HHS Public Access

Author manuscript

Leuk Lymphoma. Author manuscript; available in PMC 2019 September 27.

Published in final edited form as:

Leuk Lymphoma. 2019 March ; 60(3): 598–609. doi:10.1080/10428194.2018.1485905.

TCR αβ+/CD19+ cell depletion in haploidentical hematopoietic allogeneic stem cell transplantation: a review of current data

Kieran Sahasrabudhe[a], **Mario Otto**[b,c], **Peiman Hematti**[a,c], **Vaishalee Kenkre**[a,c]

[a]Department of Medicine, School of Medicine and Public Health, University of Wisconsin, Madison, WI, USA

[b]Division of Pediatric Hematology Oncology, and Bone Marrow Transplant, Department of Pediatrics, School of Medicine and Public Health, University of Wisconsin, Madison, WI, USA

[c]University of Wisconsin Carbone Cancer Center, Madison, WI, USA

Abstract

Allogeneic hematopoietic stem cell transplantation is a curative option for patients with a variety of diseases. Transplantation from a related haploidentical donor is being increasingly utilized for patients who lack an available human leukocyte antigen matched related or unrelated donor. One of the strategies used for haploidentical transplants involves selective depletion of T cells expressing the αβ T cell receptor and CD19+ B cells prior to transplant. This allows for the removal of cells responsible for graft-versus-host disease and post-transplant lymphoproliferative disorder but maintains hematopoietic progenitor and stem cells for engraftment (CD34+ cells), as well as cells to elicit graft-versus-tumor effect and provide anti-infective activity (such as gamma-delta T cells and natural killer cells). The aim of this review article is to present and discuss the data available to date from studies utilizing this method of transplantation.

Keywords

Graft-versus-host disease; graft rejection; clinical results; infectious complications; TCR αβ+/CD19+ depletion

Introduction

Allogeneic hematopoietic stem cell transplantation (HSCT) has become a curative option for patients with a variety of otherwise incurable diseases such as high-risk leukemias and recurrent lymphomas, as well as many nonmalignant conditions, such as severe combined immunodeficiencies (SCIDs). In 2015, approximately 8700 allogeneic transplants were performed in the US alone [1]. Unfortunately, only about one in four patients who need an allogeneic HSCT will have a human leukocyte antigen (HLA) matched sibling available to serve as the stem cell donor [2]. It has nevertheless become feasible to identify a suitable

CONTACT Vaishalee Kenkre, vpkenkre@medicine.wisc.edu, University of Wisconsin, 4037 WIMR, 1111 Highland Ave, Madison 53705, WI, USA.

Potential conflict of interest: Disclosure forms provided by the authors are available with the full text of this article online at http://dx.doi.org/10.1080/10428194.2018.1485905.

matched unrelated allogeneic donor for most of these patients [3]. However, donor search, confirmatory testing, donor evaluation, and final selection are processes which often take weeks to months. This can be too long for many patients with aggressive diseases, and it is not uncommon to miss the window of opportunity to achieve cure.

Transplantation from a haploidentical donor (most commonly a parent or child of a patient, but sometimes a sibling) has some unrivaled advantages. First, donors are highly motivated and readily available. Second, haplo-HSCT allows the selection of a donor with the highest degree of mismatch in their natural killer cell immunoglobulin-like receptor (KIR) repertoire, which can lead to better graft-versus-tumor effect [4]. Most haploidentical transplantation strategies require profound T cell depletion of the graft to minimize the risk for graft-versus-host disease (GVHD). Due to the technological progress made in graft engineering technologies and the ensuing very low numbers of remaining T cells in the graft, GVHD prophylaxis is often not necessary for a prolonged period of time in the subset of patients who receive haploidentical transplants involving T cell depletion. In some trials, GVHD prophylaxis has been entirely and safely omitted. Here, we present a summary of the evolution of a newer such strategy, TCR αβ+ and CD 19+ depletion from stem cell grafts, and a summary of the current literature. Key data are also summarized in Table 1.

Evolution of the methodology for TCR αβ+ and CD 19+ depletion

Various strategies have been utilized to increase the success rate of haploidentical allo-HSCT. Early strategies involved transplanting only CD34+ selected hematopoietic stem cells [5,6]. This allowed for the indirect removal of T cells responsible for GVHD, and for the indirect removal of B cells, which can cause post-transplant lymphoproliferative disorder (PTLD). There has also been evidence that B cells may play a role in GVHD pathogenesis, and the removal of B cells may therefore reduce the risk of GVHD as well [7–9]. In one strategy, ‘megadoses’ of mobilized peripheral blood CD34+ cells were administered [6]. In another approach, recipients were given bone marrow-derived CD34+ stem cells in addition to CD34+ peripherally-derived stem cells [5]. The CD34+ cells in these studies were selected using a soybean agglutinin and E-rosetting based technique.

Newer CD34+ cell selection techniques were subsequently introduced including the magnetic activated cell sorting (MACS) device and later the automated cliniMACS device, which allowed for more effective and efficient selection [10,11]. For these transplants, recipients underwent highly intensive conditioning regimens in order to facilitate engraftment and prevent rejection, which successfully facilitated engraftment and prevented GVHD. Studies in acute leukemia, using total body irradiation (TBI), thiotepa, fludarabine, and anti-thymocyte globulin (ATG) have shown engraftment rates of 93–95% and acute and chronic GVHD (cGVHD) ranging from 0 to 20% and 0 to 4%, respectively [5,6]. Despite the favorable rates of engraftment and GVHD, this type of transplant was associated with significant transplant-related mortality (TRM) of around 40%, in part due to the high intensity conditioning regimen, and in part due to delayed immune reconstitution caused by removal of lymphoid cells and committed hematopoietic progenitors, leading to opportunistic infections. The requirement for ‘megadoses’ of stem cells also represented a challenge [12].

Earlier investigations in the allogeneic HSCT setting had demonstrated several beneficial actions of natural killer (NK) cells in grafts including facilitating engraftment, graft-versus-tumor (GVT) effects, and activity against opportunistic infection [13–17]. The studies utilizing CD34+ stem cells, in the setting of haploidentical allo-HSCT, also led to the concept of NK cell alloreactivity. Tumor cell lysis by NK cells can be mediated by mismatch of donor NK cell killer immunoglobulin-like receptors (KIRs) and KIR ligands on recipient cells. This facilitates a graft versus leukemia effect. This phenomenon has been most pronounced in adult patients with acute myeloid leukemia (AML) but has also been reported in pediatric acute lymphoblastic leukemia (ALL) [14,18].

