

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

#### Sep 6, 2023 – 08:11 PM EDT

PDB ID	:	4FLS
Title	:	Crystal structure of Amylosucrase inactive double mutant F290K-E328Q from
		Neisseria polysaccharea in complex with sucrose.
Authors	:	Guerin, F.; Champion, E.; Moulis, C.; Barbe, S.; Tran, T.H.; Morel, S.; De-
		scroix, K.; Monsan, P.; Mulard, L.A.; Remaud-Simeon, M.; Andre, I.; Mourey,
		L.; Tranier, S.
Deposited on	:	2012-06-15
Resolution	:	2.30 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 (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:

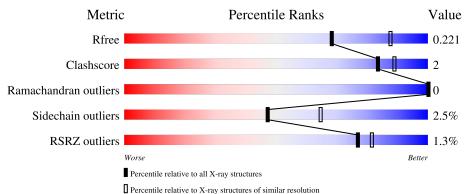
MolProbity	:	4.02b-467
Mogul	:	1.8.5 (274361), CSD as541be (2020)
Xtriage (Phenix)	:	1.13
$\mathrm{EDS}$	:	2.35
Percentile statistics	:	20191225.v01 (using entries in the PDB archive December 25th 2019)
Refmac	:	5.8.0158
CCP4	:	7.0.044 (Gargrove)
Ideal geometry (proteins)	:	Engh & Huber (2001)
Ideal geometry (DNA, RNA)	:	Parkinson et al. (1996)
Validation Pipeline (wwPDB-VP)	:	2.35

# 1 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 2.30 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	5042(2.30-2.30)
Clashscore	141614	5643 (2.30-2.30)
Ramachandran outliers	138981	5575(2.30-2.30)
Sidechain outliers	138945	5575 (2.30-2.30)
RSRZ outliers	127900	4938 (2.30-2.30)

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	628	% 	8%			
2	В	2	50%	50%			
2	С	2	10	)%			



# 2 Entry composition (i)

There are 5 unique types of molecules in this entry. The entry contains 5370 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 Amylosucrase.

Mol	Chain	Residues	Atoms			ZeroOcc	AltConf	Trace		
1	А	628	Total 5037	C 3206	N 876	0 934	S 21	0	5	0

There are 7 discrepancies between the modelled and reference sequences:

Chain	Residue	Modelled	Actual	Comment	Reference
А	1	SER	-	expression tag	UNP Q9ZEU2
А	2	PRO	-	expression tag	UNP Q9ZEU2
А	3	ASN	-	expression tag	UNP Q9ZEU2
A	4	SER	-	expression tag	UNP Q9ZEU2
А	290	LYS	PHE	engineered mutation	UNP Q9ZEU2
А	328	GLN	GLU	engineered mutation	UNP Q9ZEU2
А	537	ASP	GLY	cloning artifact	UNP Q9ZEU2

• Molecule 2 is an oligosaccharide called beta-D-fructofuranose-(2-1)-alpha-D-glucopyranose.



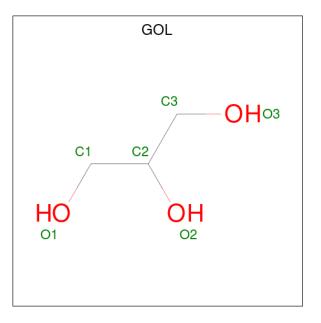
Mol	Chain	Residues	Atoms	ZeroOcc	AltConf	Trace
2	В	2	Total         C         O           23         12         11	0	0	0
2	С	2	Total         C         O           23         12         11	0	0	0

• Molecule 3 is CHLORIDE ION (three-letter code: CL) (formula: Cl).

Mol	Chain	Chain Residues Atoms		ZeroOcc	AltConf
3	А	1	Total Cl 1 1	0	0



• Molecule 4 is GLYCEROL (three-letter code: GOL) (formula:  $C_3H_8O_3$ ).



