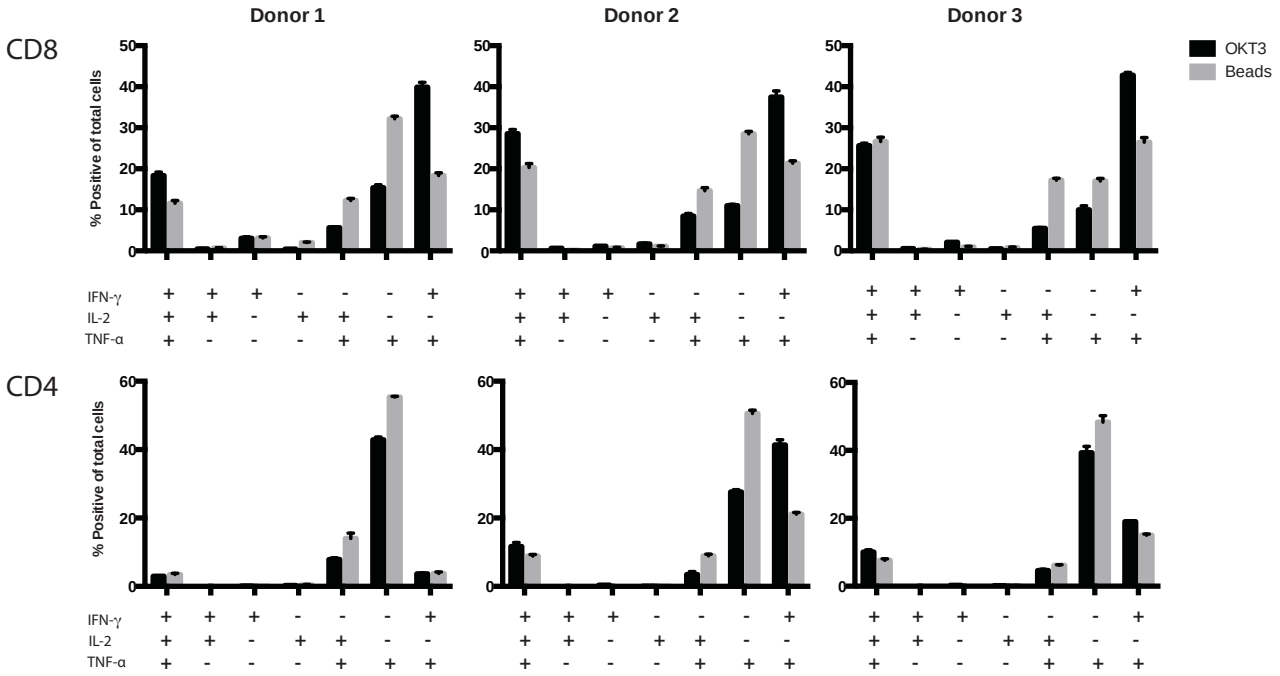
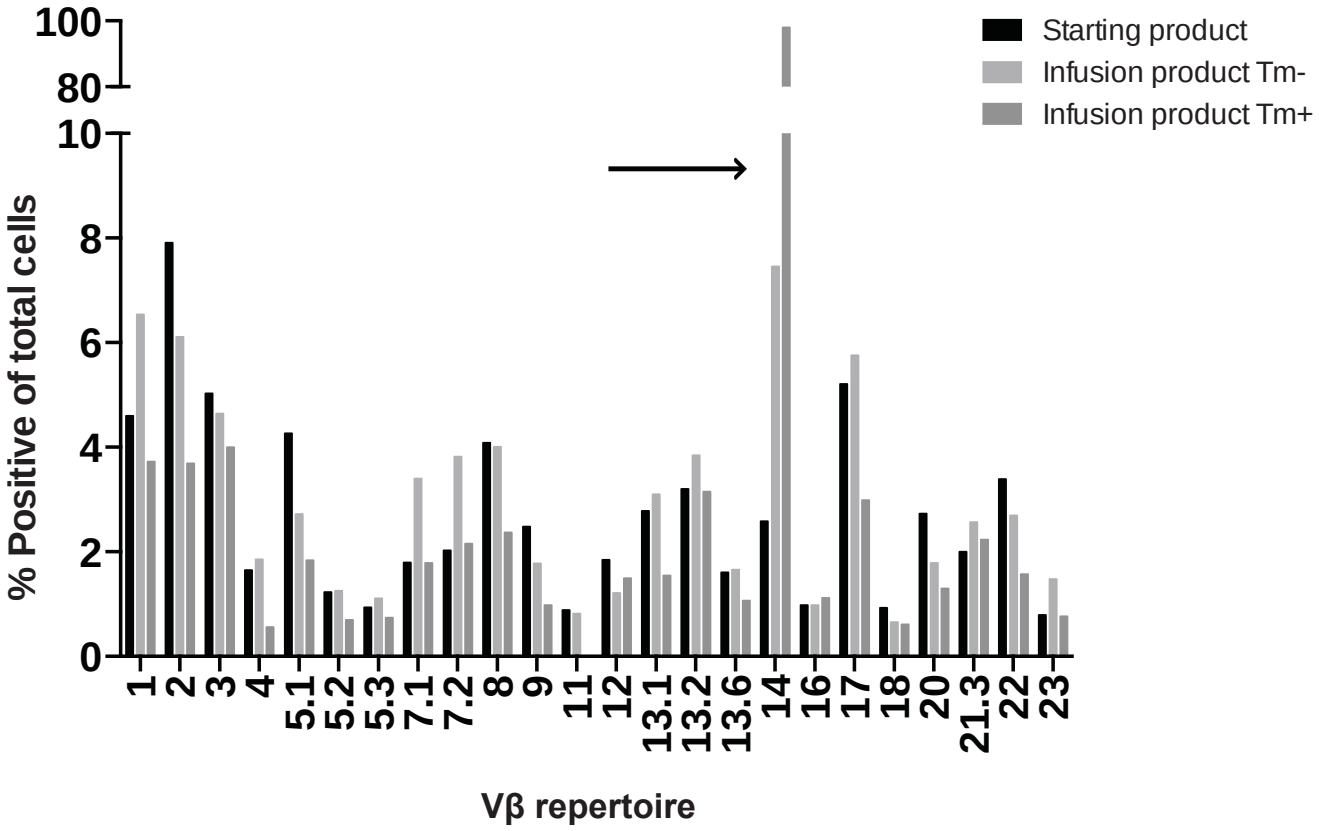
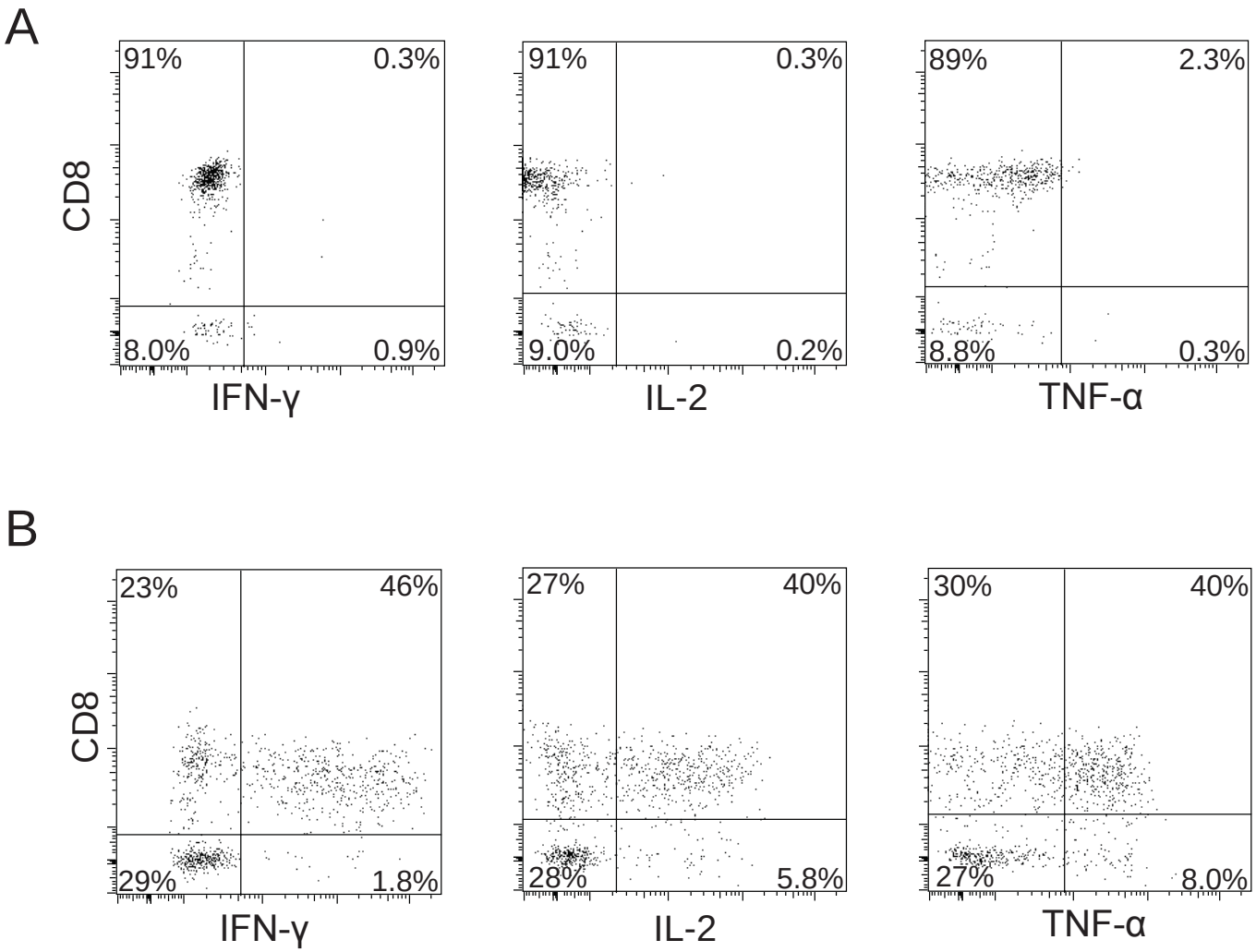
Supplementary fig. 1



Supplementary Figure 1. Intracellular cytokine production of TCR 1D3HMcys-transduced T-cells using two different activation/stimulation protocols. Three different healthy donor derived PBMC's were stimulated and subsequently transduced with the 1D3HMcys TCR and cultured for 11 days with either OKT3 (50 ng/ml) and IL-2 (200 IU/ml) or with anti-CD3/CD28 beads and IL-7/IL-15 (5 ng/ml each). TCR-transduced cells were incubated with HLA-A2$^+$ MART-1$^+$ (mel624) or HLA-A2$^-$ MART-1$^+$ (mel938) melanoma cell lines or with