Microbial Diversity and the Quality of Roof-harvested Rainwater

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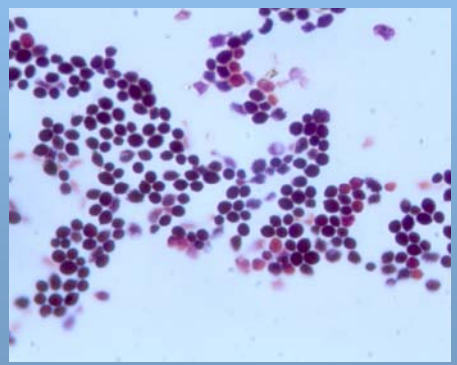

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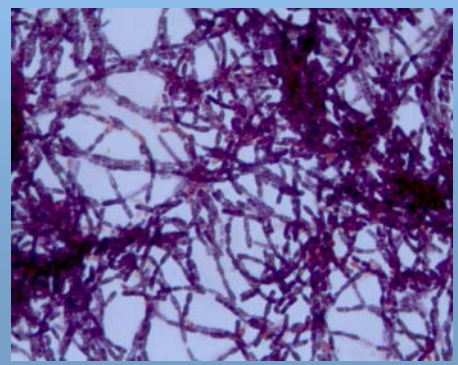
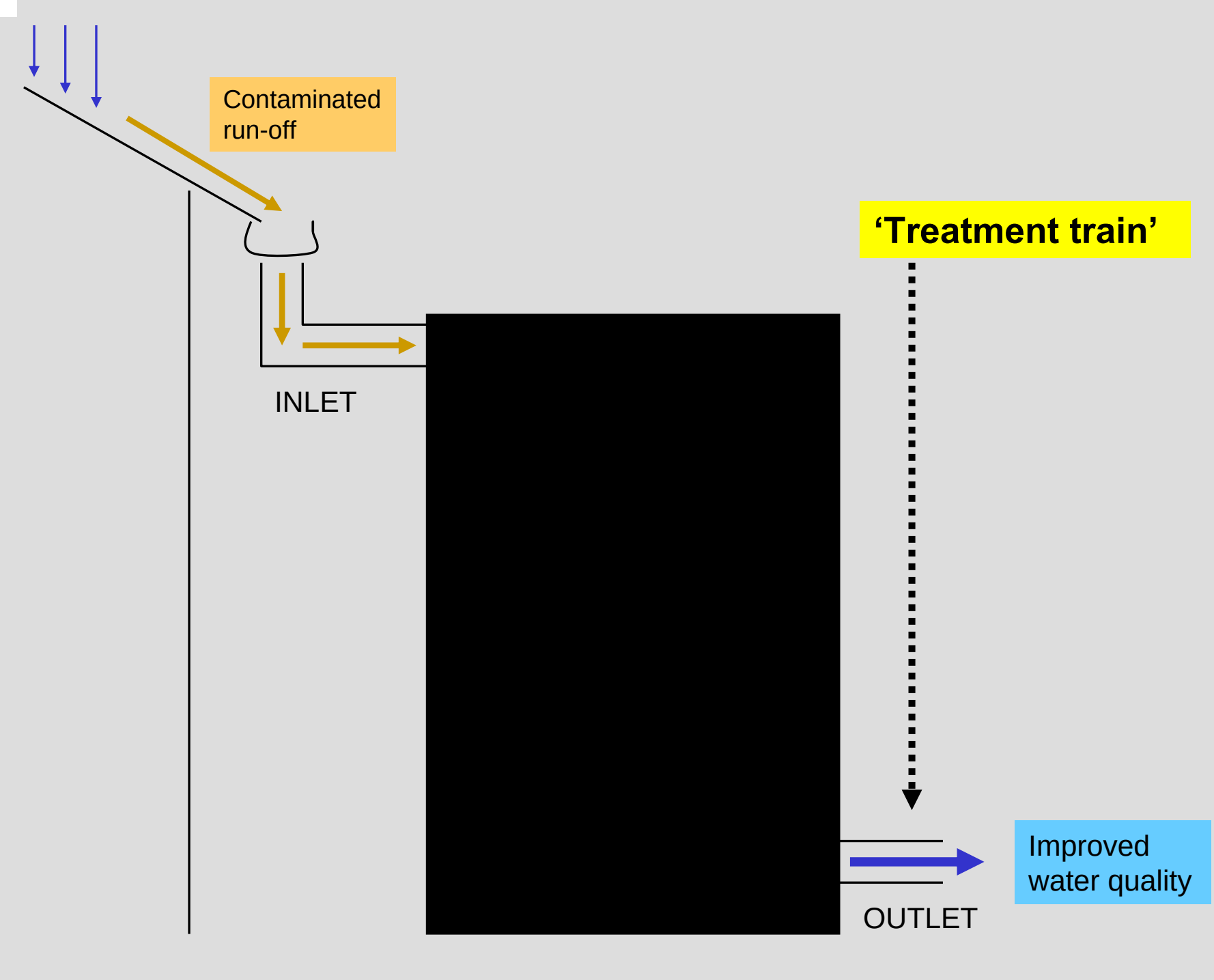


