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LABORATORY MEDICINE & PATHOLOGY

High Throughput Analysis of B Cell Dynamics and Neutralizing Antibody Development During Immunization with a Novel Clade C HIV-1 Envelope

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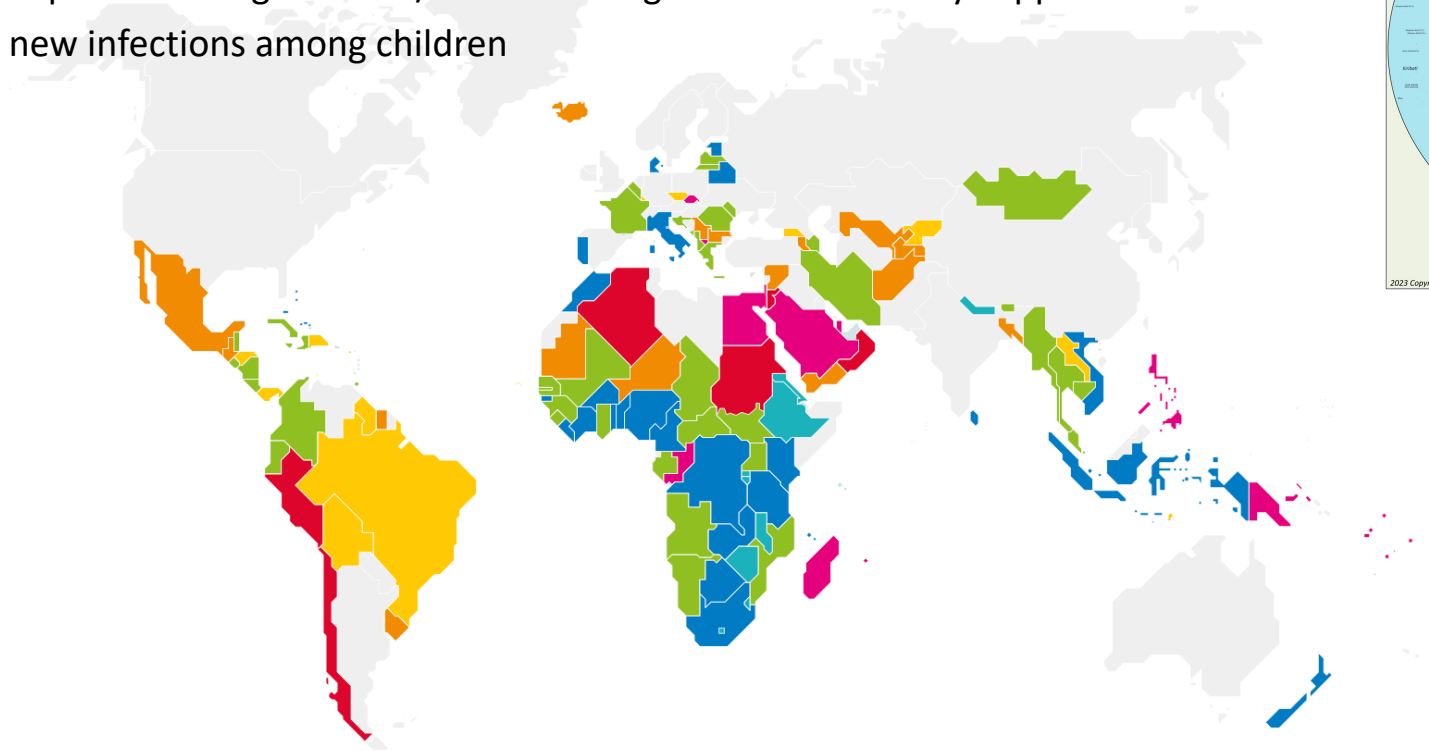
HIV/AIDS in perspective, 2022

- 39 million persons living with HIV/AIDS in 2022, 26 million in Africa
- 1.3 million new infections
- One life lost to AIDS every minute (630,000)
- 9.2 million persons living with HIV/AIDS not receiving ART
- 2.1 million persons living with HIV/AIDS receiving ART but not virally suppressed
- 130,000 new infections among children



Decline of 70% or more | 50–69% decline | 10–49% decline | Stable (−10% to 10% change) | 10–49% increase | 50–69% increase | Increase of 70% or more

Source: UNAIDS epidemiological estimates, 2023 (https://aidsinfo.unaids.org/).

Increasing infections:
Eastern Europe
Central Asia
Middle East
North Africa
Asia and the Pacific

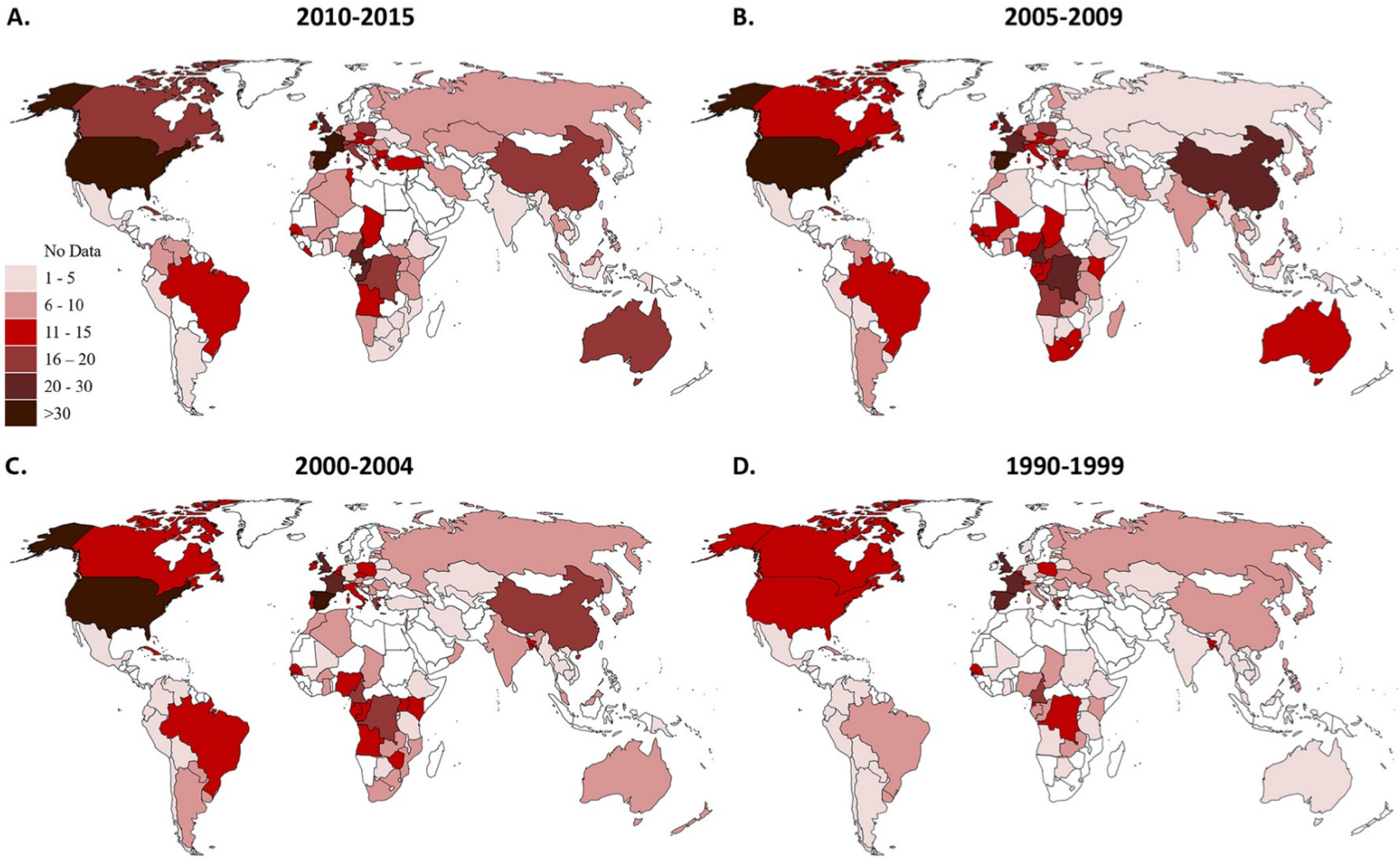
Source: 2022 data, *UNAIDS Global Update* 2023

Is a protective HIV vaccine possible?

Vaccine Trial	Main Components	Years	Locations	Cohort	Outcome
VAX 004	Bivalent Env gp120 protein, , Clade B	1999-2003	US	HIV discordant couples	No efficacy
VAX 003	Bivalent Env gp120 protein, Clade B and CRF01_AE	1999-2003	Thailand	Intravenous drug users	No efficacy
HVTN 503	Adenovirus type 5 expressing viral genes, no Env	2005-2007	US	Men who have sex with men	Stopped for futility
HVTN 504	Adenovirus type 5 expressing viral genes, no Env	2005-2007	South Africa	Heterosexual individuals	Stopped for futility
RV144	ALVAC Canarypox and Bivalent Env gp120 protein, Clade B and CRF01_AE	2004-2010	Thailand	Community-based	31% efficacy, but controversial and not reproducible
HVTN 505	DNA and Adenovirus type 5 expressing viral genes, no Env	2010-2013	US	Men who have sex with men	Stopped for futility
HVTN 702	ALVAC Canarypox and Bivalent Env gp120 protein, Clade C	2016-2020	South Africa	Heterosexual individuals	Stopped for futility
HVTN 705	Adenovirus type 26, 4 mosaic sequences, Env gp140 protein, Clade C, Imbokodo	2017-2022	Sub-Saharan Africa	Women	Stopped for futility
HVTN 706	Adenovirus type 26, 4 mosaic sequences, Env gp140 protein, Clade C, MOSAICO	2019-2024	Americas and Europe	Men and transgender persons	Stopped for futility
HVTN 703	IV infusions of bnAb VRC01, AMP study	2016-2021	Sub-Saharan Africa	Women	No efficacy
HVTN 704	IV infusions of bnAb VRC01, AMP study	2016-2021	US, Switzerland, South America	Men and transgender persons	No efficacy

AMP study finding: Most circulating strains from two major clades B and C are resistant to neutralization by bnAb VRC01, which was isolated in 2010 from a clade B subject who had been infected for more than 15 years

Global HIV-1 diversity is an increasing challenge for vaccines



AMERICAN SOCIETY FOR MICROBIOLOGY | Journal of Virology®

GENETIC DIVERSITY AND EVOLUTION

Country Level Diversity of the HIV-1 Pandemic between 1990 and 2015

Joris Hemelaar,[a,b] Shanghavie Loganathan,[a] Ramyiadarsini Elangovan,[a] Jason Yun,[a] Leslie Dickson-Tetteh,[a] Shona Kirtley,[c] WHO-UNAIDS Network for HIV Isolation and Characterization

[a]Nuffield Department of Women's and Reproductive Health, University of Oxford, Women's Centre, John Radcliffe Hospital, Oxford, United Kingdom
[b]Nuffield Department of Population Health, University of Oxford, Oxford, United Kingdom
[c]Centre for Statistics in Medicine, Nuffield Department of Orthopaedics, Rheumatology, and Musculoskeletal Sciences, University of Oxford, Oxford, United Kingdom

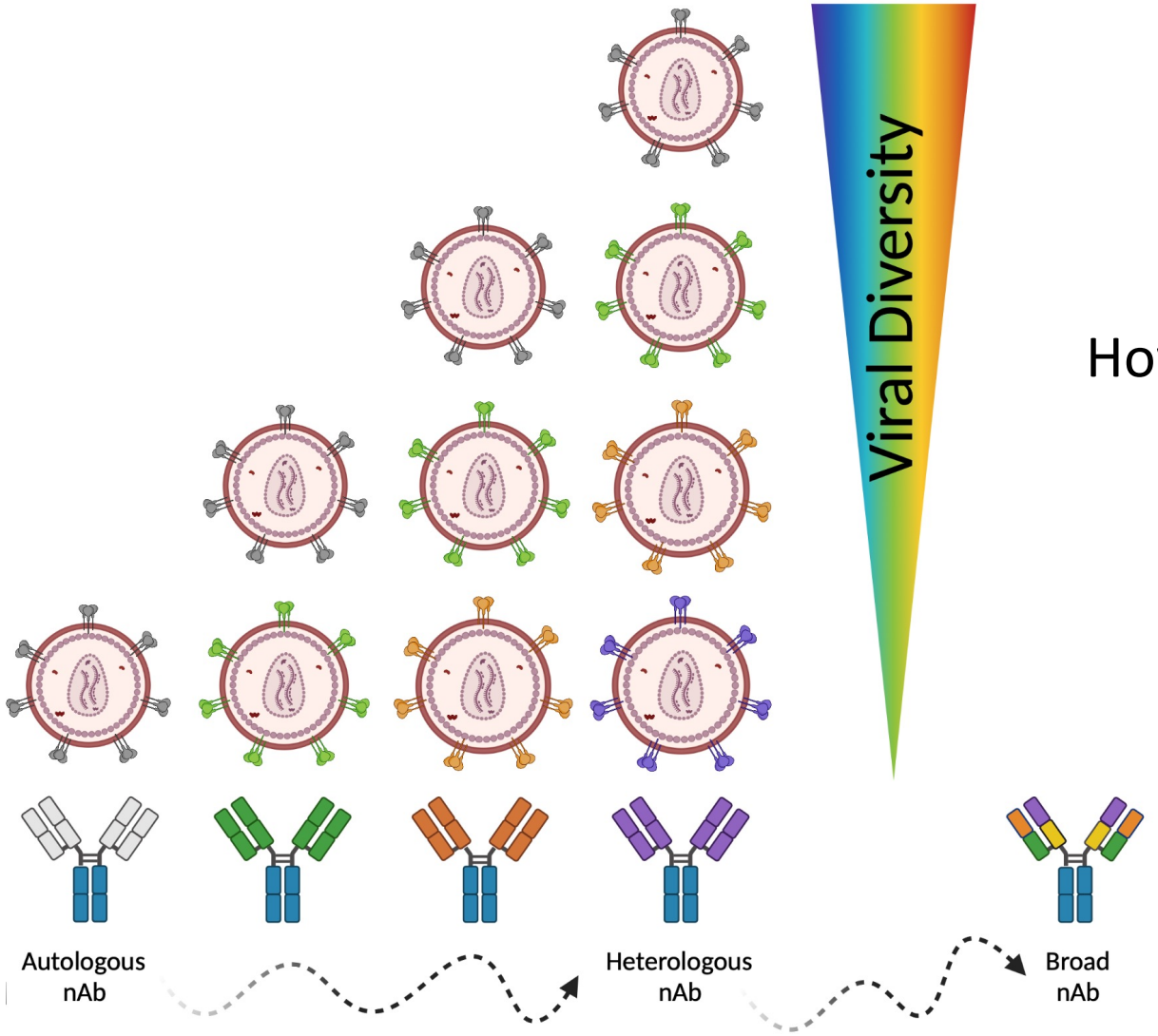
This study by Hemelaar, published in 2020, demonstrated that HIV is diversifying at a country level

