Binding Free-Energy Estimator 2 (BFEE2)

Calculate protein-ligand standard binding free energies

By Purnawan Pontana Putra

nature protocols

PROTOCOL

https://doi.org/10.1038/s41596-021-00676-1

Check for updates

Accurate determination of protein:ligand standard binding free energies from molecular dynamics simulations

Haohao Fu[1], Haochuan Chen[1], Marharyta Blazhynska[2], Emma Goulard Coderc de Lacam[2], Florence Szczepaniak[2,3], Anna Pavlova[4], Xueguang Shao[1], James C. Gumbart[4], François Dehez[2], Benoît Roux[3,5,6], Wensheng Cai[1]✉ and Christophe Chipot[2,7,8]✉

Designing a reliable computational methodology to calculate protein:ligand standard binding free energies is extremely challenging. The large change in configurational enthalpy and entropy that accompanies the association of ligand and protein is notoriously difficult to capture in naive brute-force simulations. Addressing this issue, the present protocol rests upon a rigorous statistical mechanical framework for the determination of protein:ligand binding affinities together with the comprehensive Binding Free-Energy Estimator 2 (BFEE2) application software. With the knowledge of the bound state, available from experiments or docking, application of the BFEE2 protocol with a reliable force field supplies in a matter of days standard binding free energies within chemical accuracy, for a broad range of protein:ligand complexes. Limiting undesirable human intervention, BFEE2 assists the end user in preparing all the necessary input files and performing the post-treatment of the simulations towards the final estimate of the binding affinity.

Software

- VMD 1.9.3 or later (www.ks.uiuc.edu/Research/vmd)
- NAMD 3.0 alpha or later (www.ks.uiuc.edu/Research/namd) or Gromacs 2020.4 or later (www.gromacs.org) patched with Colvars (colvars.github.io)
- Python 3.7 or later (https://www.python.org/). The use of conda https://docs.conda.io/en/latest/) is recommended
- BFEE2 (github.com/fhh2626/BFEE2)

