Published in final edited form as:

Transplantation. 2020 October ; 104(10): 2024–2034. doi:10.1097/TP.0000000000003169.

Memory T cells in transplantation: old challenges define new directions

Michael Nicosia, PhD¹, Robert L. Fairchild, PhD¹, Anna Valujskikh, PhD¹

¹Department of Inflammation and Immunity, Lerner Research Institute, Cleveland Clinic, Cleveland, OH

Abstract

Immunological memory is the ability of adaptive immune system to quickly and specifically recognize previously encountered antigens and initiate an effector response. Alloreactive memory cells can mount rapid and robust responses to the transplanted organ resulting in allograft injury.

Thus preexisting humoral or cellular memory alloresponses are typically associated with poor graft outcomes in experimental and clinical transplantation. While both B and T lymphocytes exhibit memory responses, this review discusses recent updates on the biology of memory T cells and their relevance to the field of transplantation. Three major areas of focus are the emergence and characterization of tissue resident memory T cells, manipulation of T cell metabolic pathways and the latest promising approaches to targeting detrimental T cell memory in the settings of organ transplantation.

Introduction

During protective immune responses, T cell receptor (TCR) engagement with peptide:MHC complex on the surface of antigen-presenting cells (APCs) causes extensive T cell expansion followed by effector functions and pathogen clearance. During the resolution of effector T cell (T_{EFF}) responses the antigen specific T cell populations contract, but leave behind a small subset of long-lived memory cells that are uniquely positioned and equipped for future host protection. Due to the same properties however, memory T cells with reactivity towards donor alloantigens undermine successful organ transplantation.

The obvious priming and accumulation of alloreactive memory T cells can occur via direct exposure to foreign MHCs during previous transplantations, blood transfusions and pregnancies.^{1–3} In addition, alloreactive memory cells are generated as a result of heterologous immunity occurring when the peptides of commensal bacteria or environmental antigens presented on self-MHC mimic the molecular structure of

Correspondence: Anna Valujskikh, PhD, NB3-67, Cleveland Clinic, 9500 Euclid Avenue, Cleveland, OH 44195, Tel: 216-445-5452; Fax: 216-444-8372; valuysa@ccf.org.

AUTHORSHIP PAGE

All of the authors participated in the organization and preparation of this review.

MN participated in organizing and writing the article. RLF participated in organizing and writing the article. AV participated in organizing and writing the article.

The authors declare no conflict of interest

alloantigens.⁴ In laboratory mice, which are raised in standard pathogen-free conditions,⁵ such endogenous memory T cells make up 0.05–1.00% of the total T cell pool.^{6,7} In contrast, constant exposure to pathogens and infections in nonhuman primates (NHP) and humans can lead to the acquisition of potent alloreactivity.⁸ Multiple studies demonstrated occurrences of cross-reactivity between individual pathogens and allo-MHC molecules. For instance, Adams et al. showed that prior lymphocytic choriomeningitis virus (LCMV) infection of C57BL/6 mice induces alloreactive CD8 T cells that can subsequently reject skin allografts despite mixed chimerism-costimulatory blockade based tolerance induction.⁹ Analogously, murine gammaherpesvirus 68 (MHV68) infection induced heterologous immunity in C57BL/6 mice resulting in accelerated rejection of BALB/c skin allografts.¹⁰ Bacterial infections such as *Leishmania major* can also stimulate the generation of alloreactive T cells and worsen transplant outcomes in specific mouse strain combinations.¹¹ Examples in humans have also been demonstrated such as HLA-B8⁺ patient sensitization to HLA-B4402^{8,11} following Epstein-Barr virus (EBV) infection.

Regardless of their origin, donor-reactive memory T cells are detrimental in the context of organ transplantation. Preexisting CD4 and CD8 memory T cells are rapidly reactivated and give rise to pathogenic effector subsets that can migrate into the graft and secrete proinflammatory cytokines or mediate cytotoxicity upon reactivation.¹² Memory helper T cells provide efficient help for B cells to induce generation of donor specific alloantibody (DSA) leading to antibody-mediated rejection.^{13–15} Animal studies demonstrated that within 24–48 h posttransplant, CD4 and CD8 memory T cells infiltrate allografts where they exacerbate the effects of ischemia/reperfusion injury resulting in endothelial cell activation, intra-graft chemokine production and DAMPs release.^{16,17}

Over the past twenty years, several subsets of memory T cells were defined based on the expression of adhesion molecules, chemokine and cytokine receptors (Table 1). Initially memory T cells have been divided into CD62L^{hi}/CCR7^{hi} central memory cells (T_{CM}) and CD62L^{lo}/CCR7^{lo} effector memory T cells (T_{EM}). Expression of CCR7 and CD62L in T_{CM} facilitates homing from the blood into secondary lymphoid organs, while T_{EM} can migrate from the blood to the peripheral tissues and are equipped to do so through increased expression of integrins and specific chemokine receptors.¹⁸ In the last decade a specialized noncirculating, nonlymphoid tissue resident memory T cells (T_{RM}) has been characterized.^{19,20} These localized cells provide antigen surveillance and rapid responses at sites of previous infections, especially at barrier sites such as skin, lung, intestine, and genitourinary tract.¹⁹

Newly emerging memory T cell subsets

Tissue resident memory cells - T_{RM}—Highly specialized and heterogenous T_{RM} cells are a recent addition to the family of T cell subsets. They are noncirculating, a property that is facilitated by the surface markers used to define them. While both CD4⁺ and CD8⁺ memory T cells make up T_{RM} subsets, most of the current knowledge in the field is generated by studying CD8⁺ T_{RM} cells. T_{RM} cells are thought to develop from common memory precursors that are present among activated CD127^{hi} KLRG1[−] T cells following infection.²¹ In the skin, these precursors enter the epidermis early after infection and slowly

mature into T_{RM} phenotype (Table 1) following infection resolution. Maturation results in elevated expression of CD69, an early activation marker that promotes tissue retention of T_{RM} precursors by antagonizing the functions of S1PR1,²² a chemokine receptor mediating lymphocyte egress into circulation. This is accompanied by the reduction in the expression of other known facilitators of tissue egress, CCR7 and KLF2.²³ CD69 upregulation precedes the expression of another T_{RM} associated marker – CD103, an adhesion molecule of the integrin family.²⁴ CD103 binds to E-cadherin expressed by epithelial cells and is commonly referred to as a universal T_{RM} marker. However, parabiosis studies showed the presence of CD103⁺ T_{RM} cells in a number of tissues^{25–28} and distinct cytokine output by CD103⁺ and CD103[−] T_{RM} cells.²⁹ Another marker commonly associated with T_{RM} is CD49a (Very Late Antigen-1)³⁰ and yet there are subsets that do not express CD49a,³¹ highlighting the heterogeneity of T_{RM} population. TGF- β was identified as a key cellular factor required for CD103 expression and the establishment of T_{RM} in multiple tissues.^{32,33} While other cytokines including IL-33, TNF- α , and IFN- α/β have been shown to promote T_{RM} phenotypes in vitro, their role in the development and differentiation of T_{RM} cells in vivo remains to be established.^{34,35} The use of latest tools in genomics and proteomics established that the newly matured T_{RM} cells assume a unique transcriptional profile. T_{RM} maturation is dependent upon the activities of the transcription factors Hobit and Blimp-1, both of which are necessary for tissue residence in Natural Killer (NK) T cells and liver-resident NK cells in addition to T_{RM}.³⁶ The exact requirements for T_{RM} survival are not fully understood. CD8 T_{RM} down-regulate the receptors for IL-7 and IL-15,^{35,37,38} key mediators of T cell homeostasis supporting survival and proliferation respectively.³⁹ While T_{CM} and T_{EM} CD8 cells require IL-15 for maintenance,⁴⁰ its role in T_{RM} cells appears to be location dependent.⁴¹

The presence of T_{RM} at the barrier sites enables an immediate detection and robust responses to previously encountered pathogens. Activation, proliferation and effector functions of circulating memory T cells require the drainage of peripheral antigens to the secondary lymphoid organs. This process requires time, allowing pathogen to amplify within the body. Not surprisingly, enhanced protection by T_{RM} cells compared to circulating memory cells was demonstrated for skin infections with Herpes simplex virus (HSV) and Vaccinia virus.^{19,37} The same is true of T_{RM} cells at mucosal sites, common points of entry for many infectious agents such as influenza.^{42,43} Furthermore T_{RM} cells can play an important role in the control of nonacute infections such as the reactivation of latent HSV.⁴⁴

Given rapid and robust T_{RM} responses, it is likely not a coincidence that organs containing high numbers of passenger memory T cells such as lung and intestine demonstrate some of the poorest clinical outcomes and lowest rates of graft acceptance.⁴⁵ Despite dramatic improvements in first year posttransplant attrition, long-term survival rates for these organs haven't improved since the 1980s.²¹ In the context of transplantation, T_{RM} cells may come from 2 distinct sources - donor organ and recipient. It remains unclear whether donor derived passenger leukocytes contribute to alloreactivity and rejection or are simply cleared by infiltrating recipient T cells. A recent report by de Leur and colleagues addressed the composition of T_{RM} in kidney transplants.⁴⁶ All evaluated allografts revealed the presence of both CD103⁺ and CD103[−] T_{RM} cells that were predominantly CD8⁺, and the origin of these cells dependent on time elapsed after transplantation. Grafts analyzed within one

month posttransplant had a T_{RM} population dominated by donor-derived T_{RM}. Transplants analyzed at later time points predominantly contained recipient derived CD8⁺ T_{RM}, with no detectable donor-derived T_{RM} by 5 months posttransplant.⁴⁶ One proposed theory is that certain signals generated during transplantation, such as those during ischemia reperfusion injury, trigger lymphocyte tissue egress. This has been shown in the recipients of intestinal grafts, who exhibit donor-derived T cells in circulation 7–8 weeks posttransplant.⁴⁷ In addition to the potential donor antigen delivery to recipient secondary lymphoid organs, the presence of donor-derived T_{RM} increased the risk of graft-versus host disease in this study.⁴⁷ Animals studies suggest that donor T_{RM} cells can precipitate graft tissue injury through multiple mechanisms. In a murine cardiac allograft study, donor-derived T_{RM} cells provided help through cognate interactions to recipient B cells, causing autoantibody generation and ultimately cardiac allograft vasculopathy, a major limiting factor for long term graft survival.⁴⁸

