Microbial Source Tracking with qPCR: Technology Transfer and Research

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Presentation Overview

1. Microbial Source Tracking Background

2. Technology Transfer Support

3. Research Activities

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Fecal Pollution is a Nationwide Challenge

- Fecal microbes most common biological contaminant
- Public health, economic, and ecological impacts

Top10 Causes of Impairment in U.S. Rivers and Streams

- Pathogens
- Sediment
- Nutrients
- Oxygen Depletion
- Temperature
- Metals
- Polychlorinated Biphenyls
- Mercury
- Habitat Alterations
- Turbidity

Miles of Impaired Water

https://ofmpub.epa.gov/waters10/attains_nation_cy.control#causes
EPA Responsibilities

Protect and Restore Waters for Recreational Use
- Clean Water Act 1972

Risk Assessment of Beach Contaminants
- BEACH Act (2000)
- Development of new or revised ambient water quality criteria (AWQC)

Management of Point and Non-Point Pollution Sources
- Total Maximum Daily Load (TMDL) programs
- National Pollutant Discharge Elimination System (NPDES) programs
- National Estuary Program (NEP)
- Combined Sewer Overflow (CSO) consent decrees
Common Fecal Pollution Management Tools

- Based on **general fecal indicators**
- Measure of total fecal pollution
- Presence in water is a warning signal of public health risk
- Do not discriminate between sources
Source of Fecal Pollution is Important

- Public health risk can vary by source
- Mitigation strategies can vary by source
- Source information improves management and public safety
A Microbial Source Tracking qPCR Solution

SOLUTION ... Method designed to collect, isolate, identify, and quantify a host-associated genetic marker from an environmental sample
Many Water Quality Management Applications

- Recreational beach management
- Evaluation of a best management practices
- Urban stormwater management
- Hazardous event response
- Waterborne disease outbreak response
Technology Transfer Support:
Draft EPA Methods 1696 and 1697 Available

• Human fecal pollution qPCR
• Released April 2019

Content Overview:
- Safety
- Laboratory organization
- Equipment, reagents, and supplies
- Sample collection, handling and storage
- Standardized laboratory procedures
- Quality controls
- Data analysis and calculations

Method 1696: Characterization of Human Fecal Pollution in Water by HF183/BacR287 TaqMan® Quantitative Polymerase Chain Reaction (qPCR) Assay

https://www.epa.gov/cwa-methods/other-clean-water-act-test-methods-microbiological
Technology Transfer Support: Automated Data Analysis Tool

- Simplify complex calculations
- Ensure standardized analysis
- Implement data acceptance metrics
- Concentration estimates with error

- Microsoft Excel
- Summary report
Technology Transfer Support: Self-Administered Method Proficiency Tool

- Successfully complete:
  - Prior to environmental sample testing

- Six metrics based on:
  - National laboratory validation
  - Reagent manufacturer recommendations
  - qPCR experts

- Training and management tool
Technology Transfer Support: Reference DNA Material Development

- National implementation requires a high quality reference DNA material
- Standardized and centralized source
- Not feasible for EPA to manufacture and distribute
- Interagency Agreement with National Institute of Standards and Technology
Technology Transfer Support: Technical Support and Collaboration

- **Strategic Partnership with APHL:**
  - Memorandum of Understanding (Jan 2017)
  - Enhance response to contamination events

- **Technical Support and Outreach:**
  - Public presentations
  - Regional and State workshops
  - Trained EPA Regional Lab staff

- **Research Collaborations:**
  - States, tribes, and other local communities
  - Federal agencies
Research Activities:
Large-Scale qPCR MST Implementation

Challenge: Are there spatial and temporal patterns to fecal pollution in my watershed?

- Partners:
  - OR Dept of Environmental Quality
  - OR Dept of Agriculture
  - EPA Region 10 Lab
- 1,500 km² Oregon coast
- 29 sites
- Routine sampling (n = 696)
- 8 MST qPCR methods
- E. coli testing

Research Activities:
MST qPCR and Recreational Beach Monitoring

Challenge: Find links between MST qPCR and general indicator measurements in recreational water setting?

- University of Illinois at Chicago School of Public Health
  - Sam Dorevitch (Principal Investigator)
  - Abhilasha Shrestha (PhD Candidate)
- 9 beaches sampled 5 days/week over beach season
- Enterococci and *E. coli* testing
- MST qPCR testing for human, bird, and dog sources
Research Activities: Urban Stormwater Management

Challenge: What are the sources of fecal pollution in my MS4 outfalls?

- Partners:
  - DC Department of Energy & Environment
  - EPA Region 3 Lab

- 7 catchments
- 32 MS4 outfalls
- Routine & event sampling
- Potential pollution sources (human, ruminant, bird and dog)
Research Activities: Method Development

• New Tools:
  - Cattle
  - Pinnipeds

• Performance Testing:
  - Canine
  - CrAssphage
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