

Full wwPDB X-ray Structure Validation Report (i)

Apr 4, 2023 – 10:17 am BST

PDB ID : 7ZL9

Title: Crystal structure of human GPCR Niacin receptor (HCA2)

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Deposited on : 2022-04-14

Resolution : 2.70 Å(reported)

This is a Full wwPDB X-ray Structure Validation 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/XrayValidationReportHelp
with specific help available everywhere you see the (i) 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 (1)) were used in the production of this report:

 $Mol Probity \quad : \quad 4.02b\text{--}467$

Mogul : 1.8.4, CSD as541be (2020)

Xtriage (Phenix) : 1.13

EDS : 2.32.2buster-report : 1.1.7 (2018)

Percentile statistics : 20191225.v01 (using entries in the PDB archive December 25th 2019)

Refmac : 5.8.0158

CCP4 : 7.0.044 (Gargrove)

Ideal geometry (proteins) : Engh & Huber (2001) Ideal geometry (DNA, RNA) : Parkinson et al. (1996)

Validation Pipeline (wwPDB-VP) : 2.32.2

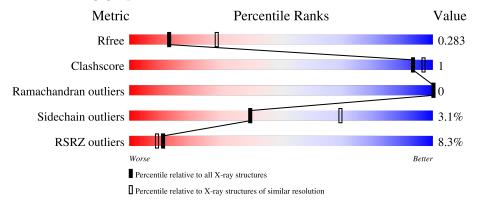


1 Overall quality at a glance (i)

The following experimental techniques were used to determine the structure: X- $RAY\ DIFFRACTION$

The reported resolution of this entry is 2.70 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.



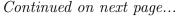
Metric	Whole archive $(\# \mathrm{Entries})$	$\begin{array}{c} {\rm Similar\ resolution} \\ (\#{\rm Entries},{\rm resolution\ range}({\rm \AA})) \end{array}$
R_{free}	130704	2808 (2.70-2.70)
Clashscore	141614	3122 (2.70-2.70)
Ramachandran outliers	138981	3069 (2.70-2.70)
Sidechain outliers	138945	3069 (2.70-2.70)
RSRZ outliers	127900	2737 (2.70-2.70)

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 of the lower 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% 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					
			8%					
1	A	431	86%	6%	8%			

The following table lists non-polymeric compounds, carbohydrate monomers and non-standard residues in protein, DNA, RNA chains that are outliers for geometric or electron-density-fit criteria:

Mol	Type	Chain	Res	Chirality	Geometry	Clashes	Electron density
5	UNX	A	1211	-	-	-	X





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Mol	Type	Chain	Res	Chirality	Geometry	Clashes	Electron density
5	UNX	A	1212	_	-	=	X



2 Entry composition (i)

There are 6 unique types of molecules in this entry. The entry contains 3196 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.

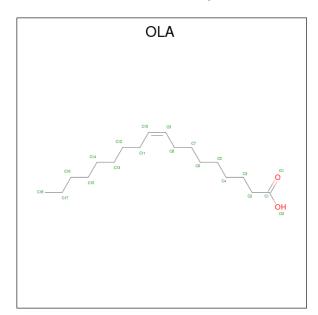
• Molecule 1 is a protein called Hydroxycarboxylic acid receptor 2, Soluble cytochrome b562.

Mol	Chain	Residues		Atoms			ZeroOcc	AltConf	Trace	
1	Λ	398	Total	С	N	О	S	0	0	0
1	A	390	3100	2041	512	524	23	0	0	

There are 5 discrepancies between the modelled and reference sequences:

Chain	Residue	Modelled	Actual	Comment	Reference
A	70	VAL	ALA	conflict	UNP Q8TDS4
A	287	VAL	SER	conflict	UNP Q8TDS4
A	1007	TRP	MET	conflict	UNP P0ABE7
A	1102	ILE	HIS	conflict	UNP P0ABE7
A	1106	LEU	ARG	conflict	UNP P0ABE7

• Molecule 2 is OLEIC ACID (three-letter code: OLA) (formula: $C_{18}H_{34}O_2$).



