Full wwPDB X-ray Structure Validation Report ⓘ

Feb 4, 2025 – 12:10 PM EST

PDB ID	:	9MDK
Title	:	Crystal Structure of E64-inhibited C0362 (TDE_0362 [TDE0362] resi 205-647)
Authors	:	Clark, N.D.; Malkowski, M.G.
Deposited on	:	2024-12-05
Resolution	:	1.93 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 ⓘ 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:

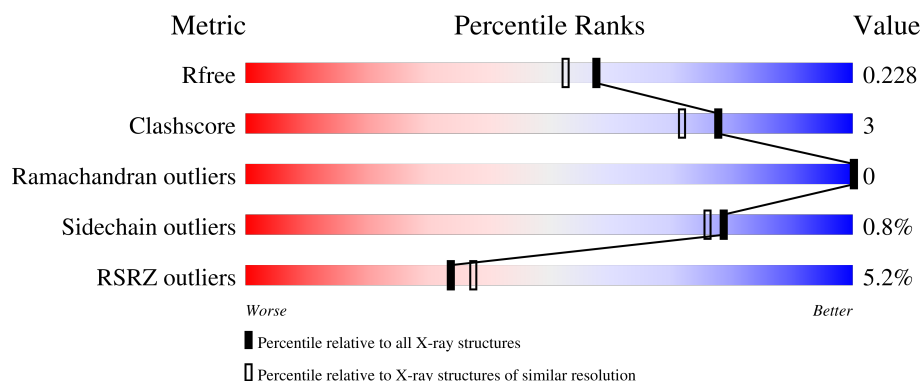|		
MolProbity	:	4.02b-467
Mogul	:	2022.3.0, CSD as543be (2022)
Xtriage (Phenix)	:	1.21
EDS	:	3.0
buster-report	:	1.1.7 (2018)
Percentile statistics	:	20231227.v01 (using entries in the PDB archive December 27th 2023)
CCP4	:	9.0.004 (Gargrove)
Density-Fitness	:	1.0.11
Ideal geometry (proteins)	:	Engh & Huber (2001)
Ideal geometry (DNA, RNA)	:	Parkinson et al. (1996)
Validation Pipeline (wwPDB-VP)	:	2.40

1 Overall quality at a glance ⓘ

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

The reported resolution of this entry is 1.93 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
Rfree | | 0.228
Clashscore | | 3
Ramachandran outliers | | 0
Sidechain outliers | | 0.8%
RSRZ outliers | | 5.2%

Worse .... *Better*

▌ Percentile relative to all X-ray structures
◨ Percentile relative to X-ray structures of similar resolution

Metric	Whole archive (#Entries)	Similar resolution (#Entries, resolution range(Å))
R$_{free}$	164625	1306 (1.94-1.94)
Clashscore	180529	1400 (1.94-1.94)
Ramachandran outliers	177936	1387 (1.94-1.94)
Sidechain outliers	177891	1387 (1.94-1.94)
RSRZ outliers	164620	1306 (1.94-1.94)

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
1	A	476	5% ▬ 86% 6% 8%

2 Entry composition ⓘ

There are 4 unique types of molecules in this entry. The entry contains 4039 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 Bacterial Ig-like domain protein C0362.

Mol	Chain	Residues	Atoms					ZeroOcc	AltConf	Trace
			Total	C	N	O	S			
1	A	439	3628	2330	600	687	11	0	11	0

There are 33 discrepancies between the modelled and reference sequences:

Chain	Residue	Modelled	Actual	Comment	Reference
A	204	MET	-	initiating methionine	UNP Q73QT1
A	648	LYS	-	expression tag	UNP Q73QT1
A	649	GLY	-	expression tag	UNP Q73QT1
A	650	GLU	-	expression tag	UNP Q73QT1
A	651	LEU	-	expression tag	UNP Q73QT1
A	652	ASN	-	expression tag	UNP Q73QT1
A	653	SER	-	expression tag	UNP Q73QT1
A	654	LYS	-	expression tag	UNP Q73QT1
A	655	LEU	-	expression tag	UNP Q73QT1
A	656	GLU	-	expression tag	UNP Q73QT1
A	657	GLY	-	expression tag	UNP Q73QT1
A	658	LYS	-	expression tag	UNP Q73QT1
A	659	PRO	-	expression tag	UNP Q73QT1
A	660	ILE	-	expression tag	UNP Q73QT1
A	661	PRO	-	expression tag	UNP Q73QT1
A	662	ASN	-	expression tag	UNP Q73QT1
A	663	PRO	-	expression tag	UNP Q73QT1
A	664	LEU	-	expression tag	UNP Q73QT1
A	665	LEU	-	expression tag	UNP Q73QT1
A	666	GLY	-	expression tag	UNP Q73QT1
A	667	LEU	-	expression tag	UNP Q73QT1
A	668	ASP	-	expression tag	UNP Q73QT1
A	669	SER	-	expression tag	UNP Q73QT1
A	670	THR	-	expression tag	UNP Q73QT1
A	671	ARG	-	expression tag	UNP Q73QT1
A	672	THR	-	expression tag	UNP Q73QT1
A	673	GLY	-	expression tag	UNP Q73QT1

