WORLDWIDE wwPDB PROTEIN DATA BANK

wwPDB X-ray Structure Validation Summary Report ⓘ

Nov 16, 2023 – 09:54 AM JST

PDB ID	:	6LHI
Title	:	Quadruple mutant (N51I+C59R+S108N+I164L) plasmodium falciparum dihydrofolate reductase-thymidylate synthase (PfDHFR-TS) complexed with C466 (compound 42) and NADPH
Authors	:	Vanichtanankul, J.; Vitsupakorn, D.
Deposited on	:	2019-12-09
Resolution	:	2.59 Å(reported)

The following versions of software and data (see references ⓘ) were used in the production of this report:

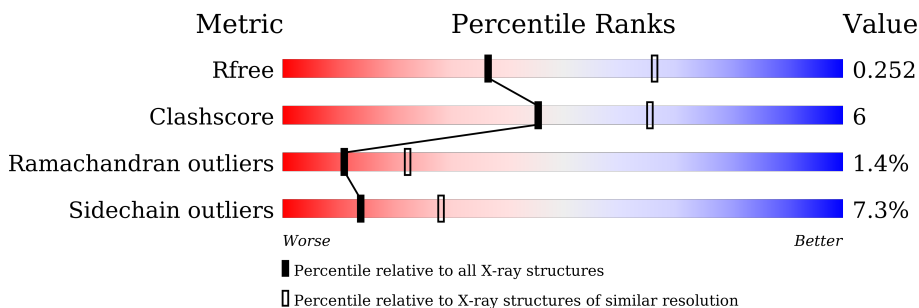|		
MolProbity	:	4.02b-467
Mogul	:	1.8.5 (274361), CSD as541be (2020)
Xtriage (Phenix)	:	1.13
EDS	:	2.36
buster-report	:	1.1.7 (2018)
Percentile statistics	:	20191225.v01 (using entries in the PDB archive December 25th 2019)
Refmac	:	5.8.0158
CCP4	:	7.0.044 (Gargrove)
Ideal geometry (proteins)	:	Engh & Huber (2001)
Ideal geometry (DNA, RNA)	:	Parkinson et al. (1996)
Validation Pipeline (wwPDB-VP)	:	2.36

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 2.59 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		0.252
Clashscore		6
Ramachandran outliers		1.4%
Sidechain outliers		7.3%

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}	130704	3163 (2.60-2.60)
Clashscore	141614	3518 (2.60-2.60)
Ramachandran outliers	138981	3455 (2.60-2.60)
Sidechain outliers	138945	3455 (2.60-2.60)

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%

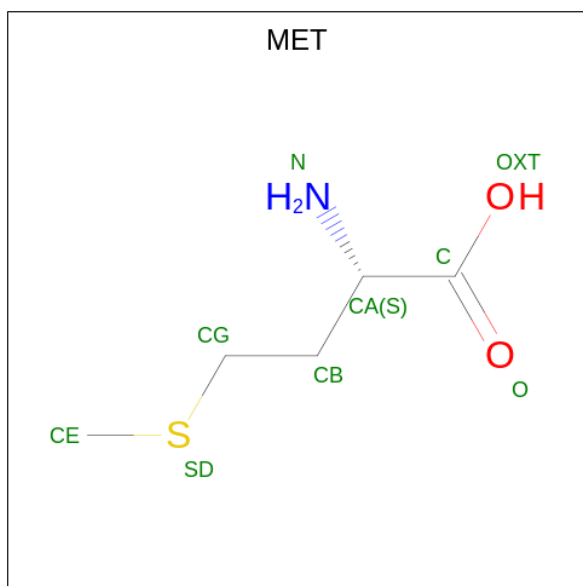| Mol	Chain	Length	Quality of chain
1	A	608	71% 15% • 12%
1	B	608	66% 18% • 13%

2 Entry composition ⓘ

There are 6 unique types of molecules in this entry. The entry contains 18078 atoms, of which 8923 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 Bifunctional dihydrofolate reductase-thymidylate synthase.
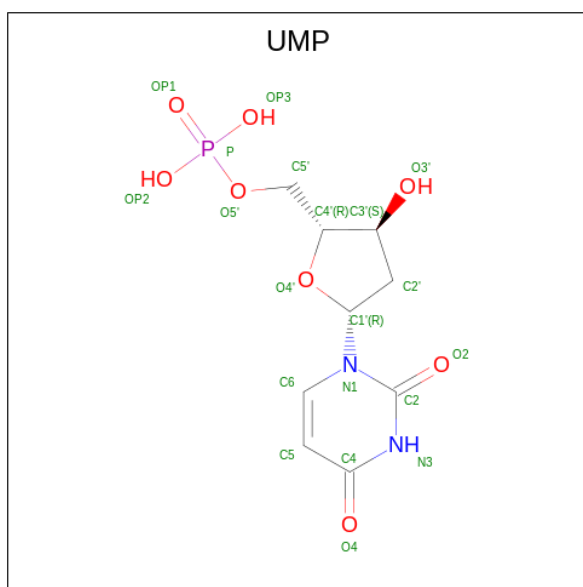
Mol	Chain	Residues	Atoms						ZeroOcc	AltConf	Trace
1	A	532	Total	C	H	N	O	S	256	0	0
			8832	2867	4399	733	809	24			
1	B	531	Total	C	H	N	O	S	254	0	0
			8813	2862	4389	730	807	25			

- Molecule 2 is METHIONINE (three-letter code: MET) (formula: $C_5H_{11}NO_2S$).

Mol	Chain	Residues	Atoms						ZeroOcc	AltConf
2	A	1	Total	C	H	N	O	S	0	0
			19	5	11	1	1	1		

- Molecule 3 is 2'-DEOXYURIDINE 5'-MONOPHOSPHATE (three-letter code: UMP) (formula: $C_9H_{13}N_2O_8P$).

