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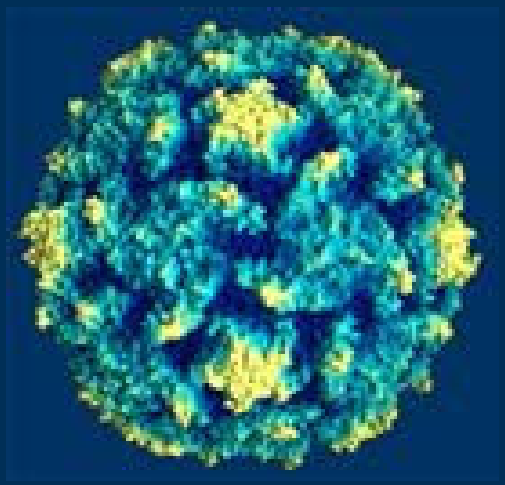
Enteric Viruses in Ground Water

Trisha Johnson



Poliovirus
www.virology.net

U.S. Department of the Interior
U.S. Geological Survey

Presentation Outline

- Background on enteric viruses
- Sampling procedure
- Detection methods
- Costs and limitations
- Case study
- Conclusions

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What are enteric viruses?

- Infect gastrointestinal tract
  - Gastroenteritis
  - Colds and respiratory tract infections
  - Hand, foot, and mouth disease
  - Myocarditis
  - Liver damage
  - Aseptic meningitis
  - Death
- Transmitted by fecal-oral route
  - 10^8 viruses per gram feces
  - < 10 virus particles to infect
  - Foodborne
  - WATERBORNE

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Enteric viruses, continued…

- Sources of viruses to ground water:
  - septic systems, leaking sewer lines, land application of wastes, livestock operations, injection wells, landfill leachates, unintentional wastewater plant overflows, and treated wastewater effluent

- Transport and Survival Characteristics
  - Small size (20 nm-350 nm)
  - Differences from bacteria and protozoa
  - Survive for weeks to months (cold temperatures)
  - Travel > 1 km from source
  - Resistance to chlorination

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Why should we be concerned about enteric viruses in ground water?

- CDC report from 1971-1996
- 58% of waterborne disease outbreaks in U.S. associated with ground water systems
  - 86% - source water contamination
  - 24% - enteric viruses
  - Unknown etiology – enteric viruses suspected
- Georgetown, TX 1980 – 7,900 people ill
  - Coxsackievirus and Hepatitis A virus in source water from a karst aquifer
- Endemic waterborne disease
- ICR Rule – 24% of source waters

