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Control of Aquatic Weeds with Hexazinone[1]

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ABSTRACT

The triazine herbicide 3-cyclohexyl-6-(dimethylamino)-1-methyl-1,3,5,triazine-2,4(1*H*, 3*H*)-dione (hexazinone) was tested for its ability to control aquatic weeds when applied at 1.0 ppmw in a 0.08-ha pond. Treatment in June completely controlled all of the following indigenous species: American pondweed (*Potamogeton nodosus* Poir.), Sago pondweed (*P. pectinatus* L.), elodea (*Elodea canadensis* Michx.), cattail (*Typha* sp.), *Spirogyra* sp., slender spikerush (*Eleocharis acicularis* (L.) Roem. & Schult.), and *Chara* sp. Dissolved oxygen declined from 8.0 ppm to 0.2 ppm within 5 days after treatment and appeared to be the cause of fish mortality observed 4 days post-treatment. Evidence for root to shoot translocation was observed in cattails and Russian olive trees (*Eleaegnus augustifolia* (L.)). A bioassay for the presence of hexazinone in water and hydrosoil revealed no phytotoxic residues 15 months after treatment. Species diversity of non-target organisms (invertebrates and periphyton) was greatly reduced in the treated pond, probably due to the loss of suitable habitat.

INTRODUCTION

Greenhouse screening tests have indicated that the triazine herbicide 3-cyclo-6-(dimethylamino)-1-methyl-1,3,5- triazine-2,4(1*H*, 3*H*)-dione (Hexazinone) may be effective against submersed macrophytes (1, 3). Under conditions of an experimental use permit issued by the Environmental Protection Agency (352-EUP-94), hexazinone was field-tested for efficacy in controlling several indigenous species of aquatic weeds in a small (0.08-ha) pond. Data were also obtained on the dissipation of hexazinone residues in water and soil, on water quality, and on nontarget species diversity.

METHODS AND MATERIALS

Two square 0.08-ha ponds, lined with butyl rubber and earth, were used for treatment and control (untreated) ponds. Each pond had a mean depth of 1.22 m, and contained the following species in similar relative abundance: cattail, elodea, American pondweed, sago pondweed, chara and *Spirogyra* spp. Ponds were maintained at 1.22 m depth by periodic filling from a municipal source. Pre-treatment plant abundance was determined as % composition of each species within each grid.

An aqueous solution of the 90% soluble powder hexazinone was applied through a 1.22-m sprayboom held 0.3 m below the surface of the water. 0.97 kg. hexazinone was applied to produce a calculated concentration of 1.0 ppm in the treated pond.

Two methods for assessing the herbicidal activity of hexazinone were used: (A) visual rating of damage to indigenous plants after treatment, and (B) visual rating of introduced flats containing *P. nodosus* and *P. pectinatus*. The introduced plants were germinated from winterbuds in 0.3 by 0.3 by 0.15 m wood flats containing soil. Flats were maintained submersed in 208 liter barrels which had been cut in half, in a greenhouse for 2 weeks following planting of winterbuds. One flat of each species was labelled and placed in each of four randomly selected 9 by 9 m grids in the control and treated ponds. Hexazinone was applied 7 days after placement of flats. The flats were observed at 2 week intervals and photographed at the end of the 100-day observation period and the number of winterbuds produced in each flat was determined.

For visual observations, a code system was used to describe the condition of each species (see Table 1). This system was used to record the symptomatic responses of the various weeds to the herbicide.

Water Quality Measurements

A portable instrument was used to make bi-weekly analyses of pH, dissolved oxygen, temperature, and conductivity (Hydrolab Surveyor 6D). These analyses were made between 12 noon and 2 p.m., and no more than 30 minutes elapsed between measurements in the control and treated ponds. Biweekly water samples were taken in duplicate for determination of hardness, alkalinity, nitrate and phosphate. Nitrate and phosphate levels were determined with an autoanalyzer (Technicon Auto Analyzer 4). Limits of detection were (mg/1): total P (.001); NO_2 (.001); NH_4 (.01); NO_3 (.004). At the time of treatment, the conditions were as follows in the treated pond: pH 7.8; dissolved oxygen, 8 ppm; total hardness, 90 mg/1; temperature 21.5 C. Conditions in the control pond were: pH, 8.6; dissolved oxygen, 9.6; total hardness, 82 mg/1; temperature 21.5 C.