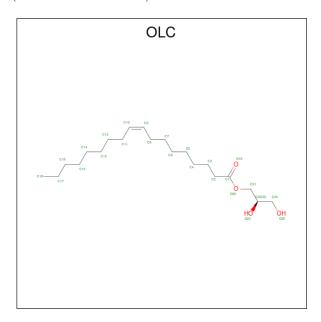
\mathbf{Mol}	Chain	Residues	Atoms			ZeroOcc	AltConf
2	A	1	Total 18	C 16	O 2	0	0

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Mol	Chain	Residues	Atoms	ZeroOcc	AltConf
2	A	1	Total C 7 7	0	0
2	A	1	Total C 7 7	0	0
2	A	1	Total C 8 8	0	0
2	A	1	Total C O 16 14 2	0	0

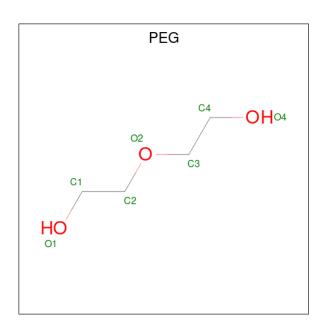
• Molecule 3 is (2R)-2,3-dihydroxypropyl (9Z)-octadec-9-enoate (three-letter code: OLC) (formula: $C_{21}H_{40}O_4$).



Mol	Chain	Residues	Atoms		ZeroOcc	AltConf	
3	A	1	Total 25	C 21	O 4	0	0

• Molecule 4 is DI(HYDROXYETHYL)ETHER (three-letter code: PEG) (formula: $C_4H_{10}O_3$).





Mol	Chain	Residues	Atoms			ZeroOcc	AltConf
4	A	1	Total 7	C 4	O 3	0	0

• Molecule 5 is UNKNOWN ATOM OR ION (three-letter code: UNX) (formula: X).

Mol	Chain	Residues	Atoms	ZeroOcc	AltConf
5	A	5	Total X 5 5	0	0

• Molecule 6 is water.

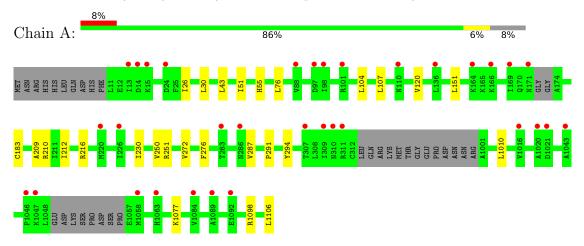
Mol	Chain	Residues	Atoms	ZeroOcc	AltConf
6	A	3	Total O 3 3	0	0



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 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: Hydroxycarboxylic acid receptor 2, Soluble cytochrome b562





4 Data and refinement statistics (i)

Property	Value	Source
Space group	P 21 2 21	Depositor
Cell constants	80.88Å 81.99Å 85.78Å	Depositor
a, b, c, α , β , γ	90.00° 90.00° 90.00°	Depositor
Resolution (Å)	30.00 - 2.70	Depositor
resolution (A)	29.64 - 2.70	EDS
% Data completeness	98.7 (30.00-2.70)	Depositor
(in resolution range)	98.7 (29.64-2.70)	EDS
R_{merge}	(Not available)	Depositor
R_{sym}	(Not available)	Depositor
$< I/\sigma(I) > 1$	1.28 (at 2.68Å)	Xtriage
Refinement program	BUSTER	Depositor
R, R_{free}	0.228 , 0.248	Depositor
it, it free	0.255 , 0.283	DCC
R_{free} test set	760 reflections (4.75%)	wwPDB-VP
Wilson B-factor (Å ²)	62.6	Xtriage
Anisotropy	0.614	Xtriage
Bulk solvent $k_{sol}(e/Å^3)$, $B_{sol}(Å^2)$	0.30,69.4	EDS
L-test for twinning ²	$< L > = 0.48, < L^2> = 0.31$	Xtriage
Estimated twinning fraction	0.008 for k,h,-l	Xtriage
F_o, F_c correlation	0.93	EDS
Total number of atoms	3196	wwPDB-VP
Average B, all atoms (Å ²)	87.0	wwPDB-VP

Xtriage's analysis on translational NCS is as follows: The largest off-origin peak in the Patterson function is 5.85% of the height of the origin peak. No significant pseudotranslation is detected.

