Water Research Commission Project

K5/2175

Evaluation of the risks associated with the use of rain-water harvested from rooftops, for domestic use and homestead food gardens; and groundwater for domestic use and livestock watering

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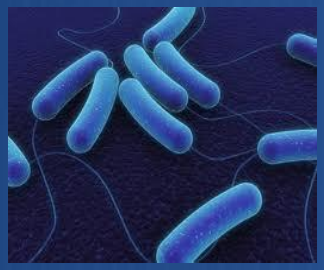
WRC Workshop Joburg
21 November 2013

WATER RESEARCH COMMISSION

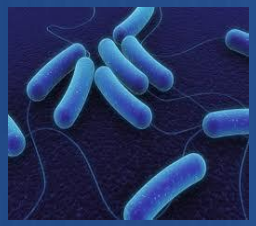
Presentation Outline

- Introduction
- Literature Review
- Research Design
- Site selection
- Sampling Strategy
- Current Results
- Future studies

Introduction: Definition



- **Rainwater harvesting** is a method used for collecting and storing rainwater from rooftops, rock surfaces using tanks and underground storage (Abdulla & Al-Shareef , 2009)



Problem statement

- Access to clean water for people and small scale farmers in the rural area of SA is a major challenge.
- &
- Rooftop harvested rain-water (RHRW) is used as an alternative source of water for personal use and food production.

- RHRW is used for drinking, and irrigation of crops without prior treatment as it is generally **assumed to be safe**.

However

- researches have reported rooftop harvested rainwater to be **contaminated** with potentially **pathogenic bacteria** including pathogenic strain of *E. coli*

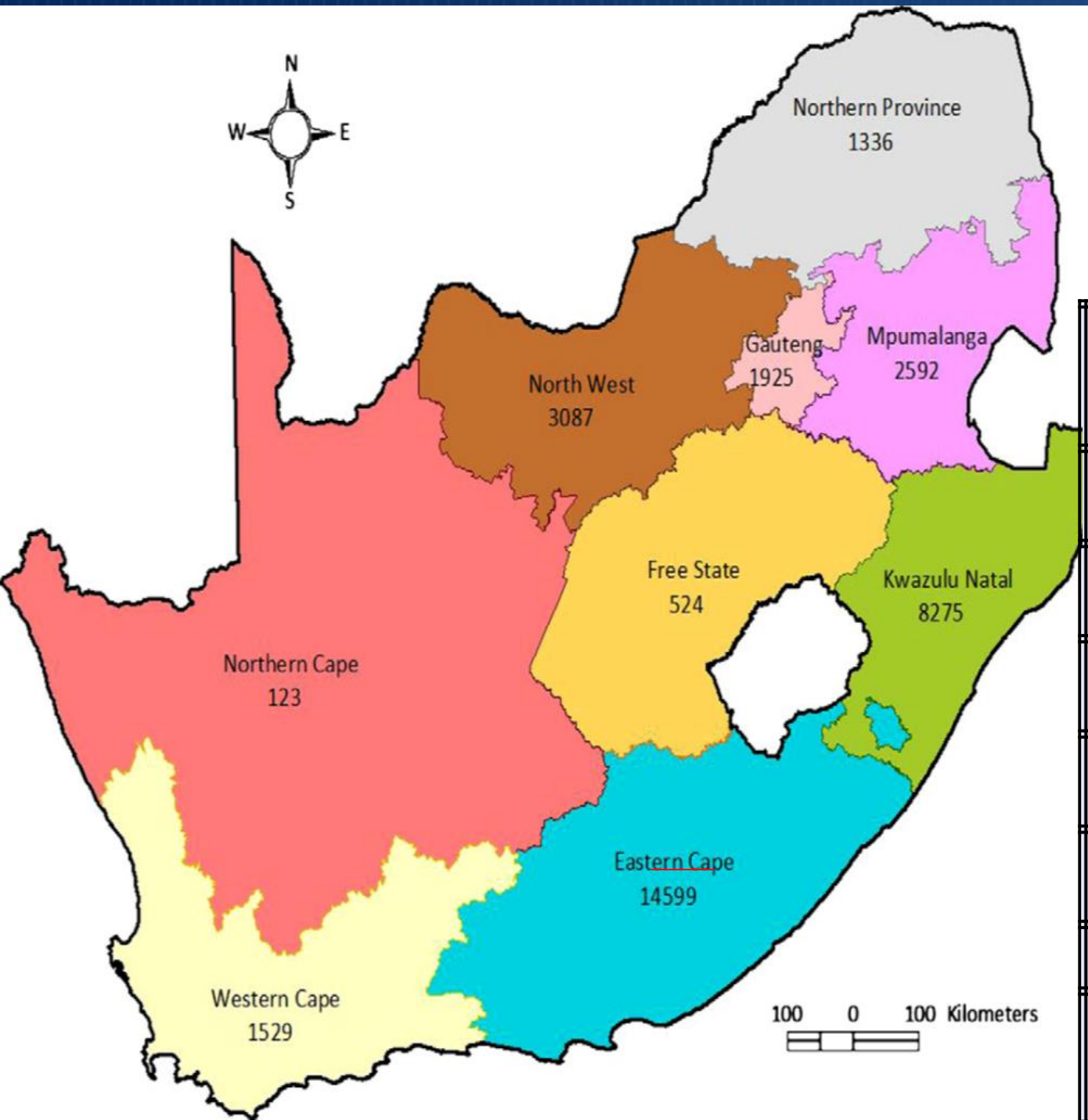
Introduction

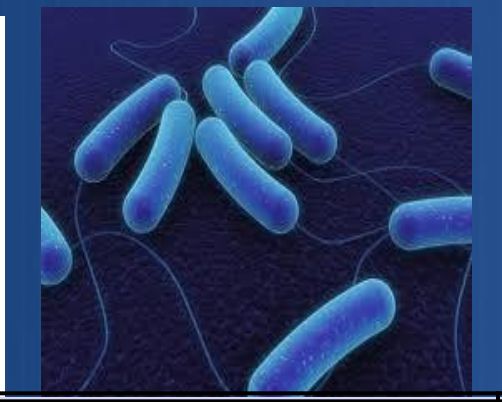
- Harvested rooftop rainwater is generally accessible to the public and is used both for domestic and subsistence agricultural purposes.
  - Likelihood exist that it is contaminated with pathogenic bacteria and / or chemicals
  - May result in disease and compromise the health and well-being of people, if directly utilised.

Introduction

- Use of rooftop rain harvested water could be to irrigate crops.
  - What is the quality of this water and what is the likelihood that it will pose a risk to the:
    - end-user
    - safety of crops irrigated with this water?
    - impact on animal health if used for drinking

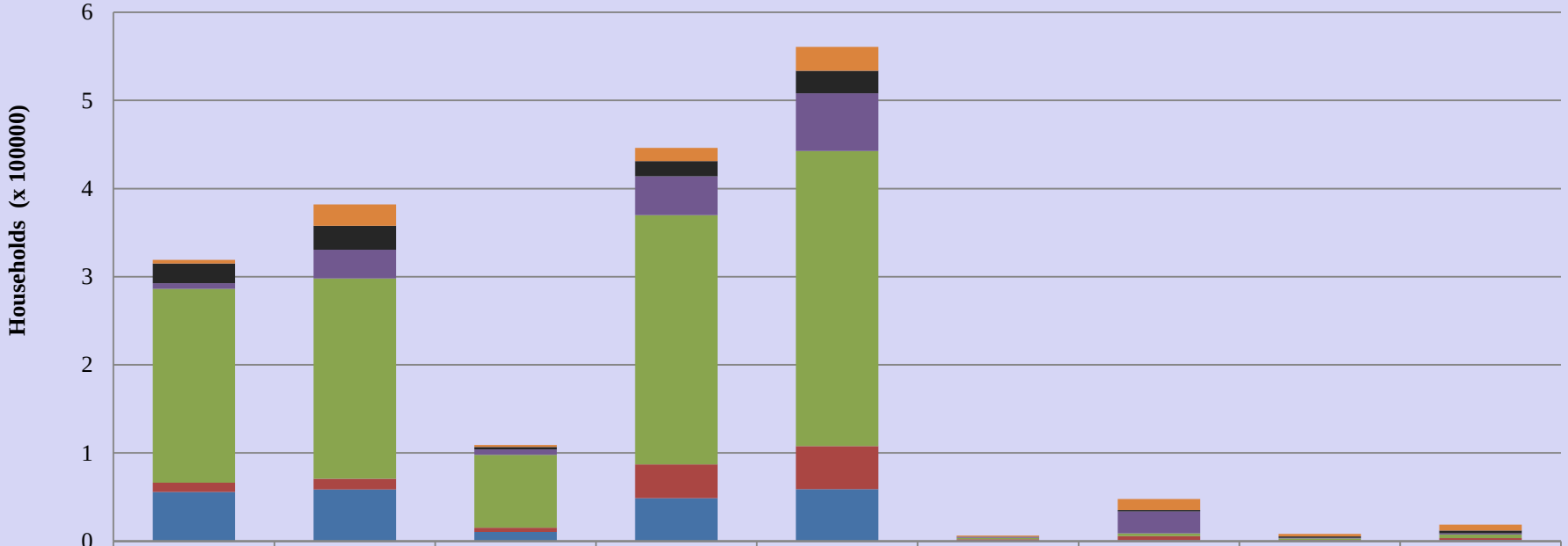
- **Therefore**: Quality of harvested rain water needs to be evaluated and specific guidelines developed.





