Alloantigen presentation and graft-versus-host disease: fuel for the fire

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Running title: Antigen presentation during transplantation
Manuscript type: Review article

Abstract Word Count: 200
Text Word Count: 3983
Figure Count: 2
Table count 1
Reference Count: 100

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Abstract

Allogeneic stem cell transplantation (SCT) is a unique procedure, primarily in patients with hematopoietic malignancies, involving chemoradiotherapy followed by the introduction of donor hematopoietic and immune cells into an inflamed and lymphopenic environment. Interruption of the process by which recipient alloantigen is presented to donor T cells to generate graft-versus-host disease (GVHD) represents an attractive therapeutic strategy to prevent morbidity and mortality after SCT and has been increasingly studied in the last 15 years. However, the immune activation resulting in GVHD has no physiological equivalent in nature; alloantigen is ubiquitous, persists indefinitely and can be presented by multiple cell types at numerous sites, often on incompatible MHC and occurs in the context of intense inflammation early after SCT. The recognition that alloantigen presentation is also critical to the development of immunological tolerance via both deletional and regulatory mechanisms further adds to this complexity. Finally, GVHD itself appears capable of inhibiting the presentation of microbiological antigens by donor dendritic cells late after SCT that is mandatory for the establishment of effective pathogen-specific immunity. Here we review our current understanding of alloantigen, its presentation by various antigen presenting cells, subsequent recognition by donor T cells and the potential of therapeutic strategies interrupting this disease-initiating process to modify transplant outcome.
Introduction

Allogeneic hematopoietic stem cell transplantation (SCT) remains an important curative therapy for hematological malignancies. The therapeutic graft-versus-leukemia (GVL) effect is immunological in nature and mediated by donor T and NK cells recognising recipient allogeneic, hematopoietic or leukemia-specific antigens. GVL is thus associated with pathogenic immune responses against non-hematopoietic tissue that manifest as graft-versus-host disease (GVHD), the major procedural limitation. GVHD presents principally as acute and chronic disease which are characterized by tissue apoptosis and fibrosis respectively. The two processes may also present simultaneously in various overlap syndromes late after transplant. Critically, 15-20% of SCT recipients will develop severe GVHD that is refractory to therapy and die. Current prevention and treatment of GVHD rely on the broad suppression of T cells and inflammation with calcineurin inhibitors and corticosteroids respectively that also impact on leukemia and pathogen-specific immunity. New approaches include post transplant cyclophosphamide and alternative immune suppression that include rapamycin. There is now an ever increasing use of alternative (as opposed to MHC matched sibling) donors for SCT. Transplants using these donors, particularly those that are HLA-mismatched such as umbilical cord or haploid (i.e. fully MHC mismatched) increase the risk of life-threatening GVHD and the necessity for intensive therapeutic interventions involving various forms of T cell depletion. Concurrently, the use of reduced-intensity (RIC) and non-ablative conditioning has increased to allow the transplantation of ever older patients and these procedures rely heavily on immunological GVL effects for their therapeutic efficacy.
It is clear that the targets of GVHD are major histocompatibility complexes (MHC) and peptides presented therein (in MHC mismatched donor-recipients) and/or minor histocompatibility antigens (mHA) in both MHC mismatched and MHC matched donor-recipient pairs. There has, however, been less clarity in regard to the nature of alloantigen presentation responsible for the initiation of immune responses in donor T cells that lead to GVHD and in particular the cells and molecular pathways involved. This review outlines recent developments in the field that improve our understanding of alloantigen presentation initiating pathological and protective immunity after SCT, highlighting a complex but highly modifiable process.

