# Uncovering a conserved vulnerability site in SARS-CoV-2 by a human antibody

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**Table of Contents**

Appendix Table S1 – S2

Appendix Fig S1 – S4

Full wwPDB X-ray Structure Validation Report

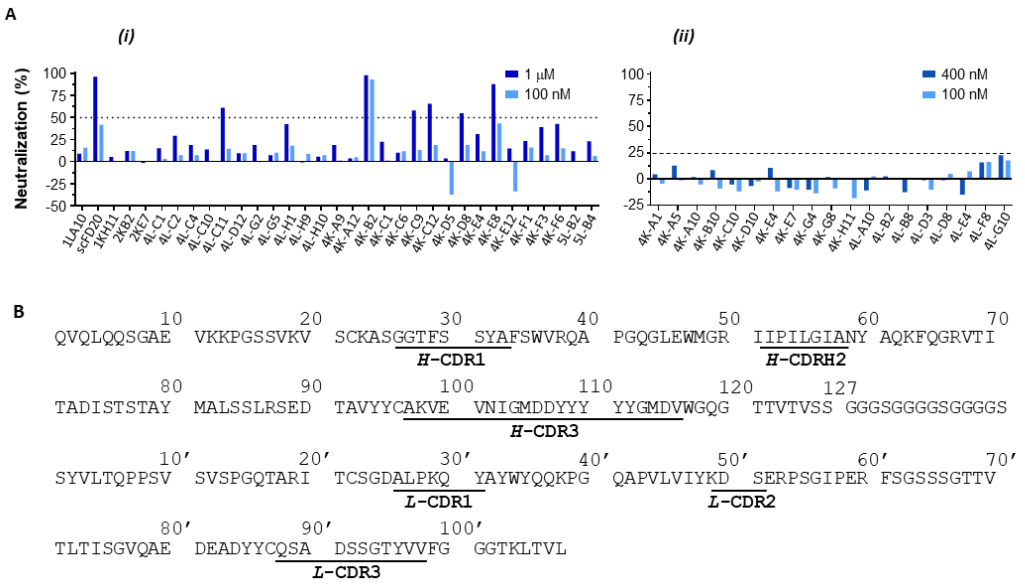
**Appendix Table S1. Amino-acid sequence of neutralizing antibodies identified in this study.**

| scFv | Sequence |
|---|---|
| scFD20 | QVQLQQSGAEVKKPGSSVKVSCKASGGTFSSYAFSWVRQAPGQGLEWMGRIIPILGIANYAQKF QGRVTITADISTSTAYMALSSLRSEDTAVYYCAKVEVNIGMDDYYYYYGMDVWGQGTTVTVSS GGGSGGGGSGGGGSSYVLTQPPSVSVSPGQTARITCSGDALPKQYAYWYQQKPGQAPVLVIYKD SERPSGIPERFSGSSSGTTVTLTISGVQAEDEADYYCQSADSSGTYVVFGGGTKLTVL |
| 4LC11 | QVQLVQSGTEVKKPGASVKVSCKASGYTFTSYYMHWVRQAPGQGLEWMGIINPGGGSTSYAQ KFQGRVTMTTDTSTSTVYMELSSLRSEDTAMYYCARDLYGVPAVGGMDVWGQGTLVTVSSGG GGSGGGGSGSGGSQSALTQPPSASGTPGQRVTISCSGSSSNIGSNSVNWYQQLPGTAPKLLIYGSS QRPSGVPARFSGSKSGTSASLAISGLRSDDEAYYYCAAWDDSLGGAVFGGGTQLTVL |
| 4KB2 | EVQLVQSGAEVTQPGASVKVSCKASGYTFTSYYMHWVRQAPGQGLEWMGIINPGGGSTSYAQ KFQGRVTMTTDTSTSTVYMELSSLRSEDTAMYYCARDLYGVPAVGGMDVWGQGTLVTVSSGG GGSGGGGSGGGGSETTLTQSPATLSVSPGERATLSCRASQTVGGNFLAWYQQKPGRAPRLLIYG TSGRAAGIPDRFTGSASGTDFTLTISRLDPEDFAVYYCQQRSKWYTFGQGTKLEIK |
| 4KC9 | QVQLVQSGTEVKKPGASVKVSCKASGYTFTSYYMHWVRQAPGQGLEWMGIINPGGGSTSYAQ KFQGRVTMTTDTSTSTVYMELSSLRSEDTAMYYCARDLYGVPAVGGMDVWGQGTTVTVSSGG GGSGGGGSGGGGPDIQMTQSPSSLSASVGDRVTITCRASQSISSYLNWYQQRPGKAPKLLIYAAS SLQSGVPSRFSGTTSGTDFTLTISSLQPEDFATYYCQQANSFPLTFGGGTKLEIK |
| 4KC12 | QVQLVQSGTEVKKPGASVKVSCKASGYTFTSYYMHWVRQAPGQGLEWMGIINPGGGSTSYAQ KFQGRVTMTTDTSTSTVYMELSSLRSEDTAMYYCARDLYGVPAVGGMDVWGQGTTVTVSSGG GGSGGGGSGGGGSDIQMTQSPSSVSASVGDRVTITCRASQYISSRFAWYQQQPGKAPKLLIYAAS TLQSGVPSRFSGTTSGTDFTLTISSLQPEDFATYYCQQASSFPLTFGGGTKVEIK |
| 4KD8 | QVQLVQSGAEVKKPGASVKVSCKASGYTFTSYYMHWVRQAPGQGLEWMGIINPGGGSTSYAQ KFQGRVTMTTDTSTSTVYMELSSLRSEDTAMYYCARDLYGVPAVGGMDVWGQGTLVTVSSGG GGSGGGGSGGGGSEIVLTQSPATLSLSPGERATLSCRASQSVSSYLAWYQQKPGQAPRLLIYDAS NRATGIPARFSGSGSGTDFTLTISSLEPEDFAVYYCQQRSNWPRVITFGQGTRLEIK |
| 4KE8 | QVQLVQSGTEVKKPGASVKVSCKASGYTFTSYYMHWVRQAPGQGLEWMGIINPGGGSTSYAQ KFQGRVTMTTDTSTSTVYMELSSLRSEDTAMYYCARDLYGVPAVGGMDVWGQGTLVTVSSGG GGSGGGGSGSGGSDIQMTQSPSSLSASVGDRVTITCRASQNIGIYLNWYQQKPGKAPELLLYTAS TLQSGVPSRFRGSGSGTDFTLTISSLQHEDFATYYCQQSYTTPPTFGGGTKVDIK |

