wwPDB EM Validation Summary Report

Oct 7, 2024 – 02:11 PM JST

PDB ID	:	8ISJ
EMDB ID	:	EMD-35692
Title	:	Pr conformer of Arabidopsis thaliana phytochrome A - AtphyA-Pr
Authors	:	Zhang, Y.; Ma, C.; Zhao, J.; Gao, N.; Wang, J.
Deposited on	:	2023-03-20
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:

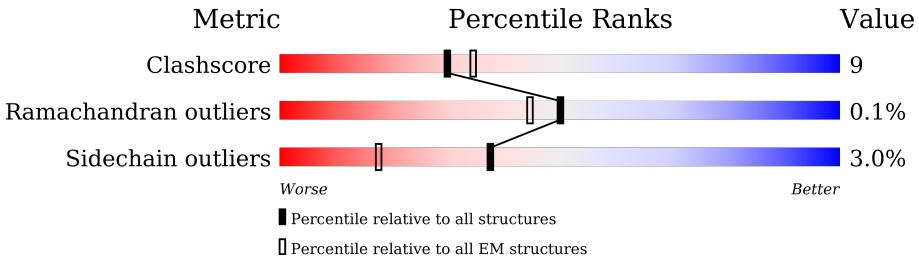
EMDB validation analysis	:	FAILED
Mogul	:	1.8.5 (274361), CSD as541be (2020)
MolProbity	:	4.02b-467
buster-report	:	1.1.7 (2018)
Percentile statistics	:	20231227.v01 (using entries in the PDB archive December 27th 2023)
MapQ	:	FAILED
Ideal geometry (proteins)	:	Engh & Huber (2001)
Ideal geometry (DNA, RNA)	:	Parkinson et al. (1996)
Validation Pipeline (wwPDB-VP)	:	2.39

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	210492	15764
Ramachandran outliers	207382	16835
Sidechain outliers	206894	16415

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	1120	62% 14% • 23%
1	B	1120	58% 18% • 23%

2 Entry composition

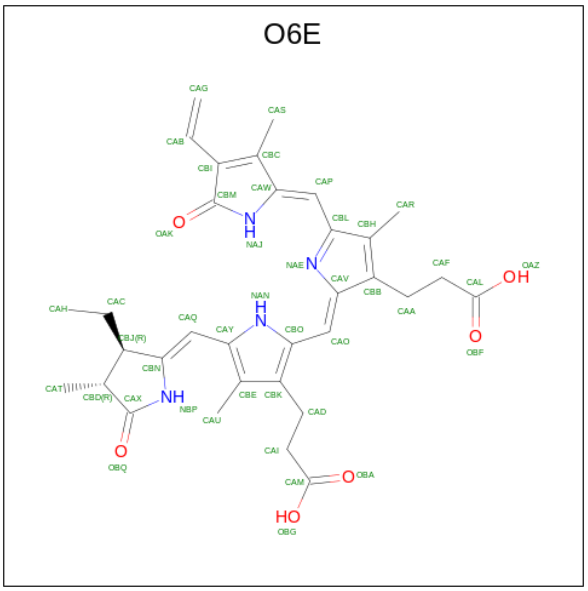
There are 2 unique types of molecules in this entry. The entry contains 13533 atoms, of which 0 are hydrogens and 0 are deuteriums.

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 Phytochrome A.

Mol	Chain	Residues	Atoms					AltConf	Trace
1	A	859	Total	C	N	O	S	0	0
			6730	4276	1156	1245	53		
1	B	857	Total	C	N	O	S	0	0
			6717	4269	1155	1240	53		

- Molecule 2 is 3-[5-[[(3 {R},4 {R})-3-ethyl-4-methyl-5-oxidanylidene-3,4-dihydropyrrol-2-yl]methyl]-2-[[5-[(4-ethyl-3-methyl-5-oxidanylidene-pyrrol-2-yl)methyl]-3-(3-hydroxy-3-oxopropyl)-4-methyl-1 {H}-pyrrol-2-yl]methyl]-4-methyl-1 {H}-pyrrol-3-yl]propanoic acid (three-letter code: O6E) (formula: $C_{33}H_{38}N_4O_6$) (labeled as "Ligand of Interest" by depositor).



