What are the Critical Elements in the Microbiology Laboratory?

Margo E. Hunt, Ph.D.

Part 9000 Coordinator, Microbiology

Standard Methods for the Examination of Water and Waste Waters (SM)

APHA, WEF, AWWA

Daubert vs. Merrell Dow

- Supreme Court decision in 1993
- Trial judges must ensure scientific evidence is relevant and reliable.

Illustrative, not definitive, list:

- Theory or technique can be or has been tested
- Peer review and publication of theory or technique
- Known or potential rate of error
- Existence and maintenance of standards controlling the technique in the scientific community
- General acceptance of the methodology or technique in the scientific community

Quality by Design (QBD)

- FDA term – process design
- EPA term – quality system

FDA

- Bacteriological Analytical Manual (BAM)
- Microbiological Best Practices, USP <11177>

USP <11177>

Personnel training
Cultures
Media preparation
Laboratory records
Laboratory layout
Interpretation of results

QC testing
Sample Handling
Laboratory equipment
Documentation
Media preparation

EPA

- 1978 Microbiological Methods for Monitoring the Environment, Water and Wastes
- Manual for the Certification of Laboratories Analyzing Drinking Water, fifth edition + 2 supplements

Standard Methods 9020

- Discusses critical elements in the microbiology laboratory and for analytical methods
- Basic Quality Assurance and Quality Control elements with minimal frequency of QC checks for most processes.

Standard Methods 9020A

- Discusses importance of Quality Assurance and the Quality Assurance Manual

9020B and 9020C

1. Facility
2. Personnel, both management and staff
3. Equipment and Supplies
4. Documentation
5. Analytical Methods
6. PT/PE (collaborative)Testing

1. Facility

- Ventilation
- Space Utilization
- Laboratory bench areas
- Walls and floors
- Cleanliness
- Electricity

2. Personnel

- Experienced in the science
- Organized
- Takes responsibility
- DOC – initial and on-going for each analyst

3. Equipment and ...

- Temperature sensing devices
- Balances
- Meters
- Autoclaves and ovens
- Incubators
- Etc.

3. Supplies

- Glassware
- Reagent water
- MF filters
- Media – purchase, storage, preparation, storage

4. Documentation

- Record everything
- Write down what you are going to do and how you are going to do it
- Follow what you said you were going to do
- SOPs
- Analytical results

5. Analytical Methods

- Run positive and negative culture controls
- Blanks
- Sterility Checks

5. Analytical Methods

- Establish analyst variability in counting
- Run duplicates at set frequency, e.g., one per batch or test run

5. Analytical Methods

- Verification – established methods
- Validation – new or non-standard methods

5. Analytical Methods

Qualitative Methods – presence/absence

1 Accuracy and precision
2. Specificity/Selectivity
3. Detection Limit
4. Robustness
5. Repeatability

5. Analytical Methods

Quantitative Methods

1. Accuracy
2. Precision/Repeatability
3. Precision/Reproducibility
4. Recovery/sensitivity
5. Detection Limit
6. Upper counting limit
7. Range

6. PE/PT Testing

- Collaborative testing

EPA and 40 CFR 136.7

- EPA's final rule, 2012, "Guidelines Establishing Test Procedures for the Analysis of Pollutants Under the Clean Water Act: Analysis and Sampling Procedures"

12 Critical Quality Control Elements

- These elements must be clearly documented in the written SOP for each analytical method not containing QA/QC procedures,

- Unless a written rationale is provided to explain why these elements are inappropriate.

12 Critical Elements for Chemistry

1. Demonstration of Capability (DOC)
2. Method Detection Limit (MDL)
3. Laboratory reagent blank (LRB), also referred to as a method blank (MB)
4. Laboratory fortified blank (LFB), also referred to as spiked blank or laboratory sample control (LCS)

12 Critical Elements for Chemistry

5. Matrix spike (MS) and matrix spike duplicate (MSD) or laboratory fortified matrix (LFM)
6. Internal standards, surrogate standards, or tracers
7. Calibration (initial and continuing), also referred to as initial calibration verification (ICV) and continuing calibration verification (CCV)
8. Control charts or other trend analysis of QC results

12 Critical Elements for Chemistry

9. Correction active (root cause analysis)
10. QC acceptance criteria
11. Definitions of preparation and analytical batches that may drive QC frequencies
12. Minimum frequency for conducting all QC elements

Are these terms applicable to microbiology?

- Some QC elements are universal, but not all chemistry QC elements are applicable to microbiology.

Are these terms applicable to microbiology?

- Microbiology has greater variability in analytical testing:

  analyst
  calibration of equipment
  materials used
  storage conditions
  equipment used
  environment
  sample matrix

  The microorganisms themselves!

AOAC – Micro. committee

- Patsy Root, Senior Regulatory Affairs Specialist, IDEXX
- Laurie Kundrat, Technical Support Specialist, Microbiologics
- Karla I. Ziegelmann-Fjeld, Technical Manager, Microbiologics
- Margo Hunt, Part 9000 Coordinator, Standard Methods

10 Critical Elements - Microbiology

1. Demonstration of Capability (DOC), initial and on-going*
2. Method blanks and sterility checks*
3. QC samples and laboratory fortified blanks*
4. Matrix spike and matrix spike duplicates*
5. Calibration*

10 Critical Elements - Microbiology

6. Control charts – trend analysis*
7. Corrective actions*
8. QC acceptance criteria*
9. Definition of a batch or test run*
10. Minimum frequency of QC checks*

1. DOC

- Each analyst must demonstrate initial and ongoing capability for each analytical they perform.
- The intent is to prove both reliability and integrity of the laboratory’s test results.

2. Method Blanks & Sterility Checks

- Ensure that unknown samples have not been compromised, contaminated or invalidated due to
  - improper handling or preparation,
  - inadequate sterilization or
  - environmental exposure.

3. QC Samples & Laboratory Fortified Blanks

- Ensure that growth media, or other method reagents/materials, are capable of supporting proper growth and/or analytical results.
- Positive and Negative Culture Controls

4. Matrix Spike & Matrix Spike Duplicates

- For difficult matrices or new methods

5. Calibration

- **Calibration of microbiological equipment (initial and continuing) Performance Qualification**

6. Control Charts/Trend Analyses

- **and Trend Analyses of Quality Control Results**

7. RCA/Correction Action

- RCA = Root Cause Analysis

8. QC Acceptance Criteria

- Used to determine if test results are acceptable
- Must be established prior to testing

9. Definitions of a Batch

- **Preparation**
- **Analytical**

10. Minimum Frequency QC Checks Lab Equipment

- Ensure precise and consistent results

What Else Should We Do?