

SUPPLEMENTARY MATERIAL (ONLINE ONLY) for

ART Interferes with Lentivirus Mediated Gene Transfer in Hematopoietic Stem Cells

Patrick M. Younan,^{1*} Christopher W Peterson,^{1*} Patricia Polacino,^{2,3} John P. Kowalski,¹
Willimark Obenza,¹ Hannah W. Miller,⁴ Brian P. Milless,¹ Phil Gafken,¹ Stephen C. DeRosa,⁴
Shiu-Lok Hu,^{2,3} and Hans-Peter Kiem.^{1,2}

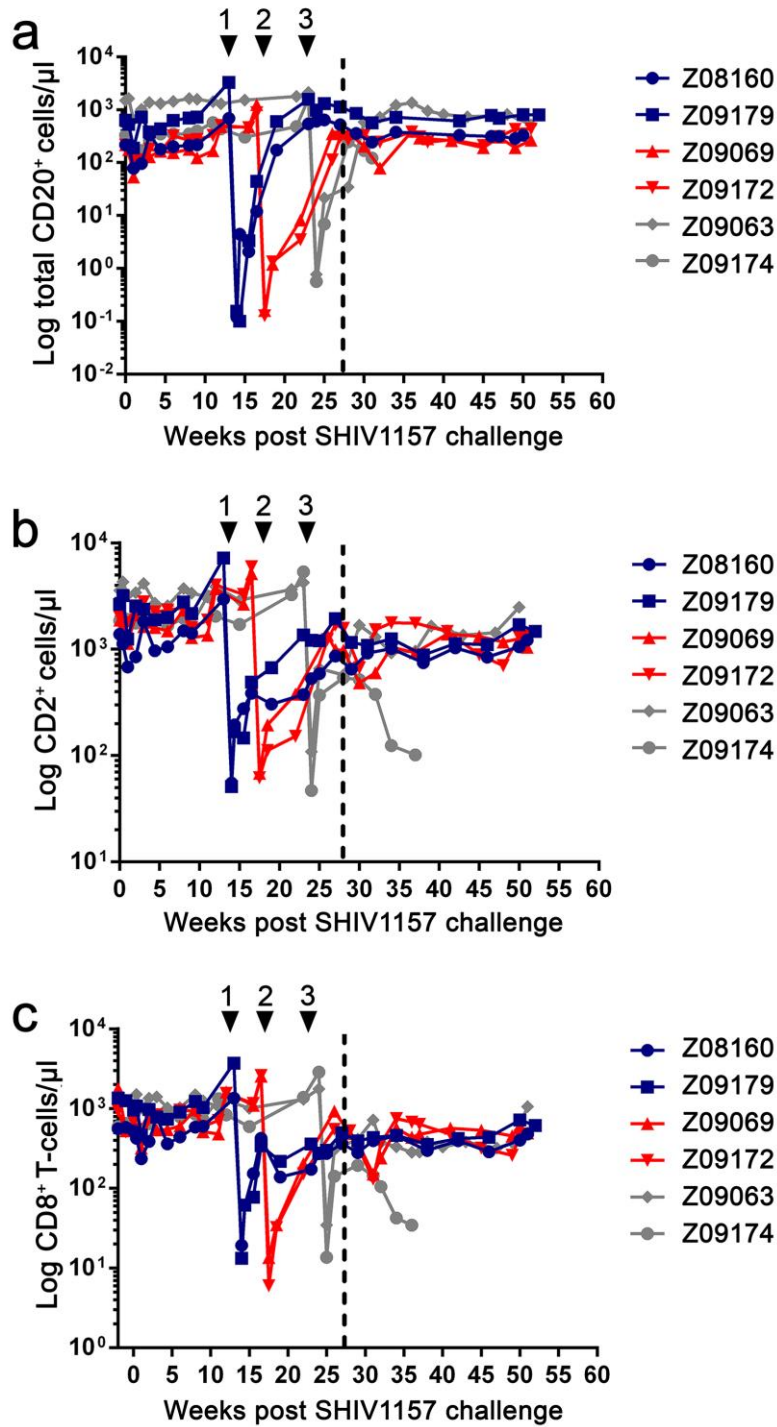
LIST OF SUPPLEMENTARY MATERIAL

Supplementary Figure S1. Lymphocyte analysis

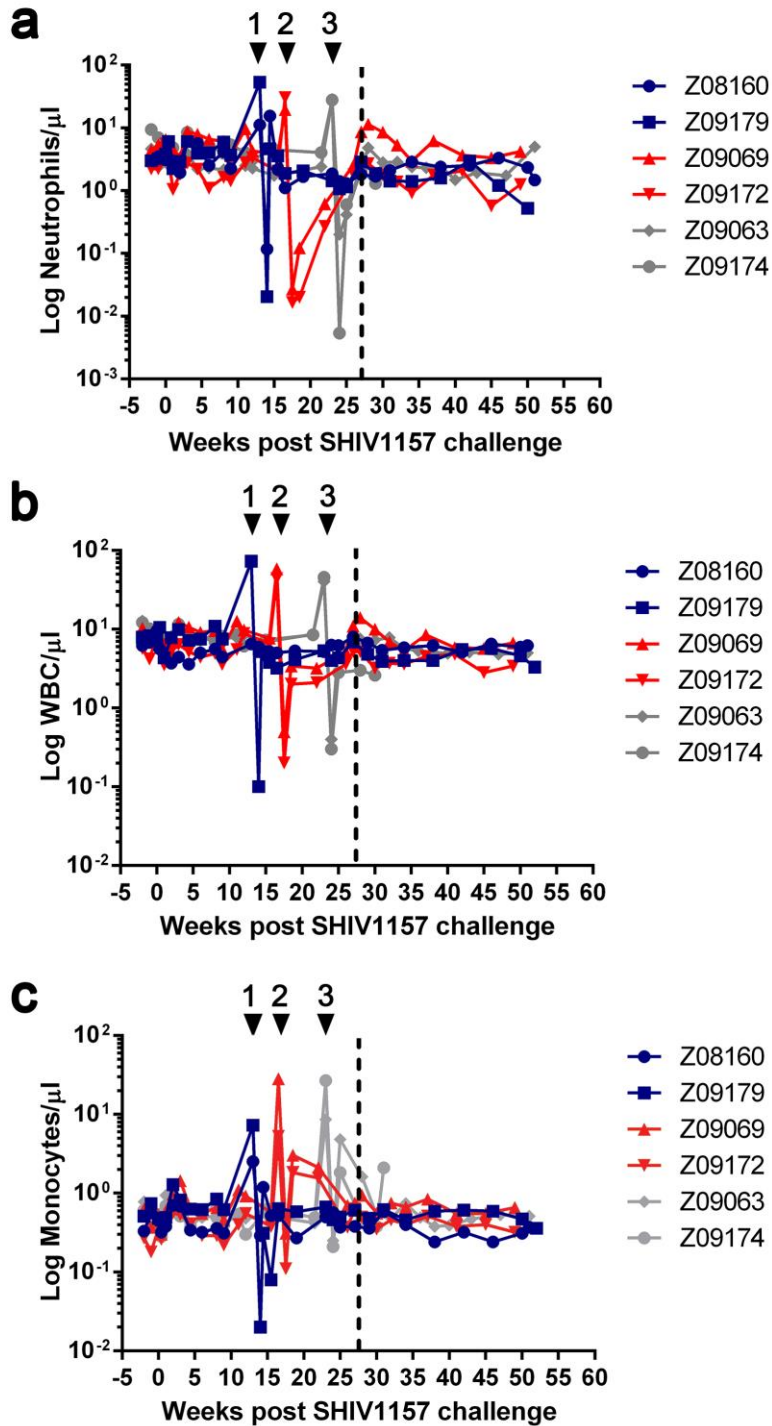
Supplementary Figure S2. ART-treated, SHIV-infected macaques exhibit normal hematopoiesis following HSCT.

