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Comprehensive Cancer Center

Viral infections in stem cell transplantation:

Lessons learned from 20 years of single-cell research in CMV immunology

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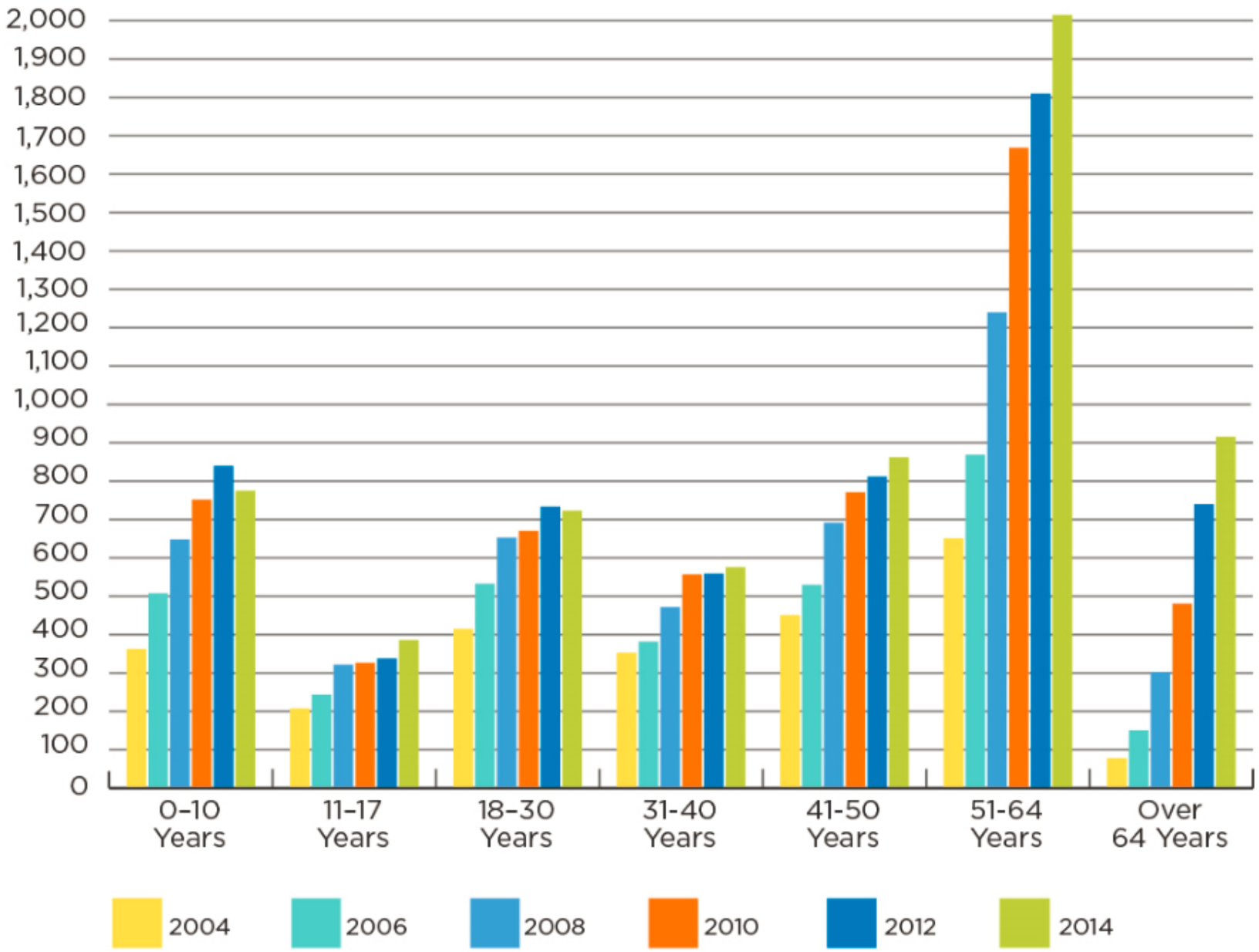
Transplants by Recipient Age



Source: National Marrow Donor Program/Be The Match FY 2014

NATIONAL MARROW DONOR PROGRAM®

BE THE MATCH®

The evolution of allogeneic SCT

Over the past 15 years:

- A major goal is to maximize T cell effects--*less* intense chemotherapy is often used
- Older patients are commonly transplanted (to 75 vs. 60)
- Peripheral blood (vs. marrow) is commonly used as a stem cell source
- 100-day mortality has decreased dramatically with nonmyeloablative or reduced-intensity conditioning
- More graft sources and donors (NMDP, worldwide registries)

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What hasn't changed much?

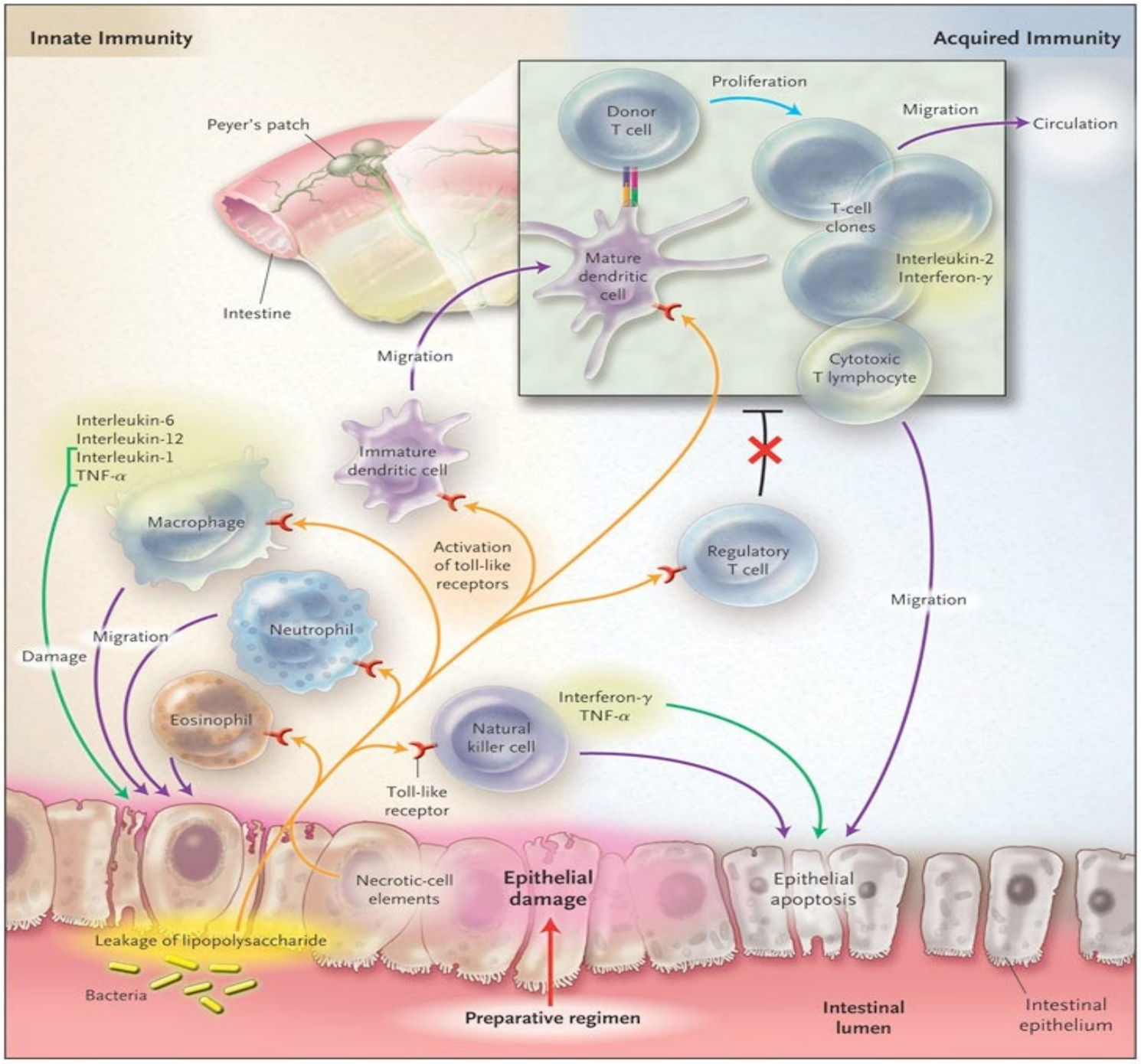
- Risk stratification for most diseases (e.g., acute myeloid leukemia, the primary indication for allo SCT) has evolved very little, despite the 'omics' revolution

- In most cases, we still infuse donor grafts as collected, without enrichment or manipulation of cell subsets

- Strategies to prevent and/or treat GVHD have not significantly evolved in over 25 years, despite many attempts and the advent of targeted and biologic therapies

- Beyond conditioning, no therapies have typically been used to reduce relapse risk

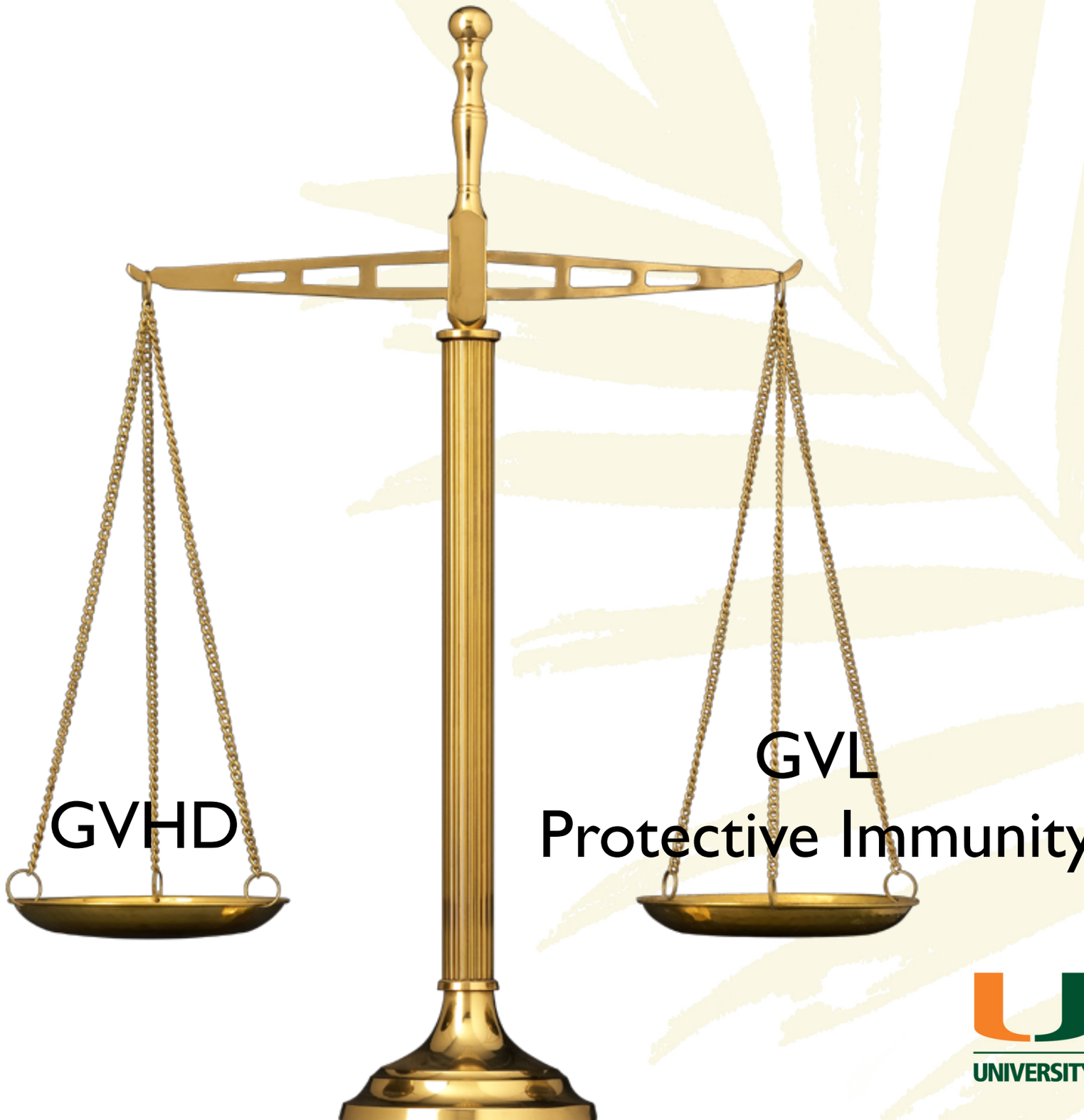
Acute GVHD: Pathophysiology



Copelan E. N Engl J Med 2006;354:1813-1826

The NEW ENGLAND JOURNAL of MEDICINE

GVHD and GVL/protective immunity are in balance



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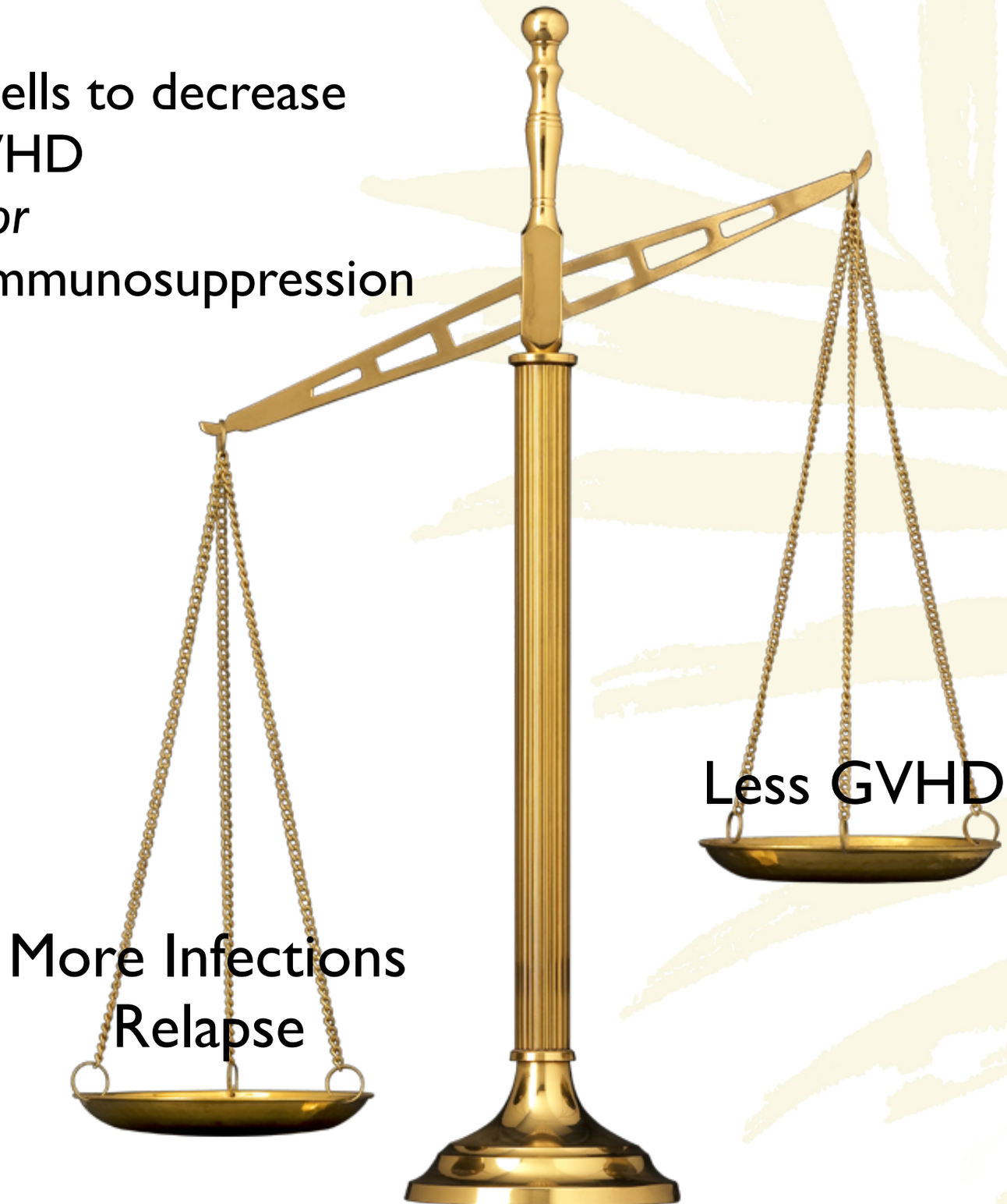
…so nonspecific T cell targeting is ineffective

Removal of T cells to decrease GVHD

or

More aggressive immunosuppression



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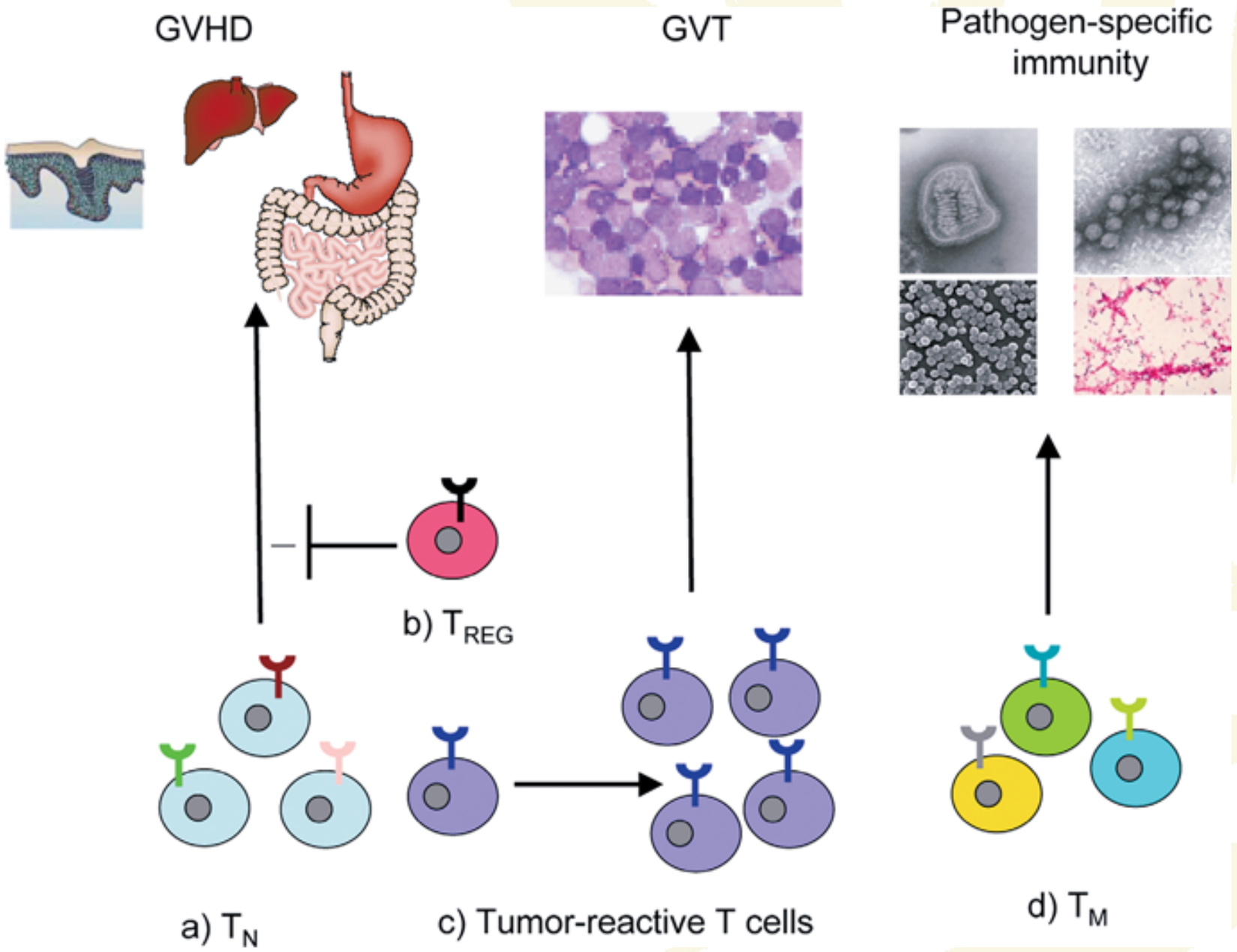
T cells in allogeneic SCT: Good and Bad



Graft-Versus-Host Disease: A Surge of Developments

Stanley R. Riddell, Frederick R. Appelbaum

PLoS MEDICINE
July 2007 | Volume 4 | Issue 7 | e198

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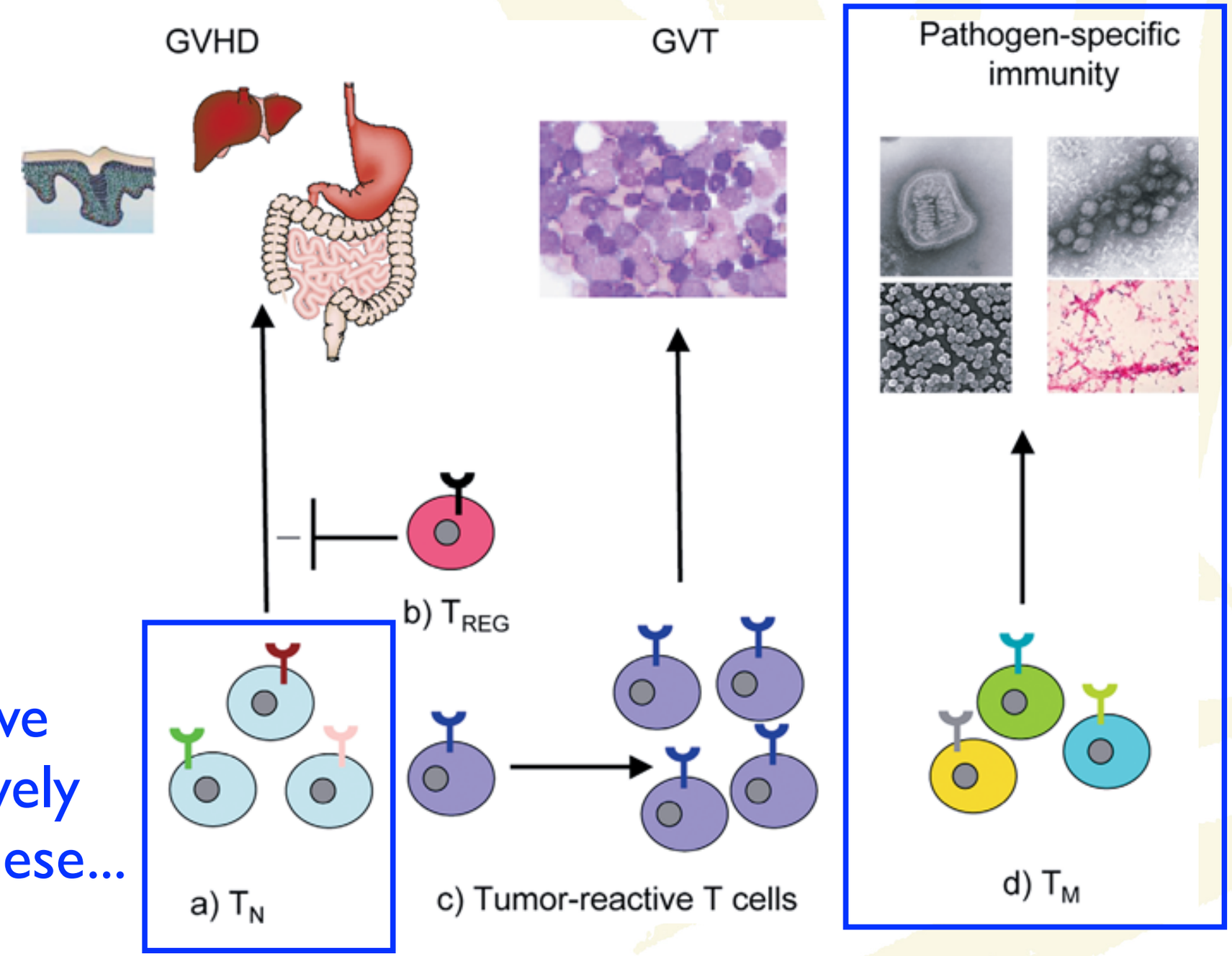
The goal: Eliminating GVHD while sparing beneficial T cells



...but not these?