Mo	l Chain	Residues	Atoms	ZeroOcc	AltConf
4	А	1	$\begin{array}{ccc} \text{Total} & \text{C} & \text{O} \\ 6 & 3 & 3 \end{array}$	0	0

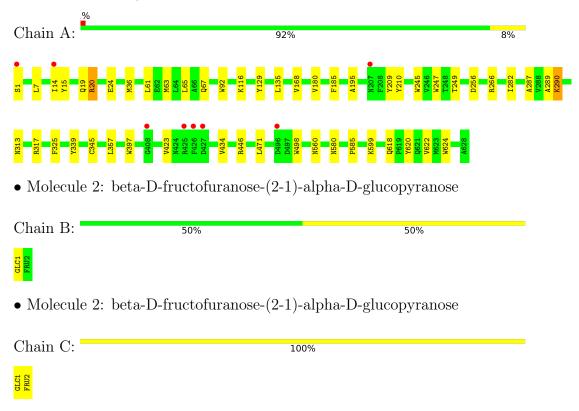
• Molecule 5 is water.

Mol	Chain	Residues	Atoms	ZeroOcc	AltConf
5	А	280	Total         O           280         280	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: Amylosucrase



## 4 Data and refinement statistics (i)

Property	Value	Source
Space group	P 21 21 2	Depositor
Cell constants	95.11Å 114.70Å 54.62Å	Depositor
a, b, c, $\alpha$ , $\beta$ , $\gamma$	$90.00^{\circ}$ $90.00^{\circ}$ $90.00^{\circ}$	Depositor
Resolution (Å)	11.98 - 2.30	Depositor
Resolution (A)	11.98 - 2.30	EDS
% Data completeness	97.9(11.98-2.30)	Depositor
(in resolution range)	98.8(11.98-2.30)	EDS
R <sub>merge</sub>	0.14	Depositor
R <sub>sym</sub>	(Not available)	Depositor
$< I/\sigma(I) > 1$	$2.62 (at 2.30 \text{\AA})$	Xtriage
Refinement program	REFMAC 5.6.0117	Depositor
$R, R_{free}$	0.176 , $0.220$	Depositor
It, Itfree	0.176 , $0.221$	DCC
$R_{free}$ test set	1340 reflections $(5.01\%)$	wwPDB-VP
Wilson B-factor $(Å^2)$	17.8	Xtriage
Anisotropy	0.052	Xtriage
Bulk solvent $k_{sol}(e/Å^3)$ , $B_{sol}(Å^2)$	0.40 , $34.8$	EDS
L-test for twinning <sup>2</sup>	$ < L >=0.47, < L^2>=0.30$	Xtriage
Estimated twinning fraction	No twinning to report.	Xtriage
$F_o, F_c$ correlation	0.94	EDS
Total number of atoms	5370	wwPDB-VP
Average B, all atoms $(Å^2)$	17.0	wwPDB-VP

Xtriage's analysis on translational NCS is as follows: The largest off-origin peak in the Patterson function is 5.75% of the height of the origin peak. No significant pseudotranslation is detected.

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



<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: CL, GLC, GOL, FRU

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	Boi	nd lengths	Bond angles		
IVIOI	Unam	RMSZ	# Z  > 5	RMSZ	# Z  > 5	
1	А	0.53	5/5181~(0.1%)	0.56	0/7051	

All (5) bond length outliers are listed below:

Mol	Chain	Res	Type	Atoms	Ζ	$\operatorname{Observed}(\operatorname{\AA})$	Ideal(Å)
1	А	92	TRP	CD2-CE2	5.28	1.47	1.41
1	А	245	TRP	CD2-CE2	5.18	1.47	1.41
1	А	624	TRP	CD2-CE2	5.15	1.47	1.41
1	А	397	TRP	CD2-CE2	5.11	1.47	1.41
1	А	498	TRP	CD2-CE2	5.02	1.47	1.41

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	5037	0	4786	24	0
2	В	23	0	21	0	0
2	С	23	0	21	0	0
3	А	1	0	0	0	0
4	А	6	0	8	0	0
5	А	280	0	0	3	0

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Mol	Chain	Non-H	H(model)	H(added)	Clashes	Symm-Clashes
All	All	5370	0	4836	24	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.

All (24) close contacts within the same asymmetric unit are listed below, sorted by their clash magnitude.