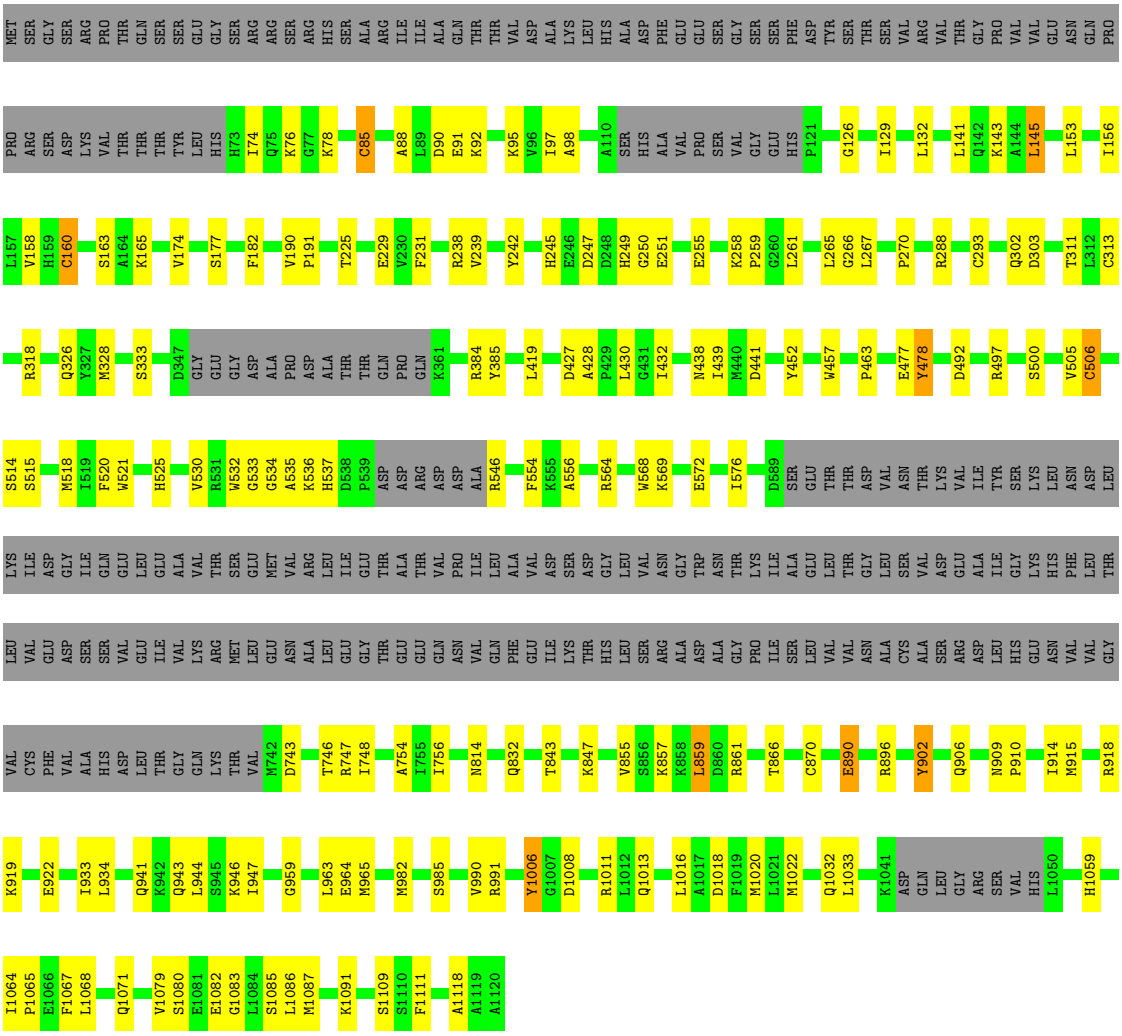
Mol	Chain	Residues	Atoms				AltConf
2	A	1	Total	C	N	O	0
			43	33	4	6	
2	B	1	Total	C	N	O	0
			43	33	4	6	

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.