Can we selectively inhibit these...

Graft-Versus-Host Disease: A Surge of Developments
Stanley R. Riddell, Frederick R. Appelbaum
PLoS MEDICINE
July 2007 | Volume 4 | Issue 7 | e198

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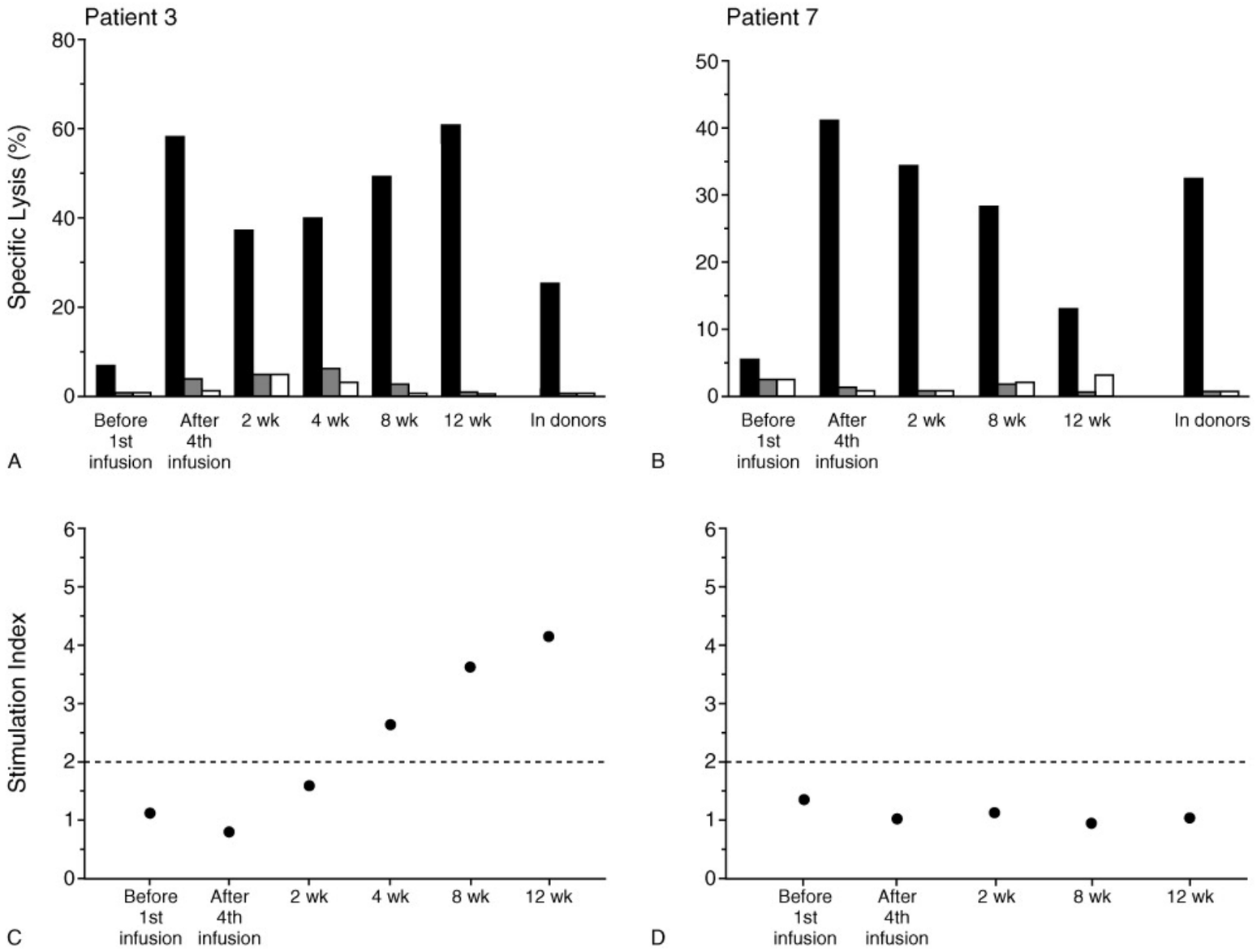
Widely held assumptions about CMV immunity in 1997

- Pathogen-specific T cell responses could not rise above frequencies of 0.5-1% of the overall repertoire
- CMV reactivation most likely occurred in the absence of pathogen-specific T cells
- Steroids are lympholytic and therefore eliminate CMV-specific T cells from the circulation

20 years ago T cell measurements were semi-quantitative



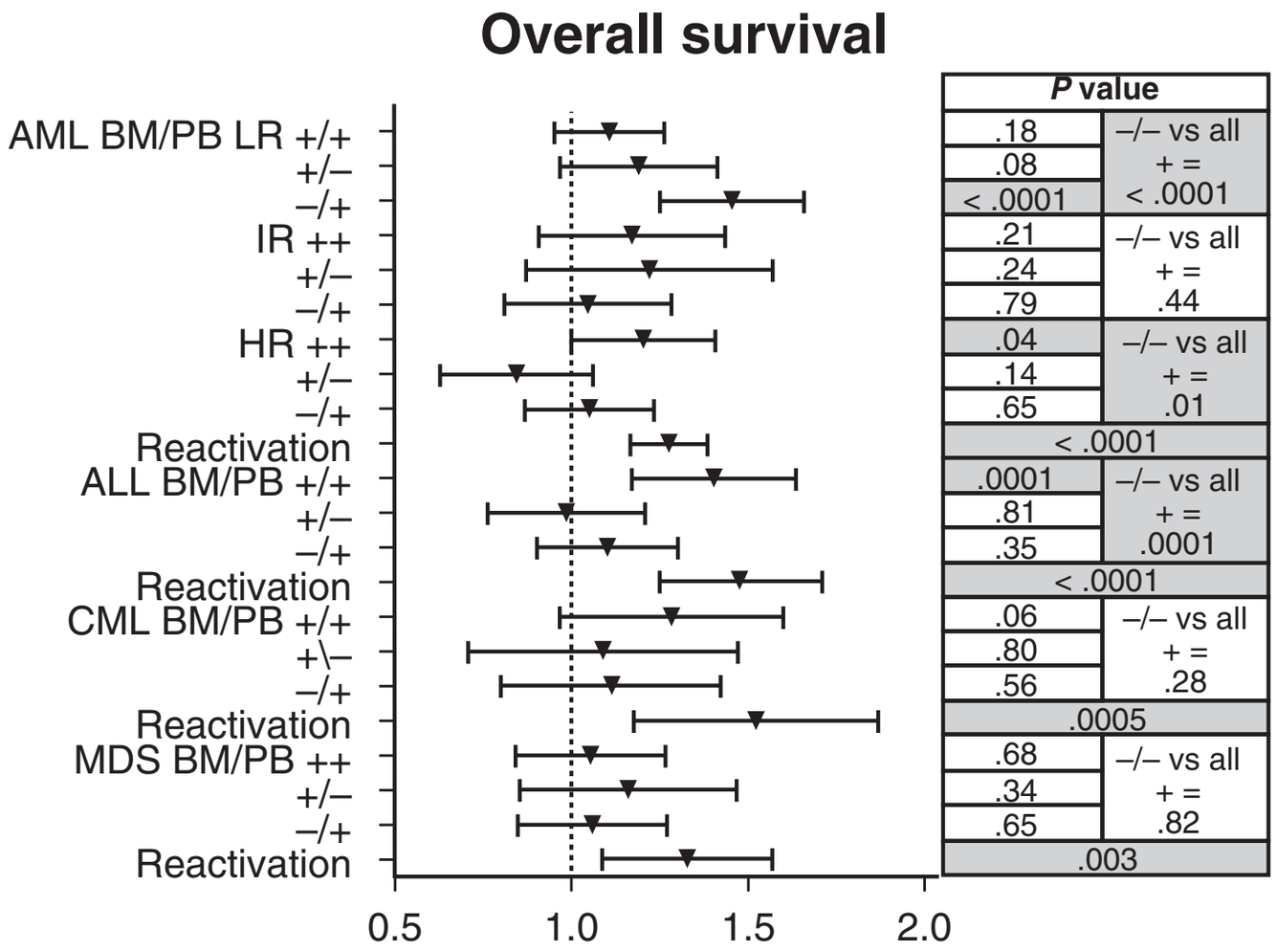
Walter, Riddell, et al N Engl J Med 1995;333:1038-1044.

CMV reactivation still matters

2016 CIBMTR Infection Working Committee analysis

Early cytomegalovirus reactivation remains associated with increased transplant-related mortality in the current era: a CIBMTR analysis

Pierre Teira,[1,*] Minoo Battiwalla,[2,*] Muthalagu Ramanathan,[3,*] A. John Barrett,[2,*] Kwang Woo Ahn,[4,5] Min Chen,[4] Jaime S. Green,[6] Ayman Saad,[7] Joseph H. Antin,[8] Bipin N. Savani,[9] Hillard M. Lazarus,[10] Matthew Seftel,[11] Wael Saber,[4] David Marks,[12] Mahmoud Aljurf,[13] Maxim Norkin,[14] John R. Wingard,[14] Caroline A. Lindemans,[15] Michael Boeckh,[16] Marcie L. Riches,[17] and Jeffery J. Auletta[18]

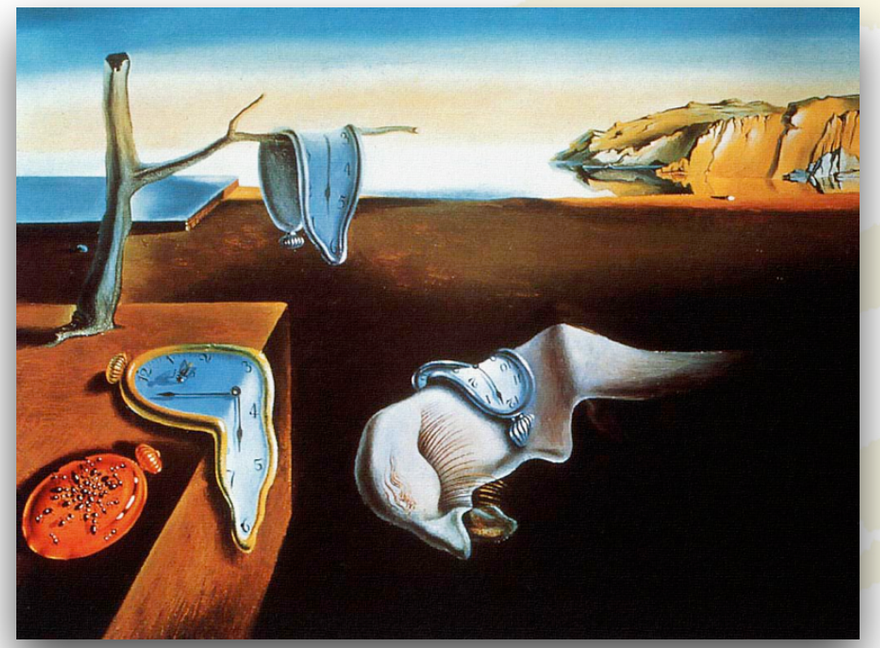


Teira, et al., CIBMTR Infection & Immune Recovery WC, *Blood* 2016

The first description of immunologic memory

"the sick and the dying were tended by the pitying care of those who had recovered, because they knew the course of the disease and were themselves free from apprehensions. For *no one was ever attacked a second time, or not with a fatal result*"

Thucydides, 430 B.C.



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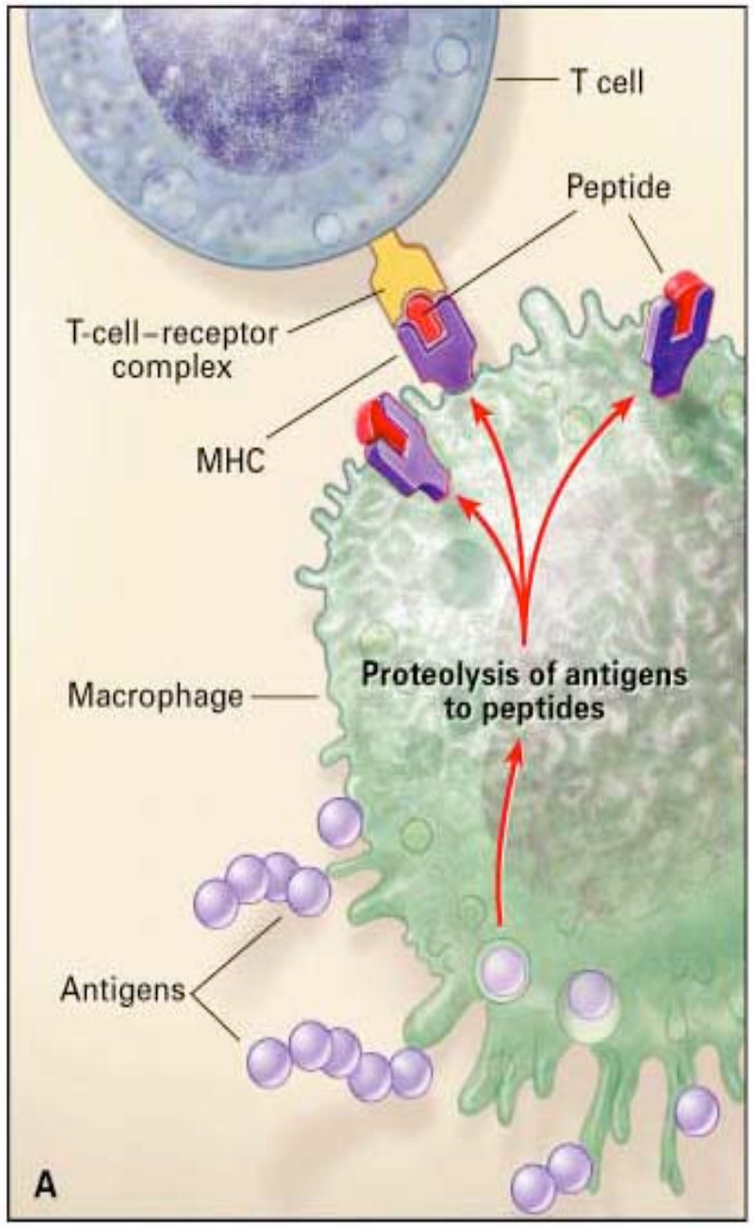
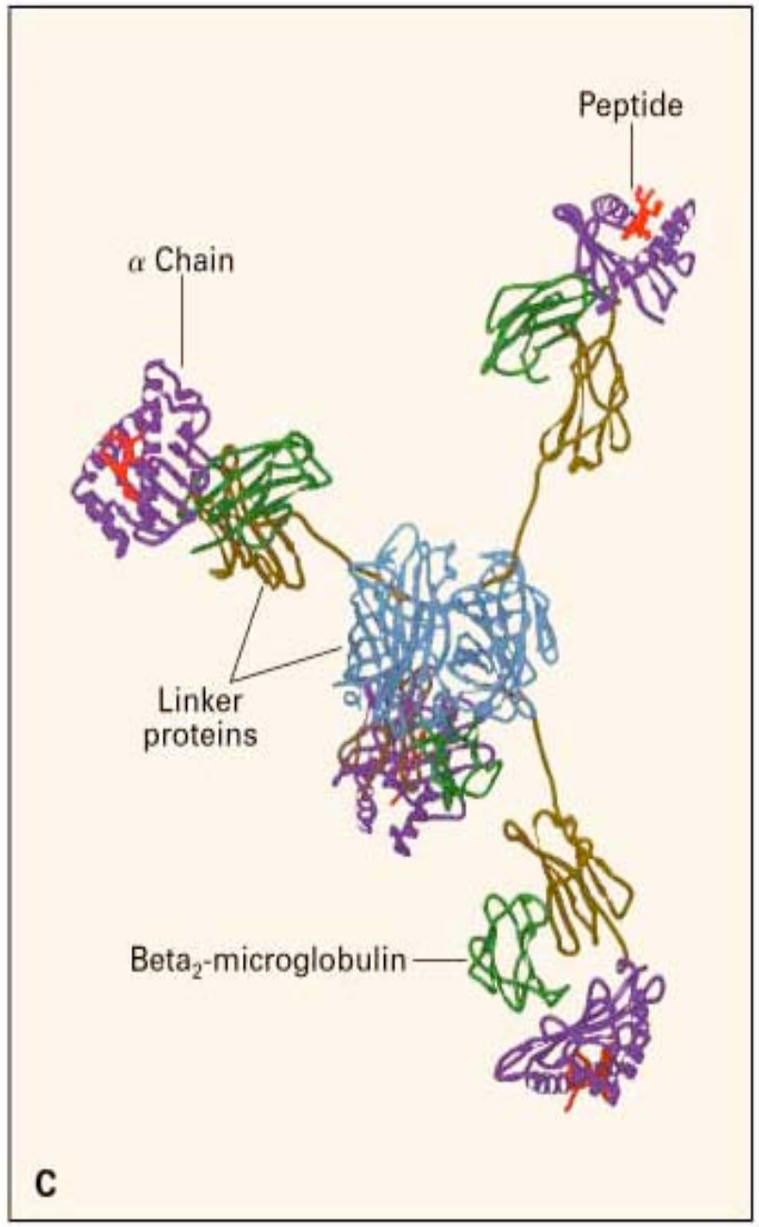
Measurement of Ag-specific CD8+ T cells with tetramers



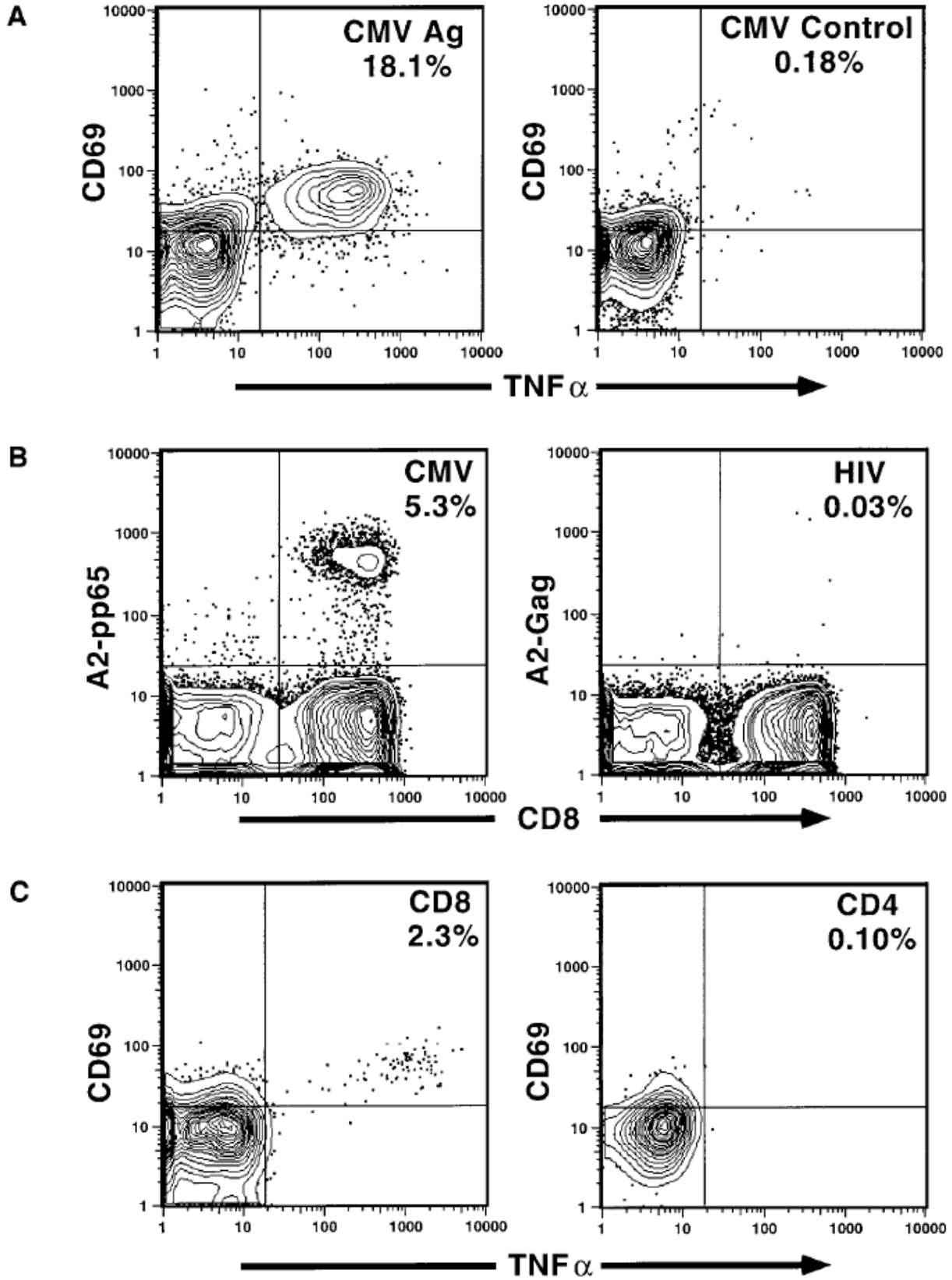


From R. Schwartz, *NEJM* 10/8/98 (original *J.Exp. Med* 187:9, 1998)

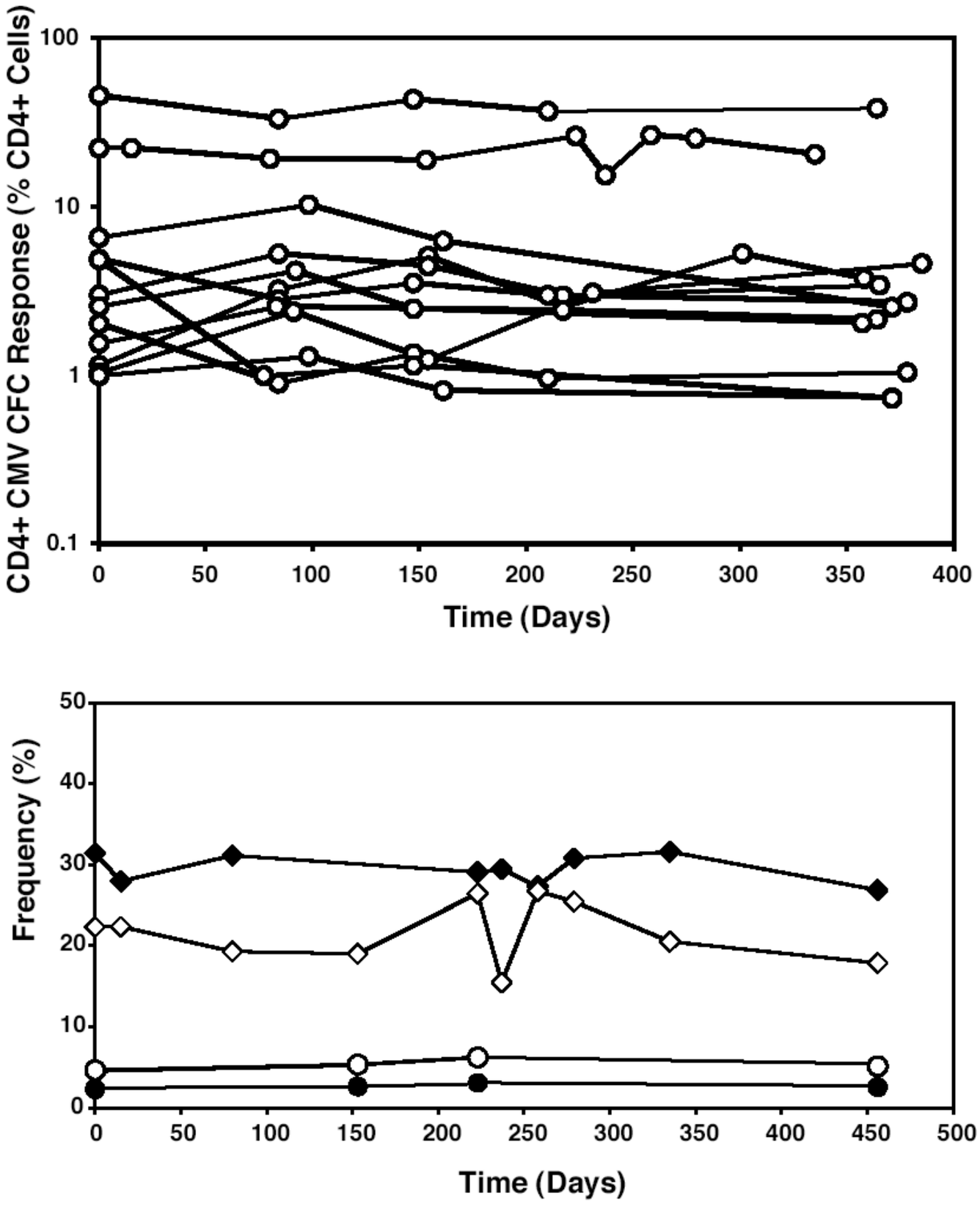
CD4+ CFC
(Functional)