A	A.L. 0	Interatomic	Clash
Atom-1	Atom-2	distance $(\text{\AA})$	overlap (Å)
1:A:618:GLN:HG2	5:A:1067:HOH:O	1.84	0.76
1:A:63:MET:HG2	1:A:67[A]:GLN:HE21	1.59	0.67
1:A:357:LEU:HB3	1:A:471:LEU:HD22	1.79	0.63
1:A:290:LYS:HE2	5:A:938:HOH:O	2.01	0.60
1:A:67[A]:GLN:HE22	1:A:266:ARG:HD3	1.67	0.59
1:A:580:ASN:O	1:A:620:TYR:HA	2.05	0.55
1:A:210:TYR:HB2	1:A:247:TRP:HB2	1.89	0.54
1:A:20:ARG:NH2	1:A:24:GLU:OE2	2.41	0.54
1:A:423:VAL:HG12	1:A:423:VAL:O	2.07	0.53
1:A:7:LEU:HD13	1:A:65:LEU:HD13	1.91	0.52
1:A:15:TYR:HB3	1:A:19:GLN:HG3	1.92	0.50
1:A:180:VAL:HG22	1:A:282:ILE:HB	1.93	0.50
1:A:423:VAL:O	1:A:423:VAL:CG1	2.59	0.50
1:A:249:THR:HG21	1:A:290:LYS:HD3	1.97	0.46
1:A:36:MET:SD	1:A:65:LEU:HD22	2.56	0.46
1:A:116:LYS:HB2	1:A:168:VAL:HG22	1.97	0.46
1:A:289:ALA:HA	1:A:339:TYR:OH	2.17	0.45
1:A:585:PRO:HG3	5:A:1015:HOH:O	2.17	0.45
1:A:287:ALA:HB1	1:A:290:LYS:HG3	2.00	0.43
1:A:67[A]:GLN:NE2	1:A:266:ARG:HD3	2.32	0.43
1:A:195:ALA:HA	1:A:209:TYR:CE1	2.55	0.41
1:A:325:PHE:HB2	1:A:345:CYS:HA	2.03	0.41
1:A:313:ASN:O	1:A:317:ARG:HG2	2.20	0.40
1:A:129:TYR:CZ	1:A:180:VAL:HG21	2.56	0.40

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	Percentiles
1	А	631/628~(100%)	616~(98%)	15~(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	527/533~(99%)	514 (98%)	13~(2%)	47 65	

All (13) residues with a non-rotameric sidechain are listed below:

Mol	Chain	Res	Type
1	А	1	SER
1	А	14	ILE
1	А	20	ARG
1	А	61	LEU
1	А	135	LEU
1	А	185	PHE
1	А	256	ASP
1	А	290	LYS
1	А	434	VAL
1	А	446	ARG
1	А	560	ASN
1	А	599	LYS
1	А	622	VAL



Sometimes sidechains can be flipped to improve hydrogen bonding and reduce clashes. All (9) such sidechains are listed below:

Mol	Chain	Res	Type
1	А	5	GLN
1	А	123	GLN
1	А	186	ASN
1	А	196	GLN
1	А	207	ASN
1	А	338	GLN
1	А	419	ASN
1	А	560	ASN
1	А	573	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)

4 monosaccharides 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	Turne	Chain	Res	Link	Bond lengths			Bond angles		
	Mol Type Chain	nes		Counts	RMSZ	# Z  > 2	Counts	RMSZ	# Z  > 2	
2	GLC	В	1	2	11,11,12	0.32	0	$15,\!15,\!17$	2.00	1 (6%)
2	FRU	В	2	2	11,12,12	0.70	0	10,18,18	0.90	0
2	GLC	С	1	2	11,11,12	0.35	0	$15,\!15,\!17$	1.09	2 (13%)
2	FRU	С	2	2	11,12,12	0.70	1 (9%)	10,18,18	0.65	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



Mol	Type	Chain	Res	Link	Chirals	Torsions	Rings
2	GLC	В	1	2	-	0/2/19/22	0/1/1/1
2	FRU	В	2	2	-	0/5/24/24	0/1/1/1
2	GLC	С	1	2	-	2/2/19/22	0/1/1/1
2	FRU	С	2	2	-	3/5/24/24	0/1/1/1

Chemical Component Dictionary. Similar counts are reported in the Torsion and Rings columns. '-' means no outliers of that kind were identified.

