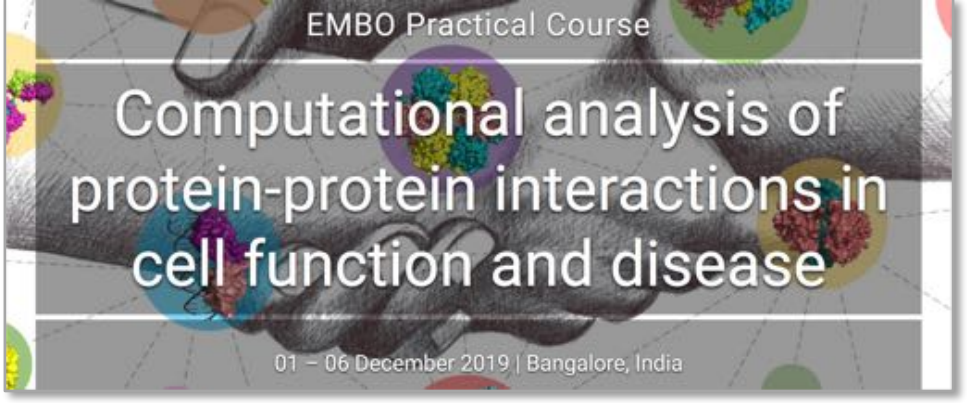


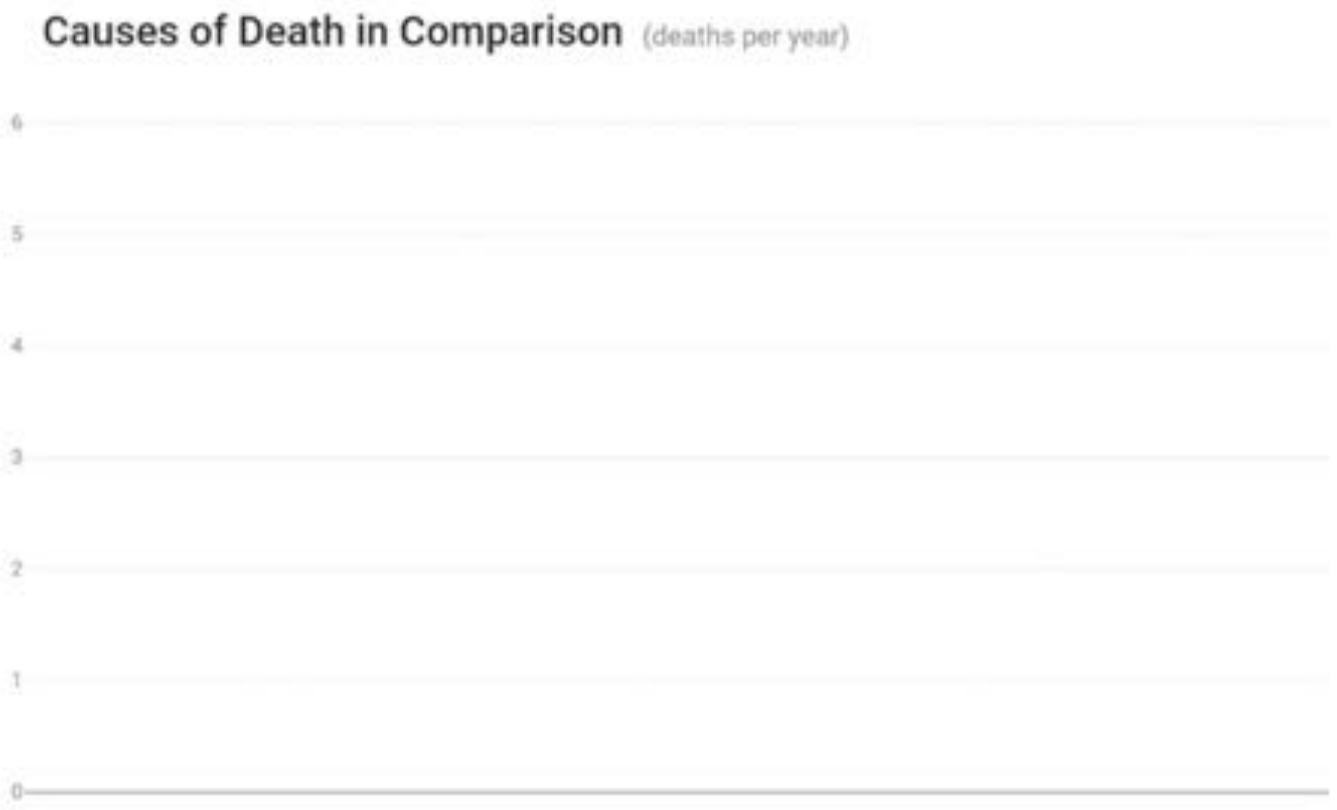
The impact of protein dynamics on the spread of viral infectious diseases:

Examples from the research in South Africa

Natasha Wood

2 December 2019

Selected Causes of Death in Comparison

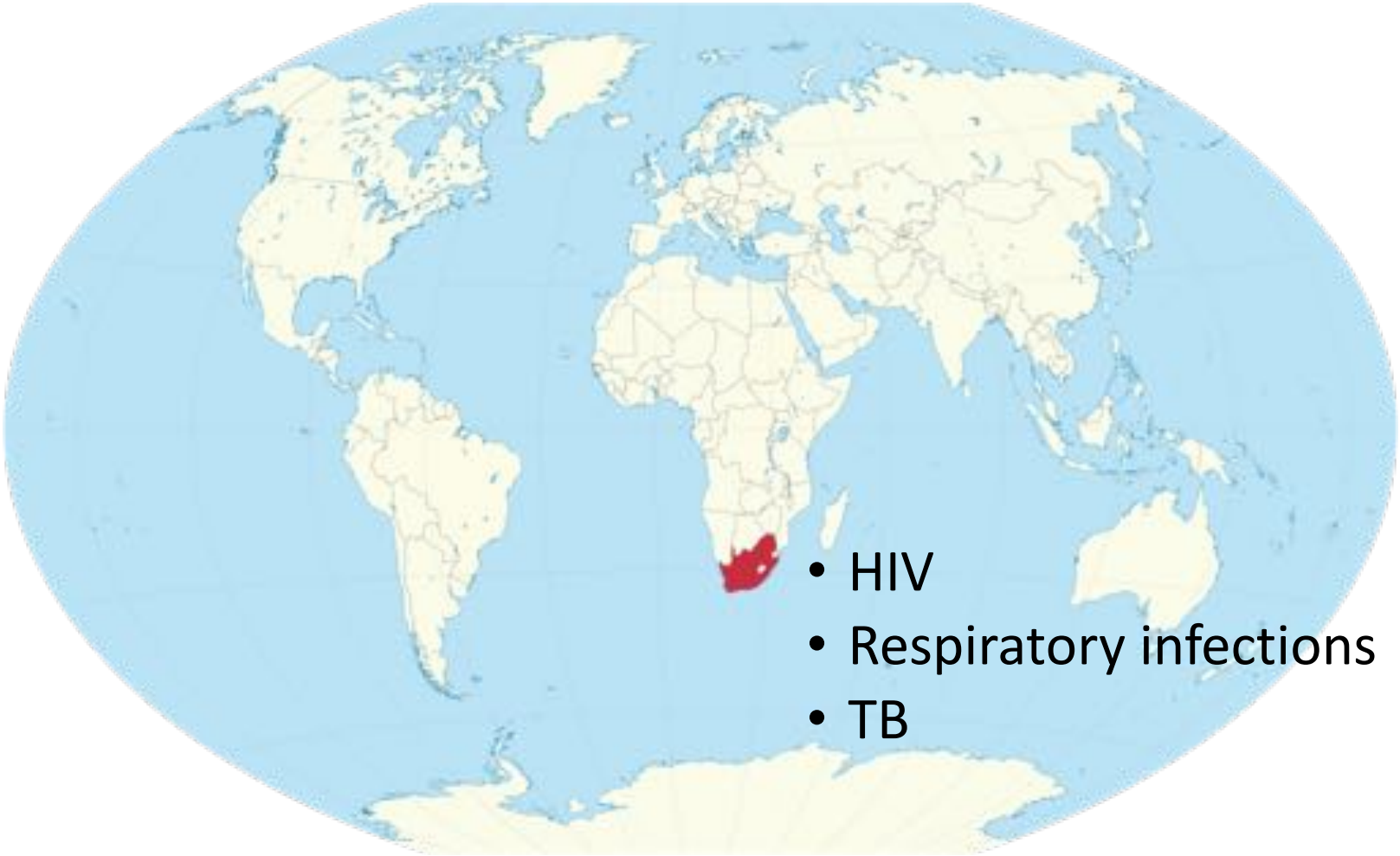


This is Data; Data sources: National Geographic, Our World In Data, Earth.com, The Guardian, BBC, crocBITE, BAAA

Selected Causes of Death in Comparison with the No. 1 - https://www.youtube.com/watch?v=X6xJVtMtLHY

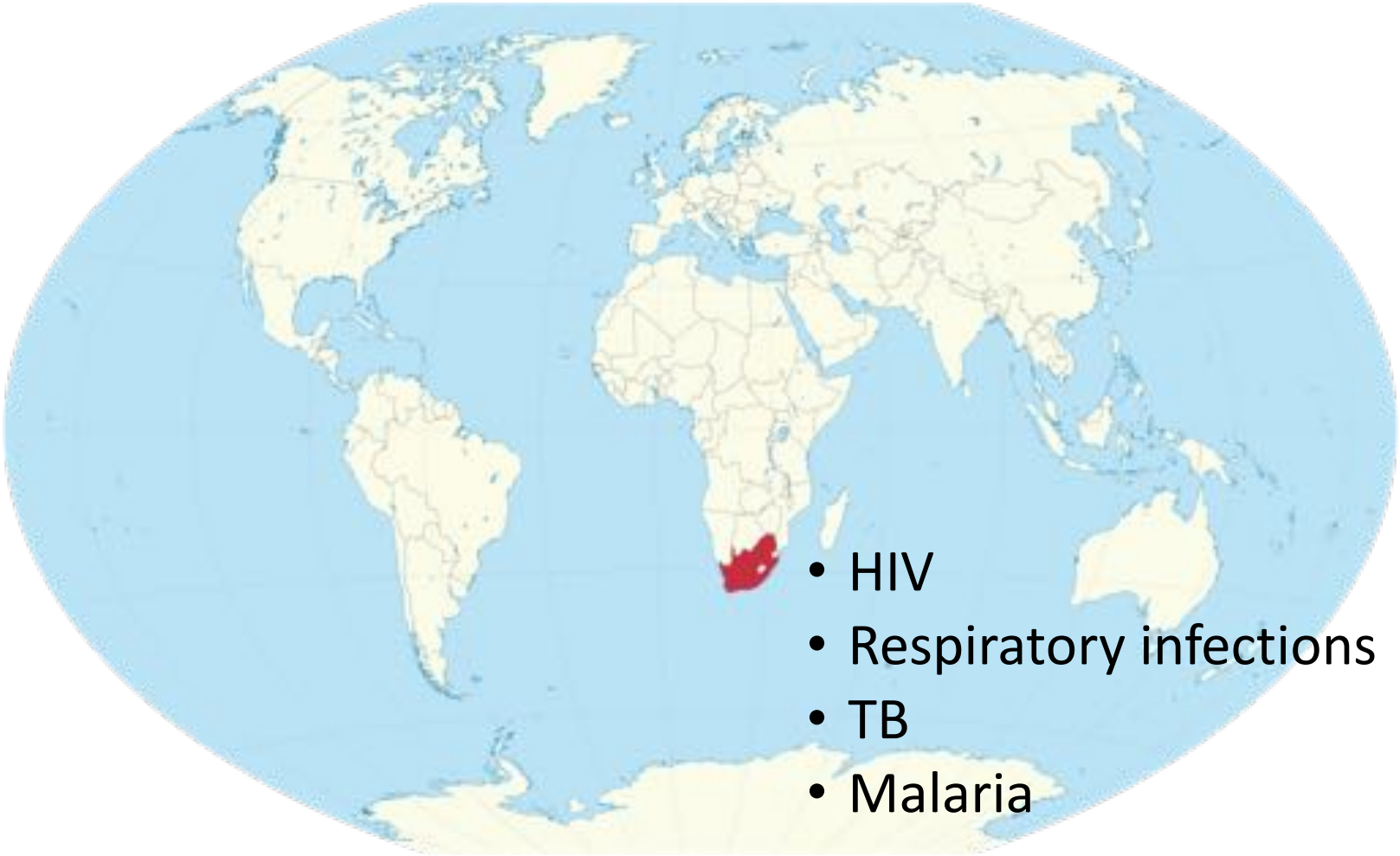
South Africa

South Africa



- HIV
- Respiratory infections
- TB

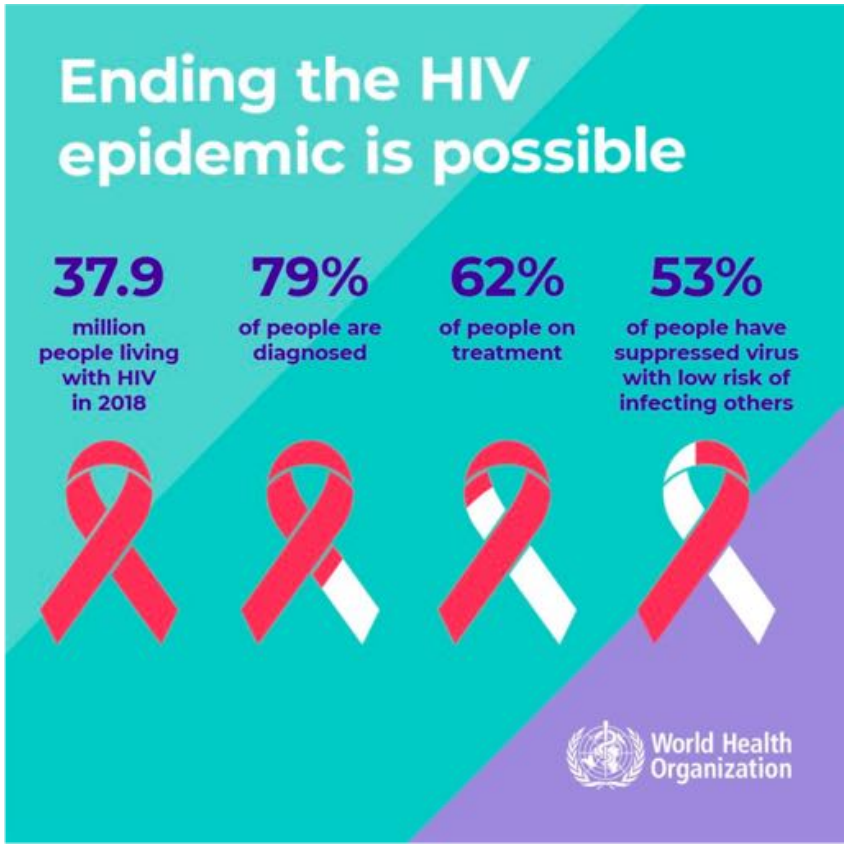
South Africa



- HIV
- Respiratory infections
- TB
- Malaria

HIV

World AIDS Day – 1 December



HIV

EDITORIAL

CRISPR's unwanted anniversary

There are key moments in the history of every disruptive technology that can make or break its public perception and acceptance. For CRISPR-based genome editing, such a moment occurred 1 year ago—an unsettling push into an era that will test how society decides to use this revolutionary technology.

In November 2018, at the Second International Summit on Human Genome Editing in Hong Kong, scientist He Jiankui announced that he had broken the basic medical mantra of "do no harm" by using CRISPR-Cas9 to edit the genomes of two human embryos in the hope of protecting the twin girls from HIV. His risky and medically unnecessary work stunned the world and defied prior calls by my colleagues and me, and by the U.S. National Academies of Sciences and of Medicine, for an effective moratorium on human claim that they did not know or were somehow operating within published guidelines. On the heels of WHO, an International Commission on the Clinical Use of Human Germline Genome Editing convened its first meeting to identify the scientific, medical, and ethical requirements to consider when assessing potential clinical applications of human germline genome editing. The U.S. National Academy of Medicine, the U.S. National Academy of Sciences, and the Royal Society of the United Kingdom lead this commission, with the participation of science and medical academies from around the world. Already this week, the commission held a follow-up meeting, reflecting the urgent nature of their mission.

Where is CRISPR technology headed? Since 2012, it has transformed basic research, drug development, diagnostics, agriculture, and synthetic biology. Fu-

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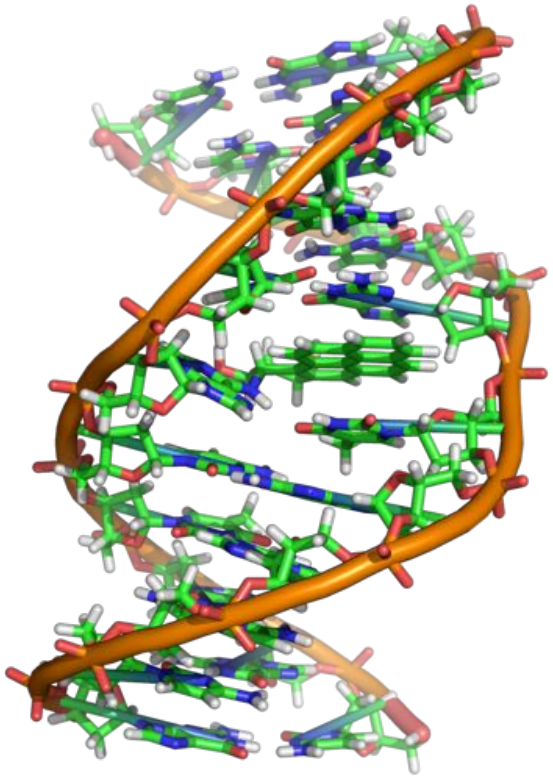
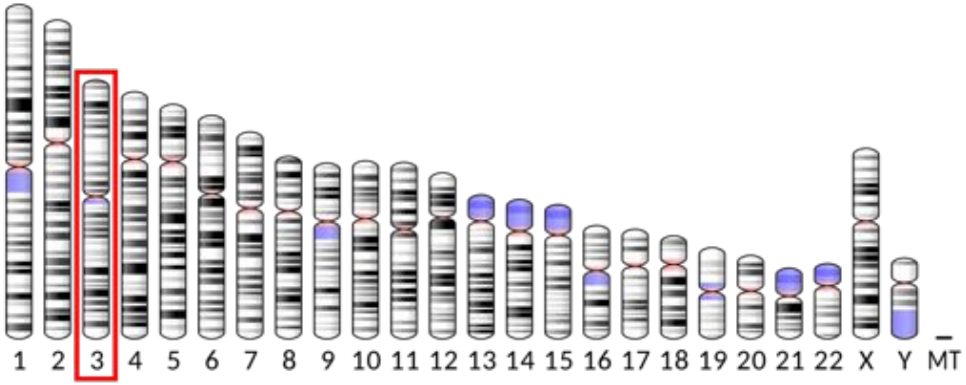
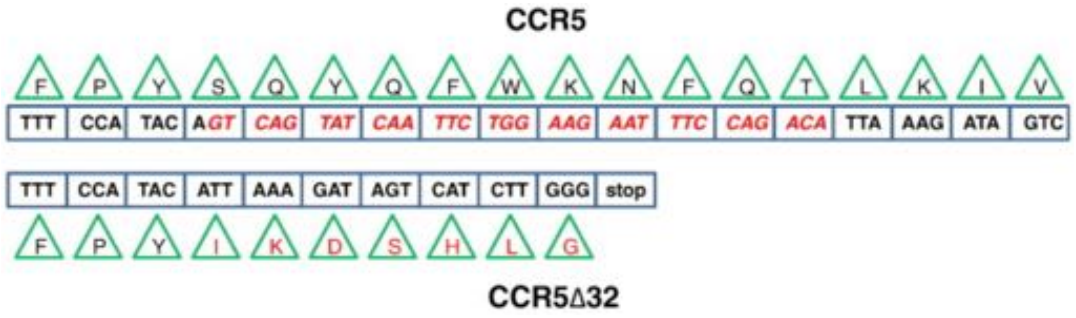
CCR5Δ32 mutation



