ABSTRACT

Kurt D. Getsinger. CHANGES IN A *Myriophyllum spicatum* L. COMMUNITY FOLLOWING 2,4-D TREATMENT. (Under the direction of Graham J. Davis) Department of Biology, July 1976.

On 13 and 14 July, 1974, approximately 350 ha of Myriophyllum spicatum (Eurasian watermilfoil) in the Kitty Hawk Bay - southern Currituck Sound area of North Carolina, were treated with a granular 20% acid equivalent formulation of the butoxyethanol ester of 2,4-dichlorophenoxyacetic acid (2,4-D) at the rate of 112 kg/ha (100 lbs/acre). Native submersed macrophytes associated with the M. spicatum stands included Ruppia maritima, Najas guadalupensis and Potamogeton pectinatus. The M. spicatum communities thrived under conditions of low ambient ortho-phosphate concentrations. Pre- and post-treatment nutrient analyses generally indicated low phosphorus and nitrogen in plant tissues. Myriophyllum spicatum was eliminated and native species density and biomass decreased six weeks after treatment. Water turbidity increased during the post-treatment period, accompanied by a cyanophytoplankton bloom. Deterioration of M. spicatum in isolated coves near the treatment area continued to occur eight weeks after the initial treatment. Accelerated growth and re-establishment of native submersed macrophytes did not occur during the 1974 growing season.

Myriophyllum spicatum and native submersed macrophytes recovered slowly during the one year, post-treatment growing season. A successional pattern in the Kitty Hawk Bay treatment area appeared to develop with the charophyte Nitella hyalina as the "pioneer" species. Observa-

tions at the end of the 1975 study pointed to the re-establishment of a vascular submersed macrophyte community dominated by *M. spicatum*.

CHANGES IN A
Myriophyllum spicatum L. COMMUNITY
FOLLOWING 2,4-D TREATMENT

A Thesis
Presented to
the Faculty of the Department of Biology
East Carolina University

In Partial Fulfillment
of the Requirements for the Degree
Master of Science in Biology

by
Kurt D. Getsinger
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TABLE OF CONTENTS

LIST OF FIGURES

LIST OF TABLES

INTRODUCTION

This study follows the effect of 2,4-dichlorophenoxyacetic acid (2,4-D) on the density, biomass and nutrient concentrations in a submersed macrophyte community which was dominated by *Myriophyllum spicatum* L. (Eurasian watermilfoil). Historically, *M. spicatum* has caused problems in waterways of North America, particularly those in the eastern United States. According to Blackburn and Weldon (1966), the plant was first reported in the Chesapeake Bay area in the late nineteenth century. During the past 80 years *M. spicatum* has become established in the lakes of the midwest and northeast U. S. reservoirs, lakes and estuaries of the South, and as far west as California (Crowell et al. 1967). Under favorable conditions, *M. spicatum* can spread very rapidly by vegetative fragmentation. Whitney et al. (1973) reported that a 40 ha stand of *M. spicatum* in Currituck Sound, N. C. in 1965 increased to nearly 27,100 ha by 1966. Kitty Hawk Bay, just south of Currituck Sound, was heavily infested with *M. spicatum* by 1968 (Anon. 1974b).

The North Carolina Department of Natural and Economic Resources (NCDNER) studied the Kitty Hawk Bay problem (Anon. 1974a) and later developed an environmental impact statement after persistent complaints about the *M. spicatum* infestation from residents. *Myriophyllum spicatum* often occurs in dense stands which limit recreation and commercial fishing. This species is not usually a significant component of waterfowl diets and it displaces desirable, native waterfowl food plants such as *Ruppia maritima* (widgeon grass), *Potamogeton pectinatus* (sago pondweed), Najas guadalupensis (southern naiad), Vallisneria americana (wild celery) and Potamogeton perfoliatus var. bupleuroides (redhead grass).

In the summer of 1974, Dare County, NCDNER, and the U. S. Army Corps of Engineers, Wilmington District, sponsored an herbicide program to treat 295 ha of M. spicatum in maximum use areas (commercial fishing, recreation, channels, docks, etc.) in Kitty Hawk Bay and 55 ha in southern Currituck Sound. A 20% active ingredient formulation of the butoxyethanol ester (BEE) of 2,4-D impregnated on baked attaclay granules was applied at the rate of 112 kg acid equivalent (ae)/ha.

Whitney et al. (1973) found that 2,4-D BEE at 112 kg ae/ha killed M. spicatum in Currituck Sound plots with a subsequent accelerated growth or "release" of valuable native rooted submersed macrophytes. Application of 2,4-D BEE at rates of 22 to 34 kg ae/ha from 1961 through 1964 in Chesapeake Bay resulted in no undesirable effects on native waterfowl food plants (R. maritima, P. pectinatus and V. americana); indeed, accelerated growth occurred in these species with the removal of the M. spicatum overstory (Rawls 1975). Wojtalik et al. (1971) found that the liquid dimethylamine (DMA) salt of 2,4-D at up to 45 kg ae/ha, did not seriously affect other submersed macrophytes in Tennessee Valley Authority (TVA) reservoirs. Beaven et al. (1962) reported that 2,4-D BEE at 67 kg ae/ha had little or no effect on native submersed macrophytes in the lower Potomac River; however, 135 kg ae/ha killed all vegetation.

Smith and Isom (1967) concluded that 2,4-D BEE at rates of 45 kg ae/ha and as high as 112 kg ae/ha did not produce significant recognizable effects on aquatic fauna or water quality in TVA reservoirs. No adverse effects of 2,4-D BEE on fish, shellfish, or blue crabs have been observed in the Chesapeake Bay - Currituck Sound area (Beaven et al. 1962; Rawls 1971; Whitney et al. 1973).

The Biology Department of East Carolina University was selected to monitor the effects of the 2,4-D treatment upon the submersed macrophyte communities at Kitty Hawk Bay and two sites in southern Currituck Sound. Inorganic nutrient levels, density and biomass of the submersed macrophytes during pre- and post-treatment periods were studied during the summers of 1974 and 1975. Simultaneous thesis research on changes in community metabolism was conducted by A. B. Hall.

This study was undertaken to (a) determine the effectiveness of 2,4-D BEE at 112 kg ae/ha on *M. spicatum* in low salinity estuarine systems, (b) determine the effect of the herbicide on certain native submersed macrophytes, (c) provide a detailed analysis of inorganic nutrients of submersed macrophytes in the Kitty Hawk Bay area, and (d) determine the type and temporal recolonization in the area following 2,4-D treatment.

DESCRIPTION OF STUDY AREA

The Kitty Hawk Bay - lower Currituck Sound area is located in northeastern, coastal North Carolina adjacent to several large, relatively shallow bodies of water which include Currituck Sound, Albemarle Sound and Kitty Hawk Bay (Fig. 1). The area is separated from the Atlantic Ocean by a barrier island extending southward from southeastern Virginia. Oregon Inlet, which marks the end of the barrier island some 40 km to the south, is the nearest direct connection to the sea. This is a shallow inlet with a low exchange volume. Thus, the Kitty Hawk Bay - lower Currituck Sound area has an extremely low salinity (less than 1 part per thousand), and is generally considered to be fresh water. At times the salinity may increase due to salt mist, wind tides and infrequent washovers across the barrier island. In the areas treated with 2,4-D the water depth varied from 1-2 m. Water turbidity was low and the bottom was easily visible most of the time. The bottom consisted of a firm sand-mud mixture, which became softer and more organic in the coves. All of the areas had a high density of submersed macrophytes, primarily dominated by *M. spicatum*. The entire area is affected by wind tides which can cause water levels to fluctuate as much as 50 cm.

Kitty Hawk Bay is a protected body of water with an irregular shoreline and numerous marsh islands. It is bounded on the east and north by the barrier island and on the south by Colington Island. The western end of the bay opens into Albemarle Sound. A study plot was selected within Kitty Hawk Bay (average depth around 1.5 m) on Colington

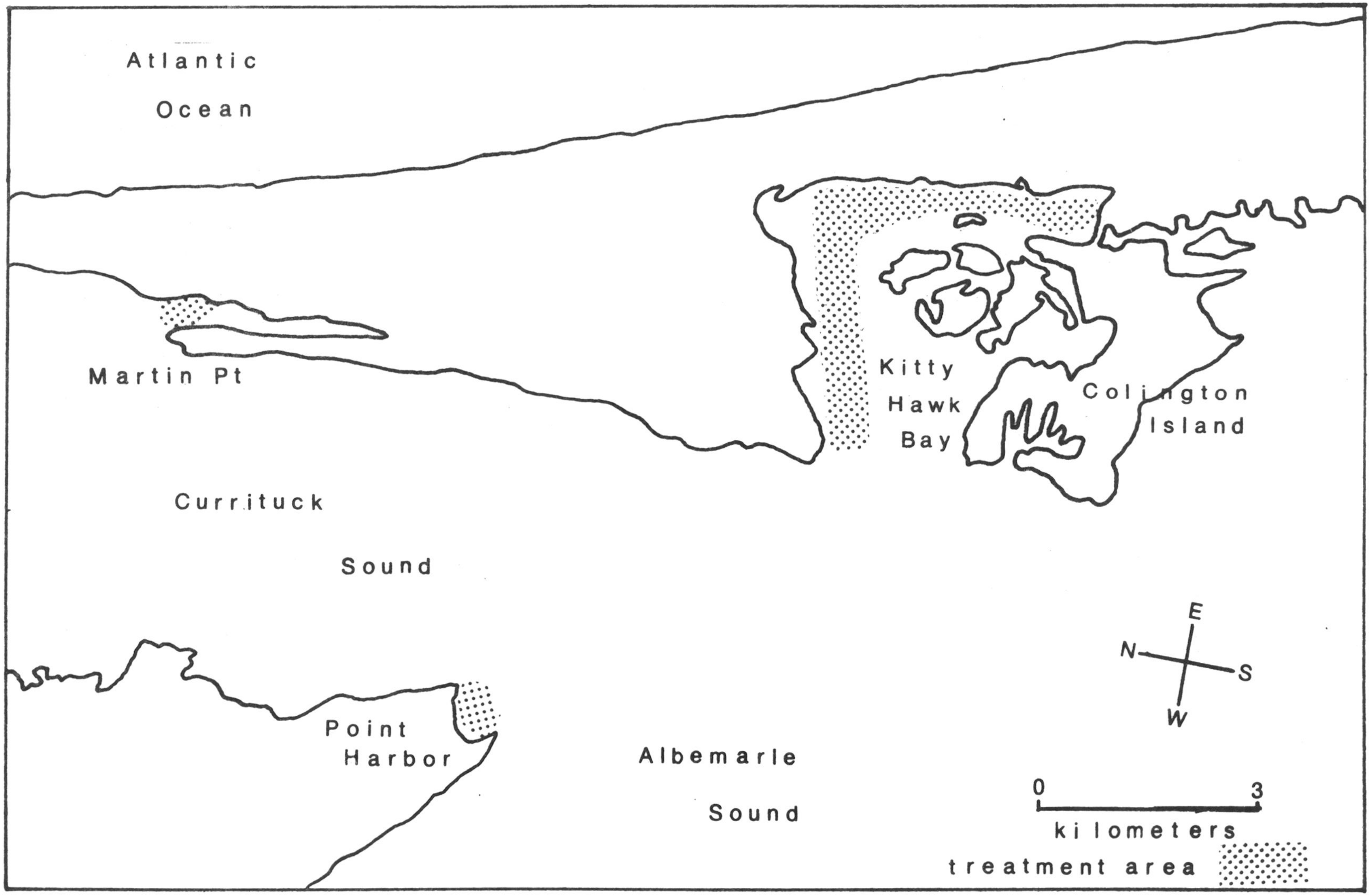

Fig. 1. The Kitty Hawk Bay - lower Currituck Sound area, North Carolina.

Creek (Fig. 2).

The Martin Point study plot (average depth around 1 m) was located at the mouth of Jean Guite Creek, which is surrounded on three sides by the barrier island, and on the north by Currituck Sound (Figs. 1 and 3). The Point Harbor study plot (average depth around 2 m) was on the southwest side of Currituck Sound (Figs. 1 and 4). Point Harbor is a cove, surrounded by sand bars which protect its waters from the wave action of Currituck and Albemarle sounds.

The Kitty Hawk Bay - lower Currituck Sound area, with its marshes and submersed vegetation, is an excellent habitat for wintering waterfowl. This wetland system supports a substantial freshwater game fish population including largemouth bass (*Micropterus salmoides*). Estuarine species such as the blue crab (*Callinectes sapidus*) and the brackish water clam (*Rangia cuneata*) are also common to the region.

Human activities in the area consist of recreation (sport fishing and hunting, skiing, sailing, etc.) and commercial fishing. Resident and vacation homes are located near all three study areas with the heaviest concentration on Colington Creek and Kitty Hawk Bay.

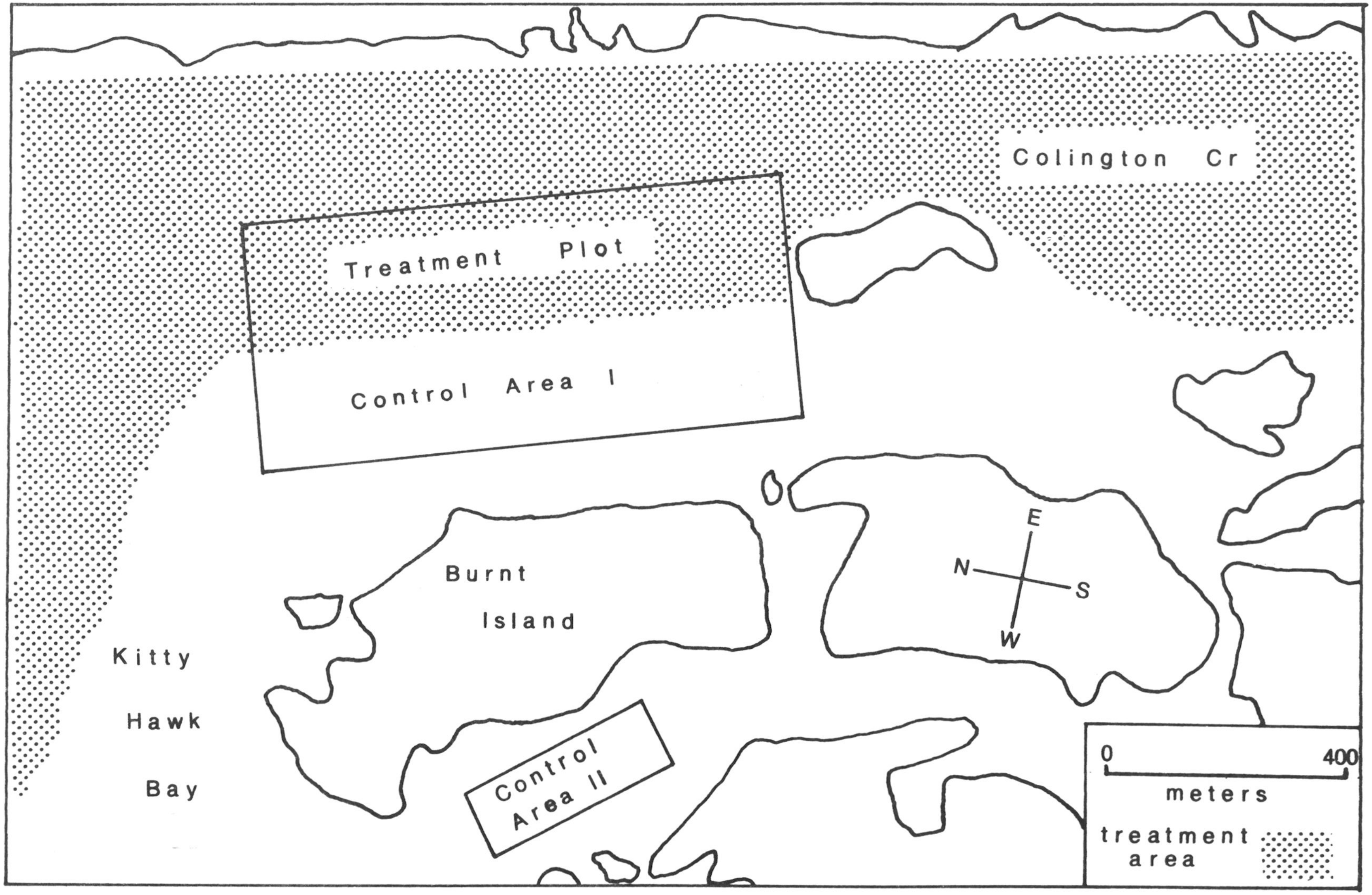

Fig. 2. The Kitty Hawk Bay study area, North Carolina.

