

Immunomodulatory properties and therapeutic application of mesenchymal stem cells

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Introduction

Mesenchymal stem cells (MSCs) are multi-potent progenitors constituting a small proportion of the bone marrow and are present in both adult and fetal tissues [1,2], including adipose tissue, umbilical cord blood, amniotic fluid and fetal lung [3–6]. They were first characterized by Friedenstein and colleagues more than 30 years ago, and were described as fibroblast-like cells with the property of adhering to plastic when cultured [7,8]. This property can be used to purify these cells and enables them to expand several-fold *in vitro* without losing their differentiation capacity. MSCs are able to differentiate into both mesenchymal and non-mesenchymal cell lineages such as adipocytes, osteoblasts, chondrocytes, tenocytes, skeletal myocytes, neurones and cells of the visceral mesoderm, both *in vitro* and *in vivo* [9,10].

Because a specific marker for human MSCs has not been identified, it is difficult to recognize these cells. The International Society for Cellular Therapy has recommended the following minimum criteria for defining multi-potent human MSCs [11,12]: (i) adherence to plastic under standard culture conditions; (ii) positive for expression of

Summary

Mesenchymal stem cells (MSCs) are multi-potent progenitor cells that are isolated from the bone marrow and several adult organs and tissues. These cells possess remarkable immunosuppressive properties and can inhibit the proliferation and function of the major immune cell populations, including T cells, B cells and natural killer (NK) cells; modulate the activities of dendritic cells (DCs); and induce regulatory T cells both *in vivo* and *in vitro*. These unique properties make MSCs ideal candidates for clinical application as immunosuppressants. The immunomodulatory effect of MSCs is mediated by a non-specific anti-proliferative action of these cells, which is dependent on cell–cell contact or secreted soluble factors such as indoleamine 2,3-dioxygenase (IDO), prostaglandin E₂ (PGE₂), nitric oxide (NO), histocompatibility leucocyte antigen-G (HLA-G), transforming growth factor (TGF)- β , interferon (IFN)- γ and interleukin (IL)-1 β . Considerable progress has been obtained in preclinical studies on MSCs, including those on their ability to activate allogeneic cells. This review examines the current understanding of the immunomodulatory properties of MSCs and its therapeutic implication for immune-mediated diseases and transplant rejection.

Keywords: immunosuppression, mesenchymal stem cells, review, therapeutic applications

CD105, CD73 and CD90 and negative for expression of the haematopoietic cell surface markers CD34, CD45, CD11a, CD19 or CD79a, CD14 or CD11b and histocompatibility locus antigen (HLA)-DR; and (iii) under a specific stimulus, differentiation into osteocytes, adipocytes and chondrocytes *in vitro*. Because of their unique regenerative potential, MSCs exhibit potential for use in tissue regeneration and repair for conditions such as cardiac anomalies or injury, bone disorders and metabolic diseases. One of the most intriguing features of MSCs is that they escape immune recognition and can inhibit immune responses [13].

In this review, we discuss the *in vivo* and *in vitro* immunomodulatory properties of MSCs, the possible mechanisms underlying the expression of these properties and the potential clinical use of MSCs *in vivo* as modulators of immune responses.

MSCs modulate T cell proliferation and function

Numerous studies have demonstrated that MSCs can suppress the T lymphocyte proliferation induced by alloantigens, mitogens and anti-CD3 and anti-CD28 antibodies *in vitro* in

humans, baboons and mice [14–20]. MSCs have a similar effect on memory and naive T cells [20], as well as CD4⁺ and CD8⁺ T cells [21], of a murine model. In addition, this suppressive effect did not require major histocompatibility complex (MHC) restriction and could also be mediated by allogeneic MSCs [15,20]. This effect may be attributed to the inhibition of cell division, which is evidenced by the accumulation of cells in the G0/G1 phase of the cell cycle [21]. At the molecular level, cyclin D2 expression is down-regulated, whereas p27 expression is up-regulated; this may explain why T cell proliferation, rather than activation, and interferon (IFN)- γ production are affected by MSCs [21]. Inhibition of T cell proliferation by MSCs appears to be mediated by both cell–cell interaction [17,22,23] and release of soluble factors such as IFN- γ and interleukin (IL)-1 β [24,25]. Some studies have indicated that soluble factors are essential for enhancing the suppressive effect of human MSCs, while the effect of rodent MSCs is mediated by cell–cell contact [14,17,20,26]. Transforming growth factor (TGF)- β 1, hepatocyte growth factor (HGF) [14], indoleamine 2,3-dioxygenase (IDO) [27] and prostaglandin E₂ (PGE₂) [28] represent MSC-derived molecules that are believed to have immunomodulatory activity on T cell responses. Neutralizing antibodies against TGF- β and HGF can restore the MSC-induced suppression of T cell proliferation [14]. Treatment with IFN- γ causes MSCs to express the protein IDO and exhibit functional activity of IDO, which in turn degrades essential tryptophan and results in kynurenine synthesis and thereby suppresses lymphocyte proliferation [27]. Co-culturing T cells with MSCs resulted in elevated levels of PGE₂, and treatment with inhibitors of PGE₂ production mitigated the MSC-mediated immune modulation [28]; however, the mechanism underlying the immunosuppressive effect of PGE₂ is poorly understood. The production of nitric oxide (NO) by MSCs has also been implicated as a potential mechanism by which MSCs inhibit T cell proliferation [29]. NO inhibits the proliferation of T cells by suppressing the phosphorylation of signal transducer and activator of transcription-5 (STAT5), a transcription factor crucial for T cell activation and proliferation [30] (Fig. 1). Ding *et al.* reported that matrix metalloproteinases (MMPs), in particular MMP-2 and MMP-9 secreted by MSCs, mediate the suppressive activity of MSCs via reduction of CD25 expression on responding T cells in a model of allogeneic islet transplant [31]. The secretion of human leucocyte antigen-G5 (HLA-G5) by MSCs is reported to be essential for the following effects of MSCs: suppression of T cell and NK cell function, shift of the allogeneic T cell response to a T helper type 2 (Th2) cytokine profile [32] and induction of CD4⁺CD25^{high}forkhead box P3 (FoxP3⁺) regulatory T cells (T_{regs}) [33].

