Efforts and advances for the Cure

Pablo Tebas, MD

Penn Medicine

Disclosures

- Consulting for Merck, Viiv and Gilead
- Write for uptodate

Lets start with definitions
The "cure" continuum

Long acting ART	Sustained Remission	Sterilizing Cure
(Control of viremia with long acting drugs)	(Control of viremia without drugs)	(No detectable infectious virus)
Absence of rebound with intermittent therapy	Absence of rebound upon cessation of ARVs	Reduction in the size of the latent, replication-competent reservoir

The only HIV sterilizing cure


BRIEF REPORT

Long-Term Control of HIV by *CCR5* Delta32/Delta32 Stem-Cell Transplantation

Gero Hütter, M.D., Daniel Nowak, M.D., Maximilian Mossner, B.S., Susanne Ganepola, M.D., Arne Mußig, M.D., Kristina Allers, Ph.D., Thomas Schneider, M.D., Ph.D., Jörg Hofmann, Ph.D., Claudia Kücherer, M.D., Olga Blau, M.D., Igor W. Blau, M.D., Wolf K. Hofmann, M.D., and Eckhard Thiel, M.D.


The New York Times Health

WORLD | U.S. | N.Y. / REGION | BUSINESS | TECHNOLOGY | SCIENCE | HEALTH

Search Health

New Hope of a Cure for H.I.V.

VIRUS-FREE Timothy Brown of San Francisco had two bone-marrow transplants to treat leukemia, and H.I.V. can no longer be detected in his body.

By ANDREW POLLACK

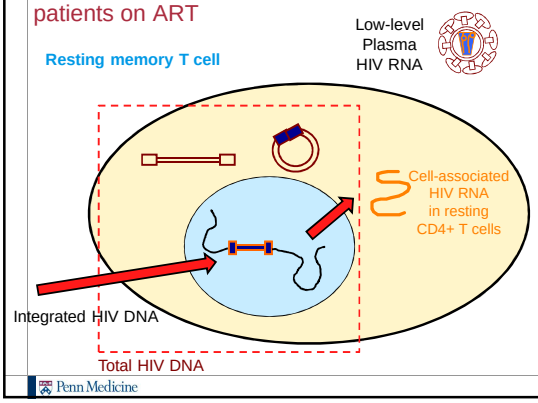
Published: November 29, 2011

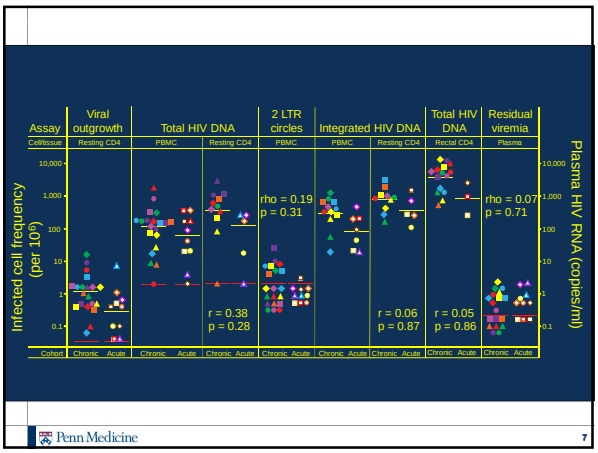
NEJM 2009; 360: 692

Barriers for HIV cure

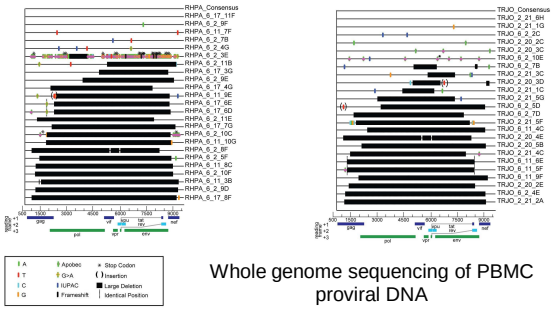
The HIV reservoir:
One in a million memory CD4 T cell has a replication competent virus

Measures of persistent HIV in fully suppressed patients on ART





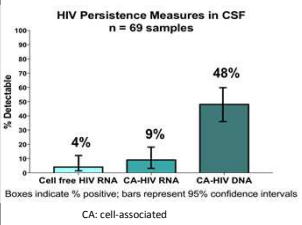
Not all proviruses are replication competent. Defective integrated virus



Whole genome sequencing of PBMC proviral DNA

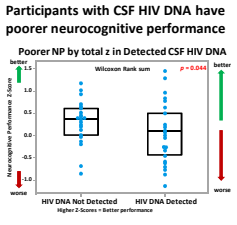
What Is New About The Reservoir?

HIV DNA Persists in CSF in Half of Those on Long-term ART



Boxes indicate % positive; bars represent 95% confidence intervals

CA: cell-associated

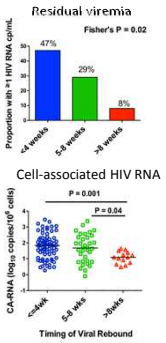


NP: neurocognitive performance

Spudich S et al, CROI 2018, #119
Robertson K et al, CROI 2018, #403LB

Biomarkers to predict HIV rebound

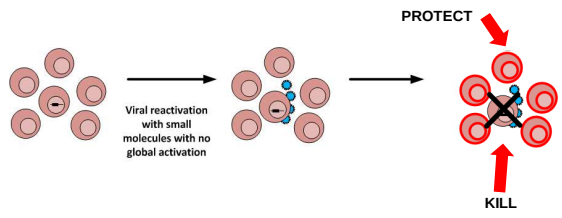
- Time to virus rebound after stopping ART may reflect size of HIV reservoir[1]
- In retrospective analysis of participants in ACTG treatment interruption studies, lower pre-TI residual viremia and cell-associated HIV RNA were associated with longer time to HIV rebound[2]
- Need to find virologic and immunologic markers that predict longer time to HIV rebound in an intensively monitored antiretroviral pause (IMAP)