CD8+ Tetramer
(Specificity)

CD8+ CFC
(Functional)



Komanduri, et al., *Virology*, 2001

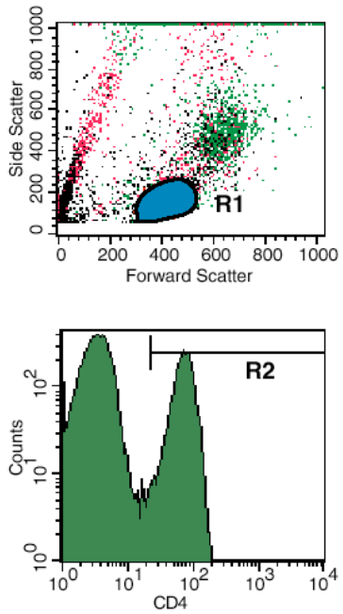


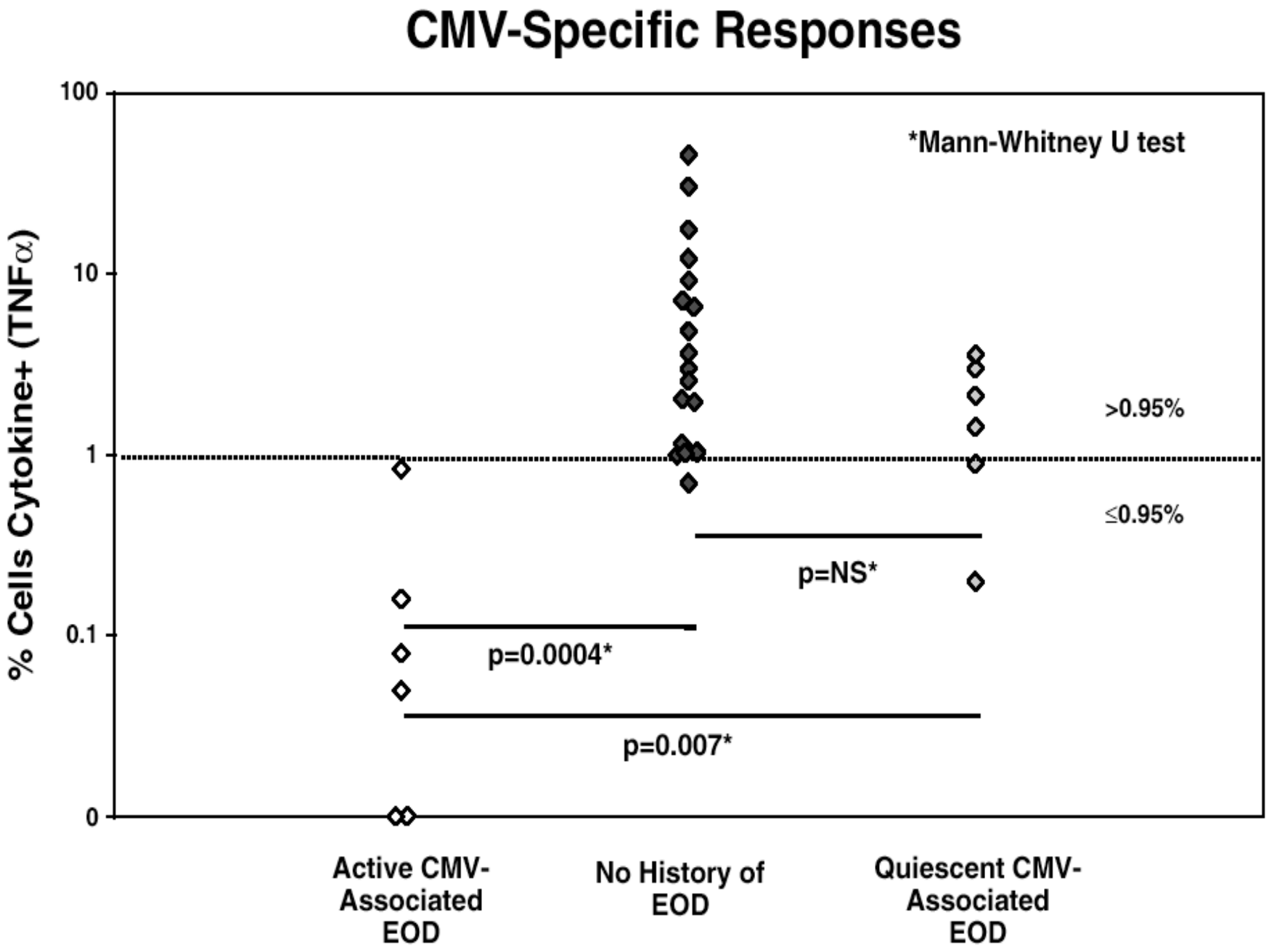
Stability and reproducibility of CD4+ and CD8+ T cell responses to CMV.

Komanduri, et al., *Virology,* 2001

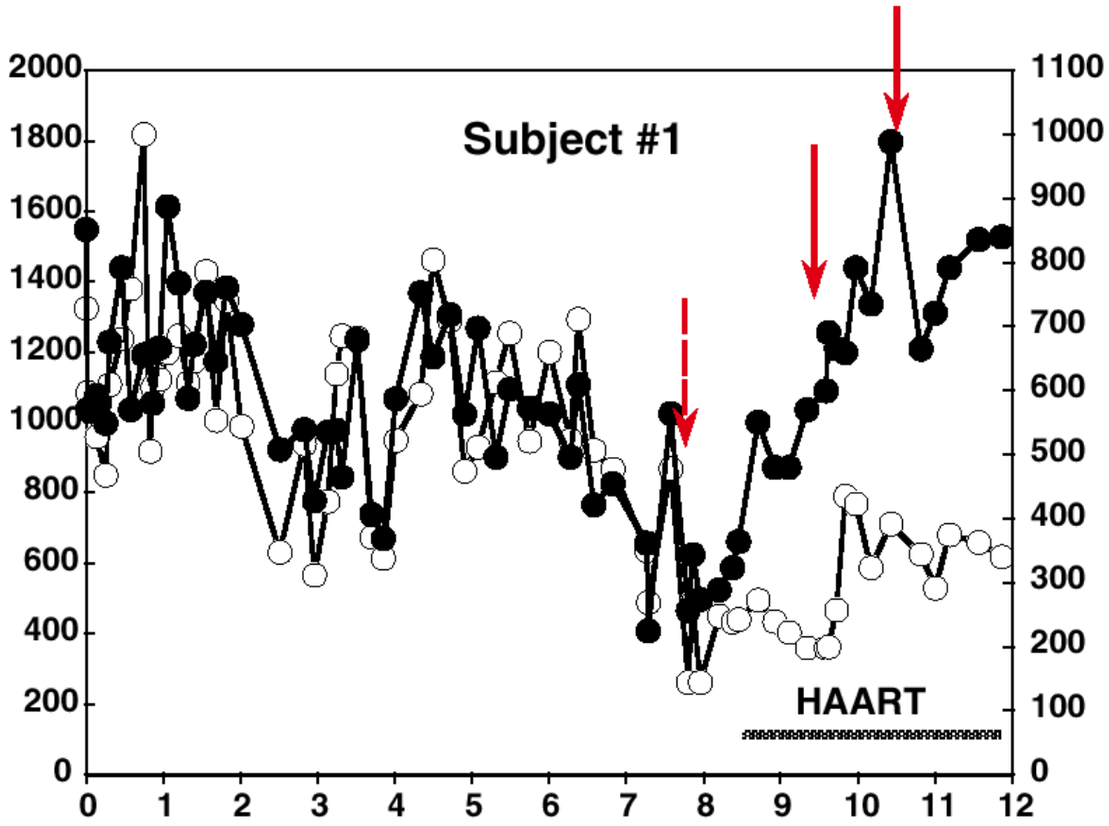
CMV Retinitis in a patient with AIDS

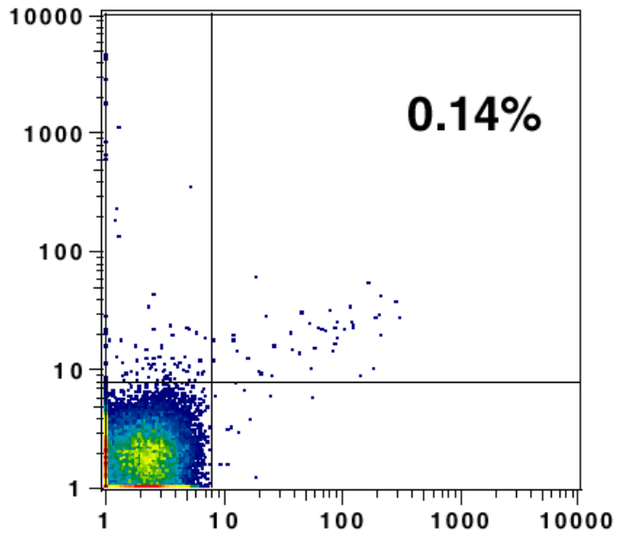
Slightly Less Ancient History

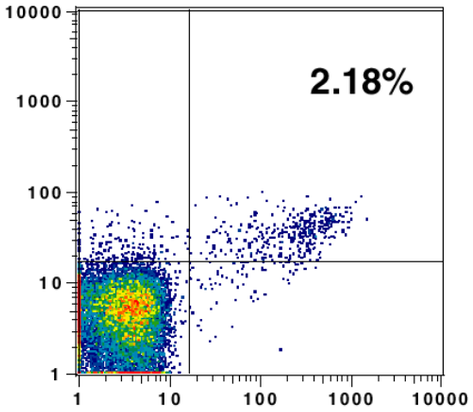


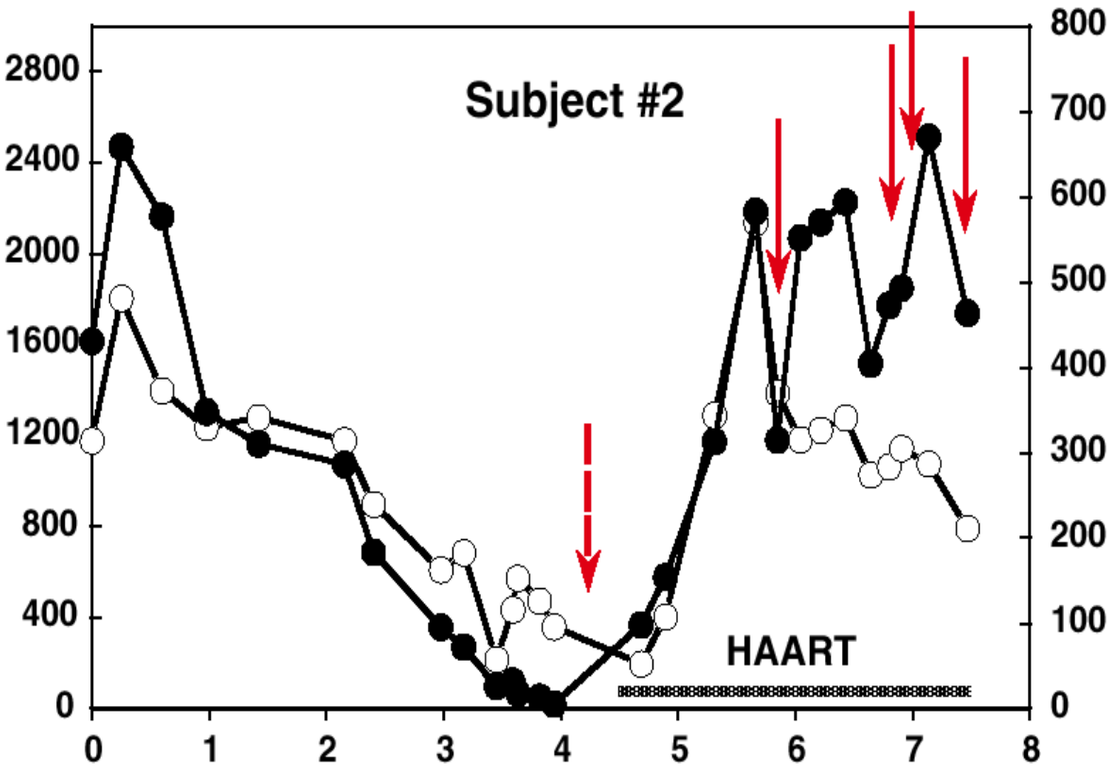


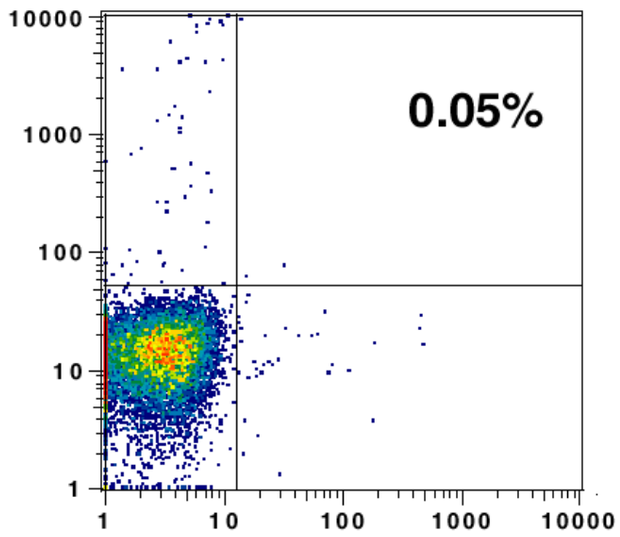
Komanduri, McCune, et al., *Nat Medicine* 4:153-7, 1998











Komanduri, et al., *J Inf Diseases*, 2001

Risk factors for late CMV reactivation in SCT

- 269 subjects transplanted 1998-2000 (alive at day 100)
- 144/269 with early reactivation (54%); 84 with late reactivation (31%)
- 65 of 144 (45%) with early reactivation had late reactivation; 15 of 125 (15%) isolated late
- Multivariate analyses of subjects with early reactivation

Ozdemir, et al, *Bone Marrow Transplantation* 2007

Risk factors for late CMV reactivation

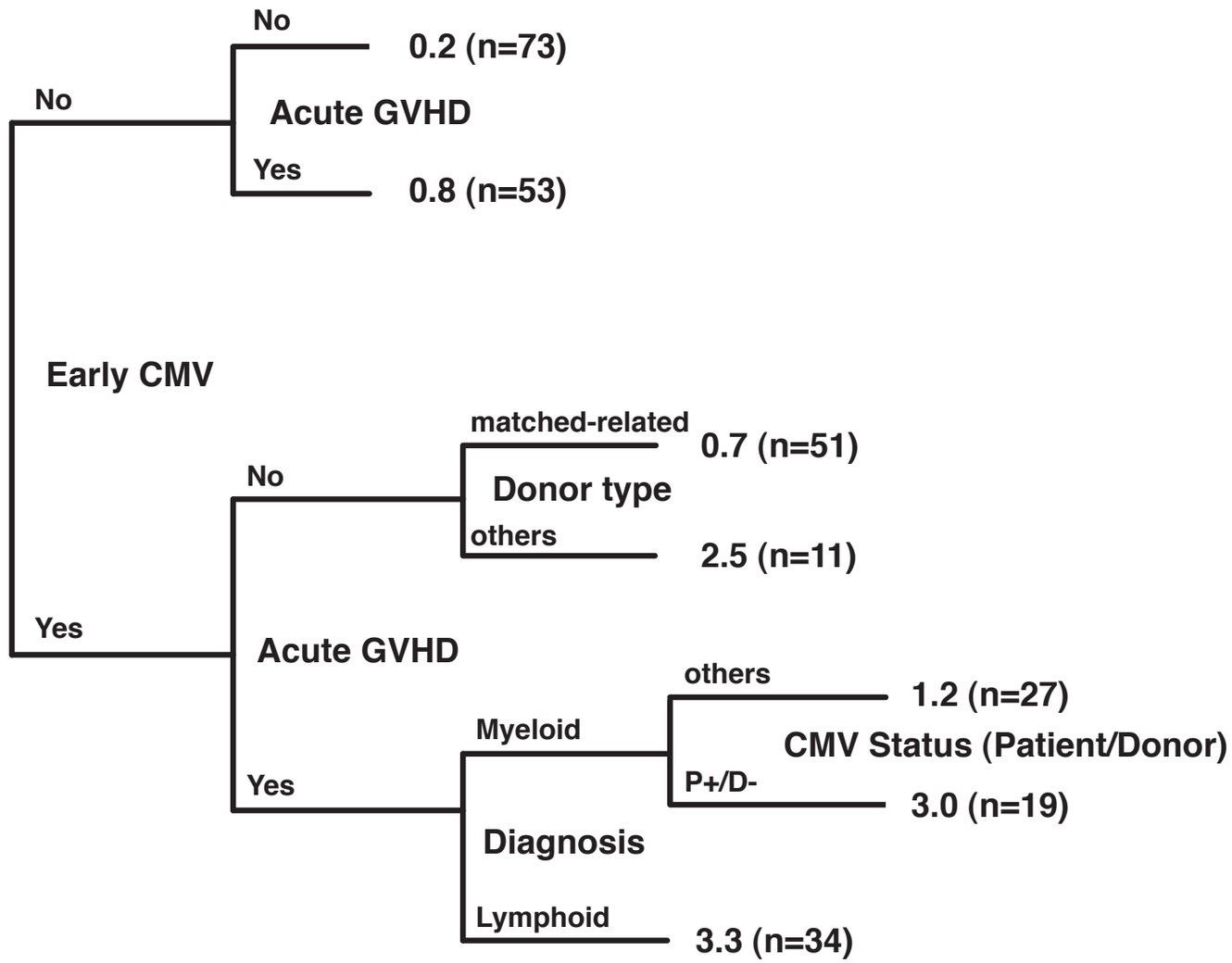
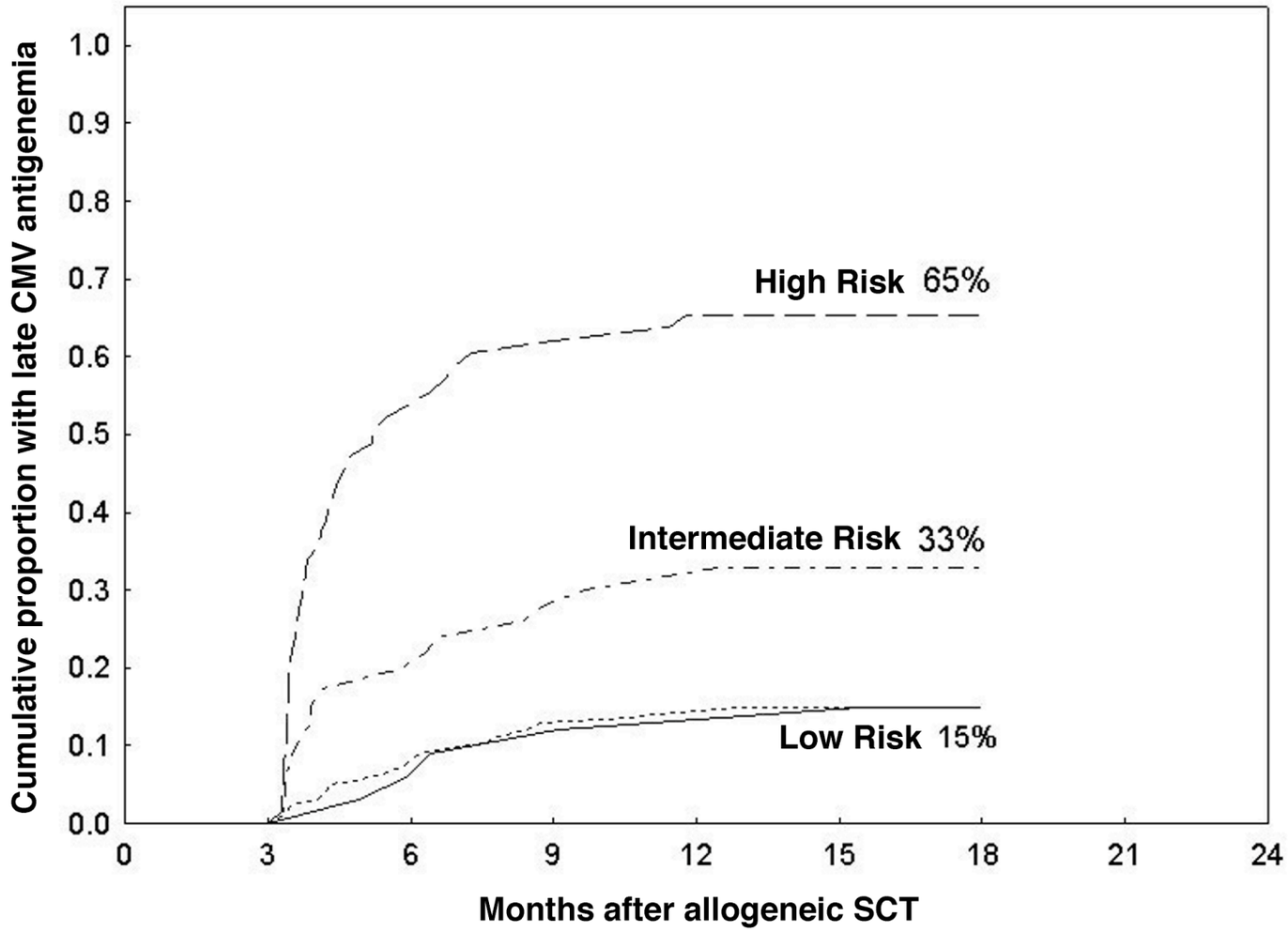


Figure 2

Ozdemir, Komanduri, et al., ASH, 2004

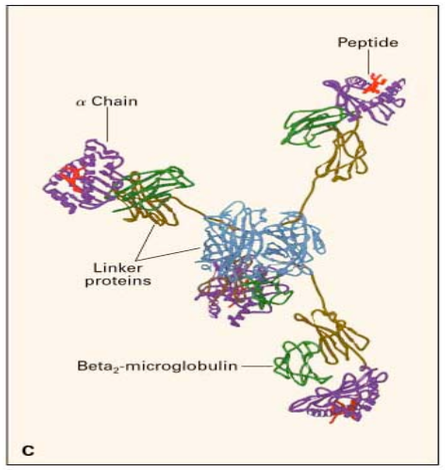
Late CMV reactivation: risk stratification

