10/2018

Flipped classroom micro 204: B cell activation and differentiation

1:10 – 1:40pm – Breakout groups (3) to prepare diagrams of 3 key steps in the B cell response; present these on the white board

1:40-2:15 – Discuss figures from 4 papers (below) that relate to B cell fate decision making; refine the diagrams based on the discussion

- please review the Introduction and Figures 1 and 2 of the Abbott et al. Immunity 2018 paper before class https://www.ncbi.nlm.nih.gov/pubmed/29287996
- this Perspective may also be helpful https://www.ncbi.nlm.nih.gov/pubmed/29343432

2:15-2:30 – Go over any remaining questions from the lecture

Breakout groups - 15 min
Prepare diagrams on white boards to show the following 3 stages of a T cell-dependent B cell response

Group 1: Initial activation of the B cell in a T-dependent immunization (showing protein-linked antigen, BCR signaling, activation/proliferation, antigen presentation, recruitment of T cell help)

Group 2: First decision the B cell makes; address short lived plasma cell vs germinal center (GC) vs death; mention class switch recombination

Group 3: Dynamics within the GC (cycling/selection/somatic hypermutation) and output of the GC (memory / long lived plasma cell)

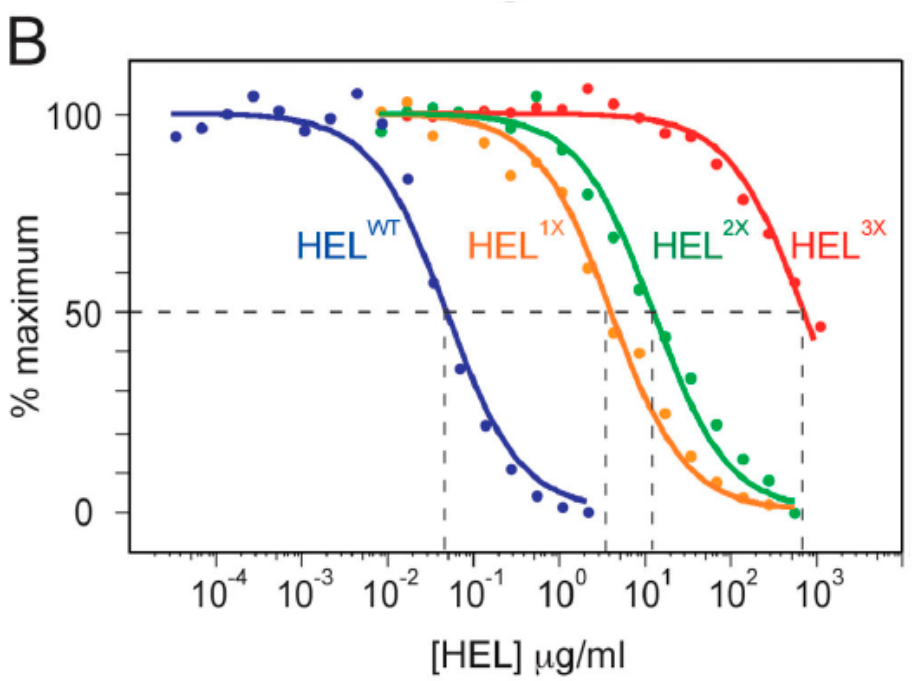
Antigen recognition strength regulates the choice between extrafollicular plasma cell and germinal center B cell differentiation

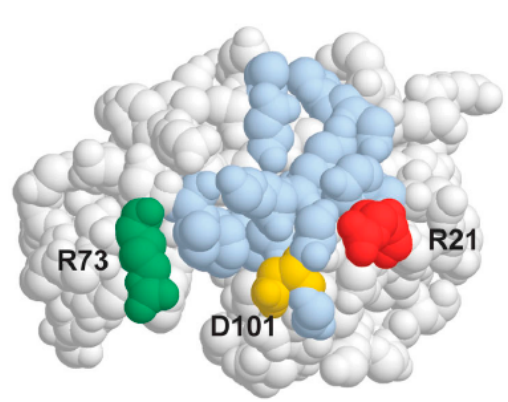
Didrik Paus,[1,2] Tri Giang Phan,[1,2] Tyani D. Chan,[1,2] Sandra Gardam,[1,2] Antony Basten,[1,2] and Robert Brink[1,2]

