zheng et al. demonstrated that MSCs isolated from RA patients exerted immunosuppressive functions, by inhibiting T-cell proliferation, blocking the secretion of several proinflammatory cytokines, and allowing the expression of anti-inflammatory IL-10 [95]. They also obtained MSCs from chondrocytes and described that, following transplantation into RA joints, these cells not only suppressed the inflammation regulating the secretion of TGF-β but also prevented joint destruction [95].

Similar to the previously described work of González et al. [92], experimental data from Augello [96] and Mao [97] confirmed the positive effects of MSCs transplantation into an animal model of collagen-induced arthritis while others did not describe any amelioration [98, 99]. Schurgers et al. showed the discrepancy between the in vitro and in vivo immunosuppression ability of MSCs. In vitro, MSCs inhibited the proliferation of T cells by regulating the levels of IFN-γ whereas in vivo transplantation of these cells into CIA animal models did not affect the progression of the disease [100]. As an explanation, due to problems during intravenously injection, MSCs could not reach the spleen and lymph nodes, so that they did not exert their functions. As MSC treatment for this pathology was not efficacious, they also suggested to focus on Treg as in mice injected with these cells the pathological signs of arthritis were significantly ameliorated [101, 102]. Another work from Bouffi et al. demonstrated that MSCs elicited their immunosuppressive effect by means of a pathway regulated by the prostaglandine-2. Moreover, they showed that MSCs operated independently from Treg cell induction. Finally they suggested that the contradictory effect of MSCs transplantation could be related to the different age of the mice used in those studies [103].

4.4.2. Systemic Lupus Erythematosus. Systemic lupus erythematosus (SLE) is an autoimmune disease that can affect any part of the body [104]. Recent findings demonstrated several defects in the hematopoietic system of SLE patients, probably due to unbalanced expression of cytokines and other growth factors. Interestingly, it was found that bone marrow–derived CD34+ stem cells overexpressed different surface markers such as CD123 and CD166 that are closely related to T-cell development inflammation [105]. Accordingly, Kushida et al. showed that transplantation of hematopoietic stem cells prevented the onset of the disease in the most commonly studied mouse model of SLE, the MRL/lpr mice [106]. Sun and colleagues determined a possible role of BM–derived MSCs in the hematological disorder typical of SLE patients [107] and suggested that MSCs transplantation could be used to ameliorate the autoimmune progression of the disease [108]. In fact they found that MSCs inhibited T-lymphocytes and Th2 proliferation and B-cell production of autoantibody, so that the pathological signs of MRL/lpr mice were drastically diminished [108].

4.4.3. Type 1 Diabetes. Type 1 diabetes is an autoimmune disease mediated by the production of auto-antibody directed against the β-cells of the pancreas. As a consequence of the destruction of these cells, the quantity of insulin produced is not sufficient to control sugar blood level. Despite the exogenous administration of insulin, long-term consequences of hyperglycemia usually occur, including vascular degeneration, blindness, and kidney failure. Islet replacement is the best way to fully reproduce the physiological release of insulin; however, both the limited availability of transplantable organs and the need for immunosuppression have limited the application of this strategy [109]. Recently it has been suggested that MSCs can overcome these problems as they can be differentiated into glucose-responsive, insulin-producing cells and they possess immunomodulatory properties. It was hypothesized that resident pancreatic MSCs could be forced to adopt a pancreatic fate in vitro. Thus, Zulewski et al. reported the isolation of nestin-positive islet-derived progenitor cells from rat pancreatic islets and their ability to differentiate in vitro toward pancreatic endocrine phenotype [110]. Similarly Huang and Tang described the correction of hyperglycemia in diabetic NOD-SCID mice thanks to nestin-positive precursors derived from human fetal pancreas [111]. However, the results of these studies were controversial and partially inconclusive [112–115] so that MSCs from other tissues could be an alternative. Among them, bone marrow derived MSCs were shown to partially differentiate into endocrine pancreatic cells [116]; furthermore, in vivo maturation of these cells partially compensated their low differentiation efficiency in vitro [117]. An intriguing option comes from studies on umbilical cord blood-derived MSCs that demonstrated the expression of pancreatic development genes in these cells. Recently a population of UCB-derived cells was shown to behave like hES cells, recapitulating the same differentiation steps from early stages to β-cells [118]. In conclusion, before MSCs clinical application in diabetes further studies are needed to improve MSCs based protocols and above all to expand our knowledge on MSCs immunogenicity in a HLA-mismatched context.

4.5. MSCs and Neurodegenerative Diseases

4.5.1. Multiple Sclerosis. Multiple sclerosis is an important cause of neurological disability in young adults. Although it is a multifactorial disease, it is known that the presence of an aberrant immunoresponsiveness leads to patches of damage throughout the brain and spinal cord. Autoreactive T cells cause myelin destruction and secondary oligodendrocyte and axonal damage [119]. Despite the efficacy of immunomodulatory or immunosuppressive drugs in controlling the number of relapses, no current therapy is effective to arrest the
progressive phase of the disease. The therapeutic potential of stem cell lies in enhancing myelin regeneration, through the replacement of lost oligodendrocytes, and therefore in preserving axons, thanks to the neurotrophic support [120]. However, the widespread distribution of lesions and the gray matter damage render the therapeutic efficacy of direct mesenchymal injection really low. Furthermore bone marrow-derived cells ability to make oligodendrocytes was low [121]. Nevertheless, bone marrow-derived MSCs have pronounced immune-modulating and immunosuppressive properties [122, 123] so as they were being tested in clinical trial for relapsing-remitting multiple sclerosis Mesenchymal Stem Cells for Multiple Sclerosis (MEEMS) (NCT01854957). Furthermore it was thought that mesenchymal stem cells promote self-repair by reducing scar formation, by stimulating the formation of new blood vessel, and by secreting growth and neuroprotective factors, such as superoxide dismutase-3 [124]. After intravenous injection, many cells entered the CNS and became widely distributed both in experimental models and in patients [125–127]: safety studies did not evidence adverse effect such as tumor formation, except for meningeval syndrome and some preliminary evidence of beneficial effects that were reported [128, 129].