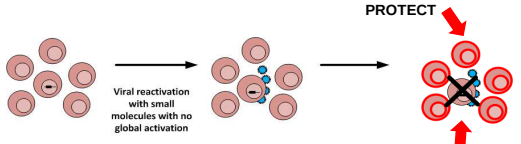
[1]Hill A et al, PNAS, 2014; [2]Li J, et al, AIDS, 2016

Current ideas about curing HIV

1. KICK
2. KILL
3. PROTECT



STEP 1. KICKING the reservoir



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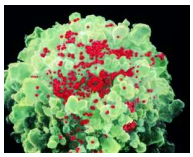
Activating the reservoir. SAHA. Proof concept



- The purpose is to make the infected cell more visible to the immune system

Archin et al, Nature 2012

LETTER

Administration of vorinostat disrupts HIV-1 latency in patients on antiretroviral therapy

RESEARCH LETTER

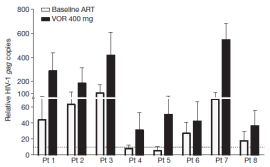


Figure 3 | VOR upregulates HIV RNA expression. The relative HIV-1 RNA copy number (mean +1 s.d.) measured in the resting $CD4^+$ T cells of eight HIV-positive patients with plasma HIV RNA BDL is shown on background ART and on ART following a single 400 mg oral dose of VOR. For each subject, the differences are significant ($P < 0.01$).

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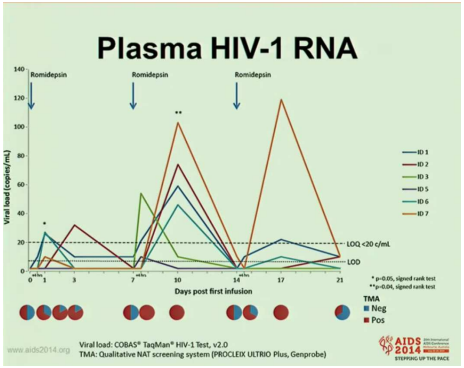
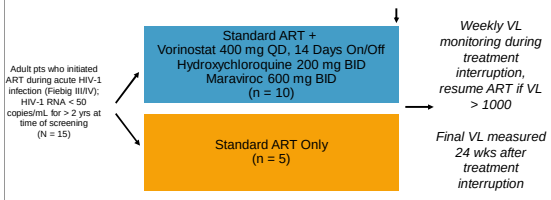
Romidepsin 5 mg/m2 (at 0,7,14 days)



Penn Medicine Ole Schmeltz Søgaard et al. TUAA0106LB 15

SEARCH 019: VHM Combination Therapy in Pts Treated During Acute HIV Infection

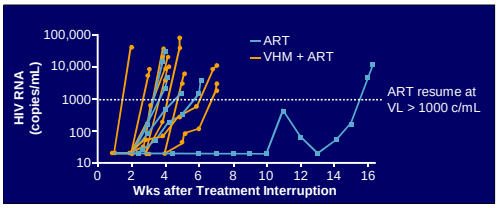
- **Prospective, randomized, open-label phase I/II trial**
  - Primary objective: frequency of pts in each arm who maintain virologic suppression (HIV-1 RNA < 50 copies/mL) at 24 wks post treatment interruption



Kroon E, et al. AIDS 2016. Abstract TUAX0101LB.

SEARCH 019: Efficacy and Safety

- **Closely monitored treatment interruption safe**
- **No change in total HIV DNA in PBMCs after treatment period and no difference in time to VL rebound after treatment interruption**
  - Median time to VL detection: 22 days (range: 14-77 days)
  - VHM-induced, low-level plasma viremia detected in some pts (*P* = .0078)

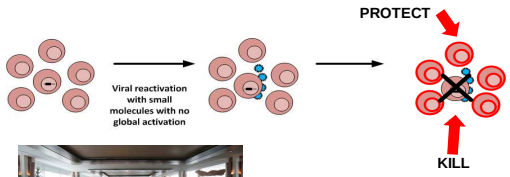


Kroon E, et al. AIDS 2016. Abstract TUAX0101LB.

What we have learned so far about kicking the reservoir?

- **Using SAHA (vorinostat), panobinostat and romidepsin activates HIV from latency**
- **Increases HIV RNA expression intracellularly**
- **Can be associated with transient HIV viremia**
- **Subsequent treatments are less effective**
- **It really does not decrease the reservoir**
- **If activating alone does not eliminate the reservoir, do you really need to do this? Or stopping therapy is enough?**

STEP 2. KILLING (eliminating) the reservoir



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Eliminating the reservoir

- **Unspecific (kill everybody)**
  - Graft vs host disease (like Timothy Brown or the Boston patients)
  - ATG effects on reservoir in HIV+ renal transplant patients (Deirdre Sawinski)
- **Specific (kill the reservoir)**
  - Boost Cellular immunity
    - Improving CD8 HIV responses (T cell vaccines)
    - Blocking the PD-1 PD-L1 pathway
    - Gene therapy (reprogrammed CD8 cells and CARs)
  - Improving Innate immunity
  - Humoral immunity
    - Using Neutralizing antibodies

