

# AQUIFER COMMUNITIES – CAN THEY PREDICT CHANGES IN AOTEAROA’S SOURCE (GROUND)WATER

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# INTRODUCTION – ‘THE WATER PLANET’

- ❖ 71 % of the earth's surface is covered with water
- ❖ 3 % of the earth's water is freshwater
- ❖ 2.5 % of the earth's freshwater is unavailable: locked up in glaciers, polar ice caps, atmosphere, soil, and deep inaccessible aquifers
- ❖ 0.5 % is available freshwater: includes groundwater



Table1. Percentages of water on earth

	%
Oceans	97.2
Ice Caps/Glaciers	2.0
Groundwater	0.62
Freshwater Lakes	0.009
Inland seas/salt lakes	0.008
Atmosphere	0.001
Rivers	0.0001
<b>TOTAL</b>	<b>99.8381</b>

- ❖ Global popn. increasing: 1950 (2.5 billion) → 2023 (< 8 billion)
- ❖ Land use practices: industrial applications, urban development and farming
- ❖ Release pollutants that infiltrate groundwater aquifers, threatening drinking water supplies
  - Metals (arsenic, manganese)
  - Petroleum products
  - Plastics
  - Pathogenic bacteria
  - Nutrients (nitrogen and phosphorus)



<https://wrytin.com/kashish4/global-population-jxniqgzz>

## Q. How are pollutants removed from groundwater?

- ❖ Aquifers naturally filter groundwater by forcing it to pass through small pores and between sediment particles
- ❖ Predatory organisms i.e., stygofauna, protozoa, bacteriophages feed on pathogenic bacteria and biofilms
- ❖ Biological (bacterial) communities utilise contaminants as energy (carbon) sources

# BIOLOGICAL – BACTERIAL COMMUNITIES

- ❖ Bacterial communities can adapt to subtle changes in the chemical composition of a groundwater system
- ❖ Changes in bacterial communities could potentially be used as an early warning of a change in water quality i.e., nitrate contamination and changes in ecosystem functioning

# INTERNATIONAL RESEARCH

## *Links found between bacterial communities and water quality*

- ❖ Overseas research has found that increased organic contamination has a negative effect on bacterial diversity
- ❖ Bacterial community compositions in polluted environments shift towards less diverse but more specialised communities
- ❖ Bacterial populations affiliated with Proteobacteria, Actinobacteria and Acidobacteria are dominant in environments polluted with organic compounds and pesticides

BALABAN, N., YANKELZON, I., ADAR, E., GELMAN, F., RONEN, Z. & BERNSTEIN, A. 2019. The spatial distribution of the microbial community in a contaminated aquitard below an industrial zone. *Water*, 11, 2128.

HOLMSGAARD, P. N., DEALTRY, S., DUNON, V., HEUER, H., HANSEN, L. H., SPRINGAEL, D., SMALLA, K., RIBER, L. & SØRENSEN, S. J. 2017. Response of the bacterial community in an on-farm biopurification system, to which diverse pesticides are introduced over an agricultural season. *Environmental Pollution*, 229, 854-862.

