

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

#### Oct 8, 2023 – 12:47 AM JST

PDB ID : 8GYE

Title : Crystal Structure of the 4-1BB in complex with ZG033 Fab

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Deposited on : 2022-09-22

Resolution : 2.30 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 : 2.35.1

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

 $Refmac \quad : \quad 5.8.0158$ 

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

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

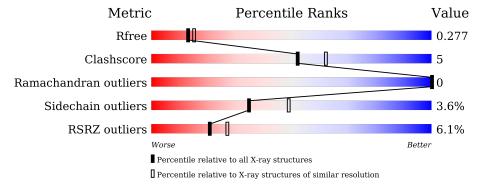
Validation Pipeline (wwPDB-VP) : 2.35.1

## 1 Overall quality at a glance (i)

The following experimental techniques were used to determine the structure: X-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	$\begin{array}{c} \textbf{Whole archive} \\ (\#\text{Entries}) \end{array}$	$\begin{array}{c} {\rm Similar\ resolution} \\ (\#{\rm Entries},\ {\rm resolution\ range}({\rm \AA})) \end{array}$
$R_{free}$	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	A	174	72%	6%		21%	_
1	D	174	9%	13%	•	21%	
2	В	220	<mark>6%</mark> 78%			18%	
2	Е	220	77%			20%	
3	С	214	84%			15%	
3	F	214	91%			8	%



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
5	GOL	E	301	-	-	-	X



# 2 Entry composition (i)

There are 6 unique types of molecules in this entry. The entry contains 8661 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 Tumor necrosis factor receptor superfamily member 9.

Mol	Chain	Residues	Atoms					ZeroOcc	AltConf	Trace
1	A	137	10001	C 597	11	O 203	S 22	0	0	0
1	D	138	Total 1019		N 191	O 204	S 22	0	0	0

There are 22 discrepancies between the modelled and reference sequences:

Chain	Residue	Modelled	Actual	Comment	Reference
A	187	LEU	-	expression tag	UNP Q07011
A	188	GLU	-	expression tag	UNP Q07011
A	189	GLY	-	expression tag	UNP Q07011
A	190	GLY	-	expression tag	UNP Q07011
A	191	GLY	-	expression tag	UNP Q07011
A	192	HIS	-	expression tag	UNP Q07011
A	193	HIS	-	expression tag	UNP Q07011
A	194	HIS	-	expression tag	UNP Q07011
A	195	HIS	-	expression tag	UNP Q07011
A	196	HIS	-	expression tag	UNP Q07011
A	197	HIS	-	expression tag	UNP Q07011
D	187	LEU	-	expression tag	UNP Q07011
D	188	GLU	-	expression tag	UNP Q07011
D	189	GLY	-	expression tag	UNP Q07011
D	190	GLY	-	expression tag	UNP Q07011
D	191	GLY	-	expression tag	UNP Q07011
D	192	HIS	-	expression tag	UNP Q07011
D	193	HIS	-	expression tag	UNP Q07011
D	194	HIS	-	expression tag	UNP Q07011
D	195	HIS	-	expression tag	UNP Q07011
D	196	HIS	-	expression tag	UNP Q07011
D	197	HIS	-	expression tag	UNP Q07011

• Molecule 2 is a protein called ZG033 Fab H chain.

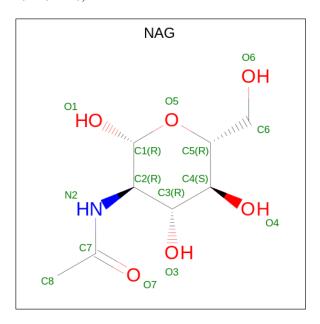


Mol	Chain	Residues	Atoms			ZeroOcc	AltConf	Trace		
2	В	216	Total 1593	C 999	1.4	O 321	S 7	0	0	0
2	Е	216	Total 1593	C 999	N 266	O 321	S 7	0	0	0

 $\bullet$  Molecule 3 is a protein called ZG033 Fab L chain.