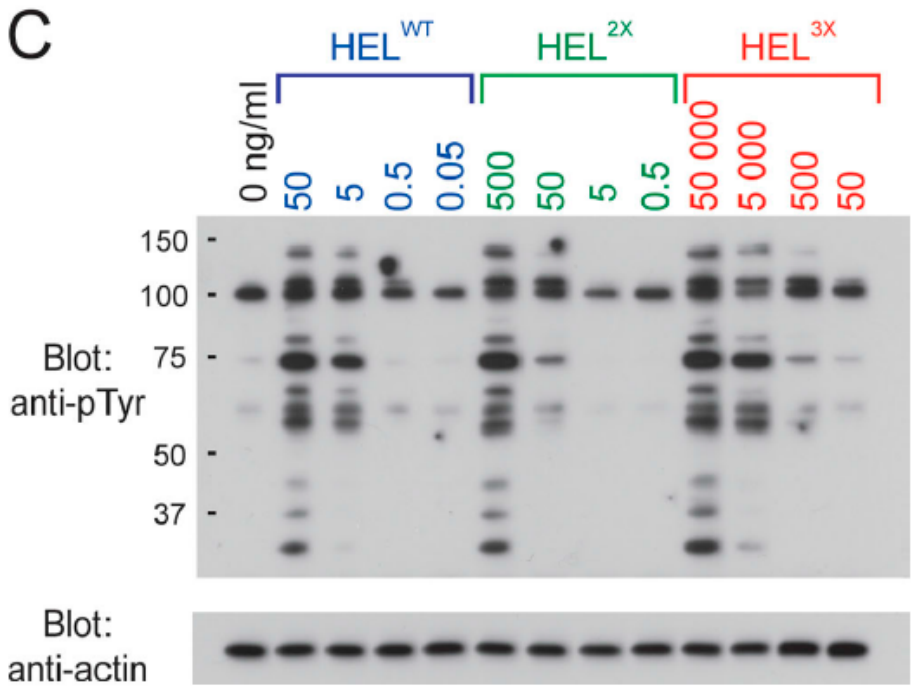
JEM © The Rockefeller University Press $8.00
Vol. 203, No. 4, April 17, 2006 1081–1091 www.jem.org/cgi/doi/10.1084/jem.20060087

Paus et al. JEM 2006 Fig 1. *Titrating Ag affinity*

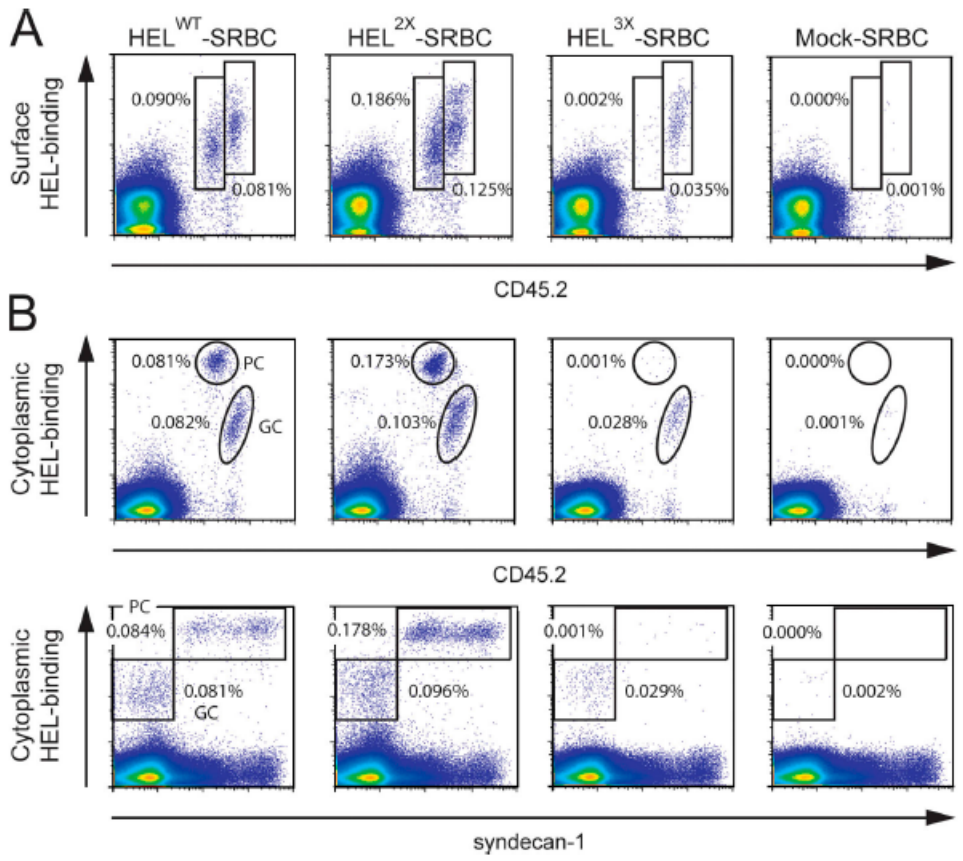
2×10^{10} M^{-1} (HELWT)
8×10^{7} M^{-1} (HEL2X = R73E, D101R)
1.5×10^{6} M^{-1} (HEL3X = R73E, D101R, R21Q)