After the initial studies utilizing purified CD34+ cells, a subsequent transplant strategy was developed that involved selectively depleting CD3 T cells (±CD19 B cells) from the graft, using immunomagnetic beads. The goal of this method was to eliminate T cells responsible for GVHD and B cells responsible for GVHD and PTLD, while maintaining CD34+ cells in addition to NK cells and other cells (including monocytes, granulocytes, and antigen presenting cells) with the potential to facilitate engraftment and provide immunologically favorable effects. Hale et al. studied this method of transplantation in 20 pediatric patients with ALL, AML, myelodysplastic syndrome (MDS), chronic myeloid leukemia (CML), or non-Hodgkin lymphoma (NHL) using a conditioning regimen of TBI, cyclophosphamide, thiotepa, and rabbit ATG [19]. Three subsequent trials were completed in patients with various hematologic malignancies using a reduced intensity conditioning regimen consisting of fludarabine, melphalan, thiotepa, and OKT3 [20–22]. Rates of engraftment were favorable in these studies ranging from 83% to 97% [20–22]. In the trial conducted by Hale et al., the rate of acute GVHD (aGVHD) was 16% (all grade I–II) with the rate of cGVHD being 5.2%. All of the cGVHD was limited stage [19]. The trials involving the reduced intensity conditioning regimens had higher rates of aGVHD, though mostly lower grade acute (grade I–II GVHD ranged from 24 to 39%, with grade III–IV GVHD seen less often (3–14%)). In these studies, the rate of cGVHD ranged from 10 to 28%. The rate of TRM in the study conducted by Handgretinger et al. was low at 2.6% [21]. However, TRM remained significant in the other studies ranging from 20 to 30% [19,20,22].

More recent studies have focused on the role of specific T cell subsets in the graft to further improve the antileukemic/anti-tumor effect of the graft while decreasing the rate of GVHD. The majority of peripheral T cells express the αβ T cell receptor and are the main drivers of GVHD [23,24]. In contrast, the subset of T cells that express the γδ T cell receptor recognize their targets in a major histocompatibility complex (MHC)-independent manner. They do not recognize allo-antigens and therefore do not contribute to GVHD. They have also been shown to have cytotoxic activity against both solid tumor and hematologic malignancies *in vitro* [25]. Studies have found a positive correlation between increased numbers of donor-derived γδ T cells and survival in patients undergoing haploidentical allo-HSCT or partially mismatched allo-HSCT [26,27]. Findings such as these have led to the development of a new method of haploidentical allo-HSCT involving the selective depletion of αβ T cells and CD 19+ B cells incorporating magnetic beads and the clini-MACS device [28]. The removal of αβ T cells and B cells significantly reduces the cells responsible for GVHD and PTLD. This type of graft maintains CD34+ hematopoietic stem cells as well as NK cells, γδ T cells, and other cell types that are thought to facilitate engraftment, elicit

GVT effect and provide anti-infective activity. This new method of transplantation has recently been utilized in pediatric and adult patients with both malignant and nonmalignant hematologic conditions and is gaining more momentum due to encouraging clinical results.

Summary of current data for TCR αβ+ and CD 19+ depletion

Graft composition

Studies have shown favorable results with respect to graft composition, and have achieved adequate depletion of TCR αβ+/CD 19+ cells and good recovery of CD34+ cells. Studies conducted with pediatric patients have shown median numbers of CD34+ cells per kg body weight in the graft from 7.9×10^6 to 18.7×10^6 [29–38]. Studies conducted with adult patients have shown comparable median numbers of CD34+ cells per kg body weight of 11×10^6 to 17.69×10^6 [39,40]. Earlier studies involving CD34+ selection for haploidentical transplants were notable for CD34+ cell counts/kg body weight ranging from 1.2×10^6 to 13.8×10^6 [5,6,10]. This suggests that the cell doses achieved with TCR αβ+/CD 19+ depletion are not inferior to CD34+ selection. Median numbers of αβ+ T cells and B cells per kg body weight range from 11×10^3 to 55×10^3 [29–37,39,40] and 40×10^3 to 300×10^3 [29,32,34,35,37,39], respectively, which indicate robust depletion, typically on the order of 3–4 log depletion. Median numbers of γδ T cells and NK cells in the graft range from 4×10^6 to 11×10^6 per kg body weight [29,32,35–41] and 22×10^6 to 81.3×10^6 [32,34–37,39], respectively. These numbers indicate that large amounts of γδ T cells and NK cells are preserved in the graft. This is particularly desirable given the favorable effects that are associated with these types of immune cells in the context of allogeneic stem cell transplantation.

Engraftment

Several studies have shown successful primary engraftment rates (83–100%) of TCR αβ +/CD 19+ depleted stem cell grafts for haploidentical allo-HSCT, with average absolute neutrophil count (ANC) recovery by day 10–15 and average platelet recovery by day 10–17 [29,33–37,39,40]. Although various conditioning regimens have been studied (possibly due to heterogeneity of diseases studied), most have commonly included alkylator-based chemotherapy, often thiotepa and melphalan, as well as ATG or radiation. In abstract format, Prezioso et al. reported results on 32 adult patients with AML or ALL, who all engrafted successfully [39]. The conditioning regimen consisted of ATG, treosulfan, fludarabine, and thiotepa. Even in the cases of engraftment failure, most patients can be successfully salvaged with a 2nd attempt. This was seen in the abstract by Lang et al. involving 41 pediatric patients with both benign and malignant conditions who received a conditioning regimen of fludarabine or clofarabine, thiotepa, and melphalan in combination with either OKT3 or ATG-Fresenius [36]. Five of the 41 patients did not engraft, yet all of these five successfully engrafted on 2nd attempt after being reconditioned and infused with grafts from different haploidentical donors that were also depleted of αβ+ T cells and B cells.

