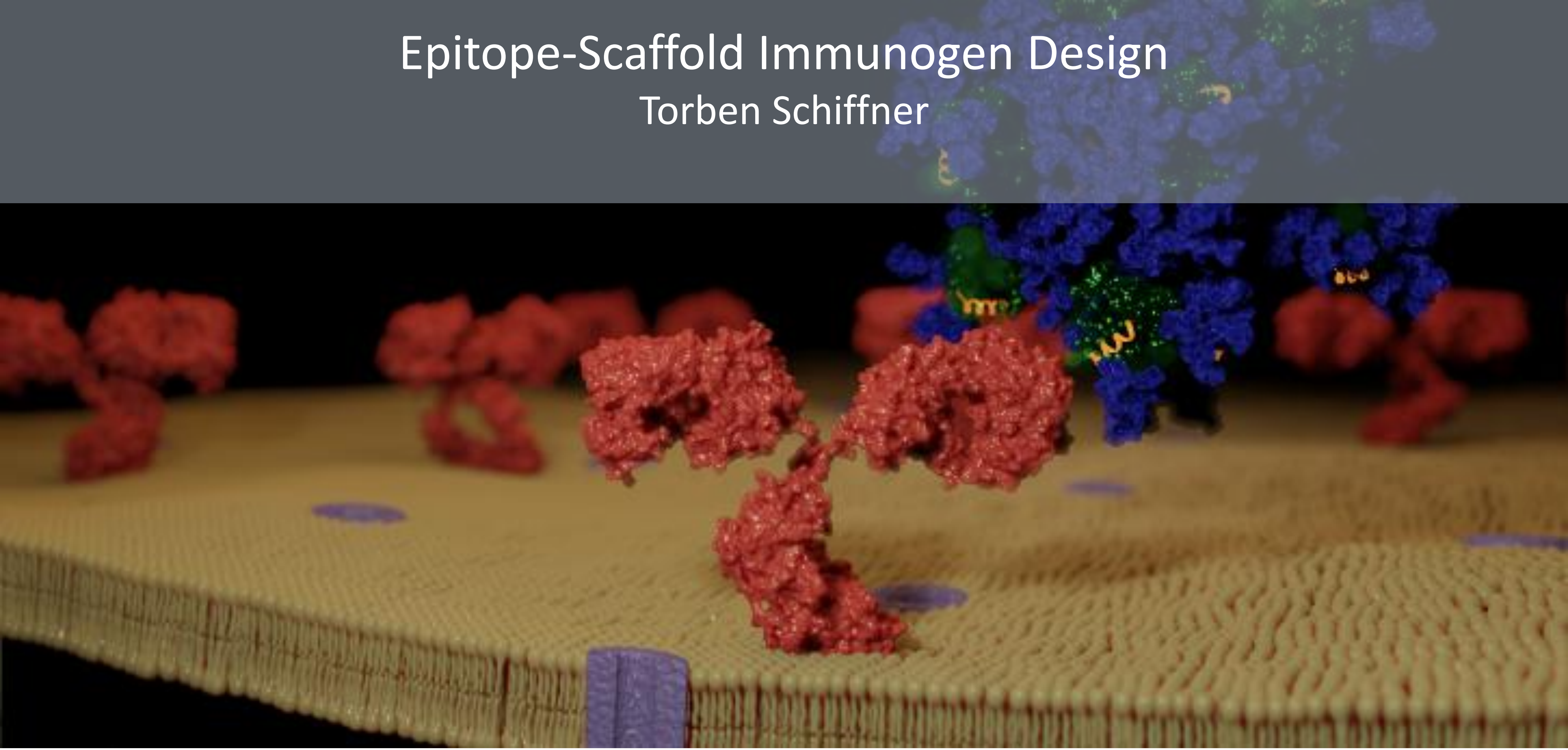
Epitope-Scaffold Immunogen Design

Torben Schiffner



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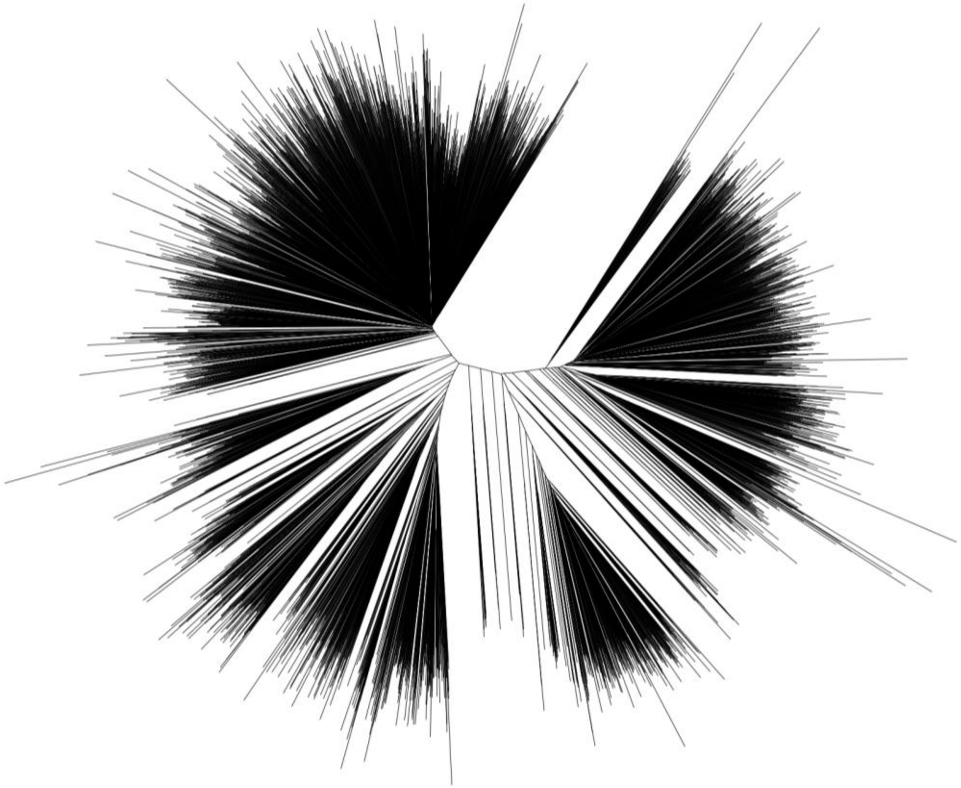
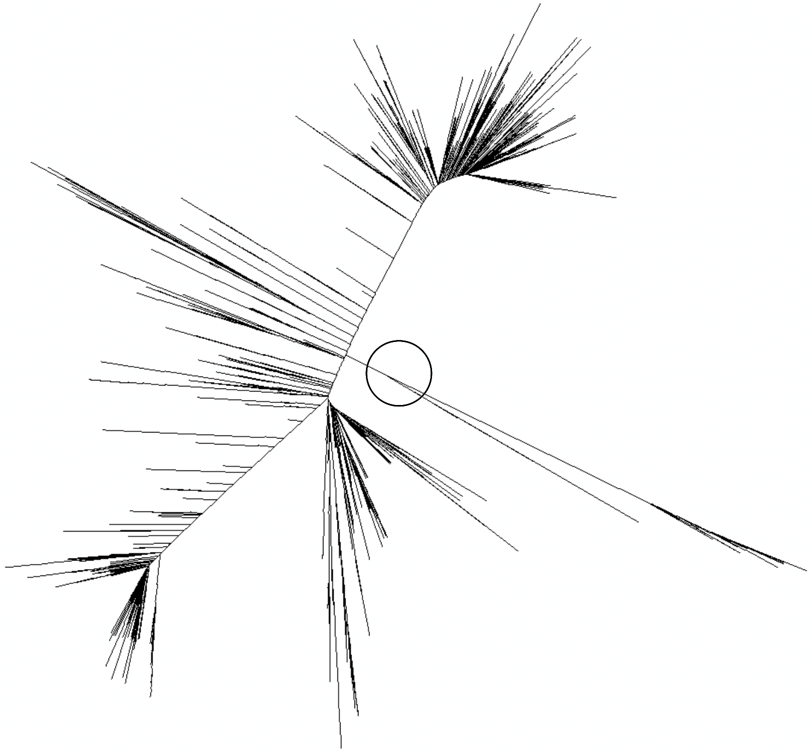
