An introduction to mesenchymal stem cell modulation of T cell-mediated immune responses

T lymphocytes (T cells) are the primary cellular effectors of the adaptive immune system and their functional properties are central to antigen specificity and memory associated with cognate immunity [1-3]. Antigen-specific activation and differentiation of naïve T cells result in the generation of a range of T-cell phenotypes that may be defined by the acquisition of characteristic cytokine secretion profiles, cytolytic mechanisms, or counter-regulatory properties [1-3]. In the wake of antigen-specific adaptive immune responses, a small proportion of activated T cells persist as memory cells and have the capacity to respond more rapidly and potently to secondary encounters with the same antigen [1,3]. These memory cells may retain the effector phenotype imprinted upon them during primary activation [1]. When these memory cells are appropriately coordinated and regulated, the diversity of T-cell effector phenotypes allows immune protection against a multitude of pathogenic microorganisms while maintaining self-tolerance and homeostasis [2]. On the other hand, overexuberant pro-inflammatory T-cell responses may lead to autoimmune and allergic diseases, including multiple sclerosis, inflammatory bowel disease, type 1 diabetes mellitus, and asthma [4-7]. Furthermore, life-saving treatments such as
allogeneic bone marrow (BM) and solid organ transplantation may be complicated by alloantigen-specific T-cell immune responses, resulting in graft-versus-host disease (GvHD) or transplant rejection [8].

Mesenchymal stem (or stromal) cells (MSCs) are a heterogeneous population of fibroblast-like progenitor cells that may be isolated and expanded from BM, umbilical cord, fat, gingiva, and other tissues [9]. They have the capacity to self-renew and differentiate into various mesodermal cell lineages, including adipocytes, osteocytes, and chondrocytes under controlled culture conditions [9]. In the past two decades, MSCs have garnered considerable attention for their potential use as regenerative therapeutic agents in a range of acute and chronic diseases [8-11]. Mechanistically, the beneficial effects of MSC therapies have been more frequently linked to their ‘trophic’ (paracrine) effects rather than their ability to transdifferentiate [11]. Specifically, MSCs are now viewed as having potent anti-inflammatory and immune-modulating properties that, in many studies, have been shown to be associated with inhibition of effector T-cell activation with or without a concomitant increase in regulatory T cell (T_{reg}) numbers [4,6,10-12]. The T-cell suppressive effects of MSCs were initially described over a decade ago [13] and have since been reported consistently for both CD4+ T helper (Th) cells and CD8+ cytotoxic T lymphocytes (CTLs) [8,11,14]. Suppression of T cells by MSCs may be direct or may occur indirectly via modulatory effects on antigen-presenting cells such as dendritic cells (DCs), resulting in altered cytokine expression and impaired antigen presentation [15-17]. MSCs themselves demonstrate a lack of stimulatory capacity toward T cells [18,19]. MSCs isolated from various sources (BM, adipose tissue, and Wharton's jelly) have been reported to equally suppress proliferation of CD4+ and CD8+ T-cell subsets in a dose-dependent fashion [20]. Reported roles for both cell-cell contact and release of soluble factors in MSC-mediated T-cell suppression are evident throughout the literature, and numerous candidate mediators have been reported: prostaglandin E_{2} (PGE_{2}), indoleamine-2,3-dioxygenase, nitric oxide, interleukin (IL)-27, transforming growth factor-beta (TGF-β), monocyte chemotactic protein 1 (MCP-1/CC12), human leukocyte antigen G, and intracellular adhesion molecule 1 among others [8,10-12,17,21-24]. The abundance of mediators identified to date suggests that MSCs exploit different immunosuppressive mechanisms under different disease conditions. MSC therapy has been successful in a range of disease models and some clinical conditions known to be associated with damaging effector T-cell responses or failure of T_{reg}-mediated counter-regulation or both [4,6,8,11,22,25-28]. Overall, it is now very well established that MSCs exert diverse and potent modulatory effects on the T-cell compartment of the immune system, most of which are suppressive in nature and of potential therapeutic value. Nevertheless, some significant controversies and a basic lack of information regarding the range of effects that MSCs have on individual T-cell effector subsets remain.

In the remaining sections of this review, we focus on the most recent data related to MSC modulation of individual well-defined Th cell and CTL effector phenotypes in vitro and in vivo. Where possible, we emphasize the relevance of current knowledge on this topic to diseases for which MSC therapy is perceived to be beneficial. We also highlight key gaps in our understanding and important unanswered questions that may be the subject of future studies.

