Supporting Information Available

The files are provided in the Supporting Information:

Prepared protein-ligand crystal structures `xtal_prepped.tar.gz`

Contains the reference protein-ligand crystal structures that have been aligned and prepared through the 'Protein Preparation Wizard' tool.

RMSD analysis: `scripts.tar.gz`

Contains the scripts `RMSD-analysis.tcl` and `plot-RMSD.py` used for RMSD analysis. A `README.txt` file has been provided which details how to generate the RMSD/time and Colormap plots found in the paper. VMD and the trajectories from the MD simulations are required to perform the analysis. Only trajectories for benzene-nhexylbenzene and benzene-nhexylbenzene simulations have been included and are within the archived data files.

FEP data: `C-I_default.tar.gz`, `C-I_pPREST.tar.gz`, `C-O_default.tar.gz`, `C-O_pREST.tar.gz`, `EXP_default.tar.gz,` and `EXP_pREST.tar.gz`

Inside each archived directory contains directories named by the ligands involved in the transformation and the protein conformation from which the simulations were started with. For example, '`C-O_default/benzene-nhexylbenzene_C/`' corresponds to the FEP calculation of benzene to n-hexylbenzene, starting from the protein closed state, while using the default protocol. Within each FEP calculation directory are the input `.mae` files containing the protein/ligand structures, the simulation configuration `.msj` files, and a `README.txt`. The `README.txt` file contains information about each FEP calculation such as the protein starting conformation, ligands involved, forcefields used, set label, free energies corresponding to the complex/solvent simulations, and the final calculated free energy. The output data for each FEP calculation is contained within the '`*_fep1/`', where inside the '`*_complex_12/`' or '`*_solvent_13/`' folders are all output files used to calculate the free energies. The final calculated free energy is writ-

ten in a '`result`' file, where extended simulations will contain two result files for the calculated free energy when analyzing the 0-5ns time frame and the 15-25ns or 40-55ns time frame.

For FEP calculations where an RMSD analysis was performed in the paper (ex.'`C-O_default/benzene` an '`RMSD-analysis/`' folder is provided. Inside are '`replica#.rmsd`' files that contain the set of RMSDs relative to each protein conformation over each replica's trajectory. The resulting RMSD/time and Colormap plots can be found inside the '`RMSD-analysis/plots/`' folders. The trajectories are also provided inside the output '`*_fep1/`' folder within the '`*_complex_10/`' directory. `C-O_pREST.tar.gz` has been split due to file size limitations. It can be rejoined by executing '`cat C-O_pREST.tar.gz.*` > `C-O_pREST.tar.gz`'

This material is available free of charge via the Internet at `http://pubs.acs.org/`.