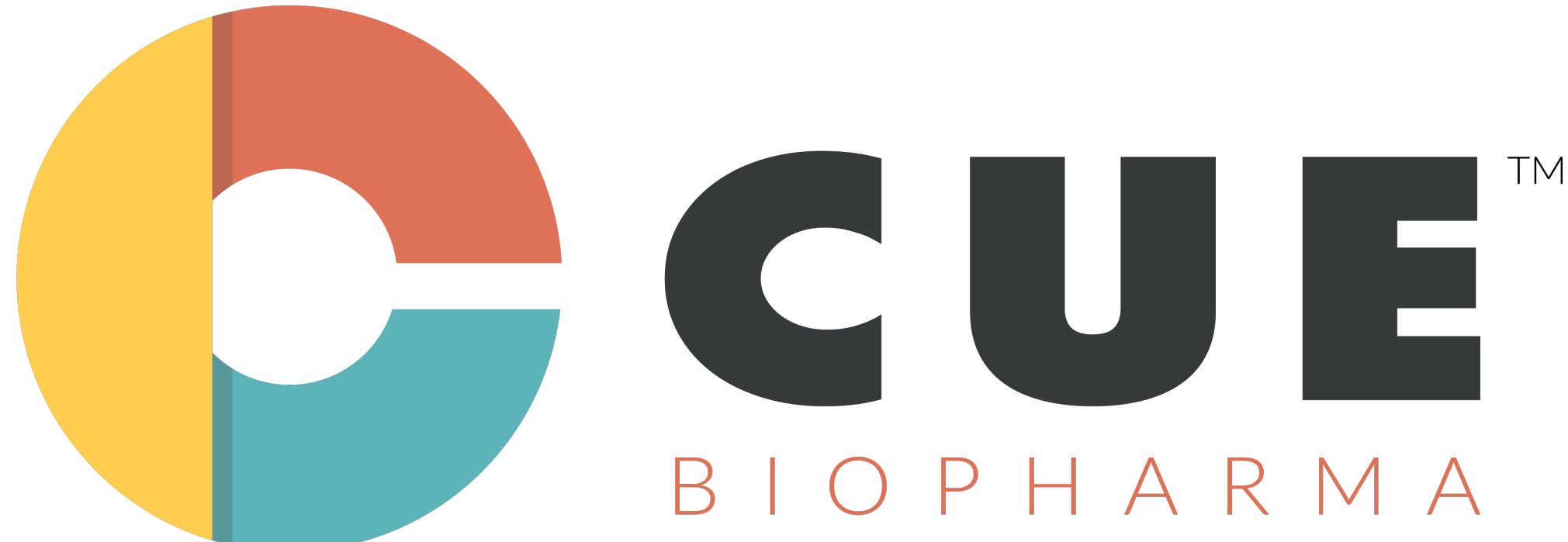


CUE-102 Selectively Activates and Expands WT1-Specific T Cells for the Treatment of Patients with WT1+ Malignancies

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Background

- Immuo-STATs™ (ISTs) are rationally engineered biologics comprised of a bivalent peptide-MHC complex and multivalent co-stimulatory molecules built on an Fc framework to enable stability, valency, favorable PK and manufacturability
- CUE-100 series ISTs are designed to selectively deliver attenuated interleukin-2 (IL-2) to tumor-specific CD8⁺ T cells (Quayle 2020; Seidel 2021)
- Wilms' Tumor 1 (WT1) was previously ranked as the highest priority antigen for therapeutic targeting in an effort by the National Cancer Institute (Cheever 2009)
- Development of novel modalities targeting WT1 provide a significant opportunity to address high unmet medical need in WT1-positive malignancies, including AML, ovarian, endometrial, breast, lung, gastric, colorectal and pancreatic cancer
- CUE-102 is being developed as a novel therapeutic fusion protein to selectively activate tumor antigen-specific T cells to treat WT1-expressing cancers

