

Microbial contamination of water and sediments

Faecal Source Tracking in the Avon / Ōtākaro River

**Institute of Environmental Science and Research Ltd.
ESR**

Specialist Science Solutions
Manaaki Tangata Taiao Hoki
protecting people and their environment through science

Acknowledgements

ESR

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Margaret MacKenzie,
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Ellen Ashmore, Andrew
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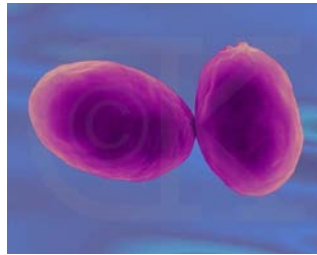
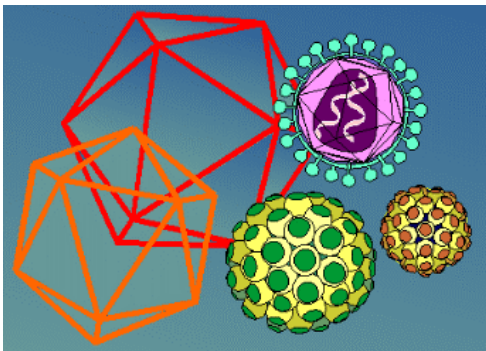


FUNDERS

- **Environment Canterbury**
- **Canterbury District Health Board**
- **Christchurch City Council**
- **Ministry of Health**
- **Ministry for the Environment**
- **FRST**
- **ESR**

Viruses

- Enteroviruses
 - Polioviruses, Echoviruses
 - Coxsackieviruses A & B
- Hepatitis A & E Viruses
 - Adenoviruses
 - Reoviruses
 - Rotaviruses
 - Caliciviruses
 - Astroviruses
- Picornavirus/parvoviruses
 - Coronaviruses



Bacteria

- Salmonellae
- Shigella spp.
- Campylobacter spp.
- Yersinia enterocolytica
- Escherichia coli
- Vibrio cholerae
- Aeromonas spp.
- Helicobacter pylori

Protozoa

- Giardia intestinalis
- Cryptosporidium parvum
- Entamoeba histolytica
 - Balantidium coli
- Cyclospora cayatenensis
- Toxoplasma gondii
 - Microsporidia

Worms

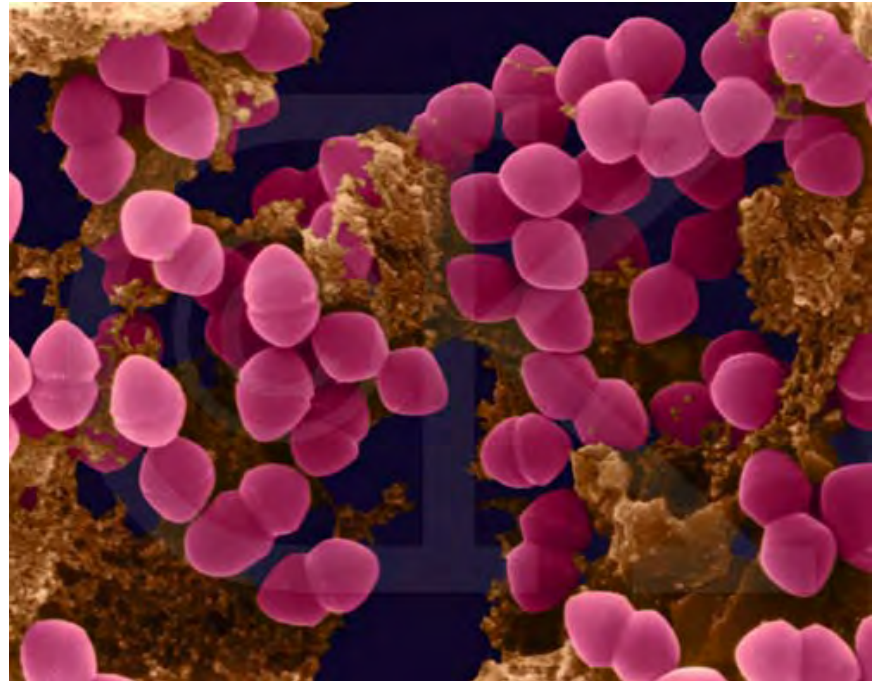
- Roundworms (Nematodes):
 - Ascaris lumbricoides
 - Trichuris trichuria
 - Hookworms:
 - Ancylostoma duodenale
 - Necator americanus
 - Strongyloides stercoralis
- Cestodes (tapeworms):
 - Hymenolepis nana



Faecal indicators



E. coli



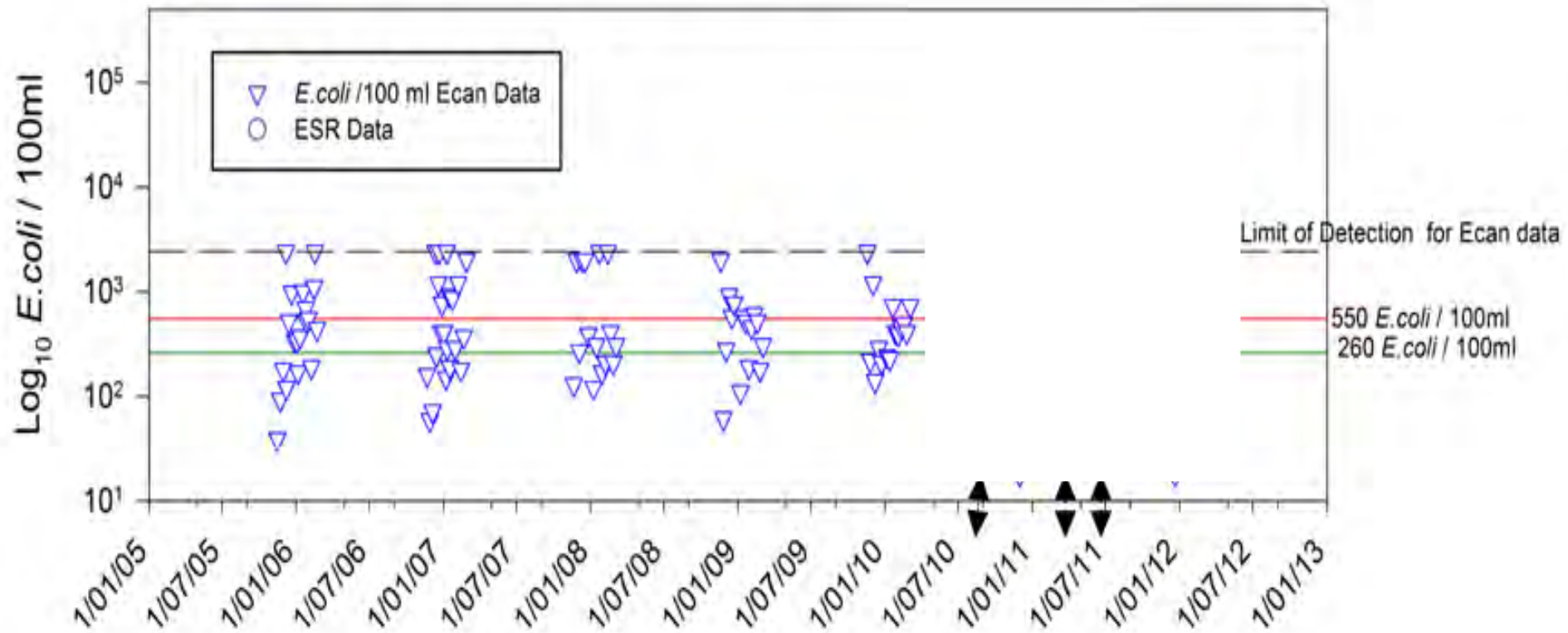
Enterococci

NZ Recreational water guidelines

- **Acceptable/Green Mode:** No single sample greater than 260 *E. coli*/100 mL.
 - Continue routine (e.g. weekly) monitoring.
- **Alert/Amber Mode:** Single sample greater than 260 *E. coli*/100 mL.
 - Increase sampling to daily.
 - Undertake a sanitary survey, and report on sources of contamination.
- **Action/Red Mode:** Single sample greater than 550 *E. coli*/100 mL.
 - Increase sampling to daily
 - Undertake a sanitary survey, and report on sources of contamination.
 - Erect warning signs.
 - Inform public through the media that a public health problem exists.