There are however, conflicting reports that donor-derived T_{RM} cells may confer a protective role in transplantation. Consistent with this scenario, irradiation of donor grafts in a rodent model of spontaneously accepted liver transplants results in allograft rejection.⁴⁹ As posttransplant infection can often precipitate rejection, the presence of protective sentinel cells in the graft may be beneficial. A study from Snyder et al. investigating T_{RM} cells in human lung transplant patients showed that grafts with higher frequencies of donor-derived T_{RM} cells experience fewer instances of primary graft dysfunction and acute cellular rejection in comparison to the grafts with fewer donor T_{RM} cells.⁵⁰ Such findings suggest that future therapies that aim to maintain donor-derived T_{RM} cells and prevent their replacement by recipient-derived T_{RM} cells may improve transplant outcome. However, considering incomplete and often controversial data on the role of T_{RM} cells in graft injury versus protection, therapies targeting donor derived T_{RM} may be better suited to certain types of solid organs, and as such should be put on hold until the role of these cells is better understood.

Terminally differentiated effector memory T cells - T_{EMRA}—Studies in humans further added a distinct subset of terminally differentiated effector memory T cells (T_{EMRA}) that re-express CD45RA surface marker typical of naïve T lymphocytes.⁵¹ T_{EMRA} cells require less IL-2 for survival and maintenance, are highly cytotoxic,⁵² and produce high levels of IFN γ ¹⁸ simultaneously with low proliferative capacity and increased sensitivity to apoptosis.⁵³ Furthermore, T_{EMRA} cells lack CCR7 expression⁵⁴ and are predominantly CD8⁺.^{55,56} These cells have been shown to make up a larger population of the T cells in blood and lymphoid tissues with aging as the proportion of naïve T cells decrease.⁵⁶ They are characterized as terminally differentiated effector cells high in IFN γ secretion,¹⁸ highly cytotoxic⁵² and highly sensitive to apoptosis with a low proliferative capacity.⁵³ As with many of populations described T_{EMRA} cells demonstrate heterogeneity within their population, as a naïve- T_{EMRA} intermediate subset has been identified as being CD27⁺CD28^{+/-}, whilst late differentiated and highly cytotoxic T_{EMRA} cells are CD27⁻CD28⁻.⁵⁷

T_{EMRA} cells have been the focus of several studies in renal disease and transplantation. Patients with end-stage renal disease show an increased frequency of CD8⁺ T_{EMRA} cells,

above the normal increase seen during aging.⁵⁸ A 2012 study on kidney transplant recipients revealed that recipients that did not suffer any biopsy-proved acute rejection episodes within the first 2 years demonstrated elevated pretransplant frequencies of CD8⁺ T_{EMRA} cells compared to those that did suffer rejection in that timeframe.⁵⁹ However, patients with elevated T_{EMRA} cell frequencies also had increased rates of cancer during immunosuppression, indicating a state of immune senescence. Kidney transplant recipients have expanded numbers of CD8⁺ T_{EMRA} cells,⁶⁰ yet despite stable long-term graft function long term persistence of this expansion is associated with a twofold increased risk of graft dysfunction in these patients.⁶¹ Transplant therapies directly targeting T_{EMRA} cells have yet to be developed but the effect of some preexisting therapies have been observed. A longitudinal study on the effects of Rituximab (αCD20 mAb) induction on T and B lymphocyte phenotype and function show stable counts and frequencies of T_{EMRA} cells for up to 24 months posttransplant regardless of Rituximab or basiliximab (αCD25 mAb) induction.⁶² Given their association with poor long-term outcomes, targeting T_{EMRA} cells may prove highly beneficial at extending allograft survival.

Memory T follicular helper cells - T_{FH}—Follicular T helper cells (T_{FH}) are a subset specialized in providing help to cognate B cells within the lymphoid follicles via expression of CD40, ICOS, IL-21, IL-4 and other molecules.^{63,64} The characteristic markers of this subset are a transcription factor Bcl6 and a chemokine receptor CXCR5 that is critical for recruitment and retention within lymphoid follicles. In the last decade, studies have shown how T cell memory and T_{FH} cells intersect.⁶⁵ Following resolution of the immune response T_{FH} cells down regulate CXCR5 expression to an intermediate level, but can quickly regain this expression upon secondary challenge.⁶⁶ Studies have shown that antigen specific memory CD4⁺ T cells are better providers of help to B cells than Ag-specific naïve CD4⁺ T cell controls, and whilst memory is not strictly lineage restricted, cell intrinsic mechanisms favor T_{FH} lineage commitment within memory cell population.^{67,68}

A number of studies have shown the effect of conventional immunosuppression on T_{FH} cells as well as detailing the benefits of T_{FH} targeted therapies. For example, Tacrolimus has been shown to partially inhibit the generation of T_{FH} cells.⁶⁹ Another study suggested that memory T_{FH} cells may be affected by tacrolimus treatment however more work is needed to confirm these findings.⁷⁰ The effects of targeting the coinhibitory or costimulatory pathways essential for T_{FH} function in animal models of transplantation have been described in another review.^{71,72} None of these studies however address the direct effects on memory T_{FH} cells, and more work is required to specifically control these cells in transplant settings.

Memory T peripheral helper cells - T_{PH}—T peripheral helper (T_{PH}) cells fulfil the role of providing help to B cells outside of the secondary lymphoid organs.⁷³ Phenotypically these cells share many common markers and effector molecules with their follicular T_{FH} counterparts including PD-1, ICOS, TIGIT, CXCL13 and IL-21, but express low levels of T_{FH} defining markers CXCR5 and Bcl6.⁷³ Instead T_{PH} cells express chemokine receptors CCR2 and CCR5 enabling their migration into sites of inflammation.⁷³ These cells are significantly expanded at the sites of peripheral inflammation such as the synovium of rheumatoid arthritis (RA) patients⁷⁴ and are found in the circulation of patients with

systemic lupus erythematosus.⁷⁵ From a functional perspective T_{PH} cells can promote B cell differentiation into plasmablasts in an IL-21 manner.⁷⁴ Vaccine studies showed that poor responders to influenza vaccination have dysfunctional T_{PH} cells with impaired IL-21 secretion and thus are unable to support IgG production by B cells.⁷⁶ In transplantation these cells may pose a significant risk by providing help to B cells in ectopic lymphoid structures (ELS) within the graft. Both kidney and heart allografts can develop ELS that contain donor-specific antibody (DSA) generating plasma cells.^{77,78} Furthermore, homogenates from kidney allografts undergoing rapid rejection revealed elevated expression of IL-21 mRNA that correlated with the increased expression of activation-induced cytidine deaminase (AID) a marker of B cell activation.⁷⁹ Taken together these data suggest a link between T_{PH} and allograft rejection via DSA production within ELS. In theory, the same principles that govern therapeutic targeting of T_{FH} should hold true for T_{PH} to prevent or alleviate antibody mediated rejection (AMR).

T memory stem cells - T_{SCM}—Stem cell like memory T cells or T memory stem cells (T_{SCM}) are a rare newly identified population of memory T cells. Mouse T_{SCM} were initially described as a population with a naïve-like phenotype, but with high expression levels of stem cell antigen 1 (SCA1), IL-2 receptor beta chain (IL-2R β) and chemokine receptor CXCR3, the last 2 being memory cell markers.^{80,81} The novel subset was termed T_{SCM} because of their ability to maintain graft-versus-host disease (GVHD) following serial transplantation into allogeneic recipients and due to their ability to reconstitute the full diversity of memory and effector T cells⁸⁰. Human and NHP T_{SCM} are phenotypically identified as co-expressing classical naïve surface markers, CD45RA⁺ and CD62L⁺, and memory associated markers including CXCR3, IL-2R β , CD58, CD11a and CD95 (Table-1).^{81,82}

T_{SCM} have been implicated in maintenance of the immune response to chronic infections, where T_{EFF} cells eventually succumb to exhaustion and thus need to be replaced.⁸³ Studies of chronic viral and parasitic infections have recently demonstrated that there is a negative correlation between disease severity and the frequency of circulating T_{SCM} cells, suggesting their presence is required to replenish exhausted cells.^{84–86} T_{SCM} have also been implicated in disease exacerbation. Elevated frequencies and activation of CD8⁺ T_{SCM} cells have been found in patients with aplastic anaemia, a condition where cytotoxic T cells target haematopoietic progenitor cells.⁸⁷ In a transplantation study, patients with elevated numbers of T_{SCM} following an immunosuppressive regimen that included anti-thymocyte globulin induction were at higher risk of treatment failure and subsequent disease relapse.⁸⁷ This raises interesting questions about the role of T_{SCM} during reconstitution of the immune repertoire following lymphoablation in transplant recipients. In particular it would be interesting to see how targeted depletion of T_{SCM} in conjunction with a clinically relevant immunosuppressive regimen effects graft outcomes as well as lymphocyte reconstitution kinetics and the reconstituted lymphocyte repertoire.

Targeting alloreactive memory T cells: updates on existing strategies and promising alternatives.

