Children's Hospital of Philadelphia

Annual Progress Report: 2011 Formula Grant

Reporting Period

July 1, 2014 – June 30, 2015

Formula Grant Overview

The Children's Hospital of Philadelphia received $3,521,179 in formula funds for the grant award period January 1, 2012 through December 31, 2015. Accomplishments for the reporting period are described below.

Research Project 1: Project Title and Purpose

Highly Active Cell Therapy of Cancer – Our purpose is to develop engineered T cell therapies for B cell malignancies, leukemias, and certain specific solid tumors such as neuroblastoma and synovial sarcoma. Using chimeric antigen receptors (CARs) which target tumors and activate T cells (CART cells), and an efficacious clinical-grade (GMP) ex vivo cell manufacturing system, we will continue our highly promising use of CAR-engineered T cells. This grant will support preclinical studies to optimize CARs in mouse xenograft models, as well as early phase clinical trials testing a variety of CAR-mediated T cell therapy approaches.

Anticipated Duration of Project

1/1/2012 – 12/31/2015

Project Overview

The overall goal of this Pennsylvania Department of Health Formula Project is to develop clinically efficacious methods of treating high-risk and relapsed leukemia, lymphoma and some solid tumors with chimeric antigen receptor (CAR)-armed T cells. The long-term goal of our cell therapy group is to establish improved treatments for hematologic malignancies and other tumors by engineering, optimizing, and clinically testing these highly active anti-cancer T cells. This treatment approach could potentially obviate the need for allogeneic stem cell transplant for some patients. Our preliminary clinical data showing cures of patients with high burdens of refractory tumor provide the first clear proof of concept and proof of mechanism for an anti-cancer cell therapy, and are potentially paradigm-shifting. They suggest that the central problems of expansion and persistence of therapeutic cells in the patient after infusion are solvable using the right cell manufacturing system and the right CAR design. With successful expansion and long-term persistence in the patient, even small doses of T cells can lyse very large tumor burdens. In order to leverage the dramatic results we have seen and continue to develop these

Note: This project is funded, in part, under a grant with the Pennsylvania Department of Health. The Department specifically disclaims responsibility for any analyses, interpretations or conclusions.

approaches for pediatric cancer patients, we are proposing a 4 year combined basic/translational cell therapy program for CHOP. We propose 3 Aims to build on these results and establish cell therapy infrastructure at CHOP:

Aim 1. Develop novel CARs targeting antigens other than CD19, and develop RNA transfection as an alternative approach to lentiviral transduction to temporarily express CARs on T cells.

Aim 2. Perform 3 cell therapy trials testing CD19-targeted CAR+ T cell approaches in patients with B cell malignancies such as acute lymphoblastic leukemia (ALL), CLL and non-Hodgkin lymphoma (NHL).

Aim 3. Using CARs and approaches developed in Aim 1, take engineered T cell-based therapy into trials enrolling non-B cell cancers, including sarcoma (target NY-ESO-1), acute myelogenous leukemia (AML; target mesothelin), and neuroblastoma (target GD2).

Principal Investigator

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Other Participating Researchers

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Expected Research Outcomes and Benefits

Expected outcomes and benefits include the following:

1. Initiate a trial to test CAR-T cell therapy in children with CD19+ malignancies. CARs are chimeric antigen receptors that redirect T cells to cancer cells and activate the T cells so they kill the tumor. Any cancer target or tumor-associated antigen which is recognized by an antibody can in principle be made into a CAR.
2. Develop mRNA-based CARs to supplement permanent modification of T cell by lentivirus. We anticipate gaining a better understanding of the performance of mRNA CAR+ T cells in animal models that will guide our use of them in the clinic.
3. A better understanding of the nature and phenotype of long-term engrafting T cells and their impact on disease control in animal models.
4. Extend the CD19 CAR concept to mRNA CARs and to the allogeneic transplant setting.
5. Develop CARs that can target non B cell tumor antigens, to test CARs against AML and solid tumors.