UMP

Mol	Chain	Residues	Atoms							ZeroOcc	AltConf
3	A	1	Total	C	H	N	O	P		1	0
			31	9	11	2	8	1			
3	B	1	Total	C	H	N	O	P		1	0
			31	9	11	2	8	1			

- Molecule 4 is 1-[3-[(4-chlorophenyl)-(phenylmethyl)amino]propoxy]-6,6-dimethyl-1,3,5-tri azine-2,4-diamine (three-letter code: E9X) (formula: $C_{21}H_{27}ClN_6O$) (labeled as "Ligand of Interest" by depositor).
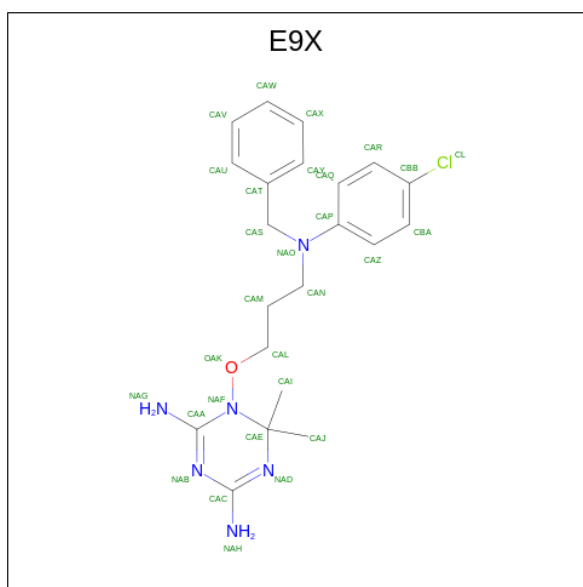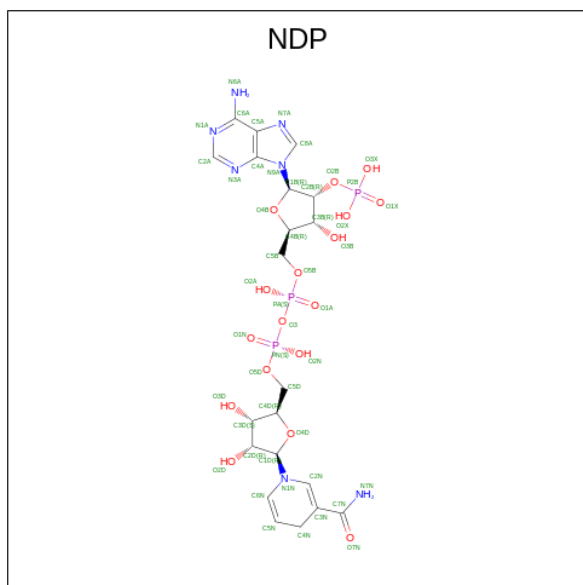
E9X

Mol	Chain	Residues	Atoms						ZeroOcc	AltConf
4	A	1	Total	C	Cl	H	N	O	4	0
			56	21	1	27	6	1		

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Mol	Chain	Residues	Atoms						ZeroOcc	AltConf
			Total	C	Cl	H	N	O		
4	B	1	56	21	1	27	6	1	4	1

- Molecule 5 is NADPH DIHYDRO-NICOTINAMIDE-ADENINE-DINUCLEOTIDE PHOSPHATE (three-letter code: NDP) (formula: $C_{21}H_{30}N_7O_{17}P_3$).

Mol	Chain	Residues	Atoms						ZeroOcc	AltConf
			Total	C	H	N	O	P		
5	A	1	72	21	24	7	17	3	3	0
5	B	1	72	21	24	7	17	3	3	0

- Molecule 6 is water.

| Mol	Chain	Residues	Atoms		ZeroOcc	AltConf
			Total	O		
6	A	47	47	47	0	0
6	B	49	49	49	0	0

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. 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: Bifunctional dihydrofolate reductase-thymidylate synthase
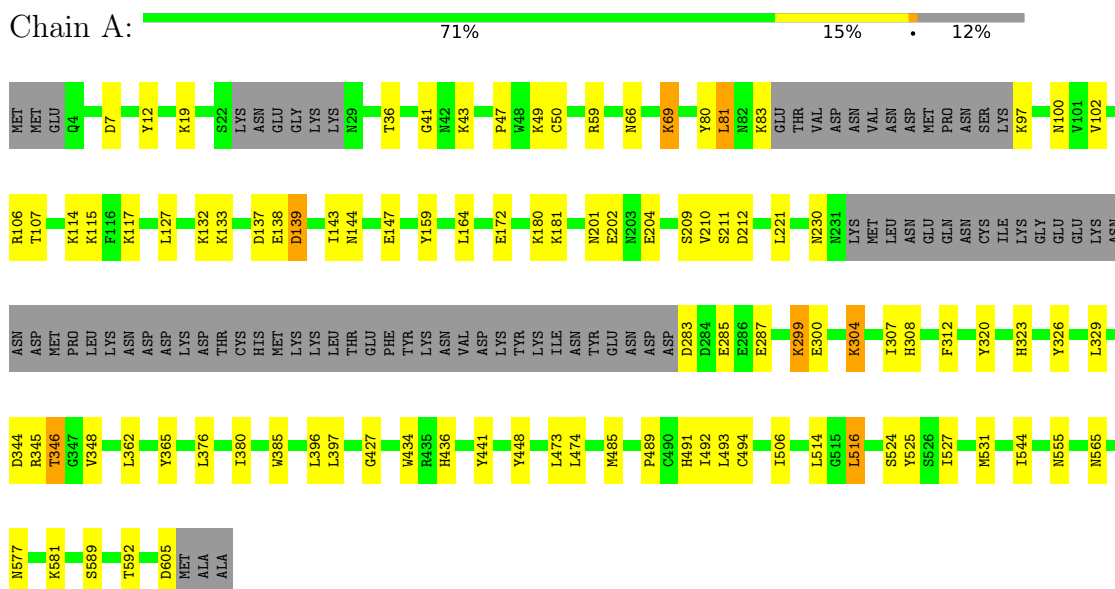
Chain A:

71%   15%   •   12%

- Molecule 1: Bifunctional dihydrofolate reductase-thymidylate synthase

Chain B:

66%   18%   •   13%
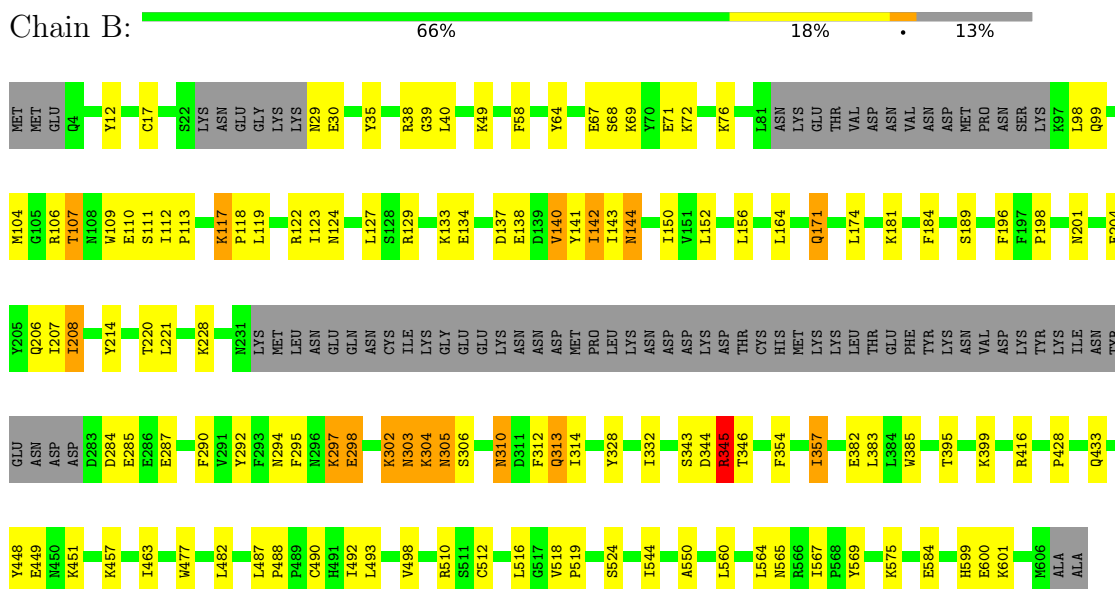
4 Data and refinement statistics ⓘ

Property	Value	Source
Space group	P 21 21 21	Depositor
Cell constants a, b, c, α, β, γ	56.31Å 154.49Å 163.94Å 90.00° 90.00° 90.00°	Depositor
Resolution (Å)	46.41 – 2.59 46.41 – 2.59	Depositor EDS
% Data completeness (in resolution range)	90.1 (46.41-2.59) 84.3 (46.41-2.59)	Depositor EDS
R_{merge}	0.07	Depositor
R_{sym}	(Not available)	Depositor
$< I/\sigma(I) >$ [1]	1.40 (at 2.58Å)	Xtriage
Refinement program	REFMAC 5.8.0258	Depositor
R, R_{free}	0.243 , 0.311 0.193 , 0.252	Depositor DCC
R_{free} test set	2081 reflections (5.08%)	wwPDB-VP
Wilson B-factor (Å^2)	29.6	Xtriage
Anisotropy	0.448	Xtriage
Bulk solvent k_{sol}(e/Å^3), B_{sol}(Å^2)	0.39 , 39.0	EDS
L-test for twinning[2]	$< |L| > = 0.48$, $< L^2 > = 0.31$	Xtriage
Estimated twinning fraction	No twinning to report.	Xtriage
F_o,F_c correlation	0.93	EDS
Total number of atoms	18078	wwPDB-VP
Average B, all atoms (Å^2)	43.0	wwPDB-VP

Xtriage's analysis on translational NCS is as follows: *The largest off-origin peak in the Patterson function is 3.90% 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: NDP, UMP, E9X

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	0.70	0/4537	0.91	2/6128 (0.0%)
1	B	0.72	0/4528	0.89	0/6116
All	All	0.71	0/9065	0.90	2/12244 (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	6
All	All	0	7

There are no bond length outliers.

All (2) bond angle outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(^{o})	Ideal(^{o})
1	A	144	ASN	CB-CA-C	-6.47	97.46	110.40
1	A	525	TYR	CB-CA-C	5.04	120.48	110.40

There are no chirality outliers.

5 of 7 planarity outliers are listed below:

Mol	Chain	Res	Type	Group
1	A	137	ASP	Peptide
1	B	113	PRO	Peptide
1	B	117	LYS	Peptide
1	B	297	LYS	Peptide

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Mol	Chain	Res	Type	Group
1	B	302	LYS	Peptide

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	4433	4399	4384	49	0
1	B	4424	4389	4374	69	0
2	A	8	11	8	1	0
3	A	20	11	11	0	0
3	B	20	11	11	2	0
4	A	29	27	0	2	0
4	B	29	27	0	0	0
5	A	48	24	26	2	0
5	B	48	24	26	4	0
6	A	47	0	0	1	1
6	B	49	0	0	0	1
All	All	9155	8923	8840	115	1

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 6.

The worst 5 of 115 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:605:ASP:C	2:A:701:MET:N	2.05	1.08
1:B:312:PHE:HA	1:B:565:ASN:HD21	1.21	1.00
1:A:312:PHE:HA	1:A:565:ASN:HD21	1.29	0.96
1:B:312:PHE:HA	1:B:565:ASN:ND2	1.94	0.81
1:B:310:ASN:ND2	1:B:310:ASN:H	1.83	0.75

All (1) symmetry-related close contacts are listed below. The label for Atom-2 includes the symmetry operator and encoded unit-cell translations to be applied.