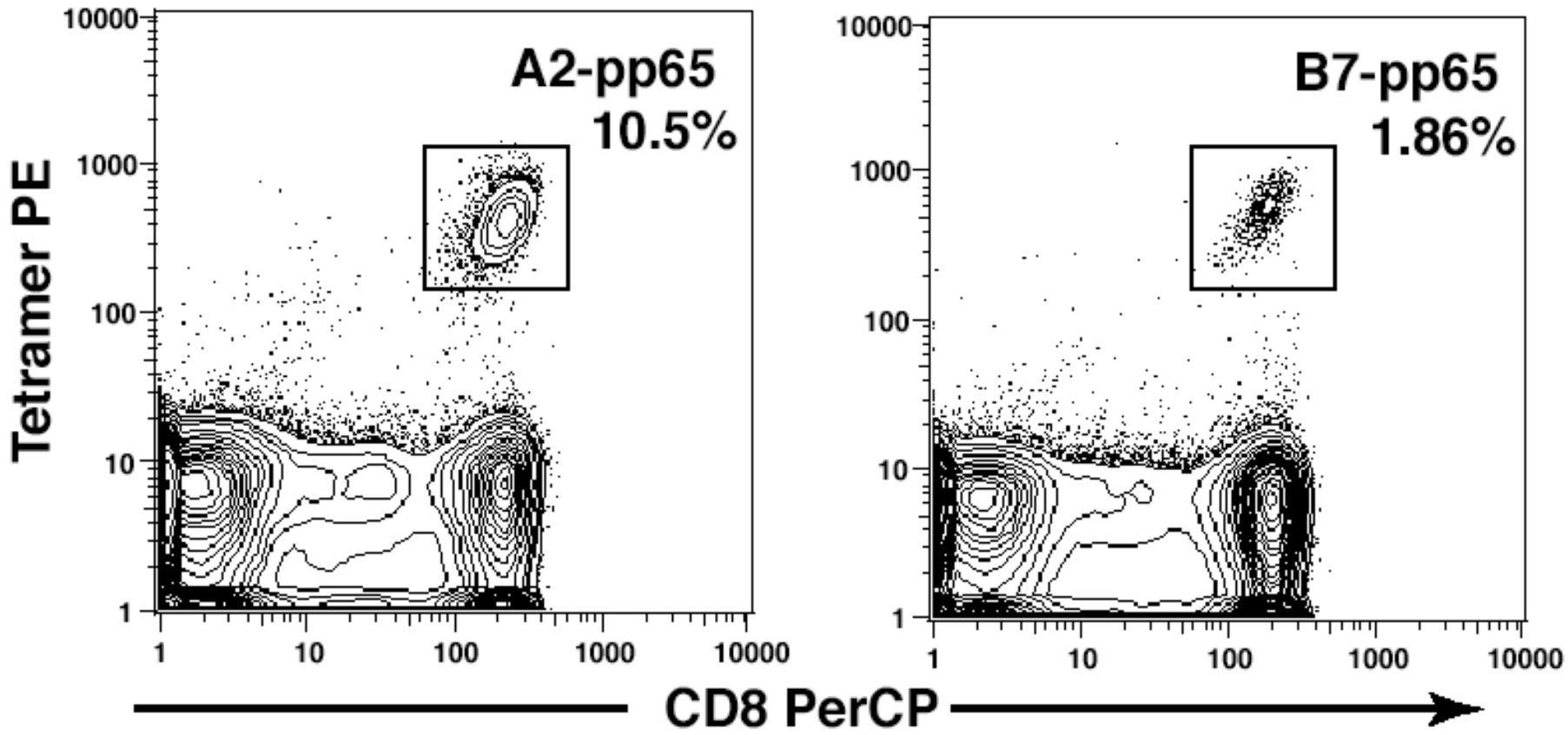
Risk Classification	Clinical Factors
Low	Patients with no antecedent early reactivation MR, no aGVHD *and* Myeloid
Intermediate	MUD/MMR/MR + aGVHD *and* Myeloid/P + D + MR, no aGVHD *and* Lymphoid
High	MUD/MMR/MR + aGVHD *and* Lymphoid/P + D + MUD/MMR/MR + aGVHD *and* Lymphoid/P + D– MUD/MMR/MR + aGVHD *and* Myeloid/P + D–



Ozdemir, et al, *Bone Marrow Transplantation* 2007

Quantitation of CMV-specific CD8+ T cells in SCT recipients





blood

2002 100: 3690-3697
Prepublished online July 5, 2002;
doi:10.1182/blood-2002-05-1387

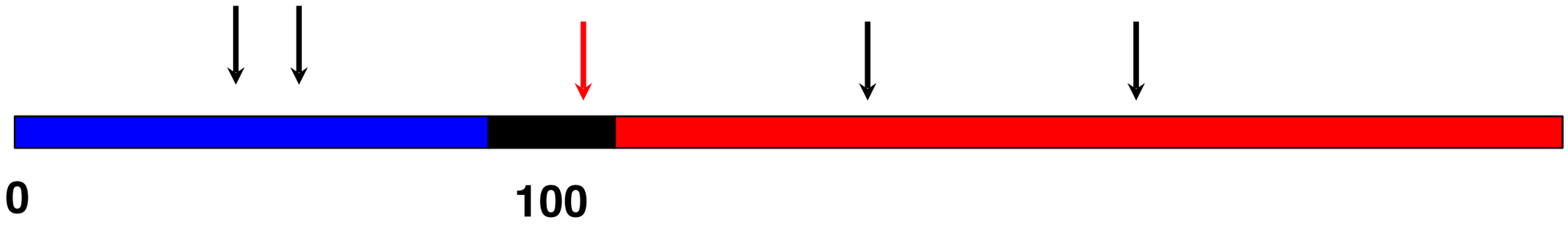
Cytomegalovirus reactivation following allogeneic stem cell transplantation is associated with the presence of dysfunctional antigen-specific CD8$^+$ T cells

Evren Özdemir, Lisa S. St. John, Geraldine Gillespie, Sarah Rowland-Jones, Richard E. Champlin, Jeffrey J. Molldrem and Krishna V. Komanduri

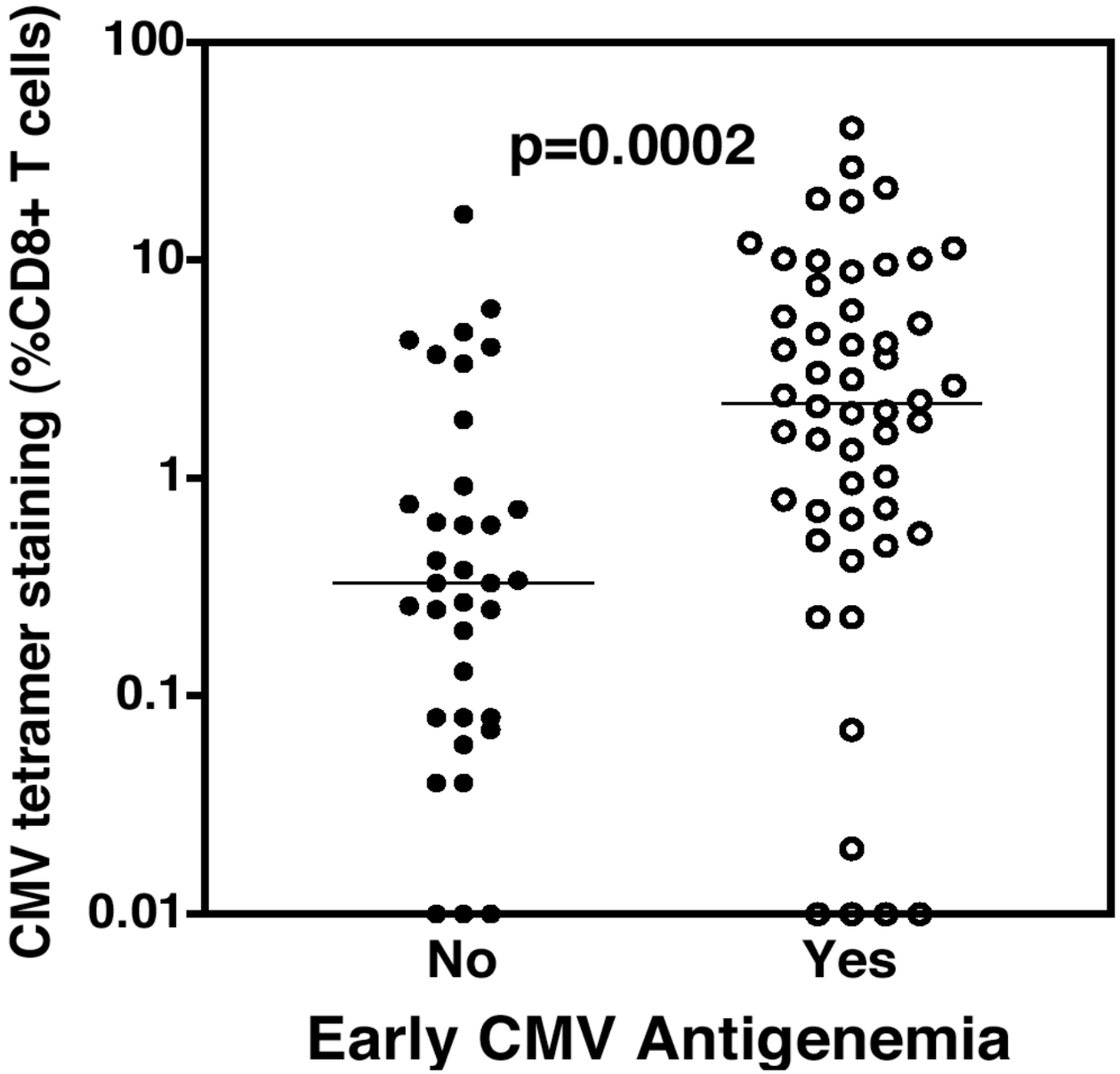
Sampling vs. Reactivation

“Early” Reactivation

“Late” Reactivation



~Day +90 Increased CMV-specific CD8+ T cells in patients with reactivation

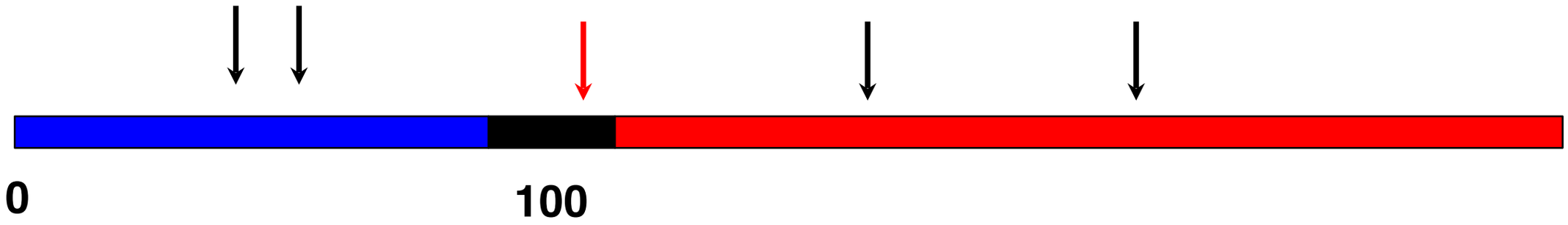


Ozdemir, Komanduri et al., *Blood*, 2002

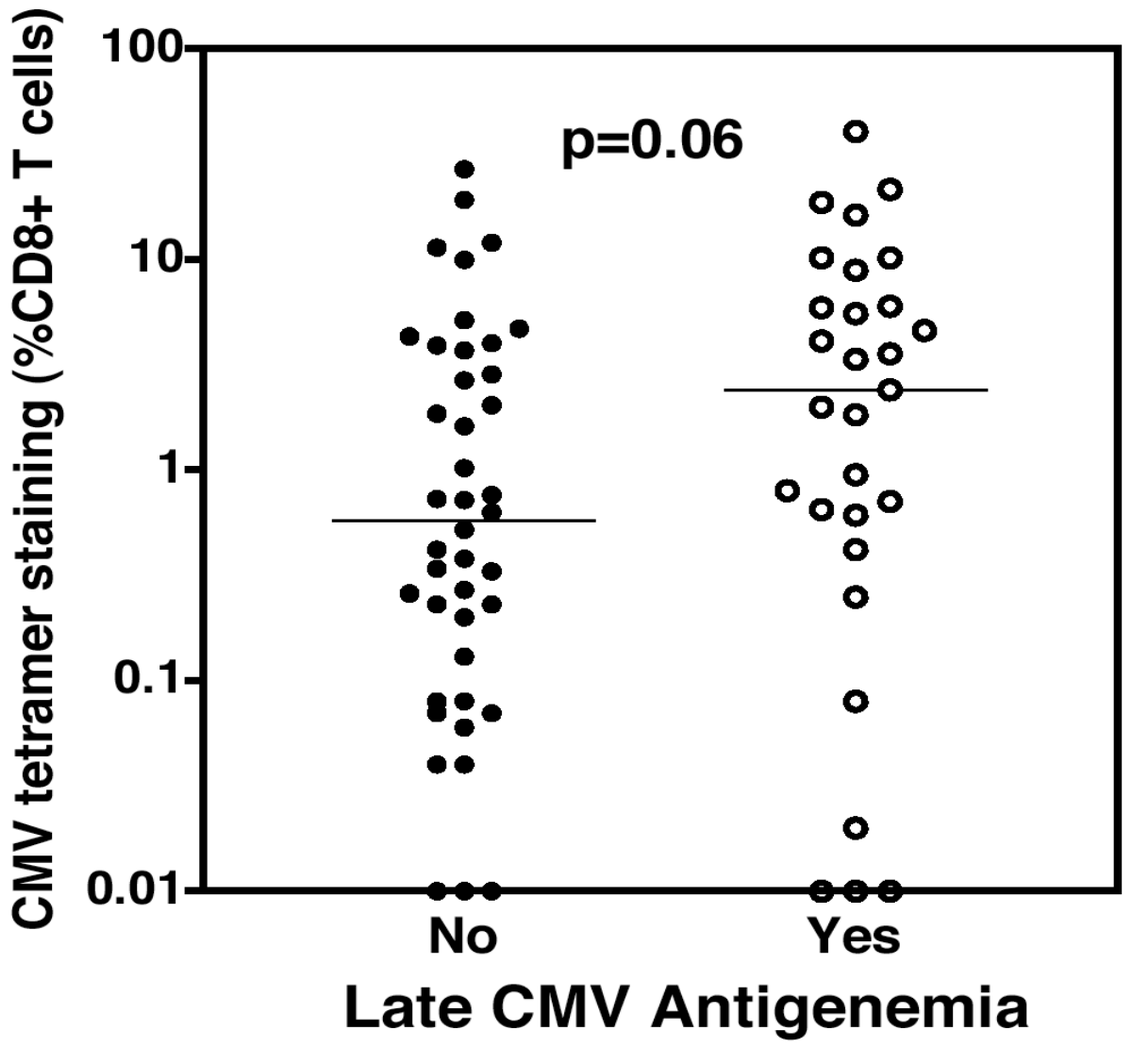
Sampling vs. Reactivation

“D100” Reactivation

“Late” Reactivation

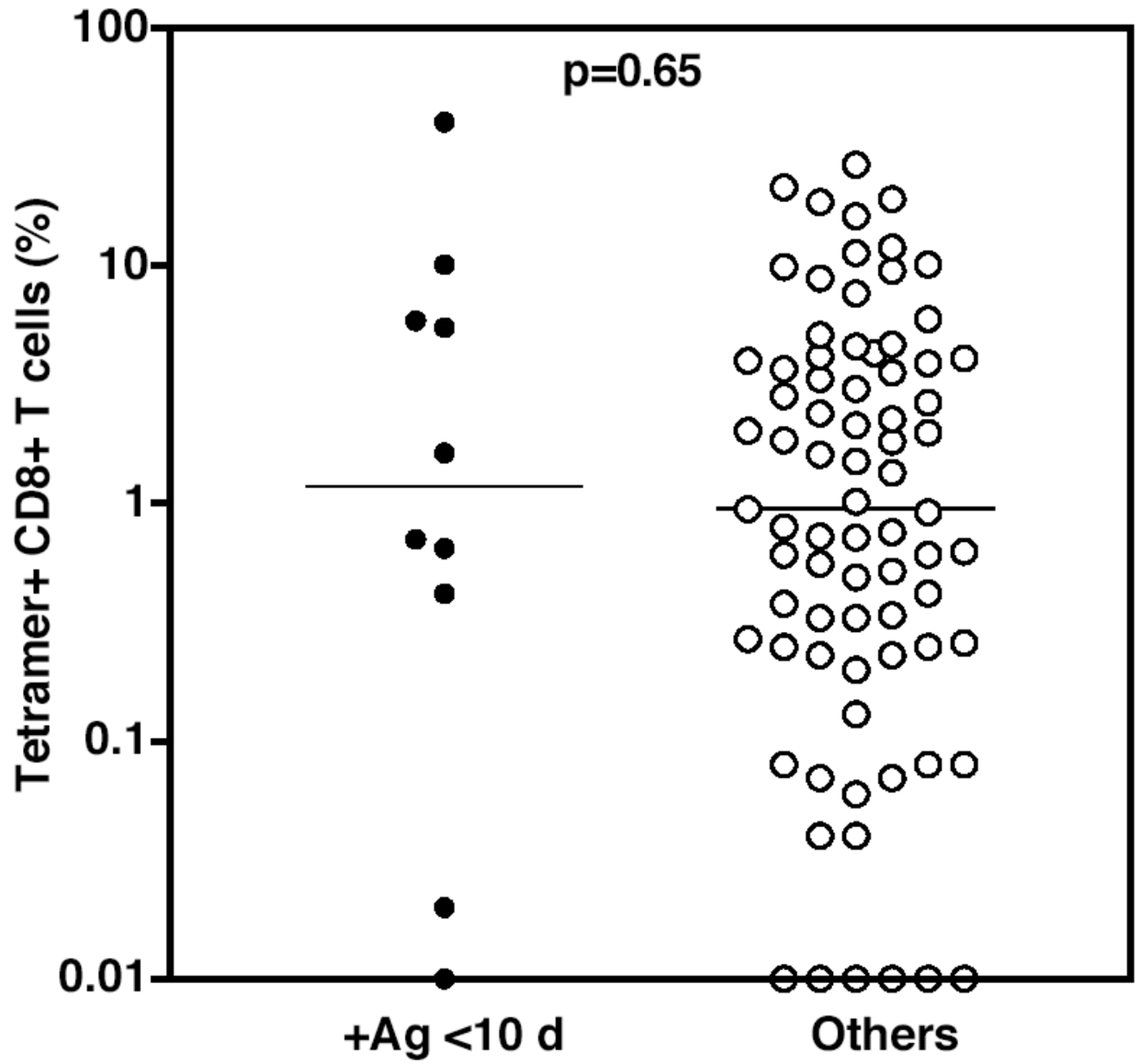


~Day +90 Increased CMV-specific CD8+ T cells in patients with early reactivation



Ozdemir, Komanduri et al., *Blood*, 2002

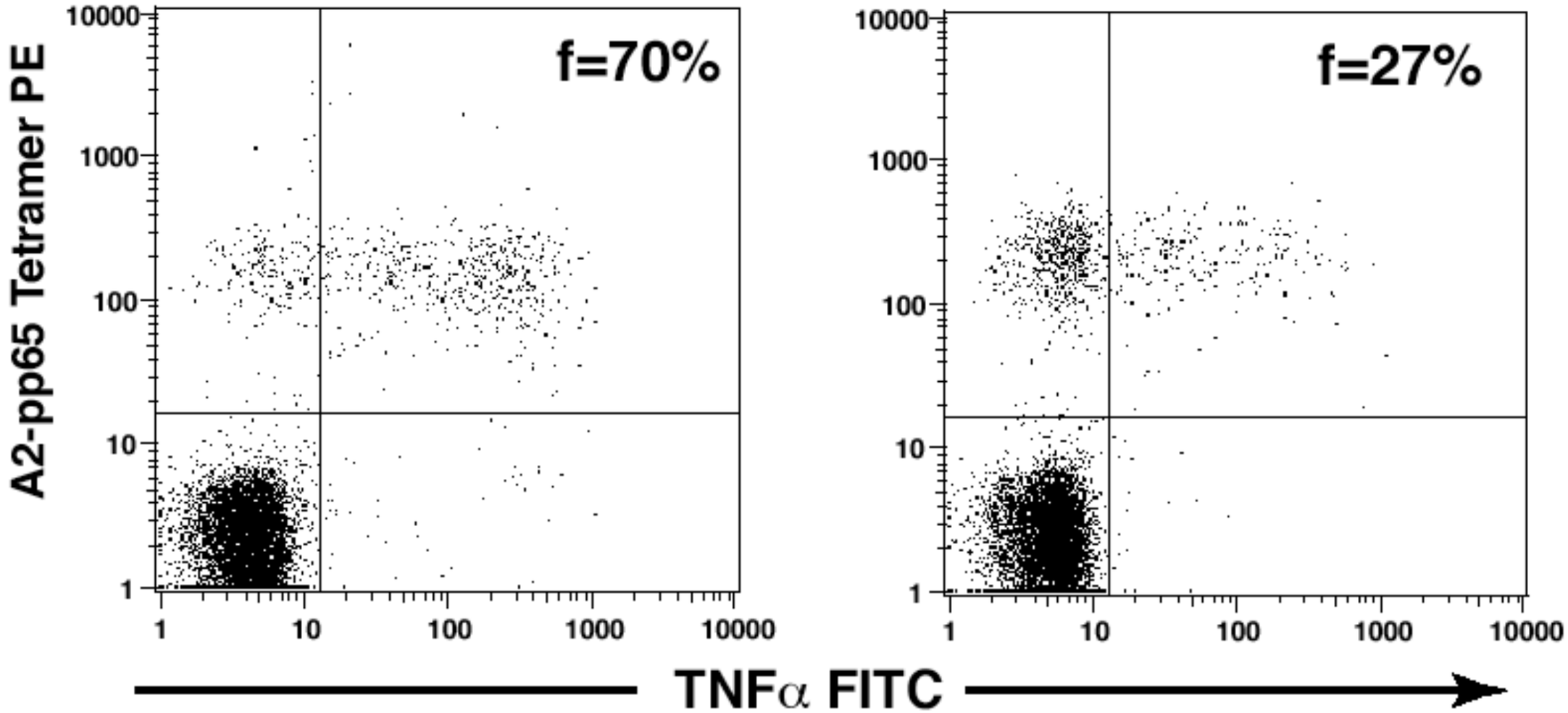
High frequencies of CMV-specific CD8+ T cells in patients assessed proximate to CMV antigenemia (4/10 on day of sampling: 40.5%, 1.6%, 10.2%, 0.7%)