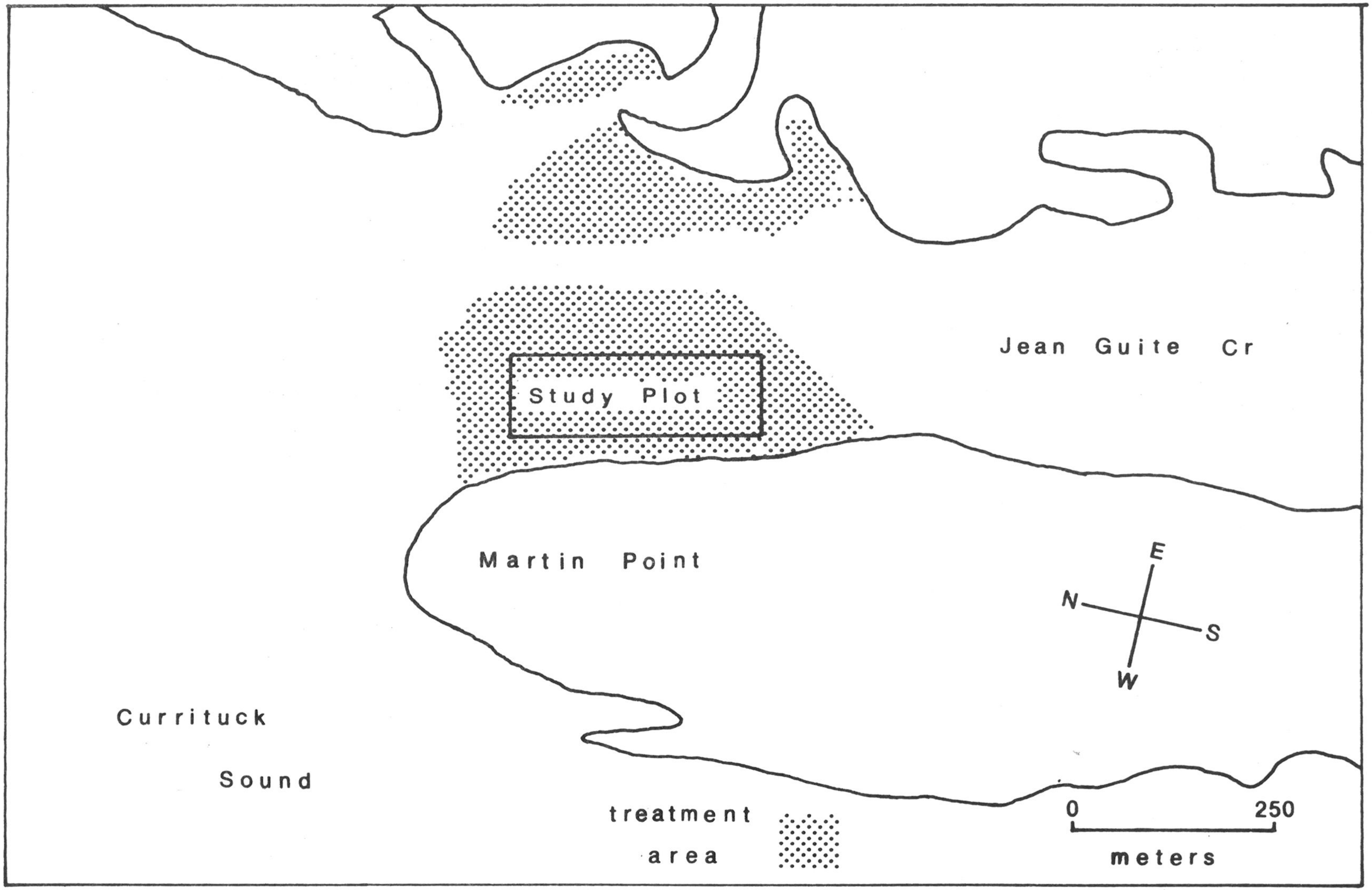

Fig. 3. The Martin Point study area, North Carolina.

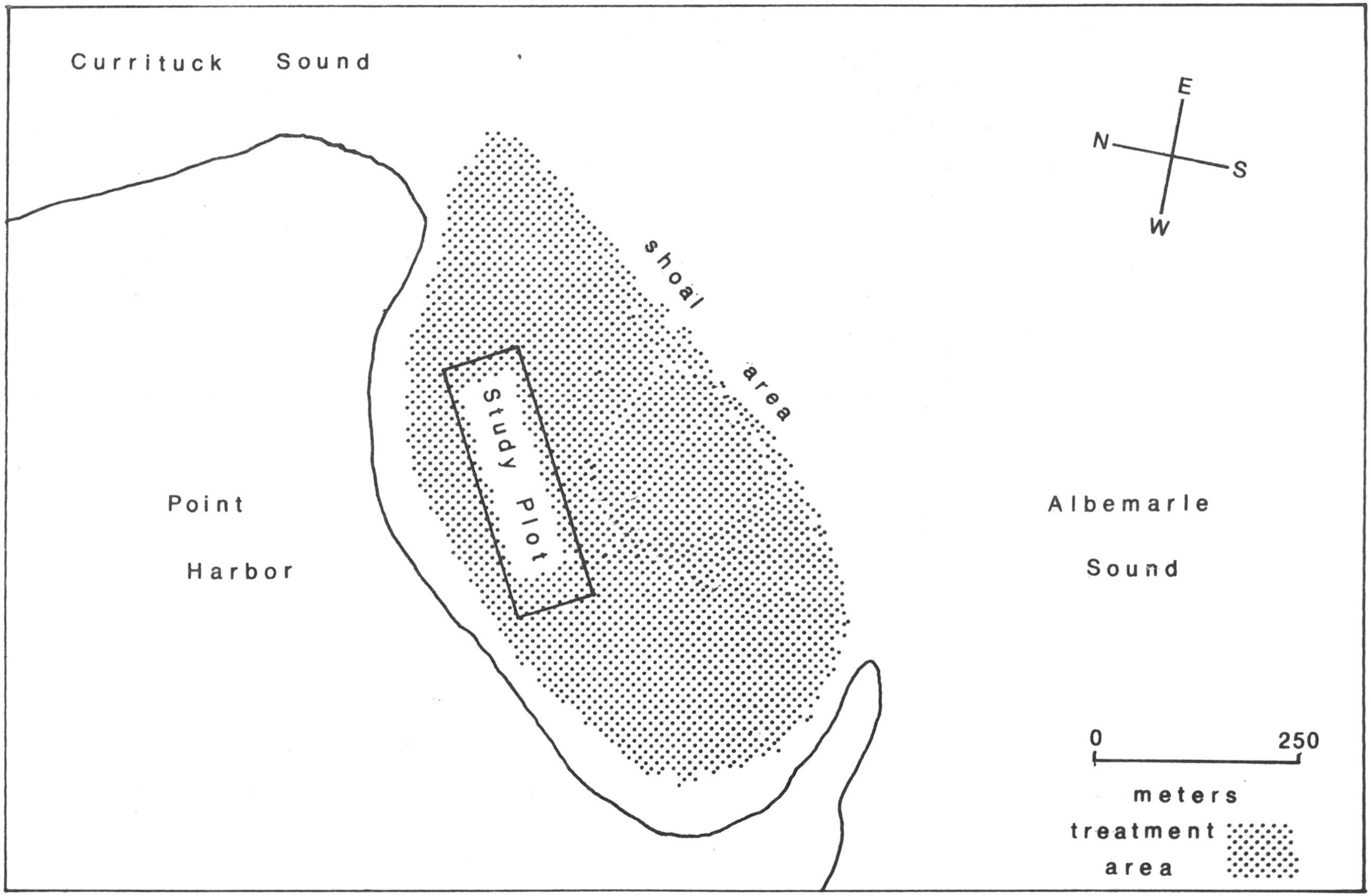

Fig. 4. The Point Harbor study area, North Carolina.

MATERIALS AND METHODS

On 13 and 14 July 1974, granular 2,4-D BEE was applied by helicopter on the three treatment areas at the rate of 112 kg ae/ha. Approximately 295 ha at Kitty Hawk Bay, 35 ha at Point Harbor and 20 ha at Martin Point were treated. Winds were northeasterly at 10 mph during application. About 3 weeks prior to treatment, study plots were established within Kitty Hawk Bay, at Martin Point and at Point Harbor (Figs. 2-4). Quadrats were chosen using a modification of the line intercept method of Lind and Cottam (1969).

In Kitty Hawk Bay a boundary line was established dividing the study plot into a treatment plot and an adjacent control plot (Control Area I). Midpoints of 10 randomly selected transects were marked with wooden stakes along this boundary line. Each transect ran perpendicular to the boundary line and was composed of 20 sample areas, indicated by anchored buoys at 25 m intervals. Ten of the buoys were in the treatment plot and 10 were in Control Area I. Buoys used in marking these transects were not permanent and were positioned by boat prior to sample collection. The total original study plot in Kitty Hawk Bay was approximately 500 x 1000 m. Control Area II (100 x 300 m) was established during the post-treatment study along the western edge of Burnt Island, adjacent to Control Area I (Fig. 2). Control Area II contained five transects, each comprised of 10 buoys at 10 m intervals.

Similar techniques were employed to establish five transects each in the Martin Point plot (100 x 300 m), and the Point Harbor plot (100 x 300 m). These transects were located entirely within the respective treatment areas and consisted of 10 buoys at 10 m intervals. Hence, no post-treatment control plots were established at these study areas.

Samples for stem counts were collected in all study areas during pretreatment (except Control Area II, which had not yet been established) and 2, 4, 6 and 52 weeks after treatment. Biomass samples were collected during pretreatment studies and 6 and 52 weeks after treatment. Since the harvest method was used, the post-treatment transects were positioned 2-3 m north of the initial (pretreatment) transects to prevent an overlapping of sample areas. Water depth was measured at all sample areas. Sample collection was facilitated by the use of skin diving apparatus or SCUBA gear depending on water depth. A square iron quadrat frame with an area of 0.1 m^2 was dropped on the bottom near the buoy and all plants rooted within the frame were harvested above the substrate. Stems were brought to the surface and counted to determine the density of each species.

Pretreatment biomass determinations were based upon the random selection of two, 0.1 m^2 quadrats along each transect in Kitty Hawk Bay giving a total of 40 quadrats, 20 in the treatment plot and 20 in Control Area I. Post-treatment biomass determinations were as above and collected on post-treatment weeks 6 and 52. Whole plants (roots and shoots) were dug from the quadrats and brought back to the laboratory. All plants from each quadrat were washed thoroughly, species were separated and counted, and the roots and shoots were separated. Samples were centrifuged in a top loading washing machine for 10 minutes on spin cycle and wet (fresh) weight determined. A 10-25 g aliquot was randomly selected from each sample and dried at 70 C for 24 h to a constant dry weight. Dried plant samples were ground in a 20 mesh Wiley mill. After grinding, each sample was sealed in a plastic bag and stored in a constant temperature room at 5 C prior to chemical analysis. Plant biomass in this paper is reported as organic dry weight (ODW). This is determined by subtracting the ash of a sample from the dry weight of a sample. Brillouin's measure H (Pielou 1966) was selected to calculate diversity. Due to the difficulty of defining individuals in vegetatively reproducing organisms, plant weight, as a measure of importance, was used in the calculation.

Inorganic nutrient and mineral analysis preparation was by ashing approximately 1 g of dried plant material at 500 C for 4 h. The ash weight was determined at room temperature. Phosphorus and cation analysis preparation was by dissolving the ash residue from 1 g of dried material in 6 N HCl. The resulting solution was brought to a boil and evaporated to near dryness on a hot plate, washed with deionized water, and filtered through Whatman # 541 filter paper. The filter paper was weighed to determine percent acid insoluble residue, presumably silica. The filtrate was diluted to 100 ml with deionized water and transferred to an acid washed polyethylene container, sealed and stored at 5 C. Phosphorus was determined by the ascorbic acid method (EPA Manual 1971) and nitrogen was determined using a Coleman Nitrogen Analyzer Model 29. Cations in the plant material were measured by atomic absorption (Ca and Mg) and flame emission (Na and K) using a Perkin-Elmer Model 305 B Atomic Absorption Spectophotometer (Perkin-Elmer Corp. 1973). The analyses were made by the Water Quality Laboratory of the Department of Biology at East Carolina University. Inorganic nutrients in this paper are expressed in microgram atoms per gram organic dry weight (µg-at/g ODW) for plant samples and microgram atoms per liter (µg-at/l) for water samples. These values are obtained by dividing the concentration of a nutrient in micrograms by its atomic weight. A summary of the number of quadrats sampled for submersed macrophyte inorganic nutrient analyses are presented in Tables 1 and 2 respectively.

Sediment samples were obtained with an Ekman dredge. Ten samples were collected from Control Area I, 10 from Control Area II and 10 from the treatment plot in Kitty Hawk Bay. The dredge was hand pushed approximately 10 cm into the substrate and the trapped materials were carried to the surface and washed. Wet, dry and ash weights of the organic material were determined in the laboratory as for the plant samples.

Surface water samples for inorganic nutrient analysis were collected weekly from 13 June to 20 August 1974, and from 18 July to 12 August 1975. Samples were taken from the treatment area and Control Areas I and II in Kitty Hawk Bay and from the treatment areas at Martin Point and Point Harbor. Filtered and unfiltered samples were frozen until analyzed by the Water Quality Laboratory. The following tests were conducted spectrophotometrically on the water samples: phosphorus, ascorbic acid method (EPA Manual 1971); nitrites, azo-method (EPA Manual 1971); and nitrates, brucine method (EPA Manual 1971). Total nitrogen was by Kjedahl digestion followed by analysis with Orion ammonia electrode (EPA Manual 1971; Orion Ammonia Electrode Instruction Manual 1971).

Table 1. Summary of the number of quadrats sampled for submersed macrophyte inorganic nutrient analysis at Kitty Hawk Bay, summers 1974 and 1975.

Species	Pretreatment	Post-treatment	
		6 weeks	52 weeks
Myriophyllum spicatum	20	8	4
Ruppia maritima	9	5	10
Najas guadalupensis	8	3	10
Potamogeton pectinatus	5	0	0
P. perfoliatus	0	0	2
Vallisneria americana	0	0	4
Nitella hyalina	0	0	32
Totals	42	16	62

Table 2. Precision for extraction and analysis of inorganic nutrients in *M. spicatum* stems.

Nutrient	$\bar{x}$ (μg-at/g ODW)	95% confidence limits (μg-at/g ODW)
N	264	± 6.7
P	239	± 1.8
Na	604	±10.6
K	283	± 3.8
Ca	35	± 1.3
Mg	67	± 2.3

Ammonia was determined with an Orion ammonia electrode (Orion Ammonia Electrode Instruction Manual 1971). Calcium, magnesium, sodium and potassium were determined with the same techniques used on the plant samples.

