

# wwPDB X-ray Structure Validation Summary Report (i)

#### Aug 25, 2020 – 03:30 PM BST

PDB ID : 4JFD

Title: Preservation of peptide specificity during TCR-MHC contact dominated affin-

ity enhancement of a melanoma-specific TCR

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Deposited on : 2013-02-28

Resolution : 2.46 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 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 : 2.13

Percentile statistics : 20191225.v01 (using entries in the PDB archive December 25th 2019)

Refmac: 5.8.0158

CCP4 : 7.0.044 (Gargrove) roteins) : Engh & Huber (2001)

Ideal geometry (proteins) : Engh & Huber (2001)
Ideal geometry (DNA, RNA) : Parkinson et al. (1996)

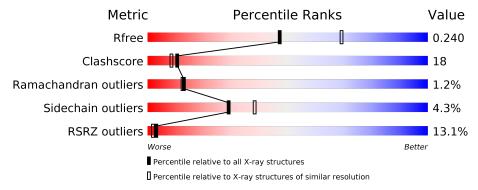
Validation Pipeline (wwPDB-VP) : 2.13

## 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 2.46 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	$\begin{array}{c} \text{Whole archive} \\ (\#\text{Entries}) \end{array}$	$\begin{array}{c} {\rm Similar  resolution} \\ (\#{\rm Entries,  resolution  range(\AA)}) \end{array}$
$R_{free}$	130704	1544 (2.48-2.44)
Clashscore	141614	1613 (2.48-2.44)
Ramachandran outliers	138981	1598 (2.48-2.44)
Sidechain outliers	138945	1598 (2.48-2.44)
RSRZ outliers	127900	1523 (2.48-2.44)

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 <=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		
			26%		
1	A	276	68%	28%	•
			7%		
2	В	100	74%	24%	•
3	С	10	60%	40%	
			11%		
4	D	196	72%	22%	5% •
			2%		
5	E	245	71%	27%	•



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
7	SO4	A	303	_	-	X	-
7	SO4	A	304	-	-	X	-
7	SO4	A	305	-	-	X	-
7	SO4	Е	305	-	-	X	-



# 2 Entry composition (i)

There are 8 unique types of molecules in this entry. The entry contains 6820 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 HLA class I histocompatibility antigen, A-2 alpha chain.

Mol	Chain	Residues		At	oms			ZeroOcc	AltConf	Trace
1	Α	276	Total	С	N	О	S	0	0	0
_	11	210	2253	1408	410	426	9			

• Molecule 2 is a protein called Beta-2-microglobulin.

Mol	Chain	Residues		At	oms			ZeroOcc	AltConf	Trace
2	R	100	Total	С	N	О	S	0	0	0
	D	100	837	533	141	159	4			0

There is a discrepancy between the modelled and reference sequences:

Chain	Residue	Modelled	Actual	${f Comment}$	Reference
В	0	MET	_	INITIATING METHIONINE	UNP P61769

• Molecule 3 is a protein called Melanoma peptide.

Mol	Chain	Residues	1	Ator	$\mathbf{n}\mathbf{s}$		ZeroOcc	AltConf	Trace
3	С	10	Total 70	C 46	N 10	O 14	0	0	0

• Molecule 4 is a protein called High Affinity TCR Alpha Chain.

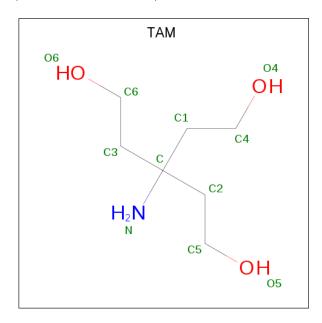
Mo	Chain	Residues		At	oms			ZeroOcc	AltConf	Trace
4	D	196	Total 1525	C 947	N 255	O 315	S 8	0	0	0

• Molecule 5 is a protein called High Affinity TCR Beta Chain.

