

wwPDB X-ray Structure Validation Summary Report (i)

Mar 25, 2024 - 02:20 PM EDT

PDB ID	:	8UP6
Title	:	Structure of atypical asparaginase from Rhodospirillum rubrum (mutant
		K19A) in complex with L-Asp
Authors	:	Lubkowski, J.; Wlodawer, A.; Zhang, D.
Deposited on		
Resolution	:	1.70 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:

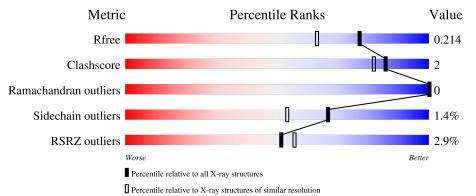
MolProbity	:	4.02b-467
Mogul	:	1.8.5 (274361), CSD as541be (2020)
Xtriage (Phenix)	:	1.13
EDS	:	2.36.1
Percentile statistics	:	20191225.v01 (using entries in the PDB archive December 25th 2019)
Refmac	:	5.8.0158
CCP4	:	7.0.044 (Gargrove)
Ideal geometry (proteins)	:	Engh & Huber (2001)
Ideal geometry (DNA, RNA)	:	Parkinson et al. (1996)
Validation Pipeline (wwPDB-VP)	:	2.36.1

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 1.70 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	4298 (1.70-1.70)
Clashscore	141614	4695 (1.70-1.70)
Ramachandran outliers	138981	4610 (1.70-1.70)
Sidechain outliers	138945	4610 (1.70-1.70)
RSRZ outliers	127900	4222 (1.70-1.70)

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	185	5%	5% • 8%
1	В	185	2% 8 3%	5% 12%
1	С	185	85%	•• 11%
1	D	185	<u>4%</u> 86%	5% 9%



2 Entry composition (i)

There are 4 unique types of molecules in this entry. The entry contains 5340 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.

Mol	Chain	Residues	Atoms			ZeroOcc	AltConf	Trace		
1	А	170	Total	С	Ν	0	S	0	1	0
	A	170	1248	788	219	235	6	0		
1	В	163	Total	С	Ν	0	S	0	1	0
	I D	105	1202	761	212	224	5			
1	С	164	Total	С	Ν	0	S	0	1	0
	U		1208	765	213	225	5	0	1	0
1	1 D	D 160	Total	С	Ν	0	S	0	1	0
	169	1241	785	218	232	6	0	1	0	

• Molecule 1 is a protein called Asparaginase.

There are 56 discrepancies between the modelled and reference sequences:

Chain	Residue	Modelled	Actual	Comment	Reference
А	-12	HIS	-	expression tag	UNP Q2RMX1
А	-11	HIS	-	expression tag	UNP Q2RMX1
А	-10	HIS	-	expression tag	UNP Q2RMX1
А	-9	HIS	-	expression tag	UNP Q2RMX1
А	-8	HIS	-	expression tag	UNP Q2RMX1
А	-7	HIS	-	expression tag	UNP Q2RMX1
А	-6	GLU	-	expression tag	UNP Q2RMX1
А	-5	ASN	-	expression tag	UNP Q2RMX1
А	-4	LEU	-	expression tag	UNP Q2RMX1
А	-3	TYR	-	expression tag	UNP Q2RMX1
A	-2	PHE	-	expression tag	UNP Q2RMX1
A	-1	GLN	-	expression tag	UNP Q2RMX1
А	0	SER	-	expression tag	UNP Q2RMX1
A	19	ALA	LYS	engineered mutation	UNP Q2RMX1
В	-12	HIS	-	expression tag	UNP Q2RMX1
В	-11	HIS	-	expression tag	UNP Q2RMX1
В	-10	HIS	-	expression tag	UNP Q2RMX1
В	-9	HIS	-	expression tag	UNP Q2RMX1
В	-8	HIS	-	expression tag	UNP Q2RMX1
В	-7	HIS	-	expression tag	UNP Q2RMX1
В	-6	GLU	-	expression tag	UNP Q2RMX1

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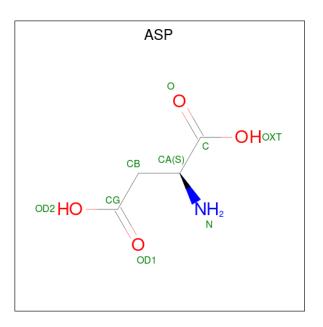


