Highly Networked CD8$^+$ T cell Epitopes have Markedly few Mutations in the Latent HIV Reservoir

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BACKGROUND

The persistent HIV latent reservoir is a major impediment to the eradication of HIV for people living with HIV (PLWH). Our laboratory identified a subset of epitopes, known as highly networked CD8+ T cell epitopes, that are constrained from mutation and preferentially targeted by individuals who naturally control HIV. In this study, we assessed whether highly networked epitopes represent a set of mutationally constrained targets for CD8+ T cells within the latent HIV reservoir to guide future T cell vaccine efforts. We hypothesize that highly networked epitopes incur markedly fewer mutations than low networked epitopes in the persistent latent reservoir.

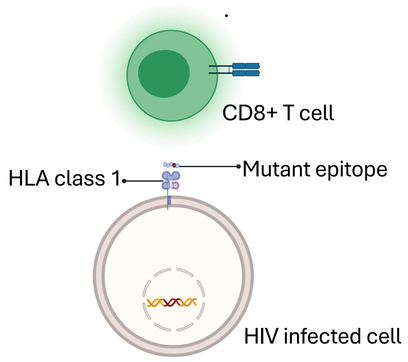


Figure 1. A mutation in viral epitope makes infected cell unrecognizable by CD8+ T cells

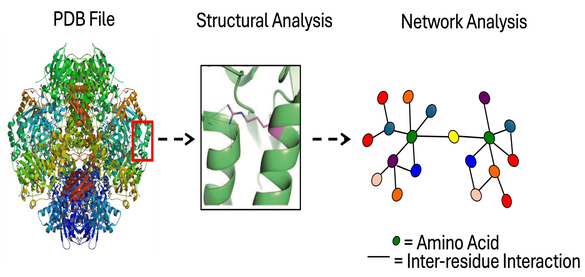


Figure 2. HIV controllers target structurally constrained epitopes that exact substantial fitness cost upon mutation. *Gaiha et al., Science 2019*

ADDITIONAL KEY INFORMATION

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Highly networked epitopes represent a set of mutationally constrained targets for therapeutic T cell vaccine approaches that may enable durable viral suppression.

RESULTS

High networked epitopes had a markedly reduced frequency of mutations than low-networked epitopes in integrated proviruses (P<0.0001) and in a sub analysis of epitopes restricted by individual participant HLA class I alleles (P<0.0002). T cell responses to highly networked epitopes demonstrated preserved recognition of mutated epitopes from proviral sequences, whereas mutated non-networked epitopes had significantly reduced recognition by existing T cell responses.

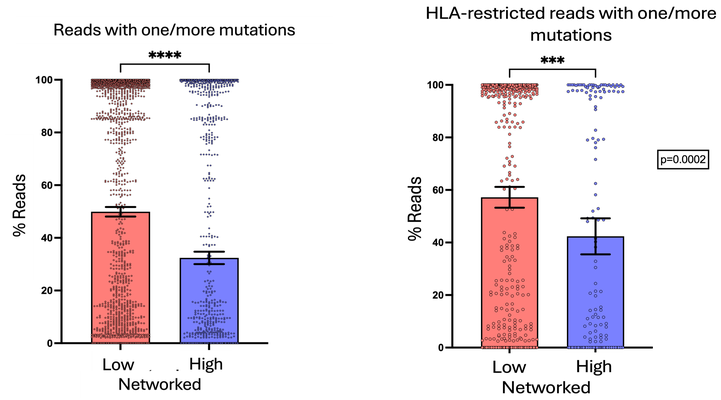


Figure 4. (**A**) High networked epitopes incurred fewer mutations than low networked epitopes. (**B**) An analysis of epitopes restricted by an individual's HLA class I alleles, high networked epitopes incurred fewer mutations than low networked epitopes.

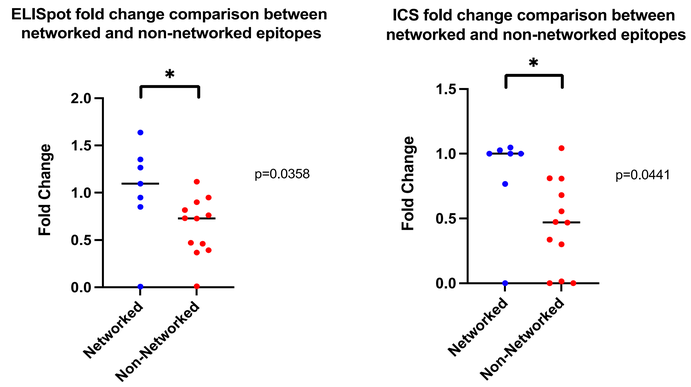


Figure 5. (**A**) Mutated networked epitopes demonstrated preserved T cell recognition whereas mutated non-networked epitopes had significantly reduced recognition by existing T cell responses as measured using ELISpot **(A)** and ICS **(B)** assays.

METHODS

To determine mutational frequencies of highly networked and non-networked epitopes in the latent reservoir, we performed proviral and near full-length individual proviral (FLIP) sequencing on genomic DNA from latently infected CD4+ T cells in 22 HIV+ individuals. All participants were fully suppressed with ART for >1 year and maintained undetectable viral loads at the time of sample collection. Sequence analysis of viral epitopes was performed using an established computational pipeline

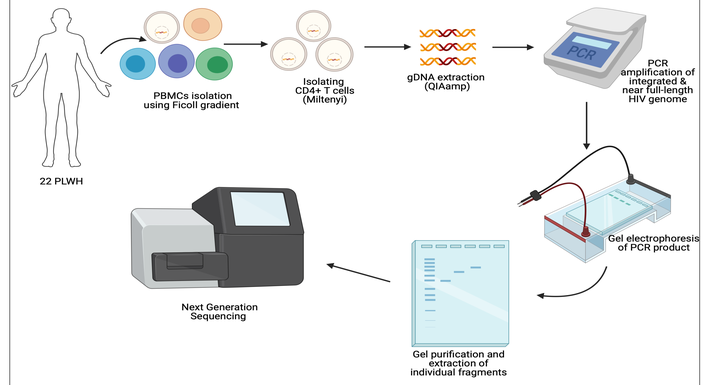


Figure 3. Schematic experimental workflow used to generate, and sequence integrated and near full-length proviral and sequences from PLWH

CONCLUSIONS

This study reveals that highly networked epitopes have markedly few mutations in the latent reservoir from ART-suppressed PLWH. CD8+ T cells recognize mutants of high networked epitopes better than mutants of non-networked epitopes. These findings therefore suggest that highly networked epitopes represent a set of mutationally constrained targets for therapeutic T cell vaccine approaches that may enable durable viral suppression following ART cessation.