**Appendix Table S2. Primers used in the construction of scFv phage display libraries.**

| Name[a] | Sequence (5'-3')[b] |
|---|---|
| **IgH 1st PCR** | |
| 5' L-VH1 | **ACAGGTGCCCACTCCCAGGTGCAG** |
| 5' L-VH3 | **AAGGTGTCCAGTGTGARGTGCAG** |
| 5' L-VH4/6 | **CCCAGATGGGTCCTGTCCCAGGTGCAG** |
| 5' L-VH5 | **CAAGGAGTCTGTTCCGAGGTGCAG** |
| 3' anti-Cγ CH1 | **TCTTGTCCACCTTGGTGTTGCT** |
| **IgH 2nd PCR** | |
| 5' phage-all-VH1 | GCCGGCCATGGCAGCCGGCTCTTCA**CAGGTGCAGCTGGTGCAGTCTGG** |
| 5' phage-all-VH6-1 | GCCGGCCATGGCAGCCGGCTCTTCA**CAGGTACAGCTGCAGCAGTCAGG** |
| 5' phage-all-VH2 | GCCGGCCATGGCAGCCGGCTCTTCA**CAGGTCACCTTGARGGAGTCTGG** |
| 5' phage-all-VH3 | GCCGGCCATGGCAGCCGGCTCTTCA**GAGGTGCAGCTGGTGGAGTCTGG** |
| 5' phage-all-VH4 | GCCGGCCATGGCAGCCGGCTCTTCA**CAGGTGCAGCTGCAGGAGTCGGG** |
| 5' phage-all-VH5 | GCCGGCCATGGCAGCCGGCTCTTCA**GAGGTGCAGCTGTTGCAGTCTG** |
| 3' GS JH1/2 /4/5 | GAGCCACCTCCGCCGCTACCGCCGCCTCCAGA**TGAGGAGACGGTGACCAG** |
| 3' GS JH3 | GAGCCACCTCCGCCGCTACCGCCGCCTCCAGA**TGAAGAGACGGTGACCATTG** |
| 3' GS JH6 | GAGCCACCTCCGCCGCTACCGCCGCCTCCAGA**TGAGGAGACGGTGACCGTG** |
| **Igλ 1st PCR** | |
| 5′ L Vλ 1 | **GGTCCTGGGCCCAGTCTGTGCTG** |
| 5′ L Vλ 2 | **GGTCCTGGGCCCAGTCTGCCCTG** |
| 5′ L Vλ 3 | **GCTCTGTGACCTCCTATGAGCTG** |
| 5′ L Vλ 4/5 | **GGTCTCTCTCSCAGCYTGTGCTG** |
| 5′ L Vλ 6 | **GTTCTTGGGCCAATTTTATGCTG** |
| 5′ L Vλ 7 | **GGTCCAATTCYCAGGCTGTGGTG** |
| 5′ L Vλ 8 | **GAGTGGATTCTCAGACTGTGGTG** |
| 3' C λ | **CACCAGTGTGGCCTTGTTGGCTTG** |
| **Igλ 2nd PCR** | |
| 5' GSI Vλ1 | AGCGGCGGAGGTGGCTCAGGCGGTGGCGGAAGT**CAGTCTGTGCTGACKCAG** |
| 5' GSVλ2 | AGCGGCGGAGGTGGCTCAGGCGGTGGCGGAAGT**CAGTCTGCCCTGACTCAG** |
| 5' GS Vλ3 | AGCGGCGGAGGTGGCTCAGGCGGTGGCGGAAGT**TCCTATGAGCTGACWCAG** |
| 5' GS Vλ4/5 | AGCGGCGGAGGTGGCTCAGGCGGTGGCGGAAGT**CAGCYTGTGCTGACTCA** |
| 5' GS Vλ6 | AGCGGCGGAGGTGGCTCAGGCGGTGGCGGAAGT**AATTTTATGCTGACTCAG** |
| 5' GS Vλ7/8 | AGCGGCGGAGGTGGCTCAGGCGGTGGCGGAAGT**CAGRCTGTGGTGACYCAG** |
| 3' phage-His-Jλ -1 | GATGGTGATGGTGGTGAGCTCTTCC**TAGGACGGTGACCTTGGTCCC** |
| 3' phage-His-Jλ -2 | GATGGTGATGGTGGTGAGCTCTTCC**TAGGACGGTCAGCTTGGTCCC** |
| 3' phage-His-Jλ -3 | GATGGTGATGGTGGTGAGCTCTTCC**GAGGACGGTCAGCTGGGTGCC** |
| **Igκ 1st PCR** | |
| 5' L Vκ1/2 | **ATGAGGSTCCCYGCTCAGCTGCTGG** |
| 5' L Vκ3 | **CTCTTCCTCCTGCTACTCTGGCTCCCAG** |
| 5' L Vκ4 | **ATTTCTCTGTTGCTCTGGATCTCTG** |
| 3' C κ 543 | **GTTTCTCGTAGTCTGCTTTGCTCA** |
| **Igκ 2nd PCR** | |
| 5' GS-all-Vκ1 | AGCGGCGGCGGCGGCTCTGGTGGTGGTGGATCC**GACATCCAGATGACCCAGTCTCC** |
| 5' GS –all-Vκ6 | AGCGGCGGCGGCGGCTCTGGTGGTGGTGGATCC**GAAATTGTGCTGACTCAGTCTCC** |
| 5' GS-all-Vκ2 | AGCGGCGGCGGCGGCTCTGGTGGTGGTGGATCC**GATGTTGTGATGACTCAGTCTCC** |
| 5' GS-all-Vκ3 | AGCGGCGGCGGCGGCTCTGGTGGTGGTGGATCC**GAAATTGTGTTGACGCAGTCTCC** |
| 5' GS-all-Vκ4 | AGCGGCGGCGGCGGCTCTGGTGGTGGTGGATCC**GACATCGTGATGACCCAGTCTCC** |
| 5' GS-all-Vκ5 | AGCGGCGGCGGCGGCTCTGGTGGTGGTGGATCC**GAAACGACACTCACGCAGTCTCC** |
| 3' phage-His-Jκ1 | GATGGTGATGGTGGTGAGCTCTTCC**TTTGATTTCCACCTTGGTCCC** |
| 3' phage-His-Jκ2 | GATGGTGATGGTGGTGAGCTCTTCC**TTTGATCTCCAGCTTGGTCCC** |
| 3' phage-His-Jκ3 | GATGGTGATGGTGGTGAGCTCTTCC**TTTGATATCCACTTTGGTCCC** |
| 3' phage-His-Jκ4 | GATGGTGATGGTGGTGAGCTCTTCC**TTTGATCTCCACCTTGGTCCC** |
| 3' phage-His-Jκ5 | GATGGTGATGGTGGTGAGCTCTTCC**TTTAATCTCCAGTCGTGTCCC** |

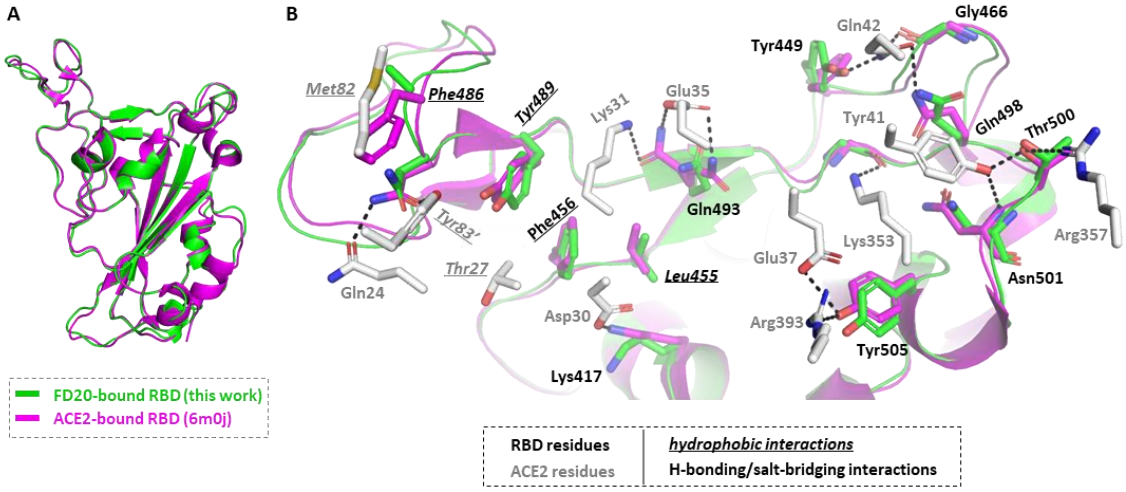
[a]Sense primers start with 5' and antisense primers start with 3'. [b]bold letters denote nucleotide sequences annealing to germline-specific regions and plain letters denote those for Gibson cloning.




**Appendix Fig S1. Identification of neutralizing antibodies.**

(**A**) Fifty-three scFv antibodies that bind to RBD on gel filtration were subjected to neutralization assays using SARS-CoV-2 pp and scFv at indicated concentrations.

