First-line cART regimen impacts the course of CD8+ T-cell counts in HIV-infected patients that achieve sustained undetectable viral load.

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First-line cART regimen impacts the course of CD8+ T-cell counts in HIV-infected patients that achieve sustained undetectable viral load.

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Abstract

The aim of the study was to investigate the impact of first-line combined antiretroviral therapy (cART) regimen on the course of CD8+ T-cell counts in human immunodeficiency virus (HIV)-infected patients.

A retrospective observational study conducted on the French DAT'AIDS Cohort of HIV-infected patients.

We selected 605 patients initiating a first-line cART between 2002 and 2009, and which achieved a sustained undetectable HIV plasma viral load (pVL) for at least 12 months without cART modification. The evolution of CD8+ T-cell counts according to cART regimen was assessed.

CD8+ T-cell counts were assessed in 572 patients treated with 2NRTIs+1PI/r (n= 297), 2NRTIs+1NNRTI (n= 207) and 3NRTIs (n= 68). In multivariate analysis, after 12 months of follow-up, the 3NRTIs regimen was associated with a significantly smaller decrease of CD8+ T-cell count compared with NNRTI-containing regimens (–10.2 cells/μL in 3NRTIs vs –105.1 cells/μL; P=0.02) but not compared with PI-containing regimens (10.2 vs –60.9 cells/μL; P=0.21). After 24 months, the 3NRTIs regimen was associated with a smaller decrease of CD8+ T-cell count and % compared with PI/r- and NNRTI-containing regimens (0.2 in 3NRTIs vs –9.9 with PI/r-regimens, P=0.001, and vs –11.1 with NNRTI-regimens, $p < 0.0001$). A focus analysis on 11 patients treated with an INSTI-containing cART regimen during the study period showed after 12 months of follow-up, a median decrease of CD8+ T-cell count of –155 [inter quartile range: –302; –22] cells/μL.

Our data highlight the fact that cART regimens have differential effects on CD8 pool down regulation.

Abbreviations: cART = combined antiretroviral therapy, HIV = human immunodeficiency virus, INSTI = integrase strand transfer inhibitor, NNRTI = non-nucleotide reverse transcriptase inhibitor, NRTI = nucleotide reverse transcriptase inhibitor, PI/r = Ritonavir-boosted protease inhibitor, pVL = plasma viral load.

Keywords: CD4:CD8 ratio, CD8, first-line cart, non nucleotide reverse transcriptase inhibitors

1. Introduction

HIV infection leads to perturbations of peripheral T-cell homeostasis with a gradual depletion of CD4+ T-cells combined with an expansion of the CD8+ T-cell compartment. After initiating combined antiretroviral therapy (cART), most HIV1-infected patients achieve sustained undetectable HIV plasma viral load (HIV-pVL) and restoration of the CD4+ T-cell compartment,[1] with an extent that depends on the level of CD4+ T-cell counts at the time of cART initiation.[2–6] In contrast, most

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patients fail to normalize CD8+ T-cell count despite long term cART, and this resilience of high $CD8^+$ T-cell counts was recently linked to increased non-AIDS-related mortality.[7] Previous studies have reported that the nature of antiretroviral drugs chosen for cART regimen affected CD4 recovery,[8,9] while the impact of cART choice on the dynamics of $CD8^+$ T-cells remains elusive.

The aim of this study was to determine the impact of cART regimen on the course of $CD8^+$ T-cell counts in HIV-infected patients who achieved sustained undetectable HIV-pVL on first-line cART.

2. Methods

2.1. Patient & Data collection

"The Dat'AIDS Cohort" is a multicenter cohort of 26959 HIV infected patients followed in fifteen major French HIV care centers using the NADIS® electronic medical record (Fedialis Medica, Marly le Roi, France).[10,11] In this cohort, patients' data are recorded during medical visits, and quality control is performed by automated checks during data capture, regular controls by clinical research associates, annual assessments and *ad hoc* processes prior to analysis.[10,11]

For this study, we selected HIV-1 infected and treatment-naive patients who initiated a first-line triple cART between Jan 2002 and Dec 2009 and who maintained an undetectable HIV plasma viral load (HIV-pVL) for at least 12 months without modification of cART regimen. Among them, we selected patients treated with a backbone of 2NRTIs combined with a ritonavir-boosted protease inhibitor (PI/r), or a non-nucleoside reverse transcriptase inhibitor (NNRTI), or a third NRTI. In addition, we analyzed separately patients treated with Integrase Strand Transfer Inhibitor (INSTI)-containing regimens, whatever the drug combination. All of these latter patients received raltegravir.

2.2. Outcome variables

Data on T-cell immunophenotyping ($CD4^+$, $CD8^+$ T-cell counts and CD4:CD8 ratio) were collected at the time of cART initiation (baseline), at the time of the first undetectable HIV-pVL (D0), and then after 12 (M12) and 24 months (M24) of follow-up with a sustained undetectable HIV-pVL without modification of cART regimen.