Graft-versus-host disease

Studies so far have shown encouraging results of TCR αβ+/CD 19+ depleted stem cell grafts for haploidentical allo-HSCT with respect to the incidence of GVHD [29,33–

37,39,40]. Data suggest that the majority of aGVHD associated with this transplant method is low grade (grade I–II). Collectively, grade I–II aGVHD was reported in 3–39% of patients and primarily limited to the skin, rather than visceral organs. A small percentage of patients in these studies developed more severe aGVHD, but the overall incidence was low (0–15%). Kaynar et al. studied 34 adults with AML or ALL who received a conditioning regimen of fludarabine, thiotepa, melphalan, and ATG [40]. Mycophenolate was administered until day 60 for GVHD suppression. Median follow-up was 176 days. Grade I–II aGVHD was observed in 26% of the patients and grade III–IV aGVHD was observed in 6%. The rate of cGVHD was 6% with 3% being limited and 3% being extensive. The authors report on at least two patients treated for GVHD using steroids, cyclosporine, and mycophenolate. Locatelli et al. studied 80 pediatric patients with AML or ALL who each received one of four conditioning regimens, two of which involved TBI [37]. There was no post-transplantation GVHD prophylaxis. Median follow-up was 46 months. The rate of aGVHD was 30% and all were grade I–II. All of these patients responded to treatment with topical or systemic steroids. The rate of cGVHD was 5% with all cases limited to the skin. Bertaina et al. studied 23 children with a variety of benign diseases who received one of four different conditioning regimens in addition to ATG and rituximab [35]. There was no post-transplantation GVHD prophylaxis. Median followup was 18 months. Grade I–II aGVHD was observed in three of 23 patients and limited to the skin. No patients experienced acute visceral or any cGVHD.

Transplant related mortality

Studies of TCR αβ+/CD 19+ depletion for haploidentical allo-HSCT have shown encouraging results regarding TRM [29,32–37,39,40]. The reported incidence of TRM is variable, and ranges between 0 and 20%. Infection was the most common cause of TRM. Lang et al. conducted a study involving 30 pediatric patients with nonmalignant conditions, hematologic malignancies, and solid tumors who received a conditioning regimen of fludarabine, thiotepa, and melphalan with either ATG or total nodal irradiation. TRM in this study was 3.3% with all TRM due to infection [34]. Bhat et al. carried out a study involving 22 pediatric patients with both malignant and nonmalignant hematologic conditions who received a conditioning regimen consisting of TBI or chemotherapy. TRM in this abstract was 14% with all TRM due to infection [29]. Giardino et al. studied 12 pediatric patients with malignant and nonmalignant conditions. Some patients received a myeloablative, while others received a non-myeloablative conditioning regimen. The rate of TRM reported in this abstract was 15% [33]. The rate of TRM in these studies is lower on average than the rate of TRM seen in earlier studies involving transplantation of ‘megadoses’ of CD34+ cells or CD3+/CD19+ depleted cells from haploidentical donors (36–40% and 20–30%, respectively) [5,6,19,20,22].

Survival

Although survival data might be difficult to interpret from studies where multiple disease histologies were included, certain studies involving only one or two disease types provide insight regarding disease efficacy.

Many studies focus on children with AML or ALL. The study conducted by Locatelli et al. in 80 children with acute leukemia who received myeloablative conditioning, including ATG, showed overall survival (OS) of 72% at 5 years and disease-free survival (DFS) of 71% at 5 years [37]. Maschan et al. utilized this transplant method in 33 pediatric patients with high risk AML with 20 receiving matched unrelated donor (MUD) transplants and 13 receiving haploidentical transplants. The conditioning regimen consisted of treosulfan, melphalan, fludarabine, and ATG. OS and DFS at 2 years were 73% and 59%, respectively, for the haploidentical cohort [32].

The studies involving adult patients with AML or ALL report an OS of 41–54% with median follow-up time ranging from 24 to 27 months [39,40], which is comparable to survival in studies utilizing 'megadoses' of CD34+ cells [5,6] and those involving CD3+/CD19+ depletion [19,20,22] which report survival ranging from 30% to 50% at median time to follow-up ranging from 2 to 3 years.

Immune reconstitution

Studies looking at immune reconstitution following TCR αβ+/CD 19+ depletion for haploidentical allo-HSCT generally show rapid early reconstitution of γδ T cells and NK cells, with more gradual appearance of αβ T cells and B cells over 3–12 months [29,34–37,42,43]. Locatelli et al. found a rapid recovery of γδ T cells and NK cells with a median of 181 cells/μL (range 1–1335) and 236 cells/μL (range 47–1813) at one month, respectively [37]. This was followed by αβ T cell reconstitution, with median 47/μL (1–672) at 1 month and 186/μL (12–1340) at 3 months, but with recovery to 573/μL (135–2146) at 6 months and 1291/μL (259–2795) at 12 months. Similarly, the B cell population also gradually increased over time with negligible amounts at 1 and 3 months; however, the number of B cells reached 160/μL (2–1609) and 291/μL (40–1616) at 6 and 12 months, respectively [37].

A similar rapid recovery of γδ T cells and NK cells was reported by Lang et al. [36]. In this study, the recovery of γδ T cells started as early as day seven after transplant, and constituted the majority of CD3+ cells in the early post-transplant period. NK cell recovery also began in the first week after transplant which were those that were co-transfused, followed by early proliferation starting in the second week, with a median of only 12.5 days needed for the NK cell population to reach >200 cells/μL. The B cell population in this study started to recover by day 30 and normalized by day 150 post-transplant. There was no sustained deficiency of B cells.

The presence of mature donor NK cells in the immediate post-transplant period and rapid expansion thereafter significantly differs from NK cell kinetics in transplants involving CD34+ cell selection. The presence of mature donor-derived NK cells is often not seen in the recipient until at least 2–3 months posttransplant in the context of CD34+ selection as seen in a study by Pende et al. [44]. This represents a significant advantage of TCR αβ+/CD 19+ depletion over CD34+ selection.

