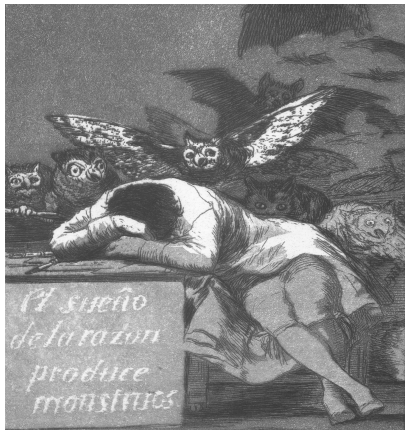


University of Florence

Dept. Experimental and Clinical Medicine

Unit Internal Medicine

GA²LEN
Global Allergy and Asthma European Network
Network of Excellence

Perchè non si parla più di AIDS ?

Paola Parronchi

XXX Congresso della Società Italiana di Allergologia, Asma ed Immunologia Clinica
Firenze 6-9 Aprile 2017

L'immunologo Ferdinando Aiuti nel famoso bacio con una donna sieropositiva

WORLD AIDS DAY


Does HIV cause AIDS?

The failure of a much sought-after vaccine against the virus has re-ignited an old debate. Mayank Tiwari explores the controversy

NCBI Resources How To

PubMed.gov

US National Library of Medicine
National Institutes of Health

PubMed

Results by year

HIV (artist illustration) could be kept at bay by editing the DNA of immune cells.

DISEASE

Closing the door on HIV

Although yet to complete clinical trials, genome editing has already shown promise against a globally important disease.

SABATO 15 OTTOBRE 2016 - SALA ARTEMISIA

15:00 *Workshop*

Immunodeficienze primitive e secondarie: approcci diagnostici e nuove modalità terapeutiche

Moderatori: Franco Dammaco (Bari), Angelo Vacca (Bari)

- L'immunità innata e i difetti funzionali e quantitativi dei granulociti neutrofili
  Baldassarre Martire (Bari)
- Malattie infiammatorie croniche dell'apparato respiratorio e di quello intestinale: il modello dei pazienti con difetti primitivi dell'immunità
  Isabella Quinti (Roma)
- Immunodeficienze secondarie: epidemiologia, diagnosi e terapia
  Carlo Agostini (Padova)

Società Italiana di Allergologia, Asma ed Immunologia Clinica

siaaic

MEETING SERIES

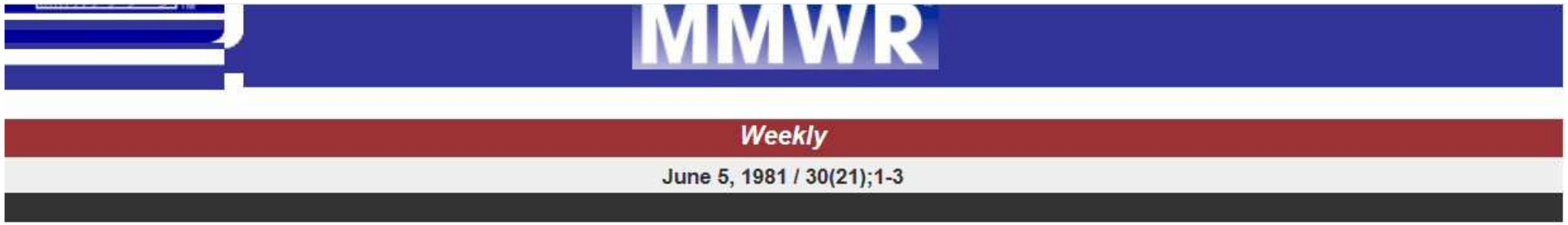
VII WINTER SCHOOL "Mario Ricci"

di Allergologia ed Immunologia Clinica

HOTEL MULINO DI FIRENZE

13 febbraio - 14 febbraio 2016, **Firenze**

The HIV epidemics: 36 years apart



Epidemiologic Notes and Reports

Pneumocystis

Pneumonia --- Los Angeles

In the period October 1980-May 1981, 5 young men, all active homosexuals, were treated for biopsy-confirmed *Pneumocystis carinii* pneumonia at 3 different hospitals in Los Angeles, California. Two of the patients died. All 5 patients had laboratory-confirmed previous or current cytomegalovirus (CMV) infection and candidal mucosal infection. Case reports of these patients follow.

"*Pneumocystis* pneumonia in the United States is almost exclusively limited to severely immunosuppressed patients. (…) The fact that these patients were all homosexuals suggests an association between some aspect of a homosexual lifestyle or disease acquired through sexual contact "

Center for Disease Control's Morbidity Mortality Weekly Report (MMWR), June 5th, 1981

HIV epidemics and Global Health Observatory (GHO) data

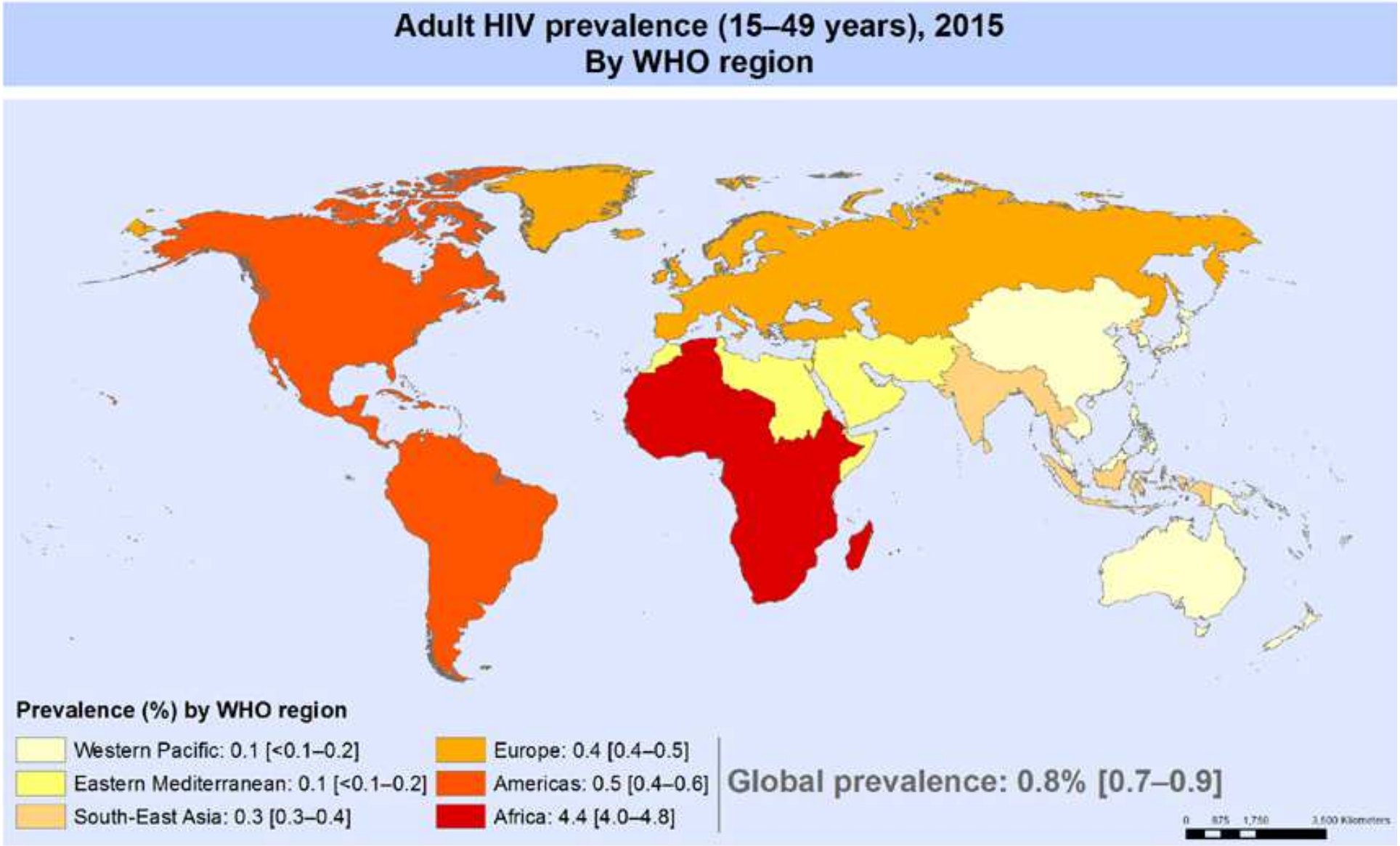
- >70 million people have been infected since the beginning

- >40 million people have died of HIV since the beginning, 1.1 million died in 2015

- 36.7 million people living with HIV (end 2015, 2.2 in West/Central Europe and USA), 0.8% of aged 15–49 years HIV+,15.8 million receiving ART (41%)

-1 HIV+ in every 25 adults in Sub-Saharan Africa (~70% of the people living with HIV worldwide), 14% in Asia and the Pacific

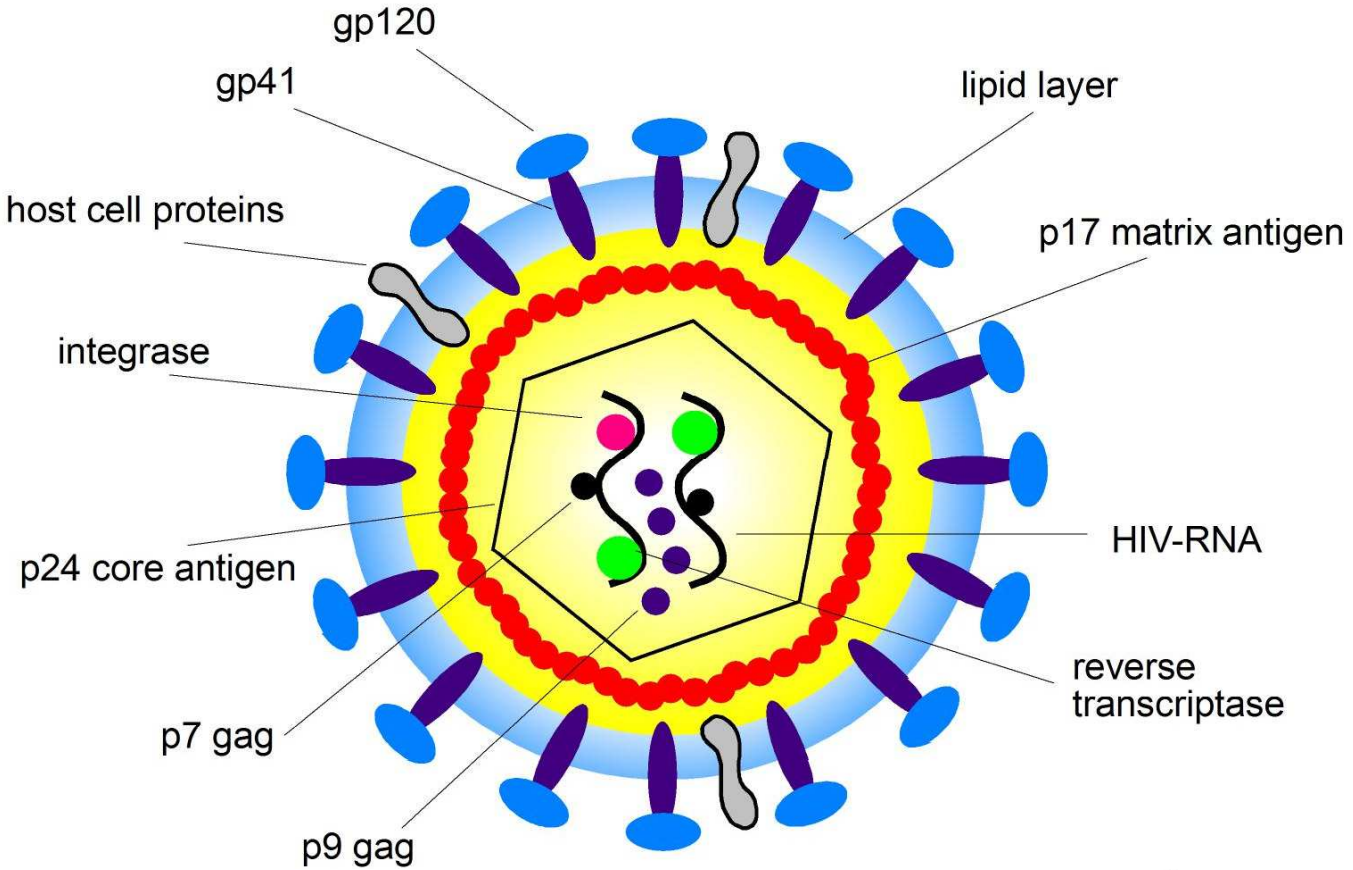
HIV epidemics and Global Health Observatory (GHO) data

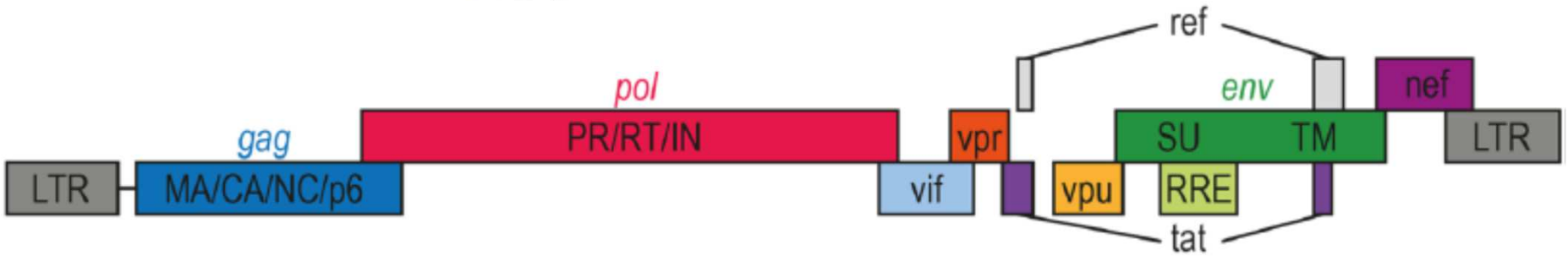


Major breakthroughs in HIV research

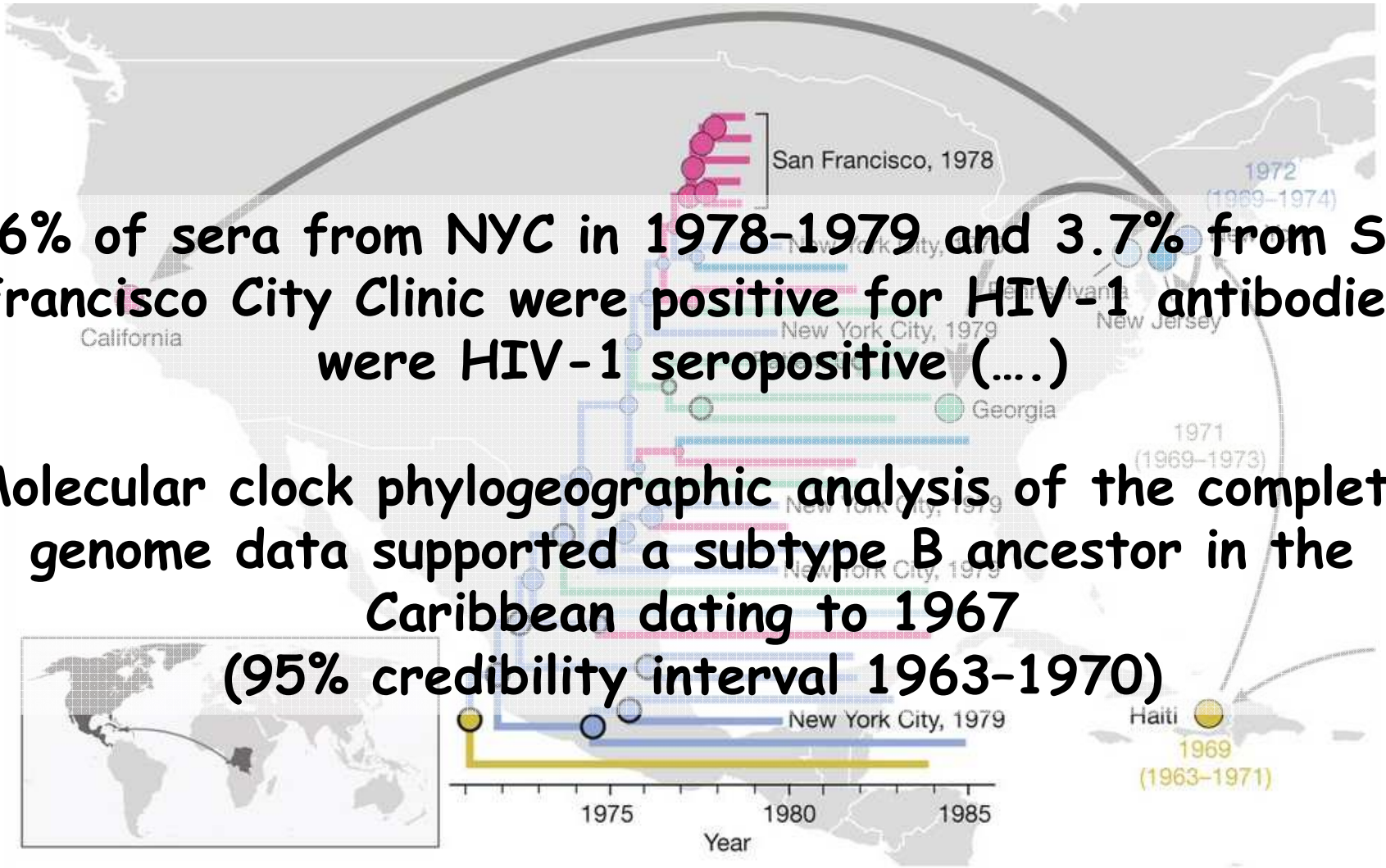
- Molecular medicine and nucleic acid technology
- Targeted antiviral therapy
- Arrest the spread of the disease with strategies for prevention
- AIDS vaccine
- Immunological studies (latency, bnAbs, animal models, cell functions….)

