Full wwPDB X-ray Structure Validation Report ⓘ

May 13, 2020 – 06:09 am BST

PDB ID	:	6NMV
Title	:	Non-Blocking Fab 218 anti-SIRP-alpha antibody in complex with SIRP-alpha Variant 1
Authors	:	Wibowo, A.S.; Carter, J.J.; Sim, J.
Deposited on	:	2019-01-11
Resolution	:	2.61 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.

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

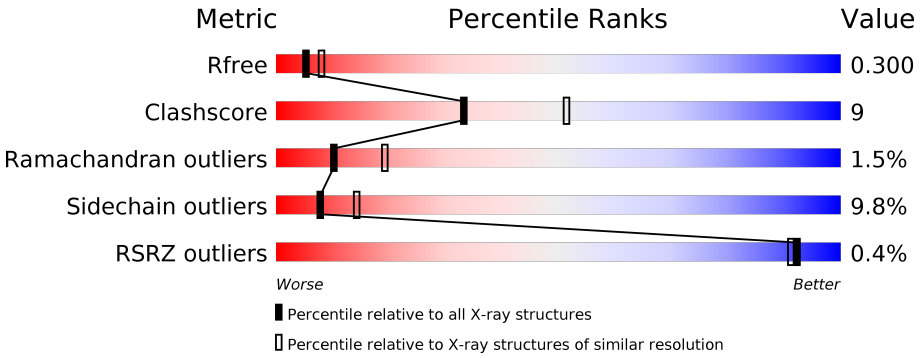
MolProbity	:	4.02b-467
Xtriage (Phenix)	:	1.13
EDS	:	2.11
Percentile statistics	:	20191225.v01 (using entries in the PDB archive December 25th 2019)
Refmac	:	5.8.0158
CCP4	:	7.0.044 (Gargrove)
Ideal geometry (proteins)	:	Engh & Huber (2001)
Ideal geometry (DNA, RNA)	:	Parkinson et al. (1996)
Validation Pipeline (wwPDB-VP)	:	2.11

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 2.61 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	3797 (2.64-2.60)
Clashscore	141614	4168 (2.64-2.60)
Ramachandran outliers	138981	4093 (2.64-2.60)
Sidechain outliers	138945	4093 (2.64-2.60)
RSRZ outliers	127900	3731 (2.64-2.60)

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 on 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	S	127	2% 72% 15% • 9%
2	H	223	68% 20% • 8%
3	L	208	74% 22% • •

2 Entry composition

There are 4 unique types of molecules in this entry. The entry contains 3959 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 Tyrosine-protein phosphatase non-receptor type substrate 1.

Mol	Chain	Residues	Atoms					ZeroOcc	AltConf	Trace
1	S	116	Total	C	N	O	S	0	0	0
			885	558	157	167	3			

There are 9 discrepancies between the modelled and reference sequences:

Chain	Residue	Modelled	Actual	Comment	Reference
S	80	ALA	ASN	conflict	UNP P78324
S	120	THR	-	expression tag	UNP P78324
S	121	ARG	-	expression tag	UNP P78324
S	122	HIS	-	expression tag	UNP P78324
S	123	HIS	-	expression tag	UNP P78324
S	124	HIS	-	expression tag	UNP P78324
S	125	HIS	-	expression tag	UNP P78324
S	126	HIS	-	expression tag	UNP P78324
S	127	HIS	-	expression tag	UNP P78324

- Molecule 2 is a protein called Fab 218 anti-SIRP-alpha antibody Variable Heavy Chain.

Mol	Chain	Residues	Atoms					ZeroOcc	AltConf	Trace
2	H	205	Total	C	N	O	S	0	0	0
			1531	967	253	306	5			

- Molecule 3 is a protein called Fab 218 anti-SIRP-alpha antibody Variable Light Chain.

Mol	Chain	Residues	Atoms					ZeroOcc	AltConf	Trace
3	L	206	Total	C	N	O	S	0	0	0
			1523	954	255	310	4			

- Molecule 4 is water.

