



wwPDB X-ray Structure Validation Summary Report ⓘ

Feb 1, 2016 – 01:43 AM GMT

PDB ID : 2E4M
Title : Crystal structure of hemagglutinin subcomponent complex (HA-33/HA-17)
from Clostridium botulinum serotype D strain 4947
Authors : Hasegawa, K.; Watanabe, T.; Suzuki, T.; Yamano, A.; Niwa, K.; Ohyama, T.
Deposited on : 2006-12-13
Resolution : 1.85 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
<http://wwpdb.org/validation/2016/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
Mogul : 1.7 (RC4), CSD as536be (2015)
Xtriage (Phenix) : 1.9-1692
EDS : rb-20026688
Percentile statistics : 20151230.v01 (using entries in the PDB archive December 30th 2015)
Refmac : 5.8.0135
CCP4 : 6.5.0
Ideal geometry (proteins) : Engh & Huber (2001)
Ideal geometry (DNA, RNA) : Parkinson et al. (1996)
Validation Pipeline (wwPDB-VP) : trunk26865

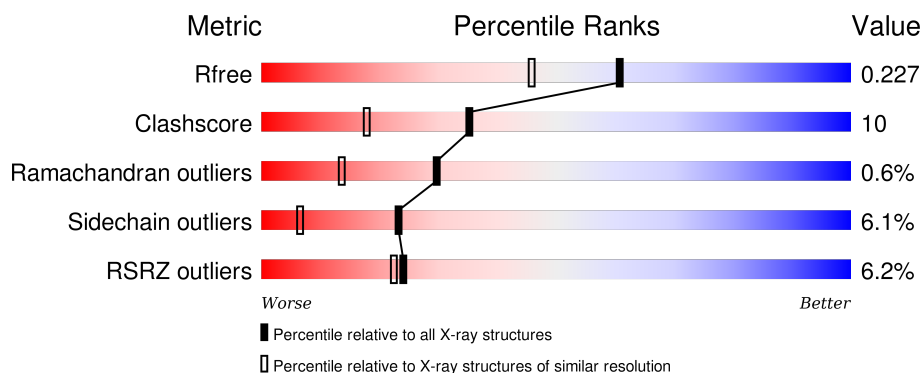
1 Overall quality at a glance

The following experimental techniques were used to determine the structure:

X-RAY DIFFRACTION

The reported resolution of this entry is 1.85 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}	91344	1745 (1.86-1.86)
Clashscore	102246	1898 (1.86-1.86)
Ramachandran outliers	100387	1875 (1.86-1.86)
Sidechain outliers	100360	1875 (1.86-1.86)
RSRZ outliers	91569	1747 (1.86-1.86)

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. 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 $\leq 5\%$. 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	286	<div> <div style="width: 100%; height: 10px; background: linear-gradient(to right, red 0%, red 26%, green 26%, green 79%, yellow 79%, yellow 97%, orange 97%, orange 100%);"></div> <div style="display: flex; justify-content: space-between; width: 100%;"> 26% 79% 17% </div> </div>
1	B	286	<div> <div style="width: 100%; height: 10px; background: linear-gradient(to right, red 0%, red 26%, green 26%, green 86%, yellow 86%, yellow 96%, orange 96%, orange 100%);"></div> <div style="display: flex; justify-content: space-between; width: 100%;"> 26% 86% 10% </div> </div>
2	C	146	<div> <div style="width: 100%; height: 10px; background: linear-gradient(to right, red 0%, red 26%, green 26%, green 68%, yellow 68%, yellow 95%, orange 95%, orange 100%);"></div> <div style="display: flex; justify-content: space-between; width: 100%;"> 26% 68% 27% </div> </div>

2 Entry composition [i](#)

There are 3 unique types of molecules in this entry. The entry contains 6631 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 Main hemagglutinin component.