No. of tanks used for Rainwater Harvesting in South Africa	
Eastern Cape	**14599**
KZN	**8275**
North West	**3087**
Mpumalanga	**2592**
Western Cape	**1529**
Limpopo	**1336**
Northern Cape	**123**

National Statistics on Water Sources



	EC	FS	GP	KZN	L	MP	NC	NW	WC
Other	4275	24109	2183	15034	27346	1250	12447	2933	6396
Rain water tank	22446	27214	3095	17225	25477	513	1639	1598	3333
Water vendor	6302	32663	6013	44158	65334	906	24772	556	1373
River/stream	219959	227019	82488	282770	335094	1746	3149	1820	3924
Dam/pool	10406	12129	4995	38309	48807	1899	4832	796	2266
Spring	55868	58752	10301	48824	58853	83	899	457	1270

Abbreviations: EC (Eastern Cape), FS (Free State), G (Gauteng), KZN (KwaZulu-Natal), L (Limpopo), MP (Mpumalanga), NC (Northern Cape), NW (Northwest), WC (Western Cape)

Number of households using non-piped water sources in South African Provinces

Knowledge gaps

- Roof harvested rainwater
  - Potential public health risks associated with various pathogens that could be present in faeces of birds, domestic animals, reptiles and insects.

- Literature on the occurrence of zoonotic bacterial and protozoa pathogens in rainwater tanks in rural communities especially in South Africa is scarce.

- No consensus on the levels of contamination and risks

Knowledge gaps

- Rural areas - domestic animals are kept within the vicinity of the homestead and faecal matter is common on the ground.
- Most research in roof harvested rainwater has been conducted in urban areas.
- Different environmental settings compared to rural settings.
- Hence the data cannot be directly extrapolated for implementation in rural communities.

Literature Review: Risk Assessment

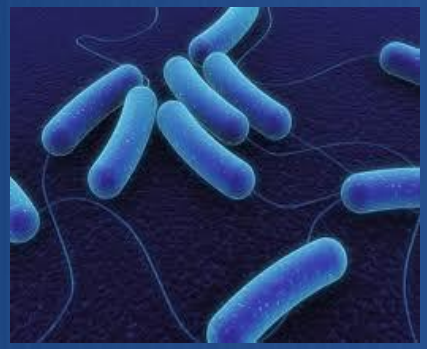
Risks associated with rain water:

- Challenges – no recent statistical data on rainwater harvesting (last comprehensive data available was published in 2007).
- Estimates have been used which at times give conflicting data.
- Risk assessment draft based on the initial published paper of Prof Norman Casey.
- Inputs were obtained from Dr Sebastian Jooste from the Department of Water Affairs (to become a working guideline).
- Additional inputs will depend on data gathered during future research.

Knowledge gaps

- Faecal matter deposited on rooftops as dust or droppings is the major source of contamination
- High solar radiation especially in sub-Saharan Africa may play a significant role in sanitising roof surfaces.
- No information currently exists on the prevalence of different pathogenic bacterial species in faces of domestic animals and birds that are the likely major sources of contamination.

Sources of microbial contamination for RHRW



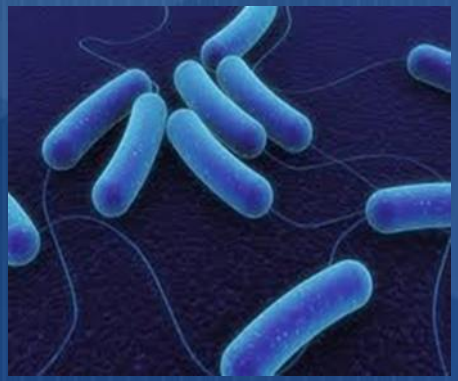
Knowledge gaps

- It is also not known how long pathogens can survive and persist on crop surfaces after irrigation with contaminated water.
- Pathogens that are transferred into rainwater storage tanks can survive and persist in biofilms.
- Biofilms offer protection to the bacterial community which may lead to their proliferation and shedding into the water.
- On the contrary, biofilm has been reported to sequester heavy metallic ions from stored rainwater.

Research Aims

1. To determine fitness for use of rainwater collected from rooftops for homestead food gardens

2. To determine fitness for use of rainwater collected from roofs for livestock production systems and domestic use



Research Aims

1. To review literature of risks associated with rooftop harvested rainwater for domestic use and for homestead gardens

2. To optimise microbiological techniques to monitor rooftop rainwater and groundwater

3. To characterise planktonic and biofilm-forming microorganisms that develop in harvested and surface water stored in 750L low density polyethylene water storage tanks

Selected Sites

Jonathan Dennison

Eastern Cape Province: Luthengele* Village

- Steep terrain- municipal water supply is too expensive to implement.
- People rely mainly on river and rain harvested water.
- Government projects: rainwater tanks installed for potable and domestic food gardening purposes.
- A non-governmental organisation (WESSA) - promoting organic agriculture in domestic food gardens.

Selected Sites

Scientific Roets

Northwest Province (Jericho)

- Rural community – impacted by the HIV/AIDS epidemic, unemployment, and poor service delivery.
- Water is pumped into reservoirs, from where it is then distributed to households.
- Municipality often cuts off water to parts or the entire village for weeks or months at a time.
- Significant number of households do not receive piped water and depend on roof-harvested rainwater as the sole source of clean water.
- In the drier months people resort to river water, which they at times mix with rooftop harvested rainwater or ground water for domestic use.

Selected Sites

Selected Sites

Limpopo Province (Ga-Molepane)

- No access to piped water – community rely on river, borehole, or roof-harvested rainwater.
- Operation Hunger (NGO) - installed a number of rainwater harvesting tanks for potable and food production purposes.
- Domestic food gardening - household and community level.
- People provided with training on gardening as well as agricultural inputs.

Selected Sites

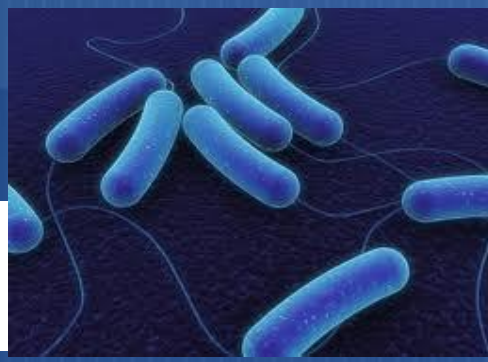
Modern Rainwater Harvesting systems

- Weltvreden* Park (Gauteng Province);
- Ifafi (North West province)*
  - Clean surrounding environment
  - Clean roof
  - First flush diverter
  - Filtration system

Escherichia coli

- *E. coli* is an inhabitant of the human and animal intestines .

- *E. coli* strain fall into seven main Phylogenetic groups (A, B1, B2, C, D, E, F) most virulent strains (group B2 and D) most non pathogenic strains belong to (group A, B1) (Clermont, et.al 2012)



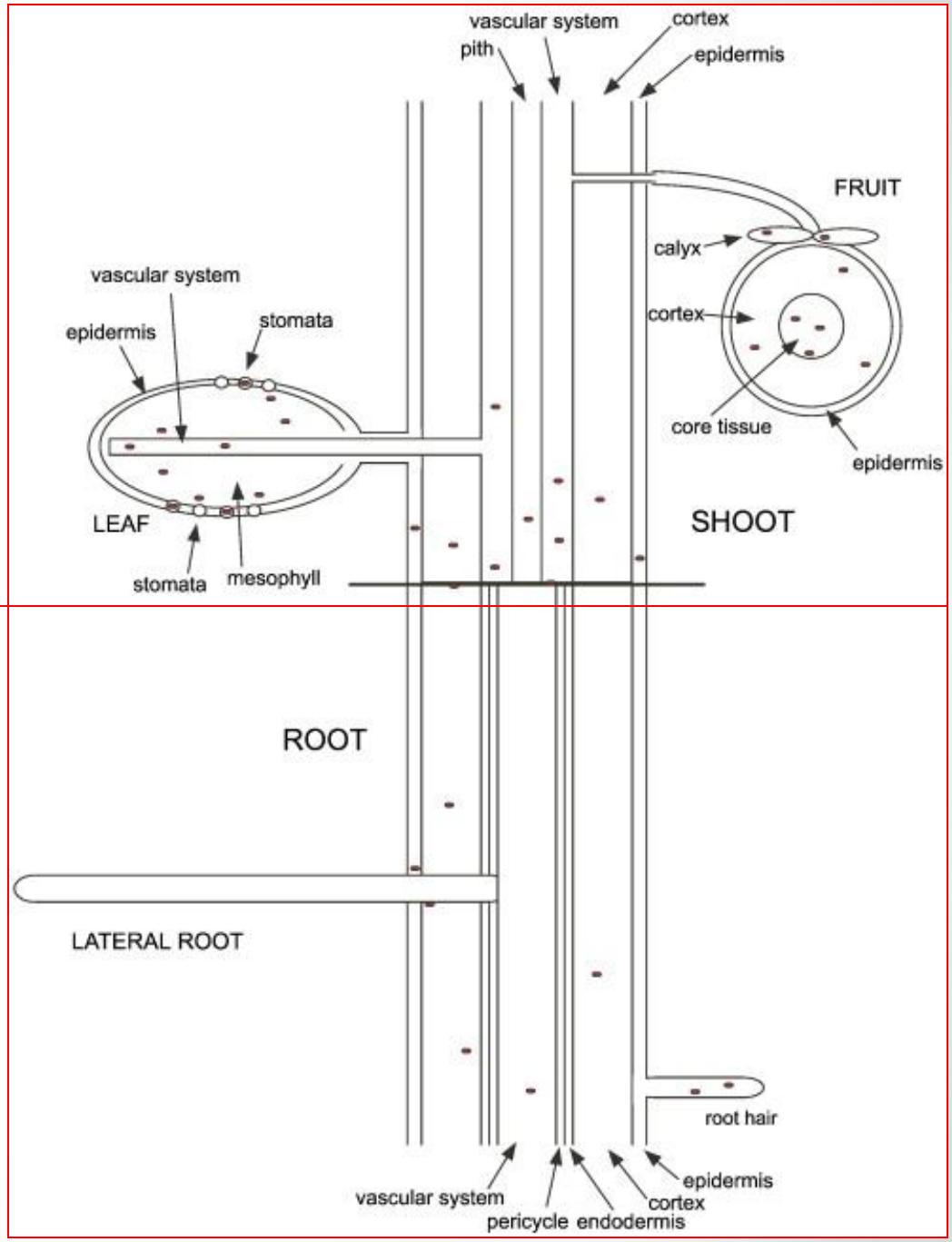
E. coli contaminated RHRW + crop irrigation

