W O R L D W I D E

wwPDB

PROTEIN DATA BANK

wwPDB EM Validation Summary Report (i)

Apr 21, 2024 – 12:08 AM JST

PDB ID	:	8J74
EMDB ID	:	EMD-36025
Title	:	Human high-affinity choline transporter CHT1 in the HC-3-bound outward-facing open conformation, dimeric state
Authors	:	Gao, Y.; Qiu, Y.; Zhao, Y.
Deposited on	:	2023-04-27
Resolution	:	3.60 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 (i)) were used in the production of this report:

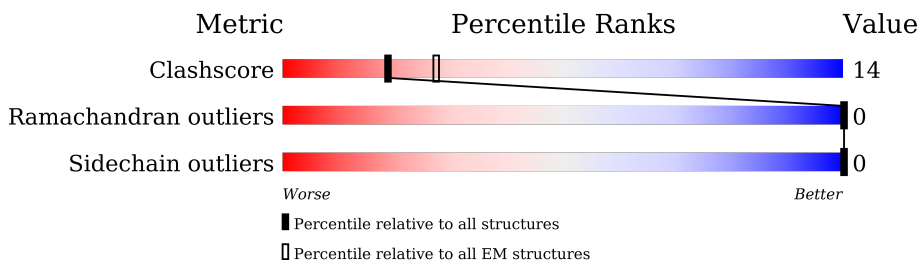|		
EMDB validation analysis	:	FAILED
Mogul	:	1.8.5 (274361), CSD as541be (2020)
MolProbity	:	4.02b-467
buster-report	:	1.1.7 (2018)
Percentile statistics	:	20191225.v01 (using entries in the PDB archive December 25th 2019)
MapQ	:	FAILED
Ideal geometry (proteins)	:	Engh & Huber (2001)
Ideal geometry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

1 Overall quality at a glance ⓘ

The following experimental techniques were used to determine the structure:
ELECTRON MICROSCOPY

The reported resolution of this entry is 3.60 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	Percentile Ranks	Value
Clashscore		14
Ramachandran outliers		0
Sidechain outliers		0

Worse   *Better*

▮ Percentile relative to all structures
▯ Percentile relative to all EM structures

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	580	64% 25% 11%
1	B	580	64% 25% 11%
2	E	3	67% 33%
2	F	3	67% 33%

WORLDWIDE
wwPDB
PROTEIN DATA BANK

2 Entry composition ⓘ

There are 6 unique types of molecules in this entry. The entry contains 8288 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 High affinity choline transporter 1.

Mol	Chain	Residues	Atoms					AltConf	Trace
1	A	515	Total 3959	C 2637	N 603	O 699	S 20	0	0
1	B	515	Total 3959	C 2637	N 603	O 699	S 20	0	0

- Molecule 2 is an oligosaccharide called beta-D-mannopyranose-(1-4)-2-acetamido-2-deoxy-beta-D-glucopyranose-(1-4)-2-acetamido-2-deoxy-beta-D-glucopyranose.
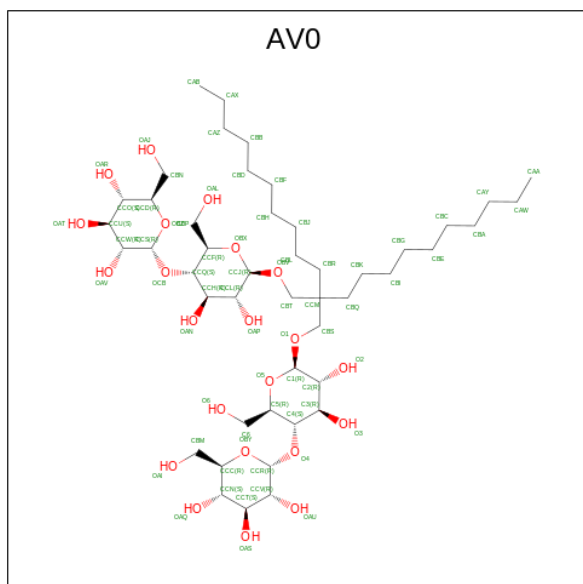

Mol	Chain	Residues	Atoms				AltConf	Trace
2	E	3	Total 39	C 22	N 2	O 15	0	0
2	F	3	Total 39	C 22	N 2	O 15	0	0

