PROCEEDINGS

1st INTERNATIONAL SYMPOSIUM ON WATERMILFOIL (MYRIOPHYLLUM SPICATUM) AND RELATED HALORAGACEAE SPECIES

JULY 23 and 24, 1985

VANCOUVER, BRITISH COLUMBIA, CANADA

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Although the research described in this document has been funded wholly or in part by the United States Environmental Protection Agency under Assistance Agreement Number R 812651 to the Aquatic Plant Management Society, Inc., it has not been subjected to the Agency's peer and administrative review and therefore may not necessarily reflect the views of the Agency and no official endorsement should be inferred.

INTRODUCTION

The impetus for organizing a symposium on milfoil species came from general discussions among the APMS Board members and officers during 1982 and 1983. Although aquatic plants such as waterhyacinth (Eichhornia crassipes) and hydrilla (Hydrilla verticillata) have received notoriety for their rampant growth and attendant impacts on water use, it was clear that plants in the Myriophyllum genus warranted more attention. The importance of these species, particularly M. spicatum, is evident from their widespread occurrence and from their encroachment in major riparian systems as well as large reservoirs and lakes.

In addition to their general distribution and often detrimental effects on aquatic sites, the milfoil group has been frustratingly difficult for taxonomists to sort out. Indeed, as the reader will soon learn from the papers on this topic, even now there is some disagreement on identification and identifying characteristics. This is an especially important limitation since the proper interpretation of field and laboratory research depends upon knowing what species is involved. With often subtle differences in gross morphology between species, coupled with the morphological variability (plasticity) within a species, our ability to predict or estimate the "weediness" of members of the group is poor.

If this were not sufficient food for discussion and research, we must add to this the environmental concerns of management strategies. One of the least expensive and most selective herbicides, 2,4-D, is quite effective on M. spicatum and even more effective on M. aquaticum, yet the public concerns over its use and its unfortunate confusion with dioxin-related problems have forced major shifts in management toward various cutting, harvesting, rototilling and dredging operations. One might reasonably question the advisability of these types of practices in new infestations and in flowing-water systems since a great deal of substrate disturbance and "seeding" via fragments results.

The Aquatic Plant Management Society with the support of the U.S. Environmental Protection Agency, brought a diverse group of scientists together to focus on some of the problems and questions mentioned here. With the publication of the papers and discussions from this Symposium, the Society hopes to kindle more interest on research on this interesting group of aquatic plants and provide a resource for scientists and aquatic plant managers.

LARS W.J. ANDERSON
Program Chairman and Editor

ACKNOWLEDGMENTS

The First International Myriophyllum Symposium and this Proceedings is a result not only of the individual contributing authors, but also a cadre of people who spent many hours assisting in the planning and conducting of the meeting. The Aquatic Plant Management Society, Inc., gratefully acknowledges the financial assistance of the U.S. Environmental Protection Agency (EPA Assistance Grant No. R 812651 to the Aquatic Plant Management Society, Inc.), the local, public support of the Ministry of Environment Canada, and diligent help with local facilities and extensive U.S. - B.C. liaison,; Bill and Gloria Rushing, who can get the impossible done - from the program to the details of room arrangements and paying bills; Bill Haller for arranging the printing and finishing touches; Max McCowen whose continual support and encouragement was felt by all; Mr. Jim Boland for his assistance in the administration of funds from the U.S. E.P.A.; and especially, Ms. Carol Kearney who patiently and efficiently handled travel arrangements for invited speakers and who assisted in proofing, indexing and the organization of these Proceedings.

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WORLDWIDE DISTRIBUTION AND TAXONOMY OF *MYRIOPHYLLUM* SPECIES

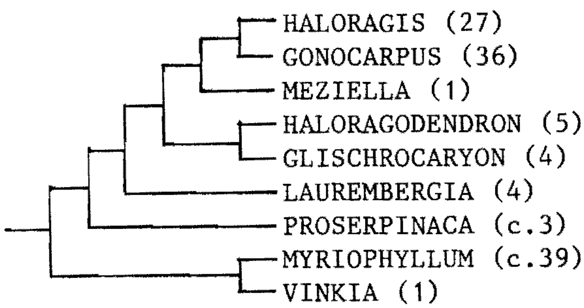
Christopher D.K. Cook
Institut fur systematische Botanik der Universitat Zürich
Zollikerstrasse 107, 8008 Zurich, Switzerland

The composition and phyletic affinities of the Haloragaceae are considered and the position of the genus *Myriophyllum* within the family is discussed. The ecology, floral specializations, fossil history and present distribution of *Myriophyllum* are reviewed.

Myriophyllum belongs to the family Haloragaceae, often misspelled Haloragidaceae, Halorrhagaceae and Halorrhagidaceae (see Bullock, 1959). The family Haloragaceae was originally based almost more on its lack of diagnostic characters than on the presence of any constant feature. During the nineteenth century it became a repository for several genera with simple or reduced floral structures. So by the beginning of this century it was a mixture of discordant elements. Gradually over the last years they have been removed. The following genera, each now the basis of a monotypic family, were once in the Haloragaceae: *Callitriche*, *Ceratophyllum*, *Gunnera*, *Hippuris* and *Trapa*. Rather strangely, the genus *Gunnera* still occasionally appears in the Haloragaceae in, otherwise, very respectable botanical works; the status of *Gunnera* is reviewed by Wagenitz (1975).

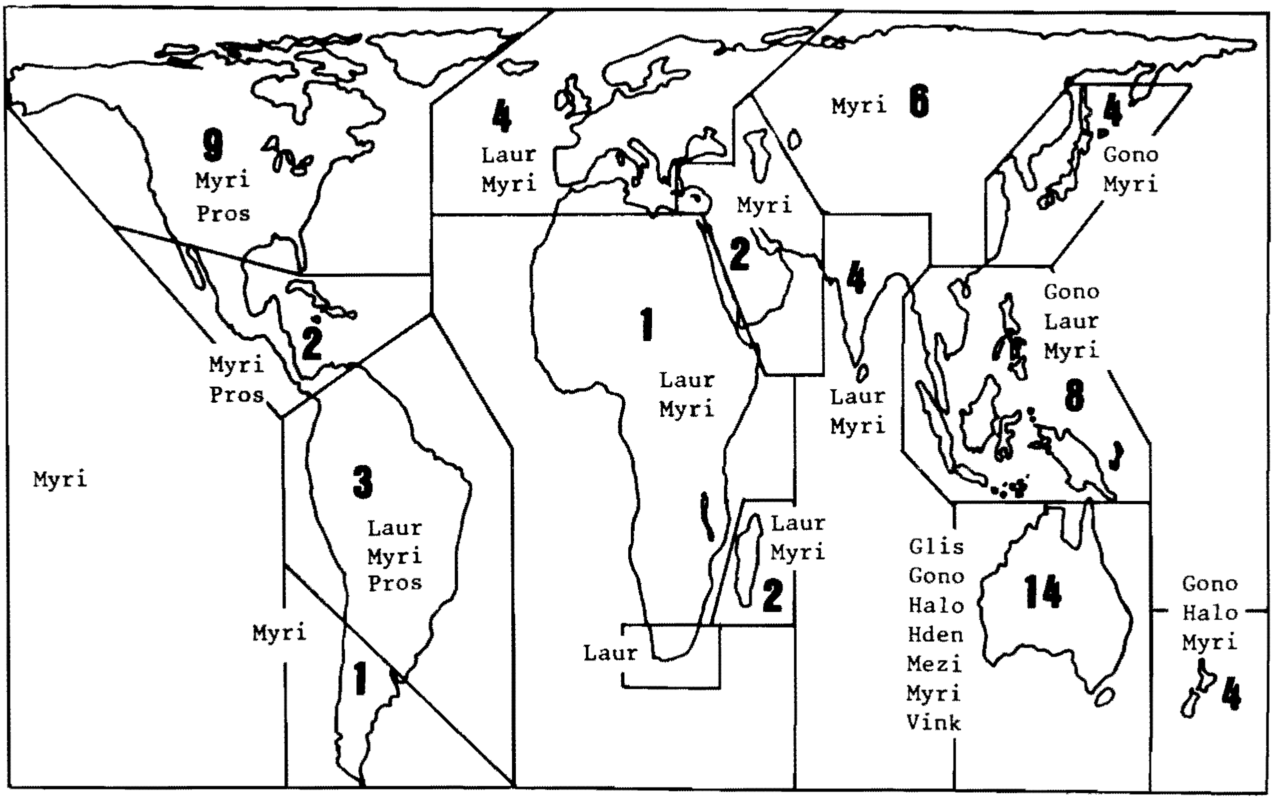
A detailed history of the taxonomic treatments of the family is given by Orchard (1975). The only attempt at a worldwide monograph of the whole family was written by Anton K. Schindler in Engler (1905). This work was based on insufficient material and is also inaccurate - taxonomic botany today would be easier had this work never been published. I have based much of this paper on the work of Orchard (1975, 1980 and 1981) and Meijden (1969).

The family is considered to contain nine genera and about 120 species. The dendrogram below lists the genera in suggested patristic groupings. The number of species in each genus is given in brackets.



With the anomalous genera removed the family is remarkably uniform and as pointed out by Orchard (1975) exhibits very constant floral vascular patterns, wood anatomy, pollen structure and embryology. The phytochemistry and chromosomes are too poorly known to be of value in accessing patristic relationships. Affinities to the Datiscaceae, Rhizophoraceae, Combretaceae, Santalaceae, Araliaceae, Saxifragales and other groups have been suggested but I feel most evidence indicates relationship to the Myrtales but it lacks the bicollateral vascular strands typical of the Myrtales. It is interesting to note in a recent review of the Myrtales by Dahlgren and Thorne (1984) that they disagreed on the affinities of the Haloragaceae; Dahlgren treats it as an order separate from but near to the Myrtales while Thorn prefers to place it in the Cornales.

The region of maximum diversity within the family is certainly Australia, see map 1. Most species are terrestrial somewhat shrubby plants but there are also ephemerals and aquatics. The family is not found in marine habitats and very few species occupy tropical rainforests. Other than some noxious weeds and decorative plants the family is of little economic value. Gonocarpus micranthus is pleasantly fragrant, it is occasionally used but apparently not specially cultivated.



Map 1. Showing the distribution of the genera of the Haloragaceae (Glis=Glischrorcaryon, Gono=Gonocarpus, Halo=Haloragis, Hden=Haloragodendron, Laur=Laurembergia, Mezi=Meziella, Myri=Myriophyllum, Pros=Proserpinaca, Vink=Vinkia) and the number of native species of Myriophyllum (indicated by numbers).

The fossil record of the Haloragaceae is reviewed by Praglowski (1970). Pollen is recorded from the Middle Eocene (c. 50.000.000 years ago) and fruits from the Miocene (c. 20.000.000 years ago). Praglowski (1970) points out that some late Cretaceous "sporomorph Stemma Normapolles" has similarities to pollen of the Haloragaceae.

The Genera

The genera Haloragis, Gonocarpus, Haloragodendron, Glischrocaryon and Meziella form a closely knit group with fruits that are 1- to 4-seeded nuts (not splitting at maturity) and all flowers bear petals. This group is confined to the Old World with Australia as the region of maximum diversity; a few species of Haloragis and Gonocarpus extend beyond Australia reaching New Zealand, South and Southeast Asia (see map 1). Excepting Haloragis brownii (Hooker fil.) Schindler which is an aquatic, the rest of the group are terrestrial plants which occupy a wide variety of habitats from desert to woodland, ranging from small ephemerals to distinctly woody shrubs. Some have conspicuous flowers (particularly Haloragodendron) and may be pollinated by insects but the greater majority of species are pollinated by wind and have large anthers borne on weak filaments and feathery stigmas.

The genus Laurembergia is patristically somewhat isolated. It has been revised by Raynal (1950, 1967); she has reduced the 18 species recognised by Schindler in Engler (1905) to four. It is found in both the Old and New Worlds (map 1) extending from tropical S. America through Africa to S.E. Asia but extraordinarily it is absent from Australasia. It is the most tropical of all genera of the Haloragaceae. It is subaquatic with divided submerged leaves and unisexual flowers, features typical for Myriophyllum but the inflorescence structure and floral features indicate affinities to Haloragis and Gonocarpus.

The genus Proserpinaca is placed somewhat away from the Haloragis group. It is the only exclusively New World genus in the family; it is found in eastern America from Nova Scotia southwards to Brazil (map 1). However, fossil fruits have been recorded from the Tertiary of Europe and Siberia which indicates a wider distribution in the past.

Superficially Proserpinaca resembles Myriophyllum with pinnately divided submerged leaves but its rhizome is somewhat woody and most of the floral characters are typical for the Haloragis group: flowers often borne in dichasia of 3 or 5 in the axils of alternate primary bracts, the flowers usually have petals (often rudimentary and caducous), and the fruit is a 3-sided, 1-seed nut. The flowers are trimerous, a feature otherwise found only in isolated species of Haloragis and Gonocarpus. There is a tendancy to the development of unisexual flowers, a feature otherwise typical for Laurembergia and Myriophyllum. The genus Proserpinaca is, rather surprisingly, poorly investigated but, inspite of this, I would place it somewhere between the Haloragis group and Myriophyllum.

The genus Myriophyllum

Myriophyllum and the very closely related genus Vinkia (recently described by Meijden, 1975 from N. Australia) are phyletically somewhat remote from the rest of the family and are placed in the tribe Myriophylleae. It is the most widespread group within the family being almost cosmopolitan. The distribution with an indication of the species density is given on map 1 and the detailed distribution in table 1.

MYRIOPHYLLUM	1	2a	2b	2c	3	4a	4b	4c	5a	5b	5c	6a	6b	6c	7a	7b
robustum Hooker fil.	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	N
triphyllum Orchard	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	N
amphibium Labill.[1]	-	-	-	-	-	-	-	-	-	-	-	-	-	-	N	-
caput-medusae Orchard	-	-	-	-	-	-	-	-	-	-	-	-	-	-	N	-
drumondii Bentham[2]	-	-	-	-	-	-	-	-	-	-	-	-	-	-	N	-
filiforme Bentham	-	-	-	-	-	-	-	-	-	-	-	-	-	-	N	-
latifolium Mueller	-	-	-	-	-	-	-	-	-	-	-	-	-	-	N	-
muelleri Sonder	-	-	-	-	-	-	-	-	-	-	-	-	-	-	N	-
porcatum Orchard	-	-	-	-	-	-	-	-	-	-	-	-	-	-	N	-
salsugineum Orchard	-	-	-	-	-	-	-	-	-	-	-	-	-	-	N	-
trachycarpum Mueller[3]	-	-	-	-	-	-	-	-	-	-	-	-	-	-	N	-
verrucosum J. Lindlay	I	-	-	-	-	-	-	-	-	-	-	-	-	-	N	-
pedunculatum Hooker fil.[4]	-	-	-	-	-	-	-	-	-	-	-	-	N		N	N
propinquum A. Cunn.	-	-	N	N	-	-	-	-	-	-	-	-			N	N
dicoccum Mueller	-	-	-	-	-	-	-	-	-	-	-	N	N	-	N	-
bonii Tardieu Blot	-	-	-	-	-	-	-	-	-	-	-	-	N	-	-	-
coronatum Meijden	-	-	-	-	-	-	-	-	-	-	-	-	N	-	-	-
pygmaeum Mattf.	-	-	-	-	-	-	-	-	-	-	-	-	N	-	-	-
siamense(Craib)Tardieu Blot	-	-	-	-	-	-	-	-	-	-	-	-	N	-	-	-
tuberculatum Roxb.	-	-	-	-	-	-	-	-	-	-	-	N	N		N	-
tetrandrum Roxb.	-	-	N	-	-	-	-	-	-	-	-	N	N	-	-	-
oliganthum (Wt.& Arn.) Mueller	-	-	-	-	-	-	-	-	-	-	-	N	-	-	-	-
axilliflorum Baker	-	-	-	-	-	-	-	-	-	-	N	-	-	-	-	-
mezianum Schindler	-	-	-	-	-	-	-	-	-	-	N	-	-	-	-	-
indicum Willd.[5]	-	-	-	-	-	-	-	-	N	-	-	-	-	-	-	-
aquaticum (Vell.) Verdc.[6]	I			I	I	I	N			I			I	I	I	I
mattogrossensis Hoehne	-	-	-	-	-	-	N	-	-	-	-	-	-	-	-	-
quitense Humb., Bonpl.& Knuth	-	-	-	-		N	N	N	-	-	-	-	-	-	-	-
pinnatum (Walt.) Britton[7]	-	-	-	-	N	N	-	-	-	-	-	-	-	-	-	-
farwellii Morong	-	-	-	-	N	-	-	-	-	-	-	-	-	-	-	-
heterophyllum Michaux	I	-	-	-	N		-	-	-	-	-	-	-	-	-	-
humile (Raf.) Morong	-	-	-	-	N	-	-	-	-	-	-	-	-	-	-	-
laxum Schuttlw. ex Chapm.	-	-	-	-	N	-	-	-	-	-	-	-	-	-	-	-
tenellum Bigelow	-	-	-	-	N	-	-	-	-	-	-	-	-	-	-	-
spicatum L.	N	N	N	N	B	-	-	-	-		-	-	-	-	-	I
alternifolium DC.	N	-	-	-	N	-	-	-	-	-	-	-	-	-	-	-
ussuriense (Regel) Maxim.[8]	-	-	N	N		-	-	-	-	-	-	-	-	-	-	-
verticillatum L	N	N	N	N	N	-	-	-	-	-	-	-	-	-	-	-
exalbescens Fernald	N	-	N		N	-	-	-	-	-	-	-	-	-	-	-

Table 1. Showing the species of Myriophyllum and their distribution (for details see text).

In table 1 I have attempted to list all the species of *Myriophyllum* and indicate their natural and introduced distribution. There is no reliable and complete taxonomic revision of *Myriophyllum*. The list has been drawn up largely from the works of Meijden (1969) and Orchard (1980, 1981). Nevertheless, there are still numerous taxonomic difficulties: 1, perhaps conspecific with *M. pedunculatum*; 2, including *M. glomeratum* Schindler and *M. integrifolium* (Hooker fil.) Hooker fil.: 3, including *M. gracile* Bentham; 4, including *M. longibracteolatum* Schindler, *M. tillaeoides* Diels and *M. votschii* Schindler; 5, including *M. intermedium* DC.; 6, including *M. brasiliense* Cambess and *M. proserpacoides* Gill.; 7, including *M. hippuroides* Nuttall, *M. scabrum* Cham. and Schlecht. and *M. sparsiflorum* Wright; 8, perhaps conspecific with *M. propinquum*. The limits of the geographical regions in this table are the same as those adopted by Cook (1985) and are indicated on map 1: 1, Europe and N. Africa; 2a, Asia Minor; 2b, C. Asia; 2c, E. Asia; 3, N. America; 4a, C. America; 4b, tropical S. America; 4c, temperate S. America; 5a, tropical Africa; 5b, S. Africa; 5c, Madagascar; 6a India; 6b, SE. Asia; 6c, Pacific; 7a, Australia; 7b, New Zealand. The symbol "N" indicates native; "I" introduced; "B" both introduced and native; "-" not present.

Unfortunately, there is no satisfactory classification of the genus *Myriophyllum* on a worldwide basis. Like so many other aquatic or amphibious plants they are phenotypically very plastic so that many taxonomic problems can best be solved after considerable field experience and, if possible, cultivation experiments. Another problem is that many species are vegetatively very alike and almost impossible to identify without flowers or ripe fruits, and ripe fruits are not common. As an example of a taxonomic mistake I myself failed to recognise *M. exalbescens* when preparing an account of *Myriophyllum* for Flora Europaea (Tutin & al., 1968) but today I am convinced that Aiken and McNeill (1980) and Faegri (1982) are correct and that it is native in Europe.

The genus is essentially aquatic and most, if not all, species can assimilate under water. However, the degree of 'aquaticness' varies from species to species. Some such as *M. alterniflorum* and *M. verticillatum* are essentially submerged species, the vegetative organs remain submerged throughout the year; in summer they can survive on wet land in the absence of strong competition but as terrestrial plants they rarely flower and, if so, very rarely succeed in developing ripe fruits. Most species, however, are essentially amphibious and usually develop some emergent leaves during the flowering and fruiting phase. Some have the majority of their assimilating tissue underwater while others such as *M. aquaticum* usually have most emergent. A few species such as *M. coronatum* and *M. pygmaeum* are essentially terrestrial plants which tolerate periods of submergence.

A feature that separates *Myriophyllum* from most of the Haloragaceae is the clear trend to the development of unisexual flowers. Very few, if any, species have exclusively bisexual flowers. Normally, at least the uppermost flowers of the inflorescence are male and sometimes the lowermost female. There is a trend to have exclusively unisexual flowers with males above the females on the same simple, spike-like inflorescence. In a few species, such as *M.*

aquaticum the trend is complete and the plants are dioecious. It is interesting that to date only female plants of M. aquaticum have been recorded outside its native range. Many other phyletically unrelated groups of aquatic plants have evolved unisexual flowers. It is likely that this evolutionary convergence is coupled more with abiotic pollination mechanisms than an indication of an adaptation to the aquatic environment. Myriophyllum is clearly wind pollinated.

Australia is the region richest in species followed by N. America, SE. Asia and Siberia. Compared to most other relatively species-rich genera of aquatic plants Myriophyllum is unusual as it has very few naturally widespread species, the majority of the species are relatively local endemics (see table 1). Among the 39 accepted species it is also remarkable that very few have become widely established outside their native ranges. A notable exception is M. aquaticum which is native in S. America but today naturalised in all continents. Myriophyllum heterophyllum a N. American species has become naturalised in Austria, England and Switzerland but does not look like becoming a pest. Myriophyllum verrucosum a native of Australia has been recorded in England but has not become naturalised. Myriophyllum spicatum which will be discussed in detail in later papers is particularly interesting as it is apparently native in Greenland (phytogeographically N. America) but has recently become a pest in parts of Canada and the USA. Most aquatic and amphibious species of Myriophyllum do not appear to be very specialized for particular habitats and virtually all species should be considered as potentially dangerous weeds. Trade and traffic in all species of Myriophyllum should be very carefully controlled.

Note added after submission of manuscript.

According to Susan Aiken (pers.comm.) Myriophyllum spicatum does not occur in Greenland. Myriophyllum farwellii is a totally submerged species. According to A. and O. Ceska (pers.comm.) M.ussuriense is distinct from M. quitense and present in N. America. The number of species in N. America (map 1) should be raised to 10 or 11.

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MYRIOPHYLLUM SPICATUM IN NORTH AMERICA

Richard Couch & Edward Nelson
Natural Sciences Department
Oral Roberts University
Tulsa, Oklahoma 74171

A search of 173 North American herbaria yielded 619 Myriophyllum spicatum L. specimens. Data from these specimen labels were combined with literature and personal communication information to map the current, known distribution of M. spicatum populations in North America. The first M. spicatum was colected October 29, 1942, in Belch Spring Pond in Washington, D. C. In 1985, 392 M. spicatum populations were verified to be present in 33 states plus the District of Columbia, and in three Canadian provinces.

Taxonomy

Myriophyllum spicatum L. is a rooted, submersed, perennial aquatic herb native to Europe, Asia, and northern Africa. The stems are long with feathery-like leaves attached along the entire stem in whorls of threes to fives and up to 35 mm long. Leaves typically have 14-21 slender, linear divisions. The flowering spikes (inflorescences) are terminal and above water level, resubmerging after the setting of fruit. Watermilfoil reproduces most effectively by way of vegetative propagation through fragmentation, rhizomes, and axillary buds (1-5).

Eurasian watermilfoil, the common name for Myriophyllum spicatum, belongs to the order Hippuridales, the family Haloragaceae R.Br.; i.e. the watermilfoil family. There are two genera of aquatic or paludal plants in this family: Proserpinaca (mermaid-weeds) and Myriophyllum. Aiken (4,5) determined there were 13 species of Myriophyllum in North America. Ceska (6) found a fourteenth species, M. ussuriense, in British Columbia, Canada. Correll and Correll (2) say there are about 45 species of Myriophyllum worldwide.

Myriophyllum spicatum was first described by Linnaeus in 1753 (4,5,7). In 1919, Fernald (8) determined an American watermilfoil differed sufficiently from the Eurasian type to be considered a new species. He named this new species Myriophyllum exalbescens Fern. He listed specimens of M. exalbescens dating back to 1865 from numerous locations across the northern United States.

Jepson (9), Hulten (10), Patten (11,12), Nichols (13), Brooks and Hauser (14), and Orchard (15) considered the differences between M. spicatum and M. exalbescens too insignificant to warrant taxa separation. Both Hulten (10) and Patten (11,12) endorsed a subspecies ranking for M. exalbescens; i.e. M. spicatum L., subsp. exalbescens (Fern) Hulten whereas Jepson (9), Nichols (13), Brooks and Hauser (14), and Orchard (15) preferred making M. exalbescens a variety of M. spicatum; i.e. M. spicatum L., var. exalbescens (Fern) Jepson. Fernald steadfastly opposed considering M. spicatum and M. exalbescens one species, but did express some willingness (16) to endorse making M. exalbescens a variety of M. spicatum.

Others, Love (17), Reed (18), Aiken (4,5,7,19) also addressed the problem of taxa distinction. They came to the conclusion that M. spicatum and the native American species M. exalbescens and M. verticillatum L. should be separate taxa based upon morphological differences, distributional patterns, and physiological differences such as the vernalization necessary for completion of *M. exalbescens* and *M. verticillatum* life cycles.

Reed (18), Aiken and Walz (20), and Aiken (4,7) said *M. exalbescens* and *M. verticillatum* are distributed only in the northern portion of North America. Reed's (18) line corresponded to Aiken and Walz's (20) 0° C January isotherm for *M. exalbescens*. Cold weather is required for vernalization and successful turion formation by *M. exalbescens* and *M. verticillatum* (20-24). This line of demarcation confines *M. exalbescens* and *M. verticillatum* north of a line extending from the Washington, D. C. area westward to the Ohio River through southern Missouri along the Kansas-Oklahoma border across northern New Mexico on to the California-Mexico border near San Diego.

Background

In 1978, Brian Welch (25) undertook a study of the distribution of *M. spicatum* in the continental United States by soliciting information from the major herbaria of the United States. He wrote 117 herbaria receiving replies from 91. The major conclusion of the study was there existed too much taxonomic confusion within the genus *Myriophyllum* for a precise distributional pattern to be determined without annotation.

Then came results of the excellent research of Susan Aiken in the late 1970's (4,5,7,19,20). Utilizing this work as our basis, we initiated this study with the following objectives: 1) annotate *Myriophyllum* collections of the major North American herbaria based upon the taxonomy of Aiken (4,5); 2) precisely distinguish *M. spicatum* from other aquatic plants, particularily *M. exalbescens* and *M. verticillatum*; 3) update information concerning the distribution of *M. spicatum* in the United States and Canada.

Methods

We borrowed collections of *Myriophyllum* spp. from leading authorities of *Myriophyllum* taxonomy in North America for study and photographing for future reference. We visited 44 herbaria in 16 states including the New York Botanical Garden and the University of Minnesota herbaria; the latter being where Susan Aiken carried out her doctoral research. We wrote more than 200 herbaria in North America asking for loans of their *Myriophyllum* collections.

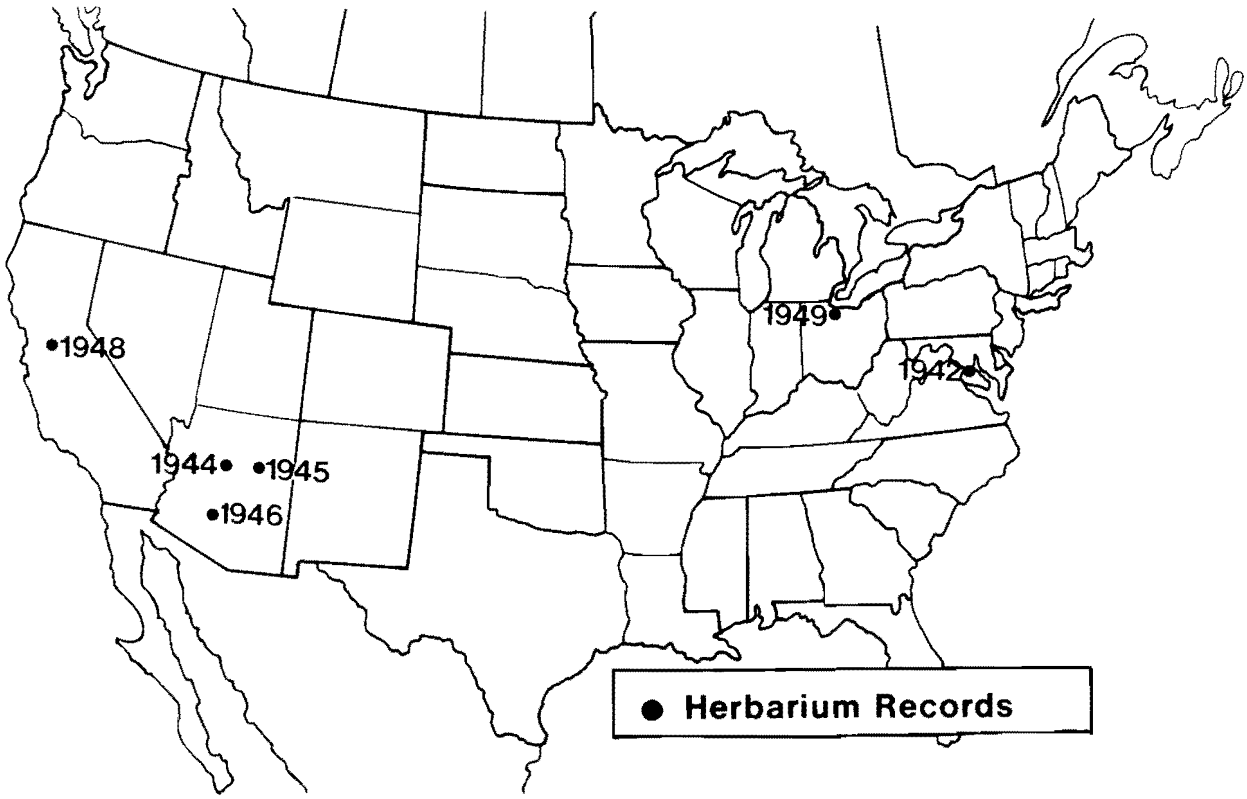
Using the appropriate keys (2,5,26) and the voucher specimen photographs, we soon became proficient at annotating the 13 species of North American *Myriophyllums* described by Aiken (5).

Results and Discussion

We have annotated about 15,000 specimens of *Myriophyllum* and have photographed about 11,000. We ascertained about 20% of all specimens not previously annotated by Susan Aiken were not correctly identified. Based upon the taxonomy refined by Aiken (5), we have established a taxonomic uniformity in the identity of those *Myriophyllum* specimens we have worked.

Introduction to North America. It is now generally conceded that *M. spicatum* is a species introduced into North America from Europe, but there are few clues to specify the exact time, place, and method of introduction. Reed (18) claimed the species was introduced in the late 1800's citing as evidence specimens he personally annotated from the Langlois Herbarium of Catholic University in Washington, D. C. Unfortunately, these specimens are not available at this time because the Langlois Herbarium is in the process of being sold. Dr. Reed is now retired and ignores correspondence. It is our opinion that if and when these specimens are made available for our inspection, we will find all of them to be M. exalbescens.

Figure 1. The introduction of Myriophyllum spicatum into North America.



Our annotation work revealed the earliest M. spicatum collected in North America came from Belch Spring Pond on October 29, 1942. Belch Spring Pond is in the District of Columbia (Figure 1). Other collection made in the 40's occurred in widely scattered sites. M. spicatum was collected at two sites near Winslow, Arizona, on the same day (September 10th) in 1945. The next year, 1946, it was collected in Encanto Park, in Phoenix, Arizona. Two years later, it was collected in Trapper's Slough of Robert's Island in San Joaquin County, California. And in 1949 at Middle Harbor of Put-in-Bay of western Lake Erie, Sandusky County, Ohio.

How was M. spicatum introduced into North America? How did it migrate to so many distant points so rapidly? No one knows, but conjecture is possible. It is commonly acknowledged that the Plant Introduction Branch of USDA has introduced hundreds of species of plants into the United States including most of our commercially important crop plants. Although we found no such notes on old M. spicatum sheets, we did find numerous reference notes on old M. aquaticum sheets from the Washington, D. C. area saying such things as "growing in greenhouse culture", "growing in pool in front of Interior Building in Washington", "appears to have escaped from cultivation", etc. The mere fact that the first confirmed population of M. spicatum growing in North America was found in the District of Columbia is fair circumstantial evidence for government officials being involved in its introduction.

A second likely source of introduction for such a species as M. spicatum involves the aquarium trade. Aquarium dealers have long advocated the utilization of M. spicatum as an attractive, useful aquarium plant (27). The dumping of an aquarium containing M. spicatum into Watts Bar Lake in east Tennessee has been ascribed the source for the introduction of M. spicatum into Tennessee Valley Authority reservoirs (28). And I remember visiting a site in Florida where it was alleged several species of widely utilized aquarium-valuable plant species were deliberately planted for harvesting as needed for the aquarium trade in that area.

Spread. Fishermen, inadvertently and sometimes deliberately, spread many aquatic species, especially those that propagate vegetatively as does M. spicatum. In moderate amounts, watermilfoil beds tend to improve fishing in a lake. Because of this, fishermen sometimes transplant watermilfoil into all their favorite fishing lakes.

Migrating waterfowl no doubt also help spread aquatic species along their flight paths. Even worm farmers have been implicated! The concession stand manager at Comanche City Lake near Comanche, Oklahoma, told us of worm farmers coming to Comanche City Lake to harvest the watermilfoil to use in their worm beds, and for packing in their worm boxes for sale to the public. Once the worms have been used, quite often the box and contents of viable watermilfoil sprigs are dumped into the body of water being fished. This could account for the large number of ponds and lakes in south-central Oklahoma having M. spicatum infestations.

Figure 2 shows the distribution of M. spicatum in the 1950's. Populations became established in the San Francisco area of California, in Hays and Travis counties of central Texas; in Comanche county of Oklahoma; in Watts Bar Lake near Knoxville, Tennessee; in several locations in eastern Ohio, western Pennsylvania, and New Jersey; and in upstate New York.

By the 1960's and 70's (Figures 3 and 4), M. spicatum populations had begun being documented in the literature as weed problems; e.g. in the Chesapeake Bay in Maryland (29-35); in the Tennessee River Valley of Tennessee and Alabama (28,36-41); in Virginia (42); in New Jersey (43-44); in Louisiana (45-46); in Florida (47-48); in North Carolina (49); in New York (18,50); in Michigan (51); in Wisconsin (13,52); in Oklahoma (53-56); in Washington state (57); in Arizona (58), in British Columbia (59); and in Ontario (60).

Figure 2. Myriophyllum spicatum in North America, 1950's.

Figure 3. Myriophyllum spicatum in North America, 1960's.

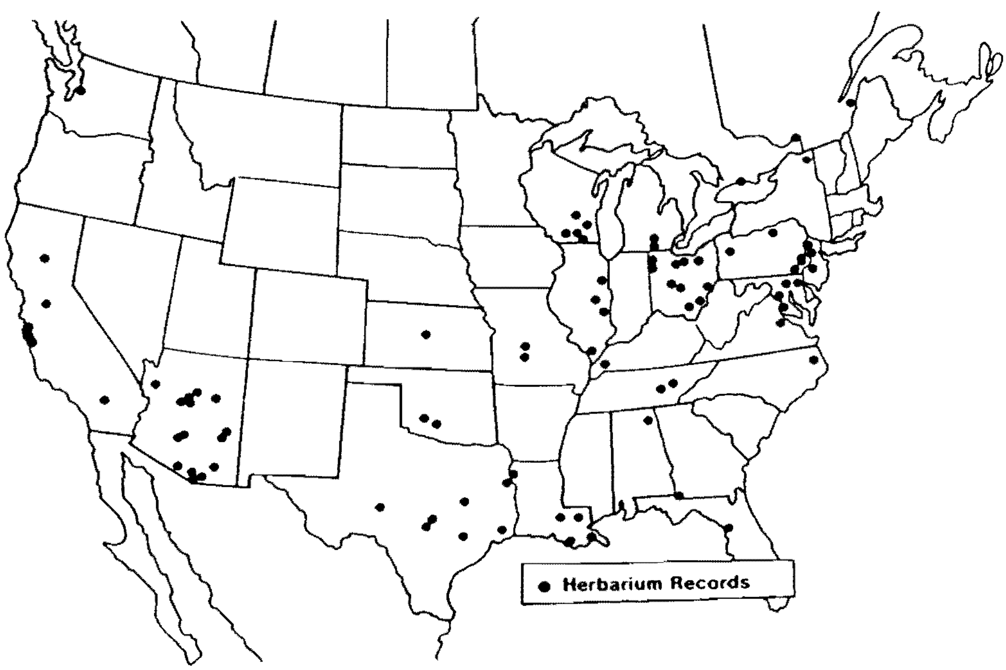


Figure 4: Myriophyllum spicatum in North America, 1970's

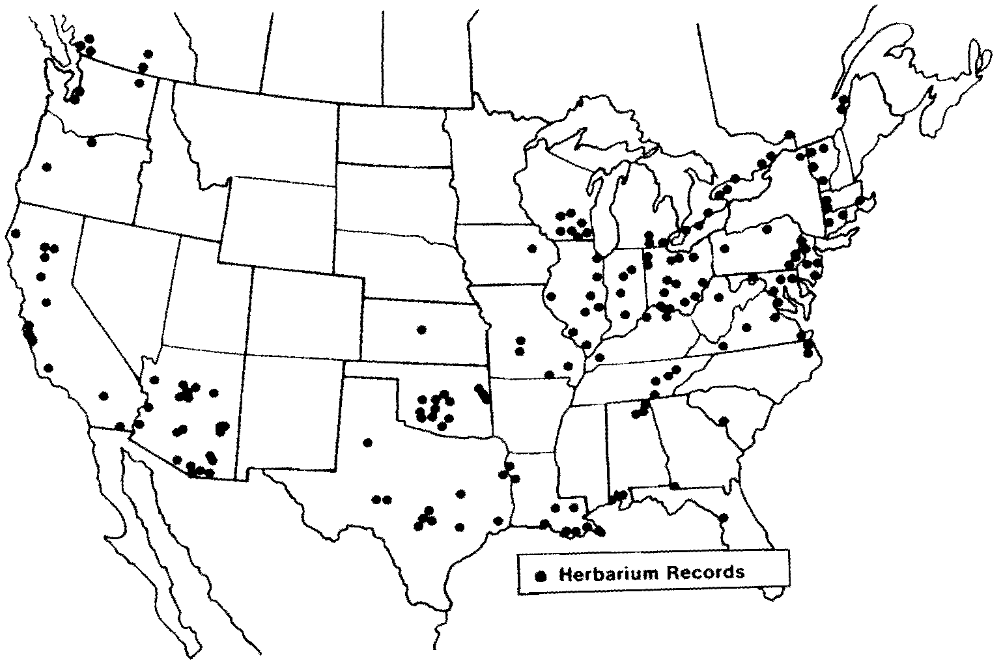
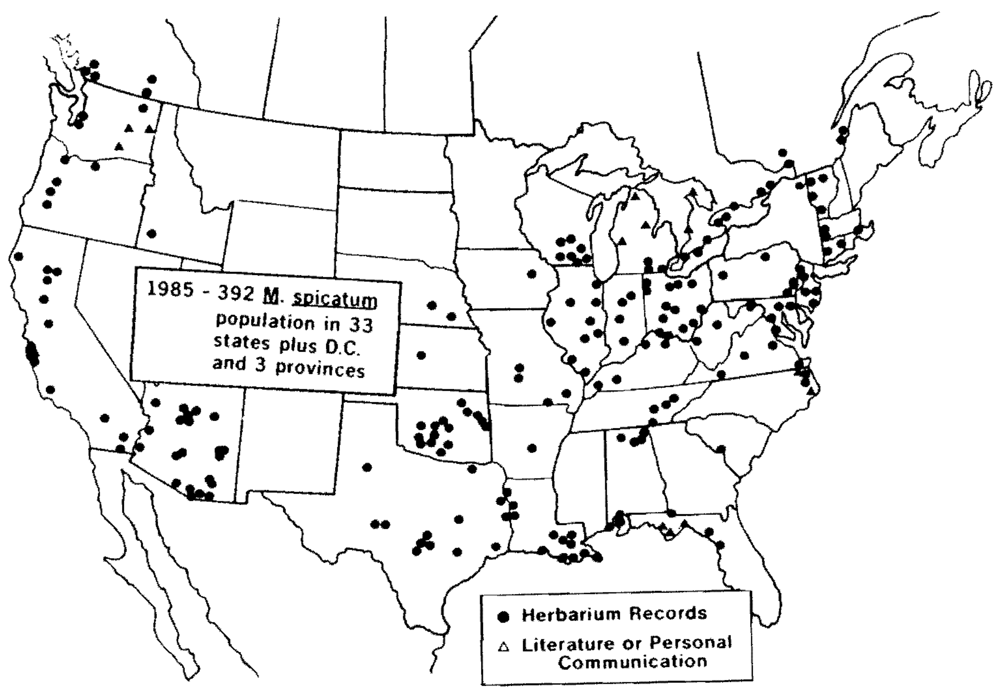


Figure 5: Myriophyllum spicatum in North America, 1980's



Present Distribution. We have verified the existence of 392 M. spicatum populations located in 33 states plus the District of Columbia, and in three Canadian provinces (Figure 5). No doubt there are more populations than these because there has been less collecting activity in recent years plus the fact that collecting traditionally lags behind a few years the actual spread of a rapidly moving species like M. spicatum.

How many sites will eventually be populated with M. spicatum is unknown. The distribution of M. spicatum in Europe and Asia provides clues, but cannot accurately predict what will eventually happen in North America.

Most of the M. spicatum populations in North America have, or are, causing weed problems of varying degrees in the ecosystems in which they are located. Without a doubt, M. spicatum is a more competitive and agressive taxa of watermilfoil as an introduced species into North America. But environmental conditions and stage of eutrophication of any water body also seem critical for the manifestation of weediness; i.e. the explosive growth (bloom) of any plant species.

The ecological principles of eutrophication (61-70) can account for the sequence of events surrounding the introduction, spread, and weed problems caused by Myriophyllum spicatum in North America. What Cronin (71) said about the Chesapeake Bay area is true for the entire continent; i.e. "that man is influencing more and more aquatic ecosystems via the introduction of large amounts of nutrients and silts into practically all water bodies in North America". And with adequate or surplus fertility, plant life of some kind will flourish -- native or exotic, algal or macrophytic, or more likely, combinations, with the most aggressive and competitive species dominating in the ecosystem.

Acknowledgments

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HISTORY OF THE INTRODUCTION AND DISTRIBUTION OF MYRIOPHYLLUM AQUATICUM IN NORTH AMERICA

Edward N. Nelson and Richard W. Couch
Natural Sciences Department
Oral Roberts University
Tulsa, Oklahoma 74171

An examination of almost 15,000 specimens of *Myriophyllum* from 173 herbaria in North America yielded 1,360 *Myriophyllum aquaticum* specimens. Data from these labels were combined with literature and personal communication information to map the currently known distribution of *M. aquaticum* in North America. The earliest specimen of *M. aquaticum* was collected April 20, 1890, from a Haddonfield, New Jersey, millpond. By 1985, 291 populations had been verified as having existed in 29 of the United States, in British Columbia, and in two states of Mexico.

Taxonomy and Natural History

Myriophyllum aquaticum (Vellozo) Verdcourt, commonly called Parrot's Feather, is a pallid grey-green, partially emersed, rooted aquatic usually erect, but sometimes trailing on mud or seepage areas. The stems are mostly simple, rarely branched. Emergent leaves are all whorled, rather stiff, oblong in outline, and 2.5-3.5 cm long. Each leaf will have 18-36 sparsely puberulent (when young), pinnately divided, pectinate linear-filiform segments. Only pistillate flowering plants are known to occur in North America. The flowers are borne in the axils of both emersed and submersed leaves (1). The pistillate flowers are about 1.5 mm long, made conspicuous by a tuft of pinkish plumose stigma lobes. Fruits never form due to lack of fertilization by staminate plants. Therefore, reproduction by *M. aquaticum* in North America is exclusively vegetative.

Endemic Distribution

Fernald (2) states that Parrot's Feather is native to Brazil. The synonym for *M. aquaticum* is *Myriophyllum braziliense* Cambess. (3). This name is firmly embedded in taxonomic literature (4,5). Most floras state simply that it is a native of South America. It is an obviously attractive plant that several authors (1,2,5,6,7) contend has been introduced into all continents worldwide by aquarists.

Methods

We personally visited visited 44 herbaria located in 16 states. In addition, we requested loans from more than 200 other herbaria recommended to us by leading aquatic plant biologists. By use of appropriate keys (1,2,5,6,7,8,9) and voucher specimens loaned to us by *Myriophyllum* authorities, we soon became proficient at identifying the species of North American *Myriophyllum* described by Aiken (1,5). We have closely examined and annotated almost 15,000 sheets of the 13 species of North American *Myriophyllum*. After annotation, each sheet was photographed for future reference and compilation into distribution maps and historical sequence. We found 15-20% of sheets not annotated previously by Aiken were not properly identified.

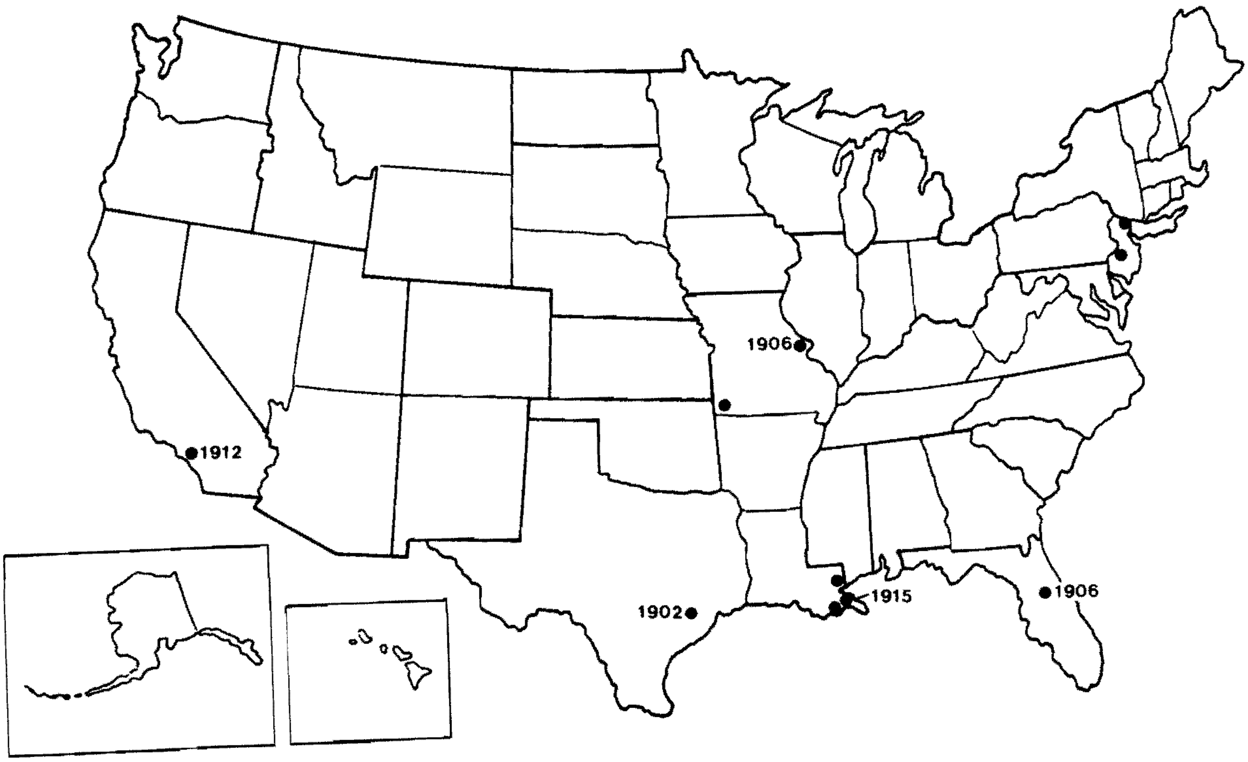
Distribution in North America

1890-1899. The earliest specimen we recorded among the 1,360 herbarium sheets of *M. aquaticum* we studied was a sheet from Cornell University Herbarium dated April 20, 1890, from Harris' Millpond in Haddonville, New Jersey. Another came from a population found near Joplin, Missouri, in 1897. A number of the collections from this decade bore the notation "cultivated". Thus it appears likely that this species was introduced as an ornamental during the late 1800's (Figure 1).

Figure 1. *Myriophyllum aquaticum* in North America, 1890-1899.

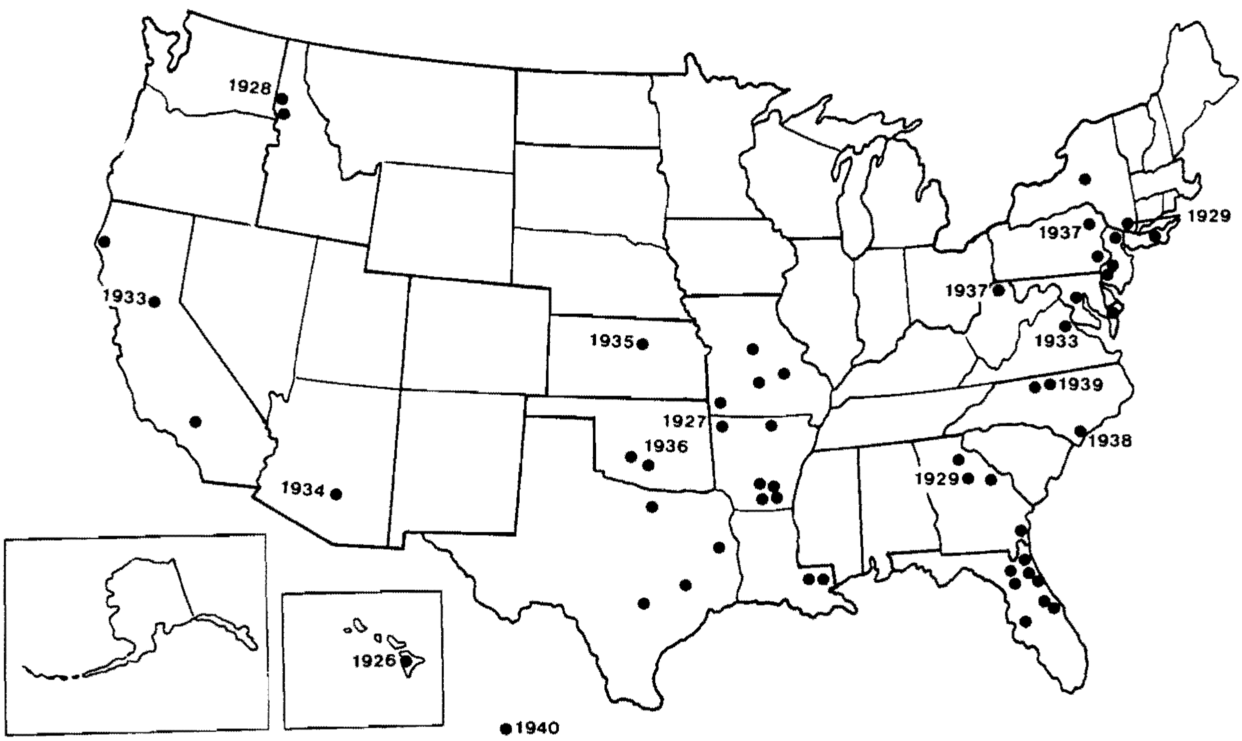
1900-1920. The New Jersey populations were still extant between 1900 and 1920. In 1902, the plant was collected in the vicinity of Houston, Texas. Additional populations were sampled in Missouri and Florida in 1906. By 1912, M. aquaticum was found in California (Figure 2).

Figure 2. Myriophyllum aquaticum in North America, 1900-1920.



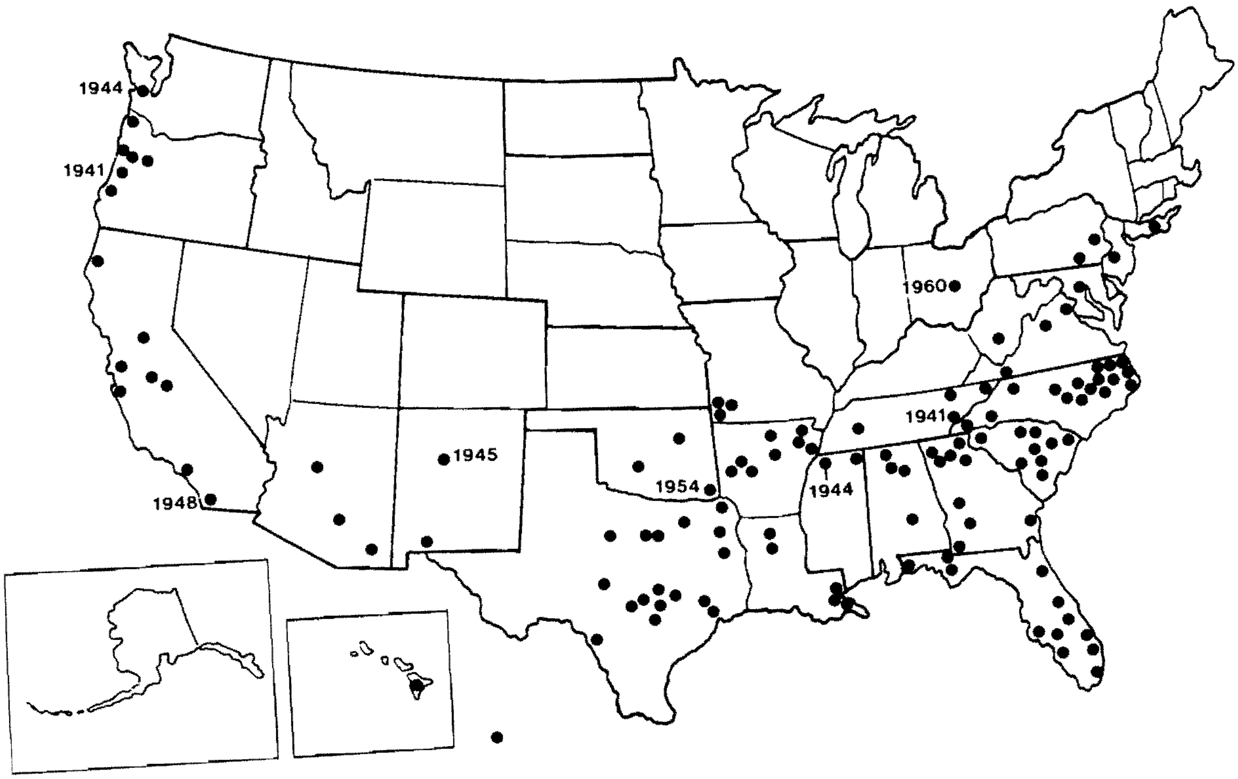
1921-1940. During these two decades, records indicated that the plant had been introduced into 12 additional states. On the east coast, New York, Pennsylvania, Maryland, West Virginia, North Carolina, and Georgia had populations. Additional habitats were occupied by M. aquaticum in the Gulf Coastal Plains states of Arkansas and Louisiana; in the Southern Great Plains states of Oklahoma and Kansas; and in the western states of Arizona, California, and Idaho. There are also records of collections made in Hawaii and Mexico for the first time during this period (Figure 3).

Figure 3. Myriophyllum aquaticum in North America, 1921-1940.



1941-1960. In the course of these twenty years, the species was collected for the first time in Ohio, Tennessee, and South Carolina in the east. Along the Gulf Coast, Alabama and Mississippi were added to its distribution. And in the west, plants had become established in scattered populations in New Mexico, Oregon, and Washington. Meanwhile, the species continued to expand within its range in a number of the southern tier of states; notably North Carolina, South Carolina, Georgia, Florida, Arkansas, and Texas. The Hawaiian population was still extant (Figure 4).

Figure 4. Myriophyllum aquaticum in North America, 1941-1960.

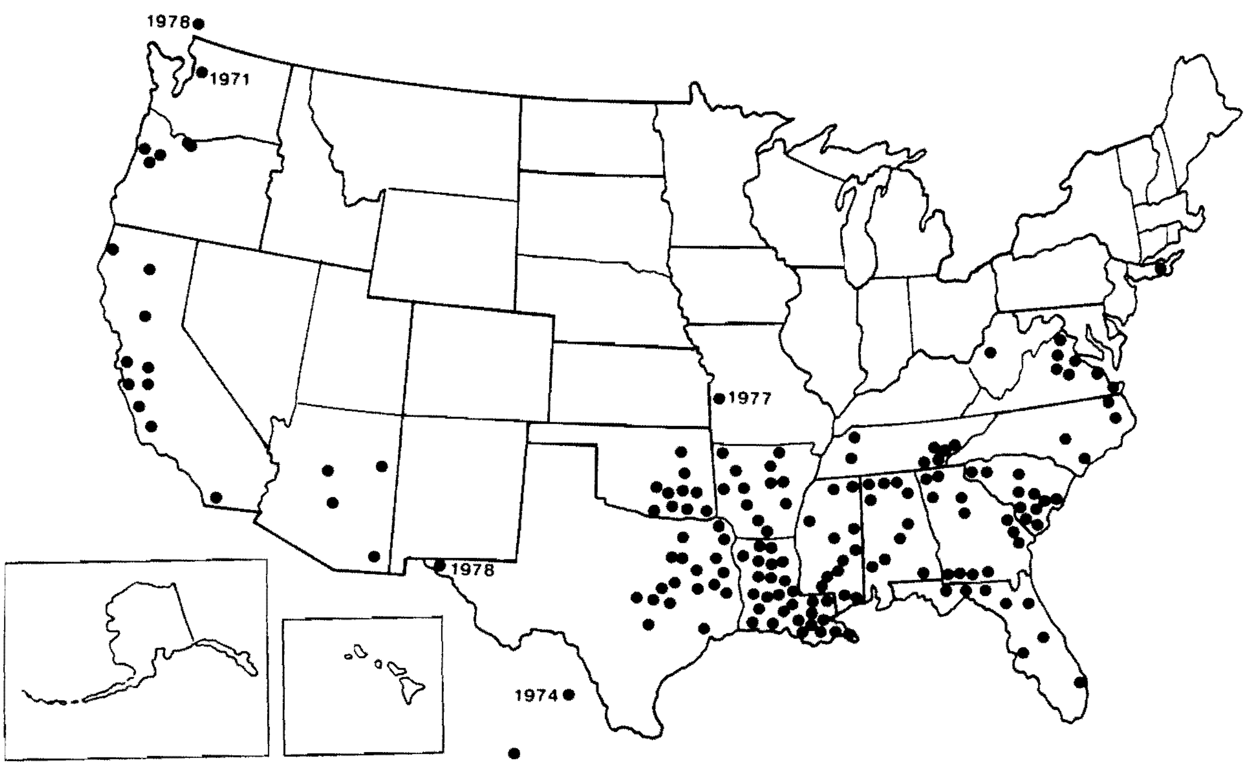


1961-1980. These two decades are notable for the apparent decline in the rate of expansion of its range. Only two new records were found, one in the Mexican state of Nuevo Leon, and the other in British Columbia. But the explosive proliferation of the species in the Gulf Coastal Plains was substantial (Figure 5). M. aquaticum expanded westward in Texas and slightly northward in Missouri. The 1971 record for Washington was reported as being a first for the State (10), but our findings show that it was in Washington 27 years earlier.

Discussion

Only the pistillate form of the diecious M. aquaticum was introduced into, and thrives in North America today. This suggests that all the populations in North America could very well be from a single clone established by a single introduction in the late 1800's on the east coast of the United States.

Figure 5. *Myriophyllum aquaticum* in North America, 1961-1980.



Likewise, the numerous herbarium label notes on the earliest collections such as "cultivated", "growing in a greenhouse", and "cultivated in a Department of Interior fountain" lend credence to it being a late 1800's introduction.

The neotropical origin of *M. aquaticum* is clearly reflected in its North American distribution. We noticed sporadic introductions into the northern temperate zone, but such populations did not persist. On the other hand, populations that became established in the subtropical parts of the continent persisted and continue to expand their range (Figures 1-5).

M. aquaticum has never been reported as a weed problem in North America. However, Jacot-Guillarmod (11) listed this exotic aquatic as one of three economically important aquatic weeds in South Africa. The other two were *Eichhornia crassipes*, the water hyacinth, and *Salvinia molesta*, the water fern.

Jacot-Guillarmod (11) speculated about the possibility that *M. aquaticum* had hybridized with the indigenous species of *Myriophyllum*. He looked for evidence of such hybridization and found none. This evidently has not happened in North America either. *M. aquaticum* appears not to have the plasticity of phenotype (1,5) that characterizes the other species of *Myriophyllum*.

Conclusion

Myriophyllum aquaticum has been among the flora of North America for almost 100 years now. Although it continues to slowly expand its range, and the number of habitats it occupies within its range, it has not been cited as a weed problem. Could this be a case of an exotic intruder from the tropics "behaving itself" -- or is it a case of the exacting environmental conditions conducive to a "weed bloom" not yet existing? The evidence to this point in time favors the conclusion that *M. aquaticum* is a successful aquatic plant introduction into North America.

