

Arch. Hydrobiol.	85	4	437-447	Stuttgart, Mai 1979
------------------	----	---	---------	---------------------

615

## Distribution potential of *Myriophyllum spicatum* (Angiospermae, Haloragidaceae) in softwater systems<sup>1</sup>

By JOHN P. GIESY, Jr. and LAURA E. TESSIER

With 6 tables in the text

### Abstract

The distribution potential of the Eurasian water milfoil (*Myriophyllum spicatum* L.) in acid, softwater systems was determined, using reciprocal transplants of sediment from 3 ponds on the Savannah River Project. Actively growing vegetative tips were suspended over sediment in plastic growing pans and rooting potential and growth measured at each station.

Par Pond, a reservoir with higher pH and alkalinities than surrounding areas due to Savannah River water inputs, has previously been colonized by *M. spicatum*. When transplanted to two other softwater ponds with total alkalinity values between 5 and 6 mg/L CaCO<sub>3</sub>, *M. spicatum* rooted and grew in one pond but not in the other with a similar pH and calcium regime. *M. spicatum* rooted and grew under pH, alkalinity and calcium conditions far below its reported tolerance. Differences in rooting and growth potential were not due to sediment differences.

### Introduction

Eurasian water milfoil (*Myriophyllum spicatum* L.) is a circumboreal aquatic angiosperm (CLAPHAM et al., 1952). *Myriophyllum* taxonomy is complicated by the strong resemblance between *M. spicatum* and *M. exalbescens* FERN, which are thought, by certain investigators, to represent the same species (MASON, 1957). Further confusion stems from a discovery of plants possessing characteristics of both species (PATTEN, 1954). AIKEN<sup>2</sup> (personal communication) concludes that *M. exalbescens* has occurred in both North America and Europe since at least the last century and has successfully produced hybrid plants (*M. spicatum*, ♀; *M. exalbescens*, ♂).

<sup>1</sup> This research was supported by Contract EY-76-C-09-0819 between the University of Georgia and the U. S. Energy Research and Development Administration and Grant EPP 75-04436 between the University of Georgia and NSF. We are indebted to L. BRIESE for field assistance and A. HARMON for assistance with laboratory analyses. J. GRACE, L. TILLY, J. WIENER and K. McLEOD kindly commented on the manuscript.

<sup>2</sup> S. AIKEN, Dept. of Botany, University of Minnesota, St. Paul, Minnesota.



BRISTOW<sup>3</sup> (personal communication) found *M. spicatum* grown in culture from axenic seeds was morphologically different from plants growing in nature and very similar in appearance to *M. exalbescens*. A more complete discussion of this controversy is given by BERGQUIST (1970). WOODENHOUSE (1933) states that *M. spicatum* has been present on the North American continent since the late Eocene while STANLEY et al. (1970) report that the species has been present in the United States only since the nineteenth century. The plants studied here will be referred to as *M. spicatum*. The distribution of *M. spicatum* has recently increased greatly. Eurasian water milfoil has rapidly dispersed throughout the northern and midwestern United States and is well established in many states (CROWELL et al., 1967). *M. spicatum* has caused serious problems in Tennessee Valley Authority reservoirs since the early nineteensixties and is established in Texas and Louisiana (SMITH et al., 1963). In the past 20 years *M. spicatum* has become widespread in the Chesapeake Bay area (STEENIS and STOTTS, 1963) and is expanding its range into the southeastern coastal United States (NICHOLS and MORI, 1971) with infestations occurring in N. Carolina, Georgia and Florida within the last 10 years (CROWELL et al., 1967). *M. spicatum* was first reported in Par Pond, on the U. S. Energy Research and Development Administration's Savannah River Plant in S. Carolina, in 1971 and has subsequently become the dominant macrophyte there.

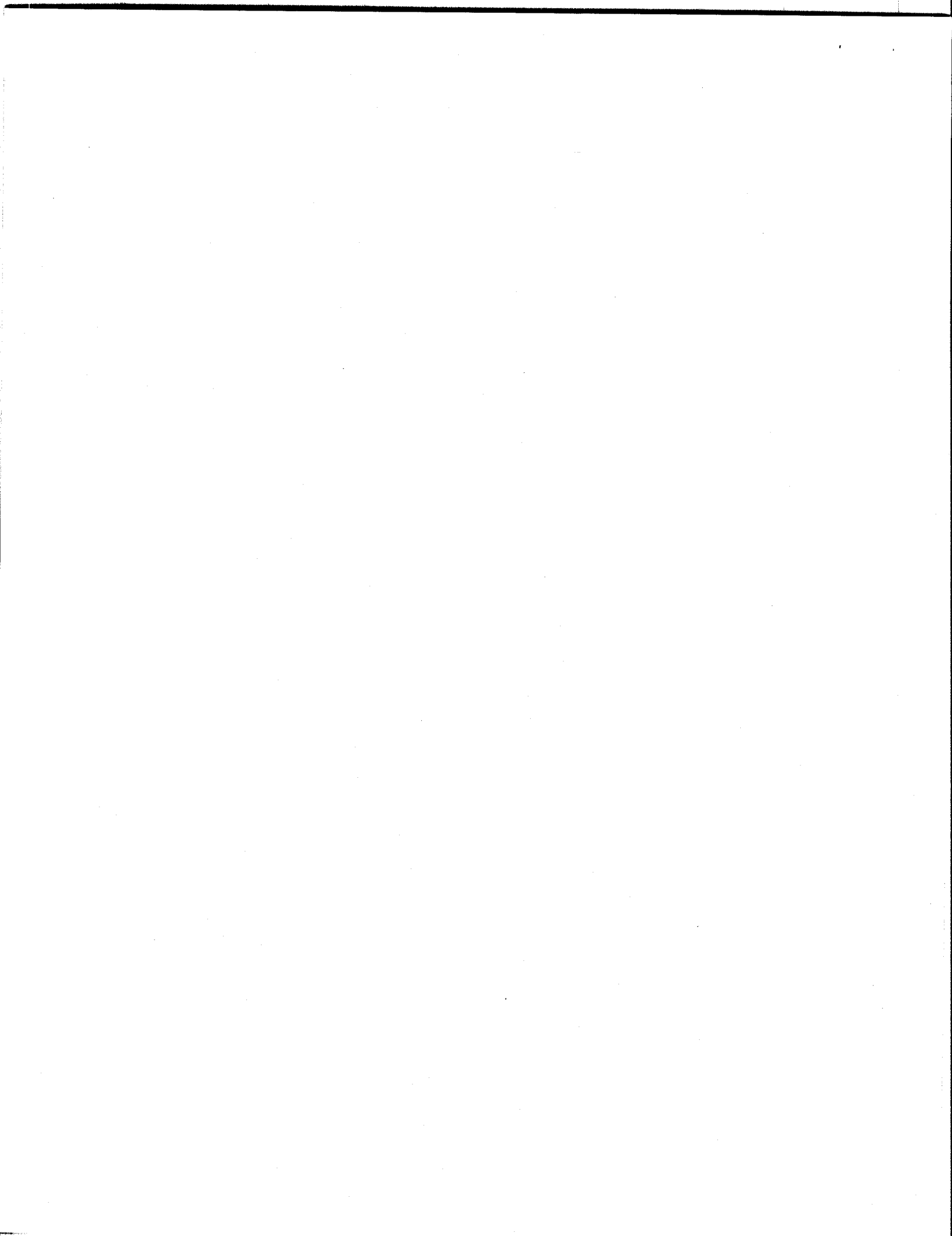
*M. spicatum* L. and *M. exalbescens* FERNALD are classified as calciphilous macrophytes (HUTCHINSON, 1970, 1975). STANLEY (1970) reported that Ca was important for  $\text{PO}_4^{3-}$  uptake and maximum photosynthesis and growth, and STEEMANN-NIELSEN (1947) indicated *M. spicatum* will not grow in small ponds of alkalinities less than  $20 \text{ mg} \cdot \text{l}^{-1} \text{ CaCO}_3$  and that low calcium content may limit growth; and SPENCE (1967) reported that *M. spicatum* distribution is directly correlated with increasing alkalinity.