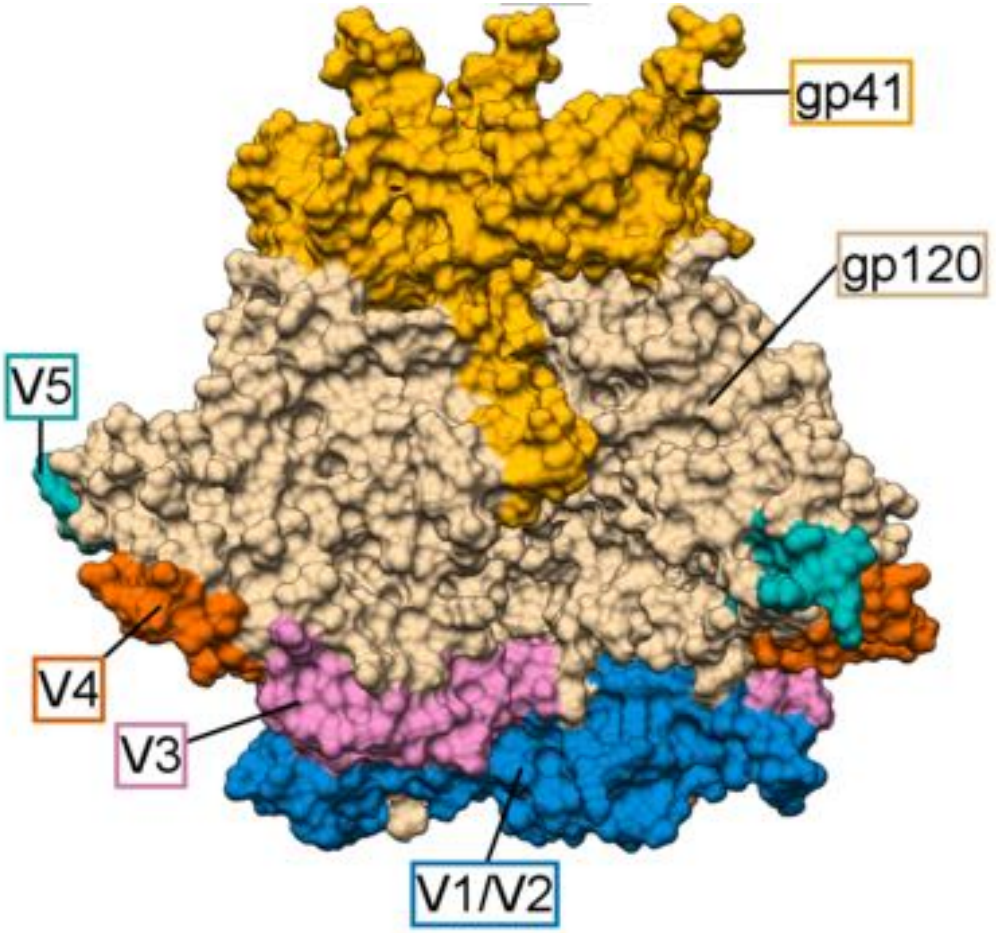


Chromosome 3: CCR5 gene

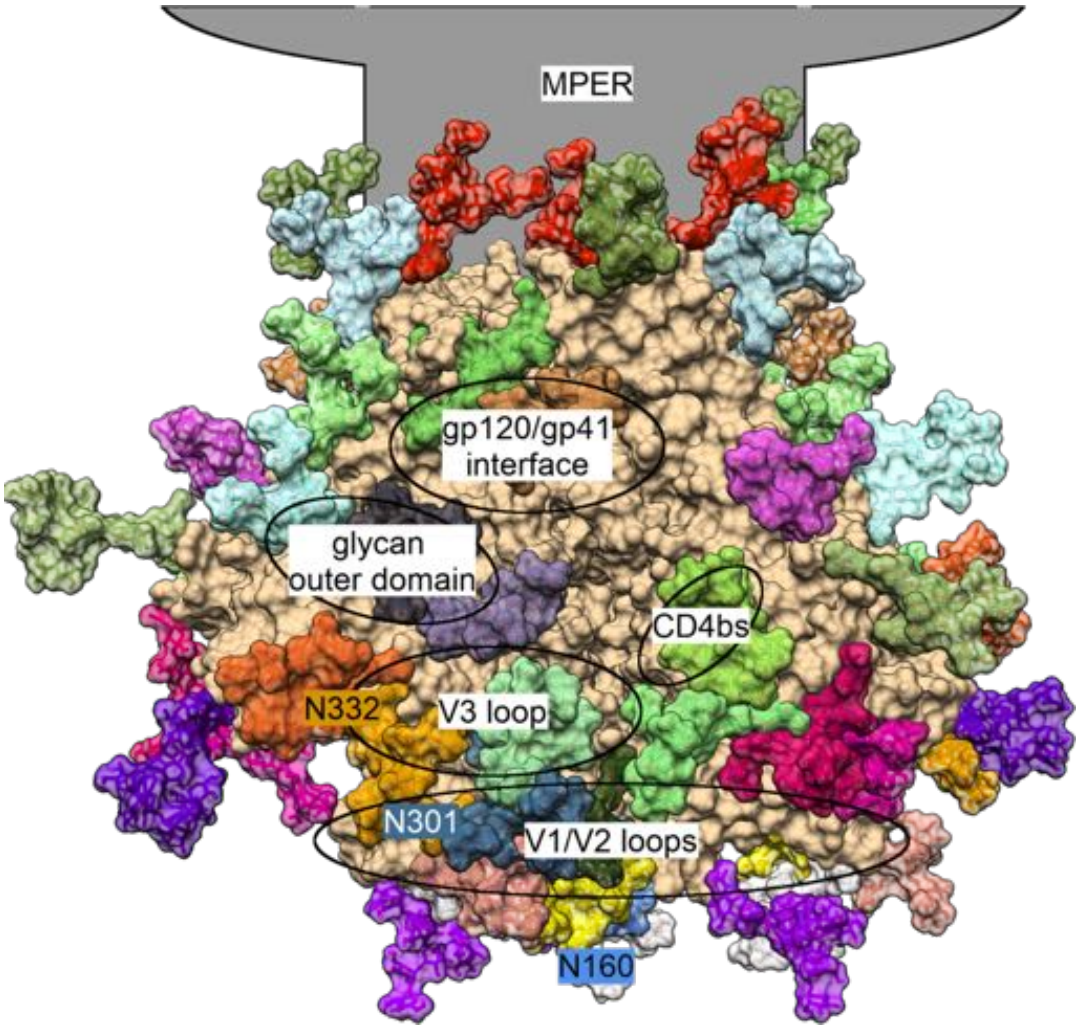


Images: 10.1016/j.atg.2013.05.004; https://en.wikipedia.org/wiki/Mutation#/media/File:Benzopyrene_DNA_adduct_1JDG.png

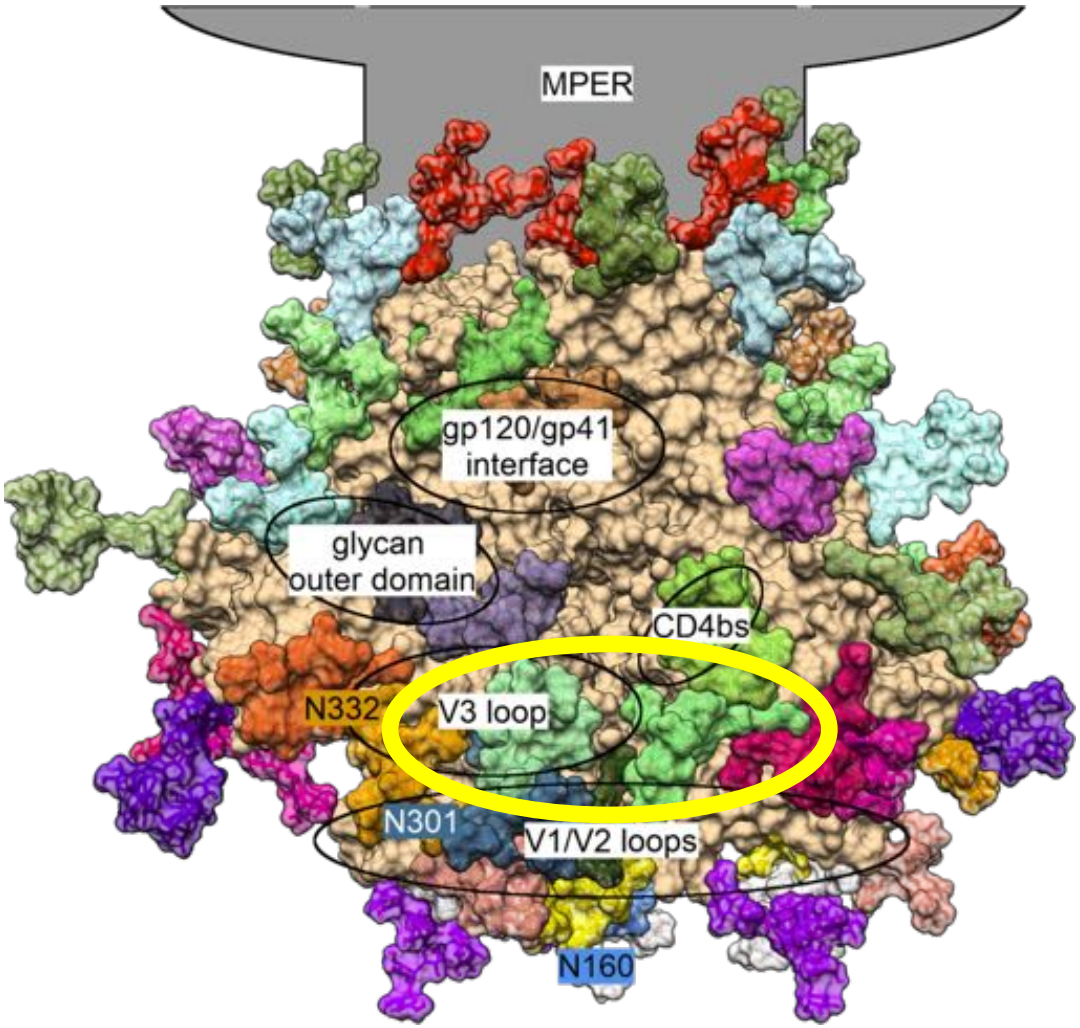
HIV-1 Env Trimer



HIV-1 Env Glycosylated Trimer

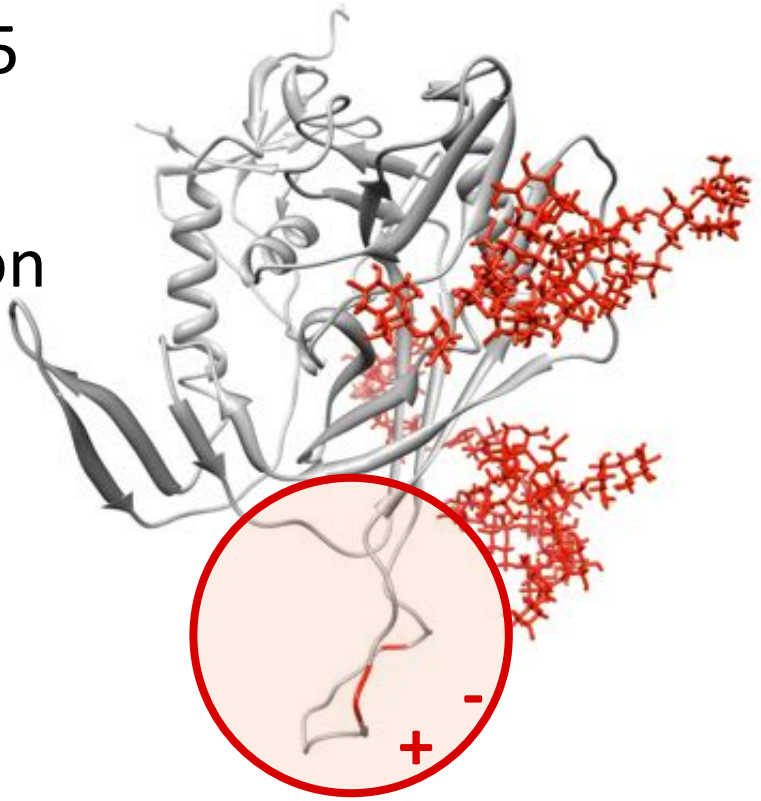


HIV-1 Env: V3 loop



Chemokine Receptors and the V3 loop

- Positions 11 / 24 / 25
- Net Charge
- N-linked Glycosylation



Chemokine Receptors and the V3 loop

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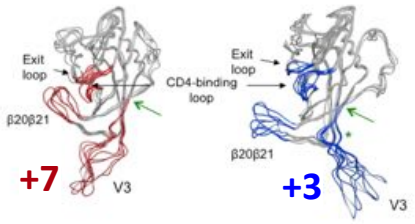
Structural Dynamics of HIV-1 Envelope Gp120 Outer Domain with V3 Loop

Masaru Yokoyama[1]*, Satoshi Naganawa[2], Kazuhisa Yoshimura[3], Shuzo Matsushita[3], Hironori Sato[1]*

1 Laboratory of Viral Genomics, Pathogen Genomics Center, National Institute of Infectious Diseases, 4-7-1 Gakuen, Musashi Murayama-shi, Tokyo, Japan, **2** Department of Microbiology and Cell Biology, Tokyo Metropolitan Institute of Medical Science, 2-1-6 Kamikitazawa, Setagaya-ku, Tokyo, Japan, **3** Division of Clinical Retrovirology and Infectious Diseases, Center for AIDS Research, Kumamoto University, 2-2-1 Honjo, Kumamoto, Japan



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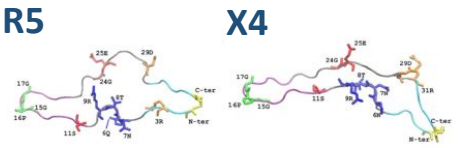
Insights into the Structure, Correlated Motions, and Electrostatic Properties of Two HIV-1 gp120 V3 Loops

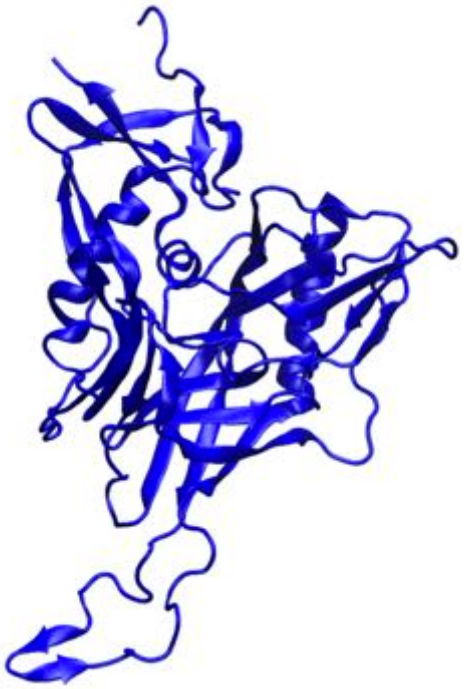
Aliana López de Victoria, Phanourios Tamamis, Chris A. Kieslich, Dimitrios Morikis*

Department of Bioengineering, University of California Riverside, Riverside, California, United States of America

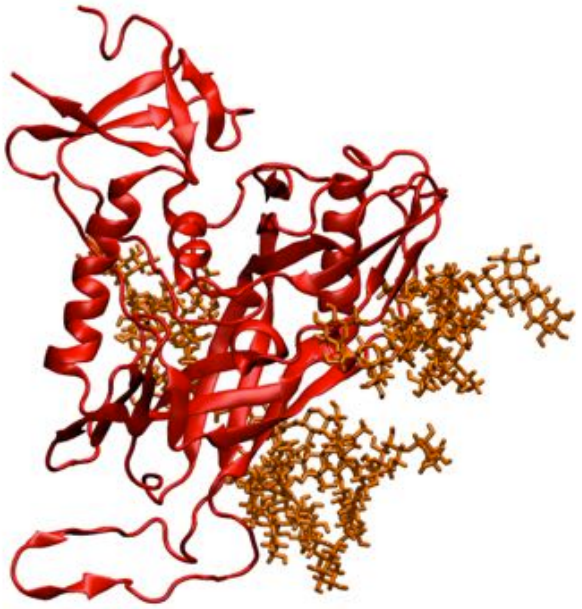


HIV-1 gp120 structures (pdb: 2B4C)

Non-Glycosylated

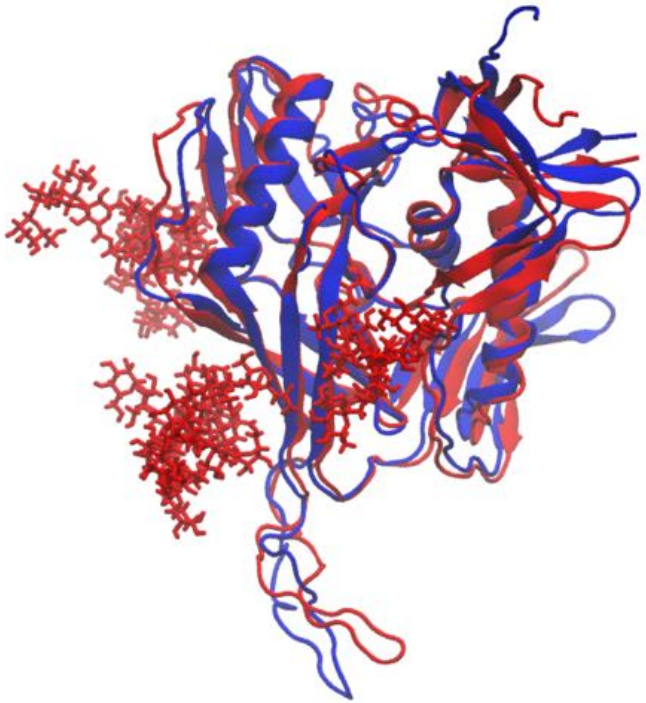


Glycosylated*5-glycans*



Non-Glycosylated and Glycosylated

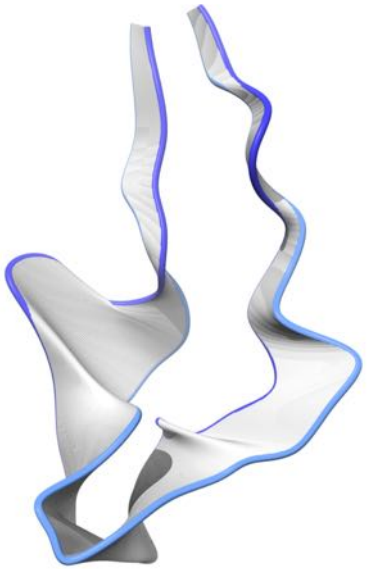
- MD Simulation, 30ns



V3-loop Movement

- Principle Component Analysis

Non-Glycosylated



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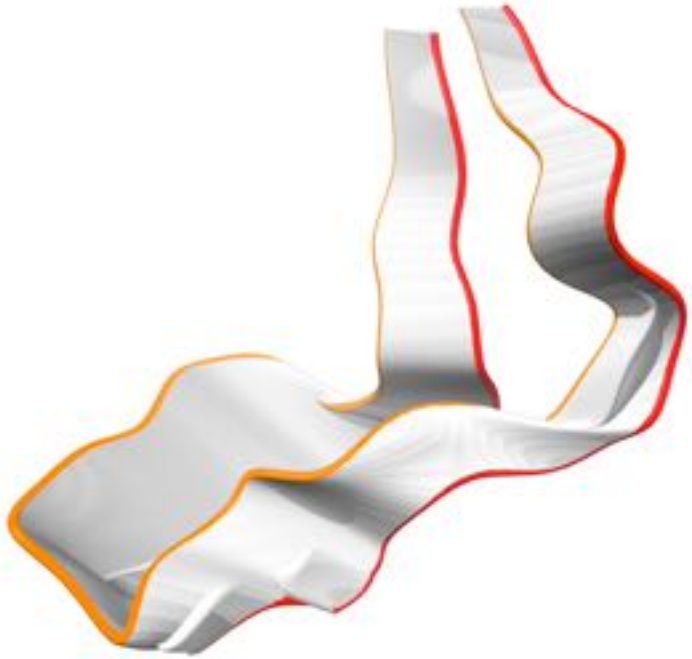