MSCs do not express MHC class II and co-stimulatory molecules such as CD80, CD86 or CD40 [9,17,34], and it is believed that T cell activity may result in anergy, which is reflected as immune tolerance. Le Blanc and co-workers reported that when MSCs are treated with IFN- γ , which is

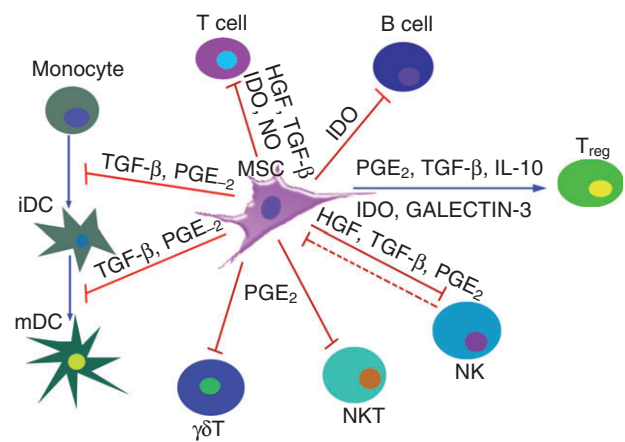


Fig. 1. Effects of mesenchymal stem cells (MSCs) on immunocytes. MSCs modulate the immune response by their interaction with a wide range of immune cells, including T cells, B cells, dendritic cells (DCs), regulatory T cells (T), natural killer (NK), NK T and $\gamma\delta$ T cells. Inhibitory role by MSCs is dependent on cell–cell contact and soluble factors released by MSCs. HGF: hepatocyte growth factor; iDC: immature dendritic cell; IDO: indoleamine 2,3-dioxygenase; IL-10: interleukin-10; mDC: mature dendritic cell; NO: nitric oxide; PGE₂: prostaglandin E₂; TGF- β : transforming growth factor β .

up-regulated in inflammation, they express MHC class II [15]. In an experimental arthritis model, MSCs decreased antigen-specific Th1/Th17 cell expansion and decreased the production of cytokines released by Th1/Th17 cells, such as IFN- γ and IL-17, and caused the Th2 cells to increase production of IL-4 [20,28,35] and IL-10 in lymph node joints [36]. T cell inhibition by MSCs is not due to the induction of apoptosis, but by the inhibition of cell division and probably by the production of soluble factors [14]. However, a recent study reported that MSCs could induce apoptosis in activated T cells [CD3(+) and bromodeoxyuridine (BrdU)(+)], but not in the resting T cells [CD3(+) and BrdU(-)]; this leads to marked attenuation of delayed-type hypersensitivity (DTH) response *in vivo* by inducing NO production [37]. Moreover, MSCs can inhibit the cytotoxic effects of antigen-primed cytotoxic T cells (CTLs) [16] by suppressing the proliferation of CTLs, rather than by direct inhibition of cytolytic activity [26,38].

A recent study showed that the negative co-stimulatory molecule B7-H4 was involved in the immunosuppressive effect of MSCs on T cell activation and proliferation via induction of cell cycle arrest and inhibition of the nuclear translocation of nuclear factor (NF)-kappa B [39]. Some studies revealed that the absence of T cell response in the presence of MSCs was transient and could be restored after the removal of MSCs [14,20]; however, others reported that T cell tolerance was induced by MSCs in murine models [35].

Although some of the mechanisms underlying the immunosuppressive effects of MSCs on T cells have been elucidated

previously, the molecular mechanisms underlying this effect remain controversial. It is believed that the mechanisms underlying the suppressive effect of MSCs may differ by species. Ren and colleagues demonstrated that mouse MSCs and human MSCs utilize different effector molecules in suppressing immune reactions [40]. Immunosuppression by human- or monkey-derived MSCs is mediated by IDO, whereas mouse MSCs exert their effect via NO under the same culture conditions. Immunosuppression by human MSCs was not intrinsic, but was induced by inflammatory cytokines and was chemokine-dependent, as it is in mouse [40]. The degree of the suppressive effect depends on the concentration of the MSCs. The high MSC/lymphocyte ratio is associated with the inhibitory effect of MSCs, while a low MSC/lymphocyte ratio is often accompanied by enhanced proliferation [41]. In this setting, MSCs may act synergistically with HLA-DR antigens upon mitogenic stimulation. However, the exact mechanisms need to be investigated further.

