wwPDB EM Validation Summary Report

Dec 24, 2023 – 12:07 AM JST

PDB ID	:	8W89
EMDB ID	:	EMD-37349
Title	:	Cryo-EM structure of the PEA-bound TAAR1-Gs complex
Authors	:	Liu, H.; Zheng, Y.; Wang, Y.; Wang, Y.; He, X.; Xu, P.; Huang, S.; Yuan, Q.; Zhang, X.; Wang, S.; Xu, H.E.; Xu, F.
Deposited on	:	2023-09-01
Resolution	:	3.00 Å(reported)

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:

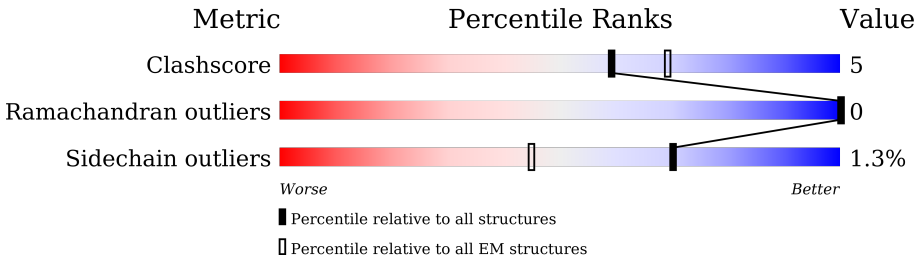
EMDB validation analysis	:	FAILED
Mogul	:	1.8.5 (274361), CSD as541be (2020)
MolProbity	:	4.02b-467
buster-report	:	1.1.7 (2018)
Percentile statistics	:	20191225.v01 (using entries in the PDB archive December 25th 2019)
MapQ	:	FAILED
Ideal geometry (proteins)	:	Engh & Huber (2001)
Ideal geometry (DNA, RNA)	:	Parkinson et al. (1996)
Validation Pipeline (wwPDB-VP)	:	2.36

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 3.00 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	158937	4297
Ramachandran outliers	154571	4023
Sidechain outliers	154315	3826

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 <=5%

Mol	Chain	Length	Quality of chain
1	A	246	89% 7% .
2	B	345	84% 14% .
3	G	71	76% . 20%
4	N	139	80% 12% 9%
5	R	339	71% 12% . 16%

2 Entry composition (i)

There are 7 unique types of molecules in this entry. The entry contains 8230 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 Guanine nucleotide-binding protein G(s) subunit alpha isoforms short.

Mol	Chain	Residues	Atoms					AltConf	Trace
			Total	C	N	O	S		
1	A	236	1934	1216	349	362	7	0	0

- Molecule 2 is a protein called Guanine nucleotide-binding protein G(I)/G(S)/G(T) subunit beta-1.

Mol	Chain	Residues	Atoms					AltConf	Trace
			Total	C	N	O	S		
2	B	338	2600	1604	467	508	21	0	0

There are 6 discrepancies between the modelled and reference sequences:

Chain	Residue	Modelled	Actual	Comment	Reference
B	-4	MET	-	initiating methionine	UNP P62873
B	-3	GLY	-	expression tag	UNP P62873
B	-2	SER	-	expression tag	UNP P62873
B	-1	LEU	-	expression tag	UNP P62873
B	0	LEU	-	expression tag	UNP P62873
B	1	GLN	-	expression tag	UNP P62873

- Molecule 3 is a protein called Guanine nucleotide-binding protein G(I)/G(S)/G(O) subunit gamma-2.

Mol	Chain	Residues	Atoms					AltConf	Trace
			Total	C	N	O	S		
3	G	57	436	273	77	83	3	0	0

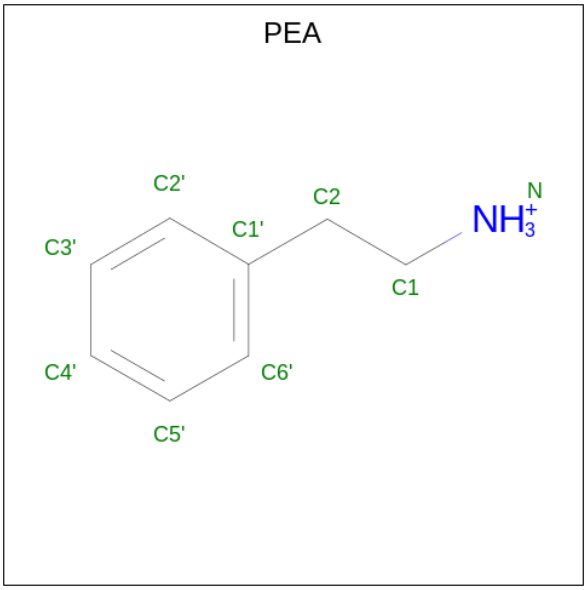
- Molecule 4 is a protein called Nanobody35.

