Clinical Infectious Diseases

REVIEW ARTICLE

CD4/CD8 ratio during HIV treatment: time for routine monitoring?

Raquel Ron[1,2*], Elena Moreno[2,3*], Javier Martínez-Sanz[1,2], Fátima Brañas[4], Talía Sainz[5], Santiago Moreno[1,2,6], Sergio Serrano-Villar[1,2,7]

[1]Department of Infectious Diseases. Hospital Universitario Ramón y Cajal and IRICYS. Madrid, Spain.; [2]CIBER de Enfermedades Infecciosas (CIBERINFEC). Instituto de Salud Carlos III. Madrid, Spain.; [3]Immunovirology Laboratory. Hospital Universitario Ramón y Cajal and IRICYS. Madrid, Spain.; [4]Department of Geriatrics. Hospital Universitario Infanta Leonor. Madrid, Spain.; [5]Department of Pediatric Infectious Diseases. Hospital Universitario La Paz. Madrid, Spain.; [6]Department of Medicine. Universidad de Alcalá, Madrid, Spain.; [7]HIV Division, Department of Medicine. University of California San Francisco, United States.

In the last decade, studies in PWH on ART have shed light on the significance of persistently high CD8 counts and low CD4/CD8 ratios. A low CD4/CD8 ratio translates increased immune activation and is associated with an increased risk of severe non-AIDS events. As a result, many clinicians now believe that the CD4/CD8 ratio can help in HIV monitoring, and many researchers now report it as an efficacy marker in interventional studies. However, the topic is more complex. Recent studies have not yielded unanimous conclusions on the ability of CD4/CD8 ratio to predict adverse outcomes, and only some clinical guidelines recommend monitoring it. Knowledge gaps remain on the best cut-off points, associated clinical events, effects of treatments, and how CD4/CD8 ratio could improve decision-making in the clinic. Here, we critically review the literature, identify knowledge gaps and discuss the role of the CD4/CD8 ratio as a marker for HIV monitoring.

---

*Equal contribution.

Corresponding author: Sergio Serrano-Villar, Department of Infectious Diseases, Hospital Universitario Ramón y Cajal, 28034 Madrid, Spain. Email: sergio.serrano@salud.madrid.org.

DOI: 10.1093/cid/ciad136

Key words: HIV monitoring; CD4/CD8 ratio; CD8 counts; non-AIDS events; mortality.

INTRODUCTION

PWH receiving antiretroviral therapy (ART) are expected to maintain viral suppression indefinitely. Historically, CD4 counts have guided diagnostic evaluations and ART decisions, leading to a widespread 'CD4 culture' . However, CD4 normalization does not reflect a complete return to health. Residual immune dysfunction persists and contributes to an excess risk of serious non-AIDS events (SNAEs), even when ART is started early[1]. While we can measure this residual immune dysfunction, the markers used in the major studies that led to these conclusions are technically challenging and too variable to be used in the clinics.

Many studies have shown that persistently low CD4 counts or elevated CD8 counts determine low CD4/CD8 ratios, which translates immune activation and is associated with an excess risk of severe non-AIDS events (SNAEs) (**Table 1**). Conversely, a high ratio during ART suggests that immune function has been restored and chronic inflammation has decreased[2]. This cumulative evidence has seeded the idea that the CD4/CD8 ratio could become an important marker for monitoring HIV, but there are still areas of controversy and knowledge gaps.

Determinants of CD8+ T cell expansion and low CD4/CD8 ratio during ART

CD4/CD8 balance is disrupted very soon after acute HIV infection, mainly because of early expansion of the CD8 pool, followed by firstly transient and then progressive CD4 count decline. These trends are only partially reversed in most patients during effective ART, even despite early ART initiation[2–4]. A low CD4/CD8 ratio during ART correlates with markers of inflammation and immunosenescence and shares associations with many putative drivers, including HIV persistence, bacterial translocation, and chronic latent coinfections (**Figure 1**).

Correlations with inflammatory and immunosenescence markers

In the general population, CD8 cell expansion reflects greater immunosenescence and independently predicts mortality, especially in older adults[5,6]. Subjects with CD4/CD8 ratio inversion (<1) showed CD8 cells with shorter telomere length, increased expression of senescence markers in CD8 cells such as CD28-, oligoclonal expansion of CMV-specific CD8+ T cells[7,8], and decreased thymic function. Later studies in PWH on ART showed that low CD4/CD8 ratios also reflect underlying inflammation (IL-6 levels)[2], oxidative stress[9], decreased thymic output[10], and inefficient control of latent viruses such as CMV[11,12], EBV[13], or active coinfections such as HCV[14,15]. Importantly, PWH and low CD4/CD8 ratios exhibit greater inflammation and immunosenescence despite otherwise successful ART[2].

Links with the HIV reservoir

A low CD4/CD8 ratio during ART correlates with larger HIV reservoirs. In two cross-sectional studies in PWH, a higher CD4/CD8 ratio after 48 and 96 weeks of effective ART was predictive of a low level of total HIV DNA in peripheral blood[16,17]. In another prospective study following ART-suppressed patients for more than seven years, there was a 0.5 decrease in HIV intact genomes for each two-fold increase in the ratio[18]. While it is tempting to assume that ongoing transcription of viral proteins could sustain CD8 cell expansion and proliferation, the current evidence only allows for establishing correlations.

