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Immune recovery after starting ART in HIV-infected patients presenting and not presenting with tuberculosis in South Africa

Michael Schomaker [1], Matthias Egger [1, 2], Mhairi Maskew [3], Daniela Garone [4], Hans Prozesky [5], Chris Hoffmann [6], Andrew Boulle [1], Lukas Fenner [2], for IeDEA Southern Africa

1 Centre for Infectious Disease Epidemiology and Research (CIDER), School of Public Health and Family Medicine, University of Cape Town, South Africa

2 Institute of Social and Preventive Medicine (ISPM), University of Bern, Switzerland

3 Health Economics and Epidemiology Research Office, Department of Internal Medicine, School of Clinical Medicine, Faculty of Health Sciences, University of the Witwatersrand, Johannesburg, South Africa

4 Khayelitsha ART Programme, Médecins Sans Frontières, Cape Town, South Africa

5 Division of Infectious Diseases, Department of Medicine, University of Stellenbosch and Tygerberg Hospital, Cape Town, South Africa

6 Aurum Institute, Johannesburg, South Africa

Correspondence:

Dr. Lukas Fenner

Institute of Social- and Preventive Medicine (ISPM), University of Bern,

Finkenhubelweg 11, CH-3012 Bern, Switzerland

Phone: +41 31 631 3867   Fax: +41 31 631 3520

E-Mail: lfenner@ispm.unibe.ch

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ABSTRACT

We studied the immune response after starting antiretroviral treatment (ART) in 15,646 HIV-infected patients with or without tuberculosis (TB) at presentation in three ART programs in South Africa between 2003-2010. Patients presenting with TB had similar increases in CD4 cells compared to all other patients (adjusted difference 4.9 cells/µl per six months, 95% CI 0.2-9.7). Younger age, advanced clinical stage, female sex, and lower CD4 cell count at ART start were all associated with steeper CD4 slopes. In South Africa HIV-infected patients presenting with TB experience immune recovery after starting ART that is no worse than in other patients.

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INTRODUCTION

In many resource-constrained settings tuberculosis (TB) is the most common AIDS-defining opportunistic infection (OI),[1] and is the leading cause of death in patients initiating antiretroviral treatment (ART) in sub-Saharan Africa.[2] TB and other OIs may play an important role in immune recovery after ART initiation.[3,4] HIV viral replication is enhanced by co-infection with *Mycobacterium tuberculosis* due to immune activation, which could in turn accelerate the depletion of CD4 cells by virus-induced immune destruction or apoptosis of lymphocytes.[5-7] TB treatment is associated with decreased immune activation, after an initial increase during the first month of treatment presumably resulting from release of mycobacterial antigens.[5]

A recent European study showed an impaired CD4 count recovery among HIV-infected patients presenting with TB at ART initiation.[4] There are limited data on the influence of different OIs on immune recovery after starting ART in sub-Saharan Africa and other resource-constrained settings.

We analyzed data from three ART programs in South Africa with access to routine CD4 cell count and HIV viral load monitoring to study the immune response after starting ART in adult patients with and without concurrent TB.

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METHODS

We analyzed data from adult patients starting ART between January 1, 2003 and December 31, 2010 in three South African treatment programs that participate in the International epidemiologic Databases to Evaluate AIDS in Southern Africa (IeDEA-SA, www.iedea-sa.org) [8] collaboration and systematically record OIs (Khayelitsha, Tygerberg and Themba Lethu Clinic). All sites provide integrated clinical care services for HIV and TB. Data are collected at each site as part of routine monitoring at enrollment and each follow-up visit. All study sites have local institutional review board or Ethics Committee approval to collect data and participate in IeDEA-SA. Inclusion criteria were age ≥16/<85 years, and no previous ART exposure. Virologically suppressed patients (<400 HIV RNA copies/ml) at ART initiation and patients with less than six months of follow-up were excluded. The selection of eligible patients is shown in the Supplemental Digital Content (see **Figure S1**, http://links.lww.com/QAI/A389).

A TB episode was defined as a diagnosis of TB (pulmonary/extrapulmonary) at the start of ART, as reported by each site. All three sites used CDC clinical and laboratory case definitions, however, whether the diagnosis of TB was confirmed or probable was not recorded in the database. To account for missing values of WHO clinical stage, CD4 cell count and viral load at baseline and during follow-up, we generated 10 imputed datasets using the Amelia II package in R.[9] The imputation model included age, sex, CD4 count and slope, viral load, observation time, WHO clinical stage, TB site, and treatment program.[10] Results were combined with Rubin's rules.[11] Patients were expected to have laboratory tests every six months for the purpose of determining missing follow-up values.