Mol	Chain	Residues	Atoms					AltConf	Trace
			Total	C	N	O	S		
4	N	127	967	602	169	190	6	0	0

- Molecule 5 is a protein called Trace amine-associated receptor 1.

Mol	Chain	Residues	Atoms					AltConf	Trace
			Total	C	N	O	S		
5	R	285	2283	1526	356	377	24	0	0

- Molecule 6 is 2-PHENYLETHYLAMINE (three-letter code: PEA) (formula: $C_8H_{12}N$) (labeled as "Ligand of Interest" by depositor).



Mol	Chain	Residues	Atoms				AltConf
			Total	C	N		
6	R	1	9	8	1		0

- Molecule 7 is water.

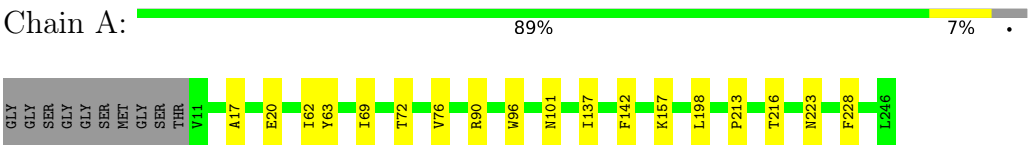
Mol	Chain	Residues	Atoms			AltConf
			Total	O		
7	R	1	1	1		0

3 Residue-property plots (i)

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.

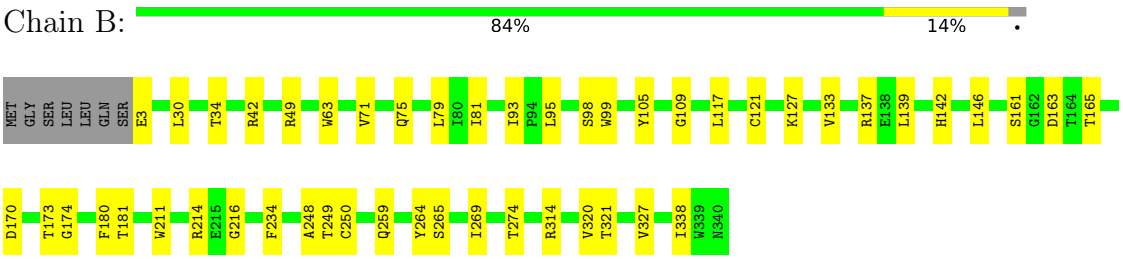
• Molecule 1: Guanine nucleotide-binding protein G(s) subunit alpha isoforms short

Chain A:



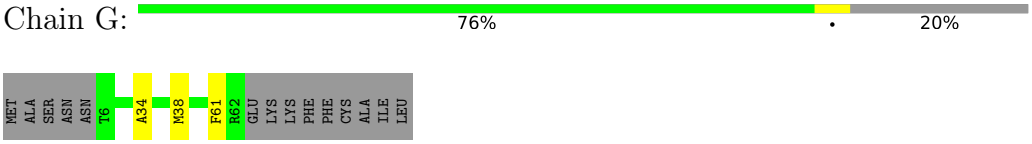
• Molecule 2: Guanine nucleotide-binding protein G(I)/G(S)/G(T) subunit beta-1

Chain B:



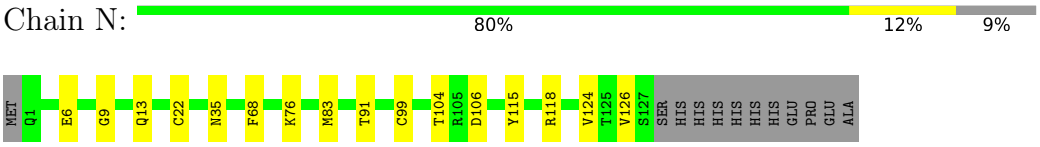
• Molecule 3: Guanine nucleotide-binding protein G(I)/G(S)/G(O) subunit gamma-2

Chain G:



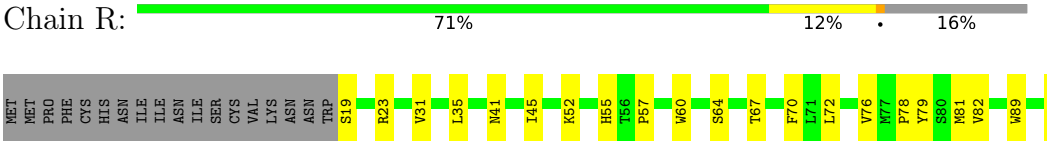
• Molecule 4: Nanobody35

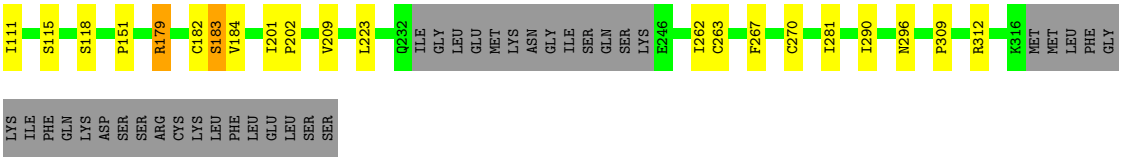
Chain N:



• Molecule 5: Trace amine-associated receptor 1

Chain R:




I111
S115
S118
P151
R179
C182
S183
V184
I201
P202
V209
L223
Q232
ILE
GLY
LEU
GLU
MET
LYS
ASN
GLY
ILE
SER
GLN
SER
LYS
E246
I262
C263
F267
C270
I281
I290
N296
P309
R312
K316
MET
MET
LEU
PHE
GLY
LYS
ILE
PHE
GLN
LYS
ASP
SER
SER
ARG
CYS
LYS
LEU
PHE
LEU
GLU
LEU
SER
SER

4 Experimental information (i)

Property	Value	Source
EM reconstruction method	SINGLE PARTICLE	Depositor
Imposed symmetry	POINT, Not provided	
Number of particles used	87235	Depositor
Resolution determination method	FSC 0.143 CUT-OFF	Depositor
CTF correction method	NONE	Depositor
Microscope	FEI TITAN KRIOS	Depositor
Voltage (kV)	300	Depositor
Electron dose ($e^-/Å^2$)	60	Depositor
Minimum defocus (nm)	1000	Depositor
Maximum defocus (nm)	2000	Depositor
Magnification	Not provided	
Image detector	GATAN K3 (6k x 4k)	Depositor

5 Model quality

5.1 Standard geometry

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

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	0.32	0/1973	0.51	0/2659
2	B	0.36	0/2647	0.57	0/3589
3	G	0.26	0/442	0.45	0/597
4	N	0.34	0/987	0.51	0/1337
5	R	0.36	0/2350	0.51	0/3194
All	All	0.34	0/8399	0.53	0/11376

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

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	1934	0	1891	14	0
2	B	2600	0	2505	32	0
3	G	436	0	448	4	0
4	N	967	0	933	10	0
5	R	2283	0	2302	25	0
6	R	9	0	12	0	0
7	R	1	0	0	0	0
All	All	8230	0	8091	78	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 5.

The worst 5 of 78 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:B:250:CYS:HB2	2:B:264:TYR:HB2	1.82	0.61
5:R:179:ARG:HG3	5:R:183:SER:HB3	1.84	0.58
5:R:108:SER:HB2	5:R:151:PRO:HB3	1.90	0.53
5:R:72:LEU:HD12	5:R:76:VAL:HB	1.90	0.53
2:B:79:LEU:HB2	2:B:95:LEU:HD21	1.92	0.53

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	234/246 (95%)	225 (96%)	9 (4%)	0	100	100
2	B	336/345 (97%)	323 (96%)	13 (4%)	0	100	100
3	G	55/71 (78%)	55 (100%)	0	0	100	100
4	N	125/139 (90%)	120 (96%)	5 (4%)	0	100	100
5	R	281/339 (83%)	273 (97%)	8 (3%)	0	100	100
All	All	1031/1140 (90%)	996 (97%)	35 (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	207/213 (97%)	206 (100%)	1 (0%)	88 96
2	B	281/287 (98%)	276 (98%)	5 (2%)	59 85
3	G	46/58 (79%)	46 (100%)	0	100 100
4	N	105/116 (90%)	105 (100%)	0	100 100
5	R	252/309 (82%)	246 (98%)	6 (2%)	49 79
All	All	891/983 (91%)	879 (99%)	12 (1%)	70 89

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

Mol	Chain	Res	Type
5	R	23	ARG
5	R	179	ARG
5	R	184	VAL
5	R	182	CYS
2	B	214	ARG

Sometimes sidechains can be flipped to improve hydrogen bonding and reduce clashes. 5 of 6 such sidechains are listed below:

Mol	Chain	Res	Type
2	B	75	GLN
2	B	88	ASN
5	R	53	GLN
1	A	146	GLN
1	A	119	GLN

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 monosaccharides in this entry.