HIV persistence, mucosal immunity impairment, and bacterial translocation

Persistent structural and functional defects of gut-associated lymphoid tissues (GALT) possibly contribute to a low CD4/CD8 ratio. Acute HIV infection causes a dramatic GALT injury, not fully reversed by ART, allowing HIV persistence in the gut, bacterial translocation, and immune activation [19]. In GALT, a low CD4/CD8 ratio correlates with the extent of CD4 depletion and CD8 expansion [2]. It also correlates with induction of the indolamine 2-3-deoxygenase pathway —an enzyme involved in tryptophane catabolism[20],—and with direct (lipopolysaccharide) or indirect (sCD14) markers of bacterial translocation[2,21]. IL-15—a cytokine induced by LPS-primed antigen-presenting cells— induces CD8 cell activation and proliferation in treated PWH[22]. Last, a lower ART adherence, which seems to contribute to inflammation even in virologically suppressed patients[23], also correlates with slower CD4/CD8 ratio recovery [24]. Altogether, these studies suggest that a low ratio result from higher HIV transcription in lymphoid tissues, allowing for bacterial translocation, chronic antigen exposure, and IDO1 induction, leading to greater inflammation and senescence,of the immune system,

Effects of sexual practices and CMV

Men who have sex with men (MSM) without HIV infection exhibit lower CD4/CD8 ratios than non-MSM[25]. Intriguingly, this population exhibits a higher risk of virus-associated cancers, including anal cancer, non-Hodgkin lymphoma, and liver cancer[26]. This raises the hypothesis that sexual practices influence the CD4/CD8 ratio. Unprotected sexual intercourse, a greater number of sex partners, and sexually transmitted infections could result in transient GALT damage, leading to bacterial translocation, inflammation, and decreased CD4/CD8 ratios[27]. Furthermore, given the effects of sexual practices and gut microbiota composition[28], and their suggested consequences on inflammation [29], it is plausible that the changes in microbiota composition related to sexual practices also contribute to CD4/CD8 ratio dynamics. In addition, CMV determines CD8 cell expansion and immunosenescence[11,12] and MSM represent a group with a greater CMV prevalence[25].

Can the CD4/CD8 ratio help identify patients at greater risk of severe non-AIDS events?

CD4/CD8 ratio shows a lower intra-individual variability (12%) than the CD4 (16%) or CD8 counts (18%) over time. CD4 and CD8 count recovery are appreciate even after ten years of ART[4,30]. Starting ART with more than 500 CD4 counts is associated with a higher probability of normalizing the CD4 counts over time, but even in this situation and despite long-term ART the CD4/CD8 ratio trajectories remain below the reference levels of people without HIV because of persisting high CD8 counts[30,31].

Studies in the past decade have reported associations between a low CD4/CD8 ratio or high CD8 counts and an excess risk of SNAEs during ART (**Table 1**). This has led many clinicians to view them as meaningful biomarkers for HIV monitoring. Indeed, the European AIDS Clinical Society recommends measuring CD4/CD8 ratio as a stronger predictor of SNAEs than CD4 counts[32]. However, there is still debate on the value of the CD4/CD8 ratio to predict mortality. Some studies[2,33,34], but not all[30,35], have reported an independent association between the CD4/CD8 ratio and non-AIDS mortality.

The topic is methodologically complex, with several sources of heterogeneity across studies. The main issues are related to the timing of CD4 and CD8 measurements with respect to ART initiation, the cut-offs defining CD4/CD8 ratio recovery and disparate definitions of clinical outcomes. In addition, the fact that the CD4 counts are included in the CD4/CD8 ratio definition pose collinearity issues in the statistical models and makes it challenging to prove an independent value. In fact, in a large cohort study with nearly 50,000 participants in the ART-CC collaboration, AIDS-related mortality declined with increasing CD4/CD8 ratios and decreasing CD8 counts, although there was little evidence that CD4/CD8 ratio or CD8 count improved the prediction of non-AIDS mortality[30]. However, recent studies, one of them in the same cohort[36], and others from the Veterans Aging Cohort Study[37] and the Swiss cohort[38] supported a connection between the CD4/CD8 ratio and cancer risk.

Which is the CD4/CD8 ratio cutoff more strongly associated with an increased risk of SNAEs? A value <1 is considered abnormal in the general population[5], although the first studies in PWH on ART found that the predictive cut-offs for a wide range of SNAEs were lower[2,33,39]. PWH and a CD4/CD8 ratio <0.4 exhibited a nearly two to three-fold increased risk of SNAEs. In the START trial, a CD4/CD8 ratio <0.5 before starting ART identified subjects who benefited most from early ART[40].

Conversely, PWH with CD4 counts >500 and CD4/CD8 ratio >1 show almost full recovery in terms of T cell activation, senescence, exhaustion[2,41], and lower risk of adverse clinical outcomes[2]. This suggests that those CD4/CD8 ratios >1 have minimal immune dysfunction and low risk of SNAEs, and might have normalized health.

Are high CD8 counts equivalent to low CD4/CD8 ratios?

Few studies have focused on the specific value of CD8 counts as markers of SNAEs risk. In the ART-CC cohort, CD8 counts showed the lowest mortality rates near the median CD8 cell count value and the highest rates at the CD8 count extremes[30]. In another cohort of 885 PWH, patients with low (<400/uL) CD8 counts exhibited an increased risk of bacterial pneumonia[42]. In the AIDS clinical trials group longitudinal linked randomized trials (ALLRT) cohort, we assessed the prognostic value of the CD8 counts for severe clinical events. While patients with increased CD8 counts at year two of ART initiation exhibited a higher risk of noninfectious non-AIDS events during the next five years, they were also protected against infectious non-AIDS events (including virus-associated cancers). In addition, marked elevations of CD8 counts were associated with an increased risk of non-AIDS mortality[43].

There is also a lack of information for the most discriminative CD8 cut-off counts. In the Copenhagen cohort, a CD8 count >1500 cells/μL after ten years of ART was associated with an 80% increased risk of non-AIDS-related mortality compared with a CD8 count of 500-1500 cells/μL[43]. We also found in the ALLRT cohort that a CD8 count >1500 cells/μL at year 2 of ART predicted a 75% increased risk of AIDS and noninfectious non-AIDS events through years 3-7. Conversely, we observed opposite effects for the risk of severe infections, which were more frequent among individuals with CD8 counts <500 cells/μL[35].