The effect of the hexazinone treatment on non-target organisms was assessed biweekly. Artificial invertebrate sub-

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TABLE 1. EFFECTS OF TREATMENT WITH 1.0 PPMW HEXAZINONE ON AQUATIC WEEDS.

Species	Pretreatment Composition 0 Days Mean Percent Coverage (aver. of 4 grids)		Post-treatment Condition[1, 2]									
			7 Days (6/30/77)		14 Days		28 Days		45 Days		100 Days	
	CONT.	TRT.	CONT.	TRT.	CONT.	TRT.	CONT.	TRT	CONT.	TRT.	CONT.	TRT.
Cattail	18	45	NC	NC	NC	BRN	NC	BRN DEC	NC	DEC	NC	NC
American pondweed	2	2	NC	DEC	FL	DEC	FL	DEAD	FLO	NC	FLO	NC
Sago pondweed	21	9	NC	BRN, DEC	GRO	DEC	GRO	DEAD	FLO	NC	FLO	NC
Chara	38	9	NC	PUR, FLT	NC	DEC	NC	DEAD	NC	NC	NC	NC
Elodea	0	9	—	NC	—	FLT, DEC	—	DEC	—	NC	—	NC
Filamentous algae (*Spirogyra* sp.)	7	<1	NC	BRN	NC	DEC	GRO	DEAD	GRO	NC	NC	NC
Needle spikerush	0	<1	—	BRN	—	DEC	—	DEAD	—	NC	—	NC

[1]Effects on individual species did not differ among grids.
[2]Codes for condition of weeds:

NC — no change from previous observation
PUR — purple coating present (probably bacteria)
BRN — browning/burning of foliage
DEC — foliage decomposing
GRO — biomass increased
DEAD — vegetation dead
FLT — vegetation now floating, detached
FLO — flowers formed
FL — floating leaves formed

strates (wire baskets containing redwood bark) were placed at three sites within each pond. Invertebrates were collected from the bark chips and preserved in 10% formaldehyde. Periphyton was sampled from paired glass microscope slides held on racks (18 by 18 cm) at three sites within each pond. Ten fields of view from each slide were observed and the number of each species determined. For both the invertebrates and periphyton, a "species diversity index" (SDI) was calculated according to the method of Shannon (14) as described by Lloyd (12).

Samples of hydrosoil and water were taken from the hexazinone-treated pond 15 months after treatment for a bioassay to detect phytotoxic residues. Winterbuds of *P. nodosus* and *P. pectinatus* were exposed to the following combinations: hexazinone-treated hydrosoil plus tap water, hexazinone-treated water plus potting soil, and potting soil plus tap water. Two replicates (five winterbuds each) for each exposure were planted in clay pots held in 19-liter glass jars in a greenhouse and observed for 4 weeks.

RESULTS AND DISCUSSION

Descriptive observations on indigenous plants are summarized in Table 1. The two species of pondweeds, followed by *Chara*, showed the earliest symptoms of phytotoxicity. Within 14 days of treatment these species were decomposing, though portions of the plants were intact. A massive and nearly complete covering of what appeared to be purple bacteria developed on *Chara* within 7 days of treatment and persisted for about 14 days.

Within 14 days, chlorosis on cattails was observed on the emerged foliar portions which had received some direct spray when the spray boom came out of the water (Figure 1a). Further phytotoxicity was evident in all stands, including those on the shore, within 28 days after treatment. In addition, two Russian olive trees (*Elaeagnus augustifolia* L.) showed similar symptoms. This indicated that hexazinone was readily translocated via rhizomes and roots to the foliar portions of these plants. The Russian olive trees were dead by the time of the final observation, 100 days post-treatment (Figure 1b).

