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Short Communication

Rapid decrease in IL-1Ra and IP-10 plasma levels following tuberculosis treatment initiation

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ABSTRACT

Objectives: Monitoring tools that could provide quick predictions of tuberculosis (TB) treatment outcomes are urgently needed. Here, we assessed whether the evolution of selected biomarkers of innate immunity may help monitoring TB treatment response within 2 weeks of treatment initiation.

Methods: ANRS12394-LILAC-TB was a proof-of-concept prospective study: adults with a rifampicin-susceptible TB who are HIV-negative and HIV-infected documented by a positive Xpert MTB/RIF test were enrolled in Cambodia and Côte d'Ivoire. Plasma concentrations of interleukin-1 receptor antagonist (IL-1Ra), interferon-γ–induced protein-10 and clusters of differentiation (CD) (scavenging CD163) were measured by commercial enzyme-linked immunosorbent assay kits. A Wilcoxon test for paired data was used for longitudinal comparisons.

Results: A total of 55 patients were enrolled (women: 31%, median age: 37 years; median CD4 count in the 10 of 13 participants with HIV: 53 cells/mm^3). Overall, 83% were considered in TB treatment success. Compared with baseline, the IL-1Ra plasma levels significantly decreased as soon as week (W) 1, independent of HIV status (−71% in HIV-positive vs −33% in HIV-negative; P <0.001). The IP-10 plasma levels significantly decreased at W1 and W2 compared with baseline (P <0.0001); however, that decrease was less marked in participants with HIV.

Conclusions: Our findings suggest that measuring IL-1Ra plasma levels with a standard enzyme-linked immunosorbent assay technique at baseline and then 1 week after TB treatment onset could help clinicians to quickly assess TB treatment response.

Introduction

Tuberculosis (TB) remains a major public health issue, with 10.6 million incident cases and 1.6 million deaths in 2022. The lack of microbiologically confirmed TB is frequent, notably in resource-limited settings and in people with advanced HIV disease. This often results in initiating empirical TB treatment, commonly used

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when the diagnosis is highly suspected. In such situations, clinicians need treatment monitoring tools that could provide quick and accurate predictions of treatment outcomes [1]. Thus, documenting an early decrease of biomarkers correlating with treatment response would be particularly useful in patients empirically treated for TB.

Monocytes and macrophages are innate immune cells that play a critical role in the pathogenesis of TB by initiating inflammatory response at the early stage of infection. Activated monocytes and macrophages release soluble forms of receptors, including a hemoglobin-haptoglobin scavenging receptor clusters of differentiation (CD) 163 (sCD163), and cytokines, such as the interleukin (IL)-1 receptor antagonist (IL-1Ra), a naturally occurring competitive inhibitor of the pro-inflammatory cytokines IL-1α and IL-1β [2]. Elevated concentrations of sCD163 have been reported in individuals infected with HIV and CD163 concentrations were suggested to predict mortality in patients with TB, independent of HIV status [3]. Interferon-γ–induced protein 10 (IP-10 or CXCL-10), a pro-inflammatory chemokine involved in trafficking immune cells to inflammatory sites, was reported to be a promising biomarker to assess early TB treatment response in patients with HIV-TB co-infection [4]. In a previous pilot study conducted in Cambodia, we reported high levels of circulating IL-1Ra at the time of TB diagnosis in severely immunosuppressed adults with HIV and a spectacular decrease of IL-1Ra after 8 weeks of TB treatment [5]. Unfortunately, 8 weeks is too late to be of real clinical interest. Clinicians need to be able to determine much more quickly whether a treatment given empirically is effective. Having that in mind, we assessed whether some soluble inflammatory biomarkers decreased within the first 2 weeks of treatment in patients with TB mono-infection and TB-HIV co-infection.

Methods

Study design and participants

LILAC-TB (ANRS 12394) was a proof-of-concept study: adults with and without HIV with a positive Xpert MTB/RIF test were enrolled in Cambodia and Côte d'Ivoire. Patients with HIV were anti-retroviral therapy–naïve, regardless of CD4 cell counts. Non-inclusion criteria included rifampin-resistant TB, as evidenced by Xpert MTB/RIF, and overt evidence of other ongoing infections. All patients received standard TB treatment according to the national guidelines.