For T-cell immunophenotyping, a standardized procedure was performed on each site, briefly, fresh EDTA-whole blood (100 μL) was incubated with combinations of fluorochrome-conjugated monoclonal antibodies specific for CD3, CD4 and CD8 (Beckman Coulter) and analyzed using a FC500 cytometer (Beckman Coulter). Normal range of $CD4^+$ and $CD8^+$ T-cell counts were established at 500 to 1200 cells/μL and 300 to 830 cells/μL, respectively according to Reichert et al.[12] Plasma HIV-RNA was quantified by successive standardized assays including Roche Cobas HIV-1 monitor, Roche Cobas Ampliprep/Cobas Taqman HIV-1 v.2 test, and Abbott RealTime HIV-1. Since the threshold value of HIV-RNA was not the same through the study period depending on the techniques available at each site (<20 copies/mL, < 50 copies/mL or <40 copies/mL), the corresponding threshold for each period was considered.

2.3. Covariates

At baseline were collected the following potential confounders: sex, age, duration of HIV exposure since diagnosis, transmission risk group, CDC stage, cytomegalovirus (Anti-cytomegalovirus IgG positive) and hepatitis C (hepatitis C virus (HCV)-RNA positive) co-infections, CD4 nadir, HIV-pVL at baseline, duration of treatment and delay to HIV suppression on cART.

2.4. Ethic statement

All subjects provided written informed consent for the use of their medical records on NADIS. This electronic medical record was approved by the French *Commission Nationale Informatique et Liberté* (Registration number: 2001/762876/nadiscnil.doc). This study was carried out in compliance with the International guidelines for human research protection as Declaration of Helsinki and International Conference on Harmonization in Good Clinical Practice (ICH-GCP).

2.5. Statistical Analysis

All analyses were performed on SPSS Advanced Statistics 20 (IBM Corp) and SAS 9.4. Patients' characteristics were described at baseline according to $CD8^+$ T-cell counts and cART regimen. Categories of patients according to $CD8^+$ T-cell counts (low < 300 cells/μL; normal 300–830 cells/μL and high > 830 cells/μL) were defined after Reichert et al.[12] Median absolute counts and percentage of $CD4^+$, $CD8^+$ T-cells and CD4:CD8 ratio as well as their variations (Delta, Δ) were compared between baseline and D0, and then between D0 and M12, D0 and M24, according to cART regimen (2NRTIs+IPI/r vs 2NRTIs+1NNRTI vs 3NRTIs). Factors associated with $CD8^+$ T-cell count and CD4:CD8 ratio were identified at each time in bivariate analysis using Kruskal–Wallis tests for qualitative variables, and Chi-2 test or simple linear regression for quantitative variables. Evolution of $CD8^+$ T-cell count and CD4:CD8 ratio at M12 and M24 by cART regimen were then studied using multiple linear regression models including the potential confounders known. The value at D0 of each outcome tested was forced in the models. Missing data were considered as such and thus, were not included in the analyses, were not replaced or extrapolated.

3. Results

Among the 5688 HIV-infected patients who initiated a first-line triple antiretroviral treatment during the study period, 2074 had cART regimen unchanged for at least 12 months, of whom 830 patients had an undetectable HIV-pVL at all assessments, and 605 patients had data on HIV-pVL, $CD4^+$ and $CD8^+$ T-cell counts available at the time of cART initiation (Supplementary Figure 1: flowchart, http://links.lww.com/MD/B358). Hence, 605 patients composed the study cohort, which had a median time of cART exposure of 32.3 (23.8–43.7) months.

3.1. Patients' characteristics at baseline according to $CD8^+$ T-cell count

At baseline, $CD8^+$ T-cell counts were high (> 830 cells/μL) in 268 (44.3%) patients and low (< 300 cells/μL) in 38 (6%) patients. After cART initiation, they were respectively 47.7% and 3.1% at D0 (viral suppression), 39.8% and 3.7% after 12 months, 43.7% and 4% after 24 months under cART and the difference for each categories was not significant. Table 1 shows the characteristics of patients distributed by ranges of $CD8^+$ T-cell count at the time of cART initiation. According to this classification, patients did not differ in age, sex, HIV transmission risk group, HCV

Table 1

Patients' characteristics according to CD8+ T-cell count at baseline.