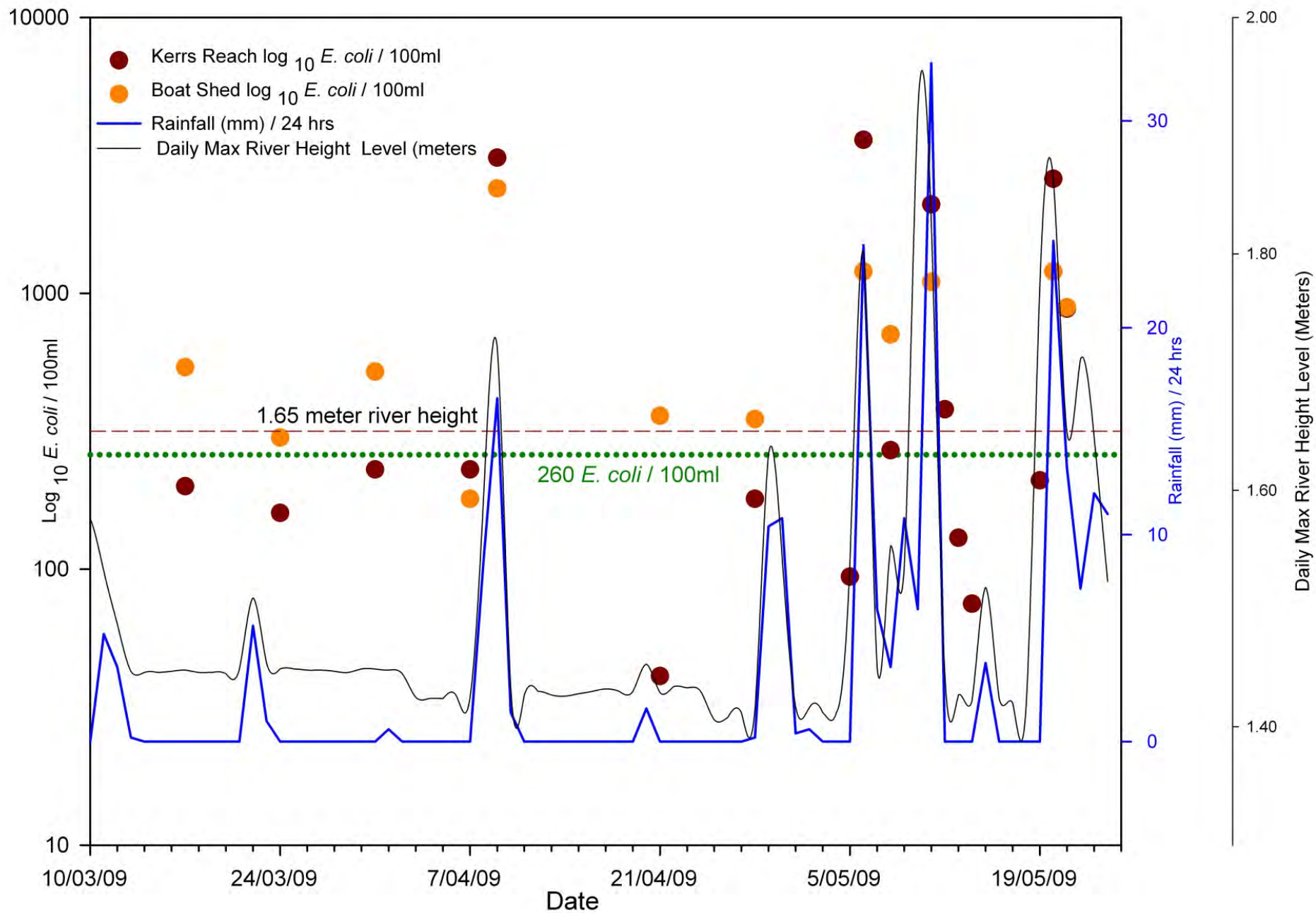
Avon River *E. coli* levels

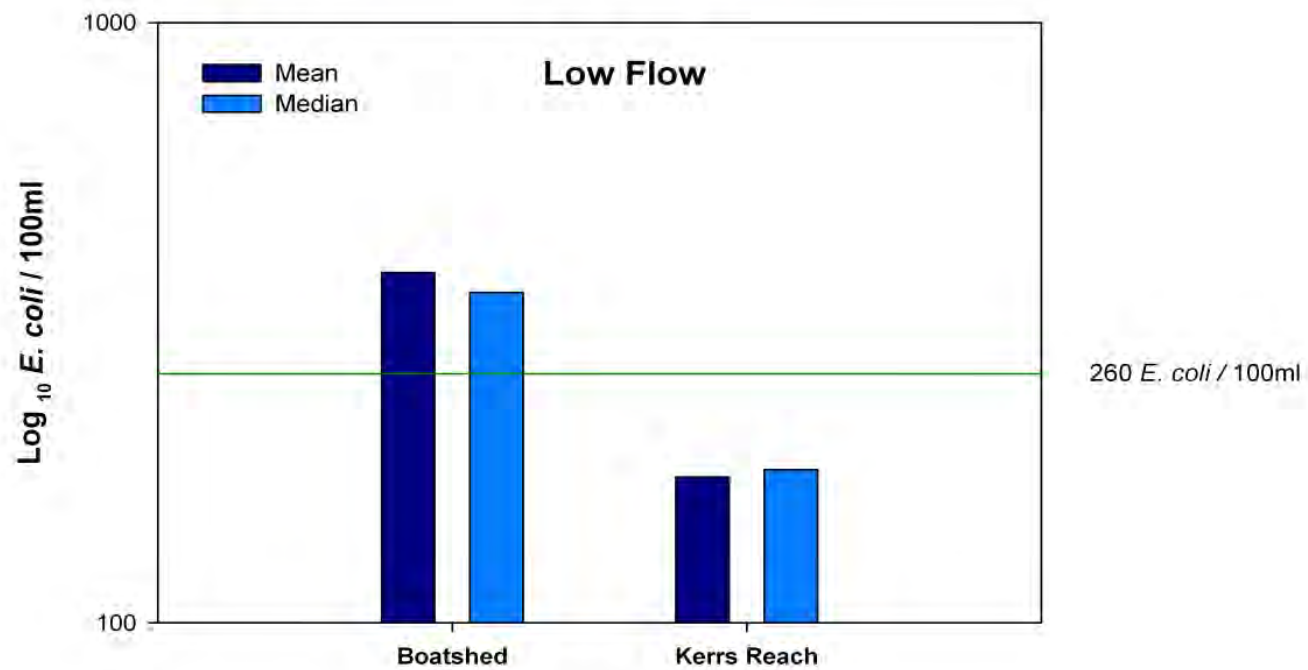
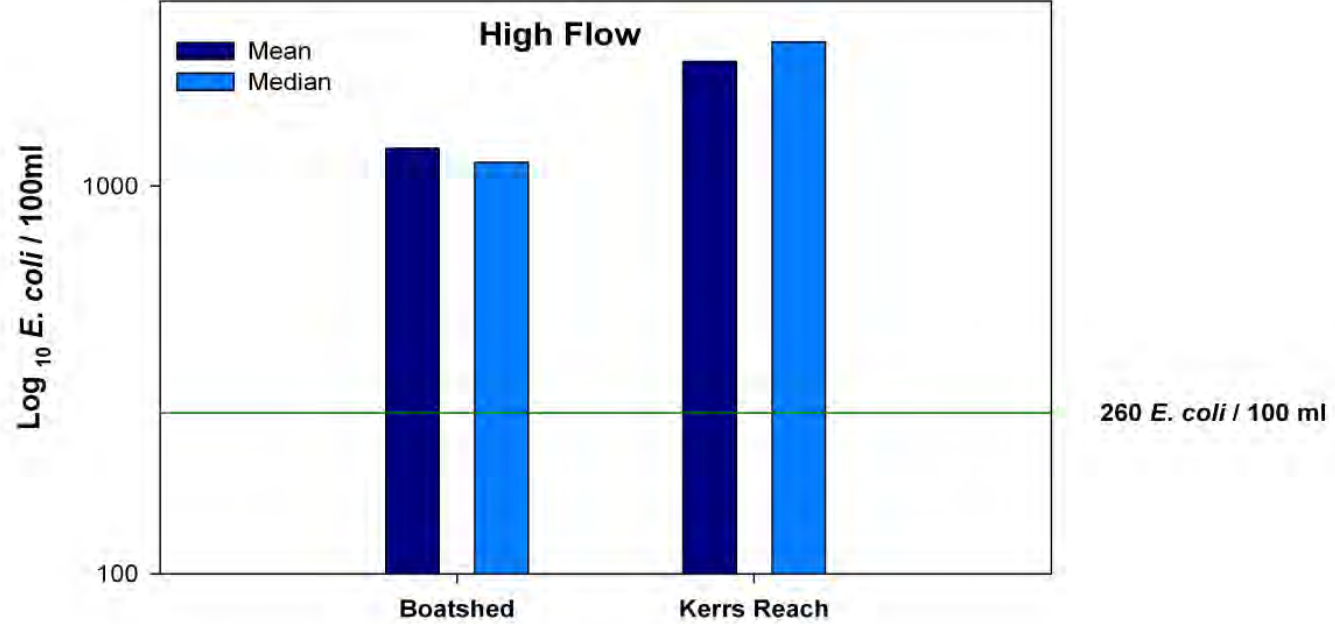
Kerrs Reach





