



Full wwPDB X-ray Structure Validation Report ⓘ

May 23, 2020 – 01:41 am BST

PDB ID : 2VEM
Title : Structure-based enzyme engineering efforts with an inactive monomeric TIM variant: the importance of a single point mutation for generating an active site with suitable binding properties
Authors : Alahuhta, M.; Salin, M.; Casteleijn, M.G.; Kemmer, C.; El-Sayed, I.; Augustyns, K.; Neubauer, P.; Wierenga, R.K.
Deposited on : 2007-10-25
Resolution : 2.20 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 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 : 2.11
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) : 2.11

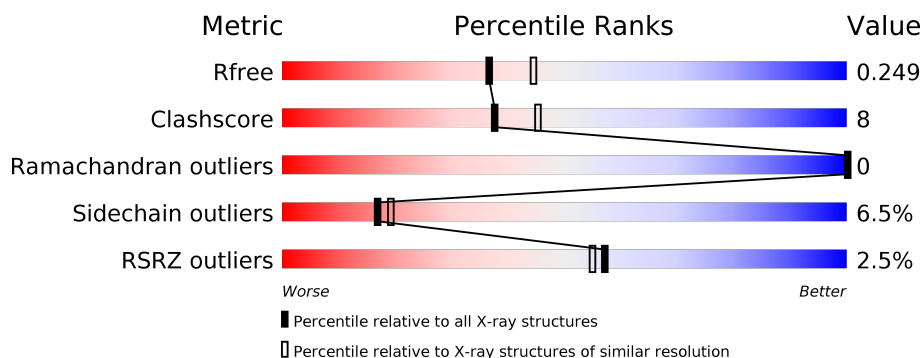
1 Overall quality at a glance

The following experimental techniques were used to determine the structure:

X-RAY DIFFRACTION

The reported resolution of this entry is 2.20 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	4898 (2.20-2.20)
Clashscore	141614	5594 (2.20-2.20)
Ramachandran outliers	138981	5503 (2.20-2.20)
Sidechain outliers	138945	5504 (2.20-2.20)
RSRZ outliers	127900	4800 (2.20-2.20)

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 on 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 $\leq 5\%$. 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	238	<div> <div>3%</div> <div> <div></div> <div>81%</div> <div>15%</div> <div>• •</div> </div> </div>
1	B	238	<div> <div>2%</div> <div> <div></div> <div>84%</div> <div>15%</div> <div>•</div> </div> </div>

The following table lists non-polymeric compounds, carbohydrate monomers and non-standard residues in protein, DNA, RNA chains that are outliers for geometric or electron-density-fit criteria:

Mol	Type	Chain	Res	Chirality	Geometry	Clashes	Electron density
2	BBR	B	1251	-	X	-	-

2 Entry composition

There are 4 unique types of molecules in this entry. The entry contains 3740 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.

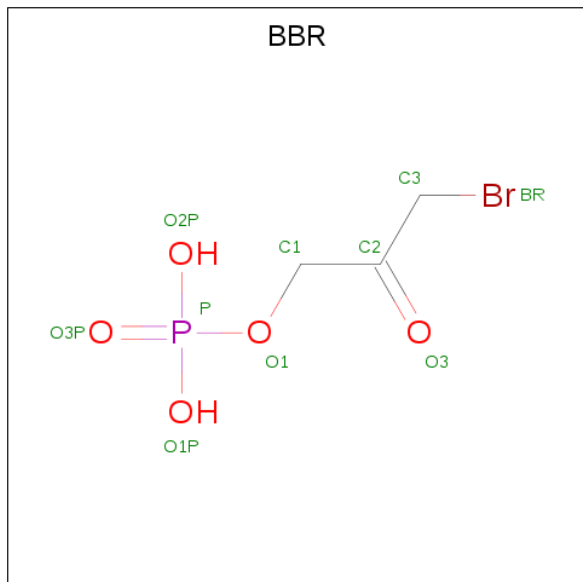
- Molecule 1 is a protein called GLYCOSOMAL TRIOSEPHOSPHATE ISOMERASE.

