**HECSP-NMRScore\_P tutorial:**

Suppose you put the folder “hecsp” at “/Users/xxx/”. Then add the following lines to bash file:

## Setting PATH for HECSP NMRScore\_P

export PYTHONPATH="/Users/xxx/hecsp:$PYTHONPATH"

export PATH="/Users/xxx/hecsp/calHcsp:$PATH"

export PATH

export HECSPHOME="/Users/xxx/hecsp"

Take the Glide docking result of 1J5I model 1 as an example. The pose viewer file generated by Glide docking is “dock\_pv.maegz”. The first molecule in the file is the receptor, and the following entries are ligand poses.

Step 1: Extract receptor from “dock\_pv.maegz” into “receptor.pdb” and ligand poses into “poses.mol2” (scripts available with Schrodinger package)

**$SCHRODINGER/utilities/pdbconvert -imae dock\_pv.maegz -opdb receptor.pdb -n 1**

Remember to delete the cap residues ACE or NMA if present in the pdb file.

**$SCHRODINGER/utilities/mol2convert -imae dock\_pv.maegz -omol2 poses.mol2 -n 2:**

Step 2: split the poses.mol2 file into separate mol2 files (script available in HECSP package)

**splitmol2.py poses.mol2 pose**

It will give you pose\_001.mol2, pose\_002.mol2 …….

Step 3: Generate mol2 file with Gasteiger charges by simply run a shell script “run.sh”, which needs the antechamber program from the Ambertools package (free)

**“for file in pose\_\*.mol2**

**do**

**base=${file%.mol2}**

**num=`echo "$file" | cut -d'.' -f1 | cut -d'\_' -f2`**

**$AMBERHOME/bin/antechamber -fi mol2 -fo mol2 -i $base.mol2 -o gas\_pose\_"$num".mol2 -j 0 -pf y -c gas -nc 0**

**done”**

It will give you gas\_pose\_001.mol2, gas\_pose\_002.mol2 …….

Step 4: Prepare “exp\_csp.txt” file in the same folder, which contains all the experimental CSPs:

Step 5:  Determine the residues or specific protons that are significantly perturbed. The result is used for the NMRScore\_P calculation in the next step (script available in the HECSP package).

**big\_H.py receptor.pdb pose\_001.mol2 10**

The last integer “10” in the command line is to define the adopted distance cutoff.

Step 6: Calculate NMRScore\_P from receptor.pdb and gas\_pose\_001.mol2 (script available in the HECSP package):

**nmrscorep.py -p receptor.pdb -l gas\_pose\_001.mol2 -s gloocv --resid 4,32,33,34,35,36,43,44,46,47,53,69,75,78,96,97,98,99,102,103,104,106,108**

Two files “NMRScore\_result.txt “ & “receptor\_gas\_pose\_001\_csp” will be generated. One can easily write a shell script to calculate hundreds of protein ligand pairs.