Correlates of Viral Control after ART-interruption in SIV-infected Rhesus Macaques

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INTRODUCTION

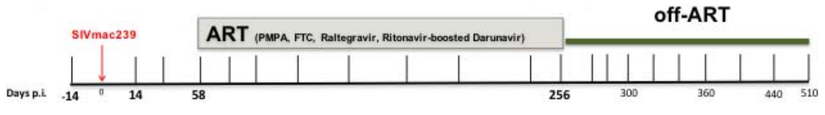
- Antiretroviral therapy (ART) does not eradicate HIV and the virus rebounds upon treatment interruption. Recently, a sustained control of HIV replication in the absence of ART has been achieved in a subset of individuals that have started ART early following infection (post-ART treatment controllers; PTC) (Saez-Cirion et al. PLoS Path 2013). Interestingly, most PTC lacked the protective HLA B alleles present in HIV elite controllers, and their $CD8^+$ T cell function was not superior to that in HIV non controllers (NC).
- The virologic and immunologic determinants of post-ART control of HIV replication are still unclear, particularly in tissues.

AIM

❖ Define the virologic and immunologic correlates of post-ART viral control using the ART-treated SIV-infected rhesus macaque (RMs) model.

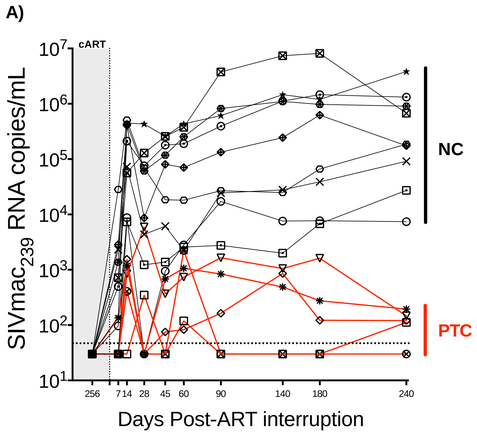
STUDY DESIGN AND METHODS

We performed a post-hoc analysis of fourteen RMs ($B*08^-$ and $B*17^-$) that show different levels of viral rebound after structured ART interruption (STI). All RMs were infected (i.v.) with $SIVmac_{239}$ (from F. Villinger, Emory University). Sixty days following infection, all RMs were treated with a 5-drug ART (PMPA, FTC, Raltegravir, and Ritonavir-boosted Darunavir) for seven months. ART was then interrupted and RMs were monitored for eight additional months. Blood (PB), lymph node (LN), and colorectal (RB) biopsies were collected throughout the study.

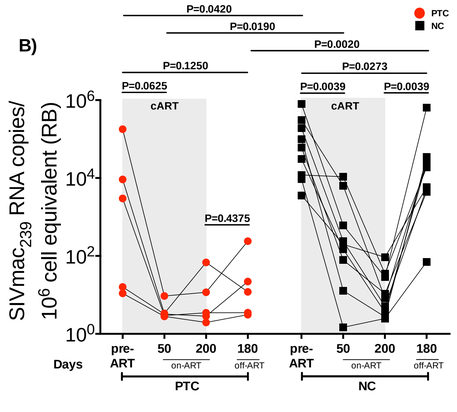


RESULTS

1.Five RMs partially controlled viral replication after STI

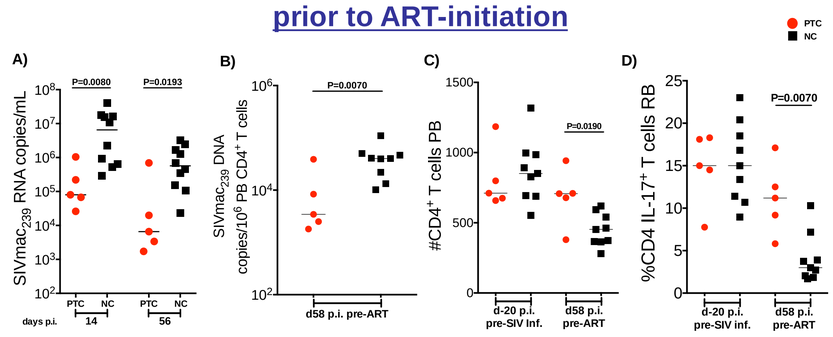


A. Longitudinal assessment of plasma viremia in ART-treated SIV-infected RMs. ART (shaded area) consisting of PMPA, FTC, Raltegravir, and Ritonavir-boosted Darunavir was very effective in suppressing SIV replication at <60 copies/mL (limit of detection) in all 14 RMs. After STI, five out of 14 RMs maintained peak plasma viral loads lower than 10^4 copies/mL, and controlled them at less than 200 copies/mL at 8 months without ART. These RMs were defined as **Post-ART Treatment Controllers (PTC,** in red).