	CD8 (cells/μL)			
	< 300 (n = 38)	300–830 (n = 299)	> 830 (n = 268)	*P*
Males	26 (68.4%)	213 (71.2%)	202 (75.4%)	0.442
Age, y	45.5 (38.5; 54.2)	42.0 (36.0; 51.0)	45.0 (38.0; 51.0)	0.065
Transmission risk group				
Heterosexual	19 (50.0%)	137 (45.8%)	126 (47.0%)	0.196
MSM	11 (28.9%)	128 (42.8%)	103 (38.4%)	
IV drug user	2 (5.3%)	7 (2.3%)	16 (6.0%)	
Others	3 (7.9%)	8 (2.7%)	7 (2.6%)	
Unknown	3 (7.9%)	19 (6.4%)	16 (6.0%)	
Follow-up of HIV infection, y	0.3 (0.1; 3.5)	0.9 (0.1; 3.1)	1.6 (0.3; 5.5)	<0.001
CDC stage C	19 (50.0%)	41 (13.7%)	28 (10.5%)	<0.001
CMV IgG+	27 (79.4%)	223 (89.2%)	194 (88.6%)	0.244
HCV-RNA+	3 (7.9%)	15 (5.1%)	16 (6.1%)	0.629
CD4 nadir, cells/μL	30 (12; 103)	210 (121; 269)	239 (177; 303)	<0.001
HIV-pVL, log cp/mL	5.2 (4.4; 5.6)	4.7 (4.3; 5.3)	4.7 (4.2; 5.2)	0.016
CD4+T-cells:				
(cells/μL)	46 (12; 108)	236 (133; 287)	279 (215; 350)	<0.001
(%)	10.2 (4.0; 20.7)	19.7 (12.7; 24.0)	14.4 (11.0; 18.9)	<0.001
CD8+ T-cells:				
(cells/μL)	217 (135; 277)	616 (477; 722)	1169 (990; 1497)	<0.001
(%)	48.5 (30.7; 57.0)	52.6 (46.0; 62.0)	66.0 (59.8; 73.3)	<0.001
CD4:CD8 ratio	0.20 (0.07; 0.58)	0.38 (0.21; 0.51)	0.22 (0.15; 0.29)	<0.001
CD4:CD8 ratio ≥1	5 (13%)	7 (2.3%)	2 (0.7%)	0.001

Data are n (%) or median (IQR).

c*ART* = combined antiretroviral therapy, CDC = centers for disease control and prevention (CDC) classification system, CMV = cytomegalovirus, HCV = hepatitis C virus, HIV-pVL = HIV plasma viral load, IQR = inter quartile range, IV = intravenous, MSM = men who have sex with men.

co-infection or prevalence of cytomegalovirus-positive serology. However, patients with $CD8^+$ T-cell count < 300 cells/μL had significantly higher HIV-pVL, lower $CD4^+$ T-cell values and lower CD4 nadir. In contrast, median duration of HIV follow-up was significantly higher in patients with $CD8^+$ T-cell count > 830 cells/μL. Median CD4:CD8 ratio was significantly higher in patients with $CD8^+$ T-cell counts within the normal range, but the fraction of patients with CD4:CD8 ratio ≥ 1 was significantly higher in patients with $CD8^+$ T-cell counts < 300 cells/μL.

3.2. Patients' characteristics at baseline according to cART regimen

Overall, the impact of cART regimen on $CD8^+$ T-cell counts was assessed on patients receiving 1 of these 3 cART regimens: 2NRTIs+1PI/r (n= 297), 2NRTIs+1NNRTI (n= 207) and 3NRTIs (n= 68). Characteristics of patients by cART regimens are reported in Table 2 and detailed antiretroviral regimens for each group are described in supplementary Table 1, http://links.lww.com/MD/B358. Eleven patients receiving an INSTI-containing cART regimen were analyzed separately. Twenty-two patients receiving other cART combinations were excluded.

At baseline, patients receiving 3NRTIs differed significantly from those receiving 2NRTIs+1PI/r or 2NRTIs+1NNRTI by their longer median duration of HIV infection follow-up, shorter delay to HIV suppression after cART initiation, lower HIV-pVL, higher CD4 nadir and median CD4:CD8 ratio. Median $CD4^+$ and $CD8^+$ T-cell counts, and CD4:CD8 ratio significantly differed between the 3 groups but the proportion of patients with a CD4:CD8 ratio ≥1 was similar at baseline. At the time of the first undetectable HIV-pVL (D0), median $CD4^+$ T-cell count was significantly higher in the 2NRTIs+1NNRTI group, while median $CD8^+$ T-cell count was no different between groups either in absolute value or in percentage. Median CD4:CD8 ratio was similar between the groups, but the proportion of patients with a CD4:CD8 ratio ≥1 was significantly lower in the 3NRTIs group.

3.3. Evolution of $CD8^+$ T-cell counts according to cART regimen after viral suppression

3.3.1. At M12. Twelve months after the first undetectable HIV-pVL, $CD8^+$ T-cell counts were available for 505 patients (2NRTIs +1PI/r group: n= 262; 2NRTIs+1NNRTI group: n= 183; 3NRTIs group: n=60). Median $CD8^+$ T-cell counts were similar in all groups after 12 months in absolute value and percentage, as were $CD4^+$ T-cell counts (Fig. 1A and B). No significant differences were observed between the groups regarding the distribution of patients by $CD8^+$ T-cell count (< 300; 300–830 or > 830 cells/μL, Fig. 2A–C), or the median CD4:CD8 ratio (Fig. 1C).

Regarding the variations between D0 and M12, patients receiving 3NRTIs displayed the smaller decrease of absolute $CD8^+$ T-cell count (median variation (ΔCD8): −15 [inter quartile range (IQR): −173; 172] vs −43 [−212; 123] cells/μL in the 2NRTIs+1PI/r group and −128 [−275; 53] cells/μL in the 2NRTIs +1NNRTI group; *P*= 0.011). Median variation of CD4:CD8 ratio (ΔCD4:CD8) was significantly smaller in the 3NRTIs group (0.12 [0.04; 0.26]) compared to 2NRTIs+1NNRTI (0.16 [0.09; 0.31]; *P*=0.05), but this difference was not significant when compared to 2NRTIs+1PI/r (0.14 [0.07; 0.26]).

