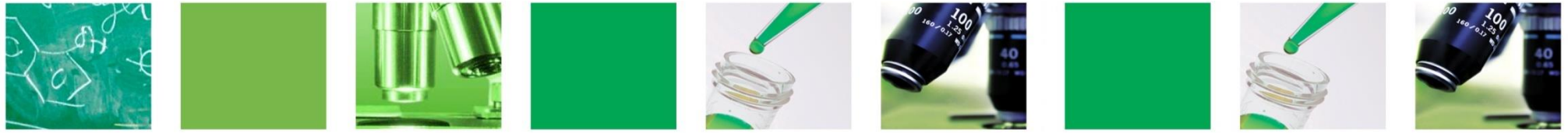


Importance of structural biology in virology research: HIV, SARS-CoV-2 and beyond

Thandeka Moyo-Gwete
Senior Medical Scientist
National Institute for Communicable Diseases
Johannesburg, South Africa

Who are we??

Medical Research Council Antibody Immunity Research Unit

Based in Johannesburg, South Africa

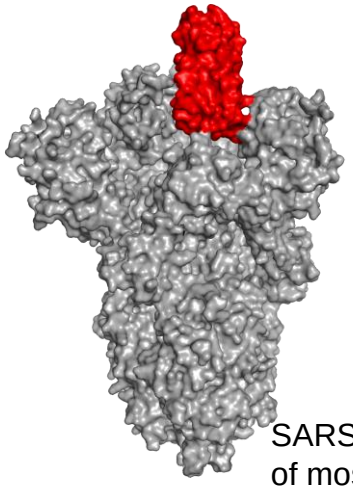
Virology, Immunology and Structure

HIV, Influenza, CMV, SARS-CoV-2

Contribution of Light Sources to Biological and Medical Sciences

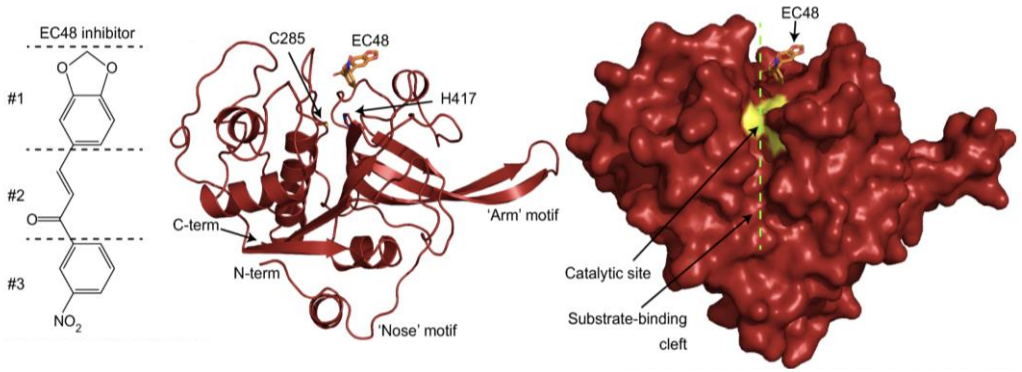
Structural biology helps us understand the **structure and function of macromolecules** including proteins, DNA and RNA

Aids in vaccine design



SARS-CoV-2 spike protein – basis of most vaccine candidates

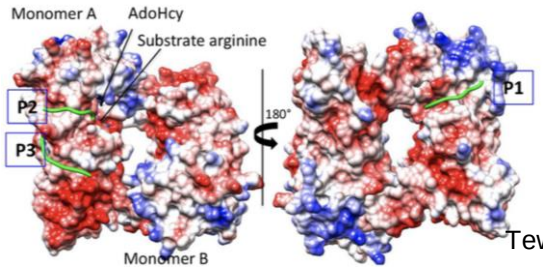
Provides information on protein-inhibitor interactions for drug, herbicide and pesticide design



Malaria protein bound by inhibitor

Machin et al 2019, *Malaria Journal*

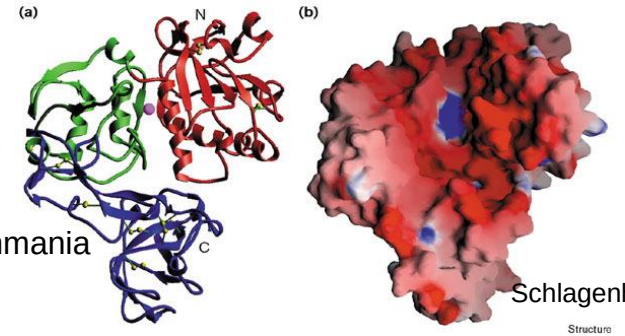
Provides insight into the mechanism of enzymes and is an enabler for industrial enzymology



Active site binding of protein arginine methyltransferases

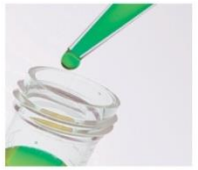
Tewaryy et al., 2019, *Cell Mol Life Sci.*

Reveal the structure and therefore vulnerable regions of proteins from pathogens



Structure of the main leishmania surface antigen

Schlagenhauf et al, 1998, *Structure*


Importance of structural biology in virology research: HIV, SARS-CoV-2 and beyond

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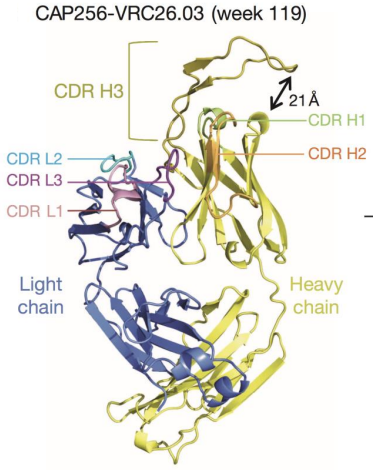
UNIVERSITY OF THE WITWATERSRAND JOHANNESBURG

Neutralizing antibodies in HIV vaccine development

- **Broadly neutralizing antibodies (bNAbs)** are required for an effective HIV vaccine

- They neutralize various global HIV-1 strains and inhibit entry into cells

- However, HIV infection has shown us:
  - Rare – 20% HIV infected individuals develop them
  - Unusual features
  - Take long to appear – chronic infection

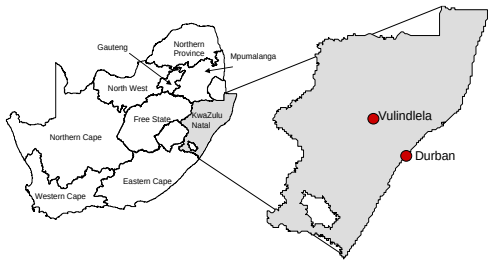


- Therefore, studying bNAbs and their targets may aid in immunogen design to elicit bNAbs

Importance of structural biology in bNAb research

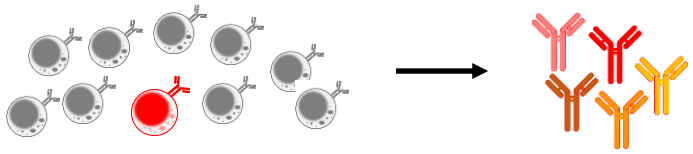
Why we study the structure of antibody-HIV Envelope complexes?

-define novel epitopes targeted by bNAbs
-discover key residues important for the neutralization of HIV by bNAbs
-define structural attributes of “special” viral strains
-inform design of immunogens which will elicit bNAbs

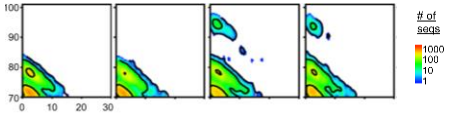


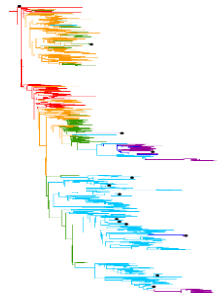
bNAb donors

i) Isolation of mAbs from memory B cells



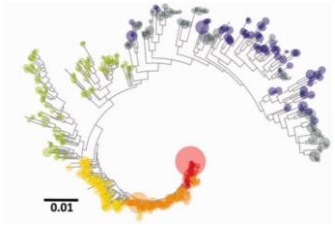
ii) Next generation sequencing of B cell lineages

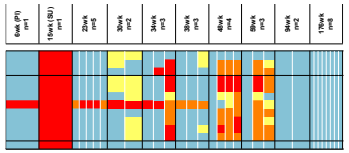




Virus antibody co-evolution pathways defined through structural analysis

iii) Viral sequencing



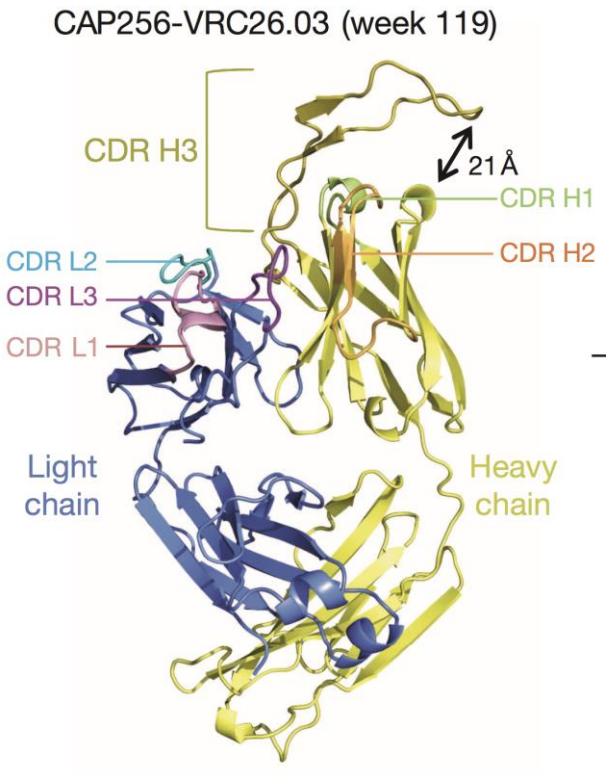


CAPRISA
CENTRE FOR THE AIDS PROGRAMME OF RESEARCH IN SOUTH AFRICA

UNAIDS
CAPRISA IS A UNAIDS COLLABORATING CENTRE FOR HIV PREVENTION RESEARCH

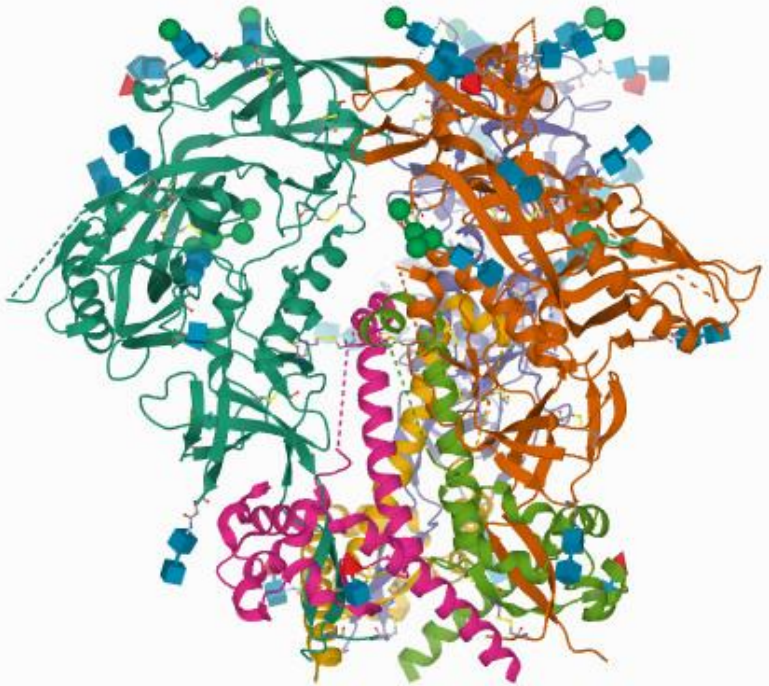
Adapted from Moore, CHIVR, 2018

Technique to obtain the high resolution structure of proteins:



Antibody structure

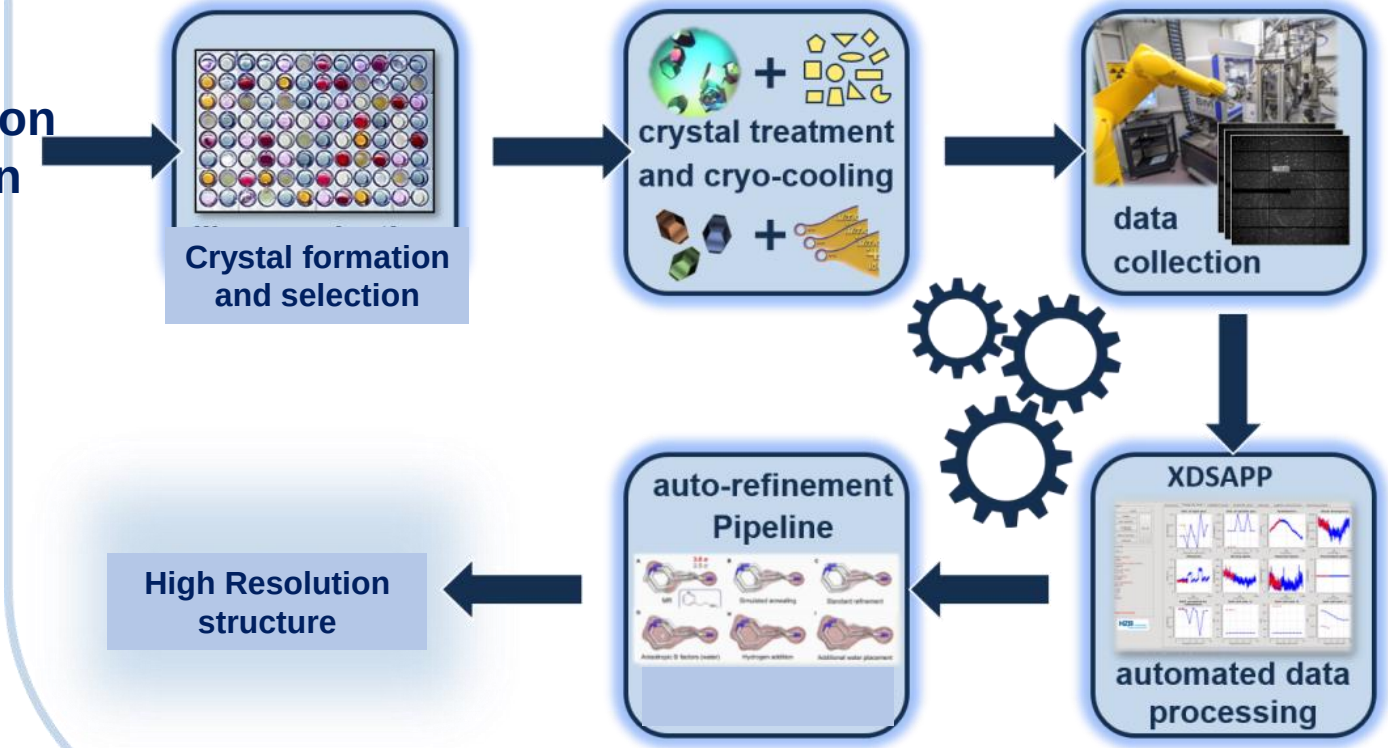
BG505



HIV Envelope structure

X-ray crystallography pipeline

Protein production and purification (HIV Env + Ab complex)



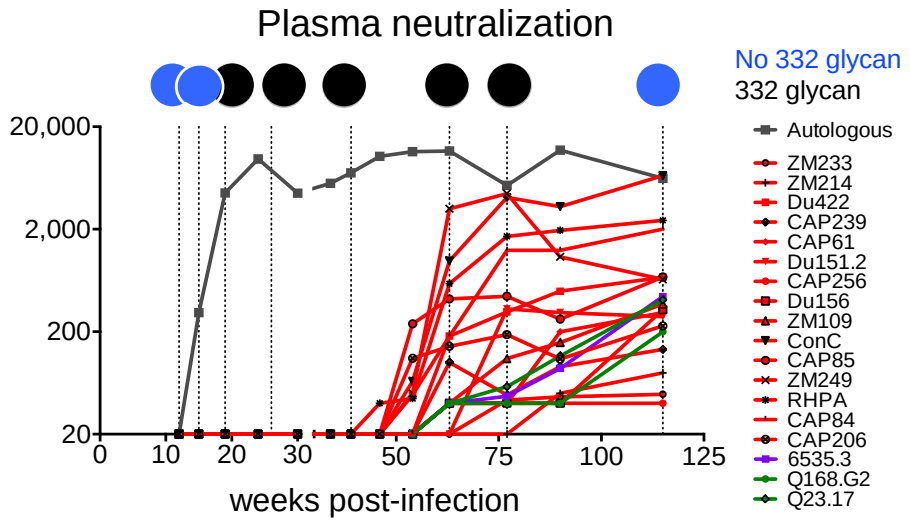
SYNCHROTRON TECHNIQUES FOR AFRICAN RESEARCH AND TECHNOLOGY
START
diamond

Adapted from: https://www.helmholtz-berlin.de/forschung/oe/np/gmx/fragment-screening/index_en.html

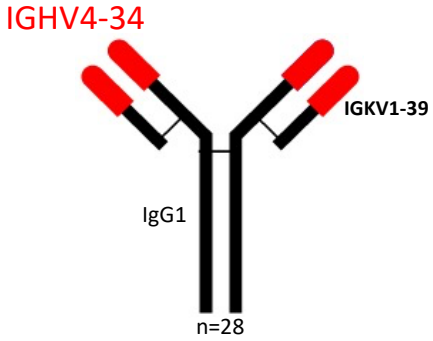
Structural characterization of antibody lineages from single donor