CUE-100 Series Immuo-STATs

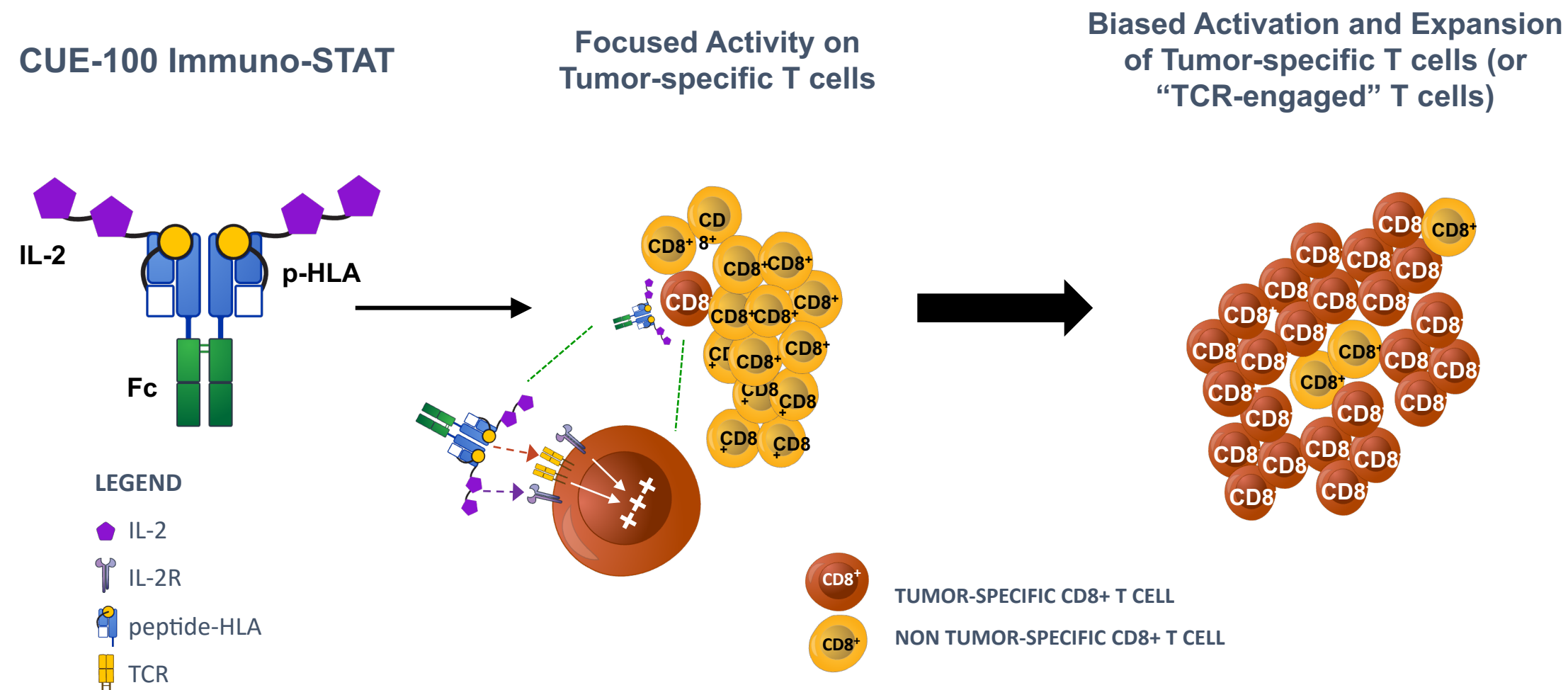
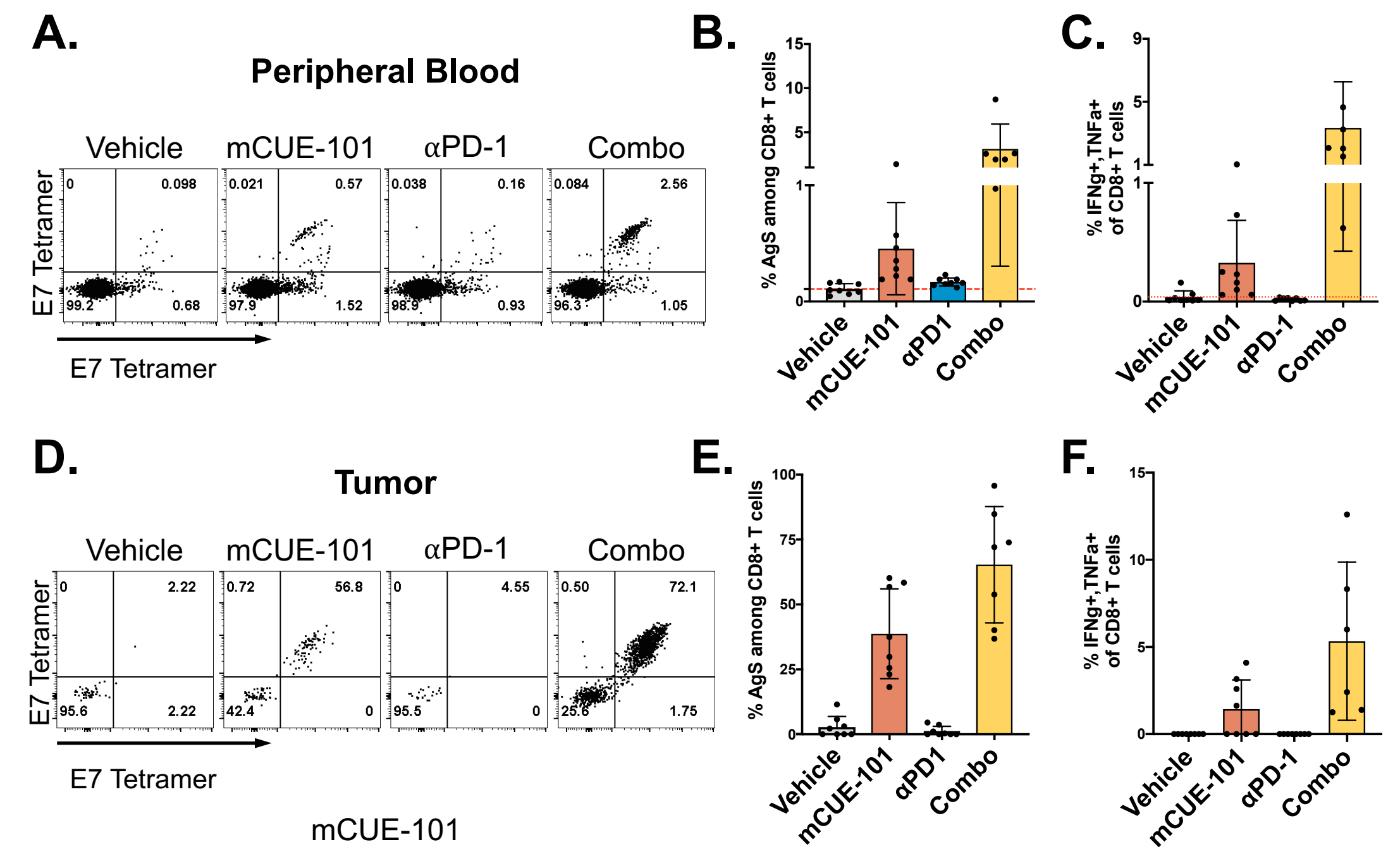


Figure 1 – The rationally engineered and modular biologics of the Immuo-STAT platform incorporate natural biological signals (“cues”) for selective engagement and modulation of disease-relevant T cells. The CUE-100 series framework is designed to selectively deliver modified IL-2 to tumor-specific T cells to drive their expansion.

CUE-100 IST Murine Surrogate (mCUE-101) Expands Antigen-Specific CD8⁺ T Cells in Blood and Tumor



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Figure 2 – mCUE-101 expands functional antigen-specific CD8⁺ T cells in the tumor and the periphery. Mice bearing established TC-1 tumors were treated with 15 mg/kg mCUE-101 alone or in combination with αPD-1. Expansion of antigen-specific cells was assessed one week after the last dose of mCUE-101. Representative flow plots show the frequency of tetramer-positive CD8⁺ T cells in the blood (A & B) and tumor (D & E). Only animals treated with mCUE-101 exhibited increased frequency of antigen-specific T cells, which was greatly increased in the tumor vs blood. mCUE-101 increased the frequency of CD8⁺ T cells that produced IFN γ and TNF α in response to peptide restimulation of splenocytes (C) and tumor-infiltrating lymphocytes (F).