PMA (50 ng/ml) and Ionomycin (1ug/ml) as positive control. Intracellular cytokine production for IFN-γ, IL-2 and TNF-α was determined, by flow cytometry, for the TCR$^+$ cells after an overnight incubation. For three different donors, boolean gating is shown for transduced CD8$^+$ cells (top row) and transduced CD4$^+$ cells (bottom row) after co-culture with mel624. Bars show average of triplicates with SD (bars).



Supplementary Figure 2. Transduced T cells have a wide TCR repertoire and are polyclonal. Starting material (before transduction) and the infusion product from the patient was stained for the TCR Vβ repertoire using an IOTest® Beta Mark kit according manufactur's protocol (Beckman Coulter). Arrow indicates the Vβ14 of the introduced TCR. Note: the % in the Tm^+ cells might be an underestimation due to downregulation of the endogenous TCR.



Supplementary Figure 3. Cytokine production of patient derived TCR modified T cells.
$2x10^6$ patient derived PBMC's (in which 9% of lymphocytes are 1D3HMcys positive), collected 7 days post infusion, were cultured alone **(A)** or in the presence of $2x10^5$ HLA-A2$^+$ MART-1$^+$ (mel624) melanoma cells **(B)**. Intracellular cytokine production for IFN-γ, IL-2 and TNF-α was determined, by flow cytometry after an overnight incubation. Plots are gated on CD3$^+$ TCR$^+$ cells.