**Mesenchymal stem cell modulation of T helper cell subsets**

Th cells are cytokine-producing CD4+ cells that recognize peptides presented to them by major histocompatibility complex (MHC) class II molecules [1-3]. Differentiation of Th cells into effector cells depends largely on the cytokine milieu present at the time of antigen presentation and activation [1-3]. In the context of this article, we will focus on reviewing recent progress (summarized in Figure 1) in understanding MSC effects on the well-described subsets Th1, Th2, and Th17 cells, T_{reg}, and CTLs with emphasis, where possible, on mechanistic and disease-specific in vivo studies.

**T helper type 1 cells**

Th1 cell induction occurs when CD4+ T cells are activated in the presence of IL-12, interferon-gamma (IFN-γ), and IL-27 [29]. IFN-γ is the characteristic cytokine produced by Th1 cells in addition to tumor necrosis factor (TNF). Known effector functions of Th1 cells include activation and recruitment of macrophages to sites of inflammation and induction of immunoglobulin (Ig) G2a production by B cells [2]. Th1 cells are responsible for the clearance of intracellular pathogens and delayed-type hypersensitivity (DTH) reactions by amplifying cellular immunity [29]. DTH reactions are mediated by both Th1 cells and CTLs [25], and, through their role as coordinators of this form of immune response, Th1 cells have the capacity to cause maladaptive tissue damage. Examples of Th1 cell-mediated inflammatory and autoimmune diseases are type 1 diabetes mellitus and Crohn's disease [7]. The literature to date indicates that MSCs exert primarily suppressive effects on Th1 cell differentiation and effector function, and evidence favors predominantly indirect mechanisms. In vitro, the generation of Th1 cells is reduced in mixed lymphocyte cultures containing MSCs or MSC-conditioned medium, likely due to inhibition of Th1 cell-stimulating properties of DCs [30]. In the in vivo setting, Lim and colleagues [25] recently demonstrated that...
infusion of MSCs attenuated cutaneous DTH in mice and that this effect was associated with reduced infiltration of CD4+ and CD8+ T cells at the challenge site and increased apoptosis of activated T cells in the draining lymph nodes. MSCs were detected close to the germinal center and paracortical region in lymph nodes [25], suggesting that they modulate immune responses directly in the area where DCs are likely to activate T cells. In experimental colitis (an animal model of inflammatory bowel disease), dose-dependent xenogenic, allogenic, and autologous adipose-derived MSCs ameliorated disease activity and were specifically associated with reduced IFN-γ-producing Th1 cells in association with increased numbers of forkhead box P3 (FOXP3)-expressing T cells (Treg) [7]. Furthermore, when treated with either total CD4+ T cells or Treg-depleted CD4+ T cells from mesenteric lymph nodes of MSC-treated colitic mice, mice with induced colitis demonstrated attenuated and enhanced colitis, respectively [7]. These data suggest that the administration of MSCs in experimental colitis dampens Th1 cell responses via induction of Treg but does not eliminate Th1 cells entirely. MSCs have also been used experimentally to effectively prevent or treat Th1 cell-mediated autoimmune diabetes mellitus in streptozotocin-treated rats and in nonobese diabetic (NOD) mice [31,32]. In the rat model, the protective effects observed following administration of MSCs were shown to be associated with increased IL-10 and IL-13 expression by T cells and with increased frequencies of both CD4+ and CD8+ FOXP3+ T cells as opposed to a direct reduction of IFN-γ-producing T cells [31]. In NOD mice, a single MSC injection minimized beta-cell destruction following

### Table:MSC Effects on Disease

| Disease Relevance | MSC Effects |
|-------------------|-------------|
| Allergic Asthma.  | Reduced IL-4 and IL-13 production in Th2-dominated allergic autoimmune disease. |
| Systemic Lupus Erythematosus. | Increased Th2 relative to Th1 and Th17 activity in non-Th2-dominated autoimmunity and allograft models. |
| Chronic GvHD.    | Potent in vitro and in vivo suppression mediated indirectly by modification of DC and induction/protection of Treg. |
| Multiple Sclerosis. | Increased proportion of FOXP3+CD4+ regulatory T-cells in primary mouse and human stimulation cultures. |
| Type I Diabetes. | Reduced generation of antigen-specific CTL in primary stimulation cultures. |
| Inflammatory Bowel Disease. | Inhibited proliferation and target cell lysis. |
| Acute GvHD.      | Low susceptibility of allo-MSC to lysis by CTL. |
| Allotransplant Rejection. | Limited or absent suppression of lysis by viral antigen-specific, memory CTL in vitro and in vivo. |

### Diagram:CD4+ and CD8+ T-cell Effector Subtypes

- **CD4+ Naïve T**: naïve CD4+ T cells activate Th2, Th1, Th17, and iTreg, respectively, via CD8+ T cells.
- **CD8+ Naïve T**: naïve CD8+ T cells activate Th17 and iTreg, respectively, via CD4+ T cells.