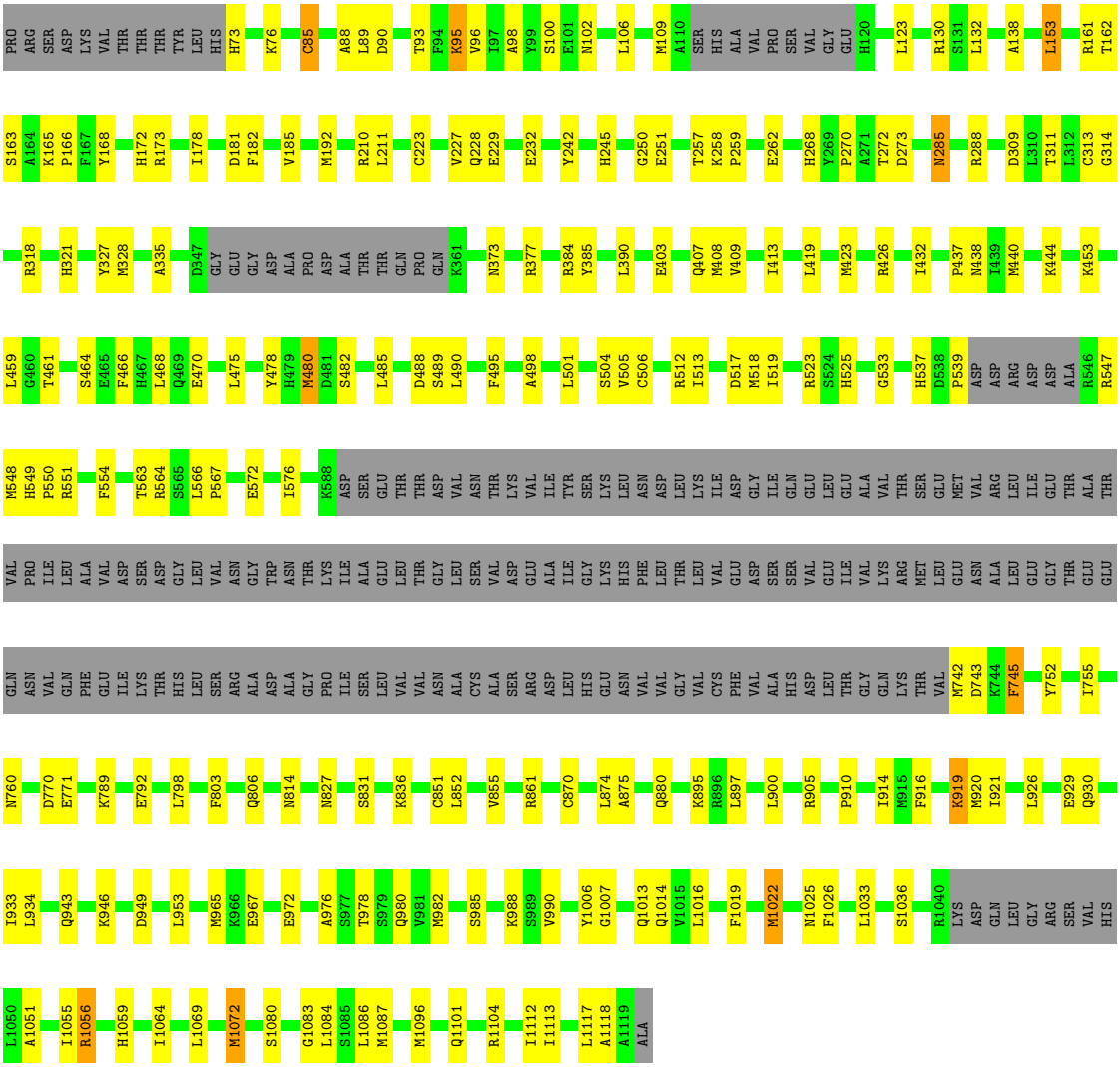
- Molecule 1: Phytochrome A

Chain A:



- Molecule 1: Phytochrome A

Chain B:


PRO ARG SER ASP LYS VAL THR THR TYR LEU HIS H73 K76 C85 A88 L89 D90 T93 F94 K95 V96 I97 A98 Y99 S100 E101 N102 L106 M109 A110 SER HIS ALA VAL PRO SER VAL GLY GLU H120 L123 R130 S131 L132 A138 L153 R161 T162
S163 A164 K165 P166 F167 Y168 H172 R173 I178 D181 F182 V185 M192 R210 L211 C223 V227 Q228 E229 E232 Y242 H245 G250 E251 T257 K258 P259 E262 H268 Y269 P270 A271 T272 D273 N285 R288 D309 L310 T311 L312 C313 G314
R318 H321 Y327 M328 A335 D347 GLY GLU GLY ASP ALA PRO ASP ALA THR THR GLN PRO GLN K361 N373 R377 R384 Y385 L390 E403 Q407 M408 V409 I413 L419 M423 R426 I432 P437 N438 I439 M440 K444 K453
L459 G460 T461 S464 E465 F466 H467 L468 Q469 E470 L475 Y478 H479 M480 D481 S482 L485 D488 S489 L490 F495 A498 L501 S504 V505 C506 R512 I513 D517 M518 I519 R523 S524 H525 G533 H537 D538 P539 ASP ASP ARG ASP ASP ALA R546 R547
M548 H549 P550 R551 F554 T563 R564 S565 L566 P567 E572 I576 K588 ASP SER GLU THR THR ASP VAL ASN THR LYS VAL ILE TYR SER LYS LEU ASN ASP LEU LYS ILE ASP GLY ILE GLN GLU LEU GLU ALA VAL THR SER GLU MET VAL ARG LEU ILE GLU THR ALA THR
VAL PRO ILE LEU ALA VAL ASP SER ASP GLY LEU VAL ASN GLY TRP ASN THR LYS ILE ALA GLU LEU THR GLY LEU SER VAL ASP GLU ALA ILE GLY LYS HIS PHE LEU THR LEU VAL GLU ASP SER SER VAL GLU ILE VAL LYS ARG MET LEU GLU ASN ALA LEU GLU GLY THR GLU GLU
GLN ASN VAL GLN PHE GLU ILE LYS THR HIS LEU SER ARG ALA ASP ALA GLY PRO ILE SER LEU VAL VAL ASN ALA CYS ALA SER ARG ASP LEU HIS GLU ASN VAL VAL GLY VAL CYS PHE VAL ALA HIS ASP LEU THR GLY GLN LYS THR VAL M742 D743 K744 F745 Y752 I755
N760 D770 E771 K789 E792 L798 F803 Q806 N814 N827 S831 K836 C851 L852 V855 R861 C870 L874 A875 Q880 K895 R896 L897 L900 R905 P910 I914 M915 F916 K919 M920 I921 L926 E929 Q930
I933 L934 Q943 K946 D949 L953 M965 K966 E967 E972 A976 S977 T978 S979 Q980 V981 M982 S985 K988 S989 V990 Y1006 G1007 Q1013 Q1014 V1015 L1016 F1019 M1022 N1025 F1026 L1033 S1036 R1040 LYS ASP GLN LEU GLY ARG SER VAL HIS
L1050 A1051 I1055 R1056 H1059 I1064 L1069 M1072 S1080 G1083 L1084 S1085 L1086 M1087 M1096 Q1101 R1104 I1112 I1113 L1117 A1118 A1119 ALA

4 Experimental information (i)

Property	Value	Source
EM reconstruction method	SINGLE PARTICLE	Depositor
Imposed symmetry	POINT, Not provided	
Number of particles used	774153	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 ($e^-/Å^2$)	50	Depositor
Minimum defocus (nm)	1500	Depositor
Maximum defocus (nm)	1800	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: O6E

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	0/6852	0.52	1/9245 (0.0%)
1	B	0.27	0/6840	0.53	0/9232
All	All	0.27	0/13692	0.52	1/18477 (0.0%)

Chiral center outliers are detected by calculating the chiral volume of a chiral center and verifying if the center is modelled as a planar moiety or with the opposite hand.A planarity outlier is detected by checking planarity of atoms in a peptide group, atoms in a mainchain group or atoms of a sidechain that are expected to be planar.

Mol	Chain	#Chirality outliers	#Planarity outliers
1	A	0	1
1	B	0	1
All	All	0	2

There are no bond length outliers.

All (1) bond angle outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(^{o})	Ideal(^{o})
1	A	145	LEU	CA-CB-CG	5.10	127.03	115.30

There are no chirality outliers.