In patients with CD4 count >500/uL, the association between a low CD4/CD8 ratio and the SNAEs risk is mainly driven by the CD8 counts. In contrast, in patients with lower CD4 counts diagnosed with advanced HIV disease, the association between the ratio and mortality depended upon the CD4 counts[2]. These results suggest that much of the harm associated with a low ratio is driven by the CD4 count (and presumably immunodeficiency) in those with low CD4 counts, while the harm associated with a low ratio in those with higher CD4 counts is driven by CD8 counts (and presumably inflammation).

The CD4/CD8 ratio in special populations

Elite controllers (EC)

In EC, the CD4/CD8 ratio correlates with immune activation[44]. Starting ART in EC decreases T cell activation and dysfunction[45] and improves the CD4/CD8 ratio[46]. Among 1,067 elite controllers in the COHERE cohort, CD4 decreased and CD8 increased before loss of virological control, suggesting that these changes may result from repeated low-level viremia. Lower CD4/CD8 ratio was predictive of loss of virological control[47]. Therefore, the ratio could help decide when to initiate ART in EC.

Children with vertical HIV infection

Although the relative risk of SNAEs has not been established yet in vertically infected patients, they exhibit increased atherosclerosis[48]. In keeping with the findings in studies in adults, their CD4/CD8 ratio inversely correlates with inflammation and immune activation, senescence, and exhaustion of T cells[49]. Therefore, a low CD4/CD8 ratio can help identify children with greater immune dysfunction, and perhaps those at increased risk of non-AIDS related comorbidities. It is unknown whether specific interventions can increase CD4/CD8 ratio recovery in children with HIV.

CD4/CD8 ratio recovery and effects of interventions

Clinical factors associated with poorer CD4/CD8 recovery include older age[39], male sex[34], lower nadir CD4 count[33], higher baseline CD4/CD8 ratio or lower baseline CD8 counts[33,3], CMV seropositivity[33,12], lower ART adherence[24] and, most significantly, late ART initiation[2,33,3]. The recovery of the CD4/CD8 ratio is greater in the first two years of ART[2,3,50], but approximately two-thirds of patients on long-term ART still have a CD4/CD8 ratio below the average levels of individuals without HIV because of high CD8 counts[2,33,3,50]. Hence, an adequate CD4 count recovery does not imply CD4/CD8 ratio normalization and identifies PWH needing additional interventions.

Because the CD4/CD8 ratio and CD8 counts were not recognized before as relevant biomarkers and the main regulatory agencies only require HIV RNA as the ART efficacy endpoint, the CD4/CD8 ratio and CD8 counts have been largely overlooked in studies assessing HIV treatment. To fill this gap, several *post hoc* analyses of clinical trials and observational studies have examined the effects of ART initiation on the CD4/CD8 ratio (**Table 2**). In STARTMRK, a faster CD4/CD8 ratio normalization was seen over five years of treatment with raltegravir compared to efavirenz[51]. In the MERIT trial, dolutegravir led to higher CD4/CD8 normalization rates than with zidovudine, which was driven by a greater CD8 count decline[52]. In the SPRING-2 study, no differences were seen in CD4/CD8 ratio recovery with raltegravir compared to dolutegravir[53]. These analyses indicated INSTI could have a stronger effect on CD4/CD8 ratio recovery and CD8 count decay than other families, but data from real-life settings were lacking. In the Spanish CORIS cohort, subjects starting INSTI-based first-line ART experienced greater rates of CD4/CD8 normalization at different cut-offs than those starting with PI or NNRTI. Strikingly, the largest differences in CD4/CD8 ratio trajectories were driven by changes in the CD8 cell counts[50].

In contrast, the number of drugs does not seem to influence the rates of CD4/CD8 ratio recovery. Previous studies evaluating ART intensification with four drugs did not find greater CD4/CD8 ratio recovery neither in peripheral blood or GALT[2]. Similarly, no differences appeared in controlled trials of ART switch when using two-drug combinations, including dolutegravir, compared to three-drug combinations[54,55]. Similarly, in CORIS, no differences appeared in

the rates of CD4/CD8 normalization between subjects starting ART initiation with dolutegravir plus lamivudine vs. dolutegravir or bictegravir-based triple antiretroviral therapy at 48 weeks[56].

The impact of ART switches on long-term CD4/CD8 ratio changes remains less studied. Large clinical trials of modern ART combinations such as dolutegravir and lamivudine or dolutegravir and rilpivirine have not found differences relative to their comparators[54,55]. However, in contrast to the ART initiation settings, studies focusing on the effects of treatment changes in suppressed individuals will require larger sample sizes and longer follow-ups, given the smaller variation of the CD4/CD8 ratio in patients virally suppressed[50].

Studies assessing therapeutic interventions beyond ART are limited. Therapies aimed at targeting lymphoid fibrosis (losartan)[57], inflammation (metformin)[58], or the microbiota (synbiotics and fecal microbiota transplants)[59,60] did not result in a detectable effect on CD4/CD8 ratios.

Clinical applications

The immediate clinical implication of CD4/CD8 ratio monitoring would be to enforce primary prevention of age-associated conditions in patients with persistently low ratios despite ART with more aggressive management of risk factors. Ratio monitoring would detect trend changes that might prompt awareness about the development of comorbidities (**Figure 1**).

Given the consistent links between a low CD4/CD8 ratio and increased risk of some preventable cancers, this population could benefit from earlier cancer screening[36,37,61]. Some examples include lung cancer, with a higher burden incidence and poorer prognosis in PWH[62], and high-grade dysplasia for anal cancer screening, provided the strong predictive value for advanced anal disease of a low CD4/CD8 ratio[61]. In addition, we should carefully check ART adherence in patients with poor CD4/CD8 ratio recovery[24].

Cardiovascular risk evaluation is challenging in PWH, as main tools are calibrated in the general population, resulting in underestimation. The excess risk linked to HIV is partially attributed to chronic immune dysfunction, and there is known association between low CD4/CD8 ratio and higher cardiovascular risk[63]. The CD4/CD8 ratio could be used as a modifying risk factor to guide the initiation of preventive measures, for example statin use in subjects with intermediate cardiovascular risk.