**Figure 1.** Diagrammatic summary of the major CD4+ and CD8+ T-cell effector subtypes (as derived from naïve CD4+ and CD8+ T cells), the reported effects of mesenchymal stem cells (MSCs) on these effectors, and some clinically important disease associations for each.

Bidirectional arrows indicate reported inter-conversion (plasticity) between Th1/Th17 phenotypes and Th17/iTreg phenotypes that may be of relevance to MSC immune modulatory effects. Relevant references are indicated numerically for individual statements. CTL, cytotoxic T lymphocyte; DC, dendritic cell; DTH, delayed-type hypersensitivity; FOXP3, forkhead box P3 transcription factor; GvHD, graft-versus-host disease; IFN-γ, interferon-gamma; IL, interleukin; iTreg, induced regulatory T cell; nTreg, natural regulatory T cell; Th1, T helper type 1 cell; Th2, T helper type 2 cell; Th17, T helper type 17 cell; Treg, regulatory T cell.
transfer of diabetogenic T cells. Protection was shown to be associated with MSC migration to pancreatic lymph nodes and with induction of IL-10-producing FOXP3+ T\textsubscript{reg} [32]. These examples from the recent literature indicate that, in clinically relevant disease settings, MSCs consistently suppress harmful autoimmune Th1 cell responses by predominantly indirect mechanisms, including modulation of antigen-presenting DCs and promotion of naturally occurring or induced FOXP3-expressing T\textsubscript{reg}.

**T helper type 2 cells**

Th2 cell differentiation occurs when CD4\textsuperscript{+} T cells are activated in the presence of IL-4, which itself is produced by Th2 cells in addition to IL-5, IL-9, IL-10, and IL-13 [2,3,29]. The role of Th2 cells in adaptive immunity is linked to host defense against extracellular parasites, to antibody class switching to IgG1 and IgE in B cells, and to recruitment of eosinophils [2,3,29,33]. Dysregulated Th2 cell responses are associated with allergic diseases such as asthma [2]. Very few studies have examined MSC effects on immune-mediated diseases in which Th2 cell responses are dominant. However, Kavanagh and Mahon [6] recently reported that allogeneic MSC administration reduced the number of infiltrating eosinophils, suppressed IgE induction, and inhibited IL-13 and IL-4 production in a mouse model of ovalbumin-induced airway inflammation. Additionally, increases in IL-10 and FOXP3 expression were observed in this study, suggesting that MSCs suppress allergen-specific Th2 cell responses in allergic airway inflammation in part via induction of T\textsubscript{reg}. As further evidence of this, depletion of T\textsubscript{reg} resulted in reversal of the protective effects of the MSCs [6]. In human subjects with chronic GvHD (which is also including modulation of antigen-presenting DCs and promotion of naturally occurring or induced FOXP3-expressing T\textsubscript{reg}).

In other circumstances, there is evidence that MSCs may favor the emergence of Th2 phenotype T cells. Bai and colleagues [4] demonstrated, for example, that mice treated with human BM-derived MSCs recovered function from limb paralysis in relapsing-remitting and chronic experimental allergic encephalomyelitis (EAE), a model for multiple sclerosis, by induction of Th2 cells. In this study, neurological improvement was associated with reduced CD45\textsuperscript{+} leukocytic infiltration of the brain and spinal cord, with increased levels of the Th2 cell-related cytokines IL-4 and IL-5, and with potent reduction in the Th1/Th17 cell-related cytokines IL-17, IFN-\gamma, TNF, and IL-12 [4]. The results suggested that MSC administration in EAE favorably altered the balance between pro-inflammatory Th1/Th17 cell and anti-inflammatory Th2 cell responses. Likewise, Fiorina and colleagues [35] reported a shift in Th1/Th2 cell balance toward Th2 cells following allogeneic MSC administration to NOD mice. Further evidence supporting a shift toward Th2 cell responses is provided by Batten and colleagues [18], who describe the use of human BM-derived MSCs for tissue engineering of a heart valve. CD4\textsuperscript{+} T cells co-cultured with MSCs expressed lower levels of IL-1-\alpha and -\beta, TNF, and IFN-\gamma but higher levels of IL-5, IL-8, and IL-3 in response to allogeneic peripheral blood mononuclear cells. Consistent with the findings of Kavanagh and Mahon [6], those of Batten and colleagues [18] additionally indicated increased expression of FOXP3 in CD4\textsuperscript{+} T cells co-cultured with MSCs, suggesting the induction of a T\textsubscript{reg} phenotype. Thus, though relatively limited, the experimental evidence to date suggests that MSCs suppress effector function of Th2 cells in Th2 cell-predominant inflammation. In other T cell-mediated immunological disorders, however, predominant MSC suppression of the Th1 and Th17 cell pathways may result in a relative skewing toward less damaging Th2 and T\textsubscript{reg} phenotypes. Whether MSCs actively induce the differentiation and expansion of Th2 cells during primary or secondary antigen-specific immune responses has not been well tested but appears less likely.

