Differences in cART-mediated immune reconstitution are revealed by distinct exhaustive phenotypes of HIV-1- and CMV-specific CD8⁺ T cells

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Introduction

Studies on long-term-non progressors (LTNP) have consistently showed that retaining a proliferative T-cell capacity is key for control of HIV-1 infection, however this is characteristically lost in the majority of HIV-1⁺ individuals (1). The mechanisms behind this are still unclear warranting further exploration. During HIV-1 infection there is an increased expression of the negative immunoregulatory molecules Programmed cell Death-1 (PD-1) and T-cell immunoglobulin mucin-3 (TIM-3) (2,3), which have been implicated in an exhaustive CD8⁺ T-cell dysfunction. Ex-vivo blockade of PD-1 to its ligand (PD-L1) has been shown to enhance the proliferative capacity of CD8⁺ T Cells (4), but it remains unclear how this impacts cART-mediated immune reconstitution. Here we assessed the effect of cART on proliferative responses to HIV-1 and CMV in parallel to the exhaustive phenotypes of HIV-1- and CMV-specific CD8⁺ T cells to better understand how this may affect immune reconstitution.

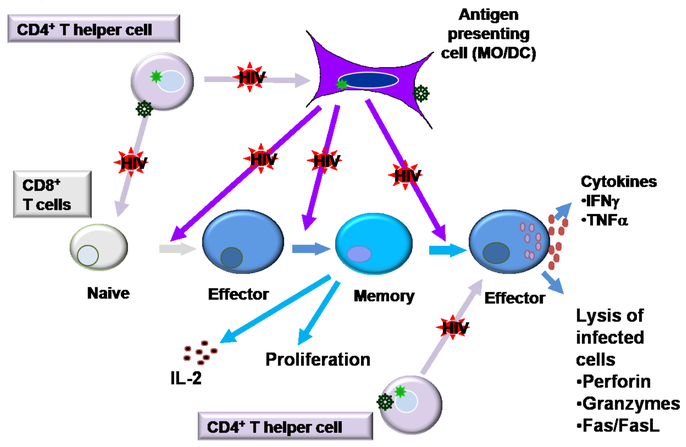

Figure 1. HIV-1 infection interferes with several key points of CD8 function, including differentiation, activation and exhaustion.

Methods

- Proliferative responses by 3H-thymidine incorporation, to CMV and HIV-1 Gag p24 were assessed over a period of 96 weeks, in HIV-1⁺ patients commencing cART.
- In addition, proliferative responses of long-term-nonprogressors (LTNP), chronically infected cART-naïve progressors and HIV seronegative individuals were assessed in parallel.
- Multimer technology was used to examine the immune activation (CD38, HLA-DR), maturation (CCR7, CD45RA) and exhaustion (PD-1, TIM-3) profiles of total, CMV- and HIV-1- specific CD8⁺ T cells *ex vivo* in cART-naïve and treated individuals.
- Statistical analysis was performed using the Kruskal-Wallis and Mann-Whitney U test with the Bonferroni correction for multiple analysis testing. Significance was defined as $p<0.05$.

References

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2. Day *et al.,* (2006) *Nature* 443, 350-354
3. Jones *et al.,* (2008) *J Exp Med* 205, 2763-2779
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Acknowledgements

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Results

- cART initiation → Increased proliferative responses to CMV but not to HIV-1 Gag p24
- LTNP → greater proliferative responses to CMV and HIV-1 Gag p24 compared to chronic progressors and healthy controls
- Higher frequencies of HIV-1-specific CD8⁺ T cells expressing PD-1 and TIM-3 compared to CD8⁺ T cells specific for CMV

Proliferative responses

Longitudinal analysis

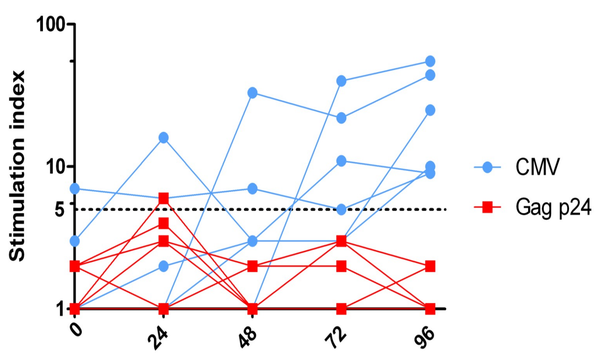


Figure 2. All six HIV-1⁺ individuals studied exhibited increased T-cell proliferative responses to CMV following 96 weeks of cART, however responses to Gag p24 remained undetectable (median stimulation index: 17.5 (9 to 55) and 1.5 (1 to 2) respectively).