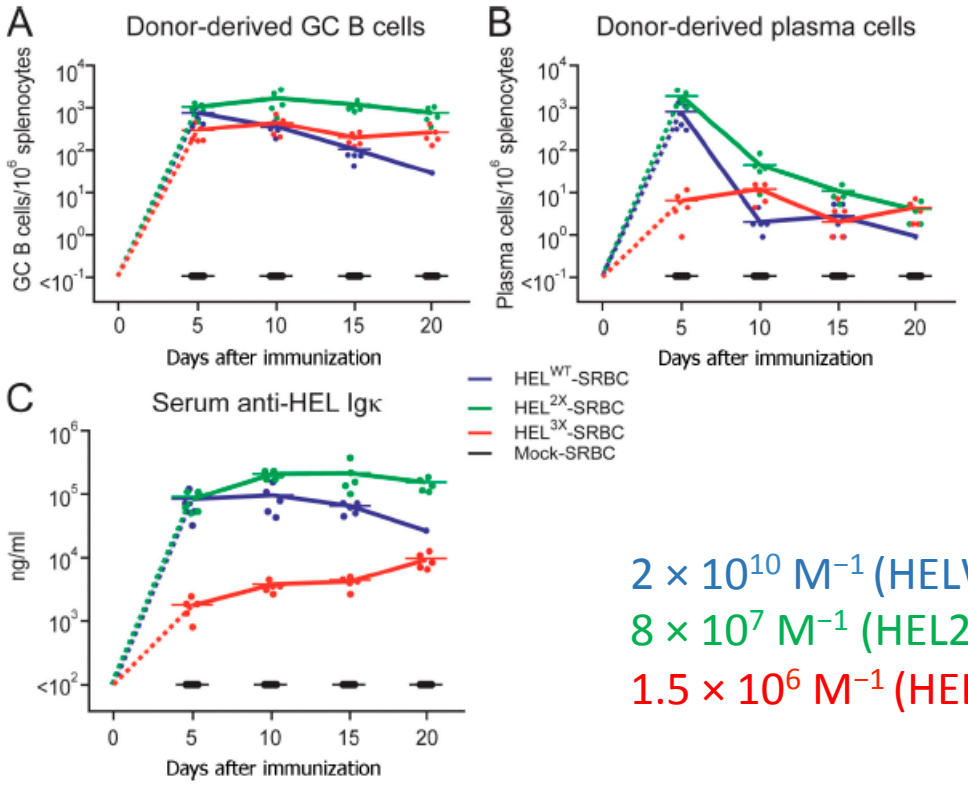






Paus et al. JEM 2006 Fig 4 / 5. *SLPC but not GC requires high affinity Ag*





2×10^{10} M^{-1} (HELWT)
8×10^{7} M^{-1} (HEL2X)
1.5×10^{6} M^{-1} (HEL3X)

Similar trends if you titrate antigen density or BCR affinity!

GC is insensitive to, but SLPC generation depends on:

-antigen AFFINITY for BCR
-antigen density
-BCR AFFINITY for Ag

Is it strange that gc response seems insensitive to ag-BCR interaction “strength”? What purpose would this serve?

What are the implications for vaccine design?

What are the possible mechanisms for these observations?

How could you test them?

-BCR signal strength could affect:

- -proliferation
- -survival
- -migration
- -differentiation (e.g. fate-defining transcription factor expression)

-Signal-independent function of the BCR to capture and present Ag?

