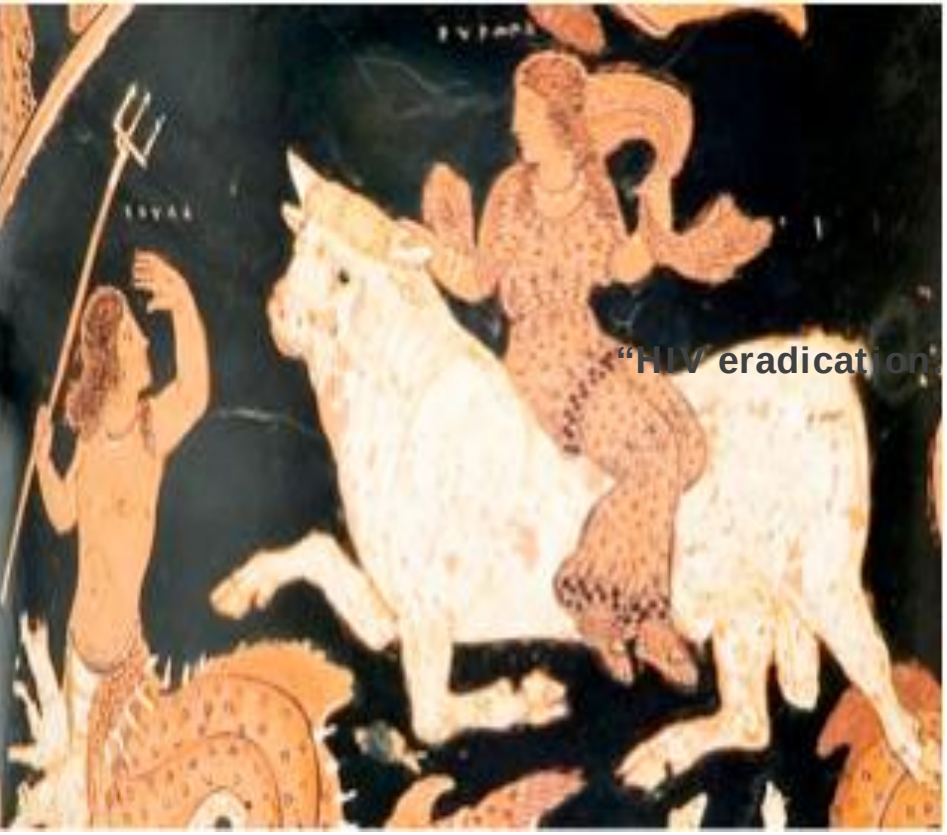
“L’infettivologia del 3° millennio:

AIDS ed altro”



VI Convegno Nazionale

Centro Congressi dell’Hotel Ariston di Paestum (SA)

15- 16 -17 maggio 2014

“HIV eradication: presente e futuro”

HIV eradication presente e futuro

Antonella Castagna
IRCCS San Raffaele

Definitions

- Functional cure

  Permanent control of HIV replication without eradicating it

- Sterilising cure

  Elimination of all HIV infected cells

Barriers to a cure for HIV: new ways to target and eradicate HIV-1 reservoirs

Christine Katlama, Steven G Deeks, Brigitte Autran, Javier Martinez-Picado, Jan van Lunzen, Christine Rouzioux, Michael Miller, Stefano Vella, Joern E Schmitz, Jeffrey Ahlers, Douglas D Richman, Rafick P Sekaly

	Intervention	Study population (patients chronically infected with HIV-1)		ClinicalTrials.gov
		Study population	Main endpoint	
ERAMUNE 01	Interleukin 7+ART intensification with raltegravir and maraviroc	29 patients with HIV RNA <50 copies per mL on ART and HIV DNA between 10 and 1000 copies per 10^6 PBMC	Decrease of 0·5 log_{10} HIV DNA from baseline at 56 weeks	NCT01019551
ERAMUNE 02	HIV rAD5 vaccine+ART intensification with raltegravir and maraviroc	28 patients with HIV RNA <50 copies per mL on ART and HIV DNA 10–1000 10^6 PBMC; negative AD5 antibody	Decrease of 0·5 log_{10} HIV DNA from baseline at 56 weeks	NCT00976404
Disulfiram	Disulfiram: 500 mg/day for 14 days	20 patients on ART with HIV RNA <50 copies per mL	2 weeks frequency of replication competent HIV-1 in resting CD4 T cells	NCT01286259
Vorinostat	Vorinostat: 200–600 mg per day	30 patients with HIV RNA <30 copies per mL	HIV expression in resting CD4 T cells	NCT01319383
Vorinostat	Vorinostat: 400 mg/day	20 patients with HIV RNA <50 copies per mL and CD4 cell >500 per mL	HIV unspliced RNA in resting CD4 T cells at 28 days	NCT01365065
Panobinostat	Panobinostat: 20 mg three times a week, every other week for 8 weeks	16 patients with suppressed viraemia and CD4 cell count >500 per mL	HIV unspliced RNA in resting CD4 T cells change from baseline	NCT01680094
CD4 T cells modified at CCR5 by zinc-finger nuclease	Autologous CD4 T cells modified at CCR5 gene by zinc-finger nuclease SB-728-T	30 patients with suppressed viraemia and CD4 cell count <300 per mL or failing ART	Safety; persistence and activation of CCR5+zinc-finger nuclease-modified autologous T cells	NCT01252641
CD4 T cells modified at CCR5 by zinc-finger nuclease	Autologous CD4 T cells modified at CCR5 gene by zinc-finger nuclease	18 patients: three cohorts of HIV-positive patients, either failing ART or with suppressed viraemia	Safety	NCT00842634
Lentivirus vector rHIV7-shI-TAR-CCR5RZ-transduced haemopoietic progenitor cells	Autologous CD34+haemopoietic cells modified by lentivirus-transduced non-functional *CCR5RZ* gene	10 patients with AIDS-related lymphoma undergoing haemopoietic stem-cell transplantation	Safety and durability of transduced cells	NCT00569985
Interferon alfa-2b	Interferon alfa-2b intensification	Recruiting, non-randomised, one-group assignment	Efficacy: viral RNA levels in blood and sequence diversification	NCT01295515

ART=antiretroviral therapy. PBMC=peripheral blood mononuclear cells.

Table 1: CURE-related clinical pilot trials in progress in 2012

HIV eradication: key points

- ART does not eliminate HIV-1 from latently infected reservoir
- An effective vaccine againsts HIV is still elusive
- HIV eradication will likely require multiple approaches

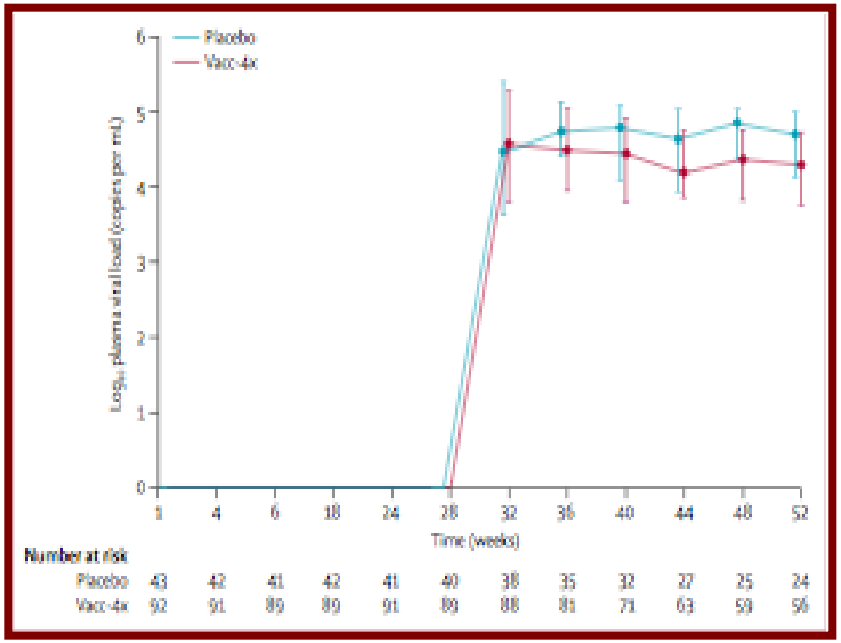
  Limiting HIV reservoir

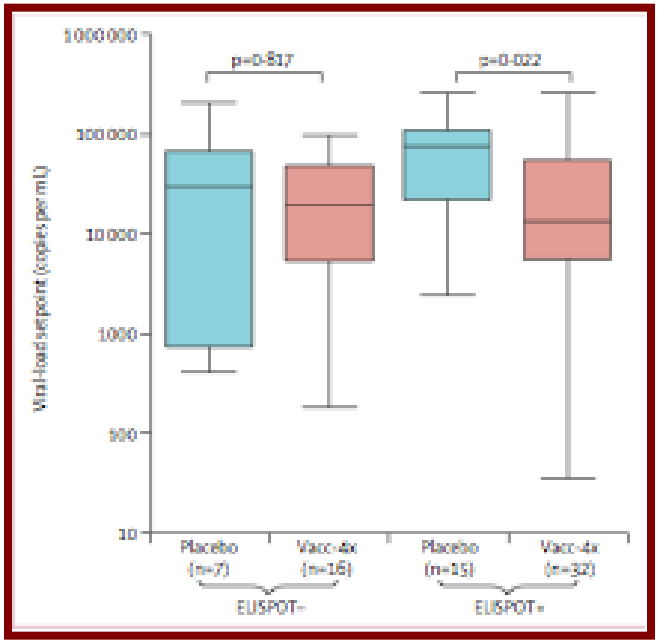
  Disrupting the state of virological latency

  Clearing residual HIV

Safety and efficacy of the peptide-based therapeutic vaccine for HIV-1, Vacc-4x: a phase 2 randomised, double-blind, placebo-controlled trial

