wwPDB EM Validation Summary Report (i)

May 2, 2024 – 06:08 PM JST

PDB ID	:	8JT6
EMDB ID	:	EMD-36634
Title	:	5-HT1A-Gi in complex with compound (R)-IHCH-7179
Authors	:	Chen, Z.; Xu, P.; Huang, S.; Xu, H.E.; Wang, S.
Deposited on	:	2023-06-21
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 (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:

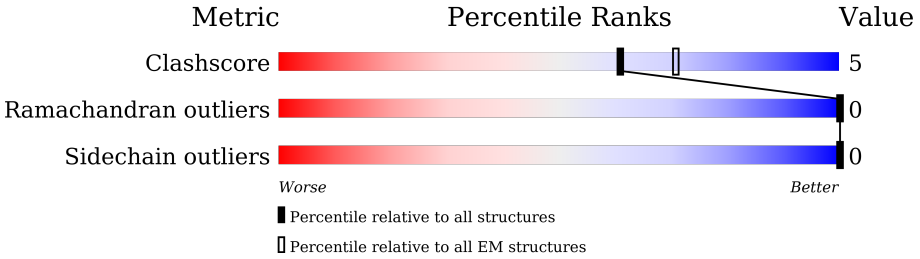
EMDB validation analysis	:	FAILED
Mogul	:	1.8.5 (274361), CSD as541be (2020)
MolProbity	:	4.02b-467
buster-report	:	1.1.7 (2018)
Percentile statistics	:	20191225.v01 (using entries in the PDB archive December 25th 2019)
MapQ	:	FAILED
Ideal geometry (proteins)	:	Engh & Huber (2001)
Ideal geometry (DNA, RNA)	:	Parkinson et al. (1996)
Validation Pipeline (wwPDB-VP)	:	2.36.2

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	354	58% • 38%
2	B	345	88% 7% 5%
3	E	269	82% • 14%
4	G	67	66% • 33%
5	R	544	46% 5% 49%

2 Entry composition (i)

There are 9 unique types of molecules in this entry. The entry contains 17718 atoms, of which 8889 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(i) subunit alpha-1.

Mol	Chain	Residues	Atoms						AltConf	Trace
			Total	C	H	N	O	S		
1	A	219	3537	1126	1771	295	333	12	0	0

There are 4 discrepancies between the modelled and reference sequences:

Chain	Residue	Modelled	Actual	Comment	Reference
A	47	ASN	SER	conflict	UNP P63096
A	203	ALA	GLY	conflict	UNP P63096
A	245	ALA	GLU	conflict	UNP P63096
A	326	SER	ALA	conflict	UNP P63096

- 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	H	N	O	S		
2	B	328	4945	1554	2429	452	489	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 ScFv16.

Mol	Chain	Residues	Atoms						AltConf	Trace
			Total	C	H	N	O	S		
3	E	230	3471	1120	1703	293	345	10	0	0

- 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
			Total	C	H	N	O	S		
4	G	45	698	218	351	60	66	3	0	0

- Molecule 5 is a protein called Soluble cytochrome b562,5-hydroxytryptamine receptor 1A.

Mol	Chain	Residues	Atoms						AltConf	Trace
			Total	C	H	N	O	S		
5	R	278	4497	1453	2300	362	365	17	0	0

There are 42 discrepancies between the modelled and reference sequences:

Chain	Residue	Modelled	Actual	Comment	Reference
R	-121	MET	-	initiating methionine	UNP P0ABE7
R	-120	ASP	-	expression tag	UNP P0ABE7
R	-119	TYR	-	expression tag	UNP P0ABE7
R	-118	LYS	-	expression tag	UNP P0ABE7
R	-117	ASP	-	expression tag	UNP P0ABE7
R	-116	ASP	-	expression tag	UNP P0ABE7
R	-115	ASP	-	expression tag	UNP P0ABE7
R	-114	ASP	-	expression tag	UNP P0ABE7
R	-113	ALA	-	expression tag	UNP P0ABE7
R	-112	LYS	-	expression tag	UNP P0ABE7
R	-111	LEU	-	expression tag	UNP P0ABE7
R	-110	GLN	-	expression tag	UNP P0ABE7
R	-109	THR	-	expression tag	UNP P0ABE7
R	-108	MET	-	expression tag	UNP P0ABE7
R	-107	HIS	-	expression tag	UNP P0ABE7
R	-106	HIS	-	expression tag	UNP P0ABE7
R	-105	HIS	-	expression tag	UNP P0ABE7
R	-104	HIS	-	expression tag	UNP P0ABE7
R	-103	HIS	-	expression tag	UNP P0ABE7
R	-102	HIS	-	expression tag	UNP P0ABE7
R	-101	HIS	-	expression tag	UNP P0ABE7
R	-100	HIS	-	expression tag	UNP P0ABE7
R	-99	HIS	-	expression tag	UNP P0ABE7
R	-98	HIS	-	expression tag	UNP P0ABE7
R	-97	HIS	-	expression tag	UNP P0ABE7
R	-96	HIS	-	expression tag	UNP P0ABE7
R	-95	HIS	-	expression tag	UNP P0ABE7
R	-94	HIS	-	expression tag	UNP P0ABE7

