WORLDWIDE wwPDB PROTEIN DATA BANK

wwPDB X-ray Structure Validation Summary Report (i)

Apr 24, 2024 – 10:05 AM EDT

PDB ID : 8T9B

Title : Structure of the CK variant of Fab F1 (FabC-F1) in complex with the C-terminal FN3 domain of EphA2

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Deposited on : 2023-06-23

Resolution : 4.20 Å(reported)

This is a wwPDB X-ray Structure Validation Summary 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 (i) symbol.

The types of validation reports are described at
http://www.wwpdb.org/validation/2017/FAQs#types.

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

|		
MolProbity	:	4.02b-467
Xtriage (Phenix)	:	1.13
EDS	:	2.36.2
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)	:	2.36.2

1   Overall quality at a glance ⓘ

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

The reported resolution of this entry is 4.20 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.
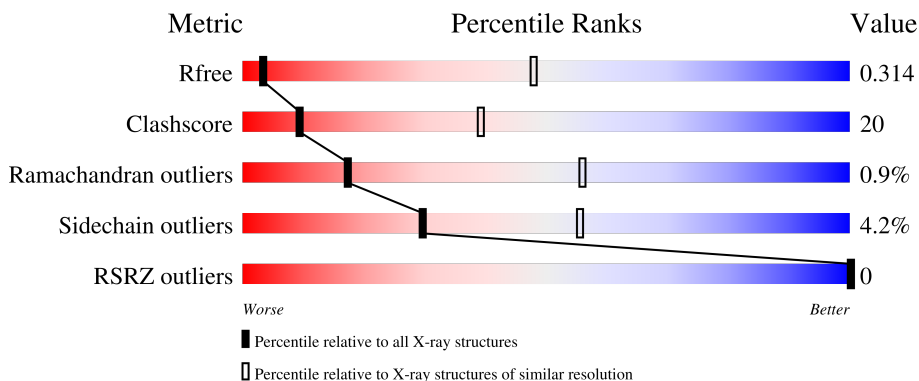
Metric	Whole archive (#Entries)	Similar resolution (#Entries, resolution range(Å))
R_{free}	130704	1005 (4.62-3.78)
Clashscore	141614	1044 (4.60-3.80)
Ramachandran outliers	138981	1000 (4.60-3.80)
Sidechain outliers	138945	1007 (4.62-3.78)
RSRZ outliers	127900	1063 (4.70-3.70)

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	A	237	61% / 36% / . .
1	C	237	57% / 38% / . . .
1	D	237	62% / 32% / . .
1	F	237	62% / 35% / .
2	B	214	62% / 35% / . .

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Mol	Chain	Length	Quality of chain
2	E	214	65% 29%
2	G	214	64% 32%
2	H	214	63% 35%
3	I	98	59% 30% 9%
3	J	98	48% 37% 11%
3	K	98	48% 40% 8%
3	L	98	48% 39% 11%

2   Entry composition ⓘ

There are 3 unique types of molecules in this entry. The entry contains 15657 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 CK variant of Fab F1 heavy chain.

Mol	Chain	Residues	Atoms					ZeroOcc	AltConf	Trace
1	C	230	Total 1707	C 1086	N 283	O 332	S 6	0	0	0
1	A	232	Total 1719	C 1091	N 284	O 337	S 7	0	0	0
1	D	230	Total 1691	C 1072	N 279	O 334	S 6	0	0	0
1	F	230	Total 1698	C 1079	N 282	O 331	S 6	0	0	0

- Molecule 2 is a protein called CK variant of Fab F1 light chain.

Mol	Chain	Residues	Atoms					ZeroOcc	AltConf	Trace
2	G	210	Total 1546	C 965	N 255	O 321	S 5	0	0	0
2	B	210	Total 1565	C 976	N 260	O 324	S 5	0	0	0
2	E	211	Total 1533	C 957	N 251	O 320	S 5	0	0	0
2	H	210	Total 1533	C 957	N 254	O 317	S 5	0	0	0

- Molecule 3 is a protein called Ephrin type-A receptor 2.

