wwPDB/EMDataBank EM Map/Model Validation Summary Report (i)

Nov 16, 2016 – 01:57 PM EST

PDB ID	:	5ME2
EMDB ID:	:	EMD-3488
Title	:	CryoEM structure of haemoglobin at 3.2 A determined with the Volta phase plate
Authors	:	Khoshouei, M.; Radjainia, M.; Baumeister, W.; Danev, R.
Deposited on	:	2016-11-14
Resolution	:	3.20 Å(reported)

This is a wwPDB/EMDataBank EM Map/Model Validation Summary Report for a publicly released PDB/EMDB entry.

We welcome your comments at *validation@mail.wwpdb.org*
A user guide is available at
http://wwpdb.org/validation/2016/EMValidationReportHelp
with specific help available everywhere you see the (i) symbol.

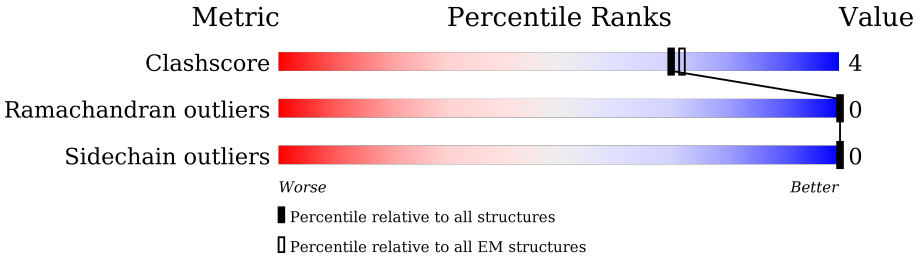
MolProbity	:	4.02b-467
Mogul	:	1.7.1 (RC1), CSD as537be (2016)
Percentile statistics	:	20151230.v01 (using entries in the PDB archive December 30th 2015)
EM map analysis	:	NOT EXECUTED
Ideal geometry (proteins)	:	Engh & Huber (2001)
Ideal geometry (DNA, RNA)	:	Parkinson et. al. (1996)
Validation Pipeline (wwPDB-VP)	:	rb-20028320

1 Overall quality at a glance (i)

The following experimental techniques were used to determine the structure:
ELECTRON MICROSCOPY

The reported resolution of this entry is 3.20 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)	EM structures (#Entries)
Clashscore	114402	924
Ramachandran outliers	111179	726
Sidechain outliers	111093	686

The table below summarises the geometric issues observed across the polymeric chains. The red, orange, yellow and green segments on the bar indicate the fraction of residues that contain outliers for >=3, 2, 1 and 0 types of geometric quality criteria. 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%

Mol	Chain	Length	Quality of chain
1	A	140	94% 6%
1	C	140	94% 6%
2	B	143	88% 12%
2	D	143	88% 12%

2 Entry composition (i)

There are 3 unique types of molecules in this entry. The entry contains 4468 atoms, of which 0 are hydrogens and 0 are deuteriums.

In the tables below, 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.

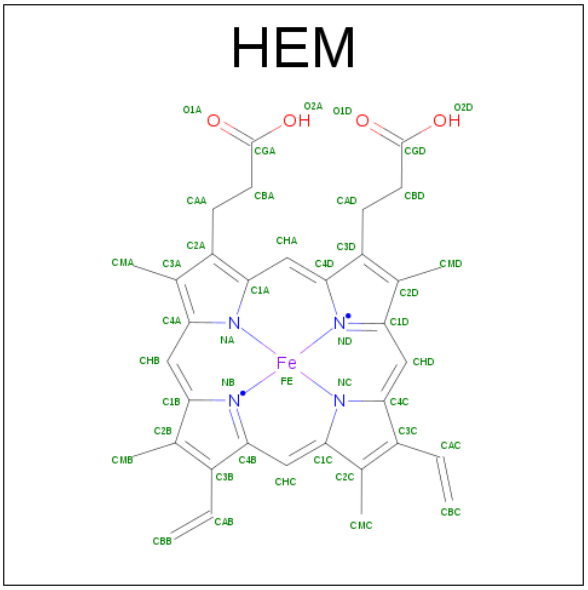
- Molecule 1 is a protein called Hemoglobin subunit alpha.

