



# Full wwPDB X-ray Structure Validation Report

Feb 26, 2014 – 09:59 PM GMT

PDB ID : 3R2X  
Title : Crystal structure of the de novo designed binding protein HB36.3 in complex  
the the 1918 influenza virus hemagglutinin  
Authors : Ekiert, D.C.; Wilson, I.A.  
Deposited on : 2011-03-14  
Resolution : 3.10 Å(reported)

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

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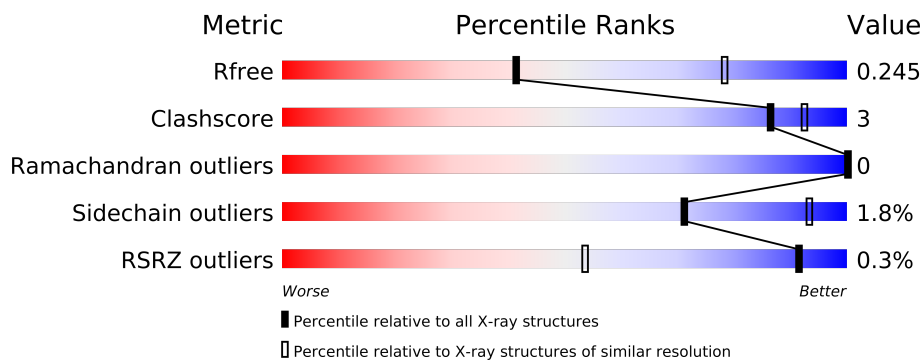
The following versions of software and data (see [references](#)) were used in the production of this report:

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 3.10 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	1007 (3.18-3.02)
Clashscore	79885	1078 (3.16-3.04)
Ramachandran outliers	78287	1044 (3.16-3.04)
Sidechain outliers	78261	1044 (3.16-3.04)
RSRZ outliers	66119	1008 (3.18-3.02)

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  $\geq 3$ , 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	331	
2	B	179	
3	C	93	

## 2 Entry composition

There are 5 unique types of molecules in this entry. The entry contains 4608 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 Hemagglutinin.

Mol	Chain	Residues	Atoms					ZeroOcc	AltConf	Trace
1	A	322	Total	C	N	O	S	0	0	0
			2500	1577	429	483	11			

There are 4 discrepancies between the modelled and reference sequences:

Chain	Residue	Modelled	Actual	Comment	Reference
A	7	ALA	-	EXPRESSION TAG	UNP Q9WFX3
A	8	ASP	-	EXPRESSION TAG	UNP Q9WFX3
A	9	PRO	-	EXPRESSION TAG	UNP Q9WFX3
A	10	GLY	-	EXPRESSION TAG	UNP Q9WFX3

- Molecule 2 is a protein called Hemagglutinin.

Mol	Chain	Residues	Atoms					ZeroOcc	AltConf	Trace
2	B	171	Total	C	N	O	S	0	0	0
			1377	860	237	274	6			

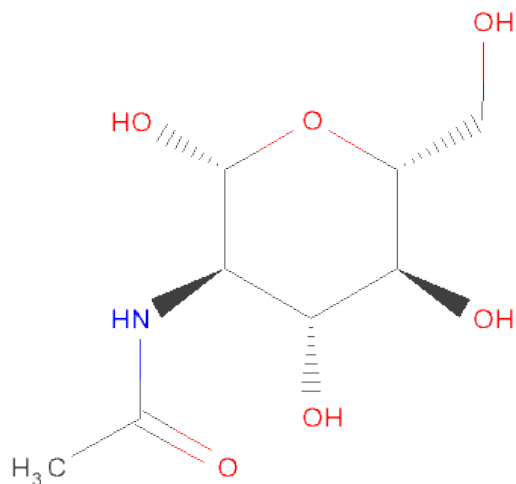
There are 3 discrepancies between the modelled and reference sequences:

Chain	Residue	Modelled	Actual	Comment	Reference
B	177	SER	-	EXPRESSION TAG	UNP Q9WFX3
B	178	GLY	-	EXPRESSION TAG	UNP Q9WFX3
B	179	ARG	-	EXPRESSION TAG	UNP Q9WFX3

- Molecule 3 is a protein called HB36.3, designed hemagglutinin binding protein.