CUE-102

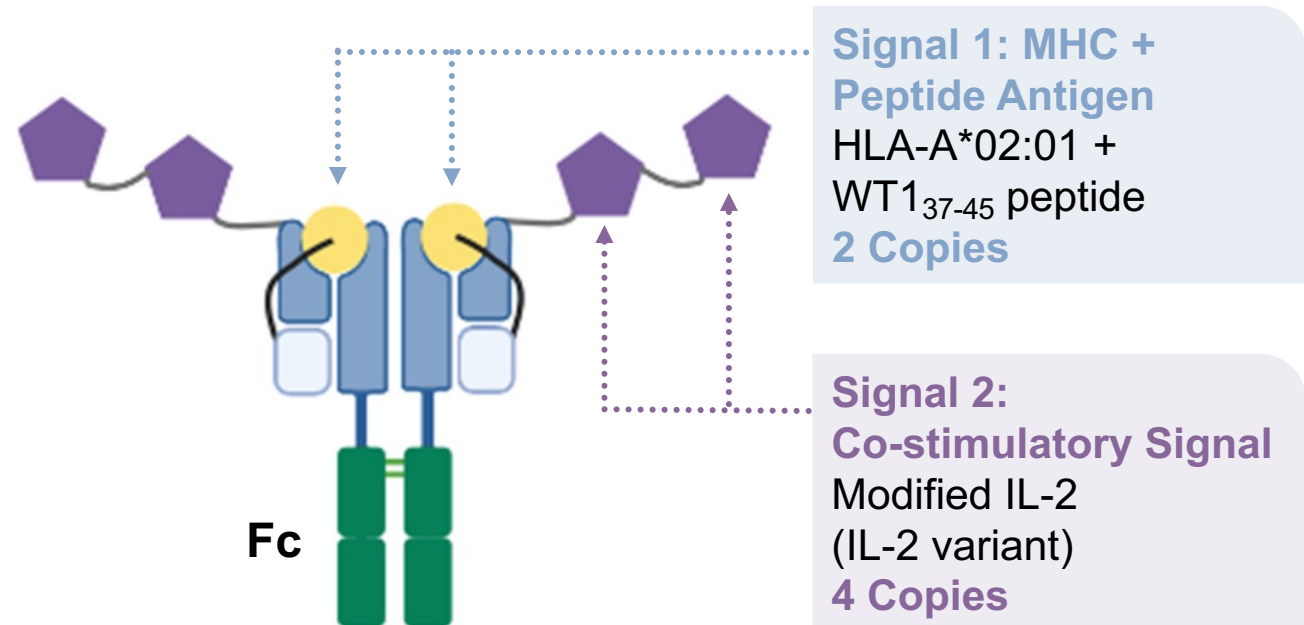


Figure 3 – **Schematic of CUE-102 molecule.** CUE-102 is comprised of a human leukocyte antigen (HLA) complex, HLA-A*0201, a peptide epitope derived from the WT1 protein, and 4 molecules of a reduced affinity human IL-2. CUE-102 is designed to bind and activate WT1-specific T cells for eradication of WT1-positive cancers.

CUE-102 Selectively Binds and Activates Antigen-Specific T Cells

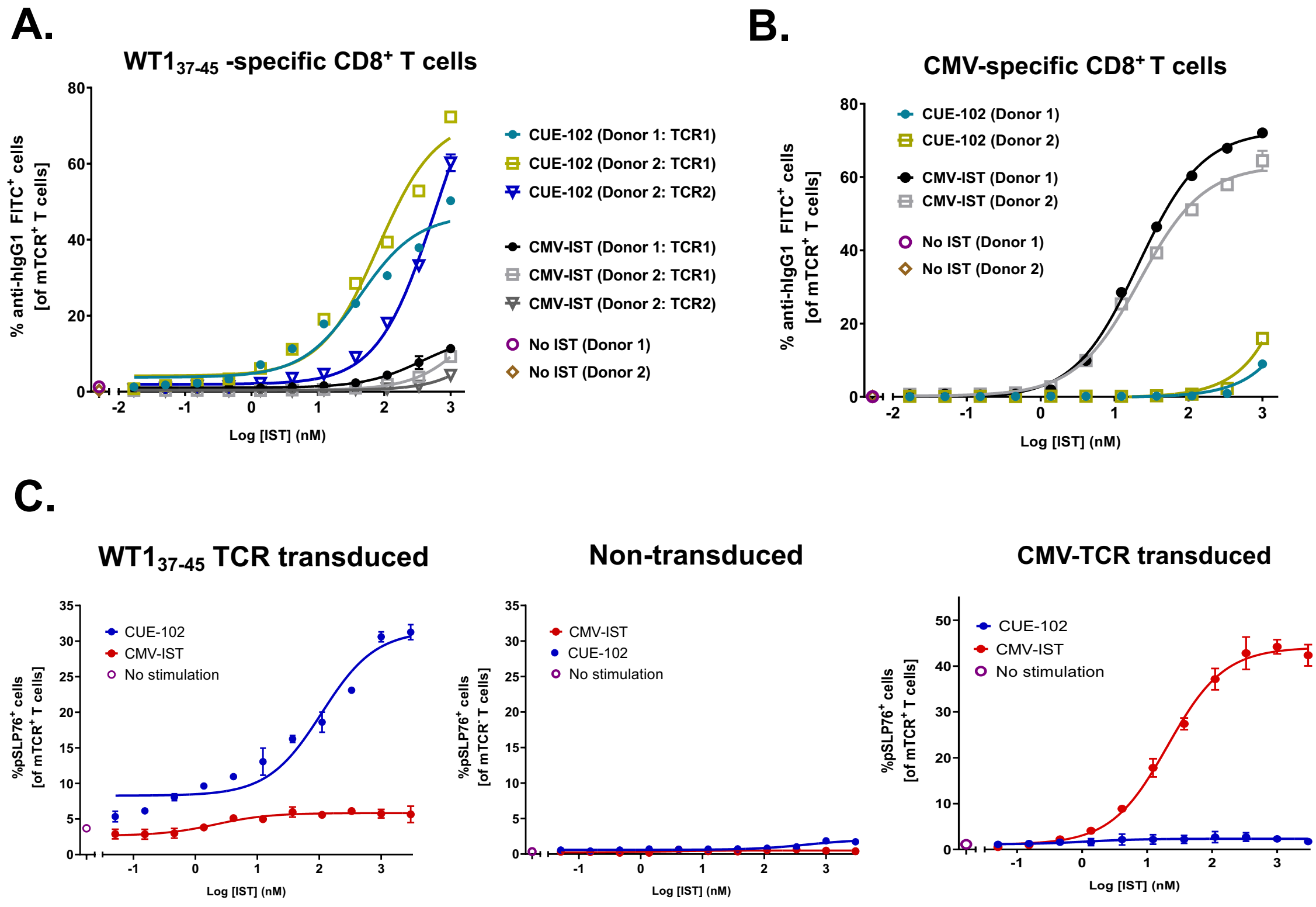


Figure 4 – CUE-102 selectively binds to (A) primary CD8⁺ T cells transduced with WT1₃₇₋₄₅-specific TCR, but not to (B) CD8⁺ T cells transduced with CMV pp65₄₉₅₋₅₀₃-specific TCR. CUE-102 selectively activated the TCR in target cells as measured by increased phosphorylation of SLP76 (pSLP76) in (C) primary CD8⁺ T cells transduced with WT1₃₇₋₄₅-specific TCR, but not in non-transduced cells or CD8⁺ T cells transduced with CMV pp65₄₉₅₋₅₀₃-specific TCRs. CMV-IST: CMV directed Immuo-STAT. TCR: transduced T Cell Receptor reactive to the respective antigen with an engineered murine constant region.

