GREAT LAKES FISHERY COMMISSION

2020 Project Completion Report[1]

DEVELOPMENT AND EVALUATION OF AN IMPROVED TFM FORMULATION FOR USE IN FEEDER STREAM TREATMENTS

Project ID – 2018_LAN_76012

by:

James A. Luoma[2], Nicholas J. Robertson[3], Nicholas A. Schloesser[2], Courtney A. Kirkeeng[2], Justin R. Schueller[2], and Erica K. Meulemans[3]

[2] U.S. Geological Survey
Upper Midwest Environmental Sciences Center
2630 Fanta Reed Road
La Crosse, WI 54602

[3] Northland College
1411 Ellis Ave.
Ashland, WI 54806

May 2020

CONTACT INFORMATION:

James A. Luoma

U.S. Geological Survey

Upper Midwest Environmental Sciences Center

2630 Fanta Reed Road

La Crosse, WI 54602

jluoma@usgs.gov

(608)781-6391

ABSTRACT:

The binational Great Lakes Fishery Commission sponsored Sea Lamprey Control Program effectively utilizes a variety of lampricide tools to keep populations of parasitic sea lampreys in the Great Lakes at levels that do not cause undue economic or ecological damage. The most widely used active ingredient used in lampricide formulations is 3-trifluoromethyl-4-nitrophenol (TFM). In typical treatments, a liquid TFM formulation is applied to lamprey producing streams continuously for 10–14 hours to produce a moving block of lampricide-treated water that kills larval lamprey before they metamorphose into their parasitic lifestage. In many smaller tributaries of dendritic streams a solid bar formulation of TFM is used to supplement the mainstem treatment block. These supplemental TFM bar applications are coordinated with the arrival of the mainstem lampricide to prevent larval sea lamprey from seeking refuge in untreated waters and surviving the treatment. TFM bars are produced from formulated surfactants and designed to release TFM over an 8–10-hour period, depending on water temperature and velocity. However, some of the surfactants have been discontinued resulting in the reformulation of the TFM bars multiple times. As a result of these reformulations, TFM bar performance has declined.

An experimental surfactant-free solid TFM tablet formulation was developed as a potential replacement for TFM bars. Release of TFM from the experimental tablets was evaluated using replicated laboratory dissolution trials conducted at three water temperatures and three water velocities. A continuous-flow laboratory flume was used for the dissolution trials and the decay of the tablets was modeled using logistic decay curves. Time required for the TFM tablet to decay 50 and 99% were compared among the groups using a two-way analysis of variance. Post-hoc Tukey Honest Significant Difference tests indicated that both water temperature and water velocity influenced the decay of the tablet; however, neither water temperature nor water velocity appeared to dramatically influence TFM release. Results from this laboratory study indicate that the next stage of evaluating the TFM tablets using field tests is warranted.

INTRODUCTION:

Treatments of sea lamprey larval nursery streams using 3-trifluoromethyl-4-nitrophenol (TFM) started in 1958 and they quickly became the primary mechanism to control sea lamprey and protect the $7B Great Lakes fishery (GLFC 2014). During typical stream treatments, TFM is continuously applied to the mainstem of the river for 10–14 hours, resulting in a moving 9-hour treated-water block at a lethal concentration (Boogaard et al. 2015, Lantz et al. 2019). During these applications, tributaries to the treated section of the stream must also be treated with lampricide to prevent sea lamprey larvae from escaping into untreated waters and surviving. A solid lampricide bar formulation (U.S. EPA registration No. 6704-86 was developed in the mid-1980s to aid in the treatment of tributaries with discharges typically less than 0.09 m^3/sec and water velocities typically less than 0.15 m/sec (Gilderhus 1985, Solomon 2019). The lampricide bar formulation was developed to contain 23% TFM as the active ingredient (A.I.) and a surfactant-based carrier system that would slowly release the TFM over an 8–10-hour period (Gilderhus 1985). Since development, several reformulations of the TFM bar carrier system have been necessitated due to discontinued surfactants. Poor bar performance, including premature dissolution and product softening, has resulted from these reformulations (Solomon 2019).

Surfactants currently used in the TFM bars have a reported 96-hour LC50 range of 1.0 – 1.8 mg/L for bluegill (*Lepomis macrochirus*) and a 48-hour EC50 of 12.2 mg/L for *Daphnia magna* (CAS No. 9016-45-9; Iofina Chemical, Inc. 2013). Additionally, degradation of some block copolymer surfactants (CAS No. 9003-11-6) is not well understood (Sigma-Aldrich 2019). Although the likelihood of acute toxicity from the inert ingredients in the TFM bar is low, a formulation with more environmentally compatible inert ingredients is desirable and aligns with the priorities of the Great Lakes Fisheries Commission's Sea Lamprey Control Research Board and the objectives of the Lampricide Control Task Force (GLFC 2019).