Mol	Chain	Residues	Atoms				ZeroOcc	AltConf	Trace
3	I	89	Total 654	C 409	N 110	O 135	0	0	0
3	J	87	Total 657	C 413	N 110	O 134	0	0	0
3	K	90	Total 682	C 426	N 119	O 137	0	0	0
3	L	87	Total 672	C 422	N 116	O 134	0	0	0

There are 16 discrepancies between the modelled and reference sequences:

Chain	Residue	Modelled	Actual	Comment	Reference
I	530	LEU	-	expression tag	UNP P29317
I	531	THR	-	expression tag	UNP P29317
I	532	PRO	-	expression tag	UNP P29317
I	533	ARG	-	expression tag	UNP P29317
J	530	LEU	-	expression tag	UNP P29317
J	531	THR	-	expression tag	UNP P29317
J	532	PRO	-	expression tag	UNP P29317
J	533	ARG	-	expression tag	UNP P29317
K	530	LEU	-	expression tag	UNP P29317
K	531	THR	-	expression tag	UNP P29317
K	532	PRO	-	expression tag	UNP P29317
K	533	ARG	-	expression tag	UNP P29317
L	530	LEU	-	expression tag	UNP P29317
L	531	THR	-	expression tag	UNP P29317
L	532	PRO	-	expression tag	UNP P29317
L	533	ARG	-	expression tag	UNP P29317

3 Residue-property plots ⓘ

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: CK variant of Fab F1 heavy chain

Chain C:

- Molecule 1: CK variant of Fab F1 heavy chain

Chain A:

- Molecule 1: CK variant of Fab F1 heavy chain

Chain D:

T215 N221 H224 K225 P226 S227 N228 T229 K233 K238 SER CYS ASP LYS THR HIS THR

• Molecule 1: CK variant of Fab F1 heavy chain

Chain F:

62%   35%   •

E1 V2 Q3 L4 V5 V13 Q14 P15 L19 C23 A24 A25 S26 G27 F28 G31 A32 Y33 L34 R35 H40 W41 V42 R43 K48 W52 V53 I56 Y57 P58 Y62 Y67 V71 T77 I78 S79 N85 Q90 M91 N92 S93 L94 R95 A96 T99 A100

C104 S107 A108 A109 S110 Y111 Y112 G113 Y114 W115 H116 W117 Y118 H119 F120 L132 V133 T134 S139 P143 S144 P147 P150 T159 A160 A161 L162 K167 D168 Y169 W178 N179 L183 T184 A192 V193 L194 Q195 G198 L199 Y200 S201 L202 V208 P209

T217 Y218 N221 S227 N228 T229 K233 K238 SER CYS ASP LYS THR HIS THR

• Molecule 2: CK variant of Fab F1 light chain

Chain G:

64%   32%   • •

ASP ILE GLN MET T5 Q6 S7 P8 L11 R18 V19 T22 C23 R24 Q27 S28 V29 A32 W41 Y42 Q43 Q44 K45 P46 G47 K48 A49 P50 K51 L52 L53 I54 L67 Y68 S69 G70 V71 P72 S74 R75 F76 S79 L89 T90 I91 S92 S93 L94 D98

Y103 C104 Y107 V108 W112 L113 I114 E123 R126 T127 V128 A129 V133 F136 V150 V151 C152 L153 Y158 A162 K163 V164 Q165 D169 Q173 N176 Q184 T196 L197 Y204 E205 K206 V209 Y210 A211 C212 E213 S220 R229 G230 E231 C232

• Molecule 2: CK variant of Fab F1 light chain

Chain B:

62%   35%   • •

ASP ILE GLN MET T5 Q6 S7 P8 D17 R18 V19 T20 I21 T22 C23 S31 A32 W41 Q44 K48 A49 L52 L53 I54 A57 S58 D66 L67 Y68 R75 F76 S77 G78 S79 R80 T88 L89 T90 I91 S92 S93 L94 F99 A100 T101 C104 L113 I114

T115 F116 G117 Q118 G119 T120 K121 R126 T127 P131 S139 V151 C152 L153 L154 F157 R160 E161 A162 K163 V164 Q165 D169 V181 T182 L193 S194 S195 T196 S200 K201 A202 D203 Y204 E205 K206 H207 K208 V209 Y210 A211 C212 E213 V214 T215 S220 V223 T224 K225