RESULTS AND DISCUSSION

Observations on Treated *Myriophyllum spicatum*

Approximately 24 h after 2,4-D application, the treated *M. spicatum* in all three study areas began to develop symptoms reported by Whitney et al. (1973). The flower spikes, which are normally several cm above the water surface in July, fell flat upon the water and the tips of the stems began to twist and curl in abnormal positions. The upper 10-15 cm of the stems became bloated and puffy. By 72 h the brittle stems were easily broken at the internodes and the leaf abscission accelerated. The stems began to brown and sink towards the bottom by 96 h. Native submersed macrophytes appeared healthy during the same period. *Myriophyllum spicatum* continued to sink and deteriorate and, by the fifteenth day, it had been eliminated in all treated areas. A few shoot fragments remained attached to the rhizomes, but most separated and formed mats along the surface. These stem mats moved with the wind tides and some were observed by commercial fishermen to have traveled from Kitty Hawk Bay into Albemarle Sound. Most of the mats, however, drifted onto the shore in the treated areas where decomposition continued. More shore mats were visible at Kitty Hawk Bay than in the other two study areas. Leaf deterioration could not be followed once the leaves fell from the stems. Leaves were not observed in the stem mats, nor were they found on the bottom. Leaf decomposition was rapid and/or the leaves were dispersed by wind tides. Slower decomposition in the stems could be expected since they contain a greater proportion of refractory materials such as cellulose and lignin. General observations and analysis of the

Ekman dredge samples gave no evidence of particulate detrital accumulation on or in the sediment.

In Kitty Hawk Bay the effectiveness of the 2,4-D beyond the treated area was much greater than expected. Myriophyllum spicatum in Control Area I began to show symptoms of herbicide application at 72 h post-treatment. The plants continued to deteriorate in Control Area I until they were eliminated, 15 days after treatment. In effect, Control Area I became a part of the Kitty Hawk Bay treatment area. Therefore, these combined areas will simply be referred to as the Kitty Hawk Bay Treatment Area. Control Area II was established at this time. Previous investigators working in Currituck Sound (Whitney et al. 1973), reported the maximum drift effect of granular 2,4-D to be approximately 46 m from the treatment edge. The drift of 2,4-D in Kitty Hawk Bay was over 300 m from the treatment edge. This large drift effect was only noted on the western side of the treated area and was not observed at the Martin Point and Point Harbor study areas. The westward drift of the herbicide in Kitty Hawk Bay was most likely due to wind tides created by the northeasterly wind during the application period. A northeasterly wind tide would tend to spread the herbicide towards the nearby western shore at Point Harbor and Martin Point; thus, the extensive drift effect did not occur at those two study areas. The treatment area at Kitty Hawk Bay, however, was bounded on the west by a 300-350 m expanse of untreated, open water. Approximately 600 ha of M. spicatum were eradicated in Kitty Hawk Bay by the application of 2,4-D, although only 290 ha were directly treated.

Once M. spicatum disappeared from the study area, wave action increased along with a corresponding increase in water turbidity as the bottom was stirred up. Secchi disc transparency was less than 0.3 m after the M. spicatum decline, as compared with greater than 1.5 m before treatment. Native submersed macrophytes began to deteriorate during the second post-treatment week. By the seventh post-treatment week a phytoplankton bloom (which also contributed to turbidity) was observed in Kitty Hawk Bay. The bloom was composed primarily of the cyanophytes Aphanizomenon and Anacystis (Hall 1976). About the time of this bloom a 30-50 ha M. spicatum die-back began in Control Area II and surrounding coves. This delayed die-back did not display the typical 2, 4-D symptoms, but had a much slower type of deterioration. The leaves remained attached to the stems as decomposition progressed and some of the plants became coated with what appeared to be an algal slime. Algal growths have been reported on Chesapeake Bay M. spicatum by Rawls (1975). This late M. spicatum decline appeared similar to that described by Bayley et al. (1968) which they attributed to "Lake Venice disease".

Young M. spicatum shoots were observed growing from fragments of deteriorated stems, which were still attached to old rhizome systems, at Point Harbor and Martin Point 8 weeks after treatment. Sprouting at 6 weeks post-treatment has been reported by Steenis and Stotts (1965). Accelerated growth and substantial re-establishment of vascular native submersed macrophytes did not occur in any of the treated areas during the study period. In fact, native macrophyte abundance dramatically declined during the first 8 weeks of the post-treatment period. The native macrophyte decline might be attributed to the relatively high concentration of 2,4-D (112 kg/ha) used in treatment. Beaven et al. (1962) reported that 2,4-D BEE at 135 kg/ha killed all submersed vegetation in the lower Potomac River, however, Whitney et al. (1973) found that 2,4-D BEE at 112 kg/ha killed only *M. spicatum* in Currituck Sound.

One year after 2,4-D application, *M. spicatum* was beginning to make a slow recovery in Kitty Hawk Bay and had become well established at Martin Point and Point Harbor. Whitney et al. (1973) reported a 50% reinfestation after 52 weeks in treated areas of Currituck Sound which were surrounded by *M. spicatum* beds. No harmful effects were observed with respect to fauna in the treatment areas following 2,4-D application and subsequent habitat changes. Secchi disc readings in 1975 (0.4 m) reflected a slight decrease in turbidity in the three study areas, although turbidity was still considerably higher than the pretreatment level of 1974 (1.5 m).

Changes in Density and Biomass of Submersed Macrophytes Following Treatment

Four species of submersed macrophytes were found in the pretreatment study areas at Kitty Hawk Bay, Martin Point and Point Harbor: *Myriophyllum spicatum* L., *Ruppia maritima* L., *Potamogeton pectinatus* L. and *Najas guadalupensis* Mangus. Earlier observations of Kitty Hawk Bay macrophytes reported by Fish (Anon. 1974a), indicated a shift in species composition from all native species in 1963 to a 75% *M. spicatum*-25% native species community in 1973 (Table 3). A U. S. Fish and Wildlife Service report (Anon. 1965) shows that beds of *P. pectinatus*, *V. americana*, *P. perfoliatus*, *R. maritima* and *N. guadalupensis* were found at Point Harbor and Martin Point at various times between 1959 and

Table 3. Relative abundance of submersed macrophytes in Kitty Hawk Bay between 1963 and 1974.[a]

Species	1963	1966	1968	1973	1974
Myriophyllum spicatum	-	-	25%	75%	58%
Ruppia maritima	50%	90%	37%	-	23%
Najas guadalupensis	-	trace	-	-	17%
Potamogeton pectinatus	45%	trace	37%	25%	2%
P. perfoliatus	5%	1%	-	-	-
Vallisneria americana	-	5%	-	-	-

[a] Data for 1963 through 1973 (Anon. 1974b).

Table 4. Percent frequency of submersed macrophytes at Point Harbor and mouth of Jean Guite Creek (Martin Point) during 1959, 1960 and 1962.[a]

	Martin Point			Point Harbor		
Species	1959	1960	1962	1959	1960	1962
Ruppia maritima	100	80	0	30	0	80
Najas guadalupensis	80	80	-	20	0	-
Potamogeton pectinatus	0	40	20	0	0	0
P. perfoliatus	0	60	40	20	20	50
Vallisneria americana	70	20	0	70	50	70

[a] Data is from Back Bay - Currituck Sound Data Report (Anon. 1965) and shows the percent frequency of each macrophyte based on 10, 0.47 m^2 samples from 83.5 ha quadrats.

1962 (Table 4).

In the summer of 1974, pretreatment densities of submersed macrophytes in Kitty Hawk Bay and Martin Point were approximately 1000 stems/m^2, while they totaled 300 stems/m^2 at Point Harbor (Tables 5 and 6). *Myriophyllum spicatum* contributed most to the density with 58% at Kitty Hawk Bay and Martin Point and 100% at Point Harbor. Although no native species were found within the immediate treatment area at Point Harbor, beds of *Potamogeton perfoliatus* var. *bupleroides* (Fernald) Farwell were observed outside of the treatment area on sand bars which separate Albemarle and Currituck sounds from the Point Harbor cove. (Henceforth *Potamogeton perfoliatus* var. *bupleroides* will be referred to as *Potamogeton perfoliatus*.) Twenty-three percent of the macrophyte stands at Kitty Hawk Bay and Martin Point consisted of *R. maritima*, while 17% was *N. guadalupensis* and 2% *P. pectinatus*. Native submersed macrophyte density was substantial in the pretreatment Kitty Hawk Bay and Martin Point areas. The native plants formed an understory beneath the taller *M. spicatum* stems. This difference in size may have been related to the high pretreatment native macrophyte density, even though biomass was low. The macrophyte stands here (>1000 stems/m^2) had a considerably greater density than the *Potamogeton*-*M. spicatum* community in a southern Wisconsin lake (192 stems/m^2) reported by Nichols and Mori (1971), or the *Myriophyllum* community of Lake Wingra (368 stems/m^2) found by Gustafson and Adams (1973).

In the weeks following 2,4-D treatment, the submersed macrophyte density decreased until the sixth post-treatment week when only 122.5 stems/m^2 at Kitty Hawk Bay and 35 stems/m^2 at Martin Point remained

Table 5. Densities of submersed macrophytes (stems/m^2) in Treatment Area and Control Area II at Kitty Hawk Bay, summers 1974 and 1975.

Species	Pretreatment	Post-treatment 2 wk	4 wk	6 wk	52 wk
		Treatment Area			
Myriophyllum spicatum	626.3	0.0	0.0	0.0	4.5
Ruppia maritima	248.7	234.7	180.7	110.5	64.9
Najas guadalupensis	188.0	11.0	7.4	7.2	41.0
Potamogeton pectinatus	17.9	1.2	0.6	4.8	0.0
P. perfoliatus	0.0	0.0	0.0	0.0	3.5
P. foliosus	0.0	0.0	0.0	0.0	1.3
Vallisneria americana	0.0	0.0	0.0	0.0	3.8
Totals	1080.9	246.9	188.7	122.5	119.0
		Control Area II			
Myriophyllum spicatum	-	381.6	219.6	136.2	7.8
Ruppia maritima	-	210.4	219.4	205.6	937.5
Najas guadalupensis	-	93.0	117.4	93.6	330.5
Potamogeton pectinatus	-	2.4	0.4	0.0	0.0
P. perfoliatus	-	0.0	0.4	0.0	1.6
P. foliosus	-	0.0	0.0	0.0	0.0
Vallisneria americana	-	0.2	0.0	0.0	12.7
Totals	-	687.6	557.2	435.4	1289.6

Table 6. Densities of submersed macrophytes (stems/m^2) in the study areas at Martin Point and Point Harbor, summers 1974 and 1975.

Species	Pretreatment	Post-treatment 2 wk	4 wk	6 wk	52 wk
		Martin Point			
Myriophyllum spicatum	584.0	0.0	0.0	0.0	284.6
Ruppia maritima	255.6	112.0	73.8	24.8	36.8
Najas guadalupensis	165.0	27.2	24.0	10.2	3.8
Potamogeton pectinatus	0.0	0.0	0.0	0.0	0.0
P. perfoliatus	0.0	0.0	0.0	0.0	0.0
P. foliosus	0.0	0.0	0.0	0.0	0.0
Vallisneria americana	0.0	1.2	1.8	0.0	10.6
Totals	1004.6	140.2	99.6	35.0	335.8
		Point Harbor			
Myriophyllum spicatum	308.0	0.0	0.0	0.0	64.8
Ruppia maritima	0.0	0.0	0.0	0.0	0.0
Najas guadalupensis	0.0	0.0	0.0	0.0	0.0
Potamogeton pectinatus	0.0	0.0	0.0	0.0	0.0
P. perfoliatus	0.0	0.0	0.0	0.0	0.0
P. foliosus	0.0	0.0	0.0	0.0	0.0
Vallisneria americana	0.0	0.0	0.0	0.0	0.0
Totals	308.0	0.0	0.0	0.0	64.8

(Tables 5 and 6). No macrophytes remained at the Point Harbor study plot. No *M. spicatum* was present in any of the treated areas, and native macrophyte densities decreased in all of the study areas. In Kitty Hawk Bay 90% of the remaining macrophytes consisted of *R. maritima*, 6% *N. guadalupensis* and 4% *P. pectinatus*. At Martin Point 70% was *R. maritima*, 29% *N. guadalupensis* and <1% *P. pectinatus*. Control Area II at Kitty Hawk Bay also declined in submersed macrophyte density (Table 5).

Approximately 52 weeks after 2,4-D treatment, *M. spicatum* had recovered to some extent in all three areas, except Control Area II. Recovery in the Kitty Hawk Bay Treatment Area reached a level of 4.5 stems/m^2, compared to the pretreatment level of 626.3 stems/m^2. During the 52 week post-treatment study, heavy stands of flowering *M. spicatum* were observed near the Kitty Hawk Bay Control Area II study plot, but were absent directly in the plot, where the *M. spicatum* density was 7.8 stems/m^2. *Myriophyllum spicatum* recovered to nearly one-half of the pretreatment density at Martin Point, from 584 stems/m^2 in 1974 to 284.6 stems/m^2 in 1975. The density may actually have been somewhat higher than the recorded values since the main stand of the macrophyte had shifted south of the 1974 treatment and study area. The Point Harbor study plot, which contained a pure stand of *M. spicatum* in 1974 (308 stems/m^2), supported another pure stand of *M. spicatum* in 1975, although in reduced amounts (64.8 stems/m^2).

Native submersed macrophytes were considerably affected by the 2, 4-D application. In Kitty Hawk Bay Treatment Area, *N. guadalupensis* decreased rapidly following treatment from 188 stems/m^2 pretreatment to a low of 7.2 stems/m^2 at six weeks post-treatment (Table 5).

In 1975, N. guadalupensis had recovered to 41 stems/m^2. Ruppia maritima decreased more slowly during the post-treatment period (248.7 stems/m^2 to 110.5 stems/m^2). One year after herbicide application in Treatment Area, R. maritima continued to decrease to 64.9 stems/m^2. Potamogeton pectinatus, a less abundant pretreatment plant, eventually seemed to disappear from the post-treatment study area. Vallisneria americana Michaux, P. perfoliatus and Potamogeton foliosus Raf. were found in small numbers in the 52 week post-treatment study areas. In Control Area II, native macrophyte densities remained fairly constant through the sixth post-treatment week in spite of the M. spicatum deterioration (Table 5). By the fifty-second post-treatment week, R. maritima and N. guadalupensis had become very abundant (937.5 stems/m^2 and 330.5 stems/m^2).

A "rooted" alga, Nitella hyalina (DC.) Ag was the dominant submersed macrophyte in the 52 week Kitty Hawk Bay Treatment Area study plots. Due to the nature of the plant, stems counts were impractical; however, most of the substrate in these two areas was covered by a dense mat of the macrophyte and biomass measurements were taken. In Control Area II N. hyalina was sparse. Traces of the alga were found in areas near the study plots during the 1974 study. Native macrophytes at Martin Point followed trends similar to the decline in abundance of the native macrophytes in the treated area of Kitty Hawk Bay (Table 6). No N. hyalina was observed at Martin Point or Point Harbor.