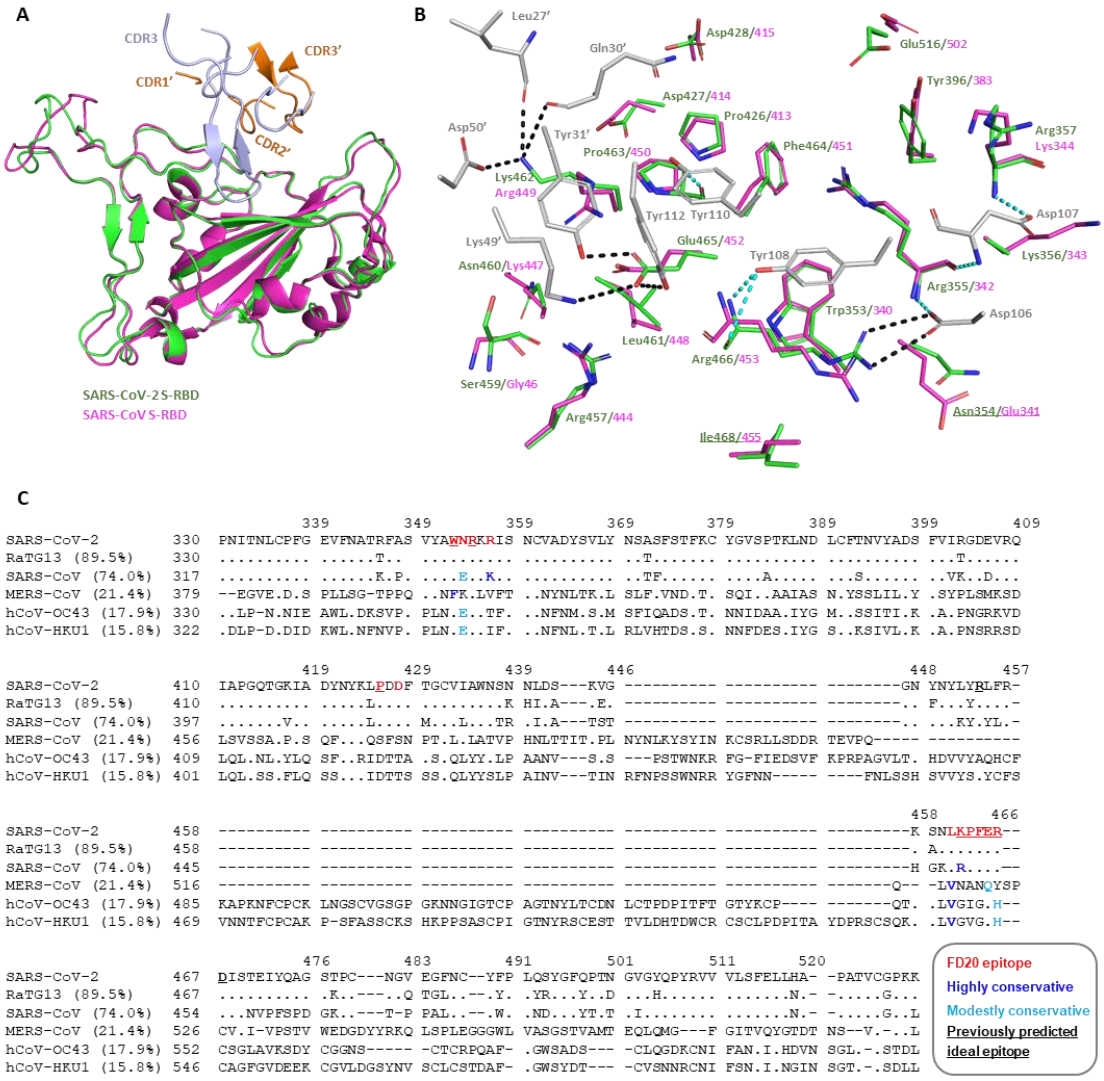
(**B**) Amino-acid sequence of scFD20. Complementarity-determining regions (CDRs) from the heavy chain (H-CDRs) and the light chain (L-CDRs) are underlined. A prime symbol denotes residues from the light chain. The mature protein contained amino acids GSSSQ at the N-terminus and SAGRAGEQKLISEEDLNSAVDHHHHHH at the C-terminus for biotinylation and affinity purification purposes.




**Appendix Fig S2. Comparison of RBD conformation between the FD20-bound form and the ACE2-bound form.**

(**A**) The overall similarity of the RBD structure between the FD20-bound form (green) and the ACE2-bound form (magenta). The Cα root mean square deviation of the two structures is 0.669 Å.

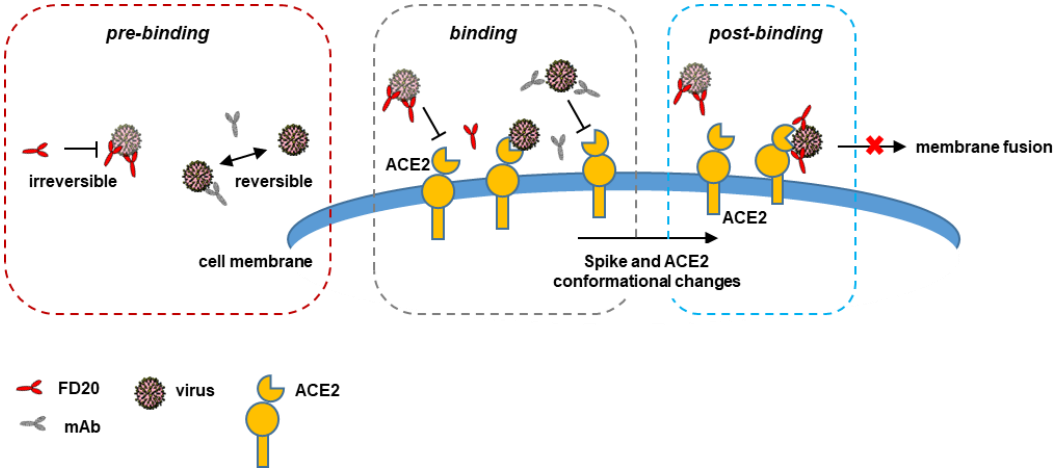
(**B**) The slight differences of the RBD structures between the FD20-bound form (green) and the ACE2-bound form (magenta). Residues from ACE2 at the receptor-binding motif (RBM) are shown as grey sticks and are also labeled with grey texts. RBM residues are labeled as black texts. Residues involved in the binding by hydrophobic interactions are labeled as underline and italic texts.




**Appendix Fig S3. Structural comparison of RBD-equivalent regions in other coronaviruses.**

(**A, B**) Aligning scFD20-RBD to the structure of SARS-CoV RBD in complex with a neutralizing antibody M396. The Cα root mean square deviation of the two structures is 0.669 Å. The similarity in the overall structure (**A**) and at the epitope region (**B**) is shown.

(**C**) The FD20 epitope (red) is conserved between SARS-CoV-2, RaTG13, and SARS-CoV. Dots indicate identical residues and dashes indicate gaps. Brackets indicate sequence identity between the aligned RBD and SARS-CoV-2 RBD. NCBI accession codes for the sequences are as follows. RaTG13, QHR63300.2; SARS-CoV, NC_004718.3; MERS-CoV, NC_019843.3; hCoV-OC43, NC_006213.1; hCoV-HKU1, NC_006577.2.




**Appendix Fig S4. Schematic mechanism diagram of antibodies that act on the *pre-binding*, *binding*, and *post-binding* steps.**

# Full wwPDB X-ray Structure Validation Report ⓘ

Sep 6, 2021 – 02:07 PM JST

| | | |
|---|---|---|
| PDB ID | : | 7CYV |
| Title | : | Crystal structure of FD20, a neutralizing single-chain variable fragment (scFv) in complex with SARS-CoV-2 Spike receptor-binding domain (RBD) |
| Authors | : | Li, Y.; Li, T.; Lai, Y.; Cai, H.; Yao, H.; Li, D. |
| Deposited on | : | 2020-09-04 |
| Resolution | : | 3.13 Å(reported) |

This is a Full wwPDB X-ray Structure Validation Report for a publicly released PDB entry.

We welcome your comments at *validation@mail.wwpdb.org*
A user guide is available at
https://www.wwpdb.org/validation/2017/XrayValidationReportHelp
with specific help available everywhere you see the ⓘ symbol.

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The following versions of software and data (see references ⓘ) were used in the production of this report:

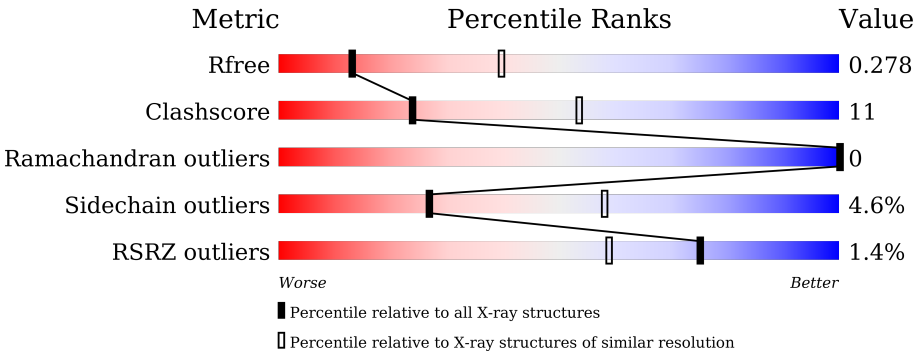
| | | |
|---|---|---|
| MolProbity | : | 4.02b-467 |
| Mogul | : | 1.8.5 (274361), CSD as541be (2020) |
| Xtriage (Phenix) | : | 1.13 |
| EDS | : | 2.23.1 |
| Percentile statistics | : | 20191225.v01 (using entries in the PDB archive December 25th 2019) |
| Refmac | : | 5.8.0158 |
| CCP4 | : | 7.0.044 (Gargrove) |
| Ideal geometry (proteins) | : | Engh & Huber (2001) |
| Ideal geometry (DNA, RNA) | : | Parkinson et al. (1996) |
| Validation Pipeline (wwPDB-VP) | : | 2.23.1 |

# 1 Overall quality at a glance (i)

The following experimental techniques were used to determine the structure:
*X-RAY DIFFRACTION*

The reported resolution of this entry is 3.13 Å.

