Full wwPDB X-ray Structure Validation Report ⓘ

Jun 18, 2024 – 03:56 PM EDT

PDB ID	:	3WII
Title	:	Crystal structure of the Fab fragment of B2212A, a murine monoclonal antibody specific for the third fibronectin domain (Fn3) of human ROBO1.
Authors	:	Nakayama, T.; Mizohata, E.; Yamashita, T.; Nagatoishi, S.; Nakakido, M.; Iwanari, H.; Mochizuki, Y.; Kado, Y.; Yokota, Y.; Sato, R.; Tsumoto, K.; Fujitani, H.; Kodama, T.; Hamakubo, T.; Inoue, T.
Deposited on	:	2013-09-12
Resolution	:	1.60 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 types of validation reports are described at
http://www.wwpdb.org/validation/2017/FAQs#types.

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

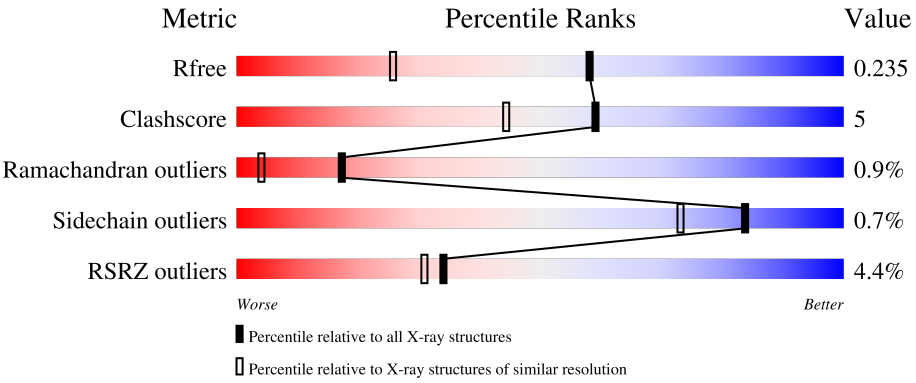
MolProbity	:	4.02b-467
Xtriage (Phenix)	:	1.20.1
EDS	:	2.37.1
Percentile statistics	:	20191225.v01 (using entries in the PDB archive December 25th 2019)
Refmac	:	5.8.0158
CCP4	:	7.0.044 (Gargrove)
Ideal geometry (proteins)	:	Engh & Huber (2001)
Ideal geometry (DNA, RNA)	:	Parkinson et al. (1996)
Validation Pipeline (wwPDB-VP)	:	2.37.1

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 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	Whole archive (#Entries)	Similar resolution (#Entries, resolution range(Å))
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	L	213	6% 85% 15%
1	M	213	2% 92% 7%
2	H	220	5% 92% 7%
2	I	220	5% 91% 8%

2 Entry composition (i)

There are 3 unique types of molecules in this entry. The entry contains 7336 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 anti-human ROBO1 antibody B2212A Fab light chain.

Mol	Chain	Residues	Atoms					ZeroOcc	AltConf	Trace
1	L	213	Total	C	N	O	S	0	1	0
			1658	1030	280	341	7			
1	M	213	Total	C	N	O	S	0	1	0
			1658	1030	280	341	7			

- Molecule 2 is a protein called anti-human ROBO1 antibody B2212A Fab heavy chain.

Mol	Chain	Residues	Atoms					ZeroOcc	AltConf	Trace
2	H	220	Total	C	N	O	S	0	2	0
			1680	1064	270	338	8			
2	I	220	Total	C	N	O	S	0	1	0
			1677	1062	270	338	7			

- Molecule 3 is water.