This highlights the increasing challenges to HIV vaccine development, use of passive bnAbs, diagnostic assays, drug resistance, and detection of virus

Neutralizing antibodies are key to vaccine induced protection against HIV-1 and other viral pathogens

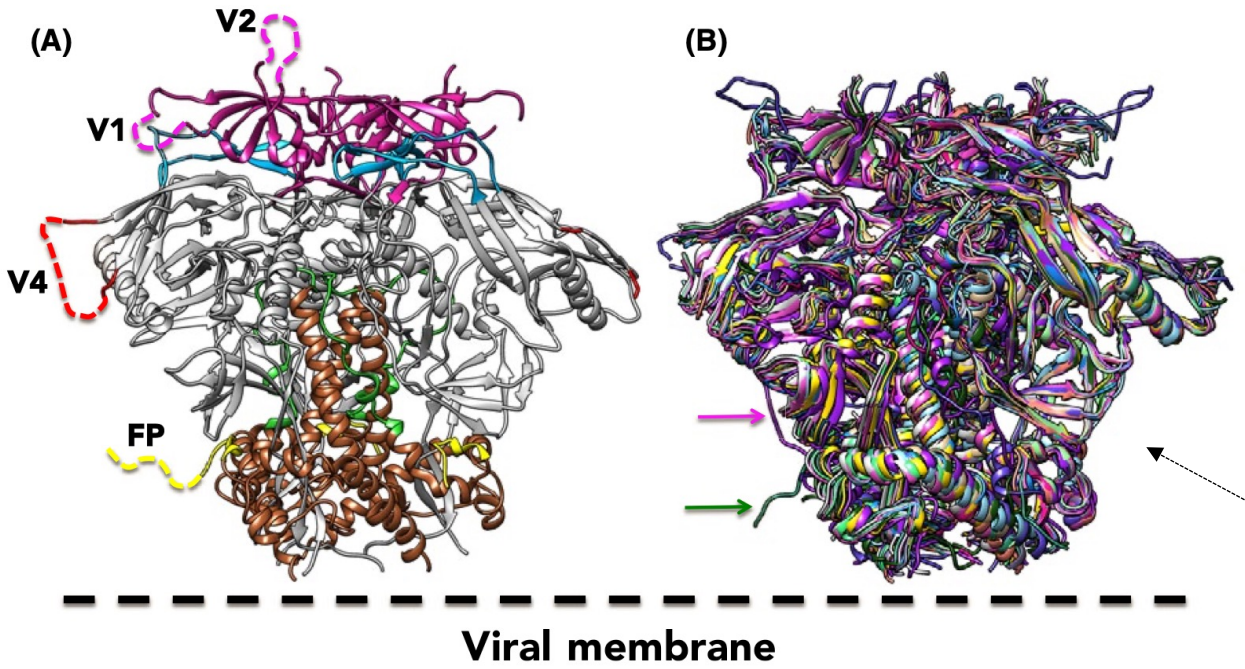
- Target the envelope (Env) glycoproteins gp120 and gp41 (ectodomain)
- Passively administered neutralizing antibodies protect against challenge with a susceptible HIV/SHIV variant in nonhuman primate models
- In vitro neutralization IC50 titer is generally predictive of passive protection
- In the AMP study, VRC01 had better efficacy against sensitive viruses
- Recent studies have shown a correlation between high levels of neutralizing antibodies elicited by trimeric envelope (Env) immunogens and protection against autologous virus (SHIV) in nonhuman primate models
- Neutralizing antibodies are a common correlate of vaccine-mediated protection against various viral pathogens (measles, SARS-CoV-2, HPV, flu, etc.)

HIV broadly neutralizing antibodies (bnAbs), which sometimes develop after many years of infection, are the 'holy grail' for a vaccine



How to elicit these?

The HIV-1 stabilized BG505 SOSIP envelope (Env) gp140 trimer (various versions) is a prototype, widely used immunogen



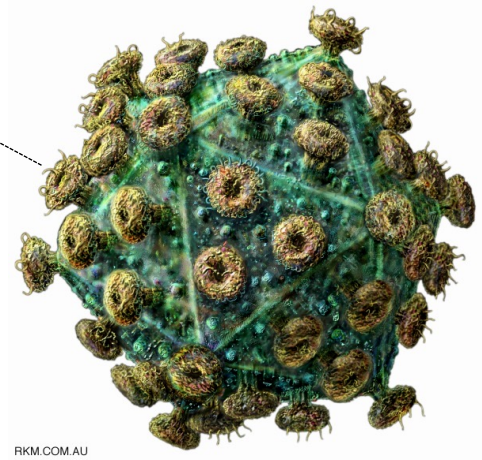
AB Ward and IA Wilson, *Immunological Reviews* 2017

This immunogen is in multiple phase I trials

16 high resolution crystal structures

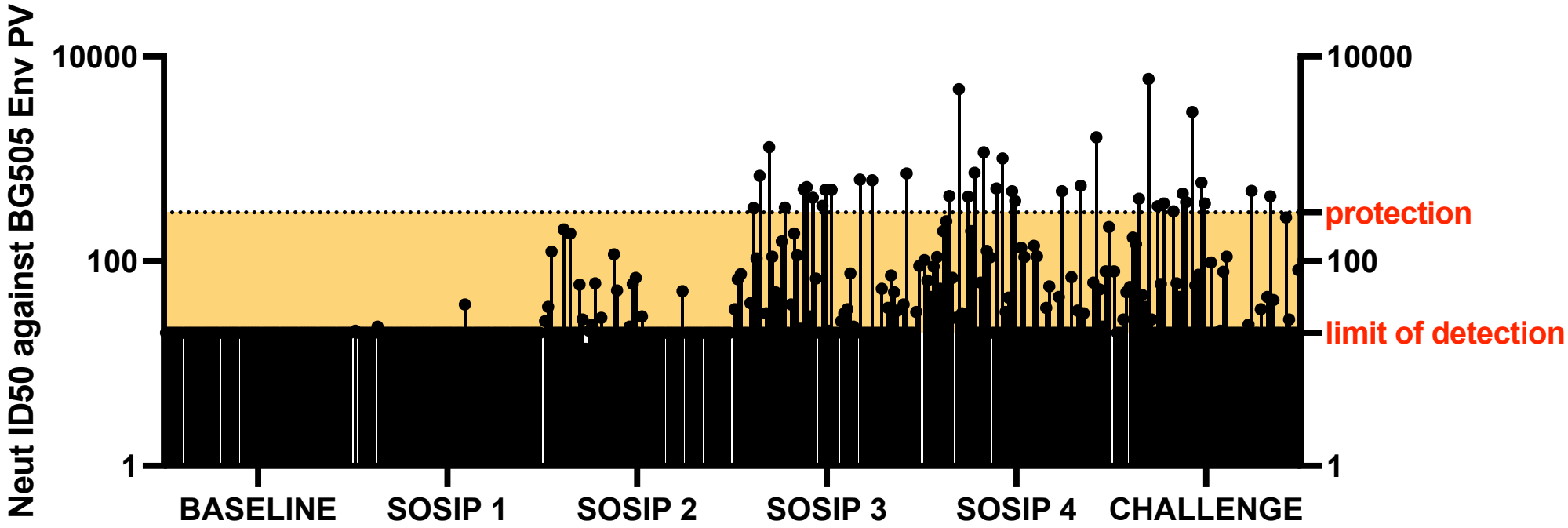
More than 100 peer reviewed articles

Early results have revealed challenges in eliciting neutralizing antibody responses in humans although only one study has published results to date (VRC)



R Sanders et al., *PLoS Pathogens* 2013; J Julien et al., *Science* 2013

BG505 SOSIP immunization of rhesus macaques leaves many with serum neutralization titers below the threshold of protection

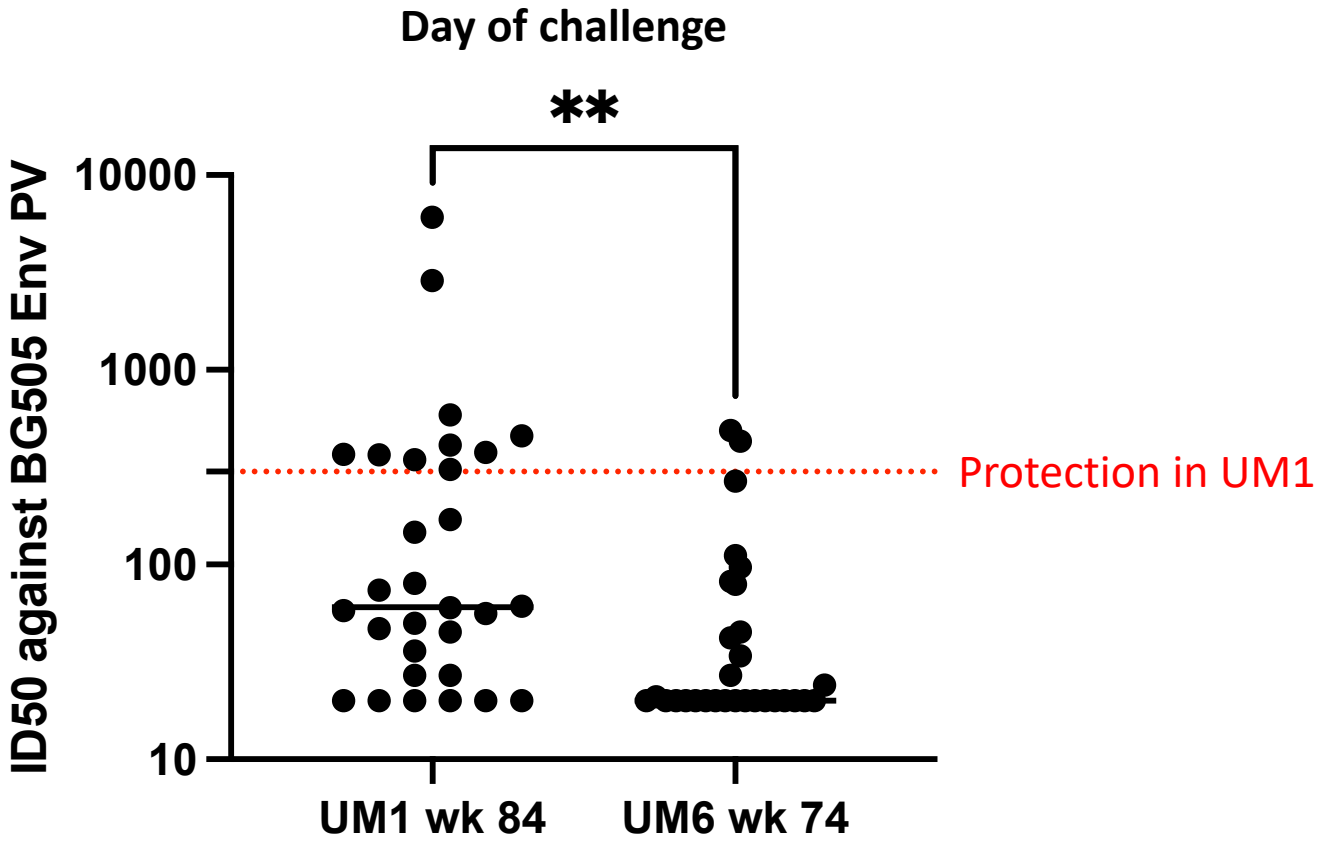


UM1 30 immunized rhesus macaques
UM6 30 immunized rhesus macaques
25/60 were at the limit of detection (ID50 titer of 20) on the day of challenge
12/60 were above the threshold of protection (ID50 of ~300) for SHIV.BG505

P Ralli-Jain, D Rosales, A Aftab, *unpublished data*

P Aruchalanam, T Charles, et al., *Nature Medicine 2020*
T Charles et al., *PLoS Pathogens 2021*

Neutralization titers using the same BG505 SOSIP immunogen and adjuvant at the same primate center vary across trials



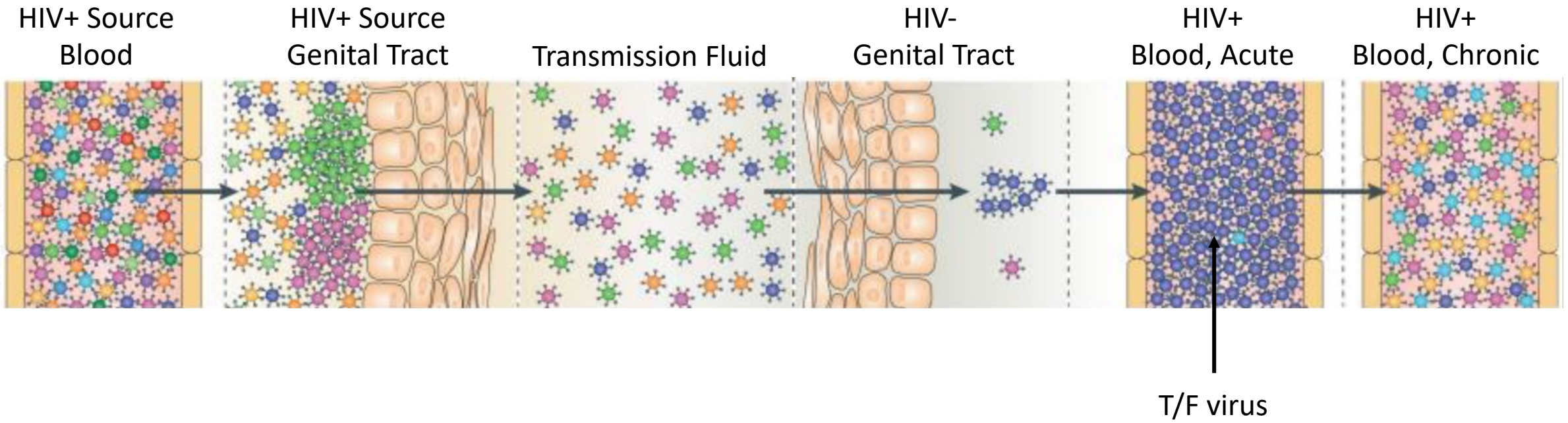
p = 0.0012, Mann-Whitney test

P Ralli-Jain, D Rosales, A Aftab, *unpublished data*

P Aruchalanam, T Charles, et al., *Nature Medicine 2020*
T Charles et al., *PLoS Pathogens 2021*