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Chain	Residue	Modelled	Actual	Comment	Reference
A	674	HIS	-	expression tag	UNP Q73QT1
A	675	HIS	-	expression tag	UNP Q73QT1
A	676	HIS	-	expression tag	UNP Q73QT1
A	677	HIS	-	expression tag	UNP Q73QT1
A	678	HIS	-	expression tag	UNP Q73QT1
A	679	HIS	-	expression tag	UNP Q73QT1

- Molecule 2 is N-[N-[1-HYDROXYCARBOXYETHYL-CARBONYL]LEUCYLAMINO-BUTYL]-GUANIDINE (three-letter code: E64) (formula: $C_{15}H_{30}N_5O_5$) (labeled as "Ligand of Interest" by depositor).
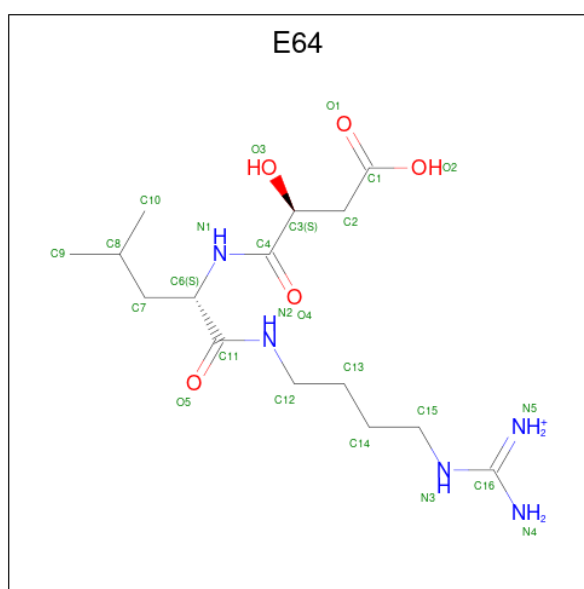
Mol	Chain	Residues	Atoms				ZeroOcc	AltConf
2	A	1	Total 25	C 15	N 5	O 5	0	0

- Molecule 3 is 1,2-ETHANEDIOL (three-letter code: EDO) (formula: $C_2H_6O_2$).

EDO

O2

OH

C1

C2

HO

O1

Mol	Chain	Residues	Atoms			ZeroOcc	AltConf
3	A	1	Total 4	C 2	O 2	0	0
3	A	1	Total 4	C 2	O 2	0	0
3	A	1	Total 4	C 2	O 2	0	0
3	A	1	Total 4	C 2	O 2	0	0

- Molecule 4 is water.

Mol	Chain	Residues	Atoms		ZeroOcc	AltConf
4	A	369	Total 370	O 370	0	1

3 Residue-property plots ⓘ

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: Bacterial Ig-like domain protein C0362
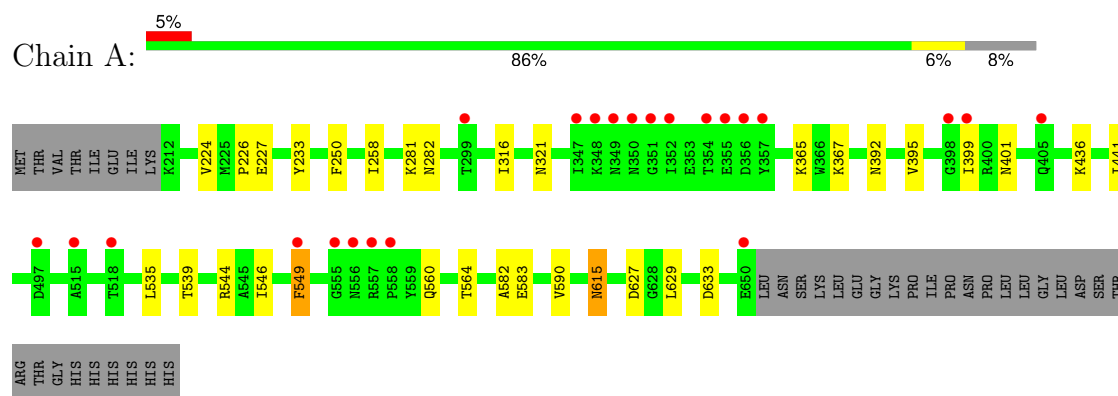
4 Data and refinement statistics (i)