Chain	Residue	Modelled	Actual	Comment	Reference
В	-5	ASN	-	expression tag	UNP Q2RMX1
В	-4	LEU	-	expression tag	UNP Q2RMX1
В	-3	TYR	-	expression tag	UNP Q2RMX1
В	-2	PHE	-	expression tag	UNP Q2RMX1
В	-1	GLN	-	expression tag	UNP Q2RMX1
В	0	SER	-	expression tag	UNP Q2RMX1
В	19	ALA	LYS	engineered mutation	UNP Q2RMX1
С	-12	HIS	-	expression tag	UNP Q2RMX1
С	-11	HIS	-	expression tag	UNP Q2RMX1
С	-10	HIS	-	expression tag	UNP Q2RMX1
С	-9	HIS	-	expression tag	UNP Q2RMX1
С	-8	HIS	-	expression tag	UNP Q2RMX1
С	-7	HIS	-	expression tag	UNP Q2RMX1
С	-6	GLU	-	expression tag	UNP Q2RMX1
С	-5	ASN	-	expression tag	UNP Q2RMX1
С	-4	LEU	-	expression tag	UNP Q2RMX1
С	-3	TYR	-	expression tag	UNP Q2RMX1
С	-2	PHE	-	expression tag	UNP Q2RMX1
С	-1	GLN	-	expression tag	UNP Q2RMX1
С	0	SER	-	expression tag	UNP Q2RMX1
С	19	ALA	LYS	engineered mutation	UNP Q2RMX1
D	-12	HIS	-	expression tag	UNP Q2RMX1
D	-11	HIS	-	expression tag	UNP Q2RMX1
D	-10	HIS	-	expression tag	UNP Q2RMX1
D	-9	HIS	-	expression tag	UNP Q2RMX1
D	-8	HIS	-	expression tag	UNP Q2RMX1
D	-7	HIS	-	expression tag	UNP Q2RMX1
D	-6	GLU	-	expression tag	UNP Q2RMX1
D	-5	ASN	-	expression tag	UNP Q2RMX1
D	-4	LEU	-	expression tag	UNP Q2RMX1
D	-3	TYR	-	expression tag	UNP Q2RMX1
D	-2	PHE	-	expression tag	UNP Q2RMX1
D	-1	GLN	-	expression tag	UNP Q2RMX1
D	0	SER	-	expression tag	UNP Q2RMX1
D	19	ALA	LYS	engineered mutation	UNP Q2RMX1

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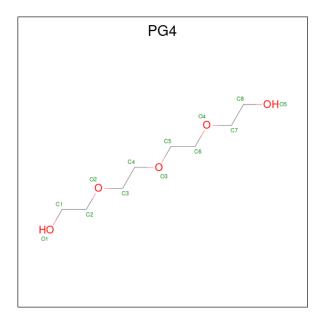
• Molecule 2 is ASPARTIC ACID (three-letter code: ASP) (formula: $C_4H_7NO_4$).





Mol	Chain	Residues	Atoms				ZeroOcc	AltConf
2	В	1	Total 9	С 4		0 4	0	0
2	С	1	Total 9	С 4	N 1	0 4	0	0

• Molecule 3 is TETRAETHYLENE GLYCOL (three-letter code: PG4) (formula: $C_8H_{18}O_5$).



Mol	Chain	Residues	Atoms	ZeroOcc	AltConf
3	В	1	Total C O 13 8 5	0	0
3	В	1	Total C O 13 8 5	0	0

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Mol	Chain	Residues	Atoms			ZeroOcc	AltConf
3	D	1	Total 13	C 8	O 5	0	0

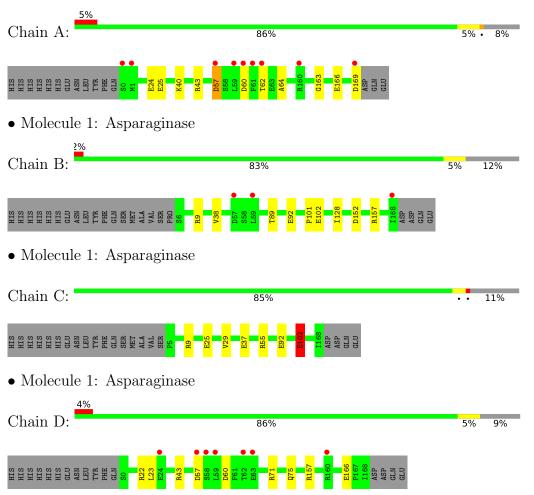
• Molecule 4 is water.

