

MEGAN6

Metagenome Analyzer

What are some different sequencing approaches?

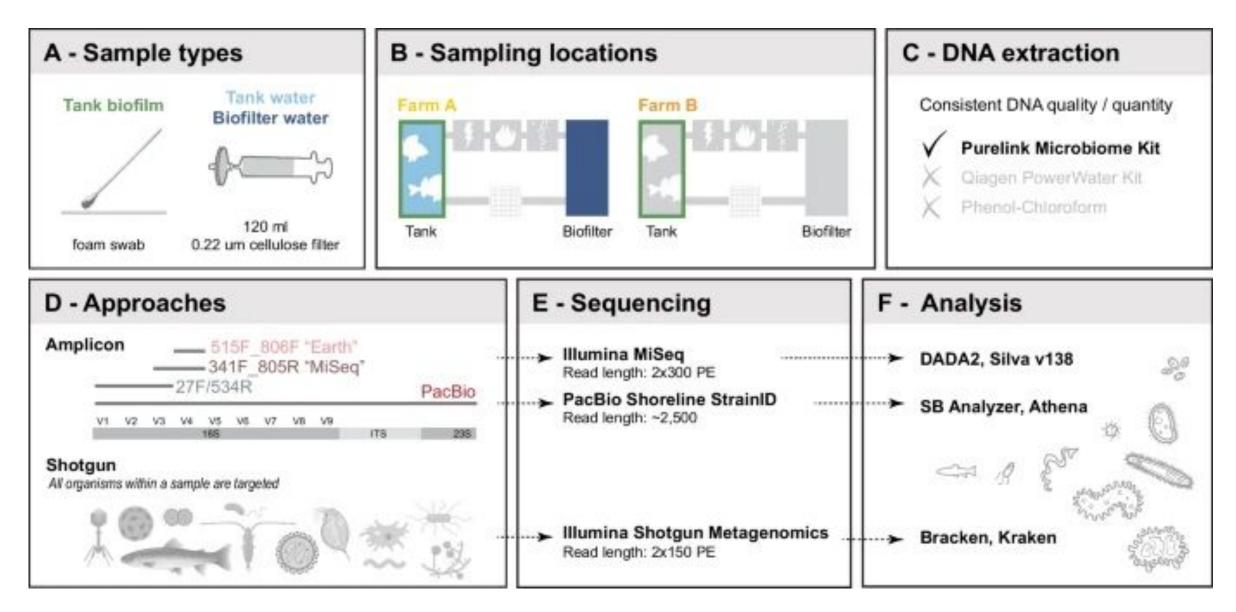


Figure Credit: Rieder et al. 2023, Environmental Microbiome

What are some different sequencing approaches?

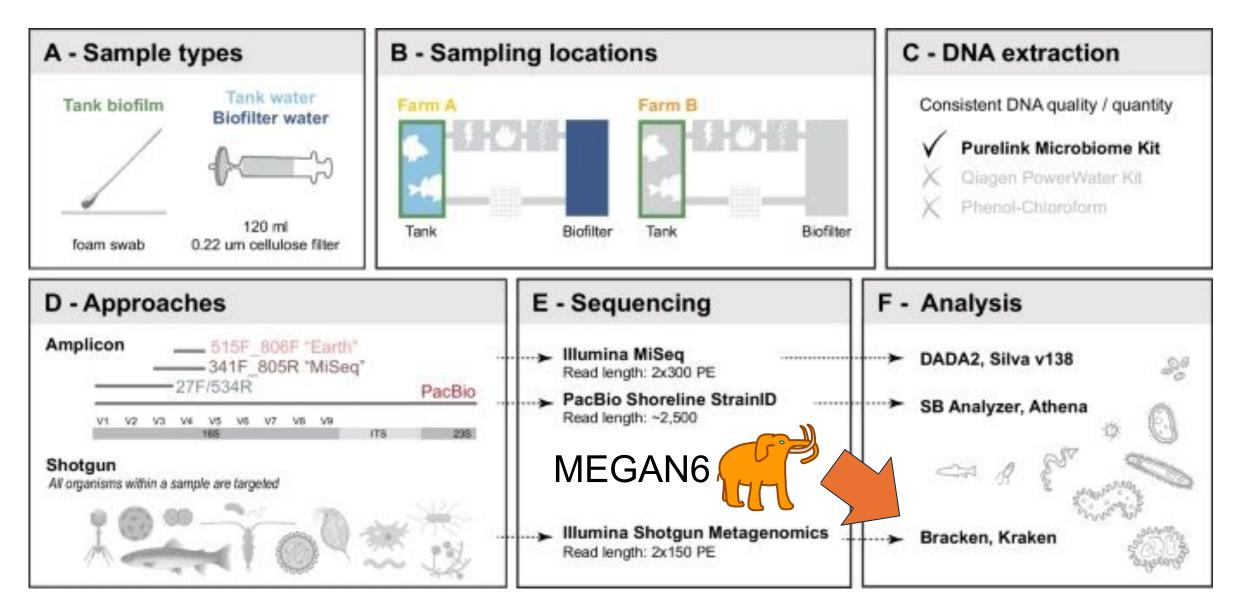


Figure Credit: Rieder et al. 2023, Environmental Microbiome

MEGAN6

 MEGAN or "MEtaGenome ANalyzer" allows for the analysis of large metagenomic datasets.