- Molecule 3 is CHOLESTEROL HEMISUCCINATE (three-letter code: Y01) (formula: $C_{31}H_{50}O_4$) (labeled as "Ligand of Interest" by depositor).
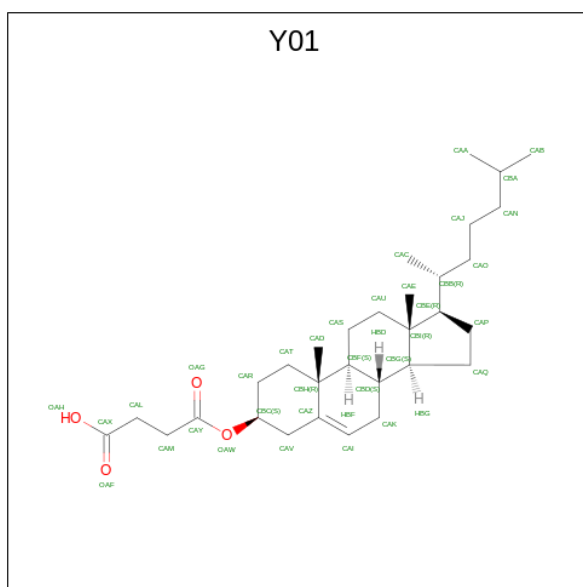
Mol	Chain	Residues	Atoms			AltConf
3	A	1	Total 35	C 31	O 4	0
3	B	1	Total 35	C 31	O 4	0

- Molecule 4 is Lauryl Maltose Neopentyl Glycol (three-letter code: AV0) (formula: $C_{47}H_{88}O_{22}$) (labeled as "Ligand of Interest" by depositor).
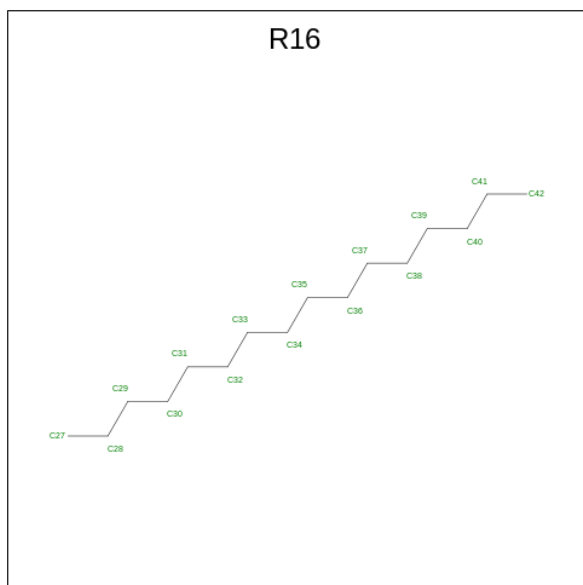

Mol	Chain	Residues	Atoms			AltConf
4	A	1	Total 69	C 47	O 22	0

Continued on next page...

Continued from previous page...

Mol	Chain	Residues	Atoms			AltConf
4	B	1	Total	C	O	0
			69	47	22	

- Molecule 5 is HEXADECANE (three-letter code: R16) (formula: $C_{16}H_{34}$) (labeled as "Ligand of Interest" by depositor).

R16

Mol	Chain	Residues	Atoms		AltConf
5	A	1	Total	C	0
			12	12	
5	B	1	Total	C	0
			12	12	

- Molecule 6 is (2S,2'S)-2,2'-biphenyl-4,4'-diylbis(2-hydroxy-4,4-dimethylmorpholin-4-ium) (three-letter code: HC6) (formula: $C_{24}H_{34}N_2O_4$) (labeled as "Ligand of Interest" by depositor).
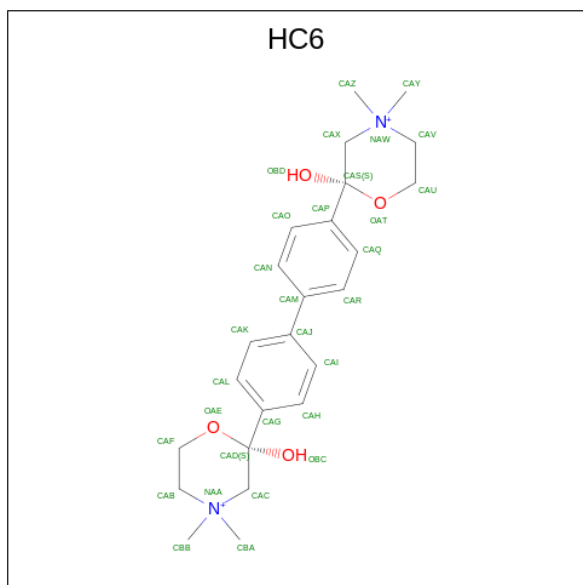
Mol	Chain	Residues	Atoms				AltConf
6	A	1	Total	C	N	O	0
			30	24	2	4	
6	B	1	Total	C	N	O	0
			30	24	2	4	