Are non-pathogenic bacteria significant?

1. The *'treatment train'* effect
   - Apparent improvements to the microbial and chemical quality of water with passage through the collection system.

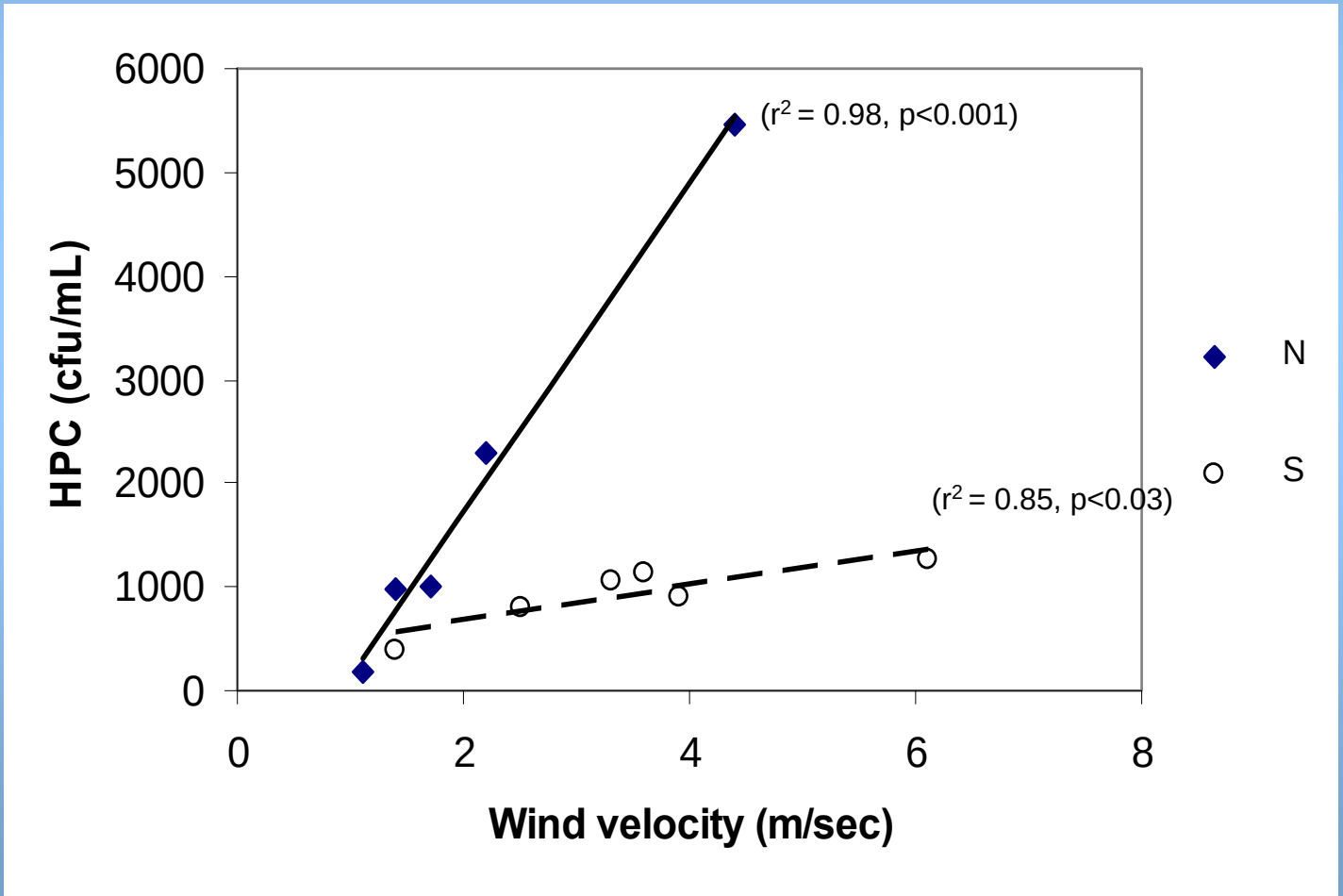

Contaminated run-off
‘Treatment train’