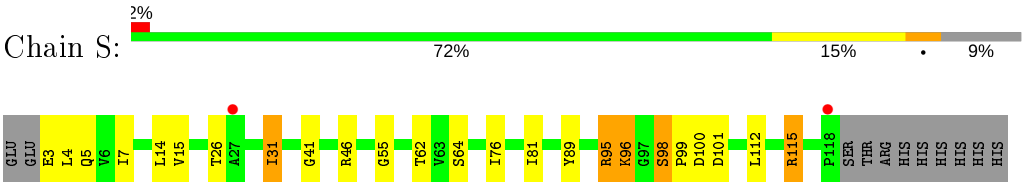
Mol	Chain	Residues	Atoms	ZeroOcc	AltConf
4	S	8	Total O 8 8	0	0
4	H	6	Total O 6 6	0	0
4	L	6	Total O 6 6	0	0

3 Residue-property plots (i)

These plots are drawn for all protein, RNA and DNA 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.

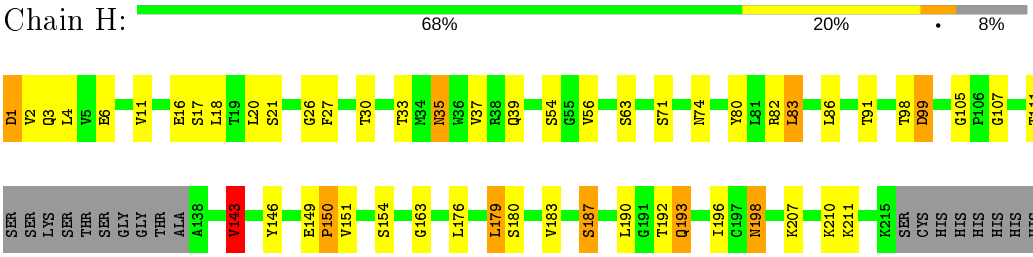
- Molecule 1: Tyrosine-protein phosphatase non-receptor type substrate 1

Chain S:



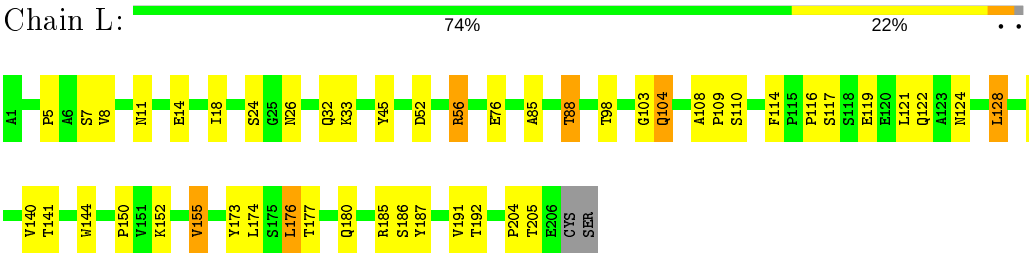
- Molecule 2: Fab 218 anti-SIRP-alpha antibody Variable Heavy Chain

Chain H:



- Molecule 3: Fab 218 anti-SIRP-alpha antibody Variable Light Chain

Chain L:



4 Data and refinement statistics (i)

Property	Value	Source
Space group	P 1 21 1	Depositor
Cell constants a, b, c, α, β, γ	44.29Å 76.09Å 72.81Å 90.00° 95.26° 90.00°	Depositor
Resolution (Å)	38.16 – 2.61 38.16 – 2.61	Depositor EDS
% Data completeness (in resolution range)	98.2 (38.16-2.61) 98.2 (38.16-2.61)	Depositor EDS
R_{merge}	(Not available)	Depositor
R_{sym}	(Not available)	Depositor
$< I/\sigma(I) >$ [1]	1.29 (at 2.61Å)	Xtriage
Refinement program	REFMAC 5.8.0230	Depositor
R, R_{free}	0.195 , 0.301 0.200 , 0.300	Depositor DCC
R_{free} test set	880 reflections (6.08%)	wwPDB-VP
Wilson B-factor (Å^2)	67.3	Xtriage
Anisotropy	0.089	Xtriage
Bulk solvent k_{sol}(e/Å^3), B_{sol}(Å^2)	0.29 , 33.3	EDS
L-test for twinning[2]	$< \|L\| > = 0.49$, $< L^2 > = 0.32$	Xtriage
Estimated twinning fraction	No twinning to report.	Xtriage
F_o,F_c correlation	0.95	EDS
Total number of atoms	3959	wwPDB-VP
Average B, all atoms (Å^2)	67.0	wwPDB-VP