Structure of the virus has been almost definitively acquired





The HIV epidemics comes from far

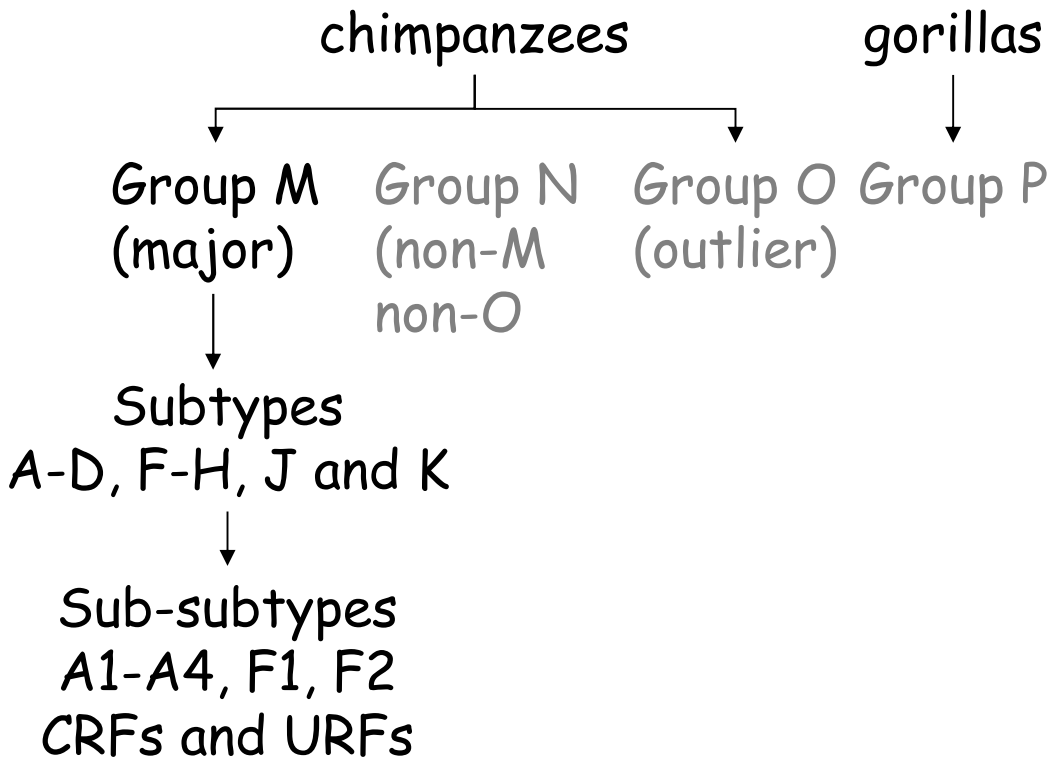


6.6% of sera from NYC in 1978–1979 and 3.7% from San Francisco City Clinic were positive for HIV-1 antibodies were HIV-1 seropositive (….)

Molecular clock phylogeographic analysis of the complete genome data supported a subtype B ancestor in the Caribbean dating to 1967 (95% credibility interval 1963–1970)

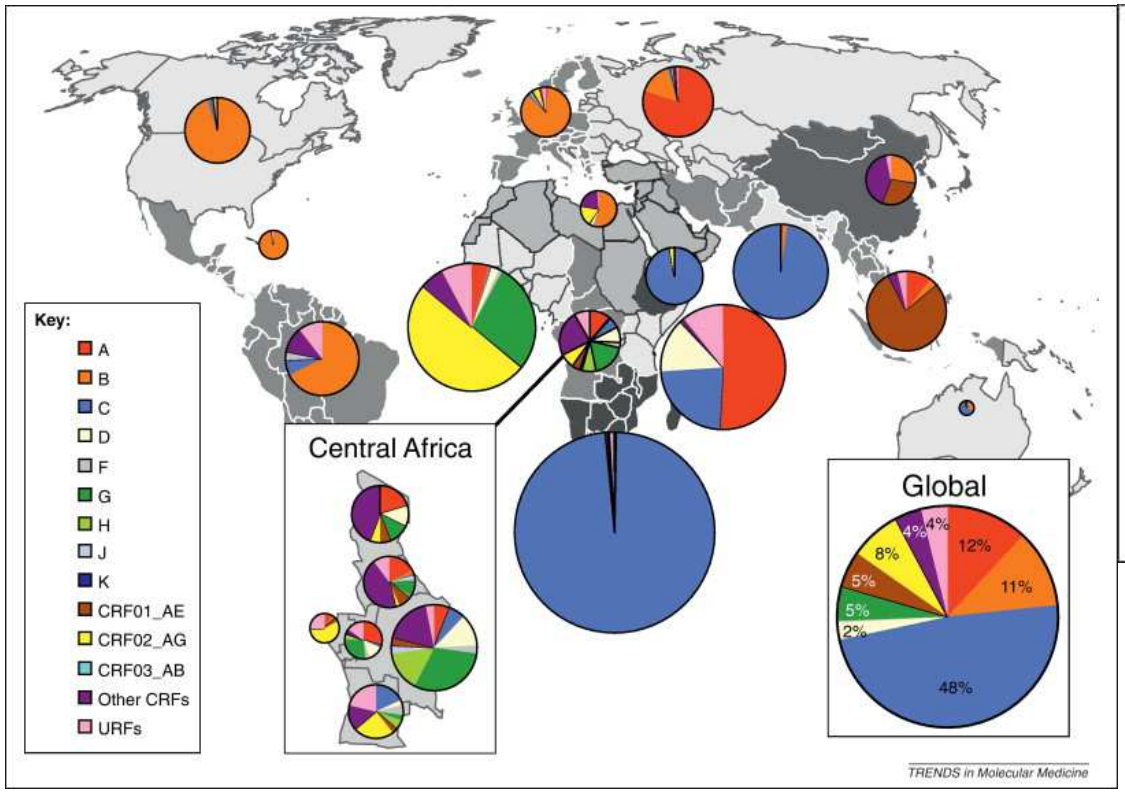
Nature 539, 98–101 (03 November 2016) doi:10.1038/nature19827

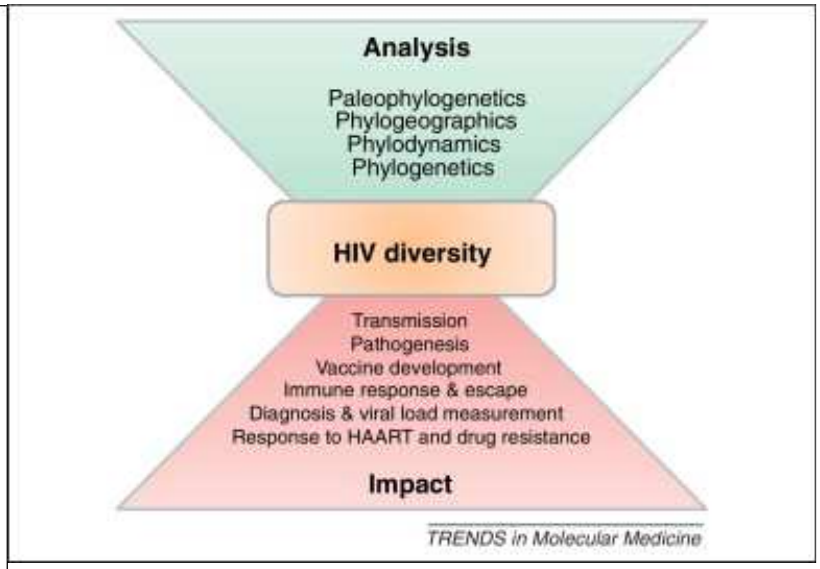
HIV viral subtypes



AIDS Rev. 2014 Jan-Mar; 16(1): 23–34.

HIV viral subtypes





fast replication cycle of the virus +
high error-prone function of its reverse transcriptase

Trends Mol Med 2012. 18:182–192

Major breakthroughs in HIV research

- Molecular medicine and nucleic acid technology
- Targeted antiviral therapy
- Arrest the spread of the disease with strategies for prevention
- AIDS vaccine
- Immunological studies (latency, bnAbs, animal models, cell functions….)

The six classes of antiretroviral agents currently available

1. Nucleoside reverse transcriptase inhibitors (NRTIs);

2. non-nucleoside reverse transcriptase inhibitors (NNRTIs);

3. protease inhibitors;

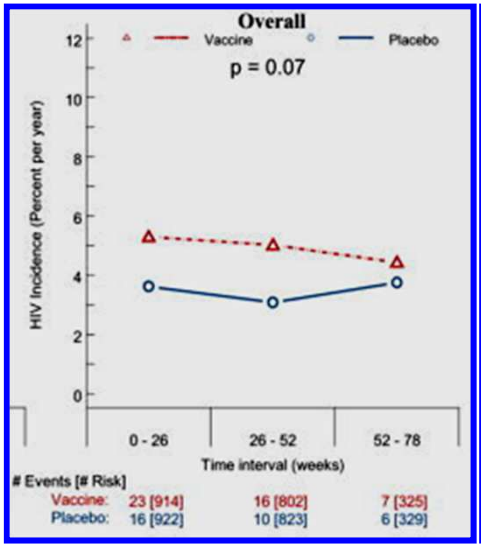
4. integrase inhibitors;

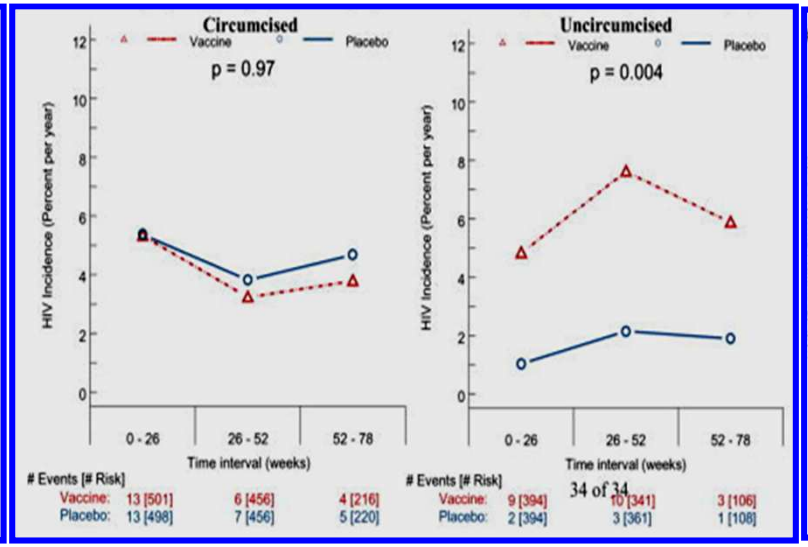
5 fusion inhibitors and chemokine receptor antagonists (CCR5 antagonists).

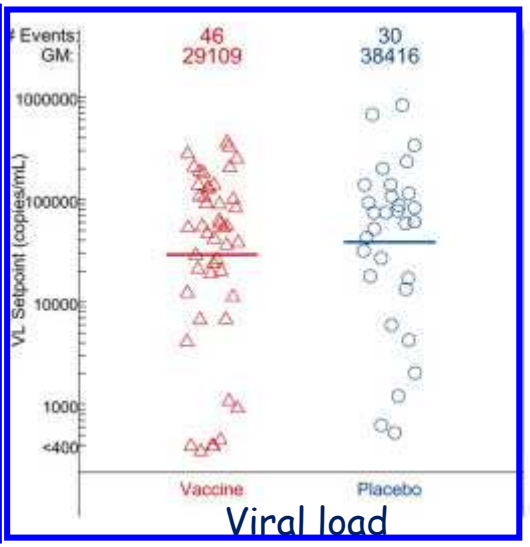
Major breakthroughs in HIV research

- Molecular medicine and nucleic acid technology
- Targeted antiviral therapy
- Arrest the spread of the disease with strategies for prevention
- AIDS vaccine
- Immunological studies (latency, bnAbs, animal models, cell functions….)

2008:Merck HIV vaccine fails, trials halted







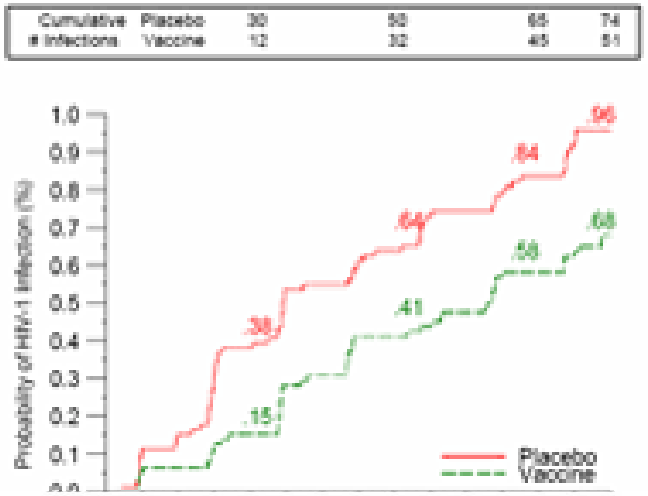
Viral load

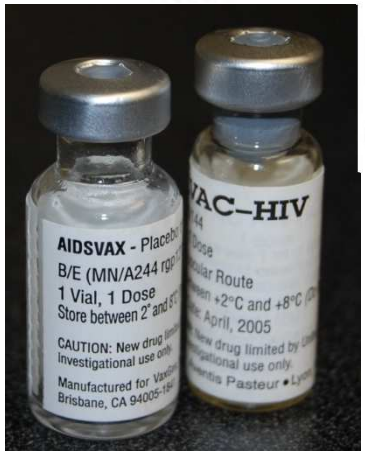
Table 1: STEP Trial Results—Number of infections and post-infection viral load levels

Trial Cohort	Vaccine: Infections	Placebo: Infec-tions	Vaccine: Post infection viral loads	Placebo: Post infection viral loads	Vaccine: Infections (men)	Placebo: Infections (men)	Vaccine: Infections (women)	Placebo: Infections (women)
First 1,500 person cohort, anti-Ad5 antibody levels <1:200	28	25	41,527 (n=25)*	26,696 (n=21)*	28 (n=522)	24 (n=536)	0	1
Second 1,500 person cohort, anti-Ad5 antibody levels >1:200	21	9	19,070 (n=21)	89,810 (n=9)	21 (n=392)	9 (n=386)	0	0
All 3,000 persons	49	34	29,109 (n=46)*	38,416 (n=30)*	49 (n=914)	33 (n=922)	0	1

Lancet 2009

The Thai vaccine (RV144)





In the face of 2 million new infections per year, the stagnation of progress towards an efficacious HIV vaccine is sobering. In 2009 a double-blind phase III HIV vaccine RV144 "Thai" trial that used a combination of a recombinant canarypox vector (ALVAC-HIV [vCP1521]) and two booster injections of a recombinant glycoprotein 120 subunit vaccine (AIDSVAX B/E) showed marginal, yet significant protection from HIV acquisition[1]

- 125 infections
  - Vaccine: 51
  - Placebo: 74
- VE: 31.2%, p=0.04
- adj. 95% CI: 1.1, 52.1

NY Acad Sci 2010

HivVaccineTrialNetwork 100 vaccine (official code NCT02404311)

Controlled
Randomized
Double-blind
vs placebo

Jan 2015-jan 2017
Results 2020

>5000 naive subjects
men/women 18-40 yrs

ALVAC
(recombinant canary pox vector replacing gp120 insert)

