Research Article

Laboratory and field comparisons of TFM bar formulations used to treat small streams for larval sea lamprey

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Abstract

A solid formulation of the pesticide TFM (4-nitro-3-(trifluoromethyl)-phenol) was developed in the 1980s for application in small tributaries during treatments to control invasive sea lamprey (*Petromyzon marinus* Linnaeus, 1758). Several initial inert ingredients were discontinued and substituted, culminating with an interim formulation that unacceptably softens and rapidly decays in warm conditions. A new TFM bar formulation was developed to resolve poor thermal stability and it was registered with the U.S. Environmental Protection Agency and Health Canada Pesticide Management Regulatory Agency in 2020. Laboratory studies compared the thermostability and dissolution (i.e., TFM release) of the interim and new formulation of TFM bars that were held at 20 °C or 45 °C for 24 hours prior to evaluation. Field tests compared the dissolution of the interim and new formulation of TFM bars when applied in three small tributaries in Michigan. Laboratory tests show that the new formulation bars remain usable when held at 45 °C for 24 hours; whereas, the interim formulation bars partially liquify and are not usable. Field tests indicate the new formulation bars have superior characteristics including a near consistent release of TFM for 10–13 hours when applied in waters with a velocity of < 0.06 m/sec. A near consistent release of TFM was observed for a maximum of about 6 hours in one field application of the interim formulation bars. Water temperature and water velocity influenced both formulations; however, the greatest effects were observed with interim formulation bars where higher initial TFM concentrations were followed by precipitous TFM concentration decreases in tributaries with the highest water temperature or velocity. Field treatment applications will provide data for refining application parameters such as the number of bars required per unit discharge at various water temperatures and the acceptable water velocity range for applications.

Key words: TFM, control, formulation, lampricide, pesticide, Great Lakes

Introduction

Improvements to the Welland Canal in the early 1900s created a route for parasitic sea lamprey (*Petromyzon marinus* Linnaeus, 1758) to bypass the geological barrier at Niagara Falls and enter the upper Great Lakes (Hartman 1973; McDonald and Kolar 2007; Nowicki et al. 2021). As a result of the invasion, the commercial and recreational fisheries in the Great Lakes collapsed within a few decades (Smith 1980; Siefkes 2017;

Wilkie et al. 2019). Efforts to remediate the devastating effects of the sea lamprey included an extensive 1950s era chemical screening program that identified the halogenated nitrophenol TFM (4-nitro-3-(trifluoromethyl)-phenol) as a selective sea lamprey pesticide (Applegate et al. 1957). Treatments of sea lamprey larval nursery streams with TFM and the construction of barriers to block sea lamprey spawning migration became the primary means to control sea lamprey populations in the Great Lakes (Birceanu et al. 2021; Christie and Goddard 2003; Wilkie et al. 2019). The combined use of routine TFM applications and barriers effectively reduced the number of spawning-phase lamprey approximately 90% (Lavis et al. 2003; Morse et al. 2003; Siefkes 2017; Wilkie et al. 2019). The sustained reduction of parasitic lamprey using these methods has allowed for the rehabilitation and the continued preservation of the multibillion-dollar Great Lakes fishery (Great Lakes Fishery Commission 2014; Birceanu et al. 2021). During stream treatments, TFM is applied continuously for approximately 12 hours, resulting in a 9-hour treated-water block at concentrations lethal to larval sea lamprey (Boogaard et al. 2015; Lantz et al. 2019). Connected waterways must be treated concurrently with the passage of the mainstem treated-water block so that larval sea lamprey do not sense and then escape into untreated waters and survive (Luoma et al. 2020; Barber and Steeves 2021; Schloesser et al. 2021). In the mid-1980s, a solid TFM bar formulation was developed as a labor-saving tool for treating low-discharge tributary streams (Gilderhus 1985). The bars were formulated to release TFM from an inert carrier matrix over an 8–10 hour period (Gilderhus 1985). Subsequently several matrix inert ingredients were discontinued, which necessitated their replacement with similar, but not identical, products (Luoma et al. 2020). TFM bars manufactured using the latest interim formulation unacceptably soften when stored at temperatures ≥ 40 °C and they have unacceptable rapid dissolution in warm waters (Barber and Steeves 2021).

Scientists at the U.S. Geological Survey Upper Midwest Environmental Sciences Center reformulated TFM bars to address storage stability at elevated temperatures and to provide improved dissolution in warm waters. This new formulation was then registered with the U.S. Environmental Protection Agency (EPA) and Health Canada Pest Management Regulatory Agency (PMRA) in 2020.

Herein, we report the results of comparative laboratory and field studies conducted with commercially manufactured TFM bars made using the interim and the new formulations. Specifically, results are presented for (1) a laboratory study that was completed to compare the thermostability and decay (i.e., dissolve and release TFM) of the commercially manufactured bar formulations and (2) a field study that was completed to compare the decay of the commercially manufactured bars applied in three low-discharge tributaries of the Rifle River, Michigan.

Materials and methods

Consistent methods and equipment were used during the laboratory and field studies; these are described upon first use and referenced thereafter. Similar methods and equipment were used to collect water samples, measure water-quality constituents, and to analyze for TFM content/concentration.

Reference Articles

Two lots of reagent grade TFM were used as reference articles for the analysis of TFM during the laboratory and field studies (99.9%, Lot MA0301602, Acros Organics, Carlsbad, California and 99.9%, Lot MKBX5239V, Sigma-Aldrich Chemical Company, Milwaukee, Wisconsin, respectively).

