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Immune regulation by non-lymphoid cells in transplantation

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Summary

Regulatory cells play a crucial role in the induction and maintenance of tolerance by controlling T cell as well as B and natural killer (NK) cell-mediated immunity. In transplantation, $CD4^+CD25^+$forkhead box $P3^+$ T regulatory cells are instrumental in the maintenance of immunological tolerance, as are several other T cell subsets such as NK T cells, double negative $CD3^+$ T cells, γδ T cells, interleukin-10-producing regulatory type 1 cells, transforming growth factor-β-producing T helper type 3 cells and $CD8^+CD28^-$ cells. However, not only T cells have immunosuppressive properties, as it is becoming increasingly clear that both T and non-T regulatory cells co-operate and form a network of cellular interactions controlling immune responses. Non-T regulatory cells include tolerogenic dendritic cells, plasmacytoid dendritic cells, mesenchymal stem cells, different types of stem cells, various types of alternatively activated macrophages and myeloid-derived suppressor cells. Here, we review the mechanism of action of these non-lymphoid regulatory cells as they relate to the induction or maintenance of tolerance in organ transplantation.

Keywords: alloreactivity, regulatory cells, transplantation

Myeloid dendritic cells (DCs) (tolerogenic DC, regulatory DC) (Fig. 1a)

Thymic DCs mediate the negative selection of T lymphocytes *in vivo* and play a role in the peripheral tolerance of self-reactive T lymphocytes [1]. Upon maturation, DCs increase their expression of major histocompatibility complex (MHC), adhesion and co-stimulatory molecules and secrete cytokines necessary to enhance T lymphocyte activation and to generate immune responses [2]. Therefore, this raises the question of whether preventing DC maturation would prevent T lymphocyte activation and promote tolerance in transplantation. This does indeed seem to be the case, as the injection of immature DCs has been shown to prolong cardiac allograft survival in mice [3] as well as rats [4,5]. Several mechanisms may be responsible for the peripheral tolerance induced by immature DCs. Some of these include T lymphocyte anergy in the absence of co-stimulatory signals, peripheral deletion of reactive T cells through Fas/Fas ligand (FasL) interactions and the capture of apoptotic cells by DCs [6], which then present antigens in a context where proinflammatory cytokine production is inhibited [7]. Other mechanisms include T cell starvation through tryptophan metabolism via indoleamine 2,3-dioxygenase (IDO) [8], T cell inhibition through the action of haem oxygenase-1 (HO-1) [9], induction of regulatory T cells (T_{reg}) (reviewed in [8]) or release of nitric oxide (NO) [5]. Certain cytokines, such as interleukin (IL)-10 [10], transforming growth factor (TGF)-β, hepatocyte growth factor (HGF) and granulocyte colony-stimulating factor, can induce immature DCs to become tolerogenic. Immature DCs induce the differentiation of T regulatory type 1 or T helper type 3 (Th3) regulatory cells following IL-10 or TGF-β production [11]. Paradoxically, mature DCs cannot induce differentiation of T_{reg} but are nevertheless required to promote their survival and function [12]. Several types of membrane molecules have been identified as markers of tolerogenic DCs, such as signalling lymphocyte activation molecule (SLAM), programmed death ligand 1 (PD-L1), DC receptor for endocytosis (DEC)-205 (CD205) and the inhibitory receptors of the immunoglobulin (Ig)-like transcript family (ILT3/4). Immature DCs exposed to $CD8^+CD28^-$ T_{reg} increase their expression of the immunoreceptor tyrosine-based inhibitory motif-containing ILT3 and ILT4 receptors, which prevents the overexpression of co-stimulatory molecules and thereby prevents the activation of $CD4^+$ T cells [13] and promotes the differentiation of T_{reg} [14]. DEC-205 appears to be a good marker for tolerogenic DCs without, however, transmitting