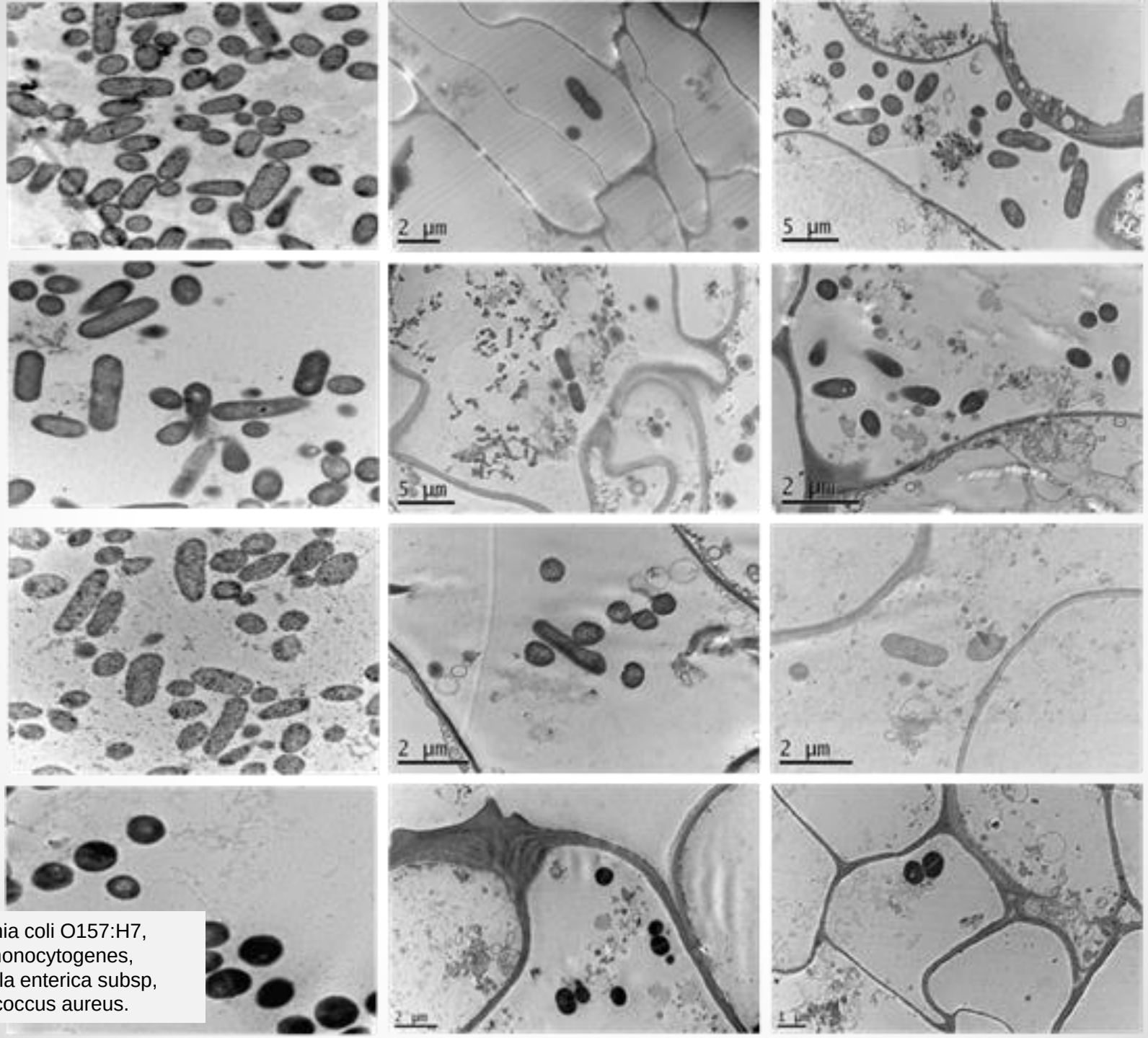
- Contaminated Irrigation water
- *E.coli* 0157:H stays in the plants by colonising the cell wall of epidermal and cortical cells of spinach.

Hypothesis

- *E. coli* strain found in contaminated RHRW are non pathogenic and the *E. coli* bacterium does not persist on cabbage and leafy greens irrigated with the contaminated water.

Deering et al, 2012





Escherichia coli O157:H7,
Listeria monocytogenes,
Salmonella enterica subsp,
Staphylococcus aureus.

Water quality and biofilm formation

Water storage

Irrigation systems

Use of Indicator Bacteria

- Use of total microbial counts, faecal coliforms and *E. Coli* has traditionally been used to evaluate levels of contamination and to determine risk.
- In roof harvested rainwater systems - traditional indicators are not necessarily correlated to the presence of pathogens.
- Pathogenic bacteria - detected in stored roof harvested rainwater presumed to be clean by traditional methods.
- Need to evaluate potential bacteria that may serve as better indicators of faecal pollution.

Use of Indicator Bacteria

- *E. Coli* - outlived by a number of pathogens including *salmonella* + *campylobacter* species.
- *Enterococcus* spp have been reported to relatively persist longer and are potentially a better indicator in harvested rainwater than *E. coli.*
- Risk assessment can therefore not be based on *E. coli* and extrapolated correlation to the presence of pathogens as used in river water systems.
- Microbial risk calculation -abundance of individual pathogenic bacterial species- (QMRA).
- Water is also contaminated when collected and stored in the household prior to use.
    - Cultural + hygiene practices

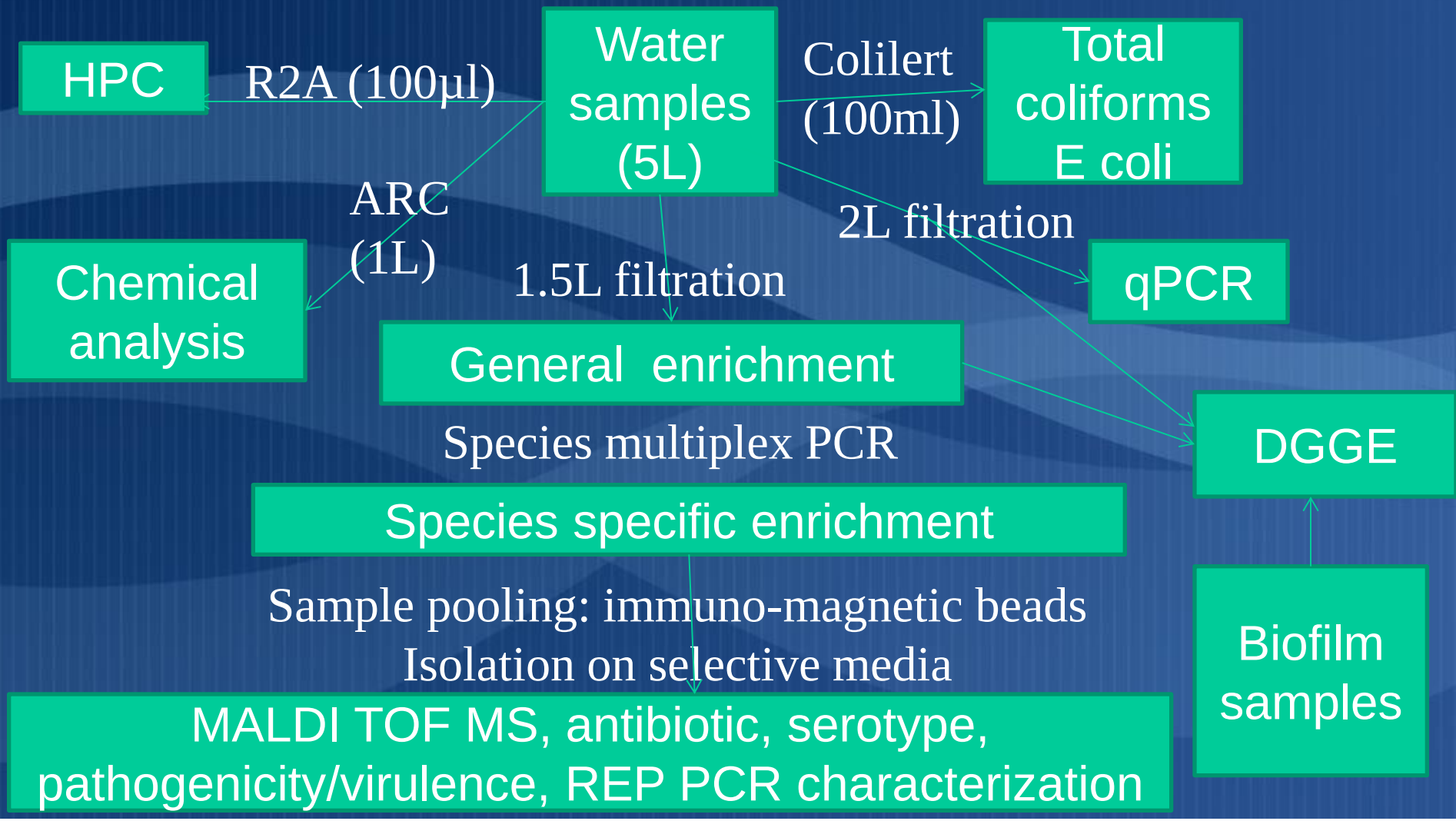
Microbiological methods

- Chromogenic media
- Multiplex PCR analysis using pathogen specific primers
- Matrix Assisted Laser Desorption Ionisation-Time of Flight (MALDI-TOF)
- 3M Molecular Detection System (3M St. Paul, Minnesota, US)