Test Articles

TFM bars for the laboratory and field trials were produced by Iofina Chemical Company (Covington, Kentucky) in 2021 using the interim and new formulations (lot numbers TFBA210430A and TFMA2103331A, respectively). The interim formulation contains 23% TFM and a proprietary inert carrier matrix consisting of two polyoxypropylene-polyoxyethylene block copolymers, an ethoxylated fatty alcohol, and a non-ionic nonylphenol ethoxylate. The new formulation contains 23% TFM and a similar proprietary inert carrier matrix consisting of equivalent polyoxypropylene-polyoxyethylene block copolymers; however, the fatty alcohol and the nonylphenol ethoxylate surfactants were replaced with an alkoylated ethylenediamine block copolymer surfactant having a higher hydrophile-lipophile balance and a higher molecular weight. The ratio of the new carrier matrix formulants was optimized for performance characteristics, and all formulants are exempt from the need for a tolerance for non-food and food use under Title 40 Part 180 of the Code of Federal Regulations (CFR 2020).

TFM bar active ingredient concentration and thermostability

Three bars of each formulation were analyzed in triplicate to determine thermostability and active ingredient content. Bar hardness was used to assess thermostability during warm weather holding and was measured using a digital fruit penetrometer (model FHT-1122, Shenzhen Graigar Technology Co., Ltd, Baoan District, Shenzhen, China). Hardness was measured on bar samples held at room temperature (20 ± 2 °C) and bar samples held at 45 ± 1 °C for 24 hours.

The active ingredient concentration in bar samples was determined using an established high-performance liquid chromatography (HPLC) method for TFM analysis as described by Luoma et al. (2020). TFM analyses were conducted using an Agilent model 1260 HPLC (Agilent Technologies Inc., Santa Clara, California) equipped with a diode array

detector. TFM concentrations were determined by comparing sample responses to a linear regression created from five analytical standards. TFM bar samples were prepared for active ingredient concentration determination by dissolving a known mass (~ 0.5 g) of bar in 500 mL of HPLC-grade methanol, resulting in a product solution of approximately 1,000 mg/L. Solutions were further diluted in ultra-pure water (i.e., 18 MΩ-cm) to yield final product concentrations of approximately 10 mg/L.

Gradient elution was applied for all HPLC analyses using two 10-millimolar ammonium acetate mobile phase constituents (A and B) that were buffered with 3 mL of acetic acid per liter. Constituent A was prepared in a 3:1 mixture of ultra-pure water and LC/MS-grade methanol and Constituent B was prepared in LC/MS-grade methanol. A 1.25 mL per minute mobile phase flow rate was maintained for the duration of the 1.5- minute run. The initial mobile phase A:B ratio was 75:25, at time 0.0 minute the A:B ratio was immediately switched to and sustained at 60:40 until 0.5 minute. At time 0.5 minute, a 0.1-minute linear gradient change in the mobile phase ratio was applied, resulting in a final A:B ratio of 75:25 for the remainder of the run. Samples were separated with a Kinetex XB-C18 column (3 × 50 mm, 2.6-µm particle size) maintained at 50 °C. Sample absorbance was determined at 295.0 nm (10.0-nm bandwidth), and normal gradient analysis influences were accounted for using a reference absorbance at 350.0 nm (80.0-nm bandwidth). Method precision was verified using laboratory-fortified samples that were prepared immediately prior to analyses of the experimental samples. Laboratory-fortified samples were prepared by diluting a known mass of reagent grade TFM in HPLC-grade methanol to create a 100-mg/L stock solution that was further diluted with ultra-pure water to achieve a final fortified sample concentration of 1.0 mg/L. The fortified samples were analyzed with the experimental samples.

Laboratory trials

Replicate samples of the interim and new bar formulations were compared for decay (i.e., TFM release) in controlled laboratory dissolution trials. To mimic exposure to warm field storage conditions, dissolution trials were also completed on bar samples that were held at an elevated temperature. All laboratory dissolution trials were completed in a 6.8-m long test flume constructed from plastic pipe as described by Luoma et al. (2020; Figure 1). Three replicate 24-hour dissolution trials were performed with each formulation in the flume using bar samples that were either (1) held at 20 ± 2 °C prior to testing, or (2) held at 45 ± 1 °C for 24 hours then cooled to ambient room temperature prior to testing. Test samples were cut from bars of each formulation using a hole saw (pilot bit removed) and a knife. Test samples were individually weighed and were approximately 6.0 cm diameter, 0.9 cm thick, and 38.0 g.
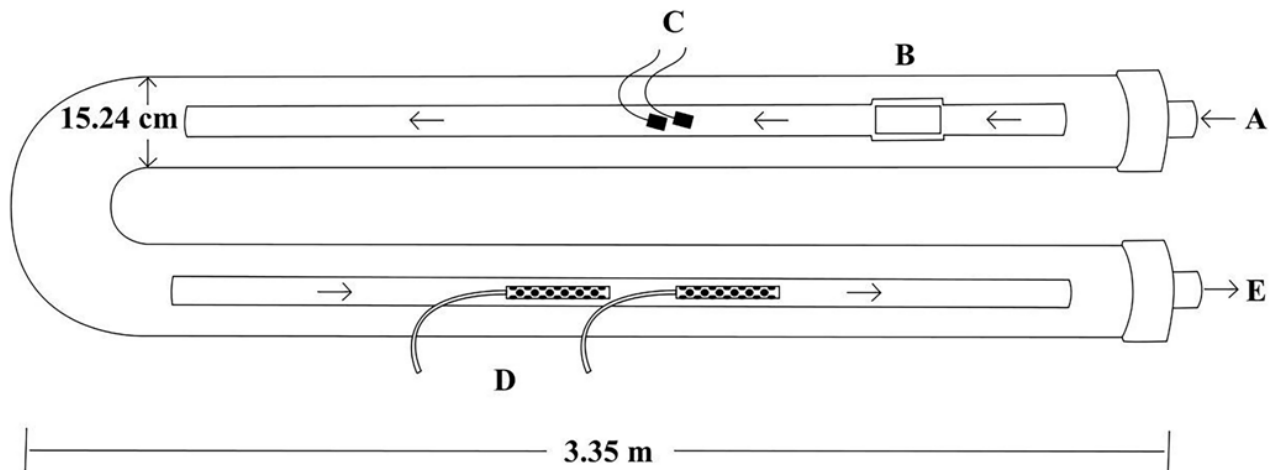
Figure 1. A schematic of the flow-through flume used for laboratory TFM bar dissolution trials showing the influent port (A), the elevated platform where the TFM bar samples were placed (B), airstones used to mix the TFM and water (C), sample collection lines (D), and the effluent port (E). (Figure from Luoma et al. 2020).