**Alloantigen**

When considering allogeneic SCT in which donor and recipient are MHC-matched, the alloantigens that initiate GVHD are defined as minor histocompatibility antigens (mHA). Broadly, these mHA are peptides generated by polymorphic genes that differ between donor and host that can in turn be presented by MHC. Most of the mHA defined to date are presented in HLA-class I, predominantly due to the molecular techniques utilized for their identification. There is increasing recent enthusiasm for identification of hematopoietic-restricted mHA since these antigens are attractive for use in immunotherapy trials to augment GVL and prevent relapse (reviewed in). Interestingly, the continued expression of mHA on non-hematopoietic cells appears to result in alloreactive T cell exhaustion and impaired GVL effects. Thus, the identification of hematopoietic-restricted mHA is highly desirable and a number of such mHA have been identified, including HA-1, HA-2, LRH-1 and ACC-2 (reviewed in). As outlined below, self-peptides (and thus not mHA) may also be seen as foreign by donor T cells when presented in a mismatched MHC.
Recognition of Alloantigen by Donor T cells

The depletion of donor T cells results in a low incidence of acute and chronic GVHD, albeit at the expense of leukemia relapse.\textsuperscript{12,13} Likewise, T cell replete autologous stem cell transplantation is associated with high relapse rates relative to allogeneic SCT, suggesting T cell stimulation by alloantigen is critical for both GVHD and GVL effects.\textsuperscript{14} GVHD can be conceptualized as MHC-class I and/or MHC-class II dependent (i.e. CD8\textsuperscript{+} and CD4\textsuperscript{+} T cell respectively). Mismatches at HLA-class I and II are significant risk factors for severe GVHD and transplant-related mortality\textsuperscript{3}; thus both CD8\textsuperscript{+} T cells and CD4\textsuperscript{+} T cells are involved and indeed the depletion of either T cell subset is insufficient to prevent GVHD.\textsuperscript{15,16} It is important to note that mHA are also presented and recognized within MHC-class I and II and so both CD4 and CD8 T cells are also involved in GVHD after MHC matched SCT. However, the origin of the antigen and the process of presentation differs in the two pathways and consideration of this is critical to the understanding GVHD as a disease process. In considering antigen presentation and T cell recognition it is also important to consider MHC-matched and MHC-mismatched SCT separately.

In MHC-matched SCT, alloantigens presented in MHC-class I are predominantly endogenous in origin (i.e. the antigen is intrinsic to the cell)\textsuperscript{17} and donor T cells recognize, via the T cell receptor, polymorphic recipient peptides presented within a MHC that is shared by both the donor and recipient (Figure 1). In regard to the process of presentation, self or viral cytosolic proteins are processed within the proteosome and then transported into the endoplasmic reticulum (ER) by the transporter associated with antigen processing (TAP). There the ER aminopeptidases (ERAP) trim peptides to 8-10 amino acids for loading into MHC-
class I before transfer to the surface (reviewed in\textsuperscript{18}). Exogenous antigens (i.e. the antigen is extrinsic to the cell) can also be presented within MHC-class I by a process termed cross-presentation and is principally thought to occur in sub-specialized dendritic cell (DC) subsets (CD8\textsuperscript{+} and/or CD103\textsuperscript{+} in mouse, BDCA3\textsuperscript{+} in humans). Here phagocytosed exogenous antigen is translocated into the proteosome for processing within MHC-class I. The importance of cross presentation to GVHD pathology remains unclear at this point although it is clearly a highly active process\textsuperscript{19} that is predicted to be critical in the generation of pathogen-specific immunity.

In contrast to MHC-class I, alloantigens presented in MHC-class II are predominantly exogenous in origin and donor T cells again recognize, via the T cell receptor, polymorphic recipient peptides presented within MHC that is shared by both the donor and recipient. Thus alloantigen may be presented in MHC-class II by recipient or donor antigen presenting cells (APC). Exogenous antigen is acquired by APC via phagocytosis of dead or necrotic cells, endocytosis or macropinocytosis. The former two processes are receptor mediated (e.g. clathrin) and these pathways exist to various levels of efficiency in all professional APC (i.e. B cells, monocytes/macrophages and DC). Exogenous proteins are processed in the lysosome and transported to the endosome for loading into MHC. MHC-class II molecules themselves are transported from the ER to the Golgi as a complex with an invariant chain which is then processed in the late endosome by HLA-DM to release the class II-related invariant chain peptide (CLIP) from the MHC, facilitating replacement by peptide of appropriate affinity (reviewed in\textsuperscript{18}) before transfer to the cell surface. Endogenous antigens can also be presented directly within MHC-class
II during periods of cellular stress in a process known as autophagy. In this process endogenous proteins of nuclear, mitochondrial and cytoplasmic origin are incorporated into autophagosomes which then fuse with the lysosome to allow antigen delivery into the MHC-class II pathway. While it has recently been demonstrated that autophagy deficient recipient DC paradoxically induce more GVHD than wild type DC, it is currently unclear whether this relates to effects on antigen presentation per se. Thus the relative contribution of autophagy to antigen presentation within MHC-class II after SCT remains unknown at this point but given the inflammation and cellular stress therein, the process itself is likely highly relevant.