All (1) bond length outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	Observed(Å)	Ideal(Å)
2	С	2	FRU	O2-C2	2.02	1.44	1.40

All (3) bond angle outliers are listed below:

Mol	Chain	Res	Type	Atoms	Z	$Observed(^{o})$	$Ideal(^{o})$
2	В	1	GLC	C1-O5-C5	6.80	121.41	112.19
2	С	1	GLC	O5-C5-C6	2.95	111.82	107.20
2	С	1	GLC	C1-O5-C5	2.25	115.25	112.19

There are no chirality outliers.

All (5) torsion outliers are listed below:

Mol	Chain	Res	Type	Atoms
2	С	1	GLC	O5-C5-C6-O6
2	С	1	GLC	C4-C5-C6-O6
2	С	2	FRU	O1-C1-C2-O5
2	С	2	FRU	O5-C5-C6-O6
2	С	2	FRU	O1-C1-C2-C3

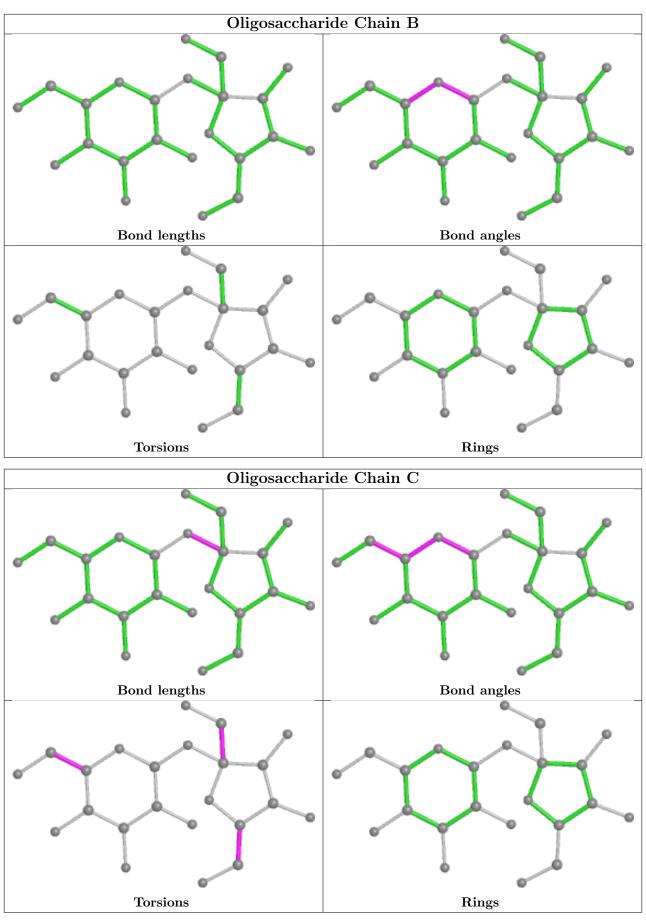
There are no ring outliers.

No monomer is involved in short contacts.

The following is a two-dimensional graphical depiction of Mogul quality analysis of bond lengths, bond angles, torsion angles, and ring geometry for oligosaccharide.









### 5.6 Ligand geometry (i)

Of 2 ligands modelled in this entry, 1 is monoatomic - leaving 1 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 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	les Link	B	ond leng	gths	Bond angles			
		туре	Unam	nes		Counts	RMSZ	# Z >2	Counts	RMSZ	# Z >2
	4	GOL	А	703	-	$5,\!5,\!5$	0.25	0	$5,\!5,\!5$	0.18	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
4	GOL	А	703	-	-	2/4/4/4	-

There are no bond length outliers.

There are no bond angle outliers.

There are no chirality outliers.

All (2) torsion outliers are listed below:

Mol	Chain	Res	Type	Atoms
4	А	703	GOL	O1-C1-C2-C3
4	А	703	GOL	O1-C1-C2-O2

There are no ring outliers.

No monomer is involved in short contacts.