Well water was temperature-adjusted to 17.0 ± 0.5 °C and supplied to the flume at a flow rate of 10.7 to 11.1 L/minute, which resulted in a water velocity of 0.040 ± 0.002 m/sec. Direct measurement of water velocity was not possible because of the small flume size. Therefore, water velocity was derived prior to trial initiation and at 6, 12, and 24 hours using two equations. Equation 1 (Gordon 2008) was used to calculate the cross-sectional surface area of the flooded portion of the flume, then equation 2 (Levesque and Oberg 2012) was used to calculate the water velocity using the result from equation 1 and the measured volume of water discharged per second.

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

Where,

A cross-sectional surface area of the flooded portion of the pipe (m²).
r pipe radius (m), and
h height of the water in the pipe (m).

$$V = \frac{q}{A} \qquad (2)$$

Where,

V water velocity (m/sec),
q discharge (m³/sec), and
A flooded cross-sectional surface area derived from equation 1 (m²).

Water sample collection and TFM concentration verification

Water samples were collected from the flume every 30 minutes over each 24-hour trial using two automated water samplers (model #3700, Teledyne-Isco, Inc, Lincoln, Nebraska). Water samples were analyzed for TFM content in triplicate using the previously described HPLC analysis methods.
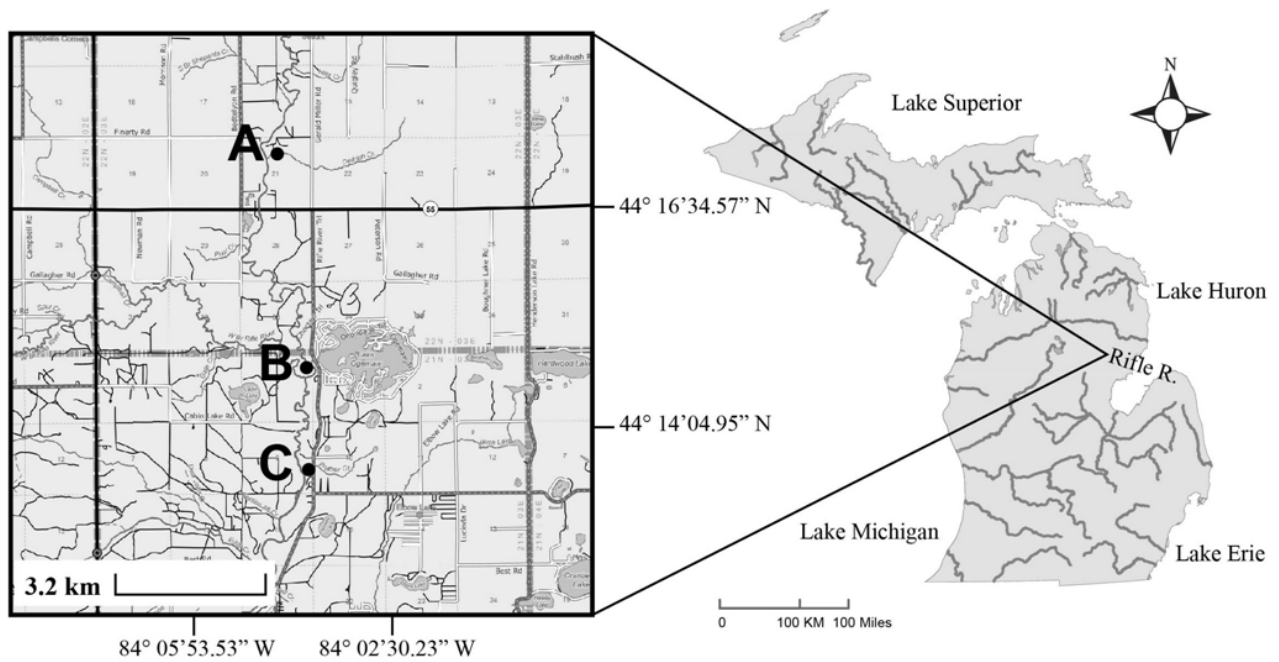
Figure 2. Location of three low-discharge tributaries of the Rifle River in Michigan used in trials to compare the interim and new formulations of TFM bars. The tributaries included Dedrich (A), Peterson (B), and Prather (C) Creeks.