Elodea canadensis exhibited the greatest tolerance to hexazinone (Table 1). Some portions of the plants remained green until the 45-day observation, and decomposition continued during the interval between 45 and 100 days. However, complete control was obtained within 3 months after treatment.

No traces of plant material remained in any of the introduced flats in the treated pond 4 months after treatment. During the same interval, the pondweeds in the flats from the control pond grew and flowered normally. No winterbuds were present in any flats from the treated pond. Flats for the control pond contained a mean (±S.E.) of 185 (± 21) American pondweed winterbuds and 176 (± 17) sago pondweed winterbuds. After a 2-month vernalization, winterbuds of each species from the control pond germinated and developed into normal plants.

Six days after hexazinone treatment, the oxygen concentration in the treated pond dropped from a pre-treatment level of 8.0 ppm to 0.2 ppm (Figure 2). This precipitous reduction was no doubt responsible for a fish kill observed 4 days after treatment. The fish killed were rainbow trout (*Salmo gairdnerii*) and suckers (*Catostomus commersoni*). The oxygen level remained extremely low

Figure 1a. Appearance of *Typha* sp. following application of hexazinone to produce a concentration of 1.0 ppmw in pond water at 14 days post-treatment. Note burning of terminal shoots.

throughout the observation period (4 months). No dead fish were observed in the control pond.

The pre-treatment levels of total phosphorus (Figure 3) were similar for the control and treated ponds. However, by the first post-treatment sampling time (6/29), total P reached nearly 0.05 mg/l in the treated pond while the level of P dropped to 0.015 in the control pond, and remained at about this level throughout the summer and fall. Total P varied considerably in the treated pond and stayed at least twice that of the control pond until 8/29. Orthophosphate concentrations were also determined but not detected in the control pond. Ortho-P ranged from 0.01-.03 mg/l in the treated pond. Since the level of P increased only in the treated pond it is probable that this resulted from decomposition of affected plants and release of phosphorus previously bound in intact plant tissue. The P released subsequently became bound and levels in the treated pond were similar to that of the control pond 2 months after treatment.

Ammonia nitrogen (Figure 4) exhibited the greatest increase following hexazinone treatment. This is not surprising since by that time most macrophytes were decomposing (Table 1). Ammonia-nitrogen (Figure 4) remained low in the control pond (less than 0.1 mg/l) throughout the post-treatment period. Nitrite-nitrogen levels exceeded detection levels (.001 mg/l) only on three sampling times. Pre-treatment level of NO_2 in the control pond was .002 and .013 in the "treatment" pond. Twenty-three days post-treatment, the control pond had .003 mg/l NO_2. In the last samples, 118 days post-treatment, the treated pond had .025 mg/l NO_2. Nitrate-nitrogen (Figure 5) was higher in the "treatment" pond before application of hexazinone (6/22), but declined below detectable level six days post-treatment. Nitrate gradually increased in the treated pond during the following three months while in the control pond it remained below detection except on 7/7 and 8/29.

The observed increase in nitrogen and phosphorus in the treated pond is consistent with other reports on the decomposition of aquatic macrophytes. Jewell (10) measured N and P levels in various submersed macrophytes as they decayed and found "regeneration rates" (release rates) of up to 4.9% and 5.8% per day, respectively. Release of N and P was most rapid during the first 10 to 20 days. Godshalk and Wetzel (7) observed high rates of release of dissolved organic material (DOM) from *Myriophyllum heterophyllum* Michx., and *Najas flexilis* (Willd.) Rosk. and Schmidt,