S226 F227 N228 R229 C232

• Molecule 2: CK variant of Fab F1 light chain

Chain E:

65%   29%   • •

ASP ILE GLN M4 V15 I21 S30 A40 W41 Y42 K45 K48 A49 L52 L53 I54 V55 S56 A57 S58 D66 L67 Y68 V71 P72 S74 R75 F76 S77 G78 S79 P96 E97 A100 C104 Q105 Q106 Y107 W112 L113 I114 T120 K121 V122 R126 T127

V133 F136 P137 P138 Q142 A148 S149 V150 L153 W166 K167 V168 D169 L172 Q173 N176 S177 Q178 E179 D188 S189 S195 T196 L197 T198 L199 E205 K206 H207 K208 V209 Y210 A211 V214 V223 S226 F227 N228 R229 G230 E231 C232

• Molecule 2: CK variant of Fab F1 light chain

Chain H:

63%	35% •

ASP ILE GLN MET T5 Q6 S10 L11 S12 A32 V39 A40 W41 Q44 G47 K48 L52 L53 I54 Y55 S56 A57 S58 D66 L67 Y68 R75 F76 S77 G78 S79 L89 T90 I91 S92 S93 L94 F99 A100 T101 C104 Q105 V108 W112 L113 K121 V122 E123

I124 K125 R126 T127 P138 S139 D140 L143 S149 V150 L154 N155 N156 R160 E161 A162 K163 V164 Q165 W166 L172 V181 T182 E183 Q184 D185 S186 K187 D188 S189 T190 Y191 S192 L193 Y204 E205 K206 Y210 A211 C212 E213 V214 T215 Q216 V223 S226 R229

C232

• Molecule 3: Ephrin type-A receptor 2

Chain I:

59%	30% •	9%

GLN THR E438 P439 S448 L452 Q462 Q463 S464 R465 V466 W467 E470 V471 T472 Y473 R474 K475 S479 N480 S481 Y482 N483 V484 R485 R486 T487 E488 G489 L494 D495 D496 L497 D500 Y503 Q506 L510 T511 Q512 E513 GLY Q515 F524 Q525 T526 L527 SER PRO LEU

THR PRO ARG

• Molecule 3: Ephrin type-A receptor 2

Chain J:

48%	37% •	11%

GLN THR E438 P439 P440 K441 V442 R443 L444 R447 S448 L452 W456 S457 I458 P459 P460 P461 R465 V466 W467 E470 V471 R474 K475 D478 S479 N480 S481 Y482 N483 V484 R485 R486 T487 E488 V492 T493 L494 L497 A498 P499 D500 T501 T502 Y503 Q506 V507 L510

T511 Q512 GLU GLY GLN G516 S519 H522 L527 SER PRO LEU THR PRO ARG

• Molecule 3: Ephrin type-A receptor 2

Chain K:

48%	40% •	8%

GLN THR E438 P439 P440 G446 L452 S463 I458 Q462 Q463 S464 W467 K468 V471 T472 Y473 R474 K475 D478 S479 N480 S481 Y482 N483 V484 R485 R486 T487 E488 G489 F490 L494 D495 D496 L497 Y503 L504 V505 Q506 A509 L510 A517 G518 S519 K520 V521 H522 E523

F524 L527 SER PRO LEU THR PRO ARG

• Molecule 3: Ephrin type-A receptor 2

Chain L:

48%	39% •	11%

GLN THR E438 P439 G446 R447 S448 S451 L452 S453 P459 P460 P461 Q462 Q463 S464 R465 V466 W467 K468 Y469 E470 V471 T472 Y473 R474 K475 K476 N480 S481 Y482 N483 V484 R485 R486 T487 E488 G489 D495 D496 L497 A498 T501 T502 Y503 V507 L510 T511 Q512 GLU GLY GLN