$\mathbf{Mol}$	Chain	Residues		$\mathbf{At}$	oms			ZeroOcc	AltConf	Trace
5	E	245	Total 1944	C 1235	N 334	O 368	S 7	0	0	0

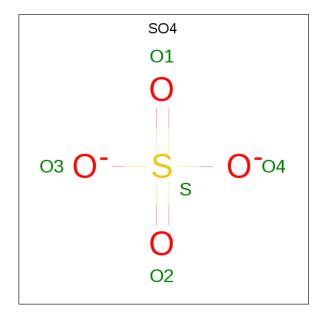


• Molecule 6 is TRIS(HYDROXYETHYL)AMINOMETHANE (three-letter code: TAM) (formula:  $C_7H_{17}NO_3$ ).



Mol	Chain	Residues	A	tor	ns		ZeroOcc	AltConf
6	A	1	Total 11	C 7	N 1	O 3	0	0

 $\bullet$  Molecule 7 is SULFATE ION (three-letter code: SO4) (formula:  $\mathrm{O_4S}).$ 



Mol	Chain	Residues	Aton	ıs	ZeroOcc	AltConf
7	A	1	Total C	) S 4 1	0	0

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Mol	Chain	Residues	Atoms	ZeroOcc	AltConf
7	A	1	Total O S 5 4 1	0	0
7	A	1	Total O S 5 4 1	0	0
7	A	1	Total O S 5 4 1	0	0
7	A	1	Total O S 5 4 1	0	0
7	Е	1	Total O S 5 4 1	0	0
7	Е	1	Total O S 5 4 1	0	0
7	Е	1	Total O S 5 4 1	0	0
7	Е	1	Total O S 5 4 1	0	0
7	E	1	Total O S 5 4 1	0	0
7	E	1	Total O S 5 4 1	0	0

### • Molecule 8 is water.

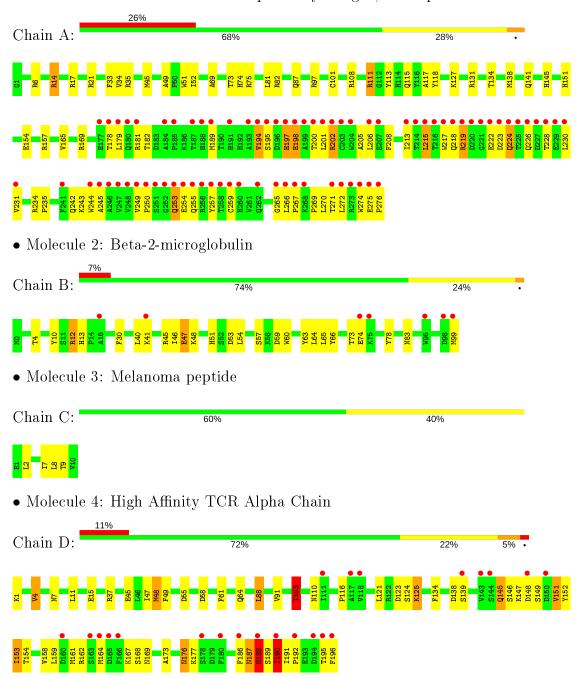
Mol	Chain	Residues	Atoms	ZeroOcc	AltConf
8	A	31	Total O 31 31	0	0
8	В	13	Total O 13 13	0	0
8	С	1	Total O 1 1	0	0
8	D	32	Total O 32 32	0	0
8	E	48	Total O 48 48	0	0



# 3 Residue-property plots (i)

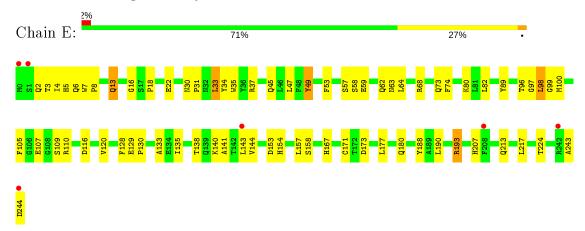
These plots are drawn for all protein, RNA, DNA and oligosaccharide chains in the entry. The first graphic for a chain summarises the proportions of the various outlier classes displayed in the second graphic. The second graphic shows the sequence view annotated by issues in geometry and electron density. 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: HLA class I histocompatibility antigen, A-2 alpha chain