Replacing the surfactant-based TFM bar with an alternate slow-release solid TFM formulation manufactured with thermally stable and readily available inert ingredients could resolve dissolution and product softening performance issues, improve environmental safety, decrease the amount of product required for treatments (i.e. increase A.I. content), and potentially reduce production costs. A tableted formulation of TFM was developed using sand, thermally stable and readily available food-grade inert ingredients, and other formulants that are included on U.S. Environmental Protection Agency lists that are not anticipated to adversely impact public health or the environment (U.S. EPA 2004, U.S. EPA 2016).

OBJECTIVES:

The objectives listed in the Technical Assistance Proposal were met and include:

(1) Develop an improved, more environmentally benign and lower-cost TFM formulation(s) with ≥ 50% TFM (w/w).

Summary result: An improved solid TFM formulation was develop that contained 68.5% high-purity TFM and inert ingredients that are of low regulatory concern (sand, calcium hydroxide, and magnesium stearate) and coated with a low regulatory concern blend of sodium alginate, psyllium hydrophilic mucilloid, and sucrose. Large scale production costs are not currently available but have the potential to be lower because of use of readily available and non-formulated inert ingredients.

(2) Optimize an improved TFM formulation(s) for controlled release over 10 – 12 hours.

Summary result: Over 100 different formulations were prepared and release rates evaluated. TFM release was optimized to the desired release profile by varying the tablet coating, dimensions, and composition. The optimized tablets are approximately 0.9 g, 14 mm diameter, and 4 mm thick.

(3) Comprehensively evaluate release rates of improved TFM formulation(s) at various water temperatures.

Summary result: A comprehensive study protocol was developed and executed to evaluate the optimized formulation at three different water temperatures and three different water velocities using replicated laboratory flume trials. This completion report contains thorough methods, data analysis, results, and discussion sections describing the development and evaluation of the TFM tablet formulation. The laboratory study had specific objectives including:

(1) Evaluate the influence of water temperature on the release of TFM from the experimental formulation.

Summary result: The objective was met and water temperature was found to affect the release of TFM from the experimental tablet formulation.

(2) Evaluate the influence of water velocity on the release of TFM from the experimental formulation.

Summary result: The objective was met and water velocity was found to affect the release of TFM from the experimental tablet formulation.

(3) Determine the suitability of the experimental formulation for continued evaluation using field trials.

Summary result: After completing the replicated laboratory trial and evaluating the data, the experimental TFM tablet formulation is deemed suitable for field investigation.

METHODS:

The experimental TFM tablet formulation was evaluated using a total of 36 independent dissolution trials that were conducted using 4 replicates at each of 3 water temperature and 3 water velocity combinations. The dissolution trials were conducted in a continuous-flow laboratory flume. TFM concentrations in the discharge water were monitored, and water chemistry data were collected during each 24-hour trial. Summary statistics were prepared for water-quality data, and TFM elution from the tablets among the experimental groups was statistically compared. Data collected for this study are available in Luoma and Schloesser (2020).

Test System

A 6.8-m long, continuous-flow test flume system constructed from 15.2-cm diameter poly vinyl chloride pipe was used for the dissolution trials (Figure 1). Well water was used for all trials and temperature acclimation was performed in six 4,000-L stainless steel tanks fitted with 1-hp heat pumps (model DSHP-9, Aqua Logic, Inc., San Diego, California, USA). Water flow to the flume was controlled using a variable-frequency-drive controlled pump and the flow rates were determined by conducting replicated manual flow rate measurements using a stopwatch and a 90-L vessel prior to beginning and terminating each trial. Prior to trial initiation, water flows were set to approximately 11, 15, and 22 L/minute for the 0.04, 0.08, and 0.12 m/sec trials, respectively. Then, water velocities were calculated and adjusted as necessary to be within 5% of target by modifying the water flow and/or the height of the water column in the flume. Water velocities were derived using two equations. First, equation 1 was used to calculate the cross-sectional surface area (cm^2) of the flooded portion of the flume (Page 2011), then equation 2 was used to calculate the water velocity within the flume using the cross-sectional area resulting from equation 1 and the measured flow rate.

