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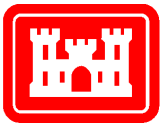

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Aquatic Plant Control Research Program

Whole-Lake Applications of Sonar™ for Selective Control of Eurasian Watermilfoil

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Final report

Prepared for U.S. Army Corps of Engineers
Washington, DC 20314-1000

and Aquatic Ecosystem Restoration Foundation
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Preface

The work reported herein was conducted as part of the Aquatic Plant Control Research Program (APCRP), Work Unit 32437, and the Aquatic Ecosystem Restoration Foundation (AERF). The APCRP is sponsored by Headquarters, U.S. Army Corps of Engineers (HQUSACE), and is assigned to the U.S. Army Engineer Research and Development Center (ERDC) under the purview of the Environmental Laboratory (EL), Vicksburg, MS. Funding was provided under Department of the Army Appropriation No. 96X3122, Construction General. The APCRP is managed under the Center for Aquatic Plant Research and Technology (CAPRT), Dr. John W. Barko, Director. Mr. Robert C. Gunkel, Jr., was Assistant Director for CAPRT. Program monitor during this study as Mr. Timothy R. Toplisek, HQUSACE.

The Principal Investigator of this study was Dr. Kurt D. Getsinger, Ecosystems Processes and Effects Branch (EPEB), Environmental Processes and Effects Division (EPED), EL, ERDC. This study was conducted and the report prepared by Dr. Getsinger, Dr. John D. Madsen, and Mr. R. Michael Stewart, EPEB; Mr. Tyler J. Koschnick. Dr. Michael D. Netherland, and Ms. Alicia G. Staddon, SePRO Corporation, Carmel, IN; Mr. David R. Honnell, University of North Texas; and Ms. Chetta S. Owens, AScI Corporation, Lewisville, TX.

Technical reviews of this report were provided by Mr. John G. Skogerboe, EPEB, and Dr. Steve Cockreham, SePRO. Special thanks is extended to Ms. Diana Klemans, Chief, Inland Lakes and Wetlands Unit (ILWU), Michigan Department of Environmental Quality, and members of the ILWU for allowing this study to occur. For assisting with various technical phases of the work, gratitude is extended to personnel at the Chemical Control and Physiological Processes Team, EPEB; the ERDC Lewisville Aquatic Ecosystem Research Facility, Lewisville, TX; the Michigan Department of Environmental Quality; the Department of Fisheries and Wildlife, Michigan State University; Aquest Corporation, Flint, MI; Progressive AE, Grand Rapids, MI; Professional Lake Management, Culedonia, MI; Environmental Lake Management, White Lake, MI; NDR Research Consultants, Plainfield, IN; and Cygnet Enterprises, Flint, MI. Sincere appreciation also goes to members of the Michigan Aquatic Managers Association, Michigan Sonar Distributors, fluridone applicators, lake managers, and riparians associated with the study lakes.

This investigation was performed under the general supervision of Dr. John W. Keeley, Director, EL; Dr. Richard Price, Chief, EPED; and Dr. Robert Kennedy, Acting Chief, EPEB.

At the time of publication of this report, Director of ERDC was Dr. James R. Houston. Commander was COL James S. Weller, EN.

This report should be cited as follows:

Getsinger, Kurt D., Madsen, John D., Koschnick, Tyler J., Netherland, Michael D., Stewart, R. Michael, Honnell, David R., Staddon, Alicia G., and Owens, Chetta S. (2001). "Whole-lake applications of Sonar™ for selective control of Eurasian watermilfoil," Technical Report ERDC/EL TR-01-7, U.S. Army Engineer Research and Development Center, Vicksburg, MS.

1 Introduction

Background

The aquatic herbicide fluridone {1-methyl-3-phenyl-5-[3-(trifluromethyl)phenyl]-4(1H)-pyridinone} is being used to control the submersed exotic weed Eurasian watermilfoil (*Myriophyllum spicatum* L.) in natural lakes and reservoirs across the northern tier states. Limiting the growth of Eurasian watermilfoil is important because the morphology and physiology of this plant enable it to form large dense stands that out compete most submersed species and displace the native plant community (Grace and Wetzel 1978; Aiken, Newroth, and Wile 1979; Madsen, Eichler, and Boylen 1988; Madsen, Hartleb, and Boylen 1991, Smith and Barko 1990). These weedy infestations also negatively impact fish and wildlife habitat, water quality, and recreational uses of water bodies (Hansen, Oliver, and Otto 1983, Newroth 1985, Ross and Lembi 1985, Nichols and Shaw 1986).

The purpose of many fluridone treatments is to selectively remove Eurasian watermilfoil, while minimizing impacts on the native plant community. Most of these treatments are utilizing the liquid aqueous suspension (AS) of fluridone, registered as Sonar® AS. In these cases, fluridone is being used in a unique manner, in that the entire water body is being managed to selectively remove an exotic pest species, rather than relying on a more traditional approach of spot-treating smaller sections of a lake to reduce the weed infestation.

Although the maximum legal concentration of fluridone in water can be up to 150 $\mu g \cdot L^{-1}$, growth chamber research has indicated that fluridone can render various levels of Eurasian watermilfoil control at initial treatment rates as low as 4 $\mu g \cdot L^{-1}$, provided that an adequate herbicide exposure period (> 60 d) is maintained (Netherland, Getsinger, and Turner 1993; Netherland and Getsinger 1995a,b). These studies have clearly shown that to provide effective control, a target plant must be exposed to some threshold level of fluridone in the initial period of exposure and then be exposed to lower levels of fluridone for an extended time period. Furthermore, results of outdoor mesocosm evaluations on mixed submersed plant communities have suggested that fluridone rates between 5 and 10 $\mu g \cdot L^{-1}$, concomitant with an adequate exposure period (> 60 days) with residues remaining above 2 $\mu g \cdot L^{-1}$, can effectively control Eurasian watermilfoil, while effects on nontarget species, such as elodea (*Elodea canadensis* L.), American pondweed (*Potamogeton nodosus* Poiret), sago

pondweed (*Potamogeton pectinatus* L.), and wild celery (*Vallisneria americana* Michaux) are minimal in the year of treatment (Netherland, Getsinger, and Turner 1997). Finally, results of these small-scale studies revealed two important points: (a) there was a significant difference in the species-selective properties of fluridone between 5 and 10 $\mu g \bullet L^{-1}$; and (b) early-season applications of fluridone provided better control of Eurasian watermilfoil and enhanced selectivity.

There is some debate among the lake management community concerning the selective plant control properties of fluridone when used in whole-lake treatment scenarios (Kenaga 1993, 1995). Although cover and diversity of native species has usually recovered by 1 to 3 years posttreatment following a whole-lake fluridone application, even at rates > 20 $\mu g \bullet L^{-1}$ (Getsinger 1993; Smith and Pullman 1997), much of the concern has focused on potential impacts to fish populations and overall lake ecology following the removal of a portion of vegetation throughout the lake in the year of treatment. Field observations and reports indicate that when fluridone is applied at water concentrations > 10 $\mu g \bullet L^{-1}$, some nontarget plant species may survive the year of treatment, while others do not (Kenaga 1993, 1995; Welling, Crowell, and Perleberg 1997; Smith and Pullman 1997). Uncertainties, however, in the aqueous fluridone concentrations achieved and maintained in these situations, have left the issue of defining optimal treatment rates for selective plant control unresolved. In addition, methods used in these studies to determine selectivity were subjective and data were not subjected to statistical analysis.

Objectives

Since reliable quantitative information linking changes in submersed plant species diversity with fluridone treatments is limited, particularly with respect to water residue records, a study was conducted in which prescription low-dose fluridone treatments were applied to selected lakes in Michigan. The primary objective of this study was to determine whether submersed plant diversity and frequency are impacted by whole-lake, low-dose fluridone applications in the year of treatment when targeting for control of Eurasian watermilfoil. Secondary objectives included: (a) determining herbicide effects on the exotic weed curly-leaf pondweed (*Potamogeton crispus* L.); (b) evaluating shifts in submersed plant species diversity at 1-year posttreatment; (c) measuring the effect of thermal stratification on water column distribution of fluridone; (d) verifying laboratory results of fluridone concentration and exposure time relationships with respect to efficacy; and (e) correlating a new immunoassay fluridone water residue technique with the conventional high-performance liquid chromatography method. A companion study, results of which are not reported here, was also conducted to determine indirect impacts of the fluridone treatments on the aquatic invertebrate and fish populations.

2 Materials and Methods

Study Sites

Eight lakes, approximately 55 to 220 ha in size and located in the eastern and western portions of southernl Michigan, were selected for the study (Figure 1). County location, surface area, depth, and littoral zone information for each lake are presented in Table 1. These lakes represented typical water bodies in the southern region of the state managed for the control of Eurasian watermilfoil and curlyleaf pondweed using herbicides. Although all of these lakes were infested with Eurasian watermilfoil, and most with curlyleaf pondweed, they also contained a total of 23 species (average per lake = seven species) of nontarget native submersed plants at the initiation of the study (Tables 2 through 5). Four of the lakes, Lobdell (221 ha), Wolverine (98 ha), Big Crooked (65 ha), and Camp (55 ha), were chosen for fluridone treatments, and an equal number, Bass (75 ha), Big Seven (68 ha), Clear (75 ha), and Heron (53 ha), were used as untreated reference lakes. The four fluridone-treated lakes were chosen from a pool of lakes that qualified under the Michigan Department of Environmental Quality's (MDEQ) permit procedures to apply Sonar® on a whole-lake basis in 1997. The untreated reference lakes were selected from a pool of lakes that would not experience major aquatic plant management activities in 1997 or 1998.

Fluridone Treatments

Using results from laboratory and mesocosm studies (Netherland, Getsinger, and Turner 1993 and 1997; Netherland and Getsinger 1995a, b), previous field experience with fluridone in Michigan lakes, and certain conditions required by MDEQ permit procedures for whole-lake Sonar applications, a prescription low-dose whole-lake fluridone treatment strategy was developed which was intended to provide control of Eurasian watermilfoil while minimizing injury to nontarget plant populations during the year of treatment. This prescription treatment was utilized on each lake, employing an initial application strategy designed to evenly distribute fluridone at a concentration of 5 $\mu g \bullet L^{-1}$ within the top 3.05 m (10 ft) of the water column over the entire lake. This initial application was followed in 2 to 3 weeks by a second, booster application, designed to reestablish a whole-lake fluridone concentration of 5 $\mu g \bullet L^{-1}$. The purpose of this initial and booster application strategy was two fold: (a) to provide maximum selectivity while controlling Eurasian watermilfoil, and (b) to compensate for low initial water

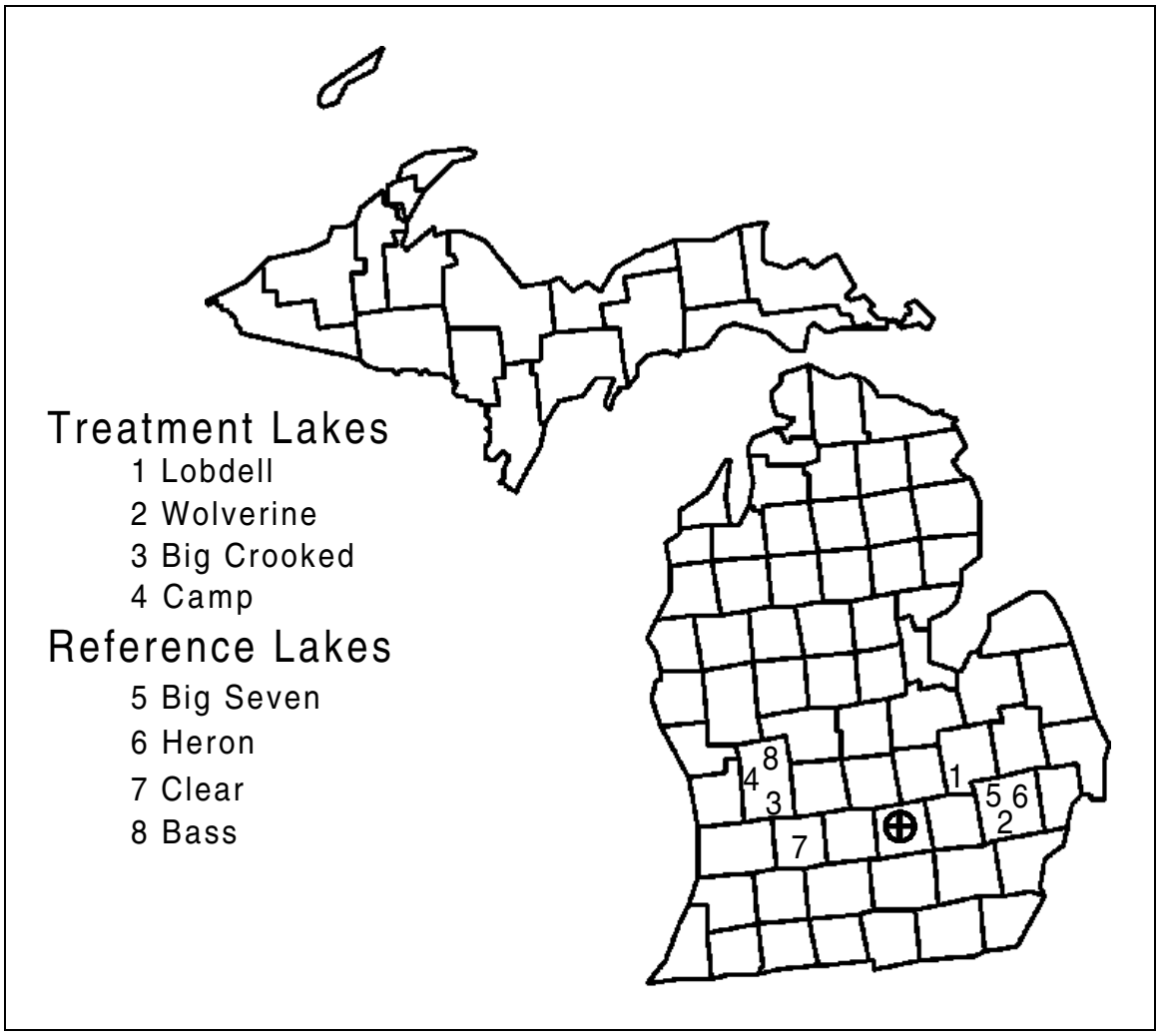

Figure 1. Lakes selected for whole-lake fluridone evaluations in Michigan, 1997-1998

Table 1
Location, Morphometry, and Extent of Littoral Zone for the Eight Study Lakes in Michigan, 1997-1998

Lake	County	Surface Area, ha	Mean Depth m	Max Depth m	Max Plant Depth,[1] m
Flurodone Treated					
Big Crooked	Kent	65	4.4	18.5	5.5
Camp	Kent	65	7.5	16.7	6.1
Lodell	Genessee/Livingston	221	3.2	24.4	5.8
Wolverine	Oakland	98	2.9	17.9	3.6
Untreated Reference					
Bass	Kent	75	3.0	10.4	7.6
Big Seven	Oakland	68	2.9	16.7	5.5
Clear	Barry	75	2.3	7.3	6.1
Heron	Oakland	53	3.5	20.1	6.4

[1] Estimate of littoral zone.

Table 2
Percent Frequency of Occurrence of Submersed Plants in Four Fluridone-Treated Lakes in Michigan, May and August 1997

Species	Pretreatreatment, May 1997				Posttreatment, August 1997			
	BIG	CAM	LOB	WOL	BIG	CAM	LOB	WOL
Cabomba caroliniana	0	0	0	0	0	0	0	0
Ceratophyllum demersum	15	4	3	0	43	4	5	8
Chara spp.	19	14	34	53	40	66	54	77
Drepanocladus spp.	0	0	0	0	<1	0	0	0
Elodea canadensis	1	32	0	0	<1	2	2	<1
Heteranthera dubia	0	0	0	0	20	71	0	5
Myriophyllum spicatum	39	74	38	46	0	4	3	35
Myriophyllum sibiricum	0	0	1	0	0	0	0	0
Myriophyllum verticillatum	0	0	1	0	0	0	0	0
Najas spp.	0	0	0	0	0	0	0	0
Najas flexilis	0	0	0	0	0	0	1	0
Najas gracillima	0	0	0	0	0	0	0	0
Najas guadalupensis	0	0	<1	0	<1	0	0	<1
Najas marina	0	0	0	0	0	0	0	0
Najas minor	0	0	0	0	0	0	0	0
Nitella spp.	0	0	0	0	0	0	0	0
Potamogeton amplifolius	35	0	8	20	34	0	19	12
Potamogeton diversifolius	0	0	0	0	0	0	0	0
Potamogeton crispus	24	68	14	17	1	33	0	0
Potamogeton foliosus	0	0	0	0	0	0	0	0
Potamogeton gramineus	0	0	0	1	0	0	0	4
Potamogeton illinoensis	0	0	<1	0	15	0	26	0
Potamogeton natans	0	0	1	0	1	0	1	0
Potamogeton nodosus	0	0	2	0	0	0	<1	0
Potamogeton pectinatus	2	0	<1	4	4	15	16	37
Potamogeton praelongus	41	11	0	1	3	28	0	0
Potamogeton pusillus	0	0	0	0	0	0	0	0
Potamogeton richardsonii	0	0	0	0	2	4	<1	0
Potamogeton robbinsii	2	0	0	0	21	0	0	0
Potamogeton zosteriformis	25	0	1	0	44	0	22	7
Potamogeton spp.	0	0	0	0	0	1	0	0
Ranunculus spp.	0	0	0	0	0	0	<1	0
Sagittaria spp.	0	0	0	0	2	0	<1	0
Sagittaria graminea	0	0	0	0	0	0	0	0
Utricularia intermedia	0	0	0	0	0	0	0	0
Utricularia minor	0	0	0	0	0	0	0	4
Utricularia purpurea	0	0	0	0	0	0	0	0
Utricularia vulgaris	0	0	1	0	<1	0	7	15
Utricularia spp.	0	0	0	0	0	0	0	0
Vallisneria americana	0	0	0	0	4	47	58	1
Zannichellia spp.	0	0	0	0	0	0	0	0

Note: BIG = Big Crooked; CAM = Camp; LOB = Lobdell; WOL = Wolverine.

residues, while extending the overall fluridone exposure period in the lakes for > 60 d.