Mol	Chain	Residues	Atoms					ZeroOcc	AltConf	Trace
1	A	234	Total	C	N	O	S	0	1	0
			1786	1137	314	331	4			
1	B	238	Total	C	N	O	S	0	7	0
			1832	1164	319	345	4			

There are 24 discrepancies between the modelled and reference sequences:

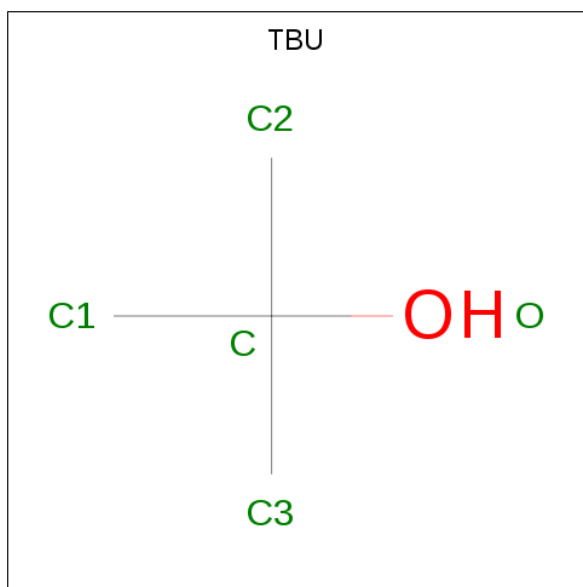
Chain	Residue	Modelled	Actual	Comment	Reference
A	233	ALA	VAL	engineered mutation	UNP P04789
B	233	ALA	VAL	engineered mutation	UNP P04789
A	15	SER	ASN	conflict	UNP P04789
A	18	PRO	GLN	conflict	UNP P04789
A	19	ASP	GLN	conflict	UNP P04789
A	68	GLY	ILE	conflict	UNP P04789
A	69	ASN	ALA	conflict	UNP P04789
A	70	ALA	LYS	conflict	UNP P04789
A	71	ASP	SER	conflict	UNP P04789
A	72	ALA	GLY	conflict	UNP P04789
A	81	ALA	PRO	conflict	UNP P04789
A	82	SER	ILE	conflict	UNP P04789
A	100	TRP	ALA	conflict	UNP P04789
B	15	SER	ASN	conflict	UNP P04789
B	18	PRO	GLN	conflict	UNP P04789
B	19	ASP	GLN	conflict	UNP P04789
B	68	GLY	ILE	conflict	UNP P04789
B	69	ASN	ALA	conflict	UNP P04789
B	70	ALA	LYS	conflict	UNP P04789
B	71	ASP	SER	conflict	UNP P04789
B	72	ALA	GLY	conflict	UNP P04789
B	81	ALA	PRO	conflict	UNP P04789
B	82	SER	ILE	conflict	UNP P04789
B	100	TRP	ALA	conflict	UNP P04789

- Molecule 2 is (3-bromo-2-oxo-propoxy)phosphonic acid (three-letter code: BBR) (formula: $\text{C}_3\text{H}_6\text{BrO}_5\text{P}$).



Mol	Chain	Residues	Atoms				ZeroOcc	AltConf
2	A	1	Total	C	O	P	0	0
			9	3	5	1		
2	B	1	Total	C	O	P	0	0
			9	3	5	1		

- Molecule 3 is TERTIARY-BUTYL ALCOHOL (three-letter code: TBU) (formula: $\text{C}_4\text{H}_{10}\text{O}$).