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: High affinity choline transporter 1

Chain A:
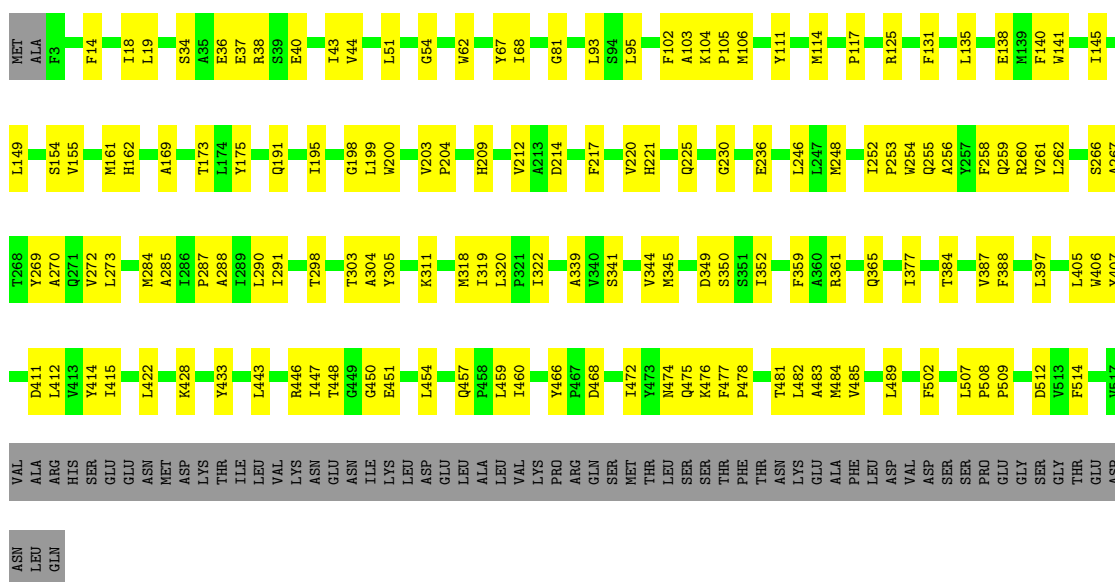
• Molecule 1: High affinity choline transporter 1

Chain B:
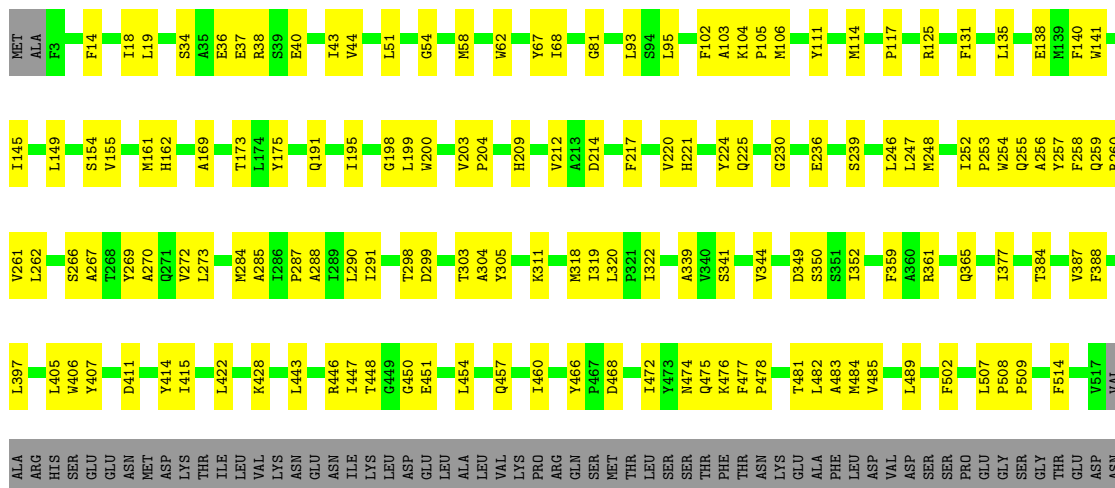
LEU
GLN

• Molecule 2: beta-D-mannopyranose-(1-4)-2-acetamido-2-deoxy-beta-D-glucopyranose-(1-4)-2-ac
etamido-2-deoxy-beta-D-glucopyranose

