Anirudh Rao
BE21B004

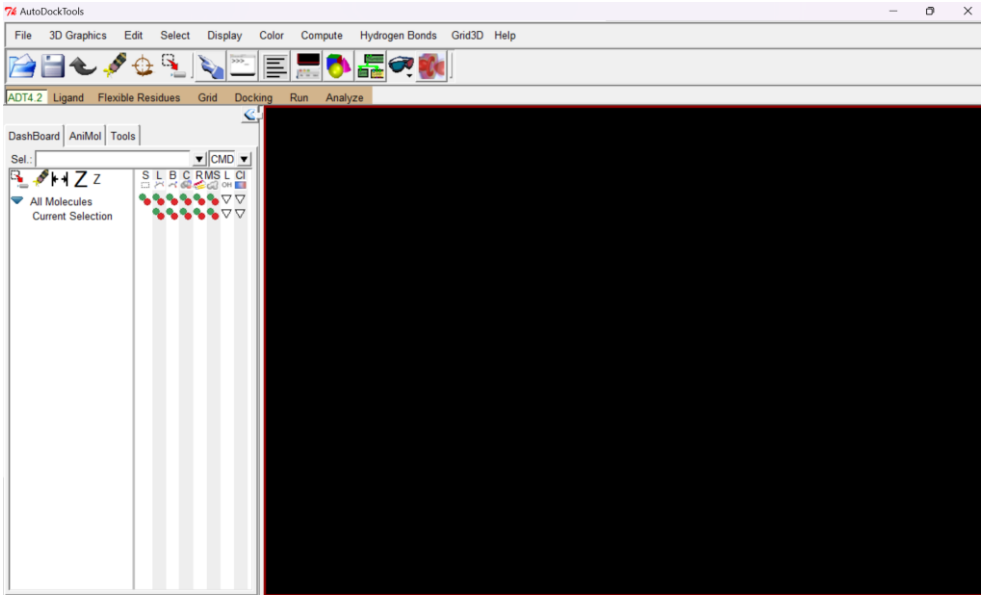
BT4110 – Computational Biology Lab

Practical 3

Rigid Docking

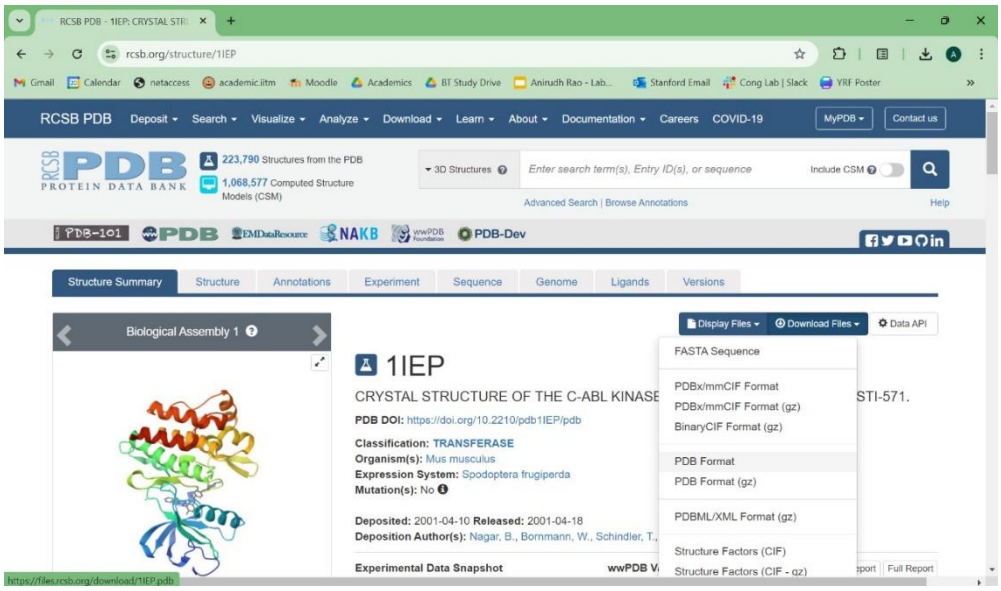
1

AutoDock was installed.



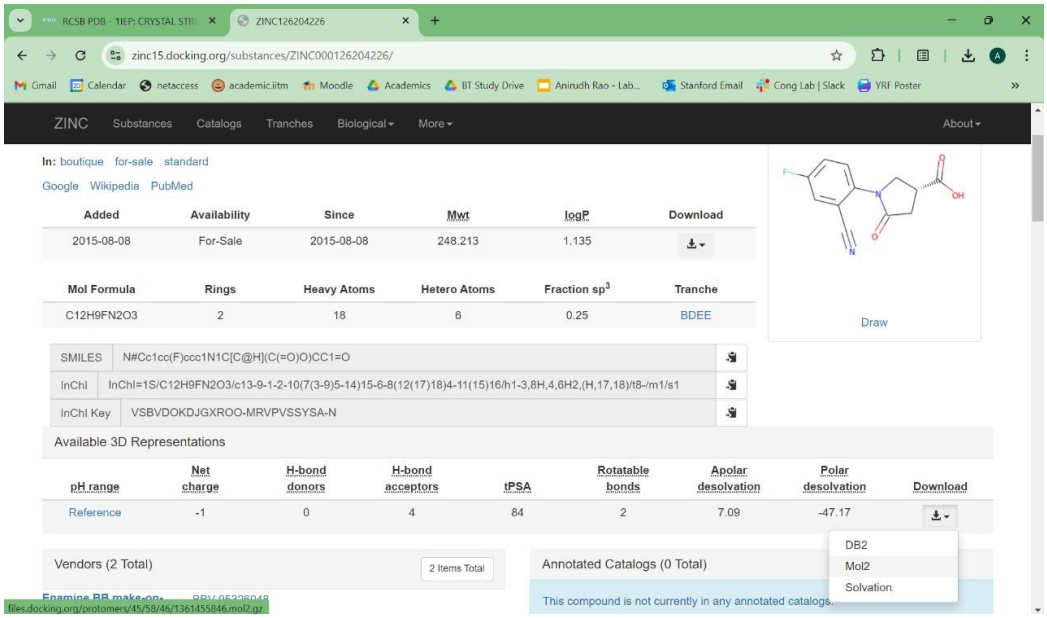
2

The structure of 1IEP was obtained from PDB in .pdb format.



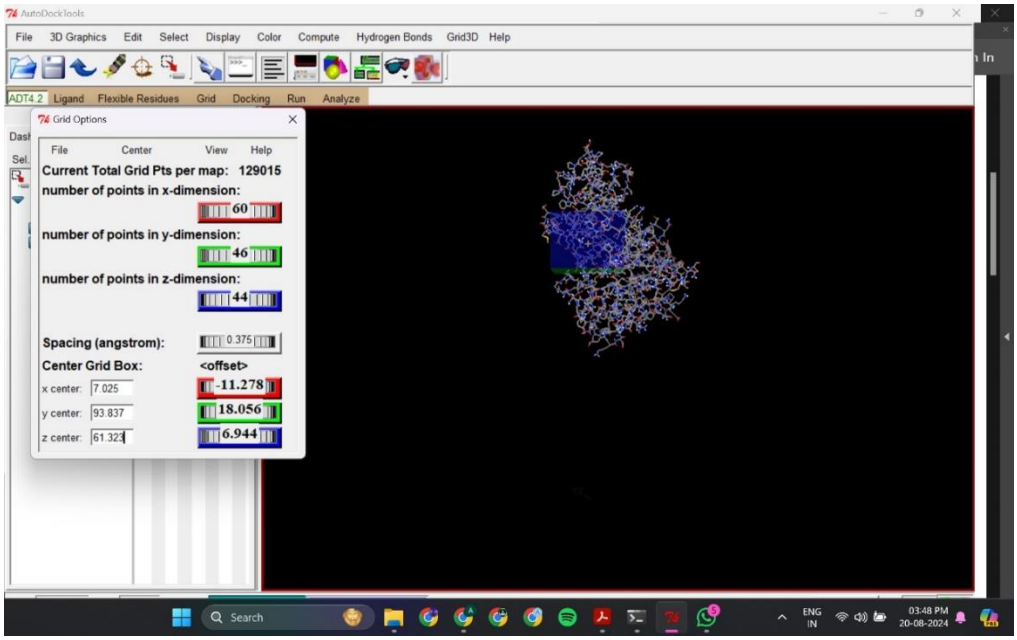
3

The structure of ZINC126204226 was obtained from the ZINC15 database in .mol2 format.

