Impact of Treatment Interruption on HIV Reservoirs and Immunologic Parameters

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Abstract

Background: Suppression of human immunodeficiency virus (HIV) and improvements in health outcomes have been achieved in infected individuals receiving antiretroviral therapy (ART). Nonetheless, the vast majority will experience plasma viral rebound upon cessation of therapy, underscoring the need for developing additional therapeutic strategies that allow durable virologic remission following the interruption of ART. Analytical treatment interruption (ATI) is an essential component of future clinical trial design to determine the efficacy of immune-based therapies in suppressing and/or eradicating HIV. Here, we investigated the effect of short-term ATI on the HIV reservoir and immunologic parameters in HIV-infected individuals.

Methods: In depth immunologic and virologic analyses were conducted using clinical specimens obtained from ten HIV-infected individuals prior to ART discontinuation, during ATI, and following reinitiation of ART. The effect of ATI on the HIV reservoir was determined by measuring the level of HIV proviral DNA, cell-associated HIV RNA, and replication-competent HIV in CD4$^+$ T cells. Characterization of intact and defective near full-length HIV proviral DNA was performed using single-genome, next-generation sequencing. Examination of immunologic parameters included longitudinal analyses of CD4$^+$ and CD8$^+$ T cells as well as cytokine and inflammation markers in plasma. Expression of activation and exhaustion markers was analyzed using flow cytometry.

Results: The median duration of the ATI phase was 57 days. All study participants experienced plasma viral rebound and resumed ART. HIV burden increased significantly in the CD4$^+$ T cells during plasma viral rebound. However, the size of the HIV reservoirs, including the frequency of CD4$^+$ T cells carrying replication-competent virus, returned to pre-ATI levels 6 to12 months after the study subjects resumed ART. Of note, the proportions of near full-length, genome-intact, and structurally defective HIV proviral DNA sequences were similar prior to ATI and following reinitiation of ART. Furthermore, no significant differences in immunologic or activation and exhaustion parameters were found between pre-ATI and post-ATI time points.

Conclusions: Our data indicate that short-term ATI is not associated with permanent expansion of the persistent HIV reservoirs nor irreversible immune system abnormalities. These findings support the inclusion of ATI in future clinical trials when evaluating strategies for achieving ART-free remission.

Background and Rationale

- The advent of antiretroviral therapy (ART) has led to improved health outcomes and the sustained suppression of human immunodeficiency virus (HIV)
- Cessation of ART results in plasma viral rebound in virtually all HIV-infected individuals
- Development of alternate therapeutic strategies aimed at achieving durable viral remission is under active investigation
- Analytic treatment interruption (ATI) is necessary to determine treatment efficacy in clinical trials testing therapeutic alternatives to ART
- Effects of short-term ATI on HIV reservoir and immunologic parameters are not well defined

Materials and Methods

Study Population: 10 HIV-infected individuals who previously participated in a passive antibody transfer study
Study Design: Specimens were collected and analyzed prior to ART discontinuation, during ATI, and following reinitiation of ART.
HIV Reservoir Quantification: Levels of HIV proviral DNA, cell-associated HIV RNA, and replication-competent HIV in CD4$^+$ T cells were measured.
Mutational Analyses: Intact and defective near full-length HIV proviral DNA were analyzed using single-genome, near full-length viral, next-generation sequencing.
Immunologic Parameters: Lymphocyte populations and CD8$^+$ T cell activation and exhaustion markers were measured using flow cytometry.

Table 1

Demographic and immunologic profiles of HIV-infected Individuals

Subject	Duration of viral suppression (years)	CD4$^+$ T cell count prior to ATI (per mm^3)	CD4$^+$ T cell % prior to ATI	CD8$^+$ T cell count prior to ATI (per mm^3)	CD8$^+$ T cell % prior to ATI	Plasma viremia prior to ATI (per ml)	Duration of ATI (days)	Duration of ART following reinitiation of ART (days)
N01	13.4	1,616	51	982	31	<40	59	373
N02	16.8	728	47	526	34	<40	46	407
N03	9.2	1,194	47	813	32	<40	55	418
N04	11.5	726	50	581	40	<40	114	343
N05	13.8	577	38	577	38	<40	69	353
N06	7.1	594	32	873	47	<40	45	386
N07	3.0	722	29	623	25	<40	22	409
N08	6.6	634	50	406	32	<40	67	302
N09	7.5	992	34	1,372	47	<40	42	305
N10	6.7	628	30	1,151	55	<40	115	140