Diversity of HIV Env dwarfs that of SARS-CoV-2 spike

HIV Env
Worldwide 2013

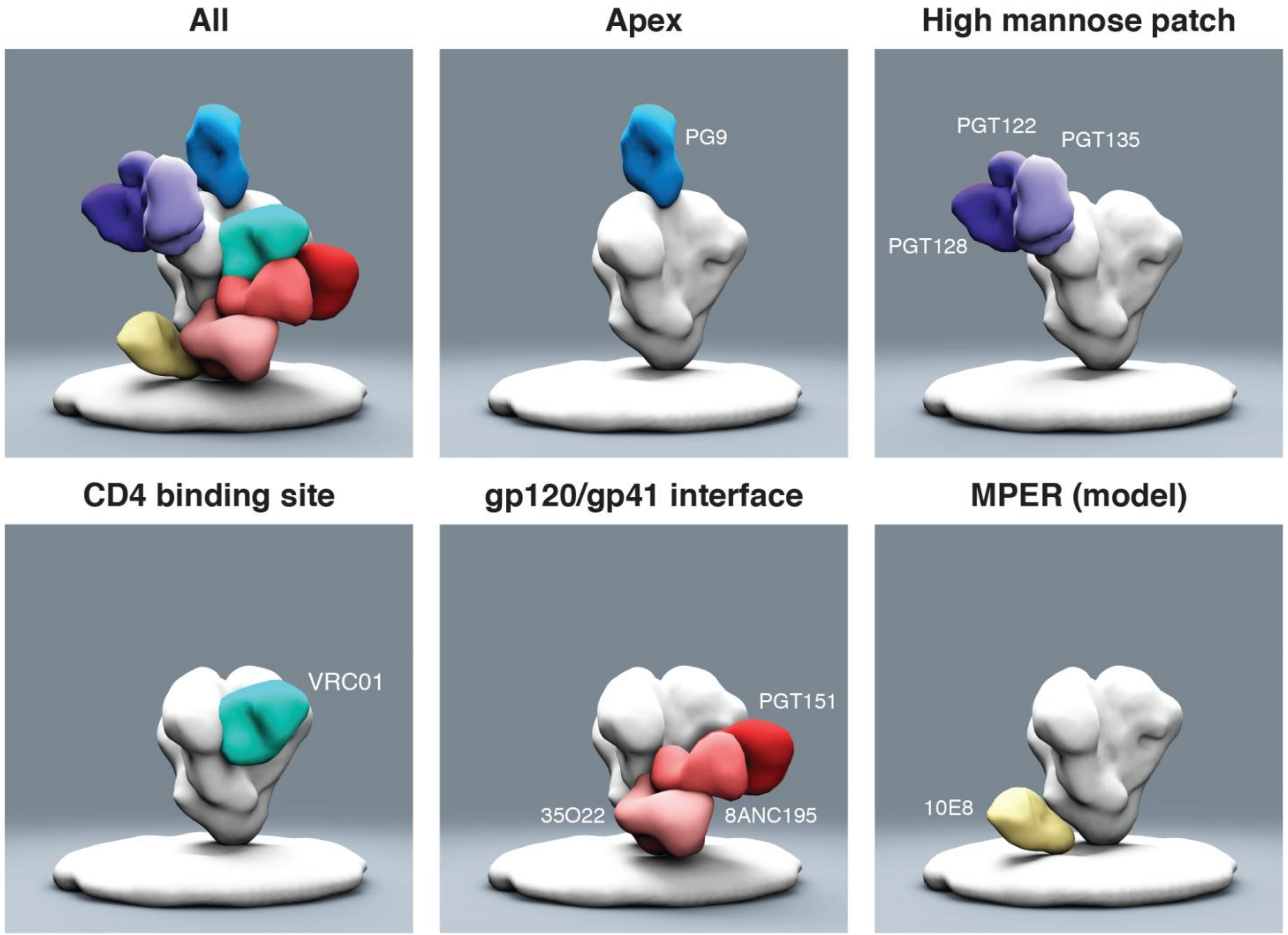


SARS-CoV-2 spike
Worldwide Oct 29, 2022



Trees show amino acid substitutions. 5000 sequences each from LANL and GISAID.