MSCs modulate B cell proliferation and function

In murine models, MSCs have been shown to inhibit the proliferation of B cells when stimulated with anti-CD40L and IL-4 [21] or pokeweed mitogen [42]. Similarly, in humans, MSCs have also been shown to inhibit the proliferation of B cells activated with anti-immunoglobulin (Ig) antibodies, anti-CD40L antibody and cytokines (IL-2 and IL-4) [43]. In addition, the B cell functions of antibody production and secretion of the chemokine receptors CXCR4, CXCR5 and CCR7, which are responsible for chemotaxis to CXCL12 and CXCL13, were impaired by MSCs; however, the expression of B cell co-stimulatory molecules and cytokine production were not affected by MSCs [43]. MSCs inhibited the proliferation of B cells only in the presence of IFN- γ , which probably implies that IFN- γ causes MSCs to produce IDO, which in turn suppressed the proliferation of effector cells through the tryptophan pathway [24] (Fig. 1).

The nature of the mechanism involved in this inhibitory effect of MSCs has not yet been elucidated completely. Thus far, the major mechanism of B cell suppression by MSCs is attributed partly to the physical contact between MSCs and B cells and in part to the soluble factors released by MSCs; this leads to the blocking of B cell proliferation in the G0/G1 phase of the cell cycle with no apoptosis [21,42,43], unlike the case with T cells. Deng *et al.* reported that allogeneic MSCs inhibited the activation, proliferation and IgG secretion of B cells in BXS mouse models of human systemic lupus erythematosus (SLE) [44]. In addition, MSCs enhanced the CD40 expression and CD40 ligand ectopic hyperexpression on the B cells of BXS mice [44].

MSCs modulate the functions of natural killer cells

Numerous studies have shown that MSCs suppress NK cell proliferation and IFN- γ production driven by IL-2 or IL-15,

but only partially inhibit the proliferation of activated NK cells [22,26,28,38,45]. Rasmusson *et al.* reported that MSCs did not inhibit the lysis of freshly isolated NK cells [26] and that MSCs were not lysed by allogeneic NK cells [26]. Conversely, Krampera *et al.* reported that NK cells cultured for 4–5 days with IL-2 in the presence of MSCs showed reduced cytolytic potential against K562 target cells and that this suppressive effect might be attributed to the IFN- γ produced by NK cells [24]. Exposure to IFN- γ did not ablate MSC-induced inhibition of T cell proliferation, but triggered the expression of HGF and TGF- β 1 secreted by MSCs at concentrations that suppressed alloresponsiveness [45] (Fig. 1). Furthermore, a study indicated that MSCs suppressed NK cell cytotoxicity against HLA class I-positive cells more effectively than HLA class I-negative cells [22]. Low HLA class I expression makes allogeneic as well as syngeneic MSCs more susceptible to lysis by activated NK cells [22,46,47]. Incubation of MSCs with IFN- γ decreased their susceptibility to NK cell-mediated lysis because of the up-regulation of HLA class I expression on MSCs [46]. The mechanisms underlying the immunosuppressive effects of MSCs are still unclear and several different, sometimes contradictory, theories have been proposed. Soluble factors such as TGF- β 1 and PGE₂ are believed to play a role in the MSC-mediated suppression of NK cell proliferation [22].

The physiological interactions between MSC and NK cells would be the reciprocal effects exerted by the two cell types, in particular the ability of activated NK cells to kill MSC. Some studies showed that IL-2-activated NK cells can effectively lyse MSCs [22,46,47] because MSCs express ligands (ULBP, PVR and nectin-2) that are recognized by activated NK receptors (NKp30, NKG2D and DNAM-1), which in turn trigger NK cell alloreactivity [46,48].

Recently, Prigione *et al.* found that the inhibitory effect of MSCs on the proliferation of invariant NK T (iNK T, V α 24⁺V β 11⁺) and $\gamma\delta$ T (V δ 2⁺) cells in the peripheral blood is mediated by releasing PGE₂, rather than IDO and TGF- β 1 (Fig. 1); however, cytokine production and cytotoxic activity of the cells were only partially affected by MSCs [49]. V δ 2⁺ cells also serve as professional antigen-presenting cells for naive CD4⁺ T cell response, and MSCs did not inhibit antigen processing/presentation by activated V δ 2⁺ T cells to CD4⁺ T cells [49].

Interaction between MSCs and dendritic cells (DCs)

MSCs impaired the differentiation of monocytes or CD34⁺ haematopoietic stem cells into dendritic cells (DCs) by inhibiting the response of the former to maturation signals, reducing the expression of co-stimulatory molecules and hampering the ability of the former to stimulate naive T cell proliferation and IL-12 secretion [23,50,51]. In addition, this inhibitory effect might be mediated via soluble factors and may be dose-dependent [50]. Spaggiari *et al.* showed that MSCs strongly inhibited the maturation and functioning of

monocyte-derived DCs by interfering selectively with the generation of immature via inhibitory mediator of MSC-derived PGE₂, but not IL-6 [52] (Fig. 1). However, the mechanism underlying the up-regulation of PGE₂ in monocyte–MSC co-cultures remains unclear. Ramasamy *et al.* reported that the cell cycle in DCs was arrested in the G0/G1 phase upon interaction with MSCs [53]. A recent study reported that MSCs isolated from human adipose tissue were more potent immunomodulators for the differentiation of human DCs than MSCs derived from the bone marrow [54].