Xtriage's analysis on translational NCS is as follows: *The largest off-origin peak in the Patterson function is 7.76% 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

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	S	0.62	0/903	0.82	0/1224
2	H	0.64	0/1567	0.81	0/2142
3	L	0.62	0/1562	0.80	1/2135 (0.0%)
All	All	0.63	0/4032	0.81	1/5501 (0.0%)

Chiral center outliers are detected by calculating the chiral volume of a chiral center and verifying if the center is modelled as a planar moiety or with the opposite hand.A planarity outlier is detected by checking planarity of atoms in a peptide group, atoms in a mainchain group or atoms of a sidechain that are expected to be planar.

Mol	Chain	#Chirality outliers	#Planarity outliers
1	S	0	2
2	H	0	2
3	L	0	1
All	All	0	5

There are no bond length outliers.

All (1) bond angle outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(^{o})	Ideal(^{o})
3	L	56	ARG	NE-CZ-NH1	5.11	122.86	120.30

There are no chirality outliers.

All (5) planarity outliers are listed below:

Mol	Chain	Res	Type	Group
2	H	143	VAL	Peptide
2	H	98	THR	Peptide
3	L	56	ARG	Sidechain
1	S	95	ARG	Sidechain
1	S	99	PRO	Peptide

5.2 Too-close contacts (i)

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	S	885	0	889	13	0
2	H	1531	0	1500	28	0
3	L	1523	0	1474	30	0
4	H	6	0	0	1	0
4	L	6	0	0	0	0
4	S	8	0	0	1	0
All	All	3959	0	3863	67	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 9.

All (67) 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)
2:H:1:ASP:O	2:H:26:GLY:HA3	1.91	0.70
2:H:82:ARG:NH2	4:H:301:HOH:O	2.29	0.66
2:H:120:PRO:HB3	2:H:146:TYR:HB3	1.77	0.66
2:H:6:GLU:OE2	2:H:105:GLY:HA3	1.95	0.65
3:L:180:GLN:O	3:L:187:TYR:OH	2.15	0.65
3:L:109:PRO:HA	3:L:135:PHE:HB3	1.79	0.63
2:H:143:VAL:HG13	2:H:179:LEU:HD23	1.81	0.62
2:H:193:GLN:HA	2:H:193:GLN:HE21	1.64	0.62
3:L:132:ILE:HG22	3:L:135:PHE:CE2	2.34	0.62
2:H:39:GLN:HE22	3:L:32:GLN:HE22	1.47	0.62
3:L:155:VAL:HA	3:L:173:TYR:O	1.99	0.62
3:L:114:PHE:HE2	3:L:131:LEU:HD12	1.65	0.61
2:H:11:VAL:HA	2:H:111:THR:O	2.01	0.60
3:L:177:THR:OG1	3:L:180:GLN:HG3	2.02	0.59
3:L:116:PRO:HD3	3:L:128:LEU:CD1	2.35	0.57
1:S:95:ARG:O	1:S:101:ASP:HA	2.05	0.57
3:L:132:ILE:HG22	3:L:135:PHE:CD2	2.42	0.54
1:S:46:ARG:HH21	3:L:88:THR:HG22	1.72	0.54
2:H:91:THR:HG23	2:H:111:THR:HA	1.90	0.54
2:H:163:GLY:O	2:H:183:VAL:HA	2.08	0.52
1:S:4:LEU:HD23	1:S:4:LEU:C	2.30	0.52