4.5.2. Amyotrophic Lateral Sclerosis. Suzuki et al. isolated MSCs from muscles and genetically modified them to constitutively express glial-derived neurotrophic factor (GDNF). Then, they transplanted the engineered MSCs into rat model of amyotrophic lateral sclerosis (ALS), a neurodegenerative disease in which patients lose motor neurons and suffer from progressive and lethal paralysis. Interestingly, ALS rats ameliorated the pathological phenotype, increasing the number of neuromuscular connections [130].

4.5.3. Parkinson’s Disease. Parkinson’s disease (PD) is a neurodegenerative disorder characterized by the progressive loss of dopaminergic neurons (DA), especially in the pars compacta of the substantia nigra. The mesostriatal dopaminergic pathway projects in the striatum and their absence causes several motor complications, including rigidity, bradykinesia, and postural instability [131, 132]. DA agonists and Levodopa (l-dopa) are effective symptomatic therapy, but unfortunately, with long-term use, they become inefficient and patients develop significant side effects. Stem cell therapy, with the aim of replacing lost neurons, is the most promising strategy for this disease [133]. It has been demonstrated that MSCs cells can enhance the levels of tyrosine hydroxylase (TH) and dopaminergic levels after transplantation in PD animal models [134]. Furthermore it has been suggested that these cells contribute to neuroprotection by secreting trophic factors, like EGF, VEGF, NT3, FGF-2, HGF, and BDNF or through antiapoptotic signalling [135] without differentiation in neuronal phenotype [136]. For these reasons new strategies, involving the genetic modification of hMSCs, arose, as a tool to induce the secretion of specific factors or to increase the percentage of DA cell differentiation [137]. For example Barzilay et al. transduced adult-derived bone marrow MSCs with a lentivirus carrying LMX1a gene; these cells showed an expression profile similar to a developing mesodiensephalic neuron and allowed DA cell differentiation [138].

4.5.4. Alzheimer Disease. Alzheimer disease (AD) is the most common form of neurodegenerative dementia; affected patients suffer from progressive loss of memory and intellectual abilities. Major anatomopathological features of AD are represented by β-amyloid deposition and neurofibillary tangles formation that ultimately end in cholinergic neurons degeneration. No treatment is currently able to stop the progression of AD [139]. Recently, different studies tried to ameliorate neuropathological deficits in animal model of Alzheimer’s disease through stem cell therapy. In particular, Shin et al. focused on clearance of amyloid plaque and they demonstrated that MSCs could enhance the cell autophagy pathway increasing neuron survival both in vitro and in vivo [140]. Similarly Ma’s group demonstrated that adipose-derived MSC, once transplanted in AD model mice, could modulate the inflammatory environment; in particular they caused an activation of the microglia that promoted the expression of alternative markers and Aβ-degrading enzymes, while decreasing expression levels of proinflammatory factors [141]. Following promising results from MSCs treatment for autoimmune diseases, it was thought to modulate the inflammatory environment of AD. In particular abnormalities of Tregs in cell number and/or function were observed [142] and it was shown that they could modulate microglial activation [143]. Yang et al. demonstrated that umbilical cord-derived MSCS activated Tregs in vitro and once transplanted in AD animal model, Tregs modulated microglia activation, increasing neuron survival [144].

5. Clinical Applications of MSCs

Before clinical applications of stem cells we need to understand their biological characteristics in order to obtain therapeutic effects. In case of MSCs, four properties are considered the most important to guarantee a clinical rescue: (i) the ability to home to the site of inflammation, following tissue injury, when injected intravenously; (ii) the ability to differentiate into various cell types; (iii) the ability to secrete multiple bioactive molecules capable of stimulating recovery of injured cells and of inhibiting inflammation; (iv) the lack of immunogenicity and the ability to perform immune-modulatory functions [87]. Moreover, the role of MSCs in therapeutic effects has still to be elucidated. MSCs have the capacity to migrate and to engraft in site of inflammation, after local or systemic administration. Various studies demonstrated that, under a variety of pathologic conditions, MSCs selectively home to sites of injury, indifferentily from the tissue. Ortiz et al. showed that murine MSCs could home to lung in response to injury, adopt an epithelium-like phenotype, and reduce inflammation in lung tissue of mice challenged with bleomycin [145]. Liu et al. found that transplanted MSCs could migrate to injured muscle tissues in mdx mice [64]. Cell migration depends on many signals, including growth factors, interleukins, and chemokines, secreted by injured cells and immune cells [146]. Recently Yagi et al.
demonstrated that the migration of MSCs is under the control of many tyrosine kinase growth factor receptors like platelet-derived growth factor (PDGF) and insulin like growth factor I (IGF-I); in addition, several chemokines such as CCR2, CCR3, CCR4, or CCL5 ameliorate MSCs migration in *in vitro* migration assays [147].

The first clinical trial using culture-expanded MSCs was performed in 1995 and 15 patients were treated with autologous stem cells [148]. After the first one, a number of clinical trials have been conducted to test the feasibility and efficacy of MSCs therapy. From 2011, 206 clinical trials using MSCs were published on the public clinical trials database (http://www.clinicaltrials.gov/) showing a very wide range of therapeutic applications. Most of these trials are Phase I studies, Phase II, and a combination of Phase I/II studies. Only a small number of these trials are in Phase III or Phase II/III. Most of the trials reported lack of adverse effects in the medium timing, although few of them showed mild and transient peri-injection effects: in general, MSCs seem to be well tolerated [87]. Very promising results were obtained by the injection of autologous and allogenic MSCs in patients suffering from osteogenesis imperfecta [76] while *in vitro* expanded MSCs were used to treat severe [149] and treatment-resistant [150, 151] GVHD patients. In addition, many completed clinical trials have demonstrated the efficacy of MSC infusion for diseases including acute myocardial ischemia (AMI), stroke, amyotrophic lateral sclerosis (ALS), and muscular dystrophies.

