Full wwPDB X-ray Structure Validation Report (i)

Sep 10, 2023 – 03:22 PM EDT

PDB ID	:	4KAA
Title	:	Crystal structure of the halotag2 protein at the resolution 2.3A, Northeast Structural Genomics Consortium (NESG) target OR150
Authors	:	Kuzin, A.; Lew, S.; Neklesa, T.K.; Noblin, D.; Seetharaman, J.; Maglaqui, M.; Xiao, R.; Kohan, E.; Wang, H.; Everett, J.K.; Acton, T.B.; Montelione, G.T.; Hunt, J.F.; Crews, C.; Tong, L.; Northeast Structural Genomics Consortium (NESG)
Deposited on	:	2013-04-22
Resolution	:	2.28 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 (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:

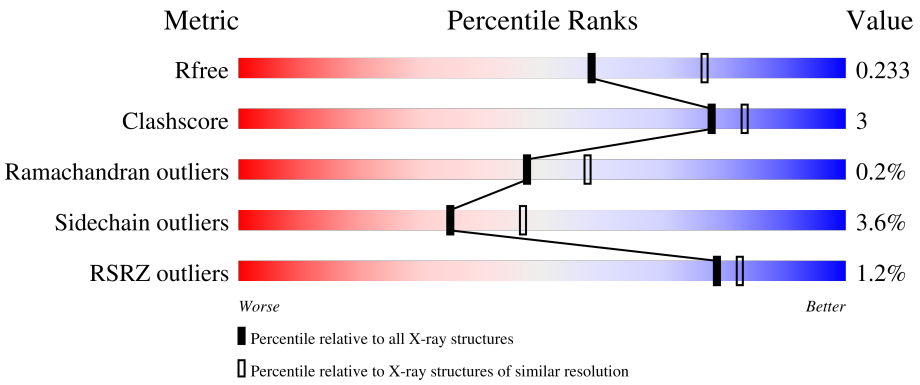
MolProbity	:	4.02b-467
Mogul	:	1.8.5 (274361), CSD as541be (2020)
Xtriage (Phenix)	:	1.13
EDS	:	2.35.1
Percentile statistics	:	20191225.v01 (using entries in the PDB archive December 25th 2019)
Refmac	:	5.8.0158
CCP4	:	7.0.044 (Gargrove)
Ideal geometry (proteins)	:	Engh & Huber (2001)
Ideal geometry (DNA, RNA)	:	Parkinson et al. (1996)

1 Overall quality at a glance (i)

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

The reported resolution of this entry is 2.28 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)	Similar resolution (#Entries, resolution range(Å))
R_{free}	130704	6980 (2.30-2.26)
Clashscore	141614	7711 (2.30-2.26)
Ramachandran outliers	138981	7597 (2.30-2.26)
Sidechain outliers	138945	7598 (2.30-2.26)
RSRZ outliers	127900	6849 (2.30-2.26)

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	306	% 85% 11% •
1	B	306	% 87% 8% • •

Validation Pipeline (wwPDB-VP) : 2.35.1

2 Entry composition (i)

There are 4 unique types of molecules in this entry. The entry contains 5061 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 Haloalkane dehalogenase.

Mol	Chain	Residues	Atoms						ZeroOcc	AltConf	Trace
			Total	C	N	O	S	Se			
1	A	294	2360	1532	399	420	3	6	0	2	0
1	B	293	2371	1539	400	422	3	7	0	4	0

There are 38 discrepancies between the modelled and reference sequences:

Chain	Residue	Modelled	Actual	Comment	Reference
A	1	MSE	-	expression tag	UNP P0A3G3
A	2	GLY	-	expression tag	UNP P0A3G3
A	3	HIS	-	expression tag	UNP P0A3G3
A	4	HIS	-	expression tag	UNP P0A3G3
A	5	HIS	-	expression tag	UNP P0A3G3
A	6	HIS	-	expression tag	UNP P0A3G3
A	7	HIS	-	expression tag	UNP P0A3G3
A	8	HIS	-	expression tag	UNP P0A3G3
A	9	SER	-	expression tag	UNP P0A3G3
A	10	HIS	-	expression tag	UNP P0A3G3
A	11	MSE	-	expression tag	UNP P0A3G3
A	12	GLY	-	expression tag	UNP P0A3G3
A	186	MSE	LYS	engineered mutation	UNP P0A3G3
A	187	GLY	CYS	engineered mutation	UNP P0A3G3
A	283	PHE	HIS	engineered mutation	UNP P0A3G3
A	284	LEU	TYR	engineered mutation	UNP P0A3G3
A	303	GLY	ALA	engineered mutation	UNP P0A3G3
A	305	ALA	-	expression tag	UNP P0A3G3
A	306	GLY	-	expression tag	UNP P0A3G3
B	1	MSE	-	expression tag	UNP P0A3G3
B	2	GLY	-	expression tag	UNP P0A3G3
B	3	HIS	-	expression tag	UNP P0A3G3
B	4	HIS	-	expression tag	UNP P0A3G3
B	5	HIS	-	expression tag	UNP P0A3G3
B	6	HIS	-	expression tag	UNP P0A3G3