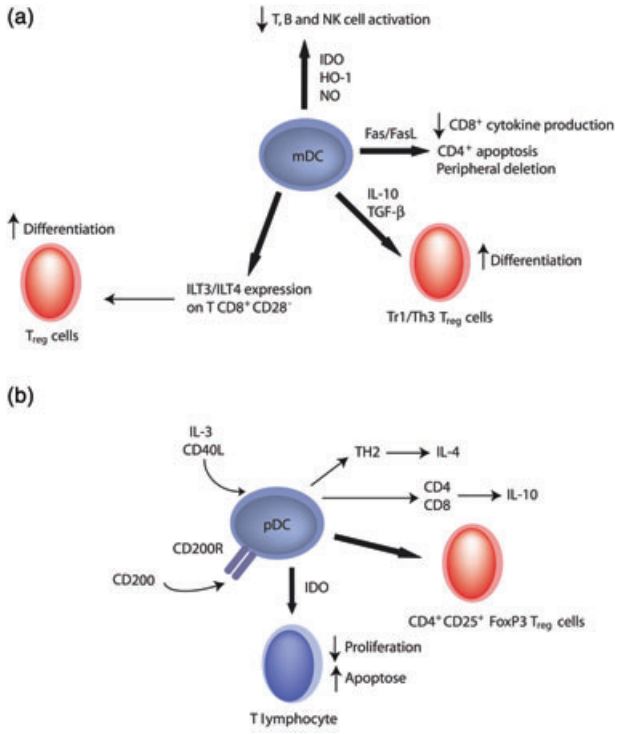

Fig. 1. Suppressive function of myeloid dendritic cells (mDC) and plasmacytoid dendritic cells (pDC). (a) mDCs induce peripheral tolerance via different mechanisms: peripheral deletion, $CD4^+$ T cell apoptosis or decrease of $CD8^+$ T cell cytokine production through Fas/Fas ligand (FasL) interaction. The synthesis of nitric oxide (NO), haem oxygenase (HO)-1 or indoleamine 2,3-dioxygenase (IDO) inhibits T, B and natural killer (NK) cell activation. After contact with $CD8^+CD28^-$ cells or through the synthesis of interleukin (IL)-10 and transforming growth factor (TGF)-β, mDC also induce regulatory T cell (T_{reg}) differentiation. (b) In the presence of IL-3 and CD40L, pDC activate IL-4 secretion by T helper type 2 (Th2) cells and IL-10 production by CD4 and CD8 T cells. They induce T_{reg} generation. Stimulation of pDC with CD200-immunoglobulin (Ig) induces IDO.

any signal to T cells [15]. $CD8^+$ DEC-205^+ DCs in mice [15] block $CD4^+$ T cell responses totally by inducing T cell apoptosis through Fas/FasL interactions [16]. They also block cytokine production by $CD8^+$ T cells without inducing their apoptosis [17]. SLAM and PD-L1, although expressed on activated DCs, generate two negative signals that cause immunosuppression. SLAM–SLAM homotypic interactions inhibit the production of IL-6, tumour necrosis factor (TNF)-α and IL-12 by CD40-stimulated DC [18], whereas DC expression of PD-L1, which shares the same B7-1 receptor as cytotoxic T lymphocyte antigen (CTLA)-4, suppresses the function of activated T cells [19]. In transplantation, interactions of B7 molecules with CTLA-4, the latter being expressed by activated T cells and T_{reg} cells, plays a critical role in peripheral tolerance by inhibiting T cell activation [20]. This effect has been attributed to signalling via signal transducer and activator of transcription 1 (STAT1), p38 mitogen-activated protein kinase (MAPK) and nuclear factor-κB, leading to the synthesis of IFN-γ which, in turn, in the absence of IL-6, leads to the induction of IDO [21,22]. IDO is responsible for down-regulating T [23–25], B and natural killer (NK) cell [26] activation and proliferation through depletion of tryptophan from the microenvironment [21] and through the immunosuppressive action of tryptophan catabolites (Kynurenin, quinolinic acid and 3-hydroxyanthranilic acid) [27]. DCs themselves escape from being destroyed via the action of IDO by a concomitant overexpression of tryptophanyl (Trp)–tRNA synthetase. The Trp–tRNA complex provides a reservoir of the amino acid in a form that is protected from IDO-mediated degradation and is directly available for protein synthesis [28].