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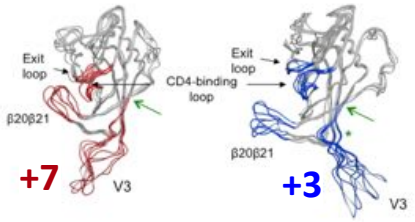

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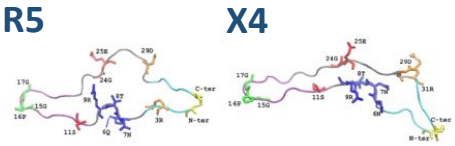

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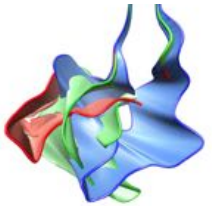
PLOS | ONE

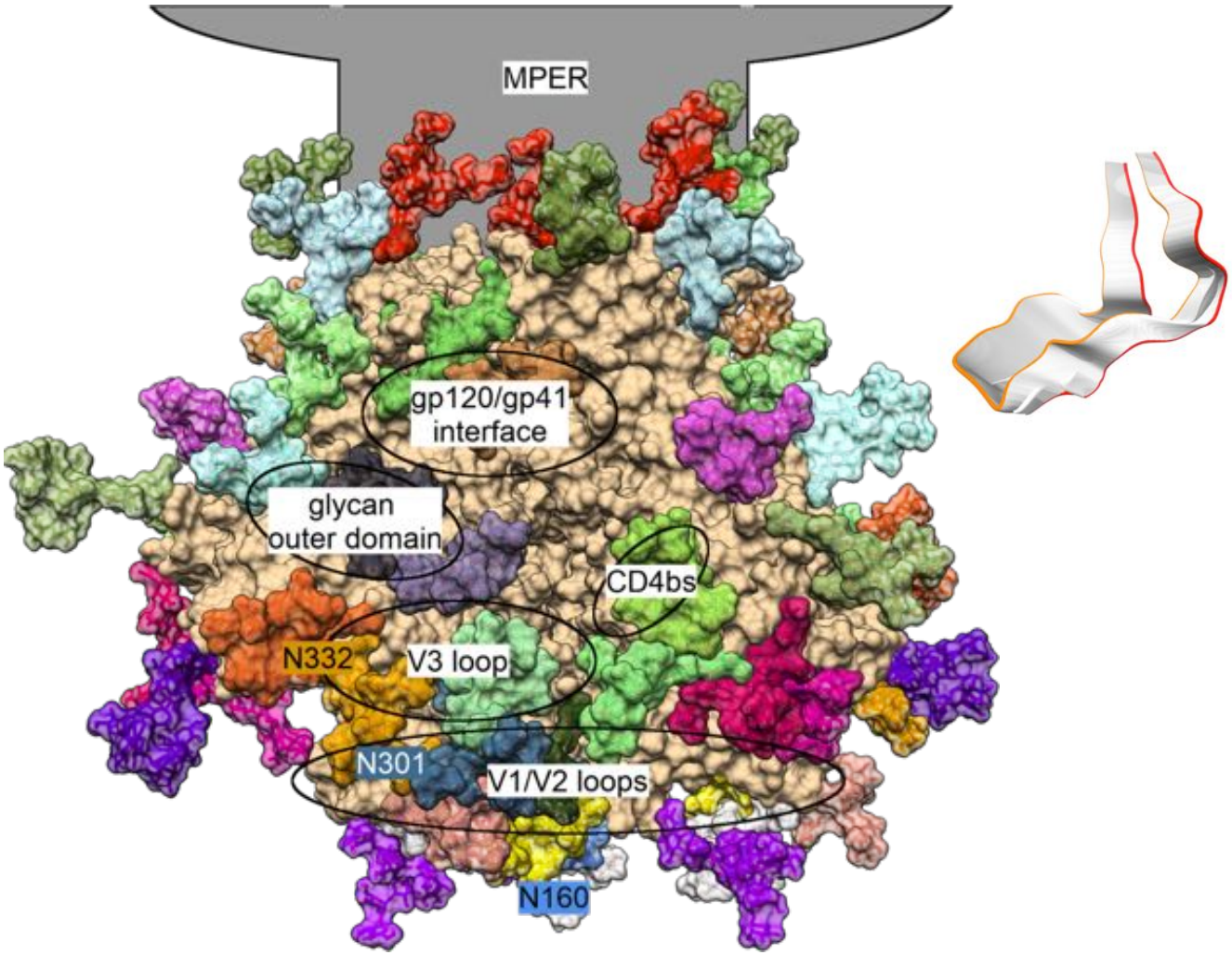
The Influence of N-Linked Glycans on the Molecular Dynamics of the HIV-1 gp120 V3 Loop

Natasha T. Wood[1], Elisa Fadda[2], Robert Davis[3], Oliver C. Grant[4], Joanne C. Martin[4], Robert J. Woods[3,4], Simon A. Travers[1]*

1 South African National Bioinformatics Institute, South African Medical Research Council Bioinformatics Unit, University of the Western Cape, Cape Town, South Africa, **2** Department of Chemistry, National University of Ireland, Maynooth, Maynooth, Ireland, **3** Complex Carbohydrate Research Centre, University of Georgia, Athens, Georgia, United States of America, **4** School of Chemistry, National University of Ireland, Galway, Galway, Ireland



HIV-1 Env Glycosylated Trimer



Glycosylation

Potential N-linked glycosylation sites (PNGS)

-----NXT|S-----

25-30 per monomer

Glycosylation

PLOS | COMPUTATIONAL BIOLOGY

RESEARCH ARTICLE

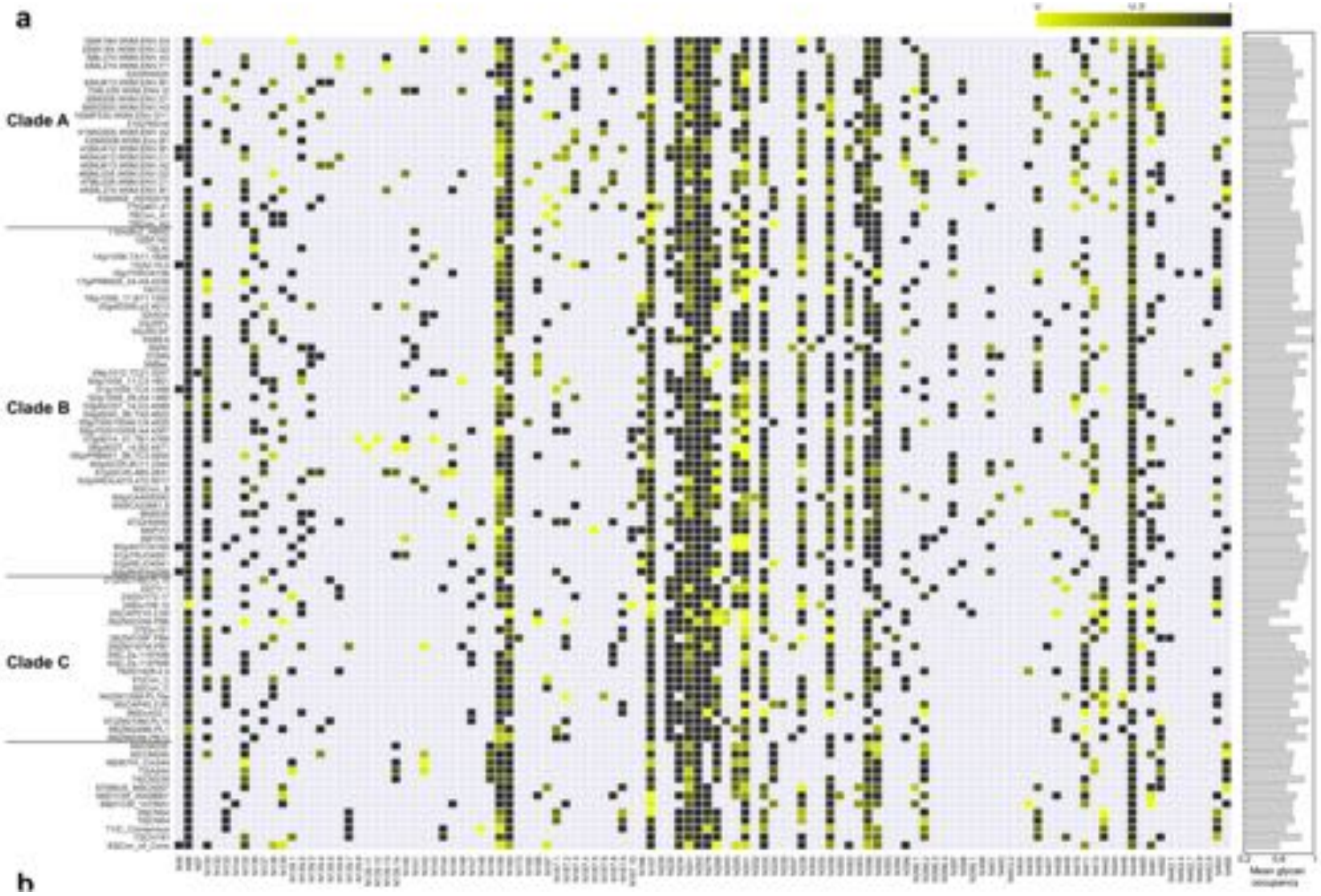
Exploiting glycan topography for computational design of Env glycoprotein antigenicity

Wen-Han Yu[1,2☯], Peng Zhao[3☯], Monia Draghi[1☯], Claudia Arevalo[1], Christina B. Karsten[1], Todd J. Suscovich[1], Bronwyn Gunn[1], Hendrik Streeck[4], Abraham L. Brass[5], Michael Tiemeyer[3], Michael Seaman[6], John R. Mascola[7], Lance Wells[3]*, Douglas A. Lauffenburger[1,2]*, Galit Alter[1]*

1 Ragon Institute of Massachusetts General Hospital, Massachusetts Institute of Technology and Harvard University, Cambridge, MA, United States of America, **2** Department of Biological Engineering, Massachusetts Institute of Technology, Cambridge, MA, United States of America, **3** Complex Carbohydrate Research Center, Department of Biochemistry and Molecular Biology, The University of Georgia, Athens, Georgia, United States of America, **4** Institute for HIV Research, University Hospital Essen, University Duisburg-Essen, Essen, Germany, **5** Department of Microbiology and Physiological Systems, University of Massachusetts Medical School, Worcester, MA, United States of America, **6** Beth Israel Deaconess Medical Center, Boston, Massachusetts, United States of America, **7** Vaccine Research Center, National Institute of Allergy and Infectious Diseases, National Institutes of Health, Bethesda, MD, United States of America

☯ These authors contributed equally to this work.
* galter@mgh.harvard.edu (GA); lauffen@mit.edu (DAL); lwells@ccrc.uga.edu (LW)

Glycosylation

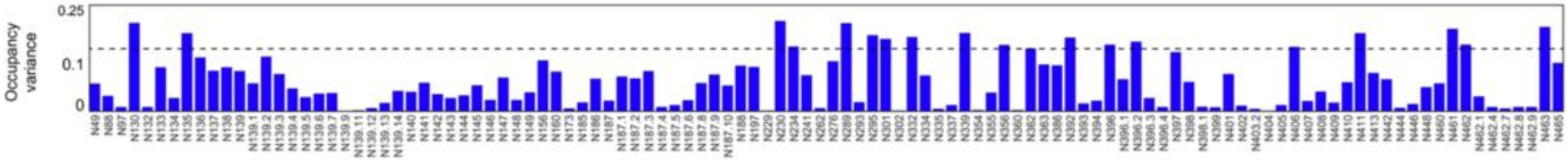


https://journals.plos.org/ploscompbiol/article?id=10.1371/journal.pcbi.1006093#sec020

Glycosylation

Potential N-linked glycosylation sites (PNGS)

-----NXT|S-----



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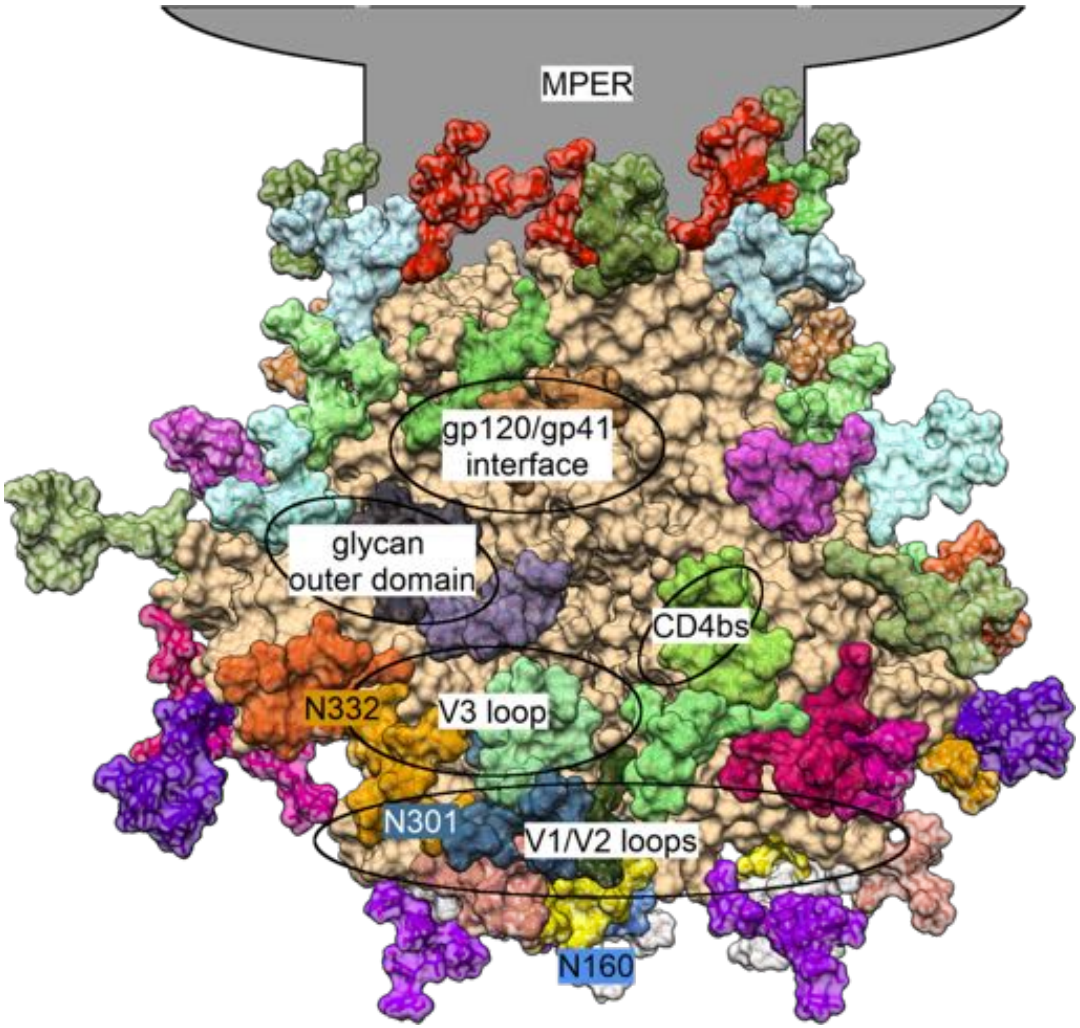
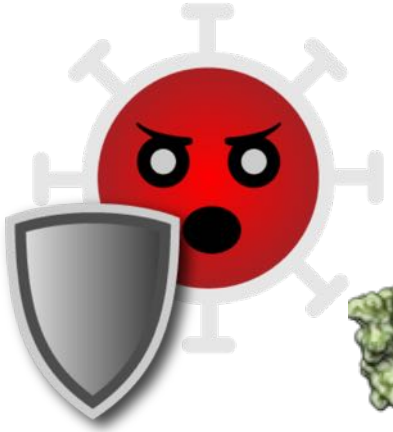
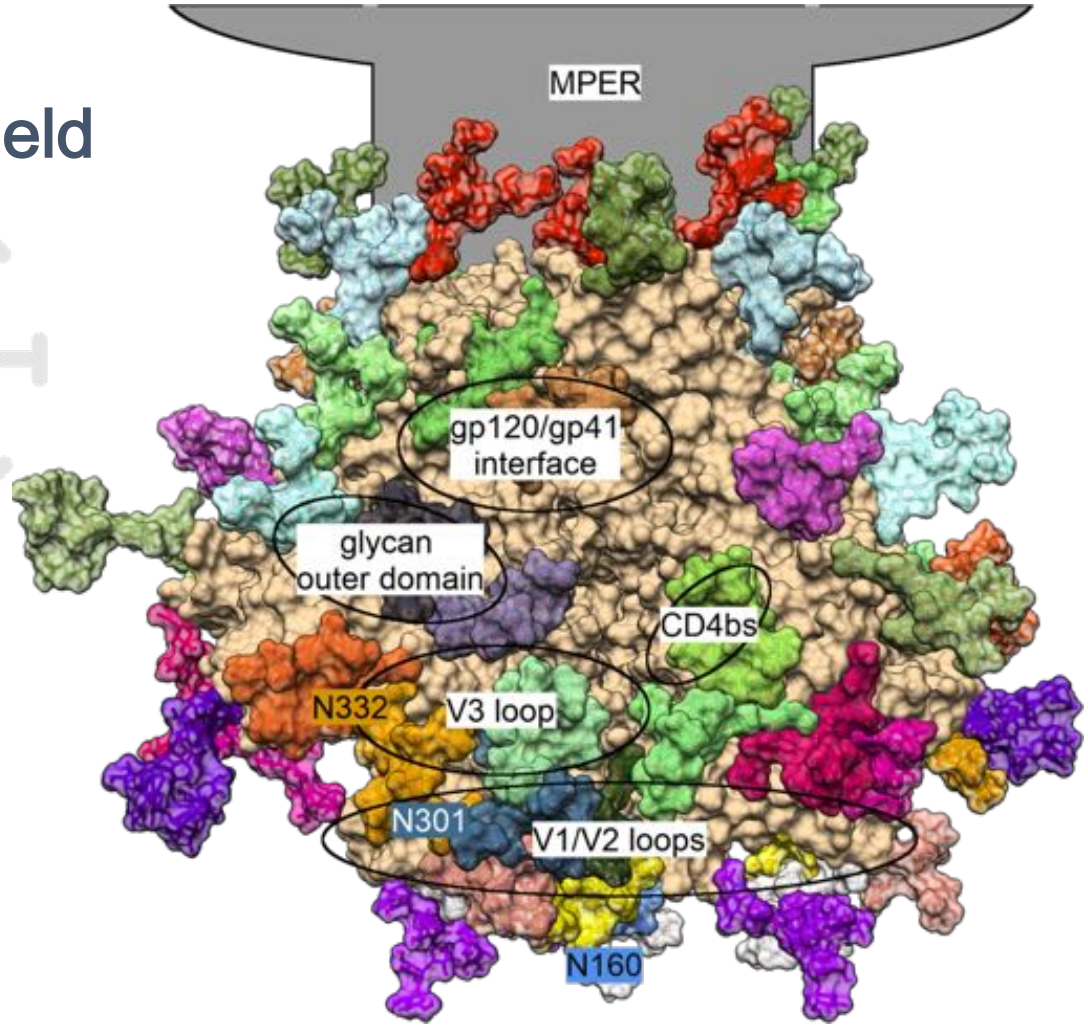