Water Quality

Water-quality constituents including dissolved oxygen (DO), pH, temperature, and conductivity were measured prior to trial initiation, hourly throughout the first 12 hours, and at 24 hours. Alkalinity and water hardness were measured prior to trial initiation and at 6, 12, and 24 hours. Temperature was measured with a digital thermometer (Thermapen model Mk4, ThermoWorks Company, American Fork, Utah). DO, pH, and conductivity were measured using a water-quality meter equipped with an optical DO probe, a digital pH probe, and a conductivity probe (models HQ40d, LDO 10101, PHC 70501, and CDC 401; Hach Company, Loveland, Colorado). Total water hardness was determined by titrating with ethylenediaminetetraacetic acid according to method 2340C, and alkalinity was determined by titrating to an endpoint of pH 4.5 with 0.02N H_2SO_4 according to method 2320B, as described in Baird et al. (2017), respectively.

Field trials

TFM bars were applied in three low-discharge tributaries of the Rifle River (Ogemaw and Arenac County, Michigan) in conjunction with a routine lampricide treatment during August 2021 (Figure 2).

A TFM concentration of 1.0 mg/L was targeted in all tributaries using the application procedures described in the Standard Operating Procedures for the Application of Lampricides in the Great Lakes Fishery Commission Integrated Management of Sea Lamprey Control Program manual (hereafter referred to as the SOP manual; Barber and Steeves 2021) with the exception that the number of bars applied was rounded to the nearest

0.5 bar instead of the nearest whole bar (i.e., bars were cut in half as necessary). The 1.0-mg/L target concentration was at or below the expected sea lamprey minimum lethal concentration in all tributaries according to the pH and alkalinity specific treatment charts in the SOP manual (Barber and Steeves 2021). The 1.0-mg/L TFM target concentration was sufficient to achieve study objectives, and the consistent target concentration eliminated potential confounding effects.

A randomization scheme was used to determine the application order of the TFM bar formulations in each tributary. Tributaries were treated with one formulation type on consecutive days, and the number of TFM bars applied was calculated using the discharge measured prior to application. TFM bars were applied directly on creek beds, which were composed of either hard sand, gravel, or small cobble, at a rate of four bars per cubic foot of discharge in accordance with procedures described in the SOP manual (Barber and Steeves 2021). A FlowTracker 2 acoustic doppler velocimeter (SonTek – a Xylem brand, San Diego, California) fitted with a top mounting wading rod was used to measure water velocity at 0.6 depth (Xylem 2019; Barber and Steeves 2021) and to determine the discharge prior to application and at 6, 12, and 24 hours. Discharge within the treated reach was determined by establishing measurement points along a tributary cross-section and then creating subsections between each measurement point. The average velocity of each subsection was calculated by averaging the velocity measured at adjacent measurement points. No more than 10% of the discharge was measured within a subsection. Discharge was calculated using the mean-section method according to equation 3 (Xylem 2019).

$$Discharge = \sum (b_1 - b_0)\left(\frac{d_1 + d_2}{2}\right)\left(\frac{\overline{V}_1 + \overline{V}_0}{2}\right) + (b_2 - b_1)\left(\frac{d_2 + d_1}{2}\right)\left(\frac{\overline{V}_2 + \overline{V}_1}{2}\right) + $$
$$(b_{n+1} - b_n)\left(\frac{d_{n+1} + d_n}{2}\right)\left(\frac{\overline{V}_{n+1} + \overline{V}_n}{2}\right) \qquad (3)$$

Where,
    $\overline{V}$ average 20-second water velocity at the measurement point (m/sec),
    d water depth at the measurement point (m), and
    b location of the measurement point.

Water sample collection and TFM concentration verification

Water samples were collected every 30 minutes during each 24-hour field trial using automated water samplers that were placed 35–75 m downstream from the application points. Sampling sites were selected based on the morphology and hydrology of the tributaries. Sampling sites were located downstream from areas such as riffles and/or constrictions that provided thorough mixing. No additional water sources were visible between the application and sampling points. Uniform mixing in the tributaries was confirmed at the sampling locations by comparing the absorbance of grab water samples collected from near the left bank, right bank, and thalwag.

Absorbance was measured at 295 nm using a portable spectrophotometer (model Genesys 20, Thermo Scientific, Waltham, Massachusetts). Triplicate subsamples of each water sample were stored at −20 °C and then analyzed for TFM using the previously described methods. Field-fortified tributary water samples were prepared, stored identically to the experimental samples, and used to verify sample stability. Field-fortified samples were prepared by diluting a known mass of reagent grade TFM in HPLC-grade methanol to create a 100-mg/L stock solution that was further diluted with tributary water to achieve a final fortified sample concentration of 1.0 mg/L.

Water quality

Water-quality constituents including DO, pH, temperature, and conductivity were measured *in situ* upstream from the application points prior to TFM bar application, then hourly for the first 12 hours, and again at 24 hours. Alkalinity and water hardness were measured on grab water samples collected prior to each application and at 6, 12, and 24 hours. Water-quality instrumentation and the methods used in the field study were the same as those described for the laboratory dissolution trials.

Data analysis

Metadata, analysis code, and data for this study are publicly available at https://doi.org/10.5066/P910SHBL (Schueller et al. 2022). Data were analyzed using R and RStudio software (version 4.1.1, R Core Team 2021 and version 2021.09.0, build 351, Rstudio Team 2021, respectively). TFM bar active ingredient concentrations, TFM bar hardness, water-quality constituents, water discharges, and water velocities were summarized using simple descriptive statistics for the laboratory and field trials. Quantile-quantile plots were used to assess data for homoscedasticity and normality prior to conducting statistical analyses and $\alpha < 0.05$ was the criterion used to designate statistical differences. TFM concentrations were normalized using the measured TFM bar active ingredient concentrations and the discharge.

Differences in active ingredient content and bar hardness between the formulations, laboratory and field water-quality constituents, water discharges, and water velocities were detected using one-way analysis of variance tests that accounted for replicates (Bates et al. 2015). *Post-hoc* Tukey Honest Significant Difference tests were used to determine which properties differed (Montgomery 2017).