March-May 2009

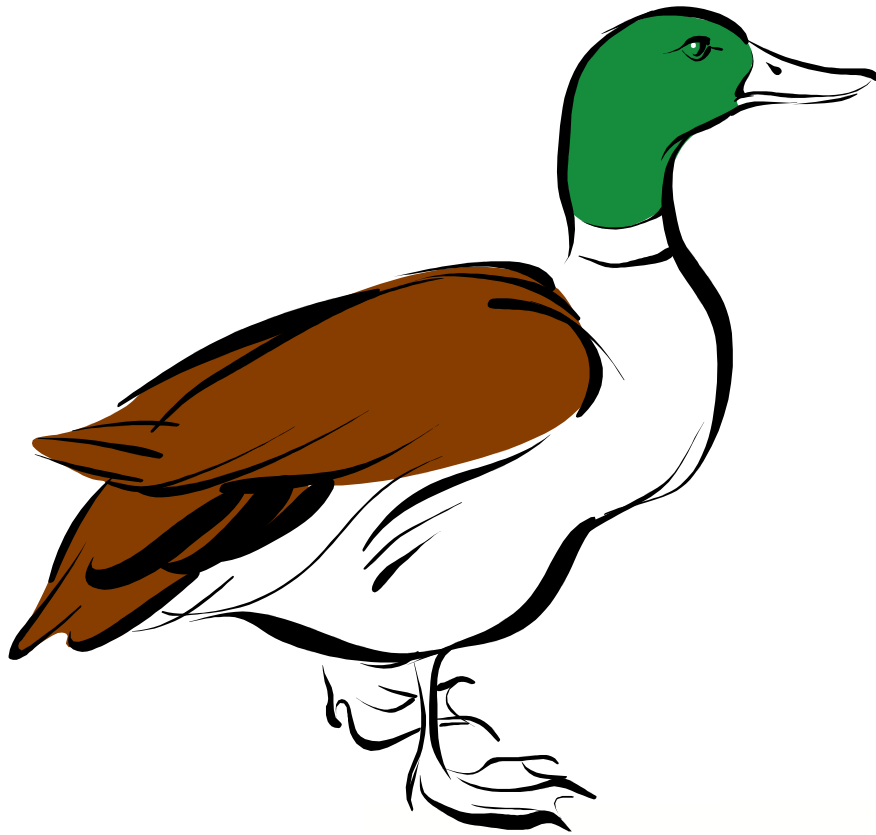
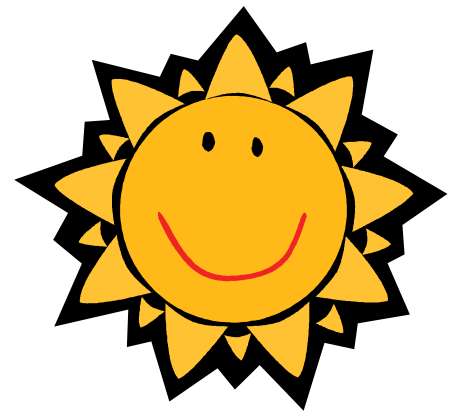




Location

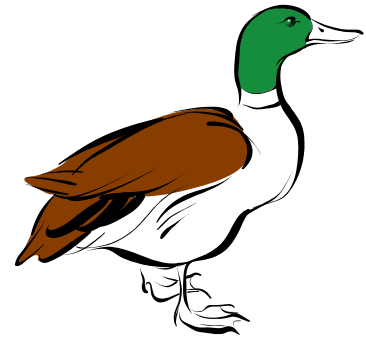
In absence of rainfall

- *E. coli* levels of up to 540 *E. coli*/100ml



Following rainfall

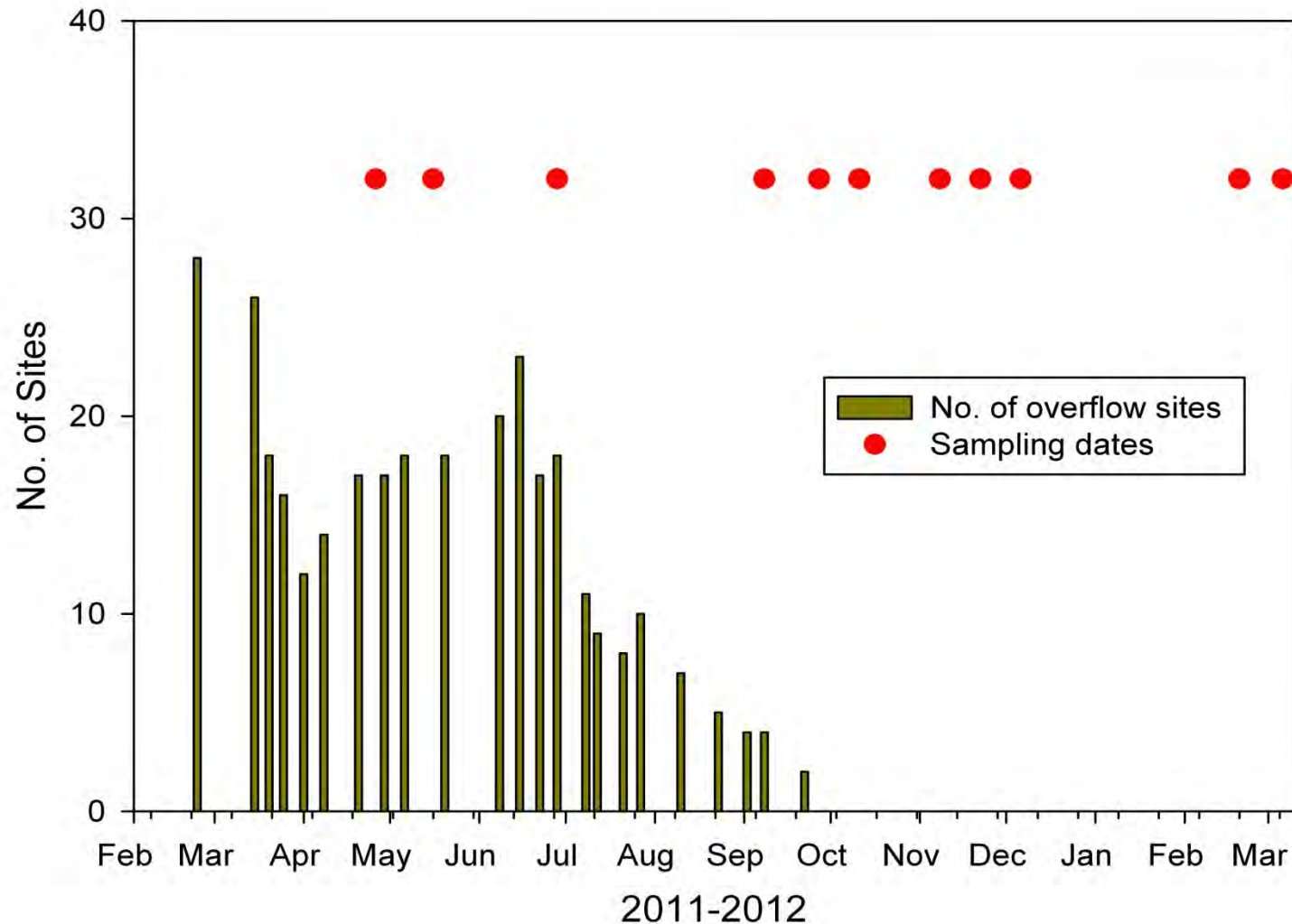
- Up to 3,600 *E. coli*/100ml.





