**PD1 TurboCAR™ T cells: PD1-resistant CAR T cells with programmable cytokine signaling outputs**

**Abstract**

CAR T cell therapy has demonstrated unprecedented efficacy in the treatment of hematological malignancies. However, clinical benefit in solid tumor indications has been limited, in part due to suppression of solid tumor microenvironment (TME) that inhibits T cell effectors and persistence. While the provision of cytokine support can help CAR T cells overcome suppressive TME, combining CAR T therapy with systemically-administered cytokines/cytokine mimetics can result in toxicities and locally activate cytokines may enhance rejection of allogeneic product by host cells. For this reason, we have previously designed and tested a novel cytokine-stimulated CAR-T cell designated a TurboCAR. TurboCAR T cells co-express a CAR and a Turbo domain (i.e., a homodimeric cytokine receptor chimera) that transmits CAR-T cell-intrinsic cytokine signals. We reported earlier that in preclinical studies, TurboCAR T cells directed against BCRAM demonstrated enhanced potency, expansion and persistence compared to the TurboCAR platform and tailor it for solid tumors, we employed a two-pronged approach aimed at inhibiting immune-suppressive PD1 signaling while simultaneously transmitting immune-potentiating cytokine signals. As a result, TurboCAR T cells, when hPD-T1 is fused to a PD1 eosinomorph that serves as a dominant-negative receptor. The PD1 eosinomorph was further modified for high-affinity binding to PD1 ligands, allowing for preferential activation of the Turbo domain. TurboCAR T cells express TurboCAR T cells directed towards PD1:exploring solid tumor target cells showed improved functionality compared to CAR T cells combined with PD1 blockade or to the parental TurboCAR T cells alone. In conclusion, PD1 TurboCARs augmented with a PD1 dominant negative eosinomorph conferred CAR T cells with resistance to PD1-mediated inhibition, while simultaneously transmitting cytokine signals in a CAR T cell-like fashion. As an in vivo product, PD1 TurboCAR T cells may obviate the need for combination therapy with anti-PD1 antibodies, while circumventing safety risks associated with systemic cytokine administration.

---

**Figure 1: The homodimeric PD1 Turbo domain activates cytokine signaling in response to PD1 ligands or anti-PD1**

---

**Figure 2: The homodimeric TpoR transmembrane and JAK-activating domain was selected for its ability to induce Turbo domain signaling**

---

**Figure 3: Ectodomain modifications improved Turbo domain signaling**

---

**Figure 4: Signal domains can be tailored to mimic diverse, combinatorial cytokine outputs**

---

**Figure 5: PD1 Turbo domain signals in human CAR T cells**

---

**Figure 6: Efficient PD1 blockade coupled to cytokine signaling underlie the superior activity of HA PD1 TurboCAR™ T cells**

---

**Figure 7: Responsiveness of the HA PD1 Turbo domain was further enhanced by modifications to the transmembrane domain**

---

**Conclusions**

- PD1 Turbo domains are designed to overcome the inherent challenges in solid tumors associated with the immune suppressive TME by blocking suppressive PD1 signaling and turning on stimulatory signals.
- PD1 Turbo domains were optimized for efficient PD1-L1 sequestration and enhanced cytokine signaling.
- PD1 Turbo domains can be tailored for diverse, programmable and combinatorial signaling outputs.

---

**References:**


Acknowledgments: Allogene's AlloBac™ program utilizes Cellera technology. The EGFRvIII AlloBac™ program is licensed exclusively from Cellera by Allogene. Allogene holds exclusive U.S. rights and has granted to Servier rights to EGFRvIII product candidates for all other countries.