**T helper type 17 cells**

The Th17 cell effector phenotype is defined by preferential secretion of IL-17A (IL-17) along with other cytokines, including IL-17F, IL-21, and IL-22. Th17 cells are pro-inflammatory and protect against extracellular pathogens, including fungi, mycobacteria, and Gram-negative bacteria, via recruitment of neutrophils [36]. Th17 cells may also be pathogenic and have been shown to have an important role in immunological diseases, including rheumatoid arthritis, multiple sclerosis, and inflammatory bowel disease [36]. TGF-\beta and IL-6, with or without IL-21, IL-23, and IL-1, are necessary for the induction and expansion of Th17 cells from naïve CD4\textsuperscript{+} precursors [2,36]. Recently, MSC effects on the Th17 cell differentiation pathway have been examined in mice and humans. Ghannam and colleagues [37] observed that human MSCs induce regulatory characteristics in Th17 cells in an inflammatory environment by downregulating the Th17 cell-specific transcription factor ROR\gamma (retinoic acid receptor-related orphan receptor gamma t) and up-regulating FOXP3. Moreover, when re-purified, these regulatory-phenotype Th17 cells suppressed proliferation of newly initiated CD4\textsuperscript{+} T cells [37]. In vivo, MSC administration has been shown to suppress the development of EAE via a reduction in IL-17 production in the central nervous system along with reduced IFN-\gamma, TNF, and IL-23 and increased TGF-\beta and IL-4 [22]. Inhibition of Th17 cell activity in EAE has also been...
Regulatory T cells

A subset of CD4+ T cells have been identified as having regulatory (suppressor) functions that are essential for the prevention of autoimmunity and the resolution of inflammatory processes. These CD4+ T_{reg} are best characterized by surface expression of the IL-2 receptor alpha chain (CD25) and, more specifically, by intracellular expression of the transcription factor FOXP3. They can be further subdivided into naturally occurring T_{reg} (nT_{reg}) that develop in the thymus or induced T_{reg} (iT_{reg}) that differentiate from naïve peripheral CD4+ T cells in the presence of TGF-β [2]. T_{reg} exert potent immunosuppressive effects via cell-cell contact and production of soluble factors and may negatively regulate the activation of each of the major Th cell subtypes as well as other immune and inflammatory cells [2]. As indicated at several points in the preceding sections, there has been a consistent theme among many in vitro and in vivo studies in support of MSC enhancement of T_{reg} number and activity [8,10]. English and colleagues [17] showed that human FOXP3+CD25^{high} T_{reg} were induced upon coculture of allogeneic MSCs and CD4+ T cells and exerted suppressive activity when re-purified and added to a newly initiated mixed lymphocyte culture. This was corroborated in a study of human adipose tissue-derived MSCs that, in addition to reducing IL-17, TNF, and IFN-γ production, induced IL-10-producing, FOXP3+ T_{reg} in vitro among collagen-specific peripheral blood T cells of patients with rheumatoid arthritis [45]. Upon re-isolation, the T_{reg} originally generated in the presence of MSCs had the capacity to inhibit IFN-γ production and proliferation of a subsequent collagen-stimulated T-cell culture [45]. In in vivo models of kidney, liver, and heart allotransplantation, several laboratories have linked the protective effects observed with MSC therapy directly to the presence of T_{reg} [26,27,46]. The importance of the induced T_{reg} populations in such transplant models, as well as in some models of allergic and autoimmune disease, is well illustrated by the induction of graft rejection or loss of therapeutic benefit following T_{reg} depletion [6,8,26]. One concern that requires further investigation regarding MSC-induced T_{reg} relates to the potential for phenotypic plasticity of pro- and anti-inflammatory CD4+ T-cell subsets under varying in vivo conditions. For example, as highlighted in a review by Afzali and colleagues [47], T_{reg} may be converted to a Th17 cell phenotype when exposed to inflammatory stimuli. In such circumstances, MSC-induced T_{reg} may exacerbate the disease state. Further human studies will be essential to fully elucidate the clinical relevance and robustness of MSC-induced T_{reg} in vivo, as evidenced by the study of Carrió and colleagues [48] in which the presence of MSC-induced T_{reg} did not alter the disease course in two patients with systemic lupus erythematosus.
Mesenchymal stem cell modulation of cytotoxic T lymphocytes

