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Immunotherapy following hematopoietic stem cell transplantation: potential for synergistic effects

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Abstract

Hematopoietic stem cell transplantation (HSCT) is a particularly important treatment for hematologic malignancies. Unfortunately, following allogeneic HSCT, graft-versus-host disease, immunosuppression and susceptibility to opportunistic infections remain among the most substantial problems restricting the efficacy and use of this procedure, particularly for cancer. Adoptive immunotherapy and/or manipulation of the graft offer ways to attack residual cancer as well as other transplant-related complications. Recent exciting discoveries have demonstrated that HSCT could be expanded to solid tissue cancers with profound effects on the effectiveness of adoptive immunotherapy. This review will provide a background regarding HSCT, discuss the complications that make it such a complex treatment procedure following up with current immunotherapeutic strategies and discuss emerging approaches in applying immunotherapy in HSCT for cancer.

Keywords

adoptive immunotherapy; antigen-presenting cell; graft-versus-host disease; graft-versus-leukemia; graft-versus-tumor; hematopoietic stem cell transplantation; myeloablative conditioning; natural killer cell; nonmyeloablative conditioning; regulatory T cell

Hematopoietic stem cell transplant (HSCT) is currently used for the treatment of a variety of life-threatening conditions, including neoplastic, autoimmune, inflammatory and genetic disorders. Early and late complications following allogeneic HSCT include graft-versus-host disease (GVHD), donor rejection, relapse of primary malignancy, conditioning-related toxicity, immunodeficiency and infections. These complications limit the efficacy of HSCT and its outcome depends on numerous variables, such as the type of disease, age of the patient, stage of disease progression and therapies used. Importantly, graft-versus-tumor (GVT) effects or recognition of cancer cells by donor T cells is the primary objective sought with this therapy. Unfortunately, GVHD and GVT are tightly associated and the pathways of one can lead to the other. This review will delineate the use of HSCT, its limitations and the pathophysiology of GVHD and GVT. We will then focus especially on current

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immunotherapeutic approaches (e.g., cellular therapies, molecular targeting, cytokine therapy and blockade of costimulatory pathways), their advantages and disadvantages and potential methods to consider in the application of HSCT.

Hematopoietic stem cell transplant

Uses

Hematopoietic stem cell transplant or bone marrow transplantation (BMT) was originally developed in the early 1950s as an outgrowth of the study of radiation exposure [1]. It was subsequently developed as a means to reconstitute the immune and hematopoietic systems and later evolved for cancer treatments involving high-dose cytoreductive therapies. Two primary sources of stem cells were used, namely autologous (from the patient) or allogeneic (from an alternative donor). Both have markedly different advantages and disadvantages that are contingent upon numerous variables, including the nature of the disease being treated. With regard to cancer treatments, it was found that allogeneic sources of hematopoietic stem cells (HSCs; either from related or unrelated donors) resulted in a significantly lower incidence of tumor relapse, although, as will be discussed later, this was not without additional complications. This ‘allogeneic effect’ of HSCT resulted in lower relapse rates in certain cancers and is among the earliest documented evidence of immunotherapy that successfully attacked the cancer. HSCT is now being used or explored for the treatment of a variety of life-threatening conditions, including hematologic, neoplastic, autoimmune, inflammatory and genetic disorders (Table 1), although its primary use is still for the treatment of hematologic malignancies.

Hematopoietic stem cells for transplantation can be derived from three different sources: bone marrow (BM), mobilized peripheral blood (peripheral blood stem cell transplant [PBSCT]) or umbilical cord blood (UCB). In the mobilization procedure, donors are treated with granulocyte colony-stimulating factor (G-CSF) [2], granulocyte-macrophage colony-stimulating factor (GM-CSF) or the combination of G-CSF and CXCR4 antagonist antibody (plerixafor) [3–5] in order to mobilize HSCs from the BM into the peripheral blood. The advantages of PBSCT are a higher yield of CD34⁺ cells, quicker engraftment and rapid hematopoietic recovery compared with BM cells (BMCs). These advantages hold true for both autologous and allogeneic transplantation [6–8]. However, increased incidence of chronic GVHD has been reported with allogeneic PBSCTs [6,7,9,10]. This method has proved to be beneficial for the treatment of patients with acute myeloid leukemia (AML) [11], as well as the treatment of other hematologic and lymphoid cancers [12,13].

Umbilical cord blood transplants (UCBTs) were initially applied to children but are now being attempted with adults. The number of stem cells that can be collected from UCB is limited and there appears to exist some qualitative differences in the stem cells and progenitors obtained. Therefore, engraftment of the transplant is a slower process compared with when the patient receives PBSCT, rendering the recipient more vulnerable to opportunistic infections owing to delayed immune reconstitution [14]. Since T-cell activity is lower in UCB, there are fewer requirements for HLA matching between donor and recipient. In addition, GVHD incidence and severity have been reported to be lower in UCBT compared with PBSCTs [15]. UCB grafts have demonstrated efficacy in hematopoietic reconstitution in children with acquired or congenital lymphohematopoietic disorders, neuroblastoma and metabolic disease [16]. In order to overcome the limitation of HSC numbers in UCB, cotransplantation of two unrelated UCB grafts has been attempted in adult patients. Recently, it has been demonstrated that double UCBT appears to have lower relapse rates than single-unit UCBT in spite of a higher incidence of acute GVHD [17]. Furthermore, double UCBT in acute leukemia patients resulted in superior survival rates and enhanced graft-versus-leukemia (GVL) effects [18]. Thus, double UCBT can be used as a

means to augment GVL, but more needs to be understood on the biology of UCB and reconstitution. While HSCT has evolved to utilize different HSC sources in combination with diverse conditioning regimens, it remains severely limited by adverse complications, such as GVHD, graft failure, toxicity and immunodeficiency of the patient, rendering them more susceptible to opportunistic infections.

Delayed immune recovery is particularly pronounced in elderly recipients. As most cancers affect individuals older than 55 years of age and HSCT is predominantly used for the treatment of cancer, more studies and insights are needed into the impact of age on the outcome of HSCT. Interest in the past years has been growing regarding haploidentical transplantation in adult patients relapsing from AML or acute lymphatic leukemia and in need of an immediate matched donor. Despite several encouraging reports demonstrating lower GVHD, in haploidentical transplantation, conditioning-related complications and leukemia relapse remain major problems with the efficacy of HSCT [19–22]. Treatment failure is mainly due to the advanced disease stage and the age at which the patient receives the transplant. Therefore, timing is an important factor in the outcome of HSCT.

Graft-versus-tumor & graft-versus-host disease: an intertwined relationship

The observations that allogeneic HSCT resulted in significantly lower relapse rates comprise perhaps the first definitive evidence that successful immune attack can occur in human cancer [23–27]. Unfortunately, this beneficial GVT activity and the complication of GVHD were tightly linked. GVHD is an immunological disorder resulting from donor lymphocytes recognizing the recipient as ‘foreign’ and mounting an immune response against host tissues in an immunoincompetent recipient. GVHD presents unique issues that impact upon the use of allogeneic HSCT in cancer. It was classically defined as occurring in acute and chronic forms initially based on time of onset; however, it has become increasingly clear that they represent fundamentally different pathobiologic entities. Acute GVHD manifests during the first 3 months following allogeneic stem cell transplantation and is associated with skin rash, nausea, diarrhea and severe liver, thymus and intestinal abnormalities. Chronic GVHD typically occurs more than 100 days after transplant and it targets glands that secrete mucous and saliva, as well as other exocrine glands [28]. This results in dysregulated production of inflammatory cytokines by T cells and other immune cells, which leads to inflammation and fibrosis, similar to autoimmune diseases [29–31]. Central to both acute and chronic GVHD are the need for donor T cells and transplantation antigen disparity between the donor and host (either MHC or minor histocompatibility antigens). The pathophysiology of acute GVHD is better understood than chronic GVHD, mainly due to the lack of experimental chronic GVHD models that include all of the features of the disease. The incidence and severity of GVHD varies based on numerous factors, including HLA compatibility, whether the donor and recipient are related, age of the recipient, disease status and type being treated, cytoreductive conditioning used, presence of donor T cells and the extent of prophylactic immunosuppression applied [32–39]. The incidence and mortality from GVHD can range from 10 to 80% depending on acute or chronic forms, risk factors and grading [40,41]. In addition, despite being different diseases, there is crossover between the etiology of acute and chronic GVHD and patients can develop both.

Graft-versus-tumor effects are reactions in which donor cytotoxic T lymphocytes (CTLs) and natural killer (NK) cells recognize the transformed cells in the recipient and target their killing. The first evidence of GVT in mice was observed in studies by Barnes and Loutit in which allogeneic BMT cured leukemia in mice, but the recipients still succumbed to GVHD [42]. These results demonstrated not only the presence of GVT effects in transplantation but also the intricate association between GVT and GVHD. One of the earliest reports of GVT in humans was found in chronic myeloid leukemia (CML) patients that relapsed following

allogeneic HSCT, but went into remission after donor lymphocyte infusion (DLI) [43]. Clinical studies have demonstrated that donor T cells are needed for GVT. T-cell depletion of donor graft, used as prophylaxis in GVHD in CML patients receiving allogeneic BMT [44,45], results in an increased risk of relapse and disease recurrence [46,47]. Although allogeneic HSCT is the only curative treatment for some hematological diseases, the potential mortality induced by complications is still a huge caveat. In addition, allogeneic HSCT was initially limited to patients under the age of 55 years owing to the extensive toxicities and complications related to the severe conditioning regimens. The advent of reduced intensity or nonmyeloablative conditioning has markedly expanded the use of HSCT to older patients [48–50]. This therapeutic procedure involves less cytoreductive conditioning and more immunosuppression of the recipient to allow for mixed donor engraftment chimerism and can be followed by delayed DLI to mediate the GVT effects. In allogeneic HSCT, there is less acute GVHD in sibling transplants. Research priorities should focus on the impact of age upon outcome in HSCT.

Pathophysiology of graft-versus-host disease

To understand the means through which to best exploit GVT, first one needs to understand GVHD, as the processes may indeed be linked. GVHD develops in four main stages: first, cytoreductive conditioning induces tissue damage; second, the induction phase in which the donor T cells become sensitized to the host cells; third, a cytokine storm in which amplification processes occur that fuel GVHD pathways; and fourth, the effector stage in which target tissues are attacked, including the tumor (Figure 1) [51–56].

Cytoreductive conditioning is applied to target the cancer in order to reduce tumor burden and to allow for donor engraftment via suppressing the recipient's immune system. Cytoreductive conditioning commonly entails radiation and/or chemotherapy, which results in tissue injury followed by the secretion of proinflammatory cytokines (i.e., TNF- α , IL-1 and IL-6), leading to activation of host antigen-presenting cells (APCs) as well as upregulation of MHC and costimulatory molecules (i.e., CD40/CD40L and B7/CD28). At this point, donor T cells recognize host APCs and interact with them via T-cell receptors (TCRs) and costimulatory molecules and their appropriate ligands. This APC/TCR interaction results in the induction phase in which donor T cells become activated, proliferate and secrete cytokines such as IL-2, TNF and IFN- γ . Interestingly, the role of any particular cytokine in GVHD seems critically dependent on the timing of their induction. Cytokines that can be deleterious in the early GVHD process have been found to be protective later on, and *vice versa* [57,58]. During the early stages of GVHD, T cells expand and induce further priming of peripheral mononuclear phagocytes and production of inflammatory cytokines such as TNF- α , IL-6 and IL-1. This 'cytokine storm' occurring after the induction phase helps direct and mobilize the various effector cells to GVHD target organs. At this level, numerous immune cell types, both adaptive and innate, become involved. Finally, during the effector stage, the production of IFN- γ , TNF- α , perforin, granzymes and upregulation of FasL and TNF-related apoptosis-inducing ligand leads to target cell death and further host tissue destruction, which begins a cycle that amplifies itself [41]. Unfortunately, these same pathways also contribute to the GVT effects. Most successful attempts to control GVHD revolve around the prevention of the induction phase since, once GVHD starts its vicious cycle, it is extremely difficult to manage.

Treatment/prevention of graft-versus-host disease: current status & development of new therapies

Therapeutic regimens

Allogeneic HSCT uses systemic immunosuppression, usually administered as prophylaxis to prevent GVHD. However, these same immunosuppressive regimens also impair the patient's immune competence, as well as GVT pathways. Numerous agents have been developed but certain types predominate. Systemic glucocorticosteroids are used in many regimens. Steroids such as methylprednisolone and prednisone are used, and suppress numerous immune pathways. Cyclosporine and tacrolimus are calcineurin inhibitors that affect T-cell proliferation. Mycophenolate mofetil and pentostatin are other immunosuppressant drugs used to suppress T-cell proliferation in the treatment of steroid-refractory GVHD [59–61]. Despite protection from GVHD, long-term immunosuppression is associated with toxicities and long-term side effects. However, attempts to taper off the immunosuppression run the risk of GVHD recurrence. Chronic GVHD presents particular problems owing to its delayed onset and the fact that it can often be refractory to steroids and other immunosuppressive agents that are effective in acute GVHD. This balance between suppressing donor T cells in GVHD and not impairing GVT represents one of the most significant issues in HSCT.