Post-Earthquake sampling

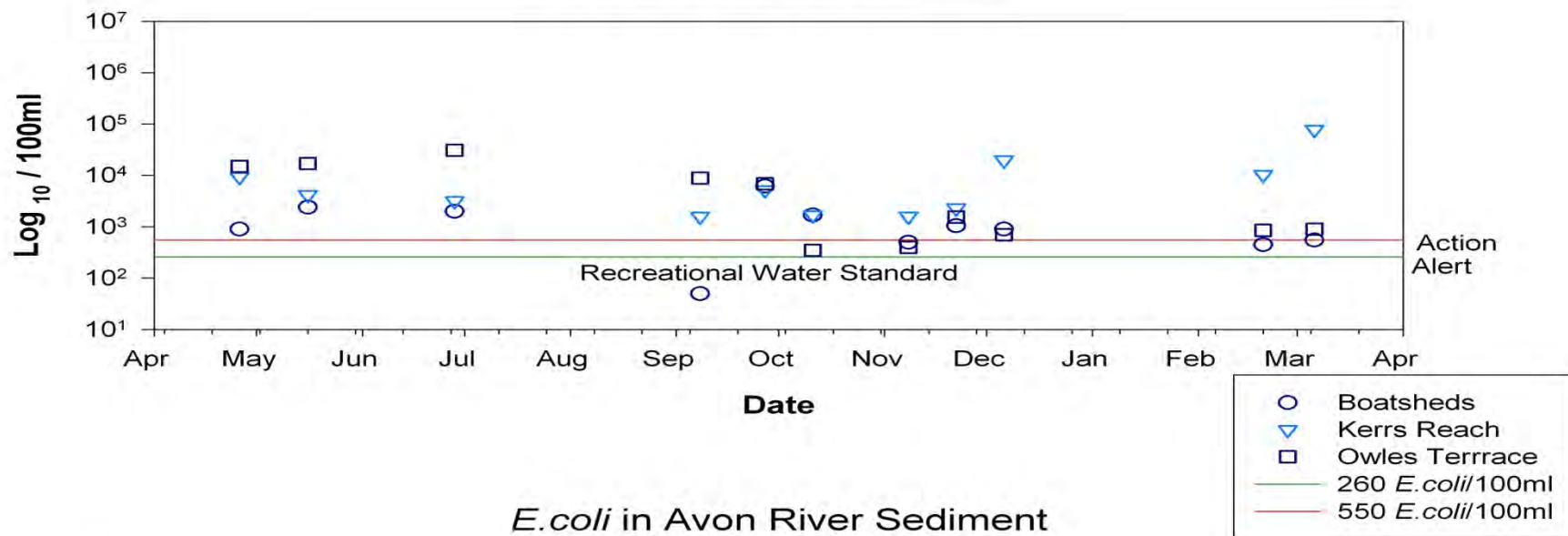
No. of overflow sites



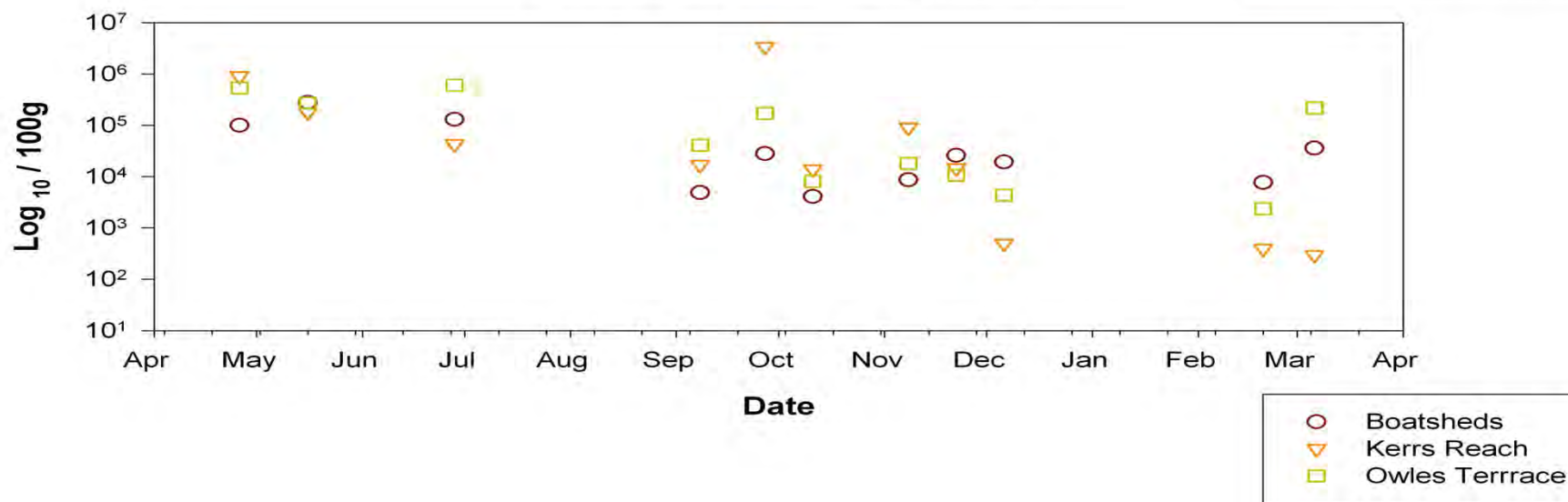




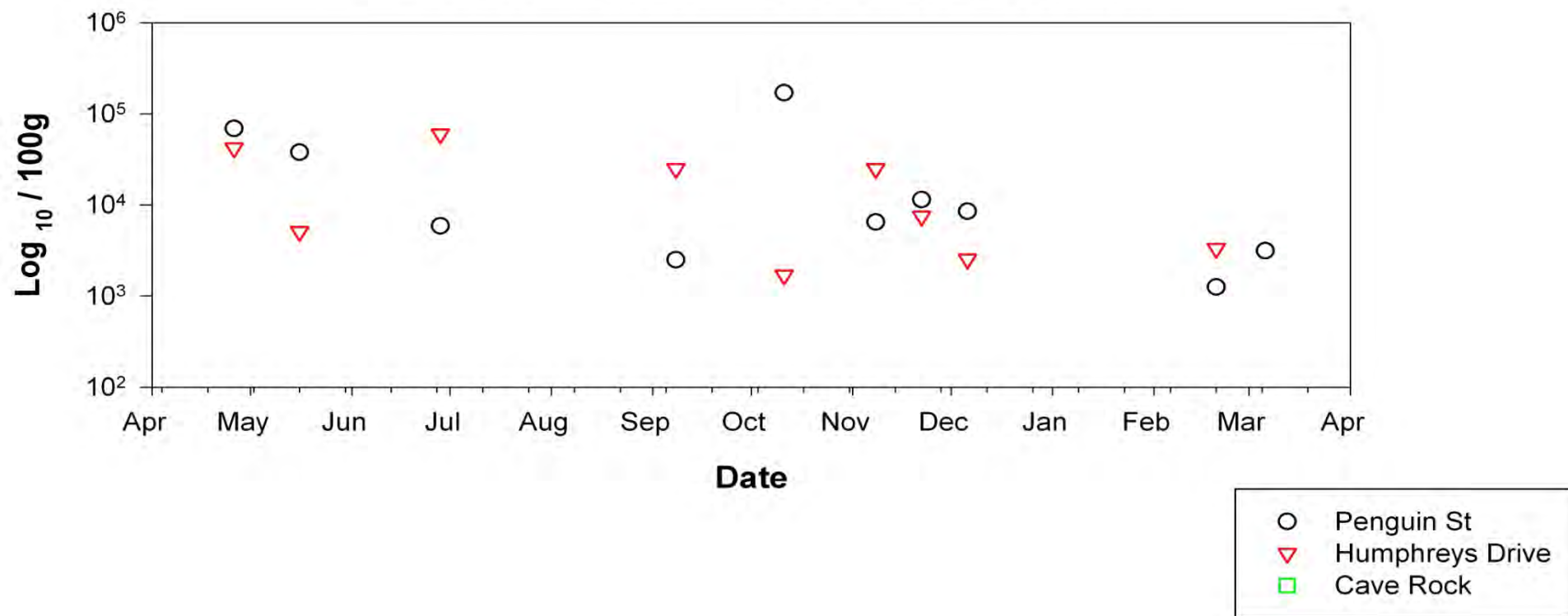
E.coli in Avon River Water



E.coli in Avon River Sediment

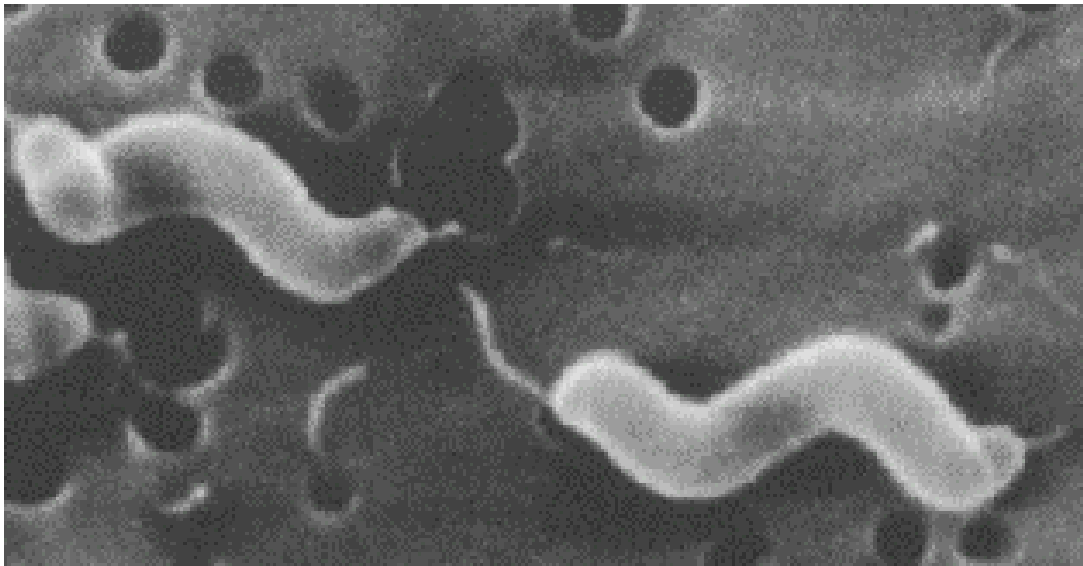


Enterococci in Estuarine and Coastal Sediment

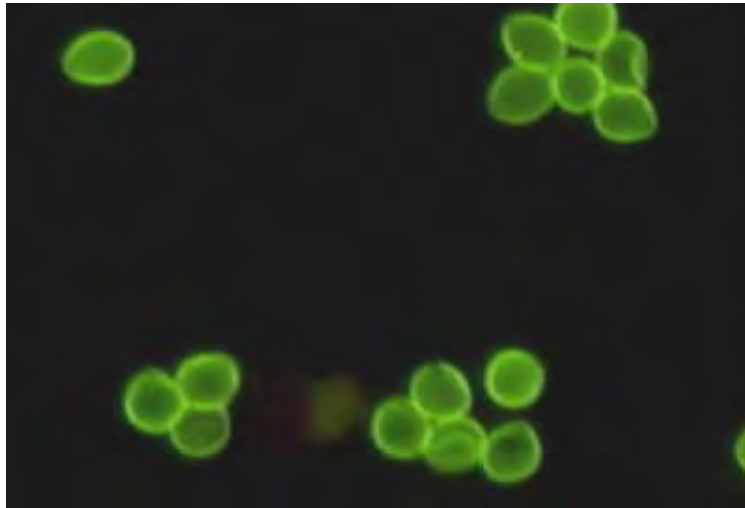
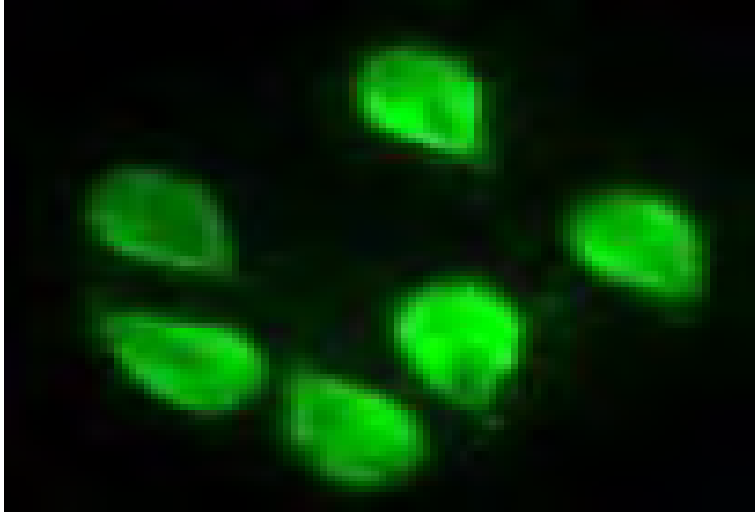