T_{regs} induced by MSCs

MSCs may also modulate immune responses via the induction of T_{regs}. MSC can induce the generation of CD4⁺CD25⁺ cells displaying a regulatory phenotype (FoxP3⁺) in mitogen-stimulated cultures of peripheral blood mononuclear cells [28,38], although the functional properties of these cells have not yet been elucidated. For example, depletion of CD4⁺CD25⁺ T_{regs} had no effect on the inhibition of T cell proliferation by MSCs [20]. However, a recent study reported that MSCs could induce kidney allograft tolerance by inducing the generation of CD4⁺CD25⁺FoxP3⁺ T_{regs} *in vivo* [55]. Additionally, MSCs have been reported to induce the formation of CD8⁺ T_{regs} that are responsible for the inhibition of allogeneic lymphocyte proliferation [19].

In a recent study, Ghannam *et al.* found that under inflammatory conditions, MSCs prevented the differentiation of naive CD4⁺ T cells into Th17 cells and inhibited the function of Th17 cells *in vitro* by secreting PGE₂. Moreover, MSCs could induce the T_{reg} phenotype in Th17 cells, which can inhibit the proliferative responses of activated CD4⁺ T cells *in vitro* [56]. Tipnis *et al.* reported that umbilical cord-derived MSCs (UC-MSCs) constitutively express B7-H1, which is a negative regulator of T cell activation. In addition, B7-H1 expression was increased and IDO expression was induced in UC-MSCs after IFN- γ treatment. Furthermore, UC-MSCs inhibited the differentiation and maturation of monocyte-derived DCs and augmented the generation of T_{regs} [57]. These immunosuppressive effects of UC-MSCs are mediated largely by cell–cell contact [57].

The induction of T_{regs} by MSCs involves not only involves direct contact between MSCs and CD4⁺ cells, but also the secretion of soluble factors such as PGE₂ and TGF- β 1 [58]. Human gingiva-derived MSCs (GMSCs) can induce IL-10, IDO, inducible NO synthase (iNOS) and cyclooxygenase 2 (COX-2) and thereby serve as immunomodulatory components in the treatment of experimental inflammatory diseases [59] (Fig. 1). A study has shown that the immunosuppressive effect of MSCs is mediated by the secretion of galectin-3, a protein known to modulate T cell proliferation, gene expression, cell adhesion and migration [60]. Madec *et al.* reported that MSCs prevent autoimmune B cell destruction and subsequent diabetes in NOD mice by induc-

ing T_{regs} [61]. The effect of MSCs in the treatment of autoimmune diseases may be through the induction of *de novo* generation of antigen-specific CD4⁺CD25⁺FoxP3⁺ T_{reg} cells [36,62].

However, a recent study reported that MSCs could sustain or suppress T cell proliferation depending on their concentration, and a low MSC/T-cell ratio might support T cell proliferation [63]. A recent study indicated that MSCs could stimulate the activation and proliferation of resting T cells and generate T_{regs} [64]. These data suggested that the culture conditions play an important role in the clinical application of MSCs [63].

Clinical application of MSCs for immune-mediated diseases

Over the past 3 decades, numerous efforts have been made to explore the therapeutic applicability of MSCs. In pathological conditions, MSCs migrate preferentially into lymphoid organs, allografts, injured and/or inflammatory tissue sites after systemic transfusion, where MSCs interact with the activated immune cells and modulate their function [65,66]. The *in vivo* immunomodulatory properties of MSCs were first described in a baboon model of skin transplantation [18]. The therapeutic potential of MSCs in immunomodulation is being explored currently in several Phase I, II and III clinical trials [67], many of which have recently been completed or are under way, as reported in the clinical trials website of the United States sponsored by the National Institutes of Health (<http://clinicaltrials.gov>). Because of their immunosuppressive properties, MSCs are believed to play a role in the maintenance of peripheral tolerance and the induction of transplantation tolerance, and they are considered potential candidates in cellular therapy for graft-*versus*-host disease (GVHD) and autoimmune diseases and in protecting solid-organ grafts from being rejected [39]. Table 1 presents data on the several applications of MSCs as immunosuppressants as studied in clinical trials (data available at the time of the preparation of this manuscript). Recently, Le Blanc *et al.* reported that MSCs obtained from HLA-identical sibling donors, haploidentical donors and third-party HLA-mismatched donors infused in 55 patients with steroid-refractory acute GVHD elicited a response in more than half the patients; the study showed that MSCs exerted their therapeutic effect in the case of both HLA-matched and HLA-unmatched donors. However, for GVHD, the use of MSCs is a double-edged sword, because the prevention of GVHD was associated with a high incidence of leukaemia relapse, which is the result of the non-specific immunosuppressive effect of MSCs on graft-*versus*-leukaemia [68,69]. Liang *et al.* reported that allogeneic MSC transplantation in patients with refractory SLE resulted in the amelioration of disease activity, improvement in the levels of serological markers and stabilization of renal function without the occurrence of serious adverse events [70].

Table 1. Clinical trials using mesenchymal stem cells (MSCs) as immunosuppressants.

