Background Chimeric antigen receptor (CAR) T cells have shown remarkable results in hematological malignancies but limited efficacy in the setting of solid tumors. GD2 is a tumor antigen expressed on neuroblastoma and osteosarcoma, and previous studies of T cells expressing 1st generation GD2-CAR were shown to be safe and mediated modest antitumor activity in patients with refractory neuroblastoma. Myeloid cells orchestrate immune responses with the ability to either activate or limit T cell responses. In the setting of solid tumors, myeloid-mediated immune suppression is known to play an important role in dampening antitumor activity. We hypothesized that myeloid cells impact CAR T cell expansion in patients with solid tumors.

Methods A phase I trial (NCT02107963) was performed to determine the feasibility and safety of administering 3rd generation GD2-CAR (GD2-CAR.OX40.28.z.ICD9) T cells in children and young adults with GD2+ neuroblastoma and osteosarcoma. Peripheral blood patient samples were analyzed retrospectively by qPCR for CAR expansion, multiplex ELISA for cytokine levels, mass cytometry (CyTOF) for phenotype analysis, ATAC-seq for epigenetic determination, and RNA-seq for transcriptomic evaluation.

Results 15 patients were enrolled on four dose levels, of which 13 patients were infused. While 76.9% of patients had stable disease by day 28, eventually all patients had disease progression. GD2-CAR T cells expanded in all patients receiving treatment, half of whom had expansion similar to that seen in clinically active CD19 and CD22 CAR T cells, but with limited persistence. To gain insight into the immune compartment in patients with good versus poor CAR T cell expansion, we evaluated immune profiles in patient pre-treatment apheresis and post-treatment peripheral blood samples. The main findings of this study indicate that a higher proportion of monocytes in pre-treatment apheresis was associated with poor CAR T cell expansion and CXCR3 expression on monocytes in pre-treatment apheresis was the most robust marker of good CAR T cell expansion in this cohort. Longitudinal analysis demonstrated that CXCR3+ monocytes were low following treatment in both good and poor CAR T cell expanders, demonstrating a transition in myeloid populations in response to GD2-CAR T cell treatment.

Conclusions Together, these data suggest that GD2-CAR T cell administration is associated with changes in the myeloid cell compartment in solid tumor patients. This study provides evidence of novel myeloid-based pre-treatment biomarkers of CAR T cell expansion and rationale for the combination of CAR T cells with myeloid-modulating therapies as a strategy to improve outcomes for patients with solid tumors.

Acknowledgements We are grateful to the study participants and their families, referring medical care teams, the faculty and staff of the NCI CCR Pediatric Oncology Branch, NCI CCR Center for Cellular Engineering, and the data managers involved in this work. Clinical trial supported in part by: Intramural Research Program, National Cancer Institute, NIH Clinical Center, National Institutes of Health. Scientific and financial support for the CIMAC-CIDC Network are provided through the National Cancer Institute (NCI) Cooperative Agreements: U24CA224331 (to the Dana-Farber Cancer Institute CIMAC), U24CA224309 (to the Stanford University CIMAC), and U24CA224316 (to the CIDC at Dana-Farber Cancer Institute). Scientific and financial support for the Partnership for Accelerating Cancer Therapies (PACT) public-private partnership (PPP).

Trial Registration NCT02107963

Ethics Approval The phase I study protocol conformed to the Declaration of Helsinki, Good Clinical Practice guidelines, and was approved by the NCI Institutional Review Board (14-C-0059) and the FDA. All patients or their legal guardians signed a document of informed consent indicating their understanding of the investigational nature and risks of this study.