The Tuberculosis Associated Microenvironment Reduces CD8+ T- Cell Control of HIV at the Site of the Coinfection

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CONFLICTS OF INTEREST

No conflicts to declare

Community Summary

The Problem:

Coinfection with tuberculosis in people living with HIV results in worsened clinical outcomes, but the mechanisms behind this are poorly understood

Our Research:

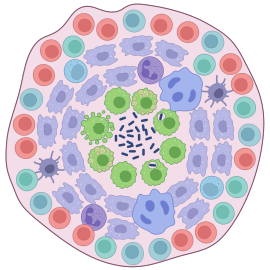
We have genetically analysed HIV sequences in people experiencing coinfection, as well as the functionality of CD8+ T cells at the site of the coinfection

Why it matters:

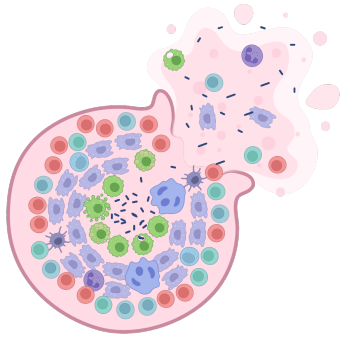
We need a deeper understanding of the immunological effects of coinfection to inform immune-mediated curative approaches and to improve clinical outcomes

Mycobacterium tuberculosis and HIV Coinfection

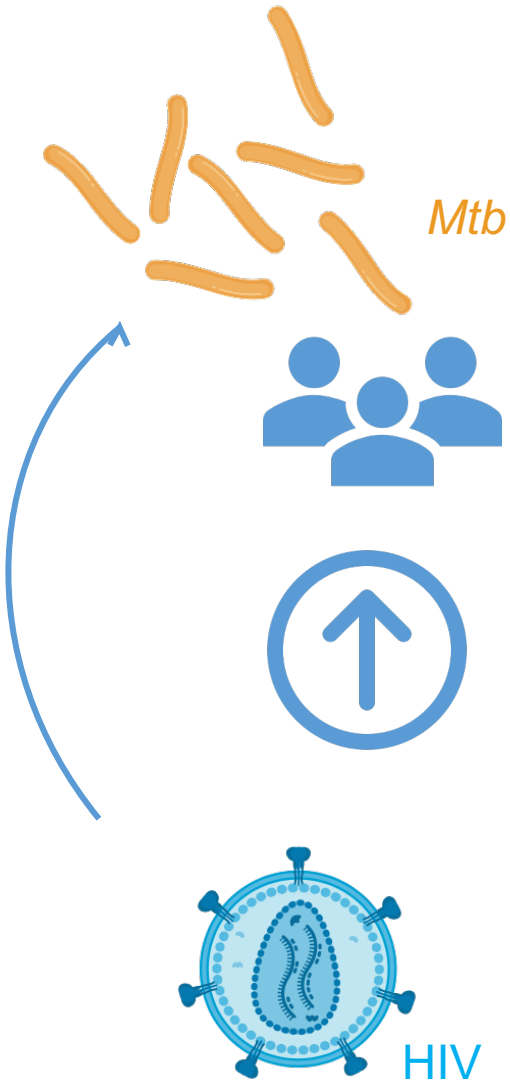
Mycobacterium tuberculosis (Mtb) is a bacterial infection which primarily affects the lungs



90% of *Mtb* infections enter a latent, non-infectious state where the bacteria is contained in a granuloma (Gideon & Flynn, 2013)



10% of latent *Mtb* infections will reactivate because the granuloma breaks down (WHO, 2024)



14 million people live with both HIV and TB (Getahun et al., 2010)

People living with HIV are **18 times** more likely to experience active TB disease (WHO, 2020)

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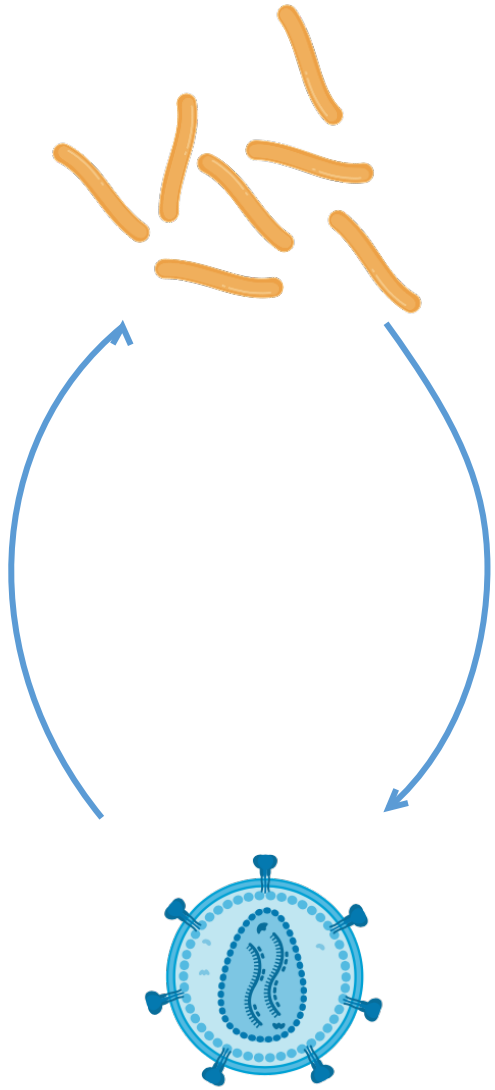

25% of HIV related deaths are attributed to tuberculosis disease (WHO, 2024)

Little is known about the effect of TB disease on HIV persistence

Studying the effects of Concurrent TB on HIV Persistence

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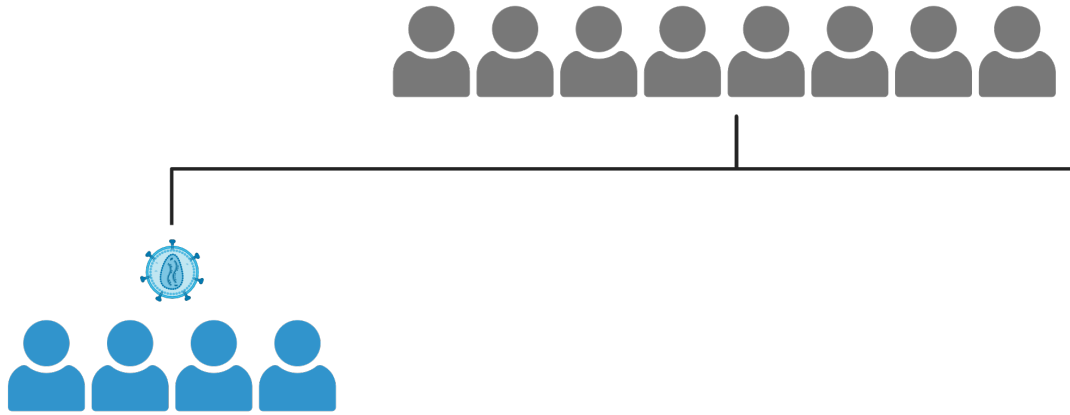


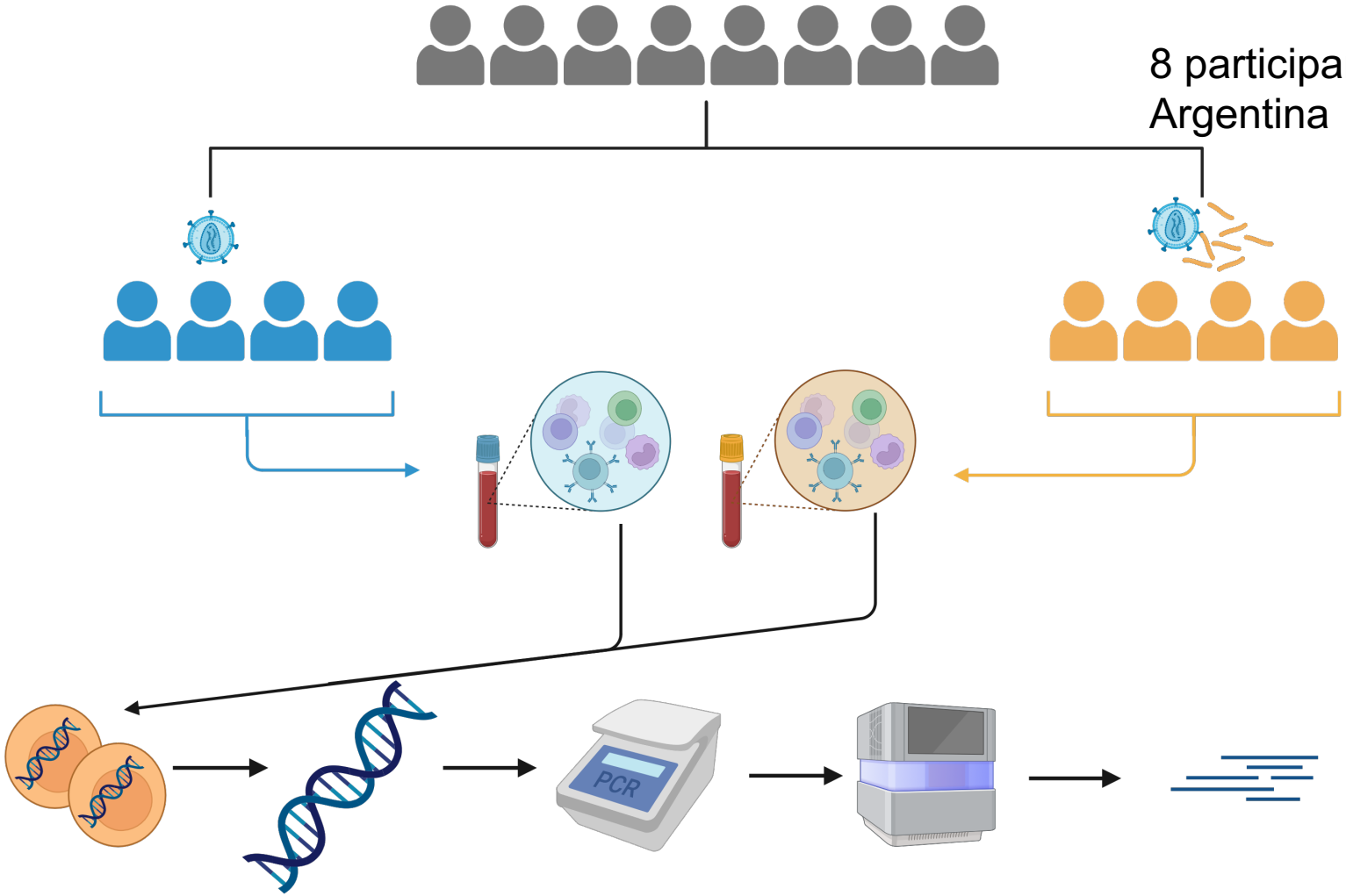
8 participant cohort from Argentina

4 participants living with HIV

4 participants living with HIV and *Mtb*

Parameter	HIV Only	TB-HIV
Mean age (range)	30.25 (24-37)	35.5 (30-43)
Male (percentage)	1 (25%)	3 (75%)
Mean time on ART in months (range)	2.5 (1-3)	2.33 (1-4)* *Note: 1 participant had been on ART for 8 years but had a detectable viral load at the time of sample collection
Mean viral load (range)	557.75 (0-1136)	836 (257-1180)
Mean CD4 count (range)	421.49 (367-467)	235 (109-362)