Richard B Pollard, Jürgen K Rockstroh, Giuseppe Pantaleo, David M Asmuth, Barry Peters, Adriana Lazzarin, Felipe Garcia, Kim Ellefsen, Daniel Podzamczer, Jan van Lunzen, Keikawus Arasteh, Dirk Schürmann, Bonaventura Clotet, W David Hardy, Ronald Mitsuyasu, Graeme Moyle, Andreas Plettenberg, Martin Fisher, Gerd Fätkenheuer, Margaret Fischl, Babafemi Taiwo, Ingebjørg Baksaas, Darren Jolliffe, Stefan Persson, Øyvind Jelmert, Arnt-Ove Hovden, Maja A Sommerfelt, Vidar Wendel-Hansen, Birger Sørensen



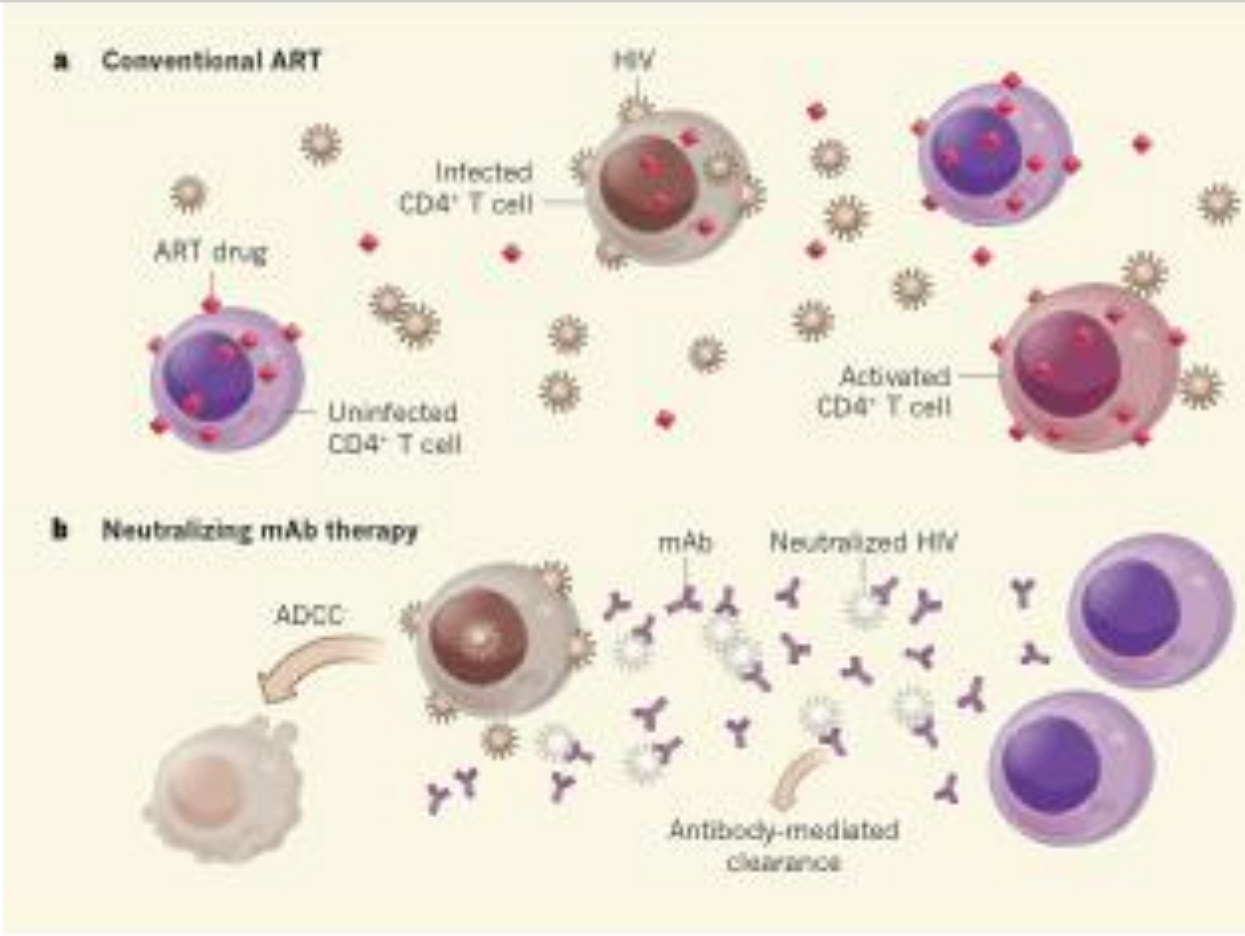


136 patients : coprimary end-points: ART resumption and CD4 change during TI

Lancet, 2014

Antibodies advance the search for a cure

Efforts to make a prophylactic HIV vaccine have identified monoclonal antibodies that potently suppress viral replication. Studies in monkeys show that these reagents effectively treat HIV infection. SEE ARTICLE P.224 & LETTER P.277