Percentile scores (ranging between 0-100) for global validation metrics of the entry are shown in the following graphic. The table shows the number of entries on which the scores are based.




| Metric | Whole archive (#Entries) | Similar resolution (#Entries, resolution range(Å)) |
|---|---|---|
| $R_{free}$ | 130704 | 1626 (3.18-3.10) |
| Clashscore | 141614 | 1735 (3.18-3.10) |
| Ramachandran outliers | 138981 | 1677 (3.18-3.10) |
| Sidechain outliers | 138945 | 1677 (3.18-3.10) |
| RSRZ outliers | 127900 | 1588 (3.18-3.10) |

The table below summarises the geometric issues observed across the polymeric chains and their fit to the electron density. The red, orange, yellow and green segments of the lower bar indicate the fraction of residues that contain outliers for >=3, 2, 1 and 0 types of geometric quality criteria respectively. A grey segment represents the fraction of residues that are not modelled. The numeric value for each fraction is indicated below the corresponding segment, with a dot representing fractions <=5% The upper red bar (where present) indicates the fraction of residues that have poor fit to the electron density. The numeric value is given above the bar.

| Mol | Chain | Length | Quality of chain |
|---|---|---|---|
| 1 | H | 281 | 2%<br>62%<br>22%<br>16% |
| 2 | B | 213 | 69%<br>20%<br>9% |
| 3 | A | 5 | 60%<br>40% |

## 2 Entry composition (i)

There are 3 unique types of molecules in this entry. The entry contains 3225 atoms, of which 0 are hydrogens and 0 are deuteriums.

In the tables below, the ZeroOcc column contains the number of atoms modelled with zero occupancy, the AltConf column contains the number of residues with at least one atom in alternate conformation and the Trace column contains the number of residues modelled with at most 2 atoms.

- Molecule 1 is a protein called The heavy chain variable region of the scFv FD20,The light chain variable region of the scFv FD20.

| Mol | Chain | Residues | Atoms | | | | | ZeroOcc | AltConf | Trace |
|---|---|---|---|---|---|---|---|---|---|---|
| | | | Total | C | N | O | S | | | |
| 1 | H | 236 | 1700 | 1085 | 275 | 332 | 8 | 0 | 0 | 0 |

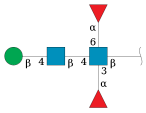
- Molecule 2 is a protein called Spike protein S1.

| Mol | Chain | Residues | Atoms | | | | | | ZeroOcc | AltConf | Trace |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | Total | C | N | O | P | S | | | |
| 2 | B | 193 | 1466 | 937 | 248 | 272 | 1 | 8 | 0 | 0 | 0 |

There are 11 discrepancies between the modelled and reference sequences:

| Chain | Residue | Modelled | Actual | Comment | Reference |
|---|---|---|---|---|---|
| B | 327 | ALA | - | expression tag | UNP P0DTC2 |
| B | 328 | GLY | - | expression tag | UNP P0DTC2 |
| B | 329 | SER | - | expression tag | UNP P0DTC2 |
| B | 532 | GLY | - | expression tag | UNP P0DTC2 |
| B | 533 | THR | - | expression tag | UNP P0DTC2 |
| B | 534 | LEU | - | expression tag | UNP P0DTC2 |
| B | 535 | GLU | - | expression tag | UNP P0DTC2 |
| B | 536 | VAL | - | expression tag | UNP P0DTC2 |
| B | 537 | LEU | - | expression tag | UNP P0DTC2 |
| B | 538 | PHE | - | expression tag | UNP P0DTC2 |
| B | 539 | GLN | - | expression tag | UNP P0DTC2 |

- Molecule 3 is an oligosaccharide called beta-D-mannopyranose-(1-4)-2-acetamido-2-deoxy-beta-D-glucopyranose-(1-4)-[alpha-L-fucopyranose-(1-3)][alpha-L-fucopyranose-(1-6)]2-acetamido-2-deoxy-beta-D-glucopyranose.



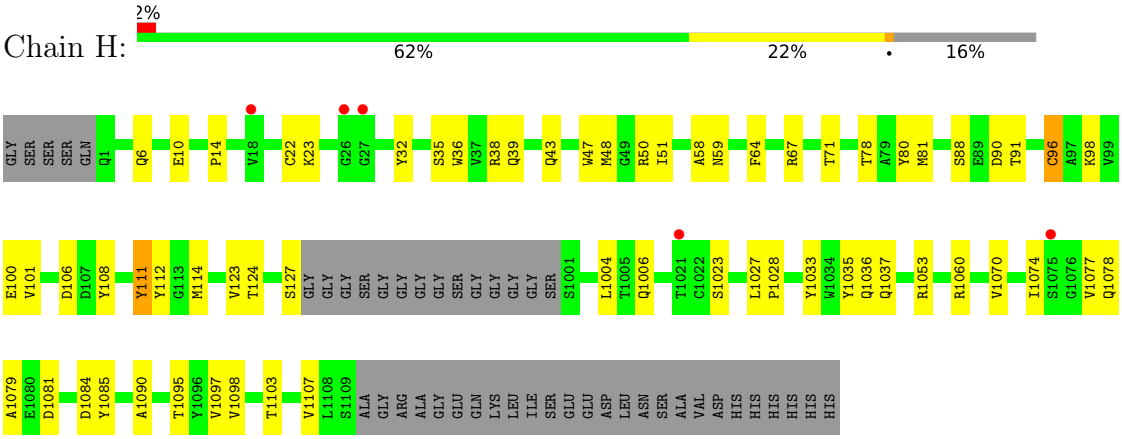
| Mol | Chain | Residues | Atoms | | | | ZeroOcc | AltConf | Trace |
|---|---|---|---|---|---|---|---|---|---|
| 3 | A | 5 | Total | C | N | O | 0 | 0 | 0 |
| | | | 59 | 34 | 2 | 23 | | | |

# 3 Residue-property plots (i)

These plots are drawn for all protein, RNA, DNA and oligosaccharide chains in the entry. The first graphic for a chain summarises the proportions of the various outlier classes displayed in the second graphic. The second graphic shows the sequence view annotated by issues in geometry and electron density. Residues are color-coded according to the number of geometric quality criteria for which they contain at least one outlier: green = 0, yellow = 1, orange = 2 and red = 3 or more. A red dot above a residue indicates a poor fit to the electron density (RSRZ > 2). Stretches of 2 or more consecutive residues without any outlier are shown as a green connector. Residues present in the sample, but not in the model, are shown in grey.

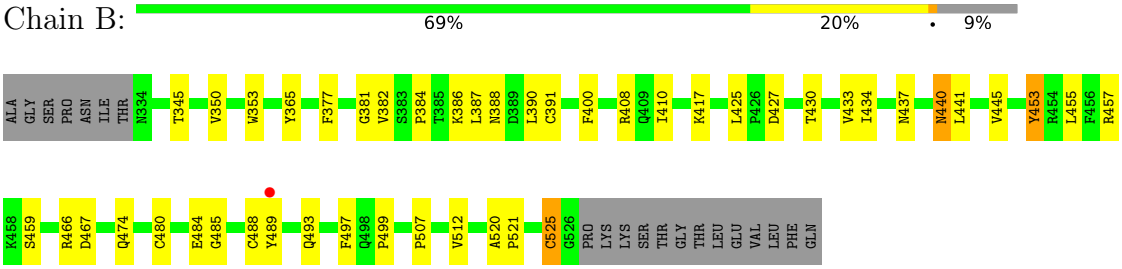
• Molecule 1: The heavy chain variable region of the scFv FD20,The light chain variable region of the scFv FD20

Chain H:




• Molecule 2: Spike protein S1

Chain B:




• Molecule 3: beta-D-mannopyranose-(1-4)-2-acetamido-2-deoxy-beta-D-glucopyranose-(1-4)-[alpha-L-fucopyranose-(1-3)][alpha-L-fucopyranose-(1-6)]2-acetamido-2-deoxy-beta-D-glucopyranose

Chain A:

# 4 Data and refinement statistics (i)

| Property | Value | Source |
|---|---|---|
| Space group | C 1 2 1 | Depositor |
| Cell constants<br>a, b, c, $\alpha$, $\beta$, $\gamma$ | 206.96Å 57.92Å 47.21Å<br>90.00° 100.43° 90.00° | Depositor |
| Resolution (Å) | 46.43 – 3.13<br>46.43 – 3.13 | Depositor<br>EDS |
| % Data completeness<br>(in resolution range) | 98.9 (46.43-3.13)<br>99.2 (46.43-3.13) | Depositor<br>EDS |
| $R_{merge}$ | 0.27 | Depositor |
| $R_{sym}$ | (Not available) | Depositor |
| $< I/\sigma(I) >$ [1] | 1.28 (at 3.12Å) | Xtriage |
| Refinement program | PHENIX 1.17.1_3660 | Depositor |
| R, $R_{free}$ | 0.252 , 0.276<br>0.253 , 0.278 | Depositor<br>DCC |
| $R_{free}$ test set | 463 reflections (4.73%) | wwPDB-VP |
| Wilson B-factor (Å$^2$) | 68.8 | Xtriage |
| Anisotropy | 0.409 | Xtriage |
| Bulk solvent $k_{sol}$(e/Å$^3$), $B_{sol}$(Å$^2$) | 0.30 , 35.9 | EDS |
| L-test for twinning[2] | $< \|L\| > = 0.48$, $< L^2 > = 0.31$ | Xtriage |
| Estimated twinning fraction | 0.030 for -h-2*l,-k,l | Xtriage |
| $F_o$,$F_c$ correlation | 0.90 | EDS |
| Total number of atoms | 3225 | wwPDB-VP |
| Average B, all atoms (Å$^2$) | 61.0 | wwPDB-VP |

Xtriage's analysis on translational NCS is as follows: *The largest off-origin peak in the Patterson function is 7.08% of the height of the origin peak. No significant pseudotranslation is detected.*

[1] Intensities estimated from amplitudes.

[2] Theoretical values of $< |L| >$, $< L^2 >$ for acentric reflections are 0.5, 0.333 respectively for untwinned datasets, and 0.375, 0.2 for perfectly twinned datasets.

# 5 Model quality

## 5.1 Standard geometry

Bond lengths and bond angles in the following residue types are not validated in this section: BMA, TPO, FUC, NAG

The Z score for a bond length (or angle) is the number of standard deviations the observed value is removed from the expected value. A bond length (or angle) with $|Z| > 5$ is considered an outlier worth inspection. RMSZ is the root-mean-square of all Z scores of the bond lengths (or angles).

| Mol | Chain | Bond lengths | | Bond angles | |
|---|---|---|---|---|---|
| | | RMSZ | $\#\|Z\| > 5$ | RMSZ | $\#\|Z\| > 5$ |
| 1 | H | 0.28 | 0/1740 | 0.50 | 0/2383 |
| 2 | B | 0.29 | 0/1495 | 0.52 | 0/2040 |
| All | All | 0.29 | 0/3235 | 0.51 | 0/4423 |

There are no bond length outliers.

There are no bond angle outliers.

There are no chirality outliers.

There are no planarity outliers.

## 5.2 Too-close contacts

In the following table, the Non-H and H(model) columns list the number of non-hydrogen atoms and hydrogen atoms in the chain respectively. The H(added) column lists the number of hydrogen atoms added and optimized by MolProbity. The Clashes column lists the number of clashes within the asymmetric unit, whereas Symm-Clashes lists symmetry-related clashes.

| Mol | Chain | Non-H | H(model) | H(added) | Clashes | Symm-Clashes |
|---|---|---|---|---|---|---|
| 1 | H | 1700 | 0 | 1560 | 42 | 0 |
| 2 | B | 1466 | 0 | 1315 | 25 | 0 |
| 3 | A | 59 | 0 | 52 | 1 | 0 |
| All | All | 3225 | 0 | 2927 | 68 | 0 |

The all-atom clashscore is defined as the number of clashes found per 1000 atoms (including hydrogen atoms). The all-atom clashscore for this structure is 11.

All (68) close contacts within the same asymmetric unit are listed below, sorted by their clash magnitude.

| Atom-1 | Atom-2 | Interatomic distance (Å) | Clash overlap (Å) |
|---|---|---|---|
| 3:A:2:NAG:H61 | 3:A:4:FUC:H63 | 1.62 | 0.81 |
| 2:B:391:CYS:HA | 2:B:525:CYS:HB3 | 1.69 | 0.73 |
| 1:H:38:ARG:NH1 | 1:H:90:ASP:OD1 | 2.22 | 0.71 |
| 1:H:39:GLN:OE1 | 1:H:1037:GLN:NE2 | 2.22 | 0.71 |
| 1:H:47:TRP:CD2 | 1:H:1097:VAL:HG22 | 2.26 | 0.71 |
| 1:H:1090:ALA:HA | 1:H:1097:VAL:HA | 1.75 | 0.67 |
| 2:B:365:TYR:HD2 | 2:B:388:ASN:HA | 1.60 | 0.67 |
| 1:H:48:MET:HG2 | 1:H:64:PHE:HE2 | 1.60 | 0.64 |
| 1:H:51:ILE:HG13 | 1:H:58:ALA:HB2 | 1.82 | 0.62 |
| 2:B:417:LYS:HD2 | 2:B:455:LEU:HD12 | 1.82 | 0.62 |
| 1:H:1060:ARG:NH1 | 1:H:1081:ASP:OD2 | 2.33 | 0.61 |
| 1:H:39:GLN:NE2 | 1:H:43:GLN:O | 2.31 | 0.61 |
| 1:H:1074:ILE:HB | 1:H:1077:VAL:HG12 | 1.84 | 0.60 |
| 1:H:1079:ALA:HA | 1:H:1107:VAL:HG11 | 1.84 | 0.59 |
| 2:B:484:GLU:OE2 | 2:B:484:GLU:N | 2.33 | 0.59 |
| 2:B:433:VAL:HG22 | 2:B:512:VAL:HG13 | 1.86 | 0.57 |
| 1:H:47:TRP:HZ2 | 1:H:50:ARG:HG2 | 1.69 | 0.57 |
| 1:H:67:ARG:NH2 | 1:H:90:ASP:OD2 | 2.38 | 0.56 |
| 1:H:35:SER:OG | 1:H:47:TRP:NE1 | 2.37 | 0.56 |
| 1:H:6:GLN:HG2 | 1:H:22:CYS:HB3 | 1.89 | 0.54 |
| 1:H:59:ASN:ND2 | 1:H:1095:THR:O | 2.40 | 0.54 |
| 1:H:100:GLU:O | 1:H:112:TYR:HB2 | 2.06 | 0.54 |
| 2:B:365:TYR:CD2 | 2:B:388:ASN:HA | 2.41 | 0.54 |
| 1:H:1036:GLN:HB2 | 1:H:1085:TYR:CE1 | 2.43 | 0.53 |
| 1:H:14:PRO:HG2 | 1:H:127:SER:HB2 | 1.90 | 0.53 |
| 1:H:50:ARG:HD2 | 1:H:111:TYR:CZ | 2.44 | 0.52 |
| 2:B:386:LYS:O | 2:B:390:LEU:HD23 | 2.10 | 0.52 |
| 2:B:457:ARG:HD3 | 2:B:459:SER:O | 2.10 | 0.52 |
| 1:H:1006:GLN:NE2 | 1:H:1085:TYR:O | 2.42 | 0.52 |
| 2:B:497:PHE:CE2 | 2:B:507:PRO:HB3 | 2.45 | 0.52 |
| 1:H:6:GLN:HA | 1:H:22:CYS:HA | 1.93 | 0.51 |
| 1:H:106:ASP:O | 1:H:108:TYR:N | 2.43 | 0.51 |
| 1:H:50:ARG:HD2 | 1:H:111:TYR:CE1 | 2.46 | 0.51 |
| 2:B:520:ALA:HB1 | 2:B:521:PRO:HD2 | 1.92 | 0.50 |
| 2:B:485:GLY:N | 2:B:488:CYS:O | 2.42 | 0.50 |
| 1:H:47:TRP:CG | 1:H:1097:VAL:HG22 | 2.46 | 0.50 |
| 1:H:112:TYR:HA | 1:H:1033:TYR:OH | 2.12 | 0.50 |
| 2:B:391:CYS:CA | 2:B:525:CYS:HB3 | 2.34 | 0.50 |
| 1:H:23:LYS:HG3 | 1:H:78:THR:HG22 | 1.94 | 0.49 |
| 1:H:1027:LEU:HD23 | 1:H:1070:VAL:HG23 | 1.93 | 0.49 |
| 2:B:474:GLN:HA | 2:B:480:CYS:SG | 2.52 | 0.49 |
| 2:B:381:GLY:HA3 | 2:B:430:THR:HA | 1.94 | 0.48 |