Chain E: 67%   33%

NAG1
NAG2
BMA3

• Molecule 2: beta-D-mannopyranose-(1-4)-2-acetamido-2-deoxy-beta-D-glucopyranose-(1-4)-2-ac
etamido-2-deoxy-beta-D-glucopyranose

Chain F: 67%   33%

NAG1
NAG2
BMA3

4 Experimental information ⓘ

Property	Value	Source
EM reconstruction method	SINGLE PARTICLE	Depositor
Imposed symmetry	POINT, C2	Depositor
Number of particles used	12269	Depositor
Resolution determination method	FSC 0.143 CUT-OFF	Depositor
CTF correction method	PHASE FLIPPING ONLY	Depositor
Microscope	FEI TITAN KRIOS	Depositor
Voltage (kV)	300	Depositor
Electron dose $(e^-/\text{Å}^2)$	60	Depositor
Minimum defocus (nm)	1200	Depositor
Maximum defocus (nm)	2200	Depositor
Magnification	165000	Depositor
Image detector	GATAN K2 SUMMIT (4k 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: R16, Y01, HC6, AV0, BMA, NAG

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	0/4071	0.46	0/5560
1	B	0.27	0/4071	0.46	0/5560
All	All	0.27	0/8142	0.46	0/11120

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	3959	0	3995	117	0
1	B	3959	0	3995	116	0
2	E	39	0	34	1	0
2	F	39	0	34	1	0
3	A	35	0	49	0	0
3	B	35	0	49	0	0
4	A	69	0	0	2	0
4	B	69	0	0	2	0
5	A	12	0	20	2	0
5	B	12	0	20	2	0
6	A	30	0	34	13	0

Continued on next page...

Continued from previous page...

Mol	Chain	Non-H	H(model)	H(added)	Clashes	Symm-Clashes
6	B	30	0	34	12	0
All	All	8288	0	8264	239	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 14.

The worst 5 of 239 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:A:141:TRP:CE2	6:A:704:HC6:HBAB	1.96	1.01
1:B:361:ARG:HH11	1:B:365:GLN:NE2	1.58	1.00
1:A:361:ARG:HH11	1:A:365:GLN:NE2	1.58	1.00
1:B:141:TRP:CE2	6:B:604:HC6:HBAB	1.96	0.99
1:B:361:ARG:HH11	1:B:365:GLN:HE21	1.08	0.97

There are no symmetry-related clashes.

5.3   Torsion angles ⓘ

5.3.1   Protein backbone ⓘ

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	513/580 (88%)	482 (94%)	31 (6%)	0	100	100
1	B	513/580 (88%)	482 (94%)	31 (6%)	0	100	100
All	All	1026/1160 (88%)	964 (94%)	62 (6%)	0	100	100

There are no Ramachandran outliers to report.

5.3.2   Protein sidechains ⓘ

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	415/474 (88%)	415 (100%)	0	100	100
1	B	415/474 (88%)	415 (100%)	0	100	100
All	All	830/948 (88%)	830 (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 (4) such sidechains are listed below:

Mol	Chain	Res	Type
1	A	365	GLN
1	A	491	ASN
1	B	365	GLN
1	B	491	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 ⓘ

6 monosaccharides 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	NAG	E	1	1,2	14,14,15	0.44	0	17,19,21	1.24	1 (5%)

Mol	Type	Chain	Res	Link	Bond lengths			Bond angles		
					Counts	RMSZ	$\#\|Z\| > 2$	Counts	RMSZ	$\#\|Z\| > 2$
2	NAG	E	2	2	14,14,15	0.31	0	17,19,21	0.52	0
2	BMA	E	3	2	11,11,12	0.54	0	15,15,17	0.83	0
2	NAG	F	1	1,2	14,14,15	0.46	0	17,19,21	1.24	1 (5%)
2	NAG	F	2	2	14,14,15	0.31	0	17,19,21	0.52	0
2	BMA	F	3	2	11,11,12	0.54	0	15,15,17	0.83	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	NAG	E	1	1,2	-	5/6/23/26	0/1/1/1
2	NAG	E	2	2	-	0/6/23/26	0/1/1/1
2	BMA	E	3	2	-	0/2/19/22	0/1/1/1
2	NAG	F	1	1,2	-	5/6/23/26	0/1/1/1
2	NAG	F	2	2	-	0/6/23/26	0/1/1/1
2	BMA	F	3	2	-	0/2/19/22	0/1/1/1

There are no bond length outliers.

