

wwPDB X-ray Structure Validation Summary Report (i)

Jun 5, 2023 – 01:43 pm BST

PDB ID	:	8A2C
Title	:	The crystal structure of the S178A mutant of PET40, a PETase enzyme from
		an unclassified Amycolatopsis
Authors	:	Costanzi, E.; Applegate, V.; Port, A.; Smits, S.H.J.
Deposited on		
Resolution	:	1.60 Å(reported)

This is a wwPDB X-ray Structure 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/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 (1)) were used in the production of this report:

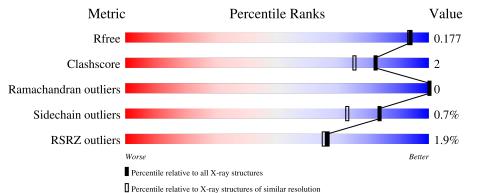
MolProbity	:	4.02b-467
Mogul	:	1.8.4, CSD as 541 be (2020)
Xtriage (Phenix)	:	1.13
EDS	:	2.33
Percentile statistics	:	20191225.v01 (using entries in the PDB archive December 25th 2019)
Refmac	:	5.8.0158
CCP4	:	7.0.044 (Gargrove)
Ideal geometry (proteins)	:	Engh & Huber (2001)
Ideal geometry (DNA, RNA)	:	Parkinson et al. (1996)
Validation Pipeline (wwPDB-VP)	:	2.33

1 Overall quality at a glance (i)

The following experimental techniques were used to determine the structure: $X\text{-}RAY \, DIFFRACTION$

The reported resolution of this entry is 1.60 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	$egin{array}{c} { m Whole \ archive} \ (\#{ m Entries}) \end{array}$	${f Similar\ resolution}\ (\#{ m Entries,\ resolution\ range}({ m \AA}))$
R_{free}	130704	3398 (1.60-1.60)
Clashscore	141614	3665 (1.60-1.60)
Ramachandran outliers	138981	3564 (1.60-1.60)
Sidechain outliers	138945	3563 (1.60-1.60)
RSRZ outliers	127900	3321 (1.60-1.60)

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% 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	А	278	90%	6%	·				
1	В	278	^{2%} 92%	•	•				



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2 Entry composition (i)

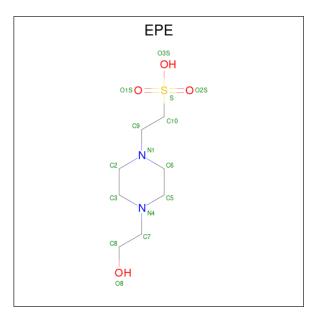
There are 6 unique types of molecules in this entry. The entry contains 8773 atoms, of which 4028 are hydrogens and 0 are deuteriums.

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 PET40 S178A.

Mol	Chain	Residues	Atoms					ZeroOcc	AltConf	Trace	
1	А	268	Total	С	Н	N	0	S	0	7	0
			4039	1282	1994	364	396	3	_		
1	В	267	Total	\mathbf{C}	Η	Ν	Ο	\mathbf{S}	0	1	0
1	D	201	3992	1268	1970	360	391	3	0	4	0

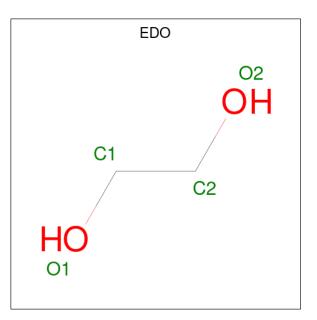
• Molecule 2 is 4-(2-HYDROXYETHYL)-1-PIPERAZINE ETHANESULFONIC ACID (three-letter code: EPE) (formula: C₈H₁₈N₂O₄S).