Total pretreatment organic dry weight (ODW) of the submersed macrophytes ranged from 120 g ODW/m^2 at Point Harbor to 238.4 g ODW/m^2 at Martin Point and 258.4 g ODW/m^2 at Kitty Hawk Bay (Fig. 5). Although

M. spicatum only accounted for 58% of the macrophyte density at Kitty Hawk Bay and Martin Point, it comprised over 95% of the biomass in the pretreatment study plots, while the remainder consisted of various native species (Tables 7 and 8). Nichols and Mori (1971) found a shallow water Myriophyllum community in Lake Wingra, composed of a solid stand of M. spicatum interspersed with other species, to have a standing crop of 385 g dry weight (DW)/m^2. The same lake also contained a Potamogeton-M. spicatum community with a standing crop of 196 g DW/m^2. Lind and Cottam (1969) reported an M. exalbescens community standing crop in Lake Mendota of 176 g DW/m^2. The maximum standing crop in macrophyte beds in the Pamlico River N. C. estuary was 57.8 g ODW/m^2 (Vicars 1976a). Most of this biomass was V. americana. One of the most productive fresh water submersed macrophyte communities in the world supported a biomass of angiosperms in a eutrophic lake near Osbysjon, Sweden of 680 g DW/m^2 (Westlake 1963). Six weeks after treatment M. spicatum had disappeared in all plots, with native macrophyte reduction in Kitty Hawk Bay from 15.5 g ODW/m^2 to 3.6 g ODW/m^2, and at Martin Point from 10.4 g ODW/m^2 to 0.8 g ODW/m^2. In Control Area II (Fig. 5) the total biomass was 76.8 g ODW/m^2, the bulk of which was M. spicatum with a standing crop of 71.9 g ODW/m^2 (Table 7).

During the fifty-second post-treatment week the total biomass had reached 22.8 g ODW/m^2 in Treatment Area and 33.6 g ODW/m^2 in Control Area II (Fig. 5). Table 7 shows that M. spicatum only accounted for 0.8 g ODW/m^2 of the standing crop in Treatment and Control Area I and 1.3 g ODW/m^2 in Control Area II. The dominant native macrophyte in Treatment Area was N. hyalina (19.8 g ODW/m^2), while the dominant macro-

Table 7. Organic dry weight (g/m^2) of submersed macrophytes in Treatment Area and Control Area II at Kitty Hawk Bay, summers 1974 and 1975.

Species	Pretreatment	Post-treatment 6 weeks	Post-treatment 52 weeks
		Treatment Area	
Myriophyllum spicatum	242.9	0.0	0.8
Ruppia maritima	7.2	2.9	1.1
Najas guadalupensis	3.5	0.1	0.5
Potamogeton pectinatus	4.8	0.6	0.0
P. perfoliatus	0.0	0.0	0.2
Vallisneria americana	0.0	0.0	0.4
Nitella hyalina	0.0	0.0	19.8
Totals	258.4	3.6	22.8
		Control Area II	
Myriophyllum spicatum	-	71.9	1.3
Ruppia maritima	-	3.0	24.6
Najas guadalupensis	-	1.9	3.8
Potamogeton pectinatus	-	0.0	0.0
P. perfoliatus	-	0.0	0.1
Vallisneria americana	-	0.0	0.4
Nitella hyalina	-	0.0	3.4
Totals	-	76.8	33.6

Table 8. Organic dry weight (g/m^2) of submersed macrophytes in the study areas at Martin Point and Point Harbor, summers 1974 and 1975.

Species	Pretreatment	Post-treatment 6 weeks	Post-treatment 52 weeks
		Martin Point	
Myriophyllum spicatum	228.0	0.0	48.4
Ruppia maritima	7.3	0.7	0.7
Najas guadalupensis	3.1	0.1	0.1
Potamogeton pectinatus	0.0	0.0	0.0
P. perfoliatus	0.0	0.0	0.0
Vallisneria americana	0.0	0.0	4.7
Nitella hyalina	0.0	0.0	0.0
Totals	238.4	0.8	53.9
		Point Harbor	
Myriophyllum spicatum	120.0	0.0	11.0
Ruppia maritima	0.0	0.0	0.0
Najas guadalupensis	0.0	0.0	0.0
Potamogeton pectinatus	0.0	0.0	0.0
P. perfoliatus	0.0	0.0	0.0
Vallisneria americana	0.0	0.0	0.0
Nitella hyalina	0.0	0.0	0.0
Totals	120.0	0.0	11.0

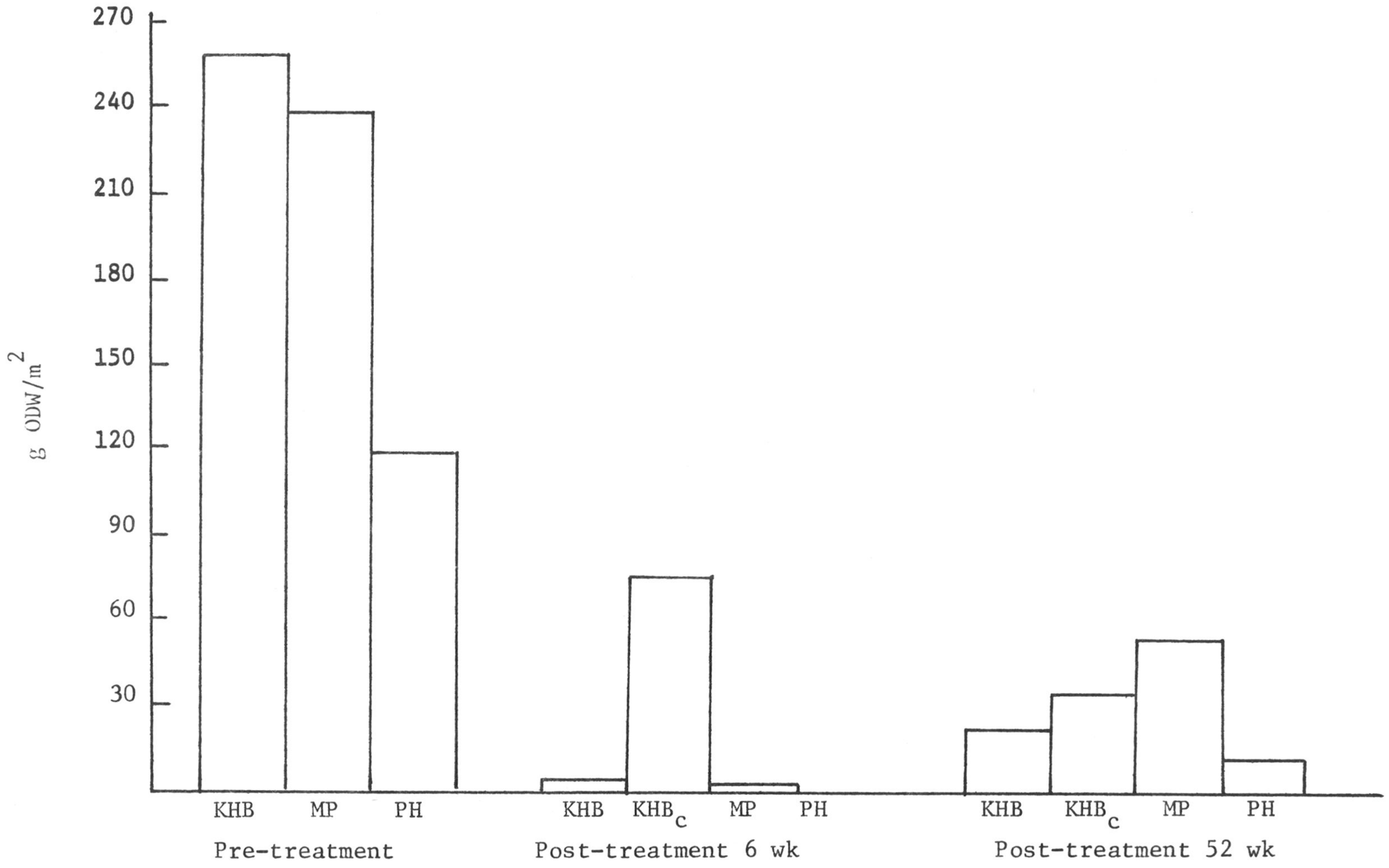

Fig. 5. Organic dry weight (ODW) of submersed macrophytes in Kitty Hawk Bay Treatment Area (KHB), Control Area II (KHB$_c$), Martin Point (MP) and Point Harbor (PH), summers 1974 and 1975.

phyte of Control Area II was R. maritima (24.6 g ODW/m^2). The Martin Point standing crop recovered to 53.9 g ODW/m^2 at 52 weeks post-treatment, with M. spicatum contributing 48.4 g ODW/m^2 (Table 8). Once again Point Harbor consisted of a pure M. spicatum bed with a standing crop of 11 g ODW/m^2. Low post-treatment native submersed macrophyte biomass confirms the previously mentioned lack of "released" native vascular macrophyte growth in the treated areas.

Submersed macrophyte species diversity, based upon biomass (Brillouin's measure H), is shown in Table 9. Kitty Hawk Bay diversity ranged from 0.11 pretreatment to 0.18 52 weeks post-treatment. Vicars (1976a), using Brillouin's measure H, found submersed macrophyte diversity as high as 2.8 in the Pamlico River, N. C. Diversity indices tended to increase during the post-treatment study period at Kitty Hawk Bay and Martin Point as the number of species increased and the biomass was more equally distributed among species. The Point Harbor study community consisted of a single species, M. spicatum.

Inorganic Nutrients of Submersed Macrophytes in Kitty Hawk Bay

Nutrient concentrations were determined for all species of submersed macrophytes collected during the summers of 1974 and 1975 (Tables 10-12). Only three species, M. spicatum, R. maritima and N. guadalupensis, were found both before and one year after 2,4-D treatment. Other species such as V. americana, P. perfoliatus and N. hyalina, appeared after herbicide application, while P. pectinatus was present before treatment but was not found during the post-treatment study period.

Shoot and root-rhizome nutrient concentrations of M. spicatum, R. maritima and N. guadalupensis from Kitty Hawk Bay Treatment Area, summers

Table 9. Diversity indices of submersed macrophytes at Kitty Hawk Bay, Martin Point and Point Harbor, summers 1974 and 1975.

Area and Date	No. of Species	Diversity (H)[a]
Kitty Hawk Bay		
Treatment Area		
Pretreatment	4	0.11
6 wk post-treatment	3	0.15
52 wk post-treatment	6	0.18
Control Area II		
6 wk post-treatment	3	0.11
52 wk post-treatment	6	0.33
Martin Point		
Pretreatment	3	0.08
6 wk post-treatment	2	0.11
52 wk post-treatment	4	0.15
Point Harbor		
Pretreatment	1	0.00
6 wk post-treatment	0	0.00
52 wk post-treatment	1	0.00

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[a] Brillouin's Index modified for biomass (Pielou 1966).

$$H=\frac{1}{N}\ln\frac{N!}{(N_1!\,N_2!\,N_3!\,\ldots\,N_s!)}$$

N=total weight of all species

N_1 N_2 N_3 ... N_s=weight of each species

Table 10. Inorganic nutrients (μg-at/g ODW) of submersed macrophytes in Treatment Area at Kitty Hawk Bay, summer 1974.

	Pretreatment					
Species	N	P	Na	K	Ca	Mg
Myriophyllum spicatum	1030[a]	18.08	462.6	272.4	213.7	91.63
	1140	26.75	286.5	337.0	269.4	153.5
Ruppia maritima	1600	43.59	387.1	802.8	246.3	184.1
	1330	32.39	280.7	515.1	224.1	183.1
Najas guadalupensis	2350	68.77	641.2	1811	370.6	183.6
	2160	48.85	742.8	1098	342.6	171.1
Potamogeton pectinatus	1210	37.48	487.3	622.7	371.7	176.9
	1030	24.27	296.1	388.6	178.5	96.08
	Post-treatment 6wk					
Ruppia maritima	1770	14.10	758.6	1142	187.8	278.2
	1540	35.10	727.5	901	194.5	237.8

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[a] $\frac{\text{shoots}}{\text{roots-rhizomes}}$

Table 11. Inorganic nutrients (μg-at/g ODW) of submersed macrophytes in Treatment Area at Kitty Hawk Bay, summer 1975.

Species	N	P	Na	K	Ca	Mg
Myriophyllum spicatum	1810[a] 1960	39.18 52.50	1300 794	635.4 745.6	158.6 332.4	175.9 280.7
Ruppia maritima	1990 1580	38.57 40.99	544.8 572.9	806.5 1048	220.9 250.5	308.6 263.5
Najas guadalupensis	2480 1730	64.41 54.21	461.3 754.9	1395 995.3	224.3 315.7	319.3 321.3
Potamogeton perfoliatus	1700 1660	57.52 41.63	453.6 1107	286.6 982.1	254.7 256.4	460.9 206.5
Vallisneria americana	1980 1970	55.45 39.17	902.2 1327	531.5 1865	692.5 330.2	238.4 245.7
Nitella hyalina "shoots and roots"	2410	48.32	487.3	531.5	692.5	238.4

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[a] $\frac{\text{shoots}}{\text{roots-rhizomes}}$

Table 12. Inorganic nutrients (μg-at/g ODW) of submersed macrophytes in Control Area II at Kitty Hawk Bay, summers 1974 and 1975.

	Post-treatment 6 wk					
Species	N	P	Na	K	Ca	Mg
Myriophyllum spicatum	590[a] 1280	16.51 28.38	562.2 293.1	361.1 184.9	163.5 291.4	158.8 160.8
Ruppia maritima	1560 1280	47.02 48.77	376.5 233.0	673.3 488.1	254.2 273.5	226.5 164.6
Najas guadalupensis	2070 2220	54.12 51.38	391.4 516.8	1361 717.3	284.1 357.1	187.2 172.6
	Post-treatment 52 wk					
Myriophyllum spicatum	1130 971	28.38 30.35	555.9 266.7	290.6 391.9	184.4 204.7	132.5 161.5
Ruppia maritima	2120 1310	66.67 45.69	536.0 571.6	954.7 1134	254.6 256.6	272.2 214.4
Najas guadalupensis	2290 1980	58.62 43.67	560.6 596.6	1564 940.0	305.2 334.4	250.1 202.4
Vallisneria americana	2330 2340	65.30 53.08	1106 1237	2076 1794	385.6 417.3	254.4 175.6
Nitella hyalina "shoots and roots"	2050	39.36	309.0	444.7	666.4	194.8

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[a] $\frac{\text{shoots}}{\text{roots-rhizomes}}$

1974 and 1975, are presented in Figs. 6-11. All of the nutrients measured in the three pretreatment plants, with the exception of sodium, tended to be highest in N. guadalupensis, followed by R. maritima and M. spicatum (Figs. 6, 7, 10-12). Fifty-two weeks after treatment, M. spicatum appeared to have the largest increase in nutrient concentrations, but the highest nutrient levels were still in N. guadalupensis. Sodium concentrations were greatest in N. guadalupensis pretreatment (Fig. 8) and M. spicatum 52 weeks post-treatment.

In most cases, nutrient levels of M. spicatum from Kitty Hawk Bay were low when compared to Myriophyllum studied by other investigators. Based on dry weight, Kitty Hawk Bay M. spicatum nitrogen was 1.16% in 1974 and 1.81% in 1975, while phosphorus was 0.06% in 1974 and 0.10% in 1975. These levels were considerably below levels of nitrogen (4.13%) and phosphorus (0.42%) of M. spicatum found by Nelson and Palmer (1938) in a Minnesota lake, or the nitrogen and phosphorus levels (3% and 0.40%) of Chesapeake Bay M. spicatum reported by Anderson et al. (1966). One exception noted was the nitrogen and phosphorus concentrations (1.51% and 0.05%) of M. spicatum reported for resevoirs of Kagawa, Japan by Umeda and Tamaki (1959).