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Taxonomy and Distribution

A recent revision of *Myriophyllum* in Australia is in press (Orchard pers. comm.). Prior to this and in preparation for it, Orchard (1,4) revised the genus in New Zealand and South America with some reference to Australian and North American material in the latter. These studies have shown that more species can be distinguished than was previously thought. Orchard (4) described six species in New Zealand: two endemic (*M. robustum* Hook.f. and *M. votschii* Schindl.) and one alien (*M. aquaticum* (Vellozo) Verdc.). Three species were found to occur naturally in S. America: *M. aquaticum*, *M. quitense* H.B.K. (the earlier, valid name for *M. elatinoides* Gaudich.) and *M. mattagrossensis* Hoehne (1). Orchard (pers. comm.) has delimited 36 species in Australia of which about 30 are endemic and one (*M. aquaticum*) is alien. Australia is the main centre of diversity for the genus (4).

Within Australia, many of the species have a restricted occurrence, being known from relatively few populations (Orchard pers. comm.). An example of this is *M. tillaeoides* Diels which is only known from the vicinity of Perth in Western Australia (2). Other species have a markedly southerly distribution. For example, *M. salsugineum* occurs in Tasmania, is frequent in southern Victoria and south eastern South Australia, but is infrequent in northern Victoria and south eastern New South Wales (1). This distribution can be contrasted with that of *M. dicoccum* F. Muell. which occurs in the Cape York Peninsula, Queensland, around the Gulf of Carpentaria and through Arnhem Land in the Northern Territory to the King Leopold Ranges in north eastern Western Australia. It has also been reported from New Guinea, Java, India and Vietnam (Orchard, pers. comm.). Another contrast is provided by *M. verrucosum*, which is recorded from every State in Australia except Tasmania, and has also been introduced to Bedfordshire England, supposedly in a wool clip (2).

The alien, *M. aquaticum*, was first reported in Australia from Sydney in 1908. Since then it has been recorded a number of times from coastal regions of New South Wales (5). It was recorded from one site in Tasmania in 1948, from several naturalised populations in Victoria between 1961 and 1966 (2) from Queensland in 1960 and 1965, and from Western Australia in 1977 (5). The plant continues to spread in Australia mostly as an escape from garden ponds and aquaria. Evans (5) recorded that one major water plant nursery in Melbourne was selling approximately 2000 pots of *M. aquaticum* annually.

Physiological Studies

The widespread occurrence of *Myriophyllum* species in Australia and their economic and presumed ecological importance have stimulated physiological studies of selected species. Denny, Orr and Erskine (6) developed apparatus to investigate carbon assimilation of submerged aquatic plants in natural waters and Orr, Pokorny and Denny (unpublished) have used this apparatus for studies of photosynthesis of *M. salsugineum*.

M. salsugineum is representative of those species in the genus which depend for the bulk of their photosynthesis on submerged leaves. Aerial stems are small and only produced during flowering and fruiting. The deeply incised, pinnatifid leaf structure of the submerged leaves of *Myriophyllum* have about double the surface area of flat entire leaves with a similar thickness. Hutchinson (7) postulated that this effectively desensitises the plant to boundary layer resistance by increasing the surface area to volume ratio. Experiments by Gessner (8) support this view by showing that the relative photosynthetic rate measured in M. heterophyllum Michx. increased by only 9% when plants were transferred from still to turbulent water.

Lloyd, Canvin, and Bristow (9) demonstrated a distinct difference between the photosynthetic response of the aerial leaves and submerged leaves of Eurasian watermilfoil, M. spicatum L. At 25°C the aerial leaves became light saturated at a photon flux density in excess of 1200 μmol m^{-2} s^{-1} PAR. The submerged leaves showed light saturation at about 200 μmol m^{-2} s^{-1}. The maximum rate of photosynthesis in the aerial leaves was about 17 μmol (CO_2) mg^{-1} chl h^{-1}, but was only about 9 μmol (CO_2) mg^{-1} chl h^{-1} in submerged leaves measured in air at zero vapour pressure deficit.

Similarly, the light compensation point was much lower in the submerged than the emergent leaves: 50 μmol m^{-2} s^{-1} PAR for the submerged and 90 μmol m^{-2} s^{-1} for the emergent leaves.

Such differences in photosynthetic capacity between aerial and submerged leaves is well known in aquatic plants. The submerged leaves of emergent species of Myriophyllum probably serve to enable the shoot to reach the surface. Thereafter the emergent leaves carry out the bulk of photosynthetic carbon assimilation, whereas in submerged species the submerged leaves must supply all photosynthate.

Orr, Pokorny, and Denny (unpublished) have shown that M. salsugineum has a light compensation point of about 10 μmol m^{-2} s^{-1} PAR, but their method of measurement was different to that of Lloyd et al. (9) and therefore makes direct comparisons invalid.

Orr et al. (unpublished) also showed that M. salsugineum at 25°C, a saturating photon flux density, an alkalinity of 1.0 meq dm^{-3} and a pH of 6.20 had a photosynthetic rate of 36.9 mg (CO_2) g^{-1} dwt h^{-1} (91 μmol (CO_2) mg^{-1} chl h^{-1}), a rate of photosynthesis that is 10 times greater than the rate measured by Lloyd et al. (9) for M. spicatum. The rate measured by M. salsugineum is 6.5 times the rate measured for a community of the floating aquatic angiosperm, Eichhornia crassipes (Mart.) Solms-Laub. with a density of 2.30 g (petiole and lamina) dm^{-2} (water surface area), of 5.7 mg (CO_2) g^{-1} dwt (petiole and lamina) h^{-1}, but is comparable to the rate of 32.9 mg (CO_2) g^{-1} dwt h^{-1} measured for a community of Salvinia molesta Mitchell at a density of 0.24 g^{-1} dwt dm^{-2} (water surface area)in winter (10).

The ability to utilise bicarbonate ion for photosynthesis confers an advantage by enabling completely submerged plants to grow in waters of pH greater than 8.3. Polarised transport, where bicarbonate is taken up by the lower leaf surface and hydroxyl ions excreted from the upper surface has been demonstrated for M. spicatum. The high pH on the upper surface causes carbonate encrustations. These encrustations are found only on species belonging to the "M. elatinoides" complex, such as M. quitense, the M. spicatum/exalbescens complex and M. verrucosum. M. salsugineum belongs to the M. elatinoides complex and should therefore be capable of using bicarbonate. Carbonate encrustations do form on the leaf in alkaline waters, but when photosynthesis was measured at high pH under conditions where the boundary layer was kept small by rapid stirring, M. salsugineum showed little photosynthetic response. This may partly explain its apparent limitation to still and slow flowing waters only.

The existence of C_4 metabolism has not been proved conclusively for any submerged plant. However, Salvucci and Bowes (11) noted the ability for submerged aquatic macrophytes to change their CO_2 compensation points (Γ) in response to temperature, pH and photoperiod. They showed that, whilst Γ will vary in M. aquaticum in response to environmental temperature and photoperiod changes, significant changes in important C_4 enzymes did not occur and some other explanation for the change needed to be found. In particular, PEP carboxylase remained low, and RuBP/PEP carboxylase ratio remained high. They also showed that emergent leaves are distinctly C_3 in nature and even when submerged in conditions promoting low Γ, the latter remained high.

Van, Haller and Bowes (12) demonstrated a temperature optimum for M. spicatum of 35°C and a Γ of 19 ppm (v:v) CO_2. Stanley and Naylor (13) also demonstrated low Γ and high temperature optima, which is similar to tropical grasses, but found little or no evidence for C_4 metabolism. It is anomolous from a physiological viewpoint though, to have high temperature optima for photosynthetic activity in plants which are primarily temperate species, are adapted to cool water, and appear to photosynthesise using the C_3 pathway.

Problems and Management

The most common problem caused by the presence of large populations of Myriophyllum species in Australia is interference with flow in the channels and drains of irrigation systems and flood mitigation schemes. Some of the emergent species in the closely related group hitherto called M. propinquum A.Cunn., are the most troublesome in this situation. The plant populations originate on the edges of the channels and grow out into the deeper water where they commonly form mixed populations with the floating pondweed, Potamogeton tricarinatus F.Muell & A. Benn., ex A. Benn., and the submerged ribbon-weed, Vallisneria spiralis L. It is in these situations that the Myriophyllum species are most troublesome because they impede flow and are capable of reducing it to substantially less than design flow. Mitchell (14) listed M. propinquum sens. lat. as one of 29 species of aquatic plants causing serious weed problems in Australia, but such problems from this species group are infrequent. Sainty (15) and Sainty and Jacobs (16) also remark on its capacity to inhibit flow although Sainty (pers. comm.) pointed out that for the most part the plant is only controlled as part of a general program to maintain a channel system, rather than because the plant is a problem per se.

The method used to manage this plant in irrigation systems is mechanical control with drag line or back actor. These are used mainly when channels and drains require reforming or desilting. On these occasions both silt and plant material including any M. propinquum sens. lat. are removed. The other form of control that is potentially available is the application of the herbicides, 2,4-D, dichlobenil and amitrole. However these are either unsuitable for use in irrigation supply water, or are not registered for use against Myriophyllum in Australia.

Two other native species cause problems in lakes and farm dams: M. verrucosum and M. salsugineum. The former is widespread and during summer is capable of occupying large areas of farm dams where it can be recognised readily by the characteristic red-purple colour of the emergent leaves. In large populations, it interferes with stock drinking and imparts a fishy taste and odour to the water, thereby degrading it for stock and domestic use. The problem is exacerbated by the difficulty of control. Application of herbicides have to take account of water volume so as to avoid dilution below the effective dose and can therefore be expensive economically and environmentally. In clear standing water, diquat is effective at concentrations of 0.5 - 1.0 mg dm^{-3} active cation but in turbid water 2,4-D ester at a rate of 3 kg ha^{-1} active ingredient has been used (C. Ripper, pers. comm.).

M. salsugineum has caused a major problem in Lake Wendouree, Ballarat, Victoria. The lake is about 238 ha in area with a mean depth of 1.7 m when full. The majority of the lake is occupied by dense stands of submerged M. salsugineum although patches of Chara also occur. The lake is mainly used for recreation and is an important aesthetic component of the City of Ballarat. In the centre of the lake there is a rowing course that was used for the Melbourne Olympic Games in 1956 and is still used for national and state regattas. About another two thirds of the lake is used for yachting. The weed interferes with both these forms of use and with other recreational uses of the lake, as it rises close to the surface when mature.

Herbicides were tested but found to have only a temporary effect and were expensive to repeat. Also the importance of recreational fishing and the proximity of houses and gardens made their use inadvisable. The City Council, who is the responsible authority, have used floating, mechanical harvesters for a number of years. The most recent is a large, complex, efficient machine designed and constructed locally. Unfortunately, although it is capable of removing large amounts of material every day, it is unable to keep up with regrowth in those years when the water is clear and temperatures are above average.

An examination of the lake followed by discussion with the users showed that the lake could be demarcated into three zones in order of importance for recreational use. The highest priority was afforded to those areas that should be clear at all times and included the rowing course. The next priority was assigned to areas which should be clear of weeds as often as possible. These comprised most of the areas used for yachting and other recreational boating. The third priority was assigned to areas to be cleared when possible but not every year. The harvesting contractor was requested to cut the highest priority area under a system designed to maximise the effect on the weed. This was to be determined by a series of experiments on frequency and timing of cutting, as carried out by Nichols (17) on M. spicatum L. Depending on the time available and the amount of growth in a particular year, second priority areas would then be cut as close as possible to times likely to have the greatest adverse impact on the weed. It was anticipated that in most years these areas could also be adequately maintained for regular recreational use.

Third priority areas would probably not be managed in those years in which the weed grew most vigorously. However, it was anticipated that, after a time, the persistence of harvesting in the highest priority areas may be sufficient to markedly decrease the weed population, thus allowing progressively more time to be spent on the other areas of lesser priority.

Some experimental harvesting indicated that in areas of bottom to surface weed growth the bulk of the material was close to the surface. Thus most of the plant material could be removed rapidly with a relatively shallow cut and the rest when convenient. However, as the pattern of regrowth following cutting was stem elongation followed by coppicing, it was necessary to cut below the point of multiple branching. Cutting experiments also showed that three cuts during the growing season effectively reduced the plant material present and that three cuts were better than two and two were better than one. Experience over the past few years since the program was initiated have shown that in most years public demand has made it difficult to cut the same area three times and priority one and most of priority two areas are now cut twice a year (R. Nuttall, pers. comm.)

M. aquaticum is not regarded as a serious weed in Australia. It inhibits flow in drains in the area for which the Dandenong Valley Authority is responsible and in small creeks in northern New South Wales and southern Queensland (Orchard, pers. comm.). However after careful consideration the National Committee on Management of Aquatic Weeds did not recommend that it be declared a noxious plant in spite of its alien status (18).

Wastewater Treatment

The possibility of using *M. aquaticum* as a means of removing plant nutrients from hyper-eutrophic wastewaters has been examined in Australia. Other alien floating species which have been used elsewhere, such as *Eichhornia crassipes*, are not available for this purpose as they are declared noxious weeds and it is an offence to cultivate them. Jackson and Gould (19) compared the efficiency of *M. aquaticum* to reduce biochemical oxygen demand (BOD) with *Spirodela*, *Salvinia molesta* (notwithstanding its noxious status) and *Typha* in synthetic sewage with a BOD of up to 200 mg dm^{-3} *M. aquaticum* was shown to be the most effective, reducing BOD by 97% after four weeks.

Rippingale and Smith (20) and Smith, Janssen and Rippingale (21) harvested *M. aquaticum* at weekly intervals from wastewater ponds and demonstrated significant reductions in total nitrogen and total phosphorus of 0.65 g m^{-2} day^{-1} and 0.15 g m^{-2} day^{-1}, respectively, over about a year.

Nuttall (22) investigated the nitrogen and phosphorus removal from secondary treated sewage effluent by *M. aquaticum*. Three lagoons 100m long, 7m wide and 1m deep were constructed below the sewage treatment plant and each received secondary treated effluent at a continuous flow rate of 0.12 ML day^{-1}. Floating roots of *M. aquaticum* were encouraged to grow over the surface of the lagoons. The first lagoon received no further treatment, other than 4 regularly spaced bubble curtains to mix the water. The second lagoon was aerated by 80 such bubble curtains and the third aerated and covered by a double skin polyethylene greenhouse canopy to reduce adverse effects of wind and winter frosts. The aerated covered lagoon was the most efficient removing 41% ammonia nitrogen, 17% total nitrogen, 4.2% total phosphorus, 65% suspended solids and 18% BOD. Based on a management system of removing half the macrophyte cover every 16 days, it was calculated that 0.78 g m^{-2} day^{-1} N and 0.31 g m^{-2} day^{-1} P could be removed from an exposed aerated lagoon. If the lagoon was also covered, at least during spring and winter, removal rates would be 0.85 g m^{-2} day^{-1} N and 0.33 g m^{-2} day^{-1} P. However an important component of the system was the presence of nitrifying bacteria which were especially associated with the plant roots. In the aerated system these bacteria were capable of converting ammonium N to nitrate N which was taken up by the macrophytes. Unfortunately harvesting the plants removed part of the bacterial population with a consequent increase in ammonium N.

Conclusions

Australia is an important centre of diversity for *Myriophyllum*. The great majority of species are well adapted to the uncertain aquatic regime which is characteristic of the continent and in which they evolved. Little is known of their ecological role in these aquatic ecosystems. Relatively few of the species provide problems for human use of aquatic resources. The only member of the genus that is seen as potentially useful is the alien, *M. aquaticum*.

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HISTORY OF THE SPREAD OF EURASIAN WATERMILFOIL THROUGH THE OKANOGAN AND COLUMBIA RIVER SYSTEMS (1978-1984)

Robert M. Rawson

U.S. Army Corps of Engineers, Seattle District
P.O. Box C-3755, Seattle, Washington 98124-2255

The spread of Eurasian watermilfoil (*Myriophyllum spicatum* L.) from British Columbia, Canada, through the Okanogan River system and into the Columbia River is discussed. Control efforts by Okanogan County, the Washington State Department of Ecology, and the U.S. Army Corps of Engineers included a fragment barrier, application of herbicides, and the use of a diver-operated suction dredge. In spite of these control efforts, milfoil has spread downstream very rapidly.

The British Columbia Ministry of Environment was the first agency in this area to become seriously involved with Eurasian watermilfoil. As the plant spread through the Okanogan Lake chain, they realized that because of the direct water connection, the problem would have serious implications for the United States as well. While they were fighting the weed in their waters, they took time to try to educate the governmental agencies in Washington State.

Due to their efforts, and information gathered from other parts of the country, the Washington State Department of Ecology became convinced that Eurasian watermilfoil did indeed pose a serious threat to Washington State waters. Therefore, in 1977, they requested the Seattle District, U.S. Army Corps of Engineers, to assist in establishing a statewide program to prevent the spread of milfoil in Washington State.

The Corps of Engineers has Congressional authority to participate in aquatic plant control programs on a cost-share basis with local governments. This authority stems from the serious water hyacinth problems in navigable waters of the southern states.

The Seattle District agreed to assist in the establishment of the statewide program, but we had no experience in dealing with aquatic plant problems. Therefore, we requested assistance from the Corps of Engineers Waterways Experiment Station in Vicksburg, Mississippi. The personnel at the Waterways Experiment Station have had extensive experience in treating aquatic plant problems, but at that time had very little experience in attempting to prevent problems.

In 1979, the Waterways Experiment Station began a Large Scale Operational Management Study to test preventive techniques. This study was focused primarily on the U.S. portion of Osoyoos Lake and the Okanogan River.

While the Seattle District office was writing the environmental impact statement and looking for alternative treatment methods, and the Waterways Experiment Station was gearing up for their research effort, the Washington State Department of Ecology and Okanogan County were monitoring the U.S. portion of Osoyoos Lake. When pioneer colonies began to show up in 1978, they treated each new area with the granular formulation of 2,4-D. Treatment at that time was done by a small hand spreader.

The Department of Ecology and the County also installed a fragment barrier across the Okanogan River at the town of Oroville to help keep fragments from moving downstream.

We knew from the start that milfoil would eventually become established in the U.S. portion of Osoyoos Lake. The main purpose of our program was to try to keep the fragments from spreading into the Okanogan River and down into the Columbia River. The chemical treatments in Osoyoos Lake were meant to eliminate pioneer colonies which would provide a fragment source for further downstream spread. The barrier was our last line of defense.

Milfoil reaches the U.S. portion of the Okanogan River

By the time the Waterways Experiment Station began their field work in 1979, milfoil colonies could be found in the upper reaches of the Okanogan River.

The existing fragment barrier was not working up to expectations, so the first project completed under the Large Scale Operational Management Test was a redesigned fragment barrier. Research on the barrier showed that it was only about 60% effective in intercepting plant fragments. This was because of problems with the contour of the river bottom and the high river flows in the spring.

Because of the large escape rate of the milfoil fragments, other treatment methods had to be used. The Waterways Experiment Station tested existing methods for effectiveness. They tested standard aquatic herbicides applied in granular formulations and liquid formulations applied with adjuvants to restrict drift. The liquid formulations were applied using trailing hoses. While the experiments were being conducted, the Osoyoos Lake treatment program continued with 2,4-D granular applications.

In spite of these efforts, milfoil was discovered at the mouth of the Okanogan River in 1980. Waterways conducted tests on milfoil fragments to determine how long they would remain viable while floating in the water. They found that the fragments could survive for several weeks. When this is taken into account along with the rate of flow in the Okanogan River, it is easy to understand how the plant spread so rapidly.

The testing and operational treatments continued in 1981, but the emphasis changed from preventing milfoil totally to preventing problem level populations from developing. The Waterways Experiment Station testing continued with controlled release herbicides and a diver-operated suction dredge which we borrowed from the British Columbia Ministry of the Environment.

Milfoil reaches the Columbia River

In 1981, we discovered the first milfoil colonies in the Columbia River. We tried chemical treatments in the Columbia, but the water flow was too great and the resulting herbicide drift was unacceptable. The suction dredge presented too many problems to use on a large scale and no other treatment method looked promising. The milfoil went untreated and rapidly spread downstream.

In 1982, it was discovered downstream of Rock Island Dam and by 1984 had advanced as far as Priest Rapids Reservoir. We will be surveying the lower reservoirs next month with the Department of Ecology. We expect to find milfoil in the Hanford Reach area or even into McNary Reservoir. We expect that milfoil will be all the way to the mouth of the Columbia within the next several years.

The milfoil in the Okanogan River is so thick now that it is actually obstructing the drainage of Osoyoos Lake. The populations in Osoyoos Lake are being kept under control by the annual treatments, but the populations in the Columbia River continue to increase.

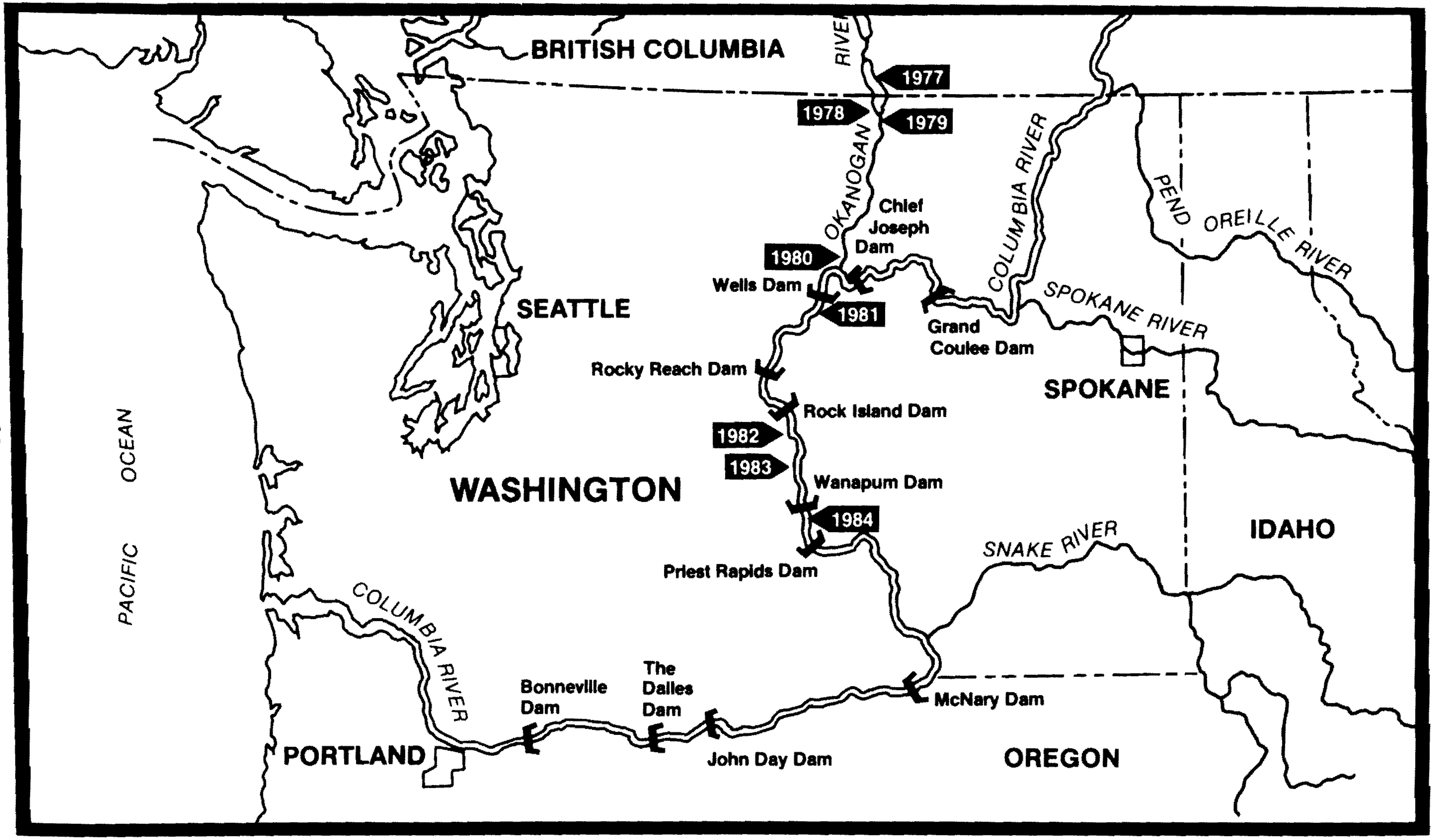
We are continuing our public information program to try to limit the amount of milfoil moved from one water system to another by boats or other human activities.

We are also supporting the Waterways Experiment Station in the development of treatment methods which would be effective in flowing water. These could be chemical treatments which would have to overcome the drifting problem, or some type of biological control agent. Grass carp research is one thing we are helping to fund. We are also interested in the work being done with plant pathogens.

Besides the Okanogan and Columbia Rivers, we are helping to fund treatments in heavily used waters to prevent the loss of public use. This program includes chemical treatment in the Pend Oreille River and the harvesting and fiberglass bottom screens being used in the Seattle area.

We believe that all of the present control methods, because of various problems, are of a stop-gap nature and that the final solution, if one can be found, will be some type of biological control. For that reason, we will continue to support research in this area.

FIGURE 1. Downstream limit of Eurasian watermilfoil by year.



MYRIOPHYLLUM HALORAGACEAE SPECIES IN BRITISH COLUMBIA: PROBLEMS WITH IDENTIFICATION

Oldriska Ceska

Biology Department, University of Victoria,
Victoria, B.C., V8W 2Y1, Canada

and

Adolf Ceska

British Columbia Provincial Museum,
Victoria, B.C., V8V 1X4, Canada

Six native species of *Myriophyllum* (i.e. *M. sibiricum*, *M. verticillatum*, *M. quitense*, *M. ussuriense*, *M. farwellii* and *M. hippuroides*) and three introduced species (*M. spicatum*, *M. heterophyllum* and *M. aquaticum*) occur in British Columbia. Thin layer chromatography of flavonoids helps to identify sterile specimens of most of these taxa of *Myriophyllum*.

The infestation of British Columbian lakes by the Eurasian water-milfoil, *Myriophyllum spicatum* L. s.str., triggered intensive surveys of aquatic macrophytes, especially in southern parts of British Columbia. These surveys led to the discovery of several species of aquatic and wetland plants new to the British Columbian flora (1), including two species of *Myriophyllum* (2). Difficulties identifying *Myriophyllum* species spurred the development of alternative identification techniques of identification based on differences in flavonoid chemistry (3). Herbarium studies (4) resulted in the name *M. exalbescens* Fern. being changed to *M. sibiricum* Kom. This paper summarizes our work (1-4) on *Myriophyllum* in British Columbia.

History of milfoil collections in British Columbia

The first list of Canadian plants (5) refers to only one species of *Myriophyllum* from British Columbia, *M. spicatum*. Henry (6) lists *Myriophyllum spicatum* as occurring in Kamloops and Sproat Lake, and *M. verticillatum* L. as "also attributed to British Columbia". Carter and Newcombe (7) report both *M. spicatum* and *M. verticillatum* on Vancouver Island. Eastham (8) uses the name *M. exalbescens* Fern. for what previous authors called *M. spicatum*, gives an additional locality for *M. verticillatum*, and includes an additional species, *M. hippuroides* Nutt., the report of which, however, was based on a specimen (Eastham 10,174 Aug. 13, 1942 - CAN, UBC) later identified (9) as *M. heterophyllum* Michx.

In 1974 Kistritz collected *M. hippuroides* in the Mission area in the lower Fraser Valley. Later surveys showed that this species is common there (10). This occurrence is, without doubt, the northern limit of the species' natural area of distribution, which ranges from California to Washington and the southern part of the British Columbian mainland. The old collections of M. hippuroides from Sumas Lake (Macoun 44,408, Aug. 21, 1901 - CAN) and Pitt River (Henry s.n. Oct. 1916 - V) strongly suggest that M. hippuroides had been overlooked in British Columbia.

In 1961 Hitchcock and Cronquist (11) misinterpreted Patten's conclusions (12) and mistakenly treated not only M. spicatum s.str. and M. exalbescens, but also M. verticillatum, as a single species. In their scheme the former two taxa were identified as M. spicatum var. exalbescens (Fern.) Jeps., and M. verticillatum was synonymous with M. spicatum var. spicatum. In all likelihood influenced by this treatment, Calder and Taylor (13) tentatively assigned sterile specimens of M. verticillatum from the Queen Charlotte Islands to M. spicatum. Hultén (14) pointed out Hitchcock and Cronquist's (11) error but Scoggan (15) followed their treatment. Scoggan also mentioned the possibility of the occurrence of M. alterniflorum DC. in British Columbia. The specimen he cited, however, is actually M. sibiricum. In addition, Scoggan's records of M. hippuroides are based on misidentified specimens of M. verticillatum.

Ceska and Warrington (16) reported an additional Myriophyllum species, M. farwellii Morong, from northern Vancouver Island. Intensive field work led to the discovery of M. ussuriense (Regel) Maxim. and M. quitense HBK in British Columbia (2). Further herbarium studies showed that both of these species were previously collected in British Columbia; M. ussuriense was collected by Henry in Pitt River (Henry s.n. Oct. 1916 - V; with M. hippuroides), and M. quitense by Macoun in Kennedy Lake (Macoun 88,556, July 23, 1909 - CAN), and Carl in Cowichan River (Carl s.n. July 1935 - UBC,V). The examination of Russian Myriophyllum collections proved that Komarov (17) described the taxon known in North America as M. exalbescens five years before Fernald (18) did. Therefore Komarov's name, M. sibiricum Kom., has priority (4).

In addition to the species mentioned above, M. aquaticum is occasionally planted in garden pools.

At the present time, nine species of Myriophyllum are known to occur in British Columbia. Six of them are native (M. sibiricum, M. verticillatum, M. quitense, M. ussuriense, M. farwellii and M. hippuroides), and three introduced (M. spicatum, M. heterophyllum, and M. aquaticum).

Identification of British Columbian Myriophyllum

The identification of flowering plants is usually relatively easy. The shape of floral bracts, number of stamens, surface of mericarps and eventual dioeciousness of plants are important characters in identification. Characters on vegetative parts of the plant (such as the presence of winter buds, stem colouration, occurrence of single alternate leaves outside of regular leaf whorls, number and position of leaflets, and presence of blackish glands on the stem and leaflets) provide additional clues. The identification of sterile plants, on the other hand, can be difficult when one depends only on vegetative characters.

In order to facilitate identification, utmost care should be taken in collecting samples and preparing herbarium specimens. One should always look for flowering or fruiting plants, since they may be scattered among sterile plants and easy to overlook. Whole plants should be collected when possible, because in some species, namely M. quitense, the basal parts have characters important for identification. The plants should be gently washed of detritus and filamentous algae. The specimens should be floated in a photographic tray onto a sheet of mounting paper or paper of a slightly lighter weight, put in a newsprint folder and dried in between corrugated cardboard, preferably in a warm air drier. One should avoid treating specimens with organic solvents before pressing. Drastic pest control methods (e.g., dipping in the mercury-chloride solution) should not be used on dried specimens because these measures impede possible chromatographic identification.

Myriophyllum sibiricum Kom. (syn.: M. exalbescens Fern., M. spicatum subsp. squamosum Laest. ex Hartman fil.)

Myriophyllum sibiricum, the most common species of Myriophyllum in British Columbia, occurs in lakes and water bodies throughout the province. It has a broad ecological amplitude. In eutrophic waters, marl lakes and slightly alkaline lakes it occurs more often than any other Myriophyllum species. It also occurs in the brackish water of the Somass River estuary (Vancouver Island).

In 1914 Komarov (17) described Myriophyllum sibiricum from Kamchatka and Siberia. In North America, Fernald (18) described the same taxon as M. exalbescens, but Komarov's name, M. sibiricum, has priority. Later Aiken and other authors (19-23) reported the species' occurrence in Scandinavia. Our study of herbarium material from Leningrad (4) shows that M. sibiricum is indeed a circumpolar species, Aiken (19) and Faegri (22) suggested.

Myriophyllum sibiricum is characterized by its entire or shallowly cut floral bracts which are shorter than fruits, whitish stems, the formation of winter buds, and leaves with up to 12 pairs of segments.

Myriophyllum spicatum L.

In British Columbia M. spicatum was introduced (during the early 1970s ?) into the lakes, sloughs, channels, and duck ponds of the southern part of the province. At present it occurs on Vancouver Island (Duncan), Pender Island, the lower Fraser River valley (sloughs and ponds in the Chilliwack area, Hatzic and Cultus Lakes, Boundary Pond), Okanagan Valley, Shuswap Lake, and Champion Lakes near Trail. It prefers eutrophic habitats.

Originally, Myriophyllum spicatum was probably distributed in Europe, northern Africa (24), and northeastern Asia (the Russian Far East and Japan). It was introduced to other continents and has become a noxious aquatic weed. In the Leningrad herbarium collection from Asia there are only a few specimens from central Siberia (the Baikal Lake area); this may indicate that the species is rare in continental Siberia. It is, however, well represented in the herbarium material from the Russian Far East and common in Japan.

Myriophyllum spicatum is characterized by entire floral bracts that are shorter than its fruits, leaves usually having more than 12 pairs of segments, and the absence of winter buds. It is generally difficult to distinguish from M. sibiricum. For the detailed account of its characters see works by Löve and Aiken et al. (25,26). The number of leaflets and the colour of stems are the best characters for distinguishing these two species. Growth experiments conducted by Aiken and Picard (27) showed that on nutrient-poor substrate, the leaves of M. spicatum converged in appearance towards the leaves of M. sibiricum grown on nutrient-rich substrate.

There is yet another character, however, which can be used to differentiate these two species. The leaflets of M. spicatum usually stand at acute angles up to about 45 degrees and are parallel to each other. The leaflets of M. sibiricum, on the other hand, stand at angles from 45 degrees to angles almost perpendicular to the rachis. They are not as regularly parallel to each other as those of M. spicatum.

Myriophyllum verticillatum L.
This species occurs throughout British Columbia. Although it clearly prefers dystrophic or oligotrophic waters, it is also found in marl lakes (providing it can root in a substrate other than precipitated calcium carbonate) and brackish water (the Somass River estuary, Vancouver Island).

Myriophyllum verticillatum is a circumpolar species. In North America it occurs from Alaska, the Yukon (28,29), British Columbia to Newfoundland, south to Florida, and from the east coast west to Minnesota and Utah (19,30). Mason (31) also reports its rare occurrence in California.

This species is clearly distinguished from the other British Columbian species by its pectinate floral bracts which are usually longer than its flowers and fruits, green stems, and characteristic clavate winter buds. Leaves are always in regular whorls, without odd alternate leaves (as in M. hippuroides), and have numerous small glands ("Myriophyllum glands", incorrectly called hydathodes in some literature).

Myriophyllum quitense HBK (syn.: M. elatinoides Gaudich.)
In British Columbia this species has been found only on Vancouver Island. It occurs in oligotrophic waters. It grows either in large lakes, in places that are exposed to wind and subjected to strong wave action, or in rivers.

Myriophyllum quitense occurs in South America and there are closely related species in Australia and New Zealand (32). In North America it was first reported (as M. elatinoides) from Oregon (18). Orchard (33) cites an additional locality in North America from Arizona and Ceska et al. (2) report it from Canada (British Columbia, Prince Edward Island), Oregon, Washington, and Wyoming.

Broad triangular floral bracts with dentate margins, a reddish tinge, and a white bloom are very characteristic of this species and there are no problems identifying flowering specimens. Unfortunately, this species flowers very rarely and when sterile it is often mistaken for one of the above three species. Two characters which help to identify M. quitense are numerous rhizomes at the base of the plant and the lowermost leaves of young shoot, which are opposite, entire, and look like bracts. The stem of M. quitense is olive green, usually darker than that of M. sibiricum. These characters are quite subtle and easily damaged when collecting. We always rely on chromatographic techniques for positive identification of this species.

Myriophyllum ussuriense (Regel) Maxim.
In British Columbia this species has been found in interior lakes (Kootenay, Mara, and Shuswap Lakes), the lower Fraser River Valley and the Lillooet River Valley (Harrison, Hatzic, and Pitt Lakes and in Pemberton), and on Vancouver Island (Kennedy Lake and Kennedy River). It grows either in shallow water close to the shore or forms large terrestrial stands in parts of the shore which are exposed in summer.

Myriophyllum ussuriense is an amphiberingian species which occurs in Asia in Taiwan, China, Primor'ye, Kamchatka, Japan. On the American side of the Pacific Ocean it has thus far only been found in British Columbia.

This species is very distinct. It is dioecious and the whole populations are formed of plants of one sex (a similar situation was observed in Japan - S. Itow, personal communication). In British Columbia all populations are formed of female plants except that in the locality of Hatzic Lake, which contains only male plants.

The habit of semiterrestrial plants of Myriophyllum ussuriense does not resemble that of any other western American species of Myriophyllum. Rather, the species can be mistaken for Crassula aquatica (L.) Schönl. It differs from Crassula in that it has leaves which are usually in whorls of three and are either pinnate or have leaflets which are reduced to short stubs. The aquatic form is similar to Myriophyllum verticillatum L. It differs from it by being more delicate, and having fewer leaves in each whorl and fewer segment pairs in the leaves. The irregular tips of the submerged leaves are quite conspicuous on fresh material. Winter buds, when developed, are very distinct in both of these species. The slender winter buds of M. ussuriense contrast with the large clavate winter buds of M. verticillatum. Terrestrial forms of Myriophyllum hippuroides Nutt. differ from M. ussuriense in that they have some leaves on the stem that are scattered outside the regular whorls.

Myriophyllum farwellii Morong

In British Columbia this species occurs on northern Vancouver Island, the lower Fraser Valley, Gray Wells Park and east Kootenay Valley (Ryan Park), usually in dystrophic or oligotrophic lakes, and rarely in more eutrophic waters (the sloughs around Agassiz).

Myriophyllum farwellii is a North American species which occurs in eastern Canada and the northeastern United States. Its occurrence in British Columbia is apparently indigenous, as there are several other species with similar distributional patterns, such as Sparganium fluctuans (Morong) Robinson, Potamogeton strictifolius Bennett, Potamogeton oakesianus Robbins and Megalodonta beckii (Torr.) Greene.

This species is easily recognized by its leaves, which are more alternate than in whorls. Flowers and fruits are scattered in the axils of the leaves, not in terminal racemes.

Myriophyllum hippuroides Nutt.

In British Columbia this species is restricted to the lower Fraser River Valley, an area from Vancouver to Hope. It occurs in lakes and sloughs. Early collections of the species (see above) indicate that it is indigenous here.

The area of distribution of this species spreads from Mexico and California to Washington and southern British Columbia. Thorough herbarium studies are needed to establish this species' eastern limits of distribution.

In its general habit and with its pectinate bracts, which are longer than flowers, Myriophyllum hippuroides resembles robust plants of M. verticillatum. It differs from it, however, in that it has a few leaves on the stem scattered outside the regular whorls. M. hippuroides lacks winter buds.

Myriophyllum heterophyllum Michx.

This species from eastern parts of North America, was introduced into British Columbia in Vancouver (in Beaver Lake in Stanley Park, where it is now extinct, and in Queen Elizabeth Park ponds).

The species is very similar to M. hippuroides, but differs from it in its fruits (19,30,34) and pollen morphology (9). Both of these characters are of little value in field identification because both species produce fruits only rarely. We have not seen enough herbarium material to be able to judge the stability of other characters, such as broader floral bracts. Unfortunately, many species of the section Tessaronia, into which this species was placed by Schindler (35) together with M. hippuroides and M. farwellii, have similar flavonoid patterns and thus chromatography is of little value as an identification tool (see below).

Myriophyllum aquaticum (Vellozo) Verdc. (syn.: M. brasiliense Cambess.)
In the Vancouver and Victoria areas this species is often planted in garden pools and ponds. It remains green throughout the winter but does die in longer frost periods (about once every three years in the Vancouver area).

The dioeciousness of plants, their robustness, floral bracts identical in shape to leaves (and just slightly smaller than leaves), and the characteristic green colour of plants are the characters by which M. aquaticum is easily identified.

Using flavonoid patterns for identification

Everyone who has dealt with the identification of Myriophyllum has come across many questionable specimens. While the identification of perfectly developed plants does not pose problems, the reliable identification of sterile, atypical, or terrestrial plants is extremely difficult.

In order to distinguish Eurasian water milfoil from native British Columbian species, the senior author applied chromatographic techniques for the routine identification of Myriophyllum samples (3). These techniques proved to be generally more reliable than identification based only on morphological characters. The combination of traditional identification with chromatography greatly improves chances of correct identification.

The aquatic environment has a strong influence on the morphological characters of aquatic plants. Anatomical and morphological structures are greatly modified and conductive tissues reduced. Leaves are either finely dissected or thin ribbon-like in order to cope with very slow gas diffusion. As a result, not only closely related species, but plants belonging to different genera or families, may have very similar habits. The chemistry of specific compounds, such as flavonoids, on the other hand, is not influenced by the aquatic environment and there is no convergence similar to that of the morphological characters.

Our studies show (10) that aquatic plants have a relatively high flavonoid content (e.g., Myriophyllum, Isoetes, Ceratophyllum). The exception is, for instance, Ranunculus subgen. Batrachium. Some flavonoid compounds may decompose in herbarium material (36), but preliminary tests showed that herbarium specimens of Myriophyllum retain all the major flavonoids contained in fresh material. Dry herbarium specimens retain their characteristic flavonoid patterns for many years.

A preliminary study of Myriophyllum samples showed that there was no difference in flavonoid content in the upper and middle portion of the plant. The inflorescence, branches terminated by the inflorescence, and the middle part of the stem have essentially the same flavonoid content. The part of the stem below the first leaves and that directly above the rhizome have very low flavonoid content. Leaves exhibit about the same flavonoid pattern as the stem but their flavonoid concentration is usually higher. As a result of this survey, leaves from the middle portion of the plant (from sections about 10 cm long) were used for extraction.

Plant material was extracted for at least 12 hours with 1% HCl in methyl alcohol. Extracts were stored at -15 °C. The crude extract was used for separation by two-dimensional TLC with Avicel S.F. Microcrystalline cellulose as a medium. Solvents were: ethyl acetate - formic acid - water (140:28:42) for the first direction, and 15% acetic acid for the second direction. For better separation the chromatograms were developed twice in the first direction.

The chromatograms were viewed under UV light (using long wavelengths). All spots were outlined on the chromatograms. A photocopy of the chromatogram was obtained using Brinkman's TLC photocopier and a chromatogram map was traced on onion-skin paper. The photocopy and traced copy were filed together as a record of the sample's pattern.

The chromatograms were then sprayed with sodium carbonate (3% solution in water). After more than 24 hours (the time necessary for the formation of coloured complexes of flavonoid compounds) they were again examined under UV light. All colour changes were noted on the traced copy of the chromatogram.

Over 500 *Myriophyllum* samples have been analyzed to date. The distribution of diagnostic spots in six *Myriophyllum* species is given in Table I. The characteristic flavonoid patterns of chromatograms (after spraying with sodium carbonate) are given in Figure 1.

Myriophyllum sibiricum, *M. spicatum*, *M. verticillatum*, and *M. quitense* have very similar patterns characterized by the presence of two different cyanidin glycosides and a predominance of flavonols. *Myriophyllum ussuriense* and *M. aquaticum* have patterns similar to each other. Like the previous group they have a predominance of flavonols, but these flavonols have a different chemical composition. *Myriophyllum farwellii*, *M. hippuroides*, and *M. heterophyllum* have only one cyanidin glycoside and a predominance of flavones. The flavonoid patterns of these three species are so similar to each other that they are of no help in distinguishing between them.

Myriophyllum sibiricum. All chromatograms of this species show two different athocyanins and major spots 1 and 2. Spots 4 and 6 are present respectively in 62% and 23% of all samples of this species, and when they occur, then they occur only in trace amounts. Minor spots 24, 32, and 30 are exclusive to this species, but they vary in occurrence. When they are present, they are excellent diagnostic spots for *M. sibiricum*. When they are absent, *M. sibiricum* can still be distinguished from *M. spicatum* by the weak development of spots 4 and 6, and from *M. verticillatum* by the lack of spots 7 and 8.

There is considerable variation in the flavonoid patterns of *M. sibiricum* due to the presence or absence of spots 1A, 4, 6, 19, 20, 24, 30, 31, 32 and 43. There is no correlation between the occurrence or absence of particular spots and this variation points out the polymorphism of the most abundant native species of *Myriophyllum* in British Columbia.

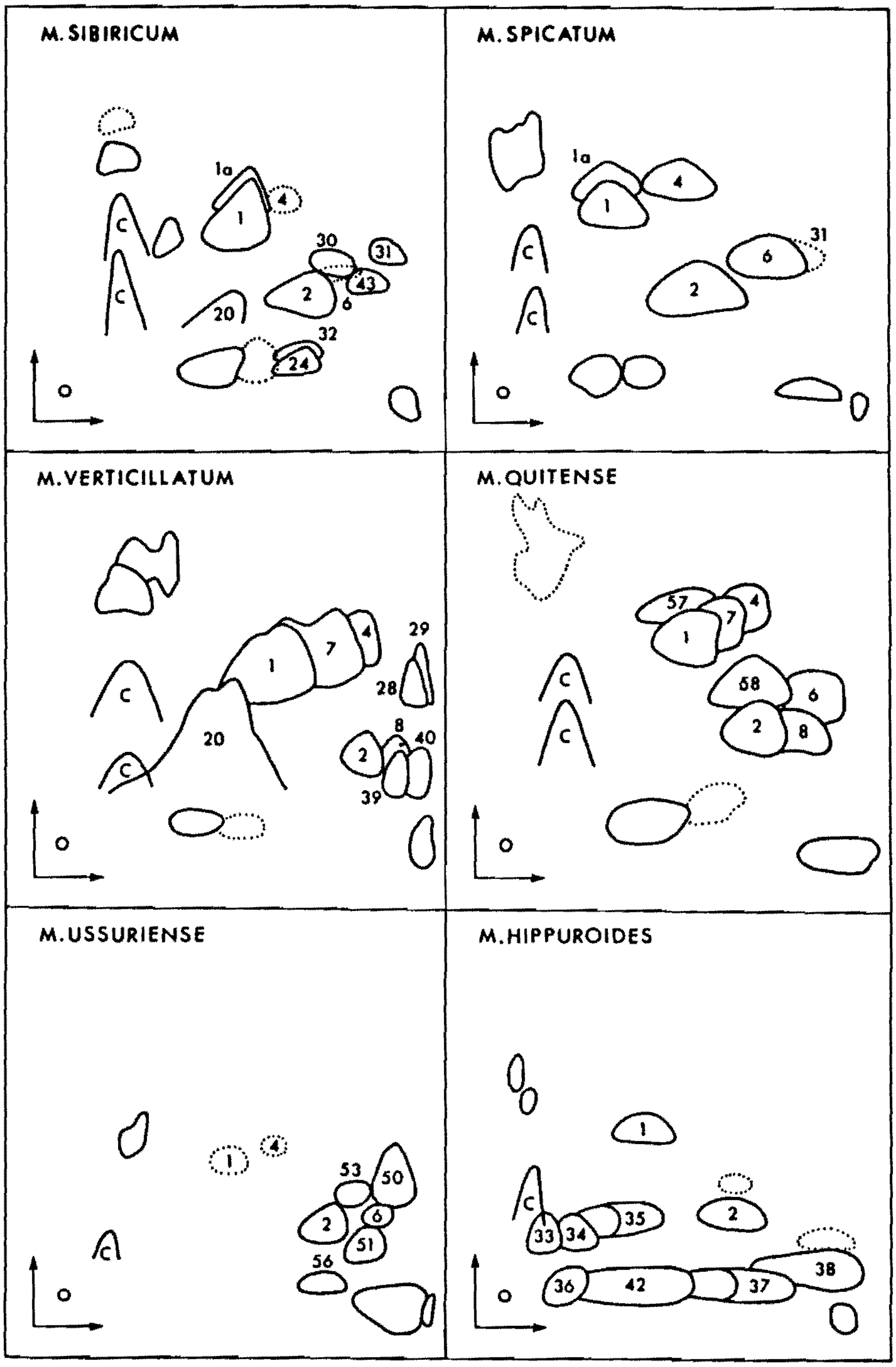
Myriophyllum spicatum. The flavonoid pattern of this species is very similar to that of *M. sibiricum*. It can be distinguished from the latter by a different concentration of spots 4 and 6. In *M. spicatum* the concentration of

Table I. Occurrence of main flavonoids in B.C. Myriophyllum species

	Myriophyllum							
Spot no	sibiricum	spicatum	verticillatum	quitense	ussuriense	hippuroides	after spray	tentative aglycone
1	+	+	+	+	tr	+	Y	quercetin
1a	+	+					Y	
2	+	+	+	+	+	+	Y	quercetin
4	tr	X	+	+	tr		G	kaempferol
6	tr	X		+	+		G	kaempferol
7			X	+			YG	isorhamnetin
8			X				YG	
20	+		+				bY	flavonoid sulphate
24	X						O	
28			X				Y	
29			X				YG	
30	X						Y	
31	+	+					Y	
32	X						lY	
33						X	dG	
34						X	BG	
35						X	dG	
36						X	lBr	
37						X	bY	
38						X	BG	
39			X				Y	
40			X				YG	
42						X	bY	
43	X						Y	
50					X		dY	
51					X		Y	
53					X		dY	
56					X		O	
57				X			dBr	
58				X			dBr	

+	present	b	bright	B	blue	Y	yellow
tr	in traces	d	dark	Br	brown	O	orange
X	diagnostic spot	l	light	G	green		

Figure 1. Flavonoid patterns of six *Myriophyllum* species.



spots 4 and 6 is usually equal to the concentration of spots 1 and 2, whereas in M. sibiricum spots 4 and 6 are present only in traces and are always much weaker than spots 1 and 2. Spots 24, 30 and 32, which occur in the majority of M. sibiricum samples, are always absent in M. spicatum. Myriophyllum spicatum can easily be distinguished from M. verticillatum by the absence of spots 7 and 8.

There is almost no variation in the flavonoid pattern of M. spicatum specimens from British Columbia. This suggests a common origin of all introductions of M. spicatum into our area.

Myriophyllum verticillatum. This species differs from M. sibiricum and M. spicatum by the presence of spots 7 and 8. Two cyanidin glycosides are always present as they are in M. sibiricum and M. spicatum. Minor spots 28, 29, 39, 40 and 41 occur only in Myriophyllum verticillatum. The absence of spots 57 and 58 distinguishes M. verticillatum from the species below. The identification value of these spots is low, however, as they occur only in a portion of the examined specimens.

Myriophyllum quitense. This species has two cyanidin glycosides and spots 1 and 2, which are common to all our species, spots 4 and 6 like M. spicatum, but usually weaker, and spots 7 and 8 like M. verticillatum. The spots characteristic of this species are spots 57 and 58, which do not occur in any other Myriophyllum.

Myriophyllum ussuriense. This species has only one spot of anthocyanin in common with the other British Columbia species. Spots 1, 2, 4 and 6 are also present in this species, but spots 1, 4 and 6 are weak. There is a number of additional spots which occur only in this species (e.g., 50, 51, 53, 56).

Myriophyllum farwellii, M. hippuroides, M. heterophyllum. The flavonoid patterns of these three species are so similar that they cannot be used to distinguish between the species. This is interesting, because M. farwellii is morphologically very distinct from the other two species in this group. Spots 1 and 2 are common to the other British Columbian species but occur here only as minor spots. The other diagnostic spots for this groups of species (33 - 37, and 42) represent a different group of flavonoids (probably flavones). Myriophyllum hippuroides, which can be mistaken for M. verticillatum when sterile, is easily identified using this chromatographic technique.

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THE EFFECTS OF ACID AND AMMONIUM DEPOSITION ON AQUATIC VEGETATIONS IN THE NETHERLANDS

C. den Hartog

Laboratory of Aquatic Ecology, Catholic University, Toernooiveld, 6525 ED Nijmegen, The Netherlands

The effect of acid precipitation on aquatic ecosystems is most spectacular in the poorly buffered soft waters on mineral sand bottoms which are practically devoid of lime. These waters are normally characterized by dominance of isoetids, particularly *Littorella uniflora*, but under influence of a decreasing pH, the inorganic carbon budget becomes disturbed, leading to suppression of *Littorella* and its companion species by *Juncus bulbosus* and/or *Sphagnum* species, and finally to the total disappearance of all submerged macrophytes. This succession becomes markedly accelerated, if the deposition contains a considerable amount of ammonia. Although the pH of the precipitation rises, under aerobic conditions NH_4^+ becomes nitrified by the activity of certain bacteria to nitrate, a process that also produces $2H^+$-ions per NO_3^--ion. Nitrification proceeds until the pH of the receiving water has reached 3.8. A further decrease of the pH can take place by direct H^+-input, and e.g. by sulphate reduction in the sediment. The lowest pH observed in The Netherlands was 2.8.

The effects of acid precipitation on hard water systems are still poorly known. In general the acid will be neutralized and the NH_4^+-input will contribute to the eutrophication. However, in some dune pools "hardening" of the water has been observed, due to dissolution of $CaCO_3$ from the bottom and the surrounding slopes. The recent appearance of *Potamogeton coloratus* in several dune pools may be a result of this water hardening.

In The Netherlands the effect of acid rain on aquatic environments has been recognized only rather recently. Of course one has noticed that the composition of a number of aquatic communities changed, and that various plant and animal species became rare or disappeared, while, quite contrary some others showed a clear increase. These changes were generally ascribed to eutrophication, or more generally regarded as a consequence of environmental degradation, an unavoidable consequence of the still expanding population.

One has to bear in mind that the number of inhabitants in The Netherlands at the beginning of the century was only ca. 5 million, in 1940 ca. 8-9 million and now has increased above 14 million. In the same time the character of the country changed from mainly agraric to highly industrialized. Even agriculture is for the main part very intensive (enlargement of scale, greenhouse culture, bio-industry), so the human impact on the environment is heavy, and pollution as a cause for the decline of the natural environment rather likely. The mechanisms of the observed changes were however hardly a subject of study, and generalizing statements about the general decline of the water quality were for that reason in many cases more based on suppositions than on evidence. Another reason why the effects of acid deposition have not been recognized at an early stage is without doubt that most waters in The Netherlands are hard waters with a high content of $Ca(HCO_3)_2$ and thus well buffered to acid additions. However, in the southern and eastern part of the country, many soft waters are found on pleistocene sandy soils which are poor in lime or almost devoid of it. These waters, locally known as "vennen" (singular "ven"), are poorly buffered and contain very little $Ca(HCO_3)_2$; they are shallow and fully mixed, with periodically fluctuating water levels; they are mainly fed by rain water; consequently they are oligotrophic, and inhabited by a highly characteristic set of biocoenoses. Most of the "vennen" are situated in nature reserves, but in spite of the management, directed towards preservation of the landscape and its natural communities, the quality of the latter has decreased seriously during the last three decades. Nevertheless the decline of the plant- and animal communities was ascribed to eutrophication and increased recreational pressure, without an indication of the eutrophicating sources.

The "vennen", which have usually a mineral sandy soil, are characterized by plant communities, classified within the Littorellion-alliance. Usually the Shore-weed, Littorella uniflora (L.) Aschers., is the dominant species covering the bottom with a fine lawn. It is accompanied by Lobelia dortmanna L., the Quill-worts Isoetes lacustris L. and I. echinospora Durieu, the Water Plantains Luronium natans (L.) Raf., Echinodorus ranunculoides (L.) Engelm. and E. repens (Lamk.) Kern et Reichg., and several other interesting species. These communities have been extensively described by Schoof-Van Pelt (1). Nowadays they are rare, and only fragmentary developed. The decline can be best illustrated by the fact that around 1950 Littorella uniflora was known from at least 227 localities of which only 35 are still existing. Lobelia dortmanna was known from 55 localities, and is at present known from one locality, where it however, has not been seen for 2 years. Isoetes lacustris is still present in one locality, where it covers a few square metres, and I. echinospora has not been seen for many years and can be considered as extinct (2).

In the years 1979-80 68 soft-water localities were selected for research on the criterion that they had in or still after 1950 Littorella uniflora as the dominant aquatic plant species (3). These 68 localities were investigated for ca. 50 physico-chemical parameters, relating to the overlying water as well as the interstitial water of the substratum. It appeared that in 53 of the 68 localities L. uniflora had disappeared or had become rare.

In 12 of the 53 localities eutrophication appeared obviously the cause of this decline. In localities where enrichment of the sediment with orthophosphate and ammonium was found, but where the water layer was relatively poor in nutrients, species like Myriophyllum alterniflorum DC and Ranunculus peltatus Schrank had come to dominance. In waters where the sediment as well as the water layer had become enriched by nutrients there were hardly bottom-rooting aquatics, but pleustophytes, particularly Lemna minor L. and Riccia fluitans L., were dominant. In some larger waters no macrophytes were found at all, but a dense plankton bloom coloured the water.

In 41 of the 53 localities, however, another development had taken place, because Littorella uniflora became as a dominant replaced by Juncus bulbosus L. and/or by Sphagnum species (among which S. cuspidatum Ehrh. was the most numerous). In some localities no plants at all were found in spite of the fact that the water was crystal clear. This successional phenomenon has been described from Scandinavia (4, 5), Belgium (6) and Germany (7, 8), but only in Scandinavia acidification of the environment has been mentioned as a possible cause. The observations in The Netherlands confirm that acidification of the water, caused by acid precipitation, leads to a decrease and the final disappearance of L. uniflora and its accompanying species, coincident with the appearance and often luxurious growth of Juncus bulbosus and Sphagnum. L. uniflora, dominant in waters with low alkalinity (0.5 meq.l^{-1}), but a pH of ca. 6.5 is replaced by J. bulbosus on sediments poor in $CaCO_3$, when the pH and the alkalinity decrease. When J. bulbosus dominates the pH is 3.8 - 4.1 and alkalinity O. On substrates which are slightly richer in $CaCO_3$ Sphagnum replaces L. uniflora. These data indicate that the inorganic carbon economy of the waters concerned is disturbed by acidification.

In poorly buffered waters, such as the "vennen", the CO_2-content is usually extremely low (6-40 μmol.l^{-1}). The species of the Littorellion alliance are well adapted for life in an environment where CO_2 is limiting. They have nearly all the isoetid growth form, which is compact, and causes a reduced surface-volume ratio. They all have a well-developed system of internal air lacunae, so that CO_2 produced during the photorespiration can be reused. Further they are able to take up CO_2 with the roots from the interstitial water, where the CO_2 level may be 10-100 times higher than in the overlying water, and to release O_2 via the roots into the substratum. In that way they can contribute themselves to the oxidation of organic matter, which provides them with CO_2. Furthermore they are equipped with a special mechanism for photosynthesis, similar to the Crassulacean Acid Metabolism (C.A.M.), and that may be interpreted as an adaption to environments, where the availability of CO_2 is precarious. Finally there is a clear relationship between the development of the underground biomass and the nutrient content of the environment. Under oligotrophic conditions the root system is well-developed, but when the nutrient availability is better, the root system is usually small compared with the aboveground biomass. This set of properties enables the Littorellion species to survive in the extreme environment of the poorly buffered, oligotrophic waters. In more alkaline waters, with a higher trophic status, these species do not stand a chance in the competition with more demanding water plants (9-16).

The addition of acid to the poorly buffered "vennen" leads to the gradual dissolution of the very low quantities of $CaCO_3$ still available in the bottom. Via $Ca(HCO_3)_2$ the $CaCO_3$ becomes transformed to CO_2, according to the following reactions:

$$CaCO_3 + H^+ \rightleftarrows Ca^{++} + HCO_3^-$$
$$HCO_3^- + H^+ \rightleftarrows H_2CO_3 \rightleftarrows H_2O + CO_2$$

The result is that CO_2 diffuses into the water layer. Littorella uniflora as well as Juncus bulbosus, which normally occurs in small quantities in this environment, profit from this CO_2-increase and increase their photosynthesis. However, the isoetid growth form, appears to be unfavourable under the improved CO_2-conditions in the water layer. The small thick leaves of Littorella have an unfavourable surface-volume ratio in comparison to the fine, almost filiform leaves of the much larger Juncus bulbosus. The latter species has a comparatively low root biomass which plays a minor role in the uptake of CO_2. It depends for CO_2 uptake almost completely on its leaves. Consequently, it is favoured by the new conditions; it grows much faster, overgrows Littorella, intercepts the light, and finally comes to absolute dominance. It has to be mentioned that saturation for photosynthesis of Juncus bulbosus is already reached at 200 $\mu mol.CO_2.l^{-1}$ in the ambient water, while for Littorella still no saturation is reached at 2000 $\mu mol.CO_2.l^{-1}$ (17).

If the carbonate content of the bottom is somewhat higher Sphagnum is strongly favoured; under these conditions it forms a thick carpet, which suffocates the isoetid lawn on the bottom, probably by light interception, but other processes may be involved as well.

If the bottom does not contain any carbonate at all, Littorella is not succeeded by other species, when acidification takes place. The plants become however smaller and finally disappear. Littorella can live under acid conditions, as is obvious from laboratory experiments, but under natural circumstances the decomposition processes in the bottom stagnate and from the organic bottom material little or no CO_2 is released, thus Littorella peters out due to CO_2-starvation.

Juncus bulbosus and Sphagnum are stimulated during the acidification process by the CO_2 released from the $CaCO_3$ in the bottom. When all $CaCO_3$ has been dissolved and the CO_2-diffusion stops, the submerged forms of these two taxa can not maintain themselves. J. bulbosus remains only in shallow water, where it develops aerial leaves and becomes independent of the water for its CO_2-need. Sphagnum also survives only in contact with the air, and may overgrow the water surface. The submerged vegetation disappears completely under the influence of acidification, due to CO_2-limitation. The bottom of such waters is covered by a layer of dead plant material, of which the decomposition has been reduced or brought to a standstill, because the communities of bacteria show little activity or no activity at all at very low pH. The water itself is crystal-clear.