Atom-1	Atom-2	Interatomic distance (Å)	Clash overlap (Å)
6:A:846:HOH:O	6:B:946:HOH:O[1_455]	1.83	0.37

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	524/608 (86%)	492 (94%)	27 (5%)	5 (1%)	15	32
1	B	523/608 (86%)	470 (90%)	43 (8%)	10 (2%)	8	15
All	All	1047/1216 (86%)	962 (92%)	70 (7%)	15 (1%)	11	22

5 of 15 Ramachandran outliers are listed below:

Mol	Chain	Res	Type
1	A	49	LYS
1	A	139	ASP
1	B	49	LYS
1	B	303	ASN
1	B	304	LYS

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	498/570 (87%)	470 (94%)	28 (6%)	21	42
1	B	497/570 (87%)	453 (91%)	44 (9%)	9	19
All	All	995/1140 (87%)	923 (93%)	72 (7%)	14	29

5 of 72 residues with a non-rotameric sidechain are listed below:

Mol	Chain	Res	Type
1	B	310	ASN
1	B	601	LYS
1	B	343	SER
1	B	487	LEU
1	A	555	ASN

Sometimes sidechains can be flipped to improve hydrogen bonding and reduce clashes. 5 of 19 such sidechains are listed below:

Mol	Chain	Res	Type
1	B	313	GLN
1	B	554	ASN
1	B	556	HIS
1	B	424	ASN
1	B	157	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 monosaccharides in this entry.

5.6 Ligand geometry ⓘ

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
5	NDP	A	704	-	45,52,52	0.68	0	53,80,80	0.97	2 (3%)
4	E9X	A	703	-	28,31,31	0.59	0	32,43,43	1.09	2 (6%)
5	NDP	B	803	-	45,52,52	0.63	0	53,80,80	0.79	0
2	MET	A	701	-	6,7,8	0.75	0	2,7,9	0.96	0
3	UMP	B	801	-	21,21,21	0.42	0	31,31,31	0.84	0
3	UMP	A	702	-	21,21,21	0.49	0	31,31,31	0.64	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
5	NDP	A	704	-	-	4/30/77/77	0/5/5/5
4	E9X	A	703	-	-	10/15/34/34	0/3/3/3
5	NDP	B	803	-	-	8/30/77/77	0/5/5/5
2	MET	A	701	-	-	4/5/6/8	-
3	UMP	B	801	-	-	2/10/22/22	0/2/2/2
3	UMP	A	702	-	-	2/10/22/22	0/2/2/2

There are no bond length outliers.

All (4) bond angle outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(^{o})	Ideal(^{o})
4	A	703	E9X	CAT-CAS-NAO	4.34	121.16	114.18
5	A	704	NDP	C5A-C6A-N6A	2.71	124.47	120.35
4	A	703	E9X	CAS-NAO-CAP	2.40	124.89	120.78
5	A	704	NDP	O2D-C2D-C1D	2.04	116.84	110.02

There are no chirality outliers.

5 of 30 torsion outliers are listed below:

Mol	Chain	Res	Type	Atoms
2	A	701	MET	O-C-CA-CB
2	A	701	MET	N-CA-CB-CG
2	A	701	MET	C-CA-CB-CG
3	B	801	UMP	O4'-C4'-C5'-O5'
4	A	703	E9X	OAK-CAL-CAM-CAN

There are no ring outliers.

5 monomers are involved in 11 short contacts:

Mol	Chain	Res	Type	Clashes	Symm-Clashes
5	A	704	NDP	2	0
4	A	703	E9X	2	0
5	B	803	NDP	4	0
2	A	701	MET	1	0
3	B	801	UMP	2	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 NDP A 704

