Rapid Immune Recovery and Graft-versus-Host Disease–like Engraftment Syndrome following Adoptive Transfer of Costimulated Autologous T Cells

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Abstract

Purpose: Previously, we showed that adoptive transfer of in vivo vaccine-primed and ex vivo (anti-CD3/anti-CD28) costimulated autologous T cells (ex-T) at day +12 after transplant increased CD4 and CD8 T-cell counts at day +42 and augmented vaccine-specific immune responses in patients with myeloma. Here, we investigated the safety and kinetics of T-cell recovery after infusing ex-T at day +2 after transplant.

Experimental Design: In this phase I/II two-arm clinical trial, 50 patients with myeloma received autografts after high-dose melphalan followed by infusions of ex-T at day +2 after transplant. Patients also received pretransplant and posttransplant immunizations using a pneumococcal conjugate vaccine only (arm B; n = 24) or the pneumococcal conjugate vaccine plus an HLA-A2–restricted multipeptide vaccine for HLA-A2+ patients (arm A; n = 26).

Results: The mean number of T cells infused was 4.26 x 10^10 (range, 1.59-5.0). At day 14 after transplant, the median CD3, CD4, and CD8 counts were 4,198, 1,545, and 2,858 cells/μL, respectively. Interleukin (IL)-6 and IL-15 levels increased early after transplant and IL-15 levels correlated significantly to day 14 T-cell counts. Robust vaccine-specific B- and T-cell responses were generated. T-cell infusions were well tolerated with no effect on hematopoietic recovery. Eight patients (16%) developed a T-cell “engraftment syndrome” characterized by diarrhea and fever that was clinically and histopathologically indistinguishable from grade 1 to 3 acute graft-versus-host disease (GVHD) of the gastrointestinal tract (seven patients) and/or grade 1 to 2 cutaneous GVHD (four patients).

Conclusions: Adoptive T-cell transfers achieve robust T-cell recovery early after transplant and induce moderate-to-severe autologous GVHD in a subset of patients.

High-dose chemotherapy followed by autologous stem cell transplantation induces complete responses and extended progression-free survival in about 20% to 40% of myeloma patients (1–3). However, even after tandem transplants, 10-year disease-free survival rates are <20% and cure rates are probably no better than 10% with long-term follow-up (4). Allogeneic transplants have been performed for myeloma with the rationale that cure rates may be increased through a T-cell–mediated “graft versus tumor” effect, but increased toxicity and mortality due to graft-versus-host disease (GVHD) largely offsets the benefit of enhanced disease control (5–8). New approaches are needed to improve on the results of autologous transplants for myeloma and other blood cancers. Some but not all retrospective studies suggest that more rapid lymphocyte recovery during the early posttransplant period may be associated with better clinical outcomes for myeloma and other hematologic neoplasms (9–11). Furthermore, immune dysfunction following high-dose chemotherapy is clearly associated with an increased risk for serious bacterial and viral infections (12–14). Thus, strategies to augment the recovery and function of autologous T cells after transplant may be beneficial.
Materials and Methods

**Trial design.** Study participants were at least 18 y old with symptomatic multiple myeloma that required systemic treatment. All patients had received initial therapy using at least three cycles of standard regimens (typically bortezomib, thalidomide, or lenalidomide plus dexamethasone) by their referring oncologist before study enrollment. On study entry, patients were required to have measurable disease (based on serum/urine electrophoresis studies or serum free light chain studies); patients in complete remission were not eligible unless they had high-risk cytogenetic features (e.g., chromosome 13 or 17 deletions, 4;14 or 14;16 translocations, or complex karyotypes). Patients were required to have adequate organ function as defined by serum creatinine levels <3.0 mg/dL, left ventricular ejection fraction >45%, and lung function parameters >40% predicted. All participants gave written informed consent on enrollment in accordance with the Declaration of Helsinki; study approval was obtained from the Institutional Review Boards of the University of Maryland and the University of Pennsylvania as well as the U.S. Food and Drug Administration.