Ozdemir, Komanduri et al., *Blood*, 2002

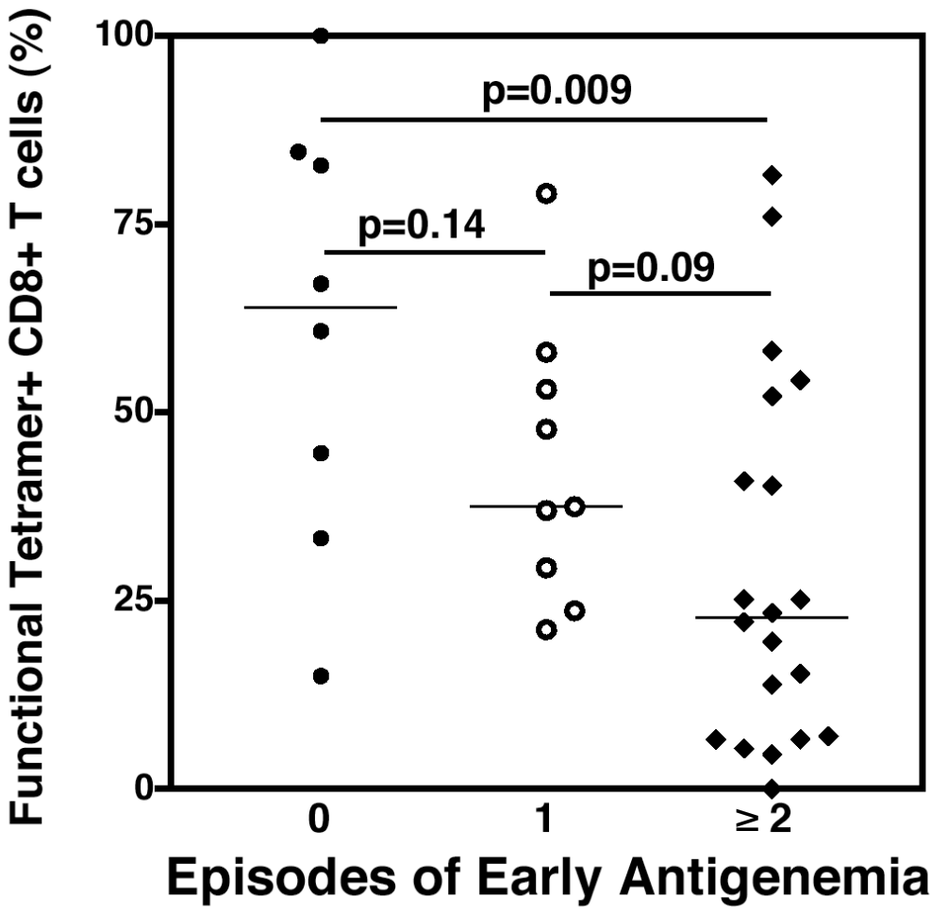
Combined tetramer/cytokine flow cytometry analysis

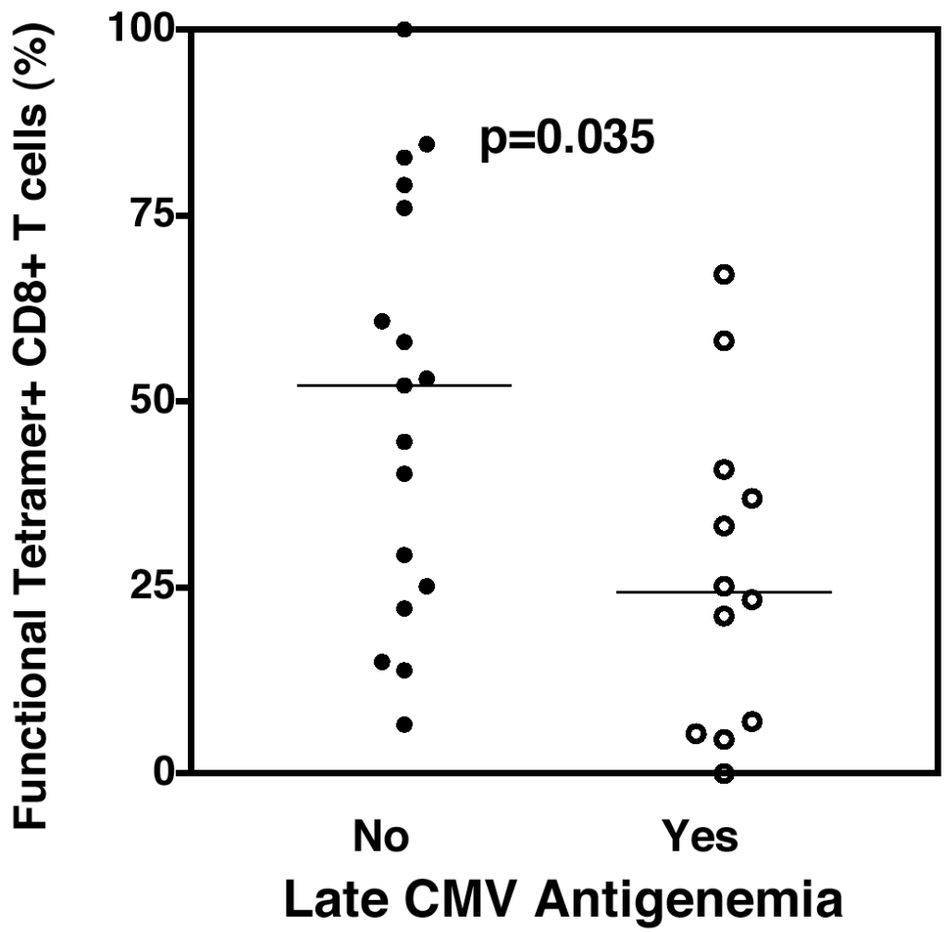
function within Ag-specific CD8+ T cells



Ozdemir, Komanduri et al., *Blood*, 2002

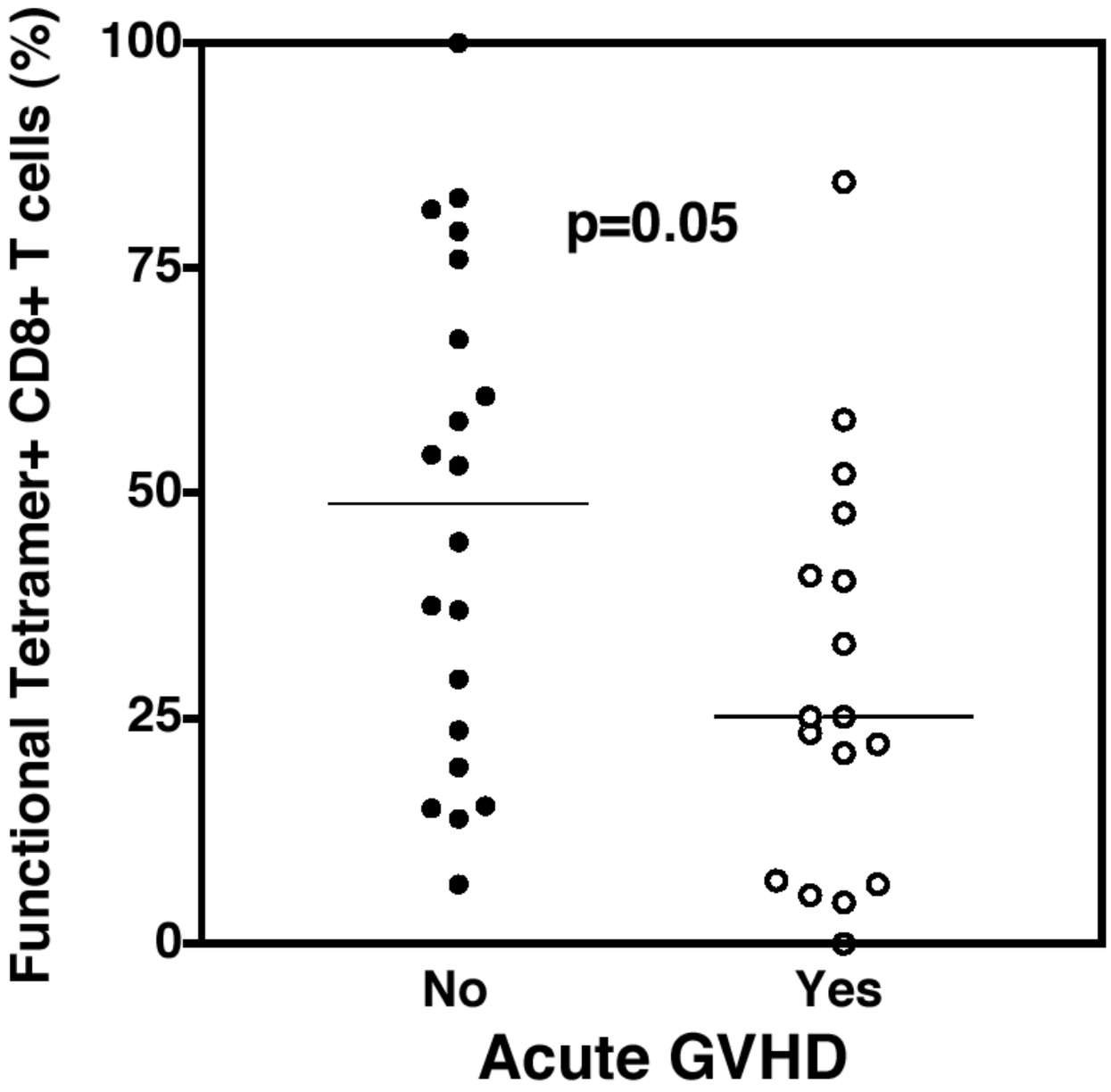
Dysfunction of CD8+ T cells is associated with CMV reactivation





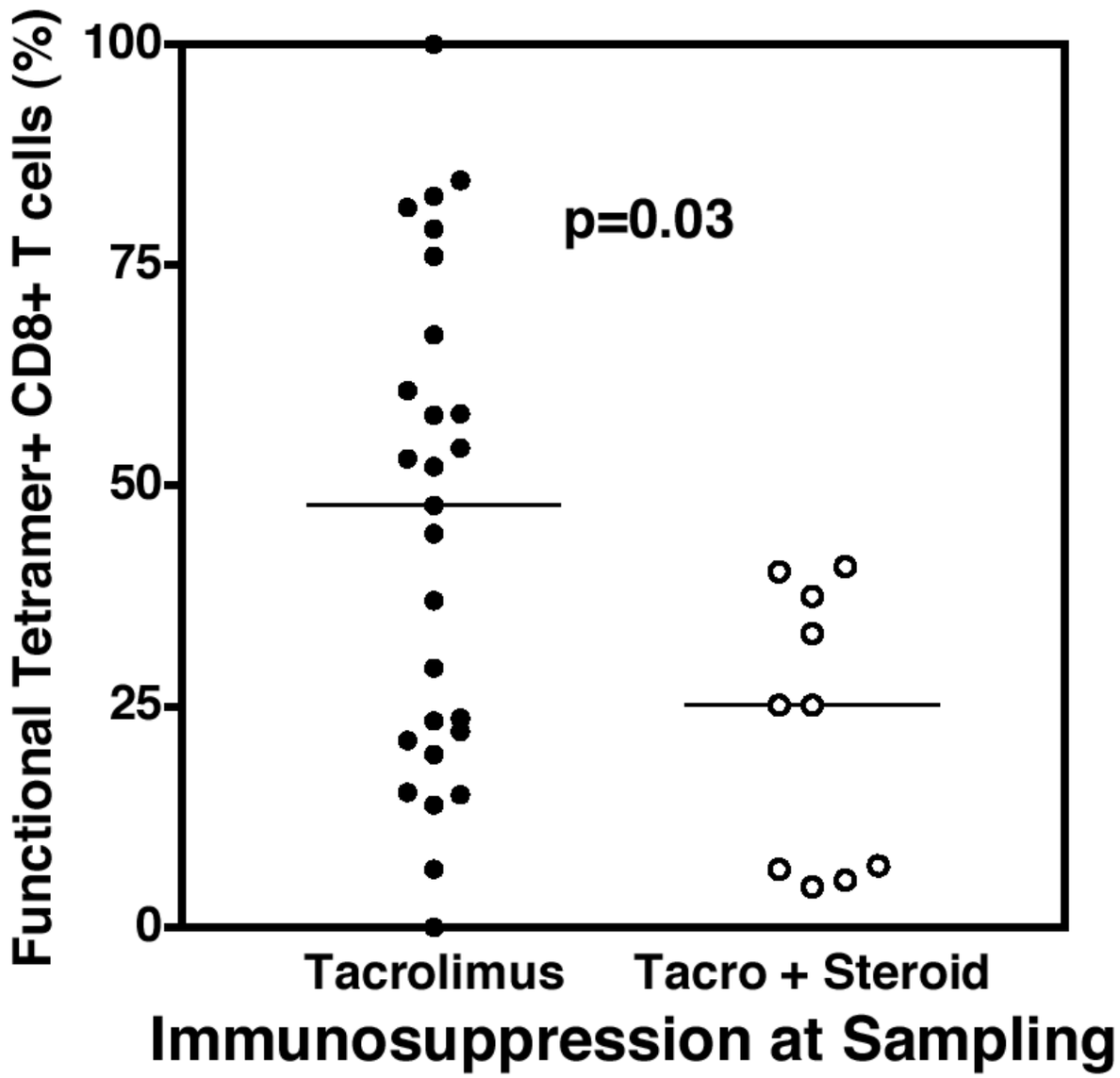
Ozdemir, Komanduri et al., *Blood*, 2002

Dysfunction *not number* of CMV-specific CD8+ T cells is associated with acute GVHD



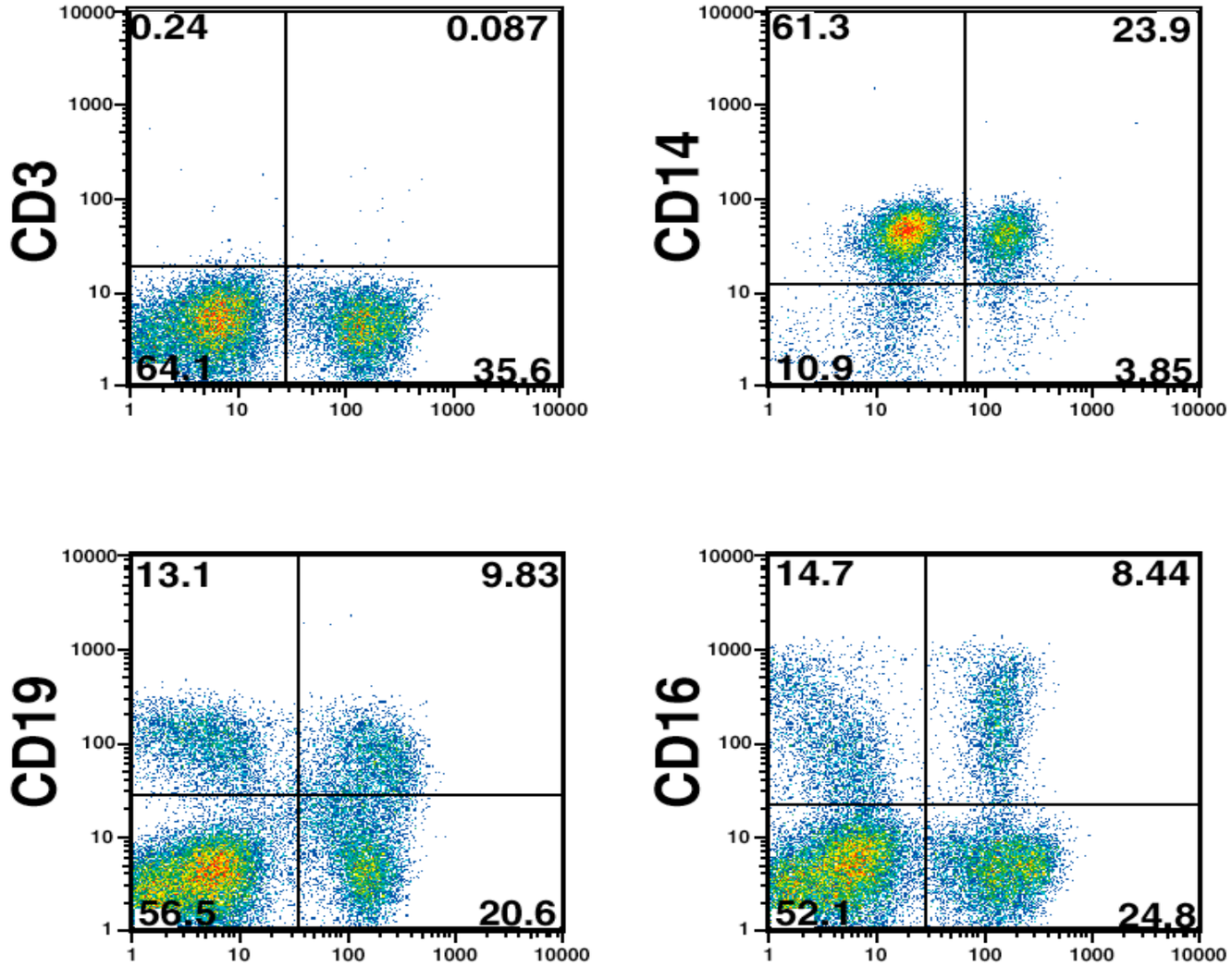
Ozdemir, Komanduri et al., *Blood*, 2002

Dysfunction (*not number)* of CD8+ T cells is associated with steroid use



Ozdemir, Komanduri et al., *Blood*, 2002

Sometimes the problem is quantitative
(Double CBT recipient with multiple life-threatening infections)



de Lima and Komanduri, *Br J Hematol* 2002

blood

2007 110: 4543-4551
Prepublished online Aug 1, 2007;
doi:10.1182/blood-2007-05-092130

Delayed immune reconstitution after cord blood transplantation is characterized by impaired thymopoiesis and late memory T-cell skewing

Krishna V. Komanduri, Lisa S. St. John, Marcos de Lima, John McMannis, Steven Rosinski, Ian McNiece, Susan G. Bryan, Indreshpal Kaur, Sean Martin, Eric D. Wieder, Laura Worth, Laurence J. N. Cooper, Demetrios Petropoulos, Jeffrey J. Molldrem, Richard E. Champlin and Elizabeth J. Shpall

- CBT recipients have profound immune deficits, but few studies have detailed immune recovery
- 12/2007: Reported results of longitudinal immune recovery studies from a group of 32 subjects
- Last interim update in 2009 (47 patients)

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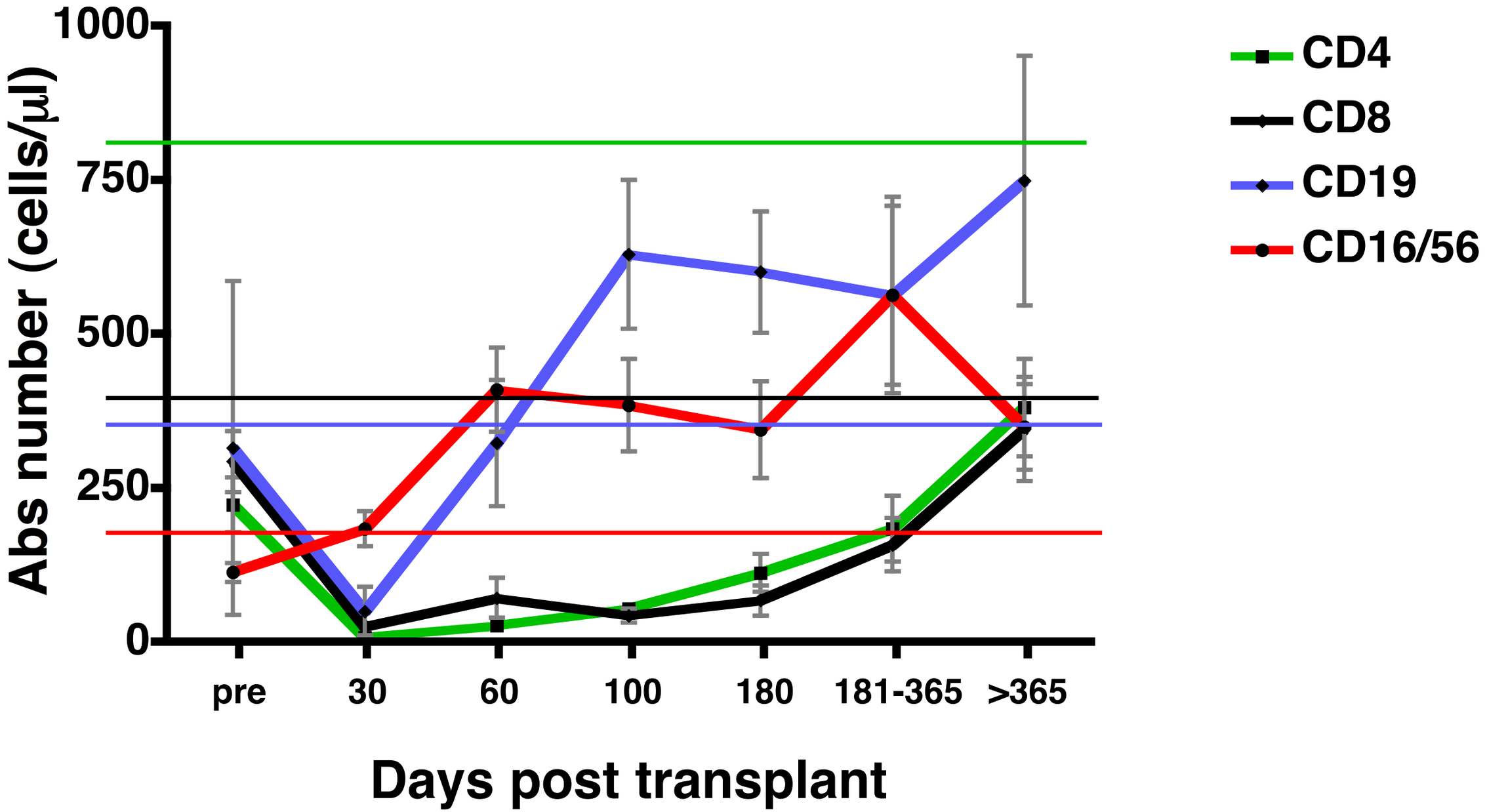