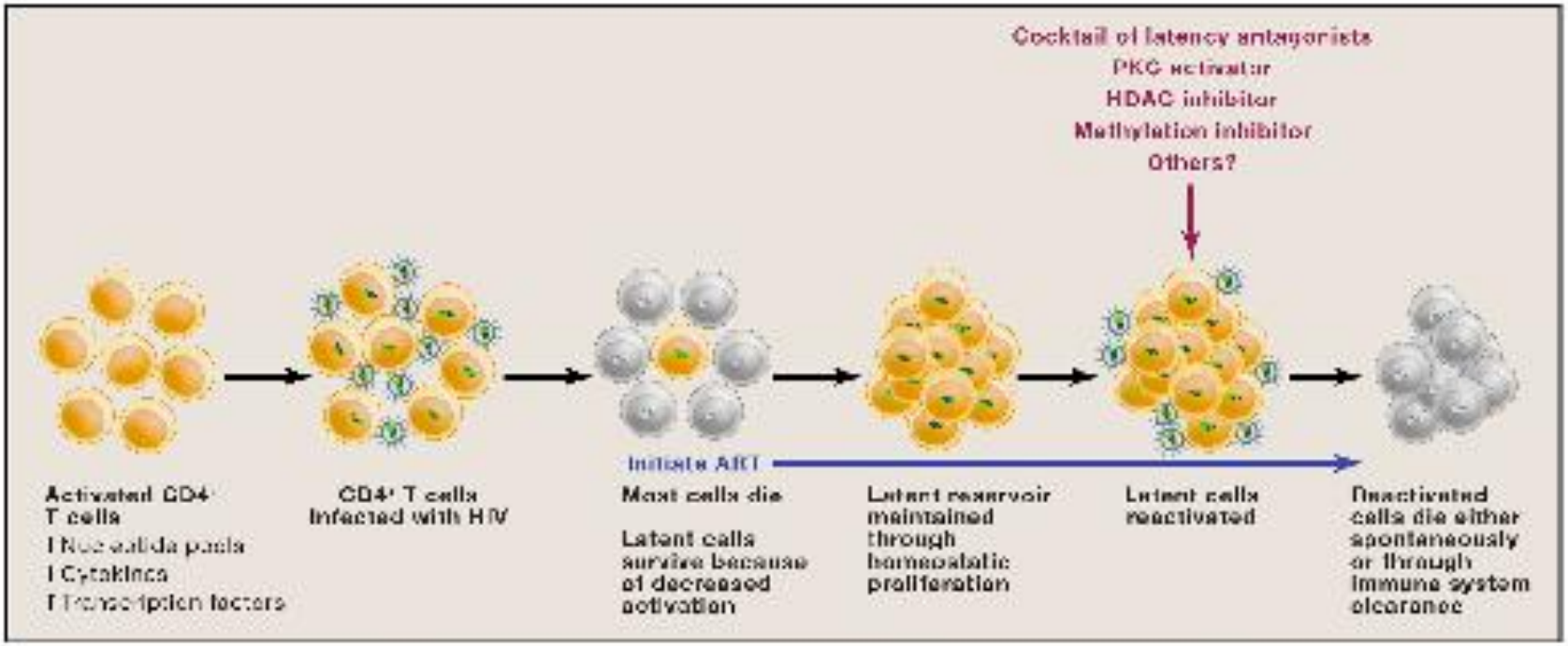
Picker LJ, Nature 2013

Leading Edge
Review

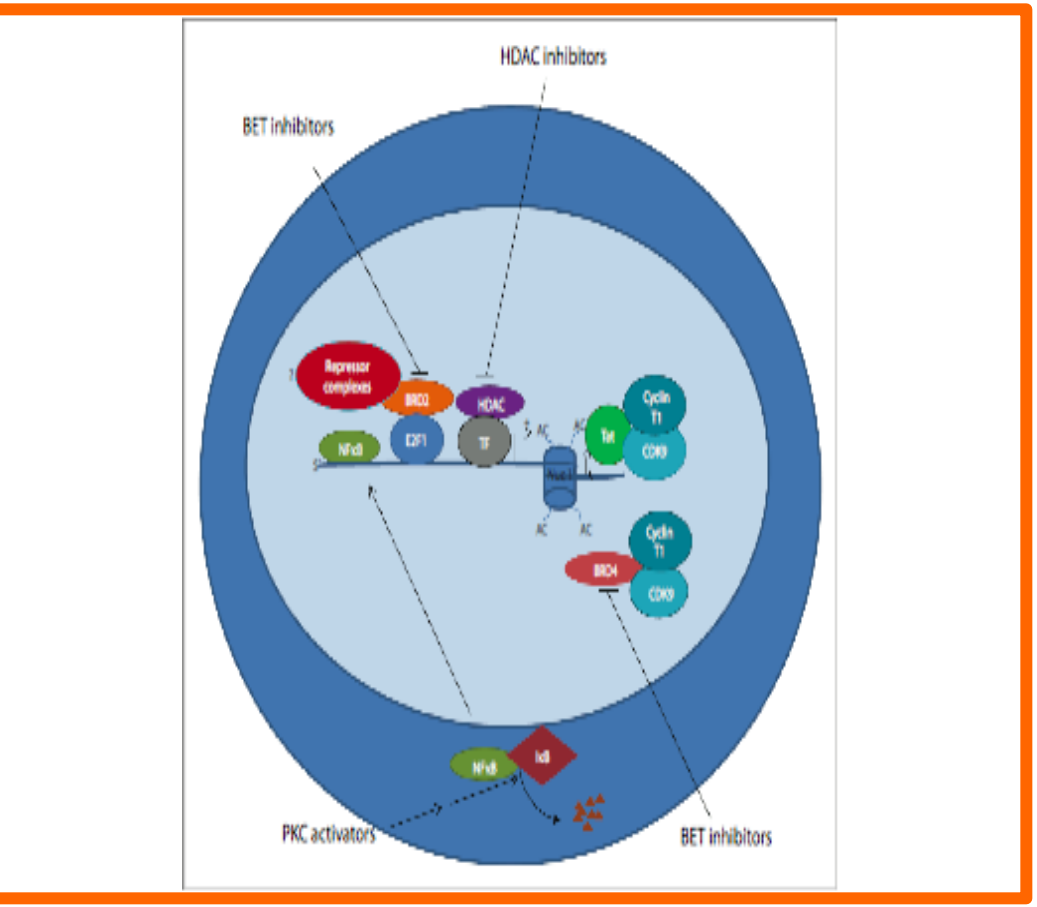
Cell

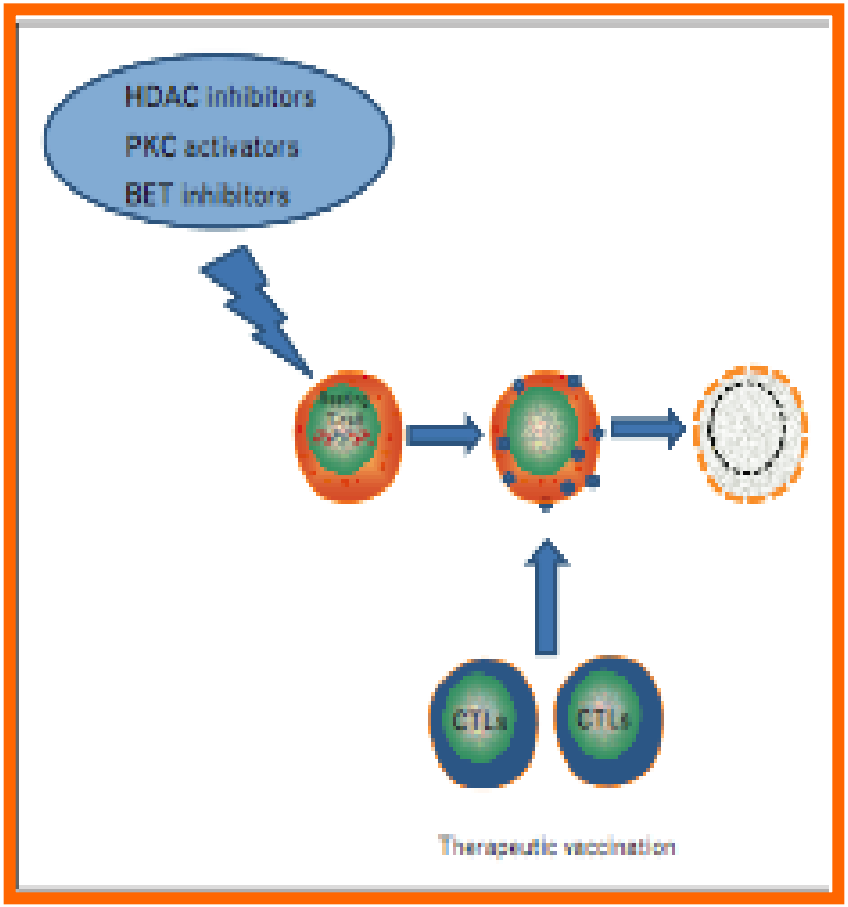
An Integrated Overview of HIV-1 Latency

Debbie S. Ruelas[1,2] and Warner C. Greene[1,3,4,*]



Pharmacologic reactivation of the virus as a cure therapy



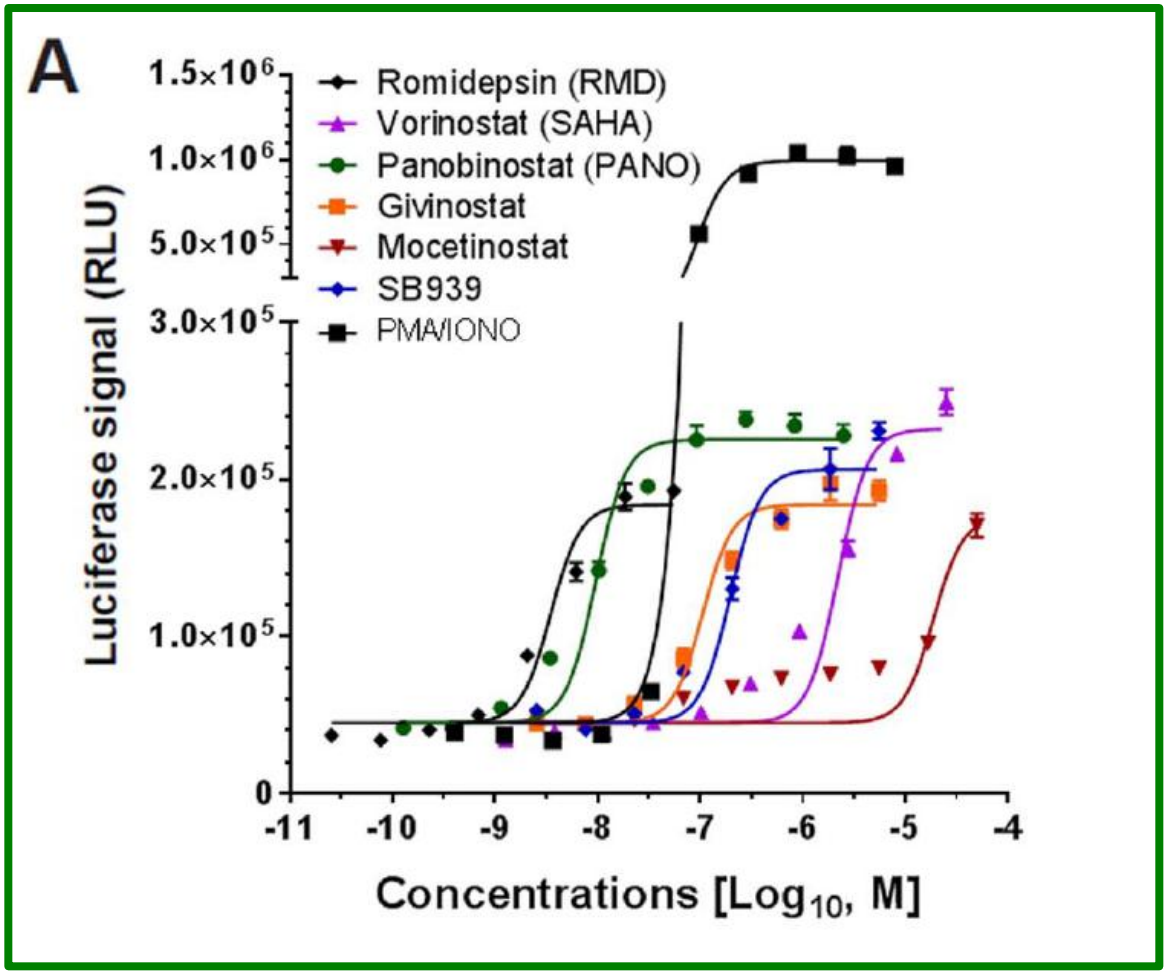


Archin NM et al, Curr Opin Infect Dis 2014

OPEN ACCESS Freely available online

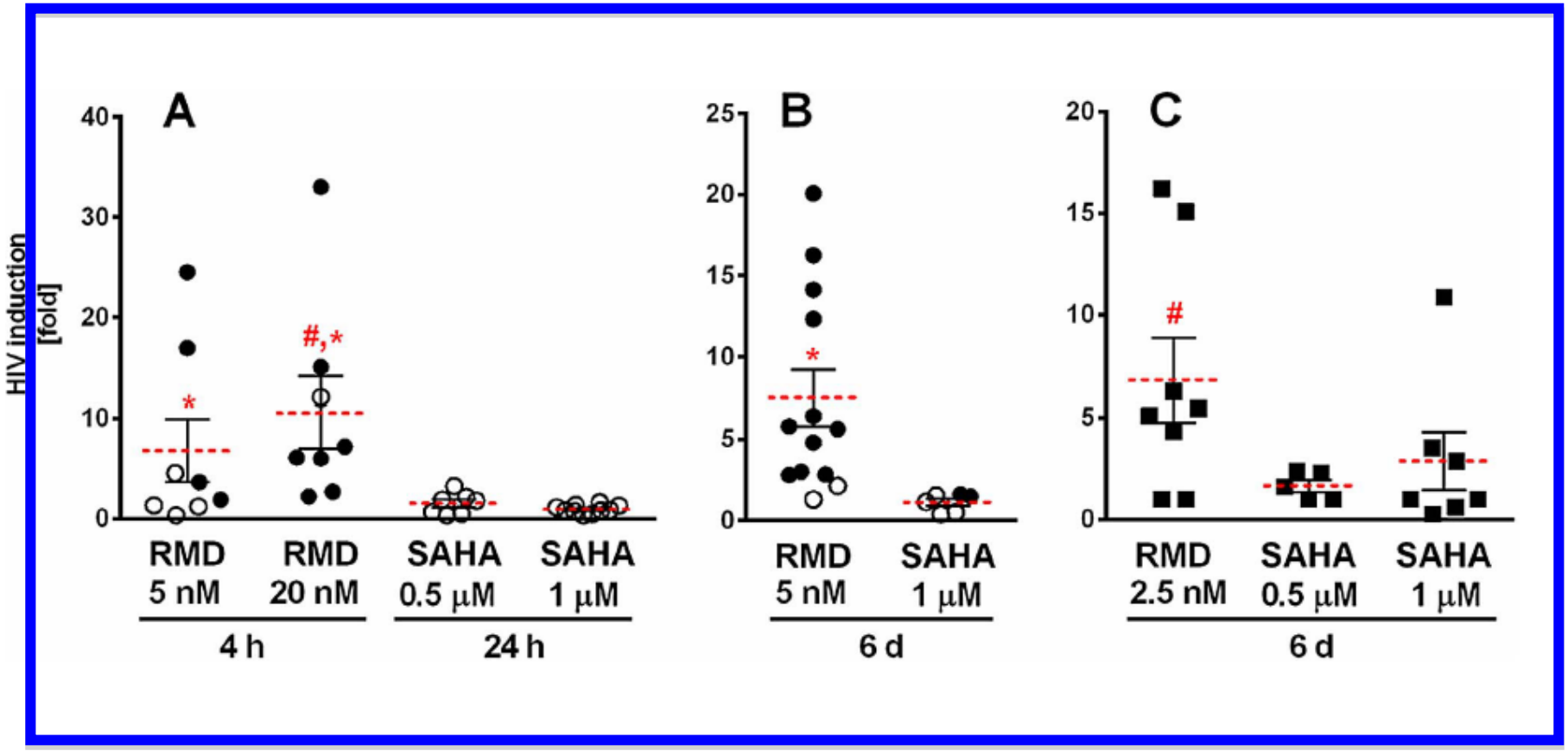
PLOS | PATHOGENS

Histone Deacetylase Inhibitor Romidepsin Induces HIV Expression in CD4 T Cells from Patients on Suppressive Antiretroviral Therapy at Concentrations Achieved by Clinical Dosing



