RoseTTAFold and Protein Structure Prediction

CCATS Group

RoseTTAFold Workflow

1. Input sequence
   - Amino Acid Sequence in FASTA format (.fa)
2. RoseTTAFold Program
   - Conda Environments (GPU, Folding; ~3.8 Gb)
   - RoseTTAFold Software (~6.9 Gb)
   - PyRoseTTA License (https://els2.comotion.uw.edu/product/pyrosetta)
3. RoseTTAFold Databases (~460 Gb)
   - Uniref30, Reduced BFD/Mgnify, Structure Templates (RCSB)

If all goes well, you get 5 predicted monomer structures

03_rosettafold

- On OSCER, copy this directory to your home:

  cp /home/van/MacroMol_Modelling_Tutorial/03_rosettafold .

- The folder is organized as follow:

> 03_rosettafold

  > Results

  > Tutorial

    > 00_monomer_pred

    > 01_complex_pred

03_rosettafold/Tutorial/00_monomer_pred

- This folder contains:
  - bax.fa (Bax)
  - rf_monomer_pred.slurm
    - SLURM script to run RoseTTAFold
  - To run:

    sbatch rf_monomer_pred.slurm

bax.fa (Bax FASTA sequence)

```
>bax
MDGSGEQPRGGGPTSSEQIMKTGALLLQGFIQDRAGRMGGEAPE
LALDPVPQDASTKKLSECLKRIGDELDSNMELQRMIAAVDTDSP
REVFFRVAADMFSDGNFNWGRVVALFYFASKLVLKALCTKVPEL
IRTIMGWTLDFLRERLLGWIQDQGGWDGLLSYFGTPTWQTVTIF
VAGVLTASLTIWKKMG
```

rf_monomer_pred.slurm

```
#!/bin/bash
#SBATCH --partition=gpu
#SBATCH --output=%j.out
#SBATCH --error=%j.err
#SBATCH --time=48:00:00
#SBATCH --job-name=rosetta

date

module load CUDA/11.3.1
ln -sf /home/van/Programs/RoseTTAFold/run_pyrosetta_ver.sh

./run_pyrosetta_ver.sh bax.fa .

date

```

SLURM Allocation requests

Load CUDA library (for GPU)

Create link of RoseTTAFold Script and run the program taking the FASTA sequence as input 1 and “.” as the working directory

run_pyrosetta.sh (made by Van)

RF *run_pyrosetta.sh* [input FASTA] [working directory]

conda activate RoseTTAFold

Prepare MSA
RF *make_msa.sh* [input FASTA] > [input.a3m]

Predict Secondary Structure
RF *make_ss.sh* [input.a3m] > [input.ss2]

Search Templates
Combine [input.a3m] [input.ss2] > [input.msa0.ss2.a3m]
HHsearch [input.msa0.ss2.a3m] > [input.hhr] & [input.atab]

Predict Distance & Orientation
RF predict_pyrosetta.py \
[input.a3m] [input.hhr] [input.a3m] > [input.3track]

conda activate folding

Monomer Prediction
RF RosettaTR.py \
[input FASTA] [input.3track > [input.pdb]

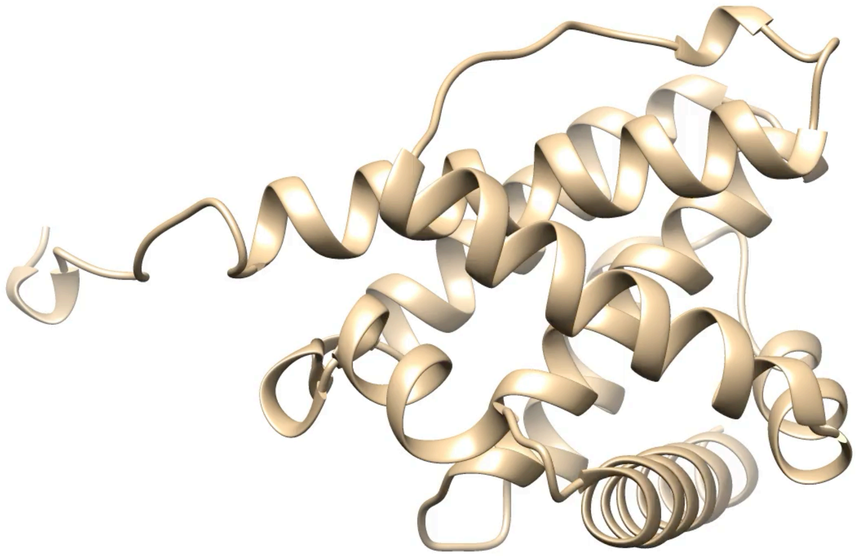
Error Estimation (DeepAccNet-MSA)
RF ErrorPredictorMSA.py \
[input.3track] > [input.crderr.pdb]

5 Predicted Models
(with separate PDB for error)

NOTE: **Bold** : program [name] : name of input (auto)
Italic : scripts RF : RoseTTAFold

Visualizing Model 1 Error on UCSF Chimera

- Download the models folder to your local computer
- Load model_1.crderr.pdb on Chimera
- Change atom coloring to error (found in B Factor column)
  - Select > Structure > protein
  - Action > Atom/bonds > Show
  - Tools > Depiction > Render by Attribute