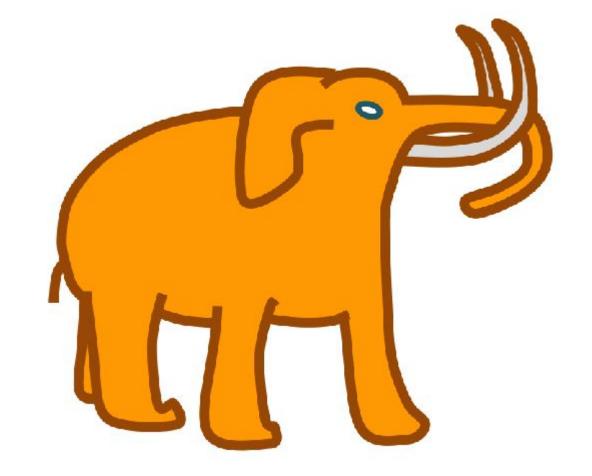


Figure Credit: MEGAN6

MEGAN History

- First version, MEGAN released in 2007 focusing on analyzing taxonomic content
- Later versions like MEGAN6 also allow for functional profiling

Research Questions that MEGAN6 can help you answer

What species are in my samples?

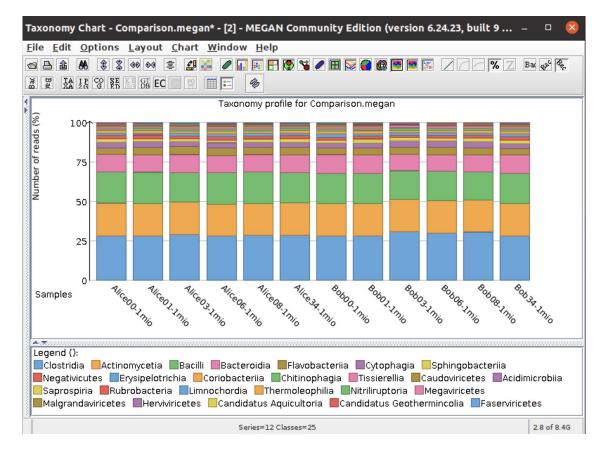


Figure Credit: MEGAN GitHub Tutorial Page

Research Questions that MEGAN6 can help you answer

What are they doing/What is the functional content of the communities?

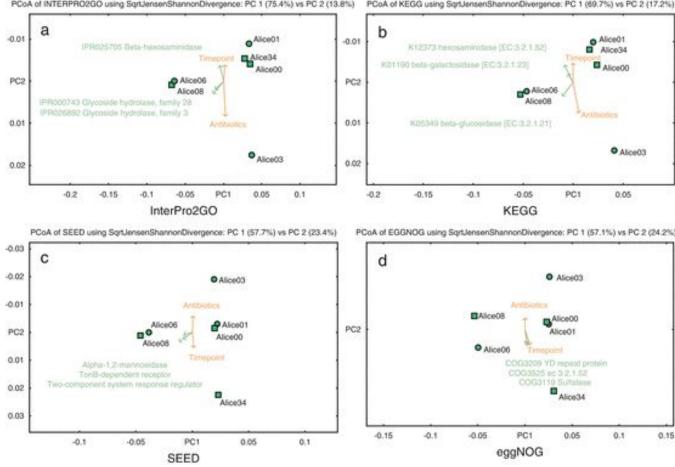


Figure Credit: Functional Metagenomics: Tools and Application (Chapter Functional Analysis in Metagenomics Using MEGAN6)

Research Questions that MEGAN6 can help you answer

 How does the taxonomic content and functional potential compare across various sample types or conditions?

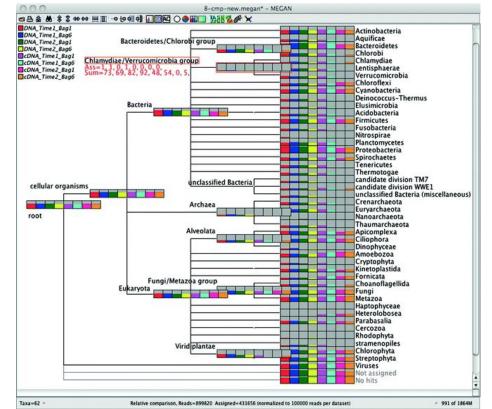


Figure Credit: MEGAN GitHub Tutorial Page

How Does MEGAN6 Work?