Critical role of T cells in graft-versus-host disease & graft-versus-tumor

T-cell depletion (TCD) of the BM graft has been found to be the most effective treatment for GVHD by limiting the number of host-reactive T cells. However, prevention of GVHD does not correlate with an improved survival rate. In fact, TCD in humans not only reduced GVHD but was also associated with higher rates of graft failure, infections, lymphoproliferative diseases and tumor relapse [44,62–66]. Attempts now focus on removing particular T-cell subsets in order to modulate the alloreactive responses.

Indirect methods of TCD use monoclonal antibodies (mAbs) alone or in combination in order to target surface antigens on lymphocytes and different T-cell subsets. T cells are usually depleted by positive selection of CD34⁺ HSCs [67]. Other studies have depleted T cells with Campath-1, which targets CD52 on mature T cells, B cells and monocytes [68,69]. Although TCD of the BM grafts may prove to be an efficient approach to prevent GVHD, clinical studies have demonstrated that TCD results in greater tumor relapse, graft failure and delayed immune reconstitution, thus making the patient more susceptible to opportunistic viral and bacterial infections as well as Epstein–Barr virus-associated B-cell lymphoproliferative disorders and cytomegalovirus [70]. Thus, the caveat that GVHD and GVT are tightly linked is perhaps no more dramatically demonstrated by the removal of the donor T cells from the allograft.

Murine preclinical models to study graft-versus-host disease/graft-versus-tumor: advantages & disadvantages

Preclinical assessment of potential therapeutics in cancer is of paramount importance as a screen to optimize therapy before clinical assessment. This is also true with HSCT. The predominant model used to delineate the immune mechanisms/pathways in GVHD is the mouse, partly owing to issues such as ease and expense, but also owing to reagent availability that allowed for a more complete characterization of the species, as well as the fact that murine models are inbred and allow for transplantable tumor studies to be performed [71,72]. Similarly to the clinical condition in humans, one needs to evaluate effects on GVHD therapeutics as well as GVT. Large animal models, such as the dog, are also used in HSCT studies, but to a much lesser extent [73–75]. The use of the mouse has

numerous cautionary notes that need to be taken into consideration before attempting to extrapolate to potential clinical relevance. Significant species differences exist between mouse and man [76]. In addition to immunological differences, there are also issues such as housing (mice are kept in specific pathogen free [SPF] conditions), age (young mice are often used in HSCT studies), sex (female mice are used most often), and significant strain differences, all of which can have a marked impact on outcome, as well as on clinical relevance. The issue of genetic drift in inbred strains from a particular colony [77,78] and the use of SPF mice has also led to an inability to reproduce data from earlier studies. Questions arise as to what is the best mouse model for GVHD or whether multiple models need to be used. Reproducibility among laboratories performing murine studies in GVHD is also affected by differences in conditioning regimens and donor cell sources (some types of additional T cells are usually administered, as the number of T cells in the BMC preparations are usually insufficient to induce acute GVHD). Murine GVHD studies must also have a GVT component, as has been demonstrated by clinical results showing the tight relationship between the two. With murine tumor studies, there are also issues regarding the use of certain transplantable tumor cell lines as a model for human cancer. These cell lines are often aggressive (which means interventions often must occur within days of implantation), extensively cultured *in vitro* and fail to mimic the slow progression and immune evasion pathways of human tumors. There are real concerns regarding the clinical relevance of studies using inbred mice that are 8–12 weeks of age (the lifespan of a mouse is approximately 2 years), housed under SPF conditions in which HSCT is used to treat a transplantable tumor cell line injected intravenously several days prior. However, the use of outbred aged mice of mixed sexes and housed in conventional facilities makes mechanistic and reproducible studies difficult to near impossible, suggesting that some balance is needed. Clearly, these caveats need to be taken into consideration when attempting to translate the preclinical findings into clinical scenarios. Finally, there are clear differences, mechanistically and pathologically, between acute and chronic GVHD; treatments that work for acute GVHD may not work in chronic GVHD. The murine models for chronic GVHD are relatively underdeveloped and this also needs to be taken into consideration in murine HSCT studies.

Augmentation of graft-versus-tumor without graft-versus-host disease: striking the balance

Chemotherapy and corticosteroids are primary standards of care for cancer diseases; however, these treatments are associated with long-term side effects, immunosuppression and toxicity [66]. Additional therapy is clearly necessary in order to prevent graft rejection and sustain disease-free survival without GVHD while preserving GVT effects. The following sections and Table 2 will outline the different immune therapies used in HSCT and their effect on GVHD and GVT.

Cellular therapies

Donor lymphocyte infusion/adoptive cell therapy using T cells—The limited efficacy of HSCT is due to immunodeficiency, severe organ toxicity, GVHD, donor graft rejection and relapse. One way to boost GVT is by reinfusing lymphocytes from a BM donor into the host that previously received a BMT. By delaying the administration of the donor cells until long after the cytoreductive conditioning, there is less toxicity, presumably owing to the reduction in inflammatory cytokines and tissue damage from the conditioning procedure. DLI of T cells is used to treat residual cancer cells or recurrent disease post-HSCT in order to induce remission of the cancer by GVT, especially in CML patients. Full BM grafts or specific subsets of lymphocytes have been adoptively transferred in order to reduce the incidence of GVHD. CD8 depletion of BM in mice, or addition of CD4⁺ T cells

after allogeneic BMT, correlated with reduced GVHD in comparison with unmanipulated grafts [79], supporting the importance of CD4 T cells in GVL. By contrast to these data, BMT with CD8 TCD but without CD4 TCD, induced a high rate of leukemia relapse in mice [80], thus demonstrating that CD8 T cells also play a role in GVL. DLI can induce remissions in patients who have relapsed following allogeneic BMT [81] and can also result in significant acute and/or chronic GVHD. The primary treatment for CML involves combination chemotherapy in order to debulk the tumor load, followed by DLI. Nevertheless, the major complication after DLI is GVHD. Selective depletion of CD8 T cells from donor peripheral blood results in a decrease in GVHD incidence in patients with CML without losing GVL. In addition, purified CD8 T cells from the donor had better engraftment capacity, greater antitumor effects and no GVHD in a murine leukemia model after a first allogeneic BMT. Clinical studies of CD4-depleted BMT and adjustment of CD8 T-cell content demonstrated that CD8 T cells appear to help in the prevention of graft rejection, but no finite dose of CD8 T cells was found to control both GVHD and graft rejection [82]. Hence, these observations prove that both CD4 and CD8 T cells play a role in GVHD as well as in GVT. It is possible that the different outcomes after DLI are due to differences in the dosages of these immune competent cells and also to the timing of DLI in accordance with disease progression and relapse.

Another means of adoptively transferring lymphocytes involves the isolation of tumor-infiltrating lymphocytes (TILs), *ex vivo* expansion of these TILs and their reinfusion back into the patient. Such treatment is referred to as adoptive cell therapy (ACT) [83,84]. The most attractive aspect of ACT is the ability to grow large numbers of antitumor lymphocytes that are capable of repopulating in the patient and seeking out and killing tumor cells *in vivo*. The efficacy of ACT was first reported in patients with metastatic melanoma receiving autologous ACT and high-dose IL-2. Cancer regression in a third of the patients was achieved regardless of whether they received conditioning therapy prior to TILs transfer and IL-2 or not [85]. Nonetheless, one of the major problems of ACT is the short persistence of the transferred lymphocytes [86]. Eventually, later trials sought to combine ACT with different regimens of lymphodepletion in metastatic melanoma. The best objective response (up to 79%) was observed in a patient receiving ablative conditioning (chemotherapy, radiation and high-bolus IL-2) in comparison with 49% in nonmyeloablative and 52% in a second ablative conditioning with lower irradiation treatment [87,88]. Higher levels of the homeostatic cytokines IL-7 and IL-15 were detected and might therefore account for the longer persistence and efficacy of the transferred TILs [87]. Remarkably, genetically engineered peripheral blood lymphocytes expressing MART-1-specific TCR- α and - β chains were able to secrete IFN- γ upon stimulation with MART-1 peptide as well as HLA-A2 melanoma and lung and breast cancer [89]. Autologous ACT of MART-1 TCR lymphocytes after lymphodepletion resulted in the regression of metastatic melanoma in two patients who remained disease free 21 and 20 months post-transfer [89]. In addition, lymphodepletion in melanoma-bearing mice combined with removal of cellular cytokine sinks (removal of NK cells and absence of endogenous regulatory T cells [Tregs]) augments the antitumor effects of adoptively transferred tumor-specific CD8⁺ T cells (pmel-1) [90,91]. Although complete lymphodepletion of the host followed by ACT demonstrates promising results, this technique of engineering lymphocytes with TCR recognizing tumor-associated antigens is labor intensive and must be specific for the patient receiving the treatment owing to MHC restriction [92,93]. Along with gene transfer of T cells, education of T cells to express MHC-restricted TCR or non-MHC-restricted chimeric antigen receptors in order to recognize tumor antigens are alternative ways to circumvent tolerance and tumor escape. However, we will not discuss these therapies in detail as they are not used following HSCT and are still being investigated [94,95].

Recent studies have also given insights into the potential mechanisms by which HSCT can be used to augment ACT using T cells even in allogeneic HSCT. The initial hypotheses regarding HSCT in order to augment ACT revolved around the use of lymphodepletion in order to facilitate donor T-cell engraftment owing to homeostatic expansion of the donor cells. However, pivotal studies by Restifo *et al.* have shed new light on this [96]. Using murine HSCT models, they have demonstrated that the activation of host APCs by the conditioning regimen via Toll receptor engagement could facilitate the engraftment and activity of transferred antigen-specific T cells. Toll receptor engagement appeared dependent on lipopolysaccharide translocation in the gut after cytoreductive conditioning [97,98]. These results suggest that HSCT may be applied when ACT is used for other types of cancer, such as breast, renal cell carcinoma or melanoma, not just hematologic malignancies. However, toxicities arising from such intensive conditioning need to be taken into consideration, as well as the effects on GVHD if allogeneic HSCT is used.

Another means to exploit T cells for ACT following HSCT would be by converting them to a ‘memory’ phenotype (CD62L^{lo}/CD44^{hi}). Studies have found that memory phenotype T cells demonstrated impaired alloresponses both *in vitro* and *in vivo* [99,100], and clinical trials are currently underway evaluating their effects after HSCT. Recent human studies have also investigated activating T cells *ex vivo* with cytokines. These cytokine-induced killer cells are predominantly CD8⁺ and kill tumor targets via NKG2D [101]. Furthermore, they have been found to inhibit GVHD in murine allogeneic HSCT models by inhibiting donor T cells, also via NKG2D [102,103]. These cells resemble NK cells with regard to non-MHC-restricted killing activity but, similar to conventional T cells, have the potential ability to home towards nonhematopoietic tissues and, therefore, may be of benefit in solid tissue cancers.

Therefore, the use of T cells following HSCT is contingent on whether autologous or allogeneic HSCT is applied and if antigen-specific versus nonspecific T cells are present. Any attempts at using T cells following allogeneic HSCT needs to take into consideration the promotion of GVHD.

Natural killer cells—Natural killer cells are lymphocytes of the innate immune system that play an important role in host rejection of tumors, as well as viral and microbial infections [104–106]. Because NK cells have the ability to distinguish allogeneic hematopoietic cells from self and can mediate potent cytolytic effector function, they represent an attractive therapy in the settings of allogeneic BMT [107]. They mediate antigen-independent cell killing as opposed to the MHC-restricted cytotoxicity of T cells. NK cells express MHC-binding activating and inhibitory receptors and can modulate the immune response [104–106,108]. The inhibitory receptor families of cell surface molecules include Ly49 (in mouse), killer cell immunoglobulin-like receptor (KIR; in human) and CD94/NKG2 (in mouse and human) receptors that recognize self-MHC-I molecules and prevent NK-cell activation and lysis of target cells. Activating receptors, such as Ly49H and Ly49D in mice, trigger cell lysis [109]. Subsequently, in the context of mismatched allogeneic transplantation, the lack of self-MHC-I leads to NK-cell activation, target cell killing and cytokine production [104].

Natural killer cell-mediated rejection of BM in mice was first discovered in the 1960s, ultimately leading to the ‘missing self’ hypothesis. The critical observation was that, although F1 hybrid mice would accept solid organ transplants from either parent, they rejected parental BMT [110–113]. This reaction is due to a mismatch between inhibitory receptors on donor NK cells (Ly49s in mouse) and their MHC-I ligands on host allogeneic targets, as well as the presence of activating ligands on the hematopoietic cells leading to NK-cell alloreactivity and graft rejection [114,115]. We have previously demonstrated that

adoptive transfer of donor-type activated NK cells following allogeneic BMT in mice with colon adenocarcinoma resulted in increased survival by promoting GVT and delaying or inhibiting GVHD [116]. This has been confirmed with clinical studies demonstrating that NK cells alloreactivity prevented GVHD and leukemia relapse in patients with AML [117,118]. Another study used delayed donor NK-cell infusions following allogeneic HSCT in patients with high-risk myeloid malignancies with promising results [119]. Murine studies in which inhibitory receptors on NK cells were blocked before NK transfer, have found increased antitumor effects [120], indicating that blocking negative or inhibitory receptors can augment the efficacy of NK-cell adoptive immunotherapy (AIT).

