

# Tips to Support Microbiological Proficiency Testing

## Agenda

- Drinking Water Microbiology Proficiency Testing
  - P/A, HPC, Quantitative Coliforms
    - Understanding the Design and Evaluation Criteria
    - Participants Common Errors
    - Tips for Success
- Waste Water Microbiology Proficiency Testing
  - Quantitative Coliforms and Enterococci
    - Understanding the Design and Evaluation Criteria
    - Participants Common Errors
    - Tips for Success
- Available Resources
- Questions

- Presence/Absence Microbiology
  - Required sample design and evaluation criteria
    - Set of 10 samples composed of 2-4 E. coli, 2-4 Enterobacter species, 1-2 Pseudomonas or other species, 1-2 containing no microorganism
    - If doing multiple methods within same study, must have unique sample set for each method
    - Participant can report presence or absence for each specific analyte(Total/Fecal/E.coli) for each of the 10 samples. Each specific result is evaluated individually as acceptable or not acceptable. Overall evaluation 9 out of 10 with no false negatives.
    - Concentration range not defined and PTRL are not applicable

Presence/Absence Microbiology

#### Common Errors

- Labelling or reporting issues when analyzing multiple series
- Poor Quality Media or Inhibitory Effects of Equipment producing false negatives
- Using buffer bottles as test vessels and misinterpreted results
- False Positives on Fecal for Enterobacter cloacae using SM9221
- Reporting analytes not appropriate for a given method
- Not following normal procedure when analyzing PT's...skipping steps.

- Presence/Absence Microbiology
  - Tips for Success
    - If running multiple series...keep them straight. Review bench work to confirm sets were labelled and recorded correctly. When entering or reporting results, have 2<sup>nd</sup> review of entry.
    - Perform all appropriate quality control on method/media prior to running samples
    - Phosphate buffer bottles are not intended to be a substitute for your normal sample bottles
    - If doing SM9221 using EC media for determination of Fecal, outgas media after media has reached incubation temperature to avoid false positives.
    - You must report values for Total Coliforms, Fecal Coliforms and E. coli for each analyte that you are seeking accreditation.
    - Only report analytes appropriate for method being used.
    - Perform all confirmation step required by procedure/method
    - Enzyme substrate methods, don't forget to add media, have process to confirm addition
    - Follow instructions provided by PT provider for product use and reporting.
    - Always use comparators for Chromogenic/Fluorescence methods when available.

- Heterotrophic Plate Count
  - Required sample design and evaluation criteria
    - Single quantitative sample containing Heterotrophic Bacteria in the manufacturing range of 5-500 CFU/mL
    - Analytes are divided into two technologies
      - Methods that determine HPC by MF or PP
      - Methods that determine HPC by MPN
    - Acceptance Criteria is Log Transformed Mean ±2SD
    - PTRLs are 2 CFU/mL

- Heterotrophic Plate Count
  - Common Errors
    - Reporting analyte not appropriate for method, switching target analytes or technologies
    - Not running appropriate dilutions to cover manufacturing range
    - Counting errors
    - Media not at appropriate temperature prior to pouring plate (low bias)
    - Sample not run within 30 minutes after hydration (high bias or low bias)
    - Results not reported to appropriate number of significant figures (PP,SP,MF Whole Number) (MPN 3 SigFig)

- Heterotrophic Plate Count
  - Tips for Success
    - Know which analyte your method determines, report appropriately
    - Determine what dilutions you will run based on the manufacturing range. Make sure potential results will be a manageable number of organisms to count. May want to consider running samples in duplicate, if not current practice.
    - Temperature of media to 44°C prior to pouring plate
    - Be prepared to run the sample with 30 minutes after hydration
    - Perform all appropriate quality control on method/media prior to running samples
    - Follow instructions provided by PT provider for product use and reporting.
    - SimPlate Unit Dose vs. Multi-Dose....use correct table
    - Make adjustment for dilution performed prior to reporting result

- Quantitative Coliforms
  - Required sample design and evaluation criteria
    - Single quantitative sample containing Escherichia coli in the manufacturing range of 20-200 CFU/100mL
    - 3 analytes divided into two technologies
      - Total, Fecal, E. coli by MF
      - Total, Fecal, E. coli by MPN
    - Acceptance Criteria is Log Transformed Mean ±2SD
    - PTRLs are 2 CFU/mL

- Quantitative Coliforms
  - Common Errors
    - Reporting analyte not appropriate for method, switching target analytes or technologies
    - Not running appropriate dilutions to cover manufacturing range
    - Poor Quality Media or Inhibitory Effects of Equipment/Supplies producing false negatives or low bias
    - Sample not run within 30 minutes after hydration (high bias)
    - Results not reported to appropriate number of significant figures (MF - Whole Number) (MPN - 3 SigFig)
    - When running Multiple Tube Methods, number of tubes selected did not cover the manufacturing range

- Quantitative Coliforms
  - Tips for Success
    - Know which analyte your method determines, report appropriately
    - Determine what dilutions you will run based on the manufacturing range. Make sure potential results will be a manageable number of organisms to count based on method being run. Run all sample provided.
    - Be prepared to run the sample with 30 minutes after hydration
    - Perform all appropriate quality control on method/media prior to running samples
    - Follow instructions provided by PT provider for product use and reporting.
    - Make adjustment for dilution performed prior to reporting result

- Quantitative Coliforms & Enterococci
  - Coliform required sample design and evaluation criteria
    - Single quantitative sample containing Escherichia coli in the manufacturing range of 20-2400 CFU/100mL
    - 3 analytes divided into two technologies
      - Total, Fecal, E. coli by MF
      - Total, Fecal, E. coli by MPN
    - Acceptance Criteria is Log Transformed Mean ±3SD
    - PTRLs are 2 CFU/mL

- Quantitative Coliforms & Enterococci
  - Enterococci required sample design and evaluation criteria
    - Single quantitative sample containing Enterococcus faecalis in the manufacturing range of 20-1000 CFU/100mL
    - 1 analyte divided into two technologies
      - Enterococci by MF
      - Enterococci by MPN
    - Acceptance Criteria is Log Transformed Mean ±3SD
    - PTRLs are 2 CFU/mL

Quantitative Coliforms & Enterococci

#### Common Errors

- Reporting analyte not appropriate for method, switching target analytes or technologies
- Not running appropriate dilutions to cover manufacturing range
- Poor Quality Media or Inhibitory Effects of Equipment/Supplies producing false negatives or low bias
- Sample not run within 30 minutes after hydration (high or low bias)
- Results not reported to appropriate number of significant figures (MF - Whole Number) (MPN - 3 SigFig)
- When running Multiple Tube Methods, number of tubes selected did not cover the manufacturing range

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Quantitative Coliforms & Enterococci

#### Tips for Success

- Know which analyte your method determines, report appropriately
- Determine what dilutions you will run based on the manufacturing range. Make sure potential results will be a manageable number of organisms to count based on method being run. Run all sample provided.
- Be prepared to run the sample with 30 minutes after hydration
- Perform all appropriate quality control on method/media prior to running samples
- Follow instructions provided by PT provider for product use and reporting.
- Make adjustment for dilution performed prior to reporting result

#### **Available Resources**

- ERA website
  - NELAC FoPT Tables
  - Webinars
  - CRM's
  - FAQ's
  - Helpful Hints
- Technical Support





#### **Contact ERA**

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