Campylobacter

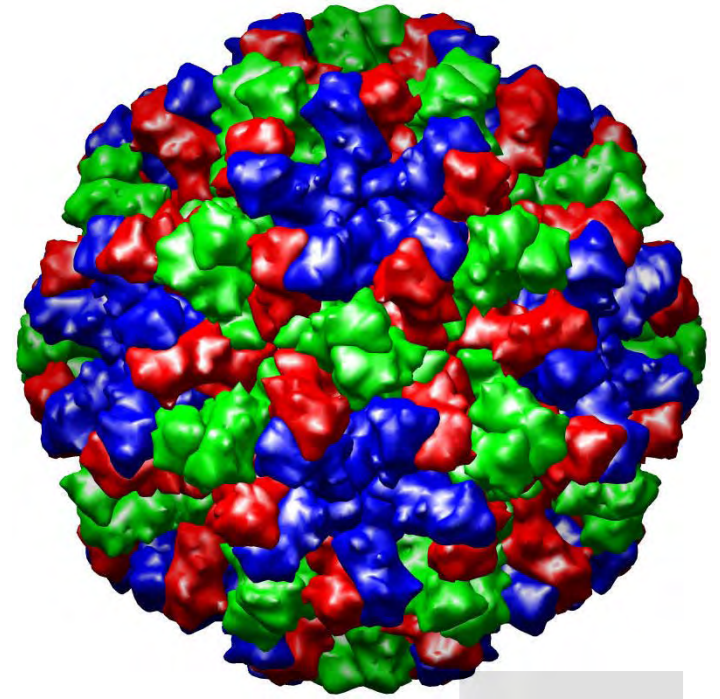
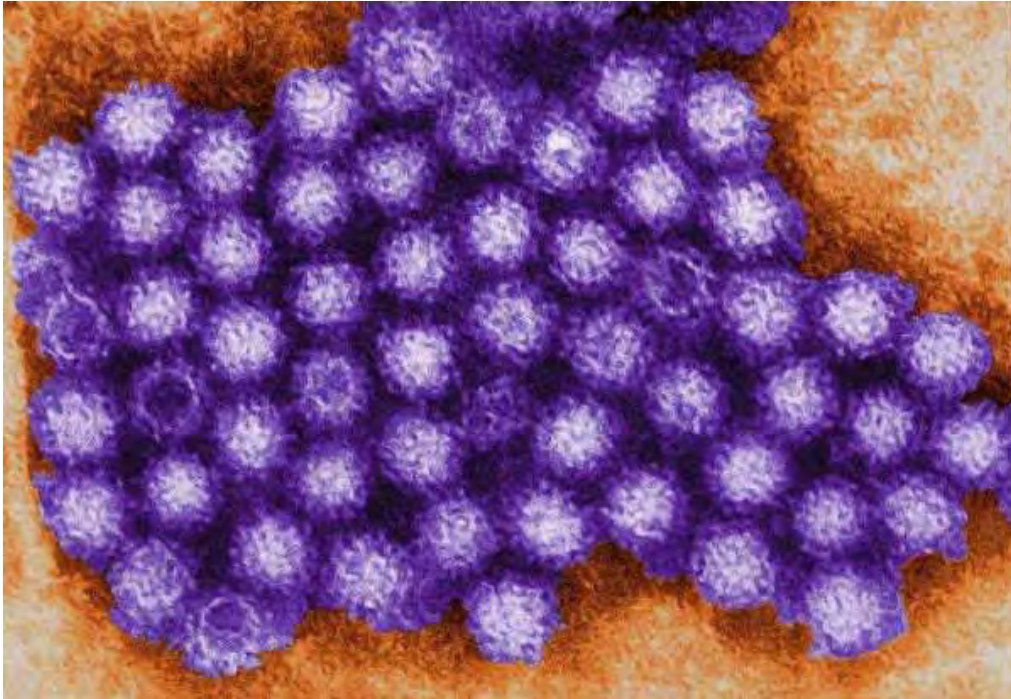
- Up to 110 Campylobacter/100ml



Protozoa



Viruses



NOROVIRUS



You don't want it.

Key findings

- **During discharges high levels of indicator bacteria and presence pathogens in the water**
 - Inform health risk estimates
- **Accumulation of micro-organisms in sediments**
 - Lack of correlation between water and sediments
 - Tidal influenced sites less accumulation
 - Heavy rainfall “wash” microorganisms out of sediments
- **Sumner Beach sediments no evidence of pathogens in beach sand**

Faecal Source Tracking in the Avon / Ōtākaro River

Methods for identifying faecal sources of contamination in waterways

**Megan Devane, Brent Gilpin,
Beth Robson, Susan Lin, Paula Scholes**

**Institute of Environmental Science and Research Ltd.
ESR**

What does it mean when you identify high levels of microbial indicators??

E. coli, Enterococci

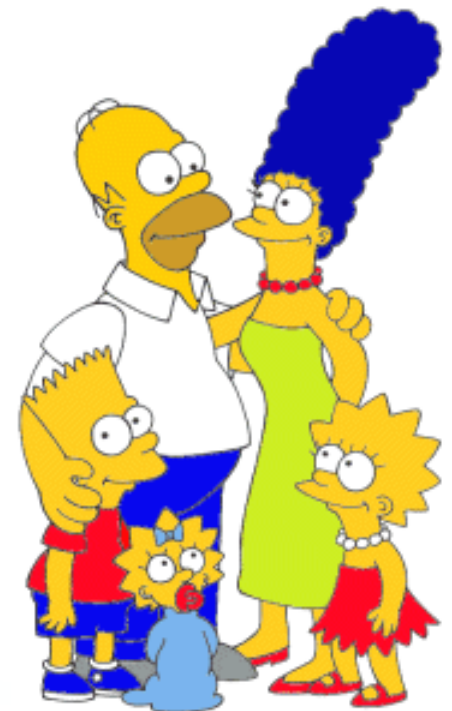
Sources of faecal
contamination



What does it mean when you identify high levels of microbial indicators??

E. coli, Enterococci

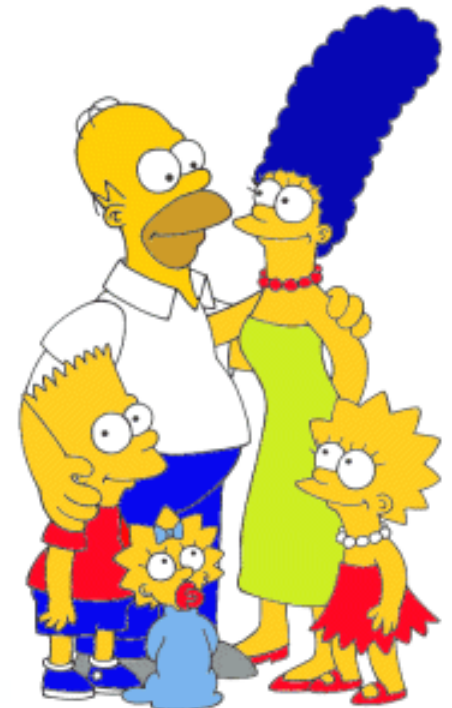
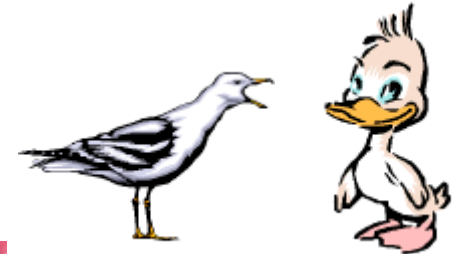
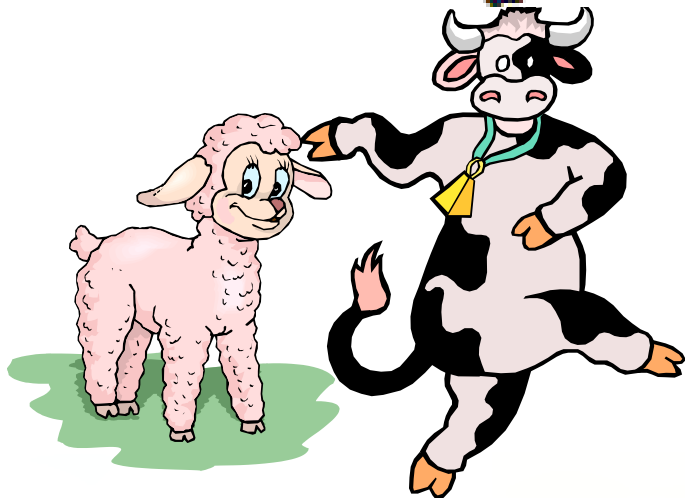
Sources of faecal
contamination



High levels of microbial indicators

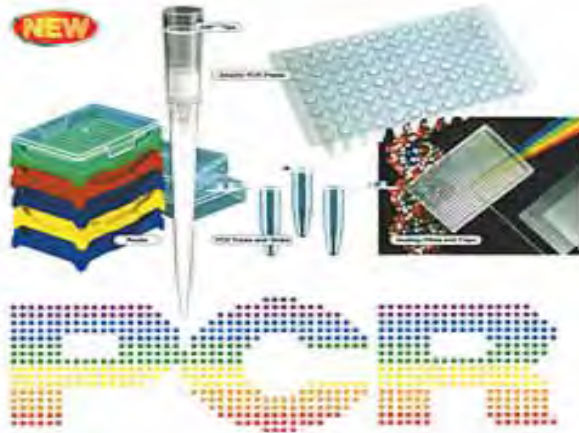
E. coli, Enterococci

Sources of faecal contamination



Toolbox for faecal source tracking

Molecular markers



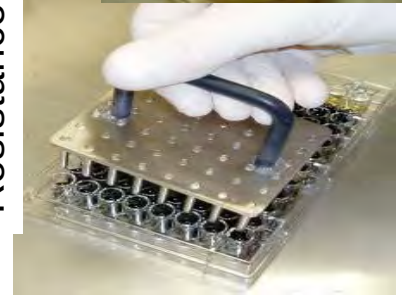
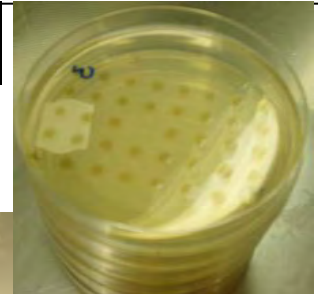
Indicative:
human,
ruminants
pigs, dog,
horses,
possum
ducks,