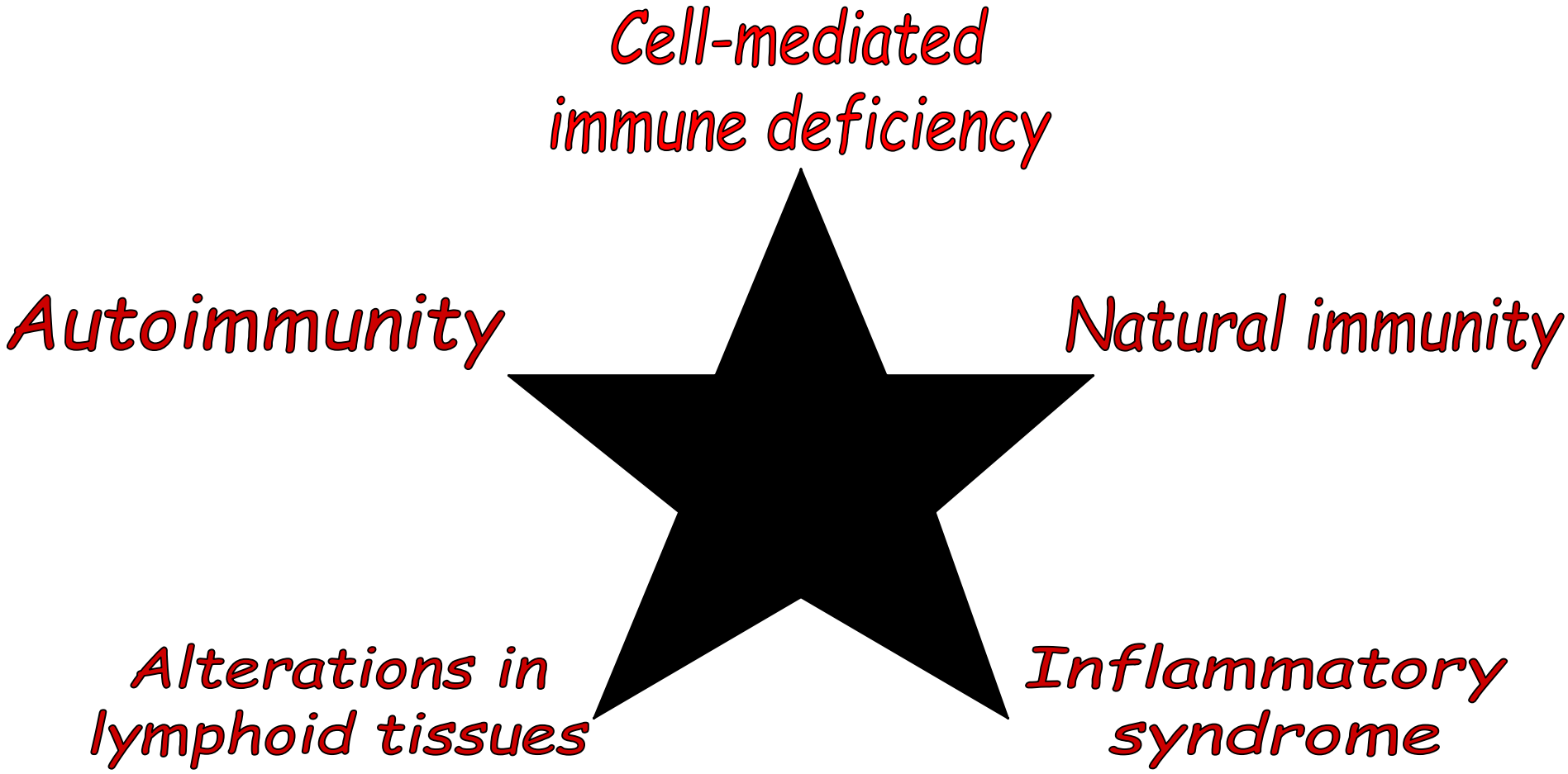
+ 2-5 IM recombinant monomeric proteins Clade C
(100 mcg each in MF59®)

Phase III efficacy trial (HVTN 702)

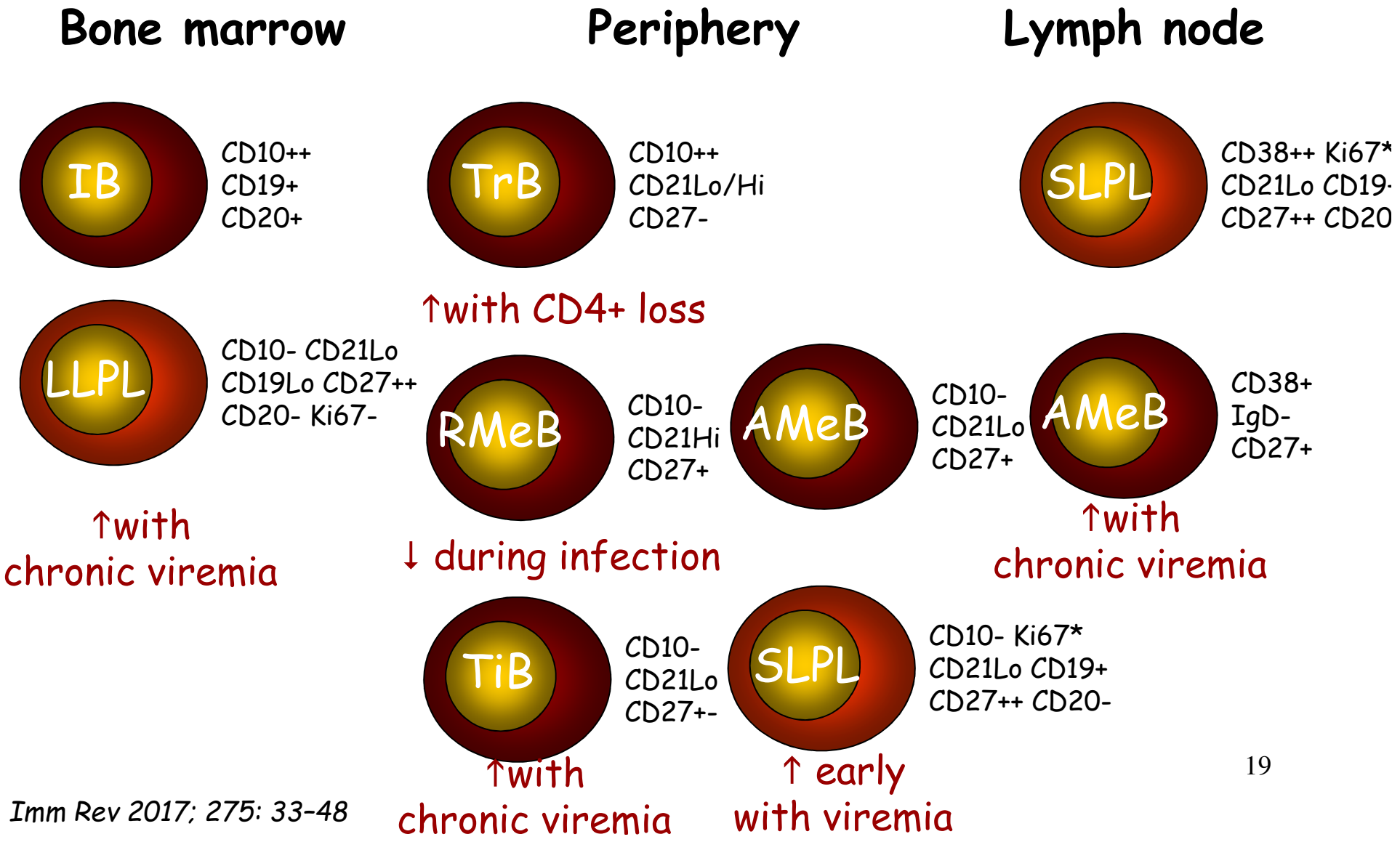
Trial Sponsors

National Institute of Allergy and Infectious Diseases (NIAID), HIV Vaccine Trials Network Bill and Melinda Gates Foundation, Medical Research Council Sanofi Pasteur, a Sanofi Company, Novartis Vaccines

Immunological hallmarks of AIDS



Alteration in B cells and passive immunization with bnAbs



Imm Rev 2017; 275: 33-48

The interplay between HIV and the immune system

HIV

Gp120/CD4
CCR5/CXCR4 ↓ Inverse transcriptase

CD4+ cells

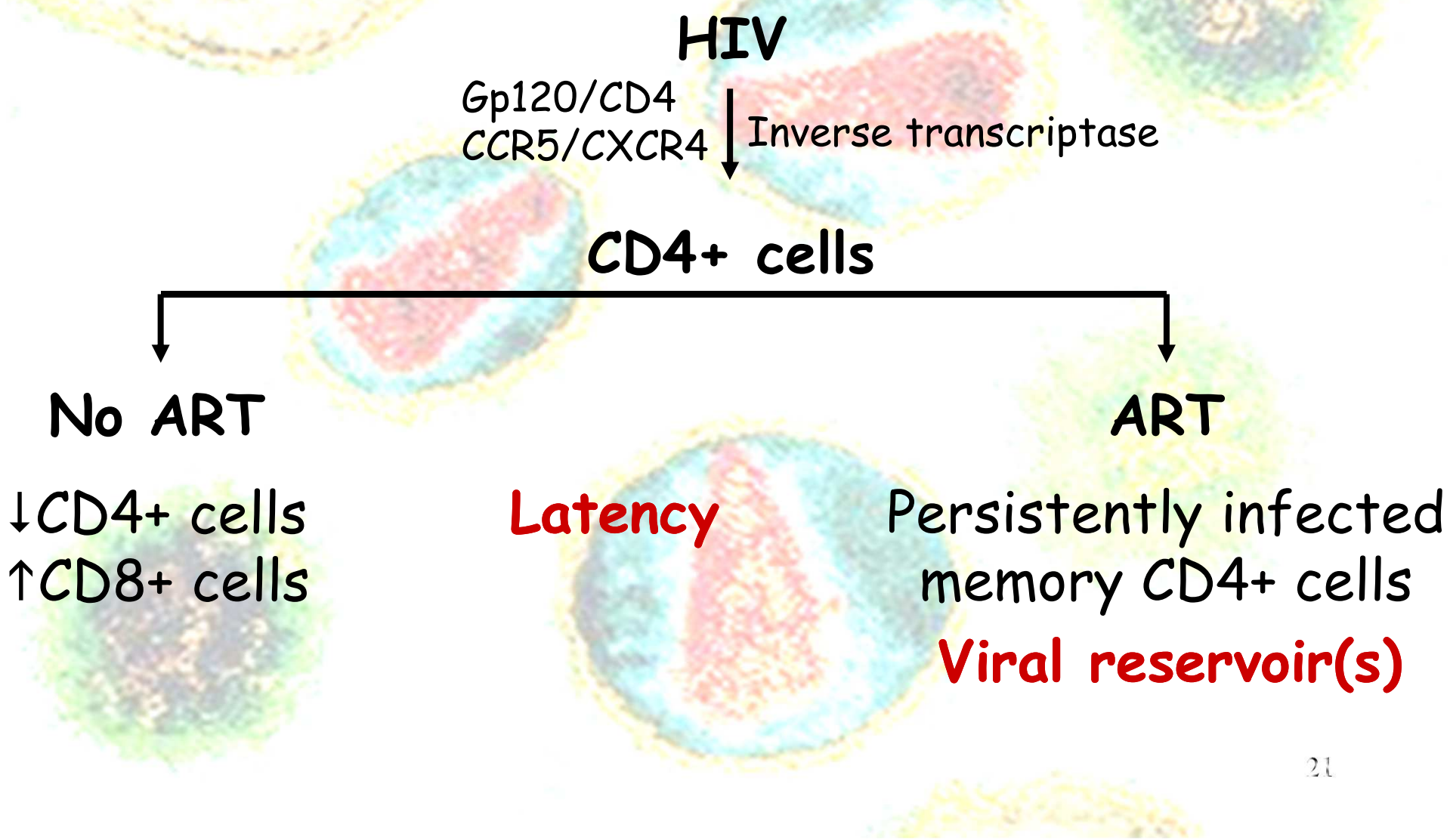
No ART

↓CD4+ cells
↑CD8+ cells

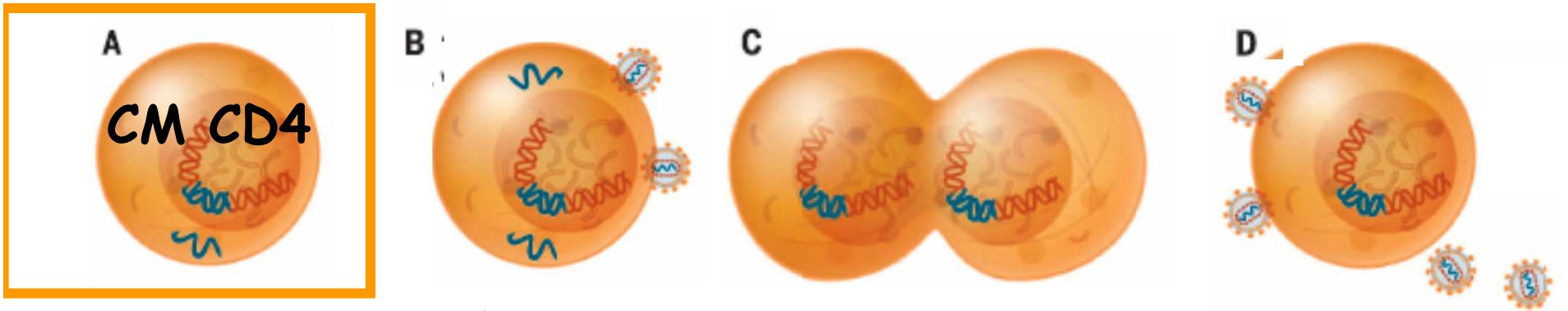
ART

Persistently infected memory CD4+ cells

The interplay between HIV and the immune system



Latency in HIV infection from the CD4 point of view



True virological and transcriptional latency

little HIV RNA expression
no detectable HIV presentation

"active latency"

ongoing production of HIV RNA and antigen

Proliferation of latently infected cells

without viral production

de novo infection despite AR

early and profound loss of splenic Tfh cells

promising as novel immunotherapeutic important for B cell differentiation/Abs production

are infected during the HIV infection represent a reservoir

Frontiers in Immunology | www.frontiersin.org 1 February 2017 | Volume 8 | Article 135