In multivariate analysis (Table 3), the 3NRTIs regimen was associated with a smaller decrease of CD8+ T-cell counts (ΔCD8) compared to 2NRTIs+1NNRTI regimens (*P*=0.02). No such significant difference was found between 3NRTIs and 2NRTIs +1PI/r regimens (*P*=0.21). Regarding ΔCD8%, the 3NRTIs regimen was associated with a smaller decrease of CD8% compared to PI/r- and NNRTI-containing regimens (*P*=0.007 and *P*=0.001, respectively). No significant difference of the variations of CD4:CD8 ratios were observed between the groups at M12 (Fig. 1C).

Table 2

Patients' characteristics by the cART regimen.

	2NRTIs+1PI/r n=297	2NRTIs+1NNRTI n=207	3NRTIs n=68	*P*
Male	211 (71.0%)	153 (73.9%)	53 (77.9%)	0.472
Age, y	43 (36; 50)	43 (36; 52)	42 (39; 51)	0.791
Transmission risk group:				
Heterosexual	145 (48.8%)	93 (44.9%)	33 (48.5%)	0.329
Homo/bisexual	115 (38.7%)	86 (41.5%)	23 (33.8%)	
IV drug use	13 (4.4%)	6 (2.9%)	6 (8.8%)	
Other	6 (2.2%)	8 (3.9%)	2 (2.9%)	
Unknown	18 (6.1%)	14 (6.8%)	4 (5.9%)	
CDC stage:				
A	201 (67.7%)	152 (73.4%)	49 (72.1%)	0.549
B	45 (15.2%)	29 (14.0%)	10 (14.7%)	
C	51 (17.2%)	25 (12.1%)	9 (13.2%)	
HCV-RNA +	18 (6.1%)	10 (4.8%)	6 (8.8%)	0.438
Anti-CMV IgG+	223 (75.1%)	150 (72.5%)	54 (79.4%)	0.505
CD4 Nadir (cells/μL)	209 (92; 275)	226 (163; 296)	239 (155; 274)	0.012
Duration of HIV follow-up, y	0.8 (0.1; 3.4)	1.1 (0.2; 4.3)	1.9 (0.2; 6.5)	0.014
Duration of cART, mo	31.2 (23.2; 41.0)	32.2 (24.1; 41.1)	48.5 (30.4; 64.1)	<0.001
Delay to HIV suppression, mo	3.2 (2.0; 5.4)	3.7 (2.0; 5.5)	2.0 (1.2; 4.4)	0.001
Pre-cART HIV-pVL (Log cp/mL)	4.9 (4.3; 5.4)	4.8 (4.4; 5.3)	4.4 (4.1; 4.8)	<0.001
Pre-cART CD4 T-cells:				
cells/μL	238 (105; 311)	261 (173; 324)	257 (191; 300)	0.015
%	16.0 (8.5; 22.3)	16.5 (12.0; 20.7)	17.0 (13.0; 21.5)	0.430
<200	117 (39.4%)	61 (29.5%)	21 (30.9%)	0.233
200–350	133 (44.8%)	116 (56.0%)	37 (54.4%)	
350–500	38 (12.8%)	26 (12.6%)	8 (11.8%)	
>500	9 (3.0%)	4 (1.9%)	2 (2.9%)	
Pre-cART $CD8^+$T-cells:				
cells/μL	748 (505; 1070)	856 (590; 1204)	733 (557; 1035)	0.014
%	59.0 (49.1; 70.0)	61.0 (52.0; 68.0)	59.7 (49.9; 67.5)	0.747
<300	23 (7.7%)	8 (3.9%)	4 (5.9%)	0.034
300–830	157 (52.9%)	90 (43.5%)	36 (52.9%)	
>830	117 (39.4%)	109 (52.7%)	28 (41.2%)	
Pre-cART CD4:CD8 ratio	0.26 (0.12; 0.41)	0.28 (0.18; 0.38)	0.32 (0.20; 0.45)	0.027
Pre-cART CD4:CD8 ≥1	8 (1.9%)	1 (0.5%)	2 (3%)	0.103
D0- $CD4^+$T-cells:				
cells/μL	354 (193; 484)	376 (285; 484)	338 (236; 412)	0.033
(%)	22.0 (13.0; 29.1)	23.0 (18.0; 28.8)	20.0 (16.3; 25.3)	0.074
D0- $CD8^+$T-cells:				
cells/μL	826 (561; 1158)	820 (622; 1133)	779 (574; 1005)	0.332
%	52.1 (43.0; 63.0)	52.0 (42.1; 61.0)	53.8 (46.7; 62.2)	0.665
D0-CD4:CD8 ratio	0.40 (0.22; 0.63)	0.44 (0.29; 0.62)	0.44 (0.28; 0.62)	0.147
D0-CD4:CD8 ≥1	12.7%	8.2%	5.8%	0.05

Data are n (%) or median (IQR). IQR= inter quartile range.

