

UNLOCKING INTRACELLULAR TARGETS WITH HIGHLY SPECIFIC TCR-LIKE ANTIBODIES



THE NEED

TCR-like antibodies for targeting intracellular proteins

Immunotherapy is a key strategy in cancer treatment but remains limited, as most (80–90%) human proteins are intracellular and inaccessible to conventional antibodies. These intracellular proteins are processed into peptides and presented on the cell surface by HLA molecules as peptide–HLA (pHLA) complexes, which are naturally recognized by T-cell receptors (TCRs). TCR-like antibodies mimic this mechanism, enabling antibody access to intracellular targets. However, the potential off-target cross-reactivity of TCRs and TCR-like antibodies poses a significant safety concern for therapeutic development.

THE SOLUTION

A novel strategy for the discovery of highly specific TCRs or TCR-like antibodies

Specificity is the key to developing safe TCR or TCR-like antibody therapeutics. To achieve this, we have designed a novel CHO-display library that presents the natural human pHLA repertoire on the cell surface (Figure 1). This library enables in-vitro discovery campaigns involving counter-selections and screenings to identify TCRs or TCR-like antibodies with minimal off-target binding at early stages.

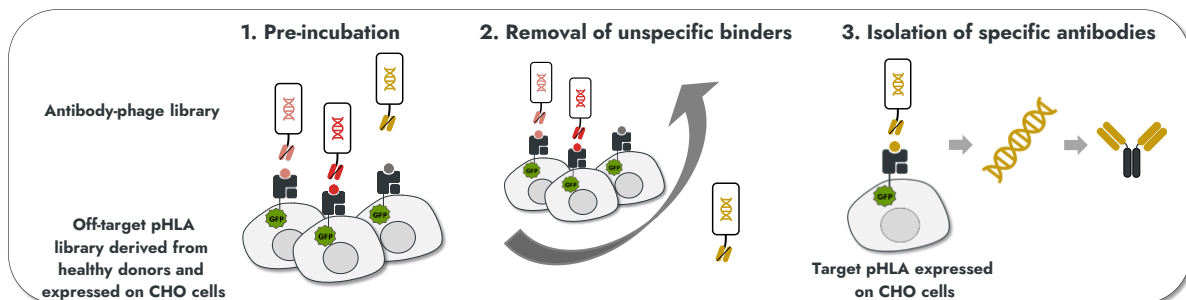


Figure 1. Workflow for discovering specific TCR-like antibodies. The antibody-phage library is incubated with the pHLA-displaying CHO cells to deplete off-target binders. Clones without binding to the healthy human pHLA repertoire are carried forward into subsequent positive selections of the cancer-specific pHLA.

THE IMPACT

Stringent counter-selections give rise to specific TCR-like antibodies

Antibody clones derived from this discovery workflow against the Wilms tumor protein 1 (WT1) pHLA complex are more specific than the reference antibodies (ESK1, 11D06), showing either no or only minimal binding to the human pHLA displaying CHO cells (Figure 2).

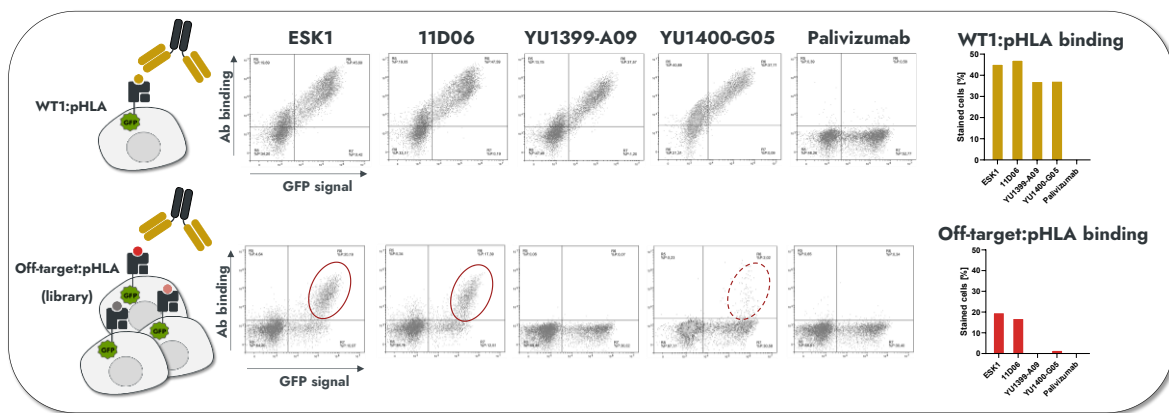


Figure 2. Flow cytometry analysis of antibody binding to the WT1:pHLA target or the pHLA off-target library displayed on CHO cells.

Being more specific, functional activity testing of our antibodies as bispecific T-cell engagers (Figure 3, A) in cell killing assays (B) showed comparable target cell killing activity as the 11D06 reference antibody (C).

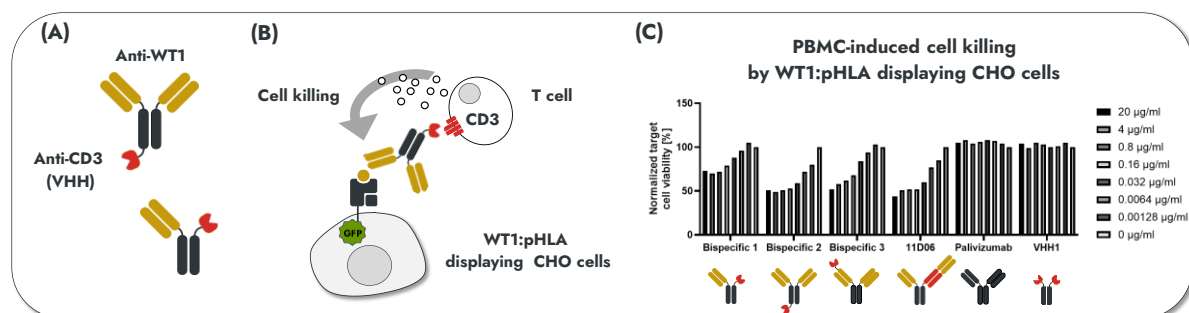


Figure 3. Activity of WT1:HLA specific antibodies in bispecific T-cell engager formats.

To accelerate the development of safe TCR-like antibodies, it is crucial to minimize—or ideally eliminate—off-target binding to pHLA of healthy human tissue. Due to the short length of the peptide sequences, there is the risk of cross-reactivity to sequence related and unrelated peptides. Implementing a counter-selection and screening strategy in discovery campaigns on the healthy human pHLA repertoire provides a solid foundation for developing safe and effective therapeutics.