After written consent, EDTA-plasma samples were collected and frozen to measure IL-1Ra, sCD163, and IP-10 at baseline (D0: TB treatment initiation), then at week (W)1, W2, W4 and W8 after TB treatment initiation. All participants were followed for 24 weeks to document TB treatment response. The primary endpoint was the change of IL-1Ra plasma concentration between baseline and W2. Secondary endpoints included the evolution of IL-1Ra at other timepoints and the evolution of IP-10 and sCD163 at all timepoints compared to baseline. The study was registered at ClinicalTrials.gov (NCT04015713).

Measurements of plasma biomarkers

Plasma samples were assessed after thawing for concentrations of IL-1Ra, IP-10, and sCD163 using commercial enzyme-linked immunosorbent assay (ELISA) kits (Human Quantikine ELISA kit, R&D system, Minneapolis, MN, USA). Each test was performed in duplicate. A single lot of ELISA kits was used in both countries to eliminate variability between lots. Quantikine Immunoassay controls for each biomarker were added in all experiments. For each participant, longitudinal plasma samples were assessed during the same experiment.

Table 1
Baseline and follow-up patients' characteristics (N = 55).

Age (median; IQR)	37 (27-53)
Male (numbers; %)	38 (69%)
Female (numbers; %)	17 (31%)
HIV-positive	13 (24%)
Clusters of differentiation 4 counts (for HIV+) (median; IQR) n = 10	53 (27-59)
TB characteristics (numbers; %)	
PTB[a]	53 (96%)
Extra PTB	2 (4%)
Country (numbers; %)	
Cambodia	28 (51%)
Ivory Coast	27 (49%)
TB outcome (numbers; %)	
Cured/Treatment success	42 (76%)
Treatment completed	4 (7%)
Treatment failed	3 (5%)
Died	2 (4%)
Lost to follow-up	0
Not evaluated	4 (7%)

IQR, interquartile range; PTB, pulmonary TB; TB, tuberculosis.
[a] Including five patients with PTB and extrapulmonary localization

Statistics

All participants were included in the analyses. To compare the plasma levels between follow-up visits and baseline, a Wilcoxon test for paired data was used. To compare plasma levels at baseline between HIV status groups, a Kruskal–Wallis test was used. Data analysis was performed using SAS software, version 9.4.

Results

Study population

Between January 2020 and May 2021, 55 patients were enrolled, including 27 participants in Côte d'Ivoire and 28 in Cambodia. Patients' characteristics are summarized in Table 1. A total of 83% of participants were considered in TB treatment success.

Biomarker measurements

Biomarker measurements were available for 52 participants. At baseline, plasma levels of IL-1Ra, IP-10, and sCD163 were significantly higher in participants who are HIV-infected than HIV-negative ones ($P = 0.0005$, $P = 0.0003$, and $P = 0.0436$, respectively). Among all participants, IL-1Ra plasma levels significantly decreased at W2 compared with baseline ($P = 0.0001$). Interestingly, the decrease of IL-1Ra was also significant at W1 ($P < 0.0001$) (Figure 1a). That decrease was significant in participants who are HIV-negative and HIV-infected (Figure 1b and 1c). Of note, the median decrease was more important in participants who are HIV-infected than participants who are HIV-negative (−71% vs −33% at W1 and −73% vs −21% at W2) (Figure 1d).

Among all participants, IP-10 plasma levels significantly decreased at W2 compared with baseline ($P < 0.0001$). That decrease was also significant from W1 ($P < 0.0001$) (Figure 1e). It was significant at W1 ($P < 0.0001$) and W2 ($P < 0.0001$) in participants who are HIV-negative and only at W1 in participants who are HIV-infected ($P = 0.0420$ at W1 and $P = 0.3203$ at W2) (Figure 1f and 1g). It was similar in participants with and without HIV at W1 (−48%) but less marked (−30%) in participants with HIV at W2 (Figure 1h). The decrease in IL-1Ra and IP-10 plasma levels was sustained until W8 (Table S1). Soluble CD163 levels did not change within the first 8 weeks of TB treatment (data not shown).