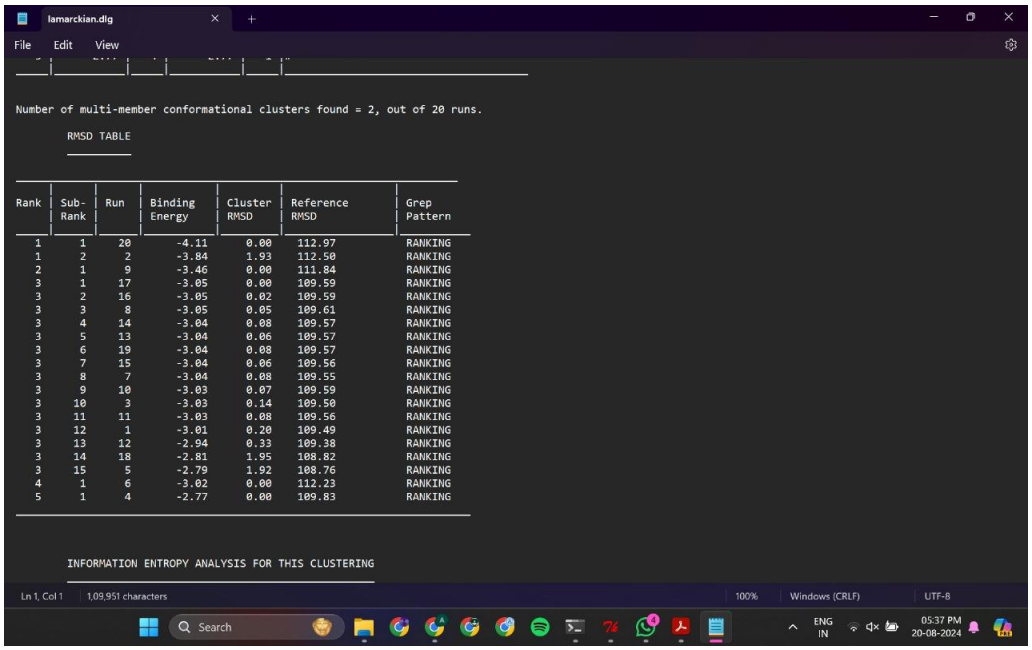


4

- The default working directory was changed to the folder containing the relevant files.
- The protein was loaded in AutoDock.
- All heteroatoms, including waters, were deleted.
- Chain A of the protein was deleted to make it monomeric.
- Polar hydrogens were added to the protein.
- All atoms were designated as ‘AD4’ type.
- Kollman charges were added to the protein.
- The protein was saved in .pdbqt format.
- After deleting the protein, the ligand was loaded into AutoDock.
- All amide bonds were made rotatable.
- The ligand was saved in .pdbqt format and then deleted from the AutoDock screen.
- The ‘Grid’ option was used to load the protein and ligand from their .pdbqt files. The ligand was loaded using ‘Set Map Types’.
- A grid box was created using the given settings and saved as a .gpf file.

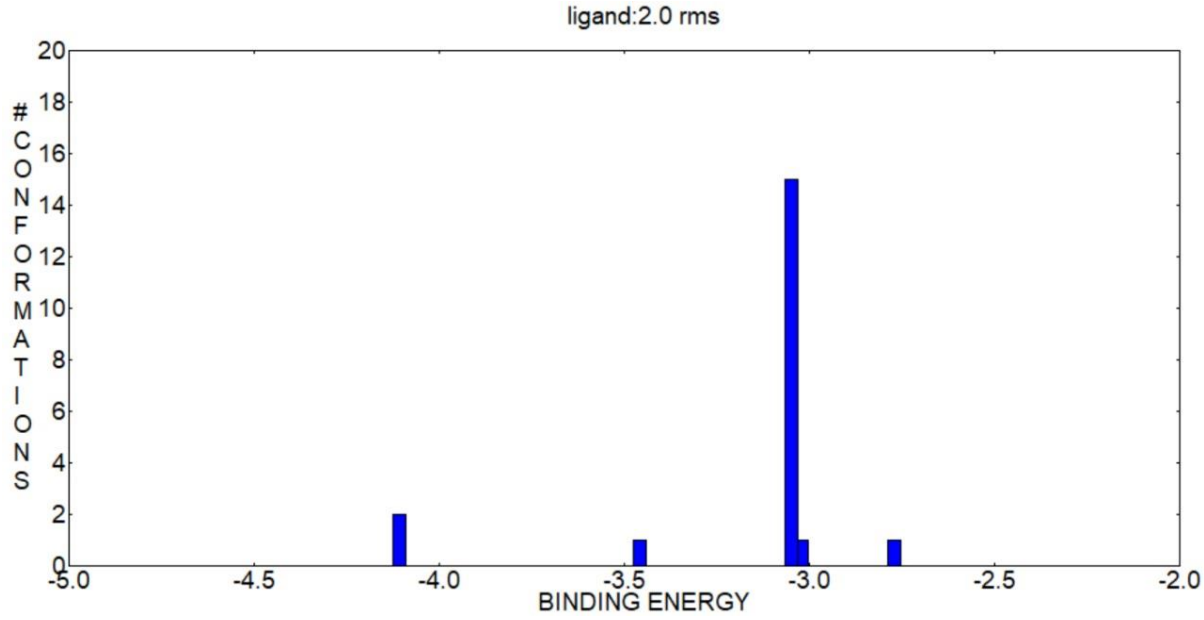


- AutoGrid was run using the .gpf file to create a .glg file and .map files.
- The protein and ligand were reloaded using the 'Docking' option. The protein was loaded using 'Set Rigid Filename'.
- The 'Search Parameters' option was set to 'Genetic Algorithm' with 20 GA runs. The output was chosen as 'Lamarckian GA', producing a .dpf file.
- AutoDock was run and the output was obtained in a .dlg file.
- The output contained 20 different orientations of the ligand in the active site.

