The Preservation of Thymic activity and the naïve T cell Compartment is a Hallmark of HIV-2 and early treated HIV-1 Individuals

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Background

Antiretroviral therapy (ART) markedly improved disease prognosis, however ART-treated subjects maintain some degree of immune activation eventually associated with co-morbidities. Importantly, HIV-2 infected individuals also feature significant immune activation, despite a relatively benign course and low to undetectable plasma viral load even in the absence of ART. To better understand the mechanisms underlying persistent immune activation, we investigated the correlation profile of immune parameters in circulating lymphocyte subsets of HIV-1 and HIV-2 infected individuals under effective ART. We specifically asked which parameters would allow the segregation of: 1) HIV-2 from HIV-1 infection; and 2) HIV-1 individuals who started ART during acute infection (early ART) and those who started during the chronic stage (late ART).

Aim

We investigated the correlation profile of high-dimensional immune parameters in the main circulating lymphocyte subsets of cohorts of HIV-1 and HIV-2 treated infected individuals, with effective ART as attested by undetectable plasma viral load, to better understand the mechanisms underlying chronic immune activation.

Conclusions

Remarkably, there was an overlap of the immune profile of early treated HIV-1 infected and seronegative cohorts, emphasizing the relevance of early ART.

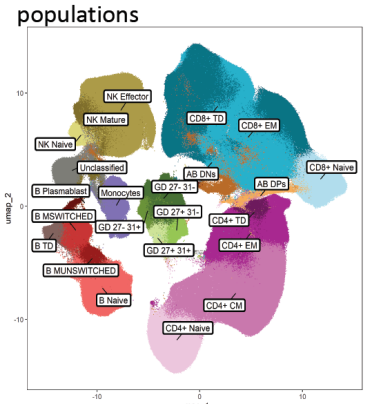
B cell clusters mainly contribute to the segregation of HIV-2 infected individuals, which may be related to the impact of the prolonged HIV-2 disease in the secondary lymphoid organs.

Regarding αβ T cell clusters these were the main segregators of HIV-1 infected patients treated early versus later. Strikingly, the CD31+ naïve CD4+ T cell cluster, which includes the recently produced T cell in the thymus (recent thymic emigrants, RTEs), distinguishes the three HIV infected cohorts, indicating that HIV-2 and early treated HIV-1 patients better preserve naïve CD4+ T cell homeostasis. Since all individuals had undetectable viremia and comparable levels of immune-activation parameters, it is likely that HIV-2 infection and early HIV-1 treatment are associated with a better preserved thymic activity and / or proliferative capacity and survival of naïve CD4 T cells. This is observed despite HIV-2 individuals being older.

Identifying underlying mechanisms may help counteracting deleterious effects of the persistent inflammation in patients under ART.

Results

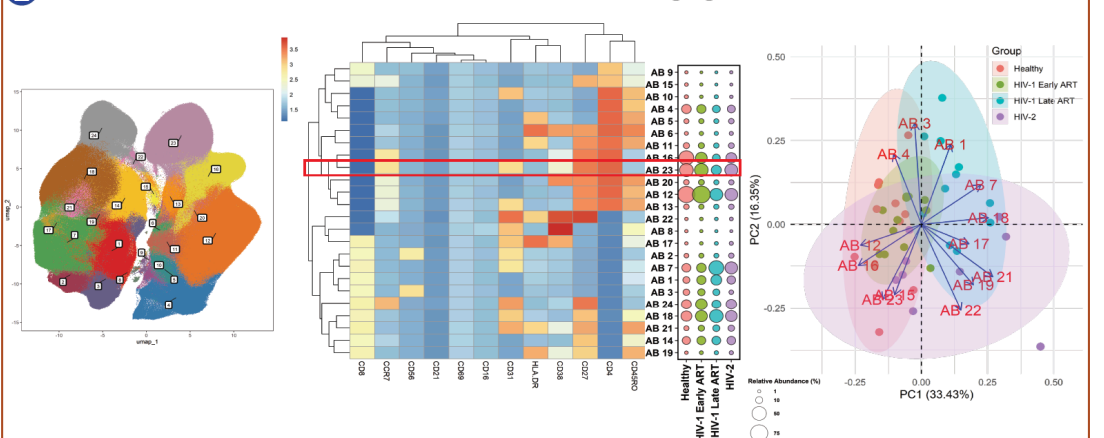
1 Selected markers allows the identification of the main populations