A third “semi-direct” pathway of antigen presentation has recently also been described. Here, donor cells can acquire recipient cell-derived cell-surface membrane including MHC-class I and class II via either trogocytosis or exosome uptake. In this process, MHC molecules loaded with alloantigen are transferred from neighbouring cells in a cell contact or an exosome-secretion dependent manner which may subsequently activate donor T cells. This process is reported to contribute to T cell activation or suppression, and is likely important as a means of antigen acquisition and potentially presentation.

In transplant settings where MHC mismatches are present, donor T cells react to recipient APC at a very high frequency (i.e. 1-10%). It is now clear that in this setting, donor T cells can cross-react to non-self (host)-MHC loaded with an antigenic peptide in a process known as molecular mimicry. Thus in MHC-mismatched transplants, recipient MHC molecules loaded with various self-peptides (i.e. peptides that may not be foreign or allogeneic to donor) may present a
conformational footprint that allows engagement of a donor TCR (Figure 1). As an example, HLA-B*4402 and HLA*4405 presenting self-peptides such as ATP-associated proteins can stimulate a TCR clone that recognizes a viral peptide presented within HLA-B*0801 (it’s self MHC). Thus there would appear an extensive capacity for conformational changes in the MHC-peptide complex to be recognized by MHC disparate T cells. In addition the TCR itself also appears capable of undergoing conformational “fine tuning” to accommodate minor conformational alterations in MHC-peptide complexes (reviewed in28). Presumably these mechanisms are responsible for the severe GVHD risk when transplanting across multiple MHC-mismatches relative to a single locus-mismatch.29,30 In the former situation, the high number of mismatched loci increase the probability of molecular mimicry of peptide-MHC complexes for numerous T cell clones. Indeed, this principal was elegantly demonstrated in 1986 by Sprent et al.31 By using B6 BMT recipients with a single mutation in MHC-class I or class II (H2-Kbm1 or H2-Ab1bm12), they demonstrated the ability of B6 donor CD8 and CD4 T cells respectively to induce lethal GVHD, despite the complete absence of any mHA disparities between donor and host, consistent the ability of recipient “self” peptides to be recognized in the context of MHC disparity. This scenario of course predicts that unlike MHC matched SCT, identification of mHA may be relatively fruitless in MHC mismatched SCT.

The requirement for host and/or donor APC for the initiation and maintenance of GVHD has been the subject of considerable study. Historically this has been referred to as direct or indirect antigen presentation. In direct antigen presentation, donor T cells recognize MHC-peptide complexes on host APC; in indirect antigen
presentation donor T cells recognize recipient-derived peptides loaded in the MHC of donor APC. A seminal study by Shlomchik et al demonstrated that host APC are necessary and sufficient for the initiation of lethal acute GVHD within MHC-class I. Subsequent studies confirmed the same held true for MHC-class II. However, the nature of the APC required appears to differ within MHC-class I and II. Within MHC-class I, hematopoietic-derived recipient APC appear to be critical although non-hematopoietic APC may play a minor role. Within MHC-class II, both hematopoietic and non-hematopoietic APC can initiate lethal acute GVHD and non-hematopoietic APC appear dominant. In contrast, donor APC appear inefficient at inducing GVHD in isolation of recipient APC regardless of the pathway.

