Evaluation of T-cell immunosenescence markers in virologically suppressed people living with HIV aged over 60 years on BIC/FTC/TAF or DTG/3TC: the Collateral study

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Abstract

Introduction: Despite successful antiretroviral therapies (ART), people living with HIV (PLWH) continue to experience higher comorbidities than uninfected subjects, suggesting premature ageing of their immune system, often referred to as immunosenescence. A critical unanswered question is whether two drugs regimens (2DR) and three drugs regimens (3DR) have different effects on immunosenescence, especially in older subjects with a low CD4 nadir. Collateral is an ongoing cohort study evaluating cellular and plasma inflammatory markers and the onset of comorbidities in PLWH on BIC/FTC/TAF or DTG/3TC. We present an interim analysis evaluating markers of T-cell immunosenescence in participants over 60 years old.

Methods: Collateral is a multicenter on-going French prospective cohort study. Inclusion criteria were participants over 40 years of age, >10 years on ART, having HIV-1 RNA <50 c/ml and taking either BIC/FTC/TAF or DTG/3TC QD for at least 6 months. HBV or HCV co-infections and archived resistance to DTG, BIC or TAF were excluded. We analysed inflammatory markers in CD4+ and CD8+ naïve (N), central memory (CM), effector memory (EM), terminally differentiated effector memory (TEMRA) and senescent (SEN-KLRG1+ 57+) T-cells in participants aged over 60 years. HIV-negative participants were included as healthy control.

Results: We included 49 PLWH (BIC/FTC/TAF=23; DTG/3TC=26) and 5 HIV-negative participants. 49% (24/49) had a CD4 nadir <200/mm^3. PLWH had higher levels of immunosenescence markers than HIV-negative participants. Among PLWH with a CD4 nadir count <200 cells/mm^3, despite similar demographic and background parameters, we found significantly lower levels of senescent and TEMRA T-cells in the BIC/FTC/TAF group (Table 1).

Conclusions: In PLWH aged over 60 years with CD4 nadir <200 cells/mm^3, we found lower levels of immunosenescence markers on BIC/FTC/TAF compared to DTG/3TC. Our results suggest that past CD4 count should be considered as criteria for treatment decision choice between 2DR and 3DR in order to reduce risks of age-associated complications.

Introduction

Despite successful antiretroviral therapies (ART), people living with HIV (PWH) continue experiencing higher comorbidities than uninfected subjects [1]. Premature ageing, often referred to as immunosenescence, has been suggested among causes for explaining the elevated prevalence of non-AIDS defining events, such as cancer, frailty and metabolic, cardiovascular, bone and neurodegenerative disorders [2].

Indeed, the current leading hypothesis is that HIV infection is associated with a chronic inflammatory condition, leading to profound inflammatory damages to the architecture of tissues involved in T-cell regeneration and function [3]. The loss of T-cell homeostasis and the accumulation of senescent cells compromise the host's ability to control a wide range of potential pathogens [4].

Possible explanations for such chronic immune activation include persistent low-level HIV replication, microbial translocation and co-infection, despite successful control of HIV replication in plasma [5,6].

Collateral is an ongoing cohort study evaluating cellular and plasma inflammatory markers and the onset of comorbidities in PWH either on triple or dual ART. Indeed, although dual ART showed their non-inferiority in terms of virological control, data are still scarce about the inflammatory consequences of treatment simplification.

We present an interim analysis evaluating markers of T-cell immunosenescence in participants over 60 years old.

Methods

Study design and participants

Collateral is a multicentre on-going French prospective cohort study, performed in Nice and Cannes, France. Inclusion criteria were participants over 40 years of age or over 10 years on ART, on stable and successful ART with either Bictegravir/Emtricitabine/Tenofovir alafenamide (BIC/FTC/TAF) or Dolutegravir/Lamivudine (DTG/3TC) for at least 6 months.

Subjects with either hepatitis B or active hepatitis C were excluded, together with those having archived mutations to DTG, BIC or TAF.

Cellular markers of immune activation and senescence

For each PWH and controls, multi-color flow cytometry on freshly isolated PBMCs was performed, allowing the identification of the following CD4+ and CD8+ subsets: naïve (N), central memory (CM), effector memory (EM) and terminally differentiated effector memory (TEMRA) cells. Indeed, on the basis of the expression of two surface molecules, CD45RA and CCR7, T-cells can be divided into four subsets, with the following characteristics: CD45RA+CCR7+ for N cells, CD45RA-CCR7+ for CM, CD45RA-CCR7- for EM and CD45RA+CCR7- for TEMRA [7].
Moreover, according to previous works, cells double expressing the co-inhibitory receptor killer-cell lectin like receptor 1 (KLRG-1) and CD57 markers were considered as senescent [4, 8].

Statistical analysis
Main background characteristics of patients were collected, including age, years from HIV diagnosis, years on ART, CD4 cell count at inclusion, CD4/CD8 ratio, nadir CD4 and number of comorbid conditions.
Differences between dual and triple ART and risk factors associated were analysed using ANOVA and t-student tests. Statistical analysis was performed using the Statview software.