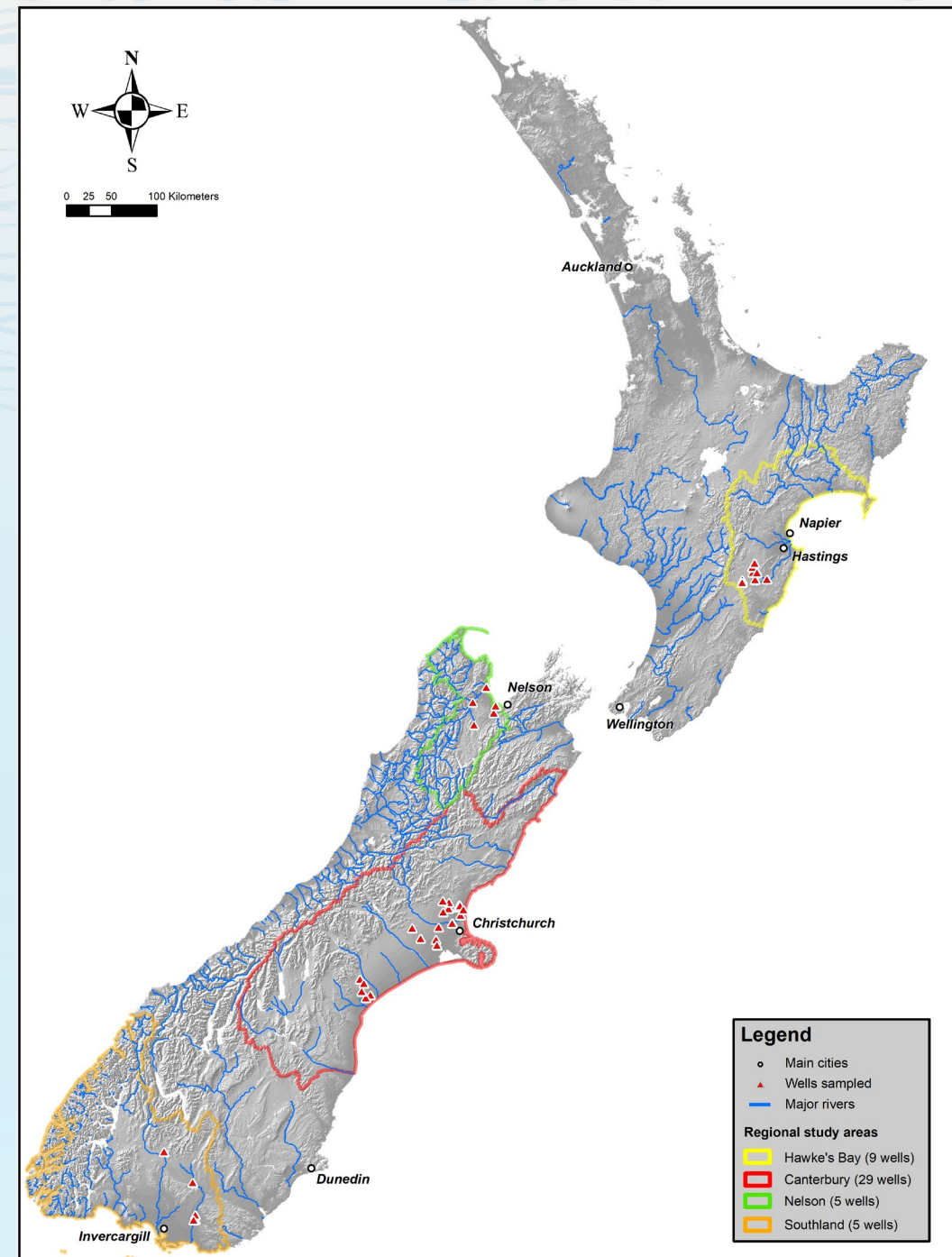
# STUDY QUESTIONS

- ❖ Can differences in bacterial communities be used to help predict changes in water quality
- ❖ Does the composition of a bacterial community change depending on aquifer geology (location)

# WHAT WE DID

## *Sampled:*

- ❖ 49 wells (shallow aquifers) in four regions
- ❖ Well parameters: pH, temp, dissolved oxygen, nitrate, total nitrogen etc.
- ❖ Extracted DNA: sequenced bacteria





# ANALYSIS

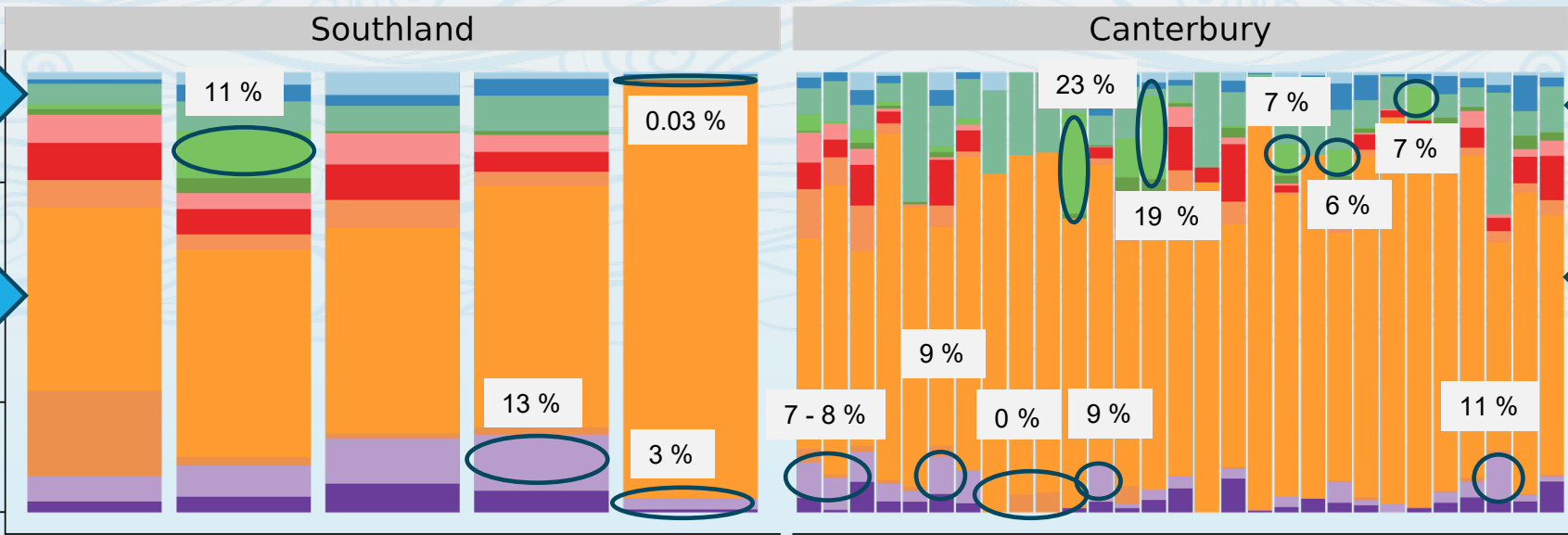
- ❖ Calc. bacterial abund. (different species within a community)
- ❖ Calc. diversity
  - ❖ Alpha - How many different bacterial species are detected within each sample
  - ❖ Beta - The difference in bacterial composition between samples (regions)
- ❖ Evaluated correlations: between bacterial abund. and well parameters
- ❖ Machine learning to assess bacterial abund. and nitrate classification

