Electronic Supplementary Material (ESI) for Environmental Science: Processes & Impacts.

Electronic Supplementary Information: Laboratory measurements underestimate persistence of the aquatic herbicide fluridone in lakes

Sydney R. Van Frost[1], Amber M. White[2], Josie M. Jauquet[1], Angela M. Magness[3], Katherine D. McMahon[1,3], and Christina K. Remucal[1,2*]*

[1]Department of Civil and Environmental Engineering

University of Wisconsin – Madison

Madison, WI 53706 USA

[2]Environmental Chemistry and Technology Program

University of Wisconsin – Madison

Madison, WI 53706 USA

[3]Department of Bacteriology

University of Wisconsin – Madison

Madison, WI 53706 USA

Contents (17 pages): Figures: S1-S7, Tables: S1-S6

* Corresponding author addresses:

Christina Remucal: 660 N. Park St., Madison, WI 53705; e-mail: remucal@wisc.edu; telephone: (608) 262-1820.

Katherine McMahon: 1550 Linden Drive, Madison WI 53706; email: trina.mcmahon@wisc.edu; telephone: (608) 890-2836.

Table of Contents

Section S1. Summary of previous fluridone studies

Table S1. Summary of literature reports of field, photodegradation, biodegradation, and sorption studies of fluridone.

Half-life	Study type	Ref. #
Field Studies		
2 – 11 days	Three ponds and one lake in MI, NY, and FL, USA; Panama Canal	1
4 – 7 days	Small ponds in Manitoba, Canada	2
5 – 60 days	Ponds in TX, WV, MO, CA, IN, and FL, USA	3
30 – 50 days	Small- medium ponds in Greenfield, IN, USA	4
17 weeks	Small artificial ponds in Manitoba, Canada	5
8 months	One lake in WI, USA	6
Photodegradation Studies		
15 – 36 hours	Ultrapure water, natural light, filtered > 297 nm	7
23 hours	Ultrapure water, simulated sunlight 280-365 nm	1
28 – 55 hours	Ultrapure water, simulated sunlight 280-365 nm	8
35 hours	Ultrapure water, natural sunlight, 325-355 nm	7
8.8 days	Ultrapure water, simulated sunlight, 310-380 nm	7
7 days	Well water, natural sunlight	9
12 days	Ultrapure & lake water, natural sunlight	8
33 days	Well water, natural sunlight, 290 - 320 nm light filtered out	9
Biodegradation Studies		
50 days	Microcosms with silty and sandy soil, saturated with tap water	10
>150 days	Microcosm with lake sediment	11
>150 days	Cultures enriched from lake sediments	11
12 months	Culture flasks with three sediment types	5
Sorption Studies		
10% sorbed in 30 days	Silty and sandy soil, saturated with tap water	10
16-27% sorbed in 28 days	Pond application in NY and FL, USA	1
14-52% sorbed in over 150 days	Pond sediments in Manitoba, Canada	5
K_{oc}: 350-2462 L kg^{-1}	Pond sediments in Manitoba, Canada	2, 12

Section S2. Field sampling methods

Lake water was collected from five lakes for bulk water chemistry measurements and photochemical irradiations. Surface water was collected from the open water and nearshore area in 4 L combusted glass amber bottles, filtered through a 0.45 μm nylon filter, and preserved at 4°C until analysis.

Table S2. Location, pH, dissolved organic carbon concentration, E_2:E_3 (absorbance at 250 nm divided by absorbance at 365 nm),[13] and $SUVA_{254}$ (specific UV absorbance at 254 nm; absorbance at 254 nm divided by [DOC])[14] for all lakes visited during field sampling.

Lake	Coordinates	pH	[DOC] (mg/L)	$SUVA_{254}$ (L mg-C^{-1} m^{-1})	E_2:E_3
Silver	45.92°N, 89.24°W	7.95	6.54	1.55	7.17
Pike	43.31°N, 88.33°W	8.21	7.18	1.83	9.24
Tomahawk	46.37°N, 91.52°W	8.02	4.52	1.06	10.06
Pleasant	42.79°N, 88.55°W	8.04	6.16	0.86	9.61
Hooker	44.56°N, 88.10°W	8.01	7.60	2.21	8.35

The parameters derived from UV-visible spectra indicate the DOM in the five study lakes is similar in composition. Specifically, the DOM is relatively low in aromaticity (i.e., low $SUVA_{254}$)[14] and in apparent molecular weight (i.e., moderate to high E_2:E_3).[13] These values are indicative of DOM that is microbial in origin or of terrestrially-derived DOM that has undergone environmental processing.

