



wwPDB NMR Structure Validation Summary Report ⓘ

Dec 25, 2024 – 01:01 AM EST

PDB ID : 2N55
BMRB ID : 25694
Title : Structure of constitutively monomeric CXCL12 in complex with the CXCR4 N-terminus
Authors : Ziarek, J.J.; Peterson, F.C.; Volkman, B.F.
Deposited on : 2015-07-07

This is a wwPDB NMR 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/NMRValidationReportHelp>

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
Percentile statistics : 20231227.v01 (using entries in the PDB archive December 27th 2023)
wwPDB-RCI : v_1n_11_5_13_A (Berjanski et al., 2005)
PANAV : Wang et al. (2010)
wwPDB-ShiftChecker : v1.2
BMRB Restraints Analysis : v1.2
Ideal geometry (proteins) : Engh & Huber (2001)
Ideal geometry (DNA, RNA) : Parkinson et al. (1996)
Validation Pipeline (wwPDB-VP) : 2.40

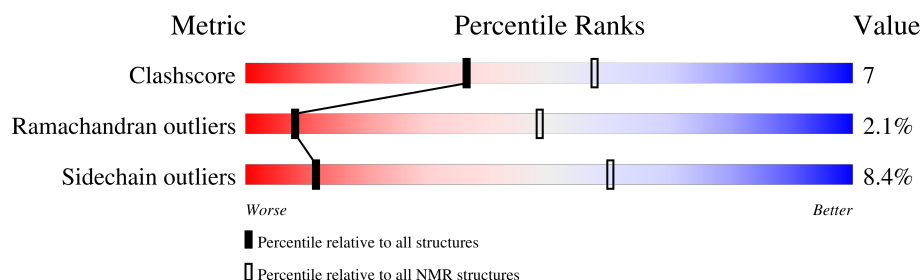
1 Overall quality at a glance

The following experimental techniques were used to determine the structure:

SOLUTION NMR

The overall completeness of chemical shifts assignment is 89%.

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)	NMR archive (#Entries)
Clashscore	210492	14027
Ramachandran outliers	207382	12486
Sidechain outliers	206894	12463

The table below summarises the geometric issues observed across the polymeric chains and their fit to the experimental data. The red, orange, yellow and green segments indicate the fraction of residues that contain outliers for ≥ 3 , 2, 1 and 0 types of geometric quality criteria. A cyan segment indicates the fraction of residues that are not part of the well-defined cores, and 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 $\leq 5\%$

Mol	Chain	Length	Quality of chain
1	A	70	
2	B	40	

2 Ensemble composition and analysis

This entry contains 20 models. Model 5 is the overall representative, medoid model (most similar to other models). The authors have identified model 1 as representative, based on the following criterion: *lowest energy*.

The following residues are included in the computation of the global validation metrics.

Well-defined (core) protein residues			
Well-defined core	Residue range (total)	Backbone RMSD (Å)	Medoid model
1	A:7-A:67, B:204-B:226 (84)	1.10	5

Ill-defined regions of proteins are excluded from the global statistics.

Ligands and non-protein polymers are included in the analysis.

The models can be grouped into 3 clusters and 3 single-model clusters were found.

Cluster number	Models
1	1, 2, 3, 5, 6, 8, 12, 13
2	10, 14, 17, 18, 19
3	4, 7, 11, 16
Single-model clusters	9; 15; 20

3 Entry composition

There are 2 unique types of molecules in this entry. The entry contains 1741 atoms, of which 860 are hydrogens and 0 are deuteriums.

- Molecule 1 is a protein called Stromal cell-derived factor 1.

Mol	Chain	Residues	Atoms						Trace
1	A	70	Total	C	H	N	O	S	0
			1153	357	587	108	94	7	

There are 4 discrepancies between the modelled and reference sequences:

Chain	Residue	Modelled	Actual	Comment	Reference
A	-1	GLY	-	expression tag	UNP P48061
A	0	MET	-	expression tag	UNP P48061
A	55	CYS	LEU	engineered mutation	UNP P48061
A	58	CYS	ILE	engineered mutation	UNP P48061

- Molecule 2 is a protein called C-X-C chemokine receptor type 4.

Mol	Chain	Residues	Atoms						Trace
2	B	40	Total	C	H	N	O	S	0
			588	190	273	49	73	3	

There are 3 discrepancies between the modelled and reference sequences:

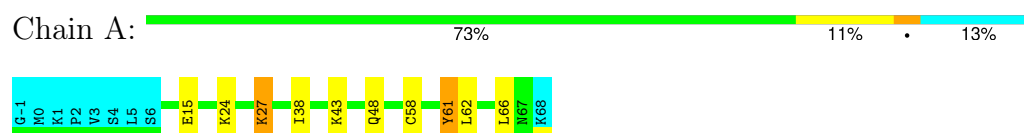
Chain	Residue	Modelled	Actual	Comment	Reference
B	199	GLY	-	expression tag	UNP P61073
B	200	SER	-	expression tag	UNP P61073
B	228	ALA	CYS	engineered mutation	UNP P61073

4 Residue-property plots [i](#)

4.1 Average score per residue in the NMR ensemble

These plots are provided for all protein, RNA, DNA and oligosaccharide chains in the entry. The first graphic is the same as shown in the summary in section 1 of this report. The second graphic shows the sequence where residues are colour-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 outliers are shown as green connectors. Residues which are classified as ill-defined in the NMR ensemble, are shown in cyan with an underline colour-coded according to the previous scheme. Residues which were present in the experimental sample, but not modelled in the final structure are shown in grey.

- Molecule 1: Stromal cell-derived factor 1



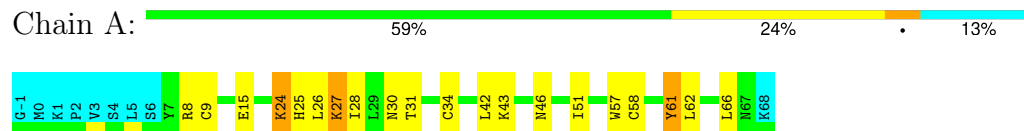
- Molecule 2: C-X-C chemokine receptor type 4



4.2 Residue scores for the representative (medoid) model from the NMR ensemble

The representative model is number 5. Colouring as in section 4.1 above.

- Molecule 1: Stromal cell-derived factor 1



- Molecule 2: C-X-C chemokine receptor type 4



5 Refinement protocol and experimental data overview

The models were refined using the following method: *torsion angle dynamics, molecular dynamics*.

Of the 100 calculated structures, 20 were deposited, based on the following criterion: *target function*.

The following table shows the software used for structure solution, optimisation and refinement.

Software name	Classification	Version
CYANA	structure solution	
CYANA	refinement	
X-PLOR NIH	geometry optimization	
X-PLOR NIH	refinement	

The following table shows chemical shift validation statistics as aggregates over all chemical shift files. Detailed validation can be found in section 7 of this report.

Chemical shift file(s)	working_cs.cif
Number of chemical shift lists	1
Total number of shifts	1315
Number of shifts mapped to atoms	1315
Number of unparsed shifts	0
Number of shifts with mapping errors	0
Number of shifts with mapping warnings	0
Assignment completeness (well-defined parts)	89%

6 Model quality [i](#)

6.1 Standard geometry [i](#)

There are no covalent bond-length or bond-angle outliers.

There are no bond-length outliers.

There are no bond-angle outliers.

There are no chirality outliers.

There are no planarity outliers.

6.2 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 each 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 averaged over the ensemble.

Mol	Chain	Non-H	H(model)	H(added)	Clashes
1	A	502	510	508	10±3
2	B	182	156	156	3±2
All	All	13680	13320	13280	202

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 7.

5 of 97 unique clashes are listed below, sorted by their clash magnitude.