CD8+ CTLs recognize cytosolic antigen-derived peptides presented by MHC class I and thus are essential for the destruction of virus-infected cells and tumor cells. Following their primary activation by professional antigen-presenting cells such as DCs, CTLs induce cell death upon secondary encounter of antigen expressed by any cell type via pro-apoptotic surface receptors or targeted release of cytotoxic granules [3,19]. CTLs also release IFN-γ, TNF, and lymphotixin-α to inhibit viral replication and to recruit macrophages to the site of infection [3]. Death receptor ligands such as TRAIL (TNF-related apoptosis-inducing ligand) and Fas ligand are upregulated on activated CTLs. In patients receiving allogeneic BM or hematopoietic stem cell transplants, activated CTLs mediate GvHD, causing damage to liver, intestine, skin, and other tissues [19]. Therapeutically administered MSCs have the potential to reduce disease severity in GvHD and other immune-mediated diseases via direct effects on CTLs as well as through inhibition of Th cell responses, which are required for full activation of CTLs [3]. For allogeneic MSC administration, the beneficial effects may also be limited by alloantigen-specific CTL-mediated MSC lysis. In a study by Rasmusson and colleagues [19], however, MSCs were resistant to CTL lysis despite the expression of MHC class I on their surface. MSCs were also unable to induce pro-inflammatory cytokine production or CD25 upregulation by CTLs [19]. This same group and others have also demonstrated that MSCs inhibit the formation of CTLs in mixed lymphocyte cultures and prevent CTL-associated lysis of target cells if added during the primary stimulation phase [49,50]. In contrast, MSCs were unable to suppress activated CTLs at the cytotoxic effector phase [50]. Such findings were corroborated by Karlsson and colleagues [5], who showed that, while MSCs potently suppressed primary alloantigen-induced proliferation and IFN-γ production by human peripheral blood leukocytes, they had no effect on cytomegalovirus (CMV)-induced proliferation or IFN-γ production. Furthermore, MSCs were unable to suppress proliferation or cytolytic killing in established CMV- or Epstein-Barr virus-specific CTL lines [5]. These observations are of particular clinical relevance to the treatment of GvHD patients who are at high risk from the reactivation of viral infections. However, in this case, administration of MSCs was ineffective toward CMV-mediated CD8+ T-cell effector functions while potently suppressing alloantigen-induced responses [5].

Interestingly, some of the protective effects of MSCs in GvHD may also result from the generation of CD8+ Treg, as demonstrated by Prevosto and colleagues [51]. It is proposed that CD8+ Treg may amplify the immune modulatory effects of MSCs because, when re-purified from peripheral blood leukocytes/MSC co-cultures, these cells potently suppressed subsequent peripheral blood leukocyte proliferation in response to alloantigen and to the non-specific mitogen phytohaemagglutinin [51]. It is also important to note that MSC-associated immune modulation may have detrimental effects in the setting of cancer. In a recent study by Patel and colleagues [52], the addition of MSCs to co-cultures of breast cancer cells and peripheral blood leukocytes resulted in enhanced Treg numbers and Th2 cell-related cytokines as well as inhibited proliferation and release of granzyme B by CTLs, all of which resulted in protection of cancer cells from immune-mediated lysis. In vivo studies to examine this phenomenon are essential to fully understand the complex interaction between MSCs, T cells, and cancer cells and to ensure that MSC administration is not associated with recurrence or rapid metastasis of cancer in some patient groups. Overall, the experimental and clinical evidence to date suggests that MSCs exert both direct and indirect suppressive effects on the generation of antigen-specific CTLs and may foster the emergence of CD8+ Treg, but do not significantly inhibit the immune surveillance functions of pre-existing CD8+ memory T cells.

