inhibiting SMAD3, PTEN, ATM, IL18 and BCL2. Consequentially, the observed network-wide changes on LDHC silencing are predicted to negatively influence cellular growth and proliferation, cell migration and cell infiltration. The LDHC-associated network indicated a higher-level regulation by miR378a-3p (p=1.4e-7, z=-3.117), affecting the downstream mechanistic in LDHC-expressing cells. Interestingly, the miR378a causal network also indicated inhibition of the immune response in LDHC-positive cells. TIDE analysis indicated that high expression of LDHC in the METABRIC Her2 breast cancer cohort (TIDE score=1.97, p=0.049), and to a lesser extent in triple negative breast cancer (TIDE score=0.466, p=0.642), decreases the beneficial effect between CTLs and overall survival observed in LDHC Low tumors. Concurrently, *LDHC*-silenced cells displayed a pro-inflammatory gene expression and cytokine profile and down-regulated the expression of PD-L1 and Gal-9 immune checkpoints.

Conclusions Our findings provide an indication of potential CTL dysfunction in breast tumors with high LDHC expression and suggests that therapeutic targeting of LDHC may inhibit tumor growth while releasing the anti-tumor immune response in breast cancer.

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P06.05 ENDOGENOUS T-CELL RESPONSES TO TEN MAJOR CANCER TESTIS ANTIGENS ARE FREQUENT IN ESOPHAGO-GASTRIC ADENOCARCINOMA AND ANTIGEN-SPECIFIC T CELLS CAN BE EXPANDED USING CD40-ACTIVATED B CELLS

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Background Tumor-associated antigens (TAAs) and especially cancer testis antigens (CTAs) are classical tumor-specific targets for immunotherapies. As TAAs are shared between patients, strategies aiming to exploit these targets are scalable and potentially applicable across different types of cancer. Loss of target antigens and other mechanisms of immune escape have limited the success of CTA-directed immunotherapy. CAR T cells and other highly effective cellular therapies have renewed the interest in TAAs. Especially combined targeting of multiple antigens appears highly promising as recently shown in lymphoma. In our study, we aimed to characterize CTA-expression patterns and their impact on endogenous T-cell responses, T-cell abundance and antigen-presentation in esophago-gastric adenocarcinoma (EGA).

Materials and Methods 41 treatment-naïve EGA patients were included in our study. RNA of tumor and patient-matched healthy tissue was isolated and used for NanoString based RNA expression analysis of 26 CTAs and 25 genes associated with antigen-presentation. Based on CTA expression, 10 peptide pools were selected and co-cultured with peripheral blood mononuclear cells (PBMCs, n=21) to determine cellular anti-tumor immune responses in a FluoroSpot assay. T-cell abundance was assessed using immunohistochemistry (CD3, CD8) and digital image analyses of tumor area and invasive margin. Autologous CD40 activated B cells were used to expand antigen-specific T cells using peptide pools of CTAs.

Results NanoString analysis revealed pronounced differences regarding CTA expression, with CEP55 and MAGEA3/6 showing strong expression, while NY-ESO-1 or MAGEA1 were only weakly expressed. 68.3% (28/41) of the patients showed expression of $\geq$ 5/10 analyzed TAAs simultaneously. In line with the frequent expression, 75.0% of the patients showed a cellular response against at least one of the TAAs. T-cell responses were most frequently detected to Survivin and NY-ESO-1 (65.0% and 52.6% of patients, respectively), while only 20.0% responded to CEP55 or TTK peptide pools. Overall, 6/20 patients showed cellular responses against $\geq$5 TAAs simultaneously. We found a strong correlation of T-cell abundance and antigen-presentation. In addition, patients with a high Immune-Score showed increased TAA expression. Finally, we demonstrate feasibility of TAA-specific T-cell expansion using CD40 activated B cells as potential strategy to induce or enhance TAA immune responses in EGA.

Conclusions Our study highlights the importance of TAAs in EGA. The identified antigens are highly relevant for immunomonitoring of clinical trials and as targets for immunotherapy. Personalized immunotherapeutic strategies targeting EGA-specific or even patient specific TAAs appear highly promising in this challenging disease.

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P06.06 ENHANCING TRAFFICKING AND RESISTANCE TO IMMUNOSUPPRESSION OF SYNTHETIC AGONISTIC RECEPTOR-TRANSDUCED T CELLS IN SOLID TUMOR MODELS

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Background Chimeric antigen receptor therapy – although very efficacious in B cell malignancies – is facing many challenges which limit its success in solid tumors, e.g. on-target off-tumor toxicities, antigen heterogeneity, lack of T cell migration into tumors and an immunosuppressive tumor microenvironment. To better control on-target off-tumor effects and address antigen heterogeneity we developed a modular approach where we equipped T cells with a synthetic agonistic receptor (SAR). The SAR is only activated in the

presence of a bispecific antibody (BiAb) cross-linking the receptor with a tumor-associated antigen. While we could show efficacy of the SAR platform in different models, limited infiltration and immune suppression still hamper its function. We could previously demonstrate that T cell infiltration can be enhanced by transduction with carefully chosen chemokine receptors like CXCR6, CCR4 and CCR8. At the same time, gene silencing of checkpoint molecules like PD-1 can make T cells more resistant to immunosuppression, thus we assumed that combining these approaches might generate a desired T cell product.