HIV-1 Env Glycosylated Trimer

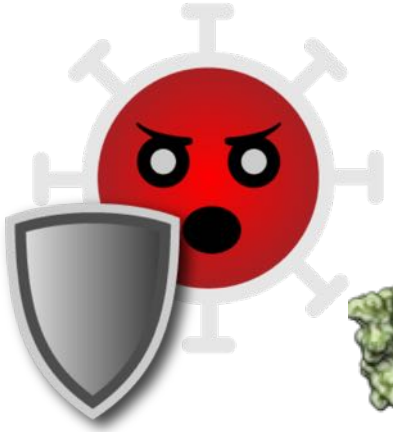
Glycan shield

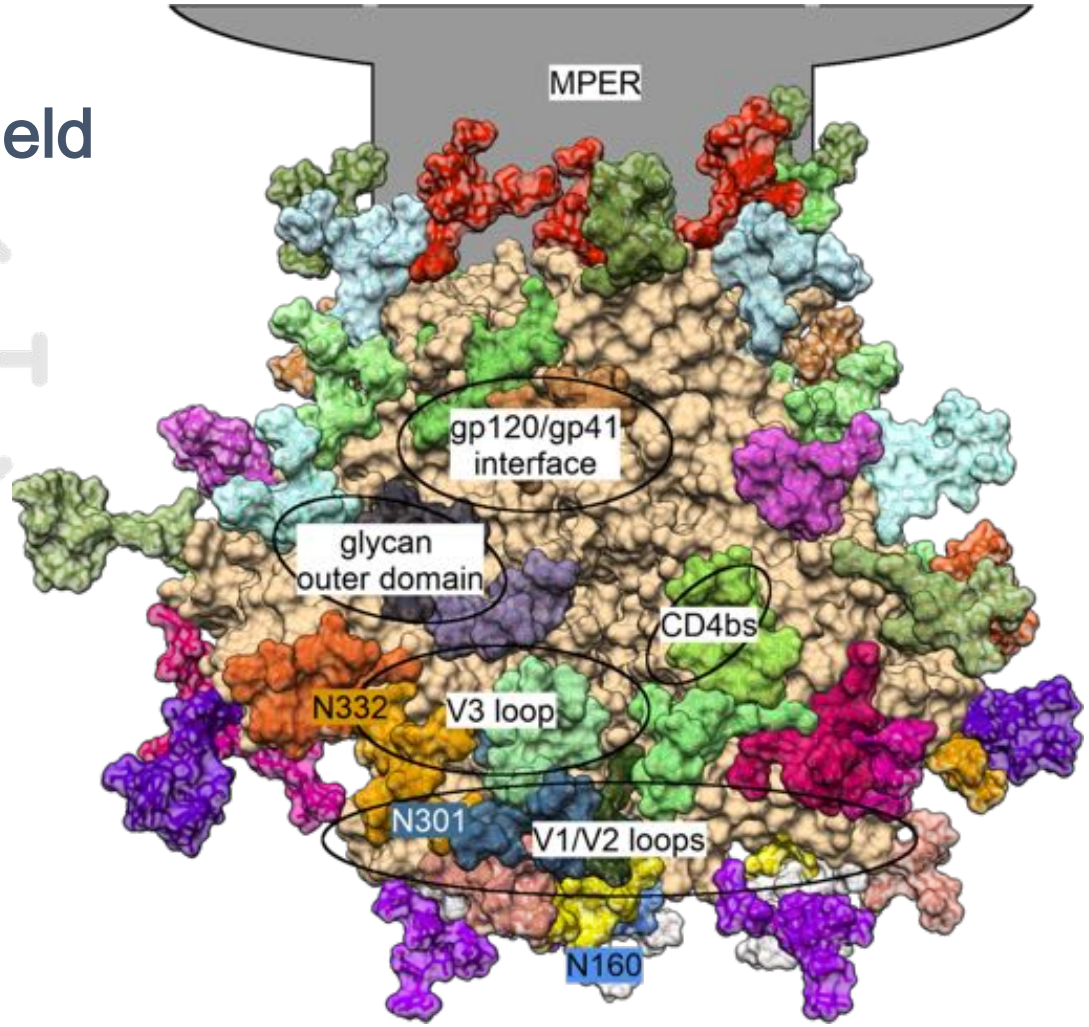


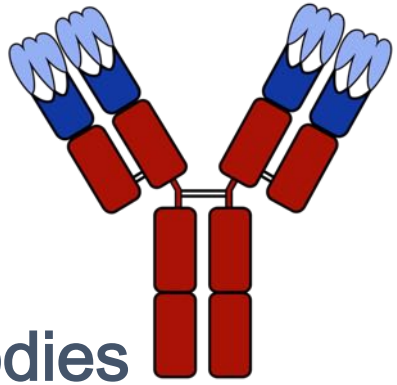


HIV-1 Env Glycosylated Trimer

Glycan shield



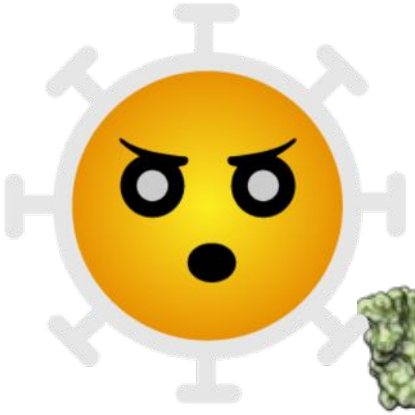


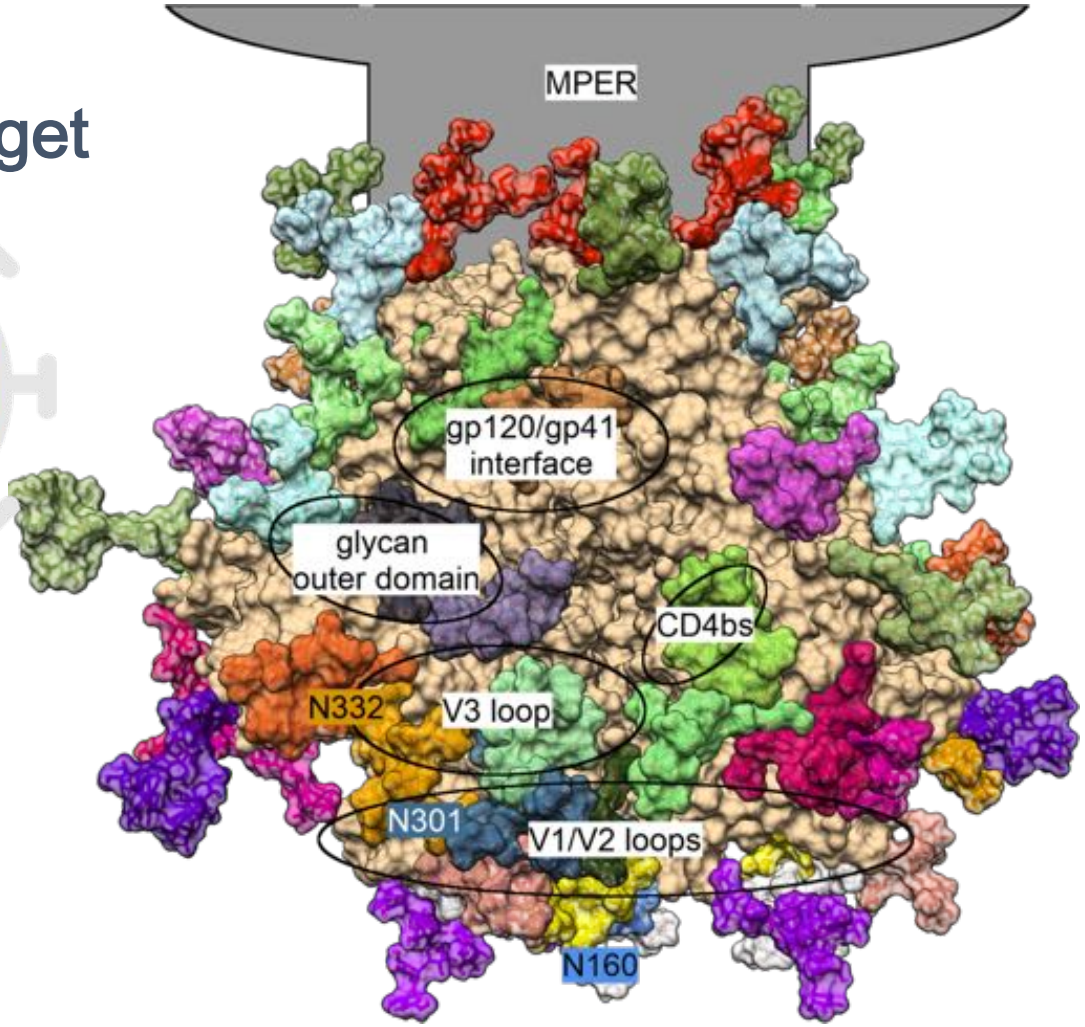


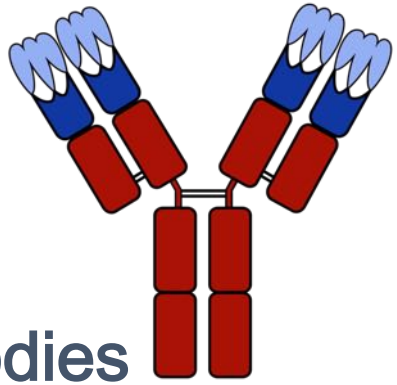
Neutralising antibodies

HIV-1 Env Glycosylated Trimer

Glycan target

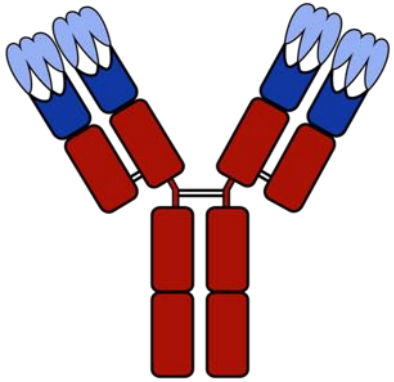






Neutralising antibodies

Neutralising antibodies: target sites



- Protein surface
- Protein surface and glycan/s
- Glycan/s

Neutralising antibodies: target sites

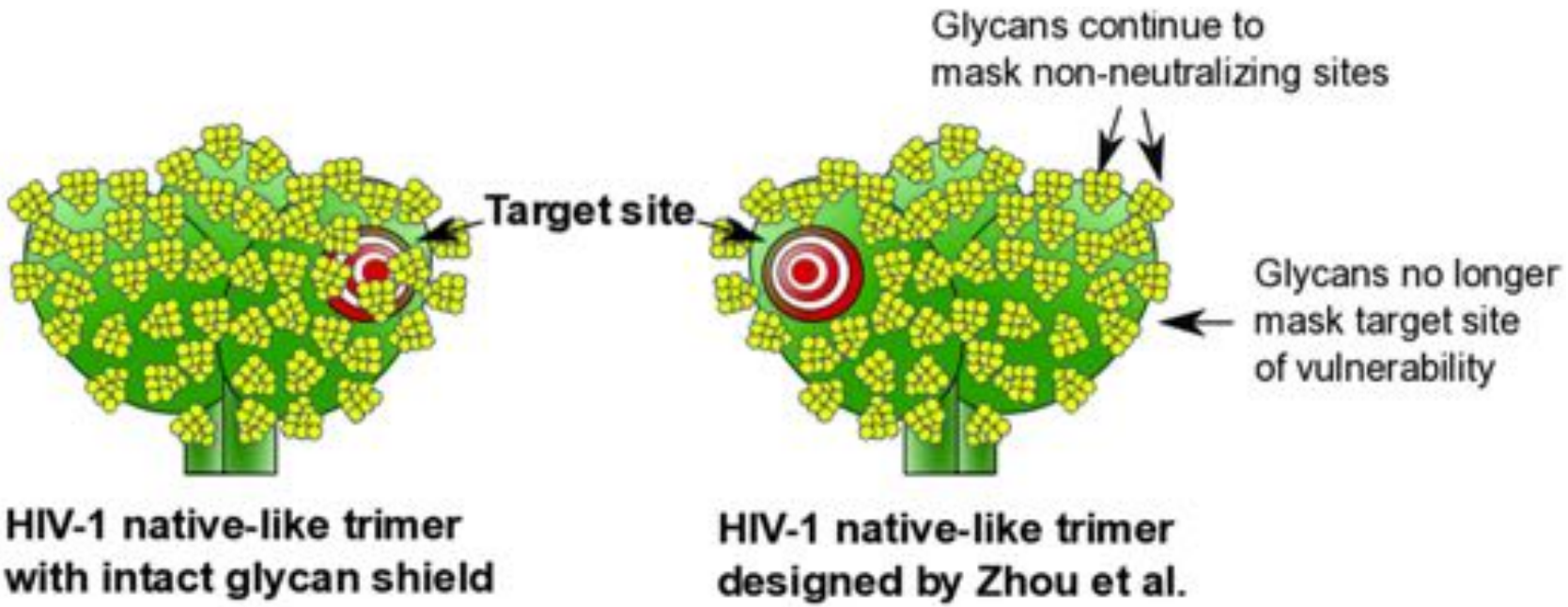
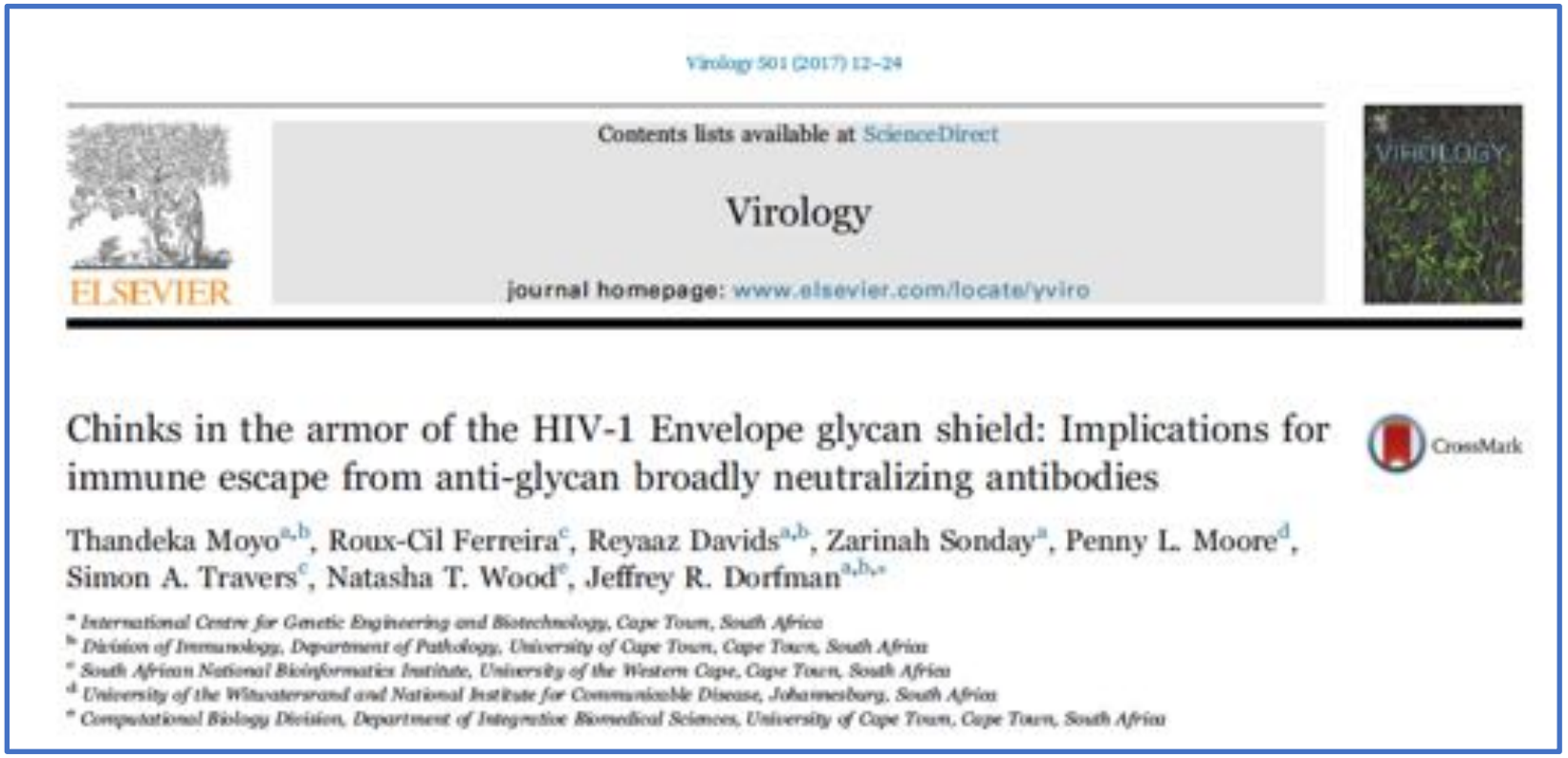


Image: https://www.sciencedirect.com/science/article/pii/S2211124717305272#fig1

HIV-1 Env and escape from bNAbs



Virology 501 (2017) 12–24

Contents lists available at ScienceDirect

Virology

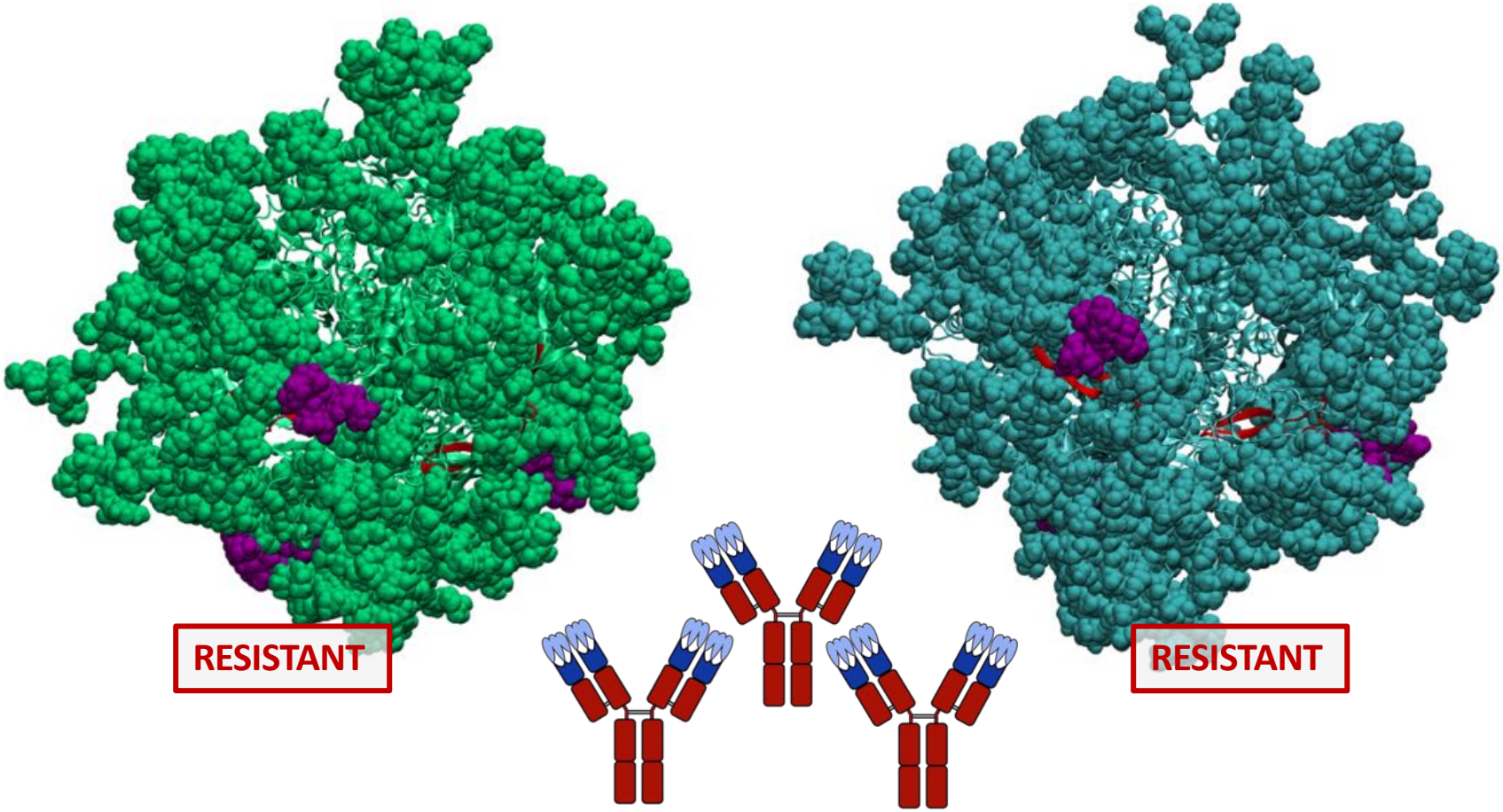
journal homepage: www.elsevier.com/locate/yviro

Chinks in the armor of the HIV-1 Envelope glycan shield: Implications for immune escape from anti-glycan broadly neutralizing antibodies

Thandeka Moyo[a,b], Roux-Cil Ferreira[c], Reyaaz Davids[a,b], Zarinah Sonday[a], Penny L. Moore[d], Simon A. Travers[c], Natasha T. Wood[e], Jeffrey R. Dorfman[a,b,*]