**Recipient APC subsets**

As aforementioned, host APC have been shown to be important for the induction of GVHD. Subsequently, many studies have attempted to identify the critical APC subsets involved, predicated on the notion that deletion may prevent GVHD. Long standing dogma has held that recipient dendritic cells (DC) are the APC responsible for GVHD, based on their ability to initiate disease when transferred into MHC deficient recipients and their transient persistence and activation after total body irradiation (TBI). In similar studies, host plasmacytoid DC (pDC) could also initiate GVHD in lethally-irradiated recipients, whereas host B cells could not. In relation to the effects of TBI and myeloablative conditioning, the incidence of acute GVHD is still high, albeit delayed, after reduced-intensity conditioning in the clinic. Thus any beneficial effects of reduced inflammation in this setting is likely countered by the persistence of recipient APC and subsequent initiation of GVHD at later time points after transplant. In this regard, conditioning intensity, inflammation and the turnover
of APC after clinical BMT is likely to be crucial in determining the relative importance of various APC subsets (i.e. donor vs. host; hematopoietic vs. non-hematopoietic) in the initiation and maintenance of GVHD. Certainly DC in skin are predominantly donor in origin by at least 40 days after BMT and this is accelerated by GVHD after reduced-intensity but not myeloablative conditioning. DCs in lymphoid tissue turnover within two weeks in mice but clear data defining this in humans is lacking. In contrast to DC, macrophage turnover in humans in tissue is considerably slower than DC and may not be complete even by one year. Together, current data suggests that host DC are short lived and donor DC are likely to predominate in relevant lymphoid organs at the time of engraftment in patients receiving ablative conditioning.

In relation to DC and GVHD, preclinical studies have established the ability of recipient DC to induce GVHD in recipients otherwise devoid of all functional APC but not their importance in recipients where all APC are competent. Recently, a new generation of transgenic mice in which CD11c+ cells can be conditionally deleted has turned this concept on its head, demonstrating that recipient DC are not required to initiate MHC-class II-dependent GVHD and may actually regulate disease with the induction of T cell apoptosis. Furthermore, recipient pDC, B cells, macrophages and Langerhans cells also appear to be redundant in isolation for the initiation of lethal acute GVHD and capable of regulating disease. The pathways involved in regulation by these professional APC subsets involve IL-10 for B cells, CD47 and donor T cell uptake for macrophages and clonal deletion for DC. In relation to DC, it may not be surprising that the APC subset specialized for the most efficient presentation of limiting amounts of antigen act to delete T cells
when antigen is ubiquitous and present in excess early after SCT following chemoradiotherapy. Nevertheless, these findings raise the possibility that considerable redundancy exists within recipient professional APC in relation to their ability to induce GVHD; that vanishing small numbers are capable of initiating severe GVHD; or alternatively that an additional, likely tissue-residing APC not depleted by the above approaches is capable of inducing GVHD.

In this vein, recipient non-hematopoietic APC, once activated by conditioning chemoradiotherapy have emerged as APCs that are highly efficient in initiating MHC-class II-dependent GVHD. Indeed, TBI induces the expression of molecules required for co-stimulation (CD40, 80, 86) on non-hematopoietic MHC-class II+ cells. To date, fibroblasts have been shown to be capable of inducing cytotoxic T lymphocyte responses, as have epithelial cells once induced to express MHC-class II by inflammation (particularly interferon-γ (IFN-γ)) (Figure 2). This concept is supported by preclinical studies whereby allogeneic non-hematopoietic but not hematopoietic APC were required for acute GVHD lethality and biopsy samples from patients demonstrating high expression of HLA-class II on colonic non-hematopoietic cells after transplantation. Recently, it has been demonstrated that a HLA-DP1 variant (rs9277534G) which results in higher protein expression than the variant (rs9277534A) is associated with an increased incidence of grade II-IV GVHD, suggesting that the levels of HLA-class II expression per se correlate with the severity of GVHD. Intriguingly, consistent with CD4+ T cell-dependent GVHD, the depletion of host cDC has no effect and may also augment MHC-class I-dependent GVHD. Thus, multiple independent studies, in multiple models have demonstrated
that recipient DC are not responsible for the initiation of GVHD and moreover, their deletion is likely to be detrimental.

