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UCARTMESO: Mesothelin (MSLN) targeting allogeneic CAR-T cells engineered to overcome tumor immunosuppressive microenvironment

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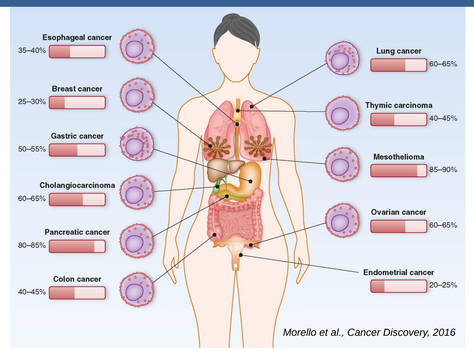
1 INTRODUCTION

- Chimeric antigen receptor (CAR) T cell therapy has provided a new therapeutic option to patients with B cell malignancies, but has thus far not shown consistent therapeutic benefit in solid tumors
- New approaches to address the challenges that have limited the efficacy of CAR-T cell therapy in solid tumors are emerging, as preventing immune suppression in the tumor microenvironment (TME)
- Factors present in the TME can impair CAR-T cell activity, with TGFβ being one of the most important regulators
- **UCARTMESO is an allogeneic CAR-T cell product that targets mesothelin (MSLN) expressing cells, gene engineered to circumvent immune suppressive signals from the TME**
- **UCARTMESO is currently under pre-clinical development by Cellectis**

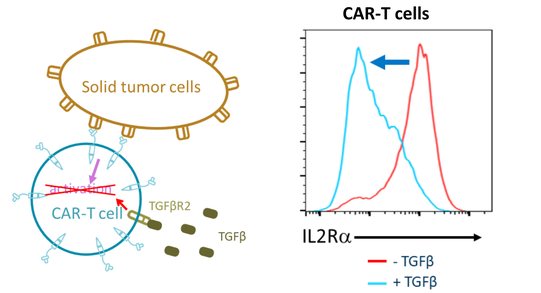
2 MESOTHELIN IS AN ATTRACTIVE SOLID TUMOR TARGET



Mesothelin (MSLN):

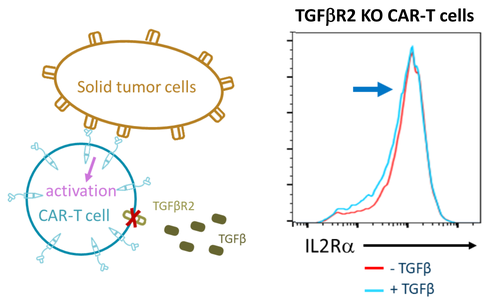
- Cell-surface antigen overexpressed in a a wide range of solid tumors
- One of the most studied targets for solid tumor treatment
- Promising preliminary clinical results have been reported using MSLN-targeted agents

3 TGFβ IMPAIRS CAR-T CELL ACTIVITY IN SOLID TUMORS



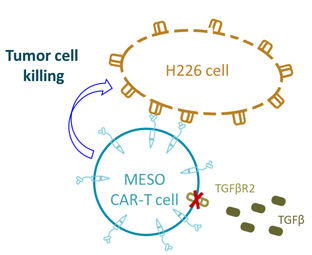
- In a TGFβ deprived TME, recognition of MSLN+ cells by CAR-T cells induces their activation
- In a TGFβ rich tumor microenvironment, binding of TGFβ to its receptor (expressed at the CAR-T cell surface) leads to inhibition of CAR-T cell activation
- **CAR-T cells exposed to tumor antigen in the presence of TGFβ display decreased expression of IL2 receptor (CD25) suggesting that these CAR-T cells could have impaired cell expansion and survival**

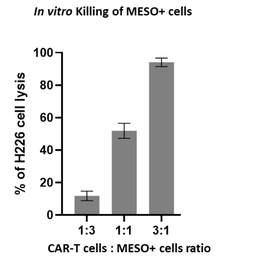
4 TGFBR2 EDITED CAR-T CELLS ARE RESISTANT TO TGFβ