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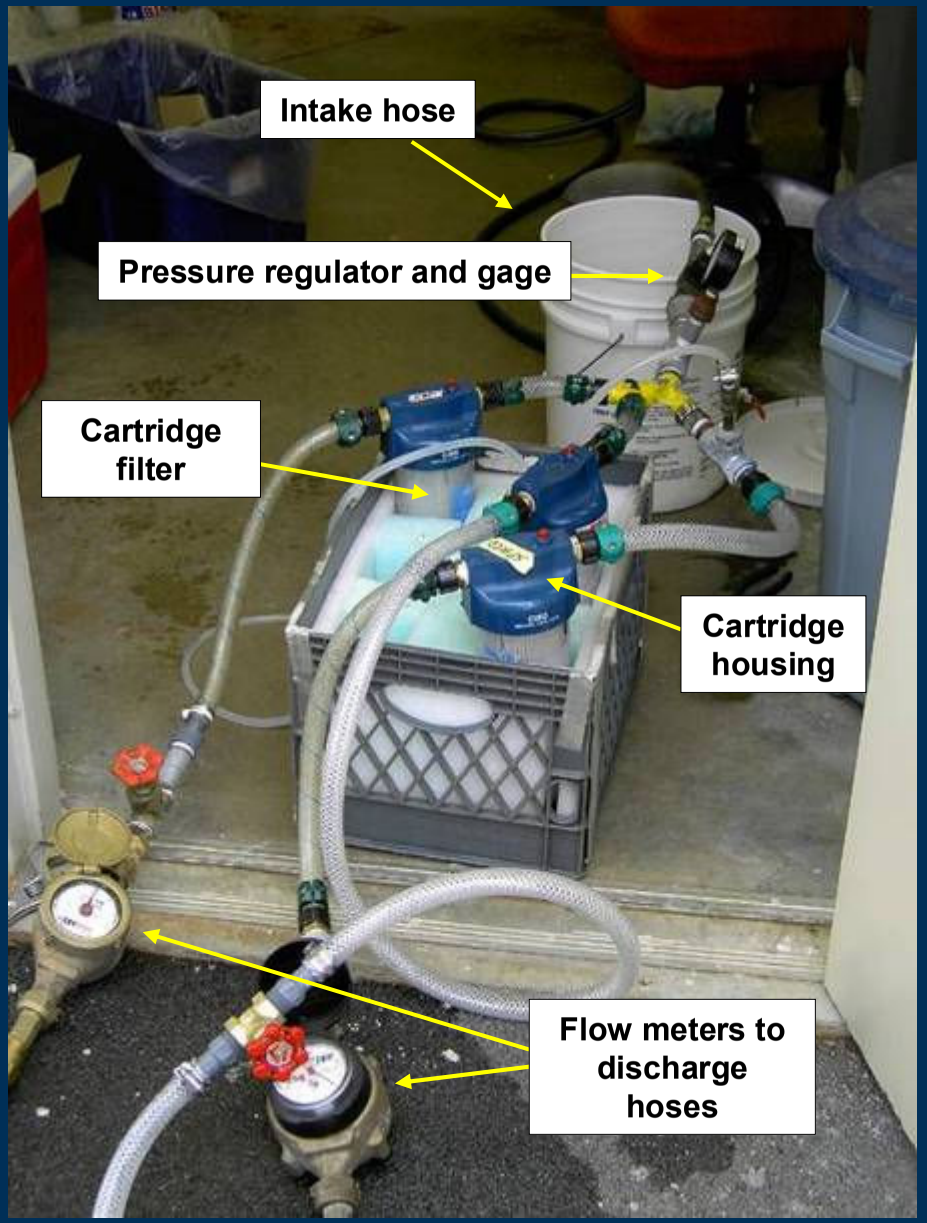
Sampling Procedure

- Sterilize apparatus
- Filter up to 2000 liters
- Field Parameters
  - Turbidity
  - pH
- Store at 4ºC
- 72-hour holding time



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Sample Processing

- Filter elution
- Concentration
- Post concentration and inhibitor removal for molecular analyses