Mol	Chain	Residues	Atoms			ZeroOcc	AltConf
3	B	1	Total	C	O	0	0
			5	4	1		

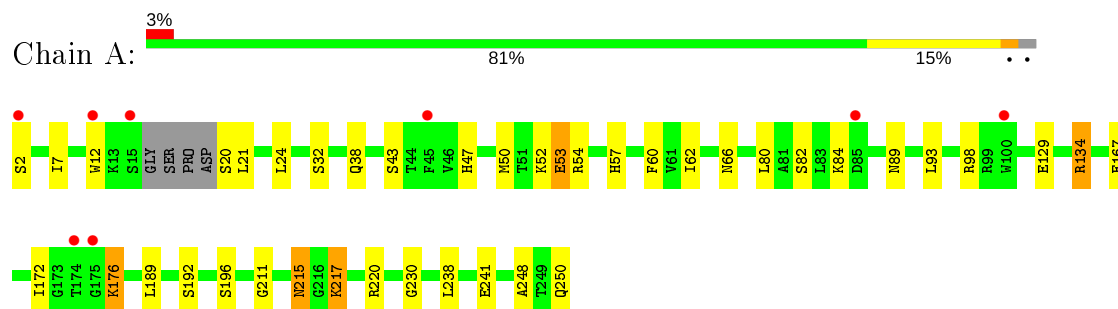
- Molecule 4 is water.

Mol	Chain	Residues	Atoms		ZeroOcc	AltConf
4	A	42	Total	O	0	0
			42	42		
4	B	56	Total	O	0	1
			57	57		

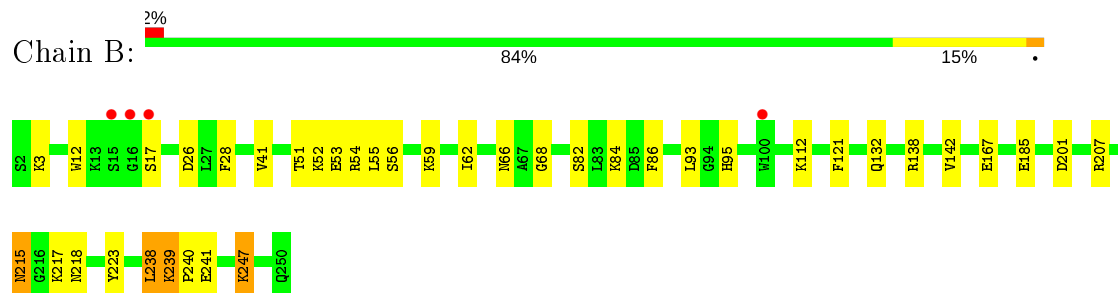
3 Residue-property plots [i](#)

These plots are drawn for all protein, RNA and DNA 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: GLYCOSOMAL TRIOSEPHOSPHATE ISOMERASE



• Molecule 1: GLYCOSOMAL TRIOSEPHOSPHATE ISOMERASE



4 Data and refinement statistics

Property	Value	Source
Space group	P 1 21 1	Depositor
Cell constants a, b, c, α , β , γ	45.80 Å 86.90 Å 56.40 Å 90.00° 97.20° 90.00°	Depositor
Resolution (Å)	19.66 – 2.20 19.66 – 2.20	Depositor EDS
% Data completeness (in resolution range)	100.0 (19.66-2.20) 100.0 (19.66-2.20)	Depositor EDS
R_{merge}	0.14	Depositor
R_{sym}	(Not available)	Depositor
$\langle I/\sigma(I) \rangle$ ¹	3.78 (at 2.19 Å)	Xtriage
Refinement program	REFMAC 5.3.0028	Depositor
R, R_{free}	0.193 , 0.247 0.194 , 0.249	Depositor DCC
R_{free} test set	1114 reflections (5.00%)	wwPDB-VP
Wilson B-factor (Å ²)	22.5	Xtriage
Anisotropy	0.172	Xtriage
Bulk solvent k_{sol} (e/Å ³), B_{sol} (Å ²)	0.34 , 41.4	EDS
L-test for twinning ²	$\langle L \rangle = 0.47$, $\langle L^2 \rangle = 0.29$	Xtriage
Estimated twinning fraction	No twinning to report.	Xtriage
F_o, F_c correlation	0.93	EDS
Total number of atoms	3740	wwPDB-VP
Average B, all atoms (Å ²)	22.0	wwPDB-VP