²Theoretical values of <|L|>, $<L^2>$ for acentric reflections are 0.5, 0.333 respectively for untwinned datasets, and 0.375, 0.2 for perfectly twinned datasets.



¹Intensities estimated from amplitudes.

5 Model quality (i)

5.1 Standard geometry (i)

Bond lengths and bond angles in the following residue types are not validated in this section: PEG, OLC, UNX, OLA

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).

Ma	.1	Chain	Bond	lengths	Bond	angles
IVIC)1	Chain	RMSZ	# Z > 5	RMSZ	# Z > 5
1		A	0.43	0/3176	0.56	0/4326

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	3100	0	3027	9	0
2	A	56	0	80	1	0
3	A	25	0	40	0	0
4	A	7	0	10	0	0
5	A	5	0	0	0	0
6	A	3	0	0	0	0
All	All	3196	0	3157	9	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 1.

All (9) close contacts within the same asymmetric unit are listed below, sorted by their clash magnitude.



Atom-1	Atom-2	Interatomic	Clash
7100111 1	7100111 2	${f distance}({f A})$	overlap(A)
1:A:120:VAL:HG21	1:A:151:LEU:HD13	1.92	0.52
1:A:104:LEU:HA	1:A:107:LEU:HD12	1.95	0.49
1:A:76:LEU:HD23	1:A:287:VAL:HG13	1.95	0.48
1:A:51:ILE:HA	1:A:55:HIS:HB3	1.96	0.47
1:A:209:ALA:HB2	2:A:1205:OLA:H32	1.97	0.46
1:A:291:PRO:HA	1:A:294:TYR:CD2	2.52	0.45
1:A:26:ILE:O	1:A:30:LEU:HB2	2.17	0.43
1:A:250:VAL:HG21	1:A:276:PHE:HA	2.01	0.43
1:A:212:ILE:HG12	1:A:230:ILE:HG23	2.03	0.40

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 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	388/431 (90%)	379 (98%)	9 (2%)	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 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	Analysed Rotameric		Percentiles	
1	A	321/384 (84%)	311 (97%)	10 (3%)	40 69	

All (10) residues with a non-rotameric sidechain are listed below:



Mol	Chain	Res	Type
1	A	43	LEU
1	A	183	CYS
1	A	210	ARG
1	A	216	ARG
1	A	251	ARG
1	A	272	VAL
1	A	1010	LEU
1	A	1077	LYS
1	A	1098	ARG
1	A	1106	LEU

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

Mol	Chain	Res	Type
1	A	1103	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)

Of 12 ligands modelled in this entry, 5 are unknown - leaving 7 for Mogul analysis.

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	Trino	Chain	Res	Link	Вс	ond leng	ths	В	ond ang	les
MIOI	Type	Chain	nes	Lilik	Counts	RMSZ	# Z > 2	Counts	RMSZ	# Z > 2
2	OLA	A	1201	-	17,17,19	0.52	0	17,17,19	0.90	1 (5%)
2	OLA	A	1204	-	7,7,19	0.27	0	6,6,19	0.47	0
3	OLC	A	1206	-	24,24,24	0.48	0	25,25,25	0.84	1 (4%)
2	OLA	A	1205	-	15,15,19	0.57	0	15,15,19	0.86	1 (6%)
2	OLA	A	1202	-	6,6,19	0.28	0	5,5,19	0.45	0
2	OLA	A	1203	-	6,6,19	0.29	0	5,5,19	0.42	0
4	PEG	A	1207	_	6,6,6	0.46	0	5,5,5	0.31	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
2	OLA	A	1201	-	-	9/15/15/17	_
2	OLA	A	1204	-	-	4/5/5/17	-
3	OLC	A	1206	-	-	12/24/24/24	-
2	OLA	A	1205	-	-	7/13/13/17	_
2	OLA	A	1202	-	-	0/4/4/17	-
2	OLA	A	1203	-	-	2/4/4/17	_
4	PEG	A	1207	-	-	4/4/4/4	-