Attenuation of IL-2 Activity in CUE-102

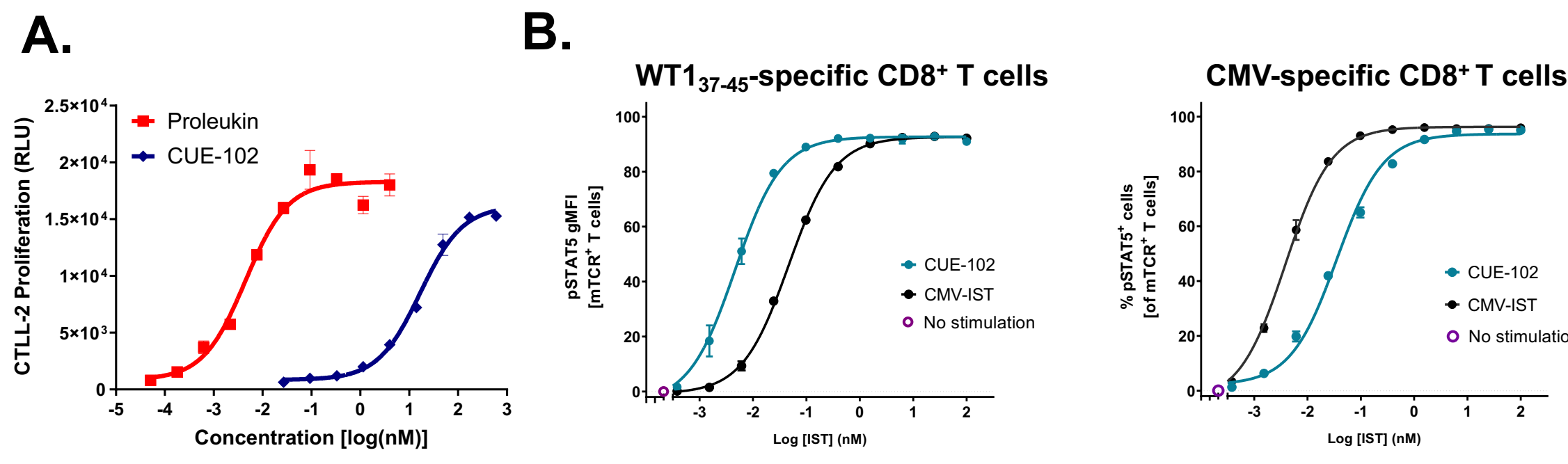


Figure 5 – (A) Human IL-2 molecules on CUE-102 are functionally attenuated and much less potent than recombinant IL-2 (Proleukin®) in a CTL-2 cell proliferation assay. (B) The pHLA specificity of CUE-102 facilitates selective phosphorylation of STAT5 (pSTAT5) immediately downstream of IL-2R on target cells. CUE-102 induces pSTAT5 with greater potency in WT1₃₇₋₄₅ specific CD8⁺ T cells than a CMV-directed IST. A CMV-directed IST induces pSTAT5 with greater potency in CMV pp65₄₉₅₋₅₀₃-specific CD8⁺ T cells than does CUE-102.