Results

From August 2023 to June 2024, we analyzed 49 PLWH, 23 on BIC/FTC/TAF and 26 taking DTG/3TC (mean age 69.5, years since HIV diagnosis 28.8, years on ART 23.6, CD4 at inclusion 583 cc/mm3, 49% with nadir CD4 < 200 cc/mm3). Five healthy individuals, matched for age, were included as controls.

PWH had higher immunosenescence markers than healthy subjects. Among PWH, no significant differences were found between those taking BIC/FTC/TAF compared to DTG/3TC (data not shown).
In case of nadir CD4 < 200 cells/mm3, those with triple ART had significantly lower levels of senescent and TEMRA CD4+ and CD8+ cells compared with those taking dual therapy, despite similar age, years since HIV diagnosis, time on ART, CD4 at inclusion and number of comorbid conditions. Moreover, a trend was found for lower CD4/CD8 ratio among those on dual ART and low CD4 nadir (Table 1).

Table 1. Characteristics of subjects included and markers of immunosenescence

	HIV-neg mean±sd n = 5	DTG/3TC mean±sd n = 26	BIC/FTC/TAF mean±sd n = 23	*p* Dual ART vs Triple ART
Demographic and background characteristics				
Age	66.4±2.07	70±6.7	69±6.7	0.84
Years from HIV diagnosis	NA	28.03±9.1	29.7±7.1	0.47
Years on ART	NA	23.1±7.8	24±6.4	0.68
CD4 at inclusion (cc/mm3)	967±126	636±302	524±243	0.165
CD4/CD8 ratio	2.65+±1.36	0.56±0.17	0.72±0.35	0.055*
Number of comorbidies	NA	2.50±1.22	2.0±1.27	0.12
Markers of immunosenescence				
SEN-CD4+	15.6±22.7	61.1± 46.3	24.7±35.1	**0.05**
SEN-CD8+	147±224	401±182	255±128	**0.039***
SEN-CD4-38+	19±23	54±42	17±18	**0.01***
SEN-TEMRA-CD4+	10±9	28±35	5.6±6.3	**0.04***
SEN-EM-CD4+	6±10	28+-24	16+-27	0.07
SEN-CD8-38+	133±208	297±138	173±120	**0.03***
SEN-TEMRA-CD8+	81.2±155	237±119	98.1±98.0	**0.02***
SEN-EM-CD8+	38±46	108±170	36±26	0.06
CD8-TEMRA	109±207	299+-168	124±109	**0.007***
CD4-TEMRA	28.2±30.8	36.2±39.8	11.9±11.2	0.056*

SEN: senescent KLRG1+ CD57+ cells
TEMRA: Terminally differentiated effector
EM: effector memory cells

*PLWH with nadir CD4 count <200 cc/mm^3

Discussion

In PLWH aged over 60 years with nadir CD4 <200 cells/mm^3, we found lower levels of immunosenescence on BIC/FTC/TAF compared to DTG/3TC, despite similar characteristics of subjects.

Senescent cells have numerous defects including a decreased capacity for proliferation, an inability to produce cytokines, short telomeres and low telomerase activity [4].

Indeed, KLRG1 plays an inhibitory role in T and NK cells and its expression typically increases with age, contributing to the inability of the immune system to respond to the antigens and mount optimum responses [8]. Studies targeting KLRG1 showed that not only it serves as a marker of T-cell senescence, but also as predictive marker of disease severity and risk for cancer. Indeed, in cancer cells the dysregulation of immune checkpoint proteins is an important mechanism of tumor immune resistance and KLRG1 has been associated with both solid and haematological malignancies [8]. Moreover, the accumulation of TEMRA, especially in CD8+ cells, has been associated with an impact on adaptive immune responses and consequently with ageing [9].

Our work shows that such markers of immunosenescence are overexpressed in PWH compared to unhealthy subjects. These results are not unusual, considering the profound modifications of the immune system during HIV infection despite successful treatment. The most intriguing results are that PWH under dual ART and with low nadir CD4 displayed higher markers of immunosenescence than those taking a triple therapy, despite similar background characteristics.

Low nadir CD4 has been associated with higher loss of naïve T-cells, increased intestinal permeability and larger reservoir size [10-13]. We suggest that in case of treatment simplification and low nadir CD4, the reduced pharmacological pressure in the anatomical reservoir, together with the dysregulated architecture of immune cells, could explain the increase of immune activation and consequently of senescent markers. Indeed, it has been shown that diffusion of integrase strand transfer inhibitors in lymph nodes is generally poor, thus potentially exposing to insufficient levels of antiretrovirals in this reservoir [14].

Moreover, we previously showed that in case of treatment simplification to dual ART, subjects with either low nadir CD4 or previous AIDS displayed higher markers of monocyte-macrophage activation measured with soluble CD163 in plasma [15].

If confirmed by larger studies, our results suggest that past CD4 count should be considered as a criteria for treatment decision choice between dual and triple ART in order to reduce risks of age-associated complications.

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