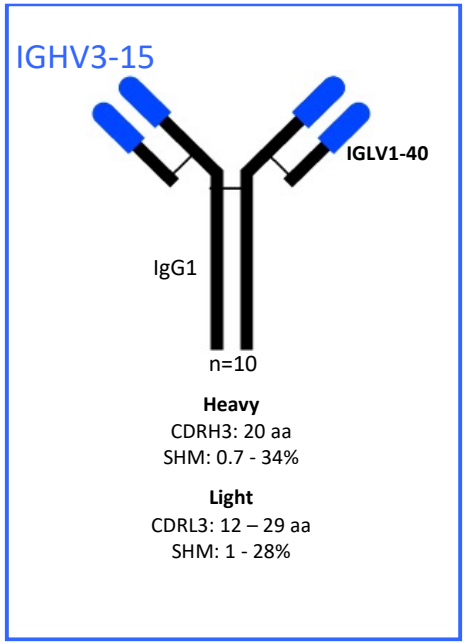
CAP314 – HIV-infected donor who developed bNAbs within 2 years post-infection

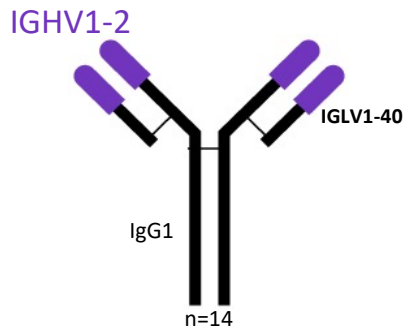
Isolated and characterized three antibody lineages (families)

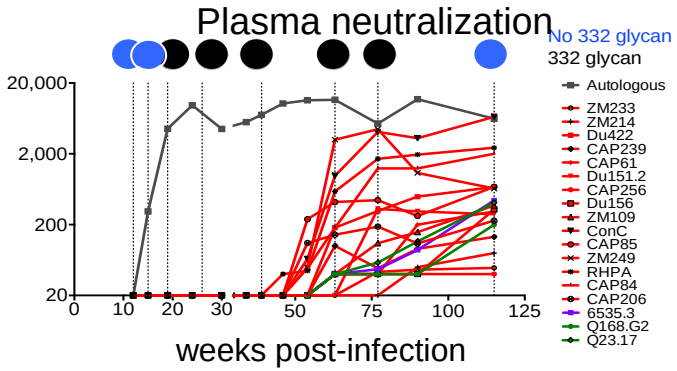


Moore et al., Nature Medicine, 2012

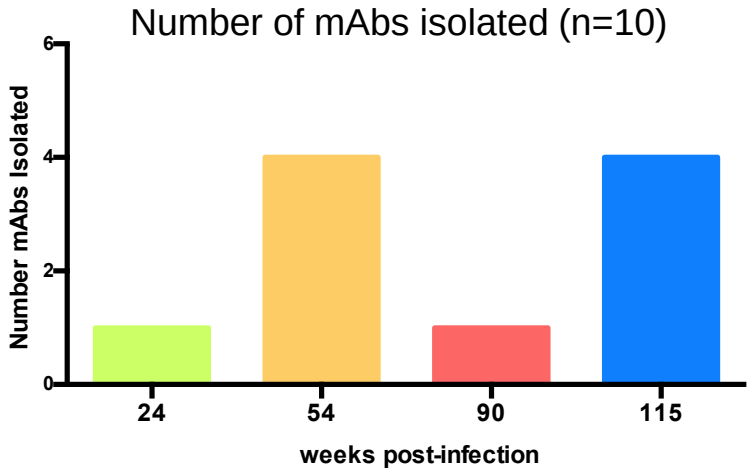


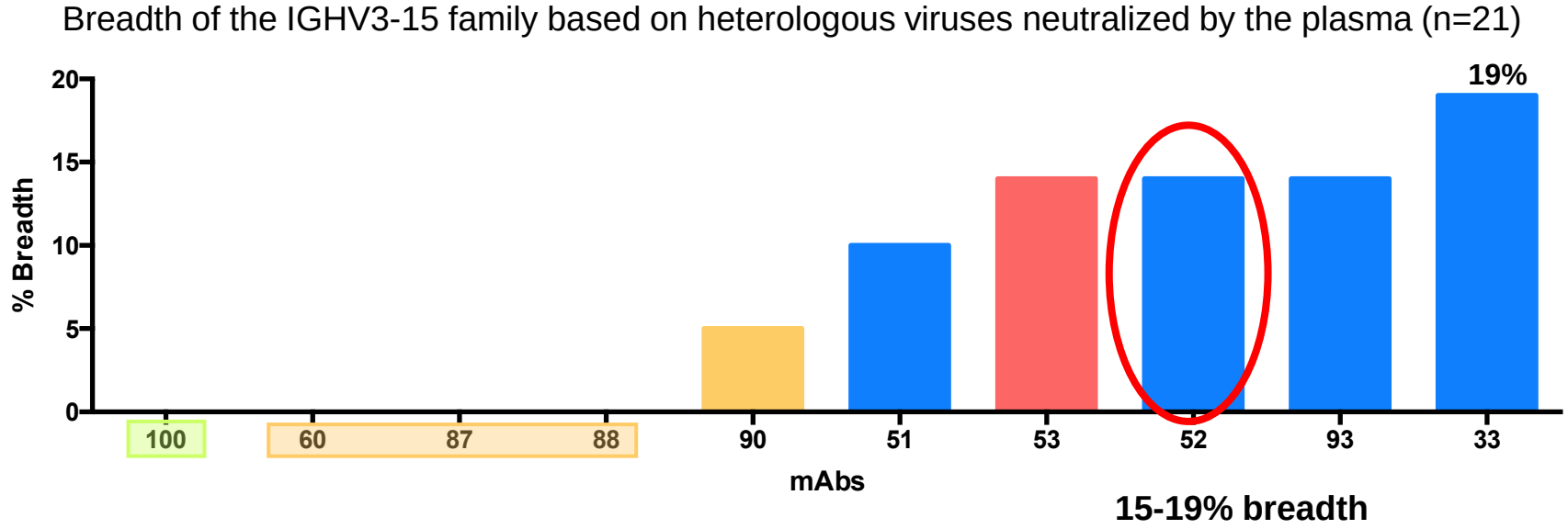




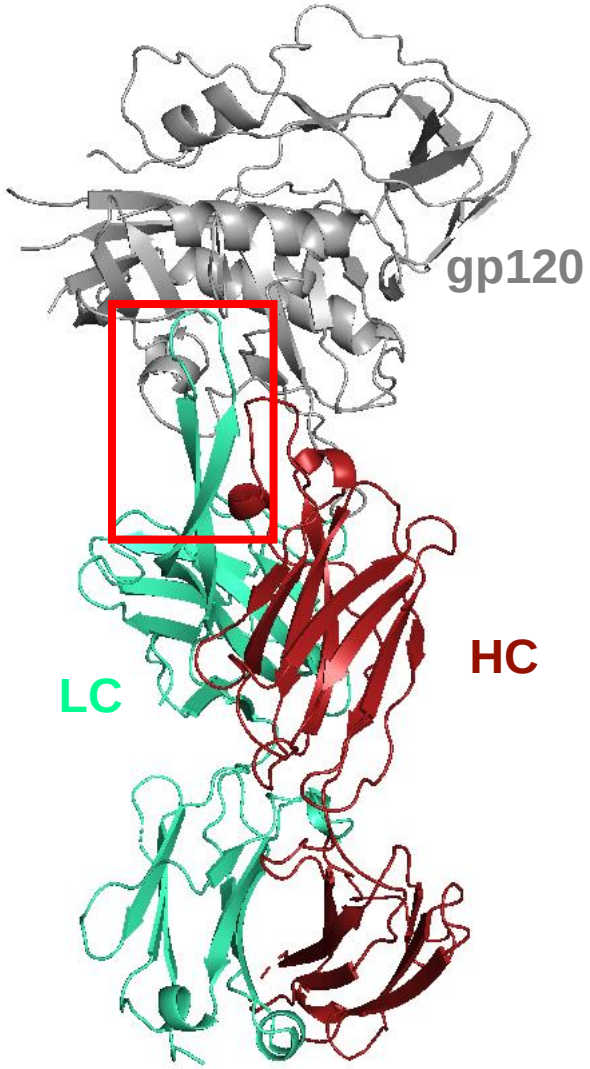


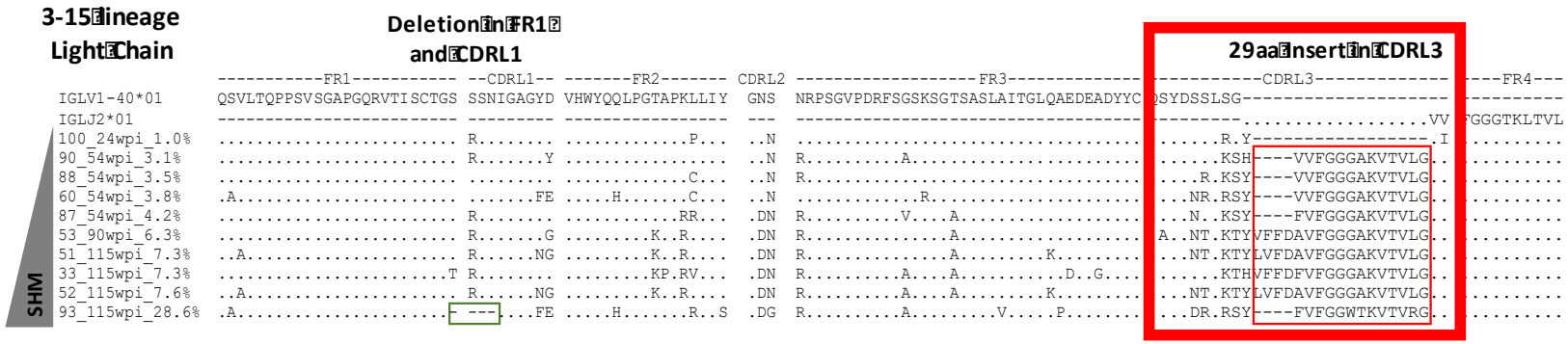
Moore et al., Nature Medicine, 2012

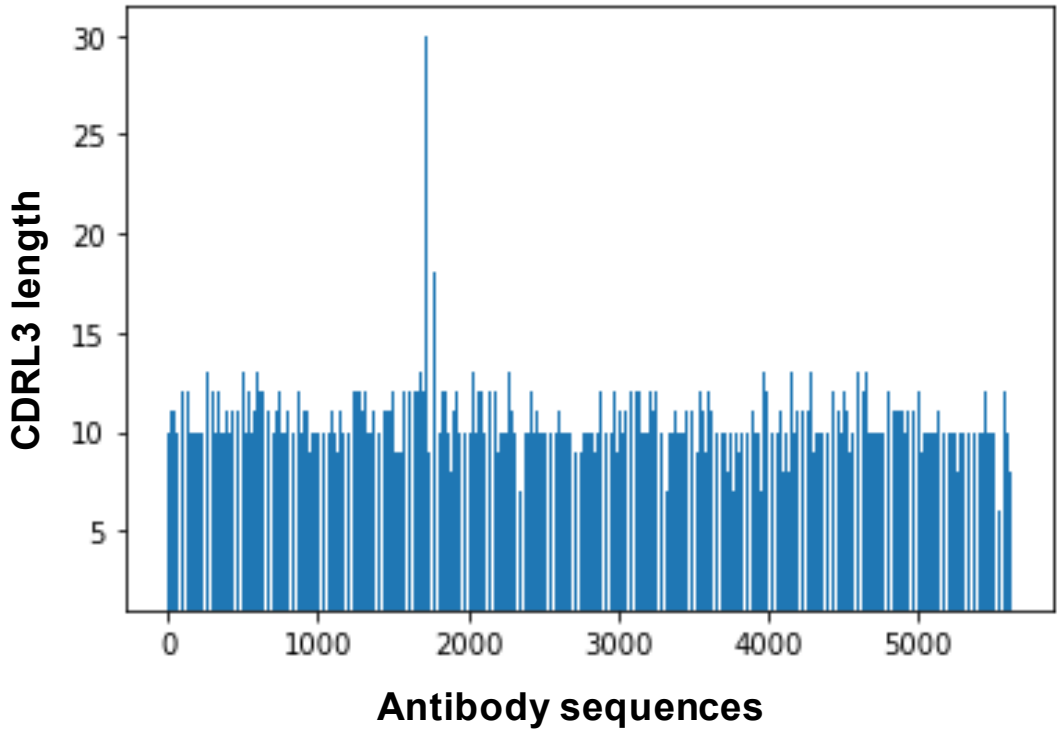




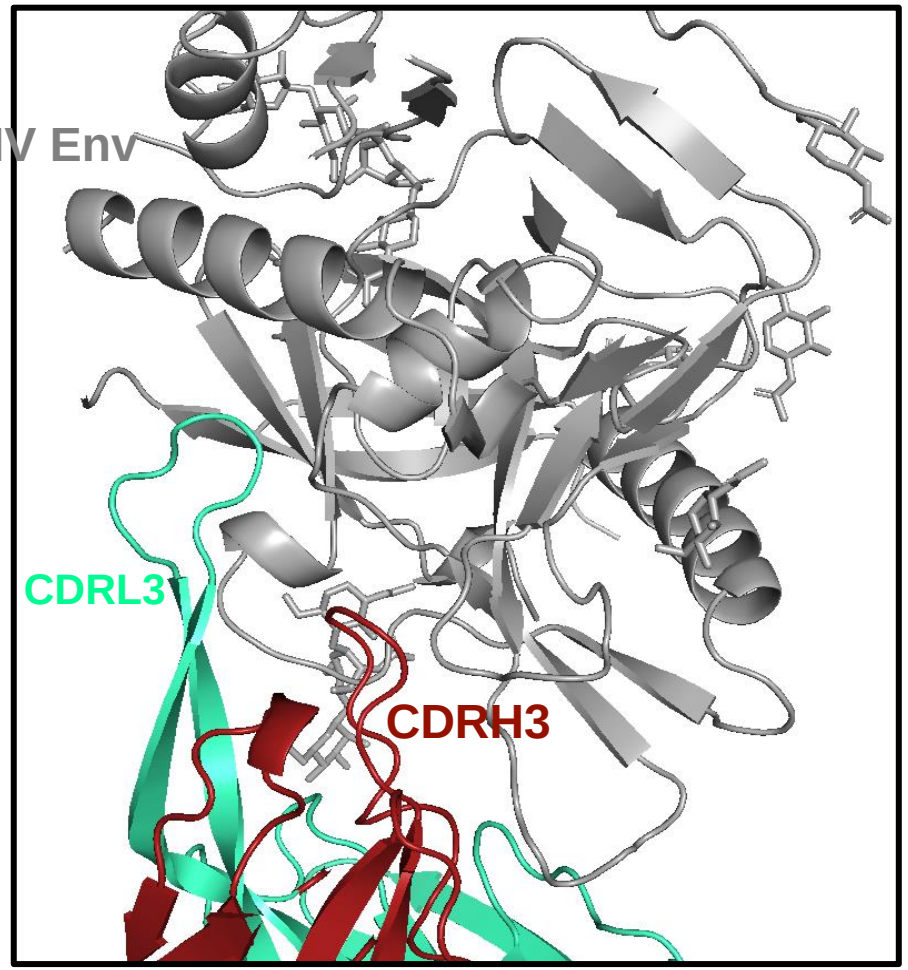
CDRL3 insertion (29 aa) associated with increased breadth over time



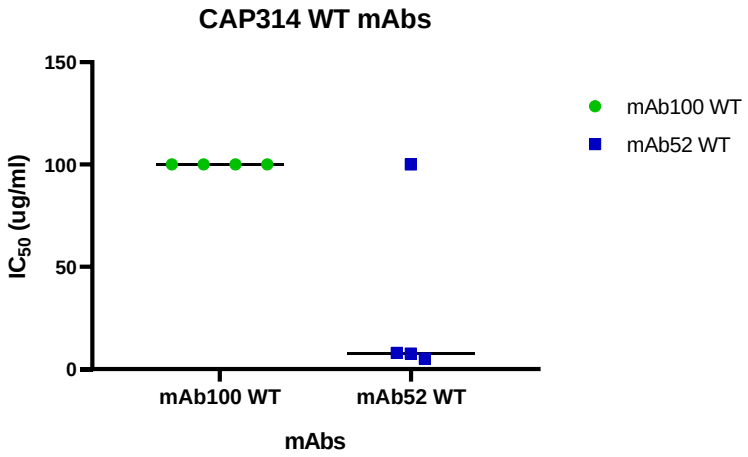


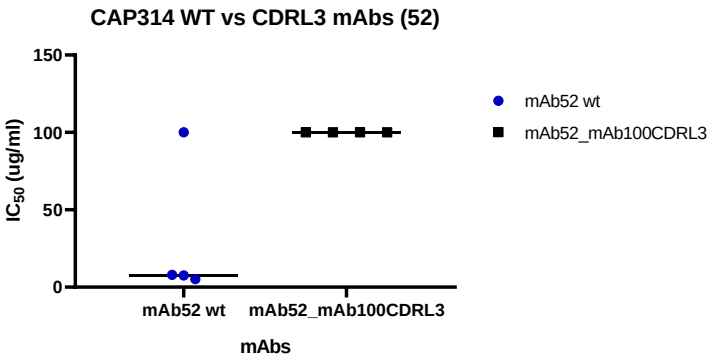


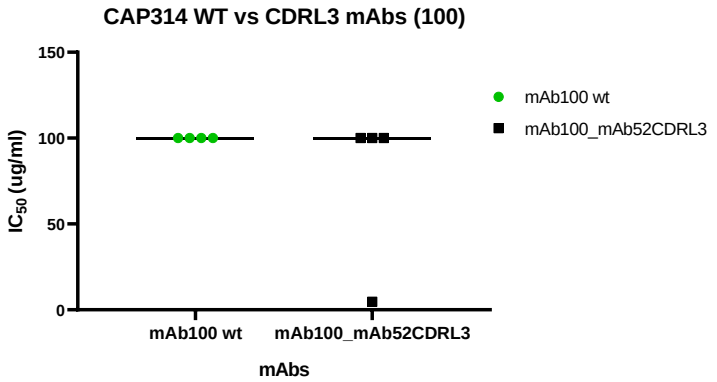
Novel mode of binding to HIV CD4 binding site



