Antigen responsive clones of CD4+ T cells contribute to the intact latent reservoir

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ABSTRACT:

Introduction: Antiretroviral therapy (ART) suppresses but does not cure HIV-1 infection due to the existence of a long-lived reservoir of latently infected cells. The reservoir is largely composed of clones of infected CD4+ T cells and its persistence appears to result in part from expansion and contraction of infected CD4+ T cell clones. However, the mechanisms that govern this process are poorly understood.

Methods: To determine whether latent proviral clones might result from, and be maintained by exposure to antigen, we analyzed viruses from latently infected cells responding to a subset of antigens from viruses associated with chronic or recurrent infections.

Result: We found clones of antigen responsive CD4+ T cells containing defective or intact latent proviruses in 7 out of 8 individuals studied.

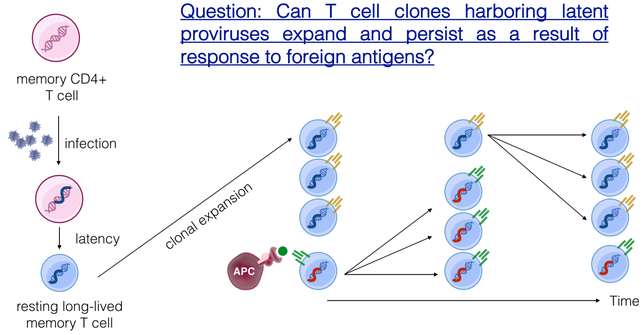
Conclusion: Chronic or repeated exposure to antigen may contribute to the longevity of the HIV-1 reservoir by stimulating the clonal expansion of latently infected CD4+ T cells.

INTRODUCTION:

The HIV-1 latent reservoir is composed of expanded clones of CD4+ T cells (~50%)[1,2,3,4,5], including HIV-1, CMV, and influenza specific clones[6,7,8]. Latent reservoir clones are dynamic, waxing and waning over time[9,10].

However, the origin of latent clones and the mechanisms that govern their persistence is not known. Thus, understanding the basis for latently infected T cell clonal expansion is important for learning how to control and potentially eliminate the reservoir. Here, we address whether latently infected cells can undergo clonal proliferation in response to cognate antigens.

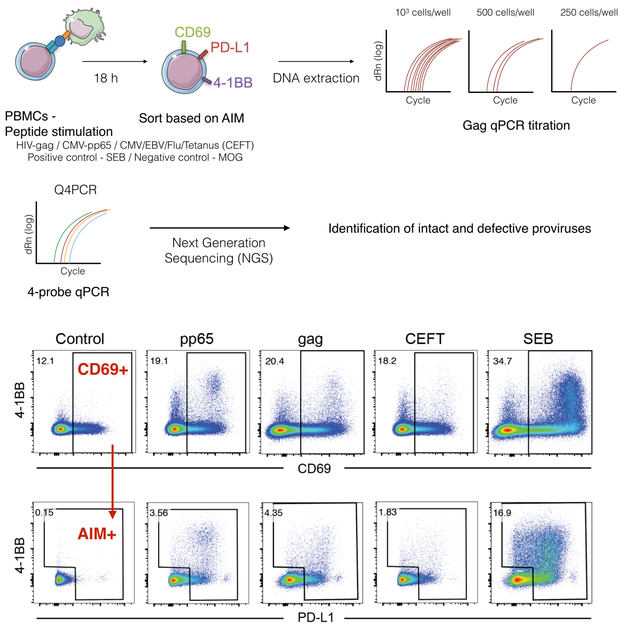
Question: Can T cell clones harboring latent proviruses expand and persist as a result of response to foreign antigens?



METHODS :

AIM Assay: CD4+ T cells from 8 ART suppressed individuals[11] were cultured with peptide pools of overlapping peptides from CMV-pp65, HIV-gag and a cocktail of peptides from CMV, EBV, Flu and Tetanus toxin (CEFT). Activated CD4+ T cells were sorted based on expression of two or more activation induced markers (AIM+: CD69 and PD-L1 or 4-1BB).

Q4PCR: Single integrated proviruses from AIM+ sorted cells are characterized as intact or defective by combining 4-probe qPCR and NGS[12].



RESULTS:

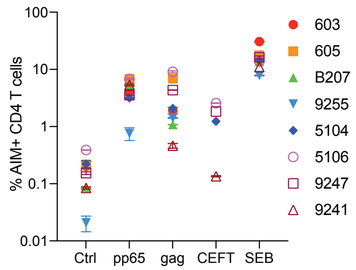


Figure 1. Percentage of antigen-specific CD4+ T cells responding to HIV-gag, CMV-pp65 and CEFT vary in magnitude among individuals.