G516 L527 SER PRO LEU THR PRO ARG

4 Data and refinement statistics (i)

Property	Value	Source
Space group	P 1 21 1	Depositor
Cell constants a, b, c, α, β, γ	76.66Å 130.65Å 223.00Å 90.00° 89.76° 90.00°	Depositor
Resolution (Å)	66.12 – 4.20 66.12 – 4.20	Depositor EDS
% Data completeness (in resolution range)	96.2 (66.12-4.20) 95.1 (66.12-4.20)	Depositor EDS
R_{merge}	0.17	Depositor
R_{sym}	(Not available)	Depositor
$< I/\sigma(I) >$ [1]	2.34 (at 4.14Å)	Xtriage
Refinement program	PHENIX 1.20.1_4487, PHENIX 1.20.1_4487	Depositor
R, R_{free}	0.296 , 0.315 0.296 , 0.314	Depositor DCC
R_{free} test set	1713 reflections (5.51%)	wwPDB-VP
Wilson B-factor (Å^2)	101.4	Xtriage
Anisotropy	1.379	Xtriage
Bulk solvent k_{sol}(e/Å^3), B_{sol}(Å^2)	0.18 , 24.6	EDS
L-test for twinning[2]	$< \|L\| > = 0.45, < L^2 > = 0.28$	Xtriage
Estimated twinning fraction	0.347 for h,-k,-l	Xtriage
F_o,F_c correlation	0.89	EDS
Total number of atoms	15657	wwPDB-VP
Average B, all atoms (Å^2)	156.0	wwPDB-VP

Xtriage's analysis on translational NCS is as follows: *The analyses of the Patterson function reveals a significant off-origin peak that is 29.53 % of the origin peak, indicating pseudo-translational symmetry. The chance of finding a peak of this or larger height randomly in a structure without pseudo-translational symmetry is equal to 1.5258e-03. The detected translational NCS is most likely also responsible for the elevated intensity ratio.*

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[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	A	0.30	0/1770	0.58	0/2424
1	C	0.30	0/1758	0.60	0/2407
1	D	0.32	0/1742	0.64	0/2391
1	F	0.30	0/1749	0.59	0/2397
2	B	0.30	0/1597	0.59	0/2177
2	E	0.33	0/1565	0.61	0/2143
2	G	0.30	0/1578	0.59	0/2154
2	H	0.30	0/1565	0.59	0/2142
3	I	0.31	0/667	0.61	0/915
3	J	0.37	0/672	0.68	0/922
3	K	0.33	0/698	0.64	0/956
3	L	0.35	0/687	0.68	0/939
All	All	0.31	0/16048	0.61	0/21967

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	A	0	1
1	D	0	3
3	I	0	1
All	All	0	5

There are no bond length outliers.

There are no bond angle outliers.

There are no chirality outliers.

All (5) planarity outliers are listed below:

Mol	Chain	Res	Type	Group
1	A	75	ARG	Sidechain

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Mol	Chain	Res	Type	Group
1	D	136	SER	Peptide
1	D	35	ARG	Sidechain
1	D	75	ARG	Sidechain
3	I	485	ARG	Sidechain

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	A	1719	0	1609	68	0
1	C	1707	0	1606	72	0
1	D	1691	0	1558	59	0
1	F	1698	0	1584	67	0
2	B	1565	0	1474	62	0
2	E	1533	0	1404	61	0
2	G	1546	0	1446	54	0
2	H	1533	0	1412	55	0
3	I	654	0	594	20	0
3	J	657	0	604	45	0
3	K	682	0	633	37	0
3	L	672	0	636	46	0
All	All	15657	0	14560	595	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 20.

The worst 5 of 595 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:E:205:GLU:HA	2:E:229:ARG:HH12	1.16	1.04
2:B:6:GLN:HG3	2:B:119:GLY:H	1.27	1.00
2:G:69:SER:HB3	2:G:72:PRO:HG3	1.43	0.97
2:B:79:SER:HB3	2:B:88:THR:HG23	1.46	0.96
1:D:28:PHE:HE1	1:D:33:TYR:HB2	1.30	0.96

There are no symmetry-related clashes.