Property	Value	Source
Space group	P 2 21 21	Depositor
Cell constants a, b, c, α, β, γ	41.68Å 99.37Å 121.22Å 90.00° 90.00° 90.00°	Depositor
Resolution (Å)	76.85 – 1.93 76.85 – 1.93	Depositor EDS
% Data completeness (in resolution range)	98.5 (76.85-1.93) 98.6 (76.85-1.93)	Depositor EDS
R_{merge}	0.14	Depositor
R_{sym}	(Not available)	Depositor
$< I/\sigma(I) >$ [1]	1.35 (at 1.92Å)	Xtriage
Refinement program	PHENIX 1.21_5207	Depositor
R, R_{free}	0.185 , 0.228 0.186 , 0.228	Depositor DCC
R_{free} test set	1972 reflections (5.05%)	wwPDB-VP
Wilson B-factor (Å^2)	27.9	Xtriage
Anisotropy	0.804	Xtriage
Bulk solvent k_{sol}(e/Å^3), B_{sol}(Å^2)	0.32 , 49.9	EDS
L-test for twinning[2]	$< |L| > = 0.49$, $< L^2 > = 0.33$	Xtriage
Estimated twinning fraction	No twinning to report.	Xtriage
F_o,F_c correlation	0.95	EDS
Total number of atoms	4039	wwPDB-VP
Average B, all atoms (Å^2)	39.0	wwPDB-VP

Xtriage's analysis on translational NCS is as follows: *The largest off-origin peak in the Patterson function is 5.08% of the height of the origin peak. No significant pseudotranslation is detected.*

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[1]Intensities estimated from amplitudes.
[2]Theoretical 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.

5 Model quality ⓘ

5.1 Standard geometry ⓘ

Bond lengths and bond angles in the following residue types are not validated in this section: E64, EDO

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	0.41	0/3723	0.58	0/5049

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	3628	0	3400	19	0
2	A	25	0	28	1	0
3	A	16	0	24	1	0
4	A	370	0	0	1	0
All	All	4039	0	3452	19	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 3.

All (19) 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:227:GLU:HB2	3:A:705:EDO:H21	1.80	0.64
1:A:535:LEU:HG	1:A:629:LEU:HD22	1.79	0.64
1:A:392:ASN:O	1:A:395:VAL:HG12	2.02	0.58
1:A:549:PHE:HD2	1:A:627:ASP:HB3	1.73	0.54
1:A:539:THR:HG21	1:A:546:ILE:HG12	1.92	0.52
1:A:560:GLN:HB3	2:A:701:E64:H71	1.91	0.52
1:A:399:ILE:C	1:A:401:ASN:H	2.14	0.51
1:A:281:LYS:NZ	4:A:808:HOH:O	2.45	0.48
1:A:436[A]:LYS:HA	1:A:441[A]:ILE:HD11	1.96	0.47
1:A:365:LYS:HD2	1:A:590:VAL:HG21	1.96	0.47
1:A:549:PHE:CD2	1:A:627:ASP:HB3	2.48	0.47
1:A:224:VAL:HB	1:A:316:ILE:HD13	1.97	0.46
1:A:564:THR:HB	1:A:582:ALA:HB3	1.99	0.43
1:A:226:PRO:HB3	1:A:233:TYR:CG	2.54	0.42
1:A:544:ARG:HG2	1:A:633:ASP:HA	2.02	0.42
1:A:250:PHE:CE2	1:A:258:ILE:HG21	2.55	0.41
1:A:282:ASN:O	1:A:321:ASN:HB2	2.21	0.41
1:A:367:LYS:HA	1:A:590:VAL:HG12	2.02	0.41
1:A:615:ASN:C	1:A:615:ASN:HD22	2.23	0.41

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 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	448/476 (94%)	435 (97%)	13 (3%)	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 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	Rotameric	Outliers	Percentiles	
1	A	365/418 (87%)	362 (99%)	3 (1%)	79	76

All (3) residues with a non-rotameric sidechain are listed below:

Mol	Chain	Res	Type
1	A	549	PHE
1	A	583	GLU
1	A	615	ASN

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	522	ASN
1	A	551	GLN
1	A	560	GLN
1	A	615	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 ⓘ

There are no oligosaccharides in this entry.