Pretreatment water from Hooker Lake used for microcosm incubations was collected with 10 L HDPE cubitainers and stored in the dark on ice until microcosm set up. Sediment for microcosm incubations and sorption experiments was collected by Eckman dredge or hand-coring at a nearshore site of each and stored in the dark on ice until microcosms set up which occurred within 24 hours.

Water samples collected during the fluridone treatment of Hooker Lake were stored on ice and in the dark until processing, which occurred no longer than 24 hours after collection.

Processing involved filtration through a 0.45 μm nylon filter and preservation at 4°C until analysis. Hooker Lake has a surface area of 0.4 km^2; therefore, water samples were collected at three sites with two being opposite nearshore sites and one location being in the center to characterize behavior throughout the lake. Samples were collected every 1-2 weeks until 60 days post-treatment.

Section S3. Photochemical irradiations

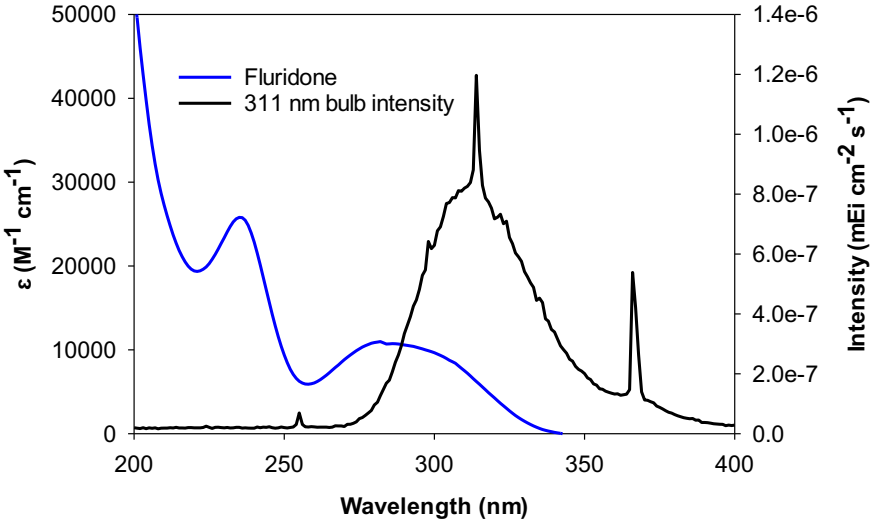

Figure S1. Molar absorptivity (ε) fluridone and the irradiance of the 311 nm bulbs used in photochemistry experiments (second y-axis).

The observed photodegradation rate constants (k_{obs}) were corrected for light screening in all solutions using by calculating a screening factor at each wavelength (S_λ):

$$S_\lambda = \frac{1-10^{-(\alpha_\lambda)(l)}}{2.303(\alpha_\lambda)(l)} \quad \text{Eq. S1}$$

where α_λ is the solution decadic absorbance measured using a UV-vis spectrophotometer and l is the pathlength of the cuvette (1 cm). An average weighted screening factor ($S_{weighted}$) was calculated from 250-455 nm and was used to correct the observed degradation rate constants for all lake waters and the direct control using Equation S2:

$$k_{screened} = \frac{k_{obs}}{S_{weighted}} \quad \text{Eq. S2}$$

The light absorbance rate constant (k_{abs}) was calculated using Equation S3:

$$k_{abs} = \Sigma \frac{2.303(I_\lambda)(\alpha_\lambda)(S_\lambda)}{([C])(j)} \quad \text{Eq. S3}$$

where I_λ is the intensity of light (mEi cm^{-2} s^{-1}), α_λ is the solution decadic absorbance, S_λ is the weighted screening factor, [C] is fluridone concentration (molar), and j is a conversion factor of 1 Einstein mol^{-1}.[15]

