

Structured treatment interruption does not regenerate HIV-specific IFN- γ and IL-2 production but enhances broader HIV-specific perforin responses in a Ugandan population.

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Background:

Antiretroviral therapy (ART) delivery strategies that maximise clinical benefit despite limited drug availability are essential in resource poor settings. We hypothesised that continuous ART (CT) would not allow regeneration of HIV-specific cellular immune responses, whilst structured ART interruption (STI) would lead to the induction of responses of potential clinical benefit.

Methods:

Participants: HIV+ adults with <200 CD4 T-cells/ μ l were recruited from a cohort initiated on ART through the Development of AntiRetroviral Therapy (DART) trial. Participants who had attained 300 CD4 T-cells/ μ l after 52 or 76 weeks on ART were randomised into either STI (12 weeks on then 12 weeks off therapy [n=12]) or CT [n=15]. Heparinised peripheral blood mononuclear cells (PBMCs) were evaluated for HIV-specific cellular immune responses at baseline, 0 and 12 weeks of STI.

Immunological assays: Whole blood was stimulated for 6 hours with Gag (clades A and D), Nef, Vif, Rev, Tat, Vpr and Vpu (clade B) peptide pools. HIV-induced CD3+CD8+IFN- γ + /perforin+ and CD3+CD4+IFN- γ +/IL-2+ release was evaluated using intracellular cytokine staining. CD4 T-cell counts were also enumerated at this point. A positive response was determined as 2 confidence intervals above the the isotypic control and the mean negative background. The gating strategy for determining HIV-specific response is as shown below.

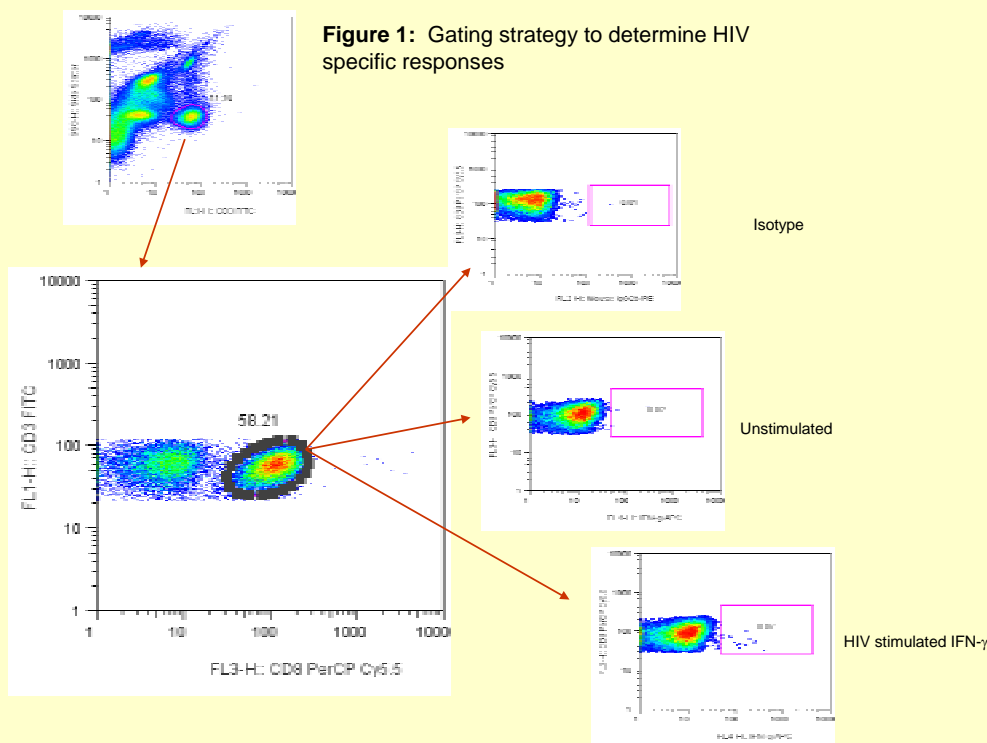


Figure 1: Gating strategy to determine HIV specific responses

A positive response was determined as HIV stimulated – (Mean Unstimulated + 2 confidence intervals) - Isotype

Statistical analysis: Statistical differences between two groups was evaluated using Fischer's Exact Chi square tests. Comparison of mean cellular immune responses between 2 groups will be evaluated using two-tailed unpaired t tests.

Results:

All peptide pools were recognised by at least one patient at pre-ART baseline with 78% (62/80) IFN-g+, 49% (39/80) perforin+ and 27% (21/77) IL-2+. CD3+CD8+. Pre-ART frequency of HIV-specific responses to the various HIV peptide pools is as shown in figure 2.