Binds to common HIV bNAb epitope (CD4 binding site) in a unique way







Using structural biology our lab can now:

i) Learn more about the structure and function of bNAbs – what unusual features do they have? how can we elicit them in a vaccine?

ii) explore vaccine elicited antibody responses – HIV trimer trials: study the structure of these Abs and how they interact with full HIV trimers and other immunogens

iii) Discover and characterize unique HIV strains – learn more about unique features of Env strains that have enhanced capacity to elicit bNAbs

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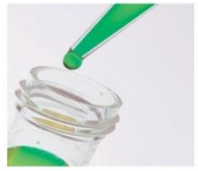

Importance of structural biology in virology research: HIV, SARS-CoV-2 and beyond

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The answer to the vaccine question: the spike protein

Corona = crown or circle of light

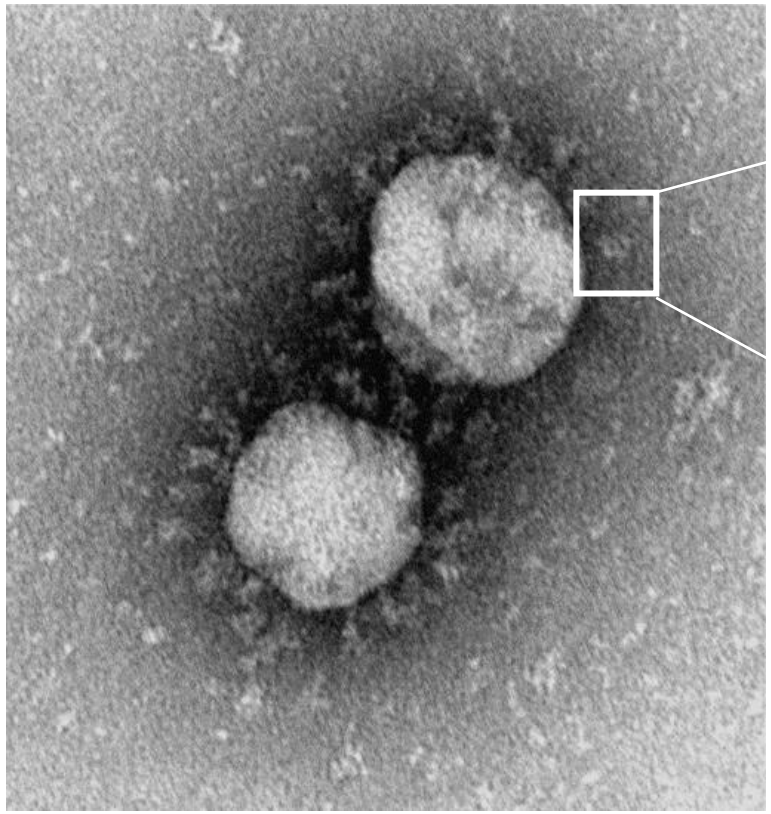
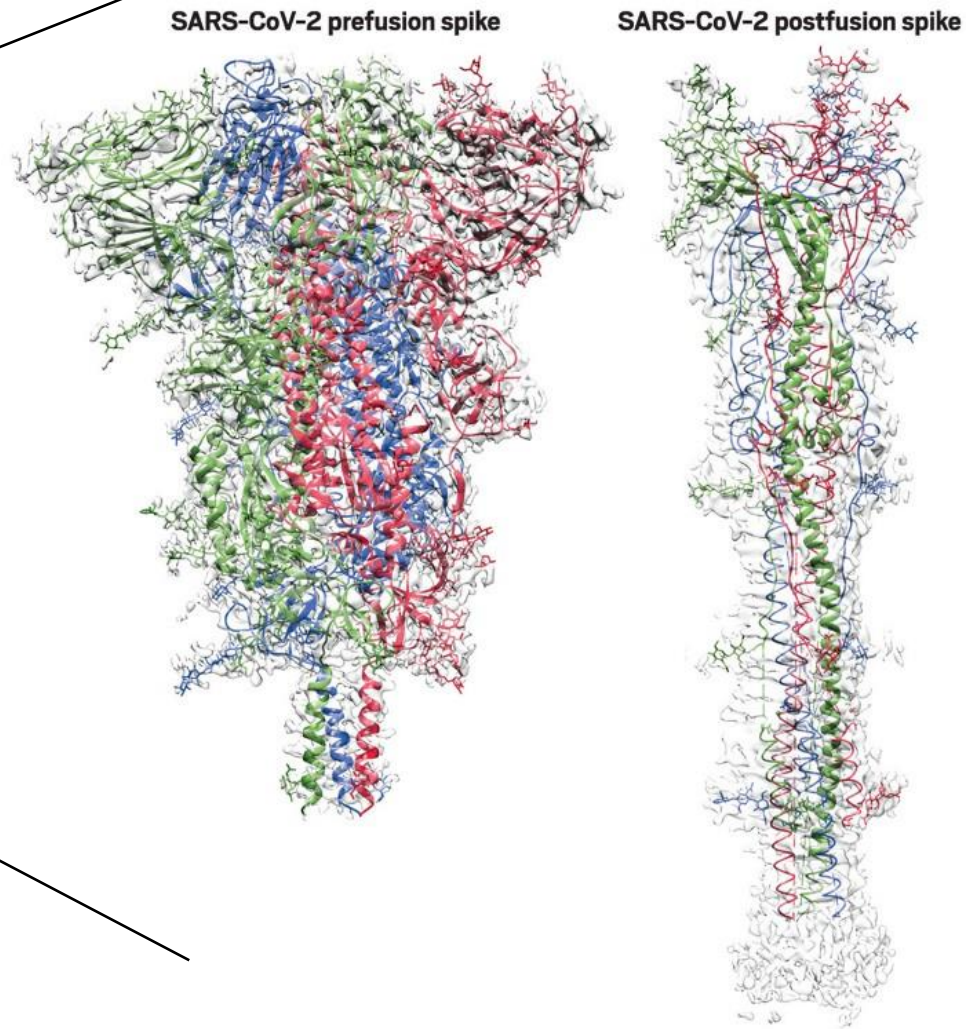
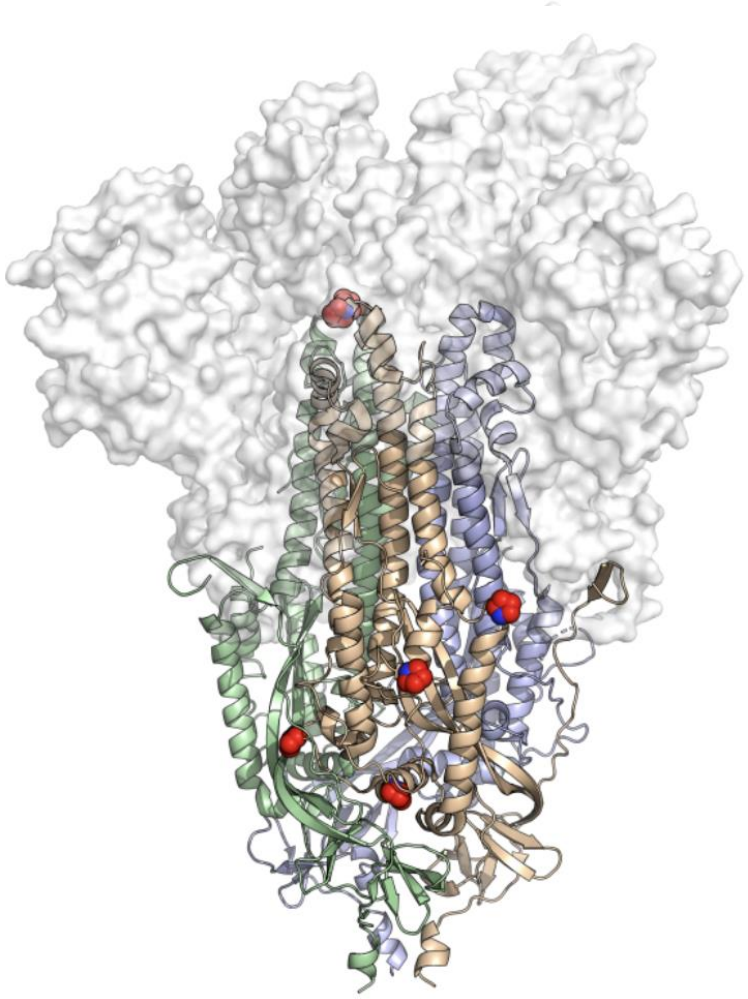


Image: Monica Birkhead
CEZPD, NICD



https://cen.acs.org/pharmaceuticals/vaccines/tiny-tweak-behind-COVID-19/98/i38

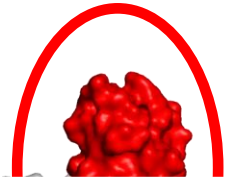
Using structural biology to make the best vaccine candidate



Jason McLellan, University of Texas

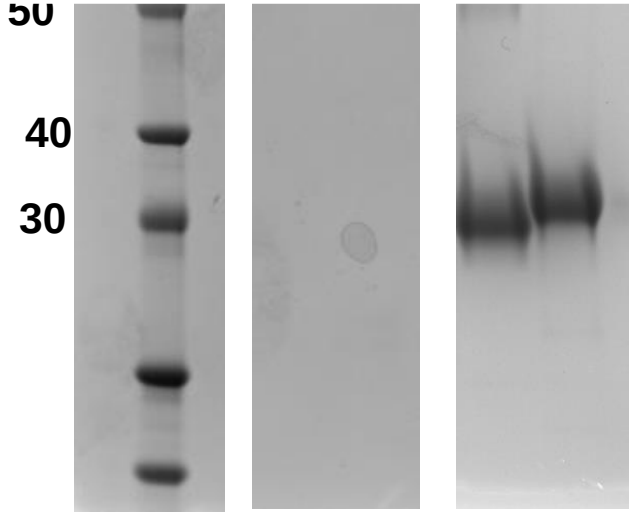
Moderna
Pfizer
JnJ
Novavax

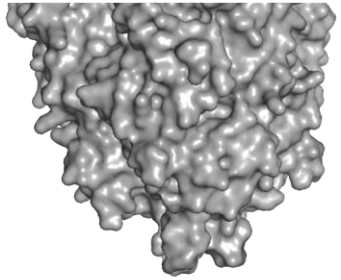
SARS-CoV-2 RBD and spike expression



Receptor binding domain (RBD)

We have used the spike protein to answer many important questions around the immune response triggered by SARS-CoV-2 infection and vaccination

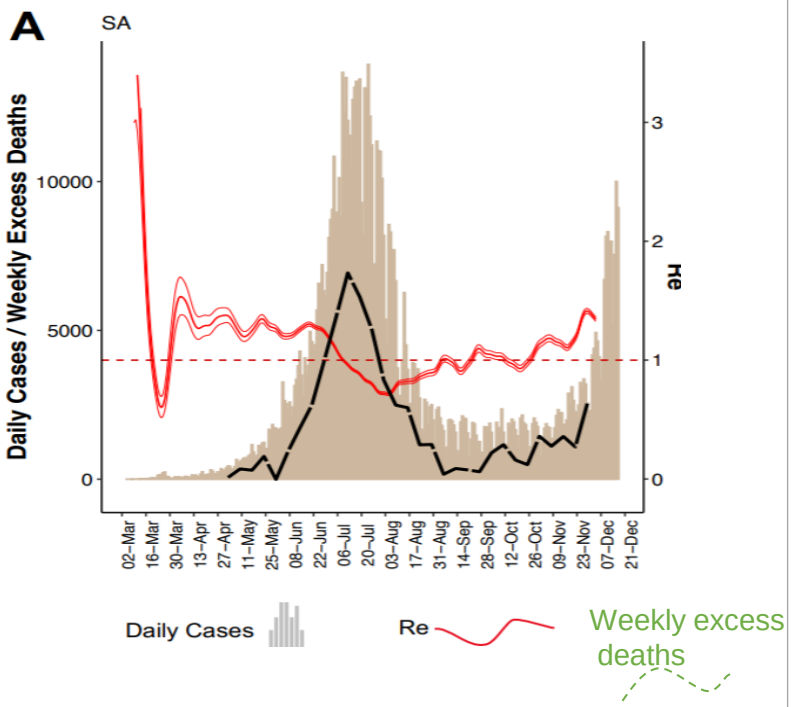




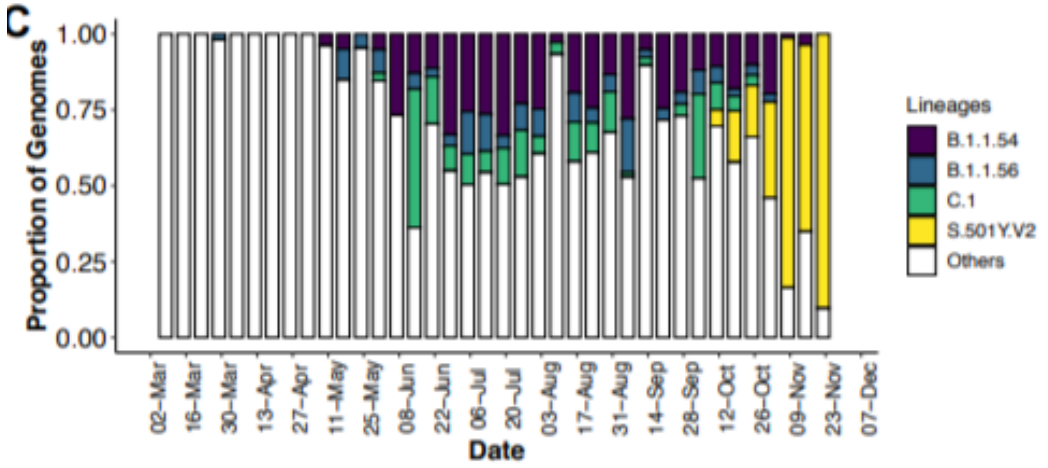
RBD and Spike constructs kindly provided by Florian Krammer and Jason McLellan

Emergence and spread of Beta variant in South Africa

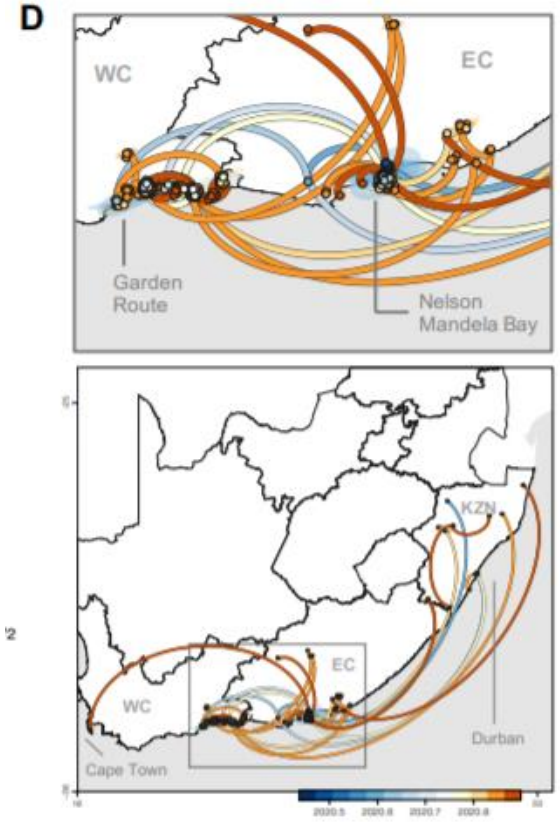
Early and rapid resurgence prompted intensified genomic surveillance in October…



.....by mid December Beta had replaced the D614G strain…..