A literature review of inorganic nutrients found in Myriophyllum is summarized in Table 13, and P. pectinatus, V. americana and Najas in Table 14. Ash content between and among species of submersed macrophytes may vary with epiphyte density, trapped sediment, water quality and other physical factors. Therefore, it must be realized that the most meaningful method of reporting results, μg-at/g ODW, was not used in Tables 13 and 14, because insufficient information was supplied by some of the

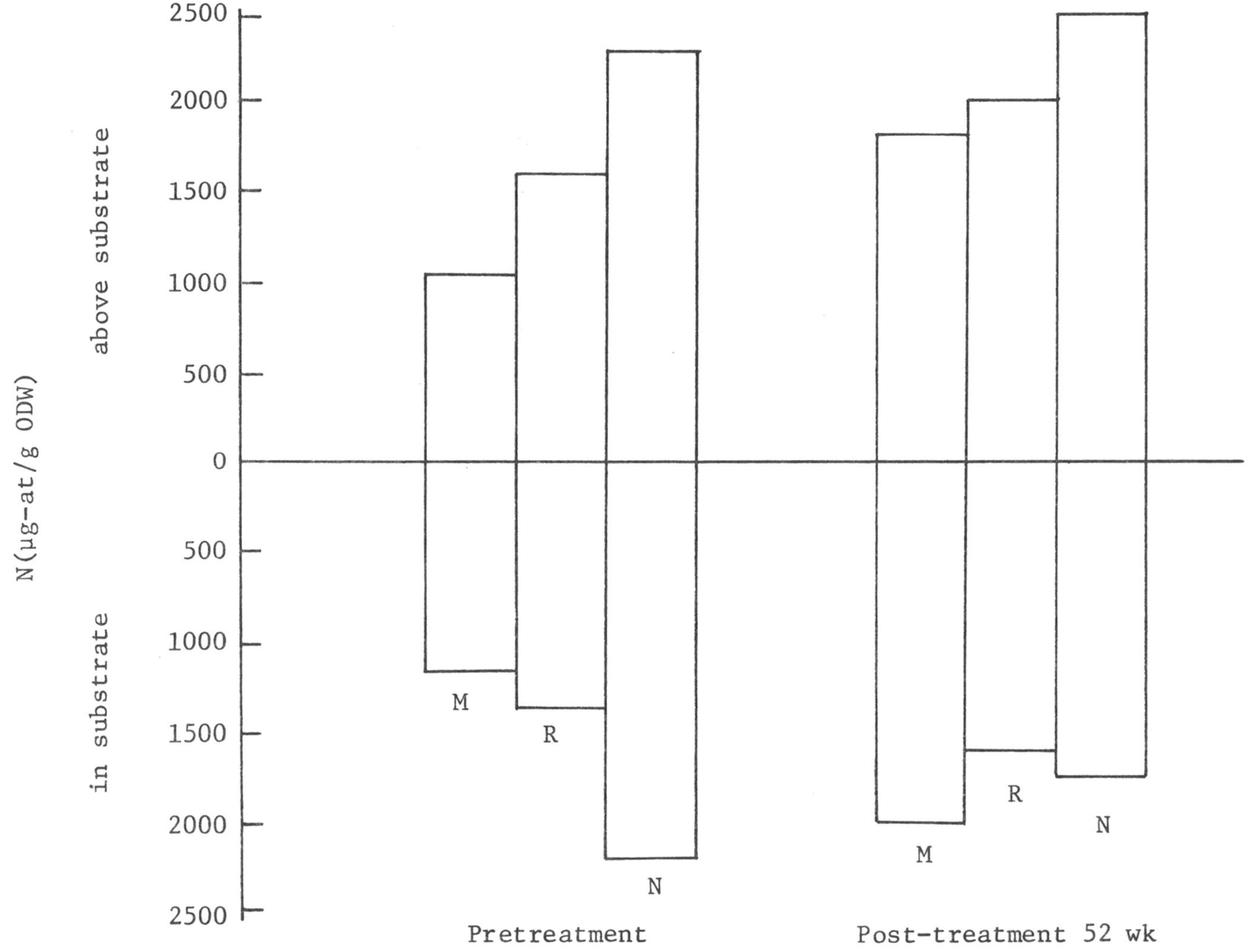

Fig. 6. Nitrogen concentration in M. spicatum (M), R. maritima (R) and N. guadalupensis (N) at Kitty Hawk Bay Treatment and Control Area I, summers 1974 and 1975.

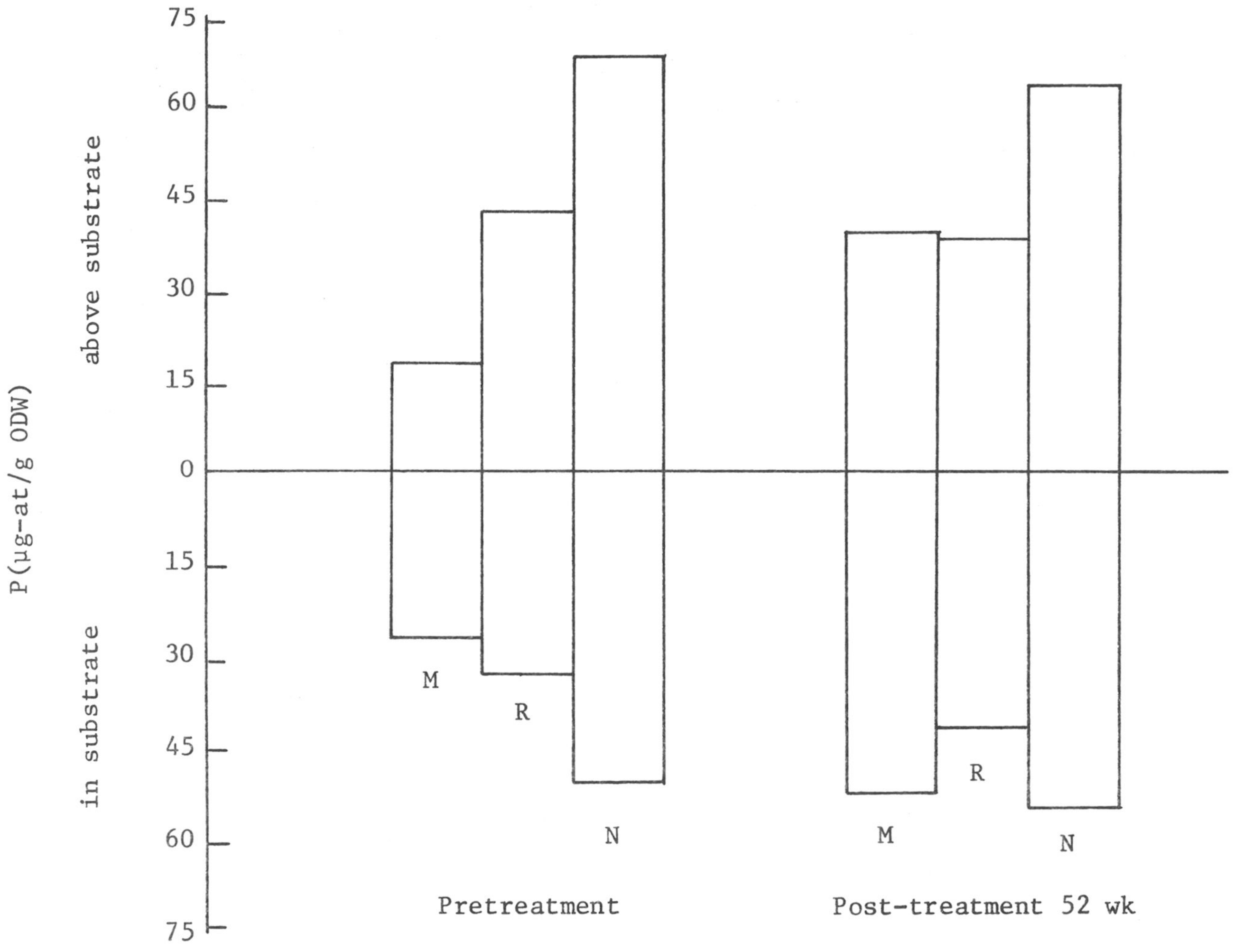

Fig. 7. Phosphorus concentration in M. spicatum (M), R. maritima (R) and N. guadalupensis (N), at Kitty Hawk Bay Treatment and Control Area I, summers 1974 and 1975.

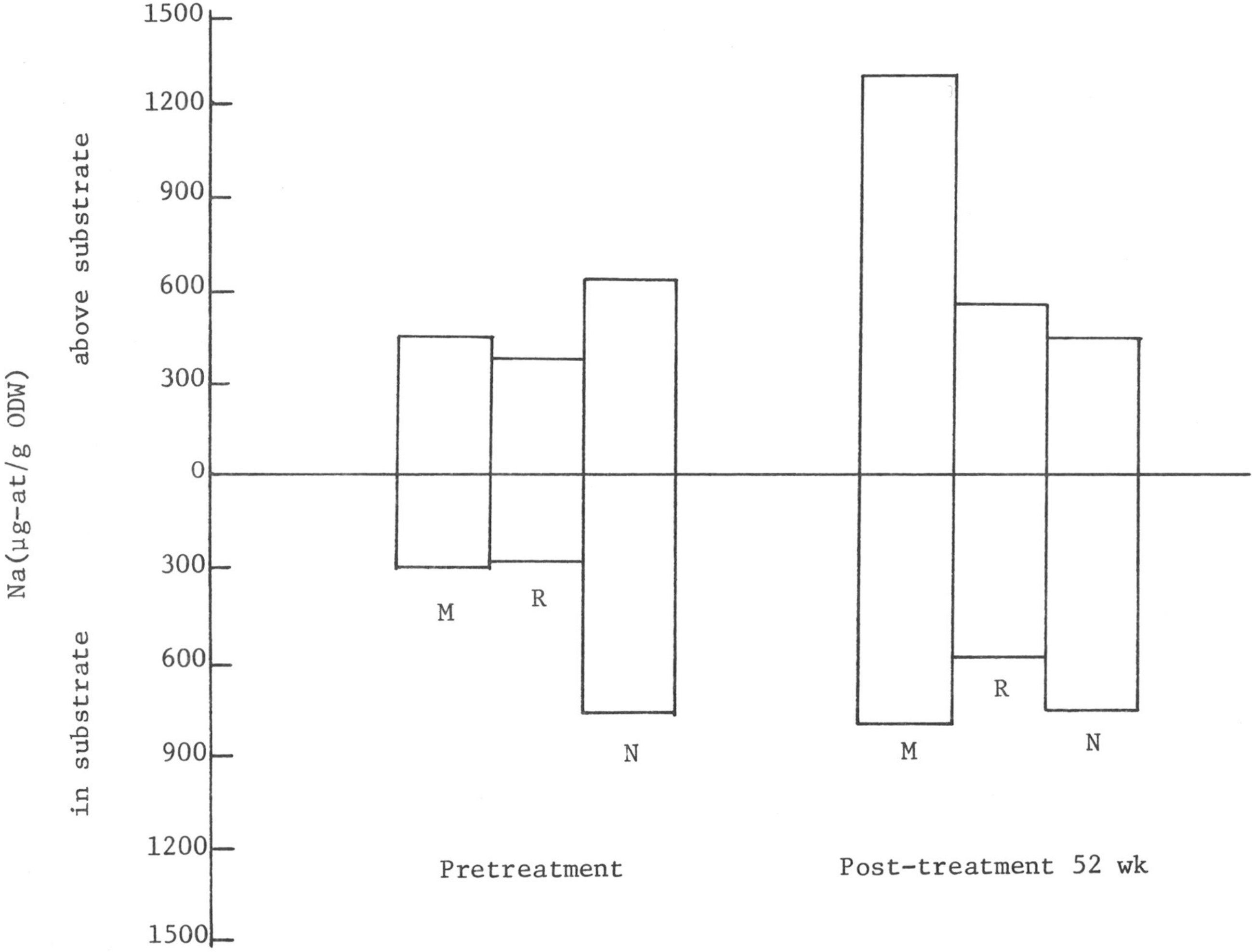

Fig. 8. Sodium concentration in M. spicatum (M), R. maritima (R) and N. guadalupensis (N), at Kitty Hawk Bay Treatment and Control Area I, summers 1974 and 1975.

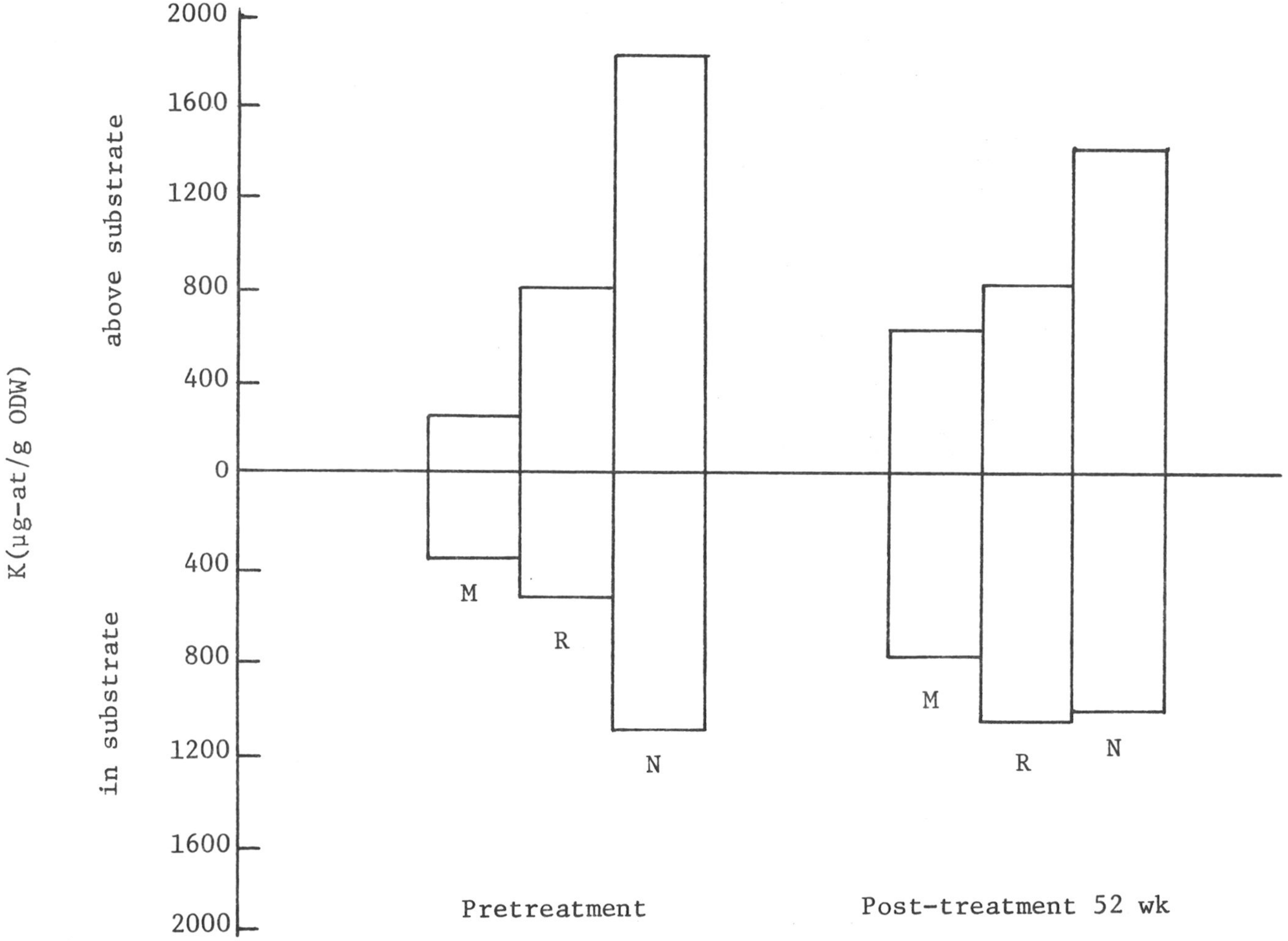

Fig. 9. Potassium concentration in _M_. _spicatum_ (M), _R_. _maritima_ (R) and _N_. _guadalupensis_ (N), at Kitty Hawk Bay Treatment and Control Area I, summers 1974 and 1975.