Observations from previous fluridone treatments in Michigan indicated that in many cases plants growing in depths > 3.05 m were not being affected by the application, even though the volume of the entire lake was used to calculate the treatment rate. Therefore, it was suspected that the establishment of a thermocline prior to application was restricting vertical mixing and dilution of the herbicide. This limited mixing could isolate and concentrate fluridone in that part of the water column located above the thermocline, resulting in the risk of increased injury to nontarget plants growing in the relatively shallow littoral

Table 3
Percent Frequency of Occurrence of Submersed Plants in Four Fluridone-Treated Lakes in Michigan, May and August 1998

Species	Pretreatreatment, May 1998				Posttreatment, August 1998			
	BIG	CAM	LOB	WOL	BIG	CAM	LOB	WOL
Cabomba caroliniana	0	0	0	0	0	0	0	0
Ceratophyllum demersum	14	0	1	3	24	12	13	1
Chara spp.	29	79	57	63	24	92	48	81
Drepanocladus spp.	0	0	0	0	0	0	0	1
Elodea canadensis	0	14	<1	3	1	9	1	<1
Heteranthera dubia	1	18	<1	<1	41	38	1	1
Myriophyllum spicatum	0	12	13	71	7	14	10	54
Myriophyllum sibiricum	<1	0	0	0	2	0	0	0
Myriophyllum verticillatum	0	0	0	0	0	0	7	0
Najas spp.	0	0	0	0	0	0	0	0
Najas flexilis	0	0	0	0	0	2	16	<1
Najas gracillima	0	0	0	0	0	0	8	25
Najas guadalupensis	0	0	0	0	33	0	0	4
Najas marina	0	0	0	0	0	0	1	0
Najas minor	0	0	0	0	0	0	0	3
Nitella spp.	0	0	0	0	0	0	0	0
Potamogeton amplifolius	49	2	25	28	41	8	7	20
Potamogeton diversifolius	0	0	0	0	0	0	0	0
Potamogeton crispus	53	86	49	36	24	34	4	1
Potamogeton foliosus	0	0	0	0	0	0	0	20
Potamogeton gramineus	<1	0	0	9	<1	0	4	0
Potamogeton illinoensis	0	1	21	3	0	0	5	12
Potamogeton natans	0	0	<1	0	0	0	0	0
Potamogeton nodosus	0	0	0	<1	0	1	0	0
Potamogeton pectinatus	2	4	14	36	<1	1	6	0
Potamogeton praelongus	29	13	0	0	23	20	0	0
Potamogeton pusillus	0	0	0	3	0	0	0	0
Potamogeton richardsonii	<1	2	0	0	0	0	0	0
Potamogeton robbinsii	14	0	0	0	11	0	0	0
Potamogeton zosteriformis	51	0	34	12	41	0	5	7
Potamogeton spp.	0	0	0	0	0	0	0	0
Ranunculus spp.	<1	13	2	0	0	1	0	0
Sagittaria spp.	<1	0	0	0	0	1	0	0
Sagittaria graminea	0	0	0	0	0	0	0	0
Utricularia intermedia	0	0	0	0	0	0	0	0
Utricularia minor	0	0	0	<1	0	0	<1	18
Utricularia purpurea	0	0	0	0	0	0	0	0
Utricularia vulgaris	<1	0	10	0	<1	0	24	14
Utricularia spp.	0	0	0	0	0	0	0	0
Vallisneria americana	0	0	6	0	3	44	39	1
Zannichellia spp.	0	1	0	<1	0	0	0	0

Note: BIG = Big Crooked; CAM = Camp; LOB = Lobdell; WOL = Wolverine.

zone. Thus, the water volume defined by the 3.05-m depth contour is the maximum volume allowed by MDEQ to be treated with fluridone in a whole-lake application scenario, and this application restriction was incorporated into the design of this study. The MDEQ's current fluridone application policy (and employment of the 3.05-m lake volume restriction) is intended to prevent excessive control of beneficial native plants (particularly in the year of treatment) and is based upon a combination of factors including: (a) the major portion of the littoral zone in central Michigan lakes supporting abundant levels of submersed plants generally occurs within waters < 3.05 m in depth; and (b) thermal stratification of these lakes is expected to occur at the time of the initial herbicide applications restricting vertical distribution.

Table 4
Percent Frequency of Occurrence of Submersed Plants in Four Untreated Lakes in Michigan, May and August 1997

Species	Pretreatreatment, May 1997				Posttreatment, August 1997			
	BAS	BIS	CLE	HER	BAS	BIS	CLE	HER
Cabomba caroliniana	0	0	2	0	0	0	14	0
Ceratophyllum demersum	0	5	0	7	1	46	2	22
Chara spp.	48	2	0	28	60	4	3	15
Drepanocladus spp.	0	0	<1	0	0	0	0	0
Elodea canadensis	<1	30	0	10	1	52	0	25
Heteranthera dubia	0	5	0	19	<1	<1	0	20
Myriophyllum spicatum	31	43	67	35	35	74	70	42
Myriophyllum sibiricum	0	0	0	0	17	<1	20	15
Myriophyllum verticillatum	0	0	0	0	0	0	0	0
Najas spp.	0	0	0	0	0	0	6	0
Najas flexilis	5	0	0	0	5	1	0	8
Najas gracillima	0	0	0	0	0	0	0	0
Najas guadalupensis	2	<1	0	0	9	0	2	3
Najas marina	0	0	0	0	0	0	0	0
Najas minor	0	0	0	0	0	0	0	0
Nitella spp.	0	0	0	0	0	0	0	0
Potamogeton amplifolius	<1	5	33	0	<1	9	44	0
Potamogeton diversifolius	0	0	0	0	0	7	0	0
Potamogeton crispus	0	49	2	21	<1	1	0	0
Potamogeton foliosus	0	0	0	0	<1	0	2	0
Potamogeton gramineus	<1	0	3	0	24	0	47	13
Potamogeton illinoensis	18	0	0	0	0	0	44	0
Potamogeton natans	0	0	0	0	2	12	0	9
Potamogeton nodosus	0	0	0	0	0	<1	0	0
Potamogeton pectinatus	0	0	0	0	6	10	0	18
Potamogeton praelongus	0	0	0	0	13	0	0	0
Potamogeton pusillus	0	0	0	0	0	<1	7	0
Potamogeton richardsonii	0	0	0	0	0	0	0	0
Potamogeton robbinsii	0	0	24	0	0	0	33	0
Potamogeton zosteriformis	2	<1	0	1	6	12	4	27
Potamogeton spp.	0	0	0	0	1	0	0	0
Ranunculus spp.	0	0	0	0	<1	0	<1	11
Sagittaria spp.	0	0	0	0	0	0	0	0
Sagittaria graminea	0	0	<1	0	0	0	6	0
Utricularia intermedia	0	0	0	0	0	0	0	0
Utricularia minor	<1	0	0	0	0	0	8	0
Utricularia purpurea	0	0	0	0	0	0	0	0
Utricularia vulgaris	<1	0	7	0	2	2	7	7
Utricularia spp.	0	0	0	0	0	0	0	0
Vallisneria americana	0	0	0	0	4	0	2	4
Zannichellia spp.	0	0	0	0	0	0	0	0

Note: BAS = Bass; BIS = Big Seven; CLE = Clear; HER = Heron.

The bathymetric maps used to determine the 3.05-m depth contours in the treated lakes were provided by the management companies responsible for weed control operations on the each lake. These maps were published prior to 1960 and contours may have changed over time as a result of sedimentation processes in the lakes, which could affect the accuracy of the 3.05-m depth, whole-lake volume calculations. Lake volume estimates were determined by the respective management companies contracted by the respective lake associations to conduct the fluridone treatments (Big Crooked and Camp by ProgressiveAE, Grand Rapids, MI, and Lobdell and Wolverine by Aquest Corporation, Flint, MI) and were made using MDEQ methods standardized for calculating such volumes

Table 5
Percent Frequency of Occurrence of Submersed Plants in Four Untreated Lakes in Michigan, May and August 1998

Species	Pretreatreatment, May 1998				Posttreatment, August 1998			
	BAS	BIS	CLE	HER	BAS	BIS	CLE	HER
Cabomba caroliniana	0	0	9	0	0	0	18	0
Ceratophyllum demersum	0	1	2	19	0	49	2	27
Chara spp.	58	2	1	33	49	0	3	44
Drepanocladus spp.	0	0	2	0	0	0	0	0
Elodea canadensis	<1	71	0	22	0	59	<1	15
Heteranthera dubia	0	0	0	1	<1	0	<1	5
Myriophyllum spicatum	34	74	69	56	44	60	66	41
Myriophyllum sibiricum	0	0	0	0	0	0	0	13
Myriophyllum verticillatum	0	0	0	0	0	0	0	0
Najas spp.	0	0	0	0	0	0	0	0
Najas flexilis	6	0	0	0	6	<1	2	0
Najas gracillima	0	0	0	0	0	0	6	0
Najas guadalupensis	0	0	0	0	9	2	2	4
Najas marina	0	0	0	0	0	0	0	0
Najas minor	0	0	0	0	0	0	0	0
Nitella spp.	2	0	<1	<1	0	0	<1	1
Potamogeton amplifolius	<1	4	25	0	<1	12	23	0
Potamogeton diversifolius	0	<1	0	0	0	5	0	0
Potamogeton crispus	0	59	0	33	0	0	0	<1
Potamogeton foliosus	10	0	13	0	0	0	7	2
Potamogeton gramineus	17	0	37	13	12	5	38	4
Potamogeton illinoensis	<1	8	0	0	0	4	31	8
Potamogeton natans	0	5	0	2	1	5	<1	4
Potamogeton nodosus	0	5	0	0	0	4	0	0
Potamogeton pectinatus	3	8	0	44	1	16	0	17
Potamogeton praelongus	23	0	0	0	11	0	0	0
Potamogeton pusillus	0	<1	<1	0	1	1	2	0
Potamogeton richardsonii	0	0	0	0	<1	0	0	0
Potamogeton robbinsii	1	0	28	0	0	0	23	0
Potamogeton zosteriformis	3	28	2	46	3	20	6	31
Potamogeton spp.	<1	0	0	0	0	0	0	0
Ranunculus spp.	0	0	3	16	0	0	0	0
Sagittaria spp.	0	0	0	0	<1	0	0	0
Sagittaria graminea	0	0	0	0	0	0	5	0
Utricularia intermedia	0	0	0	0	0	0	4	0
Utricularia minor	0	0	1	1	0	0	2	0
Utricularia purpurea	0	0	0	0	0	0	4	0
Utricularia vulgaris	1	2	11	0	1	4	6	0
Utricularia spp.	0	0	0	0	0	0	30	0
Vallisneria americana	1	0	11	4	4	0	<1	9
Zannichellia spp.	0	0	0	0	0	0	0	0

Note: BAS = Bass; BIS = Big Seven; CLE = Clear; HER = Heron.

(Appendix A). The estimated water volumes treated, as calculated using the published 3.05-m depth contours, ranged from 35 to 73 percent of the total volume of the lakes (Table 6).

Treatments were conducted from boats using various conventional liquid herbicide application equipment designed to deliver the fluridone as Sonar® AS at, or slightly below, the water surface. Spray boats were piloted across each lake in a manner to ensure even distribution of the herbicide throughout the lakes. Even distribution of fluridone was important to avoid residue "hot-spots" in the water column ensuring that nontarget plants did not receive a high initial dose,

Table 6
Pretreatment and Prebooster Thermoclines, Percent Lake Volumes by Depth Zone, and Percent Loss of Fluridone-Water Residues at 10/11 DAIT and DABT in Four Michigan Lakes, May 1997

Lake	Depth Zone Volume 0-3.05 m[1]	Depth Zone Volume 3.05-6.1 m	Depth Zone Volume 6.1-9.15 m	Pretreat Thermocline Depth	Nominal Initial Rate $\mu g \cdot L^{-1}$	Fluridone 1 DAIT $\mu g \cdot L^{-1}$	Variance from Nominal	Fluridone 10/11 DAIT $\mu g \cdot L^{-1}$	Variance from 1 DAIT
Big Crooked	58 %	29 %	10 %	8 m	5	3.8	-24 %	3.1	-18 %
Camp	35 %	25 %	20 %	8 m	5	4.2	-16 %	2.6	-38 %
Lobdell	73 %	13 %	5 %	7 m	5	5.5	+10 %	3.4	-38 %
Wolverine	54 %	22 %	13 %	6 m	5	3.4	-33 %	2.6	-24 %
Lake	**Depth Zone Volume 0-3.05 m[1]**	**Depth Zone Volume 3.05-6.1 m**	**Depth Zone Volume 6.1-9.15 m**	**Preboost Thermocline Depth**	**Nominal Booster Rate $\mu g \cdot L^{-1}$**	**Fluridone 1 DABT $\mu g \cdot L^{-1}$**	**Variance from Nominal**	**Fluridone 10/11 DABT $\mu g \cdot L^{-1}$**	**Variance from 1 DABT**
Big Crooked	58 %	29 %	10 %	2 m	5	5.0	0 %	4.5	-10 %
Camp	35 %	25 %	20 %	3 m	5	4.8	-4 %	3.9	-19 %
Lobdell	73 %	13 %	5 %	8 m	5	4.9	-2 %	5.0	+2 %
Wolverine	54 %	22 %	13 %	2 m	5	3.3	-34 %	3.2	-3 %

Note: DAIT = days after initial treatment; DABT = days after booster treatment.
[1] Treatment depth zone.

and to allow for MDEQ compliance water residue sampling at 24-hr posttreatment.

Initial fluridone applications were conducted in 1997 on 12 May (Lobdell by Aquatic Services, Inc., Goodrich, MI, and Wolverine by Environmental Lake Management, Inc., White Lake, MI), and on 14 May (Big Crooked and Camp by Professional Lake Management, Caledonia, MI). Michigan was experiencing a cooler than normal spring, and surface water temperatures of the lakes at 3 to 4 days pretreatment ranged from 10.8 to 12.4 °C. In spite of a late spring, Eurasian watermilfoil and curlyleaf pondweed were actively growing, with shoots extending 50 cm or more above the bottom. Because of the prolonged cool water temperatures, the native submersed plant communities were slightly behind their normal growth cycle pattern in some of the lakes. Applications were prescribed for mid-May to expose Eurasian watermilfoil and curlyleaf pondweed to the herbicide during periods of their most active growth. When using low doses of fluridone to selectively control Eurasian watermilfoil, it is important to treat the plant during early growth, since it is more difficult to control mature plants with low herbicide rates.

In each case, the initial treatment was followed by a second whole-lake application, conducted in an identical method as the first treatment. Sequential (booster) applications of fluridone were conducted in 1997 on 30 May (Wolverine, Big Crooked, and Camp) and on 2 June (Lobdell). These booster applications occurred 16 to 21 days after the first treatment, and were designed to reestablish the aqueous concentration of fluridone in the top 3.05 m of the water column to the 5 $\mu g \cdot L^{-1}$ level in each lake, and to maintain aqueous fluridone levels (~ 2 $\mu g \cdot L^{-1}$) for the exposure time required to control Eurasian watermifoil (> 60 days), but not injure nontarget plants. This retreatment lag time was deemed an acceptable period when considering the concentration/ exposure time requirements for fluridone efficacy and the aqueous dissipation characteristics of the product when applied in a whole-lake method. To

accurately determine the amount of fluridone to apply at the booster treatment, water samples were collected from selected locations (n=6) around each lake (Figures 2 through 5) at 10/11 days after initial treatment (DAIT) and analyzed for herbicide residues using a newly developed enzyme-linked immunosorbent assay (ELISA) technique, known as FasTEST. The mean of these six residue values were used to calculate the booster addition. Specific treatments dates and initial and booster rates are provided for each lake in Table 7.

Table 7
Initial and Booster Treatment Dates and Nominal and Actual Posttreatment Fluridone Rates ($\mu g \cdot L^{-1}$) on Michigan Lakes Treated in May and June 1997

Lake	Treatment Date	Nominal Rate[1]	Actual Rate Posttreatment[2]	Booster Date	Nominal Booster Rate[1]	Actual Rate Post Boost[2]
			1 DAIT			1 DABT
Big Crooked	5/14	5.0	3.78±0.7	5/30	1.97	5.05±0.5
Camp	5/14	5.0	4.20±1.5	5/30	2.48	4.48±0.2
			1 DAIT			1 DABT
Lobdell	5/12	5.0	5.55±2.0	6.2	2.37	4.87±0.8
Wolverine	5/12	5.0	3.35±0.3	5/30	2.42	3.29±0.2

[1] Nominal fluridone applied based on calculated volume of lake within the 3.05-m (10-ft) depth contour using previously published bathymetric maps.
[2] Fluridone residues as measured by FasTEST. Data represent mean (± 1 SE) of water samples collected posttreatment at 30-cm depth at six shallow water stations (n=6), with exception of Lobdell (n=4).

Water Residue Sampling and Analyses

To better correlate fluridone efficacy and selectivity with nominal application rates, an appropriate record of water residues is required. Previous selectivity evaluations have been hindered by a lack of rigorous water sampling protocol, and therefore these evaluations have often relied upon theoretical application rates. Field data have suggested that there can be a variation in the theoretical rate versus actual residues. Therefore, an intensive water residue sampling regime was employed in the fluridone-treated lakes in this study. In each treated lake, six water residue sampling locations (littoral zone stations) were established at regular intervals around the shoreline in a water depth of approximately 2 m, and two sampling locations (deep-zone stations) were established in deep-water regions (Figures 2 through 5). The littoral stations were positioned to provide balanced coverage of lake-wide residues, while the deep stations were designed to allow for monitoring residues above and below established thermoclines. Sampling stations were permanently fixed through the use of a global positioning system (GPS) unit and marked with anchored buoys for the duration of the study.

Water sampling regimes covered a time line of pretreatment up to 81 DAIT. Details of sampling events and collection depths for the littoral and deep stations during the initial and booster treatments are provided in Table 8. Samples from all lakes were collected from pretreatment through 10 and 11 DAIT by U.S. Army Engineer Research and Development Center (USAERDC) personnel. Remaining samples were collected on Big Crooked and Camp by ProgressiveAE personnel and on Lobdell and Wolverine by Aquest Corporation personnel. At each station, duplicate sample sets were collected at each sampling event using a

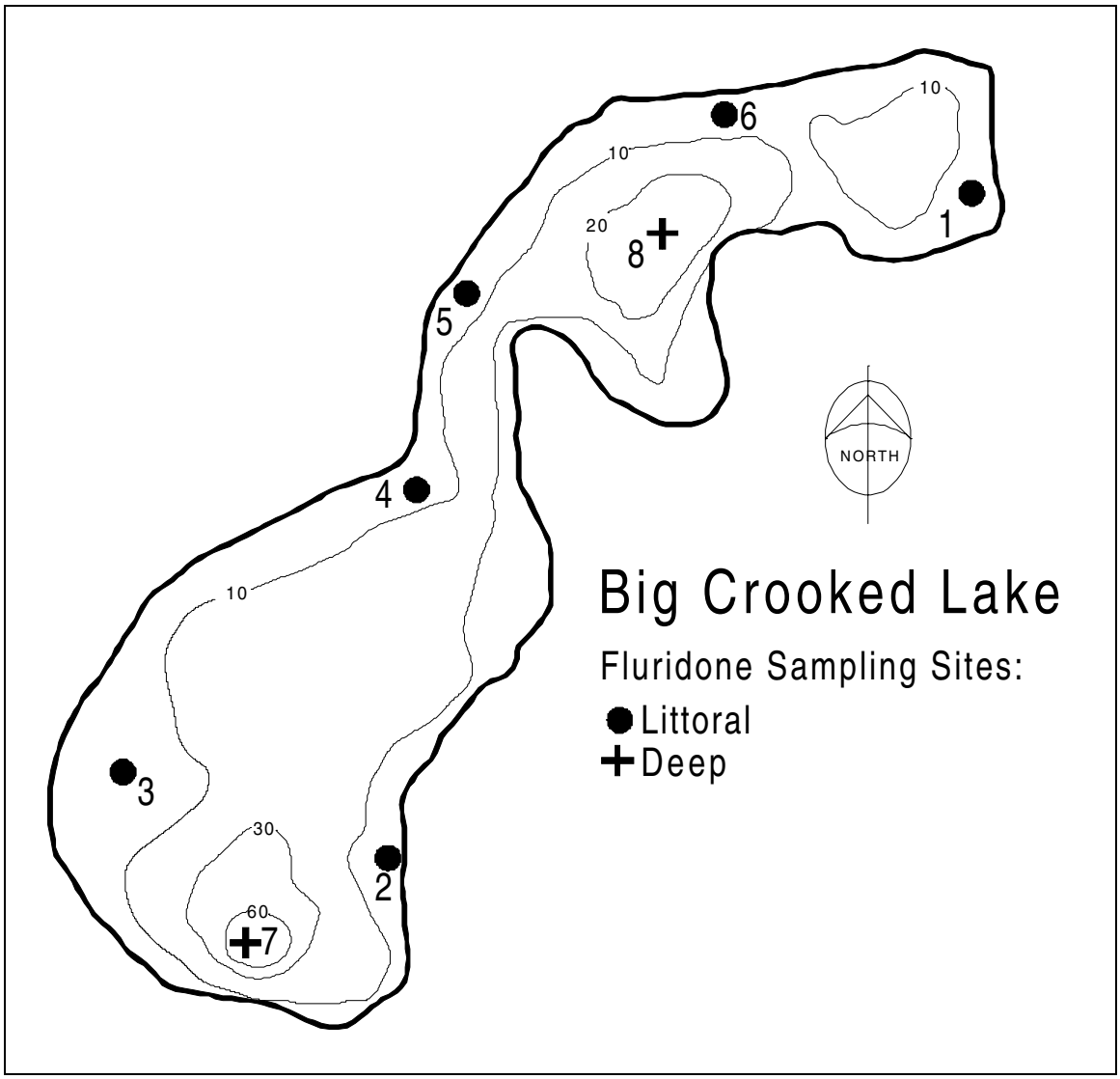

Figure 2. Fluridone water residue sampling sites in Big Crooked Lake, Kent County, Michigan, May through August 1997. Depth contours are in feet

Van Dorn water sampler. Immediately after collection, samples were transferred into amber high-density polyethylene plastic bottles and placed in an ice chest: one set to be analyzed via the ELISA and one via high performance liquid chromatography (HPLC). Samples to be analyzed by the ELISA technique were collected in 250-mL bottles (duplicates for each event), while those to be analyzed by HPLC were collected in 500-mL bottles (duplicates for each event). All samples were kept chilled, in the dark, and shipped overnight to the respective analytical laboratories where samples were stored frozen until analyzed by HPLC and stored chilled until analyzed by ELISA (<48 hr from receipt of samples). Any samples indicating questionable residue levels were reanalyzed to verify residue accuracy.