INLET
OUTLET
Improved water quality

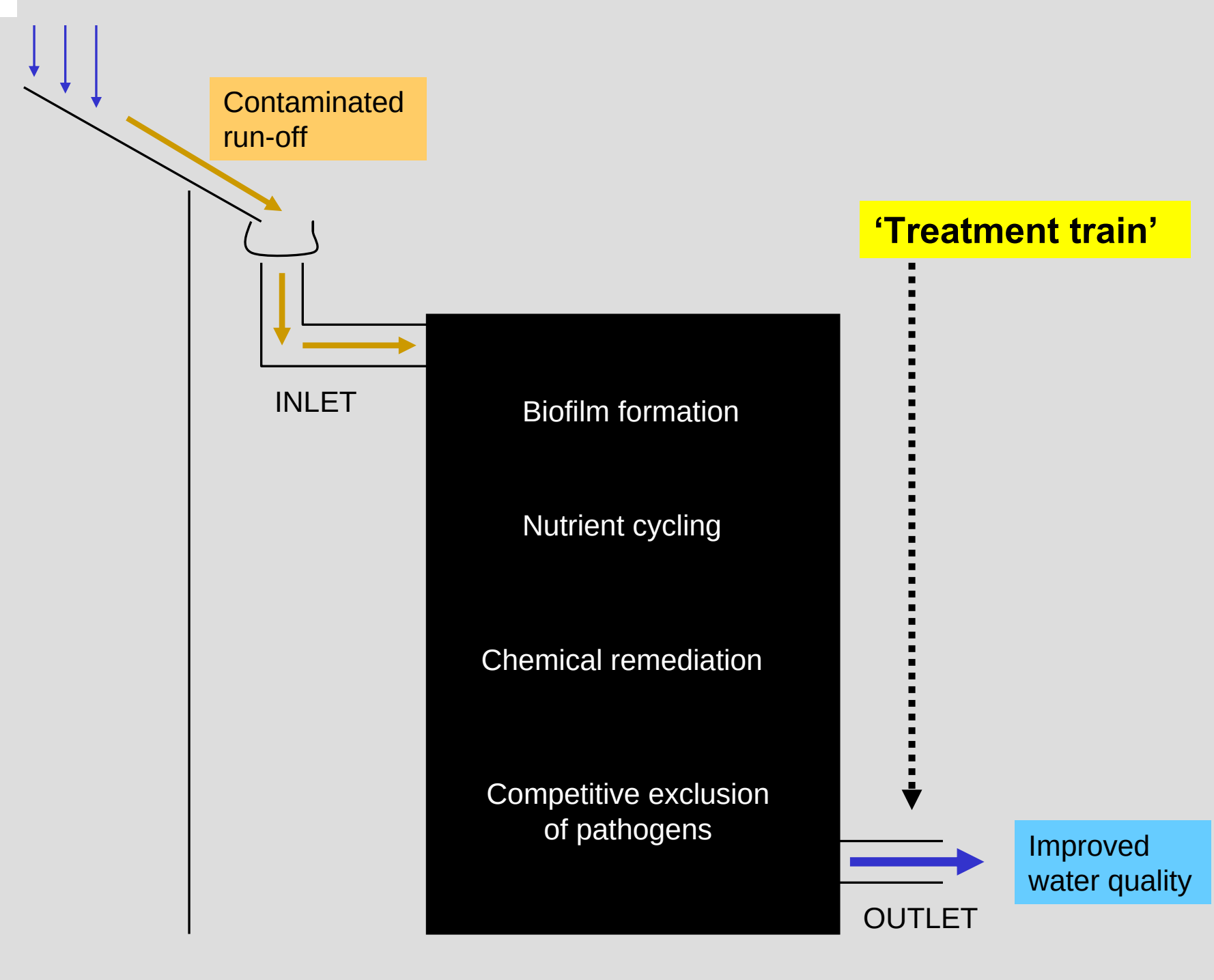
Are non-pathogenic bacteria significant?

1. The *'treatment train'* effect
   - Apparent improvements to the microbial and chemical quality of water with passage through the collection system.

