Flow Regime
• Velocity
• Volume
• Surface runoff
• Groundwater
• Variability
• High-low extremes

Habitat Structure
• Channel width/depth
• Banks
• Substrate
• Canopy cover
• Riparian vegetation
• Gradient/slope

Energy Source
• Sunlight
• Primary Production
• Secondary Production
• Organic matter inputs
• Nutrient availability

Watershed characteristics

Ecological Integrity of the River

Water Quality
• Turbidity
• Conductivity
• Temperature
• Dissolved Oxygen
• Nitrogen
• Phosphorous
• pH
• Contaminants

Biotic Interactions
• Competition
• Reproduction
• Predation
• Feeding
• Parasitism
• Disease
Outline

• Introduction
  • Microorganisms (abundance, mass, type, size, diversity etc.)
• Microbes in natural watersheds and their significance
  • Microbes in natural watersheds (planktonic and biofilms)
  • Ecological significance
  • Pathogens and potential public health concerns
  • Microbial health in streams and fecal indicator bacteria
• Agriculture and urban impacts
  • Sources (agriculture vs. urban)
  • Prevention and treatment
• Effects and efficacy of remediation and restoration (case studies)
  • BMPs (United Water/Suez)
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Micro-organisms

Microscopic; Too small to be seen by unaided eyes

Living organisms, independent

Relative sizes on a logarithmic scale

0.1 nm 1 nm 10 nm 100 nm 1 µm 10 µm 100 µm 1 mm

Light microscope

Electron microscope

https://courses.lumenlearning.com/microbiology/chapter/types-of-microorganisms/
Individual cells: Cocci-most 0.1-0.2 μm in diameter

*E. coli/Bacillus*: 0.2 μm wide, 1-5 μm long.

A few unusually large cells:

- *Thiomargarita namibiensis* (100-300 μm in diameter)
- *Epulopiscium fishelsoni* (80 μm dia, 200-700 μm long)

Some form filaments, some in sheaths
Different type
High diversity

Bacteria + Archaea: 90 million
vs. Eukaryota: ~8.7 ± 1.3 million

Woese and Fox, 1977; Pace, 1990; Woese et al., 1990; Mora et al. 2011
Earth is a microbial planet

Population size:

- Human: $7.73 \times 10^9$ (estimated Sep 2019)
- Microbes: $1.2 \times 10^{31}$ (including in both water and soils)
- ~ $1.5 \times 10^{20}$ microbes for every human

Biomass

- Humans (@70 kg) = 0.05 Gt C
- Microbes = ~ 77.2 Gt C
- Microbes “outweigh” humans ~ > 1,500 to 1

Microbes are second largest pool of living C (after plants) and the largest pool of living N and P

Whitman et al. 1998. PNAS. 95:6578-83
Human microbiome: a good analogy

Human is a microbial “body”

Cells in human body: 37.2 trillion; Bacteria: 1-10 times more

Bouslimani et al. 2015
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Microbes living in freshwater

Planktonic (free-floating) vs. Benthic (biofilm); 1 million cells/ml water

Battin et al. 2003
Photosynthetic pigments

Epifluorescence (DNA staining)
Microbes growing in soils; up to 1 billion cells/g dry soil

Biofilms: “microbial skin”
"Biofilms dominate on the surface of the Earth, except in oceans, accounting for ~80% of bacterial and archaeal cells."
Significance in ecosystems

Food web and transformation of dissolved organic matter (microbial loop)

Significance: Transformation of key elements including Carbon, Nitrogen, Phosphorus, Sulfur, etc.
Public Health Concerns

Water-representative taxa and problems

- *Pseudomonas aeruginosa*: ear infection, bathing beaches
- *Clostridium botulinum*: food poisoning
- *Legionella pneumophila*: respiratory infection and death Legionaire’s disease
- *Salmonella, Shigella, Vibrio* et al.: gi (gastrointestinal) tract, diarrhea and dysentery
- *Streptococcus, Vibrio vulnificus* et al.: Necrotizing fasciitis
- *Cryptosporidium, Giardia* etc.
- May cause a variety of diseases

Outbreaks of illness associated with recreational water (2011-2012)
Bacterial monitoring for water quality

- Clean Water Act (CWA): “to restore and maintain the chemical, physical, and biological integrity of the Nation’s waters”
- Impaired waters and TMDL (Total Maximum Daily Load) program
- Among all the TMDLs (including nutrients, sediments etc.), microbial contaminants (e.g. pathogenic bacteria) are ranked No. 1 causes for water quality degradation (U.S. EPA).