Bond lengths

Bond angles

Torsions

Rings

Ligand E9X A 703

Bond lengths

Bond angles

Torsions

Rings

Ligand NDP B 803

Bond lengths

Bond angles

Torsions

Rings

Ligand UMP B 801

Bond lengths

Bond angles

Torsions

Rings

Ligand UMP A 702

Bond lengths

Bond angles

Torsions

Rings
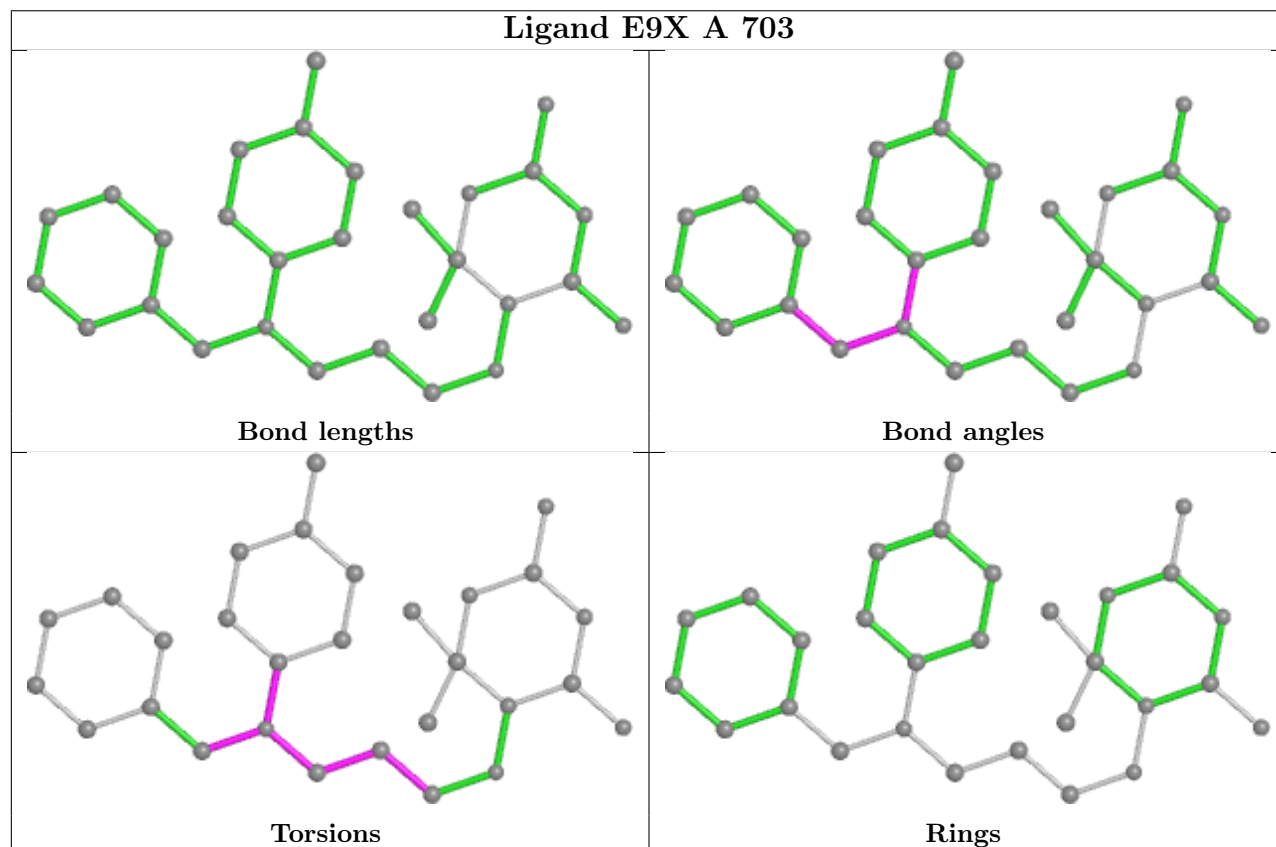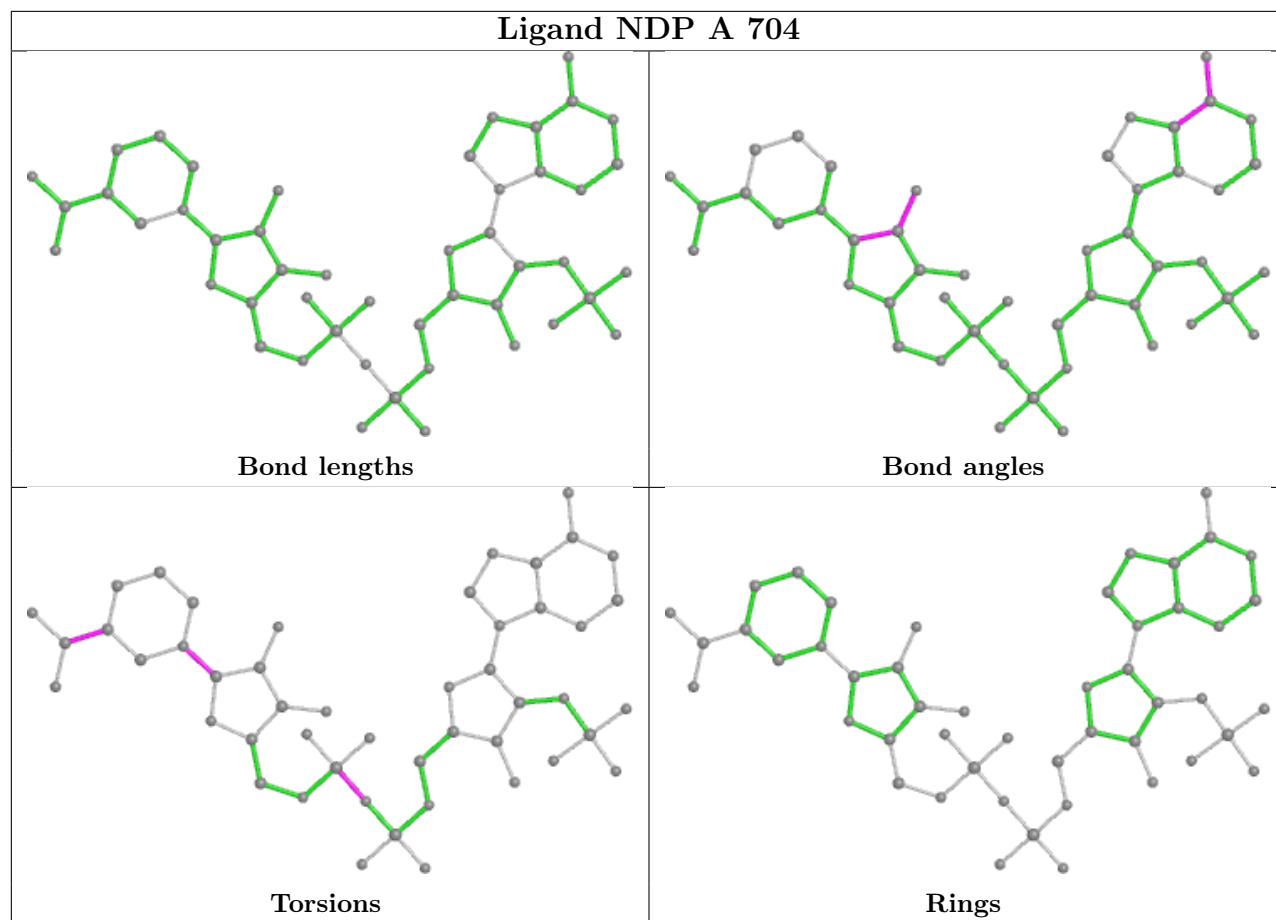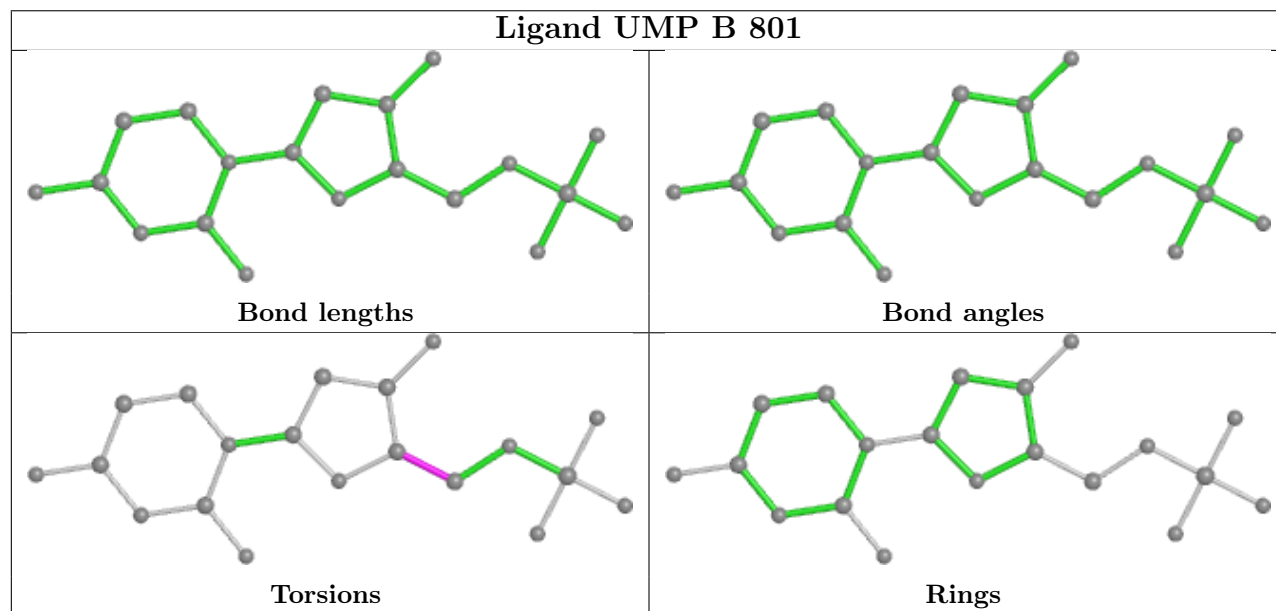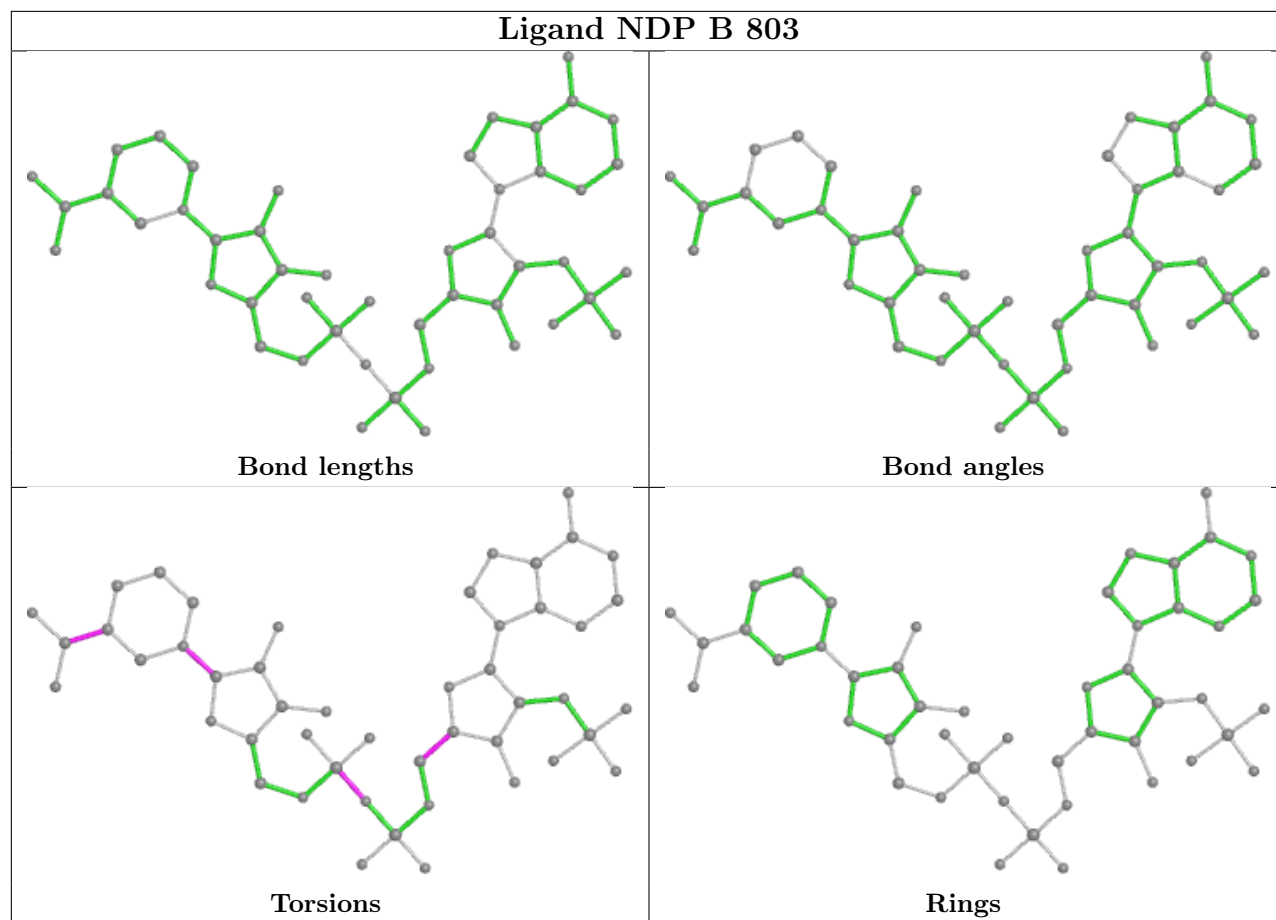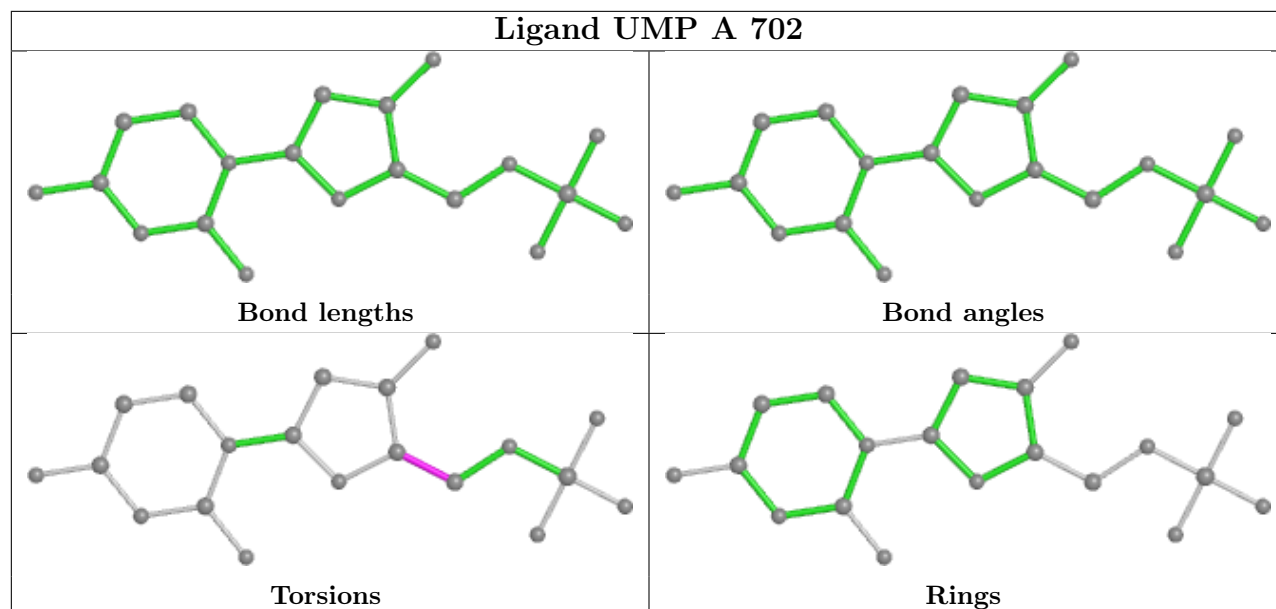
5.7 Other polymers ⓘ