Mol	Chain	Residues	Atoms					ZeroOcc	AltConf	Trace
1	A	285	Total	C	N	O	S	0	0	0
			2376	1494	408	469	5			
1	B	285	Total	C	N	O	S	0	0	0
			2376	1494	408	469	5			

There are 26 discrepancies between the modelled and reference sequences:

Chain	Residue	Modelled	Actual	Comment	Reference
A	1	MET	-	INITIATING METHIONINE	UNP P46084
A	60	THR	LYS	SEE REMARK 999	UNP P46084
A	74	ASP	ASN	SEE REMARK 999	UNP P46084
A	88	THR	GLY	SEE REMARK 999	UNP P46084
A	89	ASN	ASP	SEE REMARK 999	UNP P46084
A	98	ASP	ASN	SEE REMARK 999	UNP P46084
A	106	LEU	ILE	SEE REMARK 999	UNP P46084
A	120	THR	ILE	SEE REMARK 999	UNP P46084
A	125	ILE	MET	SEE REMARK 999	UNP P46084
A	131	ASN	SER	SEE REMARK 999	UNP P46084
A	133	ASN	SER	SEE REMARK 999	UNP P46084
A	187	THR	ILE	SEE REMARK 999	UNP P46084
A	262	THR	ILE	SEE REMARK 999	UNP P46084
B	1	MET	-	INITIATING METHIONINE	UNP P46084
B	60	THR	LYS	SEE REMARK 999	UNP P46084
B	74	ASP	ASN	SEE REMARK 999	UNP P46084
B	88	THR	GLY	SEE REMARK 999	UNP P46084
B	89	ASN	ASP	SEE REMARK 999	UNP P46084
B	98	ASP	ASN	SEE REMARK 999	UNP P46084
B	106	LEU	ILE	SEE REMARK 999	UNP P46084
B	120	THR	ILE	SEE REMARK 999	UNP P46084
B	125	ILE	MET	SEE REMARK 999	UNP P46084
B	131	ASN	SER	SEE REMARK 999	UNP P46084
B	133	ASN	SER	SEE REMARK 999	UNP P46084
B	187	THR	ILE	SEE REMARK 999	UNP P46084

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Chain	Residue	Modelled	Actual	Comment	Reference
B	262	THR	ILE	SEE REMARK 999	UNP P46084

- Molecule 2 is a protein called HA-17.

Mol	Chain	Residues	Atoms					ZeroOcc	AltConf	Trace
2	C	143	Total	C	N	O	S	0	0	0
			1161	746	185	229	1			

There is a discrepancy between the modelled and reference sequences:

Chain	Residue	Modelled	Actual	Comment	Reference
C	1	MET	-	INITIATING METHIONINE	UNP Q9LBR4

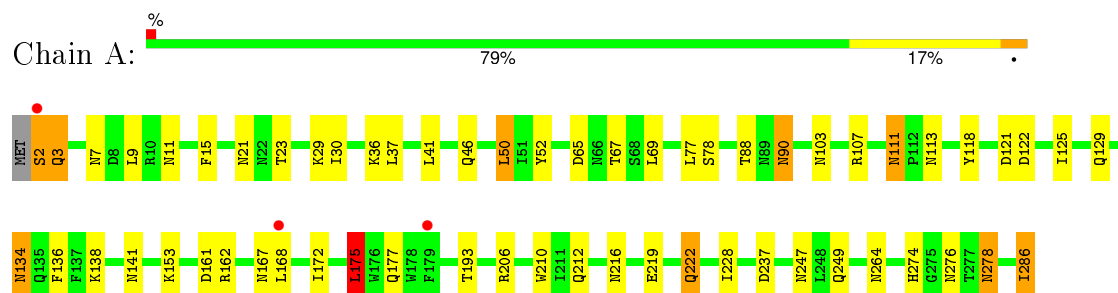
- Molecule 3 is water.