The design of the trial is depicted in Fig. 1. Briefly, patients were first tested for HLA-A2 status: HLA-A2* patients were assigned to arm A and HLA-A2* patients were assigned to arm B. Patients who were HLA-A2* (arm A) received immunizations with 100 μg of each of the following peptides: (a) hTERT I540 peptide (ILAKFLHWL; ref. 24), (b) hTERT R572Y peptide (YLFYFRKSV; ref. 25), (c) hTERT D988Y peptide (YLQVNSLQTV; ref. 26), (d) survivin Sur1M2 peptide (LMLGEFLKI; ref. 26), and (e) cytomegalovirus control peptide N495 (NLVPMMVAT; ref. 27). Vaccinations consisted of aqueous solutions of peptide (each peptide >92% pure and good manufacturing grade; Merck Biosciences AG) emulsified in the adjuvant Montanide ISA 51 (Seppic, Inc.) and delivered s.c. in the thigh (right thigh, hTERT I540, hTERT R572Y, and hTERT D988Y peptide emulsion; left thigh, Sur1M2 and cytomegalovirus N495 peptide emulsion). Sargramostim (clinical-grade granulocyte macrophage colony-stimulating factor; Berlex Laboratories, Inc.) was also given s.c. at each of the two peptide injection sites (70 μg) per vaccination. Patients in arm A also received an i.m. injection of the PCV (Prevnar, Wyeth) into the nondominant deltoid. HLA-A2* patients (arm B) received the PCV immunization only along with one injection of granulocyte macrophage colony-stimulating factor (70 μg) per injection.

About 10 d after the first set of immunizations, all patients underwent steady-state mononuclear cell collections by apheresis to collect ~1 × 10^8 mononuclear cells per kilogram body weight. Patients proceeded to stem cell mobilization using one of several regimens (most commonly cyclophosphamide at a dose of 1.5-4.5 g/m^2) followed by s.c. injections of granulocyte colony-stimulating factor (10 μg/kg). High-dose therapy consisted of melphalan at a dose of 200 mg/m^2 followed by infusions of autologous stem cells (at least 2 × 10^6 CD34+ cells/kg body weight) at day 0. Costimulated autologous T cells were infused on day +2 (see details below). Standard supportive care measures included antibiotic prophylaxis and administration of granulocyte colony-stimulating factor starting on day +5. Three additional sets of immunizations were given at days +14, +42, and +90 using the same arm-specific procedure that was used for the first immunization.
**T-cell expansion and adoptive transfer.** The mononuclear cell apheresis product was monocyte depleted by counterflow centrifugal elutriation (CaridinBCT Elutra Cell Separation System) because monocytes may inhibit lymphocyte proliferation. Monocyte-depleted mononuclear cells were then cryopreserved until 9 to 12 d before the scheduled reinfusion date (day +2 after transplant). Cells were thawed and cocultured with Dynal paramagnetic M-450 beads (DynalInvitrogen) coated with anti-CD3 (OKT3, OrthoBiotech)/anti-CD28(clone 9.3) at a ratio of three beads per cell first in a Baxter Lifecell flask and subsequently in the WAVE bioreactor system (28). The cells were grown in X-VIVO 15 (Lonza Walkersville) supplemented with 5% pooled human AB serum (ValleyBiomedical). The cultures were maintained for up to 12 d before harvesting and preparation for reinfusion. All infused T-cell products were required to meet release criteria specified for T-cell phenotype, cell viability, pyrogenicity, and freedom from bead contamination. Culture samples were tested for bacteria and fungi 2 d before harvesting and from the final cellular product. Testing of the final cellular product for bacterial endotoxin (Endosafe, Charles River) and cell phenotype by flow cytometry were also performed. Cell count, cell viability, and endotoxin testing were done to determine whether the final products met specified release criteria. In addition, the absolute number of cells with a plasma cell immunophenotype in the preharvest or final product had to be less than or equal to the absolute number of cells of the same population in the post-wash starting (apheresis) product. Plasma cells were identified by pre gating on viable cells defined by ViaProbe (Becton Dickinson) and then collecting and analyzing CD3/CD19/CD38/CD138 cells on a Becton Dickinson FACSCalibur with CellQuest software. On the designated infusion date, the T cells were harvested. The beads were removed with a Baxter Fenwal MaxSep magnetic separator, washed, and concentrated with the Baxter Fenwal Harvester System and resuspended in 100 to 500 mL of 1:1 plasmalyte A/dextrose 5%. 0.45% NaCl containing 0.5% to 1% human serum albumin. The harvested cells were transported by courier from the cell production facility to the patient and infused on the same day (day +2 of transplant). The cells were infused over 20 to 60 min without a leukocyte filter after premedication with acetaminophen and diphenhydramine. Corticosteroids were available at the bedside in the event of an allergic-type reaction. The target number of costimulated T cells for infusion was \( \sim 5 \times 10^{10} \) T cells. This target number was 5-fold greater than that used in our previous trial due to the increased expansion efficiency of the WAVE bioreactor system. Of the 50 expansions, one product failed to meet release criteria due to bacterial contamination; a second expansion was done and the cells were successfully infused at day 16 after transplant.