Mol	Chain	Residues	Atoms				ZeroOcc	AltConf	Trace	
9	C	213	Total	С	N	О	S	0	0	0
3   0	213	1641	1028	271	337	5	0			
9	E	213	Total	С	N	О	S	0	0	0
)	Г	213	1641	1028	271	337	5	0		

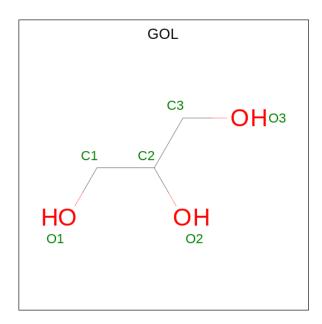
 $\bullet$  Molecule 4 is 2-acetamido-2-deoxy-beta-D-glucopyranose (three-letter code: NAG) (formula:  $\rm C_8H_{15}NO_6).$ 



Mol	Chain	Residues	Atoms	ZeroOcc	AltConf
4	A	1	Total C N O 14 8 1 5	0	0
4	D	1	Total C N O 14 8 1 5	0	0

• Molecule 5 is GLYCEROL (three-letter code: GOL) (formula: C<sub>3</sub>H<sub>8</sub>O<sub>3</sub>).





Mol	Chain	Residues	Atoms			ZeroOcc	AltConf
5	Е	1	Total 6	C 3	O 3	0	0

#### • Molecule 6 is water.

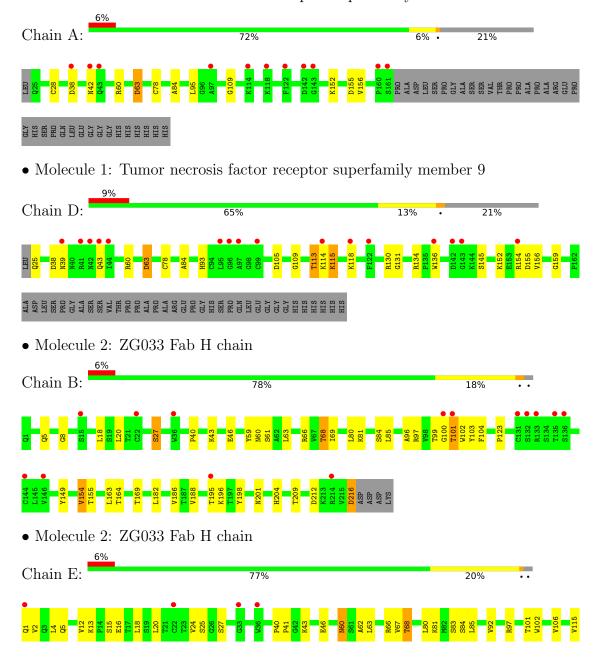
Mol	Chain	Residues	Atoms	ZeroOcc	AltConf
6	A	13	Total O 13 13	0	0
6	D	8	Total O 8 8	0	0
6	В	30	Total O 30 30	0	0
6	С	30	Total O 30 30	0	0
6	Е	28	Total O 28 28	0	0
6	F	19	Total O 19 19	0	0



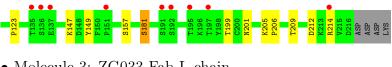
## 3 Residue-property plots (i)

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.

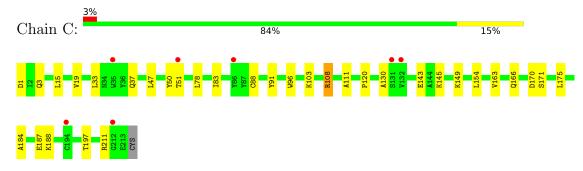
• Molecule 1: Tumor necrosis factor receptor superfamily member 9