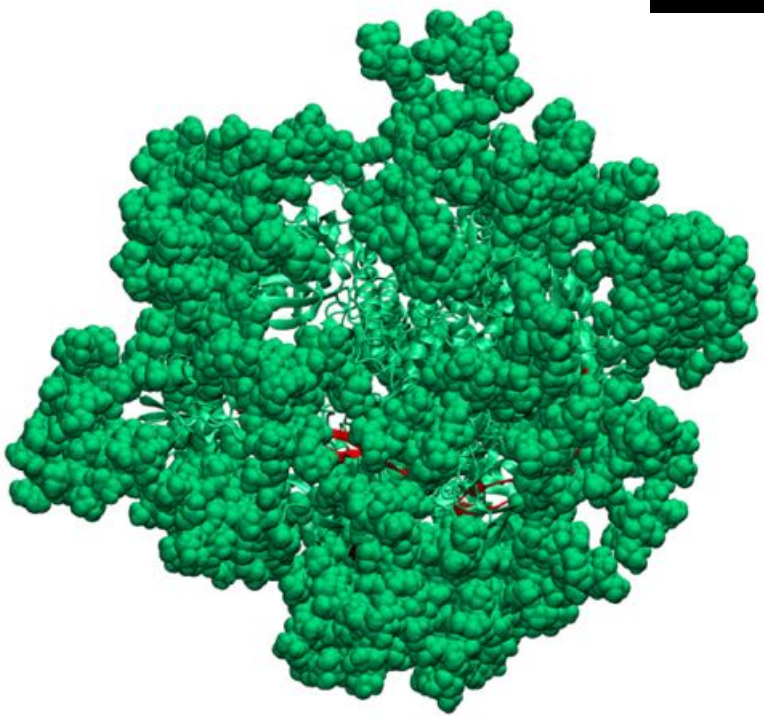
[a] International Centre for Genetic Engineering and Biotechnology, Cape Town, South Africa
[b] Division of Immunology, Department of Pathology, University of Cape Town, Cape Town, South Africa
[c] South African National Bioinformatics Institute, University of the Western Cape, Cape Town, South Africa
[d] University of the Witwatersrand and National Institute for Communicable Disease, Johannesburg, South Africa
[e] Computational Biology Division, Department of Integrative Biomedical Sciences, University of Cape Town, Cape Town, South Africa

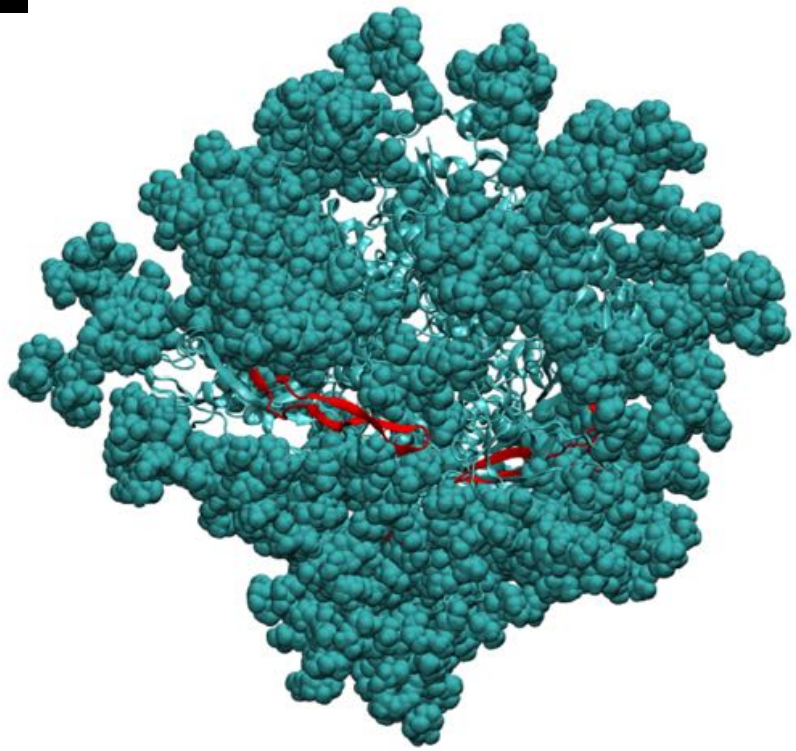
Glycosylated trimers



Glycosylated trimers

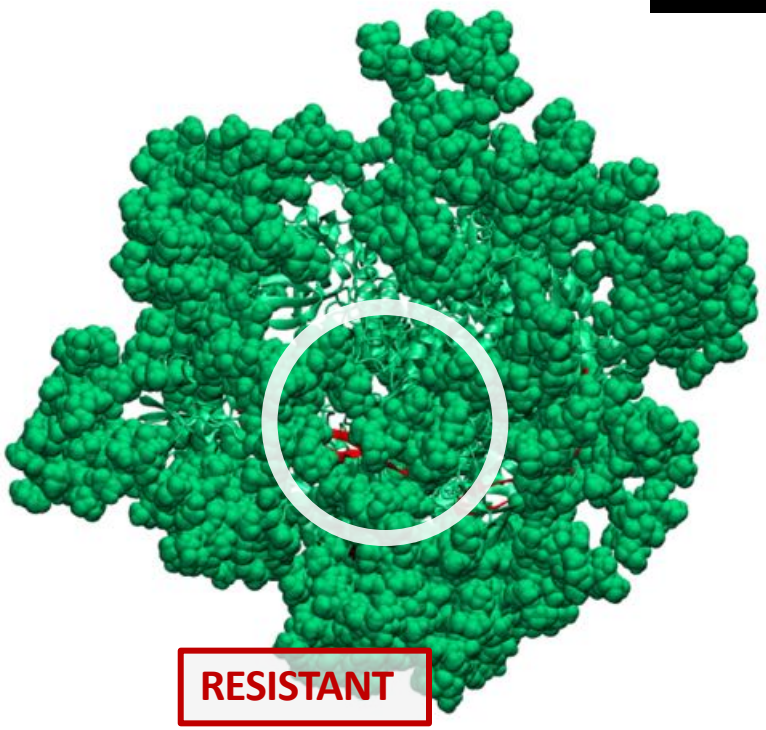
Remove N301



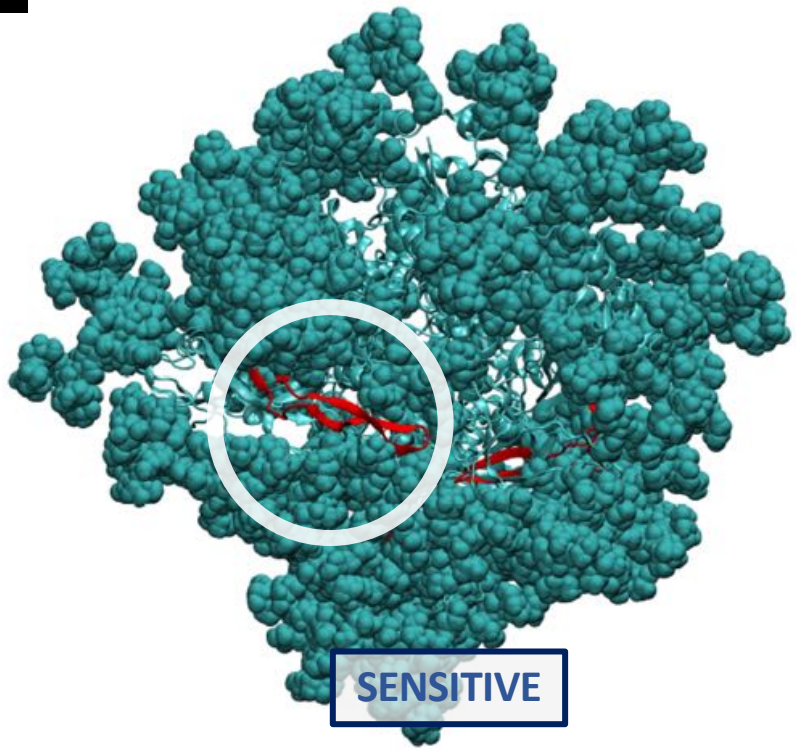


Glycosylated trimers

Remove
N301



RESISTANT



SENSITIVE

Background

CAP45

Du156

Remove N301

CAP45^{N301}

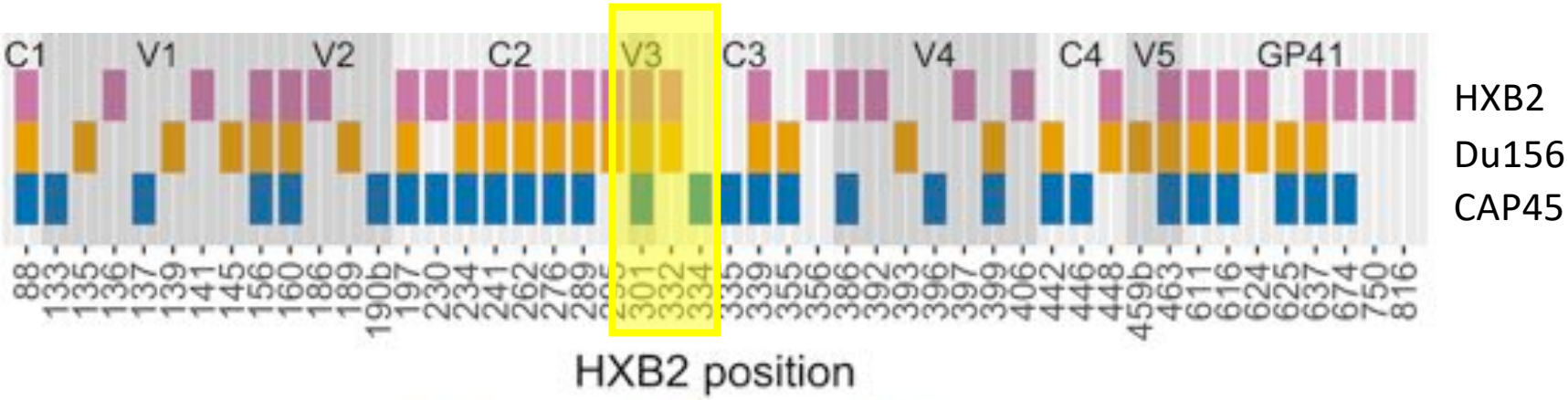
Du156^{N301}

Samples – PNGS comparison

N X S/T
(X any amino acid apart from Pro)

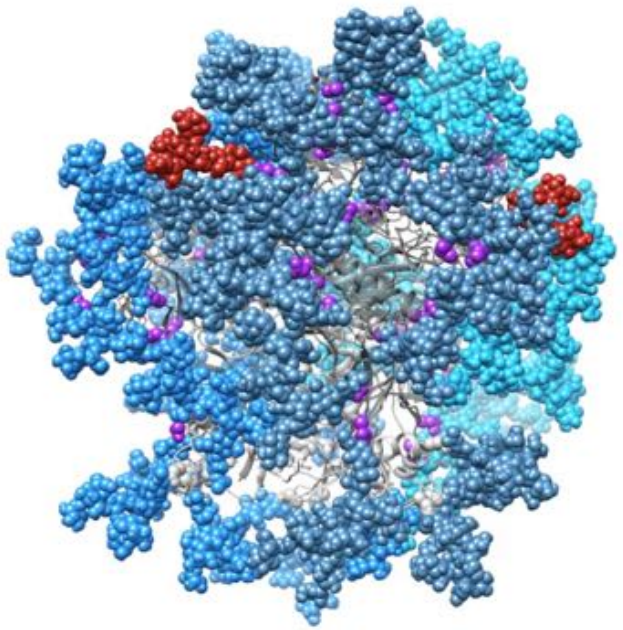
Samples – PNGS comparison

N X S/T
(X any amino acid apart from Pro)