For patients reluctant to go on therapy, a low ratio can provide a tangible argument to initiate ART, since early treatment is strongly associated with achieving a normal ratio[2,33,3]. In patients on stable ART, a normal ratio may provide some sense of reassurance that treatment is working well and that they may have achieved optimal immunologic health. Since the CD4/CD8 ratio varies slowly during ART[3,4], measuring the ratio once per year would be enough. Therefore, incorporating the ratio in routine monitoring should not result in more frequent clinical appointments.

For research, the ratio could help identify those still in need of additional interventions beyond antiretroviral drugs, evaluate strategies targeting immune dysfunction, detect patients with smaller HIV reservoirs, and select the best candidates for interventions to achieve ART-free HIV remission.

So far, the only evidence-based intervention to achieve better CD4/CD8 ratios is to start ART early, which is probably the best strategy to reduce immune dysfunction and chronic inflammation. INSTI-based ART is already the preferred combination in current clinical guidelines and also seems to increase the CD4/CD8 ratio to a greater extent than other combinations. However, the mechanisms remain to be elucidated. Conceptually, any therapy able to attenuate inflammation or immune dysfunction in treated patients could improve the CD4/CD8 ratio recovery. Finally, the ratio is a readily available biomarker in most settings. While not always reported, the CD8 counts are typically measured in most settings for a more accurate definition of the CD4+ T cell population. Given the additional prognostic information captured by this biomarker, it should be considered for routine HIV monitoring and as an exploratory outcome in interventional studies.

Future directions

Future studies should address the disease-specific relationships with CD8 counts and CD4/CD8 ratio and the most discriminative cutoffs for each type of event. Ideally, studies aimed at better defining the predictive value of the CD4/CD8 ratio should use landmark analysis to enable interpretation of time-dependent variables. In addition, the group of patients with low CD4 counts but normal CD4/CD8 ratios represents an understudied population that warrants more investigation.

We need mechanistic insight explaining how the peripheral pool of CD4+ and CD8+ T cells dynamically correlate with the respective subsets in lymphoid tissues and the implications of ART drug concentrations in tissues. It is also unclear whether the greater CD4/CD8 ratio recovery appreciated with INSTI-based ART is explained by better tolerance resulting in higher ART adherence, different penetration in tissues, or class effects. We need proof-of-concept studies evaluating the impact of specific therapeutic interventions on CD8 cell and CD4/CD8 ratio changes, especially those targeting the putative drivers of low CD4/CD8 ratios despite ART, and implementation studies evaluating whether CD4/CD8 ratio monitoring improves HIV care.

In conclusion, the CD4/CD8 ratio and CD8 counts offer the possibility to detect ongoing immune dysfunction known to contribute to the excess risk of mortality during treated HIV. Dissecting better the mechanisms driving CD8+ T cell expansion during ART will be necessary to develop innovative strategies targeted at PWH with increased CD8 counts. Until then, we can harness the associations between the ratio and CD8 counts with increased risk of SNAEs to better monitor HIV infection.

NOTES

FUNDING: This work was supported by the Instituto de Salud Carlos III, projects PI18/00154, co-funded by ERDF, "A way to make Europe", ICI20/00058 and PI21/00141, co-funded by the European Union.

CONFLICTS OF INTEREST: Outside the submitted work, J.M.-S. and R.R. have received non-financial support from ViiV Healthcare, Jannsen Cilag, and Gilead Sciences, outside the submitted work. J.M.-S. reports consulting fees from Gilead Sciences; payment or honoraria from Gilead Sciences, ViiV Healthcare, and Janssen Cilag; and travel support for attending conferences from ViiV Healthcare and Gilead. R.R. reports honoraria from Gilead Sciences for presenting at an educational event and travel support for conferences from Janssen-Cilag. S.M. reports grants, personal fees and non-financial travel support from ViiV Healthcare, grants, personal fees and non-financial support from Gilead, personal fees and non-financial travel support from Janssen, grants, personal fees and non-financial travel support from MSD, outside the submitted work. S.M. also reports participation on a Data Safety Monitoring or Advisory Board for MSD. E.M. reports non-financial and/or travel support for conferences from ViiV Healthcare and Gilead sicences, outside the submitted work. T.S. reports grants from MSD and Gilead, outside the submitted work. S. S.-V. reports personal fees including payment or honoraria from Gilead Sciences and MSD, consulting fees from Mikrobiomik and Aptatargets, non-financial and/or travel support for conferences from ViiV Healthcare and Gilead Sciences, and research grants from MSD and Gilead Sciences, paid to their institution outside the submitted work. S.S.-V. also reports planned, issued, or pending patents for Microbiome-associated biomarkers of anal cancer and SARS-CoV-2 susceptibility test in saliva. Figure 1 was generated in www.biorender.com.

REFERENCES

(first 50. Complete list in the supplementary materials)

1. Sereti I, Krebs SJ, Phanuphak N, et al. Persistent, Albeit reduced, chronic inflammation in persons starting antiretroviral therapy in acute HIV infection. Clinical Infectious Diseases **2017**; 64:124–131.
2. Serrano-Villar S, Sainz T, Lee S a., et al. HIV-Infected Individuals with Low CD4/CD8 Ratio despite Effective Antiretroviral Therapy Exhibit Altered T Cell Subsets, Heightened CD8+ T Cell Activation, and Increased Risk of Non-AIDS Morbidity and Mortality. PLoS Pathogens **2014**; 10:e1004078.
3. Caby F, Mary-Krause M, Duval X, et al. CD4+/CD8+ratio restoration in long-term treated HIV-1-infected individuals. Aids **2017**; 31:1685–1695.
4. Hughes RA, May MT, Tilling K, et al. Long terms trends in CD4+ cell counts, CD8+ cell counts, and the CD4+ : CD8+ ratio. Aids **2018**; 32:1361–1367.