$$A = r^2 \cos.^{-1}\left(\frac{r\text{-}h}{r}\right) - (r - h)\sqrt{(2rh - h^2)}$$
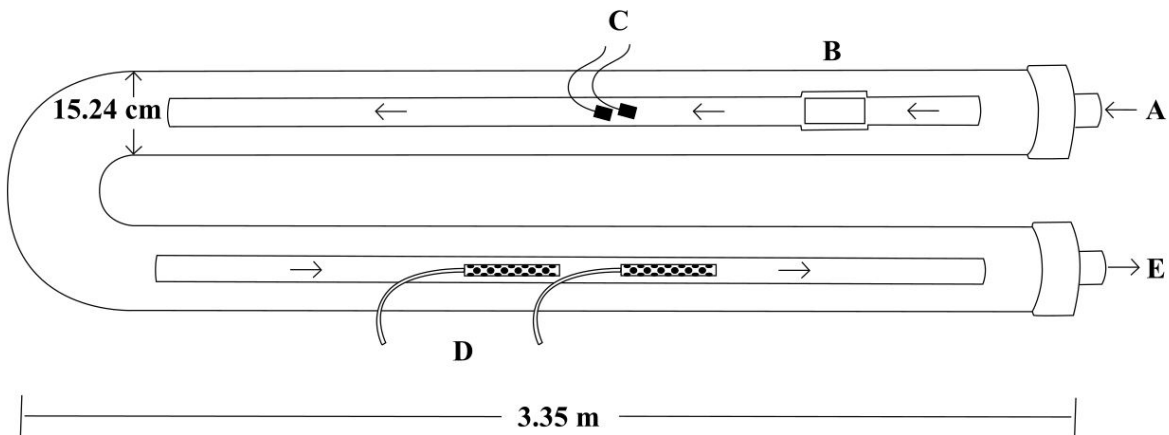
(1)

where

A   Flooded cross-section area of the flood area of the pipe (m^2).

r Pipe radius (m), and

h Height of the water in the pipe (m).

$$V = \frac{q}{A}$$

(2)

where

V Water velocity (m/sec),

q Flow rate (m^3/sec), and

A Flooded cross-sectional surface area derived from equation 1 (m^2).

Figure 1. A schematic of the flow-through flume used in the dissolution trials showing the influent port (A), the elevated platform where the experimental TFM tablets were placed (B), airstones used to mix eluted TFM (C), water sample collection lines (D), and the effluent port (E).

Experimental TFM Tablets

The experimental TFM tablets were produced on a commercial press from a mixture of high-purity TFM and inert ingredients. After pressing, the tablets were then coated and cured to control the release of TFM.

<u>High-purity TFM Preparation</u>

High-purity TFM used in the tablet production was extracted from an end-use liquid lampricide formulation (36.5% A.I.) and dehydrated until ≥ 98.5% pure. First, the sodium salt formulation was converted to the free phenol form by placing equal portions of end-use TFM lampricide (U.S. EPA registration No. 6704-45) and concentrated hydrochloric acid (36.5-38%) into a 2-L separatory funnel, which was then agitated for 5 minutes and allowed to rest for 20 minutes. The bottom layer, consisting of TFM and small amounts of isopropanol, sodium chloride, and water, was then removed and placed into a clean 2-L separatory funnel where it was washed with 1 L of deionized water to remove remaining sodium chloride and isopropanol. Lastly, the washed TFM was removed and placed into glass pans and concentrated by evaporation at 50°C in a fume hood. The extracted and dehydrated TFM was analyzed by high performance liquid chromatography (HPLC) and used for TFM tablet production after confirming the purity was ≥ 98.5%.

<u>TFM Tablet Development, Production and Coating</u>

TFM tablets were produced at Northland College (Ashland, Wisconsin, USA) using an automated desktop tablet press (Figure 2; model TDP5, LFA Machines Oxford Ltd, Chichester, West Sussex, United Kingdom). Over 100 different TFM tablet formulations were prepared and release rates evaluated. Release rates were optimized by varying the tablet coating, dimensions, and composition. The optimized formulation was prepared by mixing high-purity TFM (68.5% by weight) with inert ingredients (sand (20%), calcium hydroxide (11%), and magnesium stearate (0.5%)) into a homogenous flowable powder.