Mol	Chain	Residues	Atoms					ZeroOcc	AltConf	Trace
3	C	82	Total	C	N	O	S	0	0	0
			667	436	112	117	2			

- Molecule 4 is SUGAR (N-ACETYL-D-GLUCOSAMINE) (three-letter code: NAG) (formula: C<sub>8</sub>H<sub>15</sub>NO<sub>6</sub>).



Mol	Chain	Residues	Atoms				ZeroOcc	AltConf
4	A	1	Total	C	N	O	0	0
			14	8	1	5		

- Molecule 5 is a polymer of unknown type called SUGAR (4-MER).

Mol	Chain	Residues	Atoms				ZeroOcc	AltConf
5	A	4	Total	C	N	O	0	0
			50	28	2	20		

There are 4 discrepancies between the modelled and reference sequences:

Chain	Residue	Modelled	Actual	Comment	Reference
A	7	ALA	-	EXPRESSION TAG	UNP Q9WFX3
A	8	ASP	-	EXPRESSION TAG	UNP Q9WFX3
A	9	PRO	-	EXPRESSION TAG	UNP Q9WFX3
A	10	GLY	-	EXPRESSION TAG	UNP Q9WFX3

### 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: Hemagglutinin

Chain A: 



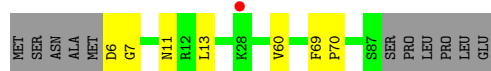
#### • Molecule 2: Hemagglutinin

Chain B: 



#### • Molecule 3: HB36.3, designed hemagglutinin binding protein

Chain C: 



## 4 Data and refinement statistics

Property	Value	Source
Space group	H 3 2	Depositor
Cell constants a, b, c, $\alpha$ , $\beta$ , $\gamma$	130.05Å 130.05Å 313.74Å 90.00° 90.00° 120.00°	Depositor
Resolution (Å)	45.74 – 3.10 45.75 – 2.87	Depositor EDS
% Data completeness (in resolution range)	99.5 (45.74-3.10) 96.6 (45.75-2.87)	Depositor EDS
$R_{merge}$	(Not available)	Depositor
$R_{sym}$	0.22	Depositor
$\langle I/\sigma(I) \rangle$ <sup>1</sup>	1.04 (at 2.86Å)	Xtriage
Refinement program	PHENIX (phenix.refine: 1.7_650)	Depositor
R, $R_{free}$	0.223 , 0.249 0.223 , 0.245	Depositor DCC
$R_{free}$ test set	1169 reflections (5.09%)	DCC
Wilson B-factor (Å <sup>2</sup> )	110.0	Xtriage
Anisotropy	0.139	Xtriage
Bulk solvent $k_{sol}$ (e/Å <sup>3</sup> ), $B_{sol}$ (Å <sup>2</sup> )	0.27 , 92.0	EDS
Estimated twinning fraction	0.016 for -h,1/3*h-1/3*k-1/3*l,-4/3*h-8/3*k+1/3*l 0.006 for -1/3*h+1/3*k+1/3*l,-k,8/3*h+4/3*k+1/3*l 0.026 for -2/3*h-1/3*k-1/3*l,-1/3*h-2/3*k+1/3*l,-4/3*h+4/3*k+1/3*l	Xtriage
L-test for twinning	$\langle  L  \rangle = 0.49$ , $\langle L^2 \rangle = 0.32$	Xtriage
Outliers	0 of 23054 reflections	Xtriage
$F_o, F_c$ correlation	0.95	EDS
Total number of atoms	4608	wwPDB-VP
Average B, all atoms (Å <sup>2</sup> )	167.0	wwPDB-VP

Xtriage's analysis on translational NCS is as follows: *The largest off-origin peak in the Patterson function is 5.38% of the height of the origin peak. No significant pseudotranslation is detected.*

<sup>1</sup>Intensities estimated from amplitudes.

