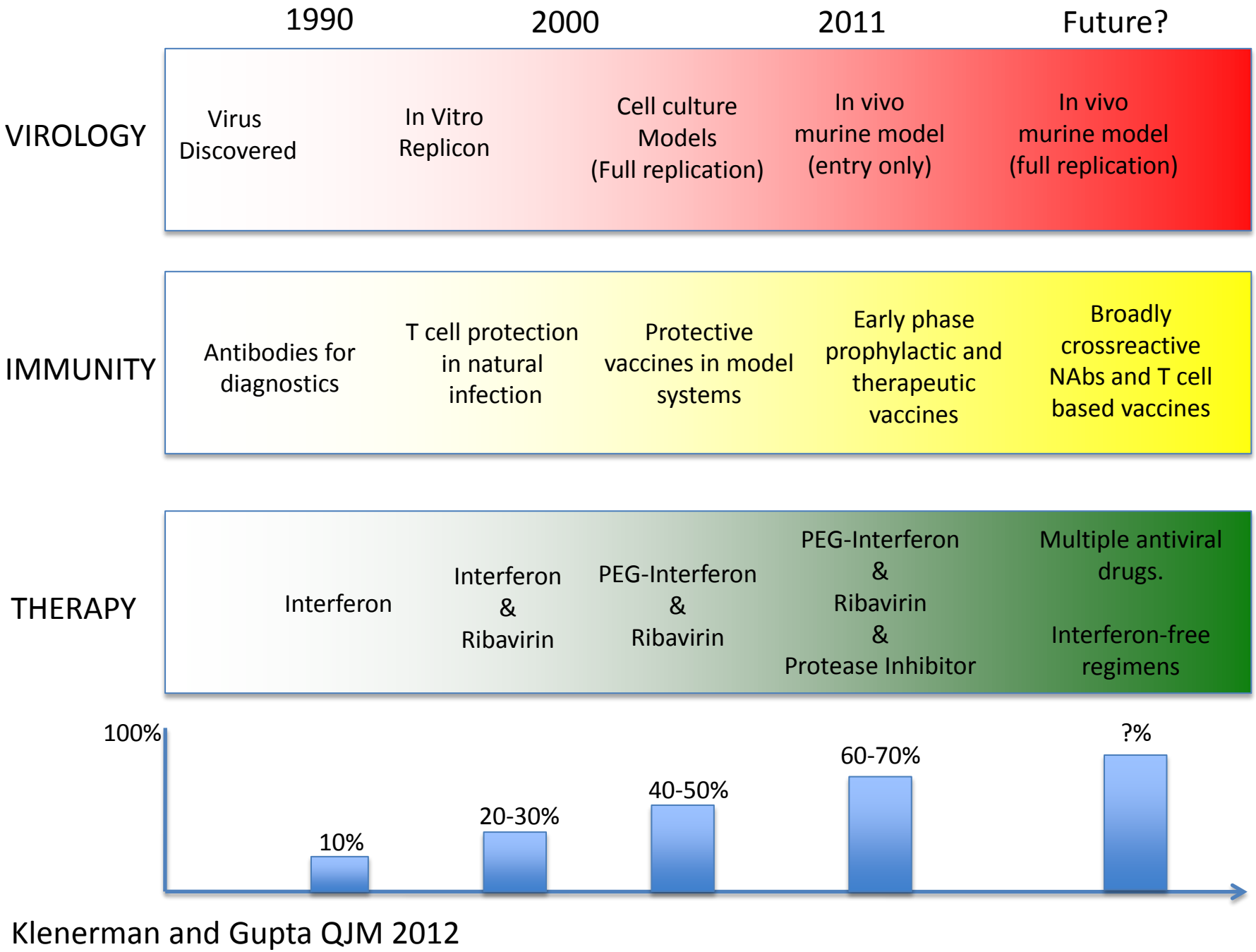
T cell memory and adenoviral vaccines

Paul Klenerman
University of Oxford

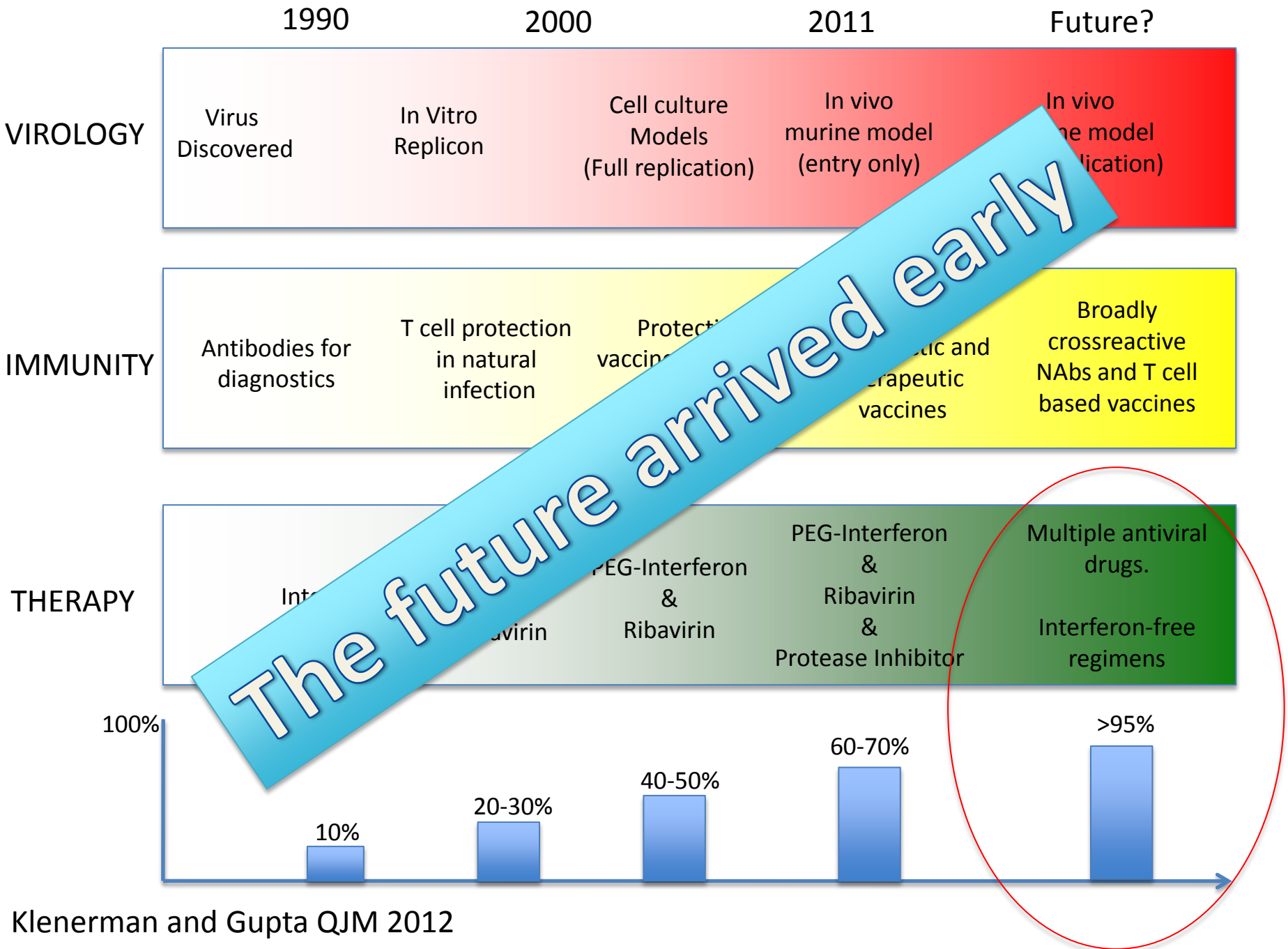
Talk Outline

- Background on chronic hepatitis C
- Development of T cell vaccines for HCV
- Adeno-vectored vaccines vs CMV

- How can we induce T cells to protect against complex infections?



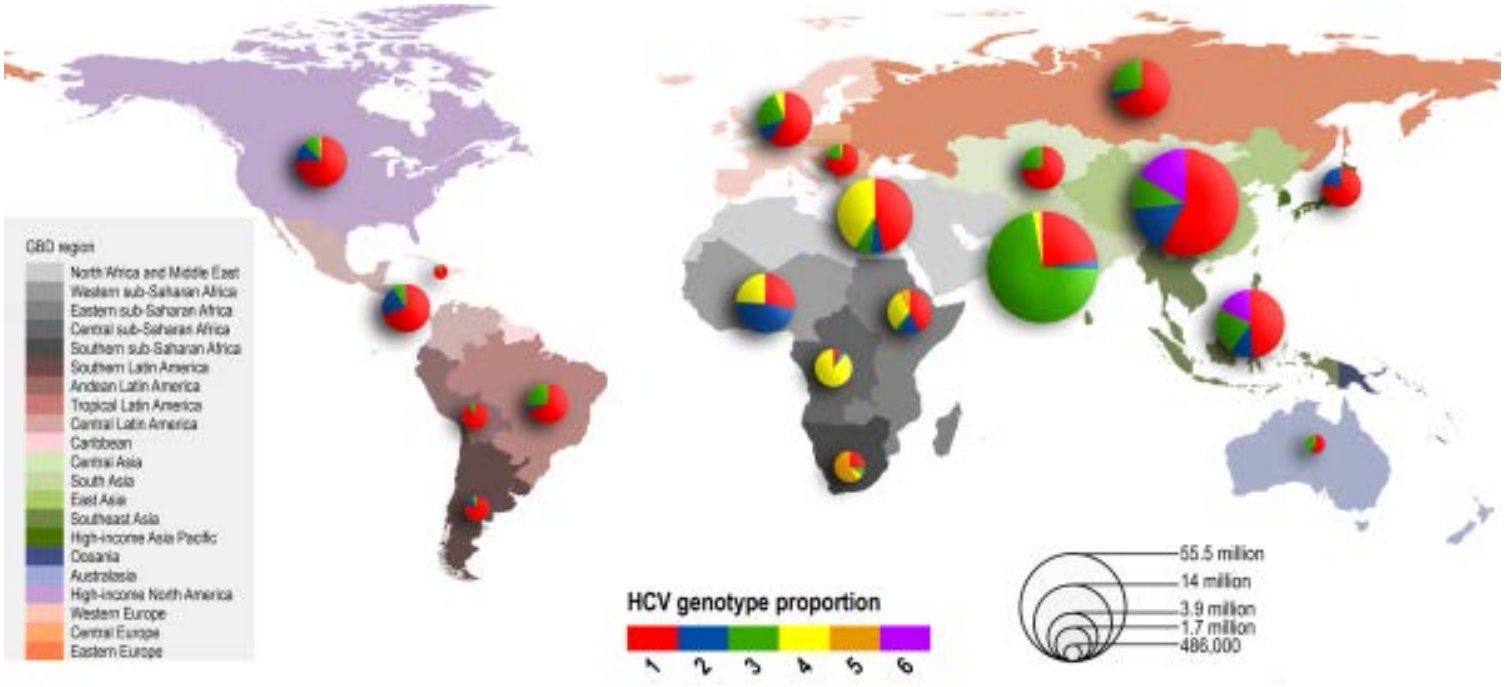
Klenerman and Gupta QJM 2012



Klenerman and Gupta QJM 2012

HCV is widespread and genetically diverse

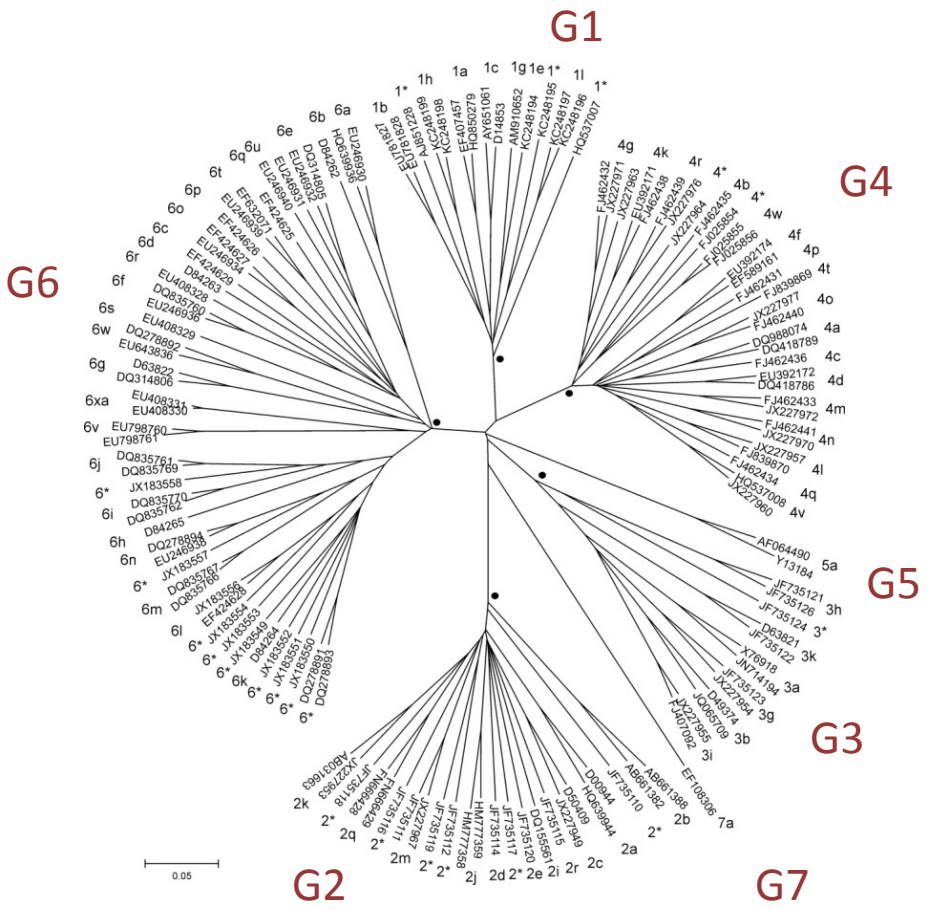
- 185 million people infected with HCV worldwide.
- Many develop liver cirrhosis or liver cancer leading to ~500,000 deaths/yr.
- HCV has 7 genotypes, 6 of which are common.
- Each responds differently to treatments and vaccines.