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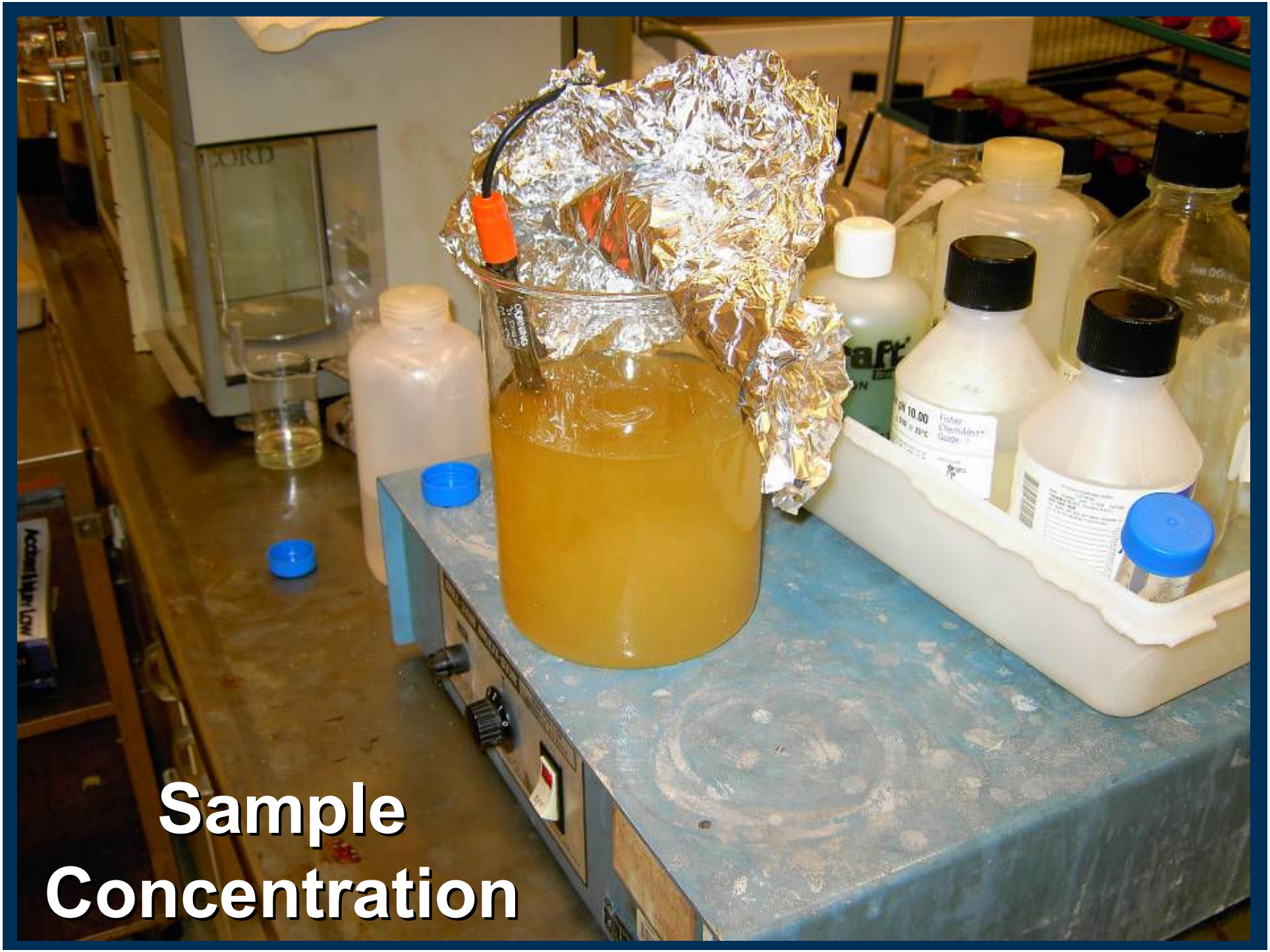
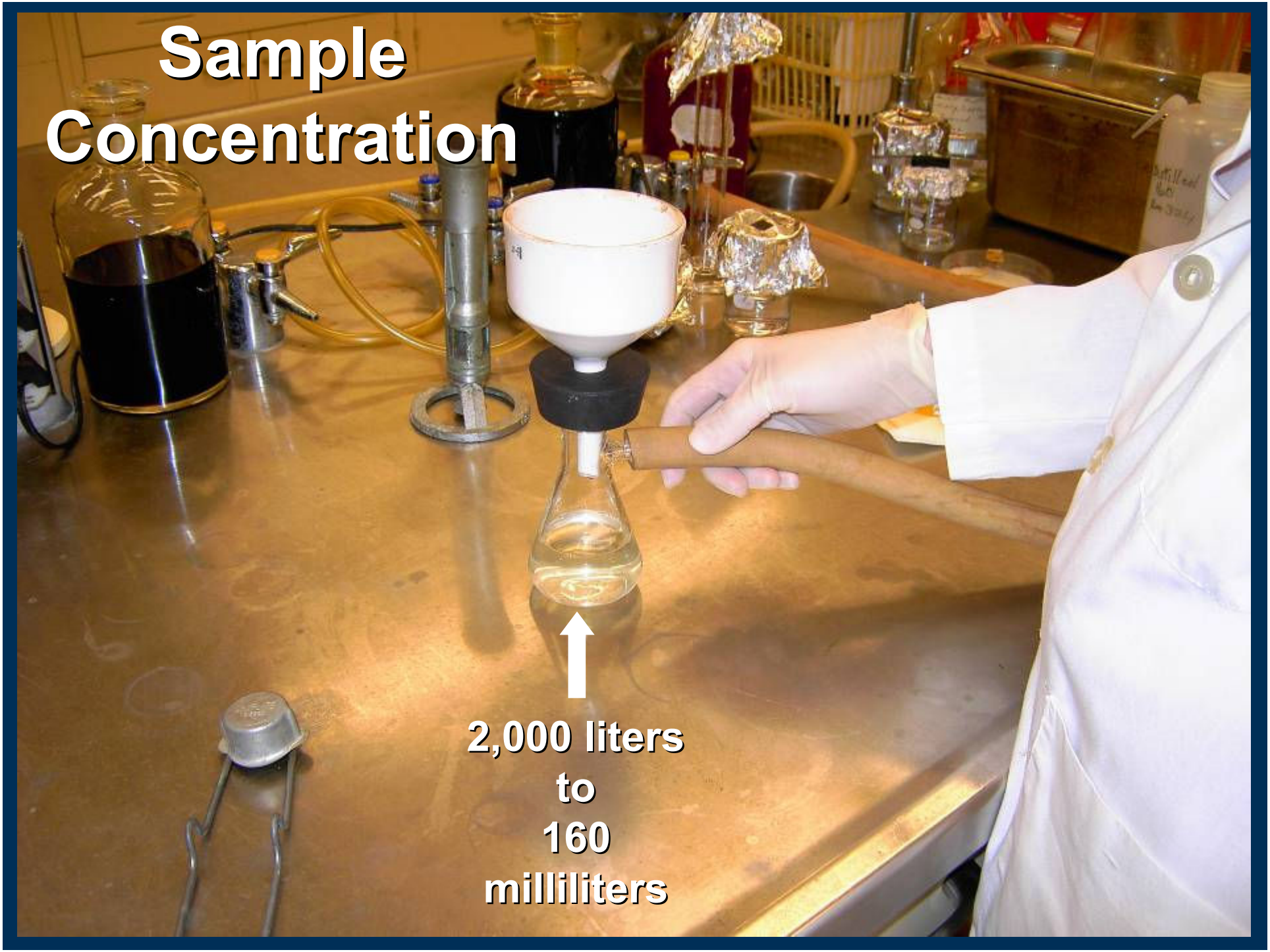
Filter Elution