Mol	Chain	Residues	Atoms					AltConf	Trace
			Total	C	N	O	S		
1	A	140	1057	679	183	192	3	0	0
1	C	140	1057	679	183	192	3	0	0

- Molecule 2 is a protein called Hemoglobin subunit beta.

Mol	Chain	Residues	Atoms					AltConf	Trace
			Total	C	N	O	S		
2	B	143	1091	703	189	196	3	0	0
2	D	143	1091	703	189	196	3	0	0

- Molecule 3 is PROTOPORPHYRIN IX CONTAINING FE (three-letter code: HEM) (formula: $C_{34}H_{32}FeN_4O_4$).



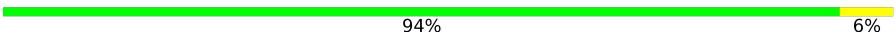
Mol	Chain	Residues	Atoms					AltConf
			Total	C	Fe	N	O	
3	A	1	43	34	1	4	4	0
3	B	1	43	34	1	4	4	0
3	C	1	43	34	1	4	4	0
3	D	1	43	34	1	4	4	0

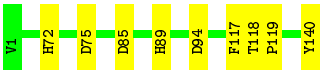
3 Residue-property plots (i)

These plots are drawn for all protein, RNA and DNA chains in the entry. The first graphic for a chain summarises the proportions of errors displayed in the second graphic. The second graphic shows the sequence view annotated by issues in geometry. 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. 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: Hemoglobin subunit alpha

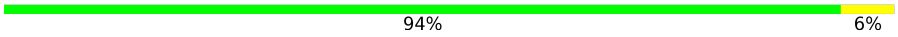
Chain A:

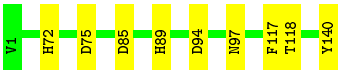




- Molecule 1: Hemoglobin subunit alpha

Chain C:



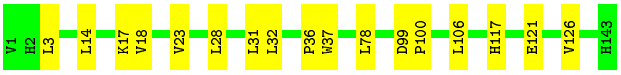


- Molecule 2: Hemoglobin subunit beta

Chain B:

- Molecule 2: Hemoglobin subunit beta

Chain D:



4 Experimental information (i)

Property	Value	Source
Reconstruction method	SINGLE PARTICLE	Depositor
Imposed symmetry	POINT, Not provided	Depositor
Number of particles used	175300	Depositor
Resolution determination method	FSC 0.143 CUT-OFF	Depositor
CTF correction method	Not provided	Depositor
Microscope	FEI TITAN KRIOS	Depositor
Voltage (kV)	300	Depositor
Electron dose $(e^-/\text{Å}^2)$	Not provided	Depositor
Minimum defocus (nm)	Not provided	Depositor
Maximum defocus (nm)	Not provided	Depositor
Magnification	Not provided	Depositor
Image detector	Not provided	Depositor

5 Model quality (i)

5.1 Standard geometry (i)

Bond lengths and bond angles in the following residue types are not validated in this section: HEM

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	$\#\|Z\| >2$	RMSZ	$\#\|Z\| >2$
1	A	0.40	0/1085	0.48	0/1477
1	C	0.40	0/1085	0.48	0/1477
2	B	0.38	0/1119	0.47	0/1522
2	D	0.38	0/1119	0.47	0/1522
All	All	0.39	0/4408	0.47	0/5998

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 (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	A	1057	0	1060	7	0
1	C	1057	0	1060	6	0
2	B	1091	0	1089	11	0
2	D	1091	0	1089	10	0
3	A	43	0	30	2	0
3	B	43	0	30	5	0
3	C	43	0	30	4	0
3	D	43	0	30	2	0
All	All	4468	0	4418	40	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 4.

