Telomere Length Predicts Immunological Recovery in Older HIV Patients Treated with cART

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Abstract

Background: Successful cART not always recover CD4 T-cells despite the effective control of HIV-1 replication. Telomeres are DNA-protein structur essential to chromosomal stability and replication. Telomere length (TL) is probably regulated by oxidative stress. Because little is known about the impact of TL on the immunorecovery we evaluated the relationship between TL and immunorecovery at 48 weeks after cART initiation in patients virologically suppressed.
Methods: CoRIS is an open, prospective, multi-centre cohort of HIV-positive subjects. We selected those on stable cART who achieved a VL<50 cop/mL after 48 weeks of their first cART. Immunorecovery was defined as an increase in CD4+ counts >100 cells/mL compared to baseline. Leukocyte TL was measured using a quantitative PCR technique and categorized into tertiles (<0.13 for the lowest, 0.13-0.27 for the middle and >0.27 for the highest tertile). Nitric oxide levels were quantified by determining nitrate/nitrite and S-nitroso compounds using an ozone chemiluminescence-based assay. Lipid peroxidation was measured by quantitation of thiobarbituric acid reactive substances. Multivariate logistic regression models were used to estimate odds ratios (OR) and its 95% confidence intervals (CI) for association between TL and the odds of achieving immunorecovery adjusting for sex, age at cART initiation (years) (<50 and ≥50) and CD4+ count at start of cART (cells/ml) (<200, ≥200). Additional adjustment for nitrosative and oxidative stress was also carried out. We assessed whether this association was different in patients aged <50 and ≥50 years at cART initiation by including interaction terms into models.
Results: Of 132 patients included, 86% were male and 81% <50 years at cART initiation. We failed to find an association between TL, and immunorecovery overall (p = 0.47) but found an interaction between TL and age at cART initiation (p = 0.008): while no association was seen in patients <50 years (p = 0.18), in those ≥50 the odds of achieving immunorecovery decreased with decreasing tertiles of TL (OR [95% CI] for middle and lowest tertiles was 0.50 [0.05-4.84] and 0.13 [0.03-0.63], respectively). Additional adjustment for nitrosative and oxidative stress did not change results.
Conclusion: Short TL in HIV-infected patients ≥50 is related to a lower immunorecovery in spite of the successful virological control.

1. Introduction

Background

- Successful combined antiretroviral therapy (cART) does not always result in complete CD4+ T-cell recovery despite the effective control of HIV-1 replication.
- Telomere dysregulation can lead to cell death, cell senescence, or abnormal cell proliferation. We hypothesized that the lack of T cell recovery may be related to telomere defects.

Objective

To evaluate the potential relationship between telomere length and immunological response at 48 weeks of cART initiation, and to assess the role of nitrosative and oxidative stress in HIV-1 immunorecovery following cART.

2. Methods

Patients & Methods

CoRIS is an open, prospective, multi-centre cohort of HIV-positive subjects that is linked to a BioBank. We selected those patients with an undetectable viral load (VL) (<50 cop. HIV RNA/ml) after 48 (± 6) wks. of their first cART and who had a serum sample available at 48 (± 12) wks.

- Leukocyte telomere length: Measured using a quantitative polymerase chain reaction technique. This technique compares signals from the telomere repeat copy number (T) to a single-copy gene 36B4 copy number (S), and allows calculation of a relative T/S ratio.
- Plasma nitrosative stress level: Nitric oxide production was indirectly quantified by determining nitrate/nitrite and S-nitroso compounds (NOx).
- Plasma oxidative stress level: Lipid peroxidation was measured by the thiobarbituric acid reactive substances (TBARS) method.

Statistical Methods

- The distribution of telomere length was divided into tertiles based on the entire study population. The cutoff values for telomere length were <0.13 for the lowest tertile (short), 0.13 – 0.27 for the middle tertile (medium) and >0.27 for the highest tertile (long).
- We calculated mean increases (95%) in CD4+ T cell counts at week 48 from cART initiation for patients with long, medium and short telomere length and used linear regression models to estimate mean differences. We also calculated the proportion of patients increasing at least 100 cells/ml at 48 weeks.
- Multivariate models were adjusted for other factors associated with immunological response.