*Continued on next page...*

*Continued from previous page...*

| Atom-1 | Atom-2 | Interatomic distance (Å) | Clash overlap (Å) |
|---|---|---|---|
| 1:H:71:THR:HG23 | 1:H:80:TYR:HB2 | 1.95 | 0.48 |
| 1:H:10:GLU:HB2 | 1:H:123:VAL:HG22 | 1.96 | 0.47 |
| 2:B:453:TYR:CE1 | 2:B:493:GLN:HB3 | 2.49 | 0.47 |
| 2:B:457:ARG:HH11 | 2:B:467:ASP:HB2 | 1.79 | 0.47 |
| 1:H:35:SER:HG | 1:H:47:TRP:HE1 | 1.59 | 0.47 |
| 2:B:350:VAL:HA | 2:B:400:PHE:HB2 | 1.97 | 0.46 |
| 1:H:1027:LEU:N | 1:H:1028:PRO:HD2 | 2.31 | 0.46 |
| 1:H:36:TRP:CE2 | 1:H:81:MET:HB2 | 2.52 | 0.45 |
| 1:H:101:VAL:HG23 | 1:H:111:TYR:HA | 1.98 | 0.45 |
| 1:H:1006:GLN:HG2 | 1:H:1103:THR:OG1 | 2.17 | 0.45 |
| 2:B:377:PHE:CD2 | 2:B:434:ILE:HG12 | 2.52 | 0.45 |
| 1:H:98:LYS:HB3 | 1:H:98:LYS:HE3 | 1.76 | 0.44 |
| 2:B:440:ASN:OD1 | 2:B:441:LEU:HG | 2.18 | 0.44 |
| 2:B:353:TRP:NE1 | 2:B:466:ARG:HB2 | 2.33 | 0.43 |
| 2:B:410:ILE:O | 2:B:425:LEU:HD13 | 2.18 | 0.43 |
| 1:H:1004:LEU:HG | 1:H:1098:VAL:HG13 | 2.00 | 0.43 |
| 1:H:32:TYR:CD1 | 1:H:98:LYS:HD3 | 2.55 | 0.42 |
| 1:H:36:TRP:CH2 | 1:H:96:CYS:HB2 | 2.55 | 0.42 |
| 1:H:91:THR:HG23 | 1:H:124:THR:HA | 2.02 | 0.41 |
| 2:B:384:PRO:HA | 2:B:387:LEU:HB2 | 2.01 | 0.41 |
| 2:B:382:VAL:HG21 | 2:B:390:LEU:HG | 2.01 | 0.41 |
| 1:H:114:MET:O | 1:H:1035:TYR:HE1 | 2.04 | 0.41 |
| 2:B:445:VAL:HG22 | 2:B:499:PRO:HG2 | 2.03 | 0.41 |
| 2:B:497:PHE:CZ | 2:B:507:PRO:HB3 | 2.54 | 0.41 |
| 1:H:35:SER:HA | 1:H:50:ARG:HA | 2.03 | 0.40 |
| 1:H:36:TRP:HB3 | 1:H:48:MET:HE2 | 2.02 | 0.40 |

There are no symmetry-related clashes.

## 5.3 Torsion angles (i)

### 5.3.1 Protein backbone (i)

In the following table, the Percentiles column shows the percent Ramachandran outliers of the chain as a percentile score with respect to all X-ray entries followed by that with respect to entries of similar resolution.

The Analysed column shows the number of residues for which the backbone conformation was analysed, and the total number of residues.

| Mol | Chain | Analysed | Favoured | Allowed | Outliers | Percentiles | |
|---|---|---|---|---|---|---|---|
| 1 | H | 232/281 (83%) | 224 (97%) | 8 (3%) | 0 | 100 | 100 |
| 2 | B | 190/213 (89%) | 181 (95%) | 9 (5%) | 0 | 100 | 100 |
| All | All | 422/494 (85%) | 405 (96%) | 17 (4%) | 0 | 100 | 100 |

There are no Ramachandran outliers to report.

### 5.3.2 Protein sidechains (i)

In the following table, the Percentiles column shows the percent sidechain outliers of the chain as a percentile score with respect to all X-ray entries followed by that with respect to entries of similar resolution.

The Analysed column shows the number of residues for which the sidechain conformation was analysed, and the total number of residues.

| Mol | Chain | Analysed | Rotameric | Outliers | Percentiles | |
|---|---|---|---|---|---|---|
| 1 | H | 164/222 (74%) | 157 (96%) | 7 (4%) | 29 | 60 |
| 2 | B | 143/182 (79%) | 136 (95%) | 7 (5%) | 25 | 56 |
| All | All | 307/404 (76%) | 293 (95%) | 14 (5%) | 27 | 58 |

All (14) residues with a non-rotameric sidechain are listed below:

| Mol | Chain | Res | Type |
|---|---|---|---|
| 1 | H | 88 | SER |
| 1 | H | 96 | CYS |
| 1 | H | 111 | TYR |
| 1 | H | 1023 | SER |
| 1 | H | 1053 | ARG |
| 1 | H | 1078 | GLN |
| 1 | H | 1084 | ASP |
| 2 | B | 408 | ARG |
| 2 | B | 427 | ASP |
| 2 | B | 437 | ASN |
| 2 | B | 440 | ASN |
| 2 | B | 453 | TYR |
| 2 | B | 489 | TYR |
| 2 | B | 525 | CYS |

Sometimes sidechains can be flipped to improve hydrogen bonding and reduce clashes. All (5) such sidechains are listed below:

| Mol | Chain | Res | Type |
|---|---|---|---|
| 1 | H | 65 | GLN |
| 1 | H | 1036 | GLN |
| 2 | B | 394 | ASN |
| 2 | B | 409 | GLN |
| 2 | B | 437 | ASN |

### 5.3.3 RNA (i)

There are no RNA molecules in this entry.

## 5.4 Non-standard residues in protein, DNA, RNA chains (i)

1 non-standard protein/DNA/RNA residue is modelled in this entry.

In the following table, the Counts columns list the number of bonds (or angles) for which Mogul statistics could be retrieved, the number of bonds (or angles) that are observed in the model and the number of bonds (or angles) that are defined in the Chemical Component Dictionary. The Link column lists molecule types, if any, to which the group is linked. The Z score for a bond length (or angle) is the number of standard deviations the observed value is removed from the expected value. A bond length (or angle) with $|Z| > 2$ is considered an outlier worth inspection. RMSZ is the root-mean-square of all Z scores of the bond lengths (or angles).

| Mol | Type | Chain | Res | Link | Bond lengths | | | Bond angles | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | | | | | Counts | RMSZ | $\#\|Z\| > 2$ | Counts | RMSZ | $\#\|Z\| > 2$ |
| 2 | TPO | B | 345 | 2 | 8,10,11 | 1.18 | 0 | 10,14,16 | 1.13 | 1 (10%) |

In the following table, the Chirals column lists the number of chiral outliers, the number of chiral centers analysed, the number of these observed in the model and the number defined in the Chemical Component Dictionary. Similar counts are reported in the Torsion and Rings columns. '-' means no outliers of that kind were identified.

| Mol | Type | Chain | Res | Link | Chirals | Torsions | Rings |
|---|---|---|---|---|---|---|---|
| 2 | TPO | B | 345 | 2 | - | 4/9/11/13 | - |

There are no bond length outliers.

All (1) bond angle outliers are listed below:

| Mol | Chain | Res | Type | Atoms | Z | Observed($^o$) | Ideal($^o$) |
|---|---|---|---|---|---|---|---|
| 2 | B | 345 | TPO | O3P-P-OG1 | 2.36 | 116.57 | 105.99 |

There are no chirality outliers.

All (4) torsion outliers are listed below:

| Mol | Chain | Res | Type | Atoms |
|---|---|---|---|---|
| 2 | B | 345 | TPO | O-C-CA-CB |
| 2 | B | 345 | TPO | CG2-CB-OG1-P |
| 2 | B | 345 | TPO | CB-OG1-P-O1P |
| 2 | B | 345 | TPO | CB-OG1-P-O3P |

There are no ring outliers.

No monomer is involved in short contacts.