Exponential decay curves were fit to the TFM concentration data for each laboratory and field trial and modeled according to OECD (2006) using the DRC package (Ritz et al. 2015). Model fitness was assessed by calculating the median absolute deviation (MAD) for each independent trial. The MAD was calculated as the median of the absolute value of the distance between the calculated and the model predicted percentage of TFM bar(s) remaining.

Table 1. Mean (standard deviation, SD) hardness (kg/cm^2) and percent active ingredient concentration of the interim and new TFM bar formulations.

Formulation	Mean (SD) Hardness at 20 °C (kg/cm^2)	Mean (SD) Hardness at 45 °C (kg/cm^2)	20 °C Treatment Active Ingredient Concentration (%)	45 °C Treatment Active Ingredient Concentration (%)
Interim	6.38[a] (0.28)	0.50 [a,c] (0.00)	22.01[d] (0.28)	21.71[d] (0.27)
New	19.06[b] (0.24)	3.60[b] (0.17)	23.39[e] (0.41)	23.63[e] (0.27)

[a, b] Hardness values within columns with same letter did not differ, $\alpha = 0.05$
[c] Samples were partially liquified.
[d, e] Active ingredient concentrations within rows with the same letter did not differ, $\alpha = 0.05$.

The predicted decay time to achieve a 25, 50, 75, and 90% reduction (DT$_{25}$, DT$_{50}$, DT$_{75}$, and DT$_{90}$, respectively) was determined from each model. Differences in decay times among treatment groups were detected using one-way analysis of variance tests, and *post-hoc* Tukey Honest Significant Difference tests were used to identify specific differences. Laboratory treatment groups were designated by formulation type and temperature treatment. Field treatment groups were designated by formulation type and stream.

Results

Method precision and retention of sample integrity were confirmed with mean TFM recovery in laboratory-fortified and field-fortified samples of 99.8 ± 1.8% and 98.4 ± 1.8%, respectively. The use of the exponential decay function to model the release of TFM from the bars during the laboratory and field trials was validated by inspecting the MAD value for each trial. Across all trials, the MAD values indicate that variations between the calculated and the predicted amount of TFM bars remaining differed by less than 4.0%.

TFM bar active ingredient concentration and thermostability

The mean percentage of active ingredient of the bars stored at room temperature was 22.01 ± 0.28% and 23.39 ± 0.41% for the interim and new bar formulations, respectively (Table 1). Analysis of the active ingredient concentrations indicated differences between formulations ($F_{(3,32)} = 86.28$, $p < 0.01$), but samples of the interim and new formulations stored at room temperature did not differ from corresponding samples that were subjected to the elevated temperature treatment ($p = 0.26$ and $p = 0.41$, respectively; Table 1).

Laboratory Trials

Water-quality constituents, water velocity, and water discharge in the laboratory trials did not differ between formulation type and temperature treatment ($F_{(3, 8)} = 0.152–1.178$), $p \geq 0.38$; Table 2). The maximum TFM concentrations measured during laboratory trials was nearly 50% more in

Table 2. Mean (standard deviation, SD) water-quality and flow characteristic by formulation and treatment type during laboratory dissolution trials[a].

Treatment Type	Formulation	Mean (SD) Temp.(°C)	Mean (SD) Discharge (m^3/min)	Mean (SD) Velocity (m/sec)	Mean (SD) Dissolved Oxygen (mg/L)	Mean (SD) pH (standard units)	Mean (SD) Hardness (as $CaCO_3$)	Mean (SD) Alkalinity (as $CaCO_3$)	Mean (SD) Conductivity (μS/cm)
20 ± 2 °C	Interim	17.0 (0.1)	0.011 (< 0.001)	0.04 (< 0.01)	8.0 (0.5)	7.82 (0.13)	184 (4)	145 (10)	413 (5)
	New	17.0 (0.2)	0.011 (< 0.001)	0.04 (< 0.01)	8.2 (0.3)	7.79 (0.08)	183 (4)	143 (5)	415 (3)
45 ± 1 °C	Interim	17.1 (0.1)	0.011 (< 0.001)	0.04 (< 0.01)	8.1 (0.6)	7.76 (0.17)	186 (6)	143 (9)	419 (7)
	New	17.0 (0.1)	0.011 (< 0.001)	0.04 (< 0.01)	8.3 (0.6)	7.85 (0.18)	187 (4)	150 (9)	420 (6)