(39% rivers and streams, 13% of lakes, reservoirs and ponds; 30% of assessed bays and estuaries)
Impaired waterways in US

Number of impaired waterways by state

- 1 - 100:
- 101 - 500:
- 501 - 1,000:
- 1,001 - 5,000:
- 5,001 or more:

EPA Water Quality Assessment, National Summary
A: The Brandywine-Christina Basin includes Brandywine, White Clay, Red Clay and Christina river subwatersheds (adapted from Water Resource Agency, Univ. of Delaware). B: Stream segments impaired by bacteria (highlighted in red) by PA DEP and DNREC (adapted from U.S. EPA Brandywine-Christina Basin nutrients and bacterial TMDL).
Public health agencies have used total coliforms and fecal coliforms as indicators since 1920s
- non-fecal origin bacterial groups
- coliforms can regrow in natural environments
- Still being used in many states and agencies

_E. coli_ and _Enterococcus_
- More specific bacterial groups
- Commonly used in these days
- Recommended by US EPA (2012)
What bacteria should I monitor?

Depends on what you want to know; Consumption vs. recreation

• Health risk from recreational water contact:
  
  Best indicators in freshwater: *E. coli* and *Enterococci*; for salt water, *Enterococci*

• Water supply or meets state water quality standards
  
  Total and/or fecal coliform
State Bacteria Monitoring

Water contact (WC): Swimming season- *E.coli* and fecal coliform; non-swimming season- fecal coliform.

Drinking Water supply (PWC): Total coliform

Total and fecal coliform

Primary: *E.coli* and *Entero*
Secondary: Fecal coliform

*E.coli* and *Entero*
E. coli monitoring at WCC

WCCLab E. coli Baseflow Samples
E. coli monitoring at WCC

WCCLab E. coli Storm Samples
Enterococci monitoring at WCC
Enterococci monitoring at WCC
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Agriculture and urban

Urbanization

Animal operation

Forested

Wastewater treatment plant

Agriculture-crop
Source of bacterial contaminants

Point and non-point sources
Sewage disposal systems: sewer breaks, sewer overflows, and sewer misconnections; on site septic systems

Agriculture: animal waste runoff, manure storage, vegetative buffer strips; livestock

Stormwater runoff: impervious surfaces, lacking catch basins and settling basins, inappropriate landscaping

Wildlife: birds and small mammals; direct contact or watershed runoff; *Giardia*, *Cryptosporidium*, *Salmonella*, *Campylobacter*, *E. coli* etc.
Water treatment

Water Treatment Process

Raw Water → Coagulation → Flocculation → Sedimentation → Disinfection → Treated water storage → Home Consumption

Coagulants
Water treatment

Coagulation

http://www.chemistry.wustl.edu/~edudev/LabTutorials/Water/PublicWaterSupply/PublicWaterSupply.html
Filtration

http://www.chemistry.wustl.edu/~edudev/LabTutorials/Water/PublicWaterSupply/PublicWaterSupply.html
Water treatment

Disinfection

http://techalive.mtu.edu/meec/module03/Sources-SurfaceWater.htm
Disinfection has proven effective and efficient against bacteria and enteric viruses, but protozoa such as *Giardia* and especially *Cryptosporidium* are very resistant to chlorination alone!

The most important and cost effective protection for water suppliers is to prevent pathogen entry into source water.
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Different approaches and BMPs
Restoration efforts: Pasture management, runoff management, riparian protection, manure management etc.

- Buffer strips
- Constructed/storm water wetland
- Sand filters
- Retention/detention ponds
- Biofiltration
Buffers can reduce bacteria by 43 to 57%, especially in agricultural watersheds.

Biofiltration can reduce >99% of the microbes.

(A. Boyer, DNREC, DE)
Retained/detention ponds can reduce bacteria by 44 to 99%.

Constructed/storm water wetlands can reduce bacteria by 78 to 90%.

(A. Boyer, DNREC, DE)
Case study: United Water/Suez
Case study: United water/Suez

BMP site

Reference site
Keep livestock out of the streams!
Fencing livestock out of streams is a highly effective method of reducing the amount of bacteria in surface waters.
Case study: Yellow Water River

Yellow Water River in Louisiana
- Poor installation/maintenance on on-site treatment systems (septic system etc.)

Approaches
- Thorough inspections on waste water treatment plants and home waste systems
- Additional restoration activities including educational outreach, sewage inspections, and water quality monitoring etc.
Case study: Yellow Water River
Case study: Upper Fishtrap Creek

Lower Nooksack River in Washington
- Exceed 100 CFU/100ml
- FC impairments: state’s CWA section 303(d) list
Case study: Upper Fishtrap Creek

- Nutrient management plans for all dairies
- Fence animals out
- Install hedgerows and filter srips
- On-site inspection and improvement on septic systems
- Farmers growing trees program
Jinjun Kan, PhD
Stroud Water Research Center
jkan@stroudcenter.org
610 268 2153 ext 280
A 30-day period geometric mean

Table 4. Recommended 2012 RWQC.