A TFM release-controlling coating was applied (1% w/w) to the tablets by immersing the tablets into the coating solution (a proprietary mixture of sodium alginate, psyllium hydrophilic mucilloid, and sucrose) which was dried at 49°C for 1 hour in a forced-air dehydrator before a second coat was applied and final curing at 49°C for 12 hours in the forced-air dehydrator. The final tablets were approximately 0.9 g, 14-mm diameter, and 4-mm thick.
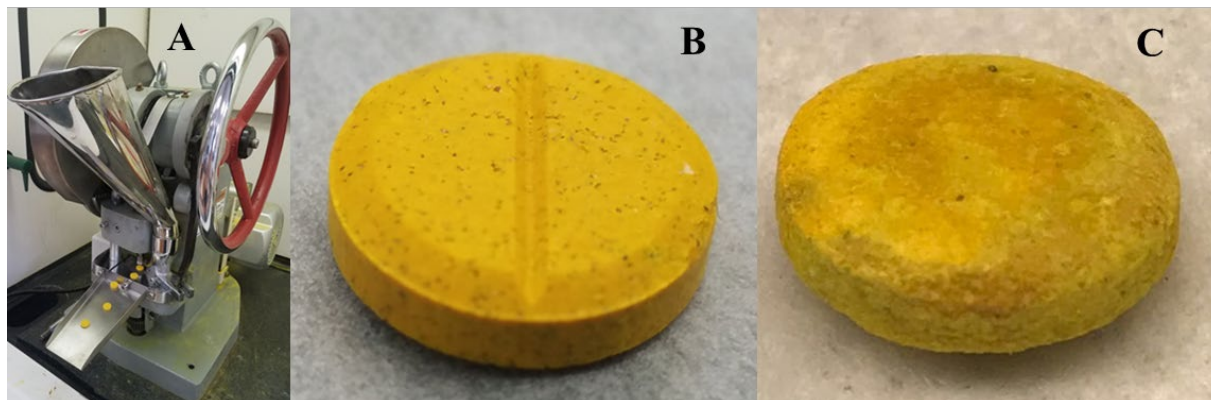
Figure 2. The model TDP5 automated press used to produce the experimental TFM tablets (A), an uncoated TFM tablet (B), and a coated TFM tablet (C).

TFM Tablet Active Ingredient Content

The mean A.I. concentration of the TFM tablets was computed from the sample TFM concentration (c_s) measured in 25 individual tablets. The mass of each tablet was determined prior to tablet bisection and elution of TFM using methanol in a 100-mL volumetric flask. The eluate was then diluted 1,000-fold by placing 1.0 mL in a 1 L volumetric flask and bringing to volume in 18 MΩ-cm water. The concentration of TFM in the final dilution was determined with HPLC and the percentage of TFM in each tablet was determined with equation 3.

$$Percent\ active\ ingredient = c_s \times \frac{1}{m} \times 10,000$$

(3)

where

c_s sample TFM concentration (mg/L), and

m tablet mass (mg).

Dissolution trials

Four replicated dissolution tests were performed for each water temperature (12, 16, and 20°C) and water velocity (0.04, 0.08 and 0.12 m/sec) combination, for total of 36 individual trials. The water temperatures and water velocities evaluated are relevant to field conditions when and where TFM bars are typically applied (Solomon 2019) and they were sufficient to achieve the study objectives. Water flow and water velocity were measured and adjusted as necessary; then, the mass of intact tablets that yielded the closest initial theoretical TFM concentration to 1.0 g of TFM per liter of water inflow was placed into the flume. The tablets were placed on an elevated sample platform approximately 60 cm downstream from the flume inflow and covered with a 0.64-cm^2 wire mesh cage. Water samples for TFM concentration analysis were collected every 30 minutes from 0 – 23.5 hours using two automated water samplers (model #3700, Teledyne-Isco, Inc, Lincoln, Nebraska, USA). Additional TFM water concentration samples were collected by hand using a pipettor every 5 minutes during the first 25 minutes to determine how quickly the tablets began to release TFM. All water samples were collected near the outflow and analyzed for TFM content using HPLC. Basic water chemistry properties (hardness, alkalinity, and pH) were measured within 30 minutes of the initiation and termination of each trial. Water pH was measured in the flume using either a Hach or an Orion Star portable water-quality meter (model HQ40d, Hach Company, Loveland, Colorado, USA; model A221, Thermo Fisher Scientific, Inc., Waltham, Massachusetts, USA, respectively). Total hardness and alkalinity were measured on water samples collected from the outfall using the EDTA titrimetric method (method 2340C; APHA 2012) and titrating to an endpoint of pH 4.5 (method 2320B; APHA 2012), respectively. Water temperature was monitored in the flume during each trial using a temperature data logger (HOBO pendant temperature/light 64K, Onset Computer Corporation, Bourne, Massachusetts, USA). Temperature verifications were also completed at the beginning and end of each trial using a hand-held digital thermometer (model Mk4; ThermoWorks Company, American Fork, Utah, USA).

Concentration Verification

TFM concentrations in the TFM extract, TFM tablets, and water samples were determined using HPLC (USGS 2019). Sample response was compared to a standard curve created from analytical standards. Standards and samples were analyzed using an Agilent model 1260 HPLC (Agilent technologies Inc., Santa Clara, California, USA) equipped with an autosampler, a Kinetex XB-C18 column (3 × 50 mm, 2.6-μm particle size) maintained at 50˚C, and a diode array detector. The flow rate was 1.25 mL/minute, injection volumes were 25-μL and gradient elution was used. The gradient elution used two 10 mM ammonium acetate mobile phase constituents (A, in a 3:1 mixture of water and methanol; B, in methanol), both of which were prepared and buffered by placing 770 mg of ammonium acetate and 3.0 mL of acetic acid per liter of solvent (USGS 2019). The percentages of A:B mobile phase constituents during each 1.5-minute sample analysis run were initially 75:25, which was transitioned to 60:40 from 0.0 to 0.5 minutes and then back to 75:25 for the remainder of the run. Sample absorbance was determined at 295.0 nm using a 10.0-nm bandwidth and the normal influences of gradient analysis were accounted for using a reference absorbance at 350.0 nm with a bandwidth of 80.0 nm.