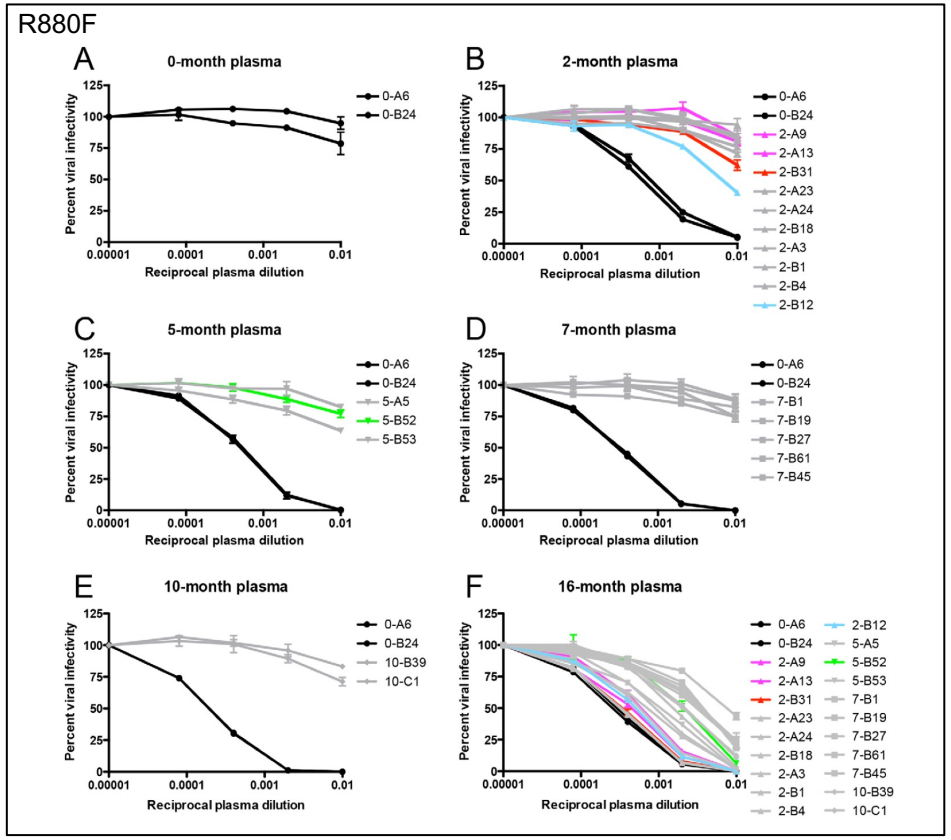
A major interest for my lab that has evolved over the years is how to consistently elicit robust autologous tier 2 neutralizing antibodies with HIV-1 Env immunogens and vaccines

Taking a step back: transmitted/founder (T/F) HIV-1 variants are those that establish a new infection and subsequently drive development of neutralizing antibody responses and viral escape

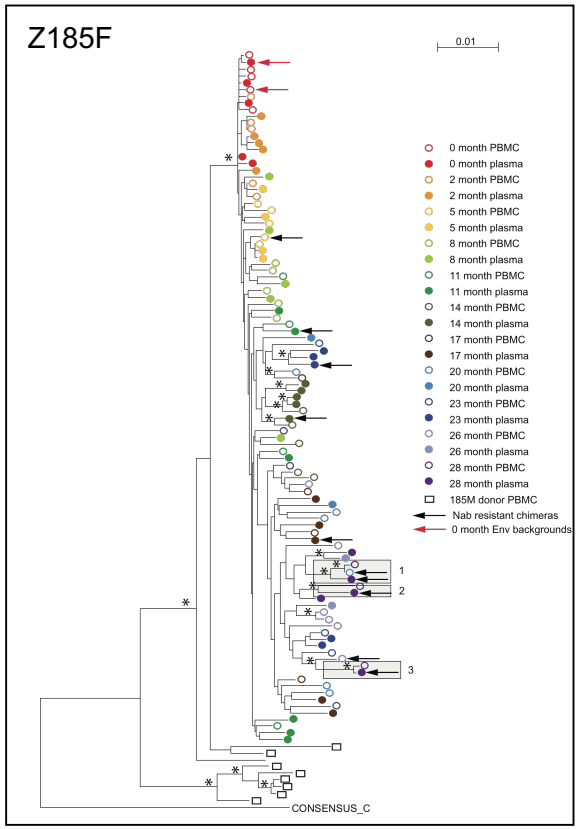


Joseph, Swanstrom, Kashuba, and Cohen, *Nature Reviews Microbiology* 2015

HIV-1 escapes from neutralizing antibodies over time by increasing diversity in the envelope (Env), mainly in gp120



M Murphy et al., *PLoS Pathogens* 2013



R Rong et al., *PLoS Pathogens* 2009

The T/F Env initiates the neutralizing antibody response, which in turn drives evolution of the viral quasispecies

Contemporaneous Env variants are resistant to plasma neutralization because they have 'escaped' the neutralizing antibody targets

Escape includes amino acid changes, insertions, deletions, and changes in N-linked glycosylation

The B cells catch up by continuing to evolve through somatic hypermutation and other mechanisms

PLWH develop vastly different levels of plasma neutralizing antibody breadth (sometimes bnAbs); what role do the T/F Env and escape variants play?

Plasma ID50 neutralization titers against a global panel of HIV-1 Envs

Persons living with HIV -1 in Africa whose plasma was tested at approx. 3 years after infection occurred

	CRF01	A	C	B	B	G	CRF01	CRF07	C	CRF07	AC	C	NEG		
Subject	**CNE8**	**398F1**	**25710**	**X2278**	**TRO11**	**X1632**	**CNE55**	**BJOX00200**	**CE1176**	**CH119**	**246F3**	**CE0217**	**VSVg**	**Median IC50**	**# neutralized**
Z1800M	2503	3940	1819	646	122	780	2035	1002	79	1058	1574	266	20	1030	12
Z1792M	263	4785	350	157	268	76	27	285	258	632	46	31	20	261	12
Z185F	1131	2503	541	158	226	184	59	182	235	313	72	54	20	205	12
R1141M	623	1590	177	681	93	121	159	210	64	506	47	41	20	168	12
Z1024F	1874	7424	313	183	235	129	124	137	63	20	88	54	20	133	11
R1142F	459	2019	275	280	319	129	24	87	36	60	34	28	20	108	12
Z221M	428	1493	178	89	262	50	22	32	138	48	69	31	20	79	12
Z1022M	274	1101	304	122	48	118	76	62	66	43	43	66	20	71	12
Z1023M	780	2048	325	73	73	53	92	54	60	68	39	28	20	70	12
Z1781M	536	1704	169	202	41	109	63	72	46	29	37	28	20	68	12
Z205F	733	1220	455	72	487	53	20	35	20	20	26	20	20	44	8
R53F	184	393	180	31	32	28	20	27	20	58	20	22	20	29	8
R283F	310	1570	55	79	43	20	20	20	20	20	20	34	20	27	6
R774F	97	324	44	44	63	20	22	20	20	20	20	20	20	21	5
R880F	565	440	183	20	84	34	20	26	20	20	20	20	20	23	5
Z153M	824	2189	305	20	20	20	20	22	20	35	30	20	20	21	5
R463F	179	876	55	31	42	25	20	20	20	20	20	20	20	22	5
Z1047M	116	96	46	20	20	20	20	20	20	20	20	20	20	20	3
Z201M	141	886	20	20	20	20	20	20	20	20	20	20	20	20	2
R66M	36	126	20	20	20	20	20	20	20	20	20	20	20	20	2
R1135M	20	20	20	20	20	20	20	20	20	20	20	20	20	20	0
Median	428	1493	180	73	63	50	22	32	20	29	30	28	20	44	8

Strong nAb

Moderate nAb

Weak or no nAb

S Smith et al., *PLoS Pathogens*, 2016

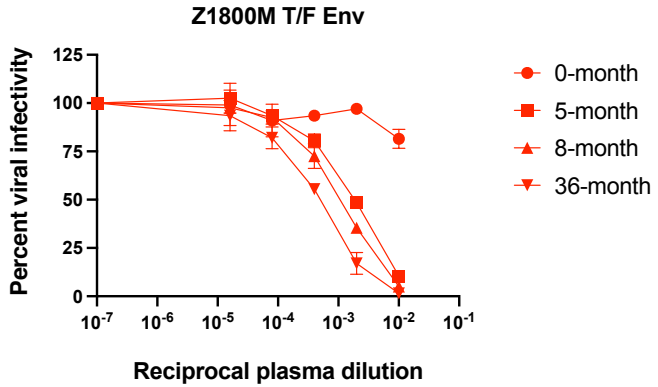
We focused on T/F Envs from two PLWH with distinct outcomes

Autologous neutralization

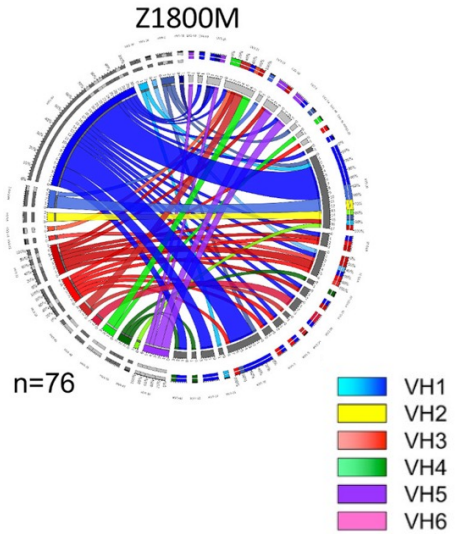
Antigen specific B cell diversity

Z1800M Tier 2 Clade C T/F Env

High neutralization breadth

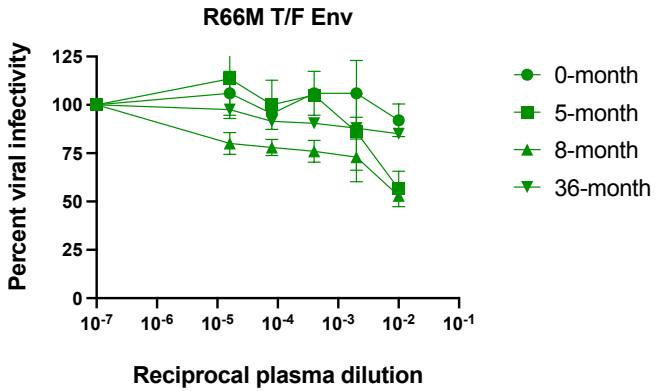


High Viral Immunotype Diversity

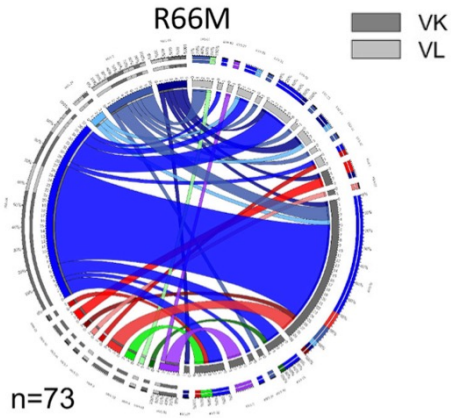


R66M Tier 2 A/C URF T/F Env

Low neutralization breadth



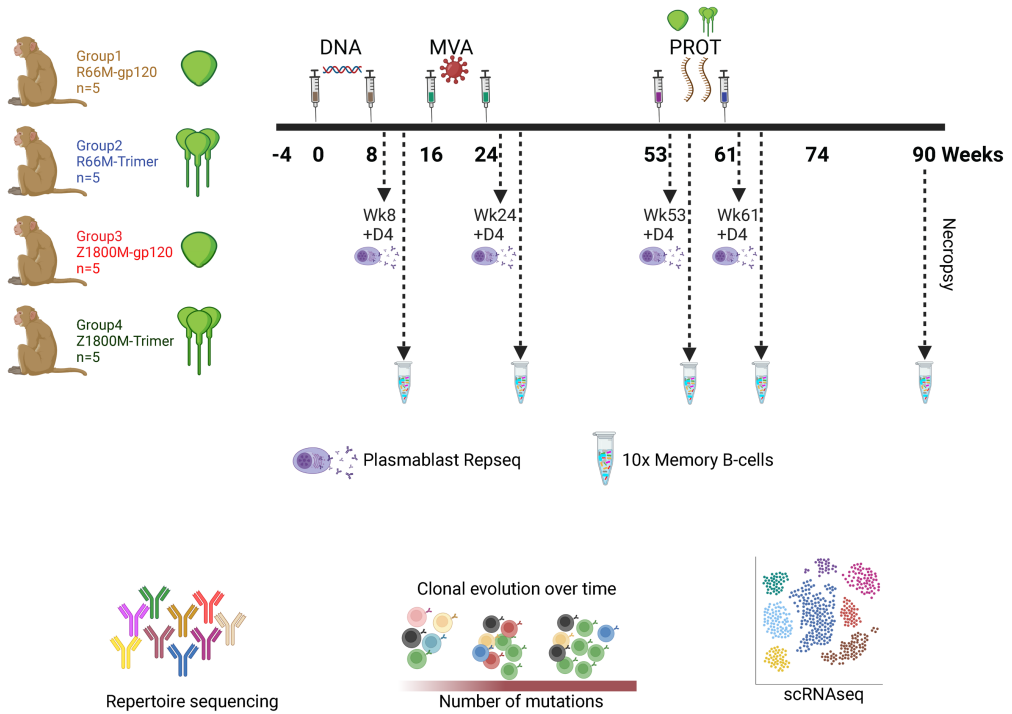
Low Viral Immunotype Diversity

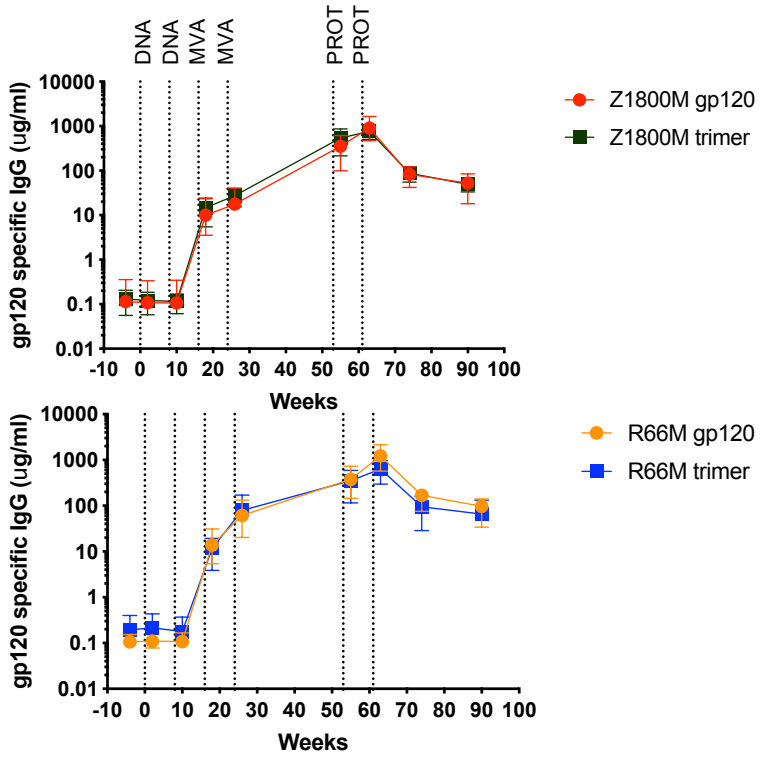


S Smith et al., *PLoS Pathogens*, 2016

S Smith et al., *Front Immunology*, 2019

A vaccination study was carried out in rhesus macaques using DNA, MVA, and Env gp140 trimer or gp120 monomer based on the two T/F Envs