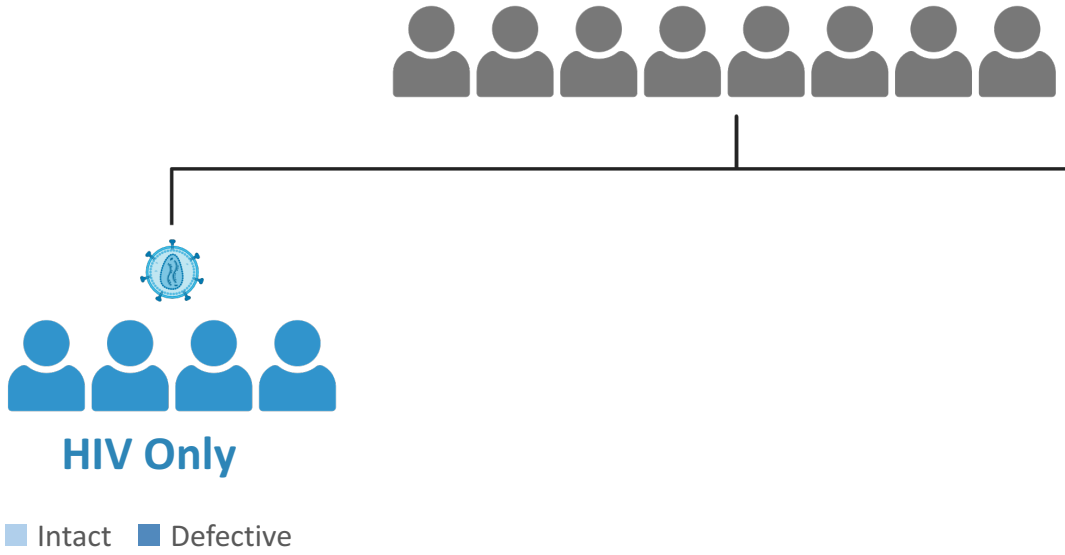
Studying the effects of Concurrent TB on HIV Persistence

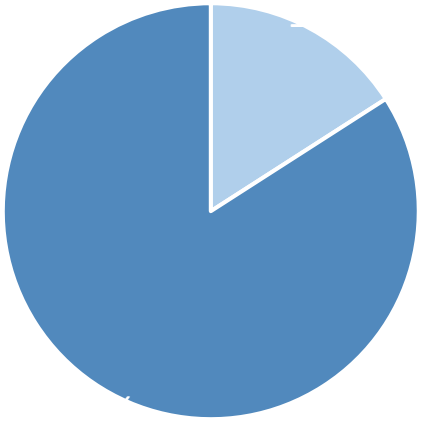


8 participant cohort from Argentina

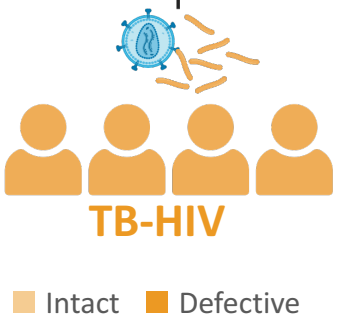
Hiener et al., 2017: Identification of Genetically Intact HIV-1 Proviruses in Specific CD4⁺ T Cells from Effectively Treated Participants

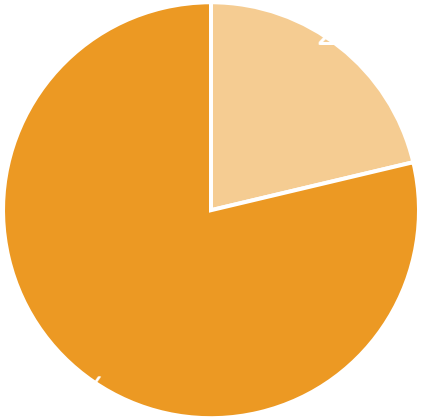
The Systemic Effect of Concurrent TB on HIV Persistence





Sequences: 157





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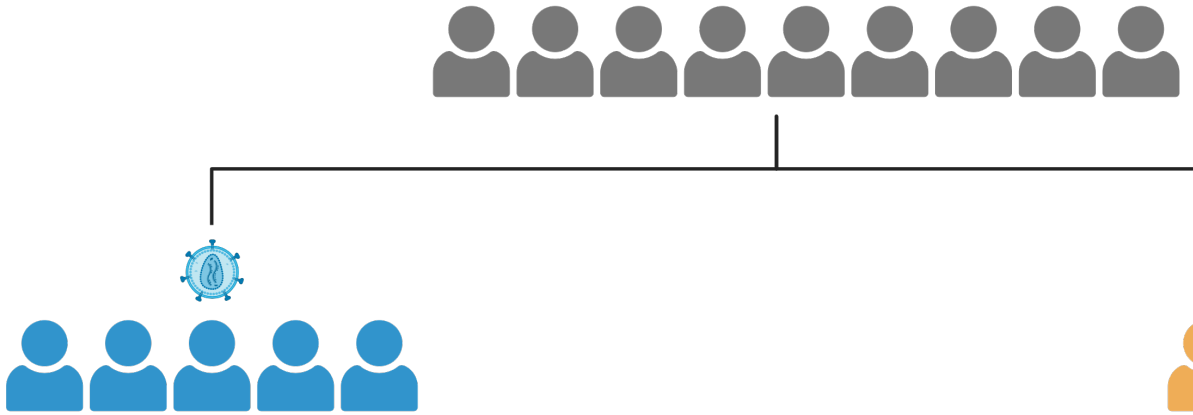
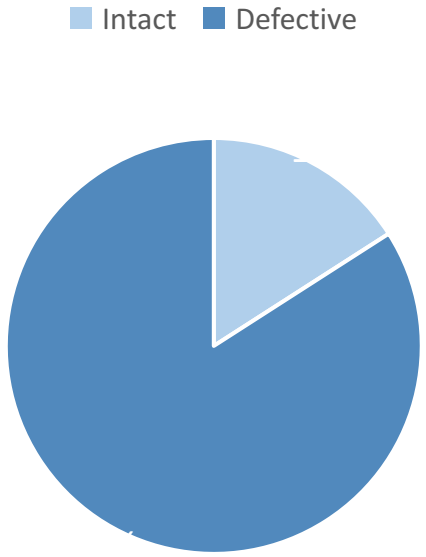
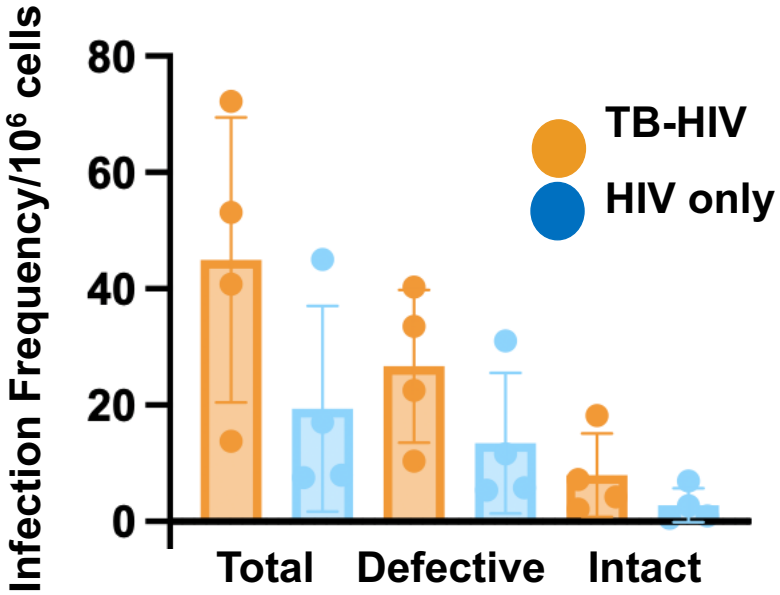
The Systemic Effect of Concurrent TB on HIV Persistence

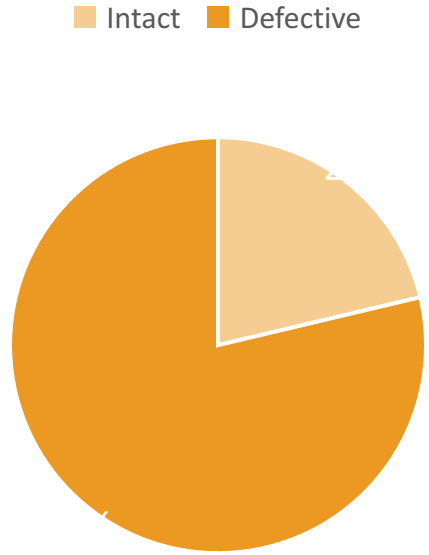


HIV Only

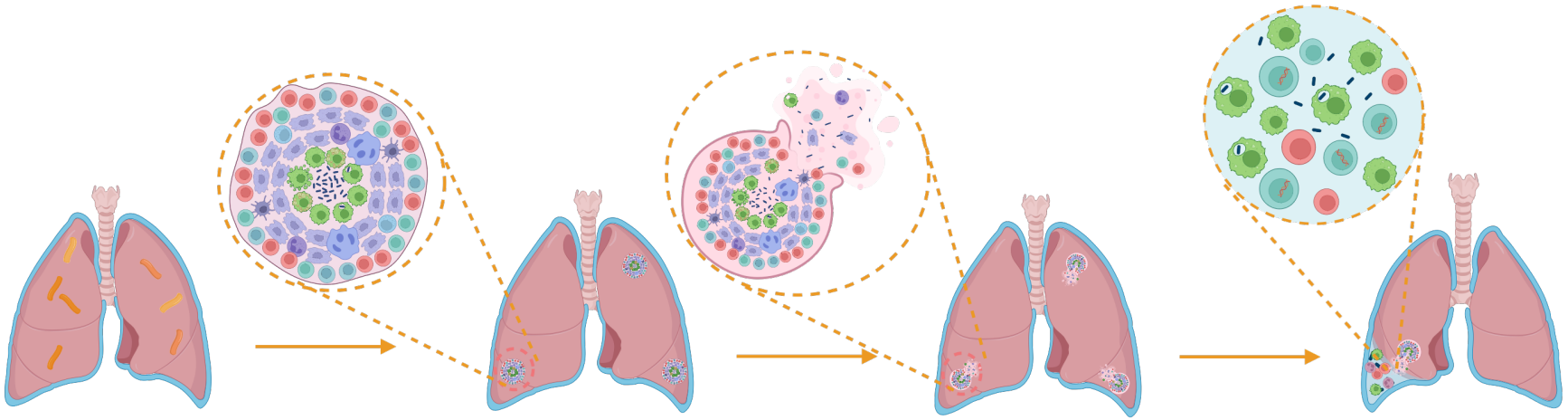




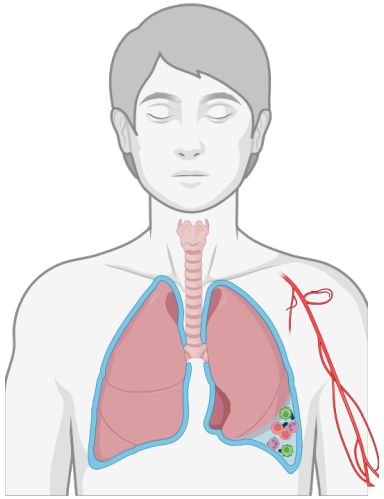
TB-HIV



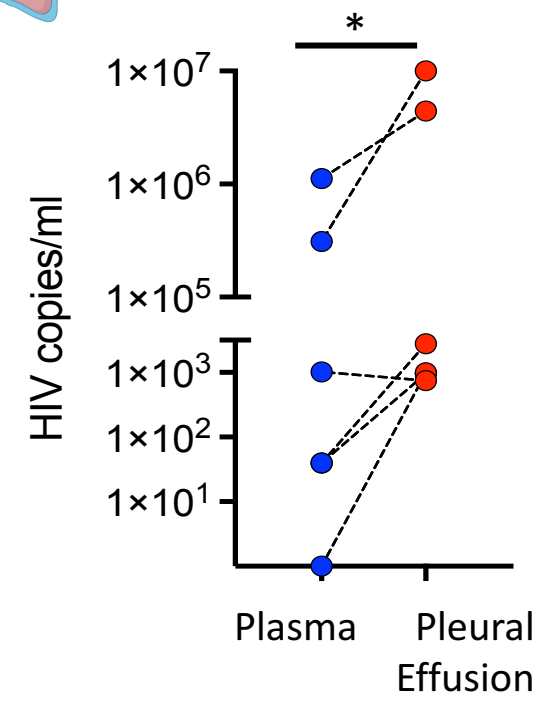
Concurrent TB and HIV at the Site of the Coinfection