It can be concluded that the acidification of soft water systems leads to the deregulation of the CO_2-metabolism of these systems. Water and sediment loose all buffer capacity, the submerged water plants finally disappear, and the decomposition processes stagnate. The succession of Littorella-stands under influence of eutrophication and acidification is summarized schematically in Table I.

In the foregoing paragraphs the phenomena, observed in acidifying waters, have been completely ascribed to the effect of the increased number of H^+-ions. Via dry and wet deposition many other compounds enter the aquatic environment, such as sulphate, nitrate, ammonium and several other air pollutants. The acidification of the environment has also consequences for the solubility of all kinds of compounds in the soil, particularly heavy metals such as Al^{+++}, Cd and Fe.

Sulphate is mainly deposited on the sediment, and as long as the environment remains aerobic, this compound plays no role. On an average the sulphate content of substrates with Juncus bulbosus is twice that of a Littorella site; however the ranges for Juncus and Littorella are not significantly different (3).

Although orthophosphate is often considered the limiting factor in the

Table I. Succession of *Littorella*-stands under influence of eutrophication and acidification

Eutrophication:

Substrate enriched, water-layer poor in nutrients

Permanently submerged:

Littorella → *Myriophyllum alterniflorum*

Fluctuating water levels, sometimes very shallow or even drying up

Littorella → *Ranunculus peltatus*

Substrate and water-layer enriched

Small water-bodies:

Littorella → *Lemna minor* and/or *Riccia fluitans*

Large-sized water-bodies:

Littorella → phytoplankton blooms

Acidification:

Substrate with very low $CaCO_3$-content

Littorella → *Juncus bulbosus* and/or *Sphagnum* → †

Substrate without $CaCO_3$ at all

Littorella → †

oligotrophic soft water systems, it appears that little effects can be noted from the aerial input, which in areas with intensive cattle- and poultry breeding may be as much as 3 $kg.ha^{-1}$ yr^{-1}. Laboratory experiments also did not show any positive influence on the growth of macrophytes under acid conditions.

The effects of the deposition of nitrogen compounds are for various reasons much more important. In the first place the quantities of deposited nitrogen are very high. According to Leuven & Schuurkes (2) the wet deposition of N-compounds (mainly NH_x) amounts to 28-52 $kg.ha^{-1}$ yr^{-1}, in areas with intensive stockbreeding. According to Van Aalst (18) the dry deposition is almost twice that amount, so that the total amount of N-deposition may be of the order of 80-150 $kg.ha^{-1}$ yr^{-1} in areas of intensive stockbreeding.

In the second place there is a great difference in the use of various N-compounds by the diverse plant species. *Littorella uniflora*, *Lobelia dortmanna*, *Luronium natans* and *Echinodorus ranunculoides*, all characteristic species of the oligotrophic soft water environments, appear to be stimulated by NO_3^-, which they absorb mainly by the roots (19). *Juncus bulbosus* and *Agrostis canina* L. are greatly enhanced by the addition of NH_4^+, which they take up by the leaves. *Littorella* is able to take up NH_4^+, but in that case it looses K^+ (15). *Juncus bulbosus* is able to take up some nitrate, *Sphagnum* however only takes up NH_4^+, and no NO_3^- at all (19), while *Potamogeton gramineus* L. can use both without discrimination (19). It follows from these data that in the acidifying "vennen" *Juncus bulbosus* is highly stimulated by the NH_4^+ input, and that this is not favourable to *Littorella*.

In the third place the increased emission of ammonia from intensive cattle and poultry farms and the overmanured cropfields has contributed in the last decade to a considerable rise of the pH of the rainwater, which now is only slightly acid. However the precipitation has become acidifying because in aerobic environments the ammonium is nitrified by a community of various bacteria, a process resulting in the formation of $2H^+$-ions per NH_4^+-ion according to the following overall formula:

$$NH_4^+ + 2O_2 \rightarrow NO_3^- + H_2O + 2H^+$$

This means that the nitrification of ammonium accelerates the acidification, particularly if it is deposited as $(NH_4)_2SO_4$ (20). This process proceeds till a pH of 3.8 is reached; at this pH the activity of the bacteria becomes inhibited. When this pH has been reached further input of NH_4^+ does not lead to further acidification, but to NH_4^+ accumulation in the environment.
A further decrease in the pH can only take place by direct H^+ input from the atmosphere, and eventually by sulphur reduction in the substratum followed by oxidation of the released free sulphides. Sphagnum is able to lower the pH by little understood exchange processes. The lowest pH measured in The Netherlands was 2.8.

The strongly acidifying effect of $(NH_4)_2SO_4$ was demonstrated in an experimental laboratory set-up, in which a number of identical mini-ecosystems simulating "ven" conditions were treated with synthetic rain of various (but realistic) composition for more than a year. During this experiment rain with H_2SO_4 addition caused a pH decrease to 4.8, while the $(NH_4)_2SO_4$ rain caused a pH decrease to 3.8, which moreover was reached in a shorter time (20).

The heavy metal contents in the waters in the southern part of The Netherlands are very high, also in comparison to other countries, but are very similar to those found in the adjacent part of Belgium (2). The high contents are a consequence of industrial pollution in the past century, and are probably not linked with the present acidification. It appears, however, that the contents of 27 $\mu mol.l^{-1}$ Al^{+++} and 7 $nmol.l^{-1}$ Cd are not harmful to the aquatic plants, but adverse to several fauna components (2).

Little is known of the influence of acid and acidifying precipitation on hard water systems which are well buffered. In these waters the H^+ input is compensated for by dissolution of $CaCO_3$ from the bottom. Consequently the amount of $Ca(HCO_3)_2$ in the water increases, and one can speak of "water hardening" or "alkalinisation". In the coastal dunes, consisting of calcareous sands, a number of pools occur with a high $Ca(HCO_3)_2$ content, but otherwise very poor in plant nutrients. As a result of the increase of $Ca(HCO_3)_2$ the water has become very suitable for Potamogeton coloratus Hornem., a species known from a few records in the former century. Recently it has appeared in various dune pools (22). However, the atmospheric deposition of N and P will enrich these waters and it can be predicted that the more opportunistic P. gramineus in time will win the competition with the strictly oligotraphent P. coloratus.

Finally it may be concluded that the effect of acid or acidifying precipitation on the aquatic vegetation depends on the composition of the rain, and on the nature of the receiving water-bodies. These properties also determine the possible counter-measures that can be taken in order to neutralize the acidifying effects. In soft waters receiving mainly H_2SO_4-containing rain, lime addition may help to keep the acidification within certain limits. In soft waters, receiving $(NH_4)_2SO_4$-containing rain, not only acidification takes place but also eutrophication. Lime addition to these waters is not advisable. In hard waters acid rain causes alkalinisation, and if NH_4^+ is involved also eutrophication; this phenomenon needs more attention. Even in a small country as The Netherlands the composition of the rain shows considerable regional differences, and the effects on the aquatic vegetation depend greatly on the properties of the receiving water-bodies.

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BIOLOGY AND ECOLOGY OF MYRIOPHYLLUM AQUATICUM

David L. Sutton
Professor

University of Florida, IFAS, Fort Lauderdale Research and Education, 3205 College Ave, Fort Lauderdale, FL 33314.

Parrot-feather (Myriophyllum aquaticum (Vell.) Verdc.), a native from South America, is a common component of indoor and outdoor aquarium landscaping because of its aesthetic attractiveness and ease of cultivation. However, it has escaped from cultivation and is naturalized in many countries throughout the world. Vegetative propagation is an important means of spread since staminate flowers are absent on plants in areas where it has become naturalized. Although it will grow submersed, it is found primarily as an emersed plant. Parrot-feather is not considered a major noxious aquatic weed, but it may cause problems in shallow ditches and enclosed bodies of water. Management of its growth can be accomplished with herbicides such as (2,4 dichlorophenoxy)acetic acid (2,4-D) or by mechanical means.

The Haloragaceae family consists of eight genera and approximately 100 species (1). The genus Myriophyllum, or watermilfoil as this group of plants are commonly called, include about 40 species widely distributed around the world. About 13 species occur in North America of which 10 are indigenous. Many of the species are extremely difficult to distinguish from each other unless flowers or seeds are present because they exhibit very similar anatomical characteristics.

Several of the watermilfoil species are notorious for their ability to dominate a body of water. Eurasian watermilfoil (Myriophyllum spicatum L.) and parrot-feather (Myriophyllum aquaticum (Vell.) Verdc.) have received considerable attention because they cause weed problems in many areas where they occur.

An understanding of the manner in which the watermilfoil plants grow, develop, and spread, and their nutritional requirements will help those individuals involved with the management of aquatic weed problems formulate plans to control these plants. This review summarizes information on one of these species, parrot-feather.

CLASSIFICATION AND NOMENCLATURE

Parrot-feather was first named in 1829 by Cambessedes (2). A nomenclature change combined Myriophyllum proserpinacoides Gill. and Myriophyllum brasiliense Camb. under its present name of Myriophyllum aquaticum (Vell.) Verdc. (3). Parrot-feather is the preferred common name for plants of this species; however, other common names found in the literature are parrot's-feather, parrotfeather, and water-feather.

NATIVE HABITAT AND AREAS OF NATURALIZATION

Parrot-feather, a dioecious plant, is indigenous to South America. In a comprehensive study of *Myriophyllum* species, Orchard (4) found only a few male flowers, and only two plants with immature fruits, on specimens collected from the lowlands of central and western South America. However, female flowers were found on plants collected from a much wider area in South America. These were primarily from low altitudes but some specimens were collected from areas as high as 3,250 m in Peru and 1,900 m in Brazil.

Little is known of the factors which regulate growth of parrot-feather in its native habitat; however, it appears that a complex of insects have evolved which feed on this plant. For example, in Argentina, the flea beetle, *Lysathia flavipes* (Boheman) was found to cause moderate damage to parrot-feather under field conditions (5). Under laboratory conditions, both the larvae and adults of this flea beetle fed on the leaves. Also, a weevil, *Listronotus marginicollis* (Hustache), was reported to be host specific only to parrot-feather in Argentina (6).

Vegetative propagation, which is characteristic of many aquatic vascular hydrophytes (7), is an important means of plant invasion by parrot-feather outside its native range. It has spread to areas outside its native range primarily through human intervention. Colonization by seed appears to be rare, and in fact, may not occur at all. Even in its native range in South America, male plants are not common (8).

Parrot-feather has been introduced throughout the world for use as an ornamental in both indoor and outdoor aquaria landscaping because of its aesthetic attractiveness and ease of cultivation. It has escaped cultivation by transportation of fragments in flowing water and by man planting it in field sites. Parrot-feather is now naturalized in many areas, especially in the warmer climates of the world.

The exact date of introduction of parrot-feather in North America is unknown, but it was probably in the late 1800's or early 1900's. Since its introduction, this plant had become established along the east coast from Florida to New York, in the South and Southwest, in California, and it also occurs sporadically in other areas of the United States.

In Australia, parrot-feather was found around Melbourne between 1961 and 1969 (9). Only female flowers have been found in Australia with flowering observed in May, September, and November. Flowering is more frequent in the warm coastal areas. In some of the warmer parts of this country, parrot-feather has shown a tendency to become a problem plant. In New South Wales, parrot-feather is a declared noxious plant but it is seldom reported as a weed problem (10); however, areas with high amounts of nutrient in the water may experience nuisance levels of plant growth.

Parrot-feather was first reported in New Zealand in 1929 (8).

Although it is found in a number of locations throughout this country, it is especially troublesome in the Manawatu drainage system. Only female plants are present, and spread of this plant is by vegetative means.

In Java, parrot-feather has naturalized in ditches and rice fields (9). It is cultivated by the Javanese as a protective cover for use in fish culture ponds (9) and the tips of the shoots are eaten as a vegetable (11).

In Japan, parrot-feather was found in 1920 in the Wakayama and Hyogo Prefectures, and it is now considered among the top six most injurious aquatic plants found there (12). Parrot-feather is particularly troublesome in shallow irrigation channels. The emersed plants are killed by the winter temperatures in Japan but the plants regrow from submerged stems in the spring. Only female plants are known to be there, hence, all new growth is by vegetative means.

In Europe, parrot-feather has been for years a favorite plant for use in aquaria. In 1976 it was found growing in a pool in West Cornwall, England (13). This was thought to be the first recorded instance of this plant growing in England since it was generally assumed that the weather there was too cold to allow for good growth of the plant outdoors; however, it apparently had been growing for several years prior to 1976 in a stream flowing from a park at Penzance. Only female plants have been found in England.

Parrot-feather was imported for use as an ornamental for fish ponds and aquaria in Southern Africa around 1918 to 1919 or perhaps slightly earlier with the first recorded specimen collected from Paarl, Western Cape Province (14). Since that time it has become more abundant than any of the indigenous species of *Myriophyllum* found in that area. Along with waterhyacinth (*Eichhornia crassipes* (Mart.) Solms), kariba weed (*Salvinia molesta* Mitchell), and the water fern *Azolla filiculoides* Lam., parrot-feather is one of the four dominant exotic aquatic weeds affecting the fresh water supplies, principally rivers and farm ponds near them, in Africa south of the Kunene and Zambezi River (15). Only female plants are present and they flower mainly during late winter and early summer.

MORPHOLOGICAL AND ANATOMICAL CHARACTERISTICS

Parrot-feather derives its name from the almost feather-like leaves which occur on this plant. The leaves are arranged in whorls of 4 to 6 around the stem. The emersed leaves are dark green and quite stiff as compared to the submersed leaves which are lighter green and more filamentous. Each leaf may have 20 or more linear-filiform divisions which gives it a feather-like appearance (16). The leaves, oval in transverse section and covered by an epidermal layer of irregularly shaped cells, are pinnately compound with a single vascular bundle, surrounded by a bundle sheath, in each leaflet (17).

The presence of emersed stems is one of the readily identifying characteristics of parrot-feather (16). These stems, generally with few branches, grow up to 30 cm or more above the surface of moist, muddy areas, or they may be attached to the bank and extend out several meters over the surface of the water. In shallow bodies of water, it is not uncommon to have submersed and emersed leaves on the same stem; however, under these conditions, the emersed portion tends to lay on the surface or extend only a few centimeters above it.

Parrot-feather is dioecious with flowers borne in the axils of the emersed leaves (16). The pistillate flowers, which are the most common since almost all naturalized populations contain female plants only, are about 1.5 mm long and appear as a tuft of white or pinkish plumose stigma lobes (18). Little is known of the appearance of male flowers, fruit, and seed. Essentially no information is available on factors affecting pollination, fruit set and development, and seed germination since the presence of male flowers is rare.

As with most aquatic macrophytes, air canals and aerenchyma are abundant in parrot-feather plants (17). Elongated air canals, radially arranged in the cortex of the stem internode, are present in the emersed portion of the plant. The air canals are interrupted at the nodes in the stem but aerenchyma is continuous from one end of a root or leaf to the other. The stem may contain 24 to 30 air canals per internode. The air canals are formed by the extension of a single layer of parenchyma cells from just under the epidermal layers of the stem to its center. This layer of cells has a storied appearance when excised plant material is viewed in longitudinal section.

Druse crystals, which occur in several genera of the Haloragaceae family, are abundant primarily on the exterior walls of the air canals in parrot-feather (17). These crystals may also be found inside the parenchyma cells, but when this occurs, only one crystal is present in each cell. Druse crystals, although less abundant than in the stems, are present both inside the cells and the intercellular spaces in the mesophyll of the leaves.

Anomocytic, sunken stomata are present on all surfaces of mature, aerial leaves of parrot-feather (17). Epidermal cells over-arch the stoma, and at times, almost close the pore opening. Spongy mesophyll occurs under the epidermal layer and is filled with numerous large air cavities. The submersed leaves do not have stomata but simple perforations occur in their leaflets which may allow water to escape slowly in response to excess root or turgor pressure (19).

The stem contains five to six collateral vascular bundles with the phloem located abaxially to the xylem (17). The center of the stem is composed of pith with primarily large parenchyma cells filled with starch granules. An endodermis is normally found in the roots of angiosperms (20); however, in the stem of parrot-feather, the pith and vascular bundles are surrounded by an endodermis characterized by the presence of a Casparian strip on the radial walls. The stem is surrounded by a single layer of collenchyma cells which appear to provide some structural support for the plant above the water.

The xylem of parrot-feather contains both annular and helical secondary wall sculpturing with a simple perforation on the end wall of each cell (17). The xylem also has narrow lumina, and, where in contact with rays, bordered pits. The phloem is located in isolated strands of two to six in number.

Arbor (21) postulates that the failure of some submersed aquatic macrophyte roots to produce root hairs may be related to a cuticularization of its epidermis. The submersed roots of parrotfeather appear to contain large amounts of cutin or suberin, or both. Root hairs may be found on roots which grow above the water but not on the submersed ones (17). Roots grow from nodes of the stems within 1 week when the lower portion of excised apical sections of the stem are submersed (22).

GROWTH AND DEVELOPMENT

Growth of parrot-feather plants may be extremely luxuriant under some conditions. Although it has naturalized in a number of locations throughout the world, parrot-feather generally is not considered a major noxious weed problem in many of them. Under natural conditions, it appears to prefer a warm climate, muddy banks or shallow bodies of water, and tends to grow in isolated patches primarily as the emersed form.

Parrot-feather has been found in static and flowing water up to 2.0 m deep in New South Wales (10). In that area it appears to grow best when high levels of nitrogen are present in the water. Also, the plant grows well there at temperatures from a low of 8 C to a high of 30 C.

Parrot-feather has adapted well to climatic conditions in California where it is becoming an increasing problem in irrigation and drainage canals (23). In shallow rivers and streams in the southern portion of Africa, parrot-feather may become so dense as to cause flooding and drainage problems (24). Also, it may interfere with recreational activities in these bodies of water.

Oxygen requirements in the root zone of emersed hydrophytes has been found to be different for various species (25-26). With some aquatic plants there are increases in growth and structural changes in plant morphology associated with aeration in the root zone. In some emersed aquatic plants, oxygen can diffuse from the root tips (27), indicating that oxygen can move through the root supplying the tissue with oxygen needed for respiration under anaerobic conditions. Oxygen in the roots is probably from photosynthesis, since it has been found to increase when shoots are exposed to light (24).

Aeration of full and one-tenth strength Hoagland's nutrient solution (28) did not influence dry weight of emersed parrot-feather plants cultured for 6 weeks under greenhouse conditions (29). Dry weight however, was lower for plants cultured in the one-tenth strength nutrient solution under these conditions as compared to similar plants in one-half or higher concentrations of nutrient solution. These data suggest that high concentrations of nutrients are required for good growth of parrot-feather under field conditions. Growth in nutrient enriched situations, such as that caused by sewage effluent, may lead to problem amounts of parrot-feather plants.

Little information is available on growth and accumulation of biomass of parrot-feather under field conditions. However, the influences of several nutrients on its growth and development have been evaluated under controlled environment conditions. Also, this plant has been used as a test aquatic species to determine uptake and translocation of various mineral elements and organic materials.

Boyd (30) found the crude protein in parrot-feather ranged from

12 to 18% on a dry weight basis for plants collected during a survey of 43 aquatic species from various areas in Alabama and Florida. This high amount of crude protein indicates the potential of parrot-feather as a possible feed for livestock and other animals. These plants also contained more that 6% tannin.

Barko and Smart (31) found that under controlled environment conditions, the submersed form of parrot-feather plants readily absorbed nitrogen and phosphorus from three sediments collected from separate locations and concentrated these nutrients in the shoots at levels well above those required for growth. These plants initially contained 8 mg of nitrogen and 1 mg of phosphorus per g of plant dry weight, after 10 weeks of growth these values increased to 33 to 36 and 3 to 5, respectively, for plants grown on the three different sediments types. Potassium decreased from an initial value of 6 mg per g of plant dry weight to 2 to 5 for the three sediments. The authors of this study theorized that potassium may have been limiting in the sediments, and thus the cause for the reduction in potassium levels after 10 weeks of growth. They suggested that because of the high root to shoot ratios found in this study, the submersed plant of the heterophyllous parrot-feather can be considered as an intermediate form between strictly submersed and emersed aquatic macrophytes.

The role which the roots play in the uptake of nutrients by aquatic macrophytes has been a subject of discussion by botanists for years (7,21) with one school of thought maintaining that the roots provide the major port of entry of nutrients by absorbing them from the sediments, while others suggest that the bulk of the nutrients are absorbed by the leaves. Parrot-feather has been used in several studies designed to provide some information on this topic.

Bristow and Whitcombe (32) reported that when P^{32} was supplied to parrot-feather plants cultured in a two-compartment apparatus designed to allow for the upper and lower portions of the stem to be kept in different nutrient solutions, over 90% of the phosphate was derived from the rooted stem portion. Furthermore, they found the amount of phosphate translocated was related to the mass of roots present. An interesting observation made during this study was that normal formation of axillary shoots occurred even though all the mineral ions were supplied by translocation from the lower compartment for their growth.

The concentrations of phosphorus on a dry weight basis were found to be significantly higher in the roots than in the shoots for emersed plants cultured in one-half strength Hoagland's No. 1 nutrient solution for 1 week under greenhouse conditions (33). The roots contained 0.7% phosphorus as compared to 0.6% in the shoots. After 2 weeks of growth, the phosphorus content of these plants was essentially the same as that found after the 1 week of growth indicating a lack of uniform distribution of phosphorus in emersed parrot-feather cultured under these conditions. Few measurements are available on the distribution of mineral elements in parrot-feather grown under field conditions.

Copper was found to be uniformly distributed throughout the shoot and roots of parrot-feather at an average concentration of 16 ppm for emersed plants cultured under greenhouse conditions (33). When these plants were exposed to 0.25 to 16.0 ppm of copper as copper sulfate pentahydrate in the root zone, copper concentrations of 52 to 6,505 ppm were found in the roots. In this study only a slight movement of copper from the roots to the shoots occurred due to an increase in copper in the treatment solution and an increase in contact time. The majority of the copper taken up by the roots occurred within the 1st week of exposure to the copper sulfate pentahydrate solutions. Growth of the plants as measured by dry weight was reduced by exposure of the roots to 4.0 ppm of copper for 1 week, but after 2 weeks of growth, dry weight was reduced for plants treated with concentrations of 1.0 ppm or higher of copper.

In a major study on the carbon dioxide gas exchange of parrot-feather, aerial leaves of this plant were found to have a much higher photosynthetic rate than the submersed ones but the dark respiration rates of the different leaves were the same (34). An irradiance of 2,000 $\mu E/m^2 X s^{-1}$ was required for saturation of net photosynthesis of the emersed leaves but the submersed ones required only 250 $\mu E/m^2 X s^{-1}$. The emersed leaves were found to exhibit characteristics similar to that of sun-adapted leaves of terrestrial plants while the submersed ones were similar to that of other submersed aquatic macrophytes.

In a study to determine the influence of salinity on growth of aquatic macrophytes, parrot-feather was found to be one of the most tolerant of 10 plants exposed to various concentrations of seawater (35). Under greenhouse conditions, seawater was toxic to growth of the emersed form of parrot-feather at salt concentrations between 10.0 and 13.2 parts per thousand (ppt), but root growth was stimulated by salt concentrations of 0.8 to 3.3 ppt. Since all concentrations of seawater reduced transpiration of the plants compared to the pond water controls, the increase in root growth was probably a response of parrotfeather to overcome a water deficit by an increase in the surface area of the roots. These data indicate the ability of parrot-feather to survive in coastal areas where salt intrusions may occur, and thus they may grow to the exclusion of other more salt sensitive aquatic macrophytes.

MANAGEMENT OF GROWTH OF PARROT-FEATHER

Herbicides. Copper at a concentration of 1.0 ppm or higher in the root zone was found to reduce growth of parrot-feather under greenhouse conditions (33). Under field conditions, the use of copper sulfate would probably not be practical as it would be difficult to maintain this concentration for the period of time necessary to kill the plants.

Combinations of certain herbicides have been shown to be more effective on some aquatic plants than the individual compounds (36-37). When the roots of emersed parrot-feather plants were placed in solutions of copper sulfate pentahydrate at 1.0 ppmw of copper plus either 0.5 or 5.0 ppmw of 7-oxabicyclo [2.2.1] = heptane-2,3-dicarboxylic acid (Aquathol) (Mention of a trademark name or a proprietary product does not constitute a guarantee or warranty of the product by the University of Florida and does not imply its approval to the exclusion of other products that also may be suitable.), the copper content of the roots was increased by 27 and 150 ppmw, respectively, over that of plants exposed to only the copper sulfate pentahydrate (38). In this study, these concentrations of Endothall reduced transpiration and inhibited growth of parrot-feather; however, the phytotoxic effect of the Endothall was not enhanced by the addition of 1.0 ppmw copper. Additional studies are needed on the effectiveness of herbicide combinations for management of parrot-feather.

Transpiration of emersed parrot-feather was significantly higher 4 days after treatment with 0.12 and 0.50 ppm of 2-chloro-4,6-bis(ethylamino)-s-triazine (simazine) as compared to control plants (22). The reason for this increase may have been related to an increase in surface area of the plants due to increased growth, since it is known that simazine can stimulate growth in certain plants under appropriate conditions (39). However, other explanations such as alteration of stomatal behavior cannot be precluded.

Simazine at 0.12 ppm resulted in a 40% reduction in dissolved oxygen in treatment solutions 24 h after treatment of the submersed form of parrot-feather under greenhouse conditions (22). After 2 days of exposure to the simazine, dissolved oxygen in the water with plants exposed to the 1.0 ppm solutions was reduced by almost 90%. In this study, dissolved oxygen was reduced to a greater extent in the treatments with parrot-feather than with either common duckweed (*Lemna minor* L.) or elodea (*Elodea canadensis* Michx.) indicating the usefulness of simazine as a potential herbicide for the control of the submersed growth forms of parrot-feather.

Growth of the emersed form of parrot-feather was inhibited by root applications of simazine at concentrations greater than 1.0×10^{-7} M (40). The amount of simazine in the upper shoot portions of the plant was closely related to transpiration. Initially, 1 and 2 h after exposure of the roots to simazine, the roots contained the highest amount of herbicide but after 192 h the shoot portion contained a much higher amount than the roots. Simazine was found to be taken up passively by the emersed form of parrot-feather as a correlation coefficient of 0.988 was calculated when the amount of simazine in the plant tissue was related to the treatment concentration over a range of 1.2×10^{-6} to 5.0×10^{-6} M. Simazine is not toxic to some plants because they deactivate this herbicide by converting it to hydroxysimazine; however, it appears that parrot-feather does not have a mechanism for deactivation of the simazine molecule. These data indicate the sensitivity of parrot-feather to simazine.

Exposure of roots of emersed parrot-feather to 0.02 ppm of 6,7 dihydrodipyrido[1,2-a:2',1'-c] pyrazinediium salt (diquat) resulted in a decrease in dry weight as compared to control plants (41). Older plants appeared more susceptible to diquat than young plants. Diquat was not translocated in parrot-feather, and its uptake was not increased by dark conditions. Darkness has been shown to result in translocation of diquat in some plants (42).

Uptake of diquat by parrot-feather appears to occur primarily by passive adsorption processes. Davies and Seaman (43) reported that metabolic accumulation was one of the mechanisms involved in the long term uptake of diquat-^{14}C in elodea; however, the metabolic inhibitor, dinitrophenol (DNP), did not increase uptake of diquat after application of this herbicide to the roots of emersed parrot-feather plants (41). A reduction of energy in the root cells by the DNP did not result in movement of diquat-^{14}C from the root portion of the plant to the shoot by way of the transpiration stream. Although emersed parrot-feather plants have a strong transpiration stream, diquat apparently did not enter the xylem where translocation could occur.

Control of parrot-feather may be achieved with (2,4-dichlorophenoxy)acetic acid (2,4-D). Blackburn and Weldon (44) suggested that the low-volatile ester of 2,4-D at rates of 4.4 to 8.9 kg per ha (4 to 8 pounds per acre) when sprayed on the leaves of emersed parrot-feather resulted in good control of this plant. Also, the granular formulation of 2,4-D was found to control parrot-feather for a year or more but no rate was given (45). Hall and Hess (46) found parrot-feather to be among 19 plants hypersensitive to 2,4-D based on studies with a number of formulations and various application rates for 63 species of plants.

When 2,4-D-1-^{14}C was applied to the emersed leaves of parrot-feather, autoradiographs showed an accumulation of labeled material occurred in the nodes accompanied by basipetal movement of carbon-14 to the roots (47). These data show that foliar applications of 2,4-D will result in translocation of the herbicide to the root zone so that growth of both the shoot and root portions of the plant will be affected. When the roots of the plant were exposed to the labeled 2,4-D, no radioactivity could be detected in the shoots, but use of the metabolic inhibitor, DNP, applied concomitantly with the 2,4-D-1-^{14}C resulted in acropetal movement in the transpiration stream of labeled material to the shoots. Although root applications of 2.5 X 10^{-6} M or higher of 2,4-D was found to inhibit growth of emersed plants, metabolic energy was necessary to provide for uptake and subsequent translocation of this herbicide.

The uptake and movement of 2,4-D in parrot-feather followed that suggested by Crafts (48) who proposed that 2,4-D or its metabolites enter the symplast of plants and is then translocated in the phloem with endogenous assimilates. Therefore, it appears that when 2,4-D is applied to young, actively growing parrot-feather plants better control will be achieved than when older, mature plants are treated with this herbicide.

Biological. Little attention has been given to the biological control of parrot-feather. The flea beetle, *L. flavipes*, and the weevil, *L. marginicollis*, have been found to cause some damage to parrot-feather, although it is not known if these insects are the primary organisms which limit growth of this plant in its native habitat.

Habeck and Wilkerson (49) found under laboratory conditions that parrot-feather was a natural host for larvae of the flea beetle *Lysathia ludoviciana* (Fall). This flea beetle is indigenous to the southern United States and the Caribbean region, and has been collected on at least 17 different plants. However, the scarcity of this flea beetle on parrot-feather plants in field sites suggest that the beetle may have additional hosts. The potential of L. ludoviciana has yet to be realized; however, these flea beetles have been collected on parrot-feather plants growing outdoors in Florida. Also, two species in the tortricidae family, Choristoneura parallela (Robison) and Argyrotaenia ivana (Fernald) were found to be present on parrot-feather during a survey for interactions of Lepidoptera and Polygonum spp. in north central Florida (50). Little is known of the feeding activity of these insects on parrot-feather.

Newly hatched larvae of the caterpillars Parapoynx allionealis Walker preferred parrot-feather leaves over Sagittaria stagorum Small, Eleocharis vivipara Link, and duckweed Lemna sp. (51). The first stage larvae mined the leaves of parrot-feather but the extent of damage which this caterpillar could cause to parrot-feather is not known.

A Rhizoctonia solani isolate (RhEa) collected from diseased anchoring waterhyacinth (Eichhornia azurea (Sw.) Kunth) plants in Panama was phytotoxic only to the tips of submersed parrot-feather which protruded above the surface of the water (52). In northern California, an isolate of Pythium carolinianum Matt. was collected from diseased emersed parrot-feather plants which showed a severe rot of underwater roots and stems (23). When mycelium of this isolate was inserted into the stems of parrot-feather cuttings, the fungi girdled the stems and caused collapse of the plants. Additional testing under field conditions showed that growth of parrot-feather plants inoculated with this isolate was lower than uninoculated controls. These studies showed that various fungi have potential for biological control of parrot-feather.

Mechanical. Excessive growth of parrot-feather may be removed by mechanical means. Little information is available however on the regrowth of plants after their removal. Since fragments will form shoots and roots under appropriate growing conditions, regrowth will occur vegetatively from them as well as any plants not removed from the harvested site. Jacot Guillarmod (24) reported difficulty in mechanically removing infestations of parrot-feather because it broke into fragments readily and many small portions of plant material could not be removed. Those plants and fragments not harvested quickly produced new growth. Studies are needed on ways to mechanically manage parrot-feather with emphasis on methods which will reduce the likelihood of reinfestation from plant fragments.

CONCLUSIONS

Parrot-feather in general may be considered more a beneficial than a noxious plant. It provides shelter and protective cover for a number of aquatic organisms, and may serve as a feed source for some herbivorous organisms. In some countries it is eaten as a vegetable by humans. Parrot-feather is an especially attractive plant for aquaria and is easy to cultivate. This plant removes nutrients, and help ameliorate wind and wave action. In some shallow bodies of water, ditches, and small rivers, parrot-feather may present serious problems when it reduces water flow and other water uses. Studies are needed on management methods which will provide effective control of parrot-feather for situations when it becomes a problem.

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SEDIMENT COMPOSITION: EFFECTS ON GROWTH OF SUBMERSED AQUATIC VEGETATION

John W. Barko and R. Michael Smart
Environmental Laboratory, Waterways Experiment Station,
Vicksburg, Mississippi 39180, U.S.A.

Sediment composition markedly affects growth rates of submersed aquatic vegetation. Hydrilla verticillata (L.f.) Royle and Myriophyllum spicatum L. grow best on fine-textured inorganic sediments with an intermediate sediment density, and poorly both on sands with a high sediment density and on organic sediments with a low sediment density. Changes in sediment density due to experimental manipulations, with or without change in sediment organic matter content, promote major changes in growth rates. Thus, the density of organic sediments, and not organic matter content per se, seems to affect the growth of rooted submersed aquatic vegetation. Mechanisms of growth regulation on sand and on organic sediments appear to be similar, and to involve nutrition. Owing to variations in the sensitivity of different aquatic macrophyte species to sediment properties affecting growth, it is postulated that alterations in sediment composition may contribute fundamentally to vegetative changes in aquatic systems.

Sediments provide an important source of nutrients, principally nitrogen, phosphorus and micronutrients (3, 12, 23), which are relatively less available to submersed macrophytes in the overlying water of most aquatic systems. The physical and chemical composition of sediments markedly affects macrophyte growth rates, and different species appear to vary in their response to sediment conditions (1, 2, 5).

Specific differences in responsiveness to sediment conditions may influence the species composition of aquatic macrophyte communities. Macrophyte community composition and the spatial distribution of individual species have been correlated with sediment organic matter content (14, 18, 21). Moreover, there is an apparent association during lake aging between increasing sediment organic matter and the decline of rooted submersed aquatic vegetation (8, 24, 25). These observations, considered collectively, suggest that the effect of sediment on submersed macrophytes may in part be related to sediment organic matter content. We earlier demonstrated that additions of organic matter to a fine-textured inorganic sediment can substantially reduce the growth of submersed macrophytes (5), but the mechanisms involved, or their applicability to growth on unaltered sediments remain unknown.

Here we summarize results of three years of experimental work involving the growth of Hydrilla verticillata (L.f.) Royle and Myriophyllum spicatum L. on a variety of different sediments from geographically widespread North American lakes. Our purpose in this article is to better elucidate relationships between growth of submersed macrophytes and sediment composition, in an attempt to increase practical understanding of the role of sediment in affecting the distribution and growth of nuisance macrophyte species. Detailed aspects of this work are elaborated elsewhere (6).

Approach

Surficial sediments were obtained with small hand-held dredges from 40 sites among 17 lakes. Lakes and sites within lakes were selected to span a broad range in sediment texture and organic matter content. Refer to (6) for details of sediment composition. Growth experiments were conducted under partially controlled environmental conditions in a greenhouse. Growth was determined as total (roots + shoots) biomass. The greenhouse facility, housing 18 large (1200 ℓ) macrophyte culture tanks and ancillary equipment, is described elsewhere (4). Light was restricted, using neutral-density shade fabric, to 50% full daylight (about 1000 $\mu E/m^2/s$ during the summer). Water temperature was maintained at 25 ± 1°C. Solution chemistry was nearly identical to that described in Table 1 of (23), except for the addition of $Ca(NO_3)_2$. The solution contained (in mg/ℓ): 32.2 Ca^{+2}, 6.8 Mg^{+2}, 16.0 Na^{+1}, 6.0 K^{+1}, 26.9 SO_4^{-2}, 44.2 Cl^{-1}, 22.1 NO_3^{-1}, and 51.8 HCO_3^{-1}. Phosphorus and micronutrients were excluded from solution to minimize algal growth in the case of the former, and because of difficulties in maintaining solubility of the latter. It was assumed that those elements excluded from solution would be obtained from sediments by root uptake (3, 9, 12, 23).

Growth in Relation to Sediment Composition

The growth of both species decreased with increasing organic matter content up to about 20%. At higher values of organic matter growth was rather uniformly reduced to the lower end of the growth range (Figure 1). At relatively low values of organic matter (<10%) sediments with greater than 75% sand (excluded from the relationship in Figure 1) also provided poor macrophyte growth. Growth on inorganic "sands" was diminished at high sediment densities while growth on "organic" sediments was diminished at low sediment densities (Figure 2). Over the range of 0 to 20% organic matter, sediment density (Figure 3) as well as macrophyte growth (Figure 1) declined with increasing organic matter content. From regression analyses, growth was somewhat better related to sediment density than to organic matter content. In close agreement with data reported for marsh soils (11) sediment density was determined nearly entirely by mineral density. Thus mineral mass contributed directly to the density of sediments with an organic matter content of less than about 20%.

Sediment Manipulations

In order to more directly examine the influences of sand, organic matter, and sediment density on macrophyte growth, separate experiments involving sediment manipulations were conducted. Results of these experiments are summarized in Table I. Additions of a fine-textured inorganic sediment (at 40% by volume) to both an organic sediment and to a sand increased the growth of *Hydrilla* dramatically, 3-fold on the organic sediment and 7-fold on the sand. These increases in growth accompanied a decrease in the organic matter content and an increase in the density of the organic sediment, and a decrease in the sand content and decrease in the density of the sand. Centrifugation of the organic sediment promoted an increase in sediment density similar to that achieved by additions of fine-textured inorganic sediment, and approximately doubled the growth of *Hydrilla*. In this latter experiment, sediment texture and organic matter content were unaffected by treatment. Thus

Table I. Summary of Results from Sediment Manipulations

Base Sediment	Treatment	Δ Organic	Δ Sand	Δ Density	Growth[a]
Organic	+ Inorganic	Decrease	--	Increase	Increase
Sand	+ Inorganic	--	Decrease	Decrease	Increase
Organic	Centrifugation	No change	--	Increase	Increase

[a] All increases in growth were statistically significant at $p < 0.05$.

sediment density or related factors, rather than organic matter content *per se* affected growth.

Nutritional Experiments

Nutrient uptake by macrophytes directly paralleled density in this investigation (6), and in this regard we considered that density affected macrophyte growth by influencing nutrient availability.

In order to examine directly the possibility that nutrition affected macrophyte growth on organic sediments, a series of experiments involving additions of P and Fe to sediments was conducted. Six organic sediments were obtained for experimental purposes from separate collections over a two-year period at or near sites of original collection. Phosphorus and Fe were added to selected sediments separately and in combination as $CaHPO_4$ at 0.1 g/l wet sediment and as Fe_2O_3 at 5.0 g/l wet sediment, respectively. In these experiments neither sediment organic matter content nor sediment density were affected by nutrient additions.

Hydrilla did not respond to the addition of Fe alone, and responded to the addition of P alone on only one of four sediments so treated (Table II). The growth of this species increased significantly, however, on both sediments amended by P in combination with Fe. In another separate experiment the effect on *Hydrilla* growth of P & Fe additions to an organic sediment (as above) was examined in a nitrogen-free solution. In that experiment, the combined addition of P and Fe had no effect on *Hydrilla* growth (data not presented), strongly suggesting a general nutrient inadequacy, i.e., multiple nutrient limitation on low density (organic) sediments.

The positive response of *Hydrilla* to "fertilization" by addition of fine-textured inorganic sediment to both sand and organic sediment (Table I), in combination with results achieved by specific nutrient additions above, indicates that the regulating effect of sediment sand and (or) organic matter content on the growth of submersed aquatic vegetation directly involves nutrition. We suggest that nutrient uptake on low density, high porosity organic sediments was limited by long diffusion distances. Indeed, increased density, and presumably decreased diffusion distances, resulted in the enhanced growth of *Hydrilla* on centrifuged organic sediment (Table I). Limited rates of nutrient diffusion and exchange in sands probably explains their poor ability also to support the growth of submersed macrophytes. In this regard, the availability in sediments of N, P, Fe and possibly other micronutrients is of critical importance, because of the significance of

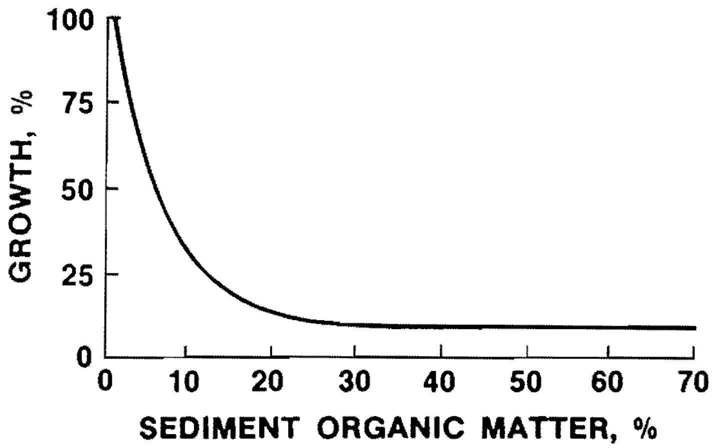

FIGURE 1. Generalized relationship between the growth of Hydrilla and Myriophyllum and the organic matter content of fine-textured sediments.

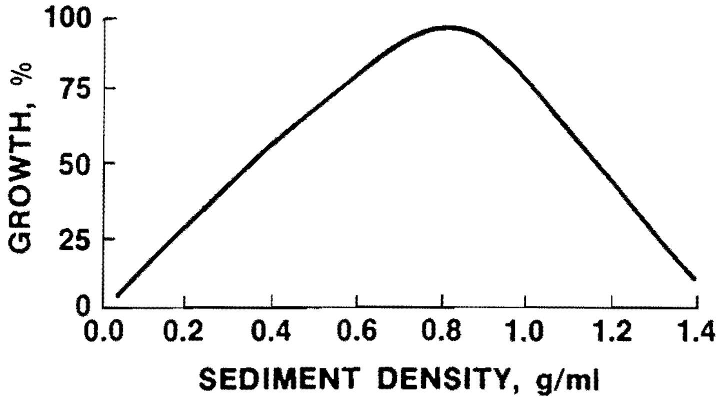

FIGURE 2. Generalized relationship between the growth of Hydrilla and Myriophyllum and sediment density. Density increase up to about 0.9 g/ml reflects decreasing sediment organic matter content. Sediments with a density > 0.9 g/ml are predominantly coarse-textured.

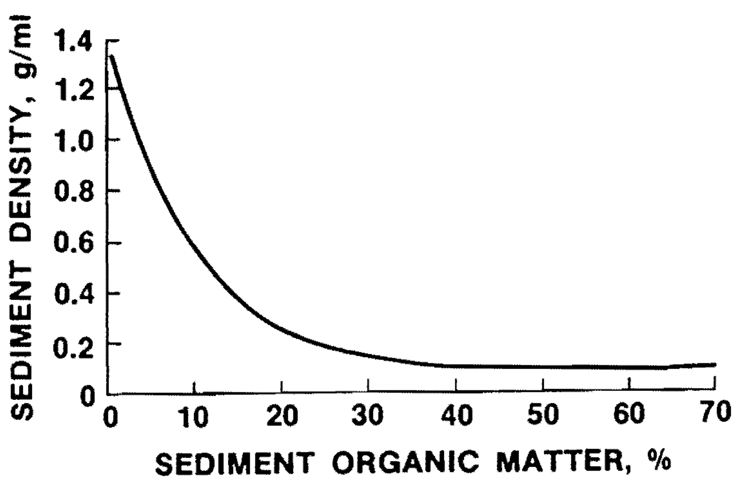

FIGURE 3. Generalized relationship between sediment density and sediment organic matter.

Table II. Effects of Nutrient Additions to Organic Sediments on the Growth of Hydrilla

Sediment	Treatment Addition	Growth Increase[a]
ORG-1	P	No
ORG-2	P	No
ORG-3	P	No
ORG-4	P	Yes
ORG-5	Fe	No
ORG-6	Fe	No
ORG-5	Fe + P	Yes
ORG-6	Fe + P	Yes

[a] Yes indicates significantly greater growth at $P < 0.05$.

sediment rather than the open water as the predominant source of these elements in most aquatic system (cf. 23).

Conclusions and Related Implications

The growth of submersed macrophytes is relatively poor on both highly organic sediments and sands compared to that on fine-textured inorganic sediments. Poor growth on sands is related to high sediment density, and on organic sediments, to low sediment density. Mechanisms of growth regulation on sand and organic sediments are similar, both involving nutrition. High concentrations of organic matter in sediments exert a negative influence on the growth of submersed macrophytes, by reducing sediment density and the associated availability of essential nutrients (notably N, P and Fe).

In nature, individual differences in the ability of aquatic macrophyte species to cope with sediment infertility or other factors associated with high sand and (or) high organic matter fractions may be important in determining the species composition of aquatic macrophyte communities. In this connection, it is notable that submersed aquatic macrophytes, particularly invasive species, are replaced by other vegetation as sediment organic matter accumulates (7, 8, 24, 25).

Sediment composition is an intrinsic component of the regional environment, but is amenable to manipulation. Various sediment covers, including sand, gravel, and plastic liners have been used in attempts to control the production of submersed macrophytes by altering sediment texture and reducing sediment nutrient uptake (10). Alternatively, dredging has been employed to both remove nutrient-rich sediments (19, 22) and to expose nutrient-poor underlying substrata, e.g., sand and gravel (10). Considered collectively, these efforts have indicated reductions in macrophyte productivity, and in nearly all cases, dramatic shifts in the species composition of submersed macrophyte communities.

We are aware of one documented occurrence of a decline in rooted submersed macrophytes following a major loading of organic matter due to watershed disturbance (13, 1). Conversely, the growth of submersed aquatic vegetation on organic sediments may be stimulated by additions of inorganic sediment (Table I). Sediment composition may be modified by aquatic plants themselves, directly, by sediment nutrient uptake (2) and contributions of their own remains to the sediment (8, 24, 25) and, indirectly, by collecting externally-loaded materials (15, 16, 17, 20). In view of these findings, we suggest that watershed disturbances, direct mechanical disturbances of bottom sediments, or autogenic processes affecting the inorganic/organic composition of sediments (and thus, sediment density and fertility) may contribute fundamentally to vegetational changes in aquatic systems.

Acknowledgments

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FACTORS ASSOCIATED WITH THE DISTRIBUTION OF *MYRIOPHYLLUM* IN BRITISH COLUMBIA

PATRICK D. WARRINGTON

Supervisor, Biological Studies
Resource Quality Section
Water Management Branch
Ministry of Environment, Parliament Buildings
Victoria, British Columbia
Canada V8V 1X5

The geographic distribution of the nine species of *Myriophyllum* L. known from British Columbia is shown on maps. Tables and figures show the distribution as a function of altitude, ecological zone, pH of the water, lake sediment chemistry and lake water chemistry. Some mechanisms of *Myriophyllum* spread within British Columbia are discussed.

The exceptionally wide diversity of habitats in British Columbia, created, in part, by variations in altitude, latitude, geology and proximity to the Pacific Ocean, offers a valuable opportunity to investigate the effects of such variables on the distribution of aquatic plants. The effects of these geographic factors on the distribution of terrestrial vegetation is fairly well known (2) but similar knowledge for aquatic plants is not available.

Nine species of *Myriophyllum* are known from British Columbia and listed in Table I. These are all readily distinguished by 2-dimensional thin layer chromatography of their flavenoid pigments (1). This technique may be needed to confirm species identifications of herbarium, fragmentary or non-flowering material but is rarely needed to identify living material in the field.

Table I. *Myriophyllum* species found in British Columbia

1. *M. aquaticum* (Vell.) Verd.
   M. *brasiliense* Camb.
2. *M. exalbescens* Fern.
3. *M. farwellii* Morong.
4. *M. heterophyllum* Michx.
5. *M. hippuroides* Nutt.
6. *M. quitense* H.B.K.
   M. elatinoides Gaud.
7. *M. spicatum* L.
8. *M. ussuriense* (Regel) Maxim.
9. *M. verticillatum* L.

Materials and Methods

For several years the author has been building a computerized dataset on the aquatic plants of British Columbia. For each lake or pond in which aquatic plants have been found, or in which they have been sought, the following information is stored, when available: aquatic plant presence or absence, water chemistry, sediment chemistry and lake morphometry parameters, growing season water temperature, drainage basin information, geographical information and the Biogeoclimatic zone (2) in which the lake is found. Similar, but less comprehensive, files exist for rivers and streams, wetlands, sloughs and ditches and park and garden pools. Data from these latter files were included to compile the distribution maps, Figures 1 and 2 and Tables 2 and 3. Figures 3 to 9 were compiled only from the lakes and pond data set since the others lack sediment and water chemistry data.

M. exalbescens Fern. has been found in 318, and *M. verticillatum* L. in 100 bodies of water. These are widespread and common species in British Columbia and sufficient water and sediment chemistry data exist to justify statistical analyses for these two species. There are not sufficient data available for the other species of *Myriophyllum*. *M. spicatum* L. has been found in 60 bodies of water in British Columbia but few of these have had water and sediment samples taken. However, due to the importance of this species as a weed, the existing data have been analyzed and tentative associations are presented.

Results and Discussion

Geographical Distribution

Figures 1 and 2 are distribution maps showing the locations in British Columbia where each species has been recorded. These maps were prepared by putting one dot in the centre of each 1:50 000 topographic map sheet where at least one distribution record exists. Some data in map sheets with many small lakes may represent several distribution records. If a species grows in a large lake, which extends over several mapsheets, then each map sheet received a dot.

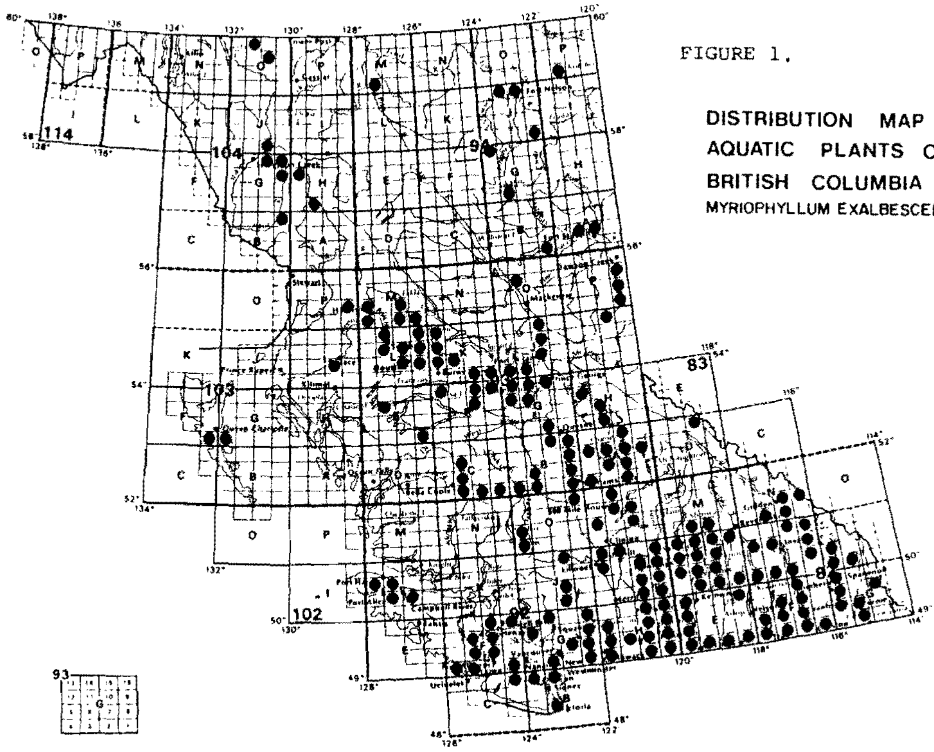
M. aquaticum (Vell.) Verde., (*M. brasilliense* Camb.), is recorded from one artificial pond in a display garden in North Vancouver. Pistillate plants have been growing there for at least eight years and flower annually. The plants overwinter in an active vegetative condition and have been observed sheathed in ice in the winter where they were sprawling up the sides of a small waterfall.

M. heterophyllum Michx. is known from four ponds in Queen Elizabeth Park in Vancouver. In one pond, where *M. spicatum* L. also occurs, the *M. heterophyllum* bed has not been replaced by the invasive *M. spicatum*, which has completely dominated the remainder of the pond over the last eight years.

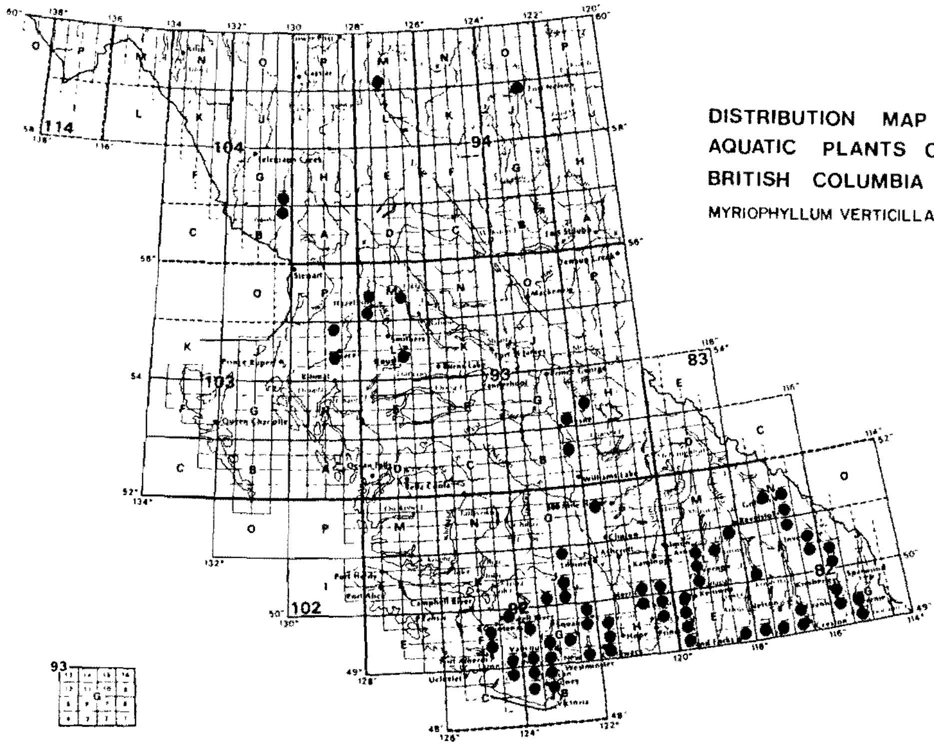
M. quitense H.B.K., (*M. elatinoides* Gaud.), has been found only on Vancouver Island; primarily in habitats with high wave action or in flowing waters. These include five lakes, one slough and two streams.

M. hippuroides Nutt. is found throughout the lower Fraser Valley in most wetlands, roadside and drainage ditches, shallow ponds and sheltered, shallow or 'weedy' water bodies. It is uncommon in large, open lake situations.

FIGURE 1.



DISTRIBUTION MAP
AQUATIC PLANTS OF
BRITISH COLUMBIA
MYRIOPHYLLUM EXALBESCENS



DISTRIBUTION MAP
AQUATIC PLANTS OF
BRITISH COLUMBIA
MYRIOPHYLLUM VERTICILLATUM

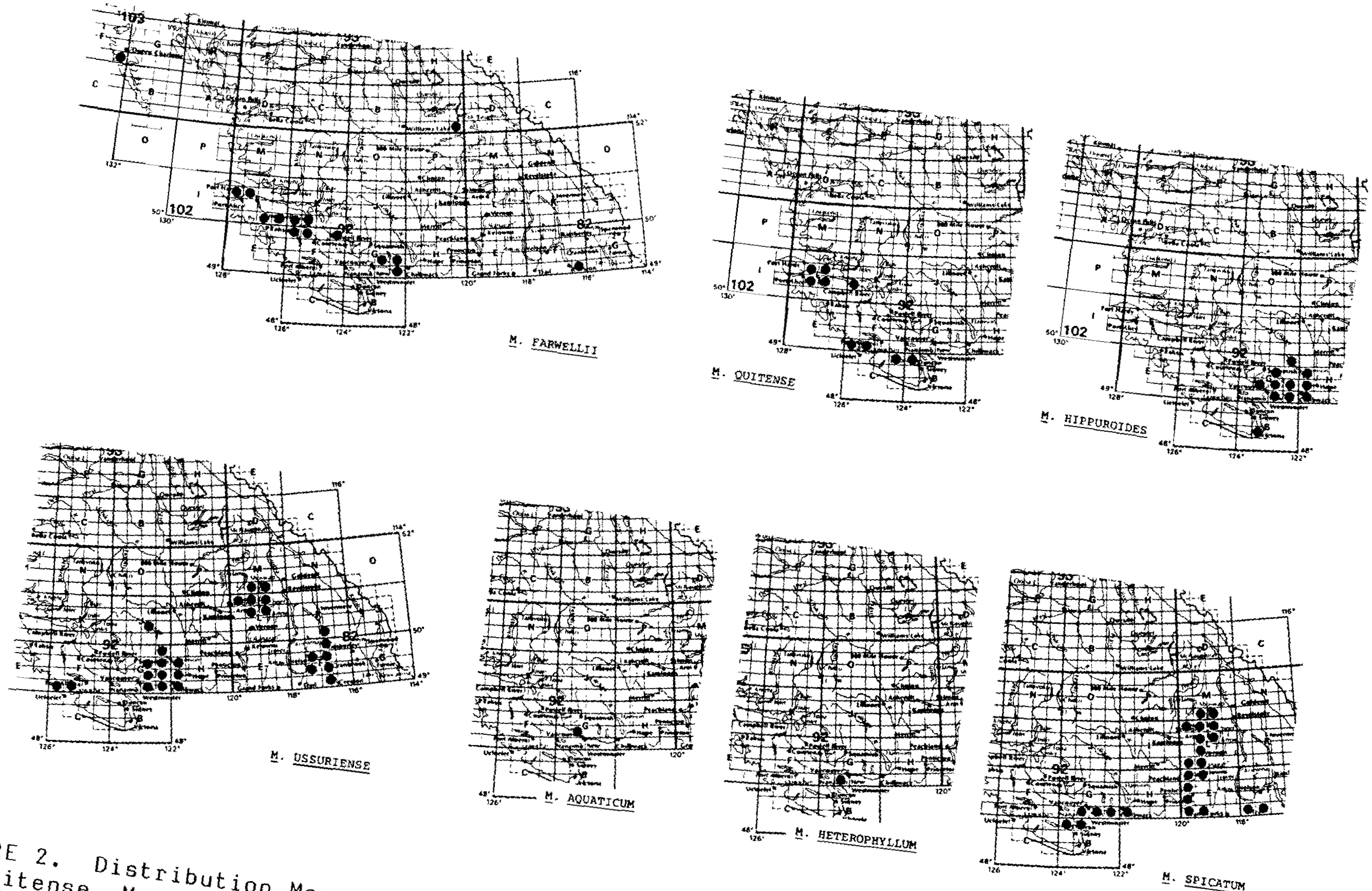


FIGURE 2. Distribution Maps of M. aquaticum, M. farwellii, M. heterophyllum, M. hippuroides, M. quitense, M. spicatum, and M. ussuriense in British Columbia.

M. ussuriense (Regel) Maxim. is most commonly found in the same habitats as M. hippuroides in the lower Fraser Valley but also in suitable, seasonally exposed habitats in the Shuswap, Kootenay and Kennedy Lake valleys. This species flowers when growing as a short, erect, terrestrial on exposed mud banks after water levels drop in late summer.

M. farwellii Morong. is found primarily on northern Vancouver Island but there are a few other isolated locations in British Columbia, including the Queen Charlotte Islands, and the lower Fraser Valley. M. farwellii habitats are often characterized by extensive amounts of wood debris, usually associated with logging.

M. spicatum is largely confined to the Okanagan and lower Fraser valleys. Other locations include Champion Lake, the Pend Oreille River, Shuswap Lake, Magic Lake on South Pender Island and a slough in Duncan on Vancouver Island. It is believed this species was first introduced to British Columbia around 1970 and has not yet had sufficient time to spread to all suitable habitats in British Columbia.

M. exalbescens and M. verticillatum are found in a wide variety of habitats throughout British Columbia. M. exalbescens is more common in alkaline conditions, with higher sodium, potassium and sulphate levels, than M. verticillatum.

Krajina's Biogeoclimatic Zones

Krajina's Biogeoclimatic zones (2) are defined by climate, geology and vegetation factors. Implicit in these zones are altitudinal ranges which are affected by aspect, exposure, latitude and proximity to the Pacific Ocean. Table II shows the distribution of Myriophyllum in British Columbia

Table II. The Distribution of Myriophyllum Species in British Columbia by Krajina's Biogeoclimatic Zones

Zones / Species	CDF	CWH	IWH	IDF	PPBG	CALP	BWBS	ESSF	SBS	SWB	AT	MH
M. exalbescens	12	31	30	111	34	58	21	18	9	2	2	
M. verticillatum	24	28	11	33	3	3	1	8	1	2		
M. spicatum	40		1	2	20							
M. ussuriense	9	6	1	3								
M. farwellii	8	15	1									
M. hippuroides	37	14										
M. quitense	3	5										
M. heterophyllum	4											
M. aquaticum	1											

Note: The numbers in the body of the Table are the number of lakes in which the given species occurs for each Krajina zone.

by biogeoclimatic zone. This Table includes lakes, rivers, garden pools, sloughs and wetlands. Species like M. exalbescens and M. verticillatum which have the widest geographical distribution are also found in the greatest variety of ecological zones. The zone with the lowest elevation, and smallest annual temperature, ranges the CDF zone, hosts the greatest variety of species; all nine species of Myriophyllum are found here.