Group	Animal	Week 18	Week 26	Week 55	Week 63	Week 74
R66M gp120	RVh17	20	20	20	20	20
	RGi17	20	20	20	20	20
	RTe17	20	20	20	20	20
	RLf17	20	20	20	20	20
	RLl17	20	20	20	20	20
R66M trimer	RWb17	20	20	20	20	20
	RPh17	20	20	20	20	20
	RVe17	20	20	20	20	20
	RTm17	20	20	20	20	20
	RVn17	20	20	20	20	20
Z1800M gp120	RRb17	20	20	20	20	20
	RPz16	20	20	20	20	20
	ROa17	20	20	77	72	20
	RLk17	20	20	136	262	46
	6S0	20	20	ND	ND	ND
Z1800M trimer	15C172	20	20	20	20	20
	RFj17	20	20	20	20	20
	RSb17	20	20	20	20	20
	RYf17	20	20	20	20	20
	MA277	20	20	20	20	20
NEG	NS	20	20	20	20	20

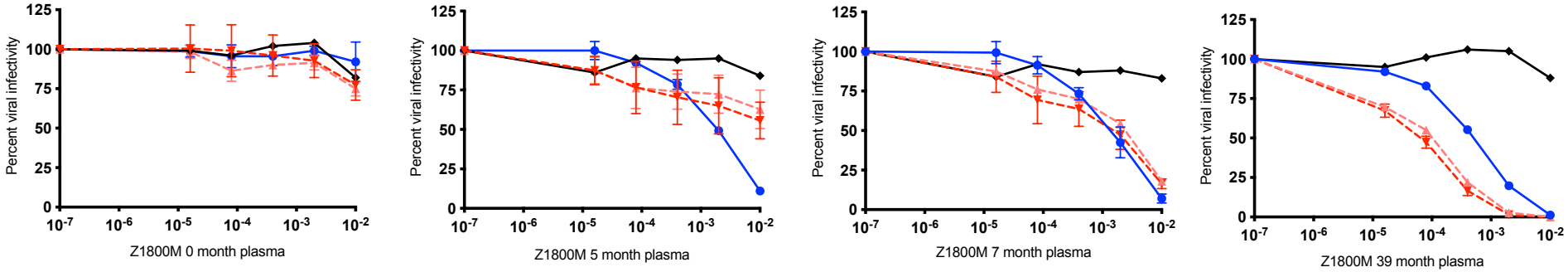
All RM developed high titers of Env-specific IgG

Only 2 RM in group 3 developed serum nAb

S Welbourn et al., *PLoS Pathogens* 2022

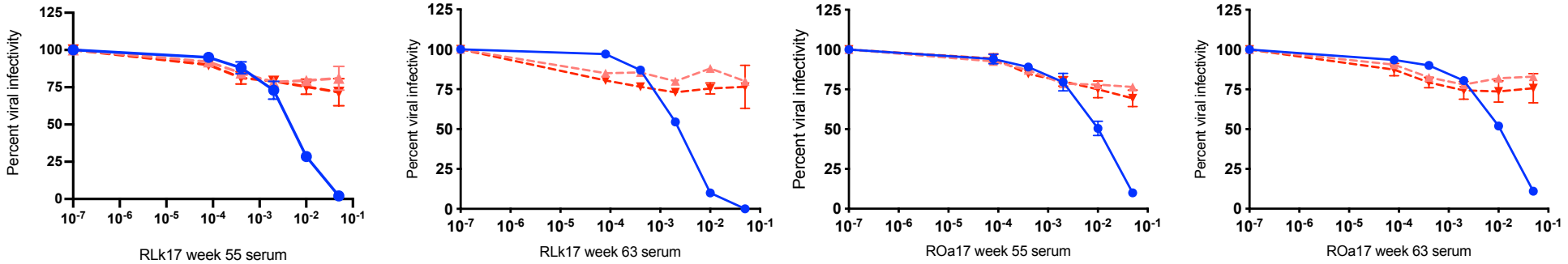
Testing two 5-month escape variants from PLWH Z1800M, Envs D10 and D11, revealed that nAb elicited by immunization mimics early HIV-1 infection

Longitudinal plasma from PLWH Z1800M shows 5-month contemporaneous viral escape



Months: 0 5 7 39

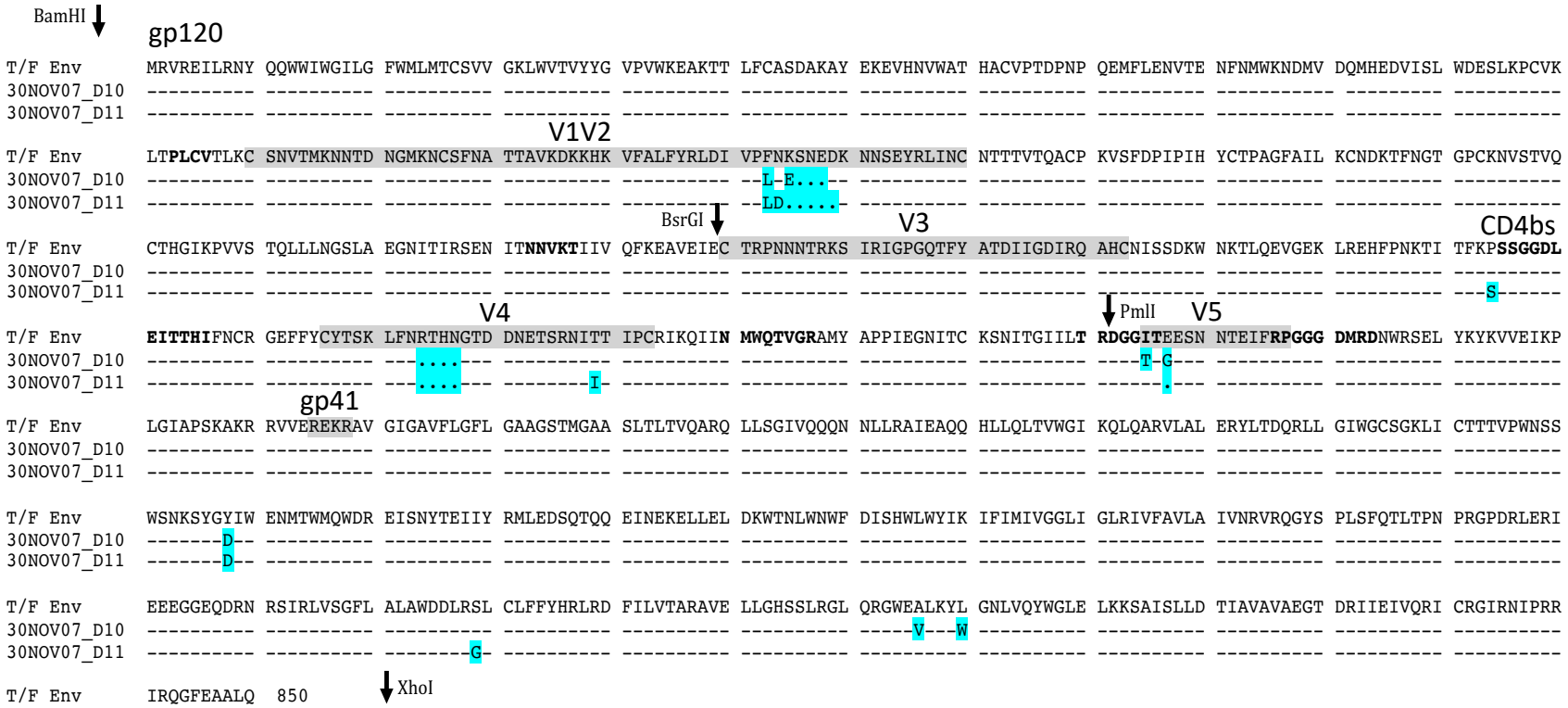
Immunized RM also can neutralize the T/F Env but not the 5-month Envs D10 and D11

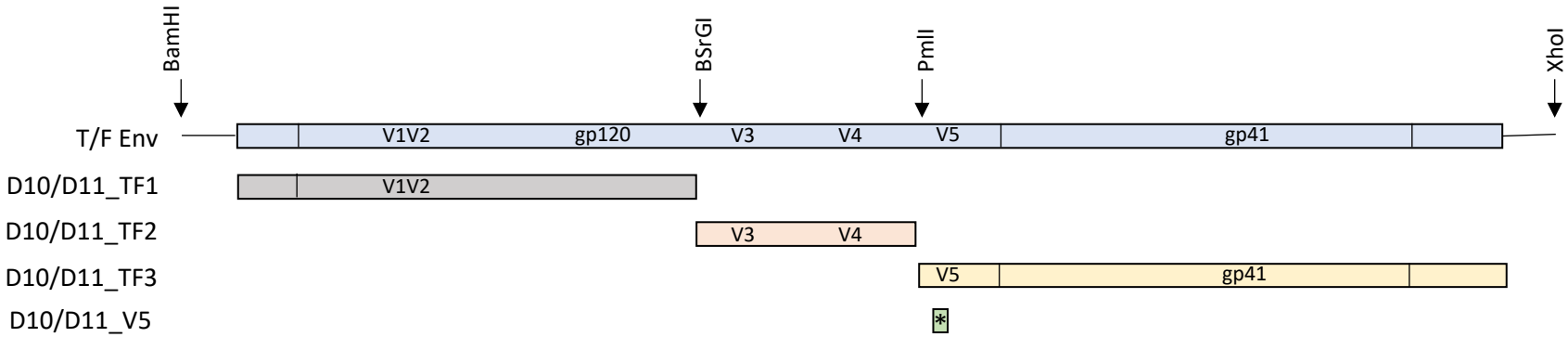


RLk17 ROa17

S Welbourn et al., *PLoS Pathogens* 2022

The Z1800M 5-month Envs D10 and D11 evolved from the T/F Env in gp120 regions V2, V4, CD4bs, V5, and gp41

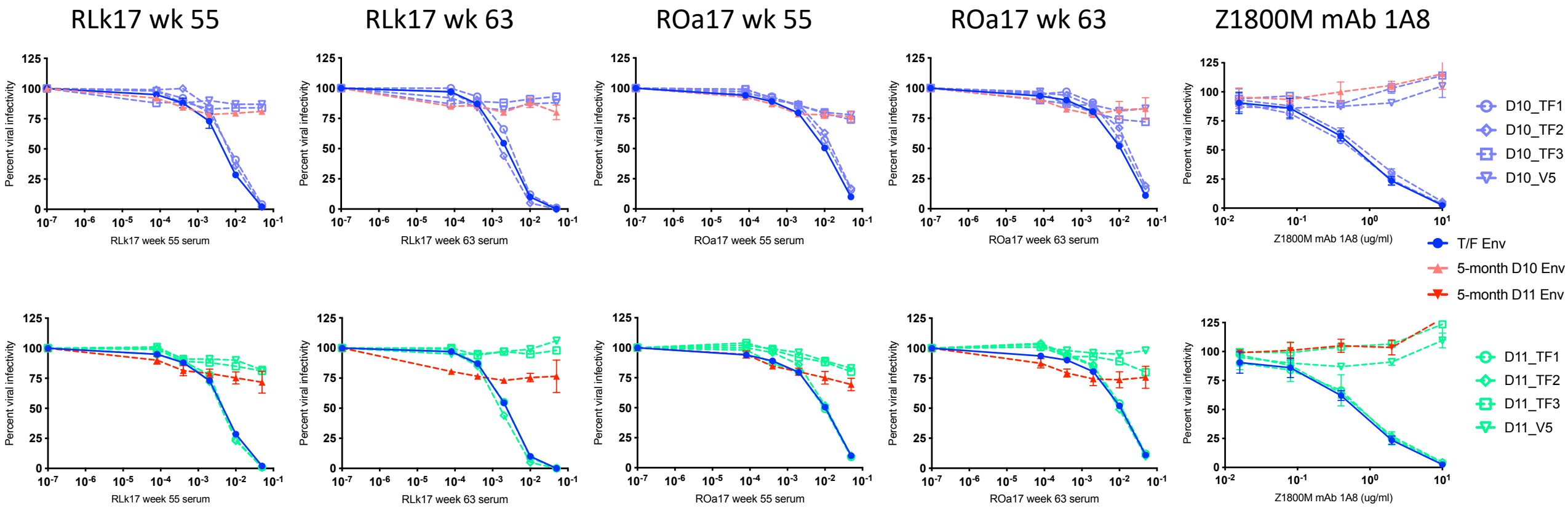




S Smith et al., *PLoS Pathogens* 2016

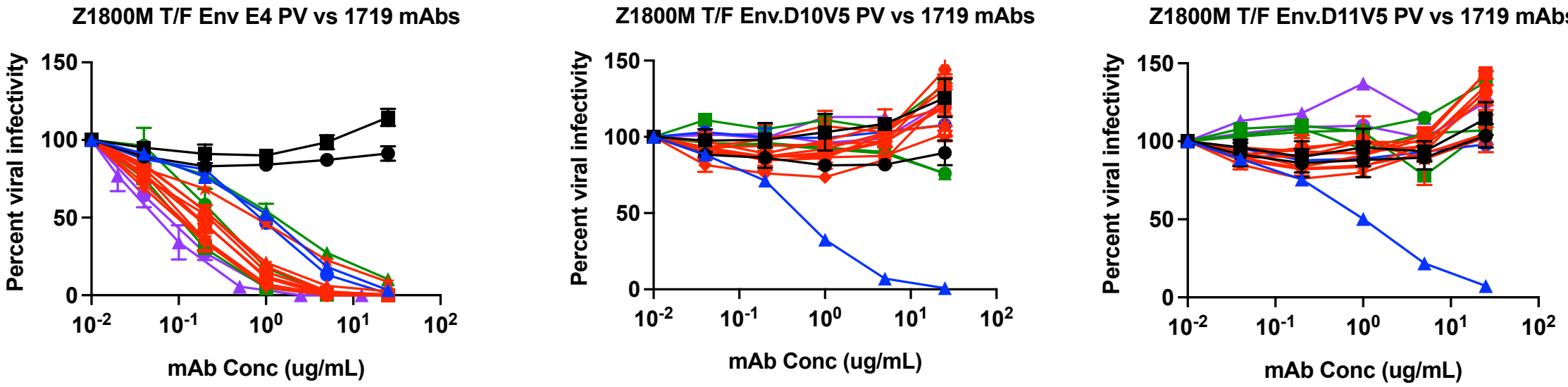
S Welbourn et al., *PLoS Pathogens* 2022