Analytical standards were prepared by diluting a known weight of reagent grade TFM with methanol in a volumetric flask. Further dilutions were made using 18 MΩ-cm water to create a series of five standards that bracketed the experimental sample(s).

TFM Recovery

The percentage of TFM placed in the flume during each trial that could be accounted for (i.e. recovered) was calculated using equation 4.

$$Percent\ Recovery = \sum_{i=0}^{24} \left[\frac{c_i \times f \times t_i}{k} \right] \times 100$$

(4)

where

i time of sample (hours),

c_i	concentration of TFM in the sample (mg/L),
f	flow of water through the flume (L/hour),
t_i	time between samples (hours), and
k	known amount of TFM placed in the flume (mg).

The percentage of TFM remaining at each sampling point was calculated by subtracting the cumulative amount of TFM eluted from the known amount of TFM placed in the flume. These values were then corrected by dividing the result by the trial specific percent recovery. The corrected percentage of TFM remaining data were analyzed to compare TFM tablet decay among the treatment groups.

DATA ANALYSIS:

Data were summarized, modeled, and analyzed using R (version 3.6.1 and 3.6.2, R Core Team 2019) and RStudio software (version 1.2.1335; RStudio 2019). Water chemistry properties and water velocities were summarized with simple descriptive statistics. A logistic decay curve was fit to the TFM tablet decay for each trial replicate and modeled according to OECD (2014) using the DRC package (Ritz et al. 2015). Model fitness was assessed by calculating the median absolute deviation (MAD) for each trial. The MAD was calculated as the median of the absolute value of the distance between the experimentally calculated and the logistic model predicted percentages of TFM remaining in the tablets. The MAD was computed for each of the four replicate trials conducted at each of the nine experimental conditions (i.e. each unique water temperature and water velocity combination) and then the mean and standard deviation were computed across the four replicates.

Differences in the model-predicted time required for the TFM tablets to decay 50 and 99% among the groups were detected using a two-way analysis of variance (ANOVA; Montgomery 2017) with both target temperature and target velocity as factors. Post-hoc Tukey Honest Significant Difference (HSD; Montgomery 2017) tests were used to determine the differences of the predicted 50 and 99% decay times among the target temperature and target velocity group means.

RESULTS:

Water quality properties (pH, alkalinity, and hardness) were similar among all trails and because they have not been attributed to TFM elution they were not evaluated further, except for temperature which was a planned covariate (Table 1). Individual pH, alkalinity, and hardness measurements from all trials ranged from 7.62 to 8.38 standard units, 139 to 154 mg/L (as $CaCO_3$), and 173 to 206 mg/L (as $CaCO_3$), respectively. Individual temperature measurements ranged from 11.8 to 12.7°C, 15.5 to 15.9°C, and 19.5 to 21.0°C for the 12, 16 and 20°C trials, respectively.

Table 1. Mean and range of basic water-quality properties collected during TFM tablet dissolution trials conducted in a laboratory flume at 12, 16, and 20°C.

| Trial Temperature (°C)	pH (standard units)	Alkalinity (as $CaCO_3$)	Hardness (as $CaCO_3$)	Measured Temperature (°C)
12	8.02 (7.87–8.12)	152 (147–154)	193 (184–196)	12.1 (11.8–12.7)
16	8.01 (7.62–8.18)	150 (139–153)	193 (173–206)	15.6 (15.5–15.9)
20	8.11 (7.96–8.38)	151 (149–154)	194 (190–196)	19.7 (19.5–21.0)

Mean water velocities for the four replicates were within 2.8, 0.6, and 0.1% of target for the 0.04, 0.08, and 0.12 m/sec trials, respectively. The water velocities for all 36 individual trials were within 5% of target.

The correlation coefficient (R) values for calibration curves used to determine TFM sample concentrations were > 0.999, indicating sample curve linearity and minimal baseline drift. The mean recovery of TFM from the 12, 16, and 20°C trials was 91.9, 94.4, and 93.4, respectively. The range of recoveries from individual trial replicates was 90.3 to 94.7, 90.7 to 96.7, and 90.9 to 97.0 in the 12, 16, and 20°C trials, respectively.