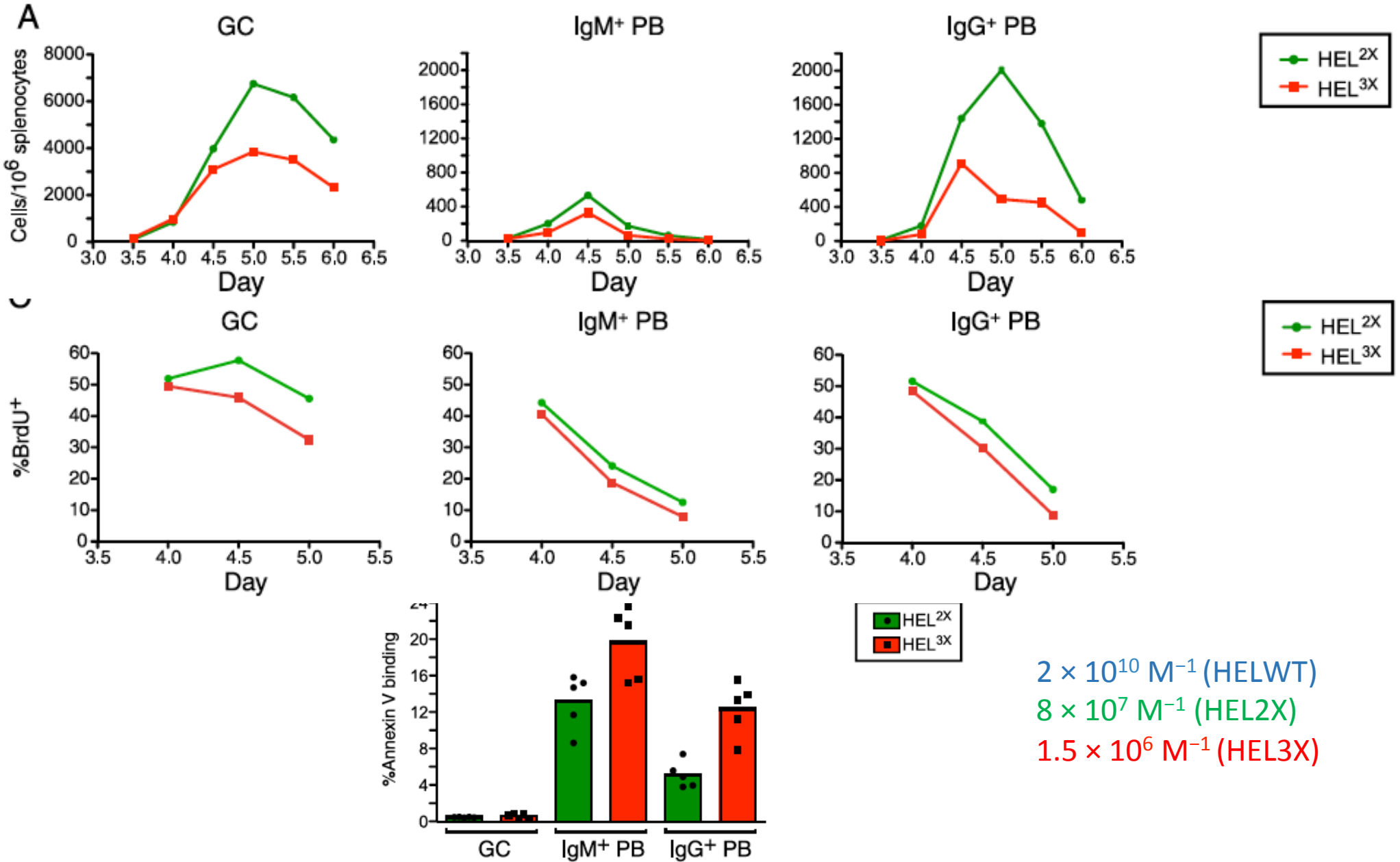
Chan et al. JI 2009

Antigen Affinity Controls Rapid T-Dependent Antibody Production by Driving the Expansion Rather than the Differentiation or Extrafollicular Migration of Early Plasmablasts

Tyani D. Chan, Dominique Gatto, Katherine Wood, Tahra Camidge, Antony Basten and Robert Brink

J Immunol 2009; 183:3139-3149; Prepublished online 7 August 2009;
doi: 10.4049/jimmunol.0901690

Chan et al. JI 2009 Fig 7. *Antigen affinity regulates proliferation and death*



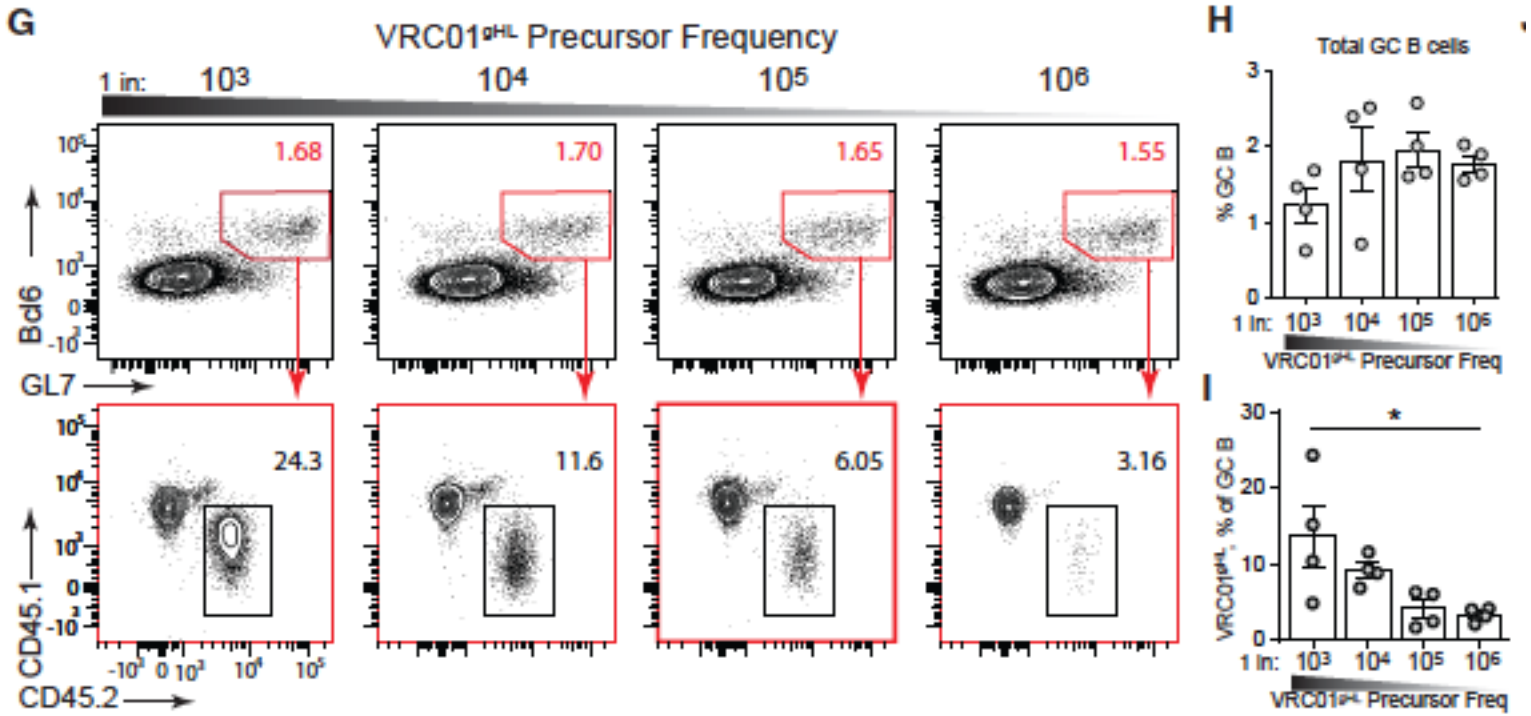
2×10^{10} M^{-1} (HELWT)
8×10^{7} M^{-1} (HEL2X)
1.5×10^{6} M^{-1} (HEL3X)