- Pleural TB is more common in PLWH
- Pleural effusion contains high HIV titres



Toossi et al., 2001
Collins et al., 2002
Toossi et al., 2003



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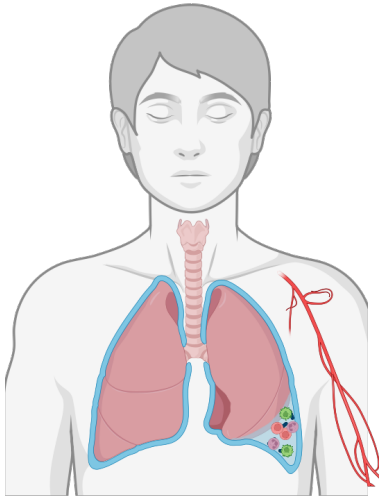
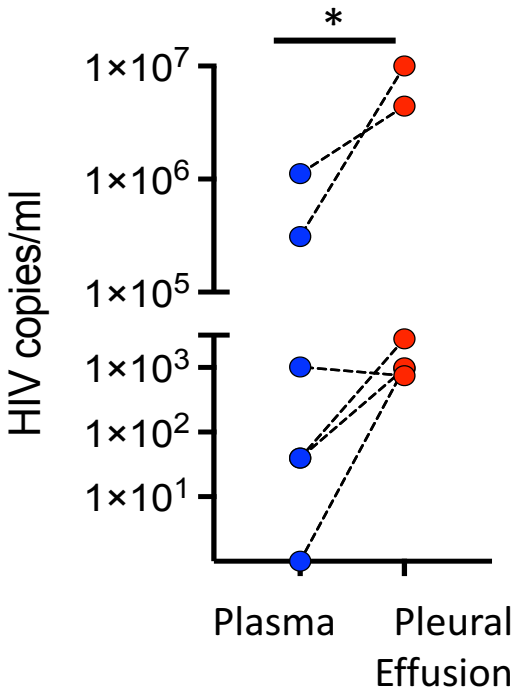
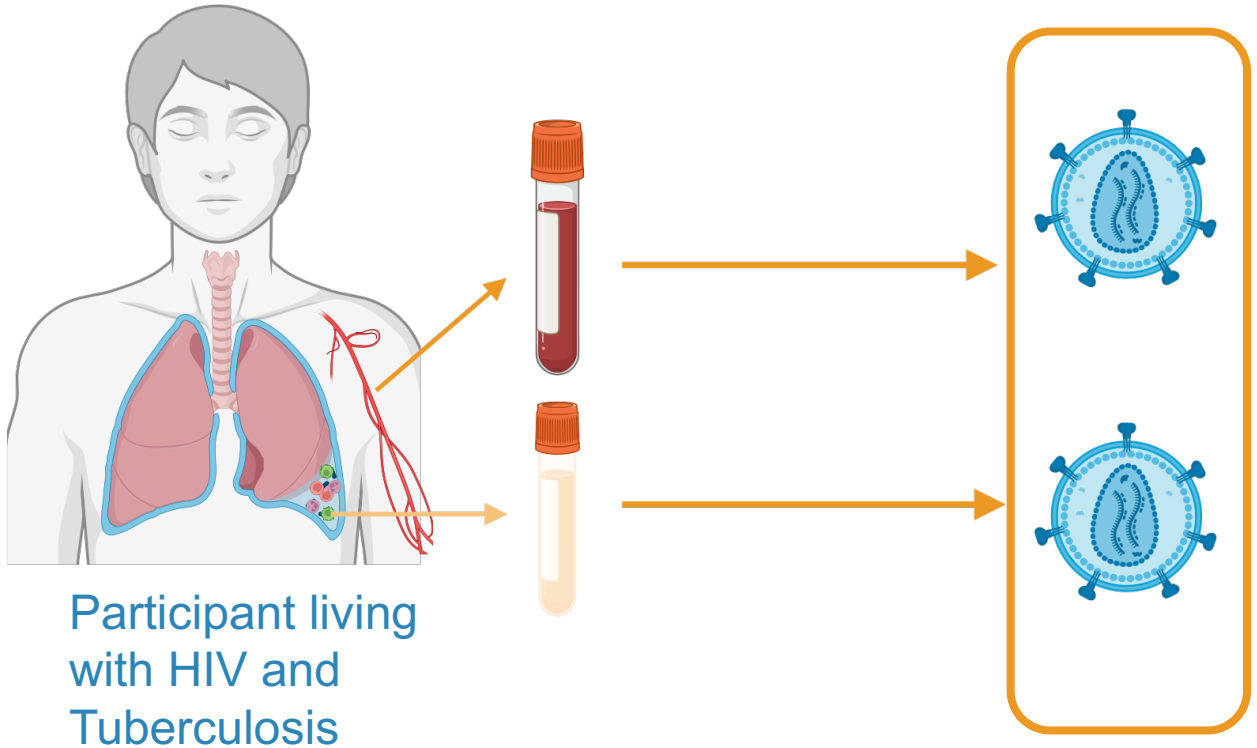


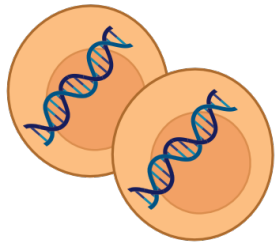


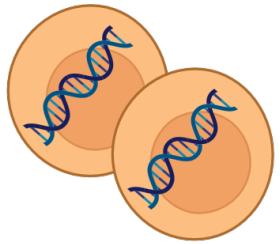
What leads to the higher viral load seen in pleural fluid?

Method: PRLS for RNA Sequencing



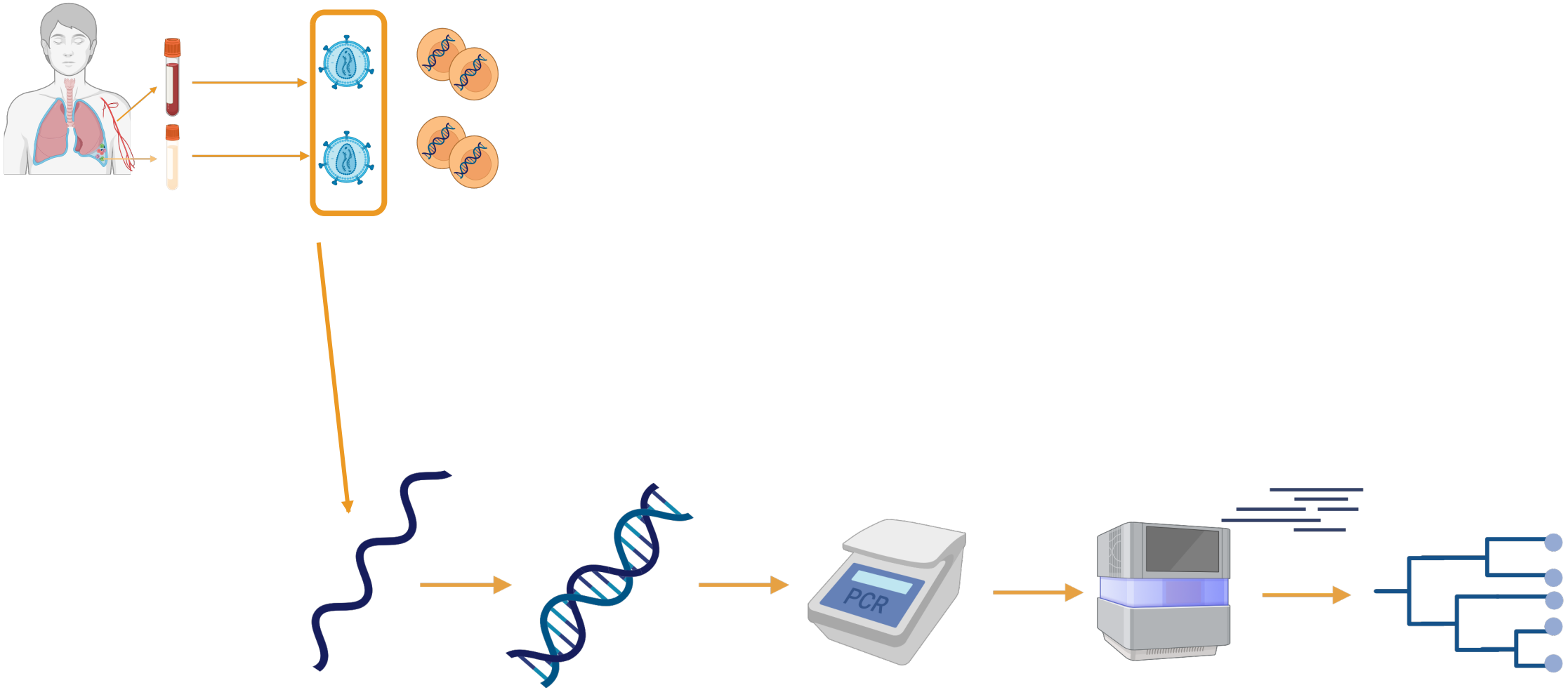
Participant living with HIV and Tuberculosis





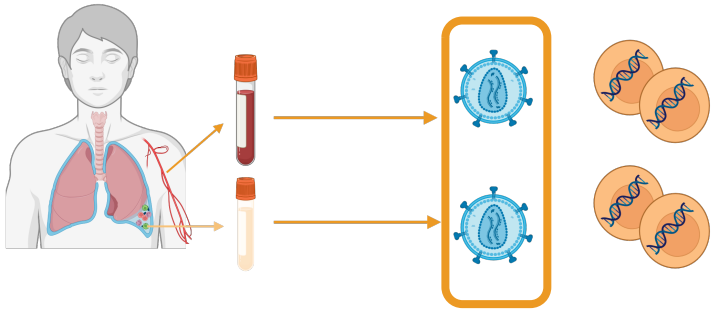
Plasma viral load: 309 909 copies/ml
Pleural effusion viral load: 10 000 000 copies/ml
Participant was not on ART at the time of sample collection