The percentage of the total number of lakes and ponds in which each of the common Myriophyllum species grows, found in each biogeoclimatic zone is given in Figure 3. M. exalbescens is found in all biogeoclimatic zones except the coastal mountain hemlock zone. In this zone the ground freezes in the winter before an insulating blanket of snow has fallen. M. verticillatum is absent only in the same coastal mountain hemlock zone and also the alpine tundra zone. Both M. exalbescens and M. verticillatum are most prevalent, 35% and 31% respectively, in the interior douglas fir zone, a valley bottom habitat. However, only 11% of the M. exalbescens is found in the two coastal zones, CDF and CWH, while 41% of the M. verticillatum is found in these two coastal zones. M. spicatum is confined almost exclusively to the two zones in which the Okanagan and lower Fraser valleys are found. This probably reflects the initial spreading phase of this recently introduced aquatic plant rather than any indication of habitat limitations. The productivity of M. spicatum is highest in the PPBG zone of the Okanagan Valley but whether this is due to limnological or ecological differences of the locations or genotypic differences in the plants is not known.

Altitude

The distribution of all the Myriophyllum species in British Columbia, in 100 m increments of altitude above sea level, is given in Table III. This Table includes lakes, rivers, garden pools, sloughs and wetlands. M. exalbescens is found 45% of the time between 700 and 1 000 m and only 10% of the time below 200 m. In contrast M. verticillatum occurs only 21% of the time between 700 and 1 000 m about 35% of the time below 200 m. This altitudinal differentiation is, in part, a reflection of the altitudinal ranges of the biogeoclimatic zones in which the two species are most prevalent.

The percentage occurrence of the common Myriophyllum species growing in British Columbia lakes and ponds, in 100 m increments of altitude above sea level, is given in Figure 4. M. verticillatum is not found above 1 300 m while M. exalbescens grows up to 1 800 m. Comparison of Table 3 and Figure 4 shows that about one half of the M. verticillatum waterbodies in the under 200 m range are not lakes; they are sloughs, ditches, backwaters and slow streams in the lower Fraser Valley. M. exalbescens does not have a similar large group of 'non-lake' habitats at low elevations.

These percentage occurrence figures are subject to bias from several sources. Some parts of British Columbia are characterized by a few large lakes (southern interior valleys) and others by many small lakes (lower Fraser Valley, Cariboo plateau). The extent and density of sampling is by no means uniform throughout the province. In one area many small lakes with virtually identical floras may be sampled, while in others only one representative lake has been sampled. Sampling density in northern British

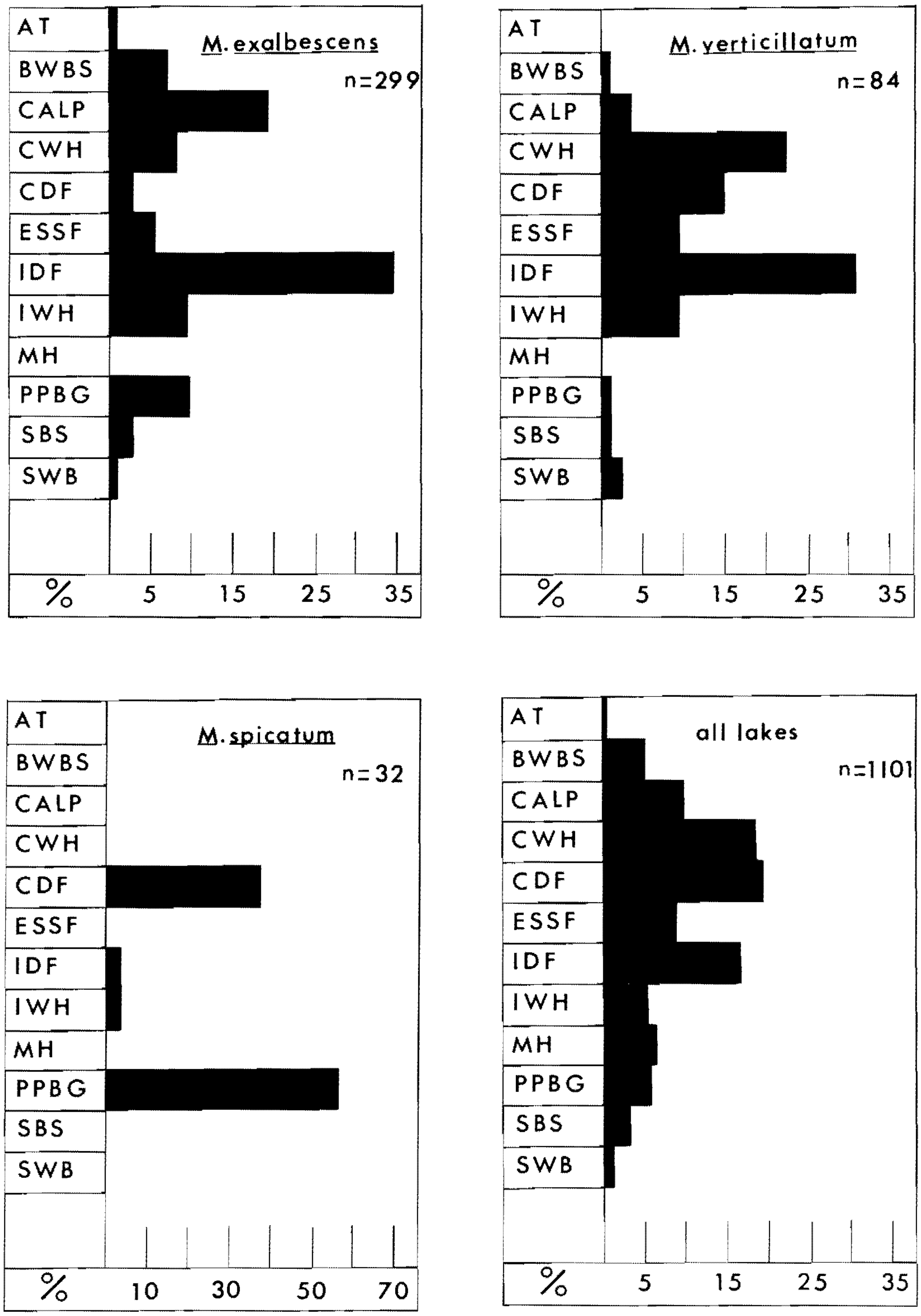


FIGURE 3. Percentage occurrence of Myriophyllum by Krajina'a Bigeoclimatic Zones of British Columbia.

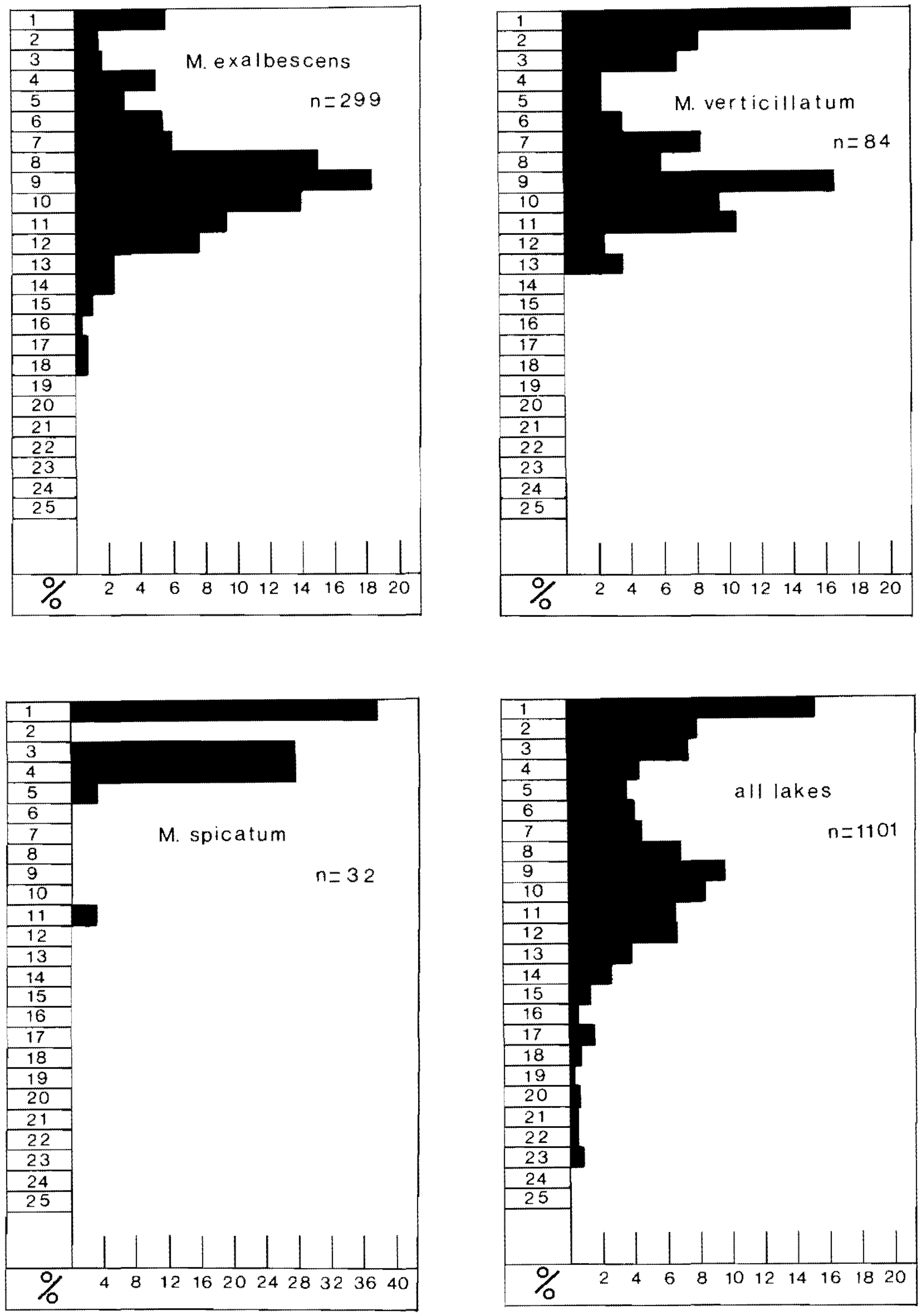


FIGURE 4. Percentage occurrence of <u>Myriophyllum</u> by altitude in British Columbia.

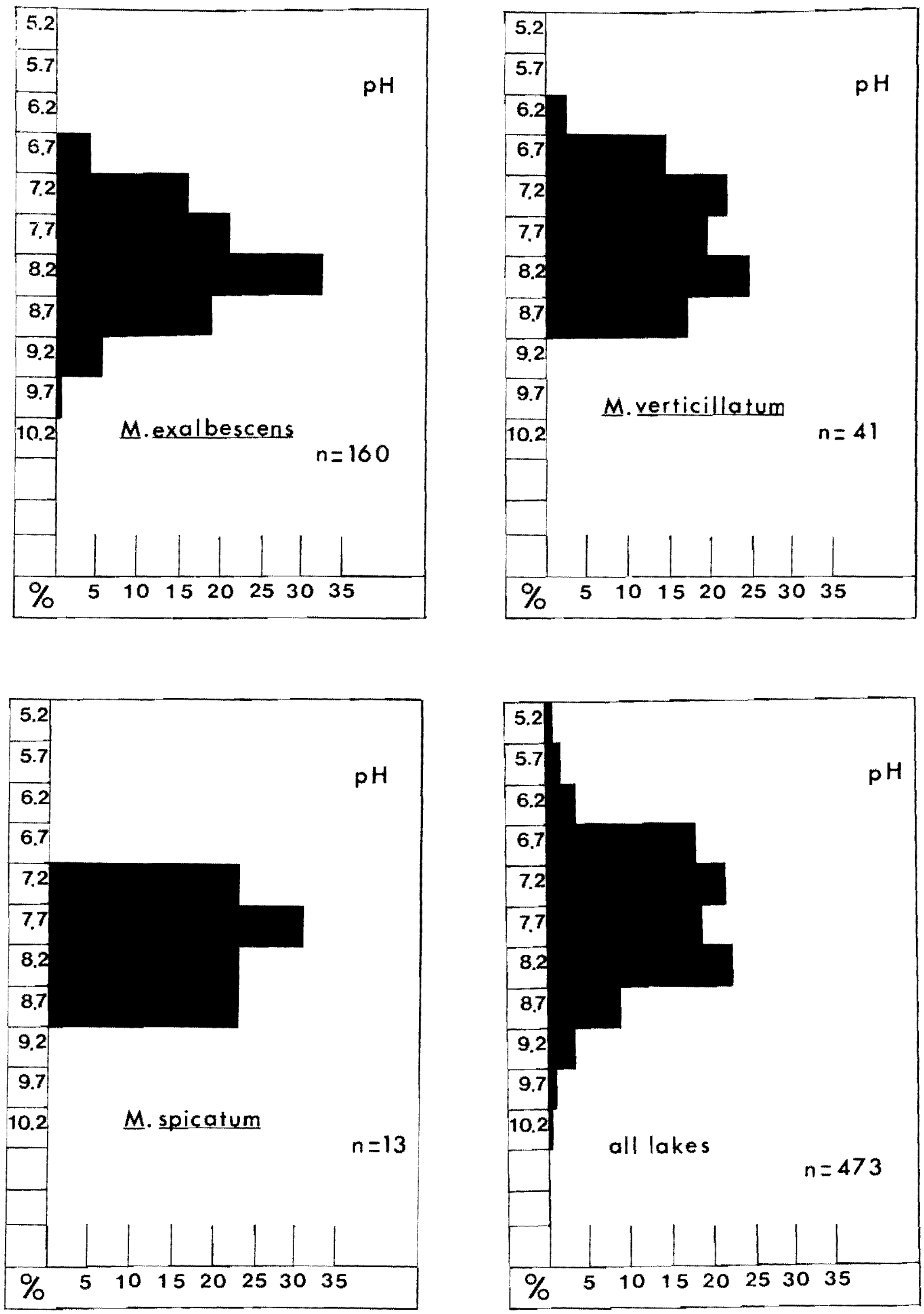


FIGURE 5. Percentage occurrence of Myriophyllum by pH in British Columbia.

Table III. The Distribution of <u>Myriophyllum</u> Species in British Columbia By Altitude in 100 m Increments

Species	Altitude																	
	100	200	300	400	500	600	700	800	900	1000	1100	1200	1300	1400	1500	1600	1700	1800
M. exalbescens	28	4	5	21	11	19	20	48	56	44	29	23	8	7	3	1	2	2
M. verticillatum	33	7	6	3	3	5	6	9	15	9	11	2	2	3				
M. spicatum	38	2	9	11	2						1							
M. ussuriense	14		1	3		1												
M. farwellii	3	10	10						1									
M. hippuroides	49		2															
M. quitense	7	1																
M. heterophyllum		4																
M. aquaticum	1																	

Note: The numbers in the body of the Table are the number of lakes in which the given species occurs for each 100 m altitude block. The first block is lakes below 100 m, i.e. from 0 to 99 m in altitude.

Columbia is very low and uncommon species and habitats have likely been missed.

pH

The pH ranges of the common <u>Myriophyllum</u> species found in British Columbia lakes are given in Figure 5. The increments are 0.5 pH units centred on the pH values listed on the axes of the Figures. The pattern of distribution of <u>M. verticillatum</u> by pH is much like that of the entire set of lakes but has a more restricted range. <u>M. exalbescens</u> grows in lakes with higher pH values. This disparity is even more pronounced if maximum pH values are plotted rather than the mean pH values in Figure 5.

Morphometry

There is a tremendous range of values for the morphometric characteristics of lakes in British Columbia. Characteristics recorded include total area and volume, littoral (shallower than 6 m water depth) area and volume, littoral area and volume as a percent of the total area and volume, mean and maximum depth, length of shoreline, and shoreline development index. No correlations were found with any of these characteristics and any species of aquatic plant. Apparently specific habitats within lakes are more significant than gross lake morphology. Personal experience indicates that markedly dissimilar lakes may still contain very similar habitats, in localized areas, which support similar plant communities.

Water Chemistry

Figures 6 and 7 show the 75th, 90th and 95th percentile limits of some standard water chemistry characteristics for the common Myriophyllum species. The characteristics displayed are hardness, alkalinity, ammonia, organic and total nitrogen, pH, conductivity, total dissolved phosphorus, dissolved chloride, sulphate, potassium and sodium and total calcium and magnesium. These star diagrams facilitate comparisons of patterns and quantities among species. M. spicatum grows in softer water than the other species and tolerates higher ammonia levels. M. exalbescens tolerates harder, more alkaline, high sulphate lakes while M. verticillatum is generally confined to carbonate lakes with lower sodium, potassium and sulphate and higher ammonia.

A correlation matrix indicates that water hardness in M. verticillatum lakes correlates at 0.94 with calcium and magnesium but much lower with potassium, 0.74, and sodium, 0.45. However, in M. exalbescens lakes the correlations with hardness are 0.53 for calcium and 0.82, 0.84, and 0.87 for potassium, sodium and magnesium respectively. In M. exalbescens lakes the correlations with sulphate are 0.63 and 0.60 for potassium and calcium respectively but very low for sodium and magnesium. All correlations with sulphate are very low in M. verticillatum lakes.

For most water chemistry parameters the full range of values may be from 2 to 8 times the 95th percentile value; the species have very wide tolerance ranges even though preferred ranges may be much more restricted. These very wide, overlapping, tolerance ranges for water chemistry characteristics mean that predicting water density from aquatic plants, or the converse, is not practical for most species and characteristics studied to date.

Sediment Chemistry

The star diagrams for some sediment characteristics are given in Figures 8 and 9. There was insufficient data to analyze M. spicatum. Both M. verticillatum and M. exalbescens have the same general pattern and grow in much higher organic carbon and kjeldahl nitrogen levels than the average sediment levels for all lakes. M. verticillatum grows in higher magnesium and total phosphorus levels at high percentiles, than M. exalbescens.

A correlation matrix shows high correlations among percent volatiles, kjeldahl nitrogen and total and organic carbon. The coefficients are generally highest in M. verticillatum lakes, next in M. exalbescens lakes but still over 0.9 in the entire lake data set. There is a 0.98 correlation of inorganic carbon with calcium in sediments of M. verticillatum lakes; this value is only 0.71 in M. exalbescens lakes.

For sediment chemistry characteristics measured, the full range of values is only up to twice the 95th percentile value.

Means of Transport

Myriophyllum distribution is also a function of the means of transport. Quarantine check stations have shown that viable fragments of Myriophyllum are moved on boat trailers and motors from one recreational lake to another (3). Boaters and water skiers also spread fragments within a lake. Ducks and geese may introduce Myriophyllum to ponds and sloughs where they regularly feed. Ducks which were suddenly flushed from a pond heavily overgrown with M. spicatum were observed to trail stem fragments from their feet as they flew away. Resident Canada goose populations have a regular circuit

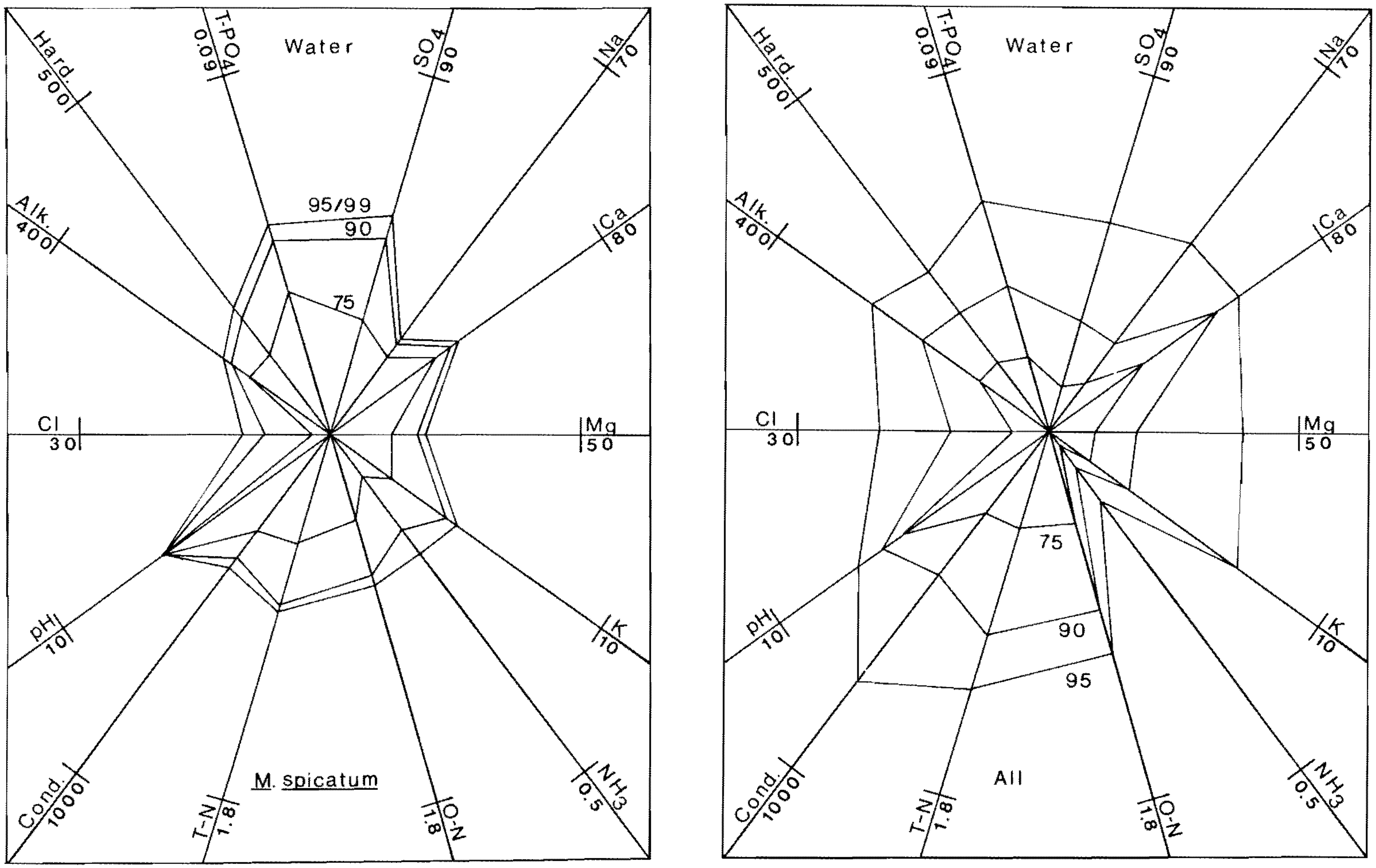


FIGURE 6. Star diagrams of water chemistry characteristics for M. spicatum and the entire lake data set.

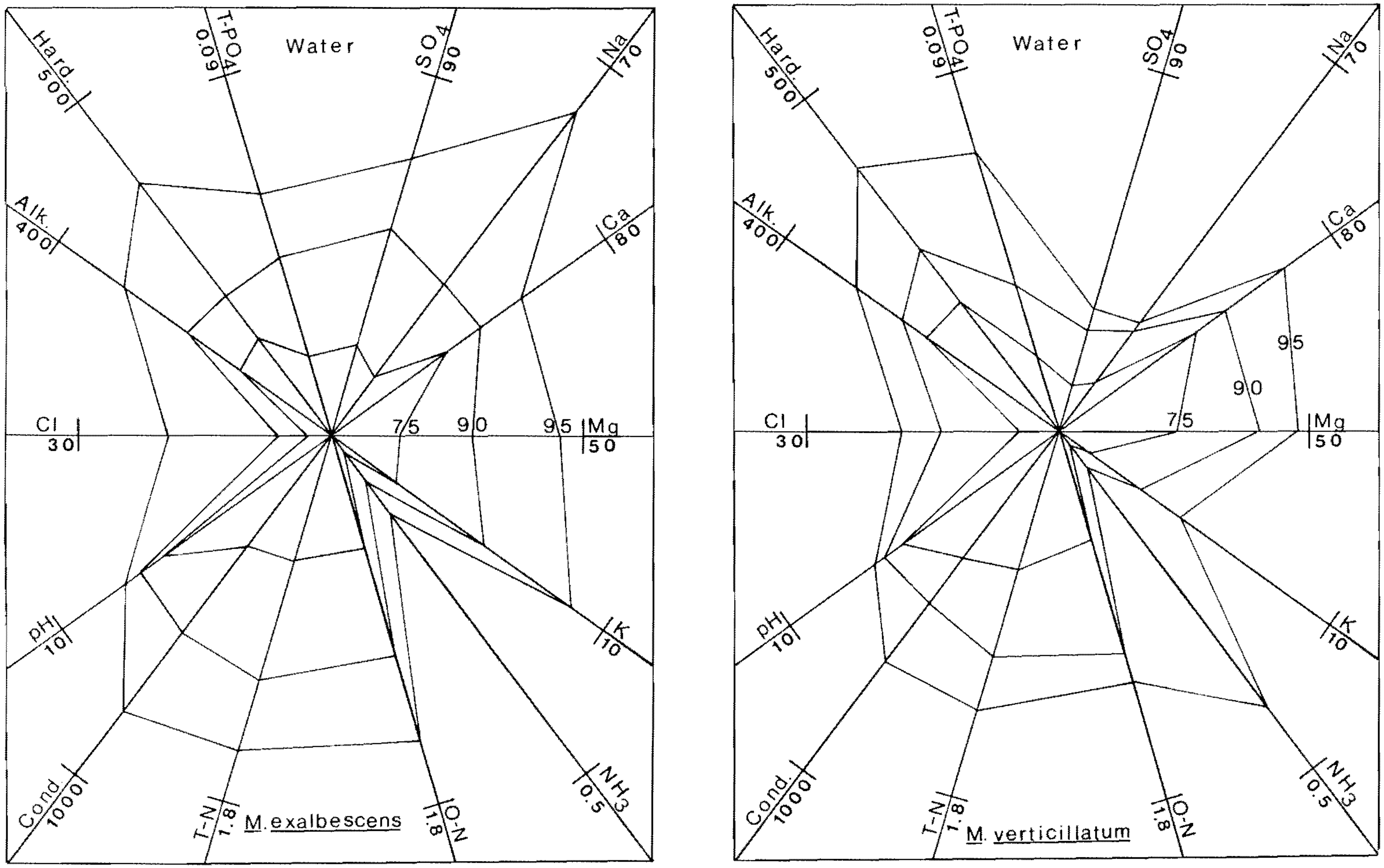


FIGURE 7. Star diagrams of water chemistry characteristics for <u>M. exalbescens</u> and <u>M. verticillatum</u>.

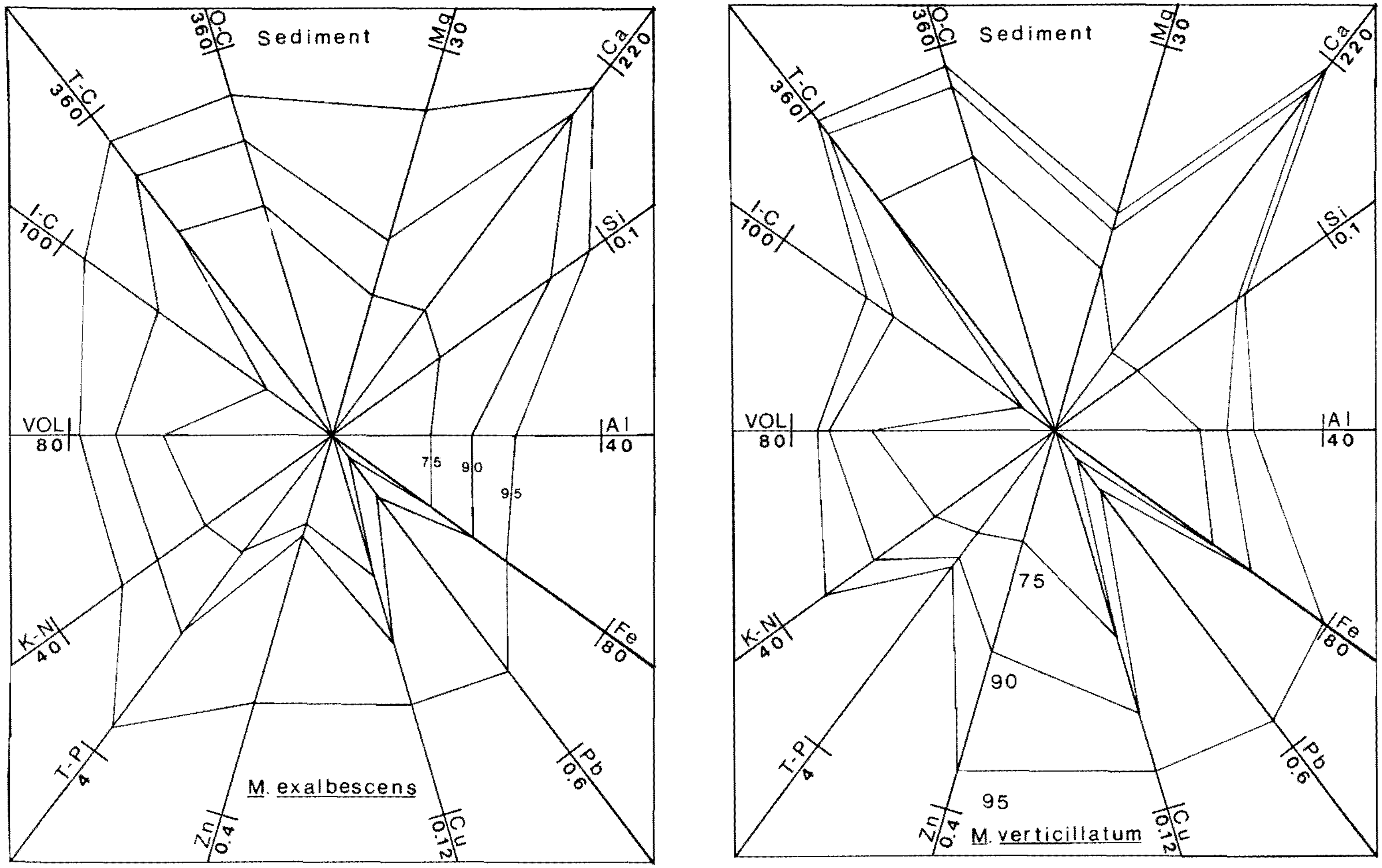


FIGURE 8. Star diagrams of sediment chemistry characteristics for M. exalbescens and M. verticillatum.

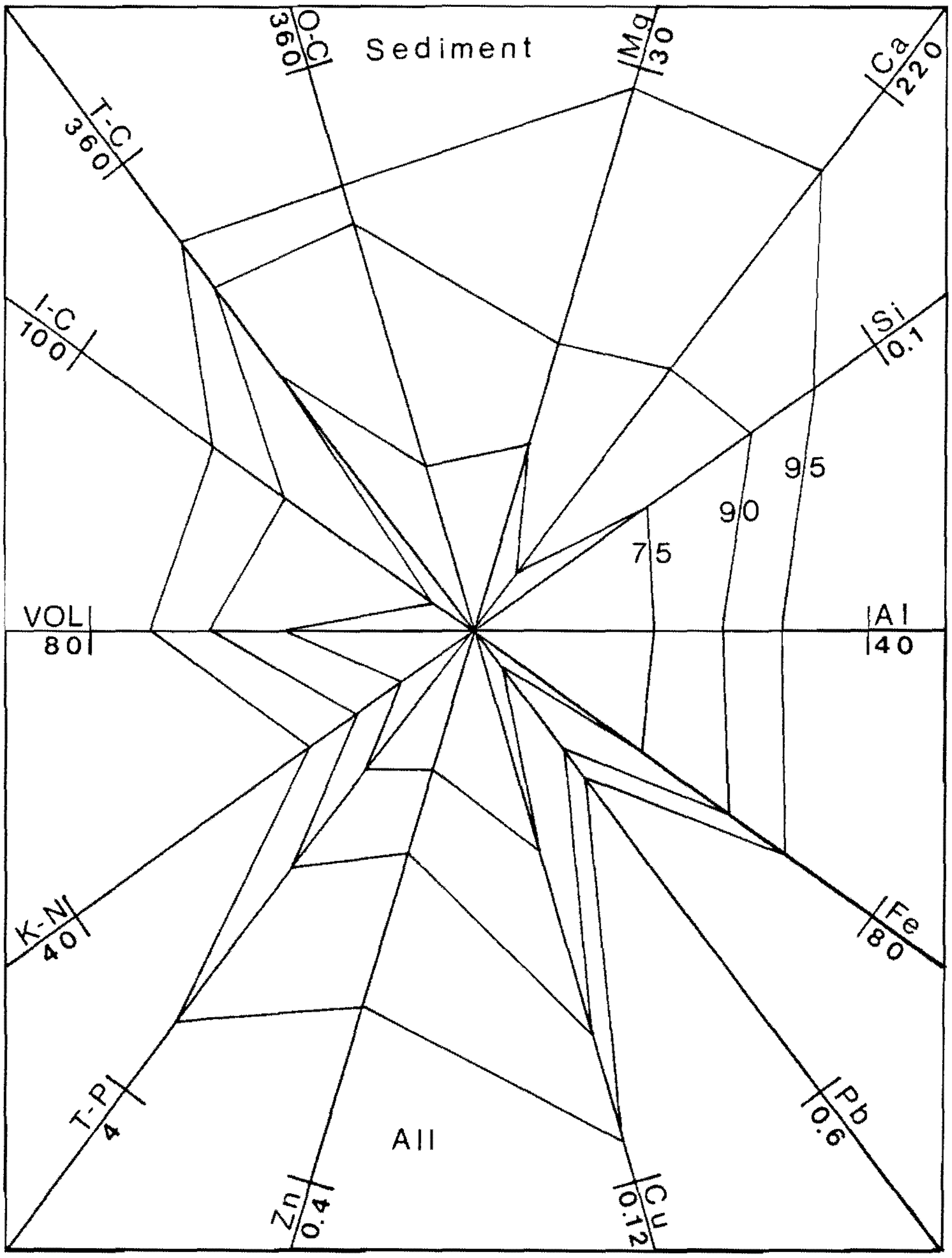


FIGURE 9. Star Diagram of Sediment Chemistry Characteristics for the entire lake data set.

of water bodies in which they forage. When one lake in the circuit becomes infected with M. spicatum the geese are believed to spread it around the whole circuit.

Heavy equipment used to clean out roadside ditches can also spread plants unless machinery is carefully cleaned after working in 'weedy' areas. Municipal equipment used for such jobs is often used to build ponds in parks or maintain beaches on lakeshores and in so doing may spread 'weeds'. When the spoils from ditch, pond and lakeshore cleaning are hauled and dumped, usually in wet or boggy areas as fill, further spread of the plants may occur.

Records show that within one or two years following construction of commercial parks and gardens, such common 'weedy' genera as Ceratophyllum, Elodea, Myriophyllum and filiform Potamogeton are present. They may be introduced by transplanting water-lilies and other aquatic plants growing in sediments from 'weedy' areas and by ducks and geese flying in from nearby sloughs and ditches.

Several instances are known of nurseries and garden shops selling aquatic plants, for ornamental ponds and pools, which are contaminated with viable fragments of M. spicatum. Several Myriophyllum species are prized aquarium plants and are actively shipped all over the world for this purpose. Some of these specimens find their way, accidentally or deliberately, into local waters. While most tropical species do not survive temperate zone winters; some do and become established. M. aquaticum and Elodea densa (Planch.) Casp., waterweed, have successfully overwintered in the lower Fraser Valley for many years. Salvinia, water fern, and Eichhornia crassipes (Mart.) Solms., water hyacinth, are occasionally introduced but do not overwinter.

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NUTRIENT LIMITATION OF *MYRIOPHYLLUM SPICATUM* GROWTH *IN SITU*

Robin M. Anderson[1] and Jacob Kalff
Lake Memphremagog Project, Limnology Research Center,
McGill University, Department of Biology,
1205 Ave. Dr. Penfield, Montreal, Quebec, H3A 1B1, CANADA

The hypothesis that the submersed macrophyte biomass in natural weedbeds is nutrient limited was tested *in situ* by an enrichment experiment. The response of *Myriophyllum spicatum* was significant and positive for N-enrichment, resulting in a 30-40% increase in biomass over controls. There was no response to phosphorus or to potassium enrichment. Plant length and number of shoots per replicate were also significantly increased by nitrogen additions but again showed no response to phosphorus and potassium. Water depth differences were also found to affect the plant responses in some cases.

Studies on lake eutrophication have shown that increased nutrient loading normally results in increased phytoplankton biomass. Spring levels of phosphorus in particular, have proven to be highly successful predictors of the summer phytoplankton biomass of north temperate lakes (1). Whole lake fertilization experiments have shown that a five to ten fold increase in phosphorus resulted in a hundred fold increase in algal biomass (2), and that the reduction of phosphorus inputs to lakes reduced their algal biomass (3). In field studies the biomass of several emergent species has been related to phosphorus (4-6). Broome *et al.* (7) found that nitrogen and phosphorus fertilizer led to increased yields of *Spartina alterniflora* Loisel. Many studies with lowland rice (*Oryza sativa* L., a cultivated emergent) have also shown increased yield in response to fertilization with nitrogen and sometimes to phosphorus and potassium (e. g. 8). The phytoplankton and the emmergent macrophyte work raised the expectation that submersed macrophyte species too are constrained by a shortage of nutrients. Yet the evidence is ambiguous. Barko and Smart (9-11) reported the biomass of several submersed species to be related to sediment characteristics in laboratory growth experiments. Langland *et al.* (12) obtained similar results with *Hydrilla verticillata* Royle. However, *in situ* studies have yet to determine first, whether sediment nutrients do indeed limit the growth of submersed macrophytes and secondly, if sediment nutrients are successful predictors of macrophyte biomass, and thus ultimately of the invertebrates and fish associated with macrophyte beds.

The present study was designed to test the hypothesis that submerged aquatic macrophytes are indeed nutrient limited. If this is the case then additions of the limiting nutrient(s) should result in an increased biomass

[1]Current adress: Département de Sciences Biologiques, Université de Québec à Montréal, C. P. 8888, Succursale 'A', Montréal, Québec, H3C 3P8, CANADA.

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of the manipulated plants. Specifically we tested the prediction that the maximum biomass attained by the submersed aquatic macrophyte Myriophyllum spicatum L. would be increased by additions to the sediment in which it is rooted, of nitrogen (N), phosphorus (P), and/or potassium (K). Nitrogen, P, and K were chosen for this experiment because they are the mineral nutrients needed in the greatest quantities by most plants (e. g. 13 for aquatic plants) and have been found to be most frequently limiting in terrestrial systems (14-16). In vivo growth experiments also suggest that these elements are likely to limit submersed macrophyte biomass (17-19).

Sediment nutrients were chosen over water column nutrients because Myriophyllum spicatum and other macrophyte species tested were shown to take only a negligeable fraction of P from the water (0.6% for M. spicatum) in Quinn Bay, Lake Memphremagog (20). Similar observations have been made elsewhere for nitrogen (21).

Myriophyllum spicatum was used for this experiment because it is easy to propagate by rooting fragments, and because the eurasian species has become a considerable nuisance in many North American lakes. Thus knowledge of factors regulating its abundance may eventually lead to effective control programs.

Materials and methods

The fertilization experiment took place in the summers of 1982 and 1983, in Quinn Bay, Lake Memphremagog, Quebec-Vermont. The natural weedbed in this bay was dominated by M. spicatum. A 10 x 10 m area at the depth of maximum biomass in the natural weedbed (2-3m), was cleared of plants in 1980. Forty galvanized iron hoops (50 cm deep and 50 cm in diameter) were sunk at random into the sediment within the cleared area. Their purpose was to prevent lateral diffusion of the fertilizer away from the experimental plants. Each hoop extended about 15 cm above the surface of the sediment, which served to protect the newly planted shoots at the beginning of the growing season.

The treatments followed a factorial design (Table 1) in which all combi-

Table 1. Factorial design treatments for 1982 and 1983. Included in the table are number of replicates (n), nutrient levels in sediments one week after treatment (ug/g dry weight of sediment), and biomass of M. spicatum (g dry weight/0.2 m^2). Numbers in parentheses are standard deviations.

Year	Treatment	n	N	P	K	Biomass
1982	Control	5	24 (2.4)	69 (27.2)	64 (53.4)	5.1 (3.02)
	N	5	579 (133.9)	80 (12.7)	73 (64.0)	8.7 (3.82)
	P	5	28 (3.8)	151 (47.8)	117 (89.0)	7.2 (2.80)
	K	5	33 (30.8)	63 (20.8)	1243 (420.1)	8.7 (3.77)
	N*P	5	403 (118.0)	187 (53.3)	43 (19.3)	8.4 (3.52)
	P*K	5	36 (16.2)	158 (59.6)	1667 (570.5)	8.3 (4.81)
	N*K	5	659 (279.8)	70 (18.8)	1301 (586.3)	10.2 (2.92)
	N*P*K	5	465 (72.3)	196 (46.1)	1118 (123.8)	8.0 (1.69)
1983	Control	13	61 (35.4)	168 (18.3)	109 (126.6)	14.3 (5.27)
	N*K	13	3446 (1539.2)	124 (47.0)	849 (83.4)	16.8 (7.21)
	P	13	65 (36.9)	422 (83.8)	68 (14.4)	14.73 (8.21)
	N*P*K	13	2266 (1227.3)	390 (68.2)	854 (57.5)	24.0 (11.32)

nations of the three nutrients were included at natural and enriched levels. There were thus seven treatments and a control. Each treatment was replicated five times (Table 1). In 1983 twelve additional hoops were added to increase replication.

The sediments in each treated hoop were fertilized by injecting 100 mls of fertilizer in solution, with a syringe at 10 points within the hoop. Each syringe was equipped with a 10 cm needle, which allowed the solution to be injected below the oxidized zone of the sediment. The fertilizer solutions were bubbled with nitrogen for twelve hours prior to injection to render them highly anoxic. This was done to prevent any oxygen-induced change in form of these nutrients which might make them unavailable for plant use (22). The forms and concentrations of the nutrients used were as follows: N-NH4Cl (324 mg/l in 1982, 300 mg/l in 1983), P-Na PO4 (saturated, 1982), and K-KCl (324 mg/l in 1982, 214 mg/l in 1983). In 1983 Na4H2PO4 2H2O (300 mg/l) was used as it is much more soluble. Ammonium-nitrogen was the only form of nitrogen used because nitrate and nitrite are undetectable below the first two to three centimeters of the sediment (M. R. Anderson, unpubl. data).

The hoops were fertilized at the end of May (Table 2) and left to equilibrate for one week. The sediments were then sampled with a long narrow core tube (2 cm internal diameter x 40 cm in length), so as to disturb the sediments as little as possible. Three cores were taken from each hoop. The sediment from the rooting zone (> 5 cm) was combined for each hoop and frozen until analysis. The sediments were analysed for chemically exchangeable N, K (23), and P (24).

After the sediments had been sampled the hoops were planted with fragments of *M. spicatum*. These shoots were collected from the natural weedbed in Quinn Bay and were pre-rooted in a shallow wading pool for ten days prior to planting. Only shoots with well developed roots were used in the experiment. The initial planting density was 15 shoots per hoop in 1982 and 20 shoots per hoop in 1983. Each shoot was 10 to 15 cm in length. The plants were harvested in mid-August (Table 2) when the biomass was near maximum for the natural

Table 2. Time sequence for growth experiments in 1982 and 1983.

	1982	1983
Fertilisation of hoops	27-29 May	28-30 May
Sediment sampled	15-16 June	10-11 June
M. spicatum planted	17 June	11-12 June
Growth measurments	-	21,28 June 16,24 July 5 August
M. spicatum harvested	23 August	15 August
Sediment sampled	24 August	16 August

population in Quinn Bay (M. R. Anderson, unpubl. data). At this time the experimental plants just reached the surface, and had not yet started flower induction. The plants were clipped at the sediment surface then returned to the laboratory where they were counted, measured, weighed, dried at 80° C to constant dry weight (48 hours) and weighed again. After the harvest the sediments were again sampled. In 1983 the plants were also counted and their length measured *in situ* at intervals over the growing season, to estimate their growth rate.

All data analyses were done using the General Linear Models (GLM) proceedure of SAS (25). In 1982 two of the hoops were used by fish as nests resulting in almost total shoot mortality in each. They were therefore not included in the analyses.

In 1983 an error was made in the treatments such that the nitrogen additions were confused with the potassium additions. Because of this, the nitrogen treatment was not independent of the potassium treatment and it was therefore difficult to separate the effects of the two treatments for that year. This was in part remedied by examining the effect of each nutrient within the two treatment levels (enriched and natural) where they were not correlated, and in part by entering them separately into the models to determine which explained a greater degree of variation.

Results

The only nutrient that significantly affected *M. spicatum* biomass in both 1982 and 1983 was nitrogen (Table 3-a). Neither phosphorus nor potassium additions resulted in increased biomass in 1982 (Table 3-a) and no interaction between nutrients was significant. The same was observed in 1983 for phosphorus (Table 3-a) and for the nitrogen-phosphorus interaction term in

Table 3. Analysis of length of plants (L), maximum length (LM), number of shoots (No), and total biomass per hoop (/0.2 m^2) (DW) in relation to -a- exchangeable nitrogen (N), phosphorus (P), and potassium (K) and -b- to N and depth (Z). (NS = not significant at the 0.05 level, * = *P* < 0.05, ** = *P* < 0.01, *** = *P* < 0.001).

-a- Dependent variable	N	P	K	F	*P*	r^2
		-1982-				
DW	*	NS	NS	7.22	0.0109	0.167
L	NS	NS	NS	4.02	0.0525	0.101
No	NS	NS	NS	—	—	—
		-1983-				
DW	*	NS	NS	4.05	0.0496	0.075
L	**	NS	NS	7.89	0.0071	0.140
No	NS	NS	NS	—	—	—
LM	**	NS	NS	10.32	0.0023	0.170

-b- Dependent variable	N	Z	F	*P*	R^2
		-1982-			
L	**	***	14.10	0.0001	0.446
No	NS	NS	—	—	—
DW	*	NS	see -a-		
		-1983-			
L	**	NS	see -a-		
LM	**	*	11.61	0.0001	0.326
No	NS	*	11.39	0.0015	0.189
DW	*	*	4.30	0.019	0.149

1983. Despite the lack of independence between potassium and nitrogen in 1983, potassium again showed no significant effect on biomass ($\underline{P}$>0.05, Table 3-a).

Nitrogen also was the only nutrient to significantly increase shoot length and maximum length (length of the longest shoot) in 1983 (Table 3-a) whereas the increase in shoot length attributed to nitrogen is almost significant in 1982 (Table 3-a). The other factor which had a significant effect on length of plants was depth (Z) (Table 3-b). Although the area chosen is within the zone of peak biomass of the natural weedbed there is nonetheless a 100 cm difference between the deepest and the shallowest hoops. This range was sufficient to induce a concommittant variation in plant length (Table 3-b) and when depth was included in the 1982 model, nitrogen was also found to significantly affect the length of the plants (Table 3-b). While depth had no effect on total biomass in 1982, it did in 1983 (Table 3-b).

In 1983 the length of the plants that were fertilized with nitrogen had become significantly greater by mid July, than those that didn´t receive N (Table 4). Between June 28th and July 16th the N-fertilized plants grew significantly faster than all others (Table 4-c). The total length of the N-fertilized plants continued to be significantly greater for the rest of the experiment. The increase in the slope of the relationship between nitrogen and length over the season (Table 4-a) indicates that the difference between the two groups (N-enriched and N-natural) continued to increase even though the growth rates were not significantly different (Table 4-c). Nitrogen additions also increased the number of shoots per hoop slightly in mid-season (Table 4-a).

Table 4. Significant relationships for length (L), maximum length (ML), and number of shoots (No) for the 1983 growing season with -a- nitrogen (N) and -b- depth (Z), and for -c- the absolute growth rates of those variables with nitrogen. Absolute growth rate is the total increase in the variable from one time to the next. ex. Absolute growth rate in length for the interval 24/7 and 5/8 is L(5/8) - L(24/7). Values are the slopes of the relationships with r^2 (in parentheses) for the significant relationships. (NS = not significant at the 0.05 level, * = $\underline{P}$ < 0.05, ** = $\underline{P}$ <0.01, *** = $\underline{P}$ < 0.001).

Date	-a- with N			-b- with Z			-c- absolute growth rate		
	L	ML	No	L	ML	No	L	ML	No
21/6	NS	—	—	—	—	—	—	—	—
28/6	NS	—	NS	-8.15* (0.12)	—	NS	0.06* (0.10)	—	NS
16/7	0.13** (0.19)	NS	NS	-11.85** (0.18)	NS	NS	NS	NS	NS
24/7	0.17** (0.13)	NS	0.001* (0.08)	-14.15* (0.10)	NS	NS	NS	NS	NS
5/8	0.25*** (0.20)	NS	0.002*** (0.20)	NS	NS	NS	NS	NS	NS
15/8	0.29** (0.14)	0.006** (0.17)	NS	NS	0.43* (0.10)	0.35** (0.19)	NS	0.003* (0.09)	NS

Depth had a negative effect on length until July (Table 4-b) followed by a lack of a significant effect until harvest time when it had become positively related to the maximum length of the shoots (Table 4-b). At that time maximum length of shoots was also positively related to nitrogen levels (Table 4-a).

Nitrogen additions resulted in an increase in the dry weight per shoot of about 25% in both years (Table 5). The number of shoots per hoop and the length of shoots were also increased by N enrichment though these increases were not statistically significant (Table 5). As a result dry weight of shoots per hoop also increased significantly by 29% in 1982 and 40% in 1983 (Table 5), reflecting increases in both the number and weight of those shoots.

Table 5. The average percent increase in dry weight per hoop (DW), number of shoots (No), length of shoots (L), and dry weight per shoot (DWIND) upon N addition. Stars indicate significant increases (* = $P < 0.05$, ** = $P < 0.01$).

Year	DW	No	L	DWIND
1982	29%*	6%	10%	23%*
1983	40%*	13%	32%**	27%*

Discussion

We have shown that increased sediment nitrogen will result in an increase in *M. spicatum* biomass. Neither phosphorus nor potassium had such an effect. Thus in Quinn Bay, Lake Memphremagog, *M. spicatum* growth was N limited. Nitrogen additions resulted in an increase of 30 to 40% in maximum biomass. Visual observation of the treatments in both 1982 and 1983 also supports the N-limitation hypothesis. The N-enriched plants appeared more vigorous throughout than the controls. They were greener and their stems were fleshier. This type of observation is common for N-enriched crop plants and is supported by a highly significant ($r2 = 0.24$, $P < 0.0017$) doubling of the wet to dry weight ratio for the N-enriched plants.

In Quinn Bay, pore water nitrogen has been found to be very low in the spring (R. Carignan, pers comm.). Thus the plants receiving extra nitrogen early in the growing season may have been able to get a head start, explaining why the growth rate of the N-enriched plants was significantly greater at that time. This early advantage was sufficient to be maintained through the rest of the growing season. Though the final effect on biomass was small over a single growing season the cumulative effect of nitrogen enrichment may be much greater. *M. spicatum* is perenial (26) and an advantage gained in one growing season will likely be carried over to the next in the overwintering biomass. High nitrogen levels by their effect on shoot length, will also allow the plants to reach the surface earlier in the growing season, allowing canopy formation (the nuisance phase) and flowering. The natural population of *M. spicatum* in Quinn Bay, did not reach the surface in time to produce canopy or flowers in either of the growing seasons (M. R. Anderson, unpubl. data). Thus the plants in fertilized areas would have a greater reproductive potential both from vegetative fragmentation of their greater biomass and from seed production.

The magnitude of the effect of nitrogen on M. spicatum is greater than that observed when nitrogen was added to Spartina alterniflora beds (15% increase in yield, 27) though less than the increase (~100%) for the emergent macrophyte Oryza sativa (28). However, the M. spicatum response to increased nitrogen is small compared to the response of the phytoplankton to nutrient additions. Thus a two-fold increse in total P in the southern portion of the lake (29) results in a 150 per cent greater algal biomass than that in our study area (30), wheras a 15 to 50 fold increase in sediment nitrogen (exchangeable N) yielded omly a 30 to 40 per cent increase in maximum macrophyte biomass.

We conclude that M. spicatum, in Lake Memphremagog at least, is limited by sediment nitrogen, whereas the phytoplankton in the same lake is primarily phosphorus limited (29, 31). However the macrophyte growth response to fertilization is small compared to that of the phytoplankton in Lake Memphremagog and elsewhere (3). Thus environmental factors other than nutrients may have a major impact on the macrophytes. One of these additional factors is water depth (light) which significantly increases tha amount of variance explained in the model even over the 100 cm range encountered (Table 3). Other such factors include underwater slope and fetch (Duarte and Kalff, unpubl.). Our findings not only show for the first time a direct in situ submerged macrophyte growth response to fertilization but also indicate the disproportionate importance, compared to the phytoplankton, of factors other than nutrients in the dynamics of submersed macrophytes.

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EURASIAN WATERMILFOIL (MYRIOPHYLLUM SPICATUM L.) IN THE TENNESSEE-VALLEY: AN UPDATE ON BIOLOGY AND CONTROL

A. Leon Bates, Earl R. Burns, and David H. Webb

Tennessee Valley Authority
Muscle Shoals, Alabama 35660

Eurasian watermilfoil, (Myriophyllum spicatum L.), has been the dominant submersed aquatic weed in Tennessee Valley Authority main stream reservoirs for 25 years and severely impedes water resource utilization. Its rapid spread after planting from an aquarium has extended the distribution over more than 840 river km and during peak infestation in 1968-69 colonized 8,900 ha. Standing crops of watermilfoil vary both spatially and temporially between reservoirs and within reservoirs. The infestation trend as determined from aerial photography has been upward for the last 15 years, reaching a recent maximum of 5,870 ha in 1982. Water level manipulation has provided the major control influence on watermilfoil, but has provided a selective advantage to annual species, such as naiads and some pondweeds. For the last 25 years, 2,4-D (2,4-dichlorophenoxy acetic acid) and various contact herbicides have been used to supplement this control technique. Herbicide treatment with 2,4-D has been less successful in colonies of mixed macrophytes wherein companion species, such as spinyleaf naiad that are more tolerant to 2,4-D, become the dominant species after selective removal of watermilfoil.

Introduction

Eurasian watermilfoil (Myriophyllum spicatum L.) is a well-established naturalized aquatic macrophyte in Tennessee Valley Authority (TVA) reservoirs. Since its introduction more than 25 years ago, watermilfoil has spread throughout the Tennessee River system with distributions in riverine habitats as well as small farm ponds. The prolific, asexual spread continues to cause excessive infestations that (a) impact water contact recreation, (b) clog intakes for raw industrial water and electric power plants, (c) depress real estate values, (d) cause depressed dissolved oxygen levels, (e) interfere with commercial fishing, and (f) cause significant increases in permanent pool mosquitoes such as Anopheles quadrimaculatus. Early attempts to eradicate watermilfoil were futile and the current integrated weed control program utilizes drawdowns and herbicide treatment to achieve maintenance control.

Bionomics

From an alleged planting of watermilfoil by a commercial dock operator in an east Tennessee reservoir about 1953, watermilfoil attained a peak infestation of about 8,900 ha in 1968-69. Extensive herbicide treatment in 1969 substantially reduced the total infestation to about 1,200 ha the following year. Since 1970 the trend has been upward, peaking at about 5,870 ha in 1982. Expenditures for herbicide maintenance of watermilfoil has exceeded $6 million since 1961. Herbicide treatment, climatic factors that indirectly affect light penetration and thus all submersed species, special drawdowns, and competition from companion species have caused annual fluctuations in infested areas as measured from aerial photography. Table 1 summarizes hectares of monotypic watermilfoil colonies by reservoir for the last five years.

Table I. HECTARES OF MONOTYPIC COLONIES OF EURASIAN WATERMILFOIL IN SIX TENNESSEE VALLEY RESERVOIRS FROM 1980-84 AS DETERMINED FROM AERIAL PHOTOGRAPHY

Reservoir	1980	1981	1982	1983	1984
Guntersville	3,025	3,970	4,445	3,695	3,020
Chickamauga	319	1,180	510	680	240
Nicakjack	170	170	270	270	105
Watts Bar	50	365	195	405	110
Melton Hill	115	85	95	85	85
Fort Loudoun	55	50	50	45	45
Total	3,734	5,820	5,565	5,180	3,605

Watermilfoil extends over 840 km of the main stream portion of the Tennessee River with about two-thirds of the infestation historically occurring in Guntersville Reservoir in northeast Alabama. Design limitations of the impounding structure, water intakes, and navigation requirements limit the amplitude of fluctuation of Guntersville Reservoir to less than a meter within an annual cycle, and create a relatively stable pool suitable for colonization by submersed species.

Other reservoirs have a greater amplitude (>3m) of fluctuation and littoral zones are less favorable for establishment of submersed macrophytes. Tributary reservoirs with more than a 5 m annual amplitude of fluctuation are usually devoid of watermilfoil and other submersed macrophytes. In such reservoirs with an absence of macrophytes, efforts have been made to artificially establish annual terrestrial or wetland species to enhance fisheries and wildlife habitat (4).

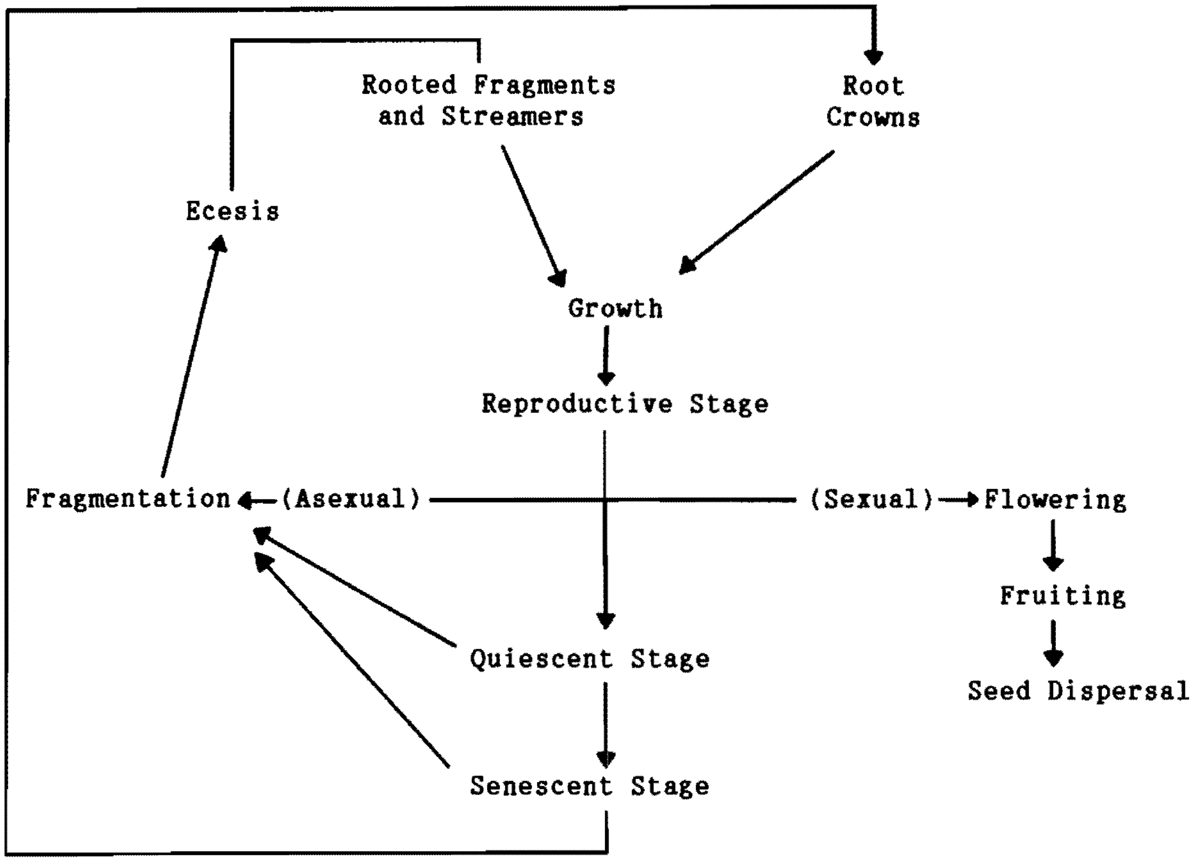
Phenology

High standing crops of watermilfoil persist at some locations in heavily infested reservoirs throughout the year, even in embayments that occasionally have several days of ice cover. More frequently, standing crop declines during winter months and regrowth occur from adventitious shoots arising from root crowns in the substrate. Although regrowth has been observed as early as January, it usually begins in March and is characterized by reddish meristems and young leaves. This rapid spring regrowth (stem elongation) phase allows the plants to reach the surface and form floating mats about July 1. Interference with water contact recreation usually begins when biomass is just below the water surface. Maximum problems occur when surface mats are formed.

Flowering has been observed as early as May 1, although peak anthesis occurs after August and continues until heavy frost. While studies (14) indicate that flowering is directly related to water temperature, unexplained variation is often noted in the initiation of flowering and peak anthesis at different locations within reservoirs and from year to year in similar habitats.