Prototype HIV broadly neutralizing antibodies (bnAbs)



All

Apex

High mannose patch

CD4 binding site

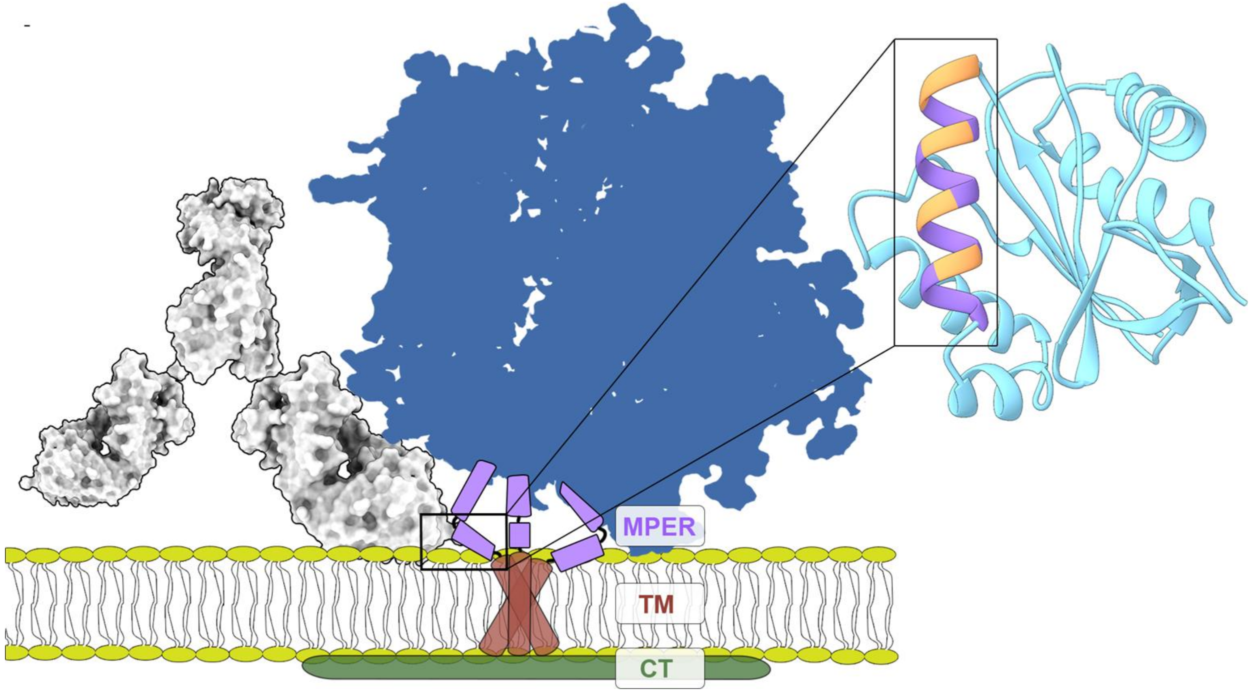
gp120/gp41 interface

MPER (model)

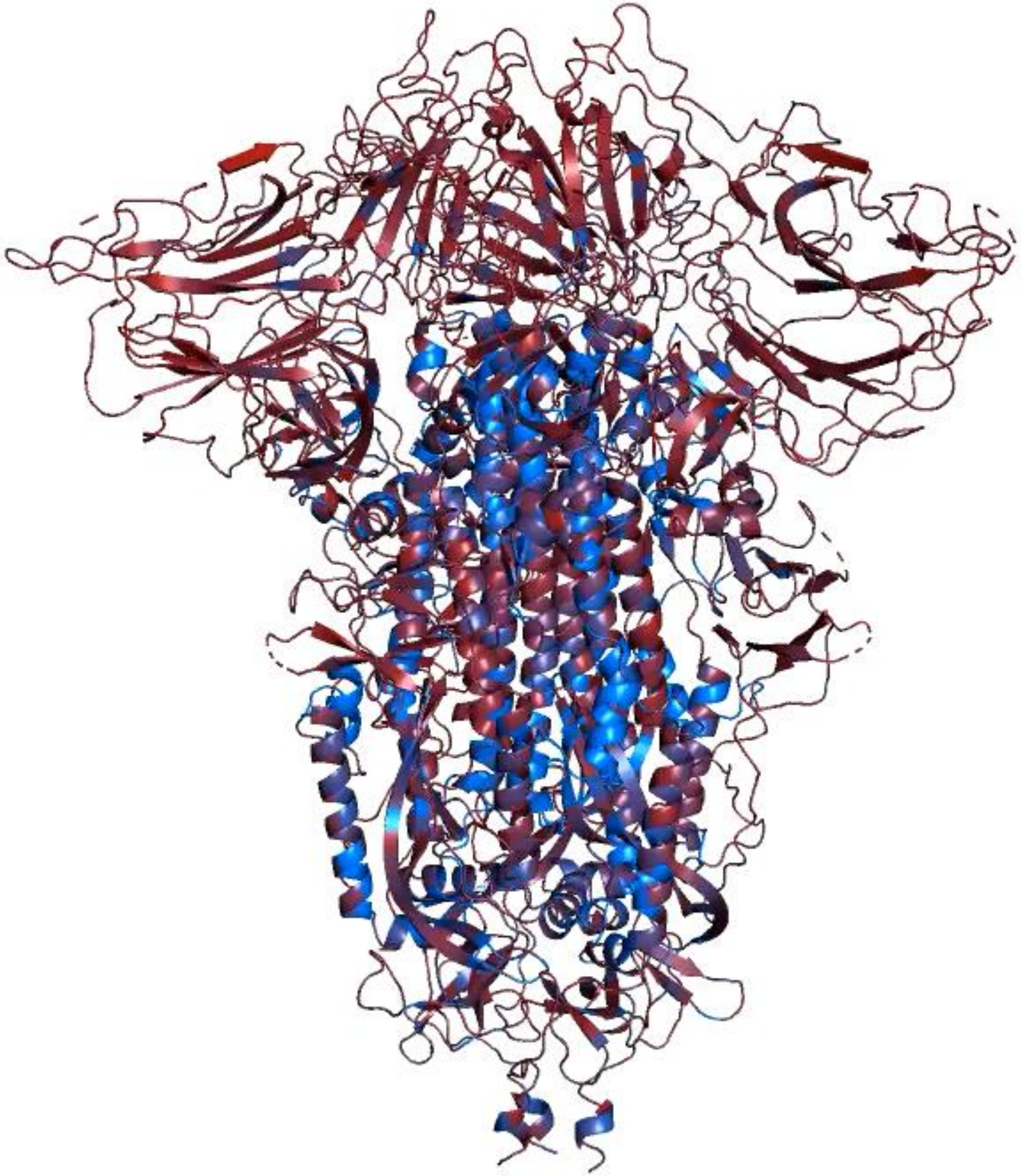
- **bnAbs neutralize diverse isolates, some up to 99% of all isolates**
- **bnAbs can provide sterilizing immunity in NHP models**
- **bnAbs can protect in humans**
- **If vaccine can elicit bnAbs, could prevent HIV infection**
- **Vaccine responses must be broad, potent, and durable**