CUE-102 Selectively Expands WT137-45-Specific CD8⁺ T Cells from Human PBMCs

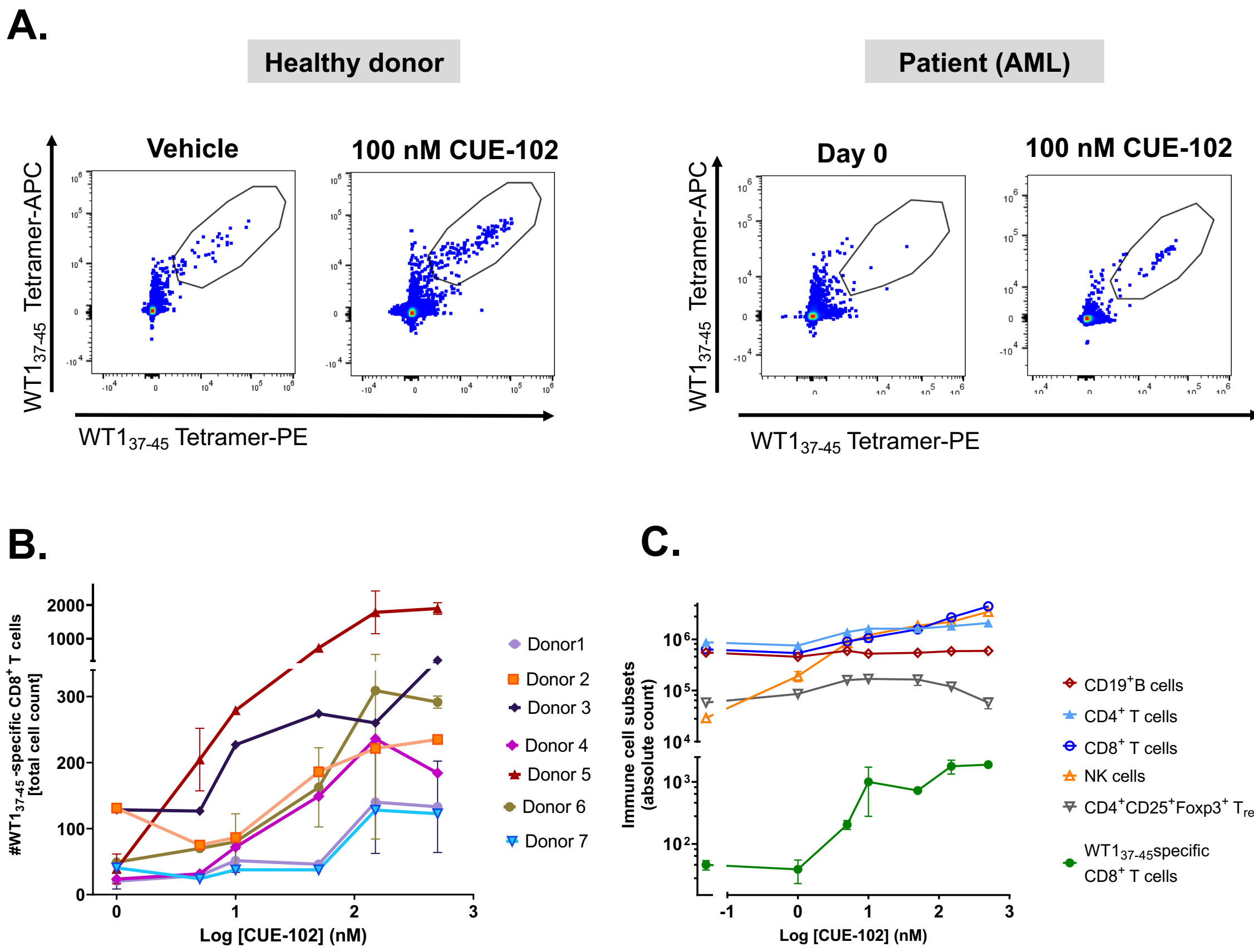


Figure 6 – CUE-102 selectively expands WT1₃₇₋₄₅-specific CD8⁺ T cells from whole human PBMCs *in vitro*. (A) Primary human PBMCs of healthy donors or acute myeloid leukemia (AML) patients were exposed to 100 nM CUE-102 for 10 days. CUE-102 expanded a population of WT1₃₇₋₄₅-specific CD8⁺ T cells as measured by double tetramer staining, while vehicle treatment did not. (B) CUE-102 induced expansion of WT1₃₇₋₄₅ specific CD8⁺ T cells in PBMCs of multiple donors in a dose-dependent manner. (C) Expansion of total NK cells was also observed in response to CUE-102 treatment. Other immune cells in donor PBMCs were not expanded, including CD4⁺ Tregs.

CUE-102 Selectively Expands Tumor Relevant T Cells

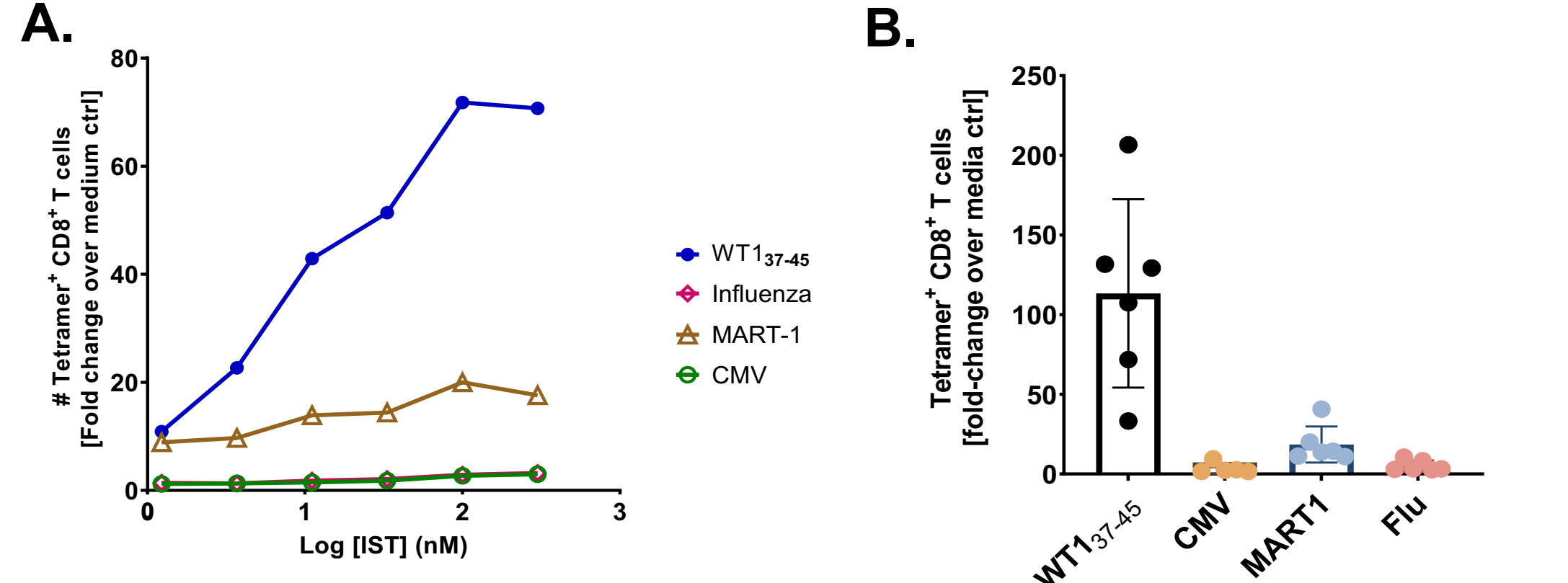


Figure 7 – CUE-102 selectively expands tumor-relevant WT1₃₇₋₄₅-specific CD8⁺ T cells but not CD8⁺ T cells specific to other antigens (CMV, MART1 or Influenza). (A) Dose-dependent and antigen specific expansion from a representative donor. (B) Antigen-selective expansion following exposure of multiple donor PBMCs to 100 nM CUE-102.

CUE-102 Expands Polyfunctional and Cytolytic WT137-45-Specific CD8⁺ T Cells from Human PBMCs