## 5.5 Carbohydrates (i)

5 monosaccharides are modelled in this entry.

In the following table, the Counts columns list the number of bonds (or angles) for which Mogul statistics could be retrieved, the number of bonds (or angles) that are observed in the model and the number of bonds (or angles) that are defined in the Chemical Component Dictionary. The Link column lists molecule types, if any, to which the group is linked. The Z score for a bond length (or angle) is the number of standard deviations the observed value is removed from the expected value. A bond length (or angle) with $|Z| > 2$ is considered an outlier worth inspection. RMSZ is the root-mean-square of all Z scores of the bond lengths (or angles).

| Mol | Type | Chain | Res | Link | Bond lengths | | | Bond angles | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | | | | | Counts | RMSZ | $\#\|Z\| > 2$ | Counts | RMSZ | $\#\|Z\| > 2$ |
| 3 | NAG | A | 1 | 3,2 | 14,14,15 | 0.25 | 0 | 17,19,21 | 1.80 | 4 (23%) |
| 3 | NAG | A | 2 | 3 | 14,14,15 | 0.64 | 1 (7%) | 17,19,21 | 0.86 | 0 |
| 3 | BMA | A | 3 | 3 | 11,11,12 | 0.73 | 1 (9%) | 15,15,17 | 1.21 | 1 (6%) |
| 3 | FUC | A | 4 | 3 | 10,10,11 | 1.38 | 1 (10%) | 14,14,16 | 1.99 | 4 (28%) |
| 3 | FUC | A | 5 | 3 | 10,10,11 | 0.69 | 0 | 14,14,16 | 0.99 | 1 (7%) |

In the following table, the Chirals column lists the number of chiral outliers, the number of chiral centers analysed, the number of these observed in the model and the number defined in the Chemical Component Dictionary. Similar counts are reported in the Torsion and Rings columns. '-' means no outliers of that kind were identified.

| Mol | Type | Chain | Res | Link | Chirals | Torsions | Rings |
|---|---|---|---|---|---|---|---|
| 3 | NAG | A | 1 | 3,2 | - | 1/6/23/26 | 0/1/1/1 |
| 3 | NAG | A | 2 | 3 | - | 0/6/23/26 | 0/1/1/1 |
| 3 | BMA | A | 3 | 3 | - | 2/2/19/22 | 0/1/1/1 |
| 3 | FUC | A | 4 | 3 | - | - | 0/1/1/1 |
| 3 | FUC | A | 5 | 3 | - | - | 0/1/1/1 |

All (3) bond length outliers are listed below:

| Mol | Chain | Res | Type | Atoms | Z | Observed(Å) | Ideal(Å) |
|---|---|---|---|---|---|---|---|
| 3 | A | 4 | FUC | C1-C2 | 3.55 | 1.60 | 1.52 |
| 3 | A | 3 | BMA | C1-C2 | 2.31 | 1.57 | 1.52 |
| 3 | A | 2 | NAG | C1-C2 | 2.08 | 1.55 | 1.52 |

All (10) bond angle outliers are listed below:

| Mol | Chain | Res | Type | Atoms | Z | Observed($^o$) | Ideal($^o$) |
|---|---|---|---|---|---|---|---|
| 3 | A | 4 | FUC | O5-C1-C2 | 4.01 | 116.96 | 110.77 |
| 3 | A | 1 | NAG | O3-C3-C4 | 3.92 | 119.41 | 110.35 |
| 3 | A | 1 | NAG | C1-O5-C5 | 3.89 | 117.46 | 112.19 |
| 3 | A | 4 | FUC | C1-C2-C3 | 3.35 | 113.78 | 109.67 |
| 3 | A | 4 | FUC | C1-O5-C5 | 3.20 | 120.04 | 112.78 |
| 3 | A | 3 | BMA | C1-O5-C5 | 2.74 | 115.91 | 112.19 |
| 3 | A | 4 | FUC | O5-C5-C4 | 2.54 | 114.08 | 109.52 |
| 3 | A | 1 | NAG | C4-C3-C2 | -2.33 | 107.60 | 111.02 |
| 3 | A | 1 | NAG | C2-N2-C7 | 2.15 | 125.97 | 122.90 |
| 3 | A | 5 | FUC | C1-O5-C5 | 2.06 | 117.45 | 112.78 |

There are no chirality outliers.

All (3) torsion outliers are listed below:

| Mol | Chain | Res | Type | Atoms |
|---|---|---|---|---|
| 3 | A | 3 | BMA | O5-C5-C6-O6 |
| 3 | A | 3 | BMA | C4-C5-C6-O6 |
| 3 | A | 1 | NAG | C3-C2-N2-C7 |

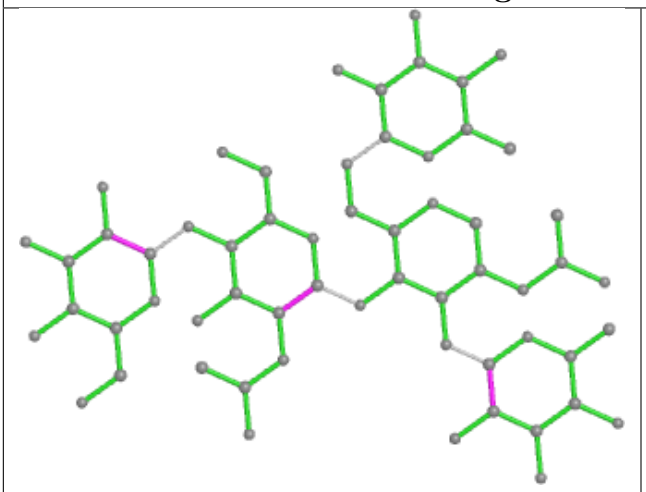
There are no ring outliers.

2 monomers are involved in 1 short contact:

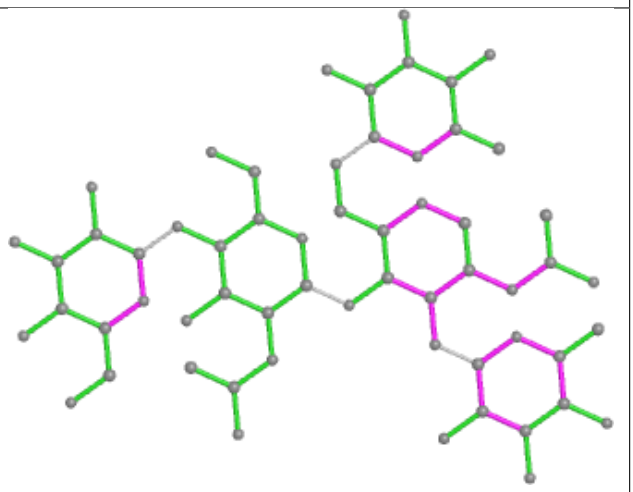
| Mol | Chain | Res | Type | Clashes | Symm-Clashes |
|---|---|---|---|---|---|
| 3 | A | 2 | NAG | 1 | 0 |
| 3 | A | 4 | FUC | 1 | 0 |

The following is a two-dimensional graphical depiction of Mogul quality analysis of bond lengths, bond angles, torsion angles, and ring geometry for oligosaccharide.

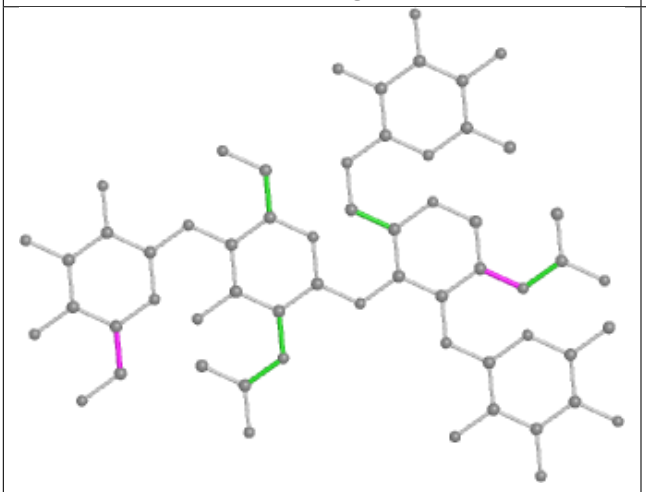
**Oligosaccharide Chain A**



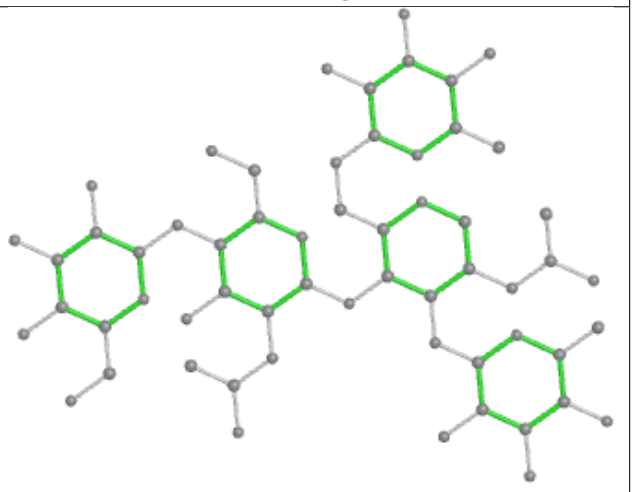
**Bond lengths**



**Bond angles**



**Torsions**



**Rings**

## 5.6 Ligand geometry (i)

There are no ligands in this entry.

