

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

#### Aug 23, 2023 – 02:13 PM EDT

PDB ID : 3BE9

Title: Structure-based design and synthesis of novel macrocyclic pyrazolo[1,5-a]

[1,3,5] triazine compounds as potent inhibitors of protein kinase CK2 and their

anticancer activities

Authors: Nie, Z.; Perretta, C.; Erickson, P.; Margosiak, S.; Lu, J.; Averill, A.; Almassy,

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Deposited on : 2007-11-16

Resolution : 2.00 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) : NOT EXECUTED

EDS : NOT EXECUTED

buster-report : 1.1.7 (2018)

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

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

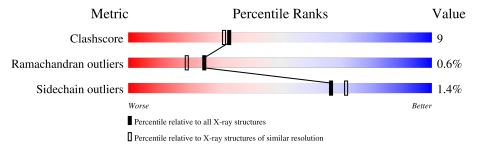
Validation Pipeline (wwPDB-VP) : 2.35

# 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 2.00 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	Similar resolution	
Metric	$(\# \mathrm{Entries})$	$(\# \text{Entries, resolution range}(\mathring{A}))$	
Clashscore	141614	9178 (2.00-2.00)	
Ramachandran outliers	138981	9054 (2.00-2.00)	
Sidechain outliers	138945	9053 (2.00-2.00)	

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%

Note EDS was not executed.

Mol	Chain	Length	Quality of chain				
1	A	352	77%	15%	• 7%		



# 2 Entry composition (i)

There are 3 unique types of molecules in this entry. The entry contains 3007 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 Casein kinase II subunit alpha.

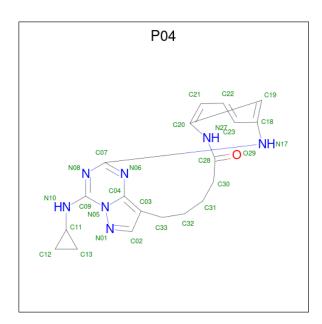
Mol	Chain	Residues	Atoms			ZeroOcc	AltConf	Trace		
1	٨	328	Total	С	N	О	S	0	0	0
1	A	320	2734	1760	471	491	12	U	U	

There are 21 discrepancies between the modelled and reference sequences:

Chain	Residue	Modelled	Actual	Comment	Reference
A	-14	MET	-	expression tag	UNP P28523
A	-13	GLY	-	expression tag	UNP P28523
A	-12	SER	-	expression tag	UNP P28523
A	-11	SER	-	expression tag	UNP P28523
A	-10	HIS	-	expression tag	UNP P28523
A	-9	HIS	-	expression tag	UNP P28523
A	-8	HIS	-	expression tag	UNP P28523
A	-7	HIS	-	expression tag	UNP P28523
A	-6	HIS	-	expression tag	UNP P28523
A	-5	HIS	-	expression tag	UNP P28523
A	-4	SER	-	expression tag	UNP P28523
A	-3	SER	-	expression tag	UNP P28523
A	-2	GLY	-	expression tag	UNP P28523
A	-1	LEU	-	expression tag	UNP P28523
A	0	VAL	-	expression tag	UNP P28523
A	1	PRO	-	expression tag	UNP P28523
A	2	ARG	-	expression tag	UNP P28523
A	3	GLY	-	expression tag	UNP P28523
A	4	SER	-	expression tag	UNP P28523
A	5	HIS		expression tag	UNP P28523
A	256	ALA	VAL	engineered mutation	UNP P28523

• Molecule 2 is 19-(cyclopropylamino)-4,6,7,15-tetrahydro-5H-16,1-(azenometheno)-10,14 -(metheno)pyrazolo[4,3-o][1,3,9]triazacyclohexadecin-8(9H)-one (three-letter code: P04) (formula:  $C_{19}H_{21}N_7O$ ).





Mol	Chain	Residues	Atoms				ZeroOcc	AltConf
9	٨	1	Total	С	N	О	0	0
	А	1	27	19	7	1	0	0

### • Molecule 3 is water.

Mol	Chain	Residues	Atoms	ZeroOcc	AltConf
3	A	246	Total O 246 246	0	0

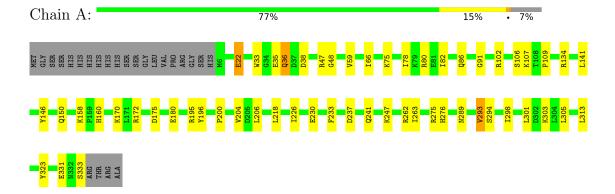


# 3 Residue-property plots (i)

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. 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. 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.