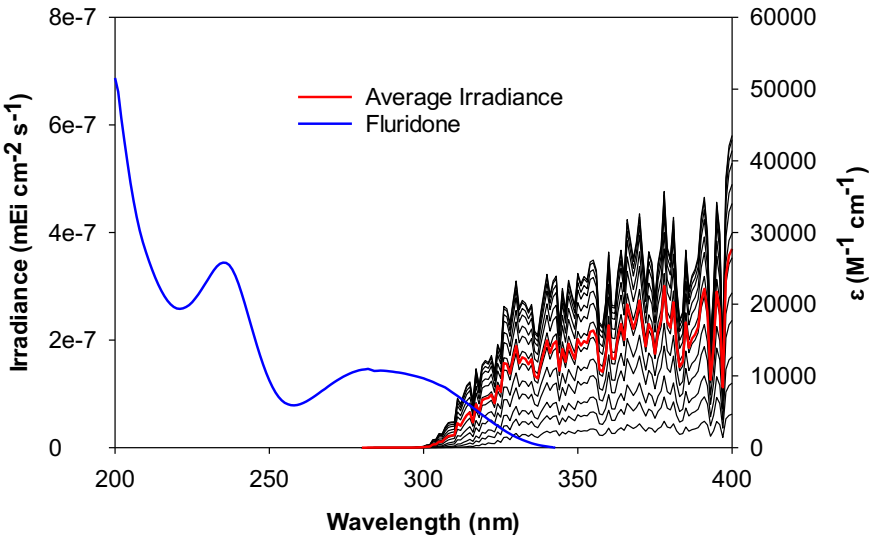

Figure S2. Average of the hourly irradiance at Hooker Lake during the day of treatment from 6 am to 6 pm (black lines). Global horizontal irradiance spectra at each time was generated using SMARTS.[16] The molar absorptivity of fluridone is plotted on the second y-axis.

Table S3. SMARTS modeling input parameters for the in-lake photolysis degradation of fluridone in Hooker Lake on the day of treatment.

Card Number/ Description	**Hooker Lake Parameters**
1. Comment	'Hooker'
1. Manually input pressure	1
2a. Pressure, surface altitude, and height	1013.25 0.229 0
3. Option to use default atmosphere	1
3a. Midlatitude Summer	'MLS'
4. Use default Water vapor	1
5. Use default ozone abundance	1
6. Use default gas abundance except CO_2	1
7. Carbon dioxide from June 2021 and 2022	417.46
7a. Use default synthetic spectrum	0
8.Use continental aerosol model	'SRA_CONTL'
9. Use aerosol optical depth of 55 nm	5
9a.	0.084
10. Select "water" for albedo	2
10b. ITILT	1
ITILT is an option for tilted surface calculations. Leave box unchecked	51 37. 180.
11. Minimal spectral range, max spectral range, variability in irradiance, and default solar constant.	280 4000 1.0 1366.1
12. Option to generate results with spreadsheet	2
12a: Interval for printing results	280 4000 1
12b. Total number of outputs	5
12c. Outputs: (1) extraterrestrial spectrum, (2) direct normal irradiance, (3) diffuse horizontal irradiance, (4) global horizontal irradiance and (5) direct horizontal irradiance	1 2 3 4 5
13. Bypass circumsolar radiation	0
14. Bypass smoothing calculation	0
15. Illuminance using CIE photopic curve	1
16. No special UV calculations	0
17. Set inputs for card 17	3
17a. Year, month, day hour, latitude, longitude, time zone.	2022 05 12 12.1 42.558952 -88.100541 -6