5.6 Ligand geometry ⓘ

5 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	EDO	A	705	-	3,3,3	0.45	0	2,2,2	0.42	0
3	EDO	A	704	-	3,3,3	0.43	0	2,2,2	0.27	0
3	EDO	A	703	-	3,3,3	0.42	0	2,2,2	0.55	0
2	E64	A	701	1	24,24,24	2.31	5 (20%)	27,30,30	0.81	0
3	EDO	A	702	-	3,3,3	0.42	0	2,2,2	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	EDO	A	705	-	-	1/1/1/1	-
3	EDO	A	704	-	-	0/1/1/1	-
3	EDO	A	703	-	-	0/1/1/1	-
2	E64	A	701	1	-	9/29/29/29	-
3	EDO	A	702	-	-	0/1/1/1	-

All (5) bond length outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(Å)	Ideal(Å)
2	A	701	E64	C16-N3	6.57	1.45	1.33
2	A	701	E64	C4-N1	5.37	1.45	1.34
2	A	701	E64	C11-N2	5.24	1.45	1.33
2	A	701	E64	C16-N4	3.12	1.45	1.34
2	A	701	E64	O4-C4	-2.14	1.19	1.23

There are no bond angle outliers.

There are no chirality outliers.

All (10) torsion outliers are listed below:

Mol	Chain	Res	Type	Atoms
2	A	701	E64	O3-C3-C4-O4
2	A	701	E64	O3-C3-C4-N1
2	A	701	E64	C6-C7-C8-C10

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Mol	Chain	Res	Type	Atoms
2	A	701	E64	C11-C6-C7-C8
3	A	705	EDO	O1-C1-C2-O2
2	A	701	E64	N1-C6-C7-C8
2	A	701	E64	C6-C7-C8-C9
2	A	701	E64	C2-C3-C4-O4
2	A	701	E64	C2-C3-C4-N1
2	A	701	E64	C12-C13-C14-C15

There are no ring outliers.

2 monomers are involved in 2 short contacts:

Mol	Chain	Res	Type	Clashes	Symm-Clashes
3	A	705	EDO	1	0
2	A	701	E64	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.