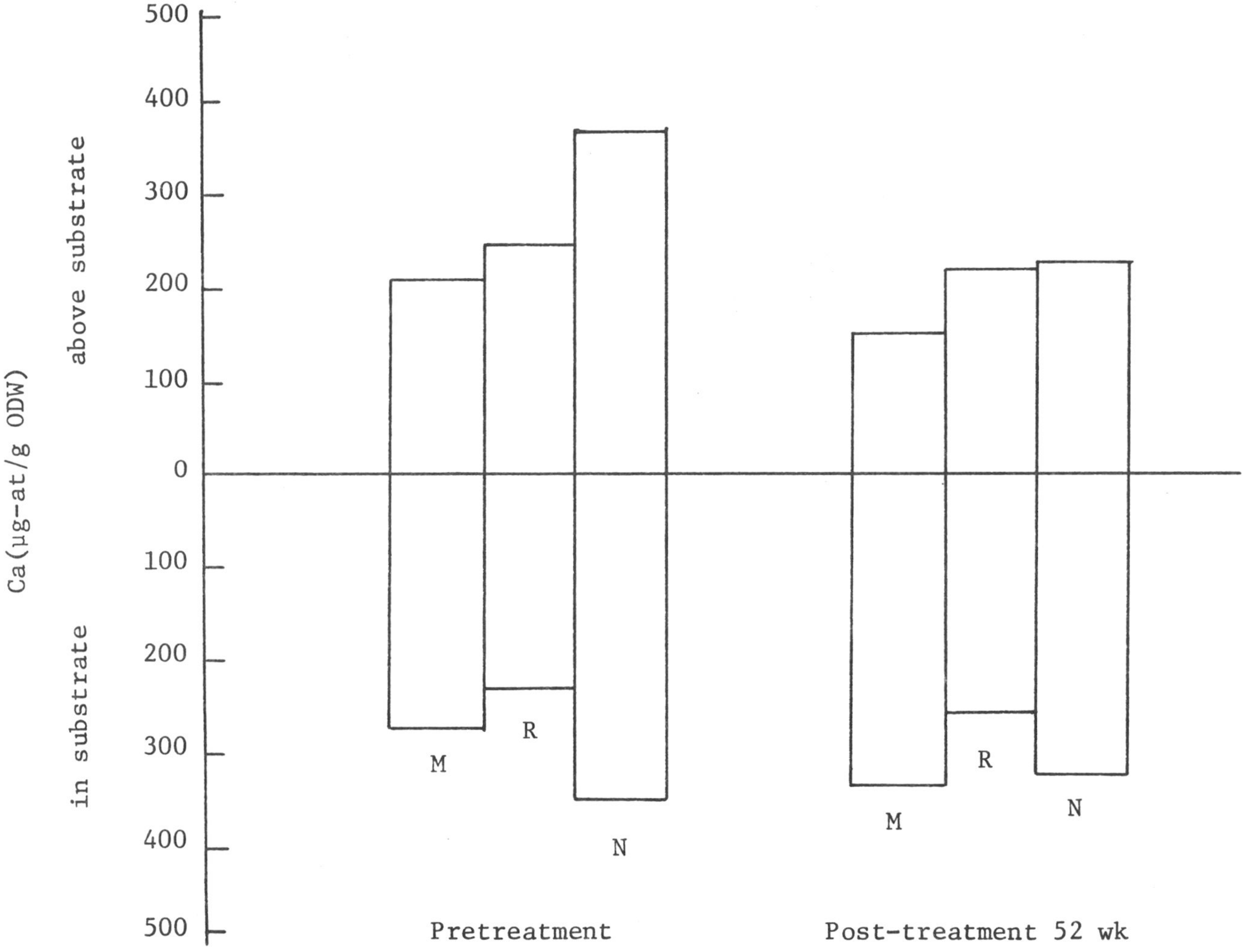

Fig. 10. Calcium concentration in M. spicatum (M), R. maritima (R) and N. guadalupensis (N), at Kitty Hawk Bay Treatment and Control Area I, summers 1974 and 1975.

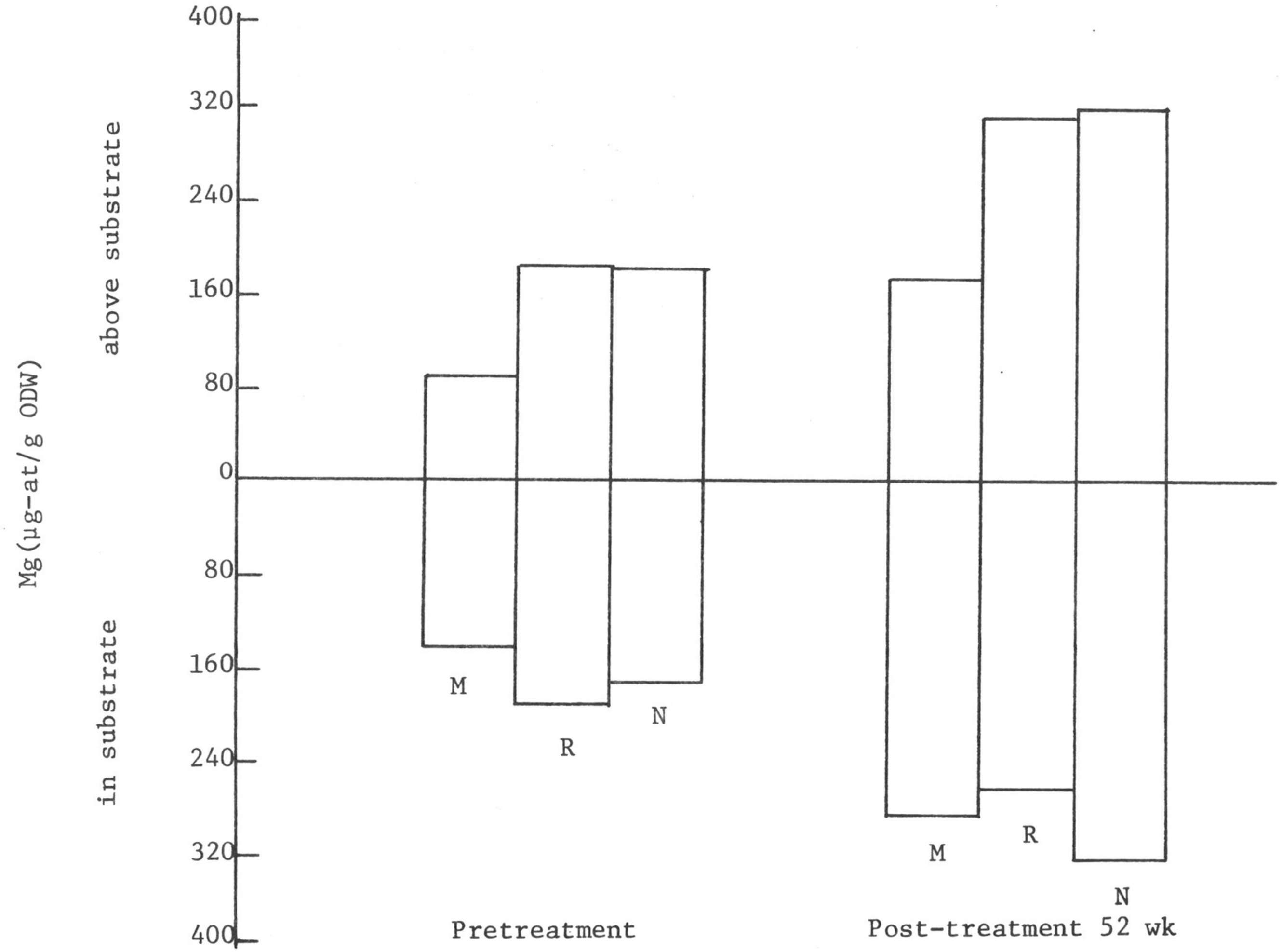

Fig. 11. Magnesium concentration in _M_. _spicatum_ (M), _R_. _maritima_ (R) and _N_. _guadalupensis_ (N), at Kitty Hawk Bay Treatment and Control Area I, summers 1974 and 1975.

Table 13. Ash and inorganic nutrients of *Myriophyllum* spp.

	Percent Dry Weight						
Species and Investigators	Ash	N	P	Na	K	Ca	Mg
Myriophyllum spicatum							
Nelson and Palmer (1938)	13.83	4.13	0.42	0.75	1.87	2.77	0.74
Umeda and Tamaki (1959)	10.03	1.51	0.05	-	1.18	1.31	-
Anderson et al. (1966)	-	3.00	0.40	1.20	2.70	0.35	0.05
Boyd (1968)	40.60	1.57	-	-	-	-	-
Reimer and Toth (1968)	-	2.89	0.50	1.01	1.68	1.31	0.35
Adams and McCracken (1974)	-	-	0.18	-	-	-	-
Gerloff (1975)	-	2.72	0.26	-	0.20	-	-
This study (1974)[a]	7.08	1.16	0.07	0.90	1.03	0.78	0.29
This study (1975)[a]	6.43	1.87	0.10	1.60	1.70	0.72	0.37
Myriophyllum exalbescens							
Daniel (1972)	-	2.34	0.41	0.89	1.71	3.15	0.80
Neel et al. (1973)	-	3.30	0.31	0.40	0.70	1.76	0.26
Myriophyllum spp.							
Schuette and Hoffman (1921)	20.72	3.00	0.51	-	-	3.04	-
Gerloff and Krombholz (1966)	-	2.42	0.35	-	-	-	-

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[a] Percent ash does not include acid insoluble residue.

Table 14. Ash and inorganic nutrients of *Potamogeton pectinatus*, *Vallisneria americana* and *Najas* spp.

Species and Investigators	Ash	N	P	Na	K	Ca	Mg
Potamogeton pectinatus							
Harper and Daniel (1934)	-	1.98	0.16	-	-	3.00	-
Allenby (1968)	-	-	-	-	-	3.20	-
Reimer and Toth (1968)	-	1.72	0.26	0.60	1.73	2.63	0.30
Adams et al. (1973)	-	-	0.30	0.20	1.33	0.86	0.38
Neel et al. (1973)	-	3.30	0.31	0.40	0.70	1.76	0.26
This study[a]	7.04	1.35	0.08	0.77	1.16	0.92	0.28
Vallisneria americana							
Schuette and Alder (1927)	25.18	1.89	0.23	0.48	4.55	5.80	1.12
Nelson and Palmer (1938)	15.64	2.42	0.21	2.39	6.77	1.55	0.76
Gerloff and Krombholz (1966)	-	2.88	0.42	-	-	-	-
Reimer and Toth (1968)	-	4.07	0.62	0.37	5.75	0.70	0.29
Neel et al. (1973)	-	3.40	0.35	0.37	2.80	1.77	0.26
Zamuda (1976a)[a]	29.00	2.80	0.60	3.10	5.80	0.80	0.70
This study (1975)[a]	17.89	2.01	0.11	1.72	4.41	0.91	0.36
Najas guadalupensis							
Boyd (1968)	18.70	3.65	-	-	-	-	-
Vicars (1976b)[a]	21.80	3.13	0.71	-	-	-	-
This study (1974)[a]	14.79	2.37	0.14	1.00	3.94	1.04	0.33
This study (1975)[a]	18.31	2.21	0.13	0.93	3.61	0.83	0.47
Najas flexilis							
Schuette and Alder (1927)	19.16	1.86	0.30	-	-	-	-
Najas spp.							
Reimer and Toth (1968)	-	1.79	0.31	0.75	1.83	14.7	0.61

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[a] Percent ash does not include acid insoluble residue.

authors cited. Nutrient levels were therefore expressed on a percent dry weight basis. Furthermore, sampling methods for the nutrient analysis varied with the investigator. For example, some used only the shoot system, stem fragments and/or growing tips, while others included the rhizome system in their analyses. Plant nutrient levels may also vary seasonally (Zamuda 1976b, Twilley 1976).

Although Kitty Hawk Bay _M_. _spicatum_ ranked low in nitrogen and phosphorus levels compared to _M_. _spicatum_ in other areas, it did contain those nutrients equal to or in excess of the critical concentration point for nitrogen (0.75%) and phosphorus (0.07%) as established for _M_. _spicatum_ by Gerloff (1975). The critical concentration is considered to be the minimum concentration of an element, or slightly less than the minimum concentration, in a plant which results in maximum plant growth (Gerloff 1975). Any amount above the critical concentration does not result in increased growth and is considered luxury accumulation.

A few cation concentrations found in Kitty Hawk Bay _M_. _spicatum_ were lower than those values for _Myriophyllum_ reported in other studies, particularly those of calcium (Table 7). The potassium level of Kitty Hawk Bay _M_. _spicatum_ (1.03% 1974 and 1.70% 1975) fell considerably above the critical concentration (0.35%) established by Gerloff. He suggested that potassium could be a key limiting nutrient in some situations.

Najas _guadalupensis_ from Kitty Hawk Bay had lower nitrogen (2.37% 1974, 2.21% 1975) and phosphorus (0.14% 1974, 0.13% 1975) levels than the same species reported by Vicars (1976b) from the Pamlico River N. C., where nitrogen and phosphorus levels were 3.13% and 0.71% (Table 14). Reimer and Toth (1968) found a lower nitrogen concentration (1.79%) in

Najas spp. from New Jersey, a higher phosphorus concentration (0.31%) and a much higher calcium concentration (14.7%) compared to a 1.04% calcium content in 1974 and a 0.83% calcium content in 1975 for Kitty Hawk Bay *Najas*.

As was the case with *M. spicatum* and *N. guadalupensis*, Kitty Hawk Bay *V. americana* ranked low in nitrogen (2.01% 1975) and phosphorus (0.11% 1975) when compared to *V. americana* reported in other areas (Table 14). Reimer and Toth (1968) found the highest nitrogen and phosphorus concentrations in *V. americana* from New Jersey (4.07% and 0.62%), while Zamuda (1976) reported *V. americana* nitrogen and phosphorus levels of 2.8% and 0.60% from the Pamlico River N. C. Gerloff and Krombholz (1966) determined the critical concentrations for nitrogen and phosphorus in *V. americana* to be 1.3% and 0.13% respectively. Kitty Hawk Bay *V. americana* exceeded the critical concentration for nitrogen, but fell below the critical concentration for phosphorus. This may be associated with the limited growth of *V. americana* in the Kitty Hawk Bay area. Table 8 indicates that cation concentrations in Kitty Hawk Bay *V. americana* were generally lower than cation concentrations reported for this plant from other areas.

Potamogeton pectinatus in Kitty Hawk Bay also had lower nitrogen (1.35% 1974) and phosphorus (0.08% 1974) levels than *P. pectinatus* studied in other areas. Harper and Daniel (1934) found *P. pectinatus* nitrogen and phosphorus levels in Oklahoma lakes to be 1.98% and 0.16%, and Neel et al. (1973) found nitrogen and phosphorus levels of *P. pectinatus* growing in Lake Sallie, Minnesota to be 3.3% and 0.31% (Table 14). Cation concentrations (especially calcium) in Kitty Hawk Bay *P. pectina-*

tus were lower than cation concentrations of P. pectinatus in other areas.

The mean concentrations of nitrogen and phosphorus for all species of submersed macrophytes found in Kitty Hawk Bay Treatment Area and Control Area II are given in Figs. 12 and 13. Species with the highest nitrogen levels were N. guadalupensis, V. americana and N. hyalina, followed by R. maritima and P. perfoliatus. Myriophyllum spicatum and P. pectinatus had the lowest nitrogen levels (Fig. 12). As for phosphorus, M. spicatum and P. pectinatus had the least, while N. guadalupensis and V. americana had the greatest amount. Potamogeton perfoliatus, R. maritima and N. hyalina accumulated intermediate amounts (Fig. 13).

Most of the submersed macrophytes in 1974 and 1975 accumulated more nitrogen and phosphorus in the shoots than in the rhizomes. Two exceptions to this general trend were M. spicatum, which accumulated less nitrogen and phosphorus in the shoots than rhizomes, and R. maritima, which accumulated less phosphorus in the shoots than rhizomes (Tables 10-12).