## 5.7 Other polymers (i)

There are no such residues in this entry.

## 5.8 Polymer linkage issues (i)

There are no chain breaks in this entry.

# 6 Fit of model and data

## 6.1 Protein, DNA and RNA chains

In the following table, the column labelled '#RSRZ> 2' contains the number (and percentage) of RSRZ outliers, followed by percent RSRZ outliers for the chain as percentile scores relative to all X-ray entries and entries of similar resolution. The OWAB column contains the minimum, median, $95^{th}$ percentile and maximum values of the occupancy-weighted average B-factor per residue. The column labelled 'Q< 0.9' lists the number of (and percentage) of residues with an average occupancy less than 0.9.

| Mol | Chain | Analysed | <RSRZ> | #RSRZ>2 | | | OWAB(Å$^2$) | Q<0.9 |
|---|---|---|---|---|---|---|---|---|
| 1 | H | 236/281 (83%) | 0.04 | 5 (2%) | 63 | 44 | 45, 61, 80, 96 | 1 (0%) |
| 2 | B | 192/213 (90%) | -0.03 | 1 (0%) | 91 | 83 | 40, 56, 101, 121 | 0 |
| All | All | 428/494 (86%) | 0.01 | 6 (1%) | 75 | 59 | 40, 60, 94, 121 | 1 (0%) |

All (6) RSRZ outliers are listed below:

| Mol | Chain | Res | Type | RSRZ |
|---|---|---|---|---|
| 1 | H | 26 | GLY | 2.5 |
| 2 | B | 489 | TYR | 2.3 |
| 1 | H | 27 | GLY | 2.3 |
| 1 | H | 18 | VAL | 2.3 |
| 1 | H | 1021 | THR | 2.3 |
| 1 | H | 1075 | SER | 2.1 |

## 6.2 Non-standard residues in protein, DNA, RNA chains

In the following table, the Atoms column lists the number of modelled atoms in the group and the number defined in the chemical component dictionary. The B-factors column lists the minimum, median, $95^{th}$ percentile and maximum values of B factors of atoms in the group. The column labelled 'Q< 0.9' lists the number of atoms with occupancy less than 0.9.

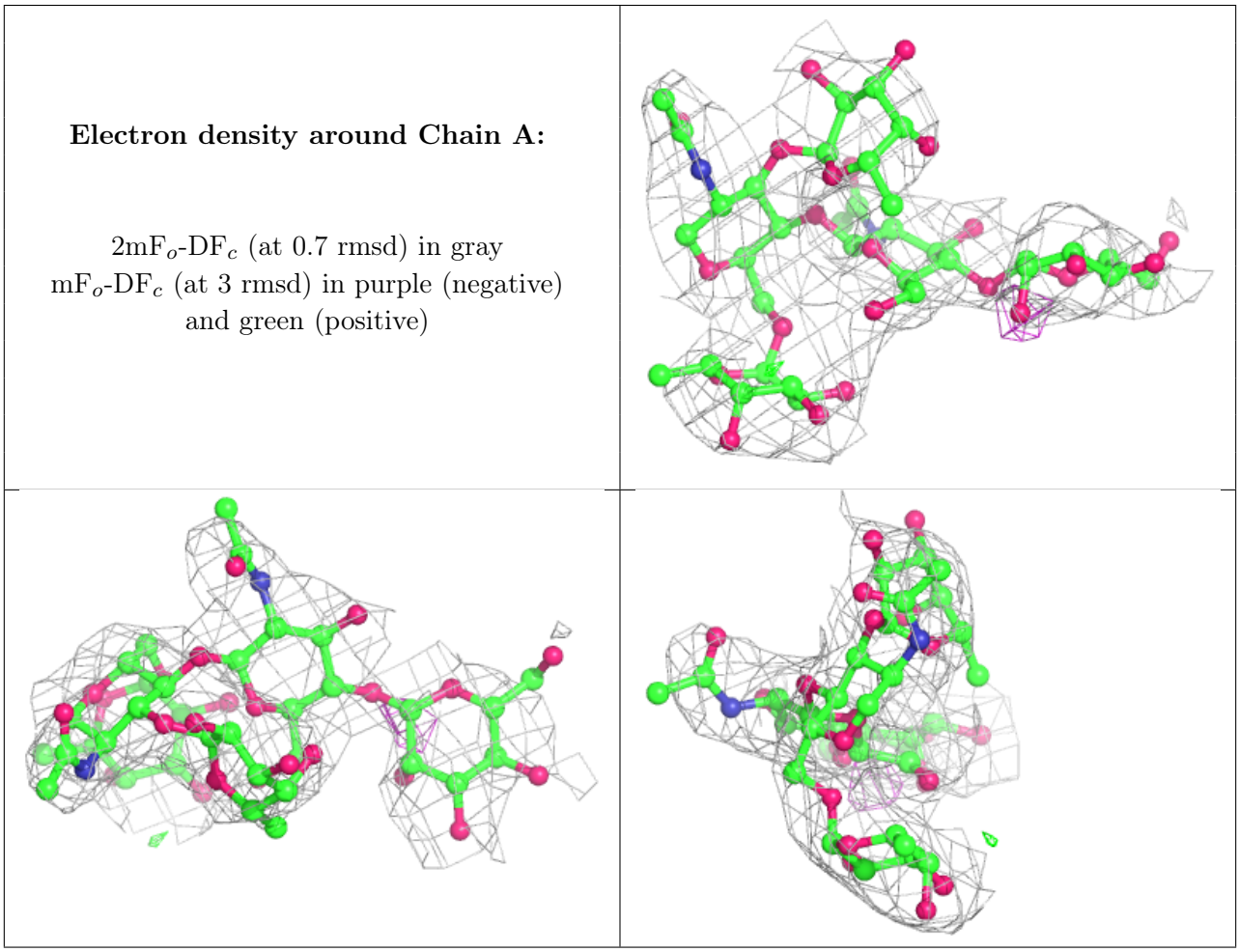
| Mol | Type | Chain | Res | Atoms | RSCC | RSR | B-factors(Å$^2$) | Q<0.9 |
|---|---|---|---|---|---|---|---|---|
| 2 | TPO | B | 345 | 11/12 | 0.83 | 0.22 | 57,57,57,57 | 0 |

## 6.3 Carbohydrates

In the following table, the Atoms column lists the number of modelled atoms in the group and the number defined in the chemical component dictionary. The B-factors column lists the minimum, median, $95^{th}$ percentile and maximum values of B factors of atoms in the group. The column labelled 'Q< 0.9' lists the number of atoms with occupancy less than 0.9.

| Mol | Type | Chain | Res | Atoms | RSCC | RSR | B-factors(Å$^2$) | Q<0.9 |
|---|---|---|---|---|---|---|---|---|
| 3 | BMA | A | 3 | 11/12 | 0.60 | 0.33 | 98,98,98,98 | 0 |
| 3 | NAG | A | 2 | 14/15 | 0.82 | 0.21 | 85,85,85,85 | 0 |
| 3 | FUC | A | 4 | 10/11 | 0.82 | 0.29 | 86,86,86,86 | 0 |
| 3 | NAG | A | 1 | 14/15 | 0.87 | 0.22 | 76,76,76,76 | 0 |
| 3 | FUC | A | 5 | 10/11 | 0.87 | 0.22 | 77,77,77,77 | 0 |

The following is a graphical depiction of the model fit to experimental electron density for oligosaccharide. Each fit is shown from different orientation to approximate a three-dimensional view.




## 6.4 Ligands

There are no ligands in this entry.

## 6.5 Other polymers

There are no such residues in this entry.