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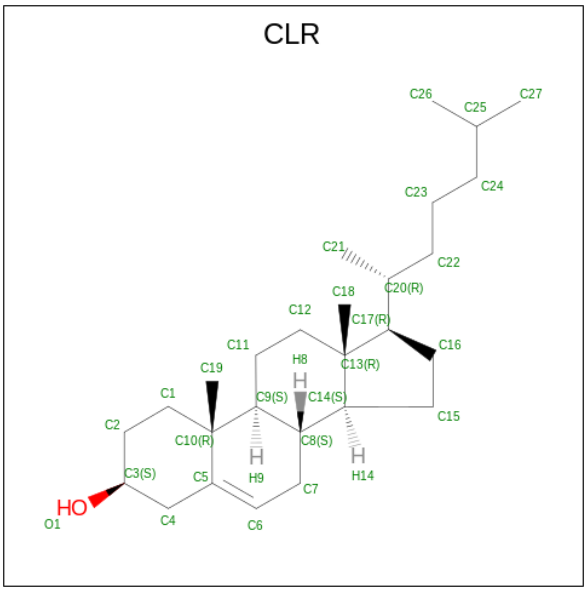
Chain	Residue	Modelled	Actual	Comment	Reference
R	-93	HIS	-	expression tag	UNP P0ABE7
R	-86	TRP	MET	conflict	UNP P0ABE7
R	9	ILE	HIS	conflict	UNP P0ABE7
R	13	LEU	-	linker	UNP P0ABE7
R	14	ALA	-	linker	UNP P0ABE7
R	15	SER	-	linker	UNP P0ABE7
R	16	GLU	-	linker	UNP P0ABE7
R	17	ASN	-	linker	UNP P0ABE7
R	18	LEU	-	linker	UNP P0ABE7
R	19	TYR	-	linker	UNP P0ABE7
R	20	PHE	-	linker	UNP P0ABE7
R	21	GLN	-	linker	UNP P0ABE7
R	24	THR	ASN	conflict	UNP P08908
R	125	TRP	LEU	conflict	UNP P08908

- Molecule 6 is CHOLESTEROL (three-letter code: CLR) (formula: $C_{27}H_{46}O$).



Mol	Chain	Residues	Atoms				AltConf
			Total	C	H	O	
6	R	1	74	27	46	1	0
			Total	C	H	O	
6	R	1	74	27	46	1	0
			Total	C	H	O	
6	R	1	74	27	46	1	0
			Total	C	H	O	
6	R	1	74	27	46	1	0

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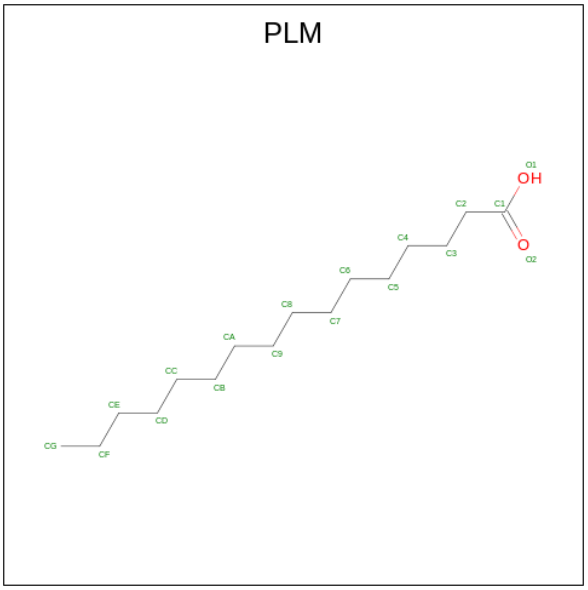
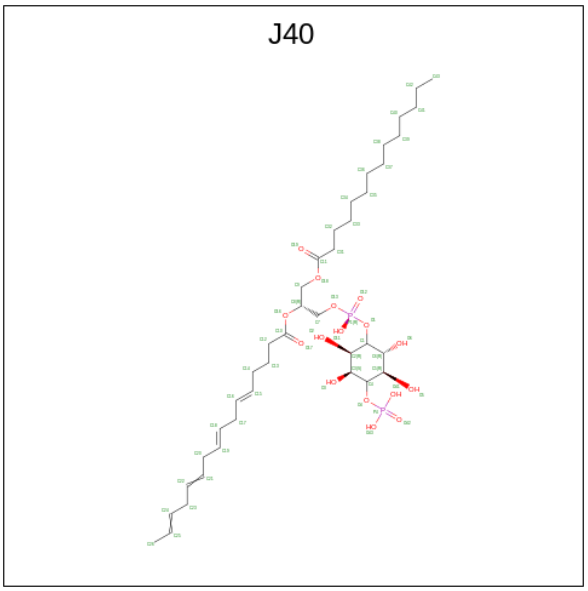
Mol	Chain	Residues	Atoms				AltConf
			Total	C	H	O	
6	R	1	74	27	46	1	0

- Molecule 7 is PALMITIC ACID (three-letter code: PLM) (formula: $C_{16}H_{32}O_2$).



