Naegleria in drinking water and pipe wall biofilms in Western Australia

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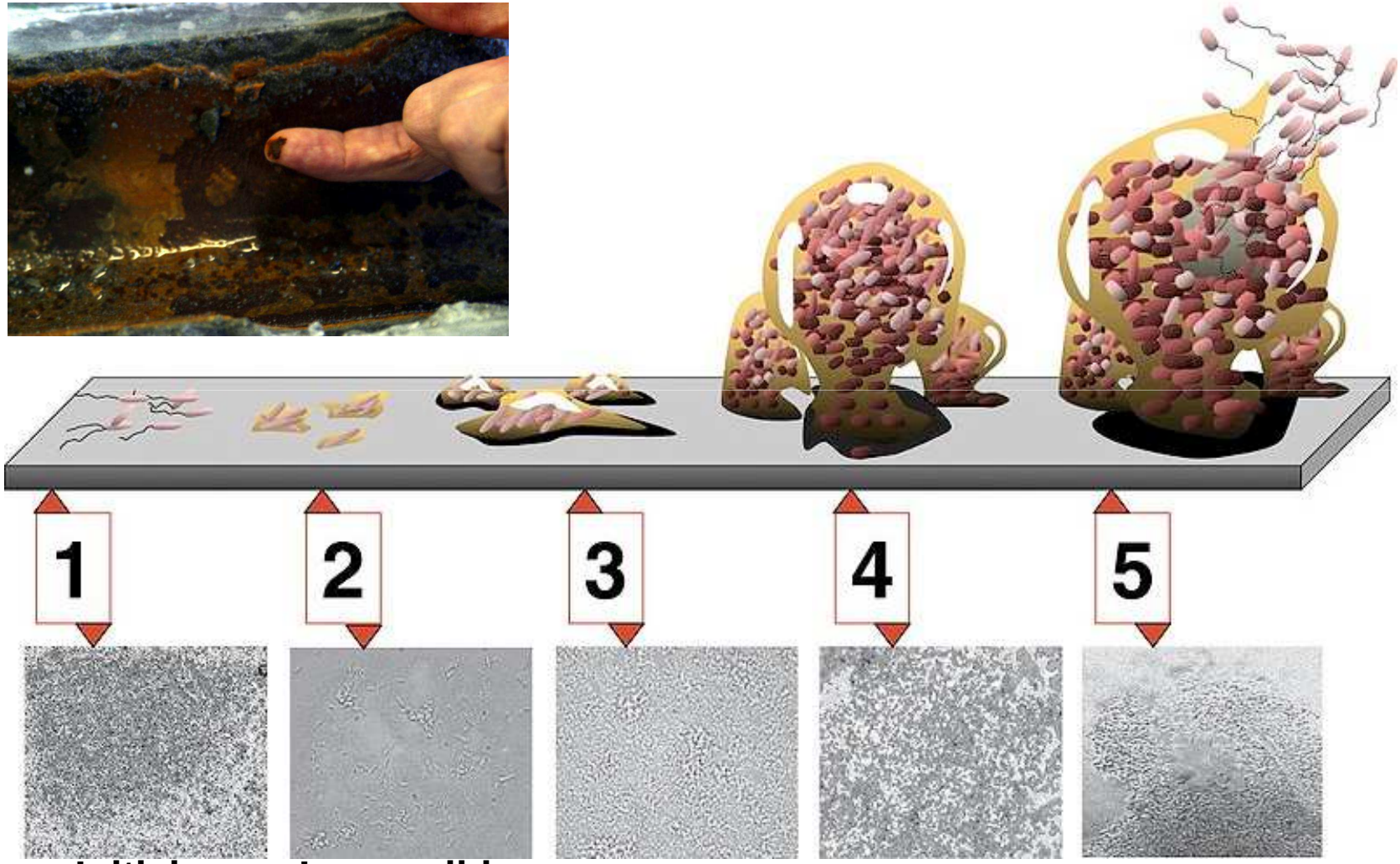
Golden Pipeline

Biofilm growth in water distribution pipes



Pipe wall biofilms and its impact

- Trap particles, harbour pathogens
- Is a sink for metals (commonly Fe & Mn) – contribute towards discoloured water incidences
- Adsorb and consume DOC, NH_4^{+-} promotes the growth of nuisance organisms
- Form diffusion barriers to disinfectants
- Promote anaerobic microenvironments – Odor
- Release film containing organisms and bound metals.

