Phenix *Python-based Hierarchical ENvironment for Integrated Xtallography*

Tutorial: Fit Biomolecules into Cryo-EM Maps using MD Simulation (commandline)

Contents

Overview

This tutorial will show you how to fit biomolecule atomic structures into cryo-EM maps using molecular dynamics simulation with PHENIX commandline

For GUI execution, please see the cryo_fit_gui_tutorial

Theoretical explanation of cryo_fit is here

For installation of cryo_fit, please see the installation notes for cryo_fit

Input files

<initial_model> and <target_map>

Initial Model

Available format: .cif and .pdb

The initial model is a guide or template structure (CIF/mmCIF/pdb) that is close to a target cryo EM map structurally.

You can use either map_to_model or UCSF chimera (Tools -> Volume Data -> Fit in Map)) to prepare the initial model.

Target Map

Available format: .ccp4 and .map (MRC style in binary file) and .sit (Situs style in text file)

Running the program

```
% phenix.cryo_fit <initial_model> <target_map>
```

example command line:

```
% phenix.cryo_fit tRNA.pdb tRNA.map
```

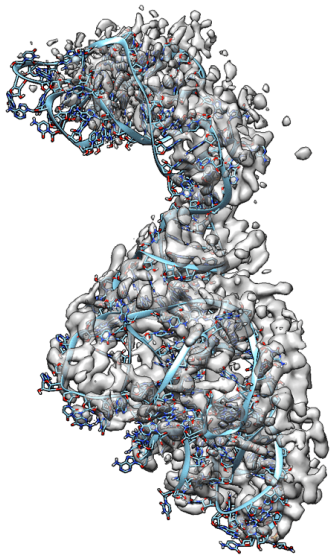
Output

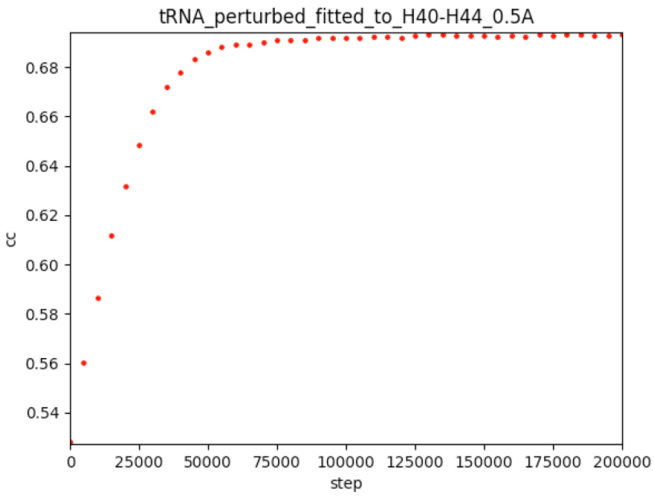
A final cryo_fitted structure: steps/8_cryo_fit/cryo_fitted.pdb (and cryo_fitted.gro for vmd visualization)

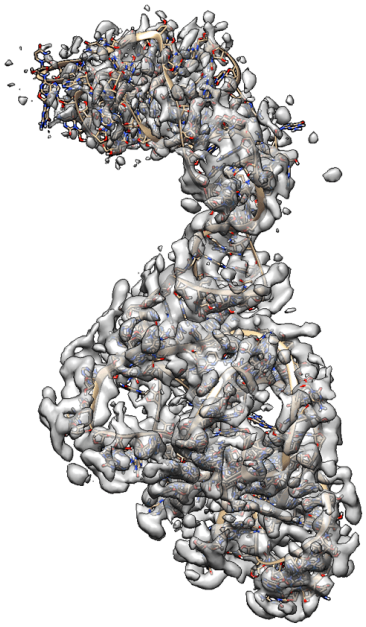
Correlation coefficients (CC) between cryo_fitted structures and cryo-EM maps: steps/8_cryo_fit/cc_record

Examples

cryo_fit example with tRNA







Before cryo_fit
(cc = 0.53)

After cryo_fit
(cc = 0.69)

Limitation

gromacs4.5.5 seems to not handle H2O (water) heteroatom. cryo_fit will remove water molecules (if any) from the input .cif/.pdb and fit to cryo electron microscopy map.

cryo_fit doesn't handle non-canonical "residue"s such as 7C4, BMA, GDP, ILX, NAG, SEP, TRX. The cryo_fit will simply erase those residues.

Reference

S. Kirmizialtin, J. Loerke, E. Behrmann, C. MT. Spahn, K. Y Sanbonmatsu, Using Molecular Simulation to Model High-Resolution Cryo-EM Reconstructions, Methods Enzymol., 558, 2015, 497-514

Options

All options will be used as default if unspecified. Gromacs expert users are welcome to customize those options if they wish.

List of most useful options

Option	Default value	Description of inputs and uses

emweight_multiply_by	8	Multiply by this number to the number of atoms for weight for cryo-EM map bias. For example, emweight = (number of atoms in gro file) x (emweight_multiply_by which is 8) The higher the weight, the stronger bias toward EM map rather than MD force field and stereochemistry preserving constraints. If user's map has a better resolution, higher value of emweight_multiply_by is recommended since map has much information. If user's map has have a worse resolution, lower value of emweight_multiply_by is recommended for more likely geometry. If CC (correlation coefficient) needs to be improved faster, higher number of emweight_multiply_by is recommended.
number_of_cores_to_use	max cores	Specify number of cores for minimization and cryo_fit. If it is not specified, or max is chosen, the cryo_fit will try to use most cores automatically (up to 16)
number_of_steps_for_cryo_fit	None	This is the initial number of steps for cryo_fit. Eventually, cryo_fit will increase it depending on molecule size and cc trend. For tutorial files, this will be 70,000
number_of_steps_for_minimization	None	Specify number of steps for minimization. If this is left blank, cryo_fit will estimate it depending on molecule size.number of steps for cryo_fit. Enough minimization will prevent "blow-up" during MD simulation later.