Messina, Humphreys, Flaxman, Brown, Cooke, Pybus and Barnes. (2014) Global distribution and prevalence of hepatitis C virus genotypes. Hepatology. doi: 10.1002/hep.27259

Genotypes and sub-genotypes

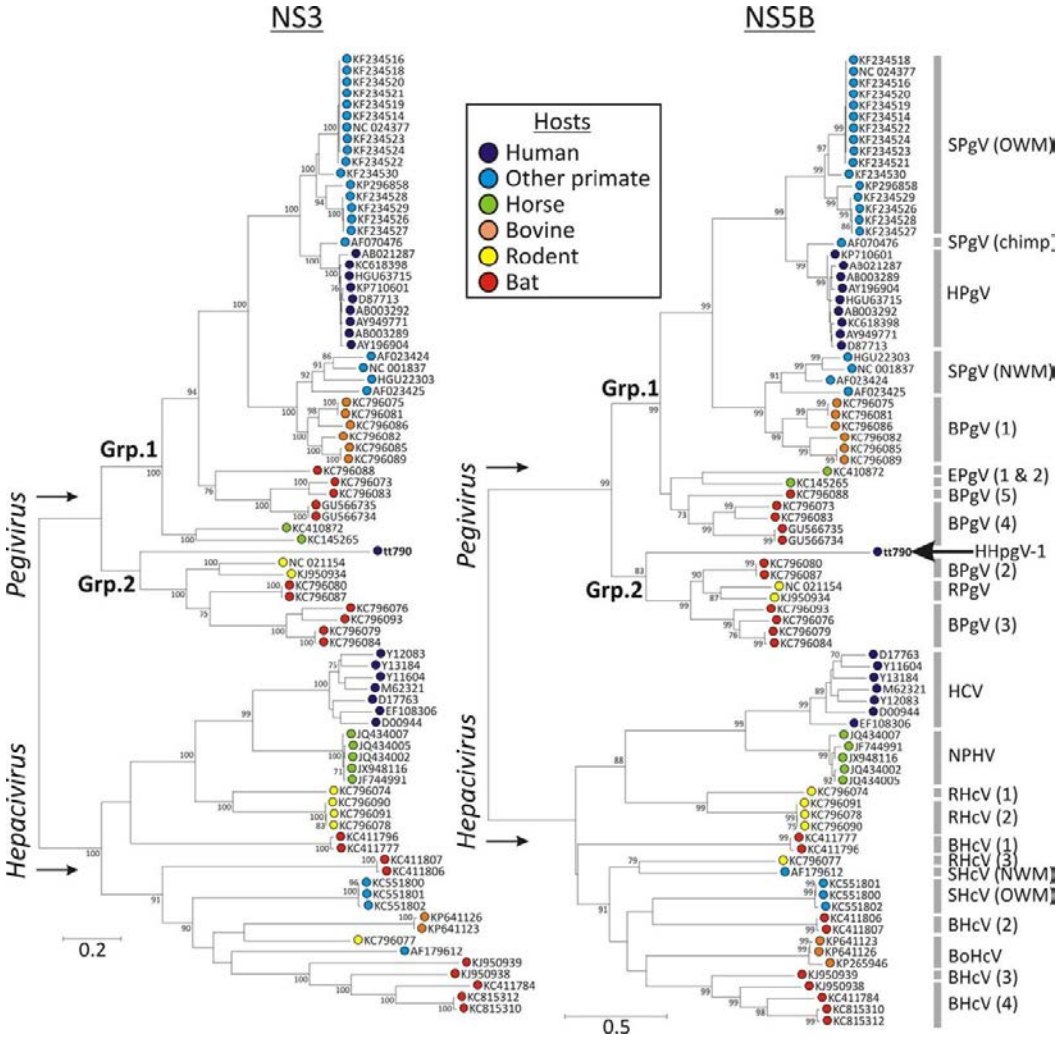
- HCV has an error-prone RNA-dependent RNA polymerase.
- The mutation rate is very high: 2.5 mutations per genome replication[1].
- There are 7 genotypes, G1 to G7, which differ by 30-35%[2].
- Within these genotypes, there are 67+ subtypes that differ by up to 20%[2].



[1] Ribeiro et al, (2012). Quantifying the diversification of Hepatitis C Virus (HCV) during primary infection: estimates of the in vivo mutation rate. PLoS Pathogens doi: 10.1371/journal.ppat.1002881

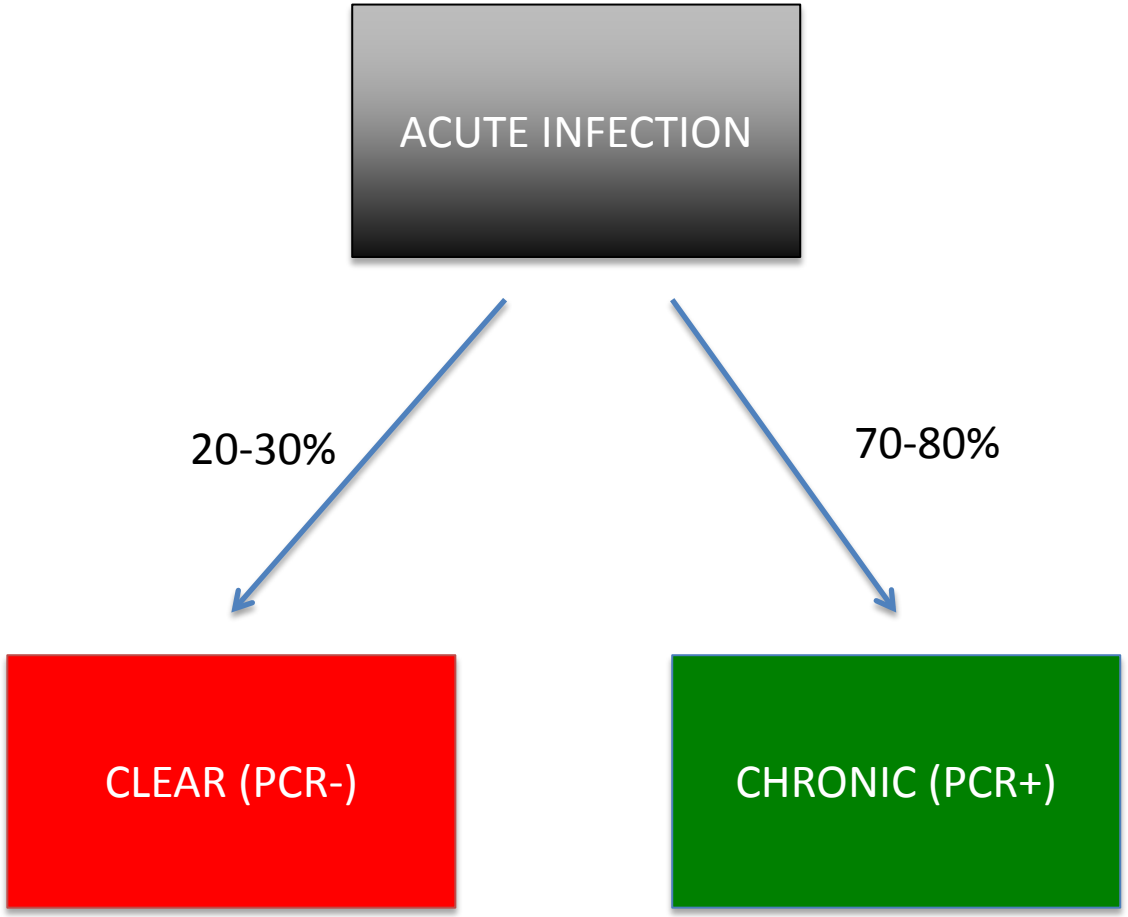
[2] Smith, Bukh, Kuiken, Muerhoff, Rice, Stapleton and Simmonds (2014). Expanded classification of hepatitis C virus into 7 genotypes and 67 subtypes: updated criteria and genotype assignment Web resource. Hepatology 59:318-327

Hepaciviruses and pegiviruses infecting different mammalian species.



Amit Kapoor et al. mBio 2015; doi:10.1128/mBio.01466-15

HCV Immunology
Genetic associations with clinical outcome



Genetic associations with clinical outcome

Explanatory Variables	OR and confidence limits	p-value
A*03	0.36 (0.15-0.89)	0.027
B*27	0.12 (0.03-0.45)	<0.001
DRB1*04:01	0.31 (0.12-0.85)	0.022
DRB1*01:01	0.2 (0.07-0.61)	0.005
DQB1*02:01	4.2 (2.04-8.66)	0.008
IFNL3 CC v T+	0.1 (0.04-0.23)	<0.001
KIR2DS3	4.36 (1.62, 11.74)	0.004

Genetic associations from a study of >300 Irish women Infected from a single source

Fitzmaurice et al Gut 2014

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HLA genes driving T cell Immunity

Fitzmaurice et al Gut 2014

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KIR genes driving NK cell Immunity

Fitzmaurice et al Gut 2014

Update on Immunology
Genetic associations with clinical outcome

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Interferon lambda gene(s) driving Innate Immunity

Fitzmaurice et al Gut 2014

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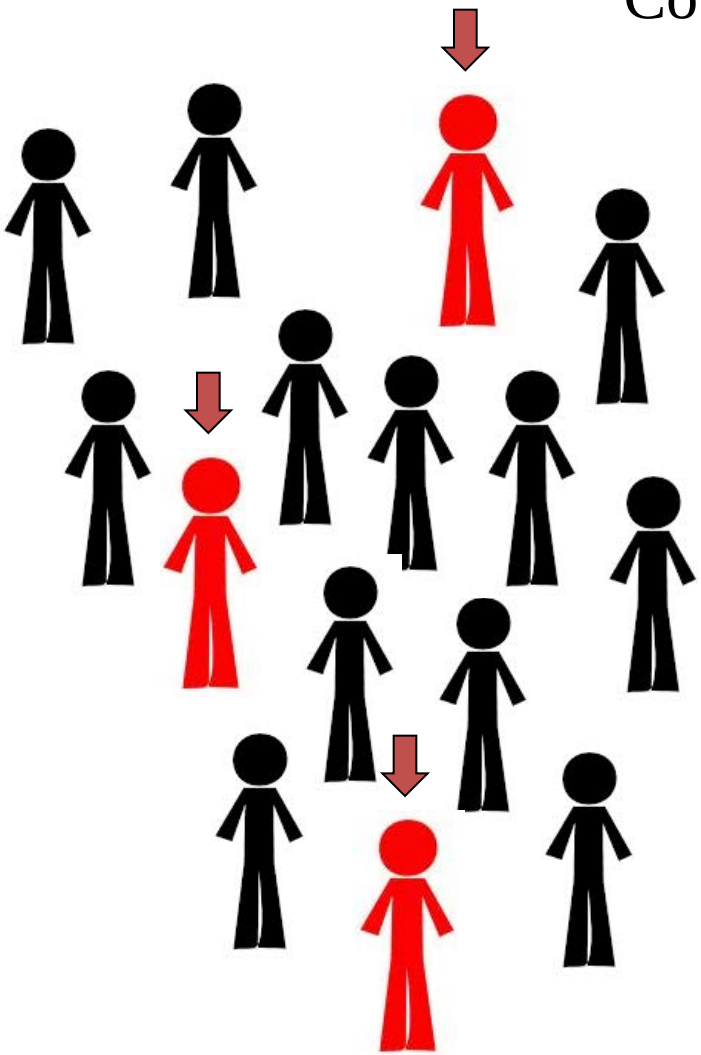
Interferon lambda gene(s) driving Innate Immunity

Large GWAS studies
also show HLA locus and IFN lambda association

Rauch et al Gastro 2010
Thomas et al Nature 2009
Duggal et al Ann Int Med 2013

Fitzmaurice et al Gut 2014

Is it worth worrying about immunity and vaccines?



Couldn’t we just treat people as needed?

Needs:

1. To identify all those infected
2. A therapy that works for all

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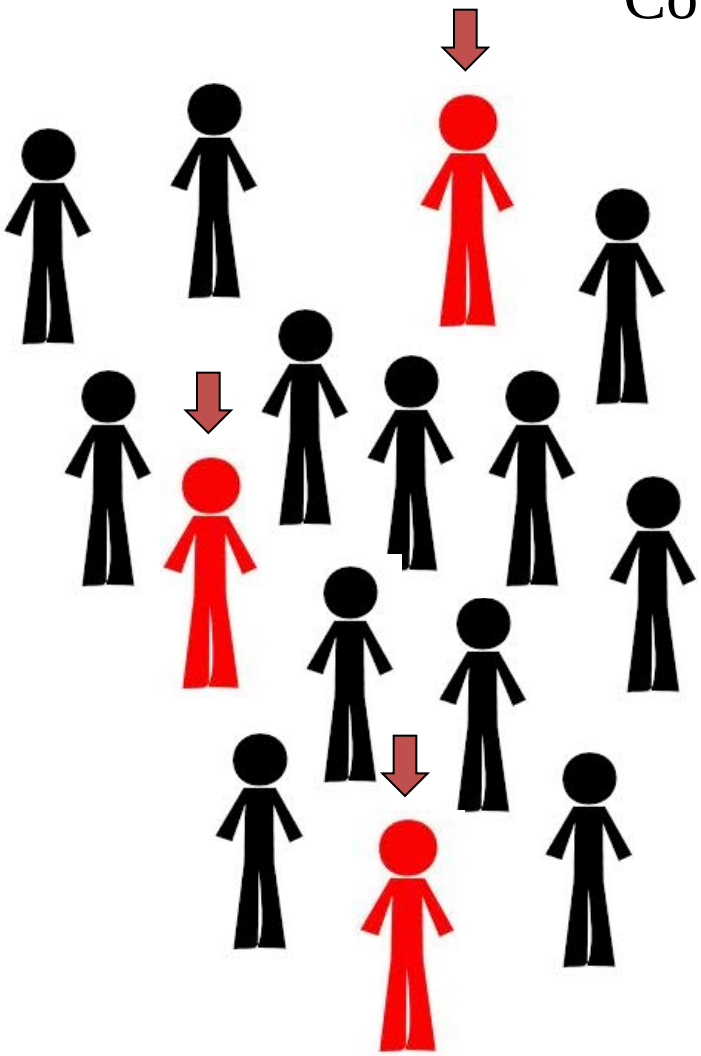

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Getting Close

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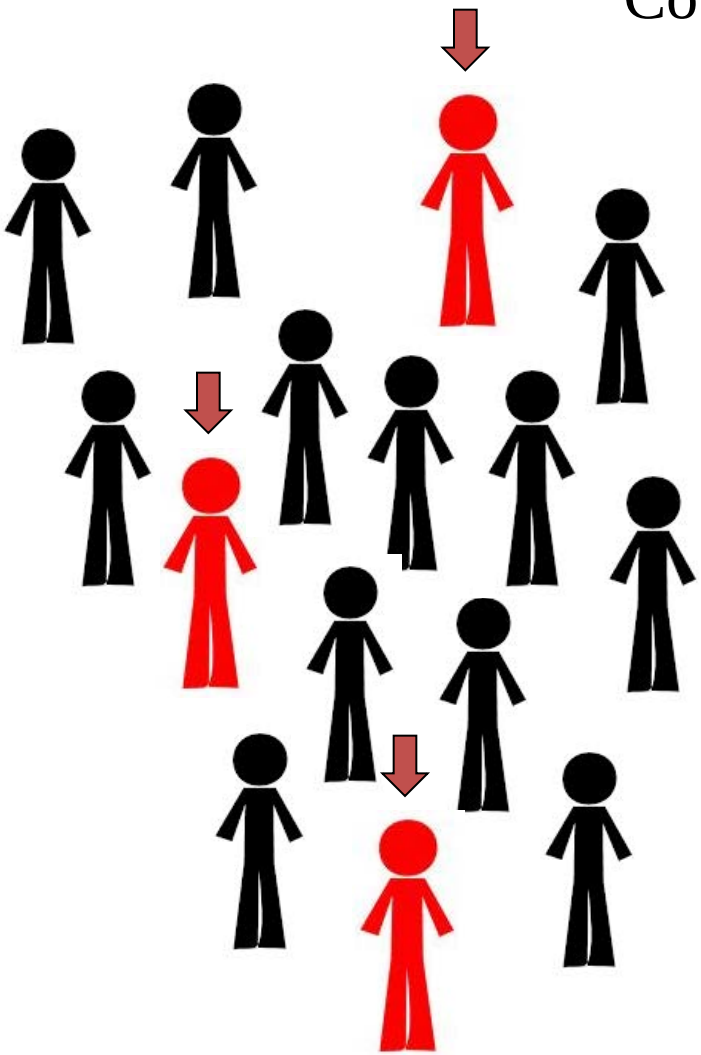

Couldn't we i… eeded?