Plasmacytoid DCs (Fig. 1b)

Plasmacytoid DCs (pDCs) are found mainly in the peripheral blood and bone marrow, and in the T cell areas of secondary lymphoid organs. pDCs are a specialized cell population that exhibit a plasma-cell like morphology and produce large amounts of type I IFN in response to viruses. These cells play a crucial role in the induction and maintenance of tolerance, properties that depend upon expression of the type I IFN receptor [8]. pDCs are considered as an immature DC subtype able to differentiate *in vitro* into mature DCs, in response to different stimuli [29]. In mice, pDCs express B220, Ly6C or CD11c but no CD123 (IL-3Rα) [30,31]. In humans, pDCs express CD4, CD123, HLA-DR, CD68, ILT-3 and CD45RA but lack CD11c, ILT-1, CD3, CD14, CD16, CD19, CD20 and CD56 [32,33]. Blood DC antigen (BDCA)-2 (CD303) and BDCA-4 (identical to neuropilin-1) are also human pDC-specific markers in the blood [34]. BDCA-2 is a type II lectin C [34] that plays a role in antigen internalization and presentation as well as in the synthesis of IL-12. In the presence of IL-3, CD40L or viruses, pDC can differentiate into mature DCs. This process involves up-regulation of MHC class II, co-stimulatory molecules and the chemokine receptor CCR7, as well as the production of IL-12, and leads in turn to an enhancement of T lymphocyte activation [29,30,35]. Activated pDCs are able to produce a large variety of proimmune cytokines including IFN-α/β, granulocyte–macrophage colony-stimulating factor, TNF-α, IL-6 and IL-8 [36]. In addition, human as well as murine mature pDCs affect T cells functions, leading to earlier activation, prolonged survival, IFN-γ production and Th1 differentiation, via different molecular mechanisms [37]. In contrast, in humans and mice, freshly isolated immature pDCs express low levels of MHC class II and co-stimulatory molecules and are consequently very limited in their capacity to present antigen to T lymphocytes and to induce cytokine polarization [29,38]. *In vivo*, the tolerogenic potential of pDCs was demonstrated initially following the administration of murine liver pDCs in a cardiac allograft model [39]. The latter cells were shown to acquire alloantigens in the allograft and then home, via the blood, to

peripheral lymph nodes where they induced the generation of $CD4^+CD25^+$forkhead box P3 (FoxP3)$^+$ T_{reg} expressing chemokine (C-C motif) receptor 4 (CCR4) [40]. Stimulation of pDCs with CD200-Ig [41], CTLA-4-Ig or glucocorticoid-induced TNF receptor family-related gene-Ig-induced IDO production and contributed to the tolerogenic state of these cells [42]. Furthermore, pDCs from tumour-draining lymph nodes express IDO constitutively, suggesting that they help to maintain the state of immunosuppression within the tumour [43].

$CD19^+$ DCs also have a plasma cell-like morphology, but unlike pDC they are found in the red pulp of the mouse spleen. These cells are normally capable of stimulating T cells. However, they can synthesize large amounts of IDO after ligation of CD80/86 by CTLA-4 bound on the surface of T_{reg} [44] or by ligation of Toll-like receptor 9 (TLR-9) [45], and thereby become immunosuppressive. As for pDC, the suppressive action of $CD19^+$ DCs is under the control of type I IFN.

Mesenchymal stem cells (Fig. 2)