There are no bond length outliers.

All (3) bond angle outliers are listed below:

Mol	Chain	Res	Type	Atoms	\mathbf{Z}	$Observed(^o)$	$\operatorname{Ideal}({}^{o})$
3	A	1206	OLC	O20-C1-C2	2.46	119.62	111.91
2	A	1201	OLA	O2-C1-C2	2.16	120.97	114.03
2	A	1205	OLA	O2-C1-C2	2.04	120.58	114.03

There are no chirality outliers.

All (38) torsion outliers are listed below:

Mol	Chain	Res	Type	Atoms
3	A	1206	OLC	C21-C22-C24-O25
3	A	1206	OLC	O20-C21-C22-C24
3	A	1206	OLC	O20-C21-C22-O23
2	A	1205	OLA	C1-C2-C3-C4
2	A	1201	OLA	C1-C2-C3-C4

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Mol	Chain	Res	Type	Atoms
2	A	1204	OLA	C4-C5-C6-C7
2	A	1205	OLA	C5-C6-C7-C8
2	A	1201	OLA	C5-C6-C7-C8
2	A	1201	OLA	C11-C12-C13-C14
2	A	1204	OLA	C2-C3-C4-C5
2	A	1205	OLA	C3-C4-C5-C6
3	A	1206	OLC	C2-C3-C4-C5
2	A	1201	OLA	C2-C3-C4-C5
3	A	1206	OLC	O23-C22-C24-O25
3	A	1206	OLC	C14-C15-C16-C17
3	A	1206	OLC	C4-C5-C6-C7
3	A	1206	OLC	C3-C4-C5-C6
4	A	1207	PEG	O2-C3-C4-O4
4	A	1207	PEG	O1-C1-C2-O2
2	A	1205	OLA	C11-C12-C13-C14
2	A	1203	OLA	C14-C15-C16-C17
2	A	1203	OLA	C13-C14-C15-C16
2	A	1204	OLA	C1-C2-C3-C4
2	A	1205	OLA	C2-C3-C4-C5
4	A	1207	PEG	C1-C2-O2-C3
4	A	1207	PEG	C4-C3-O2-C2
3	A	1206	OLC	C11-C12-C13-C14
3	A	1206	OLC	C6-C7-C8-C9
2	A	1204	OLA	C3-C4-C5-C6
2	A	1201	OLA	O2-C1-C2-C3
2	A	1201	OLA	O1-C1-C2-C3
2	A	1201	OLA	C9-C10-C11-C12
3	A	1206	OLC	C7-C8-C9-C10
2	A	1201	OLA	C4-C5-C6-C7
3	A	1206	OLC	C1-C2-C3-C4
2	A	1205	OLA	O2-C1-C2-C3
2	A	1201	OLA	C7-C8-C9-C10
2	A	1205	OLA	O1-C1-C2-C3

There are no ring outliers.