### 6. Conclusions

MSCs have many characteristics required for an optimal cell source for cell-replacement therapies, as they are easy to isolate, and retain the ability to expand over a long period of time without serious technical problem. MSCs are linearly restricted; however, there is evidence that MSCs *in vitro* can also express property of ectodermal cells [12]. One requisite of the stem cell-based therapeutic approach is to replace damaged cells. For example, in PD, many studies have focused on examining whether cells replacement therapy could be used. Although transplanted MSCs showed a low cell replacement potential, they improve the environment through the release of neuroprotective factors and they can be engineered to ensure specific expression and secretion. Moreover, MSCs promote "bystander" immunomodulation, as they can release soluble molecules and express immunorelevant receptors that are able to modify the inflammatory environment. However, many questions have to be answered both from preclinical and clinical studies using MSCs before MSCs can be used in wider clinical practice. First of all the safety: now, after MSC administration, few adverse effects have been described in terms of immediate and late effects. The relatively small number of treated patients does not permit to draw definitive conclusions on the safety of MSCs. Unfortunately, MSCs have been reported to promote tumor growth [152] and metastases [153]. Prockop et al. described that MSCs cultured with the clinical cell-therapy protocols commonly used showed potential tumorigenic transformation [154]. Chen et al. found that MSCs could aggravate arthritis in collagen-induced arthritis model by at least upregulating secretion of IL-6, which favors Th17 differentiation [155]. Secondly, quality control: *in vitro* cell amplification needs bacteriological tests (mainly in liquid medium) to face contaminations. In addition, viability and phenotype tests, oncogenic tests, and endotoxin assay should also be included. For each disease type and severity, optimal timing of MSCs administration, cell dose, and schedule of administration need to be decided. Third, clinical grade production: clinical application of MSC requires a large number of cells, so *in vitro* expansion of MSC is necessary. Studies have suggested that continuous passaging of MSCs could lead to cell transformation. Bernardo et al. found that MSCs expansion *in vitro* can be safety until passage 25 [156]. Fourth, clinical transition: it is obvious that much work remains to be done to increase our knowledge of the mechanisms regulating development, homeostasis, and tissue repair and thus provide new tools to implement the efficacy of cell therapy trials. Additionally, there is an urgent requirement to address transplantation related issues, such as engraftment, angiogenesis, tissue remodeling, and modulation of the immune response. Currently, more randomized, controlled, multicenter clinical trials are needed to find the optimal conditions for MSC therapy. In general, we think that successful cell therapy necessitates continuous interaction among biologists, clinicians, and patient working groups in the context of different tissues and diseases.

### Conflict of Interests

The authors declare that there is no conflict of interests regarding the publication of this paper.

### Authors’ Contribution

Farini Andrea and Sitzia Clementina contributed equally to this paper.

### Acknowledgments

This work was supported by Associazione La Nostra Famiglia Fondo DMD Gli Amici di Emanuele, Associazione Amici del Centro Dino Ferrari, and EU’s 7th Framework Programme Optistem 223098, Ministry of Health RF-2009-1547384.

### References

[1] A. J. Friedenstein, J. F. Gorskaia, and N. N. Kulagina, “ Fibroblast precursors in normal and irradiated mouse hematopoietic organs,” *Experimental Hematology*, vol. 4, no. 5, pp. 267–274, 1976.

[2] A. I. Caplan, “Mesenchymal stem cells,” *Journal of Orthopaedic Research*, vol. 9, no. 5, pp. 641–650, 1991.

[3] M. Owen, “Marrow stromal stem cells,” *Journal of Cell Science. Supplement*, vol. 10, pp. 63–76, 1988.

[4] S. E. Haynesworth, M. A. Baber, and A. I. Caplan, “Cell surface antigens on human marrow-derived mesenchymal cells are detected by monoclonal antibodies,” *Bone*, vol. 13, no. 1, pp. 69–80, 1992.
[5] M. F. Pittenger, A. M. Mackay, S. C. Beck et al., “Multilineage potential of adult human mesenchymal stem cells,” Science, vol. 284, no. 5411, pp. 143–147, 1999.

[6] A. Mahmood, D. Lu, M. Lu et al., “Treatment of traumatic brain injury in adult rats with intraventricular administration of human bone marrow stromal cells,” Neurosurgery, vol. 53, no. 3, pp. 697–703, 2003.

[7] J. M. Murphy, D. J. Fink, E. B. Hunziker, and F. P. Barry, “Stem cell therapy in a caprine model of osteoarthritis,” Arthritis and Rheumatism, vol. 48, no. 12, pp. 3464–3474, 2003.

[8] G. J. Block, S. Ohkouchi, F. Fung et al., “Multipotent stromal cells are activated to reduce apoptosis in part by upregulation and secretion of stanniocalcin-1,” Stem Cells, vol. 27, no. 3, pp. 670–681, 2009.

[9] L. Chen, E. E. Tredget, P. Y. G. Wu, Y. Wu, and Y. Wu, “Paracrine factors of mesenchymal stem cells recruit macrophages and endothelial lineage cells and enhance wound healing,” PLoS ONE, vol. 3, no. 4, Article ID e1886, 2008.

[10] J. M. Karp and G. S. L. Teo, “Mesenchymal stem cell homing: the devil is in the details,” Cell Stem Cell, vol. 4, no. 3, pp. 206–216, 2009.

[11] A. Alhaddaq and J. J. Mao, “Mesenchymal stem cells: isolation and therapeutic,” Stem Cells and Development, vol. 13, no. 4, pp. 436–448, 2004.

