



# wwPDB X-ray Structure Validation Summary Report

Feb 27, 2014 – 02:15 PM GMT

PDB ID : 4KRP  
Title : Nanobody/VHH domain 9G8 in complex with the extracellular region of EGFR  
Authors : Ferguson, K.M.; Schmitz, K.R.  
Deposited on : 2013-05-16  
Resolution : 2.82 Å(reported)

This is a wwPDB validation summary 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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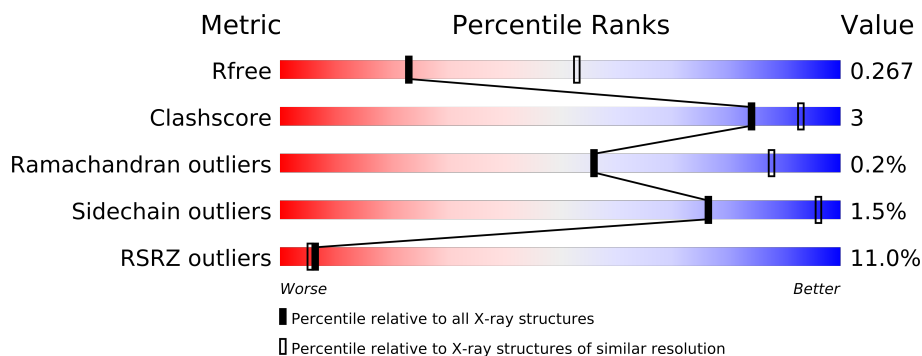
The following versions of software and data (see [references](#)) were used in the production of this report:

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 2.82 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	1963 (2.84-2.80)
Clashscore	79885	2478 (2.84-2.80)
Ramachandran outliers	78287	2429 (2.84-2.80)
Sidechain outliers	78261	2431 (2.84-2.80)
RSRZ outliers	66119	1966 (2.84-2.80)

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	625	
2	B	136	
3	C	211	
4	D	220	

## 2 Entry composition

There are 7 unique types of molecules in this entry. The entry contains 14877 atoms, of which 7017 are hydrogens 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 Epidermal growth factor receptor.

Mol	Chain	Residues	Atoms						ZeroOcc	AltConf	Trace
1	A	534	Total	C	H	N	O	S	0	0	0
			7001	2341	3252	651	712	45			

There are 7 discrepancies between the modelled and reference sequences:

Chain	Residue	Modelled	Actual	Comment	Reference
A	614	THR	-	EXPRESSION TAG	UNP P00533
A	620	HIS	-	EXPRESSION TAG	UNP P00533
A	621	HIS	-	EXPRESSION TAG	UNP P00533
A	622	HIS	-	EXPRESSION TAG	UNP P00533
A	623	HIS	-	EXPRESSION TAG	UNP P00533
A	624	HIS	-	EXPRESSION TAG	UNP P00533
A	625	HIS	-	EXPRESSION TAG	UNP P00533

- Molecule 2 is a protein called Nanobody/VHH domain 9G8.

Mol	Chain	Residues	Atoms						ZeroOcc	AltConf	Trace
2	B	116	Total	C	H	N	O	S	0	0	0
			1473	516	655	135	163	4			

- Molecule 3 is a protein called Cetuximab light chain.

Mol	Chain	Residues	Atoms						ZeroOcc	AltConf	Trace
3	C	211	Total	C	H	N	O	S	0	0	0
			3099	992	1506	270	327	4			

- Molecule 4 is a protein called Cetuximab heavy chain.

Mol	Chain	Residues	Atoms						ZeroOcc	AltConf	Trace
4	D	213	Total	C	H	N	O	S	0	0	0
			3079	1007	1494	259	314	5			

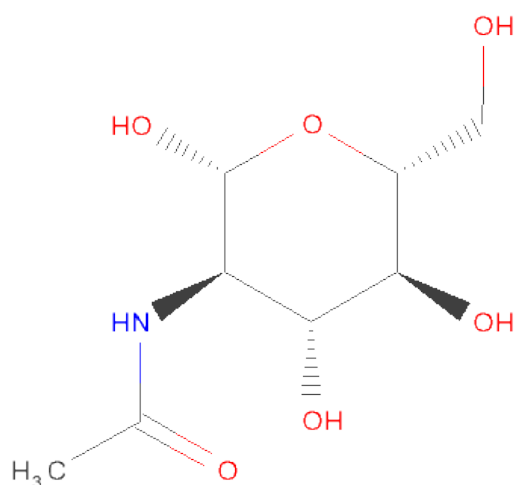
- Molecule 5 is a polymer of unknown type called SUGAR (2-MER).

