T cells co-expressing a highly potent NY-ESO-1-specific TCR and a chimeric PD1-41BB co-stimulatory switch receptor show a favorable polyclonal functional profile for the treatment of solid tumors

Andrea Coluccio, Stefanie Tippmer, Kathrin Mutze, Petra U Prinz, Maja Buerdek, Barbara Loesch, Kathrin Davari, Giulia Longino, Dolores J Schendel
Medigene Immunotherapies GmbH, a subsidiary of Medigene AG, Planegg, Germany

Background
The success of TCR-T cell therapy against solid tumors is dependent on a potent and specific TCR and on the ability of the engineered T cells to overcome the immunosuppressive signals coming from the tumor and its microenvironment.
NY-ESO-1 is a cancer-testis antigen that is highly expressed in various solid tumors, while its expression in healthy tissue is restricted to germ cells. It is a well-described target for cancer vaccines and adoptive cell therapy.

We selected an NY-ESO-1-specific TCR from a non-tolerized T repertoire of a healthy donor using Medigene’s proprietary TIS-\-ICA TCR Priming technology and high-throughput TCR isolation and characterization processes1.

PD1-41BB is a chimeric co-stimulatory switch receptor consisting of the extracellular domains of PD1 and the intracellular domain of the 41BB co-stimulatory receptor.

Results

PD1-41BB enhances activation and proliferation of TCR-T cells

- 75% of TCR-transduced T cells co-express the chimeric PD1-41BB (Fig. 2a).
- 3 fold increase in TCR-T cell co-expressing chimeric PD1-41BB after stimulation with PD-L1-expressing tumor cells (Fig. 3a). Compared to individual T cells, staining was performed using monoclonal antibodies specific for TCR, PD1, and 41BB.

PD1-41BB improves TCR-T cells polyfunctionality by increasing production of effector, stimulatory and co-stimulatory cytokines

- T cells producing multiple cytokines, so-called ‘polyfunctional’ T cells, are known to provide a more effective immune response.
- NY-ESO-1 T cells equipped with co-stimulatory receptor PD1-41BB display a 4.5-fold increase in polyfunctionality (Fig. 3b) and a 6- to 8-fold higher Polyfunctional Strength Index (PSI) (Fig. 3c).

- Polyfunctinality of NY-ESO-1 TCR+PD1-41BB-1 sharing two functional categories (see Fig. 4).

- Secretion of single proteins induced by co-culture of TCR-T cells with or without PD-I and target cell line mediated (PD-L1, PSI). Proteins are depicted and classified by function (e.g., Th1 effector, Th2 cytokines, and regulatory and inflammatory cytokines).

PD1-41BB prolongs killing activity of TCR-T cells upon repeated stimulation

- NY-ESO-1 TCR-T cells co-expressing PD1-41BB show sustained killing activity upon continuous stimulation with NY-ESO-1-200 PD1-L1 tumor cells, which is further prolonged by expression of PD-L1 on tumor cells.
- No TLR-independent killing is observed for NY-ESO-1-negative but PD-L1-positive 3D tumor spheroids.

Figure 1: PD1-41BB: A next generation co-stimulatory switch to enhance T cell functionality and overcome TME

Introduction of Medigene PD1-1 expressed by T cells with PD1-41BB on tumor cells results in induction of T cell activity and can be conducted in isolation and assemble. It must be noted that the process of the chimeric switch receptor PD1-41BB on TCR-transduced effector cells can be efficiently conducted in isolation to achieve effective functions, survival and expansion.

References

Figure 2: Improved effect function of NY-ESO-1 TCR+PD1-41BB TCR transduced T cells with NY-ESO-1 expressing tumor cells (Fig. 3a).

Figure 3: Enhanced NY-ESO-1 TCR-T cell polyfunctionality by co-expression of chimeric co-stimulatory receptor PD1-41BB.

Figure 4: Increased frequencies of secreted effector, stimulatory, and co-stimulatory proteins by TCR-T cells co-expressing specific TCR and PD1-41BB.

Conclusions
The PD1-41BB co-stimulatory switch receptor significantly increases the activity and proliferation of tumor antigen-specific TCR-T cells upon stimulation with tumor cells expressing both the target antigen NY-ESO-1 and PD1-L1.

The co-stimulatory effects of PD1-41BB are dependent on the TCR-mediated recognition of the specific antigen and on the expression of the inhibitory ligand PD-L1 on tumor cells.

Co-expression of the switch receptor leads to a striking increase in polyfunctional T cells, driven by the production of effector, stimulatory, and co-stimulatory cytokines.

The PD1-41BB co-stimulatory switch receptor is an effective molecular tool to enhance TCR-T cell functionality and resist the inhibitory signals from the TME, thus overcoming a major obstacle to the clinical efficacy of adoptive cell therapy against solid tumors.

Figure 3 Heatmap: Increased frequencies of secreted effector, stimulatory, and co-stimulatory proteins by TCR-T cells co-expressing specific TCR and PD1-41BB.