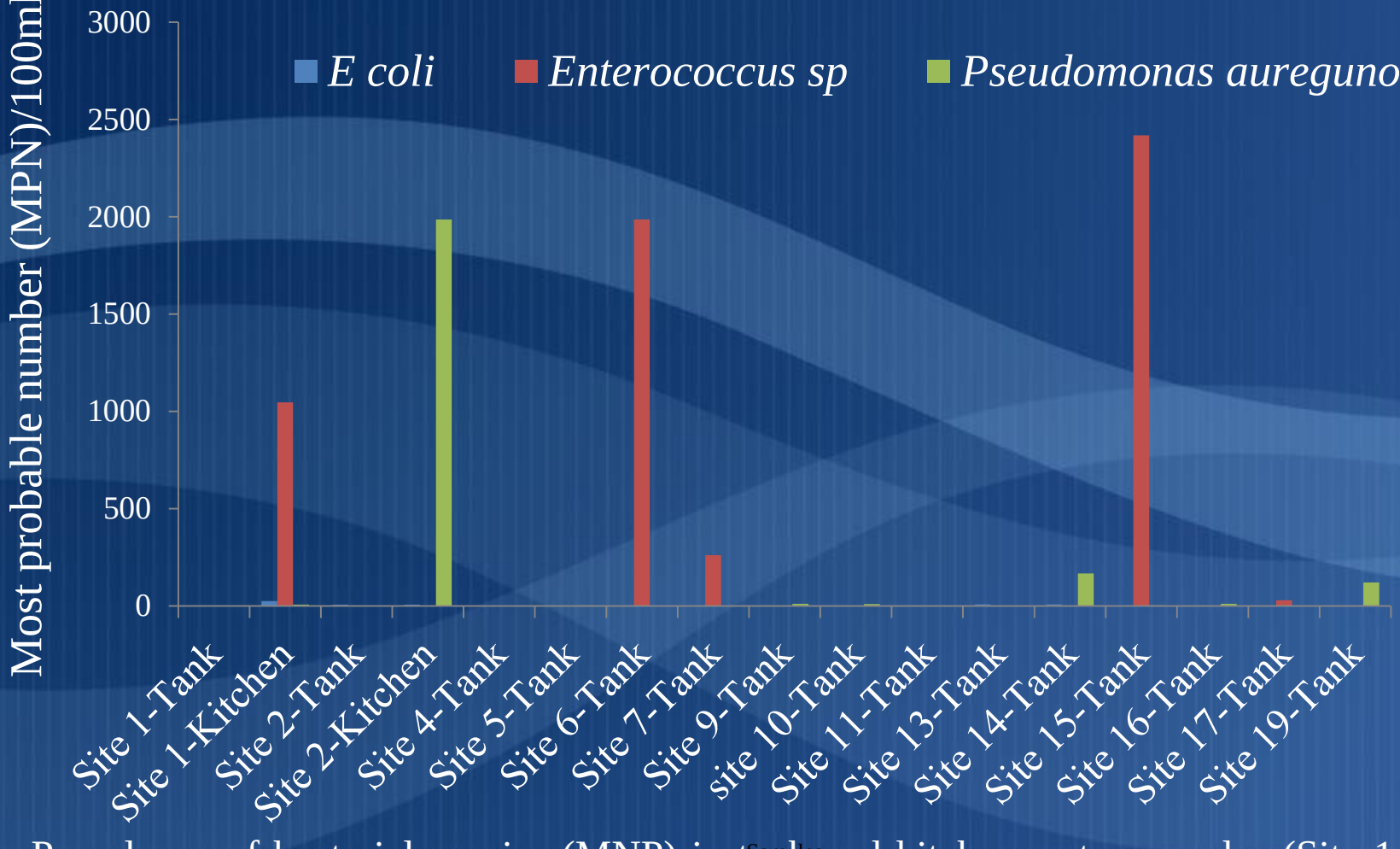
Microbial Analysis Techniques

- **Rapid detection: qPCR**
  - Based on the identified gaps in the literature, there is a need to:
    - evaluate the prevalence of different pathogenic bacteria populations in stored rainwater.
    - quantitative PCR detection of potential pathogens by targeting specific virulence genes.
  - Cultivation of most pathogens is difficult - qPCR detection + quantification of specific virulence genes circumvent the challenge
  - Enables the quantification of pathogens in viable but non-culturable state.
- Denaturant gradient gel electrophoresis (DGGE) and pyrosequencing
  - Characterise microbial populations and detection of potential sources of contamination in aquatic systems without cultivation.

Experimental Design

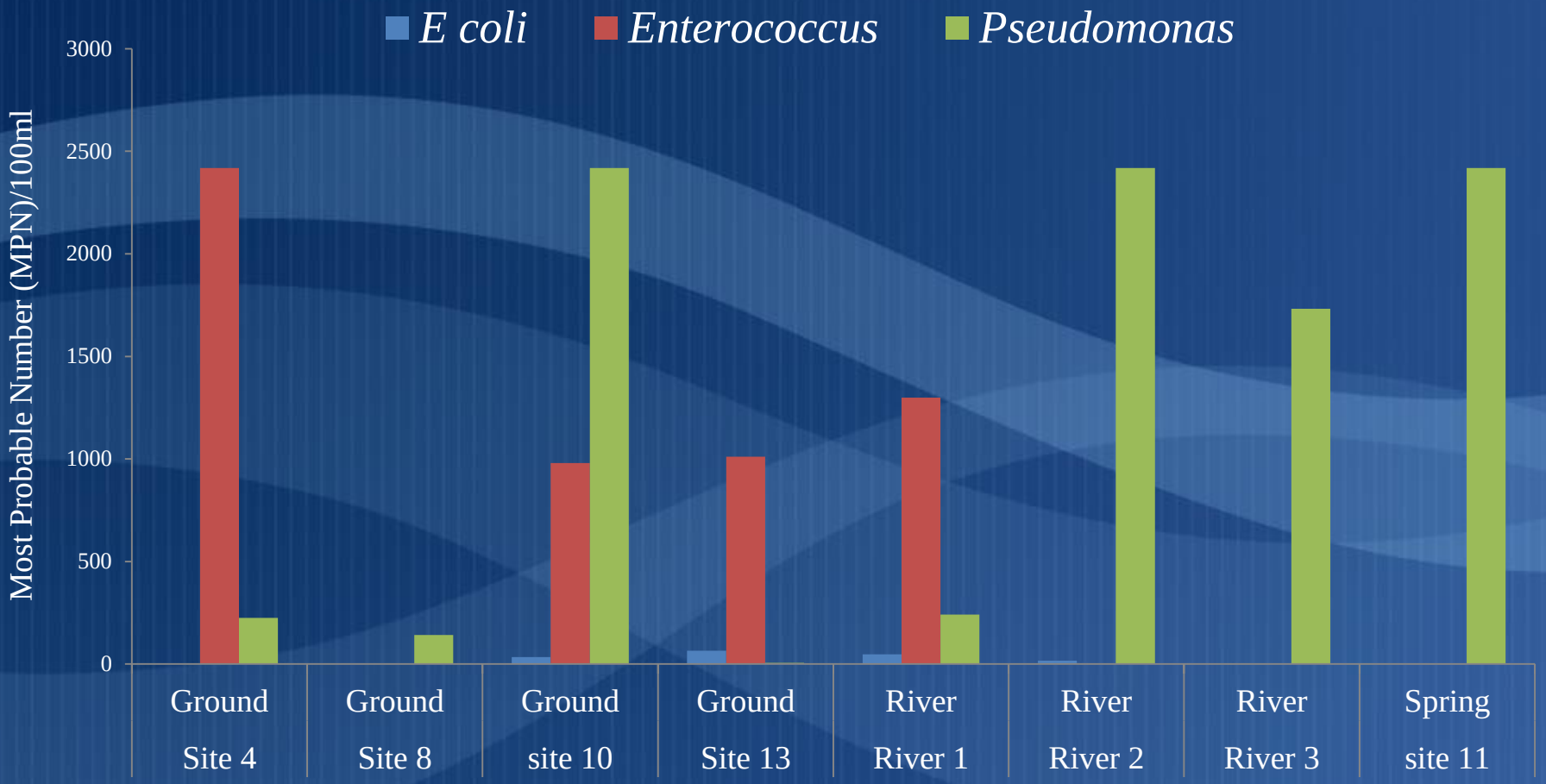


Preliminary Results



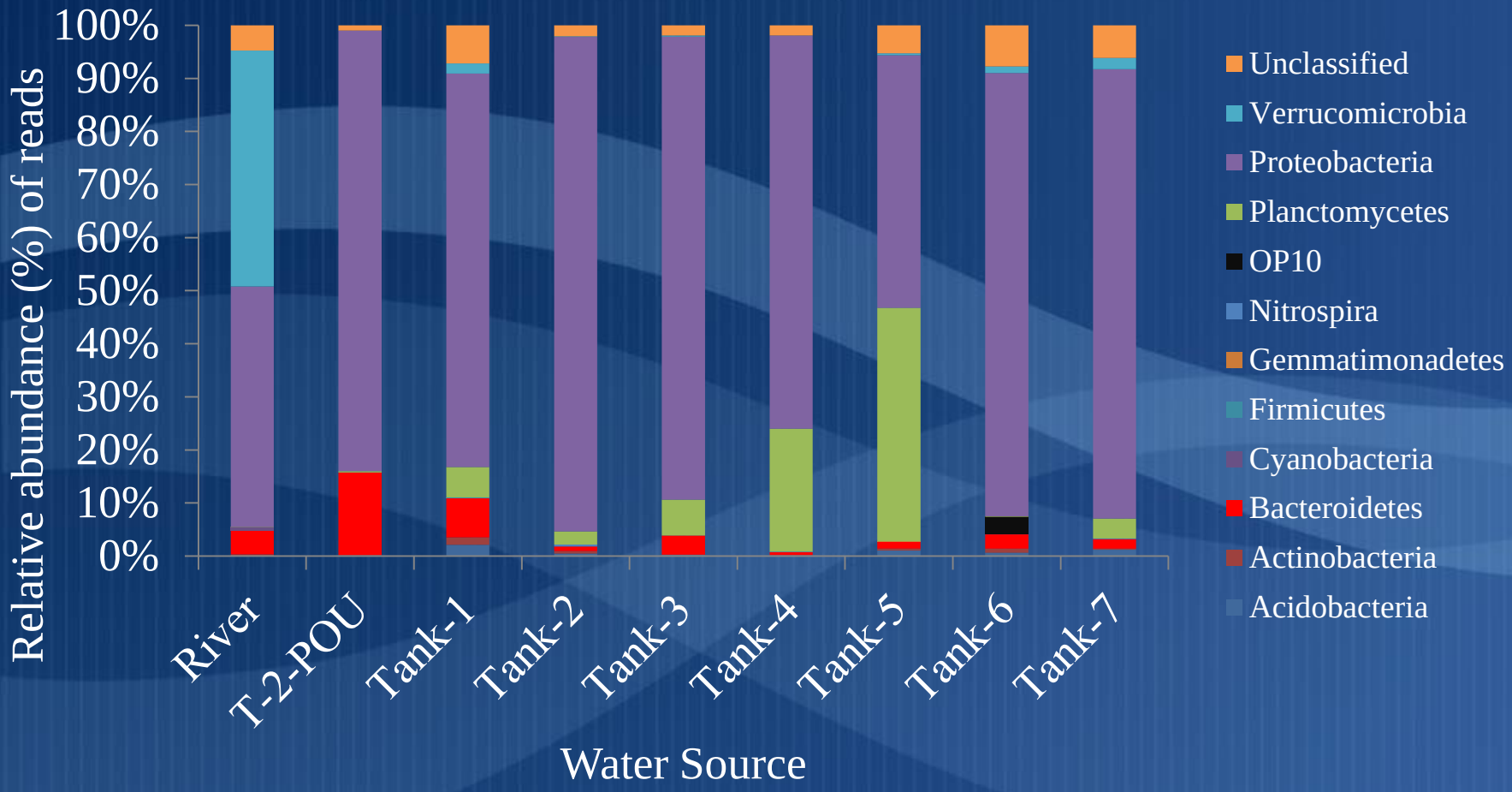
Prevalence of bacterial species (MNP) in tank and kitchen water samples (Site 1 and 2) and tank water only (Site 4-19).