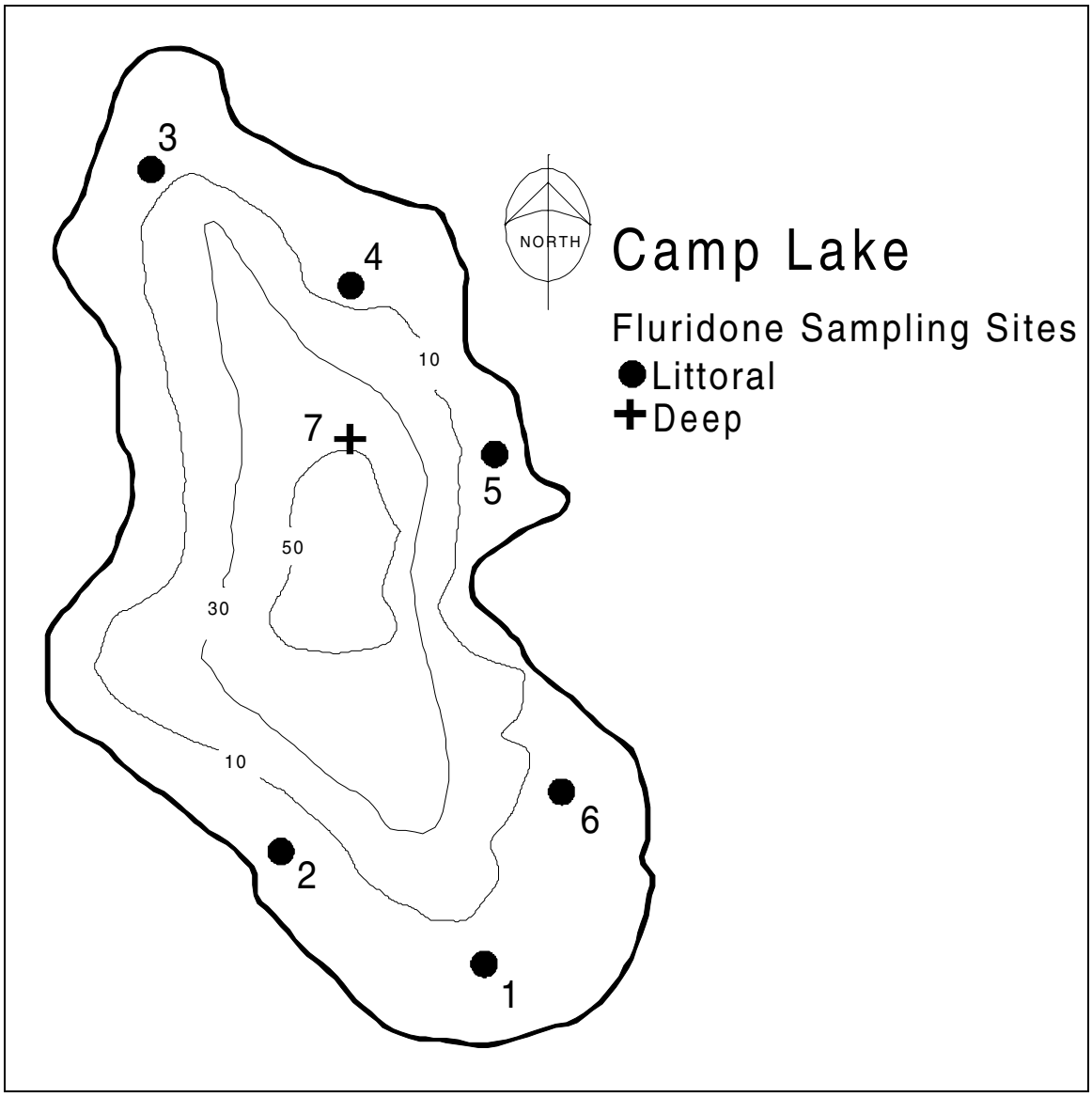

Figure 3. Fluridone water residue sampling sites in Camp Lake, Kent County, Michigan, May through August 1997. Depth contours are in feet

To compare and correlate the two analytical techniques, one complete set of water samples, comprising all samples from all lakes and all samples from individual lakes, was analyzed using both ELISA and HPLC methods. The ELISA analyses were performed by the analytical laboratory group at SePRO Corp., Carmel, IN, and the HPLC analyses were performed by the water chemistry laboratory group at the USAERDC Lewisville Aquatic Ecosystem Research Facility (LAERF), Lewisville, TX.

All HPLC procedures were conducted using a Water's HPLC system (Water's 510 pump, Water's 486 UV detector, Water's 746 data integrator, and Water's μ Bondapak C18, 125A, 10 μm, 3.9 $\times$ 300 mm HPLC column). The method employed was a modification of well-characterized techniques for

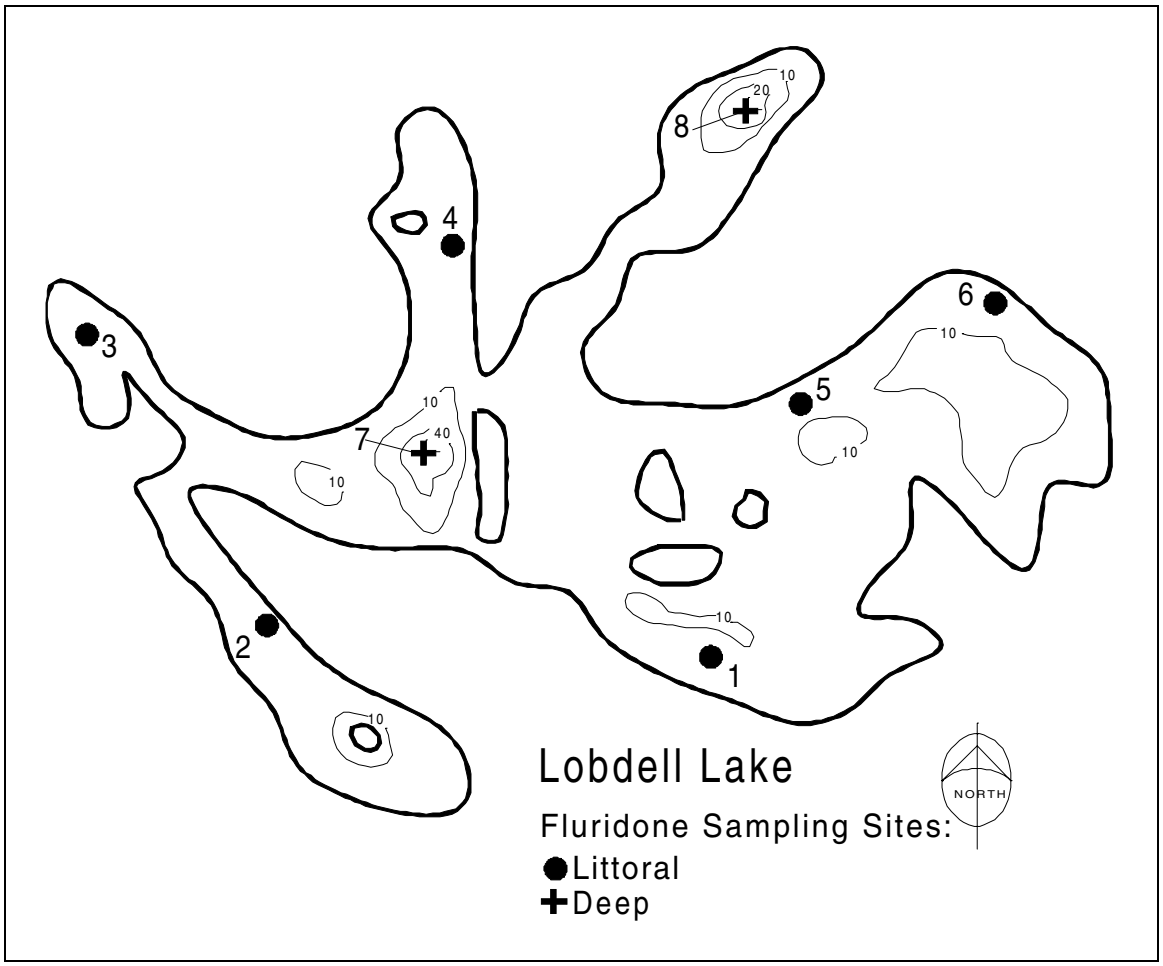

Figure 4. Fluridone water residue sampling sites in Lobdell Lake, Genessee, and Livingston Counties, Michigan, May through August 1997. Depth contours are in feet

measuring fluridone concentrations in water (West and Day 1981; Fox, Haller, and Shelling 1991). This method was modified with the use of solid phase extraction cartridges (SPE) as a pretreatment for the cleaning of the water samples and concentrating fluridone. Water's Sep-pak vac 6-cc (500-mg) C18 cartridges were placed on a 12-place Sep-Pak vacuum mainfold (JT Baker 7018-00) and a 100-mL sample volume was filtered through the SPE cartirdge. All samples were filtered though the SPE cartridges with a final elution to 2 mL with methanol. Samples were collected in a 4-mL amber glass vial and held until injection into the HPLC instrument.

Fluridone quantification in the water was determined by comparison of the detector reponse for the samples against the response obtained from direct injection of known standard concentrations of fluridone. Standards were made from analytical grade fluridone (99.1-percent purity) obtained from the SePRO Corporation. The HPLC conditions were set as follows: Eluent 65:35 methanol: water, chart speed 0.25 cm • min $^{-1}$, wavelength 313 nm, attenuation 8, flow rate

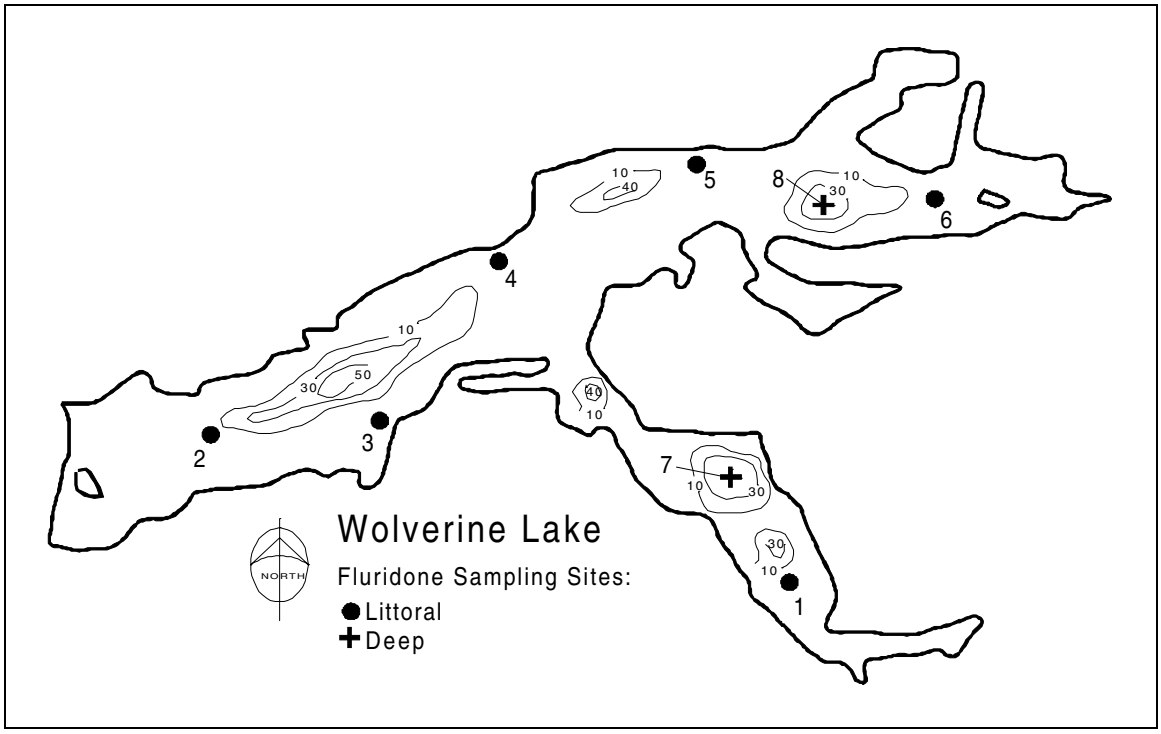

Figure 5. Fluridone water residue sampling sites in Wolverine Lake, Oakland County, Michigan, May through August 1997. Depth contours are in feet

Table 8
Water Residue Sampling Protocol for Michigan Lakes Treated with Low Doses of Fluridone, May through August, 1997

Sampling Zone	Stations	Depth[1]	Initial Treatment	Initial Sampling Event	Booster Treatment	Booster Sampling Event
Littoral	6	MID	Mid-May	Pretreat, 1, 4, 10 DAIT	Late May	1, 10, 20, 30, 60
DABT						
Deep	2	SURF		Pretreat, 4, 10 DAIT		10, 30
DABT		+T T -T BOT				

[1] Littoral zone stations collected at middepth of measured water column (~ 1 m); deep zone stations collected on depth profile to bracket thermocline where SUR = 30 cm deep, + T = 30 cm above measured thermocline, T = at measured thermocline, - T = 30 cm below measured thermocline, and BOT = 60 cm above sediment.

1.2 • mL $^{-1}$, and sample injection volume of 100 μL. Run time for a sample was approximately 10 min, with retention time for a fluridone peak at 7 min. The reporting limit for this method is 1.0 μg • L^{-1}.

The FasTEST technique applies the principle of ELISA to the determination of fluridone residues in water samples. An aliquot of the sample is mixed with an enzyme conjugated fluridone analog in a disposable test tube. Paramagnetic

particles coated with antibodies specific to fluridone are added to the tube. Both fluridone and enzyme-conjugated fluridone analogs bound to the anitbodies on the particles are held in the tube by the magnetic field, while the unbound reagents are decanted. After decanting, the particles are washed to remove the unbound enzyme conjugate. Presence of fluridone is detected by adding the enzyme substrate and chromagen, thus generating a colored product. After a 20-min incubation period, the reaction is stopped and stabilized with the addition of acid. Since the enzyme-conjugated fluridone analog competes with the unlabeled fluridone for the antibody sites, the level of color development is inversely proportional to the concentration of fluridone in the water.

Quantification of fluridone residues is accomplished through generation of a standard curve using standards supplied with the ELISA kit (Strategic Diagnostics Inc. (SDI), Newark, DE). Absorbance at 450 nm is measured in each tube using an SDI RPA-1 Photoanalyzer. The standard curve is constructed using linear regression after a log/logit transformation of the concentration and absorbance values, respectively. The equivalent fluridone concentration in unknown samples is determined by the photoanalyzer. The reporting limit for this method is 1.0 $\mu g \bullet L^{-1}$.

As a result of the low turbidity and high clarity of the water samples, no pretreatment filtering was necessary and analyses were performed on raw water. For analyses, 200 μL of sample was mixed with 800 μL of diluent for a 1-mL total sample as a 5X dilution. A 250-μL sample was withdrawn for analysis. In cases where higher dilution was necessary, then 100 μL of the 5X mix was withdrawn and mixed with 900 μL of diluent, to create a 50X dilution. A total of 60 samples can be analyzed with each set. Computer software furnished with the system provides a means of obtaining the curve and calculated results. All unknown samples were analyzed against the standard curve. A new standard curve was constructed for each set of samples analyzed.

A series of blind sample spikes and blank spikes (distilled water) were integrated into the field sample batches. Analytical grade fluridone (99.1 percent purity) was used to create the stock solution used to spike samples at 4 $\mu g \bullet L^{-1}$. percent fluridone recovery, following procedures used for spiked samples, was determined.

Water Temperture Monitoring

Water temperature was measured at all water residue sampling events using a Hydrolab Surveyor II (Hydrolab Corp, TX). Surface water temperature was measured at the littoral stations and thermal profiles were measured at the deep stations to determine the water-column stratification. Measurements recorded from the thermal profile were used to determine specific locations in the water column for the deep-station sampling events (Table 8). Deep-station samples were collected at the surface, just above the thermocline, and from the hypolimnion.

Submersed Plant Surveys

Quantitative sampling of vegetation was performed using point-based frequency of species occurrence to evaluate whole-lake distribution and diversity of the submersed plant community of all eight study lakes. This technique was implemented using grid locations determined by a geographic information system (GIS) and located on each lake using a GPS mounted on a survey boat (Madsen 1999). This type of sampling protocol allowed for a rigorous statistical analysis of the data. Point-based frequency of sampling required up to 2 days per lake to complete and was conducted in the spring (early to mid-May) and in summer (mid-August) on each lake in 1997 (year of herbicide treatment) and in 1998 (12 and 15 months posttreatment). This bimodal, 2-year sampling schedule allowed for changes in submersed plant communities to be compared within the year of treatment and across two successive growing seasons.