Note EDS was not executed.

• Molecule 1: Casein kinase II subunit alpha





# 4 Data and refinement statistics (i)

Xtriage (Phenix) and EDS were not executed - this section is therefore incomplete.

Property	Value	Source	
Space group	C 1 2 1	Depositor	
Cell constants	143.33Å 59.58Å 44.84Å	Depositor	
a, b, c, $\alpha$ , $\beta$ , $\gamma$	$90.00^{\circ}$ $103.42^{\circ}$ $90.00^{\circ}$	Depositor	
Resolution (Å)	32.70 - 2.00	Depositor	
% Data completeness	91.6 (32.70-2.00)	Depositor	
(in resolution range)	31.0 (32.10-2.00)	Depositor	
$R_{merge}$	0.08	Depositor	
$R_{sym}$	(Not available)	Depositor	
Refinement program	CNX	Depositor	
$R, R_{free}$	0.190 , 0.228	Depositor	
Estimated twinning fraction	No twinning to report.	Xtriage	
Total number of atoms	3007	wwPDB-VP	
Average B, all atoms (Å <sup>2</sup> )	26.0	wwPDB-VP	



# 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: CSO, P04

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).

Mal	Mol Chain		$\mathbf{lengths}$	Bond angles		
MIOI	Chain	RMSZ	# Z  > 5	RMSZ	# Z  > 5	
1	A	0.31	0/2794	0.56	0/3774	

There are no bond length outliers.

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	A	2734	0	2717	44	0
2	A	27	0	21	4	0
3	A	246	0	0	6	0
All	All	3007	0	2738	47	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 9.

All (47) close contacts within the same asymmetric unit are listed below, sorted by their clash magnitude.

Atom-1	Atom-2	$\begin{array}{c} {\rm Interatomic} \\ {\rm distance} \ ({\rm \AA}) \end{array}$	$\begin{array}{c} \text{Clash} \\ \text{overlap } (\text{\AA}) \end{array}$
1:A:35:GLU:HB3	1:A:38:ASP:OD2	1.80	0.81

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Continuea from prev		Interatomic	Clash	
Atom-1	Atom-2	${\rm distance}({\rm \AA})$	overlap $(\mathring{\mathrm{A}})$	
1:A:66:ILE:HG21	2:A:501:P04:H32A	1.76	0.67	
2:A:501:P04:H31	2:A:501:P04:H19	1.75	0.67	
1:A:102:ARG:NH2	1:A:107:LYS:HD2	2.13	0.63	
1:A:48:GLY:HA3	3:A:624:HOH:O	1.99	0.62	
1:A:160:HIS:H	1:A:160:HIS:CD2	2.17	0.62	
1:A:47:ARG:O	1:A:47:ARG:HD2	2.01	0.60	
1:A:134:ARG:HG2	1:A:323:TYR:CZ	2.39	0.58	
2:A:501:P04:H19	2:A:501:P04:N06	2.20	0.56	
1:A:294:SER:HB2	3:A:627:HOH:O	2.04	0.56	
1:A:33:TRP:CD2	1:A:102:ARG:HD2	2.42	0.55	
1:A:293:VAL:HG13	1:A:298:ILE:HD11	1.88	0.55	
1:A:47:ARG:HD2	1:A:47:ARG:C	2.27	0.54	
1:A:75:LYS:O	1:A:78:ILE:HG22	2.08	0.53	
1:A:293:VAL:HG12	1:A:293:VAL:O	2.08	0.52	
1:A:22:GLU:H	1:A:22:GLU:CD	2.13	0.52	
1:A:91:GLY:HA3	1:A:146:TYR:CE2	2.46	0.51	
1:A:170:LYS:HD2	1:A:172:ARG:NH2	2.27	0.50	
1:A:36:GLN:HB3	3:A:584:HOH:O	2.12	0.49	
1:A:233:PHE:HB3	1:A:241:GLN:HE22	1.78	0.48	
1:A:141:LEU:HD21	1:A:218:LEU:HD21	1.96	0.47	
1:A:293:VAL:CG1	1:A:298:ILE:HD11	2.44	0.47	
1:A:106:SER:O	1:A:107:LYS:HB2	2.14	0.47	
1:A:158:LYS:HE3	1:A:160:HIS:HB2	1.97	0.46	
1:A:275:ARG:HD3	1:A:276:HIS:N	2.31	0.46	
1:A:146:TYR:O	1:A:150:GLN:HG2	2.16	0.45	
1:A:241:GLN:HG2	3:A:733:HOH:O	2.17	0.45	
1:A:301:LEU:HD11	1:A:305:LEU:CD1	2.49	0.43	
1:A:206:LEU:HD12	1:A:263:ILE:HD11	2.00	0.43	
1:A:82:ILE:O	1:A:86:GLN:HG3	2.17	0.43	
1:A:247:LYS:NZ	1:A:247:LYS:CB	2.82	0.43	
1:A:80:ARG:HD3	3:A:729:HOH:O	2.19	0.42	
1:A:38:ASP:O	1:A:59:VAL:HG22	2.20	0.42	
2:A:501:P04:H19	2:A:501:P04:C31	2.47	0.42	
1:A:200:PRO:O	1:A:204:VAL:HG22	2.19	0.42	
1:A:262:ARG:NE	1:A:262:ARG:HA	2.34	0.42	
1:A:233:PHE:CB	1:A:241:GLN:HE22	2.32	0.42	
1:A:107:LYS:O	1:A:109:PRO:HD3	2.19	0.42	
1:A:303:LYS:HB3	1:A:313:LEU:HG	2.01	0.42	
1:A:331:GLU:C	1:A:333:SER:H	2.23	0.41	
1:A:141:LEU:HD21	1:A:218:LEU:CD2	2.50	0.41	
1:A:226:ILE:O	1:A:289:ASN:HB2	2.21	0.41	