2. Environmental bacteria comprise majority of organisms present in rainwater tanks.
   - HPC **>>>** coliform counts - (orders of magnitude)
   - Bacterial composition influenced by wind patterns

Wind direction/velocity & HPC




Contaminated run-off
‘Treatment train’
INLET
Biofilm formation
Nutrient cycling
Chemical remediation
Competitive exclusion of pathogens
OUTLET
Improved water quality

Overview

1. Assessment of microbial diversity in 2 rainwater tanks.
   - Impact of hot water systems
   - Assessment of coliforms

2. Biofilm pilot studies
   - Biofilm development and the treatment train
   - In-vitro examination of Pb^{2+} uptake

Microbial Diversity of 2 Rainwater Tanks

- Urban tank
  - Located in Newcastle (160kM Nth of Sydney)
  - Duel 2.2kL galvanized iron tanks
  - Mains water top-up system
  - Hot water service set at 60°C
- Rural tank
  - Located at Gulgong (250kM inland of Newcastle)
  - 10kL ‘aquaplate’ tank
  - Hot water service set at 50°C.

Table 1: *Plate count and diversity summary*

Tank	**Urban**		**Rural**	
Sample type	Cold	Hot	Cold	Hot
Total no. of species identified	64	20	49	39
mean no. species/sample	23 ± 3.4	8 ± 1.2	16 ± 2.3	14 ± 4.3
Mean plate count (cfu/mL)	366 ± 153	17 ± 2.6	2263 ± 1070	94 ± 31

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Fig. 2: *% composition of gram negative and gram positive bacteria in samples from rainwater tanks.*