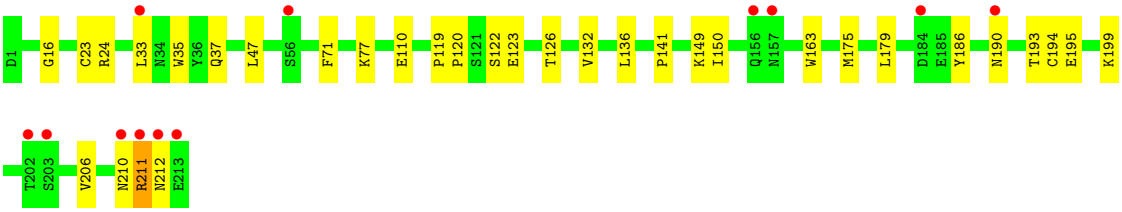
Mol	Chain	Residues	Atoms		ZeroOcc	AltConf
3	L	122	Total	O	0	0
			122	122		
3	H	174	Total	O	0	0
			174	174		
3	M	149	Total	O	0	0
			149	149		
3	I	218	Total	O	0	0
			218	218		

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.

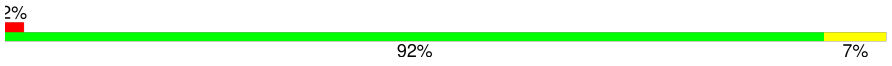
• Molecule 1: anti-human ROBO1 antibody B2212A Fab light chain

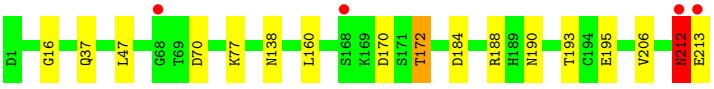
Chain L:



• Molecule 1: anti-human ROBO1 antibody B2212A Fab light chain

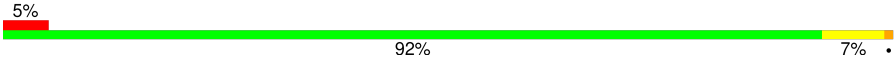
Chain M:

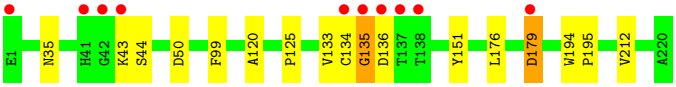




• Molecule 2: anti-human ROBO1 antibody B2212A Fab heavy chain

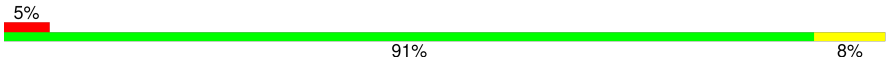
Chain H:

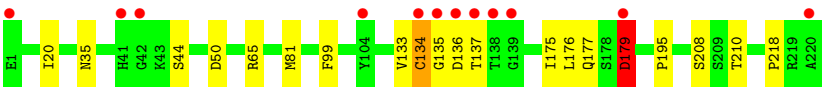




• Molecule 2: anti-human ROBO1 antibody B2212A Fab heavy chain

Chain I:





4 Data and refinement statistics (i)

Property	Value	Source
Space group	P 1 21 1	Depositor
Cell constants a, b, c, α, β, γ	41.82Å 136.49Å 77.51Å 90.00° 91.86° 90.00°	Depositor
Resolution (Å)	14.91 – 1.60 14.91 – 1.60	Depositor EDS
% Data completeness (in resolution range)	94.1 (14.91-1.60) 94.1 (14.91-1.60)	Depositor EDS
R_{merge}	0.11	Depositor
R_{sym}	(Not available)	Depositor
$< I/\sigma(I) >$ [1]	5.09 (at 1.60Å)	Xtriage
Refinement program	REFMAC 5.5.0109, PHENIX 1.8.4_1496	Depositor
R, R_{free}	0.185 , 0.230 0.190 , 0.235	Depositor DCC
R_{free} test set	5363 reflections (5.01%)	wwPDB-VP
Wilson B-factor (Å^2)	22.5	Xtriage
Anisotropy	0.191	Xtriage
Bulk solvent k_{sol}(e/Å^3), B_{sol}(Å^2)	0.39 , 52.2	EDS
L-test for twinning[2]	$< \|L\| > = 0.48$, $< L^2 > = 0.31$	Xtriage
Estimated twinning fraction	0.035 for h,-k,-l	Xtriage
F_o,F_c correlation	0.96	EDS
Total number of atoms	7336	wwPDB-VP
Average B, all atoms (Å^2)	34.0	wwPDB-VP

