

Measuring Bacteria and Viruses

California Water Quality Control Board 2022 Bacterial Summit

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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

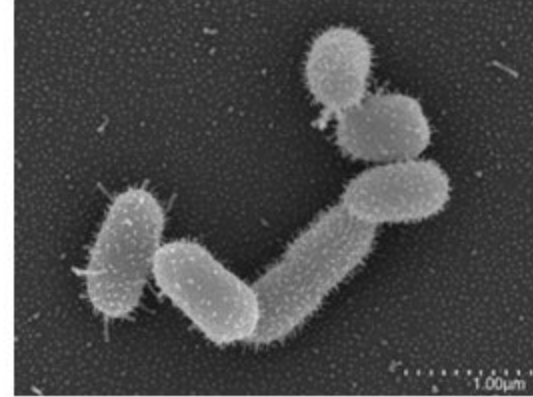


Enterococcus



Coliforms (E. coli)

Alternative Indicators

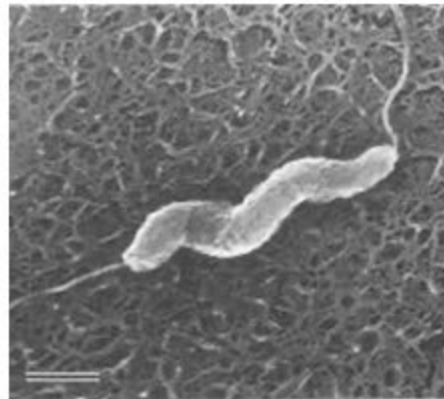


Bacteroides (HF183)



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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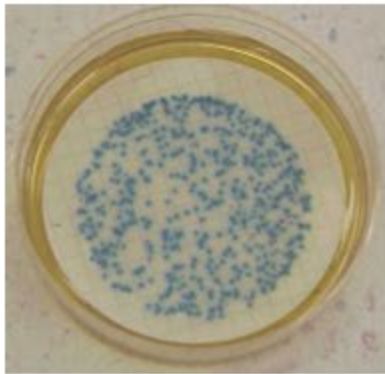
Growth-based vs. Gene-based Methods



