Pathogen Program: Molecular Tools for Environmental Management

12.2.2015

Microbial Source Tracking
A set of techniques used to determine the sources of fecal indicator bacteria in the environment.

**Microbial Source Tracking**

**State of the Science**

<table>
<thead>
<tr>
<th>RATIOS</th>
<th>CHEMICAL/ BIOCHEMICAL</th>
<th>LIBRARY DEP. METHODS</th>
<th>LIBRARY INDEP. METHODS</th>
<th>NEXT GENERATION SEQUENCING</th>
</tr>
</thead>
<tbody>
<tr>
<td>FC:FS</td>
<td>Optical Brighteners</td>
<td>PFGE</td>
<td>Bacteriophage</td>
<td>Illumina</td>
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<td></td>
<td>Ribotyping</td>
<td>Viral PCR</td>
<td>Roche 454</td>
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<td>Rep-PCR</td>
<td>Viral qPCR</td>
<td>Ion Torrent</td>
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<td>Antibiotic Resistance</td>
<td>Bacterial PCR</td>
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<td>Bacterial qPCR</td>
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host-specific qPCR
HRSD formed to protect public health
  – Contaminated oysters were a serious problem

Vision: Future generations will inherit clean waterways and be able to keep them clean

Commission approved Pathogen Program in June 2015
Focus:
1. **Microbial Source Tracking Studies**
2. **Pathogen/Virus Quantification Support**

Program Mission: To partner with localities to focus source identification efforts

Project Types: infrastructure identification, spill delineation, sewage presence confirmation
### HRSD’s MST Capabilities

<table>
<thead>
<tr>
<th>Currently</th>
<th>In the Works</th>
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<tbody>
<tr>
<td>• Fecal <em>Bacteroides</em> spp.</td>
<td>• Other sources</td>
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<tr>
<td>• HF183</td>
<td>- Gull</td>
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<tr>
<td>• Human Polyomavirus (HPyV)</td>
<td>- Dog</td>
</tr>
<tr>
<td>• EPA’s HumM2 technology</td>
<td>- Geese</td>
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<tr>
<td></td>
<td>• Pathogens</td>
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<tr>
<td></td>
<td>- Norovirus (G1 &amp; 2)</td>
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<td></td>
<td>- Adenovirus</td>
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<td>- Hepatitis A Virus</td>
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### Program Logistics

- **Partnership:** locality must commit resources to put “boots on the ground”
- **Cost:** HRSD pays for sampling and analyses
- **Contract:** Model agreement will be drafted to guide future work
- **Prioritization:** Highest priorities → chronic bacterial exceedances, public health risk
  - Project prioritization: ?
Wayne Creek: A Microbial Source Tracking Case Study

Chronic Contamination
Elizabeth River Project Monitoring

**Wet Weather:**
- **Enterococcus cells/100 mL**: 4110

**Dry Weather:**
- **Enterococcus cells/100 mL**: 2010
Novel Approach to Understand Causes

1. Alternative indicators
2. Targeted in-pipe sampling
Sampling Design

- Downstream to upstream SW pipe network investigation
- Every SW tributary intersection and outfall
- Narrow contamination origin to manageable area
Sampling Design

Sampling design based on infrastructure knowledge

City of Norfolk identified homes still using septic system

Results
**7/9/14—southern segment**

- Active septic systems—no human signal
- Evidence of northern delivery route

**9/4/14—northern segment**

- Human fecal signal found along one stormwater pipe segment
Higher spatial resolution

Human contamination signal delineated— 'hot spot' identified

City of Norfolk found a leaking sewer main
11/3/14—post repair

- Water quality improved post repair
- 2 areas of residual signal
- Areas of minimal flushing

3/19/15—post repair

- Excellent water quality:
- Low Enterococcus concentrations
- No human fecal markers detected
Questions?

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