Resistance to serum neutralization was dependent on two residue changes (D10) or one residue deletion (D11) in V5 similar to a monoclonal antibody from PLWH Z1800M



S Welbourn et al., *PLoS Pathogens* 2022

Monoclonal antibodies (mAbs) from RLk17 (lineage 1719) at weeks 26, 63, and 90 also neutralized Z1800M T/F Env by targeting V5/CD4bs



Unmutated rearranged precursor

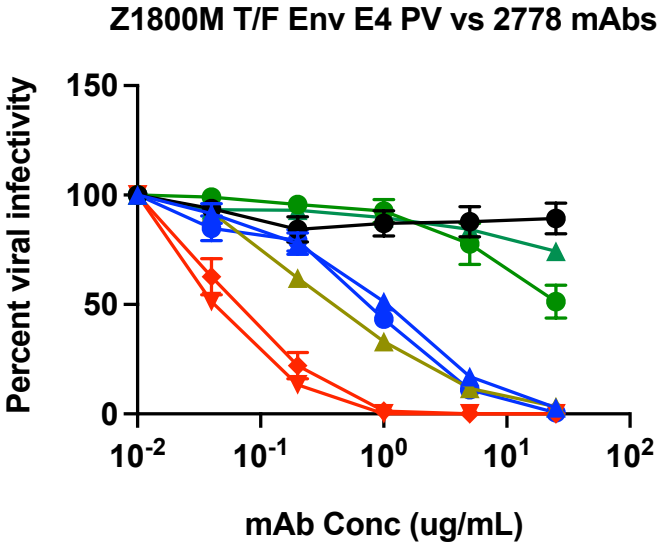
- Z1800M 1A8
- VRC01
- EM4C04
- wk 63, 2B12
- 1719 wk 26, UW4-1
- 1719, wk 26, UW4-2
- 1719, wk 26, UW4-3
- 1719, wk 63, V5.1B1
- 1719, wk 63, V5.1B3
- 1719, wk 63, V5.1C10
- 1719, wk 63, V5.1E9
- 1719, wk 63, V5.2A1
- 1719, wk 63, V5.2D10
- 1719, wk 63, V5.2D11
- 1719, wk 63, V5.1C4
- 1719, wk 363, V5.1D7
- 1719, wk 90, UW4-4
- 1719, wk 90, UW4-5

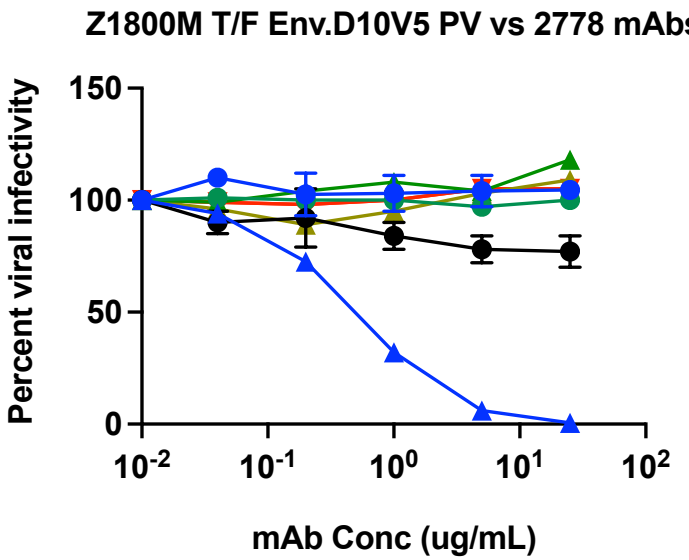
mAbs isolated by antigen specific B cell sorting, single B cell VDJ PCR, and 10X VDJ high throughput sequencing

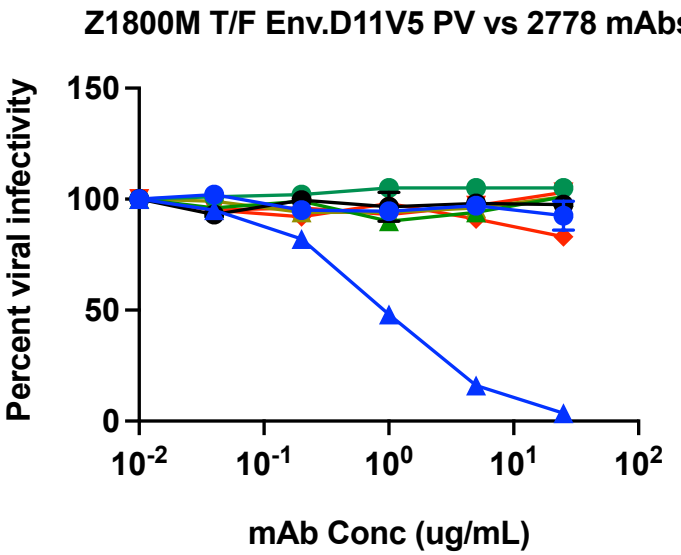
S Welbourn et al., *PLoS Pathogens* 2022

R Mopuri et al., *PLoS Pathogens* In Press

RLk17 mAbs from a second lineage (2778) also from weeks 26, 55, and 63 also neutralized by targeting V5/CD4bs







1A8

VRCO1

EM4CO4

High identity precursor

2778, wk 26, UW4-6

2778, wk 26, UW4-7

2778, wk 55, UW4-8

2778, wk 63, UW4-9

2778, wk 63, UW4-10

mAbs isolated by antigen specific B cell sorting, single B cell VDJ PCR, and 10X VDJ high throughput sequencing

S Welbourn et al., *PLoS Pathogens* 2022

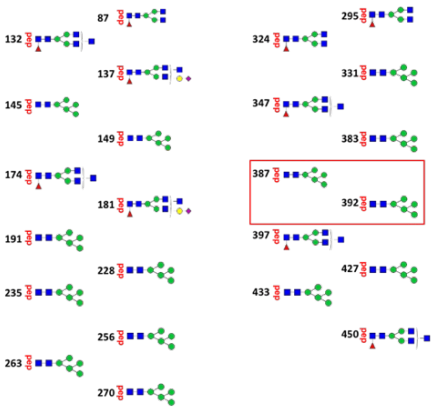
R Mopuri et al., *PLoS Pathogens* In Press

RLk17 neutralizing mAbs so far..

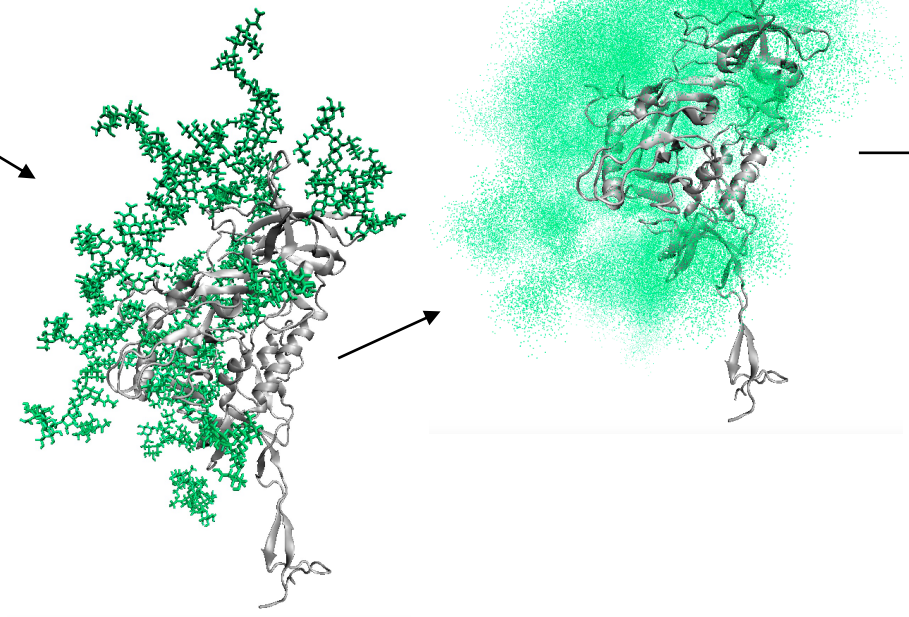
- Two independent neutralizing B cell lineages arose following MVA immunization
- Same heavy and light chain germlines
- Same target
- Unmutated rearranged or high identity precursors neutralized Z1800M T/F Env
- Neutralization potency increased with somatic hypermutation from week 26 to week 63/90
- Similar genotypically and phenotypically to the human Z1800M 1A8 monoclonal antibody (and plasma) from early infection

The V5/CD4bs region targeted by neutralizing antibodies in rhesus macaques and the PLWH is exposed in Z1800M T/F Env

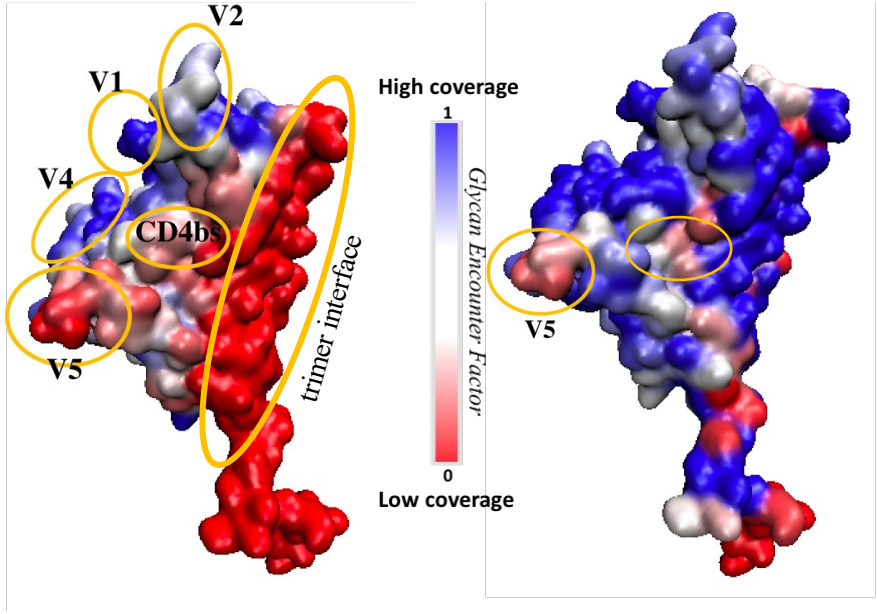
MS glycoproteomics on Z1800M T/F Env gp120



Computational glycan dynamics



Static glycan model



gp120 monomer

modeled as a trimer

CD4bs = a desirable bnAb target

Carbohydrate profile via mass spec by Drs. Anne Gleinich and Parastoo Azadi, University of Georgia Complex Carbohydrate Center

Computational modeling of the glycan shield, Drs. Srirupa Chakraborty and Gnana Gnanakaran, Los Alamos National Laboratory

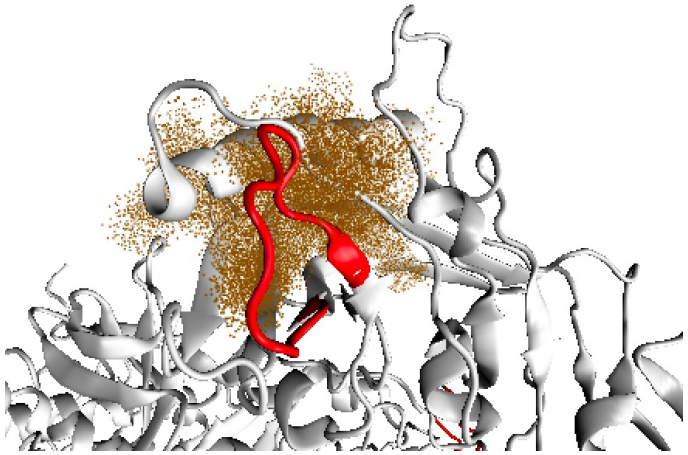
S Welbourn et al., *PLoS Pathogens* 2022

The exposed V5 loop in the T/F Env becomes shielded by the glycan at N450 when the proximal residues are mutated or deleted, representing a novel multi-pathway escape mechanism

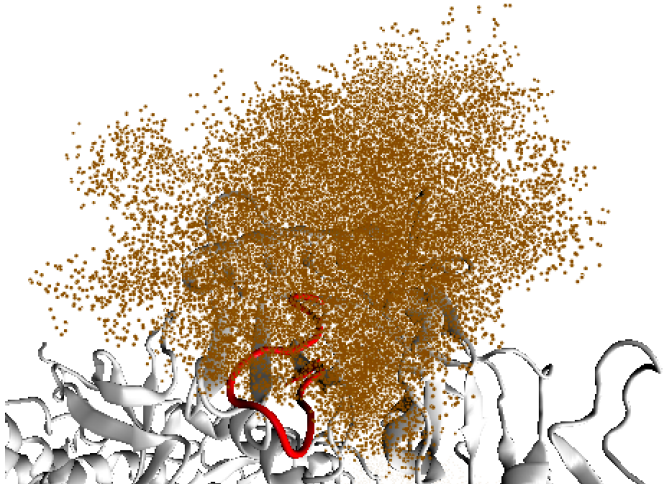
V5 loop

Glycan at N450

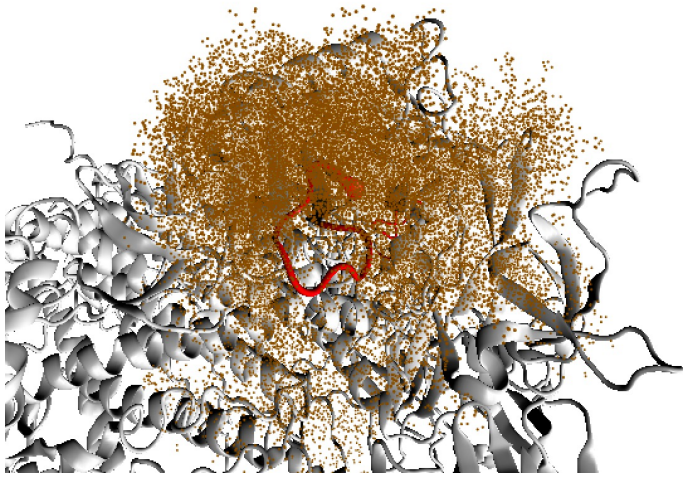
T/F Env



Env D10



Env D11



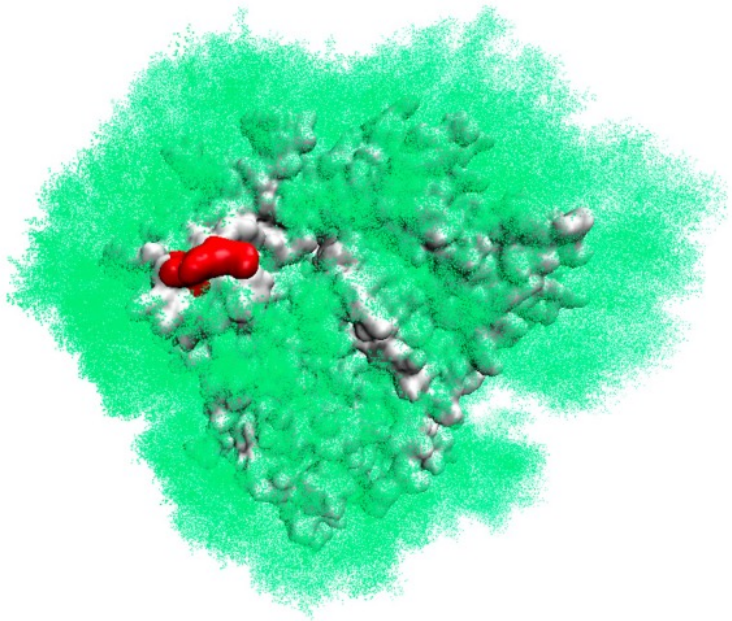
	CD4bs	V5	CD4bs
T/F Env	**TRDGGITEESNNTEIFRPGGGDMRD**		
5-month D10	-----T-G--------------		
5-month D11	-------.--------------		