D0 correspond to the time of the first undetectable HIV plasma viral load.

c*ART* = combined antiretroviral therapy, CMV = cytomegalovirus, HCV = hepatitis C Virus, HIV-pVL = HIV plasma viral load, IV = intravenous, NNRTI = non-nucleoside reverse transcriptase inhibitors, NRTI = nucleoside reverse transcriptase inhibitors, PI/r = ritonavir-boosted protease inhibitors.

3.3.2. At M24. In total 240 patients achieved 24 months of follow-up without modification of cART regimens, of which 117, 85 and 38 patients received 2NRTIs+1PI/r, 2NRTIs+1NNRTI, and 3NRTIs regimens, respectively.

Median $CD4^+$ and $CD8^+$ T-cell counts were similar between the groups at M24 (Figs. 1A and Fig. 2B). Patients receiving 3NRTIs had significantly higher CD8% (48.4 [44.1; 58.8] % vs 43.0 [36.6; 52.7] % in 2NRTIs+1PI/r group and 42 [34.5; 50.3] % in the 2NRTIs+1NNRTI group; P=0.003). Median CD4:CD8 ratio was significantly lower in patients receiving 3NRTIs (P= 0.028, Fig. 1C) but the proportion of patients with a CD4:CD8 ratio ≥1 did not differ significantly between the groups (Fig. 1D).

Median ΔCD8% was significantly smaller in patients receiving 3NRTIs (–2.0 [–8.4; 5.3] %) compared to those receiving 2NRTIs+1PI/r and 2NRTIs+1NNRTI (-8.4 [–16.2; –3.9] % and 10.5 [–16.1; –6] %, respectively; P<0.001) and median ΔCD4: CD8 was significantly smaller in patients receiving 3NRTIs (0.1 [0.1; 0.3] %) compared to 2NRTIs+1PI/r and 2NRTIs+1NNRTI (0.3 [0.2; 0.4]% and 0.3 [0.2; 0.5]%; $P < 0.01$).

In multivariate analysis (Table 3, bottom), the 3NRTIs regimen was associated with a smaller decrease of CD8% compared to PI/r- and NNRTI-containing regimens (P=0.001 and $P < 0.0001$, respectively), and to a smaller increase of CD4:CD8 ratio (P= 0.10 and P=0.01, respectively, Table 3).