Cross-sectional analysis

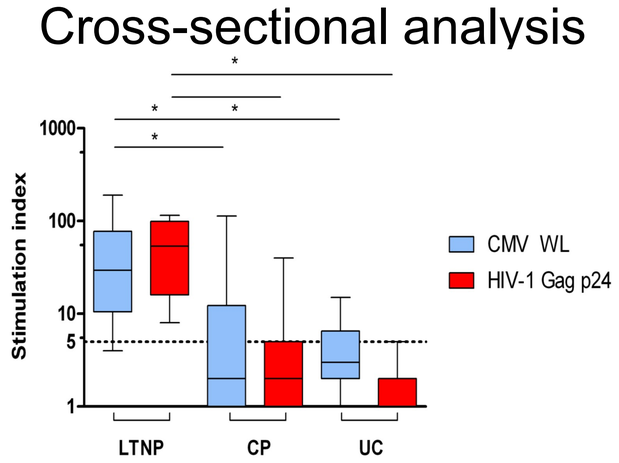
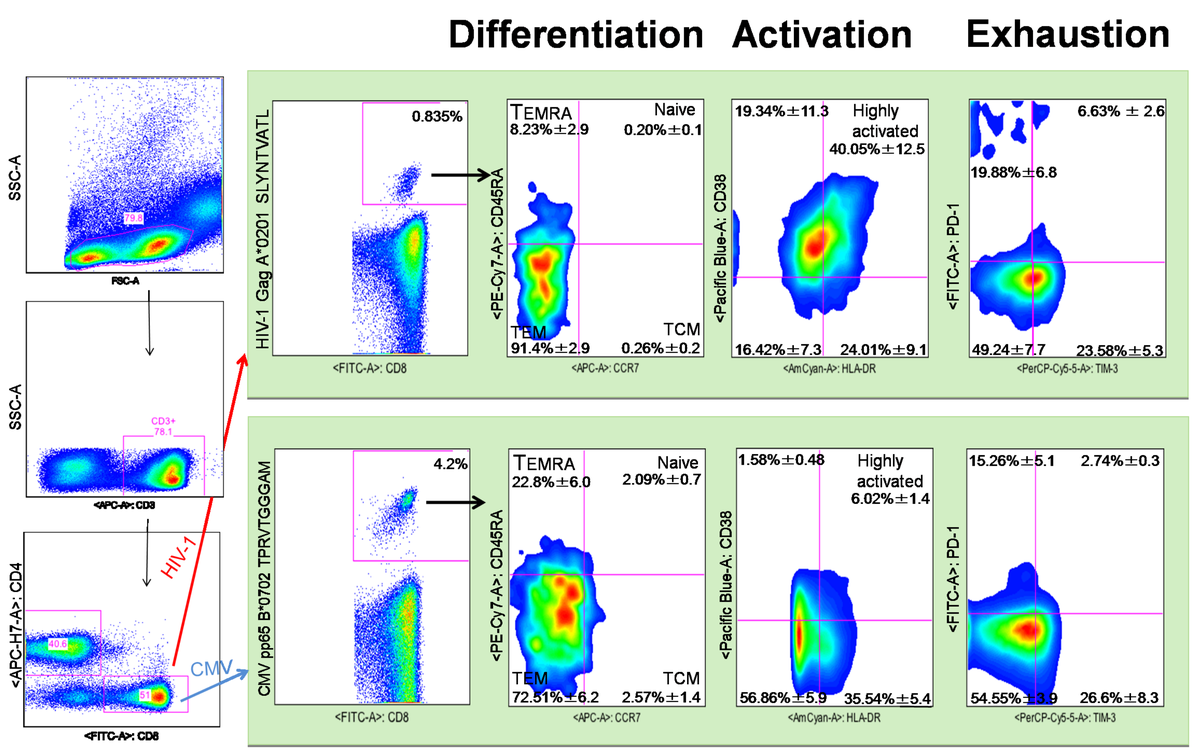


Figure 3. Significantly higher proliferative response to CMV and Gag p24 in LTNP (n=10) when compared to both healthy uninfected controls (UC; n=16) and cART-naïve HIV-1⁺ patients (chronic progressors, CP; n=54; p<0.0025 for all).

Phenotypic analysis



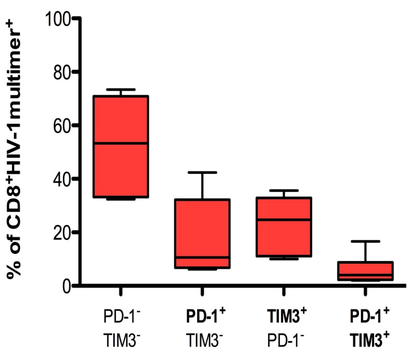


Figure 5. Exhaustive phenotype of HIV-1⁺multimer⁺ (SLYNTVATL) CD8⁺ T cells in cART-naïve HIV-1⁺ individuals (n=6).

Figure 4. Phenotypic characterization using multimer technology. Variable frequencies of CMV-specific CD8⁺ T cells expressing PD-1 (7.6 to 24.0%) and TIM-3 (12.3 to 40.9%) were observed. Furthermore, there was a trend for a higher proportion of HIV-1 Gag-specific CD8⁺ T cells to express both PD-1 and TIM-3 compared to CMV TM10-specific CD8⁺ T cells (6.63±2.59 and 2.74±0.26 respectively; n=6; two graphs furthest to the right), however a more in depth analysis is needed.

Conclusion

- The distinct cART-mediated reconstitution of CMV-specific proliferative responses compared to HIV-1 Gag p24 responses may be attributed to the observed higher frequencies of TIM-3 and PD-1 expressing CD8⁺ T cells specific for HIV-1 compared to CMV.
- This may impact the reconstitution potential of proliferative responses in HIV-1⁺ patients shown to be important for virus control.

Clinical implications

- Future therapeutic interventions should focus on targeting PD-1 and/or TIM-3 in conjunction with current cART regiments.
- This might result in improved HIV-1-specific immune reconstitution, including enhanced proliferative responses, compared to using cART alone.

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