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Induction of Immune Response after Allogeneic Wilms' Tumor 1 Dendritic Cell Vaccination and Donor Lymphocyte Infusion in Patients with Hematologic Malignancies and Post-Transplantation Relapse

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A B S T R A C T

Relapse of hematologic malignancies is the primary cause of treatment failure after allogeneic hematopoietic stem cell transplantation (HCT). Treatment for post-HCT relapse using donor lymphocyte infusion (DLI) has limited utility, particularly in the setting of acute leukemia, and can result in the development of graft-versus-host disease (GVHD). The Wilms' tumor 1 (WT1) gene product is a tumor-associated antigen that is expressed in acute leukemia and other hematologic malignancies, with limited expression in normal tissues. In this pilot trial, we assessed safety and feasibility of a WT1 peptide–loaded donor-derived dendritic cell (DC) vaccine given with DLI designed to enhance and direct the graft-versus-leukemia effect. Secondary objectives were to evaluate immunologic and clinical responses. A total of 5 subjects, median age 17 years (range, 9 to 19 years), with post-HCT relapse were enrolled. Disease subtypes included acute lymphoblastic leukemia (n = 3), acute myelogenous leukemia (n = 1), and Hodgkin lymphoma (n = 1). Successful vaccine production was feasible from all donors. DC vaccination and DLI were well tolerated. One recipient developed grade 1 skin GVHD not requiring systemic therapy. The most common adverse events included grade 1 reversible pain and pruritus at the vaccine injection and delayed-type hypersensitivity (DTH) skin testing sites. There were no grade 3 or higher adverse events related to the research. Immune responses consisted of ELISpot response in 3 recipients and positive DTH tests to WT1 peptide cocktail in 2 subjects. Our study provides 1 of the first attempts to apply tumor-specific vaccine therapy to the allogeneic setting. Preliminary results show the DC-based vaccination is safe and feasible after allogeneic HCT, with a suggestion that this approach can be used to sensitize the repopulated allogeneic-donor immune system to WT1. Future directions may include testing of vaccination strategies in the early post-transplantation setting for relapse prevention.

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INTRODUCTION

Allogeneic hematopoietic stem cell transplantation (HCT) can be curative for many hematologic malignancies with efficacy due, in part, to an allogeneic graft-versus-leukemia (GVL) effect. However, relapse remains the primary cause of treatment failure after transplantation. Post-transplantation therapies, including donor lymphocyte infusions (DLI), have variable effectiveness in the treatment of post-transplantation relapse, with especially limited potency in the setting of acute leukemia [1,2]. Novel therapies to address post-transplantation relapse are needed [3,4].

Dendritic cells (DCs) are professional antigen-presenting cells that can be readily generated from peripheral blood monocytes. Tumor vaccine trials using purified populations of DCs as antigen-presenting cells have been reported [5,6]. The Wilms' tumor 1 (WT1) gene product is a tumor-associated antigen that represents a potential target for immunotherapy in hematologic malignancies [7-11]. WT1 is expressed in most cases of acute leukemia and in many cases of chronic myelogenous leukemia and myelodysplastic syndromes [7,8].

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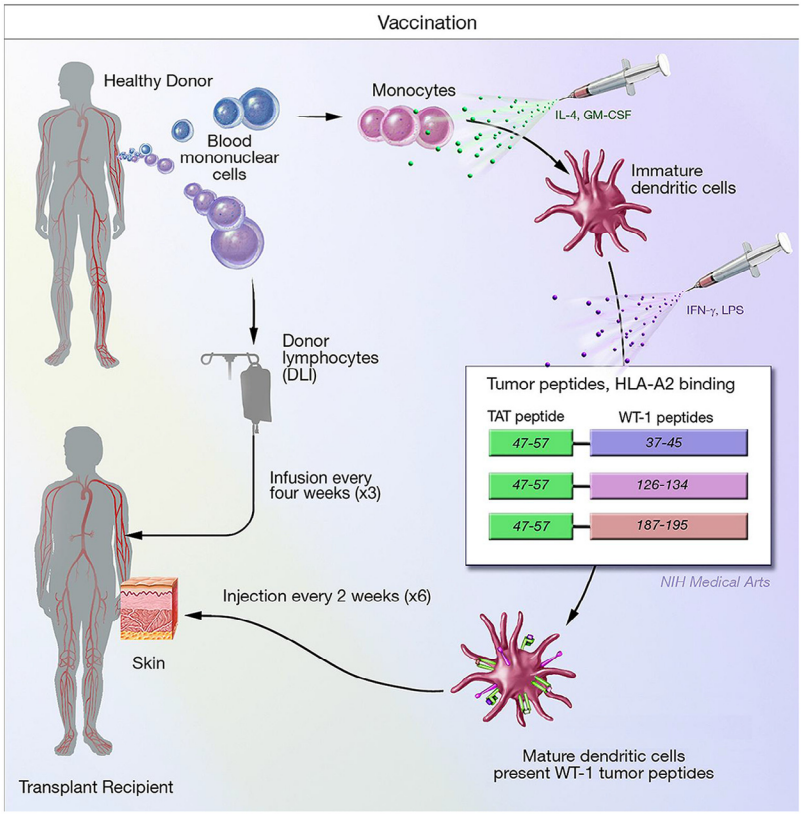

Figure 1. Protocol schema.