Airoldi et al. studied immune reconstitution in 27 pediatric patients with malignant and nonmalignant conditions and primarily focused on γδ T cell reconstitution and function [43]. In this cohort, 2–3 weeks after transplant, recipient T lymphocytes constituted

predominantly γδ T cells (mean 91.5% of CD3+ lymphocytes, range 70–100%) with the αβ T cell population increasing over time and the γδ T cell subset decreasing over time. Since aminobisphosphonates induce proliferation and activation of the Vδ2 subset of γδ T cells, the authors also studied the effect of *in vitro* exposure to zoledronic acid on the expansion of γδ T cells in a subset of 13 patients. As expected, exposure to zoledronic acid increased the expansion of the Vδ2 subset of γδ T cells. They also found that *in vitro* exposure of leukemia cell lines to zoledronic acid enhanced the cytotoxic activity of Vδ2 cells.

Subsequently, the same group assessed whether *in vivo* exposure to zoledronic acid in the post-transplant period would improve the cytotoxic activity of γδ T cells against leukemia cells in 43 pediatric patients who had undergone αβ T cell and B cell depleted haplo-HSCT [45]. Zoledronic acid was administered every 28 days for up to six doses, and the effect on γδ T cells was studied in 33 of the 43 patients until at least 7 months post-transplant. The authors report a significant decrease in the percentage of central memory Vδ2 cells over time after the first zoledronic acid infusion coupled by a significant increase in the percentage of terminally differentiated Vδ2 cells. The authors assessed the cytotoxic activity of the γδ T cells by analyzing the level of CD107a surface expression as a marker of degranulation when the γδ T cells were exposed to AML or ALL primary blasts. They found that the level of cytotoxic activity of Vδ2 cells against the leukemic cells was significantly increased after patients received one infusion of zoledronic acid compared to cytotoxic activity pretreatment and that the level of cytotoxicity continued to increase as patients received additional zoledronic acid infusions. Ultimately, patients who received three or more zoledronic acid infusions had a significantly better probability of survival than patients who received one or two treatments (86% versus 54%, respectively, $p = .008$). These important findings suggest that zoledronic acid, given post-transplant, affects the differentiation and cytotoxicity of γδ T cells, is associated with significant graft-versus-leukemia effects, and therefore has the potential to improve outcomes. It also suggests that TCR αβ +/CD 19+ depletion might be a suitable platform for post-transplant immunotherapeutic approaches given that it is associated with significant numbers of transfused NK and γδ T cells in the graft as well as rapid immune recovery.

Collectively, these studies demonstrate rapid recovery of NK and γδ T cells associated with this type of transplant, which is desirable given their important roles in protection against infections and the GVT effect. The recovery of αβ T cells and B cells is also ultimately necessary for optimal immune function and observed to occur more gradually but consistently.

Infections

Cytomegalovirus (CMV) reactivation rates ranged from 30 to 70% in various studies, however, death from CMV occurred in no more than 5% [29,33–35,39,40,46]. Two studies reported on BK virus reactivation, ranging from 17 to 25% in their cohorts, but no BK virusrelated deaths were reported [34,40]. Three studies report on adenovirus detection, ranging from 5 to 53%, with two deaths reported [29,34,35]. There were no cases of Epstein-Barr virus (EBV)-associated PTLD reported in the five studies where it was monitored, and EBV viremia was only very rarely detected [29,34,35,37,40,46]. Overall,

these data indicate that the risk of viral reactivations remains significant in the context of TCR αβ+/CD 19+ depleted haplo-HSCT. However, the reported mortality rates associated with these infections are very low. Importantly, risk of EBV reactivation and EBV-related PTLD appear to be very low in patients undergoing this type of transplant. Ultimately, larger analyses will be required to compare rates of opportunistic infections and viral reactivations and their clinical consequences in TCR αβ+/CD 19+ depletion compared to other transplant modalities.

TCR αβ+/CD 19+ depletion in non-haploidentical donor transplantation

TCR αβ+/CD 19+ depletion is also being increasingly studied in non-haploidentical transplants, including matched related donor (MRD) and MUD transplants. A study conducted by de Witte et al. utilized TCR αβ+ depleted transplants from MRD or MUD sources for 32 patients with a variety of hematologic malignancies [47]. The conditioning regimen in this study consisted of ATG + fludarabine + busulfan. Post-transplant immune suppression was accomplished by a 28-day course of mycophenolic acid. This study did not involve CD 19+ depletion. Notable results reported in this abstract include the fact that all patients in this study engrafted and that there was no grade III–IV aGVHD (grade I–II not reported) at 100 days. There were two deaths in the first few months of follow-up, one due to mucormycosis, the other related to GVHD post DLI. There was no significant difference in the rates of CMV or EBV reactivation when compared to a historical control cohort of patients receiving T cell replete allo-HSCTs (CMV incidence 54% versus 38%, p=.48, and EBV incidence 32% versus 9%, p=.148). A separate study conducted by the same group and reported in an abstract used TCR αβ+ depleted grafts from MRD and MUD sources for 14 patients with poor risk or very poor risk leukemia [48]. The rate of primary engraftment was 40%, 75%, or 100% depending on the type of conditioning regimen utilized (fludarabine + cyclophosphamide, fludarabine + busulfan, and ATG + fludarabine + busulfan, respectively). In this study, NK cell and γδ T cell recovery in the early posttransplant period showed a similar pattern as seen in the haploidentical setting.

There have also been studies using both haploidentical donors and MUD sources with TCR αβ+/CD 19+ depletion that show no significant difference between transplants when the two cohorts are compared. The study by Maschan et al. [32] notably included donor lymphocyte infusion (DLI) after transplant. Four patients in this study received therapeutic DLI for >10% increase in mixed chimerism in CD34+ cells or bone marrow on monthly surveillance. Patients with high risk disease were eligible for modified preventive DLI, and 13 patients received a median of three preventive DLIs. Engraftment was 100%. Grade II–III aGVHD was observed in 39%. The rate of cGVHD was 30%, which is higher than that reported in other trials using this transplant method. The authors suggest that this may have been due to the use of horse ATG as opposed to rabbit ATG given that horse ATG is known to be less immunosuppressive. However, the use of DLI in this trial makes GVHD rates difficult to interpret. The rate of CMV reactivation was 52% and CMV disease 6%. The rate of EBV reactivation was 50%, which is notably higher than in other trials, though the rate of EBV disease was low at 6%. Two-year TRM was 10% with two-year OS and leukemia-free survival 67% and 68%, respectively. Comparison of haploidentical and MUD transplantation

was not a primary aim of the study, but the authors report that engraftment, GVHD, and viral reactivation rates did not differ significantly between the two groups.

