Shotgun Metagenomics — Preprocessing & Host Read Removal — Hands-on

Learn how to turn raw shotgun metagenomics fastq files into clean, host-depleted readsets suitable for assembly, taxonomic and functional profiling. This module covers quality control, adapter and quality trimming, low-complexity filtering, host read removal strategies and best practices for documenting preprocessing pipelines.

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Session Index

Session 1

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Raw Shotgun Reads, QC & Trimming

Shotgun read structures and metadata linkage

- paired end / single end
- lane, run and sample IDs
- fastq and index files

Quality control reports and decisions

- FastQC / MultiQC
- adapter detection
- overrepresented sequences

Adapter and quality trimming workflows

Trimmomatic / fastp / Cutadapt length and quality thresholds sliding window concepts

Session 2
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Host Read Removal & Mapping Strategies

Why host and human read removal matters

privacy and ethics coverage of microbial reads downstream sensitivity

Building host reference databases

human (GRCh) and model organisms custom host genomes index building in Bowtie2 / BWA

Mapping-based host read depletion

Bowtie2 / BWA workflows alignment parameters keeping unaligned reads

Session 3
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Contaminant Depletion & Clean Readsets

Integrated host and contaminant removal tools

KneadData / Sunbeam / nf-core combining trimming + mapping reference panels for contaminants

Low complexity, duplicates and quality filters

complexity filters duplicate removal options length and quality cutoffs

QC of host-depleted readsets and reporting

pre vs post read counts mapping fractions logs and

MultiQC summaries

Session 4

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Mini Capstone: Preprocessing Pipeline Build

Designing a reproducible preprocessing pipeline

theory plus guided practical

From raw fastq to host-depleted clean reads

QC, trimming and host mapping | contaminant filtering | export for assembly / profiling

Deliverables and documentation checklist

pipeline script / notebook | logs and QC report bundle | FAIR-ready metadata for preprocessing