Pregnancy may be followed by an inflexion of the immune reconstitution in HIV-infected women who receive antiretroviral drugs before conception.

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*see appendix.

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Running head: Increase of CD4 after pregnancy

Summary:

Background: Whether pregnancy has an impact on evolution of CD4+ cell counts in women treated with highly potent antiretrovirals before conception remains largely unknown.

Methods: Among patients enrolled in the ANRS CO8 (APROCO/COPILOTE) cohort, we selected all women aged between 18 and 50 years at initiation of combination antiretroviral therapy (cART). Slopes of CD4+ cell counts during follow-up were estimated using mixed longitudinal models with time-dependent indicators for pregnancy and delivery.

Results: Among the 260 selected HIV-infected women, a pregnancy occurred among 39 during a median follow-up of 66 months. Women who became pregnant had higher CD4+ cell count at baseline but this difference was progressively blurred during follow-up because they had a slower increase than women who did not become pregnant. The estimated slope of CD4+ cell count decreased significantly from +2.3 cells/mm^3/month before pregnancy and in women who did not become pregnant to - 0.04 cells/mm^3/month after delivery ($p = 0.0003$).

Conclusion: A significant increase in CD4+ cell count may be preferable before pregnancy in women treated with cART, in order to overcome the evolution observed after pregnancy.

Keywords: HIV infection; pregnancy; CD4 lymphocyte count; antiretroviral therapy, highly active

Introduction

Pregnancy is common in HIV-infected women treated with antiretroviral drugs. It has been shown that the first trimester of pregnancy is temporarily associated with a decline of CD4+ cell counts due to a decrease of total lymphocytes count and that these modifications reverse soon after delivery in HIV-infected women (1). Moreover, several studies have shown that pregnancy has no unfavorable impact on HIV disease progression in developed countries (2-6). However, data remain sparse about the impact of pregnancy on CD4+ cell counts changes in women treated with antiretrovirals before conception. We studied the association between pregnancy and evolution of CD4+ cell counts in a cohort of 260 HIV-infected women treated with combination antiretroviral therapy (cART).

Patients and methods

The ANRS CO8 cohort study (APROCO/COPILOTE) enrolled 1281 HIV-1-infected patients when they initiated a protease inhibitor-containing regimen in 1997-1999 (7). Patients are prospectively followed every 4 months. In the present analysis, we selected all women of childbearing age, i.e. aged between 18 and 50 years at baseline. Slopes of CD4+ cell counts changes during follow-up were estimated using mixed longitudinal models with time-dependent indicators for pregnancy and delivery. The evolution of CD4+ cell count was modelled by two slopes: before and after the first 4 months of treatment, based on previous analyses of the same cohort (8). Modelling with time-dependent covariates further allowed the estimation of three CD4+ cell count slopes after the first 4 months: (1) before pregnancy and in women who did not become pregnant during follow-up, (2) during pregnancy, (3) after delivery.

We then performed the same analysis with censoring of data at 36 months of follow-up. This sensitivity analysis was performed since CD4+ cell count increase reached a plateau at 36 months after initiation of cART in the patients of the whole cohort who had persistent complete virological response (9). Proportions of women having CD4+ cell count > 500/mm^3 or plasma HIV RNA < 500 copies/ml after 6 years of follow-up were compared according to the occurrence of a pregnancy during preceding follow-up using multivariate logistic regression. All statistical analyses were performed using Statistical Analysis System software 9.1 (SAS Institute Inc., Cary, NC, USA).

Results

A total of 260 women aged between 18 and 50 were enrolled in the cohort. During a median follow-up of 66 months, 39 women had a total of 43 pregnancies (incidence: 3.4/100 women-years). The median delay between baseline and first pregnancy was 31 months (interquartile range: 15-53). At baseline, compared to other women enrolled in the cohort, those who became pregnant at least once during follow-up were younger (median age 29 years vs 34; $p < 0.0001$), had a higher median CD4+ cell count (364/mm^3 vs 275/mm^3; $p = 0.02$) and tended to be more frequently born in Africa (31% vs 19%; $p = 0.12$) but were not different according to presumed risk factor for HIV (infection through intravenous drug use: 26%), co-infection with hepatitis C virus (30%) and plasma HIV RNA (median: 4.2 $\log_{10}$ copies/ml). The median proportion of follow-up spent on antiretroviral therapy was 99 % in women who became pregnant and 99 % in those who did not ($p = 0.69$). Antiretroviral therapy was temporarily interrupted during the first and second trimesters during only 7 pregnancies. Among all measurements of plasma HIV RNA performed during follow-up, 73 % were < 500 copies/ml in women who became pregnant and 79 % in others ($p = 0.57$). Though women who became