Unspecific killing. The Boston cases


The New York Times
Health
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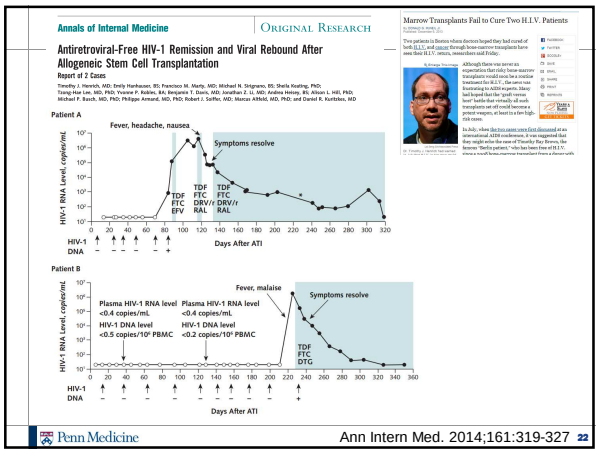
After Marrow Transplants, 2 More Patients Appear H.I.V.-Free Without Drugs

Table 1. Studies to Assess HIV-1 Reservoirs After Allogeneic HSCT and Before ART Interruption

Sample Type	Level	Total Cells Tested, *n*	Positive Wells/ Total Wells, *n/N*
Patient A (4.3 y after HSCT)			
Total HIV-1 PBMC DNA	<0.12 copies/10^6 cells	26×10^6 PBMCs	0/42
Infectious virus by viral outgrowth assay	<0.007 IUPM cells*	150×10^6 $CD4^+$ T cells	0/30
Patient B (2.6 y after HSCT)			
Total HIV-1 PBMC DNA	<0.13 copies/10^6 cells	24×10^6 PBMCs	0/42
Infectious virus by viral outgrowth assay	<0.006 IUPM cells*	160×10^6 $CD4^+$ T cells	0/32
Rectal tissue	<2.4 copies/10^6 cells	1.23×10^6 nucleated cells†	0/22

ART = antiretroviral therapy; HSCT = hematopoietic stem cell transplantation; IUPM = infectious units per million; PBMC = peripheral blood mononuclear cell.
* No replication-competent HIV-1 was recovered.
† 25% of $CD4^+$ T cells were tested using flow cytometry after tissue disaggregation.



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Ann Intern Med. 2014;161:319-327 22

EpiStem: Allogeneic Stem Cell Transplantation in HIV-1 Infected Pts

- **EpiStem Consortium: prospective, observational, cohort**
  - Eligibility: HIV-infected pts requiring allogeneic stem cell transplantation for a life-threatening hematologic condition
- **Overall goal: understand the biological determinants behind HIV-1 reservoir reduction/eradication by allogeneic stem cell transplantation**
- **Thus far, 24 HIV+ pts with diverse hematologic malignancies have been registered,15 have received stem cell transplantation and remain on ART**

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Wensing AM, et al. AIDS 2016. Abstract THAA0105. 23

EpiStem: ASCT Reduces HIV-1 Reservoirs Independent of Donor *CCR5* Status

- **Preliminary results for 3 pts showed that ASCT reduces HIV-1 reservoirs to low levels independent of procedure, donor genotype**
  - Investigators hypothesize viral clearance can be attributed to graft vs HIV-1 reservoir effect

Outcome	Pt 1	Pt 3	Pt 19
Hematologic malignancy	Burkitt NHL	NK NHL	AML
Conditioning strategy	Myoablative	Reduced intensity	Reduced intensity
Donor type	HLA-mismatched	HLA-MRD (10/10)	HLA-MUD (10/10)
Donor CCR5 status	WT	WT	Δ32
Pt chimera status	0.2% BM/0.1% PB	Full	Full
GvHD	No	Yes	Yes
HIV-1 RNA, copies/mL	5	Undetectable	--
Total HIV-1 DNA, copies/10^6 CD4+	25	Undetectable	Undetectable
qVOA, IUPM	0.034	Undetectable	Undetectable
Ileum (CD4+ cells)	--	Undetectable	Trace

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Wensing AM, et al. AIDS 2016. Abstract THAA0105. 24

Outcomes of BMT with delta 32 donors

Table 1. Men with Human Immunodeficiency Virus Type 1 (HIV-1) Infection Who Received an Allogeneic Transplant from a Stem-Cell Donor Who Was Homozygous for the *CCR5* delta32/delta32 Mutation.*

Location of Transplantation	Age of Patient	Type of Cancer	Type of Graft	Outcome after Transplantation
	yr			
Berlin†	40	Acute myeloid leukemia	HLA-matched unrelated	Alive after 7 yr, no viral rebound, no ART
Utrecht, the Netherlands‡	53	Myelodysplastic syndrome	Combined haploidentical bridge with umbilical-cord blood	Died from relapse of the myelodysplastic syndrome and pneumonia after 2 mo
Münster, Germany§	51	Non-Hodgkin's lymphoma	HLA-mismatched unrelated	Died from infection after 4 mo
Essen, Germany¶	30	Non-Hodgkin's lymphoma	HLA-matched unrelated	CXCR4-tropic HIV-1 rebound, died from relapse of non-Hodgkin's lymphoma after 12 mo
Minneapolis§	12	Acute lymphoblastic leukemia	Umbilical-cord blood	Died from GVHD after 3 mo
Santiago, Chile§	46	Non-Hodgkin's lymphoma	HLA-matched related	Died from pneumonia shortly afterward
Barcelona§	37	Non-Hodgkin's lymphoma	Combined haploidentical bridge with umbilical-cord blood	Died from relapse of non-Hodgkin's lymphoma after 3 mo

* ART denotes antiretroviral therapy, and GVHD graft-versus-host disease.
† Data are from Hütter et al.[1]
‡ Data are from Kwon et al.[3]
§ Data are from a personal communication with the transplantation center.
¶ Data are from Kordelas et al.[2]

Penn Medicine N Engl J Med 2014; 371:2437-2438December 18, 2014 25

What have we learned from these cases

- It is possible to cure HIV
- Very difficult (The score so far is 1 of 39,000000)
- Aggressive strategies to remove the reservoir (total body irradiation+Chemo+Allo BMT with GVHD) do not completely eliminate it
- Treating very very early neither…
- We may have to settle for functional cure