Library based methods



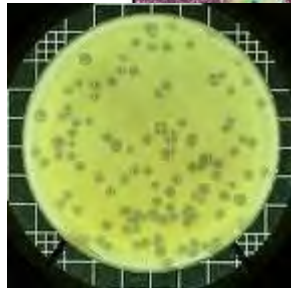
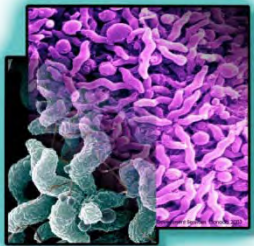
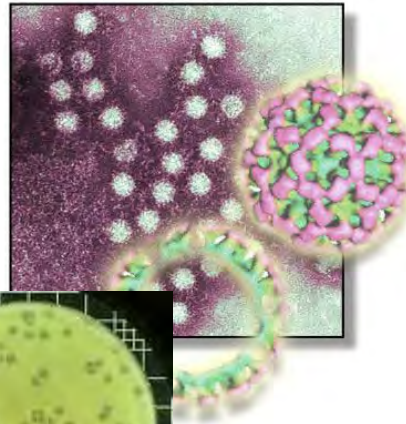
REP -PCR

Multiple Antibiotic
Resistance analysis



Pathogen Analysis

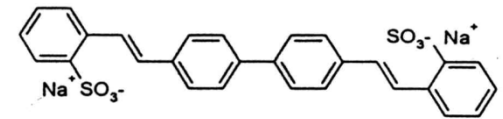
Noroviruses
Campylobacter typing
Cryptosporidium typing



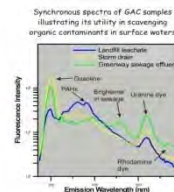
Chemical methods



FWAs



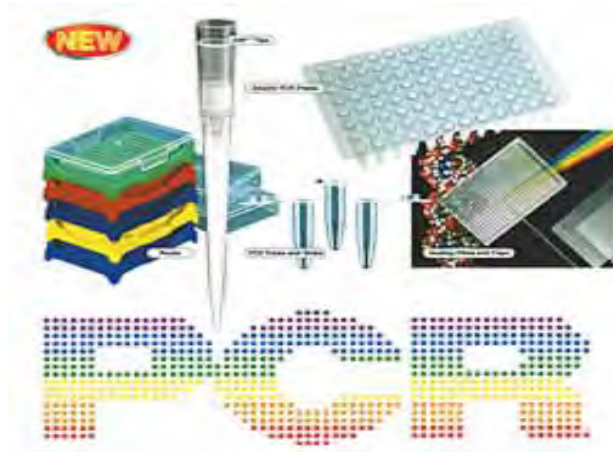
A1557 (FWA1)



Faecal sterol analysis

Toolbox for faecal source tracking

Molecular markers

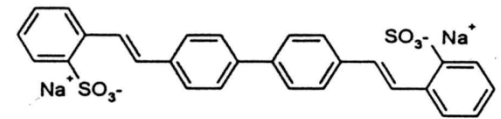


Indicative:
human,
ruminants
pigs, dog,
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ducks,

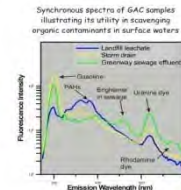
Chemical methods



FWAs



A1557 (FWA1)



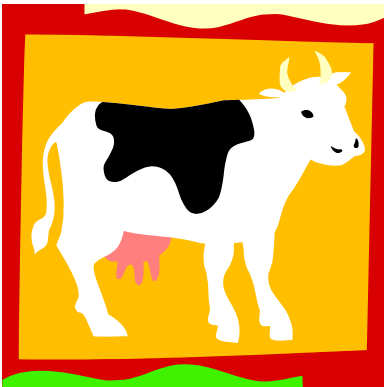
Faecal sterol analysis

Chemical Markers: Faecal Sterols

Each animal type has a similar range of sterols
but at different concentrations

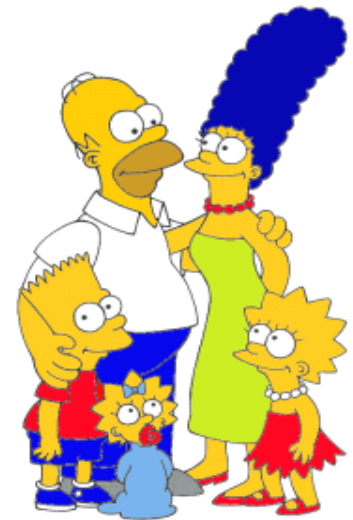
Sterol ratio analysis

Coprostanol / 24-ethylcoprostanol



>1.0 indicative of
human pollution

<1.0 indicative of pollution
from herbivores



Chemical Marker Fluorescent Whitening Agents (FWAs)

- Absorb UV-light and emit blue visible light
- Used “whiten” paper, plastic and fabrics.
- FWAs are added to washing powders
- FWAs in grey water mixed with sewage outflow
- specific indicator of human faecal contamination



Polymerase Chain Reaction (PCR)

Bacterial target	Target faecal source
<i>Bifidobacterium adolescentis</i>	Human
<i>Bacteroidales</i> - Human	Human
<i>Bacteroidales</i> - Herbivore	Ruminant
<i>Bacteroidales</i> – Dog	Dog
<i>Bacteroidales</i> – Pig	Pig
<i>Bacteroidales</i> – Horse	Horse
<i>Rhodococcus coprophilus</i>	Dairy effluent
E2 <i>Desulphovibrio</i>	Duck (Wildfowl)

Owles Terrace

**A comparison
of
FST markers
from
sediments
and overlying
water
samples**



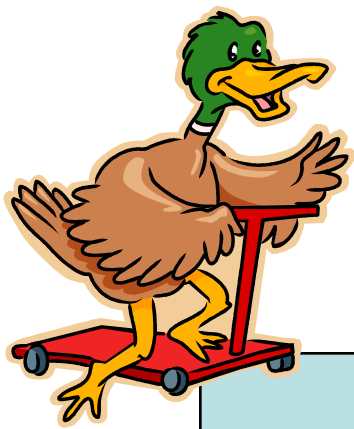
Why sample the sediments for FST markers?

- Evaluate whether sediments provide a historical record of faecal pollution
- Assess methods for aging faecal pollution

Question:

Whether resuspended sediments are likely to contribute to the FST markers identified in water samples?

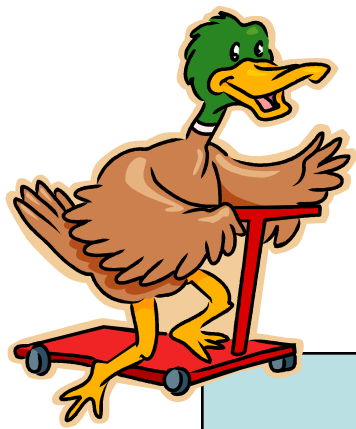




Boatshed Water



Date	<i>E. Coli</i>	Sterols	PCR Markers		
	<i>CFU/ 100 ml</i>		Human	Bird	Dog
23 Apr 2011	7,500	Wildfowl	ND	POS	POS
16 May 2011	2,350	Human	POS	POS	POS
8 Nov 2011	500	Wildfowl	ND	POS	ND
20 Feb 2012	450	Human	POS	POS	POS