The same group studied the use of TCR αβ+/CD 19+ depleted allo-HSCTs in pediatric patients with ALL and reported their results in two separate abstracts. They found no significant differences in the rates of engraftment, GVHD, TRM, or OS between the 42 patients undergoing haploidentical transplant and the 25 patients undergoing MUD transplant [31]. Similarly, this group utilized TCR αβ+/CD 19+ depleted allo-HSCTs for 59 pediatric patients with AML and found no significant difference in the rates of engraftment, GVHD, TRM, or survival between the 22 patients receiving haploidentical transplants and the 37 patients receiving MUD transplants [30].

Taken together, the data from these studies suggest that TCR αβ+/CD 19+ depleted allo-HSCT may also be advantageous in patients undergoing MUD or MRD transplants with similar outcomes compared to TCR αβ+/CD 19+ depletion for haploidentical transplantation.

Conclusions

In summary, current available data from clinical trials studying TCR αβ+/CD 19+ depletion for haploidentical allo-HSCT indicate that this modality of transplantation might be superior to other strategies involving haploidentical transplantation. Rates of engraftment, acute and cGVHD, TRM, and survival are comparable, or even better, compared to studies involving earlier strategies for haploidentical allo-HSCT including CD34+ selection and CD3+/CD19+ depletion. It will be particularly interesting to further explore how the infusion of large numbers of immunocompetent cells and the rapid immune reconstitution will affect these classical HSCT outcome parameters. Beyond the haploidentical setting, this graft engineering technique seems also suitable for the preparation of matched related and matched unrelated stem cell products. It should be noted however that the majority of the studies exploring this have involved only a small number of patients.

Allogeneic stem cell transplantation is commonly performed in the context of non-detectable or minimal residual disease, offering the patient the highest likelihood of cure. It is reasonable to believe that fast engraftment and the presence of immediately available immune cells that can facilitate a critical GVT effect is a decisive factor for decreasing relapse-related post-transplant mortality. In addition, this constellation may allow for novel early post-transplant immunotherapies (e.g. with tumor-targeting antibodies) that can take advantage of the donor-derived transplanted and rapidly reconstituting immune cells. TCRαβ+/CD19+ depleted haploidentical HSCT may therefore afford distinct immunological advantages over other allogeneic transplant methodologies and provide a unique opportunity to further augment and exploit a potent GVT immunoreactivity.

Larger studies and longer follow-up in the various patient populations that are in need of life-saving stem cell transplantation are necessary to further evaluate TCRαβ+/CD19+ depleted graft engineering. This will be particularly true for better delineating the risk of cGVHD, infections, and survival for more specific disease sub-types and conditioning

regimens, and comparing this strategy to other haploidentical and non-haploidentical allogeneic transplant approaches.

Acknowledgments

The authors thank James P Zacny, PhD for his editorial work.

Funding

This work was made possible in part by NIH/NCI 5P30CA014520-40 (University of Wisconsin Carbone Cancer Center Support Grant).

Abbreviations

aGVHD	Acute graft-versus-host disease
ALL	Acute lymphoblastic leukemia
allo-HSCT	Allogeneic hematopoietic stem cell transplantation
AML	Acute myeloid leukemia
ATG	Anti-thymocyte globulin
cGVHD	Chronic graft-versus-host disease
CML	Chronic myelogenous leukemia
CMV	Cytomegalovirus
CyA	Cyclosporin-A
DFS	Disease-free survival
EBV	Epstein-Barr virus
EFS	Event-free survival
GVHD	Graft-versus-host disease
JMML	Juvenile myelomonocytic leukemia
LFS	Leukemia-free survival
MDS/MPN	Myelodysplastic/myeloproliferative neoplasms
MM	Multiple myeloma
MRD	Matched related donor
MTX	Methotrexate
MUD	Matched unrelated donor
NHL	Non-Hodgkin lymphoma

OS	Overall survival
TBI	Total body irradiation
TRM	Transplant-related mortality

References

[1]. National Marrow Donor Program a contractor for the C.W. Bill Young Cell Transplantation Program operated through the U. S. Department of Health and Human Services, H.R.a.S. Donor Registry Transplant Data Last [updated 2017 Mar 10].

[2]. Martin PJ. In: Blume KG, Forman SJ, Appelbaum FR, editors. Overview of marrow transplantation immunology in Thomas' hematopoietic cell transplantation. Malden, MA: Blackwell Science; 2004 p. 16–30.

[3]. Gragert L, Eapen M, Williams E, et al. HLA match likelihoods for hematopoietic stem-cell grafts in the U.S. registry. N Engl J Med. 2014;371:339–348. [PubMed: 25054717]

[4]. Stringaris K, Barrett AJ. The importance of natural killer cell killer immunoglobulin-like receptor-mismatch in transplant outcomes. Curr Opin Hematol. 2017;24:489–495. [PubMed: 28817402]

[5]. Aversa F, Tabilio A, Velardi A, et al. Treatment of high-risk acute leukemia with T-cell-depleted stem cells from related donors with one fully mismatched HLA haplotype. N Engl J Med. 1998;339:1186–1193. [PubMed: 9780338]

[6]. Aversa F, Terenzi A, Tabilio A, et al. Full haplotype-mismatched hematopoietic stem-cell transplantation: a phase II study in patients with acute leukemia at high risk of relapse. JCO. 2005;23:3447–3454.