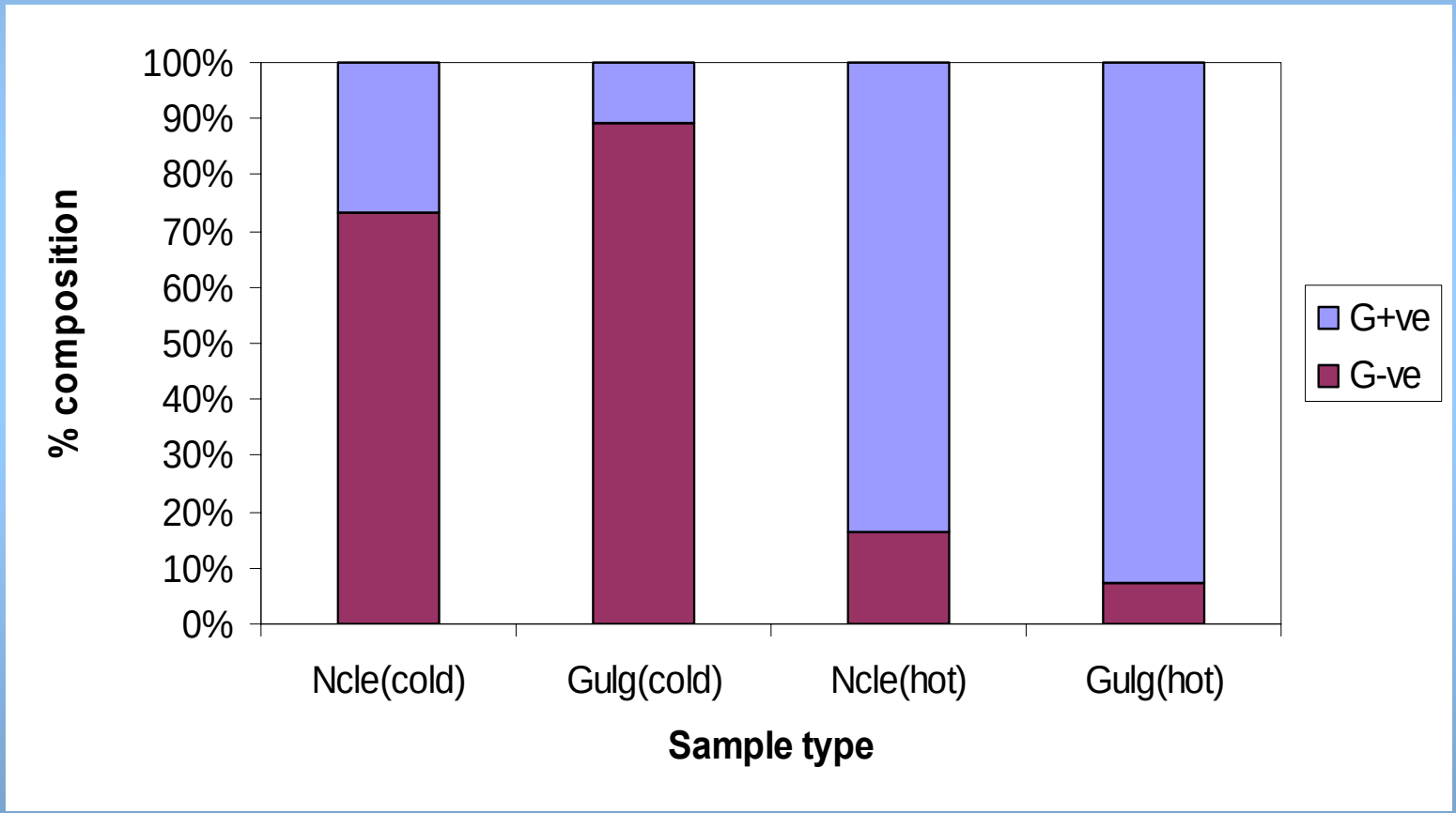
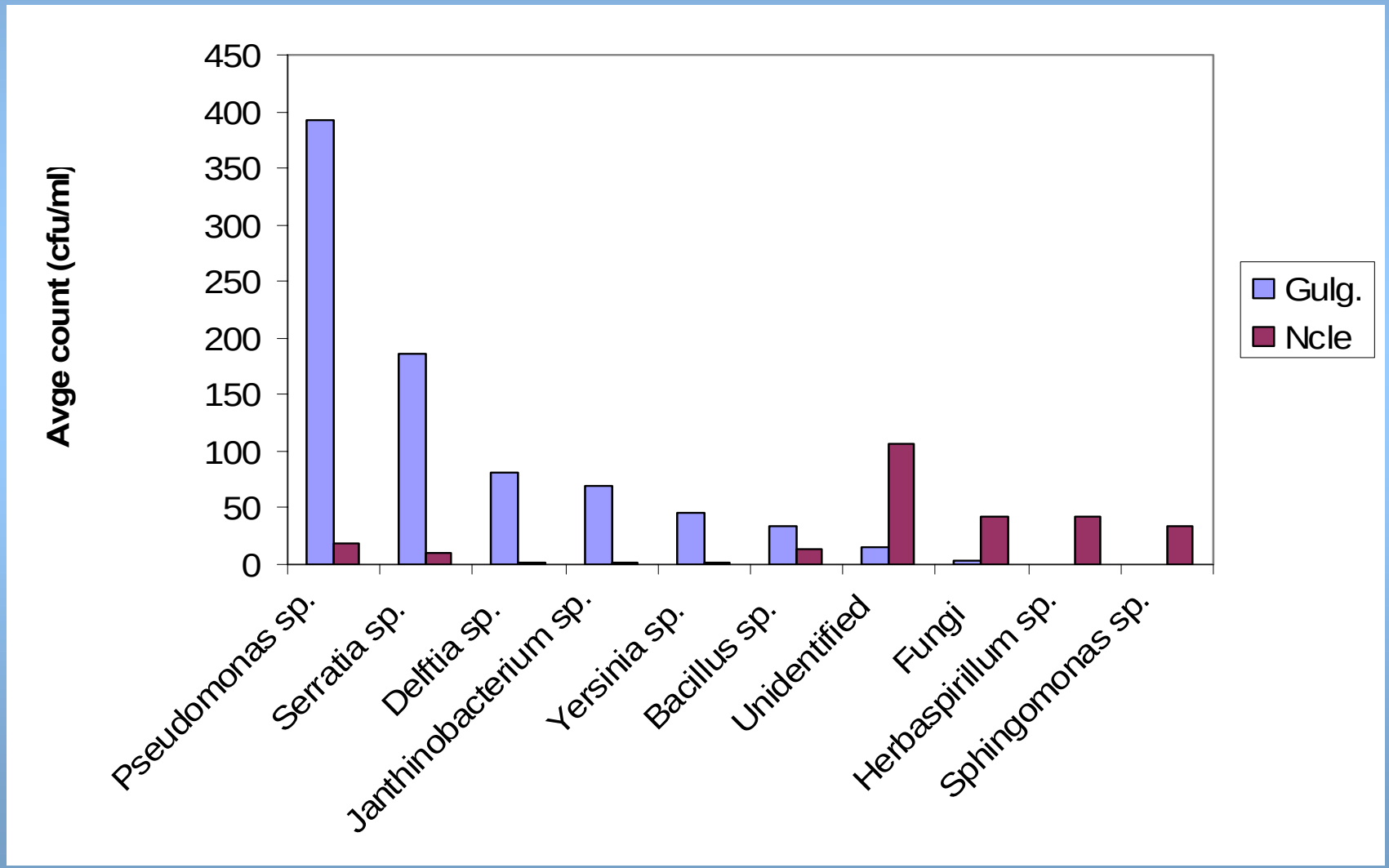