Mol	Chain	Residues	Atoms					ZeroOcc	AltConf
5	A	2	Total	C	H	N	O	0	0
			55	16	27	2	10		
5	A	2	Total	C	H	N	O	0	0
			55	16	27	2	10		

There are 14 discrepancies between the modelled and reference sequences:

Chain	Residue	Modelled	Actual	Comment	Reference
A	614	THR	-	EXPRESSION TAG	UNP P00533
A	620	HIS	-	EXPRESSION TAG	UNP P00533
A	621	HIS	-	EXPRESSION TAG	UNP P00533
A	622	HIS	-	EXPRESSION TAG	UNP P00533
A	623	HIS	-	EXPRESSION TAG	UNP P00533
A	624	HIS	-	EXPRESSION TAG	UNP P00533
A	625	HIS	-	EXPRESSION TAG	UNP P00533
A	614	THR	-	EXPRESSION TAG	UNP P00533
A	620	HIS	-	EXPRESSION TAG	UNP P00533
A	621	HIS	-	EXPRESSION TAG	UNP P00533
A	622	HIS	-	EXPRESSION TAG	UNP P00533
A	623	HIS	-	EXPRESSION TAG	UNP P00533
A	624	HIS	-	EXPRESSION TAG	UNP P00533
A	625	HIS	-	EXPRESSION TAG	UNP P00533

- Molecule 6 is SUGAR (N-ACETYL-D-GLUCOSAMINE) (three-letter code: NAG) (formula:  $C_8H_{15}NO_6$ ).

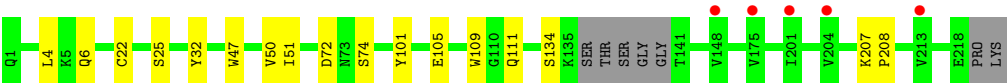


Mol	Chain	Residues	Atoms					ZeroOcc	AltConf
6	A	1	Total	C	H	N	O	0	0
			28	8	14	1	5		
6	A	1	Total	C	H	N	O	0	0
			28	8	14	1	5		
6	A	1	Total	C	H	N	O	0	0
			28	8	14	1	5		
6	D	1	Total	C	H	N	O	0	0
			28	8	14	1	5		

- Molecule 7 is water.

Mol	Chain	Residues	Atoms		ZeroOcc	AltConf
7	A	1	Total	O	0	0
			1	1		
7	C	1	Total	O	0	0
			1	1		
7	D	1	Total	O	0	0
			1	1		





## 4 Data and refinement statistics

Property	Value	Source
Space group	P 1 21 1	Depositor
Cell constants a, b, c, $\alpha$ , $\beta$ , $\gamma$	66.37Å 95.84Å 129.48Å 90.00° 99.95° 90.00°	Depositor
Resolution (Å)	44.46 – 2.82 44.46 – 2.82	Depositor EDS
% Data completeness (in resolution range)	98.0 (44.46-2.82) 98.0 (44.46-2.82)	Depositor EDS
$R_{merge}$	0.10	Depositor
$R_{sym}$	(Not available)	Depositor
$\langle I/\sigma(I) \rangle$ <sup>1</sup>	4.27 (at 2.81Å)	Xtriage
Refinement program	PHENIX (phenix.refine: 1.8.2_1309)	Depositor
R, $R_{free}$	0.215 , 0.263 0.220 , 0.267	Depositor DCC
$R_{free}$ test set	1890 reflections (5.00%)	DCC
Wilson B-factor (Å <sup>2</sup> )	51.9	Xtriage
Anisotropy	0.182	Xtriage
Bulk solvent $k_{sol}$ (e/Å <sup>3</sup> ), $B_{sol}$ (Å <sup>2</sup> )	0.31 , 25.4	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 37834 reflections	Xtriage
$F_o, F_c$ correlation	0.91	EDS
Total number of atoms	14877	wwPDB-VP
Average B, all atoms (Å <sup>2</sup> )	68.0	wwPDB-VP

Xtriage's analysis on translational NCS is as follows: *The largest off-origin peak in the Patterson function is 4.01% 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: NAG

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	0.27	0/3825	0.43	0/5230
2	B	0.26	0/838	0.39	0/1146
3	C	0.36	0/1627	0.52	0/2218
4	D	0.36	0/1627	0.52	0/2233
All	All	0.30	0/7917	0.47	0/10827

There are no bond length outliers.

There are no bond angle outliers.

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	3749	3252	86	23	0
2	B	818	655	0	4	0
3	C	1593	1506	0	11	0
4	D	1585	1494	1	9	0
5	A	56	54	0	1	0
6	A	42	42	0	1	0
6	D	14	14	0	0	0
7	A	1	0	0	0	0

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Mol	Chain	Non-H	H(model)	H(added)	Clashes	Symm-Clashes
7	C	1	0	0	0	0
7	D	1	0	0	0	0
All	All	7860	7017	87	44	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.

The worst 5 of 44 close contacts within the same asymmetric unit are listed below.