MPER bnAb 10E8

- 10E8 is very broad (>98%) with good potency (geomean IC_{50}: 0.22 μg/mL)
- Not or weakly polyreactive
- MPER not part of most soluble native-like trimers
- Epitope sterically occluded in Env trimer context
- 10E8 engages the epitope via a long (22aa) HCDR3 with a YxFW binding motif



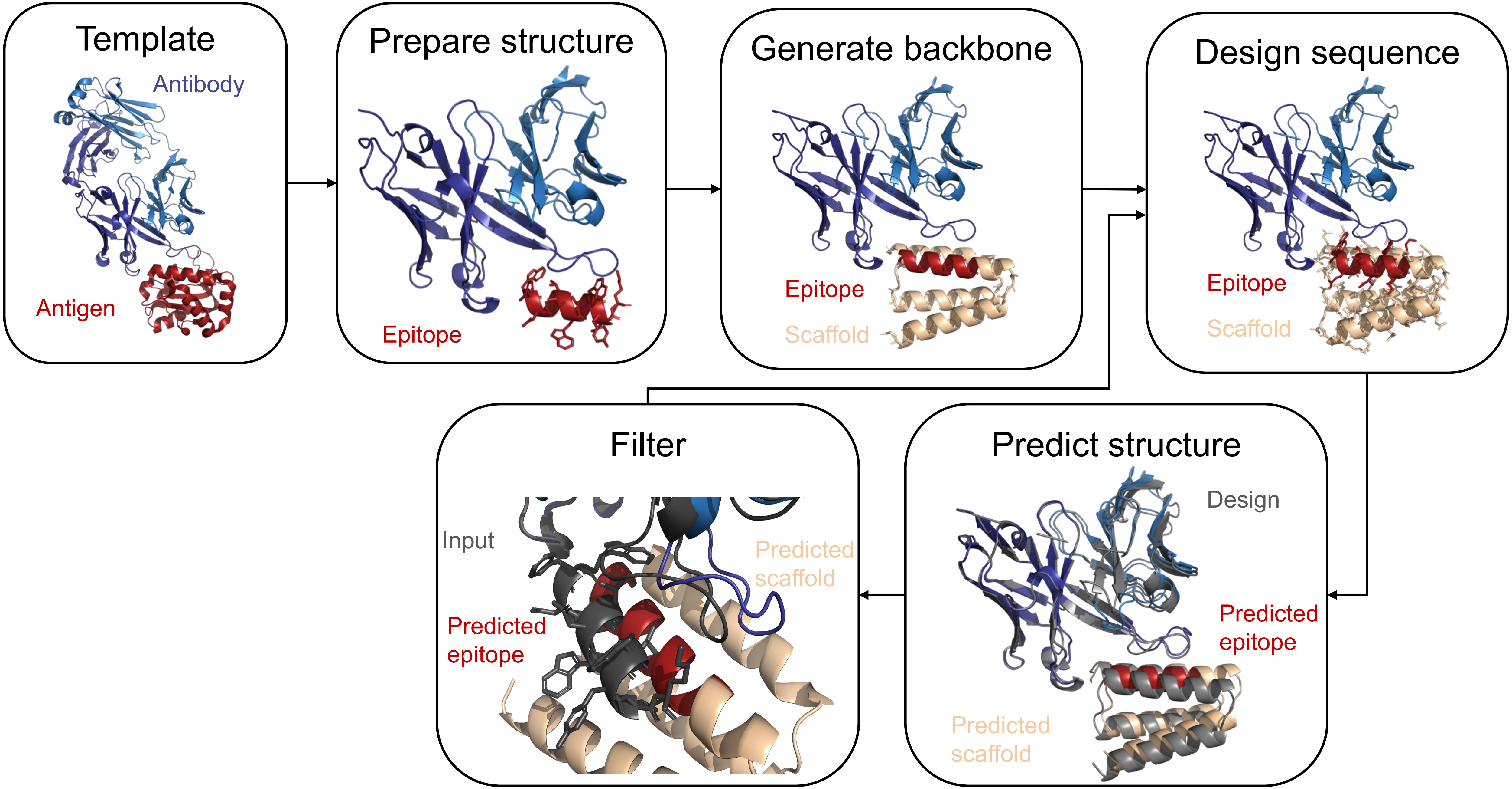
Epitope scaffolding



Sequence variability

Low High

De novo epitope-scaffold design



Template

Prepare structure

Generate backbone

Design sequence

Predict structure

Filter

RFDiffusion

Article

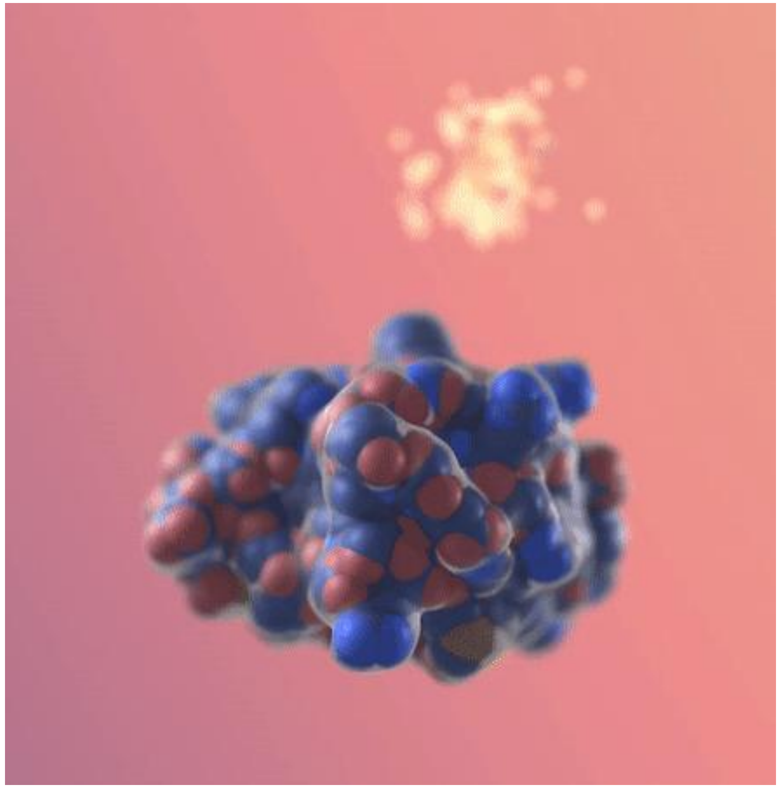
De novo design of protein structure and function with RFdiffusion

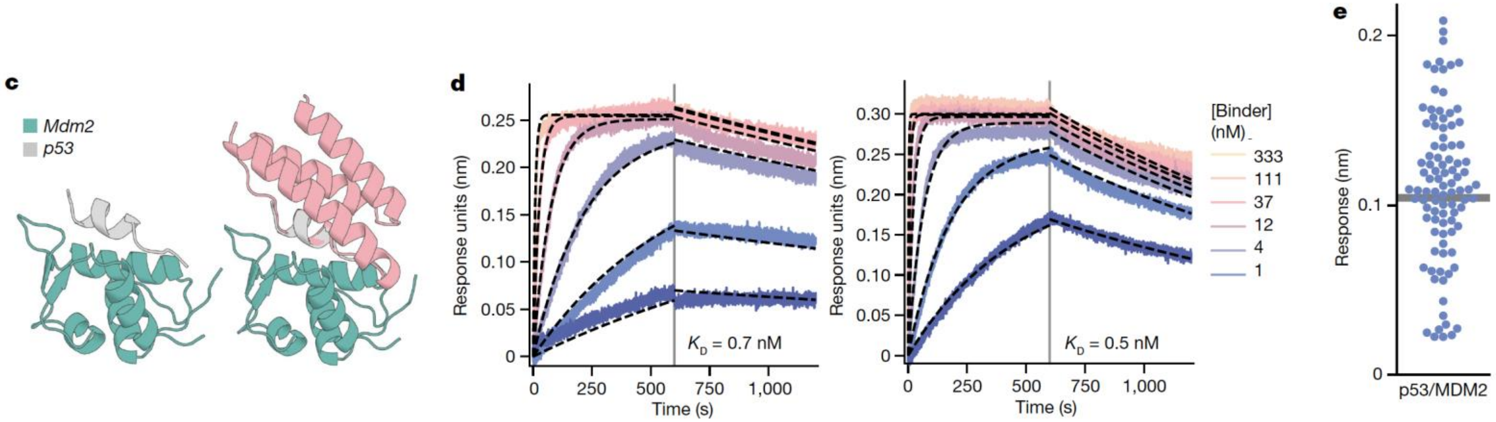
https://doi.org/10.1038/s41586-023-06415-8

Received: 14 December 2022

Accepted: 7 July 2023

Joseph L. Watson[1,2,15], David Juergens[1,2,3,15], Nathaniel R. Bennett[1,2,3,15], Brian L. Trippe[2,4,5,15], Jason Yim[2,6,15], Helen E. Eisenach[1,2,15], Woody Ahern[1,2,7,15], Andrew J. Borst[1,2], Robert J. Ragotte[1,2], Lukas F. Milles[1,2], Basile I. M. Wicky[1,2], Nikita Hanikel[1,2], Samuel J. Pellock[1,2], Alexis Courbet[1,2,8], William Sheffler[1,2], Jue Wang[1,2], Preetham Venkatesh[1,2,9], Isaac Sappington[1,2,9], Susana Vázquez Torres[1,2,9], Anna Lauko[1,2,9], Valentin De Bortoli[8], Emile Mathieu[10], Sergey Ovchinnikov[11,12], Regina Barzilay[6], Tommi S. Jaakkola[6], Frank DiMaio[1,2], Minkyung Baek[13] & David Baker[1,2,14]✉





57% of motif grafting designs bound to target sequence!!!

