



Full wwPDB X-ray Structure Validation Report

Feb 28, 2014 – 10:31 AM GMT

PDB ID : 4KG8
Title : Crystal structure of light mutant
Authors : Liu, W.; Zhan, C.; Kumar, P.R.; Bonanno, J.B.; Nathenson, S.G.; Almo, S.C.;
New York Structural Genomics Research Consortium (NYSGRG); Atoms-to-
Animals: The Immune Function Network (IFN)
Deposited on : 2013-04-28
Resolution : 2.25 Å(reported)

This is a full wwPDB validation report for a publicly released PDB entry.
We welcome your comments at validation@mail.wwpdb.org
A user guide is available at <http://wwpdb.org/ValidationPDFNotes.html>

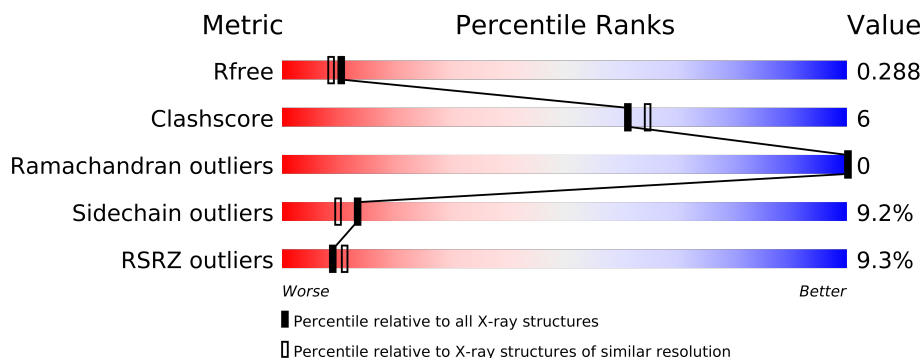
The following versions of software and data (see [references](#)) were used in the production of this report:

MolProbity : 4.02b-467
Mogul : 1.15 2013
Xtriage (Phenix) : dev-1323
EDS : stable22639
Percentile statistics : 21963
Refmac : 5.8.0049
CCP4 : 6.3.0 (Settle)
Ideal geometry (proteins) : Engh & Huber (2001)
Ideal geometry (DNA, RNA) : Parkinson et. al. (1996)
Validation Pipeline (wwPDB-VP) : stable22683

1 Overall quality at a glance

The reported resolution of this entry is 2.25 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}	66092	1108 (2.28-2.24)
Clashscore	79885	1326 (2.28-2.24)
Ramachandran outliers	78287	1291 (2.28-2.24)
Sidechain outliers	78261	1291 (2.28-2.24)
RSRZ outliers	66119	1110 (2.28-2.24)

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. The upper red bar (where present) indicates the fraction of residues that have poor fit to the electron density.

Mol	Chain	Length	Quality of chain
1	A	158	
1	B	158	
1	C	158	

2 Entry composition

There are 2 unique types of molecules in this entry. The entry contains 2843 atoms, of which 0 are hydrogen and 0 are deuterium.

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 Tumor necrosis factor ligand superfamily member 14.

Mol	Chain	Residues	Atoms					ZeroOcc	AltConf	Trace
1	A	129	Total	C	N	O	S	0	0	0
			993	639	167	186	1			
1	B	120	Total	C	N	O	S	0	0	0
			924	595	152	176	1			
1	C	115	Total	C	N	O	S	0	0	0
			901	586	149	165	1			

There are 12 discrepancies between the modelled and reference sequences:

Chain	Residue	Modelled	Actual	Comment	Reference
A	195	SER	ARG	engineered mutation	UNP O43557
A	196	ASN	VAL	engineered mutation	UNP O43557
A	198	PHE	TRP	engineered mutation	UNP O43557
A	214	GLU	LYS	engineered mutation	UNP O43557
B	195	SER	ARG	engineered mutation	UNP O43557
B	196	ASN	VAL	engineered mutation	UNP O43557
B	198	PHE	TRP	engineered mutation	UNP O43557
B	214	GLU	LYS	engineered mutation	UNP O43557
C	195	SER	ARG	engineered mutation	UNP O43557
C	196	ASN	VAL	engineered mutation	UNP O43557
C	198	PHE	TRP	engineered mutation	UNP O43557
C	214	GLU	LYS	engineered mutation	UNP O43557