All (2) planarity outliers are listed below:

Mol	Chain	Res	Type	Group
1	A	384	ARG	Sidechain
1	B	384	ARG	Sidechain

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	6730	0	6826	114	0
1	B	6717	0	6810	136	0
2	A	43	0	0	0	0
2	B	43	0	0	0	0
All	All	13533	0	13636	243	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 9.

The worst 5 of 243 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)
1:B:855:VAL:HG13	1:B:870:CYS:SG	1.52	1.50
1:B:855:VAL:CG1	1:B:870:CYS:SG	2.43	1.05
1:B:855:VAL:HG13	1:B:870:CYS:HG	1.24	0.93
1:A:439:ILE:HG23	1:A:521:TRP:CZ2	2.11	0.86
1:B:926:LEU:HB3	1:B:930:GLN:HE21	1.49	0.77

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	847/1120 (76%)	811 (96%)	35 (4%)	1 (0%)	48 81
1	B	845/1120 (75%)	796 (94%)	49 (6%)	0	100 100

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Mol	Chain	Analysed	Favoured	Allowed	Outliers	Percentiles
All	All	1692/2240 (76%)	1607 (95%)	84 (5%)	1 (0%)	50 81

All (1) Ramachandran outliers are listed below:

Mol	Chain	Res	Type
1	A	428	ALA

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	743/968 (77%)	724 (97%)	19 (3%)	41 72
1	B	742/968 (77%)	716 (96%)	26 (4%)	31 65
All	All	1485/1936 (77%)	1440 (97%)	45 (3%)	37 69

5 of 45 residues with a non-rotameric sidechain are listed below:

Mol	Chain	Res	Type
1	B	419	LEU
1	B	760	ASN
1	B	440	MET
1	B	548	MET
1	B	861	ARG

Sometimes sidechains can be flipped to improve hydrogen bonding and reduce clashes. 5 of 14 such sidechains are listed below:

Mol	Chain	Res	Type
1	B	330	ASN
1	B	372	HIS
1	B	1013	GLN
1	B	906	GLN
1	B	980	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 oligosaccharides in this entry.

5.6 Ligand geometry (i)

2 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	$\#\|Z\| > 2$
2	O6E	A	1201	1	42,46,46	1.01	3 (7%)	50,67,67	1.17	3 (6%)
2	O6E	B	1201	1	42,46,46	0.99	3 (7%)	50,67,67	1.26	4 (8%)

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	O6E	A	1201	1	-	7/25/74/74	0/4/4/4
2	O6E	B	1201	1	-	6/25/74/74	0/4/4/4

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

Mol	Chain	Res	Type	Atoms	Z	Observed(Å)	Ideal(Å)
2	A	1201	O6E	CAO-CAV	3.39	1.37	1.35

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Mol	Chain	Res	Type	Atoms	Z	Observed(Å)	Ideal(Å)
2	B	1201	O6E	CAO-CAV	3.15	1.37	1.35
2	B	1201	O6E	CAB-CBI	-2.96	1.39	1.47
2	A	1201	O6E	CAB-CBI	-2.93	1.39	1.47
2	A	1201	O6E	CBI-CBC	2.14	1.41	1.37

The worst 5 of 7 bond angle outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(^{o})	Ideal(^{o})
2	B	1201	O6E	CBO-CAO-CAV	5.12	134.92	128.81
2	A	1201	O6E	CBO-CAO-CAV	4.71	134.43	128.81
2	B	1201	O6E	CAW-CAP-CBL	3.61	136.91	128.08
2	A	1201	O6E	CAW-CAP-CBL	3.24	136.00	128.08
2	B	1201	O6E	CBD-CBJ-CBN	2.38	104.90	101.34

There are no chirality outliers.