Importantly, WT1 has limited expression in normal tissue beyond embryogenesis [12,13]. Promising clinical results, including evidence for induction of immune responses, using autologous monocyte-derived DC WT1 vaccines have been observed [14,15]. Effective antitumor immune responses after vaccination, however, may be impaired because of host immune depletion associated with standard anticancer therapies, which may be more profound in the post-transplantation setting. This might be overcome by the use of allogeneic approaches. Experience with DC vaccination, particularly in the allogeneic post-transplantation setting, is limited [16-19].

We describe results from a pilot trial that incorporates antigen-specific immunotherapy and allogeneic adoptive cell transfer for pediatric and adult patients with relapsed hematologic malignancies after allogeneic HCT using GMP methods for ex vivo generation of monocyte-derived DCs [20]. The primary objective was to assess safety and feasibility of a WT1 peptide-loaded, HLA-A2 restricted, allogeneic, donor-derived DC vaccine designed to enhance the GVL effect of coadministered DLI. A secondary objective was to determine if immunologic and clinical responses to WT1-specific peptides could be generated by this novel allogeneic vaccine strategy for treatment of post-HCT relapse. As 1 of the few experiences reporting on the use of DC vaccination in the post-allogeneic transplantation setting, our results demonstrate feasibility of this platform, which importantly helps in setting the stage for future vaccination strategies in the post-transplantation setting, potentially for relapse prevention.

MATERIALS AND METHODS

Patients

This was a single-institution pilot study for HLA-A2 positive recipients between the ages of 1 and 75 years with WT1-expressing hematologic malignancies who experienced relapse after allogeneic HCT. HLA-A2 positive, healthy, related or unrelated donors who were 5- or 6-antigen (or 8 to 10/10 allele) genotypic HLA-matched (single HLA-A or -B locus mismatch allowed) were eligible. WT1 expression of the hematologic malignancy was confirmed by either having greater than 15% of malignant cells react with anti-WT1 by immunohistochemistry or by having a positive quantitative RT-PCR of WT1 compared with a negative control using approaches that have been previously described [21]. Recipients with rapidly progressive disease, with greater than 25% marrow blasts (in the setting of acute leukemia), with active graft-versus-host disease (GVHD) greater than grade 1 or those on immunosuppression were not eligible. This study was approved by the institutional review boards of the National Cancer Institute and the National Marrow Donor Program. Parental consent with age appropriate assent was employed as needed for pediatric enrollment. This trial is registered at clinicaltrials.gov (NCT00923910).

Study Design

All recipients received a DLI product (dose at 1×10^6 CD3/kg) once every 4 weeks for a total of 3 DLIs and a DC vaccine once every 2 weeks, for a total of 6 vaccines. To manufacture the DC vaccine, a peripheral blood mononuclear cell concentrate was collected from each donor by apheresis. The peripheral blood mononuclear cell concentrates were enriched for monocytes by counterflow elutriation and the monocytes were cryopreserved. An aliquot of monocytes was thawed incubated for 3 days with granulocyte macrophage–colony stimulating factor (10 micrograms/mL) and IL-4 (2000 IU/mL) in RPMI with 10% heat-inactivated AB plasma, followed by maturation for 1 day with LPS (30 nanograms/mL) and IFN-gamma (1000 IU/mL). The mature DCs were loaded for 2 hours with a combination of 3 WT1-derived HLA A2 binding peptides (WT1 37-45, WT1 126-134, WT1 187-195). Each peptide was linked to the 11-mer HIV TAT protein transduction domain known to enhance peptide loading and antigen presentation [22-24]. Additionally, keyhole limpet hemocyanin (KLH), a neoantigen known to induce helper response, was used concurrently as a vaccine adjuvant and control antigen (Figure 1). Vaccines were administered in 2 forms: subcutaneous at a dose of 10×10^6 DCs and intradermal injection at a dose of 2×10^6 DCs.

Immune Surveillance

Immune responses were evaluated every 4 weeks after initiation of protocol therapy and was monitored by use of interferon gamma ELISpot and