

C.2 Updated Technical Abstract

Protocol K-0009 titled "Vaccination in Peripheral Stem Cell Transplant Setting for Acute Myelogenous Leukemia: The Use of Autologous Tumor Cells with an Allogeneic GM-CSF-producing Bystander Cell Line".

Acute myelogenous leukemia (AML) is relatively rare but disproportionately lethal. The approximate prevalence in the US is 2.5 in 100,000 (0.0025%), with males predominating at 1.5 to 1. The median age at diagnosis is 65 years. The etiology is primarily unknown, although prior chemotherapy, radiation exposure, and routine occupational exposure to benzene comprise a few known causes. Signs and symptoms of AML are due to progressive replacement of the normal hematopoietic cells in the bone marrow with leukemia cells, and include anemia, thrombocytopenia, hemorrhage, petechiae / ecchymoses, fever, infection, hepatosplenomegaly, persistent fatigue or malaise, weight loss, and bone or abdominal pain. If untreated, AML leads to death only months from diagnosis. Even with treatment, long-term survival (> 3 years) in adults is only ~30%. Although therapy is toxic, complete response rates to first-line induction chemotherapy are high, approximately 70%. However, in the absence of post remission therapy relapse occurs in nearly all patients.

The standard treatment for AML includes induction chemotherapy followed by post-remission therapy for those patients who obtain complete remission. Acceptable post-remission therapies include consolidation chemotherapy and autologous or allogeneic stem cell transplant (SCT). Allogeneic transplants have the advantage of inducing an immune-mediated graft-vs-leukemia (GVL) effect, but are limited in their overall efficacy and feasibility by graft-vs-host disease (GVHD) and the lack of HLA-matched donors for many patients. Autologous transplants are associated with less toxicity and correspondingly lower rates of transplant-related mortality but are associated with higher relapse rates ultimately leading to similar long term outcome to allogeneic transplant. However, autologous transplants offer the opportunity to incorporate post-transplant immunotherapy to induce a tumor-specific immune response without GVHD. This trial seeks to effectively integrate a tumor cell-based vaccine (Leukemia Bystander GVAX) into the setting of autologous SCT for the treatment of patients with acute myelogenous leukemia. This is a Phase I/II, open-label, single-arm, multicenter clinical trial in patients with newly diagnosed acute myelogenous leukemia (AML). Leukemia Bystander GVAX is a cancer vaccine consisting of two components: autologous tumor cells (leukemia cells), and CG9962 cells (K562 bystander GVAX), a genetically modified, granulocyte-macrophage colony-stimulating factor (GM-CSF)-secreting, major histocompatibility complex I/II negative, erythroleukemia cell line.

Objectives

The primary objectives of the study are to evaluate clinical and laboratory safety associated with administration of Leukemia Bystander GVAX; to determine feasibility of generation of Leukemia Bystander GVAX in subjects with AML; and to demonstrate the priming of tumor-specific immunity in response to vaccination with Leukemia Bystander GVAX using clinical and

laboratory endpoints. The secondary objectives are to evaluate additional immunologic and clinical responses to vaccination with Leukemia Bystander GVAX.

Patient Population

Fifty male and female patients, ages 18 to 60 years-old with *de-novo* AML other than M3-AML.

Study Design

Phase I/II open-label, multi-center study.

Treatment Plan and Schedule

Patients are enrolled and undergo leukemia cell harvest at the time of first diagnosis of AML. Leukemia cells are collected via peripheral blood draw, leukapheresis, or bone marrow aspiration. Eligible patients who achieve complete remission (CR) with induction chemotherapy and qualify for autologous peripheral stem cell transplant (PSCT) undergo consolidation chemotherapy and peripheral blood stem cell (PBSC) collection. Patients are then vaccinated approximately 2 weeks following the PBSC collection, and 14 days following vaccination, patients undergo leukapheresis to collect "vaccine-primed" lymphocytes for reinfusion with the PBSC graft. Patients proceed to autologous PSCT with busulfan and cyclophosphamide as the preparative regimen followed by PBSC and vaccine-primed lymphocyte infusion. On day 5 post-transplant, subjects begin GM-CSF 125 or 250 µg daily until neutrophil engraftment is documented. Eight post-transplant vaccinations are then given beginning 6 weeks post-transplant or at the time of hematopoietic recovery.

Dose

Each vaccination contains 1×10^8 tumor cells admixed with 4×10^7 CG9962 cells administered by multiple intradermal injections. Vaccinations are given once pre-transplant and every 3 weeks post-transplant beginning at week 6 for eight additional vaccinations (9 vaccinations total).

Endpoints

The primary endpoints of the trial are: 1) To evaluate clinical and laboratory safety associated with administration of Leukemia Bystander GVAX; 2) To determine the feasibility of generation and administration of Leukemia Bystander GVAX in subjects with AML; and 3) To demonstrate the priming of tumor-specific immunity in response to vaccination with Leukemia Bystander GVAX using clinical and laboratory endpoints. The secondary endpoints are: 1) To characterize the kinetics of immune reconstitution and assess immunocompetence at regular intervals before and after transplant; 2) To demonstrate the ability to generate tumor-specific immune responses prior to transplant and to demonstrate adoptive transfer of immunity through the infusion of these "primed" lymphocytes as part of the peripheral blood stem cells (PBSC) graft; 3) To monitor levels of minimal residual disease pre and post-transplant using quantitative PCR techniques; and 4) To identify relapse-free and overall survival associated with the intradermal injections of Leukemia Bystander GVAX in the PSCT setting.

Safety is being monitored by recording adverse events and laboratory evaluations. Feasibility is being assessed by the overall success rate of tumor harvest and autologous tumor cell processing. Immunologic response will be measured by DTH reaction to injections of irradiated, autologous tumor cells, in vitro monitoring of tumor-specific B and T cell responses, and overall monitoring of immune reconstitution. Clinical activity will be assessed by molecular monitoring of minimal residual disease by quantitative PCR for the leukemia-associated antigen, WT-1, and by monitoring of time to relapse and overall survival. The study, however, is not powered to draw any meaningful conclusions about the relative impact of vaccination on relapse or survival compared to historical data.

Product

Leukemia Bystander GVAX consists of two components, CG9962 cells and autologous, irradiated tumor cells mixed just prior to administration at a ratio of 5 tumor cells to 2 CG9962 cells. The CG9962 cell product was generated from a K562 erythroleukemia cell line engineered by plasmid transfection to secrete human GM-CSF. These cells are grown in suspension, irradiated at 10,000 rads to arrest cell growth, formulated in a dimethyl-sulfoxide (DMSO)-containing cryoprotectant and frozen in liquid nitrogen. Autologous tumor cells are obtained by peripheral blood draw, leukapheresis, or bone marrow aspiration, processed to isolate mononuclear cells, irradiated at 10,000 rads, frozen in autologous serum and DMSO, and stored in liquid nitrogen. Tumor and CG9962 cells are thawed just prior to administration, mixed together, and injected intradermally without further manipulation.

Trial Status

This phase I/II study of Leukemia Bystander GVAX is currently ongoing. Enrollment is nearly complete and patient treatment and data analysis is in process. There are no plans to initiate any new trials with this vaccine product this year.