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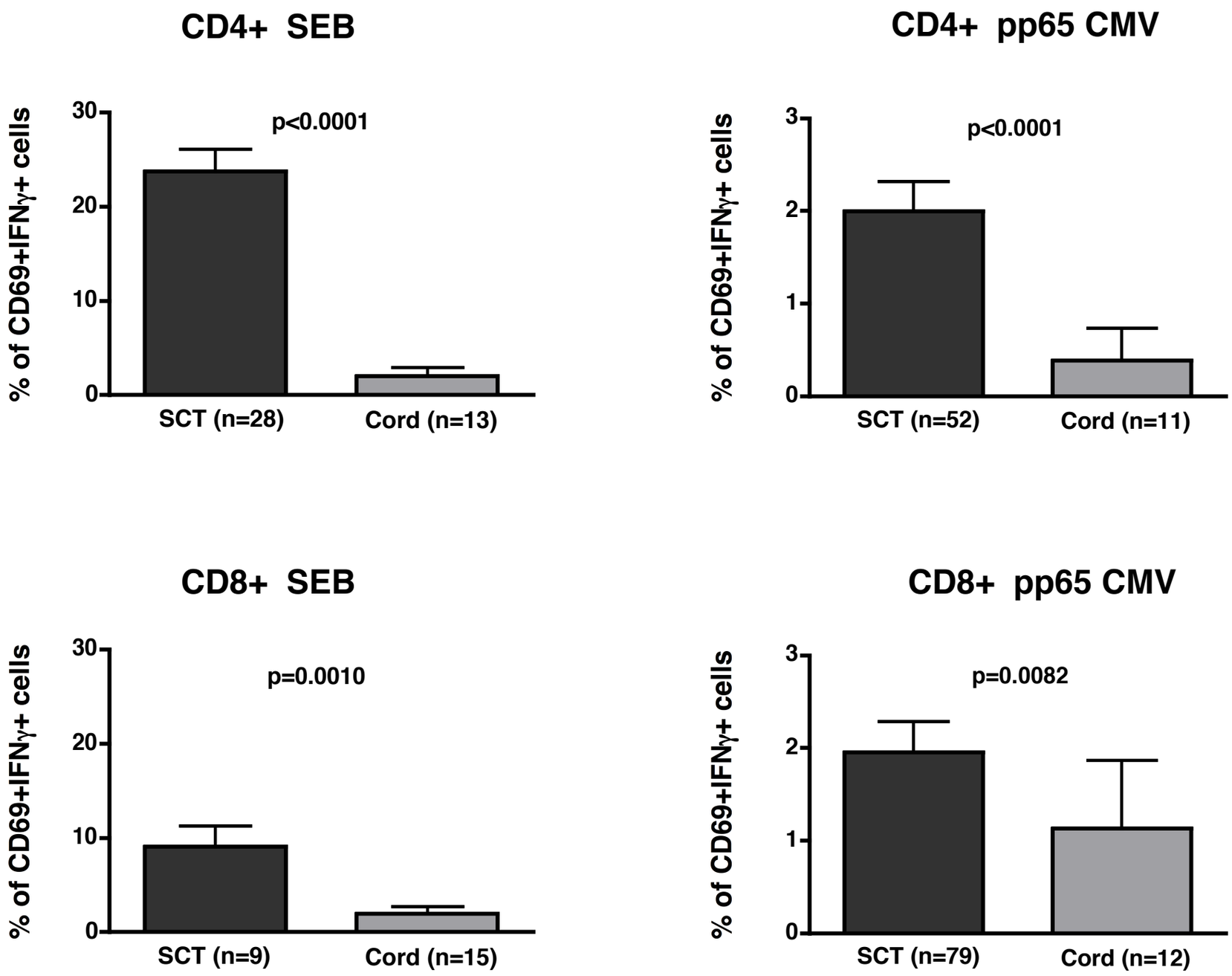
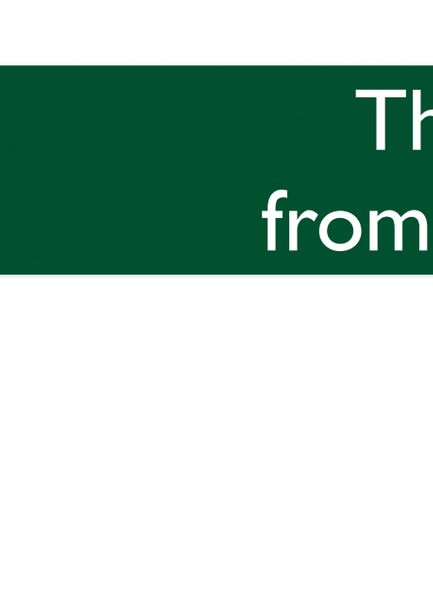
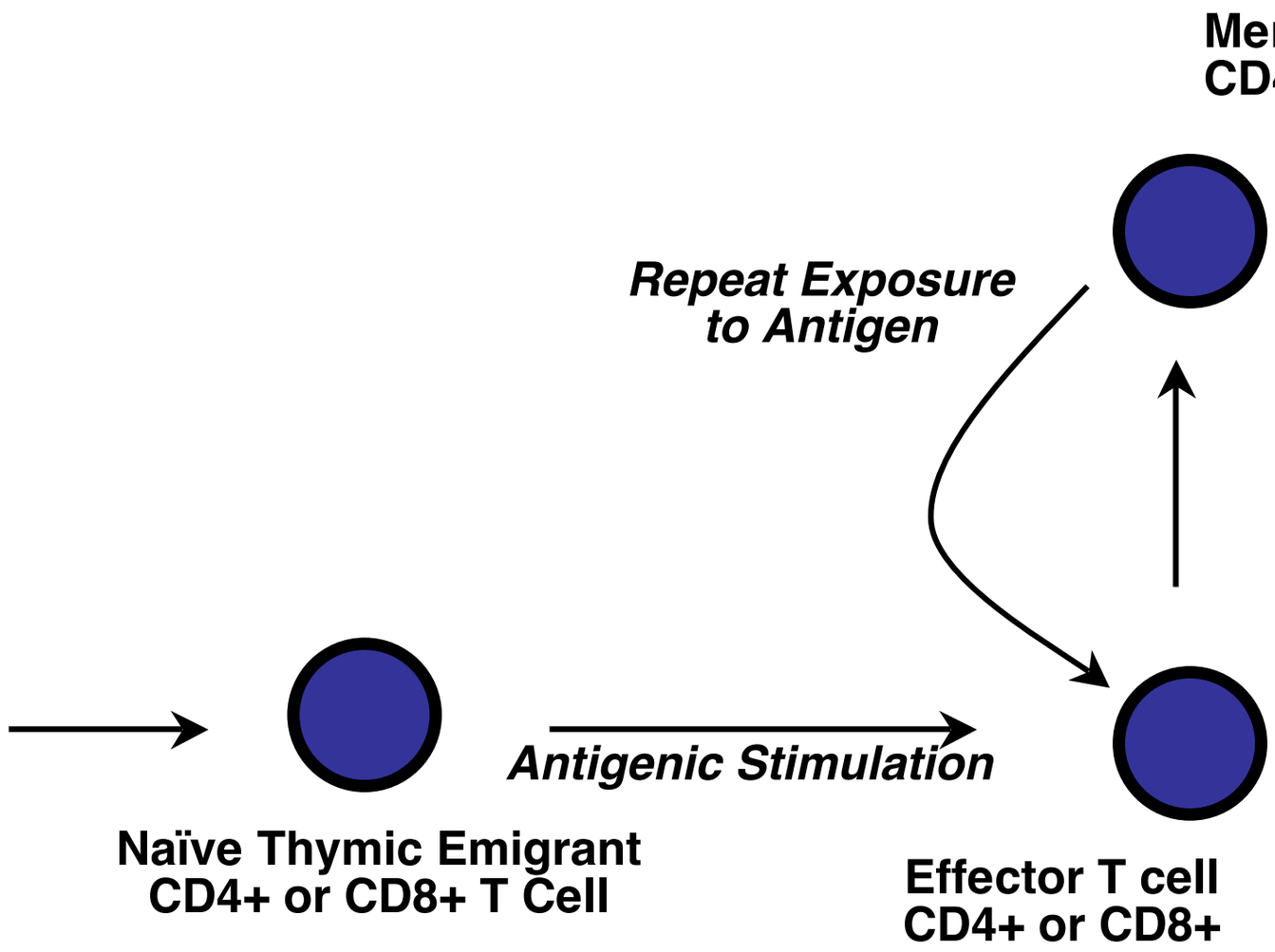


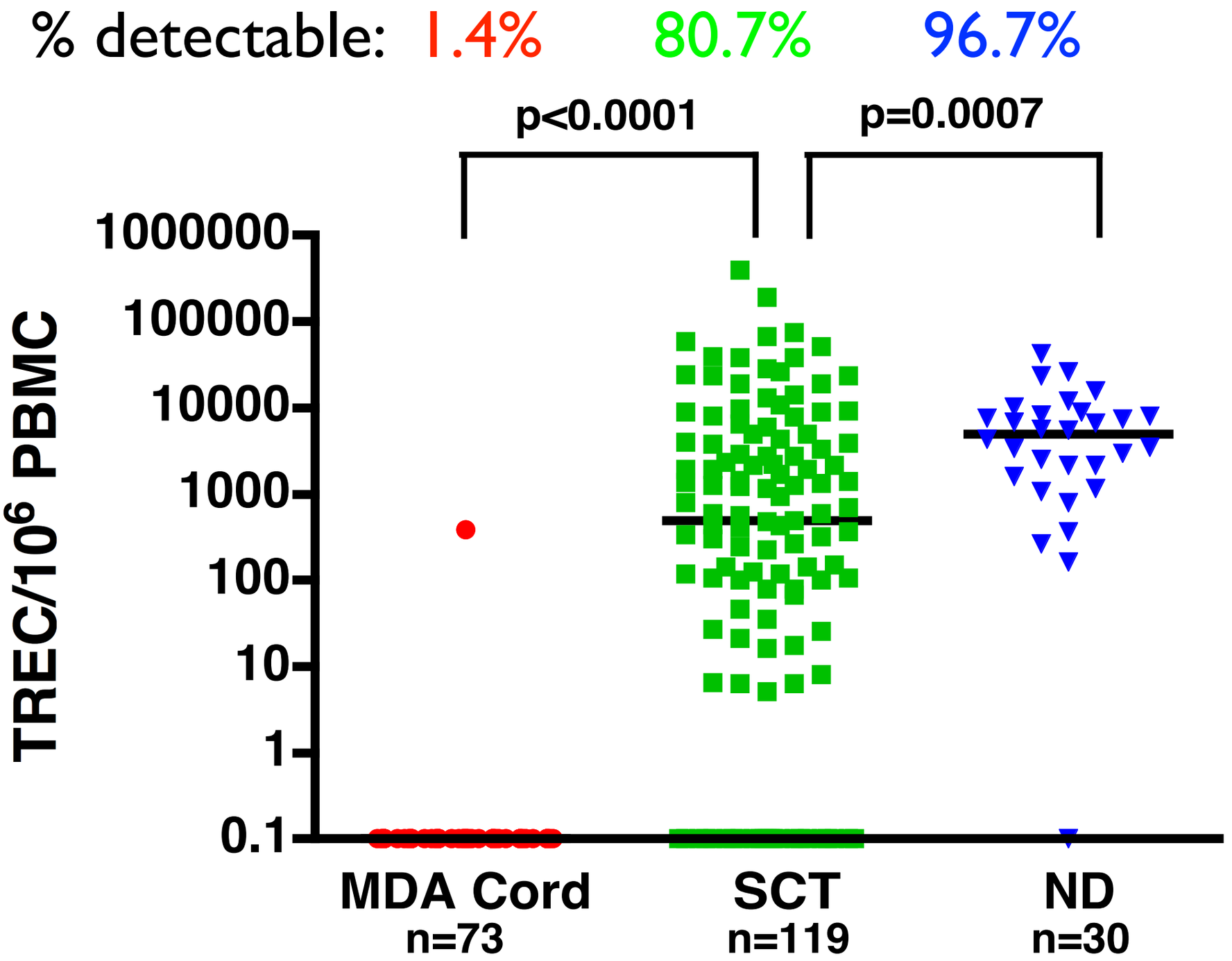
The thymus generates a naive T cell pool from which the memory repertoire is derived



Thymus



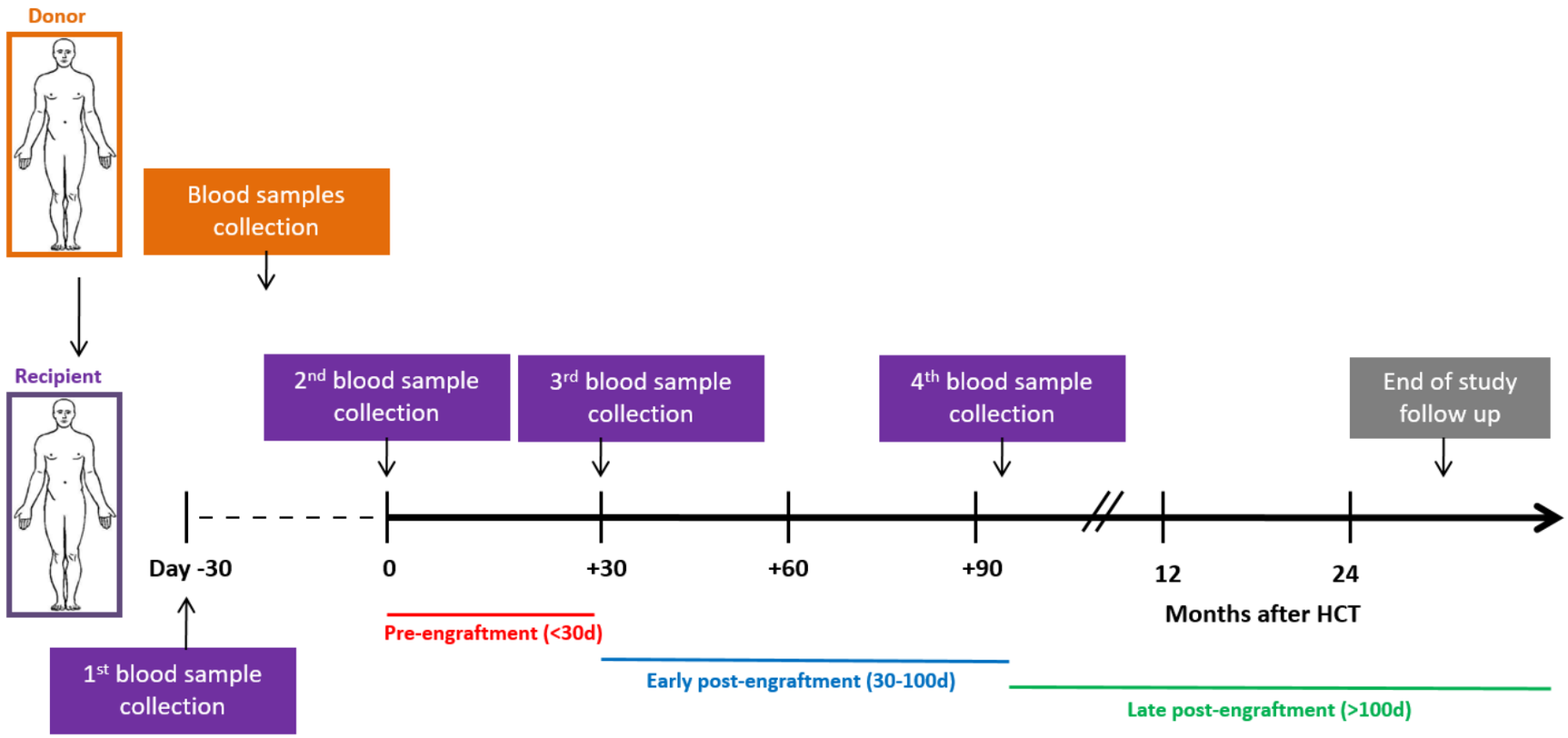
Thymopoiesis can be measured, and is impaired after adult CBT



Why bother looking again at CMV responses in 2017?

- Earlier studies were cross-sectional and not prospective
- Better understanding of function and better technology
- Looking at CD4+ and CD8+ responses to CMV pp65 and IE1, and at naive/memory/Tscm responses
- Looking at combinations of IL-2/IFN/TNF/MIP-1beta
- Correlating above more precisely with patterns of reactivation

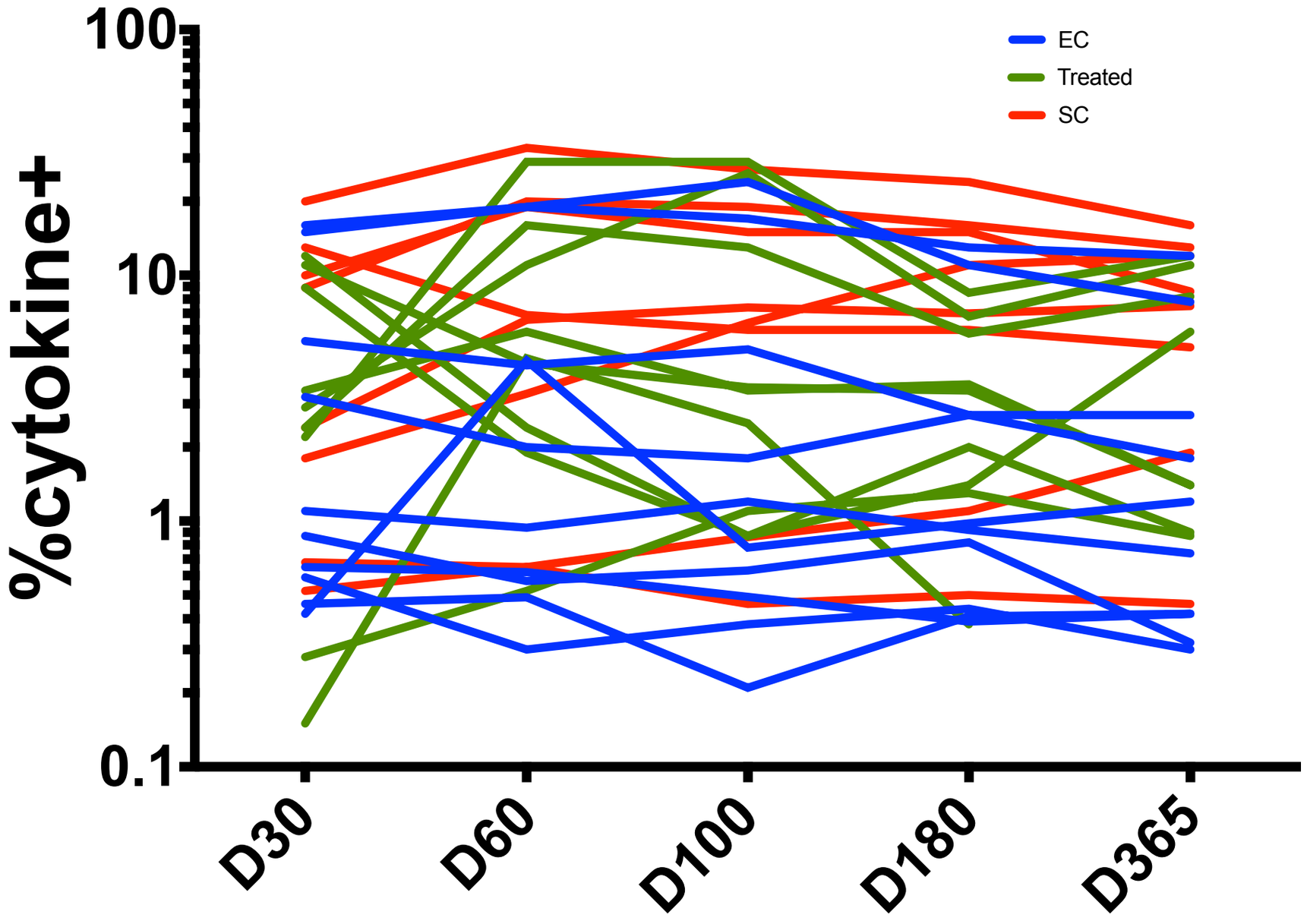
Schema for sample collection for translational research



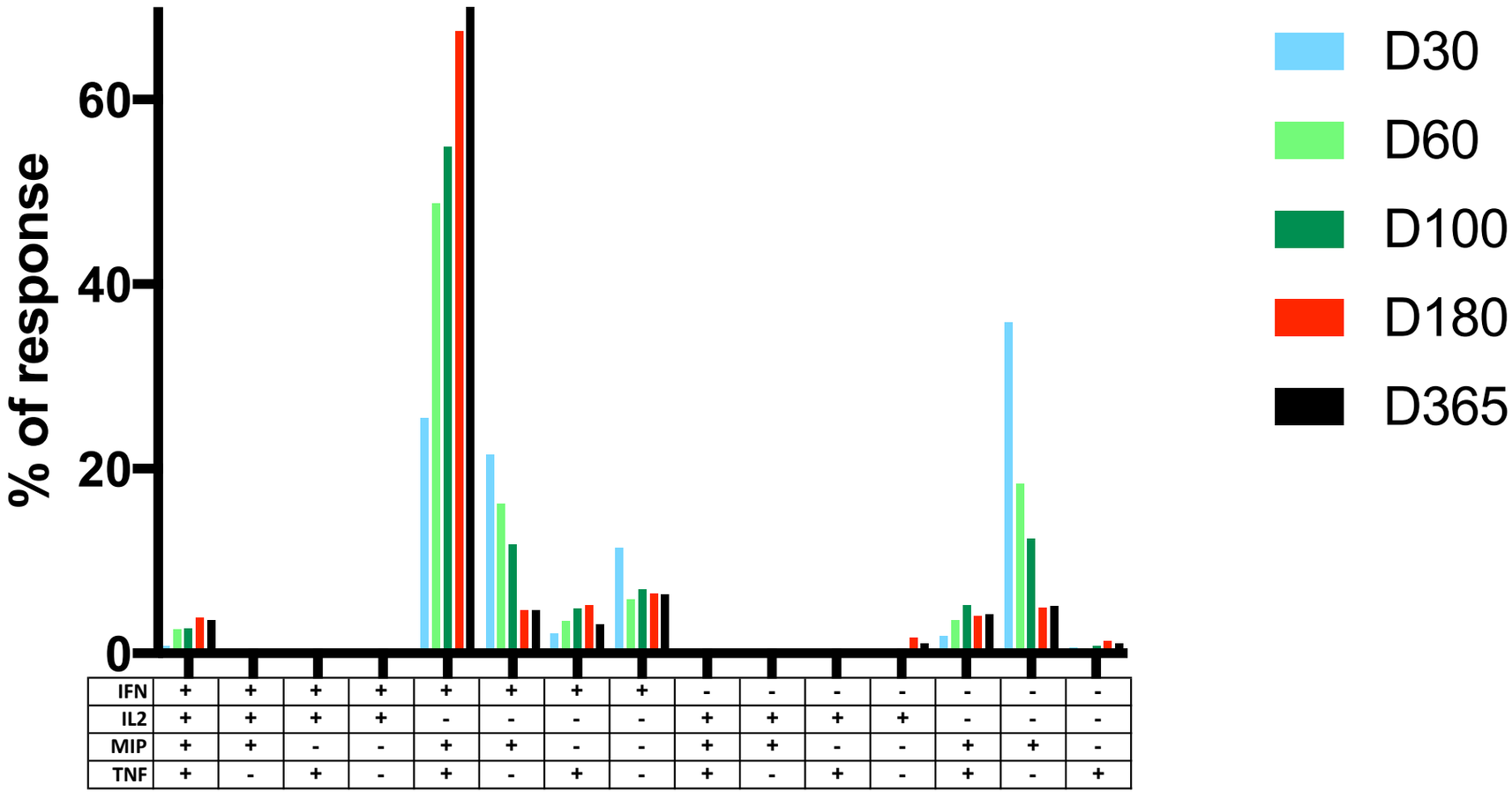
Patterns of CMV reactivation

- Three groups
  - Group 1 (EC): Elite Controller: *at risk but never reactivated*
  - Group 2 (SC) : Spontaneous control: *reactivated but control w/o therapy*
  - Group 3 (treated): *reactivated and received anti-CMV therapy*
- Here we report for the first time group 1 EC (n=10) and compare the results to groups 2 and 3 (n=8, 9)
- First day of reactivation:
  - Group 2: 3, 12, 14, 25, 33, 35, 60, 65 (median= 29)
  - Group 3: 1, 6, 13, 15, 22, 28, 32, 35, 37 (median = 22)

CD8 pp65 summary



Longitudinal CD8+ response to CMV pp65 in one patient

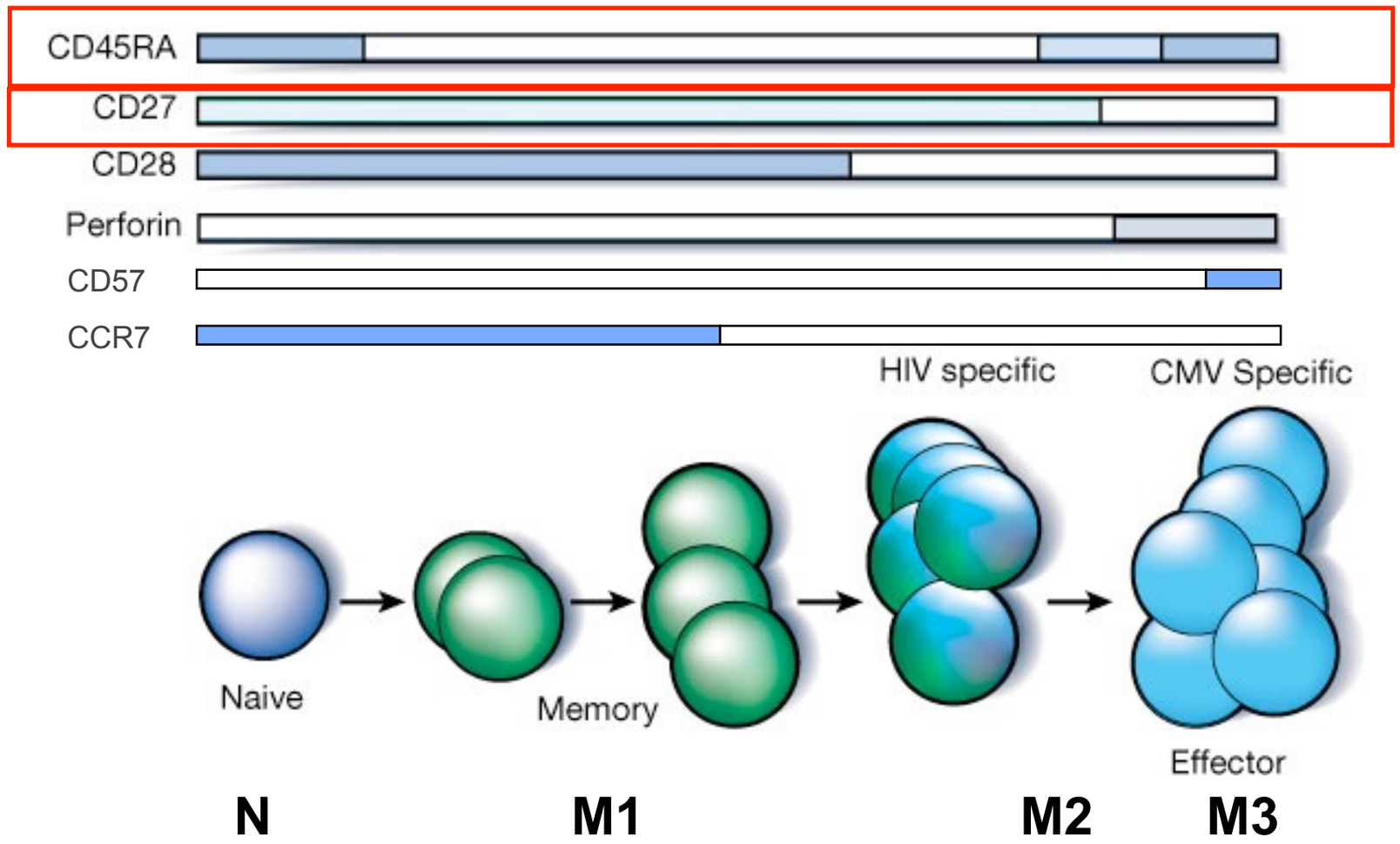


Wieder, Komanduri et al., unpublished

What is the key to immunologic control of CMV?

