Relapse prophylaxis post-haploidentical bone marrow transplantation and cyclophosphamide (Haplo/Cy) by infusion of donor-derived expanded/activated γδ T cells: A Phase I trial*

J. McGuirk, DO[1] , S. Abhyankar, MD[1], T. Goswami, MD[2], R. Soder, PhD[1], M. ter Haak[2], T. Bruns[1], S. Youngblood[2], L. Lamb[2],PhD

[1]The University of Kansas Cancer Center, Westwood, KS, United States, [2]IN8Bio, Inc., New York, NY, United States

Publication No: 3323

Introduction

Relapse post Haplo/Cyclophosphamide (Cy) bone marrow transplantation (BMT) occurs in up to ~51% of patients at one year[A]. We sought to mitigate relapse in this context by infusing expanded and activated donor-derived haploidentical gamma-delta (γδ) T cells (EAGD) early in the post-BMT setting to prevent re-emergence of residual leukemia. This single-center Phase I clinical trial represents the first systemic infusion of allogeneic EAGD cells in the post-BMT setting.

Methods

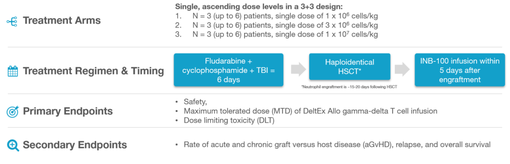

Figure 1: The trial is a standard 3+3 dose-escalation trial to evaluate safety and tolerability of EAGD infusion within 7 days of neutrophil engraftment. Primary endpoints include dose limiting toxicities (DLT) and grade 3-4 adverse events while secondary endpoints include incidence of acute and chronic graft-versus-host disease (GvHD), relapse and overall survival.

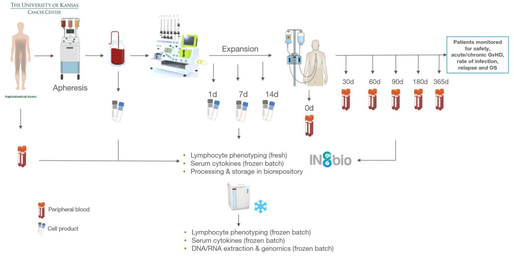

Figure 2: EAGD were manufactured from haploidentical donor apheresis product obtained 7 days prior to BMT harvest and cryopreserved. Patients received standard of care reduced intensity flu/cy/TBI conditioning (RIC) followed by an unmanipulated marrow graft and 50mg/m² Cy on days +3 and +4. Cryopreserved EAGD were thawed and infused intravenously within 7 days of neutrophil engraftment (ANC ≥500/μL X 3d). Peripheral blood for monitoring of immune status was collected at the defined intervals.

Results

Of patients enrolled (Table 1), four patients received the first dose level (DL) of 1 x 10^6 EAGD/kg and remain on study and in remission. Two patients have successfully manufactured DL2 of 3 x 10^6 EAGD/kg, have been transplanted and await neutrophil engraftment for dosing. Of other patients, one received a suboptimal dose and progressed, there was one screen failure, one product was not successfully manufactured, and one patient died prior to infusion. Patient 002 has received intermittent hypomethylating therapy for transient recipient chimerism. γδT cell related toxicities include Grade 1-2 constipation, CMV reactivation, emesis, fatigue, and hypomagnesaemia. Steroid-responsive cutaneous Grade I-II aGvHD was observed in all patients with one also experiencing Grade II intestinal GvHD. No cGvHD, DLTs, treatment-related ≥ grade 3 adverse events, cytokine release syndrome or Immune effector cell-associated neurotoxicity syndrome (ICANS) has occurred.

Table 1: Status of patients currently on study

Patient	Dose Level	Age / Sex	Cytogenetics	Prior lines	Safety Events	Morphologic CR Duration (mos)
002	1	54 / female	High-risk AML trisomy 8+ and del7; FLT3 TKD, DNMT3A,	7+3	Gr.2 skin GvHD-resolved	30.9
003	1	45 / female	High-risk AML trisomy 8+ and del7: IDH2	7+3	Gr.2 GI GvHD and Gr.2 skin GvHD Remains on Jakafi for skin GvHD	28.6
006	1	66 / male	Relapsed AML s/p 7+3, ASXL1	7+3	Gr.2 GvHD-resolved	16.9
007	1	71 / male	Relapsed AML s/p 7+3, ASXL1	Pembrolizumab	Gr.2 skin GvHD-resolved	2.5
009	2	68 / male	Ph- ALL; p53 mutated, DNMT3A, GATA2	Induction E1910, blincyto, inotuzumab x2 cycles, CAR-T with Tecartus		0.5
010	2	62 / female	AML	Hydrea; vidaza/ venetoclax x7 cycles		0.3

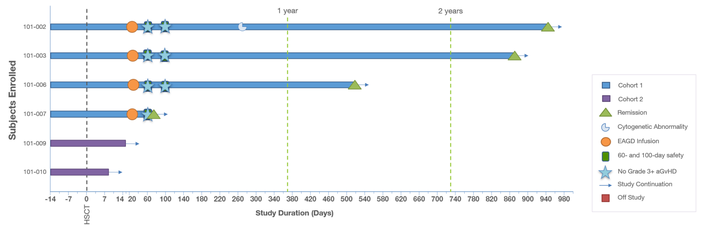

Figure 3: All three DL1 patients with more than six months of follow-up remain in morphologic complete remission (mCR) at 30.9, 28.6, and 16.9 months post-BMT as of November 11, 2022. The fourth DL1 patient remains relapse free at 2.5 months.