This study was conducted to determine the potential for *M. spicatum* colonization in southeastern softwater lacustrine systems. This was accomplished by reciprocal transplants of sediments and introduction of *M. spicatum* into the water column over each soil type. Because characteristics of both sediments and water may limit submerged aquatic macrophyte distribution (MISRA, 1968; HUTCHINSON, 1970). sediments and water were characterized in an attempt to determine parameters affecting distribution.

### Methods

**Study Sites.** Four stations were chosen to give a range of physical and chemical parameters and *M. spicatum* populations. Two stations were established on Par Pond, where *M. spicatum* occurs and one each on Pond B and Upper

<sup>3</sup> J. M. BRISTOW, Dept. of Biology, Queens University, Kingston, Canada.



Twin Pond, where *M. spicatum* does not occur. Par Pond, constructed in 1957—1958 as a cooling reservoir for production reactors has received water from the Savannah River since that time, resulting in higher hardness and alkalinity values than those of adjacent ponds (Table 1). Par Pond consists of 3 arms and covers 1069 ha with an average depth of 6.1 m. The Beyers Bay station (BB), located in the middle arm is subject to temperature elevation by reactor effluent, while temperature at the Limnology Lab station (LL) is unaffected. Pond B (PB) and Upper Twin Ponds (TP) are softwater low pH systems (Table 1). Twin Ponds, a small farm pond, is highly colored and turbid, while PB is not. Pond B was constructed in 1961 and received reactor cooling water between 1961 and 1964. There is little surface input into the pond and the only significant source of water is rainfall.

Table 1. Mean chemical and physical characteristics.

	B Pond	Twin Ponds	Limnology Lab	Beyer's Bay
Depth (cm)	48.3—142.0	50.8—73.7	50.8—60.1	68.6—96.5
Eh (mv)	280	345	310	310
pH	6.0	5.2	7.7	8.2
Total Alkalinity (ppm CaCO <sub>3</sub> )	5.2	6.4	18.6	13.8
Hardness (ppm CaCO <sub>3</sub> )	6.0	4.0	15	13
Ca (ppm)	0.6	0.7	3.6	3.2
Mg	0.43	0.38	1.05	1.05
Na (ppm)	2.2	1.8	6.1	5.9
K (ppm)	0.4	0.1	1.1	1.4
Ca + Mg	2.52	1.76	1.55	1.71
Na + K	0.09	0.73	0.05	0.20
Total Fe (ppm)	10	20	60	60
Specific Conductance ( $\mu$ mho/cm)	0.03	0.02	0.03	0.04
Total PO <sub>4</sub> (ppm)	0.03	0.02	0.02	0.02
Dissolved PO <sub>4</sub> (ppm)	27.5	7.5	37.5	45.0
Cl <sup>-</sup> (ppm)	0.08	0.07	0.04	0.05
NH <sub>4</sub> <sup>+</sup> + NH <sub>3</sub> <sup>o</sup> (ppm)	0.07	0.26	0.11	0.19
NO <sub>2</sub> <sup>-</sup> + NO <sub>3</sub> <sup>-</sup> (ppm)	8.0	18.0	13.0	14.0
Total Organic Carbon (ppm)				

**Analytical Methods.** Water samples were collected monthly, while sediment samples were collected when the study was initiated. Dissolved oxygen, redox potential, pH, conductivity and temperature were determined, using a Hydrolabs Surveyor Model 6D. Iron (Fe), magnesium (Mg), and calcium (Ca) were determined by flame atomic absorption spectroscopy. Sodium (Na) and potassium (K) were determined using flame photometry. Water samples were filtered through acidwashed 0.45  $\mu$ m membrane filters in the field. Filtered and unfiltered fractions were digested using H<sub>2</sub>SO<sub>4</sub> and K<sub>2</sub>S<sub>2</sub>O<sub>8</sub> and total dissolved orthophosphate (PO<sub>4</sub>) determined using the ascorbic acid method (APHA, 1976). Total hardness and alkalinity were measured titrimetrically in the field using EDTA and H<sub>2</sub>SO<sub>4</sub> respectively. Chlorides (Cl<sup>-</sup>) were determined using the



mercuric nitrate method (APHA, 1976). Total organic carbon concentrations in water were measured, using a Beckman Model 915 carbon analyser. Determinations of  $\text{NH}_4^+ + \text{NH}_3^0$  and  $\text{NO}_2^- + \text{NO}_3^-$  were made by the Athens, Ga. Environmental Research Laboratory of the U. S. Environmental Protection Agency. Recording thermographs, placed at each station provided a continuous temperature record. Percent carbon in the sediments was determined using a Leco total carbon analyzer. Sediment pH values were measured in the field, using a glass electrode. Ammonium acetate extractable  $\text{PO}_4^{3-}$  was determined by the ammonium molybdate-stannous chloride method (CHAPMAN & PRATT, 1961). Nitrate,  $\text{NO}_2^-$ , and  $\text{NH}_4^+$  were measured by the method of BLACK et al. (1965). Sulfide concentrations in sediments were determined using the dithizone method (GLAMMATTEO, 1975). Sediment samples were dried and separated into 7 particle size fractions between 0.105  $\mu\text{m}$  and 2.36  $\mu\text{m}$  using standard sieves.