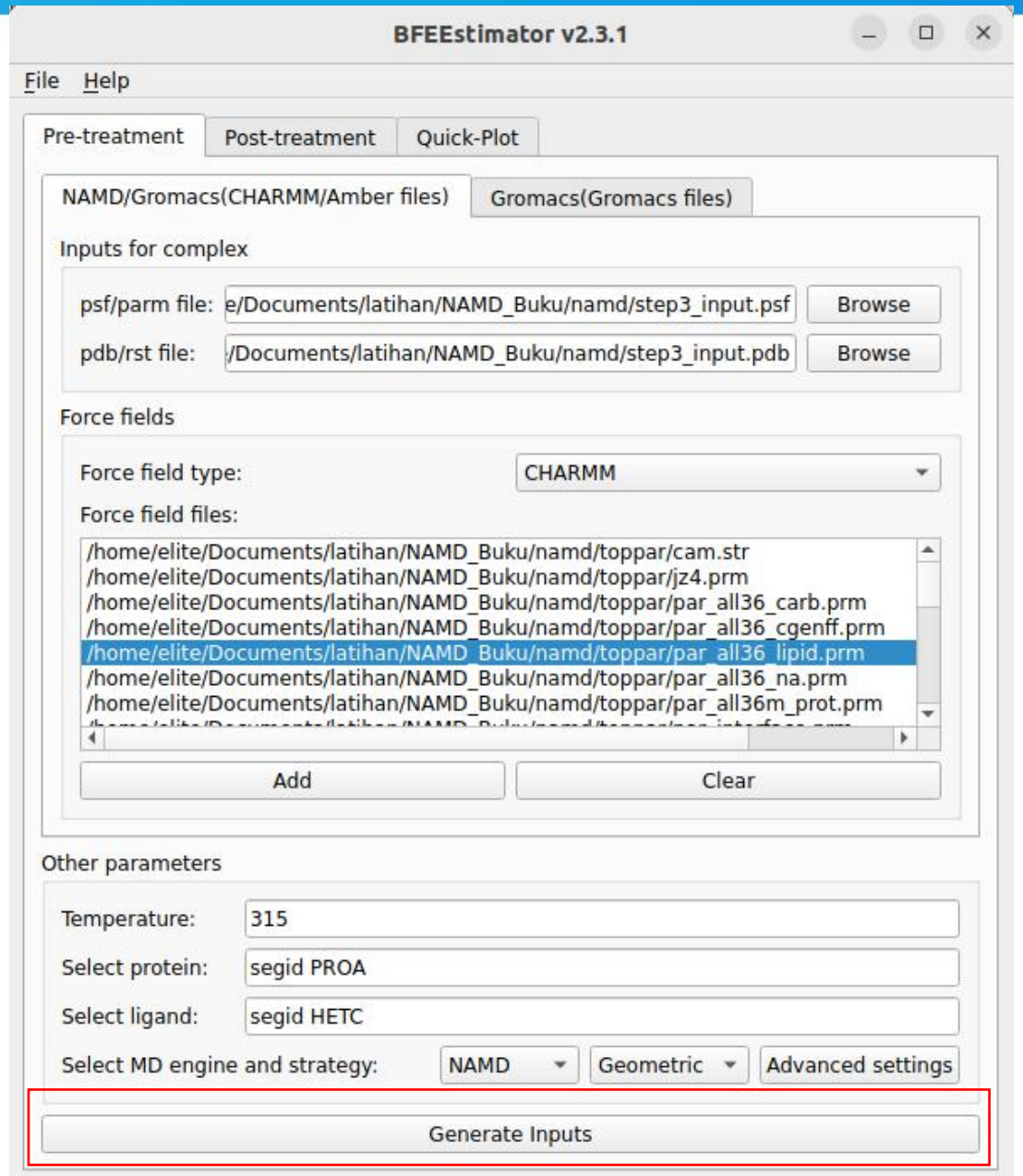
Installation BFEE2

- **conda create -n BFEE2**
- **conda activate BFEE2**
- **conda install -c conda-forge BFEE2**
- running BFEE using terminal: **BFEE2Gui.py**