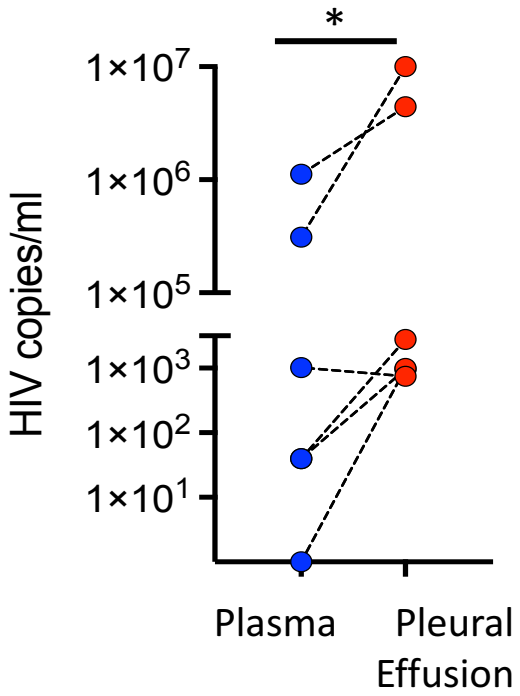
Method: PRLS* for RNA Sequencing

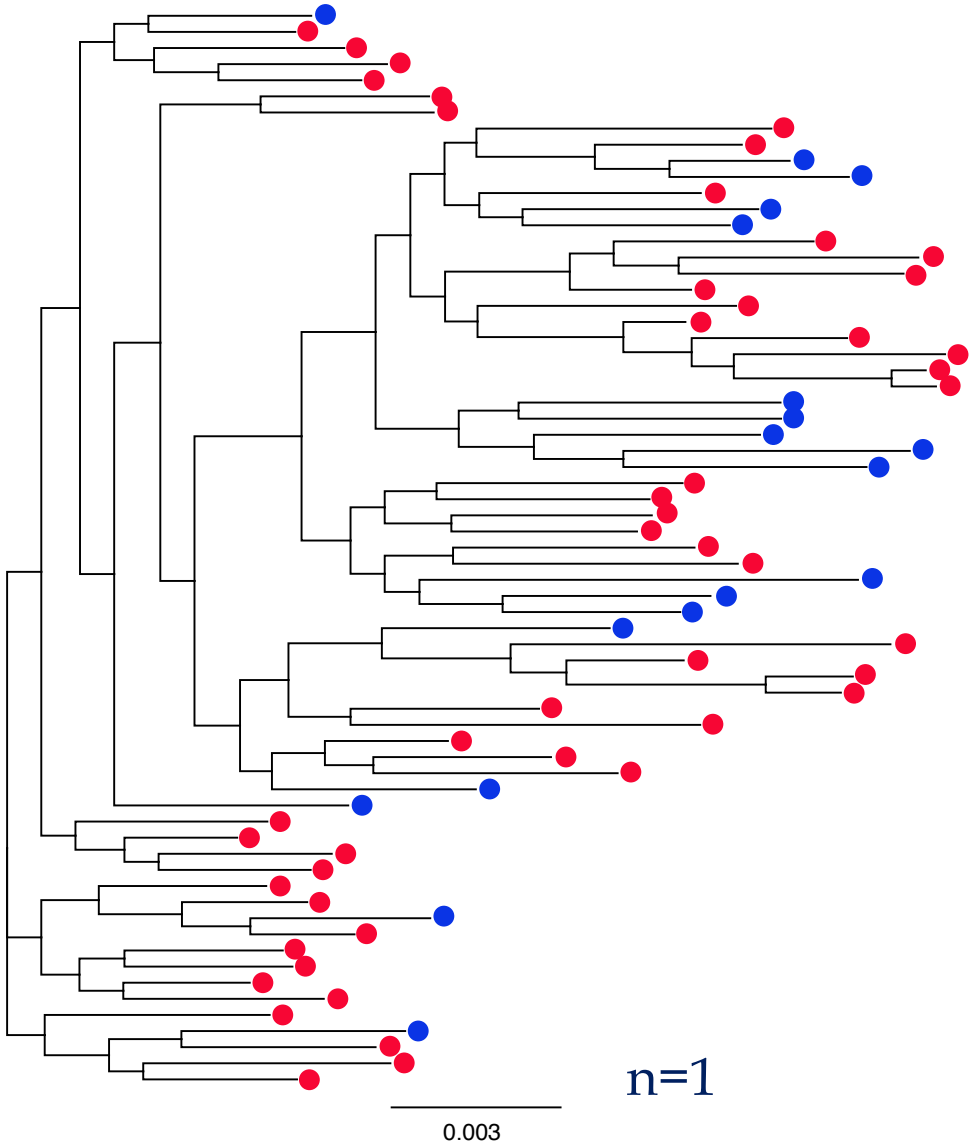


*Fisher et al., 2022 Plasma-Derived HIV-1 Virions Contain Considerable Levels of Defective Genomes

Result: RNA is not Compartmentalised in the Pleural Space







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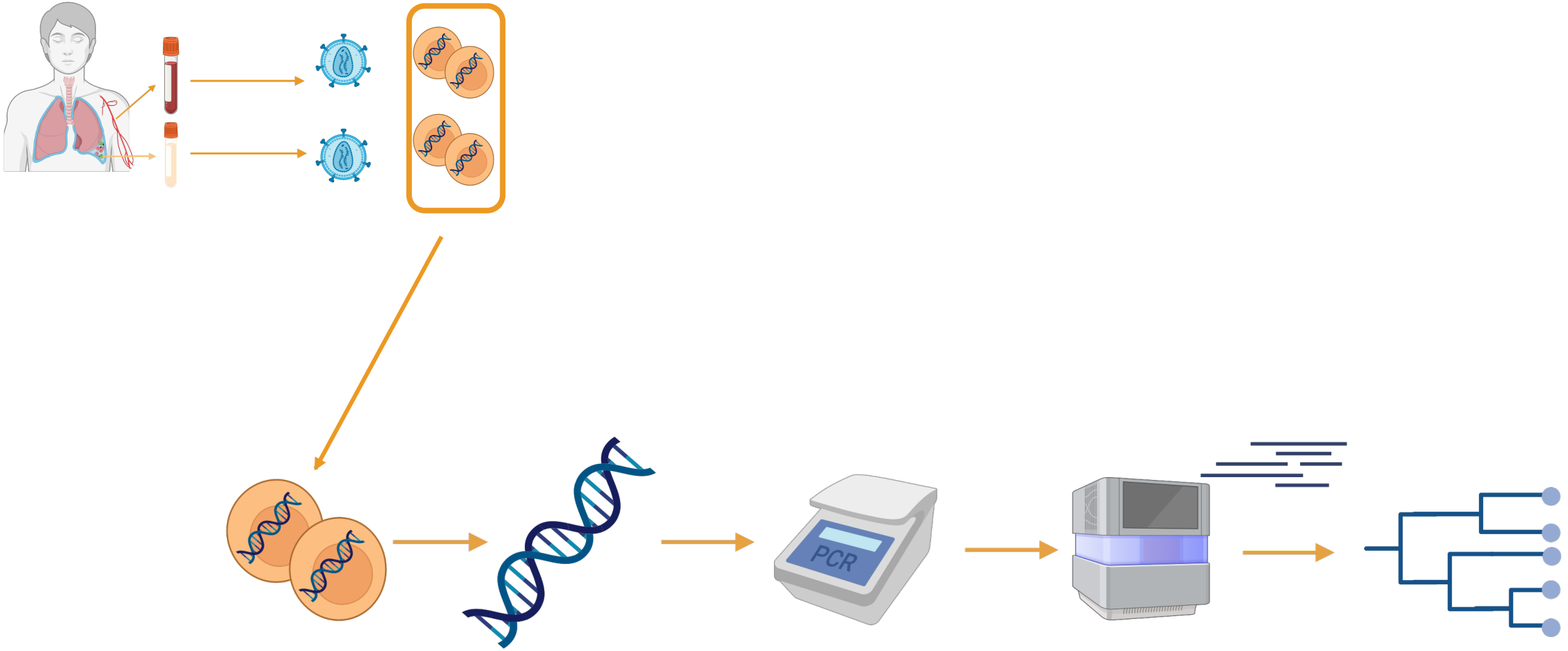

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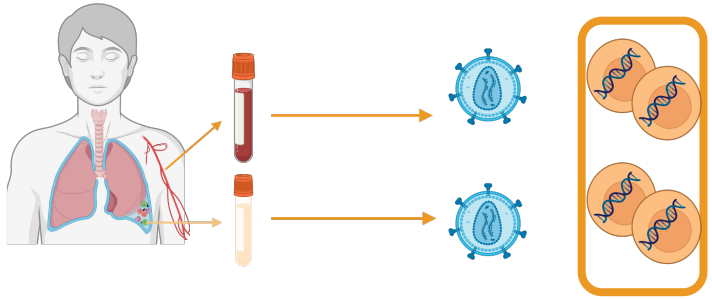
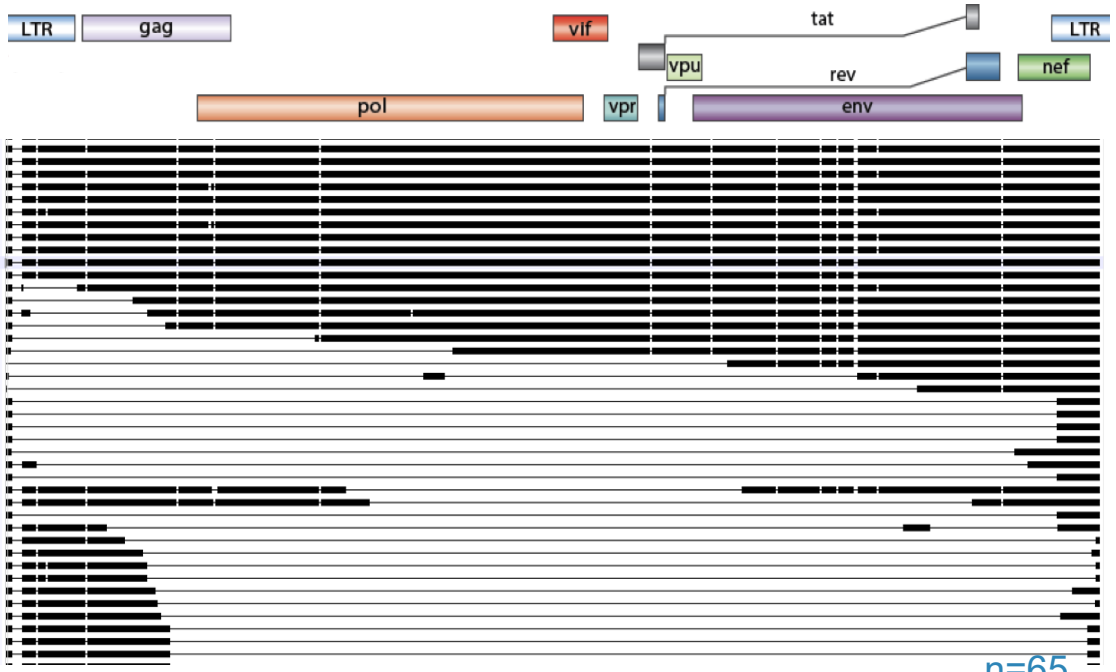
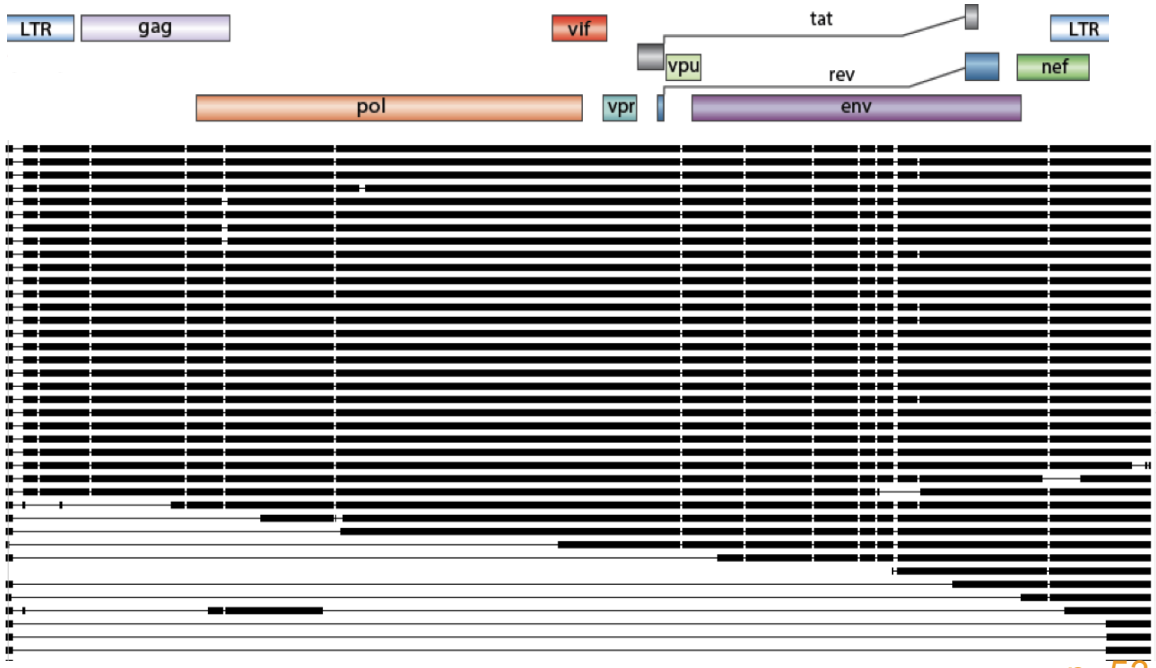