Atom-1	Atom-2	Distance(Å)	Clash(Å)
1:A:376:GLU:OE1	1:A:403:ARG:NH1	2.28	0.66
1:A:337:ASN:HD22	6:A:703:NAG:H81	1.61	0.65
1:A:455:LYS:O	2:B:27:ARG:NH2	2.31	0.64
1:A:403:ARG:NH2	2:B:100:TYR:O	2.31	0.64
2:B:54:SER:O	2:B:104:SER:OG	2.18	0.61

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	530/625 (85%)	478 (90%)	51 (10%)	1 (0%)	56	88
2	B	112/136 (82%)	96 (86%)	16 (14%)	0	100	100
3	C	209/211 (99%)	204 (98%)	4 (2%)	1 (0%)	38	75
4	D	209/220 (95%)	198 (95%)	11 (5%)	0	100	100
All	All	1060/1192 (89%)	976 (92%)	82 (8%)	2 (0%)	56	88

All (2) Ramachandran outliers are listed below:

Mol	Chain	Res	Type
1	A	214	ALA
3	C	138	ASN

### 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	368/546 (67%)	363 (99%)	5 (1%)	78	96
2	B	66/110 (60%)	64 (97%)	2 (3%)	53	86
3	C	179/188 (95%)	177 (99%)	2 (1%)	84	97
4	D	176/190 (93%)	173 (98%)	3 (2%)	73	95
All	All	789/1034 (76%)	777 (98%)	12 (2%)	76	96

5 of 12 residues with a non-rotameric sidechain are listed below:

Mol	Chain	Res	Type
2	B	27	ARG
2	B	53	TRP
4	D	25	SER
1	A	604	CYS
3	C	181	LEU

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

Mol	Chain	Res	Type
1	A	480	GLN

### 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	701	1,5	12,14,15	0.30	0	15,19,21	0.61	0
5	NAG	A	702	5	12,14,15	0.59	0	15,19,21	1.49	2 (13%)
5	NAG	A	705	1,5	12,14,15	0.58	0	15,19,21	1.51	2 (13%)
5	NAG	A	706	5	12,14,15	0.59	0	15,19,21	1.50	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
5	NAG	A	701	1,5	-	0/6/23/26	0/1/1/1
5	NAG	A	702	5	-	0/6/23/26	0/1/1/1
5	NAG	A	705	1,5	-	0/6/23/26	0/1/1/1
5	NAG	A	706	5	-	0/6/23/26	0/1/1/1

There are no bond length outliers.

The worst 5 of 6 bond angle outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(°)	Ideal(°)
5	A	706	NAG	O5-C5-C4	-3.69	105.97	110.65
5	A	705	NAG	O5-C5-C4	-3.67	105.99	110.65
5	A	702	NAG	O5-C5-C4	-3.66	106.01	110.65
5	A	705	NAG	C3-C2-N2	-2.54	107.89	111.76
5	A	702	NAG	C3-C2-N2	-2.53	107.91	111.76

There are no chirality outliers.

There are no torsion outliers.

There are no ring outliers.

## 5.6 Ligand geometry ⓘ

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	$\# Z  > 2$
6	NAG	A	703	1	12,14,15	0.25	0	15,19,21	0.47	0
6	NAG	A	704	1	12,14,15	0.59	0	15,19,21	1.49	2 (13%)
6	NAG	A	707	1	12,14,15	0.22	0	15,19,21	0.39	0
6	NAG	D	301	4	12,14,15	0.33	0	15,19,21	0.41	0

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
6	NAG	A	703	1	-	0/6/23/26	0/1/1/1
6	NAG	A	704	1	-	0/6/23/26	0/1/1/1
6	NAG	A	707	1	-	0/6/23/26	0/1/1/1
6	NAG	D	301	4	-	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(°)
6	A	704	NAG	O5-C5-C4	-3.68	105.98	110.65
6	A	704	NAG	C3-C2-N2	-2.50	107.96	111.76

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	534/625 (85%)	0.66	86 (16%) 2 2	25, 85, 151, 166	0
2	B	116/136 (85%)	1.01	28 (24%) 1 1	59, 94, 124, 137	0
3	C	211/211 (100%)	-0.18	0 100 100	18, 36, 56, 70	0
4	D	213/220 (96%)	-0.16	5 (2%) 57 58	18, 34, 53, 84	0
All	All	1074/1192 (90%)	0.37	119 (11%) 6 5	18, 53, 143, 166	0

The worst 5 of 119 RSRZ outliers are listed below:

Mol	Chain	Res	Type	RSRZ
1	A	82	ILE	8.7
1	A	49	ASN	7.6
1	A	24	PHE	7.4
1	A	126	PHE	7.3
1	A	124	VAL	7.2

### 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( $\text{\AA}^2$ )	Q<0.9
5	NAG	A	702	14/15	0.27	1.10	66,92,111,114	0
5	NAG	A	701	14/15	0.15	-0.52	53,79,96,104	0
5	NAG	A	705	14/15	0.12	-1.99	43,58,70,77	0
5	NAG	A	706	14/15	0.21	-	69,89,105,118	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( $\text{\AA}^2$ )	Q<0.9
6	NAG	D	301	14/15	0.14	0.85	64,83,97,112	0
6	NAG	A	704	14/15	0.21	0.52	71,97,116,121	0
6	NAG	A	703	14/15	0.18	0.18	59,95,116,120	0
6	NAG	A	707	14/15	0.19	-0.06	60,85,102,106	0

## 6.5 Other polymers

There are no such residues in this entry.