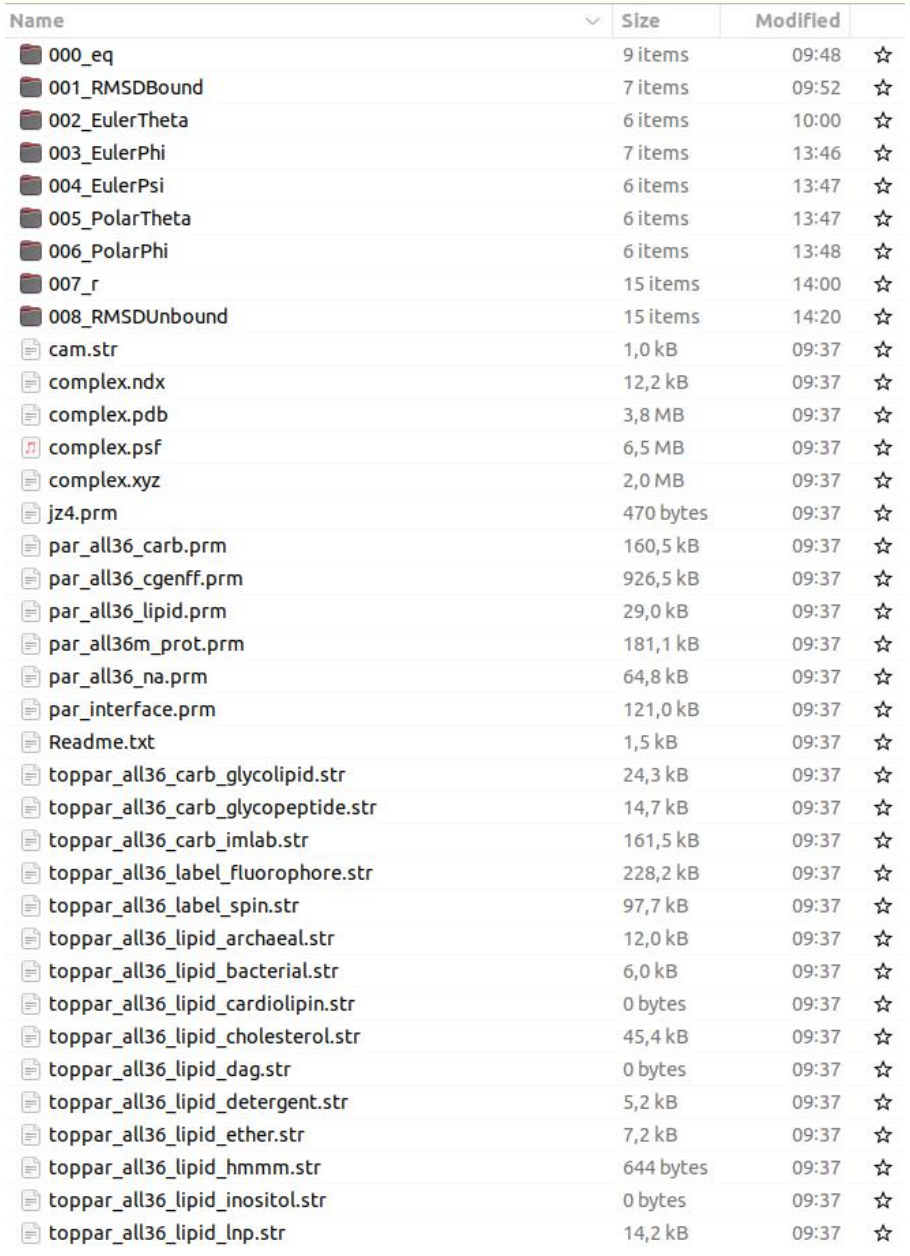
Preparation using CHARMM-GUI

- Forcefield CHARMM36M
- Watermodel TIP3P

BFEEstimator GUI



Open Folder BFEE



Name	Size	Modified
000_eq	9 items	09:48
001_RMSDBound	7 items	09:52
002_EulerTheta	6 items	10:00
003_EulerPhi	7 items	13:46
004_EulerPsi	6 items	13:47
005_PolarTheta	6 items	13:47
006_PolarPhi	6 items	13:48
007_r	15 items	14:00
008_RMSDUnbound	15 items	14:20
cam.str	1,0 kB	09:37
complex.ndx	12,2 kB	09:37
complex.pdb	3,8 MB	09:37
complex.psf	6,5 MB	09:37
complex.xyz	2,0 MB	09:37
jz4.prm	470 bytes	09:37
par_all36_carb.prm	160,5 kB	09:37
par_all36_cgenff.prm	926,5 kB	09:37
par_all36_lipid.prm	29,0 kB	09:37
par_all36m_prot.prm	181,1 kB	09:37
par_all36_na.prm	64,8 kB	09:37
par_interface.prm	121,0 kB	09:37
Readme.txt	1,5 kB	09:37
toppar_all36_carb_glycolipid.str	24,3 kB	09:37
toppar_all36_carb_glycopeptide.str	14,7 kB	09:37
toppar_all36_carb_imlab.str	161,5 kB	09:37
toppar_all36_label_fluorophore.str	228,2 kB	09:37
toppar_all36_label_spin.str	97,7 kB	09:37
toppar_all36_lipid_archaeal.str	12,0 kB	09:37
toppar_all36_lipid_bacterial.str	6,0 kB	09:37
toppar_all36_lipid_cardiolipin.str	0 bytes	09:37
toppar_all36_lipid_cholesterol.str	45,4 kB	09:37
toppar_all36_lipid_dag.str	0 bytes	09:37
toppar_all36_lipid_detergent.str	5,2 kB	09:37
toppar_all36_lipid_ether.str	7,2 kB	09:37
toppar_all36_lipid_hmmm.str	644 bytes	09:37
toppar_all36_lipid_inositol.str	0 bytes	09:37
toppar_all36_lipid_lnp.str	14,2 kB	09:37