pregnant had higher CD4+ cell count at baseline, this difference was progressively blurred during follow-up since they had a slower increase than women who did not become pregnant (Figure 1). Based on modelling, CD4+ cell count remained stable but did not increase after delivery. The estimated CD4+ cell count slope after delivery (- 0.04 cells/mm^3/month; 95% confidence interval: -1.5 to + 1.4) was not significantly different from 0 and was significantly ($p = 0.003$) lower than the slope estimated before pregnancy and in women who did not become pregnant (+2.3 cells/mm^3/month; 95 confidence interval: +1.7 to +2.9) (Table 1). Results were similar after adjustment for baseline clinical stage and plasma HIV RNA, after exclusion of the 7 women who interrupted antiretroviral therapy at any time during pregnancy and when censoring data at the second pregnancy (data not shown). The results were also similar when data were censored at 36 months of follow-up (data not shown). This latter sensitivity analysis included 22 pregancies with a median follow-up of 10 months (IQR: 2-32) between delivery and the censoring date. In the subgroup of 116 women who had both CD4+ cell count and plasma HIV RNA measured after 6 years of follow-up, among whom 22 had a pregnancy during the preceding follow-up, the proportion of women with plasma HIV RNA < 500 copies/ml or CD4+ cell count > 500/mm^3 did not differ whether or not they had a pregnancy during before 6 years of follow-up (Table 2). No progression to AIDS or death after pregnancy were reported among women who became pregnant.

Discussion

In this cohort of 260 women of childbearing age treated with cART, CD4+ cell count were stable but did not increase after delivery. Nevertheless, this inflexion of the CD4+ cell count slope, i.e. this interruption in CD4+ cell increase after delivery, did not translate in long-term immunological response, since women who became pregnant had higher CD4+ cell count at

the initiation of protease inhibitor therapy. The long-term virological response was not affected by the occurrence of a pregnancy during follow-up.

A clear biological explanation for this apparent interruption of immune reconstitution following pregnancy in cART-treated women is lacking. It has been shown that cART partially reverses the Th1 to Th2 cytokine shift induced by pregnancy (10). Conversely, it may be hypothesised that pregnancy has an unfavorable effect on the Th2 to Th1 shift induced by cART in responding patients but this hypothesis deserves in vitro confirmation.

We acknowledge that our study has potential biases due to its observational design and the relatively small number of women. Moreover, part of the results we observed may be due to the plateau in CD4+ cell count increase after 3 to 4 years of complete virological response, we and others reported (9). Our sensitivity analyses by censoring data at 36 months of follow-up however suggest that this plateau is not the only explanation for our findings. Consequently, our findings do not imply a causal relationship between this apparent inflexion of CD4+ cell count increase and pregnancy and confirmatory analyses with a longer follow-up and in other cohorts and settings are needed. Although the quite small differences we observed between slopes of CD4+ cell counts according to occurrence of pregnancy might have little clinical relevance, we believe that our findings, along with other cautionary notes concerning the fetal risk associated with cART (11,12), contributes to balance the benefits and risks of pregnancy in HIV-infected women. More specifically, women who need antiretroviral drugs for their own health should be informed that a significant increase in CD4+ cell count, for example above 350/mm^3, may be preferable before begining a pregnancy.

Apppendix: the APROCO/COPILOTE (ANRS CO8) study group.

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Figure 1. Mean CD4+ cell counts during follow-up according to the occurrence of a pregnancy in 260 women aged 18-50 at enrollment in ANRS CO8 (APROCO/COPILOTE) cohort study.

