Background

ADI-001 is a first-in-class CD20-targeted allogeneic γδ T cell therapy. ADI-001 consists of allogeneic healthy donor peripheral blood mononuclear cell (PBMC) derived γδ T cell product that has been cryopreserved, activated, and genetically engineered to express the CD20 CAR γδ T-cell that can target CD20-antigen for killing. Preclinical activity of the CD20 CAR γδ T-cell fusion protein, and expression of a chimeric antigen receptor (CAR) that is specific for CD20, has been shown to be efficacious in vitro and in animal models.

Methods

AlloCell had been analytically validated for 2-genome-based (1 donor) and 3-genome-based (2 donors) mixed cell populations (Cordis, Assay report). To additionally qualify AlloCell for ADI-001 manufacture, genomic DNA (gDNA) mixtures were prepared using 1 gDNA from the cells of two healthy individuals, or 2 gDNAs from the same PBMC sample (to establish feasibility for application in solid organs). The mixture contained cell product levels of 1%, 3%, 5%, and 10%, and 0.1%, 0.3%, 0.6%, and 0.9% as a proportion of total input.

Results

The levels of cell product measured for qualification closely matched the expected levels of donor-derived γδ T cell in gDNA mixtures, with a high degree of linearity (r2 = 0.99, which was reproduced in PPFE samples (r2 = 0.998)). We also observed that the same concentrations in triplicate, with 25% ranging from 0.31% to 1% mixture, 0.35% for the 0.1% mixture, or for PPFE samples, from 0.05% to 1% mixture to 23.6% at the lowest mixture of 0.03%.

Data from ADI-001 patient samples demonstrated AlloCell's ability to track product cell kinetics in a clinical setting and determined dose-dependent exposure across four dose levels evaluated. Finally, in one patient receiving two separate and sequential doses of ADI-001, each manufactured from a different donor, AlloCell successfully distinguished and independently quantified expansions of the first and second donor-derived cell products.

Qualification of Accurate and Precise Quantification in Cells

Figure 4. AlloCell γδ T cell qPCR assay for ADI-001 donor-derived γδ T cells (A) measured across the [1% to 0.1%] range. (B) Precision measured by coefficient of variation over [0.01% to 1%] range. (C) Evaluation of precision and sensitivity measurements using cells.

Figure 5. Detection of distinct expansions from two ADI-001 lots derived from distinct donors in a single patient using 3-genome analysis.

Detection of Distinct Expansions From Two ADI-001 Lots Derived From Distinct Donors in a Single Patient Using 3-Genome Analysis

Figure 6. A: Allelic distribution of the CD20 CAR γδ T cell population across two ADI-001 lots derived from different donors. B: The allelic distribution of the CD20 CAR γδ T cell population across two ADI-001 lots derived from different donors, measured by AlloCell qPCR analysis.

Summary & Conclusions

• Here we present qualification and clinical application of a quantitative and universal next generation sequencing solution (AlloCell) for monitoring of ADI-001, a first-in-class CD20-targeted allogeneic γδ T CAR T cell therapy.

• We show robust quantitation of dose-dependent expansion of ADI-001 in the peripheral blood using this method.

• We demonstrate assay performance assessments in both PBMC and tissue biopsy (FFPE) samples.

• We present successful distinction and quantification of two separate and distinct doses of ADI-001 infused in a single patient using 3-genome analysis.

References


Figure 5. Detection of distinct expansions from two ADI-001 lots derived from distinct donors in a single patient using 3-genome analysis.

Figure 6. A: Allelic distribution of the CD20 CAR γδ T cell population across two ADI-001 lots derived from different donors. B: The allelic distribution of the CD20 CAR γδ T cell population across two ADI-001 lots derived from different donors, measured by AlloCell qPCR analysis.