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Atom-1	Atom-2	$\begin{array}{c} {\rm Interatomic} \\ {\rm distance} \ ({\rm \AA}) \end{array}$	$\begin{array}{c} \text{Clash} \\ \text{overlap } (\text{\AA}) \end{array}$
1:A:38:ASP:HA	1:A:59:VAL:HG22	2.03	0.41
1:A:275:ARG:HD3	1:A:275:ARG:C	2.42	0.40
1:A:237:ASP:HB2	3:A:626:HOH:O	2.20	0.40
1:A:33:TRP:CE2	1:A:102:ARG:HD2	2.57	0.40
1:A:195:ARG:O	1:A:196:TYR:CB	2.69	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	A	$325/352 \ (92\%)$	309 (95%)	14 (4%)	2 (1%)	25 19	

#### All (2) Ramachandran outliers are listed below:

Mol	Chain	Res	Type
1	A	175	ASP
1	A	293	VAL

#### 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	295/315 (94%)	291 (99%)	4 (1%)	67 72	



A 11	( 4 )	• 1	• . 1				. 1 1 .		1. / 1	1 1
AH	4)	residiles	with	a	non-rotameric	3 8	sidechain	are	listed	below:

Mol	Chain	Res	Type
1	A	22	GLU
1	A	36	GLN
1	A	180	GLU
1	A	230	GLU

Sometimes sidechains can be flipped to improve hydrogen bonding and reduce clashes. All (6) such sidechains are listed below:

Mol	Chain	Res	Type
1	A	160	HIS
1	A	241	GLN
1	A	255	ASN
1	A	268	GLN
1	A	310	GLN
1	A	326	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)

1 non-standard protein/DNA/RNA residue is 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	Mol Type Chain Res Link			Link	В	ond leng	${ m gths}$	Bond angles		
	IVIOI	Type	Chain	nes	Link	Counts	RMSZ	# Z  > 2	Counts	RMSZ	# Z >2
Ī	1	CSO	A	89	1	3,6,7	0.53	0	0,6,8	-	-

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
1	CSO	A	89	1	-	1/1/5/7	-

There are no bond length outliers.

There are no bond angle outliers.

There are no chirality outliers.

All (1) torsion outliers are listed below:

Mol	Chain	Res	Type	Atoms
1	A	89	CSO	N-CA-CB-SG

There are no ring outliers.

No monomer is involved in short contacts.

## 5.5 Carbohydrates (i)

There are no monosaccharides in this entry.