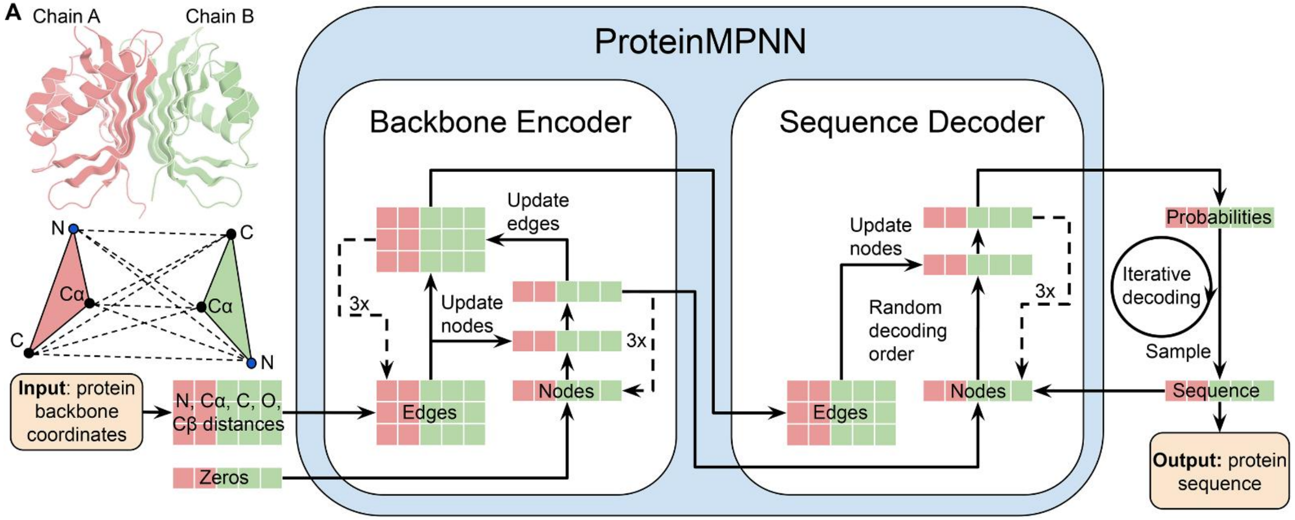
https://www.bakerlab.org/wp-content/uploads/2023/03/RFDiffusion_animation_by_Ian_C_Haydon.gif

Sequence design

- Designs goals:
  - High expression level
  - Stable protein

- Interface is kept intact
- Buried graft residues to be designed

- Suitable algorithms:
  - Rosetta Design
  - ProteinMPNN
  - ESM
  - Others
  - Combinations of the above
  - Multi-state design



The antibody problem

- “As a limitation, we observe anecdotally that AlphaFold-Multimer is generally not able to predict binding of antibodies and this remains an area for future work.”

  Evans et. al; bioRxiv “Protein complex prediction with AlphaFold-Multimer”

- Possible solution: Alphafold with “initial guess”:

Article https://doi.org/10.1038/s41467-023-38328-5

Improving de novo protein binder design with deep learning

Received: 29 July 2022

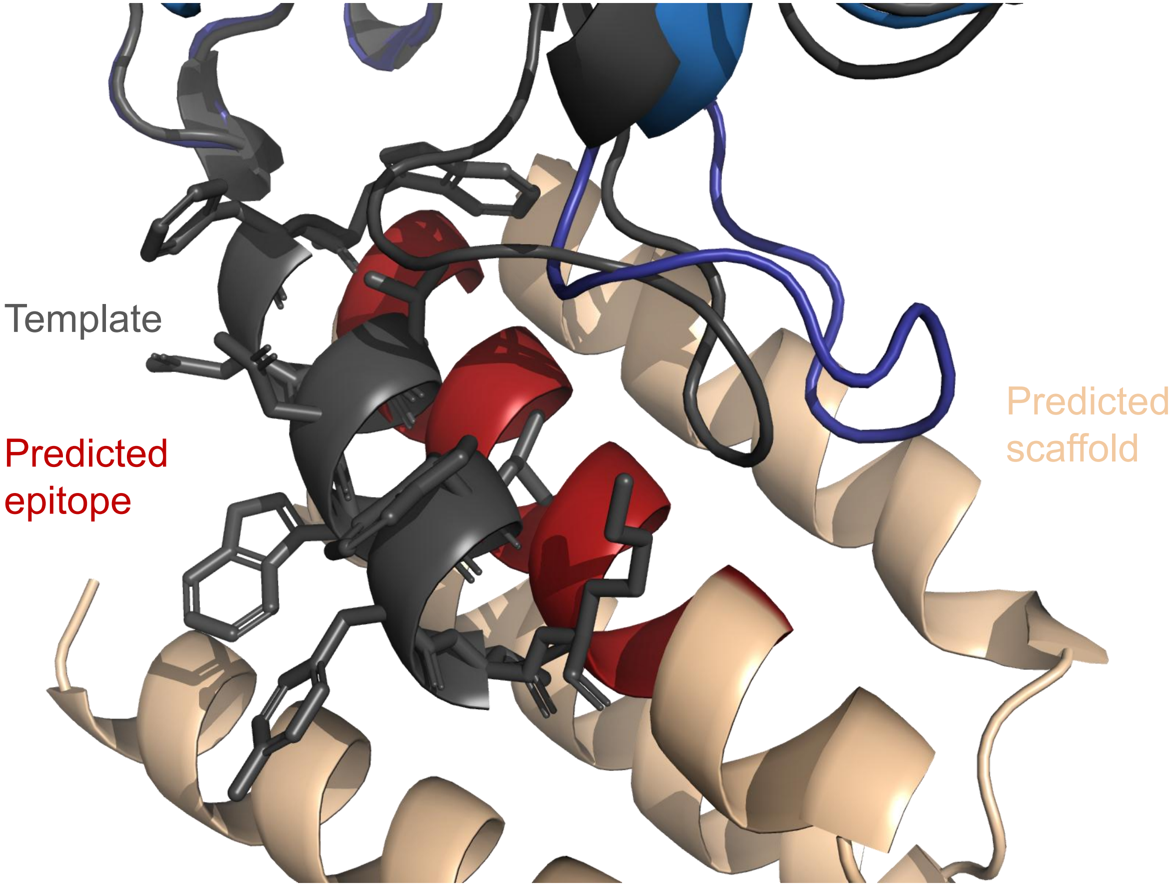
Accepted: 24 April 2023

Published online: 06 May 2023

Nathaniel R. Bennett[1,2,3,8], Brian Coventry[1,2,4,8], Inna Goreshnik[1,2], Buwei Huang[1,2,5], Aza Allen[1,2], Dionne Vafeados[1,2], Ying Po Peng[1,2], Justas Dauparas[1,2], Minkyung Baek[1,2], Lance Stewart[1,2], Frank DiMaio[1,2], Steven De Munck[6,7], Savvas N. Savvides[6,7] & David Baker[1,2,4]

Filtering

- Graft accuracy:
  - Predict structure
  - Align on the antibody
  - Measure RMSD of the graft
  - Cut-off: <3Å
- Prediction accuracy: pLDDT>90
- Interface accuracy: pTM>0.85