[7]. Ratanatharathorn V, Logan B, Wang D, et al. Prior rituximab correlates with less acute graft-versus-host disease and better survival in B-cell lymphoma patients who received allogeneic peripheral blood stem cell transplantation. Br J Haematol. 2009;145:816–824. [PubMed: 19344418]

[8]. Arai S, Sahaf B, Narasimhan B, et al. Prophylactic rituximab after allogeneic transplantation decreases B-cell alloimmunity with low chronic GVHD incidence. Blood. 2012;119:6145–6154. [PubMed: 22563089]

[9]. Iori AP, Torelli GF, De Propris MS, et al. B-cell concentration in the apheretic product predicts acute graftversus-host disease and treatment-related mortality of allogeneic peripheral blood stem cell transplantation. Transplantation. 2008;85:386–390. [PubMed: 18322430]

[10]. Handgretinger R, Lang P, Schumm M, et al. Isolation and transplantation of autologous peripheral CD34+ progenitor cells highly purified by magnetic-activated cell sorting. Bone Marrow Transplant. 1998;21:987–993. [PubMed: 9632271]

[11]. Schumm M, Lang P, Taylor G, et al. Isolation of highly purified autologous and allogeneic peripheral CD34+ cells using the CliniMACS device. J Hematother. 1999;8:209–218. [PubMed: 10349915]

[12]. Lamb LS Jr., Lopez RD. gammadelta T cells: a new frontier for immunotherapy? Biol Blood Marrow Transplant. 2005;11:161–168. [PubMed: 15744234]

[13]. Ruggeri L. Effectiveness of donor natural killer cell alloreactivity in mismatched hematopoietic transplants. Science. 2002;295:2097–2100. [PubMed: 11896281]

[14]. Ruggeri L, Mancusi A, Capanni M, et al. Donor natural killer cell allorecognition of missing self in haploidentical hematopoietic transplantation for acute myeloid leukemia: challenging its predictive value. Blood. 2007;110:433–440. [PubMed: 17371948]

[15]. Ruggeri L, Capanni M, Casucci M, et al. Role of natural killer cell alloreactivity in HLA-mismatched hematopoietic stem cell transplantation. Blood. 1999;94:333–339. [PubMed: 10381530]

[16]. Morrison BE, Park SJ, Mooney JM, et al. Chemokine-mediated recruitment of NK cells is a critical host defense mechanism in invasive aspergillosis. J Clin Invest. 2003;112:1862–1870. [PubMed: 14679181]

[17]. Cook M, Briggs D, Craddock C, et al. Donor KIR genotype has a major influence on the rate of cytomegalovirus reactivation following T-cell replete stem cell transplantation. Blood. 2006;107:1230–1232. [PubMed: 16239436]

[18]. Leung W, Iyengar R, Turner V, et al. Determinants of antileukemia effects of allogeneic NK cells. J Immunol. 2004;172:644–650. [PubMed: 14688377]

[19]. Hale GA, Kasow KA, Kwan G, et al. Haploidentical stem cell transplantation with CD3 depleted mobilized peripheral blood stem cell grafts for children with hematologic malignancies. Blood. 2005;106:2910.

[20]. Hale GA, Kasow KA, Madden R, et al. Mismatched family member donor transplantation for patients with refractory hematologic malignancies: long-term followup of a prospective clinical trial. Blood. 2006;108:3137.

[21]. Handgretinger R, Chen X, Pfeiffer M, et al. Feasibility and outcome of reduced-intensity conditioning in haploidentical transplantation. Ann N Y Acad Sci. 2007;1106:279–289. [PubMed: 17442774]

[22]. Bethge WA, Faul C, Bornhäuser M, et al. Haploidentical allogeneic hematopoietic cell transplantation in adults using CD3/CD19 depletion and reduced intensity conditioning: an update. Blood Cells Mol Dis. 2008;40:13–19. [PubMed: 17869547]

[23]. Neipp M, Exner BG, Maru D, et al. T-cell depletion of allogeneic bone marrow using anti-alphabetaTCR monoclonal antibody: prevention of graft-versus-host disease without affecting engraftment potential in rats. Exp Hematol. 1999;27:860–867. [PubMed: 10340402]

[24]. Huang Y, Cramer DE, Ray MB, et al. The role of alpha-beta- and gammadelta-T cells in allogenic donor marrow on engraftment, chimerism, and graft-versus-host disease. Transplantation. 2001;72:1907–1914. [PubMed: 11773887]

[25]. Otto M, Barfield RC, Iyengar R, et al. Human gammadelta T cells from G-CSF-mobilized donors retain strong tumoricidal activity and produce immunomodulatory cytokines after clinical-scale isolation. J Immunother. 2005;28:73–78. [PubMed: 15614047]

[26]. Godder KT, Henslee-Downey PJ, Mehta J, et al. Long term disease-free survival in acute leukemia patients recovering with increased gammadelta T cells after partially mismatched related donor bone marrow transplantation. Bone Marrow Transplant. 2007;39: 751–757. [PubMed: 17450185]

[27]. Lamb L, Henslee-Downey P, Parrish R Jr, et al. Increased frequency of TCR gamma delta + T cells in disease-free survivors following T cell-depleted, partially mismatched, related donor bone marrow transplantation for leukemia. J Hematother. 1996;5: 503–509. [PubMed: 8938522]

[28]. Chaleff S, Otto M, Barfield RC, et al. A large-scale method for the selective depletion of alphabeta T lymphocytes from PBSC for allogeneic transplantation. Cytotherapy. 2007;9:746–754. [PubMed: 17917892]

[29]. Bhat S, Ngangbam S, Iqbal W, et al. Outcomes of myeloablative haplo-identical hematopoietic stem cell transplant in pediatric patients with TCR α/β; and CD 19 depletion. Biol Blood Marrow Transplant. 2017;23: S191–S192.

[30]. Shelikhova L, Shekhovtsova Z, Balashov D. TCRAB+/CD19+-depletion in hematopoietic stem cell transplantation from matched unrelated and haploidentical donors in pediatric acute myeloblastic leukemia patients. European Hematology Association Congress; 2016; Copenhagen, Denmark. 2016. Abstract E1521.

[31]. Shelikhova L, Shekhovtsova Z, Balashov D, et al. Tcrαβ+/CD19+-depletion in hematopoietic stem cells transplantation from matched unrelated and haploidentical donors following treosulfan or TBI-based conditioning in pediatric acute lymphoblastic leukemia patients. Blood. 2016;22:S4672.

[32]. Maschan M, Shelikhova L, Ilushina M, et al. TCR-alpha/beta and CD19 depletion and treosulfan-based conditioning regimen in unrelated and haploidentical transplantation in children with acute myeloid leukemia. Bone Marrow Transplant. 2016;51:668–674. [PubMed: 26808573]

[33]. Giardino S, Caroleo A, Caviglia Iet al. Haploidentical stem cell transplantation with post-transplant cyclophosphamide or in vivo αβ+ T and CD19 + B cells depletion. A Single Center Pediatric Experience. Annual Meeting of the European Society for Blood and Marrow Transplantation; 2017; Marseille, France Abstract B149.

