

“...whoever wishes to pursue the science of medicine must first investigate the seasons of the year and what occurs in them.”

Hippocrates, 4th Century B.C.

Application of Next Generation Sequencing and Bioinformatics for Rapid and Accurate Rapid Microbial Profiling and AMR Screening

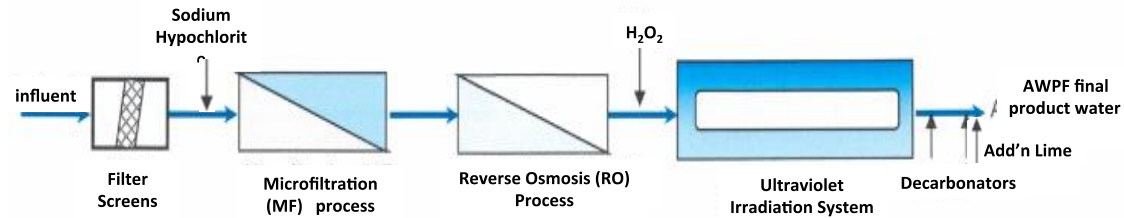
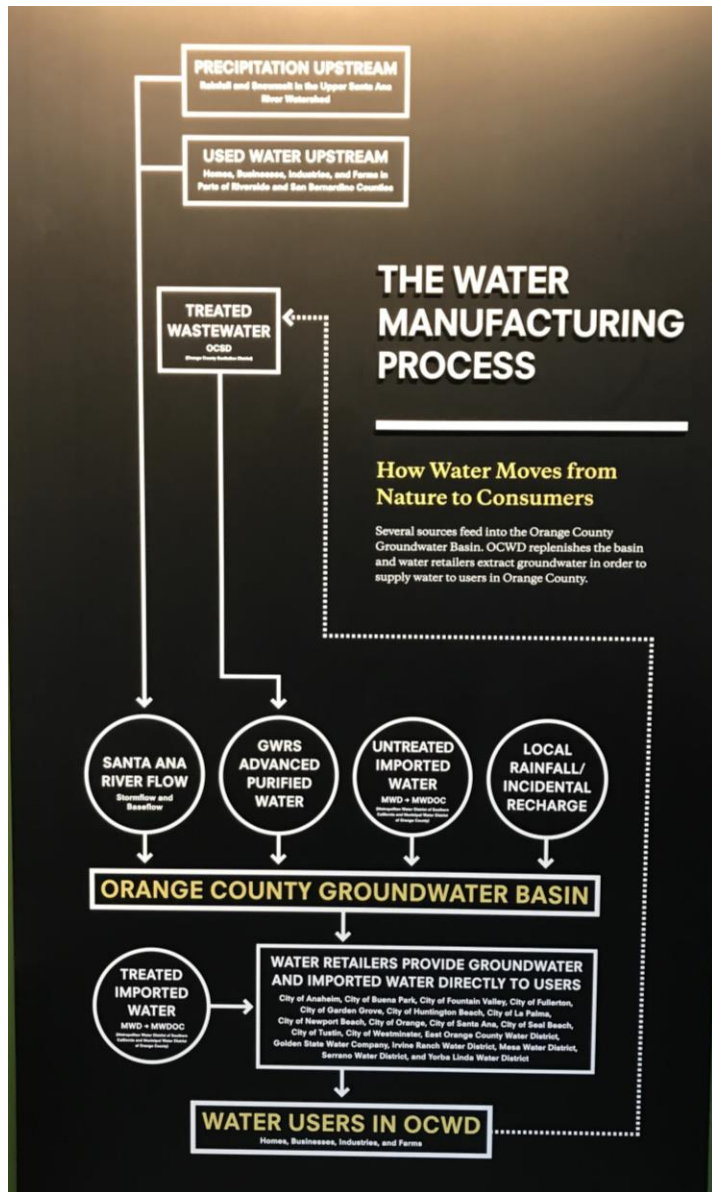
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Water Treatment Study

Orange County Water District

Metagenomics to Assess Microbial Quality Across the Water Manufacturing Process



The influent, secondary treated municipal wastewater of the AWPf treatment train is purified by a **three-step process**:

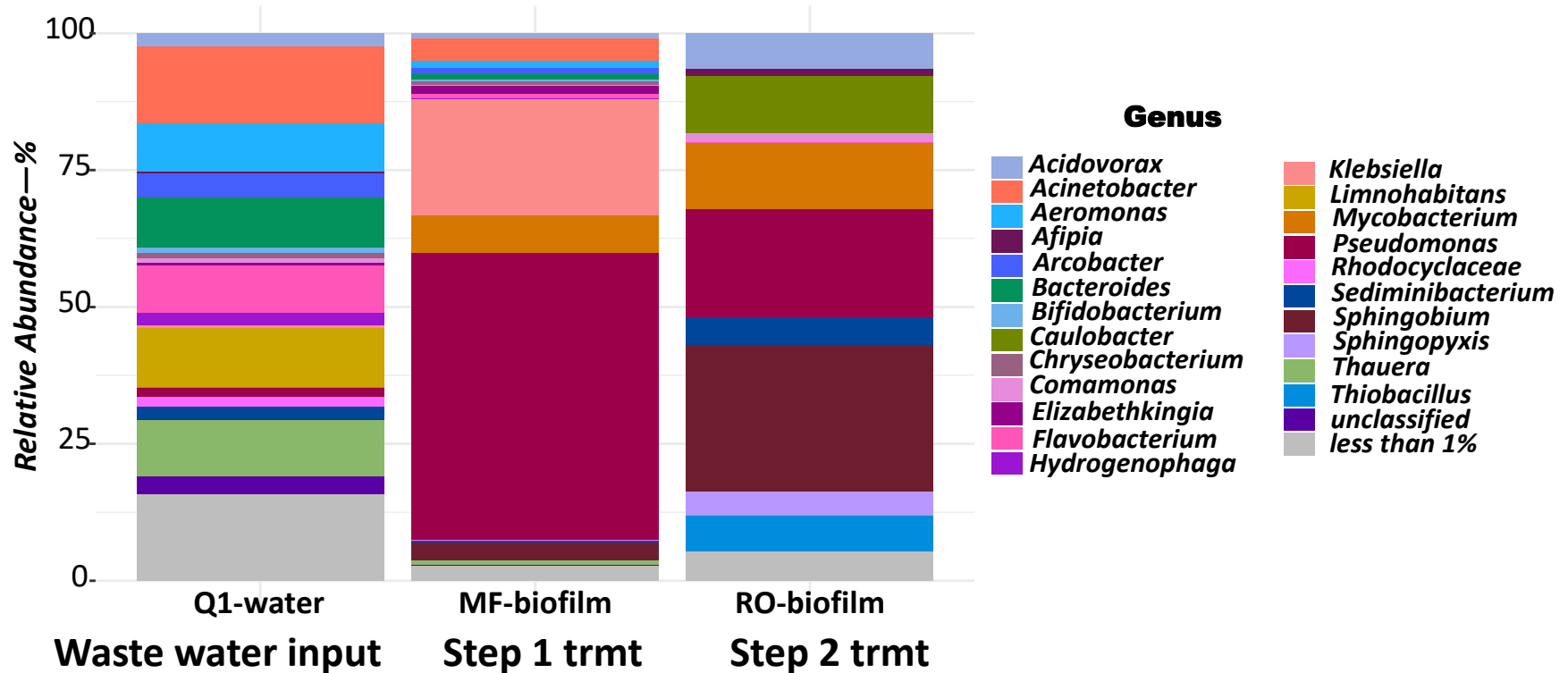
- ① microfiltration,
- ② reverse osmosis, and
- ③ ultraviolet (UV) light with hydrogen peroxide.

Initially the wastewater is screened at 4mm, Sodium hypochlorite as disinfectant is added prior to microfiltration.

Hydrogen peroxide (H_2O_2) is added before UV treatment.

