ALTERED CELLULAR IMMUNE RESPONSE IN THE CONTEXT OF SYPHILIS AND ACUTE HIV COINFECTION

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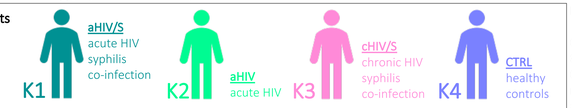
Background

The co-infection of syphilis and acute human immunodeficiency virus (HIV) is becoming increasingly significant in clinical contexts, given the escalating rates of co-infection and the intricate interplay between these two sexually transmitted infections. Syphilis enhances HIV transmission and acquisition, while HIV accelerates the progression of syphilis. This study aimed to investigate the cellular immune responses in the context of acute HIV/syphilis co-infection.

Methods

A cross-sectional analysis was conducted, comparing four cohorts of 15 patients each (total N=60): acute HIV infection with syphilis (aHIV/S), acute HIV infection without syphilis (aHIV), chronically coinfected syphilis/HIV+ patients on antiretroviral treatment (cHIV/S), and healthy controls (CTRL). Flow cytometry assessed PD-1, HLA-DR, and CD38 expression on various T-cell subsets (CD3+CD4+ (T_{CD4}), CD3+CD8+ T-cells (T_{CD8})), memory T-cells (CD4+CD45RO+ memory T-cells (T_M), CCR7±CD27± memory subsets (T_{CM}: T central memory, T_{EM}: T effector memory, T_{TM}: T transitional memory), antigen-naive cells (CD4+CD45RO- T_N)), regulatory T-cells (CD49b+LAG-3+ (TR1)), and plasmacytoid dendritic cells (BDCA2+CD123+ (pDC)). IFN gamma (IFNγ) levels were measured in plasma using Simoa technology (Quanterix).

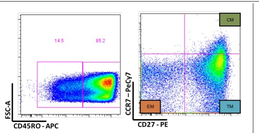
A Study cohorts



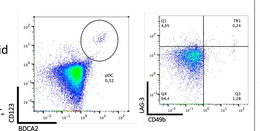
B Clinical characteristics

	K1 aHIV/S	K2 aHIV	K3 cHIV/S	K4 CTRL
sex (%)				
male	100	100	100	40
female	0	0	0	60
age (years)	36 (25-40)	35 (28-40)	45 (41-59)	37 (28-59)
HIV positive (years)	1 (0.5-2)	2.5 (2-5)	14 (10-19)	n.a.
cART (years)	n.a.	n.a.	8 (5-11)	n.a.
CD4 (cells/µl)	403 (288-599)	515 (285-638)	872 (477-974)	1230 (1020-1340)
CD4 (%)	24 (20-31)	29 (19-39)	35 (21-36)	35 (29-38)
CD4 nadir (cells/µl)	503 (369-590)	478 (285-638)	326 (258-454)	n.a.
Plasma viral load (M cps/ml	1.5 (0.3-10)	0.9 (0.5-10)	n.a.	n.a.

C Gating strategy CD4+ T-cells (T_{CD4}) and memory T-cells (T_M)



D Gating strategy plasmacytoid dendritic cells (pDC) and type1 regulatory T-cells (TR1)