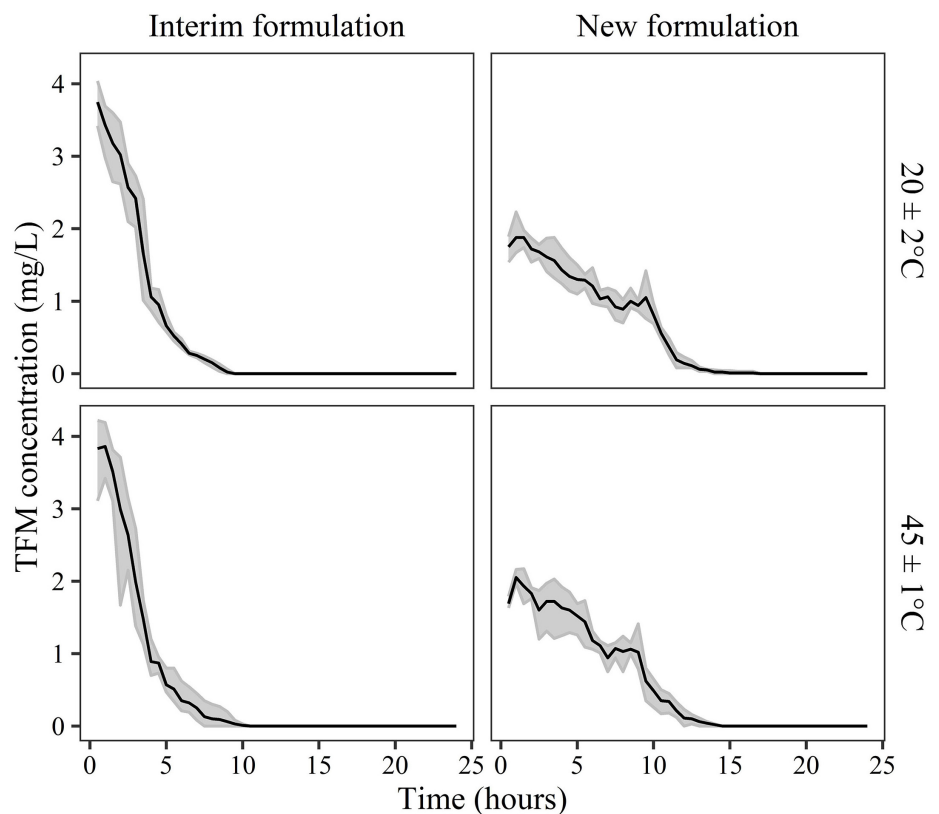[a] Values within columns did not differ, α = 0.05.

Figure 3. Mean and range (shaded area) TFM concentration measured in effluent water by formulation and treatment type for trials conducted with TFM bar samples that were either (1) held at 20 ± 2 °C or, (2) held at 45 ± 1 °C for 24 hours and then cooled to ambient room temperature, prior to testing in a laboratory flume.

trials conducted with the interim formulation, and the interim formulation also decayed faster than the new formulation (Figure 3, Table 3). The elevated temperature treatment of 45 °C for 24 hours did not appreciably alter the observed TFM concentration in the small-scale laboratory test; however, the decay of both formulations was approximately 3.4 to 5.4% faster in the elevated temperature treatment groups (Table 3).

The time for TFM bar samples to decay was considerably slower for the new formulation compared to the interim formulation (Table 3). The predicted DT_{25}, DT_{50}, DT_{75} and DT_{90} of the TFM bars differed among dissolution trial treatment groups ($F_{(3,8)}$ = 112.1–327.3). Differences were

Table 3. Predicted mean (standard deviation, SD) time to achieve 25, 50, 75, and 90% decay (DT$_{25}$, DT$_{50}$, DT$_{75}$, and DT$_{90}$, respectively) of the TFM bar samples in the laboratory flume by formulation and treatment type.

Formulation	Treatment Type	Predicted mean (SD) DT$_{25}$ (h)	Predicted mean (SD) DT$_{50}$ (h)	Predicted mean (SD) DT$_{75}$ (h)	Predicted mean (SD) DT$_{90}$ (h)
Interim	20 ± 2 °C	0.74[a] (0.06)	1.62[a] (0.10)	3.11[a] (0.15)	5.03[a] (0.20)
	45 ± 1 °C	0.70[a] (0.07)	1.55[a] (0.22)	2.99[a] (0.48)	4.85[a] (0.81)
New	20 ± 2 °C	1.77[b] (0.02)	3.74[b] (0.07)	6.92[b] (0.14)	10.58[b] (0.20)
	45 ± 1 °C	1.71[b] (0.05)	3.56[b] (0.17)	6.58[b] (0.37)	10.10[b] (0.55)

[a, b] Values within columns with the same letter did not differ, $\alpha = 0.05$.

Table 4. Mean (standard deviation, SD) water-quality and flow characteristics of Rifle River (Michigan) tributaries, by creek and formulation type, used to compare field applications of two different TFM bar formulations.

Creek	Formulation	Mean (SD) Temp. (°C)	Mean (SD) Discharge (m³/sec)	Mean (SD) Velocity (m/sec)	Mean (SD) Dissolved Oxygen (mg/L)	Mean (SD) pH (standard units)	Mean (SD) Hardness (as CaCO$_3$)	Mean (SD) Alkalinity (as CaCO$_3$)	Mean (SD) Conductivity (μS/cm)
Peterson	Interim	15.4[a] (1.1)	0.0452[a] (0.0009)	0.3147[a] (0.0241)	7.4[a] (0.2)	7.80[a] (0.06)	175[a] (6)	173[a] (3)	393[a] (3)
	New	16.2[a] (1.9)	0.0458[a] (0.0014)	0.2860[a] (0.0411)	7.4[a] (0.2)	7.90[a] (0.08)	187[a] (4)	177[a] (6)	392[a] (3)
Prather	Interim	16.3[a] (1.2)	0.0088[b] (0.0007)	0.0569[b] (0.0121)	7.9[a,b] (0.3)	8.07[a,b] (0.05)	172[a] (8)	165[a] (3)	342[b] (3)
	New	15.8[a] (0.7)	0.0088[b] (0.0006)	0.0581[b] (0.0056)	8.0[a,b] (0.2)	8.01[a,b] (0.03)	163[a] (5)	165[a] (4)	342[b] (3)
Dedrich	Interim	21.1[b] (1.6)	0.0167[c] (0.0019)	0.0466[b] (0.0089)	8.4[b] (0.9)	8.20[b] (0.12)	228[b] (6)	221[b] (3)	439[c] (7)
	New	20.5[b] (1.7)	0.0148[c] (0.0011)	0.0440[b] (0.0076)	9.0[b] (0.9)	8.31[b] (0.13)	229[b] (5)	227[b] (3)	450[c] (5)

[a,b,c] Values within columns with the same letter did not differ, $\alpha = 0.05$.

detected by formulation type ($p < 0.01$) but not between temperature treatments of the same formulation (i.e., 20 °C versus 45 °C; $p > 0.55$). Across all trials, the new formulation required 2.1–2.4 times longer to decay than the interim formulation (Table 3).