We used Chi-squared tests to test the statistical significance of differences between groups in binary variables, and the Wilcoxon rank sum test for continuous variables. We calculated CD4 slopes (defined as cells/μl/6 months) after starting ART up to four years of follow-up. We used linear mixed models to evaluate the effect of presenting with TB on slopes, adjusted for age, sex, CD4 count at ART start, current CD4 cell count, time on ART, WHO clinical stage at ART initiation, and treatment program. We further analyzed the effect of restricting the analysis to time under continuous viral suppression defined as time between ART initiation and first viral load rebound (≥1,000 copies/ml) or the last follow-up visit. We also modeled CD4 trajectories up to four years after starting ART by TB status, for two CD4 cell strata (<50, 50-199 cells/μl) using additive linear mixed models. Models included interactions of TB status with splines of time on ART and were adjusted for age, sex, WHO clinical stage, treatment site. Results for a male reference population aged 35-45 years with WHO clinical stage III from the Themba Lethu Clinic are shown.

All analyses were performed in Stata version 11.2 (Stata Corporation, College Station, USA) and R 12.1 (R Development Core Team, Austria).

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RESULTS

We included 15,646 adult patients with a median follow-up time of 710 days (interquartile range [IQR] 428-1075 days), and 624 days (IQR 364-967 days) when restricting analyses to time under viral suppression. A total of 10,622 (67.9%) patients were female, and 1,052 (6.7%) had a TB diagnosis. Median age at ART initiation was 34 years (IQR 29-40 years), median CD4 cell count 98 cells/µl (IQR 41-161 cells/µl) (see **Supplemental Digital Content, Table S1**, http://links.lww.com/QAI/A389). Patients presenting with TB had lower median CD4 cell counts (45 versus 102 cells/µl) and more advanced disease (WHO clinical stage IV 39.3% versus 26.9%) compared to all other patients, whereas distributions of age and sex were comparable between the two groups (see **Supplemental Digital Content**, **Table S1**, http://links.lww.com/QAI/A389).

Patients presenting with TB at ART start gained about five CD4 cells more per six months than patients without TB (adjusted difference 4.9 CD4 cells/µl/6months, 95% confidence interval [95% CI] 0.2-9.7, p=0.04, Table 1). During the first 6 months there was little evidence of a difference in CD4 count change (-3.3 CD4 cells, 95% CI -15.3 to 8.8, p=0.6). However, from six months onwards (up to 4 years of follow-up) TB patients were doing slightly better than patients without TB (adjusted difference 6.5 CD4 cells, 95% CI 2.0-11.1, p=0.01). The results did not substantially change when restricting the analysis to time under viral suppression (Table 1). The proportion of patients with viral load suppression at 12 months was similar between patients presenting and not presenting with TB (80.9% versus 79.1%, p=0.23).

Women had a more favorable CD4 cell response than men (adjusted difference 2.9 cells/µl, 95% CI 0.6-5.1, p=0.01), whereas the response was less favorable in older

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patients and patients with higher baseline CD4 cell counts (see **Supplemental Digital Content**, **Table S2**, http://links.lww.com/QAI/A389).

The results from the additive linear mixed model (Figure 1) showed that patients presenting with TB had a similar CD4 count recovery initially, but a slightly more favorable trajectory in the long-term, consistent with the results for the CD4 slopes. Results were similar for patients with a CD4 cell count below 50 cells/µl at ART start and patients with a CD4 cell count between 50-199 cells/µl. Immune recovery was also similar in patients with pulmonary TB (adjusted difference 5.9 cells/µl, 95% CI 0.3-11.5) and patients with extrapulmonary TB (adjusted difference 8.6 cells/µl, 95% CI 0.7-16.5), compared to patients without TB at ART start.

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DISCUSSION

We analyzed HIV-infected adult patients starting ART in three large HIV treatment programs in South Africa. We found that overall patients presenting with TB did not experience a worse CD4 cell count recovery after starting ART. The results were similar when restricting analyses to time under viral suppression.

Our results may be explained by several factors. First, TB infection causes immune activation which is associated with progression to more advanced disease.[12] Immune activation may lead to up-regulation of HIV replication, and as a result to CD4 depletion.[5] Therefore, TB treatment alone without ART has a positive effect on the immune system by reducing immune activation, maintaining stable HIV viral load and CD4 cell counts.[5;13;14] Second, a survival bias may exist as the most severely ill HIV co-infected TB patients with potentially impaired immune restoration may die before ART can be initiated. Therefore, a selection of patients with a better outcome may be reported in our dataset. Finally, patients with undiagnosed or unreported TB in the non-TB comparison group may present with an immune reconstitution syndrome, and therefore may have a rapid CD4 decline.[15]