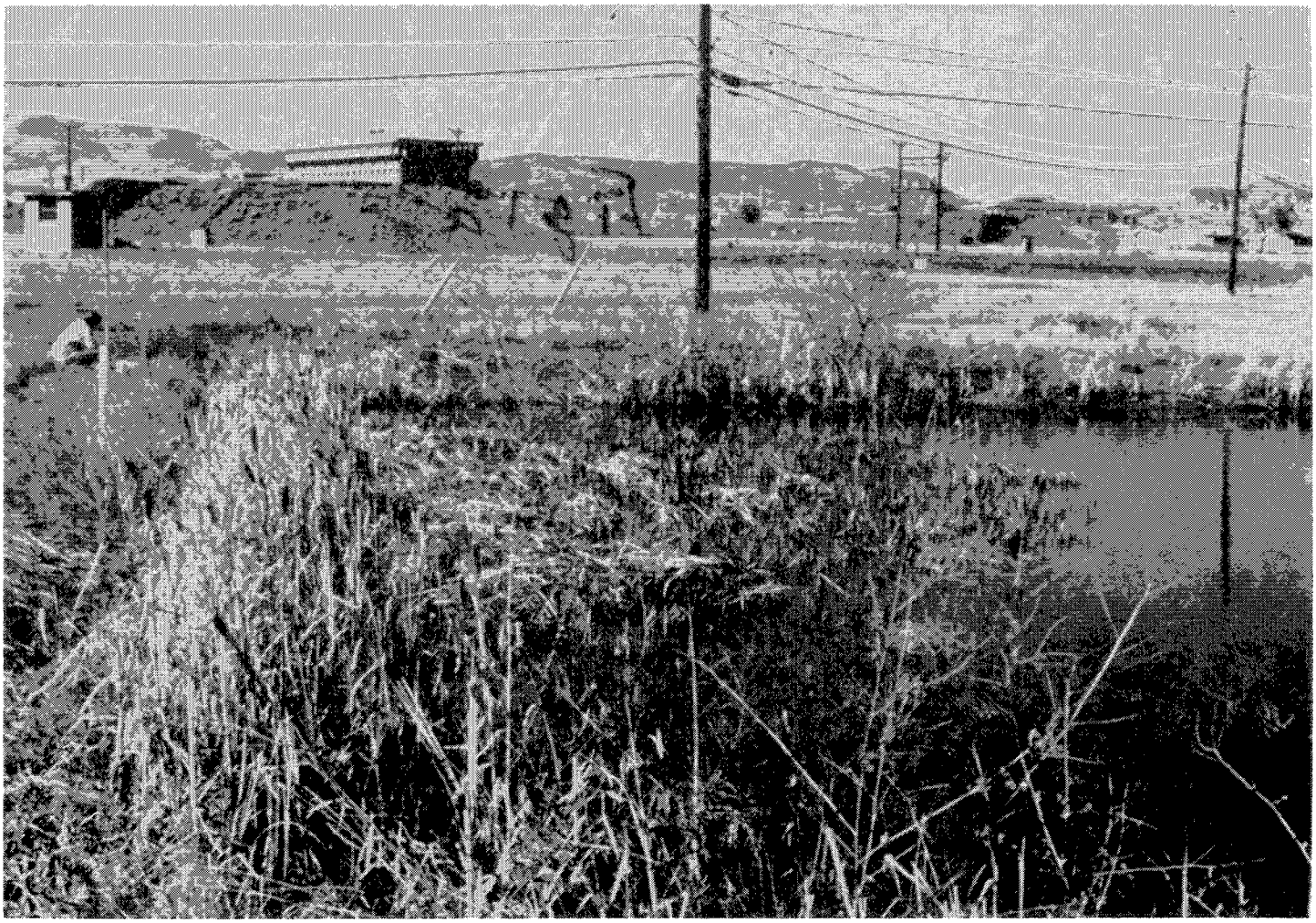

Figure 1b. At 100 days post-treatment. Note defoliated Russian olive tree on the far bank.

under anaerobic conditions at 25 C. In the present study, by the fourth day after treatment with hexazinone, conditions were nearly anaerobic and some plant decomposition was already in progress. More recently, Hill (9) described the release of nutrients from pondweed species growing in a drainage ditch. He found that detached plants lost nearly 100% of their N and P within 50 to 64 days. Most rapid loss occurred within 30 to 45 days after detachment.