❖ **Bacteroidetes** 12.42%

• Thaumarchaeota oxidise ammonia to nitrite (1<sup>st</sup> step in nitrification)

• Bacteroidetes is a phenotypically diverse group

• Within this group are spp. capable of denitrification, nitrate reduction and degradation of organic compounds such as diesel

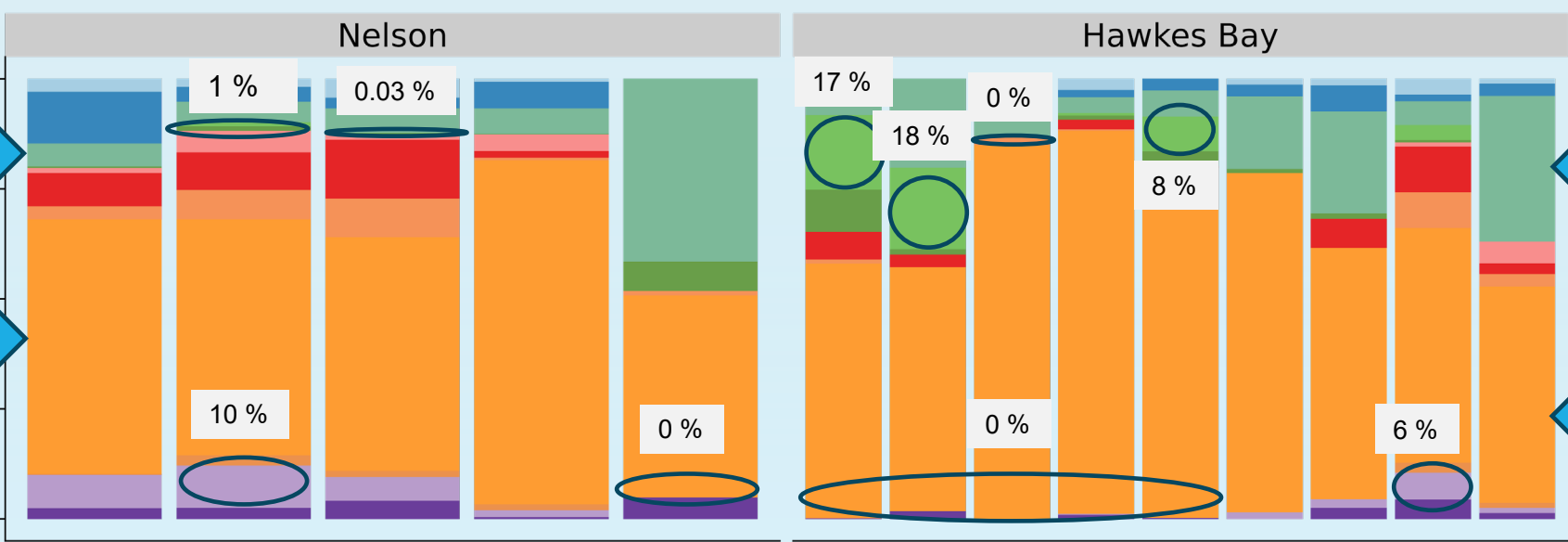


← Bacteroidetes

← Proteobacteria

• Proteobacteria remove organic pollutants (pesticides) and are involved in denitrification

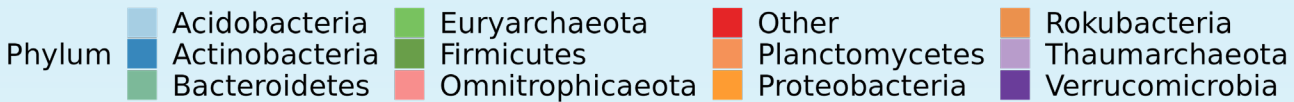
• Phylum includes multiple spp, including pathogenic bacteria and *E. coli* (an indicator of faecal contamination)



→ Bacteroidetes

→ Proteobacteria

← Proteobacteria



# SOUTHLAND WELLS (EXAMPLE)

Pristine

Contaminated

- Located in upper catchment
- 10 km upgradient is a pristine source water

- Located down gradient of three dairy farms
- Dairy wastewater is applied to land via irrigation
- Impacts shallow aquifers