Following fertilization the inflorescence sinks and ultimately releases viable mericarps. An unexplained phenomenon relates to an apparent absence of seedlings in the littoral zone based on more than 25 years of observation and sampling. The mericarps readily germinate in the laboratory without afterripening or scarification, and seed removed from lacustrine substrates subsequently have been germinated under laboratory conditions. Studies of seed germination indicate that the germination frequency is dependent upon and varies directly with light intensity. The intensity requirement is low; consequently, under natural conditions mericarps should receive sufficient light for germination. The lack of germination under natural conditions is considered to be related to the actual wavelengths of light encountered or other inhibitory mechanisms (2). Thus, sexual reproduction is considered to be insignificant in the life cycle of watermilfoil in the Tennessee Valley (figure 1).

Figure 1. SCHEMATIC LIFE CYCLE OF <u>MYRIOPHYLLUM</u> <u>SPICATUM</u> L.



Two "pulses" of auto fragmentation have been noted; one release occurring in March-April and another in September-October. Roots often form on these abscission fragments before release from the parent plant or develop on others while suspended in the water column. These fragments readily root in any substrate that provides physical anchorage even interstitial crevices in bedrock. Overfilling of reservoirs (surcharges) to strand abscission fragments and other viable stem fragments upon the shoreline is considered effective in reducing infestations.

Dewatering of watermilfoil, especially during the summer and fall, results in the formation of terrestrial "landforms." Erect apices with short internodes form at each growing tip and, provided the substrate remains moist, form roots. Extended dewatering (more than 3 weeks) usually results in desiccation (9) whereas reflooding during this interval causes regrowth from the "landform." This physiological response presumably is an adaptation to survive fluctuating water levels of short duration.

Community Dynamics

From 1960 until the mid-1970s, watermilfoil was the only submersed aquatic macrophyte that required large-scale management. Since the mid-1970s the number of species requiring control has increased substantially. Several annuals, such as southern naiad (*Najas guadalupensis* (Spreng.) Magnus), small pondweed (*Potamogeton pusillus* L.), spinyleaf naiad (*N. minor* All.), and the macro-algae, muskgrass (*Chara* spp.), are now common in the drawdown zone of most main stream reservoirs. These species, in addition to perennials such as American pondweed (*P. nodosus* Poir.), currently require large-scale management.

While a portion of the increased diversity of the aquatic flora may be related to reservoir aging, the widespread use of 2,4-D for watermilfoil control and the late fall and winter drawdowns of most main stream TVA reservoirs are considered to be major factors in the shifts of aquatic macrophyte communities. Treatments with 2,4-D exert selective removal of watermilfoil in mixed communities while allowing tolerant species, such as the naiads, pondweeds, and coontail, to persist. In impoundments with non-weedy, native aquatic macrophytes, such as certain pondweeds, the use of 2,4-D to selectively remove watermilfoil may provide an effective management tool to give a competitive advantage to more desirable aquatic macrophytes.

The late fall and winter drawdowns utilized by TVA has proven effective in controlling watermilfoil in the drawdown zone. However, these drawdowns favor the establishment and dominance of annual species, such as the naiads and some annual pondweeds. For example, planimetric measurements of aerial photographs of Chickamauga Reservoir with an annual drawdown of more than 2.5 m show spinyleaf naiad as the dominant species in a reservoir previously dominated by watermilfoil (Table II).

Table II. AREAL COVER OF SPINYLEAF NAIAD AND EURASIAN WATERMILFOIL IN CHICKAMAUGA RESERVOIR FROM 1980 TO 1984 AS DETERMINED FROM PLANIMETRIC MEASUREMENT OF AERIAL PHOTOGRAPHS

SPECIES	INFESTATION 1980	1981	1982	1983	1984
	(hectares)				
EURASIAN WATERMILFOIL	482	1,227	701	779	325
SPINYLEAF NAIAD	843	881	1,827	1,942	1,821

Monotypic stands of watermilfoil occur in Tennessee Valley streams and reservoirs; however, evidence of allelopathy is not known for watermilfoil. Slender spikerush (Eleocharis acicularis (L.)(R. & S.) has been shown (13) to have allelopathic properties and may offer a partial naturalistic control for submersed species in shallow water of reservoirs. It is important to note that spikerush is able to tolerate about 0.5 m of inundation during the summer months in the Tennessee Valley, but must be dewatered in late summer or early fall for rhizome expansion. In the littoral zone of TVA mainstream reservoirs, natural expansion of spikerush colonies is favored by late season water level manipulations and rotary mowing of emergent vegetation in the fall for mosquito control.

Habitat Relationships

Eurasian watermilfoil by virtue of its rapid spread and wide ecological amplitude of tolerance to chemical and physical parameters, expands readily into uncolonized water bodies. Water depth and factors affecting light penetration are considered to be the major limiting factors for watermilfoil in the Tennessee Valley.

The depth of colonization by watermilfoil varies between reservoirs and within reservoirs depending on the light extinction by suspended organic and inorganic fractions. Maximum colonization depths rarely exceed 5 m based on studies on Melton Hill Reservoir and observations at other locations. A shallow erosion zone occurs at depths less than 0.5 m of normal top summer pool, eliminating colonization by watermilfoil because of the wave action and deposition of suspended sediments (11). Watermilfoil in riverine habitats withstands hydraulic drag and turbulence and elongates to about 3 m if not subject to extreme current velocities.

Diverse geologic formations throughout the Valley provide parent material for substrates throughout the textural and nutrient availability ranges. Extremes of physical substrate characteristics such as substrate particle size limit standing crops. For example, high-bulk density, clay substrates are rarely colonized due to the physical limitations of the substrate as a rooting medium. Conversely, coarse-textured substrates are frequently colonized by watermilfoil and "improvement" in the substrate occurs as sediments aggrade following silt deposition along with supplemental additions of organic matter. In general, watermilfoil creates a habitat not only suitable for further colony expansion, but also for establishment of other submersed macrophytes. Aggradation of organic ooze in significant quantities is rarely observed in TVA reservoirs presumably because of the water volume exchange rates (mean flow of Tennessee River 1,800 cfs, with extremes of 130 to 12,700 cfs) and partial oxidation of organic matter during seasonal drawdowns.

Basin morphometry of TVA impoundments has been conducive to macrophyte establishment. In the Tennessee Valley, the relatively level first terrace alluvial "river bottoms" were rowcropped prior to impoundment and now support extensive colonies of submersed macrophytes. Narrow, linear colonies of watermilfoil occur along natural levees of old river banks and create valuable habitat diversity for the fisheries' resource because of the "edge effect." Watermilfoil colonies also support macroinvertebrate communities that differ from that of open littoral habitats and are important in overall reservoir diversity (8).

Impacts of Excessive Populations

Aquatic macrophytes such as Eurasian watermilfoil usually create problem situations because of overabundance. Subjective determinations of problem status ultimately depend on the primary use of the waterbody or watercourse. The main stream TVA reservoirs serve multipurpose uses wherein excessive populations usually interfere with one or more uses. The major negative impact, based on number of individuals affected, is upon water contact recreation. Economic losses are, at best, difficult to determine because of intangible evaluation of factors such as depression of real estate value or aesthetic degradation.

Infrequent negative impacts such as clogging of raw industrial water intakes at electric generating facilities or large industrial plants result in expensive delays in manufacturing or reduced generation loads. Incidences of partial clogging of intakes at TVA's steam electric generating facilities have been attributed to floating mats of watermilfoil, spinyleaf naiad, coontail, and other macrophytes during the fall senesence.

Since its introduction, Eurasian watermilfoil has provided extensive habitat for production of the permanent pool mosquito (*Anopheles quadrimaculatus*) (5). After reaching the water surface or "topping out," watermilfoil forms interstitial pools that afford protection from predation for mosquito larvae and also provide a food source by harboring algae and other organic debris. Watermilfoil and other macrophytes with similar growth forms create mosquito habitat that requires chemical larviciding to reduce anopheline mosquito populations. For the last five years, more than 6,200 ha per year have been larvicided by helicopter to reduce nuisance populations on heavily weed infested Guntersville Reservoir. Since permanent pool mosquitoes, such as *An. quadrimaculatus*, potentially serve as disease vectors (e.g. malaria, encephalitis) dense aquatic macrophytes may indirectly affect public health and welfare.

Management and Control

Management strategies for aquatic weed control generally entail procedures for (1) eradication, (2) maintenance, and (3) prevention. The strategy employed is contingent upon the stage of succession, resources available, and the intended use of the waterbody. The institutional framework for implementation of control or management of aquatic vegetation usually lags behind development of nuisance populations; thus, the water resource manager is faced with short-term temporary fixes to a persistent problem. Furthermore, development of control technology has been slow for recently introduced exotic species such as watermilfoil, and a full complement of integrated control measures cannot be exerted on the pest species.

The strategy for control of watermilfoil in TVA reservoirs followed typical pest management strategies. Initially, two attempts were made to treat all known infestations with 2,4-D herbicide. The treatments were successful for watermilfoil infestations subjected to sufficient herbicide concentrations; however, due to the impossibility of treating all colonies and the distribution of fragments in more than 93,000 ha of surface water, widespread regrowth occurred. Subsequently, a maintenance strategy was adopted about 10 years after proliferation of the original infestation, and a maintenance control strategy for watermilfoil and more recently admixtures of other weeds has continued for the last 15 years. The maintenance strategy employs the use of critical or priority area designations for determining areas of herbicide treatment.

Until control technology provides more long-term efficacious control options, maintenance control of watermilfoil is expected to continue. Prevention of spread to new tributaries or impoundments will be the key to reducing conflicts and avoiding increased levels of control effort. Various public information programs have stressed the importance of preventing spread of aquatic weeds to new areas and the public sector has been cooperative by reporting suspicious infestations. The problem is compounded because watermilfoil is now found in an upstream tributary river (Holston River) and is expected to be an upstream propagule source for most of the watershed.

Aerial remote sensing has provided an assessment and planning tool in support of TVA's maintenance control program. Color prints made from "true color film" have been used routinely since 1975 to quantify aquatic macrophyte communities in infested reservoirs. Tonal, textural, and spatial characteristics allow the photointerpreter to distinguish watermilfoil from most companion submersed species such as spinyleaf naiad on large-scale photography (1:7,200 to 1:12,000); however, certain admixtures such as coontail are more difficult to separate (1). The use of aerial photography has been invaluable for planning and assessing weed control programs and is recommended for the aquatic plant manager.

Environmental Manipulation

Water level manipulation has been TVA's most effective tool for controlling submersed species such as watermilfoil by dewatering, overwatering, strandage, or by combination of these methods. For effective control of watermilfoil and other perennial species, an amplitude (difference between high- and low-water levels) within the annual cycle of about 2 m has been effective in reducing excessive populations. Short-term dewatering for 2-3 days during periods of freezing temperatures has also been effective for reducing watermilfoil infestations in Melton Hill Reservoir; however, this technique is not always adaptable to impoundments because of lack of inflow and regulating structures (6). Studies have also shown that watermilfoil is very susceptible to freezing temperatures and multiple exposures may improve the control (10)

Variations in timing have also been effective; a 1-week drawdown of a large TVA impoundment in July 1983 desiccated about 810 ha of shallow, littoral zone macrophytes including watermilfoil, naiads, pondweeds, coontail, and macroalgae. A narrow, relatively weed-free band occurred after refilling, and control effects extended into the following two growing seasons. Supplemental benefits of the summer drawdown included a reduction of seed densities of spinyleaf naiad, mosquito populations were reduced about 50 percent, and angler success for sport fish was reportedly improved. Based on this experience, a similar 1-week drawdown has been planned for aquatic weed control in TVA's 14,300 ha Chickamauga Reservoir in July 1985. Research has demonstrated that water level management can impact shallow submersed littoral species by causing increased silt deposition on plants, decreased photosynthesis, and cause a possible disruption of physiological adaption to a specific depth (11).

The preventative nature of waterlevel management has often been overlooked. Overfilling during peak propagule dissemination followed by a concomitant rapid drop in water level strands propagules on the shoreline where they can be desiccated. Obviously, design of structures, inflows, and outflows are necessary prerequisites for this scheme of water level manipulation.

Chemical Control

Herbicidal control of watermilfoil has been integrated with water level management control since the discovery of watermilfoil. After extensive field testing of numerous herbicide formulations and more than 20 years of field application in the TVA reservoirs, 2,4-D (2,4-dichlorophenoxy acetic acid) is considered the most economical, efficacious, and environmentally compatible herbicide for watermilfoil control. Treatment rates ranging from 11.2-44.8 kg a.e./ha of 2,4-D in areas of low dilution potential provide effective control of watermilfoil. Both granular (butoxyethanol ester [BEE] of 2,4-D) and liquid formulations (dimethylamine [DMA] salt of 2,4-D) have been used widely throughout infested reservoirs. The DMA 2,4-D formulation has been used for the last 15 years because of economy and ease of application even though both formulations produce similar control responses. One of the major problems with using 2,4-D in mixed communities is the selective nature of the herbicide. Watermilfoil is highly susceptible to 2,4-D and rapidly declines following treatment, while companion "resistant" species may survive and fill the niche previously occupied by watermilfoil. In TVA's control operations, a contact herbicide (e.g., endothall or diquat) is used when species other than watermilfoil exceed 50 percent dominance by areal cover. While field determination of percent composition is limited by visibility and experience of boat crews, the determination is important for effective and economical operations.

The BEE and DMA formulations of 2,4-D were registered by EPA, and tolerances of 0.1 ppm in potable water and 1.0 ppm "in or on the raw commodity fish" were established for TVA reservoirs in 1976 (3). Limited irrigation of agronomic crops and a lack of shellfish consumption in the Tennessee Valley made tolerances for irrigation water and shellfish unnecessary. Label restrictions do not permit treatment within one-half mile of potable water intakes. After 20 years of 2,4-D use to reduce excessive watermilfoil populations, there have been no documented problems relating to toxicity of 2,4-D to nontarget aquatic organisms, severe water quality deterioration, or complaints from the public (12). The slow decomposition of watermilfoil following herbicide treatment has not caused rapid nor excessive depletion of dissolved oxygen and fish kills have not been observed.

Mechanical Control

Techniques of reducing watermilfoil using mechanical equipment have not been considered feasible in the Tennessee Valley. High capital investment and high unit cost of operation of harvesters within shallow littoral zones with high densities of obstacles, disjunct distributions of colonies, and confined control areas around piers, docks, and other manmade structures are major drawbacks. Generation of fragments and rapid regrowth of watermilfoil and other weeds are other limiting factors.

Likewise, high unit cost and annual maintenance requirements restrict use of physical barriers, such as bottom screens, to high-use recreation areas such as swimming beaches. In TVA studies, regrowth of watermilfoil stems has occurred through venting slits of one bottom screen within two weeks after installation. While limited use of bottom barriers by shoreline residents and local recreation-based businesses has occurred on TVA reservoirs, current private use is practically absent because of the high costs and annual maintenance.

Biological Control

Various studies involving the use of pathogens, insects, and herbivores for watermilfoil control are largely experimental. The use of the herbivorous fish, grass carp (*Ctenopharyngodon idella* Val.), is one of the more promising biological control technologies. The herbivory of the grass carp on watermilfoil remains a needed research area. Limited control success has been reported at a few locations, such as Deer Point Lake in Florida (7)

A comprehensive 3-year TVA study of the grass carp's potential for weed control in a reservoir habitat and the associated impacts was initiated in 1983. Although hydrilla (*Hydrilla verticillata* (L.f.) Royle) and spinyleaf naiad were present in the study embayment, watermilfoil was the dominant submersed macrophyte. Results from this study will be used to assess the potential use of grass carp in a large weed-infested impoundment. The development of sterile, triploid genotypes of grass carp plus the expansion of hydrilla has necessitated consideration of this biological control technology for future use in TVA impoundments.

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CONTROL AND MANAGEMENT OF EURASIAN WATER MILFOIL IN THE PEND OREILLE RIVER, WASHINGTON

Harry L. Gibbons, Jr.*
Maribeth V. Gibbons**

*Kramer, Chin & Mayo, Inc.
1917 First Avenue
Seattle, Washington 98101

**Water
9515 Windsong Loop NE
Bainbridge Island, Washington 98110

A 3 year study was initiated in 1982 to define a management program in the Pend Oreille River for the control of Eurasian water milfoil. The study tested the effects of two concentrations of 2,4-D DMA (dimethyl amine salt of 2,4-dichlorophenoxy acetic acid) used in conjunction with two different adjuvants-a polymer and an inverting oil. It was determined that two annual applications of the herbicide + adjuvant were required to adequately control Eurasian water milfoil in a long term management program.

In the last decade, the occurrence and spread of Eurasian water milfoil (Myriophyllum spicatum) in the Pend Oreille River has posed serious management problems. While the presence of this old world species of rooted, submersed macrophyte was not recorded in the river prior to 1976, in the years following it began to increasingly dominate littoral zone areas upstream of the Box Canyon Dam (Figure 1). M. spicatum also seemed to be spreading upstream toward the river's headwaters, Lake Pend Oreille to the south. The invasion of Eurasian water milfoil not only intensified the aquatic macrophyte problem in the river but it also increased the public's awareness of the environmental problems that existed in the river. The citizens of Pend Oreille County became concerned about two things, the rapid increase in the abundance of weeds in the shallows of the river, and the dense growths of Eurasian water milfoil that interfered with many water based activities.

Eurasian water milfoil not only appeared to be replacing the native species of water milfoil and of other plants, but it also tended to colonize more area than was previously occupied by indigenous aquatic plants. The density of this plant precluded the use of boats and fishing in some stretches of the river. In response to this developing situation, the Port of Pend Oreille became the local sponsor of the research and development of management program for the control of Eurasian water milfoil in the Pend Oreille River. The Port was cognizant of the fact that this plant was causing economic losses to area resorts and other water-based concerns. For instance, the PUD, which owned and operated the Box Canyon Dam Hydroelectric Power Plant, had estimated costs of $1,000 per day due to the constant removal of Eurasian water milfoil biomass from its trash rakes. The cost

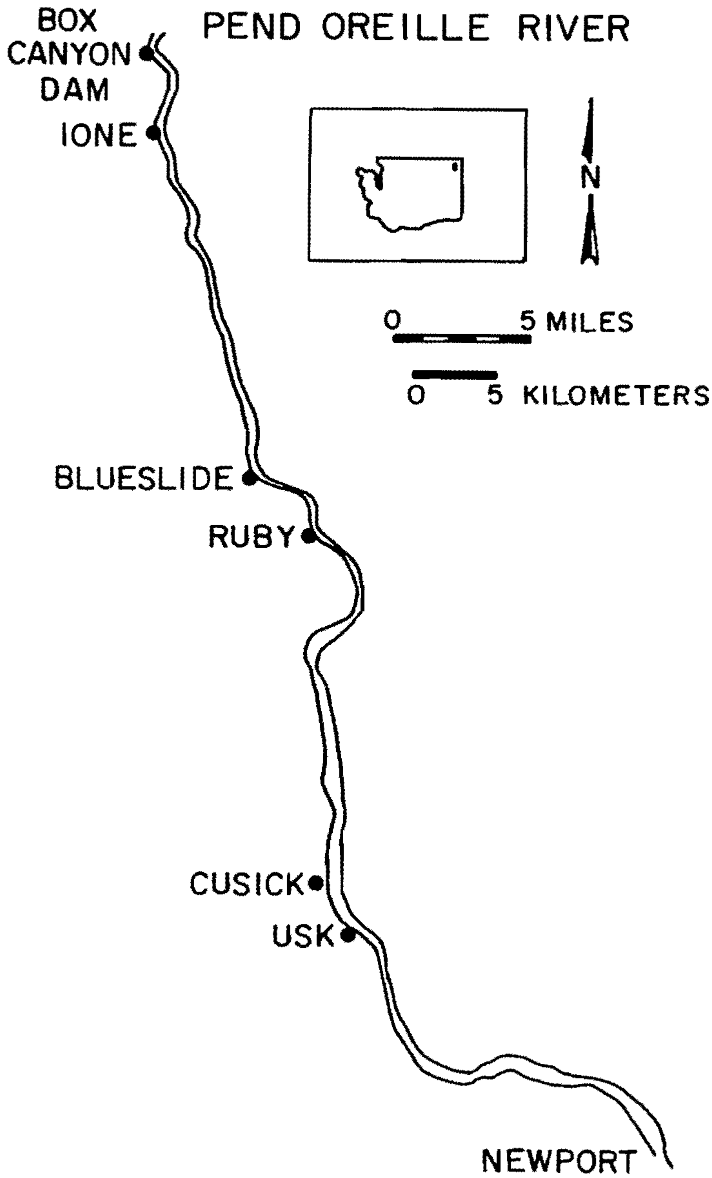

FIGURE 1. Portion of the Pend Oreille River under investigation.

estimate did not include the loss of electrical energy production that resulted from the decrease in hydraulic head over turbines.

With matching funds and technical assistance from the Washington State Department of Ecology, the Port of Pend Oreille enlisted the help of Washington State University to investigate different control options and to develop a long term management program for the control of Eurasian water milfoil in the Pend Oreille River. This work began in 1982 and carried over through 1983. In 1983, Pend Oreille County took over the local sponsorship of the program. The program grew from small test plots to the full scale treatment of 80 acres (32.4 hectares) in 1984.

The objectives of the first investigation were to review treatment alternatives for the control of Eurasian water milfoil in the Pend Oreille River, and to test the most promising control program available. The second year's effort was to refine the treatment methodology and to outline a management program. In 1984, carryover control was evaluated for the test plots treated in 1983, and the effectiveness of the full scale 1984 treatment was also assessed.

Study Area

The reach of the Pend Oreille River that was under investigation is located in Pend Oreille County, in the northeastern part of the State of Washington. The research, development, and control program covered the stretch of the river from just north of Cusick, Washington, downstream to the Box Canyon Dam (Figure 1). The test treatment plots for the 1982 2,4-D DMA application are shown in Figure 2, and the test plots for the 1983 herbicide applications are shown in Figure 3. The 1984 management applications were located in the high use areas of the river and at the southernmost extension of the Eurasian water milfoil beds near Cusick. The area near Cusick was treated in an attempt to reduce the upstream spread of this plant by stressing the uppermost populations of the plant. This will be discussed in more detail later.

Methods

After conducting a review of the available technologies for control of Eurasian water milfoil, it was decided that herbicide application was the best practicable treatment methodology for a control program in the Pend Oreille River. That conclusion was reached based primarily on the flowing water conditions in the River and the desire to limit the production of fragments that could potentially increase the spread of the plant.

In 1982, the existing coverage of Eurasian water milfoil was determined as well as the extent of upstream migration for the years between 1980 and 1982. In addition four test plots were employed in the investigation of the efficacy of 2,4-D DMA used with two different adjuvants. The two adjuvants used were an inverting oil and a polymeric carrier. Two of the plots were treated at a dose rate of 40 pounds/acre (45 kg/ha) and two plots were treated with a dose rate of 20 pounds/acre (22.5 kg/ha). Each adjuvant was used in combination with both the high and the low concentration of the herbicide.

In 1983, five test plots were used in a double application program at a dose rate of 20 lbs/ac (22.5 kg/ha) for each application. The second application was made approximately seven days after the first. The same adjuvants were used in this test as were used in the 1982 study. However, in the 1983 study, $MgSO_4$ was used in two plots in conjunction with the

TEST PLOT LOCATIONS

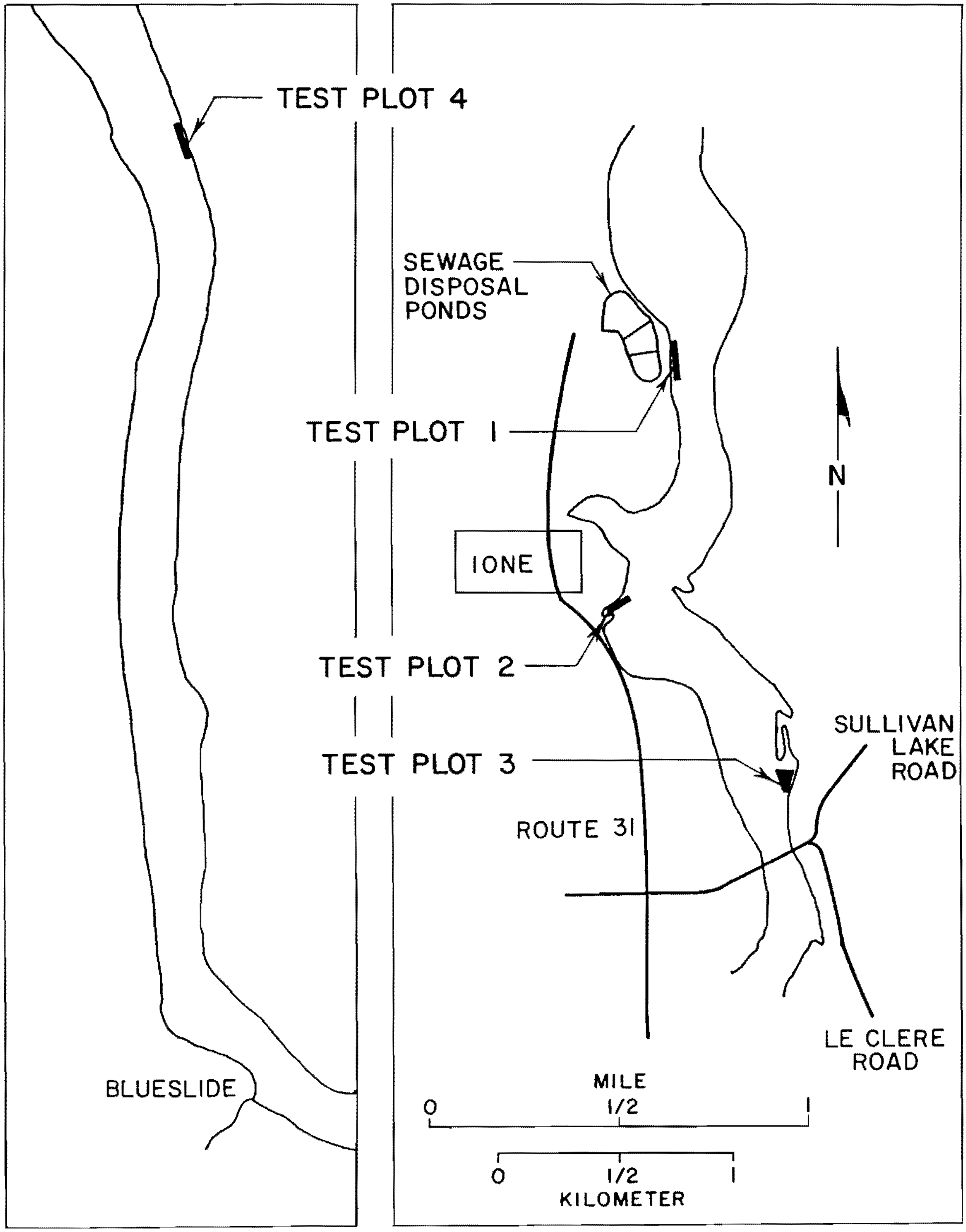

FIGURE 2. Locations of the 1982 test plots.

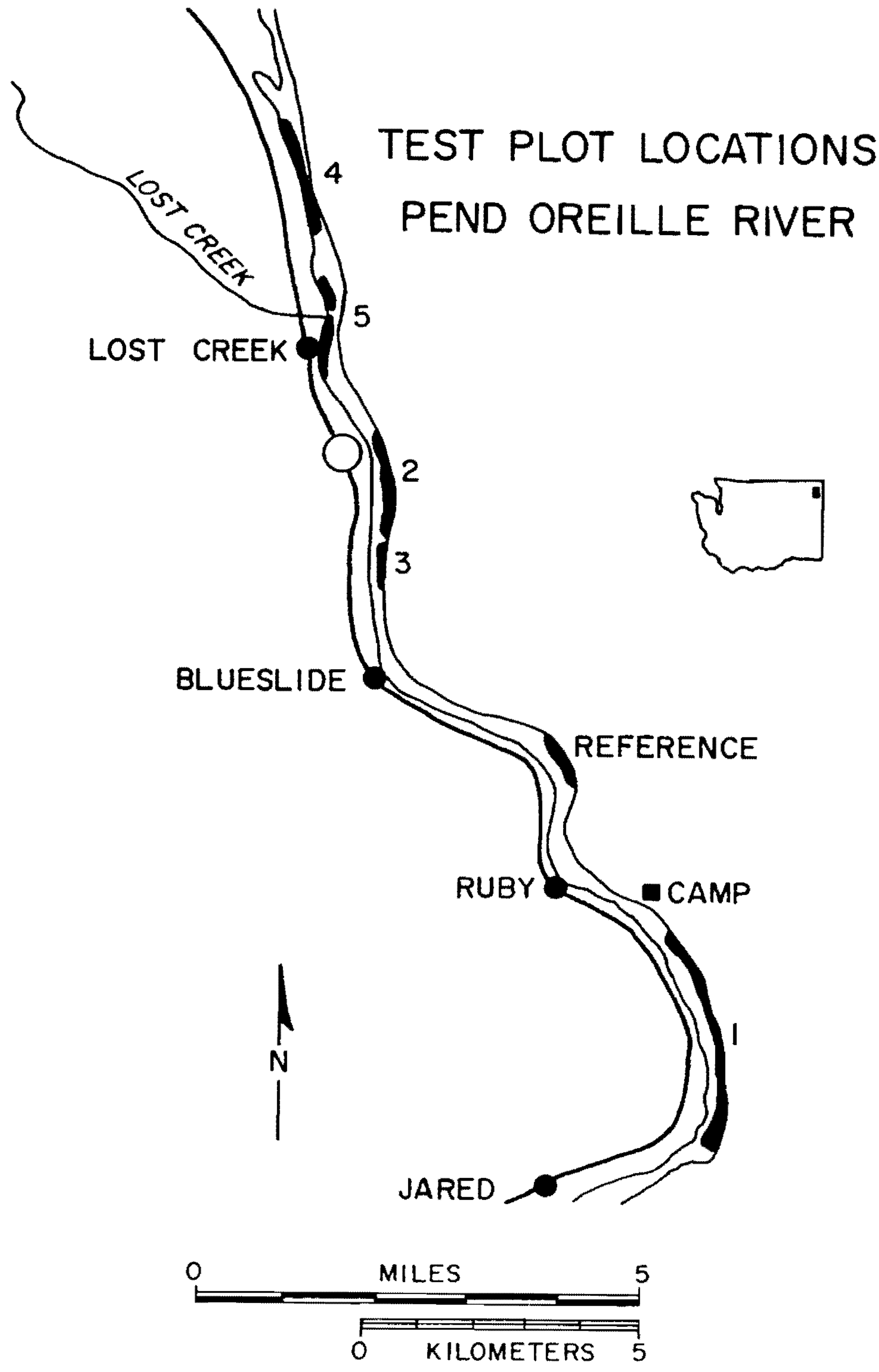

FIGURE 3. Detailed locations and configurations of reference plot and test plots identified by plot number for the 1983 tests.

inverting oil to promote sinking of this neutrally buoyant adjuvant. In one plot the $MgSO_4$ inverting oil mixture was used on both herbicide applications, while in the other plot the mixture was used only in the second application.

In 1984, all the treatment areas received the same application of 20 lbs/ac (22.4 kg/ha) of 2,4-D DMA with a 2% solution of polymer. The effectiveness of all treatments was evaluated by means of biomass measurements. For a more detailed presentation of the methods see I-5.

Results and Discussion

Within the Pend Oreille River study area, the aquatic macrophyte community is dominated by Myriophyllum spicatum, M. Exalbescens, M. verticillatum, Ceratophyllum demersum, Elodea canadensis, Potamogeton gramineus, P. pectinatus, and Ranaculus aquatilis. The littoral zone of the river is well developed along both the east and west shores of the river. It is apparent that Eurasian water milfoil is displacing the native vegetation as the ecological dominant plant within the river. In late summer of 1984 Eurasian water milfoil covered 50 to 100% of the available littoral bottom area within the study area. In 1982, it was estimated that this plant occupied 208 ac (84.3 ha). According to aerial photographs taken in 1980 by the U.S. Army Engineer Corps, Eurasian water milfoil covered 190 ac (77 ha). In the two subsequent years of observation, Eurasian water milfoil spread upstream 2.25 miles (3.6 km), to cover an additional area of 18 ac (7.3 ha). If that rate of upstream advancement remains constant, this plant will move 1.13 miles (1.8 km) upriver every year. The most likely mode of spread of Eurasian water milfoil in this river is through fragmentation, whereby plant pieces are carried upstream by wind action.

A significant finding of the 1982 study was that inadequate control of the target species was achieved with just a single application of the herbicide. This result was the same for both the low and high dose of 2,4-D DMA combined with either the polymer or the inverting oil. Two reasons may be given for this consequence. First, neither the inverting oil nor the polymer displayed the ability to hold the herbicide in the formulation on the surface of the plants for a period longer than 1 to 4 hours. Secondly and more importantly in terms of plant kill is the fact that Eurasian water milfoil was unable to absorb 2,4-D DMA after 4 hours of exposure (6). Hence, even though the herbicide may be present on the surface of the plants, it can not be taken up by the plant in significant concentrations to produce toxic effects after 4 hours of exposure. Therefore, the recommendation was made that two applications of the herbicide may be needed to more effectively impact Eurasian water milfoil. It was hypothesized that a much greater kill might be observed if a second application 2,4-D DMA was carried out during the initial period of regrowth following the first exposure to the herbicide. This was based on the assumption that the carbohydrate reserve in the root crown would be depleted resulting in the death of the plant by both direct toxicity and by exhaustion of energy stores.

In 1983 test plots were set-up to test this hypothesis and to assess the effectiveness of the adjuvants. Each of the five plots were treated twice with 20 lbs (22.5 kg/ha) of 2,4-D DMA + adjuvant, with the second application occurring approximately seven days after the first application. Table 1 presents the data from the five test plots treated in 1983.

2,4-D DMA on the plants may also become more pronounced when the density of the macrophyte beds are reduced due to the previous year's treatments. This increase in effectiveness will be a function of more even and increased contact of the herbicide with the plants.

During the carryover investigation of Eurasian water milfoil in the Pend Oreille River, research conducted by Verhalen (4) into the response of the aquatic insect community to the control program had three interesting results. One, no toxic effects of the 2,4-D DMA were observed in 1983 during the herbicide applications. Two, the aquatic insect community was found to be similar in treated and untreated areas one year following the herbicide applications. Three, the density of the insects was not related to the biomass of Eurasian water milfoil. In fact, statistical analysis showed that there was no correlation between total aquatic macrophyte biomass and insect populations. It must be concluded that the control of Eurasian water milfoil by 2,4-D DMA does not produce adverse changes in the aquatic insect community in the Pend Oreille River.

The long term management program for the control of Eurasian water milfoil began in 1984. That year 80 ac (32.4 ha) of a total coverage of more than 220 ac (89.1 ha) of Eurasian water milfoil was treated with 2,4-D DMA and the polymer adjuvant. The polymer was chosen to be used as the adjuvant in the long term control program because of the ease of employment versus the inverting oil. The overall result of the 1984 treatment was that Eurasian water milfoil biomass demonstrated a significant decline within the treatment area from 44 to 100% of the pre-treatment biomass levels. A decrease in Eurasian water milfoil was observed at the same time an increase in the densities of other aquatic macrophytes was evident. The observed increase in the native aquatic plant species was in response to the increase in available habitat area following the decline of Eurasian water milfoil. Full utilization of the littoral zone by aquatic macrophytes in the river is directly related to the nutrient characteristics of the Pend Oreille River.

It is hopes that at the end of a 5 year program that the coverage of Eurasian water milfoil will be reduced to less than 50 ac (20.3 ha). It is only through a long term management program that the desired control of Eurasian water milfoil may be achieved. This includes the dual application of 2,4-D DMA annually, until a better methodology for flowing water is developed.

Acknowledgments

The authors thank Mr. Ron Pine, Mr. Rocky Beach, and the faculty, staff and students of Washington State University for their efforts throughout these investigations. Logistical and funding support was provided by Washington State Department of Ecology, U.S. Army corps of Engineers, Washington State Water Research Center, Port of Pend Oreille, and Pend Oreille County.

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CHANGES IN THE AQUATIC PLANT COMMUNITY FOLLOWING TREATMENT WITH THE HERBICIDE 2,4-D IN CAYUGA LAKE, NEW YORK

Gary L. Miller
Environmental Studies Program
University of North Carolina
Asheville, North Carolina 28804

and

Margaret A. Trout
Department of Botany
University of New Hampshire
Durham, New Hampshire 03102

The submersed aquatic macrophyte, *Myriophyllum spicatum* L. (Eurasian water milfoil) has created a dramatic shift in aquatic plant populations in the Finger Lakes of New York as a result of explosive growth which began there in the early 1960's. In 1975 and 1977 the herbicide 2,4-dichlorophenoxy-acetic acid (2,4-D) was applied to a 36 ha experimental area to study the response of milfoil and associated plant community members. Post-application community response was then followed from 1975 through 1979.

The submersed aquatic macrophyte, *Myriophyllum spicatum* L. (Eurasian water milfoil) produced dramatic changes in the aquatic plant populations in Cayuga Lake, New York (largest of the Finger Lakes), when its explosive growth began there in the early 1960's. As in numerous other water bodies of the United States and Canada [Bayley, *et. al*. (1), Smith, *et. al*. (2), Reed (3), Aiken, *et. al*. (4)], this species rapidly increased in density and coverage and became the dominant macrophyte. By the early 1970's the northern end of Cayuga Lake was virtually a continuous bed of *Myriophyllum spicatum* [Baston and Ross (5), Miller (6)], where previously mixed populations of *Chara vulgaris* (muskgrass), *Vallisneria americana* (eelgrass), *Elodea canadensis* (waterweed), *Heteranthera dubia* (water stargrass), and various *Potamogeton* (pondweed) species occurred amid large areas of open water [Muenscher (7)]. Starting in 1975 the herbicide 2,4-dichlorophenoxyacetic acid (2,4-D) was applied to a 36 ha experimental area within the dense milfoil beds as part of a program to provide an open water channel for navigation and general recreation. The applications were made by the Seneca Co. New York Soil and Water Conservation Service.

Although the immediate effectiveness of herbicides such as 2,4-D in killing aquatic plants is well known, their long-term effects are much less predictable. Some studies report development of new macrophyte communities [Fish (8), Brooker and Edwards (9), Way, et. al. (10)], while others report continued regrowth of the target species [Goldsby, et. al. (11), Rawls (12)]. Since only a small portion of the Myriophyllum spicatum dominated area in Cayuga Lake was to be treated, it was not known if M. spicatum would continue to be the dominant macrophyte, or if changes in the community structure would occur. The purpose of this investigation was to determine whether significant shifts in macrophyte communities would result following the 2,4-D treatment.

Methods

Cayuga Lake is 60.7 km in length and 172 km^2 in surface area. The northern most 1600 ha are shallow, less than 4 m deep. At the commencement of this study, approximately 90% of this region was comprised of very dense M. spicatum populations, with the exception of a few sandy locations dominated by the macrophytic alga, Chara vulgaris (muskgrass) and intermittent mixed populations of the native species previously cited [Baston and Ross (5)].

Within the M. spicatum population, an experimental site was established consisting of three rectangular areas to be treated with 2,4-D. In addition, untreated areas, 300 m away, were established as controls (Figure 1). All of these sites were in the same water depth zone, sediment type, and macrophyte composition [Baston and Ross (5), Trout (13)]. On 25 May 1975 and 25 May 1977, the butoxyethanol ester of Aqua-Kleen 2,4-D (20% active ingredient) was applied at a rate of 100 kg/ha to the designated treatment areas.

Following the 1975 treatment and for the next five years, vegetation samples were removed from each of the two sampling sites within the treated area [the North and South Treatment Locations (NTL),(STL)] and two sites within the control area [the North and South Control Locations (NCL),(SCL]. Samples were taken from each of these four sampling sites every month during active macrophyte growth. Sampling was not conducted during the winter ice cover period. In the laboratory, plants from each sample were separated to species and the plant density, fresh weight and dry weight were determined. Simpson's diversity index [Simpson (14)], using Fager's (15) correction for direction of measurement was also calculated for each sample.

The plant density, fresh weight, dry weight, and diversity data were analysed by analysis of variance (ANOVA) and Duncan's multiple range test in order to isolate where significant differences occurred. For a more complete description of methods and statistical treatment of the data, refer to Trout (13). All references to statistical significance are based on these tests and p 0.05, unless otherwise stated.

Results

Results of the ANOVA demonstrated which factors were most important in accounting for the variation in the macrophyte density, dry weight and diversity index. For example, according to the ANOVA, significant differences were found for plant density means when they were separated by year, location, treatment, and most of the interaction terms involving the above factors (Table I). However, monthly means did not differ

Table I. Analysis of variance summary table for density.

Source	df	SS	MS	F
Locat.	1	54063.26	54063.26	9.72*
Year	4	993300.56	248325.14	44.65*
Treat.	1	42562.39	42562.39	7.65*
Month	9	52296.08	5810.68	1.04
Species	7	3160817.82	451545.40	81.18*
YxM	36	552.04	15.33	0.00
TxM	9	2650.95	294.55	0.05
YxT	4	149769.65	37442.41	6.73*
YxS	28	4018468.46	143516.73	25.80*
TxS	7	1896408.53	270915.50	48.71*
MxS	63	227199.96	3606.35	0.65
YxMxT	36	4656.52	129.35	0.02
YxMxS	252	178472.02	708.22	0.13
MxTxS	63	37158.22	589.81	0.11
YxTxS	28	1793434.80	64051.24	11.52*
YxMxTxS	252	137673.37	546.32	0.10
Error	6879	38262401.94	5562.20	
Total	7679			

*Indicates the factor is significant at alpha = 0.05.

Y - Year
M - Month
T - Treatment
S - Species
Locat. - Location
Treat. - Treatment

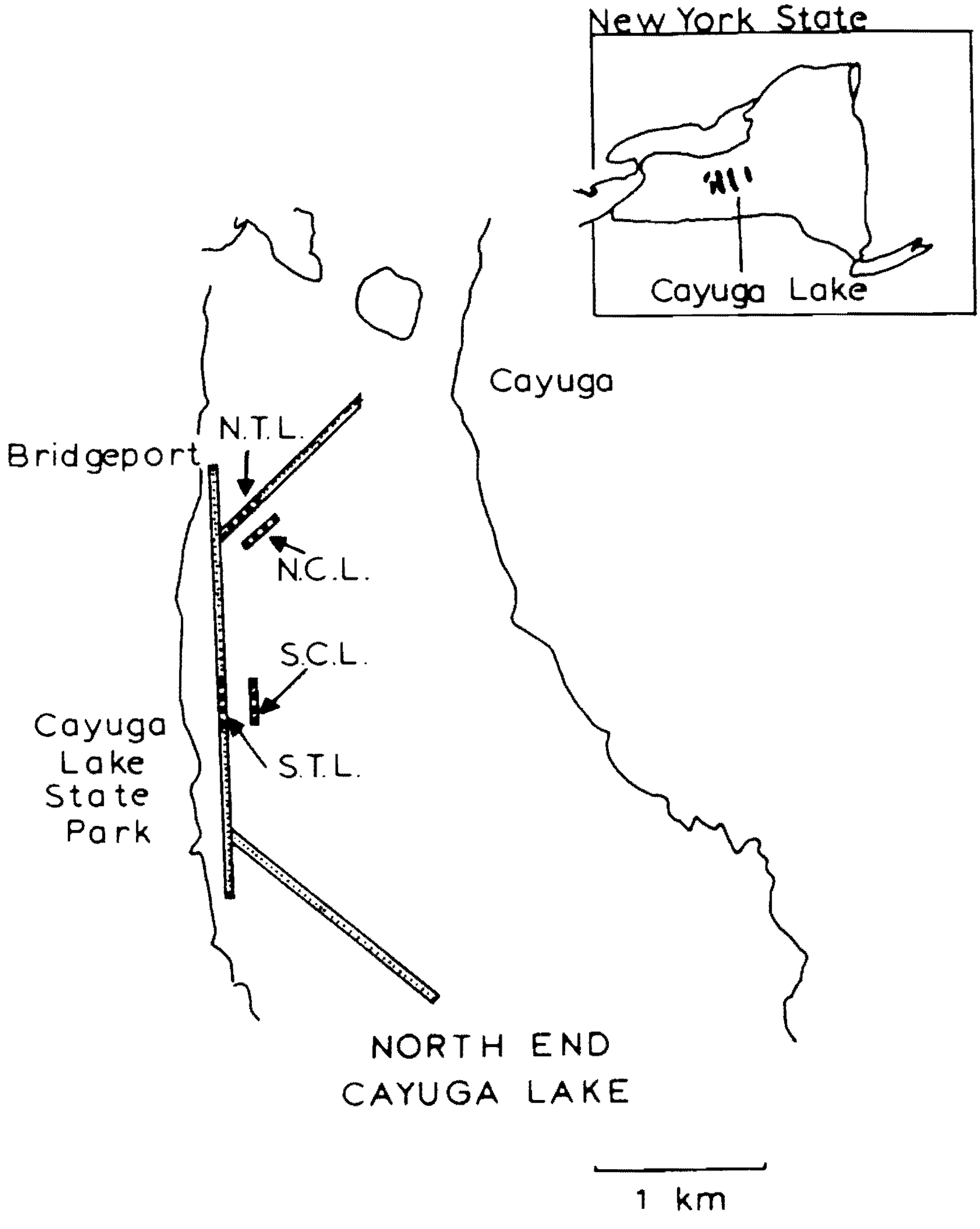


Figure 1. Experimental area (36 ha) in the northwest sector of Cayuga Lake (stipling). Vegetation samples were removed from 1.0 ha areas (large stipling) from both the North Treatment Location (NTL) and the South Treatment Location (STL). Control areas (NCL and SCL) of 1.0 ha were located approximately 300 m away.

significantly, nor did any interaction terms including month. Similar results were obtained for the dry weight and diversity index data. Also, in nearly every respect, the changes in macrophyte fresh weight agreed closely with dry weight changes and therefore will not be discussed below. Thus, the macrophyte community response to 2,4-D depended not only on the species under consideration, but also on the year of observation and the sampling site being examined.

No major shifts in the aquatic macrophyte populations of the control areas occurred during the study. M. spicatum remained the dominant macrophyte in each of the control sampling locations. Ceratophyllum demersum (coontail) was the only major species present in significantly greater dry weight in the North Control Location than in the South Control Location (t-test) (Table II). Since the dry weights in each of these sampling sites were not different for most species, all data from these two locations were combined in the figures to represent the macrophyte community in the untreated portion of the experimental area.

Table II. T-test results for dry weight analysis in control plots.

	NCL			SCL	
Species	Mean	Se		Mean	Se
M. spicatum	71.35	6.76		76.60	6.59
P. crispus	1.44	0.23		3.85	1.32
C. demersum	2.25	0.76	*	5.61	0.97
C. vulgaris	0.29	0.10		0.74	0.26
N. flexilis	3.71	1.28		2.27	0.18

* indicates a significant difference exists between the two means ($P > 0.05$) according to the t test. $N = 160$

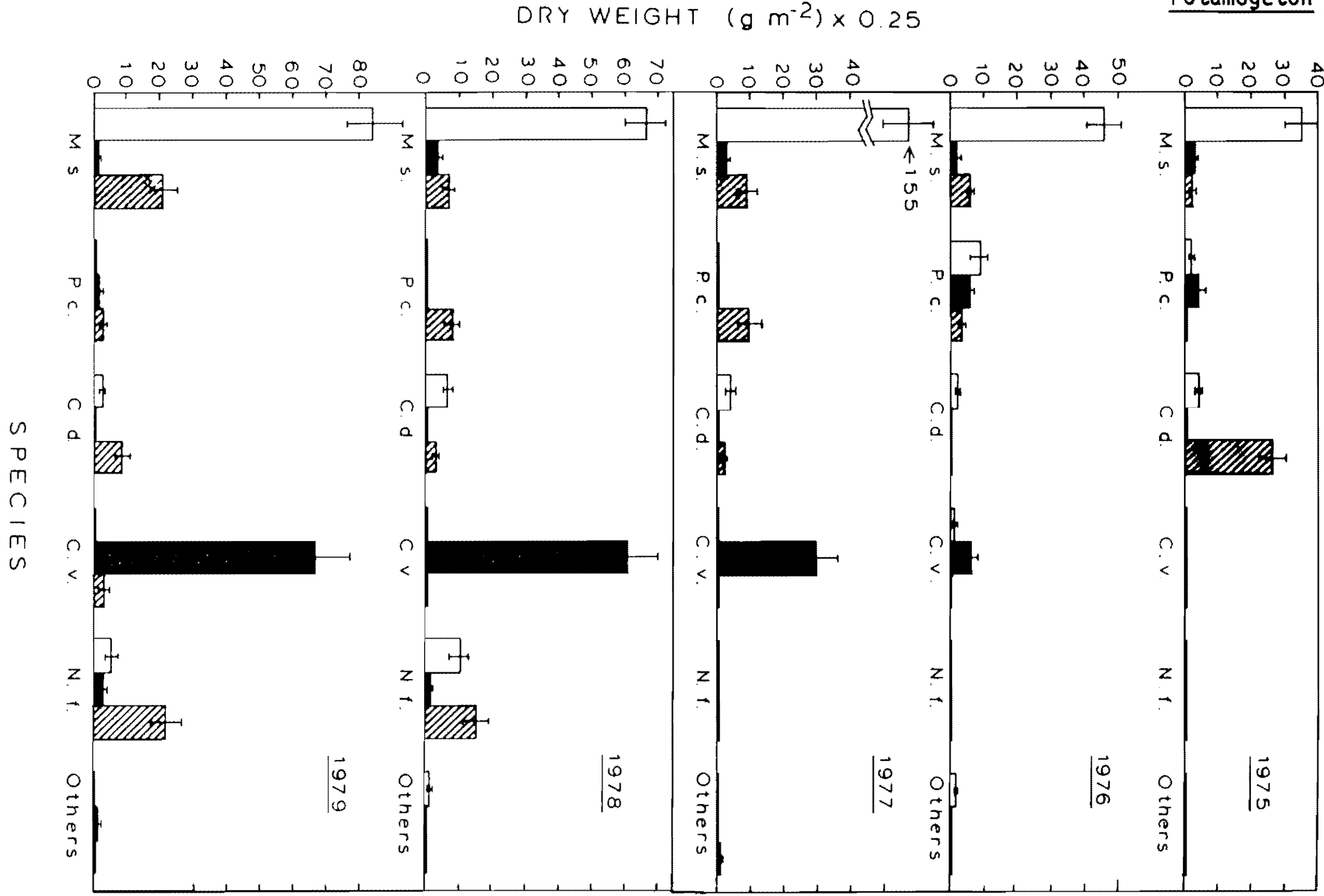
The dry weight of M. spicatum was significantly greater than any other species throughout the study in the control areas. Only one other species appeared in large amounts. Significant quantities of Najas flexilis (water naiad) appeared in control areas in 1978 and 1979 (Figure 2). It comprised 8.6% and 17% of the total macrophyte dry weight in these two years. Other species including Potamogeton crispus (curly leaf pondweed), and C. demersum occurred occasionally, but only in small amounts.

The species diversity index exhibited the tendency to remain intermediate between the North and South Treatment Location values. The mean species diversity index in the control area experienced significant yearly variations (Figure 3). The higher indices observed in 1978 and 1979 may have been influenced by the appearance of N. flexilis in those years. By maintaining normal Myriophyllum spicatum dominated populations, these control locations showed that there were no major shifts in dominance due to factors other than herbicide treatment.

In the North Treatment Location, a striking shift in macrophyte dominance occurred. Following the May 1975 application, no significant regrowth of M. spicatum or any other species occurred for the remainder of

Figure 2. Average dry weight of stems ($0.25m^2$) in control (open bars) and treatment (shaded bars) locations for the major species, 1975-1979 (a-e).

M.s. - *Myriophyllum spicatum*
P.c. - *Potamogeton crispus*
C.d. - *Ceratophyllum demersum*
C.v. - *Chara vulgaris*
N.f. - *Najas flexilis*
H.d. - *Heteranthera dubia*
P.p. - *Potamogeton pectinatus*
Others - *Najas minor*
Vallisneria americana
Elodea canadensis
Potamogeton richardsonii



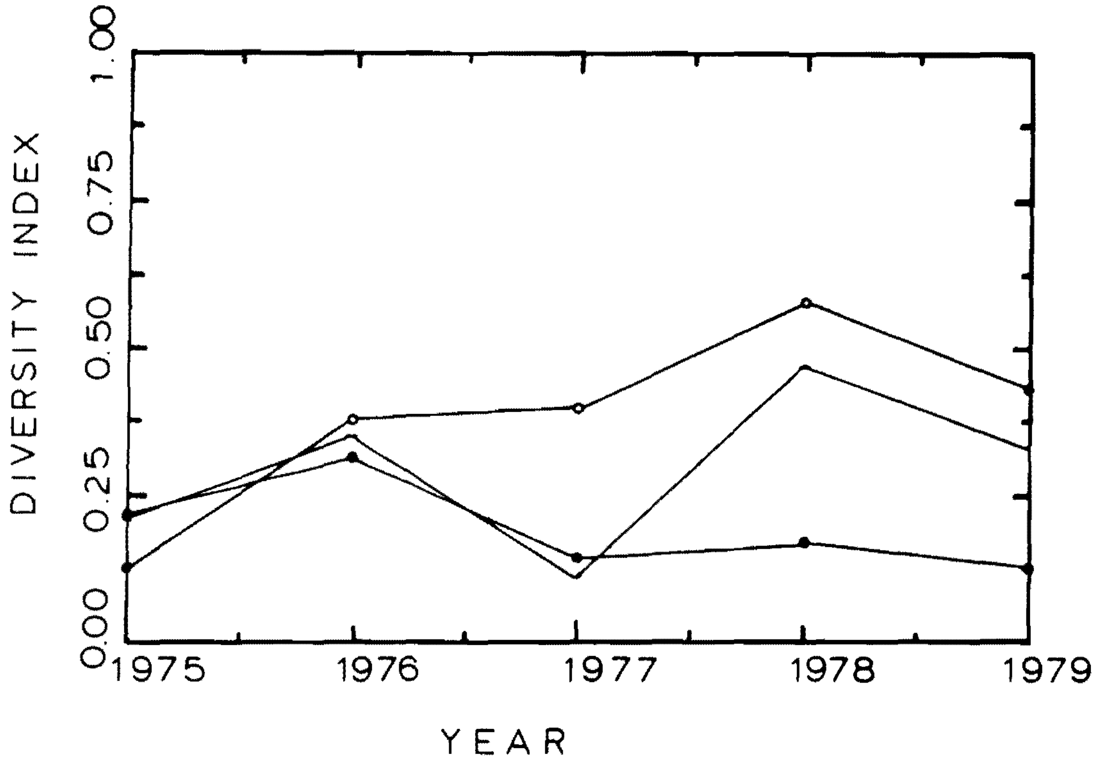


Figure 3. Mean values of Simpson's Diversity Index in the control areas (t), the North Treatment (●) and the South Treatment Locations (O) for the five study years 1975 - 1979. For each control point, n=10 and for each treatment point, n=5.

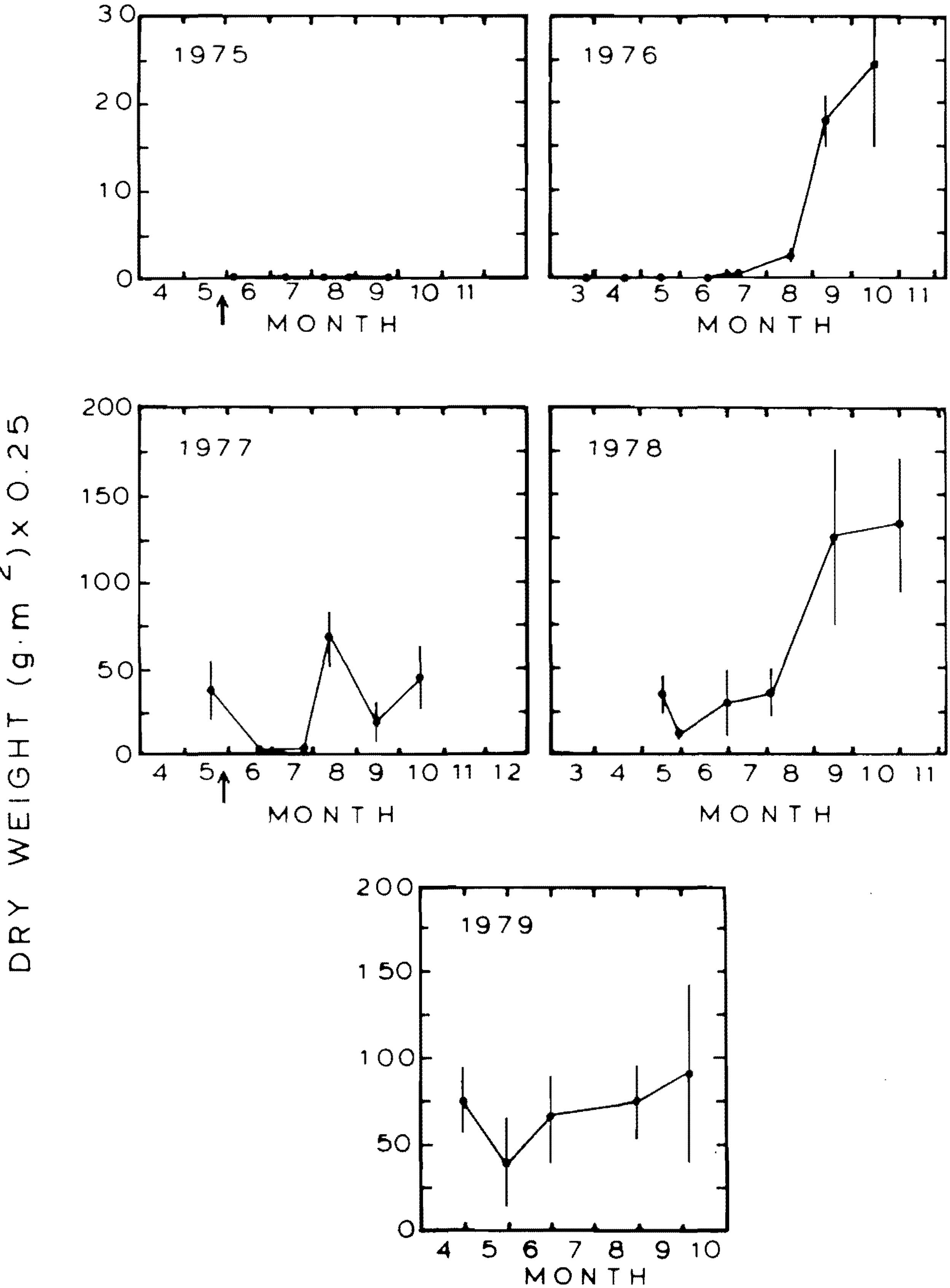


Figure 4. Mean Chara vulgaris dry weight (0.25 m^2) for each sampling period of the study 1975 - 1979. Vertical lines indicate +/- one

the year in this sampling location (Figure 2). However, in 1976, the alga, Chara vulgaris appeared and continued to appear in significantly greater quantities in each subsequent year of study. Following the first treatment, Chara remained the dominant macrophyte in the North Treatment Location throughout the study, exhibiting an average of 83% of the total macrophyte dry weight. In June, 1977, there was a decrease in C. vulgaris dry weight. This decline may have been induced by the 2,4-D application in May of that year (Figure 4). However, by mid-August, the population had recovered. M. spicatum did not recover to any degree comparable to pre-treatment or control levels. No patterns or trends were seen for any other species in the North Treatment Location following treatment. The species diversity index also remained very low throughout the study (Figure 3).

In contrast, species diversity indices were highest in the South Treatment Location. N. flexilis, C. demersum, and P. crispus were generally present in larger amounts in this sampling location than in the North (Figure 3). Significantly larger dry weights of N. flexilis appeared in 1978 and 1979 than were observed in control areas (Figure 2). Chara did not appear in significant quantities in any of the five study years in the South Treatment Location. M. spicatum showed greater recovery there than in the North Treatment Location, although still significantly below control levels. In 1979, higher dry weights of M. spicatum occurred in the South Treatment Location, perhaps suggesting a trend toward recovery of the species here. No such increase was observed in the North Treatment Location. An increase in the species diversity index was also observed in the South Treatment Location, following each 2,4-D treatment.

Discussion

While both sampling sites in the control area maintained similar aquatic macrophyte populations throughout the study period, contrasting changes occurred in the two treatment locations. Other investigators have reported shifts in macrophyte dominance following herbicide treatment [Fish (8), Brooker and Edwards (9), Newbold (16)]. An interesting ecological implication of this study was that the shift in species composition varied with site in spite of the lack of obvious differences in environmental conditions between sites. In Cayuga Lake, prior to any treatment, M. spicatum was the dominant macrophyte throughout the entire experimental area [Baston and Ross (5), Miller (6)]. C. vulgaris was not found in any of the experimental locations examined in 1975. It first appeared in August 1976 in a treated area. Similarly, Brooker and Edwards (9) and Newbold (16) reported that one year after herbicide treatment, Chara rapidly increased and became the dominant macrophyte.

Two theories have been proposed to explain how members of the Characeae become dominant plants following herbicide treatment. Fish (8) noted that large phytoplankton populations commonly develop in response to the release of nutrients from decaying macrophytes following herbicide application. Fish (8), citing Mayhew and Runkel's findings that Chara is less easily controlled by light exclusion than flowering plant species, concluded that phytoplankton densities sufficiently shade the water so that angiosperms cannot survive, thus affording Chara a competitive advantage. A contrasting hypothesis offered by Brooker and Edwards (9) proposed that high concentrations of calcium and magnesium associated with Chara surfaces prevent the entry of the herbicide. A third explanation is that Chara does not respond to the growth hormone 2,4-D as do angiosperms and thus are not affected by the treatment. In addition, in this present study, it appears that competitive interactions between C. vulgaris and M. spicatum could result in the same consequence. For example, a reduction in the M. spicatum population could give an advantage to C. vulgaris by providing more space and light. Once established, the very dense and nearly continuous layer by Chara may make it difficult for other species to anchor in the sediment (Miller - personal observations via SCUBA). In order to be certain of the reasons for the Chara shift, more experimental work is required.