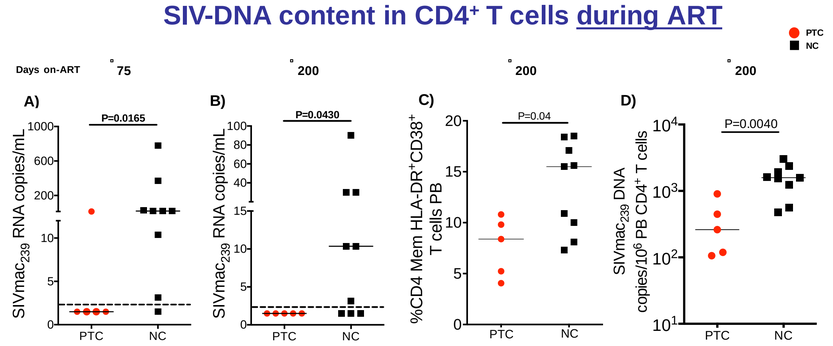
B. Longitudinal measurement of intestinal cell-associated (CA) SIV-vRNA in ART-treated SIV-infected RMs. RB CA-vRNA levels are consistent with those found in the plasma. Indeed, the number of SIVmac RNA copies significantly increased after STI in NC, but remained stable in PTC.

2. RM PTC show lower plasma viremia, SIV-DNA content, increased blood $CD4^+$ T cell counts and intestinal Th17 cells prior to ART-initiation



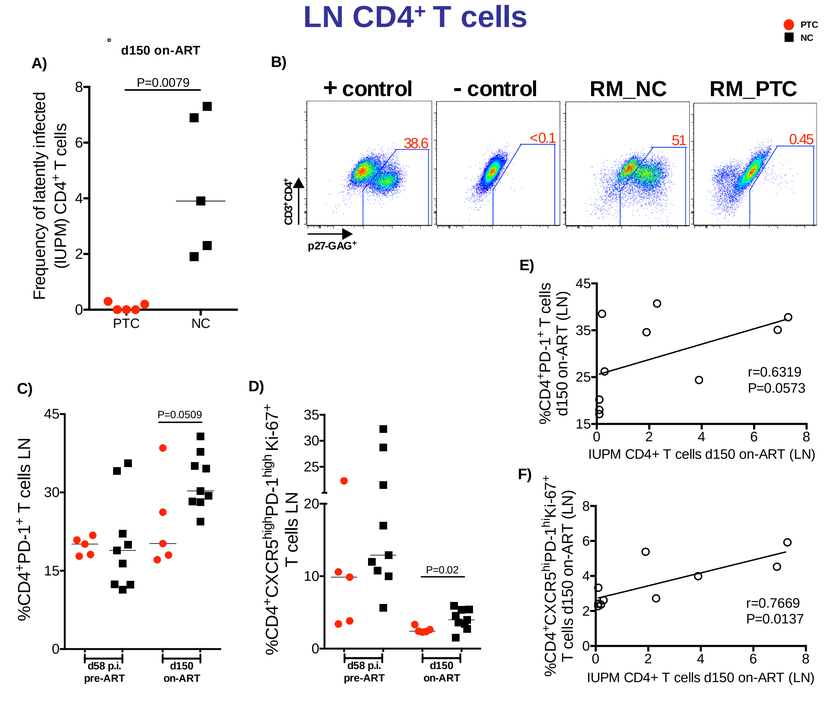
Prior to ART-initiation, PTC RMs (red) exhibited reduced peak and set-point viremia (**A**) as well as lower cell-associated SIV-DNA in purified blood $CD4^+$ T cells (**B**) than NC (black). This was associated with higher numbers of circulating $CD4^+$ T cells (**C**) and intestinal Th17 cells (**D**).