Manipulating metabolic pathways—Targeting metabolic pathways has recently emerged as a powerful approach to modulating T cell homeostasis, activation, and functions. Naïve T cells are metabolically quiescent, generating energy via breakdown of glucose, fatty acids and amino acids to drive oxidative phosphorylation (OXPHOS).^{88–91} Transitioning from a resting naïve into a highly proliferative activated state requires extensive metabolic reprogramming. This is driven by a rapid induction of aerobic glycolysis while maintaining mitochondrial OXPHOS and reactive oxygen species (ROS) output.^{92,93} While aerobic glycolysis is not the most effective means to generate ATP (2 ATP molecules generated per molecule of glucose compared to 36 molecules of ATP per glucose for OXPHOS), it does confer other metabolic advantages such as the opportunity to synthesize lipids, proteins and carbohydrates and nucleic acids.⁹⁴ Additionally, T cells can demonstrate metabolic plasticity switching between aerobic glycolysis and OXPHOS depending upon the nutrient levels of their environment.^{95,96}

Upon activation, extensive metabolic reprogramming is mediated by several signaling pathways and transcription factors. Mammalian target of rapamycin (mTOR) is one of the major regulators in this process. mTOR signaling integrates signals related to nutrient level sensing, stress responses, TCR engagement, and in turn can induce pathways linked to metabolism, cell growth and proliferation.^{97,98} mTORc1 activation induces hypoxia-inducible factor 1 α (HIF-1 α) and Myc, transcription factors that lead to increased glucose uptake and diversion of glucose from OXPHOS to aerobic glycolysis, and glutaminolysis respectively.^{99,100} Another key regulator of T cell metabolism is AMP-activated protein kinase (AMPK), that becomes activated when the ratio of AMP to ATP increases in a metabolic stress sensing function. Transient activation of AMPK upon TCR stimulation opposes mTOR function and facilitates memory T cell development by promoting catabolic pathways and energy conservation.^{101,102} Memory T cells do not heavily utilize aerobic glycolysis, and instead prominently rely on OXPHOS, in part due to the mitochondrial catabolism of fatty acids.¹⁰³ This process is dependent on free fatty acids generation from triacylglycerols (TAG). TAG synthesis in turn requires the function of a glycerol channel Aquaporin 9 which is up-regulated in activated T cells in response to IL-7R signaling and supports the survival of memory CD8 T cells.^{103,104} Recent studies have shown that inhibition of another member of aquaporin family, Aquaporin 4, reduces activation, proliferation and cytokine production in both naïve and previously sensitized T cells and significantly prolongs mouse heart allograft survival.^{105,106} Whether the effects of Aquaporin 4 inhibition are mediated through metabolic regulation remains to be elucidated. Nevertheless, targeting aquaporin channels may prove to be a useful future strategy for modulating T cell metabolism.

Memory T cells display a distinct mitochondrial morphology, with more fused networks of mitochondria compared to T effector cells with more isolate and punctate mitochondria.¹⁰⁷ However, there are subset variances with T_{CM} and T_{RM} being more reliant on mitochondrial OXPHOS than T_{EM}.¹⁰⁸ These metabolic distinctions provide basis for therapeutic manipulations of memory T cell subsets in transplantation, autoimmunity and cancer.

Depending on the type of the disease and on the desired immune response, altering the balance between glycolysis and OXPHOS should facilitate or inhibit memory T cell generation and functions. Studies in cancer have shown that inhibition of glycolysis with 2-deoxyglucose enhances memory T cell formation and provides enhanced anti-tumor activity. However, the study used a genetic approach to enforce glycolytic metabolism and this in turn limited CD8⁺ T cell memory development.¹⁰⁹ Similarly, treatment with a different glycolytic inhibitor, dichloroacetate (DCA), was shown to augment CD8 T cell activity and block tumor progression in a mouse model of melanoma.¹¹⁰

Conversely, inhibitors of fatty acid oxidation and mitochondrial OXPHOS should be utilized to control memory T cell activity. In NZB/W and MRL/lpr mouse lupus models, T cells exhibit increased mitochondrial mass and polarization and rely heavily on OXPHOS. The pharmacological inhibition of geranylgeranyl transferase, a component of fatty acid metabolism pathway, not only altered T cell metabolic phenotype but also decreased autoantibody production and lupus nephritis manifestations in these mouse strains.^{111,112} However, caution should be applied when interpreting such data as the resulting disease is often the sum of effector functions in different T cell subsets. One such example is a mouse study of experimental autoimmune encephalitis (EAE) model in which glycolysis inhibitor DCA promoted Treg functions and diminished pathogenic effector Th17 cells.¹¹³ There is accumulating evidence that T_{RM} cells have unique metabolic requirements compared to other memory T cell subsets.¹¹⁴ For example, skin-resident CD8⁺ T_{RM} cells demonstrate dependency on exogenous free fatty acid acquisition and metabolism that may be exploited therapeutically. Thus in vivo treatment with a pharmacologic fatty acid oxidation inhibitor trimetazidine decreased the survival and maintenance of T_{RM} in skin.¹¹⁵ The efficacy of this approach in targeting CD4⁺ T_{RM} cells or T_{RM} cells at different anatomic locations warrants further investigation.

Several preclinical studies evaluated the efficacy of metabolic inhibitors in bone marrow and solid organ transplantation. In a model of murine graft versus host disease (GVHD), a 2-week treatment with a fatty acid metabolism inhibitor etomoxir decreased GVHD scores up to a month after treatment cessation. Etomoxir reduced proliferation and induced apoptosis in the pathogenic CD8⁺ T cells that were highly proliferative prior to the treatment whereas nonproliferating cells were spared. In a different study, small molecule inhibitors of the F₁F₀ ATPase in the electron transport chain have also shown promise in GVHD treatment. The treatment specifically induced apoptosis in the pathogenic cell populations and markedly decreased pro-inflammatory cytokine production improving GVHD scores and survival rates.^{116,117} The study by Lee et al. demonstrated the potency of this approach in 2 mouse models of solid organ transplantation across MHC mismatch barrier. In this work, metabolic reprogramming with inhibitors of glycolysis, glutamine-derived pathways and mitochondrial OXPHOS, abrogated activation induced T cell proliferation and cytokine production leading to a significant prolongation of skin allograft survival and long-term heart allograft acceptance.¹¹⁸ Although not done in sensitized recipients this data clearly demonstrates the power of targeting metabolic pathways in transplantation. Future studies evaluating combinations of agents targeting T cell metabolism with clinically relevant immunosuppression regimens are required to fast-track these therapies from bench to bedside.

Lymphoablation—Antibody-mediated lymphocyte depletion is used as induction therapy in high risk recipients to overcome the presence of preexisting donor-reactive immunity and has been extensively reviewed previously.^{119–122} Multiple preclinical and clinical studies established that a proportion of memory/effector T cells survives the most common forms of lymphoablative induction, namely anti-CD52 mAb (CAMPATH-1) and ATG. Furthermore, the expansion of depletion-resistant memory T cells has been associated with acute rejection episodes in renal transplant recipients.¹²³ Rodent studies using the same or analogous lymphoablative reagents showed that not only do memory T cells resist depletion but in fact they thrive in the lymphopenic environment, dominate the reconstituted T cell repertoire, and are the principle mediators of allograft rejection under these conditions¹²³. Notably, memory T cells in nonlymphoid locations such as liver and lung appear to be more resistant to lymphoablation suggesting that the potential pathogenic role of T_{RM} cells in lymphopenic recipients may be amplified.¹²⁴ Studies by our group demonstrated that the recovery of pathogenic CD8⁺ T cells was critically dependent on the depletion-resistant memory CD4 helper T cells, B lymphocytes and CD40/CD154 signaling.¹²⁵ Consistent with findings in mATG treated mice, a combined therapy of anti-CD154, ATG and mycophenolate mofetil (MMF) inhibited donor reactive cellular responses in a robust xenotransplantation pig-to-baboon model.¹²⁶ Lymphoablation poses challenges of its own and was well demonstrated in a 2014 NHP kidney transplant study, which used a combined therapeutic approach of lymphoablation and belatacept treatments.¹²⁷ Animals that received combination therapy maintained graft function based on serum creatinine and blood urea nitrogen levels and demonstrated no signs of clinical rejection or detection of alloantibody, but 7 of the 8 animals did succumb to posttransplant lymphoproliferative disorders (PTLD) and infections.

One therapeutic agent that preferentially targets memory/effector T cells is Alefacept, a fusion protein combining the constant region of human IgG1 with the extracellular domain of LFA-3. LFA-3 is one of the ligands for CD2, a molecule that is upregulated on CD45RO⁺ effector/memory T cells allowing for more efficient depletion of this population compared to other therapies but still depleting all other subsets.^{128–130} Notably, Alefacept lymphoablation synergized with CTLA4-Ig in improving allograft survival in nonhuman primates presumably due to efficient depletion of CD8⁺CD2^{hi}CD28⁻ effector memory cells that are typically resistant to costimulatory blockade.¹³¹ Unfortunately, despite showing promise in the treatment of plaque psoriasis and in both solid organ and bone marrow transplantation, Alefacept was withdrawn from the market for commercial reasons.^{128,129,131,132} Nevertheless, targeting CD2 by alternative reagents remains an attractive possibility for lymphoablative induction. The efficacy of anti-CD2 rat mAb BTI-322 in treatment of acute rejection and as induction therapy was demonstrated in renal transplant patients many years ago.¹³³ Recently, the interest in this approach re-emerged due to generation of humanized BTI-322, Siplizumab.^{134–136} Siplizumab demonstrated promising safety profile in renal allograft recipients and was successfully used as part of tolerance induction trial in which the majority of HLA-mismatched kidney allograft recipients could successfully be weaned off chronic maintenance immunosuppression for at least 5 years.^{135,136} The mechanistic studies on Siplizumab were impeded due to its specificity for a binding site present only on the human and chimpanzee CD2 antigen. Most recently, the development of anti-CD2 mAbs with broad primate reactivity provided an opportunity of

investigating this approach in more accessible nonhuman primate models and is likely to facilitate clinical translation of analogous reagents.¹³⁷

Lymphoablation is a necessary part of many tolerance induction protocols in rodents and large animals. Mixed chimerism is a state where the hematopoietic cells in the recipient are made up of both donor and recipient cells as a result of bone marrow transplantation following irradiation.¹³⁸ As a result, graft reactive T cells are negatively selected in the thymus via central tolerance mechanisms. Most importantly, central tolerance can be induced by performing bone marrow transplantation several weeks after solid organ transplantation. The depletion of CD8⁺ memory T cells, that are normally resistant to radiation, is essential for delayed tolerance induction and prolonged graft survival in NHP models of kidney and lung transplantation.^{139–141} In contrast, studies in mouse models of lung transplantation reported that costimulatory blockade induced graft tolerance is dependent upon the rapid graft infiltration by CD8⁺ T_{CM} cells.¹⁴² These data suggest distinct roles memory T cells play in central vs peripheral tolerance and highlight the potential complexities of translating experimental findings to the clinic.