For each study lake, a grid of sample points was developed using MapInfo (MapInfo Corp., Troy, NY), a desk-top mapping program similar to a GIS (Figures 6 through 13). The minimum grid resolution was 50 m by 50 m. At least 200 points were visited on each lake, with a maximum of 500 points evaluated dependent upon lake size. Map lake boundaries were taken from digital county highway map database provided by MapInfo. Once on a lake, a GeoExplorer II GPS (Trimble Corp., Santa Rosa, CA) was used to accurately locate sampling points. At each point, water depth was measured, and each species present (in an area approximately one meter square) was identified and recorded. An aquascope was used to aid in underwater viewing of plants. If plants could not be clearly identified from the surface, or if plants at the bottom could not be seen, a rake-type sampling device was lowered through the water column and plants were brought to the surface for species verification. Voucher specimens representing all submersed plant species observed on the study lakes were collected and archived at the USAERDC LAERF herbarium. Any unknown or questionable species were sent to C. Barre Hellquist (North Adams State College, North Adams, MA) for verification.

During the 2-year study period, the contact herbicide diquat [6,7-dihydrodipyrido(1,2-a:2′,1′-c)pyrazinediium dibromide] and several types of chelated copper algaecides were used to control nuisance levels of native plants and algae in limited nearshore areas in some of the study lakes. This level of management was required to alleviate problems associated with excessive amounts of vegetation along the nearshore areas of lakeside residents and property owners. Since these treatments typically comprised less than 10 percent of the surface area of a lake, were in waters less than 1.3 m in depth, and only controlled some of the shoot mass of the treated vascular plants (due to the mode of action of the herbicides), they had a negligible effect on the whole-lake plant assessment results.

The maximum depth of aquatic vegetation in each lake was used to define the extent of the littoral zones (Table 1). Change in species distribution, or frequency, was evaluated using a Chi-square analysis on 2 by 2 by X tables of frequency in the littoral zone only. Change in diversity as measured by average number of species per sample site were statistically analyzed using a T-test or analysis of variance (ANOVA).

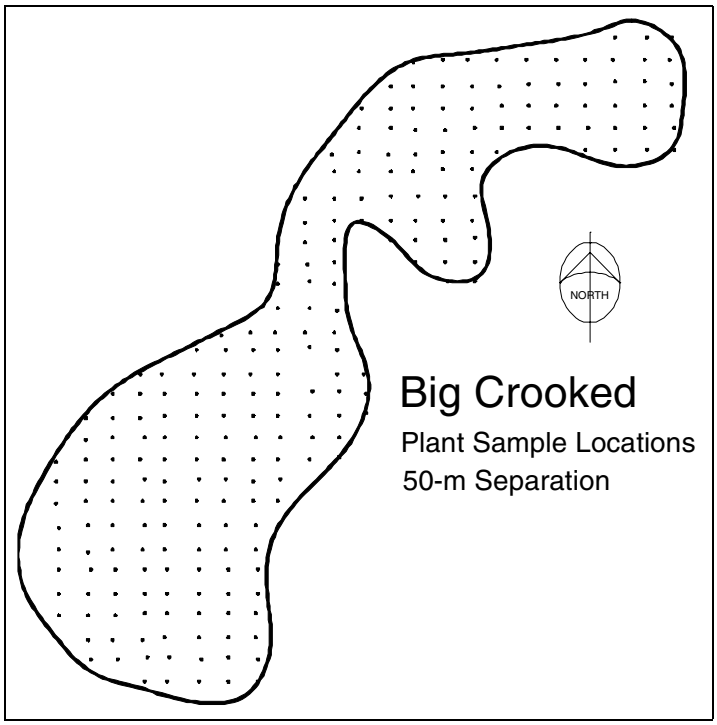

Figure 6. Plant sampling points grid for Big Crooked Lake, Kent County, Michigan, 1997-1998

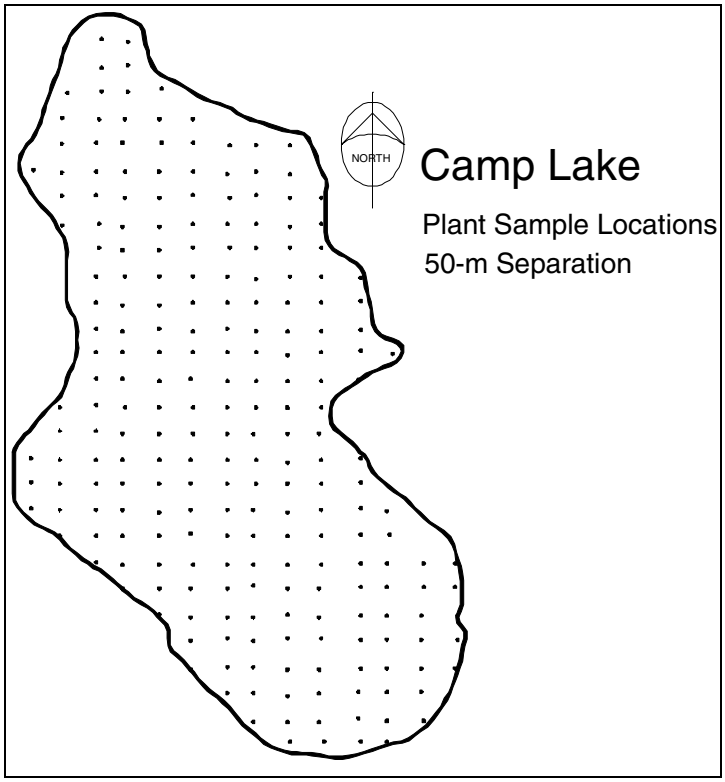

Figure 7. Plant sampling points grid for Camp Lake, Kent County, Michigan, 1997-1998

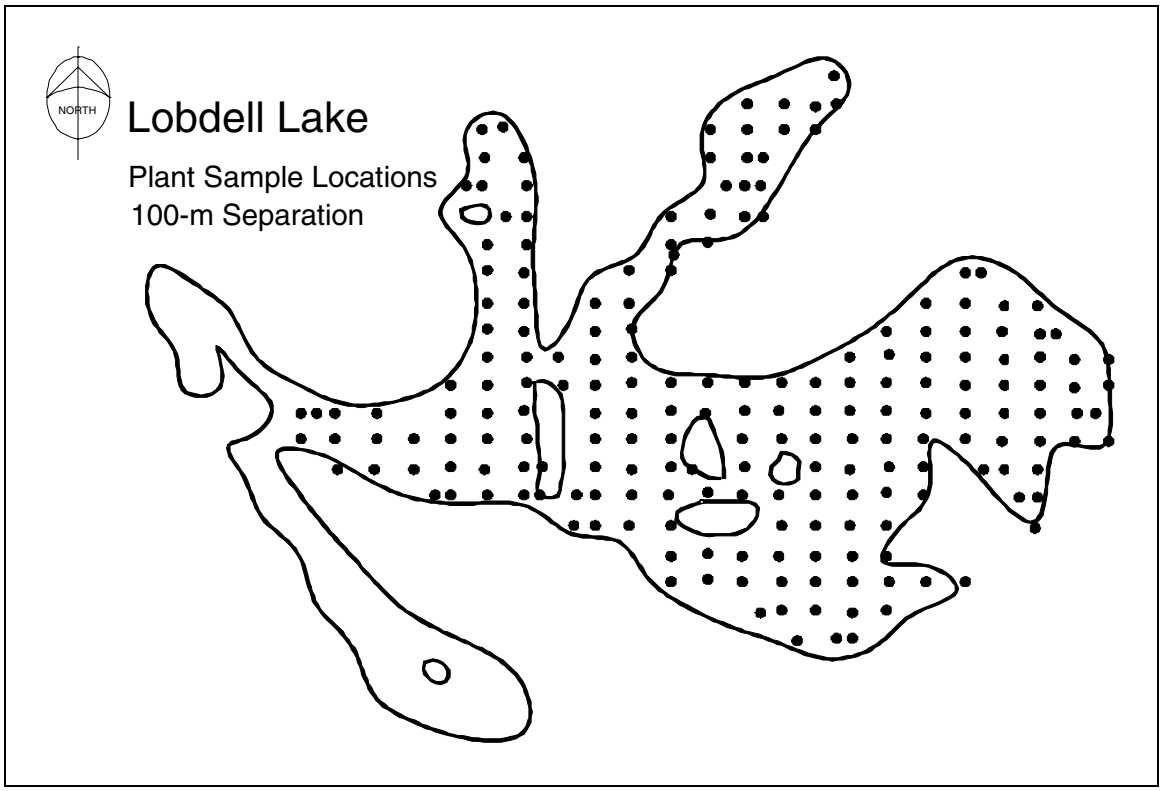

Figure 8. Plant sampling points grid for Lobdell Lake, Genessee, and Livingston Counties, Michigan, 1997-1998

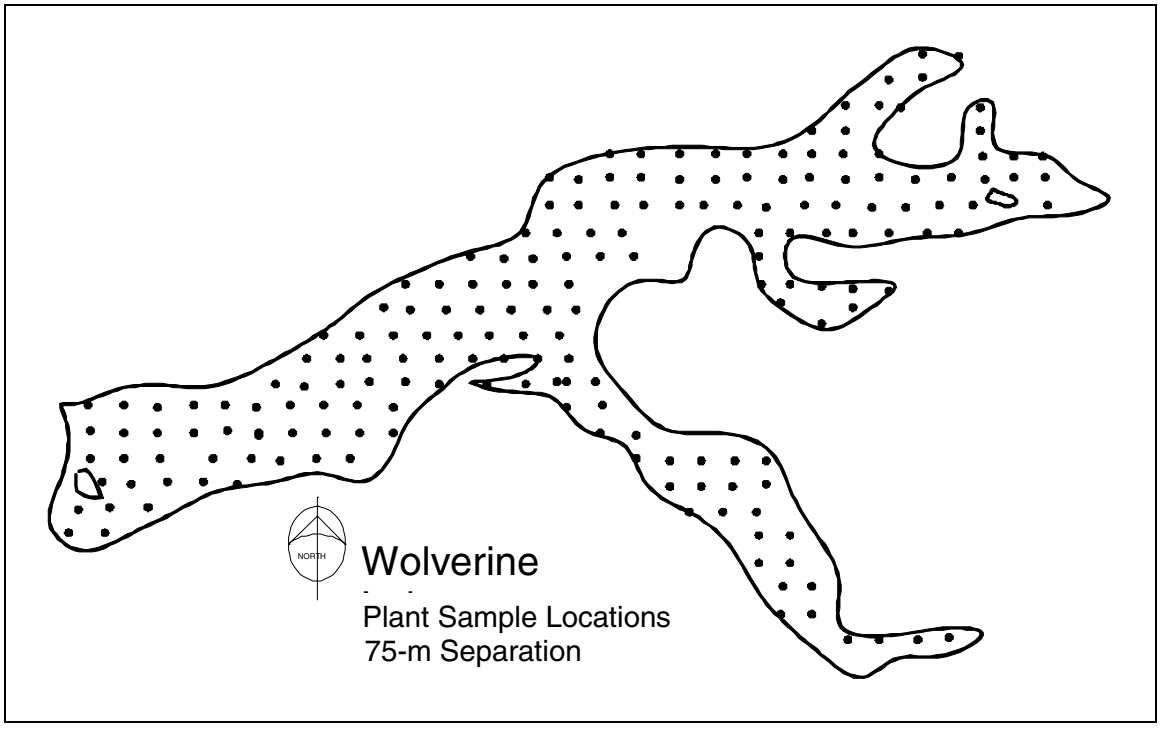

Figure 9. Plant sampling points grid for Wolverine Lake, Oakland County, Michigan, 1997-1998

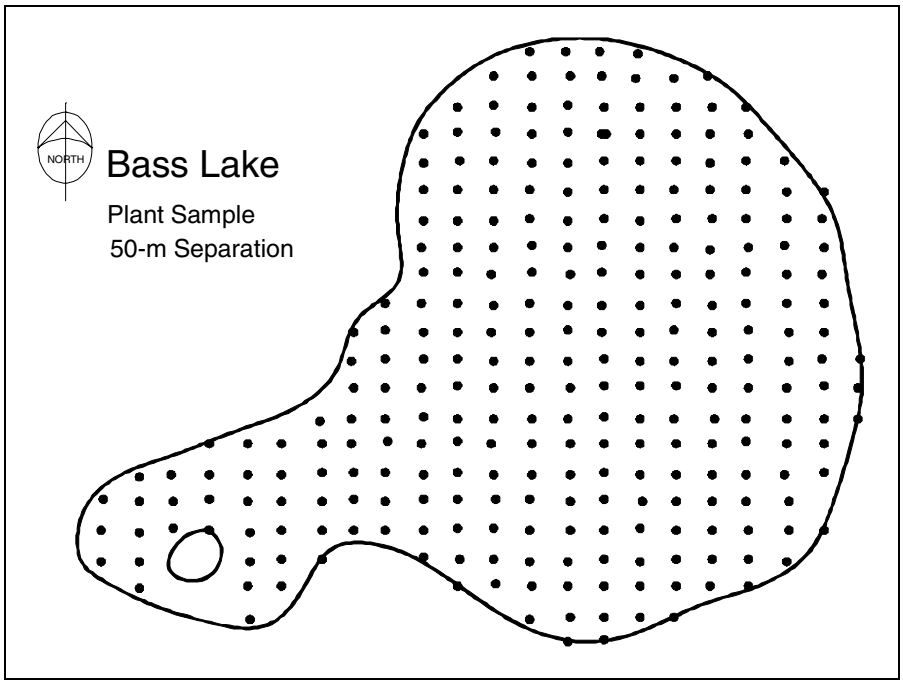

Figure 10. Plant sampling points grid for Bass Lake, Kent County, Michigan, 1997-1998

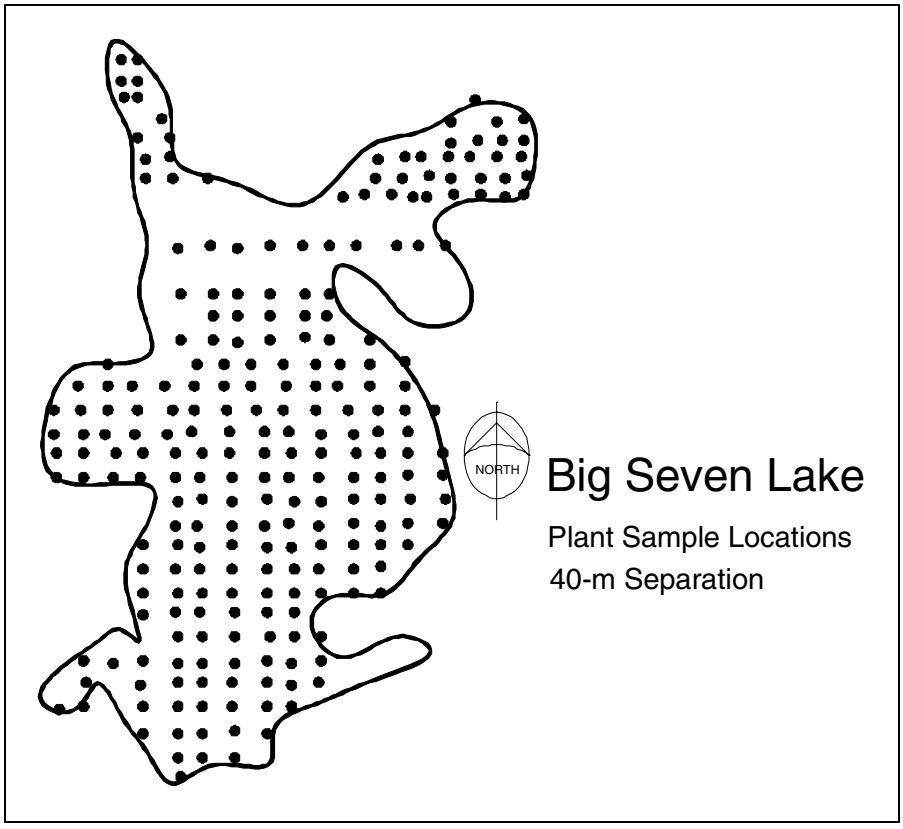

Figure 11. Plant sampling points grid for Big Seven Lake, Oakland County, Michigan, 1997-1998

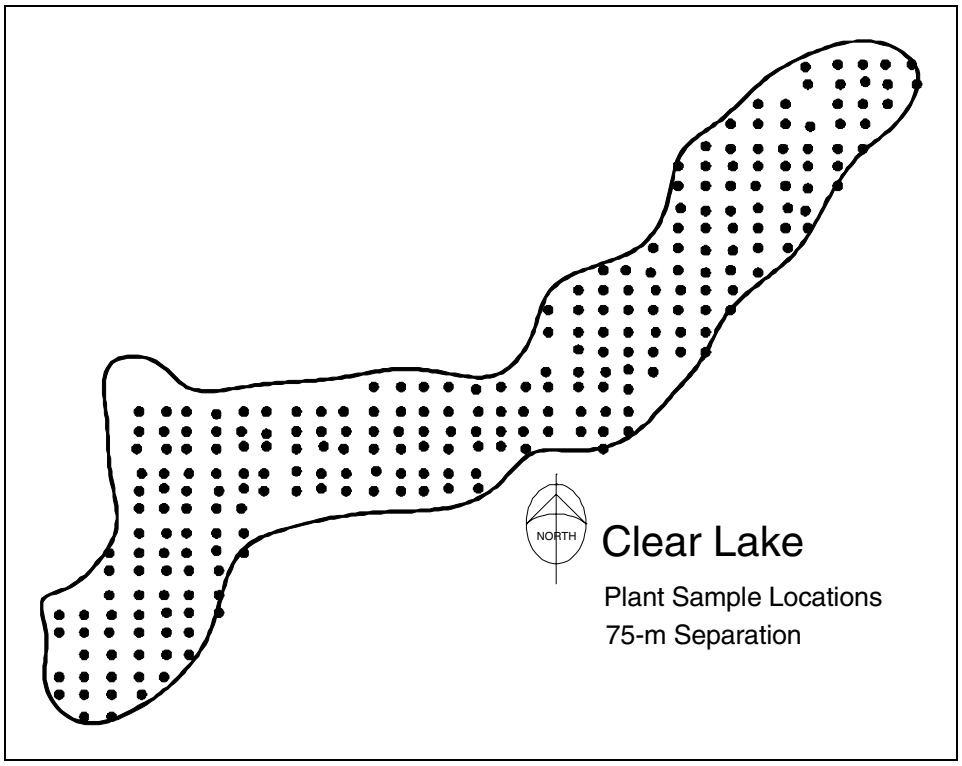

Figure 12. Plant sampling points grid for Clear Lake, Barry County, Michigan, 1997-1998

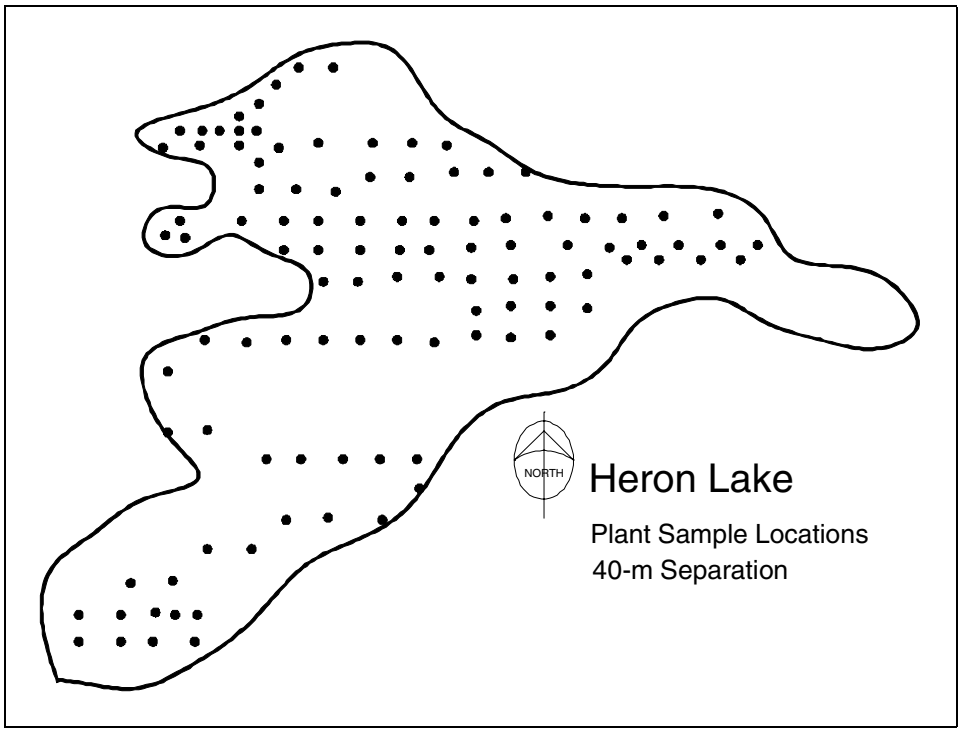

Figure 13. Plant sampling points grid for Heron Lake, Oakland County, Michigan, 1997-1998

3 Results and Discussion

Fluridone Applications

Water residues and temperature stratification

If fluridone is applied to an isothermal lake, the herbicide will become well distributed within the water column from the surface to the bottom. However, if fluridone is applied to a thermally stratified lake (warm epilminion, thermocline, cold hypolimnion), the herbicide should be well distributed throughout the isothermal epilimnion (regardless of depth) but remain isolated from the cold hypolimnetic waters below the thermocline. Weak thermal stratification that occurs on a diurnal basis in surface waters can possibly restrict the initial vertical distribution of fluridone. However, surface water turnover occurs over a period of several days, and fluridone mixes down to the well-established thermocline, which creates a barrier to further vertical mixing of the product. As the summer thermocline becomes established at shallower depths, some of the fluridone can be trapped in the hypolimnetic waters. In this study, different thermal stratification regimes occurred prior to initial fluridone applications (mid-May) and prior to the booster treatments (late May). When matched with measured fluridone residues, it becomes apparent that the vertical distribution of herbicide within the treated lakes was affected by these different stratification regimes.