All (2) bond angle outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(^{o})	Ideal(^{o})
2	E	1	NAG	C2-N2-C7	4.27	128.99	122.90
2	F	1	NAG	C2-N2-C7	4.27	128.99	122.90

There are no chirality outliers.

5 of 10 torsion outliers are listed below:

Mol	Chain	Res	Type	Atoms
2	E	1	NAG	C8-C7-N2-C2
2	E	1	NAG	O7-C7-N2-C2
2	F	1	NAG	C8-C7-N2-C2
2	F	1	NAG	O7-C7-N2-C2
2	E	1	NAG	O5-C5-C6-O6

There are no ring outliers.

2 monomers are involved in 2 short contacts:

Mol	Chain	Res	Type	Clashes	Symm-Clashes
2	F	1	NAG	1	0
2	E	1	NAG	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 oligosaccharide.

Oligosaccharide Chain E

Bond lengths

Bond angles

Torsions

Rings

Oligosaccharide Chain F

Bond lengths

Bond angles

Torsions

Rings
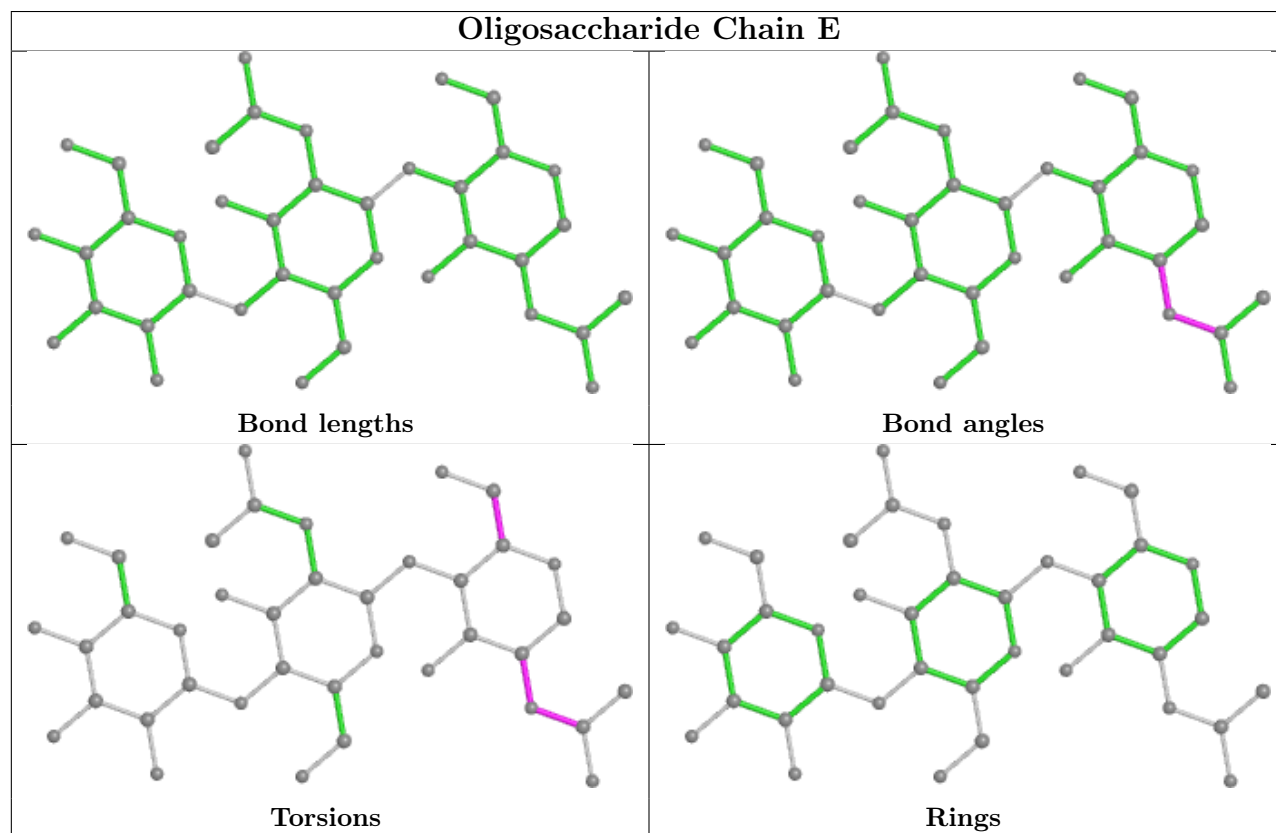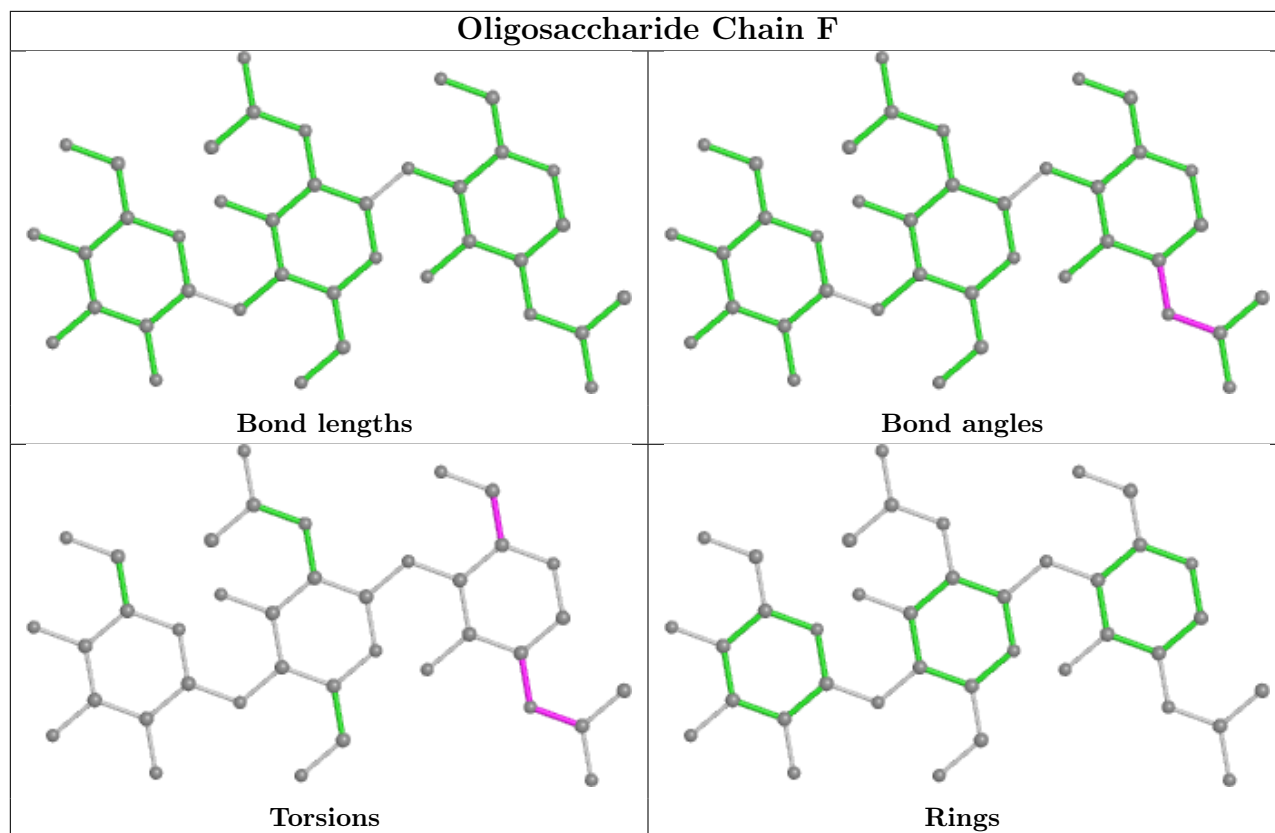