Human NK cells express a functionally comparable but structurally different class of NK-cell receptors called KIRs that bind HLA-A, HLA-B and HLA-C antigens. Most KIRs are inhibitory, and once they recognize MHC class I molecules, they suppress the cytotoxic activity of their NK cells, whereas the activating KIRs are responsible for activating NK cells. In patients suffering from AML, graft failure and relapse rates decreased with mismatched KIR ligands (missing HLA class I, which is specific for KIR), suggesting the potential use of NK-cell transplants by typing donor–recipient combinations based on KIR differences [121–123]. However, these results were not observed in patients with acute lymphocytic leukemia. Subsequently, further data demonstrated that manipulation of NK cells before transfer of HSCT or isolating NK-cell subsets that lack specific inhibitory receptors could be a promising means to augment GVT and inhibit GVHD. The issues with NK-cell transfer revolve around whether toxicities such as delayed engraftment can occur and what is the optimal means to administer NK-cell therapy (whether exogenous cytokines such as IL-2 or IL-15 also need to be given) following HSCT and whether particular NK cell subsets offer superior effects. Natural killer cells appear to play a predominant role in hematologic malignancies; therefore, AIT of NK cells may be best in these types of cancers rather than solid cancers. However, in the case of metastatic cancers, the ability of NK cells to home to the lung and liver after intravenous injection makes their use potentially promising following HSCT.

Donor regulatory T-cell infusion—CD4⁺ CD25⁺ Foxp3⁺ cells are a subpopulation of T cells that downregulate the immune system and are therefore known as Tregs. They are important in the treatment of autoimmune diseases [124] such as diabetes and inflammatory bowel disease, and also play an important role in the control of homeostatic proliferation and peripheral tolerance [125–127]. One piece of evidence supporting the importance of Tregs in alloreactivity was identified by Sakaguchi *et al.* in mice that rejected skin grafts in the absence of these suppressor cells [128]. Furthermore, the immunosuppressive effects of Tregs resulted in tissue engraftment, and these Tregs were specifically tolerant to antigens of the allograft [129–131]. While transfer of host-type Tregs can delay GVHD, mice eventually succumb to the disease. However, murine donor-type Tregs are able to protect from lethal GVHD with 100% survival at 100 days post-transplantation [132]. The mechanism of GVHD protection by Tregs is, in part, mediated by IL-10 production and control of donor antihost alloactivation [132]. Blazar *et al.* have demonstrated that depletion of CD4⁺ CD25⁺ T cells in a murine allogeneic model completely blocked the *ex vivo* tolerization to host alloantigens and resulted in GVHD [133]. The role of Tregs in preventing GVHD has been demonstrated in studies in which these cells are infused back to the patients, resulting in protection from GVHD. If Tregs are expanded and activated *ex vivo* prior to reinfusion, they can suppress alloantigen responses nonspecifically and one can also infuse a larger number of Tregs [134]. When CD4⁺ CD25⁺ cells are depleted from donor grafts, *ex vivo* tolerization to host alloantigens is abrogated and GVHD is induced. In humans, more research is needed in order to investigate the effect of Tregs *ex vivo* expansion of Tregs on GVHD. Most importantly, infusion of Tregs following allogeneic BMT prevented GVHD without losing the perforin-mediated GVT effector function in both lymphoma and leukemia mouse models

[135–137]. Questions still remain concerning the activation status of the Tregs as related to their function and longevity. As with many immune cell types, prior activation *ex vivo* leads to greater effector function but also limited expansion potential and survival when transferred to recipients. In addition, it is not clear as to how the inhibition of GVHD with retention of GVT occurs and this may be due to differences in kinetics and may also be contingent on the immunogenicity of the tumor. Depletion of Tregs is under active investigation to promote antitumor immunity, but this may involve Tregs localized in the tumor such that location of Tregs may allow discerning GVHD/GVT effects.

Manipulation of antigen-presenting cells—Cytoreductive conditioning results in host tissue damage and the secretion of proinflammatory cytokines, such as TNF- α and IL-1, which activate host APCs that in turn activate donor T cells, leading to further production of cytokines by T cells. This ‘cytokine storm’ amplifies local tissue injury and can result in damage to the gastrointestinal tract in murine and human studies [98,138–140]. Most importantly, acute GVHD is initiated by host APCs even in the absence of alloantigen on host tissues, suggesting the role of host APCs in inducing acute GVHD [141]. In murine allogeneic BMT models, depletion of host APCs using MHC class II-deficient mice prior to cytoreductive immunosuppression abrogated GVHD [142,143] and the presence of APCs, especially host DCs [144], 3 months post-BMT is associated with the severity of acute GVHD in humans [145]. Other reports demonstrate that donor APCs do not drive a GVHD response post-transplantation but can induce host tissue injury owing to cross-presentation of self-antigens from host to donor T cells [146–148]. Schlomchik *et al.* have demonstrated the importance of both donor and host APCs in inducing skin and intestinal GVHD, respectively, in a mouse model [149]. Therefore, manipulation of both donor and host APCs in the clinic by depletion of host APCs before BMT presents a potential avenue for the prevention of acute GVHD. However, little is known regarding the different types of professional APCs (e.g., myeloid DCs and plasmacytoid DCs among others) required to drive GVHD and their effect on GVT. This is important as DCs are being actively explored in tumor vaccine models and their effects on GVHD may be critically dependent on timing. Donor DCs administered long after HSCT may allow for the augmentation of GVT without GVHD.

Use of molecular targeting agents as immunomodulatory agents

Small molecules that target particular intra-cellular pathways have demonstrated great promise as direct anticancer agents. The pathways targeted can be the proteasome, the transcription factor NF- κ B or histone deacetylases (HDACs). While capable of exerting direct antitumor effects, it has become increasingly clear that these agents also exert immunomodulatory effects and can be used following HSCT, either autologous with ACT or allogeneic to modulate GVHD.

Proteasome inhibition—Important pathways that regulate cellular functions, such as cell growth/proliferation and apoptosis, are controlled by the transcription factor NF- κ B [51,150,151]. In the cytoplasm, NF- κ B dimers associate with inhibitor proteins I κ Bs and keep NF- κ B inactive. Once I κ Bs are phosphorylated, NF- κ B dissociates and enters the nucleus to carry out its functions [152]. Bortezomib (Velcade®, Millennium Pharmaceuticals, Inc., MA, USA; formerly PS-341) is a proteasome inhibitor and specifically inhibits the ubiquitin-dependent 26S proteasome unit that regulates intracellular protein degradation [153]. Proteasome inhibition leads to blockade of the NF- κ B pathway, which is constitutively activated in multiple myeloma (MM) [154]. Bortezomib has direct antitumor efficacy and is clinically approved for use in MM [155]. Our group has previously demonstrated protection against GVHD in leukemia-bearing mice that received bortezomib immediately after allogeneic BMT [156]. This protection is also accompanied by the

retention of GVT effects and a marked decrease in the secretion of proinflammatory cytokines, such as TNF- α , IL-1 and IL-6. However, efficacy of bortezomib in GVHD protection was critically dependent on the timing of administration of the drug. Delayed bortezomib treatment resulted in marked gut lesions and increased lethality after allogeneic HSCT in mice [157]. This was found to be correlated with increases in TNF-receptor I in the gut [158]. Bortezomib-GVHD dependent toxicity is also associated with different subsets of T cells in the graft. In a recent study, we have demonstrated that removal of CD4⁺ T cells from donor grafts prevented this toxicity, and transfer of CD8⁺ T cells with bortezomib administration enhanced GVT in mice. Survival was correlated with reduced secretion of TNF- α and increased IFN- γ [157]. Recently, in a Phase I clinical trial, bortezomib administration with tacrolimus and methotrexate following nonmyeloablative allogeneic HSCT prevented GVHD with minimal toxicity [159]. Questions concerning the use of bortezomib following ACT and HSCT revolve around the potential effects on immune reconstitution, as increased susceptibility to viral infections has been reported in patients receiving bortezomib [160]. In addition, the timing of administration may be critical, in that one wants to increase the tumor susceptibility to immune attack but not have increased immune cell clearance as well. Bortezomib has also been demonstrated to increase susceptibility to various tumor cells to immune-mediated attack. Finally, owing to the effects of bortezomib on plasma cells and potential roles of B cells in chronic GVHD, there is great interest in examining the potential effects of bortezomib on chronic GVHD models.

Histone deacetylase inhibitors—Administration of suberoylanilide hydroxamic acid (SAHA; vorinostat) after allogeneic BMT dampened the cytokine storm, leading to protection from GVHD and maintaining GVL effects in mice [161]. SAHA is an inhibitor of HDAC and causes cell differentiation, growth or cell arrest and apoptosis [162]. Importantly, SAHA has generated antitumor effects *in vitro* and *in vivo* in a prostate cancer model [163] and in a breast cancer model [164] with decreased production of pro-inflammatory cytokines such as TNF- α , IL-1 β , IL-6 and IFN- γ [165]. In a Phase I trial, patients with advanced solid tumors and hematological malignancies (refractory or relapsing leukemia, MM, indolent or aggressive non-Hodgkin's lymphoma, mantle cell lymphoma or Hodgkin's disease), demonstrated antitumor effects after systemic administration of SAHA [166]. However, similarly to bortezomib, late administration of SAHA and other HDAC inhibitors needs to be assessed in order to ascertain whether similar toxicities can result. Interestingly, owing to their different mechanisms of action, potential synergistic effects may result when SAHA and bortezomib are administered together, but further investigations need to be carried out in order to confirm this.

Targeting of costimulatory pathways

Host DCs are major agents in acute GVHD and therefore represent a good target for treatment. One method of GVHD treatment is to inhibit the interaction between APCs and DCs by blocking costimulatory molecules. CTL antigen-4 (CTLA-4; or CD152) is an inhibitory receptor that is upregulated on activated T cells and binds to B7.1 (CD80) and B7.2 (CD86) on APCs with higher affinity than CD28 [167–170]. CTLA-4 functions to inhibit T-cell proliferation [170–172] and CTLA-4/B7 interaction reduces GVHD morbidity in mice [173,174]. However, CTLA-4 blockade also augments antitumor immunity, suggesting that GVHD/GVT processes will cancel each other out.

Another negative costimulatory molecule is programmed death (PD)-1, which is expressed on activated T cells, B cells and macrophages [175,176]. PD-1 has two ligands: PD-L1 or B7-H1 and PD-L2 or B7-DC. PD-L1 is constitutively expressed on a variety of cells such as DCs, T and B cells, macrophages, mesenchymal stem cells and BM-derived mast cells, and is highly inducible by IFN- γ [175]. However, PD-L2 expression is more restricted to DCs,

macrophages and BM-derived mast cells. PD-1/PD-L1 interactions induce T-cell inhibition. Our group has demonstrated a loss of CD4 T cells after immunotherapy, which correlated with IFN- γ stimulation, upregulation of the surface marker B7-H1 and an increase in PD-1 on CD4⁺ T cells [177]. Targeting PD-1 after limited immune suppression with cyclosporin A or rapamycin inhibited cardiac allograft rejection in mice [178]. Blazar and colleagues have also demonstrated that blockade of the PD-1 pathway accelerates GVHD mortality in mice in an IFN- γ -dependent manner. In addition, coblockade of PD-1 and CTLA-4 resulted in even more pronounced GVHD lethality [179]. Therefore, triggering PD-1 and CTLA-4 has potential for therapeutic treatment of GVHD, but may inhibit GVT.

CD40 ligand or CD154 is expressed on activated CD4⁺ T lymphocytes and CD40 ligation by CD40L induces activation of APCs and consequently, CD4 and CD8 T-cell alloresponses. OX-40 or CD134 is a secondary costimulatory molecule and a member of the TNF-receptor family. CD134 is upregulated on activated T cells and its ligand, CD134L, is only found on activated APCs. Once CD134 is triggered, CD4⁺ T cells become activated and produce Th1 or Th2 cytokines. Antagonistic anti-OX-40L mAb reduced GVHD mortality in mice and was independent of CD28 stimulation [180]. Thus, targeting of these pathways may not only affect GVHD but also affect GVT. As with any T-cell targeting, the goal of autologous ACT is augmenting any T-cell responses, while there is a critical need to assess the effects of allogeneic HSCT on GVHD. Thus, while manipulation of costimulatory molecules may inhibit GVHD, it will also probably impair GVT responses unless alteration of timing is applied.