Mol	Chain	Residues	Atoms				ZeroOcc	AltConf		
9	Δ	1	Total	С	Η	Ν	0	\mathbf{S}	0	0
	Л	1	32	8	17	2	4	1	0	0
9	В	1	Total	С	Η	Ν	0	\mathbf{S}	0	0
	D	1	32	8	17	2	4	1	0	U

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





Mol	Chain	Residues	Atoms	ZeroOcc	AltConf
3	А	1	Total C H O	0	Ο
0	11	I	10 2 6 2	0	0
3	А	1	Total C H O	0	0
0	11	I	10 2 6 2	0	0
3	А	1	Total C H O	0	0
0	Л	T	10 2 6 2	0	
3	А	1	Total C H O	0	0
0	Л	T	10 2 6 2	0	0
3	В	1	Total C H O	0	0
0	D	1	10 2 6 2	0	0

• Molecule 4 is CHLORIDE ION (three-letter code: CL) (formula: Cl).

Mol	Chain	Residues	Atoms	ZeroOcc	AltConf
4	А	7	Total Cl 7 7	0	0
4	В	3	Total Cl 3 3	0	0

• Molecule 5 is MAGNESIUM ION (three-letter code: MG) (formula: Mg).

N	Mol	Chain	Residues	Atoms	ZeroOcc	AltConf
	5	А	1	Total Mg 1 1	0	0
	5	В	1	Total Mg 1 1	0	0



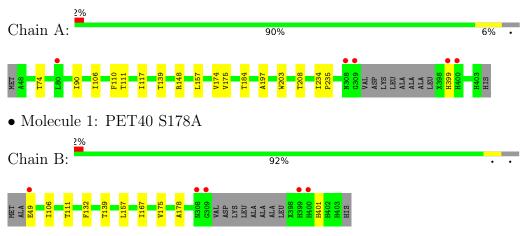
• Molecule 6 is water.

Mol	Chain	Residues	Atoms	ZeroOcc	AltConf
6	А	323	Total O 323 323	0	0
6	В	293	Total O 293 293	0	0



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: PET40 S178A



4 Data and refinement statistics (i)

Property	Value	Source
Space group	P 1 21 1	Depositor
Cell constants	46.04Å 110.00Å 47.96Å	Depositor
a, b, c, α , β , γ	90.00° 95.74° 90.00°	Depositor
Resolution (Å)	31.51 - 1.60	Depositor
Resolution (A)	36.05 - 1.60	EDS
% Data completeness	97.6(31.51-1.60)	Depositor
(in resolution range)	97.7 (36.05 - 1.60)	EDS
R _{merge}	0.07	Depositor
R _{sym}	(Not available)	Depositor
$< I/\sigma(I) > 1$	$1.76 (at 1.60 \text{\AA})$	Xtriage
Refinement program	PHENIX 1.19.2_4158, PHENIX 1.19.2_4158	Depositor
R, R_{free}	0.146 , 0.179	Depositor
10, 10 free	0.145 , 0.177	DCC
R_{free} test set	3066 reflections $(5.03%)$	wwPDB-VP
Wilson B-factor $(Å^2)$	15.9	Xtriage
Anisotropy	0.475	Xtriage
Bulk solvent $k_{sol}(e/A^3), B_{sol}(A^2)$	0.41, 52.9	EDS
L-test for $twinning^2$	$< L >=0.49, < L^2>=0.33$	Xtriage
Estimated twinning fraction	0.029 for l,-k,h	Xtriage
F_o, F_c correlation	0.98	EDS
Total number of atoms	8773	wwPDB-VP
Average B, all atoms $(Å^2)$	20.0	wwPDB-VP

Xtriage's analysis on translational NCS is as follows: The largest off-origin peak in the Patterson function is 6.20% of the height of the origin peak. No significant pseudotranslation is detected.

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.



¹Intensities estimated from amplitudes.