6. Further develop clinically relevant xenograft models of pediatric cancer, translating data from these models to clinical trial design.
7. The overall goal is improve treatment options for patients with high-risk and relapsed cancers.

Summary of Research Completed

Milestones for this year:

- *Open a 4th cell therapy trial at CHOP*. Accomplished. Current trials open at CHOP are:
    - CHP959 IRB 7706 CART19 engineered T cell trial: post stem cell transplant (SCT or allo) cohort* and non-SCT cohort*
      * combined into a single trial at FDA request
    - 12BT074 IRB 9915 biology trial – Lymphocyte functional capacity for immunotherapy
    - 13BT022 IRB 10763 – humanized CART19 trial
    - 11BT053 IRB 8648 – NY-ESO1 T cell receptor (TCR)-engineered T cells for synovial sarcoma
    - B2205J IRB 10900 –Phase 2 multisite trial of CART19 (CHOP is lead center of 9 US centers, Dr. Grupp is PI and Study Steering Committee Chair).
    - B2202 IRB 11485 – Global registration trial of CART19 (CHOP is lead center of planned 15-20 worldwide centers, Dr. Grupp PI and Study Steering Committee Chair).
    - 14BT055 IRB 11296 – RNA CART19 for Hodgkin disease. Study is open, pending first enrollment
      Trials which are written and in process of opening:
    - 15CT014 IRB 12071 – Neuroblastoma GD2 (disaloganglioside) CAR trial. IND is FDA approved, has just gone to the IRB.
    - 15CT055 – CART22 study for CD19 negative relapse. IND (Individual New Drug application) is submitted and the protocol will go to the IRB in July.
- *Accrue a total of 10 patients to the cell therapy trials (cumulative)*. Accomplished.
    - Total accrual to CHP959 on both cohorts is 55 patients
    - 12BT074, 81 patients (lymphocyte biology study)
    - 13BT022, 6 patients
    - 11BT053, 1 patient
    - B2205J, 9 enrolled at CHOP, 25 overall
    - B2202, 5 enrolled at CHOP (other sites are pending)
- *Collect biology samples on each treated/infused cell therapy patient*. Accomplished. We have had 100% of patients with samples collected and >98% individual sample collection compliance on each study.
- *Report results from lab studies at a national meeting*. Accomplished – multiple abstracts at ASH and other national meetings. Two such references are footnoted below, and all abstract references will be provided in the final report.
- *Write and submit a manuscript on lab/clinical results*. Accomplished – please see references #2, 5, 7, 10 below as well as the other New England Journal paper that we have reported in a prior grant report (Grupp et al NEJM 2013).

2015 Progress Report

The CHOP Cancer Immunotherapy Frontier Program (CIFP) has made significant progress over the past year in both clinical trials and lab work exploring T cells engineered with CARs and T cell receptors (TCRs).

Summary of current work for Aims 1 and 3:

CD123 RNA CARs for AML. We have identified CD123 as our lead clinical candidate for AML RNA CAR therapy. This work was published last grant period (Gill et al, Blood, 2014). In the last year, we have performed toxicity testing using this RNA-transfected T cell platform, and no unacceptable toxicity was observed. A trial has been written and an IND is being prepared. Enrollment of an adult patient at CHOP or Penn will be required before patients under 18 can be enrolled at CHOP. Target for opening the protocol is September.

GD2 RNA CARs for neuroblastoma: We have completed our initial preclinical studies (reported previously in our prior progress reports), and recently published a paper on GD2 CARs (see ref #2, below). We have made GMP (Good Manufacturing Practice, aka clinical grade) GD2 RNA for use in a clinical trial. The clinical trial is written and the IND is approved by the FDA, so we anticipate our first enrollment and treatment in August.

Aim 2 (ALL and NHL): Status of the clinical trials. We continue to accrue children to both prior allo SCT and non-allo SCT cohorts on CHP-959: Pilot Study of Redirected Autologous T Cells Engineered to Contain Anti-CD19 Attached to TCRz And 4-1BB Signaling Domains in Patients with Chemotherapy Resistant or Refractory CD19+ Leukemia and Lymphoma.