### 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 $>$	#RSRZ>2		$\mathbf{OWAB}(\mathbf{A}^2)$	Q<0.9	
1	А	628/628~(100%)	-0.33	8 (1%)	77	81	11, 16, 26, 41	0

All (8) RSRZ outliers are listed below:

Mol	Chain	Res	Type	RSRZ
1	А	1	SER	4.2
1	А	496	ASP	2.6
1	А	425	ARG	2.5
1	А	408	GLY	2.5
1	А	427	ASP	2.3
1	А	207	ASN	2.3
1	А	14	ILE	2.1
1	А	426	PHE	2.1

## 6.2 Non-standard residues in protein, DNA, RNA chains (i)

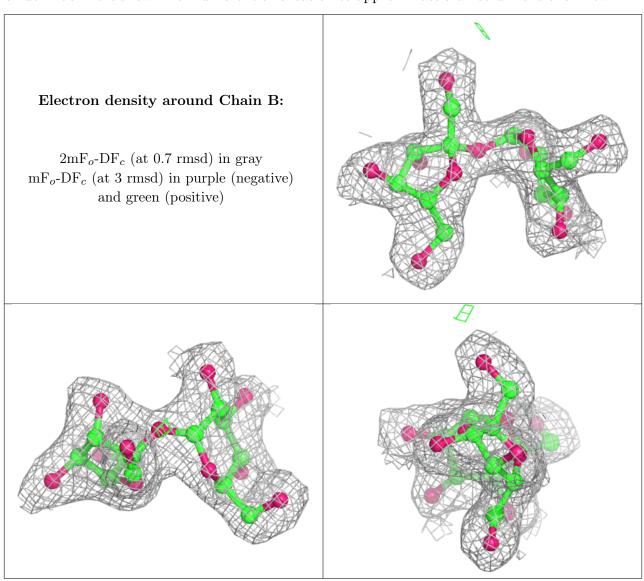
There are no non-standard protein/DNA/RNA residues in this entry.

#### 6.3 Carbohydrates (i)

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.

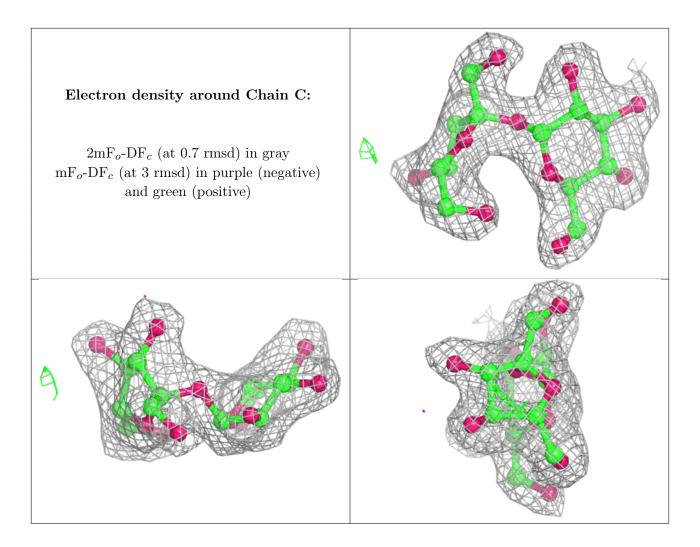
Mol	Type	Chain	Res	Atoms	RSCC	RSR	$B-factors(Å^2)$	Q < 0.9
2	GLC	С	1	11/12	0.90	0.14	$21,\!21,\!22,\!22$	0
2	FRU	С	2	12/12	0.92	0.19	21,22,23,24	0
2	FRU	В	2	12/12	0.97	0.07	13,13,14,14	0
2	GLC	В	1	11/12	0.98	0.08	13,13,13,14	0





The following is a graphical depiction of the model fit to experimental electron density for oligosaccharide. Each fit is shown from different orientation to approximate a three-dimensional view.





## 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	$B-factors(Å^2)$	Q<0.9
4	GOL	А	703	6/6	0.91	0.14	$23,\!25,\!25,\!26$	0
3	CL	А	702	1/1	0.99	0.03	18,18,18,18	0

### 6.5 Other polymers (i)

There are no such residues in this entry.