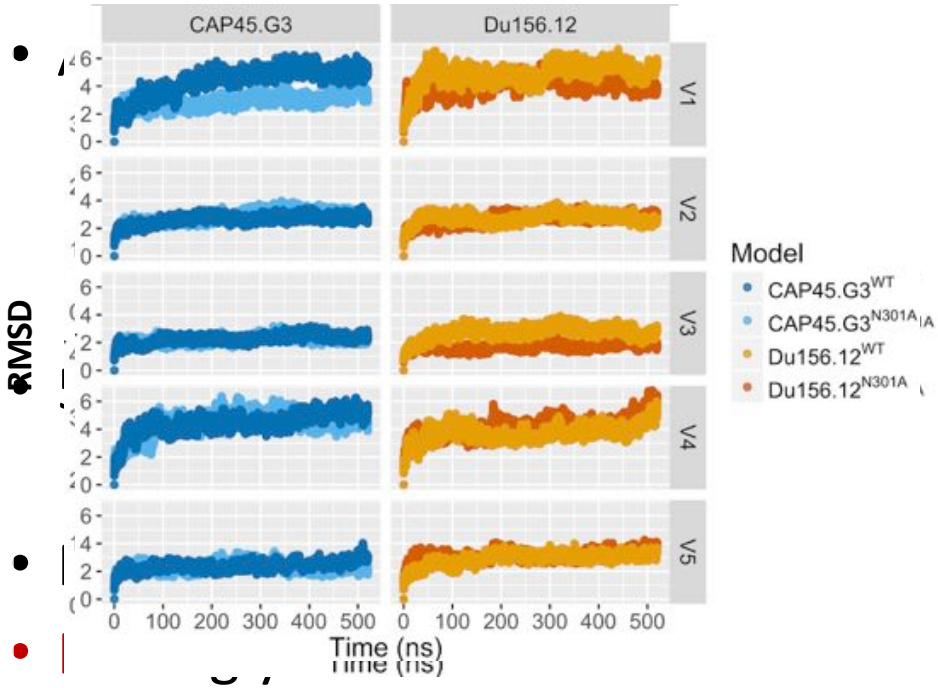


- 90% sequence identity
- 29 PNGS's

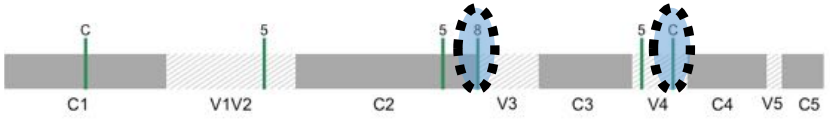
MD Simulation



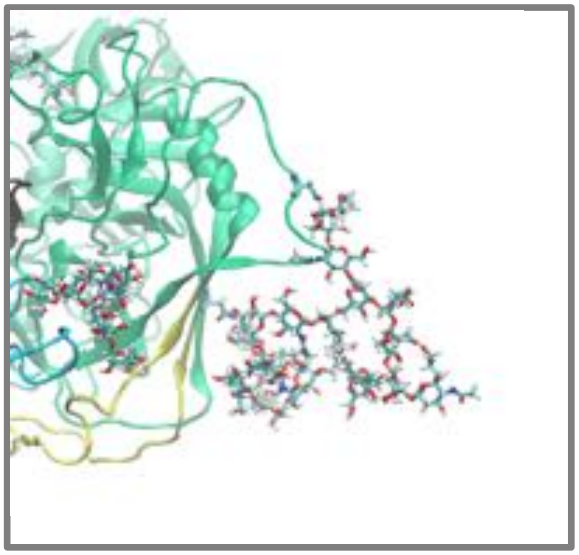
Root-Mean-Square Deviation

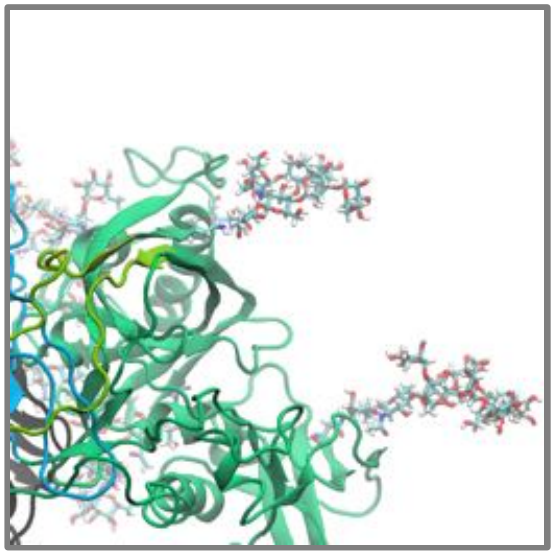


Inter-Glycan-Action

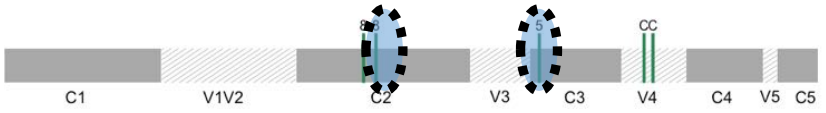


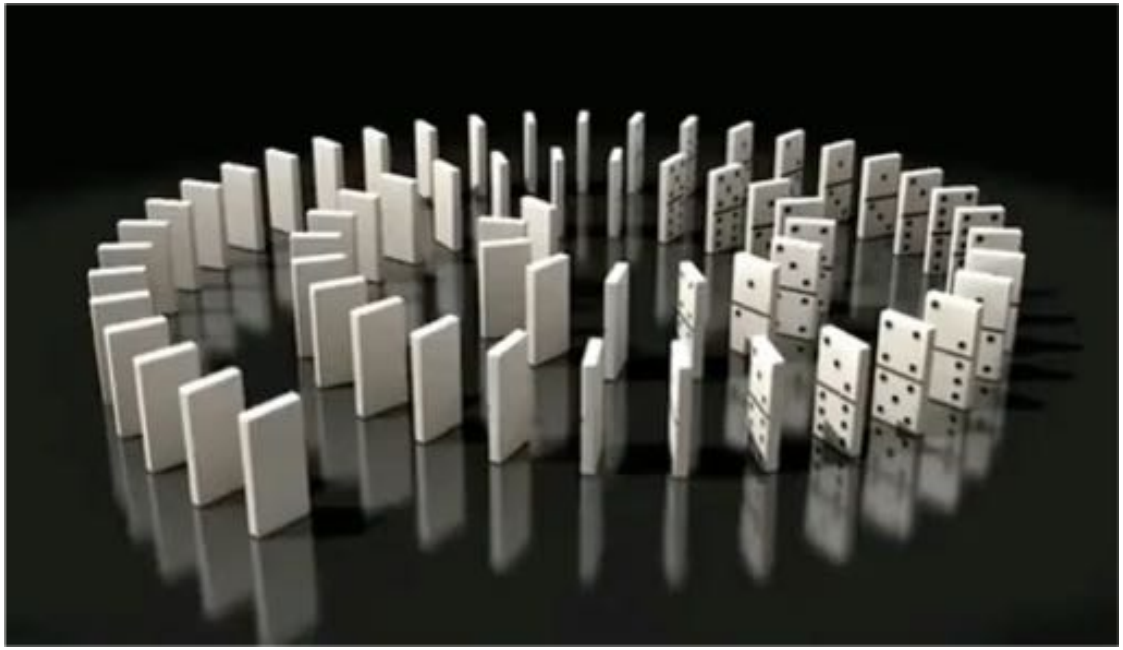
Subtype C

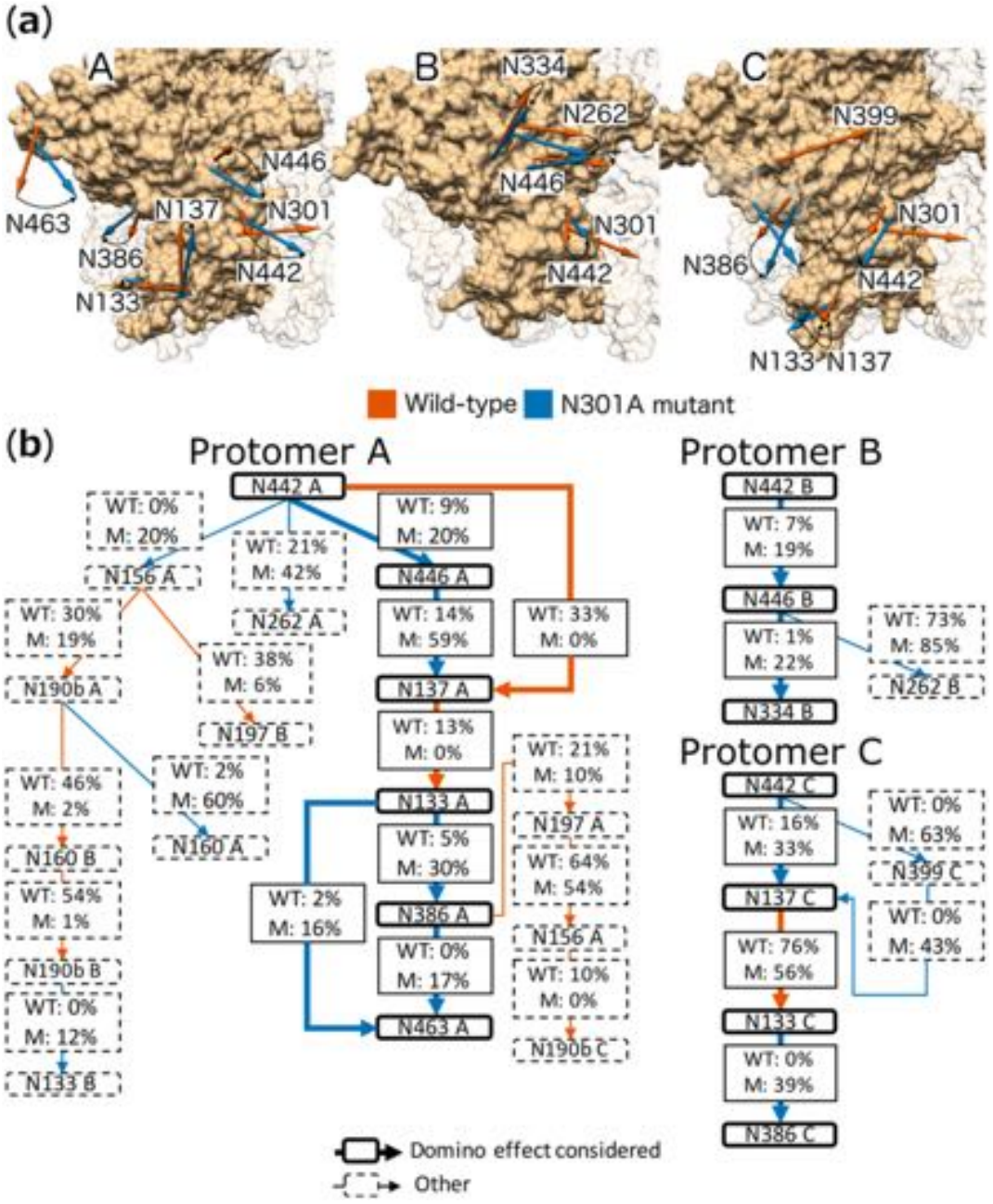




Subtype A






(a)
A
B
C
N334
N262
N399
N446
N463
N137
N301
N386
N442
N133
Wild-type
N301A mutant
(b)
Protomer A
Protomer B
Protomer C
N442 A
WT: 0%
M: 20%
N156 A
WT: 21%
M: 42%
N262 A
WT: 9%
M: 20%
N446 A
WT: 30%
M: 19%
N1906 A
WT: 38%
M: 6%
N197 B
WT: 14%
M: 59%
WT: 33%
M: 0%
N137 A
WT: 13%
M: 0%
WT: 46%
M: 2%
N160 B
WT: 2%
M: 60%
N160 A
WT: 21%
M: 10%
N197 A
WT: 5%
M: 30%
WT: 64%
M: 54%
WT: 2%
M: 16%
N386 A
N156 A
WT: 54%
M: 1%
N1906 B
WT: 0%
M: 17%
WT: 10%
M: 0%
WT: 0%
M: 12%
N463 A
N1906 C
N133 B
N442 B
WT: 7%
M: 19%
N446 B
WT: 1%
M: 22%
WT: 73%
M: 85%
N262 B
N334 B
N442 C
WT: 16%
M: 33%
WT: 0%
M: 63%
N399 C
N137 C
WT: 0%
M: 43%
WT: 76%
M: 56%
N133 C
WT: 0%
M: 39%
N386 C
Domino effect considered
Other

Neutralising antibodies: target sites

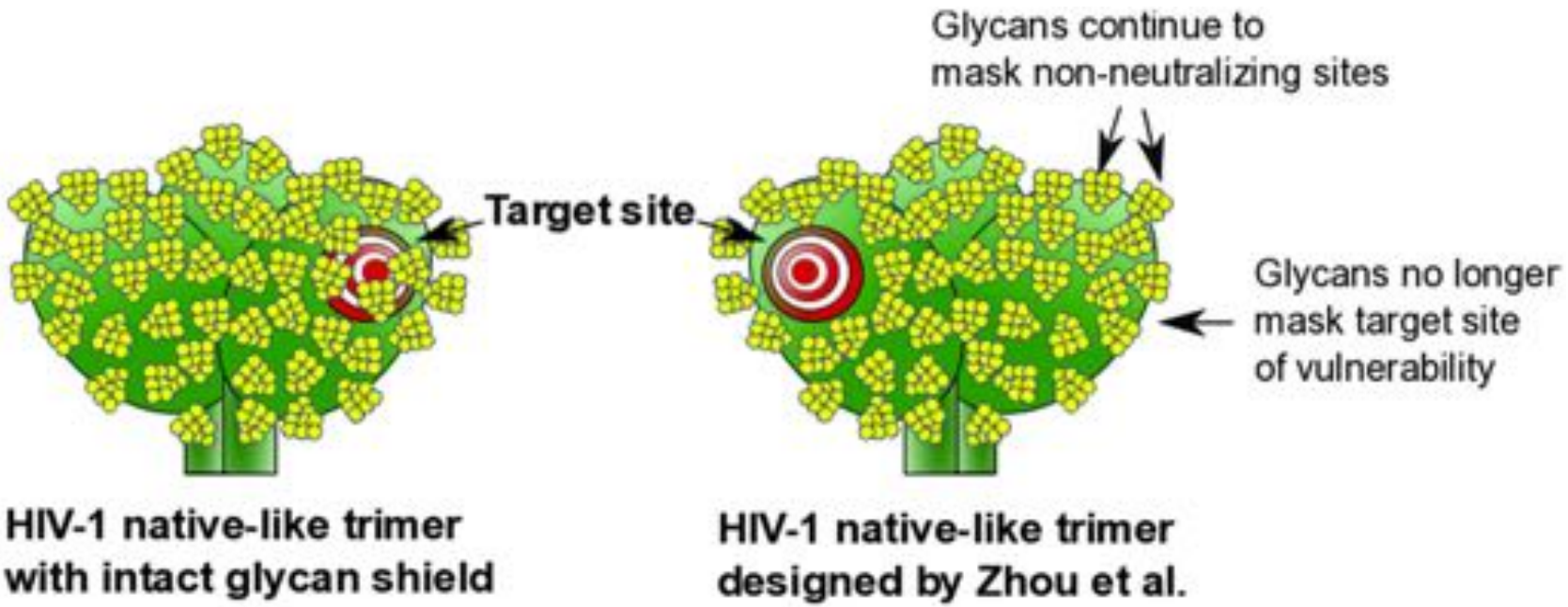
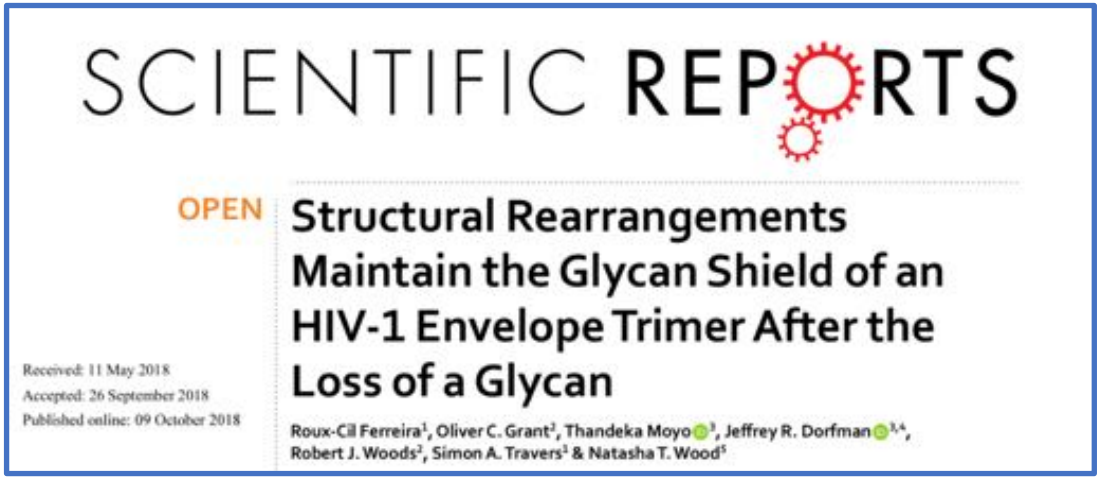


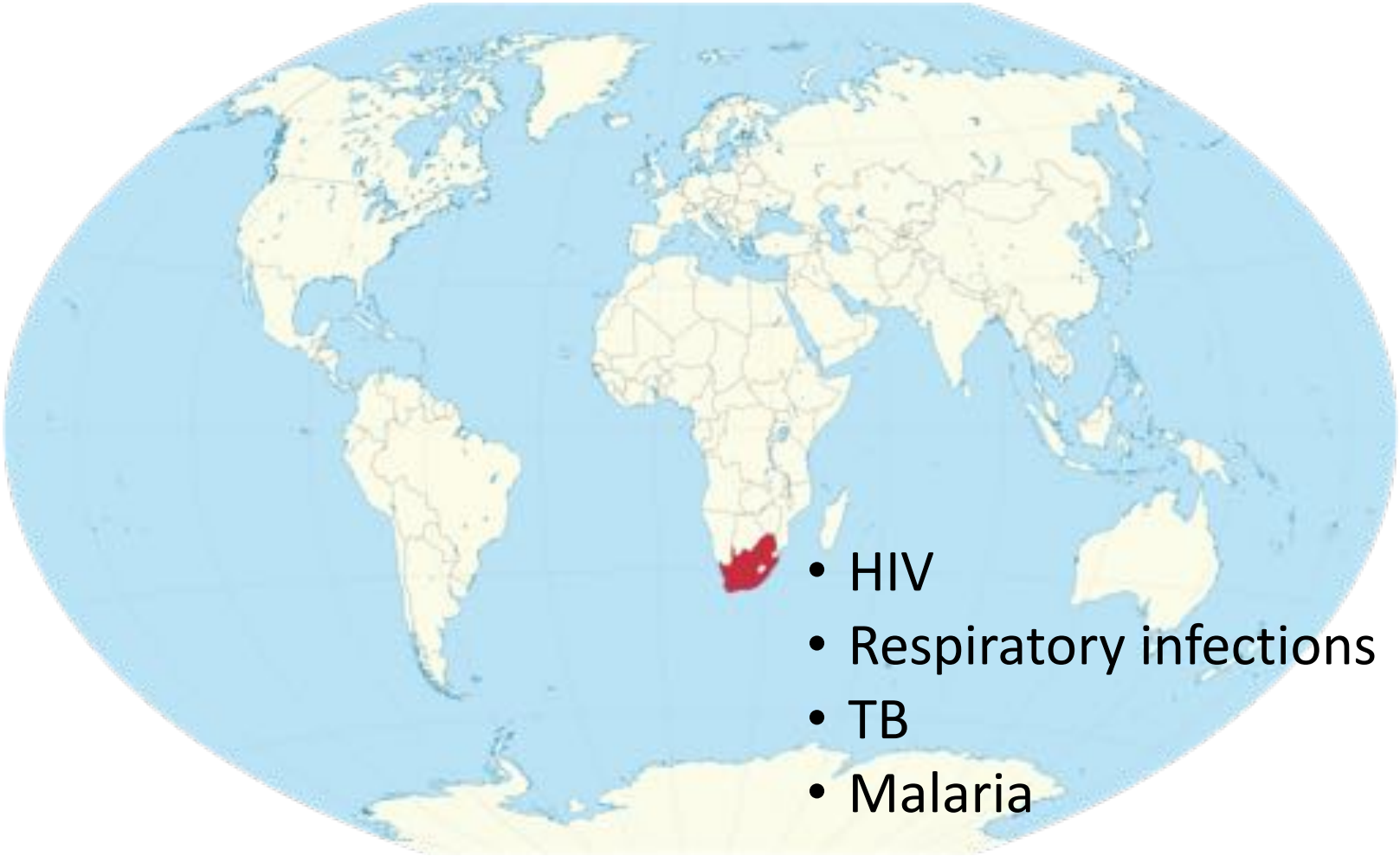
Image: https://www.sciencedirect.com/science/article/pii/S2211124717305272#fig1

Summary: MD

- Difficult to compare MD simulations
- By combining analysis methods, we can generate hypotheses for the different phenotypic observations
- The removal of a glycan can have a significant effect
- The impact of the removal of a glycan depends on the rest of the glycan landscape



South Africa



- HIV
- Respiratory infections
- TB
- Malaria

TB

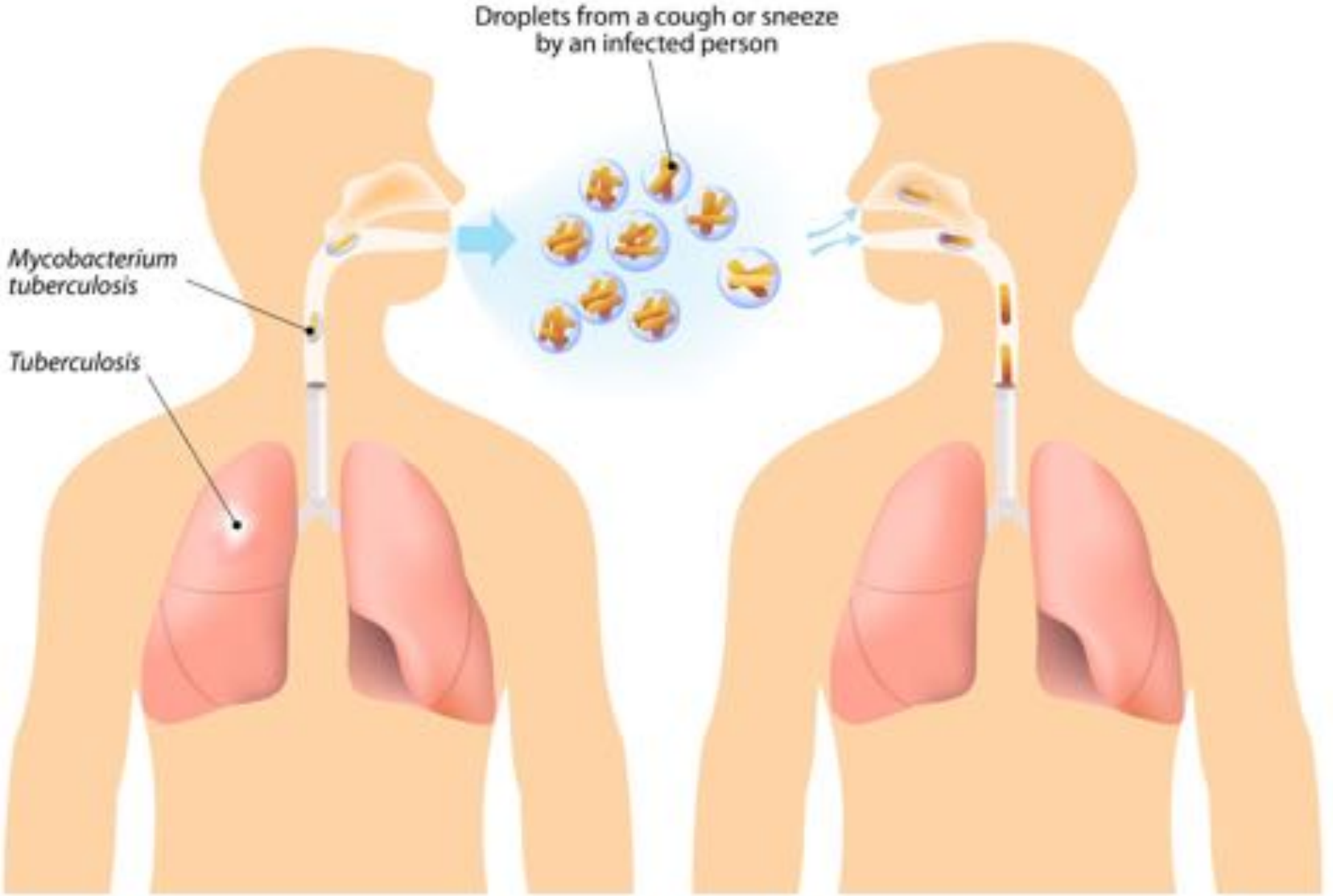
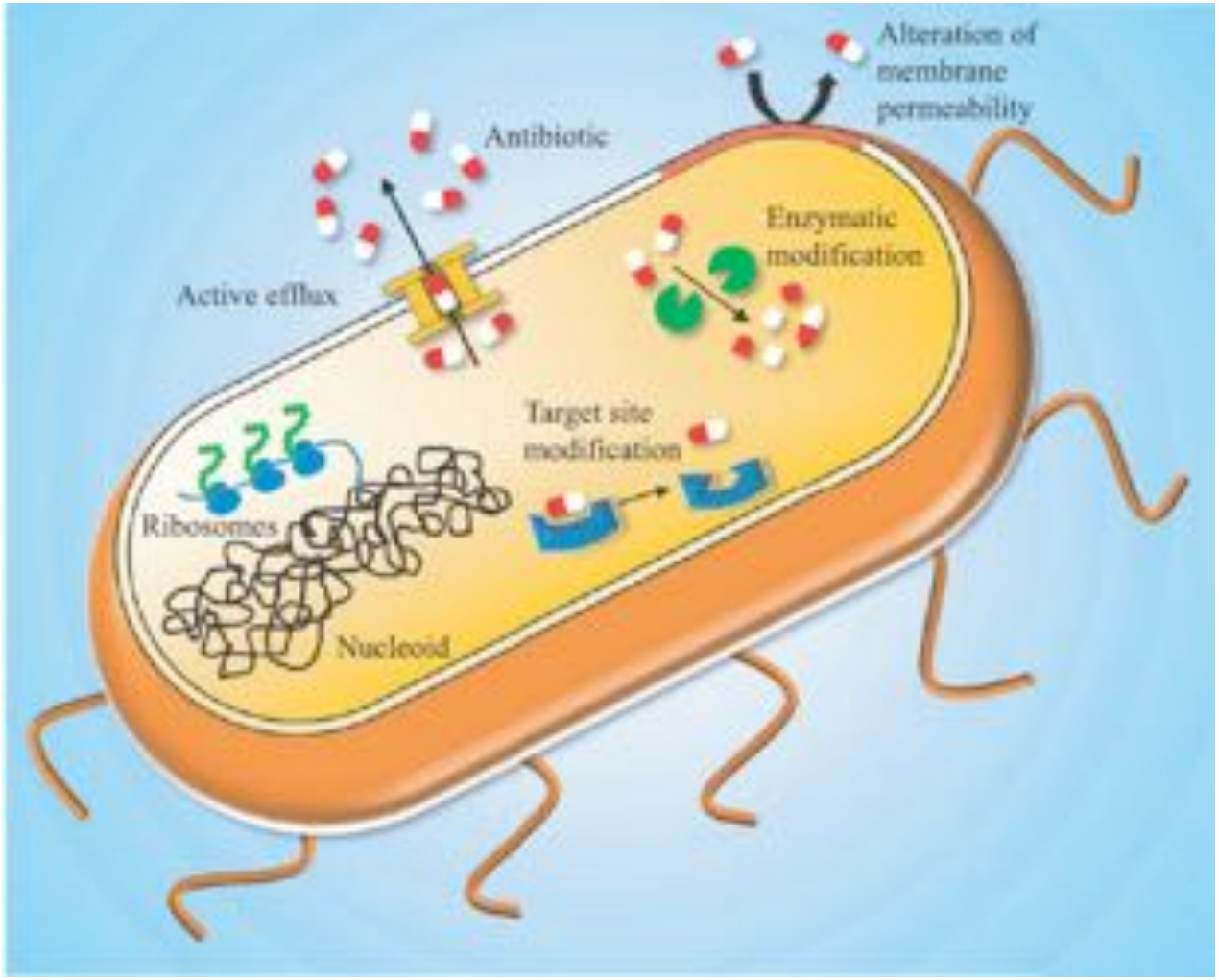