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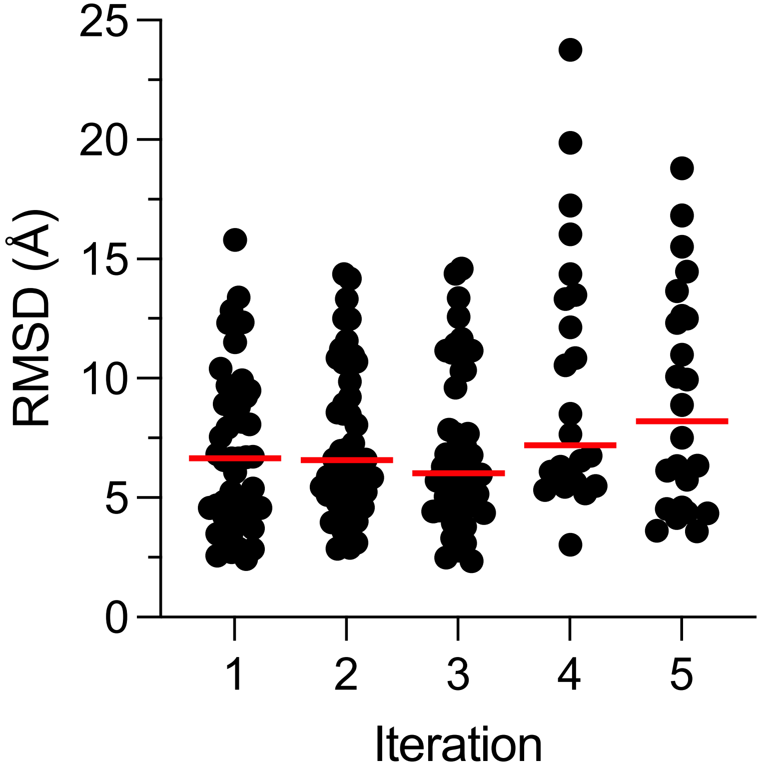
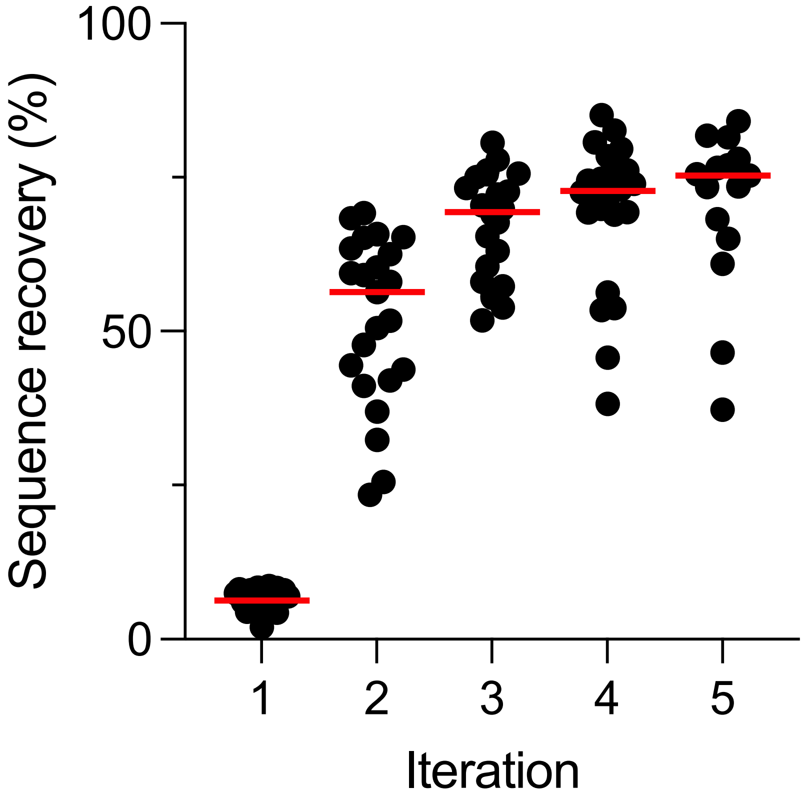
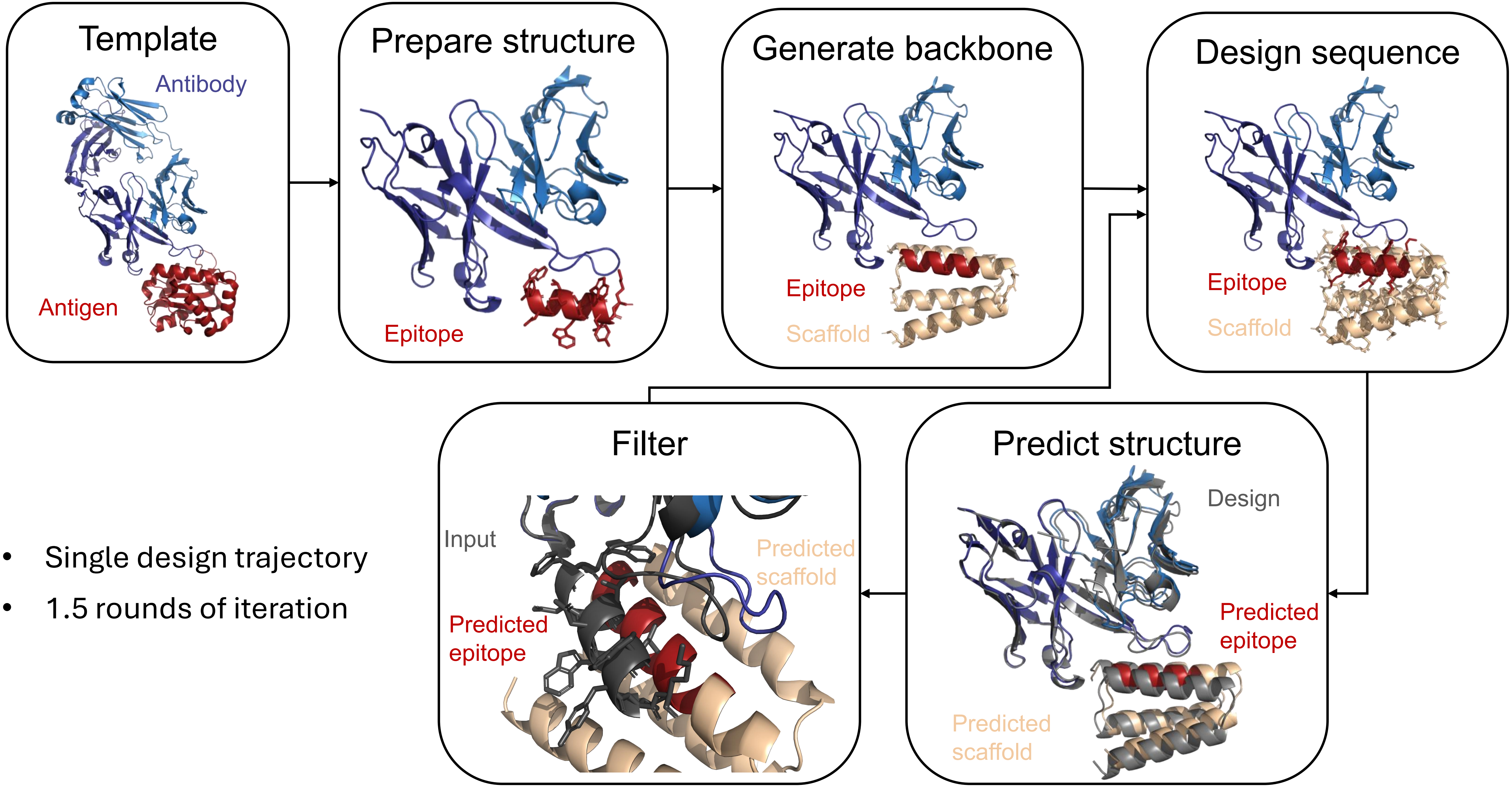