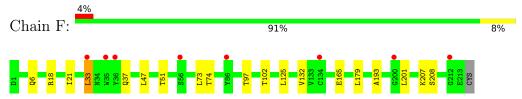




• Molecule 3: ZG033 Fab L chain



• Molecule 3: ZG033 Fab L chain





# 4 Data and refinement statistics (i)

Property	Value	Source
Space group	P 1	Depositor
Cell constants	60.02Å 65.91Å 83.64Å	Donositor
a, b, c, $\alpha$ , $\beta$ , $\gamma$	$90.00^{\circ}$ $82.50^{\circ}$ $74.82^{\circ}$	Depositor
Resolution (Å)	19.91 - 2.30	Depositor
rtesolution (A)	19.91 - 2.30	EDS
% Data completeness	97.3 (19.91-2.30)	Depositor
(in resolution range)	97.3 (19.91-2.30)	EDS
$R_{merge}$	0.09	Depositor
$R_{sym}$	(Not available)	Depositor
$< I/\sigma(I) > 1$	1.82 (at 2.30Å)	Xtriage
Refinement program	PHENIX 1.14-3260	Depositor
P. P.	0.222 , $0.277$	Depositor
$R, R_{free}$	0.222 , $0.277$	DCC
$R_{free}$ test set	2593 reflections $(4.90\%)$	wwPDB-VP
Wilson B-factor $(\mathring{A}^2)$	46.1	Xtriage
Anisotropy	0.466	Xtriage
Bulk solvent $k_{sol}(e/Å^3)$ , $B_{sol}(Å^2)$	0.29, 41.5	EDS
L-test for twinning <sup>2</sup>	$ < L >=0.50, < L^2>=0.33$	Xtriage
Estimated twinning fraction	No twinning to report.	Xtriage
$F_o, F_c$ correlation	0.95	EDS
Total number of atoms	8661	wwPDB-VP
Average B, all atoms (Å <sup>2</sup> )	62.0	wwPDB-VP

Xtriage's analysis on translational NCS is as follows: The largest off-origin peak in the Patterson function is 6.66% 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: NAG, GOL

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	Bond angles		
IVIOI		RMSZ	# Z  > 5	RMSZ	# Z  > 5		
1	A	0.25	0/1028	0.44	0/1381		
1	D	0.25	0/1036	0.45	0/1393		
2	В	0.25	0/1630	0.45	0/2229		
2	Е	0.25	0/1630	0.48	0/2229		
3	С	0.24	0/1677	0.45	0/2279		
3	F	0.25	0/1677	0.44	0/2279		
All	All	0.25	0/8678	0.45	0/11790		

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 maintenain group or atoms of a sidechain that are expected to be planar.

Mol	Chain	#Chirality outliers	#Planarity outliers
2	Ε	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	Е	15	SER	Peptide	

### 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



+ h = = = = = = = = = = = = = = = = = =	t	banaaa C-	rmana Clashas	lists stresses atm	rr malatad alaahaa
the asymme	tric unit,	wnereas 5	ymm-Clasnes	nsts symmetr	y-related clashes.

Mol	Chain	Non-H	H(model)	H(added)	Clashes	Symm-Clashes
1	A	1012	0	930	7	0
1	D	1019	0	937	12	0
2	В	1593	0	1574	23	0
2	Е	1593	0	1574	26	0
3	С	1641	0	1590	17	0
3	F	1641	0	1590	10	0
4	A	14	0	13	0	0
4	D	14	0	13	0	0
5	Ε	6	0	8	3	0
6	A	13	0	0	0	0
6	В	30	0	0	0	0
6	С	30	0	0	0	0
6	D	8	0	0	0	0
6	Ε	28	0	0	1	0
6	F	19	0	0	0	0
All	All	8661	0	8229	92	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 5.

The worst 5 of 92 close contacts within the same asymmetric unit are listed below, sorted by their clash magnitude.

Atom-1	Atom-2	$\begin{array}{c} {\rm Interatomic} \\ {\rm distance} \ ({\rm \AA}) \end{array}$	$\begin{array}{c} \text{Clash} \\ \text{overlap } (\text{\AA}) \end{array}$
2:E:16:GLU:HB3	2:E:85:LEU:H	1.32	0.94
2:E:66:ARG:HD2	2:E:84:SER:HB2	1.60	0.82
3:C:33:LEU:HB3	3:C:51:THR:HG22	1.74	0.67
1:D:115:LYS:HB2	1:D:118:LYS:HE3	1.77	0.67
2:B:27:SER:HB3	2:B:97:ARG:HH11	1.61	0.66