Precursor Frequency and Affinity Determine B Cell Competitive Fitness in Germinal Centers, Tested with Germline-Targeting HIV Vaccine Immunogens

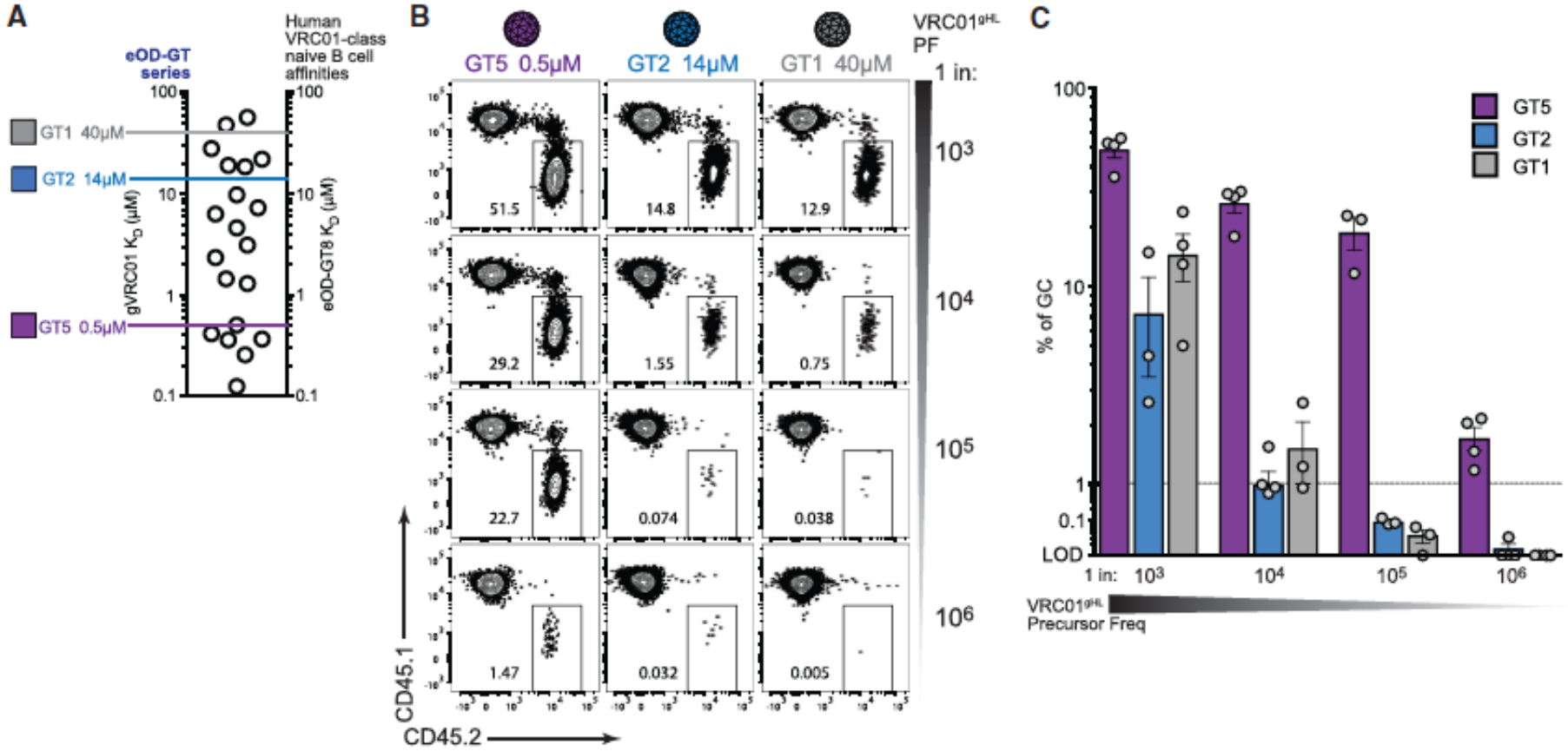
Robert K. Abbott,[1,2] Jeong Hyun Lee,[1,2] Sergey Menis,[2,3] Patrick Skog,[4] Meghan Rossi,[1,2] Takayuki Ota,[4] Daniel W. Kulp,[2,3,5] Deepika Bhullar,[4] Oleksandr Kalyuzhniy,[2,3] Colin Havenar-Daughton,[1,2] William R. Schief,[2,3,4,6,*] David Nemazee,[4,*] and Shane Crotty[1,2,7,8,*]

Immunity *48*, 133–146, January 16, 2018

Abbott et al. Immunity 2018 Fig 1. precursor frequency regulates competitive entry into the GC



Abbott et al. Immunity 2018 Fig 2. precursor frequency and antigen affinity each titrate GC entry – at “high” precursor frequency, GC entry is less sensitive to affinity!