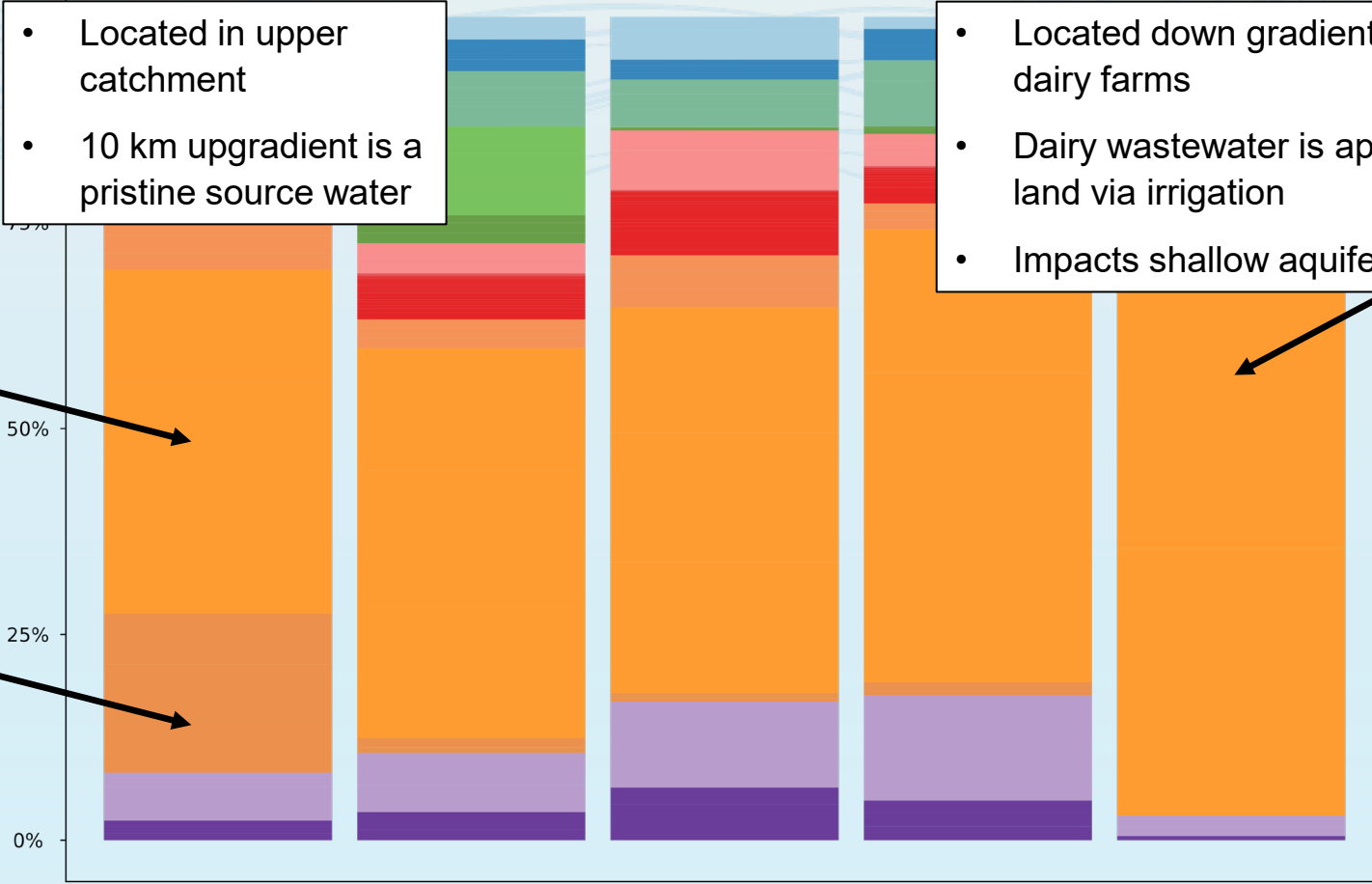
Proteobacteria 42 %

Rokubacteria 19 %

- Large genomes, adapted to survive in low nutrient envts

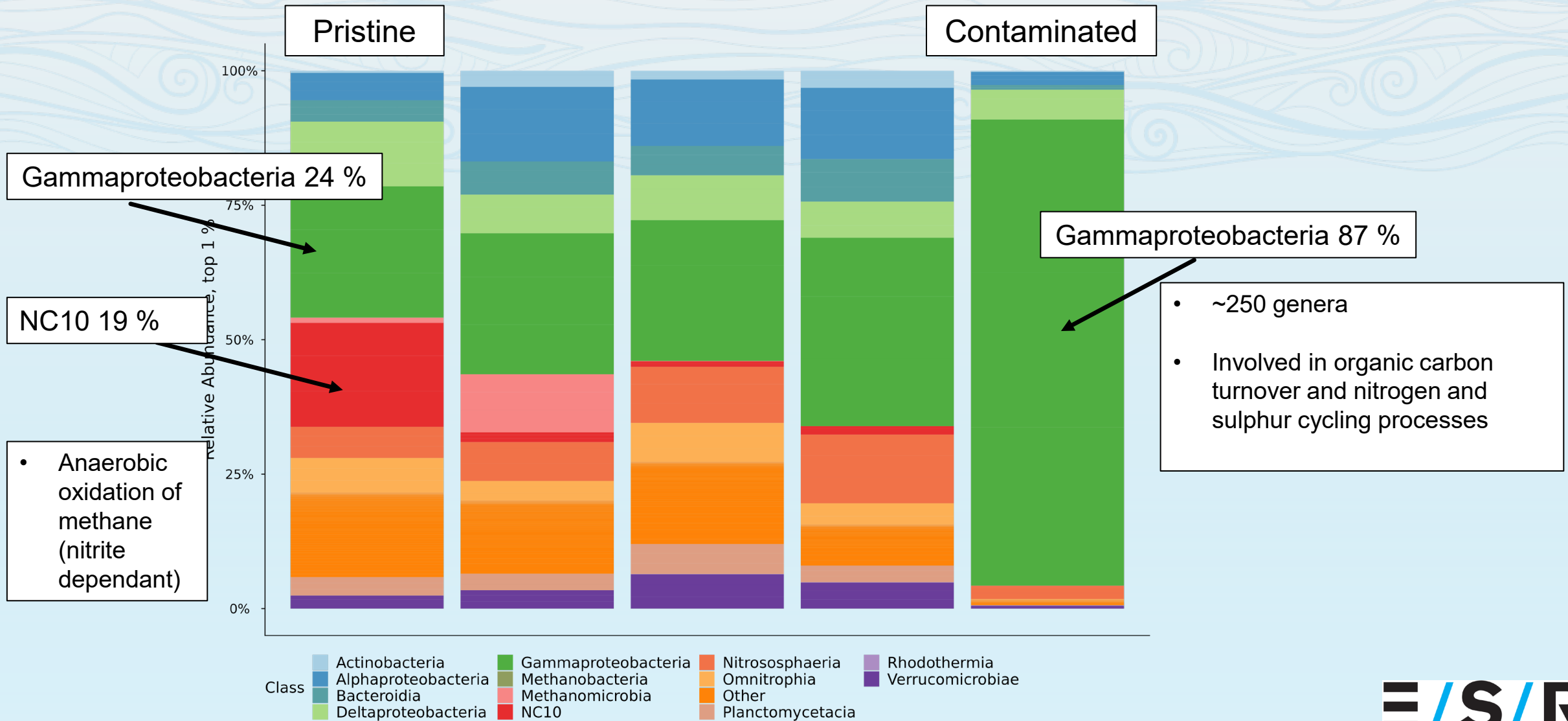
Proteobacteria 95 %

Relative Abundance, top 1 %



Phylum  