**Cytokine assays.** Serum concentrations of cytokines were measured by ELISA (R&D Systems) or by the Luminex bead assay (Invitrogen) according to the manufacturers’ instructions.

**Immunosassays and phenotyping.** Immune responses to the Prevnar(PCV) were assessed by ELISA binding assays (for antibody responses) and CFSE dilution (for T-cell proliferative responses to the CRM-197 carrier protein) as described previously (22) minus CD25 staining. T-cell responses to the hTERT and survivin peptides were assessed by in vitro stimulation followed by tetramer analysis as previously described (29), except that all tetramers were obtained from Beckman Coulter. Regulatory T (Treg) cell subpopulations (CD4/CD127/FOXp3+) were determined by intracellular staining according to the manufacturer’s instructions with anti-FOXp3 (ebiSciences) followed by surface staining with anti-CD127 and anti-CD4 (Becton Dickinson) for 15 min at room temperature. Effector T (Teff) cell subpopulations (CD3/CD8+) were stained with anti-CD3 and anti-CD8 (Becton Dickinson) for 15 min at 4°C.

**Statistical methods.** The observation times were days 14, 60, 100, and 180. Because the observation times were slightly different between the current trial and the previous trial and some observations were missing, to compare CD3, CD4, and CD8 counts between the two trials, the generalized t test was used based on the expectation-maximization algorithm (30). The nonlinear longitudinal data model was used for analyzing the absolute lymphocyte count (ALC) curve relative to time after transplant. A paired Mann-Whitney (Wilcoxon) test was used to evaluate differences in serum cytokine concentrations between enrollment and posttransplant time points. Pearson’s correlation coefficient was used for correlation analyses.
Results

From December 2006 to December 2008, 52 patients were enrolled at the two participating institutions. Table 1 shows the major clinical characteristics for the study patients. After initial immunization, two patients did not mobilize adequate stem cells to proceed to transplant, and therefore, 50 (24 arm A and 26 arm B) patients were autografted. To analyze posttransplant T-cell recovery, both arm A and arm B patients were pooled together.

Clinical effects of day +2 T-cell transfers. The mean and median numbers of T cells infused were $4.26 \times 10^{10}$ and $4.54 \times 10^{10}$, respectively (range, 1.59-5.0). The mean CD4/CD8 ratio was 2.48 (range, 0.62-11.09). The day +2 T-cell transfers were well tolerated with common adverse effects being chills/rigors (67%), nausea (21%), and low-grade fevers (18%; Supplementary Table S1). All early infusion-related toxicities were grade 1/2. Later toxicities that were possibly, probably, or definitely considered to be related to the T-cell infusions are also tabulated (Supplementary Table S1). These toxicities, also mainly grade 1/2, included mild maculopapular rashes (typically involving the face/scalp/neck/upper chest, 51%), fevers (21%), arthralgias (15%), myalgias (10%), and...
conjunctivitis (8%), which was readily responsive to glucocorticoid eye drops. One patient had incomplete platelet recovery (grade 3; \(40-50,000/\mu L\) at 6 months after transplant) considered to be possibly related to the T-cell infusion. There were no documented cases of autoantibody formation (e.g., antinuclear antibodies and thyroid antibodies) nor autoimmune endocrinopathies.