Cytokines

Cytokines play a pivotal role in the pathophysiology of GVHD. During the induction phase, donor T cells proliferate and become activated, secreting cytokines and chemokines, resulting in recruitment of other cell types and causing damage to host tissues. CD4⁺ T cells and CD8⁺ T cells can polarize into type I (IL-2 and IFN- γ) and type II (IL-4 and IL-10) and now Th17 (IL-17 and IL-22) secreting cytokines. Earlier reports have demonstrated the role of Th1 cytokines such as IL-12 and IFN- γ in the generation of GVHD. However, regulation of GVHD is also dependent on the timing and dosage of administration of exogenous IL-12, as well as on the type of body radiation used. Fas upregulation on the donor T cells is thought to be one mechanism of inhibition of GVHD by IL-12. Conversely, at the other end of the spectrum, *ex vivo* polarization of donor T cells to a Th2 profile with rapamycin inhibits murine GVHD and maintains GVT after allogeneic BMT in an IL-4-dependent fashion [181]. Transplantation of *ex vivo*-skewed Th2 T cells post-BMT resulted in suppression of GVHD and increased survival [182]. Clinical trials using Th2 or CD8 Tc2 cells are underway. The critical issue again revolves around the potential effects on GVT. Abrogation of GVHD correlates with IL-4 and IL-10 production by donor T cells and could be blocked by the administration of IL-2. The administration of IL-17 has been more controversial [183,184]. Together, these data suggest that cytokine therapy might have opposing outcomes on GVHD induction, dependent on timing, dosage and type of cytokine production by donor T cells. Clinical studies have to be carefully designed in order to use cytokine therapy or blocking antibodies such that a balance between GVHD and GVT is attained. TNF- α is another major player in the pathophysiology of GVHD in the induction and effector phase of disease. A US FDA-approved drug, infliximab, is an antibody that chimeric murine-human IgG1 inhibits the biological function of TNF- α by binding to both soluble and membrane-bound TNF- α , therefore inducing lysis of cells expressing TNF- α . Infliximab is used for autoimmune diseases. In a retrospective study, among 21 subjects with steroid-refractory acute GVHD treated with infliximab as a single agent, 67% demonstrated a response, with 62% achieving a complete response. Although patients did not experience toxicity from infliximab treatment, several complications were reported,

including an increased incidence in bacterial, viral and fungal infections [185]. In this study, TNF- α blockade, particularly in gastrointestinal steroid-refractory GVHD, demonstrates promising results, but further studies addressing effects on infectious and fungal diseases need to be better explored in clinical trials. Finally, etanercept (Enbrel®, Immunex Corporation, CA, USA) is another drug used to block TNF- α activity, sparing lysis of cells that express TNF. Enbrel is a fusion protein in which two soluble TNF receptors are fused to the Fc region of human IgG₁. Because TNF plays a central role in pathophysiology of GVHD and severe inflammation, Enbrel has been used in autoimmune diseases and rheumatoid arthritis as well as GVHD. In preliminary human studies, Enbrel demonstrates improvement in steroid-dependent chronic GVHD, but again, side effects were reported to be similar to infliximab [186]. In a Phase II study, combination Enbrel and daclizumab (a humanized mAb that targets the α -unit of the IL-2 receptor on T cells [187]) again ameliorated progression of steroid-refractory acute GVHD and mortality owing to infections [188]. In addition, Enbrel administration is possibly associated with significant improvements in idiopathic pneumonia syndrome following allogeneic HSCT [189]. The use of TNF blockade may also offer other advantages in cancer by inhibiting inflammatory processes employed by the tumor. Blockade of inflammatory cytokines, such as TNF and IL-6 (alone or combined), offers great promise in inhibiting GVH and impairing tumor progression.

Mesenchymal stem cells

Mesenchymal stem cells are non-HSCs found in the BM and can differentiate into a variety of cell types. MSCs have the ability to migrate to sites of injury and are therefore of potential use for tissue repair, including GVHD target tissues. Intravenous administration of donor MSCs to baboons before autologous skin graft demonstrated enhanced survival [190] and suppressed T-cell proliferative responses to mitogens. Furthermore, human MSCs suppressed human mixed lymphocyte reaction. This T-cell inhibition was also independent of the MHC [191].

Recent studies in rats using allogeneic HSCT with donor-derived T cells cotransplanted with BM-derived MSCs demonstrated a decrease in acute-GVHD by means of a decrease in CD4⁺ T and CD8⁺ T cells and an increase in CD4 conventional T cells (CD4⁺ CD25⁺) [192]. Most importantly, in a Phase II study, infusion of MSCs previously expanded *in vitro* reduced the risk of GVHD. Of 55 patients, 30 had a complete response independent of donor HLA match, and no side effects were observed during or after transplantation [193,194]. Nonetheless, in a randomized study, transplantation of MSCs into patients with AML had no effect on GVL, and GVHD was associated with a higher relapse rate [195]. This is not surprising given that GVHD and GVT are tightly associated. There may also be potentially deleterious effects of MSCs in cancer therapy as it has been demonstrated that MSCs can affect breast cancer progression by epigenetic means and promote metastasis [196]. Some laboratories are also examining MSCs for delivering cytokines or death receptor ligands that may augment GVT.

Regardless, effects on relapse need to be carefully monitored. Allogeneic HSCT with AIT has reached a point where broader applications to multiple cancers, including solid tissue cancers, are feasible. The critical issue revolves around attempting to 'control' the immune effectors, such that GVHD is not exacerbated.

Bibliography

Papers of special note have been highlighted as:

- of interest

■ of considerable interest

1. Kurnick NB, Montano A, Gerdes JC, Feder BH. Preliminary observations on the treatment of postirradiation hematopoietic depression in man by the infusion of stored autogenous bone marrow. *Ann Intern Med.* 1958; 49(5):973–986. [PubMed: 13595468]
2. Varas F, Bernad A, Bueren JA. Granulocyte colony-stimulating factor mobilizes into peripheral blood the complete clonal repertoire of hematopoietic precursors residing in the bone marrow of mice. *Blood.* 1996; 88(7):2495–2501. [PubMed: 8839841]
3. Plerixafor: AMD3100, AMD3100, JM 3100, SDZ SID 791. *Drugs RD.* 2007; 8(2):113–119.
4. Cashen AF, Nervi B, DiPersio J. AMD3100: CXCR4 antagonist and rapid stem cell-mobilizing agent. *Future Oncol.* 2007; 3(1):19–27. [PubMed: 17280498]
5. Uy GL, Rettig MP, Cashen AF. Plerixafor, a CXCR4 antagonist for the mobilization of hematopoietic stem cells. *Expert Opin Biol Ther.* 2008; 8(11):1797–1804. [PubMed: 18847313]
6. Blaise D, Kuentz M, Fortanier C, et al. Randomized trial of bone marrow versus lenograstim-primed blood cell allogeneic transplantation in patients with early-stage leukemia: a report from the Societe Francaise de Greffe de Moelle. *J Clin Oncol.* 2000; 18(3):537–546. [PubMed: 10653869]
7. Schmitz N, Bacigalupo A, Hasenclever D, et al. Allogeneic bone marrow transplantation vs filgrastim-mobilised peripheral blood progenitor cell transplantation in patients with early leukaemia: first results of a randomised multicentre trial of the European Group for Blood and Marrow Transplantation. *Bone Marrow Transplant.* 1998; 21(10):995–1003. [PubMed: 9632272]
8. Singhal S, Powles R, Kulkarni S, et al. Comparison of marrow and blood cell yields from the same donors in a double-blind, randomized study of allogeneic marrow vs blood stem cell transplantation. *Bone Marrow Transplant.* 2000; 25(5):501–505. [PubMed: 10713626]
9. Heldal D, Tjonnfjord G, Brinch L, et al. A randomised study of allogeneic transplantation with stem cells from blood or bone marrow. *Bone Marrow Transplant.* 2000; 25(11):1129–1136. [PubMed: 10849524]
10. Powles R, Mehta J, Kulkarni S, et al. Allogeneic blood and bone-marrow stem-cell transplantation in haematological malignant diseases: a randomised trial. *Lancet.* 2000; 355(9211):1231–1237. [PubMed: 10770306]
11. Schmitz N, Dreger P, Suttorp M, et al. Primary transplantation of allogeneic peripheral blood progenitor cells mobilized by filgrastim (granulocyte colony-stimulating factor). *Blood.* 1995; 85(6):1666–1672. [PubMed: 7534141]
12. Appelbaum FR. Haematopoietic cell transplantation as immunotherapy. *Nature.* 2001; 411(6835):385–389. [PubMed: 11357147]
13. Thomas ED. Karnofsky memorial lecture. Marrow transplantation for malignant diseases. *J Clin Oncol.* 1983; 1(9):517–531. [PubMed: 6366143]
14. Cairo MS, Wagner JE. Placental and/or umbilical cord blood: an alternative source of hematopoietic stem cells for transplantation. *Blood.* 1997; 90(12):4665–4678. [PubMed: 9389681]
15. Rubinstein P, Carrier C, Scaradavou A, et al. Outcomes among 562 recipients of placental-blood transplants from unrelated donors. *N Engl J Med.* 1998; 339(22):1565–1577. [PubMed: 9828244]
16. Wagner JE, Kernan NA, Steinbuch M, Broxmeyer HE, Gluckman E. Allogeneic sibling umbilical-cord-blood transplantation in children with malignant and non-malignant disease. *Lancet.* 1995; 346(8969):214–219. [PubMed: 7616801]
17. MacMillan ML, Weisdorf DJ, Brunstein CG, et al. Acute graft-versus-host disease after unrelated donor umbilical cord blood transplantation: analysis of risk factors. *Blood.* 2009; 113(11):2410–2415. [PubMed: 18997171]
18. Verneris MR, Brunstein CG, Barker J, et al. Relapse risk after umbilical cord blood transplantation: enhanced graft-versus-leukemia effect in recipients of 2 units. *Blood.* 2009; 114(19):4293–4299. [PubMed: 19706886]
19. Aversa F. Haploidentical haematopoietic stem cell transplantation for acute leukaemia in adults: experience in Europe and the United States. *Bone Marrow Transplant.* 2008; 41(5):473–481. [PubMed: 18176612]

20. Aversa F, Berneman ZN, Locatelli F, et al. Fourth International Workshop on Haploidentical Transplants, Naples, Italy, July 8–10, 2004. *Blood Cells Mol Dis.* 2004; 33(3):159–175. [PubMed: 15580686]
21. Aversa F, Terenzi A, Felicini R, et al. Haploidentical stem cell transplantation for acute leukemia. *Int J Hematol.* 2002; 76(Suppl):1165–1168.
22. Aversa F, Velardi A, Tabilio A, Reisner Y, Martelli MF. Haploidentical stem cell transplantation in leukemia. *Blood Rev.* 2001; 15(3):111–119. [PubMed: 11735159]
23. Gassas A, Sung L, Saunders EF, Doyle J. Graft-versus-leukemia effect in hematopoietic stem cell transplantation for pediatric acute lymphoblastic leukemia: significantly lower relapse rate in unrelated transplantations. *Bone Marrow Transplant.* 2007; 40(10):951–955. [PubMed: 17873916]
24. Khouri IF, Saliba RM, Giralt SA, et al. Nonablative allogeneic hematopoietic transplantation as adoptive immunotherapy for indolent lymphoma: low incidence of toxicity, acute graft-versus-host disease, and treatment-related mortality. *Blood.* 2001; 98(13):3595–3599. [PubMed: 11739162]
25. Schetelig J, Thiede C, Bornhauser M, et al. Reduced non-relapse mortality after reduced intensity conditioning in advanced chronic lymphocytic leukemia. *Ann Hematol.* 2002; 81(Suppl 2):S47–S48. [PubMed: 12611075]
26. Schetelig J, Thiede C, Bornhauser M, et al. Evidence of a graft-versus-leukemia effect in chronic lymphocytic leukemia after reduced-intensity conditioning and allogeneic stem-cell transplantation: the Cooperative German Transplant Study Group. *J Clin Oncol.* 2003; 21(14):2747–2753. [PubMed: 12860954]
27. Weiden PL, Flournoy N, Thomas ED, et al. Antileukemic effect of graft-versus-host disease in human recipients of allogeneic-marrow grafts. *N Engl J Med.* 1979; 300(19):1068–1073. [PubMed: 34792]
28. Deeg HJ, Storb R. Acute and chronic graft-versus-host disease: clinical manifestations, prophylaxis, and treatment. *J Natl Cancer Inst.* 1986; 76(6):1325–1328. [PubMed: 3458963]
29. Ratanatharathorn V, Ayash L, Lazarus HM, Fu J, Uberti JP. Chronic graft-versus-host disease: clinical manifestation and therapy. *Bone Marrow Transplant.* 2001; 28(2):121–129. [PubMed: 11509929]
30. Kansu E. The pathophysiology of chronic graft-versus-host disease. *Int J Hematol.* 2004; 79(3):209–215. [PubMed: 15168586]
31. Tyndall A, Dazzi F. Chronic GVHD as an autoimmune disease. *Best Pract Res Clin Haematol.* 2008; 21(2):281–289. [PubMed: 18503993]
32. Bryson JS, Jennings CD, Caywood BE, Dix AR, Lowery DM, Kaplan AM. Enhanced graft-versus-host disease in older recipient mice following allogeneic bone marrow transplantation. *Bone Marrow Transplant.* 1997; 19(7):721–728. [PubMed: 9156250]
33. Remberger M, Kumlien G, Aschan J, et al. Risk factors for moderate-to-severe chronic graft-versus-host disease after allogeneic hematopoietic stem cell transplantation. *Biol Blood Marrow Transplant.* 2002; 8(12):674–682. [PubMed: 12523580]
34. Fischer AC, Hess AD. Age-related factors in cyclosporine-induced syngeneic graft-versus-host disease: regulatory role of marrow-derived T lymphocytes. *J Exp Med.* 1990; 172(1):85–94. [PubMed: 2358786]
35. Parmar S, Del Lima M, Zou Y, et al. Donor-recipient mismatches in MHC class I chain-related gene A in unrelated donor transplantation lead to increased incidence of acute graft-versus-host disease. *Blood.* 2009; 114(14):2884–2887. [PubMed: 19654407]
36. Anasetti C, Beatty PG, Storb R, et al. Effect of HLA incompatibility on graft-versus-host disease, relapse, and survival after marrow transplantation for patients with leukemia or lymphoma. *Hum Immunol.* 1990; 29(2):79–91. [PubMed: 2249952]
37. Antin JH, Ferrara JL. Cytokine dysregulation and acute graft-versus-host disease. *Blood.* 1992; 80(12):2964–2968. [PubMed: 1467511]
38. Ferrara JL, Levine JE, Reddy P, Holler E. Graft-versus-host disease. *Lancet.* 2009; 373(9674):1550–1561. [PubMed: 19282026]
39. Loiseau P, Busson M, Balere ML, et al. HLA Association with hematopoietic stem cell transplantation outcome: the number of mismatches at HLA-A, -B, -C, -DRB1, or -DQB1 is