Boatshed Sediment

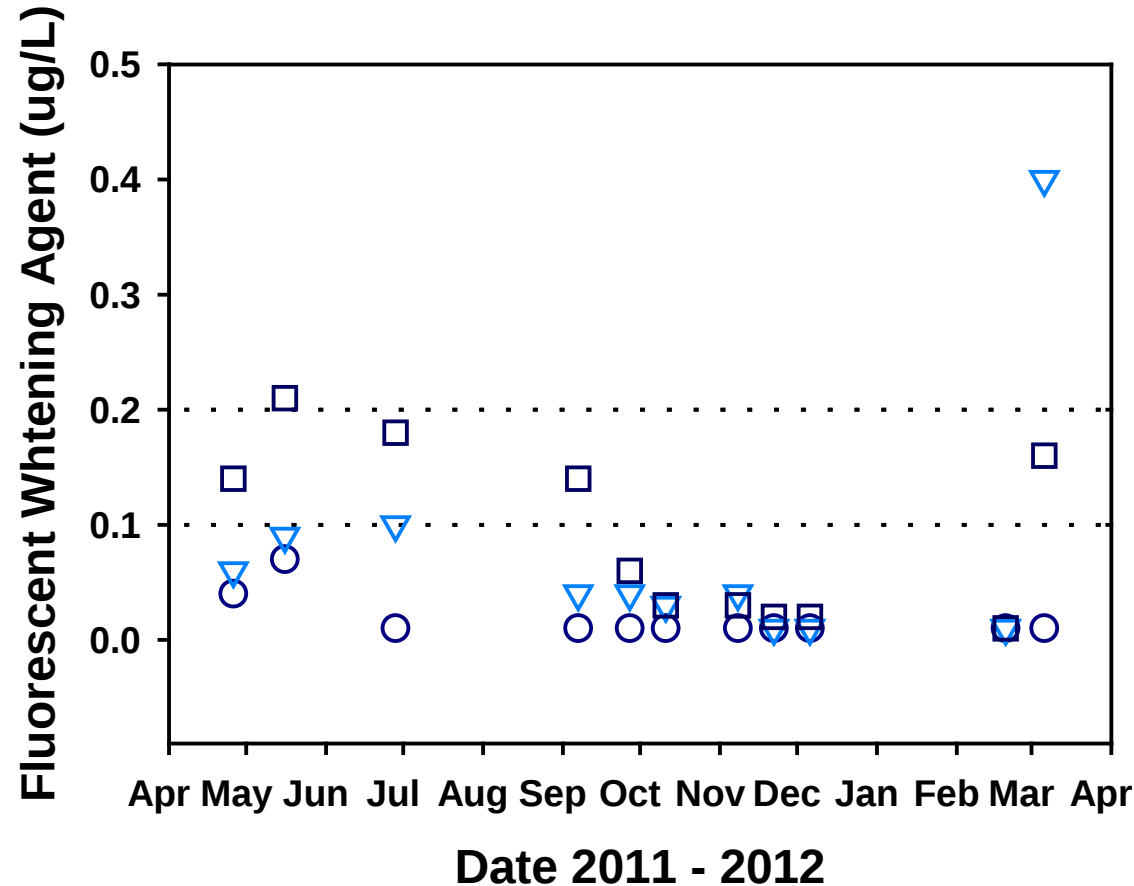


Date	<i>E. Coli</i>	Sterols	PCR Markers		
	<i>CFU/ 100 ml</i>		Human	Bird	Dog
23 Apr 2011	222,000	Wildfowl	ND	ND	ND
16 May 2011	277,000	ND	Low level	ND	ND
8 Nov 2011	8,700	ND	ND	ND	ND
20 Feb 2012	10,300	ND	ND	ND	ND

Sterol concentrations and sterol ratio analysis for Owles Terrace 26 April 2011- 20 Feb 2012

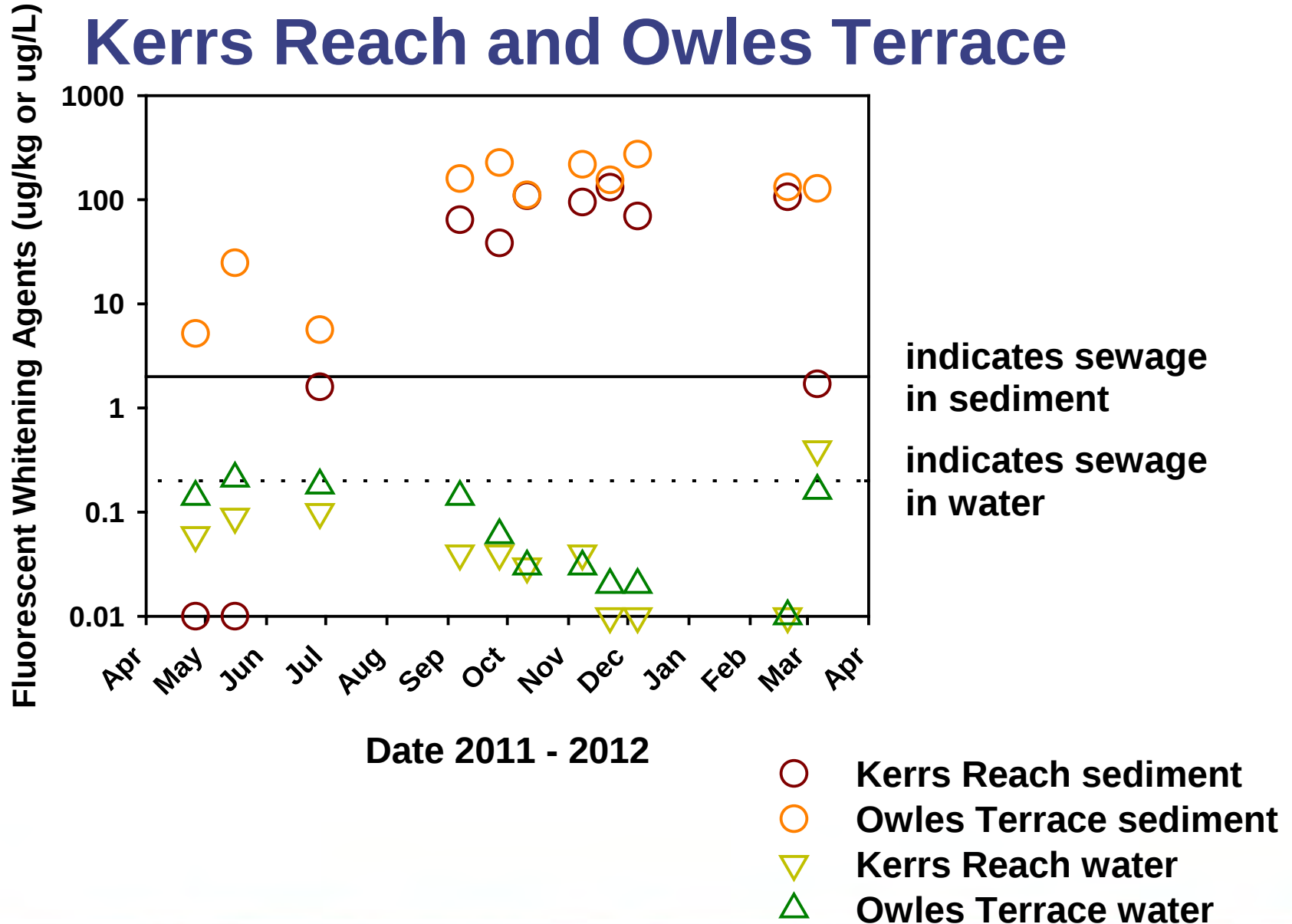
Sterol ratio analysis	Type of faecal indicator	Sediment	Water
coprostanol/cholestanol	Non-specific indicators of faecal contamination	faecal	faecal
24-ethylcoprostanol/ 24-ethylcholestanol		faecal	faecal
%Coprostanol/total sterols	 Human faecal indicators	human	human
Coprostanol/(coprostanol cholestanol)		human	human
coprostanol/24-ethylcoprostanol		human	human
%coprostanol/ coprostanol+24-ethylcoprostanol	Human versus herbivore indicator	human	human
% 24-ethylcoprostanol/total sterols	Herbivore specific indicator	ND	ND
24-ethylcholestanol/ (24-ethylcholestanol+24-ethylcoprostanol +24-ethylepicoprostanol)	Wildfowl	ND	ND