• Molecule 5: High Affinity TCR Beta Chain





# 4 Data and refinement statistics (i)

Property	Value	Source
Space group	P 43	Depositor
Cell constants	121.49Å 121.49Å 82.96Å	Domositon
a, b, c, $\alpha$ , $\beta$ , $\gamma$	$90.00^{\circ}$ $90.00^{\circ}$ $90.00^{\circ}$	Depositor
Resolution (Å)	54.33 - 2.46	Depositor
Resolution (A)	54.33 - 2.46	EDS
% Data completeness	100.0 (54.33-2.46)	Depositor
(in resolution range)	$100.0 \ (54.33-2.46)$	EDS
$R_{merge}$	0.07	Depositor
$R_{sym}$	0.07	Depositor
$< I/\sigma(I) > 1$	2.91 (at 2.45Å)	Xtriage
Refinement program	REFMAC	Depositor
D D.	0.202 , 0.248	Depositor
$R, R_{free}$	0.195 , $0.240$	DCC
$R_{free}$ test set	2225 reflections $(5.04\%)$	wwPDB-VP
Wilson B-factor (Å <sup>2</sup> )	51.1	Xtriage
Anisotropy	0.277	Xtriage
Bulk solvent $k_{sol}(e/Å^3)$ , $B_{sol}(Å^2)$	0.33,55.5	EDS
L-test for twinning <sup>2</sup>	$< L >=0.49, < L^2>=0.32$	Xtriage
Estimated twinning fraction	0.033 for h,-k,-l	Xtriage
$F_o, F_c$ correlation	0.95	EDS
Total number of atoms	6820	wwPDB-VP
Average B, all atoms (Å <sup>2</sup> )	68.0	wwPDB-VP

Xtriage's analysis on translational NCS is as follows: The largest off-origin peak in the Patterson function is 3.00% of the height of the origin peak. No significant pseudotranslation is detected.

<sup>&</sup>lt;sup>2</sup>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.



<sup>&</sup>lt;sup>1</sup>Intensities 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: TAM, SO4

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	Bond	angles
MIOI	Chain	RMSZ	# Z  > 5	RMSZ	$\mid \# Z  > 5$
1	A	0.98	$2/2319 \ (0.1\%)$	0.54	0/3149
2	В	0.87	0/860	0.57	0/1162
3	С	1.09	0/69	0.60	0/92
4	D	1.04	5/1556~(0.3%)	0.61	0/2107
5	Е	1.09	5/2003~(0.2%)	0.60	0/2732
All	All	1.02	$12/6807 \ (0.2\%)$	0.58	0/9242

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.

I	Mol	Chain	#Chirality outliers	#Planarity outliers
	4	D	0	1

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

Mol	Chain	Res	Type	${f Atoms}$	$\mathbf{Z}$	${f Observed(\AA)}$	$\mathbf{Ideal}(\mathbf{\AA})$
1	A	101	CYS	CB-SG	-17.85	1.51	1.82
4	D	103	THR	CB-CG2	-7.12	1.28	1.52
4	D	4	VAL	CB-CG1	-6.32	1.39	1.52
5	Е	180	GLN	CB-CG	5.82	1.68	1.52
4	D	91	VAL	CB-CG1	-5.60	1.41	1.52

There are no bond angle outliers.

There are no chirality outliers.

All (1) planarity outliers are listed below:



Mol	Chain	Res	Type	Group
4	D	48	MET	Mainchain

### 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	Α	2253	0	2103	99	0
2	В	837	0	803	25	0
3	С	70	0	81	5	0
4	D	1525	0	1439	60	0
5	Ε	1944	0	1848	74	0
6	A	11	0	17	2	0
7	A	25	0	0	9	0
7	E	30	0	0	5	0
8	A	31	0	0	0	0
8	В	13	0	0	1	0
8	С	1	0	0	0	0
8	D	32	0	0	0	0
8	Ε	48	0	0	3	0
All	All	6820	0	6291	240	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 18.