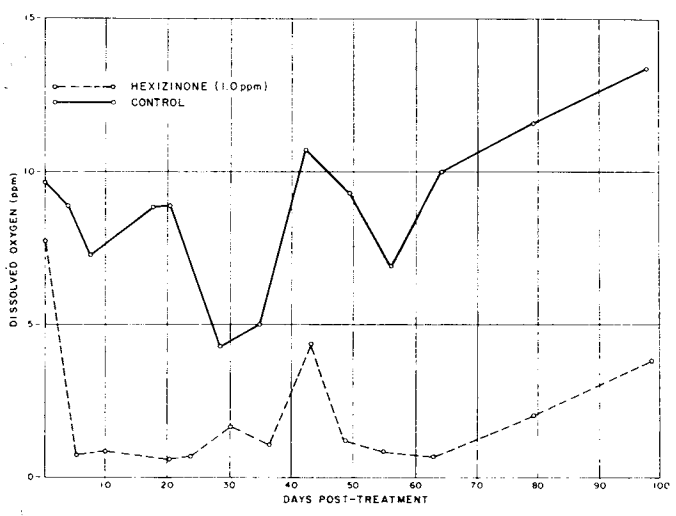

Figure 2. Dissolved oxygen in control pond and pond treated with hexazinone at 1.0 ppmw.

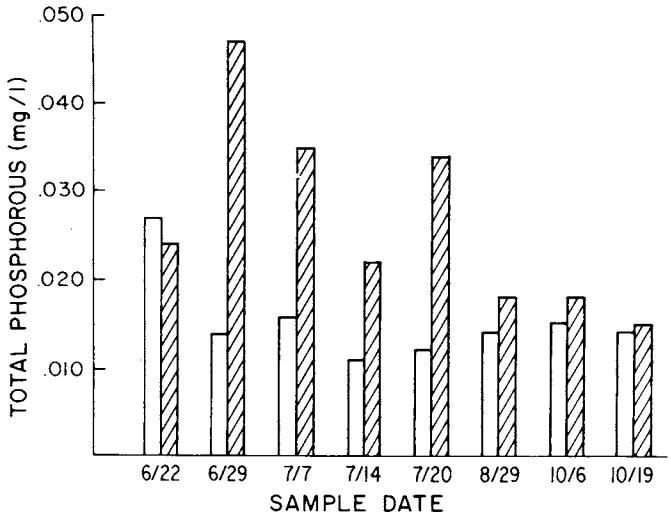

Figure 3. Total phosphorus concentration in the water of the control (open bars) and pond treated with 1.0 ppmw hexazinone (cross-hatched bars). Data are means of duplicate samples taken 25 cm below the surface of the water.

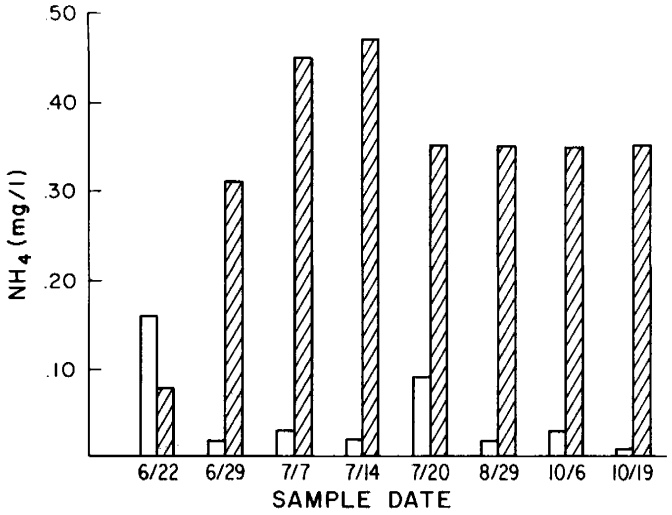

Figure 4. Ammonium concentration in control (open bars) and a treated pond (cross-hatched bars).

