Increasing Occurrence of High Fecal Indicator Bacteria (FIB) in Headwater Streams within the Lower Delaware River Watershed

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Excess sediments, nutrients and pathogenic microorganisms (USEPA).
What Bacteria are monitored

- Total coliform ➔ Fecal coliform ➔ *E. coli / Enterococcus*

- 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
  - Currently are commonly used
  - Recommended by US EPA (2012)
EPA approved

Most Probable Number (MPN) based approaches

Total coliform, *E. coli* and *Enterococci* (Colilert, Enterolert from IDEXX)
Sampling at DRW sites

Clusters Sampled
- Brandywine and Christina
- Middle Schuylkill
- New Jersey Highlands
- Poconos and Kittatinny
- Upper Lehigh
- Upstream Suburban Philadelphia
# High variability between samples

* Values outside of IDEXX readable range (>24196 MPN counts/100mL)
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Exploratory Analyses

Correlation hunting

Watershed size

Land cover – 2013 data from University of Vermont Spatial Analysis Lab
E. coli with watershed size

R (p-value): Pearson=-0.10 (0.262); Spearman=-0.40 (0.000)
Manifestation of Entero with watershed size

R (p-value): Pearson=-0.19 (0.028); Spearman=-0.71 (0.000)
Relationships with land uses

- Forest %
- Agriculture (Low vegetation %, mostly Ag but lawns, nursery plantings etc.)
- Developed %, structures and other impervious surfaces
Log₁₀(E. coli MPN counts/100 ml)

Land Cover %

E. coli vs. forest (%)

R (p-value): Pearson = -0.18 (0.042); Spearman = -0.47 (0.000)
Entero vs. forest (%)

$R$ (p-value): Pearson = -0.22 (0.028); Spearman = -0.48 (0.000)
E. coli vs. Agriculture (%)

R (p-value): Pearson=0.18 (0.038); Spearman=0.48 (0.000)
Entero vs. Agriculture (%)

R (p-value): Pearson=0.28 (0.001); Spearman=0.52 (0.000)
No significant correlations have been observed for fecal indicator bacteria (total coliform, E. coli and Enterococcus) with developed %, structures and other impervious surfaces YET.
**E. coli versus Enterococcus**

R (p-value): Pearson=0.40 (0.000); Spearman=0.7 (0.000)

Regression Equation:

\[ \text{valu1} = 227.5 + 1.124 \times \text{valu2} \]
Potential Sources

- Waste water treatment plants: sewer breaks, sewer overflows, and sewer misconnections
- On-site septic systems
- Human feces
- Livestock
- Pet waste and wildlife
- Storm runoff
Three molecular source tracking methods at Stroud: ITS-DGGE, community fingerprinting/sequencing, DNA-based host-specific PCR approach

Method presented here: bacterial host-specific genes (e.g., *Bacteroides* sp. 16S rRNA gene sequences) qPCR approach
Relative abundance of signal normalized by DNA quantity

- Human
- Human
- Cow
- All bovine

**Species**:
- HF183
- HumM2
- CowM2
- BacB2590
Summary

• Most sites had *E. coli* and *Enterococcus* concentrations that greatly exceeded EPA/PA DEP criteria.

• Total coliform, *E. coli*, and *Enterococcus* concentrations were highest in headwater streams but “diluted out” at downstream sites.

• Moderate/strong relationships with watershed size and land cover (e.g. forest%, agriculture% etc.), but no correlations with developed% have been observed.

• Source tracking is necessary to elucidate potential/probable sources of fecal contamination.
Future perspectives

- Why are the counts so high for both *E. coli* and *Entero* during the summer? Where are they coming from?

- What caused high variations? temperature, land uses, local sources, wildlife?

- Are *E. coli* and *Entero* good indicators for headwaters? Microbiomes in water column and on streambeds?

- How long can FIB survive/travel through the watershed?
Key: continuous monitoring and more data/work!


Thank you!
### 2012 EPA Recommended Water Quality Criteria (RWQC)

#### A 30-day period geometric mean

<table>
<thead>
<tr>
<th>Criteria Elements</th>
<th>Option 1</th>
<th></th>
<th>Option 2</th>
<th></th>
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<tbody>
<tr>
<td></td>
<td>Est. illness rate = 36/1,000</td>
<td>≤10% Threshold Value (CFUs/100 ml)</td>
<td>Est. illness rate = 32/1,000</td>
<td>≤10% Threshold Value (CFUs/100 ml)</td>
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<tr>
<td>Indicator</td>
<td>Geometric Mean (CFUs/100 ml)</td>
<td>Geometric Mean (CFUs/100 ml)</td>
<td>Geometric Mean (CFUs/100 ml)</td>
<td>Geometric Mean (CFUs/100 ml)</td>
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<tr>
<td>Enterococci (marine and fresh)</td>
<td>35</td>
<td>130</td>
<td>30</td>
<td>110</td>
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<tr>
<td>E. coli (fresh)</td>
<td>126</td>
<td>410</td>
<td>100</td>
<td>320</td>
</tr>
</tbody>
</table>

PA DEP Recommendation
For recreational contact season (1 May – 30 Sept)