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THE UPTAKE OF BORON
BY LEMNA MINOR

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ABSTRACT

THE UPTAKE OF BORON BY *LEMNA MINOR*

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Robert P. Glandon

Pilot analyses have suggested that *Lemna minor* L. can concentrate boron to levels an order of magnitude greater than those reported for other macrophytes. This study aimed to verify the unusual boron accumulating capability of *L. minor*. More precisely, the response in tissue boron concentration to different ambient boron concentrations was examined in the laboratory. In addition, samples of *L. minor* and *Ceratophyllum demersum* L. collected over a growing season from a wastewater pond were analyzed to follow the seasonal changes of tissue boron concentrations in the co-occurring macrophyte species.

The results showed that *L. minor* is a bio-accumulator of boron relative to other aquatic macrophyte species. Furthermore, analyses of tissue boron concentrations showed that the ambient levels under consideration were significantly different in their effect on boron levels in plant tissue. The data imply that boron was taken up from the medium at a rate reflecting the ambient boron concentration. The results of boron analyses on field samples showed that *L. minor* contained a boron concentration that was tenfold that of *C. demersum* throughout the season. Further, the results indicate that the kinetic effect of growth acts to offset the effect of uptake rate on the observed tissue concentration of boron.

THE UPTAKE OF BORON BY *LEMNA MINOR*

By

Robert P. Glandon

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TABLE OF CONTENTS

	Page
LIST OF TABLES	iv
LIST OF FIGURES	v
INTRODUCTION	1
MATERIALS AND METHODS	3
RESULTS	7
DISCUSSION	14
CONCLUSIONS	23
LITERATURE CITED	24

LIST OF TABLES

Table		Page
1	Changes in boron content and biomass over the 1973 growing season for <i>Lemna minor</i> and <i>Ceratophyllum demersum</i> in the fourth pond of the Belding, Michigan waste stabilization system.	11
2	Boron concentrations expressed as $\mu\text{g g}^{-1}$ ash-free dry weight of macrophytes collected from the Belding system in September, 1972.	21

LIST OF FIGURES

Figure		Page
1	Relationship between ambient boron concentration and mean tissue boron concentration (± 1 standard deviation) in rapidly growing cultures of <i>Lemna minor</i> .	8
2	Changes in boron concentration of <i>Lemna minor</i> and <i>Ceratophyllum demersum</i> over the growing season of 1973 at Belding, Michigan.	12
3	Stylized representation of the relationship between tissue concentration and growth rate at ambient boron concentration where $[B_1] < [B_2] < [B_3] < [B_{\text{toxic}}]$.	17

INTRODUCTION

Much of the recent limnological literature pertaining to boron has been concerned with the biogeochemical cycling of this element in freshwater. Some attention has been focused in these papers on the accumulation of boron by aquatic plants. Aquatic angiosperms require boron for growth and concentrate it relative to the ambient environment (Gerloff, 1973; Eichhorn and Augsten, 1974). Recent investigations of boron accumulation in macrophyte communities have entailed observations on the species level so that the relative significance of each species may be interpreted. Comprehensive examinations were done by Boyd and Walley (1972) working in Par Pond, South Carolina, and by Cowgill (1974) working in Linsley Pond, Connecticut. In general, the plants examined reflected the great difference in ambient boron between these two sites. Linsley Pond contained $0.485 \text{ mg l}^{-1} \text{ B}$, roughly 100 times that in Par Pond. Plants of the former were consistently higher in boron content. For example, *Nymphaea odorata* contained the highest concentration at both localities. Sixty-five $\mu\text{gm gm}^{-1} \text{ B}$ by dry weight in this species at Linsley Pond was about five and a half times the content of the plants examined by Boyd and Walley. These data are of interest in that the boron concentrations in tissues reported for the Linsley Pond specimens are among the highest in the literature for an aquatic plant species. Further, they suggest a relationship between tissue and ambient boron levels.

Our pilot analyses (cf. McNabb, in press) have suggested that *Lemna minor* L. and *Lemna trisulca* L. can concentrate boron to levels an order of magnitude greater than those reported for *N. odorata* and other macrophytes. The potential of species of *Lemna* to control the cycling of boron in an aquatic plant community engendered this investigation.

The aim of this study was to verify the unusual boron accumulating capability of *Lemna minor*. More precisely, the question of whether *L. minor* accumulates boron relative to environmental concentrations was examined. This was tested in experimental laboratory cultures by examining the response in tissue boron concentration to different ambient boron concentrations. The data were collected in such a way as to characterize this relationship in rapidly growing populations of *L. minor*. Furthermore, samples of the plant collected from a wastewater pond at Belding, Michigan were analyzed to follow the seasonal changes of the tissue boron concentrations. Samples of *Ceratophyllum demersum* L., collected over the same period at that site, were analyzed to provide a contrast between co-occurring species in the macrophyte community.