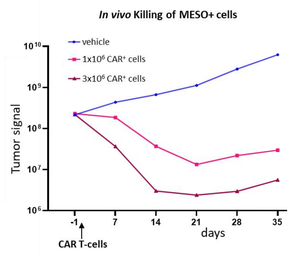
- The inhibitory effect of TGFβ on CAR-T cells can be counteracted by knocking-out its receptor: the TGFBR2 gene
- TGFβR2 edited CAR-T cells can be activated whether in the absence or presence of TGFβ
- **TGFBR2-KO MESOCAR-T cells exposed to tumor antigen in the presence of TGFβ show strong expression of IL2 receptor, suggesting they have normal cell expansion and survival**

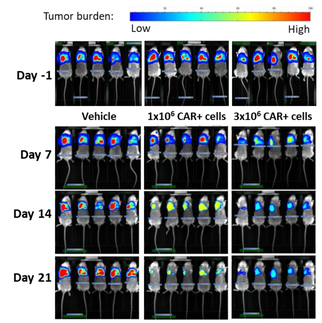
5 TGFβR2-EDITED MESOCAR-T CELLS DISPLAY POTENT ANTI-TUMOR ACTIVITY IN VITRO AND IN VIVO





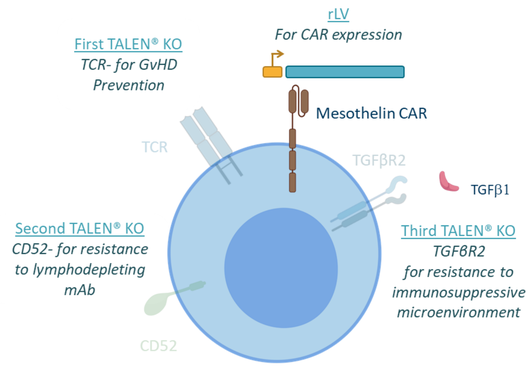
- **TGFβR2-KO MESOCAR-T cells display *in vitro* cytotoxic activity against H226 cells (a mesothelioma cell line expressing MSLN and able to produce TGFβ)**
- **Killing efficacy is proportional to the dose of CAR-T cells**
- **Cells were also able to secrete IFNγ and proliferate in response to antigenic stimulation *in vitro* (not shown)**





- *In vivo* activity of TGFβR2-KO MESOCAR-T cells was evaluated in immunodeficient mice harboring a mesothelioma tumor model
- H226 tumor burden was confirmed by bioluminescence imaging 13 days after intrapleural injection of H226 cells (Day -1 of treatment)
- IV infusion of vehicle or CAR-T cells was administered at Day 0
- **Tumor regression (and increased survival) was observed in all CAR-T treated animals**
- **Tumors were almost undetectable at the dose of 3x10⁶ CAR-T cells at the end of study (Day 70)**

6 UCARTMESO: ALLOGENEIC CAR T-CELLS IN SOLID TUMORS



UCARTMESO product candidate is composed of allogeneic non-alloreactive T cells edited with TALEN®-encoding mRNAs to disrupt TRAC, CD52 and TGFβR2 genes, and transduced ex vivo with a rLV to express a second-generation CAR targeting MSLN

- **TCRα gene KO minimizes the occurrence of GvHD**
- **CD52 gene KO improves persistence of UCARTMESO in the presence of alemtuzumab (an anti-CD52 mAb that can be used as lymphodepleting agent)**
- **TGFβR2 gene KO protects UCARTMESO from the inhibitory effect of TGFβ**

7 CONCLUSIONS & PERSPECTIVES

- Solid tumors have presented a challenge to the success of adoptive cellular therapies, due at least in part to the immunosuppressive tumor microenvironment
- Mesothelin is an attractive target for CAR-T cell therapy in solid tumors as it is overexpressed on a wide variety of tumors types
- UCARTMESO is an allogeneic “off-the-shelf” product candidate that incorporates, TCRα-KO, CD52-KO, and TGFβR2-KO, and expresses a 2nd Generation MSLN-CAR
- TGFβR2-KO MESOCAR-T cells are unable to transduce inhibitory signals through the TGFβ pathway, and therefore capable of responding to antigen stimulation in the presence of TGFβ
- Preclinical studies show potent activity of UCARTMESO *in vitro* and *in vivo* against MSLN expressing cell lines
- *In vivo* activity has been confirmed in pancreatic and pleural mesothelioma mouse models
- **TGFβR2 gene KO provides valuable additional properties to UCARTMESO, representing a very attractive strategy for their use in the treatment of solid tumors**