Mesenchymal stem cells (MSC) are multi-potent non-haematopoietic cells located in the bone marrow, and to some extent in fat tissue [46], placenta, amniotic fluid [47] and umbilical cord blood [48]. MSC can differentiate into multiple mesenchymal lineages such as bone, fat and cartilage [49], endothelial cells, neural cells or endodermic cells [50,51]. MSC are characterized by the expression of single-strand conformation analysis (SSCA)-1 [52] and SSCA-4 [53] in mice, or ganglioside GD-2 [54] in humans, but lack expression of CD11b, CD14, CD31 or CD45 haematopoietic and endothelial markers [49]. MSC possess immunomodulatory properties thought to play a role in the maintenance of peripheral tolerance, in the control of autoimmunity [55] and in fetal–maternal tolerance [56]. *In vitro*, MSC are able to suppress T lymphocyte activation and proliferation induced by mitogens and polyclonal activators as well as by cognate antigens [57]. MSC induce a cell cycle arrest at the G0/G1 phase, rendering T cells anergic [58]. TGF-β and HGF [57] are involved in the suppression of T cell proliferation by MSC. Prostaglandin E_2 (PGE_2) [59] or NO [60], both secreted constitutively by MSC, may also play a role in their suppressive activity. So far it is not entirely clear whether IDO is [61] or is not [59] involved in the immune inhibition by MSC. Recently, a pivotal role was reported for HO-1 in the immunosuppressive properties of rat and human MCS [62]. *In vivo*, MSC can also modulate immune responses by inducing $CD8^+$ T_{reg} cell generation [63] and $CD4^+CD25^+$ T_{reg} amplification [59]. Also, MSC modulate DC differentiation and maintain them in an immature state (inhibition of co-stimulatory molecule and MHC class II overexpression). This modulation takes place through the action of macrophage colony-stimulating factor [64] and IL-10 [65]. In addition to T cell suppression, the immunosuppressive action of MSC extends to B cells: their proliferation, differentiation and antibody production is prevented without modification of co-stimulatory molecule expression or cytokine production through the synthesis of PGE_2 [66]. NK cells also stop proliferating and secreting cytokines upon contact with MSC or in the presence of TGF-β or PGE_2 secreted by MSC [67]. *In vivo*, MSC delayed the T cell-mediated rejection of skin allografts in primates [68] and decreased rejection of allogeneic BM transplantation in mice [69]. MSC were also found to be protective in a rat model of kidney ischaemia/reperfusion injury [70] as well as in a model of heart transplantation [71].

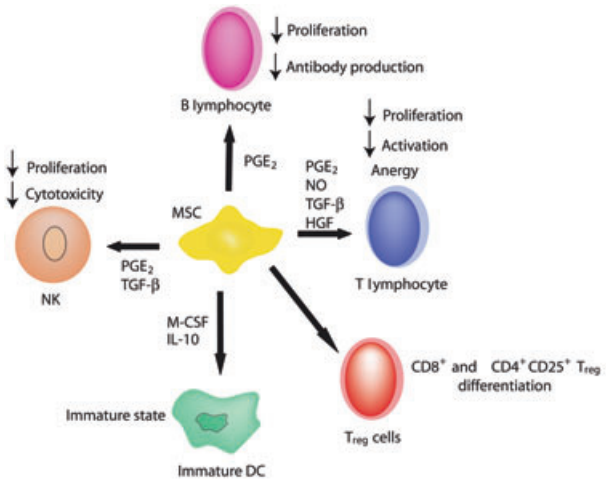

Fig. 2. Immunomodulatory properties of mesenchymal stem cells (MSC). MSC induce regulatory T cell differentiation and amplification. Prostaglandin E_2 (PGE_2), nitric oxide (NO), transforming growth factor (TGF)-β and hepatocyte growth factor produced by MSC are involved in the suppression of T cell proliferation and activation and induce anergy. PGE_2 and TGF-β block natural killer (NK) cell proliferation and cytotoxicity, whereas PGE_2 suppresses B cell proliferation and differentiation into plasmocytes. Secretion of macrophage colony-stimulating factor and interleukin (IL)-10 by MSC prevents dendritic cell maturation.

Other stem cells

The immune-privileged properties of embryonic stem cells (ESC) [72] also makes these cells candidates as a source of regulatory cells for cell therapy. ESC express low levels of MHC class I and class II molecules [73], albeit sufficient in quantity to elicit their rejection by cytotoxic T cells [74]. The reason for their immunomodulatory properties resides in their synthesis of HO-1, which reduces proliferative responses *in vitro* [75]. Neural stem cell (NSC) transplantation or transplantation of ESC-derived neural precursors has also been proposed as a means of cell replacement therapy. It was shown initially that intraventricular transplantation

of NSC attenuated brain inflammation in acute and chronic experimental autoimmune encephalomyelitis (EAE) and reduced demyelination and axonal pathology [76,77]. However, their intravenous injection also inhibited EAE and reduced central nervous system (CNS) inflammation and tissue injury, although they did not enter the CNS [78]. In these experiments, NSC were found to have migrated to lymph nodes and spleen where they inhibited the activation and proliferation of T cells and reduced markedly their encephalitogenicity. Although the mechanism of action involved has not yet been elucidated, it seems that NSC inhibit the activation of T cells without inducing their apoptosis [79].