MATERIALS AND METHODS

Lemna minor (duckweed), a floating aquatic angiosperm, is commonly composed of two or three fronds. Vegetative reproduction is accomplished when the fronds of the plant produce similar fronds of the second order from pockets on either side of the basal region. As the lateral shoots increase in size any movement of the water surface can detach them from the base of the preceding generation. A root with a conspicuous root cap hangs into the water from the underside of each plant (Arber, 1920).

Experimental plants were cultured in the laboratory from a few specimens selected from a clone in a pond at the Limnological Research Laboratory on the campus of Michigan State University. Two hundred individuals, weighing 0.0285 grams dry weight in total, were seeded into 5-liter, plastic culture vessels having a surface area of 398 cm². These vessels were located within a controlled environment chamber. Nutrient medium was conducted to each vessel through tygon^R tubing (1 mm I.D.) by a peristaltic pump with six metering heads. Each head drew from a polyethylene carboy containing medium of the desired boron concentration. The tubing fed into the bottom of a culture vessel. A constant water level was maintained within a vessel by means of a small drain at the top of the culture vessel. The flow was sustained at a rate of approximately 1.4 liters/vessel/day.

The synthetic nutrient medium used was a modification of the Hoagland solution employed by Gerloff (1973), with the exception that iron was added as a chelate of EDTA. The pH of the medium was adjusted to 8.0 by the addition of NaHCO_3 and the concentration of boron was modified according to the desired treatment levels.

The laboratory experiment was divided into four separate culture periods lasting seven days each. In preparation for each run the nutrient medium was mixed in a large holding tank from which three 20-liter carboys were filled. Boron, in the form of boric acid, was added to two of the carboys to result in concentrations of 0.1 mg l^{-1} and 1.0 mg l^{-1} B. No boron was added to the third. Two culture vessels were fed from each carboy.

Water samples taken from each vessel on the 2nd, 4th, and 6th days of each run and analyzed for boron revealed treatment levels. The mean of all samples (\pm standard deviation) taken from units to which no boron was added was $0.01 \pm .006 \text{ mg l}^{-1}$ B. The mean working concentrations in all vessels to which 0.1 and 1.0 mg l^{-1} B were added were $0.11 \pm .01$ and $1.01 \pm .015 \text{ mg l}^{-1}$ respectively.

Plants gathered from each unit at the end of the run were washed and counted. The material was dried to a constant weight at 100°C and subsequently ground in a micro-Wiley mill using a 40-mesh screen. Weighed samples were then combusted at 550°C in a muffle furnace for five hours and boron was leached from the ash material by the addition of 0.5N HCl (Dible and Truog, 1954).

Boron analyses on all water and plant samples were done colorimetrically by the curcumin method (Dible and Truog, 1954; American Public Health Association, 1971). Data were gathered from 1 ml aliquots

evaporated to dryness with 4 ml curcumin-oxalic acid solution in porcelain evaporating dishes. A temperature controlled water bath was used to maintain evaporation at $55 \pm 3^{\circ}\text{C}$. After color formation in 25 ml of 95% ethyl alcohol all samples were centrifuged for 3-5 minutes at 2400 rpm. Absorbance readings were taken at 540 nm from a Bausch and Lomb Spectronic-20 colorimeter.

Adjustments were made within the culture chamber so that each unit would receive approximately the same amount of light. The light was provided by 110 W Sylvania GRO-Lux^R florescent lamps on a 20 hour photoperiod. Light intensity at plant level varied from 10,700 to 9,200 lux through the course of the experiment. The air temperature reached 30°C during the light period and 24°C during the dark period. The vessels were suspended in a water bath which maintained the temperature of the nutrient medium between 25° and 27°C .

A randomized complete block was the statistical design chosen to assess the significance of the effect of ambient boron concentrations on the tissue concentration of the element (Sokal and Rohlf, 1969).

Growth during the experimental period was expressed as doubling time. Doubling times were calculated according to the general formula:

$$\text{D.T.} = \frac{\ln 2}{\left(\frac{\ln N_{t_1} - \ln N_{t_0}}{t_1 - t_0} \right)},$$

where N_{t_1} is the number of plants at time t_1 ; and t_1 and t_0 represent the time expressed in days (Mitchell, 1974).