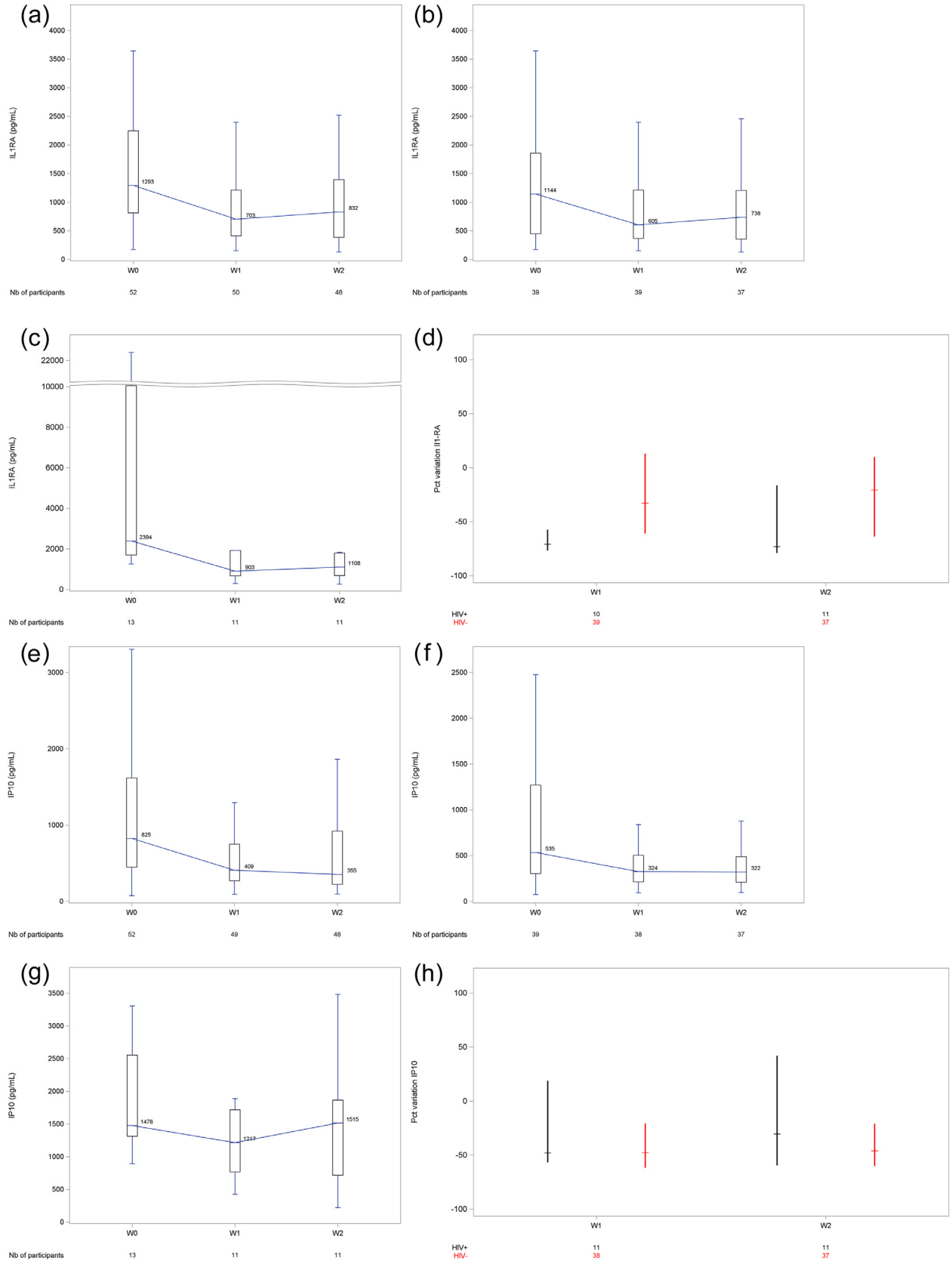

Figure 1. Longitudinal follow-up of IL-1Ra and IP-10 plasma levels from initiation of TB treatment (W0) to W2 of treatment and proportion of variation of both markers in participants with TB mono-infection and HIV-TB co-infection. Box plots of plasma IL-1Ra and IP-10 concentrations (pg per ml) are presented as median, IQR, and 1.5 IQR at initiation and after 1 and 2 weeks of TB treatment in all participants with TB (panels a and e), participants who are HIV-negative (panels b and f), and participants with TB/HIV coinfection (panels c and g). Panels d and h depict the percentages of variation (as median ± IQR) in the plasma levels of IL-1Ra and IP-10 during the follow-up under TB treatment in all participants according to their HIV status (black bars for participants who are HIV-positive and red bars for participants who are HIV-negative). IL-1Ra, interleukin-1 receptor antagonist; IP-10, interferon-γ–induced protein-10; IQR, interquartile range; TB, tuberculosis; W, week.

Discussion

This study demonstrates that IL-1Ra and IP-10 plasma levels significantly decreased as early as the 1st week of TB treatment. Most studies assessed the evolution of biomarkers after 8 weeks of TB treatment [6]. We have previously reported a spectacular decrease in IL-1Ra levels after 8 weeks of TB treatment in severely immunosuppressed patients with HIV-TB co-infection [5]. Nosik et al. [7] also reported a significant decrease in IL-1Ra plasma levels during TB treatment; however, they also assessed it after 45-60 and 180 days of TB treatment. Our present study focused on the early evolution of biomarkers after TB treatment onset. We observed that the decrease in IL-1Ra plasma levels was significant as soon as W1 in participants with and without HIV. That decrease was more substantial (approximately −70%) in participants with HIV than those without HIV. IL-1 plays a critical protective role in the early immune response during *Mycobacterium tuberculosis* infection. By blocking IL-1R1 and IL-1R2 receptors, IL-1Ra inhibits IL-1 signaling and prevents inflammation, which may result from an excess of IL-1 [8]. IL-1Ra expression and regulation are also important during mycobacterial-elicited granuloma formation.

