

Title: Algal Assay Short Course

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Objective:

To train MDWPC personnel in the theory and operation of the Printz Algal Assay: Bottle Test (AA:BT)

Procedure:

Three to five MDWPC personnel will attend a four to five day short course pertaining to the theory and performance of the AA:BT. The course will be offered three times during the three year project period and will include both lecture and laboratory sessions. Trainees will be taught to perform the AA:BT to evaluate 1) the limiting nutrient of a water, 2) the presence of algal toxicants, and 3) the sensitivity of a water to change in its nutrient status. Trainees will also be taught to use an electronic particle counter for enumerating algal cells. The AA:BT will be taught according to the EPA protocol. The course will involve the actual performance of an AA:BT under an abbreviated time period (AA:BT protocol requires 14 day incubation). The following steps will be covered in the lab:

1. sampling procedures
2. sample pretreatment (autoclaving and filtering)
3. algal culture preparation
4. test vessel preparation (chemical additions and algal inoculation)
5. cell enumeration
  - a. gravimetric
  - b. Coulter Counter
  - c. fluorometric (time allowing)
6. data generation and analysis
7. comparison of predicted and observed algal growth.

Expected Results:

Upon completion of the course the trainees will have a full understanding of the theory and operation of the AA:BT and will be able to conduct such tests independently in a sufficiently equipped algal assay laboratory.

Cost: \$21,000

Algal Assay Bottle Test: Short Course OutlineDay 1

- 8:30-9:00 Welcome, Introductions, Goals and Objectives, Review Course Outline
- 9:00-10:00 Nutrients and Aquatic Ecosystems - M. Switzenbaum
1. Food chains and balances
  2. Nutrients and trophic status
  3. Limiting nutrient theory
  4. Measurements of trophic status
  5. Eutrophication
  6. Sources of nutrients
  7. Nutrient control strategies
- 10:00-10:15 BREAK
- 10:15-11:00 Introduction to AA:BT, General Overview - K. Sheehan
1. Historical development
  2. General outline
    - a. Test alga
    - b. Spikes
    - c. Growth parameters
    - d. Test conditions
    - e. Data analysis and application
- 11:00-12:00 Overview of AA:BT Methods - R. Green
1. Sampling and sample preparation
  2. Inoculum, spikes, stock culture
  3. Experimental conditions
  4. Experimental endpoint-growth parameters
- 12:00-1:00 LUNCH
- 1:00-1:30 Lab Orientation - K. Sheehan, K. Sellers
- 1:30-5:00 Lab:
1. Preparation of inoculum, ANM, stock cultures
  2. Chemical additions
  3. Sample preparation and dispensing
  4. Microscopic examination of S. capricornutum
  5. Initiation of AA:BT, experimental conditions, glassware preparation, etc.

Wednesday, August 15

- 8:30-9:30 Theory of AA:BT - K. Sheehan
1. Liebig's law - examples
  2. Inadequacy of chemical analyses; N:P ratios
  3. Specificity of AA:BT

4. Correlation to trophic status
5. AA:BT spikes and information obtained
6. MSC and growth parameters
7. Bioavailable nutrients

9:30-10:15 Analytical Methods: N, P - D. Wagner

10:15-10:30 BREAK

10:30-11:15 Sampling Methods and Preservation - M. Switzenbaum

11:15-12:00 Methods of Algal Cell Mass Determination - K. Sheehan

1. Gravimetric
2. Fluorometric
3. Spectrophotometric
4. Coulter counter

12:00-1:00 LUNCH

1:00-1:30 Lab Orientation

1:30-5:00 Lab: Coulter counter

Thursday, August 16

8:30-9:15 AA:BT Toxicity Test - P. Austin

9:15-10:00 Case Studies - K. Sheehan, P. Austin

1. Quabbin Reservoir
2. Pittsfield WWTP and Housatonic River

10:00-10:15 BREAK

10:15-11:00 Methods of AA:BT Data  
Analysis and Interpretation - K. Sheehan

1. MSC
2. Confidence intervals
3. Statistically significant differences in growth
4. Nutrient limitation
5. N:P
6. Bioavailable nutrients
7. Yield coefficients
8. Effects of nutrient additions

11:00-12:00 Alternative Procedures - K. Sheehan, R. Green, D. Wagner

1. Other algal species
2. Alternate P-solubilization
3. AA:BT assessment of advanced WWT

12:00-1:00 LUNCH

1:00-1:30

Lab Orientation

1:30-4:00

Lab: 1. Final Coulter counting  
2. Data analysis  
3. Predicted vs. observed  
4. Bioavailable nutrients  
5. N:P  
6. Limiting nutrient analysis  
7. Graphical presentations  
8. Data interpretation  
9. Discussion of treatment alternatives and effects of  
nutrient removals/additions