Complementary information on *Lemma minor* and *Ceratophyllum demersum* was obtained from samples collected from the fourth cell of a system of

sewage oxidation ponds at Belding, Michigan. This pond had a surface area of 3.0 hectares, averaged 1.8 m in depth, and had a mean retention time of 32 days during the time of plant collection. Other features of the environment during this period are described by Lisiecki and McNabb (1975). Plant samples were collected at approximately two week intervals during the growing season. Twenty samples of each species were taken so that estimates of changes in biomass could be made. The pleustonic *L. minor* was collected by placing a 25 cm square on the water surface. All of the plants in the square were placed in a polyethylene bag. The submersed *C. demersum* was sampled either by dropping a weighted plastic cylinder (0.114 m^2) and collecting the material originating within, or by raising an unweighted cylinder (0.094 m^2) from beneath the floating mats of *C. demersum* and collecting the plants inside. A composite of the material collected on each sampling date was made for each species and analyzed for boron content. The methods for boron analyses on these plant tissues were as described above for the laboratory study.

RESULTS

In the laboratory experiments, there was no evidence that any of the three boron concentrations influenced growth. Mean doubling times of plant numbers and their standard deviations were $2.29 \pm .34$, $2.16 \pm .46$, $2.22 \pm .46$ days from the lowest to the highest ambient boron concentration respectively. These doubling times are of the same order as the 2.16 days calculated from data presented by Wołek (1973) for average exponential growth in pure cultures of *Lemna minor* with a 16 hr. photoperiod. Doubling times of 4.0 days and 4.4 days were reported for the period of rapid growth of *L. minor* in the experimental cultures of Harvey and Fox (1973) and Sutton and Ornes (1975), respectively, using a 12-hr. photoperiod. The doubling time of plant numbers in this study reflected the doubling time of organic weight biomass over the period. Mean doubling times of organic weight and their standard deviations were $2.10 \pm .30$, $1.90 \pm .32$, and $1.95 \pm .31$ from the lowest to the highest ambient boron levels respectively.

Statistical analysis of the tissue boron concentrations showed that in repeatable experiments the ambient boron levels under consideration were significantly different in their effect on the boron levels in plant tissue ($\alpha = .01$). The data from individual runs were combined at each treatment level and a mean and standard deviation were taken. The results are presented in Figure 1. Results of fourteen separate boron determinations on a single *L. minor* sample showed that up to 25%

