IL-1β Blockade Reprograms Antiviral Immunity and CD8+ T-cell Responses to Reduce the HIV Reservoir

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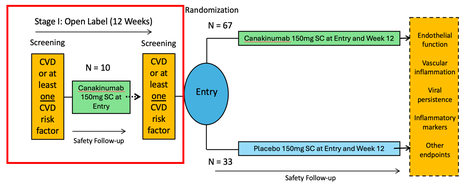
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Background and Hypothesis

- Despite effective viral control by Antiretroviral therapy (ART), PLWH exhibit chronic immune dysfunction including heightened inflammatory responses that can contribute to developing co-morbidities
- IL-1β is a potent inflammatory cytokine known to be a key mediator of acute and chronic inflammation in HIV
- IL-1β also mechanistically mediates the development of cardiovascular disease (CVD), a major co-morbidity in PLWH
- Despite effective and approved clinical therapies targeting IL-1β, nothing is known about the potential impact of blocking IL-1b on reversing immune dysfunction and reducing CVD in PWLH
- We hypothesize that IL-1β blockade reduces chronic inflammation and permits enhanced antiviral immunity, creating an environment permissive to HIV reservoir reduction.

STUDY COHORT



- Ten ART-suppressed PWH with at least one cardiovascular risk factor were enrolled.
- Participants received a single 150 mg subcutaneous dose of canakinumab.
- Plasma and PBMCs were collected at three timepoints: baseline (Week 0), Week 4, and Week 8 post-treatment.
- HIV-DNA reservoir was quantified as copies per 10^6 CD4+ T cells.
- Multi-omics profiling was performed including plasma cytokine analysis (22 analytes), sorted CD4+ T cell bulk RNA-seq, and single-cell RNA-seq of PBMCs.
- Six participants demonstrated reservoir reduction (Responders) while four showed no change or increase (Non-Responders).

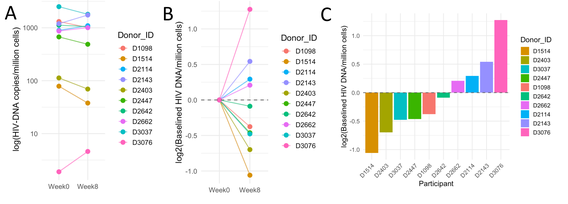


Figure 1: HIV Reservoir Response to Canakinumab. A) HIV-DNA copies per million CD4+ T cells at Week 0 and Week 8 for each participant, showing variable baseline reservoir sizes and heterogeneous treatment responses. B) Log2 baselined HIV-DNA fold change from Week 0 to Week 8, with the dashed line representing no change. Lines converging below zero indicate reservoir reduction. C) Waterfall plot of log2 baselined HIV-DNA per participant ordered by magnitude of change. Six of ten participants demonstrated reservoir reduction (Responders) while four showed no change or increase (Non-Responders).

Therapeutic blockade of the inflammatory cytokine IL-1β, using the monoclonal antibody Canakinumab, in PWHIV and CVD resulted in *reversal of pathways of chronic immune dysfunction* and *improved antiviral and cytotoxic effector immune responses*

Results

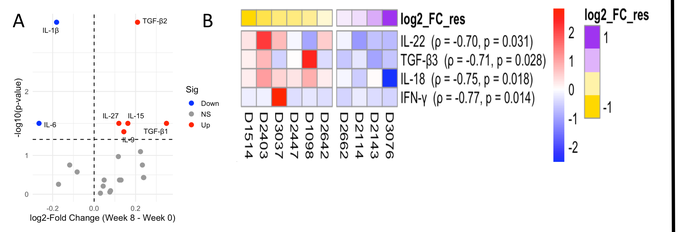


Figure 2: Canakinumab Modulates Plasma Cytokines and Cytokine Changes Track with Reservoir Reduction. A) Volcano plot of plasma cytokine changes (Week 0 vs Week 8) showing significant reduction in IL-1β (p = 0.002) and upregulation of TGF-β2. B) Heatmap of Spearman correlations between cytokine and HIV-DNA changes. Reductions in IFN-γ (ρ = -0.77), IL-18 (ρ = -0.75), TGF-β3 (ρ = -0.71), and IL-22 (ρ = -0.70) were significantly associated with reservoir reduction (all p < 0.05).