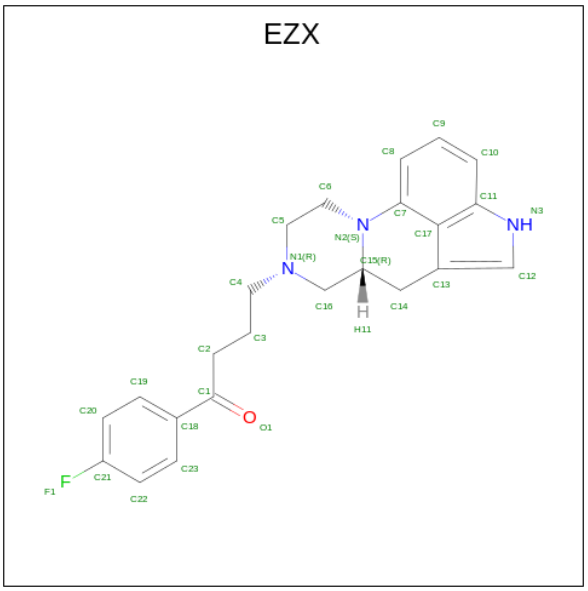
Mol	Chain	Residues	Atoms				AltConf
			Total	C	H	O	
7	R	1	49	16	31	2	0
			Total	C	H	O	
7	R	1	49	16	31	2	0

- Molecule 8 is [(2R)-1-[oxidanyl-[(2R,3R,5S,6R)-2,3,5,6-tetrakis(oxidanyl)-4-phosphonooxy-cyclohexyl]oxy-phosphoryl]oxy-3-tetradecanoyloxy-propan-2-yl] (5E,8E)-hexadeca-5,8,11,14-tetraenoate (three-letter code: J40) (formula: $C_{39}H_{68}O_{16}P_2$).



Mol	Chain	Residues	Atoms					AltConf
			Total	C	H	O	P	
8	R	1	50	13	19	16	2	0

- Molecule 9 is 1-(4-fluorophenyl)-4-[(7R)-2,5,11-triazatetracyclo[7.6.1.0^2,7.0^12,16]hexadeca-1(15),9,12(16),13-tetraen-5-yl]butan-1-one (three-letter code: EZX) (formula: $C_{23}H_{24}FN_3O$) (labeled as "Ligand of Interest" by depositor).



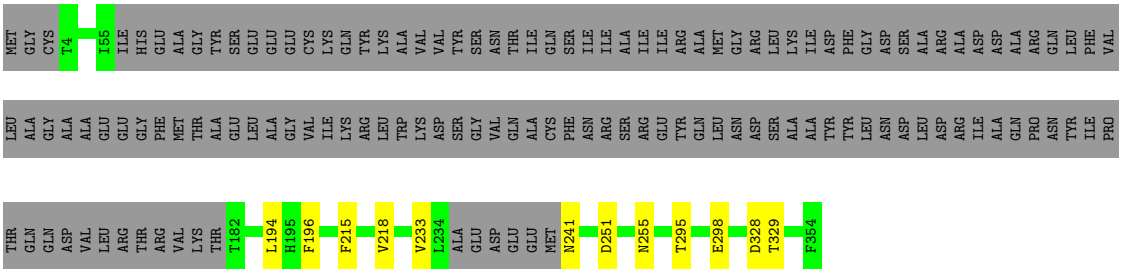
Mol	Chain	Residues	Atoms						AltConf
			Total	C	F	H	N	O	
9	R	1	52	23	1	24	3	1	0

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.

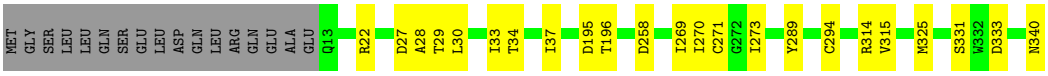
• Molecule 1: Guanine nucleotide-binding protein G(i) subunit alpha-1

Chain A: 58% • 38%



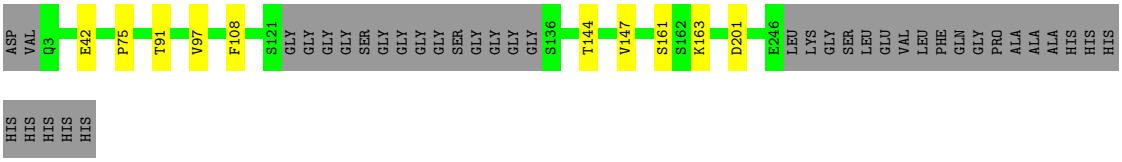
• Molecule 2: Guanine nucleotide-binding protein G(I)/G(S)/G(T) subunit beta-1

Chain B: 88% 7% 5%



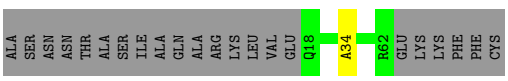
• Molecule 3: ScFv16

Chain E: 82% • 14%



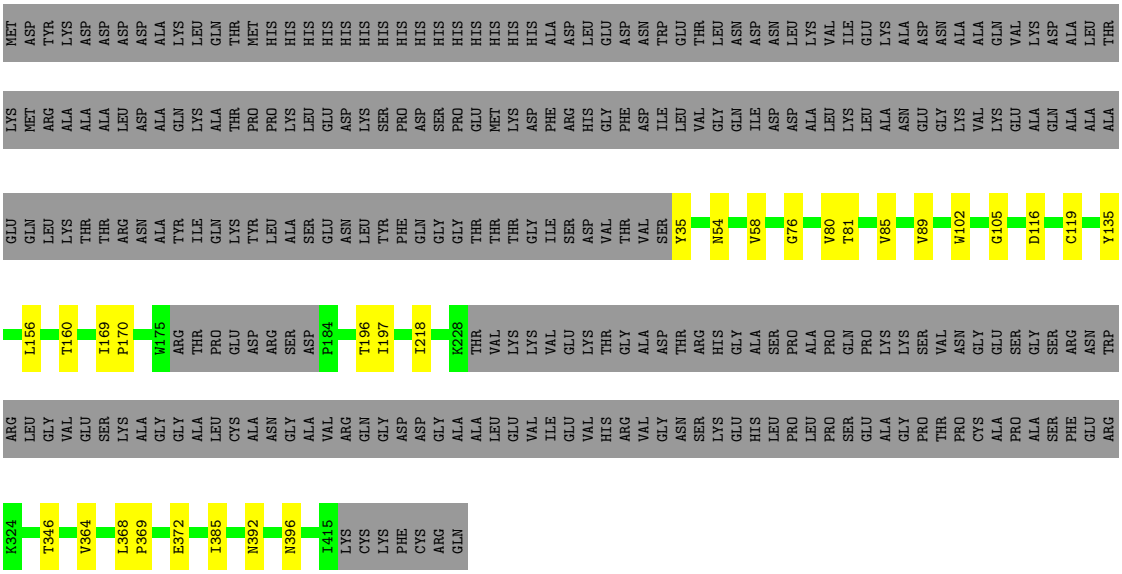
• Molecule 4: Guanine nucleotide-binding protein G(I)/G(S)/G(O) subunit gamma-2