Xtriage's analysis on translational NCS is as follows: *The largest off-origin peak in the Patterson function is 5.44% of the height of the origin peak. No significant pseudotranslation is detected.*

[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

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	L	0.36	0/1694	0.55	0/2297
1	M	0.44	0/1694	0.61	0/2297
2	H	0.40	0/1730	0.59	0/2366
2	I	0.45	0/1724	0.66	0/2358
All	All	0.42	0/6842	0.60	0/9318

Chiral center outliers are detected by calculating the chiral volume of a chiral center and verifying if the center is modelled as a planar moiety or with the opposite hand.A planarity outlier is detected by checking planarity of atoms in a peptide group, atoms in a mainchain group or atoms of a sidechain that are expected to be planar.

Mol	Chain	#Chirality outliers	#Planarity outliers
2	I	0	1

There are no bond length outliers.

There are no bond angle outliers.

There are no chirality outliers.

All (1) planarity outliers are listed below:

Mol	Chain	Res	Type	Group
2	I	179	ASP	Peptide

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	L	1658	0	1600	26	0

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Mol	Chain	Non-H	H(model)	H(added)	Clashes	Symm-Clashes
1	M	1658	0	1600	13	0
2	H	1680	0	1631	11	0
2	I	1677	0	1627	14	0
3	H	174	0	0	1	0
3	I	218	0	0	3	2
3	L	122	0	0	4	0
3	M	149	0	0	3	2
All	All	7336	0	6458	61	2

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 5.

All (61) 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:L:149:LYS:NZ	1:L:195:GLU:OE2	2.19	0.76
2:I:176:LEU:HD22	2:I:179:ASP:HA	1.67	0.74
1:L:110:GLU:OE1	3:L:375:HOH:O	2.06	0.72
2:I:133:VAL:HA	2:I:134:CYS:HB2	1.71	0.71
1:M:213:GLU:OE2	3:M:307:HOH:O	2.08	0.71
1:M:138:ASN:HA	1:M:172:THR:HG23	1.73	0.71
1:M:190:ASN:OD1	1:M:213:GLU:HG2	1.91	0.70
2:H:44:SER:OG	3:H:330:HOH:O	2.11	0.68
1:L:195:GLU:HG2	1:L:206:VAL:HG22	1.77	0.66
1:L:149:LYS:HB2	1:L:193:THR:HB	1.79	0.65
1:M:195:GLU:HG2	1:M:206:VAL:HG22	1.77	0.65
1:M:70:ASP:OD2	3:M:395:HOH:O	2.16	0.63
2:H:133:VAL:O	2:H:135:GLY:N	2.32	0.63
1:L:150:ILE:HD11	1:L:179:LEU:HD21	1.79	0.63
2:I:133:VAL:HG23	2:I:136:ASP:HB2	1.87	0.57
1:L:211:ARG:O	1:L:212:ASN:ND2	2.36	0.57
1:L:110:GLU:OE2	1:L:199:LYS:NZ	2.27	0.57
1:M:138:ASN:HA	1:M:172:THR:CG2	2.35	0.56
1:L:190:ASN:ND2	1:L:210:ASN:OD1	2.39	0.56
2:I:133:VAL:CA	2:I:134:CYS:HB2	2.35	0.56
1:L:24:ARG:NH1	3:L:333:HOH:O	2.36	0.55
1:L:210:ASN:O	1:L:212:ASN:N	2.40	0.55
2:H:212:VAL:HG22	2:I:210:THR:HG22	1.89	0.54
1:L:33:LEU:HD13	1:L:71:PHE:CG	2.43	0.54
1:L:122:SER:O	1:L:126:THR:HG23	2.07	0.53
2:I:65:ARG:NH2	3:I:457:HOH:O	2.24	0.53