Not so Easy

Ne…

1. To identify all those infected
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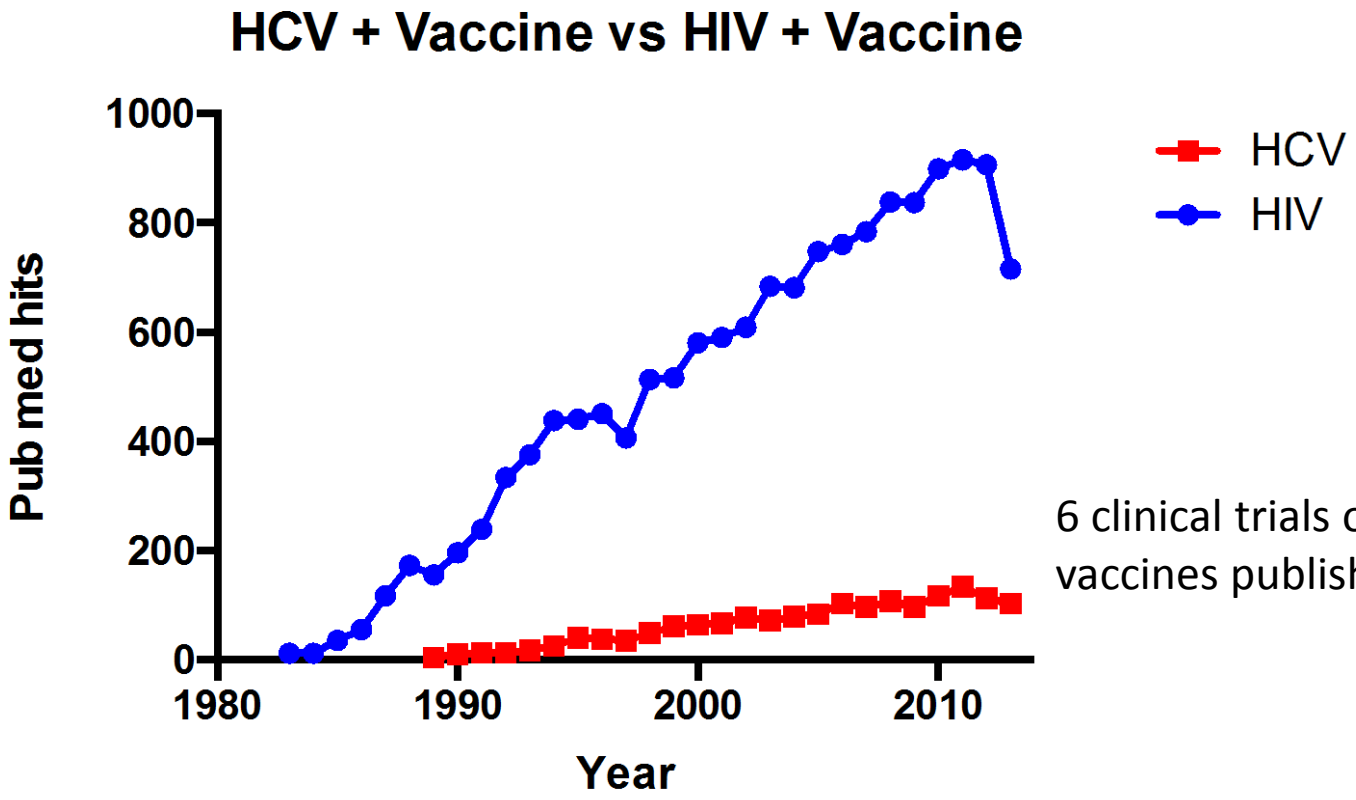
Getting Close

What is being done?



6 clinical trials of preventive vaccines published

Swadling et al ERV 2013

Some similar problems for HCV and HIV vaccines

- Complex antibody target structures
- Mutable antigenic targets for humoral response

- Potential for immune escape from T cells
- Dysfunction of T cell responses in chronic disease

- BUT HCV=clear pathway for robust defence

Humoral responses and viral control

- Evolution of HCV envelope (E1/E2) under immune selection associated with progression
- Strain-specific antibodies early – later becoming more broadly cross-reactive
- Development of Neutralising Ab has been temporally associated with clearance

Raghuraman et al, JID 2012
Von Hahn et al, Gastro 2007
Netski et al CID 2005
Farci et al Science 2000

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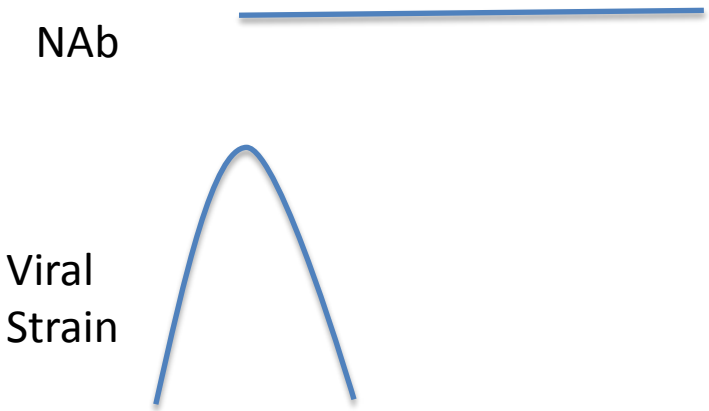

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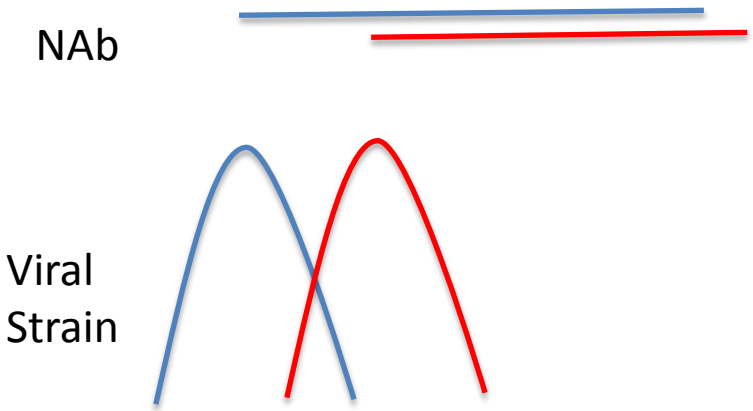

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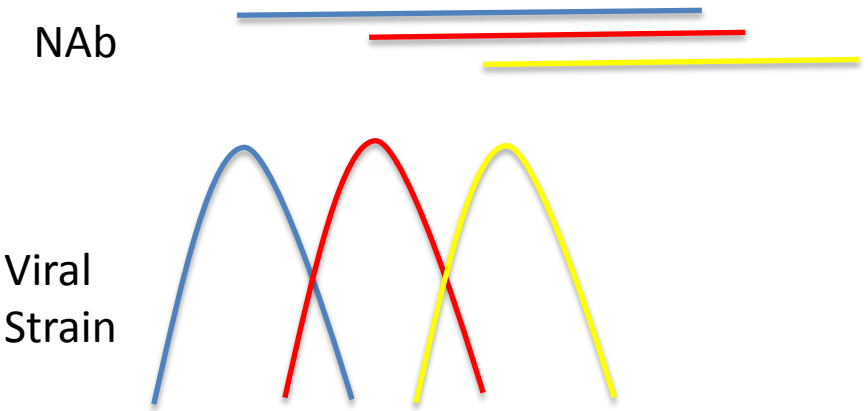

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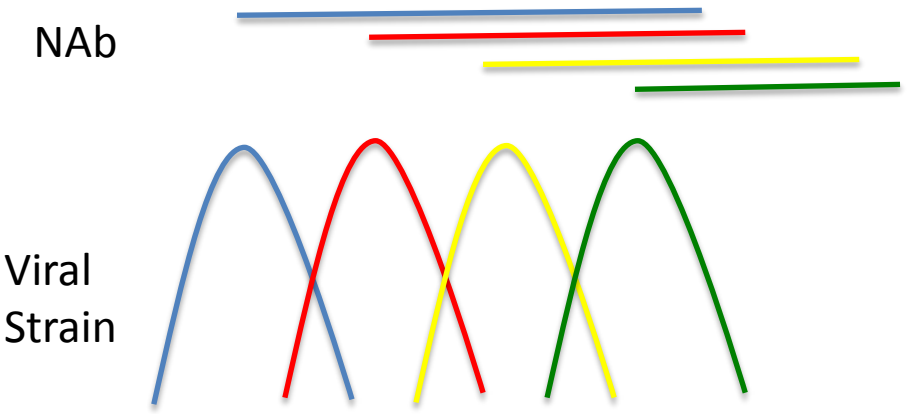

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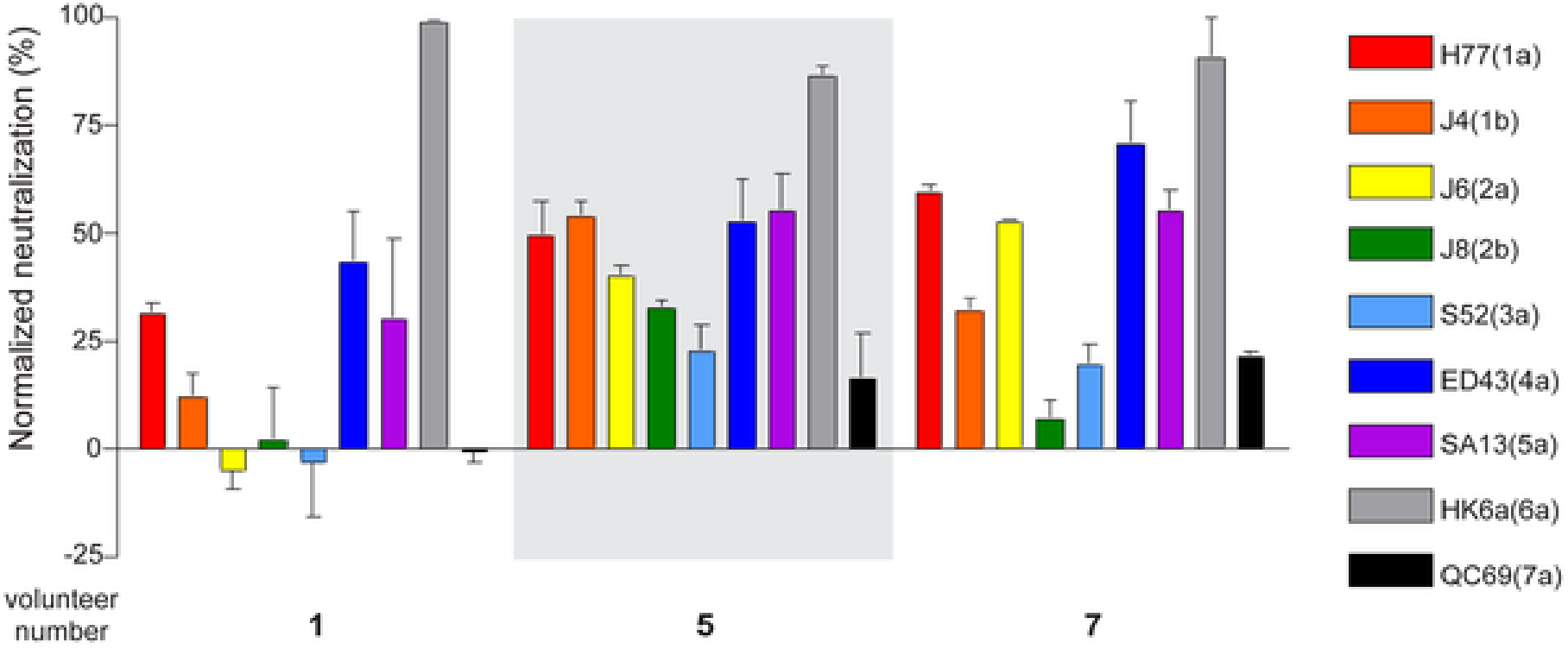
Netski et al CID 2005
Farci et al Science 2000

Can we generate broadly neutralising antibody responses by vaccination?

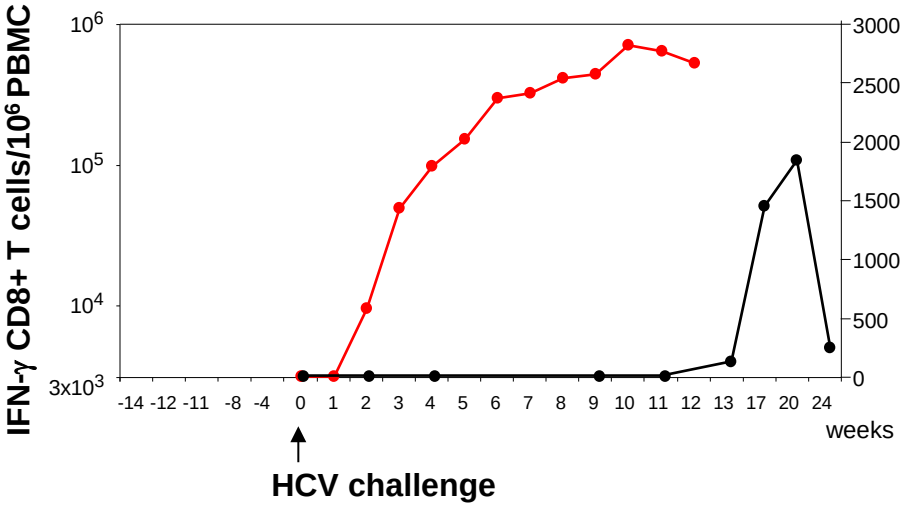


Law JLM, Chen C, Wong J, Hockman D, et al. (2013) A Hepatitis C Virus (HCV) Vaccine Comprising Envelope Glycoproteins gpE1/gpE2 Derived from a Single Isolate Elicits Broad Cross-Genotype Neutralizing Antibodies in Humans. PLoS ONE 8(3): e59776. doi:10.1371/journal.pone.0059776
http://www.plosone.org/article/info:doi/10.1371/journal.pone.0059776

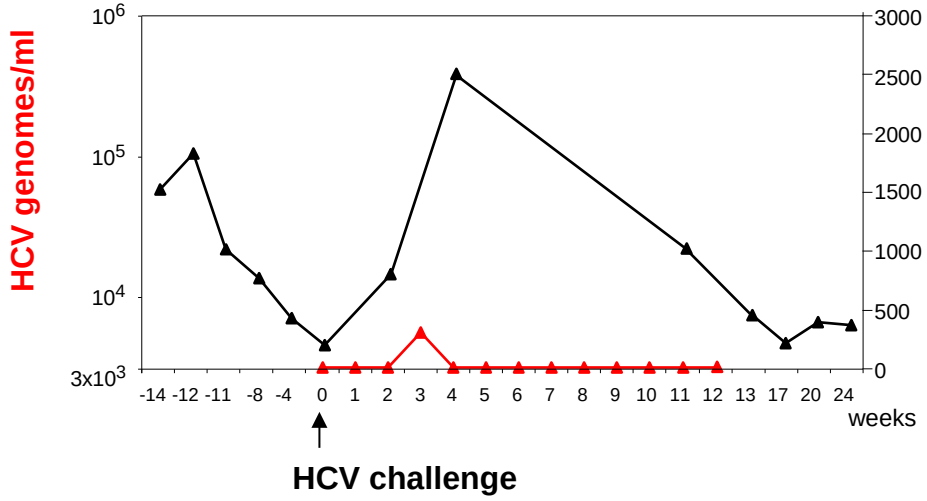
T Cell Immunity contains HCV - the evidence