3. Results

A total of 132 patients were included in the analyses, 86% male and 81% aged <50 years at cART initiation. Median (IQR) telomere length was 0.19 (0.09; 0.33) and median (IQR) NOx and TBARS were 15 (10.5; 20.6) and 25.4 (19.0; 30.7), respectively. There were no significant differences in socio-demographic and clinical characteristics of the patients according to tertiles of TL (**Table 1**).

Table 1. Socio-demographic & clinical characteristics of the patients at cART initiation according to Telomere Length tertiles

	Telomere length			
	Long	Medium	Short	p-value
	44 (33.3)	44 (33.3)	44 (33.3)	
Sex				0.18
Male	41 (93.2)	35 (79.5)	37 (84.1)	
Female	3 (6.8)	9 (20.5)	7 (15.9)	
Age (years)				0.52
Median (IQR)	40 (29 – 49)	37 (27 – 44)	38 (32 – 47)	0.45
<50	33 (75.0)	37 (84.1)	37 (84.1)	
≥50	11 (25.0)	7 (15.9)	7 (15.9)	
Transmission category				0.48
Homo/bisexual	25 (56.8)	25 (56.8)	22 (50.0)	
Injecting drug use	2 (4.6)	6 (13.6)	3 (6.8)	
Heterosexual	14 (31.8)	12 (27.3)	14 (31.8)	
Other/Unknown	3 (6.8)	1 (2.3)	5 (11.4)	
Educational level				0.22
None or Primary studies	18 (40.9)	20 (45.4)	13 (29.5)	
Secondary or University studies	25 (56.8)	19 (43.2)	26 (59.1)	
Unknown	1 (2.3)	5 (11.4)	5 (11.4)	
Country of origin				0.17
Europe, mainly Spain	33 (75.0)	35 (79.5)	37 (84.1)	
Latin America	10 (22.7)	9 (20.5)	4 (9.1)	
Africa	1 (2.3)	0	3 (6.8)	
CD4 cell counts (cells/ml)				
Median (IQR)	173 (69 – 265)	221 (102 – 290)	163 (77 – 245)	0.37
<200	26 (59.1)	20 (45.4)	28 (63.6)	0.45
200 - 350	15 (34.1)	18 (40.9)	12 (27.3)	
>350	3 (6.8)	6 (13.6)	4 (9.1)	
Viral load (copies/ml)				
Median (IQR)	81743 (34530 – 205000)	54903 (19500 – 126449)	58065 (11549 – 250569)	0.27
≤100000	24 (54.5)	31 (70.4)	24 (54.5)	0.21
>100000	19 (43.2)	12 (27.3)	16 (36.4)	
Unknown	1 (2.3)	1 (2.3)	4 (9.1)	
AIDS diagnosis				0.33
No	36 (81.8)	36 (81.8)	31 (74.5)	
Yes	8 (18.2)	8 (18.2)	13 (29.5)	
First cART employed				0.33
2 NRTIs + 1 NNRTIs	17 (38.6)	25 (56.8)	24 (54.5)	
2 NRTIs + 1 PIs	23 (52.3)	18 (40.9)	18 (40.9)	
Other	4 (9.1)	1 (2.3)	2 (4.6)	
NOx				
Median (IQR)	15.8 (11.3 – 21.4)	13.4 (10.1 – 19.5)	14.7 (9.6 – 19.8)	0.32
Lowest tertile	11 (27.5)	17 (41.5)	13 (35.1)	0.72
Middle tertile	15 (37.5)	11 (26.8)	13 (35.1)	
Highest tertile	14 (35.0)	13 (31.7)	11 (29.7)	
TBARS				
Median (IQR)	26.2 (18.9 – 31.1)	22.8 (16.3 – 28.3)	25.7 (21.8 – 30.0)	0.21
Lowest tertile	12 (30.0)	19 (46.3)	9 (24.3)	0.28
Middle tertile	13 (32.5)	12 (29.3)	15 (40.5)	
Highest tertile	15 (37.5)	10 (24.4)	13 (35.1)	

Note: cART= Combined antirretroviral regimens; NOx= S-nitroso compounds; NNRTIs= Non-nucleoside reverse transcriptase inhibitors; NRTIs= Nucleoside reverse transcriptase inhibitors; PIs= Protease inhibitors; TBARS= Thiobarbituric acid reactive substances.