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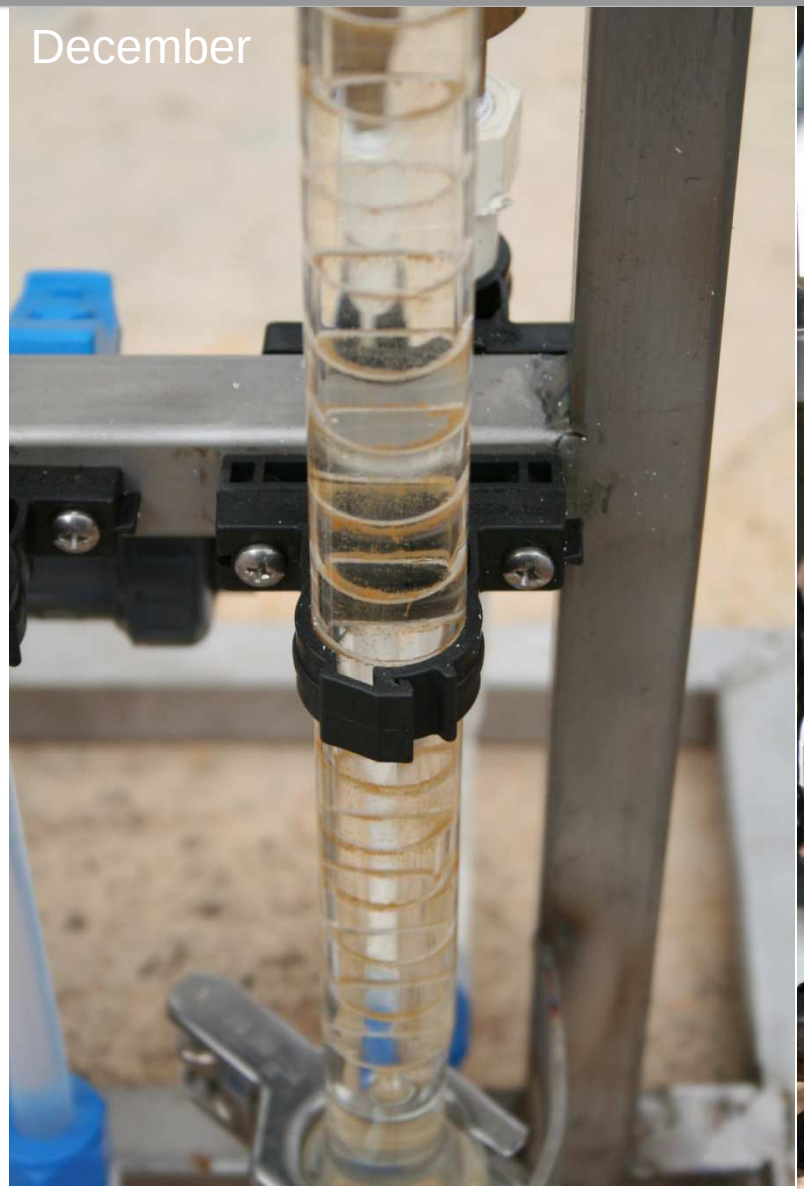
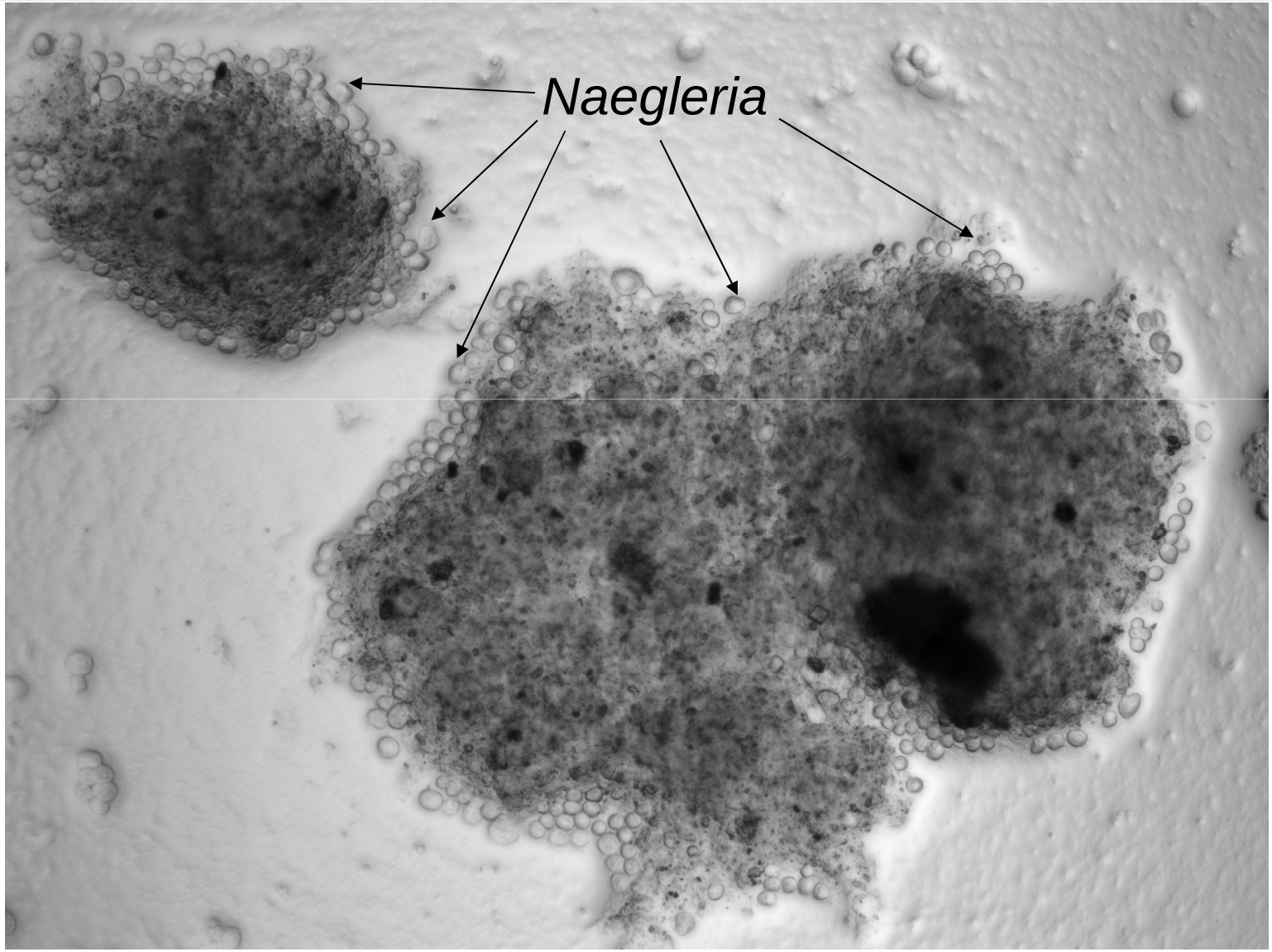
Rural Field sites