Filtering

- To find designs for which structure and sequence predictions converge, we iteratively design the sequence and predict the structure
- Sequence recovery increases substantially after the first iteration, but converges after 2-3 iterations
- Target: 95%



Today's tutorial



Template

Prepare structure

Generate backbone

Design sequence

Predict structure

Filter

- Single design trajectory
- 1.5 rounds of iteration

Questions?

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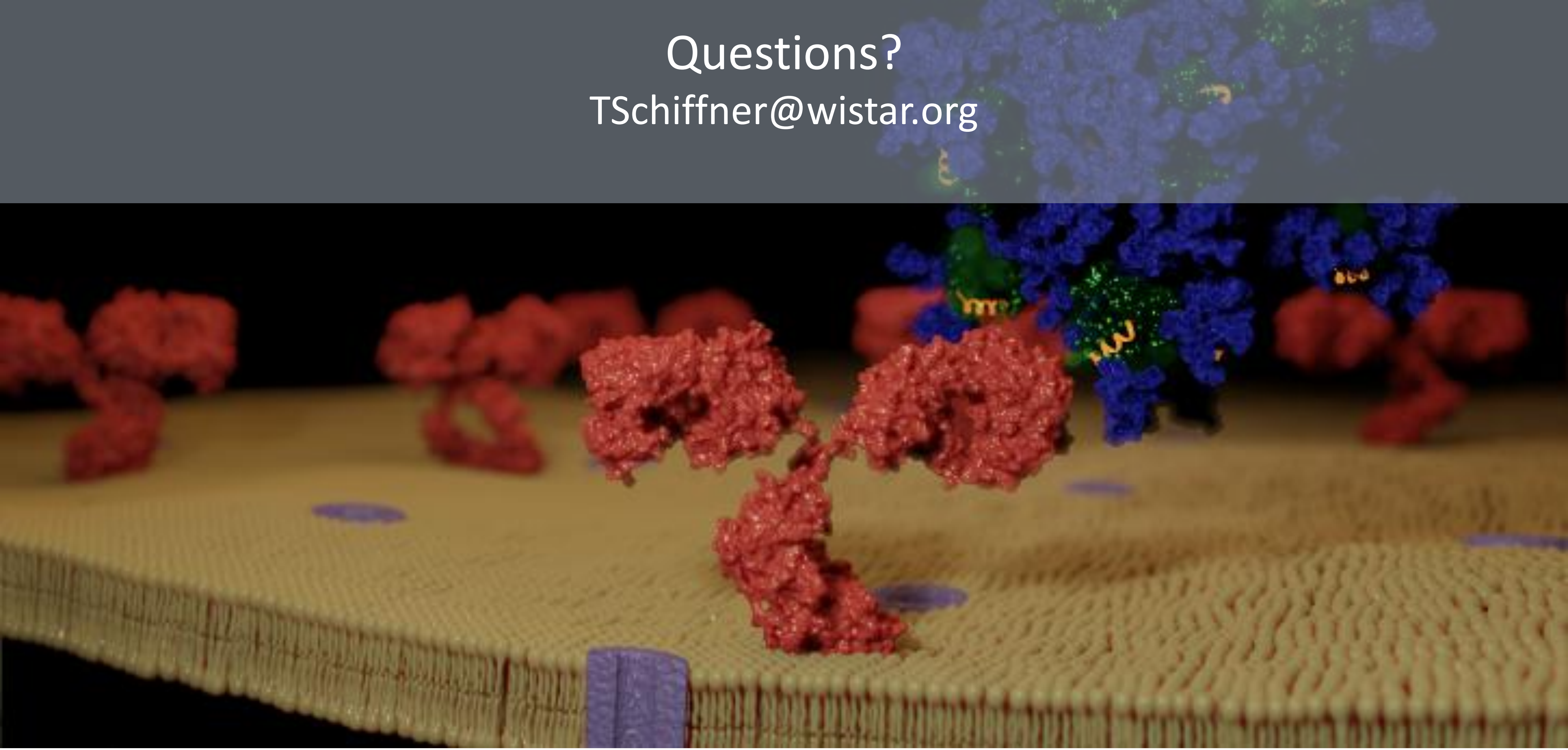

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