WEI DG, 2014

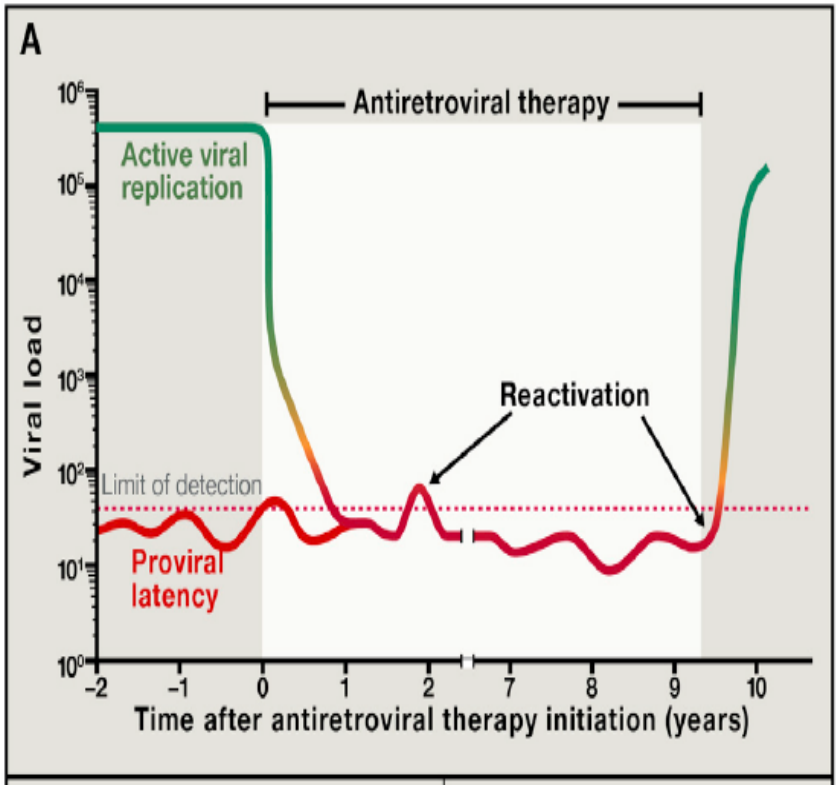
ROMIDEPSIN

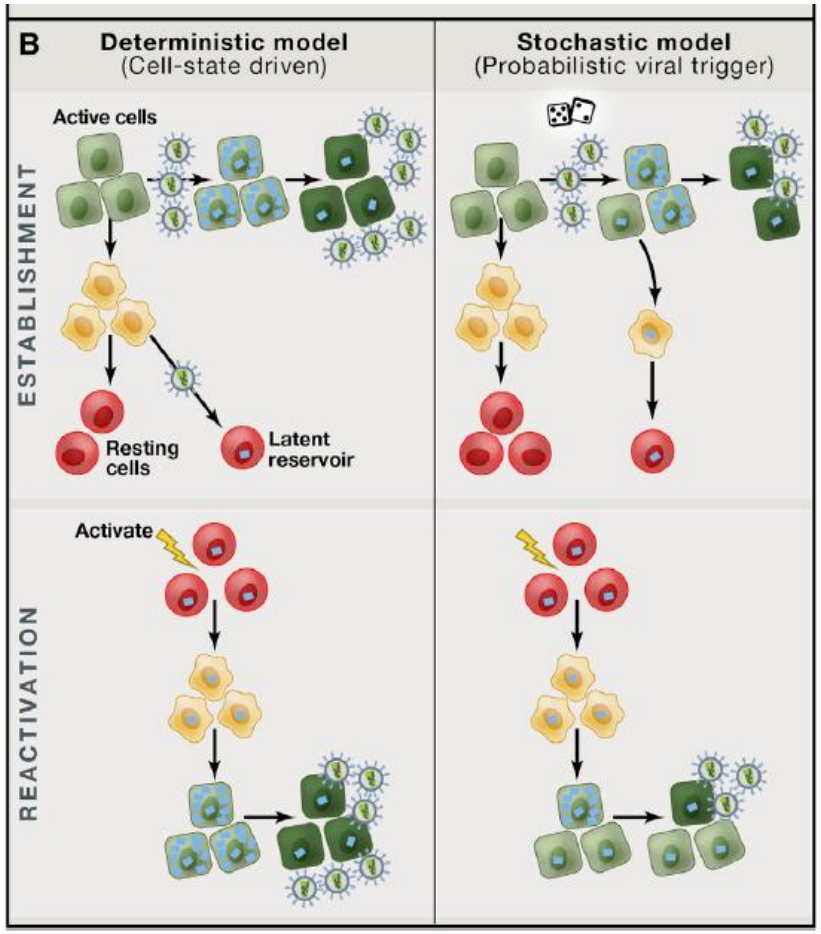


Wei DG et. al, PLOS pathogens 2014

Stochastic Fate Selection in HIV-Infected Patients

Cell



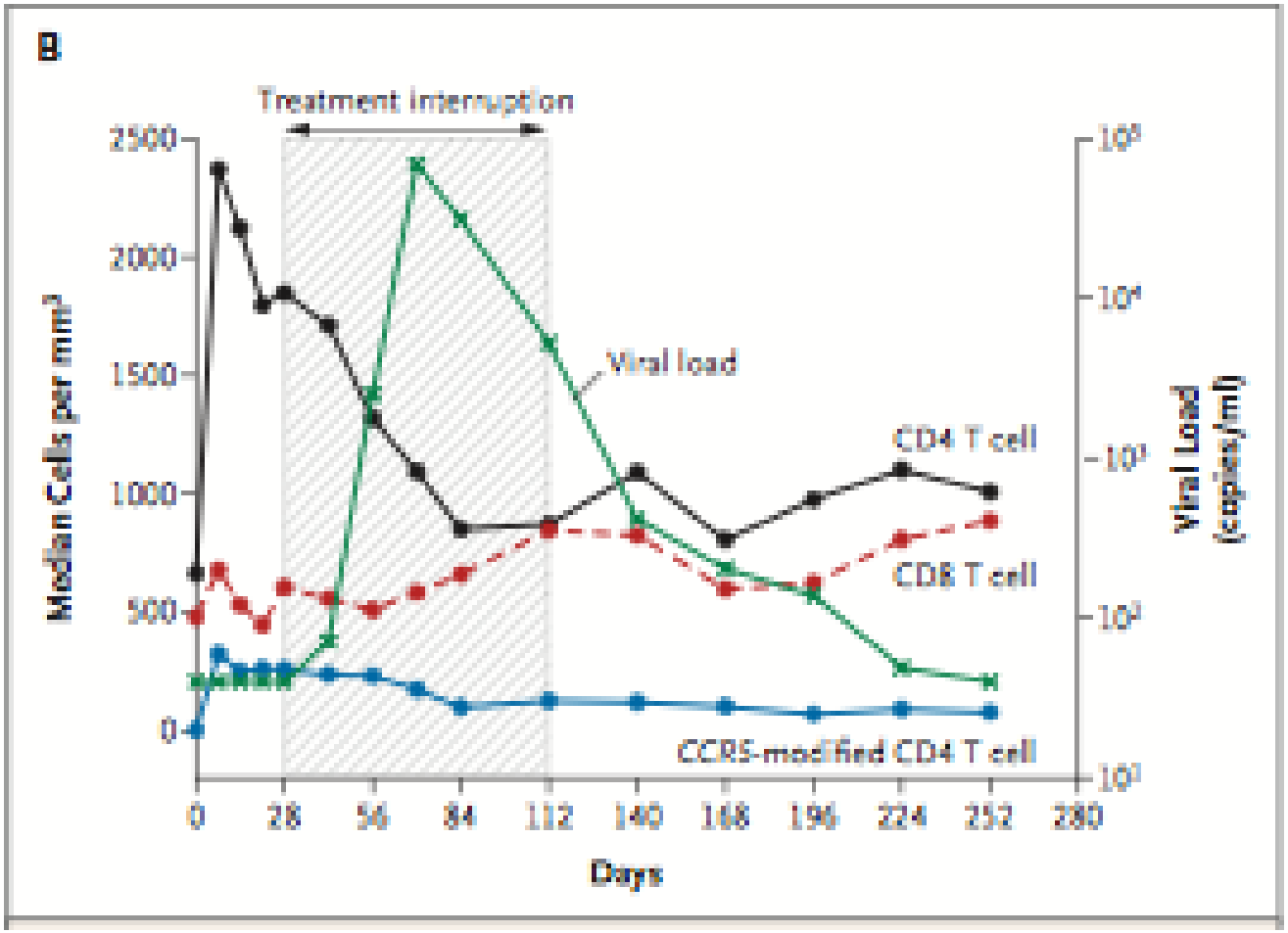


Weinberger AD, Cell 2014

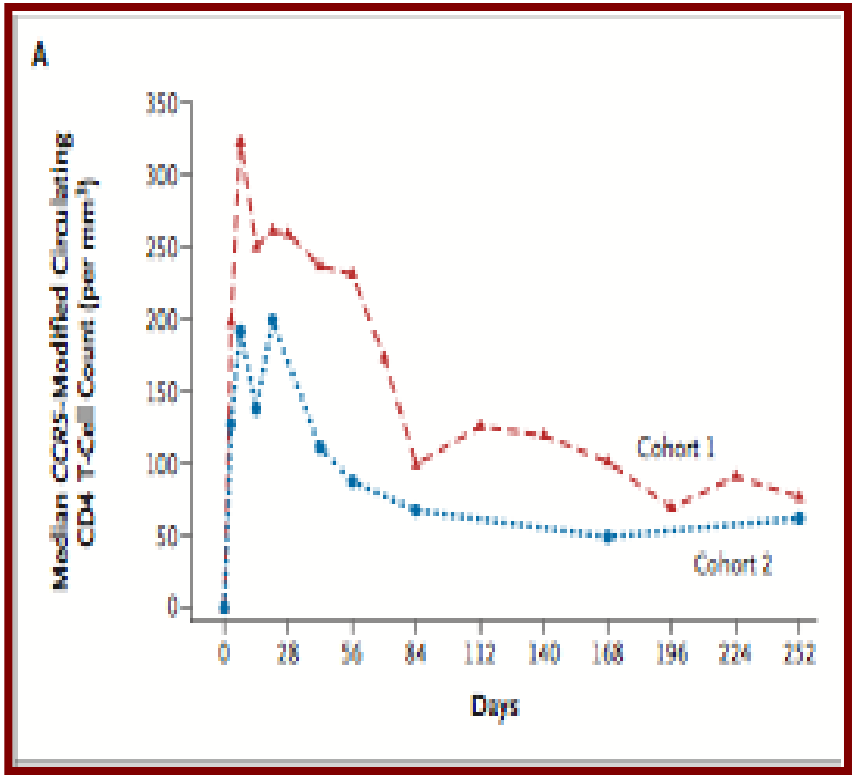
ESTABLISHED IN 1812 MARCH 6, 2014 VOL. 370 NO. 10

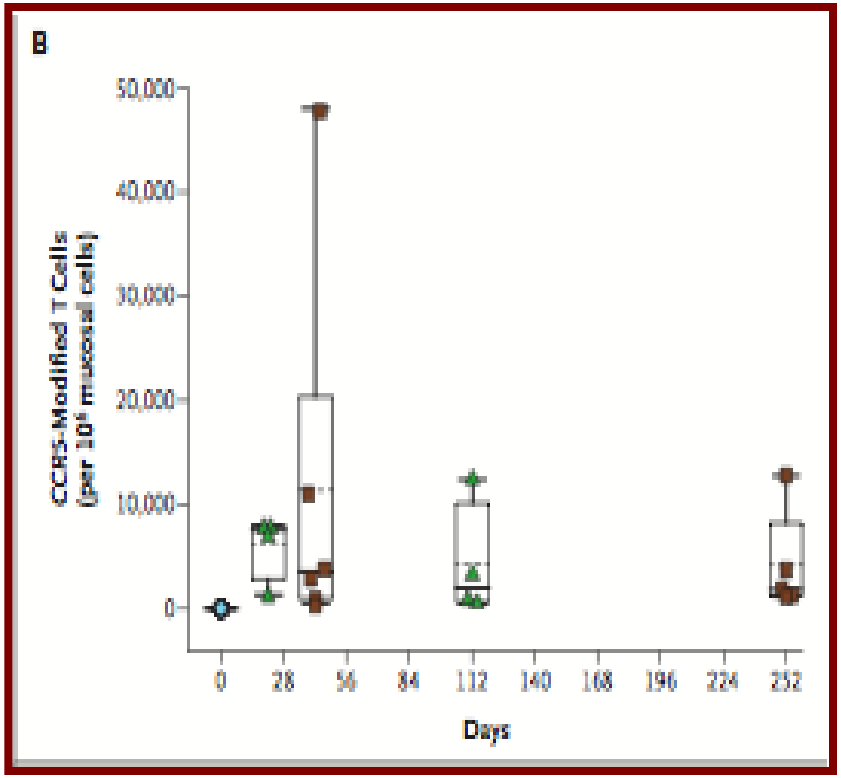
Gene Editing of *CCR5* in Autologous CD4 T Cells of Persons Infected with HIV

Pablo Tebas, M.D., David Stein, M.D., Winson W. Tang, M.D., Ian Frank, M.D., Shelley Q. Wang, M.D., Gary Lee, Ph.D., S. Kaye Spratt, Ph.D., Richard T. Surosky, Ph.D., Martin A. Giedlin, Ph.D., Geoff Nichol, M.D., Michael C. Holmes, Ph.D., Philip D. Gregory, Ph.D., Dale G. Ando, M.D., Michael