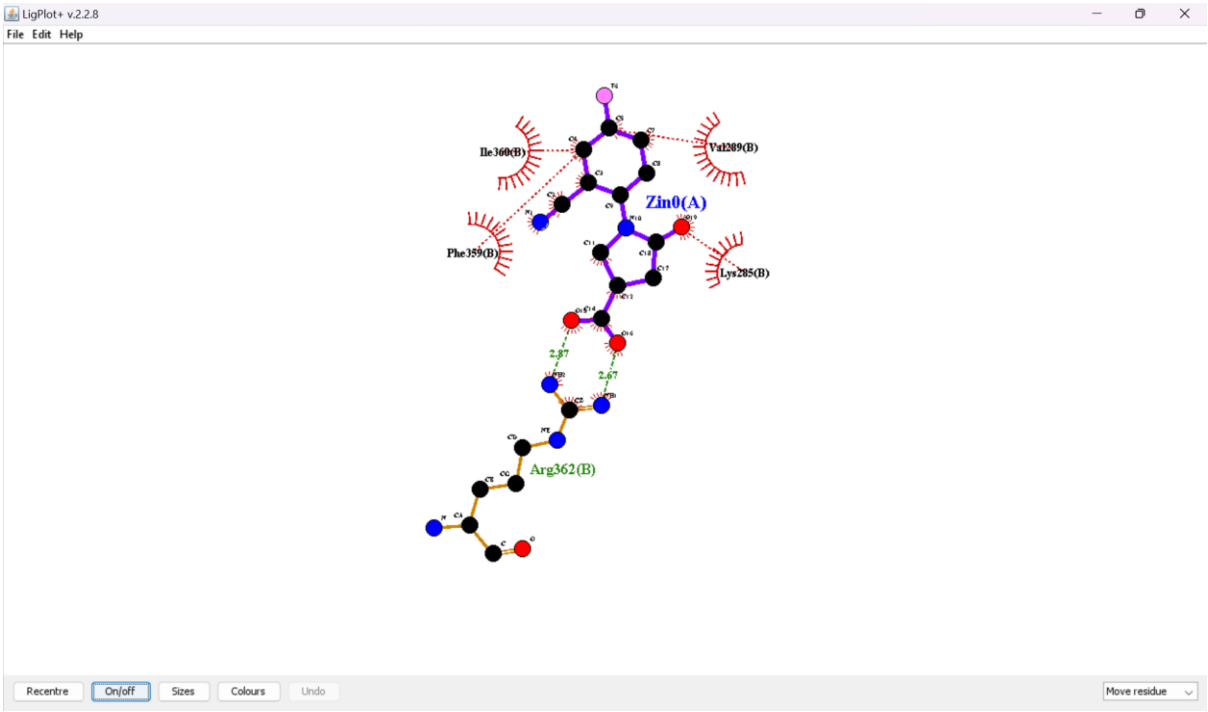


Based on the docking results, the best ligand orientation was obtained during the 20th run of the genetic algorithm. This had a binding energy of –4.11 kcal/mol.

The 'Analyze' option's 'Clusterings' tool was used to obtain the clustering histogram of the 20 ligand orientation.



The interactions were found from LigPlot after saving the complex as a .pdb file.

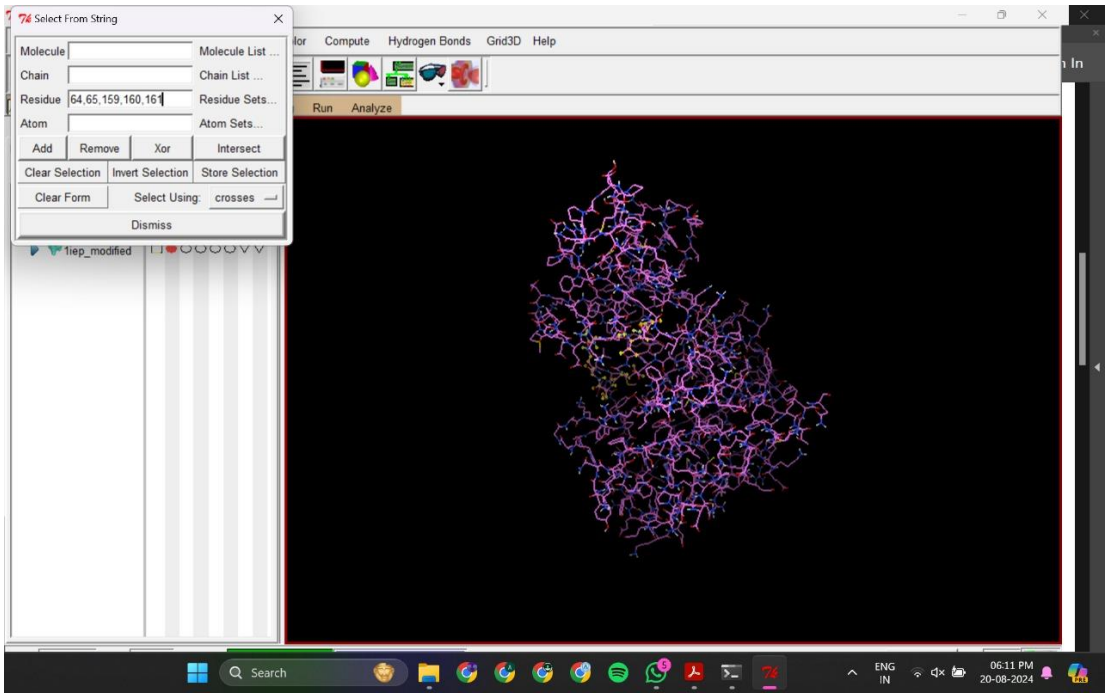


4 hydrophobic interactions and 2 hydrogen bonds were identified.