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Atom-1	Atom-2	Interatomic distance (Å)	Clash overlap (Å)
2:H:176:LEU:HG	2:H:179:ASP:HA	1.91	0.53
1:L:119:PRO:HD3	2:H:133:VAL:HG12	1.91	0.52
1:L:37:GLN:HB2	1:L:47:LEU:HD11	1.92	0.52
1:L:33:LEU:HD13	1:L:71:PHE:CD1	2.45	0.51
1:L:211:ARG:HH11	1:L:211:ARG:HG3	1.75	0.51
1:L:136:LEU:HD22	1:L:175:MET:HE1	1.93	0.51
1:M:37:GLN:HB2	1:M:47:LEU:HD11	1.93	0.50
2:H:133:VAL:HG22	2:H:136:ASP:OD2	2.11	0.50
1:M:212:ASN:OD1	1:M:213:GLU:N	2.45	0.49
1:M:16:GLY:HA2	1:M:77:LYS:HG3	1.94	0.48
1:L:211:ARG:C	1:L:212:ASN:HD22	2.16	0.48
2:I:195:PRO:HB3	2:I:218:PRO:HG3	1.96	0.48
2:H:194:TRP:CG	2:H:195:PRO:HA	2.48	0.48
1:M:170:ASP:OD1	1:M:172:THR:HB	2.14	0.48
1:L:186:TYR:HE2	1:L:211:ARG:NH1	2.12	0.47
1:L:123:GLU:OE1	3:L:323:HOH:O	2.20	0.46
2:H:120:ALA:HB2	2:H:179:ASP:OD1	2.16	0.45
1:M:160:LEU:HB3	2:I:175:ILE:HD12	1.98	0.44
2:I:177:GLN:NE2	3:I:401:HOH:O	2.50	0.44
3:M:301:HOH:O	2:I:44:SER:HB3	2.16	0.44
2:I:35:ASN:OD1	2:I:50:ASP:HB3	2.17	0.44
1:M:193:THR:HG22	1:M:195:GLU:HG3	2.00	0.44
2:I:135:GLY:HA3	3:I:315:HOH:O	2.18	0.43
1:L:23:CYS:HB2	1:L:35:TRP:CH2	2.53	0.43
1:L:163:TRP:HD1	3:L:396:HOH:O	2.01	0.43
2:I:208:SER:O	2:I:210:THR:HG23	2.19	0.43
2:H:125:PRO:HB3	2:H:151:TYR:HB3	2.01	0.43
1:L:110:GLU:HG2	1:L:141:PRO:HD3	2.00	0.42
1:M:184:ASP:O	1:M:188:ARG:HG3	2.19	0.42
1:L:16:GLY:HA2	1:L:77:LYS:HG3	2.00	0.42
1:L:211:ARG:HG3	1:L:212:ASN:HD22	1.86	0.41
2:H:43:LYS:HB3	2:H:43:LYS:HE2	1.76	0.41
2:I:20:ILE:HD11	2:I:81:MET:HE2	2.03	0.41
1:L:120:PRO:HD3	1:L:132:VAL:HG22	2.02	0.40
2:H:35:ASN:OD1	2:H:50:ASP:HB3	2.21	0.40

All (2) symmetry-related close contacts are listed below. The label for Atom-2 includes the symmetry operator and encoded unit-cell translations to be applied.