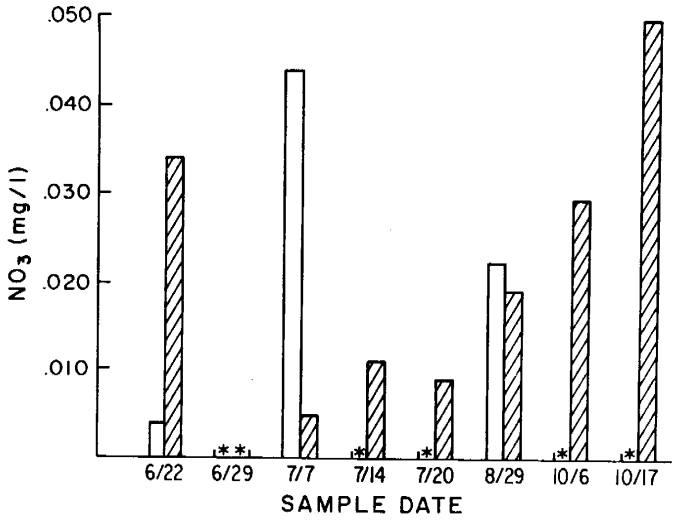

Figure 5. Nitrate nitrogen levels in control (open bars) and a pond treated with 1.0 ppmw hexazinone (cross-hatched bars). Data are means of duplicate samples taken 25 cm below the surface of the water. Asterisks indicate levels below detection limits (0.004 ppm).

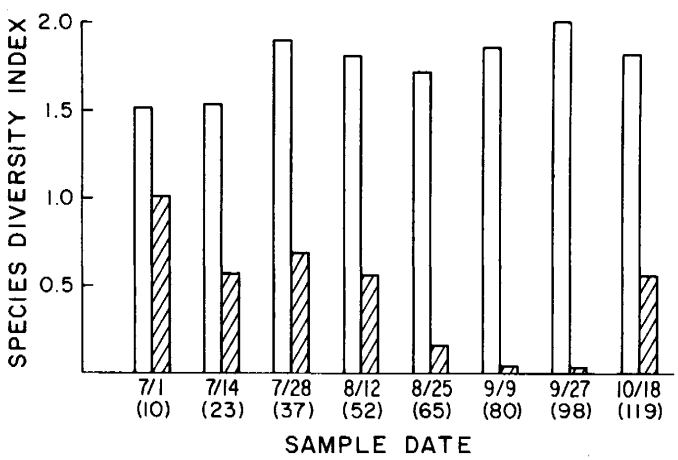

Figure 6. Species diversity index (SDI) for periphyton attached to artificial substrates in a control pond (open bars) and in a pond treated with 1.0 ppmw hexazinone (cross-hatched bars). Numbers in parentheses are days post-treatment. Values are means of three replicates within each pond.

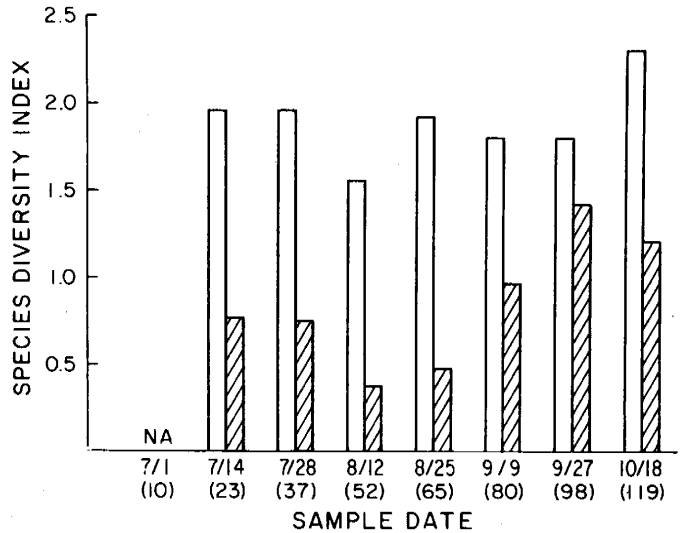

Figure 7. Species diversity index (SDI) for macroinvertebrates attached to artificial substrates in a control pond (open bars) and in a pond treated with 1.0 ppmw hexazinone (cross-hatched bars). Numbers in parentheses are post-treatment. Values are means of three replicates within each pond.