Atom-1	Atom-2	Clash(Å)	Distance(Å)	Models	
				Worst	Total
1:A:27:LYS:HG3	2:B:207:TYR:CE1	0.66	2.25	12	16
1:A:26:LEU:HD22	1:A:61:TYR:CE2	0.66	2.26	16	10
1:A:58:CYS:O	1:A:62:LEU:HG	0.62	1.95	17	13
1:A:52:ASP:OD2	1:A:54:LYS:HB3	0.60	1.96	13	1
1:A:42:LEU:O	1:A:46:ASN:HA	0.59	1.98	5	6

6.3 Torsion angles [i](#)

6.3.1 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 PDB entries followed by that with respect to all NMR

entries. 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	61/70 (87%)	55±2 (90±3%)	5±2 (9±3%)	0±0 (1±1%)	19	69
2	B	23/40 (58%)	18±1 (79±5%)	4±1 (15±6%)	1±1 (6±5%)	2	21
All	All	1680/2200 (76%)	1464 (87%)	180 (11%)	36 (2%)	8	48

5 of 13 unique Ramachandran outliers are listed below. They are sorted by the frequency of occurrence in the ensemble.

Mol	Chain	Res	Type	Models (Total)
2	B	217	GLY	5
1	A	43	LYS	4
2	B	223	SER	4
2	B	205	SER	4
2	B	218	SER	4

6.3.2 Protein sidechains ⓘ

In the following table, the Percentiles column shows the percent sidechain outliers of the chain as a percentile score with respect to all PDB entries followed by that with respect to all NMR entries. 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	56/64 (88%)	51±2 (91±3%)	5±2 (9±3%)	10	56
2	B	21/34 (62%)	20±1 (94±4%)	1±1 (6±4%)	20	72
All	All	1540/1960 (79%)	1411 (92%)	129 (8%)	11	60

5 of 25 unique residues with a non-rotameric sidechain are listed below. They are sorted by the frequency of occurrence in the ensemble.

Mol	Chain	Res	Type	Models (Total)
1	A	24	LYS	20
1	A	27	LYS	20
1	A	61	TYR	20
1	A	15	GLU	15
2	B	212	TYR	10

6.3.3 RNA [i](#)

There are no RNA molecules in this entry.

6.4 Non-standard residues in protein, DNA, RNA chains [i](#)

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

6.5 Carbohydrates [i](#)

There are no oligosaccharides in this entry.

6.6 Ligand geometry [i](#)

There are no ligands in this entry.

6.7 Other polymers [i](#)

There are no such molecules in this entry.

6.8 Polymer linkage issues [i](#)

There are no chain breaks in this entry.

7 Chemical shift validation

The completeness of assignment taking into account all chemical shift lists is 89% for the well-defined parts and 88% for the entire structure.

7.1 Chemical shift list 1

File name: working_cs.cif

Chemical shift list name: *assigned_chem_shift_list_1*

7.1.1 Bookkeeping

The following table shows the results of parsing the chemical shift list and reports the number of nuclei with statistically unusual chemical shifts.

Total number of shifts	1315
Number of shifts mapped to atoms	1315
Number of unparsed shifts	0
Number of shifts with mapping errors	0
Number of shifts with mapping warnings	0
Number of shift outliers (ShiftChecker)	1

7.1.2 Chemical shift referencing

The following table shows the suggested chemical shift referencing corrections.

Nucleus	# values	Correction \pm precision, ppm	Suggested action
$^{13}\text{C}_\alpha$	110	-0.34 ± 0.11	None needed (< 0.5 ppm)
$^{13}\text{C}_\beta$	104	-0.09 ± 0.09	None needed (< 0.5 ppm)
$^{13}\text{C}'$	99	-0.07 ± 0.06	None needed (< 0.5 ppm)
^{15}N	99	0.09 ± 0.25	None needed (< 0.5 ppm)

7.1.3 Completeness of resonance assignments

The following table shows the completeness of the chemical shift assignments for the well-defined regions of the structure. The overall completeness is 89%, i.e. 1033 atoms were assigned a chemical shift out of a possible 1165. 0 out of 10 assigned methyl groups (LEU and VAL) were assigned stereospecifically.

	Total	^1H	^{13}C	^{15}N
Backbone	406/416 (98%)	165/167 (99%)	162/168 (96%)	79/81 (98%)
Sidechain	565/656 (86%)	386/421 (92%)	169/202 (84%)	10/33 (30%)

Continued on next page...

Continued from previous page...

	Total	¹ H	¹³ C	¹⁵ N
Aromatic	62/93 (67%)	38/44 (86%)	23/44 (52%)	1/5 (20%)
Overall	1033/1165 (89%)	589/632 (93%)	354/414 (86%)	90/119 (76%)

7.1.4 Statistically unusual chemical shifts [i](#)

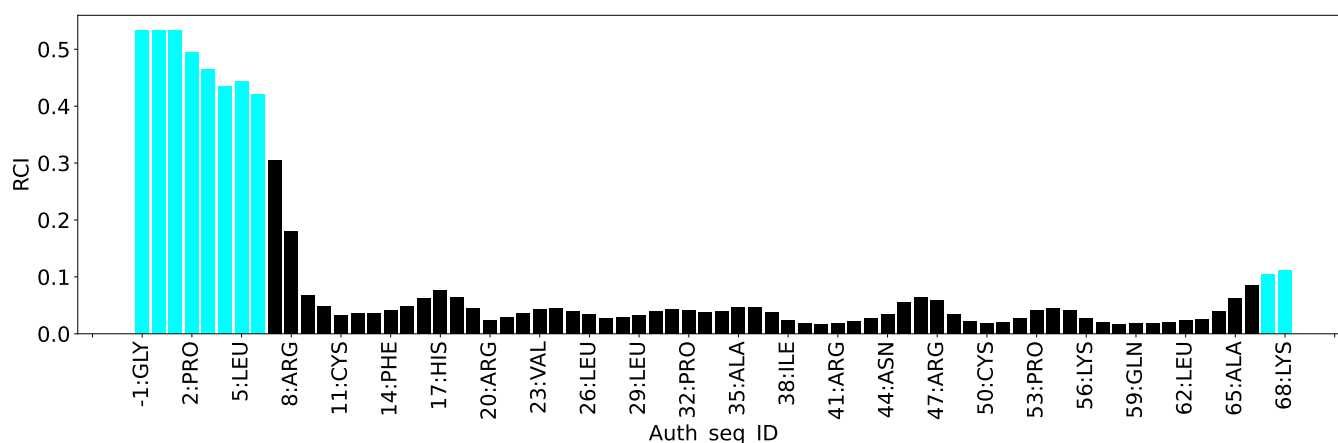
The following table lists the statistically unusual chemical shifts. These are statistical measures, and large deviations from the mean do not necessarily imply incorrect assignments. Molecules containing paramagnetic centres or hemes are expected to give rise to anomalous chemical shifts.

List Id	Chain	Res	Type	Atom	Shift, ppm	Expected range, ppm	Z-score
1	A	20	ARG	HB3	0.20	0.43 – 3.11	-5.8

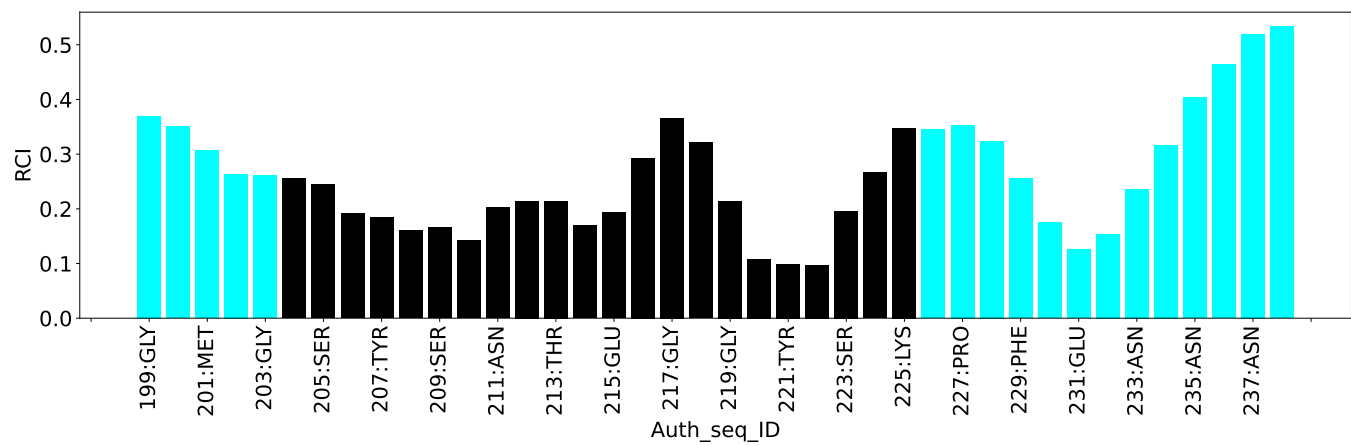
7.1.5 Random Coil Index (RCI) plots [i](#)

The image below reports *random coil index* values for the protein chains in the structure. The height of each bar gives a probability of a given residue to be disordered, as predicted from the available chemical shifts and the amino acid sequence. A value above 0.2 is an indication of significant predicted disorder. The colour of the bar shows whether the residue is in the well-defined core (black) or in the ill-defined residue ranges (cyan), as described in section 2 on ensemble composition. If well-defined core and ill-defined regions are not identified then it is shown as gray bars.