03_rosettafold/Tutorial/01_complex_pred.slurm

- This folder contains:
  - paparde.fa (from Alphafold Tutorial)
- Need to make two separate FASTA files:

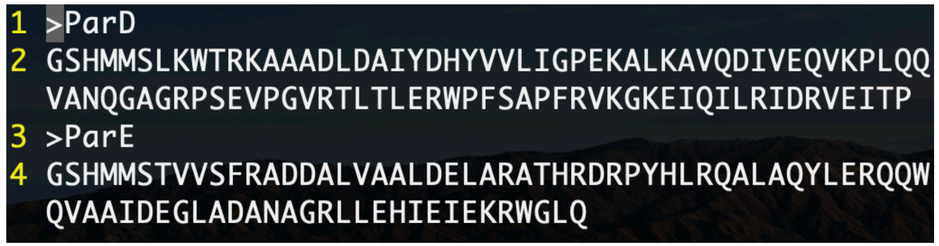
  head -n 2 paparde.fasta > pard.fa

  tail -n 2 paparde.fasta > pare.fa
  - rf_complex_pred.slurm
    - SLURM script to run RoseTTAFold

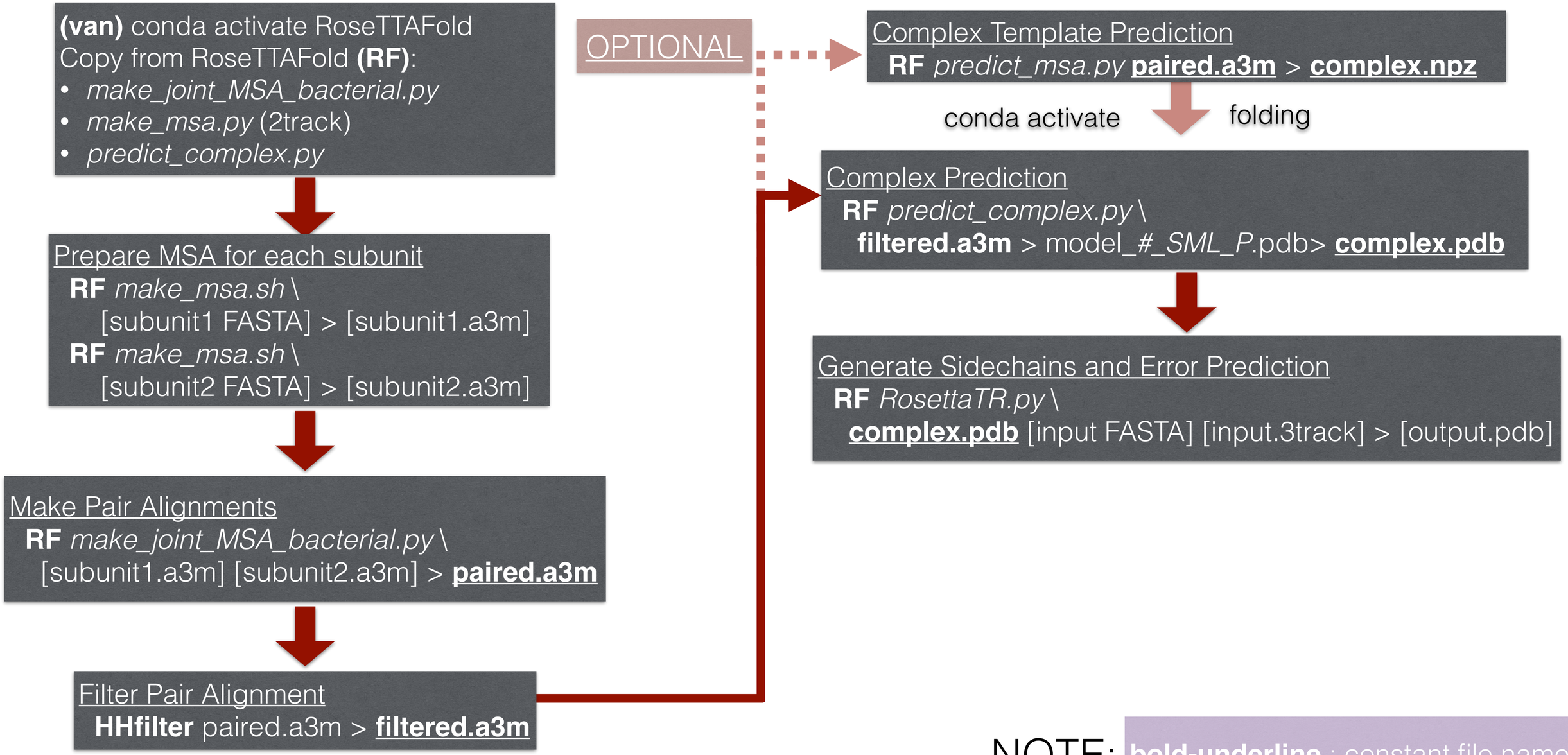
- To Run:

  sbatch rf_complex.slurm

paparde.fa (from Alphafold



rf_complex_pred.slurm (made by Van)



NOTE: **bold-underline** : constant file name