[12] N. B. Nardi and L. da Silva Meirelles, “Mesenchymal stem cells: isolation, in vitro expansion and characterization,” Handbook of Experimental Pharmacology, no. 174, pp. 249–282, 2006.

[13] K. le Blanc and M. F. Pittenger, “Mesenchymal stem cells: progress toward promise,” Cytotherapy, vol. 7, no. 1, pp. 36–45, 2005.

[14] M. Miura, S. Grontthos, M. Zhao et al., “SHED: stem cells from human exfoliated deciduous teeth,” Proceedings of the National Academy of Sciences of the United States of America, vol. 100, no. 10, pp. 5807–5812, 2003.

[15] M. J. Hoogduijn, M. J. Crop, A. M. A. Peeters et al., “Human heart, spleen, and perirenal fat-derived mesenchymal stem cells have immunomodulatory capacities,” Stem Cells and Development, vol. 16, no. 4, pp. 597–604, 2007.

[16] C. S. Sondergaard, C. J. Hodonsky, L. Khait et al., “Human thymus mesenchymal stromal stem cells augment force production in self-organized cardiac tissue,” Annals of Thoracic Surgery, vol. 90, no. 3, pp. 796–803, 2010.

[17] F. P. Barry and J. M. Murphy, “Mesenchymal stem cells: clinical applications and biological characterization,” International Journal of Biochemistry and Cell Biology, vol. 36, no. 4, pp. 568–584, 2004.

[18] A. I. Caplan, “Why are MSCs therapeutic? New data: new insight,” The Journal of Pathology, vol. 217, no. 2, pp. 318–324, 2009.

[19] P. Dore-Duffy, A. Katchev, X. Wang, and E. Van Buren, “CNS microvascular pericytes exhibit multipotential stem cell activity,” Journal of Cerebral Blood Flow and Metabolism, vol. 26, no. 5, pp. 613–624, 2006.

[20] B. Meyrick and L. Reid, “The effect of continued hypoxia on rat pulmonary arterial circulation. An ultrastructural study,” Laboratory Investigation, vol. 38, no. 2, pp. 188–200, 1978.

[21] M. K. Majumdar, M. A. Thiede, J. D. Mosca, M. Moorman, and S. L. Gerson, “Phenotypic and functional comparison of cultures of marrow-derived mesenchymal stem cells (MSCs) and stromal cells,” Journal of Cellular Physiology, vol. 176, no. 1, pp. 57–66, 1998.

[22] G. Chamberlain, J. Fox, B. Ashton, and J. Middleton, “Concise review: mesenchymal stem cells: their phenotype, differentiation capacity, immunological features, and potential for homing,” Stem Cells, vol. 25, no. 11, pp. 2739–2749, 2007.

[23] M. S. Penn, “Importance of the SDF-1/CXCR4 axis in myocardial repair,” Circulation Research, vol. 104, no. 10, pp. 1133–1135, 2009.

[24] B. Schönemeier, S. Schulz, V. Hoellt, and R. Stumm, “Enhanced expression of the CXCL12/SDF-1 chemokine receptor CXCR7 after cerebral ischemia in the rat brain,” Journal of Neuroimmunology, vol. 198, no. 1-2, pp. 39–45, 2008.

[25] B. T. G. Tan, M. M. G. Lee, and R. Ruan, “Bone marrow-derived cells that home to acoustic deafened cochlea preserved their hematopoietic identity,” Journal of Comparative Neurology, vol. 509, no. 2, pp. 167–179, 2008.

[26] S. Avniel, Z. Arik, A. Maly et al., “Involvement of the CXCL12/CXCR4 pathway in the recovery of skin following burns,” Journal of Investigative Dermatology, vol. 126, no. 2, pp. 468–476, 2006.

[27] A. Fox, J. Smythe, N. Fisher et al., “Mobilization of endothelial progenitor cells into the circulation in burned patients,” British Journal of Surgery, vol. 95, no. 2, pp. 244–251, 2008.

[28] T. Kitao, H. Ito, E. M. Schwarz et al., “Stromal cell-derived factor 1/CXCR4 signaling is critical for the recruitment of mesenchymal stem cells to the fracture site during skeletal repair in a mouse model,” Arthritis and Rheumatism, vol. 60, no. 3, pp. 813–823, 2009.

[29] J. Gao, J. E. Dennis, R. F. Muzic, M. Lundberg, and A. I. Caplan, “The dynamic in vivo distribution of bone marrow-derived mesenchymal stem cells after infusion,” Cells Tissues Organs, vol. 169, no. 1, pp. 12–20, 2001.

[30] M. B. Herrera, B. Bussolati, S. Bruno, V. Fonsato, G. M. Romanzini, and G. Camussi, “Mesenchymal stem cells contribute to the renal repair of acute tubular epithelial injury,” International Journal of Molecular Medicine, vol. 14, no. 6, pp. 1035–1041, 2004.

[31] K. A. Jackson, S. M. Majka, H. Wang et al., “Regeneration of ischemic cardiac muscle and vascular endothelium by adult stem cells,” The Journal of Clinical Investigation, vol. 107, no. 11, pp. 1395–1402, 2001.

[32] D. Orlic, K. Jajstura, S. Chimenti et al., “Bone marrow cells regenerate infarcted myocardium,” Nature, vol. 410, no. 6829, pp. 701–705, 2001.

[33] D. Orlic, J. Kajstura, S. Chimenti et al., “Mobilized bone marrow cells repair the infarcted heart, improving function and survival,” Proceedings of the National Academy of Sciences of the United States of America, vol. 98, no. 18, pp. 10344–10349, 2001.

[34] S. Gojo, N. Gojo, Y. Takeda et al., “In vivo cardiovasculogenesis by direct injection of isolated adult mesenchymal stem cells,” Experimental Cell Research, vol. 288, no. 1, pp. 51–59, 2003.