- Runnscript python in folder 000_eq:

  python 000.2_updateCenters.py

- Open each folder to running every script below:

  namd3 000.1_eq.conf > 000.1_eq.log

  namd3 001_abf_1.conf > 001_abf_1.log

  namd3 002_abf_1.conf > 002_abf_1.log

  namd3 003_abf_1.conf > 003_abf_1.log

  namd3 004_abf_1.conf > 004_abf_1.log

  namd3 005_abf_1.conf > 005_abf_1.log

  namd3 006_abf_1.conf > 006_abf_1.log

- Open folder 007_r
- Open VMD > Extension > Tkconsole
- Run this script in terminal Tkconsole:

```
source 007.0_solvate.tcl
```

- You will get file 3 file namely: complex_largeBox.pdb, complex_largeBox.xyz and complex_largeBox.psf
- running this command:

```
namd3 007.1_eq.conf > 007.1_eq.log
namd3 007.2_abf_1.conf > 007.2_abf_1.log
```

- Open folder namely 008_RMSDUnbound
- Open VMD > Extension > Tkconsole
- Run this script in terminal Tkconsole:

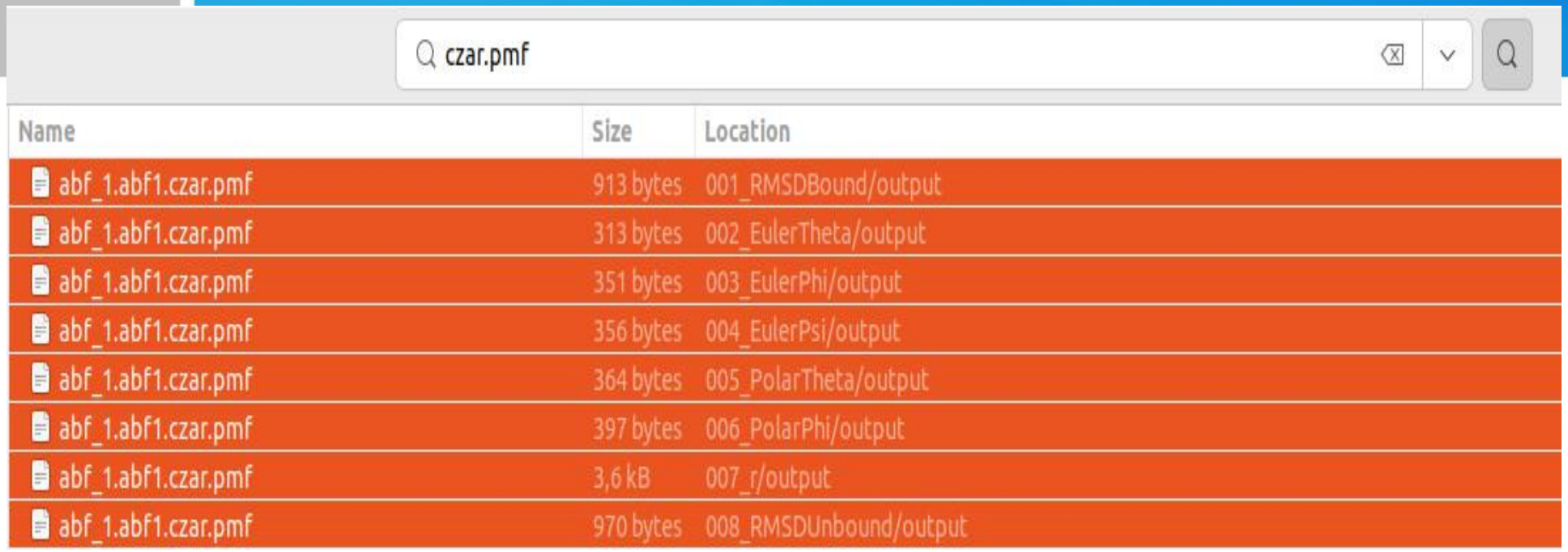
  source 008.0.1_removeProtein.tcl
- Run this command:

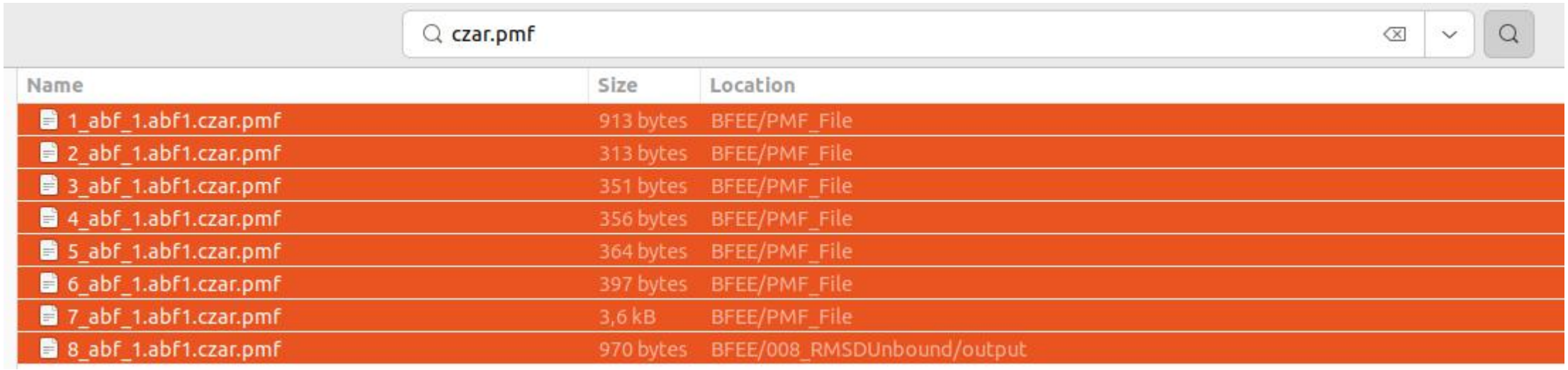
  namd3 008.1_eq.conf > 008.1_eq.log
- Open VMD > Extension > Tkconsole
- Run this script in terminal Tkconsole:

  source 008.0.2_neutrilize.tcl
- Run this command:

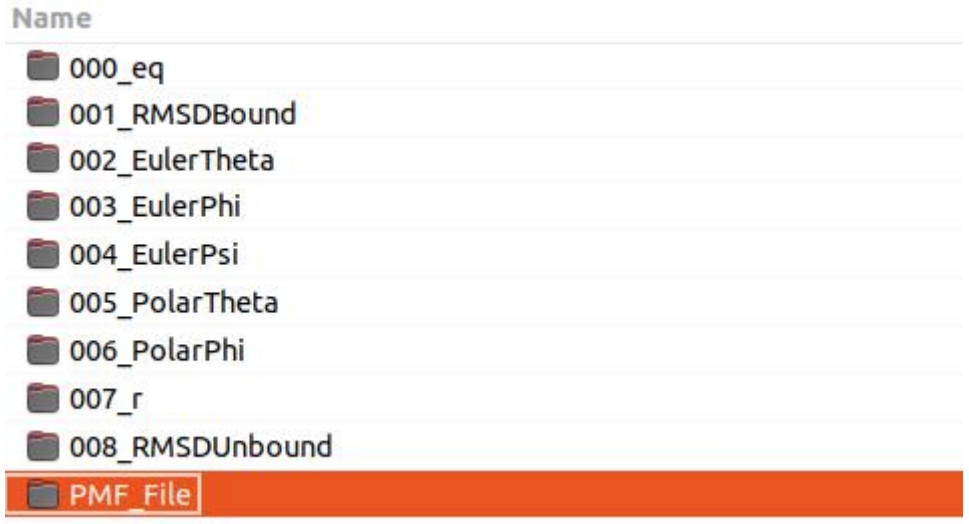
  namd3 008.2_abf_1.conf > 008.2_abf_1.log