Concluding remarks and future directions

The immune suppressive and anti-inflammatory properties of MSCs are now very well established and clearly encompass potent modulatory influences on the generation and disease-associated activity of multiple T-cell effector phenotypes [8-12]. Preclinical models provide a strong impetus for translating MSC therapy to widespread clinical use for a range of common, T-cell-mediated autoimmune diseases and for prevention or treatment of transplant complications such as rejection and GvHD [8,10]. Despite this, a critical review of our current understanding of these effects and of recent developments in MSC clinical trials [28] indicates that much remains to be learned at both the mechanistic and logistic levels. The dizzying array of potential MSC-associated mediators of T-cell suppression, the many differences between small-animal immunological models and human immune-mediated diseases, and the lack of uniformity in MSC culture and administration protocols suggest that a more focused experimental pipeline will be required for the therapeutic potential to be realized in the near future. Table 1 summarizes a number of key questions that, on the basis of the literature to date, we believe to be important for translational progress in this field. In particular, we would highlight the need to better understand conditions in which MSC administration has been found to be ineffective or even harmful during T cell-mediated disease. Zappia and colleagues [38]...
Table 1. Key unanswered or incompletely answered questions about mesenchymal stem cell effects on T-cell effector functions

1. Is there a true hierarchy of suppressive potency for MSC effects on the three major T helper cell effector pathways (for example, Th1 > Th17 > Th2 cells)?
2. Are individual T-cell effector phenotypes susceptible to different MSC mechanisms of suppression, or does suppressive mechanism vary by MSC culture condition or by disease pathogenesis (or by both)?
3. How important is cell-cell contact for the in vivo direct and indirect suppression by MSCs of harmful T-cell effector functions?
4. Is the apparent shift from Th1/Th17 to Th2/Treg responses following MSC therapy in some diseases due to true induction and expansion of anti-inflammatory T-cell effector phenotypes or to their preferential survival?
5. What are the relative effects of MSCs on memory and pre-activated T-cell effectors compared with naïve T cells undergoing primary activation?
6. Do immunosuppressive potency and mechanism of action vary among the individual cells within heterogeneous MSC cultures and can they be enhanced by MSC cloning or modification?
7. To what extent and under what circumstances are MSCs capable of promoting harmful T-cell effector functions?
8. How long-lasting is MSC modulation of antigen-specific T-cell effectors following single or multiple in vivo administration?
9. How can MSC in vivo immune suppressive potency, mechanism of action, and site of migration be better predicted for a given culture preparation prior to therapeutic administration?
10. Does the in vivo immunogenicity of allogeneic MSCs limit their long-term therapeutic benefit in autoimmune disease and allotransplantation?
11. What are the combined in vivo effects of MSC and other anti-T-cell therapies?

MSC, mesenchymal stem cell; Th1, T helper type 1; Th2, T helper type 2; Th17, T helper type 17; Treg, regulatory T cell.

demonstrated, for example, that the time of administration of MSCs was a critical parameter for successful treatment of EAE since MSC administration prior to or during the early disease course was effective whereas MSC benefit was lost once central nervous system inflammation was fully established. Similarly, in experimental arthritis, the timing of MSC administration, the relative effects of MSCs on different Th cell subsets, and the local joint conditions have been reported to critically determine the balance between therapeutic efficacy, lack of benefit, and detrimental effects [40,42,44,53]. These studies should in no way dampen enthusiasm for further preclinical and clinical applications of MSCs in disease conditions in which one or more T-cell effector pathways are known to be the primary cause for acute or chronic tissue damage. Rather, they highlight the complexity of the interactions that occur between stromal cells and cells of the immune system and the wealth of basic and therapeutic insights that can be gained from continued investigation of these interactions.
mesenchymal stem cell therapies. *Hum Gene Ther* 2010, 21:1641-1655.

11. Caplan AI: Why are MSCs therapeutic? New data: new insight. *J Pathol* 2009, 217:318-324.

12. Barry FP, Murphy JM, English K, Mahon BP: Immunogenicity of adult mesenchymal stem cells: lessons from the fetal allograft. *Stem Cells Dev* 2005, 14:252-260.

13. Bartholomew A, Sturgeon C, Siatkars M, Ferrer K, McIntosh K, Pati S, Hardy W, Devine S, Ucker D, Deans R, Moseley A, Hoffman R: Mesenchymal stem cells suppress lymphocyte proliferation in vitro and prolong skin graft survival in vivo. *Exp Hematol* 2003, 31:42-48.