HIV

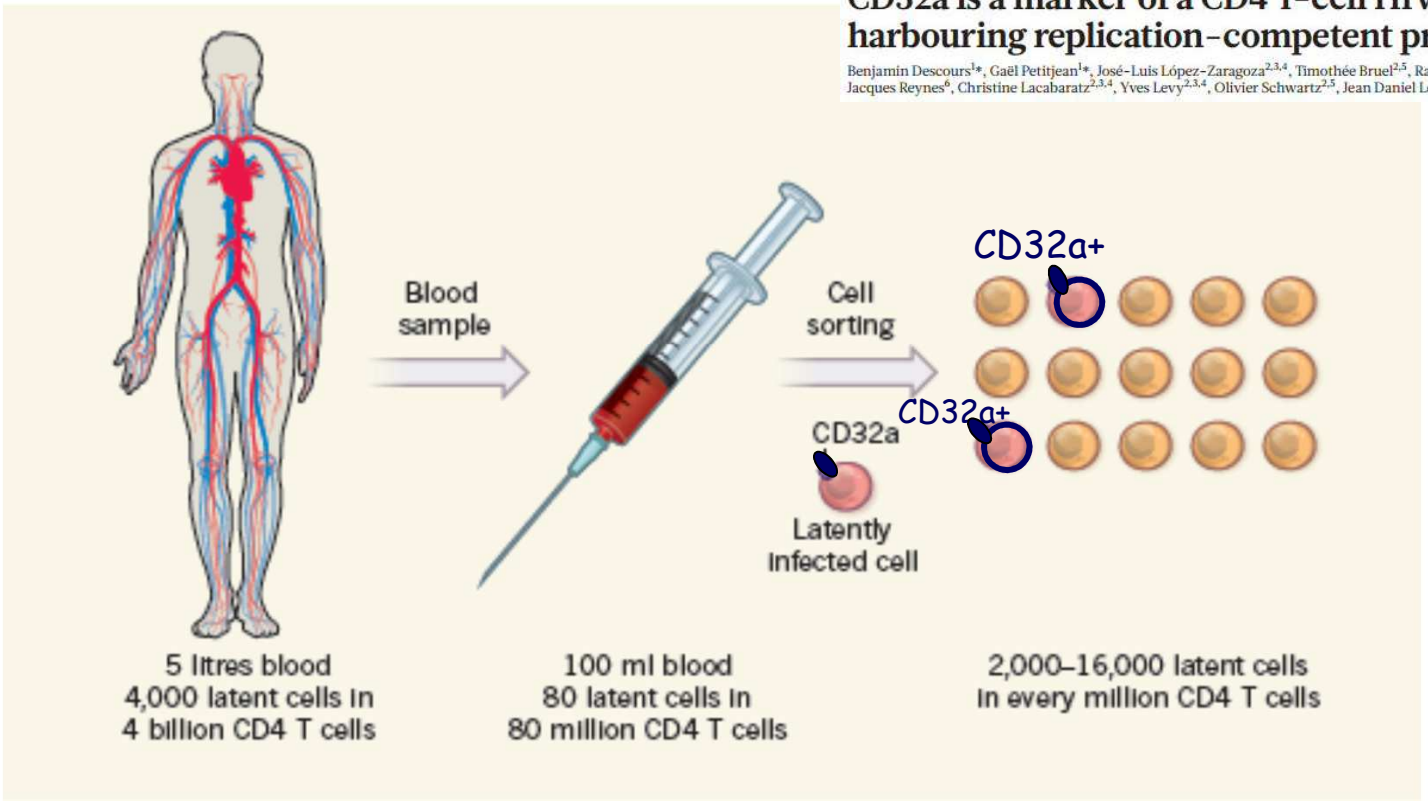
23 MARCH 2017 | VOL 543 | NATURE | 499

Finding latent needles in a haystack

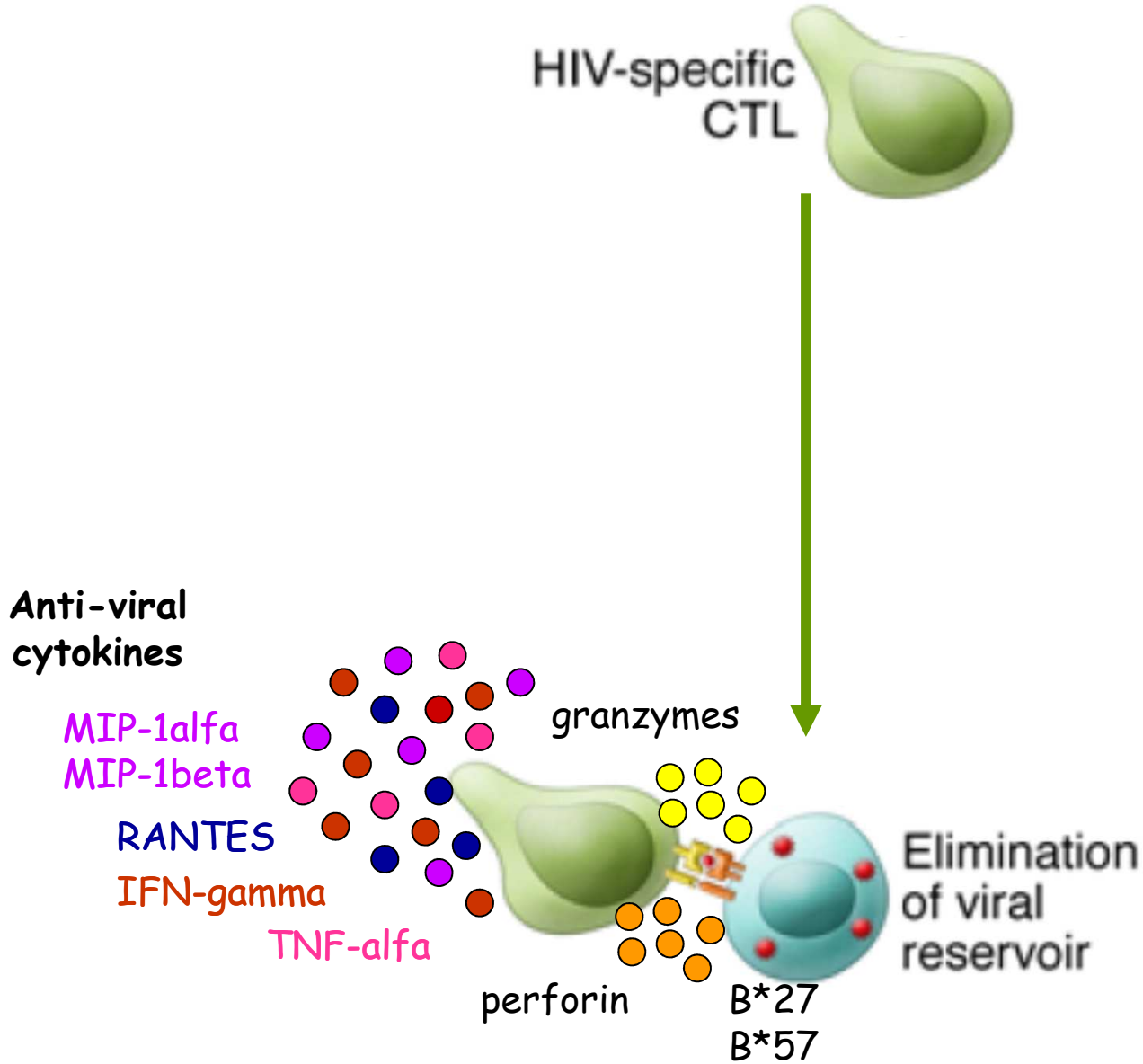
Antiretroviral therapy can keep HIV at bay, but a few cells remain infected, so the disease cannot be cured. The discovery of a protein that marks out these infected cells will facilitate crucial studies of this latent viral reservoir. SEE LETTER P.564

CD32a is a marker of a CD4 T-cell HIV reservoir harbouring replication-competent proviruses

Benjamin Descours[1]*, Gaël Petitjean[1]*, José-Luis López-Zaragoza[2,3,4], Timothée Bruel[2,5], Raoul Raffel[1], Christina Psomas[6], Jacques Reynes[6], Christine Lacabaratz[2,3,4], Yves Levy[2,3,4], Olivier Schwartz[2,5], Jean Daniel Lelievre[2,3,4] & Monsef Benkirane[1]

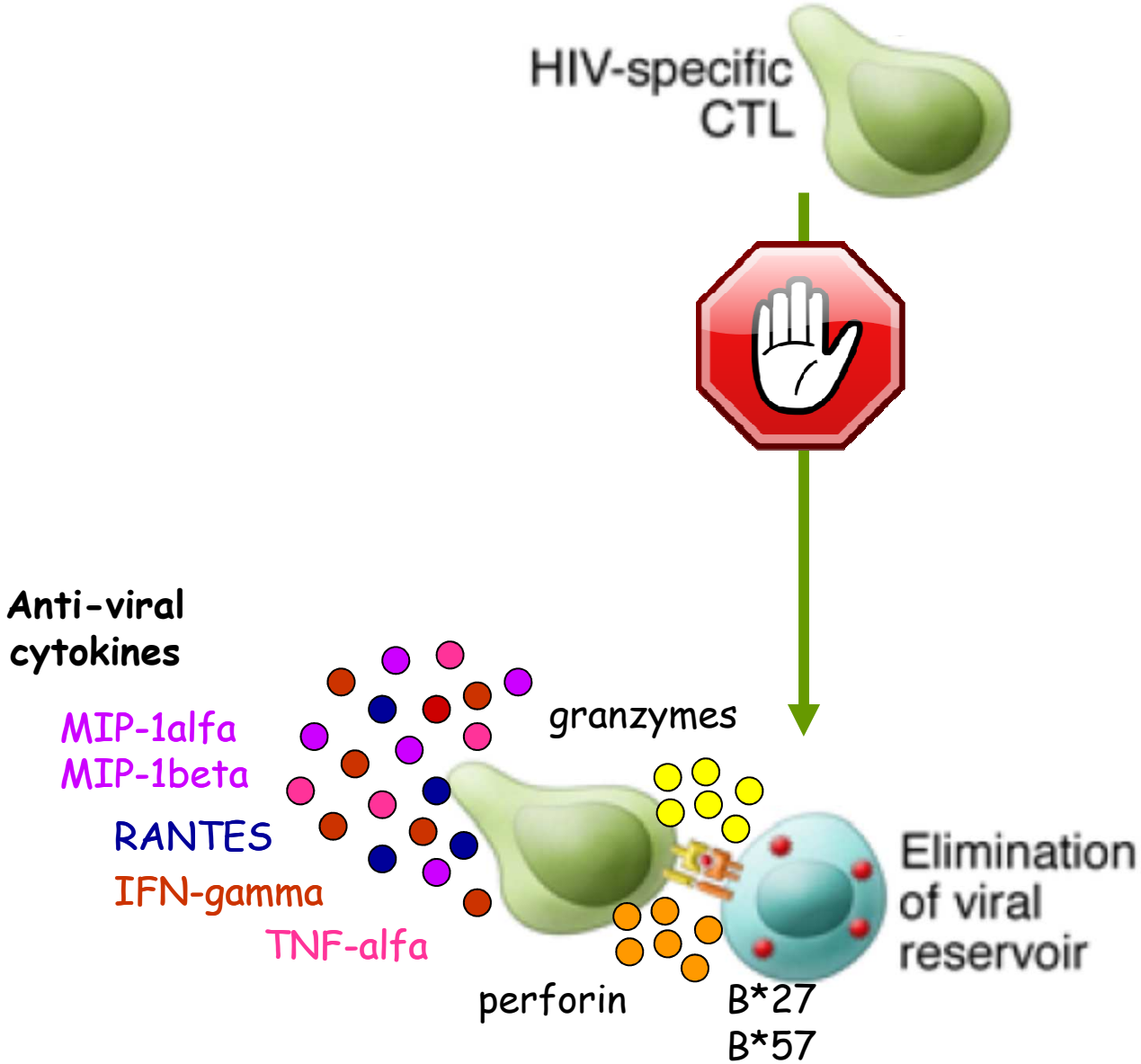


Mechanisms of latency in HIV infection



Mod from RB Jones and BD Wlaker, JCI doi:10.1172/JCI80566

Mechanisms of latency in HIV infection



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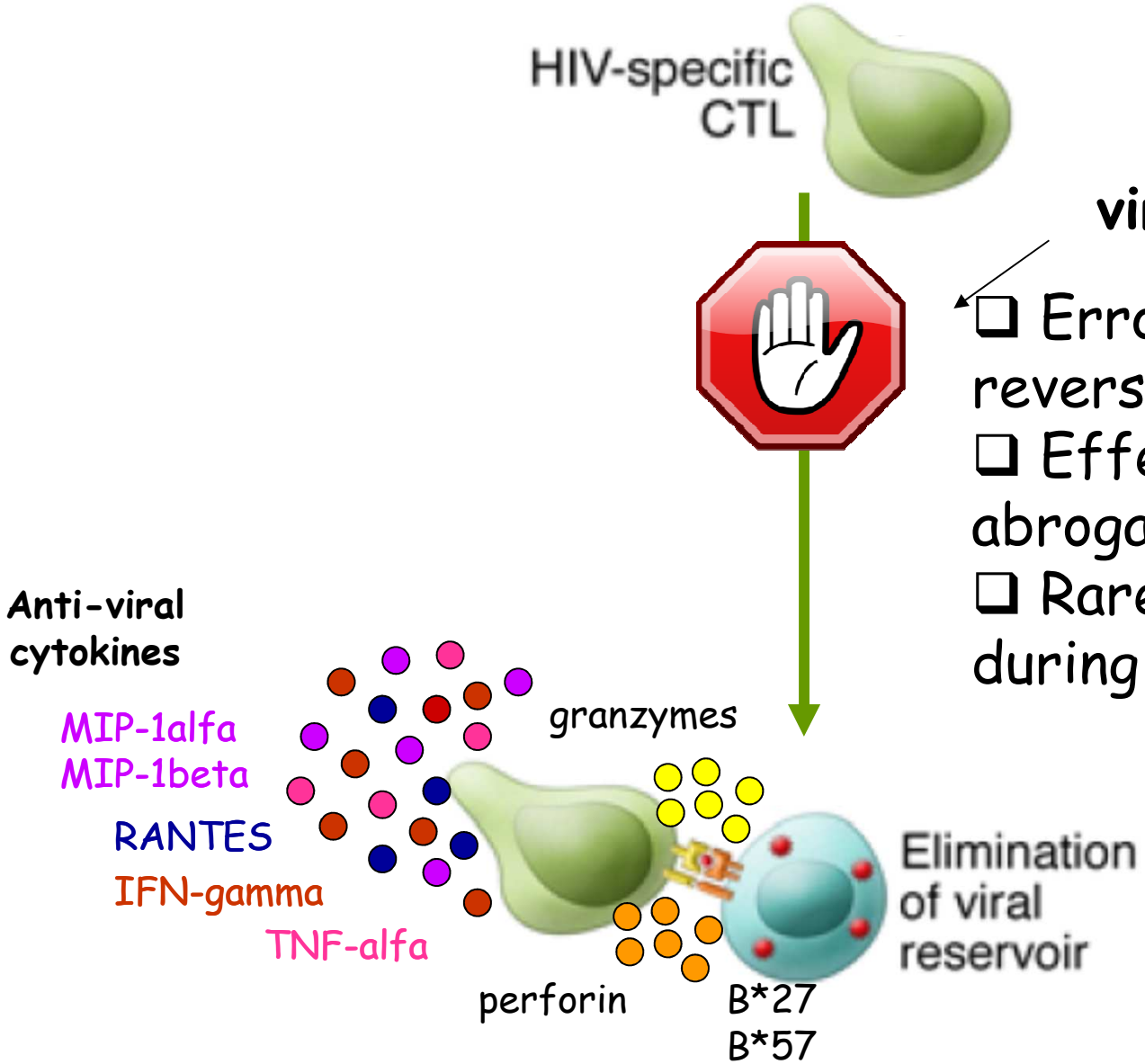
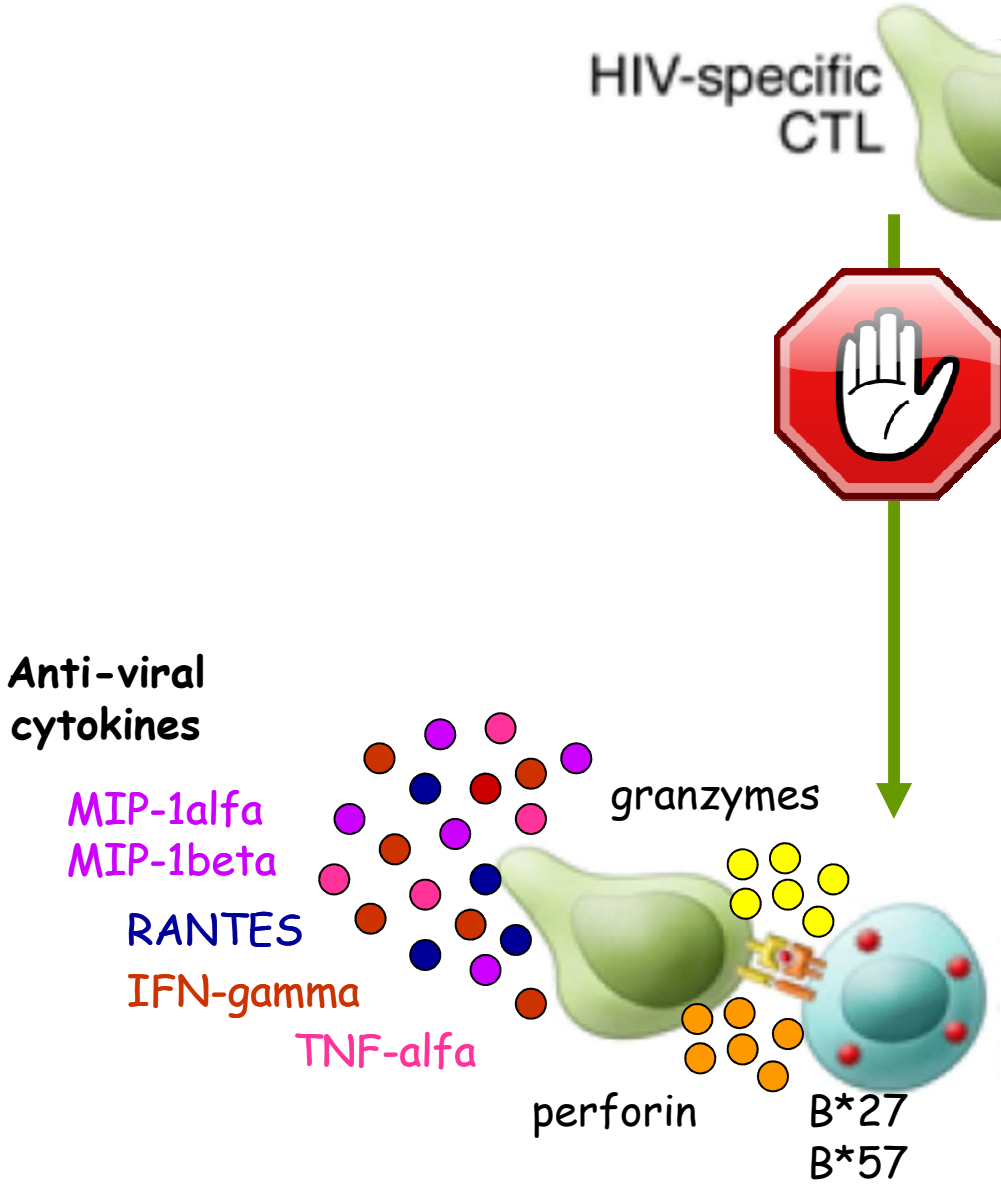
Mechanisms of latency in HIV infection