Regarding IP-10, our results are in line with those of García-Basteiro et al. [4] who reported, in a study with patients with HIV with presumptive TB, that a substantial decrease in IP-10 levels during the 1st week of TB treatment was associated with a subsequent bacteriological TB confirmation. Several reports highlighted the interest of IP-10 to monitor TB treatment response in individuals without HIV. However, in most studies, plasma levels were assessed late, i.e. after 2 months of treatment [9]. In our study, although the decrease in IP-10 levels was significant at W1 and W2 in participants who are HIV-negative, it was less marked in participants who are HIV-infected. Thus, IP-10 seems less promising to monitor early TB treatment response in patients with HIV. Other markers seem promising to monitor TB treatment response, including the RISK6 transcriptomic signature [10]. However, it must be translated into point-of-care devices before potential use in routine care.

Our findings suggest that IL-1Ra plasma concentration changes measured with a standard ELISA technique when initiating TB treatment and then 1 week later could help clinicians to quickly assess treatment response in patients empirically treated for TB. In such individuals, the next steps should include the investigation of plasma concentration changes to guide interventions such as active research of alternative diagnoses in those with a lack of rapid decrease.

Declaration of competing interest

The authors have no competing interests to declare.

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Ethical statement

The study was approved by the Cambodian National Ethics Committee for Human Research (Nº 139 NECHR) and the Côte d'Ivoire National Ethics Committee for Health Research (Nº 095-19/MSHP/CNESVS-kp).

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Author contributions

LW, DL, DG, PP, RA, and XA designed the study. PP, RA, BCS, BD, and LB collected demographic and clinical data, samples, and carried out the biomarker dosages. DG and CC performed statistical analysis. LW, GC, PP, FXB, and DL analyzed data and drafted the first version of this manuscript. All other authors read and approved the final manuscript.

Supplementary materials

Supplementary material associated with this article can be found, in the online version, at doi:10.1016/j.ijid.2024.107096.

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