

## **Solid tumor CAR T cells engineered with fusion proteins targeting PDL1 for localized IL-12 delivery against prostate cancer**

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**Background:** Chimeric antigen receptor (CAR) T cell efficacy in solid tumors, including prostate cancer, is limited due in part to the immunosuppressive solid tumor microenvironment (TME). CAR T cell engineering strategies to address the TME will be necessary to achieve robust and durable anti-tumor responses. We hypothesize that enabling CAR T cells to secrete bifunctional fusion proteins consisting of a cytokine modifier (e.g., TGFβ<sup>trap</sup>, IL15, or IL12) combined with an immune checkpoint inhibitor (e.g., αPDL1) will provide tumor localized immunomodulation to improve CAR T cell functionality.

**Methods:** Multiple solid tumor-targeted CAR T cells were transduced to secrete either a TGFβ<sup>trap</sup>, IL15, or IL12 cytokine fused to an αPDL1 scFv. These CAR + fusion combinations were assessed for effectiveness in repetitive tumor challenge assays *in vitro* as well as in syngeneic prostate and ovarian solid tumor models *in vivo*.

**Results:** CAR T cells modified with PDL1-IL12 fusions were superior in safety and efficacy to CAR T cells alone and to CAR T cells engineered with αPDL1 fused with either TGFβ<sup>trap</sup> or IL15. Further, CAR T cells engineered with PDL1-IL12 resulted in localized IFNγ production, beneficial TME modulation, potent anti-tumor responses, and reduced systemic inflammation and associated toxicities compared with other versions.

**Conclusions:** Solid tumor-targeted CAR T cells show significantly improved anti-tumor responses when engineered to secrete a bi-functional PDL1-IL12 fusion protein, and present a novel strategy to improve therapeutic efficacy. We believe our PDL1-IL12 engineering strategy in CAR T cells presents a unique opportunity to improve clinical efficacy and safety across prostate cancer and multiple solid tumor CAR targets and tumor types.

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