Mol	Chain	Residues	Atoms		ZeroOcc	AltConf
3	A	330	Total	O	0	0
			330	330		
3	B	323	Total	O	0	0
			323	323		
3	C	65	Total	O	0	0
			65	65		

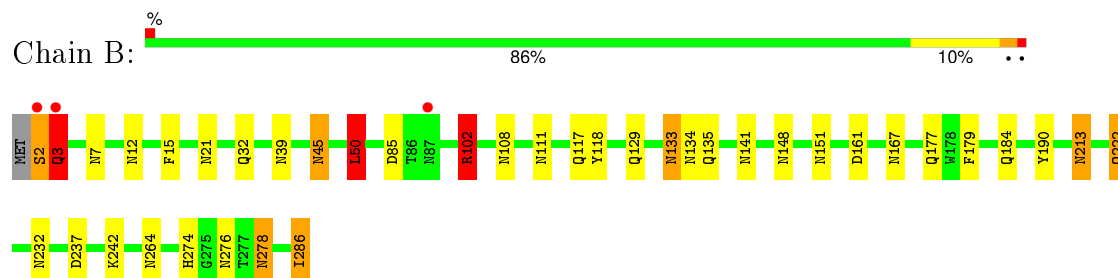
3 Residue-property plots [i](#)

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 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 ($\text{RSRZ} > 2$). 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.

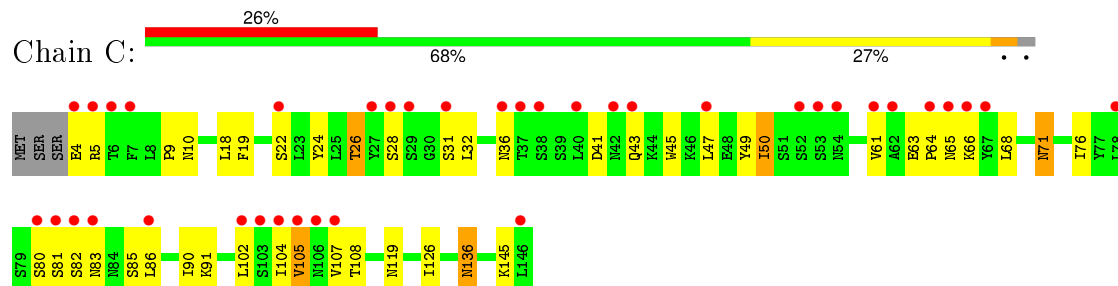
- Molecule 1: Main hemagglutinin component



- Molecule 1: Main hemagglutinin component



- Molecule 2: HA-17



4 Data and refinement statistics

Property	Value	Source
Space group	H 3 2	Depositor
Cell constants a, b, c, α , β , γ	239.13Å 239.13Å 100.02Å 90.00° 90.00° 120.00°	Depositor
Resolution (Å)	49.81 – 1.85 49.81 – 1.85	Depositor EDS
% Data completeness (in resolution range)	99.8 (49.81-1.85) 99.8 (49.81-1.85)	Depositor EDS
R_{merge}	(Not available)	Depositor
R_{sym}	(Not available)	Depositor
$\langle I/\sigma(I) \rangle$ ¹	2.86 (at 1.86Å)	Xtriage
Refinement program	REFMAC 5.2.0019	Depositor
R, R_{free}	0.194 , 0.229 0.192 , 0.227	Depositor DCC
R_{free} test set	4626 reflections (5.27%)	DCC
Wilson B-factor (Å ²)	25.0	Xtriage
Anisotropy	0.119	Xtriage
Bulk solvent k_{sol} (e/Å ³), B_{sol} (Å ²)	0.37 , 52.3	EDS
Estimated twinning fraction	No twinning to report.	Xtriage
L-test for twinning ²	$\langle L \rangle = 0.51$, $\langle L^2 \rangle = 0.34$	Xtriage
Outliers	0 of 92447 reflections	Xtriage
F_o, F_c correlation	0.96	EDS
Total number of atoms	6631	wwPDB-VP
Average B, all atoms (Å ²)	26.0	wwPDB-VP

Xtriage's analysis on translational NCS is as follows: *The largest off-origin peak in the Patterson function is 2.82% of the height of the origin peak. No significant pseudotranslation is detected.*

¹Intensities estimated from amplitudes.

²Theoretical values of $\langle |L| \rangle$, $\langle L^2 \rangle$ for acentric reflections are 0.5, 0.375 respectively for untwinned datasets, and 0.333, 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	0.77	0/2426	0.79	1/3309 (0.0%)
1	B	0.77	0/2426	0.82	4/3309 (0.1%)
2	C	0.57	0/1189	0.67	0/1620
All	All	0.74	0/6041	0.78	5/8238 (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
2	C	0	1

There are no bond length outliers.