- HLA association studies and GWAS (Class I and Class II HLA with clearance) (Neumann C, et al. Hepatology 2006;Duggal et al, Ann Int Med 2013)
- Chimpanzee CD4+ and CD8+ T cell depletion experiments *(Shoukry N J Ex Med 2003)*
- Association of breadth and magnitude of T cell response with viral clearance *(Lauer et al Gastro 2004)*
- IFN-γ HCV specific $CD8^+$ T cell responses are temporally correlated with reduced viremia after infection (Lechner F J Exp Med 2000; Thimme et al J Exp Med 2001)
- Prophylactic vaccine data (Adeno/DNA) in a chimp challenge model. *(Folgori et al Nat med 2008)*
- *BUT…NO ONE CORRELATE OF PROTECTION (BIG,BROAD,SUSTAINED, FUNCTIONAL etc)*

Vaccination = acceleration (by 3 months)



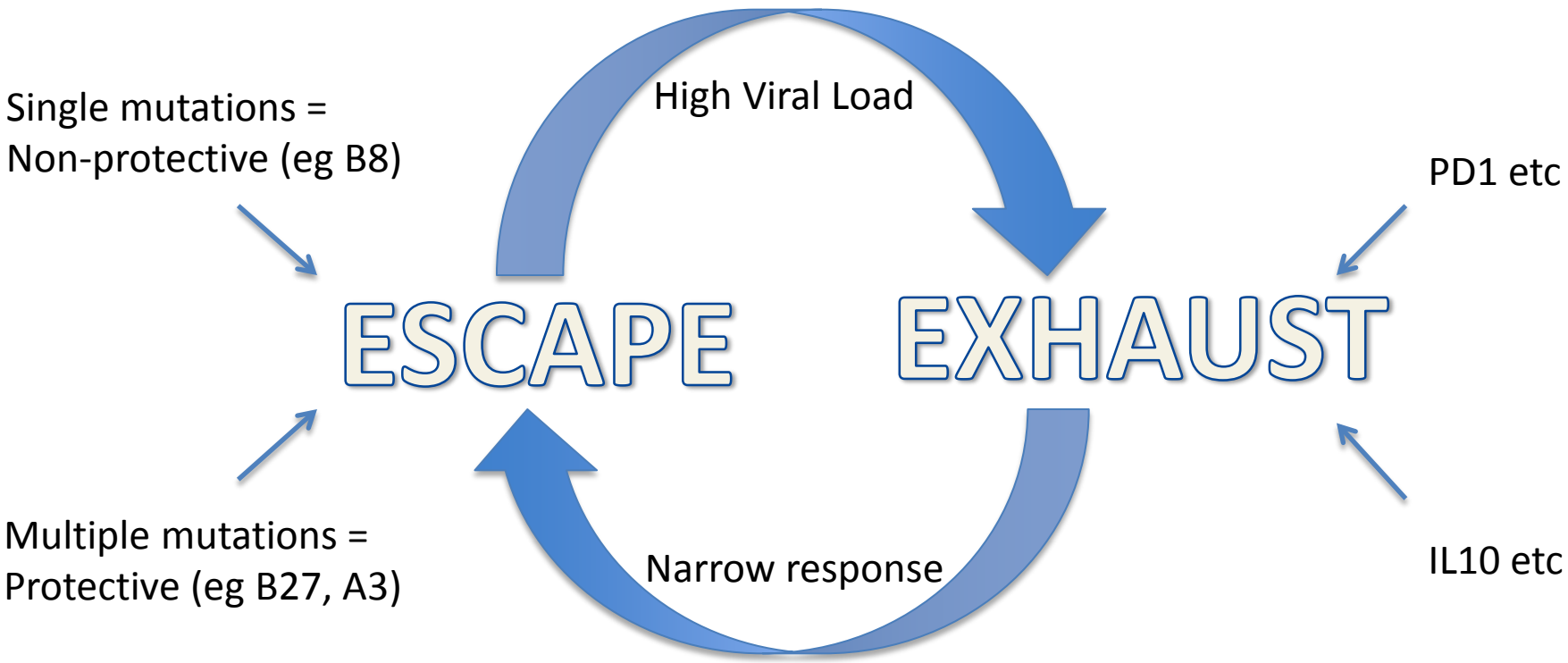
Control animals



Vaccinated HCV Ad-NS animals

Folgori et al. Nature Medicine, 2006

THE NEED FOR SPEED



Thomson et al Gut 2010

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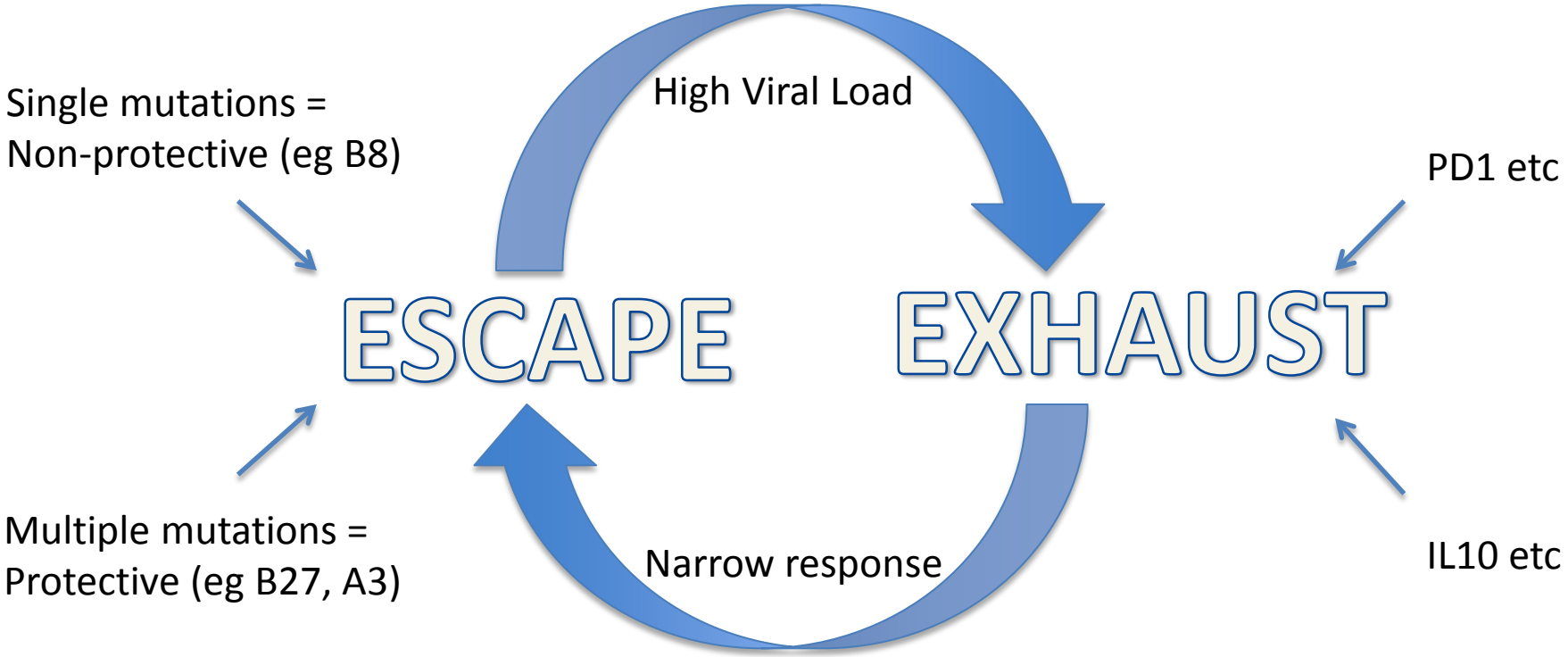

Table 2 Median viral load (VL) drop in untreated spontaneous clearers and progressors

	Spontaneous clearers (SC) (95% CI)	Progressors (PV) (95% CI)	Progressors (FV) (95% CI)	HR* (95% CI)	p Value
Maximum HCV VL $\log_{10}$ (IU/ml) drop within 100 days from first positive PCR	2.20 (1.65 to 5.03)	0.03 (−0.33 to 0.23)	0.58 (0.25 to 0.96)	1.78 (1.45 to 2.18)	<0.0001†
Maximum HCV VL $\log_{10}$ (IU/ml) drop within 200 days from first positive PCR	3.46 (1.70 to 7.05)	0.17 (−0.06 to 0.38)	0.85 (0.45 to 1.77)	1.68 (1.40 to 2.02)	<0.0001†

*HR represents change in hazard per $\log_{10}$ change in VL

Thomson et al Gut 2010

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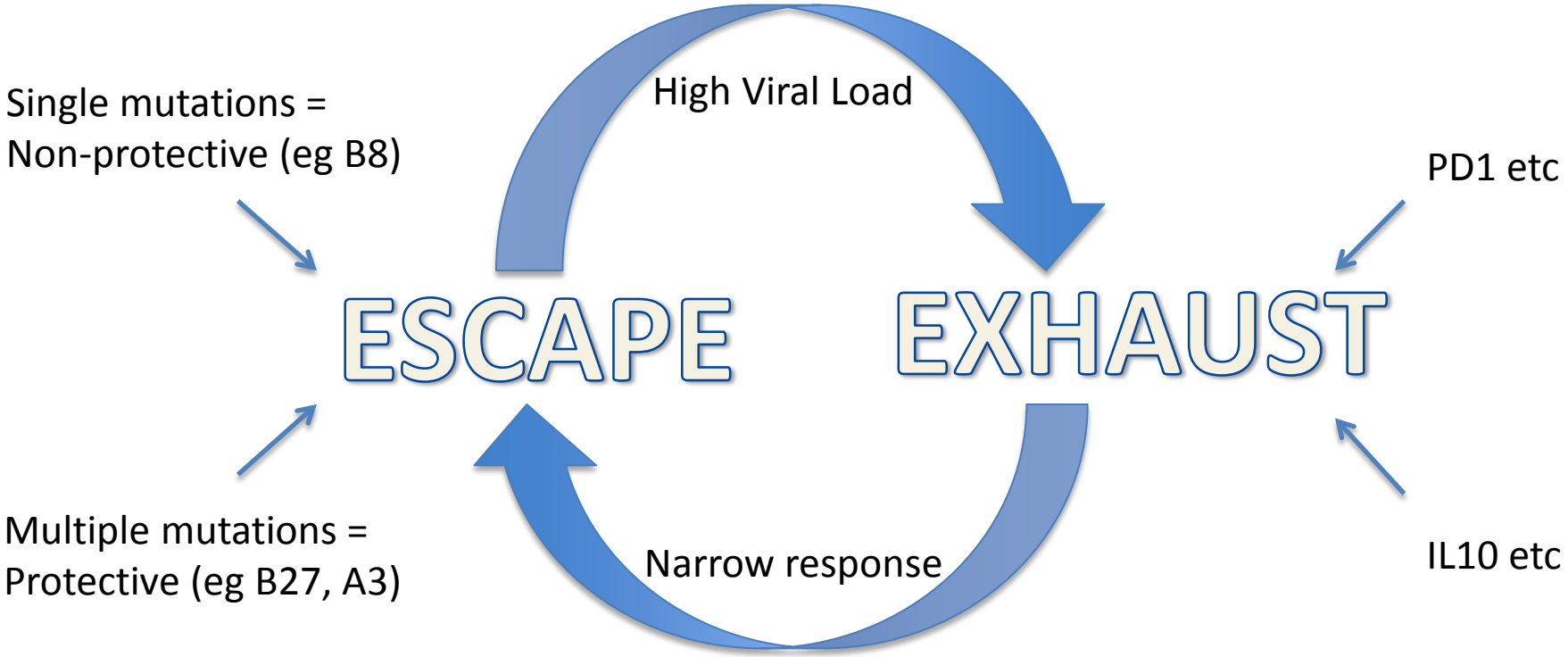

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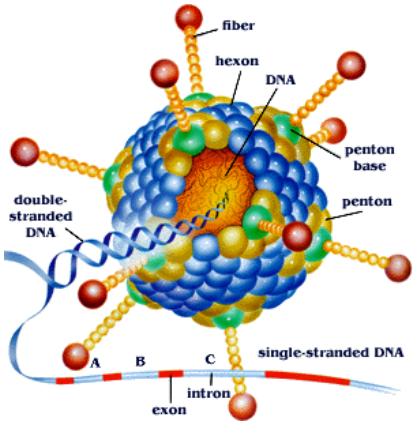
Thomson et al Gut 2010

Getting your retaliation in first*: How to experimentally induce an HCV specific T cell response?



- Adenoviral vectors disabled genetically
- Vector foreign antigens
- Induce strong cellular immunity
- combined with other vectors in heterologous prime boost regimens

Adenoviral vectors highly potent in priming antigen specific T cell responses (malaria, HIV etc)

* Carwyn James: British Lions Coach, Tour down under 1971

A problem: pre-existing anti-adenoviral immunity

Adenoviruses are shared by Humans and Chimps

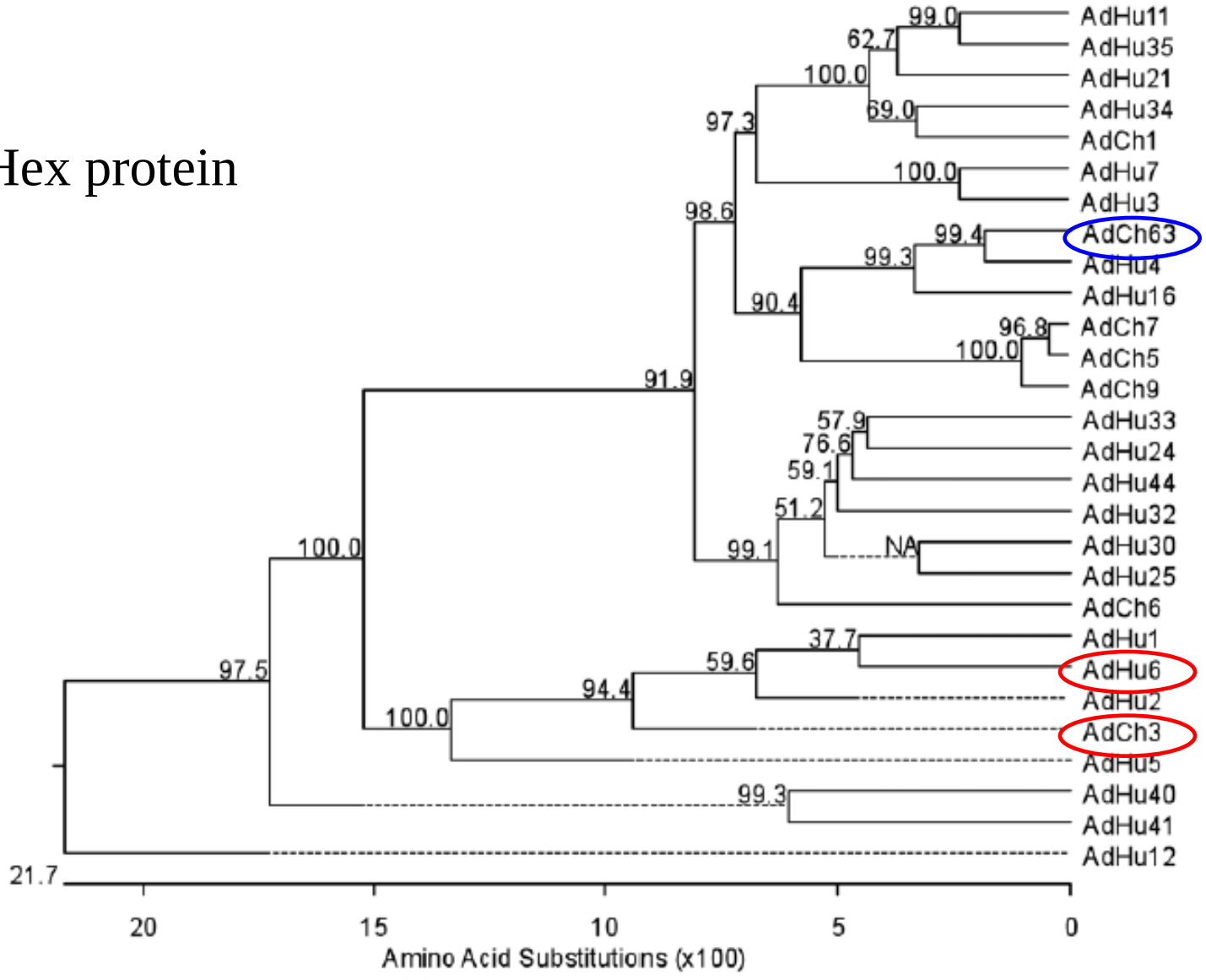
- chimps catch colds too!