Mean fluridone residues from littoral stations at all treatment lakes are presented in Table 9. Stations 2 and 3 in Lobdell Lake were not used in calculating mean residues for that lake. These stations, located in the eastern arm of Lobdell, were affected by water flow entering and exiting this arm of the lake, causing residue levels to fall below detection limits at 83 percent of the sampling dates. Mean residue data showed that by 1 DAIT, actual aqueous concentrations ranged from 10 percent above the nominal rate of 5 μg $\cdot$ L^{-1} to between 16 to 33 percent below the rate targeted for the 3.05-m-depth zone, and had declined by an additional 18 to 38 percent by 10 to 11 DAIT (Table 6). This relatively rapid loss of herbicide in the targeted treatment zone was most likely due to fluridone mixing to depths greater than 3.05 m. Water temperature measurements (Tables 10 through 13) indicate that at pretreatment, lake thermoclines ranged from 6 to 8 m in depth, with warm water in the epilimnion (10 to 11 °C) and cold water in the hypolimnion (6 to 7 °C). Under this stratification scenario, fluridone would be expected to mix and distribute throughout the nearly isothermal epilimnion (which was considerably deeper than the 3.05-m targeted

Table 9
Fluridone Water Residues (μg • L^{-1}) from Surface Stations of Four Treated Lakes in Michigan, May-August 1997. (Data represent mean values (± 1 SE), n = 6, except Lobdell where n = 4)

Lake	Pretreat	Days after Initial Treatment (DAIT) (Days after Booster Treatment (DABT))									Composite Mean Fluridone DAIT		
		1	4	7	10	17 (1)	26 (10)	36 (20)	47 (30)	75 (58)	10	20	75
Big Crooked													
Initial treat, 5/14/97	0.0	3.78	3.23	3.78	3.12	5.05	4.48	4.15	3.47	1.29	3.48	3.91	3.59
Booster treat, 5/30/97		±0.7	±0.4	±0.3	±0.3	±0.5	±0.2	±0.2	±0.1	±0.1	±0.1	±0.1	±0.3
Camp													
Initial treat, 5/14/97	0.0	4.20	2.53	2.68	2.56	4.84	3.87	3.63	2.86	2.02	2.99	3.45	3.24
Booster treat, 5/30/97		±1.5	±0.2	±0.3	±0.1	±0.2	±0.1	±0.2	±0.1	±0.4	±0.4	±0.4	±0.0
		1	**4**	**7**	**11**	**22 (1)**	**31 (10)**	**42 (21)**	**51 (30)**	**81 (60)**	**11**	**22**	**81**
Lobdell													
Initial treat, 5/12/97	0.0	5.50	4.99	4.21	3.37	4.87	5.05	3.56	2.76	1.97	4.52	4.58	4.03
Booster treat, 6/2/97		±2.0	±1.4	±0.2	±0.1	±0.8	±0.3	±0.4	±0.2	±0.2	±0.5	±0.4	±0.4
		1	**4**	**7**	**10**	**19 (1)**	**29 (11)**	**38 (20)**	**49 (31)**	**79 (61)**	**10**	**19**	**79**
Wolverine													
Initial treat, 5/12/97	0.0	3.35	3.10	2.89	2.61	3.29	3.16	2.73	1.42	1.31	2.99	3.07	2.65
Booster treat, 5/30/97		±0.3	±0.2	±0.1	±0.1	±0.2	±0.1	±0.1	±0.1	±0.1	±0.1	±0.1	±0.3

Table 10
Water Temperature and Fluridone Residues from Deepwater Stations In Big Crooked Lake, Michigan, Pretreatment through 47 Days Posttreatment, 1997. (Initial application on 14 May 1997; booster application on 30 May 1997)

Pretreatment, 10 May 1997

Station 7	Depth, m	T, C	Fluridone μg • L^{-1}	Station 8	Depth, m	T, C	Fluridone μg • L^{-1}
7.1	0.5	11.5	0.0	8.1	0.5	11.5	0.0
	1.0	11.5			1.0	11.5	
	2.0	11.4			2.0	11.5	
	3.0	11.4			3.0	11.4	
	4.0	11.3			4.0	11.4	
	5.0	11.3			5.0	11.1	
7.2	6.0	11.3	0.0	8.2	6.0	10.7	0.0
	7.0	10.9			7.0	10.0	
7.3	8.0	9.9	0.0	8.3	8.0	7.2	0.0
	9.0	7.3		8.4	9.0	6.8	0.0
7.4	10.0	6.6	0.0		10.0	6.8	
	11.0	6.4					
	12.0	6.2					
7.5	14.0	6.0	0.0	8.5	Missing		
	15.0	6.0					

Pretreatment Day 4, 18 May 1997

Station 7	Depth, m	T, C	Fluridone μg • L^{-1}	Station 8	Depth, m	T, C	Fluridone μg • L^{-1}
7.1	0.5	11.1	2.45	8.1	0.5	11.6	3.50
	1.0	11.1			1.0	11.6	
	2.0	10.7			2.0	11.3	
	3.0	10.6			3.0	10.8	
	4.0	10.5			4.0	10.7	
	5.0	10.5			5.0	10.6	
	6.0	10.4			6.0	10.5	

(Sheet 1 of 3)

Table 10 (Continued)

Pretreatment Day 4, 18 May 1997 (Continued)

Station 7	Depth, m	T, C	Fluridone µg • L^{-1}	Station 8	Depth, m	T, C	Fluridone µg • L^{-1}
7.2	7.0	10.4	2.10	8.2	7.0	10.3	3.15
	8.0	10.3			8.0	8.8	
7.3	9.0	9.4	1.65	8.3	9.0	7.9	<1
	10.0	6.9			10.0	7.0	
7.4	11.0	6.5	0.0		10.5	6.9	
	12.0	6.3		8.4	Missing		
	14.0	6.0		8.5			
7.5	15.0	6.0	0.0				
	16.0	6.0					

Posttreatment Day 10, 24 May 1997

Station 7	Depth, m	T, C	Fluridone µg • L^{-1})	Station 8	Depth, m	T, C	Fluridone µg • L^{-1}
7.1	0.5	15.1	2.30	8.1	0.5	15.5	3.20
	1.0	14.5			1.0	15.4	
	2.0	13.4			2.0	14.1	
7.2	3.0	12.9	2.15	8.2	3.0	12.9	1.35
	4.0	12.5			4.0	12.0	
	5.0	11.9			5.0	11.3	
	6.0	11.2			6.0	10.8	
7.3	7.0	10.7	1.80	8.3	7.0	10.4	3.50
	8.0	10.4			8.0	9.4	
	9.0	9.3		8.4	9.0	7.6	3.60
7.4	10.0	7.8	<1		10.0	7.3	
	11.0	6.5					
	12.0	6.3					
	14.0	6.1					
7.5	15.0	6.0	0.0	8.5	Missing		
	16.0	6.0					

Posttreatment Day 26, 9 June 1997

Station 7	Depth, m	T, C	Fluridone µg • L^{-1})	Station 8	Depth, m	T, C	Fluridone µg • L^{-1}
7.1	0.5	23.0	4.00	8.1	0.5	22.5	4.15
	1.0	22.0			1.0	21.0	
	2.0	21.0		8.2	2.0	20.5	4.10
	3.0	19.0			3.0	19.0	
	4.0	17.0		8.3	4.0	16.5	4.40
7.2	5.0	15.5	3.40		5.0	15.0	
7.3	6.0	12.5	3.55		6.0	12.0	
	7.0	11.5		8.4	7.0	10.0	4.40
	8.0	11.0			8.0	9.0	
	9.0	10.0		8.5	Missing		
	10.0	8.0					
	11.0	7.0					
	12.0	6.5					
	13.0	6.0					
7.4	14.0	6.0	0.0				
	15.0	6.0					
7.5	Missing						

Posttreatment Day 47, 30 June 1997

Station 7	Depth, m	T, C	Fluridone µg • L^{-1}	Station 8	Depth, m	T, C	Fluridone µg • L^{-1}
7.1	0.5	27.5	3.80	8.1	0.5	28.5	3.10
	1.0	27.5			1.0	28.5	
	2.0	27.5			2.0	28.0	
7.2	3.0	26.0	3.85	8.2	3.0	25.0	2.95
7.3	4.0	22.0	4.70	8.3	4.0	19.0	3.50
	5.0	18.5			5.0	16.0	
	6.0	14.5			6.0	14.0	

(Sheet 2 of 3)

Table 10 (Concluded)

Posttreatment Day 47, 30 June 1997 (Continued)							
Station 7	Depth, m	T, C	Fluridone $\mu g \cdot L^{-1}$	Station 8	Depth, m	T, C	Fluridone $\mu g \cdot L^{-1}$
7.4	7.0	12.0	3.35	8.3	7.0	12.0	
	8.0	11.0			8.0	11.0	
	9.0	10.0			9.0	10.0	
	10.0	8.5			10.0	8.5	
	11.0	8.0			11.0	8.0	
	12.0	7.0			12.0	7.0	
	13.0	7.0			13.0	7.0	
	14.0	6.5		8.4	14.0	6.5	0.0
	15.0	6.0			15.0	6.0	
7.5	Missing			8.5	Missing		

(Sheet 3 of 3)

Table 11
Water Temperature and Fluridone Residues from Deepwater Stations in Camp Lake, Michigan, Pretreatment through 47 Days Posttreatment, 1997. (Initial application on 14 May 1997; booster application on 30 May 1997)

Pretreatment, 11 May 1997							
Station 7	Depth, m	T, C	Fluridone $\mu g \cdot L^{-1}$	Station 8	Depth, m	T, C	Fluridone $\mu g \cdot L^{-1}$
7.1	0.5	10.9	0.0	8.1	0.5	10.9	0.0
	1.0	10.9			1.0	10.9	
	2.0	10.9			2.0	10.9	
	3.0	10.9			3.0	10.9	
	4.0	10.9			4.0	10.9	
	5.0	10.9			5.0	10.9	
	6.0	10.9			6.0	10.5	
7.2	7.0	10.6	0.0	8.2	7.0	10.1	0.0
	8.0	9.8			8.0	9.8	
7.3	9.0	9.4	0.0	8.3	9.0	7.6	0.0
	10.0	6.9			10.0	6.7	
7.4	11.0	6.5	0.0	8.4	11.0	6.4	0.0
	12.0	6.4			12.0	6.3	
7.5	13.0	6.3	0.0	8.5	13.0	6.2	0.0
	14.0	6.2			14.0	6.1	
Posttreatment Day 4, 18 May 1997							
Station 7	Depth, m	T, C	Fluridone $\mu g \cdot L^{-1}$	Station 8	Depth, m	T, C	Fluridone $\mu g \cdot L^{-1}$
7.1	0.5	11.3	1.95	8.1	0.5	10.8	2.45
	1.0	11.3			1.0	10.8	
	2.0	11.1			2.0	10.8	
	3.0	11.1			3.0	10.8	
	4.0	11.0			4.0	10.8	
	5.0	10.4			5.0	10.7	
	6.0	10.3			6.0	10.3	
	7.0	10.2		8.2	7.0	10.2	2.30
7.2	8.0	10.2	2.15		8.0	10.2	
	9.0	9.4		8.3	9.0	8.9	<1
7.3	10.0	8.1	<1		10.0	7.0	
7.4	11.0	6.8	0.0	8.4	11.0	6.4	0.0
	12.0	6.4			12.0	6.2	
7.5	13.5	6.1	0.0	8.5	13.0	6.1	0.0
	16.0	6.0			14.0	6.0	

(Continued)

Table 11 (Concluded)

Posttreatment Day 10, 24 May 1997							
Station 7	**Depth, m**	**T, C**	**Fluridone $\mu g \cdot L^{-1}$**	**Station 8**	**Depth, m**	**T, C**	**Fluridone $\mu g \cdot L^{-1}$**
7.1	0.5	14.9	2.60	8.1	0.5	15.0	2.20
	1.0	14.8			1.0	14.8	
7.2	2.0	14.6	2.30		2.0	14.1	
	3.0	13.2			3.0	12.6	
	4.0	12.1			4.0	11.9	
7.3	5.0	11.7	2.45	8.2	5.0	11.6	2.05
	6.0	11.3			6.0	11.3	
	7.0	10.9			7.0	11.1	
7.4	8.0	10.3	2.85		8.0	10.7	
	9.0	9.5			9.0	9.4	
	10.0	7.8		8.3	10.0	8.0	1.55
	11.0	6.9			11.0	7.1	
7.5	12.0	6.5	<1	8.4	12.0	6.8	0.00
	13.0	6.3		8.5	14.0	6.3	<1
	16.0	6.0			14.5	6.2	

Posttreatment Day 26, 9 June 1997							
Station 7	**Depth, m**	**T, C**	**Fluridone $\mu g \cdot L^{-1}$**	**Station 8**	**Depth, m**	**T, C**	**Fluridone $\mu g \cdot L^{-1}$**
7.1	0.5	21.5	3.80	8.1	0.5	22.0	4.15
	1.0	21.0			1.0	20.0	
	2.0	20.0			2.0	19.5	
7.2	3.0	19.0	4.05		3.0	19.0	
	4.0	17.5		8.2	4.0	18.0	4.50
7.3	5.0	14.0	3.85		5.0	16.0	
	6.0	12.0		8.3	6.0	13.0	3.85
	7.0	11.0			7.0	12.0	
	8.0	10.5			8.0	11.0	
7.4	9.0	10.0	2.05		9.0	9.5	
	10.0	8.5			10.0	9.0	
7.5	Missing				11.0	7.0	
					12.0	6.5	
					13.0	6.0	
				8.4	14.0	6.0	<1
				8.5	Missing		

Posttreatment Day 45, 30 June 1997							
Station 7	**Depth, m**	**T, C**	**Fluridone $\mu g \cdot L^{-1}$**	**Station 8**	**Depth, m**	**T, C**	**Fluridone $\mu g \cdot L^{-1}$**
7.1	0.5	27.0	2.65	8.1	0.5	27.0	3.85
	1.0	27.0			1.0	27.0	
	2.0	27.0			2.0	26.5	
	3.0	26.5		8.2	3.0	25.0	3.90
7.2	4.0	23.0	2.80		4.0	20.5	
7.3	5.0	21.0	3.25	8.3	5.0	17.0	2.85
	6.0	15.5			6.0	12.0	
	7.0	13.5			7.0	11.0	
	8.0	12.0			8.0	10.5	
	9.0	10.5			9.0	9.5	
	10.0	10.0			10.0	9.0	
	11.0	9.5			11.0	8.0	
7.4	12.0	9.0	1.65		12.0	8.0	
	13.0	8.0			13.0	7.5	
7.5	Missing			8.4	14.0	7.0	<1
					15.0	7.0	
				8.5	Missing		

Table 12
Water Temperature and Fluridone Residues from Deepwater Stations in Lobdell Lake, Michigan, Pretreatment through 11 Days Posttreatment, 1997. (Initial application on 12 May 1997; booster application on 2 June 1997)

Pretreatment, 8 May 1997							
Station 7	Depth, m	T, C	Fluridone $\mu g \cdot L^{-1}$	Station 8	Depth, m	T, C	Fluridone $\mu g \cdot L^{-1}$
7.1	0.5	11.9	0.0	8.1	0.5	12.6	0.0
	1.0	11.9			1.0	12.6	
	2.0	11.8			2.0	12.5	
	3.0	11.8		8.2	3.0	12.4	0.0
	4.0	11.4			4.0	11.2	
7.2	5.0	11.6	0.0	8.3	5.0	10.5	0.0
	6.0	10.9		8.4	6.0	9.6	0.0
7.3	7.0	8.8	0.0		6.2	8.7	
	8.0	7.1		8.5	Missing		
	9.0	6.4					
7.4	10.0	6.2	0.0				
	11.0	6.0					
	12.0	5.8					
	14.0	5.3					
	16.0	4.7					
	18.0	4.4					
7.5	20.0	4.3	0.0				
	24.0	4.1					
Posttreatment Day 4, 16 May 1997							
Station 7	Depth, m	T, C	Fluridone $\mu g \cdot L^{-1}$	Station 8	Depth, m	T, C	Fluridone $\mu g \cdot L^{-1}$
7.1	0.5	10.5	2.60	8.1	0.5	11.0	5.05
	1.0	10.5			1.0	11.0	
	2.0	10.5			2.0	10.9	
	3.0	10.5		8.2	3.0	10.9	5.40
	4.0	10.5			4.0	10.8	
	5.0	10.4		8.3	5.0	10.8	4.30
7.2	6.0	10.3	2.60		6.0	10.3	
	7.0	10.1			6.5	10.3	
	8.0	8.7		8.4	Missing		
7.3	9.0	7.5	<1	8.5	Missing		
	10.0	6.3					
7.4	11.0	5.9	<1				
	12.0	5.6					
	14.0	5.1					
	16.0	4.7					
	18.0	4.4					
	20.0	4.3					
7.5	22.0	4.2	0.0				
	24.0	4.2					
Posttreatment Day 11, 23 May 1997							
Station 7	Depth, m	T, C	Fluridone $\mu g \cdot L^{-1}$	Station 8	Depth, m	T, C	Fluridone $\mu g \cdot L^{-1}$
7.1	0.5	13.9	2.45	8.1	0.5	14.8	5.10
	1.0	13.9			1.0	14.7	
	2.0	13.8		8.2	2.0	14.6	3.90
	3.0	13.0		8.3	3.0	13.1	4.20
7.2	4.0	12.6	2.15		4.0	11.8	
	5.0	11.9		8.4	5.0	11.8	4.50
	6.0	11.3			6.0	10.8	
	7.0	10.6		8.5	Missing		

(Continued)

Table 12 (Concluded)

Posttreatment Day 11, 23 May 1997(Continued)							
Station 7	Depth, m	T, C	Fluridone $\mu g \cdot L^{-1}$	Station 8	Depth, m	T, C	Fluridone $\mu g \cdot L^{-1}$
7.3	8.0	9.4	1.65	8.5	Missing		
	9.0	8.0					
	10.0	6.7					
	11.0	6.0					
7.4	12.0	5.7	0.0				
	14.0	5.2					
	16.0	4.8					
	18.0	4.6					
7.5	20.0	4.4	<1				
	22.0	4.4					
	24.0	4.3					

Note: Samples for posttreatment days 26 and 47 are missing.