- strongly associated with overall survival. *Biol Blood Marrow Transplant.* 2007; 13(8):965–974. [PubMed: 17640601]
40. Antin JH. Clinical practice. Long-term care after hematopoietic-cell transplantation in adults. *N Engl J Med.* 2002; 347(1):36–42. [PubMed: 12097539]
  41. Couriel D, Caldera H, Champlin R, Komanduri K. Acute graft-versus-host disease: pathophysiology, clinical manifestations, and management. *Cancer.* 2004; 101(9):1936–1946. [PubMed: 15372473]
  42. Barnes DW, Loutit JF. Treatment of murine leukaemia with x-rays and homologous bone marrow. II. *Br J Haematol.* 1957; 3(3):241–252. [PubMed: 13460193]
  43. Bar BM, Schattenberg A, Mensink EJ, et al. Donor leukocyte infusions for chronic myeloid leukemia relapsed after allogeneic bone marrow transplantation. *J Clin Oncol.* 1993; 11(3):513–519. [PubMed: 8445426]
  44. Marmont AM, Horowitz MM, Gale RP, et al. T-cell depletion of HLA-identical transplants in leukemia. *Blood.* 1991; 78(8):2120–2130. [PubMed: 1912589]
  45. Mitsuyasu RT, Champlin RE, Gale RP, et al. Treatment of donor bone marrow with monoclonal anti-T-cell antibody and complement for the prevention of graft-versus-host disease. A prospective, randomized, double-blind trial. *Ann Intern Med.* 1986; 105(1):20–26. [PubMed: 3521427]
  46. Goldman JM, Gale RP, Horowitz MM, et al. Bone marrow transplantation for chronic myelogenous leukemia in chronic phase. Increased risk for relapse associated with T-cell depletion. *Ann Intern Med.* 1988; 108(6):806–814. [PubMed: 3285744]
  47. Marks DI, Hughes TP, Szydlo R, et al. HLA-identical sibling donor bone marrow transplantation for chronic myeloid leukaemia in first chronic phase: influence of GVHD prophylaxis on outcome. *Br J Haematol.* 1992; 81(3):383–390. [PubMed: 1390211]
  48. Couriel DR, Saliba RM, Giralt S, et al. Acute and chronic graft-versus-host disease after ablative and nonmyeloablative conditioning for allogeneic hematopoietic transplantation. *Biol Blood Marrow Transplant.* 2004; 10(3):178–185. [PubMed: 14993883]
  49. Sorror ML, Maris MB, Sandmaier BM, et al. Hematopoietic cell transplantation after nonmyeloablative conditioning for advanced chronic lymphocytic leukemia. *J Clin Oncol.* 2005; 23(16):3819–3829. [PubMed: 15809448]
  50. Alyea EP, Kim HT, Ho V, et al. Comparative outcome of nonmyeloablative and myeloablative allogeneic hematopoietic cell transplantation for patients older than 50 years of age. *Blood.* 2005; 105(4):1810–1814. [PubMed: 15459007]
  51. Vodanovic-Jankovic S, Hari P, Jacobs P, Komorowski R, Drobyski WR. NF- κ B as a target for the prevention of graft-versus-host disease: comparative efficacy of bortezomib and PS-1145. *Blood.* 2006; 107(2):827–834. [PubMed: 16174760]
  52. Welniak LA, Blazar BR, Murphy WJ. Immunobiology of allogeneic hematopoietic stem cell transplantation. *Annu Rev Immunol.* 2007; 25:139–25170.
  53. Ferrara JL. Novel strategies for the treatment and diagnosis of graft-versus-host-disease. *Best Pract Res Clin Haematol.* 2007; 20(1):91–97. [PubMed: 17336259]
  54. Ferrara JL, Cooke KR, Pan L, Krenger W. The immunopathophysiology of acute graft-versus-host-disease. *Stem Cells.* 1996; 14(5):473–489. [PubMed: 8888489]
  55. Sun Y, Tawara I, Toubai T, Reddy P. Pathophysiology of acute graft-versus-host disease: recent advances. *Transl Res.* 2007; 150(4):197–214. [PubMed: 17900507]
  56. Tsukada N, Kobata T, Aizawa Y, Yagita H, Okumura K. Graft-versus-leukemia effect and graft-versus-host disease can be differentiated by cytotoxic mechanisms in a murine model of allogeneic bone marrow transplantation. *Blood.* 1999; 93(8):2738–2747. [PubMed: 10194454]
  57. Krenger W, Hill GR, Ferrara JL. Cytokine cascades in acute graft-versus-host disease. *Transplantation.* 1997; 64(4):553–558. [PubMed: 9293864]
  58. Murphy WJ, Welniak LA, Taub DD, et al. Differential effects of the absence of interferon-gamma and IL-4 in acute graft-versus-host disease after allogeneic bone marrow transplantation in mice. *J Clin Invest.* 1998; 102(9):1742–1748. Cytokines play a pivotal role in the pathophysiology of graft-versus-host disease (GVHD) and this is dependent on the type of cytokine secreted. Surprisingly, Th1 cytokines, such as IFN- γ , appear to be protective whereas, by inducing

activation-induced cell death of the donor T cells, Th2 cytokines such as IL-4 augmented GVHD in murine models. The results also demonstrated that the particular effect of a cytokine on GVHD severity was concomitant with the stage of progression of GVHD. [PubMed: 9802888]

59. Basara N, Blau WI, Kiehl MG, et al. Efficacy and safety of mycophenolate mofetil for the treatment of acute and chronic GVHD in bone marrow transplant recipient. *Transplant Proc.* 1998; 30(8):4087–4089. [PubMed: 9865306]
60. Basara N, Kiehl MG, Blau W, et al. Mycophenolate mofetil in the treatment of acute and chronic GVHD in hematopoietic stem cell transplant patients: four years of experience. *Transplant Proc.* 2001; 33(3):2121–2123. [PubMed: 11377471]
61. Bolanos-Meade J, Jacobsohn DA, Margolis J, et al. Pentostatin in steroid-refractory acute graft-versus-host disease. *J Clin Oncol.* 2005; 23(12):2661–2668. [PubMed: 15837980]
62. Maraninchi D, Gluckman E, Blaise D, et al. Impact of T-cell depletion on outcome of allogeneic bone-marrow transplantation for standard-risk leukaemias. *Lancet.* 1987; 2(8552):175–178. [PubMed: 2885638]
63. Wagner JE, Donnenberg AD, Noga SJ, et al. Lymphocyte depletion of donor bone marrow by counterflow centrifugal elutriation: results of a Phase I clinical trial. *Blood.* 1988; 72(4):1168–1176. [PubMed: 3048436]
64. Drobyski WR, Ash RC, Casper JT, et al. Effect of T-cell depletion as graft-versus-host disease prophylaxis on engraftment, relapse, and disease-free survival in unrelated marrow transplantation for chronic myelogenous leukemia. *Blood.* 1994; 83(7):1980–1987. [PubMed: 8142664]
65. Pavletic SZ, Carter SL, Kernan NA, et al. Influence of T-cell depletion on chronic graft-versus-host disease: results of a multicenter randomized trial in unrelated marrow donor transplantation. *Blood.* 2005; 106(9):3308–3313. [PubMed: 16046530]
66. Storb R. Bone marrow transplantation. *Transplant Proc.* 1995; 27(5):2649–2652. [PubMed: 7482865]
67. Feugier P, Thevenin S, Billot M, et al. Comparison of T cell depletion strategies from bone marrow, umbilical cord and peripheral blood using five separation systems. *Hematol Cell Ther.* 1997; 39(2):67–73. [PubMed: 9168302]
68. Hale G, Xia MQ, Tighe HP, Dyer MJ, Waldmann H. The CAMPATH-1 antigen (CDw52). *Tissue Antigens.* 1990; 35(3):118–127. [PubMed: 2165283]
69. Waldmann H, Hale G. CAMPATH: from concept to clinic. *Philos Trans R Soc Lond B Biol Sci.* 2005; 360(1461):1707–1711. [PubMed: 16147535]
70. Hale G, Waldmann H. Risks of developing Epstein–Barr virus-related lymphoproliferative disorders after T-cell-depleted marrow transplants. *CAMPATH Users. Blood.* 1998; 91(8):3079–3083. [PubMed: 9531622]
71. Chu YW, Gress RE. Murine models of chronic graft-versus-host disease: insights and unresolved issues. *Biol Blood Marrow Transplant.* 2008; 14(4):365–378. [PubMed: 18342778]
72. Reddy P, Negrin R, Hill GR. Mouse models of bone marrow transplantation. *Biol Blood Marrow Transplant.* 2008; 14(1 Suppl 1):129–135. [PubMed: 18162233]
73. Storb R, Yu C, Wagner JL, et al. Stable mixed hematopoietic chimerism in DLA-identical littermate dogs given sublethal total body irradiation before and pharmacological immunosuppression after marrow transplantation. *Blood.* 1997; 89(8):3048–3054. [PubMed: 9108426]
74. Atkinson K, Shulman HM, Deeg HJ, et al. Acute and chronic graft-versus-host disease in dogs given hemopoietic grafts from DLA-nonidentical littermates. Two distinct syndromes. *Am J Pathol.* 1982; 108(2):196–205. [PubMed: 6765733]
75. Kolb H, Sale GE, Lerner KG, Storb R, Thomas ED. Pathology of acute graft-versus-host disease in the dog. An autopsy study of ninetyfive dogs. *Am J Pathol.* 1979; 96(2):581–594. [PubMed: 38670]
76. Mestas J, Hughes CC. Of mice and not men: differences between mouse and human immunology. *J Immunol.* 2004; 172(5):2731–2738. [PubMed: 14978070]
77. Stevens JC, Banks GT, Festing MF, Fisher EM. Quiet mutations in inbred strains of mice. *Trends Mol Med.* 2007; 13(12):512–519. [PubMed: 17981508]