Ligand E64 A 701

Bond lengths

Bond angles

Torsions

Rings
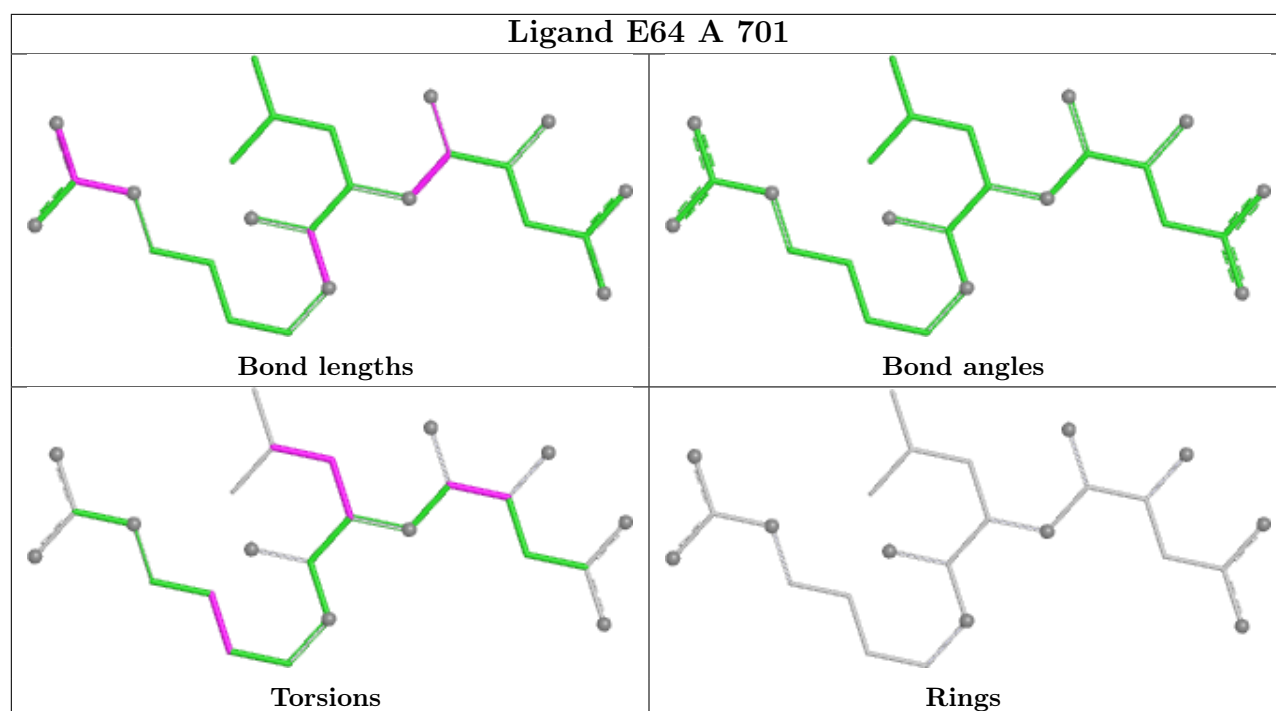
5.7 Other polymers ⓘ

There are no such residues in this entry.

5.8 Polymer linkage issues ⓘ

There are no chain breaks in this entry.

6 Fit of model and data ⓘ

6.1 Protein, DNA and RNA chains ⓘ

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, 95th 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>2			OWAB(Å^2)	Q<0.9
1	A	439/476 (92%)	0.21	23 (5%)	34	37	12, 36, 69, 100	11 (2%)

All (23) RSRZ outliers are listed below:

Mol	Chain	Res	Type	RSRZ
1	A	399	ILE	4.0
1	A	515	ALA	3.8
1	A	398	GLY	3.8
1	A	347	ILE	3.8
1	A	354	THR	3.4
1	A	356	ASP	3.3
1	A	349	ASN	3.2
1	A	350	ASN	3.0
1	A	357	TYR	3.0
1	A	556	ASN	2.9
1	A	352	ILE	2.7
1	A	351	GLY	2.6
1	A	558	PRO	2.6
1	A	497	ASP	2.6
1	A	557	ARG	2.4
1	A	650	GLU	2.4
1	A	549	PHE	2.4
1	A	518	THR	2.3
1	A	299	THR	2.2
1	A	355	GLU	2.2
1	A	555	GLY	2.2
1	A	405	GLN	2.2
1	A	348	LYS	2.1

6.2 Non-standard residues in protein, DNA, RNA chains ⓘ

There are no non-standard protein/DNA/RNA residues in this entry.

6.3 Carbohydrates ⓘ

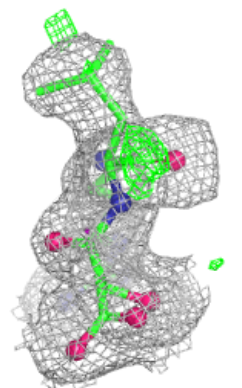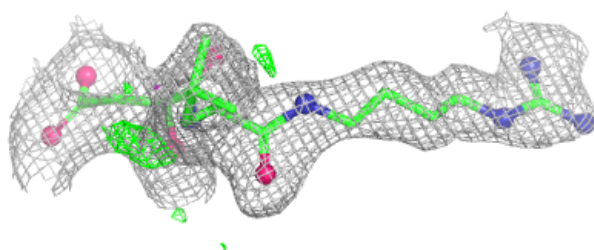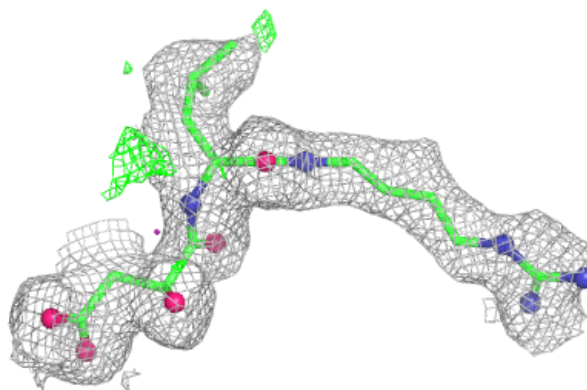There are no monosaccharides in this entry.

6.4 Ligands ⓘ

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, 95th 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	B-factors(Å^2)	Q<0.9
3	EDO	A	705	4/4	0.78	0.16	48,60,64,67	0
3	EDO	A	703	4/4	0.79	0.13	48,52,55,56	0
3	EDO	A	704	4/4	0.83	0.13	50,55,56,63	0
3	EDO	A	702	4/4	0.84	0.12	43,45,52,60	0
2	E64	A	701	25/25	0.88	0.12	38,49,62,64	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 E64 A 701:

$2mF_o\text{-}DF_c$ (at 0.7 rmsd) in gray
$mF_o\text{-}DF_c$ (at 3 rmsd) in purple (negative)
and green (positive)

6.5 Other polymers ⓘ

There are no such residues in this entry.