The logistic decay models were appropriate for the TFM tablet decay data which was demonstrated by evaluating the mean MAD values. The mean MAD values show that the variations between the experimentally calculated and the model predicted amounts of TFM remaining in the tablets differed by 0.27 to 0.95% for all trials (Figure 3). Water temperature and water velocity were factors that

influenced the time required for 50 and 99% of the TFM tablets to decay ($F_{(2,31)} = 165.3$, $p < 0.001$ and $F_{(2,31)} = 34.2$, $p < 0.001$) and ($F_{(2,31)} = 236.9$, $p < 0.001$ and $F_{(2,31)} = 42.9$, $p < 0.001$), respectively). Evaluation of the influence of water temperature on the model predicted mean time required to decay the TFM tablets at each water velocity indicated that the time required to decay 50 and 99% of the tablets took 1.0 to 1.3 hours and 3.0 to 3.8 hours longer at 12°C than at 20°C, respectively. The mean time required to decay 50 and 99% of the TFM tablets ranged from 4.2 to 4.9 hours and 12.6 to 14.5 hours at 12°C, respectively, and 3.2 to 3.6 hours and 9.6 to 10.8 hours at 20°C, respectively.

Similarly, evaluation of the influence of water velocity on the mean time required to decay the TFM tablets at each water temperature indicated that the time required to decay 50 and 99% of the tablets took 0.4 to 0.7 hours and 1.1 to 1.9 hours longer at a water velocity of 0.04 m/sec than at a water velocity of 0.12 m/sec, respectively. The mean time required to decay 50 and 99% of the TFM tablets ranged from 3.6 to 4.9 hours and 10.8 to 14.5 hours at 0.04 m/sec, respectively, and 3.2 to 4.2 hours and 9.6 to 12.6 hours at 0.12 m/sec, respectively.
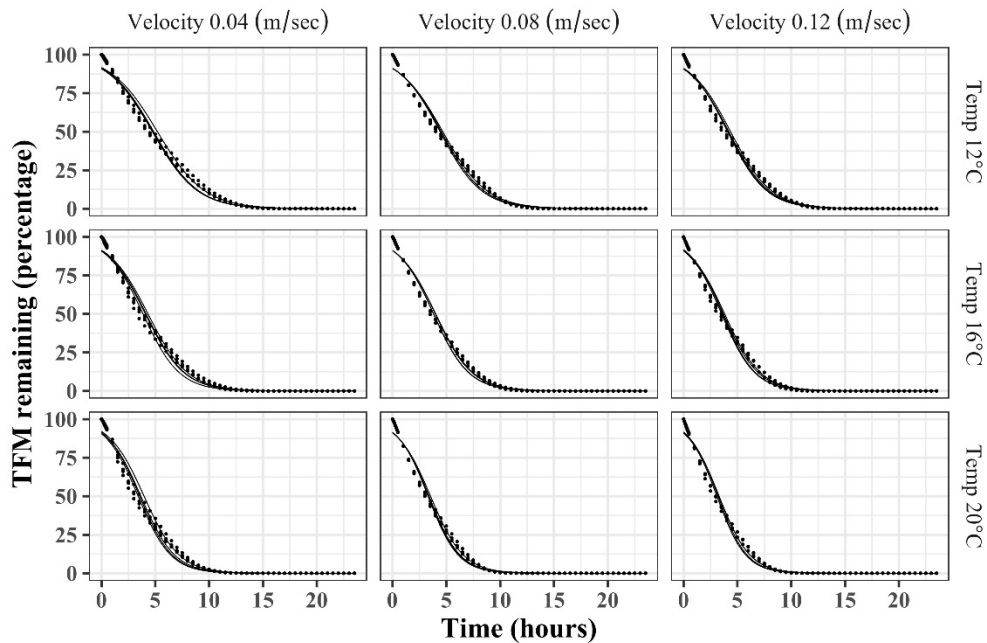
Figure 3. The percentage of TFM remaining in the TFM tablets (dashed lines) and the associated logistic decay curves (solid lines) for replicated dissolution studies conducted at nine different combinations of water temperature and water velocity.

DISCUSSION:

This study demonstrated that an alternative solid TFM tablet formulation could (1) meet TFM bar label-defined dissolution characteristics, (2) resolve the poor performance characteristics of the TFM bar, (3) reduce environmental impacts by changing inert ingredients, and (4) eliminate the need to reformulate a solid TFM formulation due to the discontinuance of formulated inert ingredients.