- have their own adenovirus strains

- little exposure of humans to these

Adenoviral vectors-phylogenetic analysis

Ad Hex protein



Colloca et al Science translational medicine Jan 2012; Barnes et al Science translational medicine Jan 2012

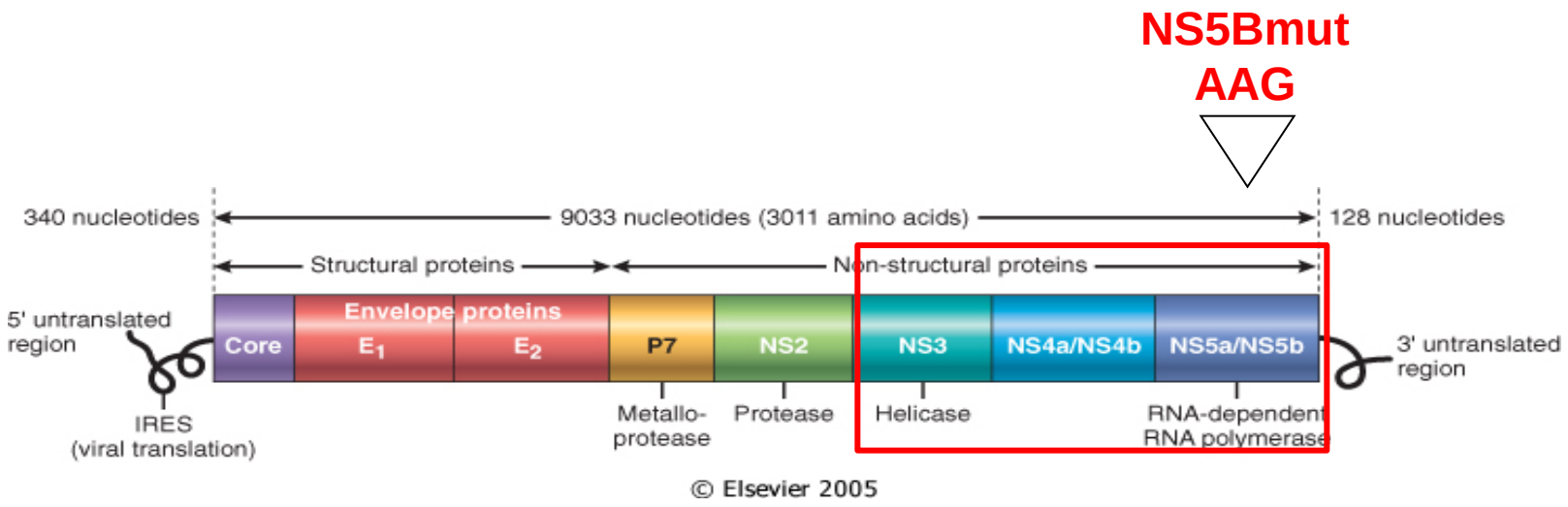
The HCV immunogen

NS3-NS5B (NS = 1985 aa)

Genotype 1, subtype 1b

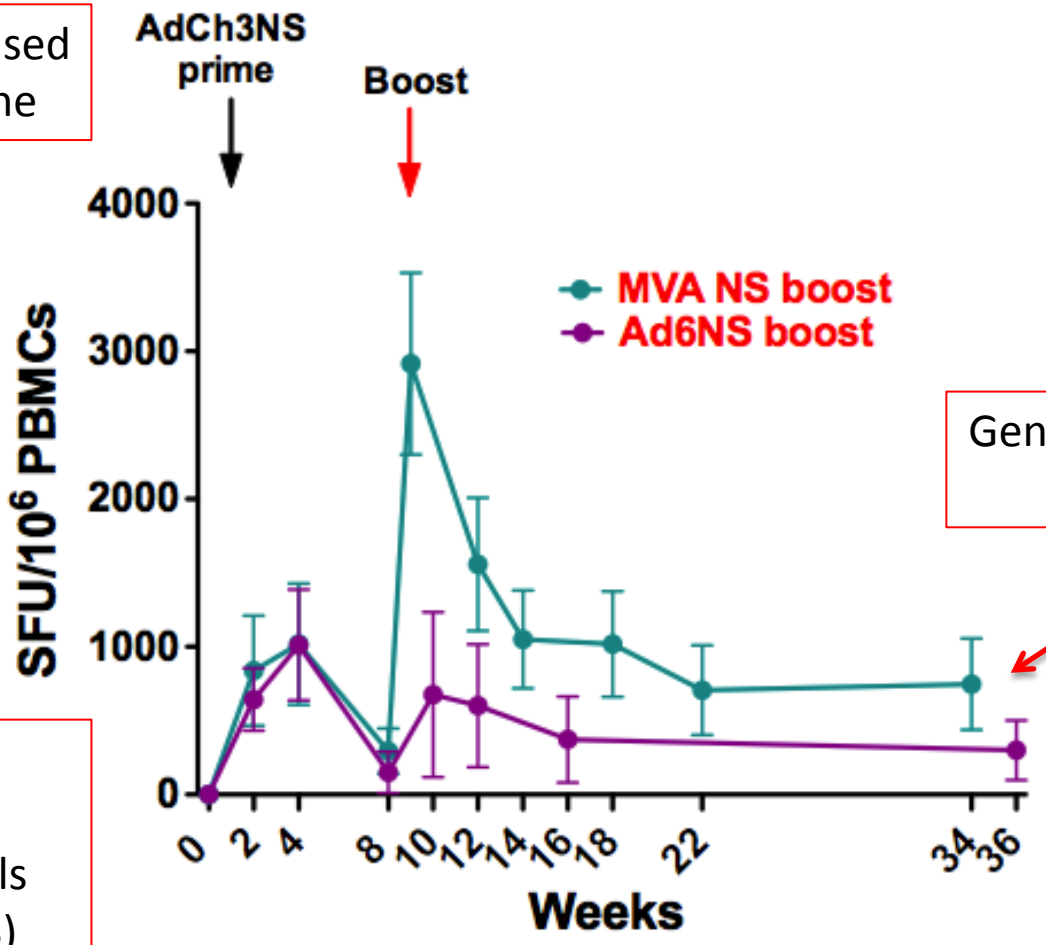
Multiple epitopes

Genetically inactivated NS5B (NSmut)



Development of T cell vaccine for HCV

Same vector as used for Ebola vaccine

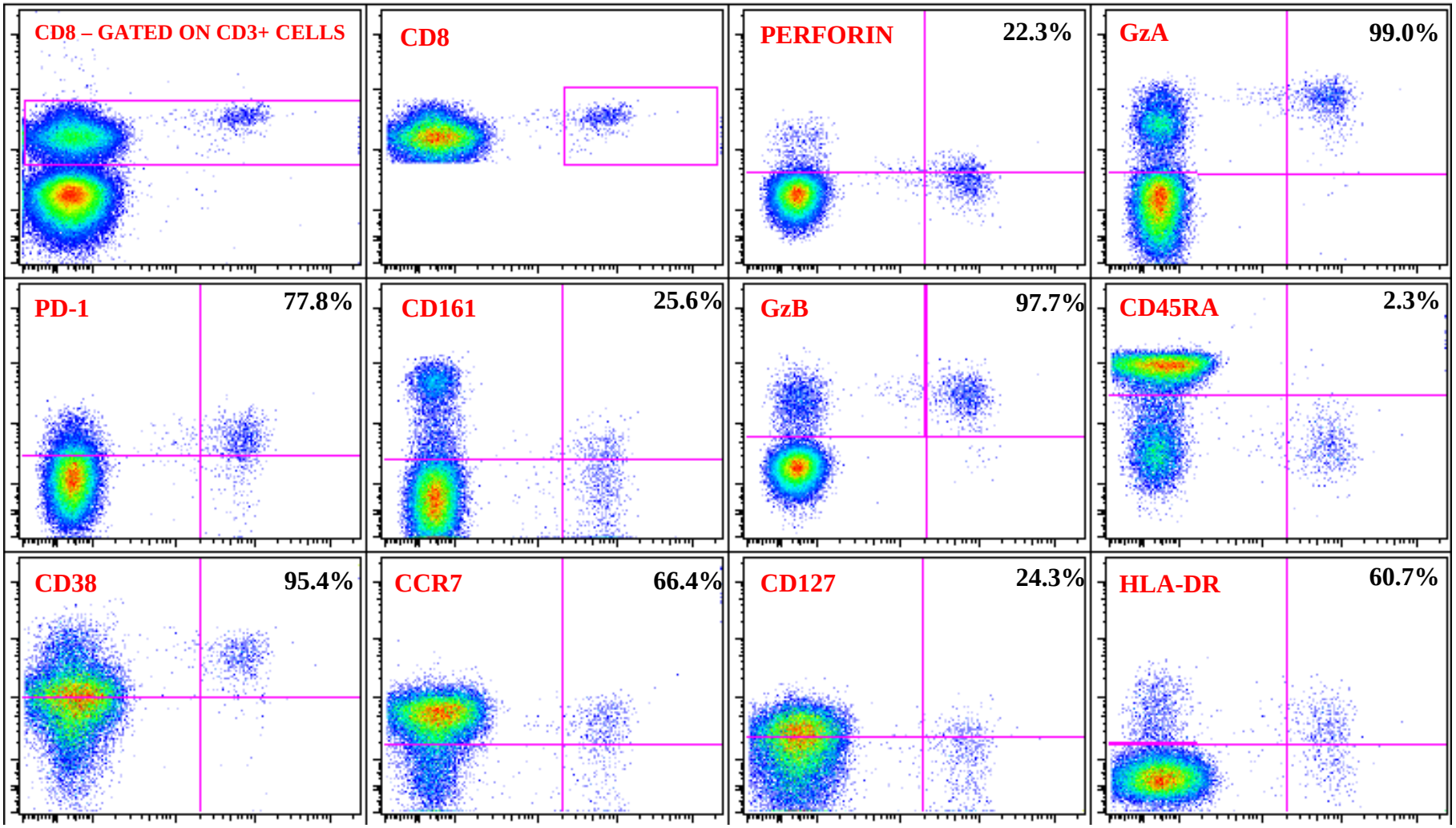


Generation of HCV-specific T cell responses

Measure IFN gamma Secreting T cells (like T-spot TB)

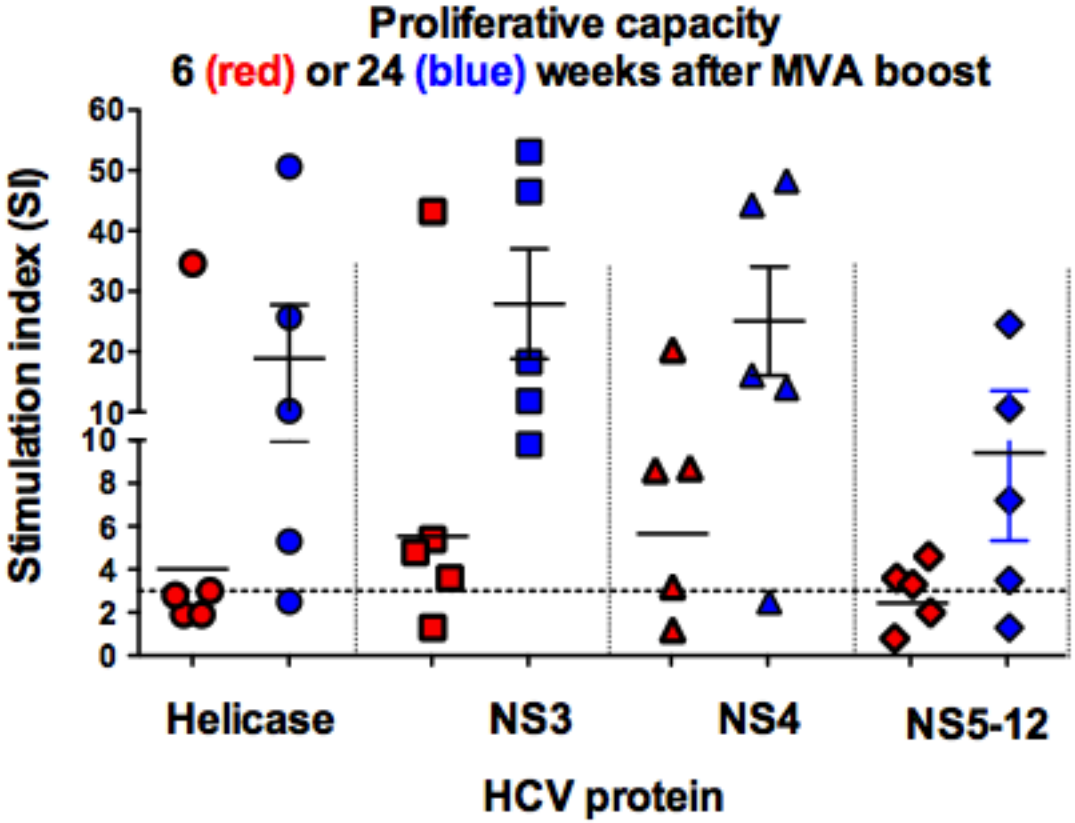
Barnes et al Science Tr Med 2012; Swadling et al Science Tr Med 2014

T cell characterisation using HLA class-I pentamers



Barnes et al Science translational medicine 2012; Swadling et al Science translational medicine 2014