The worst 5 of 240 close contacts within the same asymmetric unit are listed below, sorted by their clash magnitude.

Atom-1	Atom-2	$egin{array}{ll}  ext{Interatomic} \  ext{distance} \ ( ext{\AA}) \end{array}$	$egin{array}{c}  ext{Clash} \  ext{overlap } ( ext{Å}) \end{array}$
1:A:201:LEU:HD12	1:A:217:TRP:CZ2	1.66	1.30
5:E:63:ASN:ND2	5:E:82:LEU:HD13	1.65	1.11
1:A:131:ARG:NH2	7:A:303:SO4:O3	1.87	1.08
1:A:201:LEU:CD1	1:A:217:TRP:CZ2	2.46	0.99
1:A:215:LEU:HD11	1:A:245:ALA:HB2	1.45	0.98

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	entiles
1	A	274/276 (99%)	250 (91%)	19 (7%)	5 (2%)	8	6
2	В	98/100 (98%)	94 (96%)	4 (4%)	0	100	100
3	С	8/10 (80%)	8 (100%)	0	0	100	100
4	D	194/196 (99%)	179 (92%)	10 (5%)	5 (3%)	5	3
5	E	243/245 (99%)	229 (94%)	14 (6%)	0	100	100
All	All	817/827 (99%)	760 (93%)	47 (6%)	10 (1%)	13	12

5 of 10 Ramachandran outliers are listed below:

Mol	Chain	${f Res}$	Type
1	A	223	ASP
4	D	148	ASP
1	A	194	VAL
1	A	198	GLU
1	A	197	HIS

#### 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	A	$232/232 \; (100\%)$	223 (96%)	9 (4%)	32	42	
2	В	95/95 (100%)	91 (96%)	4 (4%)	30	39	
3	С	7/7 (100%)	7 (100%)	0	100	100	
4	D	174/174 (100%)	162 (93%)	12 (7%)	15	18	

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Mol	Chain	Analysed	Rotameric	Rotameric Outliers	
5	E	211/211 (100%)	205 (97%)	6 (3%)	43 56
All	All	719/719 (100%)	688 (96%)	31 (4%)	29 38

5 of 31 residues with a non-rotameric sidechain are listed below:

Mol	Chain	Res	Type
4	D	11	LEU
4	D	110	ASN
5	E	107	GLU
4	D	88	LEU
4	D	145	GLN

Some sidechains can be flipped to improve hydrogen bonding and reduce clashes. 5 of 28 such sidechains are listed below:

Mol	Chain	Res	Type
2	В	17	ASN
4	D	110	ASN
5	E	119	ASN
2	В	83	ASN
4	D	38	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 monosaccharides in this entry.

## 5.6 Ligand geometry (i)

12 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	Trans	Chain	Dog	Res Link	В	ond leng	$\operatorname{gths}$	Е	ond ang	gles
Mol	Type	Chain	nes	Link	Counts	RMSZ	# Z >2	Counts	RMSZ	# Z  > 2
7	SO4	Е	304	-	4,4,4	0.12	0	6,6,6	0.13	0
7	SO4	A	302	_	4,4,4	0.29	0	6,6,6	0.21	0
7	SO4	A	304	_	4,4,4	0.08	0	6,6,6	0.23	0
7	SO4	A	303	-	4,4,4	0.34	0	6,6,6	0.13	0
7	SO4	Е	303	-	4,4,4	0.24	0	6,6,6	0.08	0
7	SO4	Е	302	-	4,4,4	0.33	0	6,6,6	0.09	0
6	TAM	A	301	_	7,10,10	1.01	0	9,12,12	2.14	5 (55%)
7	SO4	Е	305	_	4,4,4	0.25	0	6,6,6	0.21	0
7	SO4	A	305	-	4,4,4	0.35	0	6,6,6	0.10	0
7	SO4	A	306	_	4,4,4	0.12	0	6,6,6	0.08	0
7	SO4	Е	306	_	4,4,4	0.21	0	6,6,6	0.14	0
7	SO4	Е	301	-	4,4,4	0.27	0	6,6,6	0.14	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.