Kalos, Ph.D., Ronald G. Collman, M.D., Gwendolyn Binder-Scholl, Ph.D., Gabriela Plesa, M.D., Ph.D., Wei-Ting Hwang, Ph.D., Bruce L. Levine, Ph.D., and Carl H. June, M.D.



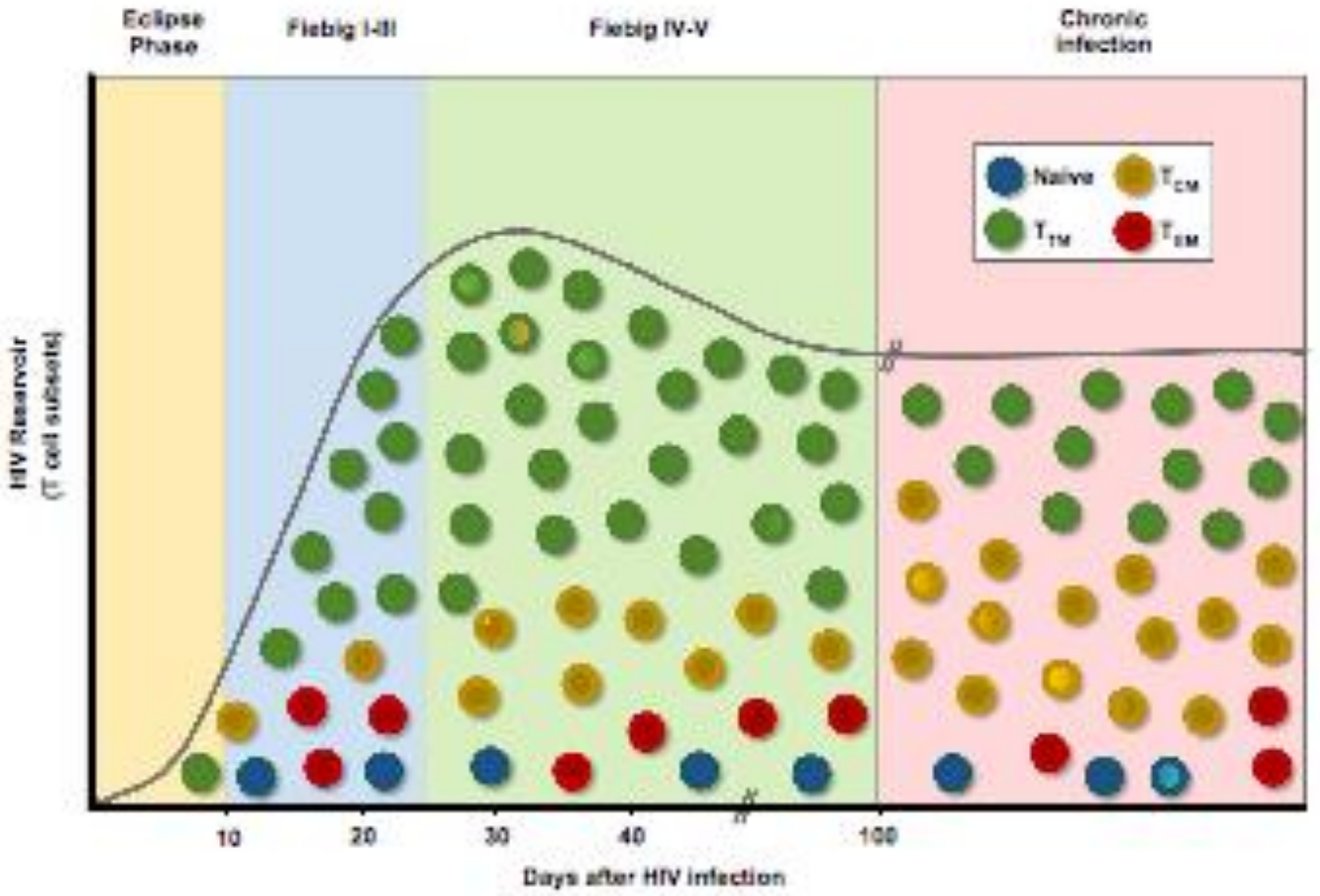
CCR5-modified T cells





Tebas P, et al , NEJM 2014

Very early ART limit the seeding of the HIV reservoir in long-lived TCM



Passaes CP, Virology 2014

The first case of " functional cure" in an infant

The NEW ENGLAND JOURNAL of MEDICINE

BRIEF REPORT

Absence of Detectable HIV-1 Viremia after Treatment Cessation in an Infant

Deborah Persaud, M.D., Hannah Gay, M.D., Carrie Ziemniak, M.S., Ya Hui Chen, B.A., Michael Piatak, Jr., Ph.D., Tae-Wook Chun, Ph.D., Matthew Strain, M.D., Ph.D., Douglas Richman, M.D., and Katherine Luzuriaga, M.D.

What do you think about the baby?

