Measuring Bacteria and Viruses

California Water Quality Control Board 2022 Bacterial Summit

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Southern California Coastal Water Research Project
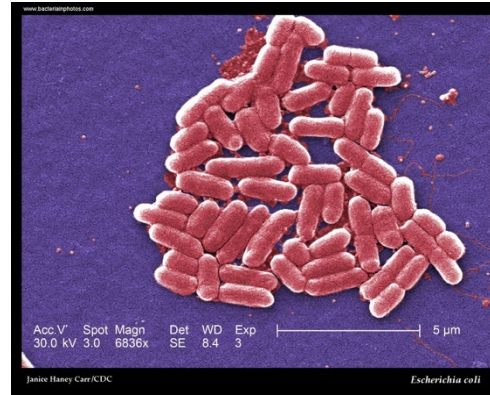
Measuring bacteria and viruses

- What are we measuring and why?

- How do we measure them?
  - Growth-Based Methods
  - Gene-Based Methods (Molecular Methods)

- What else can we measure?

- Sampling and practical considerations?
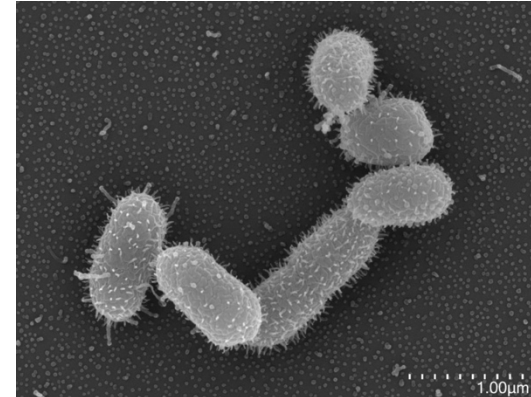
What are we measuring?
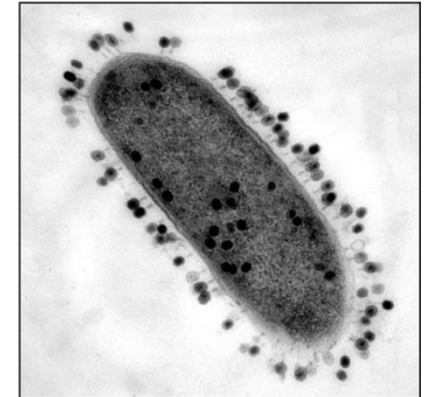
Fecal Indicator Bacteria

Enterococcus

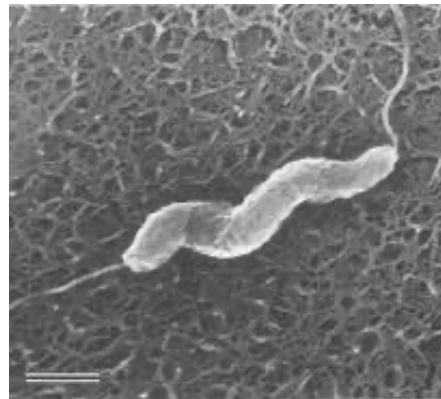Coliforms (E. coli)

Alternative Indicators

Bacteroides (HF183)

