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

Nathali Grageda*1, S J Westrop1, M Nelson2, S Mandalia1,2, A Jackson2, G Moyle2 and N Imami1

1Imperial Coll London, UK, 2Chelsea and Westminster Hosp, London, UK

Introduction

Studies on long-term non progressors (LTNP) have consistently shown 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.

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=15) when compared to both healthy uninfected controls (UC, n=16) and cART-naive HIV-1+ patients (chronic progressors, CP, n=54; p<0.0025 for all).

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

Phenotypic analysis

Figure 4. Phenotypic characterization using multimer technology. Variable frequencies of CMV-specific CD8+ T cells expressing PD-1 (7.4 to 24.6) and TIM-3 (12.3 to 40.3%) 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 ± 0.28 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 regimens.

• This might result in improved HIV-1-specific immune reconstitution, including enhanced proliferative responses, compared to using cART alone.

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-naive 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-naive 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


Acknowledgements

The authors would like to thank patients and staff at Chelsea & Westminster Hospital who participated in this study. This work was supported by the Westminster Medical School Research Trust and St Stephen’s AIDS Trust.

Introduction

Studies on long-term non progressors (LTNP) have consistently shown 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.

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=15) when compared to both healthy uninfected controls (UC, n=16) and cART-naive HIV-1+ patients (chronic progressors, CP, n=54; p<0.0025 for all).

Phenotypic analysis

Figure 4. Phenotypic characterization using multimer technology. Variable frequencies of CMV-specific CD8+ T cells expressing PD-1 (7.4 to 24.6) and TIM-3 (12.3 to 40.3%) 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 ± 0.28 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 regimens.

• This might result in improved HIV-1-specific immune reconstitution, including enhanced proliferative responses, compared to using cART alone.