T cell costimulation/coinhibition—Inhibition of CD28/CD80/CD86 costimulatory pathway at the time of antigen exposure is one of the most potent strategies to prevent T cell activation and induce tolerance.^{119,143,144} CTLA4-Ig, a fusion protein that binds to CD80/CD86 with high affinity and thus blocks CD28 engagement, is currently on the market in its second iteration, Belatacept. While Belatacept improves allograft survival rates and minimizes toxic side effects of calcineurin inhibitors,¹⁴⁵ its use is associated with the increased risk of acute cellular rejection,^{146–148} and the mechanisms of Belatacept-resistant rejection are the subject of active investigations. Over the past 2 decades, studies in animal transplant models revealed that memory T cells are more resistant to CTLA4-Ig treatment. Consistent with these findings, Espinosa and colleagues identified CD4⁺CD57⁺ T cells as possible mediators of Belatacept resistant rejection in renal transplant recipients.^{144,149} The T cells associated with rejection episodes are characterized by elevated expression of adhesion molecules LFA-1, VLA-4 and CD2, decreased surface CD28 expression and the capacity to produce high levels of effector cytokines. These findings suggested the potential benefits of simultaneously targeting CD28 costimulation and cellular adhesion molecules. Indeed, treatment with monoclonal antibodies blocking LFA-1 and/or VLA-4 prolonged allograft survival in several experimental models, warranting further investigation and potential clinical translation. Thus either anti-LFA-1 or anti-VLA-4 mAb treatment efficiently inhibited costimulatory-blockade resistant rejection of mouse skin allografts mediated by CD8⁺ memory T cells.^{150–152} Furthermore, study by Setoguchi et al. demonstrated that a short-course peritransplant treatment with anti-LFA-1 antibodies inhibited early infiltration of endogenous alloreactive CD8⁺ memory T cells into murine cardiac allografts resulting in significantly prolonged survival.¹⁵³

The clinical application of strategies targeting another key costimulatory pathway, CD40/CD154, was delayed by thromboembolic effects of early anti-CD154 antibodies.¹⁵⁴ New generation of blocking anti-CD154 and anti-CD40 antibodies have avoided this complication, and promoted acceptance of various allograft types in mice and nonhuman primates.^{155,156,127,157} While interference with CD40/CD154 pathway showed great graft-

prolonging benefits in robust animal transplantation models including pig-to baboon cardiac transplantation,¹⁵⁸ the mechanistic effects of such therapies on reactivation and functions of preexisting donor-reactive memory T cells remain to be determined.

In addition to CD28/B7 and CD40/CD154 costimulation, other costimulatory interactions including ICOS/B7RP-1, CD134/CD134L, CD70/CD27 or CD137/CD137L have been implicated in effector/memory T cell functions. The approaches targeting these pathways showed promise in donor-sensitized recipients, or after delayed administration and have been reviewed elsewhere.^{143,159} Here we would like to discuss a relatively new addition to the list of T cell coinhibitory molecules, lymphocyte-activating gene 3 (LAG-3/CD223). It is expressed following T cell activation, possibly competes with CD4 for MHC class II binding, and through as yet to be elucidated signaling pathway diminishes T cell responses.^{160–163} LAG-3 is also expressed on T regulatory cells and was shown to be key to Treg suppressor function in autoimmunity.^{164,165} Initial studies in transplant models indicate that LAG-3 deficiency in mice results in augmented T cell memory and accelerates cardiac allograft rejection.¹⁶⁶ However, further work is needed to expand on these interesting results and explore the possibility of targeting LAG-3 to ameliorate alloimmune responses. Several reagents targeting LAG-3 activity have been developed and are currently tested in preclinical studies and in clinical trials. One example is a soluble LAG-3 analog that competes with LAG-3 for MHC class II binding and augments immune responses against tumors and viral vaccines.^{167–169} Conversely, a cytolytic chimeric antibody against LAG-3 was shown to deplete LAG-3⁺ activated T cells and inhibit delayed hypersensitivity response in an NHP model.¹⁷⁰ The efficacy of such an approach for selective depletion of effector/memory alloreactive T cells in transplant recipients remains to be tested.

Conclusion

Despite rapidly expanding knowledge on memory T cells as efficient mediators of graft rejection our ability to therapeutically target them in the clinic remains limited. The main challenge is in the very nature of immune memory executed by the variety of cells with broad spectrum of effector functions. Another conceptual problem is that targeting memory T cells based on phenotypic markers lacks specificity and is likely to compromise both pathogenic and protective T cells. Acknowledging anatomical, metabolic and functional diversity of alloreactive memory T cells should identify new points for pharmacological intervention. The novel approaches targeting generation and recall of memory T cells are currently being developed and tested for applications in cancer, vaccines and autoimmune diseases and are waiting to be explored in transplant settings. In combination with modern technologies for investigating basic T cell biology, this opens exciting possibilities of scientific discovery and hopefully, therapeutic advances.

Acknowledgments

The authors receive the following grant support: R01 AI113142 (AV) and P01 AI087586 (RLF and AV) from NIAID.

ABBREVIATIONS PAGE

AMPK	AMP-activated protein kinase
AMR	antibody-mediated rejection
APC	antigen presenting cells
ATG	antithymocyte globulin
DCA	dichloroacetate
EAE	experimental autoimmune encephalitis
EBV	Epstein-Barr virus
GVHD	graft versus host disease
HSV	Herpes simplex virus
HIF-1α	hypoxia-inducible factor 1 α
LAG3	lymphocyte-activating gene 3
LCMV	lymphocytic choriomeningitis virus
MHC	major histocompatibility complex
MHV68	murine gammaherpesvirus 68
mTOR	Mammalian target of rapamycin
MMF	mycophenolate mofetil
NHP	nonhuman primates
NK	natural killer cells
OXPHOS	oxidative phosphorylation
PTLD	posttransplant lymphoproliferative disorders
RA	rheumatoid arthritis
ROS	reactive oxygen species
SCA1	stem cell antigen 1
TAG	triacylglycerols
T_{CM}	central memory cells
TCR	T cell receptor
T_{EFF}	effector T cells
T_{EM}	effector memory T cells

T_{EMRA}	Terminally differentiated effector memory T cells
T_{FH}	follicular T helper cells
T_{PH}	T peripheral helper cells
T_{RM}	tissue resident memory T cells
T_{SCM}	T memory stem cells

references

1. Hartigan CR, Sun H, Ford ML. Memory T-cell exhaustion and tolerance in transplantation. *Immunol Rev.* 2019;292(1):225–242. [PubMed: 31808170]
2. Kieffer TE, Faas MM, Scherjon SA, et al. Pregnancy persistently affects memory T cell populations. *J Reprod Immunol.* 2017;119:1–8. [PubMed: 27863266]
3. Deacock SJ, Lechler RI. Positive correlation of T cell sensitization with frequencies of alloreactive T helper cells in chronic renal failure patients. *Transplantation.* 1992;54(2):338–343. [PubMed: 1386695]
4. Lechler R, Lombardi G. The Structural Basis of Alloreactivity. *Immunol Res.* 1990;9(2):135–146. [PubMed: 2189936]
5. Beura LK, Hamilton SE, Bi K, et al. Normalizing the environment recapitulates adult human immune traits in laboratory mice. *Nature.* 2016;532(7600): 512–516. [PubMed: 27096360]
6. Lombardi G, Sidhu S, Daly M, et al. Are primary alloresponses truly primary? *Int Immunol.* 1990;2(1):9–13. [PubMed: 1708274]
7. Benichou G, Valujskikh A, Heeger PS. Contributions of direct and indirect T cell alloreactivity during allograft rejection in mice. *J Immunol.* 1999;162(1):352–358. [PubMed: 9886406]
8. Amir AL, D'Orsogna LJ, Roelen DL, et al. Allo-HLA reactivity of virus-specific memory T cells is common. *Blood.* 2010;115(15):3146–3157. [PubMed: 20160165]
9. Adams AB, Williams MA, Jones TR, et al. Heterologous immunity provides a potent barrier to transplantation tolerance. *J Clin Invest.* 2003;111(12):1887–1895. [PubMed: 12813024]
10. Beus JM, Hashmi SS, Selvaraj SA, et al. Heterologous immunity triggered by a single, latent virus in *Mus musculus*: combined costimulation- and adhesion- blockade decrease rejection. *PLoS One.* 2013;8(8):e71221.
11. Pantenburg B, Heinzl F, Das L, et al. T cells primed by *Leishmania major* infection cross-react with alloantigens and alter the course of allograft rejection. *J Immunol.* 2002;169(7):3686–3693. [PubMed: 12244161]
12. Valujskikh A, Li XC. Memory T cells and their exhaustive differentiation in allograft tolerance and rejection. *Curr Opin Organ Transplant.* 2012;17(1):15–19. [PubMed: 22186090]
13. Chen Y, Heeger PS, Valujskikh A. In vivo helper functions of alloreactive memory CD4⁺ T cells remain intact despite donor-specific transfusion and anti-CD40 ligand therapy. *J Immunol.* 2004;172(9):5456–5466. [PubMed: 15100287]
14. Zhang Q, Chen Y, Fairchild RL, et al. Lymphoid sequestration of alloreactive memory CD4 T cells promotes cardiac allograft survival. *J Immunol.* 2006;176(2):770–777. [PubMed: 16393960]
15. Gorbacheva V, Fan R, Fairchild RL, et al. Memory CD4 T cells induce antibody-mediated rejection of renal allografts. *J Am Soc Nephrol.* 2016;27(11):3299–3307. [PubMed: 27020853]
16. Su CA, Iida S, Abe T, et al. Endogenous memory CD8 T cells directly mediate cardiac allograft rejection. *Am J Transplant.* 2014;14(3):568–579. [PubMed: 24502272]
17. Tsuda H, Su CA, Tanaka T, et al. Allograft dendritic cell p40 homodimers activate donor-reactive memory CD8⁺ T cells. *JCI Insight.* 2018;3(4):e96940.
18. Sallusto F, Lenig D, Forster R, et al. Two subsets of memory T lymphocytes with distinct homing potentials and effector functions. *Nature.* 1999;401(6754):708–712. [PubMed: 10537110]