**Transplant Studies.** To distinguish between sediment and water column effects on *M. spicatum* colonization and growth, reciprocal transplants of sediments from each of the 4 stations were made during the summer of 1975. Sediments collected from each study area were autoclaved and placed in 27 × 27 × 6 cm plastic growing pans. Five pans of each sediment type were placed at each of the 4 stations. Ten cm lengths of vegetative *M. spicatum* stem tips were harvested from near the BB station and attached to wooden applicator sticks with 20 cm of monofilament line. Three stem tips were suspended above each pan by inserting the applicators into the sediments and allowing the buds to float in the water column above the sediment of interest. Plants were put in place on 5 June 1975 and determinations of rooting success and growth were made at 5-day intervals.

After 14 and 28 days the total number of plants successfully rooting in each sediment type and location were tested. Results of 5 replicates were pooled and differences between sediment type and location tested using the Friedman nonparametric 2-way test (CONOVER, 1971). Growth was determined *in situ* and reported as changes in total length. Growth data were tested with a two-way analysis of variance and multiple comparisons made, using Tukey's w-procedure.

## Results

The chemical parameters at the four stations were relatively constant across season within location. Sediment characteristics varied from station to station (Tables 2 and 3). Beyers Bay sediment had the highest organic carbon content but the lowest extractable  $\text{PO}_4^{3-}$  and Kjeldahl-N content. Pond B sediment contained the highest concentrations of Kjeldahl-N and  $\text{NH}_4^+$ . Sediment from LL was intermediate in all parameters measured except % organic carbon. This sediment was composed of sand and clay. The Kjeldahl-N in TP sediment was the second highest and extractable  $\text{PO}_4$  was equivalent to that in PB but higher than that of LL and BB sediment. Sediment pH values at the 4 stations were all very similar. Particle size distributions of the 4 stations were also similar, with most of the particles between 0.105 and 1.0  $\mu\text{m}$  (Table 3). Twin Ponds sediment had a greater proportion of its sediments in the larger fractions than the other stations. All sediments were characterized by high kaolinite contents.





Table 2. Chemical and physical sediment characteristics.

	Pond B	Limnology Lab	Beyer's Bay	Twin Ponds
% Organic Carbon	0.54	0.33	9.16	0.54
pH	5.70	5.75	5.50	6.10
Total Kjeldahl-N (ppm)	233.30	146.73	100.41	197.63
NO <sub>3</sub> <sup>-</sup> + NO <sub>2</sub> <sup>-</sup> (ppm)	14.10	11.59	9.26	23.3
NH <sub>4</sub> <sup>+</sup> + NH <sub>3</sub> <sup>0</sup> (ppm)	47.8	26.07	24.71	26.25
Extractable PO <sub>4</sub> <sup>-3</sup> (ppm)	0.034	0.022	0.016	0.035
S <sup>-</sup> (ppm)	< 2	< 2	< 2	< 2

Table 3. Sediment particle size distribution at each of the four stations.

	Pond B (%)	Limnology Lab (%)	Beyer's Bay (%)	Twin Ponds (%)
P ≥ 2.36 m	0.36	0.18	0.01	4.32
2.360 ≥ P ≥ 2.000	0.11	0.05	0.04	3.12
2.000 ≥ P ≥ 1.180	1.45	0.88	0.52	7.58
1.180 ≥ P ≥ 1.000	1.44	0.98	0.65	1.83
1.000 ≥ P ≥ 0.250	59.73	39.63	42.86	69.23
0.250 ≥ P ≥ 0.105	25.14	44.80	45.93	9.73
P ≥ 0.105	11.78	13.46	10.00	4.20

The Limnology Lab station on Par Pond had the greatest number of aquatic macrophyte species (Table 4). This station was almost completely encircled by *Typha latifolia* L. with the bottom carpeted with *Eleocharis acicularis* (L.) R & S. The BB station had a similar number of species, while PB had fewer species that did the Par Pond stations and TP had the smallest number of species. The BB station was sparse and the plants smaller. Neither PB nor TP had an established *M. spicatum* population.

Tied sprigs floated in the water above the sediments accumulating sediment as they settled to the bottom. Some lengths of *M. spicatum* settled to the bottom in a few days, while others had not settled after 14 days. After 20 days most of the sprigs had settled to the bottom. Roots formed at leaf nodes, often before plants were on the sediments. Once on the bottom sediments, sprigs become covered with a light layer of silt and rooted in the bottom sediment. Plants laying on their sides often rooted in several places along the stem.

*M. spicatum* did not root or grow when suspended in PB water but there was successful rooting at the other 3 stations. There were no significant ( $P \leq 0.90$ ) location or sediment effects on rooting at these