## 5.6 Ligand geometry (i)

1 ligand is 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	Res	Link	Bond lengths			Bond angles		
IVIOI					Counts	RMSZ	# Z  > 2	Counts	RMSZ	# Z  > 2
2	P04	A	501	-	25,31,31	1.03	3 (12%)	31,44,44	2.00	8 (25%)

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	P04	A	501	-	-	4/19/21/21	0/4/5/5



All (3) bond length outliers are listed below:

Mol	Chain	Res	Type	Atoms	$\mathbf{Z}$	Observed(A)	$\operatorname{Ideal}(\text{\AA})$
2	A	501	P04	C20-N27	-2.55	1.36	1.41
2	A	501	P04	C12-C11	2.23	1.53	1.48
2	A	501	P04	C13-C11	2.21	1.53	1.48

All (8) bond angle outliers are listed below:

Mol	Chain	Res	Type	Atoms	$\mathbf{Z}$	$Observed(^o)$	$Ideal(^{o})$
2	A	501	P04	C12-C11-N10	-5.16	109.76	118.52
2	A	501	P04	C13-C11-N10	-4.76	110.43	118.52
2	A	501	P04	C30-C28-N27	3.55	120.84	114.59
2	A	501	P04	O29-C28-C30	-2.73	117.02	122.02
2	A	501	P04	C20-N27-C28	2.68	132.18	127.50
2	A	501	P04	C02-N01-N05	2.64	108.11	103.94
2	A	501	P04	C07-N06-C04	2.29	117.88	115.28
2	A	501	P04	C09-N08-C07	2.25	118.16	115.14

There are no chirality outliers.

All (4) torsion outliers are listed below:

Mol	Chain	Res	Type	Atoms
2	A	501	P04	C04-C03-C33-C32
2	A	501	P04	O29-C28-N27-C20
2	A	501	P04	C30-C28-N27-C20
2	A	501	P04	C31-C32-C33-C03

There are no ring outliers.

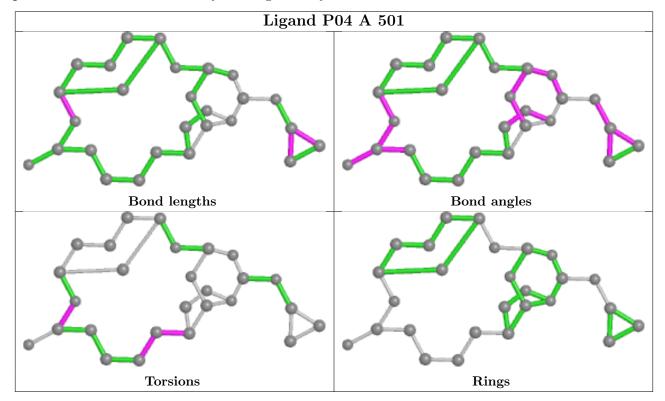
1 monomer is involved in 4 short contacts:

Mol	Chain	Res	Type	Clashes	Symm-Clashes
2	A	501	P04	4	0

The following is a two-dimensional graphical depiction of Mogul quality analysis of bond lengths, bond angles, torsion angles, and ring geometry for all instances of the Ligand of Interest. In addition, ligands with molecular weight > 250 and outliers as shown on the validation Tables will also be included. For torsion angles, if less then 5% of the Mogul distribution of torsion angles is within 10 degrees of the torsion angle in question, then that torsion angle is considered an outlier. Any bond that is central to one or more torsion angles identified as an outlier by Mogul will be highlighted in the graph. For rings, the root-mean-square deviation (RMSD) between the ring in question and similar rings identified by Mogul is calculated over all ring torsion angles. If the average RMSD is greater than 60 degrees and the minimal RMSD between the ring in question and



any Mogul-identified rings is also greater than 60 degrees, then that ring is considered an outlier. The outliers are highlighted in purple. The color gray indicates Mogul did not find sufficient equivalents in the CSD to analyse the geometry.



# 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)

EDS was not executed - this section is therefore empty.

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

EDS was not executed - this section is therefore empty.

## 6.3 Carbohydrates (i)

EDS was not executed - this section is therefore empty.

## 6.4 Ligands (i)

EDS was not executed - this section is therefore empty.

## 6.5 Other polymers (i)

EDS was not executed - this section is therefore empty.