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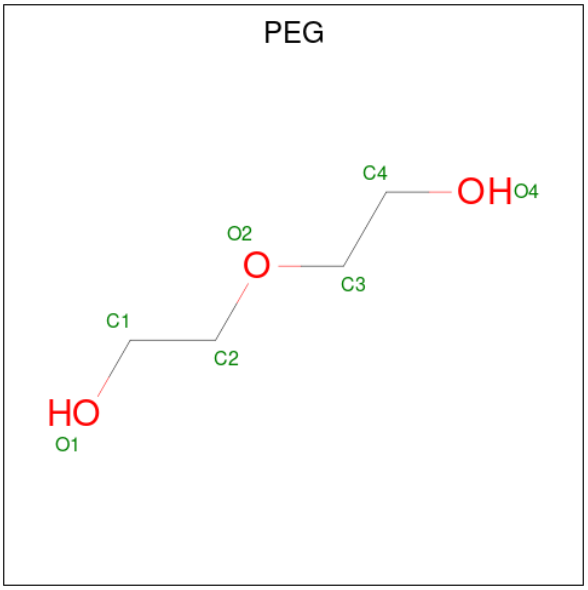
Chain	Residue	Modelled	Actual	Comment	Reference
B	7	HIS	-	expression tag	UNP P0A3G3
B	8	HIS	-	expression tag	UNP P0A3G3
B	9	SER	-	expression tag	UNP P0A3G3
B	10	HIS	-	expression tag	UNP P0A3G3
B	11	MSE	-	expression tag	UNP P0A3G3
B	12	GLY	-	expression tag	UNP P0A3G3
B	186	MSE	LYS	engineered mutation	UNP P0A3G3
B	187	GLY	CYS	engineered mutation	UNP P0A3G3
B	283	PHE	HIS	engineered mutation	UNP P0A3G3
B	284	LEU	TYR	engineered mutation	UNP P0A3G3
B	303	GLY	ALA	engineered mutation	UNP P0A3G3
B	305	ALA	-	expression tag	UNP P0A3G3
B	306	GLY	-	expression tag	UNP P0A3G3

- Molecule 2 is SODIUM ION (three-letter code: NA) (formula: Na).

Mol	Chain	Residues	Atoms	ZeroOcc	AltConf
2	A	1	Total Na 1 1	0	0
2	B	1	Total Na 1 1	0	0

- Molecule 3 is DI(HYDROXYETHYL)ETHER (three-letter code: PEG) (formula: $C_4H_{10}O_3$).



Mol	Chain	Residues	Atoms			ZeroOcc	AltConf
			Total	C	O		
3	B	1	7	4	3	0	0

- Molecule 4 is water.

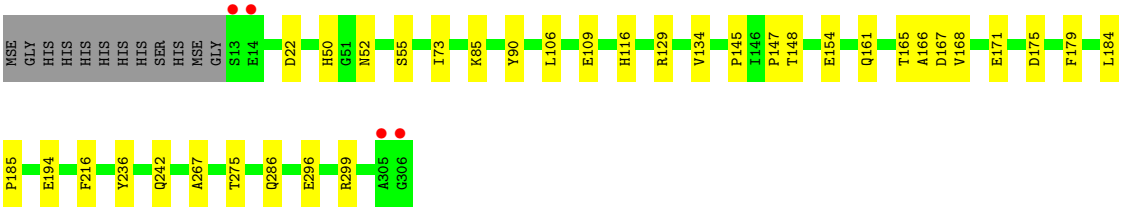
Mol	Chain	Residues	Atoms		ZeroOcc	AltConf
			Total	O		
4	A	190	190	190	0	0
			Total	O		
4	B	130	131	131	0	1

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 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.