- Error-prone HIV reverse transcriptase
- Effective cART abrogates viral evolution
- Rare escape mutations during acute infection

Mod from RB Jones and BD Wlaker, JCI doi:10.1172/JCI80566

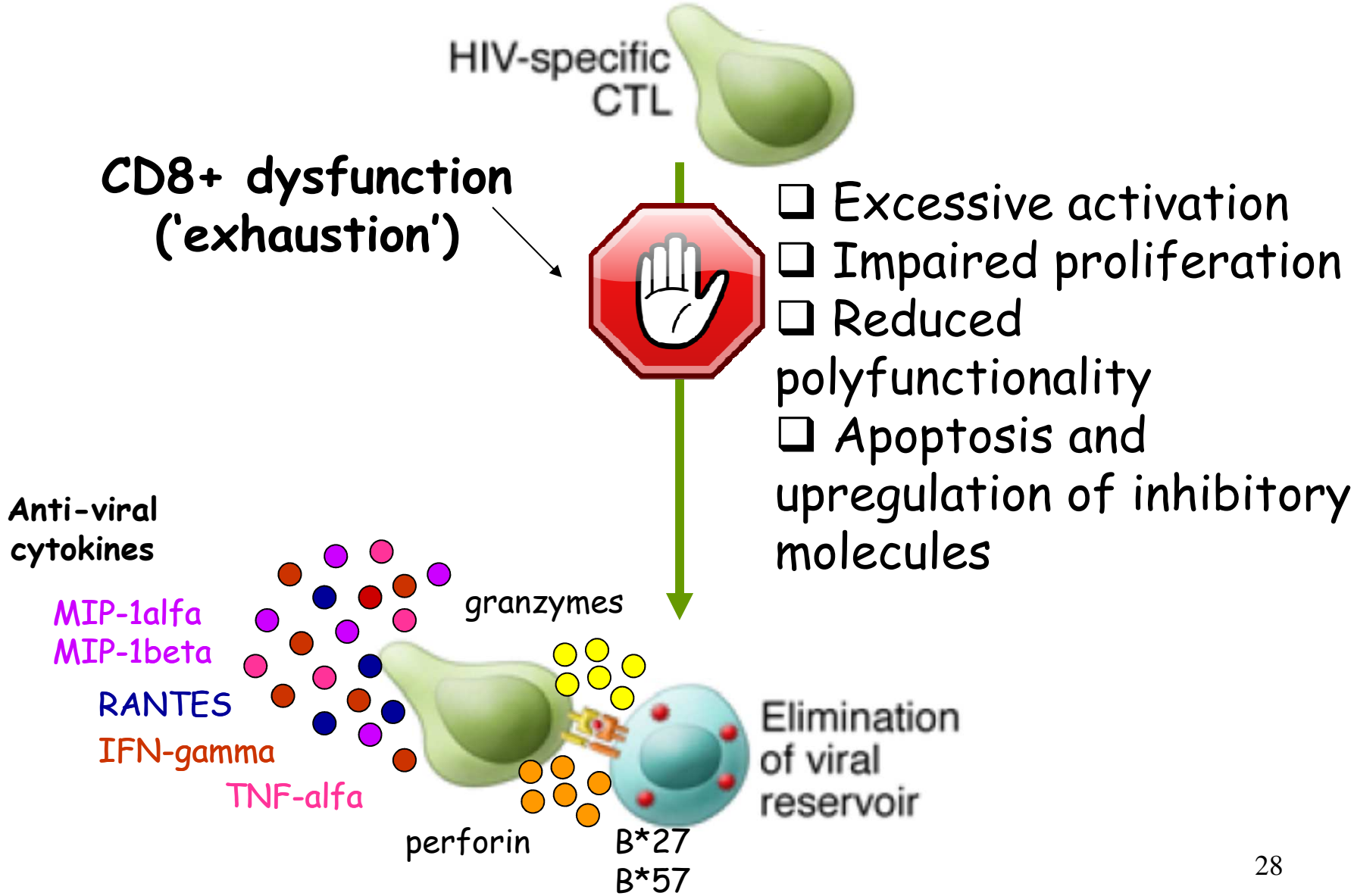
Mechanisms of latency in HIV infection



- ❑ No detectable virion production (1/10^6 CD4+)
- ❑ Highly stable reservoirs (44 mo)
- ❑ Detectable virion production induced by mitogens in vitro

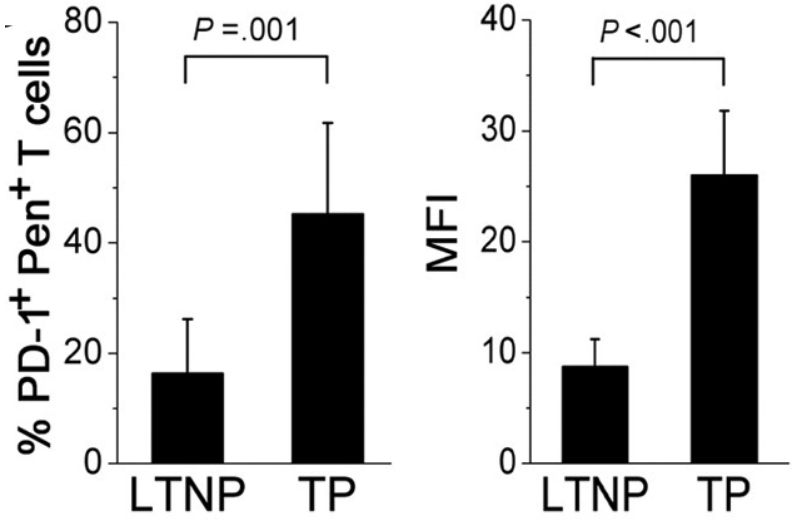
Mod from RB Jones and BD Wlaker, JCI doi:10.1172/JCI805

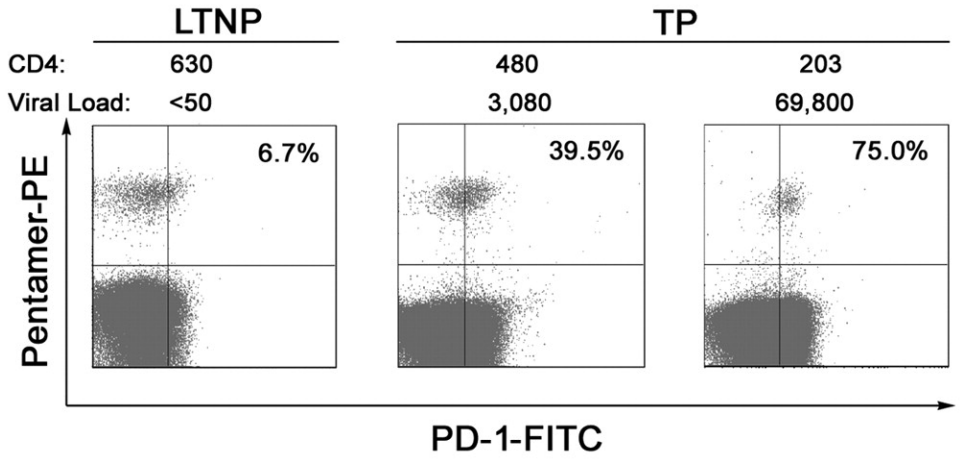
Mechanisms of latency in HIV infection

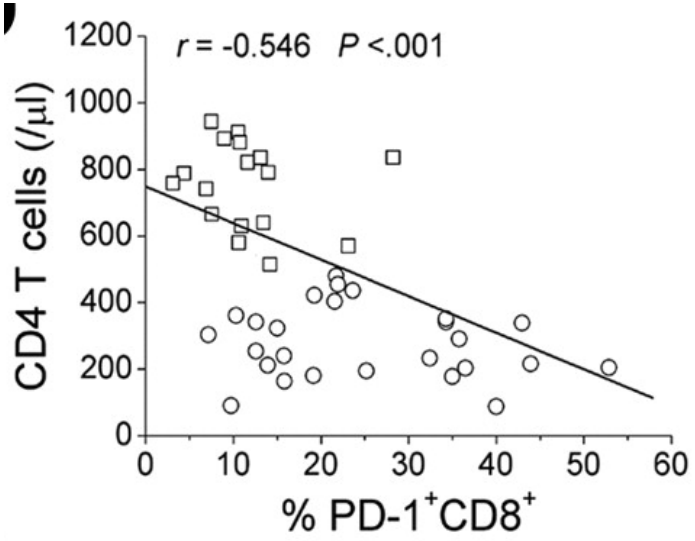


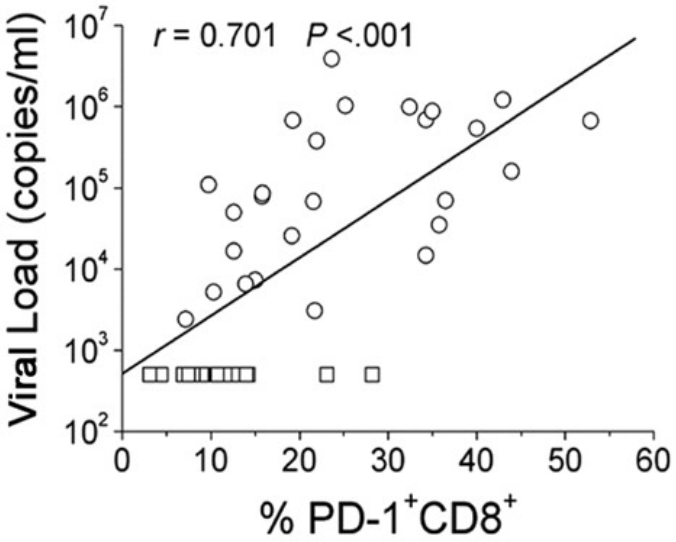
Mod from RB Jones and BD Wlaker, JCI doi:10.1172/JCI80566

PD1-related mechanims of T-cell exhaustion in HIV infection



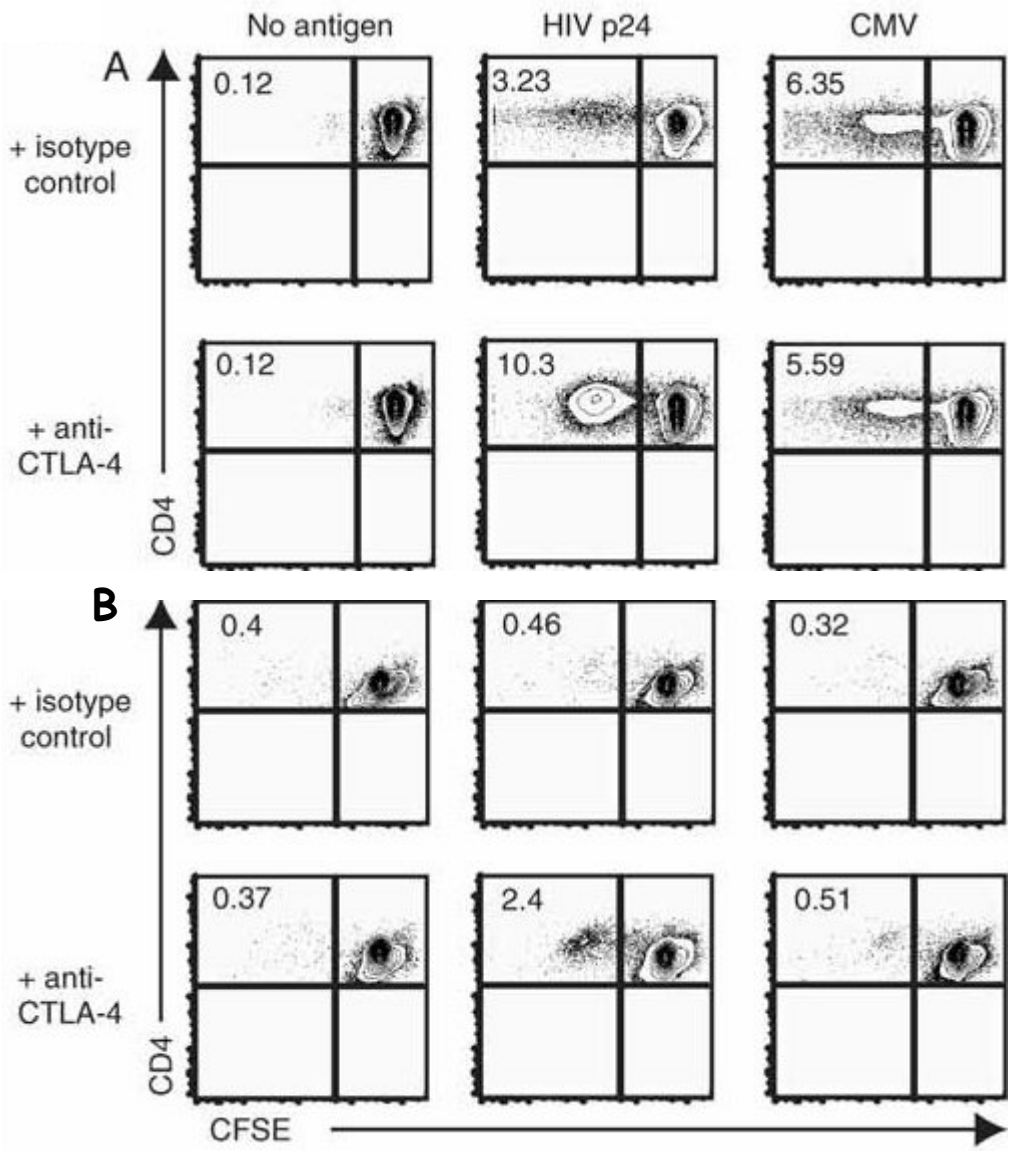




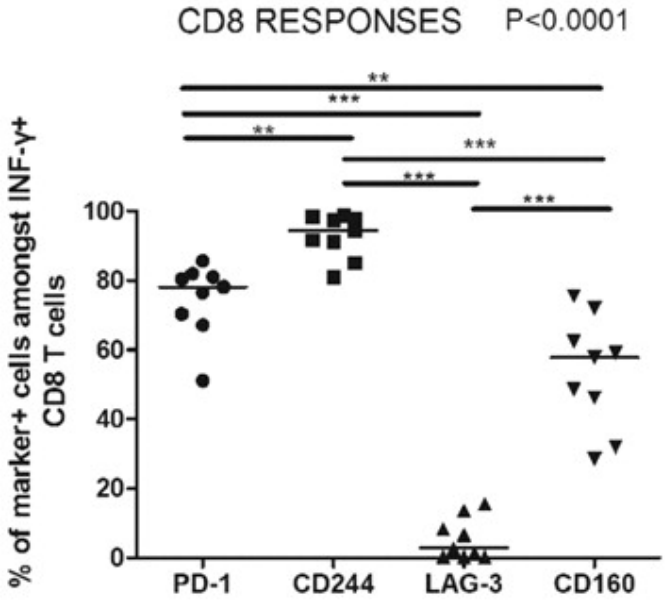


Zhang JY et al, Blood 2007. 109: 4671-8

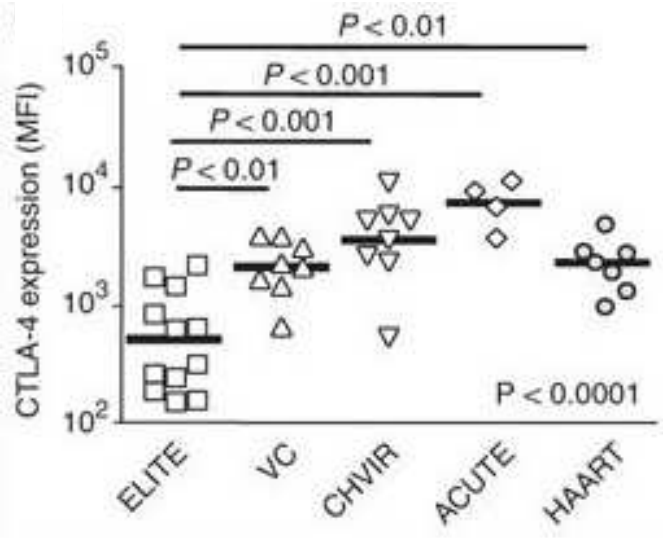
CTLA4 blockade restores HIV-specific CD4+ function