 Acidobacteria, Actinobacteria, Bacteroidetes, Euryarchaeota, Firmicutes, Omnitrophicaeota, Other, Planctomycetes, Proteobacteria, Rokubacteria, Thaumarchaeota, Verrucomicrobia

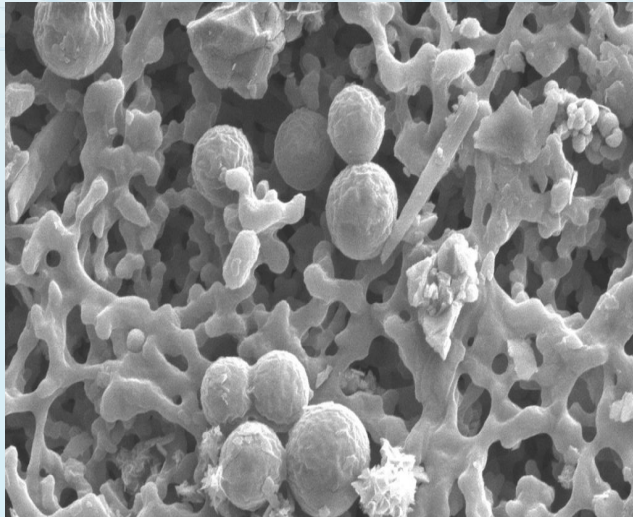
# SOUTHLAND WELLS (EXAMPLE)