Filter Water Sample

Incubate

Extract DNA or RNA



Count Colonies



Count Wells



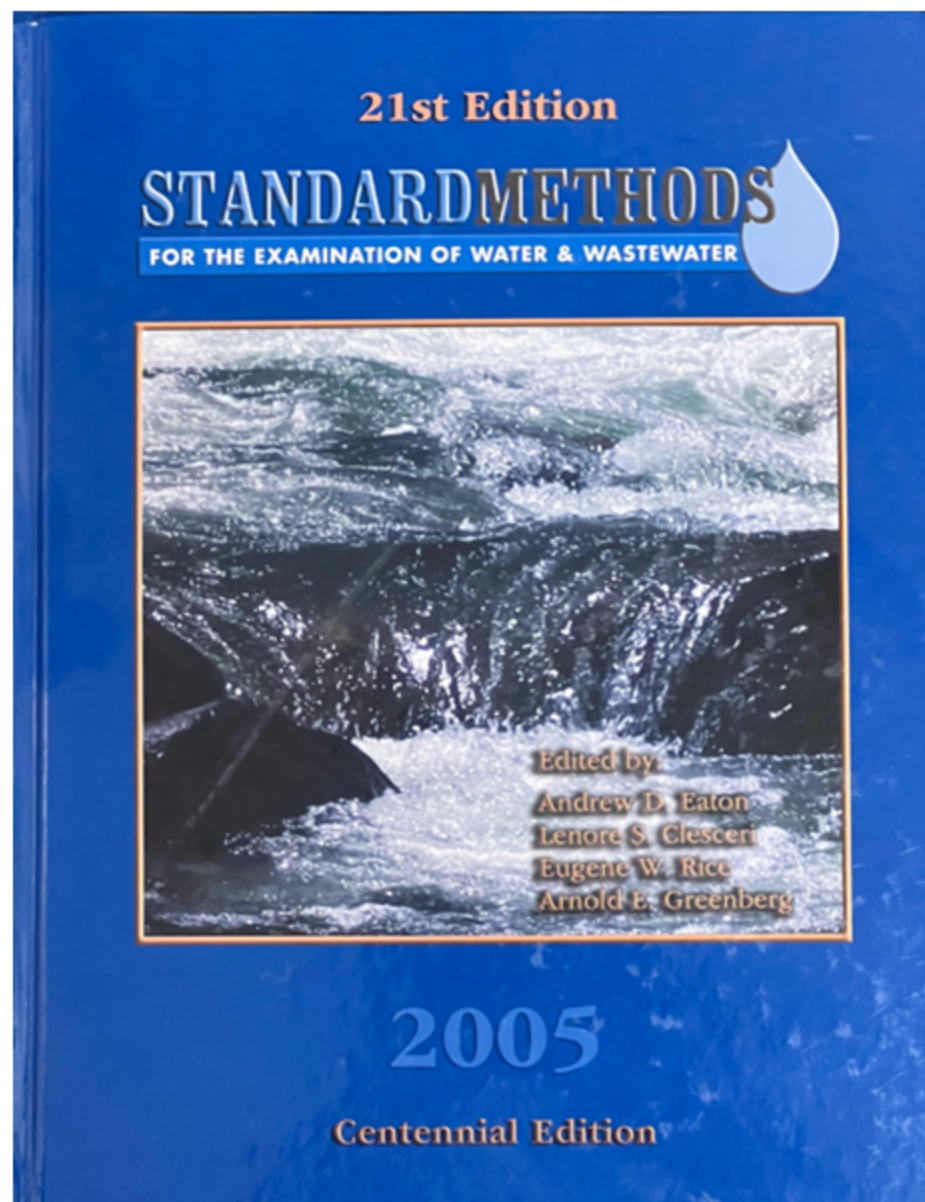
Quantify Genetic Target

Time: 1-2 Days

Time: 2-3 Hours

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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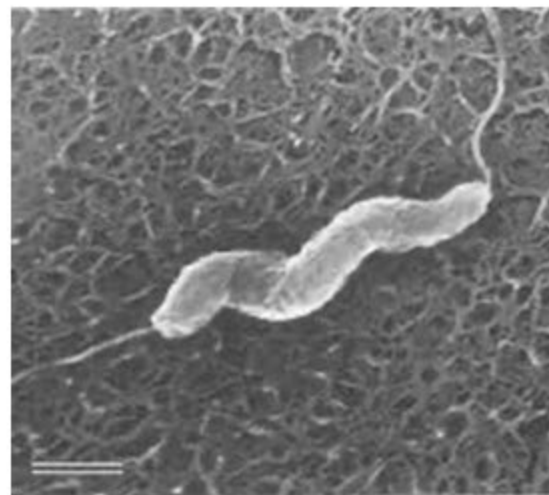
What else can we measure?

- 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

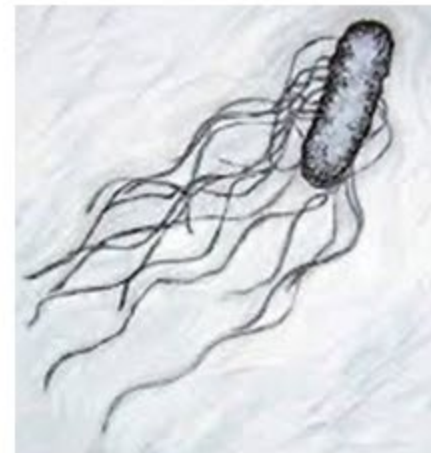
Which pathogens?