Field Trials

Water-quality constituents and streamflow characteristics were similar in trials conducted within each tributary with minimal differences observed in parameters known to influence TFM bar decay – water temperature and water velocity (i.e., within tributary differences were < 5% for temperature and < 9.6% for velocity; Table 4). In contrast, water temperature, discharge, and water velocity among tributaries differed when analyzed with consideration of formulation type ($F_{(2,3)} = 74.65–1084.70$, $p < 0.03$). Although the water temperature variance among streams was only about 5 °C, the water temperature was warmer in Dedrich Creek compared to the water temperatures in Peterson and Prather Creeks ($p < 0.01$). The water temperatures in Peterson and Prather Creeks did not differ ($p = 0.88$). Water velocity and discharge differed among all tributaries ($p < 0.01$) except the water velocity in Prather Creek did not differ from the water velocity in Dedrich Creek ($p = 0.61$). Many, but not all, of the other water-quality constituents also differed among tributaries, with the most notable
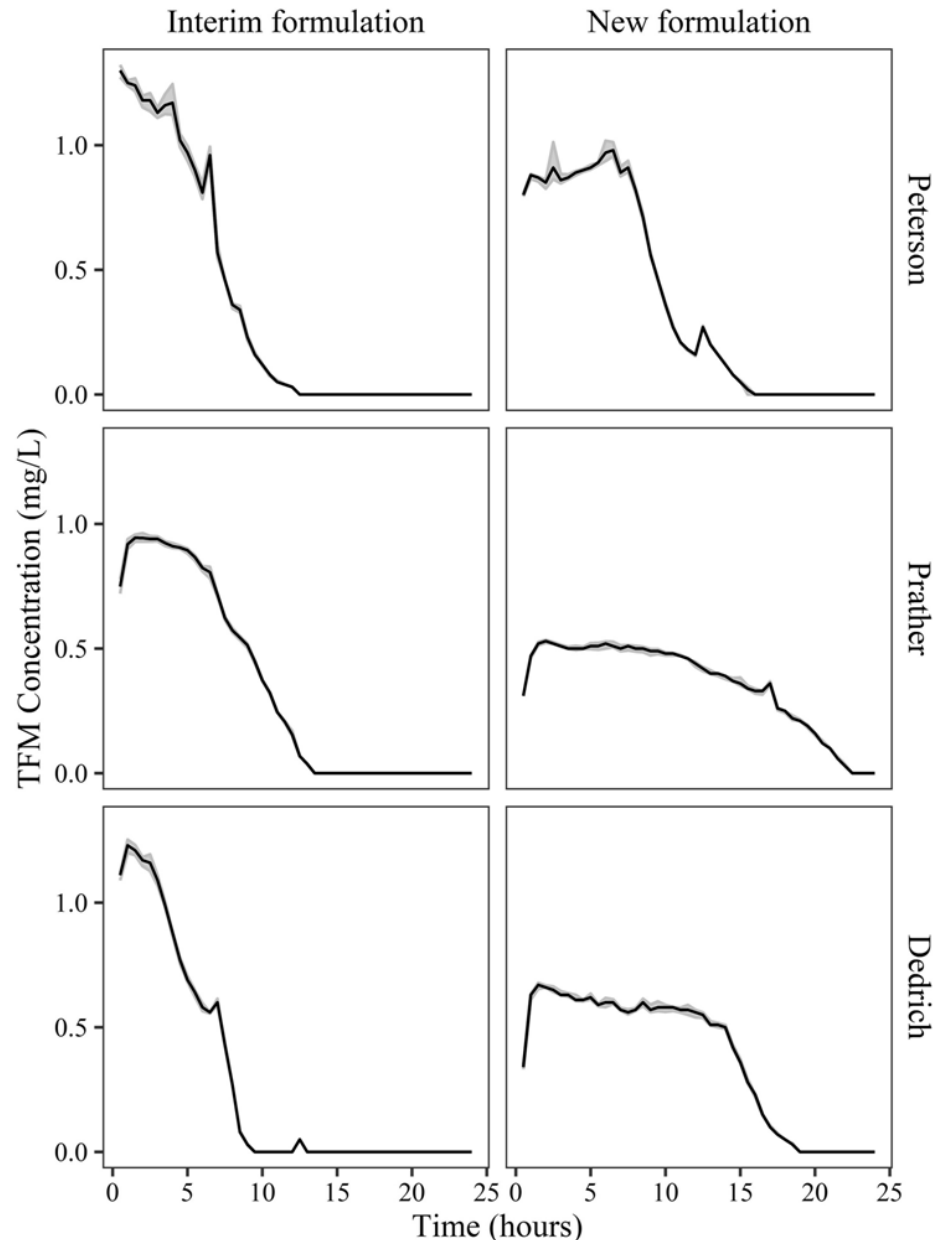
Figure 4. Mean and range (shaded area) TFM concentration measured in water samples by creek and formulation type during field trials conducted in three tributaries of the Rifle River in Michigan.

differences among comparisons of Dedrich Creek to Peterson and Prather Creeks (Table 4).

