wwPDB X-ray Structure Validation Summary Report (i)

Sep 13, 2023 – 11:16 AM EDT

PDB ID	:	4RI0
Title	:	Serine Protease HtrA3, mutationally inactivated
Authors	:	Osipiuk, J.; Glaza, P.; Wenta, T.; Lipinska, B.; Joachimiak, A.; Midwest Center for Structural Genomics (MCSG)
Deposited on	:	2014-10-03
Resolution	:	3.27 Å(reported)

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

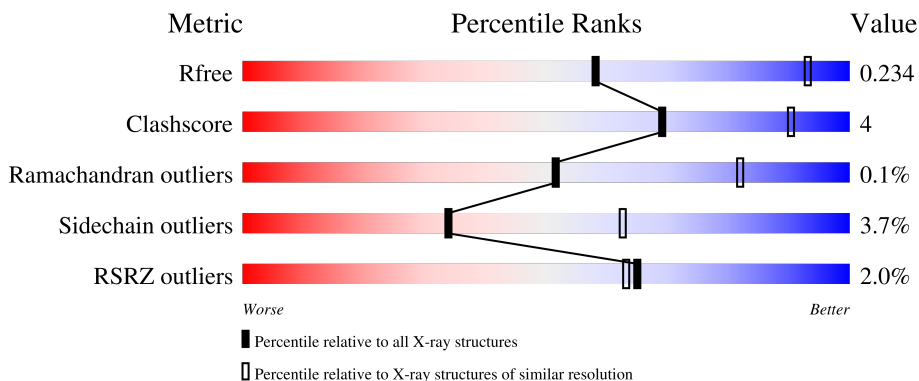|		
MolProbity	:	4.02b-467
Mogul	:	1.8.5 (274361), CSD as541be (2020)
Xtriage (Phenix)	:	1.13
EDS	:	2.35.1
Percentile statistics	:	20191225.v01 (using entries in the PDB archive December 25th 2019)
Refmac	:	5.8.0158
CCP4	:	7.0.044 (Gargrove)
Ideal geometry (proteins)	:	Engh & Huber (2001)
Ideal geometry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

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 3.27 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		0.234
Clashscore		4
Ramachandran outliers		0.1%
Sidechain outliers		3.7%
RSRZ outliers		2.0%

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	1177 (3.32-3.24)
Clashscore	141614	1044 (3.30-3.26)
Ramachandran outliers	138981	1026 (3.30-3.26)
Sidechain outliers	138945	1025 (3.30-3.26)
RSRZ outliers	127900	1141 (3.32-3.24)

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	332	% 79% 13% · 8%
1	B	332	2% 54% 7% 39%
1	C	332	% 52% 7% 41%

2 Entry composition ⓘ

There are 3 unique types of molecules in this entry. The entry contains 5380 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 Serine protease HTRA3.

Mol	Chain	Residues	Atoms					ZeroOcc	AltConf	Trace
1	A	306	Total 2343	C 1487	N 405	O 447	S 4	0	0	0
1	B	202	Total 1535	C 984	N 264	O 286	S 1	0	0	0
1	C	196	Total 1489	C 954	N 255	O 278	S 2	0	0	0

There are 27 discrepancies between the modelled and reference sequences:

Chain	Residue	Modelled	Actual	Comment	Reference
A	305	ALA	SER	engineered mutation	UNP P83110
A	454	GLY	-	expression tag	UNP P83110
A	455	GLY	-	expression tag	UNP P83110
A	456	HIS	-	expression tag	UNP P83110
A	457	HIS	-	expression tag	UNP P83110
A	458	HIS	-	expression tag	UNP P83110
A	459	HIS	-	expression tag	UNP P83110
A	460	HIS	-	expression tag	UNP P83110
A	461	HIS	-	expression tag	UNP P83110
B	305	ALA	SER	engineered mutation	UNP P83110
B	454	GLY	-	expression tag	UNP P83110
B	455	GLY	-	expression tag	UNP P83110
B	456	HIS	-	expression tag	UNP P83110
B	457	HIS	-	expression tag	UNP P83110
B	458	HIS	-	expression tag	UNP P83110
B	459	HIS	-	expression tag	UNP P83110
B	460	HIS	-	expression tag	UNP P83110
B	461	HIS	-	expression tag	UNP P83110
C	305	ALA	SER	engineered mutation	UNP P83110
C	454	GLY	-	expression tag	UNP P83110
C	455	GLY	-	expression tag	UNP P83110
C	456	HIS	-	expression tag	UNP P83110
C	457	HIS	-	expression tag	UNP P83110