**The GI tract, danger signals and acute GVHD**

The primacy of the gastrointestinal tract in severe, life-threatening acute GVHD is well established. The gastrointestinal tract and the colon in particular contains an extensive microbiome and as such acts as a reservoir for DAMP/PAMP signals, particularly in the context of high dose chemoradiotherapy. In clinical transplantation, antibiotic decontamination, particularly in regard to anaerobe eradication can reduce the incidence of acute GVHD. However, recent data suggests that some anaerobe species are in fact highly protective from GVHD and so current studies focus on qualitative rather than quantitative characteristics of the microbiome in the GI tract. Experimental models have been utilized to address the importance of DAMP/PAMP mediated activation of host and/or donor APC, including effects on antigen presentation. Although C-type lectin receptors on residual host macrophages which recognise fungal components are important in pulmonary GVHD in particular, surprisingly neither MyD88 and TRIF signals which mediate Toll-like receptor (TLR) signals nor type I IFN signals in recipient cells enhanced GVHD. On the other hand, it is well established that TLR signalling of donor cells exacerbates acute GVHD. Recently, we demonstrated that donor CD103+ DC that migrate from the colon to the mesenteric lymph nodes (mLN) under the guidance of CCR7 following activation and expansion in response to TLR (MyD88 and TRIF) or receptor for advanced glycation end products (RAGE) signals are critical in promoting GVHD lethality. Importantly, this effect is dependent on the presence of GVHD initiated by recipient APC that in turn generates a profound feed-forward loop of antigen
presentation by donor DC with subsequent pathological T cell differentiation (Th1/Th17) and (α4β7 integrin-dependent) migration to the GI tract (Figure 2).

Cytokines and acute GVHD

It is clear that allogeneic disparity induces a T cell cascade culminating in organ specific damage (GI tract, liver, skin) that manifests as acute GVHD. In the first 14 days of transplant, hyper-acute GVHD often occurs in conjunction with systemic symptoms including fever, weight gain and pulmonary edema. This syndrome commonly occurs in heavily pre-treated recipients receiving myeloablative conditioning and HLA-mismatched grafts. It is attractive to propose that this syndrome is a manifestation of cytokine dysregulation and akin to the cytokine release syndrome seen after infusion of chimeric antigen receptor T cells in which IL-6 appears to play a central role. Certainly IL-6 is dysregulated after clinical transplantation but the nature of the alloantigen presentation driving this syndrome remains unclear at this time.

Antigen presentation in chronic GVHD

Chronic GVHD is defined by fibrosis and scleroderma or bronchiolitis obliterans represent cardinal diagnostic manifestations. Current NIH criteria describes classic chronic GVHD (without acute GVHD) and an overlap syndrome in which both acute and chronic GVHD appear together. Such complexity and diversity is difficult to model experimentally although systems do exist where fibrosis is a feature. Although the role of antigen presentation in chronic GVHD remains less well understood than that in acute GVHD, emerging data suggest antigen presentation in the thymus and peripheral tissues plays an essential role in maintaining tolerance by
promoting donor regulatory T cell (T_{reg}) homeostasis.^{72-74} MHC-class II expression on donor DC within the thymus is pivotal to control autoreactive (donor-reactive) thymic-emigrant T cells and the prevention of disease in animal models.^{72,75} While the relevance to clinical chronic GVHD remains less clear, thymic dysfunction, immune suppression and autoreactive T cells are certainly a cardinal feature of human chronic GVHD. Intriguingly, the transplantation of CD80/86-deficient or CD40-deficient donor bone marrow that lacks costimulatory signals for efficient MHC-class II antigen presentation does not cause this morbidity, concordant with previous findings in which thymic clonal deletion is functionally intact in these mice.^{76,77} Instead these deficiencies result in protection from GVHD mediated by mature donor T cells within the graft.^{78} Donor DC, both in the thymus and peripherally are known to control T_{reg} development and maintenance respectively.^{79} Again, defects in T_{reg} are also a cardinal feature of chronic GVHD, occurring within the context of CD4^{+} lymphopenia.^{74} This has led to the development of immune-restorative therapeutic strategies such as low dose IL-2 to expand T_{reg}.^{80,81} Antigen presentation by DC to T_{reg} is required for T_{reg} homeostasis in general^{82,83} although direct evidence for this after SCT has yet to be established. Donor B cells^{84,85} and macrophages^{86} have also been identified as mediators of chronic GVHD, however these effects would appear likely due to their effector function (in generating alloantibodies^{87} and TGFβ^{86} respectively) rather than antigen presentation per se. Again, exclusion of a role for antigen presentation requires further testing.