S Welbourn et al., *PLoS Pathogens* 2022

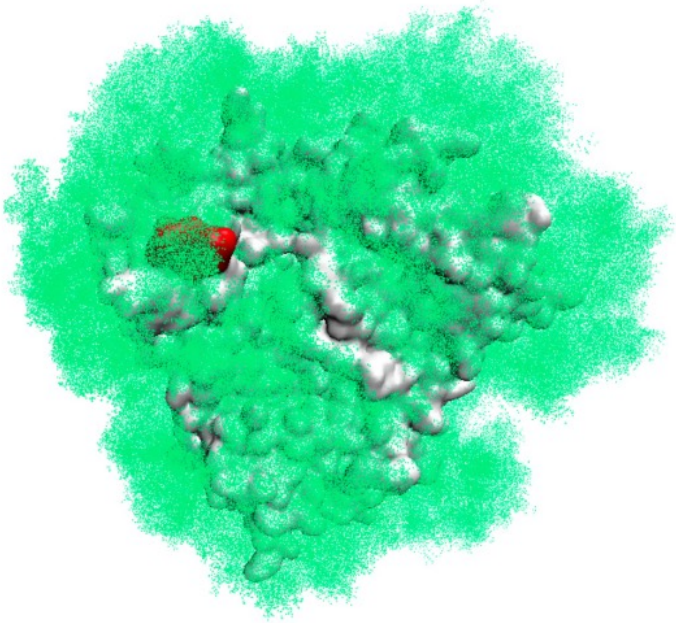
Mass Spec glycan profiling, computational glycan modeling

The dynamic glycan shield does not cover this glycan hole or the proximal CD4bs in the T/F Env leaving it vulnerable to neutralizing antibodies

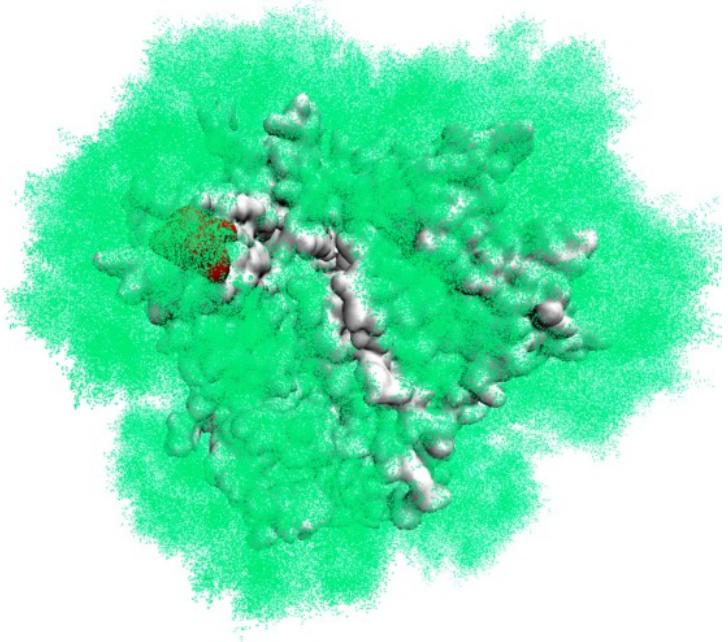
T/F Env



Env D10



Env D11

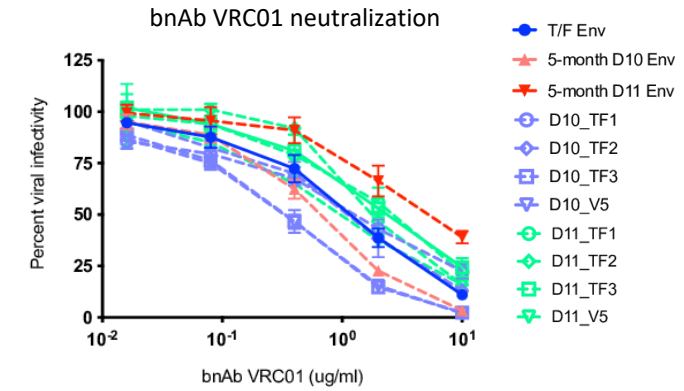


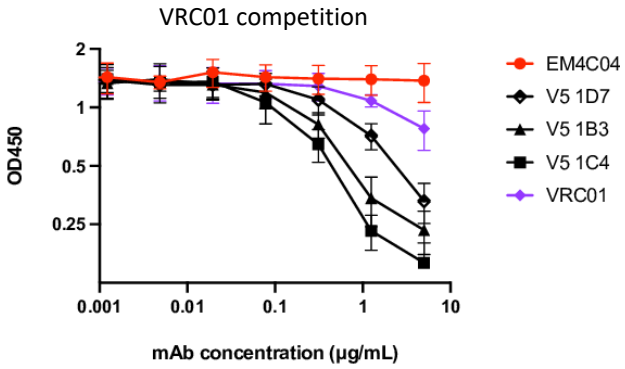
```
                    CD4bs          V5          CD4bs
T/F Env             TRDGGITEESNNTEIFRPGGGDMRD
5-month D10         -----T-G----------------
5-month D11         -------.----------------
```

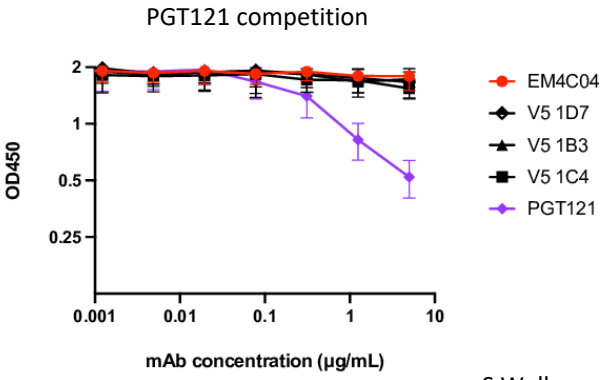
S Welbourn et al., *PLoS Pathogens* 2022

Mass Spec glycan profiling, computational glycan modeling

These vaccine elicited mAbs are not broadly neutralizing but they do target the CD4bs and potentially overlap with VRC01 contact residues

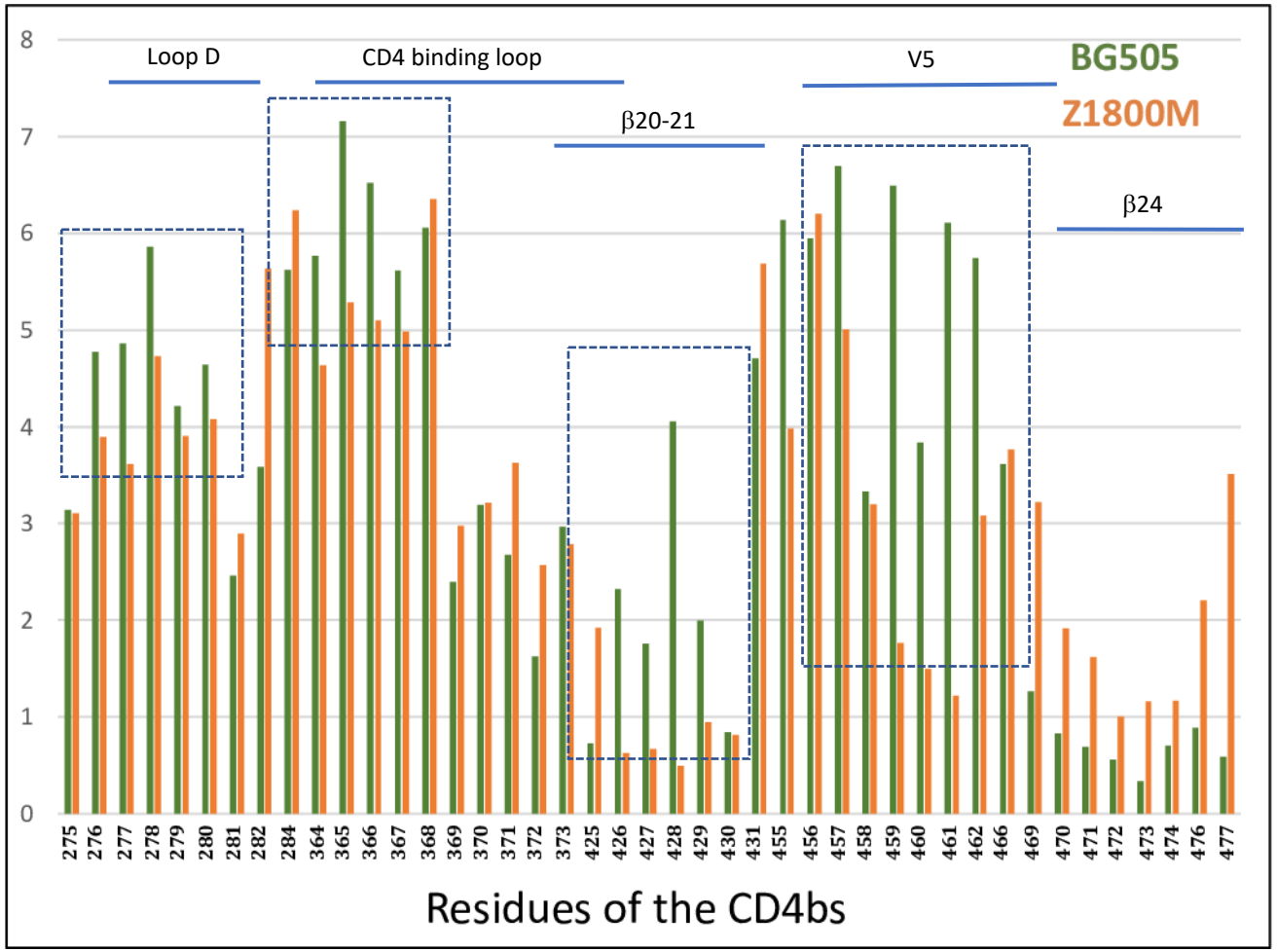




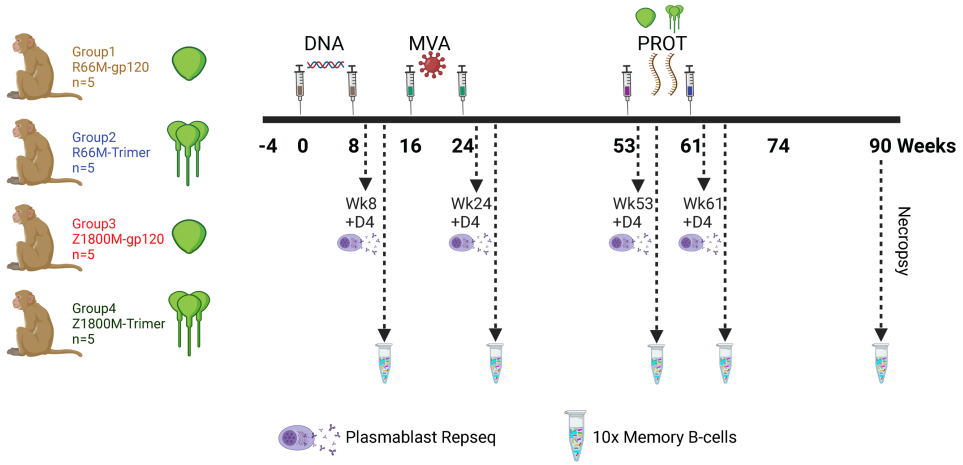


S Welbourn et al., *PLoS Pathogens* 2022

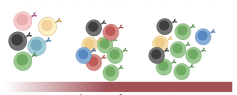
The CD4bs/VRC01 epitope is more exposed in Z1800M Env than in BG505 SOSIP

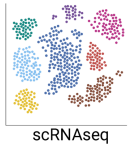


High throughput single cell VDJ analysis in all RMs revealed widespread persistence of non-neutralizing B cell lineages with a notable lack of DNA primed lineages after MVA and protein immunizations