The significant differences in the long-term regrowth response of macrophytes in the two treatment locations suggest that in addition to the possible dependent competitive advantages which Chara may have following herbicide treatment, other location-related factors must also control redevelopment of macrophyte communities. One possible explanation for this response is that one location received a lower dosage of the herbicide. This is unlikely, however, since application was continuous throughout the treated area and was conducted during very calm conditions. Other factors such as site differences in weather conditions, water depth or sediment type, do not apply here, since both locations were essentially identical in these respects.

A simplistic hypothesis is that chance introductions just after the time of treatment determine the species differences observed at each location. The probability of introduction would be most related to the type of communities adjacent to the treated areas. This could provide an opportunity for plants transported into a location to develop there. Supporting this hypothesis is the close proximity of the North Treatment Location to substantial Chara beds. Differences in plant associations in each location after treatment might be dependent on such chance events. The location of nearby proximal seed sources has been well documented for terrestrial succession by Keever (18) and others.

A general model of the effects of herbicide application on aquatic macrophyte communities, based on the findings from this study, other studies and general ecological principles, is proposed in (Figure 5). As indicated in the model, the application of an herbicide may open up the treated area for colonization by new communities to a degree comparable to catastrophic events such as fire or plowing in terrestrial systems. The pioneer community which develops depends on several factors such as nutrient release from decaying plants and increased light penetration [Newbold (16), Brooker and Edwards (9)]. Although high phytoplankton densities may follow herbicide treatment [Fish (8), Brooker and Edwards (17)] and then inhibit macrophyte development [Montgomery (19)], often there is a rapid regrowth of macrophytes [Brooker and Edwards (19), Rawls (12)].

Three general types of macrophyte regrowth communities have been reported following treatment (Figure 5). The first is regrowth of the target species in dense, monospecific stands, often creating the need for reapplication of herbicide [Rawls (12),Goldsby, et. al. (11), Hestand and Carter (20)]. The probability of this type of occurrence would increase if there was survival of rootstocks of the treated plants. The second type of

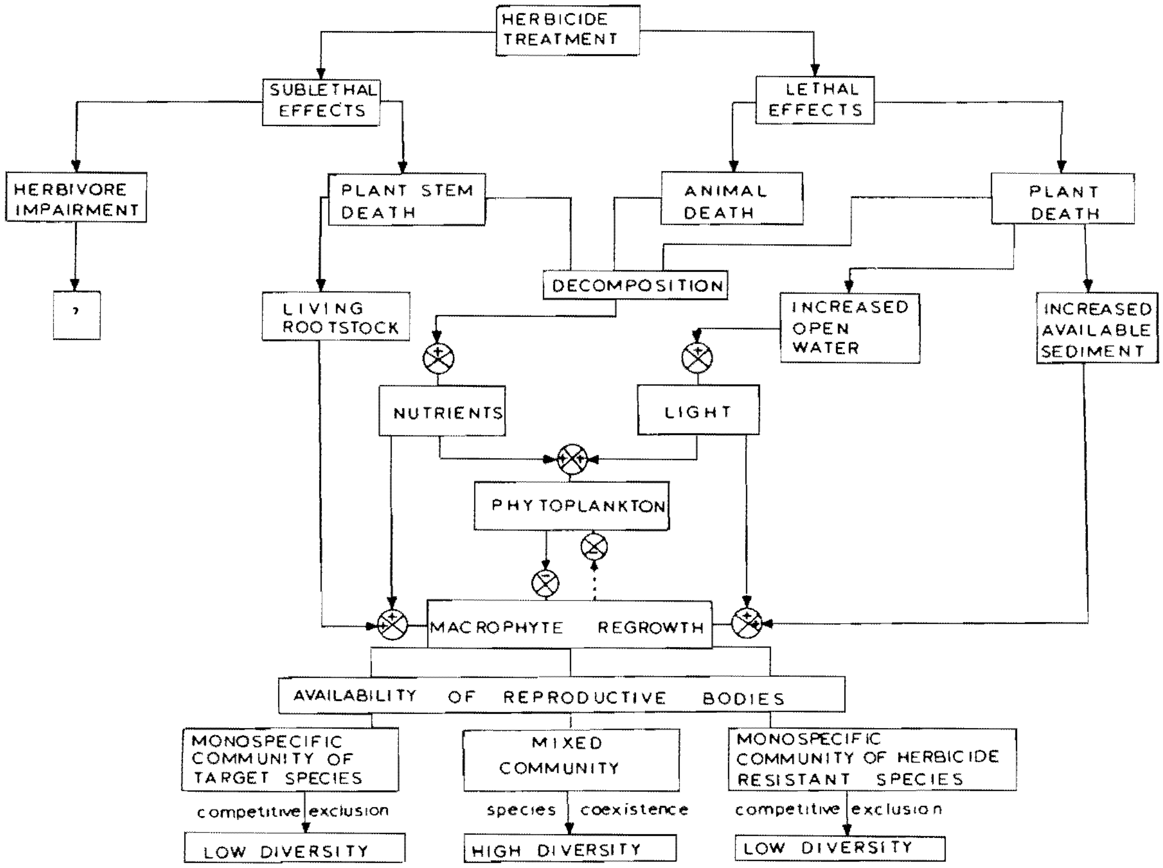


Figure 5. Model describing some major effects of herbicides on aquatic macrophytes.

macrophyte regrowth community is dominated by a species resistant to the herbicide [Fish (8), Brooker and Edwards (9), Newbold (16)]. Characteristics possessed by the dominant species in both of these communities might include the ability to rapidly disceminate reproductive bodies and the possession of a high growth potential. The dominance of Chara in the North Treatment Location appears to be the second type of community. Each of these two community types would be characterized by low species diversity. However, in a third type, a mixed macrophyte community, high species diversity would result. Here both the target species and/or resistant species, as well as other species, may be present. This type of community was present in the South Treatment Location of this study following the second 2,4-D application.

A factor which could greatly influence the type of community which develops is the temporal and spatial availability of macrophyte reproductive bodies of Chara to the treated areas. For example, if reproductive bodies of Chara in nearby untreated areas are mature at the time of treatment, they may be frequently introduced into the open region. Additionally, if any plant fragments were present at the time of treatment, the resistance factor may have increased the chances for Chara's establishment. Thus, the availability of sexual and/or asexual reproductive bodies from peripheral populations to treated area may lead to the establishment of a mixed macrophyte community. The proximity of the South Treatment Location to sites of P. crispus (whose turions are available during June in Cayuga Lake) supports this idea.

If it is the proximity of reproductive bodies to a treated area that influences the composition of the succession community, chemical management programs should consider leaving untreated areas with preferred species populations as sources of recolonizing material. Where practical the timing of herbicide applications should correspond with the maturity of reproductive bodies such as turions produced by desired species in "seed stock" areas. In addition, there exists the possibility of introducing desired species such as Chara following treatment. Other management issues which must be kept in mind are the careful management of species to avoid natural vectoring of plant propagules and human recreational activities, such as motor boat propeller cuttings which also serve as very effective vectoring agents.

In situations as described in this paper, small area treatments of 2,4-D (or other herbicides), coupled with the aggressive regrowth of native species should provide adequate open water for general recreation. By more thoroughly understanding the dynamics of plant reproduction and succession, along with the importance of the timing of the application of herbicides, aquatic plant managers may be able to significantly reduce the use of herbicides in aquatic ecosystems by as much as 50%.

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A REVIEW OF EURASIAN WATER MILFOIL IMPACTS AND MANAGEMENT IN BRITISH COLUMBIA

PETER R. NEWROTH

Manager, Littoral Resources Unit
Resource Quality Section
Water Management Branch
Ministry of Environment, Parliament Buildings
Victoria, British Columbia
Canada V8V 1X5

Eurasian water milfoil (Myriophyllum spicatum L.) was first reported from British Columbia waters about 1970. Since 1972, the B.C. Ministry of Environment has implemented comprehensive aquatic plant management throughout the province, concentrating on control of this plant and reduction of adverse impacts. This paper reviews impacts on water quality, recreation, hydrology and fisheries and describes phases in the development of control programs. The results of preventive programs, surveillance and aquatic vegetation mapping, public information and mechanical, chemical and biological control are summarized.

Public concerns about Eurasian water milfoil (Myriophyllum spicatum L.) were first voiced in British Columbia, Canada, in 1971, when populations of this plant became a nuisance in Okanagan Lake, near Vernon. Since then, this perennial, rooted aquatic plant has spread rapidly downstream through all the mainstem Okanagan Valley lakes and into the Columbia River system in the United States. Also, this plant has spread to other watersheds in British Columbia, mainly through transportation on boating equipment.

The Water Management Branch of the British Columbia Ministry of Environment has coordinated and implemented extensive aquatic plant management programs since 1972. Objectives of these management activities have included:

a) inventory and mapping of the native aquatic flora in provincial waters and documentation of spread of Eurasian water milfoil,
b) study of the ecology and biology of Eurasian water milfoil, and evaluation of habitats most conducive to its growth,
c) assessment of adverse impacts of Eurasian water milfoil on the aquatic resources of the province,
d) evaluation and documentation of the effectiveness and environmental safety of all known and practical control methods for Eurasian water milfoil, and development or encouragement of novel control technologies,

e) reducing the rate of spread of Eurasian water milfoil to previously non-infested water bodies, and
f) providing technical advice, designing cost-effective control programs and providing major funding support and aquatic plant control equipment to assist local agencies in reducing nuisance aquatic plant populations.

This paper reviews the impacts of Eurasian water milfoil in aquatic systems in the province, phases of the British Columbia aquatic plant management program and preventive and responsive management techniques that have been applied. The objectives and benefits of these approaches are discussed.

Administration, Program Phases and Policy

The Water Management Branch of the B.C. Ministry of Environment administrates the licencing of surface water throughout the province as well as providing for flood control and water quality studies. Since 1972, this Branch also has assumed responsibility for aquatic plant management and has responded to concerns about nuisance aquatic vegetation from the general public, tourism industry and regional administration agencies. Through the production of over 60 technical reports and information summaries, and participation in many public meetings, national and international workshops and technical symposia, the Water Management Branch has publicized and shared the experience gained from a diverse and comprehensive management effort.

Three major phases of the British Columbia aquatic plant management have been identified; they reflect the gradual emergence and spread of the Eurasian water milfoil problem and changes in policy over time:

1. Documentation Phase (1972-1975). During this period, major adverse impacts of Eurasian water milfoil on water use became recognized by the public, politicians and Ministry of Environment staff. Preliminary testing of mechanical harvesting and the herbicides diquat and paraquat was performed and extensive literature reviews were completed. Meanwhile, rapid spread and expansion of Eurasian water milfoil colonies was documented and the potential for widespread problems in many British Columbia water bodies was recognized.
2. Control Development Phase (1976-1980). The work force assigned to test, develop and apply control technologies expanded during this period, and major capital investments were made to acquire four harvesting machines and other mechanical control and support equipment. Thorough monitoring of trials with this equipment, experimental use of the herbicide 2,4-D, aquatic plant documentation in infested and about 1 000 non-infested water bodies and ecological work on Eurasian water milfoil was performed in this period. Also, preventive measures designed to reduce the rate of spread were applied in connection with a major public information campaign.
3. Maintenance Phase (1980-Present). Cost-share agreements were developed with several local groups to support cost-effective control programs designed to minimize public nuisances caused by Eurasian water milfoil. Under these agreements, the Ministry provides technical assistance, major control equipment and 75 percent of operational funding; local agencies provide administrative and political support and 25 percent of operating funds, and priorize treatment areas.

Total funding allocations by the Ministry of Environment for aquatic plant control and staff assigned to this project were reduced drastically in 1982-1984, with the result that research and development were suspended and only limited funding was available for capital acquisitions (new control equipment). However, expanded programs are planned in 1985.

The present policy of the Ministry of Environment has evolved so that funding assistance may be provided on a priority basis in cases where formal requests are made and local political support is present, where a serious nuisance has occurred and aquatic plant control will provide benefits, where local authorities are prepared to provide financial support and if provincial funds are available. The financial formula used at the present time requires that a minimum of 25 percent of operational funds be provided by local agencies. Experience to date indicates that the cost-share approach encourages reasonable and affordable control programs. Each year, the ongoing control programs are monitored by Ministry of Environment staff, and evaluations of local needs are made. Advisory committees include local politicians to help establish priorities for ongoing management and to encourage public support. An example of the operation of a cost-share program is provided in detail by Truelson (1).

Adverse Impacts of Eurasian Water Milfoil in British Columbia

Unwanted aquatic plants may cause a variety of nuisance problems. Although many complaints are received by the Ministry of Environment each year because of problems caused by *Potamogeton* spp., *Ceratophyllum* and *Nuphar*, Eurasian water milfoil problems are usually the most severe.

Rapid, dense growth characterizes Eurasian water milfoil, which often forms mats and clumps at the surface. Also, since this plant often prefers habitats frequented by man (shallow shorelines and embayments) or areas modified for public use (e.g.: breakwaters and marinas) it is often perceived as a major threat to traditional water use. In British Columbia waters, the main adverse impact of Eurasian water milfoil populations has related to recreational use, although other resource values have been affected; some of these concerns and experiences are reviewed in the following sections.

Impacts on Water-based Recreation. The highest priority for control of Eurasian water milfoil is in public recreation areas and especially in beach areas utilized by tourist visitors. Historically, most of the areas in the Okanagan Valley lakes, Shuswap Lake and in Cultus Lake (where most control treatments are ongoing) were not subject to nuisance aquatic plant conditions prior to the establishment of Eurasian water milfoil in the 1970's. Boating activities in some of these areas, including use of motors, sailing vessels with keels and water skiing, would be curtailed unless surfacing Eurasian water milfoil were removed or controlled. Shore-based angling is adversely affected; trollers in mid-lake also encounter mats of floating Eurasian water milfoil which catch on lines and lures.

Beach use is the most important aspect of recreation for both residents and tourists to the Okanagan region; 80 percent of tourist vacation days are spent at the beach. Surveys comparing Okanagan beach use in 1970 (prior to Eurasian water milfoil infestation) with use in 1980 indicated that aquatic weeds had become one of the main problems to residents and visitors in 1980 (2). Eurasian water milfoil adversely affects the quality of beaches by fragment accumulation along the water's edge, by spoiling the aesthetic quality of the offshore water and by increasing the safety risk for swimmers. The dense growth associated with untreated Eurasian water milfoil populations may contribute to drowning tragedies and the increased vegetation has been associated with "swimmers itch" problems.

Many desirable waterfront residential areas on Okanagan Valley mainstem lakes front on beaches affected by Eurasian water milfoil. Property assessments for taxation purposes may have been reduced in some areas because of the adverse impacts of aquatic weeds. Other examples of adverse impacts include growth of Eurasian water milfoil in Salish Park Pond (Chilliwack) and in a pond in the Kelowna Golf Club. In both cases, the surfacing plant population and associated algal mats spoiled the aesthetic quality of the ponds; in the golf club the excessive plant fragments created an additional nuisance by blocking intakes used for irrigation.

Impacts on Water Use and Hydrology. The Peachland Irrigation District utilizes water from an upland stream for domestic and irrigation supply; a small stilling basin at the point of diversion is infested with Eurasian water milfoil. A dredge was used to remove accumulated silt in 1975, and it is probable that Eurasian water milfoil was introduced with this machine, which previously had been used for Eurasian water milfoil control in Okanagan Lake. This Irrigation District has attempted control of the well established plant population because of concerns about taste and odour of the water being supplied.

Eurasian water milfoil has demonstrated ability to grow luxuriantly in rapidly flowing water and may create hydraulic resistance. In the Okanagan River and lakes system, which is regulated by dams to prevent flooding and to provide water storage, Eurasian water milfoil has interfered with regulation and control of discharges by threatening accurate measurements at stream gauging stations. Also, in a stretch of the Okanagan River downstream from Vaseux Lake, Eurasian water milfoil grows so densely that minor flooding can occur in the lake; harvesting is required annually to control the problem.

Impacts on Aquatic Ecosystems. Eurasian water milfoil now is estimated to occupy about 1 000 ha of littoral area in watersheds in the Okanagan, Shuswap and Lower Fraser River drainages. Documentation by aerial photography and ground-truth mapping confirmed that this exotic species dominated native vegetation as well as occupying habitats previously not supporting other macrophytes (3). Maxnuk confirmed that even "weedy" species such as *Potamogeton crispus* L. could be suppressed by Eurasian water milfoil within one to two growing seasons (4).

Impacts of Eurasian water milfoil on fishery resources were studied at an early stage of the Okanagan Valley infestation; at that time little evidence was obtained to confirm adverse effects on salmonid fish or waterfowl (5). Salmonid fish were not observed to frequent littoral areas during the summer period, although non-salmonid species were present. A survey in 1979 showed that substantial numbers of juvenile non-salmonid fish (e.g.: perch, crappies, sunfish, bass) are trapped in vegetation harvested in Okanagan Valley lakes; important game fish species were rarely caught. The implications of ongoing harvesting and expansion of Eurasian water milfoil to predator-prey relationships and food chains have not been explored in British Columbia.

Eurasian water milfoil has now reached densities which interfere with some shore and river spawning salmonid species. In Cultus Lake, Eurasian water milfoil populations on gravel spawning areas for sockeye salmon were treated with a rotavator in 1983; additional treatment is recommended for 1985. Unless controlled, the plants interfere with spawning by covering spawning gravels and it is possible that, over time, accumulation of organic material and gravel compaction could cause further deterioration. In the Okanagan River downstream of Okanagan Lake, salmonid spawning areas also are being occupied by Eurasian water milfoil, leading to some reduction of suitable spawning habitat.

Impacts on Water Quality. Effects of Eurasian water milfoil on nutrient levels in mainstem Okanagan Valley lakes through mobilization of sediment nutrients by uptake in vegetation have been studied (6). Phosphorus release from living tissue does not appear to have a measurable effect on water quality. However, the contributions from tissue sloughing during the growing season and year-end senescence represent a significant internal loading (ranging from 0.2 to 2.8 percent of total external phosphorus for individual Okanagan lakes). This plant has the potential to seasonally release more phosphorus to this lake system than individual sources such as storm sewers, industrial and fertilizer sources could contribute. In consideration of efforts to maintain and improve existing water quality in Okanagan Valley lakes, the control of Eurasian water milfoil growth, through management activity such as harvesting, appears to have an important role.

Control Technologies and Management Approaches Applied in British Columbia

Control Technologies. The following section provides updated information on methods and approaches applied since 1972 in British Columbia to manage Eurasian water milfoil (7-8). In addition to numerous known control methods the Ministry of Environment has solicited suggestions from the private sector, including inventors, for innovative methods to control Eurasian water milfoil. In 1977, the Ministry of Environment sponsored a National Design Contest to encourage submission of new designs for control devices. Unfortunately, this contest did not result in any practical, new equipment. More than 25 inventors have since contacted the Water Management Branch with ideas and in some cases prototypes for testing. Although the Water Management Branch cannot contribute financially to the construction of prototypes, promising designs are observed in operational testing and a preliminary evaluation is given on effectiveness and environmental impacts. Most designs have involved mechanical removal of roots and/or shoots but hydraulic, electronic and various other methods have been reviewed. One of the most promising of these approaches has been developed by Soar (9). The main criterion for further development relates to overall cost effectiveness. The new design is compared with existing control equipment, which is being used operationally, and only approaches which may be used more economically or to meet needs not now fulfilled are being encouraged.

Major control technologies being used at the present time are listed in Table I. Unfortunately, no use of the herbicide 2,4-D BEE is integrated with the mechanical control programs now organized to deal with Eurasian water milfoil problems. Water Management Branch extensively tested this herbicide in the period 1976-1982 (10). Use of 2,4-D was beneficial in treating dense infestations in Osoyoos Lake; an enclosed area was treated with 2,4-D BEE in 1981 and no harvesting was required until late in 1984. In another area (Champion Lake, Kootenay Region), application of 2,4-D BEE in 1981 controlled the initial infestation of Eurasian water milfoil. This treatment minimized spread to adjacent areas; follow-up treatments using bottom barriers and hand-picking by SCUBA divers prevented further infestation and no nuisance populations have developed.

The use of biocontrol agents for Eurasian water milfoil has been widely considered in North America, but the only promising agent for use in British Columbia was discovered during ecological research by Water Management Branch (11). Observations on the feeding behavior of a chironomid larva (*Cricotopus myriophylli* Oliver) indicated that this organism damaged and suppressed Eurasian water milfoil colonies; if larger populations of this control agent can be artifically encouraged then another important control tool will be available.

Management Approaches. In addition to the unusually broad range of control technologies considered, tested and developed, Water Management Branch has become involved in a variety of management situations requiring different approaches and combinations of technologies. Information on four major ongoing control programs is presented here.

Okanagan Valley. Table II summarizes the methods used in ongoing control programs applied since 1975 in the mainstem Okanagan Valley lakes (Wood, Kalamalka, Okanagan, Skaha, Vaseux and Osoyoos). Since 1981, the program has operated under a cost-share agreement between the Province (75 percent) and the Okanagan Basin Water Board (25 percent). The initial infestation of Eurasian water milfoil in British Columbia is believed to have occurred in Vernon Arm of Okanagan Lake, probably about 1968 or 1969. As indicated elsewhere in this paper rapid spread downstream and to adjacent water bodies occurred during the period when the potential impacts were not appreciated and practical control approaches were not understood or developed. Efforts to develop integrated control programs in the 1977-1980 period were frustrated by circumstances including:

a) Eurasian water milfoil was widely distributed over hundreds of hectares of littoral area.
b) Use of herbicides was limited to experimental test areas due to concerns about adverse impacts and lack of public and local political support.

TABLE I. Estimated Operating Costs and Treatment Rates for Selected Eurasian Water Milfoil Control Methods in British Columbia

Method	Operating Cost[1] (cost/hectare)	Average Rate[2] (hectares/day)	Comments
1. Diver Dredge	\$2 500/ha to \$19 000/ha (capital cost/dredge unit about \$12 000)	.20 ha/day extremely sparse .02 ha/day moderate .01 ha/day very dense	Treatment costs and rates are highly dependent on weed density.
2. Rototiller	\$400/ha to \$1 200/ha (capital cost/ machine about \$80 000)	0.2-0.5 ha/day	Cost and rate depend on the degree of difficulty in treating the site and the number of passes necessary to remove most of the plant material.
3. Shallow Water Tillage	\$400/ha (capital cost/ machine about \$60 000)	0.7 ha/day	Cost may increase in soft substrate where obstacles are present
4. Harvester	\$1 200/ha (capital cost/ machine about (\$55 000)	0.4 ha/day	Rate includes shore disposal of spoils.
5. Bottom Barriers	Burlap-\$12 200/ha Polyethylene- \$13 000/ha Window Screen- \$20 000/ha Texel-\$15 000/ha	.05 ha/day	Polyethylene, window screen, and Texel may be reusable and their high cost could likely be amortized over several years.

[1]Figures do not include rental or depreciation of capital costs of machinery, expenses incurred from transport/launching of machines, or administration costs. Costs in 1984 Canadian dollars.
[2]The above rates apply to an 8 hour day and include set-up time.

In most Okanagan Valley lakes, containment of Eurasian water milfoil became impossible; it was recognized about 1980 that only cosmetic or semi-intensive management was possible. In the case of Kalamalka Lake, where conditions appear to inhibit rapid growth of Eurasian water milfoil, an intensive program using 2,4-D and diver operated dredges in the 1977-1980 period was effective in restricting spread of this plant. However, because of the overall reduction in available funding for control in 1981 and until 1984, limited control employed bottom barriers and tillage. By 1984, gradual expansion of Eurasian water milfoil populations again warranted a more intensive control program; diver-operated dredging will resume in 1985 in some areas.

Aquatic plant harvesting has been the most important control method in the Okanagan Valley; however, rototilling is expected to displace harvesting in many of the most important public use areas. Although rototilling is slower, the quality of treatment is better and one tillage treatment provides more benefit than three or four harvests. Because tillage can be performed in seasons when plant biomass is reduced (fall, winter or early spring) this reduces time constraints often associated with harvesting (12).

TABLE II. Eurasian Water Milfoil Control Okanagan Valley

Control Method	Area Treated In Hectares*										
	1975	1976	1977	1978	1979	1980	1981	1982	1983	1984	Total
Harvesting	15	0	44	64	116	120	153	122	114	110	858
Rototilling	0	56	17	34	1	0	0	7	16	19	150
Shallow Cultivation	0	0	0	0	0	6	20	18	36	2**	82
Diver Dredge	0	0	8	62	35	35	0	0	0	0	140
Herbicide (2,4-D)	0	1	9	12	20	22	6	0	0	0	70
Total	15	57	78	172	172	183	179	147	166	131	1300

* Area figures may include second or third treatment of sites harvested several times during one growing season; figures are compiled by calendar year.

** Because the cultivator was accidentally damaged and could not be used in Fall, 1984, the area treated is low in 1984; the areas missed will be treated early in 1985.

Shuswap Lake. Table III provides basic information on the scope and methods used to treat Eurasian water milfoil in Shuswap Lake. This plant probably was introduced about 1977 and only was reported (despite surveys about 1980) in 1981. Because of high turbidity levels and a wide natural winter drawdown range, the growth and expansion of Eurasian water milfoil in Shuswap Lake initially was slower than that observed in most Okanagan Valley lakes. However, rapid expansion was documented in the 1983 and 1984 growing seasons despite the fact that Eurasian water milfoil remained localized in only one part of this lake (13). In recognition of the major problem in the nearby Okanagan Valley lakes, public and political support has continued for ongoing, intensive control in Shuswap Lake in 1985. The objectives of this program are:

a) Treatment of all known Eurasian water milfoil colonies, using diver-operated dredges and tillage methods.
b) Prevention of spread to non-infested areas downstream and to nearby lakes.
c) Protection of recreational areas of Shuswap Lake by control of all Eurasian water milfoil populations that threaten to be a nuisance.

Because of turbid water conditions during the early summer months, control programs are handicapped by limited visibility during the early part of the growing season and most control work is performed during August and September. In 1985, approximately $150 000 will be committed to an intensive Shuswap Lake control program; no harvesting is possible because of the sparse populations. If Eurasian water milfoil is found more widely distributed than expected when surveys are performed early this summer, then an intensive program may no longer be cost-effective and a more cosmetic effort will be planned. It is recognized that elimination of Eurasian water milfoil is impossible, but inaction is not acceptable to local residents or tourism concerns. If this plant were allowed to spread unchecked to nearby non-infested areas and permitted to become a nuisance in presently sparsely infested areas, then an even more expensive control program with purely cosmetic objectives might be needed.

TABLE III. Summary of Shuswap Lake Control Program: 1981 to 1984

	1981	1982	1983	1984
Total Infested Area (ha)	5.95	6.27	15.0	28.02
Number of Sites	9	10	16	33
Area Treated (ha)	5.93	6.27	14.26	19.86
Handpicking	-	6.25	3.75	7.60
Bottom barriers	0.06	0.02	0.31	0.03
Diver dredge	5.81	-	-	4.33
Derooting (Cultivation)	-	-	7.20	1.50
(Rototilling)	-	-	1.50	6.50
Program Cost ($)	18 500	27 000	34 000	37 919

Cultus Lake. A third cost-share program has been supported by Water Management Branch since 1978 in Cultus Lake, a heavily utilized recreational lake in the Lower Mainland near Vancouver (1). This program is more limited in scope than Okanagan or Shuswap programs because the infestation is relatively small (about 19 ha) and there is limited potential for expansion within the lake. Despite excellent water quality (including low levels of dissolved nutrients in the water column), Eurasian water milfoil would become a major problem in prime beach areas without ongoing control, mainly using rototilling and bottom barriers. Harvesting is not required on an annual basis. Adequate overall control of 6 to 8 ha of high use area is achieved by treatments of about 4 ha of this area each year in alternation.

Champion Lake. Champion Lake is located in a Provincial Park near Trail in the Kootenay area; a small Eurasian water milfoil colony was found in this 12 ha lake in 1980. At the request of, and in cooperation with the B.C. Parks and Outdoor Recreation Branch, Water Management Branch immediately initiated intensive control to prevent further spread downstream, to prevent a nuisance in this high altitude lake and to minimize risk of spread to other important, non-infested recreational lakes in the Kootenay Region. These objectives were successfully achieved by:

1) Use of a 2,4-D BEE spot treatment of about 0.6 ha (treating the single large colony) in September, 1981.
2) SCUBA surveys in June, 1982, which showed that expansion of the original colony had been arrested and abundance was reduced 50-60%.
3) Application of burlap bottom barrier over the remaining populations in 1982, with small scale follow-up treatments in 1983 and 1984.

SCUBA divers have been able to maintain the remaining populations at low density but annual re-examination and treatment is essential. The Champion Lake experience has demonstrated that considerable benefits may be achieved, at low cost, in circumstances where Eurasian water milfoil is identified at early stages of infestation. However, even in Champion Lake absolute control is not possible and maintenance is required annually.

Eurasian Water Milfoil Quarantine Project and Public Information Efforts

Introduction. By 1974, downstream spread of Eurasian water milfoil through Okanagan Lake had been documented. Recommendations were made at that time to control fragment spread downstream within the Okanagan watershed and to begin public appeals to help reduce spread of viable fragments on boating equipment to other watersheds. Recommendations included: use of signs at boat launches, a public information program, immediate efforts to attempt eradication procedures for new infestations, more effective and intensive control on existing populations and development and testing of mechanical barriers to reduce fragment spread downstream (14). Transport of Eurasian water milfoil to Kalamalka and Wood Lakes (upstream of but near the original infestation) was recorded in the summer of 1975: probably spread was caused by boaters since surveys first recorded the presence of this species in marinas. Unfortunately resources available for effective preventive action were limited, and high priority had to be placed on organizing and monitoring ongoing local control programs, aquatic plant surveys and the testing and development of control technologies during 1975-77. By 1977, Eurasian water milfoil was found in Cultus and Magic Lakes, in the Lower

Mainland. These events and extensive surveys of several hundred British Columbia lakes encouraged planning for significant preventative efforts to protect lakes not then infested with Eurasian water milfoil(15).

Results of Quarantine Projects. In 1978, an experimental boat quarantine project was initiated; more extensive projects were continued in the summers of 1979 and 1980 (16-18) with a final project in 1981 (19). Valuable information was derived from this four year project and some of the objectives and results are summarized here (see Table IV). The 1978 quarantine project was designed to gather information at 50 boat launch ramps on the transport of Eurasian water milfoil fragments by boaters and to inform the public about the hazard of spreading this plant. It was integrated with public information encouraging boaters to remove aquatic plants from boats, trailers and motors. This project revealed that about 24% of boaters leaving infested areas were carrying Eurasian water milfoil (mostly on boat trailers) and that about 2% of boaters checked entering infested lakes were already transporting this plant. Also, the questionnaire data showed that many boaters leaving infested lakes intended to visit nearby, but non-infested, recreational lakes.

The 1979 Quarantine Project was redesigned to check boaters at highway check stations; this approach was continued with minor modifications in 1980 and 1981 (Table IV). Because of the positive public response to the 1978 project, it was decided that voluntary check stations would be more economical and effective in reducing the spread of Eurasian water milfoil, informing the boating public and emphasizing the concern of government agencies about the Eurasian water milfoil problem. Over the 1979-1981 period the percentage of boaters found transporting Eurasian water milfoil fragments increased from 0.5% to 1.7%. This statistic probably reflected the gradual increase of infestation of the mainstem Okanagan Valley lakes during this period. Another discouraging aspect was the increase in boaters transporting fragments who planned to visit non-infested lakes (Table V).

Of the more than 52 000 boaters checked during the three year period, nearly 10 000 boaters (19%) originated from the neighbouring province of Alberta. In 1979 about 57% of Eurasian water milfol fragments were found on Albertan equipment; this dropped to 53% and 55% in 1980 and 1981 respectively. Because of the inordinate volume of fragments transported by boaters from Alberta, this information was provided to appropriate environmental regulation officials in that province early in 1980. Concerns were expressed about the possible spread of Eurasian water milfoil within British Columbia by Alberta boaters and also the possible spread of this plant to Alberta, where its presence was not recorded. These concerns were addressed by a cooperative exchange of technical information between the provinces, leading to posting Eurasian water milfoil warning signs at border crossing points between Alberta and British Columbia in 1980 and the establishment of a major Eurasian water milfoil public awareness program in Alberta in 1981. Boater warning cards now are routinely distributed at National Park and International Border crossing points in British Columbia.

TABLE IV. Eurasian Water Milfoil Quarantine Project (1978-1981)

Description and Number of Boaters Checked	Numbers of Boats and Trailers Found with E.W. Milfoil	Comments
1978 - launch ramps checked; 22 000 inspected during 8 week period	1998 (24%) of 8 257 boats checked leaving infested lakes and 199 (2%) of 10 637 boats checked entering infested lakes	over 16000 questionnaires were completed, providing information on boater movements; 80-90% of boats leaving infested lakes were checked. 90 summer staff were employed Project Cost - $175,000
1979 - 4 highway stations; 18 000 checked in 9 weeks	88 (.5%)	31 summer staff were employed Project Cost - $137 800
1980 - 7 highway stations; 21 942 checked in 10 weeks	197 (.9%)	45 summer staff were employed Project Cost - $215 300
1981 - 3 highway stations; 12 679 checked in 9 weeks	218 (1.7%)	21 summer staff were employed Project Cost - $130 000

Overall Benefits of Quarantine Projects. The quarantine approach evolved during a four year period in which much was learned about boater movements, origins of fragments and volume of Eurasian water milfoil transported within British Columbia. Assessment of overall effectiveness was difficult because it has been impossible to treat all potential sources of fragments or to intercept all boating equipment which might be transporting viable fragments. However, as part of extensive Ministry of Environment efforts to contain the problem and to develop appropriate control measures, the quarantine approach was highly regarded by the public and groups concerned about spread to non-infested areas. The main objective, to educate the public about the Eurasian water milfoil problem, was achieved.

In 1982, quarantine efforts were discontinued, despite strong protests from concerned groups that further efforts were required. During 1981, Eurasian water milfoil was documented for the first time in Shuswap Lake after a member of the public, encouraged by public information requests, reported this new infestation. This lake, which is north of Okanagan Lake, was reported by boaters interviewed in 1981 as their most frequent destination. Also, it was the destination of 34 of 77 boaters found with Eurasian water milfoil fragments headed for non-infested lakes. This expansion of the known infestations (particularly into a large watershed believed previously to be non-infested), reductions in the overall funding for aquatic plant management and the fact that the general public (and especially boaters) were now informed about Eurasian water milfoil, were the main reasons for termination of the quarantine approach.

Concluding Remarks

The history of Eurasian water milfoil management in British Columbia indicates that the biological capabilities of this plant were underestimated during the initial stages of infestation. Also, there were considerable delays in responding appropriately to the problem as plants were observed to establish nuisance populations and demonstrated capabilities for rapid spread. Perhaps this is understandable because of the novelty of this phenomenon in British Columbia. Although some parts of the control programs and testing of technologies appear, in hindsight, to have been ineffective, a great deal of practical experience was accumulated. The ongoing control programs which have evolved from 13 years of extensive management are streamlined, cost-effective, and environmentally safe. Development of local "action committees" to provide political and public support for control efforts and to help priorize allocation of resources has been essential to meet realistic objectives and to satisfy public needs. Also, the importance of public information efforts, to secure cooperation and provide an understanding of the need for management efforts, has been demonstrated. Ongoing control of Eurasian water milfoil and possible future problems with other exotic aquatic plants (such as *Hydrilla*) will be more effective because of these experiences.

TABLE V. Destinations of Boaters Transporting Eurasian Water Milfoil

Destination of Boater	1979 (travel within 2 weeks)	1980 (travel within 1 week)	1981 (travel within 1 week)	Total
Non-infested British Columbia Lakes	37	48	77	162
Non-infested Alberta Lakes	2	6	28	36
Non-infested Saskatchewan Lakes	0	1	2	3
Ocean	9	9	N/A	18
Not Sure/No Lake/ Previously Infested Lake	40	133	111	284
Total	88	197	218	503

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through the Okanagan Valley lakes in the southern interior during the mid-1970's. It was not surprising that the discovery of this noxious weed in Cultus Lake evoked public concern and requests from the local community for government action. Based on previous experience, the Province of British Columbia, Water Management Branch advised that control programs designed and implemented at the local level would have the best chance of receiving support and competing with other funding needs and would be less prone to disruption by potential program opponents.

This paper reviews the process by which local individuals and agencies organized themselves, arranged funding, and currently manage a nuisance lake weed infestation with cooperation from several levels of government. The approach described may have broader application to other community level aquatic plant management situations.

The Participants

During 1978, Water Management Branch staff were invited to a series of meetings by several local agencies to discuss the status of Eurasian water milfoil growth in Cultus Lake and to comment on possible control alternatives. Concurrently, there was strong public support for efforts to eradicate or at least control this nuisance plant; this sentiment did much to promote needed local political interest. As a result, the Cultus Lake Milfoil Action Committee (C.L.M.A.C.) was formed in 1978, by representative members of all agencies and groups which held shoreline jurisdiction or had a recognized vested interest in the resources of Cultus Lake (see Table 1.)

TABLE I. Representation on the Cultus Lake Milfoil Action Committee

Agency/Group	Proportion of Shoreline Jurisdiction or Other Related Concerns	
Province of British Columbia - Parks Branch	17.8 km	(74%)
Cultus Lake Park Board	4.5 km	(19%)
Lindell Beach Residents Assoc.	0.8 km	(3%)
Dept. of National Defence	0.7 km	(3%)
Lakeside Marina	0.2 km	(1%)
	24.0 km	100%
District of Chilliwack (13 km north of Cultus Lake)	Area Tourism	
Regional District of Fraser-Cheam (Regional governing body)	Area Tourism	
International Pacific Salmon Fisheries Commission (also represents Dept. of Fisheries and Oceans)	Management of sockeye salmon shore spawning grounds	

The Committee takes responsibility for program design, arranging finances, and implementing controls.

Program Design

Long-term management of aquatic weeds should be a systematic process responsive to changes in control objectives, treatment alternatives, and fluctuating availability of funds. These principal elements in the C.L.M.A.C.'s program design are discussed below.

Decision to Treat. It is the policy of the C.L.M.A.C. that the decision to initiate or proceed with control efforts be subject to annual review. Typically, this review is based on a year-end evaluation of preceeding program monitoring results. A lake-wide survey of the distribution and density of aquatic plants at the end of the growth season also has been useful in determining levels of treatment success and defining management needs for the following year. The Committee has elected to conduct annual treatments since 1978.

Define Program Objectives. Control objectives are formulated well in advance of the next growth season. The C.L.M.A.C. determines the size and location of each intended treatment site (members from each jurisdictional body at the lake are responsible for defining water uses and control needs for their own area). Planned management efforts are generally directed at (in order of decreasing priority): swim enclosures, open-water swimming and wading areas, public boating and moorage sites, and inshore salmon spawning grounds. The desired level of control also may be specified (e.g.: anticipated duration of relief from nuisance growth). However, as pointed out by Mitchell(1), care must be taken not to specify requirements which may eliminate potentially useful control alternatives. Other objectives may involve testing new control techniques. Planners are required to maintain cost-effectiveness in a field where the state of the art is undergoing continual refinement.

Develop a Detailed Treatment Plan. Once objectives are established, appropriate control methods can be selected. Details which are taken into consideration include: availability of control equipment, area and depth of target plant populations (from field surveys and air photographs), handling of spoils, substrate conditions (size of bed materials, presence of debris, etc.), and general environmental sensitivity with respect to water uses and foreshore activity. In an advisory capacity, Water Management Branch staff provide information on control equipment capabilities (all control equipment is presently owned by the Province) and make recommendations based on program objectives. The C.L.M.A.C. makes the final choice of options available (e.g. a Water Management Branch recommendation for limited use of the herbicide 2,4-D has never been approved). Typically, the best approach is an integrated program using several different methodologies appropriate to site-specific factors. Staff requirements and program duration can then be determined given the established treatment rates of the techniques to be used. A time allowance is also made for necessary monitoring activities. Proper program scheduling is critical if a particular treatment method is effective only during a specific stage of plant development. Other considerations which must be made in the Cultus Lake situation include scheduling treatments to avoid peak public use periods (weekends, special events), and any form of substrate disturbance is prohibited between

September 30 and July 1 when sockeye salmon eggs or immmature alevins may be in the gravels.

Arrange Funding to Meet Objectives. Fixed effectiveness as opposed to fixed cost is probably the most desirable approach for most aquatic weed problems because it is generally easier to be more precise about effectiveness than environmental management costs(1). Therefore, program costs are estimated once a detailed plan of activities has been prepared.

A compilation of projected expenditures by the C.L.M.A.C. for a two or three month program would normally include: personnel (machine operator(s), divers), spoils disposal if necessary, general operating expenses, administration, and rental or purchase of control-related equipment. The C.L.M.A.C. has thus far been able to obtain most mechanical control equipment on loan from the Province.

The Province also has made provision for funding assistance to local governments for the implementation of Eurasian water milfoil control on a shared cost basis, 75 percent payable by the Province and 25 percent payable by the local authorities. All requests for funding assistance are priorized; a local organization must demonstrate sufficient need to reduce the impact of Eurasian water milfoil on recreational use and the local economy. Proposed program designs also must be given approval before qualifying for assistance, to ensure cost-effectiveness in use of public funds. The C.L.M.A.C. has negotiated annual cost-share agreements with the Province since 1978.

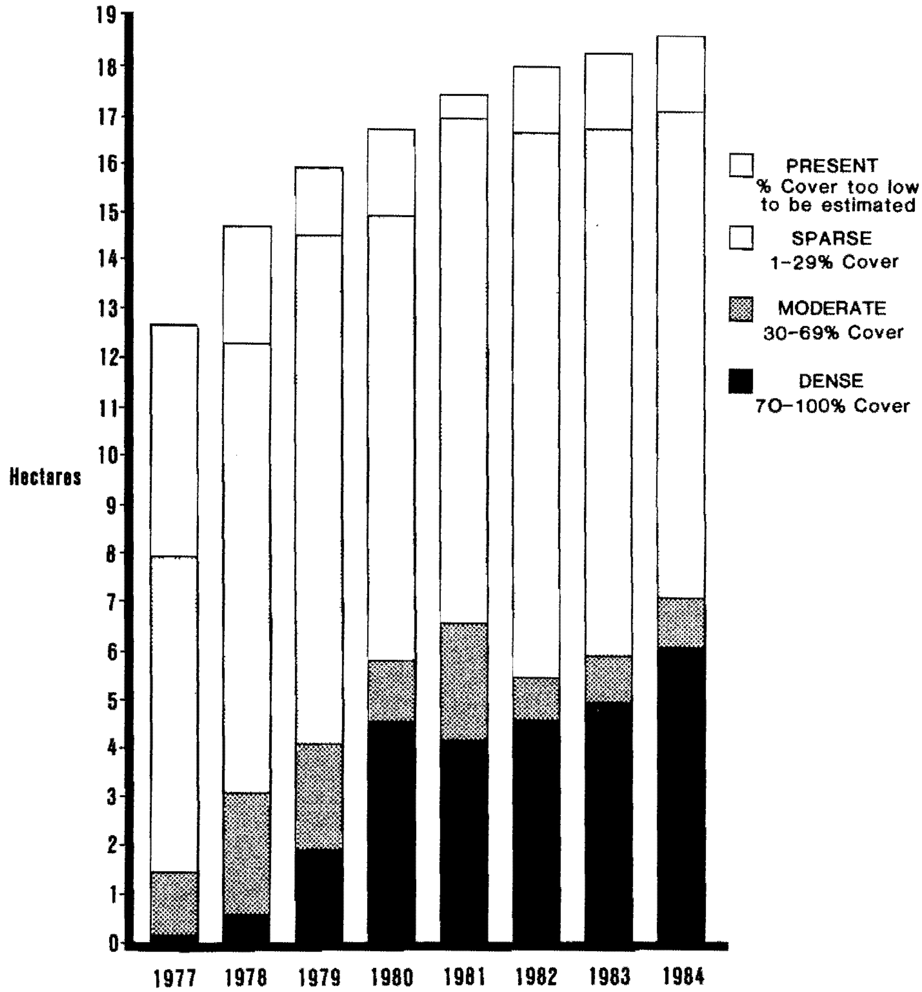
Local funding necessary to raise 25 percent of program costs is arranged by the C.L.M.A.C. The Committee's approach is to apportion funding responsibilities to its member agencies according to their length of shoreline jurisdiction. Coincidentally, Cultus Lake Provincial Park occupies 74 percent of the lake's perimeter or equivalent to the Province's cost-share contribution; therefore, the remaining shoreline jurisdictions listed in Table I (percentages) approximate the remaining fractions of the total program cost borne by the other agencies. Contributions by the District of Chilliwack and the Regional District of Fraser-Cheam are arbitrary amounts and are subject to availability of funds. Local groups may choose to provide services of equal value in place of direct funding (or any combination of cash and services) to the cost-sharing formula, where these services would otherwise incur cost to the program (e.g. administration, storage facilities, moorage). The Department of National Defense provides a support boat, heavy equipment transport, and crane services rather than funding.

The C.L.M.A.C. also has an agreement with the Department of Fisheries and Oceans, exclusive of the Provincial cost-sharing process, in which the Department provides $1 000 annually for Eurasian water milfoil removal directed specifically at rehabilitating sockeye spawning gravels along a 1 km section of shoreline.

Management Techniques: Control of Eurasian Water Milfoil in Cultus Lake

Eurasian water milfoil control in Cultus Lake has undergone several changes, primarily in response to the increase in density and distribution of this nuisance aquatic plant (Figure 2) and the development of more efficient control methods. A summary of annual program activities is

FIGURE 2. Area and Density of Eurasian Water Milfoil in Cultus Lake, 1977-1984.



presented in Table II and the following paragraphs.

TABLE II. Cultus Lake Operational Summary
Eurasian Water Milfoil Control, 1978-1984(2)

	1978	1979	1980	1981	1982	1983	1984
Total Affected Area	14.7ha	15.9ha	16.7ha	17.4ha	18.0ha	18.3ha	18.7ha
Area Treated	0.7	1.34	1.56	4.52	3.09	4.55	3.84
Method							
Rotavator				3.55	2.99	2.88	3.77
Harvester						1.4	
Diver Dredge	0.7	1.34	1.56	0.72		0.08	
Bottom Barriers				0.25	0.1	0.19	0.07
Total Program Cost	$16000	$58000	$61700	$48000	$21000	$17000	$17000

1978. The limited distribution of dense Eurasian water milfoil growth in Cultus Lake prompted local authorities to attempt intensive control beginning in late 1978, using a Province-owned, diver-operated suction dredge. Although eradication was considered to be an unrealistic goal, it was hoped that diver dredging would reduce the overall level of infestation to a point where only routine spot-treatments would be necessary in future.

1979. A major diver dredging program was initiated. However, attempts to provide lake-wide control by this method were abandoned as the spread of Eurasian water milfoil exceeded the operating limits of this machine (treatment rate of 100-300 m^2/day).

1980. Eurasian water milfoil growth continued to expand; the proportion of dense nuisance growth (>70 percent bottom cover) increased two and one half times that of the previous year. Diver dredging was still considered to be a suitable method for treatment of limited, high-use public areas.

1981. As the need for weed removal increased, the slow progress and high cost of diver dredging encouraged evaluation of other alternatives. Harvesting was considered to be of limited use due to the inaccessibility of many sites (enclosed wharves, docks, swimming impoundments) to large machines and the probable need for repetitive treatments, perhaps even in the same growing season. A proposal for selective use of the herbicide 2,4-D did not receive full approval of the C.L.M.A.C.

Three different open-mesh materials (Aquascreen, window screen, and burlap) were tested for use as bottom barriers. The light limitation and physical compression qualities of these materials provided beneficial results.

Bottom tillage (rotavating) previously had been considered impractical for use in the type of bottom terrain (log debris, rocky substrates) in Cultus Lake, although tests elsewhere using the Provincial rotavator (prototype) were successful(3). Trials conducted in Cultus Lake verified the problems with difficult substrates; however, greater than 90 percent plant removal (roots and stems) was possible at a depth range of 0.5 m to 4 m, where sufficient penetration of the rotavator tines could be achieved.

All management areas which contained nuisance weed growth were treated (4.52 ha).

1982. More efficient use of the rotavator was made possible by removal of obstacles (sunken logs) in selected management sites prior to treatments. Diver dredging was discontinued as an annual treatment as sites with immovable bottom or surface obstacles could now be intensively controlled with bottom barriers. As a result, program cost was reduced to less than half that incurred in 1981.

1983. The spread of dense Eurasian water milfoil into valuable sockeye salmon spawning grounds along the southern shoreline of Cultus Lake and a resultant avoidance response by spawners was confirmed by the International Pacific Salmon Fisheries Commission. The Federal Department of Fisheries and Oceans provided funding for rehabilitating the site by bottom tillage; rotavating was confined to mid-summer when all salmon fry had left the grounds.

A harvesting trial was largely unsuccessful due to the difficulty of treating shallow, sloping beds of Eurasian water milfoil found along much of the shoreline.

Testing of new bottom barrier materials was continued. Two private companies supplied prototype versions of their products (Tac 210, a polyester geotextile from Texel, and Dartek, a perforated nylon from Du Pont Canada) for trial use in the Cultus Lake program. Eventual production versions of both product lines incorporated design improvements recommended as a result of these trials.

The invasion of Eurasian water milfoil into non-infested areas and expansion of existing populations was minimal but nevertheless constant. However, management activities were successful at removal of all nuisance growth in the important recreational areas around Cultus Lake.

1984. Rotavating remained the most effective control method for the large majority of management sites and provided two years of relief from nuisance weed growth in many areas. Approximately two-thirds of the 6 ha management area can be treated in rotation every two years, thereby reducing the duration of annual programs. Bottom barriers continued to provide cost-effective, extended control (two to four years) wherever the rotavator could not gain access or was limited by impenetrable substrates.

Conclusion

Cost-effective, ongoing control programs which are politically and publicly supported remain a high priority because of the high recreational value of the shoreline areas of Cultus Lake. During the past seven years, practical control methods have been developed and tested and should continue to benefit lake users for some years to come.

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BOTTOM TILLAGE TREATMENTS FOR EURASIAN WATER MILFOIL CONTROL

M.D. MAXNUK

Biologist, Littoral Resources Unit
Resource Quality Section
Water Management Branch
Ministry of Environment
#3 - 4320 29th St.
Vernon, British Columbia
Canada V1T 5B7

Mechanical methods available for aquatic plant control have been largely limited to two technological types: dredging-related and harvesting/cutting related. In an attempt to improve on existing mechanical methods, the British Columbia Ministry of Environment has developed a unique technology which uses common agricultural tillage equipment to uproot aquatic plants. This paper discusses the methods, costs and effectiveness of the two derooting methods being used (rototilling and cultivating) for management of Eurasian water milfoil (Myriophyllum spicatum L.) in British Columbia, Canada.

Eurasian water milfoil (Myriophyllum spicatum L.) is a noxious aquatic weed which has become well established throughout the main lake system of the Okanagan Valley in British Columbia, Canada. In 1971, unusually dense growths of this plant were first noticed growing in the northern end of Okanagan Lake - the largest of six lakes in the system. Since that time, local agencies have made a serious attempt to control the problem by implementing a multi-disciplinary research and control program (1)

An important objective of research into possible control methods was to locate or develop an approach which could provide more effective and longer lasting control than existing control technologies (such as harvesting) and without the controversy or potential adverse environmental impact of herbicides. With this aim investigations began into methods for controlling plants by removing their roots from the substrate. In experimental trials conducted over a period of several years, two agricultural methods - rototilling and cultivating - were found to be particularly effective at dislodging water milfoil roots. Several years of operation have shown these methods to produce higher quality, longer lasting control than achieved by harvesting, at a reasonable cost.

This report describes the use of rototilling and cultivating (collectively termed bottom tillage treatments) for Eurasian water milfoil control in the Okanagan Valley, from their inclusion in the operational control program in 1980 to the present. Included are descriptions of equipment used, methods of operation, cost and rate of operation, and effectiveness.

Equipment and Operation

Rototiller: The rototiller used in the Okanagan control program is a highly modified version of agricultural rototillers used for terrestrial vegetation control. The aquatic version uses a series of L-shaped blades fixed to a shaft revolving at 150 to 250 rpm. These blades penetrate 10 to 15 cm into the substrate, slicing under water milfoil root crowns, dislodging them from their substrate. As the root crowns are caught in the rotating action of the blades, soil particles are washed off and the buoyant root crown and stem portions of the plant float to the surface and eventually to shore.

The rototiller head is approximately 2 m long and is supported by two 3.5 m long arms which are mounted on each side of a paddlewheel-driven barge. These long support arms pivot just behind the centre of the barge and are raised and lowered on a cable by a winch mounted on the barge deck. The length of the support arms permit effective operation in water depths ranging from 0.5 m to 3.5 m. Diesel engine-powered hydraulic pumps are mounted on the barge to activate the paddlewheels, winch, and rototiller head. High speed rotation of the rototiller head is accomplished with two hydraulic motors mounted on either end of the shaft.

The machine is operated in a parallel overlapping swath pattern. Two complete sets of passes are made, one parallel and the other perpendicular to shore. During operation, turbidity in the treatment area prevents the operator from seeing the lake bottom to ensure overlap or avoid underwater obstacles. Machine guidance is attempted by working within small defined blocks and fixing visual bearings to an object on shore. However, in practice the operator usually continues making overlapping parallel swaths within a discrete block until no more plant material floats to the surface. Guidance is further complicated by the difficulty in tracking, or controlling the direction and speed of the barge while the rototiller head is turning. The blades of the rototiller head tend to push the barge away from its intended course according to the contour and nature of the substrate. Without a positive guidance system, many more passes than the theoretical number (swath width multipled by width of treatment area) are required to achieve total coverage.

<u>Cultivator</u>: Cultivating is accomplished by towing an agricultural spring-shank cultivator along the lake bottom. The cultivator consists of three rows of six shanks (each with a 15 cm wide shovel or sweep) mounted on a frame between two wheels. The shanks are staggered in the rows to ensure complete overlap within the operating width. This implement is mounted by a three-point hydraulic hitch on a Bombardier "Jimmy" skidder muskeg tractor. The tractor has been modified to operate in water depths up to 1.25 m by the addition of waterproof steel skirts. The cultivator shovels are set to penetrate 10 to 15 cm into the substrate. As they slice below the root crown of the plant, they uproot and cause sufficient agitation to shake off the soil particles, allowing the root crown to float.

The machine is operated in a parallel, overlapping swath pattern. Two complete sets of passes are made, one parallel and the other perpendicular to shore. Guidance problems encountered with the floating rototiller are not as serious with the land-based cultivator because the operator has better tracking control over the direction and forward speed of the machine. As with rototilling, turbid water created during the operation does not permit the operator to see the lake bottom to overlap swaths. Although marker stakes are placed along the treatment boundary to provide a visual reference point for guidance, operation is usually continued in a discrete block or area until little or no plant material floats to the surface in the wake of the machine.

Root tillage treatments are conducted in the fall, winter, and early spring in British Columbia. During these periods massive amounts of shoot growth are not present to clog the implements and reduce operating efficiency. Viable plant material fragmented by treatments at this time of year is reduced in volume and less likely to have the potential to develop new colonies. Material which floats to shore may be frozen during the winter months and spoils disposal costs are minimal. Treatments in these periods also avoid interference with beach use during the busy summer tourist season. Late season tillage also takes advantage of lowered lake water levels, allowing both pieces of equipment to clear areas further away from shore than would be possible during normal summer water levels.

Operating Rates and Costs

This report considers bottom tillage operations conducted from April 1980 to April 1985. Areas treated with the floating rototiller are listed in Table I while areas treated with the land-based cultivator are listed in Table II. Over a three year period a total of 56.8 ha was rototilled in seven separate locations, with individual treatment sizes ranging from 1.3 to 8 ha. Over a five year period a total of 155.5 ha was cultivated in four separate locations with individual treatment sizes ranging from 2 to 21 ha. All of the areas treated with both methods receive a high degree of use by the public during the summer months, and include public beaches, boat launches and commercial lakeshore facilities.

Operating rates for bottom tillage treatments are expressed as hectares treated per seven hour day (ha/d). The number of operating days recorded for each treatment is, in most cases, the actual number of days that the machine spent on the job site as recorded by the machine operators in their daily log books. These figures include time spent in productive operation, and time spent not operating because of mechanical breakdown, preventive maintenance, machine set-up before and after moving, and paid work breaks. Theoretical operating rates based on timed trials of forward operating speed, average turn-around times, swath width, and overlap were also calculated. However, operating rates based on machine logs will be more useful to those monitoring these technologies because they reflect a more realistic picture of conditions encountered in an operational program.

Operating rates for rototilling operations shown in Table I range from a minimum of 0.12 ha/d to a maximum of 0.47 ha/d. Compared to the average operating rate of 0.26 ha/d the first two treatment rates of 0.47 and 0.38 ha/d are unusually high. The size of the areas treated in these locations may have been overestimated. Operating rates for cultivating shown in Table II range from a minimum of 0.40 ha/d to a maximum of 1.67 ha/d with an average rate of 0.81 ha/d. With a few exceptions, operating rates in areas treated for the first time with either technology are consistently lower than in subsequent treatments. This is due to the higher number of plants per unit area and is related to the need to make repeated passes because of the lack of positive guidance systems. The average operating rate for cultivating is nearly four times faster than rototilling. Higher operating speed, fewer guidance problems, and larger treatment areas suitable for

Table I. Bottom Tillage Treatments Using a Floating Rototiller to Control Eurasian Water Milfoil in the Okanagan Valley, 1982-1985

Date Treated	Location	Area (Hectares)	Cost	Cost per Hectare	Operating Days	Hectares per Day
Mar. 11-31/82	Kalamalka Beach	7.0	$3 140	$ 449	15	0.47
Feb. 25- Mar. 25/83	Kalamalka Beach	8.0	4 274	534	21	0.38
Apr. 25- May 24/83	Willow Beach	3.0	3 933	1 311	22	0.14
Nov. 2 - Dec. 1/83	Kinsman Beach	5.1			21	0.24
Feb. 8 - Mar. 15/84	Kalamalka Beach	6.5	21 810	1 253	27	0.24
Mar. 16 - April 11/84	Willow Beach	4.5			19	0.24
April 11-27/84	Rec. West	1.3			11	0.12
Oct. 3 - Nov. 8/84	Kinsman Beach	6.3			22	0.29
Jan. 29 - April 1/85	Kalamalka Beach	9.0			32	0.28
April 2-11/85	Oyama Canal	1.3	27 318	1 307	6	0.21
April 12 - May 6/85	Wood South	2.7			25	0.11
May 9-16/85	Rec. West	1.6			8	0.20

Table II. Bottom Tillage Treatments Using an Agricultural Cultivator to Control Eurasian Water Milfoil in the Okanagan Valley, 1980-1985

Date Treated	Location	Area (Hectares)	Cost	Cost per Hectare	Operating Days	Hectares per Day
April 8 - May 6/80	Motel Row	5.6	$2 585	$462	5	1.12
Feb. 25 - April 10/81	Motel Row Kelowna Foreshore Kinsman Beach	11.4 5.5 4.0	7 757	371	33	0.63
Nov. 3 - Dec. 16/81	Kelowna Foreshore	21.0	3 674	175	26	0.81
Mar. 11-26/82	Motel Row	12.0	1 779	148	12	1.00
Apr. 5-21/82	Kinsman Beach	6.0	1 698	283	11	0.55
Nov. 8 - Dec. 13/82	Kelowna Foreshore	21.0	3 341	159	25	0.84
Feb. 7-16/83	Motel Row	12.0	1 514	126	8	1.50
Feb. 23 - Mar. 3/83	Kinsman Beach	6.0	1 771	295	7	0.86
Nov. 4-25/83	Kelowna Foreshore	18.0	5 590	275	15	1.20
Feb. 13 - Mar. 2/84	Kalamalka Beach	2.0			5	0.40
Feb. 25 - April 19/85	Kelowna Foreshore	21.0	12 925	417	38	0.55
April 23-30/85	Motel Row	10.0			6	1.67

this method are the main factors which account for this difference. The theoretical operating rate for rototilling is 2.3 hectares per day, nearly ten times higher than the average actual rate of 0.27 hectares per day, while the theoretical rate for cultivating is 4.2 ha/d, four times the actual rate of 0.81 ha/d. The difference in theoretical from actual operating rates indicates the large amounts of "non-productive" time necessarily associated with most routine machine operations, and particularly the need to make multiple passes to ensure complete coverage with these methods.

Total and unit costs for rototilling and cultivating treatments carried out during the five year period under consideration are shown in Tables I and II. More detailed breakdowns of bottom tillage costs for the two most recent years are provided in Tables III and IV. All costs shown include direct operating expenditures only. Major equipment purchases, depreciation, administration, monitoring and documentation are excluded.

Unit costs for rototilling range from a minimum of $449 per hectare to a maximum of $1 311 per hectare, with an average cost of $1 074 per hectare. The costs of 1982 and 1983 rototilliilng treatments are much lower than the average because they included only the amount of money spent while the machine was on site. Major repairs amd refurbishing in the off-season were not included in the expenditure totals for those two early years, but these costs were included for the past two seasons.