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: MG, EPE, CL, 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		
	Unam	RMSZ	# Z > 5	RMSZ	# Z > 5	
1	А	0.54	0/2106	0.69	0/2877	
1	В	0.50	0/2082	0.67	0/2844	
All	All	0.52	0/4188	0.68	0/5721	

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	А	2045	1994	1976	15	0
1	В	2022	1970	1960	5	0
2	А	15	17	17	1	0
2	В	15	17	17	1	0
3	А	16	24	24	0	0
3	В	4	6	6	0	0
4	А	7	0	0	0	0
4	В	3	0	0	0	0
5	А	1	0	0	0	0
5	В	1	0	0	0	0
6	А	323	0	0	2	0

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Mol	Chain	Non-H	H(model)	H(added)	Clashes	Symm-Clashes
6	В	293	0	0	1	0
All	All	4745	4028	4000	20	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 2.

The worst 5 of 20 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:174:VAL:HG23	1:A:184[A]:THR:HG23	1.49	0.90
1:A:184[A]:THR:HG22	1:A:197:ALA:HB1	1.91	0.52
1:A:106:ILE:HG22	1:A:117:ILE:HD12	1.93	0.51
1:A:148:ARG:HH11	1:A:148:ARG:HG2	1.75	0.51
1:A:184[A]:THR:CG2	1:A:197:ALA:HB1	2.40	0.51

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	Percer	ntiles
1	А	271/278~(98%)	267~(98%)	4(2%)	0	100	100
1	В	267/278~(96%)	262 (98%)	5(2%)	0	100	100
All	All	538/556~(97%)	529~(98%)	9~(2%)	0	100	100

There are no Ramachandran outliers to report.

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	А	224/225~(100%)	222~(99%)	2(1%)	78 65		
1	В	222/225~(99%)	220 (99%)	2 (1%)	78 65		
All	All	446/450~(99%)	442 (99%)	4 (1%)	84 65		

All (4) residues with a non-rotameric sidechain are listed below:

Mol	Chain	Res	Type
1	А	157[A]	LEU
1	А	157[B]	LEU
1	В	132	PHE
1	В	401	HIS

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 19 ligands modelled in this entry, 12 are monoatomic - leaving 7 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



Mol	Type	Chain	Res	Link	Bo	ond leng	ths	Bond angles		
	Type	Ullaili	nes		Counts	RMSZ	# Z >2	Counts	RMSZ	# Z > 2
3	EDO	А	604	-	$3,\!3,\!3$	0.53	0	2,2,2	0.30	0
3	EDO	А	603	-	3,3,3	0.37	0	2,2,2	0.70	0
2	EPE	В	502	-	$15,\!15,\!15$	2.05	2 (13%)	18,20,20	2.15	<mark>6 (33%)</mark>
3	EDO	А	602	-	3,3,3	0.46	0	2,2,2	0.32	0
3	EDO	А	605	-	3,3,3	0.35	0	2,2,2	0.23	0
3	EDO	В	501	-	3,3,3	0.40	0	2,2,2	0.27	0
2	EPE	А	601	-	$15,\!15,\!15$	2.48	4 (26%)	18,20,20	2.20	<mark>6 (33%)</mark>

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).

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	А	604	-	-	0/1/1/1	-
3	EDO	А	603	-	-	1/1/1/1	-
2	EPE	В	502	-	-	8/9/19/19	0/1/1/1
3	EDO	А	602	-	-	0/1/1/1	-
3	EDO	А	605	-	-	0/1/1/1	-
3	EDO	В	501	-	-	0/1/1/1	-
2	EPE	А	601	-	-	5/9/19/19	0/1/1/1

The worst 5 of 6 bond length outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(Å)	Ideal(Å)
2	А	601	EPE	C10-S	-6.57	1.68	1.77
2	В	502	EPE	C10-S	-6.46	1.68	1.77
2	А	601	EPE	O1S-S	-4.57	1.31	1.45
2	А	601	EPE	O2S-S	-3.75	1.34	1.45
2	В	502	EPE	O3S-S	-2.88	1.37	1.47