**T-cell recovery.** Lymphocyte recovery was rapid and robust following day +2 T-cell transfers. At about day +5 after stem cell transplant, a relative and absolute lymphocytosis developed, which persisted throughout the transplant period. Hematopoietic recovery occurred simultaneously with the lymphocytosis, which “peaked” at \(\sim 4\) weeks after transplant. Thereafter, the lymphocytosis gradually subsided but the ALC remained at or above the upper limit of the reference range until at least day +100 after transplant. Figure 2A shows a statistically modeled ALC curve with 95% confidence interval for all the patients. There was significant variability in the magnitude of the CD4/CD8/CD3 lymphocyte counts, which were observed at each time point (Supplementary Table S2). Figure 2B shows the most dramatic example of posttransplant T-cell lymphocytosis (MD05). The number of CD4 cells infused was significantly and positively correlated to the day 14 and day 100 CD4 counts (Pearson’s correlation coefficient \(r = 0.285, P = 0.0318\) and \(r = 0.316, P = 0.0284\), respectively).

Figure 2C and D depicts the patterns of CD4+ and CD8+ T-cell recovery in the current myeloma trial versus the previous trial (22). In this previous study, patients were randomized to receive costimulated T cells at either day +12 (blue lines, diamonds) or day +100 (green lines, squares). These plots show that the CD4+ and CD8+ T-cell counts were significantly higher in the current study at all the time points examined, including days +14, +60, +100, and +180 (\(P < 0.001\)).

**Hematopoietic recovery.** To determine whether the early lymphocyte recovery that followed day +2 adoptive T-cell transfer had an effect on hematopoietic recovery, we compared the times to absolute neutrophil counts >500/\(\mu L\) for 2 consecutive days for the current study patients with a historical control population of 102 myeloma patients who had standard autographs without additional T cells (31). The median number of days to neutrophil recovery in the current trial was 12 days (range, 10-18) versus 12 days in the historical cohort (\(P = 0.49\)). Similarly, the median days to an unsupported platelet count >20,000/\(\mu L\) were similar for the two populations: 14 days (range, 0-28) in the current trial versus 14.5 days in the historical cohort (\(P = 0.78\)).

**T-cell engraftment syndrome.** Seven patients (MD03, MD11, MD24, MD30, UP07, UP23, and UP26) developed a posttransplant T-cell “engraftment syndrome” characterized by watery diarrhea (up to 2,000 cm³/d), abdominal pain, and fever (in six
of the patients). In six patients (MD03, MD11, MD24, MD30, UP23, and UP26), this syndrome developed at a median of 14 days after transplant (range, 9-17 days), whereas in one patient (UP07) it occurred around day 60 after transplant. Two patients including UP23 and an eighth patient, UP25, developed early bright facial rashes at about day +9 after transplant. The mean day 14 CD4 count for the seven patients who developed early GVHD-like T-cell engraftment syndromes was 2,443 (range, 863-4,049), whereas the mean day 14 CD8 count for this subset of patients was 5,822 (range, 671-11,571). There were no significant differences in the CD4 and CD8 counts for the subset of patients who developed early “GVHD” versus the group of patients who did not. Because phenotypic changes in the T-cell compartment could also contribute to development of GVHD, the balance of Teff and Treg cells was examined before and after T-cell transfers. Compared with the time of enrollment (baseline measurement), a significant increase in the ratio of Teff to Treg cells was observed at day +14 after transplant: mean log (Teff/Treg) = 2.23 at day +14 versus 1.08 at enrollment (P < 0.0001). However, perhaps due to its small size, the subgroup of patients who developed clinical GVHD did not seem to exhibit a significantly higher Teff/Treg ratio at this time point compared with the rest of the patients.