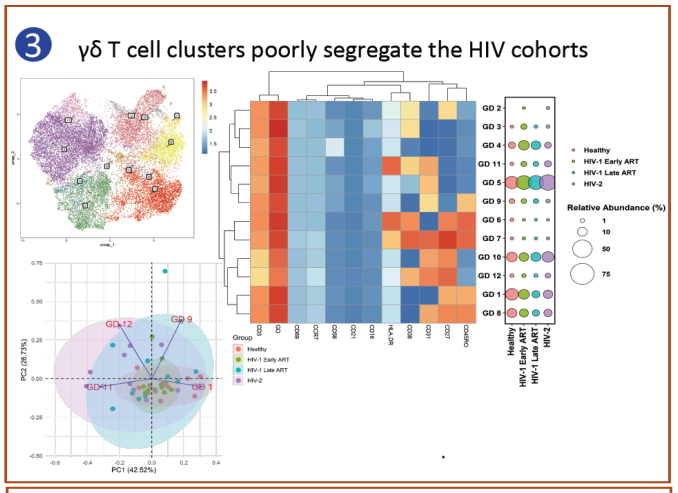
UMAP was used to embed the concadenated data of all patients into 2D space. Flow SOM clusters were then annotated into the major lymphoid populations based on marker expression

NK - Natural Killers cells
B - B cells
AB - αβ cells
GD - γδ T cells
MSWITCHED - Memory Switched
MUNSWITCHED - Memory Unswitched
TD - Terminal Differentiated
EM - Effector Memory
CM - Central Memory
DNs - Double Negatives
DPs - Double Positives

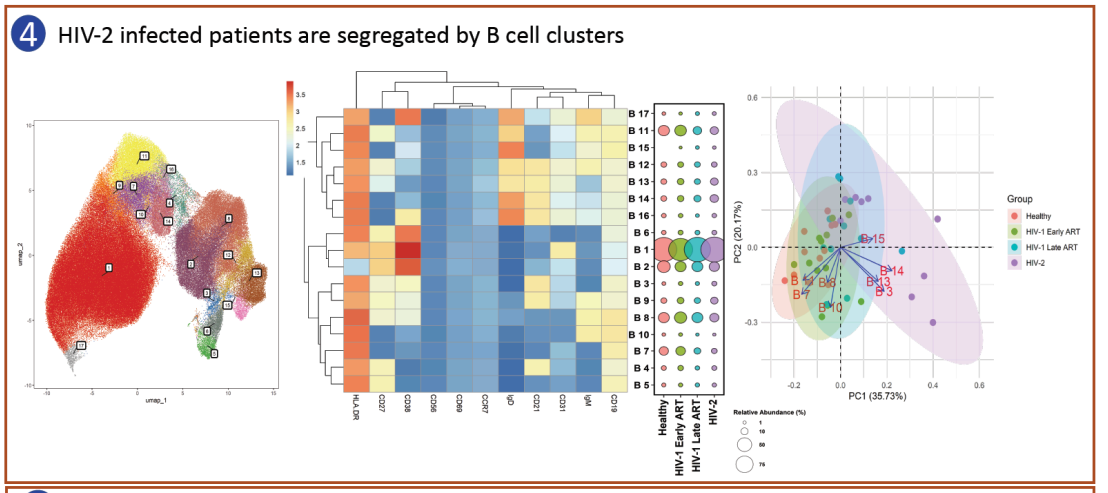
2 The CD31+ naive CD4+ T cell cluster is determinant in the segregation of the 3 HIV infected cohorts