# AQUIFER BACTERIA

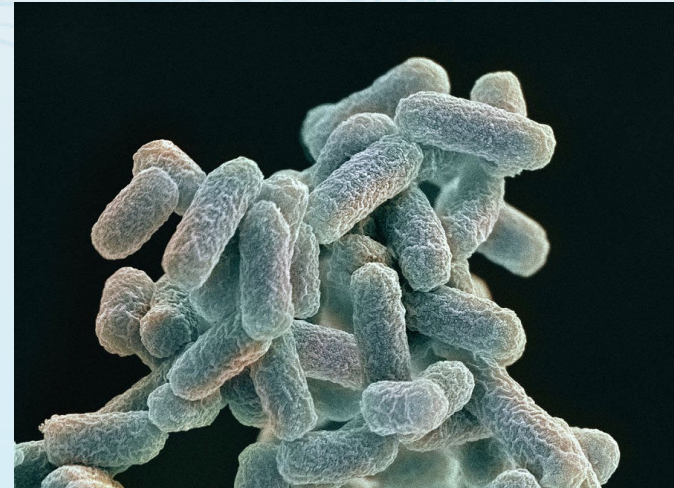
*Pseudomonas denitrificans*

## *Aquifer biofilm*



<https://www.auckland.ac.nz/en/news/2020/04/21/secret-life-of-our-aquifers.html>

## *E. coli (Proteobacteria)*



<https://pixels.com/> (Steve Gschmeissner)