Microbial Counts

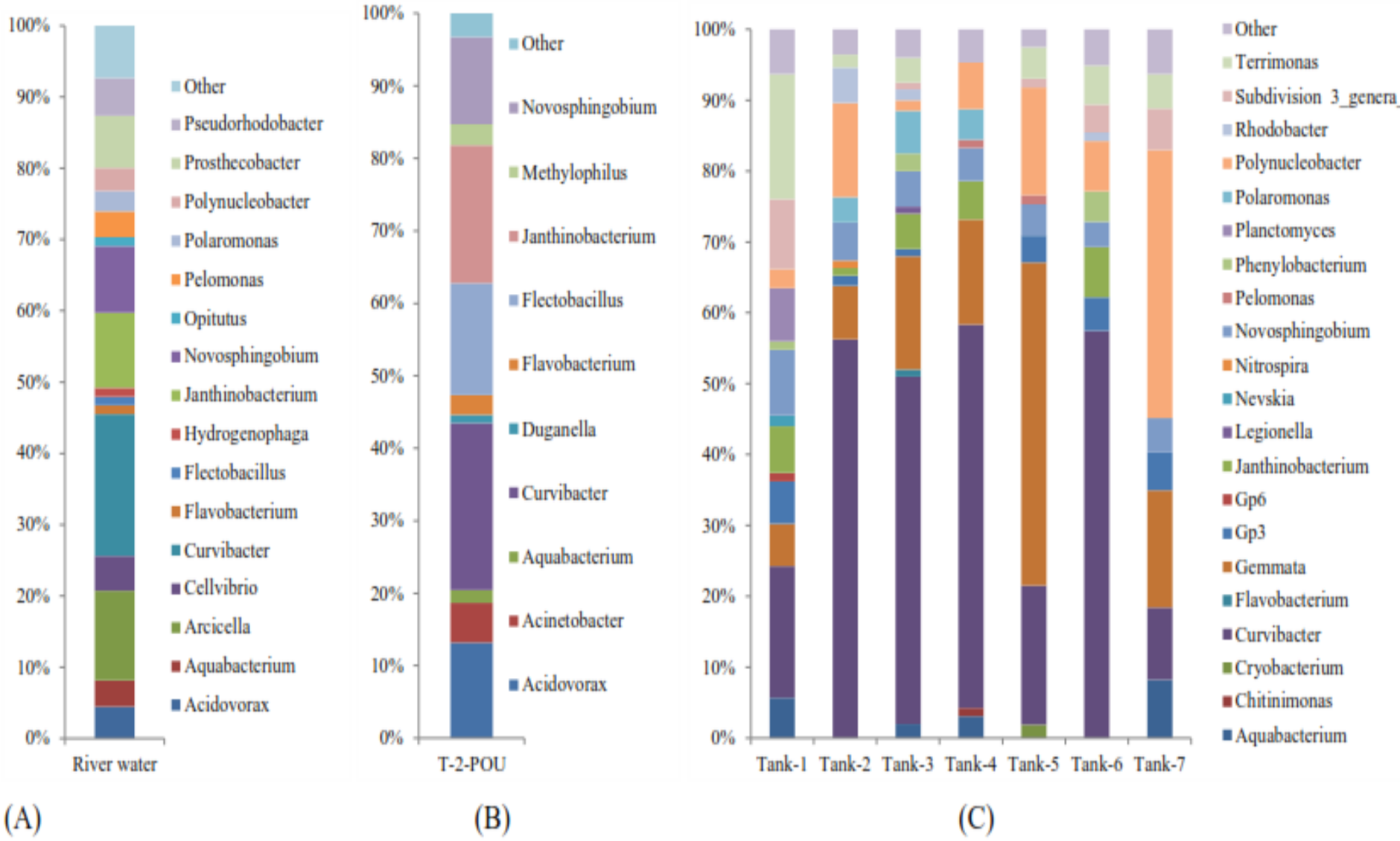


Prevalence of bacterial species (MNP) in ground harvested rainwater used for irrigation and river and spring water used for potable purposes.

