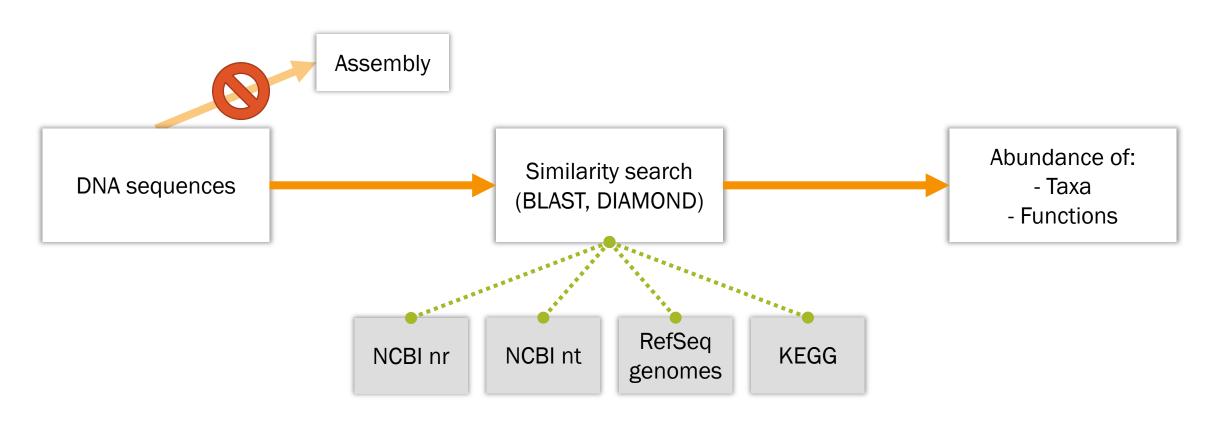
# **Environmental** metagenomics

Read-based taxonomic profiling



### What is read-based profiling?



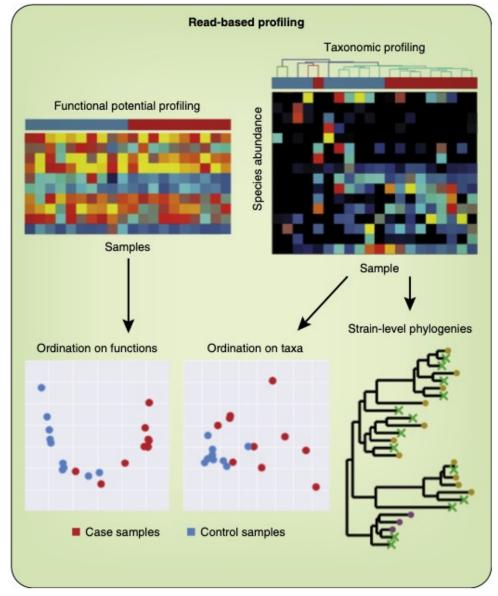
### Read-based profiling is

**Fast** 

Quantitative

Somewhat outdated

- Assembly-based are preferred
- Can give interesting preliminary insights
- Usually done as a "quick-and-dirty" estimate prior to assembly

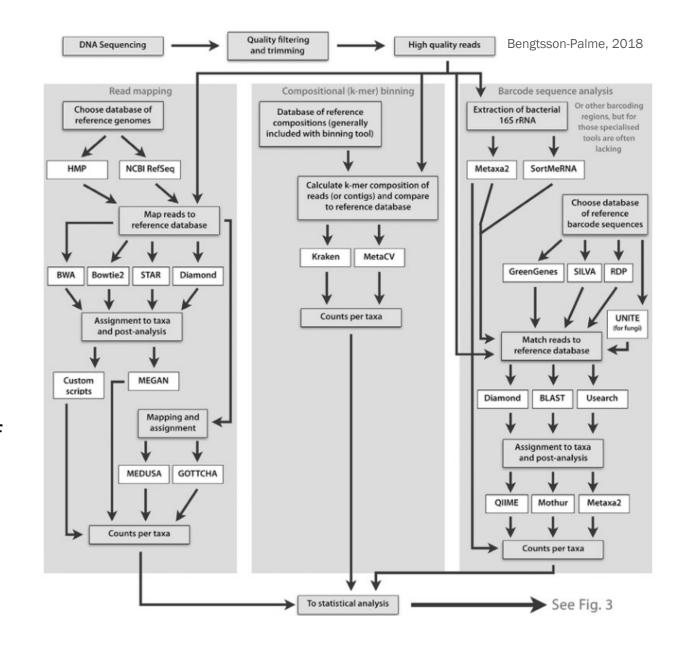




# Approaches to taxonomic profiling

## Read mapping and compositional binning

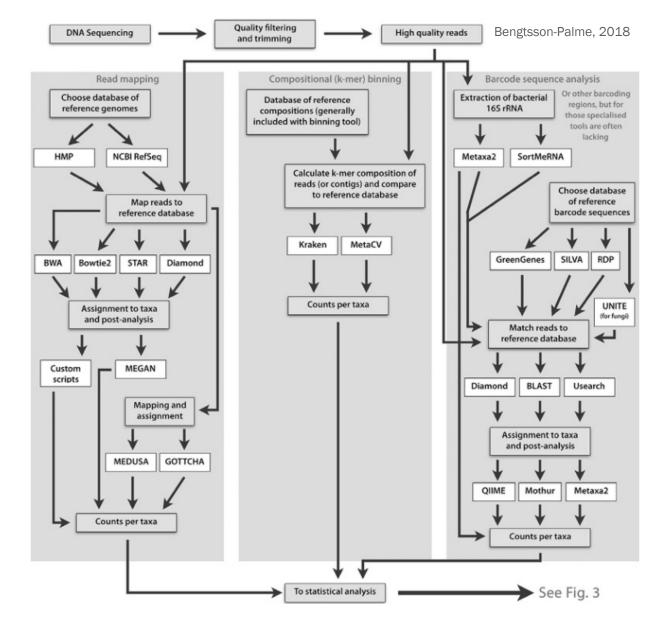
- Analysis of all reads
- Reference database of sequenced genomes
- Mapping: slow, requires lots of CPU and RAM
- Compositional binning: faster but less accurate



# Approaches to taxonomic profiling

#### Barcode sequence analysis

- Analysis of specific barcode genes (e.g. 16S rRNA)
- Curated database of barcode sequences (e.g. SILVA)
- Much faster than the other approaches, but provides lower resolution





# Approaches to taxonomic profiling: how to choose?

Analysis of all reads suffer from limited databases of reference genomes

 More suitable for environments that are better described (e.g. human gut) Analysis of barcode genes suffer from lower resolution

 More suitable for environments with a high fraction of unknown microorganisms (e.g. soil)

### Making sense of readbased analyses

#### Comparative analyses

#### **Statistics**

- Univariate (e.g. ANOVA of specific genes and taxa)
- Multivariate (e.g. PERMANOVA, ordination/clustering, Mantel test)

#### Normalization!

- Library size
- Bacterial content (e.g rpoB gene)