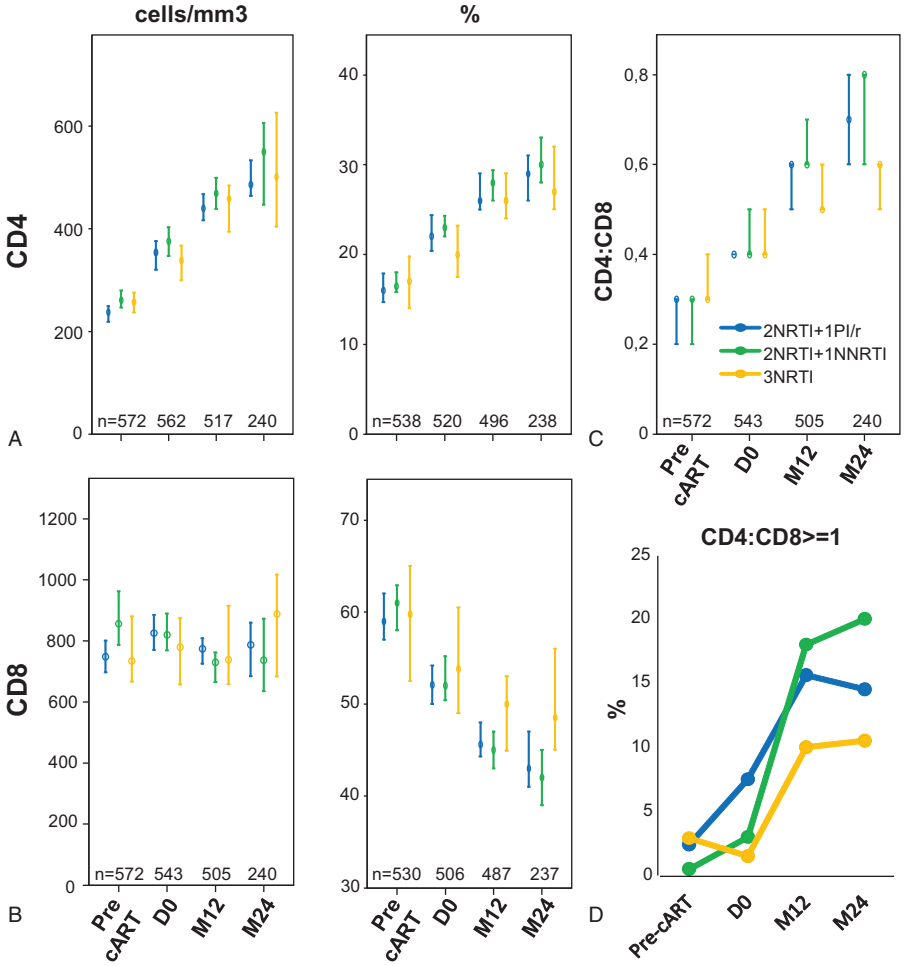

Figure 1. Comparative analysis of the impact of first-line cART regimen on T-cell compartments and CD4:CD8 ratio in HIV-infected patients. Graphs illustrate median [IQR] $CD4^+$ (A) and $CD8^+$ (B) T-cell counts (cells/μL) and percentages (%), and CD4:CD8 ratio (C) prior to cART initiation (Pre-cART), at the first undetectable HIV-pVL (D0) and after 12 (M12) or 24 (M24) months of HIV suppression, in patients receiving 2NRTIs+1PI/r, 2NRTIs+1NNRTI, or 3NRTIs as first-line cART. (D) Graph shows the fraction of patients (%) achieving normalization of CD4:CD8 ratio (≥ 1) according to first-line cART regimens. c*ART* = combined antiretroviral therapy, HIV = human immunodeficiency virus, n = number of observations, INSTI = integrase strand transfer inhibitor, NNRTI = non-nucleotide reverse transcriptase inhibitor, NRTI = nucleotide reverse transcriptase inhibitor, PI/r = Ritonavir-boosted protease inhibitor, pVL = plasma viral load.

3.4. Focus on patients receiving an Integrase Strand Transfer Inhibitor (INSTI)-containing regimen in first-line cART

Eleven patients of the study cohort received an INSTI-containing cART regimen. Most of patients were males (82%) with a median age of 43 [38; 53] year old and median time of HIV infection follow-up of 1.9 [1.2; 7.2] years. INSTI was combined to 2NRTIs for 8 patients and to 2NRTIs+1NNRTI in 3 cases. At baseline, median $CD4^+$ and $CD8^+$ T-cell counts were 340 [266; 397] cells/μL and 646 [520; 1161] cells/μL, respectively, and 36.4% of patients had $CD8^+$ T-cell count > 830 cells/μL. Median CD4:CD8 ratio was 0.48[0.27; 0.69] and < 1 in 90% of patients at baseline. At D0, median $CD4^+$ and $CD8^+$ T-cell counts had increased to reach 484 [366; 577] cells/μL and 835 [531; 1130] cells/μL, respectively. The proportion of patients with a $CD8^+$ T-cell count > 830 cells/μL dropped to 18.2%. Median CD4:CD8 ratio was 0.5 [0.41; 0.78], and CD4:CD8 ratio was < 1 in 82% of cases. All patients had their INSTI-containing first-line cART regimen unchanged after 12 months; they were only 2 patients after 24 months. After 12 months of viral suppression, median $CD4^+$ T-cell count was 541[409; 797] cells/μL with a median ΔCD4 of 133 [-26; 206] cells/μl, whereas median $CD8^+$ T-cell count decreased to 683 [539; 797] cells/μL with a median variation of −155 [−302; −22] cells/μL. The proportion of patients with $CD8^+$ T-cell count > 830 cells/μL was 20% (Fig. 2D). Conversely, median CD4:CD8 ratio improved to 0.7 [0.63; 1.02] and was ≥1 in 36.4% of patients after 12 months of viral suppression.

4. Discussion

This study performed on a large cohort of HIV-1-infected patients achieving sustained HIV suppression for at least 12 months without modification of cART regimen showed that: (i) the proportion of patients presenting with a high $CD8^+$ T-cell count remained stable despite a sustained HIV suppression for 24 months; (ii) compared to 2NRTIs+1PI/r and 2NRTIs+1NNRTI regimens, the combination of 3NRTIs was associated with the smaller decrease of $CD8^+$ T-cell counts after 24 months of viral suppression.

Our data are consistent with recent observations reporting the persistence of elevated $CD8^+$ T-cell counts after 10 years of cART.[7] In our cohort, 44% of patients presented with high $CD8^+$ T-cell counts (> 830 cells/μL) at the time of cART