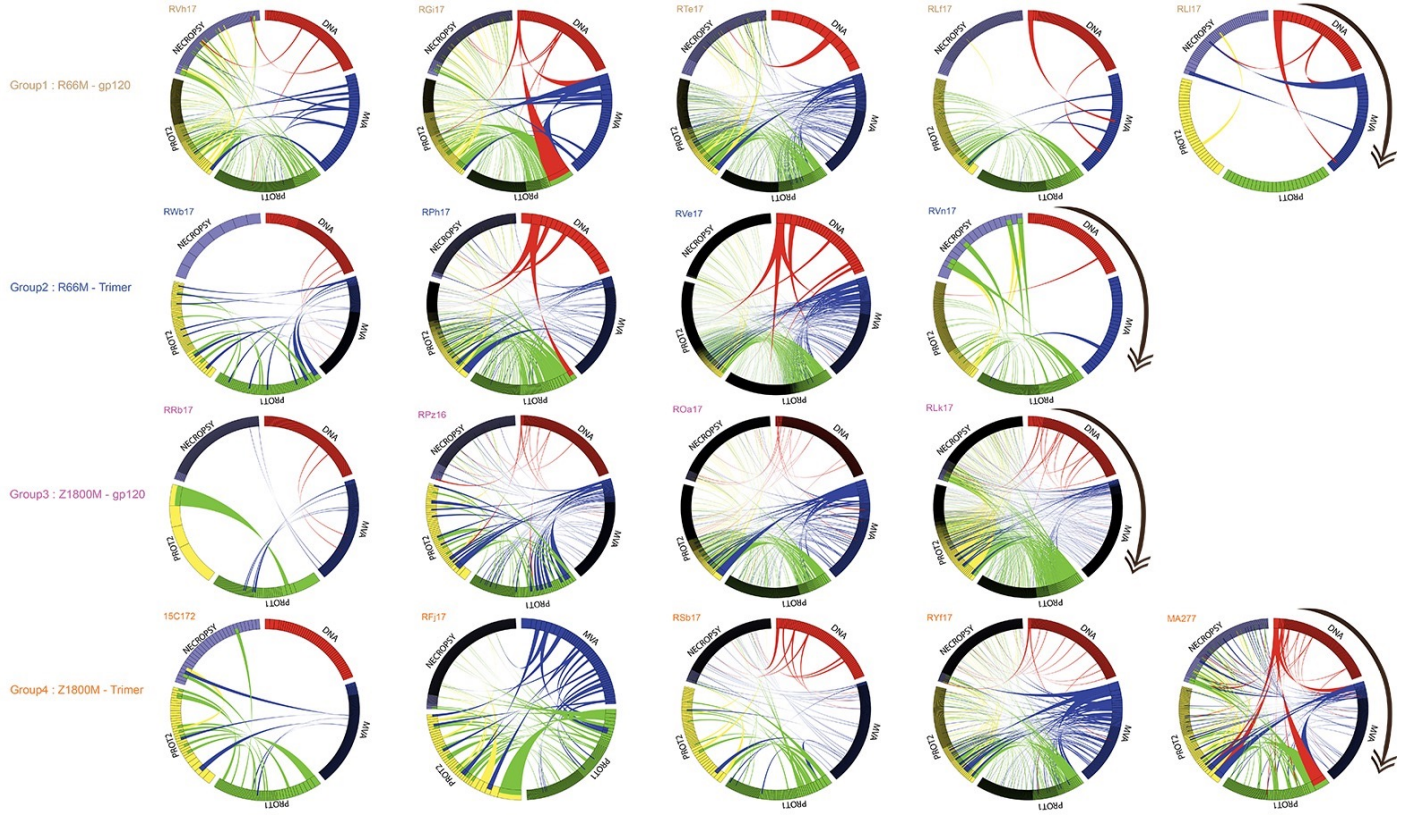


Repertoire sequencing





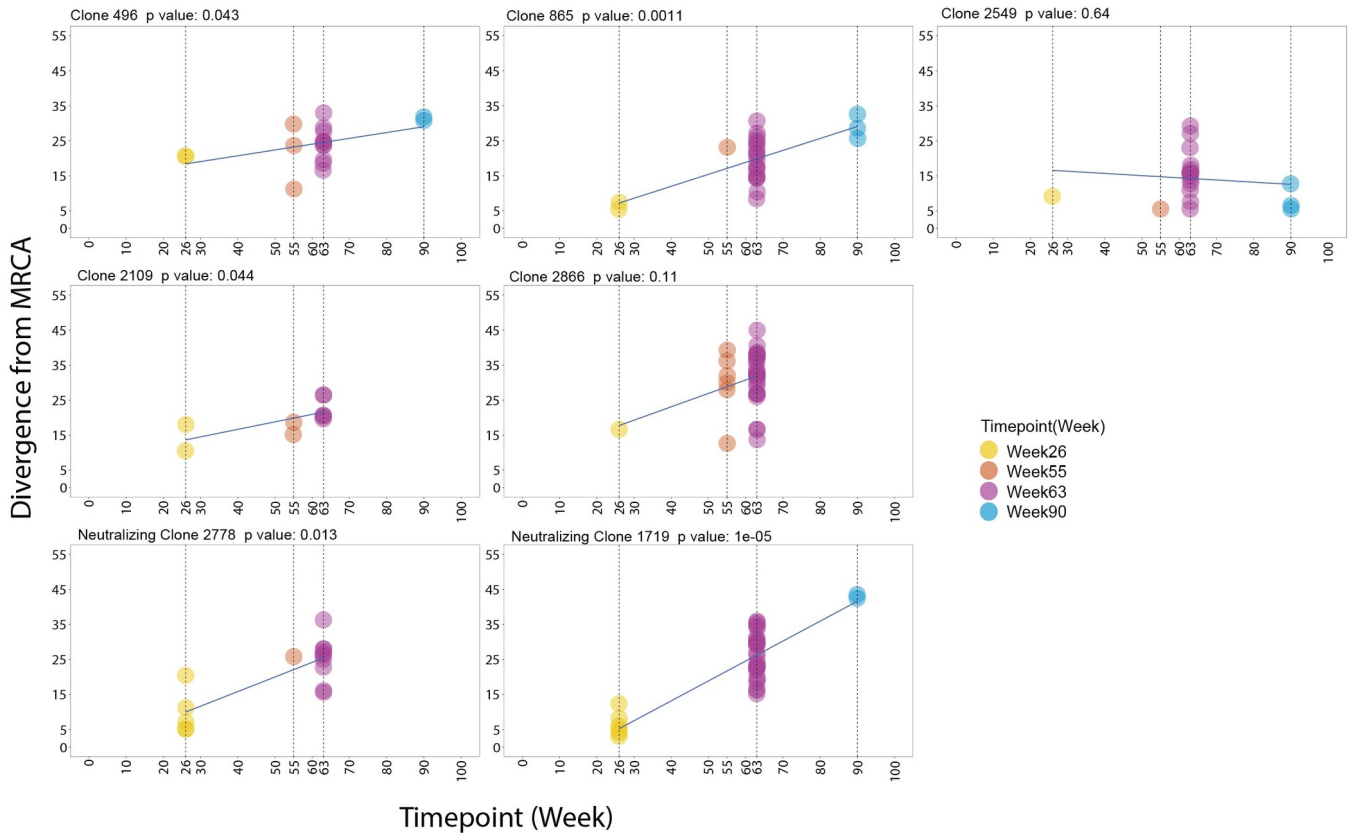
scRNAseq



R Mopuri et al., *PLoS Pathogens* In Press

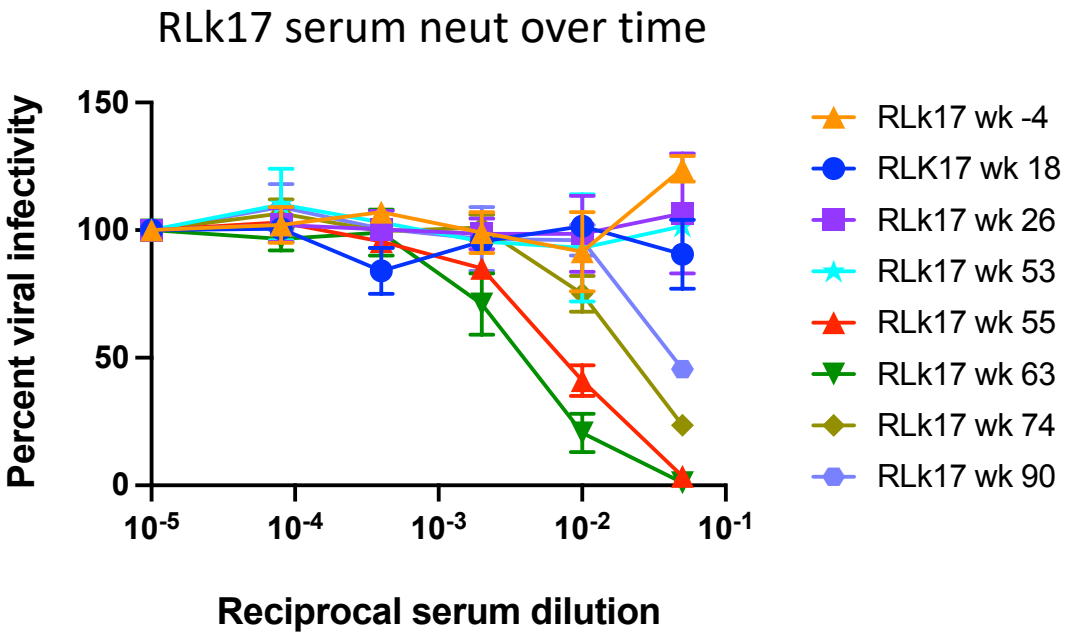
The neutralizing B cell lineages in RLk17 diverged significantly over time from their most recent common ancestor, gaining potency, but some non-neutralizing lineages also diverge at similar levels

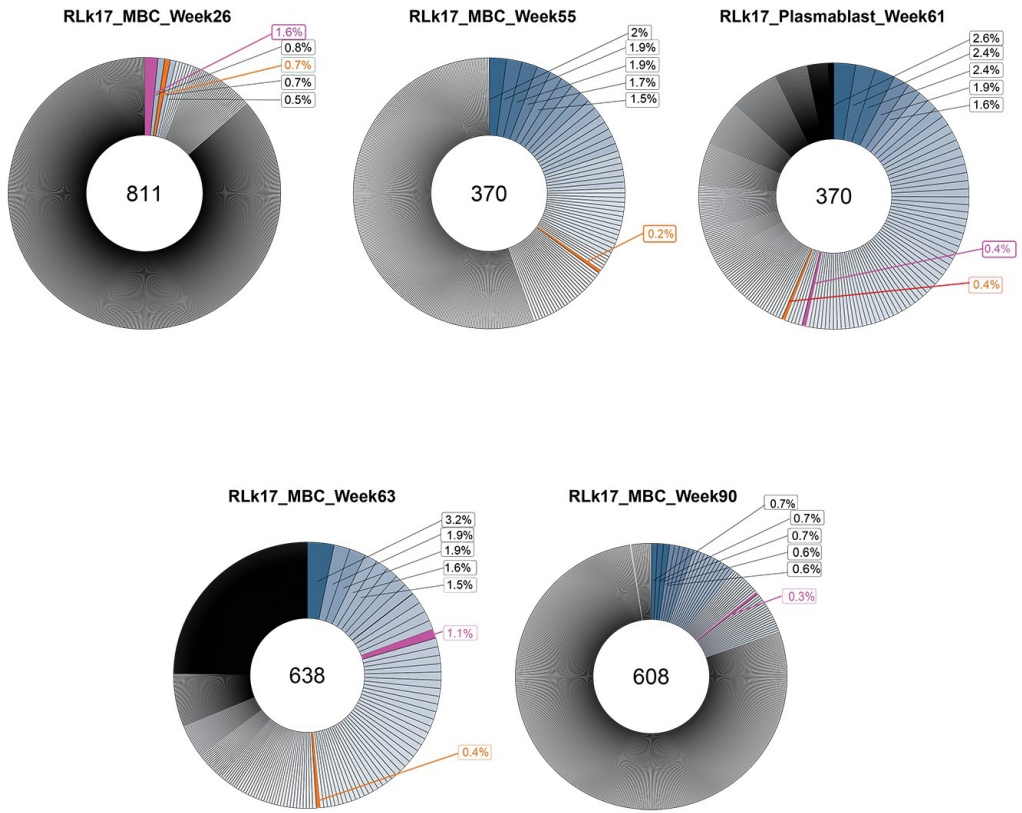
Clone Number	mAb tested	Time point	IC50 (ug/ml)	%V gene SHM
496	2D6	Week63	25	7.17%
865	2B8	Week63	25	5.02%
	2G5	Week63	25	4.68%
2549	2D1	Week63	25	5.41%
2109	V5-1A10	Week63	25	6.06%
2866	2A10	Week63	25	8.97%
2778	22UW4-6	Week26	25	1.01%
	22UW4-7	Week26	25	3.02%
	22UW4-8	Week55	0.44	6.04%
	22UW4-9	Week63	0.05	4.03%
	22UW4-10	Week63	0.07	7.72%
1719	22UW4-1	Week26	1.26	0.00%
	22UW4-2	Week26	0.27	3.69%
	22UW4-3	Week26	0.09	2.69%
	V5-1B1	Week63	0.23	7.05%
	V5-1B3	Week63	0.25	5.71%
	V5-1C4	Week63	0.16	5.71%
	V5-1C10	Week63	0.10	4.70%
	V5-1D7	Week63	0.71	7.38%
	V5-1E9	Week63	0.11	5.71%
	V5-2A1	Week63	0.12	7.38%
	V5-2D10	Week63	0.19	7.05%
	V5-2D11	Week63	0.17	5.03%
	22UW4-4	Week90	0.07	9.06%
	22UW4-5	Week90	0.05	10.40%



R Mopuri et al., *PLoS Pathogens* In Press

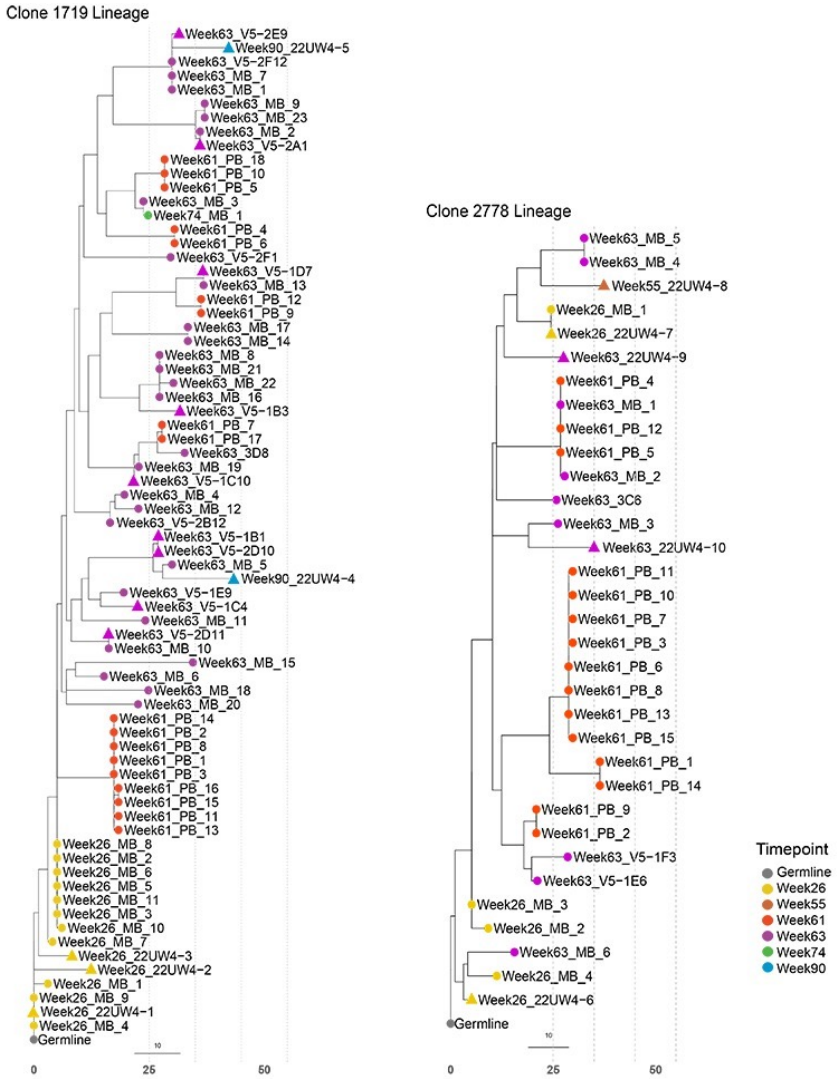
The neutralizing lineages in RLk17 were robust in their expansion and persistence within memory B cells and plasmablasts and they were highly abundant before serum neutralizing activity was detected (week 26)