<table>
<thead>
<tr>
<th>Criteria Elements</th>
<th>Estimated Illness Rate (NGI): 36 per 1,000 primary contact recreators</th>
<th>Estimated Illness Rate (NGI): 32 per 1,000 primary contact recreators</th>
</tr>
</thead>
<tbody>
<tr>
<td>Indicator</td>
<td>GM (cfu/100 mL)</td>
<td>STV (cfu/100 mL)</td>
</tr>
<tr>
<td>Enterococci</td>
<td>35</td>
<td>130</td>
</tr>
<tr>
<td>OR</td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>E. coli</em></td>
<td>126</td>
<td>410</td>
</tr>
</tbody>
</table>
| Duration and Frequency: The waterbody GM should not be greater than the selected GM magnitude in any 30-day interval. There should not be greater than a ten percent excursion frequency of the selected STV magnitude in the same 30-day interval.

*EPA recommends using EPA Method 1600 (U.S. EPA, 2002a) to measure culturable enterococci, or another equivalent method that measures culturable enterococci and using EPA Method 1603 (U.S. EPA, 2002b) to measure culturable *E. coli*, or any other equivalent method that measures culturable *E. coli*. 
# Beach Action Values

## Table 5. Beach Action Values (BAVs).

<table>
<thead>
<tr>
<th>Indicator</th>
<th>Estimated Illness Rate (NGI): 36 per 1,000 primary contact recreators</th>
<th>Estimated Illness Rate (NGI): 32 per 1,000 primary contact recreators</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>BAV (Units per 100 mL)</td>
<td>BAV (Units per 100 mL)</td>
</tr>
<tr>
<td>Enterococci – culturable (fresh and marine)&lt;sup&gt;a&lt;/sup&gt;</td>
<td>70 cfu</td>
<td>60 cfu</td>
</tr>
<tr>
<td><em>E. coli</em> – culturable (fresh)&lt;sup&gt;b&lt;/sup&gt;</td>
<td>235 cfu</td>
<td>190 cfu</td>
</tr>
<tr>
<td><em>Enterococcus</em> spp. – qPCR (fresh and marine)&lt;sup&gt;c&lt;/sup&gt;</td>
<td>1,000 cce</td>
<td>640 cce</td>
</tr>
</tbody>
</table>

<sup>a</sup> Enterococci measured using EPA Method 1600 (U.S. EPA, 2002a), or another equivalent method that measures culturable enterococci.

<sup>b</sup> *E. coli* measured using EPA Method 1603 (U.S. EPA, 2002b), or any other equivalent method that measures culturable *E. coli*.

### TABLE 3

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Symbol</th>
<th>Criteria</th>
<th>Critical Use*</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bacteria</td>
<td>Bac₁</td>
<td>([Fecal coliforms/ 100 ml]) ([<em>Escherichia coli</em>/ 100 ml]) — During the swimming season (May 1 through September 30), the maximum [fetal coliform] E. coli level shall be a geometric mean of [200] 126 per 100 milliliters (ml) based on [a minimum of five] consecutive samples, each sample collected on different days, during a 30-day period. No more than 10% of the total samples taken during a 30-day period may exceed [400] 410 per 100 ml. (Fecal coliforms/ 100 ml)—For the remainder of the year, the maximum fecal coliform level shall be a geometric mean of 2,000 per 100 milliliters (ml) based on a minimum of five consecutive samples collected on different days during a 30-day period.)</td>
<td>WC</td>
</tr>
<tr>
<td>Coliforms/100 ml — Maximum of 5,000/100 ml as a monthly average value, no more than this number in more than 20% of the samples collected during a month, nor more than 20,000/100 ml in more than 5% of the samples.</td>
<td>PWS</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
### Delaware

#### 4.5.7 Bacterial Water Quality Criteria

#### 4.5.7.1 Primary and Secondary Contact Recreation Waters:

The following criteria shall apply:

<table>
<thead>
<tr>
<th>Waterbody Type</th>
<th>Single-Sample Value (Enterococcus Colonies/100ml)</th>
<th>Geometric Mean (Enterococcus Colonies/100ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Primary Recreation Fresh Waters</td>
<td>185</td>
<td>100</td>
</tr>
<tr>
<td>Primary Recreation Waters</td>
<td>104</td>
<td>35</td>
</tr>
<tr>
<td>Secondary Recreation Fresh Waters</td>
<td>925</td>
<td>500</td>
</tr>
<tr>
<td>Secondary Recreation Waters</td>
<td>520</td>
<td>175</td>
</tr>
</tbody>
</table>

703.4 Water quality standards for coliforms.

Total and fecal coliform standards for specific classes are provided in this section.