5.3 Torsion angles ⓘ

5.3.1 Protein backbone ⓘ

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	A	230/237 (97%)	202 (88%)	26 (11%)	2 (1%)	17	56
1	C	228/237 (96%)	197 (86%)	30 (13%)	1 (0%)	34	72
1	D	228/237 (96%)	202 (89%)	23 (10%)	3 (1%)	12	48
1	F	228/237 (96%)	204 (90%)	22 (10%)	2 (1%)	17	56
2	B	208/214 (97%)	177 (85%)	31 (15%)	0	100	100
2	E	209/214 (98%)	179 (86%)	29 (14%)	1 (0%)	29	68
2	G	208/214 (97%)	179 (86%)	25 (12%)	4 (2%)	8	41
2	H	208/214 (97%)	178 (86%)	28 (14%)	2 (1%)	15	54
3	I	85/98 (87%)	67 (79%)	17 (20%)	1 (1%)	13	50
3	J	83/98 (85%)	66 (80%)	16 (19%)	1 (1%)	13	50
3	K	88/98 (90%)	72 (82%)	15 (17%)	1 (1%)	14	52
3	L	83/98 (85%)	70 (84%)	13 (16%)	0	100	100
All	All	2086/2196 (95%)	1793 (86%)	275 (13%)	18 (1%)	17	56

5 of 18 Ramachandran outliers are listed below:

Mol	Chain	Res	Type
1	C	116	HIS
1	D	173	PRO
2	H	54	ILE
2	H	56	SER
3	J	466	VAL

5.3.2 Protein sidechains ⓘ

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	A	182/195 (93%)	176 (97%)	6 (3%)	38	61
1	C	180/195 (92%)	172 (96%)	8 (4%)	28	54
1	D	177/195 (91%)	167 (94%)	10 (6%)	21	48
1	F	178/195 (91%)	174 (98%)	4 (2%)	52	70
2	B	173/187 (92%)	166 (96%)	7 (4%)	31	57
2	E	165/187 (88%)	156 (94%)	9 (6%)	21	49
2	G	170/187 (91%)	165 (97%)	5 (3%)	42	64
2	H	165/187 (88%)	158 (96%)	7 (4%)	30	55
3	I	69/89 (78%)	65 (94%)	4 (6%)	20	47
3	J	71/89 (80%)	67 (94%)	4 (6%)	21	48
3	K	73/89 (82%)	69 (94%)	4 (6%)	21	49
3	L	74/89 (83%)	72 (97%)	2 (3%)	44	66
All	All	1677/1884 (89%)	1607 (96%)	70 (4%)	30	55

5 of 70 residues with a non-rotameric sidechain are listed below:

Mol	Chain	Res	Type
3	I	485	ARG
3	I	506	GLN
3	K	482	TYR
2	B	163	LYS
2	B	126	ARG

Sometimes sidechains can be flipped to improve hydrogen bonding and reduce clashes. All (1) such sidechains are listed below:

Mol	Chain	Res	Type
1	D	40	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 monosaccharides 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, 95th 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(Å²)	Q<0.9
1	A	232/237 (97%)	-0.57	0	100	100	121, 156, 216, 248	0
1	C	230/237 (97%)	-0.63	0	100	100	115, 166, 205, 222	0
1	D	230/237 (97%)	-0.60	0	100	100	120, 152, 201, 209	0
1	F	230/237 (97%)	-0.63	0	100	100	118, 149, 200, 216	0
2	B	210/214 (98%)	-0.56	0	100	100	121, 170, 218, 239	0
2	E	211/214 (98%)	-0.61	0	100	100	123, 168, 213, 244	0
2	G	210/214 (98%)	-0.61	0	100	100	114, 159, 191, 210	0
2	H	210/214 (98%)	-0.65	0	100	100	116, 163, 194, 221	0
3	I	89/98 (90%)	-0.62	0	100	100	100, 121, 144, 170	0
3	J	87/98 (88%)	-0.57	0	100	100	100, 130, 185, 202	0
3	K	90/98 (91%)	-0.59	0	100	100	105, 127, 164, 172	0
3	L	87/98 (88%)	-0.64	0	100	100	105, 130, 178, 200	0
All	All	2116/2196 (96%)	-0.61	0	100	100	100, 153, 206, 248	0

There are no RSRZ outliers to report.

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 monosaccharides 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.