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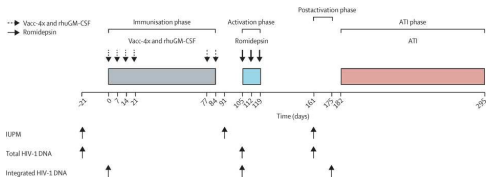
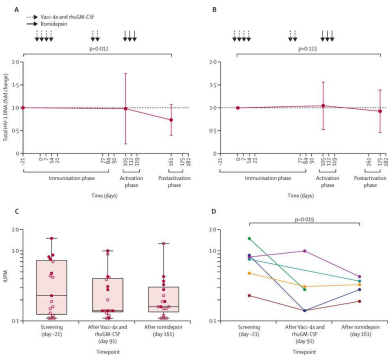
Therapeutic vaccines

Combined effect of Vacc-4x, recombinant human granulocyte macrophage colony-stimulating factor vaccination, and romidepsin on the HIV-1 reservoir (REDUC): a single-arm, phase 1B/2A trial



Penn Medicine Lancet HIV. 2016 Oct;3(10):e463-72 27

Therapeutic vaccine + romidepsin



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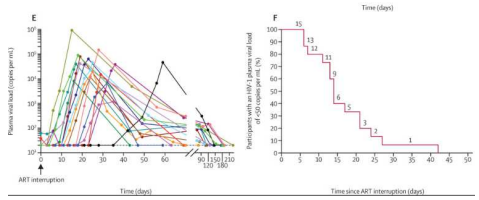
Lancet HIV. 2016 Oct;3(10):e463-72 28

Therapeutic vaccine + romidepsin



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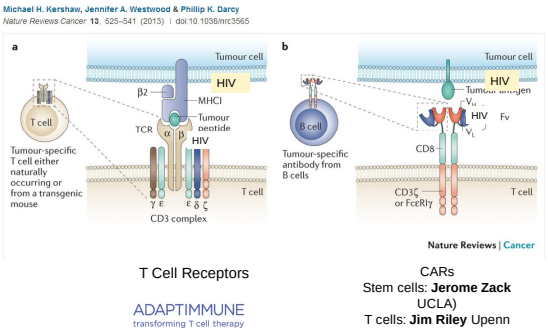
Lancet HIV. 2016 Oct;3(10):e463-72 29

Engineering the "kill"
Engineering T cells that target HIV



T Cell Receptors

ADAPTIMMUNE
transforming T cell therapy

CARs
Stem cells: **Jerome Zack** UCLA)
T cells: **Jim Riley** Upenn

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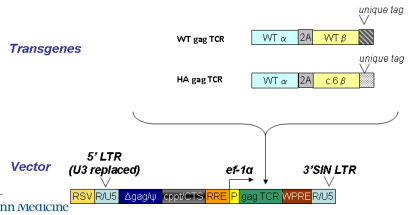
Modified CD8 cells with high affinity TCR Manufacturing

Vector is a SIN HIV derived vector developed at UPenn

Clinical grade vector was manufactured by the City of Hope, Duarte California, led by Larry Couture and David Hsu

T cells are manufactured at the Clinical Cell and Vaccine Production Facility, at UPenn, led by Bruce Levine and Carl June



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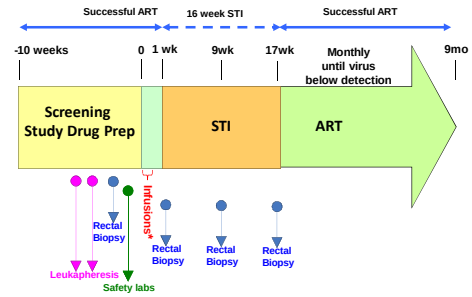
Study Schema



* Infusions split across day 0, 1, and 2 at 10%, 30%, and 60% of total dose, respectively

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Enrollment Status

Patient #	Arm	TcR	Dose	Apheresis (start of mfg)	Td %	Cell Dose	T cell infusion	Safety 1 wk (start of STI)	Safety & HIV status 13 wk (end of STI)
SL9-106	aviremic	wt-gag	low	19th July	21%	1 e8	23rd-25th Aug	Yes	Yes
SL9-118	aviremic	wt-gag	low	25th Oct	65%	1 e8	10-12 of Jan	Yes	Yes

- Patient 1 received 20 million transduced T cells
- Patient 2 received 65 million transduced T cells

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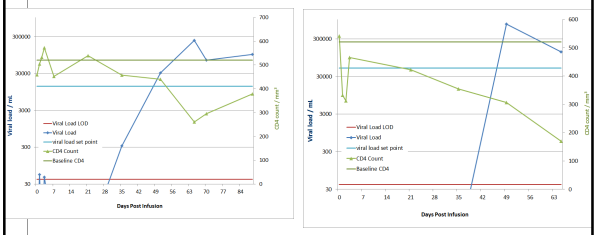
Results



- **Patient 1**: No selection of cells seen in first patient upon virus return; pre and post rectal biopsy not available
- **Patient 2**: cell persistence data pending; pre and post rectal biopsy performed
- Important to note-no CD4 cushion provided by the infusion product

Plenary Paper

blood

CLINICAL TRIALS AND OBSERVATIONS

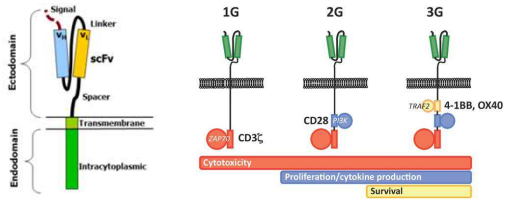
Cardiovascular toxicity and titin cross-reactivity of affinity-enhanced T cells in myeloma and melanoma