Section S4. Sorption experiments

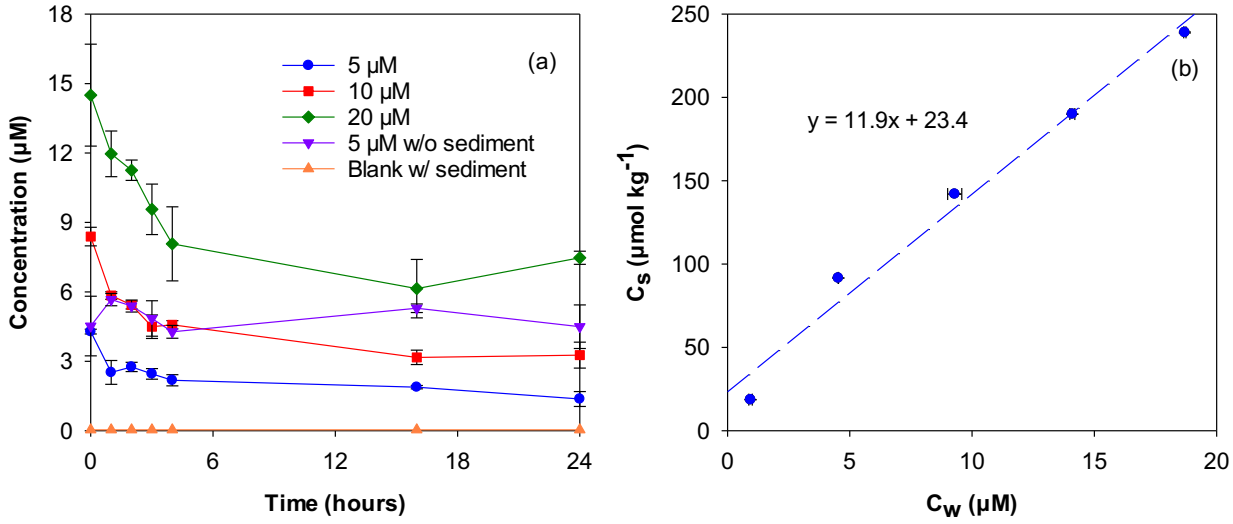

Figure S3. (a) Sorption kinetics of fluridone to lake sediment along with controls without sediment and the lake sediment with no fluridone. Fluridone with sediment shows equilibrium is reached by 6 hours of sediment interaction. Fluridone control without sediment shows little to no loss occurs through volatilization, photodegradation, or sorption to the glass bottles. Fluridone was not detected in the sediment control. (b) Fluridone concentration in sediment (C_s) versus fluridone concentration in water (C_w) after equilibration with Hooker Lake sediment for seven hours at pH 7. The slope of the isotherm is equivalent to the sediment sorption partitioning coefficient (K_d) in L kg^{-1}. Error bars represent the standard deviation of triplicate vessels.

Section S5. Microcosm incubations

The water only and water and sediment microcosms were initially combined via manual mixing and subsequently capped. Once mixed, the microcosms were incubated at room temperature (21 – 29 °C) and left stagnant. Headspace remaining in the glass jars resulted in aerobic environments and sampling introduced oxygen due to uncapping.

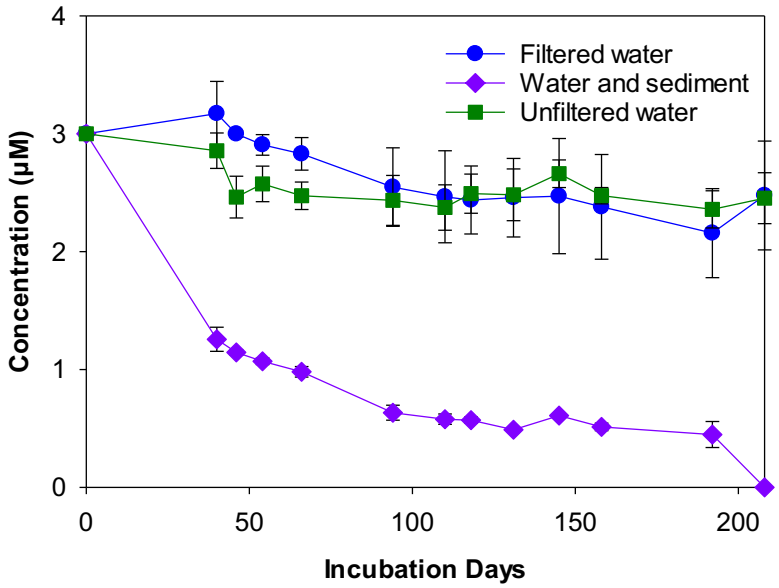

Figure S4. Aqueous fluridone concentrations (initial concentration = 3 µM) in preliminary microcosms incubated with pretreatment environmental inocula from Hooker Lake. Error bars represent the standard deviation of triplicate vessels.