5.6 Ligand geometry (i)

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	$\#\|Z\| > 2$
6	PEA	R	401	-	8,9,9	0.51	0	9,10,10	0.78	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	PEA	R	401	-	-	1/3/3/3	0/1/1/1

There are no bond length outliers.

There are no bond angle outliers.

There are no chirality outliers.

All (1) torsion outliers are listed below:

Mol	Chain	Res	Type	Atoms
6	R	401	PEA	N-C1-C2-C1'

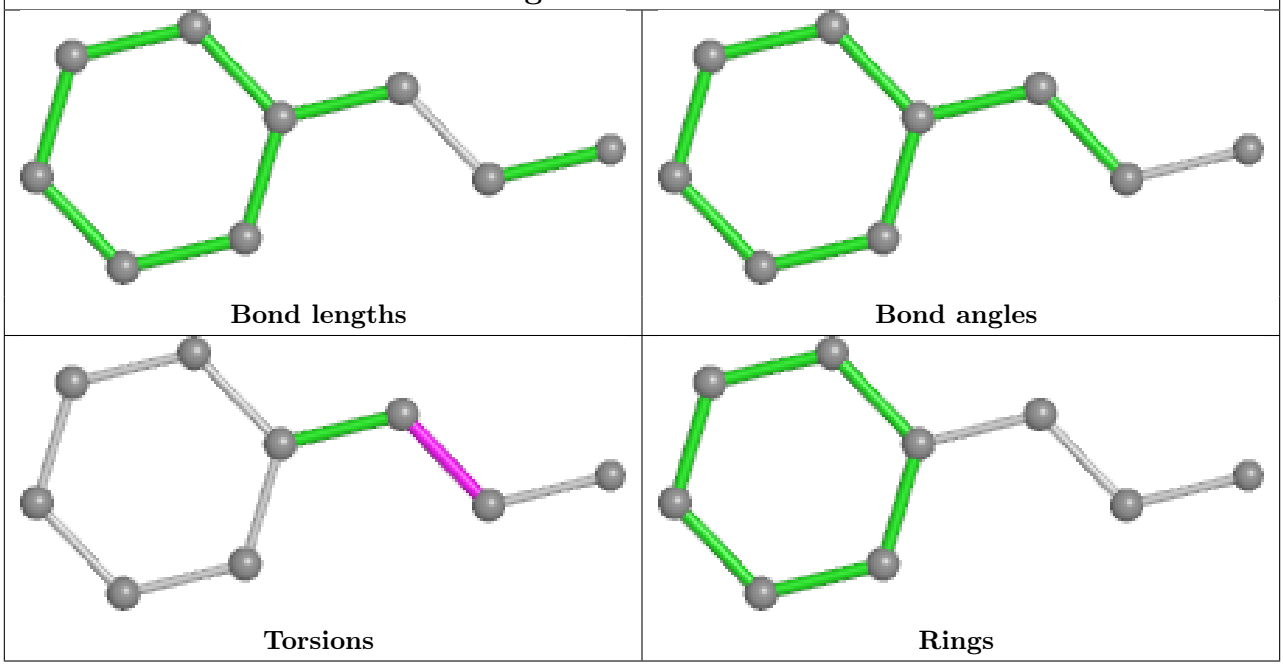
There are no ring outliers.

No monomer is involved in short contacts.

The following is a two-dimensional graphical depiction of Mogul quality analysis of bond lengths, bond angles, torsion angles, and ring geometry for all instances of the Ligand of Interest. In addition, ligands with molecular weight > 250 and outliers as shown on the validation Tables will also be included. For torsion angles, if less then 5% of the Mogul distribution of torsion angles is within 10 degrees of the torsion angle in question, then that torsion angle is considered an outlier. Any bond that is central to one or more torsion angles identified as an outlier by Mogul will be highlighted in the graph. For rings, the root-mean-square deviation (RMSD) between the ring in question and similar rings identified by Mogul is calculated over all ring torsion angles. If the average RMSD is greater than 60 degrees and the minimal RMSD between the ring in question and

any Mogul-identified rings is also greater than 60 degrees, then that ring is considered an outlier. The outliers are highlighted in purple. The color gray indicates Mogul did not find sufficient equivalents in the CSD to analyse the geometry.

Ligand PEA R 401



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.