Flexible Docking

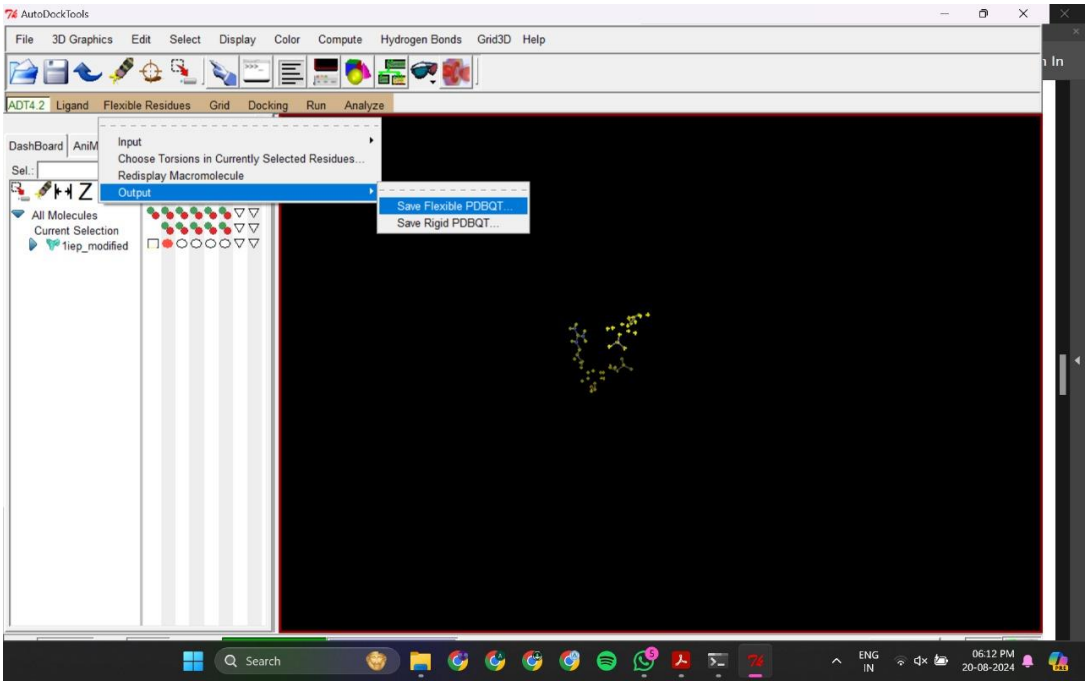
1

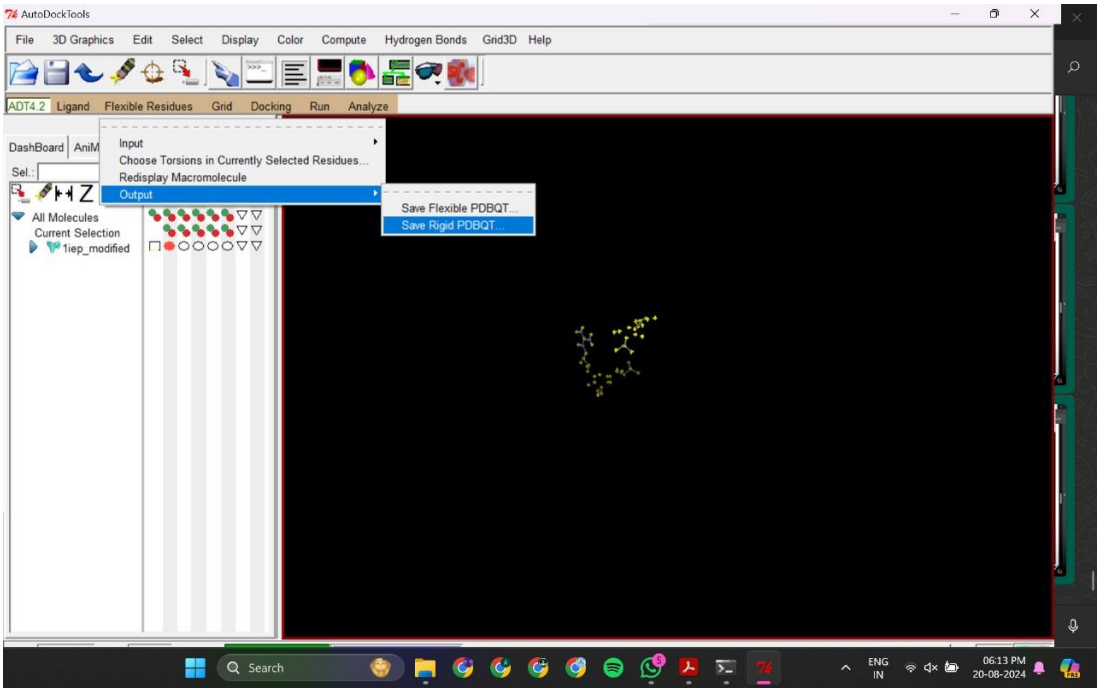
Using the .pdbqt file of the protein, the given residues were set to be flexible.



2

Two .pdbqt files were downloaded for flexible and rigid residues.





3

- The ‘Grid’ option was used to load the rigid residues, flexible residues and ligand from their .pdbqt files. The flexible residues and ligand were loaded using ‘Set Map Types’.
- The ‘Grid’ option was used to load the protein and ligand from their .pdbqt files. The ligand was loaded using ‘Set Map Types’.
- A grid box was created using the given settings and saved as a .gpf file.
- AutoGrid was run using the .gpf file to create a .glg file and .map files.
- The protein and ligand were reloaded using the ‘Docking’ option. The protein was loaded using ‘Set Rigid Filename’.
- The ‘Search Parameters’ option was set to ‘Genetic Algorithm’ with 10 GA runs. The output was chosen as ‘Lamarckian GA’, producing a .dpf file.
- AutoDock was run and the output was obtained in a .dlg file.
- The output contained 10 different orientations of the ligand in the active site.

flex_lamarckian.dlg

File Edit View

RMSD TABLE

Rank	Sub-Rank	Run	Binding Energy	Cluster RMSD	Reference RMSD	Grep Pattern
1	1	7	-3.58	0.00	112.87	RANKING
2	1	9	-2.83	0.00	108.00	RANKING
3	1	5	-2.26	0.00	109.89	RANKING
4	1	6	-1.95	0.00	106.38	RANKING
5	1	4	-1.72	0.00	108.94	RANKING
6	1	3	-1.70	0.00	109.65	RANKING
7	1	10	-1.61	0.00	108.86	RANKING
8	1	1	-1.42	0.00	108.50	RANKING
9	1	2	-1.41	0.00	108.93	RANKING
10	1	8	-1.10	0.00	110.97	RANKING

INFORMATION ENTROPY ANALYSIS FOR THIS CLUSTERING

Information entropy for this clustering = 1.00 (rmstol = 2.00 Angstrom)

STATISTICAL MECHANICAL ANALYSIS

Partition function, Q = 10.03 at Temperature, T = 298.15 K
Free energy, A ~ -1366.20 kcal/mol at Temperature, T = 298.15 K

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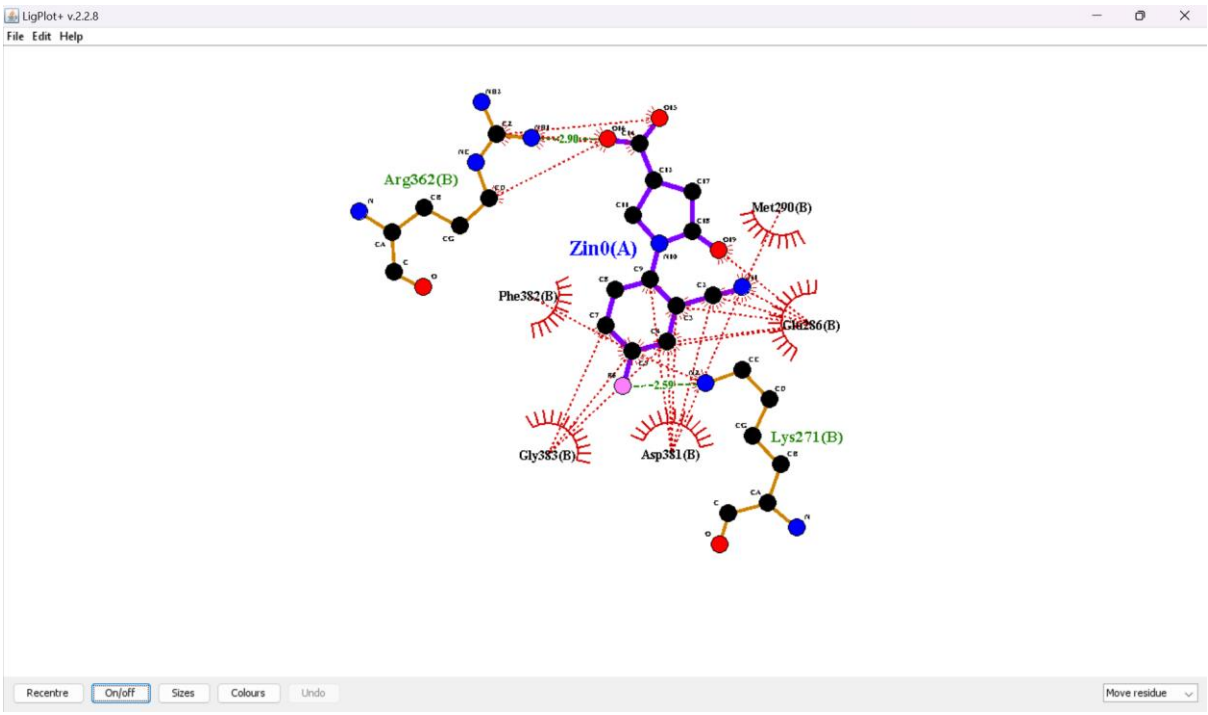
Many more hydrophobic interactions are found using flexible docking. This is due to the fact that the interacting molecules are allowed greater conformational flexibility during the docking procedure.



