THE THERAPEUTIC PEPTIDE VACCINE VACC-4X TARGETS GAG CTL EPITOPES WITH PREEXISTING MUTATIONS

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Background

Therapeutic HIV immunisation followed by latency reversal has been suggested as a strategy to achieve a functional cure for HIV.

In the clinical trial, REDUC part B, the combination of a peptide-based therapeutic HIV-1 vaccine (Vacc-4x) and the latency reversing agent romidepsin resulted in reductions in the latent HIV-1 reservoir [1], which were associated with general positive CD8+ T cell proliferation ex vivo [2]. However, any qualitative antigen-specific vaccine-induced effects remain elusive.

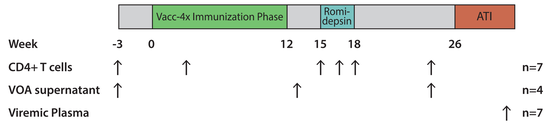
Aim & Hypothesis

The aim of this study was to investigate the phylogenetic composition of the HIV-1 regions targeted by the therapeutic vaccine Vacc-4x in participants in the clinical vaccine trial, REDUC part B [1].

We hypothesized that any cytotoxic T lymphocyte (CTL) immune-mediated effects would result in a selection of mutations in the Vacc-4x targeted domains in the rebounding virus during an analytical treatment interruption and a decrease in genetic diversity of cell-associated HIV-1 RNA and DNA following the intervention.

Study Design

In a clinical trial, 17 participants on suppressive ART were vaccinated with six doses of the therapeutic HIV gag peptide vaccine Vacc-4x (Bionor Pharma) over 12 weeks, then three doses of romidepsin over three weeks followed by an analytical treatment interruption (ATI) [1]. Seven participants were selected for sequencing analysis.



References:

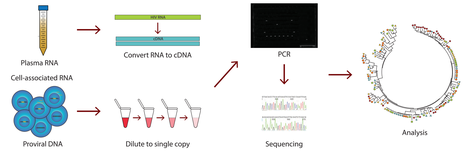
1. Leth, S. et al. Combined effect of Vacc-4x, recombinant human granulocyte macrophage colony-stimulating factor vaccination, and romidepsin on the HIV-1 reservoir (REDUC): a single-arm, phase 1B/2A trial. Lancet HIV (2016)
2. Tapia, G. et al. Sequential Vacc-4x and romidepsin during combination antiretroviral therapy (cART): Immune responses to Vacc-4x regions on p24 and changes in HIV reservoirs. The Journal of Infection (2017)

Acknowledgements:

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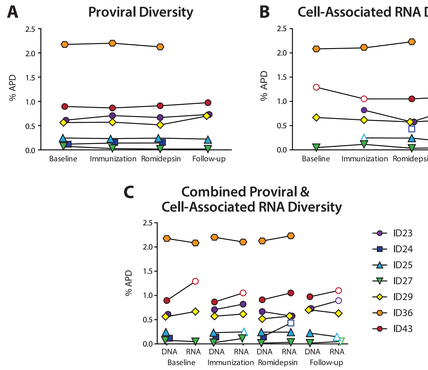
Methods

We used single-proviral/genome sequencing of a 2.1 kb region spanning the p24-RT region to determine the genetic composition of the HIV-1 DNA, cell-associated HIV-1 RNA, stimulated outgrowth viruses and plasma HIV-1 RNA during a treatment interruption. Phylogenetic analyses were conducted using MEGA 6.0.



Vacc-4x and Romidepsin Do Not Impact Proviral Diversity

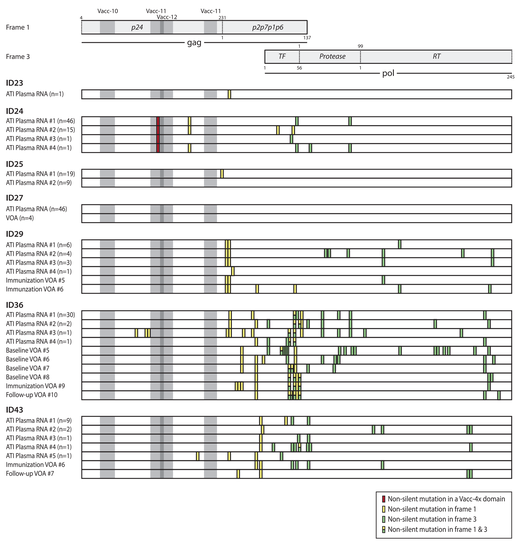
The figure below shows the genetic diversity, measured as average pairwise distances (APD). We observed no changes in proviral DNA (**A**, P = 0.09) and cell-associated HIV-1 RNA (**B**, P=0.83) genetic diversity during the intervention. There was no significant difference in the genetic diversity within each participant between the CA HIV-1 RNA and DNA sequences (**C**, P = 0.32). Statistical test: linear mixed effect model.



Non-silent Mutations Were Located Outside the Vacc-4x Domains

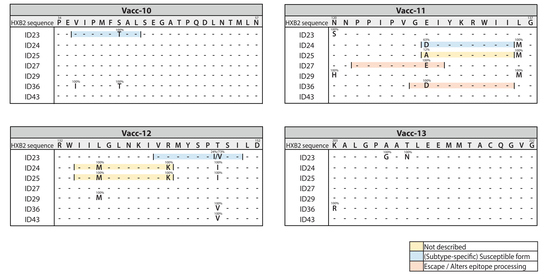
The figure below shows a sequence map of the non-silent mutations in viral outgrowth assay (VOA) and analytical treatment interruption (ATI) plasma RNA consensus sequences when compared to the majority sequence of the proviral compartment.

We identified one instance of a Vacc-4x domain-related non-silent mutation. From participant 24 in the Vacc-12 domain, the 43 proviral sequences analyzed contained 128E and 128D at a frequency of 88% and 12% respectively (of note 128D was detected both at baseline and during romidepsin therapy), whereas 100% of the 63 ATI plasma RNA sequences were 128D.



The Vacc-4x Domains Contain Pre-existing Mutations

To characterize the regions targeted by the vaccine, we identified cytotoxic T lymphocyte (CTL) epitopes within the four Vacc-4x domains based on each participant's HLA type. Identified variants were classified as the following: 1) a (sub-type specific) susceptible form; 2) alters epitope processing or documented escape according to the Los Alamos Epitope Variant and Escape Mutation Database; or 3) not described if the variant was not present in the database. Five out of the seven participants had mutations in a HLA-matched CTL epitope that were present in 100% of the sequences obtained.



CTL epitopes in Vacc-4x domains. A '–' indicates agreement with the HXB2 sequence. Any differences from the HXB2 amino acid sequence are indicated by a letter of the amino acid present. The percentages above the amino acids indicate the number of all obtained sequences from each participant which have the indicated amino acid at this site. Mutated CTL epitopes based on each individual's HLA type are highlighted and colored base on the type of variant.

Conclusions

- We detected no evidence of a selective immune pressure reflected in proviral diversity or by occurrence of specific mutation in the vaccine-targeted epitopes.
- Pre-existing CTL epitope mutations may affect the potency of this therapeutic vaccine.
- This highlights the challenges of developing effective HIV-1 therapeutic vaccines.