Table 1. Laboratory Testing and Antiretroviral Therapy Received by Mother and Child.*

Test	Result	Antiretroviral Therapy
Mother		
Rapid HIV antibody, at delivery	Positive	None
HIV ELISA and confirmatory Western blot, at 24 hr	Positive	None
Viral load, at 24 hr	2423 copies/ml	None
CD4+ T-cell count, at 14 days	644 cells/mm^3	None
HIV-1 genotype and subtype, at 14 days	Wild-type, subtype B	None
CD4+ T-cell count, at 26 mo	513 cells/mm^3	None
HIV-1 viral load, at 26 mo	6763 copies/ml	None
HLA typing, at 26 mo	A3, A23, B7, B14, Cw7, and Cw8	None
Mutation status in *CCR5* delta32, at 26 mo	Nonmutated	None
Frequency of infected cells, at 28 mo	137 IUPM	None
Child		
HIV-1 DNA, at 30 hr	Positive	Zidovudine
HIV-1 RNA, at 31 hr	19,812 copies/ml	Zidovudine, lamivudine, and nevirapine
HIV-1 RNA, at 6 days	2617 copies/ml	Zidovudine, lamivudine, and nevirapine
HIV-1 RNA, at 11 days	516 copies/ml	Zidovudine, lamivudine, and ritonavir-boosted lopinavir
HIV-1 RNA, at 19 days	265 copies/ml	Zidovudine, lamivudine, and ritonavir-boosted lopinavir
HIV-1 RNA, at 29 days	<48 copies/ml	Zidovudine, lamivudine, and ritonavir-boosted lopinavir
CD4+ T-cell percentage, at 8 days	69%	Zidovudine, lamivudine, and ritonavir-boosted lopinavir
HLA typing, at 26 mo	A3, A68, B7, B39, and Cw7	None
Mutation status in *CCR5* delta32, at 26 mo	Nonmutated	None

All specified time points are post partum. *CCR5* denotes chemokine receptor 5 gene, ELISA enzyme-linked immunosorbent assay, HIV human immunodeficiency virus, HIV-1 HIV type 1, and IUPM infectious units per 1 million resting CD4+ T cells.

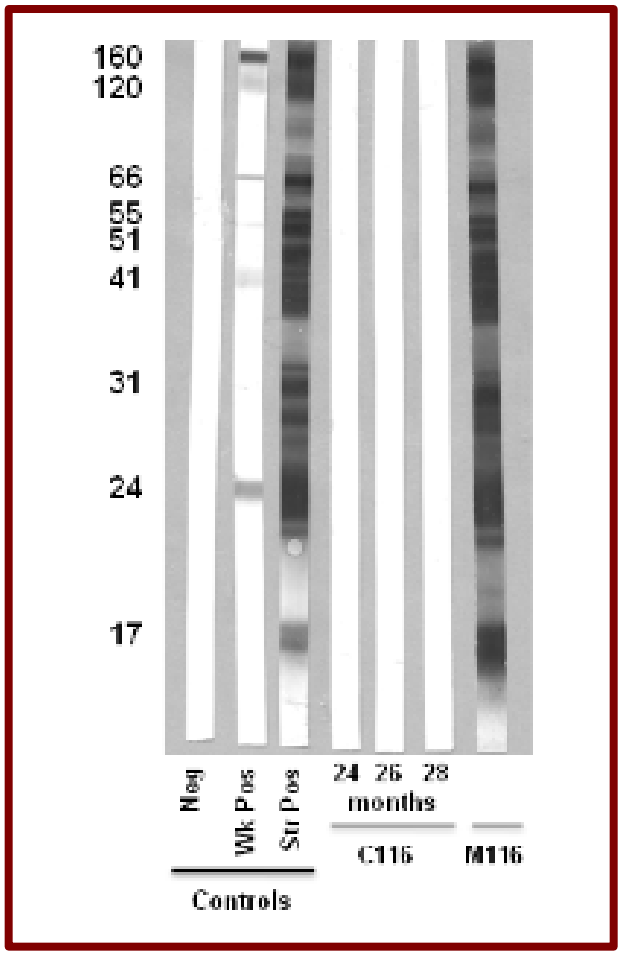
Table 2. Specialized Studies to Assess Persistence of HIV-1 Infection in the Child.*

Sample Type	Quantity	Cells Tested	
		no. of cells or plasma volume	*no. of positive replicates/ no. of wells*
Total proviral DNA			
PBMCs			
At 24 mo	<2.7 copies/10^6 cells	122,000	0/2
At 26 mo	4.2 copies/10^6 cells†	113,000	1/6
Resting CD4+ T cells			
At 24 mo	<3.5 copies/10^6 cells	96,500	0/3
At 26 mo	<2.5 copies/10^6 cells	134,000	0/6
PBMCs enriched for activated CD4+ T cells			
At 24 mo	<2.2 copies/10^6 cells	154,000	0/6
At 26 mo	<2.6 copies/10^6 cells	130,000	0/6
Monocyte-derived adherent cells			
At 24 mo	37.6 copies/10^6 cells‡	14,300	1/3
At 26 mo	<11.5 copies/10^6 cells	29,000	0/6
Residual viremia in plasma			
At 24 mo	1 copy/ml	4 ml	3/3
At 26 mo	<2 copies/ml	4 ml	0/4
Infectious virus recovery at 24 mo	<0.05 IUPM§	22 ×10^6 resting CD4+ T cells	0/22

* PBMCs denotes peripheral-blood mononuclear cells.
† The limit of detection was 2.9 copies per 10^6 cells.
‡ The limit of detection was 23.3 copies per 10^6 cells.
§ No replication-competent HIV-1 was recovered.

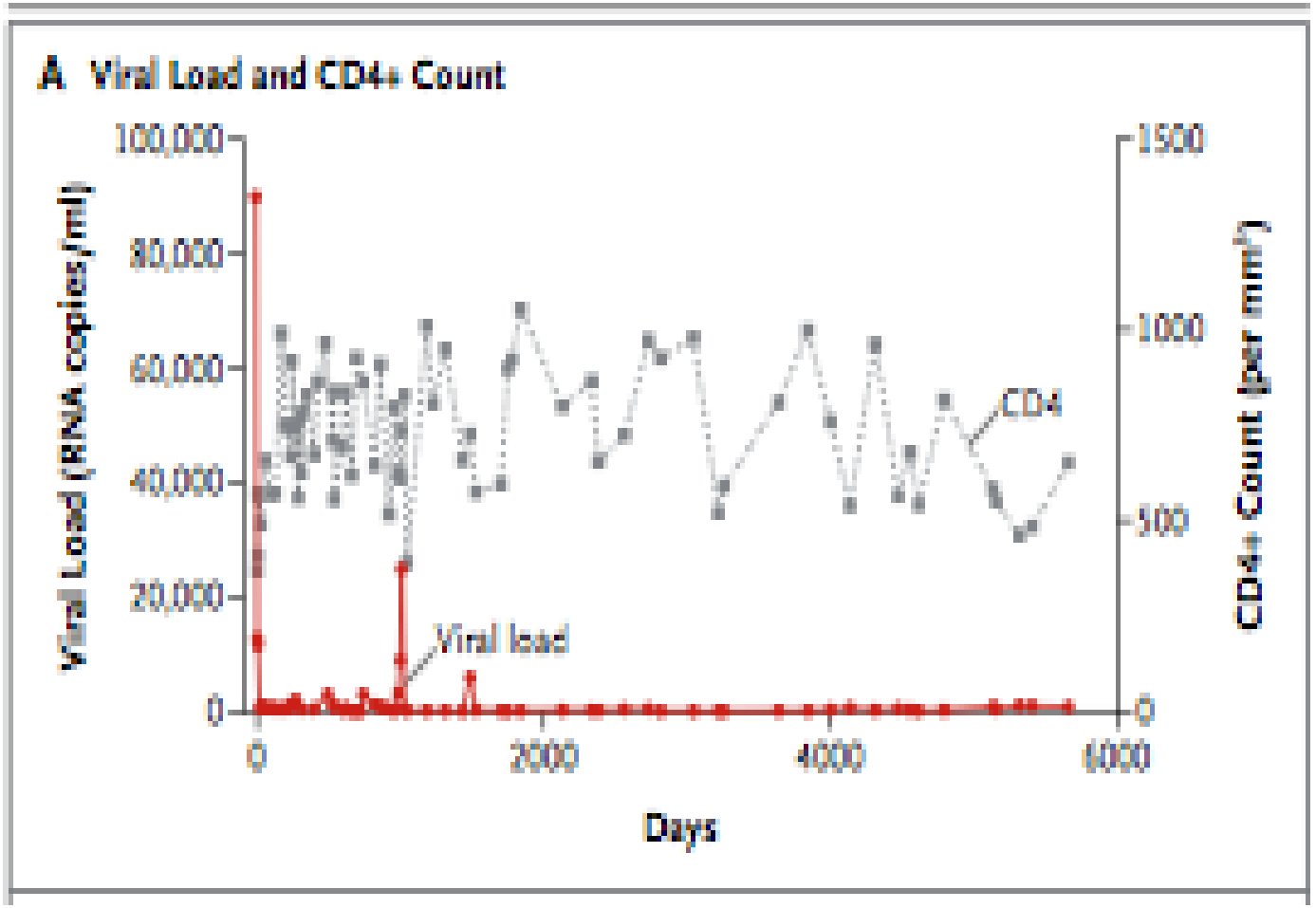
Persaud D, NEJM 2013

C116: HIV antibodies are not detected by WB



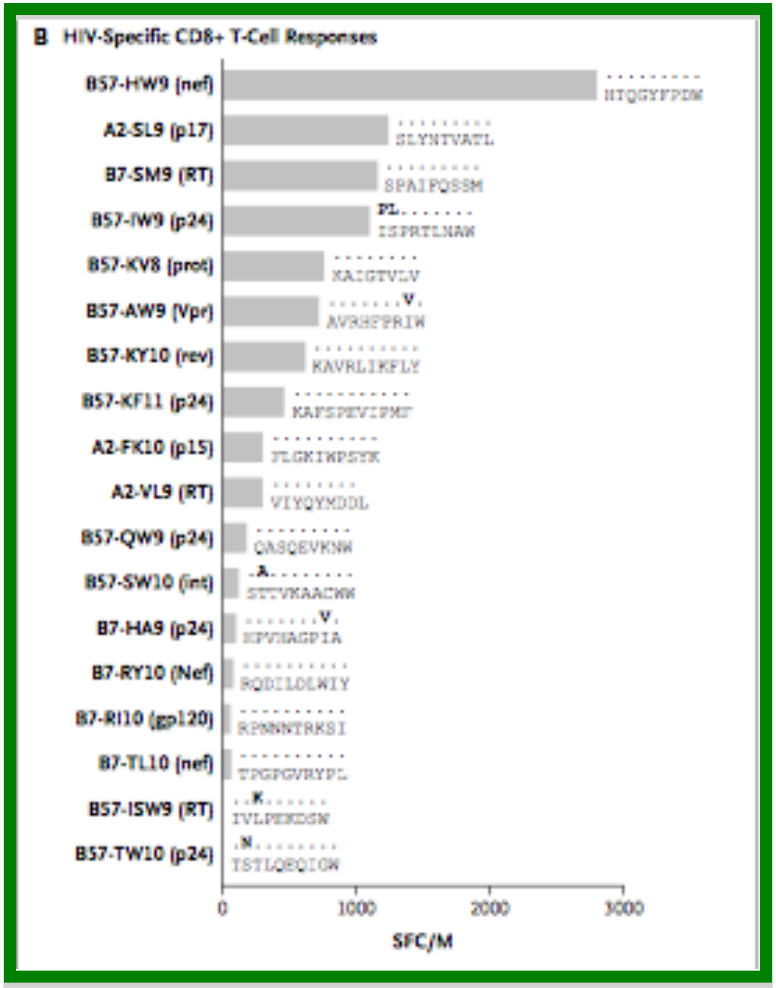
Persaud D, NEJM 2013

How a Single Patient Influenced HIV Research — 15-Year Follow-up



Jessen H, NEJM 2014

The genetic background…



He carried the highly protective HLA –B*57

Jessen H, NEJM 2014

Long-Term Reduction in Peripheral Blood HIV Type 1 Reservoirs Following Reduced-Intensity Conditioning Allogeneic Stem Cell Transplantation