Atom-1	Atom-2	Interatomic distance (Å)	Clash overlap (Å)
3:M:411:HOH:O	3:I:475:HOH:O[1_655]	2.14	0.06

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Atom-1	Atom-2	Interatomic distance (Å)	Clash overlap (Å)
3:M:410:HOH:O	3:I:476:HOH:O[1_655]	2.15	0.05

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	L	212/213 (100%)	204 (96%)	7 (3%)	1 (0%)	29 11
1	M	212/213 (100%)	206 (97%)	5 (2%)	1 (0%)	29 11
2	H	220/220 (100%)	213 (97%)	4 (2%)	3 (1%)	11 2
2	I	219/220 (100%)	209 (95%)	7 (3%)	3 (1%)	11 2
All	All	863/866 (100%)	832 (96%)	23 (3%)	8 (1%)	17 4

All (8) Ramachandran outliers are listed below:

Mol	Chain	Res	Type
1	L	211	ARG
2	I	134	CYS
2	I	137	THR
2	I	179	ASP
2	H	134	CYS
2	H	135	GLY
1	M	212	ASN
2	H	179	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	L	192/191 (100%)	190 (99%)	2 (1%)	76 61
1	M	192/191 (100%)	190 (99%)	2 (1%)	76 61
2	H	194/192 (101%)	193 (100%)	1 (0%)	88 80
2	I	193/192 (100%)	192 (100%)	1 (0%)	88 80
All	All	771/766 (101%)	765 (99%)	6 (1%)	84 70

All (6) residues with a non-rotameric sidechain are listed below:

Mol	Chain	Res	Type
1	L	194[A]	CYS
1	L	194[B]	CYS
2	H	99	PHE
1	M	172	THR
1	M	212	ASN
2	I	99	PHE

Sometimes sidechains can be flipped to improve hydrogen bonding and reduce clashes. All (2) such sidechains are listed below:

Mol	Chain	Res	Type
1	L	212	ASN
2	H	54	ASN

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)

There are no ligands in this entry.

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

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, 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	<RSRZ>	#RSRZ>2			OWAB(Å^2)	Q<0.9
1	L	213/213 (100%)	0.21	12 (5%)	24	22	23, 34, 66, 146	2 (0%)
1	M	213/213 (100%)	-0.14	4 (1%)	66	65	18, 27, 54, 135	1 (0%)
2	H	220/220 (100%)	0.09	10 (4%)	33	30	20, 28, 72, 143	0
2	I	220/220 (100%)	-0.09	12 (5%)	25	22	17, 24, 66, 168	0
All	All	866/866 (100%)	0.02	38 (4%)	34	31	17, 29, 63, 168	3 (0%)

All (38) RSRZ outliers are listed below:

Mol	Chain	Res	Type	RSRZ
2	H	42	GLY	10.1
1	M	213	GLU	9.3
2	I	135	GLY	8.6
2	H	134	CYS	8.5
2	I	134	CYS	7.8
2	H	135	GLY	7.6
1	L	212	ASN	6.8
1	M	212	ASN	6.5
2	I	138	THR	6.3
2	H	138	THR	6.0
2	I	42	GLY	5.3
2	H	41	HIS	5.1
2	I	137	THR	5.1
2	H	136	ASP	5.0
2	H	43	LYS	4.8
1	L	213	GLU	4.3
2	H	137	THR	3.9
2	H	1	GLU	3.8
1	L	211	ARG	3.5
1	L	202	THR	3.5
2	I	136	ASP	3.4

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Mol	Chain	Res	Type	RSRZ
2	I	41	HIS	3.2
1	L	56	SER	3.1
2	I	139	GLY	2.8
1	L	156	GLN	2.8
1	L	157	ASN	2.8
1	L	210	ASN	2.7
2	I	1	GLU	2.6
2	I	179	ASP	2.4
2	I	104	TYR	2.3
1	L	184	ASP	2.3
1	L	190	ASN	2.2
1	M	68	GLY	2.2
2	I	220	ALA	2.1
1	L	33	LEU	2.1
1	L	203	SER	2.0
1	M	168	SER	2.0
2	H	179	ASP	2.0

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)

There are no ligands in this entry.

6.5 Other polymers (i)

There are no such residues in this entry.