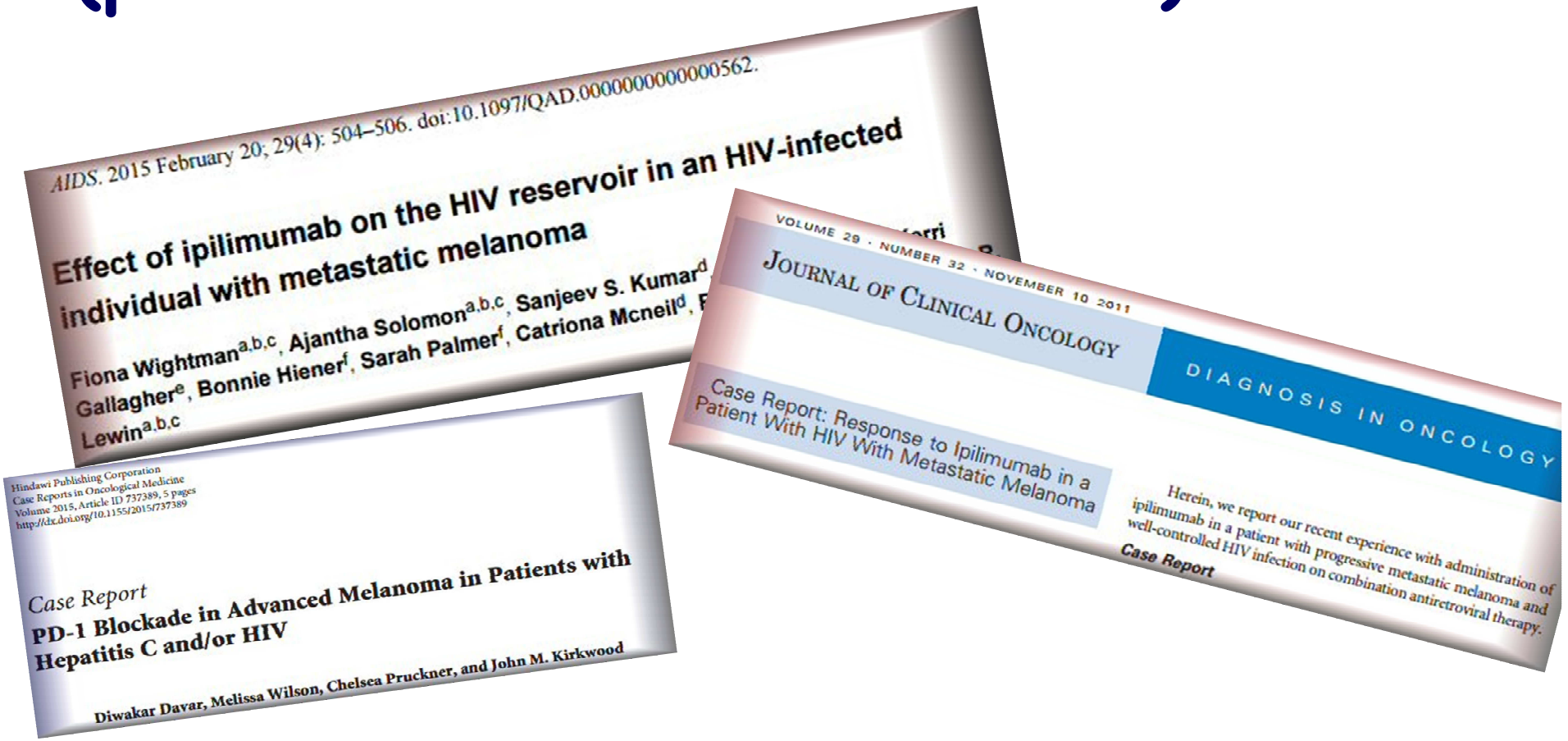
Kaufmann DE et al, Nature Imm 2007. **8**, 1246 - 1254



Porichis F et al, Blood 2011

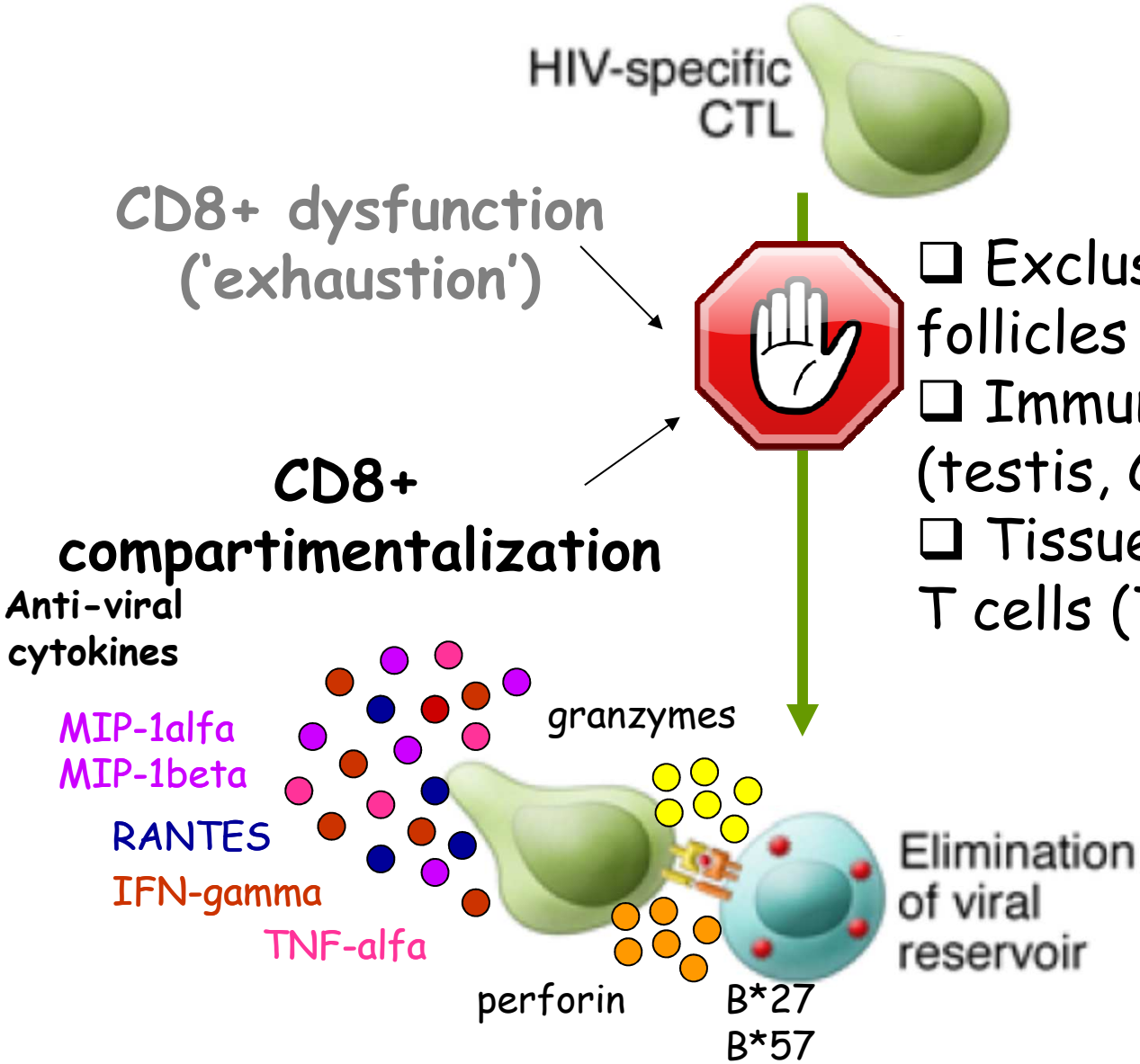


Anti-CTLA4 (ipilimumab) and anti-PD1 (pembrolizumab or nivolumab) in HIV



- 1 clinical trial phase I with ipilimumab and nivolumab in HIV and cancers
- 1 clinical trial with pembrolizumab in HIV w/ disseminated relapsing cancers

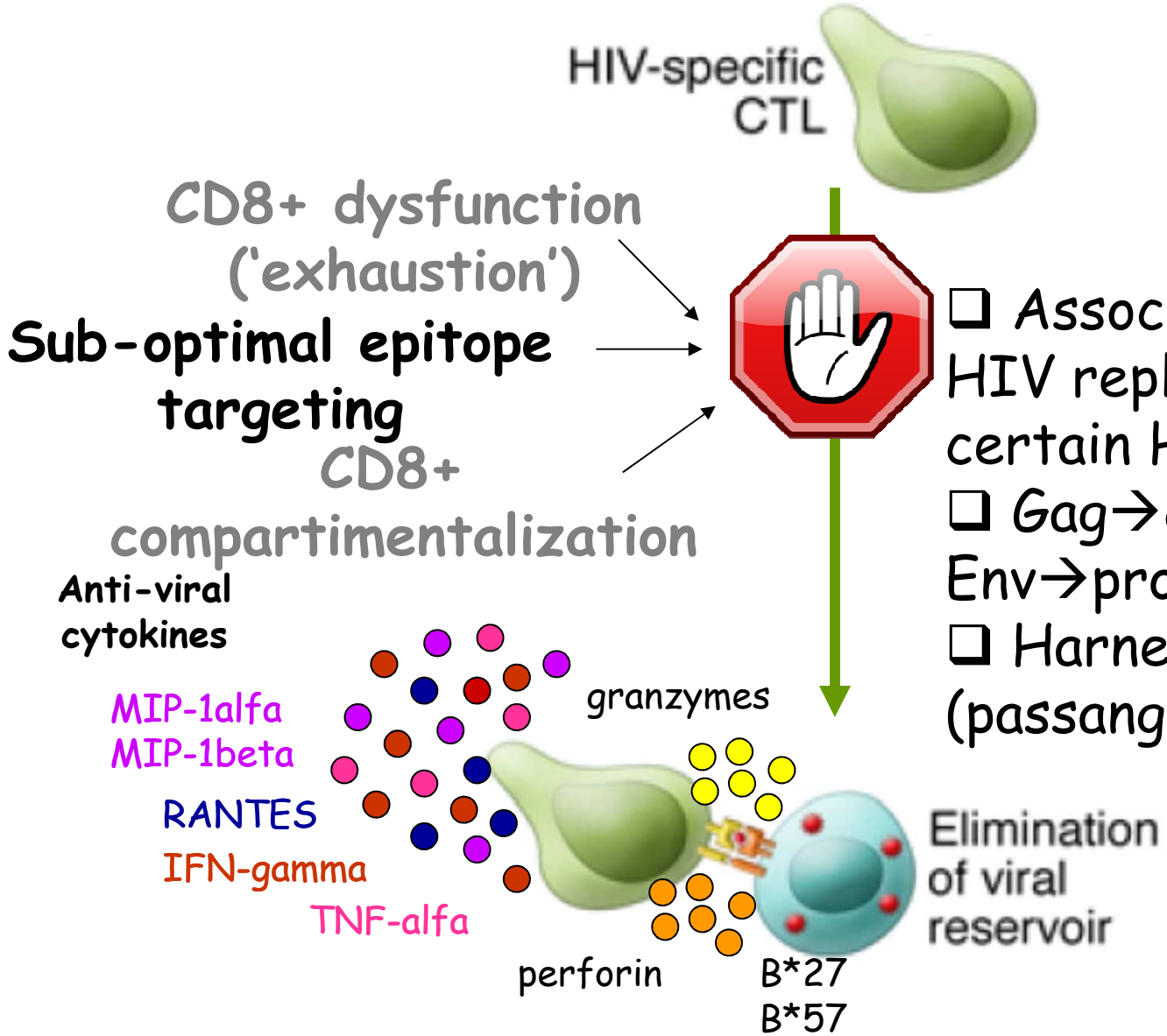
Mechanisms of latency in HIV infection



- ❑ Exclusion from linfatic follicles
- ❑ Immunopriviledged sites (testis, CNS)
- ❑ Tissue-resident memory T cells (Trm) ?

Mod from RB Jones and BD Wlaker, JCI doi:10.1172/JCI80566

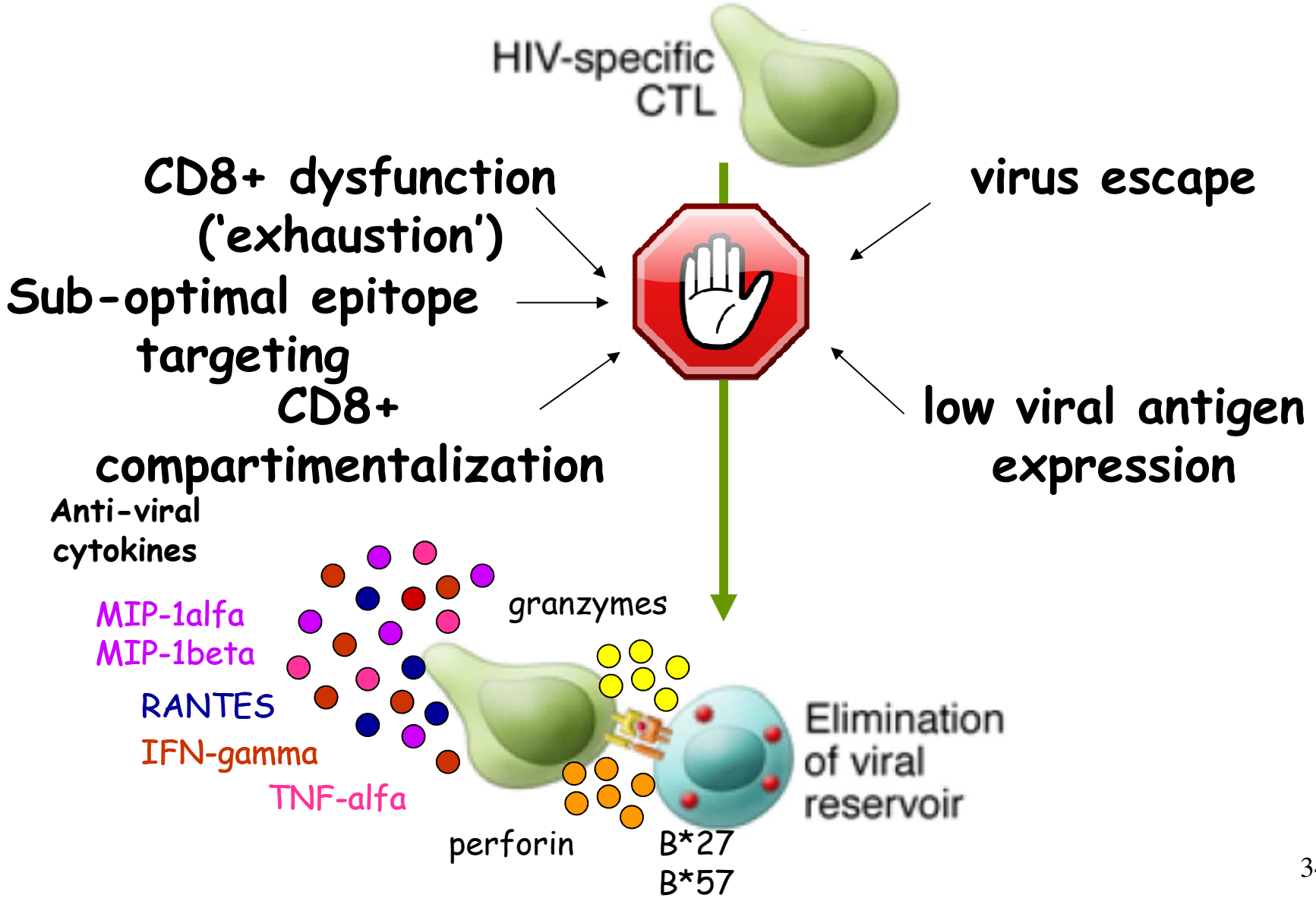
Mechanisms of latency in HIV infection



- Association between HIV replication and certain HLA class I alleles
- Gag→control, Env→progression
- Harness CD8+ T cells (passangers vs drivers)