Clinical trial	Disease	Cell type/source	Status	Sponsor	ClinicalTrials.gov identifier
Autologous mesenchymal stem cells from adipose tissue in patients with secondary progressive multiple sclerosis	Multiple sclerosis	Adipose tissue-derived autologous MSCs	Recruiting	Fundacion Progreso y Salud, Spain	NCT01056471
Mesenchymal stem cell infusion as prevention for graft rejection and GVHD	GVHD	BM-derived autologous MSCs	Recruiting	University Hospital of Liege, Belgium	NCT00504803
Mesenchymal stem cell transplantation in the treatment of chronic allograft nephropathy	Chronic allograft nephropathy	BM-derived autologous MSCs	Not yet recruiting	Fuzhou General Hospital, China	NCT00659620
Mesenchymal stem cells and subclinical rejection	Renal transplantation	BM-derived allogenic MSCs	Not yet recruiting	Leiden University Medical Center, the Netherlands	NCT00734396
Safety and efficacy study of umbilical cord blood-derived mesenchymal stem cells to promote engraftment of unrelated haematopoietic stem cell transplantation	GVHD	Human umbilical cord blood-derived MSCs	Not yet recruiting	Medipost Co. Ltd, Korea	NCT00823316
Safety and efficacy study of allogenic mesenchymal stem cells to treat extensive chronic GVHD	Chronic GVHD	BM-derived allogenic MSCs	Not yet recruiting	Guangdong General Hospital, China	NCT00972660
Evaluation of the role of mesenchymal stem cells in the treatment of GVHD	Steroid-resistant GVHD	BM-derived autologous MSCs	Recruiting	Christian Medical College, Vellore, India	NCT00314483
Mesenchymal stem cell infusion as treatment for steroid-resistant acute GVHD or poor graft function	GVHD, poor graft function	BM-derived allogenic MSCs	Recruiting	University Hospital of Liege, Belgium	NCT00603330
Safety and efficacy study of adult human mesenchymal stem cells to treat acute GVHD	GVHD	BM-derived allogenic MSC (prochymal)	Completed	Osiris Therapeutics, USA	NCT00136903
Treatment of refractory (acute or chronic) GVHD by the infusion of expanded <i>in-vitro</i> allogenic mesenchymal stem cell	GVHD	BM-derived allogenic MSCs	Recruiting	University of Salamanca, Spain	NCT00447460
Mesenchymal stem cells for treatment of amyotrophic lateral sclerosis	Amyotrophic lateral sclerosis	Adipose tissue-derived autologous MSCs	Not yet recruiting	Mayo Clinic, USA	NCT01142856
Mesenchymal stem cells under basiliximab/low-dose RATG to induce renal transplant tolerance	Kidney transplant	BM-derived autologous MSCs	Recruiting	Mario Negri Institute for Pharmacological Research, Italy	NCT00752479
Mesenchymal stem cells transplantation for refractory systemic lupus erythematosus	Refractory systemic lupus erythematosus	BM-derived allogenic MSCs	Recruiting	Nanjing Medical University, China	NCT00698191
Efficacy and safety of adult human mesenchymal stem cells to treat patients who have failed to respond to steroid treatment for acute GVHD	GVHD	BM-derived allogenic MSC (prochymal)	Completed	Osiris Therapeutics, USA	NCT00366145
Donor mesenchymal stem cell infusion in treating patients with acute or chronic GVHD after undergoing a donor stem cell transplant	GVHD	BM-derived allogenic MSCs	Not yet recruiting	Case Comprehensive Cancer Center	NCT00361049
Safety and efficacy of prochymal for the salvage of treatment-refractory acute GVHD patients	GVHD	BM-derived allogenic MSCs	Completed	Osiris Therapeutics, USA	NCT00284986
Safety and efficacy of human mesenchymal stem cells for treatment of liver failure	Liver failure	Human umbilical cord-derived MSCs	Recruiting	Beijing 302 Hospital, China	NCT01218464
Umbilical cord mesenchymal stem cells for immune reconstitution in HIV-infected patients	HIV	Human umbilical cord-derived MSCs	Recruiting	Beijing 302 Hospital, China	NCT01213186

BM: bone marrow; GVHD: graft-versus-host disease; HIV: human immunodeficiency virus; RATG: rabbit anti-human thymocyte globulin.

For solid organ transplantation, the beneficial effect of MSC-based immunosuppressive therapy is debatable. The application of calcineurine inhibitors (CNIs) would abrogate the immunosuppressive effect of MSC therapy. In addition, CNIs cause renal failure, hypertension and hyperglycaemia and increase the risk of malignancy; therefore, efforts have been made to minimize the use of CNIs treatment in organ transplantation protocols. Consequently, it may be worthwhile to compare the usefulness of combining CNI treatment and MSC therapy in organ transplantation [71]. Conversely, non-selective immunosuppression would have affected patients' antiviral immunity equally [70].

In the future, well-designed preclinical trials should be conducted to explore the clinical applicability of MSCs. Thereafter, randomized trials comparing treatment with infusions of MSCs and conventional drug-based therapies should be undertaken to confirm the therapeutic potential of these cells, as it is important not to overestimate the potential therapeutic effects of MSCs. Information gathered over such studies would help to develop innovative cell-based therapies for the treatment of diseases characterized by exaggerated immune responses.