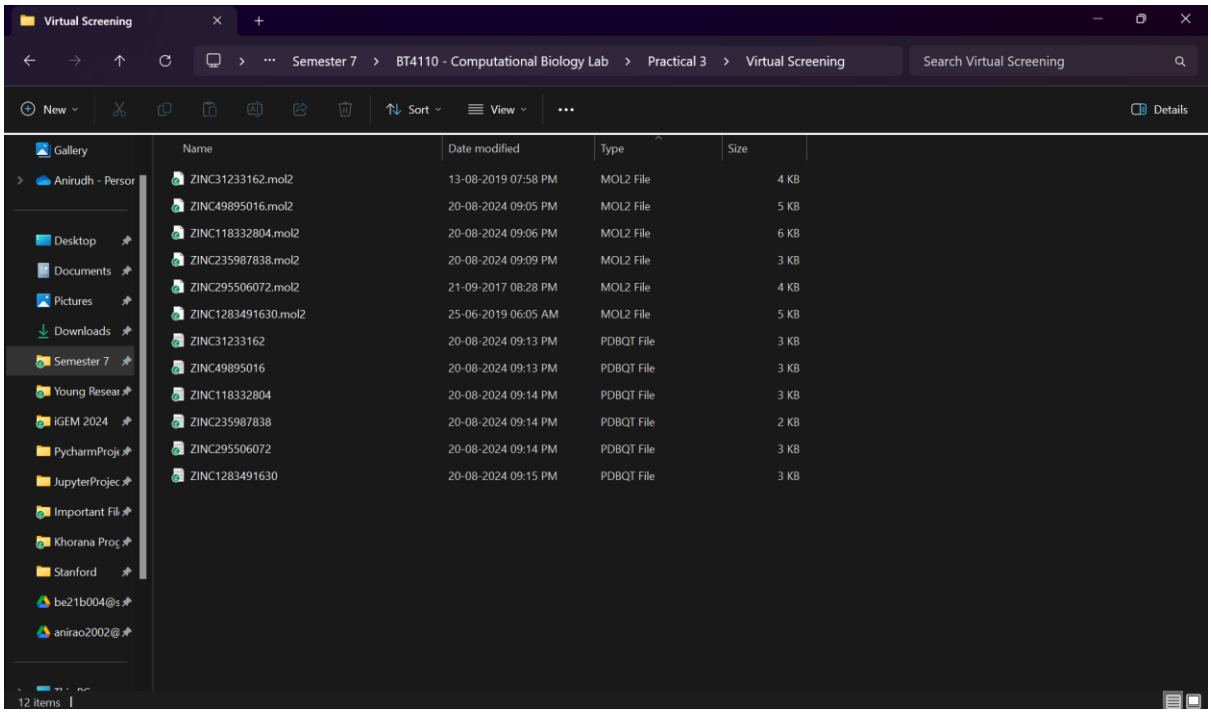
5

In general, flexible docking provides better results than rigid docking. Biomolecules in the cell constantly undergo transient changes in their conformations. Flexible docking allows us to consider these dynamic effects while docking a ligand with a receptor. Rigid docking may be more useful when computational resources are limited. For drug discovery applications, flexible docking is preferred as it more accurately reflects the behaviour of biomolecules in the cell.

Virtual Screening

1

The structures of all 6 ligands were downloaded from ZINC15 in either .mol2 or .sdf format. Using OpenBabel, all files were ultimately obtained in .mol2 format. Using AutoDock, their amide bonds were made rotatable and the ligands were saved in .pdbqt format.



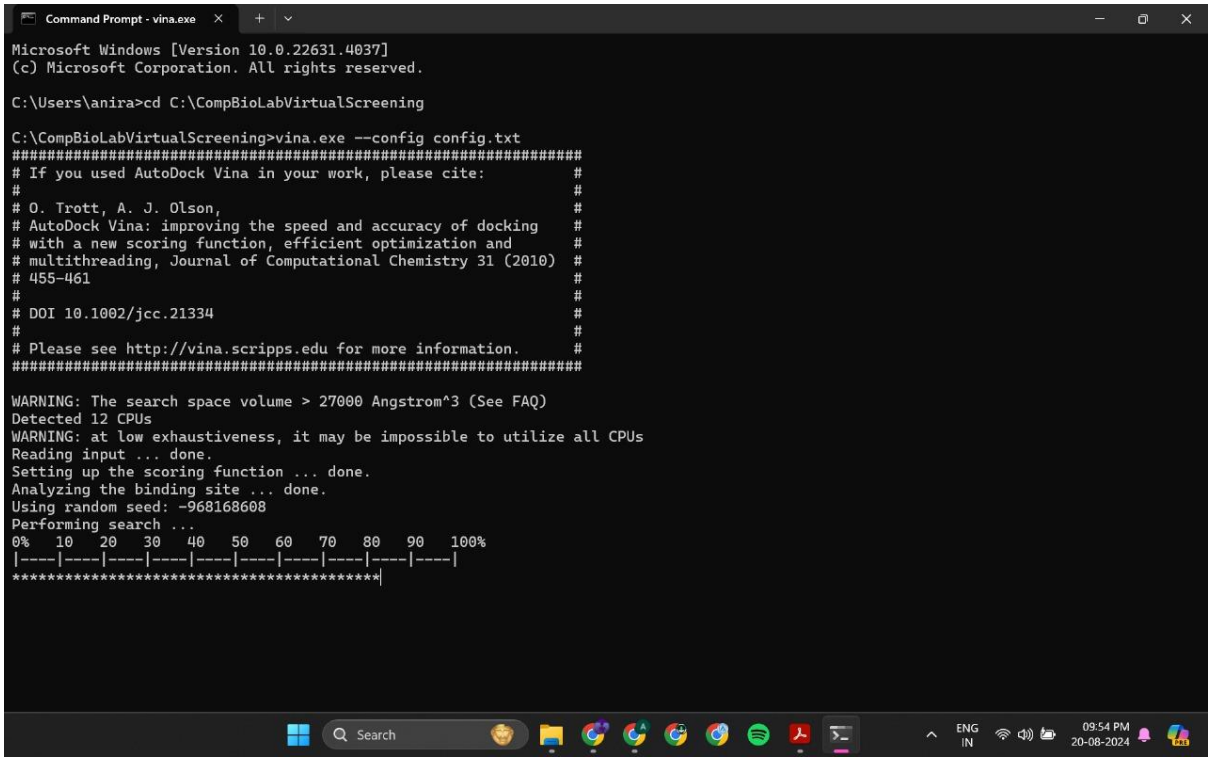
2

AutoDock Vina was used to individually screen the 6 ligands against 1IEP. A config.txt file was created to run Vina from the command line. This was done manually for each of the 6 ligands.

config.txt

File Edit View

```
receptor = 1iep_modified.pdbqt
ligand = ZINC1283491630.pdbqt

out = ZINC1283491630_out.pdbqt

center_x = 7.025
center_y = 93.837
center_z = 61.323

size_x = 60.0
size_y = 46.0
size_z = 44.0

log = log_ZINC1283491630.txt

exhaustiveness = 8
```



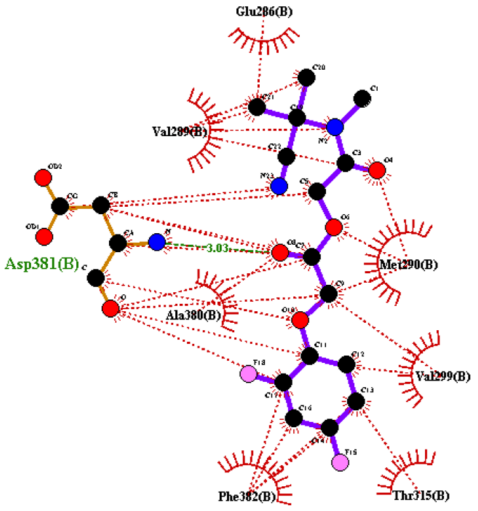
The log files obtained after the docking procedure were used to tabulate the binding energies of the ligands.