Mol	Chain	Residues	Atoms	ZeroOcc	AltConf
4	А	86	Total O 86 86	0	0
4	В	101	Total O 101 101	0	0
4	С	91	Total O 91 91	0	0
4	D	106	Total O 106 106	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: Asparaginase



4 Data and refinement statistics (i)

Property	Value	Source
Space group	P 21 21 21	Depositor
Cell constants	72.05Å 77.33Å 115.03Å	Depositor
a, b, c, α , β , γ	90.00° 90.00° 90.00°	Depositor
Resolution (Å)	38.89 - 1.70	Depositor
	38.86 - 1.70	EDS
% Data completeness	99.3 (38.89-1.70)	Depositor
(in resolution range)	99.3 (38.86 - 1.70)	EDS
R_{merge}	0.07	Depositor
R _{sym}	(Not available)	Depositor
$< I/\sigma(I) > 1$	$3.65 (at 1.70 \text{\AA})$	Xtriage
Refinement program	REFMAC 5.8.0258	Depositor
R, R_{free}	0.181 , 0.205	Depositor
It, Itfree	0.193 , 0.214	DCC
R_{free} test set	1431 reflections (2.02%)	wwPDB-VP
Wilson B-factor ($Å^2$)	13.2	Xtriage
Anisotropy	0.074	Xtriage
Bulk solvent $k_{sol}(e/Å^3)$, $B_{sol}(Å^2)$	0.37, 45.8	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	5340	wwPDB-VP
Average B, all atoms $(Å^2)$	19.0	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 52.44 % 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 4.8622e-05. The detected translational NCS is most likely also responsible for the elevated intensity ratio.

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: PG4

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	Bo	nd lengths	Bo	ond angles
IVIOI	Chain	RMSZ	# Z > 5	RMSZ	# Z > 5
1	А	0.75	0/1272	0.95	2/1723~(0.1%)
1	В	0.82	0/1224	0.90	0/1657
1	С	0.80	2/1232~(0.2%)	0.93	3/1668~(0.2%)
1	D	0.75	0/1264	0.94	1/1712~(0.1%)
All	All	0.78	2/4992~(0.0%)	0.93	6/6760~(0.1%)

All (2) bond length outliers are listed below:

Mol	Chain	Res	Type	Atoms	Ζ	$\operatorname{Observed}(\operatorname{\AA})$	Ideal(Å)
1	С	102	GLU	CD-OE1	5.54	1.31	1.25
1	С	92	GLU	CD-OE2	5.46	1.31	1.25

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

Mol	Chain	Res	Type	Atoms	Ζ	$\mathbf{Observed}(^{o})$	$\mathbf{Ideal}(^{o})$
1	А	43	ARG	NE-CZ-NH2	-6.31	117.14	120.30
1	А	43	ARG	NE-CZ-NH1	5.87	123.23	120.30
1	С	9	ARG	NE-CZ-NH2	-5.44	117.58	120.30
1	D	43	ARG	NE-CZ-NH2	-5.37	117.62	120.30
1	С	55	ARG	NE-CZ-NH2	-5.31	117.65	120.30

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



Mol	Chain	Non-H	H(model)	H(added)	Clashes	Symm-Clashes
1	А	1248	0	1269	4	0
1	В	1202	0	1226	7	0
1	С	1208	0	1230	3	0
1	D	1241	0	1269	4	0
2	В	9	0	3	0	0
2	С	9	0	3	0	0
3	В	26	0	36	2	0
3	D	13	0	18	3	0
4	А	86	0	0	1	0
4	В	101	0	0	2	0
4	С	91	0	0	2	0
4	D	106	0	0	3	0
All	All	5340	0	5054	22	0

the asymmetric unit, whereas Symm-Clashes lists symmetry-related clashes.

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 22 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:163:GLY:HA3	1:D:23:LEU:HD23	1.69	0.74	
1:D:71:ARG:O	1:D:75:GLN:HG3	1.88	0.72	
3:D:201:PG4:C2	4:D:391:HOH:O	2.42	0.68	
1:C:37:GLU:OE2	4:C:301:HOH:O	2.13	0.66	
1:A:24:GLU:HG3	4:A:242:HOH:O	1.97	0.64	