5. Wikby A, Maxson P, Olsson J, Johansson B, Ferguson FG. Changes in CD8 and CD4 lymphocyte subsets, T cell proliferation responses and non-survival in the very old: the Swedish longitudinal OCTO-immune study. Mechanisms of Ageing and Development **1998**; 102:187–198.
6. Ferrando-Martínez S, Romero-Sánchez MC, Solana R, et al. Thymic function failure and C-reactive protein levels are independent predictors of all-cause mortality in healthy elderly humans. Age (Dordrecht, Netherlands) **2013**; 35:251–9.
7. Hadrup SR, Strindhall J, Køllgaard T, et al. Longitudinal Studies of Clonally Expanded CD8 T Cells Reveal a Repertoire Shrinkage Predicting Mortality and an Increased Number of Dysfunctional Cytomegalovirus-Specific T Cells in the Very Elderly. The Journal of Immunology **2006**; 176:2645–2653.
8. Wikby A, Månsson I a., Johansson B, Strindhall J, Nilsson SE. The immune risk profile is associated with age and gender: Findings from three Swedish population studies of individuals 20-100 years of age. Biogerontology **2008**; 9:299–308.
9. Muller GC, Gottlieb MGV, Luz Correa B, Filho IG, Moresco RN, Bauer ME. The inverted CD4: CD8 ratio is associated with gender-related changes in oxidative stress during aging. Cellular Immunology **2015**; 296:149–154.
10. Rosado-Sánchez I, Herrero-Fernández I, Genebat M, Ruiz-Mateos E, Leal M, Pacheco YM. Thymic Function impacts the peripheral CD4/CD8 Ratio of HIV-infected subjects. Clinical Infectious Diseases **2017**; 64:152–158.
11. Smith DM, Nakazawa M, Freeman ML, et al. Asymptomatic CMV replication during early Human Immunodeficiency Virus (HIV) infection is associated with lower CD4/CD8 ratio during HIV treatment. Clinical Infectious Diseases **2016**; 63:1517–1524.
12. Freeman ML, Mudd JC, Shive CL, et al. CD8 T-cell expansion and inflammation linked to CMV coinfection in ART-treated HIV infection. Clinical Infectious Diseases **2016**; 62:392–396.
13. Gianella S, Moser C, Vitomirov A, et al. Presence of asymptomatic cytomegalovirus and Epstein--Barr virus DNA in blood of persons with HIV starting antiretroviral therapy is associated with non-AIDS clinical events. AIDS **2020**; 34:849–857.
14. Zaegel-Faucher O, Bregigeon S, Cano CE, et al. Impact of hepatitis C virus coinfection on T-cell dynamics in long-term HIV-suppressors under combined antiretroviral therapy. Aids **2015**; 29:1505–1510.
15. Bandera A, Lorenzini P, Taramasso L, et al. The impact of DAA-mediated HCV eradication on CD4+ and CD8+ T lymphocyte trajectories in HIV/HCV coinfected patients: Data from the ICONA Foundation Cohort. Journal of Viral Hepatitis **2021**; 28:779–786.
16. Gibellini L, Pecorini S, De Biasi S, et al. HIV-DNA content in different CD4+T-cell subsets correlates with CD4+cell : CD8+cell ratio or length of efficient treatment. Aids **2017**; 31:1387–1392.
17. Yue Y, Wang N, Han Y, et al. A higher CD4/CD8 ratio correlates with an ultralow cell-associated HIV-1 DNA level in chronically infected patients on antiretroviral therapy: A case control study. BMC Infectious Diseases **2017**; 17:771.
18. Peluso MJ, Bacchetti P, Ritter KD, et al. Differential decay of intact and defective proviral DNA in HIV-1–infected individuals on suppressive antiretroviral therapy. JCI Insight **2020**; 5:0–13.
19. Somsouk M, Estes JD, Deleage C, et al. Gut epithelial barrier and systemic inflammation during chronic HIV infection. Aids **2015**; 29:43–51.

