metagWGS: a workflow to analyse short and long HiFi metagenomic reads Taxonomic profile HiFi vs Short reads assembly

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metagWGS: a workflow to analyse short and long HiFi metagenomic reads

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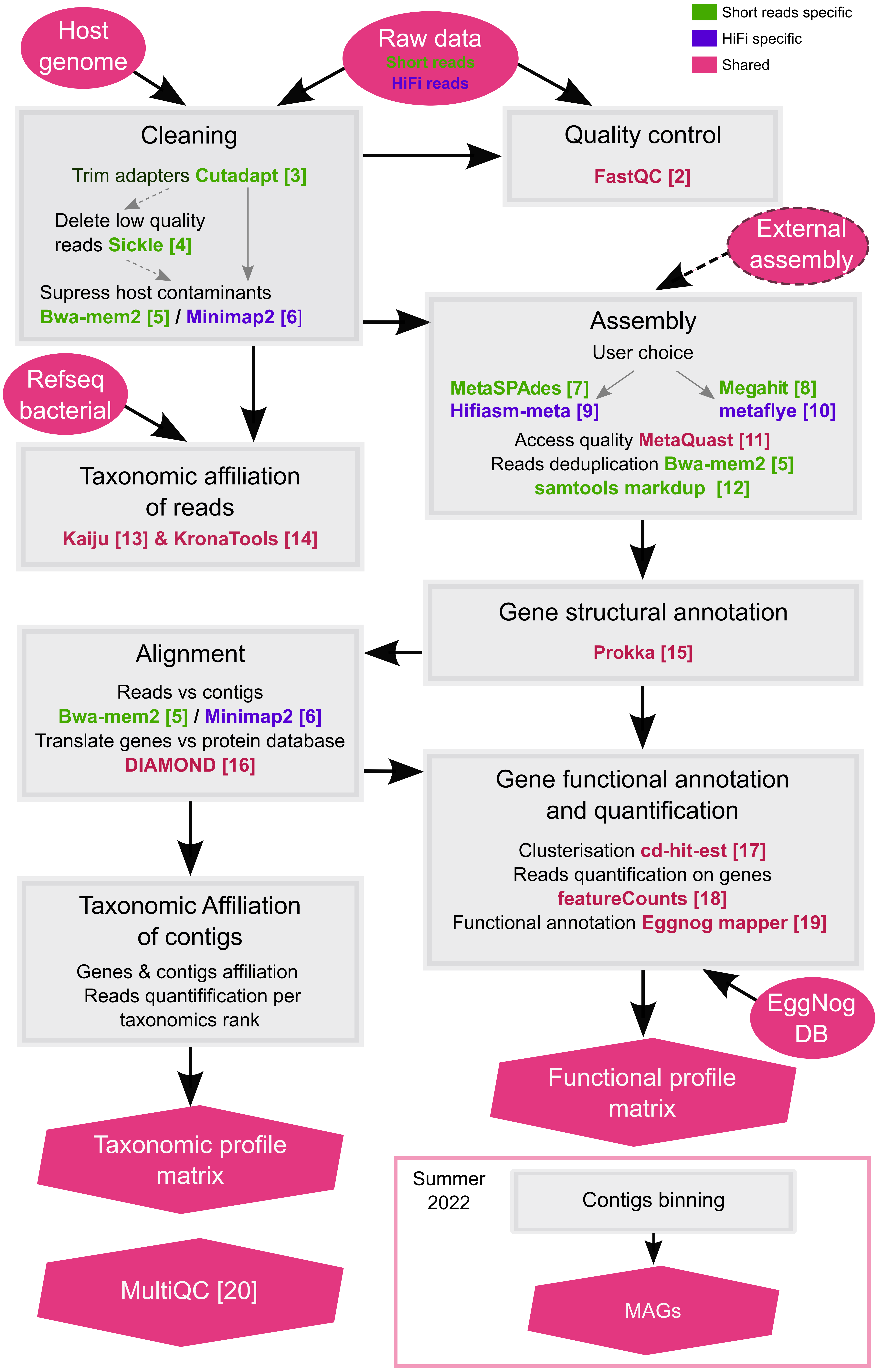
Production of whole metagenome assembly, functional and taxonomic profile



🟩 Short reads specific
🟪 HiFi specific
🟥 Shared

Host genome

Raw data
Short reads
HiFi reads

Cleaning
Trim adapters **Cutadapt [3]**
Delete low quality reads **Sickle [4]**
Supress host contaminants
Bwa-mem2 [5] / **Minimap2 [6]**

Quality control
FastQC [2]

External assembly

Assembly
User choice
MetaSPAdes [7] **Megahit [8]**
Hifiasm-meta [9] **metaflye [10]**
Access quality **MetaQuast [11]**
Reads deduplication **Bwa-mem2 [5]**
samtools markdup [12]