Chain G: 66% • 33%



• Molecule 5: Soluble cytochrome b562,5-hydroxytryptamine receptor 1A

Chain R: 46% 5% 49%


MET ASP TYR LYS ASP ASP ASP ASP ALA LYS LEU GLN THR MET HIS HIS HIS HIS HIS HIS HIS HIS HIS HIS HIS HIS HIS HIS ALA ASP LEU GLU ASP ASN TRP GLU THR LEU ASN ASP ASN LEU LYS VAL ILE GLU LYS ALA ASP ASN ALA ALA GLN VAL LYS ASP ALA LEU THR
LYS MET ARG ALA ALA ALA LEU ASP ALA GLN LYS ALA THR PRO PRO LYS LEU GLU ASP LYS SER PRO ASP SER PRO GLU MET LYS ASP PHE ARG HIS GLY PHE ASP ILE LEU VAL GLY GLN ILE ASP ASP ALA LEU LYS LEU ALA ASN GLU GLY LYS VAL LYS GLU ALA GLN ALA ALA ALA
GLU GLN LEU LYS THR THR ARG ASN ALA TYR ILE GLN LYS TYR LEU ALA SER GLU ASN LEU TYR PHE GLN GLY GLY THR THR THR GLY ILE SER ASP VAL THR VAL SER Y35 N54 V58 G76 V80 T81 V85 V89 W102 G105 D116 C119 Y135
L156 T160 I169 P170 W175 ARG THR PRO GLU ASP ARG SER ASP P184 T196 I197 I218 K228 THR VAL LYS LYS VAL GLU LYS THR GLY ALA ASP THR ARG HIS GLY ALA SER PRO ALA PRO GLN PRO LYS LYS SER VAL ASN GLY GLU SER GLY SER ARG ASN TRP
ARG LEU GLY VAL GLU SER LYS ALA GLY GLY ALA LEU CYS ALA ASN GLY ALA VAL ARG GLN GLY ASP ASP GLY ALA ALA LEU GLU VAL ILE GLU VAL HIS ARG VAL GLY ASN SER LYS GLU HIS LEU PRO LEU PRO SER GLU ALA GLY PRO THR PRO CYS ALA PRO ALA SER PHE GLU ARG
K324 T346 V364 L368 P369 E372 I385 N392 N396 I415 LYS CYS LYS PHE CYS ARG GLN

4 Experimental information (i)

Property	Value	Source
EM reconstruction method	SINGLE PARTICLE	Depositor
Imposed symmetry	POINT, Not provided	
Number of particles used	175082	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 ($e^-/Å^2$)	70	Depositor
Minimum defocus (nm)	1200	Depositor
Maximum defocus (nm)	2200	Depositor
Magnification	Not provided	
Image detector	GATAN K3 (6k x 4k)	Depositor

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: PLM, CLR, EZX, J40

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	0.24	0/1795	0.39	0/2407
2	B	0.23	0/2563	0.44	0/3475
3	E	0.26	0/1812	0.50	2/2456 (0.1%)
4	G	0.24	0/353	0.40	0/477
5	R	0.25	0/2248	0.41	1/3058 (0.0%)
All	All	0.25	0/8771	0.43	3/11873 (0.0%)

There are no bond length outliers.

All (3) bond angle outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(^{o})	Ideal(^{o})
3	E	75	PRO	CA-N-CD	-9.30	98.48	111.50
5	R	369	PRO	CA-N-CD	-5.34	104.03	111.50
3	E	75	PRO	N-CD-CG	-5.17	95.44	103.20

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	1766	1771	1768	6	0
2	B	2516	2429	2427	16	0
3	E	1768	1703	1701	6	0

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Mol	Chain	Non-H	H(model)	H(added)	Clashes	Symm-Clashes
4	G	347	351	350	2	0
5	R	2197	2300	2298	15	0
6	R	140	230	230	40	0
7	R	36	62	62	0	0
8	R	31	19	0	1	0
9	R	28	24	0	0	0
All	All	8829	8889	8836	82	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 82 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)
6:R:504:CLR:C11	6:R:504:CLR:C12	1.74	1.64
6:R:506:CLR:C12	6:R:506:CLR:C11	1.74	1.63
6:R:503:CLR:C9	6:R:503:CLR:C11	1.75	1.62
6:R:501:CLR:C9	6:R:501:CLR:C11	1.75	1.61
6:R:503:CLR:C11	6:R:503:CLR:C12	1.74	1.61

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	213/354 (60%)	212 (100%)	1 (0%)	0	100	100
2	B	326/345 (94%)	318 (98%)	8 (2%)	0	100	100
3	E	226/269 (84%)	223 (99%)	3 (1%)	0	100	100
4	G	43/67 (64%)	43 (100%)	0	0	100	100
5	R	272/544 (50%)	271 (100%)	1 (0%)	0	100	100

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Mol	Chain	Analysed	Favoured	Allowed	Outliers	Percentiles	
All	All	1080/1579 (68%)	1067 (99%)	13 (1%)	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	195/305 (64%)	195 (100%)	0	100	100
2	B	272/287 (95%)	272 (100%)	0	100	100
3	E	195/217 (90%)	195 (100%)	0	100	100
4	G	37/55 (67%)	37 (100%)	0	100	100
5	R	237/454 (52%)	237 (100%)	0	100	100
All	All	936/1318 (71%)	936 (100%)	0	100	100

There are no protein residues with a non-rotameric sidechain to report.