78. Taft RA, Davisson M, Wiles MV. Know thy mouse. *Trends Genet.* 2006; 22(12):649–653. [PubMed: 17007958]
79. Champlin R, Ho W, Gajewski J, et al. Selective depletion of CD8⁺ T lymphocytes for prevention of graft-versus-host disease after allogeneic bone marrow transplantation. *Blood.* 1990; 76(2):418–423. [PubMed: 2142440]
80. Truitt RL, Atasoylu AA. Contribution of CD4⁺ and CD8⁺ T cells to graft-versus-host disease and graft-versus-leukemia reactivity after transplantation of MHC-compatible bone marrow. *Bone Marrow Transplant.* 1991; 8(1):51–58. [PubMed: 1833016]
81. Dazzi F, Szydlo RM, Goldman JM. Donor lymphocyte infusions for relapse of chronic myeloid leukemia after allogeneic stem cell transplant: where we now stand. *Exp Hematol.* 1999; 27(10):1477–1486. [PubMed: 10517488]
82. Martin PJ, Rowley SD, Anasetti C, et al. A Phase I–II clinical trial to evaluate removal of CD4 cells and partial depletion of CD8 cells from donor marrow for HLA-mismatched unrelated recipients. *Blood.* 1999; 94(7):2192–2199. [PubMed: 10498588]
83. Rosenberg SA, Packard BS, Aebersold PM, et al. Use of tumor-infiltrating lymphocytes and interleukin-2 in the immunotherapy of patients with metastatic melanoma. A preliminary report. *N Engl J Med.* 1988; 319(25):1676–1680. [PubMed: 3264384]
84. Rosenberg SA, Yang JC, Topalian SL, et al. Treatment of 283 consecutive patients with metastatic melanoma or renal cell cancer using high-dose bolus interleukin 2. *JAMA.* 1994; 271(12):907–913. [PubMed: 8120958]
85. Rosenberg SA, Yannelli JR, Yang JC, et al. Treatment of patients with metastatic melanoma with autologous tumor-infiltrating lymphocytes and interleukin 2. *J Natl Cancer Inst.* 1994; 86(15):1159–1166. [PubMed: 8028037]
86. Rosenberg SA, Aebersold P, Cornetta K, et al. Gene transfer into humans – immunotherapy of patients with advanced melanoma, using tumor-infiltrating lymphocytes modified by retroviral gene transduction. *N Engl J Med.* 1990; 323(9):570–578. [PubMed: 2381442]
87. Dudley ME, Yang JC, Sherry R, et al. Adoptive cell therapy for patients with metastatic melanoma: evaluation of intensive myeloablative chemoradiation preparative regimens. *J Clin Oncol.* 2008; 26(32):5233–5239. [PubMed: 18809613]
88. Rosenberg SA, Dudley ME. Adoptive cell therapy for the treatment of patients with metastatic melanoma. *Curr Opin Immunol.* 2009; 21(2):233–240. [PubMed: 19304471]
89. Morgan RA, Dudley ME, Wunderlich JR, et al. Cancer regression in patients after transfer of genetically engineered lymphocytes. *Science.* 2006; 314(5796):126–129. The efficacy of adoptive transfer of lymphocytes after lymphodepletion is limited by the unspecificity of these cells to home properly and recognize residual tumor. This study demonstrates that *ex vivo* genetic manipulation of peripheral blood lymphocytes in order to express the specific T-cell receptor for melanoma-associated antigens and their reinfusion back into the patient can induce antitumor effects. Also demonstrates that adoptive immunotherapy with hematopoietic stem cell transplant (HSCT) may be a superior means to increase donor T-cell efficacy. [PubMed: 16946036]
90. Gattinoni L, Finkelstein SE, Klebanoff CA, et al. Removal of homeostatic cytokine sinks by lymphodepletion enhances the efficacy of adoptively transferred tumor-specific CD8⁺ T cells. *J Exp Med.* 2005; 202(7):907–912. [PubMed: 16203864]
91. Overwijk WW, Theoret MR, Finkelstein SE, et al. Tumor regression and autoimmunity after reversal of a functionally tolerant state of self-reactive CD8⁺ T cells. *J Exp Med.* 2003; 198(4):569–580. [PubMed: 12925674]
92. Rosenberg SA, Restifo NP, Yang JC, Morgan RA, Dudley ME. Adoptive cell transfer: a clinical path to effective cancer immunotherapy. *Nat Rev Cancer.* 2008; 8(4):299–308. [PubMed: 18354418]
93. Wrzesinski C, Restifo NP. Less is more: lymphodepletion followed by hematopoietic stem cell transplant augments adoptive T-cell-based anti-tumor immunotherapy. *Curr Opin Immunol.* 2005; 17(2):195–201. [PubMed: 15766681]
94. Berry LJ, Moeller M, Darcy PK. Adoptive immunotherapy for cancer: the next generation of gene-engineered immune cells. *Tissue Antigens.* 2009; 74(4):277–289. [PubMed: 19775368]

95. Sadelain M, Brentjens R, Riviere I. The promise and potential pitfalls of chimeric antigen receptors. *Curr Opin Immunol.* 2009; 21(2):215–223. [PubMed: 19327974]
96. Wrzesinski C, Paulos CM, Gattinoni L, et al. Hematopoietic stem cells promote the expansion and function of adoptively transferred antitumor CD8 T cells. *J Clin Invest.* 2007; 117(2):492–501. Demonstrates that cytoreductive conditioning of the recipient does much more than simply create space and allow transferred T cells to expand by homeostatic proliferation. Activation of dendritic cells via Toll-like receptors appears to play a critical role in allowing for greater survival and function of the transferred T cells. [PubMed: 17273561]
97. Cooke KR, Hill GR, Crawford JM, et al. Tumor necrosis factor- α production to lipopolysaccharide stimulation by donor cells predicts the severity of experimental acute graft-versus-host disease. *J Clin Invest.* 1998; 102(10):1882–1891. [PubMed: 9819375]
98. Hill GR, Ferrara JL. The primacy of the gastrointestinal tract as a target organ of acute graft-versus-host disease: rationale for the use of cytokine shields in allogeneic bone marrow transplantation. *Blood.* 2000; 95(9):2754–2759. [PubMed: 10779417]
99. Liang H, Liao C, Qi Z, et al. Rapamycin or tacrolimus alone fails to resist cardiac allograft accelerated rejection mediated by alloreactive CD4⁺ memory T cells in mice. *Transpl Immunol.* 2010; 22(3–4):128–136. [PubMed: 19755159]
100. Bingaman AW, Farber DL. Memory T cells in transplantation: generation, function, and potential role in rejection. *Am J Transplant.* 2004; 4(6):846–852. [PubMed: 15147417]
101. Verneris MR, Karami M, Baker J, Jayaswal A, Negrin RS. Role of NKG2D signaling in the cytotoxicity of activated and expanded CD8⁺ T cells. *Blood.* 2004; 103(8):3065–3072. Demonstrates that activated CD8⁺ T cells can become ‘natural killer (NK)-like’ with regard to effector function, and so kill targets via NKG2D. This would allow the use of activated T cells in HSCT, which would result in greater graft-versus-tumor (GVT) without GVHD. [PubMed: 15070686]
102. Kim HM, Lim J, Kang JS, et al. Inhibition of human cervical carcinoma growth by cytokine-induced killer cells in nude mouse xenograft model. *Int Immunopharmacol.* 2009; 9(3):375–380. [PubMed: 19105981]
103. Nishimura R, Baker J, Beilhack A, et al. *In vivo* trafficking and survival of cytokine-induced killer cells resulting in minimal GVHD with retention of antitumor activity. *Blood.* 2008; 112(6):2563–2574. [PubMed: 18565854]
104. Ljunggren HG, Karre K. In search of the ‘missing self’: MHC molecules and NK cell recognition. *Immunol Today.* 1990; 11(7):237–244. [PubMed: 2201309]
105. Thompson JA, Shulman KL, Benyunes MC, et al. Prolonged continuous intravenous infusion interleukin-2 and lymphokine-activated killer-cell therapy for metastatic renal cell carcinoma. *J Clin Oncol.* 1992; 10(6):960–968. [PubMed: 1588376]
106. Trinchieri G. Biology of natural killer cells. *Adv Immunol.* 1989:47187–47376.
107. Koh CY, Welniak LA, Murphy WJ. Adoptive cellular immunotherapy: NK cells and bone marrow transplantation. *Histol Histopathol.* 2000; 15(4):1201–1210. [PubMed: 11005245]
108. Uhrberg M, Valiante NM, Shum BP, et al. Human diversity in killer cell inhibitory receptor genes. *Immunity.* 1997; 7(6):753–763. [PubMed: 9430221]
109. Lanier LL. NK cell recognition. *Annu Rev Immunol.* 2005; 23:225–274. [PubMed: 15771571]
110. Bennett M. Rejection of marrow allografts: importance of H-2 homozygosity of donor cells. *Transplantation.* 1972; 14(3):289–298. [PubMed: 4404692]
111. Cudkowicz G, Bennett M. Peculiar immunobiology of bone marrow allografts. II. Rejection of parental grafts by resistant F1 hybrid mice. *J Exp Med.* 1971; 134(6):1513–1528. [PubMed: 4942407]
112. Yu YY, Kumar V, Bennett M. Murine natural killer cells and marrow graft rejection. *Annu Rev Immunol.* 1992; 10:189–213. [PubMed: 1590985]
113. Cudkowicz G, Bennett M. Peculiar immunobiology of bone marrow allografts. I. Graft rejection by irradiated responder mice. *J Exp Med.* 1971; 134(1):83–102. [PubMed: 4397663]
114. Natarajan K, Dimasi N, Wang J, Mariuzza RA, Margulies DH. Structure and function of natural killer cell receptors: multiple molecular solutions to self, nonself discrimination. *Annu Rev Immunol.* 2002; 20:853–885. [PubMed: 11861620]

115. Yu YY, George T, Dorfman JR, Roland J, Kumar V, Bennett M. The role of Ly49A and 5E6(Ly49C) molecules in hybrid resistance mediated by murine natural killer cells against normal T cell blasts. *Immunity*. 1996; 4(1):67–76. [PubMed: 8574853]
116. Asai O, Longo DL, Tian ZG, et al. Suppression of graft-versus-host disease and amplification of graft-versus-tumor effects by activated natural killer cells after allogeneic bone marrow transplantation. *J Clin Invest*. 1998; 101(9):1835–1842. [PubMed: 9576746]
117. Ruggeri L, Aversa F, Martelli MF, Velardi A. Allogeneic hematopoietic transplantation and natural killer cell recognition of missing self. *Immunol Rev*. 2006; 214:202–218. [PubMed: 17100886]
118. Ruggeri L, Capanni M, Urbani E, et al. Effectiveness of donor natural killer cell alloreactivity in mismatched hematopoietic transplants. *Science*. 2002; 295(5562):2097–2100. Adoptive transfer of alloreactive T cells not only is important for a GVT effect but can also induce GVHD. Infusion of alloreactive NK cells with killer cell immunoglobulin-like receptor ligand mismatch results in GVHD protection and eradication of leukemia in humans, as well as GVHD blockade in mice that received nonmyeloablative conditioning. The results suggest that donor-derived NK cells may be of potential clinical use in HSCT. [PubMed: 11896281]
119. Passweg JR, Tichelli A, Meyer-Monard S, et al. Purified donor NK-lymphocyte infusion to consolidate engraftment after haploidentical stem cell transplantation. *Leukemia*. 2004; 18(11):1835–1838. [PubMed: 15457184]
120. Koh CY, Raziuddin A, Welniak LA, Blazar BR, Bennett M, Murphy WJ. NK inhibitory-receptor blockade for purging of leukemia: effects on hematopoietic reconstitution. *Biol Blood Marrow Transplant*. 2002; 8(1):17–25. [PubMed: 11846352]
121. Ljunggren HG, Malmberg KJ. Prospects for the use of NK cells in immunotherapy of human cancer. *Nat Rev Immunol*. 2007; 7(5):329–339. [PubMed: 17438573]
122. Romagne F, Andre P, Spee P, et al. Preclinical characterization of 1–7F9, a novel human anti-KIR receptor therapeutic antibody that augments natural killer-mediated killing of tumor cells. *Blood*. 2009; 114(13):2667–2677. [PubMed: 19553639]
123. Sola C, Andre P, Lemmers C, et al. Genetic and antibody-mediated reprogramming of natural killer cell missing-self recognition *in vivo*. *Proc Natl Acad Sci USA*. 2009; 106(31):12879–12884. [PubMed: 19561305]
124. Shevach EM, Thornton A, Suri-Payer E. T lymphocyte-mediated control of autoimmunity. *Novartis Found Symp*. 1998; 215:200–211. discussion 211–230. [PubMed: 9760581]
125. Battaglia M, Roncarolo MG. Induction of transplantation tolerance via regulatory T cells. *Inflamm Allergy Drug Targets*. 2006; 5(3):157–165. [PubMed: 16918479]
126. Nguyen VH, Zeiser R, Negrin RS. Role of naturally arising regulatory T cells in hematopoietic cell transplantation. *Biol Blood Marrow Transplant*. 2006; 12(10):995–1009. [PubMed: 17084366]
127. Yong Z, Chang L, Mei YX, Yi L. Role and mechanisms of CD4⁺CD25⁺ regulatory T cells in the induction and maintenance of transplantation tolerance. *Transpl Immunol*. 2007; 17(2):120–129. [PubMed: 17306743]
128. Sakaguchi S, Sakaguchi N, Asano M, Itoh M, Toda M. Immunologic self-tolerance maintained by activated T cells expressing IL-2 receptor α -chains (CD25). Breakdown of a single mechanism of self-tolerance causes various autoimmune diseases. *J Immunol*. 1995; 155(3):1151–1164. [PubMed: 7636184]
129. Graca L, Cobbold SP, Waldmann H. Identification of regulatory T cells in tolerated allografts. *J Exp Med*. 2002; 195(12):1641–1646. [PubMed: 12070291]
130. Gregori S, Casorati M, Amuchastegui S, Smirardo S, Davalli AM, Adorini L. Regulatory T cells induced by 1 α ,25-dihydroxyvitamin D3 and mycophenolate mofetil treatment mediate transplantation tolerance. *J Immunol*. 2001; 167(4):1945–1953. [PubMed: 11489974]
131. Hara M, Kingsley CI, Niimi M, et al. IL-10 is required for regulatory T cells to mediate tolerance to alloantigens *in vivo*. *J Immunol*. 2001; 166(6):3789–3796. [PubMed: 11238621]
132. Hoffmann P, Ermann J, Edinger M, Fathman CG, Strober S. Donor-type CD4⁺CD25⁺ regulatory T cells suppress lethal acute graft-versus-host disease after allogeneic bone marrow transplantation. *J Exp Med*. 2002; 196(3):389–399. [PubMed: 12163567]