In this study, the TFM tablet formulation met the dissolution characteristic described in the TFM bar label (Solomon 2019), and while water temperature and water velocity did influence TFM elution, they did not influence it dramatically (Figure 3). The current TFM bars prematurely dissolve in warm water, which led to the recommendation to apply them at one-half the label rate and in some instances, additional TFM bars need to be reapplied 4 – 5 hours later (Solomon 2019). In dissolution tests using the current bar formulation at water velocities of approximately 0.04 m/sec, approximately 50% of the TFM was eluted in 2 and 5 hours at water temperatures of 20 and 12°C, respectively, and approximately 99% of the TFM was eluted in 8 and 17 hours at 20 and 12°C, respectively (Luoma and Schloesser 2020). The TFM tablets increased the mean time required to elute 50 and 99% of the TFM approximately 1.5 and 1.4 times at 20°C, a substantial improvement over the current TFM bar formulation. At 12°C the time required to elute 50% of the TFM from the tablets was similar to the current TFM bar formulation (4.9 vs. approximately 5 hours) and the time required to elute 99% of the TFM was reduced (14.5 vs. approximately 17 hours), indicating a more desirable decay pattern.

In contrast to the TFM bars, the TFM tablets are formulated with readily available inert ingredients that are environmentally compatible (U.S. EPA 2004, U.S. EPA 2016). The TFM tablets eliminate the need to reformulate the surfactant-based carrier system due to product discontinuance and they eliminate the discharge of surfactants into the environment. In addition, the low regulatory concern of the TFM tablet inert ingredients could be beneficial towards obtaining registration for the TFM tablet formulation.

This laboratory study demonstrated that the TFM tablets exhibit acceptable performance characteristics over a range of water temperatures and water velocities (Figure 3). However, field trials

are necessary to complete a comprehensive evaluation of their potential as a replacement for the TFM bar. Specific research questions that warrant investigation in a field trial include: (1) evaluating release of TFM from the tablets when applied over a range water velocities typically encountered in the field, (2) evaluating release of TFM from the tablets when they are applied over typical field substrates, and (3) assessing TFM tablet mobility when applied under various water velocity and/or substrate conditions. If acceptable performance characteristics are observed during experimental field applications, then storage stability, product chemistry, and toxicological studies will be needed to support a potential pesticide registration.

REFERENCES:

APHA (2012). American Public Health Association. Standard methods for the examination of water and wastewater. 22nd edn. American Public Health Association, Washington, D.C., 1348 pp.

Boogaard, M. A., Newton, T. J., Hubert, T. D., Kaye, C. A., and Barnhart, M. C. (2015). Evaluation of the short term 12 hour toxicity of 3-trifluoromethyl-4-nitrophenol (TFM) to multiple life stages of *Venustaconcha ellipsiformis* and *Epioblasma triquetra* and its host fish (*Percina caprodes*). Environmental Toxicology and Chemistry, 34(7), 1634-1641.

Gilderhus, P. A. (1985). Solid bars of 3-trifluoromethyl-4-nitrophenol: a simplified method of applying lampricide to small streams. Great Lakes Fishery Commission Technical Report No. 47, 6-12.

Great Lakes Fishery Commission (GLFC) (2014). Fact Sheet 2: The Great Lakes Fishery. http://www.glfc.org/pubs/factsheets/FACT%202_14-0913_HR.pdf. Accessed February 20, 2020.

Great Lake Fishery Commission (GLFC) (2019). Sea Lamprey Control Board Research Priorities. http://glfc.org/pubs/pdfs/research/SLCB_research_priorities.pdf. Accessed May 04, 2020.

Iofina Chemical, Inc. (2013). TFM bar safety data sheet. Covington, KY. http://www.glfc.org/sop2018/app/app%20f%20SDS%20TFM%20Bar.pdf. Accessed February 20, 2020.

Lantz, S. R., Kaye, C. A., Criger, L. A., Sullivan, T. J., Stephens, B., Boogaard, M. A., and Hubert, T. (2019). Comparison of continuous and interrupted lampricide block toxicity to sea lamprey and lake sturgeon. Project completion report submitted to the Great Lakes Fisheries Commission. 21pp.

Luoma, J.A., Schloesser, N.A. 2020. Data Release: Development and Evaluation of an Improved TFM Formulation for Use in Feeder Stream Treatments. U.S. Geological Survey. Data Release. https://doi.org/10.5066/P928SZH8.

Montgomery, Douglas C. (2017). *Design and analysis of experiments*. John wiley & sons.

OECD (2014). Current Approaches in the Statistical Analysis of Ecotoxicity Data: A guidance to application (annexes to this publication exist as a separate document), OECD Series on Testing and

Assessment, No. 54, OECD Publishing, Paris, France. https://doi.org/10.1787/9789264085275-en. Accessed March 03, 2020.

Page, John D. (2011). Area of a Circular Segment Given its Height. https://www.mathopenref.com/segmentareaht.html. Accessed May 16, 2020.

R Core Team. (2019). R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. https://www.r-project.org. Accessed May 26, 2020.

Ritz, C., Baty, F., Streibig, J.C., Gerhard, D. (2015) Dose-Response Analysis Using R. PLoS ONE 10(12): e0146021. https://doi.org/10.1371/journal.pone.0146021. Accessed April 20, 2020.