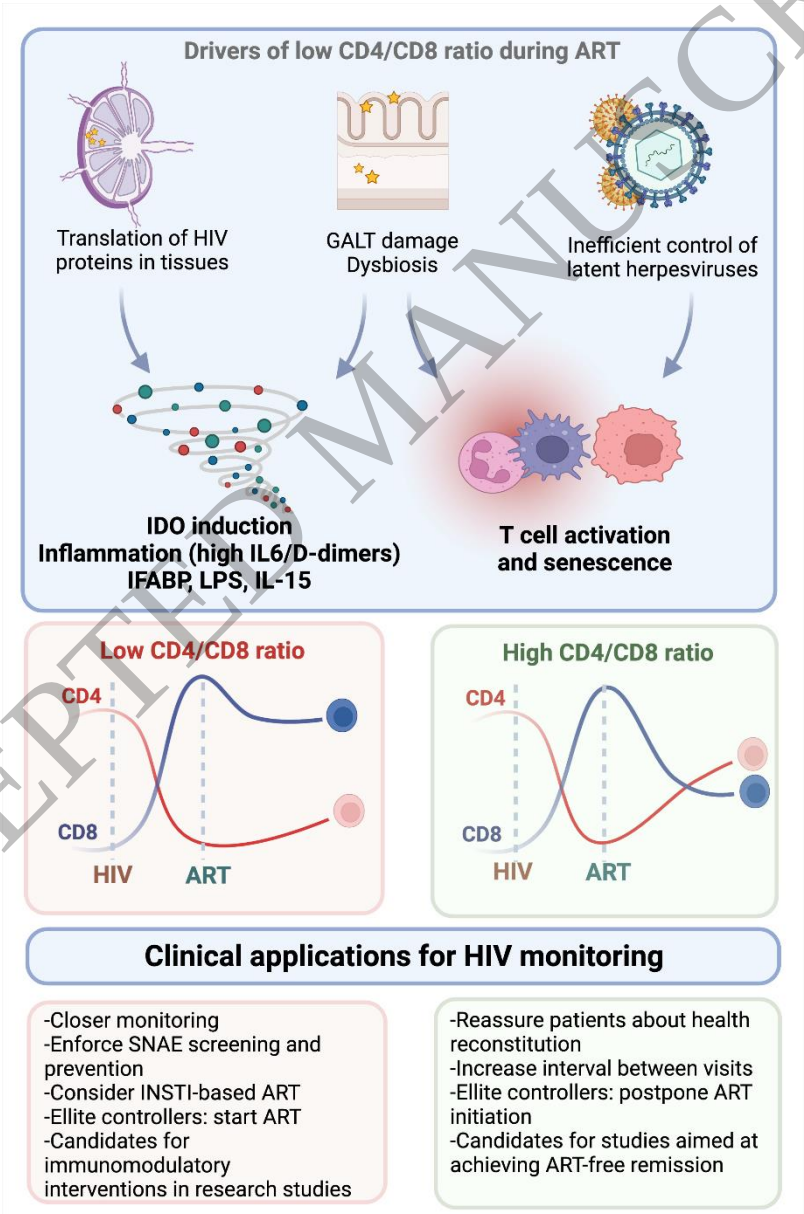
20. Boasso A, Herbeuval J-P, Hardy AW, et al. HIV inhibits CD4+ T-cell proliferation by inducing indoleamine 2,3-dioxygenase in plasmacytoid dendritic cells. Blood **2007**; 109:3351–9.
21. Tinago W, Cotter AG, Sabin CA, et al. Predictors of longitudinal change in bone mineral density in a cohort of HIV-positive and negative patients. Aids **2017**; 31:643–652.
22. Younes SA, Freeman ML, Mudd JC, et al. IL-15 promotes activation and expansion of CD8+T cells in HIV-1 infection. Journal of Clinical Investigation **2016**; 126:2745–2756.
23. Castillo-Mancilla JR, Brown TT, Erlandson KM, et al. Suboptimal Adherence to Combination Antiretroviral Therapy Is Associated With Higher Levels of Inflammation Despite HIV Suppression. Clinical infectious diseases : an official publication of the Infectious Diseases Society of America **2016**; 63:1661–1667.
24. Belaunzaran-Zamudio PF, Naranjo L, Caro-Vega Y, et al. Incomplete ART Adherence Is Associated with Lower CD4-CD8 Ratio in Virally Suppressed Patients with HIV-infection in Mexico. AIDS Research and Human Retroviruses **2023**; Available at: https://www.liebertpub.com/doi/abs/10.1089/AID.2021.0179. Accessed 7 February 2023.
25. Verboeket SO, Wit FW, Verheij E, et al. Human Immunodeficiency Virus (HIV)-Negative Men Who Have Sex with Men Have Higher CD8+T-Cell Counts and Lower CD4+/CD8+T-Cell Ratios Compared with HIV-Negative Heterosexual Men. Journal of Infectious Diseases **2021**; 224:1187–1197.
26. Dutta A, Uno H, Lorenz DR, Wolinsky SM, Gabuzda D. Low T-cell subsets prior to development of virus-associated cancer in HIV-seronegative men who have sex with men. Cancer Causes and Control **2018**; 29:1131–1142.
27. Palmer CD, Tomassilli J, Sirignano M, et al. Enhanced immune activation linked to endotoxemia in HIV-1 seronegative MSM. AIDS (London, England) **2014**; :1–4.
28. Noguera-Julian M, Rocafort M, Guillén Y, et al. Gut Microbiota Linked to Sexual Preference and HIV Infection. EBioMedicine **2016**; 5:135–146.
29. Littlefield KM, Schneider JM, Neff CP, et al. Elevated inflammatory fecal immune factors in men who have sex with men with HIV associate with microbiome composition and gut barrier function. Frontiers in Immunology **2022**; 13. Available at: https://www.frontiersin.org/articles/10.3389/fimmu.2022.1072720. Accessed 30 December 2022.
30. Trickey A, May MT, Schommers P, et al. CD4:CD8 Ratio and CD8 Count as prognostic markers for mortality in human immunodeficiency virus-infected patients on antiretroviral therapy: The antiretroviral therapy cohort collaboration (ART-CC). Clinical Infectious Diseases **2017**; 65:959–966.
31. Gras L, May M, Ryder LP, et al. Determinants of Restoration of CD4 and CD8 Cell Counts and their Ratio in HIV-1 Positive Individuals with Sustained Virological Suppression on Antiretroviral Therapy. Journal of Acquired Immune Deficiency Syndromes **2019**; 80:292–300.
32. European AIDS Clinical Society. EACS Guidelines. Version 11.0. October 2021. Available at: http://www.eacsociety.org/guidelines/eacs-guidelines/eacs-guidelines.html. Accessed April 14, 2022.
33. Mussini C, Lorenzini P, Cozzi-Lepri A, et al. CD4/CD8 ratio normalisation and non-AIDS-related events in individuals with HIV who achieve viral load suppression with antiretroviral therapy: An observational cohort study. The Lancet HIV **2015**; 2:e98–e106.