Sometimes sidechains can be flipped to improve hydrogen bonding and reduce clashes. All (2) such sidechains are listed below:

Mol	Chain	Res	Type
2	B	340	ASN
5	R	126	HIS

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)

9 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	CLR	R	502	-	31,31,31	4.14	15 (48%)	48,48,48	1.51	8 (16%)
9	EZX	R	509	-	30,32,32	4.75	17 (56%)	35,46,46	2.26	13 (37%)
8	J40	R	508	-	31,31,57	1.24	3 (9%)	43,46,72	1.53	8 (18%)
6	CLR	R	501	-	31,31,31	4.19	15 (48%)	48,48,48	1.53	8 (16%)
6	CLR	R	503	-	31,31,31	4.15	15 (48%)	48,48,48	1.49	10 (20%)
6	CLR	R	506	-	31,31,31	4.16	15 (48%)	48,48,48	1.47	6 (12%)
6	CLR	R	504	-	31,31,31	4.15	15 (48%)	48,48,48	1.54	10 (20%)
7	PLM	R	505	-	17,17,17	0.92	1 (5%)	17,17,17	0.74	2 (11%)
7	PLM	R	507	-	17,17,17	0.93	1 (5%)	17,17,17	0.74	1 (5%)

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	CLR	R	502	-	-	1/10/68/68	0/4/4/4
9	EZX	R	509	-	-	6/10/31/31	0/4/5/5
8	J40	R	508	-	-	12/25/49/77	0/1/1/1
6	CLR	R	501	-	-	4/10/68/68	0/4/4/4
6	CLR	R	503	-	-	0/10/68/68	0/4/4/4
6	CLR	R	506	-	-	1/10/68/68	0/4/4/4
6	CLR	R	504	-	-	0/10/68/68	0/4/4/4
7	PLM	R	505	-	-	2/15/15/15	-

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Mol	Type	Chain	Res	Link	Chirals	Torsions	Rings
7	PLM	R	507	-	-	2/15/15/15	-

The worst 5 of 97 bond length outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(Å)	Ideal(Å)
9	R	509	EZX	C7-C17	-18.93	1.12	1.43
6	R	502	CLR	C11-C9	13.20	1.75	1.53
6	R	504	CLR	C11-C9	13.19	1.75	1.53
6	R	501	CLR	C11-C9	13.18	1.75	1.53
6	R	506	CLR	C11-C9	13.18	1.75	1.53

The worst 5 of 66 bond angle outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(^{o})	Ideal(^{o})
6	R	504	CLR	C15-C14-C8	-5.54	109.96	119.08
9	R	509	EZX	C6-N2-C7	-5.52	108.33	118.64
9	R	509	EZX	C8-C7-N2	-5.39	119.31	123.40
6	R	506	CLR	C15-C14-C8	-5.23	110.47	119.08
6	R	503	CLR	C15-C14-C8	-5.14	110.62	119.08

There are no chirality outliers.

5 of 28 torsion outliers are listed below:

Mol	Chain	Res	Type	Atoms
8	R	508	J40	C7-O13-P1-O12
8	R	508	J40	C7-O13-P1-O11
8	R	508	J40	C7-O13-P1-O1
8	R	508	J40	C8-C7-O13-P1
8	R	508	J40	C2-C1-O1-P1

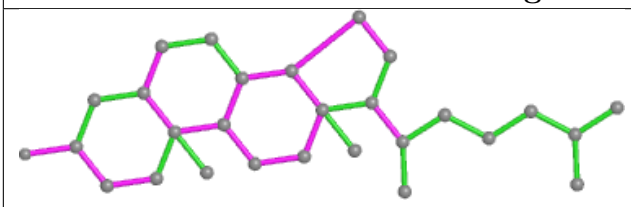
There are no ring outliers.

6 monomers are involved in 41 short contacts:

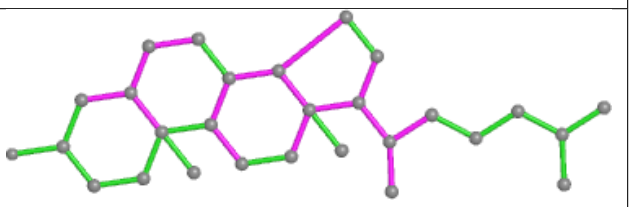
Mol	Chain	Res	Type	Clashes	Symm-Clashes
6	R	502	CLR	9	0
8	R	508	J40	1	0
6	R	501	CLR	7	0
6	R	503	CLR	8	0
6	R	506	CLR	8	0
6	R	504	CLR	8	0

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.