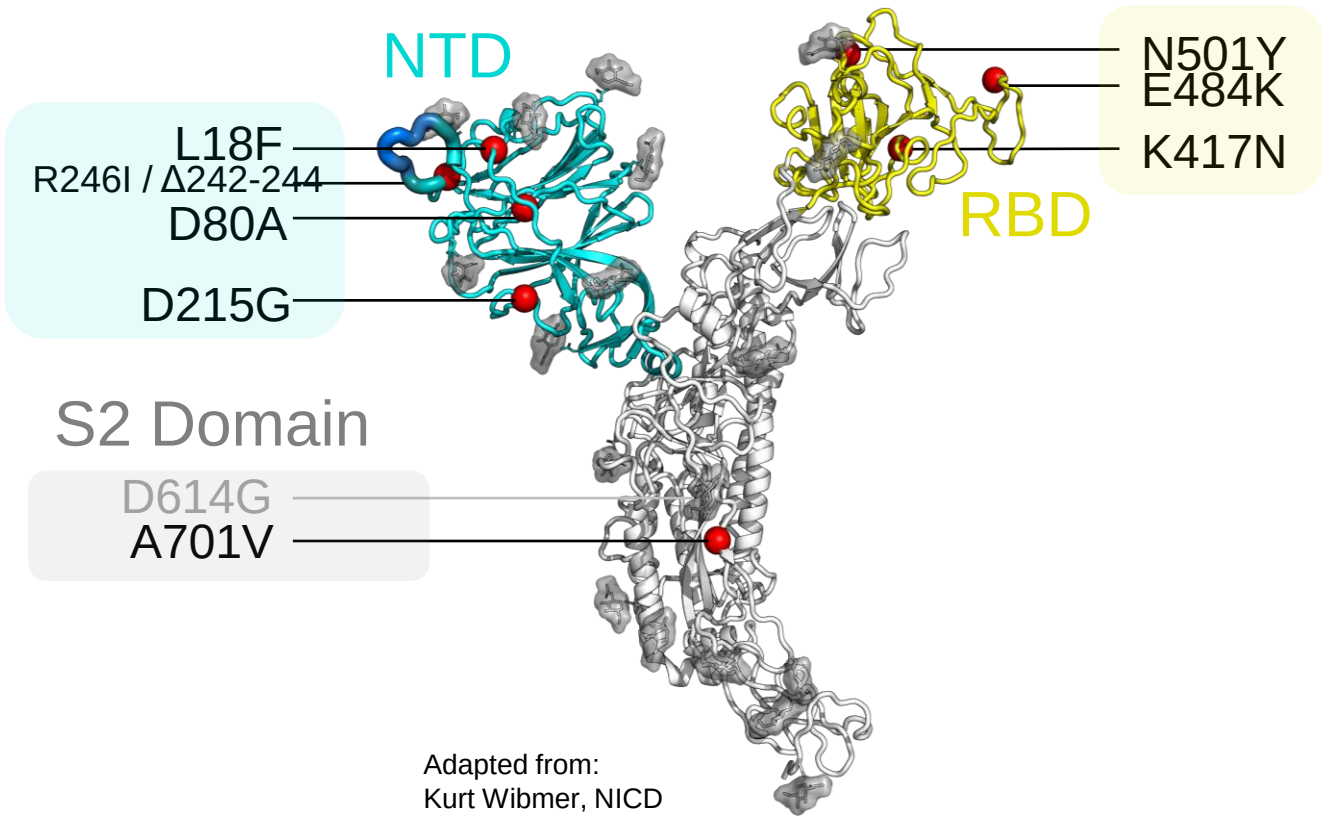


.....and spread from the Eastern Cape

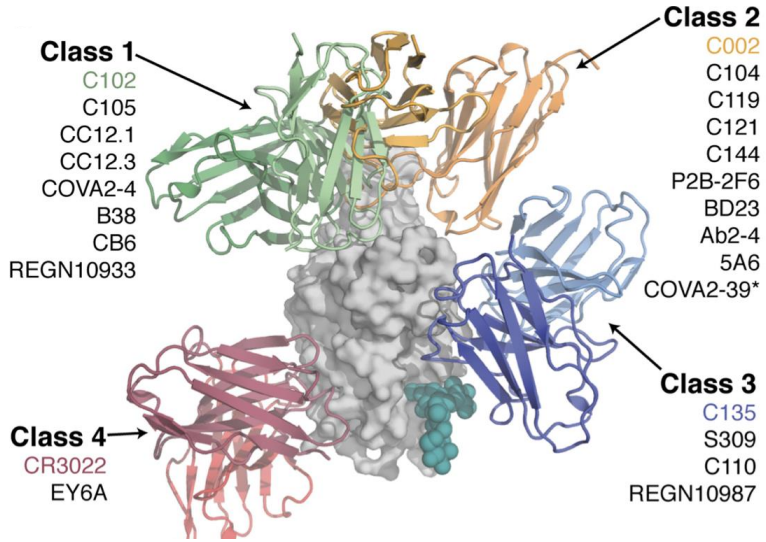


Tegally et al, Nature, 2021

Mutations Specific to the Beta variant

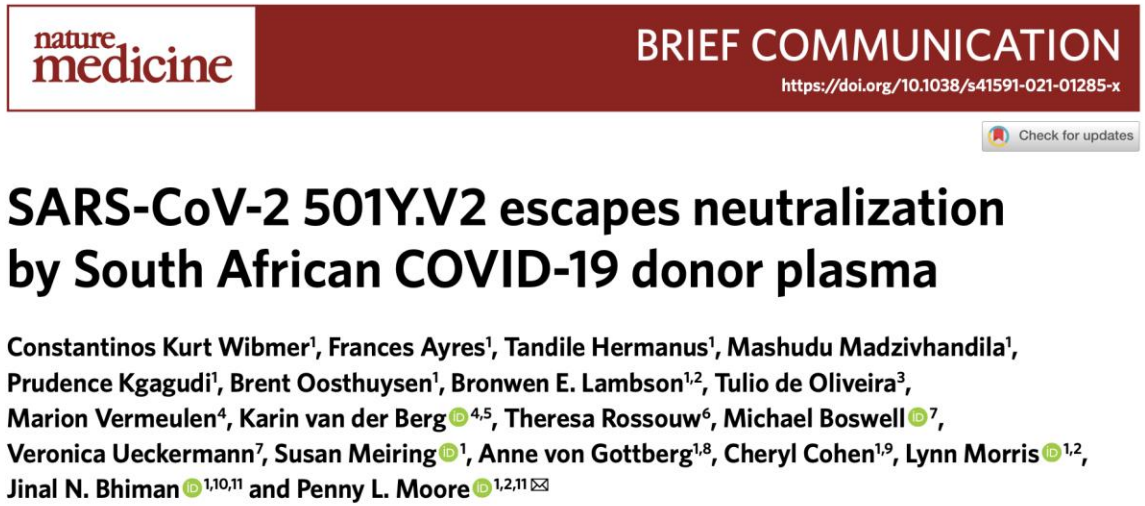


Adapted from:
Kurt Wibmer, NICD



Barnes 2020, Nature

Does the Beta variant escape neutralization by South African COVID-19 donor plasma?



nature medicine

BRIEF COMMUNICATION

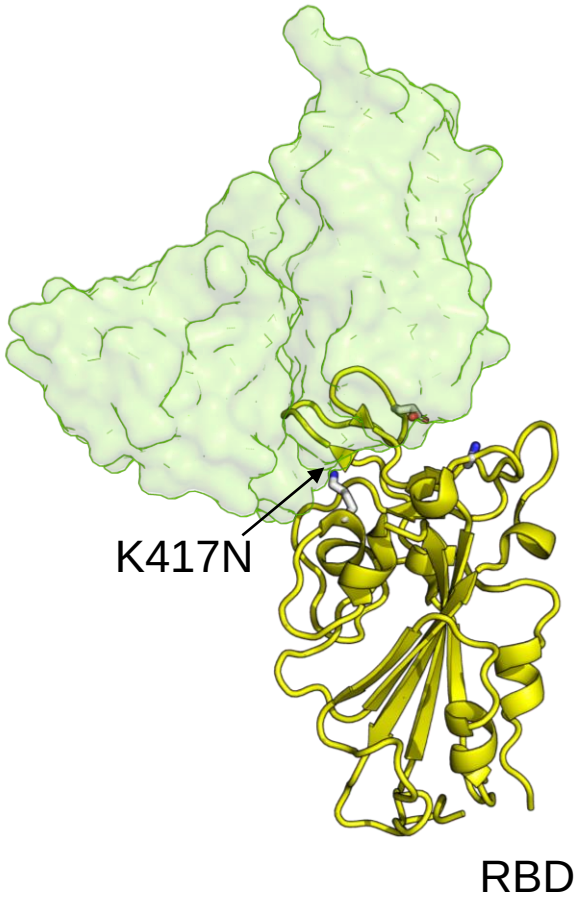
https://doi.org/10.1038/s41591-021-01285-x

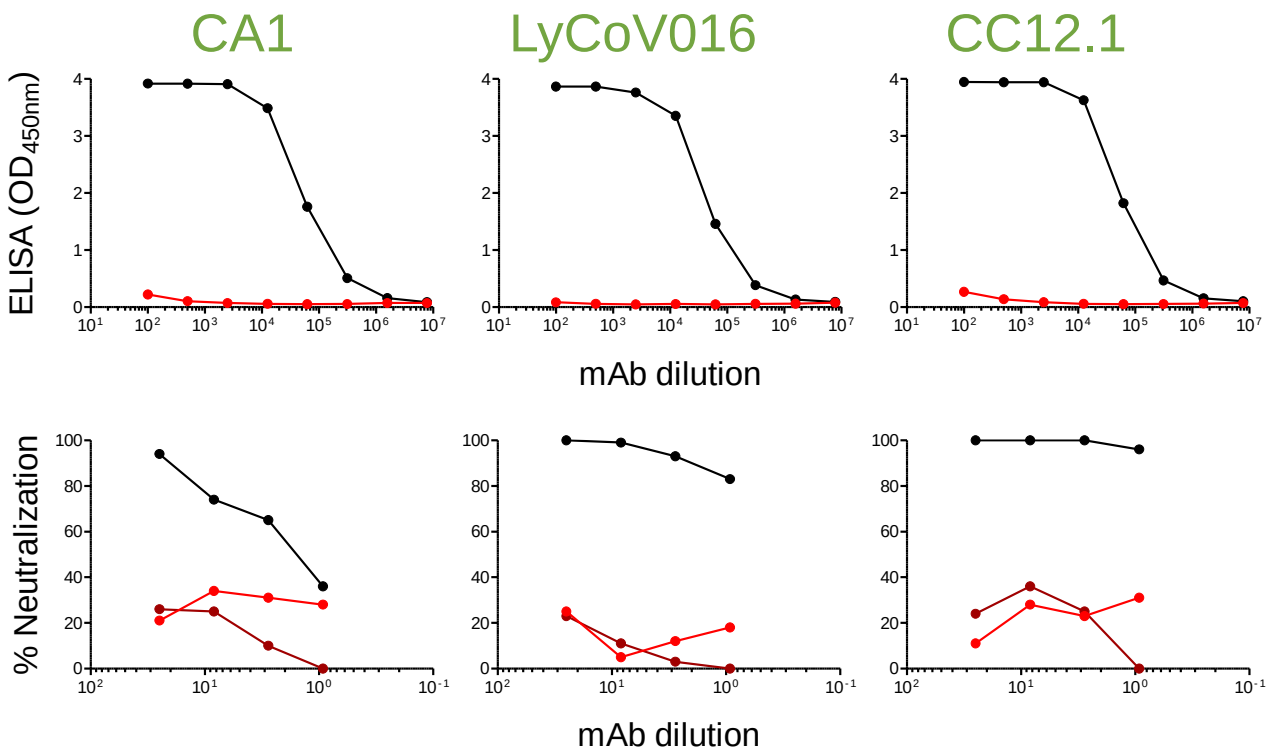
Check for updates

SARS-CoV-2 501Y.V2 escapes neutralization by South African COVID-19 donor plasma

Constantinos Kurt Wibmer[1], Frances Ayres[1], Tandile Hermanus[1], Mashudu Madzivhandila[1], Prudence Kgagudi[1], Brent Oosthuysen[1], Bronwen E. Lambson[1,2], Tulio de Oliveira[3], Marion Vermeulen[4], Karin van der Berg[4,5], Theresa Rossouw[6], Michael Boswell[7], Veronica Ueckermann[7], Susan Meiring[1], Anne von Gottberg[1,8], Cheryl Cohen[1,9], Lynn Morris[1,2], Jinal N. Bhiman[1,10,11] and Penny L. Moore[1,2,11] ✉

Beta variant is resistant to “class 1” mAbs

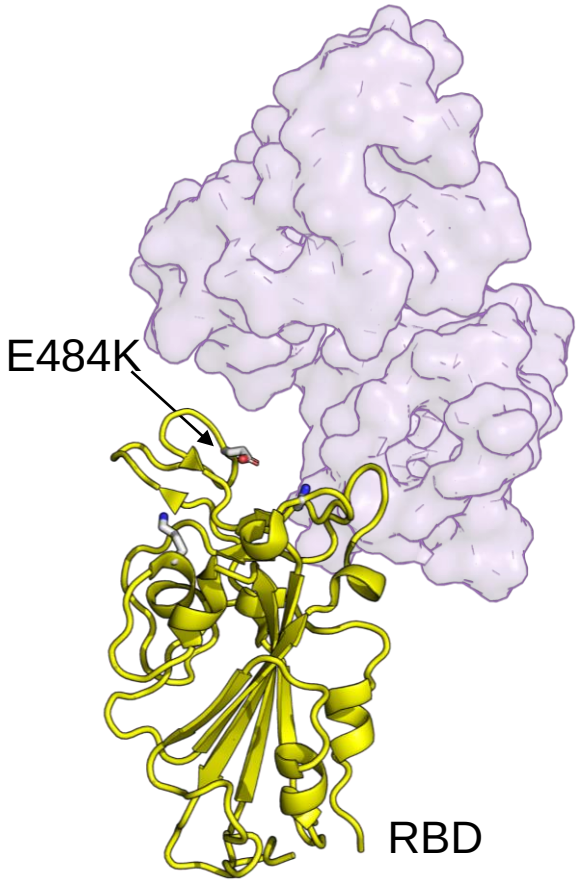


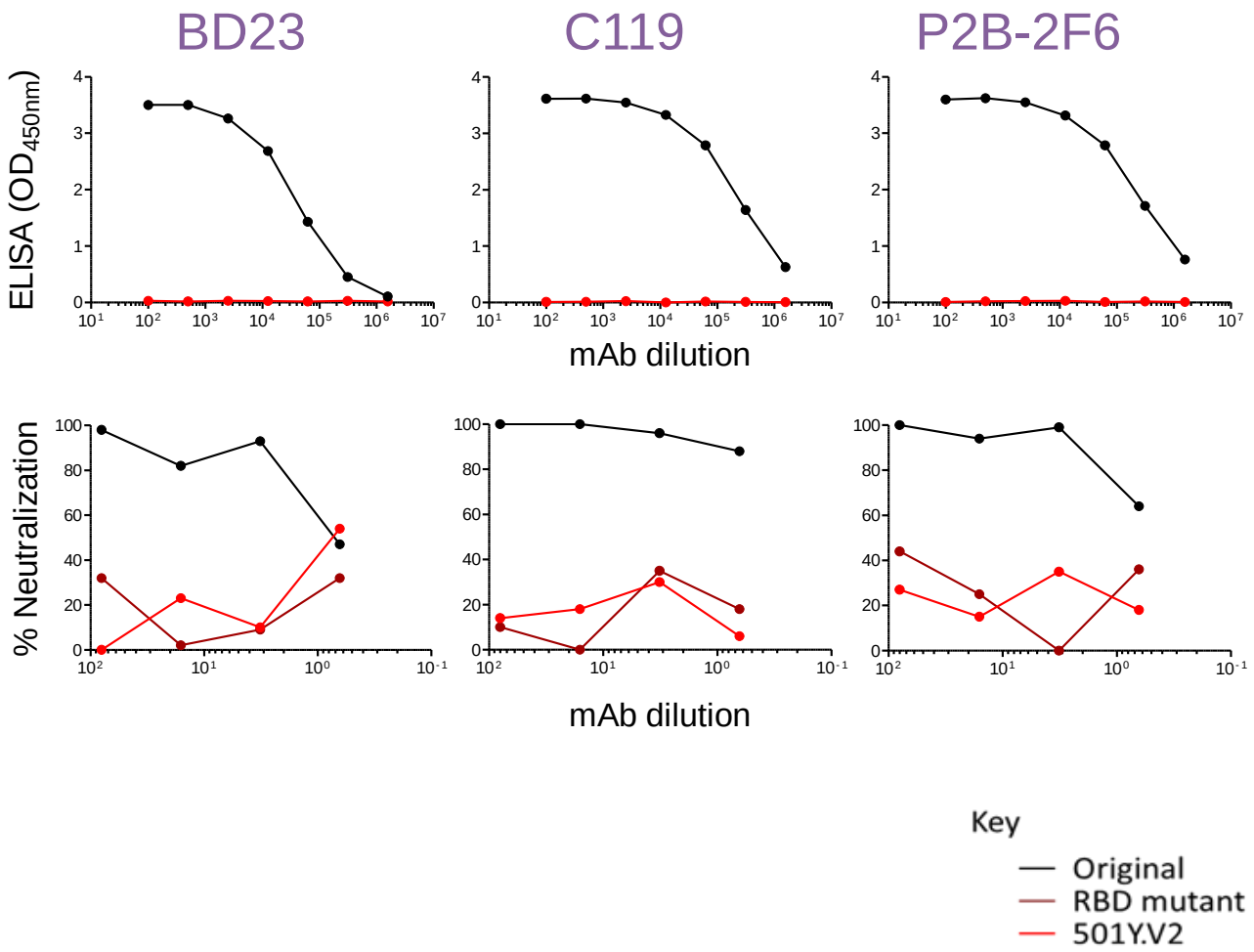


Key
— Original
— RBD mutant
— 501Y.V2

Wibmer et al, Nature Medicine, 2021

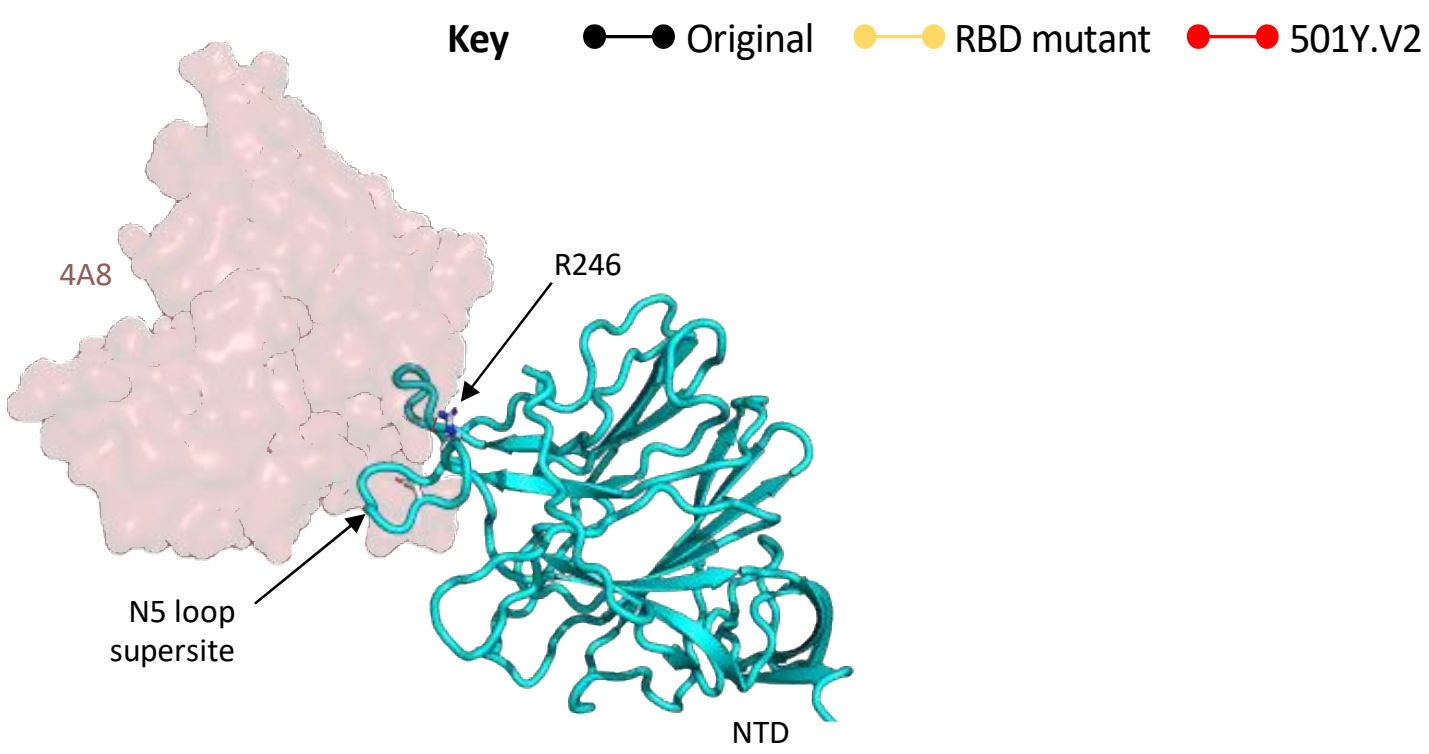
Beta variant is resistant to “class 2” mAbs