Coliphage

Pathogens

Campylobacter

Norovirus
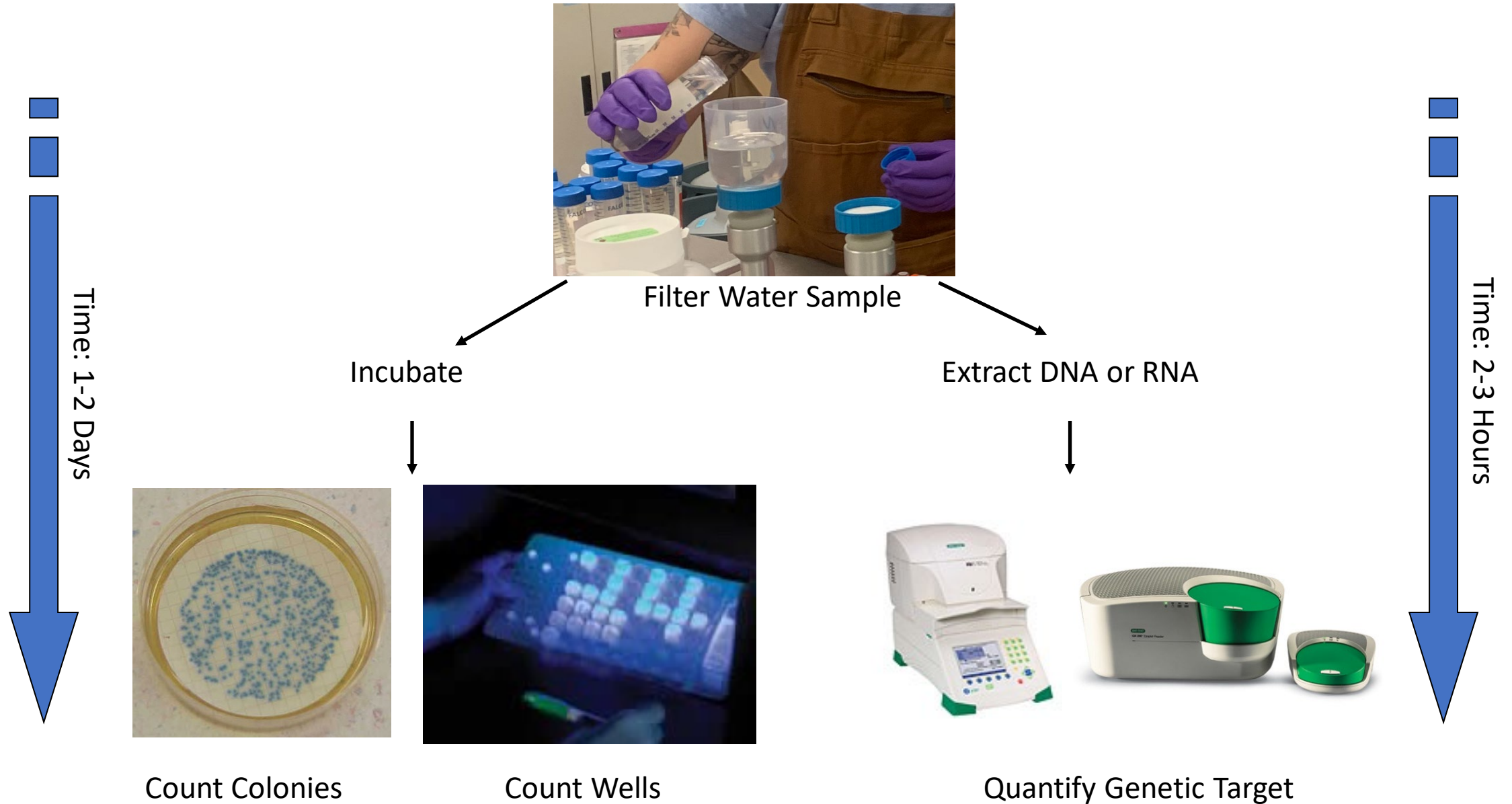
Why are we measuring them?

|	Fecal indicators	Alternative indicators	Pathogens
Abundance when there is fecal contamination	high	high	varying
Fecal source identification?	no	depends on the indicator	depends on the pathogens
Link to illness?	Statistically linked to illness risk	Statistically linked to illness risk	Cause illness

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Growth-based vs. Gene-based Methods

Filter Water Sample

Incubate

Extract DNA or RNA

Time: 1-2 Days

Time: 2-3 Hours

Count Colonies

Count Wells

Quantify Genetic Target

Growth-based methods

- Developed more than a century ago
  - Total Coliforms used in recreational waters since at least the 1950s

- Bacteria are grown overnight in a laboratory then counted by hand
  - 24-72 hours to results

- Fecal indicator bacteria have existing regulatory standards
  - Approved by EPA in 1976
  - Recreational water and shellfish