Random coil index (RCI) for chain A:



Random coil index (RCI) for chain B:



8 NMR restraints analysis

8.1 Conformationally restricting restraints

The following table provides the summary of experimentally observed NMR restraints in different categories. Restraints are classified into different categories based on the sequence separation of the atoms involved.

Description	Value
Total distance restraints	2807
Intra-residue ($ i-j =0$)	647
Sequential ($ i-j =1$)	818
Medium range ($ i-j >1$ and $ i-j <5$)	398
Long range ($ i-j \geq 5$)	798
Inter-chain	140
Hydrogen bond restraints	0
Disulfide bond restraints	6
Total dihedral-angle restraints	218
Number of unmapped restraints	0
Number of restraints per residue	27.5
Number of long range restraints per residue ¹	7.3

¹Long range hydrogen bonds and disulfide bonds are counted as long range restraints while calculating the number of long range restraints per residue

8.2 Residual restraint violations

This section provides the overview of the restraint violations analysis. The violations are binned as small, medium and large violations based on its absolute value. Average number of violations per model is calculated by dividing the total number of violations in each bin by the size of the ensemble.

8.2.1 Average number of distance violations per model

Distance violations less than 0.1 Å are not included in the calculation.

Bins (Å)	Average number of violations per model	Max (Å)
0.1-0.2 (Small)	27.9	0.2
0.2-0.5 (Medium)	2.6	0.49
>0.5 (Large)	0.4	1.02

8.2.2 Average number of dihedral-angle violations per model [i](#)

Dihedral-angle violations less than 1° are not included in the calculation.

Bins (°)	Average number of violations per model	Max (°)
1.0-10.0 (Small)	24.6	4.11
10.0-20.0 (Medium)	None	None
>20.0 (Large)	None	None

9 Distance violation analysis [i](#)

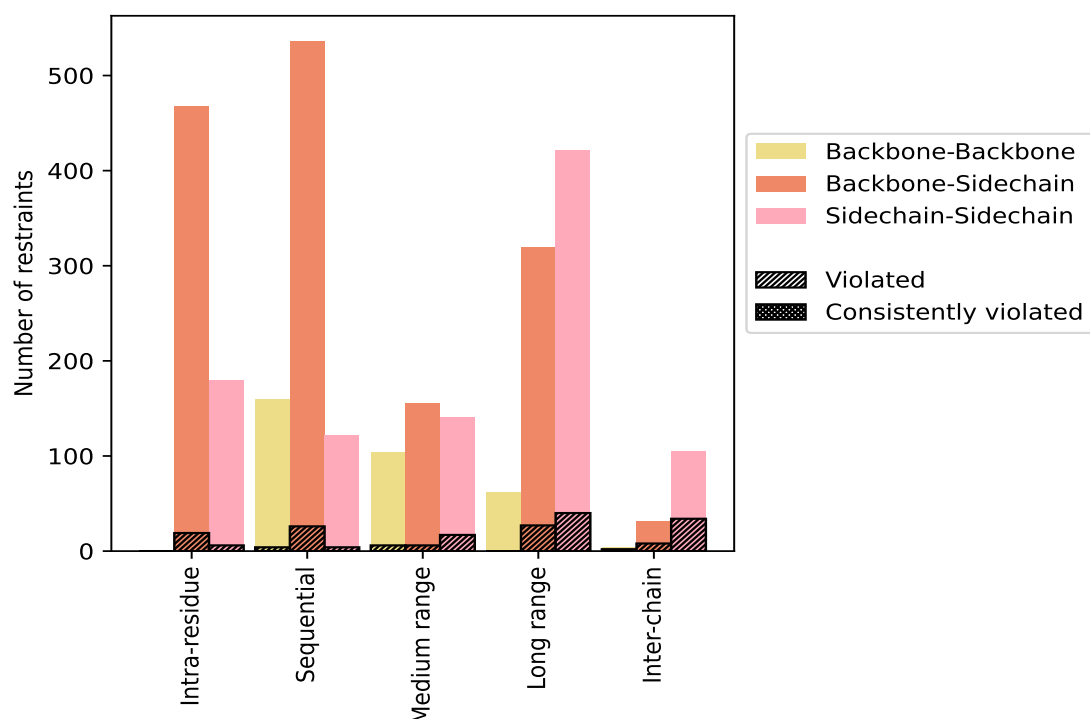
9.1 Summary of distance violations [i](#)

The following table shows the summary of distance violations in different restraint categories based on the sequence separation of the atoms involved. Each category is further sub-divided into three sub-categories based on the atoms involved. Violations less than 0.1 Å are not included in the statistics.

Restrains type	Count	% ¹	Violated ³			Consistently Violated ⁴		
			Count	% ²	% ¹	Count	% ²	% ¹
Intra-residue (i-j =0)	647	23.0	25	3.9	0.9	0	0.0	0.0
Backbone-Backbone	0	0.0	0	0.0	0.0	0	0.0	0.0
Backbone-Sidechain	468	16.7	19	4.1	0.7	0	0.0	0.0
Sidechain-Sidechain	179	6.4	6	3.4	0.2	0	0.0	0.0
Sequential (i-j =1)	818	29.1	34	4.2	1.2	0	0.0	0.0
Backbone-Backbone	160	5.7	4	2.5	0.1	0	0.0	0.0
Backbone-Sidechain	536	19.1	26	4.9	0.9	0	0.0	0.0
Sidechain-Sidechain	122	4.3	4	3.3	0.1	0	0.0	0.0
Medium range (i-j >1 & i-j <5)	398	14.2	29	7.3	1.0	0	0.0	0.0
Backbone-Backbone	104	3.7	6	5.8	0.2	0	0.0	0.0
Backbone-Sidechain	155	5.5	6	3.9	0.2	0	0.0	0.0
Sidechain-Sidechain	139	5.0	17	12.2	0.6	0	0.0	0.0
Long range (i-j ≥5)	798	28.4	67	8.4	2.4	0	0.0	0.0
Backbone-Backbone	62	2.2	0	0.0	0.0	0	0.0	0.0
Backbone-Sidechain	319	11.4	27	8.5	1.0	0	0.0	0.0
Sidechain-Sidechain	417	14.9	40	9.6	1.4	0	0.0	0.0
Inter-chain	140	5.0	44	31.4	1.6	0	0.0	0.0
Backbone-Backbone	4	0.1	2	50.0	0.1	0	0.0	0.0
Backbone-Sidechain	31	1.1	8	25.8	0.3	0	0.0	0.0
Sidechain-Sidechain	105	3.7	34	32.4	1.2	0	0.0	0.0
Hydrogen bond	0	0.0	0	0.0	0.0	0	0.0	0.0
Disulfide bond	6	0.2	0	0.0	0.0	0	0.0	0.0
Total	2807	100.0	199	7.1	7.1	0	0.0	0.0
Backbone-Backbone	330	11.8	12	3.6	0.4	0	0.0	0.0
Backbone-Sidechain	1509	53.8	86	5.7	3.1	0	0.0	0.0
Sidechain-Sidechain	968	34.5	101	10.4	3.6	0	0.0	0.0

¹ percentage calculated with respect to the total number of distance restraints, ² percentage calculated with respect to the number of restraints in a particular restraint category, ³ violated in at least one model, ⁴ violated in all the models

9.1.1 Bar chart : Distribution of distance restraints and violations [i](#)



Violated and consistently violated restraints are shown using different hatch patterns in their respective categories. The hydrogen bonds and disulfied bonds are counted in their appropriate category on the x-axis

9.2 Distance violation statistics for each model [i](#)

The following table provides the distance violation statistics for each model in the ensemble. Violations less than 0.1 Å are not included in the statistics.