Alternatively activated macrophages (Fig. 3a)

Upon activation, macrophages present a heterogeneous phenotype that divides them into two categories: 'classically' activated macrophages (CAMΦ or M1) and 'alternatively' activated macrophages (AAMΦ or M2) [80]. Such distinct differentiation depends on the presence of specific cytokines in the microenvironment. Inflammatory responses are induced by CAMΦ under the influence of microbial agent or type 1 cytokines such as IFN-γ or IL-12. This classical activation is associated with a large production of NO and proinflammatory cytokines such as IL-6 or TNF-α. In contrast, an anti-inflammatory environment is generated by the action of AAMΦ induced by anti-inflammatory agents such as IL-4, IL-10, IL-13, TGF-β, granulocyte–macrophage colony-stimulating factor or glucocorticoids [80,81]. Following administration of IL-4 or IL-13, murine AAMΦ overexpress mannose receptors (CD206) [82] as well as scavenger receptors (CD163) [83] and MHC class II molecules [84] that stimulate endocytosis and antigenic presentation. IL-4 and IL-13 also induce the expression of the co-stimulatory molecule PD-L2 [85], the surface markers CD23 [86], CD163 [87] and CD14 [88]), the chemokines CCL2 (MCP1) [89], CCL22 (MDC) [90], CCL17 (TARC) [91] and CCL18 (AMAC1) [92], the cytokines IL-1 [93], IL-10 [94] and TGF-β [95] and intracellular enzymes such as arginase I [81]. The expression and secretion of galectin-3 is also a major characteristic of AAMΦ [96]. Functionally, AAMΦ are immunosuppressive and inhibit the proliferation of activated $CD4^+$ T cells and the secretion of IL-2, IFN-γ and IL-4 in allogeneic mixed leucocyte reaction [97]. The neutralization of IL-10 with antibodies, or the blockade of NO release, fails to reverse the inhibition of AAMΦ, indicating that these factors are not essential for their suppressive activity. The suppression is also independent of co-stimulatory molecules (in contrast with CAMΦ) [98]. However, AAMΦ can be induced secondarily after B7/CD28 T cell co-stimulation blockade, suggesting that they might play a role in tolerance induction in transplantation [99]. Because they are immunosuppressive, AAMΦ are involved in a large variety of pathologies including allergy as well as cellular and humoral responses against parasites and extracellular pathogens. They are found in the placenta and in the lung, where they protect against unwanted immune reactivity and down-regulate inflammatory reactions [100,101]. Recently, a novel function was reported for $CD4^+CD25^+FoxP3^+$ T_{reg} cells in the induction of AAMϕ; T_{reg} cells produced high levels of IL-10, IL-4 or IL-13, which in turn enhanced AAMΦ differentiation [102]. Tumour-associated macrophages are a type of AAMΦ that are the most abundant immunosuppressive cells within the tumour microenvironment. They exhibit the $IL\text{-}10^{high}IL\text{-}12^{low}$ AAMΦ profile and proliferate in response to the cytokines leukemia inhibitory factor and IL-6 present in the tumour microenvironment [103].