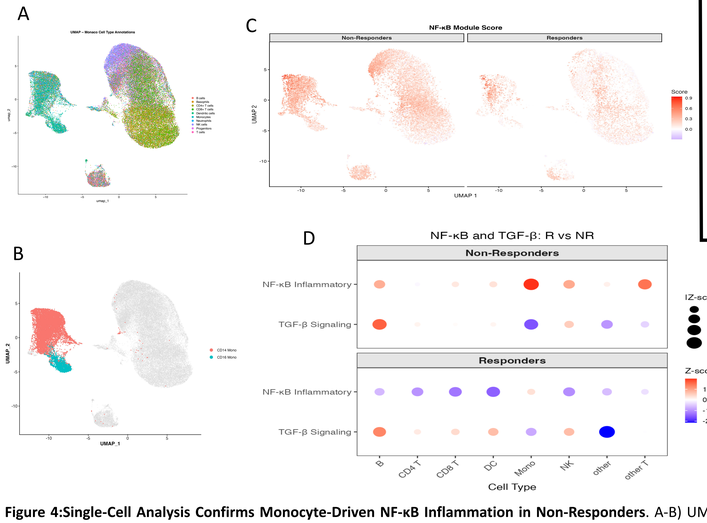


Figure 4:Single-Cell Analysis Confirms Monocyte-Driven NF-κB Inflammation in Non-Responders. A-B) UMAP of PBMCs from single-cell RNA-seq colored by cell type. C) NF-κB module scores on UMAP split by Responders and Non-Responders, showing higher NF-κB signaling in Non-Responders, particularly in monocytes. D) NF-κB and TGF-β pathway scores across cell types confirming elevated inflammation in Non-Responders and enhanced TGF-β signaling in Responders. E) Gene-level expression of NF-κB pathway genes in monocyte subsets showing Non-Responders have markedly higher expression of FOSB, JUN, FOSL1, IER3, BCL3, and IL1B in CD14+ classical monocytes. These data pinpoint monocyte-driven NF-κB inflammation as a key barrier to reservoir clearance.

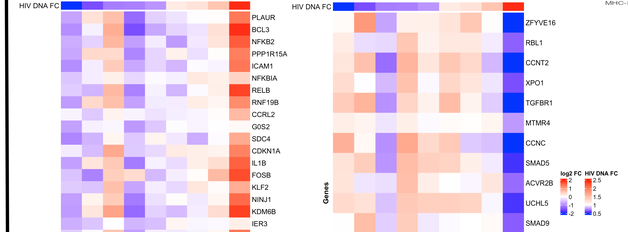


Figure 3: NF-κB Signaling Is Upregulated and TGF-β Signaling Is Downregulated in Non-Responders. A) Heatmap of NF-κB signaling gene expression changes in sorted CD4+ T cells across donors ordered by HIV-DNA fold change. Non-Responders showed upregulation of key NF-κB genes including PLAUR, BCL3, NFKB2, ICAM1, RELB, IL1B, FOSB, and JUN. B) Heatmap of TGF-β signaling gene expression showing downregulation of TGFBR1, SMAD5, SMAD9, and SMAD4 in Non-Responders. Together these data demonstrate that persistent NF-κB inflammation coupled with impaired TGF-β signaling characterizes participants who failed to reduce the reservoir.

Results

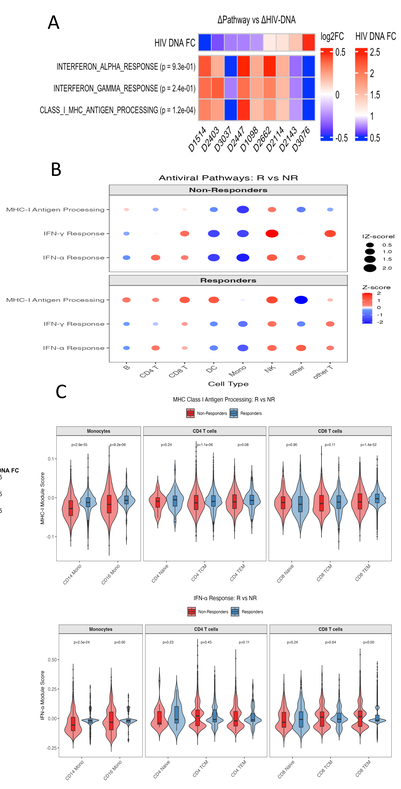


Figure 5: Responders Develop Enhanced Antiviral Immunity in the HIV Reservoir Compartment. A) Heatmap of antiviral pathway changes associated with HIV-DNA change showing IFN-α response, IFN-γ response, and MHC Class I antigen processing (p = 1.2e-04) significantly associated with reservoir reduction. B) Antiviral pathway scores (MHC-I, IFN-α, IFN-γ) between Responders and Non-Responders across cell types showing enhanced antiviral activity in Responder T cells. C) Violin plots of MHC-I and IFN-α module scores across monocyte, CD4+ T cell, and CD8+ T cell subsets confirming significantly elevated MHC Class I in Responder CD4+ central memory T cells (p = 1.1e-06), a primary HIV reservoir compartment. Bulk and single-cell data converge to show IL-1β blockade permits enhanced antiviral immunity specifically in cells harboring HIV.

Conclusions and Future Directions

- A single dose of canakinumab reduced the HIV-DNA reservoir in 60% (6/10) of ART-suppressed PWH.
- Reservoir reduction was associated with suppressed NF-κB/IL-1β-driven monocyte inflammation and enhanced TGF-β signaling.
- Responders demonstrated coordinated upregulation of MHC Class I antigen processing and interferon responses in CD4+ memory T cells.
- Non-Responders maintained persistent monocyte-driven NF-κB activation (FOSB, JUN, IL1B, BCL3).
- Future Directions: Validation in larger cohorts, evaluation of combination strategies with latency-reversing agents, and deeper characterization of monocyte-T cell crosstalk governing reservoir persistence.