

PICRUST2 Workflow

Contents

PICRUST2 wiki

1. Installing PICRUST

This was already done and does not need to be done again.

```
mamba create -n picrust2 -c bioconda -c conda-forge picrust2=2.5.2
```

2. Required input files

Two required files

- FASTA of amplicon sequences variants (ASVs; i.e. your representative sequences, not your raw reads)
- Tab-delimited table with ASV ids as the first column and sample abundances as all subsequent columns

Example of both files :

```
## PhantomJS not found. You can install it with webshot::install_phantomjs(). If it is installed,
```

3. Running the full pipeline

Running picrust through command line (not R)

a. Activate the environment

```
mamba activate picrust2
```

b. Run the full default pipeline

```
picrust2_pipeline.py -s fasta_file.fna -i asv_file.tsv -o picrust2_out -p 12
```

Main options :

- **-s** PATH - FASTA of unaligned study sequences
- **-i** PATH - Input table of sequence abundances (BIOM, TSV, or mothur shared file format)
- **-o** PATH - Output folder
- **-p** INT: Number of processes to run in parallel.
- **--skip_norm** - Skip normalizing sequence abundances by predicted marker gene copy numbers (typically 16S rRNA genes). This step will be performed automatically unless this option is specified (added in v2.2.0-b).
- **--remove_intermediate** - Remove the intermediate outfiles of the sequence placement and pathway inference steps.
- **--verbose** - If specified, print out wrapped commands to screen.

See wiki for further options.

4. Key output files

- **EC_metagenome_out** - Folder containing unstratified EC number metagenome predictions
- **pred_metagenome_unstrat.tsv.gz** sequence table normalized by predicted 16S copy number abundances (seqtab_norm.tsv.gz), and the per-sample NSTI values weighted by the abundance of each ASV (weighted_nsti.tsv.gz).
- **KO_metagenome_out** - As EC_metagenome_out above, but for KO metagenomes.
- **pathways_out** - Folder containing predicted pathway abundances and coverages per-sample, based on predicted EC number abundances.