## 5 Model quality ⓘ

### 5.1 Standard geometry ⓘ

Bond lengths and bond angles in the following residue types are not validated in this section: BMA, NAG, MAN

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	0.67	1/2564 (0.0%)	0.84	0/3493
2	B	0.69	0/1404	0.86	3/1890 (0.2%)
3	C	0.67	0/685	0.95	1/930 (0.1%)
All	All	0.68	1/4653 (0.0%)	0.86	4/6313 (0.1%)

All (1) bond length outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(Å)	Ideal(Å)
1	A	44	GLU	CG-CD	5.29	1.59	1.51

All (4) bond angle outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(°)	Ideal(°)
2	B	61	THR	N-CA-C	-5.63	95.80	111.00
2	B	98	LEU	CB-CG-CD1	-5.40	101.83	111.00
3	C	13	LEU	CA-CB-CG	5.05	126.92	115.30
2	B	89	LEU	CB-CG-CD2	-5.05	102.42	111.00

There are no chirality outliers.

There are no planarity outliers.

### 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	2500	0	0	4	0
2	B	1377	0	0	6	0
3	C	667	0	0	5	0
4	A	14	0	0	0	0
5	A	50	0	0	0	0
All	All	4608	0	0	12	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 3.

All (12) close contacts within the same asymmetric unit are listed below.

Atom-1	Atom-2	Distance(Å)	Clash(Å)
2:B:53:ASN:OD1	3:C:60:VAL:CG1	2.33	0.76
2:B:171:GLU:C	2:B:171:GLU:OE1	2.39	0.61
3:C:6:ASP:OD1	3:C:6:ASP:C	2.42	0.58
2:B:42:GLN:NE2	3:C:70:PRO:CD	2.67	0.58
1:A:48:ASN:ND2	1:A:48:ASN:C	2.58	0.57
1:A:114:SER:OG	1:A:262:ARG:NH1	2.39	0.55
3:C:7:GLY:O	3:C:11:ASN:N	2.42	0.53
2:B:38:GLN:O	2:B:42:GLN:N	2.47	0.48
1:A:118:PHE:C	1:A:118:PHE:CD1	2.91	0.44
2:B:45:ILE:CD1	3:C:69:PHE:CE1	3.04	0.41
1:A:131:GLU:OE2	1:A:133:THR:OG1	2.39	0.40
2:B:54:SER:O	2:B:58:LYS:CG	2.69	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	320/331 (97%)	313 (98%)	7 (2%)	0	100	100
2	B	169/179 (94%)	165 (98%)	4 (2%)	0	100	100
3	C	80/93 (86%)	80 (100%)	0	0	100	100
All	All	569/603 (94%)	558 (98%)	11 (2%)	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	278/285 (98%)	275 (99%)	3 (1%)	84	96
2	B	146/152 (96%)	140 (96%)	6 (4%)	41	82
3	C	66/76 (87%)	66 (100%)	0	100	100
All	All	490/513 (96%)	481 (98%)	9 (2%)	71	93

All (9) residues with a non-rotameric sidechain are listed below:

Mol	Chain	Res	Type
1	A	48	ASN
1	A	102	PHE
1	A	118	PHE
2	B	28	ASN
2	B	29	GLU
2	B	40	SER
2	B	128	ASN
2	B	145	ASP
2	B	171	GLU

Some sidechains can be flipped to improve hydrogen bonding and reduce clashes. There are no such sidechains identified.

### 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 ⓘ

4 carbohydrates 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$
5	NAG	A	601	1,5	12,14,15	0.79	1 (8%)	15,19,21	1.78	4 (26%)
5	NAG	A	602	5	12,14,15	0.56	0	15,19,21	1.61	2 (13%)
5	BMA	A	603	5	10,11,12	0.76	0	11,15,17	1.44	2 (18%)
5	MAN	A	604	5	10,11,12	0.80	0	11,15,17	1.49	3 (27%)

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
5	NAG	A	601	1,5	-	0/6/23/26	0/1/1/1
5	NAG	A	602	5	-	0/6/23/26	0/1/1/1
5	BMA	A	603	5	-	0/2/19/22	0/1/1/1
5	MAN	A	604	5	-	0/2/19/22	0/1/1/1