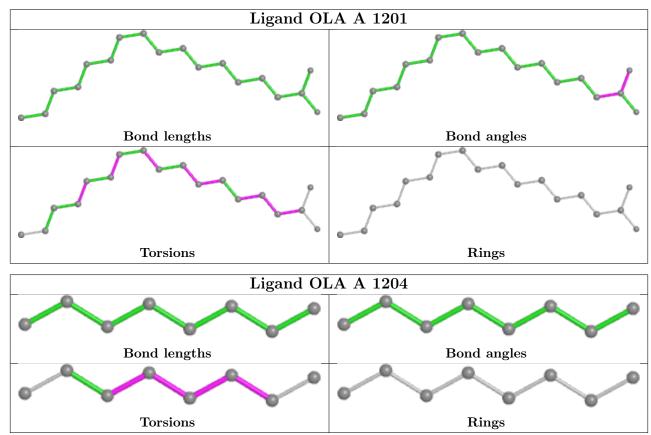
1 monomer is involved in 1 short contact:

Mol	Chain	Res	Type	Clashes	Symm-Clashes
2	A	1205	OLA	1	0

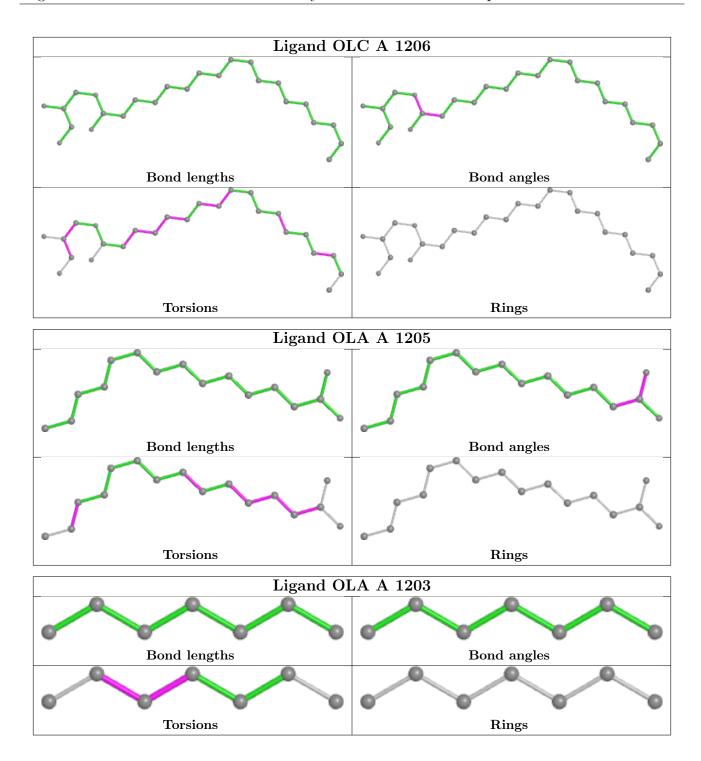
The following is a two-dimensional graphical depiction of Mogul quality analysis of bond lengths, bond angles, torsion angles, and ring geometry for all instances of the Ligand of Interest. In



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.







5.7 Other polymers (i)

There are no such residues in this entry.

5.8 Polymer linkage issues (i)

The following chains have linkage breaks:



Mol	Chain	Number of breaks
1	A	1

All chain breaks are listed below:

Model	Chain	Residue-1	Atom-1	Residue-2	Atom-2	Distance (Å)
1	A	219:GLN	С	220:MET	N	11.13



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, 95^{th} 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>	# RSRZ > 2	$OWAB(Å^2)$	Q<0.9
1	A	398/431 (92%)	0.60	33 (8%) 11 9	51, 86, 135, 182	0

All (33) RSRZ outliers are listed below:

Mol	Chain	Res	Type	RSRZ
1	A	14	ASP	6.4
1	A	1020	ALA	4.9
1	A	1089	ALA	4.5
1	A	13	ILE	4.5
1	A	97	ASP	4.3
1	A	1058	MET	4.3
1	A	164	LYS	4.2
1	A	310	ASN	3.9
1	A	169	ILE	3.8
1	A	1084	VAL	3.4
1	A	1043	ALA	3.4
1	A	88	VAL	3.3
1	A	311	ARG	3.2
1	A	98	ILE	3.2
1	A	24	ASP	3.1
1	A	1063	HIS	3.0
1	A	226	ILE	2.7
1	A	1046	PRO	2.7
1	A	15	LYS	2.6
1	A	166	LYS	2.5
1	A	1021	ASP	2.5
1	A	136	LEU	2.5
1	A	110	ASN	2.4
1	A	1016	VAL	2.4
1	A	307	THR	2.3
1	A	101	ARG	2.3
1	A	1092	GLU	2.2