Gerald P. Linette,[1] Edward A. Stadtmauer,[2] Marcela V. Maus,[2] Aaron P. Rapoport,[3] Bruce L. Levine,[2] Lyndsey Emery,[2] Leslie Litzky,[2] Adam Bagg,[2] Beatriz M. Carreno,[1] Patrick J. Cimino,[1] Gwendolyn K. Binder-Scholl,[4] Dominic P. Smethurst,[4] Andrew B. Gerry,[4] Nick J. Pumphrey,[4] Alan D. Bennett,[4] Joanna E. Brewer,[4] Joseph Dukes,[5] Jane Harper,[5] Helen K. Tayton-Martin,[4] Bent K. Jakobsen,[4,5] Namir J. Hassan,[6] Michael Kalos,[2] and Carl H. June[2]

[1]Siteman Cancer Center and Departments of Medicine and Pathology and Immunology, Washington University School of Medicine, St. Louis, MO; [2]Abramson Cancer Center, Department of Medicine, and Department of Pathology and Laboratory Medicine, University of Pennsylvania, Philadelphia, PA; [3]The Greenebaum Cancer Center, University of Maryland, Baltimore, MD; [4]Adaptimmune Ltd, Philadelphia and Abingdon, United Kingdom; and [5]Immunocore Ltd, Abingdon, United Kingdom

- Clinical testing of engineered T cells expressing an affinity-enhanced TCR against HLA-A*01–restricted MAGE-A3.
- Clinical trials in patients with melanoma and myeloma
- First two patients developed cardiogenic shock and died.
- Autopsy revealed severe myocardial damage, and histopathological analysis revealed T-cell infiltration
- No MAGE-A3 expression was detected in heart autopsy tissues

Chimeric antigen receptors



Plenary paper

Prolonged survival and tissue trafficking following adoptive transfer of CD4ζ gene-modified autologous CD4+ and CD8+ T cells in human immunodeficiency virus–infected subjects

Ronald T. Mitsuyasu, Peter A. Anton, Steven G. Deeks, David T. Scadden, Elizabeth Connick, Matthew T. Downs, Andreas Bakker, Margo R. Roberts, Carl H. June, Sayeh Jalali, Andy A. Lin, Rukmini Pennathur-Das, and Kristen M. Hege

BLOOD, 1 AUGUST 2000 • VOLUME 96, NUMBER 3

TRIAL

doi:10.1006/mthe.2002.0611, available online at http://www.idealibrary.com on IDEAL

A Phase II Randomized Study of HIV-Specific T-Cell Gene Therapy in Subjects with Undetectable Plasma Viremia on Combination Antiretroviral Therapy

Steven G. Deeks,[1] Bridget Wagner,[2] Peter A. Anton,[3] Ronald T. Mitsuyasu,[3] David T. Scadden,[4] Christine Huang,[5] Catherine Macken,[6] Douglas D. Richman,[7] Cindy Christopherson,[8] Carl H. June,[9] Richard Lazar,[10] David F. Broad,[10] Sayeh Jalali,[10] and Kristen M. Hege[10,*]

MOLECULAR THERAPY Vol. 5, No. 6, June 2002

RESEARCH ARTICLE

ADOPTIVE T CELL TRANSFER

Decade-Long Safety and Function of Retroviral-Modified Chimeric Antigen Receptor T Cells

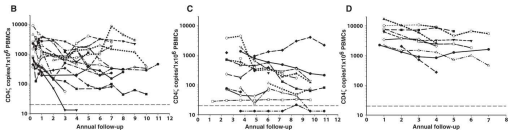
John Scholler,[1]* Troy L. Brady,[2]* Gwendolyn Binder-Scholl,[1] Wei-Ting Hwang,[3] Gabriela Plesa,[1] Kristen M. Hege,[4] Ashley N. Vogel,[1] Michael Kalos,[1] James L. Riley,[2] Steven G. Deeks,[5] Ronald T. Mitsuyasu,[6] Wendy B. Bernstein,[7] Naomi E. Aronson,[7,8] Bruce L. Levine,[1] Frederic D. Bushman,[2†] Carl H. June[1†]

Cell persisted

Persistence of CD4ζ-modified CAR T cells over 11 years after infusion.

A

Annual	1	2	3	4	5	6	7	8	9	10	11	Total
Detected	*20*	*25*	*33*	*31*	*28*	*25*	*24*	*13*	*8*	*4*	*1*	212
Tested	*20*	*25*	*35*	*33*	*29*	*26*	*24*	*15*	*9*	*4*	*1*	221



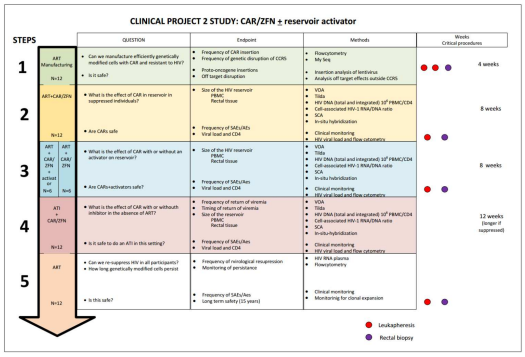
John Scholler et al., Sci Transl Med 2012;4:132ra53

Science Translational Medicine AAAS

Upenn Delaney CAR proposal Objective

- **To evaluate the safety, tolerability and antiviral activity of the combination of**
  - a genetically modified HIV resistant (CCR5 edited by zinc finger nuclease) T cell with a CD4 chimeric antigen receptor
  - with or without an activator of the HIV reservoir in the setting of well controlled HIV infection

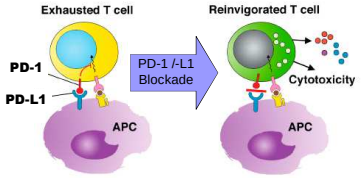
- **Hypothesis**
  - The combination of a genetically engineered CAR T with an activator of the HIV reservoir will be safe, well tolerated and will delay or prevent the return of HIV viremia and may be associated with a decrease of the size of the HIV reservoir.