Supplementary Figure S3. Activation status of peripheral T-lymphocytes.

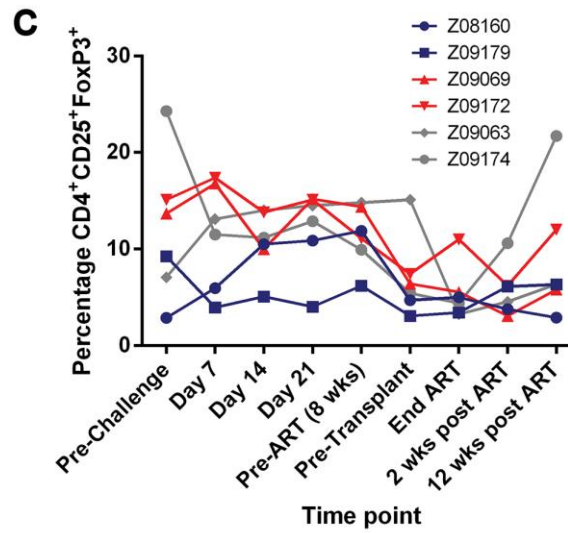
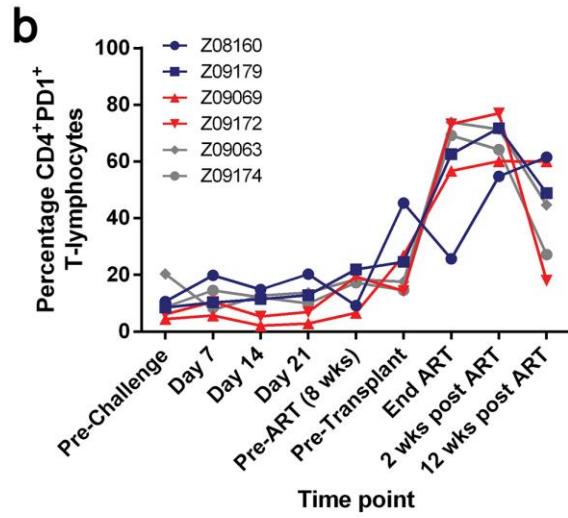
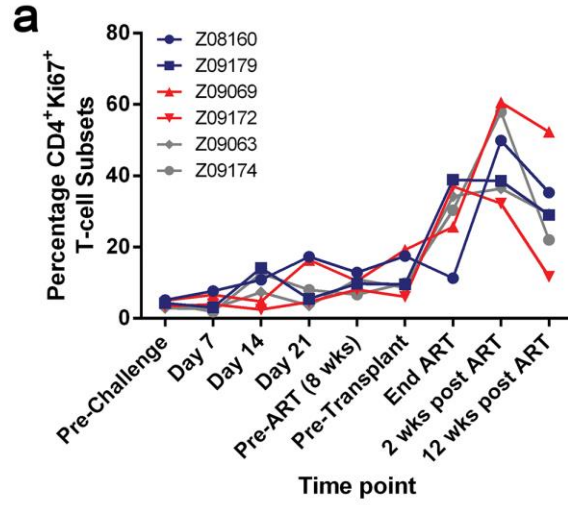
Supplementary Figure S4. Antibody production and neutralizing activity is maintained following HSCT