FWAs in Avon / Ōtākaro River water

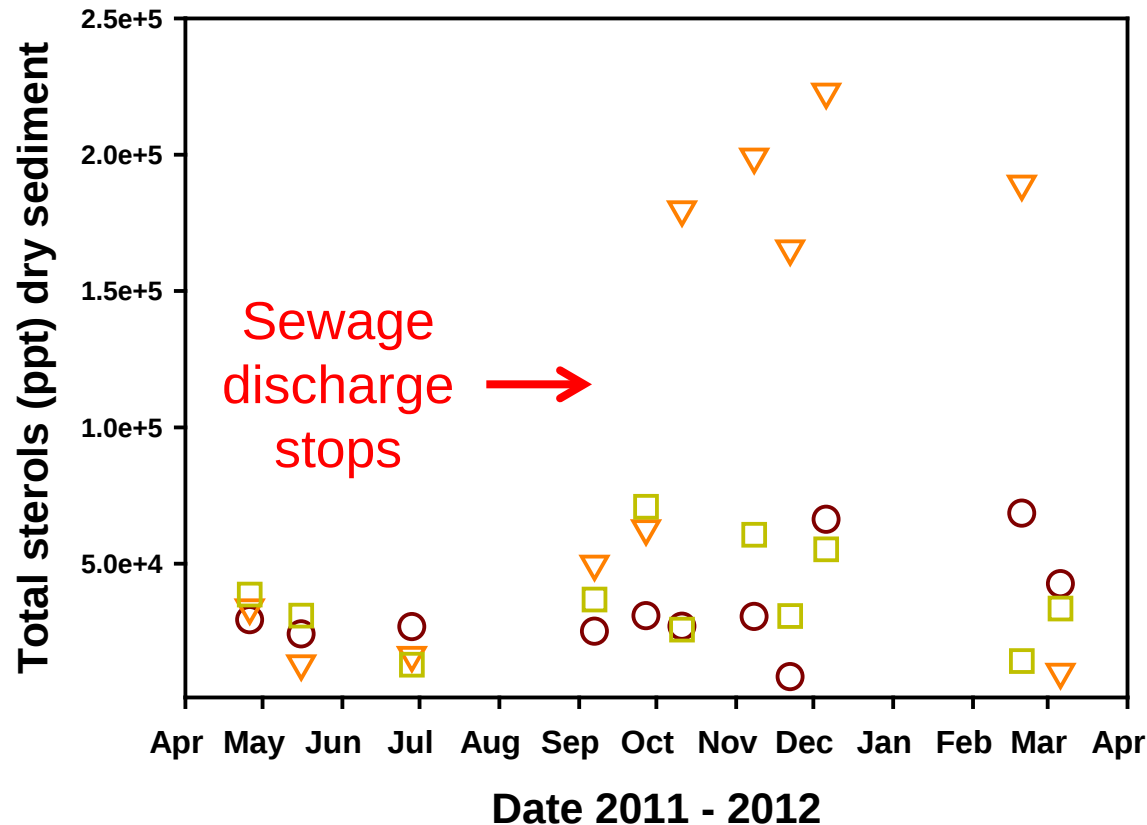


- Boatsheds water
- ▽ Kerrs Reach water
- Owles Terrace water

Accumulation of FWA in sediments at Kerrs Reach and Owles Terrace



Sterols in Avon / Ōtākaro River sediments



>10 fold less total sterols
in water column

- Boatshed sediments
- ▽ Kerrs Reach sediments
- Owles Terrace sediments

Faecal Aging Methods



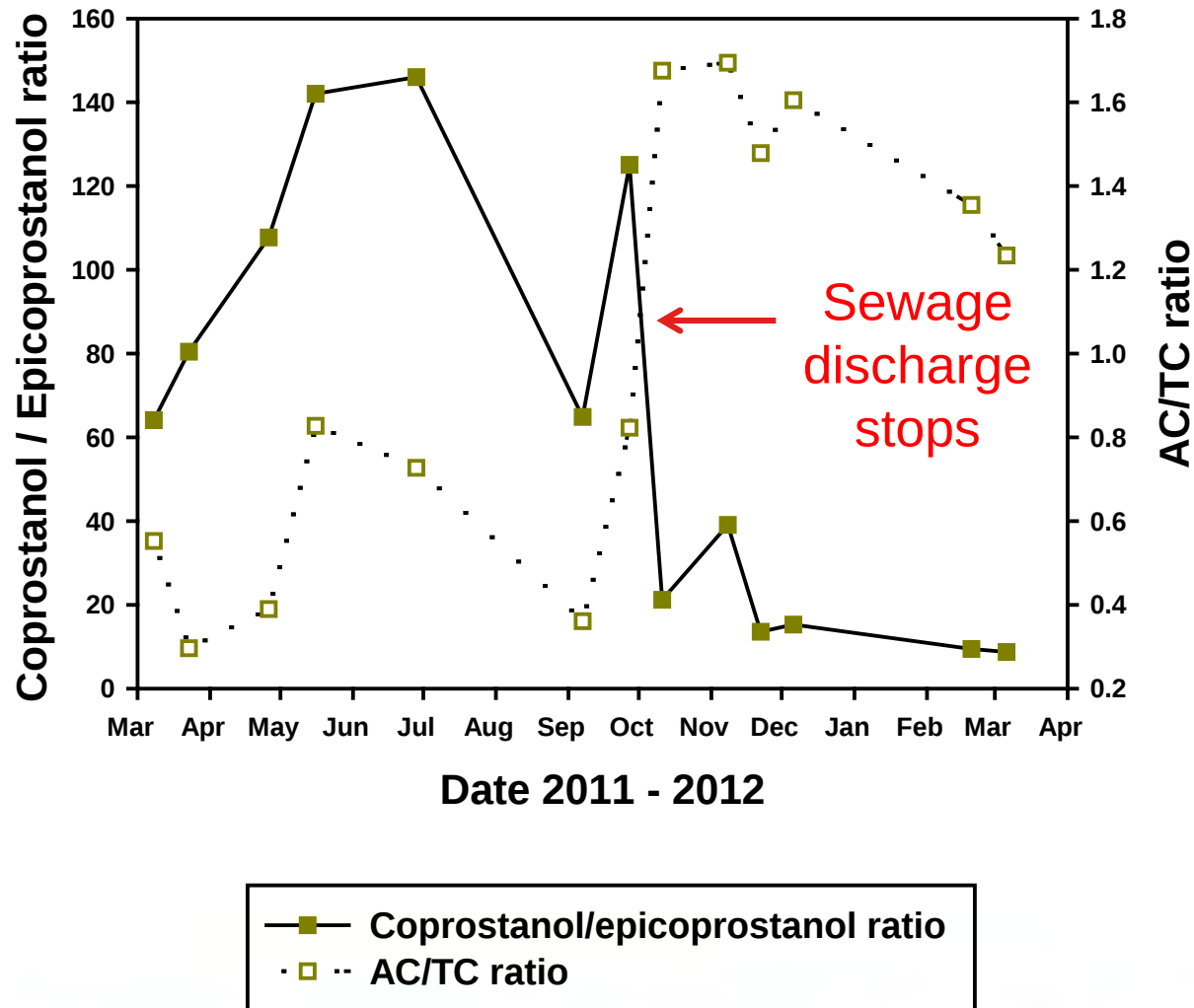
- **Bacterial Ratio** $\frac{\text{Atypical colonies}}{\text{Total Coliforms}}$
Low ratios = fresh sewage

- **Sterol ratio** $\frac{\text{Coprostanol}}{\text{Epicoprostanol}}$

high ratios = fresh sewage



Faecal aging ratios in water at Owles Terrace



Finding from this study:

Faecal aging ratios

For water impacted by human sewage:

A negative correlation was identified between the sterol aging ratio and the Total coliform ratio based on bacterial enumeration

Sterol ratio : Bacterial ratio ($P = 0.013$)



Conclusions for FST markers in Avon / Ōtākaro River sediments

	Marker accumulating in sediment
Faecal Sterols	
PCR markers	
Fluorescent Whitening Agents	

Faecal Source Tracking Conclusions

- **Sediments provide a historical record of faecal pollution**
- **Disconnect between FST markers identified in sediment and overlying water**
- **Two promising methods for aging human faecal pollution**

FUNDERS

- Environment Canterbury
- Canterbury District Health Board
- Christchurch City Council
- Ministry of Health
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- FRST

ESR

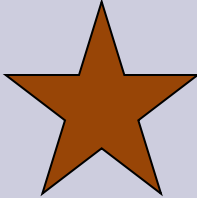
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www.waterquality.org.nz



FST markers for Avon River

	Water	Sediment	Marker accumulating in sediment
Faecal Sterols			
PCR markers			
Fluorescent Whitening Agents			

Faecal Source Tracking

Using science to help inform water management decisions

[Home](#)[Decision Trees](#)[Toolbox](#)[Case Studies](#)[Research](#)[Publications](#)[Contacts](#)

An information source for those interested in identifying sources of faecal contamination

Faecal Source Tracking:

www.waterquality.org.nz

Sophisticated Techniques to Identify and Mitigate Water Contamination Issues

Waters clean enough for humans to drink, swim in, process food with and collect food from, are indicative of a wider healthy environment. However, access to such pristine waters is diminishing as urbanisation expands and agricultural production increases, leading to faecal contamination and reduced microbiological water quality. Issues surrounding factors that may compromise water quality must be addressed and resolved.

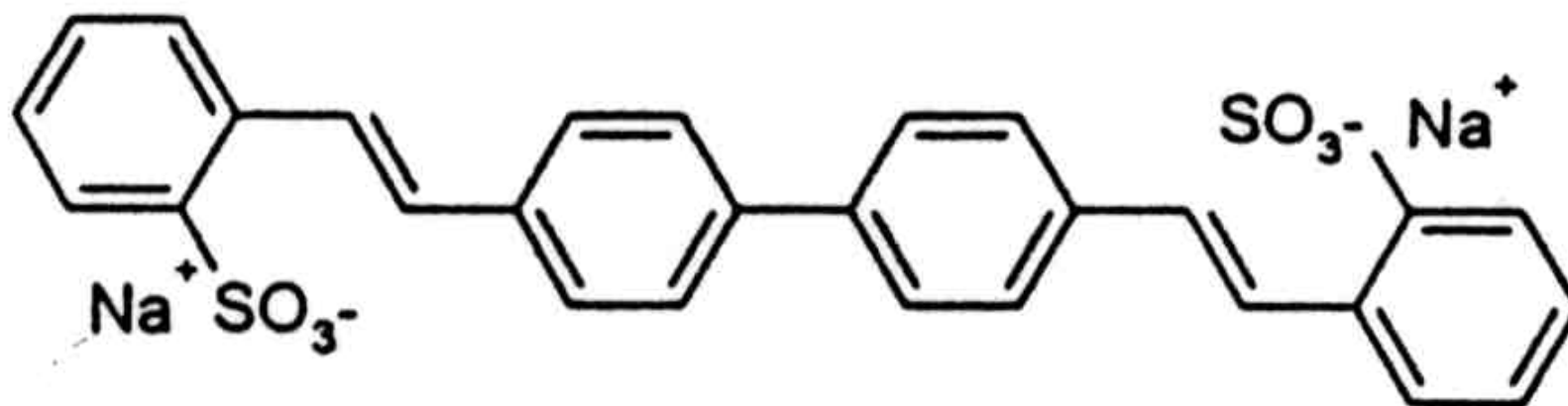
When waterways become contaminated with faeces, an important step in contamination management is identifying the source of the pollution. A research team at ESR is developing tools that will improve the microbial quality of water by discriminating among different sources of faecal contamination of water, leading to targeted environmental management interventions.

Scientists use molecular and chemical techniques to help identify the source of faecal contamination of water whether it is from farm, domestic or feral animals, humans or birds. If an initial Colilert® test indicates the presence of high counts of *Escherichia coli*, one or more assays from a 'tool box' of



FWA1

FWA 1



4,4'-bis(2-sulfostyryl)biphenyl

- Photodegradation with half life several hours
- Below photic zone, persistent since biodegradation not been observed.

Faecal Aging Methods



Second marker: **Sterol Ratio**

- Based on faecal sterols
- **Coprostanol / Epicoprostanol**
- **Higher ratios** are associated with high levels of coprostanol, the major human sterol
- **The ratio reduces over time** as sterols are degraded to epicoprostanol.