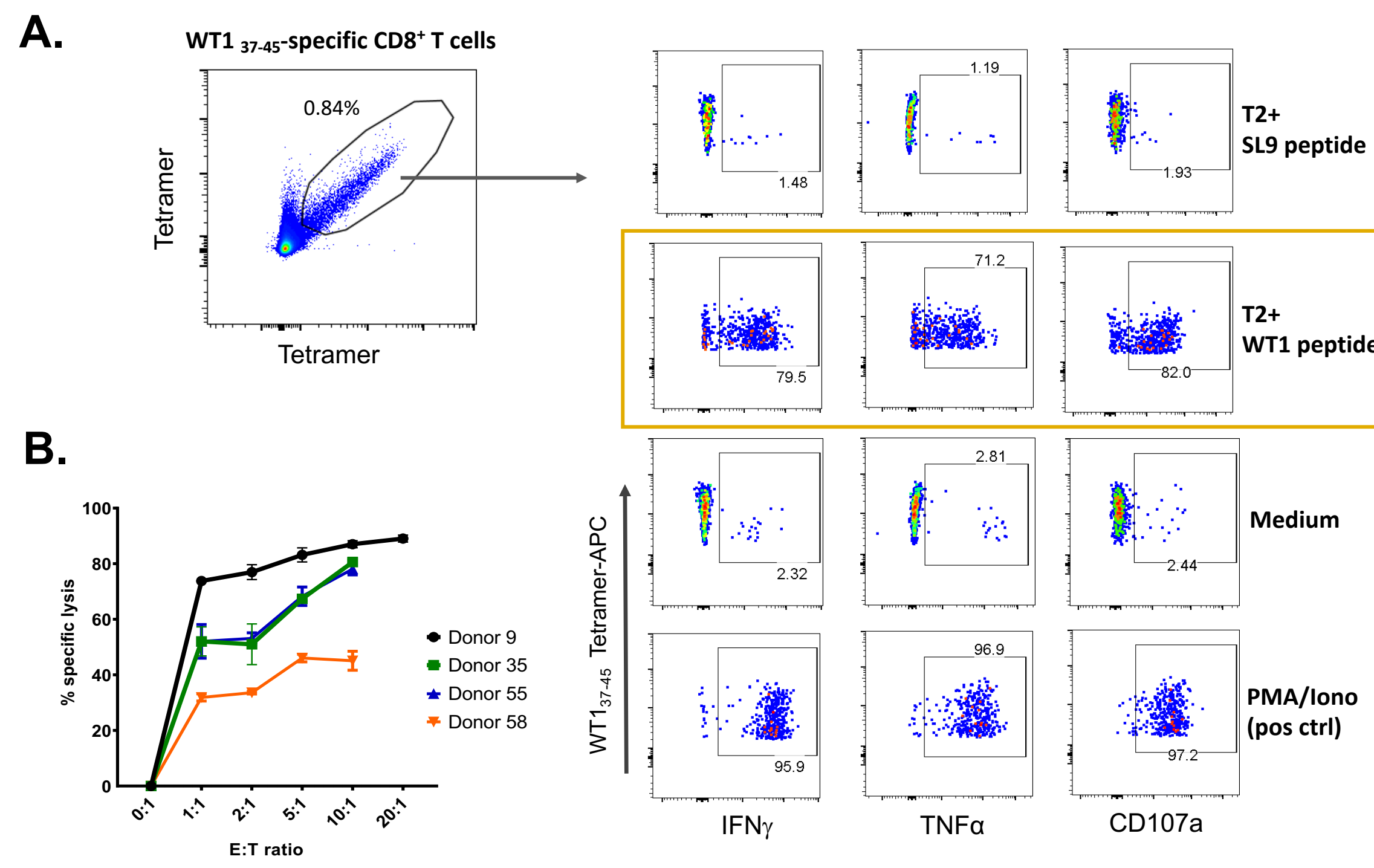


Figure 8 – CUE-102 expanded WT1₃₇₋₄₅-specific CD8⁺ T cells show (A) strong increase of intracellular IFN γ , TNF α and surface CD107a upon challenge with WT1₃₇₋₄₅-presenting T2 target cells, but minimal response towards T2 cells loaded with negative control peptide (T2 target cells). (B) CUE-102 expanded WT1₃₇₋₄₅-specific CD8⁺ T cells induce specific lysis of target T2 cells loaded with WT1₃₇₋₄₅ peptide. Percentage of specific lysis is plotted against effector: target (E:T) ratio of 1:1 - 20:1. Percentage of specific lysis is calculated as (1-(experiment ratio/control ratio)) x 100. Experiment ratio is % T2 (WT1₃₇₋₄₅ peptide) / % T2 (control peptide) in culture with WT1₃₇₋₄₅-specific CD8⁺ T cells for 18 hours. Control ratio is % T2 (WT1₃₇₋₄₅ peptide) / % T2 (control peptide) cultured in medium alone for 18 hours. Mean \pm standard deviation from triplicate wells is shown for each E:T ratio.

CUE-102 Selectively Expands WT137-45-Specific CD8⁺ T Cells in Naïve HLA-A2 Transgenic Mice

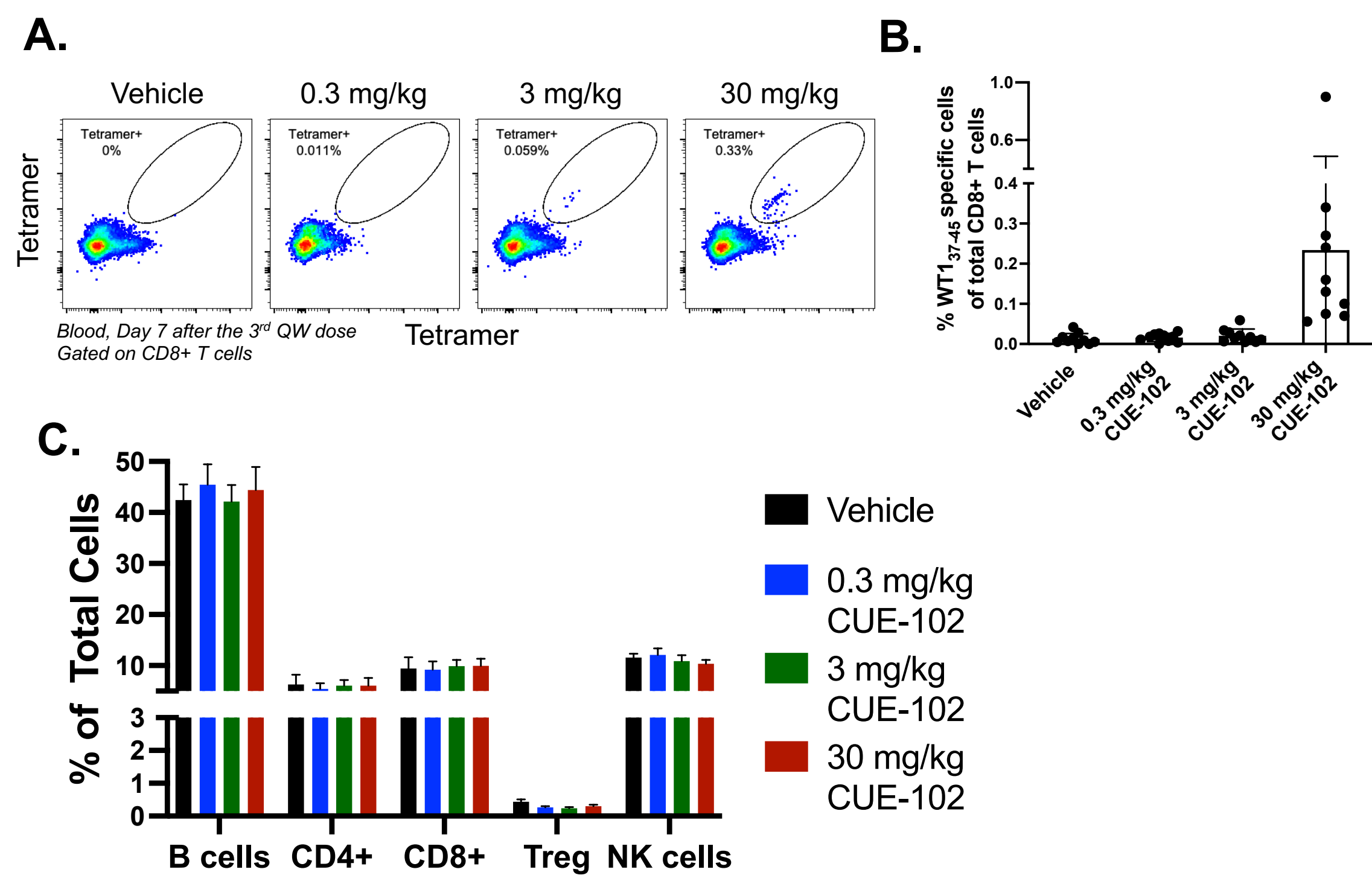


Figure 9 – Treatment of naïve HLA-A2 transgenic mice leads to selective, dose-dependent expansion of WT1₃₇₋₄₅-specific CD8⁺ T cells. Naïve HLA-A2 transgenic mice were given 3 once weekly intravenous (IV) doses of CUE-102 at the indicated dose level. The frequency of WT1₃₇₋₄₅-specific CD8⁺ T cells (A & B) and of major immune lineages (C) was assessed in peripheral blood 7 days after the last dose. Treatment with CUE-102 led to dose-dependent expansion of WT1₃₇₋₄₅ specific CD8⁺ T cells (A & B) without broadly affecting other immune lineages (C).