E Statistical analysis

Distribution *(%)*	aHIV/S acute HIV syphilis co-infection	aHIV acute HIV	cHIV/S chronic HIV syphilis co-infection	CTRL healthy controls	
T_{CD4}: CD4 positive T-cells (T_{CD4})	12.9 (3.59-18.7)	16.3 (13.6-20.7)	25.1 (20.2-33.1)	35.4 (26.4-39.2)	<0.0001
T_{CD8}: CD8 positive T-cells	10.9 (6.81-18.3)	27.2 (22.8-35.1)	22.8 (16.9-25.8)	15.0 (13.0-18.4)	<0.0001
T_N: antigen-naive T-cells (T_N)	45.3 (35.1-56.8)	32.4 (28.1-38.4)	34.6 (22.1-39.5)	45.9 (36.8-57.5)	.270
T_M: memory T-cells (T_M)	54.7 (46.9-64.9)	67.6 (54.4-72.8)	65.4 (51.9-71.6)	54.1 (48.7-64.3)	.0423
T_{CM}: central memory T-cells	59.8 (39.1-64.4)	38.7 (35.1-42.1)	34.9 (31.6-40.6)	38.1 (31.2-53.2)	.0005
T_{EM}: effector memory T-cells	10.8 (9.28-28.9)	26.1 (20.6-36.0)	23.9 (21.4-33.3)	15.8 (10.9-28.0)	.0186
T_{TM}: transitional memory T-cells	18.0 (13.4-21.4)	24.5 (22.1-27.1)	34.6 (22.5-41.2)	31.2 (28.0-36.4)	<0.0001
pDC: plasmacytoid dendritic cells	0.41 (0.32-0.52)	0.24 (0.16-0.65)	0.42 (0.2-0.46)	1.06 (0.85-1.32)	<0.0001
TR1: type 1 regulatory T-cells	2.97 (2.12-3.59)	2.89 (2.52-3.34)	4.13 (2.98-4.97)	5.94 (3.27-8.09)	.0365
Expression *(gMFI)*					
CD38 on T_{CD4}	3.0 (2.7-3.9)	4.5 (3.8-6.9)	2.3 (2.1-2.8)	2.8 (1.9-3.0)	<0.0001
CD38 on T_{CD8}	14.2 (6.3-23.3)	16.4 (9.4-33.7)	2.4 (2.2-3.3)	1.7 (1.4-1.9)	<0.0001
PD-1 on TR1	1.02 (1.01-1.05)	1.01 (1.01-1.03)	1.01 (1.0-1.02)	1.0 (1.0-1.0)	.0003

Figure 1
A Study cohorts **B** Clinical characteristics **C** Gating strategy of CD4+CD45RO+ memory T-cells (T_M) and subpopulations (central (T_{CM}), effector (T_{EM}) and transitional (T_{TM})) **D** Gating strategy of CD123+BDCA2+ plasmacytoid dendritic cells (pDC) and CD49b+LAG-3+ type 1 regulatory T-cells (TR1) **E** Statistical analysis. All values are represented as the median with interquartile range (IQR) unless otherwise stated. For statistical analysis, non-parametric Kruskal-Wallis tests with Dunn's multiple comparisons or parametric ANOVA with post-hoc analysis were used, respectively. p <0.05 shows statistical significance.
HIV human immunodeficiency virus; aHIV, acute HIV infection; cHIV, chronic HIV infection; S, syphilis; CTRL, healthy controls; IQR, interquartile range; µl, microliter; M, million; cps, copies; ml, milliliter; n.a., not applicable.

The impaired T-cell function, activation, and exhaustion observed in acute HIV/syphilis coinfection result in reduced control of HIV replication.

Results

In patients with acute HIV/syphilis co-infection, the frequency of T_{CD4} (p<0.0001) and T_{CD8} (p<0.0001) cells was significantly lower compared to other groups, while CD38 expression was highest (p <0.0001). Simultaneously, the frequency of pDC (p <0.0001) and TR1 (p=0.0365) decreased significantly in aHIV/S, aHIV, and cHIV/S groups compared to CTRL. Interestingly, T central memory cells were significantly increased in aHIV/S (p=0.0005). Noteworthy findings included the highest PD-1 expression on type 1 regulatory T cells was observed in acutely HIV-coinfected patients (p=0.0003), and it correlated positively with viral plasma load (r=0.6 p=0.0314). Furthermore, the highest levels of IFNγ were found in acutely HIV-coinfected patients.