19. Gebhardt T, Wakim LM, Eidsmo L, et al. Memory T cells in nonlymphoid tissue that provide enhanced local immunity during infection with herpes simplex virus. *Nat Immunol.* 2009;10(5):524–530. [PubMed: 19305395]
20. Steinbach K, Vincenti I, Merkler D. Resident-memory T cells in tissue-restricted immune responses: for better or worse? *Front Immunol.* 2018;9:2827. [PubMed: 30555489]
21. Beura LK, Rosato PC, Masopust D. Implications of resident memory T cells for transplantation. *Am J Transplant.* 2017;17(5):1167–1175. [PubMed: 27804207]
22. Mackay LK, Braun A, Macleod BL, et al. Cutting edge: CD69 interference with sphingosine-1-phosphate receptor function regulates peripheral T cell retention. *J Immunol.* 2015;194(5):2059–2063. [PubMed: 25624457]
23. Cyster JG, Schwab SR. Sphingosine-1-phosphate and lymphocyte egress from lymphoid organs. *Annu Rev Immunol.* 2012;30:69–94. [PubMed: 22149932]
24. Mackay LK, Rahimpour A, Ma JZ, et al. The developmental pathway for CD103(+)CD8+ tissue-resident memory T cells of skin. *Nat Immunol.* 2013;14(12):1294–1301. [PubMed: 24162776]
25. Bergsbaken T, Bevan MJ. Proinflammatory microenvironments within the intestine regulate the differentiation of tissue-resident CD8(+) T cells responding to infection. *Nat Immunol.* 2015;16(4):406–414. [PubMed: 25706747]
26. Steinert EM, Schenkel JM, Fraser KA, et al. Quantifying memory CD8 T cells reveals regionalization of immunosurveillance. *Cell.* 2015;161(4):737–749. [PubMed: 25957682]
27. Schenkel JM, Fraser KA, Masopust D. Cutting edge: resident memory CD8 T cells occupy frontline niches in secondary lymphoid organs. *J Immunol.* 2014;192(7):2961–2964. [PubMed: 24600038]
28. Fernandez-Ruiz D, Ng WY, Holz LE, et al. Liver-resident memory CD8(+) T cells form a front-line defense against malaria liver-stage infection. *Immunity.* 2016;45(4):889–902. [PubMed: 27692609]
29. Watanabe R, Gehad A, Yang C, et al. Human skin is protected by four functionally and phenotypically discrete populations of resident and recirculating memory T cells. *Sci Transl Med.* 2015;7(279):279ra239.
30. Schenkel JM, Fraser KA, Beura LK, et al. T cell memory. Resident memory CD8 T cells trigger protective innate and adaptive immune responses. *Science.* 2014;346(6205):98–101. [PubMed: 25170049]
31. Cheuk S, Schlums H, Gallais Serezal I, et al. CD49a expression defines tissue-resident CD8(+) T cells poised for cytotoxic function in human skin. *Immunity.* 2017;46(2):287–300. [PubMed: 28214226]
32. Park CO, Kupper TS. The emerging role of resident memory T cells in protective immunity and inflammatory disease. *Nat Med.* 2015;21(7):688–697. [PubMed: 26121195]
33. Carbone FR. Tissue-resident memory T cells and fixed immune surveillance in nonlymphoid organs. *J Immunol.* 2015;195(1):17–22. [PubMed: 26092813]
34. Skon CN, Lee JY, Anderson KG, et al. Transcriptional downregulation of S1pr1 is required for the establishment of resident memory CD8+ T cells. *Nat Immunol.* 2013;14(12):1285–1293. [PubMed: 24162775]
35. Casey KA, Fraser KA, Schenkel JM, et al. Antigen-independent differentiation and maintenance of effector-like resident memory T cells in tissues. *J Immunol.* 2012;188(10):4866–4875. [PubMed: 22504644]
36. Mackay LK, Minnich M, Kragten NA, et al. Hobit and Blimp1 instruct a universal transcriptional program of tissue residency in lymphocytes. *Science.* 2016;352(6284):459–463. [PubMed: 27102484]
37. Jiang X, Clark RA, Liu L, et al. Skin infection generates non-migratory memory CD8+ T(RM) cells providing global skin immunity. *Nature.* 2012;483(7388):227–231. [PubMed: 22388819]
38. Masopust D, Vezys V, Wherry EJ, et al. Cutting edge: gut microenvironment promotes differentiation of a unique memory CD8 T cell population. *J Immunol.* 2006;176(4):2079–2083. [PubMed: 16455963]

39. Goldrath AW, Sivakumar PV, Glaccum M, et al. Cytokine requirements for acute and Basal homeostatic proliferation of naive and memory CD8+ T cells. *J Exp Med.* 2002;195(12):1515–1522. [PubMed: 12070279]
40. DeGottardi MQ, Okoye AA, Vaidya M, et al. Effect of anti-IL-15 administration on T cell and NK cell homeostasis in Rhesus Macaques. *J Immunol.* 2016;197(4):1183–1198. [PubMed: 27430715]
41. Schenkel JM, Fraser KA, Casey KA, et al. IL-15-independent maintenance of tissue-resident and boosted effector memory CD8 T cells. *J Immunol.* 2016;196(9):3920–3926. [PubMed: 27001957]
42. Wu T, Hu Y, Lee YT, et al. Lung-resident memory CD8 T cells (TRM) are indispensable for optimal cross-protection against pulmonary virus infection. *J Leukoc Biol.* 2014;95(2):215–224. [PubMed: 24006506]
43. McMaster SR, Wilson JJ, et al. Airway-resident memory CD8 T cells provide antigen-specific protection against respiratory virus challenge through rapid ifn-gamma production. *J Immunol.* 2015;195(1): 203–209. [PubMed: 26026054]
44. Khanna KM, Bonneau RH, Kinchington PR, et al. Herpes simplex virus-specific memory CD8+ T cells are selectively activated and retained in latently infected sensory ganglia. *Immunity.* 2003;18(5):593–603. [PubMed: 12753737]
45. Lodhi SA, Lamb KE, Meier-Kriesche HU. Solid organ allograft survival improvement in the United States: the long-term does not mirror the dramatic short-term success. *Am J Transplant.* 2011;11(6):1226–1235. [PubMed: 21564524]
46. de Leur K, Dieterich M, Hesselink DA, et al. Characterization of donor and recipient CD8+tissue-resident memory T cells in transplant nephrectomies. *Sci Rep.* 2019;9(10):5984. [PubMed: 30979940]
47. Zhang Y, Ruiz P. Solid organ transplant-associated acute graft-versus-host disease. *Arch Pathol Lab Med.* 2010;134(8):1220–1224. [PubMed: 20670147]
48. Win TS, Rehakova S, Negus MC, et al. Donor CD4 T cells contribute to cardiac allograft vasculopathy by providing help for autoantibody production. *Circ Heart Fail.* 2009;2(4):361–369. [PubMed: 19808360]
49. Zhang Y, Zhao H, Bo L, et al. Total body irradiation of donors can alter the course of tolerance and induce acute rejection in a spontaneous tolerance rat liver transplantation model. *Sci China Life Sci.* 2012;55(9):774–781. [PubMed: 23015125]
50. Snyder ME, Finlayson MO, Connors TJ, et al. Generation and persistence of human tissue-resident memory T cells in lung transplantation. *Sci Immunol.* 2019;4(33):pii: eaav5581.
51. Sallusto F, Geginat J, Lanzavecchia A. Central memory and effector memory T cell subsets: function, generation, and maintenance. *Annu Rev Immunol.* 2004;22:745–763. [PubMed: 15032595]
52. Hamann D, Baars PA, Rep MH, et al. Phenotypic and functional separation of memory and effector human CD8+ T cells. *J Exp Med.* 1997;186(9):1407–1418. [PubMed: 9348298]
53. Geginat J, Lanzavecchia A, Sallusto F. Proliferation and differentiation potential of human CD8+ memory T-cell subsets in response to antigen or homeostatic cytokines. *Blood.* 2003;101(11):4260–4266. [PubMed: 12576317]
54. Willinger T, Freeman T, Hasegawa H, McMichael AJ, Callan MF. Molecular signatures distinguish human central memory from effector memory CD8 T cell subsets. *J Immunol.* 2005;175(9):5895–5903. [PubMed: 16237082]
55. Tilly G, Doan-Ngoc TM, Yap M, et al. IL-15 harnesses pro-inflammatory function of TEMRA CD8 in kidney-transplant recipients. *Front Immunol.* 2017;8:778. [PubMed: 28713392]
56. Thome JJC, Yudanin N, Ohmura Y, et al. Spatial map of human T cell compartmentalization and maintenance over decades of life. *Cell.* 2014;159(4):814–828. [PubMed: 25417158]
57. Rufer N, Zippelius A, Batard P, et al. Ex vivo characterization of human CD8+ T subsets with distinct replicative history and partial effector functions. *Blood.* 2003;102(5):1779–1787. [PubMed: 12750165]
58. Betjes MG, Langerak AW, van der Spek A, et al. Premature aging of circulating T cells in patients with end-stage renal disease. *Kidney Int.* 2011;80(2):208–217. [PubMed: 21525849]