Ligand	Binding Energy (kcal/mol)
ZINC1283491630	–8.1
ZINC49895016	–8.6
ZINC118332804	–5.7
ZINC31233162	–6.7
ZINC235987838	–7.8
ZINC295506072	–5.8

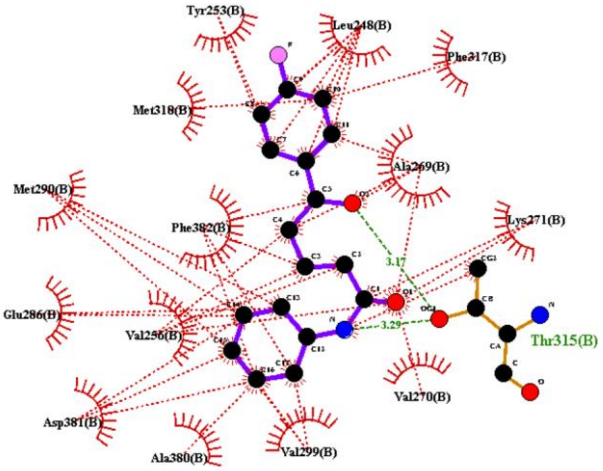
From these results, **ZINC49895016** seems to have the highest affinity for 1IEP.

3

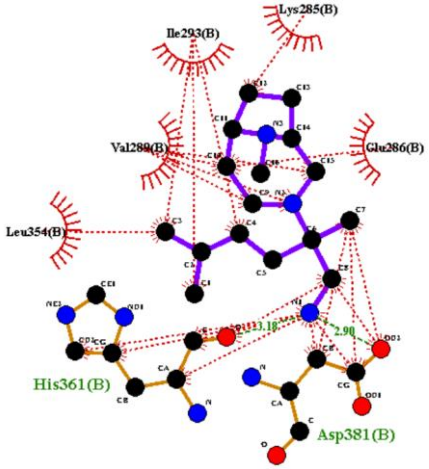
LigPlot was used to determine the protein-ligand interactions.



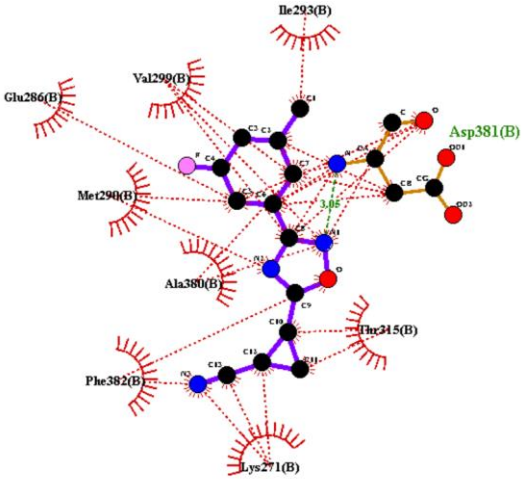
ZINC31233162_docked



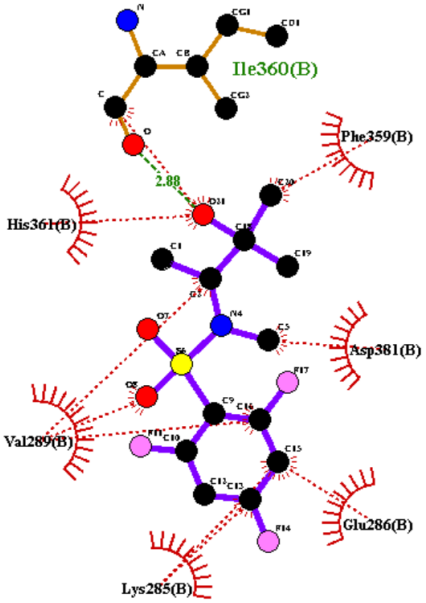
ZINC49895016_docked



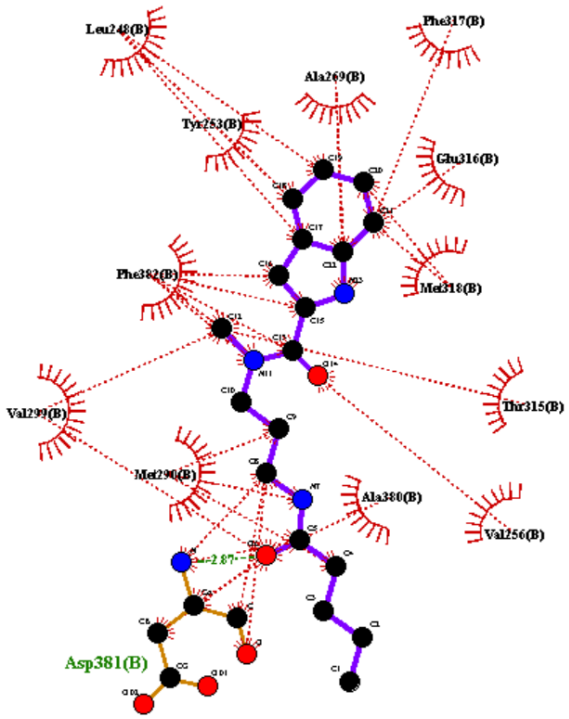
ZINC118332804_docked



ZINC235987838_docked



ZINC295506072_docked



ZINC1283491630_docked

The interactions are tabulated below:

ZINC31233162

	Interaction type	Atom 1 name	Res 1 name	Res 1 no.	Atom 2 name	Res 2 name	Res 2 no.	Distance
1	Hydrogen bond	N	ASP	381	O8	ZIN	0	3.032
2	Hydrophobic	C14	ZIN	0	CE2	PHE	382	3.846
3	Hydrophobic	C13	ZIN	0	CE2	PHE	382	3.808
4	Hydrophobic	C17	ZIN	0	CD2	PHE	382	3.805
5	Hydrophobic	C16	ZIN	0	CD2	PHE	382	3.688
6	Hydrophobic	C14	ZIN	0	CD2	PHE	382	3.84
7	Hydrophobic	N23	ZIN	0	CG	ASP	381	3.649
8	Hydrophobic	O8	ZIN	0	CB	ASP	381	3.243
9	Hydrophobic	C7	ZIN	0	CB	ASP	381	3.762
10	Hydrophobic	C5	ZIN	0	CB	ASP	381	3.377
11	Hydrophobic	C17	ZIN	0	O	ASP	381	3.724
12	Hydrophobic	C11	ZIN	0	O	ASP	381	3.622
13	Hydrophobic	C9	ZIN	0	O	ASP	381	3.888
14	Hydrophobic	C7	ZIN	0	O	ASP	381	3.738
15	Hydrophobic	O10	ZIN	0	C	ASP	381	3.789
16	Hydrophobic	O8	ZIN	0	CA	ASP	381	3.648
17	Hydrophobic	C7	ZIN	0	N	ASP	381	3.694
18	Hydrophobic	O8	ZIN	0	C	ALA	380	3.635

19	Hydrophobic	O8	ZIN	0	CA	ALA	380	3.885
20	Hydrophobic	C13	ZIN	0	OG1	THR	315	3.634
21	Hydrophobic	C12	ZIN	0	CG1	VAL	299	3.799
22	Hydrophobic	C9	ZIN	0	CG1	VAL	299	3.708
23	Hydrophobic	C9	ZIN	0	CE	MET	290	3.475
24	Hydrophobic	C9	ZIN	0	SD	MET	290	3.799
25	Hydrophobic	O6	ZIN	0	SD	MET	290	3.834
26	Hydrophobic	O6	ZIN	0	CG	MET	290	3.665
27	Hydrophobic	O4	ZIN	0	CG	MET	290	3.516
28	Hydrophobic	C21	ZIN	0	CG2	VAL	289	3.89
29	Hydrophobic	C21	ZIN	0	CG1	VAL	289	3.764
30	Hydrophobic	C20	ZIN	0	CG1	VAL	289	3.734
31	Hydrophobic	O4	ZIN	0	CG1	VAL	289	3.477
32	Hydrophobic	N2	ZIN	0	CG1	VAL	289	3.858
33	Hydrophobic	C21	ZIN	0	CB	VAL	289	3.681
34	Hydrophobic	C21	ZIN	0	CG	GLU	286	3.732
35	Hydrophobic	C21	ZIN	0	CA	GLU	286	3.748

