510 ES008, A HIGH AFFINITY LILRB1 SPECIFIC BLOCKING ANTIBODY ACTIVATES MULTIPLE IMMUNE CELLS TO FIGHT CANCERS

Xiaofeng Niu, Haixia Jiang, Chunnian Wang , Dawei Sun , Rui Gao, Quan Qiu, Hongtao Lu*. *Elpiscience Biopharma, Ltd., Shanghai, China*

Background Leukocyte Immunoglobulin-like receptor B1 (LILRB1), the most broadly expressed member of LILRB family, is an immunoreceptor tyrosine inhibitory motif (ITIM)-containing inhibitory receptor expressed on various immune cells including monocytes, macrophages, DCs, B cells and subsets of NK cells and T cells. LILRB1 was shown to bind to β2M-associated classical and non-classical MHC-class I molecules, and non-HLA ligands such as S100A8/S100A9 and UL18. Upon ligand ligation, LILRB1 elicits inhibitory signaling by phosphorylation of the ITIMs present on the cytoplasmic tail, which subsequently recruit and activate SHP-1/SHP-2 phosphatase to suppress downstream tyrosine phosphorylation-dependent signaling pathways, that normally promote effector functions. LILRB1 ligation renders the tolerance of DCs, inhibits B cell responses, prevents tumor cell destruction by macrophages, inhibits the activation and cytotoxicity of LILRB1-expressing NK cells, and downregulates TEMRA $CD8^+$ T cell function. In addition, LILRB1 competes with CD8 for binding to the MHC class 1 molecules, leading to inhibition of antigen-presentation, and downstream T cell responses. Indeed, high LILRB1 expression correlates with reduced survival in both hematopoietic and solid tumor patients. Blocking LILRB1 augments macrophage phagocytosis of tumor cells, restores cytotoxic function of NK cells, and enhances tumor cell killing by effector $CD8^+$ T cells. We have therefore developed a high affinity LILRB1 specific blocking antibody that can activate multiple immune cells to fight cancers.

Methods LILR family homologue binding properties were evaluated by ELISA and FACS. Antigen binding affinity was determined by SPR system (Biacore). Blocking activity was determined by FACS-based competition assays and LILRB1/SHP-1 recruitment assay. *In vitro* function activity was evaluated by macrophage phagocytosis assay and NK killing assay. Epitope analysis was performed by competitive ELISA. Lead clone was humanized via CDR grafting and back mutation screening.

Results ES008-a is a high affinity human LILRB1 specific antibody, which can completely block HLA-G/LILRB1 and HLA-A2/LILRB1 interactions, thereby inhibiting HLA-G and classical MHC class I molecules-induced LILRB1 inhibitory signaling. ES008-a can effectively synergize with CD47/SIRPα inhibitors to enhance macrophage phagocytosis of tumor cells. ES008-a can also potently potentiate NK cells killing of tumor cells.

Conclusions We have developed a functional anti-LILRB1 monoclonal antibody ES008-a that has great potential to activate multiple immune effector cells in the tumor microenvironment, leading to strong anti-tumor immunity.

http://dx.doi.org/10.1136/jitc-2023-SITC2023.0510