



wwPDB EM Validation Summary Report ⓘ

Apr 26, 2026 – 12:15 AM JST

PDB ID : 9VI9 / pdb_00009vi9
EMDB ID : EMD-65081
Title : Cryo-EM Structure of NPFFR1 in complex with peptide RFRP-3
Authors : Na, M.; Xu, F.
Deposited on : 2025-06-17
Resolution : 3.02 Å(reported)
Based on initial model : .

This is a wwPDB EM Validation Summary Report for a publicly released PDB entry.

We welcome your comments at validation@mail.wwpdb.org

A user guide is available at

<https://www.wwpdb.org/validation/2017/EMValidationReportHelp>
with specific help available everywhere you see the ⓘ symbol.

The types of validation reports are described at

<http://www.wwpdb.org/validation/2017/FAQs#types>.

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

EMDB validation analysis : **FAILED**
MolProbity : 4-5-2 with Phenix2.0
Percentile statistics : 20250101.v01 (using entries in the PDB archive January 1st 2025)
EM percentile statistics : **NOT EXECUTED**
MapQ : **FAILED**
Ideal geometry (proteins) : Engh & Huber (2001)
Ideal geometry (DNA, RNA) : Parkinson et al. (1996)
Validation Pipeline (wwPDB-VP) : 2.49

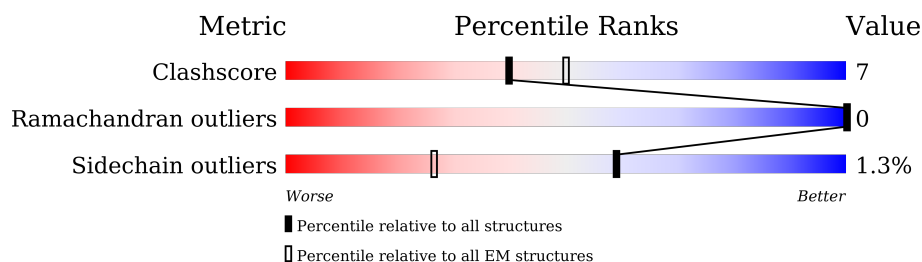
1 Overall quality at a glance

The following experimental techniques were used to determine the structure:

ELECTRON MICROSCOPY

The reported resolution of this entry is 3.02 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	229148	23984
Ramachandran outliers	224038	23583
Sidechain outliers	223484	23102

The table below summarises the geometric issues observed across the polymeric chains and their fit to the map. The red, orange, yellow and green segments of the bar indicate the fraction of residues that contain outliers for ≥ 3 , 2, 1 and 0 types of geometric quality criteria respectively. 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 $\leq 5\%$.

Mol	Chain	Length	Quality of chain
1	A	310	
2	B	349	
3	C	338	
4	D	54	
5	P	9	
6	S	248	

2 Entry composition

There are 6 unique types of molecules in this entry. The entry contains 9025 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 Neuropeptide FF receptor 1.

Mol	Chain	Residues	Atoms					AltConf	Trace
1	A	298	Total	C	N	O	S	0	0
			2394	1582	405	386	21		

There is a discrepancy between the modelled and reference sequences:

Chain	Residue	Modelled	Actual	Comment	Reference
A	132	TRP	PHE	conflict	UNP Q9GZQ6

- Molecule 2 is a protein called Guanine nucleotide-binding protein G(i) subunit alpha-1.

Mol	Chain	Residues	Atoms					AltConf	Trace
2	B	217	Total	C	N	O	S	0	0
			1754	1117	293	331	13		

There are 3 discrepancies between the modelled and reference sequences:

Chain	Residue	Modelled	Actual	Comment	Reference
B	47	ASN	SER	conflict	UNP P63096
B	203	ALA	GLY	conflict	UNP P63096
B	326	SER	ALA	conflict	UNP P63096

- Molecule 3 is a protein called Guanine nucleotide-binding protein G(I)/G(S)/G(T) subunit beta-1.

Mol	Chain	Residues	Atoms					AltConf	Trace
3	C	338	Total	C	N	O	S	0	0
			2601	1604	467	509	21		

- Molecule 4 is a protein called Guanine nucleotide-binding protein G(I)/G(S)/G(O) subunit gamma-2.

Mol	Chain	Residues	Atoms					AltConf	Trace
4	D	54	Total	C	N	O	S	0	0
			413	260	71	79	3		

- Molecule 5 is a protein called RFRP-3.