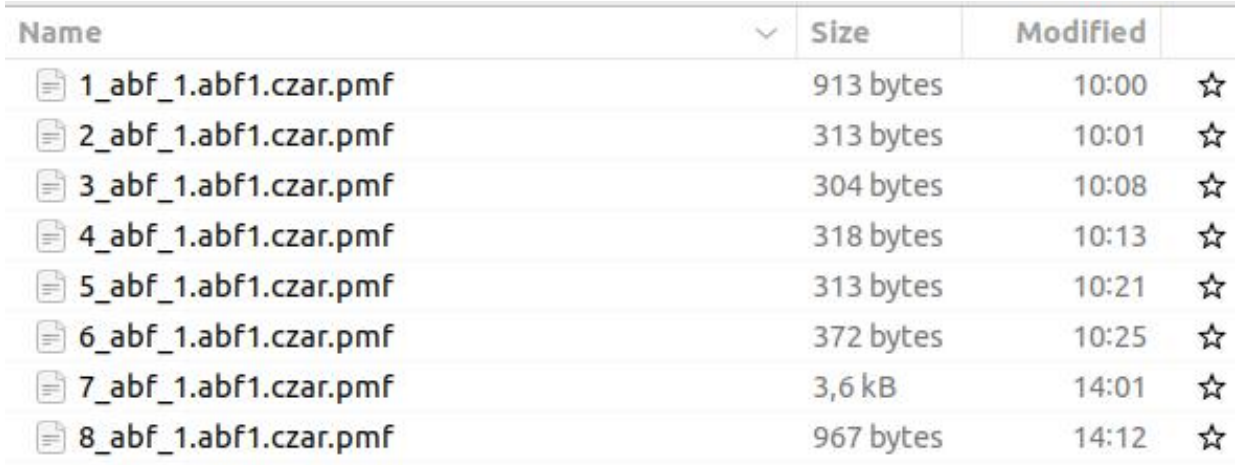
- Open output folder each folder, search file namely: **abf_1.abf1.czar.pmf** and rename file such as: 8_abf_1.abf1.czar.pmf the number 8 comes from the folder name 008_RMSDUnbound.


czar.pmf
Name
Size
Location
abf_1.abf1.czar.pmf 913 bytes 001_RMSDBound/output
abf_1.abf1.czar.pmf 313 bytes 002_EulerTheta/output
abf_1.abf1.czar.pmf 351 bytes 003_EulerPhi/output
abf_1.abf1.czar.pmf 356 bytes 004_EulerPsi/output
abf_1.abf1.czar.pmf 364 bytes 005_PolarTheta/output
abf_1.abf1.czar.pmf 397 bytes 006_PolarPhi/output
abf_1.abf1.czar.pmf 3,6 kB 007_r/output
abf_1.abf1.czar.pmf 970 bytes 008_RMSDUnbound/output

