P09 Young researchers session

P09.01 SINGLE-CELL TRANSCRIPTOMIC ATLAS-GUIDED DEVELOPMENT OF CHIMERIC ANTIGEN-RECEPTOR (CAR) T CELLS FOR THE TREATMENT OF ACUTE MYELOID LEUKEMIA

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Background While chimeric-antigen receptor (CAR) T cells have revolutionized the treatment of refractory B cell malignancies, they have yet to achieve success in the treatment of acute myeloid leukemia (AML).[1] In AML, development of CAR therapy is hindered by expression of AML-associated antigens also on pivotal healthy tissues (e.g. hematopoietic stem and progenitor cells, HSPC). The revolution in single-cell technologies has generated massive expression data, providing precise information on the transcriptomic anatomy of healthy and malignant cells.[2] However, these resources have rarely been used for *de novo* antigen predictions. We hypothesized that we could use these technologies to establish high resolution antigen projections, enabling the identification of novel target structures. Hence, we leveraged an atlas of RNA sequencing data of over 500,000 single cells from AML patients and healthy human tissues for target identification and subsequent testing of novel target structures for CAR T cell therapy.

Materials and Methods 12 single cell data sets were harmonized and used for target prediction. Anti-murine and anti-human CAR constructs targeting the lead candidate - colony-stimulating factor 1 receptor (CSF1R) - were generated and transduced into primary murine and human T cells. AML cell lines and primary AML samples were used to verify expression of CSF1R and as target cell lines *in vitro* and *in vivo*. Off-target toxicities of CAR were analyzed *in vitro* and *in vivo* using a variety of different models.

Results Using a newly developed single-cell RNA sequencing-based screening algorithm, CSF1R was identified as a promising target antigen for CAR T cell therapy in AML. Expression of CSF1R was verified on a large panel of AML cell lines and in primary AML samples. Newly developed anti-CSF1R-CAR T cells efficiently lysed AML target cells *in vitro*. *In vivo*, anti-CSF1R-CAR T cells induced strong and sustained remissions in cell line- and patient-derived xenograft models. Compared to anti-CD33-CAR T cells, anti-CSF1R-CAR T cells did not lyse healthy HSPC and proved to be safe when used in fully syngeneic mice models.

Conclusions Aided by our screening algorithm, we identified CSF1R as a new promising target for CAR therapy in AML and proved the efficacy of newly developed CAR T cells. Our results highlight the remarkable translational potential of unbiased, high-resolution target screenings for cancer entities and warrant further clinical investigations of newly developed anti-CSF1R-CAR T cells.

REFERENCES

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