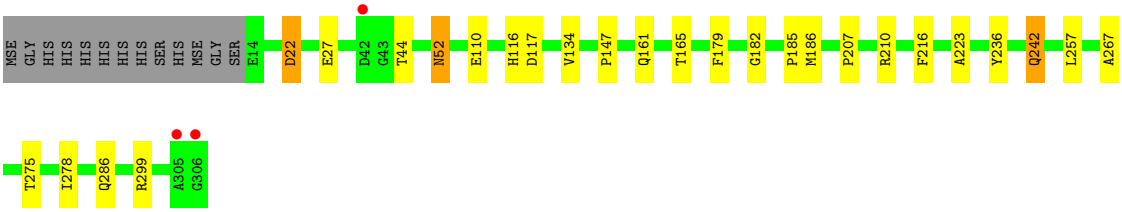
• Molecule 1: Haloalkane dehalogenase

Chain A:



• Molecule 1: Haloalkane dehalogenase

Chain B:



4 Data and refinement statistics (i)

Property	Value	Source
Space group	P 21 21 21	Depositor
Cell constants a, b, c, α, β, γ	69.63Å 91.26Å 98.16Å 90.00° 90.00° 90.00°	Depositor
Resolution (Å)	29.61 – 2.28 29.61 – 2.28	Depositor EDS
% Data completeness (in resolution range)	94.5 (29.61-2.28) 94.6 (29.61-2.28)	Depositor EDS
R_{merge}	0.09	Depositor
R_{sym}	(Not available)	Depositor
$< I/\sigma(I) >$ [1]	9.23 (at 2.29Å)	Xtriage
Refinement program	PHENIX dev_1269	Depositor
R, R_{free}	0.167 , 0.233 0.167 , 0.233	Depositor DCC
R_{free} test set	1394 reflections (5.03%)	wwPDB-VP
Wilson B-factor (Å^2)	21.1	Xtriage
Anisotropy	0.945	Xtriage
Bulk solvent k_{sol}(e/Å^3), B_{sol}(Å^2)	0.30 , 31.5	EDS
L-test for twinning[2]	$< \|L\| > = 0.49$, $< L^2 > = 0.32$	Xtriage
Estimated twinning fraction	No twinning to report.	Xtriage
F_o,F_c correlation	0.95	EDS
Total number of atoms	5061	wwPDB-VP
Average B, all atoms (Å^2)	24.0	wwPDB-VP

Xtriage's analysis on translational NCS is as follows: *The analyses of the Patterson function reveals a significant off-origin peak that is 21.39 % of the origin peak, indicating pseudo-translational symmetry. The chance of finding a peak of this or larger height randomly in a structure without pseudo-translational symmetry is equal to 7.0798e-03. The detected translational NCS is most likely also responsible for the elevated intensity ratio.*

[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 (i)

5.1 Standard geometry (i)

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

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	0.47	2/2436 (0.1%)	0.63	3/3322 (0.1%)
1	B	0.46	2/2447 (0.1%)	0.64	3/3336 (0.1%)
All	All	0.46	4/4883 (0.1%)	0.63	6/6658 (0.1%)

All (4) bond length outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(Å)	Ideal(Å)
1	A	216	PHE	CG-CD1	9.60	1.53	1.38
1	A	216	PHE	CG-CD2	9.32	1.52	1.38
1	B	216	PHE	CG-CD2	9.05	1.52	1.38
1	B	216	PHE	CG-CD1	8.97	1.52	1.38

All (6) bond angle outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(^{o})	Ideal(^{o})
1	B	216	PHE	CB-CG-CD1	-13.67	111.23	120.80
1	A	216	PHE	CB-CG-CD2	-13.62	111.27	120.80
1	A	216	PHE	CB-CG-CD1	-13.34	111.46	120.80
1	B	216	PHE	CB-CG-CD2	-12.39	112.12	120.80
1	B	216	PHE	CD1-CG-CD2	-6.96	109.25	118.30
1	A	216	PHE	CD1-CG-CD2	-6.28	110.14	118.30