Mod from RB Jones and BD Wlaker, JCI doi:10.1172/JCI80566

Mechanisms of latency in HIV infection



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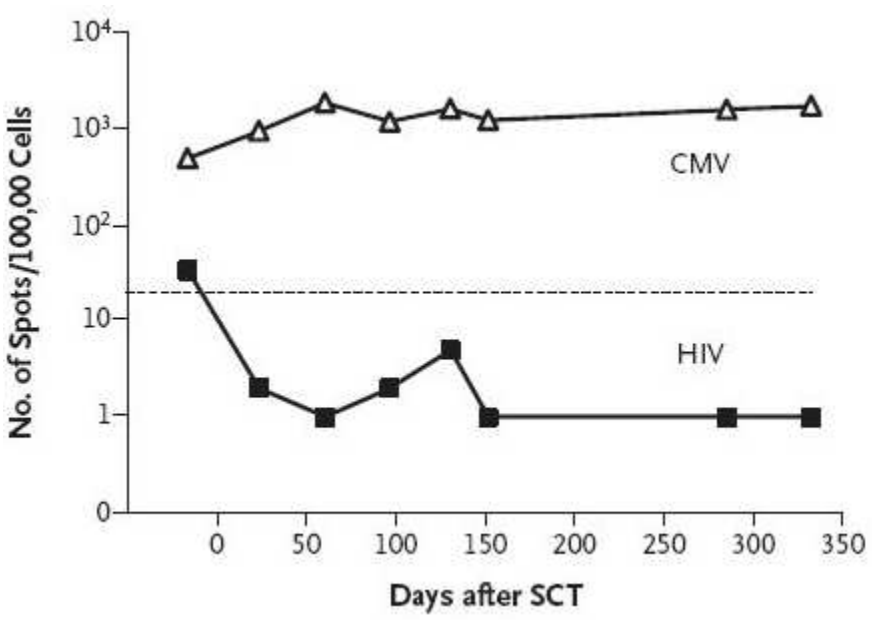
The "real-life" experiments of HIV eradication: the Berlin patient

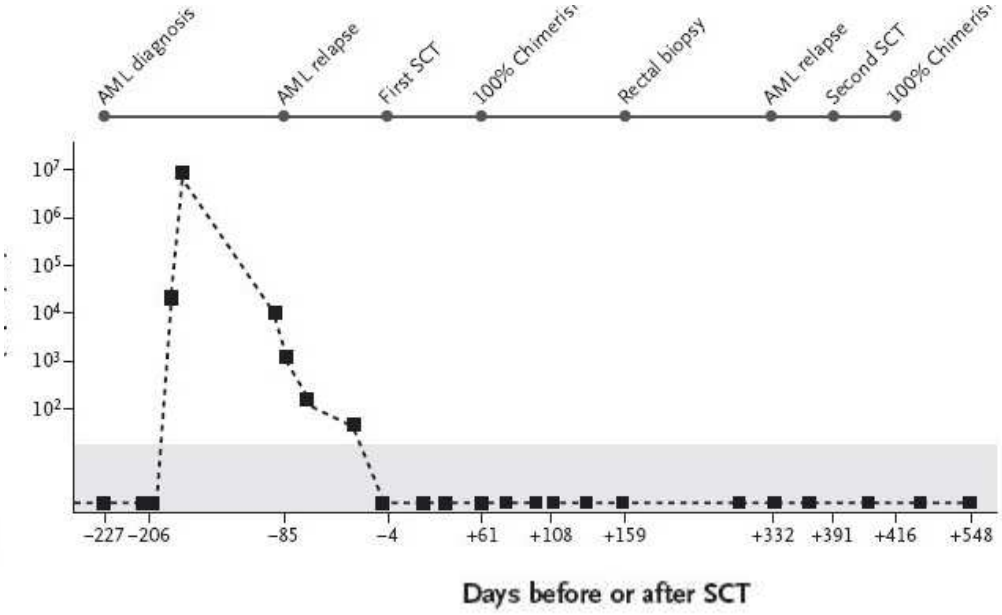
BRIEF REPORT

The NEW ENGLAND JOURNAL of MEDICINE

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

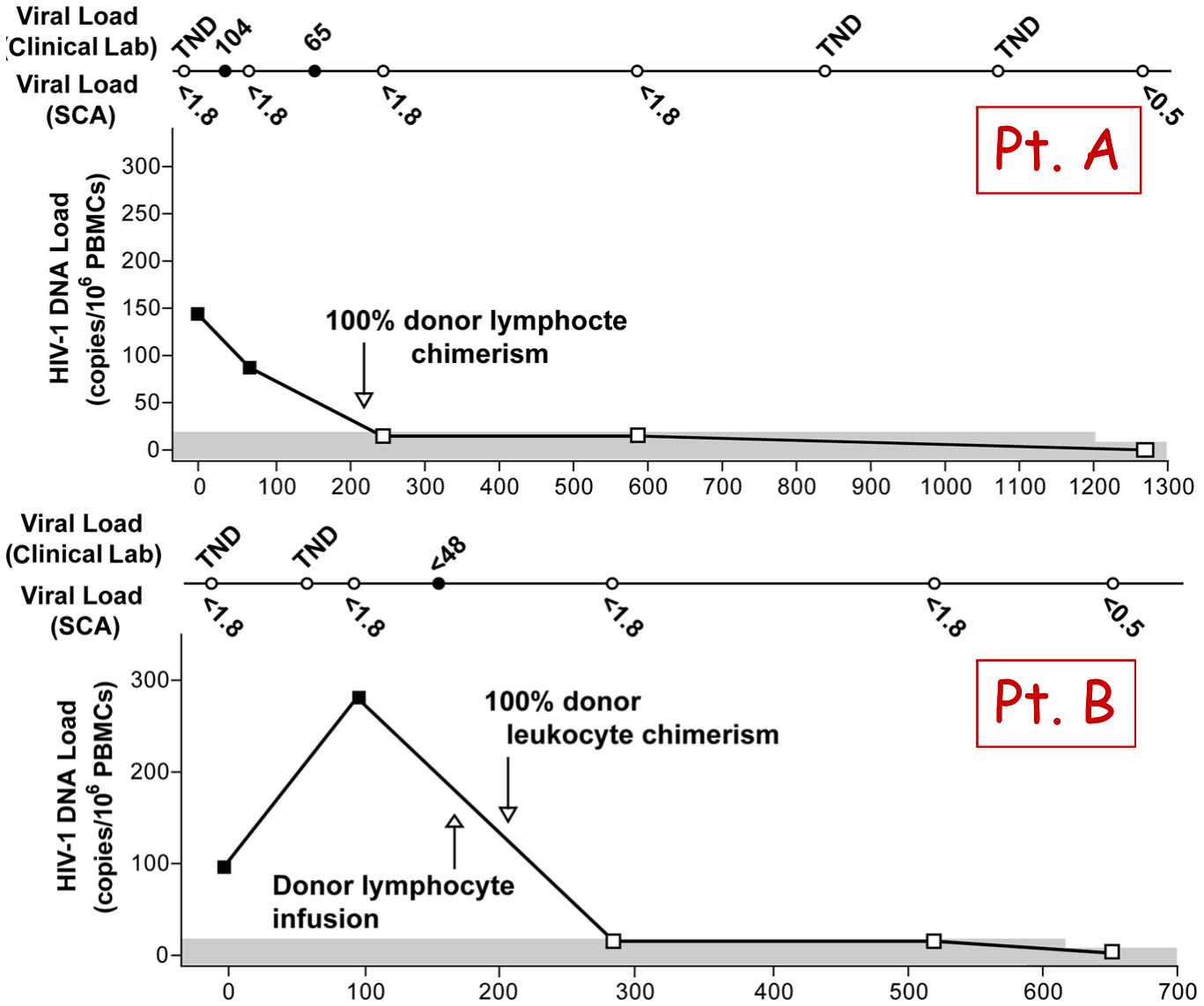
Gero Hütter, M.D., Daniel Nowak, M.D., Maximilian Mossner, B.S., Susanne Ganepola, M.D., Arne Müßig, M.D., Kristina Allers, Ph.D., Thomas Schneider, M.D., Ph.D., Jörg Hofmann, Ph.D., Claudia Kücherer, M.D., Olga Blau, M.D., Igor W. Blau, M.D., Wolf K. Hofmann, M.D., and Eckhard Thiel, M.D.





free of cancer and HIV-1 after > 5 yrs

The "real-life" experiments of HIV reservoirs reduction: the Boston patients



Henrich TJ et al, J Infect Dis 2013. 207:1694-702

The Mississippi child example

THE HUFFINGTON POST

975x96

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HIV-positive mother (no treatment during pregnancy)

child started treatment by the time she was 30 hours old

antiretroviral therapy stopped at 18 months

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remained off the drugs for 27 months
with no signs of the virus in the blood

HIV re-emerges in 'cured' Mississippi girl

10 July 2014 | Health

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Novel approaches for eradication of HIV infection

- **Shock and kill**

LRAs (i.e. histone deacetylase inhibitors [HDACIs] + cytokines + TLR agonists (or others) + CD8+ T cells [or other immune effectors])

→ induce antigen expression from quiescent cells and eliminate exposed targets

- **Therapeutic immunization**

In vitro short-term expansion of CD8+ T cell lines with HIV antigens or de novo priming of novel HIV-specific T cells or DC vaccines

→ increased CTL-mediated killing of HIV-infected cells

- **Cell therapy**

In vitro ex vivo expansion and reinfusion of virus-specific CTLs (+ homing reagents) or transgenic HIV-specific TCRs or chimeric antigen receptors (CARs)

→ increased CTL-mediated killing of HIV-infected cells

Novel approaches for eradication of HIV infection

- **Co-inhibitory blockade**

ART+ mAbs against PD1 or CTLA4

→ enhance the abilities of CD8+ T cells to clear persistent viral reservoirs

- **Gene editing**

Zinc finger nucleases and transcription activator-like effector nucleases, CRISPR/Cas9-mediated strategies

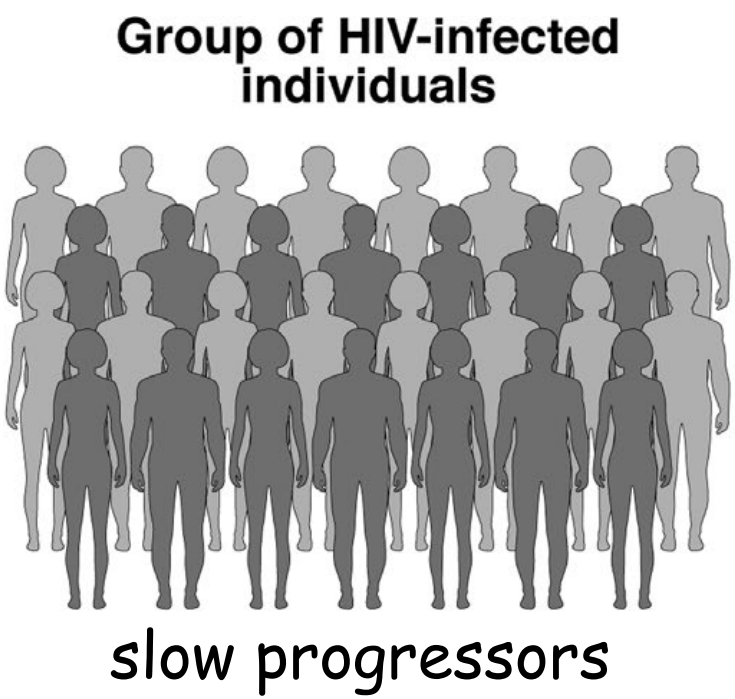
→ disrupt CCR5, Block of pre-integrated proviral dsDNA, cleavage or reactivation of latent provirus

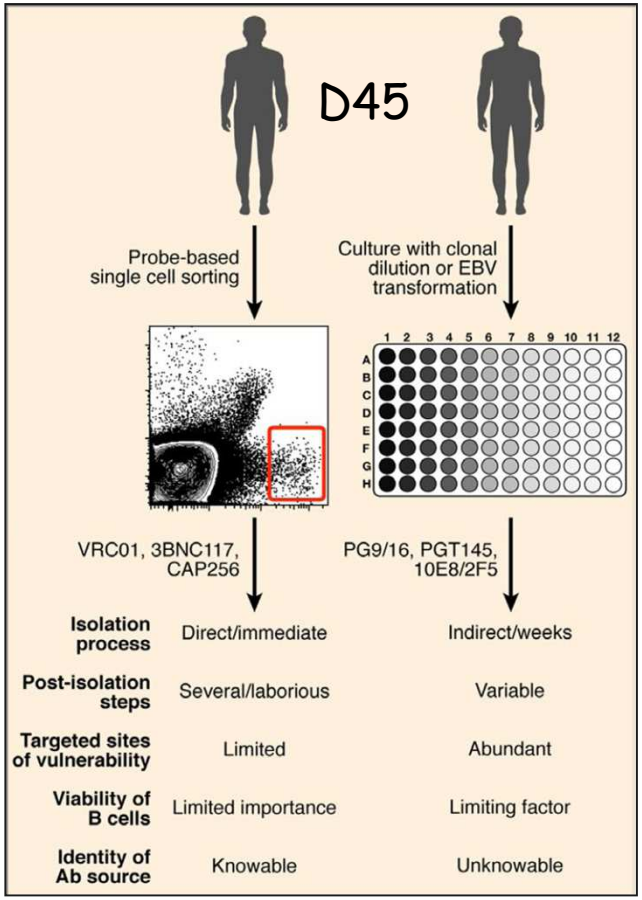
- **Additional immunotherapeutics**

cART+IL-15, cART+IL-15 superagonists (ALT-803), cART+TLR2 agonists, cART+ agonistic anti-CD40 mAbs

→ reverse blocking mechanisms

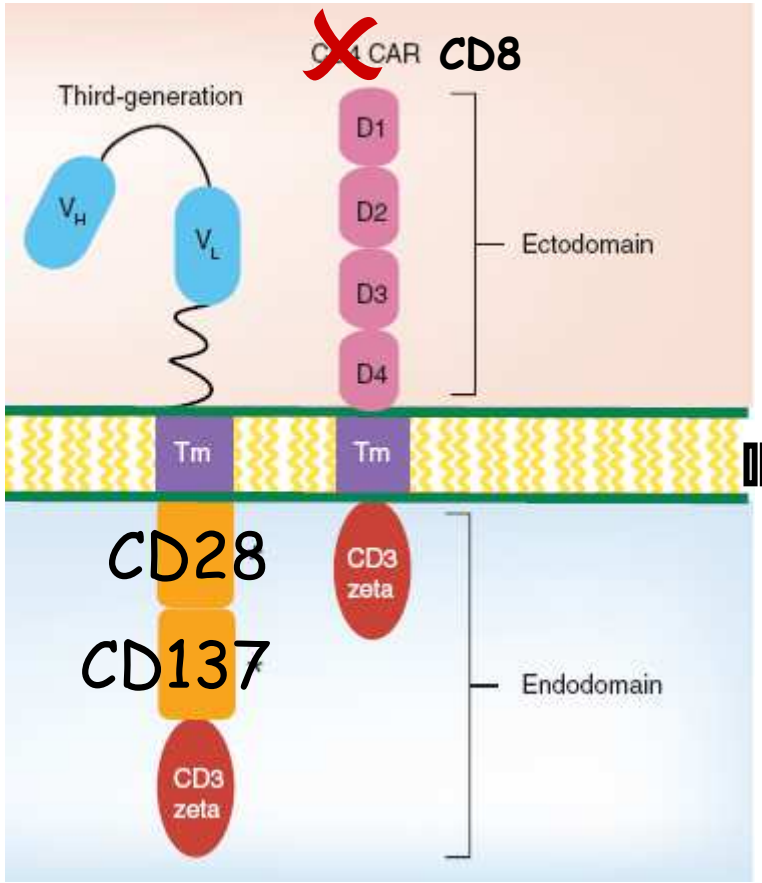
Alteration in B cells and passive immunization with bnAbs





Imm Rev 2017; 275: 33–48

CAR-T CD8 carrying VRC01 bnAb may exert potent antiviral activities



effectively induced cytolysis of reactivated latently infected CD4+ T cells isolated from infected individuals

Long time lasting ?
Immune escape ?
Viral escape ?
Generating HIV-resistance ?