WT137-45-Specific Cells Expanded In Vivo by CUE-102 are Polyfunctional and Cytolytic

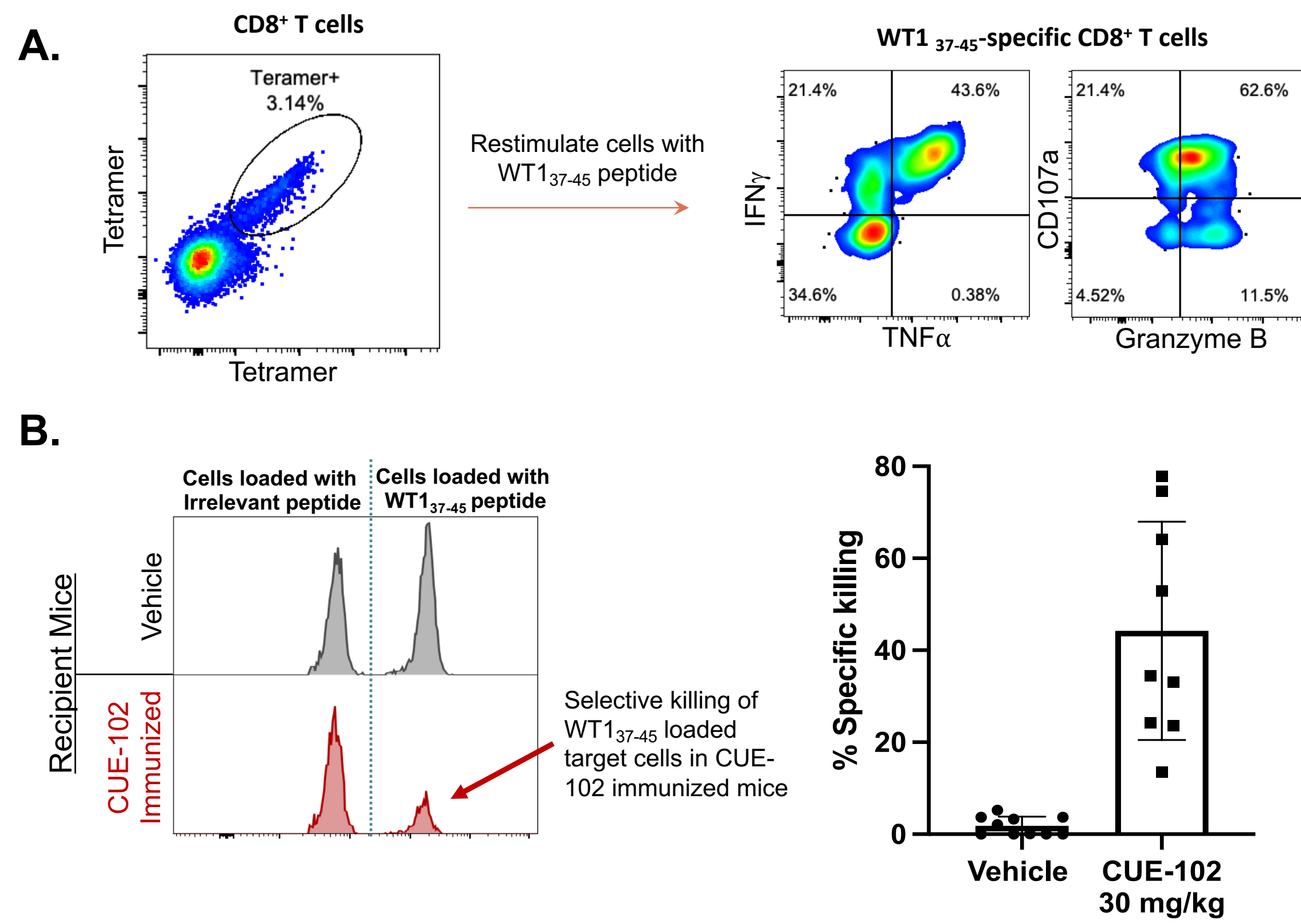


Figure 10 – CUE-102 expands functional CTLs in vivo. (A) The majority of WT1₃₇₋₄₅-specific CD8⁺ T cells (tetramer+) isolated from the spleens of naïve HLA-A2 transgenic mice immunized with 4 doses of CUE-102 produced IFN γ , TNF α , CD107a and Granzyme B in response to ex vivo restimulation. (B) HLA-A2 mice immunized with CUE-102 show antigen-specific in vivo killing of HLA-A2+ target cells pulsed with WT1₃₇₋₄₅ peptide vs. an irrelevant peptide, as shown by the loss of WT1₃₇₋₄₅-labeled target cells in CUE-102 immunized HLA-A2 transgenic mice (red), but not in naïve mice (black).

Repeated Treatment with CUE-102 Results in Cumulative Expansion of WT1-Specific T Cells