We have reported results from 25 pediatric and 5 adult patients, which was published in the New England Journal of Medicine during this grant period (reference #1 below; reported as being in press previously). We recently updated these data at the American Society of Hematology and European Bone Marrow Transplant meetings over the past year. This trial of chimeric antigen receptor (CAR)-modified T cells targeting CD19 was developed for patients with relapsed/refractory ALL. Autologous T cells transduced with a CD19-directed CAR (CART19/CTL019) lentiviral vector were infused into patients with relapsed/refractory ALL. Patients were monitored for response, toxicity, expansion and persistence of circulating CART19 T cells. At our most recent update, 48 patients with relapsed/refractory ALL received CART19. Complete responses were achieved in 45/48 patients (94%), including 3 blinatumomab-refractory patients and 30 with prior SCT. CART19 cells proliferated in vivo and were detectable in blood, bone marrow, and cerebrospinal fluid of responding patients. Sustained remissions were achieved with 6-month overall survival of 81%, and 12 month overall survival of 78%. Probability of 6-month CART19 persistence was 68% and relapse-free B cell aplasia was 73%. All but one patient experienced grade 1-4 cytokine release syndrome (CRS). Severe (grade 4) CRS, seen in 31% of patients, was associated with higher disease burden and effectively treated with the anti-IL-6 receptor antibody tocilizumab. These results from the ongoing CART19 study show that CAR-modified T-cell therapy against CD19 is effective for relapsed/refractory ALL. CART19 achieved a high remission rate even in patients for whom previous SCT had failed, and

durable remissions up to 3 years have been observed without subsequent therapy such as bone marrow transplant.

Phase 2 US multisite trial. These study results have resulted the development of a phase 2 multicenter trial in pediatric ALL, which opened on 7/16/14 and which has now enrolled 25 patients at 8 pediatric centers, 9 of which were treated at CHOP. No results of this trial have been published yet, but the CR rate is similar to what we have been seeing in CHP959.

Breakthrough Therapy designation for CART19 (CTL019 or tisagenlecluecel-T) and Global registration trial. More recently, we have worked with the drug company Novartis, who has licensed this technology, to design a global multisite trial with the goal of providing registration data to the Food and Drug Administration. This is the final step toward our ultimate goal of being able to turn this CURE-supported cell therapy into an FDA approved therapy. This is supported by the decision last summer by the FDA to award this therapy with the Breakthrough Therapy designation, the first cell therapy treatment to receive this designation and the first time an academic group has received such a designation. To have gone from treating our first patient in 2012 with CURE grant support, a young lady who remains in remission 3 years later and who has met the President (Figure 1), to a potential registration trial with regulatory submission that could occur to the FDA by the end of 2016, is significant progress. This also represents economic activity in the Commonwealth of Pennsylvania that is vastly in excess of the CURE Grant support, due to tens of millions of dollars of investment from the drug company Novartis in CTL019 development at Penn and CHOP, as well as >$30M in clinical activity at CHOP, 90% coming from patients from outside the Philadelphia area.