Mol	Chain	Residues	Atoms				AltConf	Trace
5	P	9	Total	C	N	O	0	1
			69	45	14	10		

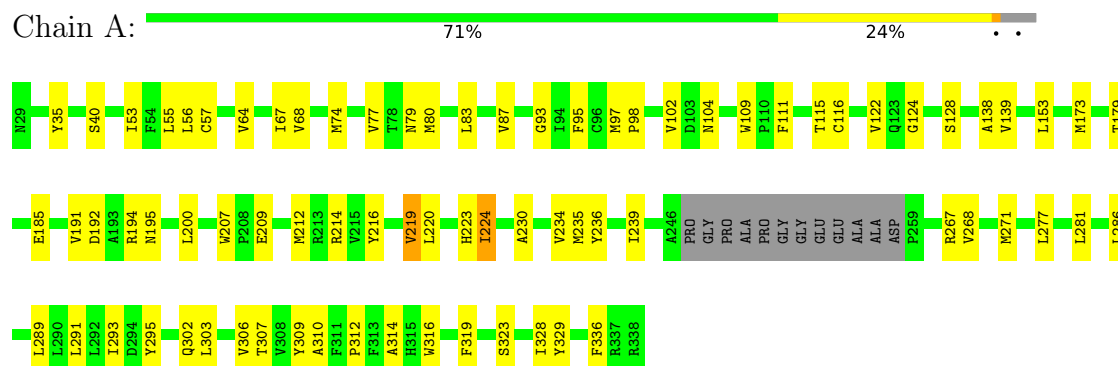
- Molecule 6 is a protein called scFv16.

Mol	Chain	Residues	Atoms					AltConf	Trace
6	S	233	Total	C	N	O	S	0	0
			1794	1138	297	349	10		

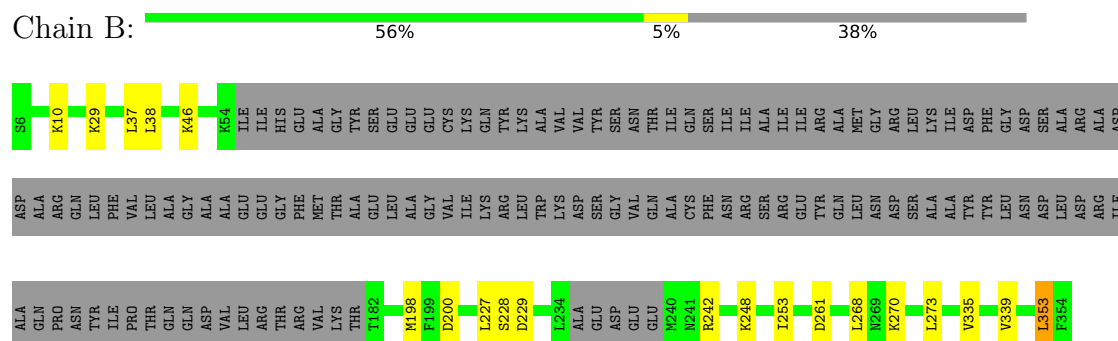
3 Residue-property plots

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

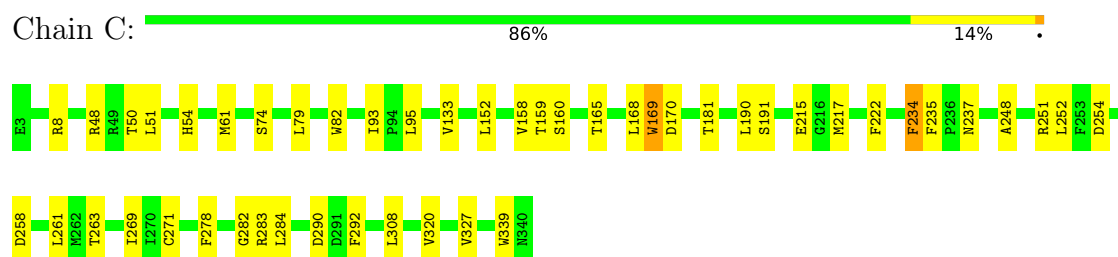
• Molecule 1: Neuropeptide FF receptor 1