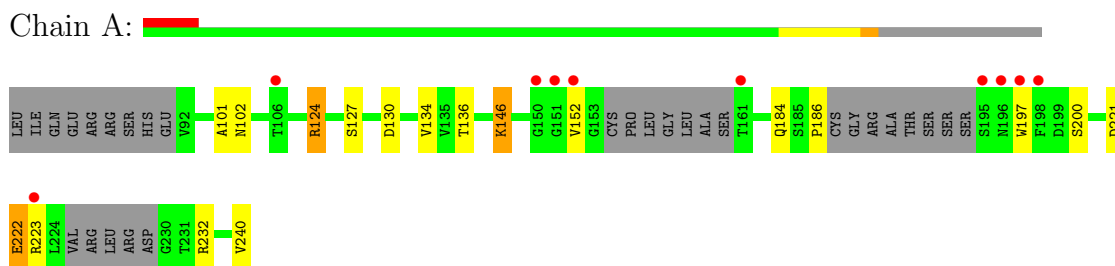
- Molecule 2 is water.

Mol	Chain	Residues	Atoms		ZeroOcc	AltConf
2	A	16	Total	O	0	0
			16	16		
2	B	4	Total	O	0	0
			4	4		
2	C	5	Total	O	0	0
			5	5		

3 Residue-property plots

These plots are drawn for all protein, RNA and DNA chains in the entry. The first graphic for a chain summarises the proportions of errors 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.

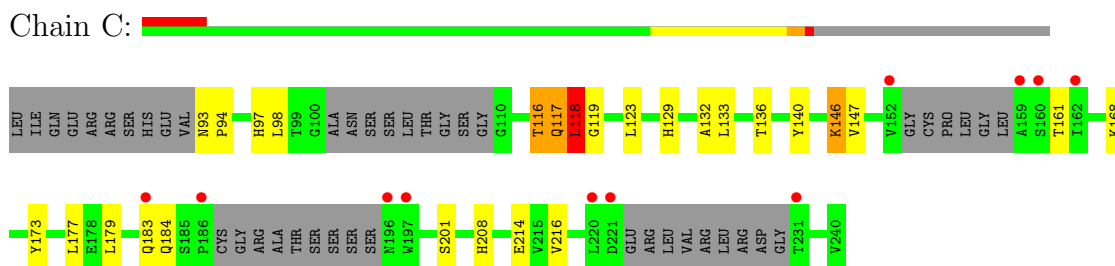
- Molecule 1: Tumor necrosis factor ligand superfamily member 14



- Molecule 1: Tumor necrosis factor ligand superfamily member 14



- Molecule 1: Tumor necrosis factor ligand superfamily member 14



4 Data and refinement statistics

Property	Value	Source
Space group	P 1 21 1	Depositor
Cell constants a, b, c, α , β , γ	59.64Å 46.91Å 70.40Å 90.00° 98.02° 90.00°	Depositor
Resolution (Å)	20.00 – 2.25 42.25 – 2.25	Depositor EDS
% Data completeness (in resolution range)	99.6 (20.00-2.25) 99.6 (42.25-2.25)	Depositor EDS
R_{merge}	0.08	Depositor
R_{sym}	0.06	Depositor
$\langle I/\sigma(I) \rangle$ ¹	5.88 (at 2.24Å)	Xtriage
Refinement program	REFMAC 5.7.0029	Depositor
R, R_{free}	0.211 , 0.286 0.216 , 0.288	Depositor DCC
R_{free} test set	949 reflections (5.39%)	DCC
Wilson B-factor (Å ²)	34.9	Xtriage
Anisotropy	0.364	Xtriage
Bulk solvent k_{sol} (e/Å ³), B_{sol} (Å ²)	0.33 , 29.8	EDS
Estimated twinning fraction	No twinning to report.	Xtriage
L-test for twinning	$\langle L \rangle = 0.49$, $\langle L^2 \rangle = 0.33$	Xtriage
Outliers	0 of 18557 reflections	Xtriage
F_o, F_c correlation	0.94	EDS
Total number of atoms	2843	wwPDB-VP
Average B, all atoms (Å ²)	41.0	wwPDB-VP

Xtriage's analysis on translational NCS is as follows: *The largest off-origin peak in the Patterson function is 7.48% of the height of the origin peak. No significant pseudotranslation is detected.*

¹Intensities estimated from amplitudes.

