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HIV latency is associated with changes in the Wnt1, β-catenin, TGF-β and BCL-2 signalling pathways

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Introduction

Although HAART has significantly improved the survival of people living with HIV (PLWH), lifelong treatment remains essential because it does not completely eradicate the infection. HIV persists in resting CD4+ T cells and can reappear when therapy is interrupted. The molecular mechanisms underlying latency are indeed numerous and complex. Recent evidence has highlighted the critical role of CD8 T cells in controlling HIV DNA levels, although data on this aspect remain limited. Of note, Wnt secretion by CD8+ T cells has observed to stimulate the Wnt/β-catenin pathway in CD4+ T cells, which regulates HIV latency by inhibiting its transcription. In addition, TGF-β enhances HIV-specific CD8+ T-cell function, which is negatively correlated with HIV DNA levels. Both pathways affect the regulation of apoptosis, with Wnt/β-catenin inhibiting apoptosis and TGF-β promoting it in the context of HIV latency.

Study Design

On the light of these consideration, the aim of this study was to investigate the role of Wnt1, β-catenin, TGF-β pathway and the anti-apoptotic gene BCL-2 in HIV latency by examining their expression levels in both CD4+ and CD8+ T cells.

Methods

Peripheral blood mononuclear cells (PBMCs) were collected from HAART-naïve (n=10) and HAART-treated (n=19) PLWH recruited at the Department of Public Health and Infectious Diseases of "Sapienza" University of Rome (Italy). Total HIV DNA amount was determined using primers and probe that recognize the gag gene by RT/real-time PCR. CD4+ and CD8+ T cells were isolated from PBMCs by positive immunomagnetic selection using CD4 or CD8 micro beads (Miltenyi Biotec). For each cell population, mRNA levels of Wnt1, β-catenin, TGF-β, SMAD2, SMAD3, SMAD4 and BCL-2 were evaluated using RT/real-time PCR. Statistical analysis was performed using PRISM and $p<0.05$ were considered statistically significant.

Results

As expected, HAART-treated individuals had reduced HIV DNA levels compared to HAART-naïve PLWH ($p<0.0001$). Wnt1 mRNA expression in CD8+ T cells increased in HAART-treated compared to HAART-naïve individuals ($p=0.0368$). β-catenin ($p=0.0017$; $p=0.0096$), TGF-β ($p=0.0003$; $p=0.0167$), SMAD2 ($p=0.0006$ for both), SMAD3 ($p=0.0021$; $p=0.0019$), SMAD4 ($p<0.0001$; $p=0.0002$) and BCL-2 ($p=0.0005$; $p=0.0003$) mRNA expression levels in both CD4+ and CD8+ T cells were increased in HAART-naïve compared to HAART-treated individuals. Moreover, HIV DNA levels were correlated negatively with the expression levels of all analyzed genes in CD4+ T cells and Wnt1, SMAD4 and BCL-2 mRNA expression in CD8+ T cells.

Figure 1 **Comparison of HIV-DNA levels between HAART-naïve and HAART-treated PLWH**

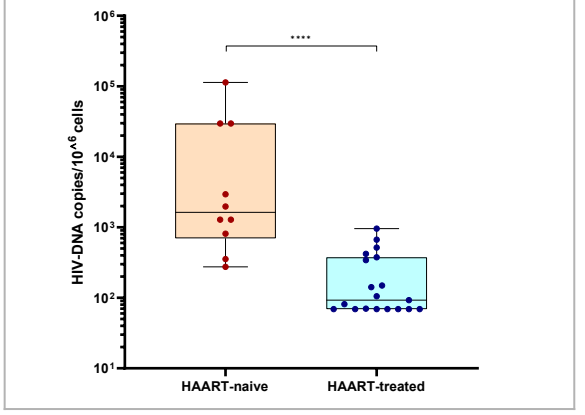
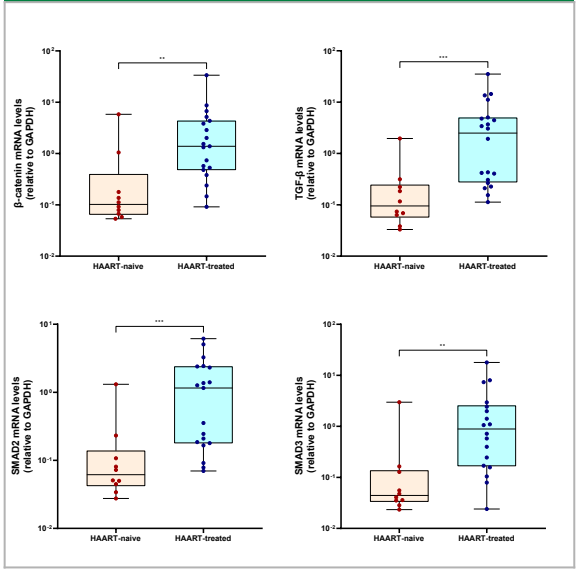