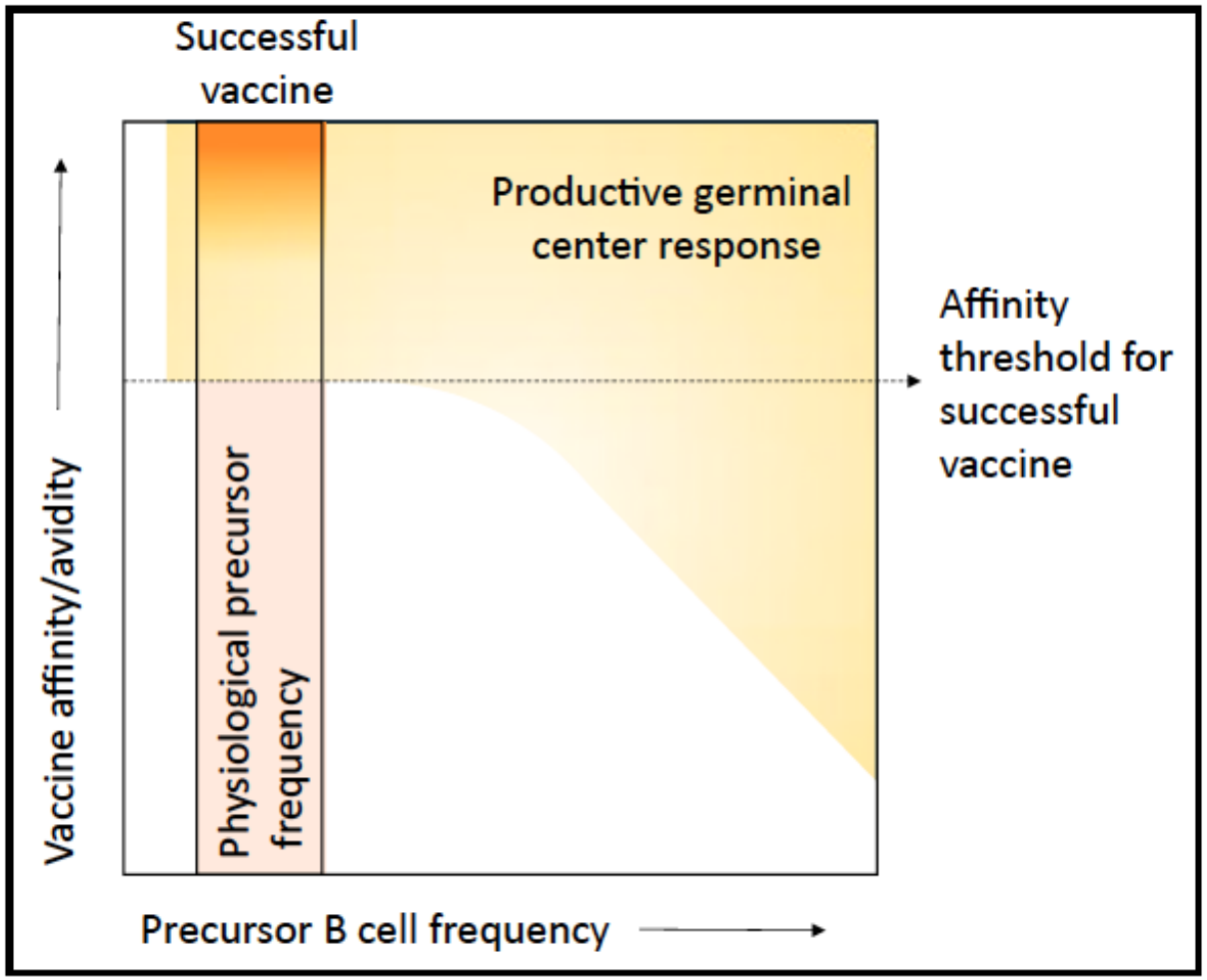


Figure 1. Diagram Depicting the Response of Varying Numbers of Precursor B Cells Capable of Producing bNAbs to HIV Constructs that Stimulate the B Cells with Varying Affinities

Formation of germinal centers in which the precursor B cells can outcompete other endogenous B cells allow acquisition of the desired somatic mutations over time. At the physiologically low precursor frequencies found in humans, an affinity threshold appears essential for a successful immunogen.