Rescue the immune response against HIV

Blockade of the PD-1/PD-L1 pathway is a viable therapeutic strategy in chronic HIV

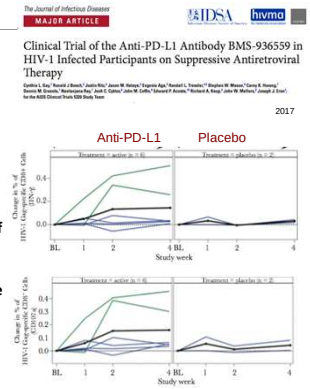
- **Restores HIV-specific immune functions *in vitro* and *ex vivo***
- **Reduces viremia and prolongs survival in animal models**



Adapted from Freeman G. et.al., *J Exp Med* 2006.

ACTG 5326

- ACTG A5326: randomized trial of single infusion of anti-PDL1 Ab
- 6 participants received Ab, 2 received placebo
- Increased HIV-specific CD8+ T cell responses in 2 of 6 who received anti-PD-L1
- No sustained changes in measures of HIV persistence (but small numbers)
- 1 participant developed asymptomatic hypophysitis (immune related AE)



Improving the innate immune system

An interferon trial

Subjects on ART, vl<50 copies/ml, CD4>450 cells/ml, nadir>200 cells/ml

Visit week	0	4	8	10	13	17	21	25	29	33	37	41	45	49
	ART											ART		

Arm A: Peg-IFN-α2A 180 µg/week

Arm B: Peg-IFN-α2A 90 µg/week

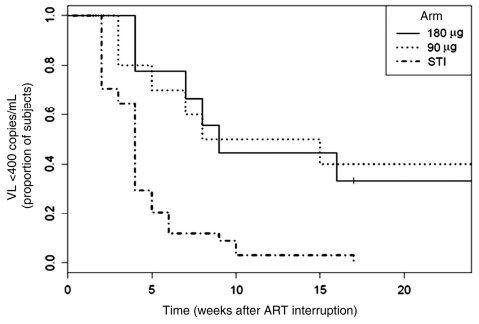
Proportion of subjects with viral load<400 copies/ml at 12 weeks of monotherapy

Penn Medicine

Azzoni L et al. J Infect Dis. 2013;207:213-222

The Journal of Infectious Diseases

Viral load (VL) response to Peg–interferon alfa-2a.

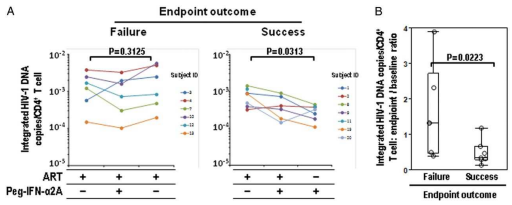


Penn Medicine

Azzoni L et al. J Infect Dis. 2013;207:213-222

The Journal of Infectious Diseases

Decreases of the HIV reservoir Associated with improvement of NK function



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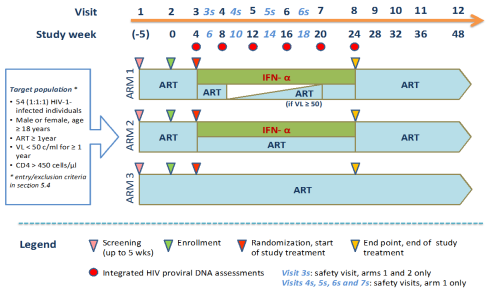
Azzoni L et al. J Infect Dis. 2013;207:213-222

The Journal of Infectious Diseases

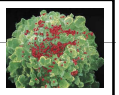
Study design

WISTAR / BEAT HIV Study



The Wistar Institute

Improving the humoral response
Broadly neutralizing antibodies



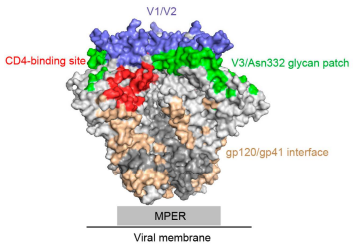
- **Several novel monoclonal antibodies that have been derived from cells from HIV-infected persons combine extraordinarily high potency with breadth of neutralization**

n engl j med 370;4 january 23, 2014

Nature 303; November 14, 2013

Penn Medicine 48

There has been an explosion of bNABs against HIV



CD4-binding site
b12, VRC01, VRC07, NIH45-46, 3BNC117, VRC-PG04

V1/V2
PG9, PG16, CH01-04, PGT141-145, PGDM1400

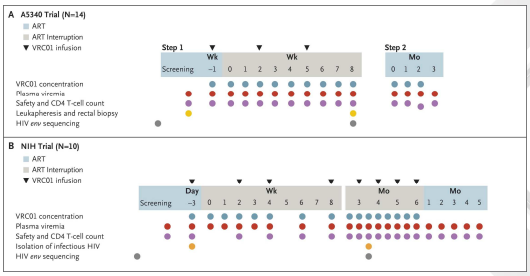
V3/Asn332 glycan patch
PGT121-123, PGT125-131, PGT135, 10-1074, 2G12

gp120/gp41-interface
PGT151, 35O22, 8ANC195

MPER
2F5, 4E10, 10E8

The paper was the combination of 2 studies

Would the presence of passively given VRC01 would prevent the rebound of viremia during an ATI?