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Chain	Residue	Modelled	Actual	Comment	Reference
C	458	HIS	-	expression tag	UNP P83110
C	459	HIS	-	expression tag	UNP P83110
C	460	HIS	-	expression tag	UNP P83110
C	461	HIS	-	expression tag	UNP P83110

- Molecule 2 is PHOSPHATE ION (three-letter code: PO4) (formula: O_4P).
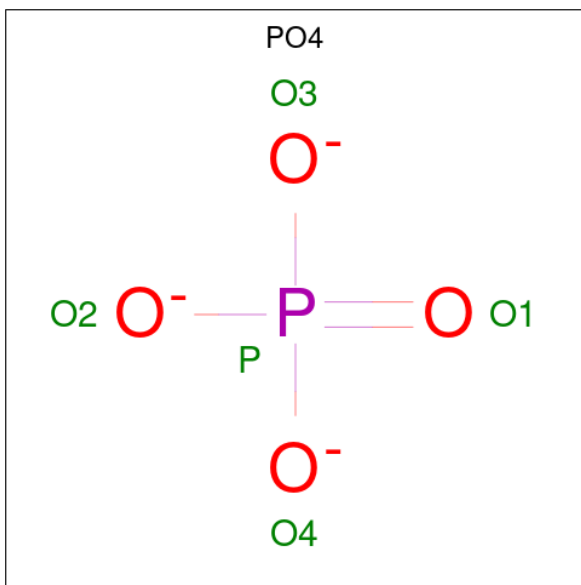
Mol	Chain	Residues	Atoms			ZeroOcc	AltConf
2	A	1	Total 5	O 4	P 1	0	0
2	A	1	Total 5	O 4	P 1	0	0

- Molecule 3 is water.