Xtriage's analysis on translational NCS is as follows: *The largest off-origin peak in the Patterson function is 5.91% of the height of the origin peak. No significant pseudotranslation is detected.*

¹Intensities estimated from amplitudes.

²Theoretical values of $\langle |L| \rangle$, $\langle L^2 \rangle$ 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: TBU, BBR

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	0.61	0/1823	0.67	1/2472 (0.0%)
1	B	0.66	0/1894	0.66	2/2569 (0.1%)
All	All	0.63	0/3717	0.67	3/5041 (0.1%)

There are no bond length outliers.

All (3) bond angle outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(°)	Ideal(°)
1	A	134	ARG	NE-CZ-NH2	-6.07	117.26	120.30
1	B	201[A]	ASP	CB-CG-OD1	5.15	122.93	118.30
1	B	201[B]	ASP	CB-CG-OD1	5.15	122.93	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	1786	0	1809	29	0
1	B	1832	0	1847	28	0
2	A	9	0	2	0	0
2	B	9	0	2	1	0
3	B	5	0	10	2	0
4	A	42	0	0	2	0

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Mol	Chain	Non-H	H(model)	H(added)	Clashes	Symm-Clashes
4	B	57	0	0	2	0
All	All	3740	0	3670	56	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 8.

All (56) 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:12:TRP:HB2	1:B:238:LEU:HD23	1.51	0.93
1:B:59:LYS:HZ2	3:B:1252:TBU:H23	1.51	0.75
1:B:59:LYS:NZ	3:B:1252:TBU:H23	2.03	0.73
1:A:12:TRP:HB2	1:A:238:LEU:HD22	1.75	0.69
1:A:220:ARG:HD2	1:A:248:ALA:HA	1.77	0.67
1:B:26:ASP:OD1	1:B:54:ARG:NH2	2.27	0.66
1:B:84:LYS:HG2	1:B:121:PHE:CE1	2.31	0.65
1:A:167:GLU:HG2	1:A:211:GLY:HA3	1.79	0.64
1:B:66:ASN:HA	1:B:93:LEU:HD23	1.84	0.60
1:A:12:TRP:CZ3	1:A:21:LEU:HD22	2.38	0.59
1:A:215:ASN:ND2	1:A:217:LYS:HB2	2.17	0.58
1:A:220:ARG:NH1	1:A:250:GLN:O	2.36	0.58
1:B:66:ASN:OD1	4:B:2019:HOH:O	2.18	0.56
1:B:185:GLU:HB2	4:B:2044:HOH:O	2.05	0.55
1:A:43[B]:SER:OG	1:A:47:HIS:HB2	2.07	0.55
1:A:53:GLU:HG2	1:A:54:ARG:HG2	1.90	0.54
1:A:57:HIS:HB3	1:A:60:PHE:CD1	2.42	0.54
1:B:247:LYS:NZ	1:B:247:LYS:HB2	2.22	0.54
1:A:215:ASN:HD21	1:A:217:LYS:CB	2.21	0.54
1:B:28:PHE:HE2	1:B:238:LEU:HD11	1.73	0.54
1:A:215:ASN:HD21	1:A:217:LYS:HB2	1.73	0.53
1:B:51:THR:HG22	1:B:62:ILE:HG12	1.89	0.53
1:B:239:LYS:HB2	1:B:240:PRO:HD2	1.90	0.53
1:B:239:LYS:HG3	1:B:241:GLU:HG2	1.90	0.52
1:B:239:LYS:HB2	1:B:240:PRO:CD	2.39	0.52
1:A:38:GLN:NE2	4:A:2010:HOH:O	2.42	0.51
1:A:32:SER:OG	1:B:56:SER:O	2.29	0.49
1:A:52:LYS:HA	1:A:62:ILE:CD1	2.43	0.48
1:B:138:ARG:O	1:B:142:VAL:HG23	2.13	0.48
1:A:66:ASN:HA	1:A:93:LEU:HD23	1.95	0.48
1:A:129:GLU:OE2	1:A:134:ARG:HD2	2.16	0.46
1:A:52:LYS:HA	1:A:62:ILE:HD11	1.98	0.46