**GVHD and the inhibition of antigen presentation**

While alloantigen presentation by donor DC in the GI tract is markedly enhanced early after SCT in the presence of acute GVHD,^{42} chronic GVHD is highly
immunosuppressive and indeed the major cause of mortality and morbidity in these patients is opportunistic infection. While there are undoubtedly significant intrinsic T cell defects during chronic GVHD, recent animal data has demonstrated defective antigen presentation, predominantly by donor DC within MHC-class II. Subsequent work demonstrates this results in overwhelming cytomegalovirus (CMV) disease with hepatic necrosis and lethality due to the inability to generate pathogen-specific immunity. Conversely, CMV reactivation may be correlated with reduced relapse of acute myeloid leukaemia. Although CMV is detected in leukemic blasts and this is speculated to provide virus-derived antigens as GVL targets, the mechanisms involved will need further careful experimentation. While the nature of the defect in antigen presentation (e.g. self or allogeneic) within donor DC remains poorly defined, it appears to be the result of chronic inflammation and immune activation.

**Antigen presentation and the separation of GVHD and GVL**

Recipient hematopoietic APC are crucial for the induction of both MHC-class I and MHC-class II-dependent GVL in MHC-matched mHA-mismatched models. Donor APC are either not required, or have limited capacity (relative to recipient APC) to promote GVL, depending on the model systems used. Recently the CD8+ DC subset has been suggested as a critical APC for the induction of GVL although this appears somewhat at odds with data in relation to GVHD. On the other hand, MHC expression on leukemia cells is clearly critical for effective GVL responses. Thus, the expression of MHC-class I on target cells is required for CD8+ T cell-dependent GVHD and GVL, whereas MHC-class II expression is required for CD4+ T cell-dependent GVL but not GVHD. The latter is due to the fact that inflammatory cytokines produced by CD4+ T cells can induce target tissue damage independent of
cognate TCR-MHC interactions. Targeting presentation of not only mHA but also leukemia-associated antigens (LAA) (e.g. Wilms tumor antigen-1 or proteinase 3) are also promising approaches to enhance GVL effects without attendant risks of GVHD. Since donor APC are minimally required, if at all, for GVL, transient interruption of alloantigen presentation by donor CD103+ DC migrating from the colon would appear a cellular cascade central to the effective separation of GVHD and GVL. Antibodies with activity against molecules that are expressed on activated cells including DC may be effective if they can be administered in the right window after SCT when donor DC are active and recipient DC have been eliminated. Of note, since donor APCs are likely crucial for the generation of pathogen-specific immunity, targeting donor DC or APC subsets will need to be studied with caution in the clinic. In addition, understanding the nature and mechanisms by which recipient non-hematopoietic APC function to initiate GVHD may be an important key to separating GVHD and GVL in future. Whether this defect in antigen presentation plays a role in the Treg deficiency seen during chronic GVHD remains to be elucidated but nonetheless serves as an attractive hypothesis.

Conclusions and Future directions

Alloantigen presentation is central to both the therapeutic (GVL) and adverse effects (GVHD) of allogeneic SCT and as such represents a pathway of primary importance to clinical outcome. There would appear multiple therapeutic strategies worthy of exploration at this time which are described in Table 1. Firstly, the adoptive transfer or induction of defined mHA-specific donor T cells as well as LAA-specific T cells would appear an attractive immunotherapy approach to prevent and treat disease relapse. As long as the mHA is restricted to hematopoietic cells, donor vaccination
could also be contemplated. Secondly, interruption of alloantigen presentation by donor DC within the GI tract, at a time when recipient DC have been eliminated, should separate GVHD effects and could be undertaken by inhibition of the chemokine signal (CCR7) required for migration of these DC to primary lymphoid organs. The role of the microbiome in generating PAMPs that drive antigen presentation and cytokine secretion in the GI tract also makes it a highly attractive target for modulation after transplantation. It is important to note that many of our standard immune suppressants have also been described to impact on antigen presentation directly (Table 1 and reviewed in). Finally, a better understanding of the recipient non-hematopoietic APC that induce GVHD and the pathways that are required for their pathogenicity, particularly autophagy, may allow for therapeutic intervention.