| Mol	Chain	Residues	Atoms		ZeroOcc	AltConf
3	A	2	Total 2	O 2	0	0
3	B	1	Total 1	O 1	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 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.
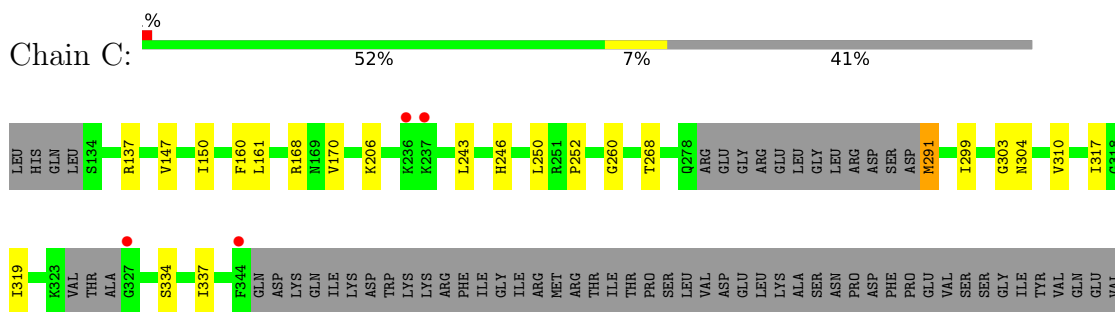
• Molecule 1: Serine protease HTRA3
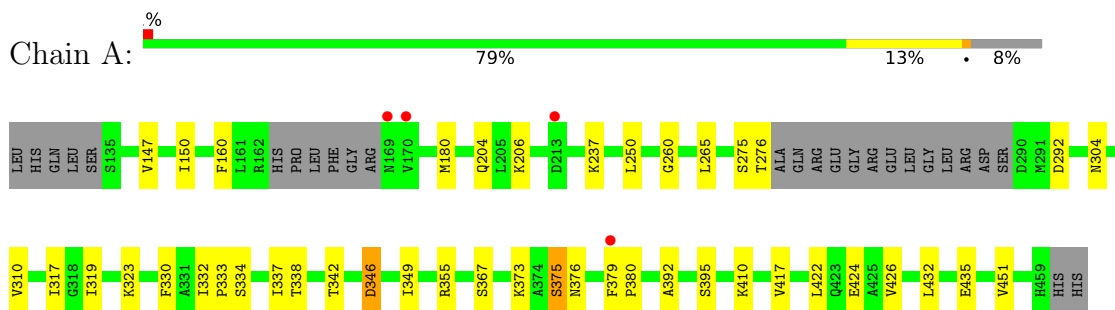
• Molecule 1: Serine protease HTRA3
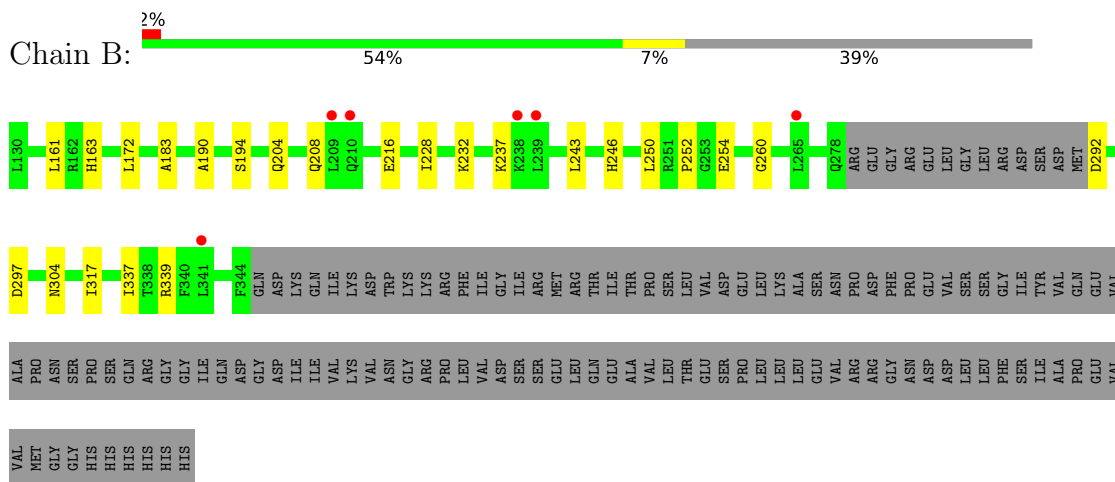
• Molecule 1: Serine protease HTRA3

ALA PRO ASN SER PRO SER GLN ARG GLY GLY ILE GLN ASP GLY ASP ILE ILE VAL LYS VAL ASN GLY ARG PRO LEU VAL ASP SER GLU GLN GLN ALA VAL LEU THR GLU SER PRO LEU LEU GLU VAL ARG ARG GLY ASN ASP LEU LEU PHE SER ILE ALA PRO GLU VAL

VAL MET GLY GLY HIS HIS HIS HIS HIS HIS

4 Data and refinement statistics ⓘ

Property	Value	Source
Space group	P 43 21 2	Depositor
Cell constants a, b, c, α, β, γ	118.98Å 118.98Å 167.86Å 90.00° 90.00° 90.00°	Depositor
Resolution (Å)	40.76 – 3.27 40.76 – 3.27	Depositor EDS
% Data completeness (in resolution range)	98.4 (40.76-3.27) 98.4 (40.76-3.27)	Depositor EDS
R_{merge}	0.09	Depositor
R_{sym}	(Not available)	Depositor
$< I/\sigma(I) >$ [1]	2.61 (at 3.25Å)	Xtriage
Refinement program	PHENIX 1.8.2_1309	Depositor
R, R_{free}	0.199 , 0.233 0.200 , 0.234	Depositor DCC
R_{free} test set	935 reflections (4.94%)	wwPDB-VP
Wilson B-factor (Å²)	109.4	Xtriage
Anisotropy	0.142	Xtriage
Bulk solvent k_{sol}(e/Å³), B_{sol}(Å²)	0.32 , 65.9	EDS
L-test for twinning[2]	$< |L| > = 0.50, < L^2 > = 0.34$	Xtriage
Estimated twinning fraction	No twinning to report.	Xtriage
F_o,F_c correlation	0.95	EDS
Total number of atoms	5380	wwPDB-VP
Average B, all atoms (Å²)	112.0	wwPDB-VP