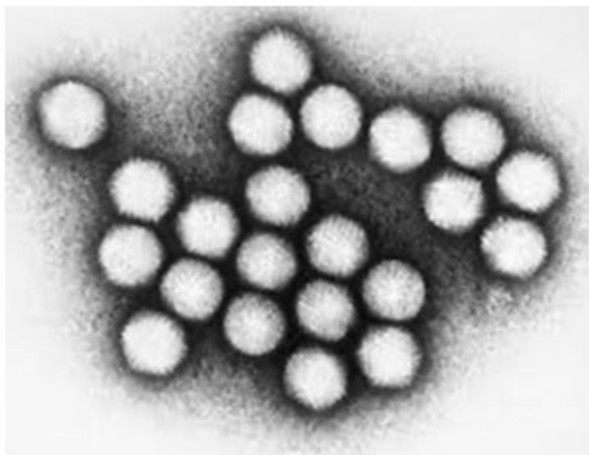
Norovirus



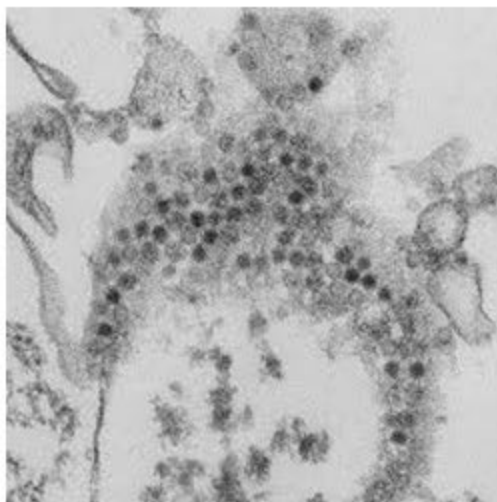
Campylobacter



Salmonella



Adenovirus



Enterovirus



**Shiga Toxin
E. coli**



Vibrio

Sampling and other practical considerations



Sampling

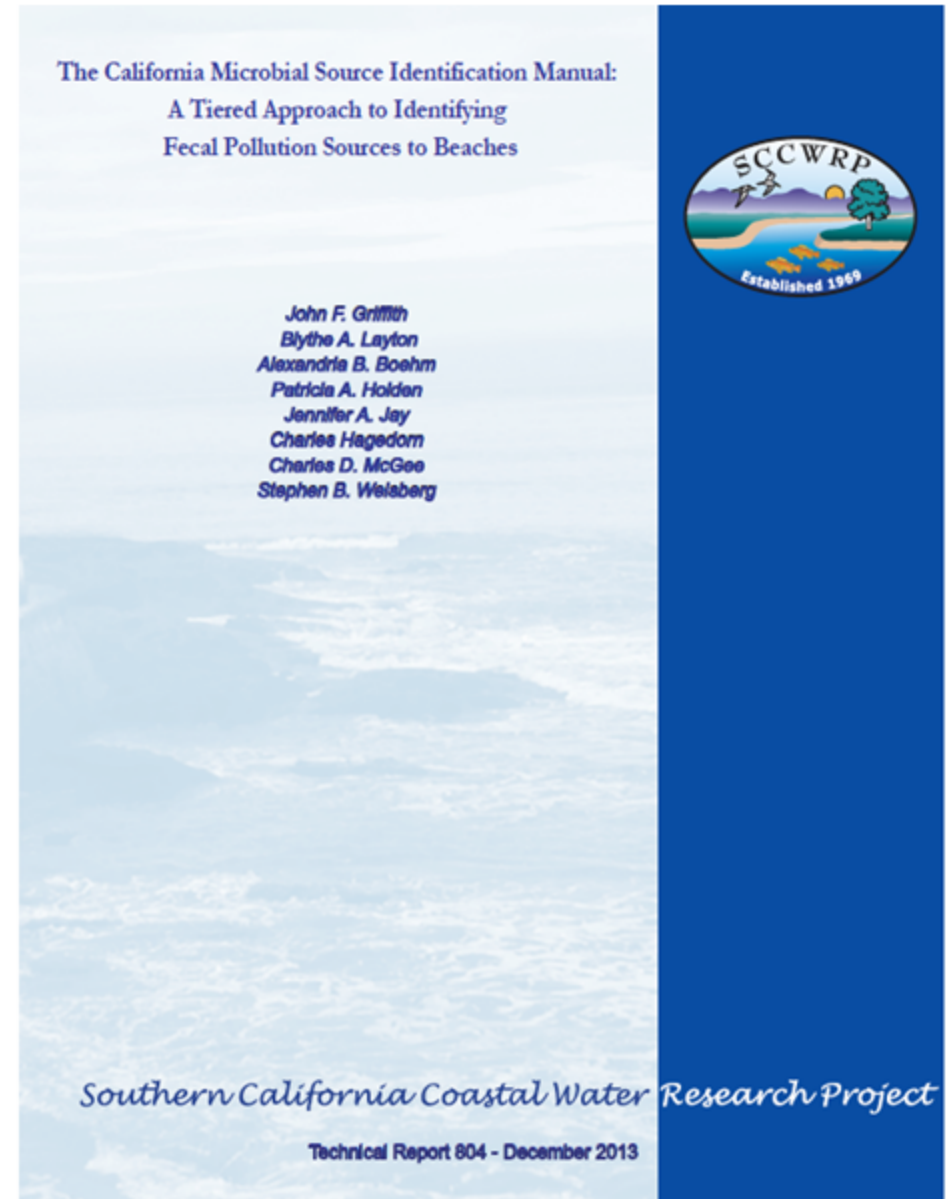
- EPA allows two sampling approaches to determine bacterial water quality
 - Single sample
 - Short-term assessment
 - Can miss intermittent contamination
 - Rolling 30-day geometric mean (weekly sampling)
 - Long-term trends
 - Can smooth out short-term increases

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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