Timothy J. Henrich,[1,2] Zixin Hu,[1,2] Jonathan Z. Li,[1,2] Gaia Scliaranghella,[3,a] Michael P. Busch,[6,7] Sheila M. Keating,[6,7] Sebastien Gallien,[1,8] Nina H. Lin,[2,4] Francoise F. Giguel,[4] Laura Lavoie,[4] Vincent T. Ho,[2,5] Philippe Armand,[2,5] Robert J. Soiffer,[2,5] Manish Sagar,[1,2,a] Ann S. LaCasce,[2,5] and Daniel R. Kuritzkes[1,2]

[1]Divison of Infectious Diseases, Brigham and Women's Hospital, [2]Harvard Medical School, [3]Ragon Institute of MGH, MIT, Harvard, [4]Division of Infectious Diseases, Massachusetts General Hospital, and [5]Department of Medical Oncology, Dana-Farber Cancer Institute, Boston, Massachusetts; [6]Blood Systems Research Institute, and [7]University of California–San Francisco, San Francisco; and [8]Hopital Saint-Louis, Paris, France

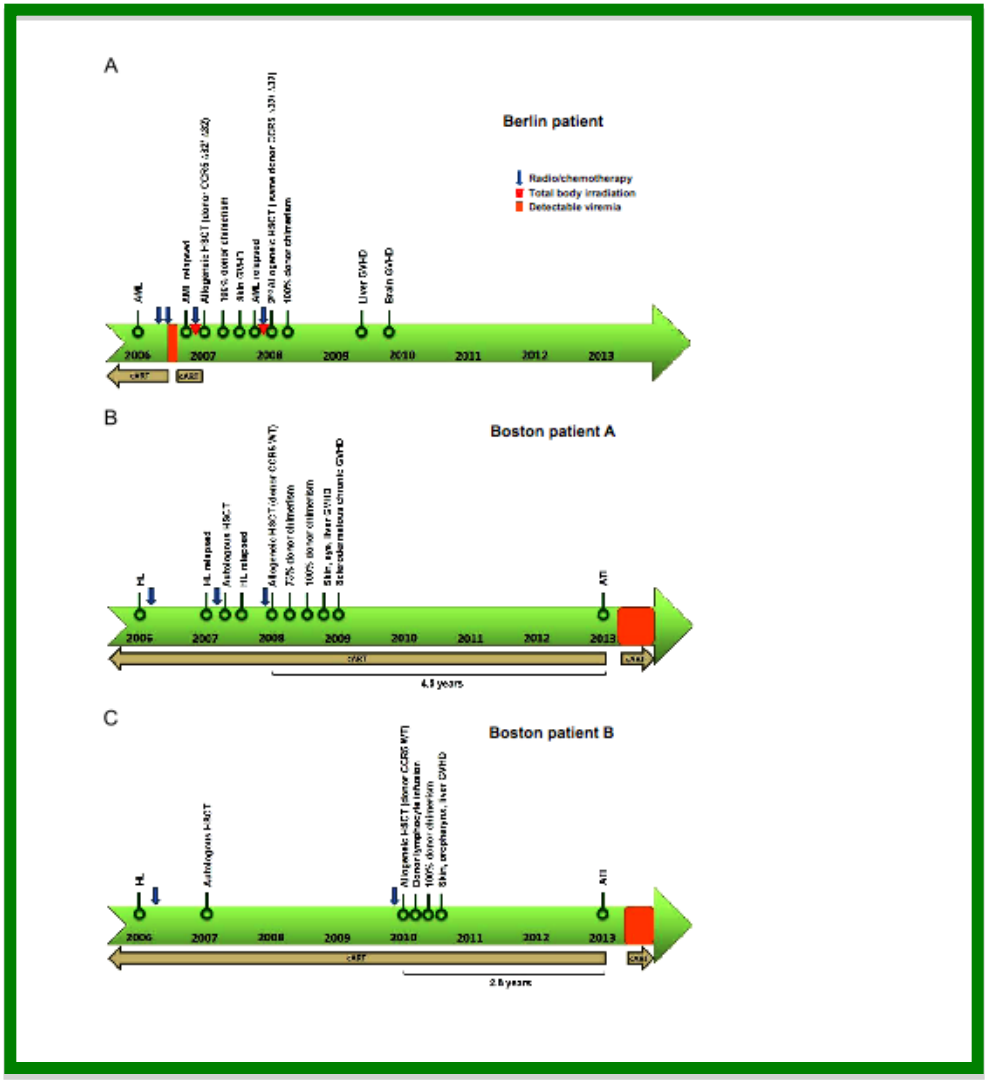
Background. The long-term impact of allogeneic hematopoietic stem cell transplantation (HSCT) on human immunodeficiency virus type 1 (HIV-1) reservoirs in patients receiving combination antiretroviral therapy (cART) is largely unknown.

Methods. We studied the effects of a reduced-intensity conditioning allogeneic HSCT from donors with wild-type–CCR5$^+$ cells on HIV-1 peripheral blood reservoirs in 2 patients heterozygous for the *ccr5Δ32* mutation. In-depth analyses of the HIV-1 reservoir size in peripheral blood, coreceptor use, and specific antibody responses were performed on samples obtained before and up to 3.5 years after HSCT receipt.

Results. Although HIV-1 DNA was readily detected in peripheral blood mononuclear cells (PBMCs) before and 2–3 months after HSCT receipt, HIV-1 DNA and RNA were undetectable in PBMCs, CD4$^+$ T cells, or plasma up to 21 and 42 months after HSCT. The loss of detectable HIV-1 correlated temporally with full donor chimerism, development of graft-versus-host disease, and decreases in HIV-specific antibody levels.

Conclusions. The ability of donor cells to engraft without evidence of ongoing HIV-1 infection suggests that HIV-1 replication may be fully suppressed during cART and does not contribute to maintenance of viral reservoirs in peripheral blood in our patients. HSCTs with wild-type–CCR5$^+$ donor cells can lead to a sustained reduction in the size of the peripheral reservoir of HIV-1.

The first case of “steriling cure”



Passaes CP, Virology 2014

PLOS | PATHOGENS

Challenges in Detecting HIV Persistence during Potentially Curative Interventions: A Study of the Berlin Patient

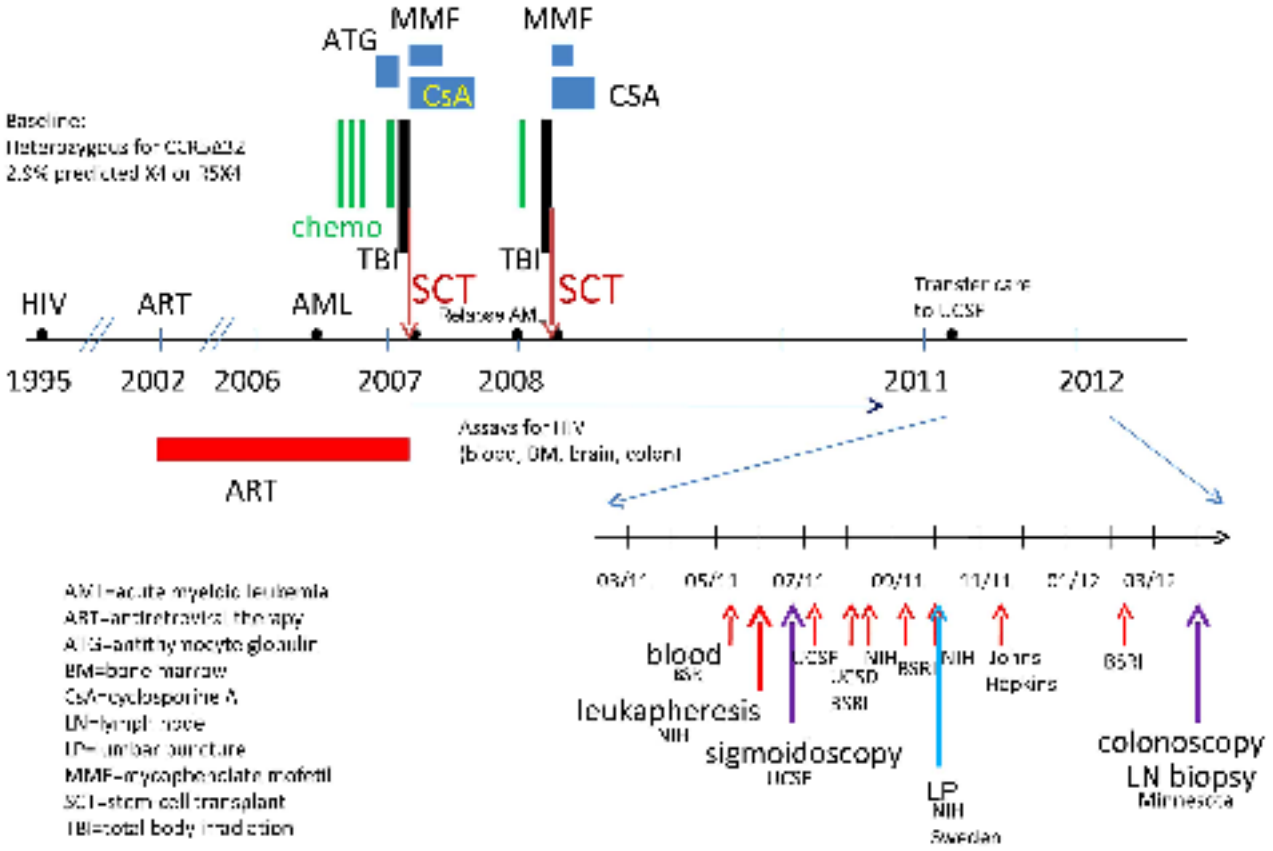


Figure 1. Timeline for clinical treatments and study samples.
doi:10.1371/journal.ppat.1003347.g001