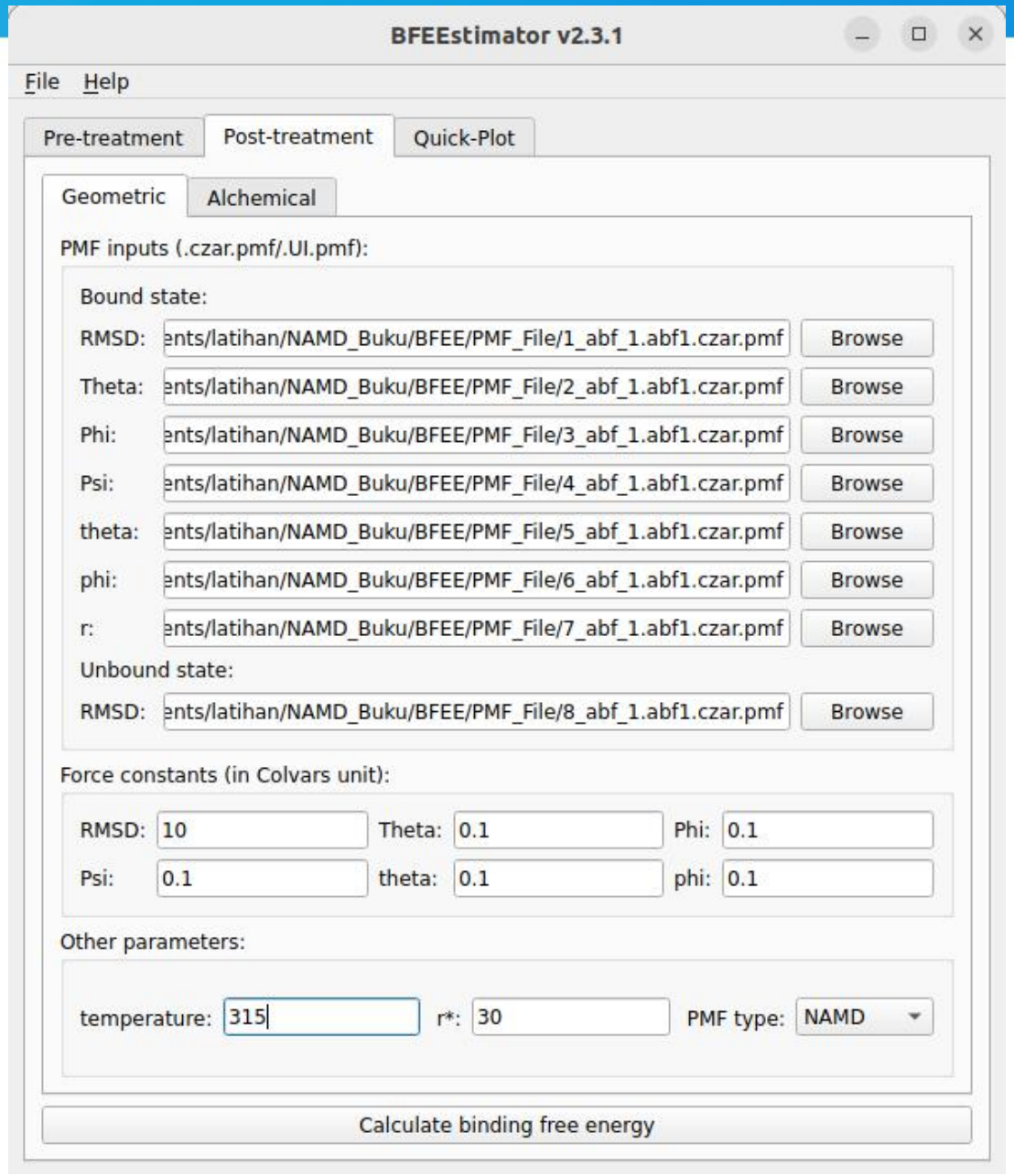

czar.pmf
Name
Size
Location
1_abf_1.abf1.czar.pmf 913 bytes BFEE/PMF_File
2_abf_1.abf1.czar.pmf 313 bytes BFEE/PMF_File
3_abf_1.abf1.czar.pmf 351 bytes BFEE/PMF_File
4_abf_1.abf1.czar.pmf 356 bytes BFEE/PMF_File
5_abf_1.abf1.czar.pmf 364 bytes BFEE/PMF_File
6_abf_1.abf1.czar.pmf 397 bytes BFEE/PMF_File
7_abf_1.abf1.czar.pmf 3,6 kB BFEE/PMF_File
8_abf_1.abf1.czar.pmf 970 bytes BFEE/008_RMSDUnbound/output