Xtriage's analysis on translational NCS is as follows: *The largest off-origin peak in the Patterson function is 2.56% 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: PO4

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	0.23	0/2385	0.44	0/3235
1	B	0.24	0/1564	0.45	0/2124
1	C	0.24	0/1516	0.47	0/2055
All	All	0.24	0/5465	0.45	0/7414

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	2343	0	2379	19	0
1	B	1535	0	1573	12	0
1	C	1489	0	1523	13	0
2	A	10	0	0	0	0
3	A	2	0	0	0	0
3	B	1	0	0	0	0
All	All	5380	0	5475	40	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 4.

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The worst 5 of 40 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:346:ASP:OD1	1:A:346:ASP:N	2.30	0.63
1:A:260:GLY:HA3	1:A:304:ASN:HA	1.81	0.61
1:A:392:ALA:HB3	1:A:395:SER:HB3	1.88	0.56
1:A:319:ILE:HG13	1:A:337:ILE:HD11	1.90	0.54
1:C:319:ILE:HG13	1:C:337:ILE:HD11	1.91	0.53

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	300/332 (90%)	288 (96%)	11 (4%)	1 (0%)	41	72
1	B	198/332 (60%)	191 (96%)	7 (4%)	0	100	100
1	C	190/332 (57%)	181 (95%)	9 (5%)	0	100	100
All	All	688/996 (69%)	660 (96%)	27 (4%)	1 (0%)	51	82

All (1) Ramachandran outliers are listed below:

Mol	Chain	Res	Type
1	A	375	SER

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	262/284 (92%)	250 (95%)	12 (5%)	27	58
1	B	169/284 (60%)	163 (96%)	6 (4%)	35	63
1	C	164/284 (58%)	160 (98%)	4 (2%)	49	73
All	All	595/852 (70%)	573 (96%)	22 (4%)	34	62

5 of 22 residues with a non-rotameric sidechain are listed below:

Mol	Chain	Res	Type
1	B	243	LEU
1	B	339	ARG
1	B	292	ASP
1	C	168	ARG
1	A	346	ASP

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

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 ⓘ

2 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	PO4	A	502	-	4,4,4	0.95	0	6,6,6	0.46	0
2	PO4	A	501	-	4,4,4	0.92	0	6,6,6	0.44	0

There are no bond length outliers.

There are no bond angle outliers.

There are no chirality outliers.

There are no torsion outliers.

There are no ring outliers.

No monomer is involved in short contacts.

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.

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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	306/332 (92%)	0.18	4 (1%)	77	76	71, 101, 153, 188	0
1	B	202/332 (60%)	0.22	6 (2%)	50	49	73, 107, 145, 185	0
1	C	196/332 (59%)	0.23	4 (2%)	65	63	81, 114, 161, 232	0
All	All	704/996 (70%)	0.21	14 (1%)	65	63	71, 106, 156, 232	0

The worst 5 of 14 RSRZ outliers are listed below:

Mol	Chain	Res	Type	RSRZ
1	C	236	LYS	4.2
1	C	237	LYS	3.6
1	C	327	GLY	3.5
1	A	170	VAL	2.9
1	B	239	LEU	2.8

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
2	PO4	A	501	5/5	0.77	0.26	190,190,190,190	0
2	PO4	A	502	5/5	0.96	0.17	102,102,102,102	0

6.5   Other polymers ⓘ

There are no such residues in this entry.