Drop in CD4+ counts below 200 cells/µL after reaching (or starting from) values higher than 350 cells/µL in HIV-infected patients with virological suppression.

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Abstract

Background. Aim of the study was to quantify the risk of a drop in CD4+ counts below 200 cells/μL after reaching values >350 cells/μL on antiretroviral therapy (or after starting antiretroviral therapy with CD4+ count >350 cells/μL) in the absence of virological failure.

Setting. Ambulatory care services in Italy.

Methods. Prospective cohort study of patients enrolled in the ICONA Foundation Study cohort. All patients who started ART with >350 CD4+/μL, or with ≤350 CD4+/μL, but reached values >350 cells/μL after virological suppression (VS: two consecutive viral loads ≤50 copies/mL). The date of CD4 count>350 was the baseline for the analysis and those with ≥1 viral load and CD4+ count after baseline were included. The primary end-point was the cumulative risk (Kaplan-Meier estimates) of a CD4+ drop below 200 cells/μL over follow-up, which was censored at the date of virological failure (confirmed HIV-RNA >50 copies/mL), death or last visit.

Results: Six thousand six hundred sixty-three patients were included. A confirmed CD4+ drop below 200 cells/μL was never observed over a median follow-up of 45 (Q1: 21, Q3: 89) months, as long as VS was maintained. Upper limits of the 97.5%CI of rates of confirmed CD4+ drop below 200 cells/μL were 0.28 and 0.38/1,000 PYFU for patients with ≤350 and >350 CD4+ cells/μL at starting ART, respectively.

Conclusions: In patients who started ART in Italy with >350 CD4+ cells/μL, or reached >350 CD4+ cells/μL after VS, the risk of a CD4+ drop below 200 cells/μL in those maintaining VS was negligible.

Keywords: CD4+ cells count; CD4+ count dipping; CD4+ count monitoring; virological suppression; antiretroviral therapy.

Introduction

CD4+ T-lympocytes count is the strongest predictor of disease progression in HIV-infected patients and prophylaxis of opportunistic infection is recommended when it drops below 200 cells/µL [1-6]. Frequent CD4+ cell count monitoring has been highly recommended for many years, but a great debate on the utility of their monitoring arose more recently [7, 8]. In HIV-infected people receiving antiretroviral therapy (ART), CD4+ counts tend to increase or remain stable as long as viral replication is controlled [9-12] and clinical events seldom occur after ART introduction [10, 13]. Furthermore, there are no treatment strategies to increase CD4+ cell count during periods of viral suppression. A CD4+ count drop below 200 cells/µL is currently seldom observed during successful antiretroviral therapy [14-17] and frequent testing may cause unnecessary anxiety in patients with clinically inconsequential fluctuations: thus, the utility of their monitoring has become a matter of debate [5, 6, 17]. Indeed, treatment decisions clinician can take in response to CD4+ monitoring is starting or restarting prophylaxis of opportunistic infections when counts drop below 200 cells/µL [1-4] or, in clinical settings were viral load testing is not routinely implemented, triggering viral load testing [18]. In this context, some guidelines recommend optional or stop monitoring CD4+ cell counts once virological suppressions is sustained, and the immunological status is stabilized [1,3, 18].

Randomized comparisons of individuals following different CD4+ count monitoring strategies are difficult to be performed, and currently lacking, and, although it is well established that CD4+ cells dipping below 200 cells/µL occur infrequently while viral load is suppressed [14-17, 19], robust estimates of CD4+ cells dipping below 200 cells/µL over long follow-up are limited. Furthermore, relevant differences might emerge when considering countries with different organization of the social health system.

Aim of this analysis was to quantify the probability of a drop in CD4+ counts below 200 cells/µL after reaching values >350 cells/µL on ART (or when starting ART with CD4+ count >350 cells/µL) in the absence of virological failure (VF), in a cohort of Italian HIV-infected patients.

Methods

The ICONA Foundation Study (ICONA) is a multi-center prospective observational study of HIV-1-infected adult patients, which was set up in 1997. Eligible patients are those starting ART when they are naive to antiretrovirals. The ICONA Foundation study has been approved by IRB of all the participating centers; sensitive data from patients are seen only in aggregate form. All patients sign a consent form to participate in ICONA, in accordance with the ethical standards of the committee on human experimentation and the Helsinki Declaration (1983 revision). Demographic, clinical and laboratory data and information on therapy are collected for all participants and recorded using electronic data collection [www.icona.org].

