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and

Fisheries and Oceans Canada
Sea Lamprey Control Centre
1219 Queen Street East
Sault Ste. Marie, Ontario P6A 2E5
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INSTRUMENT OPERATING PROCEDURE

INSTRUMENT:

Spectrophotometer

MODEL:

DR/2800

MANUFACTURER:

Hach

PRECAUTIONS:

POTENTIAL INTERFERENCES

None listed in manual

SAFETY

No special safety precautions

PROCEDURES:

I. Background

A. The Hach DR/2800, the factory-suggested replacement for the DR/2400, is capable of direct readout of concentrations of TFM, however, this option does not support the

requirements of methods of analysis developed for line-powered spectrophotometers.

B. The procedures for analysis with the DR/2800 closely parallel those used with the Turner model SP-830 spectrophotometers.

1. Similarities

a. Pre-formulated standards with concentrations of 0, 4, 8, and 12 mg/L TFM are used.
b. The slope of the instrument response is determined through measurement of the absorbance of the TFM standards.
c. Base/Acid measurements of background absorbance are conducted to assure accuracy of analyses.
d. Water samples are buffered to pH ~9.0 before measurement of absorbance.
e. Water samples are filtered before measurement of absorbance.
f. Record keeping requirements are the same.

2. Difference

a. Water samples normally are not heated before analysis.

C. The accuracy of measurements depends to some degree on the comparative absorbencies of the optically-matched sample cells in each DR/2800 kit. The method of data interpretation used during TFM analyses demands that special attention is given to compensation for poor matches when they cannot be avoided.

II. Preparation

A. Set up the spectrophotometer in a shaded location. Always use the black outdoor light shield when making absorbance measurements.

B. Use line power if available, however, use of this instrument is normally reserved for situations in which line power is not available.

C. After all peripheral equipment for sample preparation is ready, press the **POWER** button on the back of the instrument. The instrument will conduct internal tests.

D. The **Main Menu** will automatically be displayed once the status check is complete. Select the **Single Wavelength** option on the touch pad screen.

E. With the **Single Wavelength** main screen active, check the wavelength for the desired wavelength of **395** nm for TFM.

F. Match the 10 mL sample cells

1. Open a new 0.0 mg/L standard and rinse and fill two factory-matched 10 mL sample cells.

2. Place the first cell in the sample cell holder with the 10 mL mark facing to the right and close the black outdoor light shield. Press **ZERO** on the touch pad screen. **"ZEROING..."** will be displayed until the instrument is zero calibrated. **0.000 ABS** will appear on the display when the instrument is zero calibrated.

3. Place the second cell in the sample cell holder with the10 mL mark facing to the

right and close the black outdoor light shield.

4. Press **READ** on the touch pad screen.

5. The sample cell should produce an absorbance ± less than 0.008 (considered an acceptable match; maximum ~ 0.05 mg/L error). If the difference is greater, clean the cell or check the absorbencies of spare sets of matched cells. If an acceptable match cannot be obtained and another matched set is not available, note the absorbance difference so absorbencies of standards can be corrected. **The cell with the lesser absorbance is used as the blank.**

III. Calibration–standards

A. Open the remainder of the new set of TFM standards. Do not allow the standards to sit in sunlight while in use and store the standards in the dark. If the standards are very cold (apt to fog), warm them in a water bath or in the hands.

B. Insert the cuvette that contains the blank and press zero. Wait until **0.000 ABS** is displayed.

C. Insert the 0.0 mg/L TFM standard and close the black outdoor light shield. The 0.0 standard may produce a reading other than 0.000 because of an imperfect match of cuvettes. If the cells are not suitably matched, subtract the difference due to cell mismatch (noted above) and record in log book.

D. Again insert the cuvette containing the blank and close the black outdoor light shield. Wait until **0.000 ABS** is displayed, insert the 4.0 mg/L standard, and close the black outdoor light shield. Press **READ** on the touch pad screen. Again adjust the result for differences in absorbance between blank and sample cells if necessary. Record the corrected absorbance in the instrument log book. Repeat the procedure with 8.0 and 12.0 mg/L TFM standards

E. Divide the recorded absorbance of each standard by the concentration of TFM (mg/L; 4.0, 8.0, and 12.0). Average the results and record the mean in the log book and on the analysis data sheet.

IV. Calibration–stream water sample

A. Measure the background absorbance and B/A ratio of the stream water (TOP:018.x). Do not interchange the cells. Always use the same cell for the 0.0 standard (blank).

B. Prepare the TFM-free water sample for analysis

1. Add 1 mL sodium tetraborate buffer to a 250 mL stream water sample and shake.

2. Filter the sample into the cuvette with a syringe filter. Generally syringe filters should be replaced after being used between 4 to 6 times. Filters can be used until resistance is felt which may be one use on a stream with high total suspended solids to many uses on a stream with low total suspended solids.

3. Adjust the sample temperature by placing the10 mL cuvette into a water bath or by holding in the hands.

C. Insert the blank into the sample cell holder.

D. Press the **ZERO** key and wait until the instrument is zero calibrated.

E. Dry the sample cuvette with a tissue, insert into the sample cell holder, close the black outdoor light shield, and press **READ** on the touch screen.

F. Note the measured result on the analysis form. This absorbance includes both the background absorbance of the stream water and the difference in absorbance between the blank and sample cells. Do not correct this value for differences between cells even if it is significant. If the difference between cells was considered significant when checked, compensation was made when producing the calibration curve.

G. Determine the B/A ration for the stream water at the site according to procedures outlined in TOP:018.x.

V. Sample measurement

A. Collect a stream water sample containing TFM.

B. Prepare the water sample for analysis.

1. Add 1 mL sodium tetraborate buffer to a 250 mL stream water sample and shake.

2. Filter the sample into the cuvette with a syringe filter.

3. If problems with cuvette fogging occur, adjust the sample temperature by placing the 10 mL cuvette into the water bath or by holding in the hands.

C. Insert the blank into the sample cell holder.

D. Press the **ZERO** key and wait until the instrument is zero calibrated.

E. Dry the sample cuvette with a tissue and insert into the sample cell holder.

F. Close the black outdoor light shield, and press **READ** on the touch screen.

G. Record the resulting absorbance on the lampricide analysis data sheet.

H. Subtract the background absorbance (base blank on the data sheet) and record.

I. Divide the resulting absorbance by the calculated slope of the calibration curve.

J. Record the result as the concentration of TFM (mg/L) in the stream water sample.

VI. Documentation

A. Make entries into instrument log book each time instrument is used.

B. Record results of analysis on LAMPRICIDE ANALYSIS data sheet (Appendix M).

MAINTENANCE:

I. Cleaning instrument -- see page 73, Section 7.1 in the instrument manual.

II. Replacing batteries -- See page 74, Section 7.2 in the instrument manual.

III. Replacing Lamp -- See page 76, Section 7.3 in the instrument manual.

REFERENCE:

Hach DR/2800 Spectrophotometer Instrument Manual

This procedure has been reviewed and approved by the undersigned representative of the U.S. Fish and Wildlife Service and Fisheries and Oceans Canada.

REVIEWED/APPROVED Jessica M. Barber Digitally signed by JESSICA BARBER Date: 2020.03.10 11:06:34 -04'00' DATE_____
Field Supervisor (U.S.)

REVIEWED/APPROVED Lula Spires DATE 05MAR2020
Program Manager (Canada)