59. Betjes MG, Meijers RW, de Wit EA, et al. Terminally differentiated CD8+ Temra cells are associated with the risk for acute kidney allograft rejection. *Transplantation*. 2012;94(1):63–69. [PubMed: 22691956]
60. Meijers RW, Litjens NH, de Wit EA, et al. Uremia-associated immunological aging is stably imprinted in the T-cell system and not reversed by kidney transplantation. *Transpl Int*. 2014;27(12):1272–1284. [PubMed: 25082296]
61. Yap M, Boeffard F, Clave E, et al. Expansion of highly differentiated cytotoxic terminally differentiated effector memory CD8+ T cells in a subset of clinically stable kidney transplant recipients: a potential marker for late graft dysfunction. *J Am Soc Nephrol*. 2014;25(8):1856–1868. [PubMed: 24652799]
62. Kamburova EG, Koenen HJ, van den Hoogen MW, et al. Longitudinal analysis of T and B cell phenotype and function in renal transplant recipients with or without rituximab induction therapy. *PLoS One*. 2014;9(11):e112658.
63. Kwun J, Manook M, Page E, et al. Crosstalk between T and B cells in the germinal center after transplantation. *Transplantation*. 2017;101(4):704–712. [PubMed: 27906827]
64. Adrianto I, Wen F, Templeton A, et al. Association of a functional variant downstream of TNFAIP3 with systemic lupus erythematosus. *Nat Genet*. 2011;43(3):253–258. [PubMed: 21336280]
65. Hale JS, Ahmed R. Memory T follicular helper CD4 T cells. *Front Immunol*. 2015;6:16. [PubMed: 25699040]
66. Weber JP, Fuhrmann F, Hutloff A. T-follicular helper cells survive as long-term memory cells. *Eur J Immunol*. 2012;42(8):1981–1988. [PubMed: 22730020]
67. Macleod MK, David A, McKee AS, et al. Memory CD4 T cells that express CXCR5 provide accelerated help to B cells. *J Immunol*. 2011;186(5):2889–2896. [PubMed: 21270407]
68. Hale JS, Youngblood B, Latner DR, et al. Distinct memory cd4(+) t cells with commitment to t follicular helper- and t helper 1-cell lineages are generated after acute viral infection. *Immunity*. 2013;38(4):805–817. [PubMed: 23583644]
69. De Bruyne R, Bogaert D, De Ruyck N, et al. Calcineurin inhibitors dampen humoral immunity by acting directly on naive B cells. *Clin Exp Immunol*. 2015;180(3):542–550. [PubMed: 25682989]
70. Wallin EF, Hill DL, Linterman MA, et al. The calcineurin inhibitor tacrolimus specifically suppresses human T follicular helper cells. *Front Immunol*. 2018;9:1184. [PubMed: 29904381]
71. de Graav GN, Hesselink DA, Dieterich M, et al. Belatacept does not inhibit follicular T cell-dependent B-cell differentiation in kidney transplantation. *Front Immunol*. 2017;8:641. [PubMed: 28620390]
72. Yan L, de Leur K, Hendriks RW, et al. T follicular helper cells as a new target for immunosuppressive therapies. *Front Immunol*. 2017;8:1510. [PubMed: 29163552]
73. Rao DA, Gurish MF, Marshall JL, et al. Pathologically expanded peripheral T helper cell subset drives B cells in rheumatoid arthritis. *Nature*. 2017;542(7639):110–114. [PubMed: 28150777]
74. Rao D, Bocharnikov A, Fonseka C, et al. T peripheral helper cells are expanded in the circulation of active SLE patients and correlate with CD21low B cells. *Arthritis Rheumatol*. 2018;70.
75. Bocharnikov AV, Keegan J, Wacleche VS, et al. PD-1hiCXCR5- T peripheral helper cells promote B cell responses in lupus via MAF and IL-21. *JCI Insight*. 2019;4(20):pii: 130062.
76. Pallikkuth S, de Armas LR, Rinaldi S, et al. Dysfunctional peripheral T follicular helper cells dominate in people with impaired influenza vaccine responses: Results from the FLORAH study. *PLoS Biol*. 2019;17(5):e3000257.
77. Thauan O, Field AC, Dai J, et al. Lymphoid neogenesis in chronic rejection: evidence for a local humoral alloimmune response. *Proc Natl Acad Sci U S A*. 2005;102(41):14723–14728.
78. Huibers MM, Gareau AJ, Beerthuijzen JM, et al. Donor-specific antibodies are produced locally in ectopic lymphoid structures in cardiac allografts. *Am J Transplant*. 2017;17(1):246–254. [PubMed: 27428759]
79. Deteix C, Attuyl-Audenis V, Duthey A, et al. Intragraft Th17 infiltrate promotes lymphoid neogenesis and hastens clinical chronic rejection. *J Immunol*. 2010;184(9):5344–5351. [PubMed: 20357253]
80. Zhang Y, Joe G, Hexner E, et al. Host-reactive CD8+ memory stem cells in graft-versus-host disease. *Nat Med*. 2005;11(12):1299–1305. [PubMed: 16288282]

81. Gattinoni L, Speiser DE, Lichterfeld M, et al. T memory stem cells in health and disease. *Nat Med.* 2017;23(1):18–27. [PubMed: 28060797]
82. Lugli E, Gattinoni L, Roberto A, et al. Identification, isolation and in vitro expansion of human and nonhuman primate T stem cell memory cells. *Nat Protoc.* 2013;8(1):33–42. [PubMed: 2322456]
83. Im SJ, Hashimoto M, Gerner MY, et al. Defining CD8+ T cells that provide the proliferative burst after PD-1 therapy. *Nature.* 2016;537(7620):417–421. [PubMed: 27501248]
84. Vigano S, Negron J, Ouyang Z, et al. Prolonged antiretroviral therapy preserves HIV-1-specific CD8 T cells with stem cell-like properties. *J Virol.* 2015;89(15):7829–7840. [PubMed: 25995260]
85. Ribeiro SP, Milush JM, Cunha-Neto E, et al. The CD8(+) memory stem T cell (T(SCM)) subset is associated with improved prognosis in chronic HIV-1 infection. *J Virol.* 2014;88(23):13836–13844.
86. Mateus J, Lasso P, Pavia P, et al. Low frequency of circulating CD8+ T stem cell memory cells in chronic chagasic patients with severe forms of the disease. *PLoS Negl Trop Dis.* 2015;9(1):e3432.
87. Hosokawa K, Muranski P, Feng X, et al. Memory stem T cells in autoimmune disease: high frequency of circulating CD8+ memory stem cells in acquired aplastic anemia. *J Immunol.* 2016;196(4):1568–1578. [PubMed: 26764034]
88. O'Sullivan D, Pearce EL. Targeting T cell metabolism for therapy. *Trends Immunol.* 2015;36(2):71–80. [PubMed: 25601541]
89. Fox CJ, Hammerman PS, Thompson CB. Fuel feeds function: energy metabolism and the T-cell response. *Nat Rev Immunol.* 2005;5(11): 844–852. [PubMed: 16239903]
90. Yusuf I, Fruman DA. Regulation of quiescence in lymphocytes. *Trends Immunol.* 2003;24(7):380–386. [PubMed: 12860529]
91. van der Windt GJ, Pearce EL. Metabolic switching and fuel choice during T-cell differentiation and memory development. *Immunol Rev.* 2012;249(1):27–42. [PubMed: 22889213]
92. Sena LA, Li S, Jairaman A, et al. Mitochondria are required for antigen-specific T cell activation through reactive oxygen species signaling. *Immunity.* 2013;38(2):225–236. [PubMed: 23415911]
93. Michalek RD, Gerriets VA, Jacobs SR, et al. Cutting edge: distinct glycolytic and lipid oxidative metabolic programs are essential for effector and regulatory CD4+ T cell subsets. *J Immunol.* 2011;186(6):3299–3303. [PubMed: 21317389]
94. Macintyre AN, Rathmell JC. Activated lymphocytes as a metabolic model for carcinogenesis. *Cancer Metab.* 2013;1(1):5. [PubMed: 24280044]
95. Chang CH, Curtis JD, Maggi LB, Jr., et al. Posttranscriptional control of T cell effector function by aerobic glycolysis. *Cell.* 2013;153(6):1239–1251. [PubMed: 23746840]
96. Jacobs SR, Herman CE, MacIver NJ, et al. Glucose uptake is limiting in T cell activation and requires CD28-mediated akt-dependent and independent pathways. *J Immunol.* 2008;180(7):4476–4486. [PubMed: 18354169]
97. Laplante M, Sabatini DM. mTOR Signaling in Growth Control and Disease. *Cell.* 2012;149(2):274–293. [PubMed: 22500797]
98. Powell JD, Pollizzi KN, Heikamp EB, et al. Regulation of immune responses by mTOR. *Ann Rev Immunol.* 2012;30:39–68. [PubMed: 22136167]
99. Doedens AL, Phan AT, Stradner MH, et al. Hypoxia-inducible factors enhance the effector responses of CD8(+) T cells to persistent antigen. *Nat Immunol.* 2013;14(11):1173–1182. [PubMed: 24076634]
100. Kim JW, Tchernyshyov I, Semenza GL, et al. HIF-1-mediated expression of pyruvate dehydrogenase kinase: A metabolic switch required for cellular adaptation to hypoxia. *Cell Metab.* 2006;3(3):177–185. [PubMed: 16517405]
101. Tamas P, Hawley SA, Clarke RG, et al. Regulation of the energy sensor AMP-activated protein kinase by antigen receptor and Ca²⁺ in T lymphocytes. *J Exp Med.* 2006;203(7):1665–1670. [PubMed: 16818670]
102. Rolf J, Zarrouk M, Finlay DK, et al. AMPK alpha 1: A glucose sensor that controls CD8 T-cell memory. *Eur J Immunol.* 2013;43(4):889–896. [PubMed: 23310952]