Results of the species diversity analyses are presented in Figures 6 and 7. Sampling for periphyton growth was initiated 10 days post-treatment, while the first macroinvertebrate samples from artificial substrates were taken 23 days post-treatment. The diversity of periphyton in the control pond (Figure 6) and macroinvertebrates (Figure 7) remained relatively high and stable throughout the 3½-month observation period. At 23 days post-treatment, macroinvertebrate diversity in the treated pond was less than half that of the control pond, and remained low through the 65-day sample. There was an increase in diversity in the treated pond 80 and 98 days post-treatment, but the SDI remained lower than that of the control pond.

The SDI of periphyton in the treated pond fell from 10 days post-treatment (1.0) through the 98-day sample (0.02). The populations on the slides in the two ponds consisted mainly of diatoms and a few species of filamentous green algae. Since hexazinone is a potent inhibitor of photosynthesis, it is not surprising that the numbers of species (as well as total numbers of algae) decreased so dramatically following treatment. One hundred nineteen days post-treatment some recovery of the epiphyte population occurred as the SDI increased to over 0.5.

The immediate decline in diversity of macroinvertebrate was probably caused by the precipitous reduction in dissolved oxygen, which could have led to mortality or emigration from bark substrates to more aerated soil-water-air interfaces along the shorelines. Diversity of macroinvertebrates continued to decline and this might be expected as the habitat and food sources deteriorated with destruction of macrophytes and epiphytes. Partial recovery of macroinvertebrate diversity was evident after the 65-day sample time. Since the sampling method for macroinvertebrates relied upon colonization of bark substrate, these data are not necessarily indicative of species diversity in other benthic habitats such as the hydrosoil or rhizomes of macrophytes.

However, the constancy of SDI in the control pond compared to the hexazinone-treated pond suggests that the method can detect perturbations in the benthic populations.

No phytotoxicity was evident in either species of pondweed grown in soil or water taken from the hexazinone-treated pond 15 months after treatment. All plants germinated and produced normal foliage and secondary plants.

The results of this small pond test clearly indicate that hexazinone at 1.0 ppmw effectively controls a variety of aquatic weeds, including algae. It is likely that lower concentrations might also be effective, but may require a longer time for complete control. There was evidence for root-to-shoot translocation and such translocation seems reasonable in that acropetal movement of other triazine herbicides has been demonstrated in terrestrial and aquatic plants (4, 5, 7, 15, 16). Root to shoot translocation is a desirable characteristic for herbicides for control of submersed aquatic weeds, but caution should be exercised because of the possibility of damage to near-shore non-target plants (e.g. trees, shrubs, turf) if their roots reach the hydrosoil.

The observed precipitous decline in dissolved oxygen is not surprising since the extensive weed biomass treated would support decomposition and since it is well-known that triazine herbicides are potent inhibitors of photosynthesis (2, 17, 18). A similar reduction in dissolved oxygen during the first five days following hexazinone treatment of *Myriophyllum verticillatum* was reported by Fowler (3). Under normal circumstances, use of a lower concentration of hexazinone and partial treatment probably could alleviate some of the adverse effects on dissolved oxygen. In this regard, a pelleted or granular formulation might be useful in localizing the treatment.

Observation of the pond one year after treatment with hexazinone revealed no submersed aquatic macrophyte regrowth and no regrowth of of cattails. This finding indicates that hexazinone completely killed the rhizomatous portions of the plants. The only regrowth observable after 12 months was a phytoplankton population and an algal layer on the hydrosoil. If hexazinone has soil persistence characteristics similar to those of other triazines, it is not surprising that weed control continued for at least one year. Simazine and atrazine persist for 10 to 12 months after terrestrial application (11), and hexazinone reportedly lasts 8 to 10 months (6). No data could be located on the persistence of hexazinone in hydrosoil. Recently, Rhodes (13) reported that hexazinone was stable in water up to four weeks (the limit of his sampling) and that bluegill (*Lepomis macrochirus*) did not exhibit abnormal behaviour or mortality when exposed to 1.0 ppm hexazinone. However, the results of the bioassay indicate that a phytotoxic residue did not persist in water or hydrosoil 15 months after treatment.

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