Table III. Cost* of Bottom Tillage Treatments Carried Out Between November 2, 1983 and April 27, 1984

Operator's Wages	
Rototilling	$ 5 400
Cultivating	2 500
Equipment Transport	2 900
Pick-up Rental and Operation	3 500
Fuel and Lubricants	1 600
Maintenance, Repairs**,Supplies	11 500
Total	$27 400

*These include direct operating costs only. Capital costs, supervision, and administration are excluded.
**Includes two major breakdowns: Bombardier differential and final drive rebuild ($4 500) and rototiller hydraulics ($2 500).

Table IV. Cost* of Bottom Tillage Treatments Carried Out Between October 3, 1984 and May 16, 1985

Operator's Wages	
Rototilling	$11 744
Cultivating	5 556
Equipment Transport	3 260
Pick-up Rental and Operation	6 135
Fuel and Lubricants	4 000
Maintenance, Repairs, Supplies	9 548
Total	$40 243

*These include direct operating costs only. Capital costs, supervision, and administration are excluded.

Unit costs for cultivating range from a minimum of $126.00 per hectare to a maximum of $462.00 per hectare, with an average cost of $274.00 per hectare. For the same reasons as for rototilling, cultivating costs shown in Tables III and IV should be considered a more accurate estimation of expenditures required for this kind of treatment.

Effectiveness

The effectiveness of bottom tillage treatments was determined by comparing changes in either stem or whole plant density before and after operation. SCUBA divers counted and recorded the number of Eurasian water milfoil stems or plants occurring in 0.25 m^2 quadrats or 1.0 m^2 quadrats thrown randomly throughout a treatment site. The approximate number of quadrats sampled per treatment site ranged from 25 to 50. Fewer quadrats were sampled (n= 6 to 10) when pre-treatment conditions were observed to be uniformly dense. Only firmly rooted plants or stems attached to roots firmly embedded in the bottom were counted. Stem fragments lying on the bottom or starting to root into the substrate were not included. All post-treatment sampling was done well into the first growing season after treatment so that all remaining root material had a chance to produce new shoots and before recolonizing fragments from outside the treatment area could develop into established plants. These observations confirmed the presence or absence of roots remaining after treatment.

Because this was an on-going treatment program, it was not possible to perform detailed quantitative evaluations of each site listed in Tables I and II. Also, as most sites were being treated on an annual basis it was virtually impossible to determine the effectiveness of a single treatment beyond one growing season. In many locations only post-treatment assessments could be performed. For these sites, quadrats were thrown randomly into nearby untreated areas to estimate pre-treatment density. Visual observations of effectiveness also were made for most treatments.

A detailed study of 1977 and 1978 experimental rototilling trials described immediate and long-term effectiveness, as well as effectiveness in different substrate types (2).

Changes in plant density following rototilling and cultivating treatments are shown in Tables V and VI respectively. Single treatments consisting of multiple overlapping passes parallel and perpendicular to shore reduce uniformly dense stands of Eurasian water milfoil by 80 to 97% for rototilling and 49 to 98% for cultivating. Most of the remaining plant material observed consisted of small clumps or strips completely missed due to guidance problems or because of proximity to obstacles. Large portions of treated areas were typically devoid of Eurasian water milfoil. Areas where lower quality treatment was observed frequently were found to have hard-packed substrate which resisted the penetration of the blades or shovels, or extremely soft silty substrates where the root clumps were partially reburied. Where visual observations were made, tilled areas

Table V. Reductions in Eurasian Water Milfoil Stem Density Immediately Following Rototilling

Treatment Location and Date	Average Stem Density Before Treatment	Average Stem Density After Treatment	Percent Reduction
Kalamalka Beach* Feb.-April/77	40/0.25 m^2**	4/0.25 m^2	90
Skaha Public Beach Aug.-Sept./77	52/0.25 m^2	11/0.25 m^2	80
Willow Beach (A)*** Apr.-May/83	172/0.25 m^2**	6/0.25 m^2	96.5
Kinsman Beach Nov.-Dec./83	74/0.25 m^2**	10/0.25 m^2	86.5
Willow Beach (B) March-April/84	54/0.25 m^2	2/0.25 m^2	96
Rec. West Marina April/84	61/0.25 m^2	2/0.25 m^2	97
Wood Lake South April-May/85	62/0.25 m^2	5/0.25 m^2	92

*After one full growing season the average stem density increased to 12/0.25 m^2 while the percent reduction increased to 70%.

**Pre-treatment density was estimated by sampling an adjacent untreated area.

***After one full growing season the average stem density increased to 9/0.25 m^2 while the percent reduction increased to 95% .

Table VI. Reductions in Eurasian Water Milfoil Stem Density Immediately Following Cultivating

Treatment Location and Date	Average Density Before Treatment	Average Density After Treatment	Percent Reduction
Motel Row Experimental Site. Mar./80	190 plants/m^2	14 plants/m^2	92.5%
Kelowna Foreshore Nov.-Dec/81	29 plants/0.25 m^2	15 plants/0.25 m^2	49%
Motel Row Mar./82	190 plants/m^2*	7 plants/m^2	96%
Kelowna Foreshore Nov./83	109 stems/0.25 m^2*	2 stems/0.25 m^2	98%

*Pre-treatment density was estimated by sampling adjacent untreated area.

typically were described as very sparse to moderate plant cover while adjacent untreated areas were mostly uniformly dense.

Because these methods remove roots, treated areas remain free of Eurasian water milfoil throughout the first growing season following treatment. Although reinfestation from nearby untreated areas and regrowth of remnant root material begins almost immediately, plant density remains sufficiently reduced to permit normal water-based recreational activities throughout a second growing season following treatment.

Conclusions

Eurasian water milfoil problems in British Columbia, and particularly in the Okanagan Valley, are unique when compared to the rest of North America. Most of British Columbia lakes now infested with this plant are large, deep, mesotrophic to oligotrophic lakes with a relatively narrow littoral shelf. Much of the surface areas of these lakes is well beyond the depth of rooted aquatic plants. However, the shallow littoral shelves provide ideal habitat for rooted aquatic plants; coincidentally, these areas are used and appreciated most by the public. Harvesting the narrow littoral shelves in areas of high public use is a largely unsatisfactory method of controlling nuisance aquatic plant growth. Not only must this work be done when plants are near the surface and probably already a nuisance to beach users, but harvested areas often grow back to nuisance proportions within the same growing season. Harvesting also generates considerable cut material that is costly to dispose of, produces floating fragments, leaves a stubble of cut stems on the bottom and uncut plants in less than 1 m water depth.

After five years of operational experience, which followed two years of experimentation, bottom tillage treatments have proven to be a viable alternative to harvesting. Properly conducted tillage operations do not leave a stubble of uncut plants, are effective virtually up to the water's edge, and control Eurasian water milfoil growth for at least one entire growing season. Tillage treatments also can be conducted in the colder periods of the year, before the tourist season starts.

Bottom tillage will never entirely replace harvesting as a control method. The much faster operating rate for harvesting, ranging from about 1 to 3 ha/day, will be required when treating large infested areas for relatively rapid, inexpensive vegetation removal. Where higher quality and longer lasting treatments are needed, bottom tillage is becoming more widely used and favored for aquatic plant control.

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LABORATORY INVESTIGATIONS ON ULTRASONIC CONTROL OF EURASIAN WATER MILFOIL

ROGER J. SOAR

Lambda Technology
1150 Woodstock Avenue
Victoria, British Columbia
Canada V8V 2R1

The results presented in this report demonstrate that high power ultrasound and associated cavitational streaming have the ability to disrupt the structure of Eurasian water milfoil (*Myriophyllum spicatum* L.). Biological effects of ultrasound on plant cells, documented in laboratory tests, include; cell death, damage or destruction of cellular structures, changes in osmotic potential and chemical changes within the plant. The exposure time required to facilitate these damages range from instantaneous to several seconds. Modern advances in high power electronics and magnetostrictive transducers promise economical applications of ultrasound as a feasible control technology. As an aquatic plant management tool, ultrasound would have numerous advantages over present technologies particularly in reducing adverse environmental impacts.

During the last few years theoretical and practical implications of using high intensity ultrasound as a method of aquatic plant control have been investigated. Experimental work has concentrated on Eurasian water milfoil (*Myriophyllum spicatum* L.), a nuisance plant found in abundance in southern British Columbia. Due to the proprietory nature of the ongoing research, this paper is a brief review on the observed effects of ultrasound on Eurasian water milfoil. Exact equipment description remains confidential; however, experimental details are given where possible.

None of the techniques now utilized for aquatic plant control can economically provide 100 percent weed removal; some may not be publicly or environmentally acceptable (herbicides), some may be hazardous to the operator (diver dredging) and some are only cosmetic (harvesting). At present there is no adequate tool available for comprehensive aquatic plant management. Ultrasound as an aquatic plant management tool would have numerous advantages over present technologies and could resolve long-standing environmental issues with the public. Advantages of the successful application of this technology would include:

- aquatic plants could be controlled before they develop into a large nuisance biomass;
- dormant or overwintering plants could be treated, yielding high root crown mortality;
- treated weeds would decay *in situ* so that waste disposal would be unnecessary;
- the method has immediate on/off capabilities with nearly instantaneous effects;

- treatment areas are defined, and possible adverse effects to non-target aquatic life would be localized;
- no foreign materials would be introduced during or be produced after treatment;
- treatments would result in no adverse effects on drinking or irrigation water;
- ultrasound components have a superior life expectancy as there are no moving parts;
- purchase cost of an ultrasound array would be the same as a large harvester; maintenance costs would be reduced.

Conceivably this technology could be developed in modules adaptable to large or small scale applications and attached to existing support platforms on water or land.

High Intensity Ultrasound and Cavitation

Ultrasound is a high frequency (10 kHz-200 MHz) sound wave that can travel through any molecular medium. The sound wave is usually generated by an electro-mechanical device, a transducer, which produces a mechanical vibration within the medium causing the medium particles to oscillate. The oscillations of particles subjected to a sound wave produce regions of high and low pressure at any given point, as the wave passes it. Parameters that affect the transmission of ultrasound through a liquid medium include the frequency and intensity of the ultrasound and the temperature, density and quantity of dissolved gases in the medium.

Under certain conditions within a medium, ultrasound with a sufficiently high amplitude can produce a phenomenon known as cavitation. Cavitation can be observed in two forms (1)

A. Stable Cavitation - or non-collapsing bubble formation is important at low ultrasound intensities. It can produce microstreaming in plant cells that can disrupt the internal organization of the cell (2).
B. Collapse Cavitation - a more violent phenomenon that results in erosion of cell walls and mass disruption of both plant and animal cells.

Effects of Ultrasound and Cavitation on Plants

Common biological effects of intense ultrasound on plant cells are chromosonal anomalies, cell death, damage to or destruction of cellular structures, reduced growth rates and mitotic indices. These effects have been reported after moderate exposures of 1-10 w/cm^2 for several minutes using medically relevant frequencies of 0.8-2.3 MHz (3)

Effects documented during previous studies by the author included the characterization of progressive damage to Eurasian water milfoil leaf and stem epidermal cells; the duration of exposure over which this occurred varied from instantaneous to several seconds. The progressive stages include chloroplast deformation, chloroplast homogenization to cell rupture.

Other effects produced by ultrasound and cavitation are changes in the osmotic potential of cells and chemical reactions within the liquid being cavitated. It has been documented that the permeability of cellular material in plants and animals is changed both during and after treatment with ultrasound. Chemical changes that occur as a result of ultrasound have also been widely documented (4,5,6). For example, hydrogen peroxide is reported to be formed from water during collapse cavitation by dissociation of water into atomic hydrogen and the hydroxyl radical.

Experimental Methods

Apparatus. The experimental system consisted of a B+K 3010 function generator with a useable frequency range of 0.1Hz to 1MHz; a laboratory built tone burst generator capable of producing pulsed signals with durations ranging from 0.01 to 20.0 seconds and a variable duty cycle from 5-100%. The signals were amplified by an ENI 1140L broadband (9-250kHz) amplifier connected to an impedance matching transformer. A 12 amp variable D.C. volt power supply provided a biasing current to the transducer and a choke-capacitor filter network was constructed to provide the integrated AC-DC power to the transducers. Exposure period was controlled by a one shot electronic timer.

Beam patterns, intensity distribution and signal monitoring were measured using an Atlantic Research BC-10 hydrophone and direct probe connections to a Tektronics 2015 oscilloscope. Total energy flux (7) was measured using an Ohaus chemical micro balance.

A 230 L tank with sound absorbing foam was used for transducer output measurement and another tank of 2 300 L capacity was used for plant testing. A third tank (200 L capacity) was constructed so that it could be pressurized to simulate transducer operating conditions to a depth of 8 m. The transducer's cavitation stream was monitored by visual observation through a window in the tank or by hydrophone. The tank facilitated detailed examination of the transducer's performance at all operational depths.

Transducer Evaluation. When considering the biological action and comparing different ultrasound beams it is preferable to know the physical parameters of the beam. The general approach in most research has been to combine measurements of total energy flux through the cross section of the beam, using an absolute device such as a radiation balance, and making relative measurements within the cross-section of the beam using a hydrophone device.

Measurements of the transducers were performed in the 230 L barrel; hydrophone measurements were taken at points on a grid covering the transducer face at 1 cm intervals, absolute radiation measurements were taken at intervals of 5 cm from the transducer face. Measurements were converted to atmospheres of pressure to express the transducer output.

Plant Growth Conditions. Eurasian water milfoil apical shoots in prime condition were collected from Cultus Lake during summer and winter growth periods. The shoots were placed in aquaria where they were kept prior to and after treatment. Aquarium water was bubbled with air and heated to maintain temperatures between 20-23°C; a 15.5 hour photo period was provided by eight Sylvania "gro-lux" fluorescent tubes.

Sonnication of Eurasian water milfoil. Apical fragments were randomly selected from the aquaria and placed in the required exposure and control groups. All fragments were cut 10-12 cm in length and examined for infections or damage before being used for experiments. The number of internodes per fragment was documented and the fragment tagged for future identification. The base internodes (not included in the treatment evaluation) of each fragment were inserted into a rubber tube clamped on a sound absorbing platform 15 cm from the transducer face. Treatment duration was controlled by a one-shot electronic timer. All exposure sets for each test group were conducted on the same day. Fragments that were pressurized during treatment were treated two at a time. Both pressurization and depressurization was accomplished in approximately 5 seconds with the fragments maintained under pressure for about 1 minute. Control fragments for the pressure tests were maintained under pressure for 5 minutes. Tap water that had been allowed to stand overnight was used for all treatments.

Post-treatment observations were made at three intervals; immediate, 10 day and 20 days, at each observation period the degree of damage and amount of new growth were recorded. Immediate damage consisted of partial to complete flooding of the lacunar chambers of an internode. Damage was expressed as a percentage of fragment internodes that were flooded. Ten day damage was usually characterized by internodes that had lost original colour, rigidity, and had generally begun to deteriorate. Twenty day damage was usually identified by brown-black internodes that became rotten, and to a lesser extent internodes that exhibited the ten day damage. New growth (the number of new internodes formed on each fragment) was expressed as a percentage of the original number of internodes before treatment.

Transducer Placement. The three transducers used in the experiments were placed in various configurations and operating modes; here they are differentiated as operating modes A-C.

Results

Transducer Performance. A review of the physical parameters surrounding the phenomenon of cavitation isolated variables considered to be important in both experimental and field applications. The two most important variables in this research application are bubble size (nuclei) in the medium, and the intensity of the sound field related to the static pressure of the water.

Measurements showed that the visual indicators of cavitational change closely corresponded with measured outputs and theoretical thresholds. The theoretical threshold to create collapse cavitation with nuclei of sizes found in tap water ($5x10^{-3}$ to $5x10^{-5}$ cm dia.) is 2-3.5 atm. Transducer measurements documented pressures of 2.5-3.0 atm. which were within the required threshold level but not exceeding them. To confirm the occurrence of collapse cavitation two further tests were conducted. The first documented the rate of erosion of sheet aluminum foil by collapse cavitation, the second involved observation of the cavitational stream under dark conditions. A sound field producing collapse cavitation emits a very faint light visible to a dark adapted observer. This radiation is called sono-luminescence and is a clear physical indicator of collapse cavitation. Observations found that stable cavitation was created at the theoretical threshold of 1 atm. and that collapse cavitation was observed at an intensity of 2.5 atm.

When the transducer was placed under static pressure, both light emission and erosion of aluminum foil began to decline at a depth of 1.5 m

(2 psi, 0.15 atm.) and were 95% eliminated at 7 m (10 psi, 0.65 atm.). The sound energy at higher pressure creates the lower form of cavitation (stable cavitation) which has a threshold of 1 atm. Nuclei subjected to stable cavitation oscillate around their equilibrium size, causing a streaming motion in the medium around the bubble.

Plant Treatment. Initial tests conducted with aquarium-grown apical fragments and the transducer in operating mode A showed that 80% of the fragment internodes were immediately damaged with exposures of 5 seconds or greater (Figure 1). Twenty days post-treatment, 85% of the internodes exposed to 2 seconds or more ultrasonic treatment were dead. Twenty days after the first treatment the fragments were retreated using the same exposures. After a further 16 days, no nodes survived or regrew in the 20 and 30 second exposure groups, in the 10, 5 and 2 second exposure groups, all original nodes had died and a substantial amount of new growth was killed. In the 0.5 second exposure group many of the original nodes were dead; however, many fragments continued to support new growth. During the same 36 day period, the control group had grown by 80%.

Close observation of fragments exposed to ultrasound showed that bubbles of water would instantly appear within the lacunae of the smaller stems (2 mm dia.). The bubbles oscillated rapidly, gradually increasing in size while slowly migrating up and down the lacunae until the entire lacunar chamber became flooded. The next test series used fragments collected during an active growing season in Cultus Lake. These fragments were hardier than the aquarium plants and the stems were noted to be more resilient and less likely to be crushed. Tests made on these fragments used transducer configuration C.

The immediate effects of the treatment appeared to be less pronounced than previous tests (Figure 2); however, observations 10 day post-treatment indicated that damage to original plant material had increased. Twenty days after treatment, 93% of original fragment material had died, with exposures of two seconds of greater. Stronger new growth was documented in the lake plants than in the aquarium plants, with the 15 second treatment having a higher rate of new growth, although still considerably lower than the new growth produced by the control fragments (77.1%).

A second collection of apical material was taken in August for lab testing using configuration C. At this time of the year the lake plants were no longer actively producing stem material but were about to auto fragment and develop adventitious roots. Immediate damage to the fragments was extensive (90%) after 10 seconds of exposure; a higher rate of damage than observed in summer growth fragments. Also, long term damage was more extensive, essentially all original fragment internodes were killed and the amount of new growth was considerably less than the previous treatment groups (Figure 3).

Incidental observations made during the course of these tests indicated that fragment deterioration was dissimilar. After 20 days post-treatment the fragments exposed to the transducer in operating mode A became dark brown with an occasional hardened internode, the expected rotting cycle. Fragments exposed to operating mode C became slimy and translucent with no structural integrity. Fragments exposed to a 50/50 split treatment (operating mode A+B=C) showed both forms of deterioration. Observations of

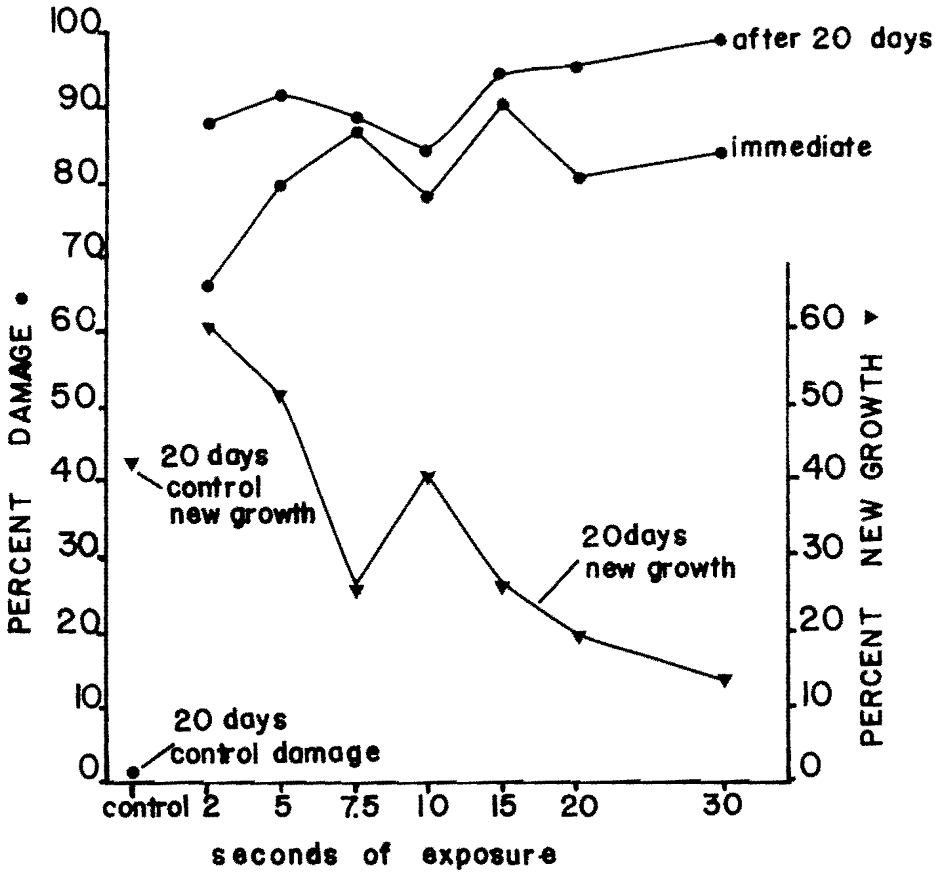

FIGURE 1. Percent damage (•) and percent new growth (▼) of aquarium grown fragments after treatment with transducers in configuration A. (n=10).

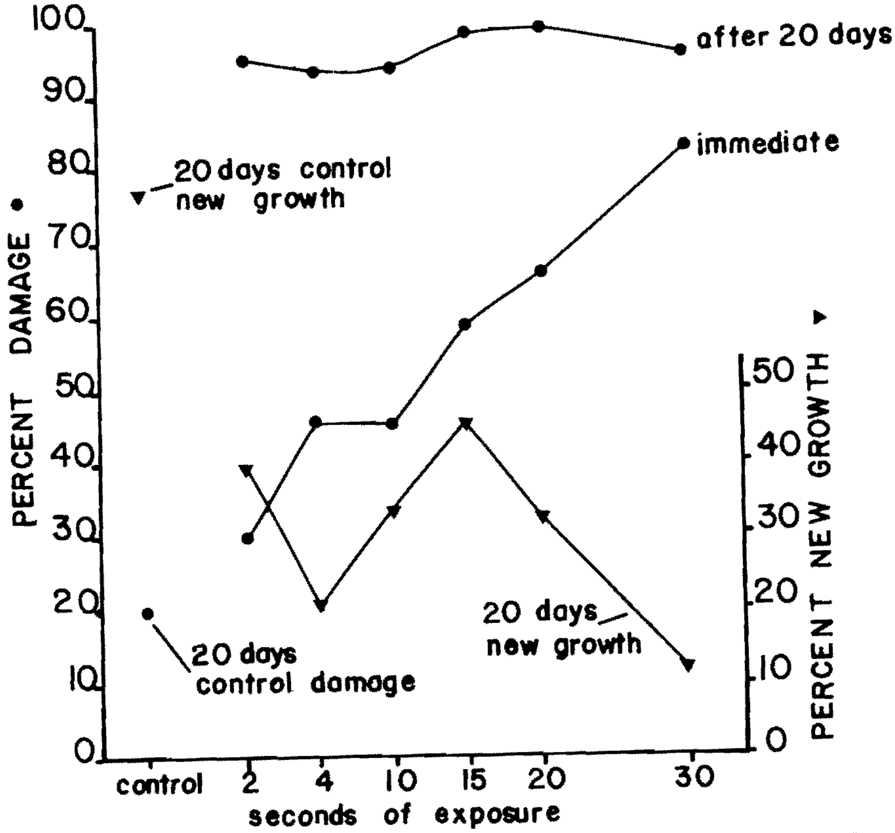

FIGURE 2. Percent damage (•) and percent new growth (▼) of Cultus Lake fragments

fragments during exposure to mode B also showed that smaller lacunae were flooded instantly. As the exposure duration increased over 10 seconds, many fragments began to disintegrate with nothing more than a fiber skeleton remaining after 20-30 seconds.

During the winter months (February-April) dormant apical tips were again collected from Cultus Lake. At this time of the year the apical fragment internodes were almost solid cellular material, in most cases having no visible lacunar structures. The stem was extremely resistant to crushing, and generally had a woody appearance and texture. After placement in the warm aquaria the fragments produced extensive new growth. The over-wintering fragments were used to determine the effect of treatment at increasing depths and the subsequent change to the threshold of collapse cavitation and plant damage. Test fragments were treated at pressures of ambient, 1.5 m, 3.0 m and 4.5 m equivalent depth, with control sets at ambient and 4.5 m. Three exposure groups of 2, 5 and 10 seconds were used at each depth with the transucer operating in mode A.

Initial post-treatment observations of the ambient test groups indicated little observed damage (Table I). Subsequent analysis of treatments at 1.5, 3.0 and 4.5 m depths showed that damage increased considerably with depth. Again, no lacunar flooding was apparent but extensive epidermal cell disruption was evident around the nodes and randomly along the internodes. At the 4.5 m depth even the 2 second treatment incurred extensive epidermal damage, and on occasion the internodes were almost cut in two. Ten days after treatment the ambient exposures had 24-31% damage while the 4.5 m exposures had 76-80% damage. By the 20th day post treatment the same pattern was still evident but the longer exposure times produced greater mortality. Figure 4 shows a plot of percent damage vs. depth for the 2 second exposure group.

Depth of treatment also had an effect on the amount of new growth produced by treatment fragments. Ten days post-treatment all three exposure levels had lower new growth production after treatment at greater depth (Figure 5). The ambient and 4.5 m controls had almost identical levels of new growth production indicating that pressurization had little physiological effect on the fragments.

In all the tests conducted, the method used to quantify the amount of new growth after treatment (used as an index of treatment effectiveness) introduced two ambiguities. The primary problem is that a count of the number of internodes does not distinguish between sizes. The internode size of post-treatment new growth is always very small in relation to the original fragment size. Therefore, in Figure 5 the 80% new growth of the 20 day ambient treatment actually represents only 5-10% new biomass when compared to the biomass of the original fragment. The control fragments which are documented as producing 85-95% new growth produced apporoximately 60% new biomass, the internodes being longer and thicker than treated fragment new growth. The second ambiguity is that the viability of the new growth is not represented in the data. For the most part, new growth of the controls is much more robust than the treatment fragment new growth. The new growth therefore must be used only as a relative index of effectiveness.

Table I. Post-Treatment Observations to Assess the Percent Damage to Overwintering Apical Fragments Treated at Equivalent Depths of Ambient, 1.5 m, 3.0 m, 4.5 m (n=20)

Observation Period	Immediate			10 Day			20 Day		
Exposure in Seconds	2 Damage %	5 Damage %	10 Damage %	2 Damage %	5 Damage %	10 Damage %	2 Damage %	5 Damage %	10 Damage %
Equivalent Depth of of Treatment									
Ambient	10	3	10	31.0	23.9	29.0	50.4	47.0	62.6
1.5 m	13	16	20	32.9	31.5	45.9	64.6	55.7	60.0
3.0 m	35	55	56	51.2	72.0	79.9	72.4	71.3	91.0
4.5 m	59	69	68	77.3	76.6	79.4	71.8	86.0	91.7
Control							Ambient	18%	
							4.5 m	15%	

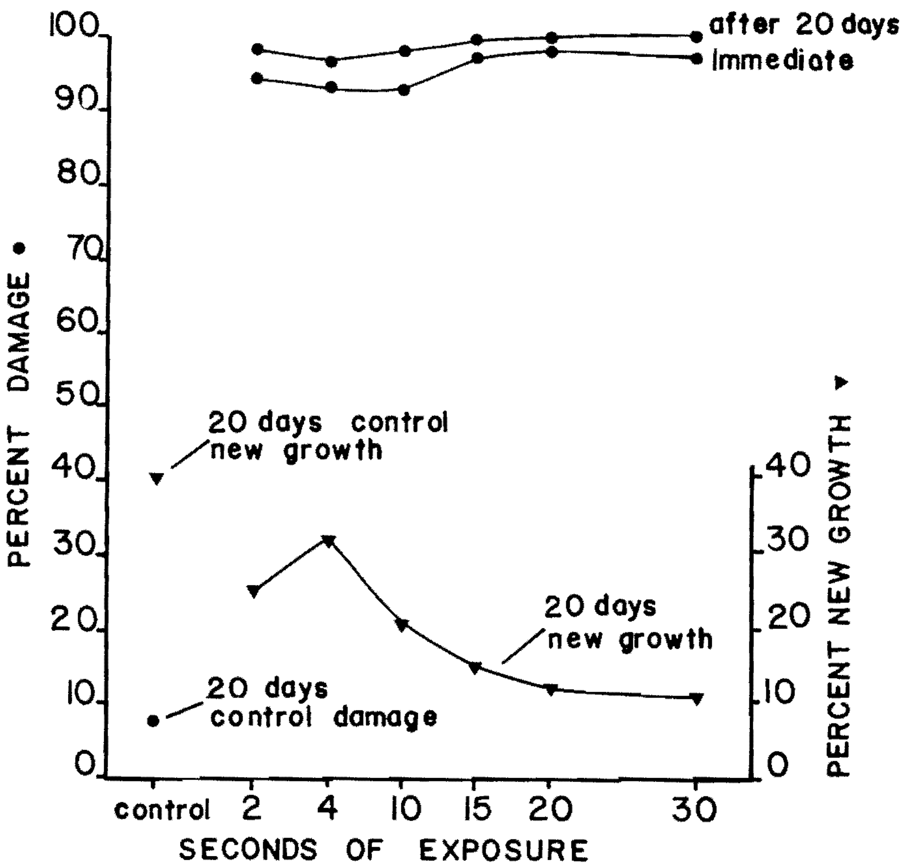

FIGURE 3 Percent damage (●) and percent new growth (▼) of Cultus Lake fragments (August) after treatment with transducers in configuration C

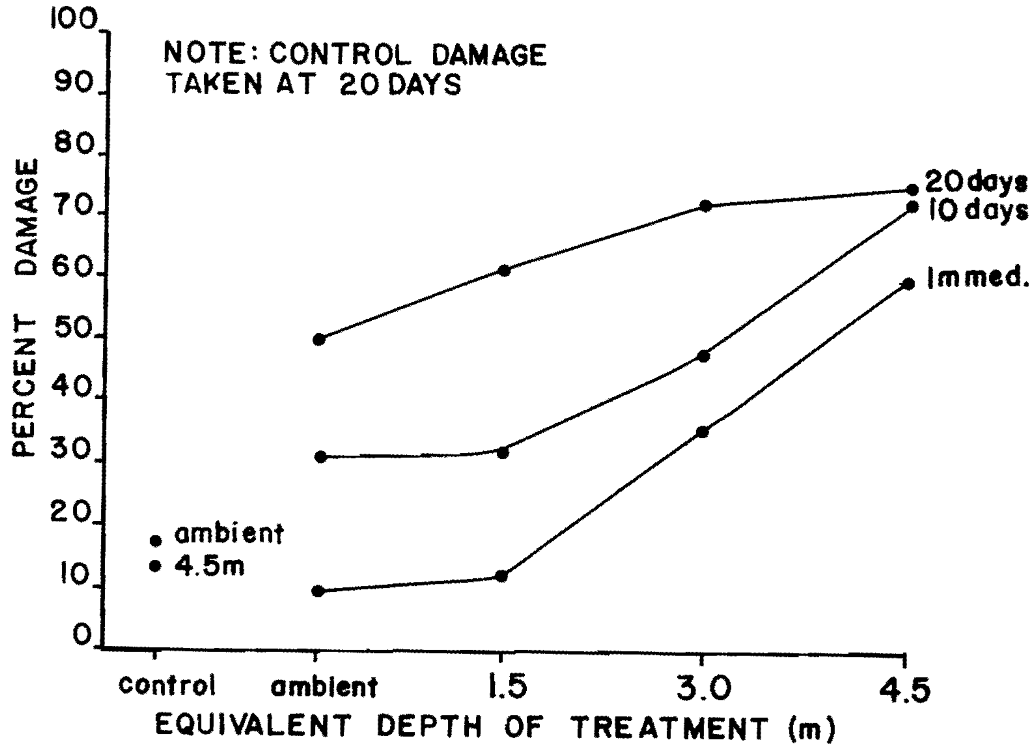

FIGURE 4 Percent damage to Cultus Lake fragments (February) treated for 2 secon
at static pressures equivalent to depths of ambient 1.5, 3.0, and 4,5 m, (n=20)
control n=40)

Table II. Post-Treatment Observations to Assess New Growth Production of Overwintering Apical Fragments Treated at Equivalent Depths of Ambient, 1.5 m, 3.0 m and 4.5 m (n=20)

Observation Period	10 day			20 Day		
Exposure/Seconds	2 Damage %	5 Damage %	10 Damage %	2 Damage %	5 Damage %	10 Damage %
Equivalent Depth Of Treatment						
Ambient	39.0	51.0	46.8	80.5	117.0	76.0
1.5 m	36.4	43.7	34.9	65.0	70.5	73.0
3.0 m	35.5	27.25	25.6	74.0	58.9	51.6
4.5 m	19.6	11.5	15.9	47.2	30.5	35.5
Control	Ambient 85% new growth			4.5 m equivalent depth 95% new growth after 20 days		

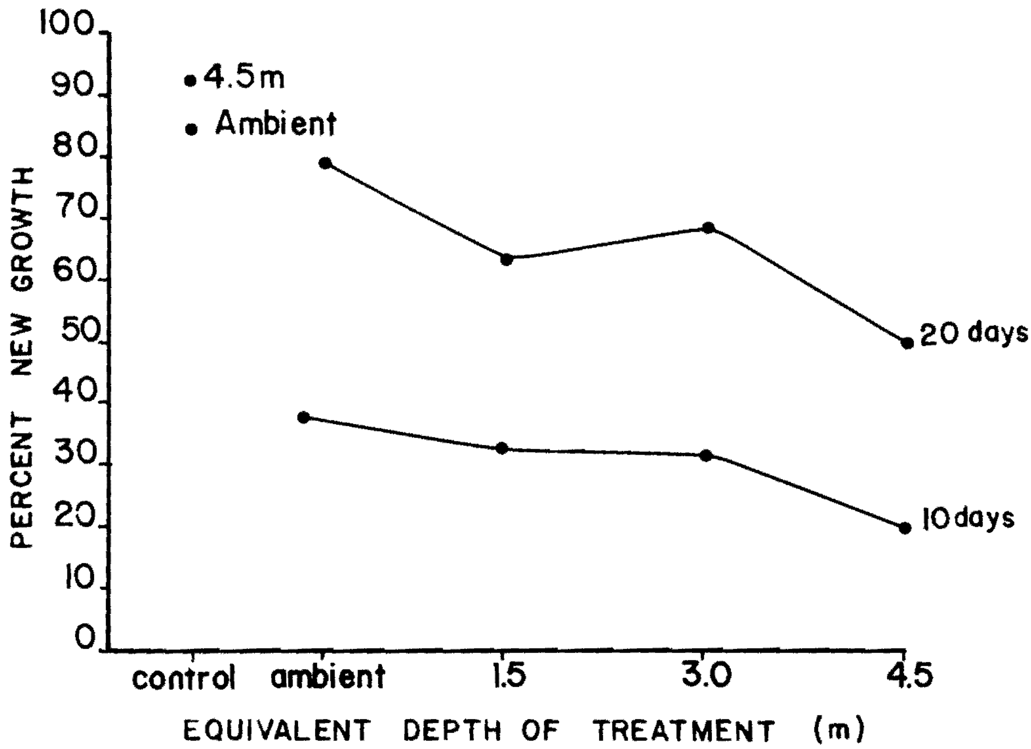

Figure 5. Post treatment new growth produced by Cultus Lake fragments (February) treated for 2 seconds at static pressures equivalent to depths of ambient, 1.5, 3.0 and 4.5 m. (n=20, control n=40).

Tests have been conducted to determine the minimum exposure to obtain 100% fragment mortality with no new growth production. Tests at ambient pressure using configuration B and an exposure of 45 seconds caused massive disruption of the meristematic cellular structure and left only a translucent fibrous skeleton of the fragment. Tests conducted at equivalent pressures of 4.5 m required only a 20 second exposure using configuration A, to obtain 100% mortality within eight days of treatment.

Root Crowns. Preliminary tests conducted on root crown meristems and rhizome material (unpigmented subterranean apical shoots) indicated that these dense cellular structures are more resistant to the effect of ultrasound and will require more intense exposures.

Adventitious Roots. Occasionally during the treatment of a fragment adventitious roots were present; those exposed to ultrasound were affected and showed deterioration. Microscopic examination of treated roots most often found massive disruption and removal of the epidermal cell layers. When the xylem of the root was examined through these "holes" the xylem tracheae were often seen to be physically dislocated. The roots invariably rotted within a few days of treatment.

Discussion

The purpose of this project has been to conduct laboratory studies on the effectiveness of high powered ultrasound to be used as a device to control aquatic plants.

The results presented here indicate that Eurasian water milfoil is susceptible to high powered ultrasonics. Although only limited testing has been possible, some definitive conclusions can be drawn from the data collected.

Of primary interest is the variation in level of mortality dependent on the season in which the apical fragments were collected. The hardiest or most difficult fragments to damage are the overwintering apical tips; the more "tender" August apical tips are the most susceptible to damage. Also, aquarium grown fragments are particularly susceptible to physical disruption and instantaneous flooding of the lacunae. The present effectiveness of treatments can be directly related to the outward physical toughness of the fragments. Overwintering fragments that possess thick epidermal structures with negligible air lacunae formation are the most resistant. As the immediate effects of lacunar flooding are suggested to be a result of changed osmotic potential, the rigid cell structure of older overwintering plants must be resistant to this immediate effect. As considerable post-treatment damage was still observed on overwintering apices, other destructive processes are obviously pervading.

The most marked post-treatment effect on summer growth apical material was the occurrence of brown spots within the stem usually within 18 hours of treatment. These spots nearly always corresponded with the location of lacunar flooding. After several days the brown spots would darken and enlarge until they emerged through the epidermal cell layer of the stem. The rotting of the stem was observed to spread through an internode but would not jump a node to a neighbouring unaffected internode. Microscopic examination of stem material in areas affected showed no mechanical damage to the surrounding cell structures. It is tentatively suggested that the lacunar flooding followed by internal stem deterioration is caused by chemical reactions. The most probable reaction would be the breakdown of water within the lacunae, under sonnication, forming aqueous free radicals that would recombine to form H_2O_2. This molecularly weak compound may still break down the plant's internal structure and may cause the brown spots. The actual mechanism of deterioration must be determined through additional research.

With the increase of ambient pressure relative to the transducer pressure wave and the subsequent reduction in the amount of collapse cavitation generated at depth, it was expected that less damage would occur to plants treated at depth. However, this was not observed experimentally since treatments conducted at 1.5 m were marginally more effective than those at ambient, whereas tests conducted at 3.0 and 4.5 m equivalent depth were considerably more effective. As the amount of collapse cavitation is decreased at this depth and has been replaced by stable cavitation it is thought that a very disruptive micro-streaming of cell contents is occurring that is sufficient to disrupt the cells. Another favourable observation about treatment damage at depth was the noticeable cell disruption around the nodes and leaf stalks. The prevalence and severity of damage for this exposure duration had not been previously observed. The cause of this effect is still unknown.

Reduction of new growth from nodes or apical material appears to be the most important approach to achieving control of aquatic plants. Treatment of the root crown, the eventual goal to allow feasible control of overwintering aquatic plants, will be investigated as soon as control of nodal regrowth has been accomplished satisfactorily in the lab. It is expected that once this is achieved there will be few complications in treating the exposed root crown. At this time, treatment effects of the sediment penetrating qualities of ultrasound will be studied through analysis of disruption in the root mass. Also, it is expected that new growth production of treated _in situ_ plant material will be reduced compared to that documented in the lab. Treatment of overwintering plant material that is still subject to frigid water temperatures and short daylight hours may in fact, completely inhibit any recovery potential of plants such as that exhibited by aquaria plants.

The first generation transducers tested during this study demonstrated that ultrasound and cavitational streaming have the ability to disrupt, and with longer exposures, to totally disintegrate aquatic plant material. To produce effective _in situ_ treatments that would allow for coverage of 1 ha per day by an ultrasonic array, effective static exposures in the lab must be reduced to the two second level. With the design and construction of second generation transducers almost complete, it is anticipated that these objectives soon will be accomplished. Using the same input power, these designs concentrate from five to twelve times the cavitational energy of to the surrounding cell structures. It is tentatively suggested that the lacunar flooding followed by internal stem deterioration is caused by chemical reactions. The most probable reaction would be the breakdown of water within the lacunae, under sonnication, forming aqueous free radicals that would recombine to form H_2O_2. This molecularly weak compound may still break down the plant's internal structure and may cause the brown spots. The actual mechanism of deterioration must be determined through additional research.

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Summary

1. The development of ultrasound as a plant control method would have many advantages over present control methods; they include: treatment of dormant plants, no plant disposal, immediate on/off capabilities, no drift, long equipment life expectancy and elimination of environmental conflicts.
2. The plants most susceptible to damage by ultrasound are new summer shoots. Winter plant material is more resistent to treatments due to its dense cell structure.
3. Exposure to ultrasound changes the osmotic potential of the epidermal cells allowing lacunar flooding.
4. The formation of H_2O_2 is tentatively suggested to occur and cause damage within irradiated plant material.
5. For any exposure duration, increased depth of treatment causes substantially greater mortality of the plant fragment and reduce new growth potential.
6. Using first generation transducers, considerable damage to plant material,has been incurred at target exposures of two seconds or more.
7. Second generation transducers now being constructed will be 5-12 times more powerful than transducers used to date and promise to provide practical control of all meristematic plant parts.

Acknowledgments

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Short-term Impact of Harvesting of Eurasian Watermilfoil

D.S. Painter and J.I. Waltho

National Water Research Institute
Environment Canada
Burlington, Ontario, Canada, L7R 4A6

The short-term efficacy of harvesting of Eurasian watermilfoil in Buckhorn Lake, Ontario was dramatically influenced by the timing of the cut. Nineteen harvesting scenarios were examined for their effects on milfoil regrowth and tissue chemistry as well as the amount of open water created. A June/August or June/September double cut would appear to be the most desirable scenario with very little advantage in a triple cut. Milfoil biomass was significantly affected in the second year by a cut in October of the preceding year.

Shoot and root phosphorus, nitrogen, carbon and carbohydrates were altered by harvesting. The tissue chemistry was altered in the spring of the second year, particularly if a September or October cut was performed. However, by the summer of the second year no differences in tissue chemistry were observed except in root total non-structural carbohydrates which were significantly reduced.

The ecological consequences of mechanical harvesting have been studied and no adverse impacts have been reported (1-7). Harvesting is, however, criticized since multiple harvests may be required each growing season and no long-term effect on regrowth may be apparent. A review of 13 reported harvesting projects concluded that *Myriophyllum spicatum* was controlled most effectively by harvests that remove as much shoot material as possible several times during the growing season and the evidence suggested that harvests in late September or early October should most effectively reduce biomass the following year (8). This paper examines the short-term effect of many harvesting scenarios on milfoil regrowth and tissue chemistry.

Methods

The experiment was designed to determine the most effective harvesting schedule from 19 possible schedules. Twenty-four 2 X 10 meter plots were established in 1.6 to 1.75 meters of water on the west side of Nichol Island in Buckhorn Lake, Ontario, Canada on June 5-7, 1979. Single, double, and triple cuts were performed from June to October 1979. A cut was also performed in May 1980 in a plot that had been cut in October 1979. The plots were cut at 0.5 meters above the sediment using Scuba equipment and small sickles. Monthly sampling was performed from June to December 1979 and April to August 1980. All plant and sediment samples were obtained using Scuba. Fresh weight and dry weight were measured at the beginning and end of the study based on one 0.25 m^2 quadrat per plot at the beginning and triplicates per plot at the end. Plant height was determined by measuring the length of 25 random stems which achieved an allowable 95% confidence error of 5 cm. Shoot and root samples were analyzed for % water, % organic content, total non-structural carbohydrates, total nitrogen, total phosphorus and total carbon. Sediment cores were obtained and the 0 to 40 cm section was analyzed for loss on ignition, total phosphorus and total nitrogen.

Water content was determined by weight difference after samples were dried for 16 hours at 75°C. Loss on ignition (% organic content) was determined on dried plant material which was muffled at 550°C for two hours. Total non-structural carbohydrates were determined by enzymatic extraction with amyloglucosidase for conversion of starches to glucose and glucose analysis using the phenol-sulphuric acid colorimetric method (9). Total phosphorus, total nitrogen and total carbon were determined as per the Analytical Methods Manual (10). The loss on ignition values were used to correct the chemical analysis, initially expressed on a dry weight basis, to an ash-free dry weight basis (AFDW).

Results and Discussion

Short term effect of harvesting on milfoil growth. Plant height was chosen as the most appropriate indicator of the impact of harvesting and subsequent milfoil regrowth because the goal of harvesting is to create an unobstructed water column for recreational use. The error involved in quadrat sampling and the small size of the plots necessitated the use of a non-destructive sampling method such as plant height. The efficacy of a particular harvesting schedule was evaluated by determining the number of days the water column remained unobstructed in the top 50 cm. The 50 cm unobstructed water depth was chosen as the criteria for suggesting whether the area was useable for recreation. The days open was also subdivided into days during the tourist season (June 1 to October 15) and total days (May 15 to November 15). Figure 1 illustrates the days open created by the 18 harvesting schedules performed in the first year. The control plot had 0 days open. Striking differences in impact are apparent fo the various harvesting scenarios. The

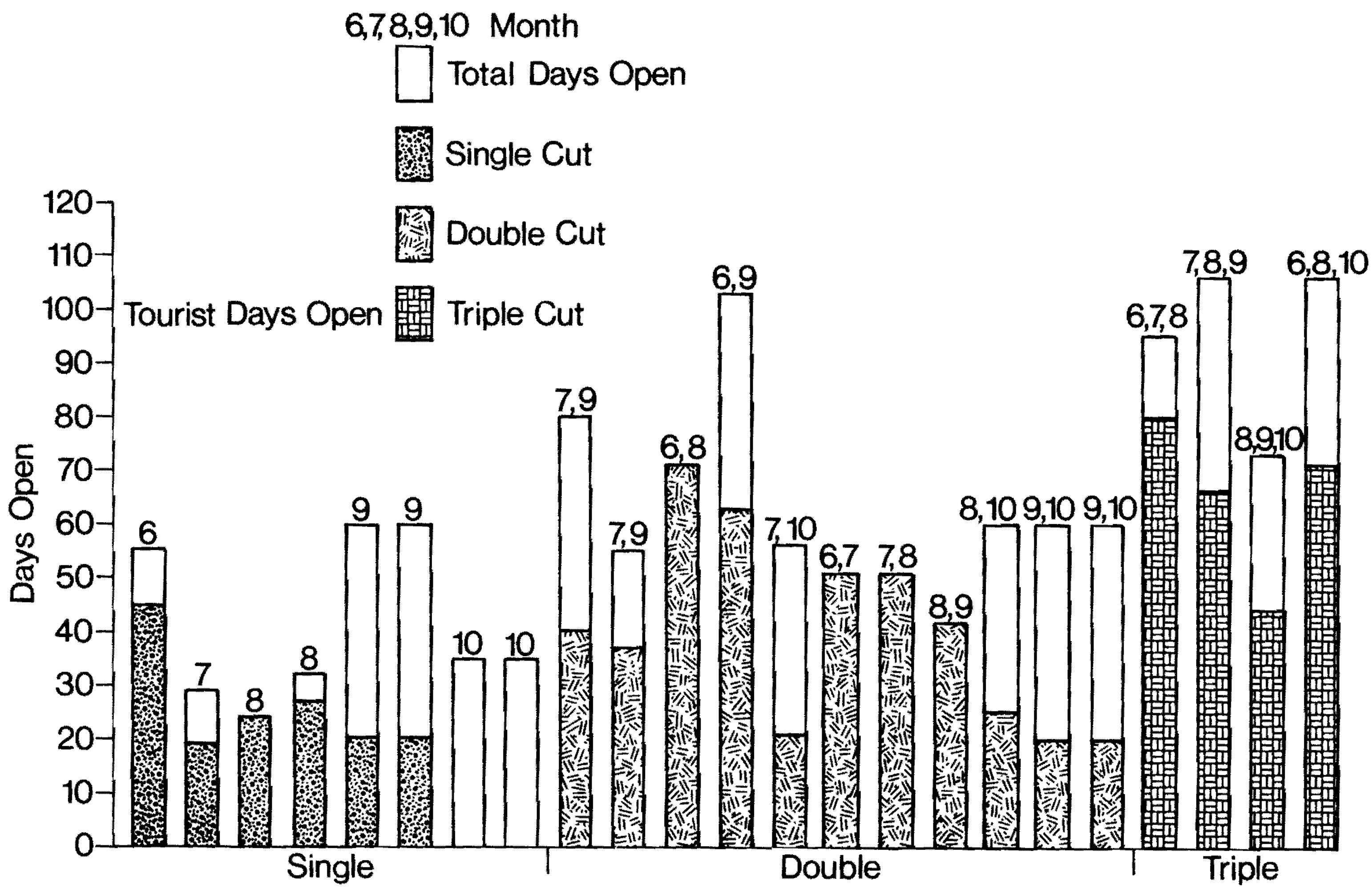

Figure 1 DAYS OPEN CREATED BY 18 HARVESTING SCHEDULES

usefulness of the informaton to lake managers in planning harvesting timetables or determining equipment requirements is obvious. For example, a single cut should be performed early in June at the beginning of the tourist season to maximize effect per unit effort. A single cut in July has a very short impact. In fact, the properly timed June single cut was as effective or more effective than many of the double cuts except for June/August, June/September and July/September. If the 45 days of open water during the tourist season that a single June cut creates is insufficient in the eyes of the lake manager and equipment is available, then the double cuts just mentioned particularly June/August or June/September could increase the days open during the tourist season to 60-70 days and total days open to 104 in the case of June/September. The effort involved in a triple cut scenario would be wasted provided a properly timed double cut was possble. Figure 2 illustrates the actual plant heights observed in 1979, the harvesting year, and in 1980, the recovery year, for several harvesting scenarios.

Another approach to illustrate the impact of harvesting would be to measure the areas in Figure 2 where the plant height was below the surface. If the plants were at the surface then the percent open water would be 0% and as the harvesting impact on plant height increased, the percentage open water would increase. Table 1 summarizes the % open water and days open, both tourist and total, for the harvesting scenarios tested. The control plot had only 10.6% open water. The best single cut (June) increased the % open water to 28.2%. The best double cut (June/September) had 40.7% open water and the triple cuts could only increase the % open water to 43.5%. The % open water and days open for replicate plots were similar.

The impact of the 1979 harvesting on regrowth during the 1980 season was also determined using the same approach. Table 1 also summarizes the % open water and days open during the 1980 season up to July 15. The control area had 31.5% open water and 19 days open. The best single cut (June 79) had 39.7% open water and 26 days open. The best double cut (July/September 79) had 40.2% open water and 26 days open. Triple cuts had no increased effect on the 1980 regrowth compared to the best double cut. The cut performed during May 1980 on the plot that had also been cut in October 1979 (10-5 in Table 1) resulted in 41.9% open water and 30 days open. Although the 1979 cuts did affect regrowth in 1980, the effect was minimal with an increase in % open water of only 10% and an increase in days open of only 10 days. The early May cut in 1980 appeared to have no advantage based on plant height.

The impact of the harvesting scenarios on the plant biomass during the second season was also determined by sampling plant biomass directly by quadrat sampling in August 1980. Figure 3 illustrates the dry weight of the milfoil per m^2 in August 1980 of the 19 harvested plots and the control plot. A one way analysis of variance was performed on the plant dry weights in each plot and the plots which were found to be significantly different (95%) from the control have the standard error bars included on the figure. The only significant trend discernible is that those harvesting schedules that included a harvesting in October 1979 had significantly less biomass in

Table I. % Open Water and Days Open for 1979 and 1980

Cut	1979			1980	
	%Open	Days Open		%Open	Days Open
		Tourist	Total		
Single Cuts					
6	28.2	45	55	39.7	26
7	20.3	19	29	37.9	24
8	16.0	24	24	36.0	22
8	18.9	27	32	36.2	22
9	20.0	20	60	33.0	21
9	20.0	20	60	36.0	23
10	17.9	0	35	34.0	21
10-5	19.9	0	35	41.9	30
Double Cuts					
7-9	30.7	40	80	40.2	26
7-9	25.9	37	55	40.3	25
6-8	33.5	71	71	36.8	23
6-9	40.7	63	103	35.5	22
7-10	27.5	21	56	35.0	21
6-7	32.7	51	51	34.5	22
7-8	28.7	51	51	35.0	22
8-9	22.8	42	42	37.8	24
9-10	27.3	20	60	36.0	24
9-10	26.7	20	60	37.5	24
8-10	29.0	25	60	35.3	22
Triple Cuts					
6-7-8	38.6	80	95	40.3	27
7-8-9	43.3	66	106	39.0	26
8-9-10	35.3	44	73	36.7	23
6-8-10	43.5	71	106	39.0	24
Control	10.6	0	0	31.5	19

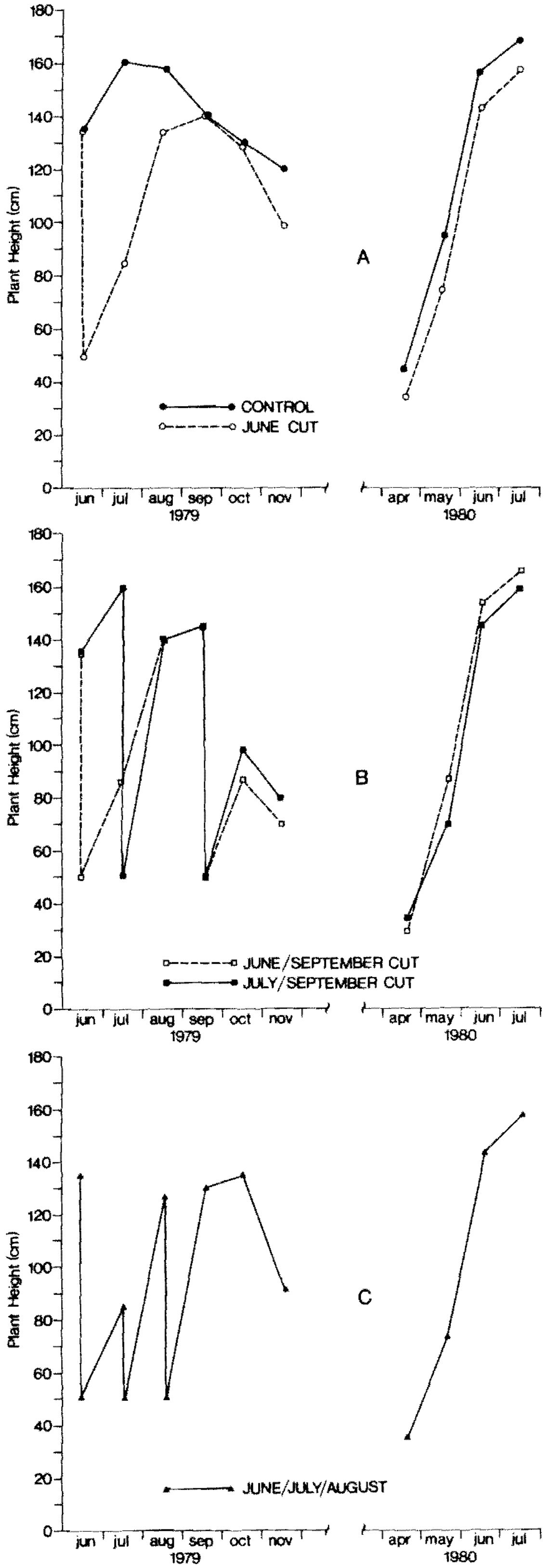

Figure 2 PLANT HEIGHT RESPONSE TO SINGLE (A), DOUBLE (B), AND TRIPLE (C) CUTS

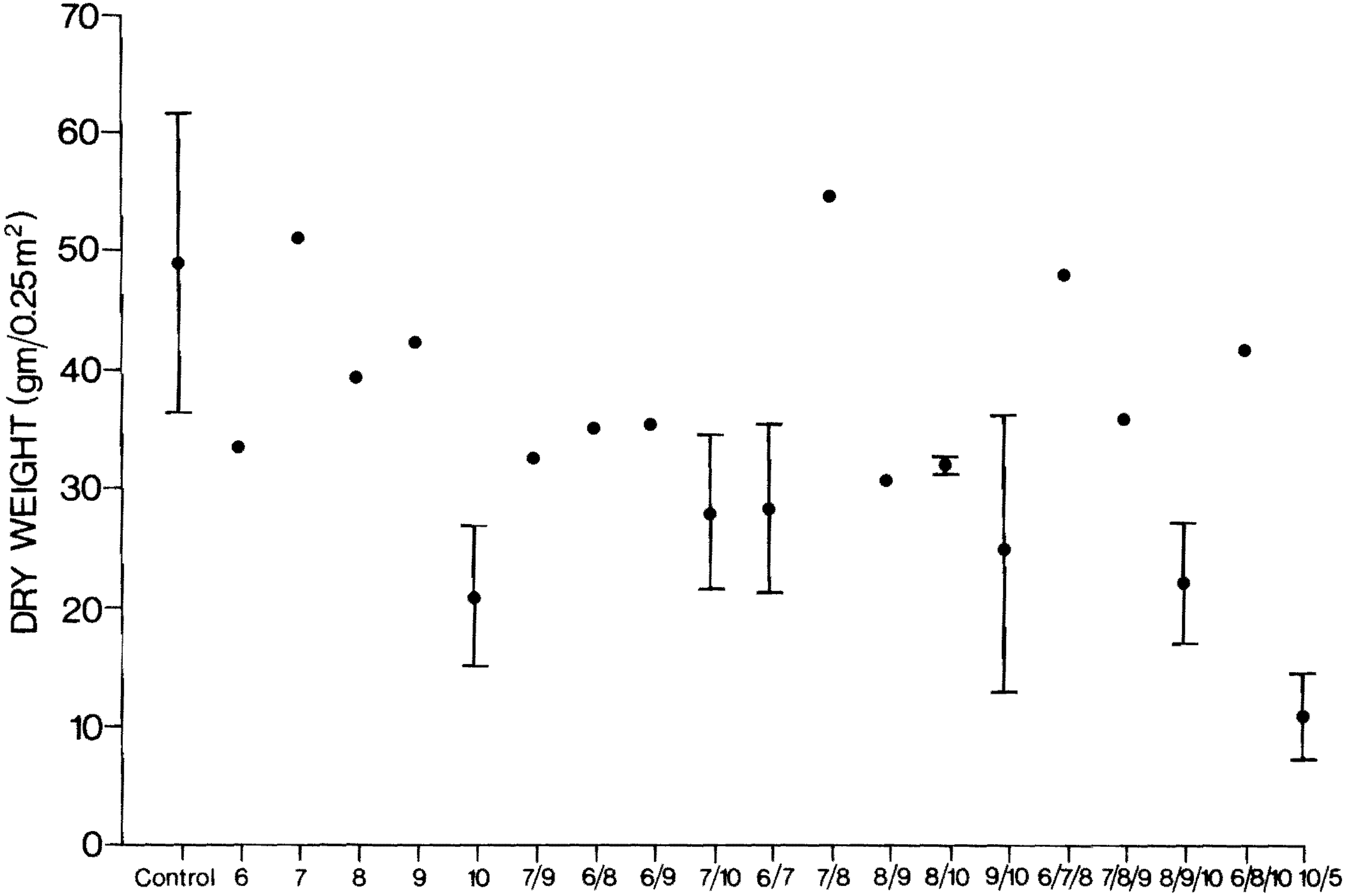

Figure 3 MEAN DRY WEIGHT IN AUGUST 1980 VERSUS CUTTING SCHEDULE
(Error bars are one standard deviation, and are provided for those plots which are significantly different from control.)

August 1980. The harvesting in October79/May80 also appears to have had a significant impact of biomass when the results are interpreted on an areal dry weight basis instead of plant height.

Short term effects of harvesting on milfoil tissue chemistry. Shoot and root tissue samples were analyzed for phosphorus, nitrogen, total carbon and total non-structural carbohydrates (TNC). The tissue chemistry was measured to determine if the harvesting effects on regrowth could be explained by an analysis of the tissue chemistry and then exploiting the effects on tissue chemistry in a long-term harvesting experiment. Figures 4 through 7 illustrate the seasonal trends of tissue chemistry of the control and the June/August/October harvest. The effect of the triple harvest on shoot phosphorus can be observed in Figure 4a. Tissue phosphorus increased in the month following harvesting. However, shoot phosphorus returned to values similar to control in the second month following harvest, except for the October cut, where the effect on shoot phosphorus continued through to the spring of the second year. Shoot phosphorus increased in the month following harvesting compared to the control tissue phosphorus in 24 of 36 cases or 66.7%. The average increase in tissue phosphorus was 364 ug P/g AFDW. The mean seasonal phosphorus concentration was 1934 ug P/g AFDW, so the phosphorus increase due to harvesting was 19% of the seasonal mean. Figure 4b illustrates the effect of the triple cut on root phosphorus. Root phosphorus also increased in the month following harvest and returned to values similar to control in the second month. The October cut affected root phosphorus in the spring of the second year but the effect did not extend into the summer. Root phosphorus increased in the month following harvesting in 21 of 36 cases or 58%. The average increase was 187.5 ug P/g AFDW. The mean seasonal root phosphorus was 1227.5 ug P/g AFDW, so the phosphorus increase due to harvesting was 15.3% of the seasonal mean.