QUANTIFYING HIV PERSISTENCE

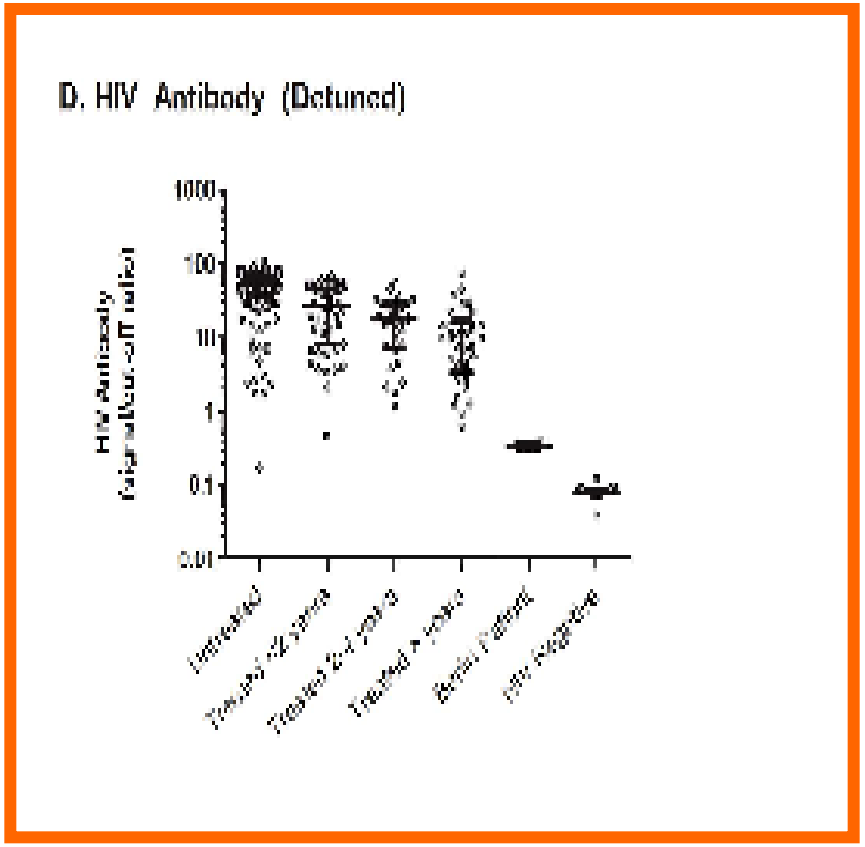
Table 3. HIV in tissues.

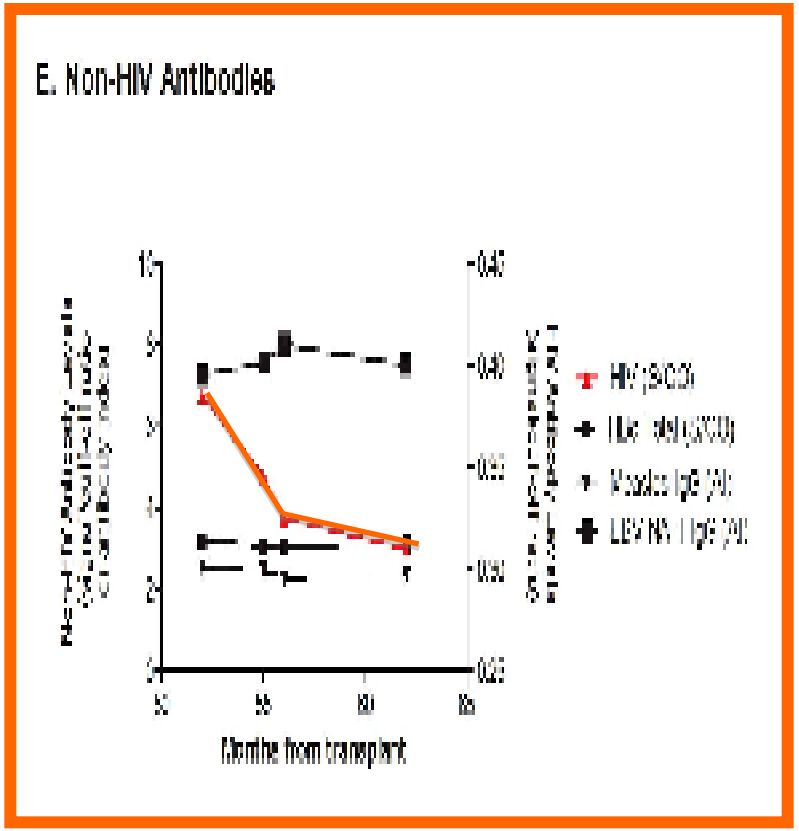
Sample	Date	Lab	Measure	Input	Assay	Detection Limit	False Positive Rate[1]	Result
CSF cells	10/3/11	NIH	HIV DNA	8,400 cells	qPCR [19] for LTR	0.5–2.6 copy/μg	0/72 (<1.4%)	ND[2]
CSF	10/3/11	Sweden	HIV RNA	7 ml	SCA [7]	0.3 copy/ml [7]		ND (≤0.1 copy/ml)
CSF	10/3/11	NIH	HIV RNA	10 ml	Roche Ampliprep [19]	1–2 copy/ml	1/179 (0.6%)	ND (≤0.1 copy/ml)
Lymph Node	3/27/12	Vaccine Research Center	HIV DNA	No cell counts available	Fluorescence- assisted clonal amplification of Env	1 copy/reaction		ND
Lymph Node	3/27/12	Vaccine Research Center	HIV RNA	No cell counts available	RNA sequencing	1 read/sample		ND
Lymph Node	3/27/12	Univ. of Minnesota	HIV RNA		ISH [13]	10^{3-4} cells/g		ND
Rectal biopsy	6/20/11	SFVA/UCSF	HIV DNA	DNA from 2.7×10^6 cells (from 3×2 biopsies)	qPCR for LTR [24,56]	1–2 copy/reaction [56]	1/130 (0.8%)	2 of 15 wells positive1 in 10^6 cells
Rectal cells (collagenase digestion)	6/20/11	SFVA/UCSF	HIV DNA	DNA from 1.5×10^6 cells (out of $2\times2.4\times10^6$)	qPCR for LTR [24,56]	1–2 copy/reaction [56]	1/130 (0.8%)	1 of 10 wells positive1 in 10^6 cells
4 sorted cell populations from rectum	3/27/12	Vaccine Research Center	HIV DNA	3.1×10^5 $CD45^-$ cells	Fluorescence-enabled clonal amplification of Env	1 copy/reaction		ND
				3.1×10^4 $CD45^+$ non-T cells				ND
				1.3×10^4 $CD4^+$ T cells				ND
				3.7×10^4 other T cells				ND
4 sorted cell populations from rectum	3/27/12	Vaccine Research Center	HIV RNA	8.5×10^5 $CD45^-$ cells	RNA sequencing	1 read/sample		ND
				3.2×10^4 $CD45^+$ non-T cells				ND
				1.3×10^4 $CD4^+$ T cells				ND
				3.8×10^4 other T cells				ND
Rectal biopsy	6/20/11	SFVA/UCSF	HIV RNA	RNA from 7.8×10^6 cells (from 3×2 biopsies)	qRT-PCR for LTR [24,56]	1–2 copy/reaction [56]	0/83 (<1.2%)	ND (≤1 in 7.8×10^6 cells)
Rectal cells (collagenase digestion)	6/20/11	SFVA/UCSF	HIV RNA	RNA from 2.5×10^6 cells (out of $2\times2.4\times10^6$)	qRT-PCR for LTR [24,56]	1–2 copy/reaction [56]	0/83 (<1.2%)	ND (≤1 in 2.5×10^6 cells)
Rectal Biopsy	3/27/12	Univ. of Minnesota	HIV RNA		ISH [13]	10^{3-4} cells/g		Rare signal, Partifact
4 sorted cell populations from ileum	3/27/12	Vaccine Research Center	HIV DNA	8.6×10^5 $CD45^-$ cells	Fluorescence- enabled clonal amplification of Env	1 copy/reaction		ND (≤1 in 10^6 cells)
				4.9×10^4 $CD45^+$ non-T cells				ND
				2.9×10^4 $CD4^+$ T cells				ND
				1.0×10^5 other T cells				ND
4 sorted cell populations from ileum	3/27/12	Vaccine Research Center	HIV RNA	8.8×10^5 $CD45^-$ cells	RNA sequencing	1 read/sample		ND
				5.0×10^4 $CD45^+$ non-T cells				ND
				3.0×10^4 $CD4^+$ T cells				ND
				1.1×10^5 other T cells				ND
Ileal Biopsy	3/27/12	Univ. of Minnesota	HIV RNA		ISH [13]	10^{3-4} cells/g		ND

[1]Historical, as determined from the number of positive wells out of all wells containing samples from HIV- subjects/donors that had been processed and run along with positive samples.
[2]ND = not detected.

Yukl AS, PLOS Pathogens 2014

The Berlin patient





Yukl SA et al, PLOS Pahogens 2014

“ Ogni grande viaggio inizia con un singolo passo”

Lao-tzu