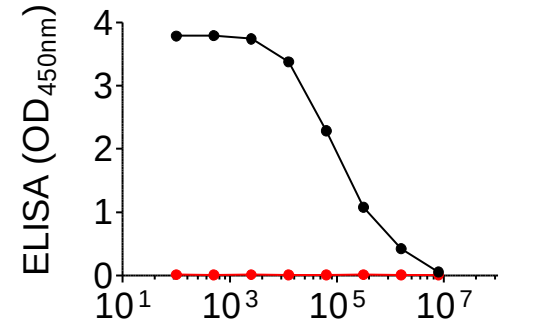


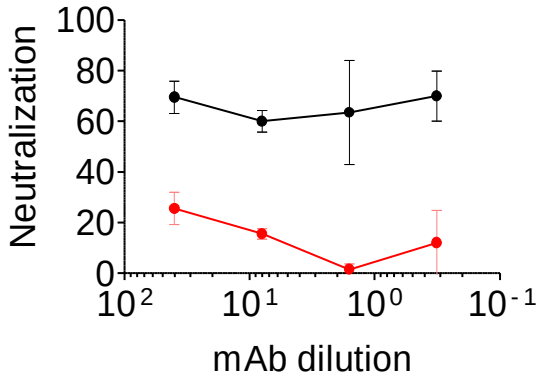


Wibmer et al, Nature Medicine, 2021

Beta variant is resistant to NTD-directed mAbs



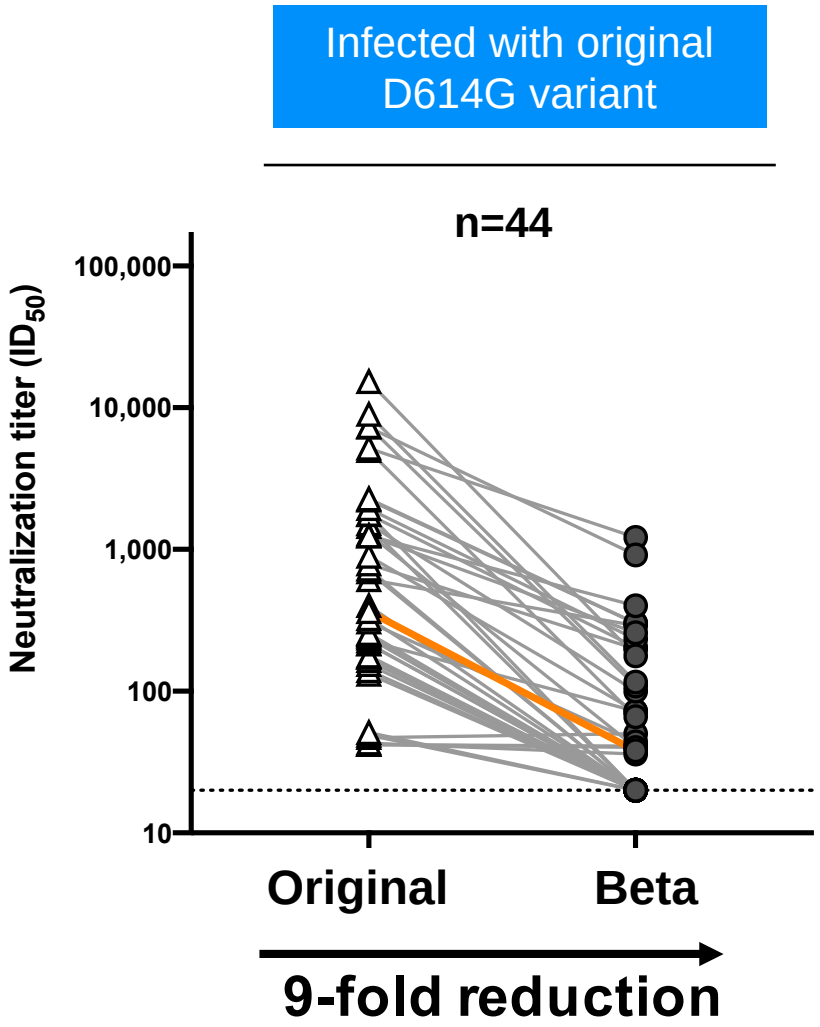


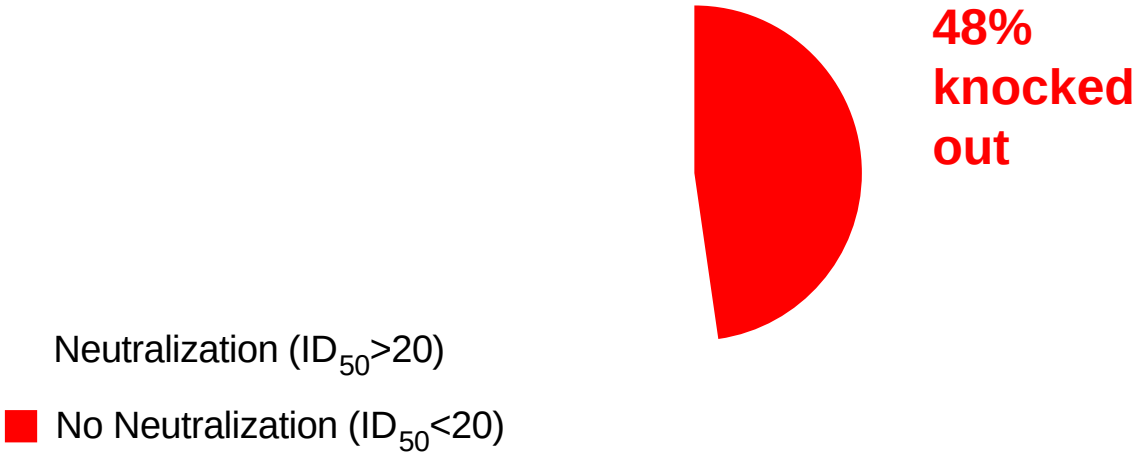


The Beta variant exhibits escape from three classes of therapeutically relevant monoclonal antibodies.

Wibmer et al, Nature Medicine, 2021

Beta escapes neutralization by South African COVID-19 donor plasma

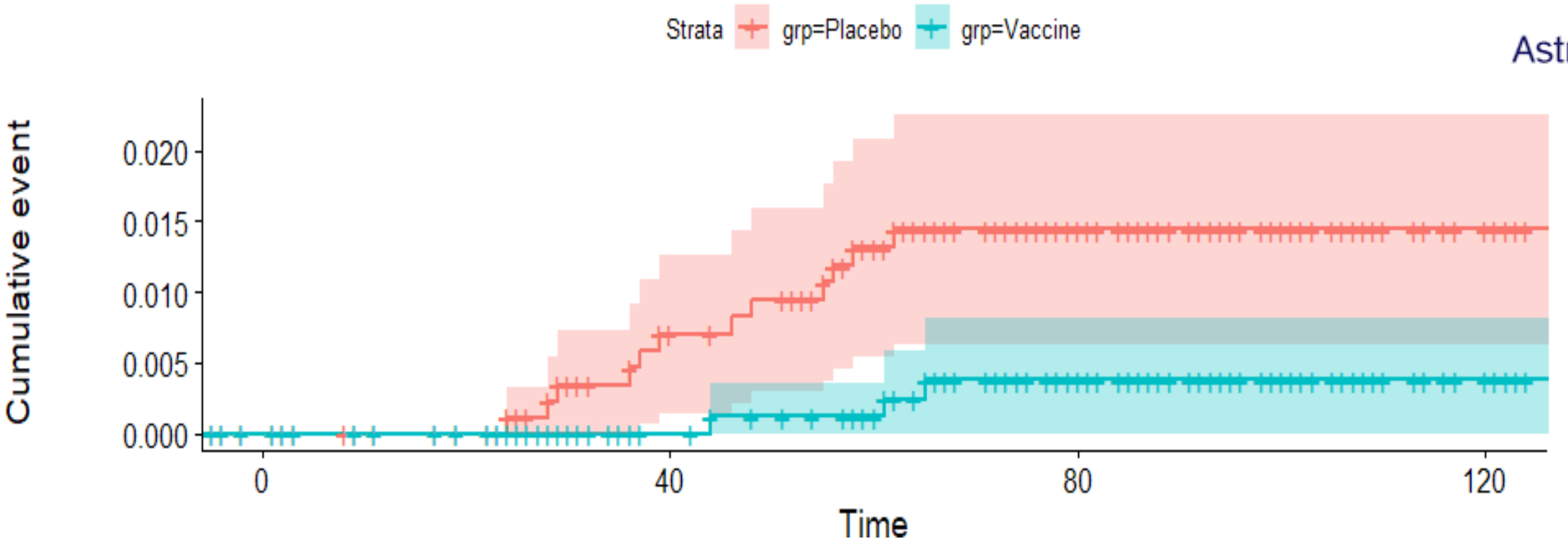




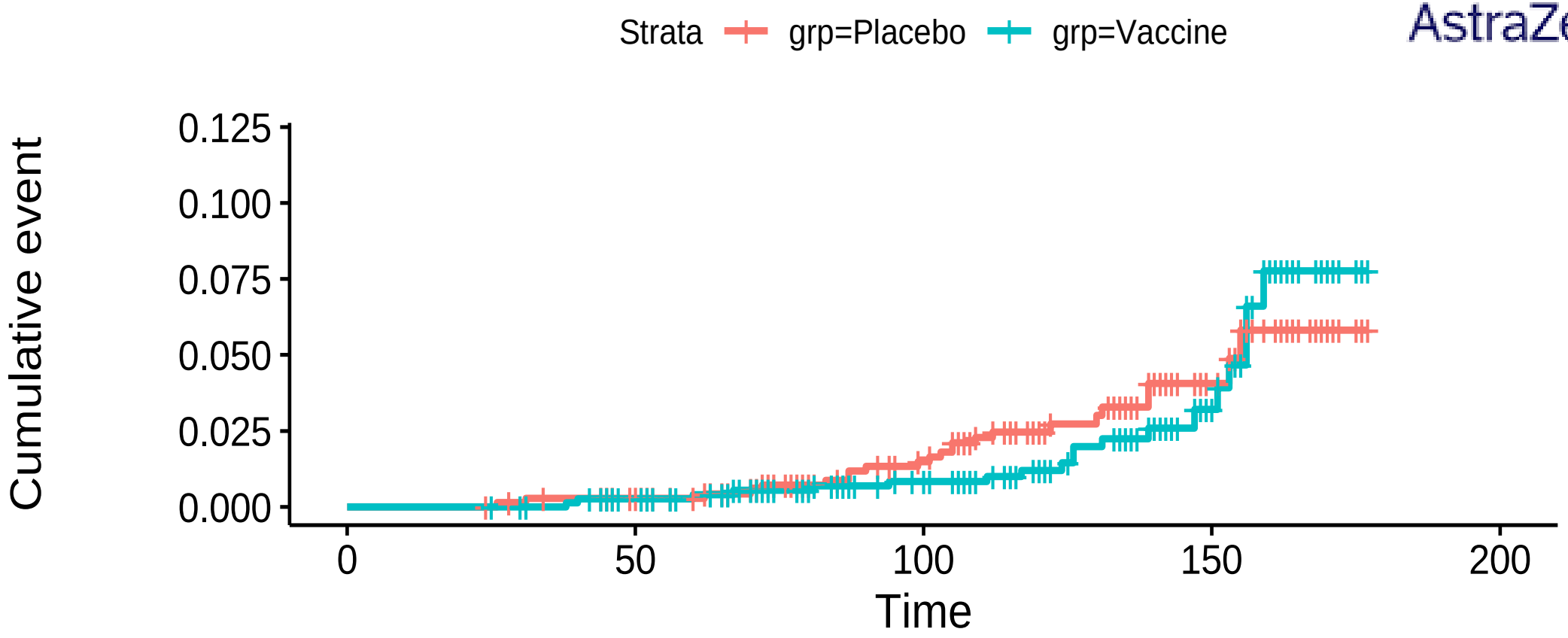
Implications of immune escape for vaccine efficacy?

Wibmer et al, Nature Medicine, 2021

How well does the AstraZeneca vaccine neutralize Beta?

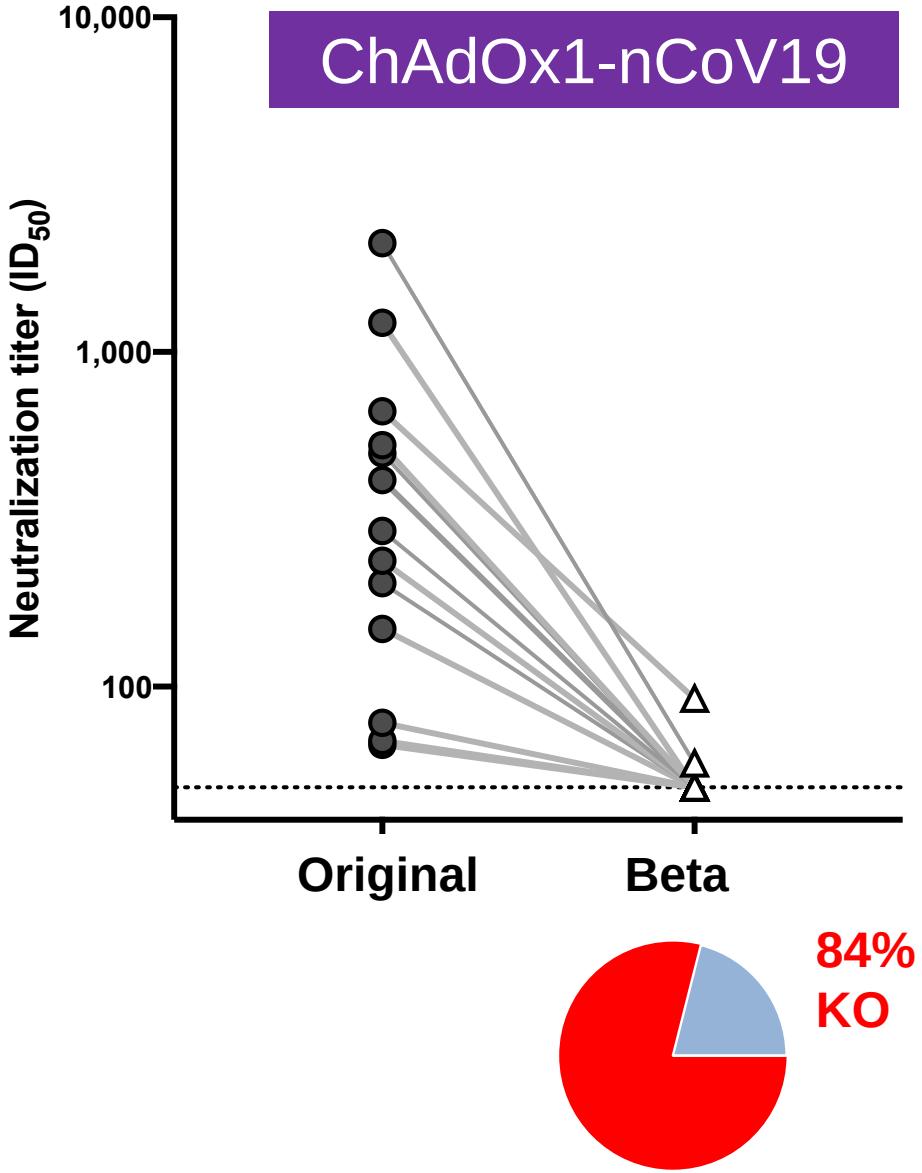


75% risk reduction in mild-moderate Covid-19 occurring at least 14 days after single dose of ChAdOx1/nCoV19 prior to evolution of the Beta variant in South Africa.