~2.0 x 0.5 microns

Indicator of faecal contamination

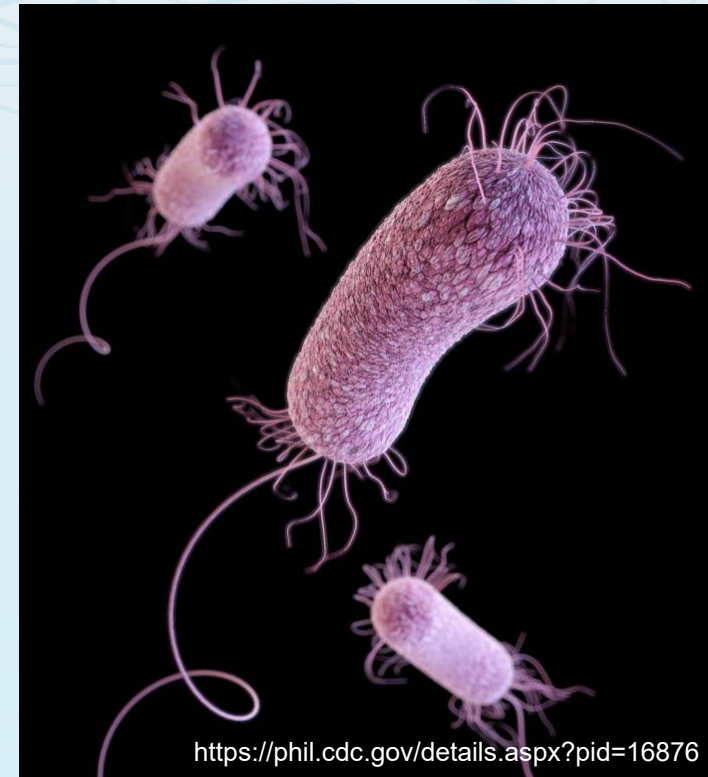
# PROTEOBACTERIA

*Pseudomonas denitrificans*



~ 1.05 x 0.8 microns

*Pseudomonas aeruginosa*

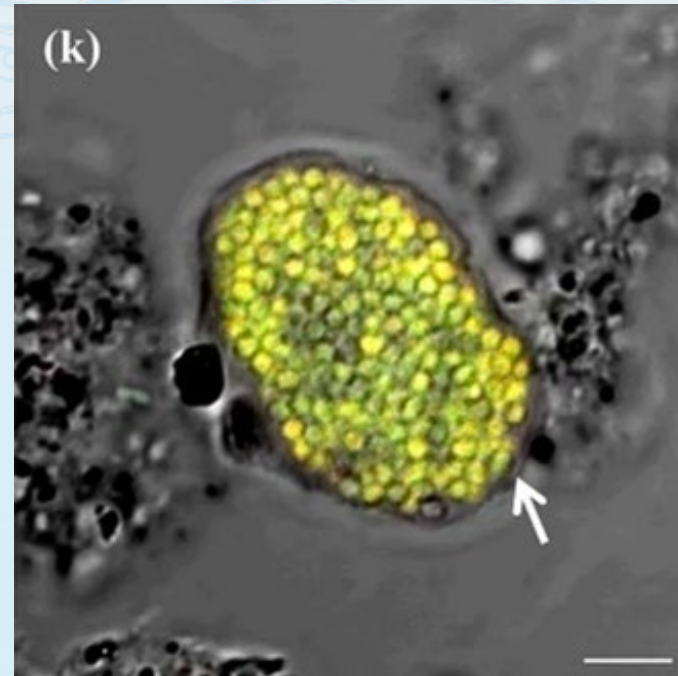


1.5-3 x 0.5-0.8 microns

Denitrification

# ROKUBACTERIA

NC10



*10 - 30 microns, round or oval, honeycomb-shaped microcolonies*

Denitrifying methanotroph

# ALPHA DIVERSITY - SOUTHLAND

Less contamination = more bacterial diversity

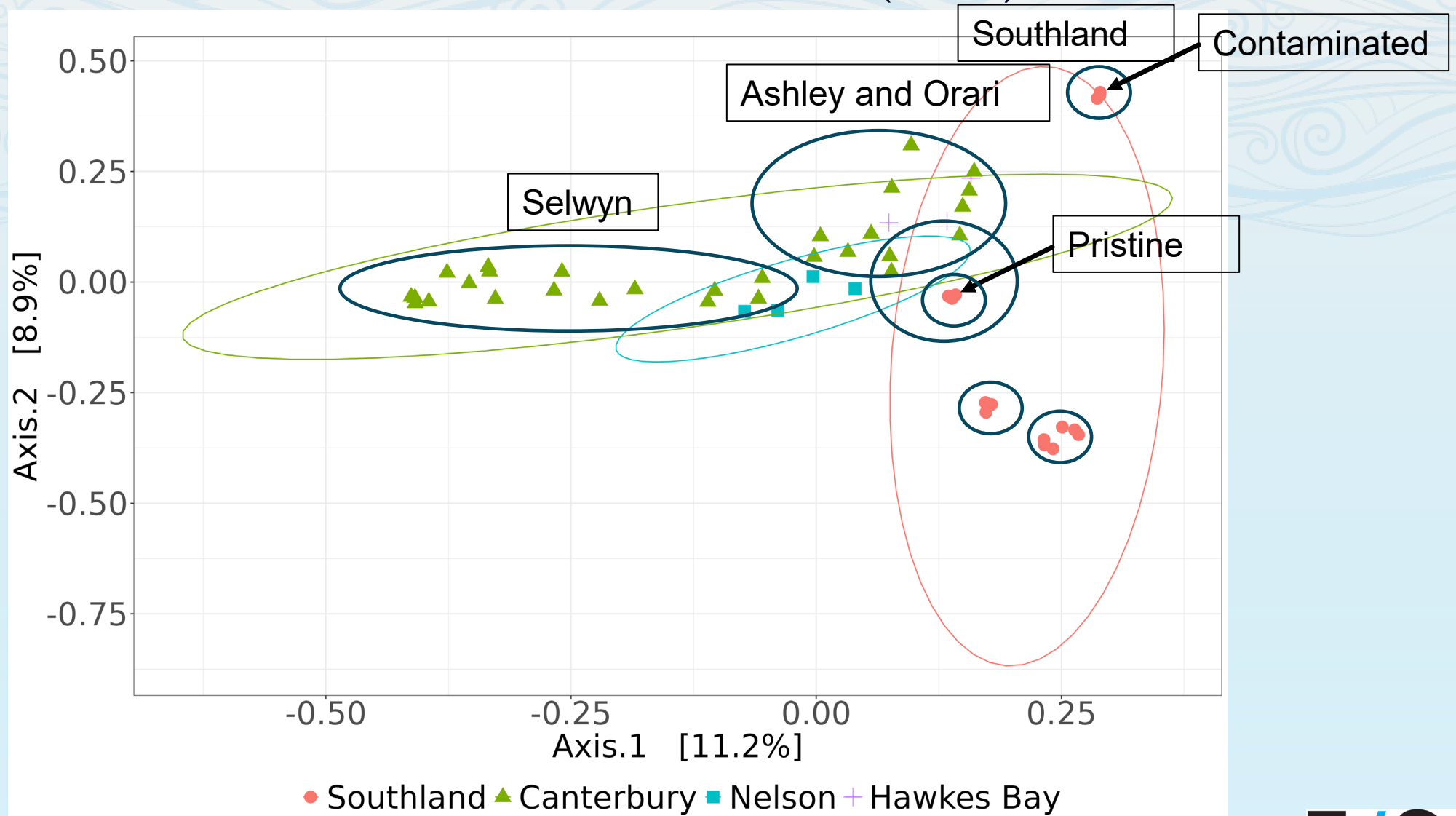
Higher contamination = less bacterial diversity

	Shannon Index (spp. diversity)
Pristine	5.6
Contaminated	1.8

	Observed Index (# of spp.)
Pristine	530
Contaminated	55



# BETA DIVERSITY - PRINCIPAL COORDINATE ANALYSIS (PCOA)



- Axes – show the percent of total variation explained by each axis
- The closer the two sample points are, the more similar their bacterial composition

## DIFFERENCES - *Southland*

- ❖ Cooler climate
- ❖ Aquifers have a unique geology, being thin, long and shallow with alluvial deposits
- ❖ The alluvial deposits - underlain with Gore lignite sedimentary rock
  - ❖ Formed from naturally acidic compressed peat
- ❖ Average thickness of alluvial deposit ~10 – 15 m
- ❖ In contrast: Canterbury Plains
  - ❖ Thickness of alluvial gravel commonly exceeds 100 m