Proliferative responses increase over time



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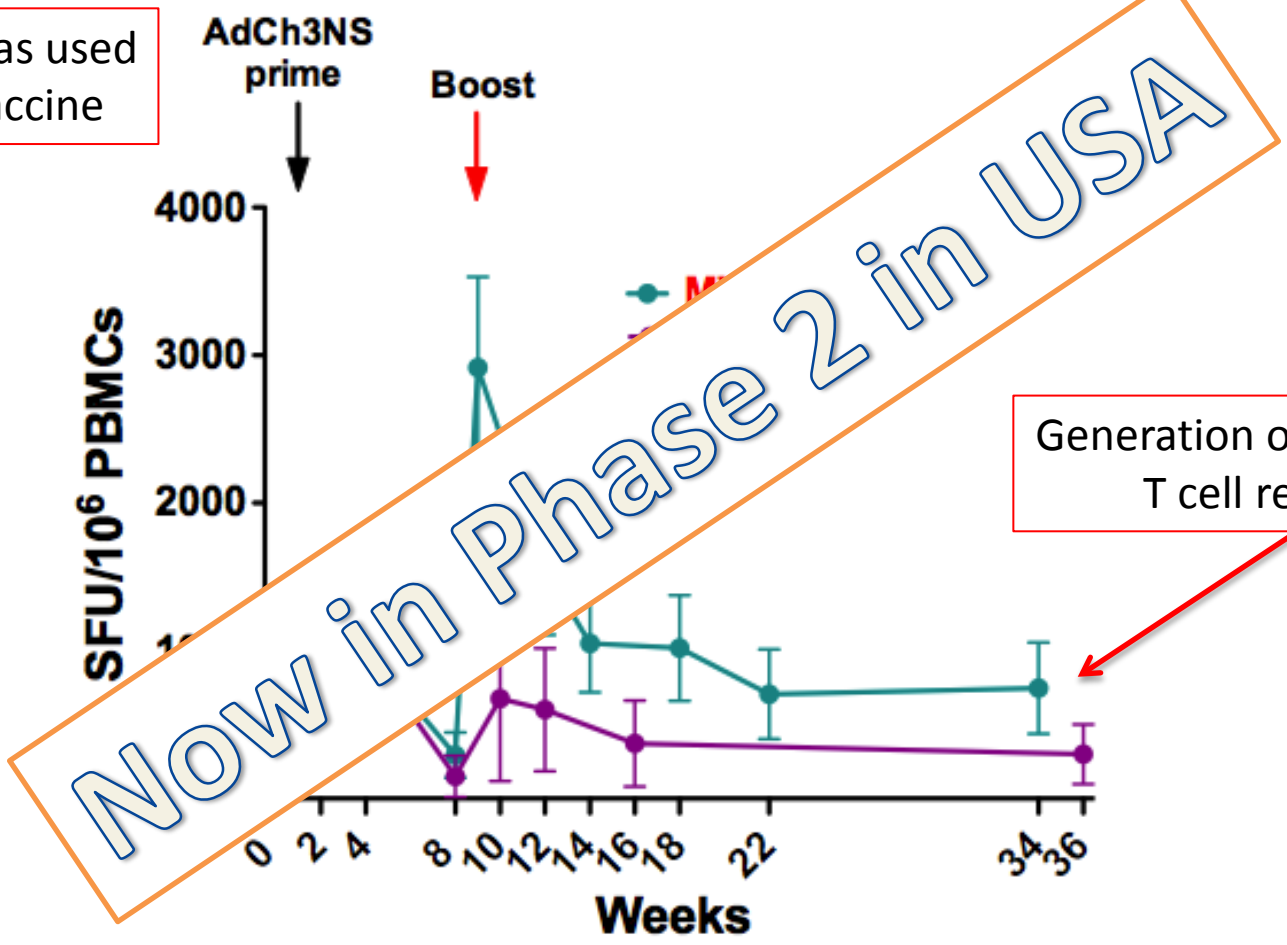


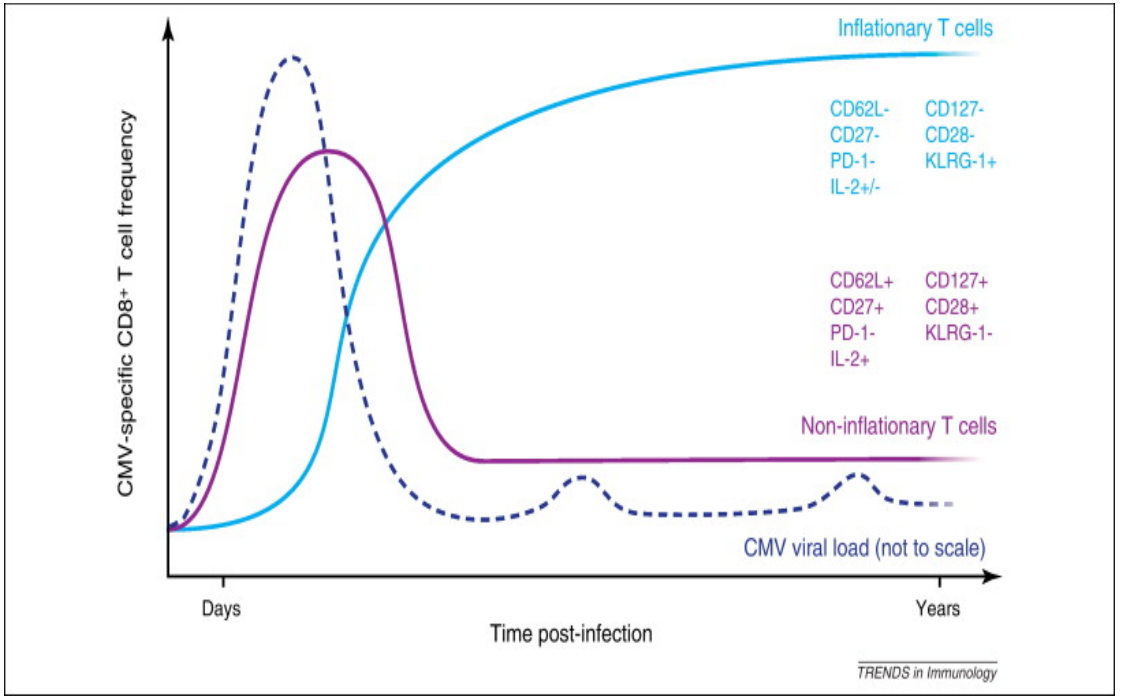
Now in Phase 2 in USA

Generation of HCV-specific T cell responses

Barnes et al Science Tr Med 2012; Swadling et al Science Tr Med 2014

Memory Inflation - Overview

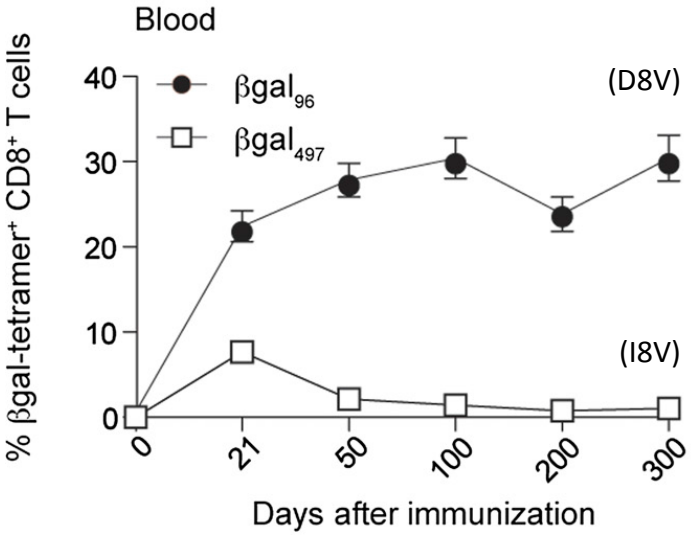
- Memory inflation was first noted in MCMV, with up to 30% of $CD8^+$ T cells being accounted for by this subset
  (Karrer *et al*. J Immunol 2003)
- MHC Class I tetramers can be used to track these populations *in vivo*
- Inflationary T cells have an T_{EM} phenotype, are found in the periphery and remain functional

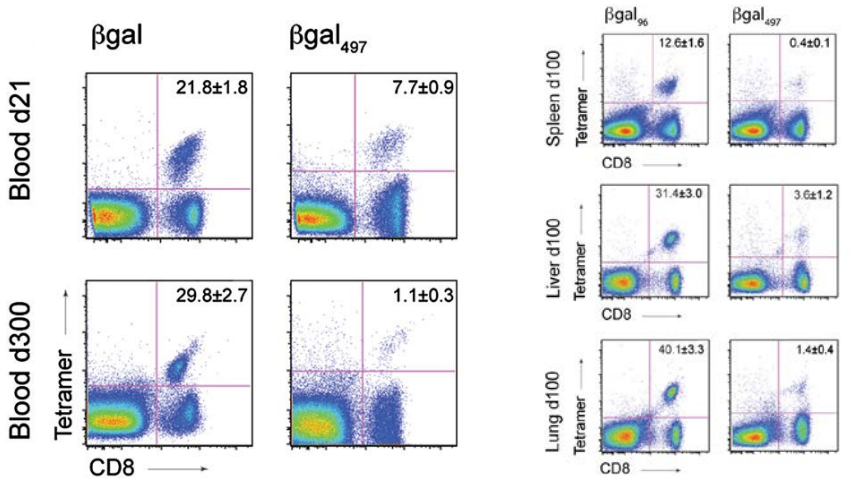


O'Hara *et al*. Trends in Immunology 2012

The Adenoviral model for memory inflation

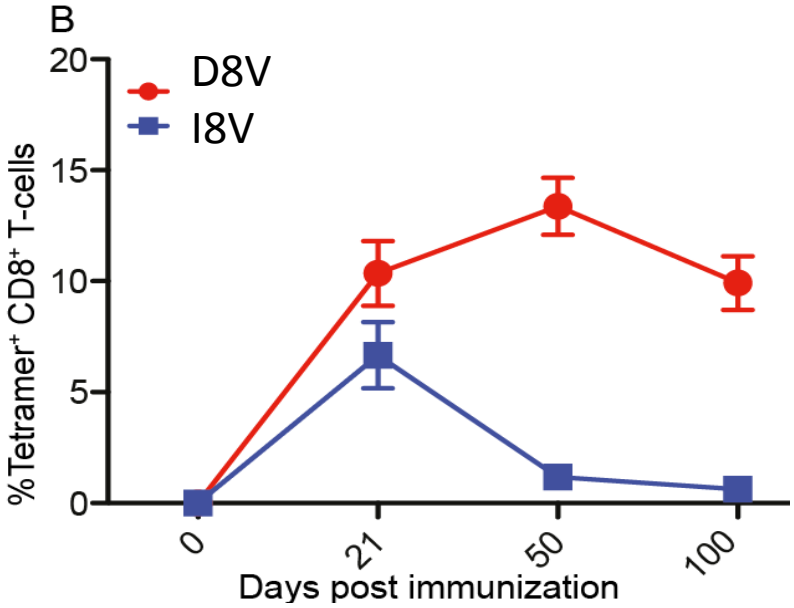
- Recombinant adenovirus expressing the βgal protein, human CMV promoter and lacking E1 and E3 genes
- Two co-dominant responses, elicited from the same protein
- High level antigen expression, largely in the liver
- The model recapitulates all features of memory inflation, including frequency, function, phenotype, distribution and transcriptome