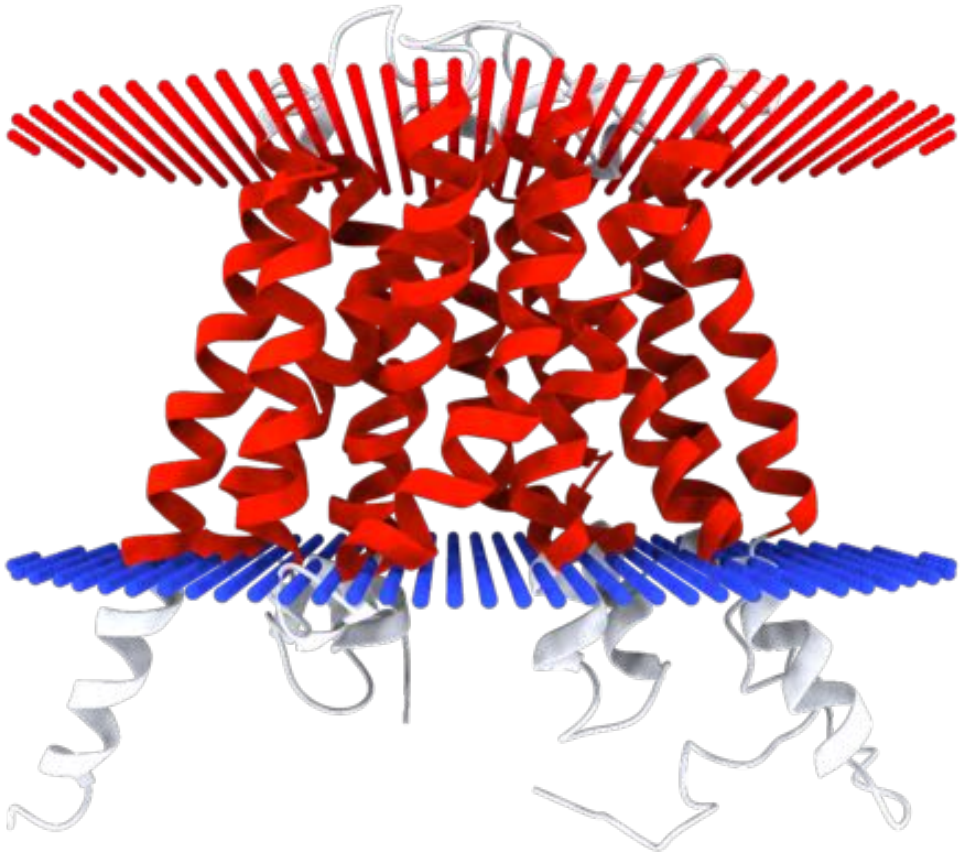
Image from: https://www.anokacounty.us/2935/Tuberculosis-TB

The four major mechanisms by which the bacterial cells develop multiple drug resistance



Sharma A, Gupta VK, Pathania R. Efflux pump inhibitors for bacterial pathogens: From bench to bedside. Indian J Med Res 2019;149:129-45

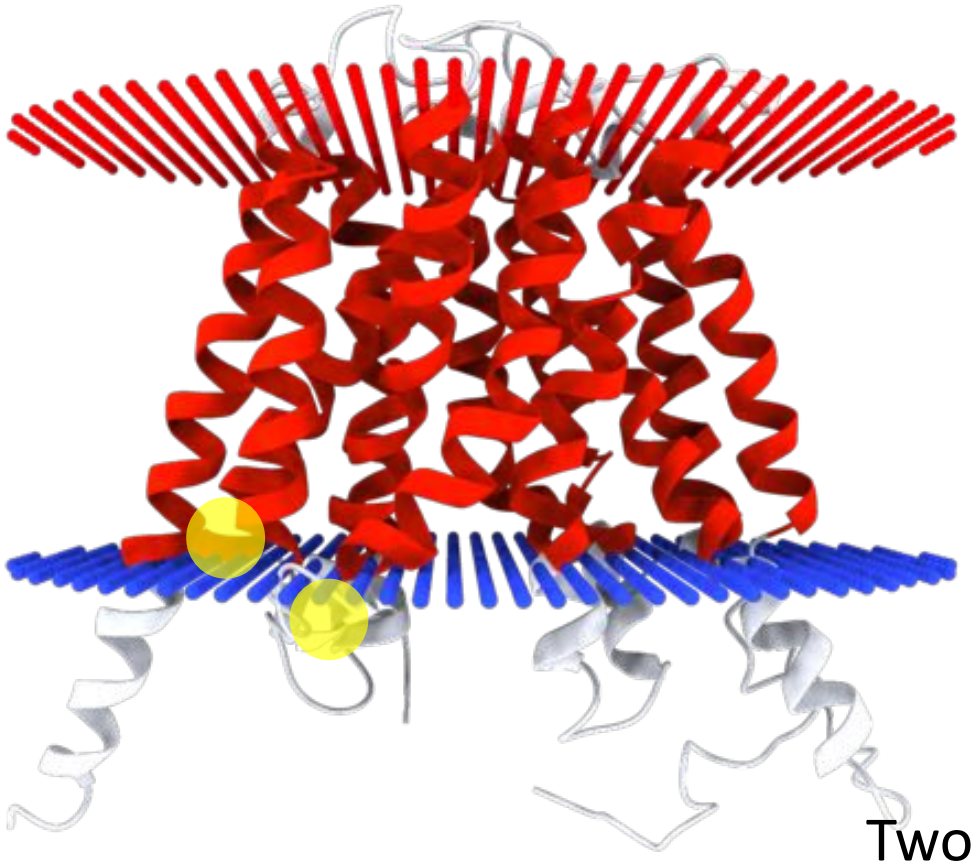
Efflux pumps



TAP (Rv1258c) efflux pump

Liu J, Shi W, Zhang S, Hao X, Maslov DA, Shur KV, Bekker OB, Danilenko VN and Zhang Y (2019) Mutations in Efflux Pump Rv1258c (Tap) Cause Resistance to Pyrazinamide, Isoniazid, and Streptomycin in M. tuberculosis. Front. Microbiol. 10:216. doi: 10.3389/fmicb.2019.00216

Efflux pumps

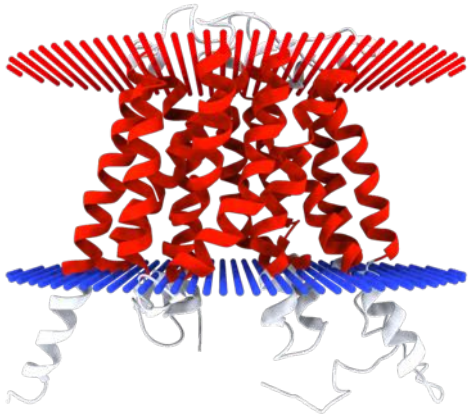


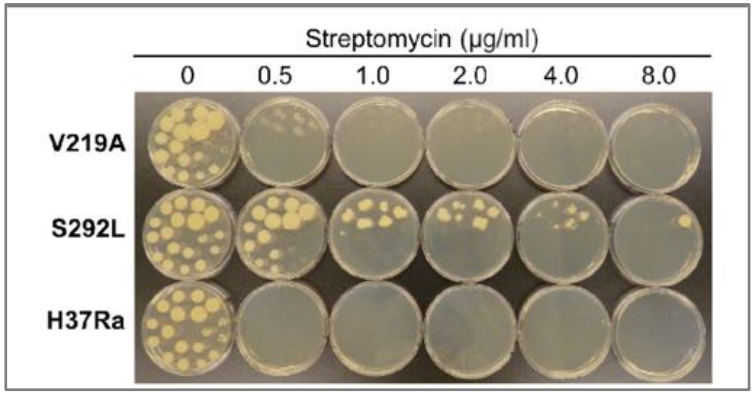
Two point mutations
V219A and S292L

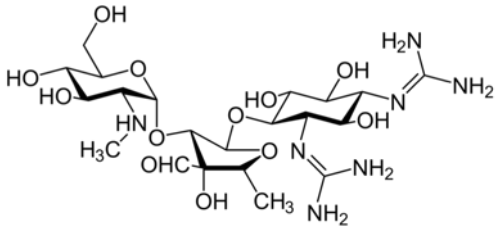
TAP (Rv1258c) efflux pump

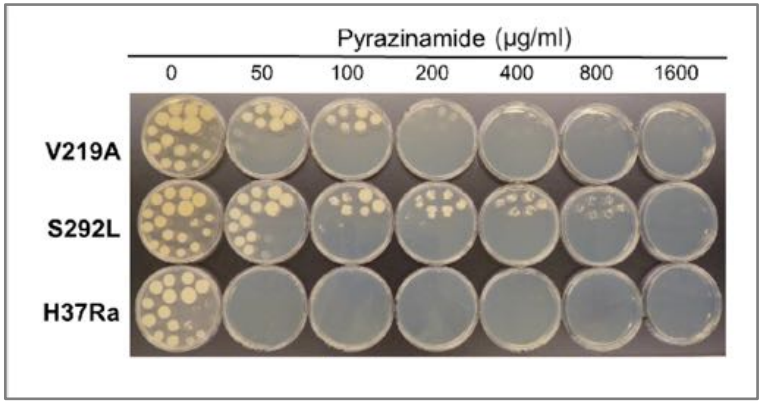
Liu J, Shi W, Zhang S, Hao X, Maslov DA, Shur KV, Bekker OB, Danilenko VN and Zhang Y (2019) Mutations in Efflux Pump Rv1258c (Tap) Cause Resistance to Pyrazinamide, Isoniazid, and Streptomycin in M. tuberculosis. Front. Microbiol. 10:216. doi: 10.3389/fmicb.2019.00216

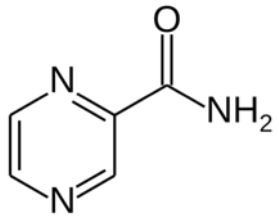
Efflux pumps





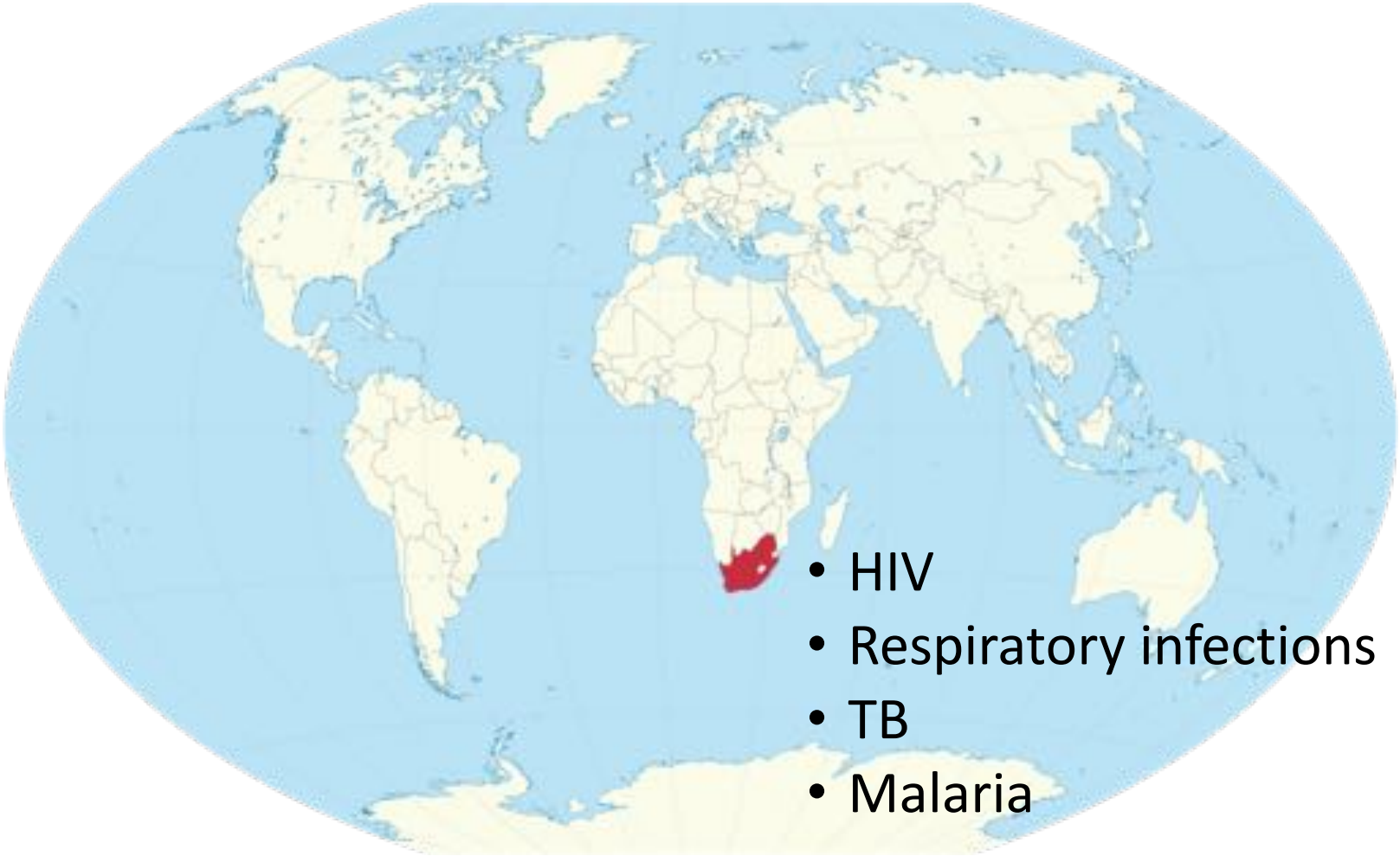






Liu J, Shi W, Zhang S, Hao X, Maslov DA, Shur KV, Bekker OB, Danilenko VN and Zhang Y (2019) Mutations in Efflux Pump Rv1258c (Tap) Cause Resistance to Pyrazinamide, Isoniazid, and Streptomycin in M. tuberculosis. Front. Microbiol. 10:216. doi: 10.3389/fmicb.2019.00216

South Africa



- HIV
- Respiratory infections
- TB
- Malaria

Malaria

Malaria: infection and transmission

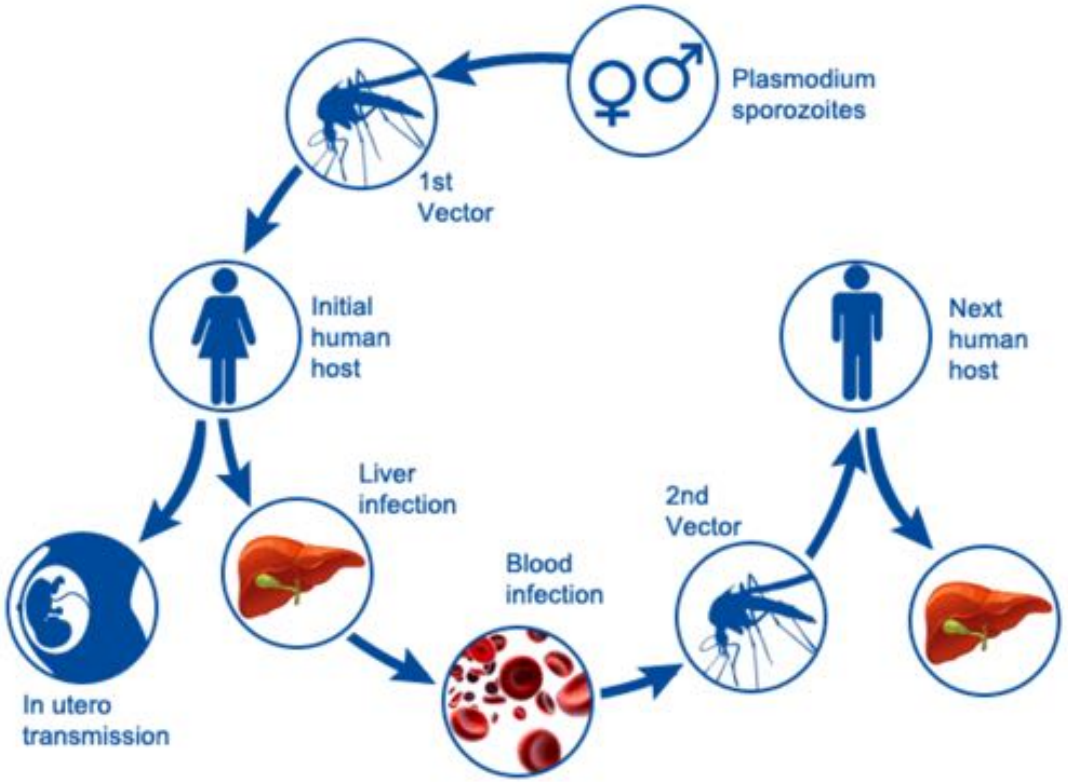
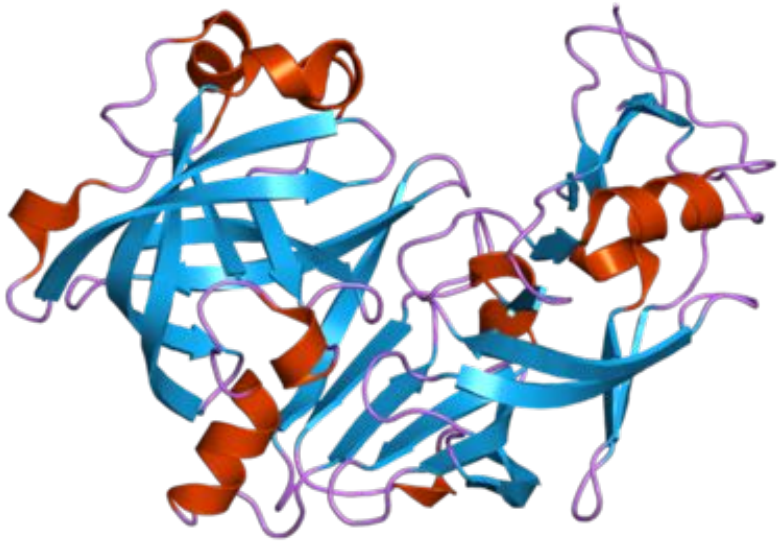


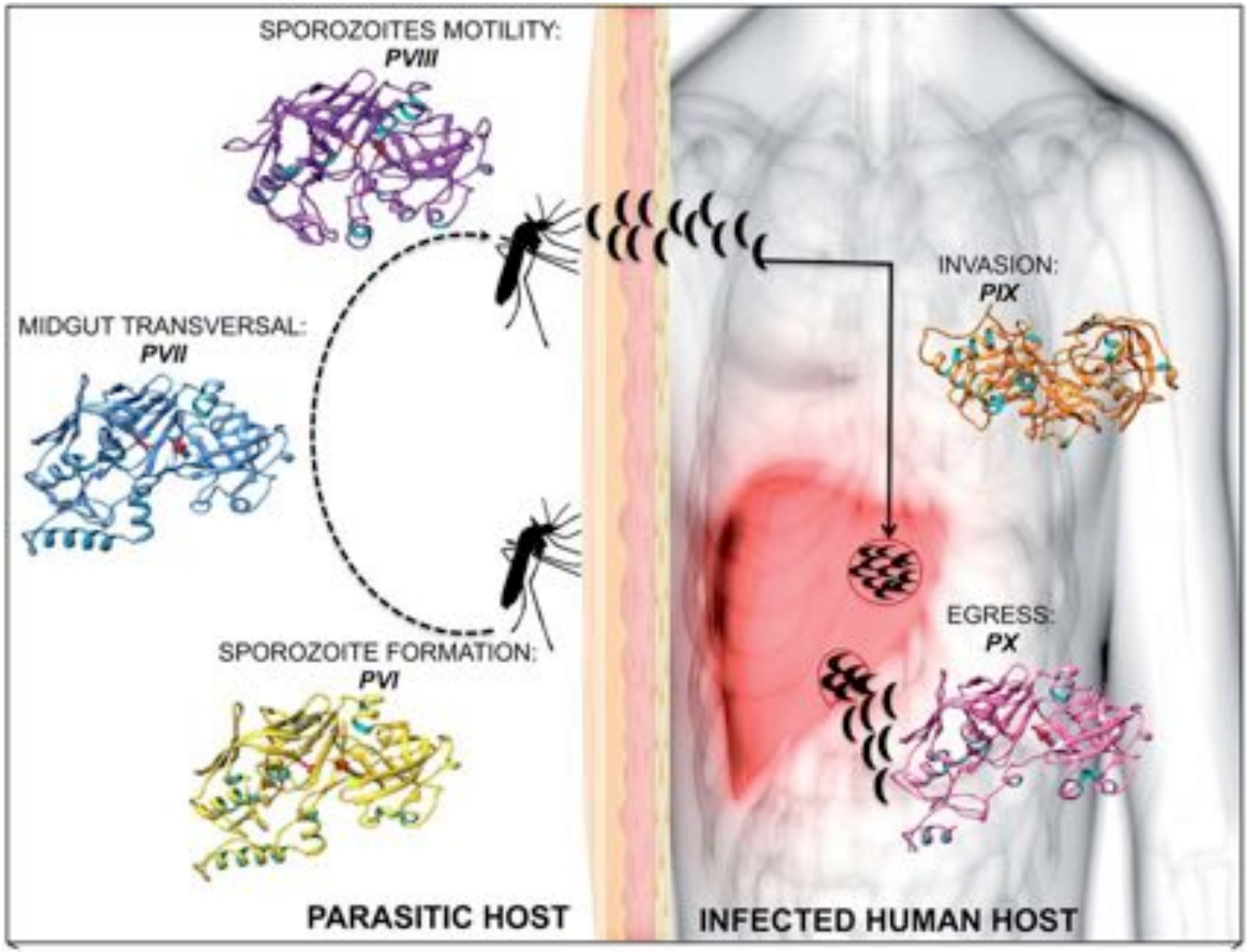
Image: https://www.jenner.ac.uk/about-malaria