Model ID	Number of violations						Mean (Å)	Max (Å)	SD ⁶ (Å)	Median (Å)
	IR ¹	SQ ²	MR ³	LR ⁴	IC ⁵	Total				
1	7	5	2	12	7	33	0.14	0.51	0.07	0.13
2	6	4	7	9	12	38	0.14	0.64	0.08	0.12
3	4	4	0	16	12	36	0.14	0.41	0.05	0.13
4	5	3	0	11	4	23	0.15	0.21	0.04	0.13
5	8	3	4	13	12	40	0.15	0.58	0.08	0.13
6	5	3	7	5	8	28	0.15	0.37	0.05	0.14
7	2	3	10	10	9	34	0.14	0.57	0.08	0.12
8	4	3	1	17	6	31	0.15	0.41	0.06	0.13
9	2	5	3	10	6	26	0.17	0.96	0.16	0.13
10	4	1	4	15	13	37	0.13	0.54	0.07	0.11

Continued on next page...

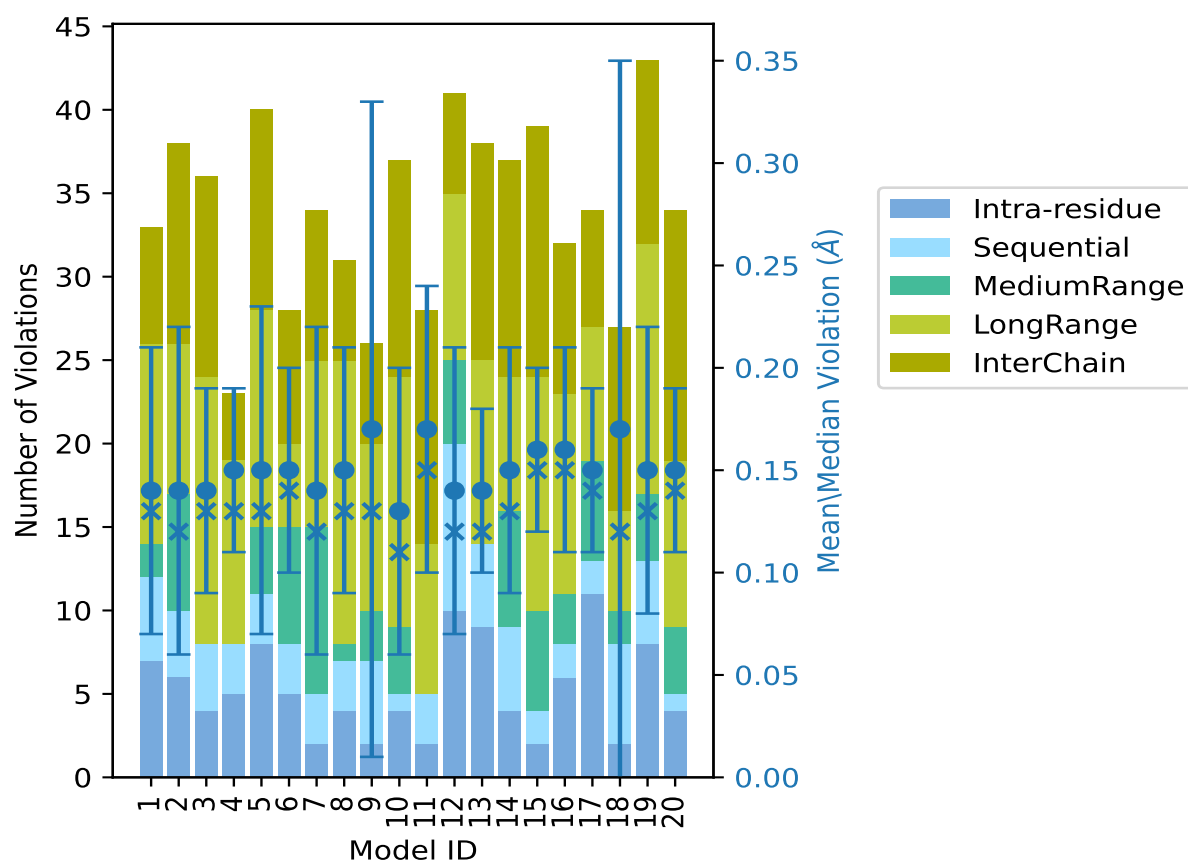
Continued from previous page...

Model ID	Number of violations						Mean (Å)	Max (Å)	SD ⁶ (Å)	Median (Å)
	IR ¹	SQ ²	MR ³	LR ⁴	IC ⁵	Total				
11	2	3	0	9	14	28	0.17	0.44	0.07	0.15
12	10	10	5	10	6	41	0.14	0.44	0.07	0.12
13	9	5	0	11	13	38	0.14	0.24	0.04	0.12
14	4	5	7	8	13	37	0.15	0.49	0.06	0.13
15	2	2	6	14	15	39	0.16	0.28	0.04	0.15
16	6	2	3	12	9	32	0.16	0.37	0.05	0.15
17	11	2	6	8	7	34	0.15	0.22	0.04	0.14
18	2	6	2	6	11	27	0.17	1.02	0.18	0.12
19	8	5	4	15	11	43	0.15	0.52	0.07	0.13
20	4	1	4	10	15	34	0.15	0.28	0.04	0.14

¹Intra-residue restraints, ²Sequential restraints, ³Medium range restraints, ⁴Long range restraints,

⁵Inter-chain restraints, ⁶Standard deviation

9.2.1 Bar graph : Distance Violation statistics for each model ⓘ



The mean(dot),median(x) and the standard deviation are shown in blue with respect to the y axis on the right

9.3 Distance violation statistics for the ensemble

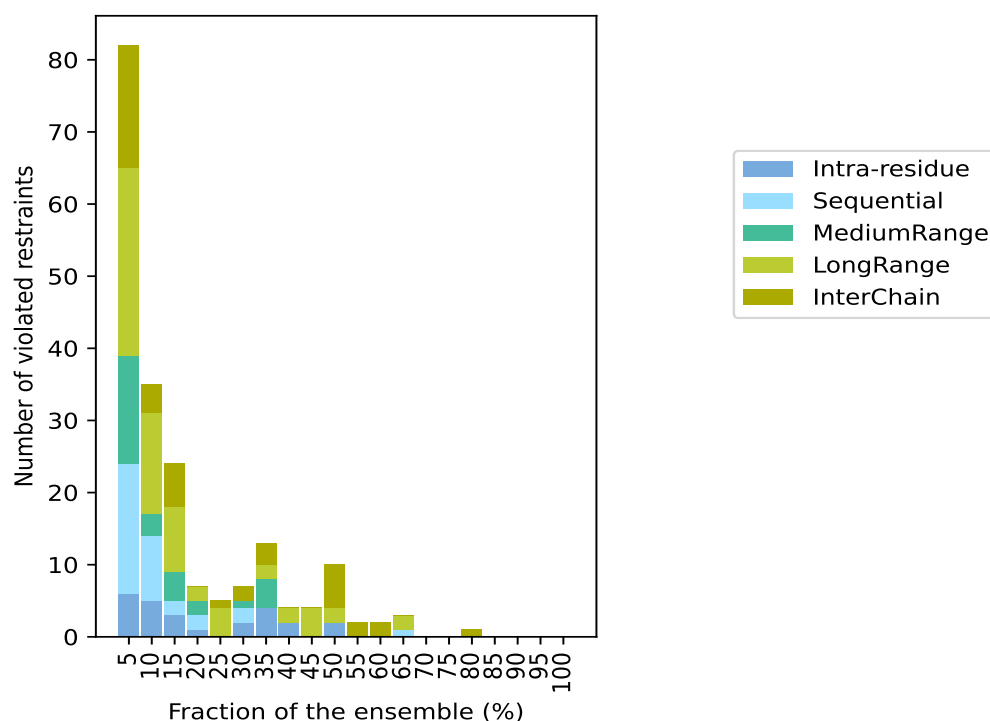
Violation analysis may find that some restraints are violated in few models and some are violated in most of models. The following table provides this information as number of violated restraints for a given fraction of the ensemble. In total, 2602(IR:622, SQ:784, MR:369, LR:731, IC:96) restraints are not violated in the ensemble.