Mononuclear Cells from Blood



n=65

Mononuclear Cells from Pleural Effusion



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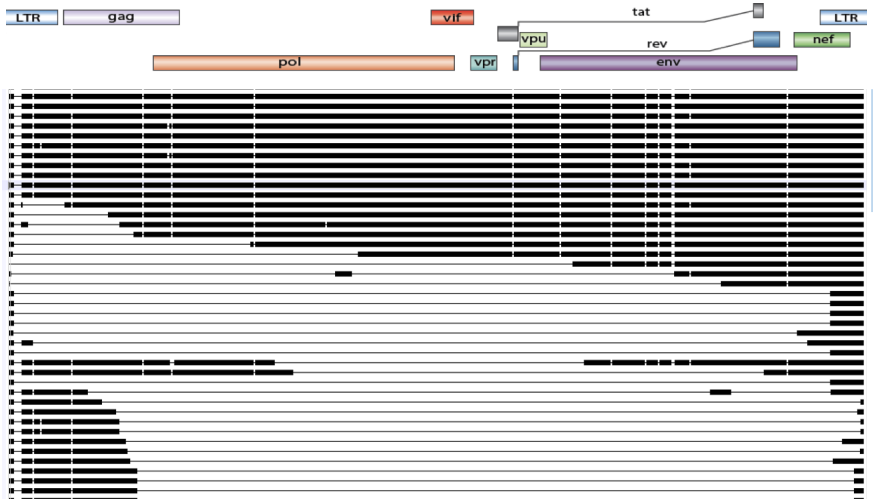
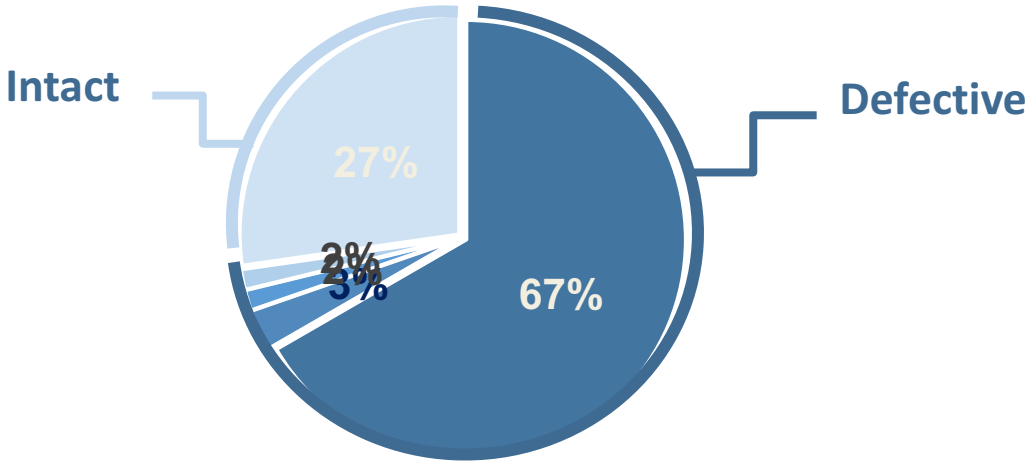
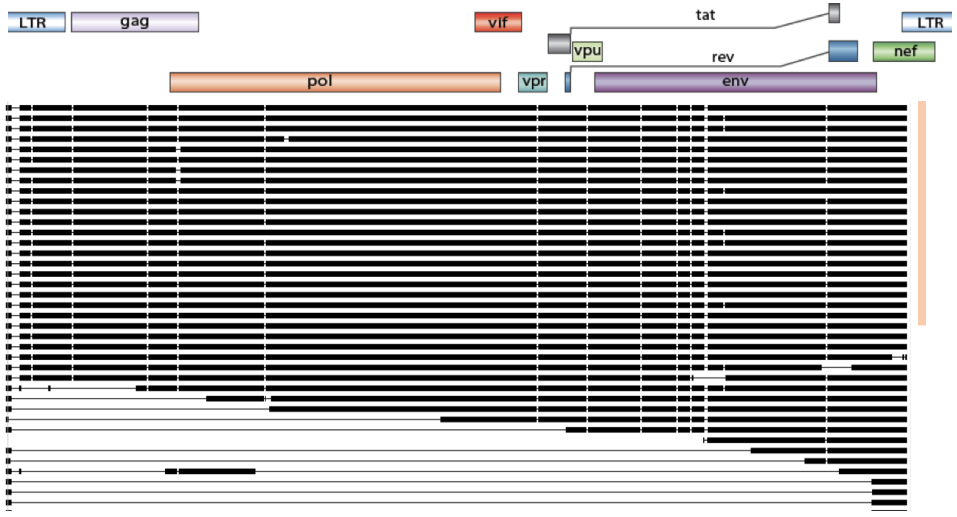


Mononuclear Cells from Blood

Defective (LID) Defective (INV) Hypermut Frameshift/Stop codon Intact





Mononuclear Cells from Pleural Effusion

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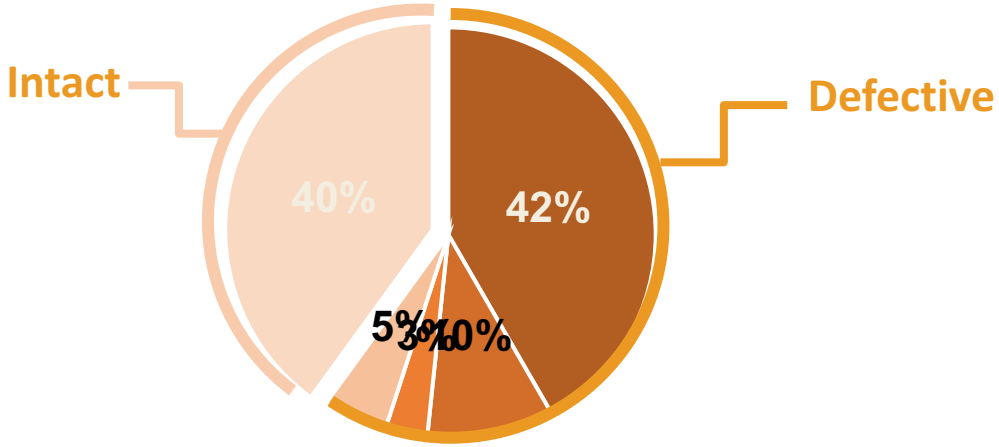

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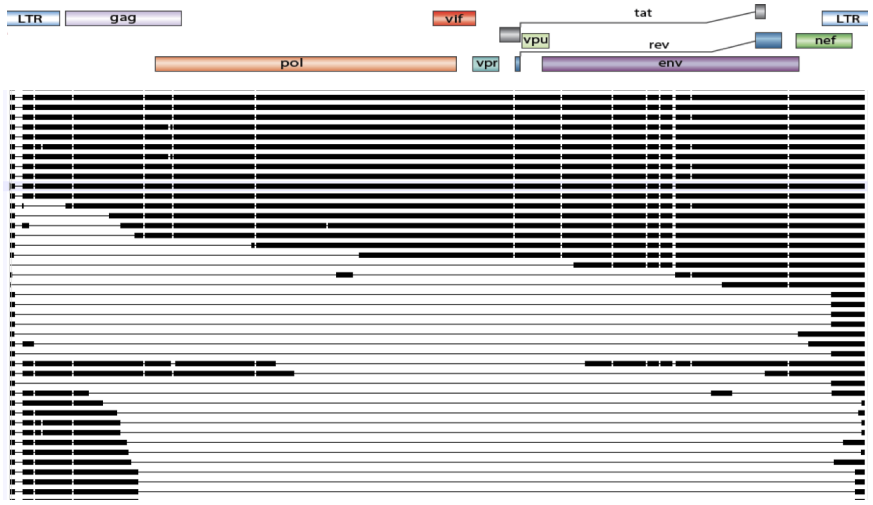

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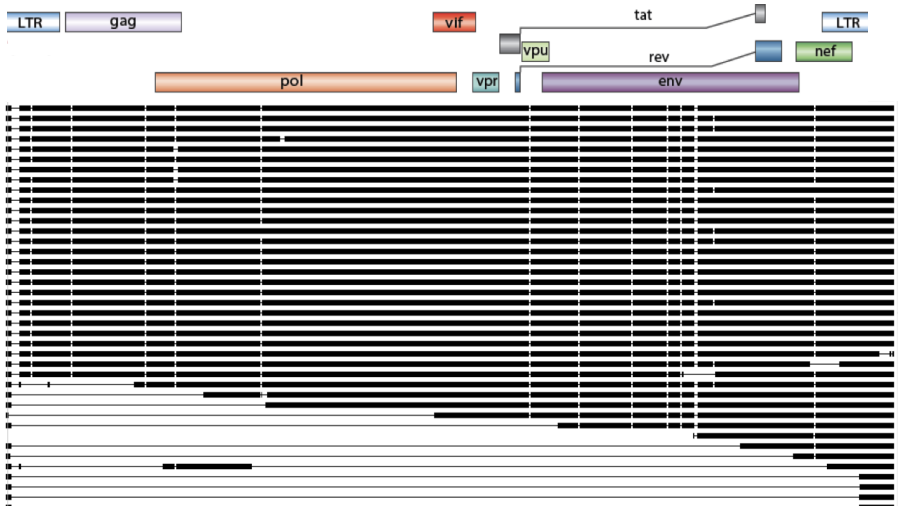
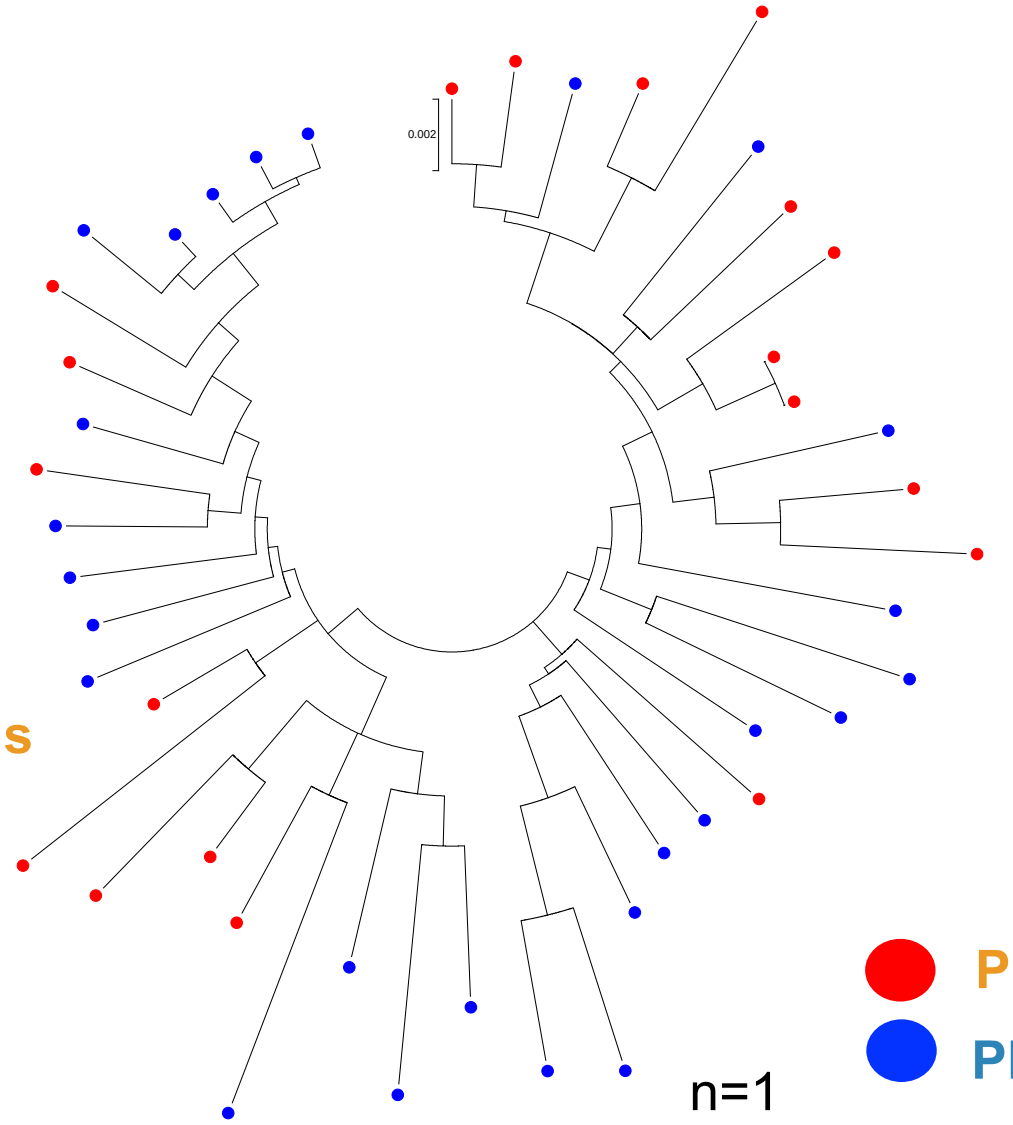


