Genetic Modification of lovance’s TIL through TALEN®-mediated knockout of PD-1 as a strategy to empower TIL therapy for cancer

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Introduction

Study Objectives
• To genetically and functionally characterize PD-1 KO TILs, and assess their therapeutic value

Background
• Despite the existence of various tumour-infiltrating lymphocytes (TILs) and their reported efficacy results in metastatic melanoma and cervical cancer patients, reflect on G7 or G9, respectively.
• One potential limitation of TIL therapy is T cell exhaustion, illustrated by the low viability of TILs harvested from breast cancer patients. Development of trypan blue dye exclusion, a semi-quantitative technique, to detect viable cells, is described here.
• Tryptophan and arginine catabolism, pathways that are involved in the degradation of immune cells. Tryptophan is metabolized by tryptophanase (TPH1), which is expressed in breast cancer cells.

Experimental Design
• Three examples of breast, lung, and ovarian cancers were used in this study.
• Several rounds of TIL expansion with PD-1 KO TILs were tested for optimization of the PD-1 KO TIL expansion process. The optimal conditions were determined for the expansion of breast TILs (Table 2).

PD-1 KO TIL Expansion Process
• The PD-1 KO TILs were highly functional compared to control TILs, as demonstrated by the robust level of IFN-γ production and killing capacity.

Results

Figure 1. Addition of a PD-1 TALEN® electroporation step to lovance’s Generation 2 process mediated efficient PD-1 KO in TIL

Figure 2. PD-1 KO did not affect either TIL expansion or viability

Figure 3. PD-1 KO TIL exhibited comparable phenotype relative to control TIL

Figure 4. PD-1 KO TIL were highly functional compared to control TILs

Figure 5. Degree of reactivity of PD-1 KO TIL was slightly elevated relative to unmodified TILs

Conclusion

• An improved protocol for the generation of TALEN®-mediated PD-1 KO TILs, and their expansion to therapeutically relevant numbers was successful. The generation of PD-1 KO TILs in 28 days, followed by Expansion in 4 weeks, and in vitro and in vivo characterization, is described here.

Disclosures
• The authors declare no potential conflicts of interest.

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References