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Atom-1	Atom-2	Interatomic distance (Å)	Clash overlap (Å)
3:L:135:PHE:CE1	3:L:140:VAL:CG1	2.93	0.51
2:H:6:GLU:OE1	2:H:107:GLY:N	2.41	0.50
2:H:179:LEU:HD23	2:H:179:LEU:C	2.32	0.50
3:L:109:PRO:CA	3:L:135:PHE:HB3	2.41	0.50
3:L:204:PRO:O	3:L:205:THR:HG23	2.12	0.50
1:S:96:LYS:NZ	1:S:101:ASP:OD1	2.46	0.49
2:H:149:GLU:CD	2:H:150:PRO:HA	2.33	0.49
2:H:35:ASN:ND2	2:H:35:ASN:N	2.62	0.48
2:H:39:GLN:NE2	3:L:32:GLN:HE22	2.12	0.47
3:L:26:ASN:O	3:L:85:ALA:HA	2.15	0.47
2:H:17:SER:HA	2:H:83:LEU:O	2.16	0.46
1:S:4:LEU:HB2	1:S:31:ILE:O	2.15	0.46
2:H:2:VAL:HG13	2:H:27:PHE:CD1	2.51	0.46
3:L:11:ASN:HB2	3:L:14:GLU:OE1	2.15	0.46
3:L:108:ALA:HB1	3:L:109:PRO:HD2	1.99	0.45
2:H:54:SER:OG	2:H:56:VAL:HG22	2.17	0.45
3:L:117:SER:HB2	3:L:119:GLU:OE1	2.17	0.45
3:L:176:LEU:N	3:L:176:LEU:HD23	2.32	0.45
2:H:179:LEU:HG	2:H:180:SER:N	2.32	0.45
3:L:135:PHE:CE1	3:L:140:VAL:HG13	2.52	0.44
2:H:20:LEU:O	2:H:80:TYR:HA	2.18	0.44
3:L:103:GLY:O	3:L:104:GLN:HB3	2.17	0.44
2:H:143:VAL:HG13	2:H:179:LEU:CD2	2.47	0.44
2:H:83:LEU:HB3	2:H:86:LEU:HD21	2.00	0.43
2:H:196:ILE:HD13	2:H:211:LYS:HA	1.99	0.43
3:L:176:LEU:N	3:L:176:LEU:CD2	2.81	0.43
1:S:14:LEU:HD11	1:S:115:ARG:HD3	2.00	0.43
2:H:16:GLU:O	2:H:86:LEU:HD23	2.18	0.42
3:L:174:LEU:HA	3:L:174:LEU:HD12	1.89	0.42
1:S:15:VAL:CG1	1:S:112:LEU:HD11	2.49	0.42
1:S:41:GLY:HA2	4:S:206:HOH:O	2.19	0.42
3:L:173:TYR:N	3:L:173:TYR:CD1	2.88	0.42
1:S:7:ILE:HD11	1:S:26:THR:OG1	2.19	0.42
3:L:33:LYS:NZ	3:L:76:GLU:O	2.39	0.42
2:H:3:GLN:O	2:H:4:LEU:HD12	2.20	0.41
3:L:121:LEU:O	3:L:124:ASN:N	2.51	0.41
3:L:5:PRO:HG2	3:L:18:ILE:HG23	2.01	0.41
1:S:81:ILE:HD13	1:S:81:ILE:HA	1.92	0.41
1:S:62:THR:HG23	1:S:64:SER:O	2.21	0.41
2:H:30:THR:HG22	2:H:74:ASN:HB3	2.02	0.41
1:S:46:ARG:HE	3:L:88:THR:HG22	1.86	0.40