Cation concentrations in Kitty Hawk Bay plants in 1974 tended to be higher in shoots than in rhizomes, while this condition was reversed in the plants collected during the summer of 1975.

Inorganic Nutrients in the Waters of Kitty Hawk Bay and Southern Currituck Sound

Water samples were collected and analyzed for inorganic nutrients during the summers of 1974 and 1975 in all three treatment areas (Tables 15-18). The cations sodium, potassium, calcium and magnesium were found in ratios similar to those in sea water. The change in cation concentration from week to week appeared to be related to prevailing

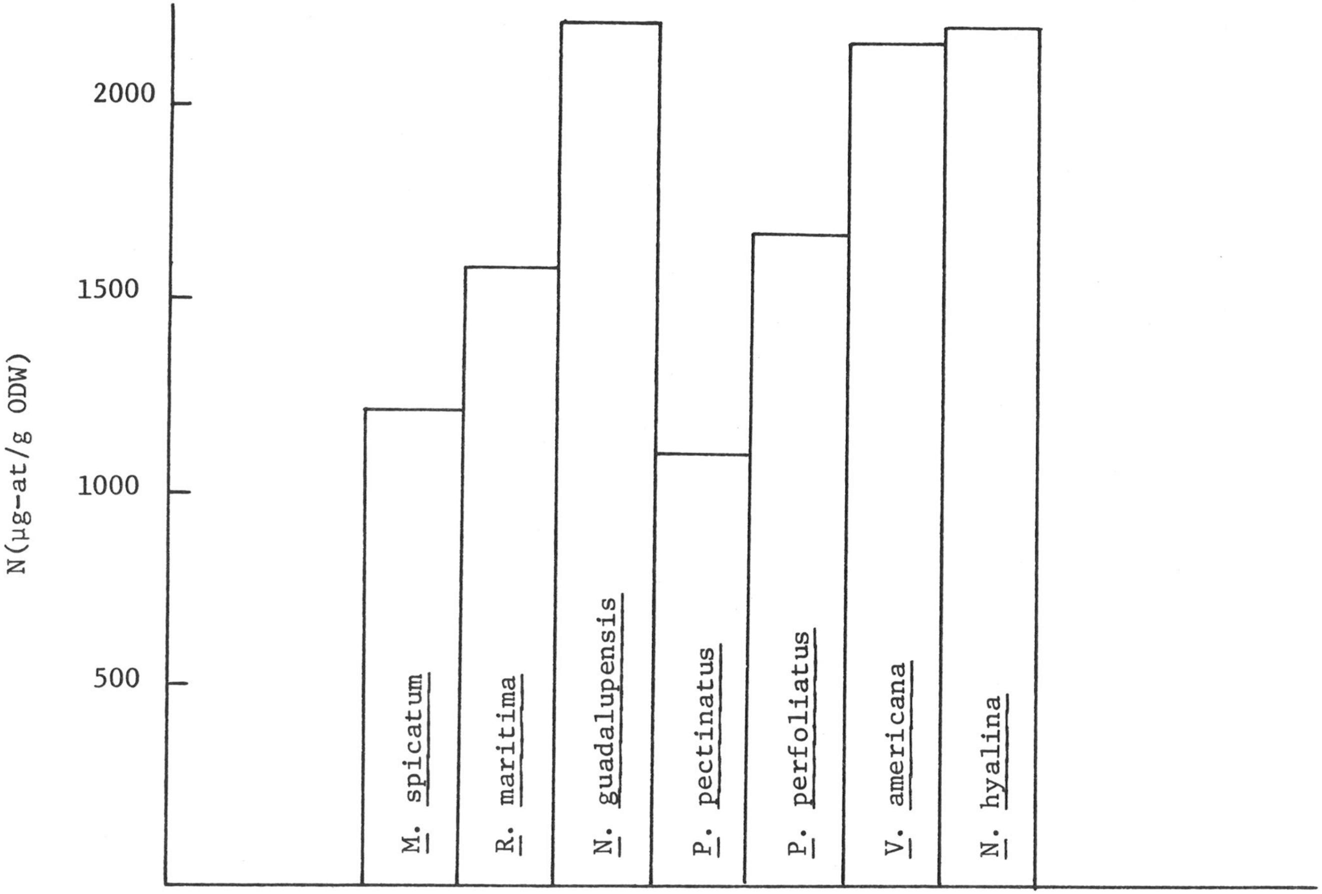

Fig. 12. Mean concentration of nitrogen in *M. spicatum*, *R. maritima*, *N. guadalupensis*, *P. pectinatus*, *P. perfoliatus*, *V. americana* and *N. hyalina* at Kitty Hawk Bay Treatment and Control Area II, summers 1974 and 1975.

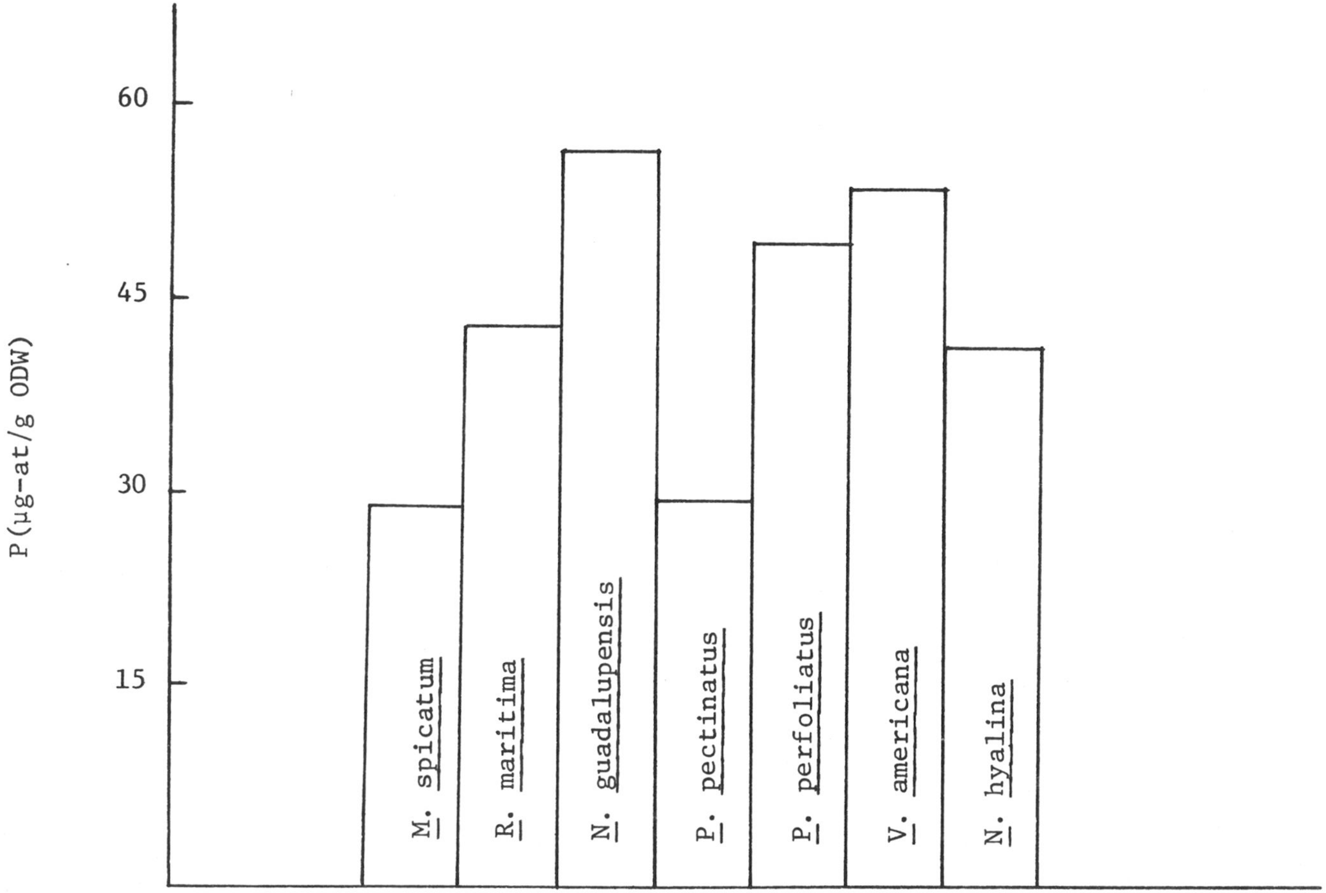

Fig. 13. Mean concentration of phosphorus in *M. spicatum*, *R. maritima*, *N. guadalupensis*, *P. pectinatus*, *P. perfoliatus*, *V. americana* and *N. hyalina* at Kitty Hawk Bay Treatment Area and Control Area II, summers 1974 and 1975.

Table 15. Inorganic nutrients (μg-at/l) in water at Kitty Hawk Bay, summers 1974 and 1975.

Date	Wind (mph)[a]	Na	K	Ca	Mg
		Treatment Area			
13 Jun 74	NW 10	23,490	627	762	2930
20 Jun 74	NE 15	23,050	599	750	2880
8 Jul 74	SW 10	22,620	499	625	2570
13 Jul 74[b]	NE 10	-	-	-	-
16 Jul 74	NE 10	23,490	499	712	2750
25 Jul 74	SW 10	26,100	538	762	2920
5 Aug 74	SW 20	43,500	875	625	5300
13 Aug 74	SE 10	24,790	435	500	2000
20 Aug 74	SE 5	19,570	422	612	2080
18 Jul 75	NW 5	59,810	1020	1050	5340
25 Jul 75	SW 15	69,600	1200	1220	5920
1 Aug 75	NE 5	17,400	461	725	750
12 Aug 75	NW 5	27,190	640	662	3250
		Control Area II			
13 Jul 74[b]	NE 10	-	-	-	-
25 Jul 74	SW 10	26,100	543	717	3250
5 Aug 74	SW 20	25,230	532	210	2840
13 Aug 74	SE 10	24,790	435	500	2000
20 Aug 74	SE 10	28,270	648	687	3590
18 Jul 75	NW 5	36,970	691	512	3130
25 Jul 75	SW 15	71,770	1150	1237	5840
1 Aug 75	NE 5	35,890	896	950	1460
12 Aug 75	NW 5	27,190	691	625	2170

[a] estimated velocity

[b] treatment date

Table 16. Inorganic nutrients (μg-at/l) in water at Martin Point and Point Harbor, summers 1974 and 1975.

		Nutrients			
Date	Wind (mph)[a]	Na	K	Ca	Mg
		Martin Point			
13 Jun 74	NW 10	26,100	366	525	1670
20 Jun 74	NE 15	8,700	358	538	1740
8 Jul 74	SW 10	17,400	397	575	2120
14 Jul 74[b]	NE 10	-	-	-	-
16 Jul 74	NE 10	18,270	410	625	2120
25 Jul 74	SW 10	19,140	363	575	1880
5 Aug 74	SW 20	26,100	538	225	3000
13 Aug 74	SE 10	19,570	422	612	2080
20 Aug 74	SE 5	18,700	538	562	2540
18 Jul 75	NW 5	16,310	333	300	1350
25 Jul 75	SW 15	51,110	947	925	4500
1 Aug 75	NE 5	9,790	307	350	500
12 Aug 75	NW 5	31,540	691	712	3130
		Point Harbor			
13 Jun 74	NW 10	20,440	461	525	2230
20 Jun 74	NE 15	20,440	481	512	2410
8 Jul 74	SW 10	16,530	358	268	1670
14 Jul 74[b]	NE 10	-	-	-	-
16 Jul 74	NE 10	14,790	230	170	1040
25 Jul 74	SW 10	20,010	410	412	2290
5 Aug 74	SW 20	32,190	627	287	3630
13 Aug 74	SE 10	16,960	248	212	1080
20 Aug 74	SE 5	14,350	346	290	1680
18 Jul 75	NW 5	40,240	768	687	3340
25 Jul 75	SW 15	29,360	563	550	2840
1 Aug 75	NE 5	20,660	512	500	830
12 Aug 75	NW 5	13,050	384	362	1670

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[a] estimated velocity

[b] treatment date

winds (Tables 15-16). A southwest wind moved brackish water from Oregon Inlet, northward into the Kitty Hawk Bay-lower Currituck Sound area which, in turn, caused an increase in cation concentrations in the water. A northeast wind moved fresh water southward from upper Currituck Sound which caused a decrease in the cation concentrations in the water of the Kitty Hawk Bay-lower Currituck Sound area.

Generally, cation concentrations appear to have been higher in the summer of 1975 than in the summer of 1974 in all of the study areas. This increase in water cation levels was reflected to some extent in the concentrations of cations in the submersed macrophytes during the two summers. In Kitty Hawk Bay Treatment Area, sodium, potassium and magnesium concentrations increased in both the shoots and roots from 1974 to 1975 in *M. spicatum* and *R. maritima* (Figs. 8, 9 and 11). Calcium decreased in the stems, but increased in the roots of the same two species (Fig. 10). *Najas guadalupensis*, however, decreased in sodium, potassium and calcium in the roots and shoots from 1974 to 1975 and increased in magnesium.

Tables 17 and 18 give nitrogen and phosphorus levels in the waters of Kitty Hawk Bay, Martin Point and Point Harbor during the study period. Nitrites decreased for several weeks following 2,4-D treatment, but recovered somewhat in the summer of 1975, except at Kitty Hawk Bay Treatment Area where the nitrite levels remained low. After a short decline, nitrates increased in the weeks following treatment and remained above pretreatment levels during the summer of 1975. Nitrogen concentrations were generally higher in submersed macrophytes in 1975 than in 1974, particularly in the shoots of *M. spicatum*, *R. maritima* and

Table 17. Nitrogen and phosphorus (µg-at/l) in water at Kitty Hawk Bay, summers 1974 and 1975.[a]

Dates	Nutrients					
	NO_2-N[b]	NO_3-N[b]	NH_3-N	Kjel N	Ortho P[b]	Total P
	Treatment Area					
13 Jun 74	0.79	<2.57	<0.71	286	0.13	2.71
20 Jun 74	0.71	<2.57	<0.71	257	0.32	<1.15
8 Jul 74	0.43	7.14	<0.71	86	<0.07	1.93
13 Jul 74[c]	-	-	-	-	-	-
16 Jul 74	<0.11	2.86	1.43	57	<0.07	<1.15
25 Jul 74	<0.11	2.86	2.14	200	<0.07	1.61
5 Aug 74	<0.11	7.86	<0.71	68	0.29	<1.15
13 Aug 74	0.14	5.71	<0.71	257	0.48	2.86
20 Aug 74	<0.11	-	<0.71	228	<0.07	<1.15
18 Jul 75	0.14	5.36	25.71	247	0.32	<1.15
25 Jul 75	<0.11	3.21	15.00	565	0.32	<1.15
1 Aug 75	0.14	3.21	14.28	655	<0.07	<1.15
12 Aug 75	<0.11	11.07	25.71	726	0.32	1.61
	Control Area II					
13 Jul 74[c]	-	-	-	-	-	-
25 Jul 74	<0.11	2.86	8.57	171	<0.07	1.61
5 Aug 74	<0.11	7.86	<0.71	71	<0.07	<1.15
13 Aug 74	0.14	5.71	<0.71	143	0.68	<1.15
20 Aug 74	<0.11	8.57	<0.71	183	<0.07	<1.15
18 Jul 75	<0.11	3.57	26.43	289	<0.07	<1.15
25 Jul 75	0.43	3.71	19.28	610	0.32	<1.15
1 Aug 75	0.57	4.86	16.43	751	0.32	<1.15
12 Aug 75	0.29	10.00	16.43	982	0.32	1.29

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[a] The limit of detection was calculated as 3 standard deviations of 10 replicates near the detection limit for the test.