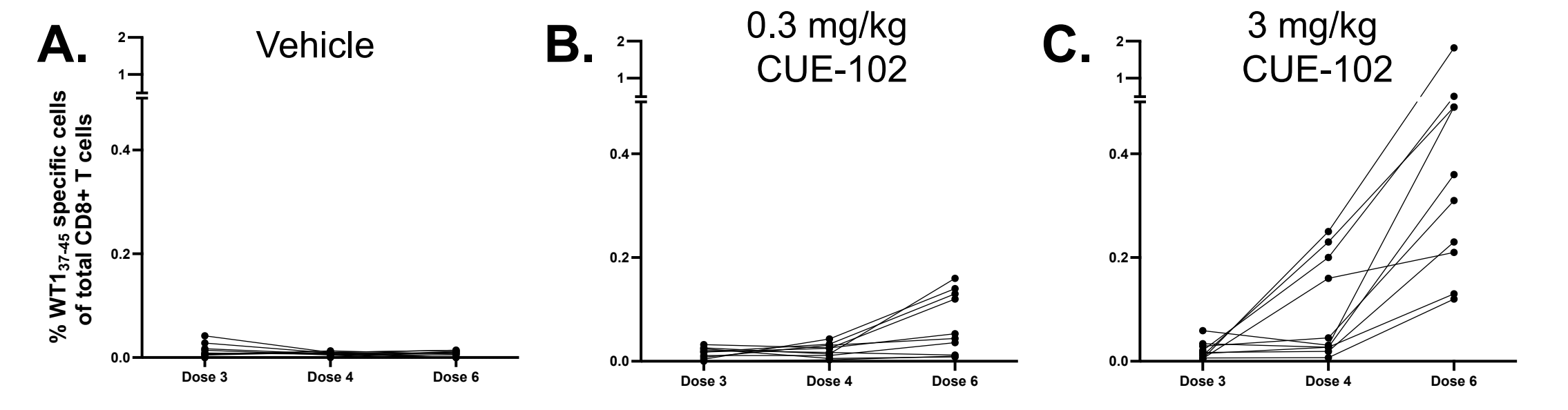
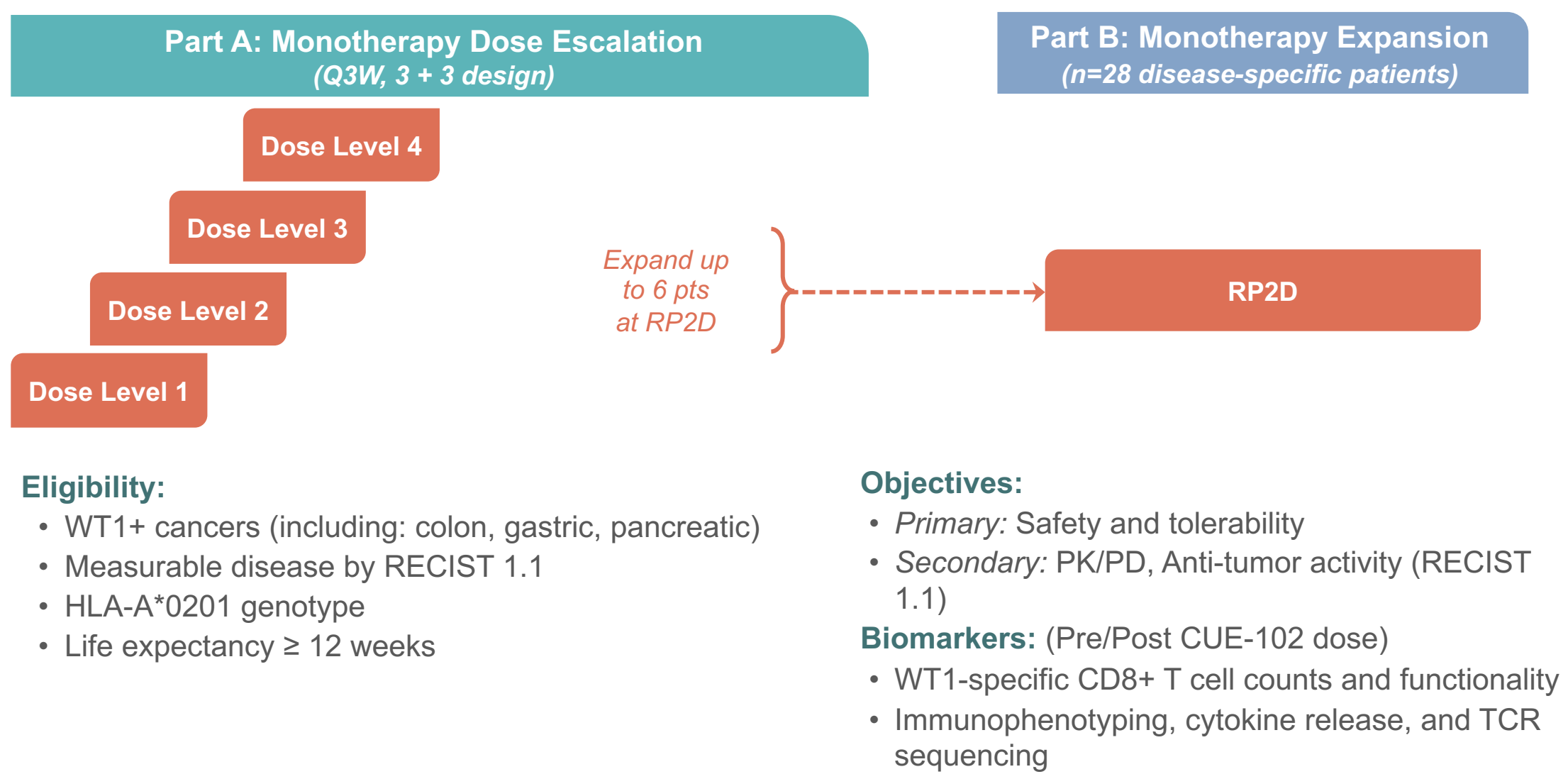


Figure 11 – Repeated treatment with CUE-102 expands WT1-specific CD8⁺ T cells. Graphs display frequencies of WT1₃₇₋₄₅-specific cells among total CD8⁺ T cells in PBMCs from mice that received 3, 4, or 6 total doses of (A) Vehicle, (B) 0.3 mg/kg CUE-102, or (C) 3 mg/kg CUE-102. WT1₃₇₋₄₅-specific cells become detectable in blood at both the 3 mg/kg and 0.3 mg/kg dose levels as the number of doses increases.

CUE-102 First-in-Human Clinical Trial

CUE-102-01 is a Phase 1, FIH study to characterize the safety, tolerability, PK, PD, immunogenicity, and preliminary antitumor activity of CUE-102 in subjects who are HLA-A*0201-positive, have WT1-positive, recurrent/metastatic cancers, and have failed conventional therapies.



Conclusions

- CUE-102, developed in collaboration with LG Chem (Korea), is a novel fusion protein designed to selectively deliver attenuated IL-2 to tumor-specific CD8⁺ T cells
- CUE-102 demonstrates selective binding, activation, and expansion of polyfunctional and cytolytic WT1₃₇₋₄₅-specific primary human CD8⁺ T cells from healthy and cancer patient samples
- Treatment of naïve HLA-A2 transgenic mice with CUE-102 elicits and selectively expands WT1₃₇₋₄₅ specific T cells that are polyfunctional and cytotoxic in vivo
- The novel mechanism of action of CUE-102, namely targeted activation of tumor-antigen-specific CD8⁺ T cells via delivery of reduced affinity mutant IL-2, supports its potential for anti-cancer efficacy in a Phase 1 clinical trial in WT1+ relapsed/metastatic cancers