Our finding extends those of a previous study from one of the included cohorts (the Khayelitsha ART program) which showed that receiving TB treatment at ART initiation was associated with an improved CD4 gain.[3] In contrast to the previous analysis, we used mixed linear models, modeled non-linear trajectories, and imputed missing values. Moreover, our analyses included a larger and more representative dataset. Another study from South Africa also failed to show a worse immunological outcome among patients with TB,[16] but the analysis was not restricted to time under viral suppression. We found no evidence for a worse immune recovery in patients with

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extrapulmonary TB. Finally, we also demonstrated that the immune recovery was poorer in men and older patients, as previously shown.[3;17] Interestingly, our study contrasts with a recent report from a high-income setting which showed that patients presenting with TB had impaired CD4 cell count responses to ART.[4] The impaired immune recovery was mainly driven by a delay in achieving viral suppression.[4] While studies from high-income settings suggested a decrease in viral load after TB treatment in patients with or without additional ART,[18;19] we and others failed to show such an effect in sub-Saharan settings in patients receiving TB treatment on ART.[20;21] The situation in European countries, however, differs from settings in sub-Saharan Africa with regard to the public health systems, TB screening practices, clinical follow-up, and patient characteristics.

The favorable CD4 cell count immune recovery in patients with concurrent TB remained even when restricting analyses to time under viral suppression. Viral suppression is indicative of adherence to ART. HIV-infected patients on TB treatment may be more adherent to ART, particularly when TB treatment is directly observed. Similarly, the provision of cotrimoxazole or isoniazid preventive therapy has been shown to improve retention in care among pre-ART patients.[22;23] However, we found no evidence, as assessed by HIV RNA suppression, for an effect of adherence.

A limitation of our study was the potential under-ascertainment of TB as shown by the low prevalence of TB cases, and the lack of a uniform case definition. The under-ascertainment of TB may have masked a worse immune recovery after starting ART in patients with TB compared to patients starting without known TB. However, we restricted our study to cohorts that systematically collect information on OIs. Our study may also have been influenced by OIs that occurred during follow-up, but results were similar when restricting analyses to time under viral suppression during which an OI episode is unlikely. Another limitation was the heterogeneity between the included ART

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programs.[24] To address this, we calculated relative differences adjusted for the most important confounding factors.

In summary, we found that HIV-infected patients presenting with TB did not have a worse immune recovery after ART initiation compared to other patients. This is possibly due to a favorable long-term effect of the TB treatment, or factors not captured in our dataset, such as more frequent counseling offered to this patient group or more frequent clinical visits. We stress that immune recovery does not necessarily mean that CD4 cell function is fully restored.[25] Our study underscores the benefit of co-treatment of HIV and TB, and indicates that management and integration of services may have a positive long-term effect on the immune recovery in resource-constraint settings.

Our study needs to be confirmed in observational data from HIV cohorts with a high degree of diagnostic ascertainment of OIs. Further studies need to focus on integrated TB-HIV management and its effect on OI incidence and immune recovery after ART initiation.

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Figure 1. Modeled CD4 trajectories defined as CD4 cell count increase from baseline by tuberculosis (TB) status at ART start, according to CD4 cell count at the start of ART of CD4 cell count <50 (A), and CD4 cell count between 50-199 (B).

Table 1. Multivariate associations with CD4 cell count changes after starting antiretroviral treatment (ART) comparing HIV-infected patients presenting with TB at ART start compared to all other patients. Results are presented as coefficient (CD4 cell count/µl/6 months), overall and restricted to time under viral suppression (HIV RNA viral load <1,000 copies/ml).

Characteristic	Crude			Adjusted		
	Coefficient	(95% CI)	*P* value	Coefficient	(95% CI)	*P* value
Unrestricted						
Overall	-5.35	(-10.67 to -0.02)	0.049	4.94	(0.20-9.68)	0.042
First 6 months after ART start	-32.97	(-46.30 to -19.64)	<0.0001	-3.25	(-15.32 to 8.81)	0.58
>6 months after ART start	0.20	(-5.06 to 5.46)	0.94	6.51	(1.97-11.06)	0.006
Restricted to time under viral suppression						
Overall	-7.08	(-12.56 to -1.60)	0.012	4.89	(0.06-9.73)	0.048
First 6 months after ART start	-38.86	(-52.19 to -25.53)	<0.0001	-1.29	(-13.47 to 10.87)	0.83
>6 months after ART start	0.05	(-5.39 to 5.50)	0.99	6.22	(1.53-10.91)	0.010

Models were adjusted for age, sex, CD4 cell count at ART start, current CD4 cell count, time on ART, WHO clinical stage, and treatment site. An interaction term between time on ART and presenting with TB at ART start was included in the model.

ART, antiretroviral therapy; TB, tuberculosis; 95% CI, 95% confidence interval

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