103. O'Sullivan D, van der Windt GJW, Huang SCC, et al. Memory CD8(+) T cells use cell-intrinsic lipolysis to support the metabolic programming necessary for development. *Immunity*. 2014;41(1) 75–88. [PubMed: 25001241]
104. Cui G, Staron MM, Gray SM, et al. IL-7-induced glycerol transport and TAG synthesis promotes memory CD8+ T cell longevity. *Cell*. 2015;161(4):750–761. [PubMed: 25957683]
105. Ayasoufi K, Kohei N, Nicosia M, et al. Aquaporin 4 blockade improves survival of murine heart allografts subjected to prolonged cold ischemia. *Am J Transplant*. 2018;18(5):1238–1246. [PubMed: 29243390]
106. Nicosia M, Miyairi S, Beavers A, et al. Aquaporin 4 inhibition alters chemokine receptor expression and T cell trafficking. *Sci Rep*. 2019;9(1):7417. [PubMed: 31092872]
107. Buck MD, O'Sullivan D, Klein Geltink RI, et al. Mitochondrial Dynamics Controls T Cell Fate through Metabolic Programming. *Cell*. 2016;166(1): 63–76. [PubMed: 27293185]
108. Phan AT, Doedens AL, Palazon A, et al. Constitutive glycolytic metabolism supports CD8(+) T cell effector memory differentiation during viral infection. *Immunity*. 2016;45(5):1024–1037. [PubMed: 27836431]
109. Sukumar M, Liu J, Ji Y, et al. Inhibiting glycolytic metabolism enhances CD8+ T cell memory and antitumor function. *J Clin Invest*. 2013;123(10):4479–4488. [PubMed: 24091329]
110. Ohashi T, Akazawa T, Aoki M, et al. Dichloroacetate improves immune dysfunction caused by tumor-secreted lactic acid and increases antitumor immunoreactivity. *Int J Cancer*. 2013;133(5):1107–1118. [PubMed: 23420584]
111. Caza TN, Talaber G, Perl A. Metabolic regulation of organelle homeostasis in lupus T cells. *Clin Immunol*. 2012;144(3):200–213. [PubMed: 22836085]
112. Caza TN, Fernandez DR, Talaber G, et al. HRES-1/Rab4-mediated depletion of Drp1 impairs mitochondrial homeostasis and represents a target for treatment in SLE. *Ann Rheum Dis*. 2014;73(10):1888–1897. [PubMed: 23897774]
113. Gerriets VA, Kishton RJ, Nichols AG, et al. Metabolic programming and PDHK1 control CD4+ T cell subsets and inflammation. *J Clin Invest*. 2015;125(1):194–207. [PubMed: 25437876]
114. Pan Y, Kupper TS. Metabolic reprogramming and longevity of tissue-resident memory T cells. *Front Immunol*. 2018;9:1347. [PubMed: 29967608]
115. Pan Y, Tian T, Park CO, et al. Survival of tissue-resident memory T cells requires exogenous lipid uptake and metabolism. *Nature*. 2017;543(7644):252–256. [PubMed: 28219080]
116. Gatza E, Wahl DR, Oipari AW, et al. Manipulating the bioenergetics of alloreactive T cells causes their selective apoptosis and arrests graft-versus-host disease. *Sci Transl Med*. 2011;3(67):67ra68.
117. Glick GD, Rossignol R, Lyssiotis CA, et al. Anaplerotic metabolism of alloreactive T cells provides a metabolic approach to treat graft-versus-host disease. *J Pharmacol Exp Ther*. 2014;351(2):298–307. [PubMed: 25125579]
118. Lee CF, Lo YC, Cheng CH, et al. Preventing allograft rejection by targeting immune metabolism. *Cell Rep*. 2015;13(4):760–770. [PubMed: 26489460]
119. Nicosia M, Valujskikh A. Total recall: can we reshape T cell memory by lymphoablation? *Am J Transplant*. 2017;17(7):1713–1718. [PubMed: 27888576]
120. Fehr T, Sykes M. Tolerance induction in clinical transplantation. *Transpl Immunol*. 2004;13(2):117–130. [PubMed: 15380542]
121. Golshayan D, Pascual M. Tolerance-inducing immunosuppressive strategies in clinical transplantation: an overview. *Drugs*. 2008;68(15):2113–2130. [PubMed: 18840003]
122. Haudebourg T, Poirier N, Vanhove B. Depleting T-cell subpopulations in organ transplantation. *Transpl Int*. 2009;22(5):509–518. [PubMed: 18980625]
123. Pearl JP, Parris J, Hale DA, et al. Immunocompetent T-cells with a memory-like phenotype are the dominant cell type following antibody-mediated T-cell depletion. *Am J Transplant*. 2005;5(3):465–474. [PubMed: 15707400]
124. Ayasoufi K, Fan R, Fairchild RL, et al. CD4 T cell help via B cells is required for lymphopenia-induced CD8 T cell proliferation. *J Immunol*. 2016;196(7):3180–3190. [PubMed: 26912319]

125. Ayasoufi K, Fan R, Fairchild RL, et al. CD4 T cell help via B cells is required for lymphopenia-induced CD8 T cell proliferation. *J Immunol.* 2016;196(7):3180–3190. [PubMed: 26912319]
126. Ezzelarab MB, Eksler B, Echeverri G, et al. Costimulation blockade in pig artery patch xenotransplantation - a simple model to monitor the adaptive immune response in nonhuman primates. *Xenotransplantation.* 2012;19(4):221–232. [PubMed: 22909135]
127. Kim EJ, Kwun J, Gibby AC, et al. Costimulation blockade alters germinal center responses and prevents antibody-mediated rejection. *Am J Transplant.* 2014;14(1):59–69. [PubMed: 24354871]
128. Chamian F, Lin SL, Lee E, et al. Alefacept (anti-CD2) causes a selective reduction in circulating effector memory T cells (Tem) and relative preservation of central memory T cells (Tcm) in psoriasis. *J Transl Med.* 2007;5:27. [PubMed: 17555598]
129. Chamian F, Lowes MA, Lin SL, et al. Alefacept reduces infiltrating T cells, activated dendritic cells, and inflammatory genes in psoriasis vulgaris. *Proc Natl Acad Sci U S A.* 2005;102(6):2075–2080. [PubMed: 15671179]
130. Larsen R, Ryder LP, Svejgaard A, et al. Changes in circulating lymphocyte subpopulations following administration of the leucocyte function-associated antigen-3 (LFA-3)/IgG1 fusion protein alefacept. *Clin Exp Immunol.* 2007;149(1):23–30. [PubMed: 17403057]
131. Weaver TA, Charafeddine AH, Agarwal A, et al. Alefacept promotes co-stimulation blockade based allograft survival in nonhuman primates. *Nat Med.* 2009;15(7):746–749. [PubMed: 19584865]
132. Shapira MY, Abdul-Hai A, Resnick IB, et al. Alefacept treatment for refractory chronic extensive GVHD. *Bone Marrow Transplant.* 2009;43(4):339–343. [PubMed: 18850020]
133. Mourad M, Besse T, Malaise J, et al. BTI-322 for acute rejection after renal transplantation. *Transplant Proc.* 1997;29(5):2353. [PubMed: 9270759]
134. Pruett TL, McGory RW, Wright FH, et al. Safety profile, pharmacokinetics, and pharmacodynamics of sipilizumab, a humanized anti-CD2 monoclonal antibody, in renal allograft recipients. *Transplant Proc.* 2009;41(9):3655–3661. [PubMed: 19917362]
135. Kawai T, Cosimi AB, Spitzer TR, et al. HLA-mismatched renal transplantation without maintenance immunosuppression. *N Engl J Med.* 2008;358(4):353–361. [PubMed: 18216355]
136. Kawai T, Sachs DH, Sprangers B, et al. Long-term results in recipients of combined HLA-mismatched kidney and bone marrow transplantation without maintenance immunosuppression. *Am J Transplant.* 2014;14(7):1599–1611. [PubMed: 24903438]
137. Berglund E, Alonso-Guallart P, Danton M, et al. Safety and pharmacodynamics of anti-CD2 monoclonal antibody treatment in cynomolgus macaques - an experimental study. *Transpl Int.* 2020;33(1):98–107. [PubMed: 31523849]
138. Zuber J, Sykes M. Mechanisms of mixed chimerism-based transplant tolerance. *Trends Immunol.* 2017;38(11):829–843. [PubMed: 28826941]
139. Aoyama A, Tonsho M, Ng CY, et al. Long-term lung transplantation in nonhuman primates. *Am J Transplant.* 2015;15(5):1415–1420. [PubMed: 25772308]
140. Tonsho M, Lee S, Aoyama A, et al. Tolerance of lung allografts achieved in nonhuman primates via mixed hematopoietic chimerism. *Am J Transplant.* 2015;15(8):2231–2239. [PubMed: 25904524]
141. Koyama I, Nadazdin O, Boskovic S, et al. Depletion of CD8 memory T cells for induction of tolerance of a previously transplanted kidney allograft. *Am J Transplant.* 2007;7(5):1055–1061. [PubMed: 17286617]
142. Krupnick AS, Lin X, Li W, et al. Central memory CD8+ T lymphocytes mediate lung allograft acceptance. *J Clin Invest.* 2014;124(3):1130–1143. [PubMed: 24569377]
143. Benichou G, Gonzalez B, Marino J, et al. Role of memory t cells in allograft rejection and tolerance. *Front Immunol.* 2017;8:170. [PubMed: 28293238]
144. Espinosa JR, Samy KP, Kirk AD. Memory T cells in organ transplantation: progress and challenges. *Nat Rev Nephrol.* 2016;12(6):339–347. [PubMed: 26923209]
145. Larsen CP, Pearson TC, Adams AB, et al. Rational development of LEA29Y (belatacept), a high-affinity variant of CTLA4-Ig with potent immunosuppressive properties. *Am J Transplant.* 2005;5(3):443–453. [PubMed: 15707398]