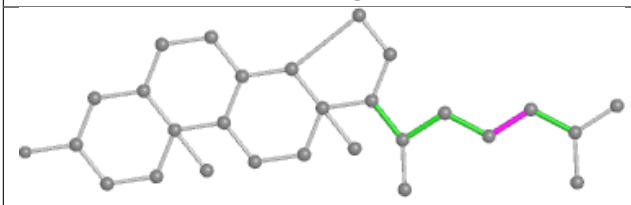
Ligand CLR R 502



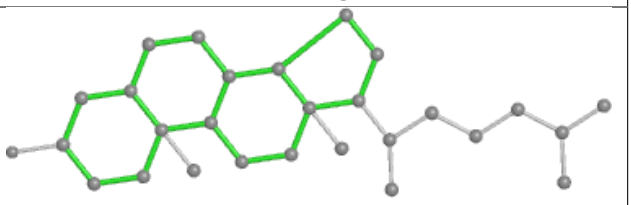
Bond lengths



Bond angles

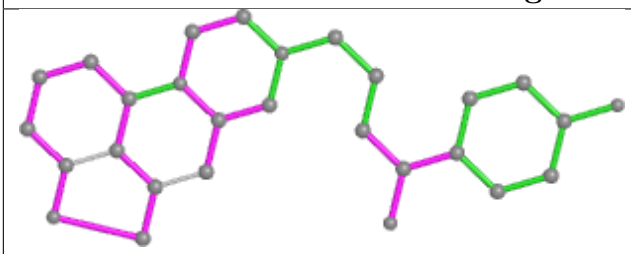


Torsions

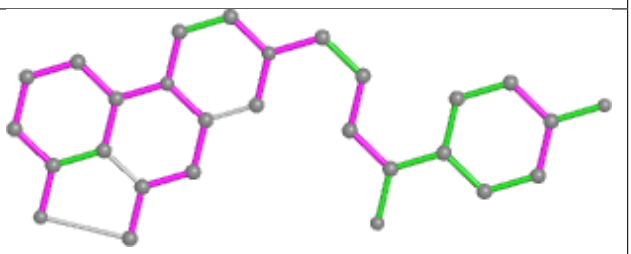


Rings

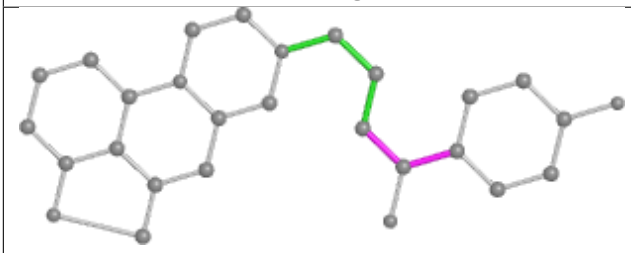
Ligand EZX R 509



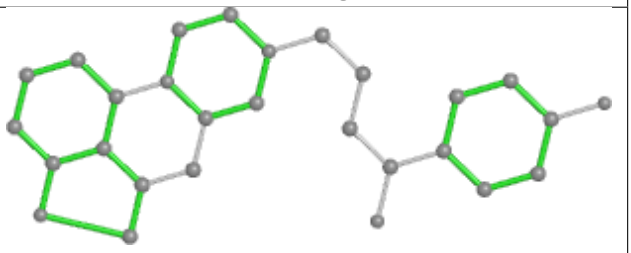
Bond lengths



Bond angles

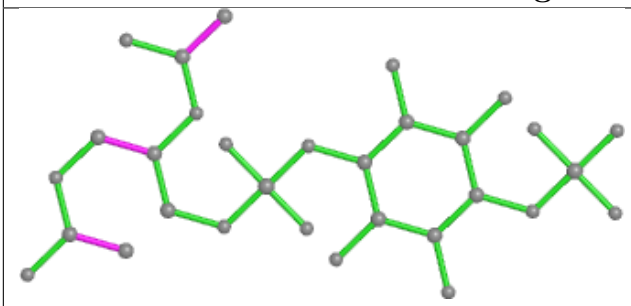


Torsions

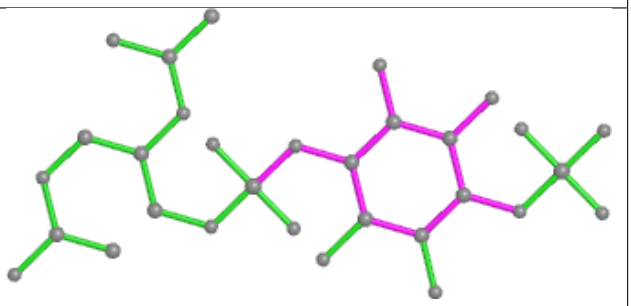


Rings

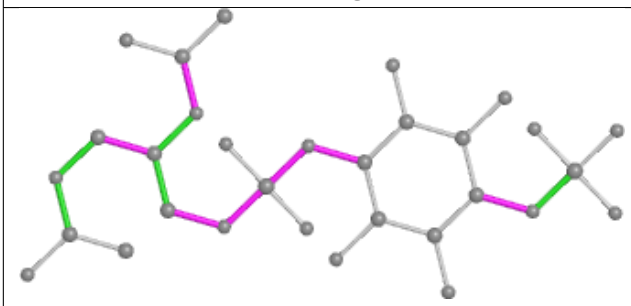
Ligand J40 R 508



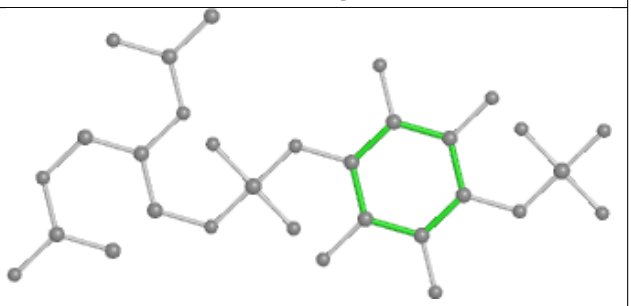
Bond lengths



Bond angles

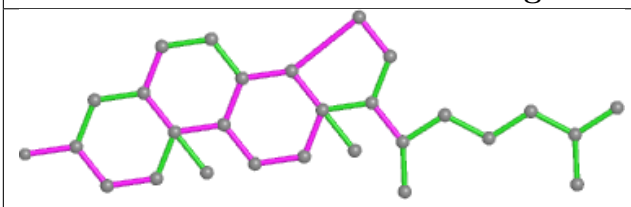


Torsions

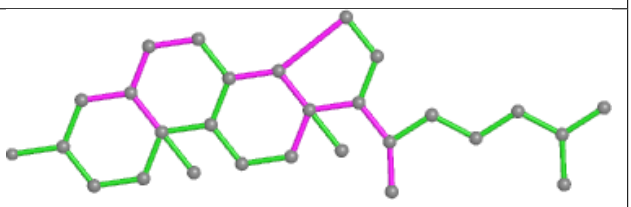


Rings

Ligand CLR R 501



Bond lengths



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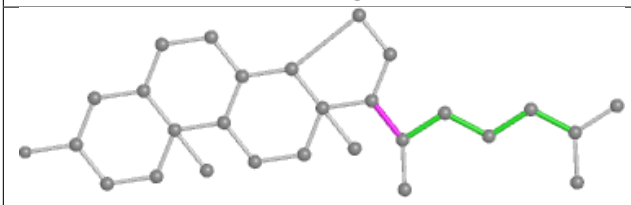