Results

Figure 1

Levels of HIV DNA and plasma viremia in study participants

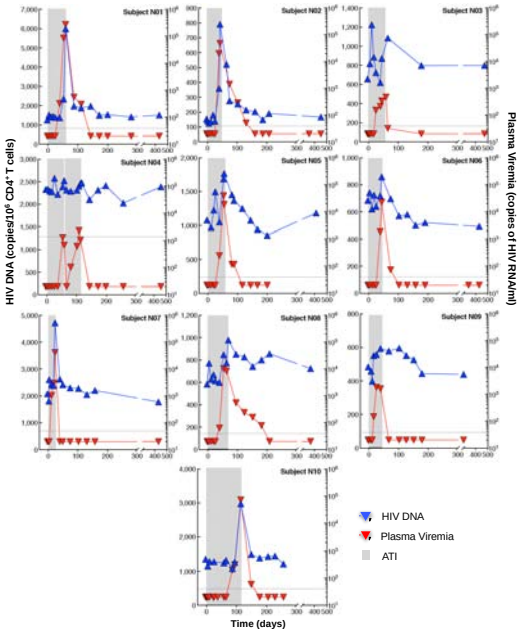


Figure 2

Impact of ATI on HIV Reservoirs

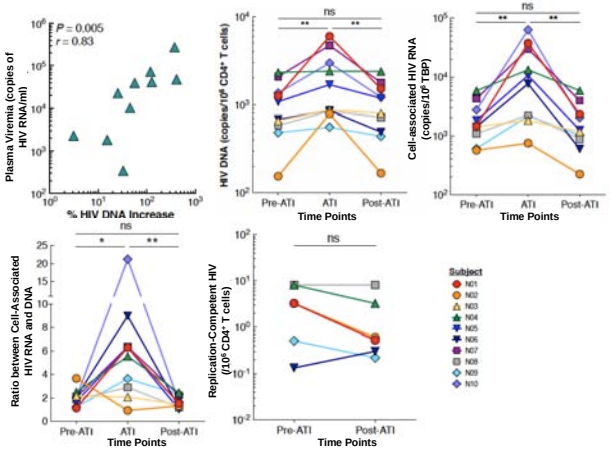


Figure 3

Structural characteristics of HIV DNA sequences

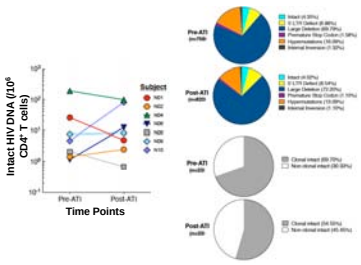


Figure 4

Longitudinal measurement of lymphocyte populations

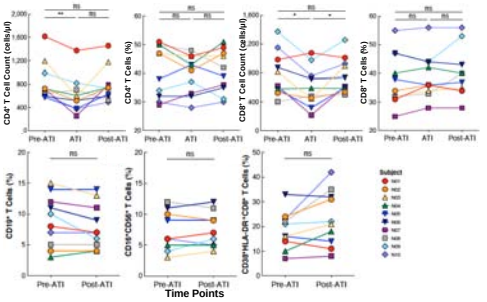
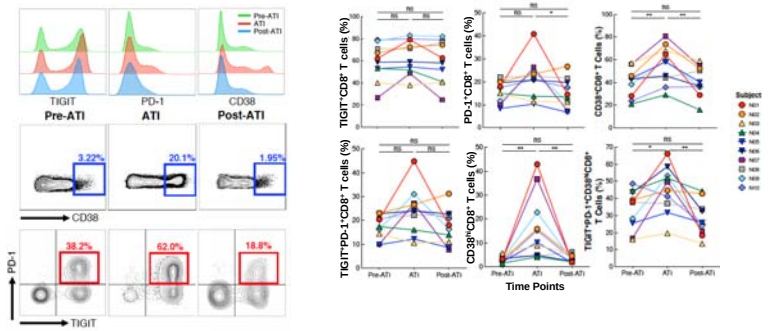


Figure 5

Levels of exhaustion and activation marker expression on CD8$^+$ T cells



Conclusions and Future Directions

- Short-term ATI does not result in permanent expansion of the HIV reservoir.
- Immunologic abnormalities were not found after reinitiation of ART.
- Our findings support the inclusion of ATI in future clinical trials investigating new therapeutic strategies for achieving ART-free remission.

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