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	0.84	0/1015	0.94	1/1374 (0.1%)
1	B	0.78	0/943	0.93	1/1273 (0.1%)
1	C	0.69	0/922	0.88	2/1248 (0.2%)
All	All	0.78	0/2880	0.92	4/3895 (0.1%)

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	2
1	B	0	1
All	All	0	3

There are no bond length outliers.

All (4) bond angle outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(°)	Ideal(°)
1	C	118	LEU	CA-CB-CG	7.94	133.56	115.30
1	A	124	ARG	NE-CZ-NH2	-7.29	116.65	120.30
1	C	118	LEU	N-CA-C	-5.86	95.17	111.00
1	B	177	LEU	CA-CB-CG	5.56	128.08	115.30

There are no chirality outliers.

All (3) planarity outliers are listed below:

Mol	Chain	Res	Type	Group
1	A	101	ALA	Peptide
1	A	222	GLU	Peptide
1	B	173	TYR	Peptide

5.2 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 hydrogens added by MolProbity. The Clashes column lists the number of clashes within the asymmetric unit, and the number in parentheses is this value normalized per 1000 atoms of the molecule in the chain. The Symm-Clashes column gives symmetry related clashes, in the same way as for the Clashes column.

Mol	Chain	Non-H	H(model)	H(added)	Clashes	Symm-Clashes
1	A	993	0	971	7	0
1	B	924	0	888	13	0
1	C	901	0	880	14	0
2	A	16	0	0	0	0
2	B	4	0	0	0	0
2	C	5	0	0	1	0
All	All	2843	0	2739	32	0

Clashscore is defined as the number of clashes calculated for the entry per 1000 atoms (including hydrogens) of the entry. The overall clashscore for this entry is 6.

All (32) close contacts within the same asymmetric unit are listed below.

Atom-1	Atom-2	Distance(Å)	Clash(Å)
1:C:118:LEU:HG	1:C:119:GLY:HA2	1.54	0.90
1:B:146:LYS:HD3	1:B:200:SER:OG	1.87	0.74
1:B:170:THR:OG1	1:B:213:GLU:OE2	2.08	0.64
1:A:146:LYS:HD3	1:A:200:SER:OG	1.96	0.64
1:B:103:SER:CB	1:B:104:SER:HB3	2.33	0.58
1:A:232:ARG:NH1	1:C:183:GLN:OE1	2.37	0.57
1:B:103:SER:HB3	1:B:104:SER:CB	2.34	0.57
1:B:103:SER:HB3	1:B:104:SER:HB3	1.87	0.56
1:C:116:THR:HB	1:C:123:LEU:HD11	1.89	0.55
1:C:168:LYS:HB2	1:C:179:LEU:HD11	1.88	0.55
1:C:118:LEU:CG	1:C:119:GLY:HA2	2.32	0.53
1:B:109:GLY:N	1:B:110:GLY:HA2	2.24	0.52
1:B:143:ILE:O	1:B:204:GLY:HA2	2.11	0.51
1:C:133:LEU:O	1:C:214:GLU:HA	2.10	0.50
1:B:103:SER:CB	1:B:104:SER:CB	2.91	0.49
1:B:173:TYR:N	1:B:174:PRO:C	2.66	0.49
1:A:186:PRO:HG2	1:A:197:TRP:CZ3	2.51	0.46
1:C:132:ALA:HB2	1:C:216:VAL:HG12	1.99	0.45
1:C:116:THR:O	1:C:117:GLN:HG2	2.16	0.45
1:A:222:GLU:O	1:A:223:ARG:HG3	2.16	0.45
1:A:240:VAL:HG12	1:A:240:VAL:OXT	2.15	0.45

Continued on next page...

Continued from previous page...