CD4+ monitoring in the cohort was performed at least twice yearly, according to study protocol and to Italian guidelines [2].

Antiretroviral regimens used by the studied patients were not predefined by the study protocol (which is strictly observational), but were prescribed according to current Italian guidelines [2].

Virological suppression (VS) was defined as having achieved on two consecutive occasions a viral load $\leq$50 copies/mL. All patients enrolled in the ICONA Foundation Study cohort who started ART with >350 CD4+/µL, or with ≤350 CD4+/µL, but reached values >350 cells/µL after VS, and had at least one VL and CD4+ count assessed after baseline, were included in this analysis. In patients who started ART with >350 CD4+/µL, baseline was defined as the date of VS after at least 6 months since initiation of ART. In those who started ART with ≤350 CD4+/µL, baseline was the date of first achieving a CD4+ value >350 CD4+/µL after VS.

Viral load was assessed in each center according to local procedures. Yearly change in CD4+ cell counts after baseline were estimated from fitting a linear mixed model.

The primary end-point was the cumulative risk (Kaplan-Meier estimates) of a confirmed CD4+ drop below 200 cells/µL during follow-up, which was censored at the date of VF (defined as a confirmed HIV-RNA >50 copies/mL), death or last clinical visit. A CD4+ count drop below 200 cells/µL was defined as the occurrence

of two consecutive CD4+ count below the threshold of 200 cells/μL. All analyses were repeated also using an alternative definition of viral rebound (with a cut-off of 400 copies/mL).

All analyses were performed using SAS version 9.4 (SAS Institute, Cary, NC, USA).

Results

Six thousand six hundred sixty-three adult patients were included in the analysis (Table 1). The main differences between patients who started ART with <350 CD4+/µL and those who started ART with >350 CD4+/µL regarded gender, risk factor, nationality, CD4+ nadir, CD4+/CD8+ ratio, HCV co-infection and HIV-RNA load.

Over a median (Q1, Q3) follow-up of 45 (21, 89) months, AIDS-defining events occurred in 124 (2%) patients, serious non-AIDS-defining [20] in 306 (5%), death (21 AIDS related, 132 non-AIDS related) in 153 (2%) and VF>50 copies/mL in 1,796 (27%). The median (Q1, Q3) CD4+ count at the time the AIDS-defining events occurred was 458 (228, 671) cells/µL.

The median CD4+ measurements in the follow-up from baseline were five (2, 12), with an average of 2.6 (2, 3.4) measurements/year, and the median prospective HIV-RNA measurements from baseline were five (2, 11), with 2.6 (2, 3.4) measurements/year.

Baseline CD4+ cell counts were 646 (611, 682)/µL in patients who started ART with ≤350 CD4+/µL and 757 (708, 807)/µL in those who started ART with >350 CD4+/µL (difference at baseline: 110.9 [50.4, 171.5] cells/µL; p<.001). Estimated yearly changes in CD4+ cell counts were 27.44 (-18.2, 73.12) cells/µL in patients who started ART with ≤350 CD4+/µL and 31.99 (-8.83, 72.81) cells/µL in those who started ART with >350 CD4+/µL (difference: 4.55 [-56.5, 65.64] cells/µL; p=0.884) (supplementary figure).

A confirmed CD4+ drop below 200 cells/µL was never observed; unconfirmed CD4+ drops below 200 cells/µL occurred in nine patients and were not associated with clinical events. The Kaplan-Meier estimate of the cumulative risk of unconfirmed dropping below 200 cells/µL was 0.24% (0.07%, 0.40%) by 4 years from baseline; no further events (drops) were observed over the following 9 years and 249 individuals were still at risk by year 12. Upper limits of the 97.5% CI of rates of confirmed CD4+ drop below 200 cells/µL were 0.28 and 0.38/1,000 PYFU for patients with ≤350 and >350 CD4+ cells/µL at ART start. Upper limits of the 97.5% CI of rates of confirmed CD4+ drop below 200 cells/µL according to type of ART started, HCV co-infection,

calendar year of baseline, baseline CD4/CD8 >0.3 or ≤0.3, time from HIV diagnosis and HIV-RNA at ART start > or ≤5 log10 copies/mL, are detailed in table 2. Upper limits of this CI were always below two per 1,000 PYFU. Only patients with calendar year baseline between 1997 and 2001 and those with a CD4+/CD8+ ≤0.3 at ART start had an upper limit of the 97.5% CI above one per 1000 PYFU. Results were similar when viral rebound was defined using the more conservative threshold of 400 copies/mL for the definition of VF (data not shown).