34. Han WM, Apornpong T, Kerr SJ, et al. CD4/CD8 ratio normalization rates and low ratio as prognostic marker for non-AIDS defining events among long-term virologically suppressed people living with HIV. AIDS Research and Therapy **2018**; 15:13.
35. Serrano-villar S, Wu K, Hunt PW, et al. Predictive value of CD8 + T cell and CD4 / CD8 ratio at two years of successful ART in the risk of AIDS and non-AIDS events. eBioMedicine **2022**; 80:104072.
36. Castilho JL, Bian A, Jenkins CA, et al. CD4/CD8 Ratio and Cancer Risk among Adults with HIV. JNCI: Journal of the National Cancer Institute **2022**; 114:854–862.
37. Sigel K, Wisnivesky J, Crothers K, et al. Immunological and infectious risk factors for lung cancer in US veterans with HIV: a longitudinal cohort study. The Lancet HIV **2017**; 4:e67–e73.
38. Clifford GM, Lise M, Franceschi S, et al. Lung cancer in the Swiss HIV Cohort Study: role of smoking, immunodeficiency and pulmonary infection. British journal of cancer **2012**; 106:447–52.
39. Serrano-Villar S, Pérez-Elías MJ, Dronda F, et al. Increased risk of serious non-AIDS-related events in HIV-infected subjects on antiretroviral therapy associated with a low CD4/CD8 ratio. PLoS ONE **2014**; 9:e85798.
40. Molina JM, Grund B, Gordin F, et al. Which HIV-infected adults with high CD4 T-cell counts benefit most from immediate initiation of antiretroviral therapy? A post-hoc subgroup analysis of the START trial. The Lancet HIV **2018**; 5:e172–e180.
41. Tinago W, Coghlan E, Macken A, et al. Clinical, immunological and treatment-related factors associated with normalised CD4+/CD8+ T-cell ratio: effect of naïve and memory T-cell subsets. PloS one **2014**; 9:e97011.
42. Gohil SK, Heo M, Schoenbaum EE, Celentano D, Pirofski LA. CD8 + T cells and risk for bacterial pneumonia and all-cause mortality among HIV-infected women. Journal of Acquired Immune Deficiency Syndromes **2012**; 60:191–198.
43. Helleberg M, Kronborg G, Ullum H, Ryder LP, Obel N, Gerstoft J. Course and Clinical Significance of CD8+ T-Cell Counts in a Large Cohort of HIV-Infected Individuals. The Journal of infectious diseases **2015**; 211:1726–34.
44. Sajadi MM, Pulijala R, Redfield RR, Talwani R. Chronic immune activation and decreased CD4 cell counts associated with hepatitis C infection in HIV-1 natural viral suppressors. Aids **2012**; 26:1879–1884.
45. Hatano H, Yukl SA, Ferre AL, et al. Prospective Antiretroviral Treatment of Asymptomatic, HIV-1 Infected Controllers. PLoS Pathogens **2013**; 9:e1003691.
46. Kim CJ, Kovacs C, Chun TW, et al. Antiretroviral therapy in HIV-infected elite controllers: Impact on gut immunology, microbial translocation, and biomarkers of serious non-AIDS conditions. Journal of Acquired Immune Deficiency Syndromes **2014**; 67:514–518.
47. Chereau F, Madec Y, Sabin C, et al. Impact of CD4 and CD8 dynamics and viral rebounds on loss of virological control in HIV controllers. PLoS ONE **2017**; 12:e0173893.
48. Sainz T, Álvarez-Fuente M, Navarro ML, et al. Subclinical Atherosclerosis and Markers of Immune Activation in HIV-Infected Children and Adolescents. JAIDS Journal of Acquired Immune Deficiency Syndromes **2014**; 65:42–49.
49. Carrasco I, Tarancon-Diez L, Vázquez-Alejo E, et al. Innate and adaptive abnormalities in youth with vertically acquired HIV through a multicentre cohort in Spain. J Int AIDS Soc **2021**; 24:e25804.

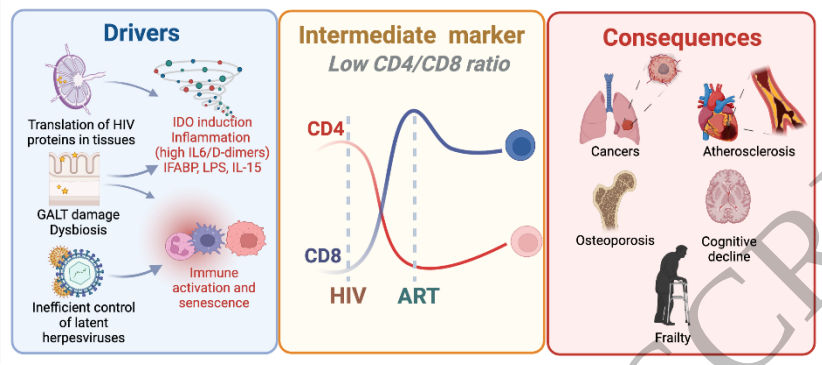
50. Serrano-Villar S, Martínez-Sanz J, Ron R, et al. Effects of first-line antiretroviral therapy on the CD4/CD8 ratio and CD8 cell counts in CoRIS: a prospective multicentre cohort study. The Lancet HIV **2020**; 7:e565–e573.

FIGURE LEGEND

Figure 1. Factors influencing CD4/CD8 ratio dynamics during ART and clinical applications of CD4/CD8 ratio monitoring during treated HIV infection.



Abbreviatures: ART, antiretroviral therapy; GALT, gut-associated lymphoid tissue, IDO, indoleamine 2,3-dioxygenase-1.



Graphical Abstract

Tables

Table 1. Principal studies associating a low CD4/CD8 ratio with increased risk of serious non-AIDS events

Study	Study design	Sample size	Follow up	Biomarker and timing of measurement	Clinical outcomes analyzed	Main findings
Castilho JL. AIDS 2016 [63]	Retrospective cohort	2006	14 years	CD4/CD8 after one year of viral suppression	Cardiovascular Cancer Hepatopathy Renal	Ratio increase as protective factor of cardiovascular events in patients <50 years old
Mussini C. Lancet HIV 2015 [33]	Prospective multicentric cohort	3236	16 years	CD4/CD8 nearest to the event	Cancer Cardiovascular (MI, coronary disease) Cerebrovascular (stroke) Hepatopathy (cirrhosis, encephalopathy) Renal (AKI) Death	Ratio <0.30 associated with high risk of clinical events or death
Serrano-Villar S. PLoS One 2014 [39]	Case-control	407	13 years	CD4/CD8 nearest to the event	Cardiovascular Cerebrovascular Chronic renal disease Cancer	CD4/CD8 ratio <0.4 associated with high risk
Serrano-Villar S. Ebiomedicine 2022 [35]	Pooled randomized control trials	5133	7 years	CD4/CD8 at year 2 of viral suppression	AIDS-defining events Non-AIDS-defining events (liver, cardiovascular, renal disease, non-AIDS cancers, fractures, diabetes, serious bacterial infection, and non-accidental death)	CD4/CD8 ratio <0.4 not associated with high risk CD8 ≥1500 associated with high risk
Sanger CB. Dis Colon Rectum	Retrospective multicentric cohort	2267	18 years	CD4/CD8 nadir and nearest to the event	Anal high-grade squamous intraepithelial lesions and anal	Ratio increment associated with risk reduction