Number of violated restraints						Fraction of the ensemble	
IR ¹	SQ ²	MR ³	LR ⁴	IC ⁵	Total	Count ⁶	%
6	18	15	26	17	82	1	5.0
5	9	3	14	4	35	2	10.0
3	2	4	9	6	24	3	15.0
1	2	2	2	0	7	4	20.0
0	0	0	4	1	5	5	25.0
2	2	1	0	2	7	6	30.0
4	0	4	2	3	13	7	35.0
2	0	0	2	0	4	8	40.0
0	0	0	4	0	4	9	45.0
2	0	0	2	6	10	10	50.0
0	0	0	0	2	2	11	55.0
0	0	0	0	2	2	12	60.0
0	1	0	2	0	3	13	65.0
0	0	0	0	0	0	14	70.0
0	0	0	0	0	0	15	75.0
0	0	0	0	1	1	16	80.0
0	0	0	0	0	0	17	85.0
0	0	0	0	0	0	18	90.0
0	0	0	0	0	0	19	95.0
0	0	0	0	0	0	20	100.0

¹Intra-residue restraints, ²Sequential restraints, ³Medium range restraints, ⁴Long range restraints,

⁵Inter-chain restraints, ⁶ Number of models with violations

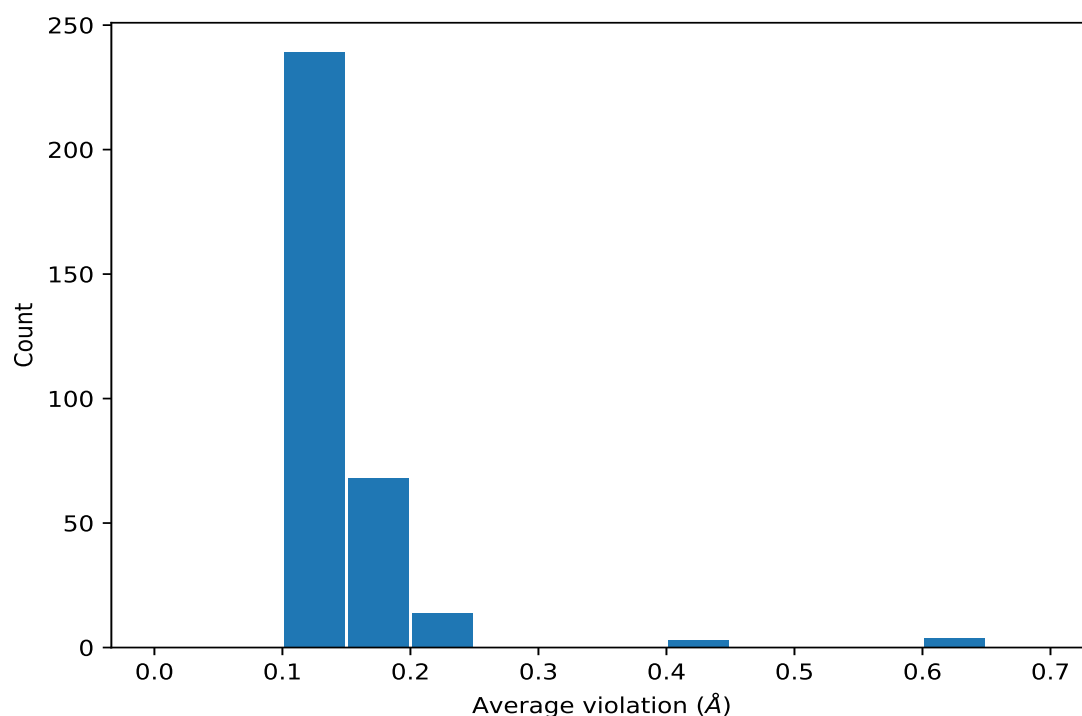
9.3.1 Bar graph : Distance violation statistics for the ensemble [i](#)



9.4 Most violated distance restraints in the ensemble [i](#)

9.4.1 Histogram : Distribution of mean distance violations [i](#)

The following histogram shows the distribution of the average value of the violation. The average is calculated for each restraint that is violated in more than one model over all the violated models in the ensemble



9.4.2 Table: Most violated distance restraints [i](#)

The following table provides the mean and the standard deviation of the violations for the 10 worst performing restraints, sorted by number of violated models and the mean violation value. The Key (restraint list ID, restraint ID) is the unique identifier for a given restraint. Rows with same key represent combinatorial or ambiguous restraints and are counted as a single restraint.

Key	Atom-1	Atom-2	Models ¹	Mean (Å)	SD ¹ (Å)	Median (Å)
(3,992)	1:61:A:TYR:HB3	2:204:B:ILE:HG22	16	0.43	0.13	0.41
(3,992)	1:61:A:TYR:HB3	2:204:B:ILE:HG21	16	0.43	0.13	0.41
(3,992)	1:61:A:TYR:HB3	2:204:B:ILE:HG23	16	0.43	0.13	0.41
(1,1027)	1:30:A:ASN:HD21	1:65:A:ALA:HB1	13	0.15	0.03	0.16
(1,1027)	1:30:A:ASN:HD21	1:65:A:ALA:HB2	13	0.15	0.03	0.16
(1,1027)	1:30:A:ASN:HD21	1:65:A:ALA:HB3	13	0.15	0.03	0.16
(3,575)	1:30:A:ASN:HD21	1:65:A:ALA:HB1	13	0.15	0.03	0.16
(3,575)	1:30:A:ASN:HD21	1:65:A:ALA:HB2	13	0.15	0.03	0.16
(3,575)	1:30:A:ASN:HD21	1:65:A:ALA:HB3	13	0.15	0.03	0.16
(1,563)	1:54:A:LYS:HB3	1:55:A:CYS:H	13	0.15	0.03	0.14
(1,1025)	1:68:A:LYS:H	2:207:B:TYR:HA	12	0.16	0.05	0.14
(3,1089)	1:68:A:LYS:H	2:207:B:TYR:HA	12	0.16	0.05	0.14
(1,1333)	1:49:A:VAL:HG11	2:221:B:TYR:HD1	11	0.14	0.04	0.13
(1,1333)	1:49:A:VAL:HG11	2:221:B:TYR:HD2	11	0.14	0.04	0.13
(1,1333)	1:49:A:VAL:HG12	2:221:B:TYR:HD1	11	0.14	0.04	0.13
(1,1333)	1:49:A:VAL:HG12	2:221:B:TYR:HD2	11	0.14	0.04	0.13

Continued on next page...

Continued from previous page...

