

TECHNICAL OPERATING PROCEDURE

PROCEDURE TITLE: Continuous Flow Diluter

APPLICABILITY: Branch of Aquatic Ecosystem Health

ORIGINAL

PRINCIPLE: Calibration procedure for the continuous flow diluter

PRECAUTIONS

A. Potential Interferences

1. Clean system before use to minimize the possibility of contamination. Clean diluter with soap and water and flush system with water overnight.

B. Safety

1. Standard laboratory safety devices (lab coat, gloves, and safety glasses) should always be worn when handling solvents or chemicals or when carrying out the described procedures.
2. Follow the Material Safety Data Sheets for chemicals used in this procedure.

PROCEDURE

A. Calibration

1. Initiate water flow to diluter and fill to overflow standpipe level in the headbox.
2. Adjust water flow until a slight overflow in the headbox is achieved (Figure 1).
3. Determination of water volume
 - a. Measure the volume of water entering the cell where the chemical stock solution will enter the diluter box (Cell 1) for 1 minute (Figure 1). Repeat four times and calculate the average volume in L/min. This volume will be used to determine the chemical stock solution to be prepared to achieve the highest concentration for the system. Record this volume in the study log.
 - b. Repeat Step 3.a for the volumes entering each cell of the diluter box and sum all volumes to determine total volume of the diluter in L/min. The volume of water entering every cell should have approximately the same flow rate (except Cell 1). The volume of water entering each cell should be equal to the water leaving each cell (except Cell 1). If the flow rates are not the same, adjusting the standpipes in the headbox corresponding to the cells in question will slightly alter the flow rates. Raising the standpipes will decrease the flow rates and, vice versa, lowering the standpipes will increase the flow rates. This also applies to the standpipes

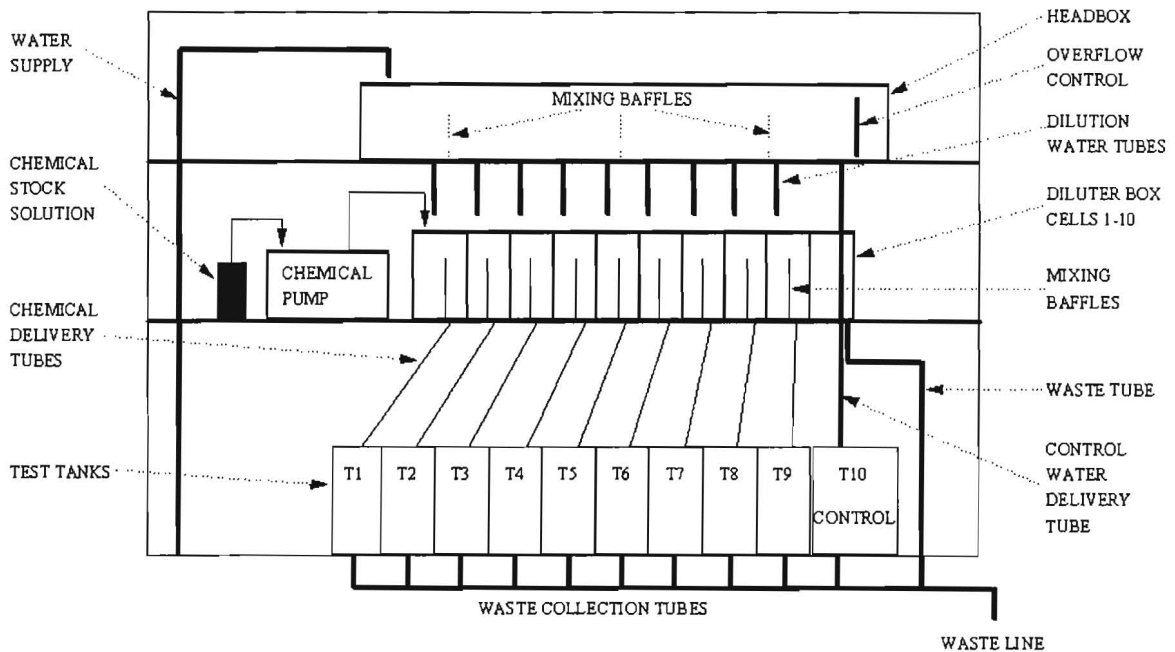


Figure 1. Diagram of Continuous Flow Diluter

in the diluter box for flow rates leaving each cell. If adjustments are necessary, start with flow rate adjustments in the headbox until all flow rates are equal entering each cell (except Cell 1) and then proceed with adjustments of flow rates in the diluter box, if necessary. Record the total diluter volume in the study log. This will help determine the amount of dilution water required for the duration of the test.

4. Determination of the dilution ratios for each cell

- a. Prepare a chemical stock solution for the desired concentration for chemical Cell 1. The concentration of the chemical stock solution should be based on the amount of chemical stock solution being delivered to Cell 1 and the calculated average of dilution water entering Cell 1. The Masterflex automatic pipettor is used to deliver the chemical stock solution to Cell 1.

(1.) Example: Desired highest test concentration = 5 mg/L

Calculated average dilution water = 3.0 L/min
 Rate entering Cell 1
 Chemical stock solution = 2.0 mL/min

Calculate chemical stock solution concentration (A):

$$(A \times 2.0 \text{ mL/min}) \div 3,000 \text{ mL/min} = 5 \text{ mg/L}$$

$$A = 7,500 \text{ mg/L or } 7.5 \text{ g*/L}$$

* assuming 100% active ingredient

- b. Initiate pumping the chemical stock solution into Cell 1 at 2.0-mg/L Masterflex automatic pipettor setting.
- c. After 1 hour, sample each aquaria delivery tube and determine chemical concentration using an appropriate analytical method for the desired chemical.
- d. Calculate dilution ratios from chemical concentrations from Step 4.c and record in the study log.

$$(1 - C_2/C_1 \dots C_n/C_{n-1}) \times 100 = \% \text{ dilution}$$

where C = chemical concentration of the cell

REFERENCE

A. None.

ORIGINAL

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DATE: 24 May 2010

APPROVED BY: [Signature]
Branch Chief, Aquatic Ecosystem Health

DATE: 26 May 2010