Plasmepsin proteins: produced by *Plasmodium falciparum*

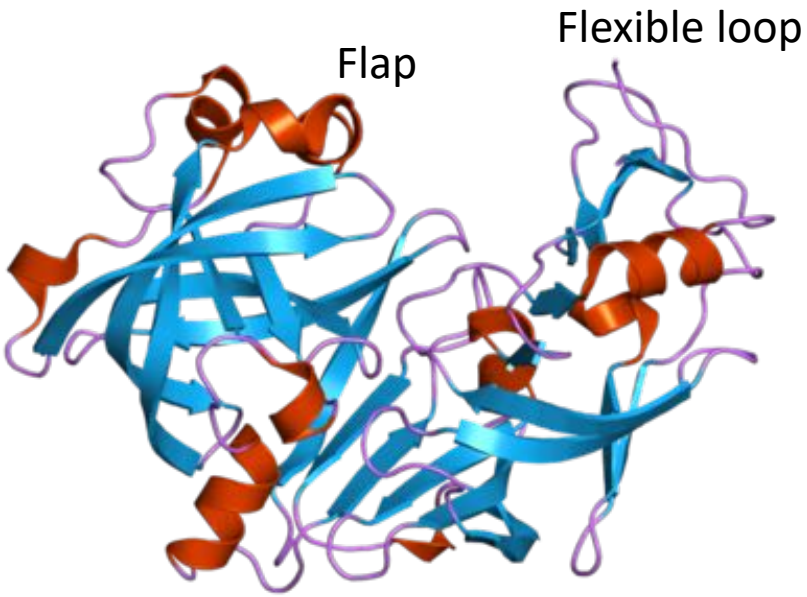


https://en.wikipedia.org/wiki/Plasmepsin_II

Plasmepsin proteins

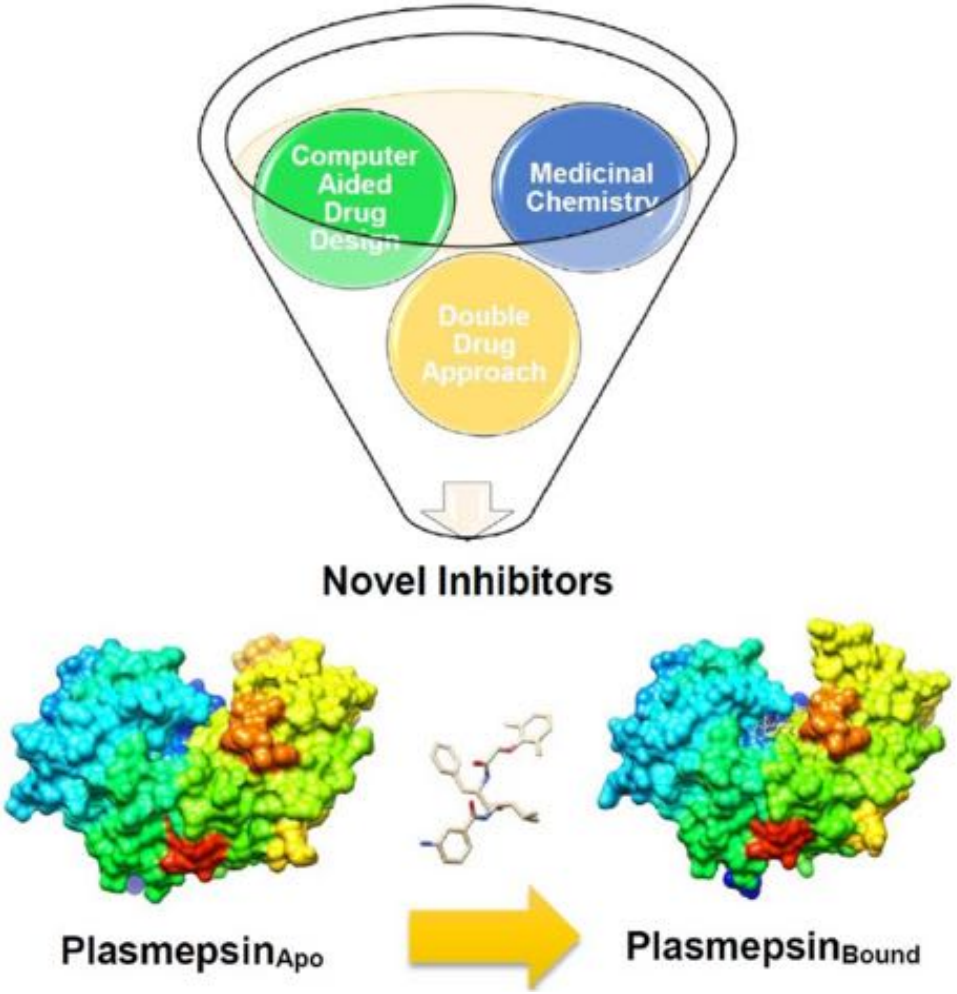


Plasmepsin



https://en.wikipedia.org/wiki/Plasmepsin_II

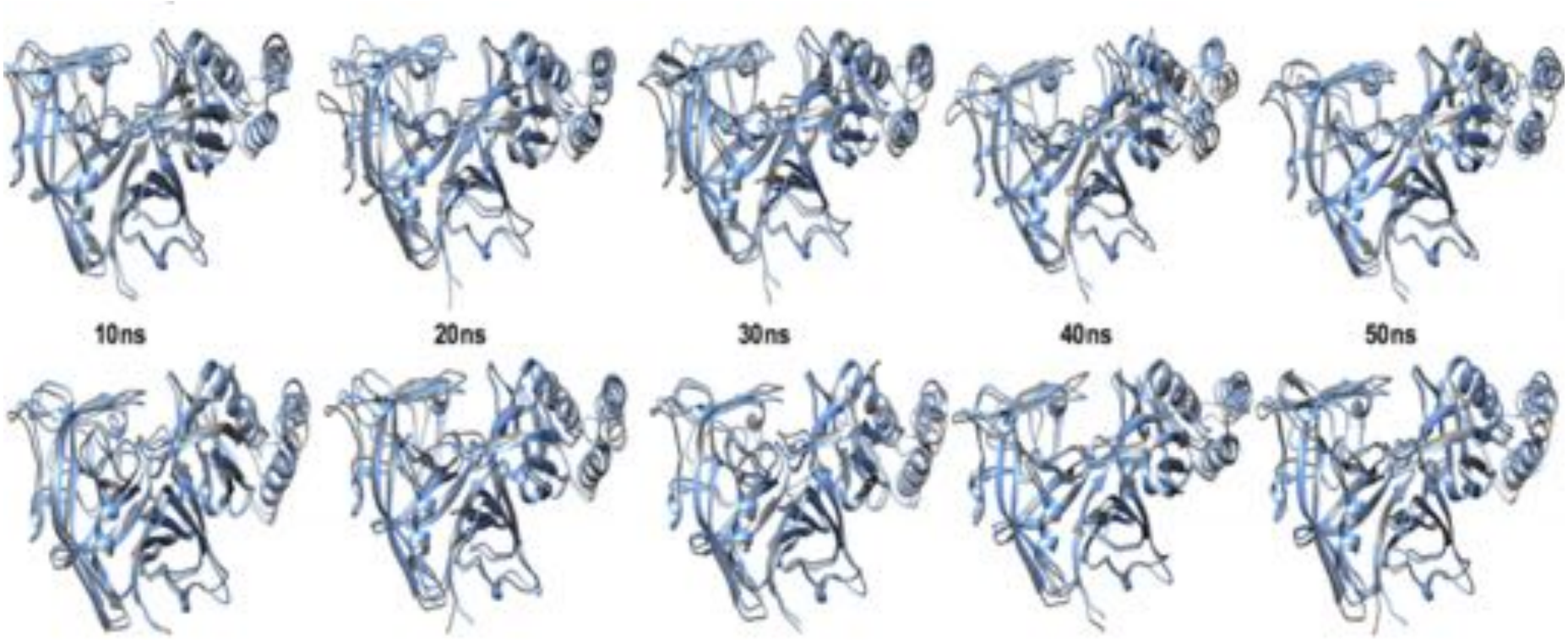
Plasmepsin proteins



N. Dan, S. Bhakat, New paradigm of an old target: an update on structural biology and current progress in drug design towards Plasmepsin II, European Journal of Medicinal Chemistry (2015), doi: 10.1016/j.ejmech.2015.03.049.

Plasmepsin V: MD simulations

Unbound

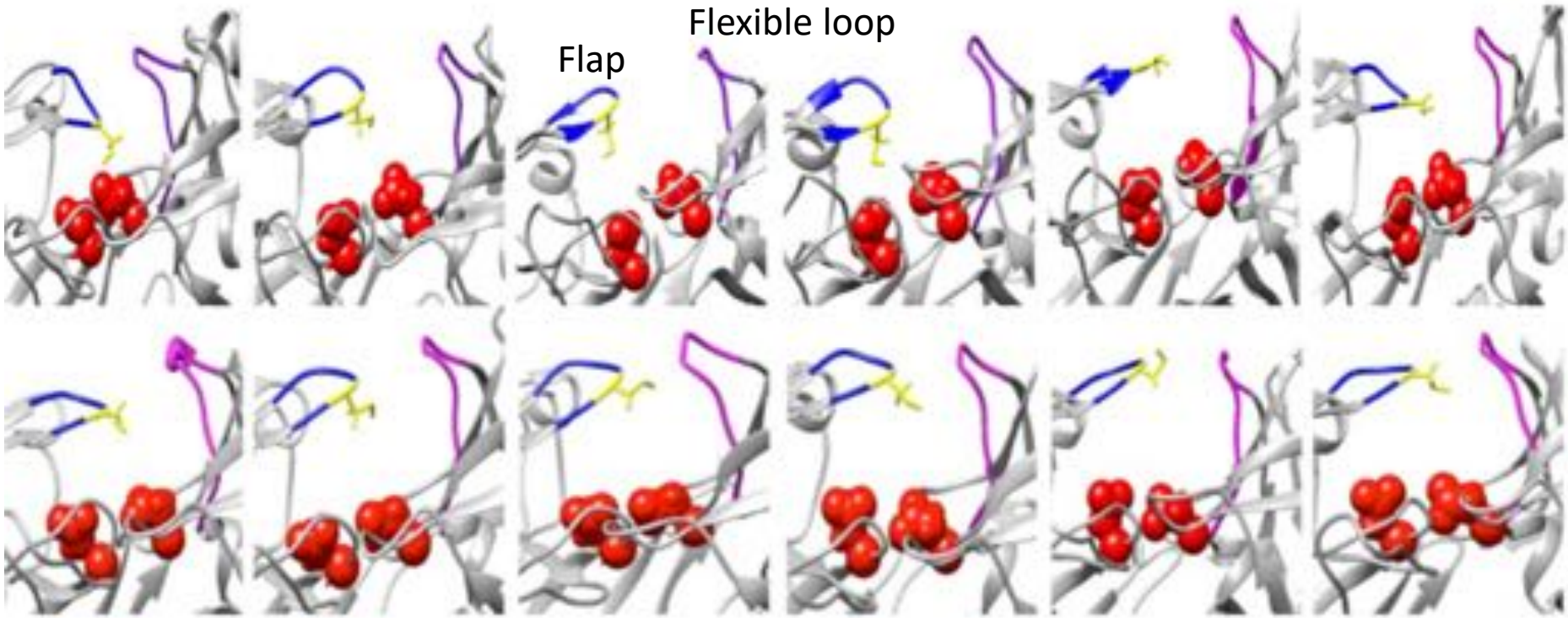


Bound: Inhibitor - WEHI-842

L. McGillewie and M. E.S Soliman, *Mol. BioSyst.*, 2016, DOI: 10.1039/C6MB00077K.

Plasmepsin V: MD simulations

Unbound



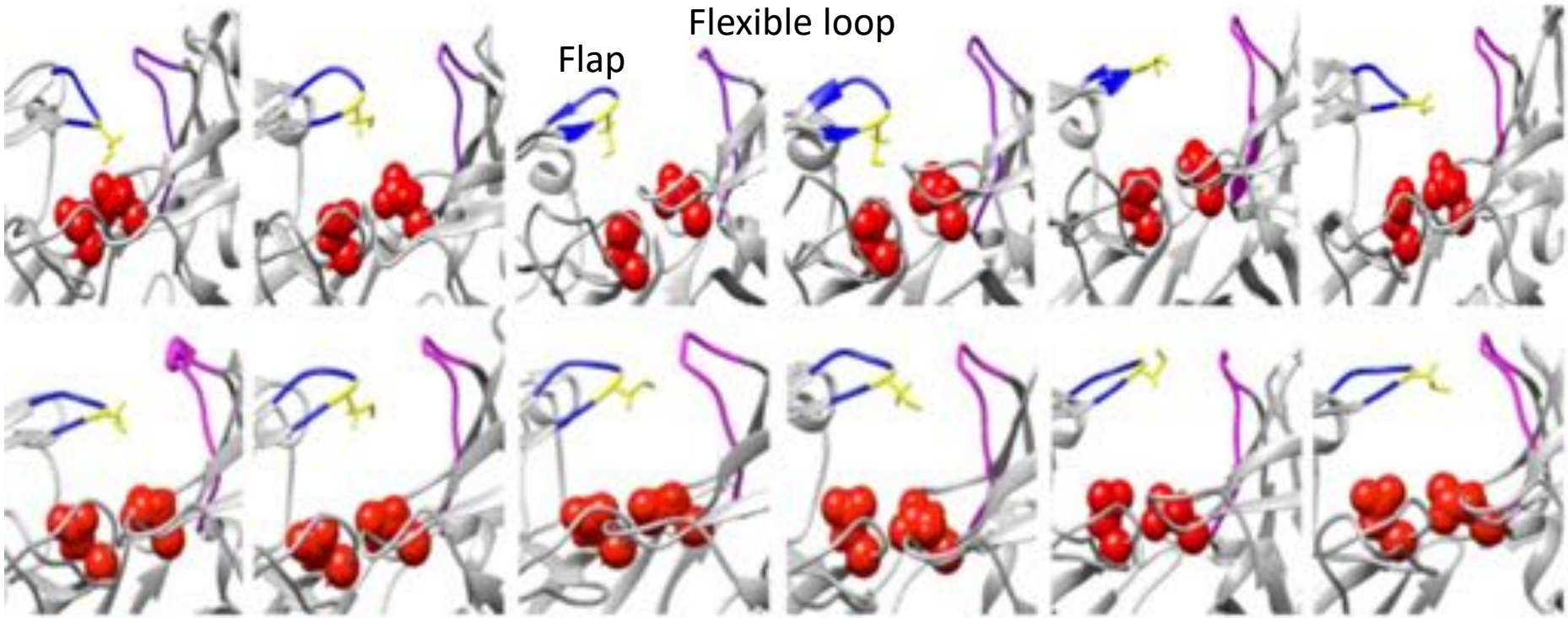
Bound: Inhibitor - WEHI-842

L. McGillewie and M. E.S Soliman, *Mol. BioSyst.*, 2016, DOI: 10.1039/C6MB00077K.

Plasmepsin V: MD simulations

In an unbound conformation, the flap region moves freely opening and closing the active site.

Unbound



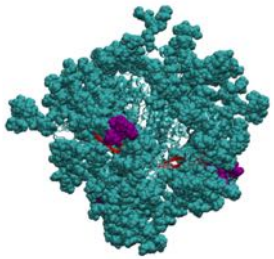
Bound: Inhibitor - WEHI-842

Potential for finding compounds that reduce the flexibility even further.

L. McGillewie and M. E.S Soliman, *Mol. BioSyst.*, 2016, DOI: 10.1039/C6MB00077K.

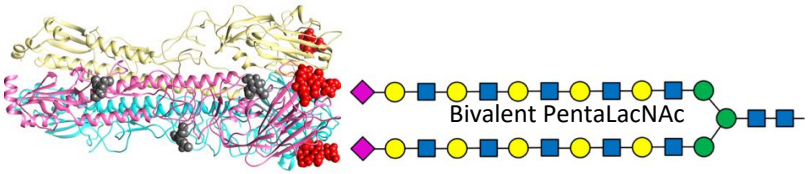
Summary

HIV



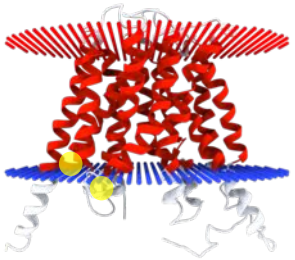
The impact of a loss of a glycan depends on the surrounding landscape.

Influenza A



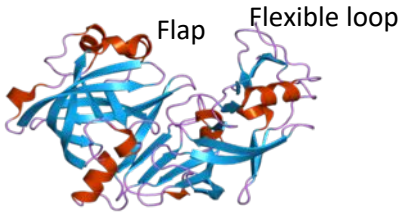
Glycans can rotate and bind in various way.

TB



Point mutations can influence the function of an efflux pump.

Malaria



Inhibitors affect the dynamics of flaps and loops.

Summary

A single mutation can have a substantial influence on the dynamics of a protein.

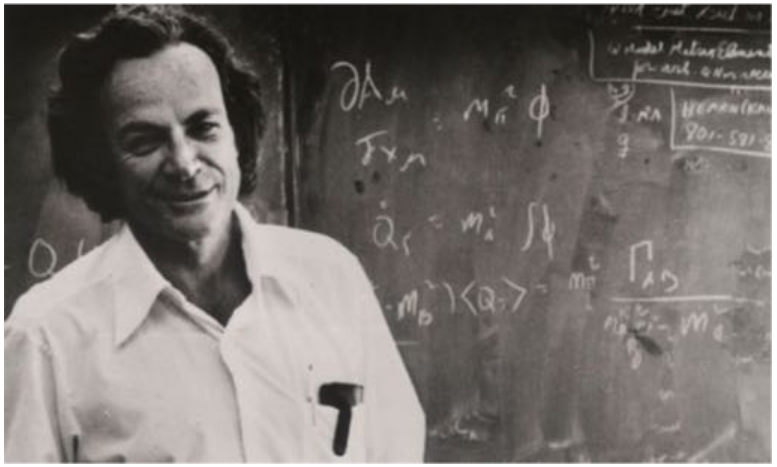
When the protein dynamics is altered, so too is the functioning of the protein.

This, in turn, impacts the spread of infectious diseases.

Final thought

"everything that living things do can be understood in terms of the jiggling and wiggling of atoms."

The Feynman Lectures in Physics vol. 1, 3-6 (1963)



Thanks!

- Roux-Cil Ferreira (University of the Western Cape)
- David Matten (UCT)
- Clare Garrard (UCT)
- Oliver Grant (Complex Carbohydrate Research Center, Atlanta)
- Simon Travers (University of the Western Cape)
- Darren Martin (UCT)
- Nicky Mulder (UCT)