Refseq bacterial

Taxonomic affiliation of reads
Kaiju [13] & KronaTools [14]

Gene structural annotation
Prokka [15]

Alignment
Reads vs contigs
Bwa-mem2 [5] / **Minimap2 [6]**
Translate genes vs protein database
DIAMOND [16]

Gene functional annotation and quantification
Clusterisation **cd-hit-est [17]**
Reads quantification on genes
featureCounts [18]
Functional annotation **Eggnog mapper [19]**

Taxonomic Affiliation of contigs
Genes & contigs affiliation
Reads quantification per taxonomics rank

EggNog DB

Functional profile matrix

Taxonomic profile matrix

MultiQC [20]

Summer 2022

Contigs binning

MAGs

Workflow features

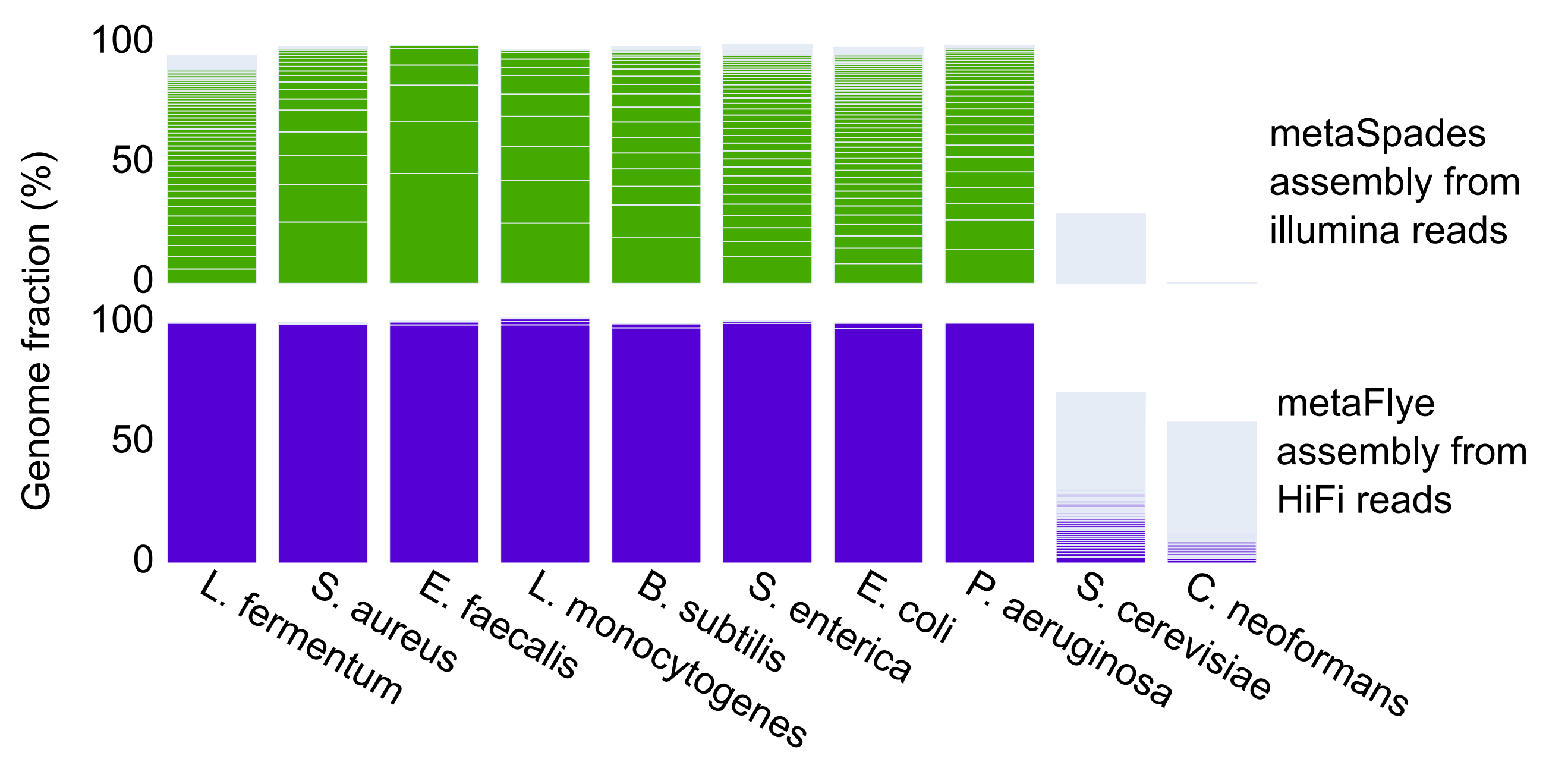
Type of NGS data:
whole genome shotgun sequencing (Illumina HiSeq3000 or NovaSeq, paired, 2*150bp ; PacBio HiFi reads, single-end)