ZINC49895016

	Interaction type	Atom 1 name	Res 1 name	Res 1 no.	Atom 2 name	Res 2 name	Res 2 no.	Distance
1	Hydrogen bond	OG1	THR	315	O2	UNL	1	3.171
2	Hydrogen bond	N	UNL	1	OG1	THR	315	3.294
3	Hydrophobic	C17	UNL	1	CE2	PHE	382	3.714
4	Hydrophobic	C16	UNL	1	CE2	PHE	382	3.652
5	Hydrophobic	C5	UNL	1	CE2	PHE	382	3.646
6	Hydrophobic	C3	UNL	1	CE2	PHE	382	3.662
7	Hydrophobic	C16	UNL	1	CD2	PHE	382	3.737
8	Hydrophobic	C3	UNL	1	CD2	PHE	382	3.467
9	Hydrophobic	C15	UNL	1	O	ASP	381	3.735
10	Hydrophobic	C14	UNL	1	O	ASP	381	3.62
11	Hydrophobic	C16	UNL	1	N	ASP	381	3.587
12	Hydrophobic	C15	UNL	1	N	ASP	381	3.825
13	Hydrophobic	C16	UNL	1	CB	ALA	380	3.387
14	Hydrophobic	C10	UNL	1	N	MET	318	3.425
15	Hydrophobic	C10	UNL	1	CD1	PHE	317	3.711
16	Hydrophobic	N	UNL	1	CG2	THR	315	3.534
17	Hydrophobic	C17	UNL	1	CG1	VAL	299	3.739
18	Hydrophobic	C16	UNL	1	CG1	VAL	299	3.448
19	Hydrophobic	C15	UNL	1	CG1	VAL	299	3.569
20	Hydrophobic	C15	UNL	1	CE	MET	290	3.835
21	Hydrophobic	C14	UNL	1	CE	MET	290	3.757
22	Hydrophobic	C13	UNL	1	CE	MET	290	3.645
23	Hydrophobic	C14	UNL	1	SD	MET	290	3.721
24	Hydrophobic	C13	UNL	1	SD	MET	290	3.879
25	Hydrophobic	C14	UNL	1	OE2	GLU	286	3.006
26	Hydrophobic	C13	UNL	1	OE2	GLU	286	3.699
27	Hydrophobic	O1	UNL	1	CB	LYS	271	3.572

28	Hydrophobic	C1	UNL	1	CB	LYS	271	3.878
29	Hydrophobic	O1	UNL	1	CA	LYS	271	3.85
30	Hydrophobic	O1	UNL	1	C	VAL	270	3.696
31	Hydrophobic	O1	UNL	1	CA	VAL	270	3.898
32	Hydrophobic	O2	UNL	1	CB	ALA	269	3.405
33	Hydrophobic	O1	UNL	1	CB	ALA	269	3.703
34	Hydrophobic	C11	UNL	1	CB	ALA	269	3.81
35	Hydrophobic	C4	UNL	1	CB	ALA	269	3.777
36	Hydrophobic	O1	UNL	1	C	ALA	269	3.476
37	Hydrophobic	C3	UNL	1	CG2	VAL	256	3.888
38	Hydrophobic	C2	UNL	1	CG2	VAL	256	3.367
39	Hydrophobic	O1	UNL	1	CG1	VAL	256	3.634
40	Hydrophobic	C4	UNL	1	CG1	VAL	256	3.326
41	Hydrophobic	C3	UNL	1	CG1	VAL	256	3.691
42	Hydrophobic	C2	UNL	1	CG1	VAL	256	3.441
43	Hydrophobic	C8	UNL	1	OH	TYR	253	3.788
44	Hydrophobic	C8	UNL	1	CZ	TYR	253	3.855
45	Hydrophobic	C7	UNL	1	CZ	TYR	253	3.853
46	Hydrophobic	C8	UNL	1	CE1	TYR	253	3.793
47	Hydrophobic	C11	UNL	1	CD2	LEU	248	3.808
48	Hydrophobic	C10	UNL	1	CD2	LEU	248	3.8
49	Hydrophobic	C9	UNL	1	CD2	LEU	248	3.787
50	Hydrophobic	C8	UNL	1	CD2	LEU	248	3.779
51	Hydrophobic	C7	UNL	1	CD2	LEU	248	3.788
52	Hydrophobic	C6	UNL	1	CD2	LEU	248	3.841

ZINC118332804

	Interaction type	Atom 1 name	Res 1 name	Res 1 no.	Atom 2 name	Res 2 name	Res 2 no.	Distance
1	Hydrogen bond	N1	UNL	1	OD2	ASP	381	2.901
2	Hydrogen bond	N1	UNL	1	O	HIS	361	3.18
3	Hydrophobic	C8	UNL	1	OD2	ASP	381	3.353
4	Hydrophobic	C7	UNL	1	OD2	ASP	381	3.66
5	Hydrophobic	C7	UNL	1	CG	ASP	381	3.477
6	Hydrophobic	N1	UNL	1	CG	ASP	381	3.638
7	Hydrophobic	C7	UNL	1	CB	ASP	381	3.378
8	Hydrophobic	N1	UNL	1	CB	ASP	381	3.68
9	Hydrophobic	N1	UNL	1	CD2	HIS	361	3.773
10	Hydrophobic	N1	UNL	1	CG	HIS	361	3.664
11	Hydrophobic	C8	UNL	1	O	HIS	361	3.594
12	Hydrophobic	N1	UNL	1	C	HIS	361	3.777
13	Hydrophobic	N1	UNL	1	CA	HIS	361	3.523
14	Hydrophobic	C3	UNL	1	CD2	LEU	354	3.655
15	Hydrophobic	C3	UNL	1	CD1	LEU	354	3.657
16	Hydrophobic	C4	UNL	1	CD1	ILE	293	3.826
17	Hydrophobic	C3	UNL	1	CD1	ILE	293	3.64
18	Hydrophobic	C1	UNL	1	CD1	ILE	293	3.844
19	Hydrophobic	C10	UNL	1	CG2	VAL	289	3.539

20	Hydrophobic	C15	UNL	1	CG1	VAL	289	3.627
21	Hydrophobic	C9	UNL	1	CG1	VAL	289	3.797
22	Hydrophobic	N2	UNL	1	CG1	VAL	289	3.738
23	Hydrophobic	C9	UNL	1	CB	VAL	289	3.82
24	Hydrophobic	C10	UNL	1	CA	GLU	286	3.7
25	Hydrophobic	C12	UNL	1	CE	LYS	285	3.703
26	Hydrophobic	C12	UNL	1	CG	LYS	285	3.637