Anti–HIV-1 B cell responses are dependent on B cell precursor frequency and antigen-binding affinity

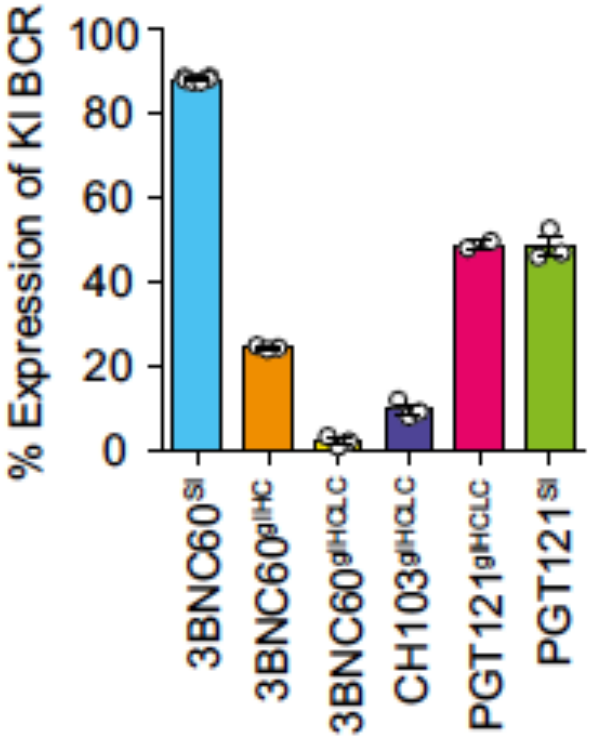
Pia Dosenovic[a], Ervin E. Kara[a], Anna-Klara Pettersson[a], Andrew T. McGuire[b], Matthew Gray[b], Harald Hartweger[a], Eddy S. Thientosapol[a], Leonidas Stamatatos[b], and Michel C. Nussenzweig[a,c,1]

PNAS | May 1, 2018 | vol. 115 | no. 18 | 4743–4748

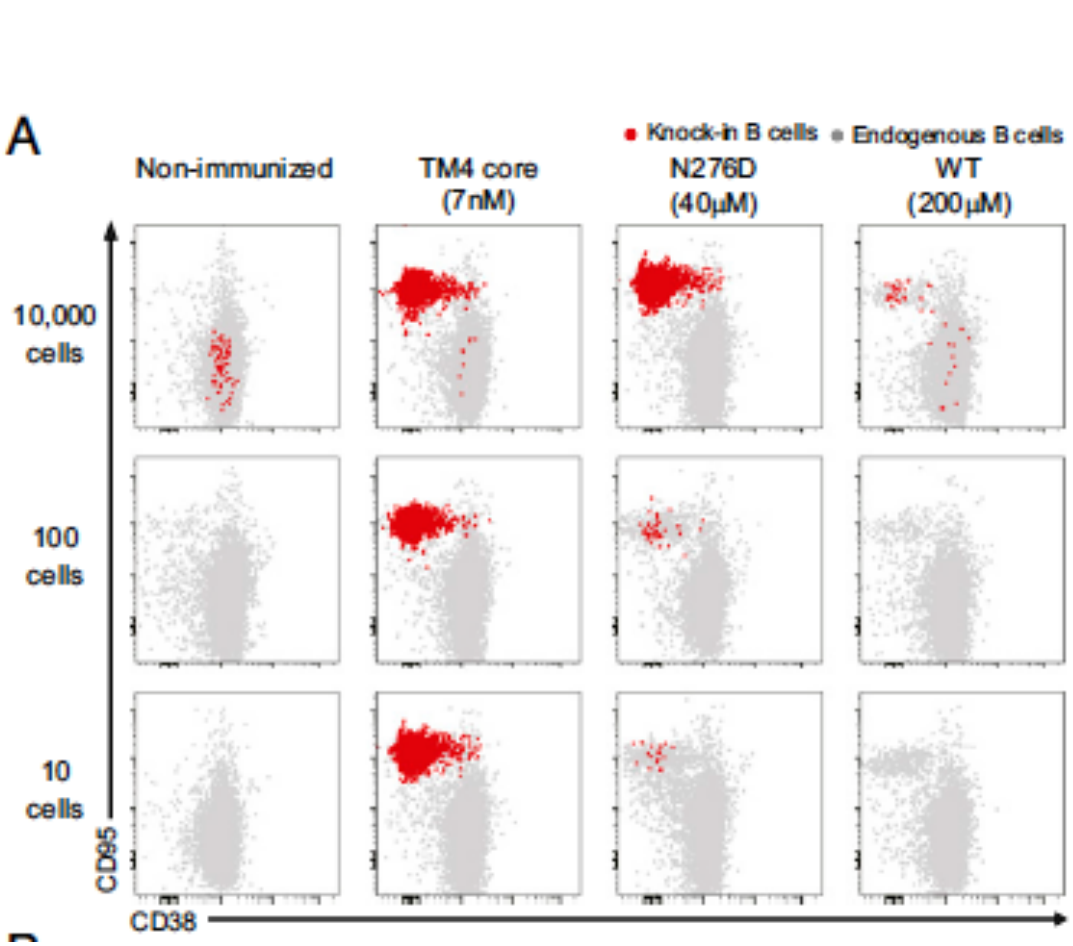
Prior BCR Tg generated from HIV bNabs: F5, 4E10, 3BNC60 germline, and VRC01 germline show autoreactivity and trigger tolerance mechanisms… this may affect response to immunization