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Mol	Chain	Res	Type	RSRZ
1	A	1047	LYS	2.2
1	A	309	ILE	2.2
1	A	171	ASN	2.2
1	A	220	MET	2.2
1	A	283	THR	2.1
1	A	286	ASN	2.1

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 monosaccharides in this entry.

6.4 Ligands (i)

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. The B-factors column lists the minimum, median, 95^{th} 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.

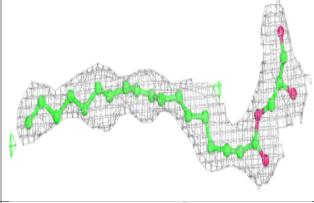
Mol	Type	Chain	Res	Atoms	RSCC	RSR	$oxed{ \mathbf{B\text{-}factors}(\mathbf{\mathring{A}}^2) }$	Q<0.9
5	UNX	A	1212	1/1	0.59	0.74	48,48,48,48	0
5	UNX	A	1211	1/1	0.73	0.51	51,51,51,51	0
3	OLC	A	1206	25/25	0.74	0.24	80,86,95,95	0
2	OLA	A	1202	7/20	0.76	0.32	72,73,73,73	0
2	OLA	A	1205	16/20	0.77	0.26	86,89,90,91	0
2	OLA	A	1203	7/20	0.78	0.23	68,69,69,70	0
4	PEG	A	1207	7/7	0.82	0.34	95,96,97,97	0
5	UNX	A	1209	1/1	0.82	1.15	54,54,54,54	0
2	OLA	A	1201	18/20	0.83	0.22	65,72,80,80	0
2	OLA	A	1204	8/20	0.83	0.32	57,59,61,61	0
5	UNX	A	1208	1/1	0.93	2.06	62,62,62,62	0
5	UNX	A	1210	1/1	0.93	1.31	59,59,59,59	0

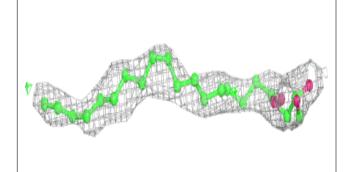
The following is a graphical depiction of the model fit to experimental electron density of all instances of the Ligand of Interest. In addition, ligands with molecular weight > 250 and outliers as shown on the geometry validation Tables will also be included. Each fit is shown from different orientation to approximate a three-dimensional view.

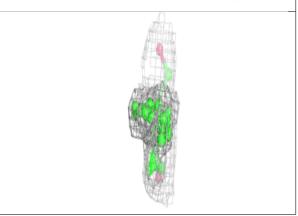


Electron density around OLC A 1206:

 $2 {\rm mF}_o\text{-}{\rm DF}_c$ (at 0.7 rmsd) in gray ${\rm mF}_o\text{-}{\rm DF}_c$ (at 3 rmsd) in purple (negative) and green (positive)

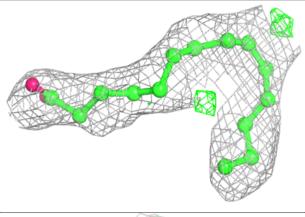


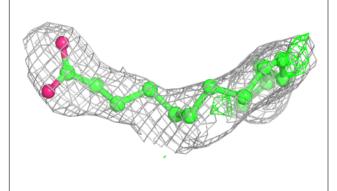


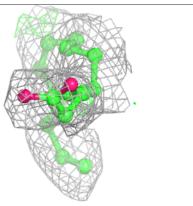


Electron density around OLA A 1205:

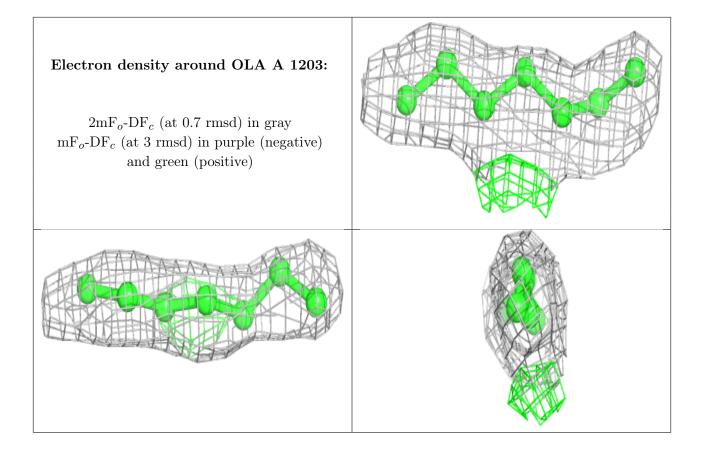
 $2 \mathrm{mF}_o\text{-}\mathrm{DF}_c$ (at 0.7 rmsd) in gray $\mathrm{mF}_o\text{-}\mathrm{DF}_c$ (at 3 rmsd) in purple (negative) and green (positive)



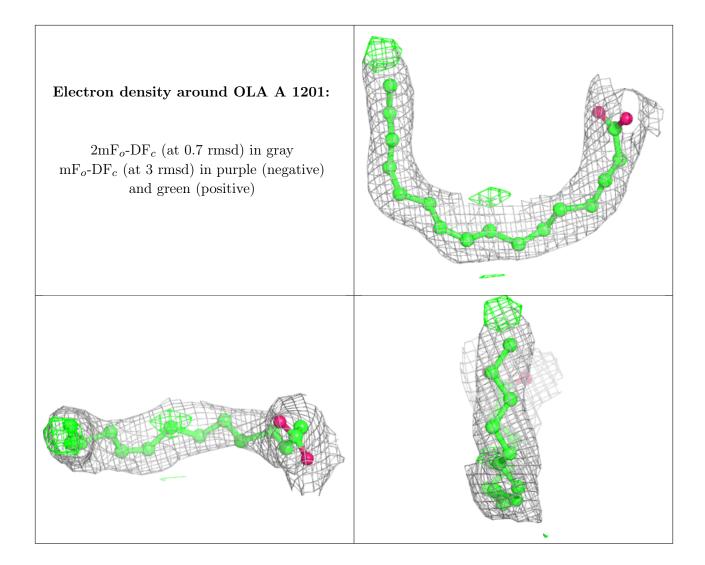




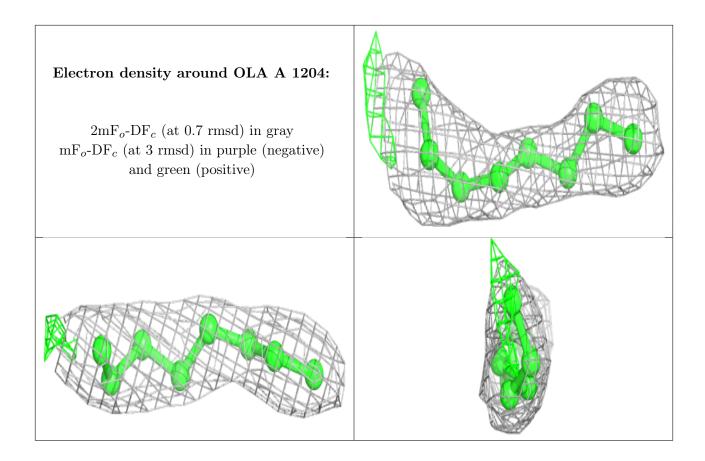












6.5 Other polymers (i)

There are no such residues in this entry.