Field sites

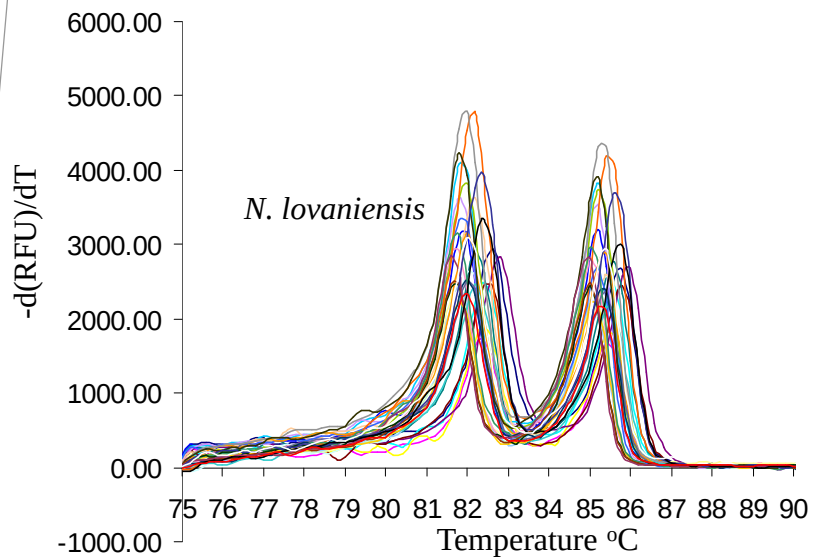
Influent Biomonitors at Field site

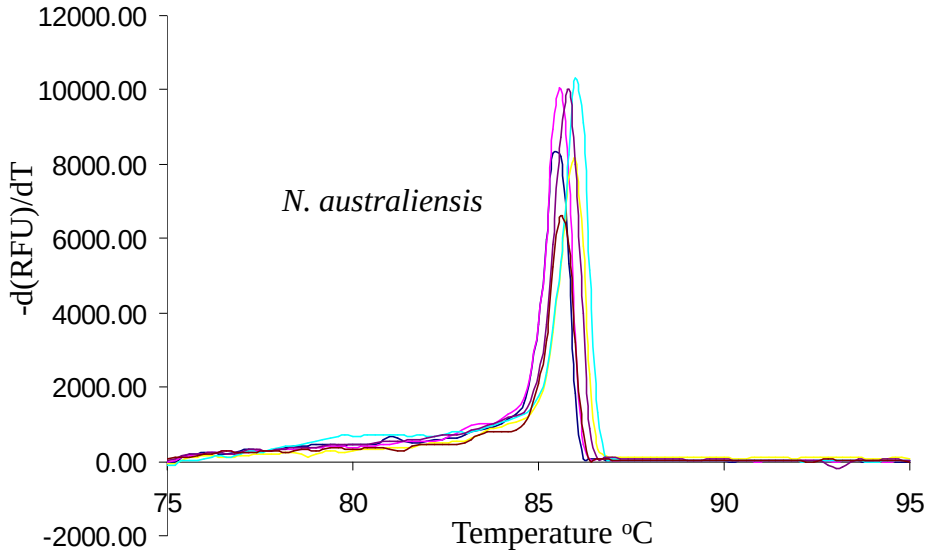


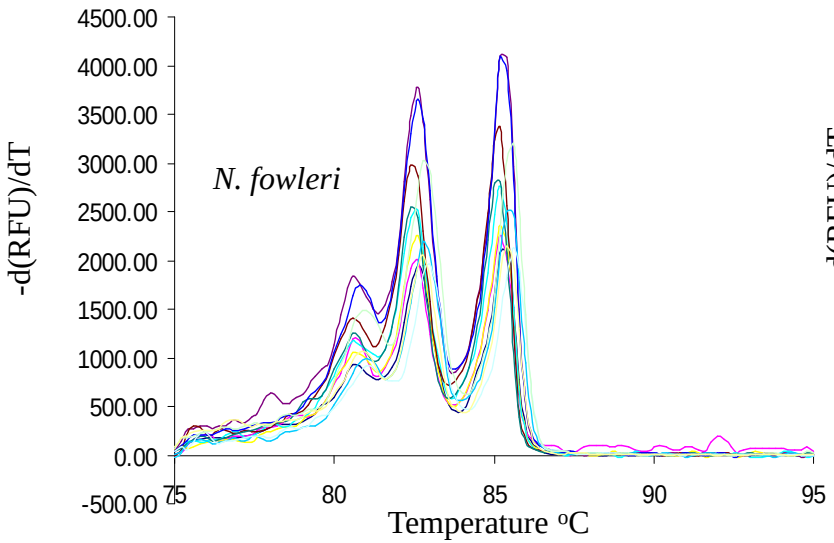
Naegleria isolated from biofilm

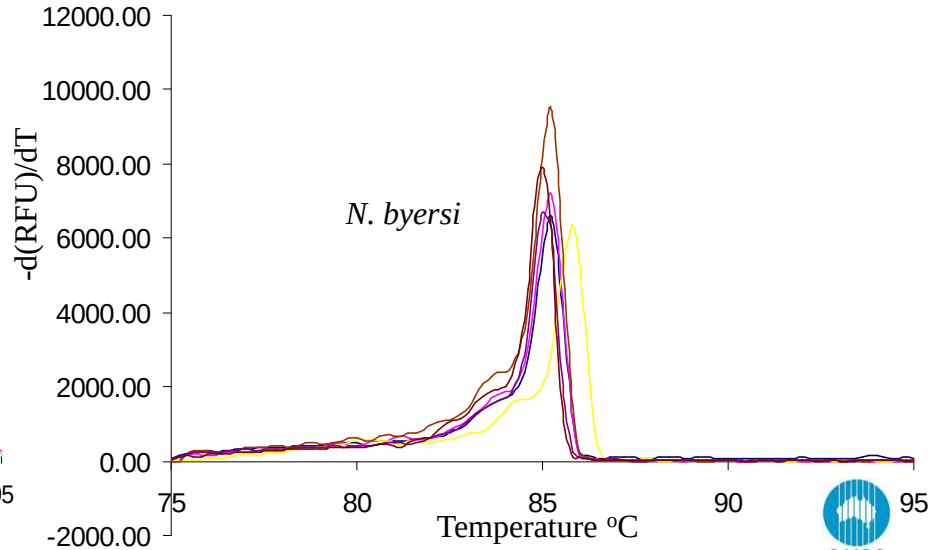


Species Specific Identification- Consensus Primers

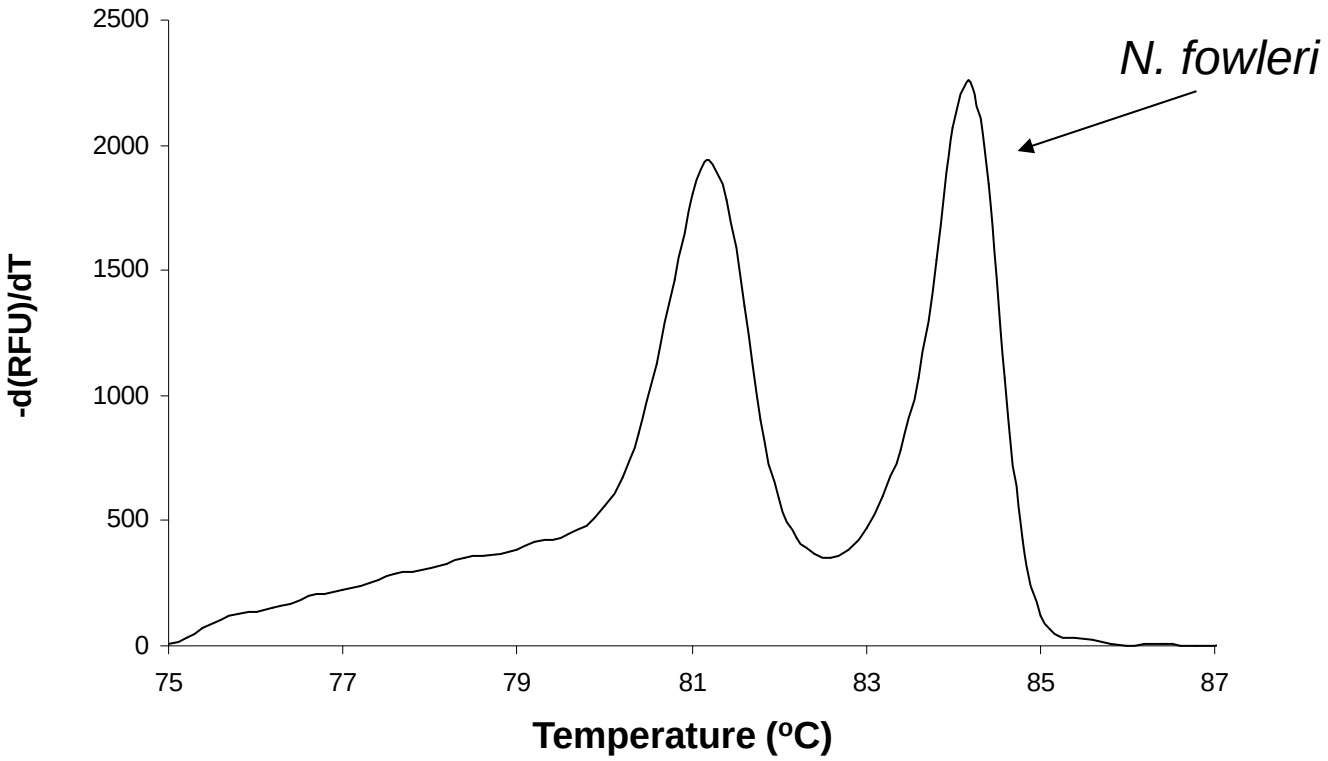








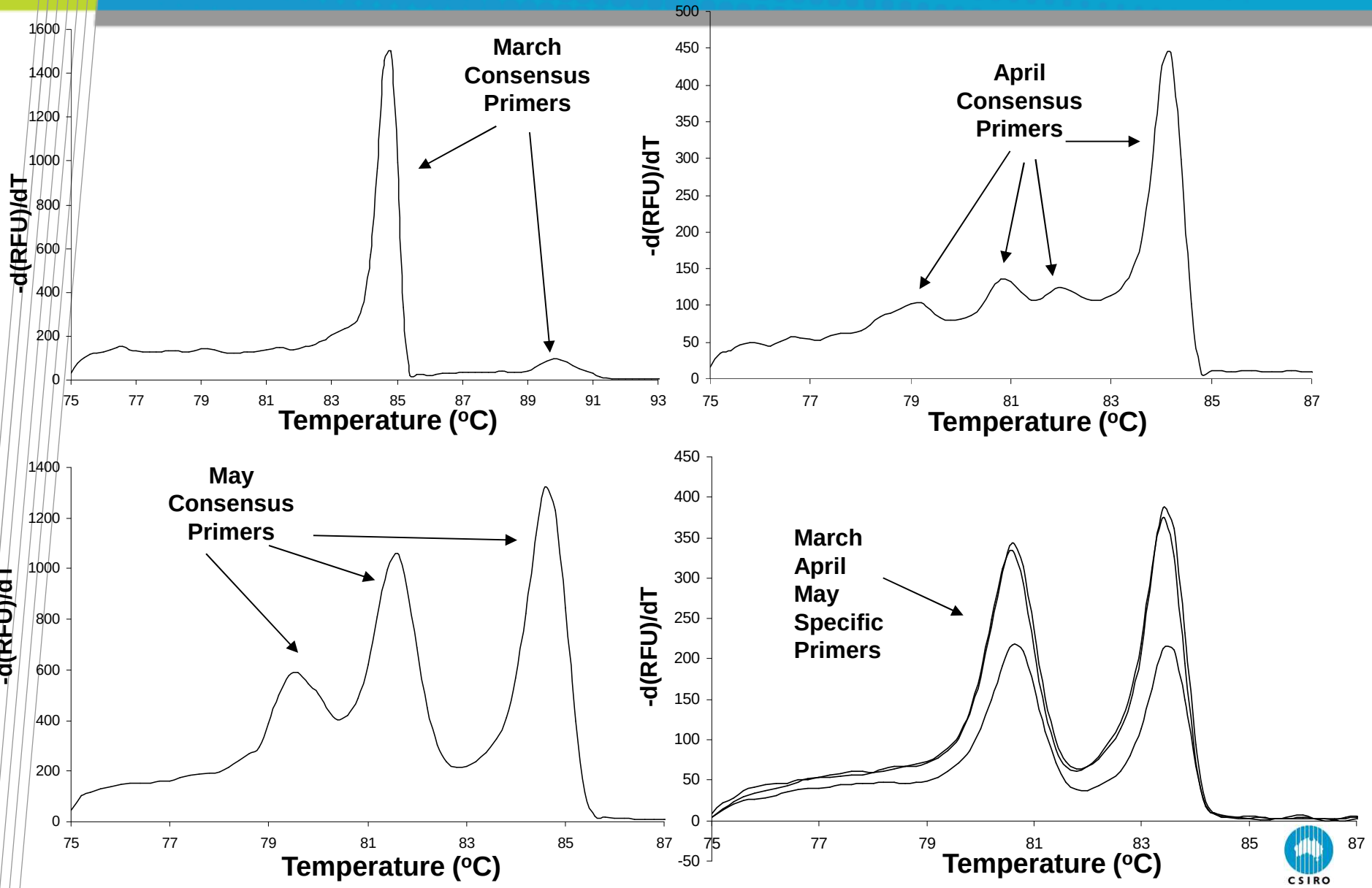
N. fowleri specific primers



No detection of other *Naegleria* sp., *Willaertia*, *Hartmannella*, or bacteria

Puzon G. J., J. A. Lancaster, J. T. Wylie, and J. J. Plumb. 2009. Rapid detection of *Naegleria fowleri* in water distribution pipeline biofilms and drinking water samples. (Environ. Sci. Technol 2009)

Comparison of detection methods for *N. fowleri* in field biofilm samples



Molecular method advantages

- Fast
  - Reduced time, 6h v. 48+ for traditional method (NNA - *E. coli* plating)
- Direct detection
  - Reduce culturing bias
    - No need for subculturing
  - Detects in bulk water and biofilm
- Specific
  - Detects only *N. fowleri* (No bias/loss by competing *Naegleria* spp.)