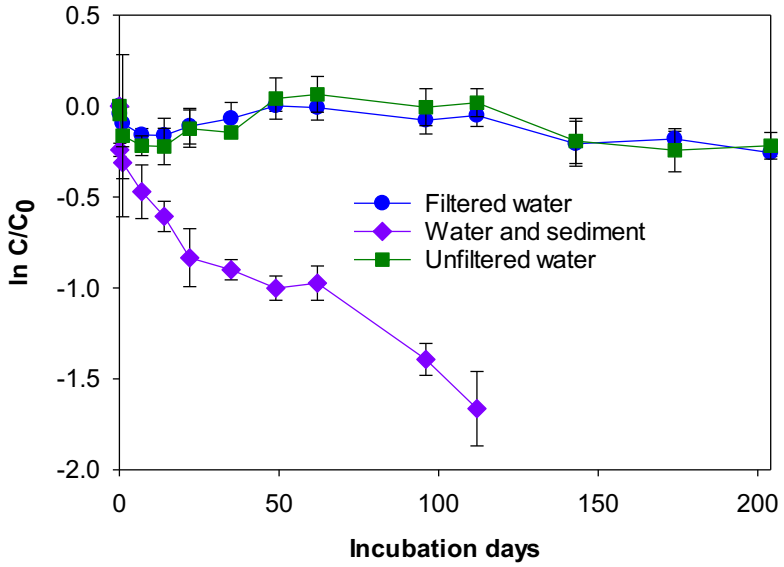

Figure S5. Pseudo-first-order microcosm kinetics for fluridone in the second set of microcosms. Error bars represent the standard deviation of triplicate vessels.

Section S6. Sediment extractions

To determine the validity of the solution used in the sediment extractions, a recovery test was performed (**Figure S6**). Triplicate samples of 100 mg of Ottawa test sand were placed in Falcon tubes. Each sample was spiked with a known amount of fluridone that would result in 1, 5, 10, 15, and 20 µM of fluridone with the extraction solution added. Samples were then introduced to 7 mL of a 50:50 methanol:water extraction solution, shaken in an incubator shaker for two hours, centrifuged, and syringe filtered (0.45 µm) into clean 2 mL glass amber vials for analysis. While fluridone recoveries may have been lower in lake sediment due to the presence of natural organic matter, total fluridone recovery of >90% was observed in initial microcosm timepoints (**Figure 3c** in main manuscript). In addition, it is important to note that the same lake sediment was used for all experiments and field measurements (**Figure S7**) and thus any matrix interferences were the same in all extractions.

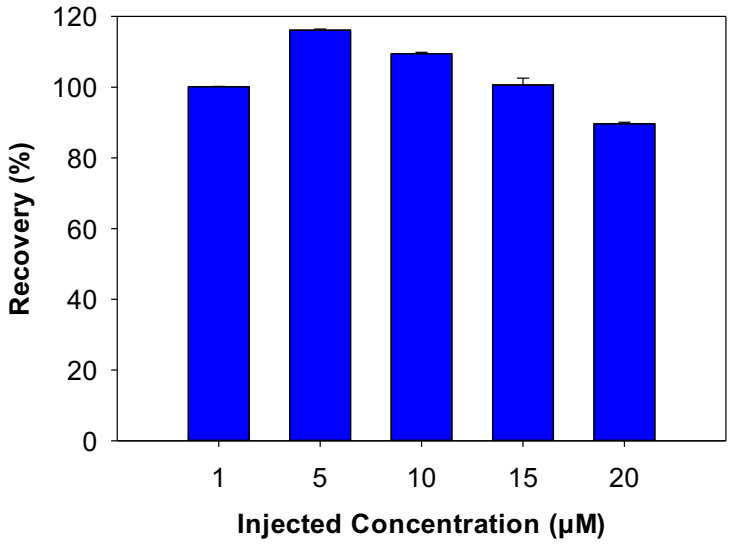

Figure S6. Percent recovery of fluridone from sediment samples using a 50:50 methanol:water extraction solution. Error bars represent the standard deviation of triplicate vessels.