[34]. Lang PJ, Schlegel PG, Meisel R, et al. TCR-alpha/beta and CD19 depleted haploidentical stem cell transplantation following reduced intensity conditioning in children: first results of a prospective multicenter phase I/II clinical trial. Blood. 2016;128:389.

[35]. Bertaina A, Merli P, Rutella S, et al. HLA-haploidentical stem cell transplantation after removal of alphabeta + T and B cells in children with nonmalignant disorders. Blood. 2014;124:822–826. [PubMed: 24869942]

[36]. Lang P, Feuchtinger T, Teltschik H-M, et al. Improved immune recovery after transplantation of TCRalphabeta/CD19-depleted allografts from haploidentical donors in pediatric patients. Bone Marrow Transplant. 2015;50:S6–S10. [PubMed: 26039210]

[37]. Locatelli F, Merli P, Pagliara D, et al. Outcome of children with acute leukemia given HLA-haploidentical HSCT after αβ T-cell and B-cell depletion. Blood. 2017;130:677–685. [PubMed: 28588018]

[38]. Li Pira G, Malaspina D, Girolami E, et al. Selective depletion of αβ T cells and B cells for human leukocyte antigen-haploidentical hematopoietic stem cell transplantation. A three-year follow-up of procedure efficiency. Biol Blood Marrow Transplant. 2016; 22:2056–2064. [PubMed: 27519279]

[39]. Prezioso L, Bonomini S, Schifano C, et al. Good immunological reconstitution in adults with acute leukemia after alpha-beta TCR/CD19+ depleted haploidentical hematopoeitic stem cell transplantation (HSCT). European Hematology Association Congress; 2017; Madrid, Spain. Abstract E1497.

[40]. Kaynar L, Demir K, Turak EE, et al. TcRαβ-depleted haploidentical transplantation results in adult acute leukemia patients. Hematology. 2017;22:136–144. [PubMed: 27724812]

[41]. Schumm M, Lang P, Bethge W, et al. Depletion of T-cell receptor alpha/beta and CD19 positive cells from apheresis products with the CliniMACS device. Cytotherapy. 2013;15:1253–1258. [PubMed: 23993299]

[42]. Park M, Im HJ, Koh KN, et al. Reconstitution of T cell subsets after haploidentical hematopoietic cell transplantation using alpha beta T cell-depleted graft and the clinical implication of γδ T cells. Bone Marrow Transplant. 2016;51:S29; Abstract O044.

[43]. Airoldi I, Bertaina A, Prigione I, et al. γδ T-cell reconstitution after HLA-haploidentical hematopoietic transplantation depleted of TCR-αβ+/CD19+ lymphocytes. Blood. 2015;125:2349–2358. [PubMed: 25612623]

[44]. Pende D, Marcenaro S, Falco M, et al. Anti-leukemia activity of alloreactive NK cells in KIR ligand-mismatched haploidentical HSCT for pediatric patients: evaluation of the functional role of activating KIR and redefinition of inhibitory KIR specificity. Blood. 2009;113: 3119–3129. [PubMed: 18945967]

[45]. Bertaina A, Zorzoli A, Petretto A, et al. Zoledronic acid boosts γδ T-cell activity in children receiving αβ(+) T and CD19(+) cell-depleted grafts from an HLA-haploidentical donor. Oncoimmunology. 2017;6:e1216291. [PubMed: 28344861]

[46]. Laberko A, Bogoyavlenskaya A, Shelikhova L, et al. Risk factors for and the clinical impact of cytomegalovirus and Epstein-Barr virus infections in pediatric recipients of TCR-α/β- and CD19-depleted grafts. Biol Blood Marrow Transplant. 2017;23:483–490. [PubMed: 28039080]

[47]. de Witte M, Fleurke G, Timmerman L, et al. Immune reconstitution and clinical outcome after α/β T-cell depleted allogeneic stem cell transplantation from matched related and unrelated donors. Blood. 2015;126:4313.

[48]. DeWitte MA, te Boome L, van der Wagen L, et al. Development of an alpha/beta T-cell depleted allogeneic stem cell transplantation protocol form matched related and unrelated donor grafts in patients with poor risk leukemia. Blood. 2014;124:3885.

Table 1.

Summary of key data for TCR αβ+/CD 19+ depletion for haploidentical transplantation.

Reference[a]	Number of patients Ages: median (range)	Disease types	Conditioning regimen and immunosuppression	Median CD34+ cells/kg body weight	Median αβ + T cells/kg body weight	Engraftment	aGVHD	cGVHD	TRM	Survival
29. Bhat et al. (abstract)	22 62 months (3–184)	32% ALL 5% AML 5% CML 5% Juvenile myelomonocytic leukemia 53% Benign diseases	*Conditioning regimen:* TBI or chemotherapy	18.7×10^6	0.055×10^6	95%	Grade II–IV 22% Grade III–IV 4.5%	0%	14% total, all from infection	EFS-77% at median 402 days
30. Shelikhovad (abstract)	59 9.1 years (0.6–22)	100% AML	*Conditioning regimen:* Treosulfan, melphalan, fludarabine *GVHD prophylaxis*: 1. ATG +/− post-transplant tacrolimus/MTX 2. ATG, rituximab, and post-transplant bortezomib or tacrolimus/MTX	7.9×10^6	15×10^3	98.3%	Grade ≥ II 20.3% Grade III–IV 8.5%	28%	10.8% total at 3 years, all from infection	OS-64% at 3 years EFS-63% at 3 years
31. Shelikhova and coworkers[d] (abstract)	67 9.3 y ears, (0.15–20)	100% ALL	*Conditioning regimen*: Treosulfan based versus TBI based *GVHD prophylaxis*: 1. ATG +/− post-transplant tacrolimus/MTX 2. ATG, rituximab, bortezomib, and post-transplant bortezomib or tacrolimus	10×10^6	19×10^3	96%	Grade II–IV-23.9% Grade II–IV-7.5%	22.9%	17% total at 24 months	OS-50% at 2 years EFS-49.6% at 2 years
32. Maschan et al.[d] (manuscript)	33 9.3 years (1–23)	100% AML	*Conditioning regimen*: Treosulfan, melphalan, fludarabine, ATG *Post-transplant immunosuppression*: - Tacrolimus and MTX in 21 - Tacrolimus in 5 -MTX in 2 - None in 5	9×10^6	20×10^3	100%	Grade II–III 39% Grade IV-0%	30% total Limited-20% Extensive-10%	13% total at 2 years	OS-67% at 2 years LFS-68% at 2 years
33. Giardino et al.[b] (abstract)	12 6.4 years (1.8–11.5)	17% malignant conditions 83% benign conditions	*Conditioning regimen*: Myeloablative versus non-myeloablative with ATG and rituximab *Post-transplant immunosuppression*: CyA in two patients who received $>1 \times 10^4$ αβ + T cells after depletion. None for the other 10 patients	15.5×10^6	0.53×10^5	67%	Grade I–III-0% Grade IV-8.3%	0%	15% total	OS-85% at 3 years

Leuk Lymphoma. Author manuscript; available in PMC 2019 September 27.