Increases in CD4+ T cell counts at 48 weeks from cART initiation were greater in patients with long telomere length than in those with medium and short telomere length (p = 0.007). After adjustment for sex, age, CD4 T cell counts, and viral load at cART initiation in a multivariate linear regression model, differences in mean CD4 T cell count increases remained statistically significant (p = 0.02) (**Table 2**). Additional adjustment for NOx and TBARS did not change the results.

Differences in mean CD4+ T cell count increases according to tertiles of NOx were in the limit of statistical significance (p = 0.056). However, we failed to find an association between tertiles of TBARS and CD4 T cell count increase at 48 weeks from cART initiation (p = 0.37).

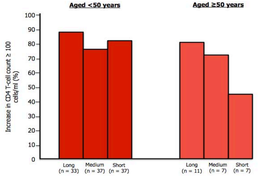
Table 2. Crude and adjusted analyses for the associations between telomere length and increase in CD4 T cell counts at 48 weeks after cART initiation

	Telomere length			
	Long	Medium	Short	p-value
Increase in CD4 T cell counts at 48 weeks from cART initiation				
Mean (95%) CD4 T cell counts increase, cells/ml	266 (236; 297)	189 (144; 235)	189 (114; 265)	
Crude mean difference (95% CI)	Ref.	-77.0 (-134.1; -19.6)	-76.8 (-139.0; -14.7)	0.007
Adjusted mean difference *	Ref.	-72.7 (-128.0; -17.4)	-82.2 (-155.3; -9.0)	0.02
Adjusted mean difference **	Ref.	-71.4 (-124.1; -18.7)	-88.9 (-153.1; -24.8)	0.01

* Adjusted for sex (male, female), age (<50, ≥50 years), CD4 T cell counts (<200, 200 – 350, >350 cells/ml) and viral load (≤100000, >100000 cop/ml) at cART initiation
** Additionally adjusted for tertiles of Nox and TBARS

When analyzing the association between telomere length and the increase of at least 100 cells/ml at 48 wks. from cART, a similar pattern was observed (OR [95% CI] for medium and short telomere length, compared to long telomere length, were 0.46 [0.10; 2.16] and 0.51 [0.10; 2.57]) (p = 0.61). We found an interaction between telomere length and age at cART initiation (P = 0.004): there was a suggestion of a higher impact of telomere length on the odds of increasing at least 100 cells/ml in patients aged ≥ 50 year s(OR [95% CI]) for medium and short telomere length, compared to long telomere length, were 0.48 [0.04; 5.10] and 0.10 [0.02; 0.43]) than in those aged <50 years (**Figure 1**).

Figure 1. Crude and adjusted analyses for the associations between telomere length and the increase of at least 100 cells/ml at 48 weeks from cART initiation, according to age at cART initiation



Increase in the CD4 T cell counts ≥100 cells/ml at 48 wks. from cART initiation	p-value			p-value			
Number (%)	29 (87.9)	29 (78.4)	32 (86.5)		9 (81.8)	5 (71.4)	3 (42.9)	
Crude OR (95% CI)	Ref.	0.50 (0.14; 1.84)	0.88 (0.15; 5.05)	0.38	Ref.	0.56 (0.05; 5.69)	0.17 (0.04; 0.79)	0.02
Adjusted OR (95% CI) *	Ref.	0.51 (0.10; 2.70)	0.95 (0.13; 6.97)	0.36	Ref.	0.48 (0.04; 5.10)	0.10 (0.02; 0.43)	<0.001
Adjusted OR (95% CI) **	Ref.	0.51 (0.09; 2.95)	0.76 (0.10; 5.69)	0.62	Ref.	0.61 (0.03; 13.52)	0.10 (0.02; 0.52)	0.02

* Adjusted for sex (male, female), age (<50, ≥50 years), CD4 T cell counts (<200, 200 – 350, >350 cells/ml) and viral load (≤100000, >100000 copies/ml) at cART initiation
** Additionally adjusted for tertiles of Nox and TBARS.

4.Conclusions

- A lower immunological response despite a successful virological response is associated with a shorter telomere lenght. This effect is not related with NOx or TBARS.
- Telomere length may be a useful marker of immunological response in HIV-infected patients ≥50 years.

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