Materials and Methods All constructs had been generated previously by overlap-extension cloning. The EGFRvIII (E3) SAR consists of extracellular EGFRvIII, transmembrane CD28 and intracellular CD28 and CD3ζ. Human CXCR6-GFP, CCR4-GFP and CCR8-GFP are composed of the chemokine receptors fused to GFP via a 2A sequence. Primary human T cells were retrovirally transduced to stably express the SAR and chemokine receptors. We analyzed migration, cytotoxicity and activation of the single and double (E3 SAR and chemokine receptor) transduced T cells. In addition, PD-1 was knocked out using CRISPR-Cas9 and killing kinetics of target cells and T cell activation were assessed.

Results Co-transduction with chemokine receptors significantly increased migration of E3 SAR T cells to their respective ligand while lysis of target-expressing tumor cell and T cell activation in the presence of BiAb were not affected *in vitro*. Additionally knocking out PD-1 enhanced killing kinetics and activation of E3 SAR and E3 SAR + CXCR6-GFP transduced T cells compared to corresponding mock electroporated T cells.

Conclusions Using the controllable and modular SAR – BiAb platform SAR T cell activation can be limited by stopping BiAb dosing if adverse events occur. In addition, SAR T cells can be redirected to an alternative tumor-associated antigen by exchanging the BiAb in the case of antigen escape. Here we present add-ons to this approach for increased tumor infiltration and resistance to immunosuppression. Since migration is enhanced upon co-transduction with chemokine receptors and target cell lysis is accelerated upon PD-1 knockout in vitro these two additional modifications seem very promising options to further improve tumor control in vivo.

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P06.07 *IN VIVO* STUDIES OF IMMUNOMODULATORY A-CTLA-4 ANTIBODY IN A HUMANIZED MOUSE MODEL

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Background Recent findings in cancer immunotherapy have reinforced the hypothesis that the immune system is able to control most cancers. Immunomodulatory antibodies can enhance immune responses, having the potential to generate anti-cancer immunity.[1–4]

Materials and Methods Most current studies addressing this question are performed in murine mouse model systems or use *in vitro* culture systems, which do not reflect the human in vivo situation, potentially leading to results that cannot be fully translated into human cancer therapy. Therefore, it is necessary to establish a new mouse model, which allows the study of cancer immunotherapy in the context of a human immune system. We focused on the establishment of a humanized mouse model, in which different immunomodulatory antibodies can be tested in the presence of a human immune system.

Results First experiments concerning the suitability to test immunomodulatory antibodies in the humanized mouse model, revealed that effects of checkpoint-control antibody a-CTLA-4 were similar to the effects seen in patients of clinical studies. To analyse the anti-tumor activities of immunomodulatory antibodies in vivo we are establishing a human melanoma-like tumor model in humanized mice.

Conclusions This enables us to test the efficacy of immunomodulatory agonistic antibodies (such as CP-870,893) and checkpoint control antibodies (such as anti-CTLA-4) in eliminating a melanoma-like tumor. Furthermore, parameters like tumor infiltrating human cells und cytokine/chemokine production can be analysed.

REFERENCES

1. Schuster M, Nechansky A, Loibner H. Cancer immunotherapy. *Biotechnol J* 2006;**1**:138–147.
2. Mellman I, Coukos G, Dranoff G. Cancer immunotherapy comes of age. *Nature rev* 2011;**480**:480–489.
3. Finn OJ. Immuno-oncology: understanding the function and dysfunction of the immune system in cancer. *Annals of Oncology* 2012;**23**:vii6–vii9.
4. Langer LF, Clay TM, Morse MA. Update on anti-CTLA-4 in clinical trials. *Expert Opin Biol Ther* 2007;**8**:1245–1256.

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P07 Cell therapy in haematologic diseases

P07.01 A MODULAR AND CONTROLLABLE T CELL THERAPY PLATFORM FOR AML

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Background Targeted immunotherapies have shown limited success in the context of acute myeloid leukemia (AML). The mutational landscape, heterogeneity attributed to this malignancy and toxicities associated with the targeting of myeloid lineage antigens, it has become apparent that a modular and controllable cell therapy approach with the potential to target multiple antigens is required. We propose a controlled ACT