[b] filtered samples

[c] treatment date

Table 18. Nitrogen and phosphorus (µg-at/l) in water at Martin Point and Point Harbor, summers 1974 and 1975.[a]

	Nutrients					
Dates	NO_2-N[b]	NO_3-N[b]	NH_3-N	Kjel N	Ortho P[b]	Total P
	Martin Point					
13 Jun 74	1.64	<2.57	<0.71	200	1.39	1.74
20 Jun 74	-	-	-	-	-	-
8 Jul 74	3.43	7.14	<0.71	86	0.30	2.10
14 Jul 74[c]	-	-	-	-	-	-
16 Jul 74	<0.11	2.86	1.43	57	<0.07	1.29
25 Jul 74	<0.11	2.86	2.86	143	<0.07	1.93
5 Aug 74	<0.11	10.71	<0.71	71	<0.07	<1.15
13 Aug 74	<0.11	5.71	<0.71	183	0.48	1.55
20 Aug 74	<0.11	10.71	<0.71	228	<0.07	<1.15
18 Jul 75	0.14	8.93	30.00	263	0.32	<1.15
25 Jul 75	0.64	6.07	15.00	494	0.32	<1.15
1 Aug 75	0.79	4.86	12.86	413	0.64	<1.15
12 Aug 75	0.71	4.86	23.58	642	0.32	1.29
	Point Harbor					
13 Jun 74	1.00	<2.57	<0.71	171	0.48	<1.15
20 Jun 74	-	-	-	-	-	-
8 Jul 74	1.07	7.14	<0.71	86	<0.07	2.90
14 Jul 74[c]	-	-	-	-	-	-
16 Jul 74	0.11	2.86	<0.71	57	<0.07	1.29
25 Jul 74	1.43	2.86	2.14	200	<0.07	1.61
5 Aug 74	<0.11	7.14	<0.71	88	<0.07	<1.15
13 Aug 74	<0.11	4.64	<0.71	68	0.68	<1.15
20 Aug 74	<0.11	8.57	<0.71	166	<0.07	<1.15
18 Jul 75	<0.11	3.21	26.43	327	0.32	<1.15
25 Jul 75	0.29	5.36	14.28	655	0.64	<1.15
1 Aug 75	0.57	4.86	17.86	436	1.29	2.58
12 Aug 75	0.36	4.86	29.28	643	<0.07	1.61

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[a] The limit of detection was calculated as 3 standard deviations of 10 replicates near the detection limit for the test.

[b] filtered samples

[c] treatment date

N. guadalupensis (Fig. 6). Ammonia increased from <0.71 μg-at/l before treatment to 1.43-2.86 μg-at/l 2 weeks after treatment. Increased ammonification during submersed macrophyte decay may have caused the higher ammonia levels. Ammonia dropped to <0.71 μg-at/l during the last 3 weeks of the 1974 study concomitant with a phytoplankton bloom in the same period. In the summer of 1975, however, ammonia increased to over 10 times its highest level in 1974 in all three areas. At one point in 1975 ammonia was measured at 30 μg-at/l at Martin Point. Nitrates are released typically from estuarine sediments rather than ammonia (Harrison and Hobbie 1974), yet, Hobbie et al. (1972) reported that ammonia is released from the sediments of the Pamlico River estuary, N. C. A similar situation may occur in Kitty Hawk Bay. Once the biomass was removed by 2,4-D treatment, the increased wave action and turbulence may have caused a greater exchange of ammonia between the sediments and the water column than had existed before treatment. Hobbie and Smith (1975) and Hobbie (1974) reported high ammonia levels (20-60 μg-at/l) in the water column in the Neuse River estuary and the Pamlico River estuary following a hurricane in 1971. The increased ammonia levels in 1975 could also be related to increased ammonification and/or nitrification inhibition associated with ecosystem perturbation. Also, an ammonia "sink" was removed from the ecosystem with the death of the macrophytes.

Kjeldahl (total) nitrogen increased 2 to 4 fold in the study areas from 1974 to 1975. Since Kjeldahl nitrogen data was based on unfiltered samples, much of this difference may have been due to an increase in particulate suspended material from the sediments, which occurred following M. spicatum deterioration.

Ortho-phosphate was below limits of detection (<0.07 μg-at/l) for 2 to 3 weeks following treatment in all of the study areas, but increased in the summer of 1975 (0.32-1.29 μg-at/l). Despite increased phosphate in the water column in 1975, phosphorus levels in R. maritima and N. guadalupensis remained fairly constant. Phosphorus levels in M. spicatum, however, increased in both the shoots and rhizomes from 1974 to 1975 (Fig. 7). Phosphate levels in Kitty Hawk Bay were considerably lower than phosphate levels of 1-7 μg-at/l found by Hobbie and Smith (1975) in the Neuse River estuary, N. C., or the phosphate levels of 1-100 μg-at/l reported by Hobbie et al. (1972) in the Pamlico River estuary, N. C. Total phosphorus decreased during post-treatment weeks to levels below limits of detection (<1.15 μg-at/l). One year after 2,4-D application, total phosphorus levels were still lower than levels found in the pre-treatment study.

SUMMARY AND CONCLUSIONS

Environmental Impact of 2,4-D Application

Application of the herbicide 2,4-D BEE on the *M. spicatum* beds in Kitty Hawk Bay and southern Currituck Sound distinctly altered the submersed macrophyte communities in the treated areas. A model for recovery of the submersed macrophyte community in Kitty Hawk Bay, based on general observations and data, is shown in Fig. 14.

Two weeks after treatment, *M. spicatum* had deteriorated completely with a sharp decrease in density and biomass of native submersed macrophytes. Turbidity increased following macrophyte decline. This was related, at least in part, to wave induced resuspension of sediments. A seventh post-treatment week cyanophytoplankton bloom contributed to the already high water turbidity. *Myriophyllum spicatum* in areas well away from the treated zone began to deteriorate during the eighth post-treatment week. An algal-like slime was observed on *M. spicatum* stems in the areas which experienced delayed deterioration. Steenis and Stotts (1965) and Rawls (1975) reported that algal growths on *M. spicatum* may be significant in delayed *M. spicatum* kills, since applied herbicides may accumulate in the growth with release later. Wojtalik et al. (1971) found that plankton in TVA reservoirs sorbed large amounts of 2,4-D and retained it for extended periods. Such 2,4-D dynamics and/or high turbidity may have been factors in the delayed *M. spicatum* die-back at Kitty Hawk Bay.

One year after herbicide application, the Kitty Hawk Bay treatment area was covered with a dense bottom mat of the charophyte *Nitella*

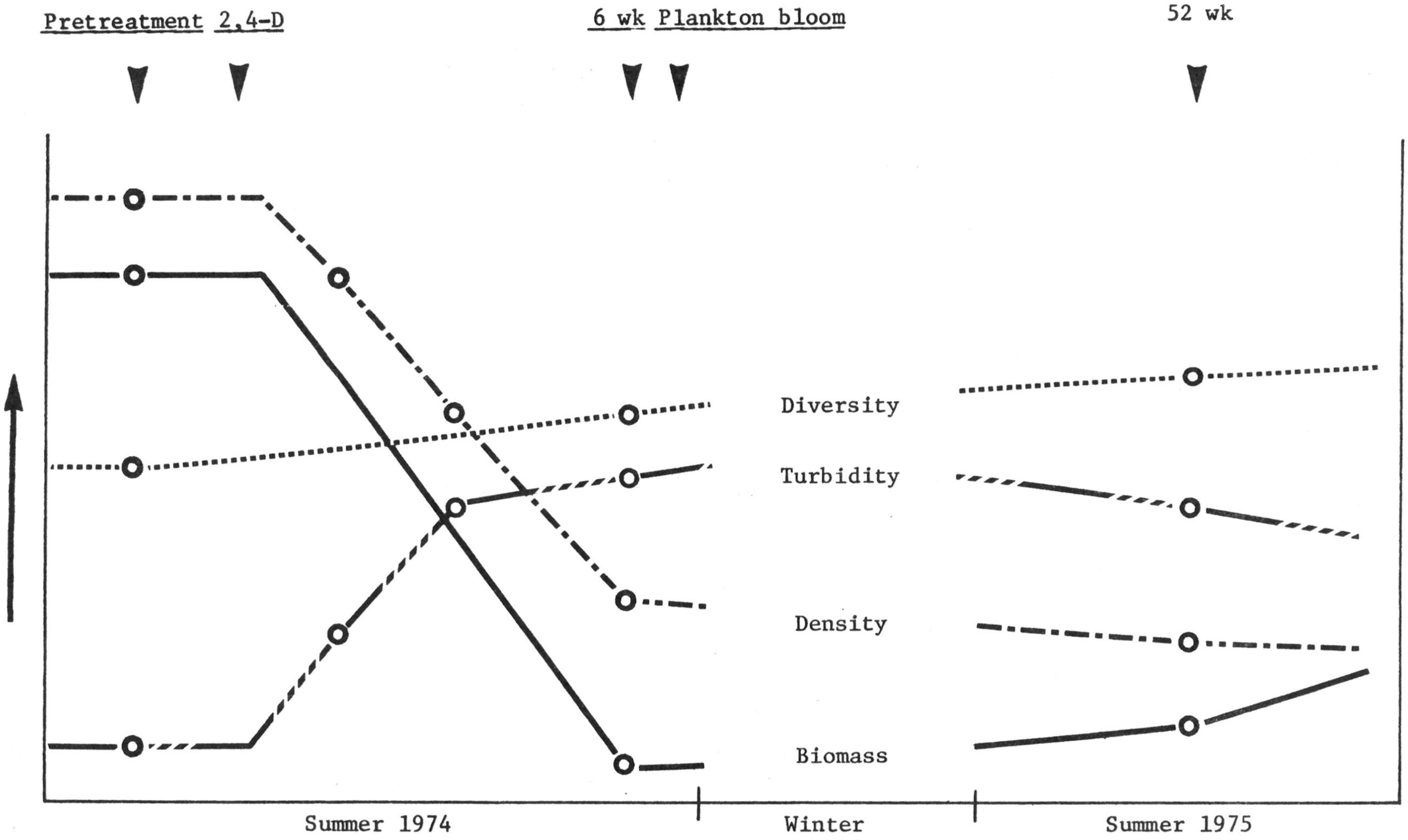

Fig. 14. A model for the recovery of the submersed macrophyte community in Kitty Hawk Bay, based on general observations and data, summers 1974 and 1975.

hyalina. This accounts for the increase in macrophyte biomass shown in the model. This "rooted" alga had been observed near the study plots during the pretreatment collection period. The model also indicates a low 52 week post-treatment macrophyte density compared with pretreatment values. Because of the fragile structure and twisted shoot growth of *N. hyalina* stem counts were impossible. Water turbidity had decreased by the summer of 1975. Diversity increased slightly during the 1974 and 1975 post-treatment study periods. An increase in available nutrients and lower light intensities, due to roiled sediments, may have led to the establishment of *N. hyalina* in the Kitty Hawk Bay treatment area. The *N. hyalina* "stems" were only 5 to 10 cm above the substrate; therefore, wave dampening effectiveness was limited. Yet, these charophyte beds may function as a "pioneer" stage in submersed macrophyte community succession by competitive inhibition of phytoplankton blooms through nutrient utilization and by reduction of water turbidity through plankton reduction and substrate stabilization. Young stems of *M. spicatum* and native vascular species were observed growing in the *N. hyalina* beds. Recovery of the treated area in Kitty Hawk Bay to a *M. spicatum* dominated community closely resembling the pretreatment macrophyte community should occur 2 to 3 years after 2,4-D application.

Myriophyllum spicatum as a Low Phosphate Plant

Ortho-phosphate levels in Kitty Hawk Bay were low, being below the limits of detection in seven of the 12 water sample periods in the summer of 1974. The lowest measured level was <0.07 μg-at/l and the highest was 0.68 μg-at/l. Similar low levels were found at Martin Point and Point Harbor. Phosphate levels in all of these areas in the summer of 1975 were somewhat higher.

Phosphorus levels in *M. spicatum*, *R. maritima* and *N. guadalupensis* before 2,4-D treatment and in scattered plants in the treated area one year later are shown in Fig. 7. *Myriophyllum spicatum* was dominant and thriving with lower levels of plant phosphorus than the other two species which had much lower biomass. The phosphorus levels of *M. spicatum* were closer to those of the other two species in 1975 when phosphate levels were somewhat higher in the water column and biomass of all species was low. *Myriophyllum spicatum*, then, was the dominant where phosphate levels, at least in the water column, were low and predominated with significantly lower plant phosphate levels. This suggests that *M. spicatum* predominance may be due, in part, to lower critical tissue levels of phosphorus in reference to requirements for survival and growth. The comparatively low critical tissue concentration of phosphorus for *M. spicatum* (0.07%), reported by Gerloff (1975) is consistent with the Kitty Hawk Bay studies. These observations suggest that *M. spicatum* is a "low phosphate plant."

Gerloff (1975) also studied laboratory growth in mixed cultures at high and extremely high phosphate levels. *Myriophyllum spicatum* competed somewhat better than *Elodea occidentalis* at the lowest of the two phosphate levels. *Myriophyllum spicatum* grew more poorly than *Elodea canadensis* and *Potamogeton crispus* when pond water was supplemented with phosphate in the laboratory (Mulligan and Baranowski 1969). In further research in outdoor cement ponds, Mulligan et al. (1976) found that *Chara* and *M. spicatum* were eliminated in 1969 in ponds fertilized in 1968 while *E. canadensis* and *P. crispus* flourished. These controlled studies indicate that *M. spicatum* does poorly under high nutrient conditions generally and under high phosphate conditions specifically. Stanley (1970) pointed out that *M. spicatum* was less dense in the TVA area where ambient phosphate was higher and he suggested that the phosphate levels may affect distribution of the species. Stanley and Goode (1972) found that high levels of phosphate actually inhibited root growth of *M. spicatum* cuttings.

The *M. spicatum* infestation of the Kitty Hawk Bay - southern Currituck Sound area may be related to low available phosphate. However, optimum *M. spicatum* growth may be triggered by a combination of nutrient conditions. Stanley and Goode (1972) reported that the micronutrient borate might stimulate *M. spicatum* growth in low phosphate areas of TVA lakes. More research is needed on the limiting nutrients of *M. spicatum* growth, in addition to a continued study of water and sediment nutrients in the Kitty Hawk Bay-Currituck area. Perhaps ecosystem modification in the Kitty Hawk Bay-Currituck Sound area will shed further light on the phosphate - *M. spicatum* growth relationship.

Myriophyllum spicatum Biomass and 2,4-D

Application of 2,4-D BEE at 112 kg ae/ha on *M. spicatum* communities in the Kitty Hawk Bay-southern Currituck Sound area appears to yield excellent short-term (1-2 year) control of *M. spicatum*. In areas where root-rhizome destruction is complete, *M. spicatum* control may last as long as three growing seasons. Under certain wind conditions in Kitty Hawk Bay, 2,4-D BEE can directly kill up to twice the target area. When large areas (300 ha) of Kitty Hawk Bay *M. spicatum* are destroyed by herbicide, adjacent *M. spicatum* stands may have delayed die-backs. Factors related to increased water turbidity may be responsible for the delayed deterioration. Long-term control with herbicide is unlikely because of the vast stands of *M. spicatum* in the area. These beds are the source of vegetative fragments which can lead to early reinfestation of a treated area. Apparently 2,4-D at 112 kg ae/ha is potent enough to severely damage native submersed macrophytes found in the Kitty Hawk Bay-southern Currituck Sound area. It is also apparent that *M. spicatum* will outcompete vascular native submersed macrophytes in an area denuded following 2,4-D treatment. However, as reported for the Kitty Hawk Bay treatment area, native charophytes may be "pioneer" species in community succession, eventually replaced by vascular macrophytes dominated by *M. spicatum*.

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