



## Leukemia-associated antigen reactive T-cells in ATIR101, a recipient-specific allodepleted T-cell product facilitating haploidentical HSCT

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**Introduction:** Graft-versus-leukemia (GvL) relies on donor T-cells killing host leukemia cells post-transplant. T-cells with preferential recognition of leukemia cells have been well documented but their clinical relevance remains challenged because thymic selection prevents a high-affinity interaction between T-cells and antigen presenting cells (APC) expressing regular antigens in self-HLA. ATIR101, a personalized T-cell immunotherapeutic selectively depleted of HLA-haplotype mismatched T-cells, provides a unique platform to study leukemia-reactive T-cells as high-affinity interactions with antigens expressed in the mismatched haplotype may occur, whereas T-cells responding to the antigen-presenting foreign HLA-molecule have been eliminated.

**Material (or patients) and methods:** Two out of the first 10 ATIR101 batches manufactured in clinical phase 2 study CRAIR- 007 (NCT01794299) met the requirement of having a mismatched haplotype known to be able to express a known leukemia-associated antigen; these batches were used to screen for the presence of leukemia-associated antigen reactive T-cells. Because the frequency of leukemia-associated antigen reactive T-cells is expected to be very low, we used peptide-MHC monomers of the mismatched HLA-haplotype presenting leukemia-associated antigens and established a stimulation platform with artificial APCs (aAPC). Those aAPC consisted of streptavidin coated microspheres loaded with a biotinylated anti-CD28 antibody and the respective biotinylated peptide HLA monomer.

**Results:** In one of the two batches, leukemia-associated antigens specific T-cells were detected: ATIR101 cells were stimulated with Myb628 /HLA-B44 aAPCs and Myb628 specific T-cell expansion was assessed after one or two rounds of stimulation; an irrelevant HLA-B44 multimer was used as negative control. Clearly, we were able to detect CD8+ T-cells with specific reactivity against one HLA-B44- restricted leukemia-associated HLA ligand derived from the MYB gene (figure 1).

**Conclusion:** These data show that T-cells recognizing leukemia-associated antigens expressed in the mismatched HLA haplotype are retained in ATIR101 from which the T-cells





responding to the antigen-presenting foreign HLA-molecule have been eliminated. Conceivably, these cells may contribute to the Graft-versus-leukemia (GvL) effect of ATIR101.

**Figure 1:**