Fig. 1: *Profiles of the Newcastle and Gulgong tanks based on average counts of most abundant genera present.*



Faecal indicator presence and abundance relative to total plate count

Sample type	COLD		HOT	
	Presence (% of samples)	Avge % of plate count	Presence (% of samples)	Avge % of plate count
E.coli	50	0.06 ± 0.03	0	0
Enterococci	23	0.05 ± 0.03	28	0.62 ± 0.41
Faecal col.	77	0.52 ± 0.27	28	0.62 ± 0.41
Total col.	100	3.77 ± 2.08	43	3.36 ± 2.21

Biofilms and the treatment train

Location: Newcastle on east coast of Australia (approx. 160kM Nth of Sydney)

Tank	Biofilm development	Comment
A *(G.I.)*	Advanced	heavy slime layer with extensive clumping
B *(Plastic)*	Moderate	uniform slime layer around interior

Table 3: *Variation in bacterial counts between the tank water column and tap outlet of tanks A and B.*

Tank	Sample type	Bacterial count	
		E.coli (cfu/100ml)	HPC (cfu/mL)
A	Tank water column	48	610
	Tap outlet	29	390
	Improvement	**34%**	**36%**
B	Tank water column	70	200
	Tap outlet	60	160
	Improvement	**14%**	**20%**

Table 3: *Variation in bacterial counts between the tank water column and tap outlet of 2 tanks.*

Tank	Sample type	Bacterial count	
		E.coli (cfu/100ml)	**HPC** (cfu/mL)
A	Tank water column	48	610
	Tap outlet	29	390
	Improvement	**34%**	**36%**
B	Tank water column	70	200
	Tap outlet	60	160
	Improvement	**14%**	**20%**

Pb^{2+} uptake by in-vitro cultivated biofilm

- *E.coli* biofilms cultivated in wells of perspex tissue culture plates for 72 hours.
- Incubation broth replaced with sterile milli-Q water spiked with Pb^{2+} (100ppb).
- Pb^{2+} determined by ICP-MS at time = 0, 5, 10 and 20 hours.