All seven patients with apparent “gastrointestinal GVHD” had colonoscopic biopsies at a median of 17 days after transplant (range, 15-81). In six patients (MD03, MD11, MD24, MD30, UP23, and UP26), the biopsies were interpreted to show histopathologic grade 2 to 3 GVHD of the gut. The biopsy for the patient with late-onset symptoms (UP07) was considered to be nonspecific, although intraepithelial lymphocytosis and rare apoptotic bodies were observed. Figure 3A depicts a representative computed tomography scan of the abdomen taken at the time of symptom onset for MD11, which shows bowel wall thickening consistent with GVHD. Figure 3B shows a representative colon biopsy from MD11, which shows crypt cell apoptosis, whereas Fig. 3C shows crypt dropout (red arrow) and neuroendocrine hyperplasia (black arrow), typical features of gut GVHD. Immunohistochemical staining for T-cell subsets (Fig. 3D; Supplementary Fig. S1) shows a prominent lymphocytic infiltrate along with intraepithelial localization of CD3+, CD4+, and CD8+ T cells. Figure 4A also shows the grade 3 GVHD histopathology for MD30—the patient with the most severe clinical syndrome that was observed. All seven patients were initially treated with 1 to 2 mg/kg of methylprednisolone and/or oral budesonide followed by a rapid taper over ~1 month and exhibited rapid and complete clinical responses.

Four patients, including MD30, UP23, and UP26 (who also had gastrointestinal GVHD) and UP25 (no gastrointestinal symptoms), developed early skin rashes including generalized erythroderma (MD30) or bright facial rashes (UP23, UP25, and UP26) at about day +9 after transplant. A biopsy of one of these rashes (MD30) showed typical grade 2 skin GVHD with apoptotic keratinocytes, basal vacuolization, and CD3+ T-cell infiltration of the dermis and epidermis (Fig. 4B and C), whereas the skin biopsy of a second patient with facial rash alone (UP25) showed a follicular and eccrine duct-centric infiltration of lymphocytes in the superficial dermis (data not shown). These rashes resolved in ~1 week with (MD30) or without (UP23, UP25, and UP26) systemic glucocorticoids.

**Mechanisms of T-cell recovery.** To explore the mechanisms that may contribute to early posttransplant recovery and expansion of transferred T cells, serum levels of various cytokines were measured serially before and after transplant. Serum levels of interleukin (IL)-2, IL-4, IL-6, IL-7, IL-12, IL-17, and IFN-γ were measured for 21 consecutive patients at enrollment, T-cell collection, day +14 after transplant, day +60, day +100, and day +180 using the Luminex multiplex cytokine assay system. By this method, only IL-6 levels showed a significant increase (P < 0.03) during the early posttransplant period, whereas IFN-γ levels showed a modest but significant decrease at day +14 compared with enrollment (P = 0.03); the levels of IL-2, IL-4, IL-7, IL-12, and IL-17 did not significantly change between these time points. Figure 5 depicts the cytokine level changes for IL-6 and IFN-γ in terms of percentage change compared with enrollment, although

![Fig. 4. A, grade 3 GVHD of gut (MD30) showing apoptotic crypt cells (thin arrow) and complete crypt dropout (thick arrow). B, grade 2 GVHD of skin (MD30) with apoptotic keratinocytes (black arrow) and basal vacuolization and degeneration. C, intraepidermal and i.d. CD3+ T cells (MD30).](https://www.aacrjournals.org/clinica/content/15/13/4504/F4.large.jpg)
the statistical analysis was based on the serum cytokine concentrations. No significant correlation between IL-6 levels and T-cell recovery parameters could be established. Thirty consecutive sets of serum samples were also assayed for IL-15 levels using a standard ELISA assay, and a statistically significant increase (+2-fold) was observed at day +14 compared with enrollment (P < 0.0001; Fig. 5). In addition, highly significant inverse correlations were found between the day 14 IL-15 levels and the day 14 ALC (r = -0.452, P = 0.0054), day 14 CD3 count (r = -0.399, P = 0.0177), day 14 CD4 count (r = -0.467, P = 0.0071), and day 14 CD8 count (r = -0.386, P = 0.0234). About midway through the study, the amendment was mandated to allow collection of serum samples at day +2 after transplant (just before T-cell transfers), and thus, a limited number of patients could also be assayed for day +2 IL-15 levels. The mean IL-15 level at day +2 was more than five times higher than at enrollment (P < 0.0001). Furthermore, a positive correlation was found between day +2 IL-15 levels and day +14 CD8 counts (Pearson’s correlation coefficient r = 0.6583, P = 0.038). Positive correlations between day +2 IL-15 levels and day +14 CD3 and CD4 counts were marginally or nonsignificant (CD3: r = 0.548, P = 0.059; CD4: r = 0.5168, P = 0.095) perhaps due to the small numbers of patients who were tested.