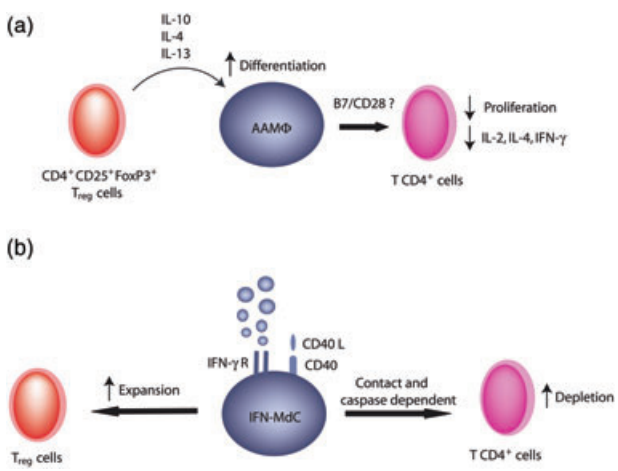

Fig. 3. Suppressive function of alternatively activated macrophages (AAMΦ) and interferon-γ-stimulated monocyte-derived cells (IFN-γ-MdC). (a) Cytokines secreted by regulatory cells induce AAMΦ differentiation. Interaction with $CD4^+$ T cells induces suppression of proliferation and inhibition of cytokine production. (b) IFN-γ-MdC induce depletion of activated $CD4^+$ T cells through contact and caspase-dependent mechanisms. On the other hand, IFN-γ-MdC induce regulatory T cell expansion.

The IFN-γ-stimulated monocyte-derived cells (IFN-γ-MdC; Fig. 3b)

The IFN-γ-MdC are a macrophage subset arising when the latter are cultured in the presence of CD40L-expressing $CD4^+$ T cells, macrophage colony-stimulating factor and IFN-γ. IFN-γ-MdC express F4/80, CD11b/c, CD86 and CD274, but lack CD4, CD8, Gr1, CD19, CD80 and CD207. Functionally, IFN-γ-MdC induce a cell contact and caspase-dependent depletion of activated T cells and an expansion of $CD4^+CD25^+FoxP3^+$ regulatory cells. Although their mechanism of action has not yet been identified, the expression of IFN-γ receptor and CD40 has been shown to be necessary for their functional activity. Contrary to expectation, IDO is not thought to be involved in the immunosuppressive activities of IFN-γ-MdC [104]. In mice, IFN-γ-MdC delivered intravenously can migrate to gut-associated peripheral lymphoid

tissues and promote the clinical and histological resolution of chronic colitis and dampen acute rejection of allogenic heart transplants [104].

Myeloid-derived suppressor cells (Fig. 4)

In the 1980s, cells named natural suppressor cells, distinct from T and NK cells, were described in mice bearing transplantable tumours [105]. These cells, originating from the bone marrow, derive from a heterogeneous mixture of myeloid cells at different stages of differentiation. They were defined initially as immature myeloid cells or myeloid suppressor cells (MSC) because of their capacity to suppress immune responses [106]. To minimize confusion with MSC, Gabrilovitch proposed to name these cells 'myeloid-derived suppressor cells' (MDSC) [107]. These cells accumulate in mice in various lymphatic organs [108] in pathological conditions, including bacterial infections [109], chronic inflammation, tumour progression, graft-*versus*-host disease [110] or immune stress following activation by superantigens [111]. They are characterized by the co-expression of Gr-1 (Ly-6G) and CD11b [106] together with the immature cell marker CD31 [112]. Other markers that could be correlated with the suppressive function of these cells include CD80 [113], F4/80, CD115 (macrophage colony-stimulating factor receptor) [114] and CD16 [115]. MDSC also express major histocompatibility class I but not class II molecules [116]. In humans, MDSC are defined by the expression of immature markers such as $CD34^+$, $CD33^+$, $CD15^+$, $CD14^-$ and $CD13^+$ and are increased in the peripheral blood of cancer patients [117]. MDSC are able to suppress T and B cell proliferation and cytokine production by blocking entry into the cell cycle, without killing target cells [118], in a contact-dependent manner [116]. They are also able to suppress NK cell activity [119]. Several studies showed that the interaction itself between MDSC and T cells, as well as IFN-γ secretion by T lymphocytes, are instrumental in the induction of MDSC suppressive activity [118]. Indeed, in tumour-bearing mice, both release of IFN-γ by T lymphocytes and stimulation through CD40 [118] induce MDSC to produce IFN-γ and IL-13, which are utilized in an autocrine manner to enhance their suppressive properties [120]. To control T lymphocyte responses, activated MDSC enhance the production and activity of two enzymes involved in the metabolism of L-arginine, a non-essential amino acid that plays a role in immune responses and regulation of T lymphocyte function [121]. These enzymes are inducible NO synthase (iNOS), which induces NO production [122], and arginase-1 (ARG-1), which depletes arginine from the microenvironment [123]. ARG-1 converts L-arginine into urea and L-ornithine whereas iNOS oxidizes L-arginine into L-citrulline and NO [124]. The activation of ARG-1 and iNOS suppresses T cell proliferation by interfering with intracellular signals involved in the transduction pathways leading to T cell apoptosis [125]. Indeed, reversal of the inhibitory effect on T cell proliferation was achieved following addition of N(G)-monomethyl-L-arginine, an inhibitor of iNOS [122]. Although low concentrations of NO act in synergy with the T cell receptor (TCR) to stabilize p53 and allow for IL-2 synthesis by effector T cells [126], higher doses inhibit phosphorylation and activation of signalling molecules such as Janus kinases, STAT5, extracellular-regulated kinase and protein kinase B (Akt) [118], and therefore block the IL-2 and MAPK signalling pathways. The use of phosphodiesterase-5 inhibitors, which down-regulate ARG 1 and iNOS expression, was shown to reduce tumour recruited-MDSC expansion and indirectly enhance T cell proliferation [127]. Inhibition of T cell proliferation by MDSC is also characterized by the loss of the TCR ζ chain [108] in a L-arginine-dependent manner [128].

