SCIENTIFIC ABSTRACT

Most pediatric patients with B cell acute lymphoblastic leukemias (B-ALL) are cured with standard chemotherapy regimens. However, a subset of patients will experience relapsed disease and require further therapy with allogeneic bone marrow transplantation (allo-BMT). While this additional therapy may successfully cure a number of these patients, too many will ultimately relapse and die from their disease. This latter patient population has a very poor prognosis in the setting of current treatment strategies and urgently requires novel therapeutic approaches to treat their refractory disease.

We have previously generated a chimeric antigen receptor (CAR) specific to the CD19 antigen (19-28z) expressed on normal B cells and most B cell malignancies. T cells retrovirally modified to express the 19-28z CAR demonstrate significant preclinical anti-tumor activity in murine tumor models. The 19-28z CAR is a second-generation receptor that is composed of the anti-human CD19 single-chain variable fragment, CD28 transmembrane and signaling domain, and the CD3 ζ signaling domain. We have cloned the 19-28z CAR into the SFG retroviral vector, which was derived and adapted from the Moloney murine leukemia retrovirus and permits the transduction of human T cells. *In vitro*, retrovirally transduced 19-28z CAR^+ T cells from healthy donors and patients with leukemia efficiently targeted and lysed $CD19^+$ B cell tumors. *In vivo*, treatment with 19-28z CAR^+ T cells in SCID-Beige mice eradicated systemic human B cell tumors and achieved a long-term survival. Lastly, we have demonstrated that pretreatment of mice with cyclophosphamide enhances 19-28z gene-modified T cell survival and function.

Our preclinical studies have led to the development of currently ongoing clinical trials for patients with relapsed CLL (ORB protocol #0507-721) and acute lymphoblastic leukemia (ORB protocol #0907-985) with autologous $19\text{-}28z^+$ gene-modified T cells. We propose to extend CAR modified T cell therapy to pediatric patients with relapsed $CD19^+$ B-ALL following allo-BMT. In contrast to the use of autologous patient derived T cells, this protocol following allo-BMT, is restricted to donor derived T cells which carry a significant risk of graft vs host disease (GVHD). To mitigate this risk we will utilize EBV specific donor T cells. Prior clinical trials have demonstrated that donor derived EBV specific T cells do not induce graft versus host disease, therefore treatment of relapsed B-ALL patients with EBV specific donor T cells further modified to express the 19-28z CAR will result in minimal toxicity yet allow for optimal CD19 targeted anti-tumor effect.

This is a standard phase I dose-escalation study treating relapsed pediatric B-ALL following allo-BMT with donor derived Epstein-Barr virus (EBV) specific T cells further modified to express the 19-28z CAR. Primary objective of this study is to assess the safety of 19-28z gene-modified T cells in patients with relapsed B-ALL with a secondary objective to determine the anti-tumor efficacy and persistence of 19-28z gene-modified T cells. This clinical protocol represents a novel promising and much therapeutic approach for the treatment of pediatric patients with post allo-BMT relapsed B-ALL.