Functional immune studies. One purpose of the current study was to confirm and perhaps extend the observations of robust and early recovery of B- and T-cell function that were made in the previous study of combined vaccine and T-cell immunotherapy (22). Based on 29 patients analyzed to date, the estimated mean total pneumococcal antibody response (sum of titers for serotypes 6B, 14, 19F, and 23F) at day 100 after transplant was 98.7 μg/mL in the current trial versus 138.6 μg/mL in the earlier trial (P = 0.45). At day 100 after transplant, the mean percentage of CD4+/CD25+ T cells that proliferated (CFSE<sup>dim</sup>) in response to the CRM-197 carrier protein was 6.0% (n = 22) in the current trial versus 7.1% at day 114 (n = 8) in the previous trial (P not significant). Based on 15 total arm A (HLA-A2<sup>+</sup>) patients analyzed to date, 6 patients (40%) had positive tetramer responses at one or more time points after immunization, defined as tetramer staining by flow cytometry >0.1% (and >3-fold increase versus enrollment/baseline).

Discussion

High-dose therapy in conjunction with autologous stem cell rescue can achieve a minimal disease state and improve disease-free survival for patients with myeloma and other hematologic malignancies. However, relapses are common due to the lack of an effective antitumor immune response to eliminate residual disease. In principle, the period following high-dose therapy should be conducive to the application of immunotherapy given the low tumor burden and the potential reduction in Treg populations. Unfortunately, following standard autologous transplants, the immune system is typically characterized by immune cell depletion and impaired immune cell function, which may persist for up to 4 to 10 years (32, 33).

In a previous randomized study, we found that a single infusion of ~8 × 10<sup>9</sup> costimulated autologous T cells at day +12 after transplant accelerated the numerical and functional recovery of both CD4<sup>+</sup> and CD8<sup>+</sup> T cells and could provide help for vaccination during the early posttransplant period (22). Passenger lymphocytes in the autologous peripheral blood stem cell products alone were insufficient for these purposes.

We hypothesized that adoptive transfer of costimulated T cells at day +2 after transplant might take greater advantage of early posttransplant homeostatic lymphocyte expansion mechanisms because lymphopoietic cytokine levels peak very soon after lymphodepleting chemotherapy (34). Indeed, results from the current study showed that robust CD4<sup>+</sup> and CD8<sup>+</sup> T-cell counts developed as early as day 14 after transplant. T-cell counts were significantly higher at days 14, 60, 100, and 180 after transplant than in the previous study when costimulated T cells were infused at day +12 or day +100 after transplant. These data indicate that T-cell recovery was augmented by earlier T-cell transfer, although the 5-fold greater number of T cells that were infused in the present study may also have contributed to the enhanced recovery, which was observed particularly at later time points.