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Atom-1	Atom-2	Interatomic distance (Å)	Clash overlap (Å)
1:S:89:TYR:N	1:S:89:TYR:CD1	2.89	0.40
2:H:187:SER:HA	2:H:190:LEU:HD11	2.02	0.40
2:H:154:SER:C	2:H:198:ASN:HD22	2.24	0.40
3:L:144:TRP:CE3	3:L:174:LEU:HD22	2.57	0.40
3:L:7:SER:HB3	3:L:98:THR:HG22	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	S	114/127 (90%)	106 (93%)	5 (4%)	3 (3%)	5	8
2	H	201/223 (90%)	184 (92%)	15 (8%)	2 (1%)	15	30
3	L	204/208 (98%)	186 (91%)	15 (7%)	3 (2%)	10	19
All	All	519/558 (93%)	476 (92%)	35 (7%)	8 (2%)	10	19

All (8) Ramachandran outliers are listed below:

Mol	Chain	Res	Type
1	S	55	GLY
1	S	100	ASP
1	S	98	SER
2	H	99	ASP
3	L	104	GLN
3	L	185	ARG
2	H	63	SER
3	L	150	PRO

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	S	95/106 (90%)	88 (93%)	7 (7%)	13 26
2	H	176/191 (92%)	156 (89%)	20 (11%)	5 9
3	L	169/171 (99%)	153 (90%)	16 (10%)	8 15
All	All	440/468 (94%)	397 (90%)	43 (10%)	8 14

All (43) residues with a non-rotameric sidechain are listed below:

Mol	Chain	Res	Type
1	S	3	GLU
1	S	5	GLN
1	S	31	ILE
1	S	76	ILE
1	S	96	LYS
1	S	98	SER
1	S	115	ARG
2	H	1	ASP
2	H	18	LEU
2	H	21	SER
2	H	33	THR
2	H	35	ASN
2	H	37	VAL
2	H	71	SER
2	H	83	LEU
2	H	99	ASP
2	H	143	VAL
2	H	150	PRO
2	H	151	VAL
2	H	176	LEU
2	H	179	LEU
2	H	187	SER
2	H	192	THR
2	H	193	GLN
2	H	198	ASN
2	H	207	LYS

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Mol	Chain	Res	Type
2	H	210	LYS
3	L	8	VAL
3	L	24	SER
3	L	45	TYR
3	L	52	ASP
3	L	88	THR
3	L	110	SER
3	L	122	GLN
3	L	128	LEU
3	L	133	SER
3	L	141	THR
3	L	152	LYS
3	L	155	VAL
3	L	176	LEU
3	L	186	SER
3	L	191	VAL
3	L	192	THR

Some sidechains can be flipped to improve hydrogen bonding and reduce clashes. All (8) such sidechains are listed below:

Mol	Chain	Res	Type
1	S	70	ASN
2	H	193	GLN
2	H	198	ASN
3	L	11	ASN
3	L	32	GLN
3	L	163	GLN
3	L	165	ASN
3	L	193	HIS

5.3.3 RNA

There are no RNA molecules in this entry.

5.4 Non-standard residues in protein, DNA, RNA chains

There are no non-standard protein/DNA/RNA residues in this entry.

5.5 Carbohydrates

There are no carbohydrates in this entry.

5.6 Ligand geometry

There are no ligands in this entry.

5.7 Other polymers

There are no such residues in this entry.

5.8 Polymer linkage issues

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	S	116/127 (91%)	-0.27	2 (1%) 70 66	45, 72, 105, 133	0
2	H	205/223 (91%)	-0.41	0 100 100	42, 62, 89, 115	0
3	L	206/208 (99%)	-0.29	0 100 100	41, 65, 93, 111	0
All	All	527/558 (94%)	-0.33	2 (0%) 92 91	41, 65, 97, 133	0

All (2) RSRZ outliers are listed below:

Mol	Chain	Res	Type	RSRZ
1	S	118	PRO	2.7
1	S	27	ALA	2.5

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

There are no non-standard protein/DNA/RNA residues in this entry.

6.3 Carbohydrates

There are no carbohydrates in this entry.

6.4 Ligands

There are no ligands in this entry.

6.5 Other polymers

There are no such residues in this entry.