5.6 Ligand geometry (i)

8 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	$\#\|Z\| > 2$
3	Y01	B	603	-	38,38,38	0.37	0	57,57,57	0.43	0
4	AV0	B	601	-	72,72,72	0.14	0	96,98,98	0.53	0
5	R16	B	602	-	11,11,15	0.15	0	10,10,14	0.12	0
6	HC6	A	704	-	30,33,33	0.56	0	36,52,52	0.43	0
4	AV0	A	702	-	72,72,72	0.14	0	96,98,98	0.53	0
5	R16	A	703	-	11,11,15	0.15	0	10,10,14	0.12	0
6	HC6	B	604	-	30,33,33	0.56	0	36,52,52	0.43	0
3	Y01	A	701	-	38,38,38	0.37	0	57,57,57	0.43	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
3	Y01	B	603	-	-	6/19/77/77	0/4/4/4
4	AV0	B	601	-	-	9/50/130/130	0/4/4/4
5	R16	B	602	-	-	1/9/9/13	-
6	HC6	A	704	-	-	10/12/44/44	0/4/4/4
4	AV0	A	702	-	-	9/50/130/130	0/4/4/4
5	R16	A	703	-	-	1/9/9/13	-
6	HC6	B	604	-	-	10/12/44/44	0/4/4/4
3	Y01	A	701	-	-	6/19/77/77	0/4/4/4

There are no bond length outliers.