146. Rostaing L, Vincenti F, Grinyo J, et al. Long-term belatacept exposure maintains efficacy and safety at 5 years: results from the long-term extension of the BENEFIT study. *Am J Transplant.* 2013;13(11):2875–2883. [PubMed: 24047110]
147. Vincenti F, Charpentier B, Vanrenterghem Y, et al. A phase III study of belatacept-based immunosuppression regimens versus cyclosporine in renal transplant recipients (BENEFIT study). *Am J Transplant.* 2010;10(3):535–546. [PubMed: 20415897]
148. Vincenti F, Larsen CP, Alberu J, et al. Three-year outcomes from BENEFIT, a randomized, active-controlled, parallel-group study in adult kidney transplant recipients. *Am J Transplant.* 2012;12(1):210–217. [PubMed: 21992533]
149. Espinosa J, Herr F, Tharp G, et al. CD57(+) CD4 T cells underlie belatacept-resistant allograft rejection. *Am J Transplant.* 2016;16(4):1102–1112. [PubMed: 26603381]
150. Kitchens WH, Larsen CP, Ford ML. Integrin antagonists for transplant immunosuppression: panacea or peril? *Immunotherapy.* 2011;3(3):305–307. [PubMed: 21395371]
151. Kitchens WH, Haridas D, Wagener ME, et al. Integrin antagonists prevent costimulatory blockade-resistant transplant rejection by CD8(+) memory T cells. *Am J Transplant.* 2012;12(1):69–80. [PubMed: 21942986]
152. Isobe M, Suzuki J, Yamazaki S, et al. Acceptance of primary skin graft after treatment with anti-intercellular adhesion molecule-1 and anti-leukocyte function-associated antigen-1 monoclonal antibodies in mice. *Transplantation.* 1996;62(3):411–413. [PubMed: 8779692]
153. Setoguchi K, Schenk AD, Ishii D, et al. LFA-1 antagonism inhibits early infiltration of endogenous memory CD8 T cells into cardiac allografts and donor-reactive T cell priming. *Am J Transplant.* 2011;11(5):923–935. [PubMed: 21466654]
154. Koyama I, Kawai T, Andrews D, et al. Thrombophilia associated with anti-CD154 monoclonal antibody treatment and its prophylaxis in nonhuman primates. *Transplantation.* 2004;77(3):460–462. [PubMed: 14966427]
155. Adams AB, Shirasugi N, Jones TR, et al. Development of a chimeric anti-CD40 monoclonal antibody that synergizes with LEA29Y to prolong islet allograft survival. *J Immunol.* 2005;174(1):542–550. [PubMed: 15611281]
156. Badell IR, Thompson PW, Turner AP, et al. Nondepleting anti-CD40-based therapy prolongs allograft survival in nonhuman primates. *Am J Transplant.* 2012;12(1):126–135. [PubMed: 21920020]
157. Pascual J, Pirsch JD, Odorico JS, et al. Alemtuzumab induction and antibody-mediated kidney rejection after simultaneous pancreas-kidney transplantation. *Transplantation.* 2009;87(1):125–132. [PubMed: 19136902]
158. Mohiuddin MM, Singh AK, Corcoran PC, et al. Role of anti-CD40 antibody-mediated costimulation blockade on non-Gal antibody production and heterotopic cardiac xenograft survival in a GTKO.hCD46Tg pig-to-baboon model. *Xenotransplantation.* 2014;21(1):35–45. [PubMed: 24164510]
159. Valujskikh A The challenge of inhibiting alloreactive T-cell memory. *Am J Transplant.* 2006;6(4):647–651. [PubMed: 16539619]
160. Triebel F, Jitsukawa S, Baixeras E, et al. LAG-3, a novel lymphocyte activation gene closely related to CD4. *J Exp Med.* 1990;171(5):1393–1405. [PubMed: 1692078]
161. Workman CJ, Cauley LS, Kim IJ, et al. Lymphocyte activation gene-3 (CD223) regulates the size of the expanding T cell population following antigen activation in vivo. *J Immunol.* 2004;172(9):5450–5455. [PubMed: 15100286]
162. Macon-Lemaître L, Triebel F. The negative regulatory function of the lymphocyte-activation gene-3 co-receptor (CD223) on human T cells. *Immunology.* 2005;115(2):170–178. [PubMed: 15885122]
163. Maeda TK, Sugiura D, Okazaki IM, et al. Atypical motifs in the cytoplasmic region of the inhibitory immune co-receptor LAG-3 inhibit T cell activation. *J Biol Chem.* 2019;294(15):6017–6026. [PubMed: 30760527]
164. Do JS, Visperas A, Sanogo YO, et al. An IL-27/Lag3 axis enhances Foxp3+ regulatory T cell-suppressive function and therapeutic efficacy. *Mucosal Immunol.* 2016;9(1):137–145. [PubMed: 26013006]

165. Kim D, Le HT, Nguyen QT, et al. Cutting edge: IL-27 attenuates autoimmune neuroinflammation via regulatory T cell/Lag3-dependent but IL-10-independent mechanisms in vivo. *J Immunol.* 2019;202(6):1680–1685. [PubMed: 30700587]
166. Erfe J, Yang C, Ndishabandi D, et al. Deleting LAG-3 Accelerates cardiac allograft rejection and augments T cell memory in mice. *Am J Transplant.* 2017;17:516–517.
167. Brignone C, Grygar C, Marcu M, et al. IMP321 (sLAG-3), an immunopotentiator for T cell responses against a HBsAg antigen in healthy adults: a single blind randomised controlled phase I study. *J Immune Based Ther Vaccines.* 2007;5:5. [PubMed: 17394654]
168. Brignone C, Grygar C, Marcu M, et al. IMP321 (sLAG-3) safety and T cell response potentiation using an influenza vaccine as a model antigen: A single-blind phase I study. *Vaccine.* 2007;25(24):4641–4650. [PubMed: 17493710]
169. Brignone C, Escudier B, Grygar C, et al. A phase I pharmacokinetic and biological correlative study of IMP321, a novel MHC class II agonist, in patients with advanced renal cell carcinoma. *Clin Cancer Res.* 2009;15(19):6225–6231. [PubMed: 19755389]
170. Poirier N, Haudebourg T, Brignone C, et al. Antibody-mediated depletion of lymphocyte-activation gene-3 (LAG-3(+)) -activated T lymphocytes prevents delayed-type hypersensitivity in non-human primates. *Clin Exp Immunol.* 2011;164(2):265–274. [PubMed: 21352204]
171. Ahmadzadeh M, Hussain SF, Farber DL. Heterogeneity of the memory CD4 T cell response: persisting effectors and resting memory T cells. *J Immunol.* 2001;166(2):926–935. [PubMed: 11145669]
172. Sathaliyawala T, Kubota M, Yudanin N, et al. Distribution and compartmentalization of human circulating and tissue-resident memory T cell subsets. *Immunity.* 2013;38(1):187–197. [PubMed: 23260195]
173. Surh CD, Sprent J. Homeostasis of naive and memory T cells. *Immunity.* 2008;29(6):848–862. [PubMed: 19100699]
174. Larbi A, Fulop T. From “Truly Naive” to “Exhausted Senescent” T cells: when markers predict functionality. *Cytometry A.* 2014;85(1):25–35. [PubMed: 24124072]
175. Schenkel JM, Masopust D. Tissue-resident memory T cells. *Immunity.* 2014;41(6): 886–897. [PubMed: 25526304]
176. Meli AP, King IL. Identification of mouse T follicular helper cells by flow cytometry. *Methods Mol Biol.* 2015;1291:3–11. [PubMed: 25836297]

Table 1.

Memory T cell subsets and the surface markers used experimentally to define them.

SUBSET	HUMAN MARKERS	MOUSE MARKERS	COMMON MARKERS	REFS
NAIVE	CD45RA ^{hi} CD45RO ^{lo}	CD44 ^{lo}	CD62L ^{hi} CCR7 ^{hi} CD28 ^{hi}	143
CENTRAL MEMORY (T _{CM})	CD45RA ^{lo} CD45RO ^{hi}	CD44 ^{hi}	CD62L ^{hi} CCR7 ^{hi} CD28 ^{hi}	18,51,171,172
EFFECTOR MEMORY (T _{EM})	CD45RA ^{lo} CD45RO ^{hi}	CD44 ^{hi}	CD62L ^{lo} CCR7 ^{lo} CD28 ^{hi}	18,51,171,172
TERMINALLY DIFFERENTIATED EFFECTOR MEMORY (T _{EMRA})	CD45RA ^{hi} CD45RO ^{lo} CD62L ^{lo} CCR7 ^{lo} CD28 ^{lo} CD57 ^{hi} KLRG1 ^{hi}			172–174
RESIDENT MEMORY (T _{RM})	CD45RA ^{lo} CD45RO ^{hi}	CD44 ^{hi}	CD62L ^{lo} CCR7 ^{lo} CD103 ^{hi} CD69 ^{hi} S1PR1 ^{lo}	20,171,175
T FOLLICULAR HELPER (T _{FH})			CD4 ⁺ CXCR5 ⁺ PD-1 ^{hi} CD40, ICOS, IL-21	63,65,176
T PERIPHERAL HELPER (T _{PH})			CD4 ⁺ CXCR5 ⁻ PD-1 ^{hi} CD40, ICOS, IL-21	73
T MEMORY STEM CELL (T _{SCM})	SCA1 IL-2Rb CXCR3 CD45RA ⁺ CD62L ⁺ CD58 CD11a CD95			80–82