5 of 13 torsion outliers are listed below:

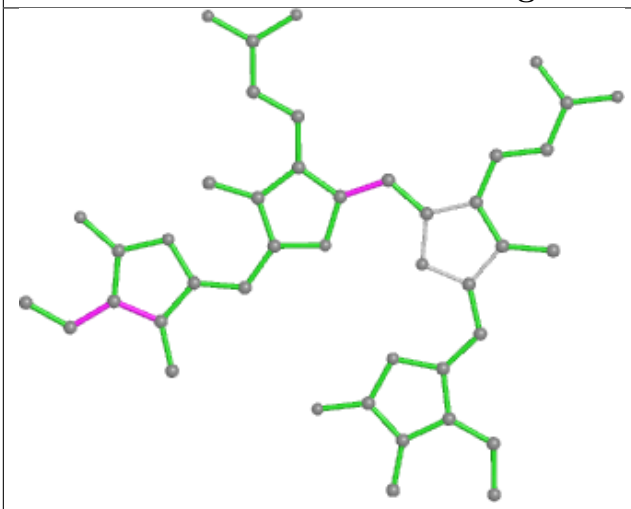
Mol	Chain	Res	Type	Atoms
2	A	1201	O6E	CAW-CAP-CBL-NAE
2	B	1201	O6E	CAW-CAP-CBL-NAE
2	A	1201	O6E	CAW-CAP-CBL-CBH
2	B	1201	O6E	CAW-CAP-CBL-CBH
2	A	1201	O6E	CAH-CAC-CBJ-CBN

There are no ring outliers.

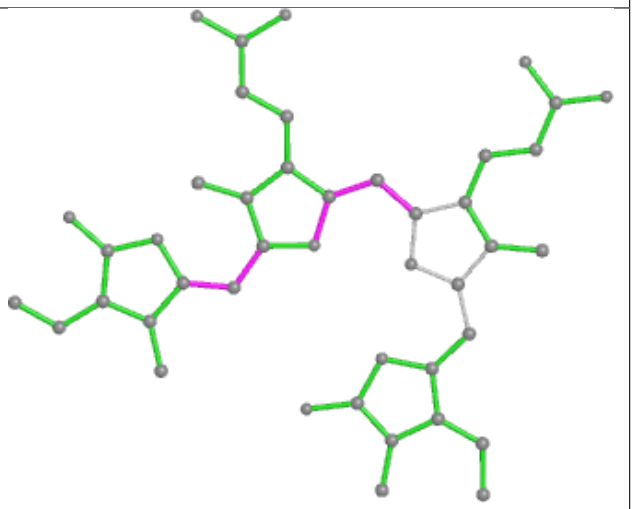
No monomer is involved in short contacts.

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.

