HMMR-redirected adoptive T cell therapy for treatment of refractory acute myeloid leukemia in context of mismatched stem cell transplantation

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Background: The hyaluronic acid-mediated motility receptor (RHAMM/HMMR) is an interesting antigen for adoptive T cell transfer. This is due to two observations: firstly, high expression of RHAMM/HMMR mRNA in AML patients is correlated with poor prognosis and secondly, the existence of immune response against HMMR is linked to better clinical outcome. We previously used dendritic cell priming to obtain allo-restricted HMMR-specific T cells restricted to HLA-A2. The redirected T cells recognized HLA-A2-positive HMMR-positive tumor cells well *in vitro*. *In vivo*, leukemia outgrowth was strongly retarded in humanized xenograft mice. We now present the safety profile of HMMR-redirected T cells. **Methods:** We assessed the impact of HMMR-redirected T cells on normal human stem cells (HSC) *in vitro* using a colony forming unit assay (CFU) and *in vivo* using HLA-A2-transgenic mice (HHDII?). Cross-reactivity of HMMR-redirected T cells for other peptides and HLA allotypes (30 LCL cell lines with common HLA allotypes) were tested using mouse B3Z cells and HMMR-specific TCR-transgenic T cells (generated by CD3/CD28 activation of human peripheral blood T cells and transduction with an MP71 retroviral vector carrying codon-optimized murine HMMR-specific TCR). **Results:** Co-cultures of human CD34+ HSC from HLA-A2+ donors with HMMR-redirected T cells strongly reduced colony forming units when compared to co-cultures derived from HLA-A2-

donors. When HSC-containing bone marrow cells of transgenic mice were pre-cultured with HMMR-redirected T cells before transplantation, they failed to reconstitute hematopoiesis in irradiated mice; the animals died at the same time as the non-transplanted controls. No cross-recognition was seen with more than 100 common HLA-A2-restricted self-peptides. Reactivity of B3Z mouse cells and of HMMR-redirected T cells was tested against a panel of allogenic LCLs and showed no evidence of allo-cross reactivity. Recognition was specifically confirmed for LCL lines expressing the HLA*02:01, HLA*02:07 and HLA*A2:09 subtypes; all three express the same peptide binding groove. It is mandatory to consider this information when designing exclusion criteria of donors, as patients with all three HLA-A2 subtypes could be included in the clinical trial. **Conclusions:** Adoptive T cell transfer of HMMR-redirected T cells for HLA-A2 mismatched stem cell transplantation promises two clinical benefits: Firstly, targeted residual autologous HLA-A2 HSCs lead to faster donor chimerism and secondly the elimination of residual HLA-A2+ leukemic cells may allow curative treatment.