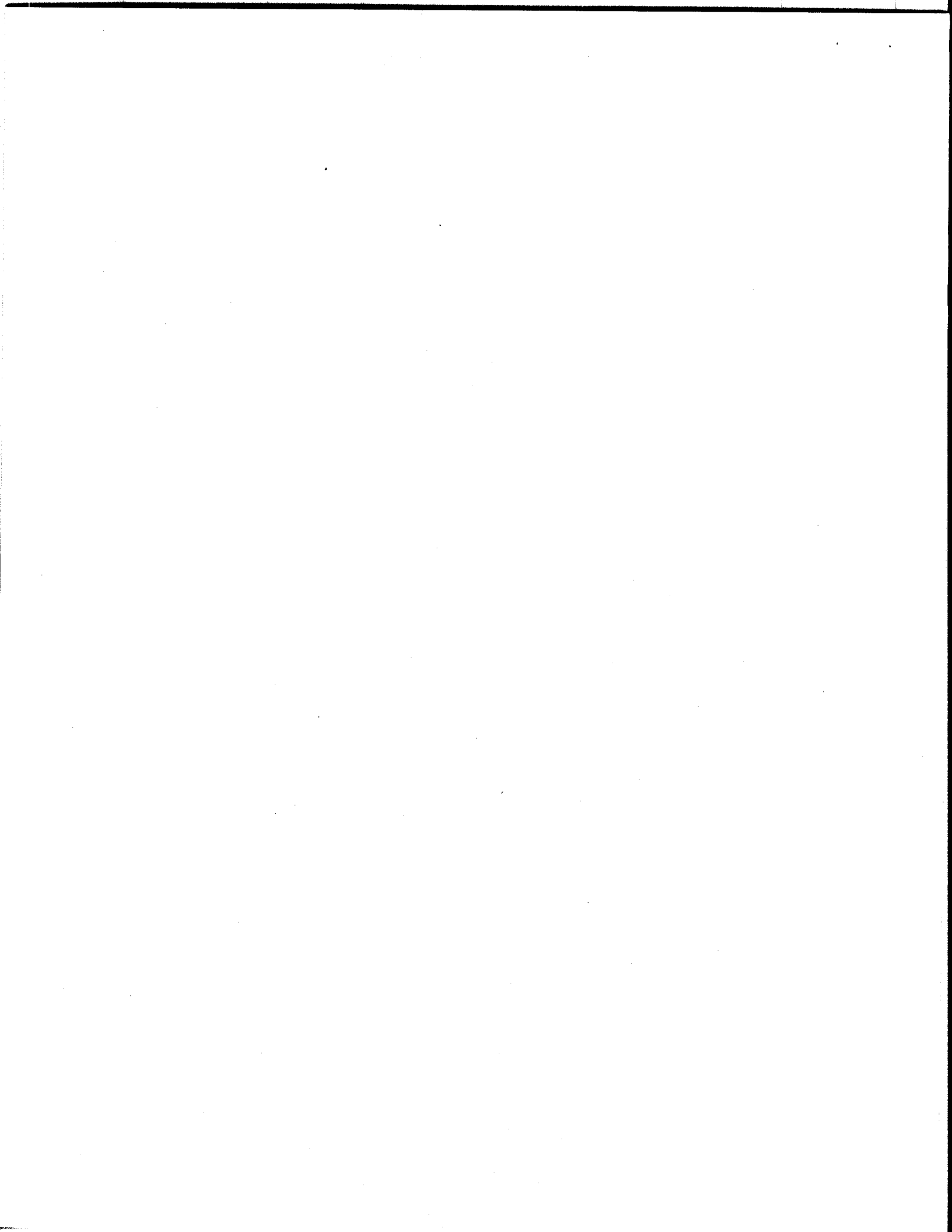


Table 4. Occurrence of aquatic macrophytes. LL = Limnology Lab Par, BB = Beyers Bay Par, PB = Pond B, TP = Twin Ponds.

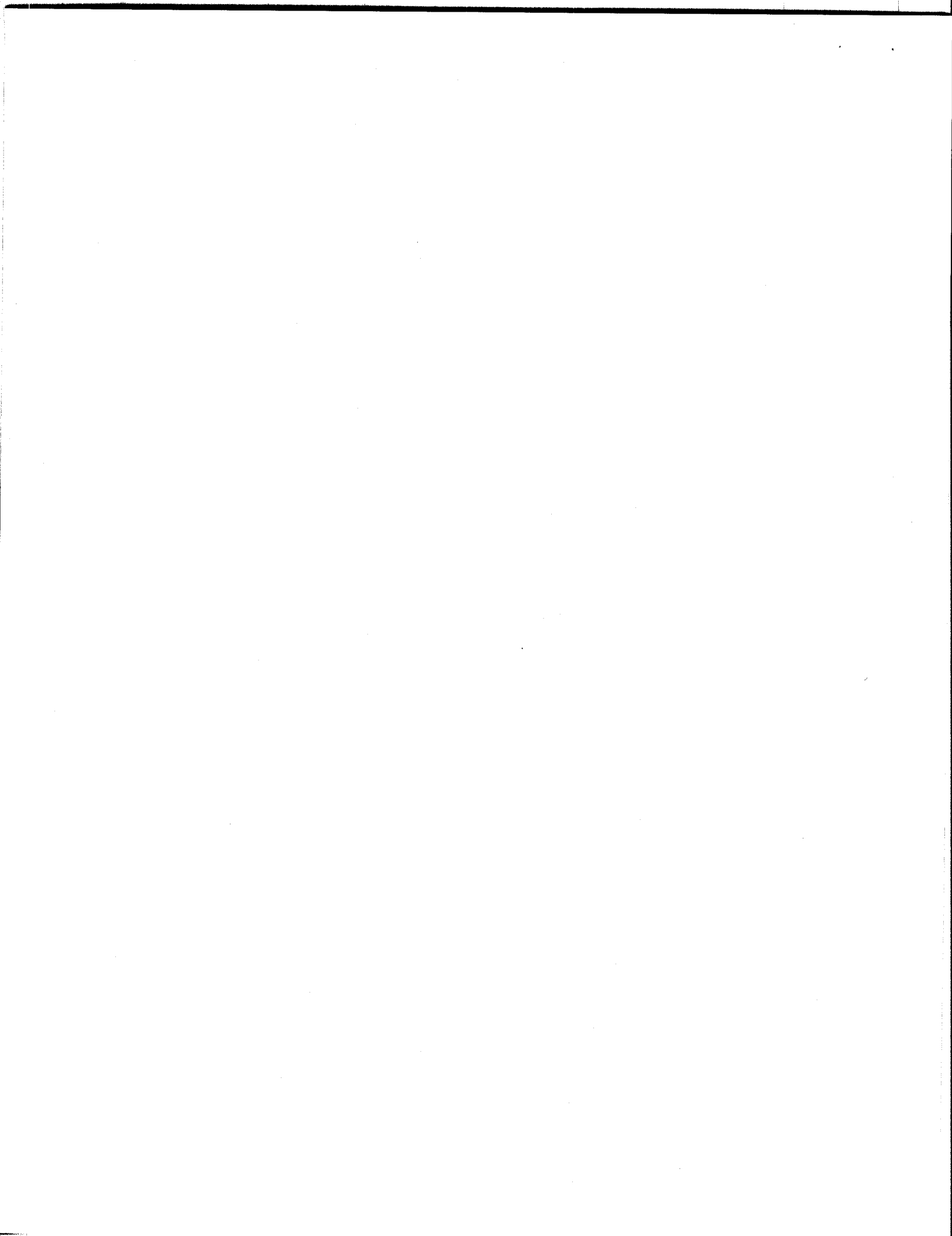
Species	LL	BB	PB	TP
<i>Typha latifolia</i> L.	+	+	+	+
<i>Eleocharis acicularis</i> (L.) R. & S.	+	+	+	+
<i>Hydrocotyle umbellata</i> L.	+	+	+	
<i>Typha domingensis</i> PERS.	+	+		
<i>Najas gracillima</i> (A. BR) MAGNUS	+	+		
<i>Potamogeton pusillus</i> L.	+			
<i>Vallisneria americana</i> MICHX.	+			
<i>Najas guadalupensis</i> (SPRENG.) MAGNUS		+		
<i>Utricularia</i> sp.			+	
<i>Ceratophyllum demersum</i> L.			+	
<i>Elodea nuttallii</i> (PLANCH.) ST. JOHN				+

stations. When *M. spicatum* was suspended over PB sediments at locations other than PB, there was no difference in rooting success, indicating lack of growth in PB was not due to sediment effects.