RStudio Team (2019). RStudio: Integrated Development for R. RStudio, Inc., Boston, MA. http://www.rstudio.com. Accessed May 26, 2020.

Sigma-Aldrich (2019). Pluronic® F-127 Safety Data Sheet. St. Louis, MO. https://www.sigmaaldrich.com/MSDS/MSDS/DisplayMSDSPage.do?country=US&language=en&productNumber=P2443&brand=SIGMA&PageToGoToURL=https%3A%2F%2Fwww.sigmaaldrich.com%2Fcatalog%2Fsearch%3Fterm%3D9003-11-6%26interface%3DCAS%2520No.%26N%3D0%2B%26mode%3Dpartialmax%26lang%3Den%26region%3DUS%26focus%3Dproduct. Accessed February 21, 2020.

Solomon, B., Ed (2019). Standard operating procedures for the application of lampricides in the Great Lakes Fishery Commission Integrated Management of Sea Lamprey (*Petromyzon marinus*) Control Program (No. SCL 04-001.10). Marquette Biological Station, U.S. Fish and Wildlife Service, Marquette, MI. http://www.glfc.org/sop.php. Accessed May 04, 2020.

U.S. Environmental Protection Agency (U.S. EPA) (2004). List of Inert Pesticide Ingredient - List 4B. 18 pp. https://www.epa.gov/sites/production/files/2015-10/documents/inerts_list4bname.pdf. Washington, D.C. Accessed February 21, 2020.

U.S. Environmental Protection Agency (U.S. EPA) (2016). Inert Ingredients Eligible for FIFRA 25(b) Pesticide Products. https://www.epa.gov/sites/production/files/2016-11/documents/minrisk_inert_ingredients_w_tolerances_2016-11-16.pdf. Washington, D.C. Accessed February 21, 2020.

U.S. Geological Survey (USGS) (2019). Determination of Percent Active Ingredient of Technical Grade Manufacturing Use and End-use Bayluscide (Niclosamide, NIC) and 3-trifluoromethyl-4-Nitrophenol (TFM) in Lampricide Formulations. Internal Standard Operating Procedure AEH 409. Upper Midwest Environmental Sciences Center. 7pp. http://iwww.umesc.er.usgs.gov:81/media_archives/documents/standard_operating_procedures/aquatic_ecosystem_health/aeh409.05.pdf. Accessed March 04, 2020.

ACKNOWLEDGEMENTS:

DELIVERABLES:

Reports

Luoma, J.A., Robertson, N. J., Schloesser, N.A., Kirkeeng, C.A., Schueller, J.R., and Meulemans E.K. (2020). Development and evaluation of an improved TFM formulation for use in feeder stream treatments. Project completion report submitted to the Great Lakes Fisheries Commission. 19 pp.

Presentations

Kirkeeng, C.A. and Schueller, J.R. (2020). TFM pellet and bar reformulation update. Presented at the 24th Sea Lamprey Annual Workshop. Traverse City, MI, January 22-23, 2020.

Data and Analysis Code

Luoma, J.A., Schloesser, N.A. 2020. Data Release: Development and Evaluation of an Improved TFM Formulation for Use in Feeder Stream Treatments. U.S. Geological Survey. Data Release. https://doi.org/10.5066/P928SZH8.

Students Trained

Ariah Law and Samuel Paulson (Northland College) were trained in formulation development, TFM tablet manufacturing and data interpretation.

RESEARCH HIGHLIGHTS:

- An improved and optimized tableted TFM formulation was developed as a potential replacement for TFM bars that contains 68.5% TFM and environmentally compatible inert ingredients that are of low regulatory concern.

- A laboratory study was conducted to evaluate the potential use of the tableted TFM formulation as a potential replacement for the TFM bars. In this study the release of TFM from the tablets observed during replicated laboratory flume trials conducted at different water temperatures and water velocities was evaluated and compared.

- The laboratory study demonstrates that TFM tablets may be a suitable replacement for the TFM bars that could improve TFM release profiles, eliminate the discharge of formulated surfactants into the environment, and eliminate the need for reformulation due to inert ingredient availability.

- Water temperature and water velocity were both found to be factors that influence the release of TFM from the tablets; however, the influences did not appear to be dramatic and the tablets appear to be less affected than the current TFM bar formulation.

- This study demonstrates the suitability of the experimental TFM formulation for further evaluation using a field study. Objectives of a field study could include: (1) evaluating release of TFM from the tablets when applied over a range water velocities typically encountered in the field, (2) evaluating release of TFM from the tablets when applied over various substrates, and (3) determining the mobility of TFM tablet mobility when applied under various water velocity and/or substrate conditions.