14. Geisler F, Bohlinger J, Bussolari R, Dominici M, Hardergreyter R, Muller I: Human multipotent mesenchymal stromal cells use galectin-1 to inhibit immune effector cells. *Blood* 2010, 116:3770-3779.

15. Cutler AJ, Limbani V, Girdleston E, Navarrete CV: Umbilical cord-derived mesenchymal stromal cells modulate monocyte function to suppress T cell proliferation. *J Immunol* 2010, 185:6617-6623.

16. Wang Q, Sun B, Wang D, Ji Y, Kong Q, Wang G, Wang J, Zhao W, Jin L, Li H: Murine bone marrow mesenchymal stem cells cause mature dendritic cells to promote T-cell tolerance. *Stem Cells* 2008, 26:607-615.

17. English K, Ryan JM, Tobin L, Murphy MJ, Barry FP, Mahon BP: Cell contact, prostaglandin E2 and transforming growth factor beta1 play non-redundant roles in human mesenchymal stem cell induction of CD4+CD25(High)forkhead box P3+ regulatory T cells. *Clin Exp Immunol* 2009, 159:149-160.

18. Batten P, Samrudchandha B, Antonow JW, Tay SS, Lovell M, Taylor PM, Yacobov MH: Human mesenchymal stem cells induce T cell anergy and downregulate T cell allo-reponses via the Th2 pathway: relevance to tissue engineering human heart valves. *Tissue Eng* 2011, 17:2263-2273.

19. Rasmusson I, Uhmilin M, Le Blanc K, Levitsky V: Mesenchymal stem cells fail to trigger effector functions of cytotoxic T lymphocytes. *J Leukoc Biol* 2007, 82:687-693.

20. Najar M, Raicevic G, Boufker HI, Fayyad Kazan H, De Bruyn C, Meuleman N, Constandse M, Deboosere P, Meuleman N: Umbilical cord-derived mesenchymal stem cells treated GVHD and promotes vascularized skin allograft survival in rats. *Stem Cells Dev* 2008, 17:1321-1328.

21. Atkinson J, Le Blanc K, Bell D, Flucke M, Cooley J, Diabetes 2008, 32:1391-1399.

22. Coffman RL: The origin of TH2 responses. *Science* 2010, 328:116-1117.

23. Zhou H, Guo M, Bhan C, Sun Z, Yang Z, Zeng Y, A H, Zhao R: Efficacy of bone marrow-derived mesenchymal stem cells in the treatment of sclerodermatous chronic graft-versus-host disease: clinical report. *Biol Blood Marrow Transplant* 2010, 16:403-412.

24. Fiorina P, Jurewicz M, Angello A, Vergani A, Dama S, Adala S, Selig M, Godwin J, Law K, Pasclaci C, Smith RN, Capella C, Rodig S, Adra CN, Atkinson M, Sayegh MH, Abdi R: Immunomodulatory function of bone marrow-derived mesenchymal stem cells in experimental autoimmune type 1 diabetes. *J Immunol* 2009, 183:993-1004.

25. Awasthi A, Kuchroo VK: TH17 cells: from precursors to players in inflammation and infection. *Immunol Rev* 2009, 218:499-498.

26. Ghanem S, Pene J, Torcy-Moquet G, Jorgensen C, Yssel H: Mesenchymal stem cells inhibit human TH17 cell differentiation and function and induce TH1 regulatory cell phenotype. *J Immunol* 2010, 185:302-312.

27. Zappia E, Casazza S, Pedemonte E, Benvenuto F, Bonanni L, Gori S, Ceravolo A, Cazzanti F, Frassoni F, Mancardi G, Uccelli A: Mesenchymal stem cells ameliorate experimental autoimmune encephalomyelitis inducing T-cell anergy. *Blood* 2005, 106:755-761.

28. Rafeh M, Campeau P, Aguilar-Mahecha A, Buchman M, Williams P, Birman E, Yuan S, Young YK, Bovlin MN, Forner K, Baski M, Galipeau J: Mesenchymal stem cells ameliorate experimental autoimmune encephalomyelitis by inhibiting CD4+TH17 T cells in a CC chemokine ligand 2-dependent manner. *J Immunol* 2009, 182:5994-6002.

29. Carrión F, Nova E, Luz P, Aparabala F, Figueras F: Opposing effect of mesenchymal stem cells on TH1 and TH17 cell polarization according to the state of CD4(+) T cell activation. *Immunol Lett* 2011, 135:10-16.