All (5) bond angle outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(°)	Ideal(°)
1	B	102	ARG	NE-CZ-NH1	-14.62	112.99	120.30
1	B	102	ARG	NE-CZ-NH2	11.63	126.12	120.30
1	B	50	LEU	CB-CG-CD1	5.58	120.49	111.00
1	A	175	LEU	CB-CG-CD1	5.33	120.06	111.00
1	B	85	ASP	CB-CG-OD1	5.28	123.05	118.30

There are no chirality outliers.

All (1) planarity outliers are listed below:

Mol	Chain	Res	Type	Group
2	C	80	SER	Peptide

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	2376	0	2279	53	0
1	B	2376	0	2279	34	0
2	C	1161	0	1123	33	0
3	A	330	0	0	4	0
3	B	323	0	0	6	0
3	C	65	0	0	0	0
All	All	6631	0	5681	114	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 10.

The worst 5 of 114 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:C:82:SER:H	2:C:83:ASN:HA	0.96	1.11
1:A:125:ILE:HD11	3:A:364:HOH:O	1.56	1.04
2:C:63:GLU:HG2	2:C:66:LYS:HD2	1.47	0.97
1:A:41:LEU:H	1:A:46:GLN:HE22	1.09	0.96
2:C:82:SER:N	2:C:83:ASN:HA	1.73	0.94

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	283/286 (99%)	277 (98%)	4 (1%)	2 (1%)	26 11

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Mol	Chain	Analysed	Favoured	Allowed	Outliers	Percentiles	
1	B	283/286 (99%)	274 (97%)	7 (2%)	2 (1%)	26	11
2	C	141/146 (97%)	128 (91%)	13 (9%)	0	100	100
All	All	707/718 (98%)	679 (96%)	24 (3%)	4 (1%)	30	13

All (4) Ramachandran outliers are listed below:

Mol	Chain	Res	Type
1	A	3	GLN
1	B	237	ASP
1	A	237	ASP
1	B	3	GLN

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	274/275 (100%)	257 (94%)	17 (6%)	23	6
1	B	274/275 (100%)	260 (95%)	14 (5%)	29	11
2	C	135/138 (98%)	124 (92%)	11 (8%)	15	3
All	All	683/688 (99%)	641 (94%)	42 (6%)	23	6

5 of 42 residues with a non-rotameric sidechain are listed below:

Mol	Chain	Res	Type
1	B	3	GLN
1	B	102	ARG
2	C	71	ASN
1	B	21	ASN
1	B	45	ASN

Some sidechains can be flipped to improve hydrogen bonding and reduce clashes. 5 of 63 such sidechains are listed below:

Mol	Chain	Res	Type
1	B	66	ASN
1	B	133	ASN
2	C	88	ASN
1	B	89	ASN
1	B	108	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](#)

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](#)

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 [i](#)

6.1 Protein, DNA and RNA chains [i](#)

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	285/286 (99%)	-0.06	3 (1%) 82 82	15, 22, 31, 41	0
1	B	285/286 (99%)	-0.31	3 (1%) 82 82	14, 21, 31, 42	0
2	C	143/146 (97%)	1.29	38 (26%) 1 0	19, 41, 60, 80	0
All	All	713/718 (99%)	0.11	44 (6%) 24 22	14, 23, 49, 80	0

The worst 5 of 44 RSRZ outliers are listed below:

Mol	Chain	Res	Type	RSRZ
2	C	82	SER	9.1
2	C	103	SER	8.5
2	C	104	ILE	8.5
2	C	7	PHE	6.9
2	C	83	ASN	6.7

6.2 Non-standard residues in protein, DNA, RNA chains [i](#)

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

6.3 Carbohydrates [i](#)

There are no carbohydrates in this entry.

6.4 Ligands [i](#)

There are no ligands in this entry.

6.5 Other polymers [i](#)

There are no such residues in this entry.