Table 13
Water Temperature and Fluridone Residues from Deepwater Stations in Wolverine Lake, Michigan, Pretreatment through 10 Days Posttreatment, 1997. (Initial application on 12 May 1997; booster application on 30 May 1997)

Pretreatment, 9 May 1997							
Station 7	Depth, m	T, C	Fluridone $\mu g \cdot L^{-1}$	Station 8	Depth, m	T, C	Fluridone $\mu g \cdot L^{-1}$
7.1	0.5	11.5	0.0	8.1	0.5	11.7	0.0
	1.0	11.5			1.0	11.7	
	2.0	11.5			2.0	11.7	
	3.0	11.5			3.0	11.7	
7.2	4.0	11.5	0.0		4.0	11.4	
	5.0	11.5		8.2	5.0	10.7	0.0
7.3	6.0	8.0	0.0		6.0	10.4	
	7.0	6.8		8.3	7.0	9.0	0.0
7.4	8.0	6.3	0.0		8.0	7.4	
	9.0	6.2		8.4	9.0	6.6	0.0
	10.0	5.9			10.0	6.5	
7.5	12.0	5.8	0.0	8.5	Missing		
	13.0	5.8					
Posttreatment Day 4, 16 May 1997							
Station 7	Depth, m	T, C	Fluridone $\mu g \cdot L^{-1}$	Station 8	Depth, m	T, C	Fluridone $\mu g \cdot L^{-1}$
7.1	0.5	11.5	3.00	8.1	0.5	11.1	3.80
	1.0	11.4			1.0	11.0	
	2.0	11.3			2.0	11.0	
	3.0	11.2			3.0	10.9	
	4.0	11.1			4.0	10.9	
7.2	5.0	11.1	3.00		5.0	10.7	
	6.0	10.8		8.2	6.0	10.5	4.45
7.0	7.0	9.8	2.15		7.0	10.1	
	8.0	6.9		8.3	8.0	9.9	3.65
7.4	9.0	6.4	0.0		9.0	8.0	
	10.0	6.1		8.4	10.0	6.6	0.0
	11.0	5.9		8.5	Missing		
7.5	12.0	5.8	0.0				
	14.0	5.7					

(Continued)

Table 13 (Concluded)

Posttreatment Day 10, 22 May 1997							
Station 7	Depth, m	T, C	Fluridone $\mu g \cdot L^{-1}$	Station 8	Depth, m	T, C	Fluridone $\mu g \cdot L^{-1}$
7.1	0.5	15.8	2.30	8.1	0.5	16.0	2.70
	1.0	15.4			1.0	15.9	
	2.0	13.9			2.0	14.4	
7.2	3.0	13.1	2.45	8.2	3.0	13.6	2.90
	4.0	12.7			4.0	13.3	
	5.0	11.0			5.0	12.3	
	6.0	10.5		8.3	6.0	11.1	3.40
7.3	7.0	9.9	2.35		7.0	10.8	
	8.0	8.0			8.0	10.4	
7.4	9.0	6.4	0.0	8.4	9.0	8.5	1.55
	10.0	6.0			10.0	6.8	
	11.0	5.9		8.5	Missing		
7.5	12.0	5.8	<1				
	13.0	5.7					

treatment zone), and to not penetrate below the thermocline into the hypolimnetic zone. Comparisons of depth zone volumes in the lakes, water temperature profiles, and fluridone residues support this expected fluridone dilution in the upper 3.05-m-depth zone of the epilimnion, and the lack of residues in the hypolimnion.

Calculated volumes for the 3.05-m-depth zone, and deeper reaches, are presented as a proportion of total lake volume in Table 6. Calculated volumes from the 3.05- to 9.15-m (10- to 30-ft-) depth zone account for 26 to 45 percent of lake volumes in Big Crooked, Camp, and Wolverine, which could explain the 16- to 33-percent loss of fluridone in the 0.0- to 3.05-m (0- to 10-ft) depth zone in those lakes. The exception was Lobdell, which shows residues 10 percent greater than nominal. However, this lake contained the smallest calculated 3.05- to 9.15-m depth zone volume (18 percent), and by far the greatest 0- to 3.05-m-depth zone volume (73 percent), the zone targeted for the nominal rate. Measured residues from deep stations at 4 and 10 DAIT (Tables 10 through 13) show that fluridone did mix to depths well below 3.05 m, and were absent, or nearly so, below the established thermoclines. With the epilimnion (0.0 to 8.0 m) nearly three times deeper than the targeted application depth zone (0.0 to 3.05 m) and comprising a significant proportion of the lake volume, the fluridone mixed below the targeted zone and the nominal application rate were not achieved in three of the lakes.

Since a different thermal stratification pattern existed in the lakes just prior to booster treatments, vertical mixing of fluridone was also somewhat different than what occurred at the initial applications. At 10 DAIT (just prior to booster treatments), temperature profiles (Tables 10 through 13) showed that thermoclines were becoming established at 2 to 4 m, almost completely within the 3.05-m targeted treatment zone. Water temperatures in the 2- to 4-m-depth zone ranged from 14 to 16 °C. A gradient of temperatures (8 to 13 °C) extended below the 2- to 4-m zone, reaching depths of 9 to 10 m in depth, below which a nearly isothermal (6 to 7 °C) hypolimnetic zone still existed. Under this

stratification scenario, booster fluridone residues should not have mixed much below the 3.05-m-depth zone, and the nominal booster application rate should have been achieved; which effectively occurred in three of the lakes (Table 6). In Big Crooked and Camp, fluridone residues in littoral stations were reduced by only 0 to 4 percent at 1 day after booster treatment (DABT) and by an additional 10 to 19 percent at 10 to 11 DABT. In Lobdell, residues were reduced by 2 percent at 1 DABT but showed a slight increase (2 percent) at 11 DABT. However, in Wolverine, residues declined by 34 percent at 1 DABT (similar to the decline observed following the initial treatment) and decreased by an additional 3 percent at 10 DABT. The low residues in Wolverine, which exhibited a 2-m deep thermocline at booster treatment, cannot be adequately explained. Perhaps the out-dated contour map used to determine lake volumes in Wolverine was inaccurate. As expected, water residues from deep-station profiles (Tables 10 through 13) showed that very little, if any, fluridone was present in the cold hypolimnion through 47 DAIT.

Big Crooked Lake

Mean fluridone levels from littoral stations in Big Crooked Lake, pretreatment to 75 DAIT, are presented Table 9. Levels measured 3.78±0.7 $\mu g \cdot L^{-1}$ at 1 DAIT, fell to 3.12±0.3 $\mu g \cdot L^{-1}$ at 10 DAIT, and were boosted back to 5.05±0.5 $\mu g \cdot L^{-1}$ by 1 DABT. Levels slowly declined to 1.29±0.1 $\mu g \cdot L^{-1}$ by 58 DABT (75 DAIT). Fluridone levels averaged 3.48+0.1 $\mu g \cdot L^{-1}$ over the 10 DAIT period, 3.91+0.1 $\mu g \cdot L^{-1}$ over the 20 DAIT period, and 3.59±0.3 $\mu g \cdot L^{-1}$ over the 75 DAIT period (Table 9). Results from deepwater sampling locations (Table 10) showed no measurable fluridone residues were found in the cold (6 to 6.5 °C), hypolimnetic-water layer through the 47 DAIT collection period.

Camp Lake

Mean fluridone levels from littoral stations in Camp Lake, pretreatment to 75 DAIT, are presented in Table 9. Levels measured 4.20±1.5 $\mu g \cdot L^{-1}$ at 1 DAIT, fell to 2.56±0.1 $\mu g \cdot L^{-1}$ at 10 DAIT and were boosted back to 4.84±0.2 $\mu g \cdot L^{-1}$ by 1 DABT. Levels slowly declined to 2.02±0.4 $\mu g \cdot L^{-1}$ by 58 DABT (75 DAIT). Fluridone levels averaged 2.99+0.4 $\mu g \cdot L^{-1}$ over the 10 DAIT period, 3.45±0.4 $\mu g \cdot L^{-1}$ over the 20 DAIT period, and 3.24±0.3 $\mu g \cdot L^{-1}$ over the 75 DAIT period (Table 9). Results from the deep-water sampling locations (Table 11) showed little to no measurable fluridone residues found in the cold (6 to 7 °C), hypolimnetic-water layer through the 47 DAIT collection period.

Lobdell Lake

Mean fluridone levels from littoral stations in Lobdell Lake, pretreatment to 81 DAIT, are presented in Table 9. Levels measured 5.5±2.0 $\mu g \cdot L^{-1}$ at 1 DAIT, fell to 3.37±0.1 $\mu g \cdot L^{-1}$ at 11 DAIT, and were boosted back to 4.87±0.8 $\mu g \cdot L^{-1}$ by 1 DABT. Levels slowly declined to 1.97±0.2 $\mu g \cdot L^{-1}$ by 60 DABT

(81 DAIT). Fluridone levels averaged 4.52±0.5 μg • L^{-1} over the 11 DAIT period, 4.58+0.4 μg • L^{-1} over the 22 DAIT period, and 4.03±0.4 μg • L^{-1} over the 81 DAIT period (Table 9). Results from the deepwater sampling locations (Table 12) showed little to no measurable fluridone residues found in the cold (4 to 6 °C), hypolimnetic-water layer through the 11 DAIT collection period.

Wolverine Lake

Mean fluridone levels from littoral stations in Wolverine Lake, pretreatment to 79 DAIT, are presented in Table 9. Levels measured 3.35±0.30 μg • L^{-1} at 1 DAIT, fell to 2.61±0.1 μg • L^{-1} at 10 DAIT, and were boosted back to 3.29±0.2 μg • L^{-1} by 1 DABT. Levels slowly declined to 1.31±0.1 μg • L^{-1} by 61 DABT (79 DAIT). Fluridone levels averaged 2.99±0.1 μg • L^{-1} over the 10 DAIT period, 3.07±0.1 μg • L^{-1} over the 19 DAIT period, and 2.65±0.3 μg •L^{-1} over the 79 DAIT period (Table 9). Results from the deep-water sampling locations (Table 13) showed little to no measurable fluridone residues found in the cold (5 to 6 °C), hypolimnetic-water layer through the 10 DAIT collection period.

Correlation of HPLC and ELISA Techniques

Comparisons obtained from the HPLC and ELISA measurements for each fluridone-treated lake and a composite for all fluridone-treated lakes combined are presented in Figures 14 through 18. ELISA results compare well to HPLC results indicating that the ELISA method maintained a good linear estimate of fluridone concentrations throughout the sampling period. Since a good correlation ($r^2 = 0.80$) was established between the two analytical techniques, results from the ELISA method were used to report residue data in this manuscript.

Analysis of blank spike samples (4.0 μg • L^{-1}) resulted in a 98-percent recovery range (93 to 107 percent) of fluridone using the ELISA technique, and a 93-percent recovery range (83 to 102 percent) using the HPLC technique. Analysis of spiked field samples (4.0 μg • L^{-1}) resulted in an 89-percent recovery range (84 to 103 percent) using ELISA and an 88-percent recovery range (75 to 102 percent) using HPLC. Although problems with cross reactivity have been noted in some ELISA methods (Lydy, Carter, and Crawford 1996), this problem is not expected when measuring fluridone because of its unique chemical structure and the sole use of this product as an aquatic herbicide.

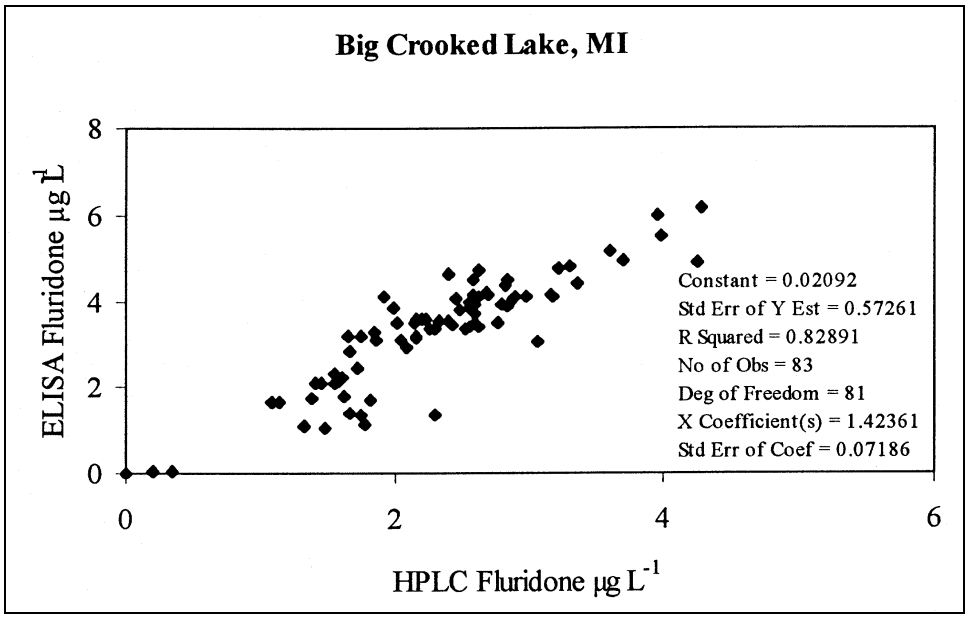

Figure 14. Comparison of ELISA and HPLC analytical methods for fluridone residues in water, Big Crooked Lake, Michigan, 1997

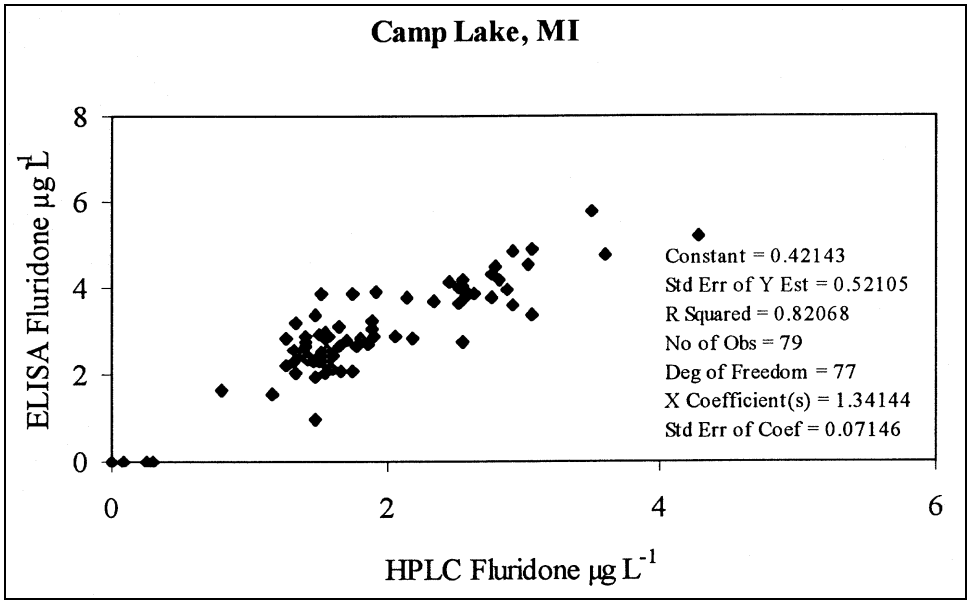

Figure 15. Comparison of ELISA and HPLC analytical methods for fluridone residues in water, Camp Lake, Michigan, 1997

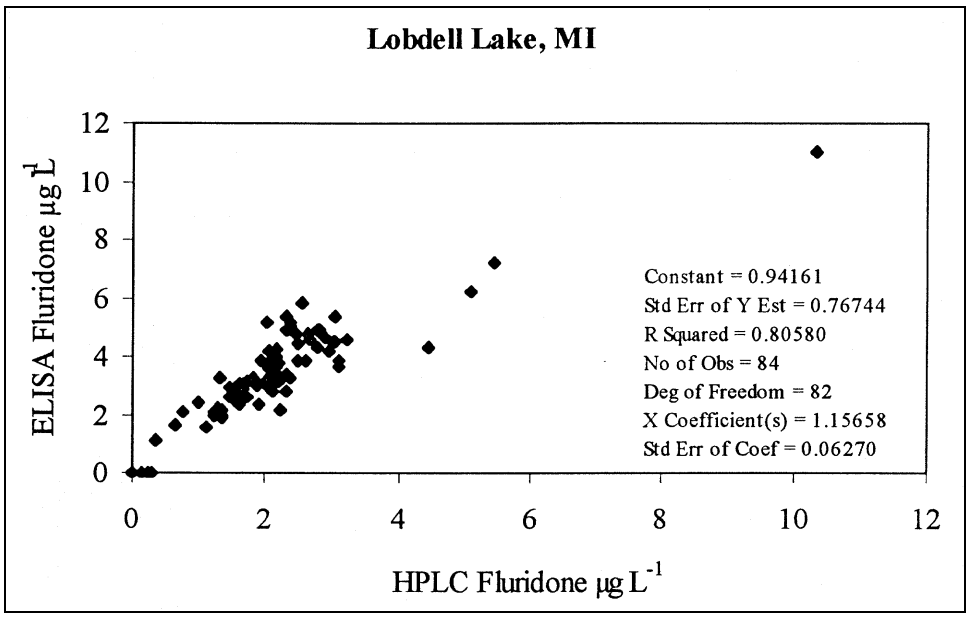

Figure 16. Comparison of ELISA and HPLC analytical methods for fluridone residues in water, Lobdell Lake, Michigan, 1997