30. Darlington P, Bovlin MN, Renouco C, Francois M, Galipeau J, Freedman MS, Atkins HL, Cohen JA, Solchaga L, Bar-Or A: Reciprocal TH1 and TH17 regulation by mesenchymal stem cells: implication for multiple sclerosis. *Ann Neurol* 2010, 68:540-545.

31. Bouffi C, Bong C, Courties G, Jorgensen C, Noël D: IL-6-dependent PGE2 secretion by mesenchymal stem cells inhibits local inflammation in experimental arthritis. *PLoS ONE* 2010, 5:e14247.

32. Kong O, Liao Z, Shao C, Shi Y, Pham H, Hoffman R: IL-6-dependent PGE2 expression by allogeneic bone marrow stromal cells ameliorates diabetes in NOD mice through modulating the distribution of CD4+ T cell subsets. *Cell Immunol* 2008, 253:23-30.

33. Lim JH, Kim JS, Yoon WH, Shin JS, Nam MH, Yang SH, Kim SJ, Park CG: 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, 185:4022-4029.

34. Geisler F: Azzollini N, Cassis P, Simr S, Morigi M, Cugini D, Cavinato RA, Todeschini M, Solini S, Sonzogni A, Perico N, Remuzzi G, Notni M: Pretransplant infusion of mesenchymal stem cells prolongs the survival of a semiallogeneic heart transplant through the generation of regulatory T cells. *J Immunol* 2008, 181:3933-3946.

35. Anknur J, Park J, Karp JM: Mesenchymal stem cell therapy: two steps forward, one step back. *Trends Mol Med* 2010, 16:203-209.

36. Hermann-Kleiter N, Baier G: NFT pull the strings during CD4+ T helper cell effector functions. *Blood* 2010, 115:2989-2997.

37. Aksu AE, Honber E, Sacks J, Ikeguchi R, Breitinger J, Szocho M, Unadkat J, Feili-Hariri M: Co-infusion of donor bone marrow with host mesenchymal stem cells treats GVHD and promotes vascularized skin allograft survival in rats. *Clin Immunol* 2008, 127:348-358.

38. Boumaz A, Srinivasan S, Witt WT, Feghali-Bostwick C, Dai Y, Garcia-Ocana A, Feili-Hariri M: Autologous bone marrow-derived rat mesenchymal stem cells promote PD-1+T-cell immunity in the islets, after T cell cytokine pattern and preserve regulatory T cells in the periphery and kidney. *J Autoimmun* 2009, 32:333-42.

39. Madec AM, Mallone R, Aouno G, Abou Mard E, Messier A, Eljaafari A, Thivolet C: Mesenchymal stem cells protect NOD mice from diabetes by inducing regulatory T cells. *Diabetologia* 2009, 52:1391-1399.

40. Coffman RL: The origin of TH2 responses. *Science* 2010, 328:116-1117.

41. Zhou H, Guo M, Bhan C, Sun Z, Yang Z, Zeng Y, A H, Zhao R: Efficacy of bone marrow-derived mesenchymal stem cells in the treatment of scleroderma-type chronic graft-versus-host disease: clinical report. *Biol Blood Marrow Transplant* 2010, 16:403-412.

42. Fiorina P, Jurewicz M, Angello A, Vergani A, Dama S, Adala S, Selig M, Godwin J, Law K, Pasclaci C, Smith RN, Capella C, Rodig S, Adra CN, Atkinson M, Sayegh MH, Abdi R: Immunomodulatory function of bone marrow-derived mesenchymal stem cells in experimental autoimmune type 1 diabetes. *J Immunol* 2009, 183:993-1004.
inhibit the formation of cytotoxic T lymphocytes, but not activated cytotoxic T lymphocytes or natural killer cells. Transplantation 2003, 76:1208-1213.

51. Prevosto C, Zancolli M, Canevali P, Zocchi MR, Poggi A. Generation of CD4+ or CD8+ regulatory T cells upon mesenchymal stem cell-lymphocyte interaction. Haematologica 2007, 92:881-888.

52. Patel SA, Meyer JR, Greco SJ, Corcoran KE, Bryan M, Rameshwar P. Mesenchymal stem cells protect breast cancer cells through regulatory T cells: role of mesenchymal stem cell-derived TGF-beta. J Immunol 2010, 184:5885-5894.

53. Tso GH, Law HK, Tu W, Chan GC, Lau YL. Phagocytosis of apoptotic cells modulates mesenchymal stem cells osteogenic differentiation to enhance IL-17 and RANKL expression on CD4+ T cells. Stem Cells 2010, 28:939-954.