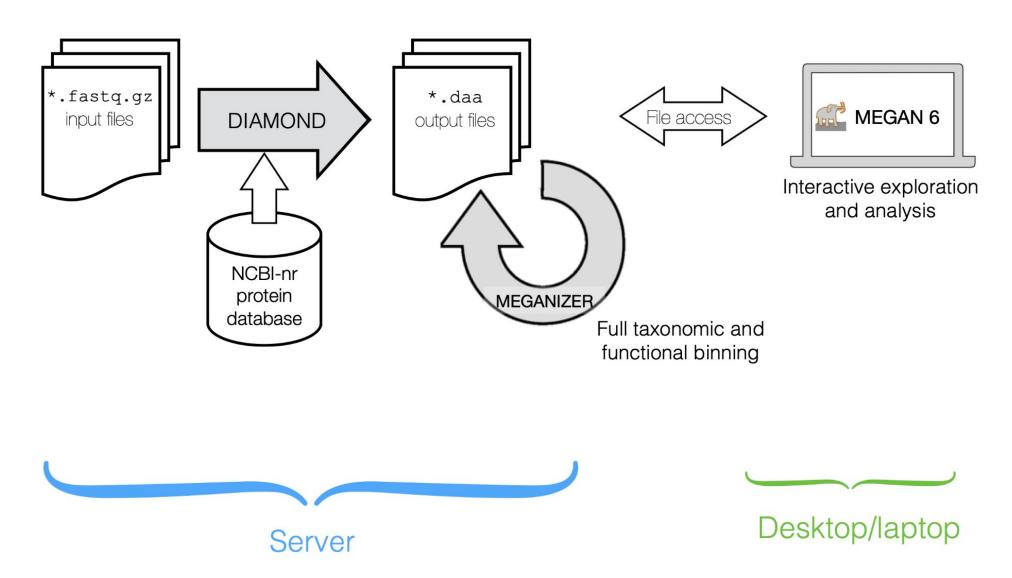


Figure Credit: MEGAN GitHub Tutorial Page

Download MEGAN/DIAMOND files needed

DIAMOND

mkdir MEGAN

cd MEGAN

wget https://github.com/bbuchfink/diamond/releases/download/v2.0.9/diamond-linux64.tar.gz

tar xzf diamond-linux64.tar.gz

wget ftp://ftp.ncbi.nlm.nih.gov/pub/taxonomy/taxdmp.zip

wget ftp://ftp.ncbi.nlm.nih.gov/pub/taxonomy/accession2taxid/prot.accession2taxid.gz

wget ftp://ftp.ncbi.nlm.nih.gov/blast/db/FASTA/nr.gz

EVERYTHING CAN BE FOUND HERE TOO

/storage/home/dkb5890/scratch/MEGAN_workshop/
This folder is 205 G → make sure you create your folder where you have enough space

DIAMOND

Build database

salloc -A open --nodes=1 --ntasks-per-node=1 --mem-per-cpu=16gb --time=12:00:00

./diamond makedb --in nr.gz -d nr_nov --taxonmap prot.accession2taxid.gz --taxonnodes nodes.dmp --taxonnames names.dmp

DIAMOND

blastx

~/MEGAN/diamond blastx -f 100 \

- -d ~/MEGAN/nr_nov.dmnd \
- -q ~/raw_metagenomes/240131_VH00707_131_AAF2G52M5_MG/D1_DC2_1_bulkDNA_S1_R1_001.fastq \
- -o ~/MEGAN/alignments/D1_DC2_1_bulkDNA_S1_R1_001.daa

EVERYTHING CAN BE FOUND HERE TOO

/storage/home/dkb5890/scratch/MEGAN_workshop/

Download MEGAN

From the MEGAN website: https://software-ab.cs.uni-tuebingen.de/download/megan6/welcome.html

The following file maps NCBI-nr accessions to taxonomic and functional classes (NCBI, GTDB, EC, eggNOG, InterPro2GO, SEED), unzip before use:

megan-map-Feb2022.db.zip

wget https://software-ab.cs.uni-tuebingen.de/download/megan6/megan-map-Feb2022.db.zip

wget https://software-ab.cs.uni-tuebingen.de/download/megan6/MEGAN_Community_unix_6_25_10.sh

unzip MEGAN_Community_unix_6_25_10.sh

Install the GUI and command line tools by running:

./MEGAN_Community_unix_6_25_10.sh

EVERYTHING CAN BE FOUND HERE TOO

/storage/home/dkb5890/scratch/MEGAN_workshop/

MEGANIZATION

/storage/home/dkb5890/megan/tools/daa2rma -i ~MEGAN/alignments/D1_DC2_1_bulkDNA_S1_R1_001.daa \

- -mdb ~MEGAN/megan-map-Feb2022.db \
- --out ~MEGAN/alignments/D1_DC2_1_bulkDNA_S1_R1_001.rma6

EVERYTHING CAN BE FOUND HERE TOO

/storage/home/dkb5890/scratch/MEGAN_workshop/

→ rma file to MEGAN!!