Immunotherapy (2017) 9(5), 401–410

The interplay between HIV and the immune system

HIV

Gp120/CD4
CCR5/CXCR4 ↓ Inverse transcriptase

CD4+ cells

No ART

↓CD4+ cells
↑CD8+ cells

Immune activation

Latency

ART

Persistently infected memory CD4+ cells

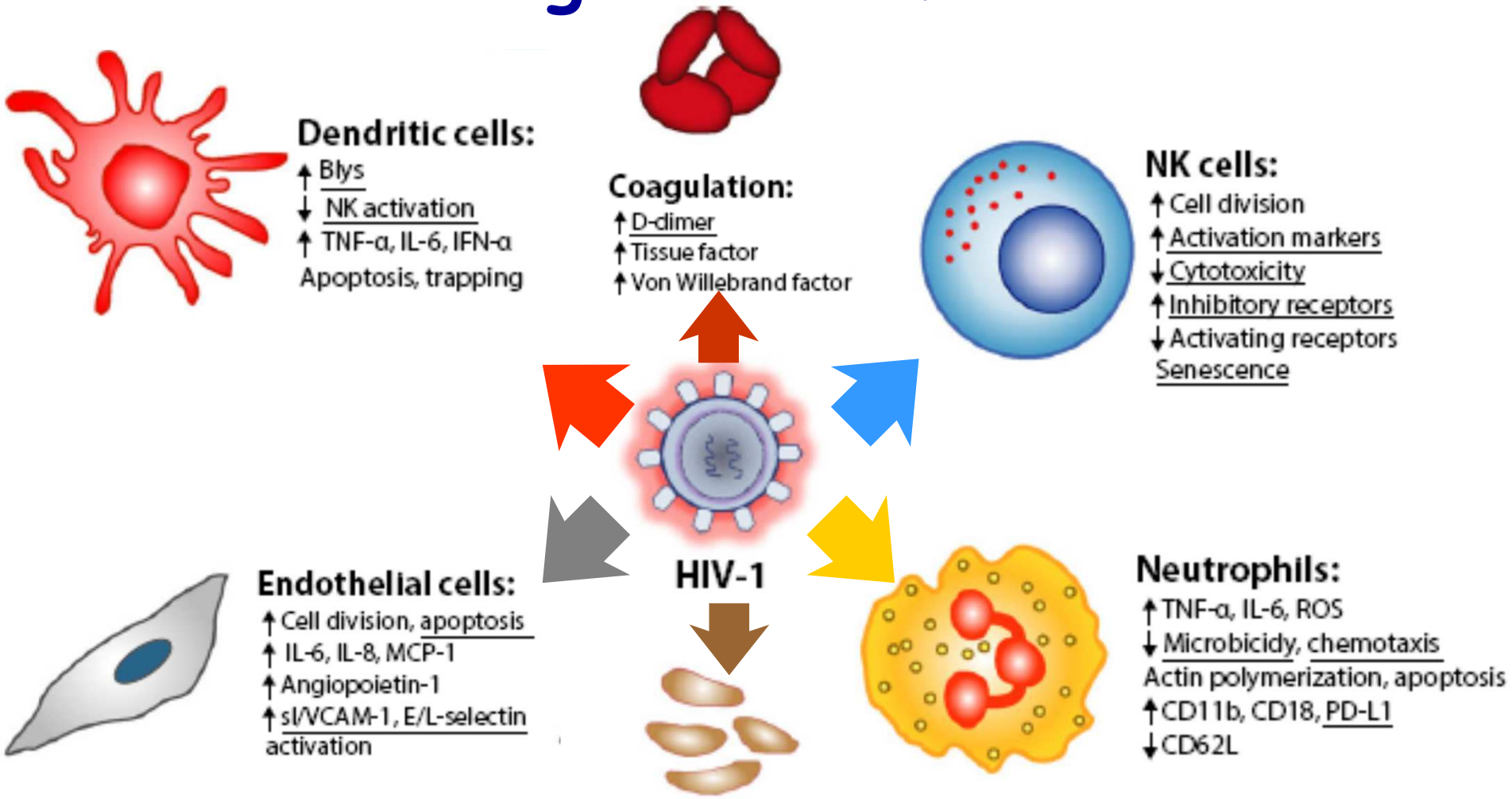
Viral reservoir(s)
Abnormalities in immune system

Autoimmunity in AIDS

- **Excess of humoral immunity**
  - B lymphocyte proliferation and B cell lymphoma
  - Autoantibodies produced by expanded B lymphocytes
- **Altered Tregs**
  - Low Treg counts (CD4+CD25hi+Foxp3+)
  - Decreased IL-2 levels in addition to IL-10 and TGF-beta
  - Decreased TLR4 expression
- **Elevated cytotoxic T cells**
- **Improvement of HIV infection with solid organ and SC transplantation**

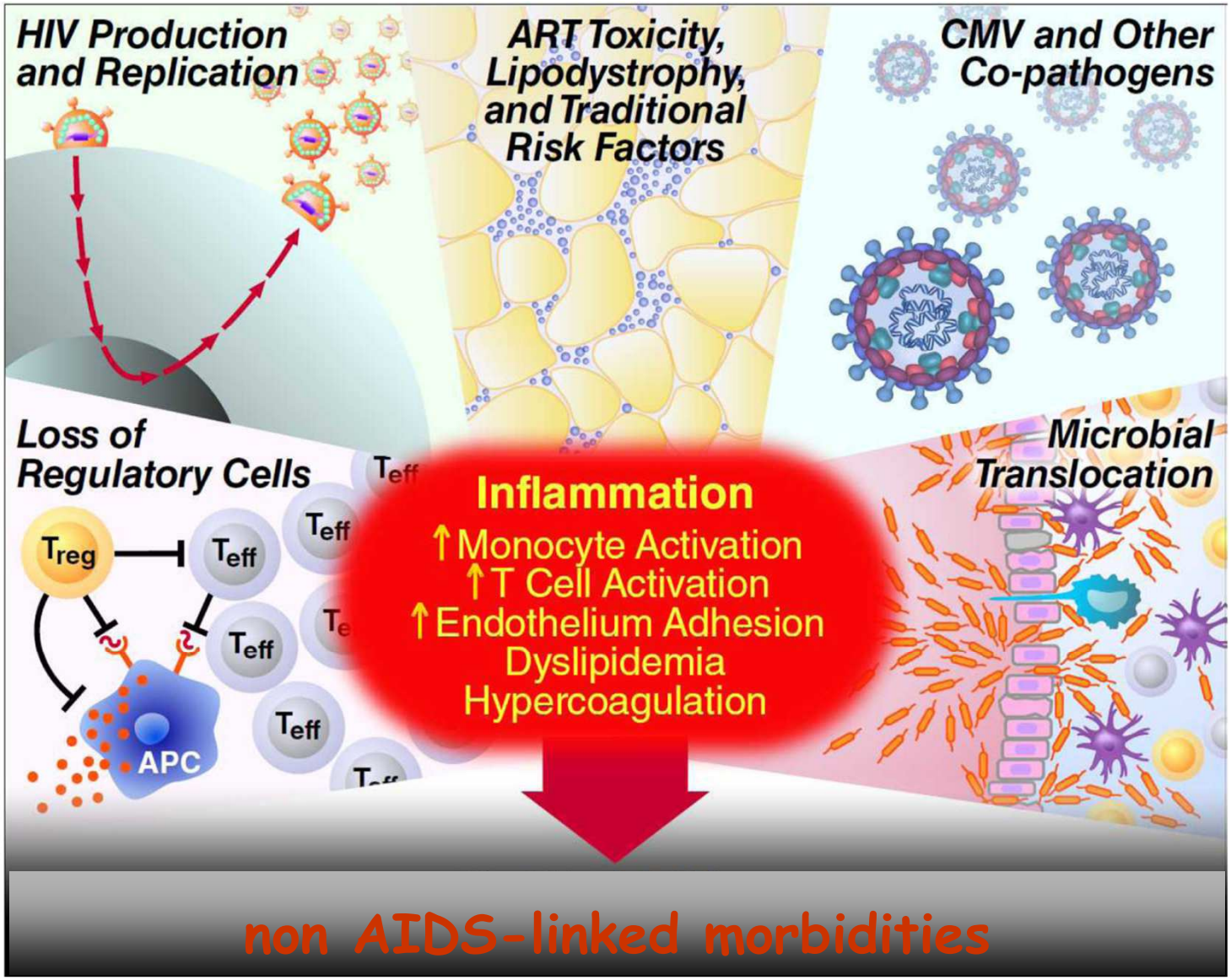
Autoimmune disorders
(SLE, PM, ITP, arthritis, APS, Grave's)

Persistence of immune activation through HIV infection



HIV Medicine (2016), 17, 89–105

The End of AIDS ?
HIV Infection as a Chronic Disease

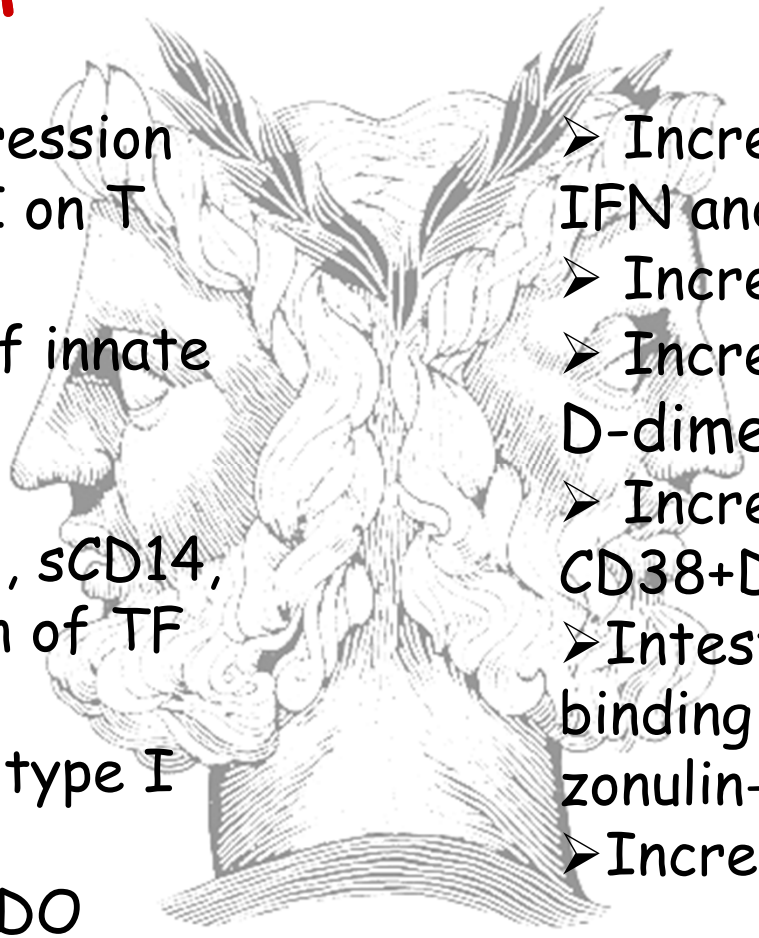


Lancet. 2013 November 2; 382(9903): 1525–1533.

Persistence of immune activation through HIV infection

HIV infection

- CD38 and DR expression with increased MIFI on T cells
- Soluble markers of innate immune activation (neopterin, β2-MG)
- Increased sCD163, sCD14, increased expression of TF on monocytes
- High dysregulated type I IFN production
- Dysregulation in IDO
- Uncertain role of Th17
- Increased TNF



ART therapy

- Increased levels of type I IFN and CXCL10
- Increased sCD163
- Increased IL-6, hsCRP, D-dimer, sTNFR-1
- Increased CD38+DR+CD8+ cells
- Intestinal-fatty acid binding protein (I-FABP) and zonulin-1
- Increased TNF

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Membrane tumour necrosis factor-α is involved in the polyclonal B-cell activation induced by HIV-infected human T cells

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INFECTION of CD4$^+$ T cells by human immune deficiency virus-1 (HIV-1) causes severe dysfunction of cellular immunity[1–3], but paradoxically results in intense polyclonal activation of B cells, possibly accounting for both hypergammaglobulinaemia and frequent development of B-cell malignancies seen in HIV-infected patients[4–7]. We have reported that human CD4$^+$ T-cell clones infected with HIV *in vitro* markedly stimulate immunoglobulin synthesis by B cells through a non-cognate, contact-dependent

T cell clone	TNF-α RNA expression		TNF-α secretion (U ml^{-1})	
	HIV$^-$	HIV$^+$	HIV$^-$	HIV$^+$
TT52			<50 (874)	518 (2,640)
TT 7			<50 (1,066)	638 (7,638)
TT8			<50 (130)	250 (1,840)

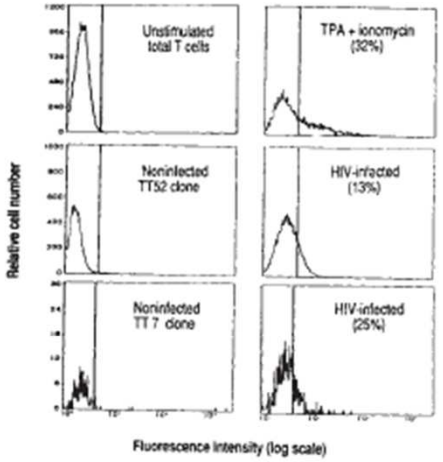
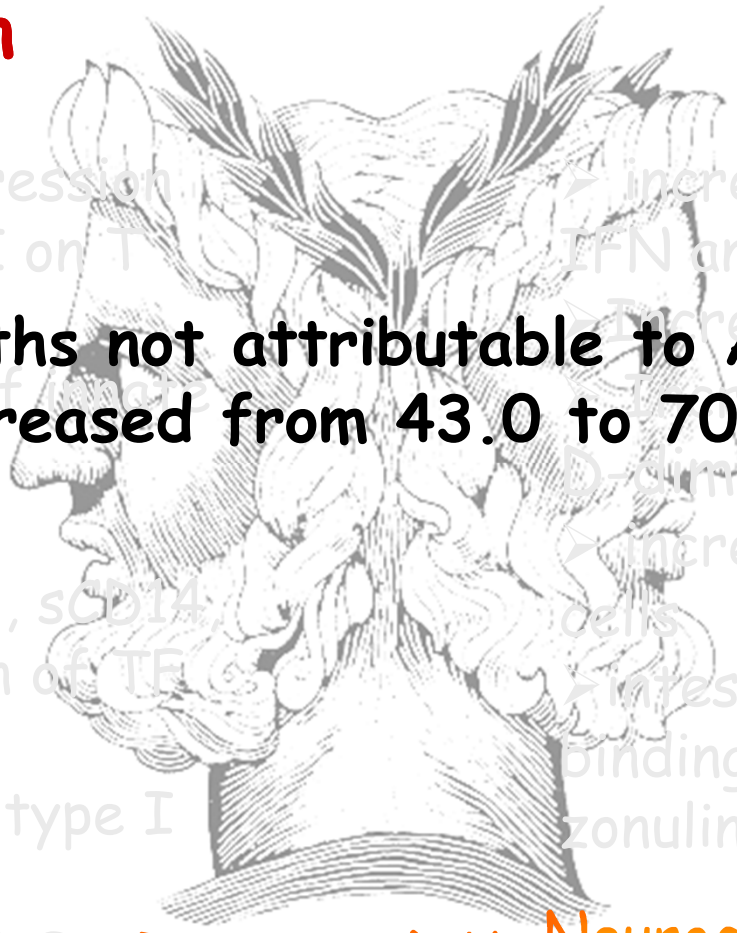


FIG. 1 TNF-α secretion and expression of membrane TNF-α by HIV-infected T-cell clones. CD4$^+$ T-cell clones specific for tetanus toxoid or recombinant Poa pratensis group IX (Poa p IX) allergen were generated from peripheral blood lymphocytes (PBL) of HIV-seronegative donors by limiting-dilution cloning as described[R,16]. T blasts from each clone were then divided into two equal aliquots and co-cultured with irradiated (6,000r) HIV-infected or non-infected PBL, respectively. Co-culturing was done in the presence of phytohaemagglutinin (PHA) (1% v/v), interleukin-2 (20 u ml^{-1}) and polybrene (5 μg ml^{-1})[R]. On day 14, HIV infection of T-cell clones was assessed by

Persistence of immune activation through HIV infection

HIV infection **ART therapy**



Deaths not attributable to AIDS increased from 43.0 to 70.5%

mortality CAD Type II DM Neurocognitive dysfunction end-organ disease

Conclusions

- De-regulated immuno-metabolism represents a central element to the biased immunity against HIV-1 infection that leads to viral dissemination and pathogenesis
- In spite of a full virological response to treatment, immune activation often persists as well and may impair the immune recovery and favour non AIDS-linked morbidities
- From early interventions characterized by high toxicity and lack of efficacy, the ultimate goal of a durable ART-free virologic remission have not achieved but ongoing trials are aimed to perturb the reservoir
- No human vaccine trial conducted to date has elicited high titer broadly neutralizing antibody responses

Conclusions

- De-regulated immuno-metabolism represents a central element to the biased immunity against HIV-1 infection that leads to viral dissemination and pathogenesis

- In spite of a full virological response to treatment, immune activation often persists as well and may impair the immune recc

- Fror ty and lack T-free viro als are aimed to perturb the reservoir

Now this is not the end. It is not even the beginning of the end. But it is, perhaps, the end of the beginning.

- No human vaccine trial conducted to date has elicited high titer broadly neutralizing antibody responses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