[35] D. Zhang, F. Zhang, Y. Zhang et al., “Combining erythropoietin infusion with intramyocardial delivery of bone marrow cells is more effective for cardiac repair,” Transplant International, vol. 20, no. 2, pp. 174–183, 2007.

[36] C. P. Hofstetter, E. J. Schwarz, D. Hess et al., “Marrow stromal cells form guiding strands in the injured spinal cord and promote recovery,” Proceedings of the National Academy of Sciences of the United States of America, vol. 99, no. 4, pp. 2199–2204, 2002.

[37] J. A. Kode, S. Mukherjee, M. V. Joglekar, and A. A. Hardikar, “Mesenchymal stem cells: immunobiology and role in immunomodulation and tissue regeneration,” Cytotherapy, vol. 11, no. 4, pp. 377–391, 2009,
G. M. Spaggiari, A. Capobianco, S. Becchetti, M. C. Mingari, J. Stagg, “Immune regulation by mesenchymal stem cells: two
Z.-G. Zhao, W.-M. Li, Z.-C. Chen, Y. You, and P. Zou, “Immuno-suppressive properties of mesenchymal stem cells derived from
bone marrow of patients with chronic myeloid leukemia,” Immunological Investigations, vol. 37, no. 7, pp. 726–739, 2008.
A. Nasef, C. Mazurier, S. Bouchet et al., “Leukemia inhibitory factor: role in human mesenchymal stem cells mediated
immunosuppression,” Cellular Immunology, vol. 253, no. 1-2, pp. 16–22, 2008.
H. Sheng, Y. Wang, Y. Jin et al., “A critical role of IFNγ in priming MSC-mediated suppression of T cell proliferation through
up-regulation of B7-H1,” Cell Research, vol. 18, no. 8, pp. 846–857, 2008.
G. Ren, L. Zhang, X. Zhao et al., “Mesenchymal stem cell-mediated immunosuppression occurs via concerted action of
chemokines and nitric oxide,” Cell Stem Cell, vol. 2, no. 2, pp. 141–150, 2008.
G. M. Spaggiari, A. Capobianco, S. Becchetti, M. C. Mingari, and L. Moretta, “Mesenchymal stem cell-natural killer cell inter-
actions: role that activated NK cells are capable of killing MSCs, whereas MSCs can inhibit IL-2-induced NK-cell prolif-
eration,” Blood, vol. 107, no. 4, pp. 1484–1490, 2006.
A. Poggi, C. Prevosto, A.-M. Massaro et al., “Interaction between human NK cells and bone marrow stromal cells induces
NK cell triggering: role of NKP30 and NKG2D receptors,” The Journal of Immunology, vol. 175, no. 10, pp. 6352–6360,
2005.
J. Stagg, “Immune regulation by mesenchymal stem cells: two sides to the coin,” Tissue Antigens, vol. 69, no. 1, pp. 1–9, 2007.
A. Jewett, A. Arasteh, H.-C. Tseng et al., “Strategies to rescue mesenchymal stem cells (MSCs) and dental pulp stem cells
(DPSCs) from NK cell mediated cytotoxicity,” PloS ONE, vol. 5, no. 3, Article ID e9874, 2010.
M. Giuliani, A. Bennaceur-Griscelli, A. Nanbakhsh et al., “TLR ligands stimulation protects MSC from NK killing,” Stem Cells,
vol. 32, no. 1, pp. 290–300, 2014.
B. Zheng, B. Cao, M. Crisan et al., “Prospective identification of myogenic endothelial cells in human skeletal muscle,” Nature
Biotechnology, vol. 25, pp. 1025–1034, 2007.
L. J. Nesti, W. M. Jackson, R. M. Shanti et al., “Differentiation potential of multipotent progenitor cells derived from
warratmannized muscle tissue,” Journal of Bone and Joint Surgery A, vol. 90, no. 11, pp. 2390–2398, 2008.
M. A. F. V. Gonçalves, A. A. F. de Vries, M. Holkers et al., “Human mesenchymal stem cells ectopically expressing full-
length dystrophin can complement Duchenne muscular dystrophy myotubes by cell fusion,” Human Molecular Genetics, vol. 15,
no. 2, pp. 213–221, 2006.
K. Németh, A. Leeelahavanichkul, P. S. T. Yuen et al., “Bone marrow stromal cells attenuate sipsis via prostaglandin E 2-
dependent reprogramming of host macrophages to increase their interleukin-10 production,” Nature Medicine, vol. 15, no.
1, pp. 42–49, 2009.
A. Asakura, P. Scale, A. Girgis-Gabardo, and M. A. Rudnicki, “Myogenic specification of side population cells in skeletal
muscle,” The Journal of Cell Biology, vol. 159, no. 1, pp. 123–134, 2002.
Z. Qu-Petersen, B. Deasy, R. Jankowski et al., “Identification of a novel population of muscle stem cells in mice: potential for
muscle regeneration,” The Journal of Cell Biology, vol. 157, no. 5, pp. 851–864, 2002.
C. de Bari, F. Dell’Accio, F. Vandenabeele, J. R. Vremeech, J.-M. Raymackers, and F. P. Luyten, “Skeletal muscle repair by adult
human mesenchymal stem cells from synovial membrane,” The Journal of Cell Biology, vol. 160, no. 6, pp. 909–918, 2003.
E. J. Gang, J. A. Jeong, S. H. Hong et al., “Skeletal myogenic differentiation of mesenchymal stem cells isolated from human
umbilical cord blood,” Stem Cells, vol. 22, no. 4, pp. 617–624, 2004.
M. Rafei, P. M. Campeau, A. Aguilar-Mahcka et al., “Mesenchymal stromal cells ameliorate experimental autoimmune
encephalomyelitis by inhibiting CD4 Th17 T cells in a CC chemokine ligand 2-dependent manner,” The Journal of Immunology,
vol. 182, no. 6, pp. 5994–6002, 2009.
Y.-W. Yang, H. Bai, C.-B. Wang, M. Lin, and L.-Q. Wu, “Experimental study on influence of bone marrow mesenchymal stem
cells on activation and function of mouse peritoneal macrophages,” Zhonghua Xue Ye Xue Za Zhi, vol. 29, no. 8, pp. 540–
543, 2008.
F. Casiraghi, N. Azzollini, P. Cassis et al., “Pretransplant infusion of mesenchymal stem cells prolongs the survival of a semiallo-
geneic heart transplant through the generation of regulatory T cells,” The Journal of Immunology, vol. 181, no. 6, pp. 3933–3946,
2008.
D. Campioni, R. Rizzo, M. Stignani et al., “A decreased positivity for CD90 on human mesenchymal stromal cells (MSCs)
is associated with a loss of immunosuppressive activity by MSCs,” Cytometry B: Clinical Cytometry, vol. 76, no. 3, pp. 225–230,
2009.
B. J. Jones, G. Brooke, K. Atkinson, and S. J. McTaggart, “Immunosuppression by placent al indoleamine 2,3-dioxyn-
genase: a role for mesenchymal stem cells,” Placenta, vol. 28, no. 11-12, pp. 1174–1181, 2007.
J. Chan, S. N. Waddington, K. O’Donoghue et al., “Widespread distribution and muscle differentiation of human fetal mes-
enchymal stem cells after intrauterine transplantation in dystrophic mdx mouse,” Stem Cells, vol. 25, no. 4, pp. 875–884, 2007.
T. C. MacKenzie, G. P. Kobinger, N. A. Kootstra et al., “Efficient transduction of liver and muscle after in utero injection of
tentiviral vectors with different pseudotypes,” Molecular Therapy, vol. 6, no. 3, pp. 349–358, 2002.
Y. Liu, X. Yan, Z. Sun et al., “Flk-1” adipose-derived mesenchy-
mal stem cells differentiate into skeletal muscle satellite cells and ameliorate muscular dystrophy in MDX Mice,” Stem Cells and
Development, vol. 16, no. 5, pp. 695–706, 2007.
S.-W. Feng, X.-L. Lu, Z.-S. Liu et al., “Dynamic distribution of
bone marrow-derived mesenchymal stromal cells and change of
pathology after infusing into mdx mice,” Cytotherapy, vol. 10,
no. 3, pp. 254–264, 2008.
K. Y. Kong, J. Ren, M. Kraus, S. P. Finkenstein, and R. H. Brown
Jr., “Human umbilical cord blood cells differentiate into muscle
in sjl muscular dystrophy mice,” Stem Cells, vol. 22, no. 6, pp.
981–993, 2004.
J. Rauh, F. Milan, K.-P. Günther, and M. Stiehler, “Bioreactor
systems for bone tissue engineering,” Tissue Engineering B:
Reviews, vol. 17, no. 4, pp. 263–280, 2011.
[68] R. S. Tuan, “Role of adult stem/progenitor cells in osseointegration and implant loosening,” The International Journal of Oral & Maxillofacial Implants, vol. 26, supplement, pp. 50–69, 2011.