*M. spicatum* exhibited measurable growth after 14 and 21 days at all stations except PB, where all of the material disintegrated after 14 days of exposure (Table 5). Analysis of variance of growth, where growth occurred, indicated significant main effects due to sediment type and location ( $P > 0.90$  and  $P > 0.95$ , respectively) and a highly significant interaction between sediment type and location ( $P > 0.999$ ). Because of this interaction, one-way analyses of variance were conducted for soil type within location and locational effects across soil type. There were significant effects ( $P > 0.90$ ) in all classes except soil type at the Limnology lab stations.

Table 5. Effect of location on growth of *M. spicatum* by sediment type. Mean growth (cm) at each location after 21 days is listed from left to right in ascending order. Means which are not significantly different at  $\alpha = 0.05$ , using Tukey's w-procedure are underlined. N = 14, P = 3, error df = 156.

Sediment	Location		
	TP	LL	BB
LL	<u>7.21</u>	<u>10.17</u>	11.27
BB	LL	BB	TP
	10.68	<u>17.27</u>	18.57
TP	LL	TP	BB
	8.05	8.28	13.39
PB	LL	BB	TP
	6.97	7.56	9.85



Plants grown at LL exhibited the poorest growth regardless of sediment type (Table 5). The best growth was observed at Beyers Bay and TP, which represented an area of natural colonization and good growth and an area which was not colonized and assumed to represent adverse conditions for growth.

The effect of sediment on growth was much less than that of location, however, sediment from LL supported the poorest growth (Table 6). The effects of location at LL suppressed any sediment effects. Beyers Bay sediment supported the best growth regardless of station.

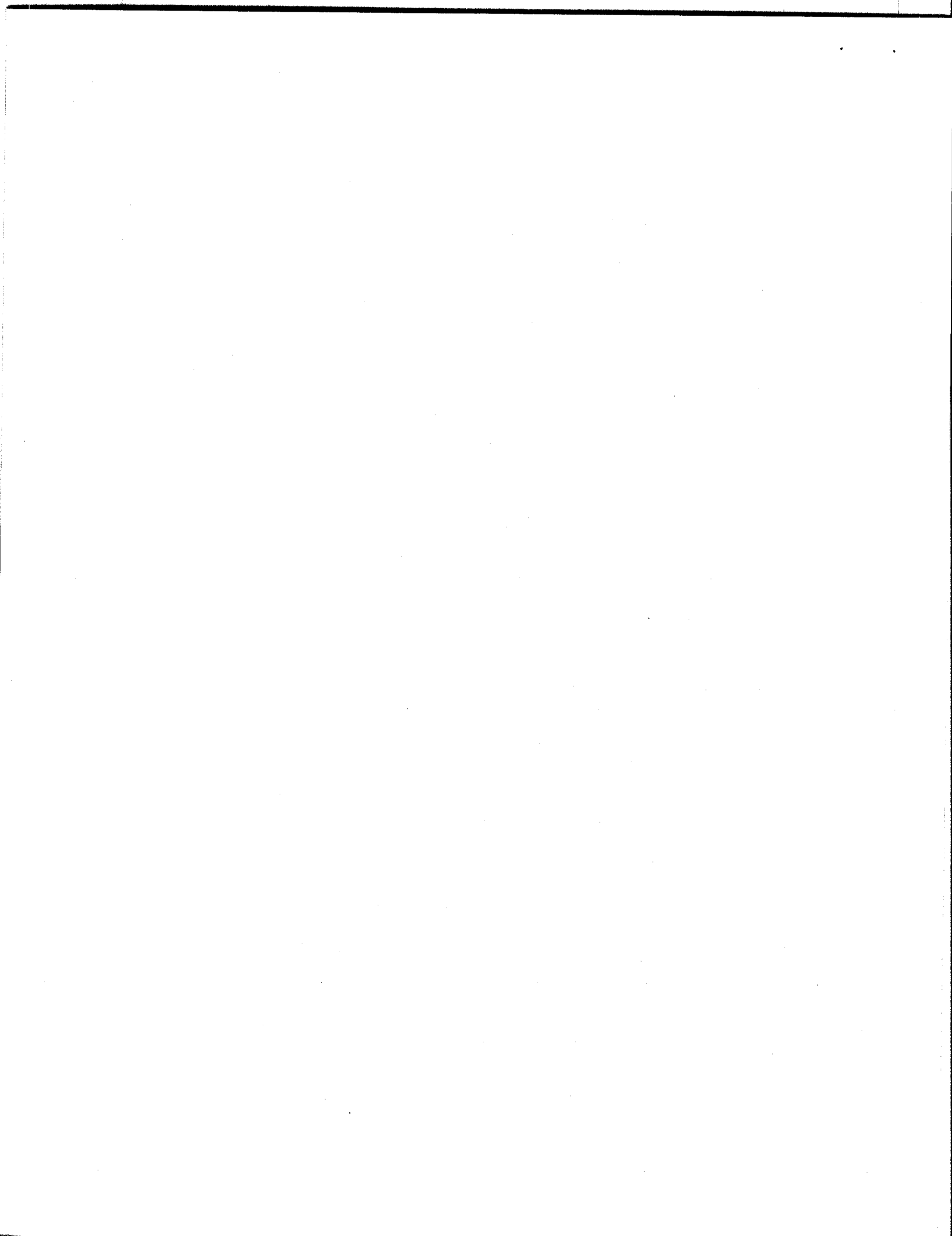
Table 6. Effect of sediment type on growth of *M. spicatum* by sediment type. Mean growth (cm) at each sediment type after 21 days is listed from left to right in ascending order. Means which are not significantly different at  $\alpha = 0.05$  using Tukey's w-procedure are underlined.  $N = 14$ ,  $P = 4$ , error  $df = 156$ .

Location	Sediment type			
LL	PB	TP	LL	BB
	<u>6.97</u>	<u>8.05</u>	<u>10.17</u>	<u>10.68</u>
BB	PB	LL	TP	BB
	<u>7.56</u>	<u>11.27</u>	<u>13.39</u>	<u>17.27</u>
TP	LL	TP	PB	BB
	<u>7.21</u>	<u>8.28</u>	<u>9.85</u>	<u>18.57</u>

### Discussion

Although viable seeds are formed in large quantities and viable seeds can be found in the water and on the bottom at all times of the year, the most efficient method of reproduction and dispersion of *M. spicatum* is by fragmentation (SMITH, 1963). Terminal pieces of *M. spicatum* are abscised and float in the water column before losing buoyancy and settling to the bottom. Abscission buds which settle to the bottom develop roots which anchor the plant. These buds may be transported from one body of water to another by waterfowl or human activities.