$\mathbf{Mol}$	Type	Chain	Res	Link	Chirals	${f Torsions}$	Rings
6	TAM	A	301	_	-	7/12/12/12	_

There are no bond length outliers.

All (5) bond angle outliers are listed below:

Mol	Chain	Res	Type	Atoms	${f Z}$	$Observed(^o)$	$\mathbf{Ideal}(^o)$
6	A	301	TAM	C2-C-C1	3.20	116.15	110.50
6	A	301	TAM	C3-C-C1	3.06	115.90	110.50
6	A	301	TAM	C3-C-C2	2.70	115.27	110.50
6	A	301	TAM	C3-C-N	-2.33	101.44	108.09
6	A	301	TAM	C1-C-N	-2.10	102.10	108.09

There are no chirality outliers.

5 of 7 torsion outliers are listed below:



Mol	Chain	Res	Type	Atoms
6	A	301	TAM	C2-C-C1-C4
6	A	301	TAM	C3-C-C1-C4
6	A	301	TAM	N-C-C1-C4
6	A	301	TAM	C3-C-C2-C5
6	A	301	TAM	N-C-C2-C5

There are no ring outliers.

8 monomers are involved in 16 short contacts:

Mol	Chain	Res	Type	Clashes	Symm-Clashes
7	Е	304	SO4	1	0
7	A	302	SO4	1	0
7	A	304	SO4	2	0
7	A	303	SO4	4	0
7	E	303	SO4	1	0
6	A	301	TAM	2	0
7	E	305	SO4	3	0
7	A	305	SO4	2	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	<RSRZ $>$	$\#\mathrm{RSRZ}{>}2$	$OWAB(\AA^2)$	Q < 0.9
1	A	$276/276 \ (100\%)$	1.32	73 (26%) 0 0	26, 59, 196, 217	0
2	В	100/100 (100%)	0.44	7 (7%) 16 13	26, 66, 153, 169	0
3	С	10/10 (100%)	-0.20	0 100 100	25, 28, 34, 38	0
4	D	$196/196 \; (100\%)$	0.59	22 (11%) 5 3	21, 49, 135, 163	0
5	E	$245/245 \ (100\%)$	0.01	6 (2%) 59 54	24, 44, 107, 151	0
All	All	827/827 (100%)	0.63	108 (13%) 3 2	21, 52, 164, 217	0

The worst 5 of 108 RSRZ outliers are listed below:

Mol	Chain	$\operatorname{Res}$	Type	RSRZ
1	A	201	LEU	10.6
1	A	196	ASP	10.2
1	A	249	VAL	10.0
1	A	248	VAL	8.3
1	A	250	PRO	8.2

### 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	${f B\text{-factors}}({f \AA}^2)$	Q < 0.9
7	SO4	E	304	5/5	0.76	0.19	46,53,65,73	5
6	TAM	A	301	11/11	0.82	0.30	66,80,80,80	0
7	SO4	Е	302	5/5	0.83	0.22	36,44,52,62	5
7	SO4	E	303	5/5	0.85	0.18	47,53,67,73	5
7	SO4	E	306	5/5	0.88	0.27	31,34,46,54	5
7	SO4	A	306	5/5	0.93	0.16	40,41,53,62	5
7	SO4	E	305	5/5	0.93	0.22	35,39,47,53	5
7	SO4	Е	301	5/5	0.93	0.19	63,76,80,80	1
7	SO4	A	305	5/5	0.94	0.15	33,41,44,52	5
7	SO4	A	302	5/5	0.96	0.09	67,74,80,80	0
7	SO4	A	303	5/5	0.96	0.10	41,64,76,77	0
7	SO4	A	304	5/5	0.99	0.10	37,40,48,52	0

## 6.5 Other polymers (i)

There are no such residues in this entry.