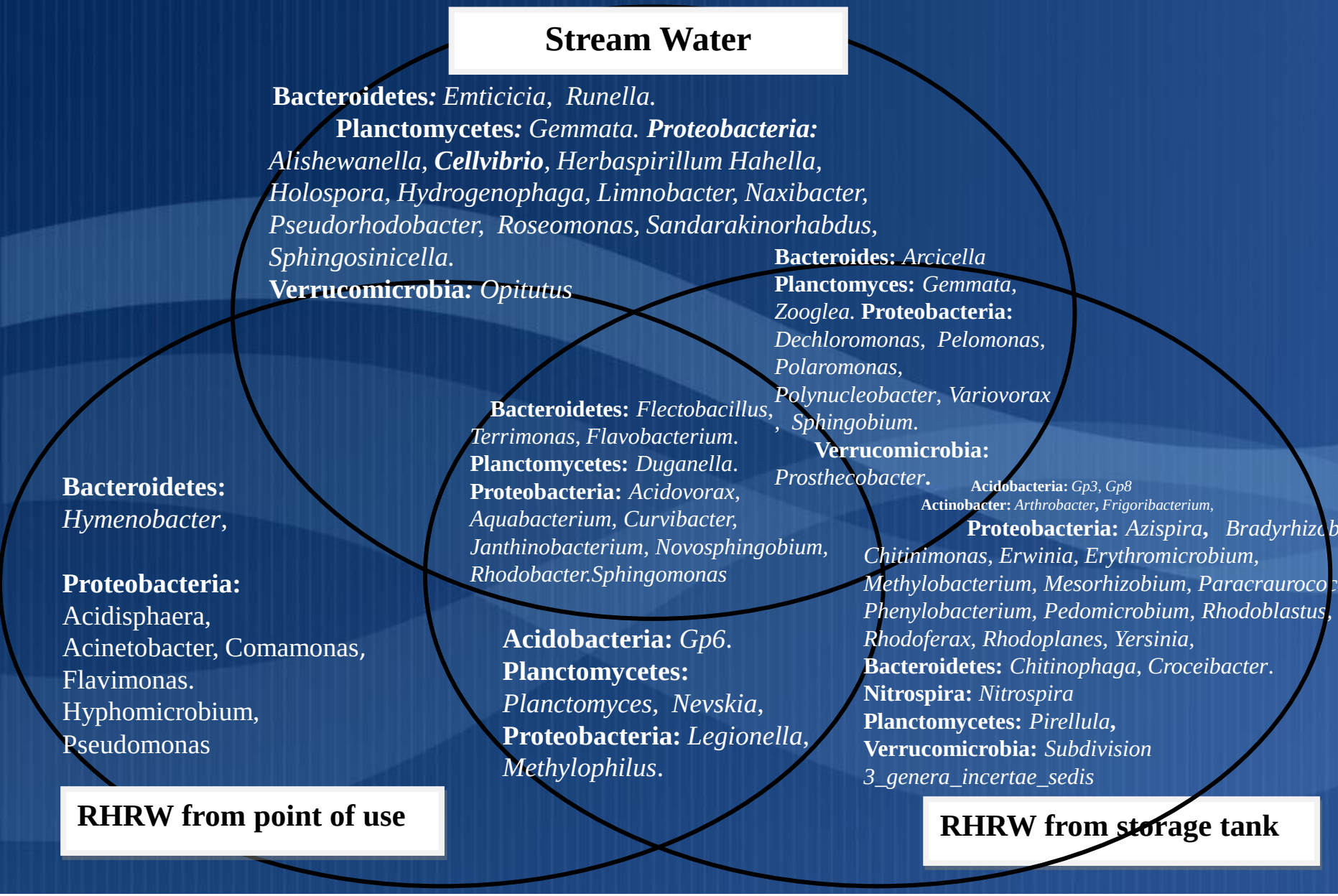
Pyrosequencing

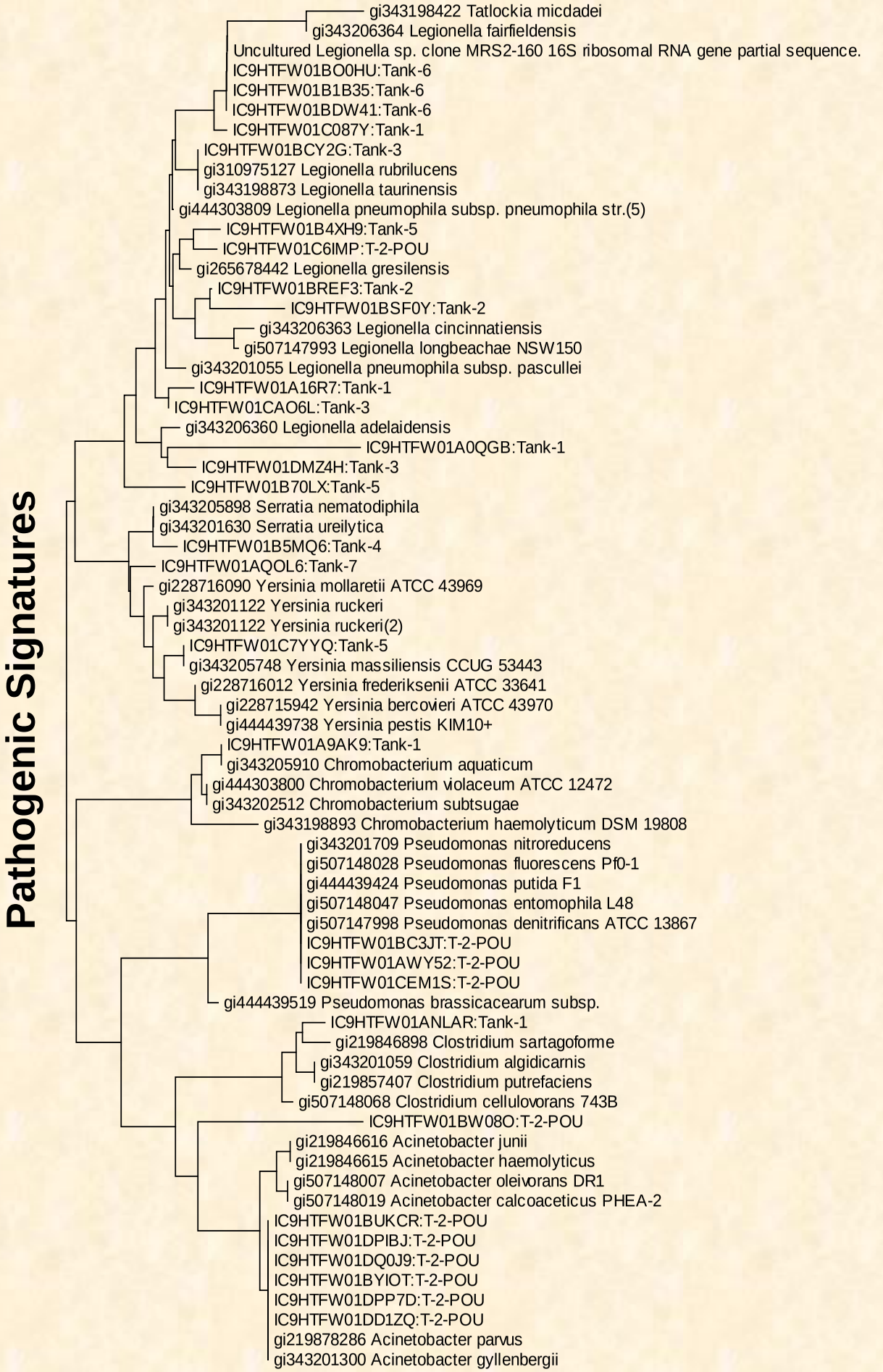


Average relative abundance (%) of the reads classified into the predominant microbial phyla by individual samples.

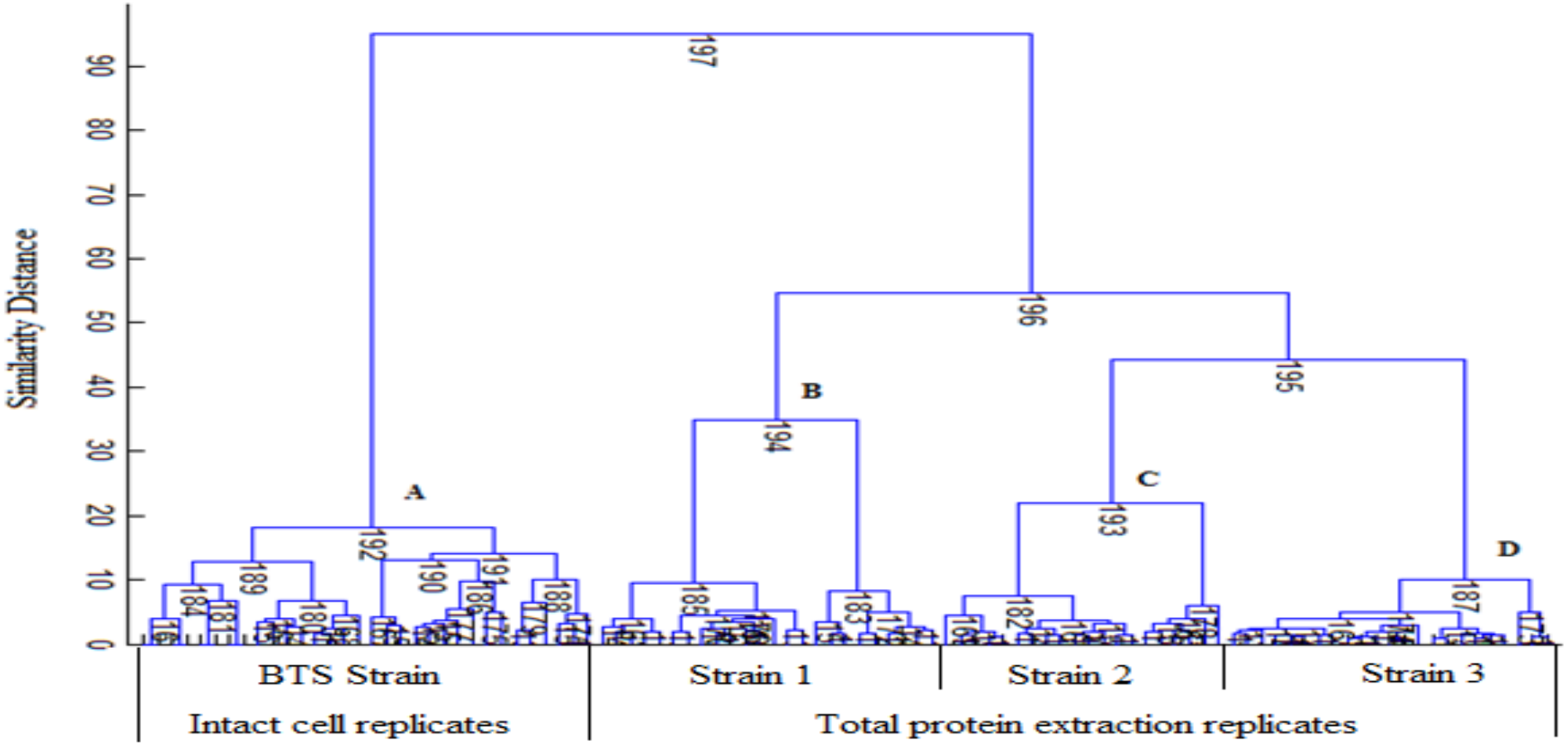


Bacterial communities of river water (A), kitchen water (B) and tank water (C).