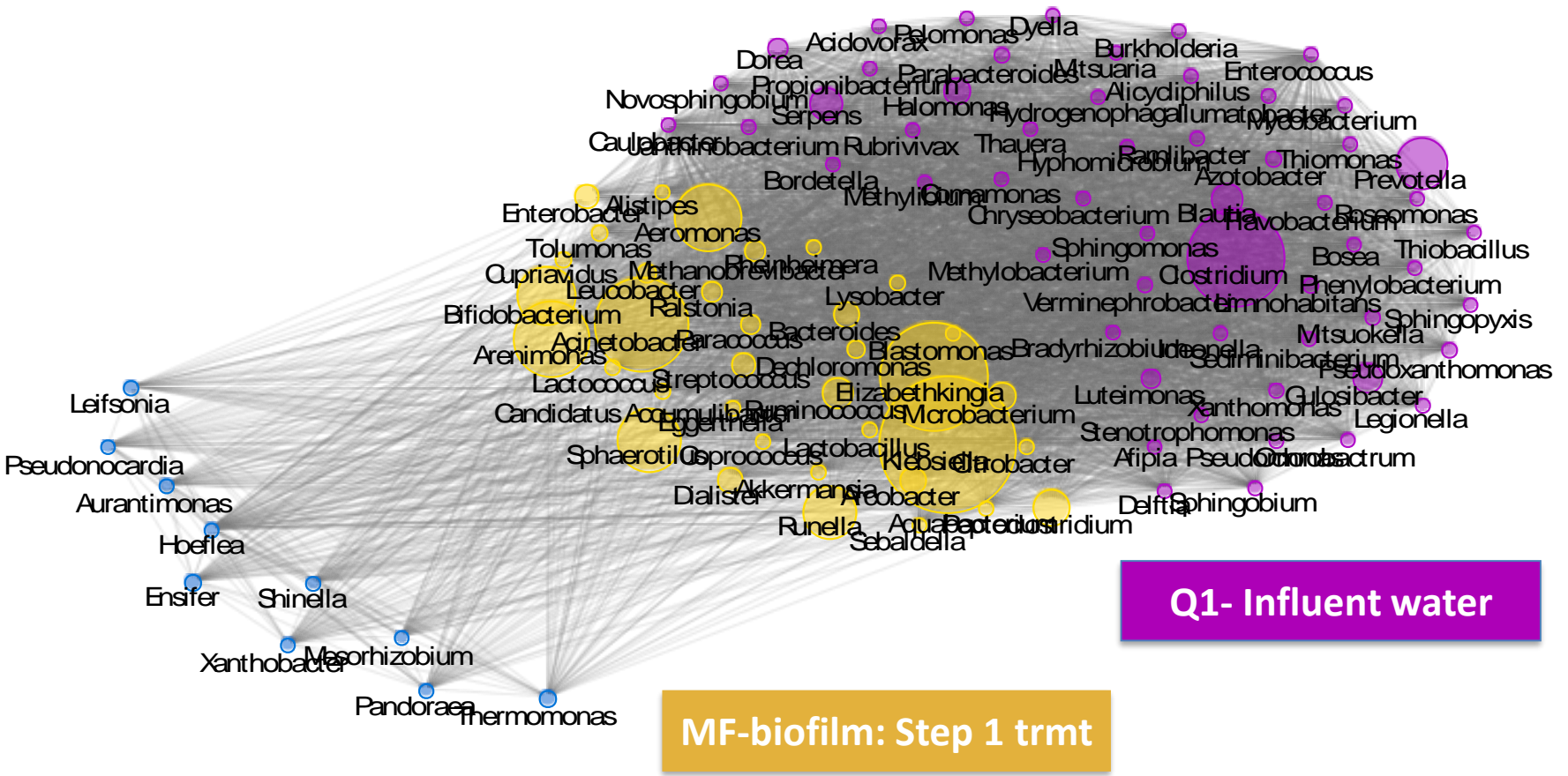
The decarbonators remove CO_2 and raise the pH; addition of lime further stabilizes the purified water.

Relative abundance of bacteria genera



Relative abundance of bacterial genera in membrane filter and reverse osmosis-biofilms and in the influent water, Q1. Genera with less than 1% relative abundance are presented in gray.