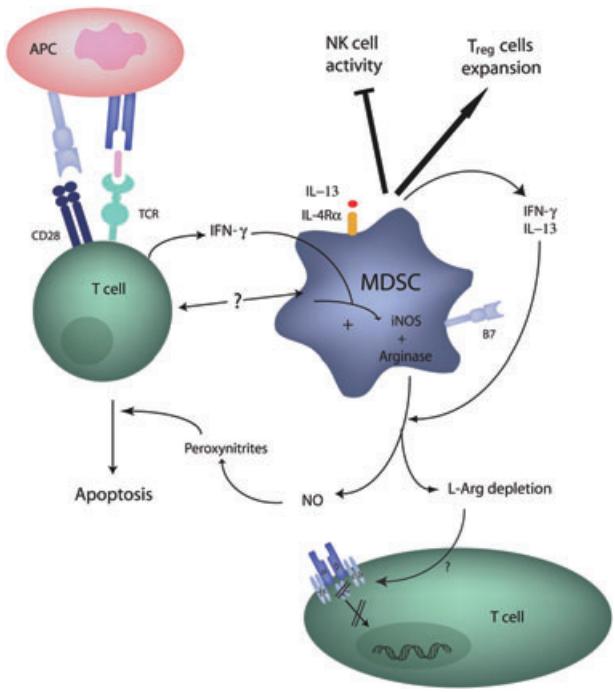

Fig. 4. Immunoregulatory properties of myeloid-derived suppressor cells (MDSC). The release of interferon (IFN)-γ by T lymphocytes and the stimulation through CD40 trigger IFN-γ and interleukin (IL)-13 production by MDSC, which leads secondarily to the production of both inducible nitric oxide synthase (iNOS) and arginase I. These enzymes are able to suppress T cell receptor signalling and induce T cell apoptosis. In addition, MDSC are also able to enhance regulatory T cell expansion and suppress natural killer (NK) cell activity.

In addition to their direct activity on T cells, MDSC are able to down-regulate T cell proliferation indirectly by enhancing the development of $CD4^+CD25^+FoxP3^+$ T_{reg} [114]. Tumour growth in mice was shown to be delayed by the action of anti-CTLA-4 antibodies that block the CTLA-4/CD80 interactions occurring between T_{reg} and MDSC [129]. Also, the cross-talk between MDSC and macrophages of tumour-bearing mice results in a reduction in IL-12 release by macrophages and an increase in IL-10 production by

MDSC, reorientating the response towards a type 2 response that favours tumour progression [130].

In transplantation, a role for MDSC was described recently for the first time in a rat model of kidney allograft tolerance induced by anti-CD28 antibodies [131]. In this model, MDSC were found to accumulate in the graft and blood of recipient animals and the production of NO was found to be responsible for the maintenance of tolerance.