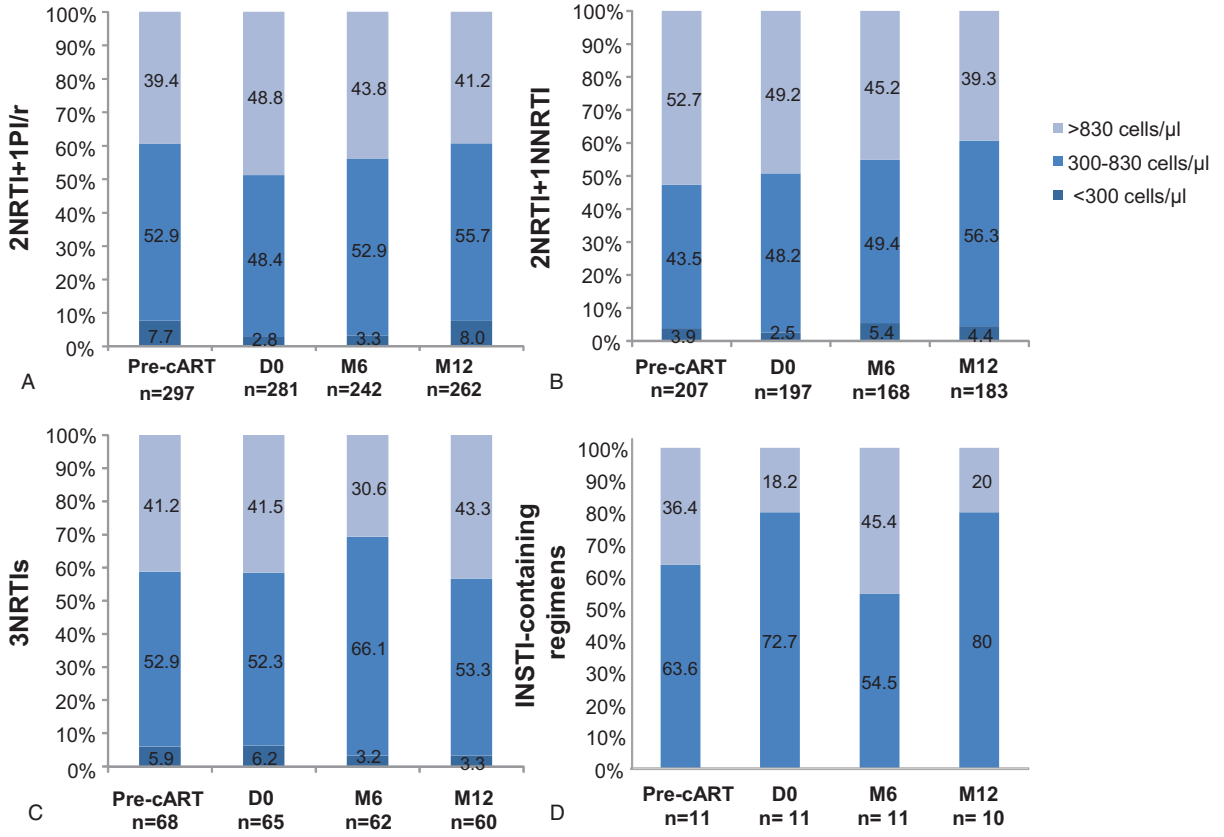

Figure 2. Distribution of patients by ranges of CD8+ T-cell counts according to first-line cART regimen. Stacked bars represent the proportions of patients with high (>830 cells/μL), normal (300–830 cells/μL), and low (< 300 cells/μL) $CD8^+$ T-cell counts among patients receiving 2NRTI+1PI/r (A), 2NRTI+1NNRTI (B), 3NRTIs (C), or in patients receiving an INSTI-containing regimen (D). c*ART* = combined antiretroviral therapy, HIV = human immunodeficiency virus, n = number of observations, INSTI = integrase strand transfer inhibitor, NNRTI = non-nucleotide reverse transcriptase inhibitor, NRTI = nucleotide reverse transcriptase inhibitor.

initiation. These patients had significantly longer HIV follow-up, higher CD4 nadir and higher $CD4^+$ T-cell counts, but their HIV-pVL did not differ from patients with normal or low $CD8^+$T-cell counts. Hence, a high HIV-pVL does not seem to be a pivotal factor for CD8 hyperlymphocytosis. The association between the duration of HIV follow-up and $CD8^+$ T-cell count was no longer significant after 6 months of HIV suppression regardless of cART regimens (data not shown). However, other factors such viral coinfections may affect the down regulation of the CD8 compartment upon viral suppression. Indeed, we have previously shown that HCV coinfection is associated with high $CD8^+$ T-cell counts in long-term HIV suppressors on cART.[13] Nevertheless, in this cohort the number of HCV coinfected patients was too small for being a confounder.

The persistence of high $CD8^+$ T-cell counts despite a durable control of HIV replication might be explained by a premature immune senescence of $CD8^+$ T-cells, as described for naïve HIV-infected patients.[14] Indeed, senescent $CD8^+$ T-cells accumulate over time, which is likely to counter the normalization of CD4:CD8 ratio.[15] Unfortunately, immune senescence could not be evaluated in our study. However, evidences of the association between expanded CD8 immunosenescent and/or activated subpopulations and accelerated progression of HIV disease keep on accumulating.[16,17,18–21]

Up to date, the impact of cART regimen on immune recovery during clinical trials and observational cohorts was mostly evaluated in terms of $CD4^+$ T-cell recovery, while data on $CD8^+$ T-cells are scarce. Risk of non-AIDS related events and death was reported to correlate to high $CD8^+$ T-cell counts (>1500 cells/μL) in patients achieving sustained undetectable HIV-pVL.[7] Although CD4:CD8 ratio was shown to be a reliable predictor of the evolution of HIV-disease,[22–24] its improvement does not always correlate with recovery of the CD4 compartment. Altogether, these data and ours stress the need to consider not only the evolution of CD4:CD8 ratio but also $CD8^+$ T-cell count, in absolute value and percentage, for the evaluation of immune restoration under cART.