- Methods being developed and tested for coliphage as indicator viruses
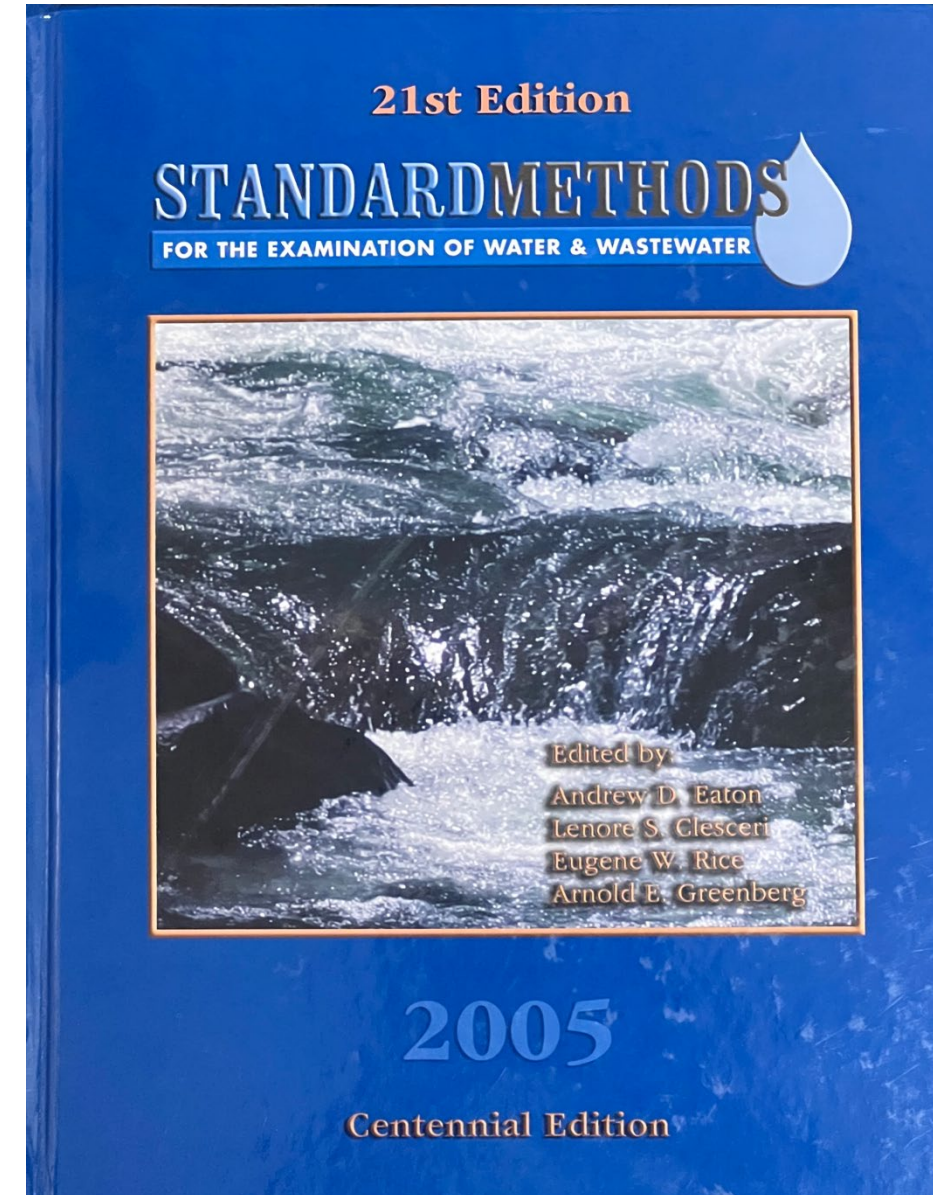
Limitations of Growth-Based methods

- General fecal indicators, not specific
  - No source identification

- Long wait for results

WARNING

OCEAN WATER CONTACT MAY CAUSE ILLNESS

¡AVISO!

EL CONTACTO CON AGUA DEL OCEANO PUEDE CAUSAR ENFERMEDADES LOS NIVELES DE BACTERIAS EXCEDEN LOS ESTANDARES DE SALUD

ORANGE COUNTY ENVIRONMENTAL HEALTH DIVISION FOR FURTHER INFORMATION, CALL (714) 667-3752 www.ocbeachinfo.com.

WARNING

OCEAN WATER QUALITY WAS BAD YESTERDAY AND YOU SHOULD NOT HAVE SWUM

WATER QUALITY MIGHT BE OK TODAY

PLEASE CHECK OUR WEBSITE TOMORROW

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Limitations of Growth-Based Methods

- General fecal indicators, not specific
  - No source identification

- Long wait for results

- Complex environmental samples can interfere with growth

- Fecal indicator bacteria or viruses do not cause illness

Gene-Based methods (molecular methods)

- Developed 15-20 years ago

- Bacterial DNA is isolated in a laboratory and quantified
  - As little as 3 hours to results

- Fecal indicator bacteria have existing regulatory standards
  - Approved by EPA in 2012
  - Recreational water

- Greater specificity
  - Can measure other targets
  - Alternative indicators

Alternative indicators

Considerations When Implementing Gene-Based Methods

- Higher materials cost

- Complex environmental samples can interfere with chemistry

- Shorter history of use
  - Fewer long-term datasets
  - Fewer locations

- Fecal indicator bacteria and alternative indicators do not cause illness

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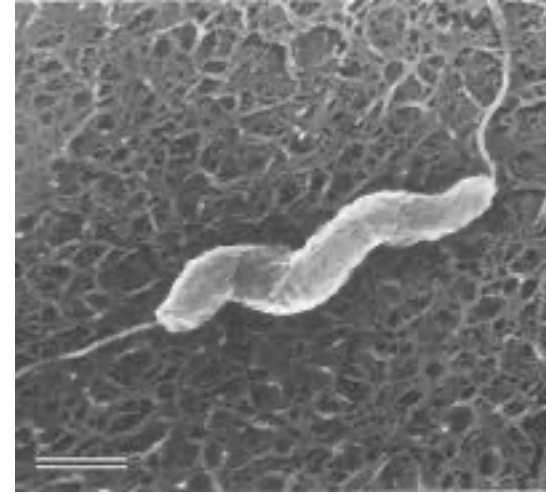- Sampling and practical considerations

What else can we measure?