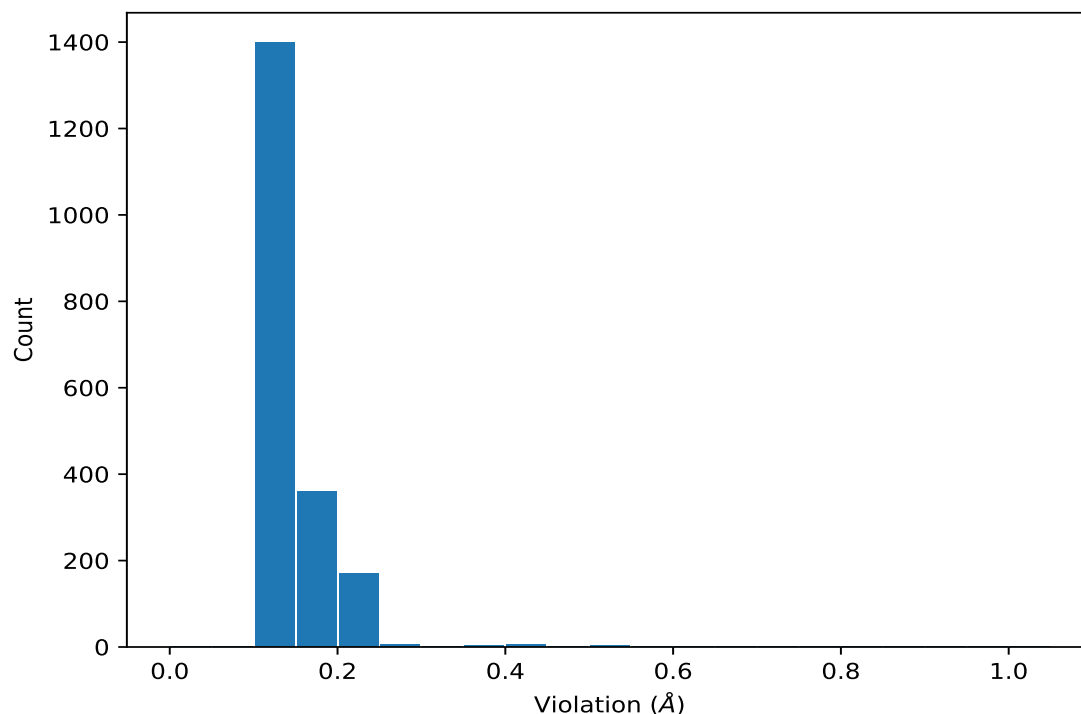
Key	Atom-1	Atom-2	Models ¹	Mean (Å)	SD ¹ (Å)	Median (Å)
(1,1333)	1:49:A:VAL:HG13	2:221:B:TYR:HD1	11	0.14	0.04	0.13
(1,1333)	1:49:A:VAL:HG13	2:221:B:TYR:HD2	11	0.14	0.04	0.13
(3,815)	1:49:A:VAL:HG11	2:221:B:TYR:HD1	11	0.14	0.04	0.13
(3,815)	1:49:A:VAL:HG11	2:221:B:TYR:HD2	11	0.14	0.04	0.13
(3,815)	1:49:A:VAL:HG12	2:221:B:TYR:HD1	11	0.14	0.04	0.13
(3,815)	1:49:A:VAL:HG12	2:221:B:TYR:HD2	11	0.14	0.04	0.13
(3,815)	1:49:A:VAL:HG13	2:221:B:TYR:HD1	11	0.14	0.04	0.13
(3,815)	1:49:A:VAL:HG13	2:221:B:TYR:HD2	11	0.14	0.04	0.13
(1,1316)	1:27:A:LYS:HE2	2:207:B:TYR:HE1	10	0.18	0.04	0.2
(1,1316)	1:27:A:LYS:HE2	2:207:B:TYR:HE2	10	0.18	0.04	0.2
(1,1316)	1:27:A:LYS:HE3	2:207:B:TYR:HE1	10	0.18	0.04	0.2
(1,1316)	1:27:A:LYS:HE3	2:207:B:TYR:HE2	10	0.18	0.04	0.2
(3,481)	1:27:A:LYS:HE2	2:207:B:TYR:HE1	10	0.18	0.04	0.2

¹Number of violated models, ²Standard deviation

9.5 All violated distance restraints [i](#)

9.5.1 Histogram : Distribution of distance violations [i](#)

The following histogram shows the distribution of the absolute value of the violation for all violated restraints in the ensemble.



9.5.2 Table : All distance violations

The following table provides the 10 worst performing restraints, sorted by the violation value. The Key (restraint list ID, restraint ID) is the unique identifier for a given restraint. Rows with same key represent combinatorial or ambiguous restraints and are counted as a single restraint.

Key	Atom-1	Atom-2	Model ID	Violation (Å)
(3,172)	1:16:A:SER:HB3	2:224:B:MET:HB2	18	1.02
(3,172)	1:16:A:SER:HB3	2:224:B:MET:HG2	9	0.96
(3,992)	1:61:A:TYR:HB3	2:204:B:ILE:HG22	2	0.64
(3,992)	1:61:A:TYR:HB3	2:204:B:ILE:HG23	5	0.58
(3,992)	1:61:A:TYR:HB3	2:204:B:ILE:HG22	7	0.57
(3,992)	1:61:A:TYR:HB3	2:204:B:ILE:HG23	10	0.54
(3,992)	1:61:A:TYR:HB3	2:204:B:ILE:HG22	19	0.52
(3,992)	1:61:A:TYR:HB3	2:204:B:ILE:HG22	1	0.51
(3,992)	1:61:A:TYR:HB3	2:204:B:ILE:HG23	14	0.49
(3,172)	1:16:A:SER:HB3	2:224:B:MET:HB3	11	0.44

10 Dihedral-angle violation analysis [i](#)

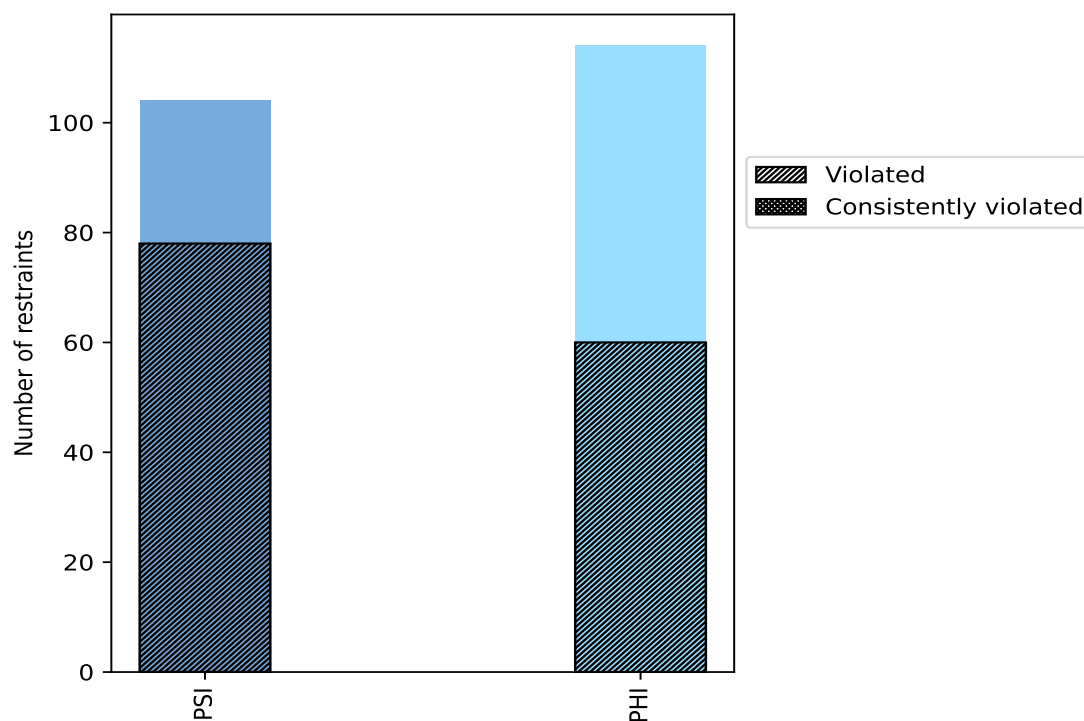
10.1 Summary of dihedral-angle violations [i](#)

The following table provides the summary of dihedral-angle violations in different dihedral-angle types. Violations less than 1° are not included in the calculation.

Angle type	Count	% ¹	Violated ³			Consistently Violated ⁴		
			Count	% ²	% ¹	Count	% ²	% ¹
PSI	104	47.7	78	75.0	35.8	0	0.0	0.0
PHI	114	52.3	60	52.6	27.5	0	0.0	0.0
Total	218	100.0	138	63.3	63.3	0	0.0	0.0

¹ percentage calculated with respect to total number of dihedral-angle restraints, ² percentage calculated with respect to number of restraints in a particular dihedral-angle type, ³ violated in at least one model, ⁴ violated in all the models