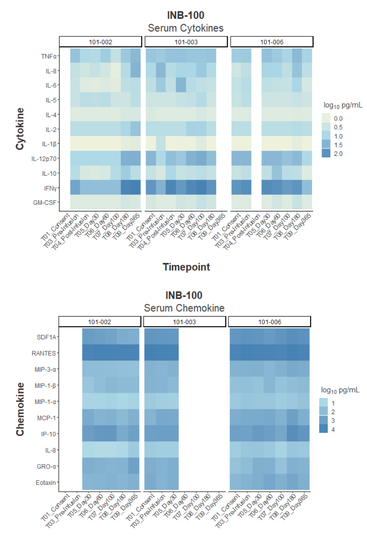

Figure 4: The serum cytokine/chemokine environment reveals an initial inflammatory environment that gradually normalizes. Cytokine and chemokine levels were determined in parallel with lymphocyte phenotyping using Ayoxxa's Human Cytokine 11-plex assay. Ongoing analysis revealed an initial inflammatory environment with predominant expression of IFNγ and TNFα that gradually declines as recovery progressed. In addition, a dip in IL-6, IL-8 and TNF-α at day 100 for subjects 101-002 and 101-006 was observed, with recovery after 180 days at which time cytokine levels increase overall to moderate levels.

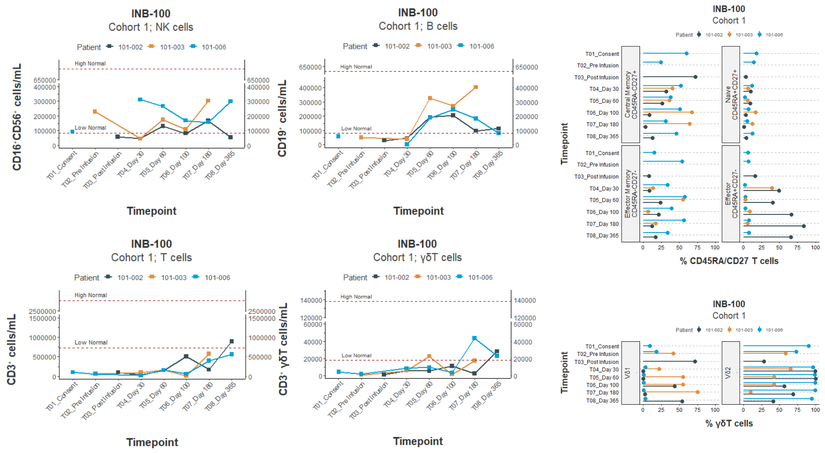

Figure 5: Lymphocyte subset analysis revealed significant T and B cell depletion throughout patient enrollment with B cell recovery initiating approximately two months post treatment, and T cell subsets three-to-six months post treatment with γδ T cells (primarily Vδ2+ subtype) slowly increasing toward normal levels. T cells transitioned from a CD45+CD27- effector phenotype to CD45RA CD27+ central to effector memory phenotype as recovery progressed. NK cells, known to be the first to recover in the haploidentical setting, remain generally within normal ranges throughout treatment.

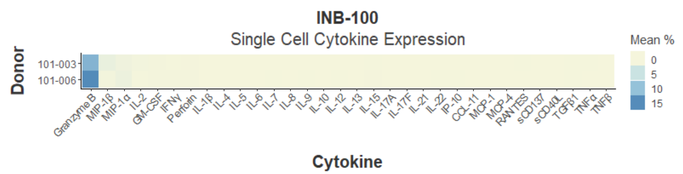

Figure 6: Haploidentical EAGD products were evaluated for single cell cytokine expression. Due to limited graft material single cell cytokine expression could be obtained from only two EAGD grafts. Cellular cytokine expression was determined by IsoPlexis Isocode system using the human adaptive immune panel. Both EAGD grafts demonstrate similar cytokine profiles and principally express Granzyme B, MIP1α, MIP1β and IL-2. As this is a novel product in the transplant setting, specific cell-based cytotoxicity assays remain in development

Conclusions

Early indications suggest that post-Haplo HSCT post-Cy EAGD infusion with the initial dose level of 1 x 10^6 EAGD/kg has manageable toxicity and an appropriate immune recovery profile. Four of four patients treated with a full 1 x 10^6 EAGD/kg remain in mCR, alive and progression-free.

* NCT03533816; Expanded/Activated Gamma Delta T-cell Infusion Following Hematopoietic Stem Cell Transplantation and Post-transplant Cyclophosphamide, https://clinicaltrials.gov/ct2/show/NCT03533816

A. Luznik et al., Biology of Blood and Marrow Transplantation, Vol. 14 (6), June 2008 - https://doi.org/10.1016/j.bbmt.2008.03.005