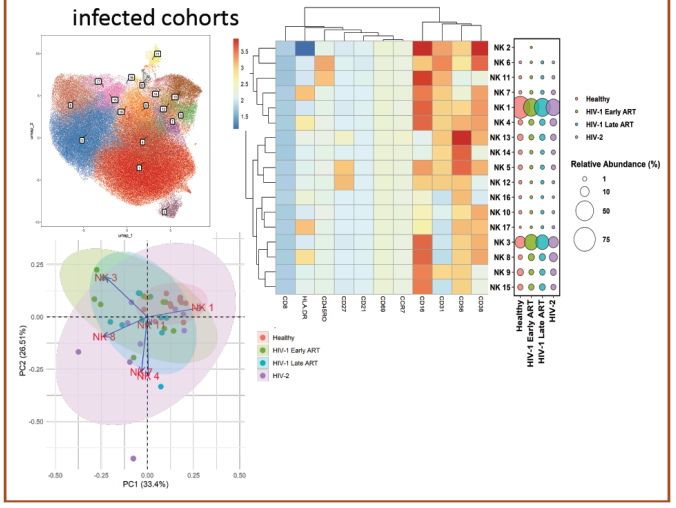
3 γδ T cell clusters poorly segregate the HIV cohorts



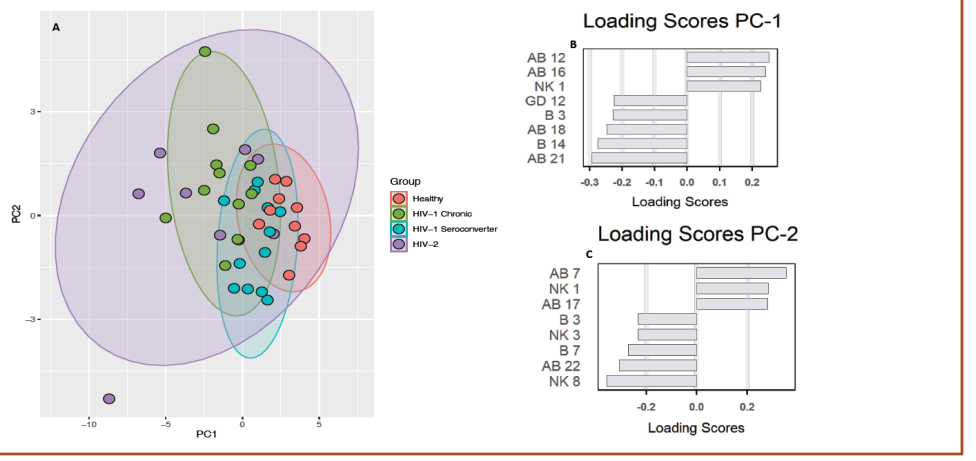
4 HIV-2 infected patients are segregated by B cell clusters



5 NK clusters mainly distinguish seronegative form HIV infected cohorts



6 Combined analysis of all the identified significant clusters



Patients and Methods

To profile chronic immune activation, we enrolled patients who started ART while seroconverting (early-ART, n=12), late-ART HIV-1 infected individuals (n=11), HIV-2 infected individuals (n=9) along with seronegative individuals (n=11). We stained 5 million circulating leukocytes in whole blood, with a panel of combined 18 markers allowing the discrimination of cell differentiation stages and levels of activation of CD4+ and CD8+ αβ and γδ T cells, NK and B cells by spectral flow cytometry. For a clear and comprehensive visualization of these populations and their respective phenotypes, we applied dimension reduction and clustering algorithms. We then generated correlograms and PCA analysis to uncover patterns of immune activation, integrating all main lymphocyte subsets and correlate the resulting profile with clinical parameters.

Table 1. Clinical and epidemiological data

Clinical variables	HIV-1 early ART	HIV-1 late ART	HIV-2	Healthy
N (Male/Female)	12 (9/2)	11 (3/8)	9 (2/7)	11 (4/6)
Age, years	43 (27-75)	51 (25-73)	61 (28-76)	54 (34-62)
Genetic Background	Latino - 25%	Latino – 9.09%	Latino – 0% *	Latino - 0%
	European 66.(6)%	European - 54.54%	European 22.(2)%	European - 100%
	African – 8.(3)%	African – 27.27%	African – 77.(7)%	African – 0%
Start of ART, years	2015 (2011-2019)	2011 (1997-2021)	2017 (2001-2021)	NA
Years on ART	6.5 (3-11)	11 (1-25)	5 (1-21)	NA
CD4+ T cells/µl	754 (361-1684)	421 (242-708)	426 (104-116)	
CD8+ T cells/µl	450 (198-789)	839 (554-1351)	501 (362-1569)	
CD4/CD8 ratio	1.57 (1.07-3.30)	0.45 (0.30-0.99)	0.89 (0.21-1.99)	

Data shown as median (interquartile range). NA- Not Applicable.

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