10.1.1 Bar chart : Distribution of dihedral-angles and violations [i](#)



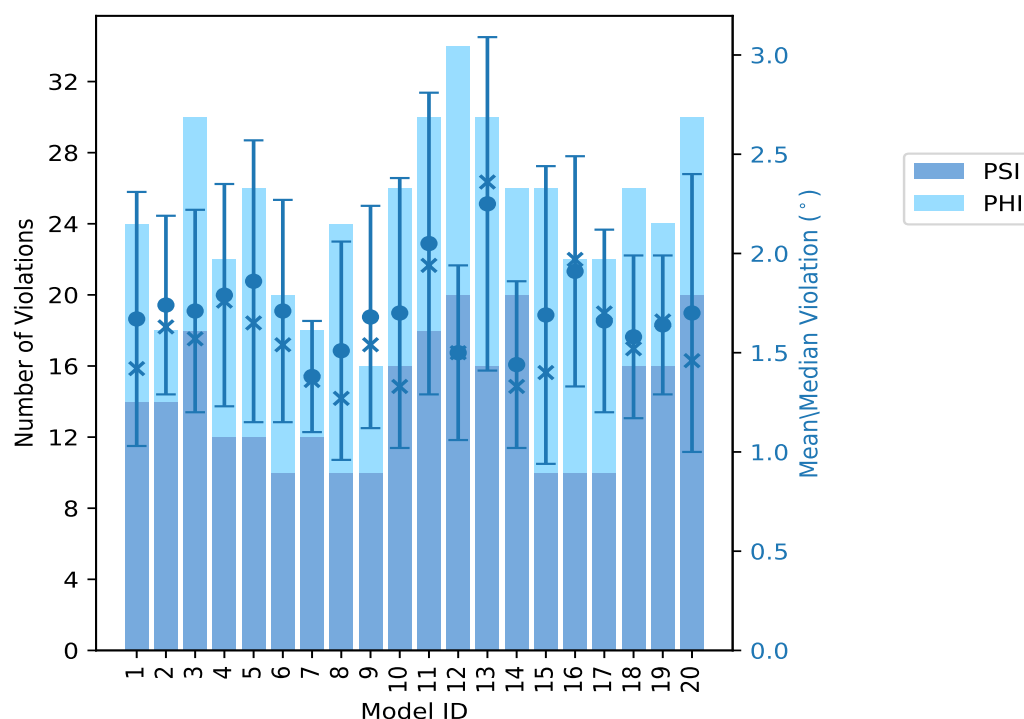
Violated and consistently violated restraints are shown using different hatch patterns in their respective categories

10.2 Dihedral-angle violation statistics for each model

The following table provides the dihedral-angle violation statistics for each model in the ensemble. Violations less than 1° are not included in the statistics.

Model ID	Number of violations			Mean (°)	Max (°)	SD (°)	Median (°)
	PSI	PHI	Total				
1	14	10	24	1.67	3.12	0.64	1.42
2	14	4	18	1.74	2.87	0.45	1.63
3	18	12	30	1.71	2.85	0.51	1.57
4	12	10	22	1.79	2.92	0.56	1.76
5	12	14	26	1.86	3.44	0.71	1.65
6	10	10	20	1.71	2.65	0.56	1.54
7	12	6	18	1.38	2.01	0.28	1.36
8	10	14	24	1.51	2.6	0.55	1.27
9	10	6	16	1.68	2.91	0.56	1.54
10	16	10	26	1.7	3.26	0.68	1.33
11	18	12	30	2.05	3.82	0.76	1.94
12	20	14	34	1.5	2.51	0.44	1.5
13	16	14	30	2.25	4.11	0.84	2.36
14	20	6	26	1.44	2.52	0.42	1.33
15	10	16	26	1.69	3.51	0.75	1.4
16	10	12	22	1.91	2.92	0.58	1.97
17	10	12	22	1.66	2.33	0.46	1.7
18	16	10	26	1.58	2.66	0.41	1.52
19	16	8	24	1.64	2.2	0.35	1.66
20	20	10	30	1.7	3.18	0.7	1.46

10.2.1 Bar graph : Dihedral violation statistics for each model [i](#)



The mean(dot),median(x) and the standard deviation are shown in blue with respect to the y axis on the right

10.3 Dihedral-angle violation statistics for the ensemble [i](#)

Violation analysis may find that some restraints are violated in very few models and some are violated in most of models. The following table provides this information as number of violated restraints for a given fraction of ensemble.

Number of violated restraints			Fraction of the ensemble	
PSI	PHI	Total	Count ¹	%
18	16	34	1	5.0
20	18	38	2	10.0
14	2	16	3	15.0
12	8	20	4	20.0
6	2	8	5	25.0
0	4	4	6	30.0
0	2	2	7	35.0
2	4	6	8	40.0
0	2	2	9	45.0
0	0	0	10	50.0
0	2	2	11	55.0

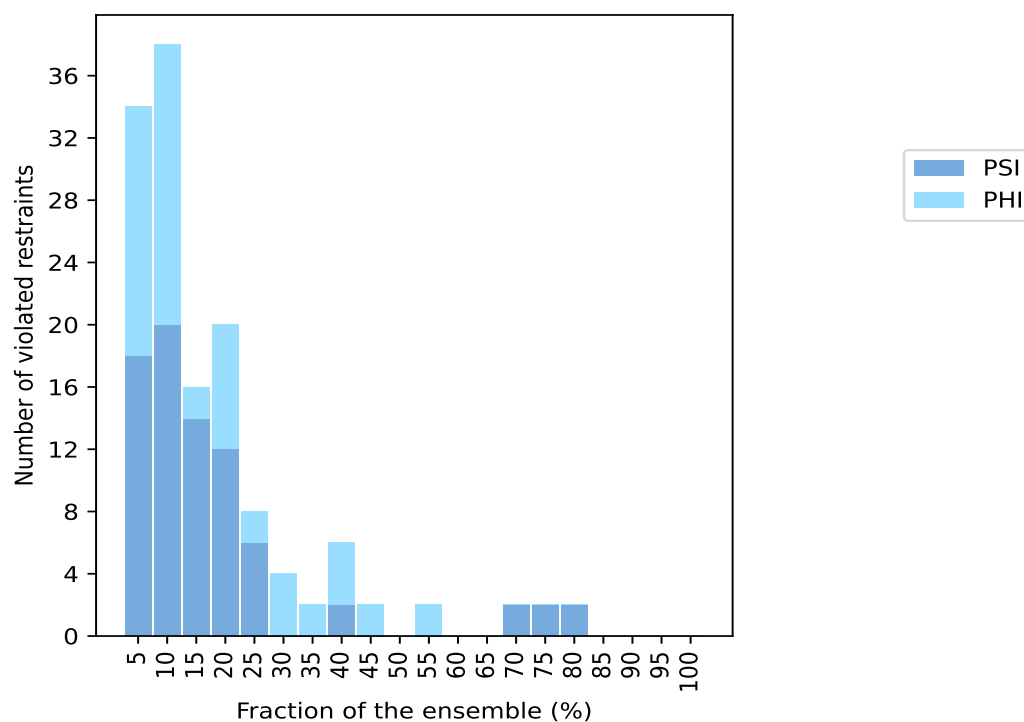
Continued on next page...

Continued from previous page...

Number of violated restraints			Fraction of the ensemble	
PSI	PHI	Total	Count ¹	%
0	0	0	12	60.0
0	0	0	13	65.0
2	0	2	14	70.0
2	0	2	15	75.0
2	0	2	16	80.0
0	0	0	17	85.0
0	0	0	18	90.0
0	0	0	19	95.0
0	0	0	20	100.0