2021 [61]					cancer	
Hema MN. Plos One 2016 [64]	Prospective multicentric cohort	1227	9 years	CD4/CD8 nearest to the event	Cancer: gastrointestinal, hepatic, ORL, urologic, lung, skin, Hodgkin lymphoma	CD4/CD8 ratio <0.5 associated with high risk of non-AIDS cancers
Sigel K. Lancet HIV 2017 [37]	Prospective multicentric cohort	21666	14 years	CD4/CD8 on the last year	Lung cancer	Low CD4/CD8 ratio associated with high risk
Castilho JL. J Natl Cancer Inst 2022 [36]	Retrospective multicentric cohort	83893	8.5 years	CD4/CD8 ratio 6, 12, 18, and 24 months lagged	AIDS and non-AIDS defining cancer	CD4/CD8 ratio of 0.30 associated with increased risk of any incident cancer

Table 2. Main studies comparing the effects of different ART regimens at initiation or switch on CD4/CD8 ratio changes.

Study	ART regimen	Study design	Sample size	Follow up (weeks)	Main findings
First-line ART					
Martínez-Sanz J. Frontiers in immunology 2022 [56]	Lamivudine + dolutegravir vs. 2NRTI + dolutegravir or bictegravir	Multicentric prospective cohort (CORIS)	2214	48	No differences between 2DC and 3DC in CD4/CD8 ratio normalization at 0.5, 1.0 and 1.5 cut-offs
Serrano-Villar S. Lancet HIV 2020 [50]	2 NRTI + INSTI or NNRTI or PI	Multicentric prospective cohort (CORIS)	6804	196	INSTI regimens associated with greater CD4/CD8 gain, more significant during the first year
Figueroa MI. HIV Medicine 2021 [65]	Lopinavir/ritonavir or darunavir/ritonavir-based 2DC vs. 3DC	Pooled analysis or randomized double-blind clinical trial (GARDEL and ANDES, posthoc analysis)	567	48	No differences observed in CD4/CD8 ratio or in the proportion of patients with CD4/CD8 ratio > 1
Fabbiani M. Journal of the Acquired Immune Deficiency Syndrome 2021 [66]	2 NRTI + INSTI or NNRTI or PI	Multicentric retrospective cohort	1428	144	INSTI-based regimens had a higher probability of CD4/CD8 ratio normalization (≥1) and optimal immunological recovery both in total population and in advanced disease
Blanco JR. Plos One 2020 [53]	2 NRTI + raltegravir or dolutegravir	Randomized double blind clinical trial (SPRING-2, posthoc analysis)	822	96	No differences between groups.
Herrera S. Journal of Antimicrobial Chemotherapy 2018 [67]	2 NRTI + INSTI or NNRTI or PI	Multicentric retrospective cohort (CNICS)	3971	48	INSTI-based regimens had a higher probability of CD4/CD8 ratio normalization (≥1) compared with the PI group
Blanco JR. Clinical Microbiology and Infection 2018 [68]	Tenofovir difumarate/emtricitabine/efavirenz vs.	Randomized double blind clinical trial (SINGLE, posthoc analysis)	721	96	Greater increases in CD4/CD8 ratio >1 with tenofovir difumarate/emtricitabine/efavirenz than abacavir/lamivudine/dolutegravir

	abacavir/lamivudine/dolutegravir				
Serrano-Villar S. Journal of Antimicrobial Chemotherapy 2017 [51]	2 NRTI + efavirenz or raltegravir	Randomized double blind clinical trial (STARTMRK, posthoc analysis)	563	240	Raltegravir showed faster CD4/CD8 ratio normalization compared with efavirenz
Serrano-Villar. Antimicrobial Agents Chemotherapy 2017 [52]	Zidovudine and lamivudine + efavirenz or maraviroc	Randomized clinical trial (MERIT, posthoc analysis)	721	240	Higher rates of CD4/CD8 ratio normalization with efavirenz.
De Salvador-Guillouët F. Plos One 2015 [69]	2 NRTI + INSTI or NNRTI or PI	Prospective cohort	567	52	Association between INSTI based regimens and ratio normalization (>1)
ART switch					
Trujillo-Rodríguez M.Clinical Microbiology and Infection 2022 [55]	2DC (lamivudine + dolutegravir or darunavir/cobicistat) vs. maintaining 3DC tenofovir alafenamide/emtricitabine/elvitegravir/cobicistat or lamivudine/abacavir/dolutegravir	Open-label randomized control trial	151	96	Similar changes in CD4/CD8 ratio and inflammatory markers.
Llibre JM. OFID 2022 [54]	Dolutegravir + rilpivirine vs. TAF-based 3DC	Randomized clinical trial (TANGO, posthoc analysis)	741	48	No differences in CD4/CD8 ratio between groups
Llibre JM. OFID 2022 [54]	Dolutegravir + lamivudine vs. TAF-based 3DC	Randomized clinical trial (SALSA, posthoc analysis)	493	48	No differences in CD4/CD8 ratio between groups

Abbreviatures: NRTI, nucleoside reverse transcriptor inhibitor; NNRTI, non-nucleoside reverse transcriptor inhibitors; PI, protease inhibitors; INSTI, integrase strand-transfer inhibitors; 2DC, two-drug ART combinations; 3DC, three-drug ART combinations