All (1) bond length outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(Å)	Ideal(Å)
5	A	601	NAG	O5-C5	-2.07	1.41	1.45

All (11) bond angle outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(°)	Ideal(°)
5	A	602	NAG	O5-C5-C6	5.02	112.25	106.98
5	A	601	NAG	O5-C5-C6	3.86	111.03	106.98
5	A	603	BMA	O3-C3-C2	-3.45	103.62	109.94
5	A	601	NAG	C3-C2-N2	-3.20	106.89	111.76
5	A	604	MAN	O5-C5-C6	3.03	110.16	106.98
5	A	601	NAG	O3-C3-C2	-2.88	103.05	109.09

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Mol	Chain	Res	Type	Atoms	Z	Observed(°)	Ideal(°)
5	A	604	MAN	O5-C5-C4	-2.49	107.49	110.65
5	A	604	MAN	C4-C3-C2	-2.37	107.33	110.50
5	A	602	NAG	C6-C5-C4	2.30	118.56	113.00
5	A	601	NAG	C6-C5-C4	-2.25	107.57	113.00
5	A	603	BMA	O5-C5-C6	2.12	109.20	106.98

There are no chirality outliers.

There are no torsion outliers.

There are no ring outliers.

## 5.6 Ligand geometry ⓘ

1 ligand is 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	$\# Z  > 2$
4	NAG	A	501	1	12,14,15	0.71	0	15,19,21	1.90	2 (13%)

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
4	NAG	A	501	1	-	0/6/23/26	0/1/1/1

There are no bond length outliers.

All (2) bond angle outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(°)	Ideal(°)
4	A	501	NAG	O5-C5-C6	6.06	113.34	106.98
4	A	501	NAG	O3-C3-C2	2.68	114.71	109.09

There are no chirality outliers.

There are no torsion outliers.

There are no ring outliers.

## 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<sup>th</sup> 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 <sup>2</sup> )	Q<0.9
1	A	322/331 (97%)	-0.19	0 100 100	102, 153, 235, 317	0
2	B	171/179 (95%)	-0.03	1 (0%) 86 36	90, 181, 254, 303	0
3	C	82/93 (88%)	-0.03	1 (1%) 75 20	119, 173, 219, 301	0
All	All	575/603 (95%)	-0.12	2 (0%) 91 53	90, 163, 247, 317	0

All (2) RSRZ outliers are listed below:

Mol	Chain	Res	Type	RSRZ
3	C	28	LYS	3.2
2	B	134	GLY	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 ⓘ

In the following table, the Atoms column lists the number of modelled atoms in the group and the number defined in the chemical component dictionary. LLDF column lists the quality of electron density of the group with respect to its neighbouring residues in protein, DNA or RNA chains. The B-factors column lists the minimum, median, 95<sup>th</sup> percentile and maximum values of B factors of atoms in the group. The column labelled ‘Q< 0.9’ lists the number of atoms with occupancy less than 0.9.

Mol	Type	Chain	Res	Atoms	RSR	LLDF	B-factors(Å <sup>2</sup> )	Q<0.9
5	MAN	A	604	11/12	0.09	-	272,275,279,284	0
5	BMA	A	603	11/12	0.07	-	221,227,232,235	0
5	NAG	A	602	14/15	0.10	-	194,197,200,201	0
5	NAG	A	601	14/15	0.10	-	156,159,163,164	0

## 6.4 Ligands

In the following table, the Atoms column lists the number of modelled atoms in the group and the number defined in the chemical component dictionary. LLDF column lists the quality of electron density of the group with respect to its neighbouring residues in protein, DNA or RNA chains. The B-factors column lists the minimum, median, 95<sup>th</sup> percentile and maximum values of B factors of atoms in the group. The column labelled 'Q< 0.9' lists the number of atoms with occupancy less than 0.9.

Mol	Type	Chain	Res	Atoms	RSR	LLDF	B-factors(Å <sup>2</sup> )	Q<0.9
4	NAG	A	501	14/15	0.20	-	202,208,215,216	0

## 6.5 Other polymers

There are no such residues in this entry.