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Atom-1	Atom-2	Interatomic distance (Å)	Clash overlap (Å)
1:A:220:ARG:CD	1:A:248:ALA:HA	2.46	0.44
1:B:215:ASN:HD22	1:B:215:ASN:C	2.21	0.44
1:B:52[A]:LYS:HA	1:B:62:ILE:HD13	1.99	0.44
1:A:12:TRP:HB2	1:A:238:LEU:CD2	2.46	0.44
1:B:215:ASN:ND2	1:B:218:ASN:H	2.15	0.44
1:A:215:ASN:ND2	1:A:217:LYS:CB	2.80	0.44
1:B:52[B]:LYS:HA	1:B:62:ILE:HD13	1.99	0.44
1:A:215:ASN:HD22	1:A:217:LYS:N	2.15	0.43
1:A:241:GLU:HG2	4:A:2042:HOH:O	2.18	0.43
1:A:12:TRP:CZ3	1:A:47:HIS:HB3	2.53	0.43
1:B:55:LEU:HD23	1:B:62:ILE:HG13	2.01	0.42
1:A:84:LYS:NZ	1:A:89:ASN:HD22	2.18	0.42
1:B:52[A]:LYS:HD3	1:B:86:PHE:CZ	2.55	0.42
1:A:176:LYS:HA	1:A:176:LYS:HE3	2.02	0.42
1:A:66:ASN:ND2	1:A:98:ARG:HG3	2.35	0.42
1:A:167:GLU:HB3	1:A:172:ILE:HD11	2.02	0.41
1:A:167:GLU:HG2	1:A:211:GLY:CA	2.49	0.41
1:A:7:ILE:O	1:A:230:GLY:HA3	2.20	0.41
1:B:28:PHE:HE2	1:B:238:LEU:CD1	2.33	0.41
1:B:68:GLY:HA3	1:B:112:LYS:HE2	2.02	0.41
1:B:41:VAL:HG13	1:B:238:LEU:HD21	2.02	0.41
1:B:95:HIS:NE2	2:B:1251:BBR:O3	2.42	0.40
1:B:3:LYS:NZ	1:B:223:TYR:O	2.54	0.40
1:B:215:ASN:HD21	1:B:218:ASN:H	1.68	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	231/238 (97%)	226 (98%)	5 (2%)	0	100 100

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Mol	Chain	Analysed	Favoured	Allowed	Outliers	Percentiles	
1	B	243/238 (102%)	238 (98%)	5 (2%)	0	100	100
All	All	474/476 (100%)	464 (98%)	10 (2%)	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	185/187 (99%)	172 (93%)	13 (7%)	15	16
1	B	194/187 (104%)	183 (94%)	11 (6%)	20	24
All	All	379/374 (101%)	355 (94%)	24 (6%)	17	20

All (24) residues with a non-rotameric sidechain are listed below:

Mol	Chain	Res	Type
1	A	2	SER
1	A	20	SER
1	A	24	LEU
1	A	50	MET
1	A	53	GLU
1	A	80	LEU
1	A	82	SER
1	A	176	LYS
1	A	189	LEU
1	A	192	SER
1	A	196	SER
1	A	215	ASN
1	A	217	LYS
1	B	17	SER
1	B	53	GLU
1	B	82	SER
1	B	132	GLN
1	B	167	GLU