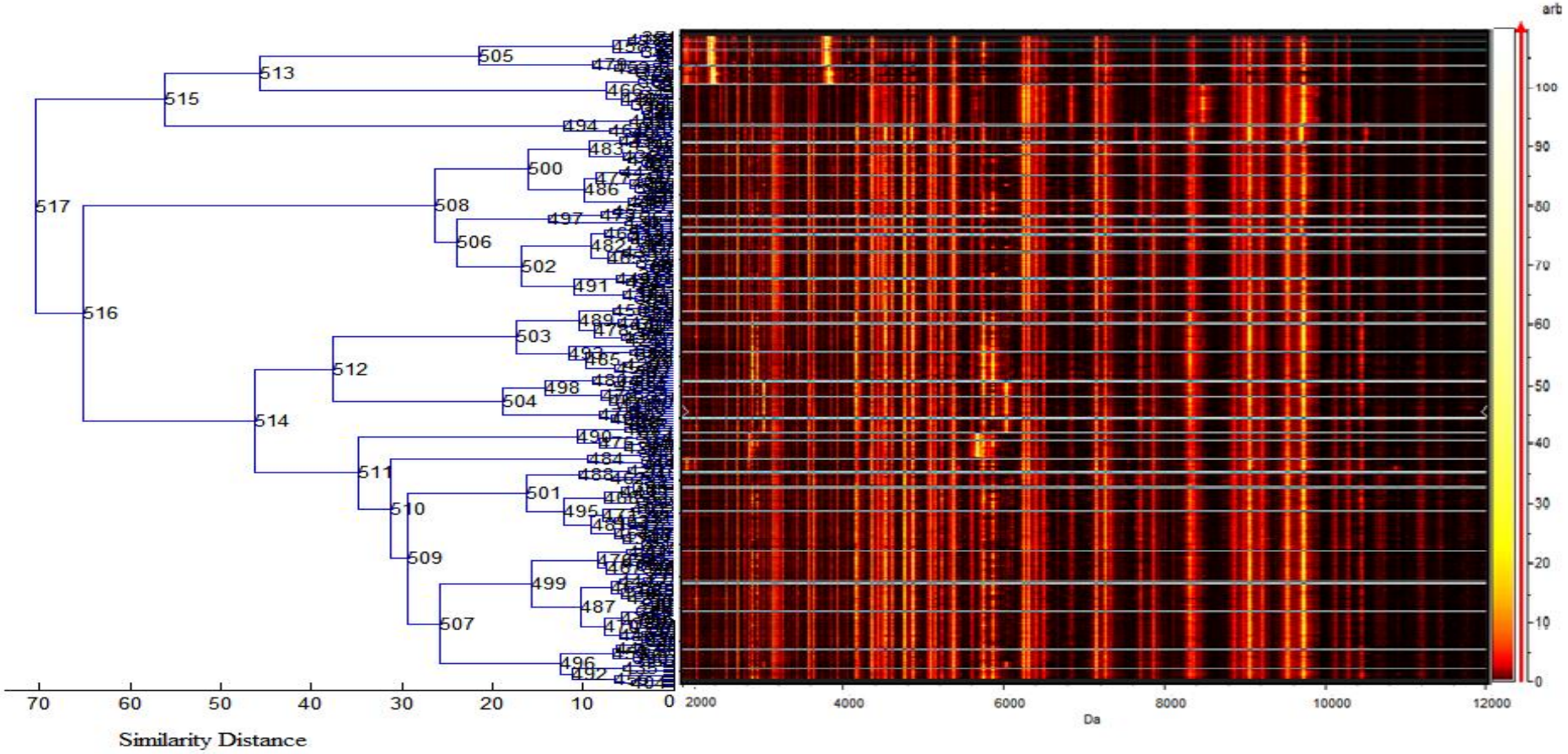

Stream Water
Bacteroidetes: Emticicia, Runella.
Planctomycetes: Gemmata. Proteobacteria: Alishewanella, Cellvibrio, Herbaspirillum Hahella, Holospora, Hydrogenophaga, Limnobacter, Naxibacter, Pseudorhodobacter, Roseomonas, Sandarakinorhabdus, Sphingosinicella.
Verrucomicrobia: Opitutus
Bacteroides: Arcicella
Planctomyces: Gemmata, Zooglea. Proteobacteria: Dechloromonas, Pelomonas, Polaromonas, Polynucleobacter, Variovorax, Sphingobium.
Verrucomicrobia: Prosthecobacter.
Bacteroidetes: Flectobacillus, Terrimonas, Flavobacterium.
Planctomycetes: Duganella.
Proteobacteria: Acidovorax, Aquabacterium, Curvibacter, Janthinobacterium, Novosphingobium, Rhodobacter.Sphingomonas
Bacteroidetes: Hymenobacter,
Proteobacteria: Acidisphaera, Acinetobacter, Comamonas, Flavimonas. Hyphomicrobium, Pseudomonas
Acidobacteria: Gp6.
Planctomycetes: Planctomyces, Nevskia,
Proteobacteria: Legionella, Methylophilus.
Acidobacteria: Gp3, Gp8
Actinobacter: Arthrobacter, Frigoribacterium,
Proteobacteria: Azispira, Bradyrhizobium, Chitinimonas, Erwinia, Erythromicrobium, Methylobacterium, Mesorhizobium, Paracraurococcus, Phenylobacterium, Pedomicrobium, Rhodoblastus, Rhodoferax, Rhodoplanes, Yersinia,
Bacteroidetes: Chitinophaga, Croceibacter.
Nitrospira: Nitrospira
Planctomycetes: Pirellula,
Verrucomicrobia: Subdivision 3_genera_incertae_sedis
RHRW from point of use
RHRW from storage tank



MALDI TOF MS: BACTERIAL CHARACTORISATION

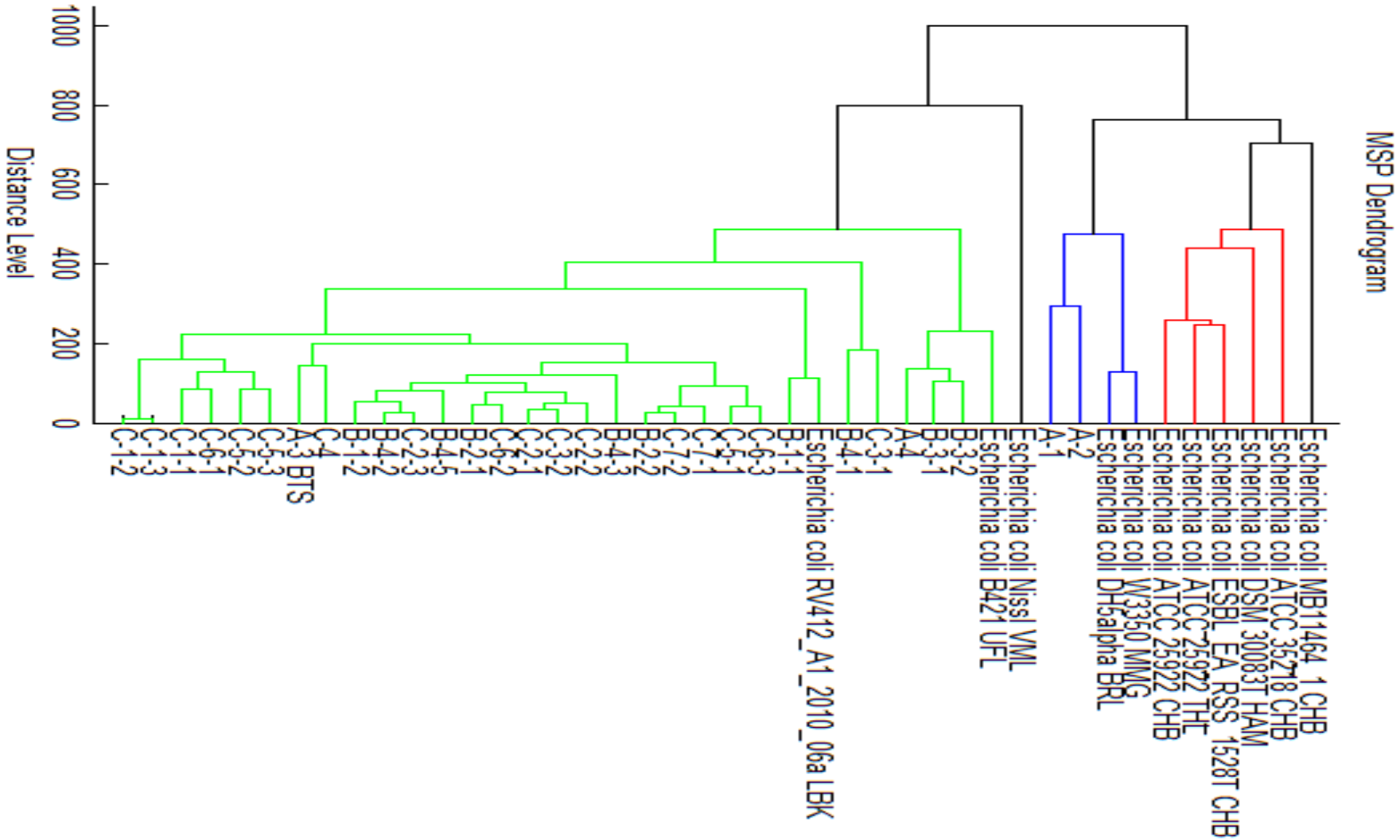


MALDI TOF MS spectra reproducibility showing (a) Ward Euclidian cluster dendrogram for 4 strain groups spectra obtained with intact bacteria (A) and total protein extraction (B, C and D).

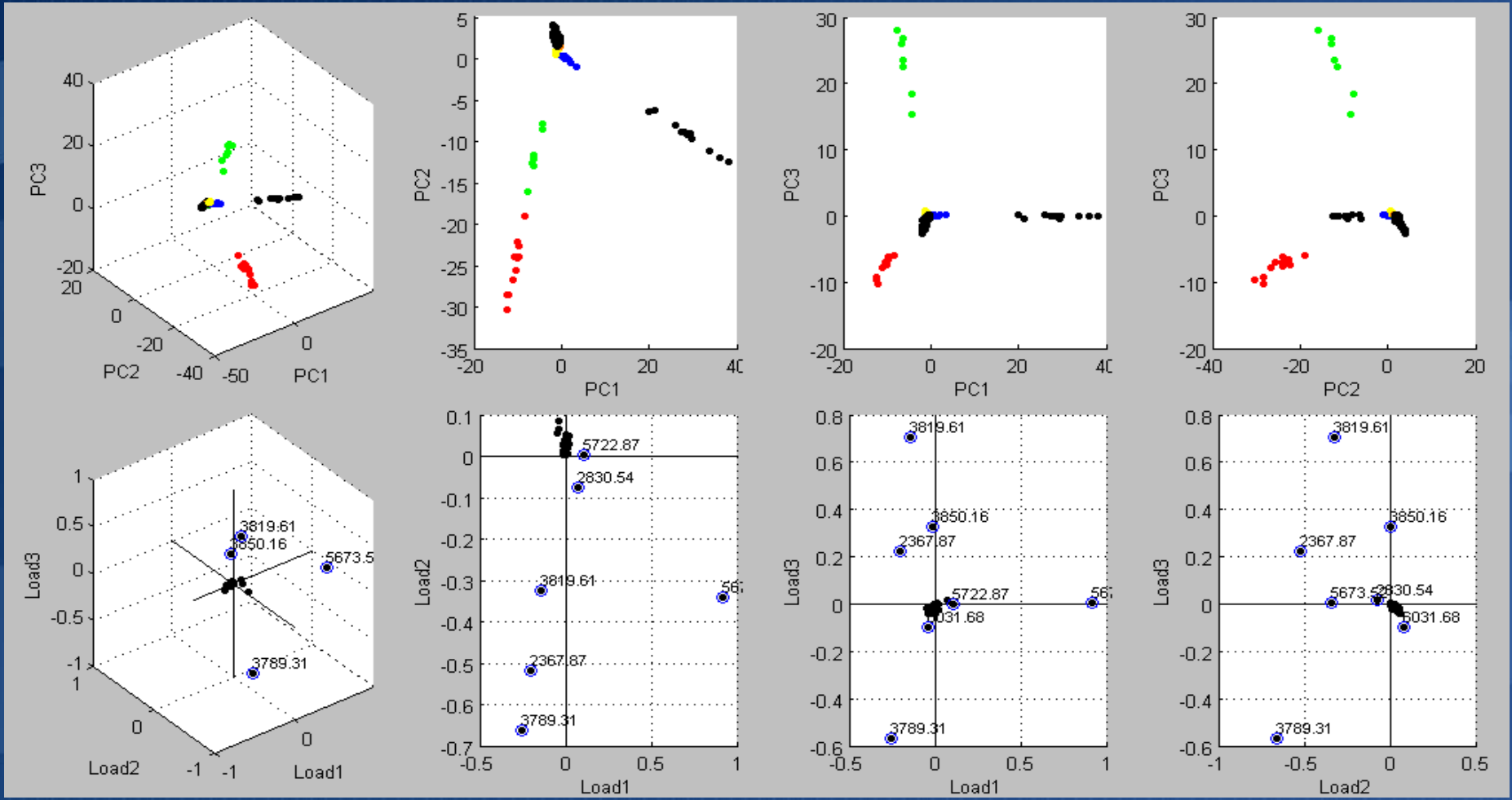


- Two-dimensional Unsupervised hierarchical cluster analysis of 238 bacterial isolates (506 spectra) versus 100 m/z values (rows) dendrogram and associated chromatic gell vie of the mass spectra profiles.
- Normalized expression value for each protein is indicated by a colour with arbitrary units represented by varying shades of brightness.
- Absolute intensities of the ions are shown on the key to the right and the masses (in Da) of the ions are shown on the x axis.