[69] P. Bianco and P. G. Robey, “Stem cells in tissue engineering,” Nature, vol. 414, no. 6859, pp. 118–121, 2001.

[70] R. Cancedda, G. Bianchi, A. Derubeis, and R. Quarto, “Cell therapy for bone disease: a review of current status,” Stem Cells, vol. 21, no. 5, pp. 610–619, 2003.

[71] K. Kawate, H. Yajima, H. Ohgushi et al., “Tissue-engineered bone grafts for bone regeneration: in vitro and in vivo,” Artificial Organs, vol. 30, no. 12, pp. 960–962, 2006.

[72] U. Nöth, J. Reichert, S. Reppenhagen et al., “Cell based therapy for the treatment of femoral head necrosis,” Orthopade, vol. 36, no. 5, pp. 466–471, 2007.

[73] S. G. Pneumatics, G. K. Triantafyllopoulos, S. Chatzioannou, E. K. Basdara, and A. G. Papavassiliou, “Biomolecular strategies of bone augmentation in spinal surgery,” Trends in Molecular Medicine, vol. 17, no. 4, pp. 215–222, 2011.

[74] D. Neen, D. Noyes, M. Shaw, S. Gwilym, N. Fairlie, and N. Birch, “Haloos and bone marrow aspirate used for lumbar spine fusion: a case controlled study comparing halos with autograft,” Spine, vol. 31, no. 18, pp. E656–E640, 2006.

[75] P. Zhang, Y.-K. Gan, J. Tang et al., “Clinical study of lumbar fusion by hybrid construct of stem cells technique and biodegradable material,” Zhonghua Wai Ke Za Zhi, vol. 46, no. 7, pp. 493–496, 2008.

[76] E. M. Horwitz, P. L. Gordon, W. K. K. Koo et al., “Isolated allogeneic bone marrow-derived mesenchymal stem cells engraft and stimulate growth in children with osteogenesis imperfecta: implications for cell therapy of bone,” Proceedings of the National Academy of Sciences of the United States of America, vol. 99, no. 13, pp. 8932–8937, 2002.

[77] E. M. Horwitz, D. J. Prockop, L. A. Fitzpatrick et al., “Transplantability and therapeutic effects of bone marrow-derived mesenchymal cells in children with osteogenesis imperfecta,” Nature Medicine, vol. 5, no. 3, pp. 309–313, 1999.

[78] S. Makino, F. Fukuda, S. Miyoshi et al., “Cardiomyocytes can be generated from marrow stromal cells in vitro,” The Journal of Clinical Investigation, vol. 103, no. 5, pp. 697–705, 1999.