(a) Total coliforms (number per 100 ml).

<table>
<thead>
<tr>
<th>Classes</th>
<th>Standard</th>
</tr>
</thead>
<tbody>
<tr>
<td>AA</td>
<td>The monthly median value and more than 20 percent of the samples, from a minimum of five examinations, shall not exceed 50 and 240, respectively.</td>
</tr>
<tr>
<td>A, B, C, D, SB, SC, I, SC</td>
<td>The monthly median value and more than 20 percent of the samples, from a minimum of five examinations, shall not exceed 2,450 and 5,000, respectively.</td>
</tr>
<tr>
<td>SA</td>
<td>The median most probable number (MPN) value in any series of representative samples shall not be in excess of 70.</td>
</tr>
<tr>
<td>A-Special</td>
<td>The geometric mean, of not less than five samples, taken over not more than a 30-day period shall not exceed 1,000.</td>
</tr>
<tr>
<td>GA</td>
<td>The maximum allowable limit is 50.</td>
</tr>
</tbody>
</table>

(b) Fecal coliforms (number per 100 ml).

<table>
<thead>
<tr>
<th>Classes</th>
<th>Standard</th>
</tr>
</thead>
<tbody>
<tr>
<td>A, B, C, D, SB, SC, I, SC</td>
<td>The monthly geometric mean, from a minimum of five examinations, shall not exceed 200.</td>
</tr>
<tr>
<td>A-Special</td>
<td>The geometric mean, of not less than five samples, taken over not more than a 30-day period shall not exceed 200.</td>
</tr>
</tbody>
</table>

(c) The total and fecal coliform standards for classes B, C, D, SB, SC and I shall be met during all periods:

1. when disinfection is required for SPDES permitted discharges directly into, or affecting the best usage of, the water; or
2. when the department determines it necessary to protect human health.

8 CRR-NY 703.4
Current through September 15, 2016
A. Criteria for Class I Waters — Water Contact Recreational and Protection of Nontidal Warmwater Aquatic Life.

(1) Bacteriological.

(a) Table 1. Bacteria Indicator Criteria for Frequency of Use.

<table>
<thead>
<tr>
<th>Indicator</th>
<th>Freshwater (Either apply)</th>
<th>All-Area</th>
<th>Single Sample Maximum Allowable Density</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Frequent Full Body Contact Recreation (Upper: 75% CL)</td>
<td>Moderate</td>
<td>Occasional Full Body Contact Recreation (Upper: 50% CL)</td>
</tr>
<tr>
<td>Enterococci</td>
<td>33</td>
<td>61</td>
<td>78</td>
</tr>
<tr>
<td>E. coli</td>
<td>126</td>
<td>235</td>
<td>258</td>
</tr>
<tr>
<td>Marine water</td>
<td>Enterococci</td>
<td>35</td>
<td>104</td>
</tr>
</tbody>
</table>

CL = confidence level

All numbers are counts per 100 milliliters

(b) In freshwater for E. coli, the following formula is used to calculate the upper 75 percent confidence interval for single sample maximum allowable density: antilog(log(126) + 0.675 * log(SD)).

c) In freshwater for enterococci, the following formula is used to calculate the upper 75 percent confidence interval for single sample maximum allowable density: antilog[[log(33)] + 0.675 * log(SD)], where log(SD) is the standard deviation of the log transformed E. coli or enterococci data. If the site data are insufficient to establish a log standard deviation, then 0.4 is used as the log standard deviation for both indicators. At the default log standard deviation, the values are 235 for E. coli and 61 for enterococci.

d) In saltwater, for enterococci, the following formula is used to calculate the upper 75 percent confidence interval for single sample maximum allowable density: antilog[[log(35)] + 0.675 * log(SD)], where log(SD) is the standard deviation of the log transformed enterococci data. If the site data are insufficient to establish a log standard deviation, then 0.7 is used as the log standard deviation. At the default log standard deviation, the value is 104.
1. Bacterial quality (Counts/100 ml)
   i. Shellfish Harvesting: Bacterial Indicators shall not exceed, in all shellfish waters, the standard for approved shellfish waters as established by the National Shellfish Sanitation Program as set forth in its current manual of operations.

ii. Primary Contact Recreation:
   (1) Enterococci levels shall not exceed a geometric mean of 35/100 ml, or a single sample maximum of 104/100 ml.
   (2) E. Coli levels shall not exceed a geometric mean of 126/100 ml or a single sample maximum of 235/100 ml.

iii. Secondary Contact Recreation:
   (1) Fecal coliform levels shall not exceed a geometric mean of 770/100 ml.
   (2) Fecal coliform levels shall not exceed a geometric mean of 1500/100 ml.

Shellfish Waters
SE1 and SC
All FW2
SE2
SE3