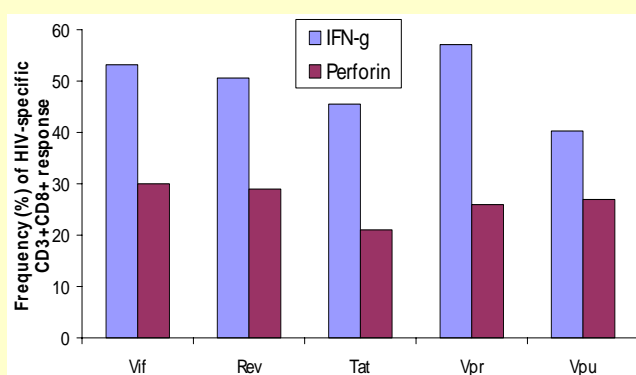


Fig. 2: Frequency of pre-ART baseline HIV-specific responses

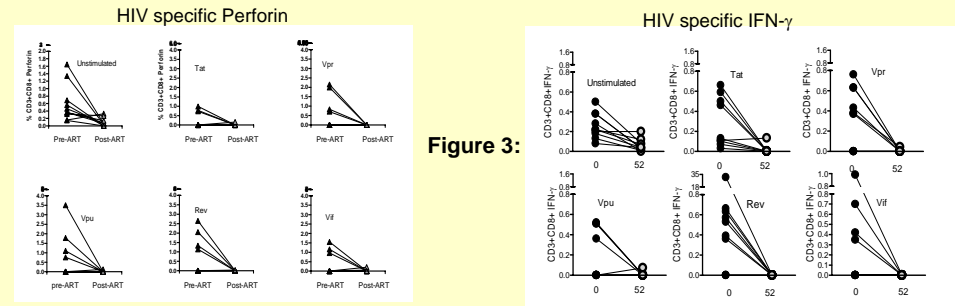


Figure 3:

Overall, there was decline in HIV-specific T cell responses following 52/76 of ART (Fig. 3)

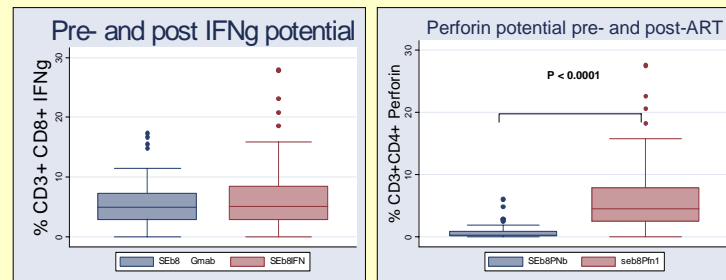


Figure 4: Potential for T-cell responses before and after ART as evaluated by stimulation with SEB

Overall, there was no difference in IFN- γ release potential following 52 or 76 weeks of ART as evaluated by stimulated with SEB. However the potential to release perforin significantly increased after the same period of treatment. ($P < 0.001$), Fig.4

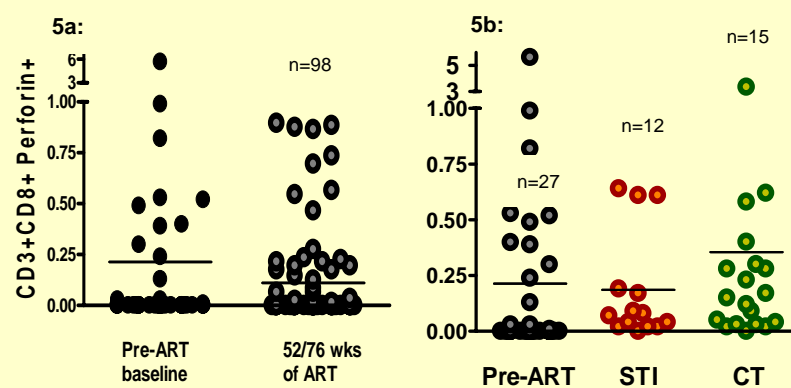


Fig 5: Potential for T cell Perforin responses as evaluated by stimulation with HIV peptide pools

There was no significant difference in IFN- γ release potential following 52 or 76 weeks of ART as evaluated by stimulated with HIV peptide pools, $p=0.19$ (5a). Additionally, there was no significant difference in perforin release potential between STI and CT participants when evaluated at the end of 12 weeks of randomisation (5b)

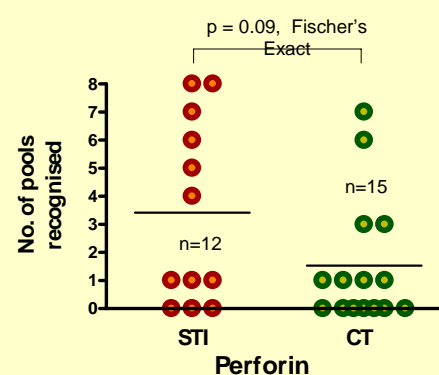


Figure 6: Breadth of T cell Perforin responses

Preliminary data reveals that STI participants tended to respond to more HIV peptide pools (6 of 12) compared to CT participants (2 of 15).

Conclusions:

Following the first round of STI, participants in the STI arm tended to broaden of HIV-specific CD3+CD8+ perforin+ but did not regenerate HIV-specific CD4 T cell responses. STI randomisation in this study has now been discontinued due to concerns related to clinical outcome.

Acknowledgement:



Study participants



The Development of AntiRetroviral Therapy (DART) trial team



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