Workflow:
a scalable and reproducible metagenomic analysis with a nextflow [1] pipeline using Singularity [21] container

Fully documented
https://forgemia.inra.fr/genotoul-bioinfo/metagwgs

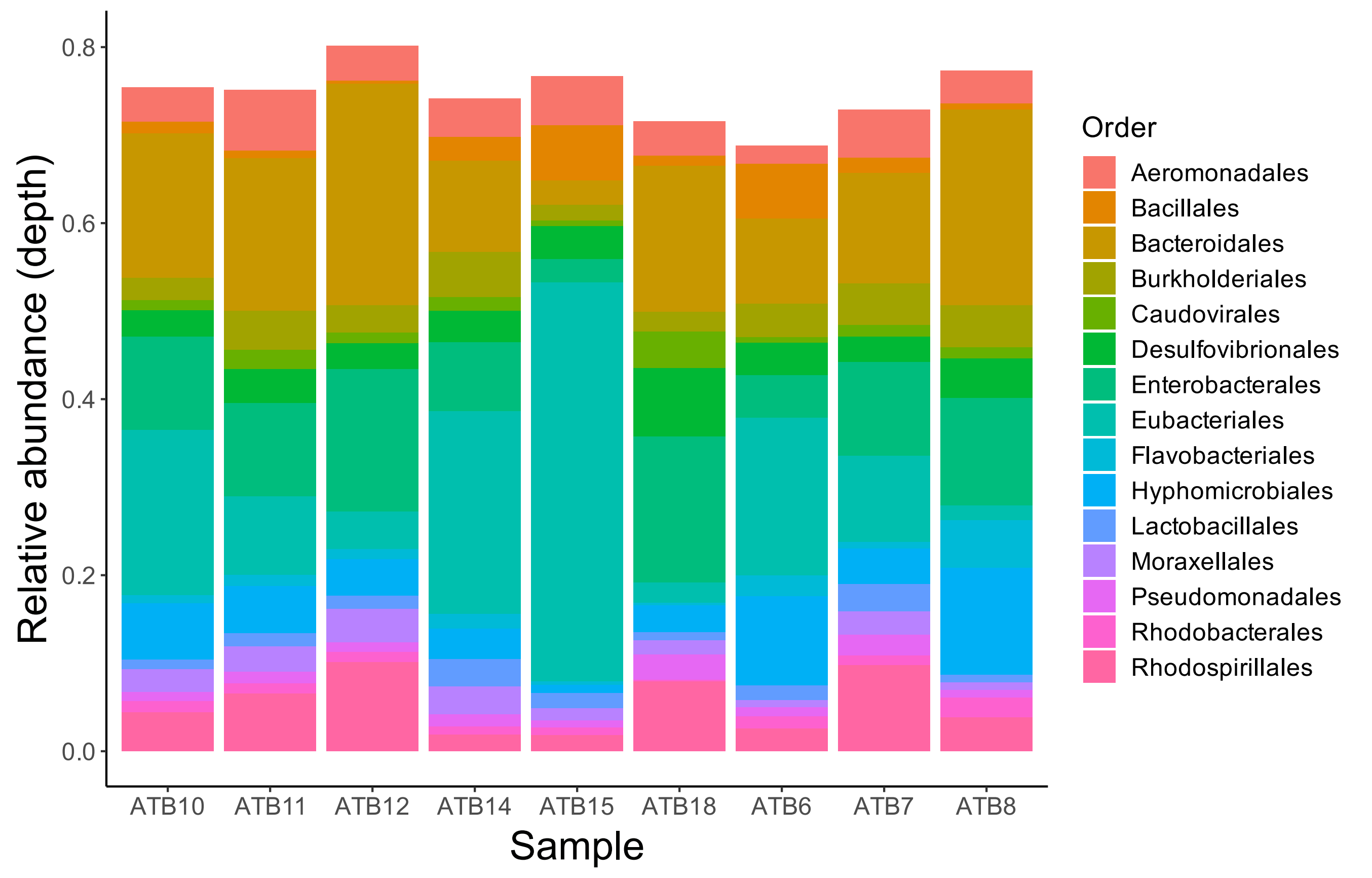
Results examples

HiFi vs Short reads assembly



Bacterial genomes of mock Zymobiomics are assembled in one contig by HiFi reads. The genome fraction is the fraction of the reference genomes covers by the assembly. Each block in a bar represents a contig. Bacterial genomes are well covered by all assemblies but Illumina assembly is much more fragmented.

Taxonomic profile



Relative abundance of the 15 major orders of nine biofilms grown in bioreactors from sewage (example from ATB biofilm project). Graph made from the taxonomic profile matrix generated by metagWGS. ATB15 is a sample that was exposed to ciprofloxacin (antibiotic) during the 14 days of culture.

Perspectives

- Annotation of Antibiotic Resistant Genes
- Annotation of mobilome genes
- Co-assembly

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