The alkalinity of Par Pond is higher than that of the surrounding ponds and sloughs, but is still relatively low. The alkalinity value determined at BB, where *M. spicatum* is well established, is only  $13.8 \text{ mg} \cdot \text{l}^{-1} \text{ CaCO}_3$ , (Table 1), which is below the range where *M. spicatum* is generally found (SPENCE, 1967). The Ca concentration in Par Pond is between  $3.0$  and  $4.0 \text{ mg} \cdot \text{l}^{-1}$ , which is far below the *M. spicatum* range indicated by HUTCHINSON (1970). Establishment and growth of *M. spicatum* in this very soft water raises the question of what lower extremes of Ca concentration and pH the plant can tolerate. *M. spicatum* rooted and

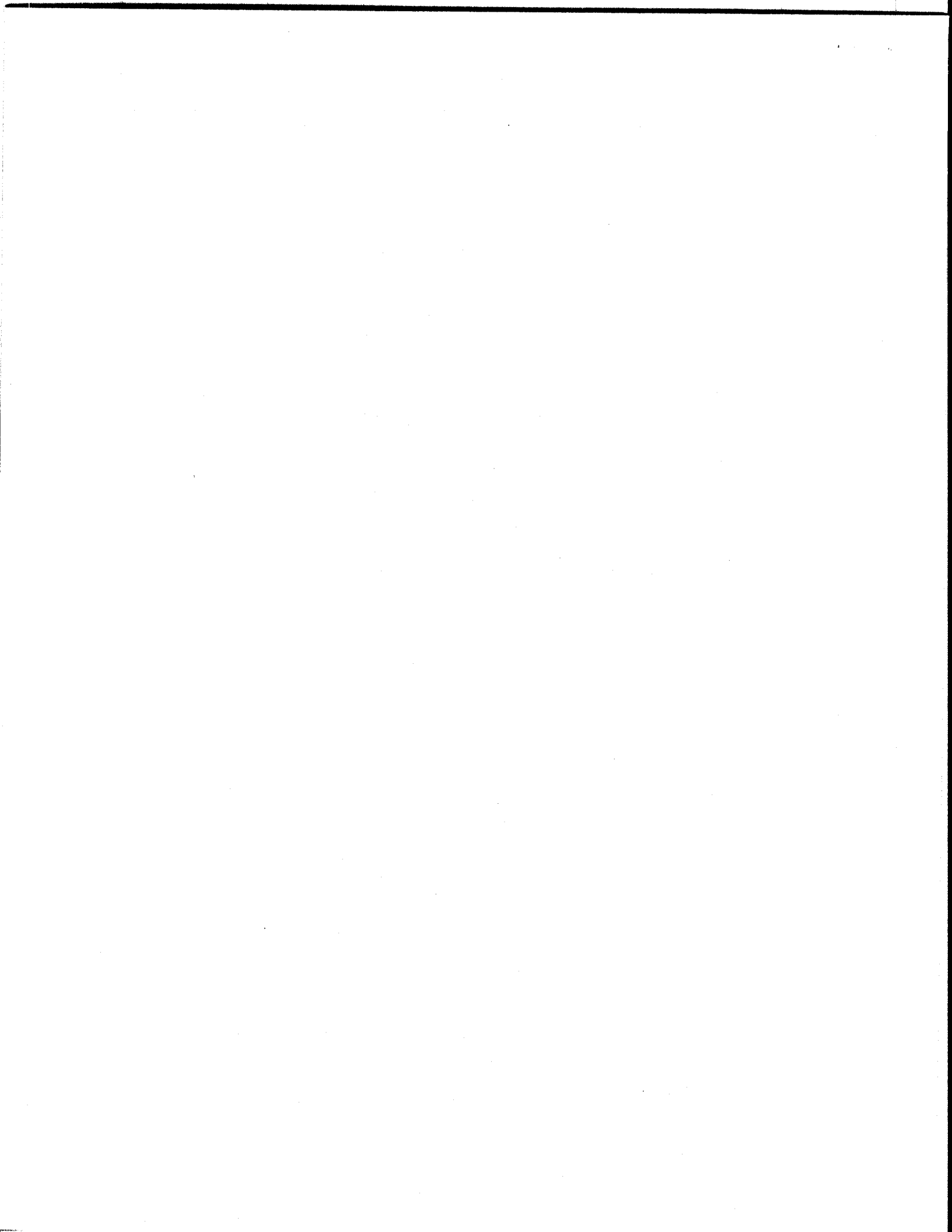


grew in TP which has a total alkalinity of only  $6.4 \text{ mg} \cdot \text{l}^{-1} \text{ CaCO}_3$  and total Ca concentration of  $0.7 \text{ mg} \cdot \text{l}^{-1}$ . No growth occurred in PB which has total alkalinity and Ca concentrations similar to TP suggesting that total alkalinity and Ca concentration alone were not controlling rooting potential and growth.

Laboratory studies by STANLEY (1970) indicated a pH optimum of 8.4 for *M. spicatum* and SPENCE (1967) found that survival of *M. spicatum* is directly correlated with high pH. ANDERSON, et al. (1965) reported the pH range of *M. spicatum* to be 5.8 to 9.5, while STEEMANN-NIELSEN (1947) indicated the photosynthetic processes of *M. spicatum* are poisoned by pH conditions above 10 and below 2.2 and that respiration was 20% lower at pH 4.5 than at 9.3. HUTCHINSON (1970) developed occurrence envelopes of 3 species of *Myriophyllum* which indicated the relationship between the pH and Ca determining their distribution. While the pH of Par Pond is within the optimum range for *M. spicatum*, the pH in TP is well below that where *M. spicatum* is reported to be successful. However, colonization and growth in TP was comparable to that at the Par Pond stations, indicating the ability of *M. spicatum* to grow in low pH situations.

The low growth observed at the LL station for all soil types was probably due to the morphometry of the area. Although the station proper was protected by stands of *T. latifolia* L. and *T. domingensis* PERS., it was subject to more wave action than the other stations. The *M. spicatum* which was naturally present in the area was small, due to fragmentation. There was little wave action in TP and both PB and BB were protected. The bottom morphometry at LL was much flatter. This, coupled with increased wave action, also reduced the amount of fine organic material overlying the bottom sediments. Reduced superficial organic sediment and continuous movement of sprigs reduces the potential for establishing *M. spicatum* in the area.

