U.S. Fish and Wildlife Service
Marquette Biological Station
3090 Wright Street
Marquette, Michigan 49855
U.S.A.

and

U.S. Fish and Wildlife Service
Ludington Biological Station
5050 Commerce Drive
Ludington, Michigan 49431
U.S.A.

and

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

TECHNICAL OPERATING PROCEDURE

PROCEDURE TITLE:

Procedures for Conducting Rhodamine WT Dye Dilution Studies using fluorescent dye feeds.

APPLICABILITY:

Procedure applies to the use of Rhodamine WT dye to estimate the dilution of lampricides prior to application to streams.

PRINCIPLE:

Rhodamine WT dye is applied with a continuous addition technique to simulate a lampricide application. The application is made at a uniform rate over a period of time to ensure a stable concentration of dye at a downstream location. The loss of dye concentration is used to predict lampricide losses during stream treatments. For
measurement of time-of-travel of stream water using fluorescent dye slugs see TOP:002.x.

SAMPLE COLLECTION AND PRESERVATION:

Samples of water containing fluorescent dye are collected by hand or by automatic water sampler (IOP:002.x and IOP:003.x). Water samples are usually analyzed immediately, but if storage is necessary the samples are labeled and refrigerated. Subsequent analysis is completed as soon as possible.

EQUIPMENT REQUIRED:

I. Sample collection

A. Sample bottles of sufficient volume for measurement of samples. Maximum requirement is about 500 mL for a Turner Designs fluorometer utilizing a flow-through cell

configuration.

B. Sigma or ISCO brand automatic sampler

II. Fluorescent Dye

A. Rhodamine WT (20% solution)

III. Analytical equipment

A. Fluorometer (IOP:004.x, , IOP:004.xB, and IOP:004.xC)

POTENTIAL INTERFERENCES:

There can be significant losses to adsorption of Rhodamine WT on certain clays. Ambient air temperatures below 32^0 F will cause freezing in the suction hose of the automatic sampler if left exposed. If an intake is not suspended above stream bottom, silt and other debris will obstruct hose. If two units are set next to each other electro-magnetic interference may occur.

SAFETY:

No special safety precautions

DISPOSAL:

Not applicable

REAGENTS:

Not applicable

PROCEDURES:

I. Calibration

Note: Rhodamine WT Dye is considered 100% active for this procedure

A. Preparation of a 1000 uL/L (part per million) Rhodamine W.T. standard solution

1. Pipette 1 mL Rhodamine W.T. into a 1000 mL volumetric flask

2. Fill to the 1000 mL mark with deionized water

3. Label the flask

B. Preparation of a 1 uL/L Rhodamine W.T. standard solution

1. Pipette 1 mL of a 1000 uL/L Rhodamine W.T. into a 1000 mL volumetric flask

2. Fill to the 1000 mL mark with deionized water

3. Label the flask

C. Preparation of a 100 nL/L (part per billion) Rhodamine W.T. standard solution

1. Dispense 100 mL of 1 uL/L standard stock solution into a 1000 mL volumetric flask

2. Fill to the 1000 mL mark with deionized water

3. Label the flask

D. Calibrate the fluorometer with the 100 nL/L standard solution (IOP:004.x, IOP:004.xB, and IOP:004.xC)

II. Dye dilution investigation must be indexed to the discharge of the stream section under study. A stream discharge measurement is conducted prior to the application of dye (TOP:001.x).

III. The application rate of dye is determined by the formula:

$qd = Q * C/10^9$

Where:

qd = application rate (cc/min)
Q = discharge (cc/min)
C = desired concentration of dye (ppb)

Note: 1. Concentrations are based on volume:volume relationships.
2. Discharge must be multiplied by a factor of 10^6 (cubic meter to cubic centimeter) and 60 (minutes to seconds).

Example: $Q = 20\ m^3/s$
$C = 2$ ppb

$Qd = Q * C$
$= 20 m^3/sec * 10^6\ cc/m^3 * 60\ sec/min * 2\ parts/10^9$
$= 2.4$ cc/min

IV. An application rate of 0.06 cc/min will yield 1.0 ppb in 1 cubic meter per second. Diluted mixtures of dye are required for application. A 1:1000 dilution is appropriate to determine dye concentrations on small to medium discharge streams; a rate of 60 mL/min yields 1 ppb in 1 cubic meter per second discharge.

V. The addition of dye is usually made at a proposed site for application of lampricide. A sufficient volume of dye solution is made to ensure an appropriate application time. It is desirable to feed the dye for the same period of time that lampricides are to be applied, however, shorter feed times can be used. Pertinent application data (start time of application, feed rates, amount of dye used, staff gauge readings, application location) are recorded.

VI. Dye is applied to the stream by methods similar to those used to apply lampricides (IOP:005.x).

VII. Concentrations of dye are measured to determine the downstream progress of the block at pre-selected sites. Water samples are taken by hand or collected by automatic water samplers. Sampling continues until a profile of dye concentrations is complete.

VIII. Concentrations of dye can be measured with a fluorometer either on site (with battery power) or in a lab (line power). All sample times are recorded.

IX. The time of travel is also determined and is considered the time necessary for the dye to reach maximum concentration at each sampling site.

REFERENCES:

Replogle, J.A., L.E. Myers and J.B. Brust. Flow Measurements with Fluorescent Tracers. Proceedings of the American Society of Civil Engineers. 1966.

Wilson, J.F. Jr. Time-of Travel Measurements and Other Applications of Dye Tracing. Internat. Assoc. Sci. Hydrol. Pub. No. 76. 1968.

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

REVIEWED/APPROVED Jessica M. Barber Digitally signed by JESSICA BARBER Date: 2021.02.11 14:06:18 -05'00' DATE_____

Field Supervisor (U.S.)

REVIEWED/APPROVED Steeves, Todd 0 Digitally signed by Steeves, Todd 0 DN: C=CA, O=GC, OU=DFO-MPO, CN="Steeves, Todd 0" Reason: I agree to the terms defined by the placement of my signature in this document Location: Sault Ste Marie Date: 2021-02-09 13:07:40 Foxit PhantomPDF Version: 10.0.1 DATE_____

Program Manager (Canada)

Revision No.	Date	Person(s) Responsible	Description
31	2/1/21	Benson Solomon, Lauren Freitas, Shawn Robertson, Chris Gagnon	No significant changes, minor wording