Figure 1 CHOP Cell Therapy patient Emily Whitehead meets the President

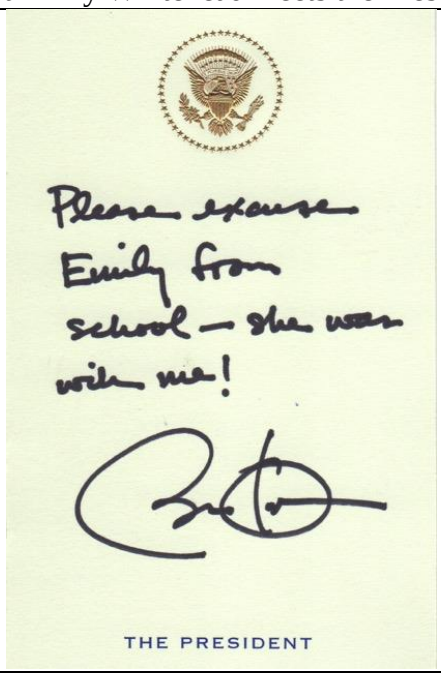

Figure 2 referrals for CTL019 therapy for our first clinical trial

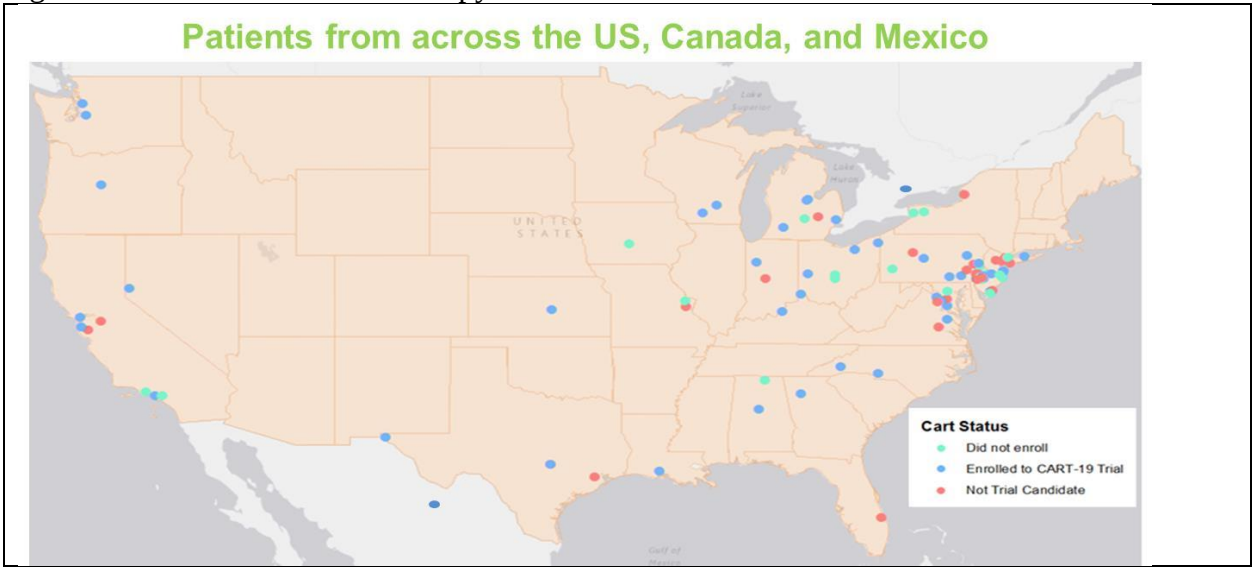

Table 1: Papers published in this period with CURE grant support:

1. Maude, SL N Frey, PA Shaw, R Aplenc, DM Barrett, NJ Bunin, A Chew, VE Gonzalez, Z Zheng, SF Lacey, BL Levine, YD Mahnke, JJ Melenhorst, SR Rheingold, A Shen, DT Teachey, CH June, DL Porter, and **SA Grupp**. 2014. Sustained Remissions with Chimeric Antigen Receptor T Cells For Leukemia. New England Journal of Medicine, 371:1507-17. PMID: 25317870. NIHMSID: 640298.
2. Singh, NA, X Liu, J Hulitt, S Jiang, CH June, **SA Grupp**, DM Barrett, and Y Zhao. 2014. Nature of tumor control by permanently and transiently-modified GD2 chimeric antigen receptor T cells in xenograft models of neuroblastoma. Cancer Immunology Research. DOI: 10.1158/2326-6066.CIR-14-0051.
3. June, CH, MV Maus, G Plesa, LA Johnson, Y Zhao, BL Levine, **SA Grupp**, and DL Porter. 2014. Engineered T cells for cancer therapy. Cancer Immunol Immunother., DOI: 10.1007/s00262-014-1568-1. Sep673(9):969-75. Epub 2014 June 19. PMID: 24943274
4. Lankester, AC, F Locatelli, P Bader, E Rettinger, M Egeler, S Katewa, MA Pulsipher, S Nierkens, K Schultz, R Handgretinger, **SA Grupp**, JJ Boelens, and CM Bollard. 2014. Will post transplant cell therapies for pediatric patients become standard of care? Biol Blood Marrow Transplant. DOI: 10.1016/j.bbmt.2014.07.018; S1083-8791(14)0044-3. PMID: 25064748.
5. Lee, DW, R Gardner, DL Porter, CU Louis, N Ahmed, M Jensen, **SA Grupp**, and CL Mackall, C. L. 2014, Current concepts in the diagnosis and management of cytokine release syndrome. Blood 124(2): 188-195. PMID: 24876563.
6. **Grupp, SA**. 2014. Advances in T-cell therapy for ALL. Best Practice & Research Clinical Haematology, doi:10.1016/j.beha.2014.10.014