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References

- Herzog EL, Chai L, Krause DS. Plasticity of marrow-derived stem cells. *Blood* 2003; **102**:3483–93.
- Prockop DJ, Gregory CA, Spees JL. One strategy for cell and gene therapy: harnessing the power of adult stem cells to repair tissues. *Proc Natl Acad Sci USA* 2003; **100** (Suppl. 1):11917–23.
- Gronthos S, Franklin DM, Leddy HA, Robey PG, Storms RW, Gimble GM. Surface protein characterization of human adipose tissue-derived stromal cells. *J Cell Physiol* 2001; **189**:54–63.
- Erices A, Conget P, Minguell JJ. Mesenchymal progenitor cells in human umbilical cord blood. *Br J Haematol* 2000; **109**:235–42.
- in't Anker PS, Scherjon SA, Kleijburg-van der Keur C *et al.* Amniotic fluid as a novel source of mesenchymal stem cells for therapeutic transplantation. *Blood* 2003; **102**:1548–9.
- in't Anker PS, Noort WA, Kruisselbrink AB *et al.* Nonexpanded primary lung and bone marrow-derived mesenchymal stem cells promote the engraftment of umbilical cord blood-derived CD34(+) cells in the NOD/SCID mice. *Exp Hematol* 2003; **31**:881–9.
- Friedenstein AJ, Petrakova KV, Kurolesova AI, Frolova GP. Heterotopic of bone marrow. Analysis of precursor cells for osteogenic and hematopoietic tissues. *Transplantation* 1968; **6**:230–47.
- Friedenstein AJ. Precursor cells of mechanocytes. *Int Rev Cytol* 1976; **47**:327–59.
- Pittenger MF, Mackay AM, Beck SC *et al.* Multilineage potential of adult human mesenchymal stem cells. *Science* 1999; **284**:143–7.
- Reyes M, Lund T, Lenvik T, Aguiar D, Koodie L, Verfaillie CM. Purification and *ex vivo* expansion of postnatal human marrow mesodermal progenitor cells. *Blood* 2001; **98**:2615–25.
- Horwitz EM, Le Blanc K, Dominici M *et al.* Clarification of the nomenclature for MSC: the International Society for Cellular Therapy position statement. *Cytotherapy* 2005; **7**:393–5.
- Dominici M, Le Blanc K, Mueller I *et al.* Minimal criteria for defining multipotent mesenchymal stromal cells. The International Society for Cellular Therapy position statement. *Cytotherapy* 2006; **8**:315–17.
- Rasmusson I. Immune modulation by mesenchymal stem cells. *Exp Cell Res* 2006; **312**:2169–79.
- Di Nicola M, Carlo-Stella C, Magni M *et al.* Human bone marrow stromal cells suppress T lymphocyte proliferation induced by cellular or nonspecific mitogenic stimuli. *Blood* 2002; **99**:3838–43.
- Le Blanc K, Tammik L, Sundberg B, Haynesworth SE, Ringden O. Mesenchymal stem cells inhibit and stimulate mixed lymphocyte cultures and mitogenic responses independently of the major histocompatibility complex. *Scand J Immunol* 2003; **57**:11–20.
- Potian JA, Aviv H, Ponzio NM, Harrison JS, Rameshwar P. Veto-like activity of mesenchymal stem cells: functional discrimination between cellular responses to alloantigens and recall antigens. *J Immunol* 2003; **171**:3426–34.
- Tse WT, Pendleton JD, Beyer WM, Egalka MC, Guinan EC. Suppression of allogeneic T-cell proliferation by human marrow stromal cells: implications in transplantation. *Transplantation* 2003; **75**:389–97.
- Bartholomew A, Sturgeon C, Siatskas M *et al.* Mesenchymal stem cells suppress lymphocyte proliferation *in vitro* and prolong skin graft survival *in vivo*. *Exp Hematol* 2002; **30**:42–8.
- Djouad F, Plence P, Bony C *et al.* Immunosuppressive effect of mesenchymal stem cells favors tumor growth in allogeneic animals. *Blood* 2003; **102**:3837–44.
- Krampera M, Glennie S, Dyson J *et al.* Bone marrow mesenchymal stem cells inhibit the response of naive and memory antigen-specific T cells to their cognate peptide. *Blood* 2003; **101**:3722–9.
- Glennie S, Soeiro I, Dyson PJ, Lam EW, Dazzi F. Bone marrow mesenchymal stem cells induce division arrest anergy of activated T cells. *Blood* 2005; **105**:2821–7.
- Sotiropoulou PA, Perez SA, Gritzapis AD, Baxeivanis CN, Papamichail M. Interactions between human mesenchymal stem cells and natural killer cells. *Stem Cells* 2006; **24**:74–85.
- Jiang XX, Zhang Y, Liu B *et al.* Human mesenchymal stem cells inhibit differentiation and function of monocyte-derived dendritic cells. *Blood* 2005; **105**:4120–6.
- Krampera M, Cosmi L, Angeli R *et al.* Role for interferon-gamma in the immunomodulatory activity of human bone marrow mesenchymal stem cells. *Stem Cells* 2006; **24**:386–98.
- Groh ME, Maitra B, Szekely E, Koc ON. Human mesenchymal stem cells require monocyte-mediated activation to suppress alloreactive T cells. *Exp Hematol* 2005; **33**:928–34.
- Rasmusson I, Ringden O, Sundberg B, Le Blanc K. Mesenchymal stem cells inhibit the formation of cytotoxic T lymphocytes, but not activated cytotoxic T lymphocytes or natural killer cells. *Transplantation* 2003; **76**:1208–13.
- Meisel R, Zibert A, Laryea M, Gobel U, Daubener W, Dilloo D. Human bone marrow stromal cells inhibit allogeneic T-cell