Reference[a]	Number of patients Ages: median (range)	Disease types	Conditioning regimen and immunosuppression	Median CD34+ cells/kg body weight	Median αβ + T cells/kg body weight	Engraftment	aGVHD	cGVHD	TRM	Survival
34. Lang et al. (abstract)	30 7 years (1–17)	30% ALL 27% AML 20% solid tumors 10% MDS/MPN 13% other benign conditions	*Conditioning regimen*: Fludarabine, thiotepa, and melphalan with either ATG or total nodal irradiation Post-transplant immunosuppression: Mycophenolate mofetil through day 30	14.6×10^6	14×10^3	77%	Grade II-3.3% Grade III–IV-0%		3.3% total, all from infection	OS 93% 100 days after transplant
35. Bertaina et al. (manuscript)	23 3.3 years (0.4–12)	100% benign diseases	*Conditioning regimen*: 1. Busulfan, thiotepa, fludarabine 2. Treosulfan, thiotepa, fludarabine 3. Treosulfan, fludarabine 4. Fludarabine, cyclophosphamide +/− single dose TBI All regimens also involved ATG-Fresenius and rituximab	15.8×10^6	4×10^4	83%	Grade I–II 13% Grade III–IV 0%	0%	9.3% total, all from infection	OS and DFS 91.1% at 24 months
36. Lang et al. (manuscript)	41 9 years (2–18)	49% ALL 22% AML 7% MDS/ JMML 10% relapsed solid tumors 12% benign diseases	*Conditioning regimen*: 1. Fludarabine or clofarabine, thiotepa, and melphalan with OKT3 2. Fludarabine or clofarabine, thiotepa, and melphalan with ATG-Fresenius Post-transplant immunosuppression: Mycophenolate through day 30	14.9×10^6	16.9×10^3	88%	Grade 0–39% Grade I-36% Grade II-10% Grade III–IV 15%	Total-22% Limited-15% Extensive-7%		
37. Locatelli et al. (manuscript)	80 9.7 years (0.9–20.9)	AML 30% ALL 70%	*Conditioning regimen*: 1. TBI + Thiotepa+-Fludarabine 2. TBI + Thiotepa+-Melphalan 3. Thiotepa + busulfan + fludarabine 4. Busulfan + cyclophosphamide + melphalan	13.93×10^6	0.047×10^6	98%	30%, all grade I–II	5% all limited to the skin	5% total 2.5% idiopathic pneumonitis 1.25% infection 1.25% cardiac insufficiency	OS 72% at 5 years DFS 71% at 5 years
39. Prezioso and Bonomini (abstract)	32 51 years (19–74)	84% AML 16% ALL	*Conditioning regimen*: Treosulfan, fludarabine, thiotepa, ATG	11×10^6	4.9×10^4	100%	Grade II-25% Severe-6.25%	6.25%-all mild	15% total	OS 41% at 27 months
40. Kaynar et al. (manuscript)	34 28 years (1st–3rd quartile 21–38)	AML 71% ALL 29%	*Conditioning regimen*: Fludarabine, thiotepa, melphalan, ATG *Post-transplant immunosuppression*:	12.69×10^6	11.72×10^3	91%	Grade I–II 26% Grade III–IV 6%	Total-6% Limited-3% Extensive-3%	21% total 15% infection 3% GVHD 3% graft failure	OS 54% at 1 year DFS 42% at 1 year

Reference[a]	Number of patients Ages: median (range)	Disease types	Conditioning regimen and immunosuppression	Median CD34+ cells/kg body weight	Median αβ + T cells/kg body weight	Engraftment	aGVHD	cGVHD	TRM	Survival
			Mycophenolate through day 60							
47. de Witte et al.[c] (abstract)	32 patients	100% hematologic malignancies-AML, ALL, MM, NHL	*Conditioning regimen*: ATG, fludarabine, busulfan *Post-transplant immunosuppression*: mycophenolic acid for 28 days			100%	Grade > II 0%		6.25% total 3.125%-infection 3.125%-GVHD	OS-94% and LFS 88% at 1–14 months
48. DeWitte et al.[c] (abstract)	14 patients	100% poor risk or very poor risk leukemia	*Conditioning regimen*: Cohort I-fludarabine, cyclophosphamide Cohort II-fludarabine, busulfan Cohort III-ATG, fludarabine, busulfan	Recovery of ~75%	~4 log depletion	Cohort I-40% Cohort II-75% Cohort III-100%				

aGVHD: acute graft-versus-host disease; ALL: acute lymphocytic leukemia; AML: acute myeloid leukemia; ATG: antithymocyte globulin; cGVHD: chronic graft-versus-host disease; CML: chronic myeloid leukemia; DFS: disease-free survival; EFS: event-free survival; JMML: juvenile myelomonocytic leukemia; LFS: leukemia-free survival; MDS: myelodysplastic syndrome; MM: multiple myeloma; MPN: myeloproliferative neoplasm; MRD: matched related donor; MTX: methotrexate; MUD: matched unrelated donor; NFIL: non-Hodgkin lymphoma; OS: overall survival; TBI: total body irradiation; TCR: T cell receptor; TRM: transplant-related mortality.

[a] Number represents citation number in references section.

[b] Incroporates only data from alpha/beta T cell and B cell depletion.

[c] Incorporates data from both MRD and MUD transplants.

[d] Incorporates data from both haploidentical and MUD transplants.