133. Blazar BR, Taylor PA, Noelle RJ, Vallera DA. CD4⁺ T cells tolerized *ex vivo* to host alloantigen by anti-CD40 ligand (CD40L:CD154) antibody lose their graft-versus-host disease lethality capacity but retain nominal antigen responses. *J Clin Invest.* 1998; 102(3):473–482. [PubMed: 9691083]
134. Godfrey WR, Spoden DJ, Ge YG, et al. Cord blood CD4⁺CD25⁺-derived T regulatory cell lines express FoxP3 protein and manifest potent suppressor function. *Blood.* 2005; 105(2):750–758. [PubMed: 15374887]
135. Edinger M, Hoffmann P, Ermann J, et al. CD4⁺CD25⁺ regulatory T cells preserve graft-versus-tumor activity while inhibiting graft-versus-host disease after bone marrow transplantation. *Nat Med.* 2003; 9(9):1144–1150. Adoptive transfer of donor-derived regulatory T cells (CD4⁺CD25⁺) after bone marrow transplant (BMT) in leukemia- and lymphoma-bearing mice induces suppression of GVHD without losing GVT effect. These studies demonstrate that regulatory T cells can suppress GVHD without compromising GVT, suggesting that the kinetics of GVHD and GVT are indeed different. [PubMed: 12925844]
136. Jones SC, Murphy GF, Korngold R. Post-hematopoietic cell transplantation control of graft-versus-host disease by donor CD425 T cells to allow an effective graft-versus-leukemia response. *Biol Blood Marrow Transplant.* 2003; 9(4):243–256. [PubMed: 12720217]
137. Trenado A, Charlotte F, Fisson S, et al. Recipient-type specific CD4⁺CD25⁺ regulatory T cells favor immune reconstitution and control graft-versus-host disease while maintaining graft-versus-leukemia. *J Clin Invest.* 2003; 112(11):1688–1696. [PubMed: 14660744]
138. Hill GR, Crawford JM, Cooke KR, Brinson YS, Pan L, Ferrara JL. Total body irradiation and acute graft-versus-host disease: the role of gastrointestinal damage and inflammatory cytokines. *Blood.* 1997; 90(8):3204–3213. [PubMed: 9376604]
139. Hill GR, Teshima T, Gerbitz A, et al. Differential roles of IL-1 and TNF- α on graft-versus-host disease and graft versus leukemia. *J Clin Invest.* 1999; 104(4):459–467. [PubMed: 10449438]
140. van Bekkum DW, Roodenburg J, Heidt PJ, van der Waaij D. Mitigation of secondary disease of allogeneic mouse radiation chimeras by modification of the intestinal microflora. *J Natl Cancer Inst.* 1974; 52(2):401–404. [PubMed: 4150164]
141. Teshima T, Ordemann R, Reddy P, et al. Acute graft-versus-host disease does not require alloantigen expression on host epithelium. *Nat Med.* 2002; 8(6):575–581. [PubMed: 12042807]
142. Duffner UA, Maeda Y, Cooke KR, et al. Host dendritic cells alone are sufficient to initiate acute graft-versus-host disease. *J Immunol.* 2004; 172(12):7393–7398. [PubMed: 15187116]
143. Shlomchik WD, Couzens MS, Tang CB, et al. Prevention of graft versus host disease by inactivation of host antigen-presenting cells. *Science.* 1999; 285(5426):412–415. [PubMed: 10411505]
144. Reddy V, Iturraspe JA, Tzolas AC, Meier-Kriesche HU, Schold J, Wingard JR. Low dendritic cell count after allogeneic hematopoietic stem cell transplantation predicts relapse, death, and acute graft-versus-host disease. *Blood.* 2004; 103(11):4330–4335. [PubMed: 14962904]
145. Auffermann-Gretzinger S, Lossos IS, Vayntrub TA, et al. Rapid establishment of dendritic cell chimerism in allogeneic hematopoietic cell transplant recipients. *Blood.* 2002; 99(4):1442–1448. [PubMed: 11830498]
146. Chakraverty R, Sykes M. The role of antigen-presenting cells in triggering graft-versus-host disease and graft-versus-leukemia. *Blood.* 2007; 110(1):9–17. [PubMed: 17327406]
147. Edinger M, Powrie F, Chakraverty R. Regulatory mechanisms in graft-versus-host responses. *Biol Blood Marrow Transplant.* 2008; 15(Suppl 1):2–6. [PubMed: 19147068]
148. Matte CC, Liu J, Cormier J, et al. Donor APCs are required for maximal GVHD but not for GVL. *Nat Med.* 2004; 10(9):987–992. [PubMed: 15286785]
149. Anderson BE, McNiff JM, Jain D, Blazar BR, Shlomchik WD, Shlomchik MJ. Distinct roles for donor- and host-derived antigen-presenting cells and costimulatory molecules in murine chronic graft-versus-host disease: requirements depend on target organ. *Blood.* 2005; 105(5):2227–2234. [PubMed: 15522961]
150. Pahl HL. Activators and target genes of Rel/NF- κ B transcription factors. *Oncogene.* 1999; 18(49):6853–6866. [PubMed: 10602461]

151. Siebenlist U, Franzoso G, Brown K. Structure, regulation and function of NF-kappa B. *Annu Rev Cell Biol.* 1994;10:405–10455.
152. Dejardin E. The alternative NF-kappaB pathway from biochemistry to biology: pitfalls and promises for future drug development. *Biochem Pharmacol.* 2006; 72(9):1161–1179. [PubMed: 16970925]
153. Richardson PG, Mitsiades C, Hideshima T, Anderson KC. Bortezomib: proteasome inhibition as an effective anticancer therapy. *Annu Rev Med.* 2006; 57:33–47. [PubMed: 16409135]
154. Markovina S, Callander NS, O'Connor SL, et al. Bortezomib-resistant nuclear factor- κ B activity in multiple myeloma cells. *Mol Cancer Res.* 2008; 6(8):1356–1364. [PubMed: 18708367]
155. Kane RC, Bross PF, Farrell AT, Pazdur R. Velcade: US FDA approval for the treatment of multiple myeloma progressing on prior therapy. *Oncologist.* 2003; 8(6):508–513. [PubMed: 14657528]
156. Sun K, Welniak LA, Panoskaltis-Mortari A, et al. Inhibition of acute graft-versus-host disease with retention of graft-versus-tumor effects by the proteasome inhibitor bortezomib. *Proc Natl Acad Sci USA.* 2004; 101(21):8120–8125. Administration of bortezomib postallogeic BMT in leukemia-bearing mice not only prevents GVHD but is also associated with greater survival. These studies demonstrate that molecular targeting agents, such as proteasome inhibition, can result in immunomodulatory effects outside of the direct antitumor effects by the agent. [PubMed: 15148407]
157. Sun K, Li M, Sayers TJ, Welniak LA, Murphy WJ. Differential effects of donor T-cell cytokines on outcome with continuous bortezomib administration after allogeneic bone marrow transplantation. *Blood.* 2008; 112(4):1522–1529. [PubMed: 18539902]
158. Sayers TJ, Brooks AD, Koh CY, et al. The proteasome inhibitor PS-341 sensitizes neoplastic cells to TRAIL-mediated apoptosis by reducing levels of c-FLIP. *Blood.* 2003; 102(1):303–310. [PubMed: 12637321]
159. Koreth J, Stevenson KE, Kim HT, et al. Bortezomib, tacrolimus, and methotrexate for prophylaxis of graft-versus-host disease after reduced-intensity conditioning allogeneic stem cell transplantation from HLA-mismatched unrelated donors. *Blood.* 2009; 114(18):3956–3959. [PubMed: 19713456]
160. Lonial S, Waller EK, Richardson PG, et al. Risk factors and kinetics of thrombocytopenia associated with bortezomib for relapsed, refractory multiple myeloma. *Blood.* 2005; 106(12):3777–3784. [PubMed: 16099887]
161. Reddy P, Maeda Y, Hotary K, et al. Histone deacetylase inhibitor suberoylanilide hydroxamic acid reduces acute graft-versus-host disease and preserves graft-versus-leukemia effect. *Proc Natl Acad Sci USA.* 2004; 101(11):3921–3926. Administration of suberoylanilide hydroxamic acid post-BMT decreases the secretion of proinflammatory cytokines, therefore minimizing GVHD pathology without abrogating the efficacy of T-cell responses and GVT effect. [PubMed: 15001702]
162. Marks P, Rifkind RA, Richon VM, Breslow R, Miller T, Kelly WK. Histone deacetylases and cancer: causes and therapies. *Nat Rev Cancer.* 2001; 1(3):194–202. [PubMed: 11902574]
163. Butler LM, Agus DB, Scher HI, et al. Suberoylanilide hydroxamic acid, an inhibitor of histone deacetylase, suppresses the growth of prostate cancer cells *in vitro* and *in vivo*. *Cancer Res.* 2000; 60(18):5165–5170. [PubMed: 11016644]
164. Cohen LA, Marks PA, Rifkind RA, et al. Suberoylanilide hydroxamic acid (SAHA), a histone deacetylase inhibitor, suppresses the growth of carcinogen-induced mammary tumors. *Anti Cancer Res.* 2002; 22(3):1497–1504.
165. Leoni F, Zaliani A, Bertolini G, et al. The antitumor histone deacetylase inhibitor suberoylanilide hydroxamic acid exhibits antiinflammatory properties via suppression of cytokines. *Proc Natl Acad Sci USA.* 2002; 99(5):2995–3000. [PubMed: 11867742]
166. Kelly WK, Richon VM, O'Connor O, et al. Phase I clinical trial of histone deacetylase inhibitor: suberoylanilide hydroxamic acid administered intravenously. *Clin Cancer Res.* 2003; 9(10 Pt 1):3578–3588. [PubMed: 14506144]

167. Linsley PS, Greene JL, Brady W, Bajorath J, Ledbetter JA, Peach R. Human B7-1 (CD80) and B7-2 (CD86) bind with similar avidities but distinct kinetics to CD28 and CTLA-4 receptors. *Immunity*. 1994; 1(9):793–801. [PubMed: 7534620]
168. Linsley PS, Greene JL, Tan P, et al. Coexpression and functional cooperation of CTLA-4 and CD28 on activated T lymphocytes. *J Exp Med*. 1992; 176(6):1595–1604. [PubMed: 1334116]
169. van der Merwe PA, Bodian DL, Daenke S, Linsley P, Davis SJ. CD80 (B7-1) binds both CD28 and CTLA-4 with a low affinity and very fast kinetics. *J Exp Med*. 1997; 185(3):393–403. [PubMed: 9053440]
170. Walunas TL, Lenschow DJ, Bakker CY, et al. CTLA-4 can function as a negative regulator of T cell activation. *Immunity*. 1994; 1(5):405–413. [PubMed: 7882171]
171. Carreno BM, Bennett F, Chau TA, et al. CTLA-4 (CD152) can inhibit T cell activation by two different mechanisms depending on its level of cell surface expression. *J Immunol*. 2000; 165(3):1352–1356. [PubMed: 10903737]
172. Krummel MF, Allison JP. CD28 and CTLA-4 have opposing effects on the response of T cells to stimulation. *J Exp Med*. 1995; 182(2):459–465. [PubMed: 7543139]
173. Blazar BR, Taylor PA, Linsley PS, Vallera DA. *In vivo* blockade of CD28/CTLA4: B7/BB1 interaction with CTLA4-Ig reduces lethal murine graft-versus-host disease across the major histocompatibility complex barrier in mice. *Blood*. 1994; 83(12):3815–3825. [PubMed: 7515723]
174. Blazar BR, Taylor PA, Panoskaltsis-Mortari A, Sharpe AH, Vallera DA. Opposing roles of CD28:B7 and CTLA-4:B7 pathways in regulating *in vivo* alloresponses in murine recipients of MHC disparate T cells. *J Immunol*. 1999; 162(11):6368–6377. [PubMed: 10352249]
175. Fife BT, Bluestone JA. Control of peripheral T-cell tolerance and autoimmunity via the CTLA-4 and PD-1 pathways. *Immunol Rev*. 2008;224:166–182.
176. Greenwald RJ, Freeman GJ, Sharpe AH. The B7 family revisited. *Annu Rev Immunol*. 2005;23:515–23548.
177. Alderson KL, Zhou Q, Berner V, et al. Regulatory and conventional CD4⁺ T cells show differential effects correlating with PD-1 and B7-H1 expression after immunotherapy. *J Immunol*. 2008; 180(5):2981–2988. [PubMed: 18292520]
178. Ozkaynak E, Wang L, Goodearl A, et al. Programmed death-1 targeting can promote allograft survival. *J Immunol*. 2002; 169(11):6546–6553. [PubMed: 12444166]
179. Blazar BR, Carreno BM, Panoskaltsis-Mortari A, et al. Blockade of programmed death-1 engagement accelerates graft-versus-host disease lethality by an IFN- γ -dependent mechanism. *J Immunol*. 2003; 171(3):1272–1277. [PubMed: 12874215]
180. Blazar BR, Sharpe AH, Chen AI, et al. Ligation of OX40 (CD134) regulates graft-versus-host disease (GVHD) and graft rejection in allogeneic bone marrow transplant recipients. *Blood*. 2003; 101(9):3741–3748. [PubMed: 12521997]
181. Jung U, Foley JE, Erdmann AA, et al. *Ex vivo* rapamycin generates Th1/Tc1 or Th2/Tc2 Effector T cells with enhanced *in vivo* function and differential sensitivity to post-transplant rapamycin therapy. *Biol Blood Marrow Transplant*. 2006; 12(9):905–918. [PubMed: 16920556]
182. Foley JE, Jung U, Miera A, et al. *Ex vivo* rapamycin generates donor Th2 cells that potently inhibit graft-versus-host disease and graft-versus-tumor effects via an IL-4-dependent mechanism. *J Immunol*. 2005; 175(9):5732–5743. [PubMed: 16237064]
183. Kappel LW, Goldberg GL, King CG, et al. IL-17 contributes to CD4-mediated graft-versus-host disease. *Blood*. 2009; 113(4):945–952. [PubMed: 18931341]
184. Yi T, Zhao D, Lin CL, et al. Absence of donor Th17 leads to augmented Th1 differentiation and exacerbated acute graft-versus-host disease. *Blood*. 2008; 112(5):2101–2110. [PubMed: 18596226]
185. Couriel D, Saliba R, Hicks K, et al. Tumor necrosis factor- α blockade for the treatment of acute GVHD. *Blood*. 2004; 104(3):649–654. [PubMed: 15069017]
186. Chiang KY, Abhyankar S, Bridges K, Godder K, Henslee-Downey JP. Recombinant human tumor necrosis factor receptor fusion protein as complementary treatment for chronic graft-versus-host disease. *Transplantation*. 2002; 73(4):665–667. [PubMed: 11889452]