Shoot nitrogen response is illustrated in Figure 5a and again the response is similar to phosphorus. The shoot nitrogen rose in the month following harvest but dropped to values similar to control in the second month. The October cut influenced the spring shoot nitrogen but the effect did not last into the summer. Shoot nitrogen increased in 19 of 28 cases or 67.9% following harvesting. The average increase was 0.324% N (AFDW) and the seasonal mean was 2.32% N, so the nitrogen increase due to harvesting was 14% of the seasonal mean. The effect of the triple cut on root nitrogen is illustrated in Figure 5b. The response of root nitrogen to cutting was similar to those previously described with increases occurring in the month following harvesting. Root nitrogen increased in 24 of 28 cases or 85.7% following harvesting. The average increase was 0.555% (AFDW) and the seasonal mean was 1.744% N, so the nitrogen increase due to harvesting was 32% of the seasonal mean.

Figure 6a illustrated the response of shoot carbon to the triple harvest. Shoot carbon decreased in the months following harvesting but the effect did not extend into the second season. Shoot carbon decreased in 23

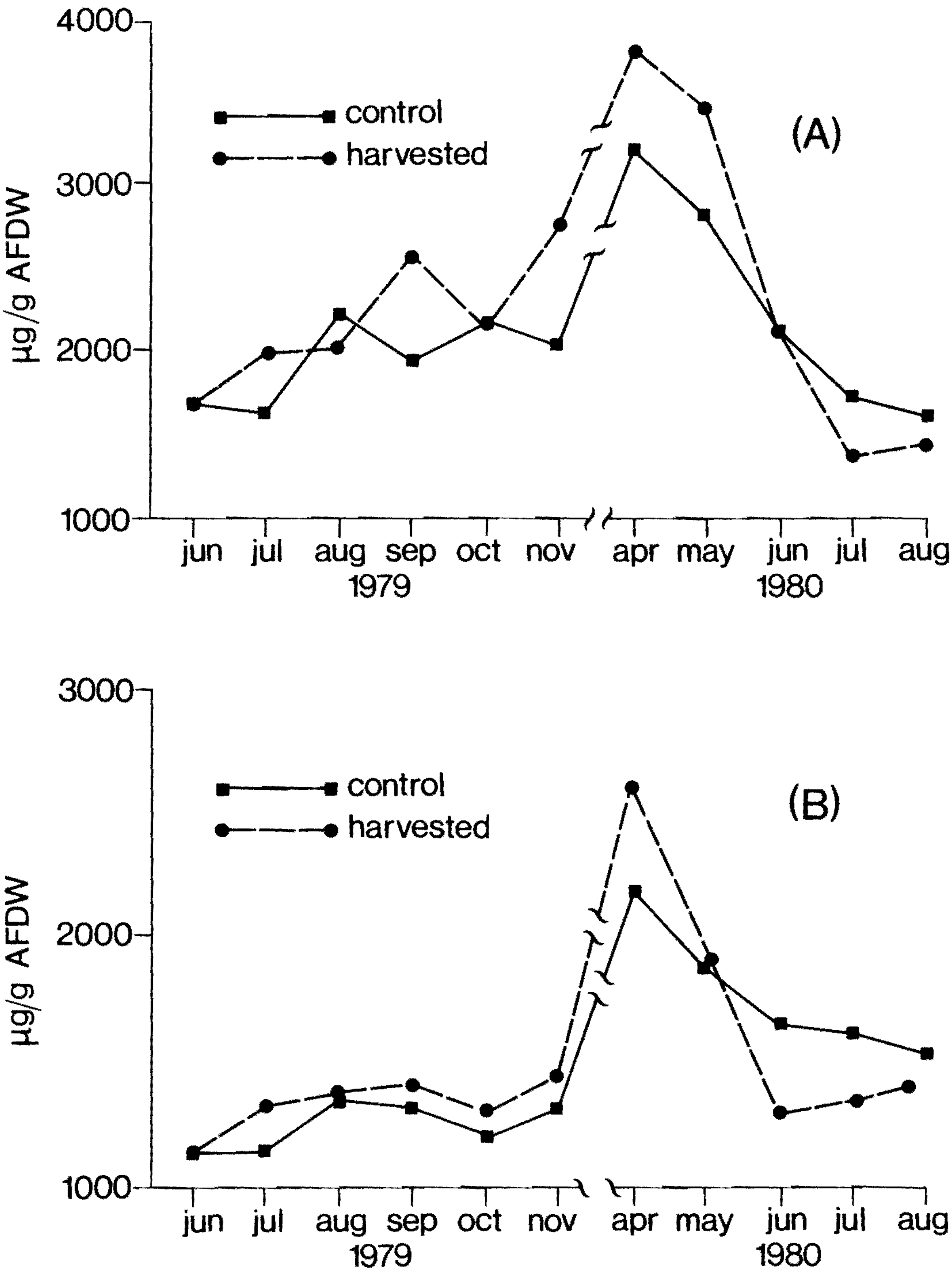

Figure 4 SHOOT PHOSPHORUS (A) AND ROOT PHOSPHORUS (B) FOR THE JUNE/AUGUST/OCTOBER CUT

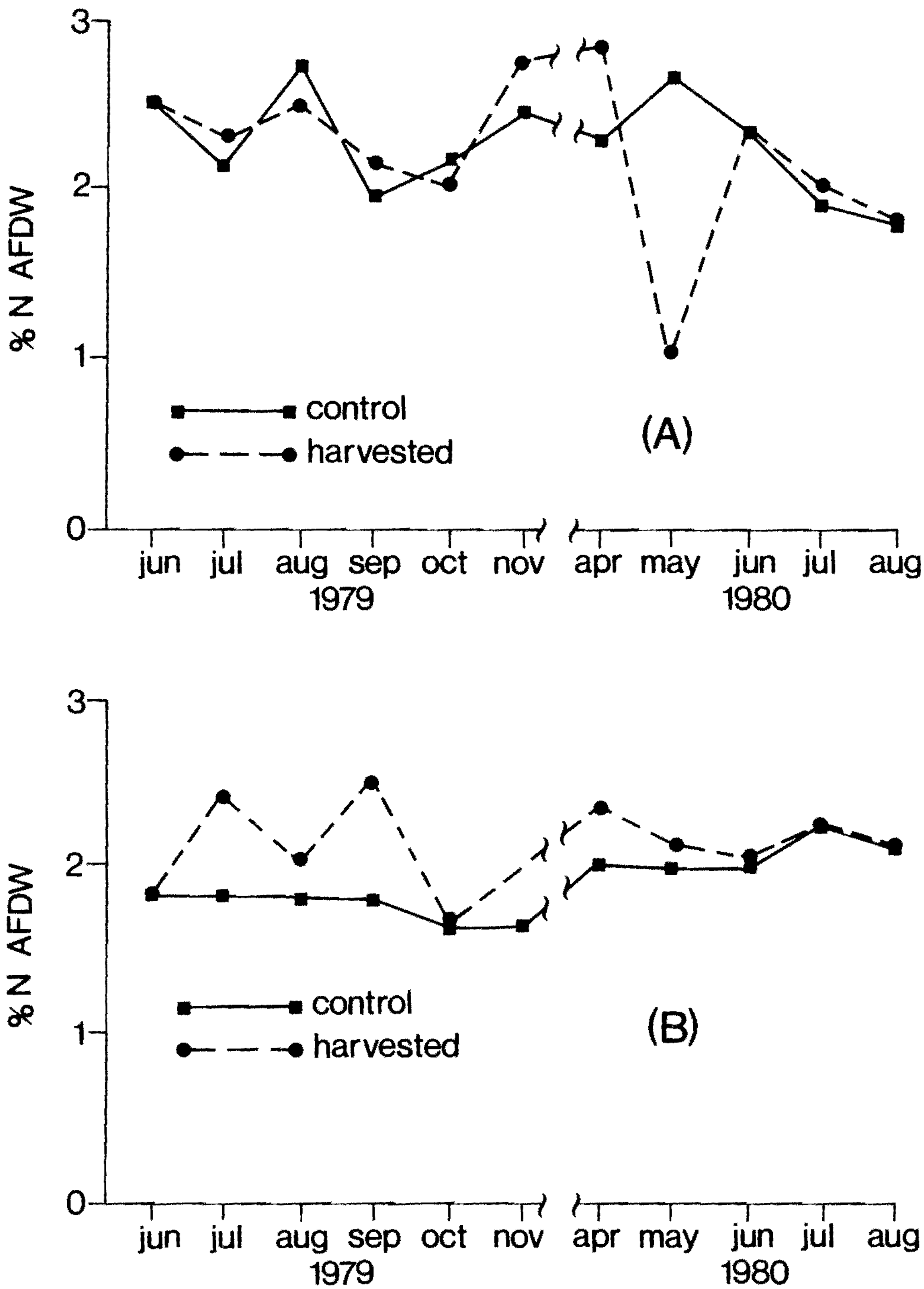

Figure 5 SHOOT NITROGEN (A) AND ROOT NITROGEN (B) FOR THE JUNE/AUGUST/OCTOBER CUT

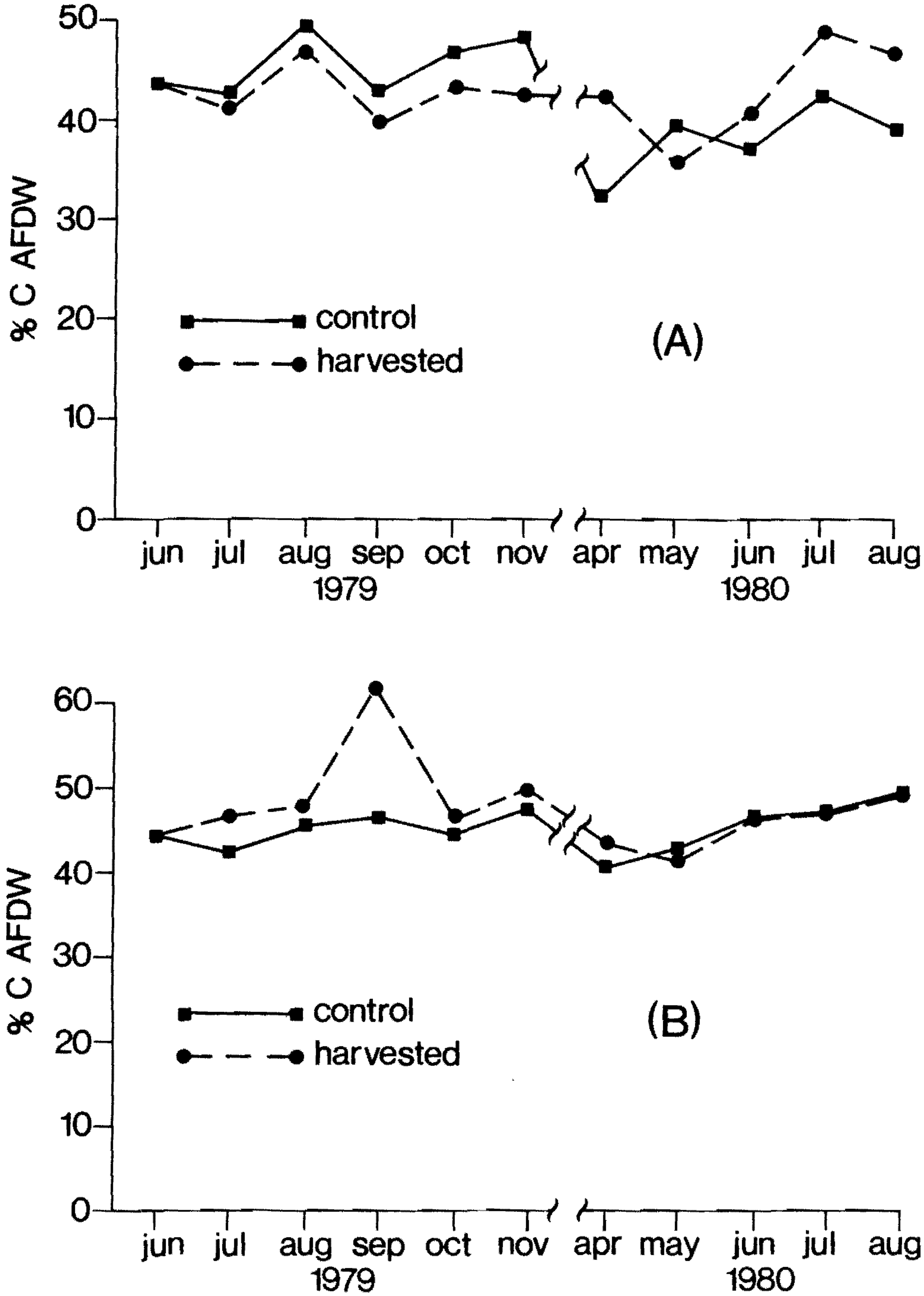

Figure 6 SHOOT CARBON (A) AND ROOT CARBON (B) FOR THE JUNE/AUGUST/OCTOBER CUT

of 28 cases or 82.1% following harvesting. The average decrease was 2.9% C (AFDW) and the seasonal mean was 45.37% C, so the carbon decrease due to harvesting was 6.4% of the seasonal mean. Figure 6b illustrates the response of root carbon to harvesting. The root carbon increased in the month following harvesting and returned to values similar to control root carbon in the second month. Root carbon increased in 20 of 28 cases or 71.4% following harvesting. The average increase was 4.1% and the seasonal mean was 45.28%, so the root carbon increase due to harvesting was 9% of the seasonal mean.

The effect of the triple cut on shoot total non-structural carbohydrates (TNC) can be observed in Figure 7a. Shoot TNC decreased following harvesting and the effect extended throughout the first season becoming progressively more pronounced towards the end of the season. Shoot TNC in the spring of the second season was similar to control shoot TNC. However, the June shoot TNC was much reduced compared to the control. A reduction in shoot TNC in June of the second season occurred in this example but was not the norm amongst the other harvesting schedules tested. Shoot total non-structural carbohydrates decreased in 32 of 37 cases or 86.5% following harvesting. The average decrease was 11% TNC (AFDW) and the seasonal mean was 40.67% TNC, so the decrease due to harvesting was 27% of the seasonal mean. Figure 7b illustrates the response of root TNC to the triple cut. Root TNC decreased following harvest especially in the fall. Root TNC in the spring of the second season was lower than the control in 18 of 19 examples. The mean root TNC in June was 15.4% compared to the control root TNC of 28.6%, a reduction of 46.3%. Root TNC decreased in 28 of 37 cases or 75.7% following harvesting. The average decrease was 9.9% TNC (AFDW) and the seasonal mean was 27.3% TNC, so the decrease due to harvesting was 36.3% of the seasonal mean.

The effect of harvesting on milfoil tissue chemistry was evident in 1979 and in some cases carried through to April of 1980. Effects on spring 1980 tissue chemistry were particularly evident if a cut was performed in September or October of 1979 but by the summer of 1980 no differences in tissue chemistry except root TNC were evident in any of the harvested plots.

Decreases in root total non-structural carbohydrates have been previously observed following harvesting and are probably a result of less photosynthetic tissue available for carbohydrate production and mobilization of root reserves to support new growth (11). The increase in tissue phosphorus and nitrogen in both the shoots and roots are most probably due to accumulation in tissues as a result of a much reduced demand due again to a reduction in shoot material. The reduction in shoot total carbon of 3% C from a season mean of 46.9% C probably reflects the decrease in shoot carbohydrates of 11 % TNC from a seasonal mean of 40.7% TNC. The decrease in TNC is, however, larger than the decrease in total carbon. Therefore, it appears that the structural carbon of the harvested plants probably increased on a percentage basis. Since the plant stems remaining after cutting are the stouter stems at the base of the plant, the conjectured increase in structural carbon after harvesting seems reasonable. The increase in root

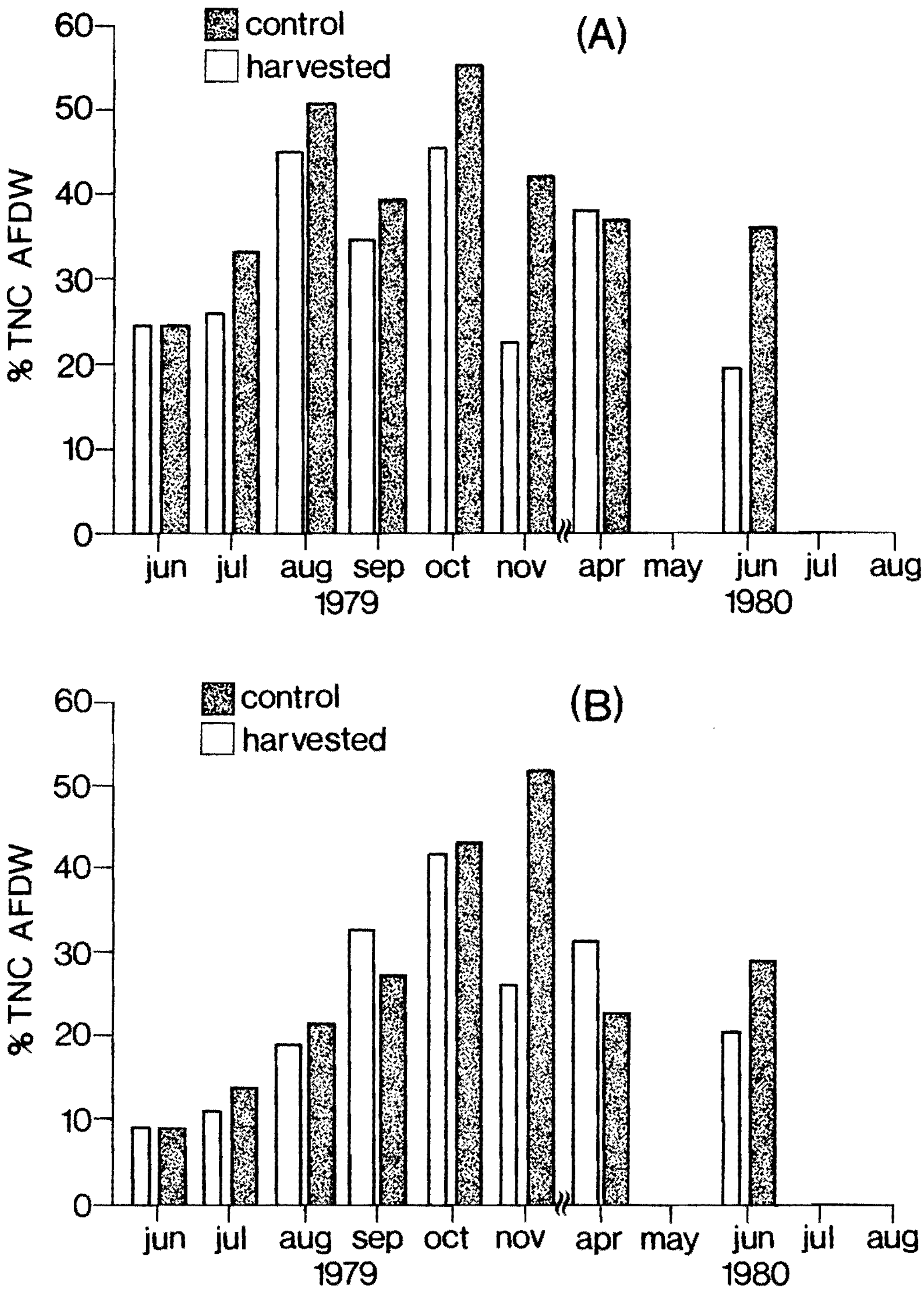

Figure 7 SEASONAL SHOOT TNC (A) AND ROOT TNC (B) FOR THE JUNE/AUGUST/OCTOBER CUT

carbon following harvesting is rather surprising considering the 36.3% drop in root total non-structural carbohydrates. As in the argument with the shoot carbon, the root structural carbon probably increased but even more dramatically than the shoots. The root masses were visually observed to be much smaller after harvesting. Therefore, it would appear that a certain amount of root death occurred leaving only the stouter roots which would explain the increase in structural carbon but a decrease in root carbohydrates.

Short term effects of harvesting on sediment chemistry. No observable changes in sediment total phosphorus or nitrogen occurred in the first or second season within the rooting depth of milfoil (0-40 cm). Total phosphorus averaged 1000 ug P/g and total nitrogen averaged 2.5% N. The total phosphorus and total nitrogen values exhibited very little change throughout the season indicating that the milfoil growth demands were supplied by a sediment pool size much smaller than the total pool.

Conclusions

The timing of a harvesting program was observed to dramatically influence the short-term efficacy of the cutting when judged by the duration of open water created. Proper timing of cuts can ensure efficient use of equipment and resources. A June/August or June/September double cut would appear to be most desirable with very little advantage in a triple cut. Plant height appeared not to be affected in the second year by any harvesting schedule; however, plant biomass on an areal basis was significantly affected in the second summer by a cut in October of the preceding year.

Tissue chemistry was altered by harvesting. Total non-structural carbohydrates of both shoots and roots decreased. Shoot and root phosphorus and nitrogen increased following harvesting. Shoot carbon decreased and root carbon increased following harvesting. The tissue chemistry was altered in the spring of the second year particularly if a September or October cut was performed; however, by the summer of the second year, no differences in tissue chemistry were observed except in root TNC which was significantly reduced.

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Fate and Impact of 2,4-D in a Pond Ecosystem

E. Nagy, D.S. Painter, and B.F. Scott

National Water Research Institute
Environment Canada
Burlington, Ontario, Canada, L7R 4A6

Experimental pond ecosystems were used to monitor the fate and effect of two 2,4-D formulations in a two-year study. At an application rate that produced a 1 ppm nominal concentration in the water phase, the chemical was effective in milfoil control. The treatments did not produce significant effects on the bacteria, fungi, phytoplankton and zooplankton communities. White sucker fry exhibited some mortality during the first few days following treatment in the first year, but none in the second year. Adult fish were not affected in either year. After the collapse of the milfoil, increased bacteria populations were observed, clams and snails showed enhanced growth, and the zoobenthos shifted to oligochaete dominated populations. The 2,4-D persisted in the water and sediment for most of the summer and fall seasons, with its disappearance from the water showing half lives of 17 and 35 days in the two years. Some of the observed effects were attributed to the presence of 2,4-DCP in the system.

The recent invasion into Canadian waters of Eurasian watermilfoil, _Myriophyllum spicatum_, the chemical control of which relies on the use of 2,4-dichlorophenoxyacetic acid (2,4-D), has resulted in both an increased use of the chemical and increased public concern with the safety of its use. In Canada, hundreds of tonnes of 2,4-D are used annually for the control of terrestrial and aquatic weeds. In addition to water weed control, the chemical can enter the aquatic environment from terrestrial sources (_1_).

The chemical, 2,4-D, can be used in several forms such as esters, acid salts and amines. The only commercial formulation approved in Canada for aquatic weed control is the butoxyethyl ester in a slow release form (AQUA-KLEEN, by Union Carbide Agricultural Chemicals). In the late 1970s, Agriculture Canada had banned the terrestrial use of the butylester form because of drift problems, was reviewing the use of butoxyethyl ester, and was considering the N,N-dimethylamine formulation for specific aquatic uses. This study was designed to determine the fate and impact of the latter two forms in an aquatic ecosystem.

Previous studies have found that 2,4-D does not persist long in the aquatic environment (2). Although a laboratory study reported some effects on phytoplankton (3), field studies generally show no effect on phytoplankton, zooplankton, clams, or fish (4,5). For a perspective we may note that the 96-hour LD_{50} of the 2,4-D acid to bluegill is 350 ppm, whereas the recommended treatment concentration is 1 ppm (6). A laboratory study inferred that the decomposing vegetation after 2,4-D treatment could produce anoxic conditions, presenting a secondary hazard to fish (7).

Ecosystem studies on 2,4-D (4,8,9,10) have investigated several components of the food chain to determine uptake and persistence of the chemical in the biota, and possible toxic effects. Our approach utilized experimental ponds to study the impact of 2,4-D on the components of the ecosystem, and on community structures. The ponds, as closed systems, were considered well suited for the study of the fate and persistence of the chemical. A critique of using such ponds is given elsewhere (11).

This paper summarizes the results of a two-year study (1980/81) and is based on two internal reports which document all findings (12,13).

Experimental

Site Preparation . Six ponds, about 10x20 m each with a depth of 1.5-1.8 m, were excavated in an isolated location near Winona, Ontario, a year before the 2,4-D experiments. The ponds were lined with four layers of 6 mil black polyethylene, with sediment placed over the bottom and the sides. They were filled with water from a nearby pond, in a way that insured the introduction of both planktonic and benthic organisms. Milfoil was planted in the level sediment area of each pond during the fall prior to the treatment year. Shortly thereafter, twenty common shiner (*Notropis cornutus*) were added to each pond.

Pisidiid clams (*Sphaerium rhomboideum*) were collected from a natural pond just prior to the experiment. Ten clams were placed in each of 72 containers with 12 placed in each pond.

Environmental Sample Collection and Analyses. Samples were collected from mobile bridges constructed for this study. A diving platform was attached to each bridge so that a Scuba diver lying on the platform could be moved about the pond for sediment sampling and plant growth measurements without agitating the sediment.

Composite water samples were subsampled for bacteria, water column fungi, phytoplankton, protozoa, water chemistry, and particulate material. Samples were generally collected fortnightly during periods of open water, monthly during the winter, but a more frequent sampling schedule was used immediately before and after the chemical treatments.

On June 25, 1980, two ponds were treated with the N,N-dimethylamine formulation of 2,4-D, and two with the butoxyethyl ester (AQUA-KLEEN). The additions were calculated to produce nominal concentrations of about 1 ppm, the recommended dosage for milfoil control. The two remaining ponds were used as controls. On July 5, 1981, the two former control ponds were treated with the amine and the ester, respectively.

The water chemistry parameters of nitrate, nitrite, ammonia, TKN, particulate nitrogen, filtered, unfiltered and reactive phosphorus, particulate and dissolved organic carbon, alkalinity, calcium, magnesium, chloride and sulphate ions were determined according to the methods of IWD, Environment Canada (14).

Water samples for 2,4-D analysis were collected in 1 L amber bottles containing 2 g each of XAD-2 and XAD-7 ion exchange resin. The samples were acidified with 4 mL concentrated sulphuric acid and stored at $4^{o}C$ in the dark. The 2,4-D was desorbed from the resin with ethyl ether. The extract was dried, reduced in volume, diazotized, and analyzed on a gas chromatograph with an electron capture detector (12,13,15).

Sediment samples were first extracted with 50 mL of 0.1M Na_3PO_4, then the centrifuged and filtered extract was acidified and extracted with ethyl ether. The extract was then treated as those from the water samples and analyzed by GC.

Biological Sample Collection and Analyses. Clam and fish samples were digested in concentrated HCl, then extracted with benzene. The extract was treated as above. Milfoil samples were Soxhlet extracted with a 60:40 benzene-methanol mixture. The extracts was washed with 0.5M NaOH and the aqueous phase was extracted, after acidification, with ethyl ether. The extract was handled in the same way as the water extracts.

Acute toxicity tests were conducted on clams and white sucker fry. Young clams from a permanent pond were used in 120-hr laboratory test to determine the acute toxicity curves for 2,4-D acid at pH 7.9 and 2,4-DCP at pH°s of 6.8 and 8.8. Acute toxicity to white sucker fry was determined by placing several mesh-covered containers of the fry in the experimental ponds daily, for one week, after the 2,4-D additions, and recording the mortalities for each 24-hour period.

Bacteria and fungi were quantified in the 1980 study season, using standard sampling and plating techniques. Phytoplankton samples were collected in both years to determine both populations and community structures. Equally intensive efforts were made to analyze zooplankton (protozoans and mesozooplankton) and zoobenthos (including clams and snails).

Milfoil growth was monitored by measurements of total stem length and carbon dioxide uptake. General observations were made about the makeup of the whole macrophyte communities before and after the milfoil treatments.

Results and Discussion

Both herbicide formulations killed the milfoil in 2 to 3 weeks at the nominal concentration of 1 ppm in the water. Actual concentrations in the water column were significantly below 1 ppm during most of the study period. Attempts were made to recolonize the treated ponds with fresh milfoil 55 and 86 days after the treatments. Residual 2,4-D concentrations above 0.1 ppm killed the new milfoil, confirming reported laboratory results (16). Concentrations just below 0.1 ppm caused excessive sublethal effects (fused leaves). The nominal 1 ppm 2,4-D concentration recommended for milfoil control thus provides a tenfold margin to allow for drift and dilution in natural water bodies.

The main difference between the two formulations was the immediate availability of 2,4-D in the water column from the amine, compared to its slow release from the ester pellets. The spraying of the amine (in pond 2) resulted in aerial drift of the chemical, causing sublethal effects on the milfoil in an adjacent control pond one week later. Analysis of the water showed no 2,4-D in the affected pond, but 0.075 ppm 2,4-DCP was detected for about 1 hour after the drift event. Applying the amine under the water surface (in pond 5) eliminated the drift and resulted in a more immediate distribution of the chemical in the water column.

The 2,4-D concentrations in the water are shown, for the two years of the study, in Figures 1 and 2. The short term data show that 2,4-D was almost immediately available from the amine, but was more slowly released from the ester-containing pellets. After this initial variation, the disappearance of the 2,4-D from the water followed similar patterns.

The 2,4-D remained detectable in the water phase for about four months after application, with the concentrations remaining above the recommended drinking water quality standards of 0.1 ppm for most of this period. The disappearance of 2,4-D from the water column followed first order kinetics, with the calculated half lives of 17 and 35 days in the two years.

Dichlorophenol (2,4-DCP) was present in the system both as an impurity in the formulations and as a degradation product of 2,4-D. Its presence was observed in the water column for most of the study period in 1980. In the following year, probably due to the higher water pH, it was present at lower concentrations and for a shorter period. The 2,4-DCP concentrations were generally about ten times lower than that of the parent acid, and were more variable during the study.

The 2,4-D concentrations in the sediment are shown in Figures 3 and 4. In 1980, the chemical appeared in the sediment of the ester ponds immediately after treatment, but was found in the amine pond sediments only after the milfoil's collapse. The release of 2,4-D from decomposing milfoil appeared to be a significant source. The chemical persisted in the sediment for about as long as it was detected in the water column. The 2,4-DCP was also observed in the sediments, at concentrations about an order of magnitude lower than that of the parent acid, as long as the 2,4-D was detected.

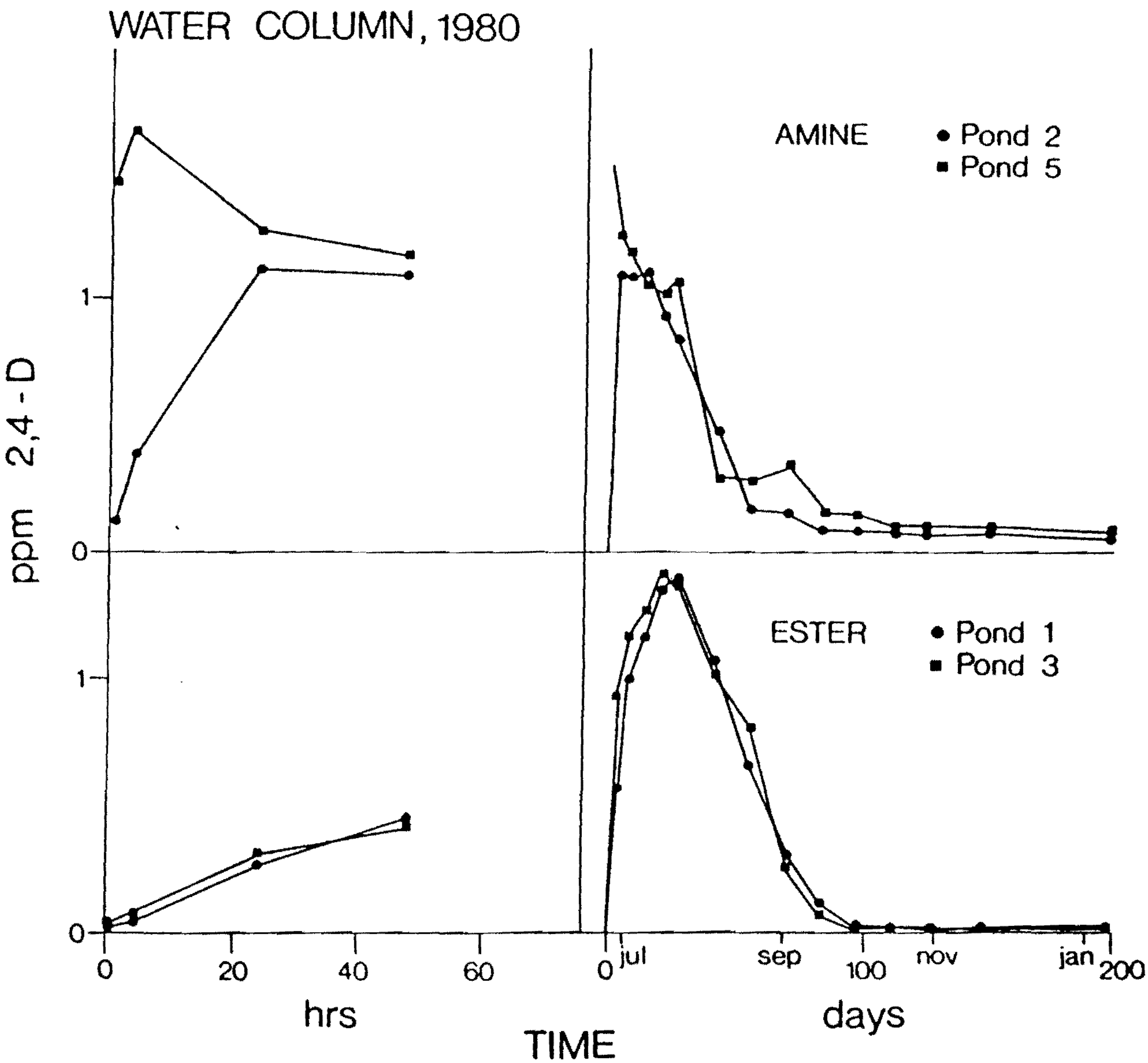

FIGURE 1. 2,4-D concentrations in the water (1980).

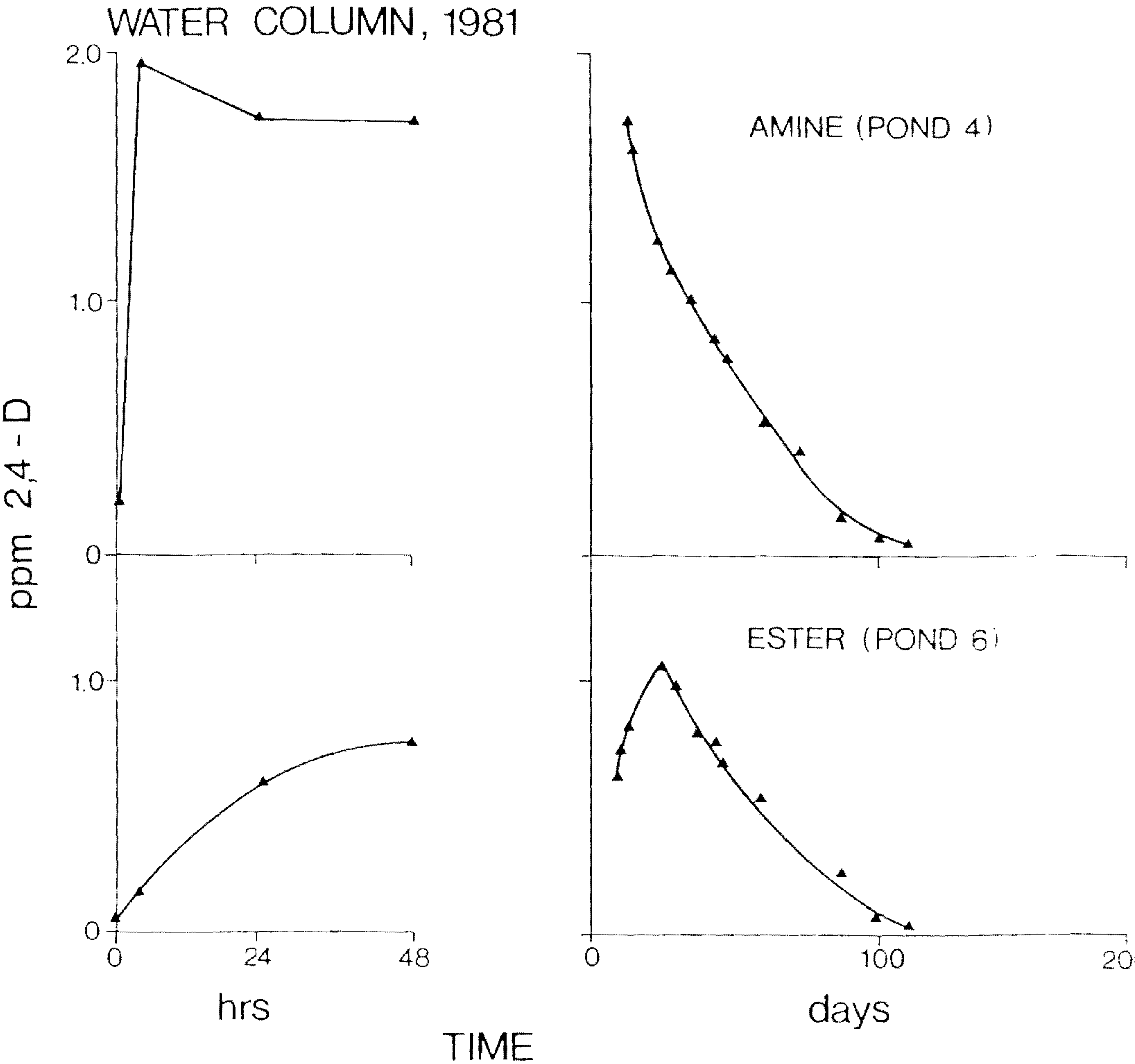

FIGURE 2. 2,4-D concentrations in the water (1981).

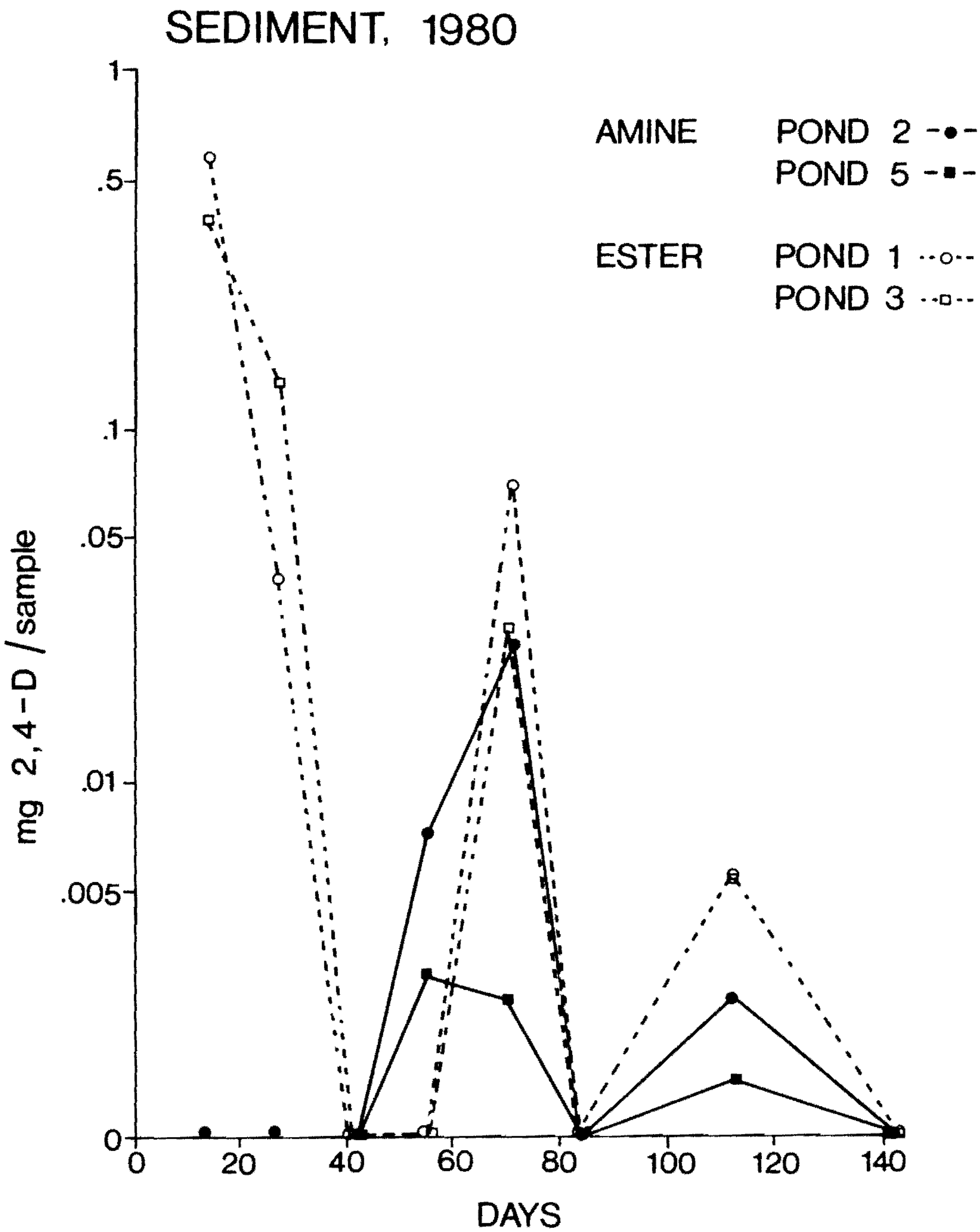

FIGURE 3. 2,4-D in the sediment (1980) (per 10 cm^2 of bottom).

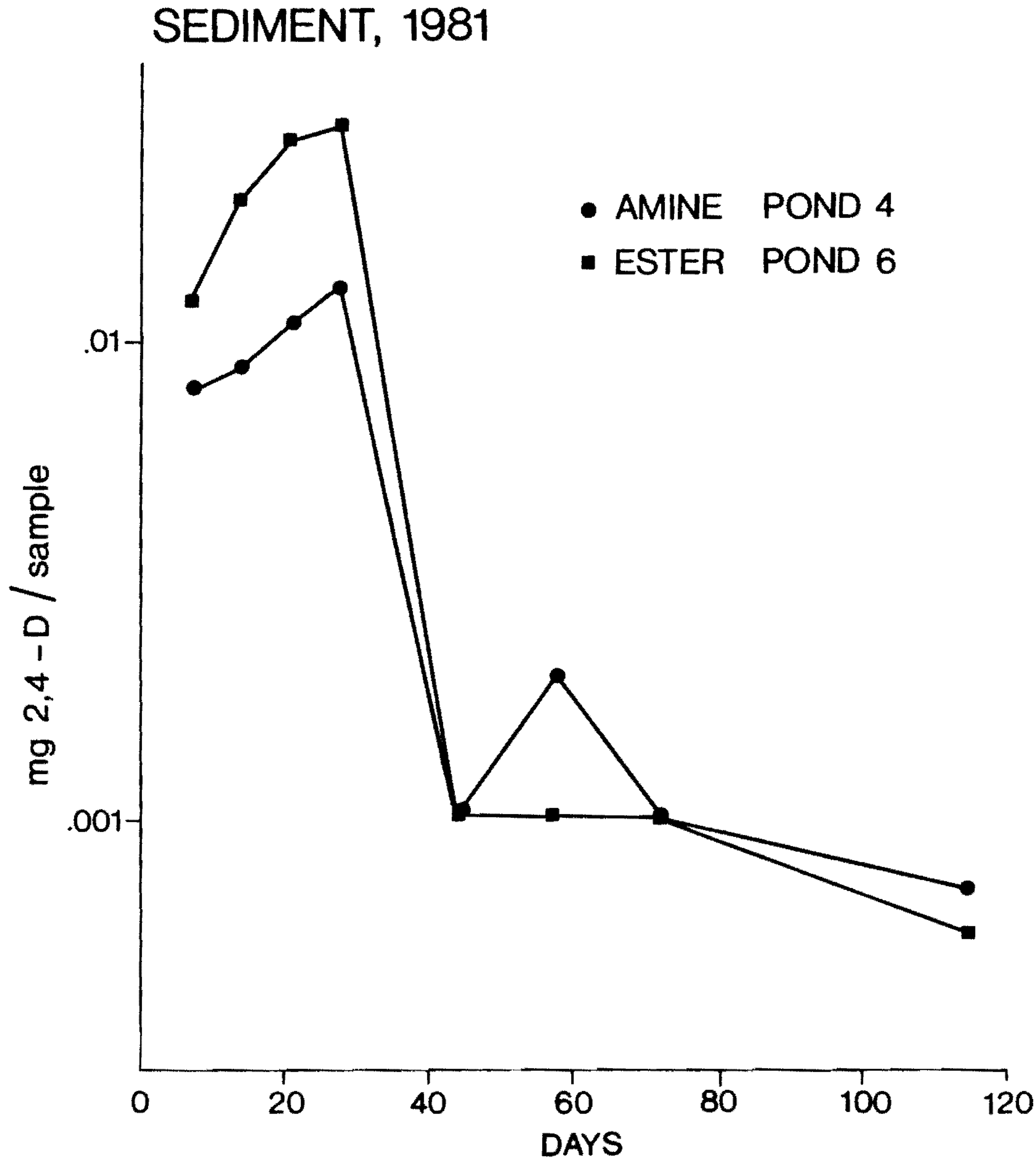

FIGURE 4. 2,4-D in the sediment (1981) (per 10 cm^2 of bottom).

Fish and clam tissues contained some 2,4-D and 2,4-DCP in all treated and control ponds, but no significant trends were observed. Milfoil from the treated ponds contained measurable amounts of 2,4-D, but no 2,4-DCP was detected.

Laboratory toxicity tests showed that 2,4-D was not toxic to clams at a concentration of 250 ppm. The 2,4-DCP, on the other hand, was toxic with a 96 hr LC_{50} of 23.4 and 65.9 ppm at pH 6.8 and pH 8.8, respectively, indicating that the phenol was more toxic than the phenolate ion (17). White sucker fry exhibited limited mortality during the first six days in the treated ponds in the first treatment year. No mortality was observed after six days. On the first day only, the amine ponds produced a higher mortality than the ester ponds (18). The lower water pH in the first year shifted the phenol-phenolate equilibrium for DCP in favour of the more toxic phenol form, resulting in the observed mortalities, in agreement with the laboratory clam studies. Adult common shiners in the ponds were not affected.

The various biological components of the pond ecosystems showed minimal or no response to the treatments. The bacteria populations were unaffected following the treatment, but showed some increases in the treated ponds after the collapse of the milfoil beds. Phytoplankton and zooplankton biomasses and diversity were generally unaffected, although the 1981 samples indicated possible marginal effects on a few of the phytoplankton species (19). The zoobenthos were not directly affected by the treatment. After the collapse of the milfoil, a community shift occurred from chironomid to tubificid dominated communities as a secondary effect of the treatment. The number of snails was observed to increase as a secondary effect, i.e. after the milfoil collapse. The growth of the clams was inhibited in the treated ponds during the first one to three weeks after the applications. Their subsequent growth, on the other hand, was enhanced by the increased food supply. No effects were observed on the reproductive capacity of the clams.

The macrophytes predominant in the control ponds (*Elodea* , *Potamogeton* and *Typha*) were inhibited in the treated ponds, in which *Chara* dominated after the milfoil treatments.

The collapse of the milfoil produced anoxic conditions in the sediment, but did not result in significant oxygen depletion or nutrient enrichment in the water column. Calcium concentrations and alkalinity were observed to increase in 1981, once milfoil decomposition began, probably due to dissolution of calcium carbonate from the milfoil surfaces. Young plants of cattails (*Typha*) did not colonize the shoreline of the treated ponds until the 2,4-D concentrations had declined.

The pond ecosystems used in this study showed remarkably little variation from pond to pond with respect to water quality parameters and most of the biological components. Phytoplankton populations, on the other hand, were extremely variable, masking possible subtle effects on community structures. The year allowed for the stabilization of the ponds appeared to be sufficient for the establishment of indigenous ecosystems.

Summary

Both 2,4-D formulations were shown to be effective in milfoil control at the recommended 1 ppm nominal concentration.

The 2,4-D concentrations in the water were halved in 17 to 35 days, probably dependent on water pH. Concentrations decreased to non-detectable levels in both water and sediment by late fall.

The 2,4-DCP was present both as an impurity in the formulations and a degradation product of 2,4-D. Although its concentrations were generally an order of magnitude lower than that of the parent acid, it was probably responsible for an initial fish fry mortality, an initial inhibition of clam growth, and some sublethal effects on milfoil in an aerial drift event.

The six individual ponds were similar and were "in phase" in the variations of several water quality parameters. Significant water quality changes included increased pH in all ponds during the second year due to increased macrophyte growth, and increased alkalinity and calcium ion concentrations in the treated ponds in the second year arising from the dissolution of calcium carbonate from the decaying milfoil.

The ponds were also similar in terms of most biological variables, but were often "out of phase" with respect to abundances of individual species of phytoplankton.

The treatment showed no measurable effect on bacteria, fungi, phytoplankton and zooplankton, with possible secondary effects on bacteria and phytoplankton. Some fish fry mortality was observed in the first days of the 1980 treatment, probably due to the presence of 2,4-DCP and to low water pH. An initial inhibition of clam growth was also attributed to the presence of 2,4-DCP. The growth of other water weeds was also inhibited and Chara became the dominant macrophyte in the treated ponds.

Secondary effects of the treatment included increased bacteria populations, enhanced growth of clams and snails after the collapse of the milfoil beds, due to an increased food supply, and a shift in the zoobenthic community from chironomid to oligochaete populations.

The pond study allowed a thorough survey of chemical and biological variables in a closed ecosystem. The absence of drift and dilution processes produced a "worst-case" scenario. The observed primary and secondary effects of 2,4-D application would probably be reduced or undetected in more open ecosystems.

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The authors wish to recognize the contributions of all participants in this multidisciplinary study:

J. Hart (NWRI) - pond construction, sampling and analyses;
B.J. Dutka and A. Kwan (NWRI) - bacteria;
J. Sherry (NWRI) - fungi;
M. Dickman (Brock U.) - phytoplankton;

W.D. Taylor (U. of Waterloo) - protozoa;
J. Wood (NWRI) - milfoil;
J. Mackie and M. Stephenson (U. of Guelph) - clams;
A.J. Niimi (GLFRB) - fish toxicity;
M.N. Charlton (NWRI) - diurnal DO study.

One of the authors, B.F. Scott, was responsible for the pond design and overall coordination of the study.

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PANEL DISCUSSION

Moderator: Lars W.J. Anderson

Participants:	A. Leon Bates	David Mitchell
	John Barko	Peter Newroth
	Christopher Cook	David Sutton
	Richard Couch	

Editor's Note: Following formal presentations of papers, a discussion session was held. The panel discussion was moderated by Lars Anderson; panel participants were: Leon Bates, John Barko, Christopher Cook, Richard Couch, David Mitchell, Peter Newroth and David Sutton.

The discussion lasted almost an hour and stimulated comments and questions from the floor. The following is excerpted from transcription of most of the discussion.

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Moderator: In light of problems and confusion of the taxonomy in *Myriophyllum* species, how can this be resolved?

Cook: The biggest problem - and this relates also to obtaining funds for taxonomic research - is that it must be handled *internationally*, but one is usually only funded *nationally*. one needs to break this "border". For example, questions like: should *M. exalbescens* be called *M. sibiricum* cannot be answered until you start looking "worldwide". I think a lot of "species" are based on "patriotism" rather than "patristics". The problem is that one can get money to work on plants in one's *own* country, but not for plants in other countries.

Moderator: What are the implications of being able to break down these barriers?

Cook: If you look at work that's being done - a Dutchman has just done work on species in Southeast Asia, a Tasmanian has worked on South America and North Australia - but this is not a world perspective. Then there's the approach- where can one grow all these species? They must be cultivated to look at them properly.

Moderator: That brings up the question of environmental influences on morphology.

Couch: Within the U.S., we've found Susan Aiken's papers very helpful in separating species with perhaps 90% accuracy. You have to keep in mind that there are "lumpers" and "splitters"; Dr. Aiken is a "splitter"; for example, there are some groups that could be put together, such as *hippuroides*, *heterophyllum*, *M. ussuriense*. As Dr. Aiken points out, you've got to grow them out. I agree with Dr. Cook that you cannot use a hit and miss approach - you need a separate study. There are also problems with historical records. We found that maybe 20% of the herbarium specimens we examined (that Dr. Aiken had not) were misidentified. Also, there are many more older collections than recent ones. In the past 20 years, few aquatic plant collections had been made.

Moderator: Does chemical taxonomy offer any help in confirming relationships? For example, secondary compounds on isozyme patterns have received attention.

Cook: The answer is yes - but as an adjunct to other methods. You've got to grow and cross these plants and deduce something of their relationships. You won't get this information from small molecules - you might from large molecules. but the taxonomy has got to serve the people - something that a lot of taxonomists tend to forget. You need a system where species are identifiable without a computer, but below the species level. (varieties, races, etc.), you can use thin layer chromatography, electrophoresis, or whatever you wish, but I think you need two levels: big species (broad) and lots of varieties (specific). For example, that's what you've got in Hydrilla at the moment - thank God nobody has decided to call the monoecious one (in the U.S.) a new species - that would be crazy. So we have Hydrilla verticillata with two 'races" in the U.S. and I've got at least 5 in cultivation in Switzerland now (and I hope they stay there as well!).

Barko: As an example of confused taxonomy (or misidentification), people working on Lake Wingra during the IBP considered the dominant plant to be M. exalbescens and a problem even though it is native. Well it was later correctly identified as M. spicatum, yet the problem didn't increase at all, so is there really a distinction between these two species (or perhaps others) in terms of the problem they create.

Cook: Clearly, yes. M. exalbescens is no problem here or in Europe; M. spicatum is no problem in Europe, but it is a problem here.

Barko: Is it due to more rapid spread or different growth form in M. spicatum?

Cook: My guess is that its due in part to a "release" where a species is transferred to a new geographic location., The M. spicatum here (North America) is an old world species (or race) and it is relieved of burden or stresses (e.g. microorganisms, fungae, specific competitors).

Couch: I would agree, and mention that Barry Helquist - a taxonomist in the Massachusetts area, tells me that M. heterophyllum is the problem species in that area, not M. spicatum.

Mitchell: You're really asking what makes a species weedy and I don't think there are simple answers to that. In Australia, there are a couple of species with weedy potential, e.g. *M. salsugineum* and *M. verrucosum* that respond to disturbance, grow rapidly and prolifically, even in their "own" environment. if these species got to another country - in the right conditions -they would be dangerous. on the other hand there are many probably "good" species - perhaps 36 - most of which have a very small distribution. For example, *M. tillaeoides* has been recognized as a "good" species and known from half a dozen localities is unlikely to become a weedy species. It's one of the so-called semi-terrestrial forms. But, if a plant shows the potential of being a weed in its own (native) environment, it should be treated with caution, if it gets out anywhere else.

Cook: Another point is that even a slight change in the conditions can shift species dominance. A case is in the River Rhine (Switzerland) where there has always been *Ranunculus fluitans* but as soon as phosphate levels go up, this plant becomes a problem.

Newroth: We previously discussed "practical" taxonomy, and in 1974 here in B.C., we had a very practical need since it was clear there were a number of species of *Myriophyllum* but we didn't know which ones. We looked for a "fingerpointing" method such as that presented by Adolf Ceska. We found it useful for us. More research is needed in this area. This kind of information is needed so that management decisions, which may involve $100,000, can be made correctly and quickly.

Moderator: Would it be practical and of value to put together an international "collective" through which funding and research could be facilitated - perhaps a 3-5 year program to straighten-out taxonomy?

Cook: It's been done once - but it never really worked. A panel of experts was formed during the International Hydrobiological Decade to address these problems and it went for a couple of years - then funding ran out. But it's possible.

Painter: I cultivated *M. hippuroides* and *M. heterophyllum* under similar conditions and found that *hippuroides* is much more aggressive than *heterophyllum*. In the early 1970's we in Ontario had *M. exalbescens* and *M. spicatum* growing side by side in the field and had no problems distinguishing them. I would like the panel to comment on the "natural declines" in populations: can we explain these and conduct research on this phenomena so as to learn how to encourage it where we would like?

Barko: These declines are not unique to *M. spicatum*. It has occurred in Europe with *Elodea canadensis*, which was exported from the U.S. It's also occurring in some parts of Florida with *Hydrilla verticillata*. The best information is on *M. spicatum* and Susan Bailey and John Andrews at Wisconsin suggest it is related to a kind of "wasting" disease reported for seagrass throughout the world. Other theories invoke gradual eutrophication and attendant increases in phytoplankton and attached algae which can reduce light availability to macrophytes, coupled with a negative feed-back on reproduction. (See a recent paper by Robert Twilly in Marine Biology). So you have light attenuation, diseases, reproductive effects, possibly allelopathathic interactions. Our own work suggests the change in the composition of sediments due to the plants themselves may also cause a stress. Brian Moss (Great Britain) has reports that in addition to increased nutrient loadings, there are changes in zooplankton behavior and in fish-use of plant beds, changes in boating use and related effects on sedimentation. But it's probably a combination of all these variables which are part of successional changes -either natural or caused by man's activities. I think we need to emphasize, in our research, the redevelopment of these areas by employing plants with more desirable characteristics.

Bates: We've had *M. spicatum* in the TVA for the past 25 years, but the total has decreased in the past few years to 14,000 -16,000 acres. However, the total for all species is still around 22,000 acres so there has been some "niche-snitching" by other species as the milfoil declined, such as spiny leaf naiad. So this is similar to what John (Barko) has discussed.

Moderator: It seems that this points to a need for more work on ecological questions that we have about aquatic macrophytes in general.

On another topic, I'd like some discussion on the extent of milfoil infestations - what is the area involved?

Newroth: For B.C., based on surveys done around 3 years ago, we have around 1,00 ha, however, the potential is much greater because many of the important lake systems have not got Eurasian watermilfoil. So for B.C., potentially we have 2,000 -3,000 ha at most. For the rest of Canada, perhaps Scott Painter can provide some estimates.

Painter: There have been no studies to document rigorously the acreage - just from personal experience, I would say that for Ontario alone there may be about 60,000 acres now. We have documented most of the St. Lawrence to Quebec City and for both Ontario and Quebec probably there are $100,000 acres severely infested.

Couch: For the U.S., if you take our estimate of 392 population in 1985 and add perhaps 50 to that, then multiply by an average per acre per site, you could arrive at an estimate. So if you use a 500-acre average, you would get a reasonable estimate (221,100 acres).

Bates: One of the "unknowns" here are the private lakes and ponds - that's a gap we don't really know about. TVA has ca. 16,000 acres right now.

Moderator: Regarding control of *Myriophyllum*, what is the Panel's feeling on the potential for biological control. It appears that with milfoil there hasn't been a tremendous success with insects or pathogens, though perhaps grass carp represents a "general" control agent.

Sutton: Where you have only Eurasian watermilfoil, the grass carp will probably eat it, even though its (*M. spicatum*) low on the grass carp's preference. But in large areas, it may be difficult to use since the fish may not feed in the areas where plants need to be removed such as beaches or swimming areas. In small farm ponds certainly it could be used.

Moderator: A question from the floor was raised on the potential rototilling as practised in B.C. Peter, would you care to comment?

Newroth: Our rototilling is quite selective - aimed at high-use areas where leaving a stubble is not acceptable to beach users. One advantage is that it can be used in the fall or early spring before the tourists arrive. For us, it facilitates a continuity in the program. But of course, it impacts the sediment and we have not investigated that in detail to see how many worms are killed or what kind of changes there are in the organic material. Perhaps we're really resuspending a lot of fine particles and organic material, which drift away and into deep parts of the lake. We may be making a more mineralized, less organic sediment and even encouraging milfoil by rototilling. I'd like to see more research on this. For now, I don't see a major expansion of rototilling in our program. We're only treating ca. 200 ha. in the Okanogan with all mechanical methods and perhaps tillage would increase to 70-80 ha, to 100 ha by next year. The method is too slow for 100's of ha at once. But shallow tillage as part of shore-base equipment could cover ca. 1.5 -2 ha. per day, and would be practical on large areas especially where drawdown is feasible such as the TVA. There are limitations such as unsuitable substrate, pipelines, cables, powerline cables or other obstacles.

Barko: The points that Peter raised on rototilling further emphasize the need for a broader, ecological perspective in aquatic plant management. We too often examine individual plant responses and ignore the fact that these species are acting in -and reacting to - the whole system. Disturbance like rototilling or dredging are really major changes in the plant's environment and we need to learn how the system changes and what it means for the success or failure of various species.

Moderator: I wish we could continue but our time is gone. I would like to thank all the participants of the Symposium and in particular the invited speakers and Panel participants.

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THE AQUATIC PLANT MANAGEMENT SOCIETY, Incorporated
POST OFFICE BOX 16
VICKSBURG, MISSISSIPPI 39180