Atom-1	Atom-2	Distance(Å)	Clash(Å)
1:C:129:HIS:O	1:C:129:HIS:CD2	2.69	0.44
1:C:97:HIS:O	1:C:98:LEU:HD23	2.17	0.43
1:C:208:HIS:CD2	2:C:304:HOH:O	2.70	0.43
1:B:173:TYR:N	1:B:174:PRO:CA	2.81	0.43
1:A:127:SER:HB2	1:A:134:VAL:HB	2.01	0.42
1:B:163:THR:O	1:B:219:VAL:HA	2.19	0.41
1:B:103:SER:HB3	1:B:104:SER:HB2	2.00	0.41
1:B:163:THR:HB	1:B:220:LEU:HG	2.02	0.41
1:C:93:ASN:ND2	1:C:94:PRO:HD3	2.36	0.41
1:C:146:LYS:HA	1:C:201:SER:O	2.21	0.41
1:A:124:ARG:NE	1:C:140:TYR:OH	2.54	0.40

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	121/158 (77%)	116 (96%)	5 (4%)	0	100	100
1	B	108/158 (68%)	101 (94%)	7 (6%)	0	100	100
1	C	105/158 (66%)	98 (93%)	7 (7%)	0	100	100
All	All	334/474 (70%)	315 (94%)	19 (6%)	0	100	100

There are no Ramachandran outliers to report.

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	104/129 (81%)	97 (93%)	7 (7%)	23	21
1	B	96/129 (74%)	86 (90%)	10 (10%)	10	7
1	C	94/129 (73%)	84 (89%)	10 (11%)	10	6
All	All	294/387 (76%)	267 (91%)	27 (9%)	13	10

All (27) residues with a non-rotameric sidechain are listed below:

Mol	Chain	Res	Type
1	A	102	ASN
1	A	130	ASP
1	A	136	THR
1	A	146	LYS
1	A	152	VAL
1	A	184	GLN
1	A	221	ASP
1	B	103	SER
1	B	146	LYS
1	B	147	VAL
1	B	160	SER
1	B	173	TYR
1	B	175	GLU
1	B	177	LEU
1	B	219	VAL
1	B	221	ASP
1	B	232	ARG
1	C	116	THR
1	C	117	GLN
1	C	118	LEU
1	C	136	THR
1	C	146	LYS
1	C	147	VAL
1	C	161	THR
1	C	173	TYR
1	C	177	LEU
1	C	184	GLN

Some sidechains can be flipped to improve hydrogen bonding and reduce clashes. All (4) such sidechains are listed below:

Mol	Chain	Res	Type
1	A	93	ASN
1	C	93	ASN

Continued on next page...

Continued from previous page...

Mol	Chain	Res	Type
1	C	129	HIS
1	C	208	HIS

5.3.3 RNA ⓘ

There are no RNA chains 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, 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(Å ²)	Q<0.9
1	A	129/158 (81%)	0.16	10 (7%) 13 16	15, 25, 61, 83	0
1	B	120/158 (75%)	0.53	13 (10%) 6 7	19, 42, 78, 105	0
1	C	115/158 (72%)	0.61	11 (9%) 8 10	18, 44, 79, 90	0
All	All	364/474 (76%)	0.42	34 (9%) 9 11	15, 37, 77, 105	0

All (34) RSRZ outliers are listed below:

Mol	Chain	Res	Type	RSRZ
1	B	159	ALA	6.4
1	A	152	VAL	5.0
1	A	196	ASN	4.8
1	B	170	THR	4.2
1	C	221	ASP	4.2
1	A	151	GLY	4.1
1	B	110	GLY	4.1
1	C	197	TRP	3.8
1	B	173	TYR	3.8
1	C	159	ALA	3.8
1	B	230	GLY	3.6
1	B	108	SER	3.5
1	B	160	SER	3.5
1	A	195	SER	3.3
1	C	220	LEU	3.2
1	A	198	PHE	3.2
1	C	160	SER	3.1
1	C	196	ASN	3.1
1	B	221	ASP	3.0
1	C	186	PRO	2.9
1	B	220	LEU	2.6
1	C	162	ILE	2.6
1	A	161	THR	2.6

Continued on next page...

Continued from previous page...

Mol	Chain	Res	Type	RSRZ
1	C	231	THR	2.6
1	B	107	GLY	2.5
1	A	197	TRP	2.5
1	B	196	ASN	2.4
1	C	152	VAL	2.4
1	A	223	ARG	2.4
1	B	103	SER	2.4
1	A	106	THR	2.3
1	C	183	GLN	2.1
1	B	140	TYR	2.1
1	A	150	GLY	2.0

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.