ZINC235987838

	Interaction type	Atom 1 name	Res 1 name	Res 1 no.	Atom 2 name	Res 2 name	Res 2 no.	Distance
1	Hydrogen bond	N	ASP	381	N1	UNL	1	3.052
2	Hydrophobic	O	UNL	1	CE2	PHE	382	3.479
3	Hydrophobic	O	UNL	1	CD2	PHE	382	3.496
4	Hydrophobic	N3	UNL	1	CD2	PHE	382	3.704
5	Hydrophobic	N3	UNL	1	CA	PHE	382	3.415
6	Hydrophobic	C7	UNL	1	CB	ASP	381	3.812
7	Hydrophobic	C6	UNL	1	CB	ASP	381	3.853
8	Hydrophobic	C2	UNL	1	CB	ASP	381	3.853
9	Hydrophobic	C8	UNL	1	O	ASP	381	3.442
10	Hydrophobic	C6	UNL	1	O	ASP	381	3.59
11	Hydrophobic	C5	UNL	1	O	ASP	381	3.637
12	Hydrophobic	N1	UNL	1	C	ASP	381	3.736
13	Hydrophobic	C8	UNL	1	N	ASP	381	3.646
14	Hydrophobic	C7	UNL	1	N	ASP	381	3.366
15	Hydrophobic	C6	UNL	1	N	ASP	381	3.582
16	Hydrophobic	N1	UNL	1	CB	ALA	380	3.301
17	Hydrophobic	C7	UNL	1	C	ALA	380	3.868
18	Hydrophobic	N1	UNL	1	C	ALA	380	3.872
19	Hydrophobic	C7	UNL	1	CA	ALA	380	3.891
20	Hydrophobic	N1	UNL	1	CA	ALA	380	3.71
21	Hydrophobic	C11	UNL	1	OG1	THR	315	3.743
22	Hydrophobic	C10	UNL	1	CG2	THR	315	3.718
23	Hydrophobic	C8	UNL	1	CG1	VAL	299	3.836
24	Hydrophobic	C7	UNL	1	CG1	VAL	299	3.85
25	Hydrophobic	N1	UNL	1	CG1	VAL	299	3.525
26	Hydrophobic	C1	UNL	1	CD1	ILE	293	3.865
27	Hydrophobic	N2	UNL	1	CE	MET	290	3.406
28	Hydrophobic	C5	UNL	1	SD	MET	290	3.818
29	Hydrophobic	N2	UNL	1	SD	MET	290	3.598
30	Hydrophobic	C5	UNL	1	CG	MET	290	3.881
31	Hydrophobic	C5	UNL	1	OE2	GLU	286	3.309
32	Hydrophobic	C13	UNL	1	NZ	LYS	271	3.774
33	Hydrophobic	C13	UNL	1	CD	LYS	271	3.625
34	Hydrophobic	C12	UNL	1	CD	LYS	271	3.87
35	Hydrophobic	N3	UNL	1	CD	LYS	271	3.796
36	Hydrophobic	C12	UNL	1	CB	LYS	271	3.691

ZINC295506072

	Interaction type	Atom 1 name	Res 1 name	Res 1 no.	Atom 2 name	Res 2 name	Res 2 no.	Distance
1	Hydrogen bond	O21	ZIN	0	O	ILE	360	2.881
2	Hydrophobic	C5	ZIN	0	OD2	ASP	381	3.414
3	Hydrophobic	C5	ZIN	0	CG	ASP	381	3.561
4	Hydrophobic	O21	ZIN	0	CA	HIS	361	3.551
5	Hydrophobic	O21	ZIN	0	C	ILE	360	3.86
6	Hydrophobic	C20	ZIN	0	CD2	PHE	359	3.743
7	Hydrophobic	C20	ZIN	0	CB	PHE	359	3.78
8	Hydrophobic	C16	ZIN	0	CG2	VAL	289	3.887
9	Hydrophobic	O8	ZIN	0	CG1	VAL	289	3.421
10	Hydrophobic	C2	ZIN	0	CG1	VAL	289	3.629
11	Hydrophobic	C15	ZIN	0	CA	GLU	286	3.54
12	Hydrophobic	C15	ZIN	0	N	GLU	286	3.604
13	Hydrophobic	C15	ZIN	0	CG	LYS	285	3.715
14	Hydrophobic	C13	ZIN	0	CG	LYS	285	3.715

ZINC1283491630

	Interaction type	Atom 1 name	Res 1 name	Res 1 no.	Atom 2 name	Res 2 name	Res 2 no.	Distance
1	Hydrogen bond	N	ASP	381	O6	ZIN	0	2.869
2	Hydrophobic	C16	ZIN	0	CE2	PHE	382	3.489
3	Hydrophobic	C15	ZIN	0	CE2	PHE	382	3.571
4	Hydrophobic	C13	ZIN	0	CE2	PHE	382	3.654
5	Hydrophobic	C12	ZIN	0	CE2	PHE	382	3.467
6	Hydrophobic	N11	ZIN	0	CE2	PHE	382	3.547
7	Hydrophobic	C13	ZIN	0	CD2	PHE	382	3.753
8	Hydrophobic	N11	ZIN	0	CD2	PHE	382	3.7
9	Hydrophobic	O6	ZIN	0	CB	ASP	381	3.65
10	Hydrophobic	C8	ZIN	0	O	ASP	381	3.26
11	Hydrophobic	C8	ZIN	0	C	ASP	381	3.855
12	Hydrophobic	O6	ZIN	0	CA	ASP	381	3.74
13	Hydrophobic	C8	ZIN	0	N	ASP	381	3.743
14	Hydrophobic	C5	ZIN	0	N	ASP	381	3.759
15	Hydrophobic	O6	ZIN	0	C	ALA	380	3.436
16	Hydrophobic	O6	ZIN	0	CA	ALA	380	3.448
17	Hydrophobic	C20	ZIN	0	O	MET	318	3.896
18	Hydrophobic	C21	ZIN	0	N	MET	318	3.601
19	Hydrophobic	C20	ZIN	0	N	MET	318	3.575
20	Hydrophobic	C21	ZIN	0	CA	PHE	317	3.886
21	Hydrophobic	C21	ZIN	0	O	GLU	316	3.895
22	Hydrophobic	C12	ZIN	0	OG1	THR	315	3.576
23	Hydrophobic	C12	ZIN	0	CG2	VAL	299	3.453
24	Hydrophobic	O6	ZIN	0	CG1	VAL	299	3.647
25	Hydrophobic	C9	ZIN	0	CE	MET	290	3.778
26	Hydrophobic	N7	ZIN	0	CE	MET	290	3.558

27	Hydrophobic	N7	ZIN	0	SD	MET	290	3.581
28	Hydrophobic	C4	ZIN	0	SD	MET	290	3.878
29	Hydrophobic	C4	ZIN	0	CG	MET	290	3.341
30	Hydrophobic	N23	ZIN	0	CB	ALA	269	3.277
31	Hydrophobic	C22	ZIN	0	CB	ALA	269	3.668
32	Hydrophobic	O14	ZIN	0	CG2	VAL	256	3.641
33	Hydrophobic	O14	ZIN	0	CG1	VAL	256	3.458
34	Hydrophobic	C18	ZIN	0	OH	TYR	253	3.893
35	Hydrophobic	C18	ZIN	0	CZ	TYR	253	3.727
36	Hydrophobic	C18	ZIN	0	CE1	TYR	253	3.696
37	Hydrophobic	C19	ZIN	0	CD2	LEU	248	3.76
38	Hydrophobic	C18	ZIN	0	CD2	LEU	248	3.643
39	Hydrophobic	C17	ZIN	0	CD2	LEU	248	3.72

In summary,

Ligand	Hydrogen Bonds	Hydrophobic Interactions	Total No. of Interactions
ZINC1283491630	1	38	39
ZINC49895016	2	50	52
ZINC118332804	2	24	26
ZINC31233162	1	34	35
ZINC235987838	1	35	36
ZINC295506072	1	13	14

ZINC49895016, which has the highest affinity for 1IEP, also has the highest number of interactions among the ligands. It has a higher number of hydrogen bonds and hydrophobic interactions. Thus, **ZINC49895016** binds most effectively to the protein.