No significant risk reduction in mild-moderate Covid-19 from the Beta variant occurring at least 14 days after 2nd dose of ChAdOx1/nCoV19.

How well do vaccinee sera neutralize the Beta variant?



Protection against severe disease caused by Beta is unknown

Madhi et al, NEJM, 2021

The GRAND ARRIVAL of the AstraZeneca vaccine in SA

AstraZeneca

The AZ vaccine roll-out was halted in SA

Conclusion I

The Beta variant shows substantial and complete escape from:

- Three classes of therapeutically relevant mAbs
- Neutralizing antibodies in COVID-19 convalescent plasma
- Vaccinee sera, including Astrazeneca but questions remain about level and mechanism of protection from severe infection

The original variant doesn't make good cross-reactive Ab responses. What about the Beta variant?

- Does the Beta variant elicit robust antibody responses?

- Is there cross-reactivity in binding and neutralization by antibodies triggered by Beta?

The NEW ENGLAND JOURNAL of MEDICINE

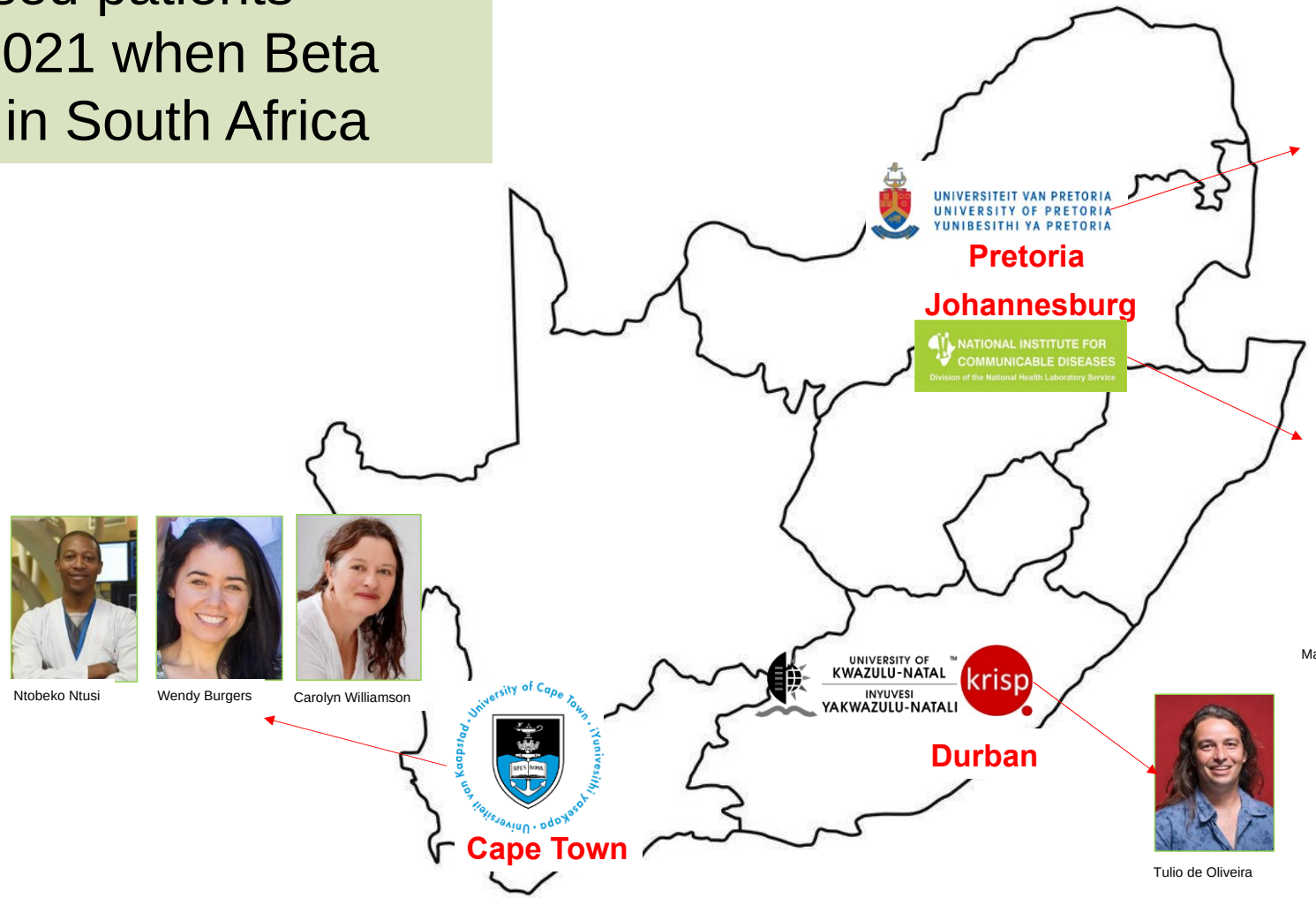
CORRESPONDENCE

Cross-Reactive Antibody Responses Elicited by the 501Y.V2 Variant

T. Moyo-Gwete and Others

Cohort of 89 hospitalised patients recruited in Jan/Feb 2021 when Beta was dominant variant in South Africa

Groote Schuur Hospital



Ntobeko Ntusi

Wendy Burgers

Carolyn Williamson

Tulio de Oliveira


Theresa Rossouw


Michael Boswell


Penny Moore

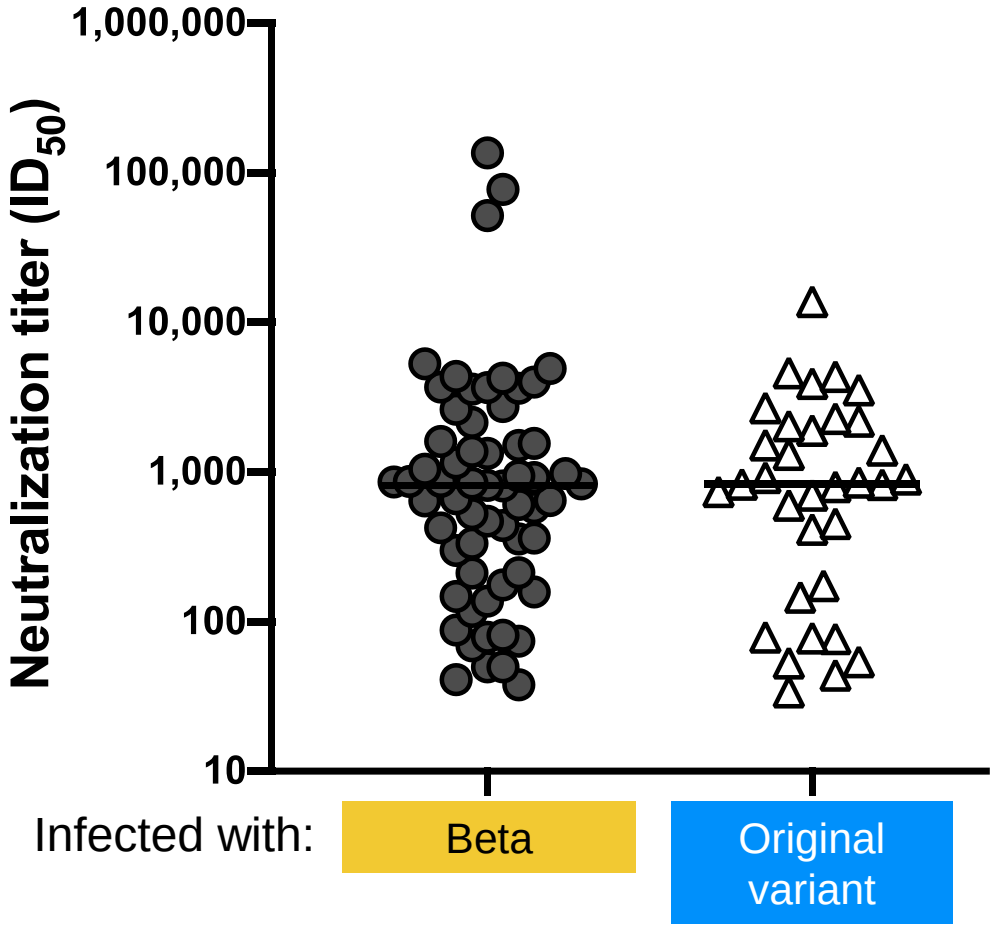

Thandeka Moyo-Gwete


Mashudu Madzivhandila


Lynn Morris

Moyo-Gwete, Madzivhandila et al, NEJM, 2021

Does Beta infection elicit potent neutralizing antibodies?

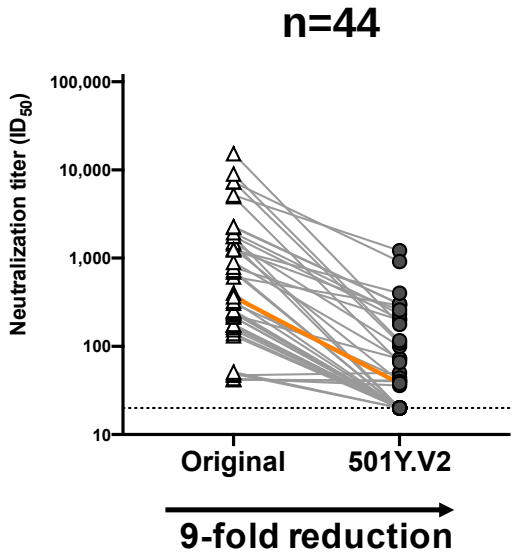


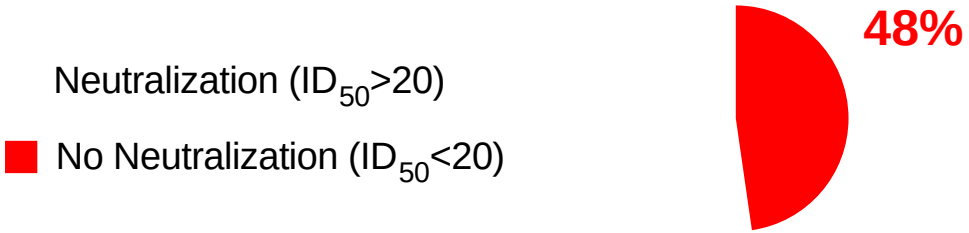
Beta infection elicits potent binding (not shown) and neutralizing antibodies

Moyo-Gwete, Madzivhandila et al, NEJM, 2021

Are neutralizing antibodies to Beta (501Y.V2) cross-reactive?

Infected with original variant



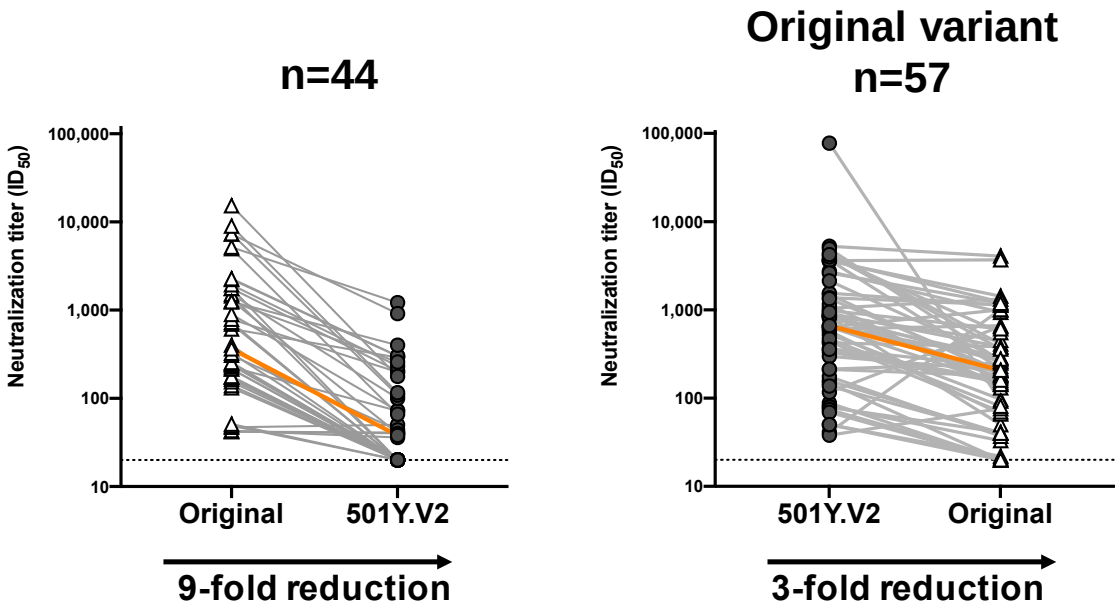


Moyo-Gwete, Madzivhandila et al, NEJM, 2021

Are neutralizing antibodies to 501Y.V2 cross-reactive?

Infected with original variant

infected with Beta



48%

7%

Neutralization (ID_{50}>20)

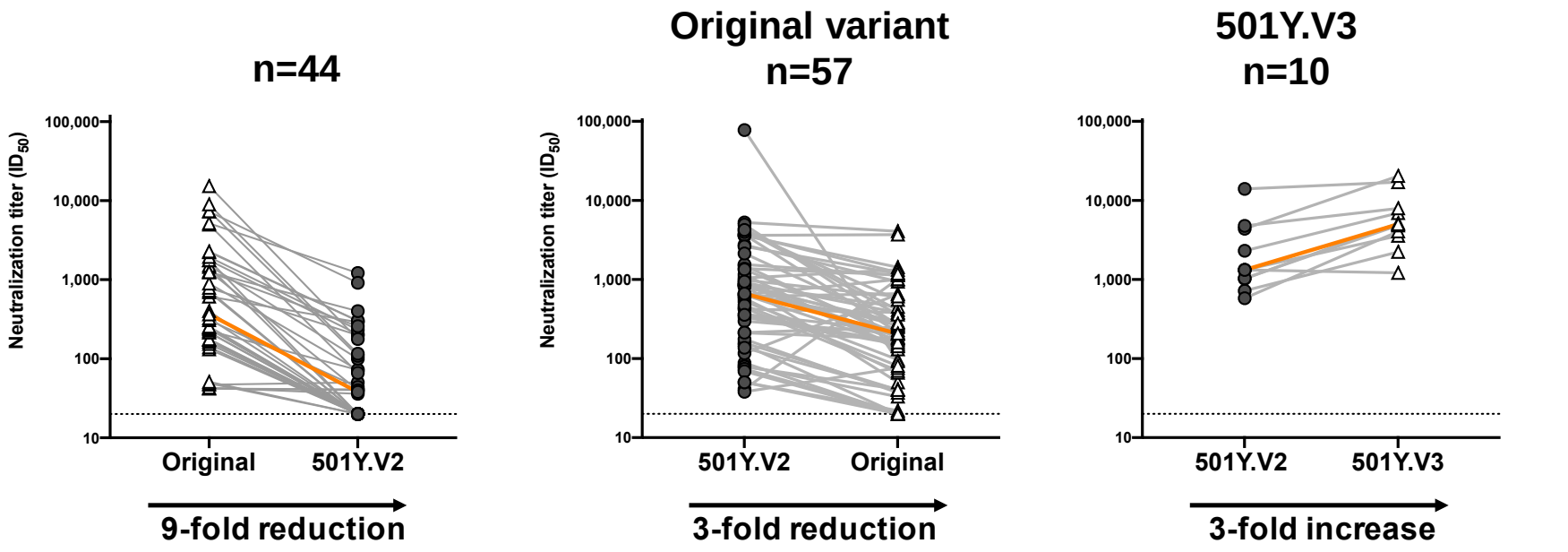
No Neutralization (ID_{50}<20)

Moyo-Gwete, Madzivhandila et al, NEJM, 2021

Are neutralizing antibodies to Beta (501Y.V2) cross-reactive?