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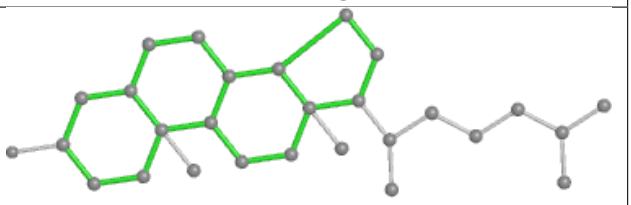
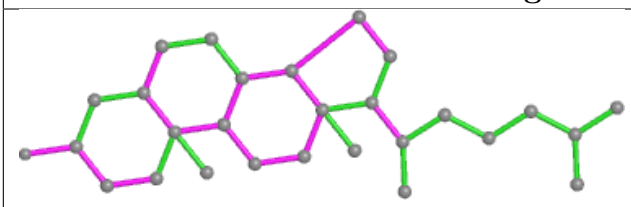


Rings

Ligand CLR R 503



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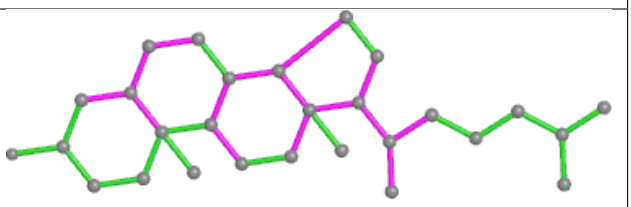

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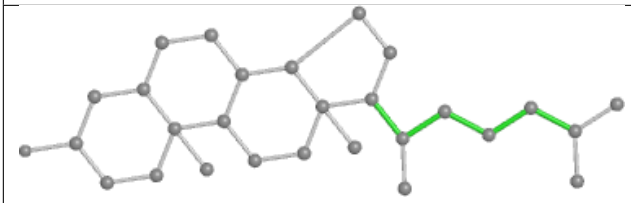

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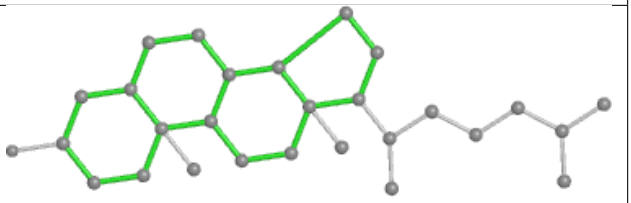

Rings

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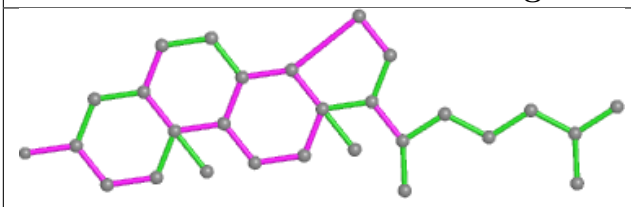

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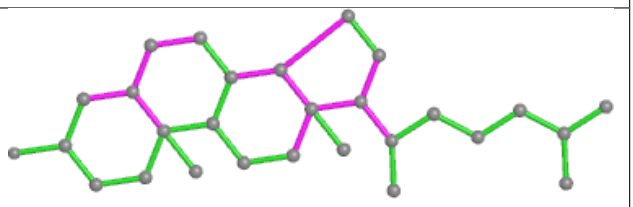

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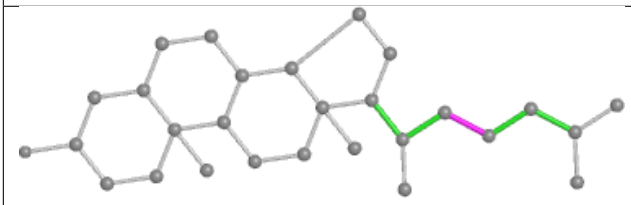