The mechanisms responsible for the posttransplant T-cell recovery are unclear but may include T-cell growth stimulation by host-derived cytokines. IL-7 and IL-15 play important roles in lymphocyte homeostasis, and in animal models of allogeneic transplantation, IL-7 and IL-15 administration improved posttransplant immune reconstitution (34–36). Of the cytokines assayed, only IL-6 and IL-15 showed significant increases between the baseline/enrollment time point and day 14. IL-6 is known to increase early after allogeneic stem cell transplantation in conjunction with infections or GVHD (37, 38). To further analyze whether the increased IL-6 or IL-15 levels might contribute to the early T-cell recovery, a correlation analysis was done between day 14 IL-6 or IL-15 levels and day 14 T-cell counts. Notably, IL-6 levels did not correlate to any T-cell recovery parameter, whereas IL-15 levels correlated significantly but inversely to day 14 ALC, CD3<sup>+</sup>, CD4<sup>+</sup>, and CD8<sup>+</sup> counts. The inverse correlation between IL-15 levels and T-cell levels is consistent with the model that IL-15 activity is regulated by host-derived cytokines. IL-7 and IL-15 play important roles in lymphocyte homeostasis, and in animal models of allogeneic transplantation, IL-7 and IL-15 administration improved posttransplant immune reconstitution (34–36). Of the cytokines assayed, only IL-6 and IL-15 showed significant increases between the baseline/enrollment time point and day 14. IL-6 is known to increase early after allogeneic stem cell transplantation in conjunction with infections or GVHD (37, 38). To further analyze whether the increased IL-6 or IL-15 levels might contribute to the early T-cell recovery, a correlation analysis was done between day 14 IL-6 or IL-15 levels and day 14 T-cell counts. Notably, IL-6 levels did not correlate to any T-cell recovery parameter, whereas IL-15 levels correlated significantly but inversely to day 14 ALC, CD3<sup>+</sup>, CD4<sup>+</sup>, and CD8<sup>+</sup> counts. The inverse correlation between IL-15 levels and T-cell levels is consistent with the model that IL-15 activity is regulated...
through binding to circulating T cells so that large numbers of T cells act like a cytokine “sink” for IL-15. Thus, IL-15 levels, which rise within days after completion of high-dose therapy, drive T-cell expansion but fall quickly by day 14 as the T cells recover (39, 40). Indeed, the lowest value that was measured for IL-15 at day 14 after transplant (1.0 pg/mL) occurred in the patient MD05 who had the most rapid and robust T-cell recovery (Fig. 2B). In keeping with this model, studies of IL-15 levels at day +2 after transplant (just before T-cell transfers) showed a significant positive correlation with day +14 CD8 counts.

One potential concern about the day +2 transfers of costimulated T cells was the effect on hematopoietic recovery. However, a comparison of neutrophil and platelet recovery in the current study with a large cohort of myeloma patients who received autographs without costi-
mulated T cells showed no significant differences in re-
covery kinetics.

An unexpected observation was that a subset of patients (16%) developed an early T-cell engraftment syndrome characterized by watery diarrhea and fever that was clinically and histopathologically indistinguishable from moderate to severe acute GVHD of the gastrointestinal tract and mild to moderate GVHD of the skin. All patients with diarrhea responded rapidly and durably to courses of moderate-dose systemic or nonabsorbable glucocorticoids. Autologous GVHD of the skin has been previously recognized in autograft patients who received autographs without costi-
mulated T cells showed no significant differences in re-
covery kinetics.

Autologous GVHD of the gastrointestinal tract is much rarer than autologous GVHD of the skin. In one prospective report of 97 autotransplant patients who received cyclosporine A, only 4 (4%) developed gastrointestinal GVHD; all cases were mild (grade 1) and only 2 cases were confirmed by biopsy (45). A larger retrospective series of 681 autograft recipients reported a frequency of gastrointestinal GVHD of 13%, but most of these cases involved the upper gastrointestinal tract causing nausea and vomiting: classic symptoms of diarrhea occurred in only 5% of patients and were mild in all cases (46). In addition, biopsy confirmation of gastrointestinal GVHD in this series occurred at mean of 42 days after transplant. This previously published experience contrasts with the present study, which chiefly involved the face, scalp, neck, and chest.

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Disclosure of Potential Conflicts of Interest

R.H. Vonderheide declares a potential financial conflict of interest related to inventorship on a patent about hTERT as a tumor-associated antigen for cancer immunotherapy. C.H. June has patents and patent applications in the field of adoptive immunotherapy but has been divested of financial benefit from this technolo-
gy. This arrangement is under compliance with the policies of the University of Pennsylvania. The other authors disclosed no potential conflicts of interest.

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