## DIFFERENCES - *Southland*

Parameter	Unit	Southland	Other regions (range)
*pH		5.8	6.6 - 7.0
*Temperature	°C	10.4	13.1 - 13.9
DOC	mg/L	0.6	1.7 - 2.2
DO	mg/L	6.3	1.4 - 5.7
Specific conductance	uS/cm	340	149 - 175
Nitrate	mg/L	5.5	1.1 - 2.5
Total nitrogen	mg/L	5.7	1.1 - 2.6
* p-value < 0.002			

Median

Lower

Higher

## CORRELATIONS

- ❖ Significant positive correlations were found between several bacterial spp. and pH
- ❖ Moderate correlations were found between bacterial spp. and total nitrogen, organic reactive phosphorus and nitrate

# MACHINE LEARNING – RANDOM FOREST

- ❖ Used a small subset of groundwater data
- ❖ Assessed bacterial abund. and nitrate classification using Random Forest

NZ Drinking Water Std	Nitrate-N (mg/L)	Category
WHO MAV	0 – 1	Pristine
	1 – 11.3	Below MAV
	< 11.3	Above MAV

Correctly predicted: **90 %** of nitrate classifications using Rel. Abund. of Thaumarecaeota and Proteobacteria

# CONCLUSIONS

## Q. Can differences in bacterial communities be used to help predict changes in water quality

- ❖ Contaminated wells had more Proteobacteria than pristine wells and less bacterial diversity
- ❖ Correlations were identified between bacteria and pH, total nitrogen, organic reactive phosphorus and nitrate
- ❖ Random Forest: correctly predicted 90 % of nitrate classifications using abundances of Thaumarecaeota and Proteobacteria

# CONCLUSIONS

## Q. Does the composition of a bacterial community change depending on the well geology (location)

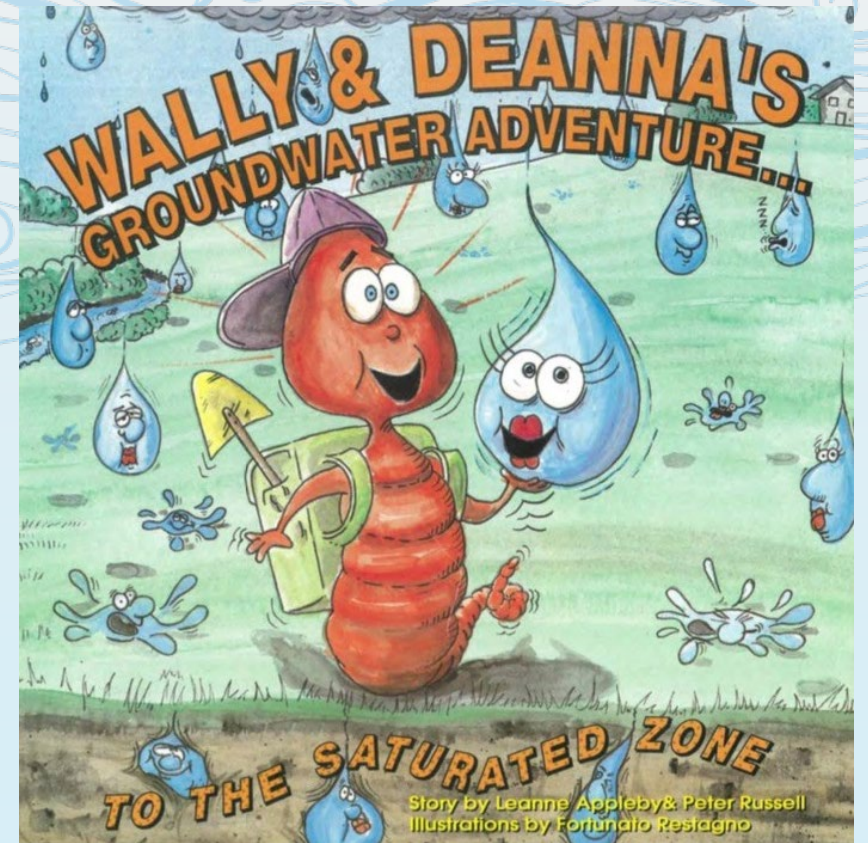
- ❖ The Southland region had a different geology and a different bacterial composition (community) compared to the other regions

## OVERALL CONCLUSION:

- ❖ While differences in the bacterial community were not apparent from abundance data (at level of phyla or genera)
  - Diversity data indicated differences in bacterial composition based on geology
  - Correlation and machine learning pipelines showed links between bacterial taxa and groundwater parameters, including nitrate

# ACKNOWLEDGEMENTS

- ❖ Funding: ESR SSIF funding
- ❖ Thanks for listening!



<https://gw-project.org/groundwater-education-for-children/>