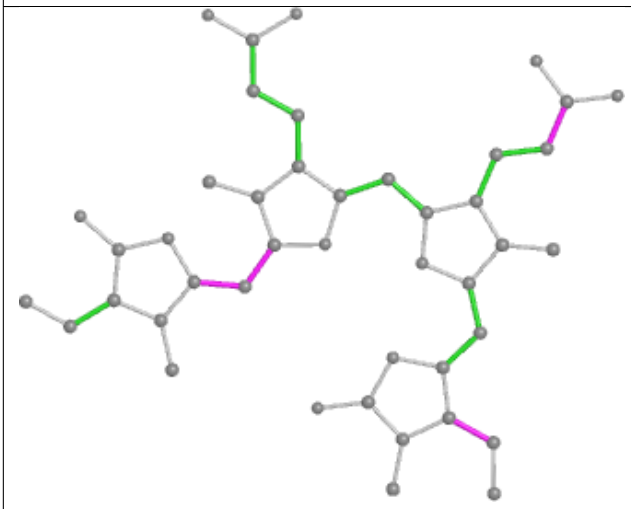
Ligand O6E A 1201



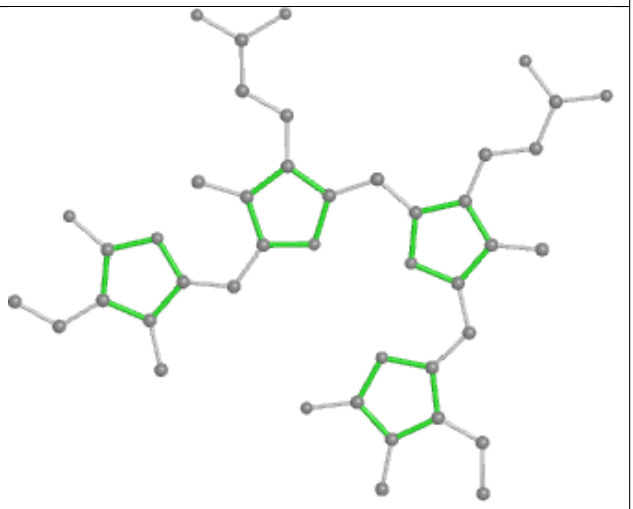
Bond lengths



Bond angles

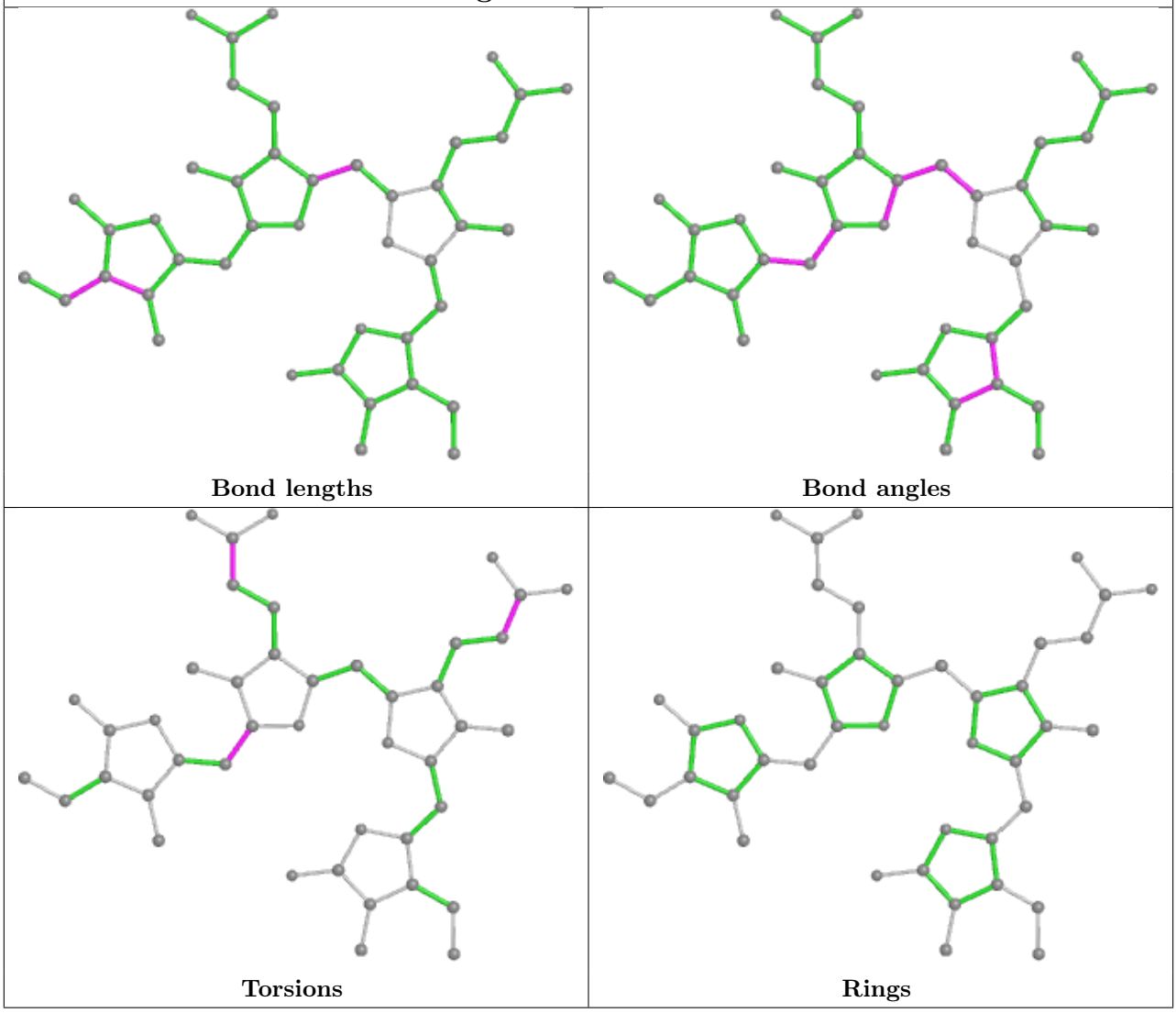


Torsions



Rings

Ligand O6E B 1201



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.