OVEREXPRESSION OF THE CHEMOKINE RECEPTOR CCR7 CONFRS ANTI-CXCR5 CAR T CELLS WITH INCREASED HOMING TO LYMPH NODES AND ENHANCED CYTOTOXICITY TOWARD B CELL LYMPHOMA

Maria Zschummel, Jara Joedicke, Anca Margineanu, Susanne Blachut, Eric Lindberg, Norbert Hüblner, Armin Rehm, Uta Höpken*. Max-Delbrück-Center for Molecular Medicine (MDC), Berlin, Germany

Background The efficacy of CAR T cell therapy correlates with the ability of the transferred therapeutic T cells to traffic to the respective tumor niche. We observed that engineered T cells downregulate the lymphoid homing receptor CCR7 during the manufacturing process. Consequently, in-vitro and in-vivo migration of CAR T cells toward the corresponding chemokine ligands was severely impaired. To improve nodal CAR T cell trafficking and anti-lymphoma efficacy of B-NHL-targeting anti-CXCR5 CAR T cells, we engineered CAR T cells which co-expressed the homing receptor CCR7.

Methods The anti-human CXCR5 CAR and anti-mouse CXCR5 CAR, encoded within a retroviral MP71 backbone, were generated essentially as described. In case of the human and mouse CXCR5 CAR-CCR7 vectors, hCCR7 or mCCR7 cDNAs without stop codons were introduced upstream of the CXCR5 CAR cassette via P2A linkage. To test the functionality of CCR7-engineered CAR T cells in-vitro, transwell migration, cytotoxicity, and cytokine release assays were employed. CAR T cell killing dynamics were determined by microscopy-based methods. Mouse leukemia and lymphoma models served as tools to study in-vivo CCR7-modulated CAR T cell homing to lymph nodes and target cell killing.

Results CCR7 overexpression on human and mouse anti-CXCR5 CAR T cells restored their migratory capacity and improved lymph node homing. In addition, we observed an enhanced killing capacity of CCR7-equipped CAR T cells in-vitro, which is likely supported by the costimulatory role of CCR7 in the formation of an immunological synapse. We showed that CCR7 accumulated in mature CAR synapses, an observation that is supported by the transcriptional upregulation of genes associated with cytoskeletal rearrangement, relevant for immunological synapse formation and migration in CCR7-engineered CAR T cells.

Conclusions Therapeutically, CCR7-enforced nodal recruitment and enhanced killing kinetics of CAR T cells conferred improved lymphoma eradication in a mouse lymphoma model. Beyond improved anti-tumor responses in nodal hematological malignancies, we envisage that this CCR7-engineering approach might be useful to improve CAR T cell therapies that specifically target solid tumor entities metastasized to lymph nodes.

Acknowledgements This work was funded by grants from the HGF “Zukunftsthema Inflammation & Immunology”.

REFERENCE