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	Perce	ntiles
1	А	169/185~(91%)	169 (100%)	0	0	100	100
1	В	162/185~(88%)	162 (100%)	0	0	100	100
1	\mathbf{C}	163/185~(88%)	162 (99%)	1 (1%)	0	100	100
1	D	168/185~(91%)	167~(99%)	1 (1%)	0	100	100
All	All	662/740~(90%)	660 (100%)	2~(0%)	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	127/141~(90%)	122~(96%)	5(4%)	32 13		
1	В	121/141~(86%)	121 (100%)	0	100 100		
1	С	122/141~(86%)	121~(99%)	1 (1%)	81 74		
1	D	126/141~(89%)	125~(99%)	1 (1%)	81 74		
All	All	496/564~(88%)	489~(99%)	7(1%)	67 53		

5 of 7 residues with a non-rotameric sidechain are listed below:

Mol	Chain	Res	Type
1	А	166	GLU
1	А	169	ASP
1	D	22	ARG
1	С	102	GLU
1	А	57	ASP

Sometimes side chains can be flipped to improve hydrogen bonding and reduce clashes. There are no such side chains 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)

5 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).

Mal	Mol Type		Res	Link	Bo	Bond lengths			Bond angles		
	туре	Chain	nes		Counts	RMSZ	# Z > 2	Counts	RMSZ	# Z > 2	
2	ASP	В	201	-	6,8,8	1.24	1 (16%)	8,10,10	1.73	2 (25%)	
2	ASP	С	200	-	6,8,8	0.97	0	8,10,10	1.22	0	
3	PG4	В	203	-	12,12,12	0.33	0	11,11,11	0.26	0	
3	PG4	В	202	-	12,12,12	0.22	0	11,11,11	0.32	0	
3	PG4	D	201	-	12,12,12	0.20	0	11,11,11	0.56	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
2	ASP	В	201	-	-	2/8/8/8	-
2	ASP	С	200	-	-	2/8/8/8	-
3	PG4	В	203	-	-	5/10/10/10	-
3	PG4	В	202	-	-	3/10/10/10	-
3	PG4	D	201	-	-	6/10/10/10	-

All (1) bond length outliers are listed below:



Mol	Chain	Res	Type	Atoms	Z	Observed(Å)	Ideal(Å)
2	В	201	ASP	OD1-CG	2.20	1.29	1.22

All (2) bond angle outliers are listed below:

Mol	Chain	Res	Type	Atoms	Ζ	$\mathbf{Observed}(^{o})$	$Ideal(^{o})$
2	В	201	ASP	OXT-C-CA	2.92	123.33	113.38
2	В	201	ASP	OXT-C-O	-2.81	117.72	124.09

There are no chirality outliers.

5 of 18 torsion outliers are listed below:

Mol	Chain	Res	Type	Atoms
2	В	201	ASP	O-C-CA-N
2	С	200	ASP	O-C-CA-N
2	В	201	ASP	OXT-C-CA-N
3	В	203	PG4	O4-C7-C8-O5
2	С	200	ASP	OXT-C-CA-N

There are no ring outliers.

3 monomers are involved in 5 short contacts:

Mol	Chain	Res	Type	Clashes	Symm-Clashes
3	В	203	PG4	1	0
3	В	202	PG4	2	0
3	D	201	PG4	3	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	170/185~(91%)	0.19	9 (5%) 26 29	6, 18, 43, 58	0
1	В	163/185~(88%)	-0.01	3 (1%) 68 72	5, 14, 36, 47	0
1	С	164/185~(88%)	-0.05	0 100 100	5, 13, 31, 42	0
1	D	169/185~(91%)	0.10	7 (4%) 37 41	5, 15, 44, 55	0
All	All	666/740~(90%)	0.06	19 (2%) 51 56	5, 15, 40, 58	0

The worst 5 of 19 RSRZ outliers are listed below:

Mol	Chain	Res	Type	RSRZ	
1	А	61	PHE	3.9	
1	D	59	LEU	3.4	
1	А	169	ASP	3.0	
1	D	63	GLU	3.0	
1	А	59	LEU	3.0	

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.



Mol	Type	Chain	Res	Atoms	RSCC	RSR	$\mathbf{B} ext{-factors}(\mathrm{\AA}^2)$	Q < 0.9
3	PG4	В	203	13/13	0.69	0.21	$38,\!46,\!50,\!51$	0
3	PG4	В	202	13/13	0.88	0.17	25,38,47,48	0
3	PG4	D	201	13/13	0.90	0.17	23,36,47,51	0
2	ASP	В	201	9/9	0.92	0.18	19,25,31,37	0
2	ASP	С	200	9/9	0.96	0.14	18,20,24,25	0

6.5 Other polymers (i)

There are no such residues in this entry.