• Molecule 2: Guanine nucleotide-binding protein G(i) subunit alpha-1



• Molecule 3: Guanine nucleotide-binding protein G(I)/G(S)/G(T) subunit beta-1

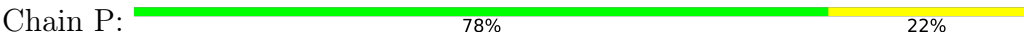


• Molecule 4: Guanine nucleotide-binding protein G(I)/G(S)/G(O) subunit gamma-2

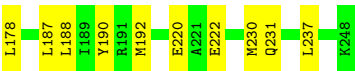
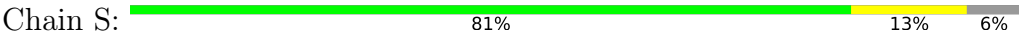




● Molecule 5: RFRP-3



● Molecule 6: scFv16



4 Experimental information

Property	Value	Source
EM reconstruction method	SINGLE PARTICLE	Depositor
Imposed symmetry	POINT, Not provided	
Number of particles used	140756	Depositor
Resolution determination method	FSC 0.143 CUT-OFF	Depositor
CTF correction method	PHASE FLIPPING ONLY	Depositor
Microscope	FEI TALOS ARCTICA	Depositor
Voltage (kV)	300	Depositor
Electron dose ($e^-/\text{\AA}^2$)	60	Depositor
Minimum defocus (nm)	700	Depositor
Maximum defocus (nm)	2200	Depositor
Magnification	Not provided	
Image detector	FEI FALCON IV (4k x 4k)	Depositor

5 Model quality [i](#)

5.1 Standard geometry [i](#)

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	0.19	0/2462	0.43	0/3358
2	B	0.14	0/1783	0.36	0/2390
3	C	0.17	0/2648	0.43	0/3589
4	D	0.20	0/419	0.51	0/566
5	P	0.36	0/71	0.61	0/97
6	S	0.13	0/1838	0.38	0/2491
All	All	0.17	0/9221	0.41	0/12491

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	2394	0	2456	50	0
2	B	1754	0	1749	14	0
3	C	2601	0	2505	33	0
4	D	413	0	423	13	0
5	P	69	0	72	1	0
6	S	1794	0	1736	20	0
All	All	9025	0	8941	127	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 7.

The worst 5 of 127 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)
3:C:254:ASP:HB2	3:C:261:LEU:HD11	1.68	0.74
3:C:284:LEU:HD12	4:D:79:ASP:HB3	1.68	0.74
6:S:220:GLU:HG3	6:S:222:GLU:H	1.55	0.70
1:A:173:MET:HE1	1:A:220:LEU:HD11	1.75	0.69
1:A:267:ARG:HH12	1:A:271:MET:HB2	1.59	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	294/310 (95%)	288 (98%)	6 (2%)	0	100	100
2	B	211/349 (60%)	206 (98%)	5 (2%)	0	100	100
3	C	336/338 (99%)	325 (97%)	11 (3%)	0	100	100
4	D	52/54 (96%)	50 (96%)	2 (4%)	0	100	100
5	P	7/9 (78%)	6 (86%)	1 (14%)	0	100	100
6	S	229/248 (92%)	214 (93%)	15 (7%)	0	100	100
All	All	1129/1308 (86%)	1089 (96%)	40 (4%)	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	259/266 (97%)	253 (98%)	6 (2%)	44	72
2	B	194/302 (64%)	192 (99%)	2 (1%)	68	83
3	C	281/281 (100%)	278 (99%)	3 (1%)	65	82
4	D	44/44 (100%)	42 (96%)	2 (4%)	24	57
5	P	8/8 (100%)	8 (100%)	0	100	100
6	S	198/201 (98%)	198 (100%)	0	100	100
All	All	984/1102 (89%)	971 (99%)	13 (1%)	59	80

5 of 13 residues with a non-rotameric sidechain are listed below:

Mol	Chain	Res	Type
2	B	353	LEU
3	C	50	THR
4	D	77	LYS
3	C	234	PHE
4	D	69	MET

Sometimes sidechains can be flipped to improve hydrogen bonding and reduce clashes. 5 of 10 such sidechains are listed below:

Mol	Chain	Res	Type
4	D	75	HIS
4	D	90	ASN
5	P	6	GLN
2	B	204	GLN
2	B	331	ASN

5.3.3 RNA ⓘ

There are no RNA molecules 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 [i](#)

There are no oligosaccharides 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.