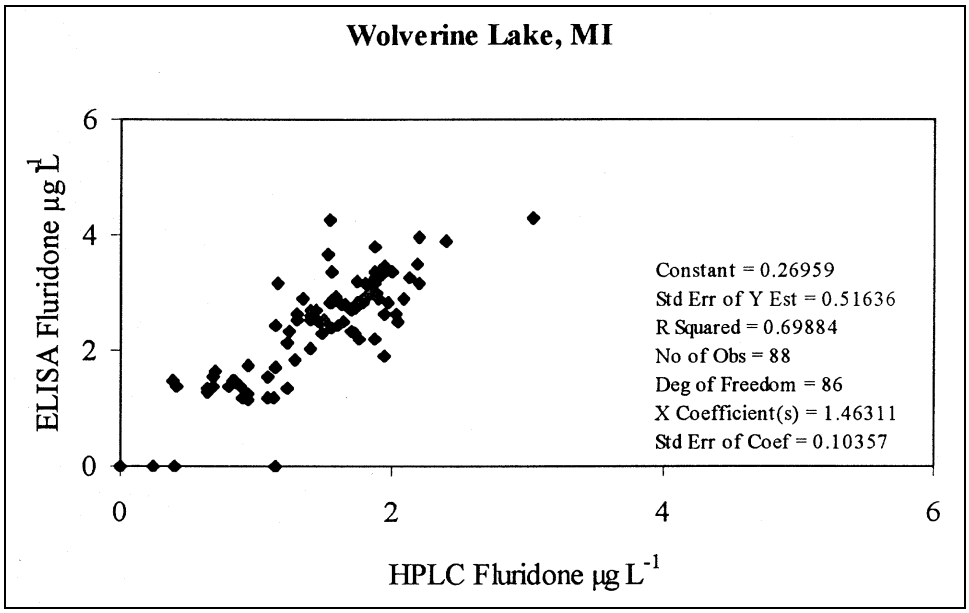

Figure 17. Comparison of ELISA and HPLC analytical methods for fluridone residues in water, Wolverine Lake, Michigan, 1997

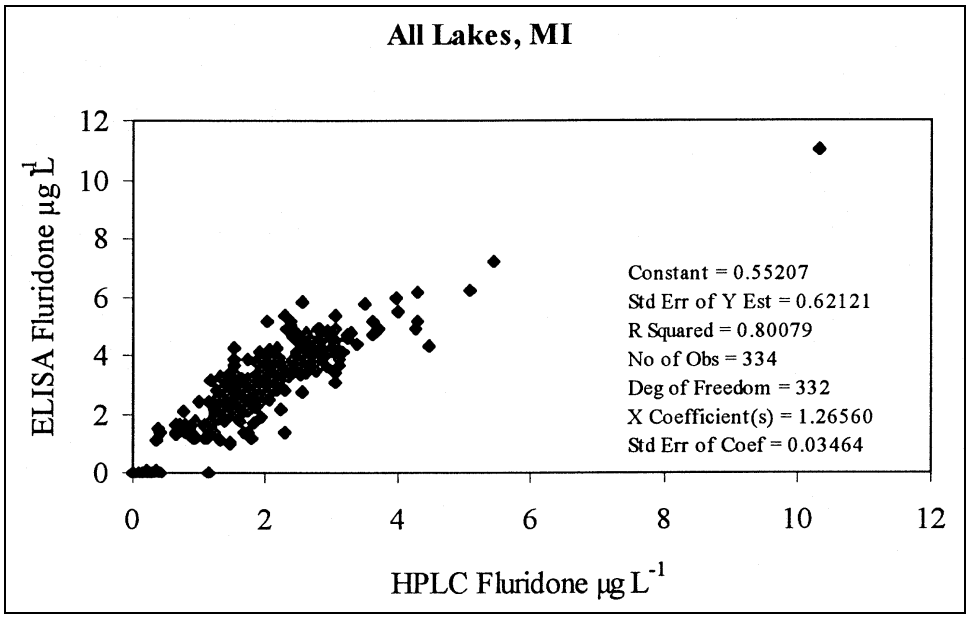

Figure 18. Comparison of ELISA and HPLC analytical methods for fluridone residues in water, all treated lakes, Michigan, 1997

As a result of the required extraction phase, HPLC methods are costly and time consuming, especially when measuring fluridone at lower concentrations. These factors reduce the effective use of HPLC-derived residue data for low-dose fluridone application strategies that must be implemented within short time periods. A major advantage of employing the ELISA method is the ability to analyze either a small or very large number of samples and provide data within 24 to 48 hr to aquatic plant managers, who can then use that information to modify application strategies, if required. Flexibility in application strategy is a critical component of selective weed management when using low-dose, whole-lake treatment schemes. To date, the major disadvantage of using the ELISA method for measuring fluridone in water has been the uncertainty of the accuracy of results compared to traditional HPLC methods. However, results from this study show that the methods are statistically compatible.

Fluridone Treatment Effects on the Submersed Plant Communities

Eurasian watermilfoil control

Eurasian watermilfoil was controlled in all fluridone-treated lakes, except for Wolverine (Figure 19). Control was excellent in Big Crooked, with a 100-percent reduction in frequency measured in the year of treatment (1997) and only a 7-percent frequency of occurrence measured by August 1998, after 15 months posttreatment. Control in Camp was also very good, with a 95-percent reduction in frequency in the year of treatment and slight recovery (14-percent frequency of occurrence) observed by August 1998. Fluridone

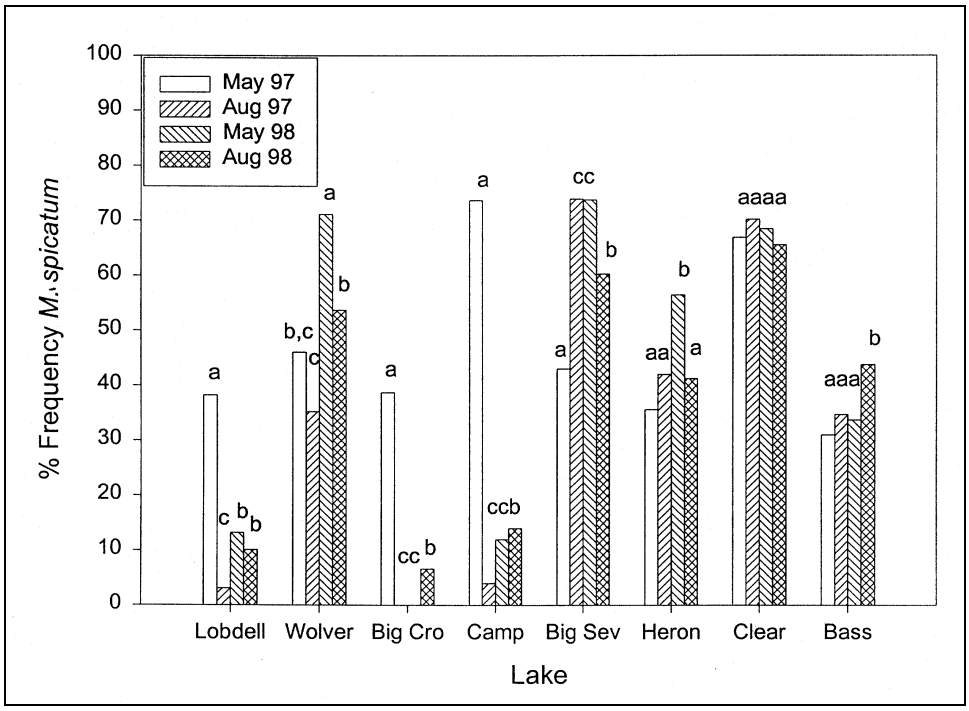

Figure 19. Percent frequency of Eurasian watermilfoil (*Myriophyllum spicatum*) in fluridone-treated and untreated lakes in Michigan, 1997-1998

provided a 93-percent reduction in frequency of Eurasian watermilfoil in the year of treatment in Lobdell, but growth of that plant had recovered to 10-percent frequency of occurrence by August 1998.

In these three lakes, removal of Eurasian watermilfoil from upper levels of the water column did not take place until 8 to 12 weeks after initial herbicide application. This slow "knockdown" and collapse of the canopy were most likely caused by the low fluridone rates administered in these treatments and by the advanced growth stage of the plants at the time of treatment. An application earlier in the plant's growth cycle might have provided a more rapid knockdown. Field observations in other Michigan lakes, and in other states, have indicated that higher rates of fluridone (> 10 $\mu g \bullet L^{-1}$) can knock down standing beds of the plant in less than 6 weeks posttreatment. This low-dose, boost treatment regime, however, was considered an operational success on Big Crooked, Camp, and Lobdell, since the plant was effectively removed as a nuisance species in the lakes for two growing seasons. As predicted from earlier growth chamber and mesocosm studies (Netherland, Getsinger, and Turner 1993; Netherland and Getsinger 1995a,b), water residues in these lakes reached a high enough level and remained in contact with the plant for a long enough period of time to provide effective control.

In Wolverine, the treatment regime was considered an operational failure per adequately controlling Eurasian watermilfoil. In that lake, the frequency of the plant was only reduced by 27 percent in the year of treatment. And by August 1998, the frequency of occurrence was measured at 54 percent, which was

8 percent greater than that recorded at the pretreatment evaluation period the previous spring. The low residue levels measured in that lake through the posttreatment period, indicate that lethal levels of fluridone and adequate exposure periods of those levels were never achieved. Although the plant was not controlled, some opening of the plant canopy and smaller, stunted shoots were observed in most of the lake, particularly during the year of treatment.

In contrast to the treated lakes, frequency of Eurasian watermilfoil significantly increased in two of the untreated reference lakes, Big Seven and Bass, while levels of the plant remained essentially unchanged in the other two water bodies, Heron and Clear (Figure 19). The stable or increased growth of Eurasian watermilfoil in these reference lakes provided strong evidence that the decline of the plant in the treated lakes was a direct result of the herbicide application and was not a consequence of any natural or seasonal phenomena.

Curlyleaf pondweed control

The unique life cycle of curlyleaf pondweed allows this plant to grow rapidly in the early spring, form a dense canopy by May, and then decline naturally by late June (Nichols 1999). Moreover, in May and June, curlyleaf pondweed produces numerous compact axillary turions that serve as the source of growth and reinfestation the following spring. Therefore, fluridone applications in mid-May 1997 were likely conducted during peak biomass and just prior to a natural senescence of curlyleaf pondweed. However, under this scenario the timing of treatments would not have prevented production of turions. As exepcted for a plant that senesces in early summer, posttreatment evaluations in August 1997 showed a significant reduction in curlyleaf pondweed for both treated and untreated lakes.

Curlyleaf pondweed frequency did increase in all fluridone-treated lakes between May 1997 and May 1998 (Figure 20). This expanded growth represented 1.3- to 3.5-fold increase in curlyleaf pondweed frequency. Anecdotal evidence has suggested that curlyleaf pondweed growth can be stimulated in lakes that have been treated the previous spring with herbicides, including fluridone. This growth and is probably related to reduced competition via removal of Eurasian watermilfoil and the condition and abundance of the curlyleaf turion bank on a lake-specific basis. Although this growth release seems to have occurred on the treated lakes in this study, it must be noted that an increase, albeit to a lesser extent (1.2- to 1.6-fold), in curlyleaf pondweed frequency also occurred in two of the untreated reference lakes, Big Seven and Heron (Figure 20). This curlyleaf pondweed growth release was not measured in the other two reference lakes, Bass and Clear, because these lakes had an extremely low proportion of that plant (< 1-percent frequency) in their respective plant communities. The increase in curlyleaf pondweed frequency in untreated reference lakes indicates that some of the expanded growth observed in herbicide-treated lakes may have been related to natural or seasonal events. The warmer-than-normal temperatures experienced in southern Michigan during winter and spring 1998, for instance, could have contributed to boost growth rates typically exhibited by curlyleaf pondweed.

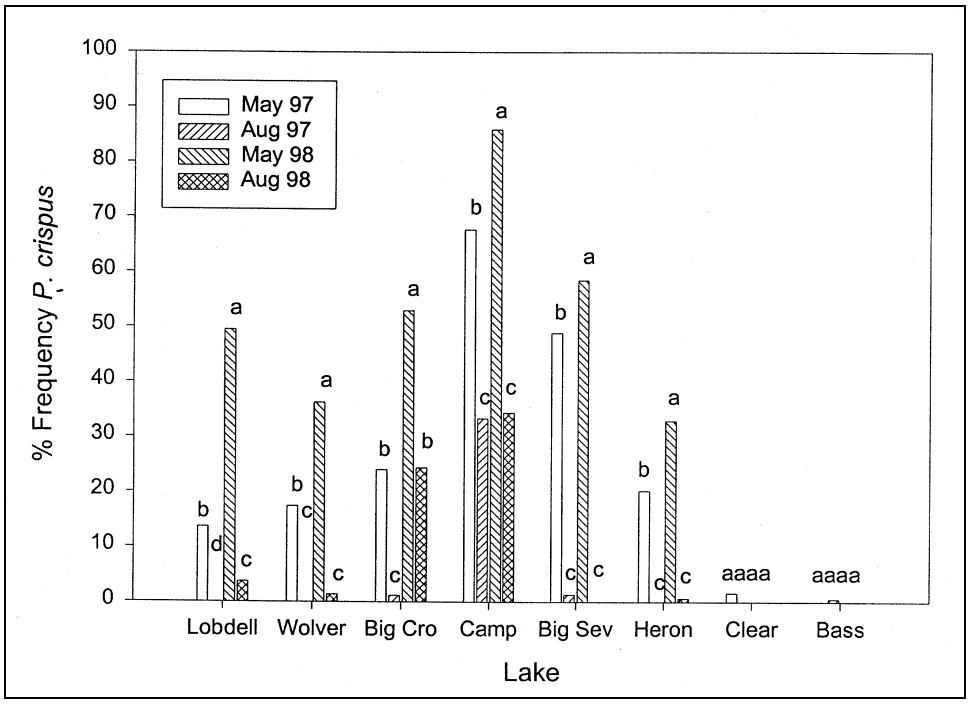

Figure 20. Percent frequency of curlyleaf pondweed (*Potamogeton crispus*) in fluridone-treated and untreated lakes in Michigan, 1997-1998

The increased proliferation of curlyleaf pondweed following fluridone applications suggests that timing of treatments can be critical when managing a lake for the invasive exotics curlyleaf pondweed and Eurasian watermilfoil. Fall fluridone applications and early spring treatments (late March through mid-April), conducted at rates that control Eurasian watermilfoil, can also control curlyleaf pondweed (authors' unpublished data). These early season treatments have the added benefit of controlling curlyleaf pondweed prior to formation of turions. Disrupting the life cycle of curlyleaf pondweed by preventing production of new turions is currently being investigated as a strategy to provide long-term control of this invasive weed. Data collected from Indiana and Michigan lakes treated with fluridone in the fall and early spring have demonstrated near 100-percent control of curlyleaf pondweed biomass, as well as a great reduction (60 to 90 percent) in viable turions (authors' unpublished data).

Total and native species diversity

Data presented in Figure 21 indicate that total submersed plant diversity (with Eurasian watermilfoil and curlyleaf pondweed included in the analyses) was significantly greater in the fluridone-treated lakes, both within the year of treatment and between pretreatment and 1-year posttreatment. The greatest change occurred between May 1997 and May 1998, where total diversity increased 1.5 to 2.3 fold. Total species diversity remained above the May 1997 levels through August 1998. Likewise, a significantly greater total species diversity was measured in the untreated reference lakes during the study period

(Figure 21). Diversity levels increased by factors similar to those measured in the treated lakes. While it is clear that the fluridone treatments did not reduce total plant species diversity, the reference lake data suggest that some of the increase in species diversity measured in the treated lakes could have been caused by natural or seasonal events.

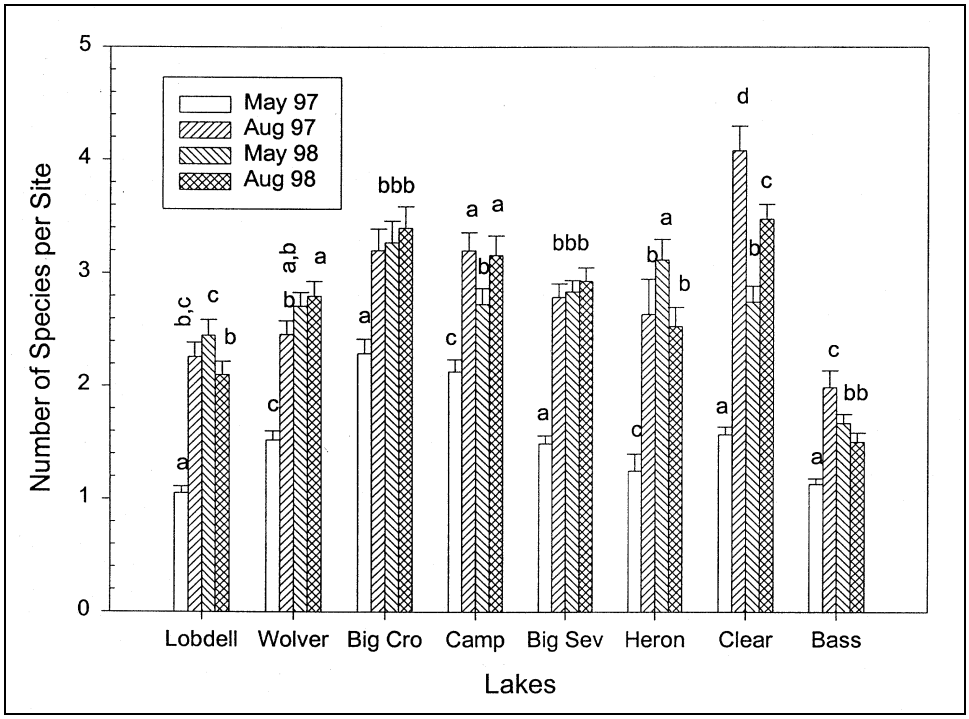

Figure 21. Number of all submersed plant species per site in fluridone-treated and untreated lakes in Michigan, 1997-1998

Native plant species diversity, calculated without the presence of Eurasian watermilfoil and curlyleaf pondweed, was roughly equivalent between the treated and untreated lakes (Figure 22) and exhibited posttreatment increases very similar to those seen for total species diversity. The greatest increase in species diversity occurred between May 1997 and May 1998, with species numbers still elevated above pretreatment levels in August 1998. Again, these data clearly show that fluridone treatments did not have a negative impact on species diversity, but the increases observed might have been related to natural events between seasons. Natural shifts in plant community assemblages occur in Northern tier lakes from early spring to late summer, and comparisons within a growing season could be confounded by these seasonal effects.