3. RM PTC show lower residual viremia, immune activation and SIV-DNA content in $CD4^+$ T cells during ART



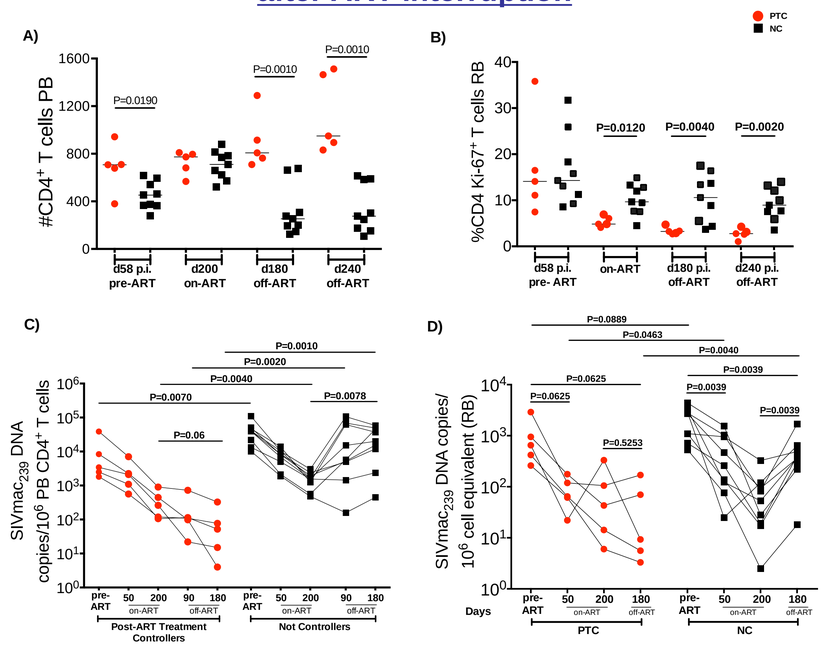
Longitudinal assessment of residual plasma viremia (**A, B**; limit of detection 3 copy/mL – dotted line). Levels of peripheral $CD4^+$ T cells HLA-DR^+CD38^+ (**C**) and cell-associated SIV-DNA in purified blood $CD4^+$ T cells (**D**) prior to ART-interruption in PTC (red) and NC (black).

4. RM PTC show lower frequencies of latently infected LN $CD4^+$ T cells



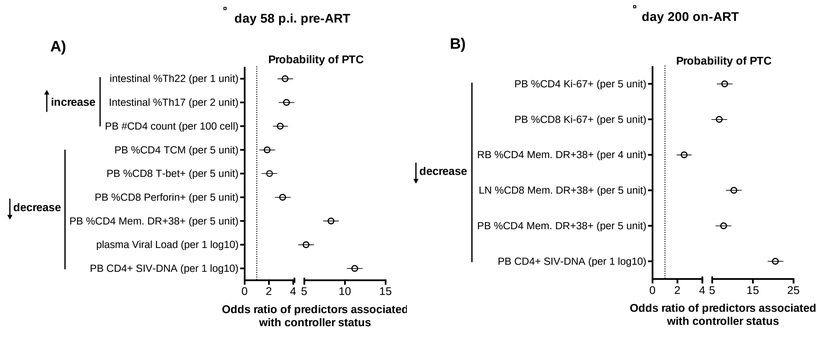
Frequencies (infectious unit per million; IUPM) of latently infected LN CD4+ T cells at d150 on-ART in PTC (red) and NC (black) (**A**). Representative staining for $CD4^+p27^+$ in QVOA assay (**B**). Levels of $CD4^+PD\text{-}1^+$ T cells (**C**) and proliferating TFH cells ($CXCR5^{hi}PD\text{-}1^{hi}Ki\text{-}67^+$; **D**) in the LN at d150 on-ART. In the LN, the frequency of latently infected $CD4^+$ T cells positively correlates with the levels of $CD4^+PD\text{-}1^+$ T cells (**E**) and proliferating TFH cells **(F)**.

5. RM PTC exhibit higher $CD4^+$ counts, reduced immune activation, and gradual decline of SIV-DNA content after ART-interruption



PTC maintained significantly higher numbers of $CD4^+$ T cells in blood (cells/μL; **A**) and lower levels of Ki-67 expression in intestinal $CD4^+$ T cells (**B**) up to eight months off-ART. Longitudinal levels of SIV-DNA content (**C,D**): PTC RMs showed a gradual decline of cell-associated SIV-DNA in blood $CD4^+$ T cells (**C**) and intestinal cells (**D**) after ART-discontinuation. PTC in red and NC in black.

6. Immunologic and virologic parameters associated with increased probability of becoming PTC



Correlates associated with likelihood of PTC status were calculated using univariable logistic regression analysis at pre-ART (**A**) and ART-interruption (**B**), and are expressed as odds ratios for the measure variations indicated for each value.

CONCLUSIONS

In SIV_{mac239}-infected RMs, sustained viral control after ART interruption was associated with:

- A less progressive SIV infection, with reduced viremia, lower SIV-DNA content, higher $CD4^+$ T cell counts, as well as lower levels of T cell activation and proliferation. Of note, CTL responses were comparable between PTC and NC.
- Lower levels of residual plasma viremia, SIV-DNA content, and frequencies of latently infected cells during ART. In LN, the frequency of latently infected cells associates with the levels of PD-1 expression in $CD4^+$ T cells and TFH proliferation.
- Virologic and immunologic benefits after ART interruption, with preserved blood and tissue $CD4^+$ T cells, limited immune activation, and gradual decline of SIV-DNA content.

ACKNOWLEDGEMENTS

We acknowledge the animal care and veterinary staff at YNPRC and the Emory CFAR Virology Core. This work was supported by NIH grants RO1-AI116379, amfAR 109109-57-RGRL, and in part by ORIP/OD P51OD011132 to YNRPC.