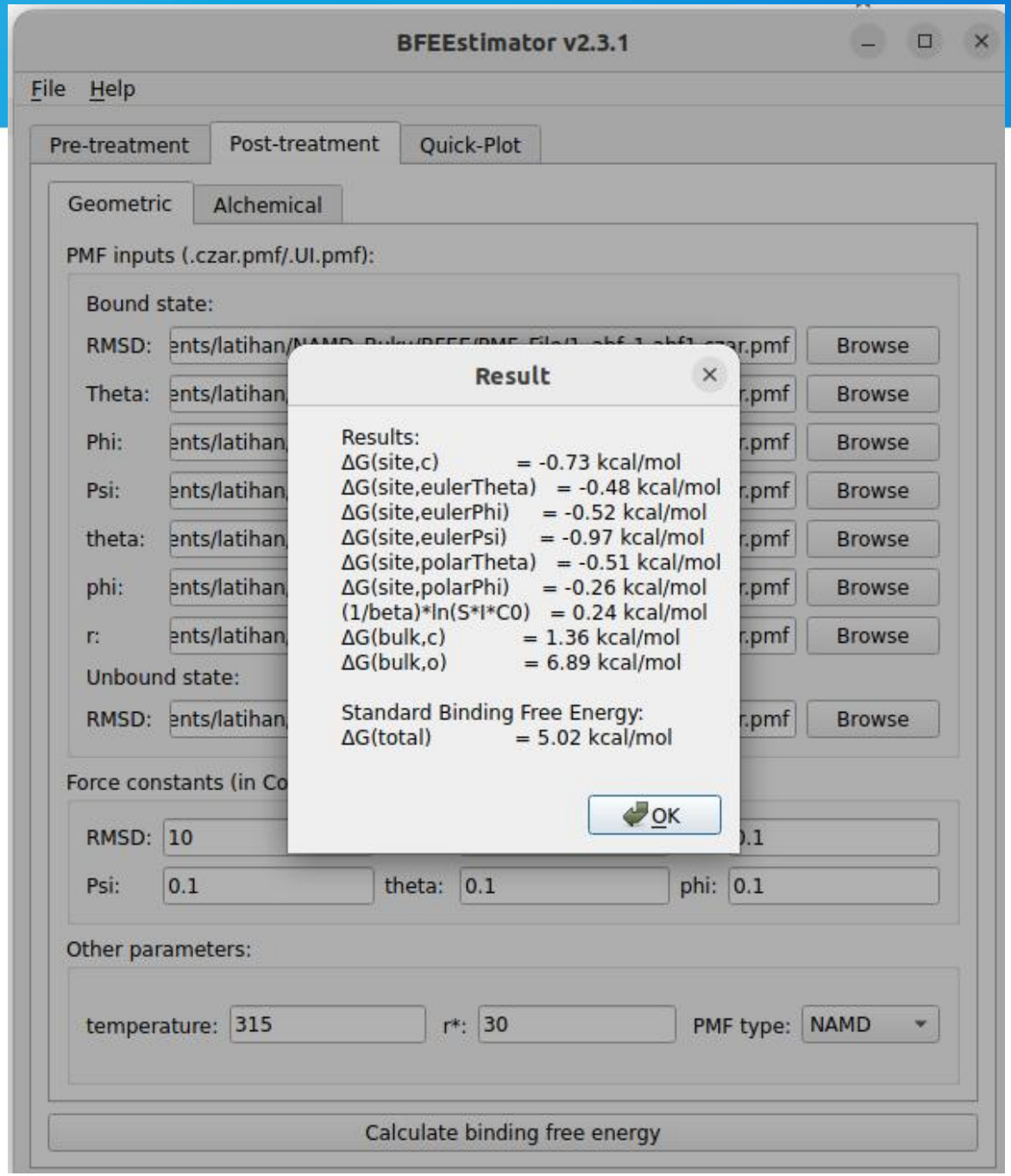
- Make folder namely PMF_file to store all file .czar.pmf file, After changing the name of the file abf_1.abf1.czar.pmf each folder collect the files in one folder as shown below





Click Calculate binding free energy




BFEEstimator v2.3.1
File
Help
Pre-treatment
Post-treatment
Quick-Plot
Geometric
Alchemical
PMF inputs (.czar.pmf/.UI.pmf):
Bound state:
RMSD:
Theta:
Phi:
Psi:
theta:
phi:
r:
Unbound state:
RMSD:
Browse
Result
Results:
ΔG(site,c) = -0.73 kcal/mol
ΔG(site,eulerTheta) = -0.48 kcal/mol
ΔG(site,eulerPhi) = -0.52 kcal/mol
ΔG(site,eulerPsi) = -0.97 kcal/mol
ΔG(site,polarTheta) = -0.51 kcal/mol
ΔG(site,polarPhi) = -0.26 kcal/mol
(1/beta)*ln(S*I*C0) = 0.24 kcal/mol
ΔG(bulk,c) = 1.36 kcal/mol
ΔG(bulk,o) = 6.89 kcal/mol
Standard Binding Free Energy:
ΔG(total) = 5.02 kcal/mol
OK
Force constants (in Co
RMSD: 10
Psi: 0.1
theta: 0.1
phi: 0.1
Other parameters:
temperature: 315
r*: 30
PMF type: NAMD
Calculate binding free energy