Clinical potential of regulatory 'non-T' cells

Regulatory 'non-T' cells basically present non-antigen-specific immunosuppressive properties. They usually do not induce dominant tolerance [132]. However, they co-operate with T_{reg} cells and reinforce an antigen-specific and dominant immunosuppression driven by these T_{reg} cells. The therapeutic potential of regulatory 'non-T' cells is best exemplified in animal models where transplant tolerance or the improvement of immunoinflammatory diseases could be obtained after adoptive transfer of MDC [4], pDC [39], NSC [78] or IFN-γ DC [104]. In man, the immunoregulatory potential of regulatory 'non-T' cells is currently being tested. Transplant acceptance-inducing cells (TAIC) are immunoregulatory macrophages with the capacity to specifically dampen allogeneic rejection [104]. In a safety clinical trial study, the pre-transplant infusion of these donor-derived regulatory cells in kidney allograft recipients did not provide conclusive evidence of a beneficial effect. However an alloantigen-specific unresponsiveness was observed which suggested that TAIC injection was not immunogenic but immunoregulatory, and could allow minimization of pharmacological immunosuppression. The transfer of autologous *ex vivo* expanded MSCs (NCT00752479 phase I/II study; available at: http://clinicaltrials.gov/) to kidney allograft recipients under a standard immunosuppressive regimen is also tested to suppress immune rejection further and improve donor kidney survival. In type I diabetes patients, autologous monocyte-derived DC are also used in a phase I clinical trial (NCT00445913). The cells are first treated *ex vivo* with antisense phosphorothioate-modified oligonucleotides targeting the primary transcripts of the CD40, CD80 and CD86 co-stimulatory molecules to produce iDC and infused to assess their diabetes-suppressive potential.

Concluding remarks

In addition to T_{reg}, a series of other cell types have been described that share the common feature of being able to modulate the reactivity of T and other immune cells. The most common mechanism of action of these cells is a direct induction of target T cell death or growth arrest, mediated by immunosuppressive enzymes, cytokines or cytotoxic mechanisms. In addition, a consistent finding has been the non-T cell-mediated induction of T_{reg} cells, which secondarily mediate the immune suppression. Inversely, T_{reg} cells can also trigger immunoregulation by non-T cells (as is the case for AAMΦ; Table 1). Therefore, immune regulation should not be seen as a consequence of the action of one regulatory cell type but rather of the synergistic interactions of T and regulatory 'non-T' cells.

Table 1. Non-lymphoid cells with immunosuppressive activity.

Cell type	Mechanism of action	Transplant or inflammation model
Myeloid DC	Fas/FasL [16], TGF-β, IL-10, induction of T cell anergy, induction of T_{reg} [6] IDO [25], HO-1 [9], NO [5]	Cardiac allograft survival in mice [3] and rats [4,5]
Plasmacytoid DC	Controls T cell polarization [38], IDO [133], induction of T_{reg} [40]	Cardiac allograft survival in mice [39]
MSC	PGE_2 [59], TGF-β[57], NO[60], IDO [61], HO-1 [62] Induction of T cell anergy [58], induction of T_{reg} [59].	Heart transplantation in mice [71]
ESC	Low MHC expression [73] Production of HO-1 [75]	Transplantation of ESC for tissue repair [134]
NSC	n.d.	Intracerebral injection of neuronal precursors in EAE [78]
AAMΦ	TGF-β [80] Arginase I [80]	Suppression of T cell proliferation [99]
IFN-γ–MdC	Induction of T_{reg}, CD40 and IFN-γ-dependent [104]	Heart transplantation in mice Mouse DSS-induced colitis [104]
MDSC	Release of nitric oxide [122] Action of arginase I [123]	Kidney transplantation in rats [131]

AAMΦ, alternatively activated macrophages; DC, dendritic cell; DSS, dextran sulphate sodium; EAE, experimental autoimmune encephalomyelitis; ESC, embryonic stem cells; IFN-γ–MdC, interferon (IFN)-γ-stimulated monocyte-derived cells; MDSC, myeloid-derived suppressor cells; MSC, mesenchymal stem cells; n.d., not defined; NSC, neural stem cells; IDO, indoleamine 2,3-dioxygenase; HO-1, haem oxygenase; PGE_2, prostaglandin E_2; IL, interleukin; IFN, interferon; TGF, transforming growth factor; T_{reg}, T regulatory cells.

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