Mononuclear Cells from Pleural Effusion



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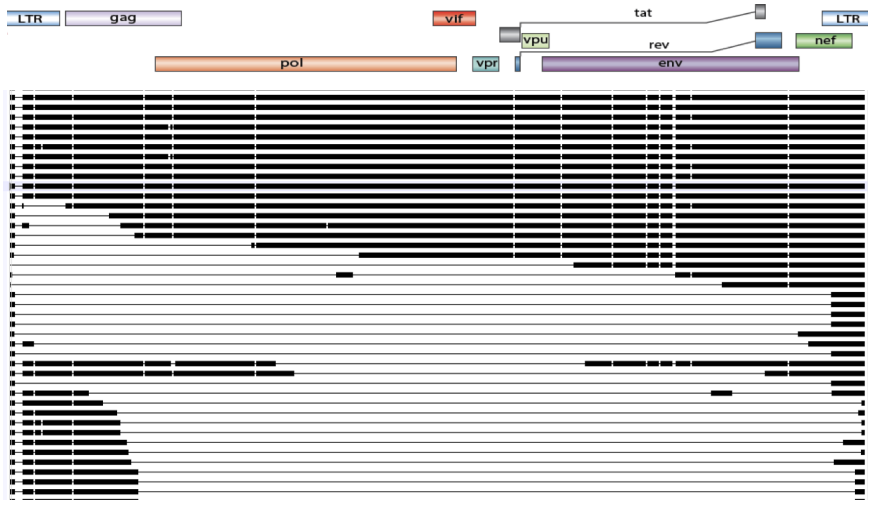

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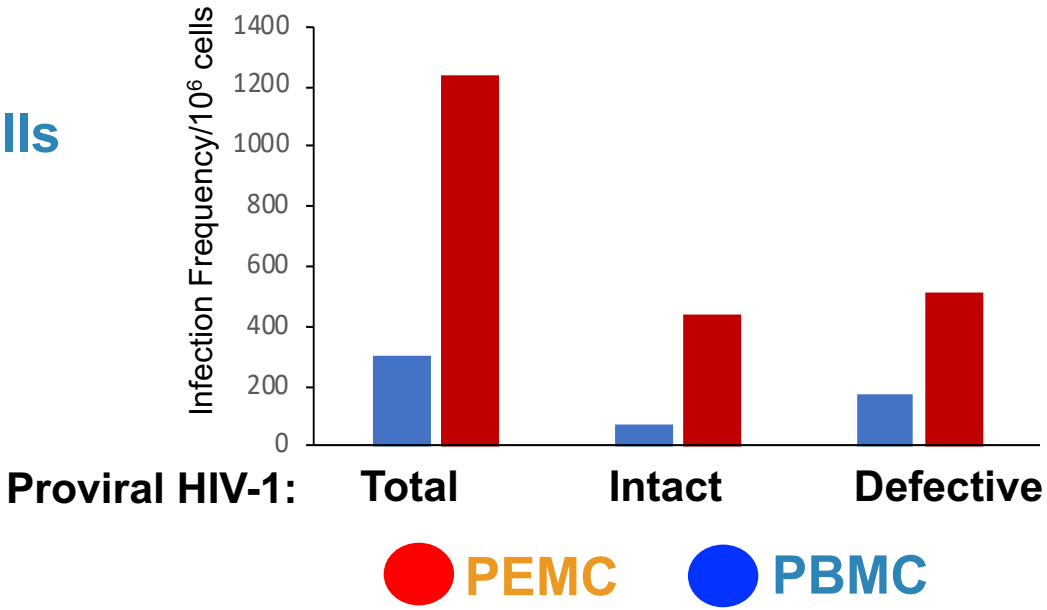

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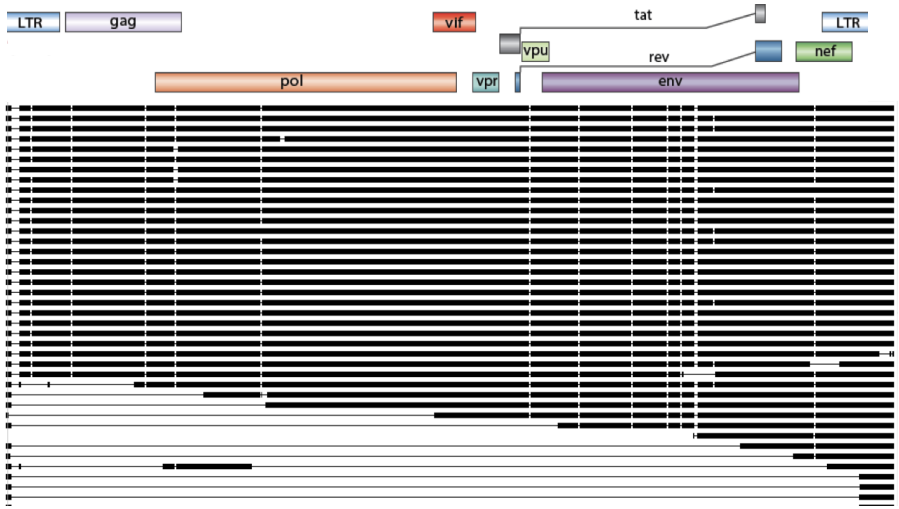

Mononuclear Cells from Pleural Effusion

If there's no compartmentalisation, why are there more genetically-intact proviruses in the pleural fluid?

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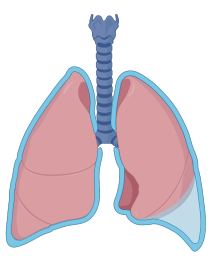
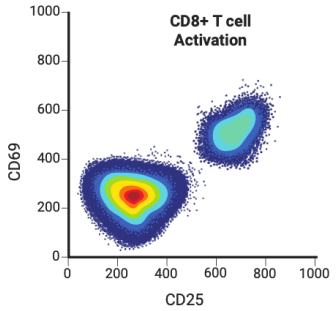
Hypothesis: The CD8+ T cell-mediated anti-HIV response is impaired at the site of the coinfection.

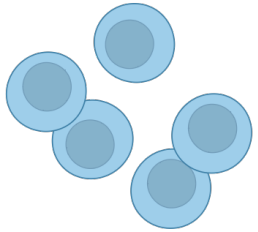
Method: CD8+ T cell Activation in the Presence of TB-PE

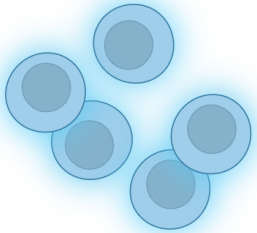


Pleural effusion isolated from people with pleural tuberculosis



Flow cytometry analysis of activation markers





Healthy donor blood

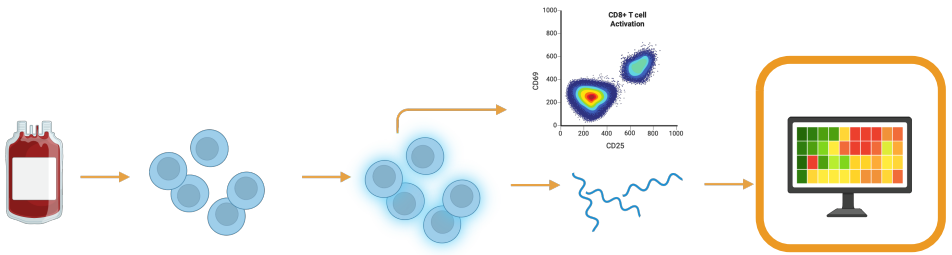
CD8+ T cell isolation

CD8+ T cell activation +/- TB-PE

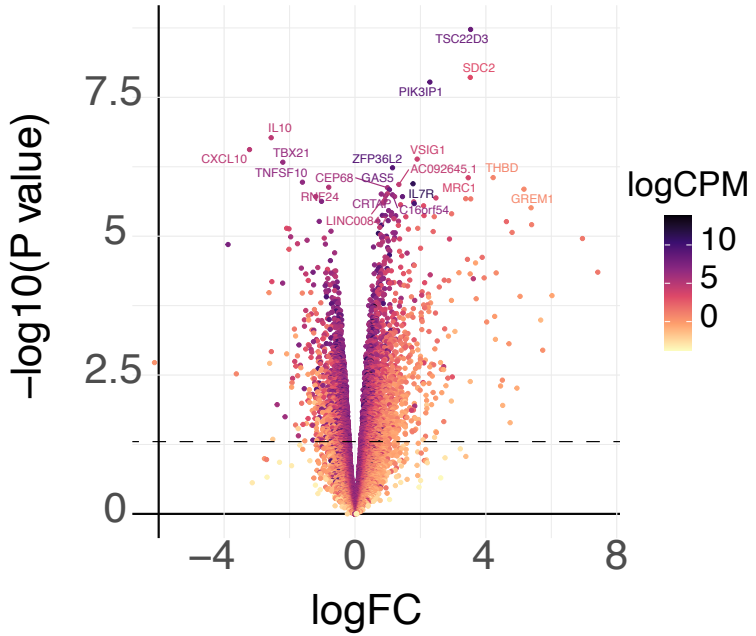
Bulk RNAseq

Analysis

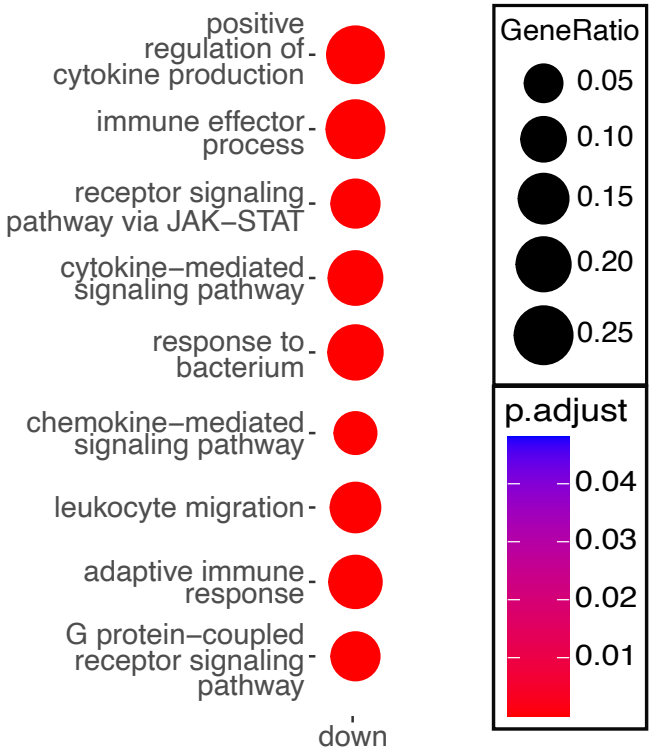
Result: CD8+ T cell Activation is Downmodulated by TB-PE