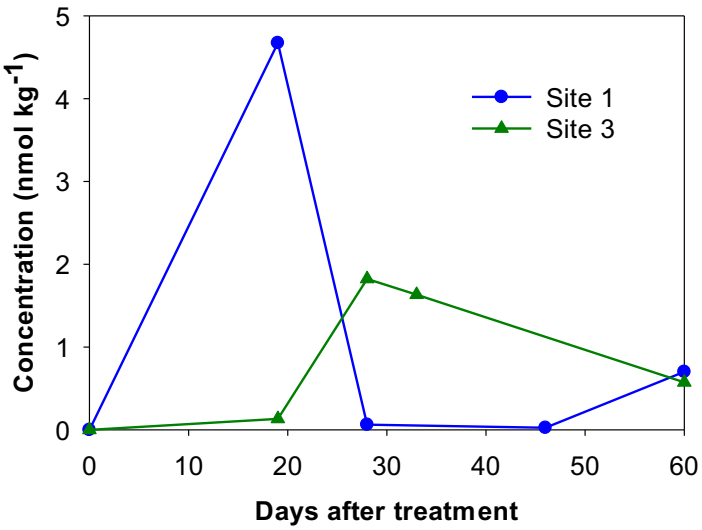

Figure S7. Sediment fluridone concentrations in Hooker Lake during treatment. Concentrations are reported as nmol of fluridone per kg of dried sediment. Note that only one site was sampled on certain days.

Section S7. Analytical methods

Fluridone from laboratory experiments, 3-trifluoromethyl benzoic acid, and 2-nitrobenzaldehyde were analyzed via high performance liquid chromatography (HPLC). All methods used an Agilent Technologies 1260 Infinity instrument equipped with a diode array detector, an Agilent InfinityLab C-18 Poroshell 120 column, an aqueous buffer composed of 10% acetonitrile and 0.1% formic acid in ultrapure water for the aqueous phase (A), and 100% acetonitrile for the mobile phase (B). Fluridone and 3-trifluoromethyl benzoic acid were analyzed using a gradient method (**Table S4**)[17] and 2-nitrobenzyaldehyde was analyzed using an isocratic method (**Table S5**). Fluridone analyzed through HPLC had a limit of detection (LOD) of 0.6 μM.

Table S4. Time segments for gradient method used to analyze fluridone and 3-trifluoromethyl benzoic acid on HPLC.

Time (minutes)	A%	B%
0	60	40
0.8	0	100
1.25	0	100
1.30	60	40
3.5	60	40

Table S5. Instrument parameters for detection of fluridone, 3-trifluoromethyl benzoic acid, and 2-nitrobenzaldehyde in water by HPLC.

Compound	% Aqueous Buffer	Flow (mL min^{-1})	Detection wavelength (nm)	Retention Time (min)	Purpose	LOD (μM)
2-nitrobenzaldehyde	80	0.5	231	2.9	311 nm actinometer	0.5
fluridone	See Table S4	0.8	313	1.8	herbicide	0.6
3-trifluoromethyl benzoic acid	See Table S4	0.8	222	1.1	degradation product	0.01
unknown product	See Table S4	0.8	313	1.04	degradation product	-

Fluridone in the field samples was quantified using an Agilent Triple Quad 6460 liquid chromatograph-tandem mass spectrometer (LC-MS/MS) using positive mode electrospray ionization which had an LOD of 2.6 nM. Fluridone was quantified using a calibration curve generated using an authentic standard of the parent compound. Internal standards or surrogates were not used due to the lack of commercially available labeled fluridone. All field samples were from the same lake and interpreted based on the percent change in fluridone concentration; therefore, any matrix interference from the lake water samples was the same in all cases and does not impact interpretation of the data. Samples were analyzed using a gradient method (**Table S6**) of aqueous buffer (A: 10% acetonitrile and 0.1% formic acid in ultra-pure water) and organic phase of 100% acetonitrile (B) at 0.4 mL min^{-1} on an Agilent InfinityLab C-18 Poroshell 120 column with a column temperature control of 40°C.

LC-MS/MS running conditions and method details for fluridone:

Mode: Positive electrospray ionization	Retention time: 2.8 min
Scan type: MRM	Precursor *m/z*: 330
Gas temp: 300°C	Product *m/z*: 309, 259
Nebulizer pressure: 45 psi	Fragmentor voltage: 312
Sheath gas temperature: 250°C	Collision energy: 41, 56
Sheath gas flow rate: 11 L/min	Cell accelerator voltage: 4
Capillary voltage: 3500 V	LOD: 2.6 nM
Dwell: 80 msec	

Table S6. Time segments for gradient method used to analyze fluridone on LC-MS/MS.