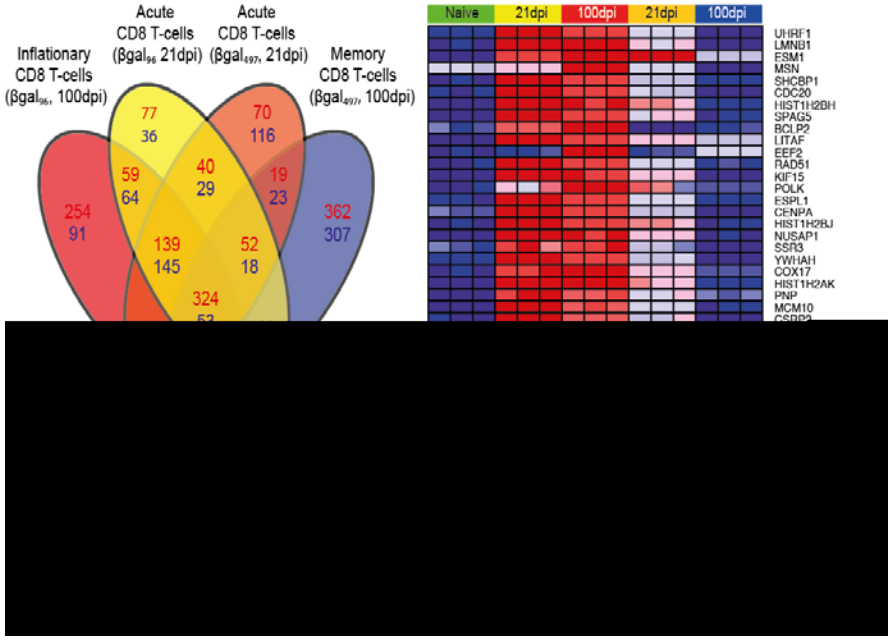
Bolinger *et al*, J Immunol 2013

Persistent Tem population vs conventional memory



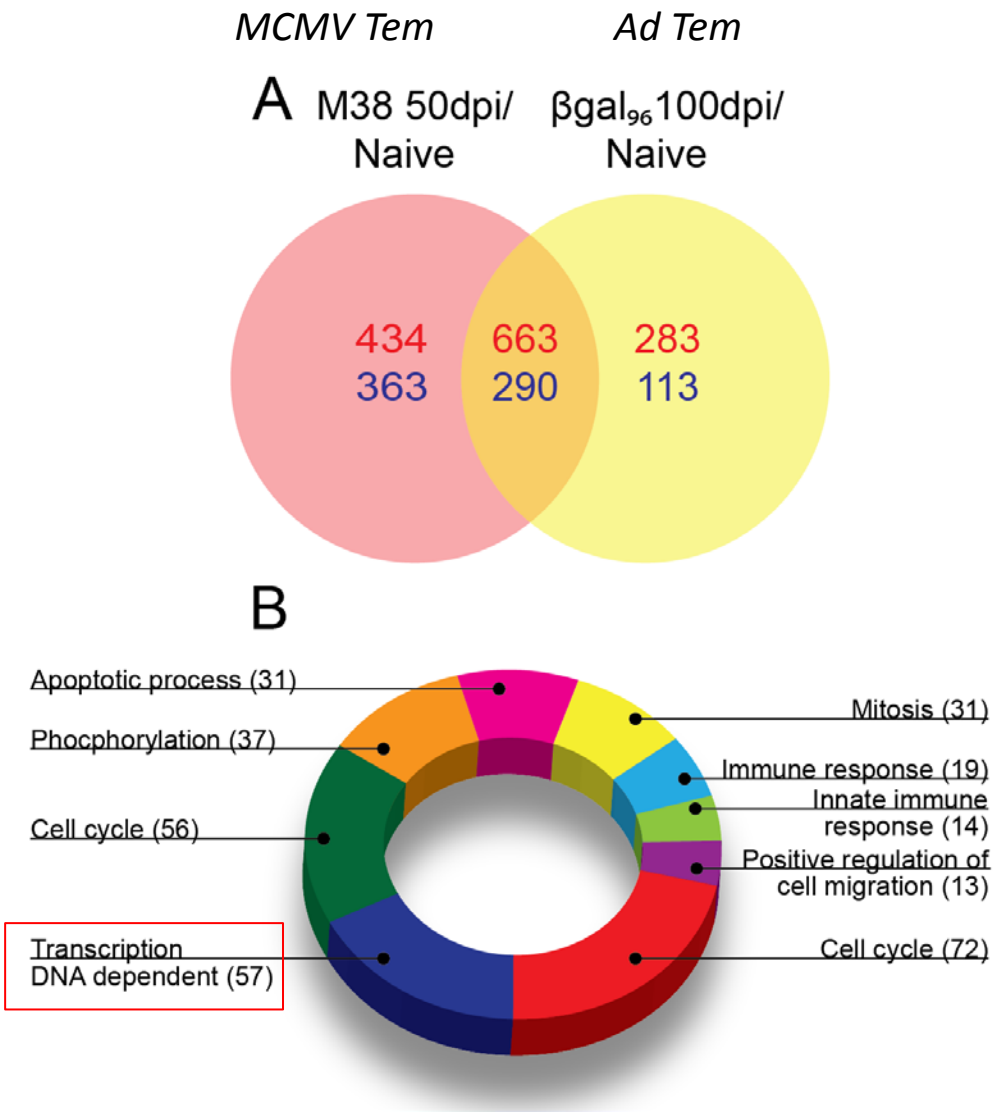
Adenovirus

Inf *Non-Inf*



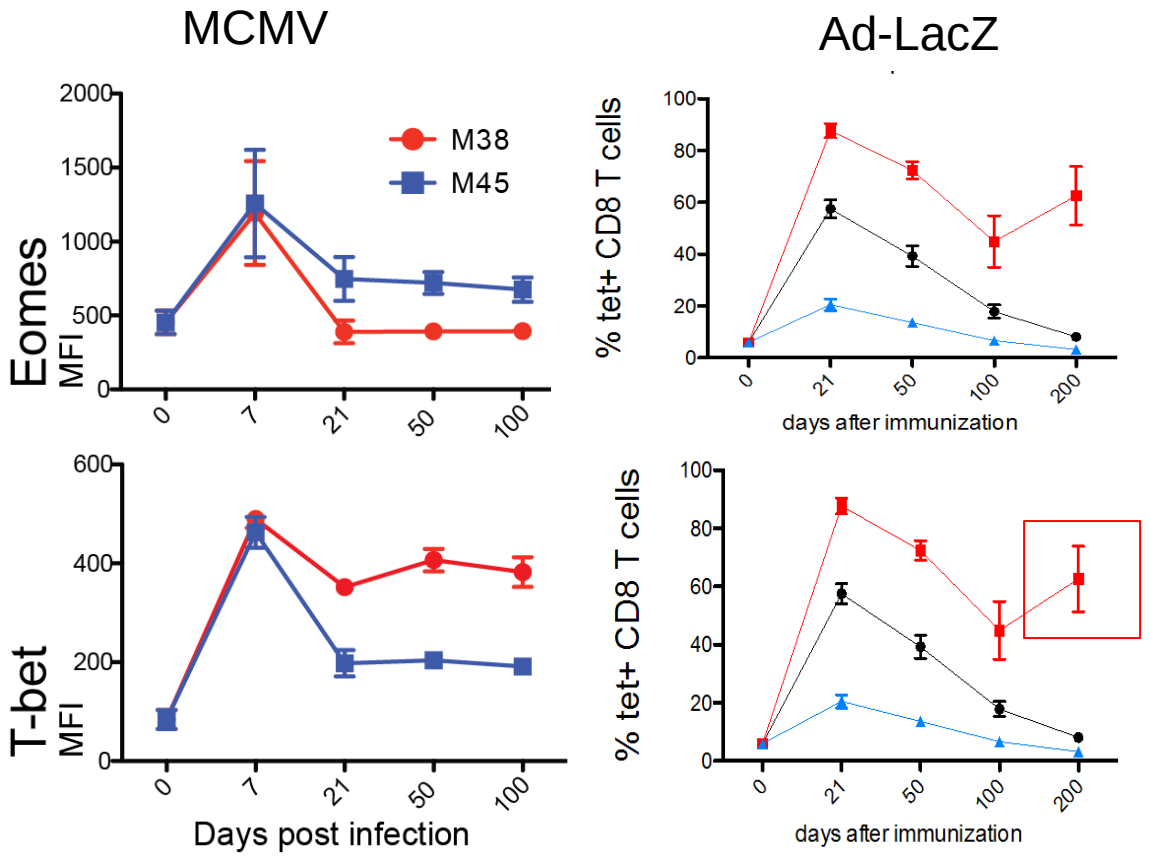
- 2 T cell responses induced in parallel (same transgene) - Ad5-LacZ
- D8V (inf) : T-cells acquire 'inflation-specific gene expression from acute to memory
- I8V (non-inf): regresses back towards Naïve T-cell gene exp

Stuart Sims *Bea Bolinger* *Emanuele Marchi*



MCMV- and Adeno-induced inflationary populations share a core transcriptional programmme

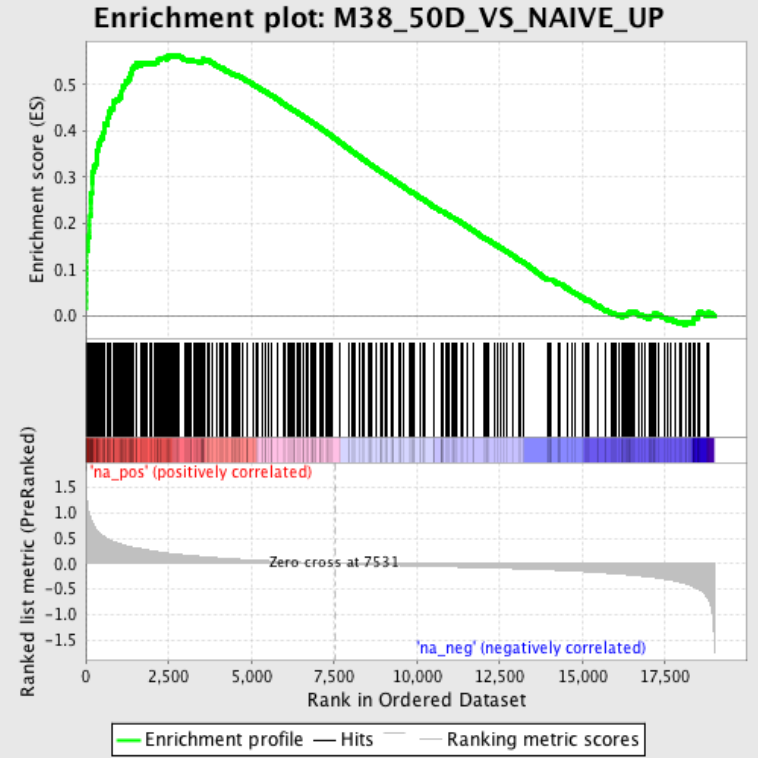
Tbet (TBX21) is reproducibly sustained in inflationary populations



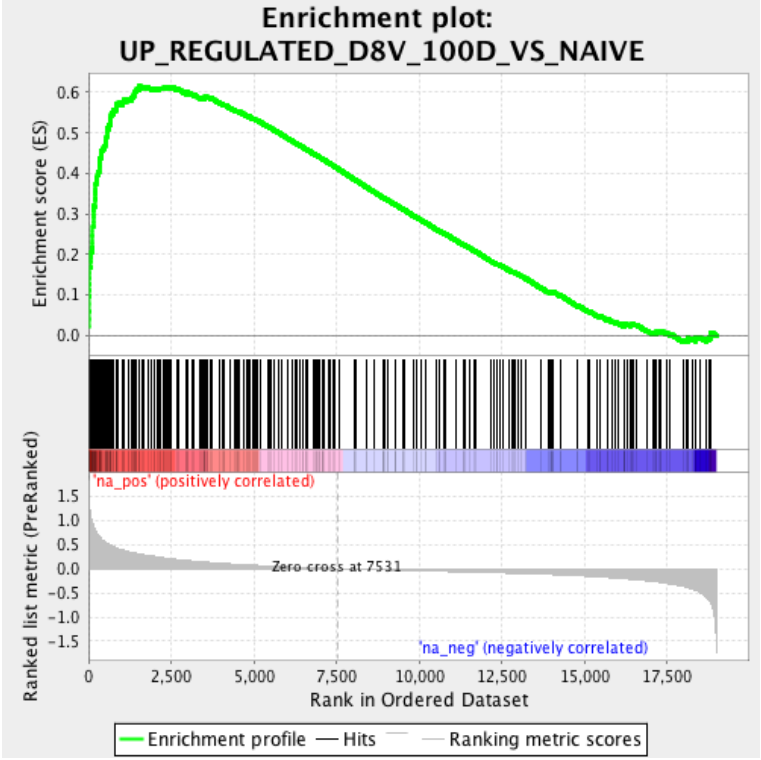
- Inflating/ persistent Tem
- Non-inflating/ conventional
- Bulk CD8

GSEA: Cells resemble Human CMV responses

MCMV vs HCMV



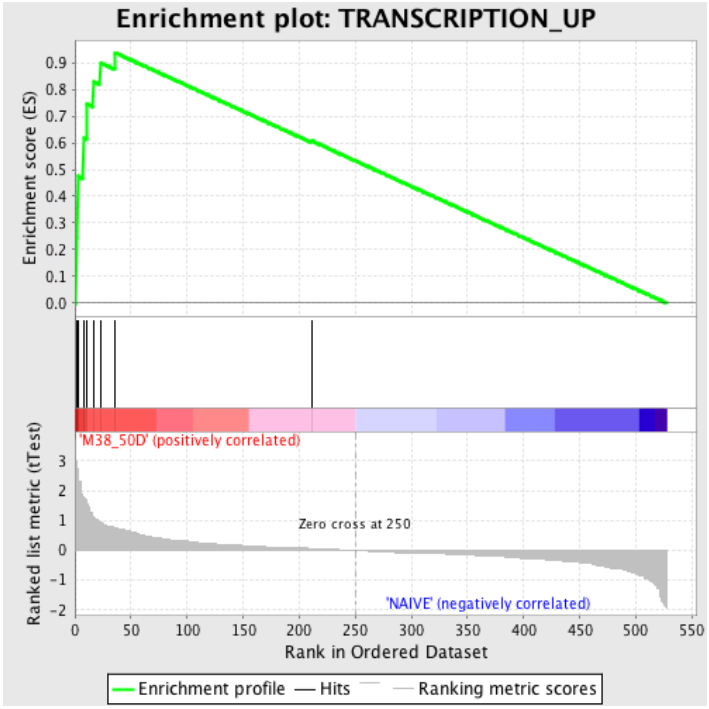
Ad vs HCMV



Sims, Bolinger et al Cell reports 2015

GSEA: Cells resemble Human CMV responses (TFs only)

Top Gene = Tbet



SO…WE CAN INDUCE “INFLATED” POPULATIONS IN MICE USING ADENO…

Is this at all relevant to human adenovectors?

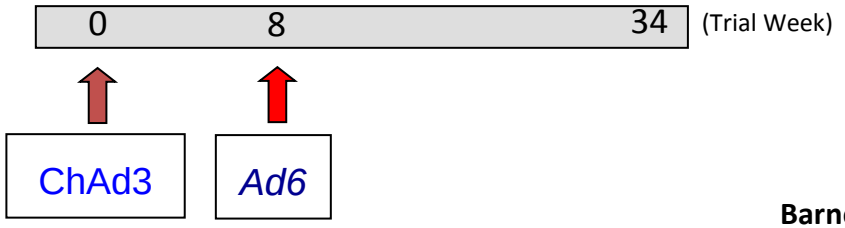
- 1. Do the responses have a “CMV” like phenotype?
- 2. Do they express high Tbet?

Virally vectored vaccines for HCV

HCV Immunogen

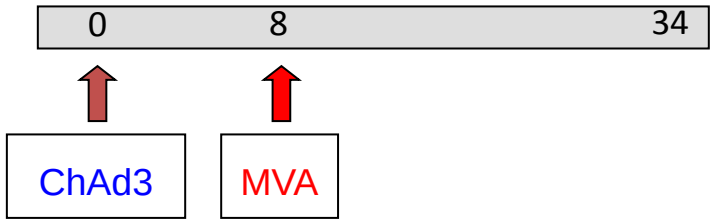
- NS3-NS5B (NS = 1985 aa). Genotype 1b. Highly conserved HCV region containing multiple epitopes