¹ Number of models with violations

10.3.1 Bar graph : Dihedral-angle Violation statistics for the ensemble [i](#)

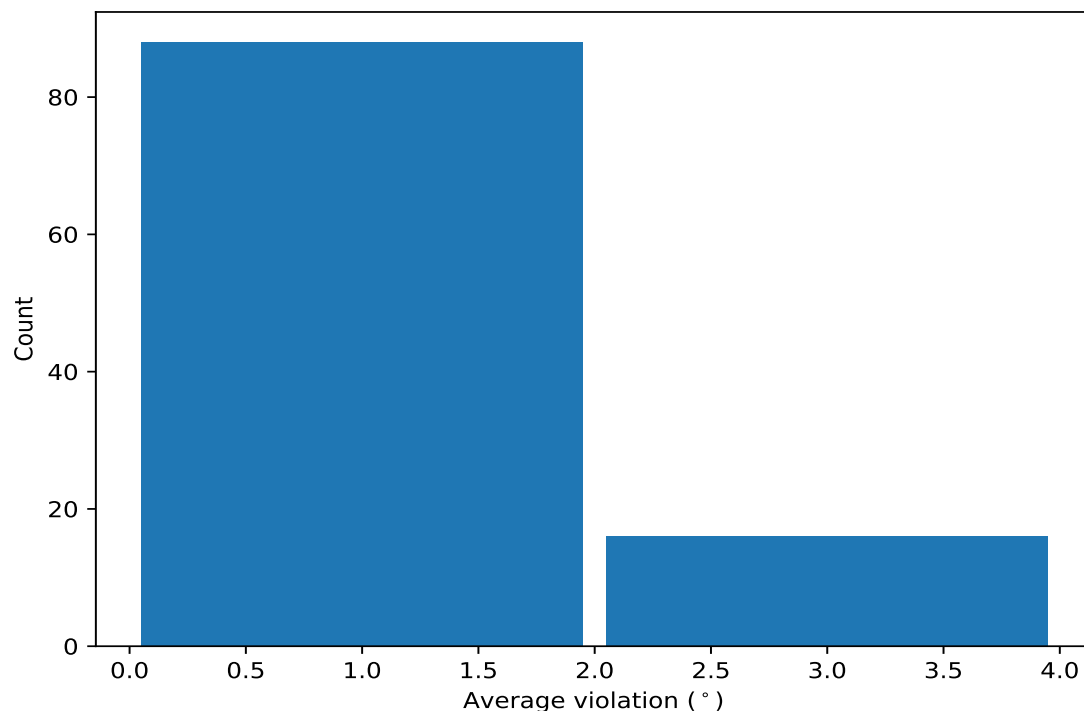


10.4 Most violated dihedral-angle restraints in the ensemble [i](#)

10.4.1 Histogram : Distribution of mean dihedral-angle violations [i](#)

The following histogram shows the distribution of the average value of the violation. The average is calculated for each restraint that is violated in more than one model over all the violated models

in the ensemble



10.4.2 Table: Most violated dihedral-angle restraints [i](#)

The following table provides the mean and the standard deviation of the violations for the 10 worst performing restraints, sorted by number of violated models and the mean violation value. The Key (restraint list ID, restraint ID) is the unique identifier for a given restraint.

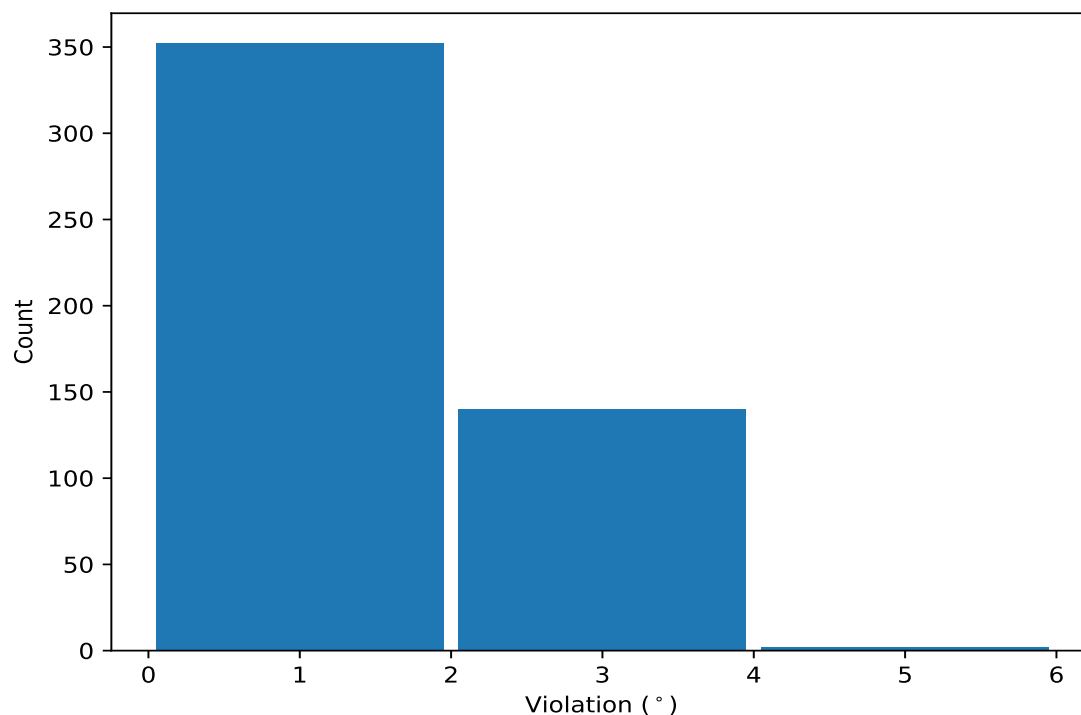
Key	Atom-1	Atom-2	Atom-3	Atom-4	Models ¹	Mean	SD ²	Median
(1,75)	1:58:A:CYS:N	1:58:A:CYS:CA	1:58:A:CYS:C	1:59:A:GLN:N	16	1.91	0.57	1.98
(2,75)	1:58:A:CYS:N	1:58:A:CYS:CA	1:58:A:CYS:C	1:59:A:GLN:N	16	1.91	0.57	1.98
(1,71)	1:56:A:LYS:N	1:56:A:LYS:CA	1:56:A:LYS:C	1:57:A:TRP:N	15	1.59	0.39	1.72
(2,71)	1:56:A:LYS:N	1:56:A:LYS:CA	1:56:A:LYS:C	1:57:A:TRP:N	15	1.59	0.39	1.72
(1,14)	1:19:A:ALA:N	1:19:A:ALA:CA	1:19:A:ALA:C	1:20:A:ARG:N	14	2.03	0.67	2.04
(2,14)	1:19:A:ALA:N	1:19:A:ALA:CA	1:19:A:ALA:C	1:20:A:ARG:N	14	2.03	0.67	2.04
(1,37)	1:34:A:CYS:C	1:35:A:ALA:N	1:35:A:ALA:CA	1:35:A:ALA:C	11	2.4	0.56	2.48
(2,37)	1:34:A:CYS:C	1:35:A:ALA:N	1:35:A:ALA:CA	1:35:A:ALA:C	11	2.4	0.56	2.48
(1,4)	1:11:A:CYS:C	1:12:A:ARG:N	1:12:A:ARG:CA	1:12:A:ARG:C	9	1.71	0.51	1.75
(2,4)	1:11:A:CYS:C	1:12:A:ARG:N	1:12:A:ARG:CA	1:12:A:ARG:C	9	1.71	0.51	1.75

¹ Number of violated models, ²Standard deviation, All angle values are in degree (°)

10.5 All violated dihedral-angle restraints [i](#)

10.5.1 Histogram : Distribution of violations [i](#)

The following histogram shows the distribution of the absolute value of the violation for all violated restraints in the ensemble.



10.5.2 Table: All violated dihedral-angle restraints [i](#)

The following table provides the list of violations for the 10 worst performing restraints, sorted by the violation value. The Key (restraint list ID, restraint ID) is the unique identifier for a given restraint.

Key	Atom-1	Atom-2	Atom-3	Atom-4	Model ID	Violation (°)
(2,52)	1:42:A:LEU:C	1:43:A:LYS:N	1:43:A:LYS:CA	1:43:A:LYS:C	13	4.11
(1,52)	1:42:A:LEU:C	1:43:A:LYS:N	1:43:A:LYS:CA	1:43:A:LYS:C	13	4.11
(2,58)	1:47:A:ARG:N	1:47:A:ARG:CA	1:47:A:ARG:C	1:48:A:GLN:N	11	3.82
(1,58)	1:47:A:ARG:N	1:47:A:ARG:CA	1:47:A:ARG:C	1:48:A:GLN:N	11	3.82
(2,97)	2:210:B:ASP:N	2:210:B:ASP:CA	2:210:B:ASP:C	2:211:B:ASN:N	15	3.51
(1,97)	2:210:B:ASP:N	2:210:B:ASP:CA	2:210:B:ASP:C	2:211:B:ASN:N	15	3.51
(2,52)	1:42:A:LEU:C	1:43:A:LYS:N	1:43:A:LYS:CA	1:43:A:LYS:C	5	3.44
(1,52)	1:42:A:LEU:C	1:43:A:LYS:N	1:43:A:LYS:CA	1:43:A:LYS:C	5	3.44
(2,106)	2:222:B:ASP:N	2:222:B:ASP:CA	2:222:B:ASP:C	2:223:B:SER:N	10	3.26
(1,106)	2:222:B:ASP:N	2:222:B:ASP:CA	2:222:B:ASP:C	2:223:B:SER:N	10	3.26