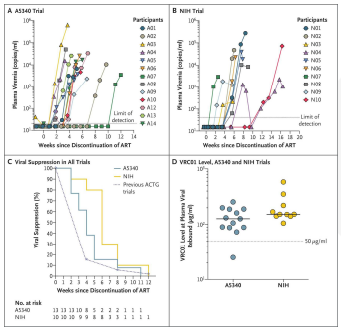
Bar, et al. New England Journal of Medicine 375.21 (2016): 2037-2050

Table 1. Characteristics of the Participants at Baseline.*

Characteristic	A5340 Trial (N = 14)	NIH Trial (N = 10)	Historical Controls from Previous ACTG Studies (N = 61)
Sex — no. (%)			
Male	14 (100)	8 (80)	53 (87)
Female	0	2 (20)	8 (13)
Age — yr			
Median (IQR)	38 (34–44)	51 (44–56)	44 (40–50)
Range	27–52	33–59	27–73
Race or ethnic group — no. (%)†			
White non-Hispanic	6 (43)	6 (60)	41 (67)
Black non-Hispanic	6 (43)	3 (30)	13 (21)
Hispanic, regardless of race	2 (14)	1 (10)	7 (11)
Weight — kg			
Median (IQR)	86 (77–102)	83 (78–89)	NA
Range	60–115	75–100	NA
HIV RNA — copies/no. (%)			
<50 copies/ml	13 (93)	10 (100)	61 (100)
≥50 copies/ml	1 (7)	0	0
CD4 T-cell count — cells/mm^3			
Median (IQR)	896 (579–1053)	724 (630–926)	852 (686–1048)
Range	470–1586	577–1616	350–1667
Nadir CD4 T-cell count — no. (%)			
<201 cells/mm^3	0	2 (20)	3 (5)
201–500 cells/mm^3	12 (86)	3 (30)	39 (64)
>500 cells/mm^3	2 (14)	4 (40)	16 (26)
Unknown	0	1 (10)	3 (5)
Duration from initiation of ART to study entry — yr			
Median (IQR)	4.7 (3.8–6.0)	10.0 (7.7–13.3)	5.6 (4.1–6.7)
Range	2.7–14.5	7.0–17.2	0.7–16.8

The answer was simple: just a little bit…

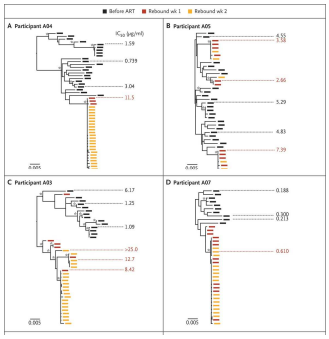
Despite high levels of antibody, everybody rebound. Some participants took longer

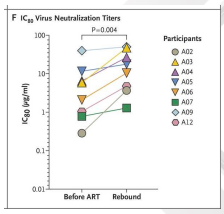


Bar, et al. New England Journal of Medicine 375.21 (2016): 2037-2050

Why?

Most likely preexisting resistance

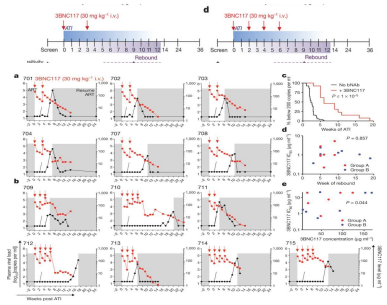




Bar, et al. New England Journal of Medicine 375.21 (2016): 2037-2050

LETTER

HIV-1 antibody 3BNC117 suppresses viral rebound in humans during treatment interruption



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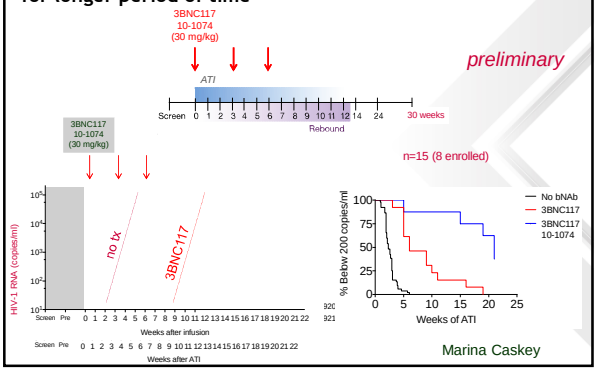
J F Scheid *et al. Nature* **535,** 556–560 (2016)

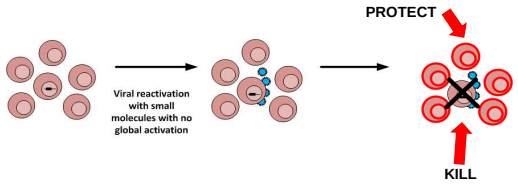
55

First combo bNABs studies
3BNC117 plus 10-1074 appear to delay viral rebound for longer period of time



Marina Caskey

Step 3. Protecting the uninfected cells



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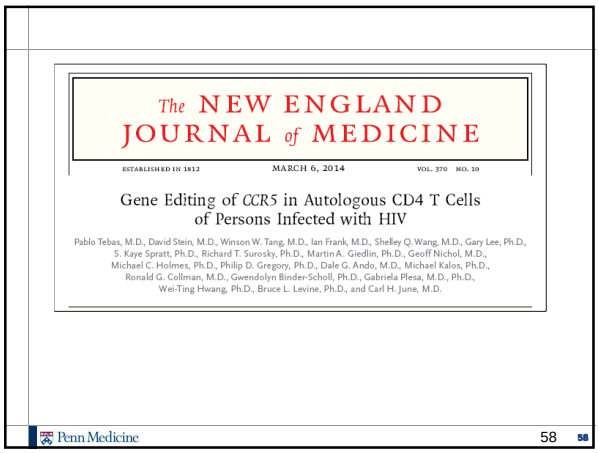