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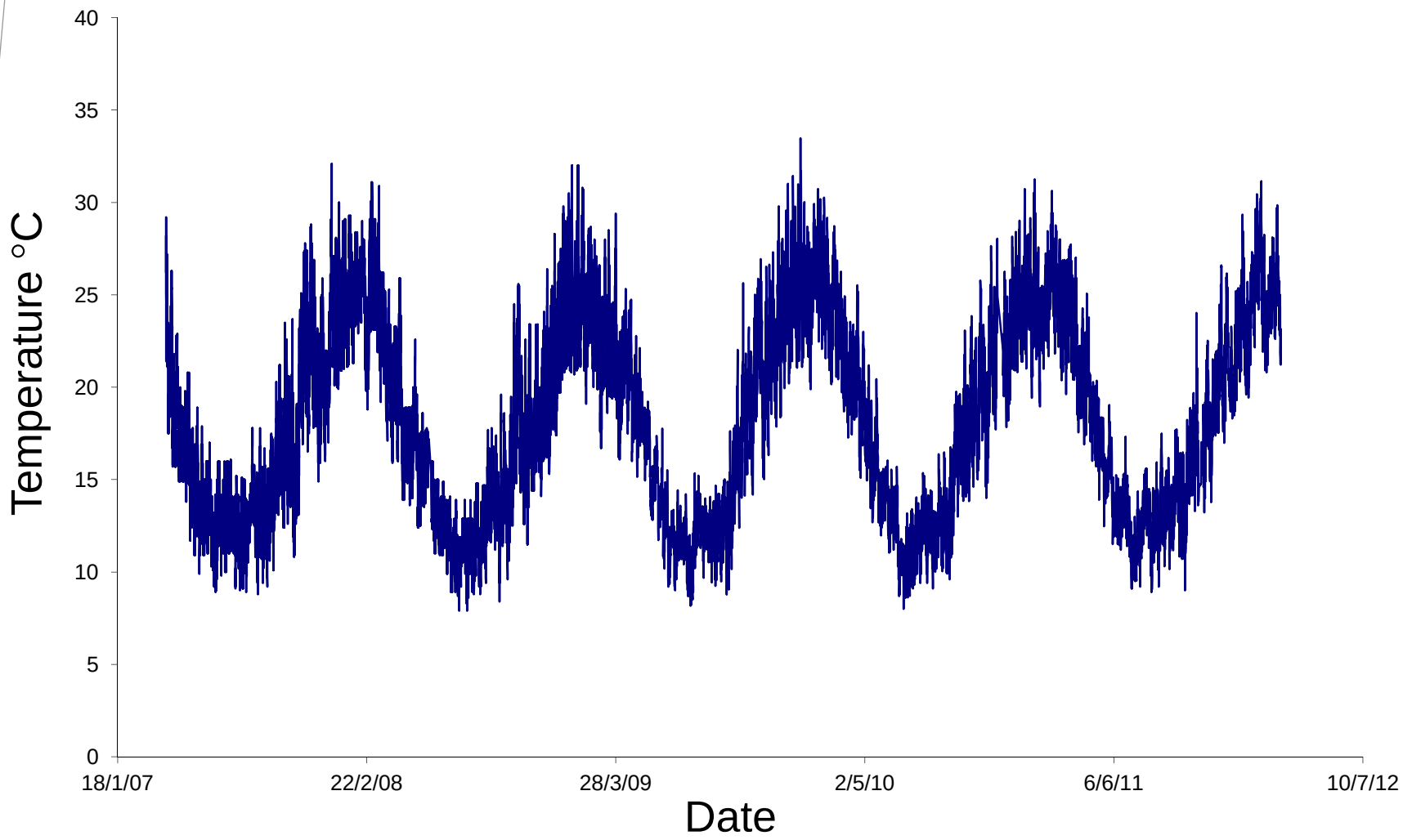
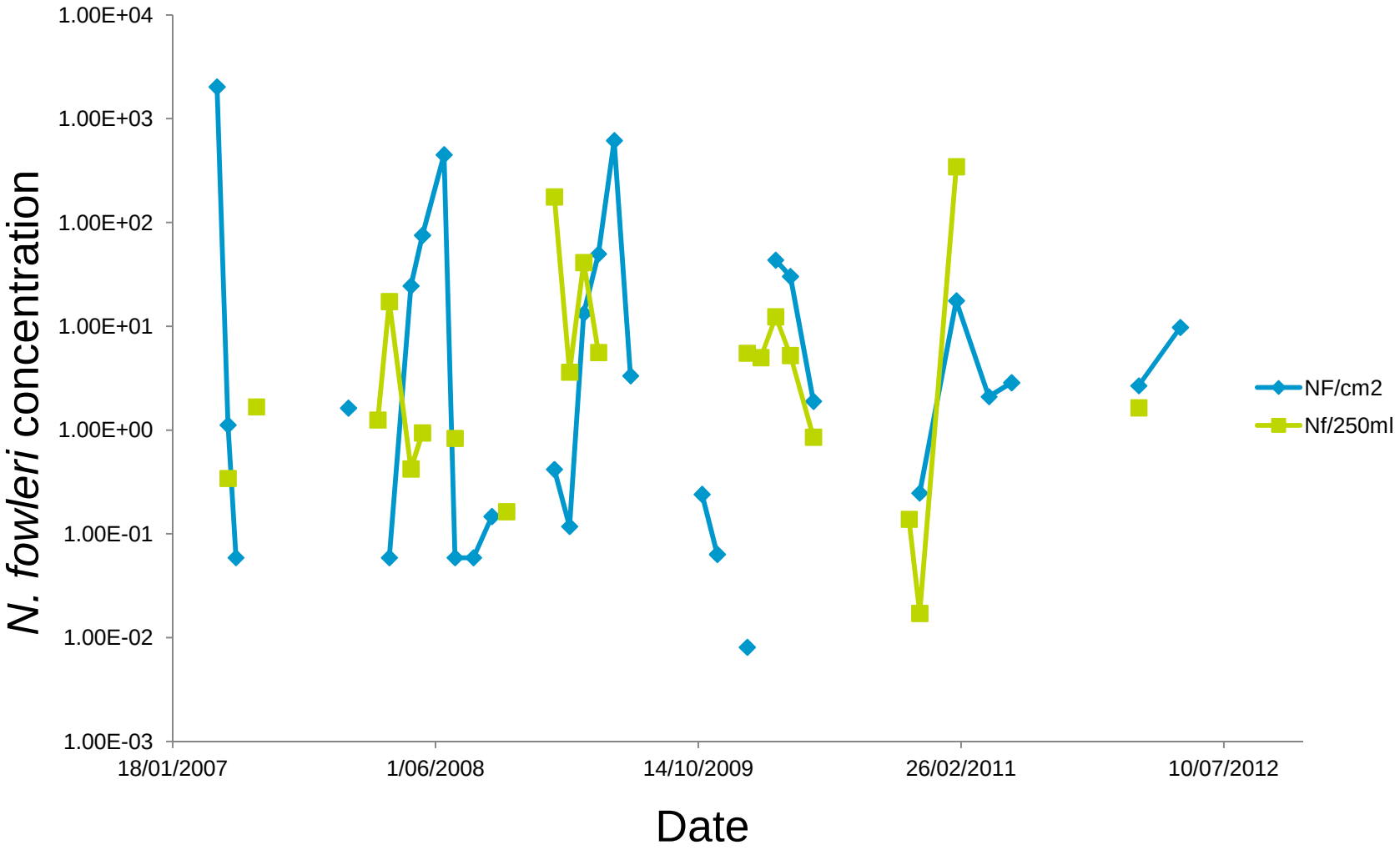
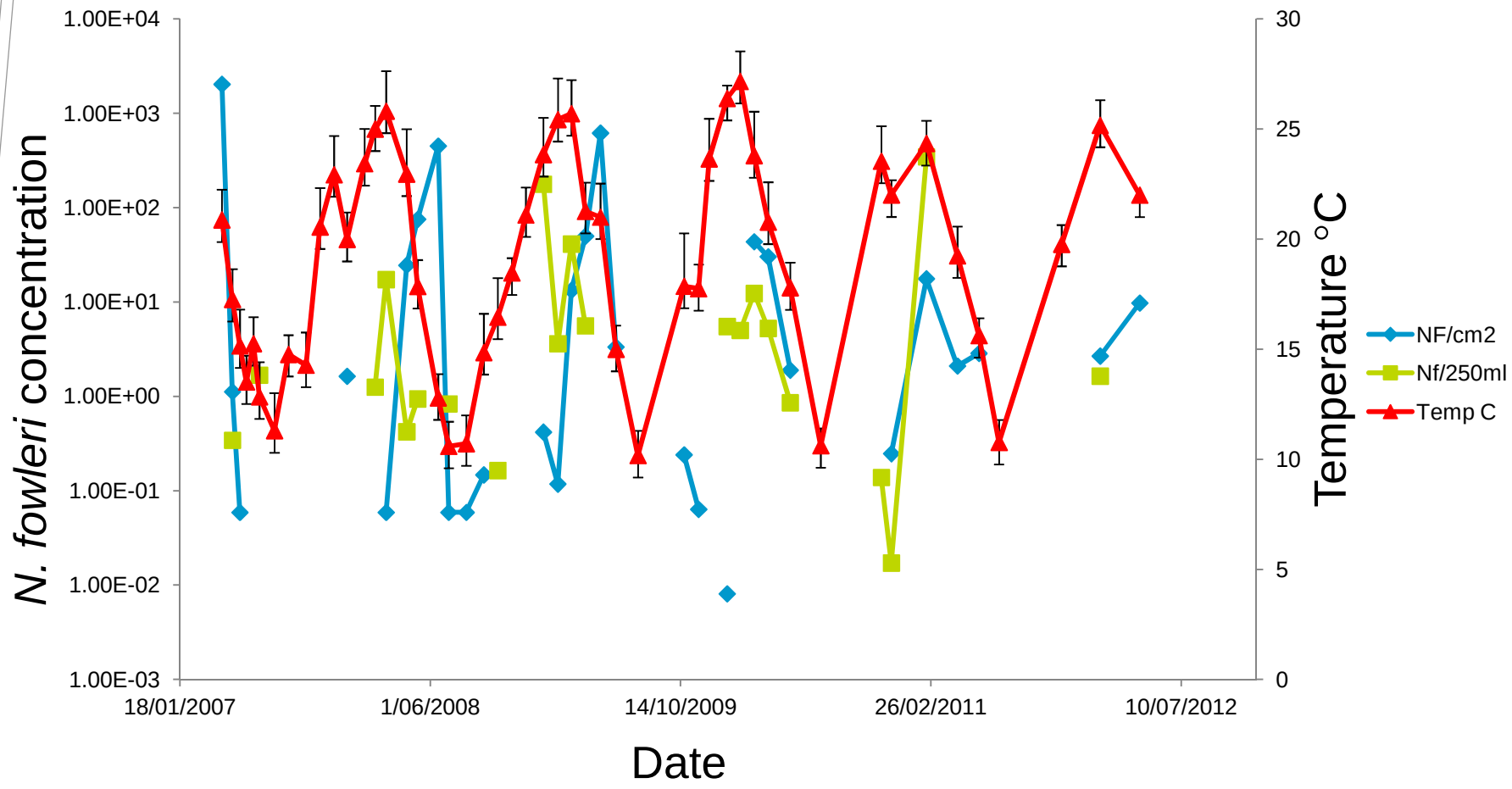
Water temperature at *N. fowleri* field site