[79] W. Xu, X. Zhang, H. Qian et al., “Mesenchymal stem cells from adult human bone marrow differentiate into a cardiomyocyte phenotype in vitro,” Experimental Biology and Medicine, vol. 229, no. 7, pp. 623–631, 2004.

[80] N. Nagaya, T. Fujii, T. Iwase et al., “Intravenous administration of mesenchymal stem cells improves cardiac function in rats with acute myocardial infarction through angiogenesis and myogenesis,” American Journal of Physiology—Heart and Circulatory Physiology, vol. 287, no. 6, pp. H2670–H2676, 2004.

[81] J. G. Shake, P. J. Gruber, W. A. Baumgartner et al., “Mesenchymal stem cell implantation in a swine myocardial infarct model: engraftment and functional effects,” The Annals of Thoracic Surgery, vol. 73, no. 6, pp. 1919–1926, 2002.

[82] N. Nagaya, K. Kawahata, T. Itó et al., “Transplantation of mesenchymal stem cells improves cardiac function in a rat model of dilated cardiomyopathy,” Circulation, vol. 112, no. 8, pp. 1128–1135, 2005.

[83] K. C. Wollert, G. P. Meyer, J. Lotz et al., “Intracoronary autologous bone-marrow cell transfer after myocardial infarction: the BOOST randomised controlled clinical trial,” The Lancet, vol. 364, no. 9429, pp. 141–148, 2004.

[84] D. G. Katritsis, P. A. Sotiropoulou, E. Karvouni et al., “Transcoronary transplantation of autologous mesenchymal stem cells and endothelial progenitors into infarcted human myocardium,” Catheterization and Cardiovascular Interventions, vol. 65, no. 3, pp. 321–329, 2005.

[85] S.-L. Chen, W.-W. Fang, F. Ye et al., “Effect on left ventricular function of intracoronary transplantation of autologous bone marrow mesenchymal stem cell in patients with acute myocardial infarction,” The American Journal of Cardiology, vol. 94, no. 1, pp. 92–95, 2004.

[86] M. Takahashi, T.-S. Li, R. Suzuki et al., “Cytokines produced by bone marrow cells can contribute to functional improvement of the infarcted heart by protecting cardiomyocytes from ischemic injury,” American Journal of Physiology—Heart and Circulatory Physiology, vol. 291, no. 2, pp. H886–H893, 2006.

[87] S. Wang, X. Qu, and R. C. Zhao, “Clinical applications of mesenchymal stem cells,” Journal of Hematology & Oncology, vol. 5, article 19, 2012.

[88] D. van Poll, B. Parekkadan, C. H. Cho et al., “Mesenchymal stem cell-derived molecules directly modulate hepatocellular death and regeneration in vitro and in vivo,” Hepatology, vol. 47, no. 5, pp. 1634–1643, 2008.

[89] M. Mohamadnejad, K. Alimoghaddam, M. Mohyeddin-Bonab et al., “Phase I trial of autologous bone marrow mesenchymal stem cell transplantation in patients with decompenated liver cirrhosis,” Archives of Iranian Medicine, vol. 10, no. 4, pp. 459–466, 2007.

[90] P. Kharazhi, P. M. Hellström, B. Noorinayer et al., “Improvement of liver function in liver cirrhosis patients after autologous mesenchymal stem cell injection: a phase I-II clinical trial,” European Journal of Gastroenterology and Hepatology, vol. 21, no. 10, pp. 1199–1205, 2009.

[91] N. H. Riordan, K. Chan, A. M. Marleau, and T. E. Ichim, “Cord blood in regenerative medicine: do we need immune suppression?” Journal of Translational Medicine, vol. 5, article 8, 2007.

[92] M. A. González, E. González-Rey, L. Rico, D. Büscher, and M. Delgado, “Treatment of experimental arthritis by inducing immune tolerance with human adipose-derived mesenchymal stem cells,” Arthritis and Rheumatism, vol. 60, no. 4, pp. 1006–1019, 2009.

[93] M. Walmsley, P. D. Katsikis, E. Abney et al., “Interleukin-10 inhibition of the progression of established collagen-induced arthritis,” Arthritis and Rheumatism, vol. 39, no. 3, pp. 495–503, 1996.

[94] E. J. Wehrens, B. J. Prakken, and F. van Wijk, “T cells out of control—impaired immune regulation in the inflamed joint,” Nature Reviews Rheumatology, vol. 9, pp. 34–42, 2013.

[95] Z. H. Zheng, X. Y. Li, J. Ding, J. F. Jia, and P. Zhu, “Allogeneic mesenchymal stem cell and mesenchymal stem cell-differentiated chondrocyte suppress the responses of type II collagen-reactive T cells in rheumatoid arthritis,” Rheumatology, vol. 47, no. 1, pp. 22–30, 2008.

[96] A. Augello, R. Tasso, S. M. Negriti, R. Cancetta, and G. Penazzi, “Cell therapy using allogeneic bone marrow mesenchymal stem cells prevents tissue damage in collagen-induced arthritis,” Arthritis and Rheumatism, vol. 56, no. 4, pp. 1175–1186, 2007.

[97] F. Mao, W.-R. Xu, H. Qian et al., “Immunosuppressive effects of mesenchymal stem cells in collagen-induced mouse arthritis,” Inflammation Research, vol. 59, no. 3, pp. 219–225, 2010.

[98] J.-J. Choi, S.-A. Yoo, S.-J. Park et al., “Mesenchymal stem cells overexpressing interleukin-10 attenuate collagen-induced
relapsing-progressive multiple sclerosis," Clinical Pharmacology and Therapeutics, vol. 87, no. 6, pp. 679–685, 2010.

[129] B. Yamout, H. Hourani, H. Salti et al., “Bone marrow mesenchymal stem cell transplantation in patients with multiple sclerosis: a pilot study,” Journal of Neuroimmunology, vol. 227, no. 1-2, pp. 185–189, 2010.