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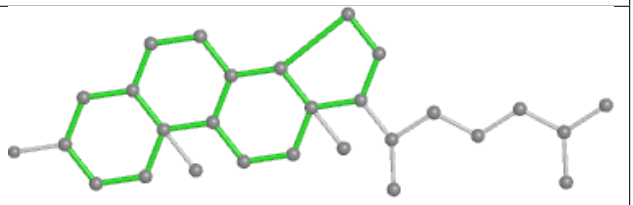
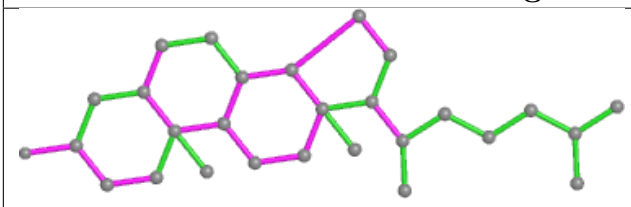


Rings

Ligand CLR R 504



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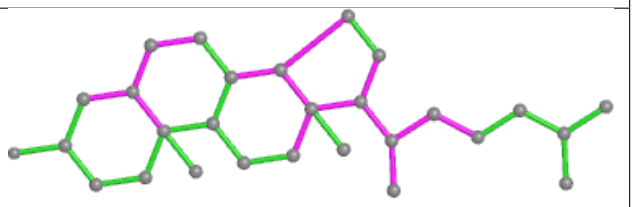
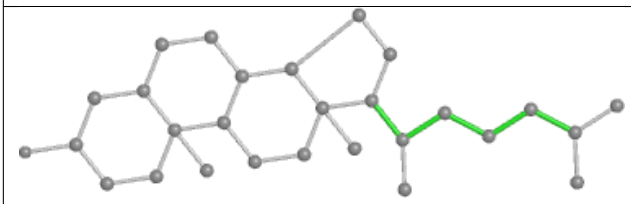


Bond angles



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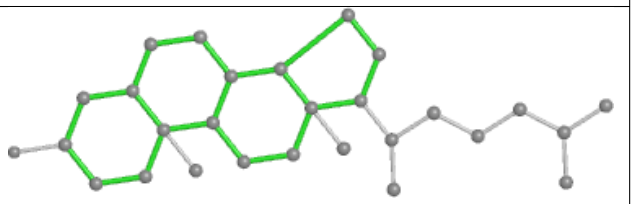
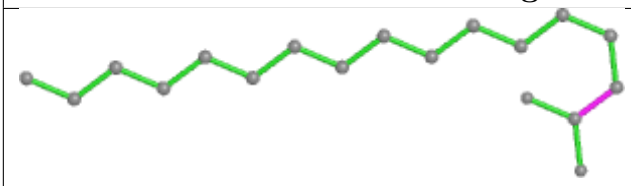
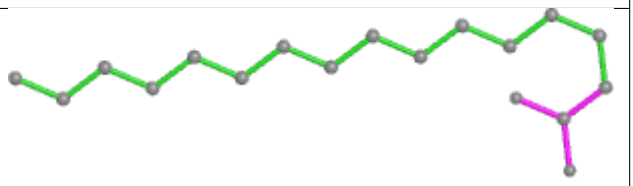


Rings

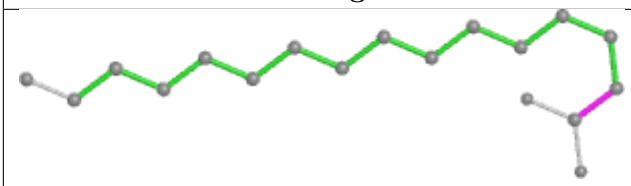
Ligand PLM R 505



Bond lengths

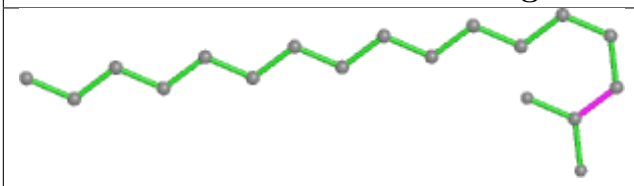
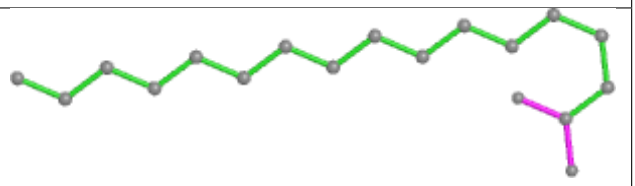
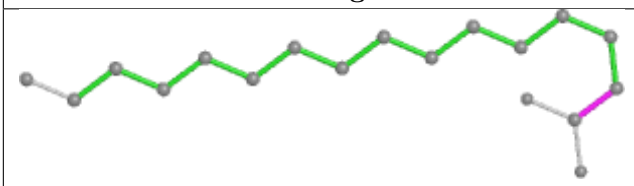
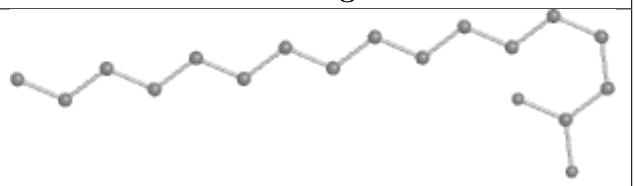


Bond angles



Torsions

Rings

Ligand PLM R 507	
 Bond lengths	 Bond angles
 Torsions	 Rings

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.