- responses by indoleamine 2,3-dioxygenase mediated tryptophan degradation. *Blood* 2004; **103**:4619–21.
- 28 Aggarwal S, Pittenger MF. Human mesenchymal stem cells modulate allogeneic immune cell responses. *Blood* 2005; **105**:1815–22.
- 29 Sato K, Ozaki K, Oh I *et al.* Nitric oxide plays a critical role in suppression of T-cell proliferation by mesenchymal stem cells. *Blood* 2007; **109**:228–34.
- 30 Bingisser RM, Tilbrook PA, Holt PG, Kees UR. Macrophage-derived nitric oxide regulates T cell activation via reversible disruption of the Jak3/STAT5 signaling pathway. *J Immunol* 1998; **160**:5729–34.
- 31 Ding Y, Xu D, Feng G, Bushell A, Muschel RJ, Wood KJ. Mesenchymal stem cells prevent the rejection of fully allogeneic islet grafts by the immunosuppressive activity of matrix metalloproteinase-2 and -9. *Diabetes* 2009; **58**:1797–806.
- 32 Carosella ED, Moreau P, Le Maoult J, Le Discorde M, Dausset J, Rouas-Freiss N. HLA-G molecules: from maternal–fetal tolerance to tissue acceptance. *Adv Immunol* 2003; **81**:199–252.
- 33 Selmani Z, Naji A, Zidi I *et al.* Human leukocyte antigen-G5 secretion by human mesenchymal stem cells is required to suppress T lymphocyte and natural killer function and to induce CD4⁺CD25^{high}FOXP3⁺ regulatory T cells. *Stem Cells* 2008; **26**:212–22.
- 34 Deans RJ, Moseley AB. Mesenchymal stem cells: biology and potential clinical uses. *Exp Hematol* 2000; **28**:875–84.
- 35 Zappia E, Casazza S, Pedemonte E *et al.* Mesenchymal stem cells ameliorate experimental autoimmune encephalomyelitis inducing T cell anergy. *Blood* 2005; **106**:1755–61.
- 36 González MA, Gonzalez-Rey E, Rico L, Büscher D, Delgado M. Treatment of experimental arthritis by inducing immune tolerance with human adipose-derived mesenchymal stem cells. *Arthritis Rheum* 2009; **60**:1006–19.
- 37 Lim JH, Kim JS, Yoon IH *et al.* Immunomodulation of delayed-type hypersensitivity responses by mesenchymal stem cells is associated with bystander T cell apoptosis in the draining lymph node. *J Immunol* 2010; **185**:4022–9.
- 38 Maccario R, Podesta M, Moretta A *et al.* Interaction of human mesenchymal stem cells with cells involved in alloantigen-specific immune response favors the differentiation of CD4⁺ T-cell subsets expressing a regulatory/suppressive phenotype. *Haematologica* 2005; **90**:516–25.
- 39 Sensebé L, Krampera M, Schrezenmeier H, Bourin P, Giordano R. Mesenchymal stem cells for clinical application. *Vox Sang* 2010; **98**:93–107.
- 40 Ren G, Su J, Zhang L *et al.* Species variation in the mechanisms of mesenchymal stem cell-mediated immunosuppression. *Stem Cells* 2009; **27**:1954–62.
- 41 Le Blanc K, Ringdén O. Immunomodulation by mesenchymal stem cells and clinical experience. *J Intern Med* 2007; **262**:509–25.
- 42 Augello A, Tasso R, Negrini SM *et al.* Bone marrow mesenchymal progenitor cells inhibit lymphocyte proliferation by activation of the programmed death 1 pathway. *Eur J Immunol* 2005; **35**:1482–90.
- 43 Corcione A, Benvenuto F, Ferretti E *et al.* Human mesenchymal stem cells modulate B-cell functions. *Blood* 2006; **107**:367–72.
- 44 Deng W, Han Q, Liao L, You S, Deng H, Zhao RC. Effects of allogeneic bone marrow-derived mesenchymal stem cells on T and B lymphocytes from BXSb mice. *DNA Cell Biol* 2005; **24**:458–63.
- 45 Ryan JM, Barry F, Murphy JM, Mahon BP. Interferon-gamma does not break, but promotes the immunosuppressive capacity of adult human mesenchymal stem cells. *Clin Exp Immunol* 2007; **149**:353–63.
- 46 Spaggiari GM, Capobianco A, Becchetti S, Mingari MC, Moretta L. Mesenchymal stem cell natural killer cell interactions: evidence that activated NK cells are capable of killing MSCs, whereas MSCs can inhibit IL-2-induced NK-cell proliferation. *Blood* 2006; **107**:1484–90.
- 47 Poggi A, Prevosto C, Massaro AM *et al.* Interaction between human NK cells and bone marrow stromal cells induces NK cell triggering: role of NKP30 and NKG2D receptors. *J Immunol* 2005; **175**:6352–60.
- 48 Moretta A, Bottino C, Vitale M *et al.* Activating receptors and coreceptors involved in human natural killer cell-mediated cytotoxicity. *Annu Rev Immunol* 2001; **19**:197–23.
- 49 Prigione I, Benvenuto F, Bocca P, Battistini L, Uccelli A, Pistoia V. Reciprocal interactions between human mesenchymal stem cells and gammadelta T cells or invariant natural killer T cells. *Stem Cells* 2009; **27**:693–702.
- 50 Nauta AJ, Kruisselbrink AB, Lurvink E, Willemze R, Fibbe WE. Mesenchymal stem cells inhibit generation and function of both CD34⁺-derived and monocyte-derived dendritic cells. *J Immunol* 2006; **177**:2080–7.
- 51 Zhang W, Ge W, Li C *et al.* Effects of mesenchymal stem cells on differentiation, maturation, and function of human monocyte-derived dendritic cells. *Stem Cells Dev* 2004; **13**:263–71.
- 52 Spaggiari GM, Abdelrazik H, Becchetti F, Moretta L. MSCs inhibit monocyte-derived DC maturation and function by selectively interfering with the generation of immature DCs: central role of MSC-derived prostaglandin E2. *Blood* 2009; **113**:6576–83.
- 53 Ramasamy R, Fazekasova H, Lam EW, Soeiro I, Lombardi G, Dazzi F. Mesenchymal stem cells inhibit dendritic cell differentiation and function by preventing entry into the cell cycle. *Transplantation* 2007; **83**:71–6.
- 54 Ivanova-Todorova E, Bochev I, Mourdjeva M *et al.* Adipose tissue-derived mesenchymal stem cells are more potent suppressors of dendritic cells differentiation compared to bone marrow-derived mesenchymal stem cells. *Immunol Lett* 2009; **126**:37–42.
- 55 Ge W, Jiang J, Arp J, Liu W, Garcia B, Wang H. Regulatory T-cell generation and kidney allograft tolerance induced by mesenchymal stem cells associated with indoleamine 2,3-dioxygenase expression. *Transplantation* 2010; **90**:1312–20.
- 56 Ghannam S, Pène J, Torcy-Moquet G, Jorgensen C, Yssel H. Mesenchymal stem cells inhibit human Th17 cell differentiation and function and induce a T regulatory cell phenotype. *J Immunol* 2010; **185**:302–12.
- 57 Tipnis S, Viswanathan C, Majumdar AS. Immunosuppressive properties of human umbilical cord-derived mesenchymal stem cells: role of B7-H1 and IDO. *Immunol Cell Biol* 2010; **88**:795–806.
- 58 English K, Ryan JM, Tobin L, Murphy MJ, Barry FP, Mahon BP. Cell contact, prostaglandin E(2) and transforming growth factor beta 1 play non-redundant roles in human mesenchymal stem cell induction of CD4⁺CD25(High) forkhead box P3⁺ regulatory T cells. *Clin Exp Immunol* 2009; **156**:149–60.
- 59 Zhang Q, Shi S, Liu Y *et al.* Mesenchymal stem cells derived from human gingiva are capable of immunomodulatory functions and ameliorate inflammation-related tissue destruction in experimental colitis. *J Immunol* 2009; **183**:7787–98.
- 60 Sioud M, Mobergslén A, Boudabous A, Fløisand Y. Evidence for the involvement of galectin-3 in mesenchymal stem cell