Figure 2 **Comparison of β-catenin, TGF-β, SMAD2, SMAD3, SMAD4 and BCL-2 mRNA levels in CD4⁺ cells between HAART-naive and HAART-treated PLWH**



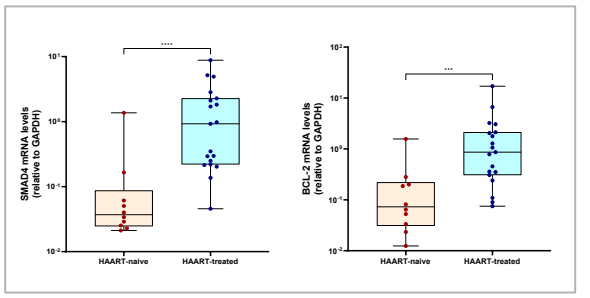
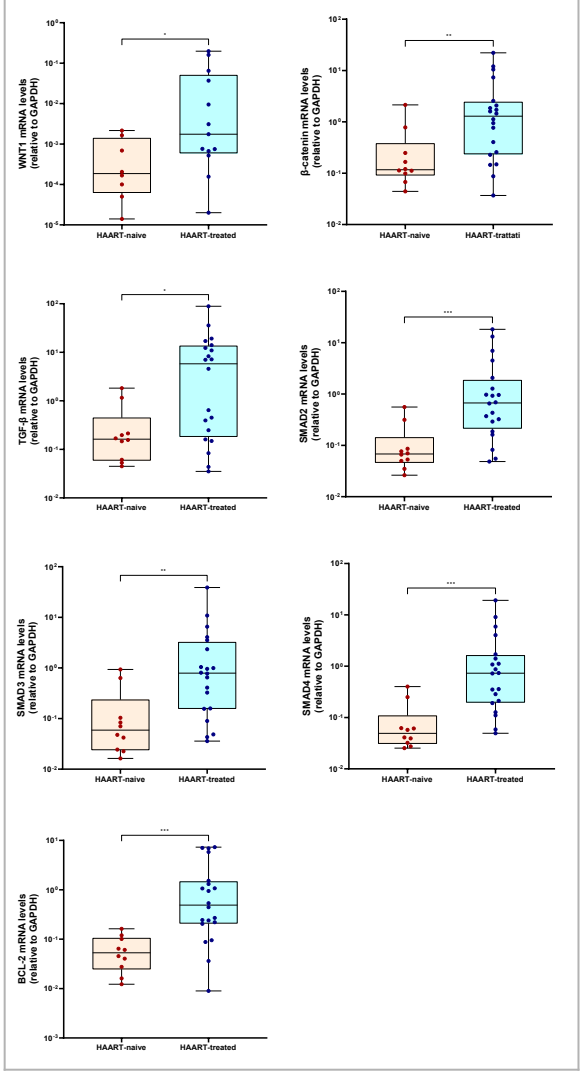


Figure 3 **Comparison of β-catenin, TGF-β, SMAD2, SMAD3, SMAD4 and BCL-2 mRNA levels in CD8⁺ cells between HAART-naive and HAART-treated PLWH**



Conclusion

These results suggest that low HIV DNA levels correlate with increased expression of Wnt1, β-catenin, TGF-β pathway genes and BCL-2, suggesting a potential role for these cellular pathways in controlling HIV latency.

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