The worst 5 of 40 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)
1:C:85:ASP:HA	1:C:89:HIS:HD2	1.48	0.76
1:A:85:ASP:HA	1:A:89:HIS:HD2	1.48	0.76
3:C:201:HEM:HBD1	3:C:201:HEM:HHA	1.71	0.70
3:A:201:HEM:HHC	3:A:201:HEM:HBB2	1.72	0.70
2:D:17:LYS:NZ	2:D:121:GLU:OE2	2.28	0.67

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 PDB entries followed by that with respect to all EM entries.

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	138/140 (99%)	134 (97%)	4 (3%)	0	100	100
1	C	138/140 (99%)	134 (97%)	4 (3%)	0	100	100
2	B	141/143 (99%)	137 (97%)	4 (3%)	0	100	100
2	D	141/143 (99%)	137 (97%)	4 (3%)	0	100	100
All	All	558/566 (99%)	542 (97%)	16 (3%)	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 PDB entries followed by that with respect to all EM entries.

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	112/112 (100%)	112 (100%)	0	100	100
1	C	112/112 (100%)	112 (100%)	0	100	100
2	B	115/115 (100%)	115 (100%)	0	100	100
2	D	115/115 (100%)	115 (100%)	0	100	100
All	All	454/454 (100%)	454 (100%)	0	100	100

There are no protein residues with a non-rotameric sidechain to report.

Some sidechains can be flipped to improve hydrogen bonding and reduce clashes. 5 of 9 such sidechains are listed below:

Mol	Chain	Res	Type
1	C	20	HIS
2	D	131	GLN
1	C	89	HIS
1	A	89	HIS
1	C	72	HIS

5.3.3 RNA (i)

There are no RNA molecules in this entry.

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

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

5.5 Carbohydrates (i)

There are no carbohydrates in this entry.

5.6 Ligand geometry (i)

4 ligands 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	HEM	A	201	1	24,50,50	2.19	4 (16%)	16,82,82	1.31	1 (6%)
3	HEM	B	201	2	24,50,50	1.99	4 (16%)	16,82,82	1.44	3 (18%)
3	HEM	C	201	1	24,50,50	2.04	4 (16%)	16,82,82	1.68	5 (31%)
3	HEM	D	201	2	24,50,50	1.99	4 (16%)	16,82,82	1.48	3 (18%)

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	HEM	A	201	1	-	0/6/54/54	0/0/8/8
3	HEM	B	201	2	-	0/6/54/54	0/0/8/8
3	HEM	C	201	1	-	0/6/54/54	0/0/8/8
3	HEM	D	201	2	-	0/6/54/54	0/0/8/8

The worst 5 of 16 bond length outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(Å)	Ideal(Å)
3	A	201	HEM	C3B-C2B	-6.68	1.31	1.40
3	C	201	HEM	C3B-C2B	-5.41	1.33	1.40
3	C	201	HEM	C3C-C2C	-5.39	1.33	1.40
3	A	201	HEM	C3C-C2C	-5.38	1.33	1.40
3	D	201	HEM	C3C-C2C	-5.15	1.33	1.40

The worst 5 of 12 bond angle outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(^{o})	Ideal(^{o})
3	D	201	HEM	CBD-CAD-C3D	-2.98	107.24	112.47
3	C	201	HEM	CAD-CBD-CGD	-2.52	107.89	112.78
3	C	201	HEM	C3B-CAB-CBB	-2.47	121.43	126.40
3	D	201	HEM	CAA-CBA-CGA	-2.46	108.00	112.78
3	B	201	HEM	CAD-CBD-CGD	-2.45	108.01	112.78

There are no chirality outliers.

There are no torsion outliers.

There are no ring outliers.

4 monomers are involved in 13 short contacts:

Mol	Chain	Res	Type	Clashes	Symm-Clashes
3	A	201	HEM	2	0
3	B	201	HEM	5	0
3	C	201	HEM	4	0
3	D	201	HEM	2	0

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.