Time (minutes)	A%	B%
0	40	60
0.75	40	60
0.80	0	100
1.15	0	100
1.25	40	60
3.00	40	60

Dissolved organic carbon (DOC) concentrations were measured using a GE Sievers M5310 TOC analyzer. Calibration check solutions were made from analytical grade potassium hydrogen phthalate ranging from 0 - 10 mg-C L^{-1}. Ultraviolet-visible light spectra for each lake were

collected using a Shimadzu 2401PC recording spectrophotometer in 1 nm increments from 200-800 nm.

References

(1) West, S. D.; Day, E. W.; Burger, R. O. Dissipation of the experimental aquatic herbicide fluridone from lakes and ponds. *J. Agric. Food Chem.* **1979**, *27 (5)*, 1067- 1072.

(2) Muir, D.; Grift, N. P.; Blouw, A. P.; Lockhart, W. L. Persistence of fluridone in small ponds. *J. Environ. Qual.* **1980**, *9* (1), 151–156.

(3) West, S. D.; Burger, R. O.; Poole, G. M.; Mowrey, D. H. Bioconcentration and field dissipation of the aquatic herbicide fluridone and its degradation products in aquatic environments. *J. Agric. Food Chem.* **1983**, *31* (3), 579–585.

(4) Arnold, W. R. Fluridone: A new aquatic herbicide. *J. Aquat. Plant Manage*. **1979**, *17*, 30–33.

(5) Muir, D. C. G.; Grift, N. P. Fate of fluridone in sediment and water in laboratory and field experiments. *J. Agric. Food Chem.* **1982**, *30* (2), 238–244.

(6) Onterra. Silver Lake Management Plan https://www.townshipofmarion.com/wp-content/uploads/2018/01/SilverWaushara_HWMmonitorReport2017_v1-1.pdf (accessed May 4, 2023).

(7) Mossler, M. A.; Shilling, D. G.; Haller, W. T. Photolytic degradation of fluridone. *J. Aquat. Plant Manage.* **1989**, *27*, 69–73.

(8) Saunders, D. G.; Mosier, J. W. Photolysis of the aquatic herbicide fluridone in aqueous solution. *J. Agric. Food Chem.* **1983**, *31* (2), 237–241.

(9) Macdonald, G. E.; Haller, W. T.; Shilling, D. G. UV-B filtration to reduce photolysis of fluridone in experimental tanks. *J. Aquat. Plant Manage.* **1996**, *34* (2), 78–80.

(10) Marquis, L. Y.; Comes, R. D.; Yang, C. P. Degradation of fluridone in submersed soils under controlled laboratory conditions. *Pestic. Biochem. Physiol.* **1982**, *17*, 68–75.

(11) Mossler, M. A.; Shilling, D. G.; Albrecht, S. L.; Haller, W. T. Microbial degradation of fluridone. *J. Aquat. Plant Manage.* **1991**, *29*, 77–80.

(12) Fluridone Human Health and Ecological Risk Assessment https://www.dec.ny.gov/docs/lands_forests_pdf/seraflurid.pdf (accessed May 4, 2023).

(13) Helms, J. R.; Stubbins, A.; Ritchie, J. D.; Minor, E. C.; Kieber, D. J.; Mopper, K. Absorption spectral slopes and slope ratios as indicators of molecular weight, source, and photobleaching of chromophoric dissolved organic matter. *Limnol. Oceanogr.* **2009**, *53*, 955-969.

(14) Weishaar, J. L.; Aiken, G. R.; Bergamaschi, B. A.; Fram, M. S.; Fujii, R.; Mopper, K. Evaluation of specific ultraviolet absorbance as an indicator of the chemical composition and reactivity of dissolved organic carbon. *Environ. Sci. Technol.* **2003**, *37* (20), 4702–4708.

(15) A. Leifer, *The Kinetics of Environmental Aquatic Photochemistry: Theory and Practice*, American Chemical Society, Washington, DC, **1988**, 25–28.

(16) Gueymard, C. A. Interdisciplinary applications of a versatile spectral solar irradiance model: A review. *Energy* **2005**, *30* (9), 1551–1576.

(17) Cozzola, A. J.; Dehnert, G. K.; White, A. M.; Karasov, W. H. Effects of subchronic exposure to environmentally relevant concentrations of a commercial fluridone formulation on fathead minnows (Pimephales promelas). *Aquat. Toxicol.* **2022**, *244*, 106098.