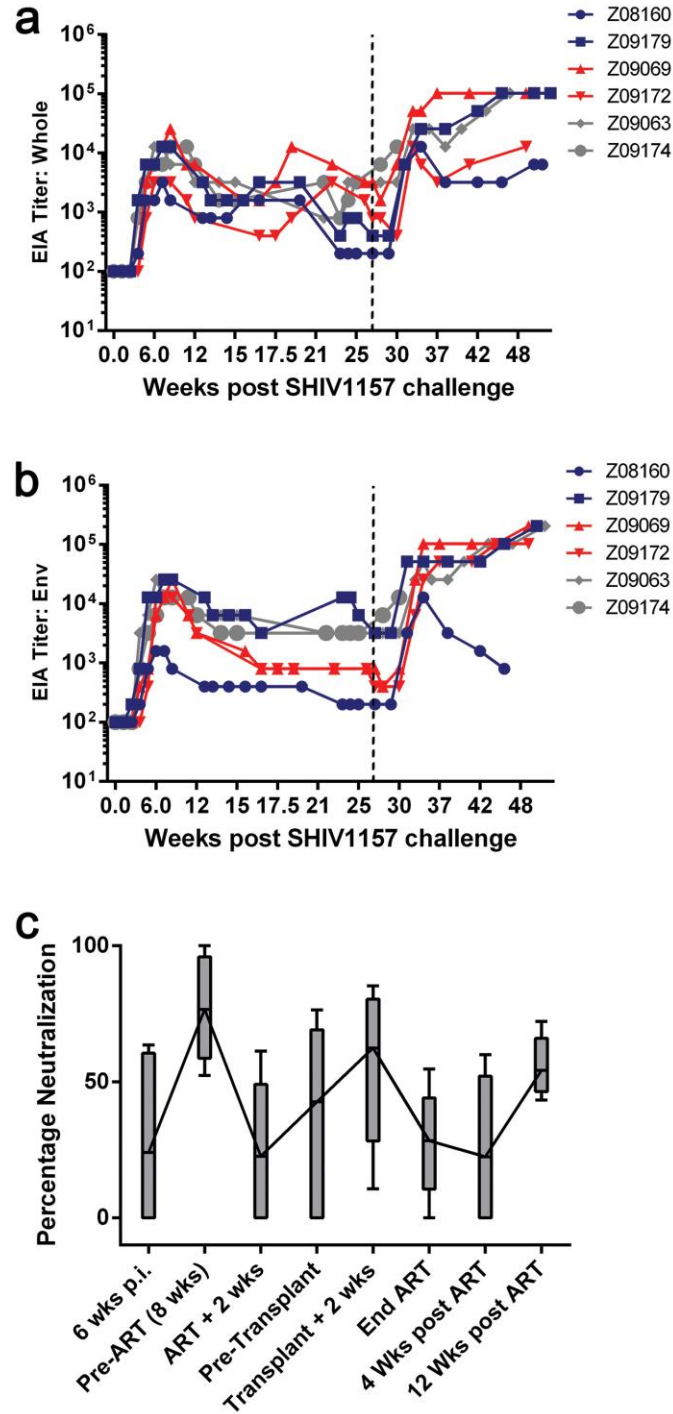
Supplementary Figure S1: Lymphocyte analysis. The absolute number of B-cells (CD2⁺CD20⁺), total lymphocytes (CD2⁺) and CD2⁺CD8⁺ T-cells was determined throughout the time course by flow cytometry. Arrows indicate HSCT and dotted line indicates end of ART.



Supplementary Figure S2: ART-treated, SHIV-infected macaques exhibit normal hematopoiesis following HSCT. The absolute number of (a) neutrophils, (b) white blood cells and (c) monocytes was determined prior to and following HSCT.



Supplementary Figure S3: Activation status of peripheral T-lymphocytes. (a) The percentage of activated CD4⁺ T-cells was determined using the antibody staining panel described in the *Materials and Methods* section. CD4⁺ T-cells co-staining for the activation marker Ki67 were detected by flow cytometry. (b) The relative percentage of PD-1 expression was used to determine the extent of immune exhaustion at the indicated time points. (c) The percentage of regulatory CD4⁺ T-cell (T_{regs}) was determined by staining for CD25 and FoxP3.



Supplementary Figure S4: Antibody production and neutralizing activity is maintained

following HSCT. Virus specific antibody responses were determined by ELISA against (a) HIV Env and (b) SIVmac 239 whole virus. Endpoint titer was determined based on an optical absorbance value of at least 3-fold higher than average values obtained from SHIV-negative

macaque serum. (c) Serum neutralization activity was determined in samples collected at the indicated time points and compared to neutralization activity in pre-infection serum.