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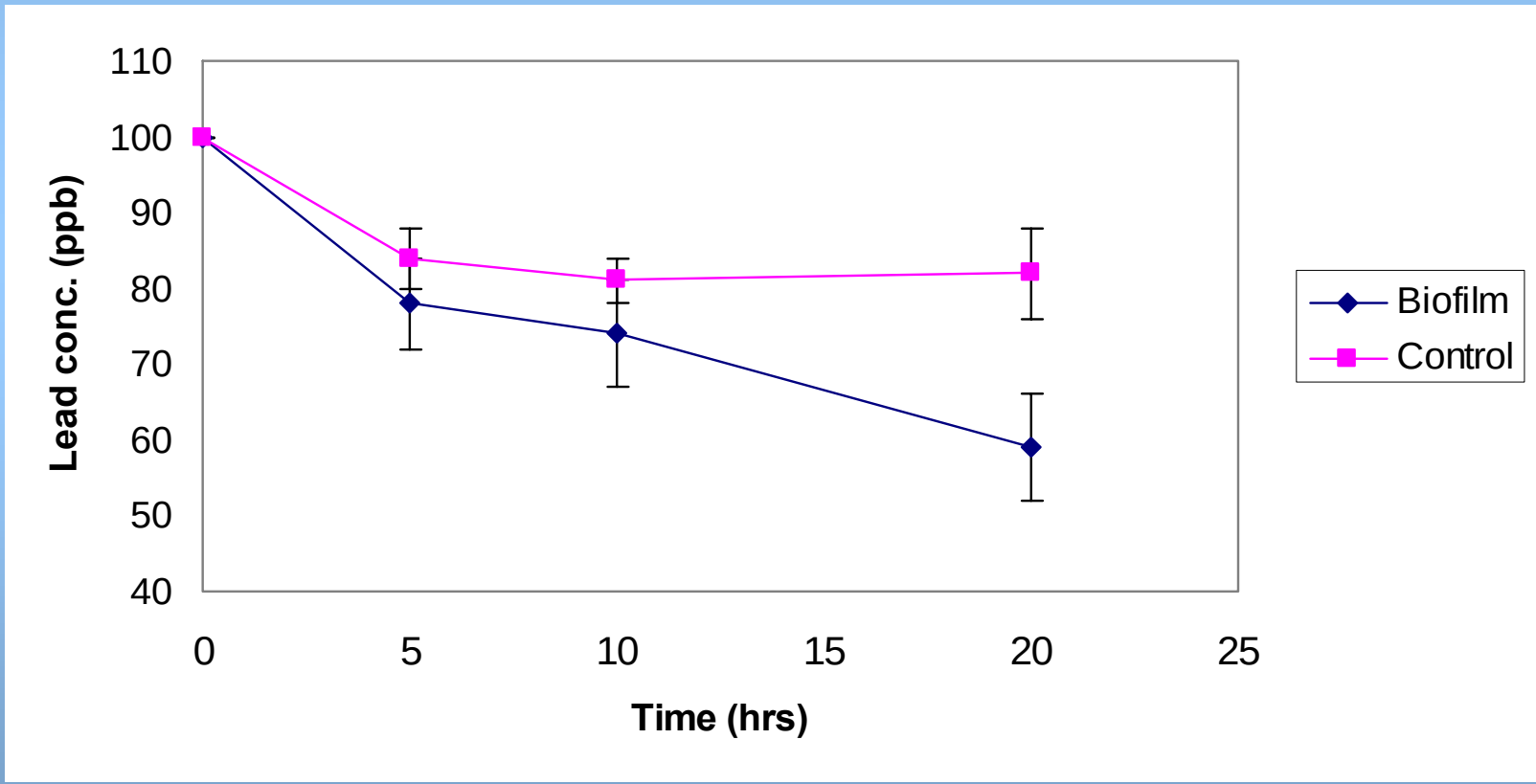
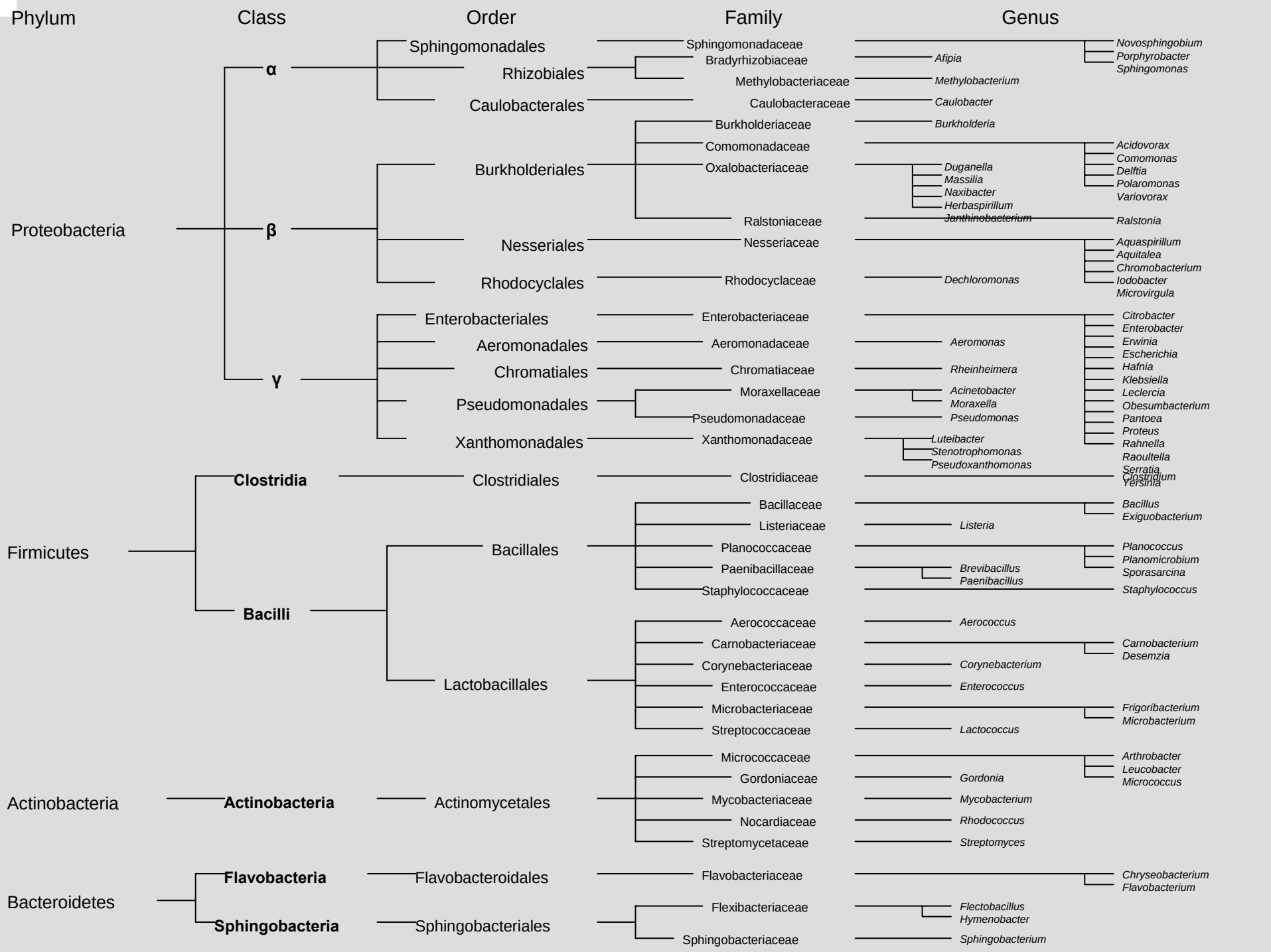


