Hackathon Problem: Protein-Ligand Docking and Binding

Reminder, run these calculations in your /gscratch!

In the class, we did protein-ligand docking and binding for the protein kinase (RCSB PDB code: 3QKK) with the ligand fisetin. For the hackathon, you will be running a similar system and running identical calculations from the in-class lecture.

To get started, we have provided information regarding the protein and various ligands with binding free-energy information (see the table below). **Your deliverable for this project is a final document and a presentation that includes information about docking multiple-ligands on the protein (ADRB2) at Trp286 site; justifying the choice of one ligand for binding that you will choose to simulate next; choice of collective variable; running a funnel metadynamics simulation to get the binding affinity; plotting free energy as a function of collective variable; and discuss its comparison with the experimental values shown below.** Find relevant publications and site them and perform the calculations on the GPU for the faster calculations.

Target	Ligand	ΔG_{exp} (kcal/mol)
ADRB2	ICI-118551	−12.8
ADRB2	Alprenolol	−12.7
ADRB2	Carvedilol	−12.7
ADRB2	Isoprenaline	−9.9
ADRB2	BI-167107	−14.3
ADRB2	Adrenaline	−9.8

Follow the following steps to achieve your deliverables:

- You can find protein PDB at: **https://alphafold.com/entry/P07550**
- You can fetch the ligands from PubChem database.

Step 1. Multiple-ligand molecular docking

1.1. Choose one pair of protein from the given table along with the ligand.
1.2. Once you have chosen the protein and ligand find the binding pocket and perform the protein-ligand docking using Chimera and AutoDock Vina as we discussed in workshop Day 3.
1.3. Once the docking is completed get the PDB file of the protein-ligand complex for further processing.
1.4. Along with the PDB file of the protein–ligand complex gets the binding affinity from the Auto Dock vina and compare it with the experimental value mentioned in the above table.
1.5. In addition, to the binding affinity observe the hydrogen bonding interactions of the protein–ligand docking in chimera and post the results.

Step 2. MD Simulations – to set your system for funnel metadynamics

2.1. Once the protein-ligand complex is prepared, create the topology file using CHARMM force field and TIP3P water model.
2.2. Once the topology is created, create a cubic box where the minimum distance between the molecule and the edge of the box is 3.0 nm.
2.3. Once the box is created follow the protocol as discussed in the workshop, create box, solvate the molecule, ionize the molecule and do the energy minimization and plot how total energy changes with time.
2.4. After the energy minimization, perform the NVT equilibration and plot the Temperature vs Time.
2.5. After performing NVT equilibrium, perform NPT equilibration and plot how density changes with time.

Step 3. <u>Funnel Metadynamics Simulation</u>

3.1. Using the equilibrated structure as above (nvt.gro) construct the funnel restraint in VMD and obtain the funnel meta dynamics configuration file (plumed.dat) and create the reference PDB structure as well (ref.pdb) with “grep command”. Here, make sure you have provided all the information correctly.
3.2. Once the configuration file is prepared perform the funnel meta dynamics simulation (as it is very difficult to reach the convergence in few days, run your simulation as much as you can in the allocated hackathon time and go to the next step).
3.3. Using the graphical user interface analysis tool in VMD and attach the screenshots.
3.4. Extract the free energy data from the HILLS file and plot it.
3.5. Plot how free energy changes with time.
3.6. Finally, calculate the binding free energy and compare it with the experimental values.