Interestingly, we did not observe a significant difference of the variation of the CD8 compartment between patients receiving PI/r and NNRTI-containing regimens. The small number of patients receiving an INSTI-containing cART regimen in our cohort did not allow us to compare this group to other. However, after 12 months of viral suppression, this subpopulation of patients receiving INSTI displayed the most important decrease of $CD8^+$ T-cell counts observed in this study and, moreover, the highest $CD8^+$ T-cell count at baseline (>830 cells/mL), the biggest drop after 12 months of HIV suppression. A recent study performed in 39 naive patients receiving a combination of raltegravir and TDF/FTC showed that CD4 recovery was associated with an early and transient increase of $CD8^+$ T-cell count, but was not different from baseline after 12 months of follow-up.[25]

In our study, 38 patients displayed low $CD8^+$ T-cell counts (<300 cells/μL) at cART initiation, which has previously been associated with bad prognosis.[7] According to Helleberg's study,

Table 3

Evolution at M12 and M24 of CD8 according to the cART regimen. Multivariate linear regression.

At M12	2NRTI+1PI/r N = 250	2NRTI+1NNRTIs N = 177	3NRTIs N = 58
Δ CD8-M12-D0			
Cells/mm^3 (SE)[†]	−60.9 (17.1)	−105.1 (20.3)[*]	−10.2 (36.1)
% (SE)[‡]	−6.8 (0.5)[**]	−7.6 (0.7)[**]	−2.7 (1.4)
ΔCD4:CD8ratioM12-D0 (SE)[§]	0.19 (0.01)	0.22 (0.02)	0.15 (0.03)
At M24	**N = 117**	**N = 85**	**N = 38**
Δ CD8-M24-D0			
Cells/mm^3 (SE)[‖]	−99.7 (31.5)	−121.0 (36.2)	−15.3 (55.8)
% (SE)[¶]	−9.9 (1.1)[***]	−11.1 (1.3)[***]	0.2 (2.3)
ΔCD4:CD8 ratio M24-D0 (SE)[#]	0.28 (0.02)	0.33 (0.03)[**]	0.20 (0.04)

NNRTI = non-nucleoside reverse transcriptase inhibitors, NRTI = nucleoside reverse transcriptase inhibitors, PI/r = ritonavir-boosted protease inhibitors, SE = standard error.

[†] Model adjusted for confounders: duration of HIV at treatment initiation, CD4 nadir, pre-cART HIV-pVL, CD8 at D0.

[‡] Model adjusted for confounders: duration of HIV at treatment initiation, duration of treatment, delay to HIV suppression, CD8% at D0.

[§] Model adjusted for confounders: CD4 nadir, pre-cART HIV-pVL, CD4/CD8 ratio at D0.

[‖] Model adjusted for confounders: CD4 nadir, duration of HIV at treatment initiation, duration of treatment, CD8 at D0.

[¶] Model adjusted for confounders: duration of treatment, CD8% at D0.

[#] Model adjusted for confounders: CD4 nadir, duration of treatment, delay to HIV suppression, CD4/CD8 ratio at D0.

[*] *P*-value <0.05.

[**] *P*-value <0.01.

[***] *P*-value <0.001; ref = 3NRTIs.

patients that display a CD8$^+$ T-cell count < 500 cells/μL a year after cART initiation, are at higher risk of mortality due to AIDS-related conditions. In our study, patients with low CD8+ T-cell count had higher HIV-pVL, lower CD4$^+$ T-cell values and lower CD4 nadir, which was also observed after 10 years of cART treatment in Helleberg's study. During the study period, the proportion of these patients remained stable, whatever the cART regimens. Unfortunately, this retrospective study did not allow us to evaluate morbidities and mortality rate.

One limitation of this study is that, despite the large number of patients of the Dat'AIDS cohort, the fraction of patients receiving an unchanged first-line cART regimen that could be analyzed after 24 months of viral suppression is rather small. Nevertheless, it should be considered that the rate of cART regimen modification and interruption is particularly high during the first year of cART, as observed in our study and other contemporary studies.[26] Secondly, due to the period of patient selection, the potential impact of last generation antiretroviral drugs currently recommended for cART initiation, like INSTI, was investigated in a very small number of patients. Finally, regarding the retrospective design of the study, only the potential confounders available in our database could be used in the multiple linear regression models. Thus, we cannot rule out the role of others factors than cART regimen, such as comorbidities, to explain the differences observed between the different drug combinations.

In conclusion, our study showed that the number of CD8$^+$ T-cells remains high despite a sustained control of HIV replication on first-line cART, and that cART regimen has an impact on the course of CD8$^+$ T-cells. NRTI-only regimens in naive patients may be associated with poor immune restoration, as reflected by a poor down regulation of the CD8 compartment, while patients receiving INSTI-containing regimens displayed encouraging results regarding CD8 down regulation. Hence, the immune balance leading to CD4:CD8 recovery does not only depend on the time of cART initiation but might also be conditioned by the effect of drug regimen on the evolution of CD8$^+$ T-cell count.

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