Strain group similarity evaluation



Principal Component Analysis

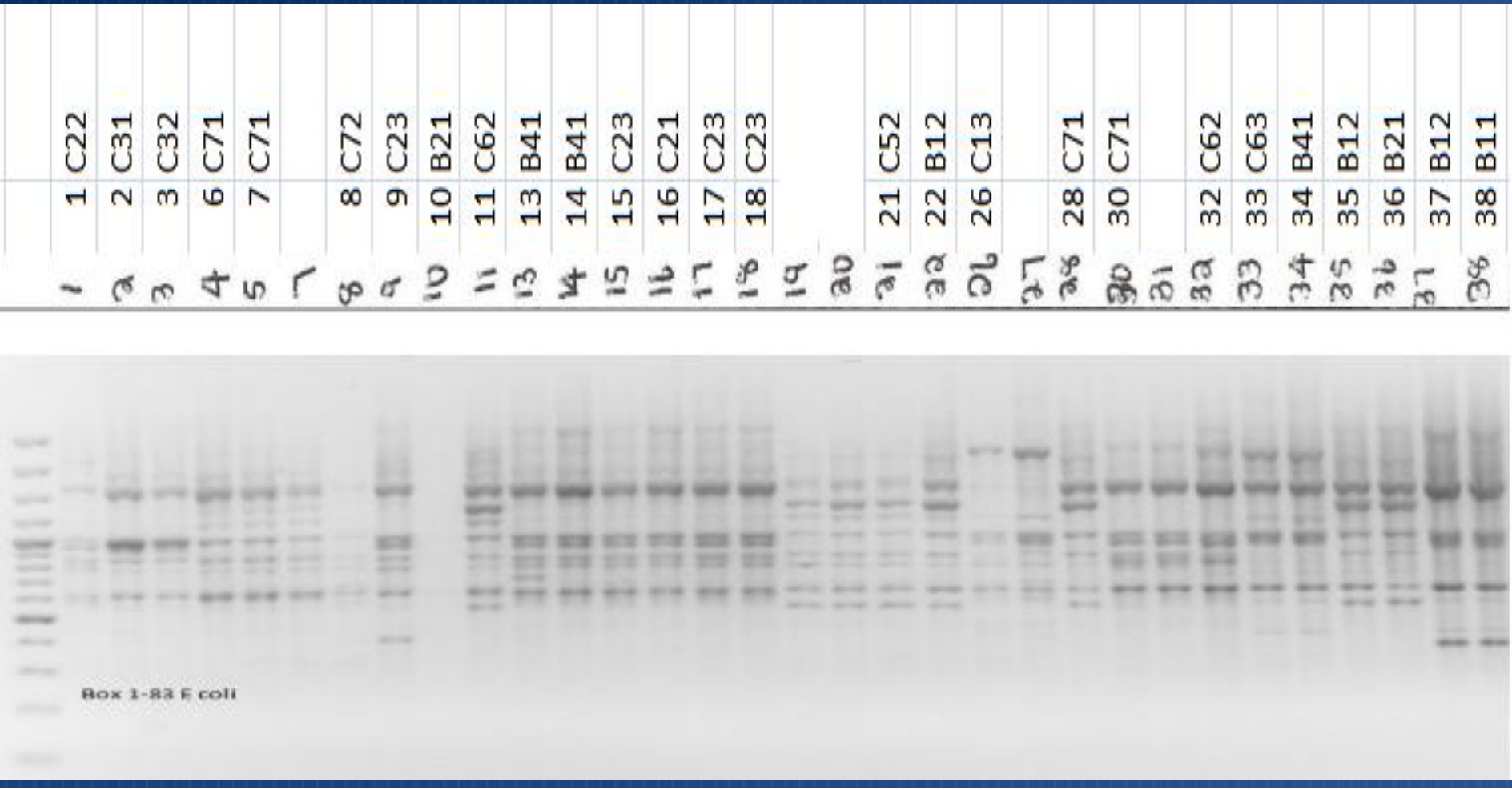


Results of a support vector machine (SVM) model on the binary peak data of 31 unique strain (cluster) groups.
Rows denote the observed strain groups and the columns the predicted strain groups.
Actual group membership is shown in the table. Identification % given at the bottom of the table.

	A-1	A-2	A-3	A-4	B-1-1	B-1-2	B-2-1	B-2-2	B-3-1	B-3-2	B-4-1	B-4-2	B-4-3	B-4-5	C-1-1	C-1-2	C-1-3	C-2-1	C-2-2	C-2-3	C-3-1	C-3-2	C-4	C-5-1	C-5-2	C-5-3	C-6-1	C-6-2	C-6-3	C-7-1	C-7-2
A-1	1	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
A-2	-	7	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
A-3	-	-	1	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
A-4	-	-	-	7	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
B-1-1	-	-	-	-	5	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
B-1-2	-	-	-	-	-	8	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
B-2-1	-	-	-	-	-	-	1	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
B-2-2	-	-	-	-	-	-	-	6	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
B-3-1	-	-	-	-	-	-	-	-	4	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
B-3-2	-	-	-	-	-	-	-	-	-	3	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
B-4-1	-	-	-	-	-	-	-	-	-	-	7	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
B-4-2	-	-	-	-	-	-	-	-	-	-	1	1	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
B-4-3	-	-	-	-	-	-	-	-	-	-	-	-	5	1	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
B-4-5	-	-	-	-	-	-	-	-	-	-	-	-	-	7	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
C-1-1	-	-	-	-	-	-	-	-	-	-	-	-	-	-	5	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-
C-1-2	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	9	-	-	-	-	-	-	-	-	-	-	1	-	-	-	-
C-1-3	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	1	-	-	-	-	-	-	-	-	-	-	-	-	-	-
C-2-1	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	6	-	-	-	-	-	-	-	-	-	-	-	-	-
C-2-2	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	1	-	1	6	-	-	-	-	-	-	-	-	-	-	-	-
C-2-3	-	-	-	-	-	-	-	-	-	-	-	-	-	1	-	-	-	-	-	5	-	-	-	-	-	-	-	-	-	-	-
C-3-1	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	3	1	-	-	-	-	-	-	-	-	-
C-3-2	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	7	-	-	-	-	-	-	-	-	-
C-4	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	5	-	-	-	-	-	-	-	-
C-5-1	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	6	-	-	-	-	-	-	-
C-5-2	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	9	-	-	-	-	-	-
C-5-3	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	1	-	-	-	-	-
C-6-1	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	1	-	-	-	-
C-6-2	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	1	-	-	-
C-6-3	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	1	-	-
C-7-1	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	8	-
C-7-2	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	5

Class. accuracy (%) = 100.000(Train), 89.06(Test), 97.23(Overall)
Cross-validation accuracy (%) = 86.77

MALDI TOF MS Comparison with REP PCR



Bacterial Source Tracking

	A-1	A-2	A-3	A-4	B-1-1	B-1-2	B-2-1	B-2-2	B-3-1	B-3-2	B-4-1	B-4-2	B-4-3	B-4-5	C-1-1	C-1-2	C-1-3	C-2-1	C-2-2	C-2-3	C-3-1	C-3-2	C-4	C-5-1	C-5-2	C-5-3	C-6-1	C-6-2	C-6-3	C-7-1	C-7-2	Total
PTA-1	-	-		-	1	1	1	-	-	1	-	1	1	2	-	1	-	-	3	-	-	-	-	1	1	2	1	-	-	-	-	17
PTA-2	-	-		-	2	-	-	1	-	1	2	3	-	-	-	2	3	-	-	3	-	2	-	-	-	-	-	1	3	2	2	27
PTA-3	-	-		9	-	1	4	2	4	-	1	3	-	-	-	2	2	2	1	-	-	2	-	5	3	10	4	2	2	-	3	62
JHB-1	-	-		-	1	1	1	-	-	-	-	-	2	1	1	3	2	-	2	-	1	2	-	1	2	3	-	4	2	-	-	29
JHB-2	-	-		-	1	-	-	1	-	1	1	1	1	2	1	-	-	1	3	-	-	-	-	-	-	3	-	1	-	1	2	20
Water total	**-**	**-**	**-**	**9**	**5**	**3**	**6**	**4**	**4**	**3**	**4**	**8**	**4**	**5**	**2**	**8**	**7**	**3**	**9**	**3**	**1**	**6**	**-**	**7**	**6**	**18**	**5**	**8**	**7**	**3**	**7**	**155**
Bird	-	-	-	-	4	10	8	5	-	-	9	5	4	8	4	9	5	3	-	4	3	5	-	2	5	9	10	5	9	8	2	136
Sewage	-	-	-	-	-	6	-	-	-	-	6	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	12
human	-	-	-	-	-	-	-	-	-	-	-	-	-	-	2	2	-	-	-	-	-	-	46	3	-	-	-	-	-	-	-	53
Control Strain	-	-	16	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	16
Milk	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	4	-	-	-	-	-	-	-	1	-	-	-	-	-	-	5
River	11	7	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	18
Total	**11**	**7**	**16**	**9**	**9**	**19**	**14**	**9**	**4**	**3**	**19**	**13**	**8**	**13**	**8**	**19**	**16**	**6**	**9**	**7**	**4**	**11**	**46**	**12**	**12**	**27**	**15**	**13**	**16**	**11**	**9**	**395**

Acknowledgents

WRC