Discussion

We investigated the risk of CD4+ drop below 200 cells/μL in patients who started ART with >350 CD4+ cells/μL, or reached >350 CD4+ cells/μL after virological suppression, and we observed none of these events over a median follow-up of roughly four years, as long as virological suppression was maintained. We then tried to picture the worst scenario, by calculating the upper limit of the confidence interval of rates of CD4+ cells drop, we found that, at worst, this incidence is above 1 per 1,000 PYFU but always less than 2 per 1000 PYFU. This estimate is consistent with those previously found elsewhere. Among 1,820 HIV-infected patients in another cohort, those with HIV-1 RNA <200 copies/mL and CD4+ counts ≥300 cells/μL had a 97.1% probability of maintaining durable CD4+ counts ≥200 cells/μL for 4 years [14]. Similarly, in the ARTEMIS trial, only 1.1% of 449 patients with sustained HIV-1 RNA suppression below 400 cpies/mL dipped below 200 CD4+/μL on two consecutive visits [15]. Furthermore, in 7,250 patients in South Africa, after 10 years of ART, 92.9% of patients with ongoing virologic suppression maintained CD4+ cell counts continuously above 200 cells/μL [19]. In an Asian cohort, among 1,538 patients virologically suppressed over an unreported follow-up, the rate of confirmed dipping below 200 CD4+/μL was 0.77/100 patient-years. There was no significant difference in time to confirmed dipping below 200 CD4+/μL between biannual and annual CD4+ measurement [21]. Finally, in the PISCIS Cohort study, after a median follow-up of almost two years, CD4+ cell counts fell to <200 cells/μL in 7.4% of 8,695 patients [16]. However, these proportions reflect unconfirmed dipping below 200 CD4+/μL.

It can be argued that dipping below 200 CD4+/μL is not necessarily clinically relevant, because clinical events can occur also with counts of >200 cells/μL; however, routine CD4+ cell count monitoring did not have an impact on mortality rates among 39 283 HIV-infected patients with VLs <1000 copies/mL or CD4+ cell counts ≥350/μL beyond 12 months after ART initiation [13].

Monitoring CD4+ count might be only a source of patients' anxiety for a result that does not change treatment decision [7, 8]; reducing the frequency of CD4+ monitoring would also reduce costs related to care of HIV-infected patients [8, 17]. However, knowing when CD4+ drop below 200 cells/µL is important, because in these cases (re)starting prophylaxis of opportunistic infections is recommended [1-4]. In absence of randomized comparisons of CD4+ count monitoring strategies, our analysis provides solid estimates that should help designing such studies.

Caniglia and coll. tried to define the best timing for CD4+ cell counts monitoring in a very large cohort using an inverse probability of weighting approach to mimic a randomized trial, but most of the 39,029 eligible patients changed strategy during follow-up (mostly in the first six months): indeed, less than 1/10 of them did not change the initial strategy and could be maintained in the analysis over two years of follow-up, thus limiting the power of the study [10].

The results of our analysis confirm and expand, over a long follow-up, also in a context (i.e. the Italian) of universal, free access to ART, the results of previous analyses. They also lend support to optional monitoring of CD4+ cell counts in patients with satisfactory virological and immunological response to ART.

As the analysis was censored at virological failure, the results of this study apply only as long as virological suppression is maintained. It must be also underlined that we excluded from the analysis some immunological non-responders (i.e. those patients who started ART with ≤350 CD4+/µL and never attained values >350 cells/µL), because less frequent CD4+ cell counts monitoring is not an option in these patients; in other words, the results of this analysis do not apply to immunological non-responders.

The main limitations of this study are the relatively small sample size and the relatively short follow-up; however, all events occurred over the first 3 years, so it is unlikely that results will be different by repeating the analysis after further follow-up have been cumulated.

In summary, in patients who started ART in Italy with >350 CD4+ cells/µL, or reached >350 CD4+ cells/µL after VS, a confirmed CD4+ drop below 200 cells/µL was never observed as long as virological suppression was

maintained. The results of this study support optional monitoring of CD4+ cell counts, in a setting in which viral load assessment is easily available and in patients with these characteristics; they are also useful to help designing randomized trials comparing CD4+ count monitoring strategies in virologically suppressed populations.

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