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Unlocking the Potential of Antibodies Against SARS-COV-2: A Pipeline for Fast and Accurate Mapping of Interaction Sites with Free Energy Analysis

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Introduction

In this study, we employed free energy interaction analysis to identify key interaction sites between SARS-COV-2 and antibody structures. Specifically, we generated a pipeline for efficient mapping of these interaction sites that can be used for multiple system pairs, or for a single pair using parallelized computing. Our pipeline significantly reduces the time and effort required for this analysis, making it a feasible approach for large-scale studies. Our results demonstrate that the interaction between SARS-COV-2 and antibody structures can be mapped with high accuracy using free energy interaction analysis [1]. The technology can also be used to study protein-protein interactions in general. By mapping the key interaction sites between proteins, researchers can gain insights into the mechanism of interaction and the structural features of the protein complex. This information can help to identify potential drug targets and facilitate the design of peptide inhibitors that disrupt the protein-protein interaction [2].

Input data

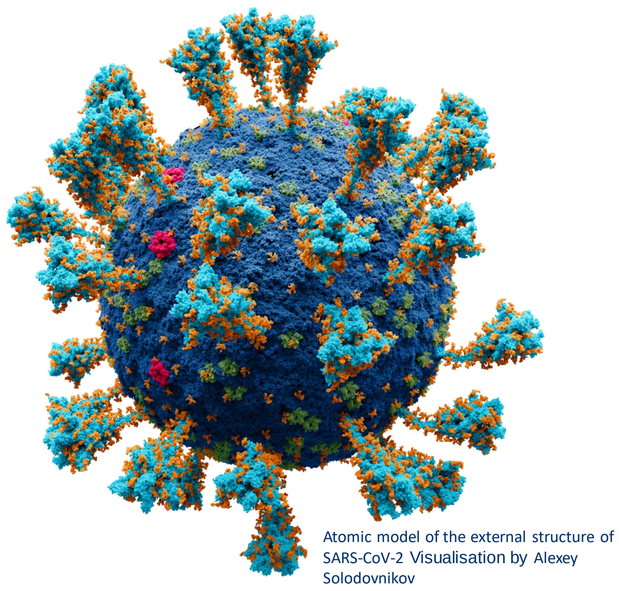
- ✓NCBI PDB
- ✓Any valid (protein) structure file in PDB format
- ✓Identify interacting Chains

Pre-processing of Structures

- ✓Fix Side Chain
- ✓Fix Numbering
- ✓Fix Clashes
- Detect Gaps

Free Energy analysis (Ala scan)

- ✓ΔΔG antibody
- ✓ΔΔG antigen
- ✓Clean structure files



Atomic model of the external structure of SARS-CoV-2 Visualisation by Alexey Solodovnikov

Aims

- ❖From PDB to ΔΔG without any manual input
- ❖Fix or detect PDB inherent flaws (gaps, res numbering, clashes)
- ❖Create workflows deployable for HPC high throughput or accelerated analysis (deployability, parallelisation)

Results

Our pipeline predicted the effect of AA to Ala substitutions utilising the FoldX forcefield on protein protein interactions in SARS-CoV-2 antibodies and for a small antiviral peptide (LCB1). Pre-processing of PDB structure files produces clean structures with completed side chains for further processing as well as a list of gaps in insufficiently resolved structures. The pipeline identified key residues involved in protein-protein interactions of SARS-CoV-2 antibodies and provided insights into the peptide's binding mechanism (Fig 1). Results for a small antiviral peptide against SARS-COV-2 (LCB1) [2] were consistent with experimental data (Fig. 2). The automated workflow removes the need for manual inputs and potential for human error, allowing for high-throughput analyses.