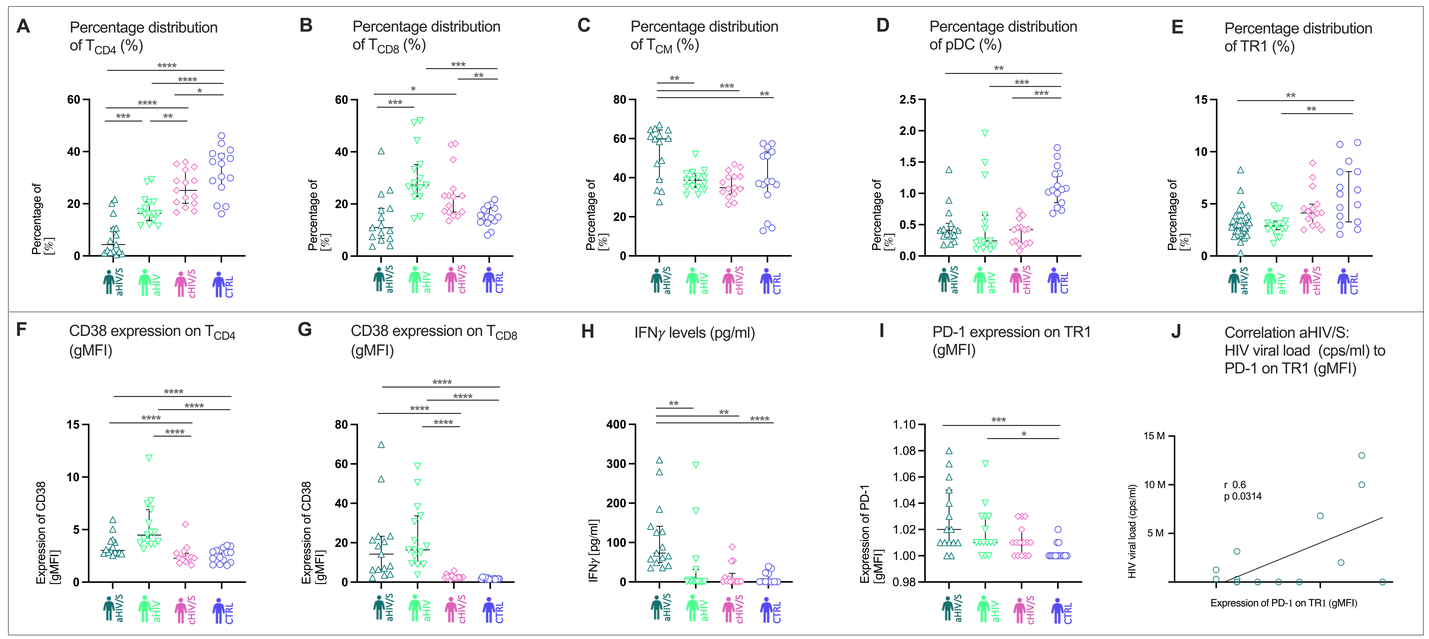


Figure 2 Preliminary results of T-cell distribution, activation and exhaustion by cohort.
Percentage distribution (%) of **A** T_{CD4} **B** T_{CD8} **C** T_{CM} **D** pDC **E** TR1 in all cohorts, respectively. CD38 expression (gMFI) on **F** T_{CD4} and **G** T_{CD8}. **H** Interferon gamma levels (IFNγ, pg/ml) of study cohorts **I** PD-1 expression (gMFI) on TR1 **J** Correlation of plasma viral load (cps/ml) and PD-1 on TR1 in acutely HIV/syphilis-coinfected patients. In patients acutely coinfected with HIV/syphilis, the frequency of T_{CD4} and T_{CD8} was lowest (T_{CD4}: aHIV/S < aHIV < cHIV/S < CTRL; T_{CD8}: aHIV/S < CTRL < aHIV < cHIV/S), while the expression of CD38 was highest. The frequency of pDC and TR1 decreased significantly in the blood of aHIV/S, aHIV and cHIV/S compared to CTRL. Highest PD-1 expression on TR1 was observed from acutely HIV-coinfected patients, which correlated positively with viral plasma load (r0.6; p0.0314).

Data information: All values are represented as the median with interquartile range (IQR) unless otherwise stated. For statistical analysis, non-parametric Kruskal-Wallis tests with Dunn's multiple comparisons or parametric ANOVA with post-hoc analysis were used, respectively. Spearman's r was used to describe non-parametric correlations. p <0.05 shows statistical significance: *p ≤0.05, **p ≤0.01, ***p ≤0.001, ****p ≤0.0001.
T_{CD4}, CD4 positive T-cells; T_{CD8}, CD8 positive T-cells; TCM, T central memory T-cells; pDC, plasmacytoid dendritic cells; TR1, type 1 regulatory T-cells; PD-1, programmed-death-1; HIV, human immunodeficiency virus; aHIV, acute HIV infection; cHIV, chronic HIV infection; S, syphilis; CTRL, healthy controls; gMFI, geometric mean fluorescence intensity; %, percentage; pg, picogram; ml, milliliter.

Conclusions

The impaired T-cell function, activation, and exhaustion observed in acute HIV/syphilis coinfection result in reduced control of HIV replication. The interplay between these two infections underscores the necessity for longitudinal investigations into patients coinfected with HIV and syphilis, particularly in the post-initiation phase of antiretroviral treatment. Such studies are imperative for gaining insights into the enduring effects of this coinfection on the HIV reservoir, providing valuable information for optimizing therapeutic strategies and enhancing our understanding of the immunological dynamics in these co-affected individuals. Longitudinal studies of patients coinfected with HIV and syphilis, especially after initiating antiretroviral treatment, are warranted to investigate their effects on the HIV reservoir.

References

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