Total and native species plant cover (frequency of presence)

As an additional method for determining shifts in species diversity, the percent frequency of plant presence (plant cover) for total plant species (including Eurasian watermilfoil and curlyleaf pondweed) and for native plant

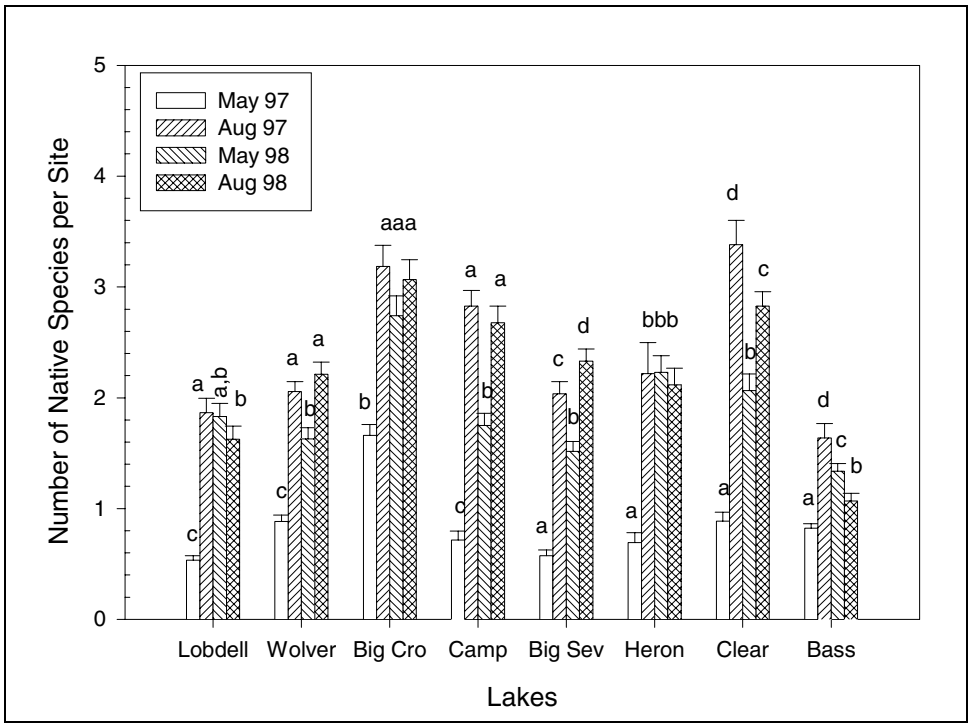

Figure 22. Number of native submersed plant species per site in fluridone-treated and untreated lakes in Michigan, 1997-1998

species (without the inclusion of Eurasian watermilfoil and curlyleaf pondweed) were compared for all lakes within the year of treatment (1997), and between years (1997 and 1998). These analyses indicate that total plant cover, and native plant cover, significantly increased or remained the same in all of the lakes, including those treated with fluridone (Figures 23 and 24). These data demonstrate that fluridone treatments did not have a negative impact on plant cover, but because of similar shifts in the untreated reference lakes, the increases observed might have been related to natural events between seasons. In all cases, posttreatment plant cover was maintained at levels above 60 percent, which exceeds the plant cover amounts (20 to 40 percent) in the littoral zone considered optimal for healthy fisheries in Northern tier lakes (Savino and Stein 1982; Wiley et al. 1984; Wiley, Tazik, and Sobaski 1987).

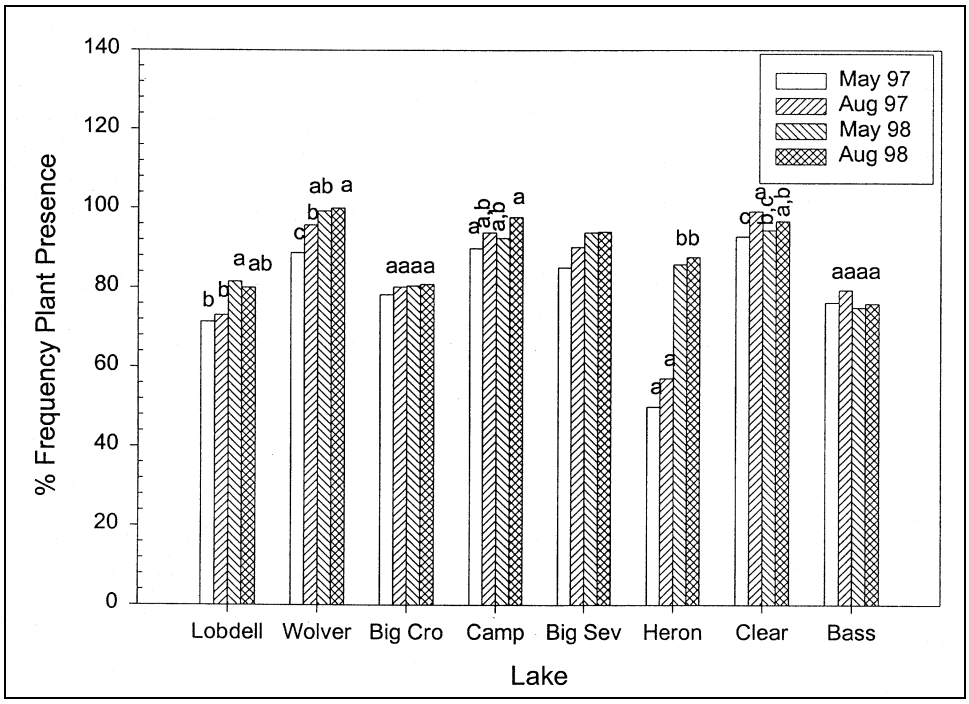

Figure 23. Percent frequency of all submersed plant species presence in fluridone-treated and untreated lakes in Michigan, 1997-1998

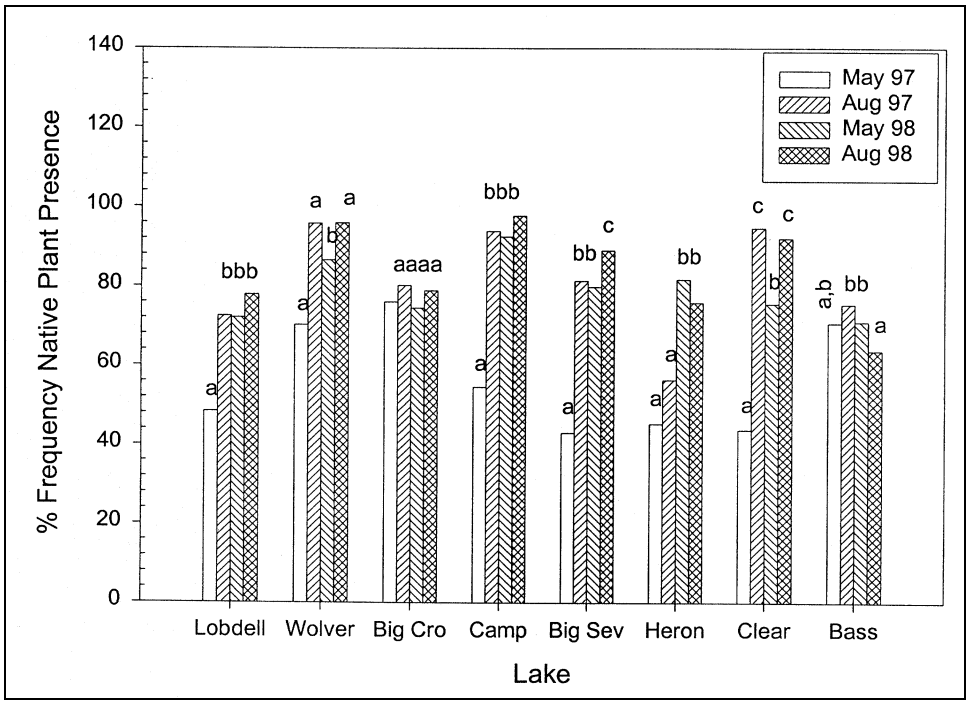

Figure 24. Percent frequency of native submersed plant species presence in fluridone-treated and untreated lakes in Michigan, 1997-1998

4 Conclusions and Recommendations

Conclusions

Based on the results of this investigation, the following conclusions can be drawn:

a. The low-dose, 5 μg • L^{-1} boost to 5 μg • L^{-1} whole-lake fluridone-treatment strategy can provide up to 100-percent control of Eurasian watermilfoil in the year of treatment and near 90-percent control through 15 months posttreatment, provided that adequate aqueous fluridone concentration/exposure time (CET) relationships are maintained. The appropriate fluridone CET relationship was maintained in three of the treated lakes (Big Crooked, Camp, Lobdell), and Eurasian watermilfoil was controlled (> 93 percent, year of treatment; > 86 percent, 15 months posttreatment).

b. If the required aqueous fluridone CET relationship is not maintained, failure to control Eurasian watermilfoil can also occur under the treatment strategy employed in this study. This was observed on Wolverine Lake where the required CET relationship was not met and limited control of Euarsian watermilfoil occurred in the year of treatment (27 percent), and increased in abundance (54 percent) by 1 year posttreatment.

c. The treatment strategy used in this study will not significantly impact the native plant species diversity or cover in the year of treatment, or through 15 months posttreatment.

d. The timing of fluridone applications were not conducive for long-term control of the invasive exotic plant curlyleaf pondweed. The increase in curlyleaf pondweed biomass in May 1998 was likely related to several factors including: 1) removal of Eurasian watermilfoil as a competitor; 2) mid-May 1997 fluridone applications did not prevent prolific formation of new turions that serve as a source of curlyleaf pondweed reinfestation the following year; and 3) increases in curlyleaf pondweed frequency in both treated and untreated lakes in May 1998 suggest that environmental

conditions may have been favorable for growth and expansion of that plant.

e. Fluridone residues will become well-mixed throughout depth zones exhibiting isothermal conditions; however, thermal stratification can restrict mixing of residues into deeper and colder waters (hypolimnion).

f. If the depth zone targeted for treatment is above the thermocline, fluridone residues will mix throughout the isothermal epilimnion, thereby reducing the nominal application rate in the targeted zone.

g. Results from this work demonstrate that the ELISA technique for aqueous fluridone measurements can be used as an accurate real-time tool for precision application enhancement.

Recommendations

Based on the results of this investigation, the following recommendations are provided:

a. If the 3.3-m-depth contour is employed to define lake volume used for calculating target treatment rates of fluridone and that contour is significantly above the level of the measured thermocline, the treatment strategy should be shifted to a higher initial and booster application rate, such as 6 $\mu g \bullet L^{-1}$. This slightly greater level of fluridone should mitigate some of the loss of aqueous residues from the targeted treatment zone, provide good control of Eurasian watermilfoil, and result in no significant impact on the native plant community.

b. If threshold fluridone CET requirements for selective control of Eurasian watermilfoil are to be successfully used in the field, managers should incorporate accurate bathymetric contours and temperature profiles into volume calculations used to determine whole-lake treatment rates. Without precise and up-to-date morphometric and thermal information, the finely tuned and narrowly defined laboratory-based CET principles are likely to be offset by unrefined field estimates. Ideally, water temperature profiles should be measured immediately prior to initial and booster applications, and that information should be used to determine the depth contour utilized for calculating the lake volume to be treated.

c. Since the expedient and reliable immunoassay water residue technique, FasTEST, can allow for flexible application strategies to be developed and used to ensure adequate and selective control of Eurasian watermilfoil, this diagnostic tool should be routinely used in whole-lake fluridone treatments.

d. Whole-lake treatment techniques for managing curlyleaf pondweed should be explored that emphasize timing of fluridone application with respect to phenological events of that target plant. Treatments in the early

spring or in fall may have the potential to disrupt the curlyleaf life cycle and provide control of that plant. These types of treatments may have the ability to control both curlyleaf pondweed and Eurasian watermilfoil when they co-exist in lakes.

e. Additional mesocosm studies should be conducted to more accurately determine the potential of fluridone to selectively control Eurasian watermifloil at rates between 5 and 10 $\mu g \bullet L^{-1}$.

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Appendix A
Procedure for Calculating Lake Volumes for Proposed Sonar Treatments[1]

This is the standard procedure used to calculate the volume of water within the upper 10 feet of a lake and to determine the appropriate amount of Sonar to apply. The goal of this procedure is to achieve rapid and uniform distribution of a given concentration of Sonar by treating water within the 0- to 5-foot-depth contour and within the 5- to 10-foot-depth contours separately with different amounts of Sonar. This procedure determines the amount of Sonar necessary to treat an entire lake to a depth of 10 feet at a given concentration.

Volume Calculations

a. Determine the surface acres of the 0-, 5-, and 10-foot-depth contours. Example: 0-, 5-, and 10-foot-depth contours are 239, 189, and 71 acres, respectively.

b. Use the following lake volume formula to calculate the volume of the lake between the surface and 5-foot depth.

V(ac/ft) = h/3 (A1 + A2 + [sq. rt. (A1 × A2)]), where V = volume, h = height of the water column, A1 = area of the lake surface, A2 = area of the 5-foot contour, A3 = area of the 10-foot contour. Results are in acre-feet. The volume of water to the 5-foot-depth contour = 5/3 (239 + 189 [sq. rt. (239 × 189)]) = 1,069 af.

c. Multiply the area of the 5-foot contour by 5 feet. 189a × 5f = 945 af.

d. Subtract Step 3 from Step 2. This equals the acre-feet in the 0- to 5-foot deep "donut" area. 1,069 - 945 = 124 af.

[1] DEQ – Michigan Department of Environmental Quality Land and Water Management Division.

e. Multiply Step 4 by 2.72. Then multiply that figure by the target concentration in parts per million. $124 \times 2.72 \times 0.005 = 1.7$ pounds (or quarts) of Sonar. A 1-quart container of Sonar AS contains 1 pound of active ingredient. Distribute this amount evenly in the 0- to 5-foot "donut" area.

f. Enter the 5- and 10-foot areas into the volume formula to find the volume of water between the 5- and 10-foot depths.
Volume = 5/3 (189 + 71 + [sq. rt. of (189×71)]) = 628 af.

g. Add Steps 3 and 6 to get the volume of the "donut hole" area below the 5-foot-depth contour to a depth of 10 feet (628 + 945 = 1,573 af).

h. Multiply Step 7 by 2.72. Then multiply by the target concentration in parts per **million**. $1{,}573 \times 2.72 \times 0.005 = 21.4$ pounds or quarts of Sonar. Distribute this amount evenly in the 0- to 10-foot "donut hole" area.

REPORT DOCUMENTATION PAGE

Form Approved
OMB No. 0704-0188

1. REPORT DATE *(DD-MM-YYYY)*	2. REPORT TYPE	3. DATES COVERED *(From - To)*
February 2001	Final Report	

4. TITLE AND SUBTITLE
Whole-Lake Applications of Sonar™ for Selective Control of Eurasian Watermilfoil

5a. CONTRACT NUMBER

5b. GRANT NUMBER

5c. PROGRAM ELEMENT NUMBER

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5d. PROJECT NUMBER

5e. TASK NUMBER

5f. WORK UNIT NUMBER

7. PERFORMING ORGANIZATION NAME(S) AND ADDRESS(ES)

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8. PERFORMING ORGANIZATION REPORT NUMBER

ERDC/EL TR-01-7

9. SPONSORING / MONITORING AGENCY NAME(S) AND ADDRESS(ES)
U.S. Army Corps of Engineers
Washington, DC 20314-1000;

Aquatic Ecosystem Restoration Foundation
Lansing, MI 48917-4048

10. SPONSOR/MONITOR'S ACRONYM(S)

11. SPONSOR/MONITOR'S REPORT NUMBER(S)

12. DISTRIBUTION / AVAILABILITY STATEMENT
Approved for public release; distribution is unlimited

13. SUPPLEMENTARY NOTES

14. ABSTRACT

The aquatic herbicide Sonar AS (fluridone) is being used in Northern tier states to selectively control the submersed exotic species Eurasian watermilfoil (*Myriophyllum spicatum* L.) growing in lakes and reservoirs. Reliable quantitative information linking changes in the submersed plant community following fluridone applications is limited, particularly with respect to water residue records; therefore, a study was conducted to investigate the impact of low-dose fluridone treatments on the aquatic plant communities in eight lakes of southern Michigan.

The main objective of the study was to determine whether submersed plant species diversity and frequency were impacted by low-dose fluridone applications in the year of treatment, when targeting for Eurasian watermilfoil control. Secondary objectives included (a) determining fluridone effectiveness on the exotic submersed species curlyleaf pondweed (*Potamogeton crispus* L.), (b) evaluating shifts in plant species diversity at 1-year posttreatment, (c) measuring the effect of thermal stratification on water column distribution of fluridone residues, and (d) verifying laboratory-derived results of fluridone concentration and exposure time relationships with respect to efficacy against Eurasian watermilfoil.

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15. SUBJECT TERMS
Aquatic herbicide | Fluridone | Low-dose herbicide
Aquatic plant control | Invasive species

16. SECURITY CLASSIFICATION OF:			17. LIMITATION OF ABSTRACT	18. NUMBER OF PAGES	19a. NAME OF RESPONSIBLE PERSON
a. REPORT	b. ABSTRACT	c. THIS PAGE			19b. TELEPHONE NUMBER *(include area code)*
UNCLASSIFIED	UNCLASSIFIED			52	

14. ABSTRACT

Study lakes were 55 to 220 ha in size and contained an average of nine species of submersed plants, including Eurasian watermilfoil and curlyleaf pondweed. Four lakes (Big Crooked, Camp, Lobdell, and Wolverine) were treated in mid-May 1997 with Sonar AS to yield a target concentration of 5 g • L^{-1} (ppb) fluridone in the upper 3.05 m (10 ft) of each lake. A sequential (booster) application of Sonar AS was conducted on each lake at 16 to 21 days after initial treatment (DAIT). This whole-lake booster application was intended to reestablish the target concentration of fluridone (5 g • L^{-1}) in the upper 3.05 m of each lake. Four water bodies (Bass, Big Seven, Clear, and Heron) received no fluridone applications and served as untreated reference lakes.

Water residue samples were collected on prescribed intervals on each of the fluridone-treated lakes from pretreatment up to 81 DAIT. Samples were collected from six littoral zone stations and from two deep locations throughout the lakes. Water temperature profiles were measured at the deep stations at each water-sampling event. Fluridone residues were analyzed using two separate techniques; (1) the newly developed enzyme-linked immunosorbent assay (ELISA), and (2) the standard high-performance liquid chromatography (HPLC) method.

Quantitative sampling of vegetation was performed using point-based frequency of species occurrence to evaluate whole-lake distribution and diversity of the submersed plant community of all eight study lakes. The technique was implemented using global positioning and geographic information systems, with a minimum grid resolution of 50 m by 50 m. Plant surveys were conducted in early to mid-May and in mid-August in 1997 (year of treatment) and 1998 (12 and 15 months posttreatment).

Aqueous fluridone levels on three of the treated lakes met the laboratory-derived criteria for achieving good control of Eurasian watermilfoil by providing a peak concentration of approximately 5 g • L^{-1} during the first 2 weeks of posttreatment, and by maintaining a concentration > 2 g • L^{-1} through 60 DAIT. This fluridone concentration and exposure time (CET) relationship resulted in good to excellent control of Eurasian watermilfoil through 15 months of posttreatment on these lakes. On a fourth lake, however, the required CET relationship was not maintained and poor control of Eurasian watermilfoil was observed. There was no strong evidence of long-term curlyleaf pondweed control in any of the fluridone-treated lakes.

The herbicide application strategy used in this study did not significantly impact the native plant species diversity or cover in the year of treatment, or through 15 months of posttreatment, in any of the fluridone-treated lakes. Native plant cover was maintained at levels > 70 percent in the year of treatment and at 1 year of posttreatment; a level above the range (20 to 40 percent) recommended for healthy fish and wildlife habitat. The selective control of Eurasian watermilfoil achieved in this study verified results from previously conducted laboratory and outdoor mesocosm evaluations.

Fluridone residues became well mixed in the water column under isothermal conditions, and thermal stratification prevented mixing of fluridone into deeper and colder waters. Thermal stratification, or the lack thereof, at the time of herbicide application can impact target concentrations of fluridone. Using the volume of a preselected depth zone to calculate the amount of fluridone needed to achieve a particular target concentration can result in an over- or under-dosing of a water body, leading to poor or nonselective control of Eurasian watermilfoil.

Higher initial and booster rates of fluridone (e.g. 6 g • L^{-1}) could mitigate the loss of product to mixing processes associated with deep thermoclines likely to occur in early spring treatments, if 3.05-m-depth contour is preselected for volume calculations. These slightly higher treatment rates should provide for more consistent control of Eurasian watermilfoil, while still maintaining selective control properties per nontarget native plants.