Supplemental Data 1

Detailed flow cytometric analysis of expanded cells from children had not been previously performed. Therefore, we analyzed the expanded products form the children reported in this study and from 5 adult products for comparison. We did not find any substantive differences.

Tfh cells were defined as: CD4+CXCR5+PD1+Bcl6+ICOS+ (expressed as a percent of CD3)

CD8 Tscm cells were defined as: CD3+CD8+CCR7+CD45RA+CD45RO-CD62L+CD27+ CD28+CD127+CD95+CD122+ (expressed as a percent of CD8)

CD4 Tscm cells were defined as: CD4+CCR7+CD45RA+CD45RO-CD62L+CD27+CD28+CD127+CD95+CD122+ (expressed as a percent of CD4)

Treg cells were defined as: CD4+CD25hiCD127-FoxP3+ (expressed as a percent of CD4)

Tregi were defined as: CD4+CD25hiCD127-FoxP3+CD45RO+PD1+ (expressed as a percent of CD4)

16 Parameter flow cytometry was optimized and performed on a modified LSR II equipped with 4 lasers (Red, Blue, Violet and Green)

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| **Non-intrinsic Parameters** | **Fluor** | **Ab Clone** | **Vendor** | **Catalog Number** | **Isotype** |
| CD3 | APC-Cy7 | SK7 | BD Bioscience | 557832 | Ms IgG1,k |
| CD8 | AF700 | RPA-T8 | BD Bioscience | 557945 | Ms IgG1,k |
| Violet Viability Dye | PacBlue | na | Invitrogen | L34955 | na |
| CD4 | BV605 | OKT-4 | Biolegend | 317437 | Ms IgG2a, k |
| CD14 | PacBlue | M5E2 | BD Bioscience | 558121 | Ms IgG2a, k |
| CD16 | PacBlue |  | BD Bioscience | 558122 | Ms IgG1,k |
| CD19 | PacBlue | HIB19 | Biolegend | 302223 | Ms IgG1,k |
| CD127 | BV650 | A019D5 | Biolegend | 351325 | Ms IgG1,k |
| CD27 | APC | M-T271 | BD Bioscience | 558664 | Ms IgG2a,k |
| CD28 | PerCPCy5.5 | CD28.2 | Biolegend | 302922 | Ms IgG2a,k |
| CD45RA | BV570 |  | Biolegend | 304131 | Ms IgG2a,k |
| CD45RO | ECD |  | Beckman Coulter | IM2712U | Ms IgG2a,k |
| CD95 | PECy7 | DX2 | Biolegend | 305622 | Ms IgG2a,k |
| CD62L | FITC | CREG-56 | Biolegend | 304804 | Ms IgG2a,k |
| CD122 (IL2R beta) | PE |  | Biolegend | 339006 | Ms IgG2a,k |
| CCR7 (CD197) | BV711 | G043H7 | Biolegend | 353227 | Ms IgG2a,k |

Healthy donor peripheral blood



Expanded products from a pediatric patient



**Comparison of adult and pediatric expanded products**

|  |  |  |  |
| --- | --- | --- | --- |
| Population | Adults (n=5) | Children (n=18) | P value |
|  |  |  |  |
|  |  |  |  |
| **CD4 Naive** | **1.6 + 1.4** | **3.2 + 8.5** | **<0.05** |
| CD8 Naive | 3.5 + 5.3 | 3.5 + 3.1 | 0.11 |
| Tfh | 7.4 + 5.5 | 5.0 + 4.0 | 0.31 |
| CD4 Tscm | 0.79 + 1.4 | 1.1 + 1.5 | 0.69 |
| CD8 Tscm | 0.56 + 0.82 | 0.62 + 1.0 | 0.91 |
| Treg | 7.6 + 3.9 | 4.4 + 4.1 | 0.16 |
| Tregi | 5.6 + 3.3 | 3.4 + 4.0 | 0.30 |