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Mol	Chain	Res	Type
1	B	207	ARG
1	B	215	ASN
1	B	217	LYS
1	B	238	LEU
1	B	239	LYS
1	B	247	LYS

Some sidechains can be flipped to improve hydrogen bonding and reduce clashes. All (9) such sidechains are listed below:

Mol	Chain	Res	Type
1	A	38	GLN
1	A	89	ASN
1	A	215	ASN
1	B	65	GLN
1	B	89	ASN
1	B	132	GLN
1	B	215	ASN
1	B	224	GLN
1	B	250	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 carbohydrates in this entry.

5.6 Ligand geometry [i](#)

3 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	BBR	A	1251	1	8,8,9	1.96	1 (12%)	9,11,12	1.92	3 (33%)
3	TBU	B	1252	-	4,4,4	0.58	0	6,6,6	0.70	0
2	BBR	B	1251	1	8,8,9	1.94	1 (12%)	9,11,12	1.92	4 (44%)

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	BBR	A	1251	1	-	0/6/6/8	-
2	BBR	B	1251	1	-	5/6/6/8	-

All (2) bond length outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(Å)	Ideal(Å)
2	A	1251	BBR	O3-C2	5.28	1.42	1.21
2	B	1251	BBR	O3-C2	5.06	1.41	1.21

All (7) bond angle outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(°)	Ideal(°)
2	A	1251	BBR	O3-C2-C3	-3.72	111.94	121.40
2	B	1251	BBR	O2P-P-O1P	3.10	119.47	107.64
2	A	1251	BBR	C3-C2-C1	3.06	126.19	116.88
2	B	1251	BBR	O3-C2-C3	-2.83	114.22	121.40
2	B	1251	BBR	C3-C2-C1	2.69	125.06	116.88
2	B	1251	BBR	O1P-P-O1	-2.37	100.42	106.73
2	A	1251	BBR	O2P-P-O1P	2.12	115.74	107.64

There are no chirality outliers.

All (5) torsion outliers are listed below:

Mol	Chain	Res	Type	Atoms
2	B	1251	BBR	C1-O1-P-O1P
2	B	1251	BBR	C1-O1-P-O2P

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Mol	Chain	Res	Type	Atoms
2	B	1251	BBR	C1-O1-P-O3P
2	B	1251	BBR	O1-C1-C2-O3
2	B	1251	BBR	O1-C1-C2-C3

There are no ring outliers.

2 monomers are involved in 3 short contacts:

Mol	Chain	Res	Type	Clashes	Symm-Clashes
3	B	1252	TBU	2	0
2	B	1251	BBR	1	0

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	234/238 (98%)	-0.09	8 (3%) 45 43	14, 21, 33, 43	0
1	B	238/238 (100%)	-0.22	4 (1%) 70 68	14, 21, 32, 42	0
All	All	472/476 (99%)	-0.16	12 (2%) 57 55	14, 21, 33, 43	0

All (12) RSRZ outliers are listed below:

Mol	Chain	Res	Type	RSRZ
1	A	15	SER	3.8
1	B	17	SER	3.6
1	B	16	GLY	3.3
1	A	2	SER	3.3
1	A	12	TRP	2.9
1	A	175	GLY	2.7
1	A	45	PHE	2.4
1	B	15	SER	2.3
1	B	100	TRP	2.3
1	A	100	TRP	2.3
1	A	174	THR	2.1
1	A	85	ASP	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 carbohydrates 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(\AA^2)	Q<0.9
2	BBR	A	1251	9/10	0.84	0.25	40,45,51,52	0
3	TBU	B	1252	5/5	0.91	0.17	25,26,26,27	0
2	BBR	B	1251	9/10	0.96	0.09	20,22,26,27	0

6.5 Other polymers [i](#)

There are no such residues in this entry.