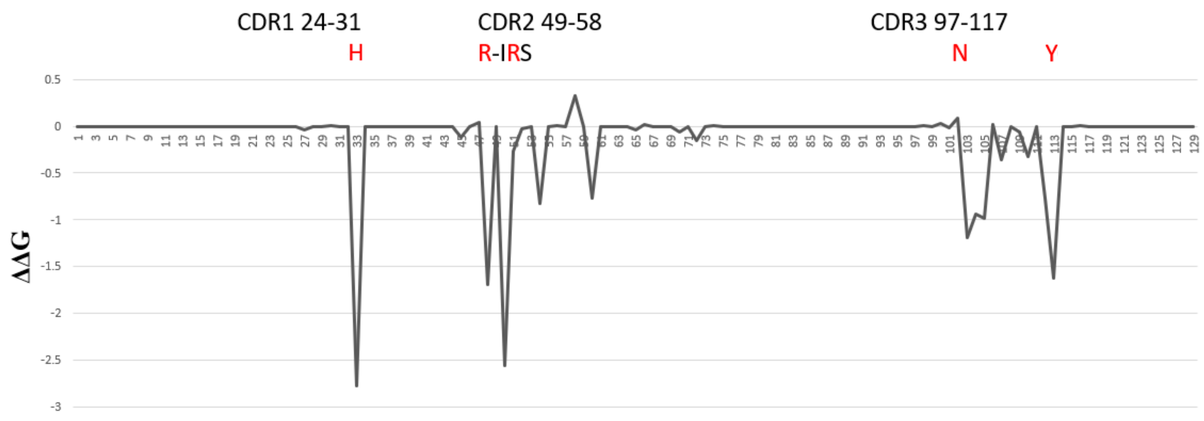
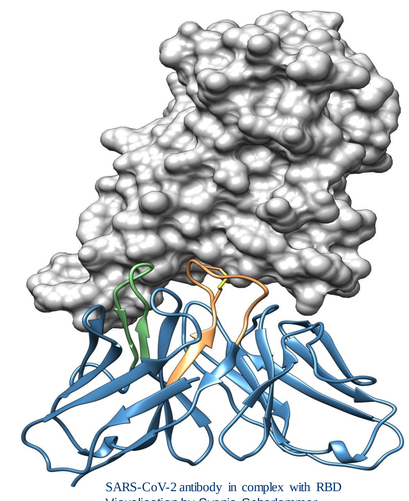


Fig. 1 *Free energy mapping of an Anti-SARS-CoV-2 antibody binding interface. The most influential residues for each position are marked red and visualized on the antibody's paratope. On the Y axis ΔΔG and on the X axis the protein residue for the antibodies heavy chain.*



SARS-CoV-2 antibody in complex with RBD
Visualisation by Svenja Schorlemmer

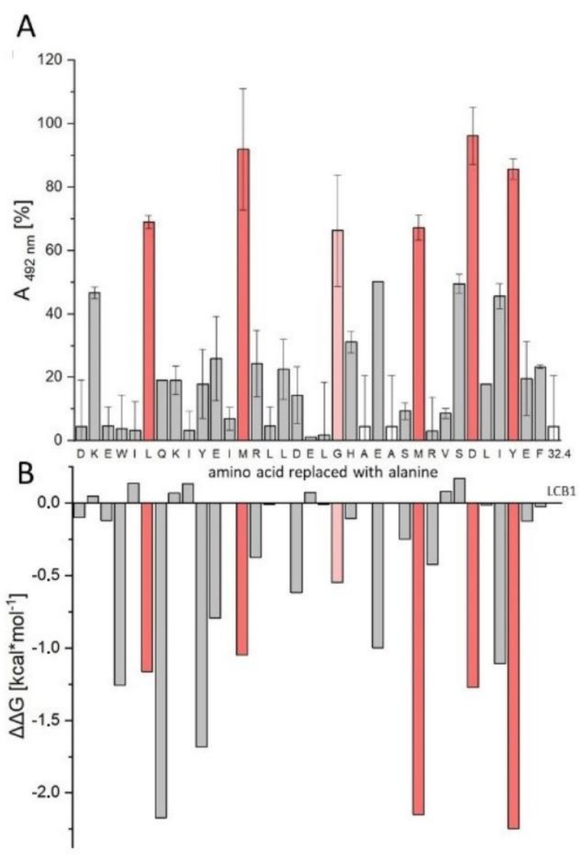


Fig. 2 *Experimental and computational alanine scan of LW32.4 and LCB1, respectively. (**A**) Inhibition of ACE2–RBD interaction by alanine exchange variants of LW32.4 (% remaining interaction at 12 nM peptide). Red bars indicate residues whose replacement with alanine resulted in more than 50% loss of inhibitory activity; white bars denote identical peptides. (**B**) Loss of binding free energy of the LCB1–RBD complex upon in silico replacement of individual LCB1 positions with alanine* [2]

Discussion

Our pipeline provides a promising approach for predicting the effect of AA to Ala substitutions on protein-protein interactions in SARS-CoV-2 antibodies and a small antiviral peptide with free energy analysis. We have shown that it can provide consistent results with experimental data using alanine scanning mutagenesis. Further refinement and validation of the pipeline will show its usefulness to a wide range of systems and experimental settings. Currently the system is deployed for the analysis of anti-HIV and SARS-CoV-2 antibodies. The remaining necessity to identify interacting chains in complex structures prevents a fully automated workflow for now. The system is only bottlenecked by the availability of high-resolution structures. This may be overcome in the future as advances in AI assisted de-novo structure prediction will provide ready access to many new structures without the inherent need for experimental structure generation [3].

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[2] Weißenborn L, Richel E, Hüseman H, Welzer J, Beck S, Schäfer S, Sticht H, Überla K, Eichler J. Smaller, Stronger, More Stable: Peptide Variants of a SARS-CoV-2 Neutralizing Miniprotein. Int J Mol Sci. 2022 Jun 4;23(11):6309. doi: 10.3390/ijms23116309
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