Infected with original variant

infected with 501Y.V2



48%

7%

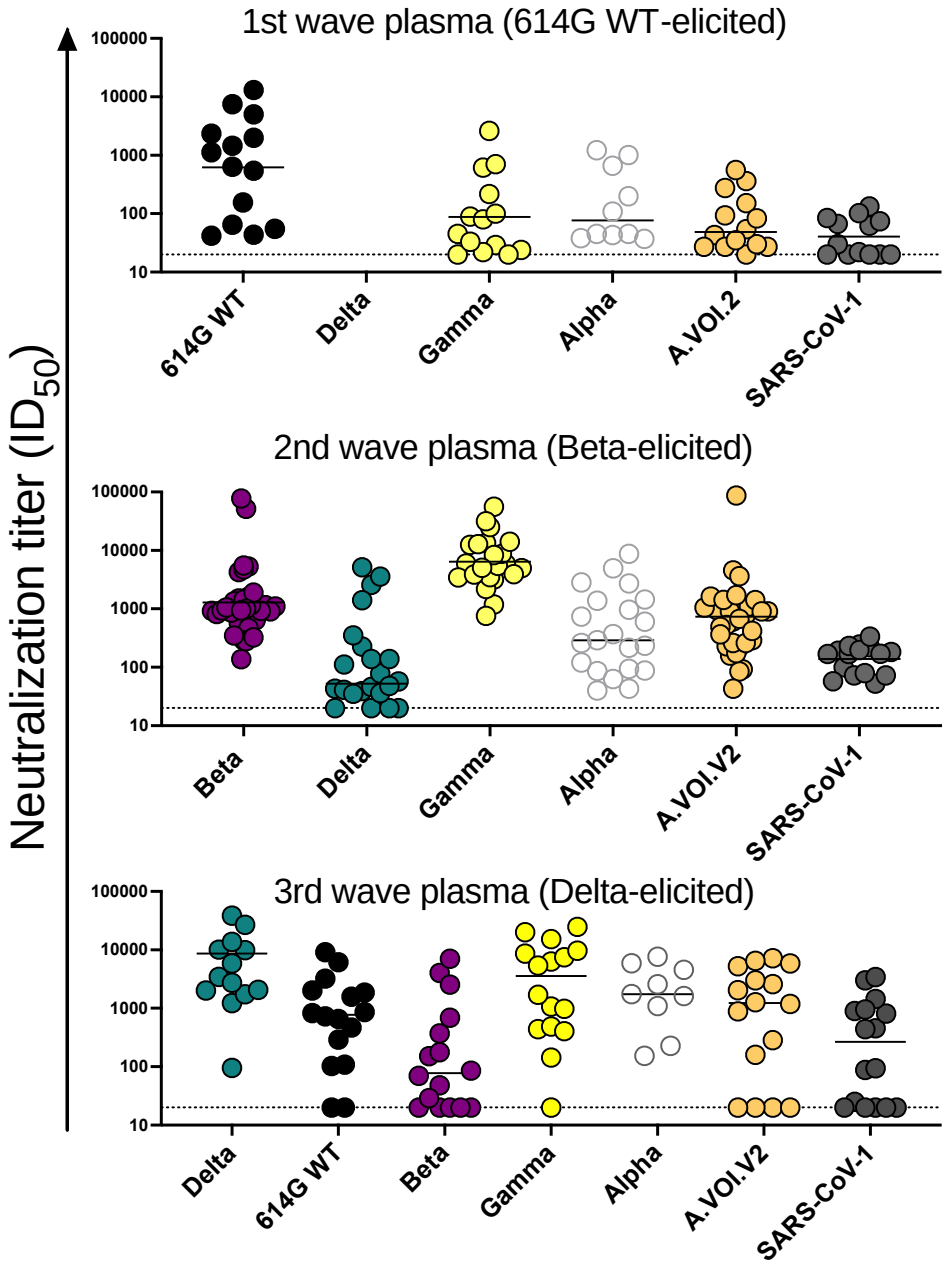
0%

Neutralization (ID_{50}>20)

No Neutralization (ID_{50}<20)

Moyo-Gwete, Madzivhandila et al, NEJM, 2021

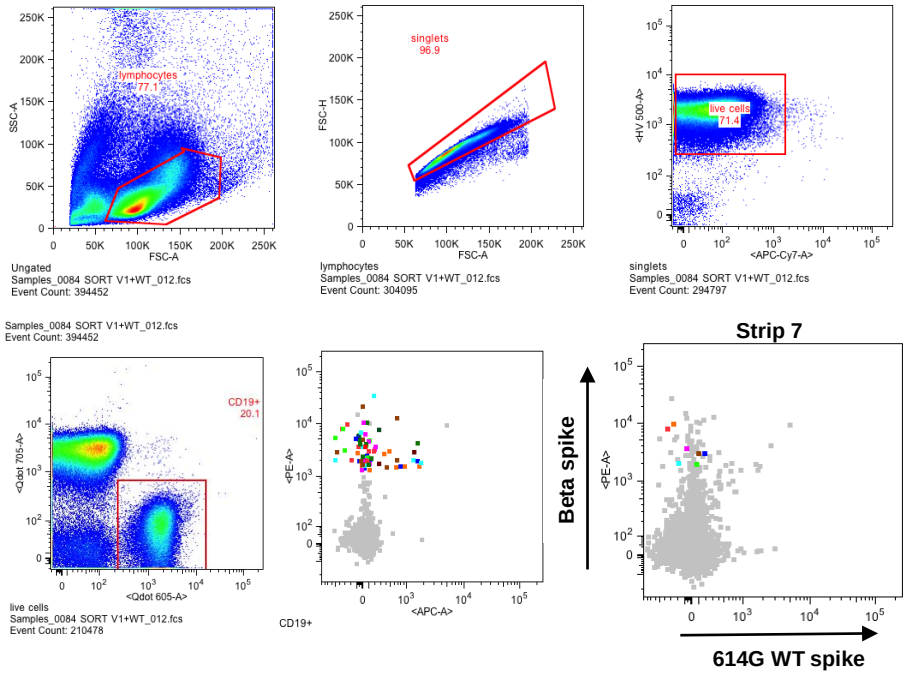
Beta and Delta-elicited plasma is more cross-reactive than orginal (WT) plasma

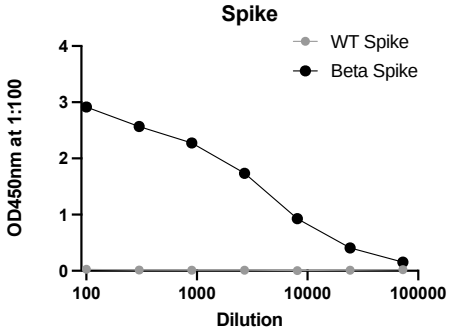


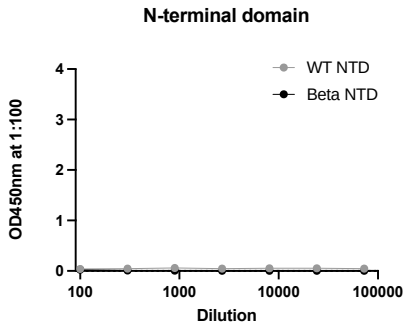
Moyo-Gwete, et al, manuscript in preparation

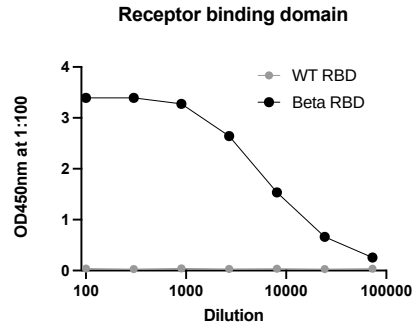
Isolation of a Beta-specific antibody (K417N-dependent)

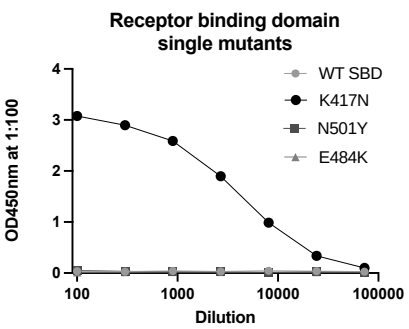
Pseudovirus	084 Plasma
614G WT	20
Beta	7613
Delta	20
Delta+ (N417K)	62893
Gamma	26
A.VOI.V2	89
SARS-CoV-1	20











Pseudovirus	084-7D
614G WT	20
Beta	0,1
Delta	20
Delta+ (N417K)	0,01
Gamma	20
A.VOI.V2	20
SARS-CoV-1	20

In the process of obtaining the structure in collaboration with Trevor Sewell at the University of Cape Town

Vaccine-elicited immunity following SARS-CoV-2 infection (ancestral and Beta)

In collaboration with Wendy Burgers and Ntobeko Ntusi, University of Cape Town

Cell Host & Microbe

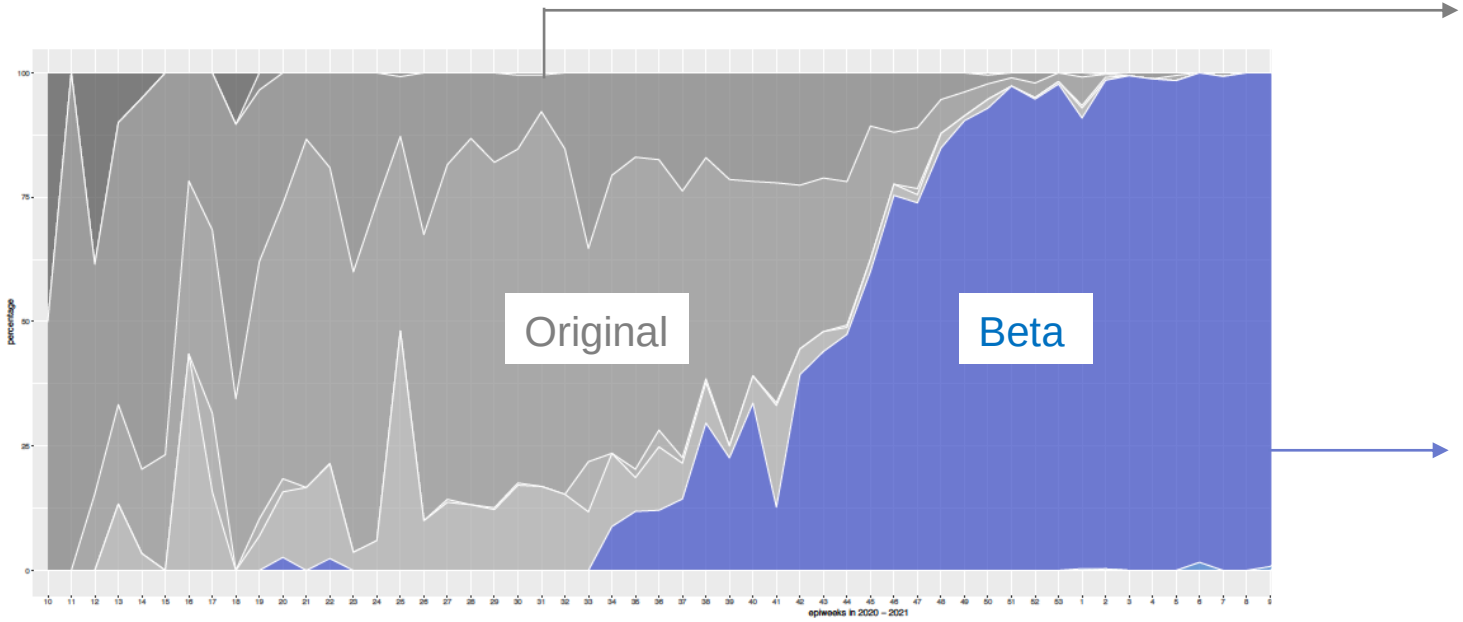
CellPress

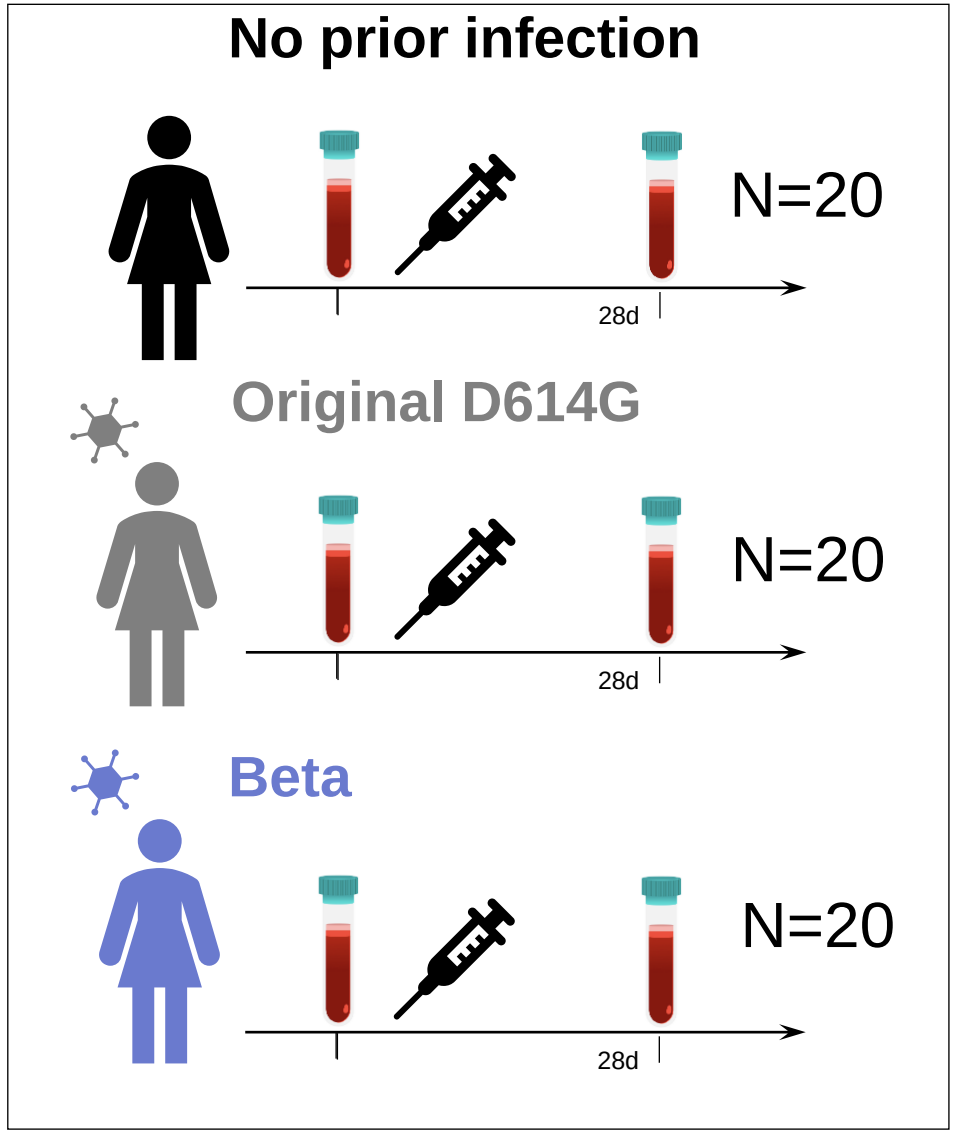
Short article

Prior infection with SARS-CoV-2 boosts and broadens Ad26.COV2.S immunogenicity in a variant-dependent manner

Roanne Keeton,[1,2,20] Simone I. Richardson,[3,4,20] Thandeka Moyo-Gwete,[3,4,20] Tandile Hermanus,[3,4,20] Marius B. Tincho,[1,2] Ntombi Benede,[1,2] Nelia P. Manamela,[3,4] Richard Baguma,[1] Zanele Makhado,[3,4] Amkele Ngomti,[1,2] Thopisang Motlou,[3,4] Mathilda Mennen,[5] Lionel Chinhoyi,[5] Sango Skelem,[5] Hazel Maboreke,[1,6] Deelan Doolabh,[1,2] Arash Iranzadeh,[1,2] Ashley D. Otter,[7] Tim Brooks,[7] Mahdad Noursadeghi,[8] James C. Moon,[9,10] Alba Grifoni,[11] Daniela Weiskopf,[11] Alessandro Sette,[11,12] Jonathan Blackburn,[1,6] Nei-Yuan Hsiao,[2,13] Carolyn Williamson,[1,2,14] Catherine Riou,[1,2,14] Ameena Goga,[15] Nigel Garrett,[16,17] Linda-Gail Bekker,[1,18] Glenda Gray,[15] Ntobeko A.B. Ntusi,[1,5,19,21,*] Penny L. Moore,[3,4,21,*] and Wendy A. Burgers[1,2,14,21,22,*]

What is the effect of previous infection on responses after JnJ vaccination?