- Fecal indicators and alternative indicators still have the limitations of being indicators
  - They're not measuring what actually makes you sick

- Culturing pathogenic viruses and bacteria is an option in some cases
  - Not for all pathogens

- The same gene-based technologies we've developed for source tracking can measure pathogenic viruses and bacteria
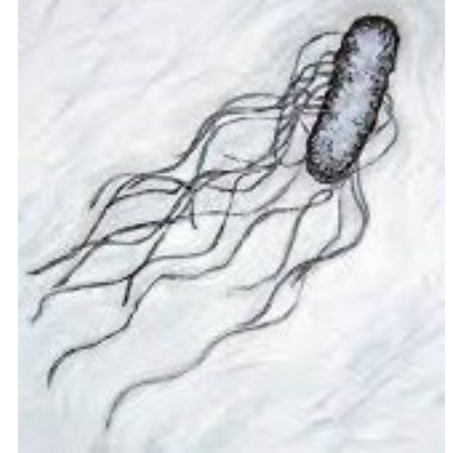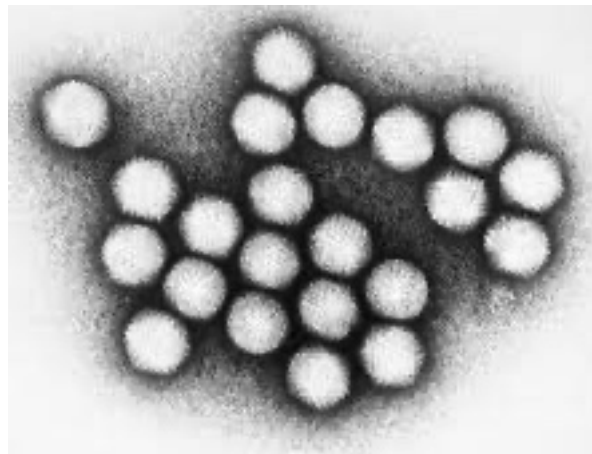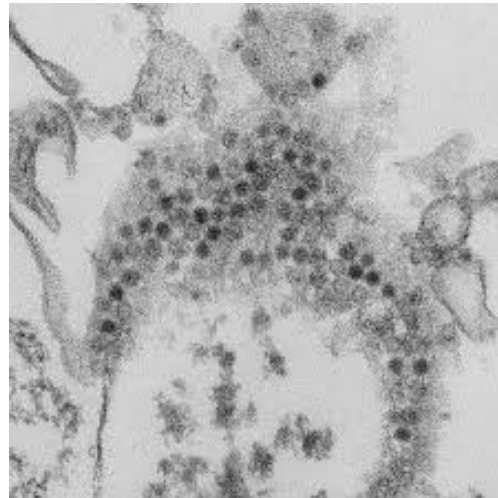
Which pathogens?
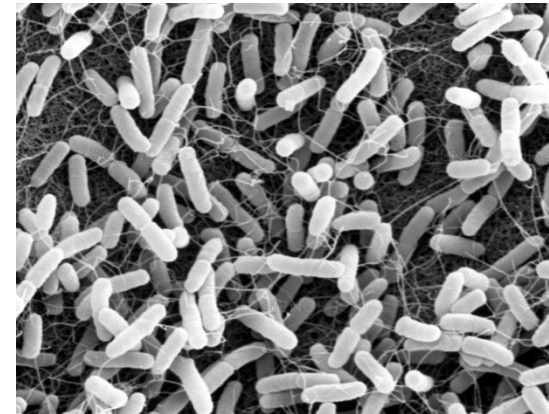

Norovirus

Campylobacter

Salmonella

Adenovirus

Enterovirus

Shiga Toxin
E. coli

Vibrio

Sampling and other practical considerations

Sampling

- EPA allows two sampling approaches to determine bacterial water quality

    - Single sample
        - Short-term assessment
        - Can miss intermittent contamination

    - Rolling 30-day geometric mean (weekly sampling)
        - Long-term trends
        - Can smooth out short-term increases

