

## Install

- **Native installation**
  - Install **Miniconda**
  - Install **QIIME 2**
  - Install **Snakemake & dependencies**

OR

- **Docker container**
  - Install **Docker Desktop**
  - **Download** Docker image
  - **Run** Docker container

## Setup

- **Clone** Tourmaline repository (directory) from GitHub
- **Initialize** directory from previous Tourmaline run (optional)
- Edit **config.yaml** file
- Link to **reference database**
- Organize **sequence files** and edit **fastq manifest file**
- Edit and link to **metadata file**

## Run

- **x\_denoise**
  - **x\_taxonomy\_unfiltered**
  - **x\_diversity\_unfiltered**
  - **x\_report\_unfiltered**
  - **x\_taxonomy\_filtered**
  - **x\_diversity\_filtered**
  - **x\_report\_filtered**
- (optional)

x = dada2\_pe | dada2\_se | deblur\_se

Example command:  
\$ snakemake dada2\_pe\_denoise

## Input

- **./ (top-level directory)**
- Snakefile
- config.yaml
- scripts/

- **./00-data/**
- metadata.tsv
- manifest\_pe.csv
- repseqs\_to\_filter\_{method}.tsv

## Output

- **./01-imported/**
- refseqs.qza
- reftax.qza
- fastq\_pe.qza
- fastq\_summary.qzv

- **./02-output-{method}-{filter}/**
- **00-table-repseqs/**
- table.qza
- table.qzv
- repseqs.qza
- repseqs.qzv

- **./02-output-{method}-{filter}/**
- **01-taxonomy/**
- taxonomy.qza
- taxonomy.qzv
- taxa\_barplot.qzv

- **./02-output-{method}-{filter}/**
- **02-alignment-tree/**
- aligned\_repseqs.qza
- rooted\_tree.qza
- rooted\_tree.qzv
- repseqs\_properties.tsv
- repseqs\_properties.pdf
- repseqs\_to\_filter\_outliers.tsv
- repseqs\_to\_filter\_unassigned.tsv

- **./02-output-{method}-{filter}/**
- **03-alpha-diversity/**
- rarefied\_table.qza
- alpha\_rarefaction.qzv
- \*\_vector.qza
- \*\_group\_significance.qzv

- **./02-output-{method}-{filter}/**
- **04-beta-diversity/**
- \*\_distance\_matrix.qza
- \*\_pcoa\_results.qza
- \*\_emperor.qzv

- **./03-reports/**
- metadata\_summary.md
- report\_{method}\_{filter}.md
- report\_{method}\_{filter}.html

Legend:

- Orange arrow = sequence of steps
- Purple arrow = output of manual setup
- Green arrow = output of Snakemake commands
- {method} = dada2-pe | dada2-se | deblur-se
- {filter} = unfiltered | filtered
- .qza = QIIME 2 artifact file
- .qzv = QIIME 2 visualization file