Activated CD8+ T cells vs Activated CD8+ T cells + TB-PE



Pathway analysis of pathways associated with CD8+ T cell activation



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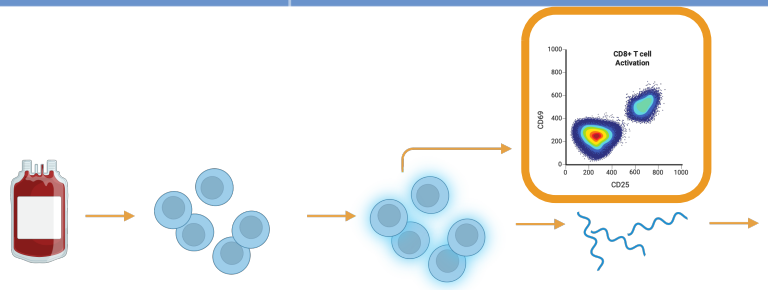

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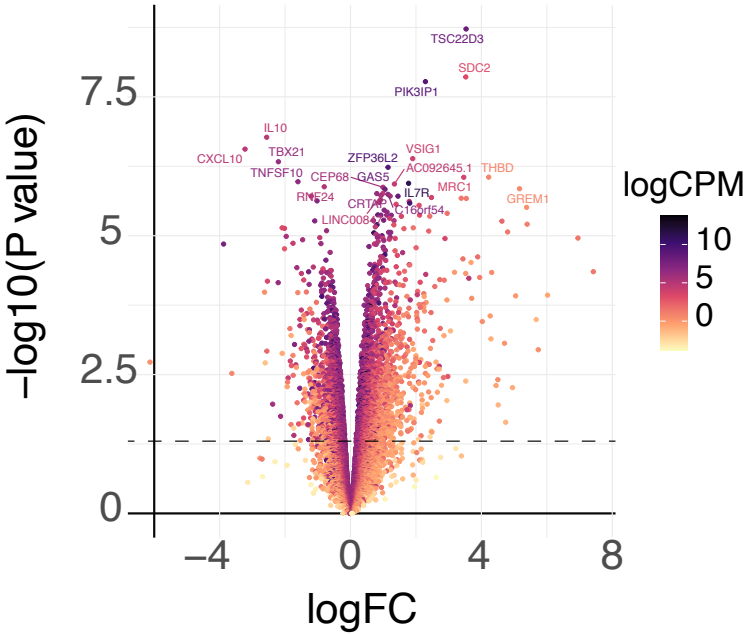

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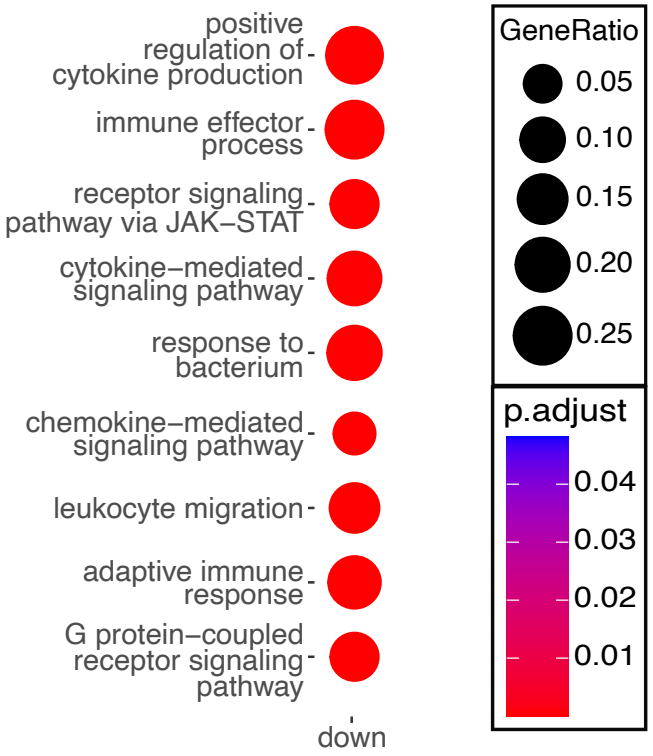
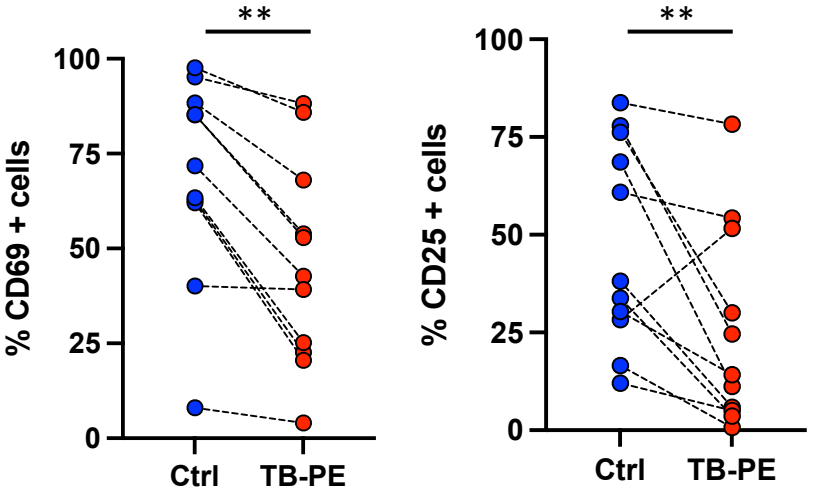
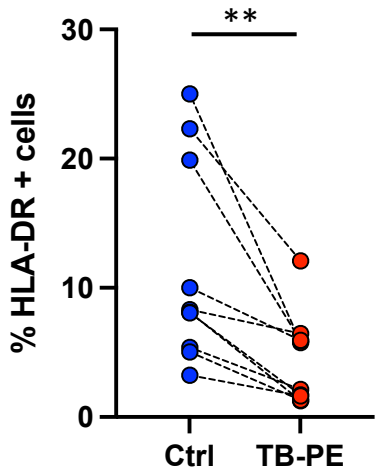
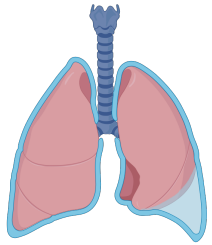




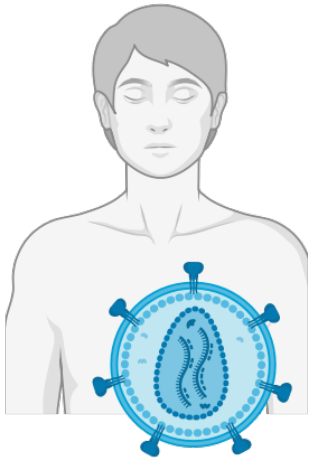


*p<0.05, **p<0.01, ***p<0.001

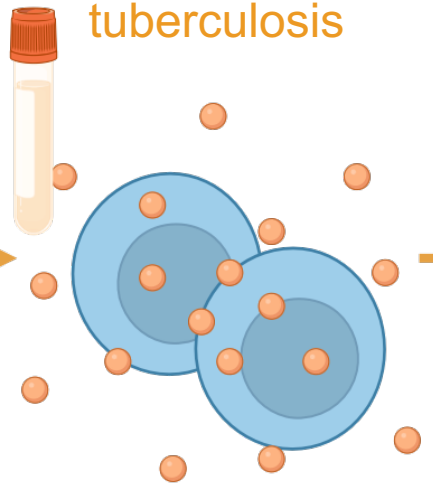
Method: Assessing CD8+ T cell Functionality in the Presence of TB-PE