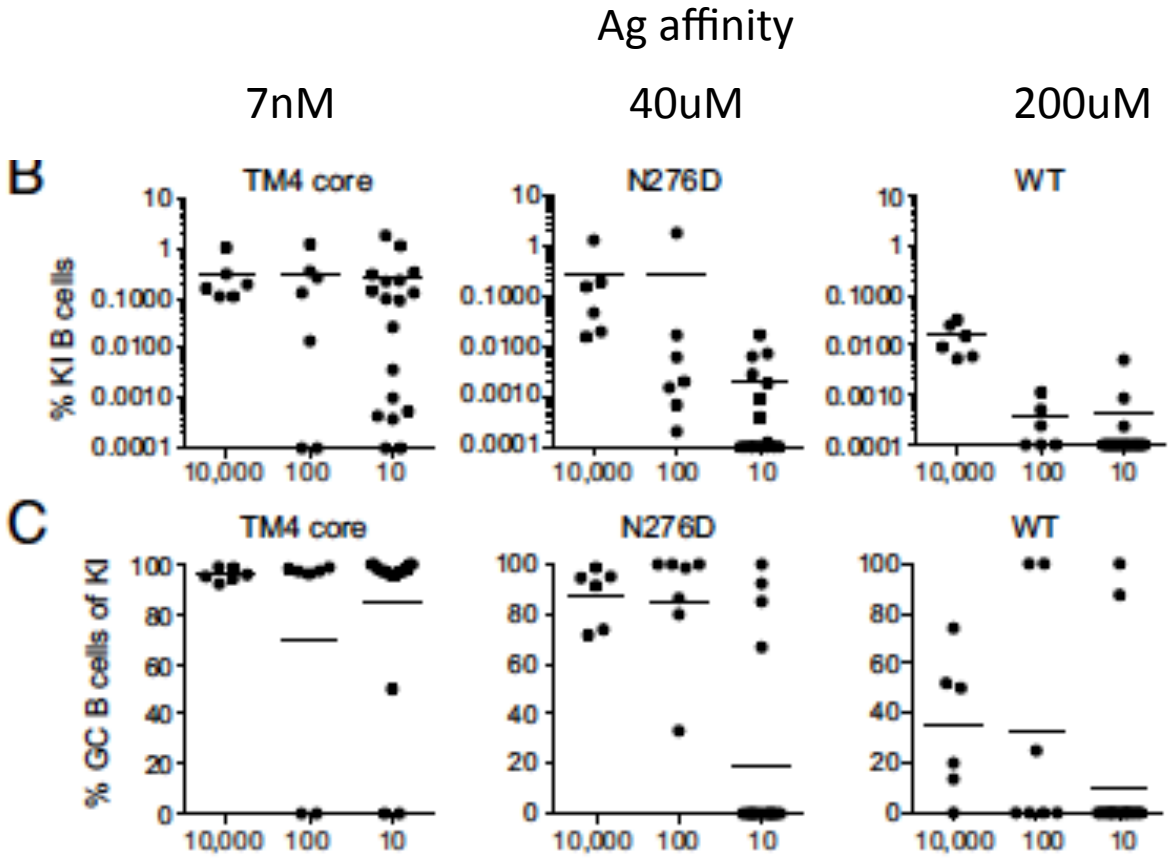
Other bNAb BCR Tgs include 3BNC60, CH103 and PGT12

3BNC60^{SI} used in this paper: HC is mutated but light chain is reverted to germline : NOT autoreactive



Dosenovic et al. PNAS 2018 Fig 4. GC size regulated by precursor frequency and antigen affinity





GC generation is in fact sensitive to antigen affinity unlike report from Paus et al, where only SLPC generation seems to be sensitive to antigen affinity. How can you reconcile these data?

What effect might a really high affinity/avidity ligand be expected to have on SLPC responses, and how might that be relevant for GC responses?

What is the significance of precursor frequency? By what mechanism could competition with other clones affect GC entry?

What might be the impact of self-reactivity of the precursor clone on response to foreign antigens?

How do all these considerations determine optimal vaccine design?

Questions posed during taped lecture for discussion

- What is the frequency of B cells specific for a typical viral antigen?
- What sets the number of B cells?
- Flu and HIV antigens are heavily glycosylated. Can we make antibodies against carbohydrate antigens? Can we make them against the ones on viruses? How?
- Do we want ideal vaccines to drive a T-independent or T-dependent response?
- Do viruses promote the B cell response solely be engaging the BCR or can other receptors on the B cell be involved? Which ones?
- Ultimately the vaccine needs to induce plasma cell differentiation. Does it matter what type of plasma cell is induced?
- Which Ig isotype would be best for an anti-Flu response? Or anti-HIV?
- What is meant by antibody-mediated opsonization? How does this differ from complement-mediated opsonization?
- Why do infants get repeated rounds of a vaccine within months of each other?
- Why do we get boosters of some vaccines and not others?
- How does pre-existing antibody against one part of the virus affect the ability to make antibody against another?