Conclusions

- Rainwater tanks harbour a wide diversity of microbial species.
- Species composition may vary considerably between tanks at different locations.
- Species composition varies temporally at any given site.
- Minimal evidence of faecal contamination.
- Hot water systems deliver water of quality suitable for bathing.
- Biofilms have capacity to chemically remediate water.


Phylum
Class
Order
Family
Genus
Proteobacteria
α
β
γ
Sphingomonadales
Rhizobiales
Caulobacterales
Burkholderiales
Nesseriales
Rhodocyclales
Enterobacteriales
Aeromonadales
Chromatiales
Pseudomonadales
Xanthomonadales
Sphingomonadaceae
Bradyrhizobiaceae
Methylobacteriaceae
Caulobacteraceae
Burkholderiaceae
Comomonadaceae
Oxalobacteriaceae
Ralstoniaceae
Nesseriaceae
Rhodocyclaceae
Enterobacteriaceae
Aeromonadaceae
Chromatiaceae
Moraxellaceae
Pseudomonadaceae
Xanthomonadaceae
Novosphingobium
Porphyrobacter
Sphingomonas
Afipia
Methylobacterium
Caulobacter
Burkholderia
Acidovorax
Comomonas
Delftia
Polaromonas
Variovorax
Duganella
Massilia
Naxibacter
Herbaspirillum
Janthinobacterium
Ralstonia
Aquaspirillum
Aquitalea
Chromobacterium
Iodobacter
Microvirgula
Dechloromonas
Citrobacter
Enterobacter
Erwinia
Escherichia
Hafnia
Klebsiella
Leclercia
Obesumbacterium
Pantoea
Proteus
Rahnella
Raoultella
Serratia
Yersinia
Aeromonas
Rheinheimera
Acinetobacter
Moraxella
Pseudomonas
Luteibacter
Stenotrophomonas
Pseudoxanthomonas
Firmicutes
Clostridia
Bacilli
Clostridiales
Bacillales
Lactobacillales
Clostridiaceae
Bacillaceae
Listeriaceae
Planococcaceae
Paenibacillaceae
Staphylococcaceae
Aerococcaceae
Carnobacteriaceae
Corynebacteriaceae
Enterococcaceae
Microbacteriaceae
Streptococcaceae
Clostridium
Bacillus
Exiguobacterium
Listeria
Planococcus
Planomicrobium
Sporasarcina
Brevibacillus
Paenibacillus
Staphylococcus
Aerococcus
Carnobacterium
Desemzia
Corynebacterium
Enterococcus
Frigoribacterium
Microbacterium
Lactococcus
Actinobacteria
Actinobacteria
Actinomycetales
Micrococcaceae
Gordoniaceae
Mycobacteriaceae
Nocardiaceae
Streptomycetaceae
Arthrobacter
Leucobacter
Micrococcus
Gordonia
Mycobacterium
Rhodococcus
Streptomyces
Bacteroidetes
Flavobacteria
Sphingobacteria
Flavobacteroidales
Sphingobacteriales
Flavobacteriaceae
Flexibacteriaceae
Sphingobacteriaceae
Chryseobacterium
Flavobacterium
Flectobacillus
Hymenobacter
Sphingobacterium