- suppression of allogeneic T-cell proliferation. *Scand J Immunol* 2010; **71**:267–74.
- 61 Madec AM, Mallone R, Afonso G *et al.* Mesenchymal stem cells protect NOD mice from diabetes by inducing regulatory T cells. *Diabetologia* 2009; **52**:1391–9.
- 62 Gonzalez-Rey E, Gonzalez MA, Varela N *et al.* Human adipose-derived mesenchymal stem cells reduce inflammatory and T cell responses and induce regulatory T cells *in vitro* in rheumatoid arthritis. *Ann Rheum Dis* 2010; **69**:241–8.
- 63 Najjar M, Rouas R, Raicevic G *et al.* Mesenchymal stromal cells promote or suppress the proliferation of T lymphocytes from cord blood and peripheral blood: the importance of low cell ratio and role of interleukin-6. *Cytotherapy* 2009; **11**:570–83.
- 64 Crop M, Baan CC, Korevaar SS, Ijzermans JN, Weimar W, Hoogduijn MJ. Human adipose tissue-derived mesenchymal stem cells induce explosive T-cell proliferation. *Stem Cells Dev* 2010; **19**:1843–53.
- 65 Ge W, Jiang J, Baroja ML *et al.* Infusion of mesenchymal stem cells and rapamycin synergize to attenuate alloimmune responses and promote cardiac allograft tolerance. *Am J Transplant* 2009; **9**:1760–72.
- 66 Hoogduijn MJ, Crop MJ, Peeters AM *et al.* Donor-derived mesenchymal stem cells remain present and functional in the transplanted human heart. *Am J Transplant* 2009; **9**:222–30.
- 67 Salem HK, Thiernemann C. Mesenchymal stromal cells: current understanding and clinical status. *Stem Cells* 2010; **28**:585–96.
- 68 Ning H, Yang F, Jiang M *et al.* The correlation between cotransplantation of mesenchymal stem cells and higher recurrence rate in hematologic malignancy patients: outcome of a pilot clinical study. *Leukemia* 2008; **22**:593–9.
- 69 Vianello F, Dazzi F. Mesenchymal stem cells for graft-versus-host disease: a double edged sword? *Leukemia* 2008; **22**:463–5.
- 70 Liang J, Zhang H, Hua B *et al.* Allogenic mesenchymal stem cells transplantation in refractory systemic lupus erythematosus: a pilot clinical study. *Ann Rheum Dis* 2010; **69**:1423–9.
- 71 Popp FC, Renner P, Eggenhofer E *et al.* Mesenchymal stem cells as immunomodulators after liver transplantation. *Liver Transpl* 2009; **15**:1192–8.