Because the number of growing pans and number of *M. spicatum* shoots was limited, growth was determined using length instead of biomass so that *in situ* measurements could be made without plant removal. Plants in experimental pans grew rapidly, then fragmented, with the fragments floating away. Some *M. spicatum* grew as much as 35 cm in 21 days. Measurements taken after 28 days showed reductions in length of as much as 20 cm over those taken at 21 days. This made growth determinations after 28 days impossible and may be responsible for some of the experimental variability. Growth quantification was further complicated by invasion of growing pans by *M. spicatum* previously present at the LL and BB stations. However, the fact remains that considerable growth occurred in TP, which is outside the generally accepted range for successful *M. spicatum* growth.





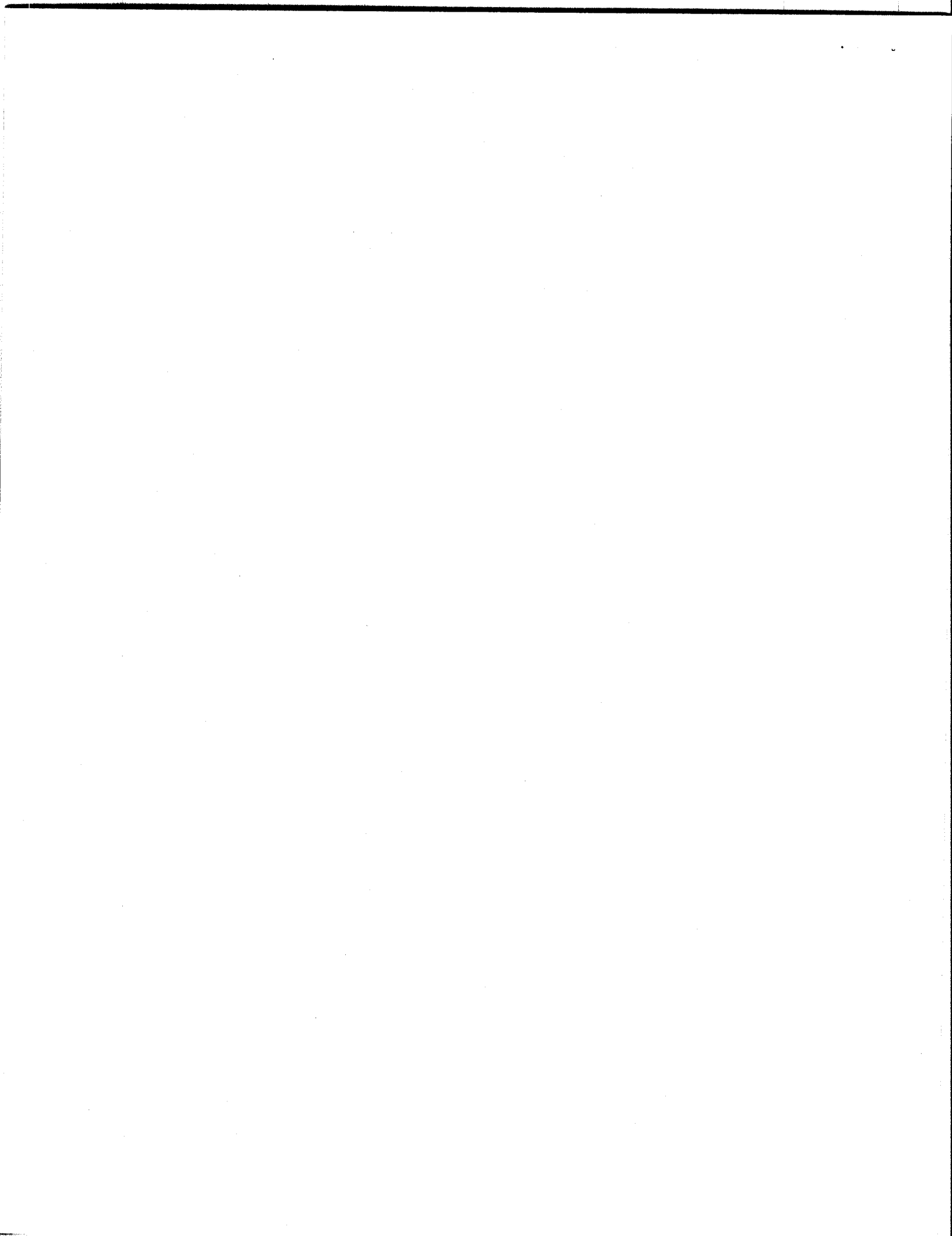
Sediment texture is important in determining the distribution of *M. spicatum* (PELTIER & WELCH, 1970). The sediment particle size distribution of *M. spicatum* at all 4 stations is within the range preferred by *M. spicatum* (PATTEN, 1955). The most striking difference between sediments from the 4 locations was the percentage of organic carbon. Although the BB sediment was underlain by sand and clay similar to that at the other stations, the upper few cm were highly organic. The general trend toward better growth on BB soil is probably due to this increased organic carbon content since both total  $\text{PO}_4^{3-}$  and Kjeldahl nitrogen were relatively low in this sediment. Although sediment preference descriptions by many authors indicate that *M. spicatum* is an adaptable plant, the best growth generally occurs on muck and fine ooze (STEENIS, et al., 1962). The extractable  $\text{PO}_4^{3-}$ , Kjeldahl nitrogen and  $\text{NO}_3^-$  and  $\text{NO}_2^-$  concentrations in BB sediment were all less than in the other three sediments, indicating that limitation by these nutrients was not responsible for the poorer growth on the other sediments.

*M. spicatum* has naturally invaded Par Pond which has a total alkalinity which is below the range generally reported for areas where *M. spicatum* grows. When *M. spicatum* sprigs were transferred to two ponds with softer, more acid waters than those of Par Pond, successful rooting and growth occurred in one but not the other. The chemical and physical parameters of both ponds were similar and no single outstanding parameter limiting growth in PB could be identified. The most outstanding features of PB, which may restrict *M. spicatum* are high  $\text{Cl}^-$  combined with low pH and  $\text{CO}_2$  or low Fe. *M. spicatum* has the potential to invade softwater systems in the Southeastern United States which are below the ranges of pH and alkalinity previously reported. AIKEN (Personal communication) has observed the *M. spicatum* is expanding its range into waters in Canada with essentially no bicarbonate alkalinity. Therefore, care should be taken to restrict its introduction into these areas.

Because many factors, such as interspecific competition may influence the invasion success of *M. spicatum* into a particular system long range studies should be conducted in acid-soft water systems. The expansion of the range of *M. spicatum* into areas where it has previously been restricted from may be due to increased human activity and concomitant increased vegetative dissemination, genetic changes in the species or changes in the chemical nature of surface waters.