Pleural effusion isolated from people with pleural tuberculosis



PBMCs isolated from a person living with HIV



Cells stimulated with HIV peptides +/- TB-PE

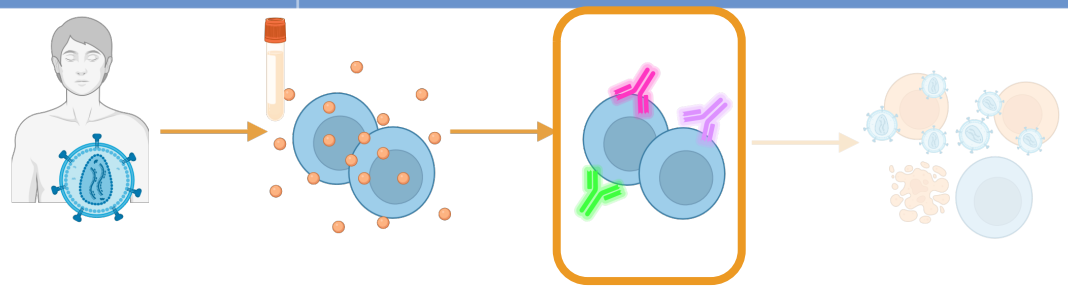
α-CD107a/b

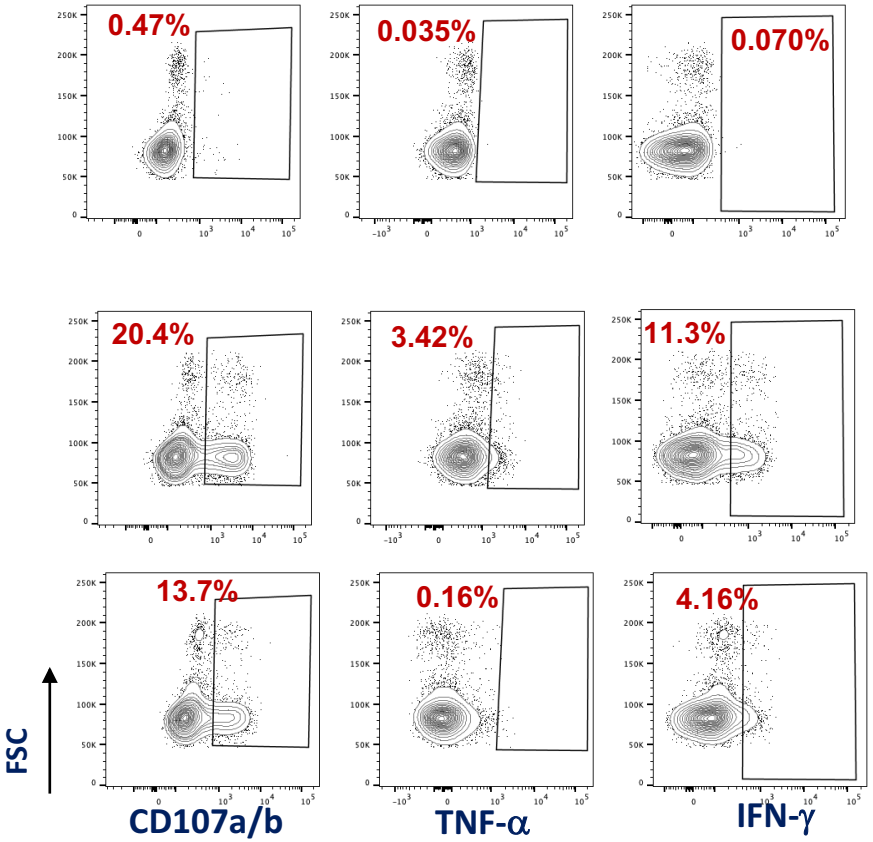
α-IFNγ

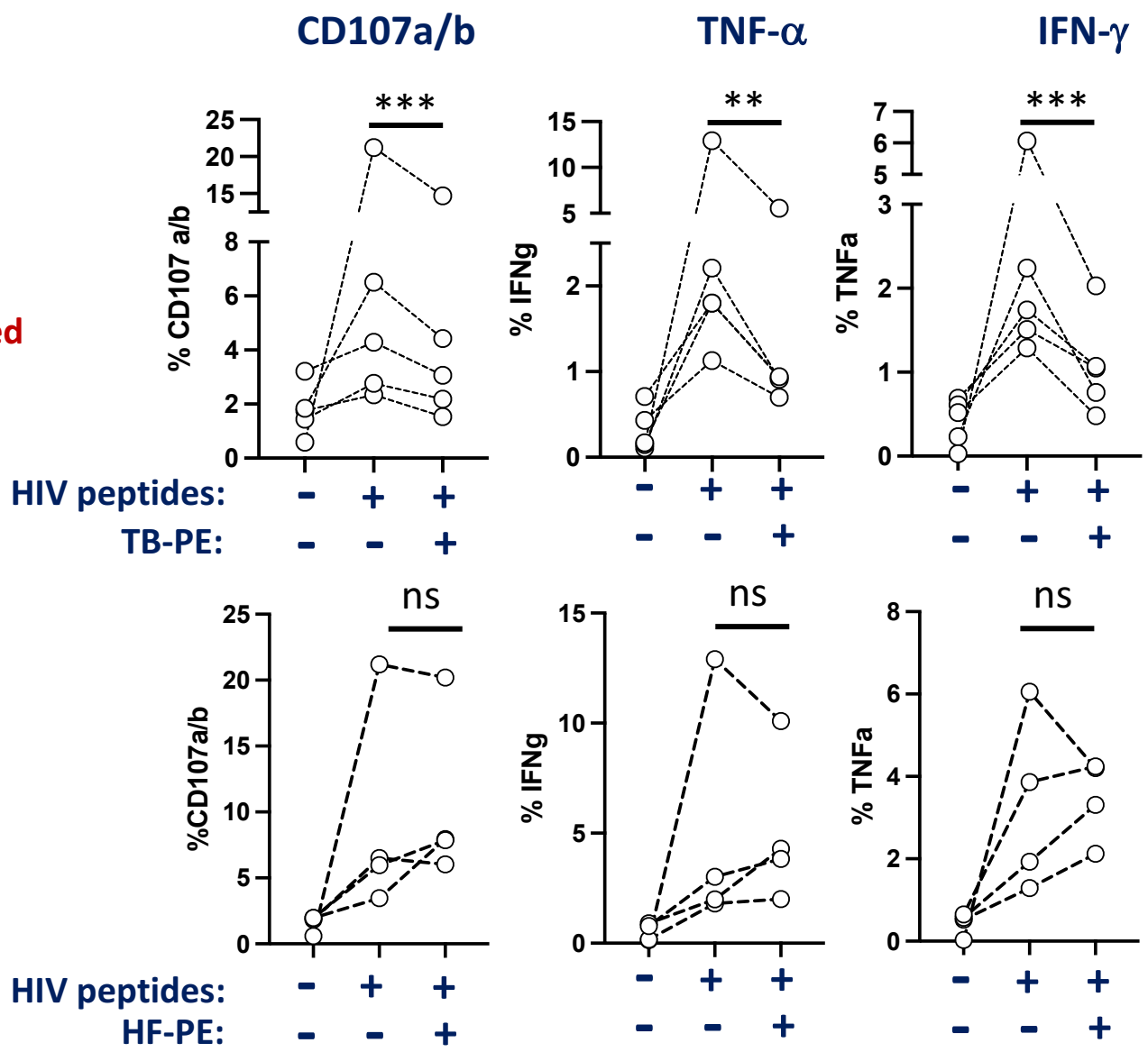
α-TNF α

Markers of CD8+ T cell functionality assessed after 14 days of expansion

Result: CD8+ T cell Functionality is Impaired by TB-PE

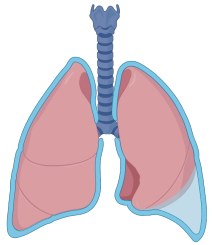




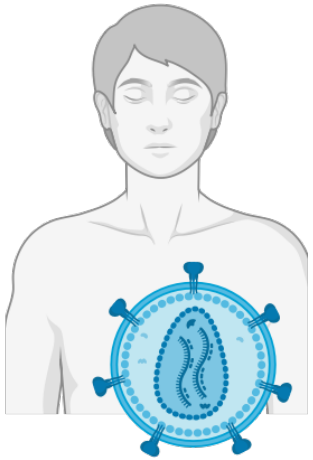


*p<0.05, **p<0.01, ***p<0.001

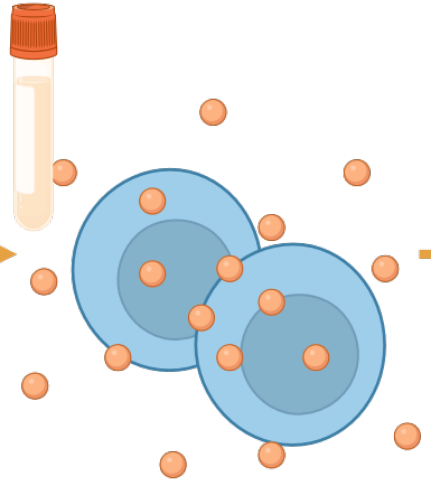
Method: Assessing CD8+ T cell Killing Capacity in the Presence of TB-PE



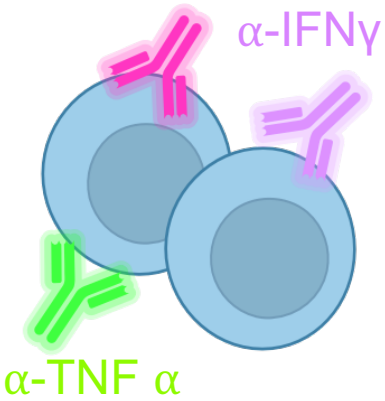
Pleural effusion isolated from a person with pleural tuberculosis



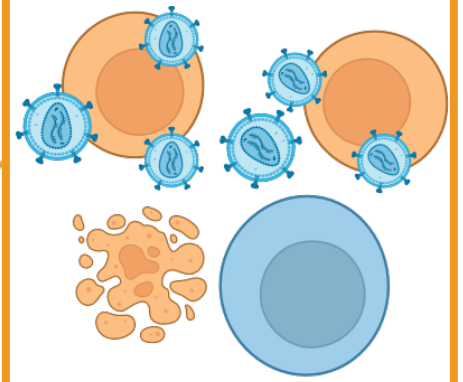
PBMCs isolated from a person living with HIV



Cells stimulated with HIV peptides +/- TB-PE

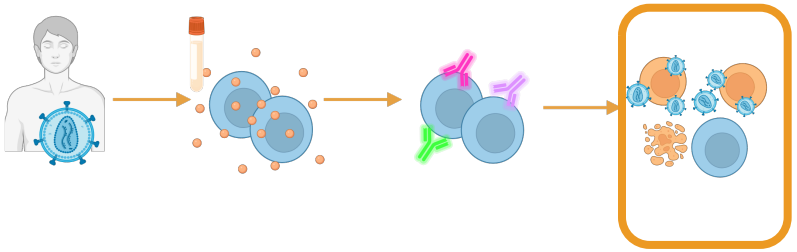


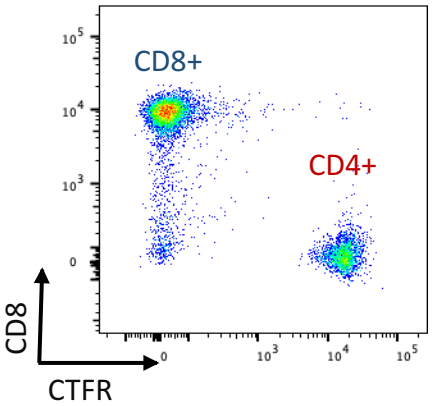
Markers of CD8+ T cell functionality assessed after 14 days of expansion

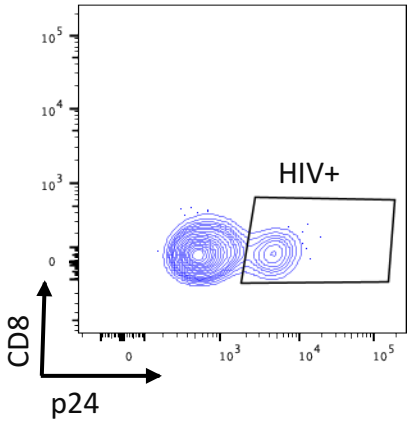


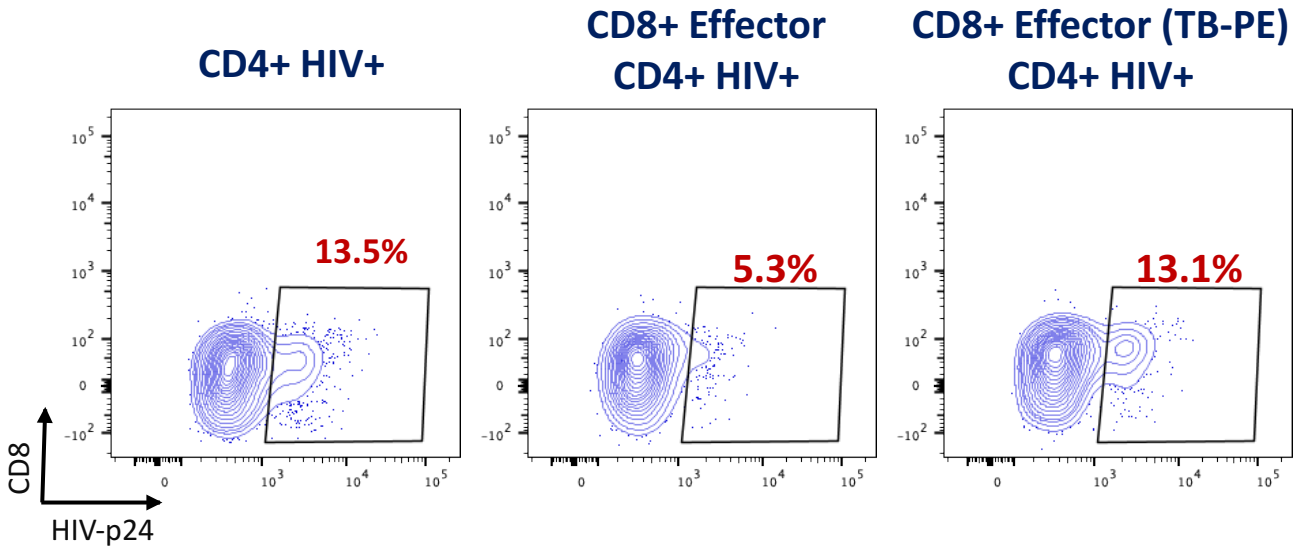
CD8+ T cells are co-cultured with HIV-infected CD4+ T cells

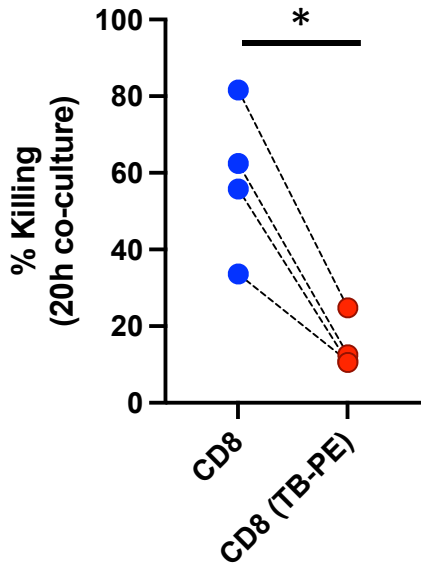
Method: Assessing CD8+ T cell Functionality in the Presence of TB-PE





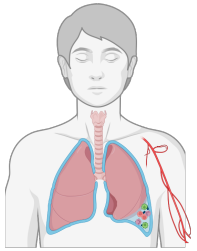


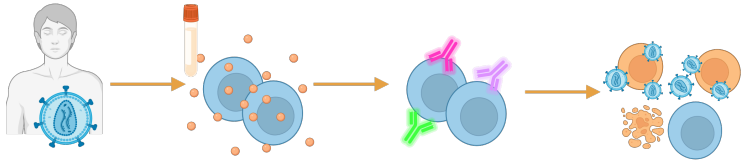




*p<0.05

Conclusions





1. The HIV infection frequency in PBMCs from people living with HIV and TB is higher.

2. More genetically intact provirus identified at the site of the coinfection.

3. TB-PE reduces effector functionality of HIV-specific CD8+ T cells

The tuberculosis-associated microenvironment impacts CD8+ T cell functionality, leading to reduced viral control at the site of the coinfection.

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INBIRS


IPBS Institut de Pharmacologie et de Biologie Structurale

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