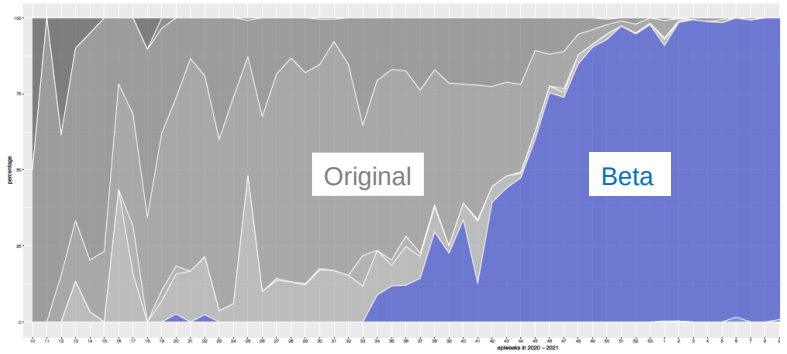


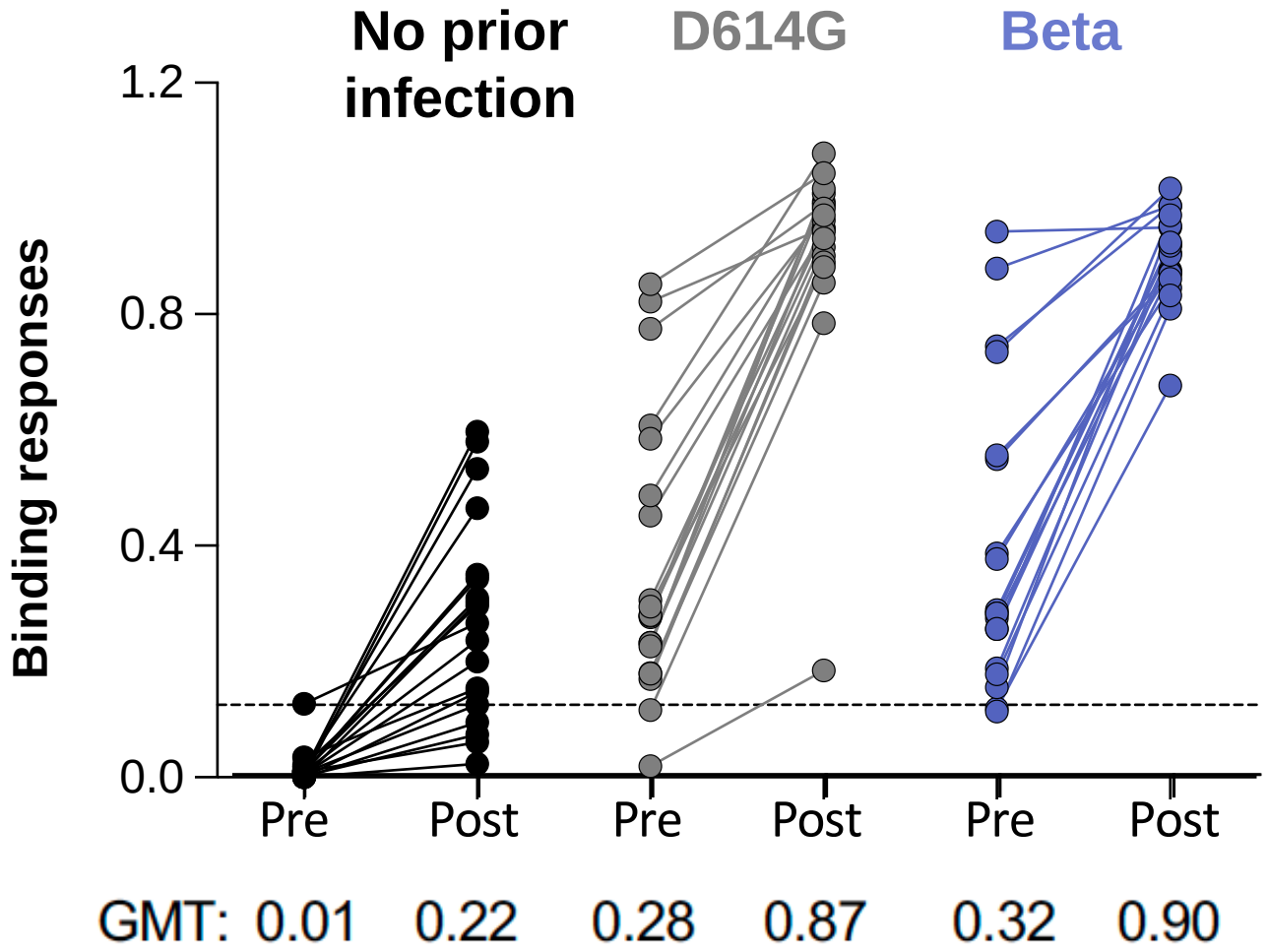


With Wendy Burgers and Ntobeko Ntusi

Keeton, Richardson, Moyo-Gwete et al, CHM

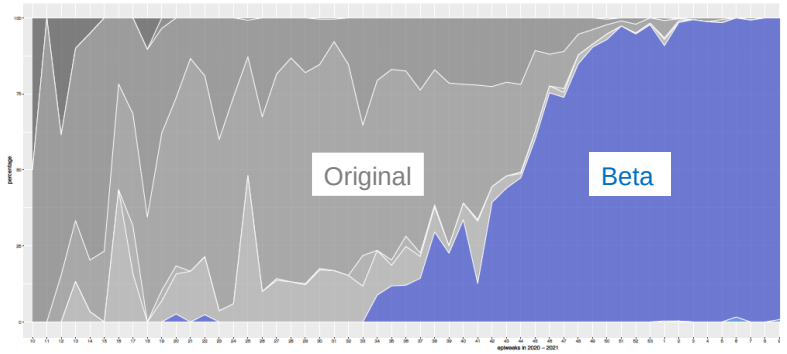
Previous infection results in Ad26.COV2.S boosting of binding antibodies

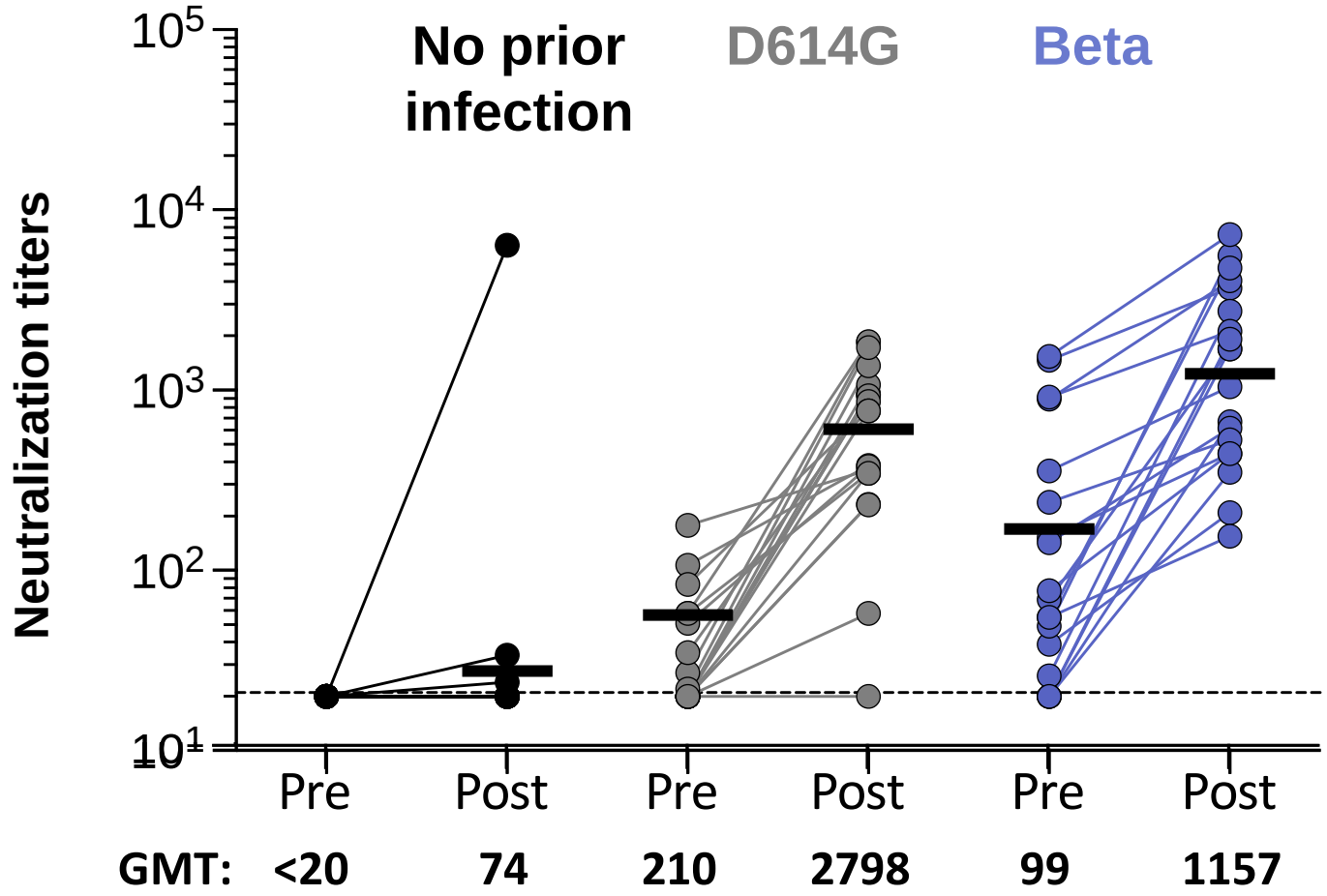




Keeton, Richardson, Moyo-Gwete et al, CHM

Previous infection results in Ad26.COV2.S boosting of neutralizing antibodies





Similar to other vaccines, SARS-CoV-2 infection before Ad26.COV2.S boosts neutralizing antibodies

Conclusion II

- SARS-CoV-2 infection before Ad26.COV2.S boosts binding and neutralizing antibody activity
- In high SARS-CoV-2 seroprevalence areas, such as South Africa, this will significantly impact vaccine efficacy – especially with booster vaccines available

Immune responses elicited by vaccination in people living with HIV

- 7.5 million South Africans are HIV positive
- 19% adult HIV prevalence ages 15-49)

Safety and immunogenicity of the ChAdOx1 nCoV-19 (AZD1222) vaccine against SARS-CoV-2 in people living with and without HIV in South Africa: an interim analysis of a randomised, double-blind, placebo-controlled, phase 1B/2A trial

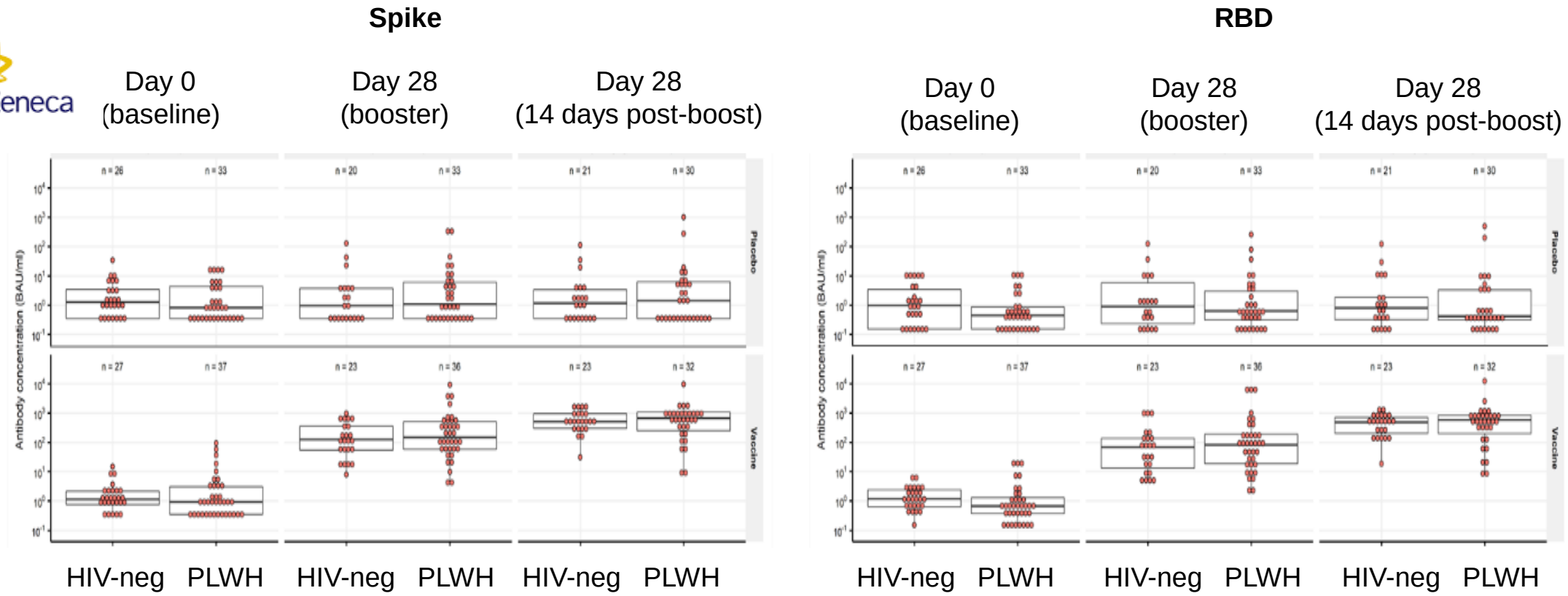
Shabir A Madhi, Anthonet L Koen, Alane Izu, Lee Fairlie, Clare L Cutland, Vicky Baillie, Sherman D Padayachee, Keertan Dheda, Shaun L Barnabas, Qasim Ebrahim Bhorat, Carmen Briner, Parvinder K Aley, Sutika Bhikha, Tandile Hermanus, Elizea Horne, Aylin Jose, Prudence Kgagudi, Teresa Lambe, Masebole Masenya, Mduduzi Masilela, Nonhlanhla Mkhize, Andrew Moultrie, Christian K Mukendi, Thandeka Moyo-Gwete, Amit J Nana, Ayanda Nzimande, Faeezah Patel, Sarah Rhead, Carol Taoushanis, Asha Thombrayil, Samuel van Eck, Merryn Voysey, Tonya L Villafana, Johan Vekemans, Sarah C Gilbert, Andrew J Pollard, Penny L Moore, Gaurav Kwatra*, on behalf of the Wits VIDA COVID team†*

Divergence of Delta and Beta variants and SARS-CoV-2 evolved in advanced HIV disease into two serological phenotypes

Sandile Cele[1,2], Farina Karim[1,2], Gila Lustig[3], San Emmanuel James[4], Tandile Hermanus[5,6], Eduan Wilkinson[4,7], Jumari Snyman[1,8], Mallory Bernstein[1], Khadija Khan[1,2], Shi-Hsia Hwa[1,9], Houriiyah Tegally[4], Sasha W. Tilles[10], Lavanya Singh[4], Jennifer Giandhari[4], Ntombifuthi Mthabela[1], Matilda Mazibuko[1], Yashica Ganga[1], Bernadett I. Gosnell[11], Salim Abdool Karim[3], Willem Hanekom[1,9], Wesley C. Van Voorhis[10], Thumbi Ndung'u[1,8], COMMIT-KZN Team[§], Richard J. Lessells[2,3,4], Penny L. Moore[3,5,6], Mahomed-Yunus S. Moosa[11], Tulio de Oliveira[2,3,4,7,12], Alex Sigal[1,2,13*]

Binding to SARS-CoV-2 spike and RBD similar in PLWH and HIV-uninfected

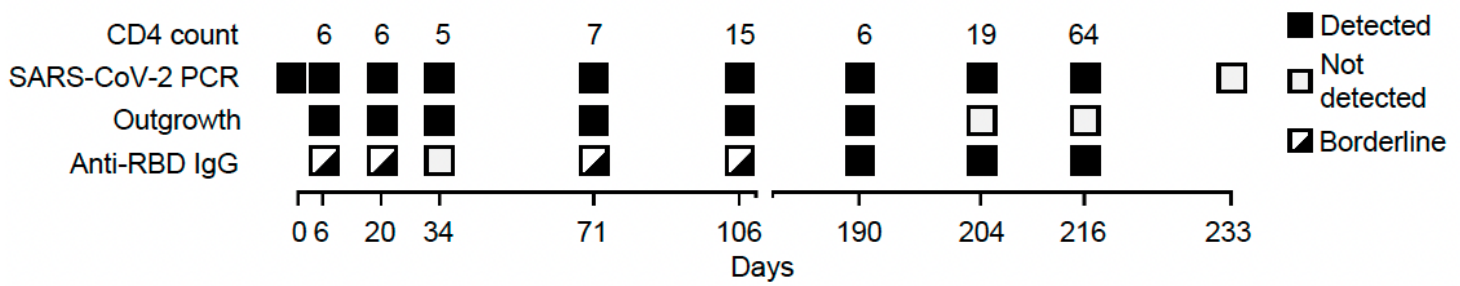
Randomised, double-blind, placebo-controlled phase 1b/2a trial in South Africa

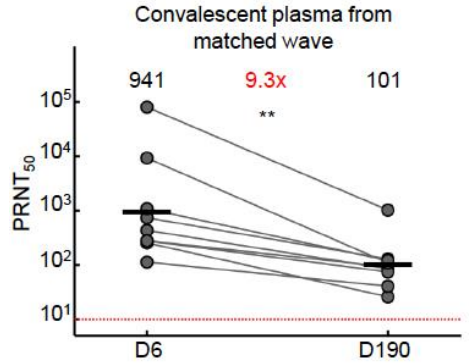


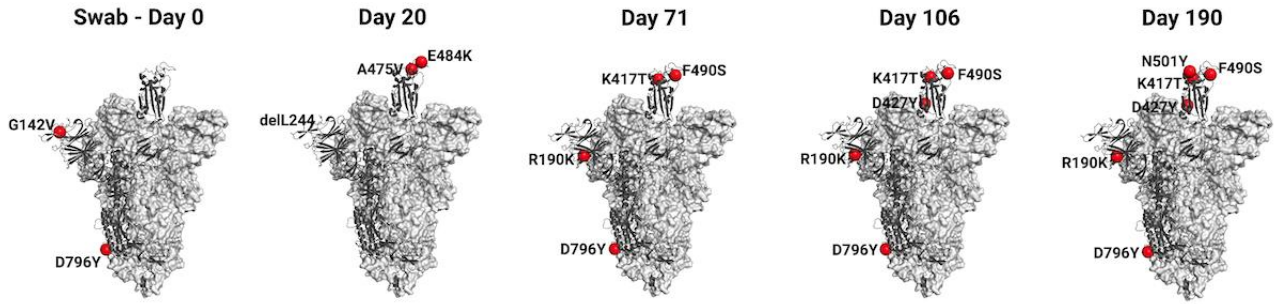
Madhi 2021, Lancet HIV

Emergence of resistant variants within long-term shedders

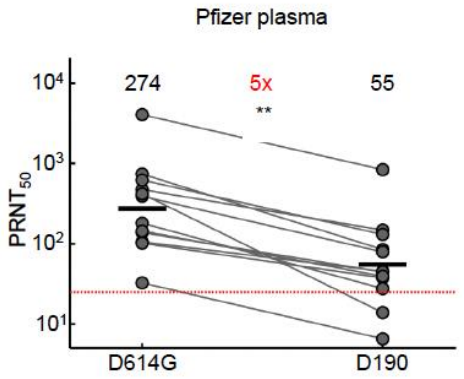
HIV unsuppressed individuals may be long-term SARS-CoV-2 shedders







Accumulation of mutations over time



Cele and Sigal, CHM, under revision

Final conclusions

- Structural biology is a key field for both virology and immunology – **without it we would not have a SARS-CoV-2 vaccine**
- Understanding the structure of a protein helps us understand its function and how to manipulate the protein to get a desired effect
- Although there is still no HIV vaccine, major strides have taken place over the last 40 years bringing us closer to this goal
- Structural biology will continue to be a relevant even in the face of the next global pandemic

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Ntobeko Ntusi
Carolyn Williamson
Roanne Keeton

UKZN
Tulio De Oliveira
Alex Sigal
Houriiyah Tegally
Sandile Cele

501Y.V2 consortium of South African scientists, chaired by Drs W Hanekom and Dr T De Oliveira

Clinical teams and participants