Acknowledgements

The authors thank Ms Madeleine Flynn of QIMR Berghofer for generation of the graphics.

Authorship

Contribution: M.K. and G.R.H wrote the manuscript.

The authors apologize to the authors of the many important articles that were unable to be referenced due to space restrictions.

Conflict of interest disclosure: M.K. declares no competing financial interests. G.R.H has received funding from Roche for clinical studies of IL-6 inhibition.
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Table 1. Therapeutic agents and their potential effects on alloantigen presentation in transplantation

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<th>Commercial examples</th>
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<td>PF-04494700</td>
<td>Inhibit donor DC activation and expansion in the GI tract$^{42}$</td>
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<td>TLR4 inhibitor</td>
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<td>Inhibit donor DC activation and expansion in the GI tract$^{42}$</td>
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<td>Prevent migration of alloantigen bearing tissue donor DC into lymph nodes$^{42}$</td>
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<td>Anti-a4b7 integrin Ab</td>
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<td>Inhibit T cell expansion and Th17 differentiation in response to APC derived IL-6$^{42}$</td>
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<td>Janus Kinase (JAK)</td>
<td>Ruxolitinib</td>
<td>Impaired DC activation and migration$^{1}$</td>
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**Currently used immune suppressants**

|                              |                            |                     |                                                           |
| calcineurin inhibition       | Cyclosporin                | Tacrolimus          | Inhibition of antigen presentation in DC$^{100}$          |
| mTOR inhibition              | Rapamycin                  |                     | Promotion of tolerogenic DC phenotype$^{109}$              |
| Post-transplant cyclophosphamide | Cyclophosphamide         |                     | ? Depletion of APC                                         |
Figure legends

Figure 1. Alloantigen presentation and recognition in MHC matched and mis-matched transplantation

Left) peptide recognition in MHC-matched transplant; Since donor T cells are selected through positive and negative selection in donor thymus, the donor T cell receptor recognizes host-derived (non-self) minor histocompatibility antigens (mHA, green) within a MHC (red) that is shared between donor and host. Right) Molecular mimicry in MHC mis-matched transplantation; the donor T cell receptor can recognize a mis-matched host MHC (blue) loaded predominantly with antigenic-peptide derived from ubiquitous self-proteins (yellow) rather than polymorphic mHA.

Figure 2. The initiation and amplification of GVHD by alloantigen presentation

Naive donor T cells contaminating the stem cell graft migrate into recipient LNs and/or GVHD target organs, particularly the GI tract. They encounter recipient alloantigen presented by recipient hematopoietic APC in the LNs and/or by recipient non-hematopoietic APC in target tissue (e.g. fibroblasts, epithelial cells). The latter obtain functional antigen presentation capacity following tissue damage induced by chemoradiotherapy during conditioning and cytokines generated following donor T cell activation. Hereafter donor T cells proliferate and differentiate into Th1 and Th17 cells and induce GVHD in the target tissues, particularly the GI tract. Tissue damage within the colon at this point allows microbiome-derived DAMP/PAMP signals to expand and activate donor CD103+ DC in situ which subsequently migrate into the mesenteric LN (mLN) under the influence of CCR7. Within the mLN these donor DC secrete IL-12 and present alloantigen to donor T cells to further drive pathological Th1 and Th17 differentiation whilst imprinting gut-homing α4β7 integrins, permissive
of massive secondary migration into the gut, resulting in fulminant disease.
Schematic highly modified from$^{42}$. 
Alloantigen presentation and graft-versus-host disease: fuel for the fire

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