187. Przepiorka D, Kernan NA, Ippoliti C, et al. Daclizumab, a humanized anti-interleukin-2 receptor alpha chain antibody, for treatment of acute graft-versus-host disease. *Blood*. 2000; 95(1):83–89. [PubMed: 10607689]
188. Wolff D, Roessler V, Steiner B, et al. Treatment of steroid-resistant acute graft-versus-host disease with daclizumab and etanercept. *Bone Marrow Transplant*. 2005; 35(10):1003–1010. [PubMed: 15806135]
189. Yanik G, Hellerstedt B, Custer J, et al. Etanercept (Enbrel) administration for idiopathic pneumonia syndrome after allogeneic hematopoietic stem cell transplantation. *Biol Blood Marrow Transplant*. 2002; 8(7):395–400. [PubMed: 12171486]
190. 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(1):42–48. [PubMed: 11823036]
191. 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(1):11–20. [PubMed: 12542793]
192. Tian Y, Deng YB, Huang YJ, Wang Y. Bone marrow-derived mesenchymal stem cells decrease acute graft-versus-host disease after allogeneic hematopoietic stem cells transplantation. *Immunol Invest*. 2008; 37(1):29–42. [PubMed: 18214798]
193. Barrett AJ, Le Blanc K. Prophylaxis of acute GVHD: manipulate the graft or the environment? *Best Pract Res Clin Haematol*. 2008; 21(2):165–176. [PubMed: 18503984]
194. Le Blanc K, Frasson F, Ball L, et al. Mesenchymal stem cells for treatment of steroid-resistant, severe, acute graft-versus-host disease: a Phase II study. *Lancet*. 2008; 371(9624):1579–1586. [PubMed: 18468541]
195. 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(3):593–599. [PubMed: 18185520]
196. Karnoub AE, Dash AB, Vo AP, et al. Mesenchymal stem cells within tumour stroma promote breast cancer metastasis. *Nature*. 2007; 449(7162):557–563. [PubMed: 17914389]
197. Gesundheit B, Shapira MY, Resnick IB, et al. Successful cell-mediated cytokine-activated immunotherapy for relapsed acute myeloid leukemia after hematopoietic stem cell transplantation. *Am J Hematol*. 2009; 84(3):188–190. [PubMed: 19105234]
198. Huang XJ, Liu DH, Liu KY, et al. Treatment of acute leukemia with unmanipulated HLA-mismatched/ haploidentical blood and bone marrow transplantation. *Biol Blood Marrow Transplant*. 2009; 15(2):257–265. [PubMed: 19167686]
199. Kebriaei P, Detry MA, Giralt S, et al. Long-term follow-up of allogeneic hematopoietic stem-cell transplantation with reduced-intensity conditioning for patients with chronic myeloid leukemia. *Blood*. 2007; 110(9):3456–3462. [PubMed: 17652620]
200. Poire X, Artz A, Larson RA, et al. Allogeneic stem cell transplantation with alemtuzumab-based conditioning for patients with advanced chronic myelogenous leukemia. *Leuk Lymphoma*. 2009; 50(1):85–91. [PubMed: 19142796]
201. Wrench D, Gribben JG. Stem cell transplantation for non-Hodgkin's lymphoma. *Hematol Oncol Clin North Am*. 2008; 22(5):1051–1079. XI. [PubMed: 18954751]
202. Boyiadzis M, Foon KA, Pavletic S. Hematopoietic stem cell transplantation for chronic lymphocytic leukemia: potential cure for an incurable disease. *Expert Opin Biol Ther*. 2007; 7(12):1789–1797. [PubMed: 18034645]
203. Gribben JG. Stem cell transplantation in chronic lymphocytic leukemia. *Biol Blood Marrow Transplant*. 2008; 15(Suppl 1):53–58. [PubMed: 19147079]
204. Laport GG, Sandmaier BM, Storer BE, et al. Reduced-intensity conditioning followed by allogeneic hematopoietic cell transplantation for adult patients with myelodysplastic syndrome and myeloproliferative disorders. *Biol Blood Marrow Transplant*. 2008; 14(2):246–255. [PubMed: 18215785]
205. Warlick ED, Cioc A, Defor T, Dolan M, Weisdorf D. Allogeneic stem cell transplantation for adults with myelodysplastic syndromes: importance of pretransplant disease burden. *Biol Blood Marrow Transplant*. 2009; 15(1):30–38. [PubMed: 19135940]

206. Armand P, Antin JH. Allogeneic stem cell transplantation for aplastic anemia. *Biol Blood Marrow Transplant.* 2007; 13(5):505–516. [PubMed: 17448909]
207. Kim HJ, Park CY, Park YH, et al. Successful allogeneic hematopoietic stem cell transplantation using triple agent immunosuppression in severe aplastic anemia patients. *Bone Marrow Transplant.* 2003; 31(2):79–86. [PubMed: 12621487]
208. Min CK, Kim DW, Lee JW, Han CW, Min WS, Kim CC. Hematopoietic stem cell transplantation for high-risk adult patients with severe aplastic anemia; reduction of graft failure by enhancing stem cell dose. *Haematologica.* 2001; 86(3):303–310. [PubMed: 11255278]
209. Bourquin JP, Thornley I, Neuberg D, et al. Favorable outcome of allogeneic hematopoietic stem cell transplantation for relapsed or refractory acute promyelocytic leukemia in childhood. *Bone Marrow Transplant.* 2004; 34(9):795–798. [PubMed: 15354207]
210. Friedrich W, Muller SM. Allogeneic stem cell transplantation for treatment of immunodeficiency. *Springer Semin Immunopathol.* 2004; 26(1–2):109–118. [PubMed: 15368077]
211. Gennery AR, Cant AJ. Advances in hematopoietic stem cell transplantation for primary immunodeficiency. *Immunol Allergy Clin North Am.* 2008; 28(2):439–456. X–XI. [PubMed: 18424341]
212. Gratwohl A. Allogeneic hematopoietic stem cell transplantation for severe autoimmune diseases. *Autoimmunity.* 2008; 41(8):673–678. [PubMed: 18958760]
213. Nash RA. Allogeneic HSCT for autoimmune diseases: conventional conditioning regimens. *Bone Marrow Transplant.* 2003; 32(Suppl 1):S77–S80. [PubMed: 12931249]
214. Forrest DL, Thompson K, Nevill TJ, Couban S, Fernandez LA. Allogeneic hematopoietic stem cell transplantation for progressive follicular lymphoma. *Bone Marrow Transplant.* 2002; 29(12):973–978. [PubMed: 12098065]
215. Childs RW, Clave E, Tisdale J, Plante M, Hensel N, Barrett J. Successful treatment of metastatic renal cell carcinoma with a nonmyeloablative allogeneic peripheral-blood progenitor-cell transplant: evidence for a graft-versus-tumor effect. *J Clin Oncol.* 1999; 17(7):2044–2049. [PubMed: 10561256]
216. Takahashi Y, Childs RW. Nonmyeloablative transplantation: an allogeneic-based immunotherapy for renal cell carcinoma. *Clin Cancer Res.* 2004; 10(18 Pt 2):S6353–S6359.
217. Nakashima Y, Shiratsuchi M, Abe Y, et al. Sustained molecular remission by non-myeloablative stem cell transplantation after autologous hematopoietic stem cell transplantation in a patient with multiple myeloma. *Leuk Lymphoma.* 2005; 46(8):1217–1222. [PubMed: 16085565]
218. Zeiser R, Bertz H, Spyridonidis A, Houet L, Finke J. Donor lymphocyte infusions for multiple myeloma: clinical results and novel perspectives. *Bone Marrow Transplant.* 2004; 34(11):923–928. [PubMed: 15361911]
219. Claviez A, Sureda A, Schmitz N. Haematopoietic SCT for children and adolescents with relapsed and refractory Hodgkin's lymphoma. *Bone Marrow Transplant.* 2008; 42(Suppl 2):S16–S24. [PubMed: 18978738]
220. Vose JM, Sharp G, Chan WC, et al. Autologous transplantation for aggressive non-Hodgkin's lymphoma: results of a randomized trial evaluating graft source and minimal residual disease. *J Clin Oncol.* 2002; 20(9):2344–2352. [PubMed: 11981006]
221. Drabko K, Choma M, Zaucha-Prazmo A, et al. Megachemotherapy and autologous hematopoietic stem cell transplantation in children with solid tumours excluding neuroblastoma – experience of Polish paediatric centres. *Med Wieku Rozwoj.* 2006; 10(3 Pt 1):785–792. [PubMed: 17317909]
222. Manilay JO, Sykes M. Natural killer cells and their role in graft rejection. *Curr Opin Immunol.* 1998; 10(5):532–538. [PubMed: 9794835]

Figure 1. Pathophysiology of graft-versus-host disease and graft-versus-tumor

–: Inhibition; +: Promotion; CTL: Cytotoxic T lymphocyte; DC: Dendritic cell; GVHD: Graft-versus-host disease; GVT: Graft-versus-tumor; iDC: Immature dendritic cell; NK: Natural killer; PMN: Peripheral mononuclear phagocyte; TCR: T-cell receptor; TRAIL: TNF-related apoptosis-inducing ligand.

Table 1

Common diseases treated with hematopoietic stem cell transplantation

Type of HSCT	Disease	Ref.
Allogeneic	Acute myeloid leukemia	[197,198]
	Chronic myeloid leukemia	[199,200]
	Non-Hodgkin's lymphoma	[201]
	Chronic lymphocytic leukemia	[202,203]
	Myelodysplastic syndrome	[204,205]
	Severe aplastic leukemia	[206–208]
	Refractory leukemia	[209]
	Severe immunodeficiency syndromes	[210,211]
	Autoimmune disease	[212,213]
	Progressive large-cell lymphoma	[214]
	Renal cell carcinoma	[215,216]
Autologous	Multiple myeloma	[217,218]
	Progressive Hodgkin's disease	[219]
	Large-cell lymphoma	[220]
	Relapsed germ-cell tumor	[221]

HSCT: Hematopoietic stem cell transplantation.

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Table 2
Immunotherapeutic strategies for cancer treatment in allogeneic hematopoietic stem cell transplant

Immune therapy	Hematologic disease		Solid cancer		Ref.
	GVHD	GVT	GVHD	GVT	
TCD	Decrease	Decrease			[82]
DLI of CD4 T cells	Decrease/increase	Increase			[79]
DLI of CD8 T cells	Decrease/increase	Increase			[80]
ACT of TILs	?	Increase	Decrease	Increase	[15,83–85,87]
ACT of genetically engineered PBLs	?	Increase	?	Increase	[89,92,93]
NK cells	Decrease	Increase			[120,222]
Tregs	Decrease	Preserved			[134–137]
APC manipulation	Decrease/increase	?			[142,143,146,148,149]
Proteasome Inhibition	Decrease/increase	Increase			[156,157,160]
HDAC inhibitors	Decrease	Increase			[161,166]
Costimulatory pathways	Decrease	Decrease			[179,180]
Cytokines	Decrease/increase	Decrease/increase			[181,182,186,188,189]
MSCs	Decrease	?			[194]

?: Unknown; ACT: Adoptive cell therapy; APC: Antigen-presenting cell; DLI: Donor lymphocyte infusion; GVHD: Graft-versus-host disease; GVT: Graft-versus-tumor; HDAC: Histone deacetylase; HSCT: Hematopoietic stem cell transplant; MSC: Mesenchymal stem cell; NK: Natural killer; PBL: Peripheral blood lymphocyte; TCD: T-cell depletion; TIL: Tumor-infiltrating lymphocyte; Treg: Regulatory T cell.