Other practical considerations

- Time of sampling
  - Early morning to get results faster
  - Before UV rays reduce bacterial numbers

- Event based sampling
  - tides, storms, spills

- Contamination source
  - Raw vs treated sewage

- Fate and transport
  - Contamination through sand or groundwater

- Deciding which methods to use and when

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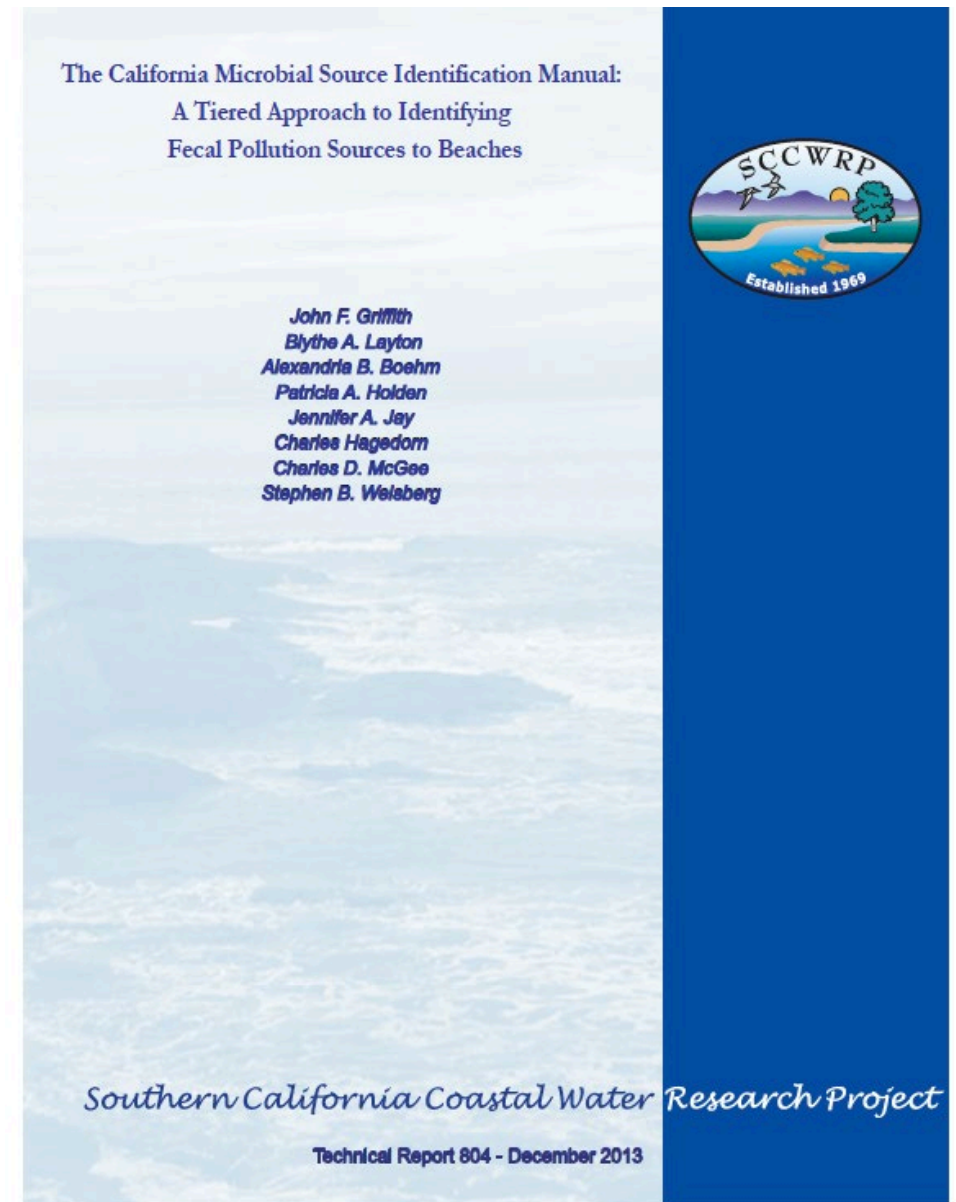
The California Microbial Source Identification Manual:
A Tiered Approach to Identifying
Fecal Pollution Sources to Beaches

John F. Griffith
Blythe A. Layton
Alexandria B. Boehm
Patricia A. Holden
Jennifer A. Jay
Charles Hagedorn
Charles D. McGee
Stephen B. Weisberg

SCCWRP
Established 1969

Southern California Coastal Water Research Project

Technical Report 804 - December 2013

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