The worst 5 of 12 bond angle outliers are listed below:

Mol	Chain	Res	Type	Atoms	Ζ	$\mathbf{Observed}(^{o})$	$Ideal(^{o})$
2	В	502	EPE	O3S-S-C10	5.69	114.97	105.77
2	А	601	EPE	O1S-S-C10	5.23	113.21	106.92
2	А	601	EPE	C2-C3-N4	4.03	118.91	110.64
2	А	601	EPE	C9-N1-C2	3.77	120.87	111.23

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Mol	Chain	Res	Type	Atoms	Z	$Observed(^{o})$	$Ideal(^{o})$
2	А	601	EPE	C6-N1-C2	-3.36	101.27	108.83

There are no chirality outliers.

5 of 14 torsion outliers are listed below:

Mol	Chain	Res	Type	Atoms
2	А	601	EPE	C10-C9-N1-C2
2	А	601	EPE	C9-C10-S-O2S
2	В	502	EPE	C10-C9-N1-C2
2	В	502	EPE	C8-C7-N4-C5
2	В	502	EPE	C9-C10-S-O1S

There are no ring outliers.

2 monomers are involved in 2 short contacts:

Mol	Chain	Res	Type	Clashes	Symm-Clashes
2	В	502	EPE	1	0
2	А	601	EPE	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, 95^{th} 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	$\langle RSRZ \rangle$	#RSRZ>2	$OWAB(Å^2)$	Q<0.9
1	А	267/278~(96%)	-0.44	5 (1%) 66 65	10, 15, 30, 79	0
1	В	266/278~(95%)	-0.51	5 (1%) 66 65	11, 15, 30, 72	0
All	All	533/556~(95%)	-0.47	10 (1%) 66 65	10, 15, 31, 79	0

The worst 5 of 10 RSRZ outliers are listed below:

Mol	Chain	Res	Type	RSRZ
1	А	309	GLY	13.3
1	А	308	ASN	6.1
1	В	309	GLY	5.9
1	В	308	ASN	4.1
1	В	399	HIS	3.9

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, 95^{th} 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.



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Mol	Type	Chain	Res	Atoms	RSCC	RSR	B-factors(Å ²)	Q<0.9
3	EDO	А	604	4/4	0.77	0.13	$25,\!31,\!42,\!50$	0
3	EDO	А	603	4/4	0.84	0.15	33,40,52,53	0
3	EDO	А	602	4/4	0.86	0.21	34,43,52,57	0
3	EDO	А	605	4/4	0.94	0.12	20,26,32,32	0
5	MG	А	613	1/1	0.94	0.15	37,37,37,37	0
3	EDO	В	501	4/4	0.96	0.10	19,24,29,29	0
4	CL	В	505	1/1	0.96	0.04	34,34,34,34	0
2	EPE	В	502	15/15	0.96	0.23	15,49,73,78	0
5	MG	В	506	1/1	0.96	0.16	32,32,32,32	0
4	CL	А	608	1/1	0.97	0.04	26,26,26,26	0
2	EPE	А	601	15/15	0.97	0.09	13,37,47,50	0
4	CL	А	610	1/1	0.98	0.07	33,33,33,33	0
4	CL	А	612	1/1	0.98	0.05	22,22,22,22	0
4	CL	В	504	1/1	0.98	0.05	24,24,24,24	0
4	CL	А	607	1/1	0.99	0.04	$25,\!25,\!25,\!25$	0
4	CL	А	611	1/1	0.99	0.04	20,20,20,20	0
4	CL	А	606	1/1	0.99	0.07	19,19,19,19	0
4	CL	В	503	1/1	0.99	0.09	$15,\!15,\!15,\!15$	0
4	CL	А	609	1/1	1.00	0.04	13,13,13,13	0

6.5 Other polymers (i)

There are no such residues in this entry.