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	2360	0	2293	15	0
1	B	2371	0	2301	12	0
2	A	1	0	0	0	0
2	B	1	0	0	0	0
3	B	7	0	10	0	0
4	A	190	0	0	5	0
4	B	131	0	0	0	0
All	All	5061	0	4604	27	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 (27) 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:161:GLN:O	1:B:165:THR:HG23	2.00	0.61
1:A:90:TYR:HB3	4:A:665:HOH:O	2.02	0.59
1:A:161:GLN:O	1:A:165:THR:HG23	2.05	0.56
1:A:129:ARG:NH1	4:A:550:HOH:O	2.40	0.55
1:B:52:ASN:ND2	1:B:117:ASP:OD2	2.41	0.54
1:A:50:HIS:CE1	1:A:55:SER:HA	2.46	0.51
1:B:182:GLY:O	1:B:185:PRO:HD2	2.12	0.50
1:B:22:ASP:OD2	1:B:22:ASP:N	2.44	0.48
1:A:267:ALA:HB2	1:A:275:THR:HG21	1.98	0.46
1:B:147:PRO:O	1:B:223:ALA:HA	2.16	0.45
1:B:44:THR:HG23	1:B:110:GLU:HG2	1.99	0.45
1:A:73:ILE:HD13	1:A:106:LEU:HD23	1.99	0.44
1:A:145:PRO:O	1:A:147:PRO:HD3	2.18	0.43
1:B:52:ASN:HD22	1:B:52:ASN:C	2.22	0.42
1:B:257:LEU:HD23	1:B:257:LEU:HA	1.88	0.42
1:A:171:GLU:O	1:A:175:ASP:HB2	2.20	0.42
1:A:116:HIS:CG	1:A:286:GLN:HE22	2.38	0.41
1:B:116:HIS:CG	1:B:286:GLN:HE22	2.39	0.41
1:B:207:PRO:HA	1:B:210:ARG:HD2	2.03	0.41
1:A:184:LEU:HB3	1:A:185:PRO:HD3	2.02	0.41
1:A:194:GLU:OE1	4:A:579:HOH:O	2.22	0.41
1:A:166:ALA:O	1:A:168:VAL:N	2.46	0.41
1:A:296:GLU:OE2	1:A:299:ARG:NH1	2.54	0.41
1:A:148:THR:HB	4:A:626:HOH:O	2.21	0.40

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Atom-1	Atom-2	Interatomic distance (Å)	Clash overlap (Å)
1:A:85:LYS:HD3	4:A:599:HOH:O	2.22	0.40
1:B:267:ALA:HB2	1:B:275:THR:HG21	2.02	0.40

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 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	294/306 (96%)	284 (97%)	9 (3%)	1 (0%)	41	49
1	B	295/306 (96%)	284 (96%)	11 (4%)	0	100	100
All	All	589/612 (96%)	568 (96%)	20 (3%)	1 (0%)	47	57

All (1) Ramachandran outliers are listed below:

Mol	Chain	Res	Type
1	A	167	ASP

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 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	251/251 (100%)	242 (96%)	9 (4%)	35	47
1	B	252/251 (100%)	239 (95%)	13 (5%)	23	30
All	All	503/502 (100%)	481 (96%)	22 (4%)	35	37

All (22) residues with a non-rotameric sidechain are listed below:

Mol	Chain	Res	Type
1	A	22	ASP
1	A	52	ASN
1	A	109	GLU
1	A	134	VAL
1	A	154	GLU
1	A	179	PHE
1	A	236	TYR
1	A	242[A]	GLN
1	A	242[B]	GLN
1	B	22	ASP
1	B	27	GLU
1	B	52	ASN
1	B	134	VAL
1	B	179	PHE
1	B	186[A]	MSE
1	B	186[B]	MSE
1	B	236	TYR
1	B	242[A]	GLN
1	B	242[B]	GLN
1	B	278	ILE
1	B	299[A]	ARG
1	B	299[B]	ARG

Sometimes sidechains can be flipped to improve hydrogen bonding and reduce clashes. There are no such sidechains identified.

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)

Of 3 ligands modelled in this entry, 2 are monoatomic - leaving 1 for Mogul analysis.

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	PEG	B	401	-	6,6,6	0.61	0	5,5,5	0.51	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	PEG	B	401	-	-	1/4/4/4	-

There are no bond length outliers.

There are no bond angle outliers.

There are no chirality outliers.

All (1) torsion outliers are listed below:

Mol	Chain	Res	Type	Atoms
3	B	401	PEG	C1-C2-O2-C3

There are no ring outliers.

No monomer is involved in short contacts.

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.

6 Fit of model and data (i)

6.1 Protein, DNA and RNA chains (i)

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	288/306 (94%)	-0.52	4 (1%)	75	79	14, 22, 40, 68	0
1	B	287/306 (93%)	-0.40	3 (1%)	82	86	13, 23, 39, 80	0
All	All	575/612 (93%)	-0.46	7 (1%)	79	82	13, 22, 40, 80	0

All (7) RSRZ outliers are listed below:

Mol	Chain	Res	Type	RSRZ
1	B	306	GLY	7.9
1	B	305	ALA	3.4
1	A	13	SER	3.3
1	B	42	ASP	2.7
1	A	306	GLY	2.7
1	A	14	GLU	2.4
1	A	305	ALA	2.1

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

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

6.3 Carbohydrates (i)

There are no monosaccharides in this entry.

6.4 Ligands (i)

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	PEG	B	401	7/7	0.84	0.13	38,40,48,48	0
2	NA	A	401	1/1	0.97	0.15	37,37,37,37	0
2	NA	B	402	1/1	0.98	0.13	27,27,27,27	0

6.5 Other polymers (i)

There are no such residues in this entry.