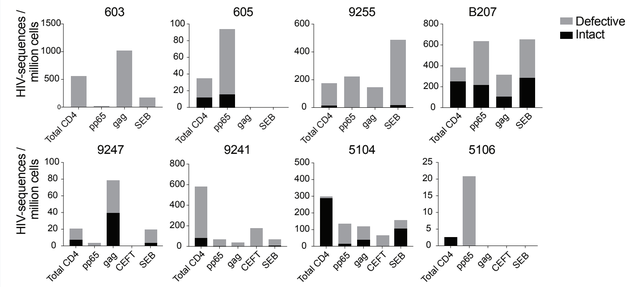


Figure 2. Overall frequency of intact (black) and defective (grey) integrated proviruses contained within CMV-pp65, HIV-gag and CEFT antigen responsive (AIM+) cells vary among individuals.

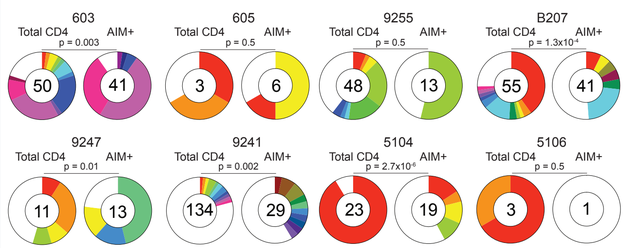


Figure 3. Clonal distribution of HIV-1 sequences in antigen responsive (AIM+) cells differs from the total CD4+ T cell control in 5 of 6 individuals for whom we obtained sufficient data.

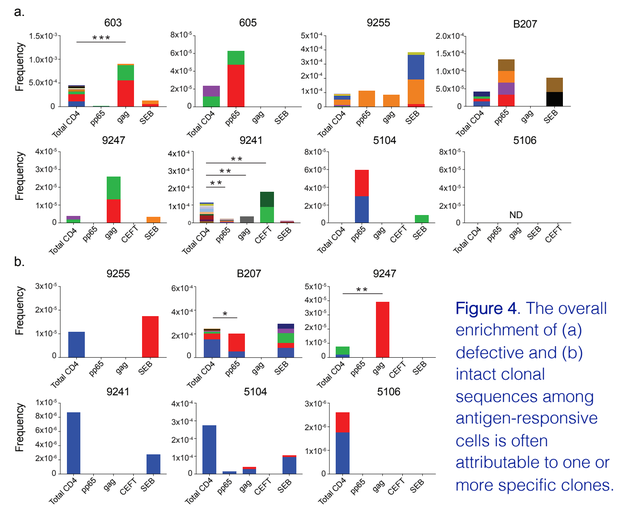


Figure 4. The overall enrichment of (a) defective and (b) intact clonal sequences among antigen-responsive cells is often attributable to one or more specific clones.

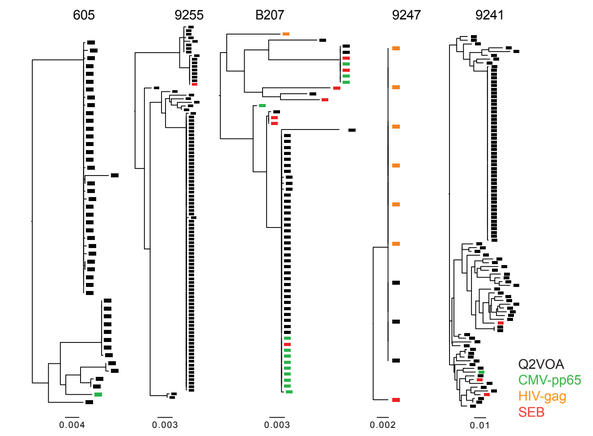
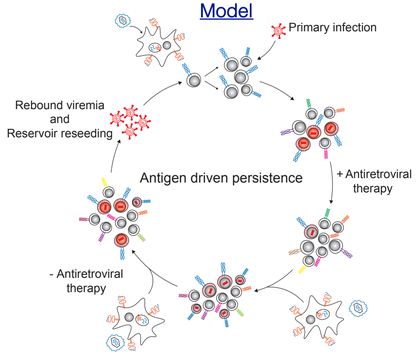


Figure 5. Some intact *env* sequences from AIM+ cells (colored) are identical to replication competent viruses obtained by outgrowth (black) in 3 of 5 donors assayed.



CONCLUSIONS:

1. Both intact and defective HIV-1 proviruses can persist in clones of CD4+ T cells that respond to CMV and HIV antigens.
2. Infected clones of CD4+ T cells may respond to diverse pathogens in HIV-1 infected individuals.
3. Intermittent exposure to common antigens found in the virome and microbiome may account for the reported waxing and waning of individual clones of latently infected cells and their persistence over time.

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