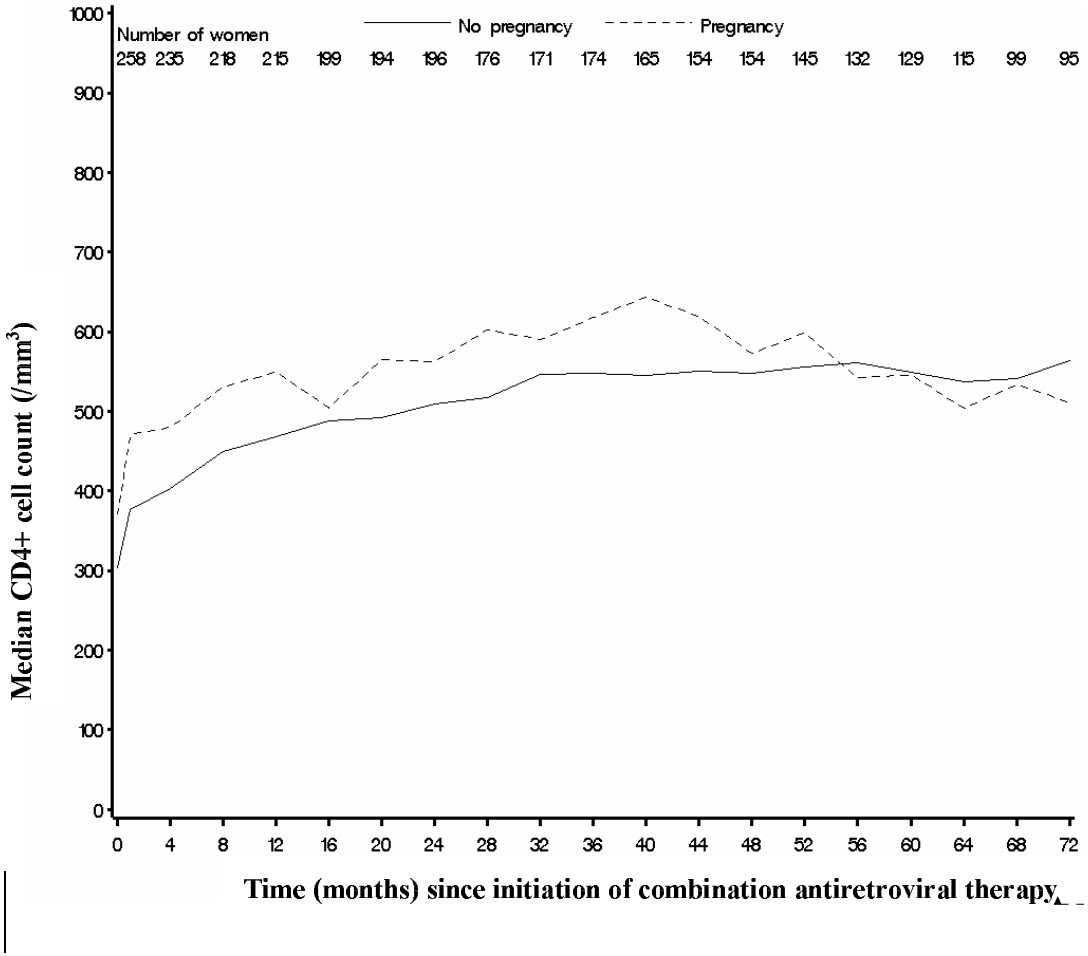

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Table 1. Modelling of CD4+ cell counts after the first 4 months of follow-up (M4) in 260 women aged 18-50 at enrollment in ANRS CO8 (APROCO/COPILOTE) cohort study.

	Estimated values (/mm^3/month)	95% CI
CD4 at M4 in women who became pregnant	488	[414;561]
CD4 at M4 in women who did not become pregnant	420	[387;452]
CD4 slope between M4 and before pregnancy in women who became pregnant and after M4 in women who did not become pregnant	+2.3	[+1.7; +2.9]
CD4 slope during pregnancy	-0.80	[-7.8;+6.3]
CD4 slope after delivery	-0.04	[-1.5;+1.4]

Abbreviations: CD4: CD4+ cell count; CI: confidence interval

Table 2. Immunological and virological response 6 years after initiation of combination antiretroviral therapy in 160 women aged 18-50 years at enrollment in ANRS CO8 (APROCO/COPILOTE) cohort study.

Response at 6 years	Pregnancy during follow-up n = 22	No pregnancy during follow-up n = 138	Adjusted odds-ratio*	p
Plasma HIV RNA < 500 copies/ml	73%	73%	1.09	0.88
CD4+ cell count > 500/mm^3	51%	52%	0.91	0.86

* Odds-ratio were estimated with multiple logistic regression models adjusted for naivete of antiretroviral drugs at baseline, baseline CD4+ cell counts, prescription of nelfinavir at baseline for virological response and for naivete of antiretroviral drugs at baseline and cumulative duration of antiretroviral therapy during follow-up for immunological response.