Sample
Concentration


Sample
Concentration


Sample Concentration
2,000 liters to 160 milliliters

Sample Cleanup

Analysis Methods – Cell Culture

- "Total Culturable Virus" Assay
  - Inoculate thin layer of BGMK cells with concentrated water sample, analyzed microscopically
- Advantages
  - Large volume of sample (25%) is analyzed
  - Detects infectious enteric viruses
  - Low detection limit – 1 infectious virus
- Disadvantages
  - Not specific for any virus
  - Not all viruses can be detected
  - Takes a few to several weeks for results

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Analysis Methods – RT-PCR

- Reverse - Transcription Polymerase Chain Reaction
  - Molecular method - detects presence of viral RNA
- Advantages
  - Detect specific viruses (HAV, Norwalk, Rotavirus)
  - Results in a few days
- Disadvantages
  - Small sample volume (2.5%)
  - Detects infectious and noninfectious viruses
  - Prone to false negatives from inhibition

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Cost

- Up to $2,000 per sample
- Strict lab requirements
- Options for Analysis
  - Federal and State Agencies
  - Universities
  - Commercial Labs

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Limitations

- Monitoring frequency
- Sample volume
- Analyses have limitations
- QA/QC samples

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Case Study – PWS Wells and Springs in Karst Aquifers in East Tennessee

- Sampled PWS serving 750 to 55,000 people
- Karst aquifers under base flow conditions
- 88% of PWS and 71% of samples positive for infectious enteric viruses by cell culture
- Only 29% of PWS were positive for enteric viruses each time they were sampled
- No samples positive for enteric viruses by RT-PCR
- 2 sites always negative for indicator bacteria

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Conclusions

- Enteric viruses are a major public health concern
- Important to focus on source water as a multi-barrier approach
- Sampling only for indicators or one type of pathogen in source waters does not give the complete picture
- Sampling one time for viruses is only a snapshot
- Sources of information
  - Ground Water Rule
  - ICR Rule

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