7. Maude, SL, DT Teachey, DL Porter, and **SA Grupp.** 2015. CD19-targeted Chimeric Antigen Receptor T cell Therapy for Acute Lymphoblastic Leukemia. Blood, in press.
8. Pulsipher, MA, B Langholz, DA Wall, KR Schultz, N Bunin, W Carroll, E Raetz, S Gardner, RK Goyal, J Gastier-Foster, M Borowitz, D Teachey, and **SA Grupp**. 2015. Risk factors and timing of relapse after allogeneic transplantation in pediatric ALL: for whom and when should interventions be tested? Bone Marrow Transplantation. In press. doi:10.1038/bmt.2015.103
9. Pulsipher, MA, C Carlson, B Langholz, DA Wall, KR Schultz, N Bunin, I Kirsch, JM Gastier-Foster, M Borowitz, C Desmarais, D Williamson, M Kalos, and **SA Grupp.** 2015. IgH-V(D)J NGS-MRD measurement pre- and early post-allotransplant defines very low- and very high-risk ALL patients. Blood, in press. DOI 10.1182/blood-2014-12-615757.
10. Haiying, Q, M Cho, W Haso, L Zhang, S Tasian, H Zarni Oo, G Neri, Y Lin, J Zhou, B Mallon, S Maude, D Teachey, D Barrrett, R Orentas, M Daugaard, P Sorensen, **SA Grupp**, and T Fry. 2015. Eradication of pre B cell ALL using chimeric antigen receptor-expressing T cells targeting the TSLPR oncoprotein. Blood. In press.
11. Porter, DL, WT Hwang, NV Frey, SF Lacey, PA Shaw, AW Loren, A Bagg, KT Marcucci, A Shen, V Gonzalez, D Ambrose, **SA Grupp**, A Chew, Z Zheng, MC Milone, BL Levine, JJ Melenhorst, CH June. 2015. Chimeric Antigen Receptor T Cells Persist and Induce Sustained Remissions in Relapsed Refractory Chronic Lymphocytic Leukemia. Science Translational Medicine. In press.

Table 2: Presentations

Grupp, SA, N Frey, R Aplenc, B Levine, S Maude, S Rheingold, C. Strait Barker, D Teachey, Y Mahnke, D Porter, C June. T cells engineered with a CAR targeting CD19 (CTL019 cells) produce significant in vivo proliferation, complete responses and long-term persistence without GVHD in children and adults with relapsed, refractory ALL. 40th European Bone Marrow Transplant Meeting, April 2014, Milan, Italy, Abstract #1. * Winner, van Bekkum Prize 1st Plenary Abstract Presentation
Grupp, SA, SL Maude, P Shaw, R Aplenc, DM Barrett, C Callahan, A Chew, SF Lacey, BL Levine JJ Melenhorst, L Motley, SR Rheingold, A Shen, DT Teachey, PA Wood, DL Porter and CH June. T cells engineered with a chimeric antigen receptor targeting CD19 (CTL019 cells) produce significant in vivo proliferation, complete responses and long-term persistence without GVHD in children and adults with relapsed, refractory ALL. 56th Annual American Society of Hematology, December 6-9, 2014, San Francisco, CA. Blood, Abstract #380. *****Platform presentation by Dr. Grupp *Featured in ASH Press Program and featured in Best of ASH