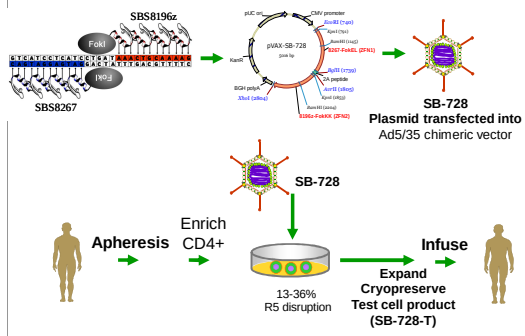
The NEW ENGLAND JOURNAL of MEDICINE

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

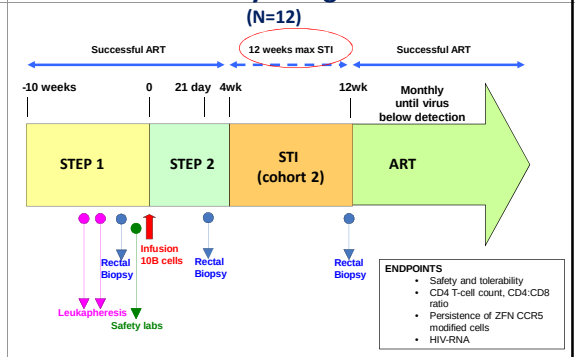
Gene Editing of *CCR5* in Autologous CD4 T Cells of Persons Infected with HIV

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.

Autologous CCR5 Modified CD4+ T-cells

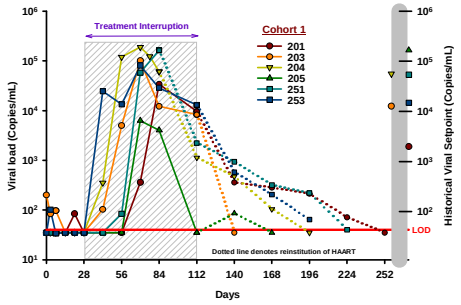


Study Design

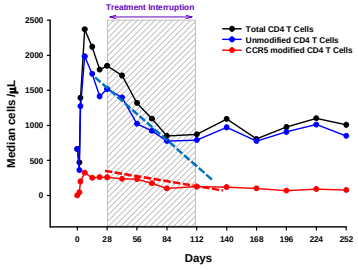


HIV + doing well with high nadir and current CD4 counts

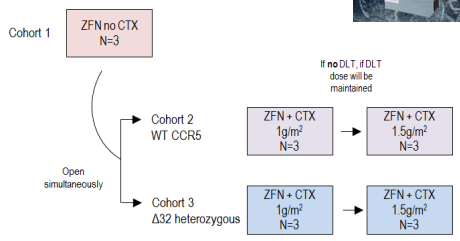
What happened to the viral load during ATI?



Did the modified cells have a survival advantage?



Next study at UPenn (IND approved). Electroporation with CTX



Another strategy for protection at Penn
Blocking R5 and X4 tropic virus using C34

HIV-1 Entry as a Therapeutic Target

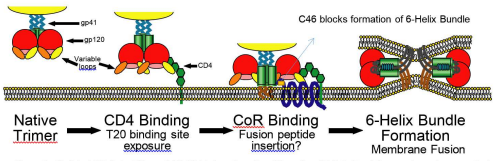
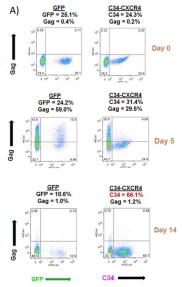
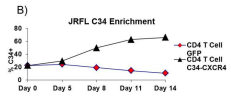


Figure 1. Model of HIV Entry This model highlights how disruption of formation of 6-Helix Bundle) expression make an excellent therapeutic target

Cloning C34 into CXCR4





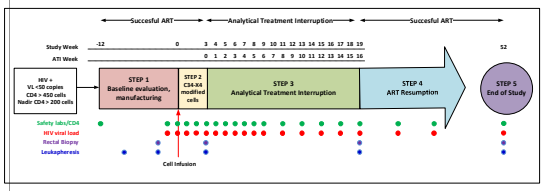
C)

Enrichment of C34:CXCR4 exposed to various HIV strains

Virus	Enrichment C34:X4 Start	Final
JRFL (R5-topic)	25%	66%
BaL (R5 Tropic)	27%	53%
US1 (R5-topic)	27%	60%
CMU01 (X4-topic)	25%	59%
MN (X4-topic)	27%	78%
R3A (R5/X4-topic)	27%	76%
SF2 (R5/X4-topic)	25%	69%

Figure 2. Selective Enrichment of C34:CXCR4 T cells. CD4 cells transduced with either GFP or C34:CXCR4 were diluted with untransduced CD4 cells to a ratio of 1:4 (A, 1st row), and infected with JRFL HIV. Cells were monitored for enrichment using flow cytometry for GFP or C34, and also monitored for HIV infection by intracellular gag staining. Enrichment over time is shown graphically in panel B. This experiment was replicated with multiple strains of HIV with various tropisms, each resulting in dramatic enrichment of the C34:CXCR4-modified cells

C34-X4 modified cells
Proposed study



UPENN gene therapy collaborative group

Penn ACTU
- Larisa Zifchak/Amber/Jenna/Mark
- Joe Quinn
- Pablo Tebas
- Rob Roy MacGregor

Jacoby Medical Center
- David Stein
- Angelo Seda

U. Penn Abramson Inst.
- **Carl June**
- Bruce Levine
- **Jim Riley**
- Richard Carroll
- Gwen Binder
- Liz Veloso

Penn CFAR
- **Clinical Core**
  - Ian Frank
- **Immunology Core**
  - Jean Boyer
- **Viral/Molecular core**
  - Farida Shaheen
  - Ron Collman
  - Rick Bushman
  - Jim Hoxie

ViRxSys

Sangamo

Adaptaminue

Penn CTRC

NIH-NIAID

ACTG

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