ChAd3/Ad6

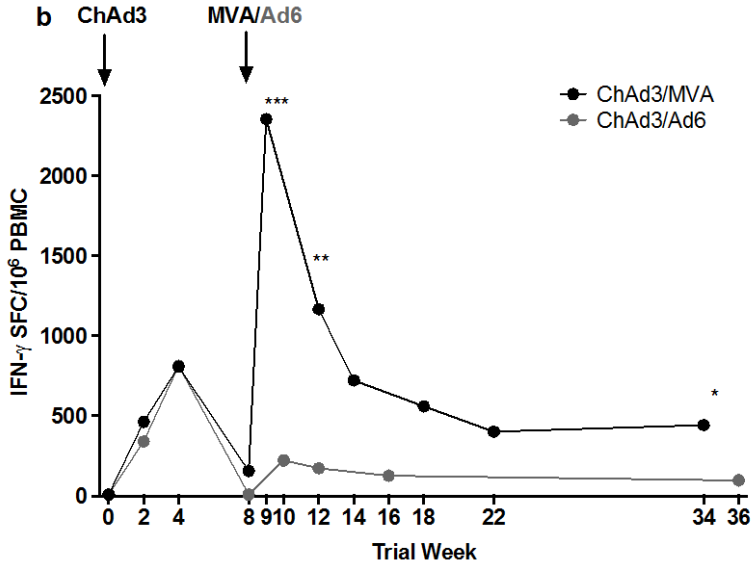


Barnes et al 2012 *Sci Trans Med*

ChAd3/MVA

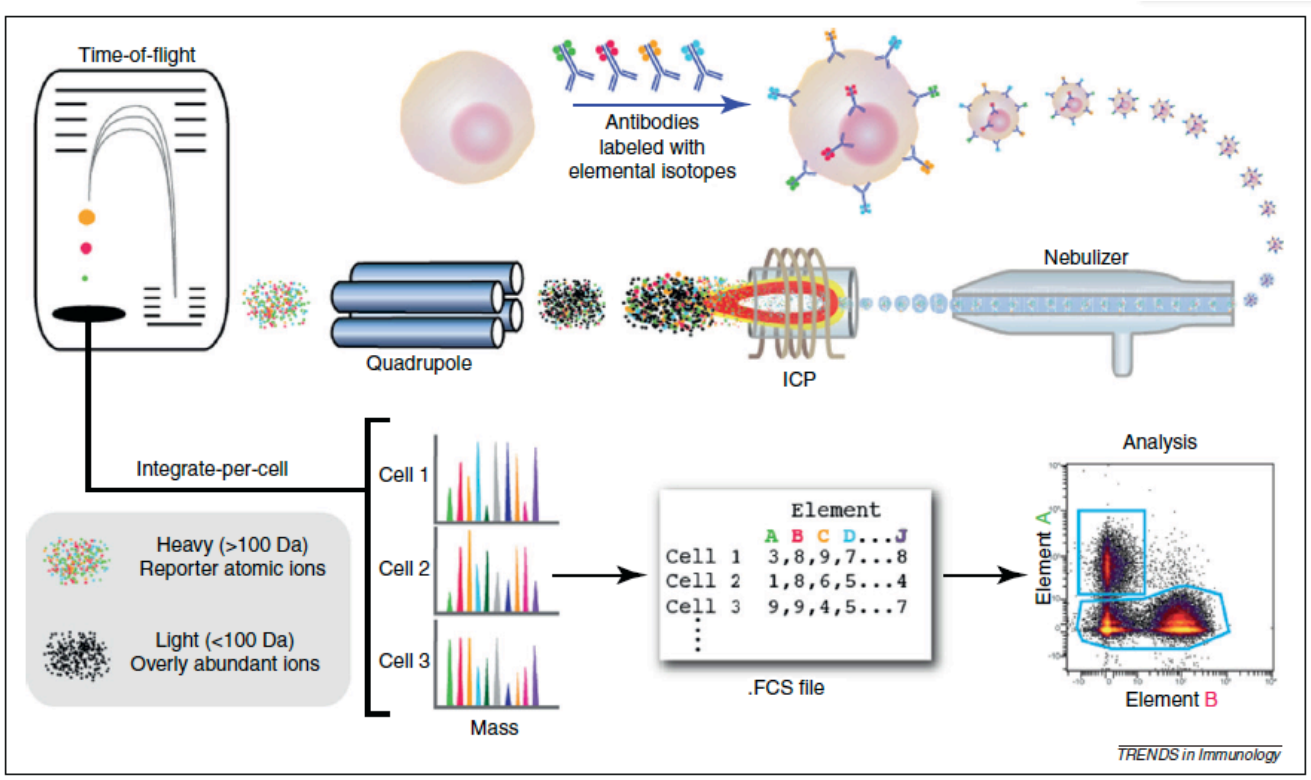


Swadling et al 2014 *Sci Trans Med*

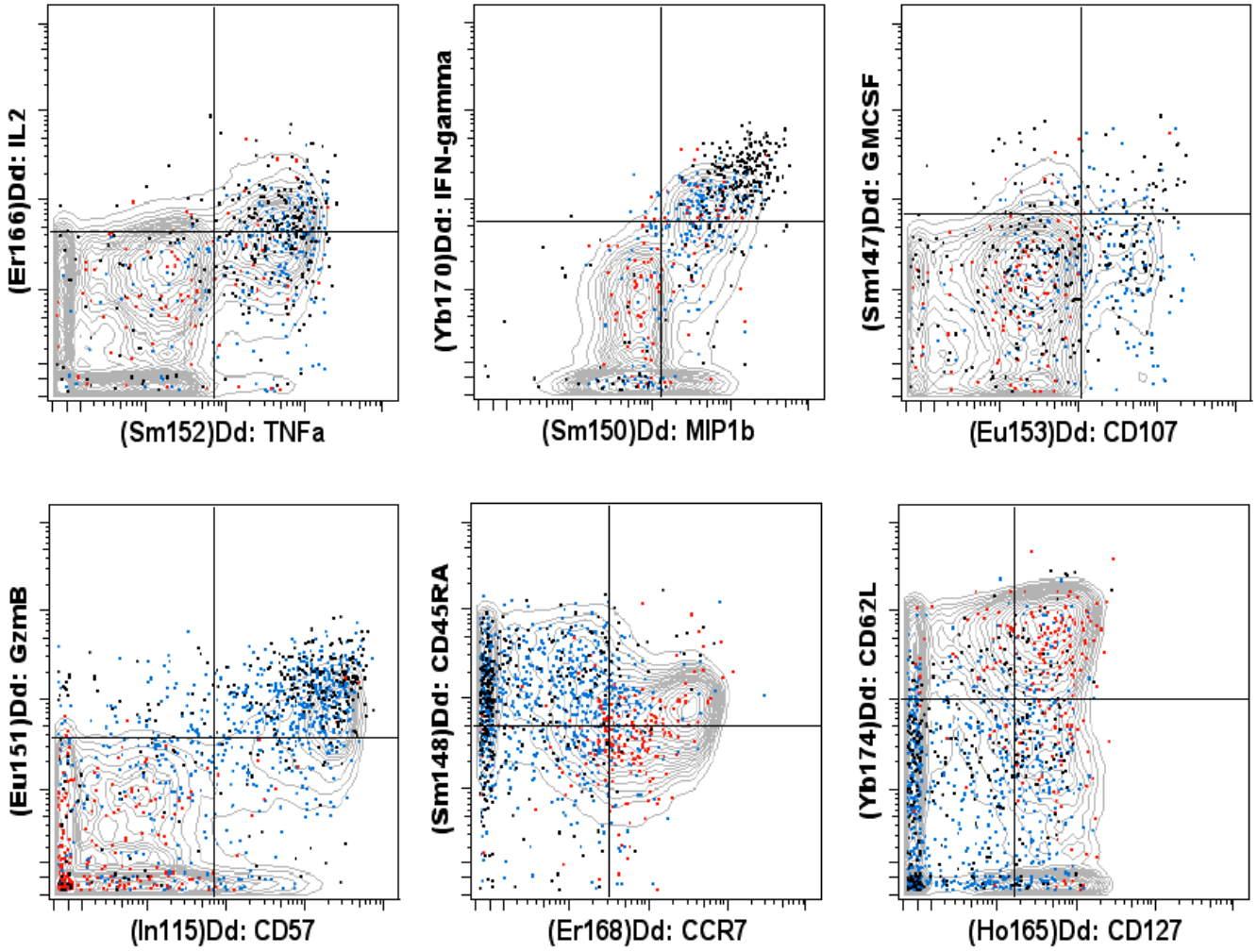


CyTOF – Inductively coupled plasma Mass Spectrometer (ICP-MS)

33 antibodies, 3 cell parameters (viability, DNA content, cell size) single cell analysis



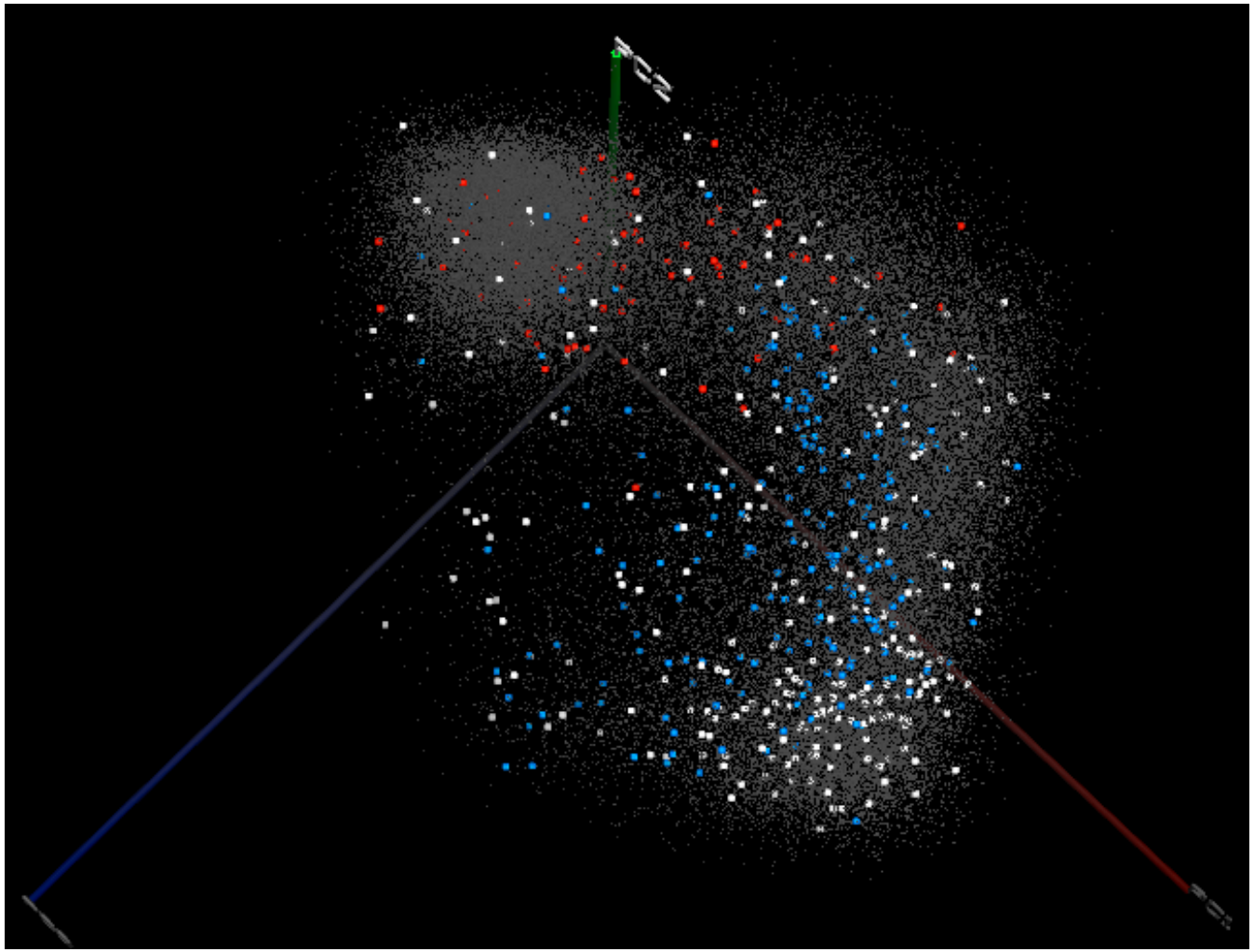
Mass cytometry – Shared phenotype CMV and Ad-induced



Flu-specific
CMV-specific
Ad-Induced

Sims, Bolinger et al Cell reports 2015

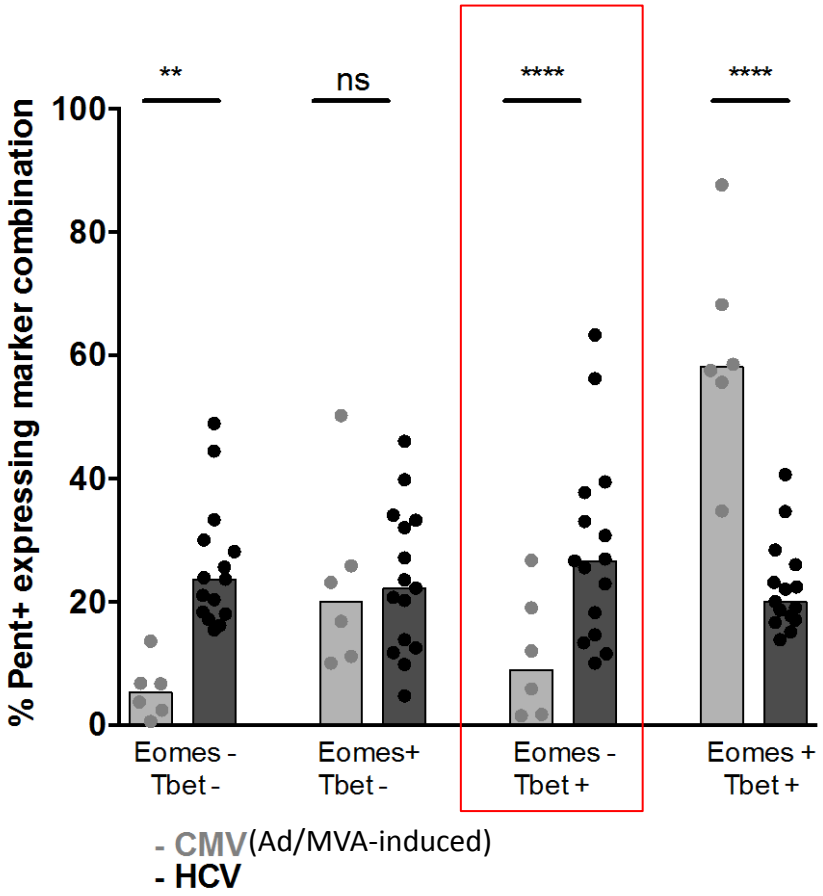
ADENOVECTOR INDUCED CELLS HAVE “CMV” PHENOTYPE



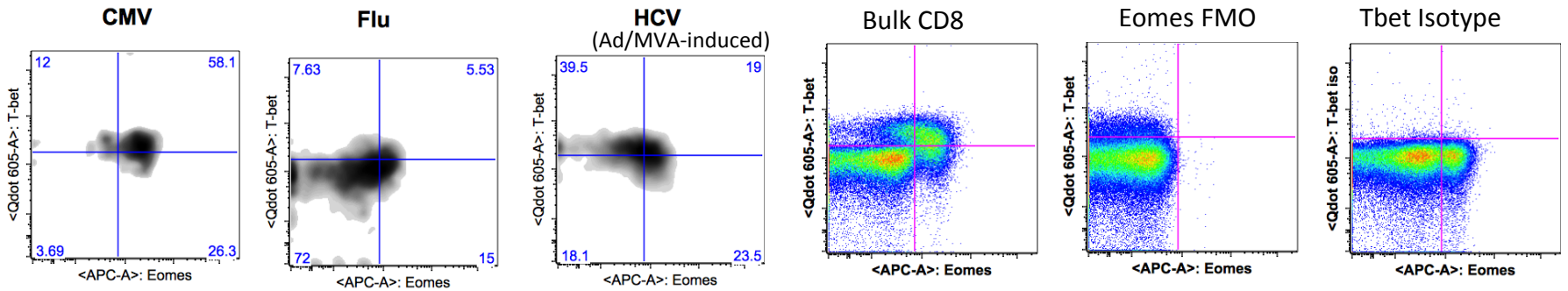
Red = Flu
Blue = CMV
White = HCV (TW22)

Sims, Bolinger et al Cell reports 2015

TFs on Ag-specific cells



- Ad/Ad and Ad/MVA induced CD8 T-cells show a mixture of Tbet/Eomes co-expression patterns
- Tbet+Eomes – enriched in those who have spontaneous resolved HCV relative to Chronics (Paley *et al* Science 2012).

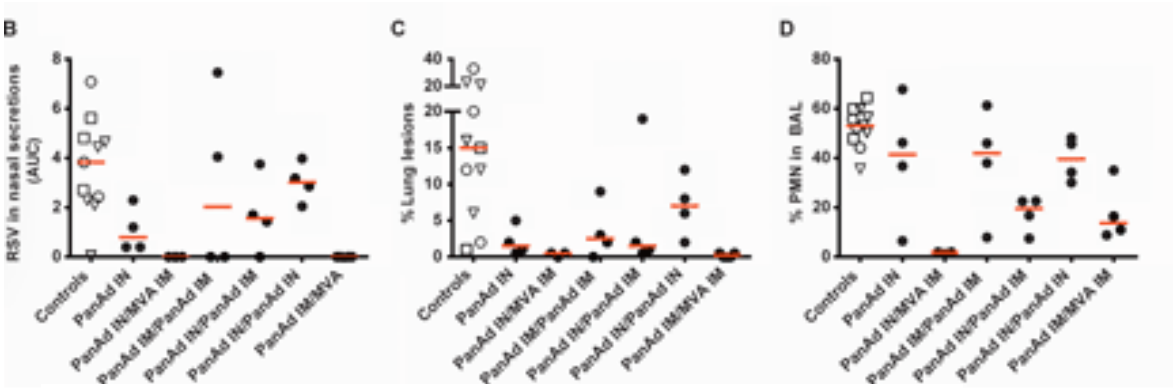


Sims, Bolinger et al Cell reports 2015

Conclusions

- MCMV and Adeno-induced “inflationary” memory share common features
- We still need to understand further where the antigen presentation is occuring, on what cell and at what timepoint.
- There are some shared features of memory with emerging human adeno-vectored vaccines.
- Such vectors (+/- boosting) may effectively induce “tissue homing” T cell populations for protection against persistent and complex pathogens.

?Next example: RSV



Calves (protection)

B

Fa peptide pool

Fb peptide pool

M peptide pool

N peptide pool

Adult Humans (immunogenicity)

(P) IN PanAd3-RSV (B) IM PanAd3-RSV

Taylor et al Science Tr Med 2015
Green et al Science Tr med 2015

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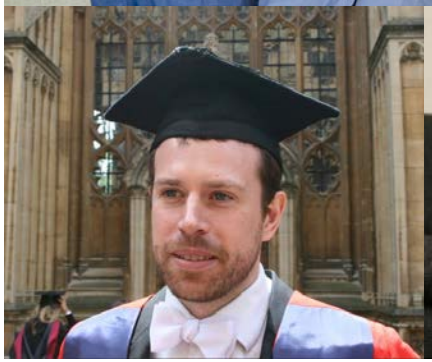

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