R Mopuri et al., *PLoS Pathogens* In Press

RLk17 neutralizing lineage 1719 continued to undergo SHM for 29 weeks after the final immunization while lineage 2778 was not detected at this time

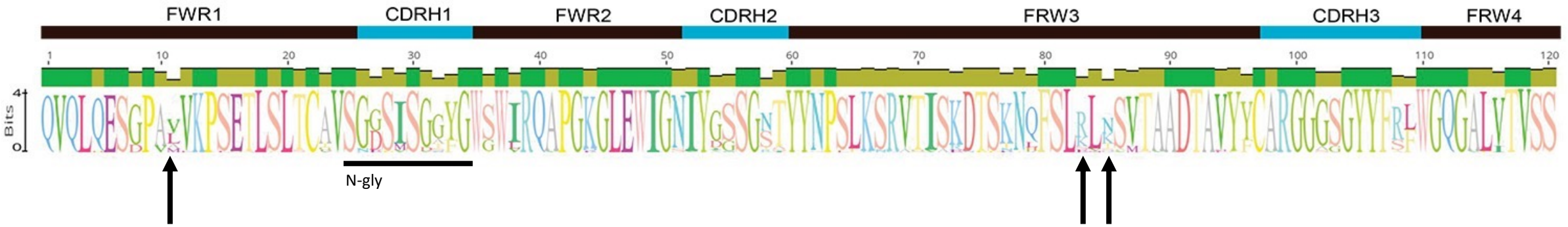


R Mopuri et al., *PLoS Pathogens* In Press

The RLk17 neutralizing lineages underwent extensive SHM/toggling over time in some framework regions as well as the CDR regions

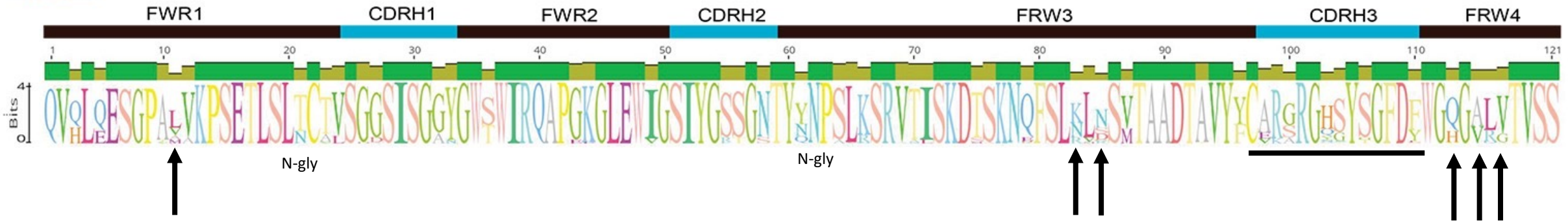
Clone 1719

Weeks 26 to 90



Weeks 26 to 63

Clone 2778



R Mopuri et al., *PLoS Pathogens* In Press

Using individualized germline databases for all RMs, RLk17 was the only Z1800M immunized RM that harbored the neutralizing VH germline allele

RM ID	Vaccine / Group	Neut germline	Closest germline to IGHV4-NL_21*01_S4478	% identity
RGi17	R66M gp120		IGHV4-NL_22*01_S2393	96.56
RLf17	R66M gp120	IGHV4-NL_21*01_S4478		100
RLl17	R66M gp120		IGHV4-NL_21*01_S0138	95.88
RTe17	R66M gp120		IGHV4-NL_22*01_S2393	96.56
RVh17	R66M gp120	IGHV4-NL_21*01_S4478		100
RPh17	R66M trimer		IGHV4-NL_22*01_S2393	96.56
RTm17	R66M trimer		IGHV4-NL_21*01_S0138	95.88
RVe17	R66M trimer	IGHV4-NL_21*01_S4478		100
RVn17	R66M trimer		IGHV4-NL_21*01_S0138	95.88
RWb17	R66M trimer		IGHV4-NL_20*01_S1055	96.23
ROa17	Z1800M gp120		IGHV4-NL_21*01_S0138	95.88
RPz16	Z1800M gp120		IGHV4-NL_23*01_S1494	96.22
RRb17	Z1800M gp120		IGHV4-NL_21*01_S0138	95.88
RLk17	Z1800M gp120	IGHV4-NL_21*01_S4478		100
15C172	Z1800M trimer		IGHV4-NL_21*01_S0138	95.88
MA277	Z1800M trimer		IGHV4-NL_21*01_S0138	95.88
RFj17	Z1800M trimer		IGHV4-NL_21*01_S0138	95.88
RSb17	Z1800M trimer		IGHV4-NL_21*01_S0138	95.88
RYf17	Z1800M trimer		IGHV4-NL_21*01_S5333	99.66

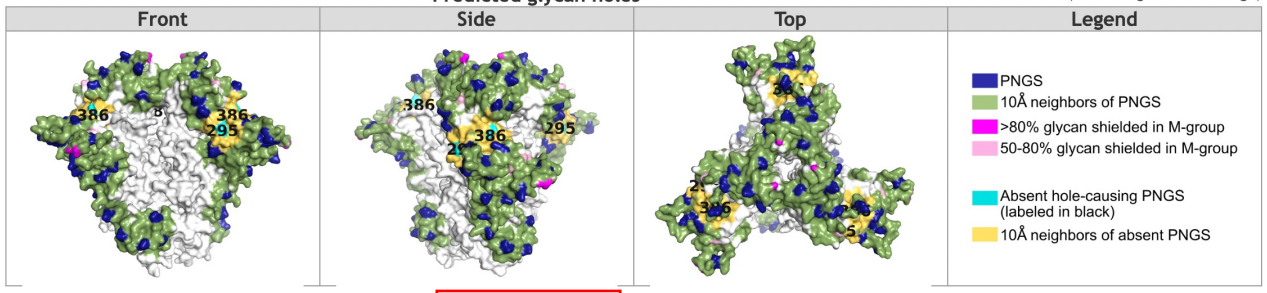
R Mopuri et al., *PLoS Pathogens* In Press

Conclusions from the Z1800M project to date

- Z1800M T/F Env has a uniquely exposed V5/CD4bs region that makes it a candidate for further development and strategic manipulation
- Neutralizing B cell precursors in RLk17 were activated by MVA immunization; early engagement is important to not be competed out by the many other non-neutralizing specificities that are also elicited
- Neutralizing B cell precursors in RLk17 could neutralize at various levels prior to SHM
- Certain germlines may be more amenable to developing neutralization than others; also dependent on the Env immunogen
- SHM in the framework regions appear to be important for affinity maturation and gaining neutralization potency
- HIV-1 T/F Env proteins do contain ‘blueprints’ for neutralizing antibody development, likely based on glycan coverage, which follows a similar path in humans and rhesus macaques

Where is this project going from here? Use evolution to inform Z1800M Env immunogen design to try to elicit some heterologous neutralization breadth by targeting CD4bs

Predicted glycan holes (click images to view large)



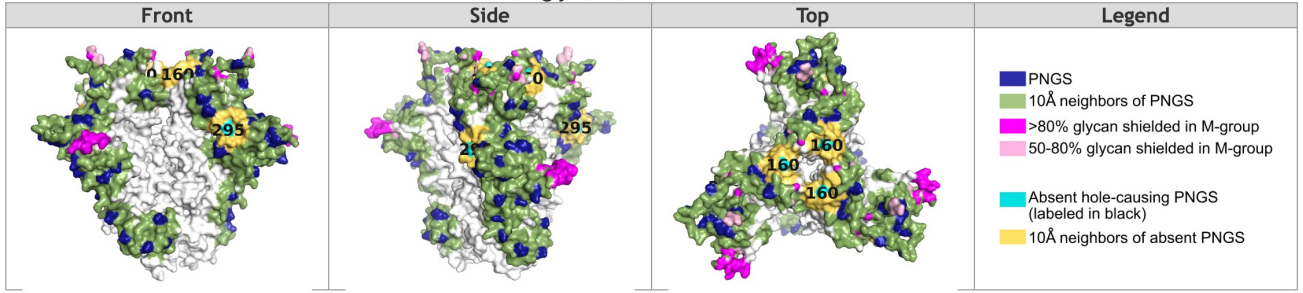
Glycan hole positions: 258 259 286 326 328 359 364 369 372 373 374 375 376 378 383 384 386 387 417 418 419 445 457 467 470

Absent PNGS	Glycan hole area due to absent PNGS	Glycan hole area due to cumulative addition of PNGS	Glycan hole positions covered by absent PNGS
N386	744.27	744.27	258 259 328 364 369 372 373 374 375 383 384 386 387 417 418 419 470
N295	157.9	157.9	376 378 445

The T/F Env has a uniquely exposed V5/CD4bs region that makes it an attractive candidate for further development and strategic manipulation

This exposed region is targeted by autologous neutralizing antibodies

Predicted glycan holes (click images to view large)



Glycan hole positions: 124 125 127 153 159 160 161 162 166 167 168 169 170 171 172 286 307 308 326 376 378 445 459 466

Absent PNGS	Glycan hole area due to absent PNGS	Glycan hole area due to cumulative addition of PNGS	Glycan hole positions covered by absent PNGS
N160	1356.9	1356.9	124 125 127 159 160 161 162 166 167 168 169 170 171 172 307 308
N295	157.9	157.9	376 378 445

42 months later (~6-12 months after high plasma breadth developed) CD4bs coverage has changed

Interestingly, these Env variants also lost a glycan critical to V2 apex targeted bnAbs, which could also have been involved in breadth

Longitudinal Env sequence analysis by Elena Giorgi, FHCC
https://www.hiv.lanl.gov/content/sequence/GLYSHIELDMAP/glyshieldmap.html

Determine if/how the V5 domain regulates CD4bs exposure in Z1800M Envs

	V00	V02	V06	V12	V18	V24	V30	V36	V39	V42	
GITE----------ESNNT-EIFRPG	0.994	0.996	0.001	0	0	0	0	0	0	0	
GTTG----------ESNNT-EIFRPG	0	0	0.245	0	0	0	0	0	0	0	
GIT----------ESNNT-EIFRPG	0	0	0.113	0	0	0	0	0	0	0	Single PNG at 466
GDTG----------ESNNTEEIFRPG	0	0	0	0.326	0	0	0	0	0	0	
GDTA----------ESNNTEEIFRPG	0	0	0	0.283	0	0	0	0	0	0	
G--------------TNNT-EIFRPG	0	0	0	0	0.812	0.011	0	0	0	0	
GKSNNT--------ESNNT-EIFRPG	0	0	0	0	0.034	0.314	0.157	0.001	0	0	
GTTESNNT------ESNNT-EIFRPG	0	0	0	0	0.039	0.459	0	0	0	0	
GTTESNNT------ESNNT-ETFRPG	0	0	0	0	0	0.039	0.484	0	0	0	PNG Gain at 464
GKSNNT--------ESNNT-ETFRPG	0	0	0	0	0	0	0.115	0.002	0	0	
GTPESNNT------ESNNT-ETFRPG	0	0	0	0	0	0	0.037	0.792	0.143	0	
GTPESNNP------ESNNT-ETFRPG	0	0	0	0	0	0	0	0.041	0.545	0.140	Loss of 1 PNG
GTP----------ESNNP-ETFRPG	0	0	0	0	0	0	0	0	0.007	0.427	Loss of both PNGs
GTP----------ESN-T-ETFRPG	0	0	0	0	0	0	0	0	0	0.104	
Mean Number of PNGs	1.00	1.00	0.98	1.05	1.08	1.99	1.98	1.87	1.22	0.53	
Mean V5 Length	14.00	14.00	13.96	14.59	10.81	17.18	17.30	17.63	17.90	14.98	

→ V5 influences VRC01 and autologous mAb neutralization

→ V5 to be tested

Transitory increase in length and PNGs observed prior to breadth could be driven by escape from V5/CD4bs neutralizing antibodies

Longitudinal sequence analysis by Elena Giorgi, FHCC

Other Research Projects and Directions

- Developing and testing Z1800M based immunogens using viral evolution, trimer stabilization, computational modeling (Kelly Lee, Gnana Gnanakaran, Noah Sather, Elena Giorgi)
- Tracking and structurally characterizing neutralizing antibodies and their epitopes elicited in BG505 SOSIP immunized rhesus macaques (Marzena Pazgier and Steve Bosinger)
- Structure, function, and modeling of HIV-1 Env-Matrix interactions in the viral particle (Kelly Lee, Gnana Gnanakaran, Michael Zwick, Gaurav Bhardwaj)
- Development and testing of novel tenofovir-based analogues modified for improved bioavailability and potency; evaluating viral diversity and resistance (Eric Miller, Nicole Pribut)
- Developing novel nucleic acid-based HIV vaccines (Amit Khandar, Noah Sather, Deb Fuller, Elena Giorgi)

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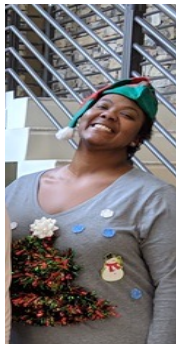

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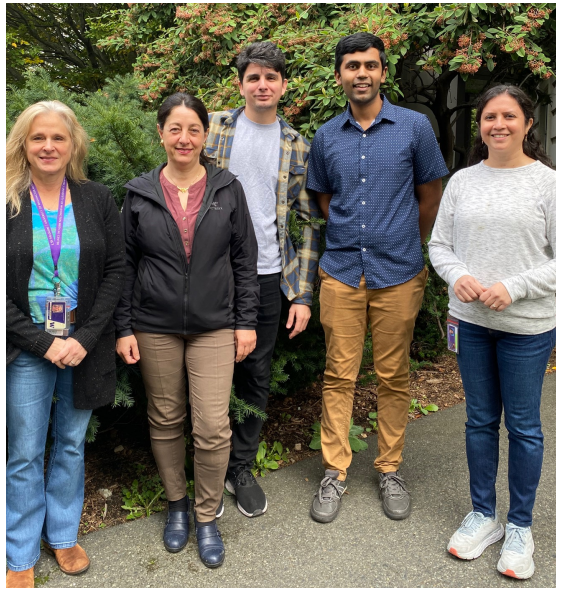

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