There are no bond angle outliers.

There are no chirality outliers.

5 of 52 torsion outliers are listed below:

Mol	Chain	Res	Type	Atoms
4	A	702	AV0	OBX-CCJ-OBV-CBT
4	B	601	AV0	OBX-CCJ-OBV-CBT
6	A	704	HC6	CAC-CAD-CAG-CAL
6	A	704	HC6	CAC-CAD-CAG-CAH
6	A	704	HC6	OBC-CAD-CAG-CAL

There are no ring outliers.
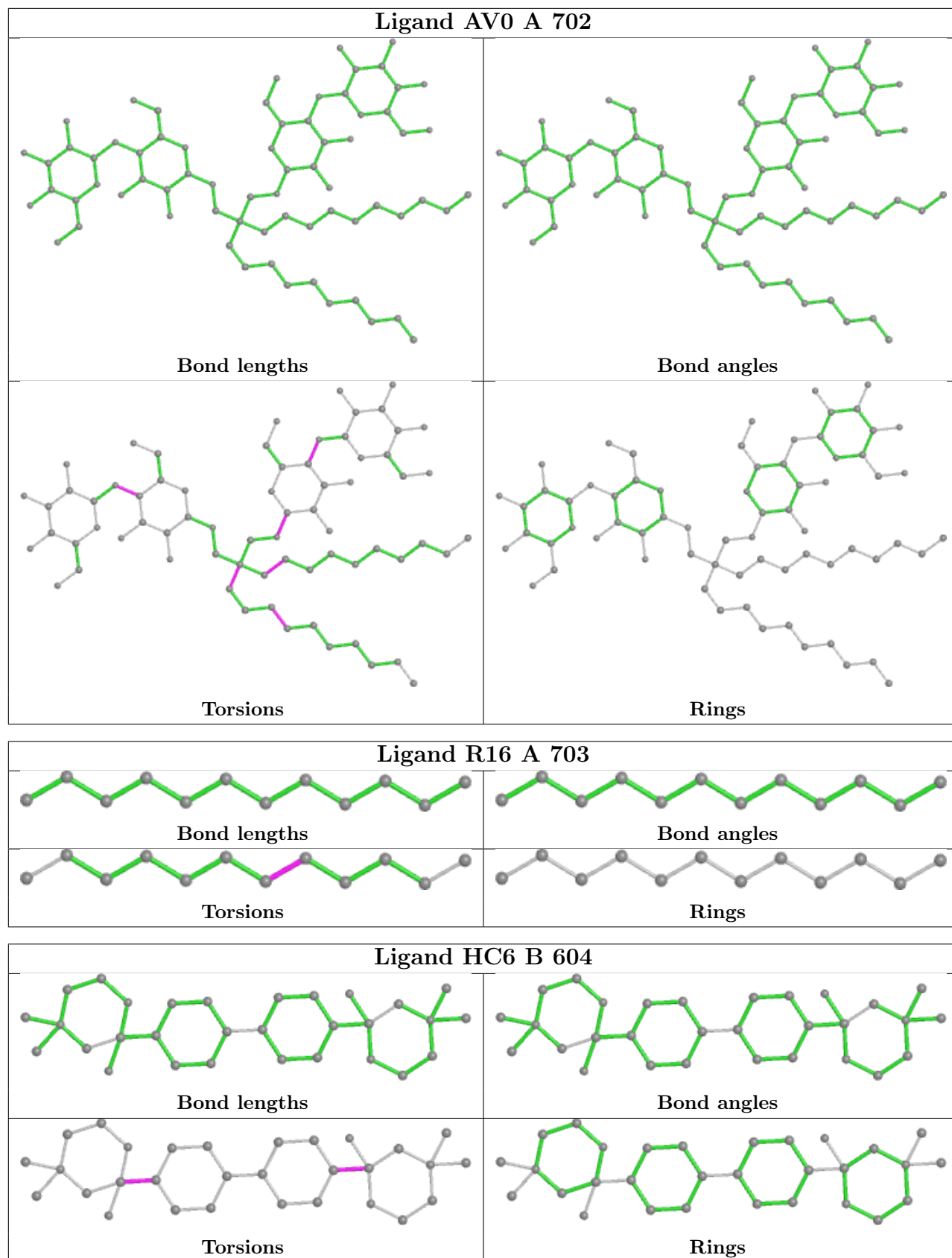
6 monomers are involved in 33 short contacts:

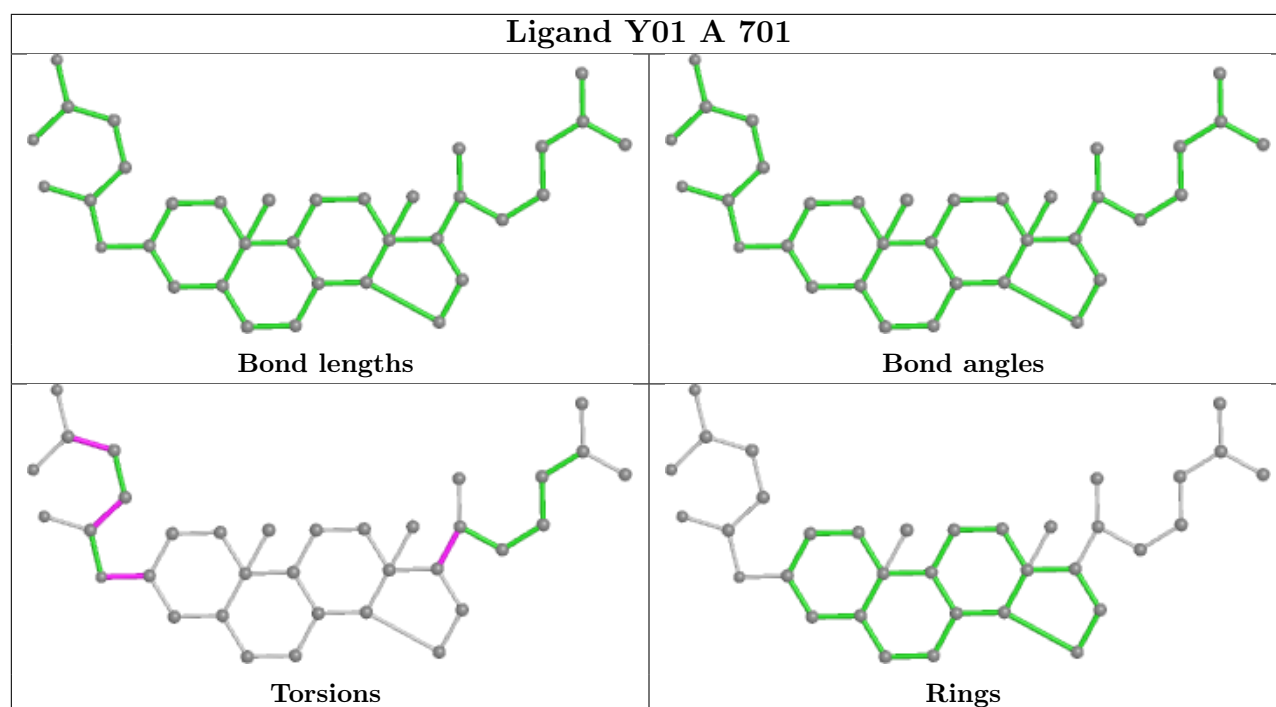| Mol	Chain	Res	Type	Clashes	Symm-Clashes
4	B	601	AV0	2	0
5	B	602	R16	2	0
6	A	704	HC6	13	0
4	A	702	AV0	2	0
5	A	703	R16	2	0
6	B	604	HC6	12	0

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 Y01 B 603

Bond lengths

Bond angles

Torsions

Rings

Ligand AV0 B 601

Bond lengths

Bond angles

Torsions

Rings

Ligand R16 B 602

Bond lengths

Bond angles

Torsions

Rings

Ligand HC6 A 704

Bond lengths

Bond angles

Torsions

Rings

Ligand AV0 A 702

Bond lengths

Bond angles

Torsions

Rings

Ligand R16 A 703

Bond lengths

Bond angles

Torsions

Rings

Ligand HC6 B 604

Bond lengths

Bond angles

Torsions

Rings

Ligand Y01 A 701

5.7 Other polymers ⓘ

There are no such residues in this entry.

5.8 Polymer linkage issues ⓘ

There are no chain breaks in this entry.