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 r	number of residues.
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Mol	Chain	Analysed	Favoured	Allowed	Outliers	Perce	ntiles
1	A	135/174 (78%)	135 (100%)	0	0	100	100
1	D	136/174 (78%)	133 (98%)	3 (2%)	0	100	100
2	В	214/220 (97%)	211 (99%)	3 (1%)	0	100	100
2	E	214/220 (97%)	210 (98%)	4 (2%)	0	100	100
3	С	211/214 (99%)	208 (99%)	3 (1%)	0	100	100
3	F	211/214 (99%)	207 (98%)	4 (2%)	0	100	100
All	All	1121/1216 (92%)	1104 (98%)	17 (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	Perce	ntiles
1	A	119/147 (81%)	117 (98%)	2 (2%)	60	76
1	D	120/147 (82%)	112 (93%)	8 (7%)	16	21
2	В	186/190 (98%)	176 (95%)	10 (5%)	22	30
2	E	186/190 (98%)	179 (96%)	7 (4%)	33	47
3	С	189/190 (100%)	184 (97%)	5 (3%)	46	63
3	F	189/190 (100%)	185 (98%)	4 (2%)	53	70
All	All	989/1054 (94%)	953 (96%)	36 (4%)	35	49

5 of 36 residues with a non-rotameric sidechain are listed below:

Mol	Chain	Res	Type
2	Ε	92	VAL
3	F	201	LEU
2	Е	181	SER
3	F	33	LEU
2	В	61	SER



Sometimes sidechains can be flipped to improve hydrogen bonding and reduce clashes. All (1) such sidechains are listed below:

Mol	Chain	Res	Type
1	D	42	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)

3 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	Chain	Chain	Chain	Chain	Chain	Chain	Chain	Chain	Chain	Chain	Chain	Chain	Chain	Chain	Chain	Chain	Chain	Chain	Chain	Chain	Chain	Res	Link	Bo	ond leng	ths	В	ond ang	les
WIOI	Mol Type Chain		nes	Lilik	Counts	RMSZ	# Z  > 2	Counts	RMSZ	# Z >2																					
5	GOL	Е	301	-	5,5,5	0.90	0	5,5,5	0.98	0																					
4	NAG	A	201	1	14,14,15	0.31	0	17,19,21	0.43	0																					
4	NAG	D	201	1	14,14,15	0.47	0	17,19,21	0.43	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
5	GOL	Е	301	-	-	0/4/4/4	-

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$\mathbf{Mol}$	$\mathbf{Type}$	Chain	$\operatorname{Res}$	Link	Chirals	Torsions	Rings
4	NAG	A	201	1	-	3/6/23/26	0/1/1/1
4	NAG	D	201	1	-	2/6/23/26	0/1/1/1

There are no bond length outliers.

There are no bond angle outliers.

There are no chirality outliers.

All (5) torsion outliers are listed below:

Mol	Chain	Res	Type	Atoms
4	A	201	NAG	O5-C5-C6-O6
4	D	201	NAG	C8-C7-N2-C2
4	D	201	NAG	O7-C7-N2-C2
4	A	201	NAG	C4-C5-C6-O6
4	A	201	NAG	C1-C2-N2-C7

There are no ring outliers.

1 monomer is involved in 3 short contacts:

Mol	Chain	Res	Type	Clashes	Symm-Clashes
5	E	301	GOL	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			$OWAB(A^2)$	Q<0.9
1	A	137/174 (78%)	0.42	11 (8%) 12 16	41, 64, 91, 99	0
1	D	138/174 (79%)	0.65	16 (11%) 4 6	45, 70, 99, 105	0
2	В	216/220 (98%)	0.37	14 (6%) 18 24	39, 56, 87, 121	0
2	E	216/220 (98%)	0.48	13 (6%) 21 28	41, 61, 99, 122	0
3	С	213/214 (99%)	0.35	7 (3%) 46 53	42, 58, 77, 85	0
3	F	213/214 (99%)	0.48	8 (3%) 40 47	43, 60, 85, 94	0
All	All	1133/1216 (93%)	0.45	69 (6%) 21 27	39, 60, 89, 122	0

The worst 5 of 69 RSRZ outliers are listed below:

Mol	Chain	Res	Type	RSRZ
2	В	135	THR	5.0
2	Ε	135	THR	5.0
1	A	42	ASN	4.2
1	D	142	ASP	3.8
2	В	101	THR	3.8

### 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	$\mathbf{B} ext{-}\mathbf{factors}(\mathbf{\mathring{A}}^2)$	Q<0.9
5	GOL	Е	301	6/6	0.61	0.43	52,59,65,79	0
4	NAG	D	201	14/15	0.67	0.26	83,96,105,109	0
4	NAG	A	201	14/15	0.81	0.24	63,90,94,99	0

## 6.5 Other polymers (i)

There are no such residues in this entry.