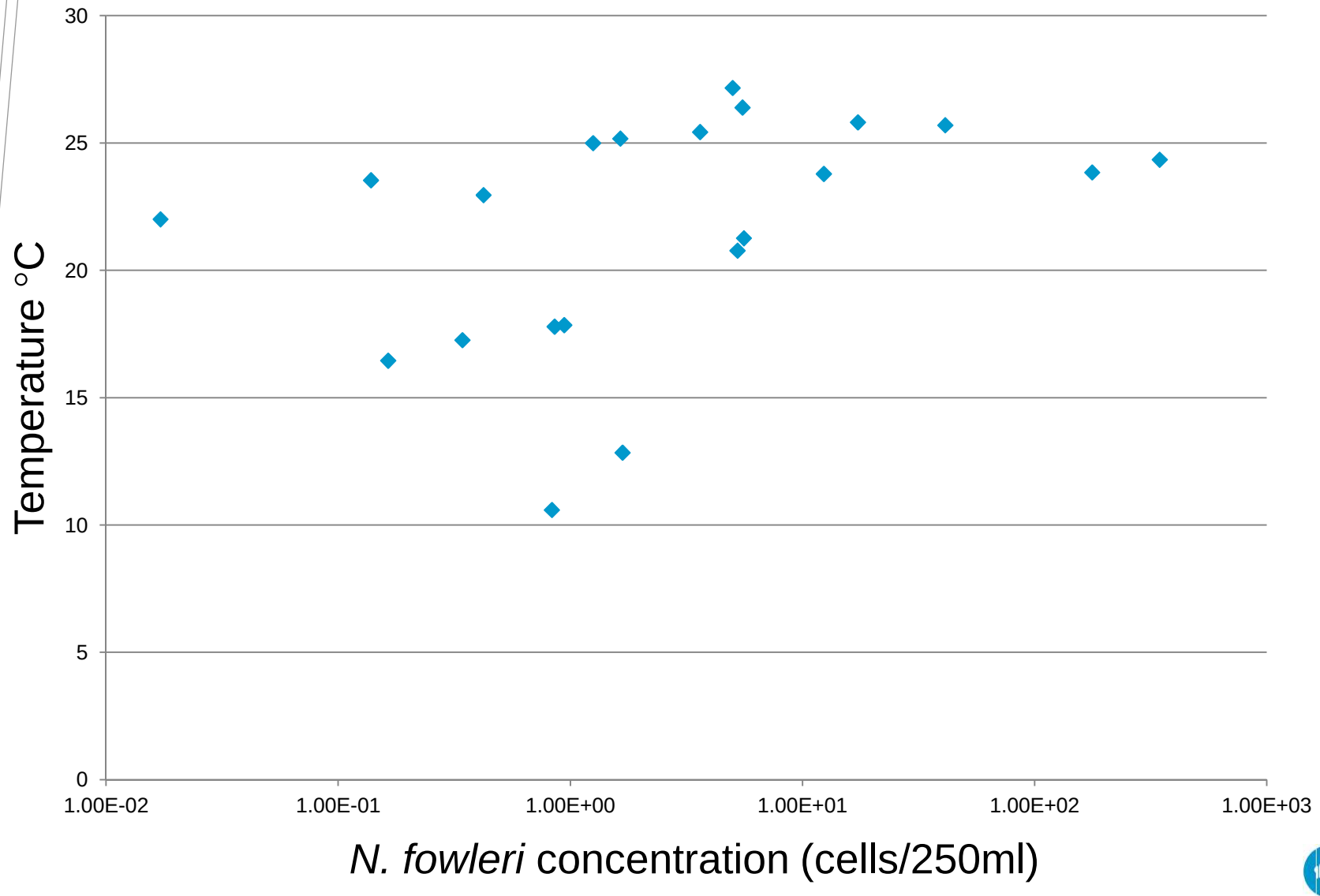
N. fowleri in biofilm and bulk water



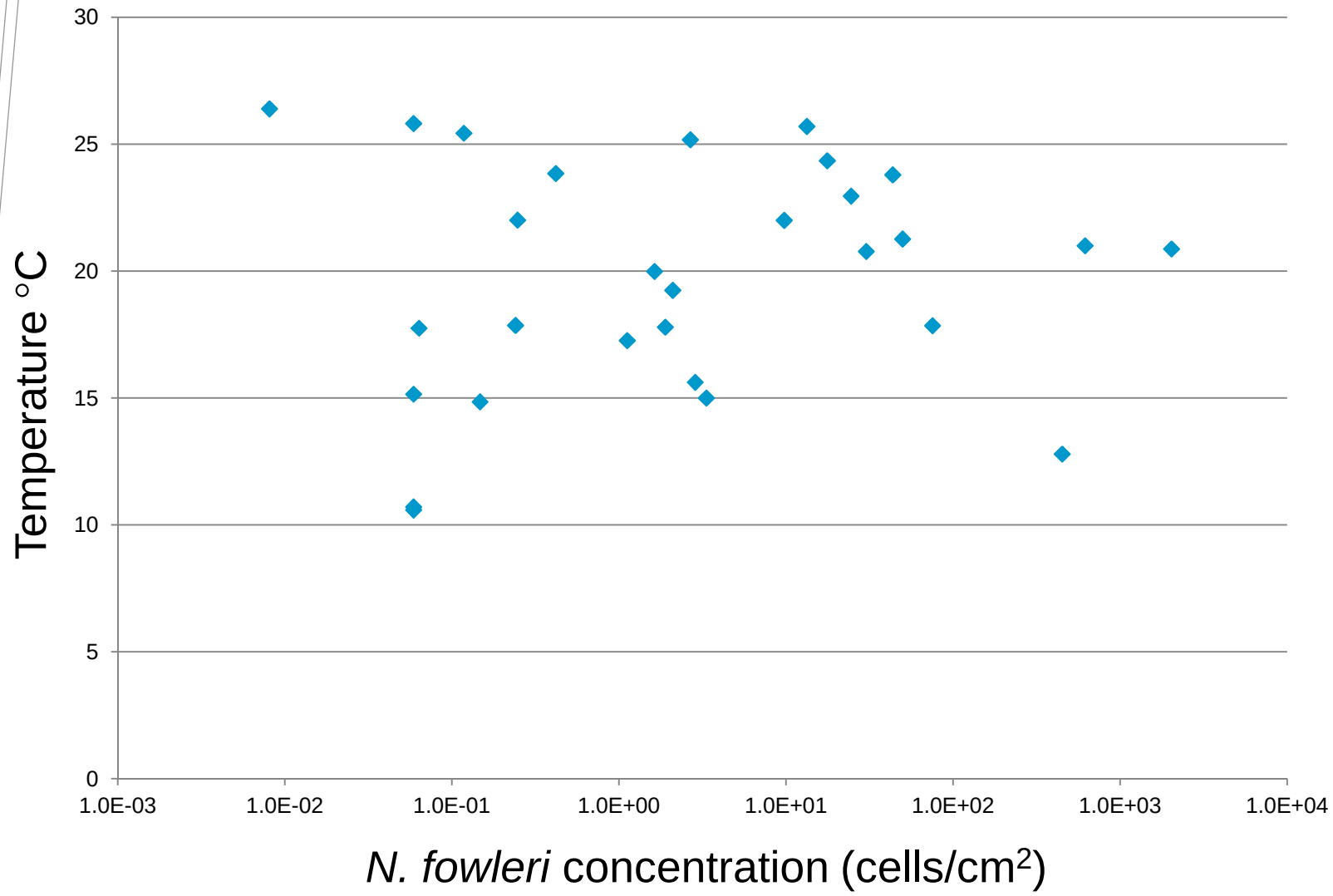
N. fowleri and Water Temperature



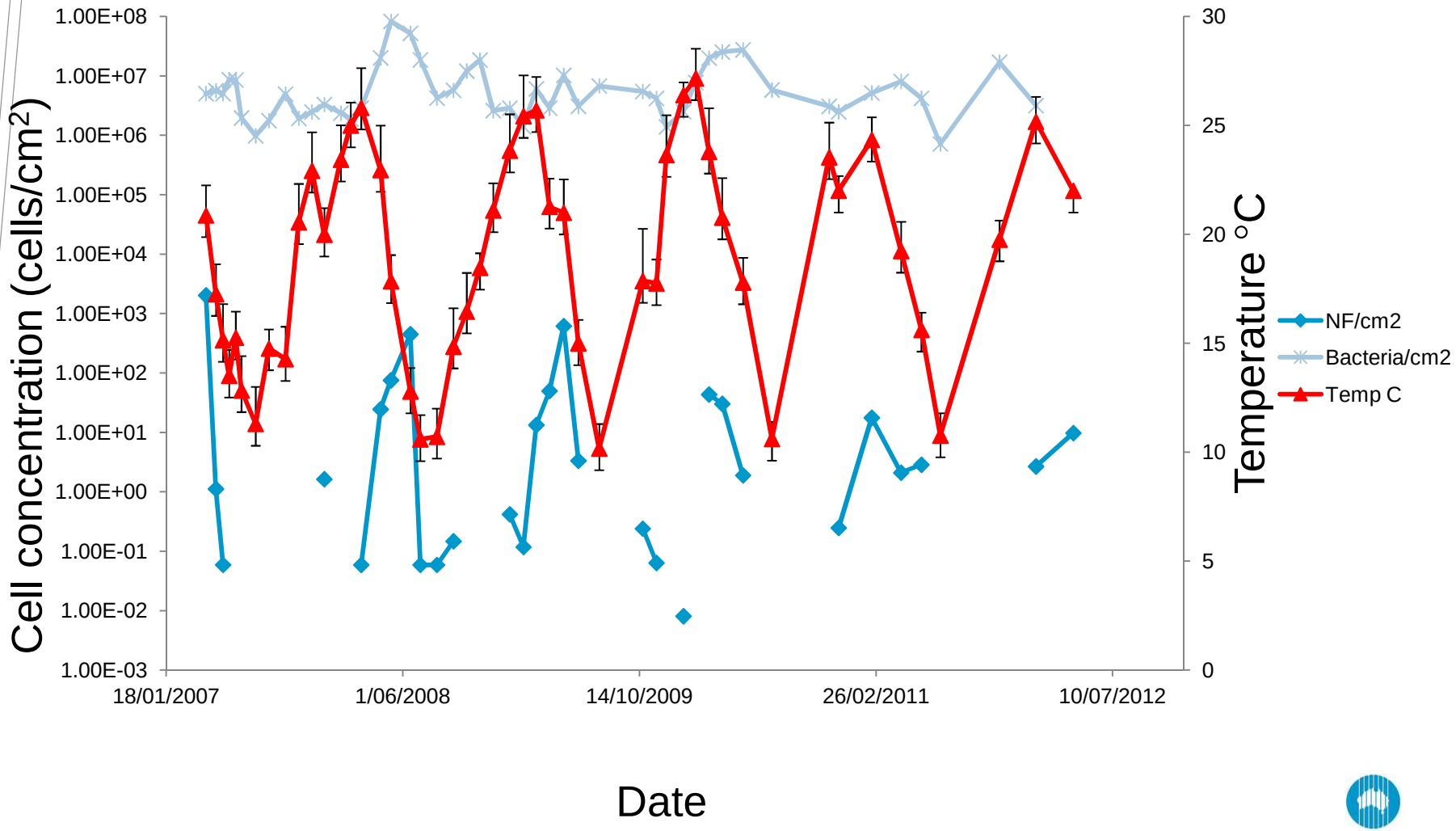
N. fowleri in bulk water v Temperature



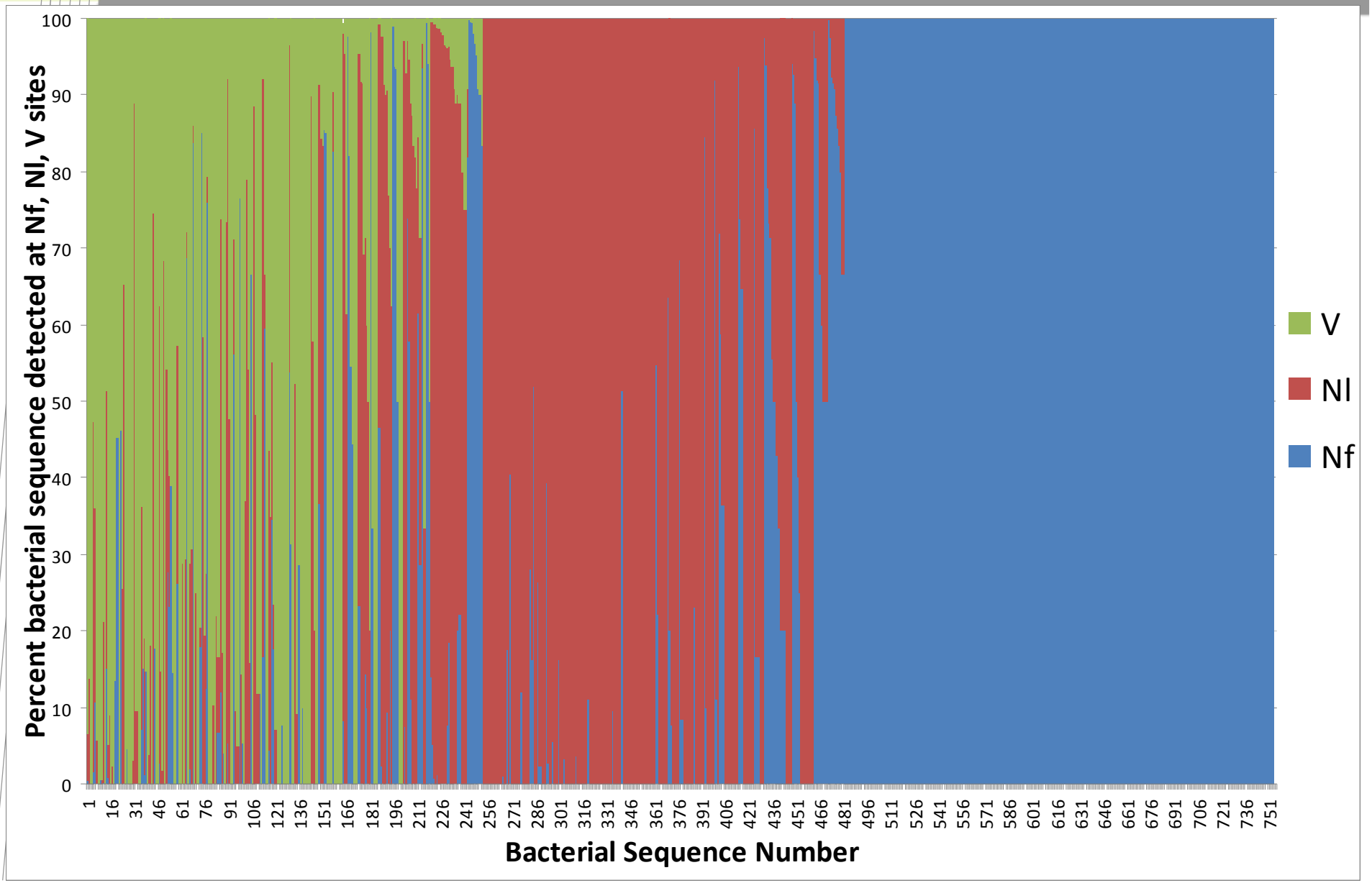
N. fowleri in biofilm v Temperature



N. fowleri- Water Temp, biofilm bacterial concentration, and *N. fowleri* in biofilm