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 ⓘ

Unable to reproduce the depositors R factor - this section is therefore empty.

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

Unable to reproduce the depositors R factor - this section is therefore empty.

6.3 Carbohydrates ⓘ

Unable to reproduce the depositors R factor - this section is therefore empty.

6.4 Ligands ⓘ

Unable to reproduce the depositors R factor - this section is therefore empty.

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 UMP A 702:

$2mF_o$-DF_c (at 0.7 rmsd) in gray
mF_o-DF_c (at 3 rmsd) in purple (negative)
and green (positive)
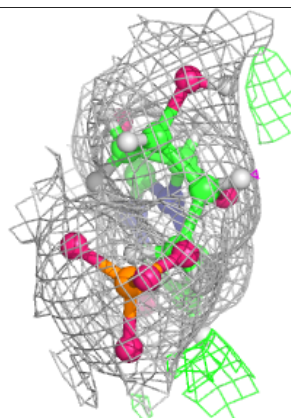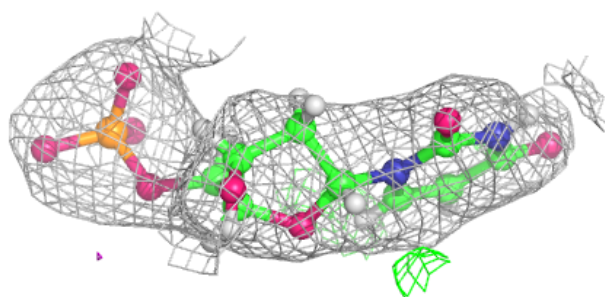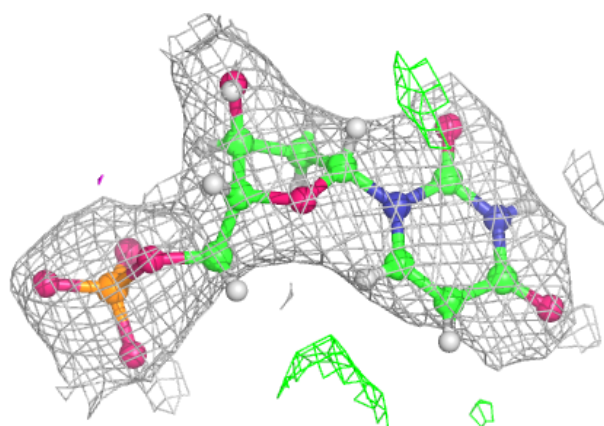
Electron density around UMP B 801:

$2mF_o$-DF_c (at 0.7 rmsd) in gray
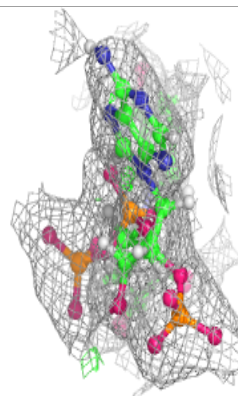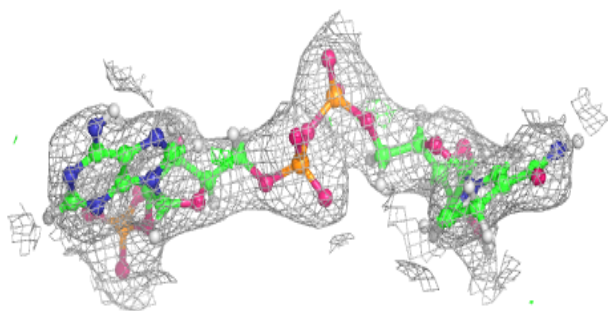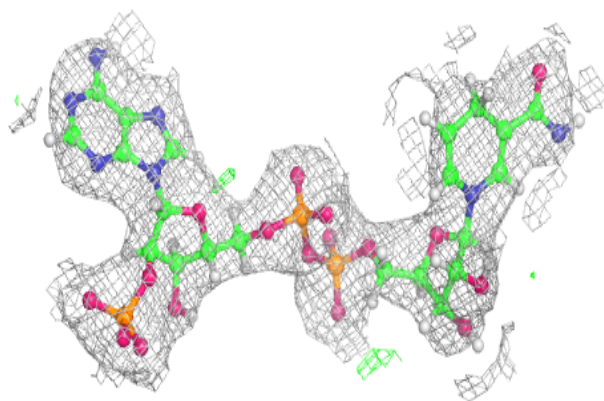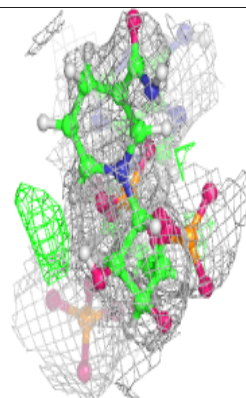mF_o-DF_c (at 3 rmsd) in purple (negative)
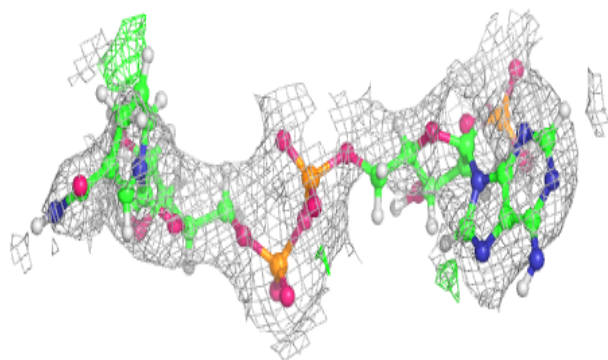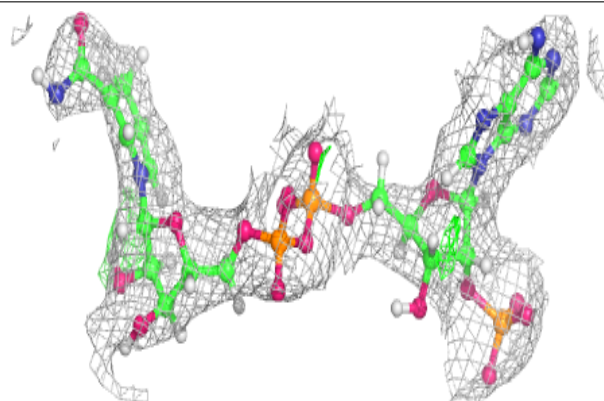and green (positive)
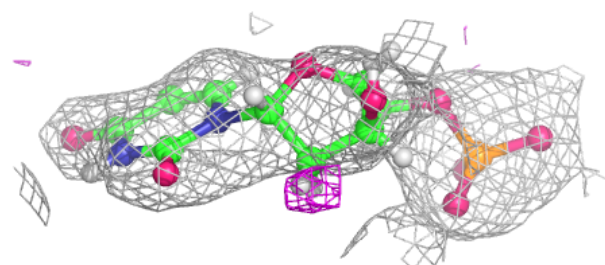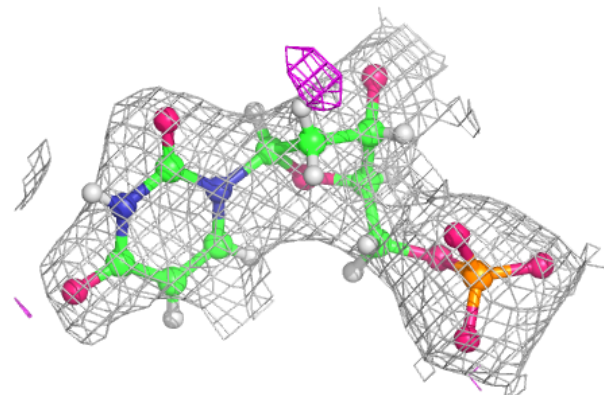
Electron density around E9X A 703:

$2mF_o$-DF_c (at 0.7 rmsd) in gray
mF_o-DF_c (at 3 rmsd) in purple (negative)
and green (positive)
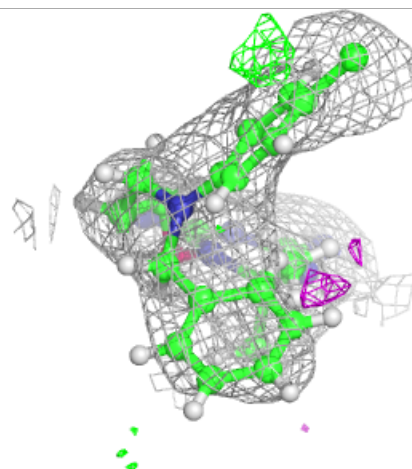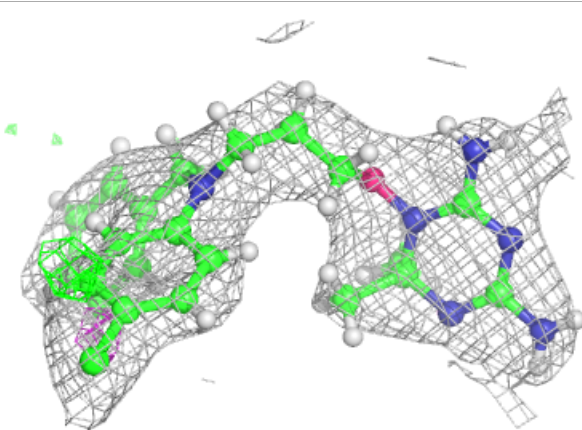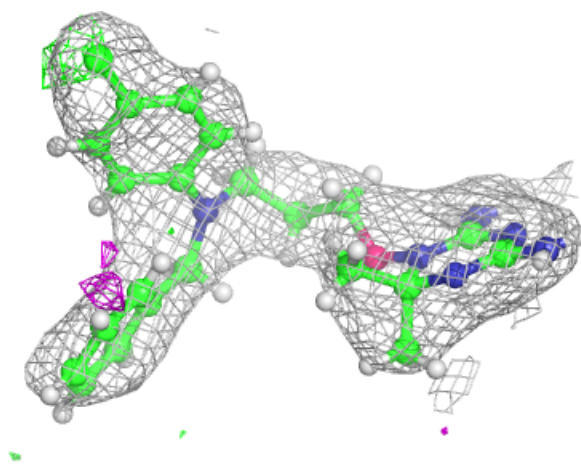
Electron density around NDP A 704:

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

Electron density around NDP B 803:

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

6.5 Other polymers ⓘ

Unable to reproduce the depositors R factor - this section is therefore empty.