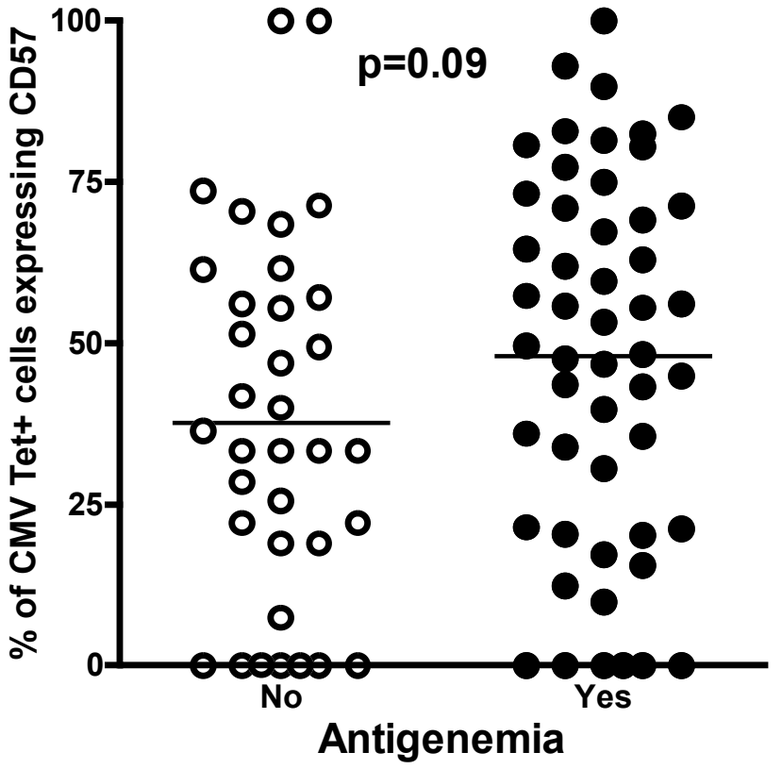
- Differentiation status of responders? Tscm?
- Functional focusing of the response? Clonal diversity?
- Virological?
- Perhaps driven mostly by dysfunction? GVHD? Steroid?
- Lots of work ahead

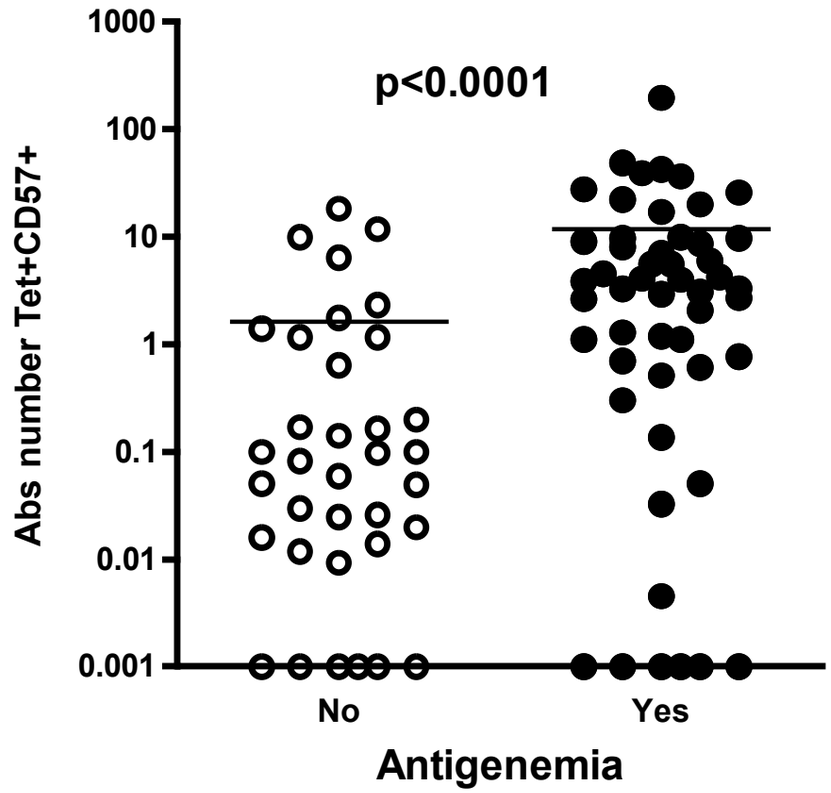
Surface phenotypic markers identify naive and memory T cells



Adapted from McMichael AJ & Rowland-Jones SL, *Nature* 2001

CMV reactivation is associated with an abundance of late effector CD57+ CMV-specific CD8+ T cells

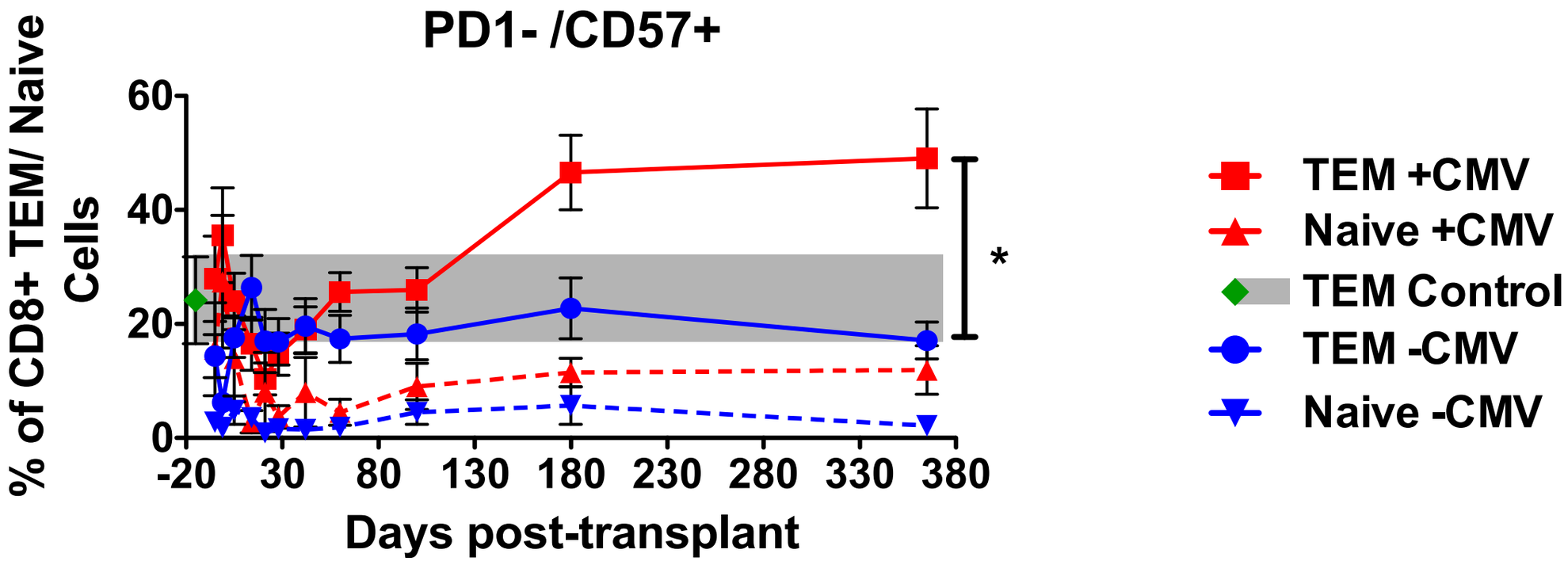




Ozdemir et al., *ASH* 2005

Impact of CMV Reactivation on TCR Repertoire

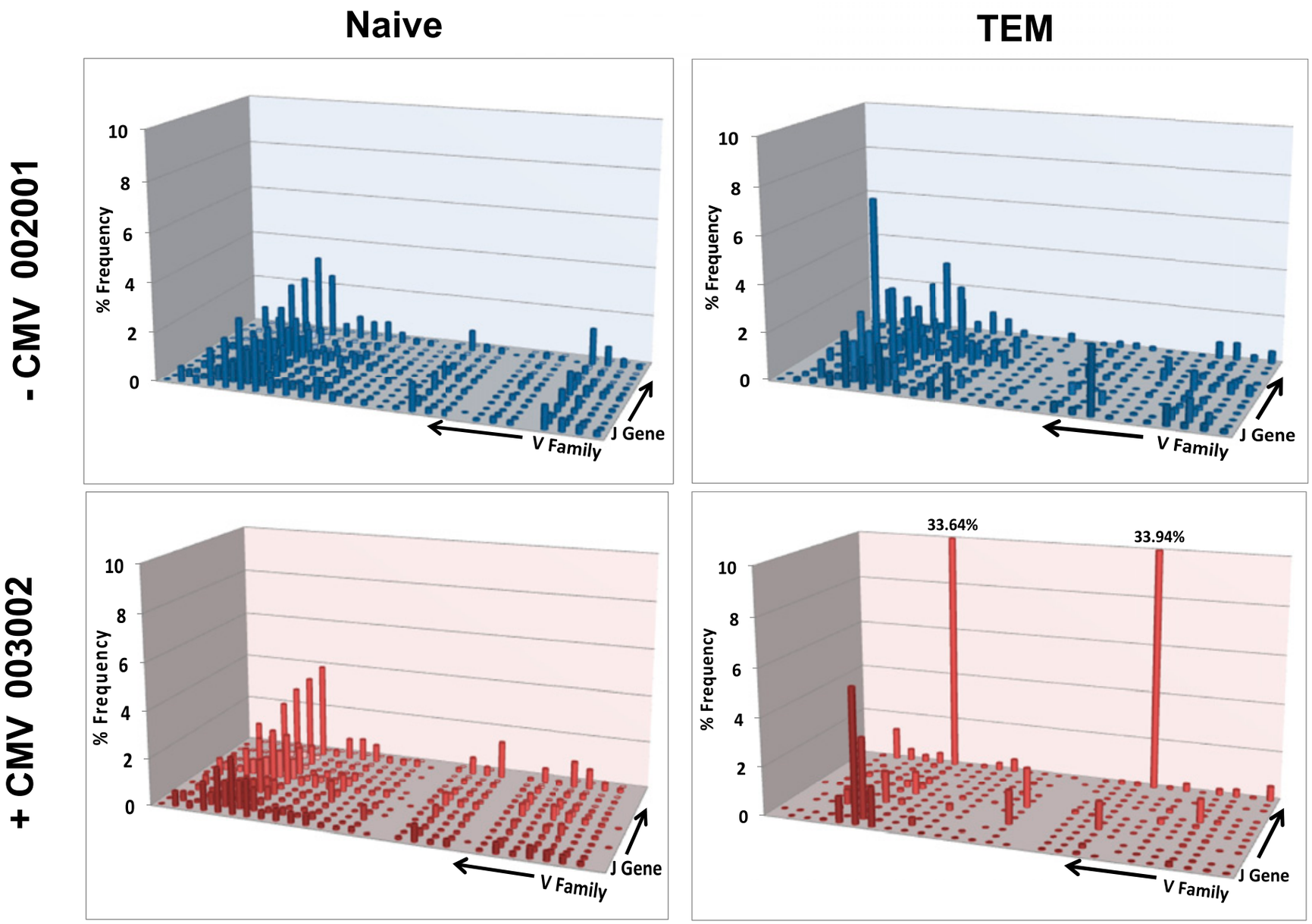
CMV-driven Exhaustion (PD1-/CD57+ CD8+ T cells)



Suessmuth, Kean et al., *Blood* Plenary Paper, 2016

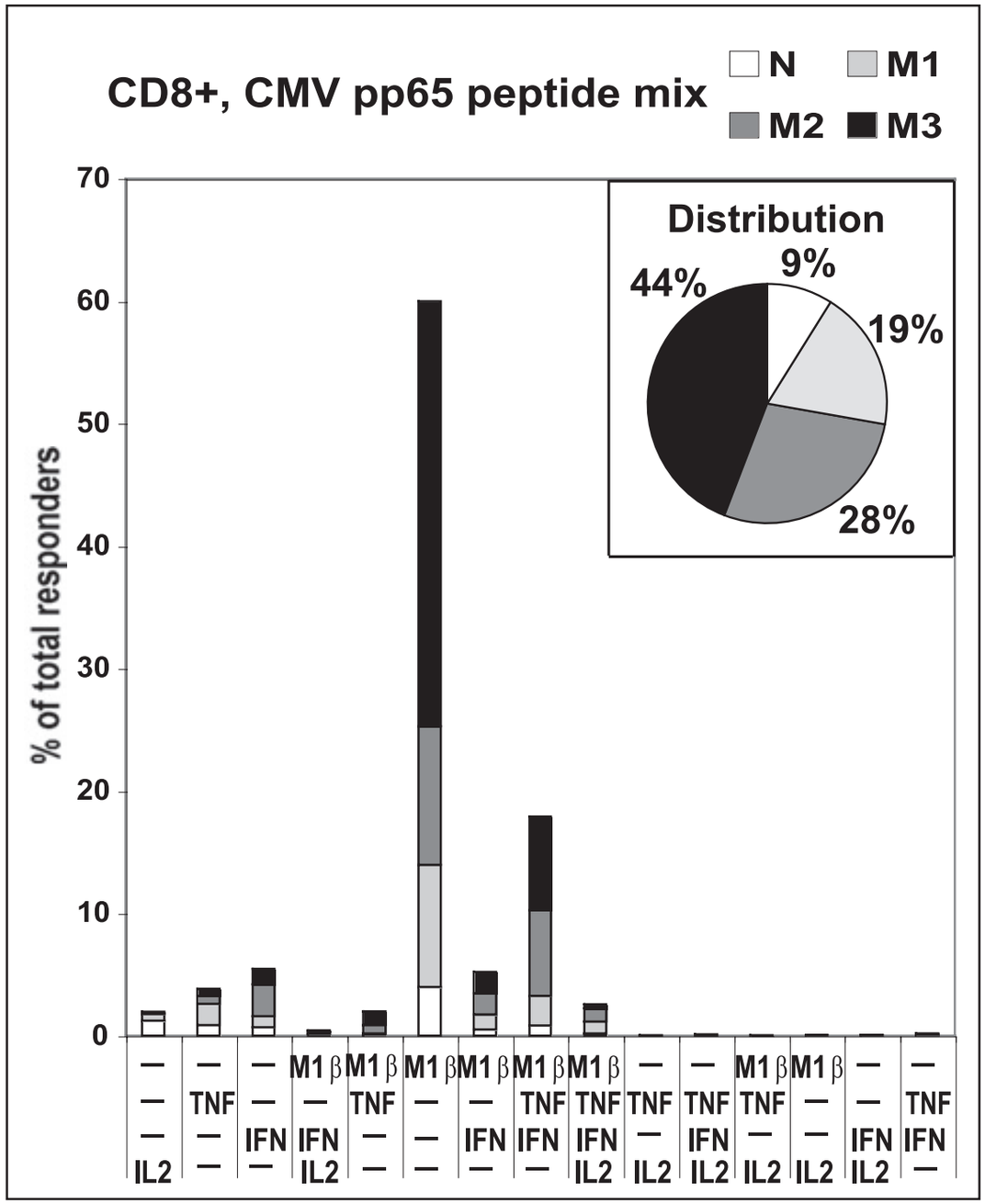
Impact of CMV Reactivation on TCR Repertoire

CMV-driven Exhaustion (Repertoire Skewing in TEM but not T_N cells)



Suessmuth, Kean et al., *Blood* Plenary Paper, 2016

Human CMV and other virus-specific T cells are highly differentiated



The Journal of Immunology

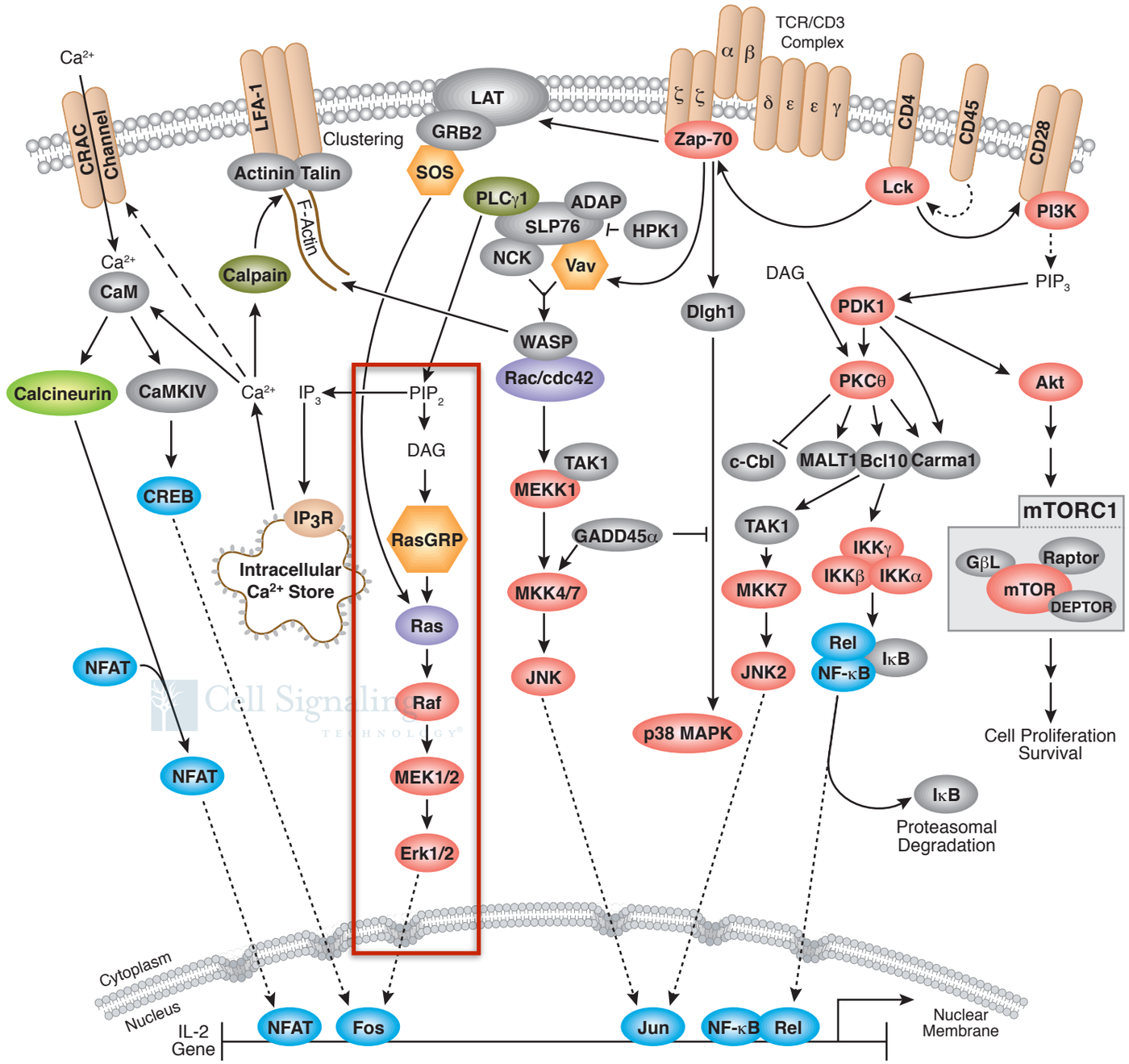
Human Late Memory CD8⁺ T Cells Have a Distinct Cytokine Signature Characterized by CC Chemokine Production without IL-2 Production

Tae Kon Kim, Lisa S. St. John, Eric D. Wieder, Jahan Khalili, Qing Ma and Krishna V. Komanduri

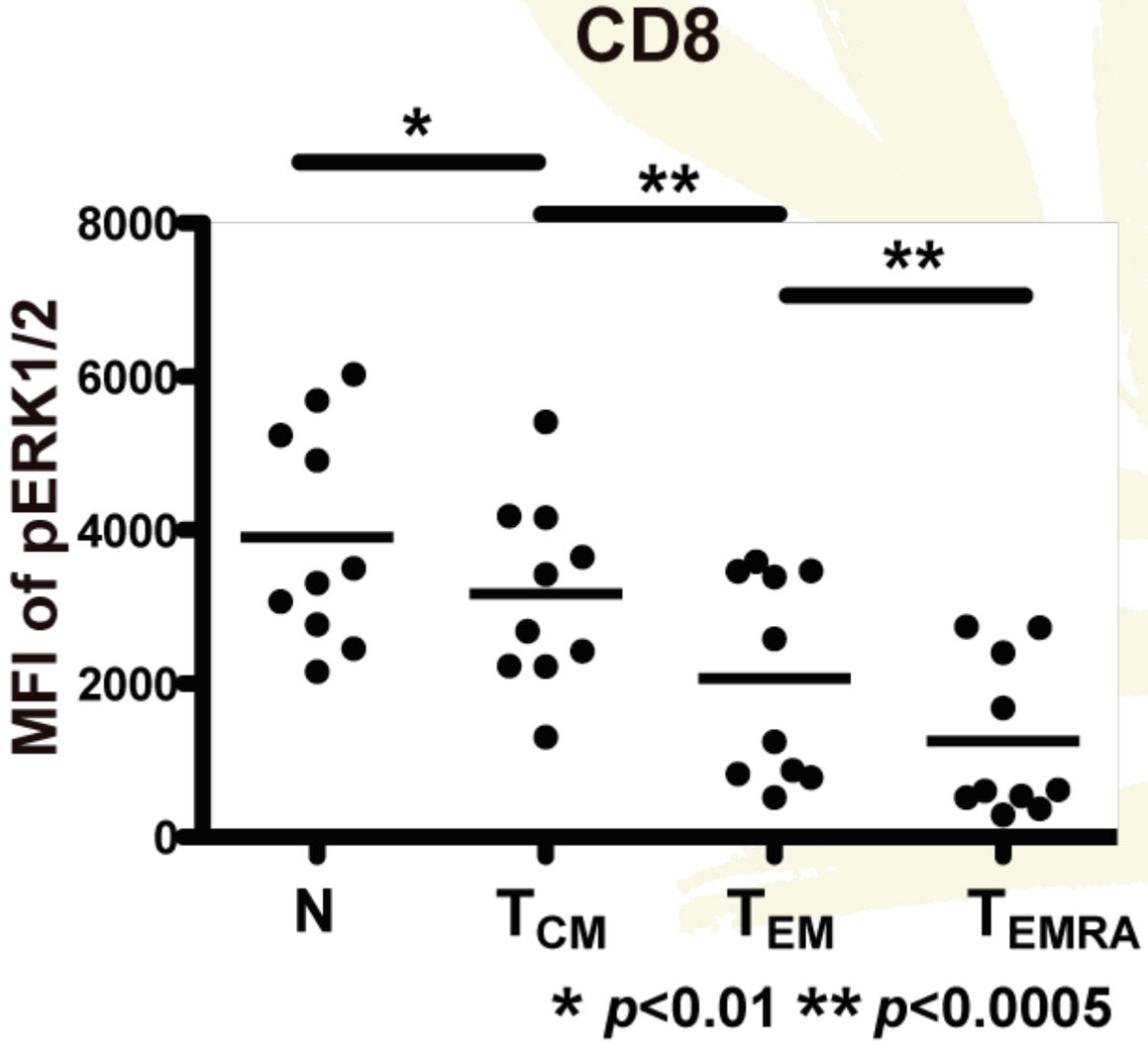
J Immunol 2009; 183:6167-6174; Prepublished online 19 October 2009;
doi: 10.4049/jimmunol.0902068
http://www.jimmunol.org/content/183/10/6167