Stepwise reduction of bacteria genera



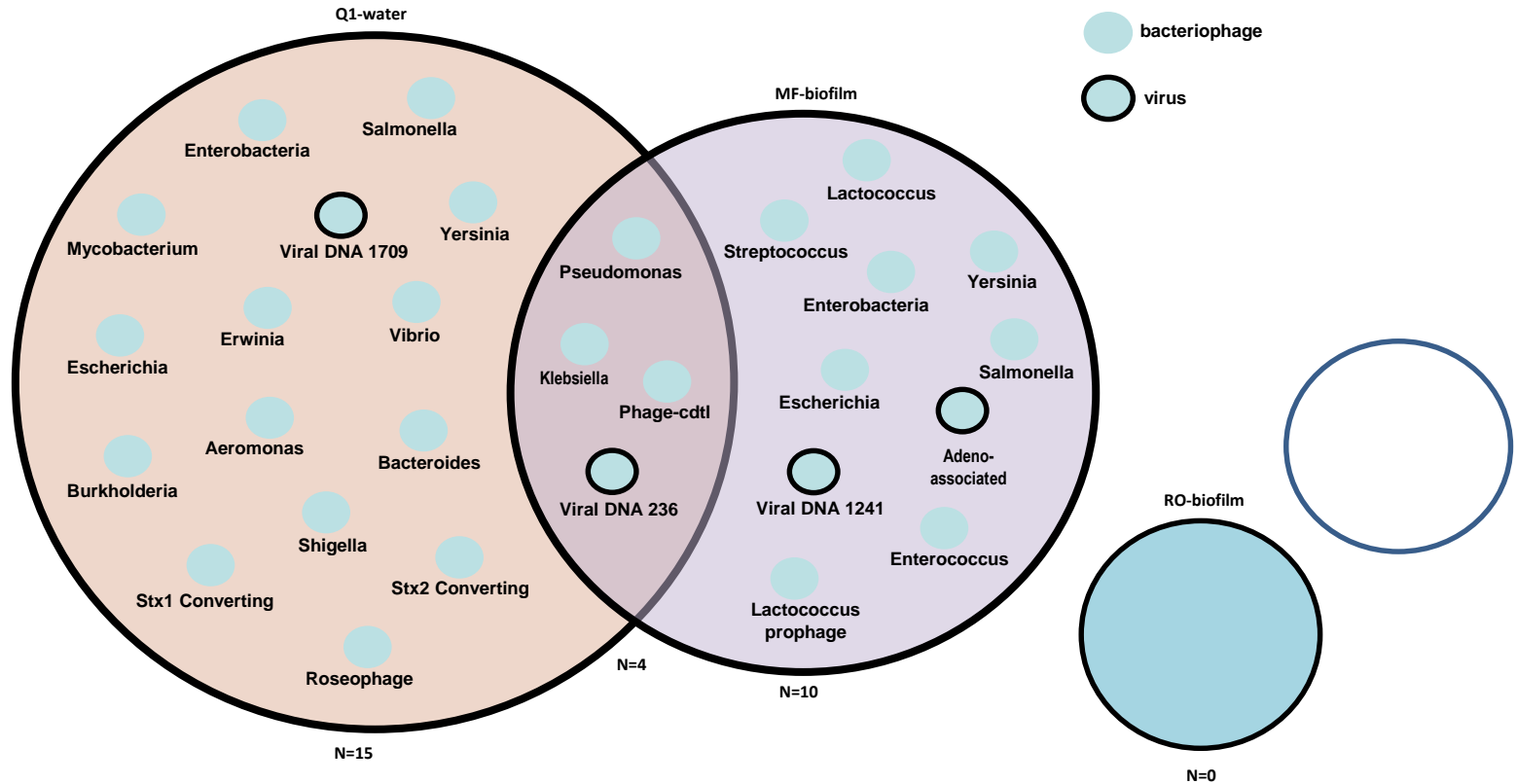
RO-biofilm: Step 2 trmt

MF-biofilm: Step 1 trmt

Q1- Influent water

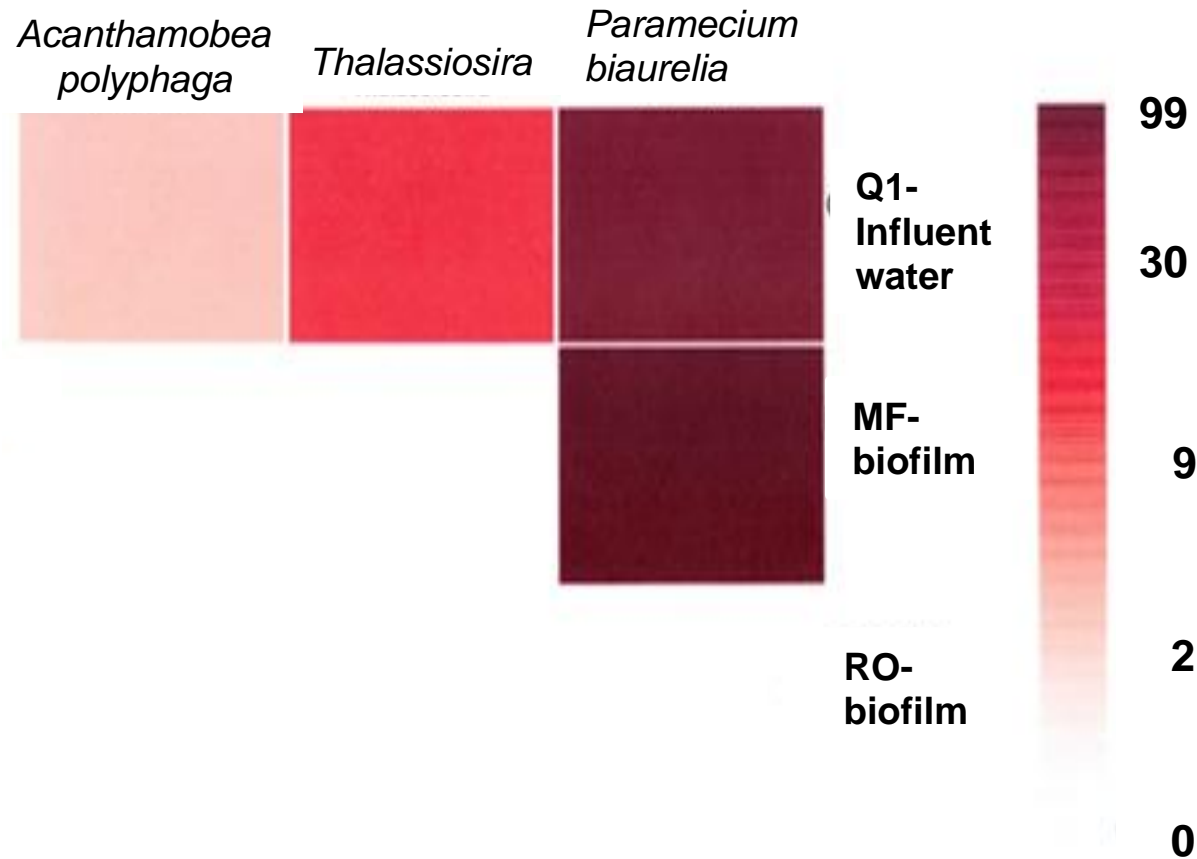
Relative Abundance—%

Comparison of virus and bacteriophage DNA sequences



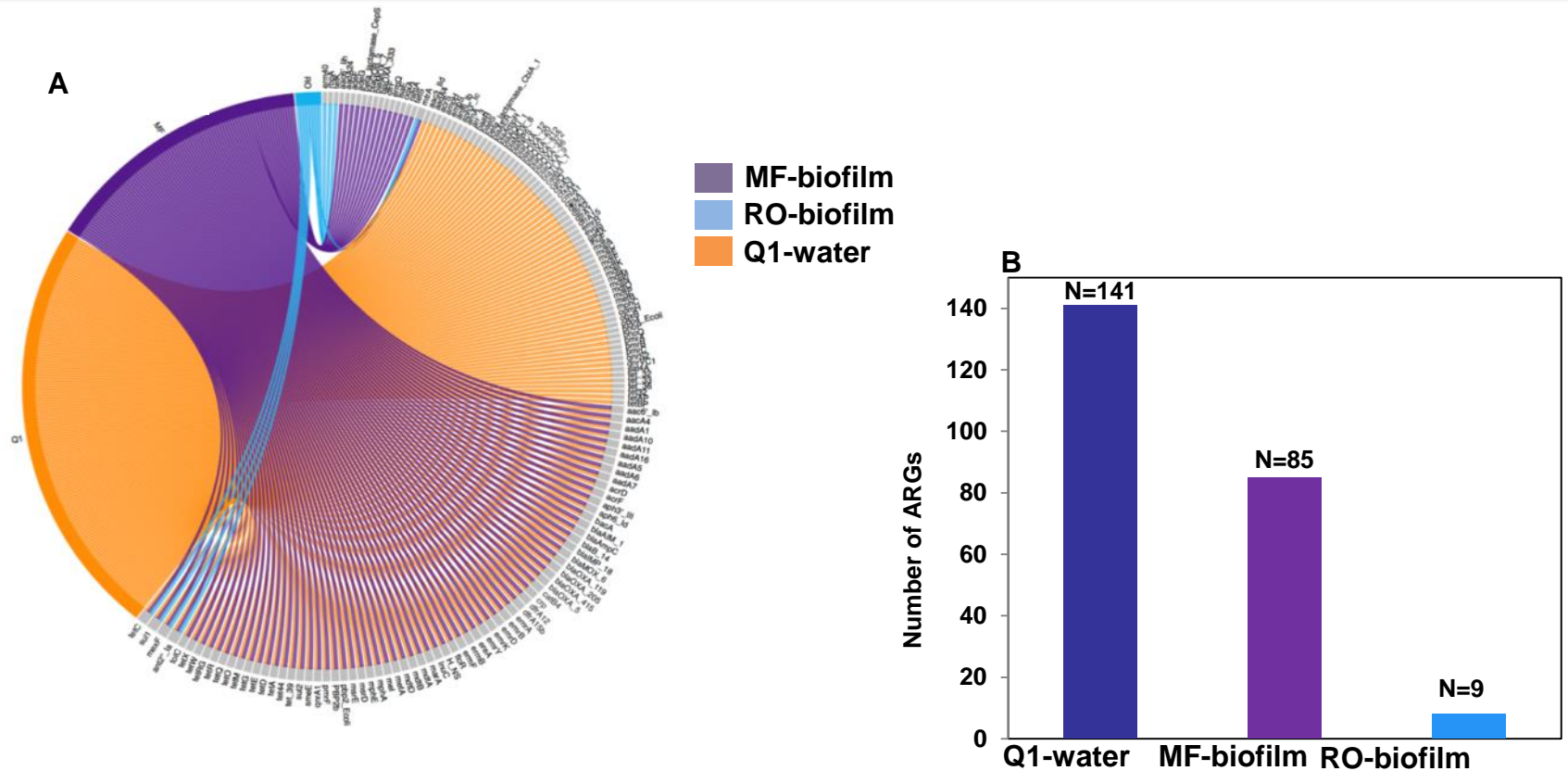
Virus and bacteriophage DNA sequences comparison demonstrate the presence of bacteriophages and virus DNA in the membrane filter (MF)-biofilm and in the influent water, Q1. Note, the absence of bacteriophages and DNA viruses in the reverse osmosis (RO)-biofilm. Presence and absences of sequences (partial or complete) related to bacteriophage and viruses in MF-biofilm were compared to the Q1 water.

Relative abundance of parasite DNA in MF-biofilm and Q1-water.



The approximate relative abundance heat map was simplified, using the CosmosID bioinformatics algorithm and curated databases. No parasite specific sequences were found in the RO-biofilms.

Distribution of antibiotic resistance genes (ARGs) in MF and RO-biofilms



(A) Distribution of antibiotic resistance genes (ARGs) in influent Q1-water, membrane filter (MF) and reverse osmosis (RO)-biofilm samples. Presence, absence, and five shared ARG sequences were identified in the MF and RO-biofilm. All ARG sequences compared represent complete and/or partial gene coverage for identification of ARGs.

(B) Stepwise reduction of antibiotic resistance coding genes (ARGs) identified in MF and RO-biofilm and in influent Q1.