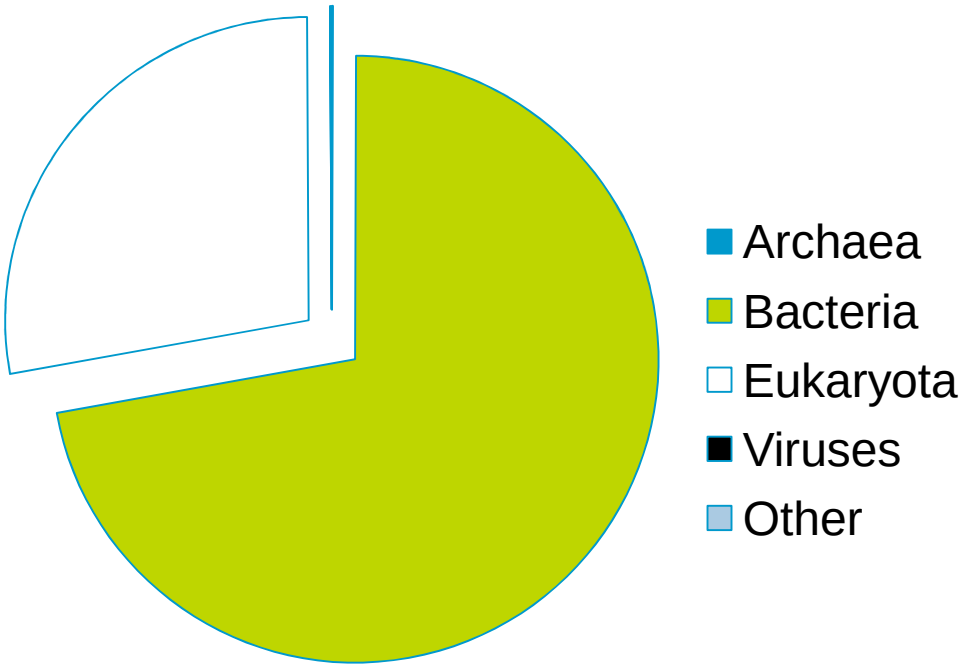


Ecogenomic analysis- pyrosequencing



Sequence analysis

Analysis Method	Total Bacterial sequences (All sites)	Unique Bacterial sequences (All sites)
DGGE	42	38
454-Pyro	753	153



Principle Component Analysis of Sequences

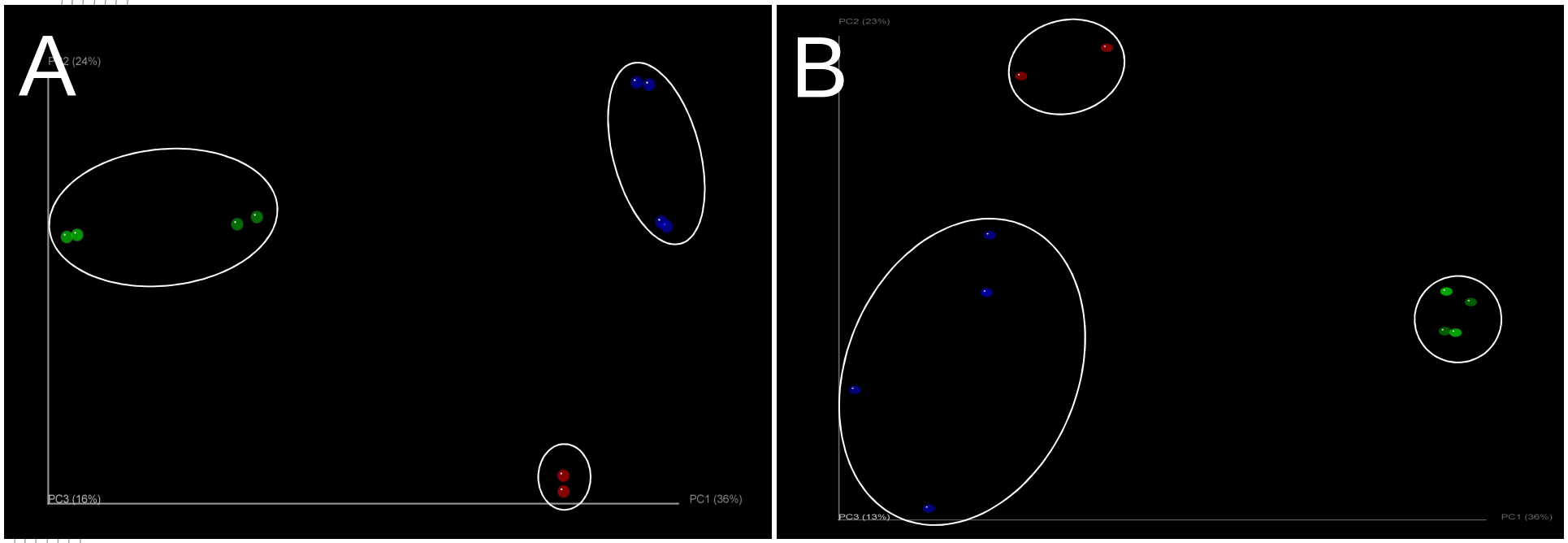


Figure 3: PCA analysis of 16s (A) and 18S (B) sequences by site. Green are *N. fowleri* positive sites, blue and red are *N. fowleri* negative locations. Each colour represents a different pipeline (circled). The pipeline that the sequences originate from explains 70 and 69% of the variance between the samples. However, those samples that are *N. fowleri* positive (green) are separate from both *N. fowleri* negative pipelines.

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