TCR activation and the RAS/MEK/ERK pathway

T Cell Receptor Signaling



T cell signaling differs in "naive" and more mature T cells



Plenary Paper

TRANSPLANTATION BLOOD, 6 JUNE 2013 • VOLUME 121, NUMBER 23

MEK inhibitors selectively suppress alloreactivity and graft-versus-host disease in a memory stage-dependent manner

Takero Shindo,[1] Tae Kon Kim,[1] Cara L. Benjamin,[1] Eric D. Wieder,[1] Robert B. Levy,[2] and Krishna V. Komanduri[1,2]

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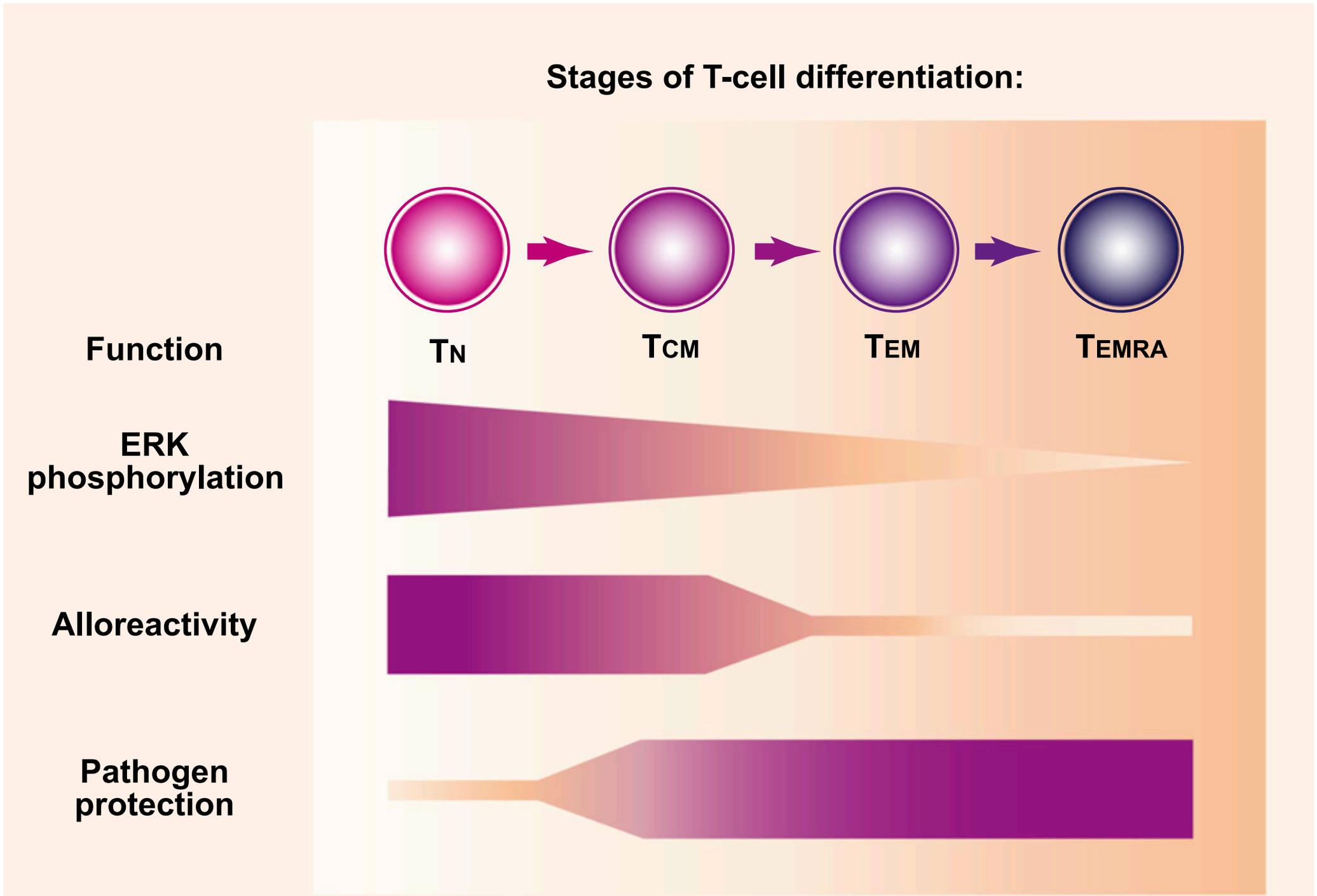


Professional illustration by Paulette Dennis

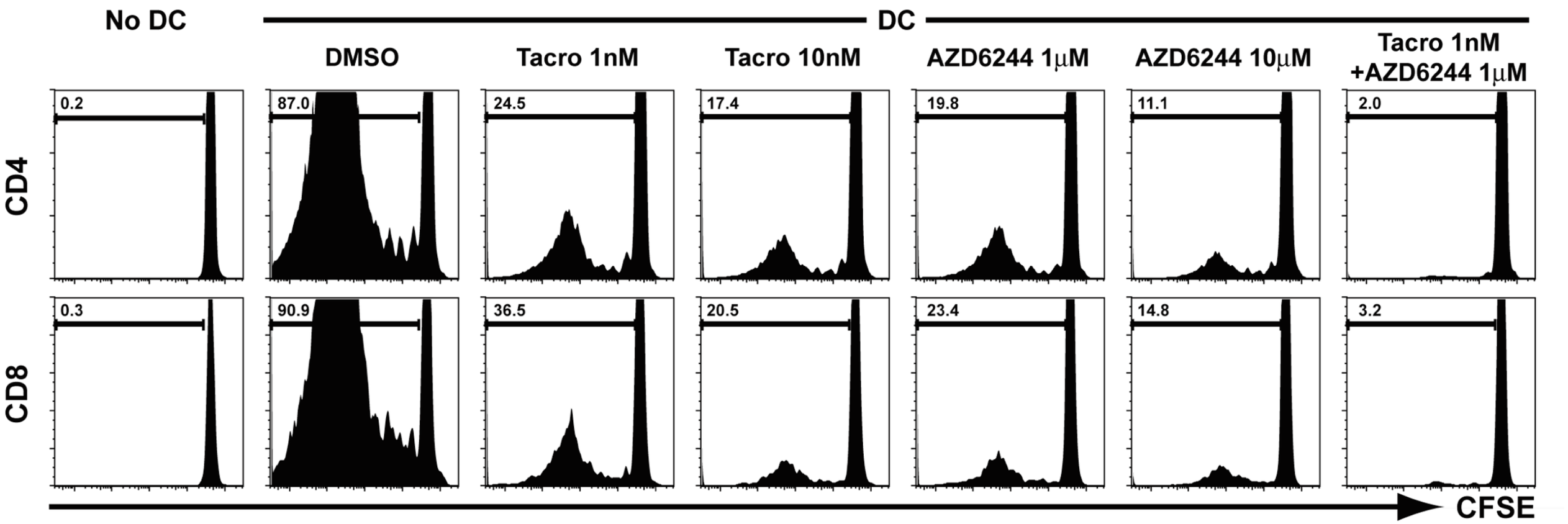
Comment on Shindo et al, page 4617

MEKing it easier to prevent GVHD

Paul J. Martin[1] [1]FRED HUTCHINSON CANCER RESEARCH CENTER

insideblood®
6 JUNE 2013 | VOLUME 121, NUMBER 23

Selumetinib inhibits alloreactivity, is synergistic with tacrolimus



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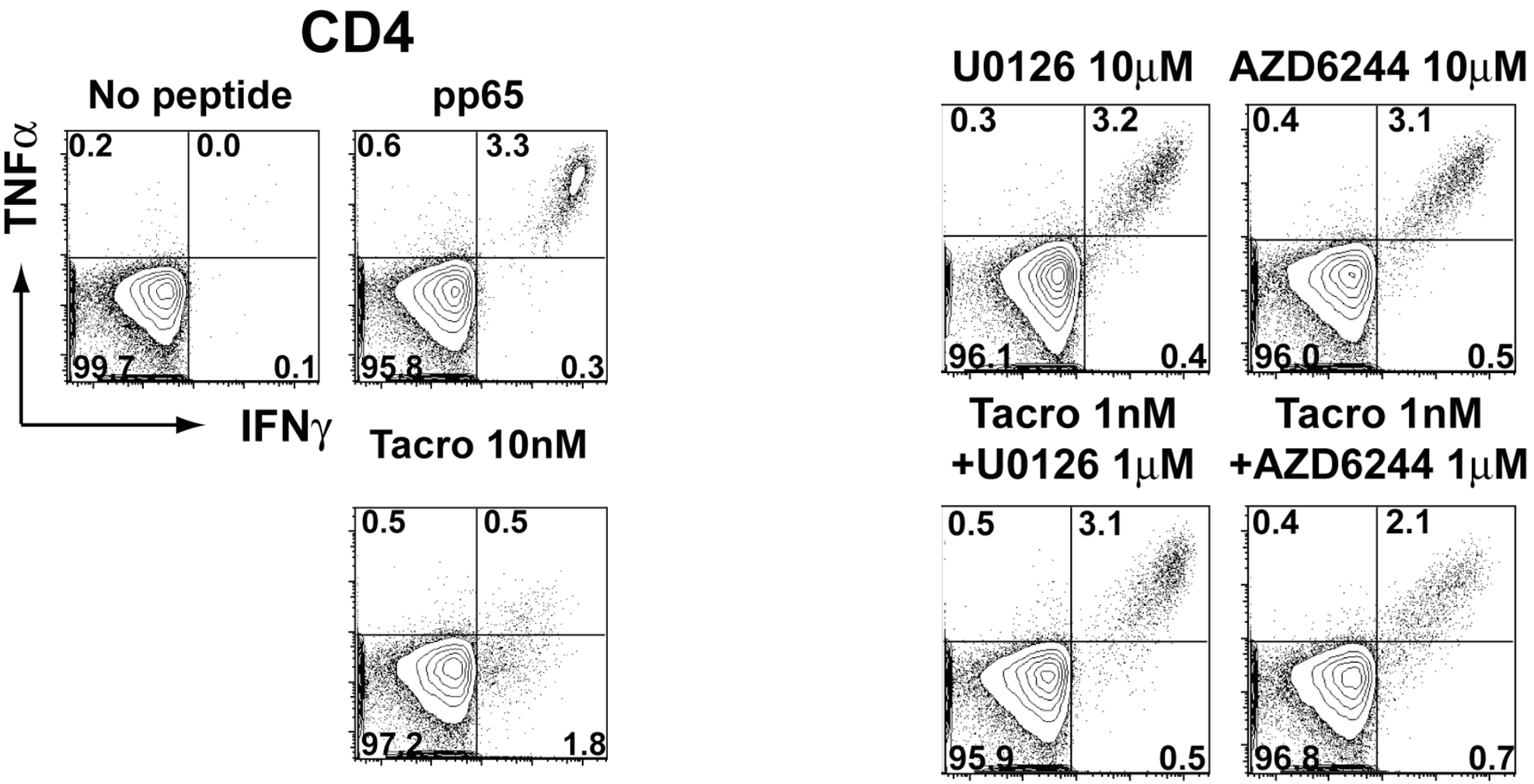
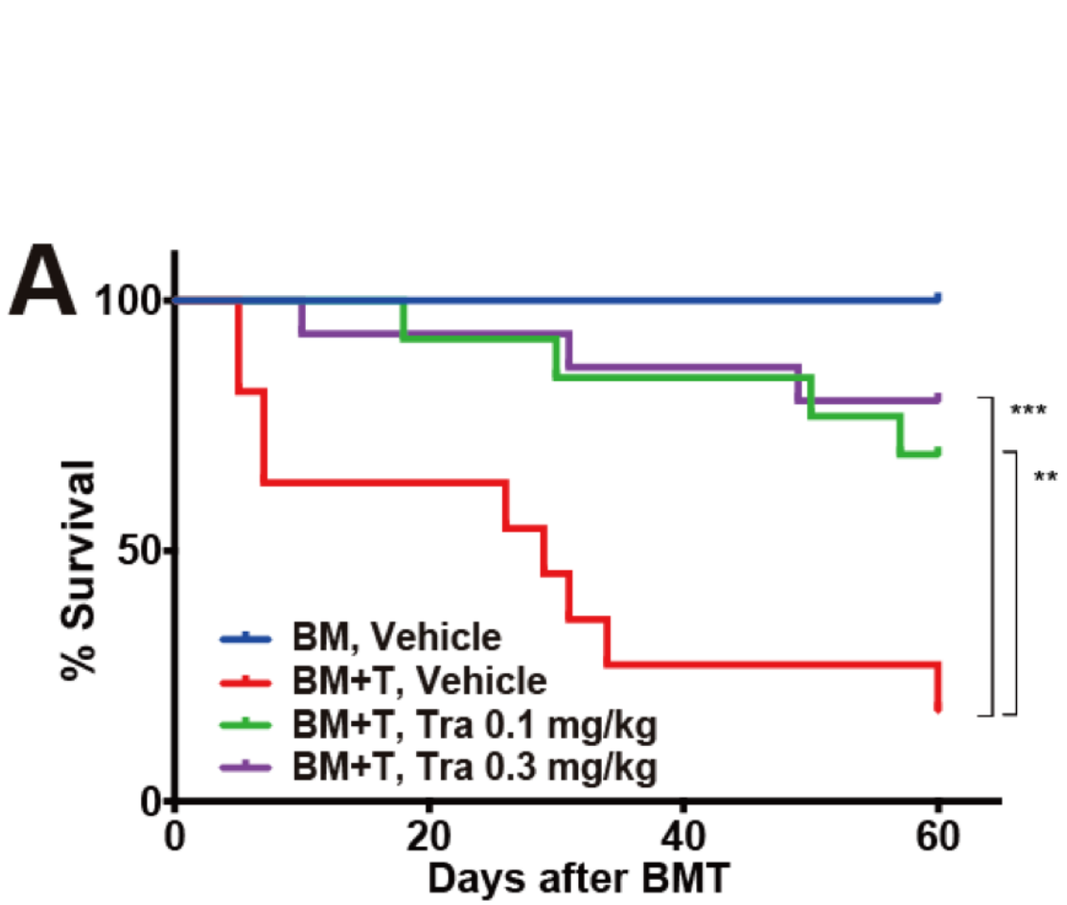
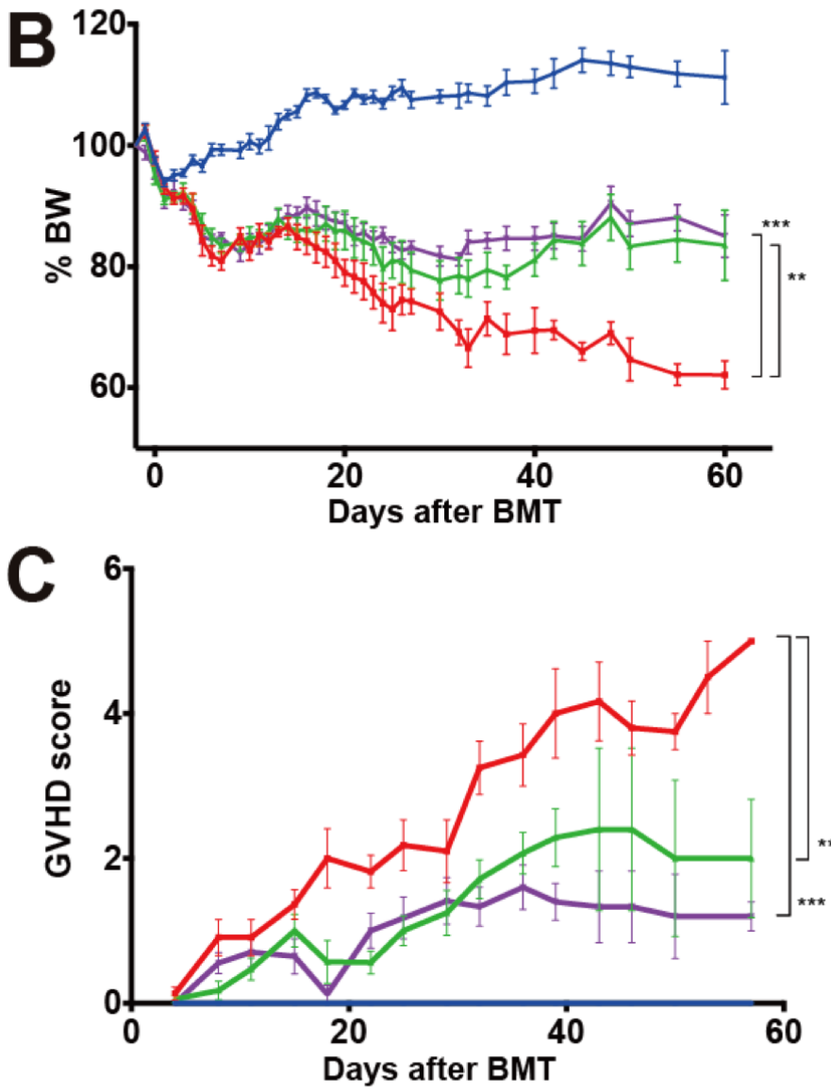
MEK inhibition spares CMV-specific polyfunctional T cells



Shindo, Kim, et al., Blood 2013

Trametinib also spares CMV immunity and inhibits GVHD *in vivo*





Itamura, Shindo, et al., *JCI Insight* 2016

Selective immunosuppression by MEK inhibition

- MEK inhibition may selectively target alloreactivity while sparing pathogen-specific immunity (CMV, EBV-specific T cells)
- Evidence of a class effect and potential synergy with CNI
- Immune recovery may be protected via more selective inhibition
- RAS/MEK/ERK signaling also important in some cancers (relapse?)

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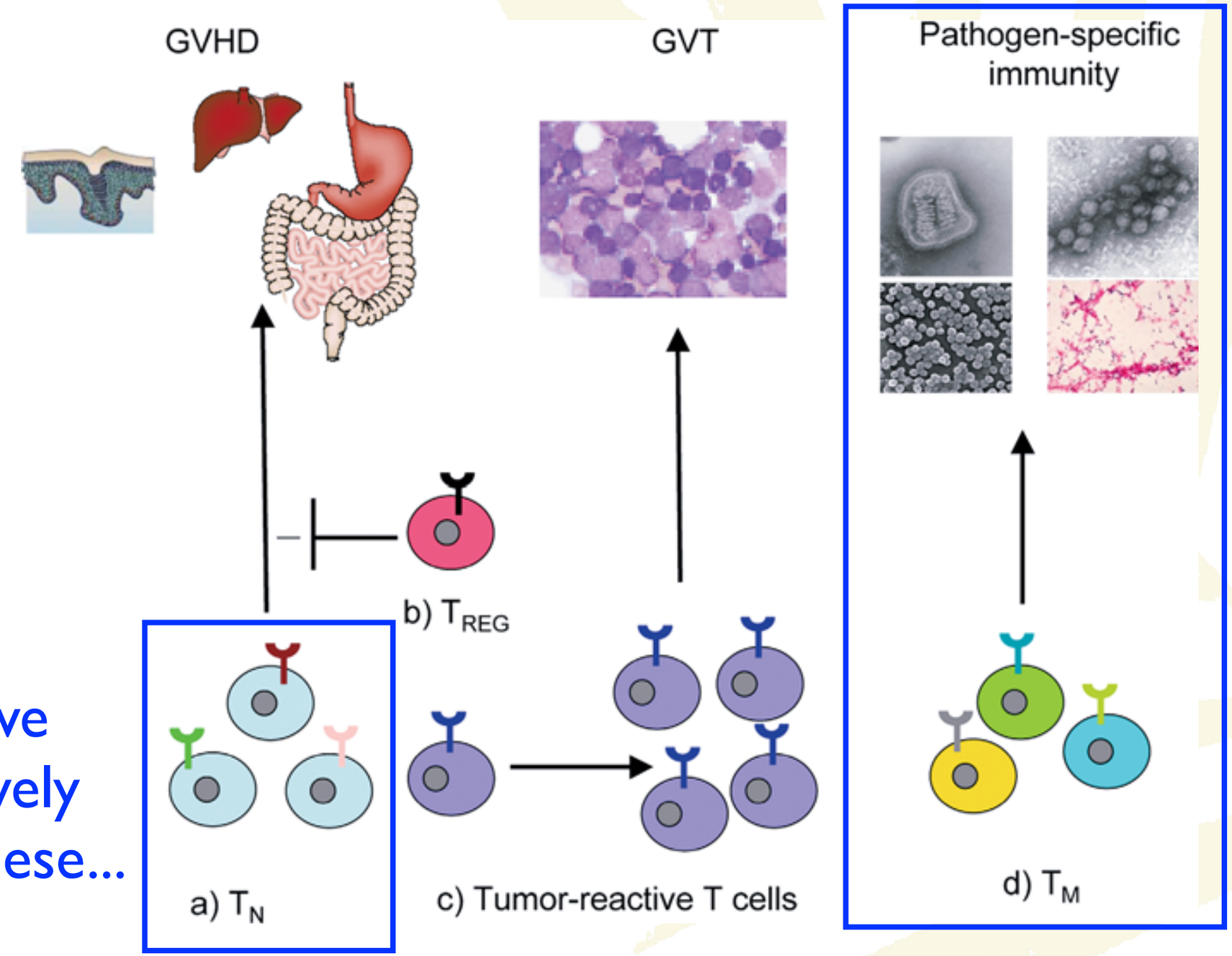
What have we learned from 20 years of CMV immunology?

- Pathogen-specific T cell responses could not rise above frequencies of 0.5-1% of the overall repertoire
  - Not true (routinely massively expanded in healthy and in patients)
- CMV reactivation most likely occurred in the absence of pathogen-specific T cells
  - True after CBT and TCD, but not in most SCT recipients
- Steroids are lympholytic and therefore eliminate CMV-specific T cells from the circulation
  - Not true (biggest issue is dysfunction of T cell responses)

A key challenge will be to develop better and more selective immunosuppression to facilitate pathogen-specific immune recovery

The goal: Eliminating GVHD while sparing beneficial T cells



...but not these?

Can we selectively inhibit these...

Graft-Versus-Host Disease: A Surge of Developments
Stanley R. Riddell, Frederick R. Appelbaum
PLoS MEDICINE
July 2007 | Volume 4 | Issue 7 | e198

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