

Certificate of Analysis

Cell count	1.7 million per vial
Viability	80%
Sterility	Negative for Bacteria, Yeast, and Fungi

Donor Information

Donor ID	213
Age	27
Gender	Female
Race	Caucasian
Height	5'10"
Weight	210
ABO Type	O negative

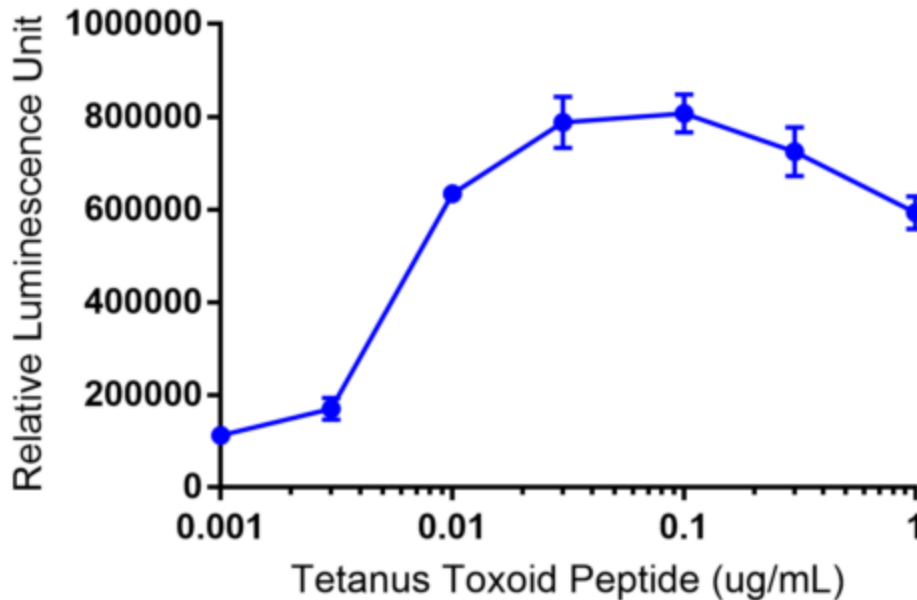
HLA typing

	Allele 1	Allele 2
HLA-A	*0201	*0201
HLA-B	*35	*56
HLA-C	*01	*04
HLA-DRB1	*11	*01

Donors are tested for the blood borne pathogens HIV-1 and 2, Hepatitis B, Hepatitis C and HTLV-1 and are negative. Cells should still be handled as if potentially infectious following biosafety level 2 procedures

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Specific Proliferation of Tetanus Toxoid-Specific T Cells in Response to Increasing Tetanus Toxoid Peptide Concentration



Method. A vial of Tetanus Toxoid-specific T cells was thawed and the cells were cultured for 24 hours in 2 mL of X-VIVO15 medium (Lonza, Walkersville, MD) in a 24 well plate prior to use in proliferation assay.

The proliferation culture was prepared as follows. 50 uL peptide dilutions in X-VIVO 15 at 4X the final concentration were plated into a 96 well round-bottom plate. 20,000 mitomycin-C treated autologous B-Lymphoblastoid cell line (B-LCL) in 50 uL X-VIVO15 were added to each well. The cell suspension was incubated for >30 minutes in a 37C/ 6%CO₂ incubator prior to adding 20,000 T cells (see above) in 100 uL X-VIVO 15. The culture was incubated for 4 days in a 37C/ 6%CO₂ incubator.

Using a multichannel pipetter, 50 uL of cell suspension from each well was transferred into a white flat-bottom 96 well plate. 50 uL of CellTiter-Glo Cell Proliferation Reagent (Promega, Madison, WI) was added to each well. After 10 minutes of incubation at room temperature, the relative luminescence was read in an automatic plate reader. Data is presented as the mean relative luminescence unit (RLU) +/- standard deviation value vs the tetanus toxoid peptide concentration.

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