#### Zusammenfassung

Die Ausbreitungsmöglichkeiten des europäischen Tausendblattes (*Myriophyllum spicatum* L.) in sauren weichen Gewässern wurden durch reciproke Sediment-Verpflanzungen von 3 Teichen des Savannah River Projekts unter-



sucht. Aktiv wachsende vegetative Spitzen wurden über dem Sediment in Plastik-Pfannen verteilt und die Bewurzelungs-Fähigkeit und das Wachstum an jeder Station gemessen. Par Pond, ein Reservoir mit höheren pH- und Alkalinitäts-Werten als die umgehenden Gewässer infolge des Wassereinflusses vom Savannah River, ist kürzlich durch *M. spicatum* besiedelt worden. *M. spicatum* wurde in 2 andere Teiche mit weichem Wasser und mit pH- und Gesamtalkalinitäts-Werten zwischen 5 und 6 mg/l CaCO<sub>3</sub> verpflanzt. Sie wurzelte und wuchs in dem einen Teich, aber nicht in dem anderen trotz ähnlicher pH- und Calcium-Werte. *M. spicatum* wurzelte und wuchs unter pH-, Alkalinitäts- und Calcium-Bedingungen, die weit unter den bisher bekannten Toleranzgrenzen lagen. Die Unterschiede in der Fähigkeit zur Bewurzelung und Wachstum wurden nicht durch Sediment-Unterschiede verursacht.

### References

- American Public Health Association (1976): Standard methods for the examination of water and wastewater, 14th ed. 1193 p.
- ANDERSON, R. R., BROWN, R. G. & RAPPLEYE, R. D. (1965): Mineral composition of Eurasian water milfoil, *Myriophyllum spicatum* L. — Chesapeake Sci. 6 (1): 68—72.
- BERGQUIST, E. T. (1970): Ecological and morphological effects of 2, 4-D on *Myriophyllum spicatum* L. in a Tennessee reservoir. — Ph. D. Dissertation. Univ. Tennessee, Knoxville.
- BLACK, C. A., EVANS, D. D., WHITE, J. C., ENSMINGER, L. E. & CLARK, F. E. (1965): Methods of soil analysis; Part 2: Chemical and microbial properties. — Amer. Soc. Agron. Inc., Madison Wisc. 1572 p.
- CHAPMAN, H. D. & PRATT, P. F. (1961): Methods of analysis for soils, plants and waters. — Univ. Calif., Berkeley. 309 p.
- CLAPHAM, A. R., TUTIN, T. G. & WARBURG, E. F. (1952): Flora of the British Isles. — Cambridge Univ. Press, Cambridge. 1591 p.
- CONOVER, W. J. (1971): Practical nonparametric statistics. — John Wiley & Sons, Inc., New York, 462 p.
- CROWELL, T. E., STEENIS, J. H. & SINCOCK, J. L. (1967): Recent observations of Eurasian water milfoil in Currituck Sound, North Carolina and other coastal and southeastern states. — Proc. 12th Ann. Meeting South. Weed Conf. 348—352.
- GIAMMATTEO, P. (1975): A method for the direct determination of sulfide in water and sediments. — M. S. Thesis. Univ. Georgia, Athens, Ga.
- HUTCHINSON, G. E. (1970): The chemical ecology of three species of *Myriophyllum* (Angiospermae, Haloragidaceae). — Limnol. Oceanogr. 15 (1): 1—5.
- (1975): A Treatise on Limnology III: Limnological Botany. — John Wiley and Sons, New York, 660 p.
- MASON, H. L. (1957): A flora of the marshes of California. — Univ. Calif., Berkeley. 878 p.
- MISRA, R. D. (1968): Edaphic factors in the distribution of aquatic plants in the English lakes. — J. Ecol. 26: 411—451.
- NICHOLS, S. A. & MORI, S. (1971): The littoral macrophyte vegetation of Lake Wingra: An example of a *Myriophyllum spicatum* invasion in a southern Wisconsin lake. — Trans. Wis. Acad. Sci. Arts. Lett. 59: 107—119.



- PATTEN, B. C., Jr. (1954): The status of some American species of *Myriophyllum* as revealed by the discovery of intergrade material between *M. exalbes-cens* FERN and *M. spicatum* L. in New Jersey. — *Rhodora*. 56: 213—225.
- PATTEN, B. C. (1955): Germination of the seed of *Myriophyllum spicatum* L. — *Bull. Torrey Bot. Club*. 82 (1): 50—56.
- PELTIER, W. H. & WELCH, E. B. (1970): Factors affecting Growth of rooted aquatic plants in a reservoir. — *Weed Sci.* 18: 7—9.
- SMITH, G. E. (1963): Control of Eurasian water milfoil (*M. spicatum*) in TVA reservoirs. *Proc. Southeastern Weed Control Conf.* 16: 351—353.
- SMITH, G. E., HALL, T. F. & STANLEY, R. A. (1967): Eurasian water milfoil in the Tennessee Valley. — *Weeds*. 15 (2): 95—98.
- SPENCE, D. H. N. (1967): Factors controlling the distribution of freshwater macrophytes with particular reference to the lochs of Scotland. — *J. Ecol.* 55: 147—170.
- STANLEY, R. A. (1970): Studies on nutrition, photosynthesis and respiration of *Myriophyllum spicatum* L. — Ph. D. Dissertation. Duke Univ., Durhan, N. C.
- STANLEY, R. A., HALL, T. F. & SMITH, G. E. (1966): Studies on the biology and control of Eurasian water milfoil in the Tennessee Valley. — *Proc. South-eastern Weed Control Conf.* 19: 396.
- STEEMANN-NIELSEN, E. (1947): Photosynthesis of aquatic plants with special reference to the carbon sources. — *Dansk Bot. Ark.* 12 (8): 5—71.
- STEENIS, J. H. & STOTTS, V. D. (1963): Progress report on distribution and control of Eurasian water milfoil in the Chesapeake Bay region, 1962. — *Proc. South Weed Conf.* 16: 341—342.
- STEENIS, J. H., STOTTS, V. D. & GILLETTE, C. R. (1962): Observations on distribution and control of Eurasian water milfoil in Chesapeake Bay, 1961. — *Proc. New England Weed Control Con.* 16: 442—448.
- WOODENHOUSE, R. P. (1933): Tertiary pollen. II. The oil shales of the Eocene Green River formation. — *Bull. Torrey Bot. Club*. 60: 479—524.

## Address of the authors:

JOHN P. GIESY, Jr. & LAURA E. TESSIER, Savannah River Ecology Laboratory, Drawer E, Aiken, SC 29801.