[130] M. Suzuki, J. McHugh, C. Tork et al., “Direct muscle delivery of GDNF with human mesenchymal stem cells improves motor neuron survival and function in a rat model of familial ALS,” Molecular Therapy, vol. 16, no. 12, pp. 2002–2010, 2008.

[131] Y. Agid, “Parkinson’s disease: pathophysiology,” The Lancet, vol. 337, no. 8753, pp. 1321–1324, 1991.

[132] S. J. Kish, K. Shannak, and O. Hornykiewicz, “Uneven pattern of dopamine loss in the striatum of patients with idiopathic Parkinson’s disease. Pathophysiological and clinical implications,” The New England Journal of Medicine, vol. 318, no. 14, pp. 876–880, 1988.

[133] C. F. P. Teixeira, S. R. Zamunér, J. P. Zuliani et al., “Neutrophils do not contribute to local tissue damage, but play a key role in skeletal muscle regeneration, in mice injected with Bothrops asper snake venom,” Muscle and Nerve, vol. 28, no. 4, pp. 449–459, 2003.

[134] I. Kan, T. Ben-Zar, Y. Barhum et al., “Dopaminergic differentiation of human mesenchymal stem cells: Utilization of bioassay for tyrosine hydroxylase expression,” Neuroscience Letters, vol. 419, no. 1, pp. 28–33, 2007.

[135] F. Wang, T. Yasuhara, T. Shingo et al., “Intravenous administration of mesenchymal stem cells exerts therapeutic effects on parkinsonian model of rats: focusing on neuroprotective effects of stromal cell-derived factor-1α,” BMC Neurosciences, vol. 11, article 52, 2010.

[136] A. Wilkins, K. Kemp, M. Ginty, K. Hares, E. Mallam, and N. R. Barzilay, T. Ben-Zur, S. Bulvik, E. Melamed, and D. Offen, “Ex vivo expansion and subsequent infusion of human bone marrow-derived stromal progenitor cells (mesenchymal progenitor cells): implications for therapeutic use,” Bone Marrow Transplantation, vol. 16, no. 4, pp. 557–564, 1995.

[137] K. le Blanc, I. Rasmusson, B. Sundberg et al., “Treatment of severe acute graft-versus-host disease with third party haploidentical mesenchymal stem cells,” The Lancet, vol. 363, no. 9419, pp. 1439–1441, 2004.

[138] K. le Blanc, F. Frassoni, L. Ball et al., “Mesenchymal stem cells for treatment of steroid-resistant, severe, acute graft-versus-host disease: a phase II study,” The Lancet, vol. 371, no. 9624, pp. 1579–1586, 2008.

[139] O. Ringdén, M. Uzunel, I. Rasmusson et al., “Mesenchymal stem cells for treatment of therapy-resistant graft-versus-host disease,” Transplantation, vol. 81, no. 10, pp. 1390–1397, 2006.

[140] F. Djouad, P. Pience, C. Bony et al., “Immunosuppressive effect of mesenchymal stem cells favors tumor growth in allogeneic animals,” Blood, vol. 102, no. 10, pp. 3837–3844, 2003.

[141] A. E. Karnoub, A. B. Dash, A. P. Vo et al., “Mesenchymal stem cells within tumour stroma promote breast cancer metastasis,” Nature, vol. 449, no. 7162, pp. 557–563, 2007.

[142] D. J. Prockop, M. Brenner, W. E. Fibbe et al., “Defining the risks of mesenchymal stem cell therapy,” Cytotherapy, vol. 12, no. 5, pp. 576–578, 2010.

[143] B. Chen, J. Hu, L. Liao et al., “Flk-1+ mesenchymal stem cells aggravate collagen-induced arthritis by up-regulating interleukin-6,” Clinical and Experimental Immunology, vol. 159, no. 3, pp. 292–302, 2010.

[144] M. E. Bernardo, F. Locatelli, and W. E. Fibbe, “Mesenchymal stem cells: a novel treatment modality for tissue repair,” Annals of the New York Academy of Sciences, vol. 1176, pp. 101–117, 2009.

[145] J. T. Walsh and J. Kipnis, “Regulatory T cells in CNS injury: the simple, the complex and the confused,” Trends in Molecular Medicine, vol. 17, no. 10, pp. 541–547, 2011.

[146] H. Yang, H. Yang, Z. Xie, L. Wei, and J. Bi, “Systemic transplantation of human umbilical cord derived mesenchymal stem cells-educated T regulatory cells improved the impaired cognition in AbetaPPswe/PS1dE9 transgenic mice,” PLoS ONE, vol. 8, Article ID e69129, 2013.

[147] L. A. Ortiz, F. Gambelli, C. McBride et al., “Mesenchymal stem cell engraftment in lung is enhanced in response to bleomycin exposure and ameliorates its fibrotic effects,” Proceedings of the National Academy of Sciences of the United States of America, vol. 100, no. 14, pp. 8407–8411, 2003.

[148] E. Spaeth, A. Klop, J. Dembinski, M. Andreeff, and F. Marinì, “Inflammation and tumor microenvironments: defining the migratory itinerary of mesenchymal stem cells,” Gene Therapy, vol. 15, no. 10, pp. 730–738, 2008.

[149] T. Yagi, D. Ito, Y. Okada et al., “Modeling familial Alzheimer’s disease with induced pluripotent stem cells,” Human Molecular Genetics, vol. 20, no. 23, Article ID ddr394, pp. 4530–4539, 2011.

[150] H. M. Lazarus, S. E. Haynesworth, S. L. Gerson, N. S. Rosenthal, and A. I. Caplan, “Ex vivo expansion and subsequent infusion of human bone marrow-derived stromal progenitor cells (mesenchymal progenitor cells): implications for therapeutic use,” Bone Marrow Transplantation, vol. 16, no. 4, pp. 557–564, 1995.