The new formulation yielded considerably lower maximum TFM concentrations, periods of relatively consistent TFM release, and longer durations of detectable amounts of TFM (Figure 4). The time for the TFM bars to decay 25–90% was greater for the new formulation compared to the interim formulation ($F_{(1/4)} = 7.87$–9.42, $p < 0.05$; Table 5). The new TFM bar formulation required an average of about 1.5–2.5 times longer to decay than the interim formulation when applied in the same tributary (Table 5).

Table 5. Predicted time required to decay 25, 50, 75, and 90% (DT$_{25}$, DT$_{50}$, DT$_{75}$, and DT$_{90}$, respectively) of the TFM bars in field trials by creek and formulation.

Creek	Formulation	Predicted DT$_{25}$ (h)	Predicted DT$_{50}$ (h)	Predicted DT$_{75}$ (h)	Predicted DT$_{90}$ (h)
Peterson	Interim	1.44	2.96	5.49	8.54
	New	2.20	4.41	7.93	11.78
Prather	Interim	1.95	3.93	7.12	10.74
	New	4.06	8.24	13.65	17.87
Dedrich	Interim	1.23	2.56	4.78	7.51
	New	3.38	6.78	11.57	15.76

Discussion

The new formulation for the TFM bars improved environmental compatibility by replacing the nonylphenol ethoxylate surfactant in the carrier matrix with a tolerance exempt alkoylated ethylenediamine block copolymer surfactant. Nonylphenol ethoxylate surfactants are widely used in commercial and consumer products; however, they are under increased scrutiny because of the aquatic toxicity of their degradants (EPA 2012).

The range of water temperatures during the field trials was limited (i.e., ~ 15–21 °C), and performance at lower water temperatures would need further evaluation to determine cold water application strategies required to achieve desired concentrations. Cold water application strategies could include (1) increasing the number of bars applied, (2) applying the bars in higher velocity areas, or (3) increasing the exposed surface area of the bars by breaking the bars or producing and applying smaller bars. Although additional evaluation of cold water applications would be beneficial, this study did demonstrate that the new TFM bar formulation has improved warm weather performance characteristics compared to the interim formulation. For example, when held under a simulated mid-summer high air temperature of 45 °C for 24 hours, the new formulation was softened but remained intact and suitable for use, whereas the interim formulation was partially liquified and unsuitable for use (Table 1). Furthermore, new formulation bars released TFM for nearly twice the duration as the interim formulation in laboratory and field dissolution trials conducted at water temperatures of 17 °C and ~ 20 °C, respectively (Tables 3 and 5).

The slower decay of the new formulation resulted in lower initial TFM concentrations, which were followed by slower concentration declines (Figure 3). In the laboratory trials, the DT$_{75}$ and DT$_{90}$ with the interim and new TFM bar formulations were approximately 3 and 5 hours and 7 and 10 hours, respectively (Table 3). The laboratory trials indicate that the new formulation could maintain effective TFM concentrations for at least 7 hours in systems with similar flow and temperature conditions. The laboratory trials with the interim formulation confirmed prior poor field performance and indicate that sustaining efficacious TFM concentrations for a suitable period is unlikely when applied in tributaries with similar conditions (Figure 3, Table 3).

The field trials enabled evaluation of the formulations under differing water temperature and water flow conditions. Water temperatures in Peterson and Prather Creeks were similar during the field trials ($\bar{x}$ = 15.4–16.3 °C) but colder than those in Dedrich Creek ($\bar{x}$ = 20.5 and 21.1 °C). Peterson Creek had considerably higher discharge and water velocity compared to Prather and Dedrich Creeks (Table 4). Water velocity and temperature had the most influence on TFM bar decay. The positive correlation of water velocity on TFM bar decay is demonstrated by the higher initial TFM concentrations and the faster concentration declines observed in Peterson Creek compared to Prather Creek (Figure 4). The positive correlation of water temperature on TFM bar decay is demonstrated by the higher initial TFM concentrations and the sharper concentration declines observed in Dedrich Creek compared to Prather Creek (Figure 4). The TFM concentration profiles for Dedrich and Prather Creeks demonstrate that the new formulation can release TFM relatively consistently for about 10–13 hours when applied in waters with a velocity of < 0.06 m/sec (Figure 4). Application of the new formulation bars in waters with a velocity of approximately 0.3 m/sec in Peterson Creek reduced the time the TFM release was relatively consistent to approximately 8 hours (Figure 4). The interim formulation bars demonstrated relatively consistent TFM release for a maximum of about 6 hours in Prather Creek (mean velocity ~ 0.06 m/sec; Figure 4). The more rapid decreases in the TFM concentration observed during the interim formulation trials can be attributed to the higher water velocity in Peterson Creek and the higher water temperature in Dedrich Creek (Figure 4).

The comparative laboratory and field studies between the interim and new TFM bar formulations demonstrated that (1) the new bar formulation is stable when stored at temperatures up to 45 °C, and (2) the new bar formulation can provide a relatively consistent release of TFM for ≥ 8 hours when applied in low-velocity waters. Additional field applications and data collection by Sea Lamprey Control Program personnel would allow for determining the optimal application rates (i.e., number of bars per cubic meter of discharge) required to achieve the desired TFM concentrations over a range of water velocities and water temperatures.

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Authors' contribution

James A. Luoma: research conceptualization, sample design and methodology, data collection, data analysis and interpretation, manuscript preparation and editing; Justin R. Schueller: sample design and methodology, data collection, data analysis and interpretation, manuscript preparation and review; Nicholas A. Schloesser: sample design and methodology, data collection, manuscript preparation and review; Todd A. Johnson: sample design and methodology, data collection, manuscript preparation and review; Courtney A. Kirkeeng: sample design and methodology, data collection, manuscript preparation and review.

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