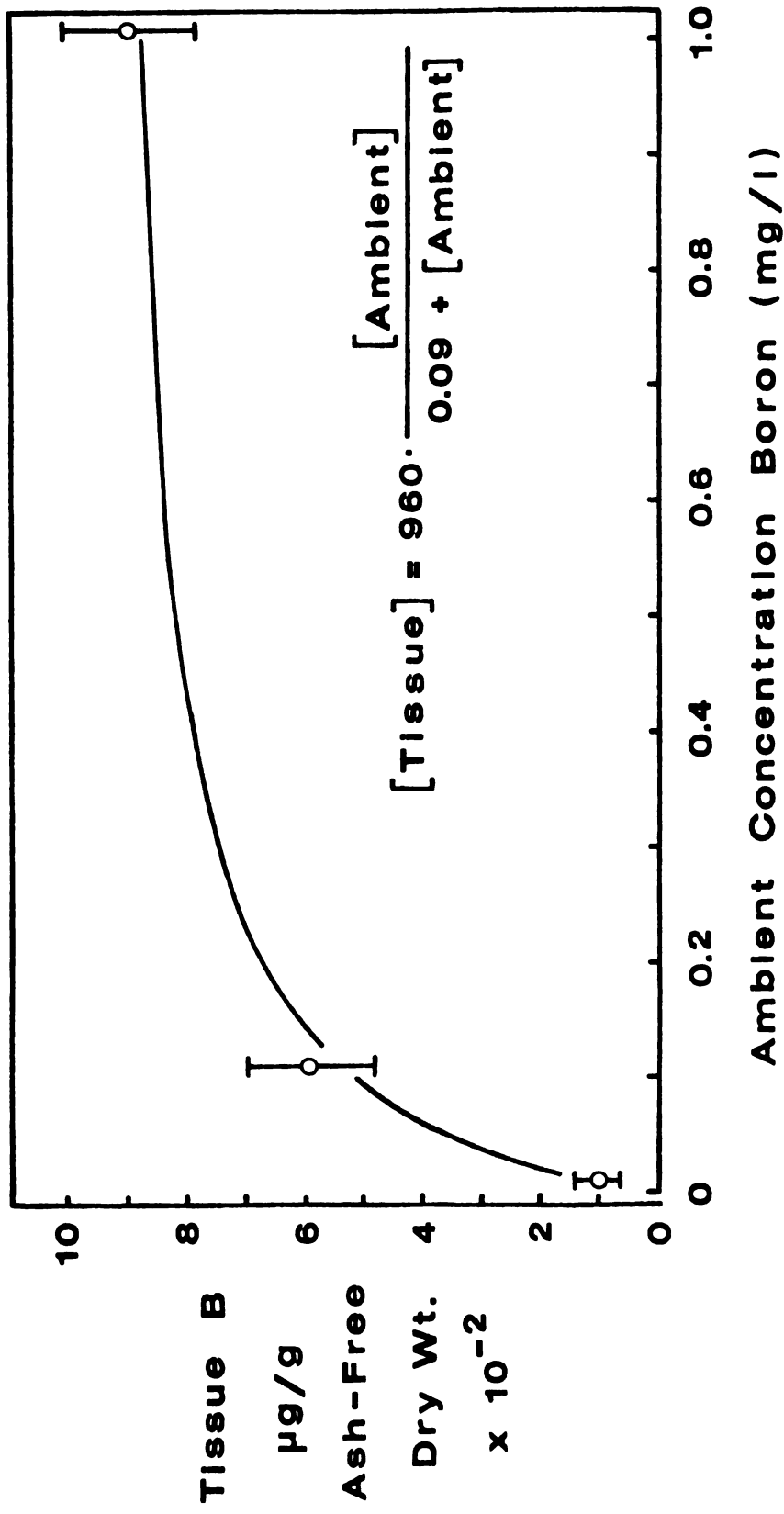


Figure 1. Relationship between ambient boron concentration and mean tissue boron concentration (± 1 standard deviation) in rapidly growing cultures of *Lemna minor*.

of the observed variability in the tissue levels can be attributed to analytical technique.

The plot of the plant tissue concentration on a parts per million organic weight basis against ambient boron concentration displayed a non-linear relationship. The curve representing the fit of the data points approaches a limit as ambient boron concentration increases and approaches the origin as ambient levels decrease. Estimation of the constants characterizing the equation was facilitated by performing a linear transformation and solving for the slope and y-intercept using the experimental data after the method of Dowd and Riggs (1964).

The curvilinear relationship suggests that changes in ambient concentrations below about 0.3 mg l^{-1} B produce a marked response in tissue boron levels. This general relationship is not unique to boron. A curvilinear response was observed when *L. minor* tissue phosphorus concentration was plotted against ambient orthophosphate levels (Sutton and Ornes, 1975). An unusual aspect of the data is the high tissue concentration at each ambient level relative to tissue boron levels of other species of aquatic plants.

Using the data of Figure 1, an arithmetic plot of the relative accumulation of boron (tissue boron + ambient boron) as a function of ambient boron could be made. Such a plot would indicate an active uptake of boron from the dilute medium and a relatively passive response at higher concentrations. This relationship to ambient boron was observed by Cowgill (1974) for other species of aquatic plants existing in localities that differed as to the available supply of boron.

The results of boron analyses on field samples of *Lemna minor* and *Ceratophyllum demersum* from the wastewater pond at Belding, Michigan and estimates of standing biomass of each species over the growing season are presented in Table 1. The seasonal change in boron concentrations of the two species is presented graphically in Figure 2.

Ceratophyllum demersum was the dominant submerged aquatic angiosperm found in the Belding pond. The plants have no roots. Basal branches sink or grow into the soft sediments between seasons of maximum growth. As the summer progressed, the bouyancy of new vegetation and the decomposition of basal portions of the stem combined to produce free-floating mats of the plant. During the summer of 1973, formation of the surface mat commenced during the first week in June, reached a maximum on 8/15 and declined in the interval of 9/5 to 9/26. As the floating vegetation developed, the population of *L. minor* expanded to cover it.

An obvious phase of Figure 2 is the precipitous decline in tissue boron experienced by both species during the latter half of June when concentrations fell to nearly fifty percent of their previous level. It can be noted from Table 1 that this interval corresponds to the period of most rapid increase of *L. minor* biomass. From June 20 to July 4 the doubling time of *L. minor* biomass was calculated to be 2.00 days.

Boron analyses show that throughout the season *L. minor* contained a boron concentration that was tenfold that of *C. demersum* (Table 1). Furthermore, toward the end of the season the two species exhibited divergent trends with respect to boron concentration. The concentration

Table 1. Changes in boron content and biomass over the 1973 growing season for *Lemna minor* and *Ceratophyllum demersum* in the fourth pond of the Belding, Michigan waste stabilization system.

Date	<i>Lemna minor</i>		<i>Ceratophyllum demersum</i>	
	mg B/Kg Ash-Free Dry Weight	Pond Standing Crop Kg Ash-Free Dry Weight ¹	mg B/Kg Ash-Free Dry Weight	Pond Standing Crop Kg Ash-Free Dry Weight ²
5/23	--	--	69	--
6/6	--	nil	87	281
6/20	1948	1	195	769
7/4	988	126	101	1685
7/18	2031	189	190	2660
8/1	2380	171	154	3377
8/15	2658	154	85	3973
9/5	3249	39	72	3565
9/26	3034	15	137	2623

¹Ash-free dry weight is 79% of dry weight.

²Ash-free dry weight is 78% of dry weight.

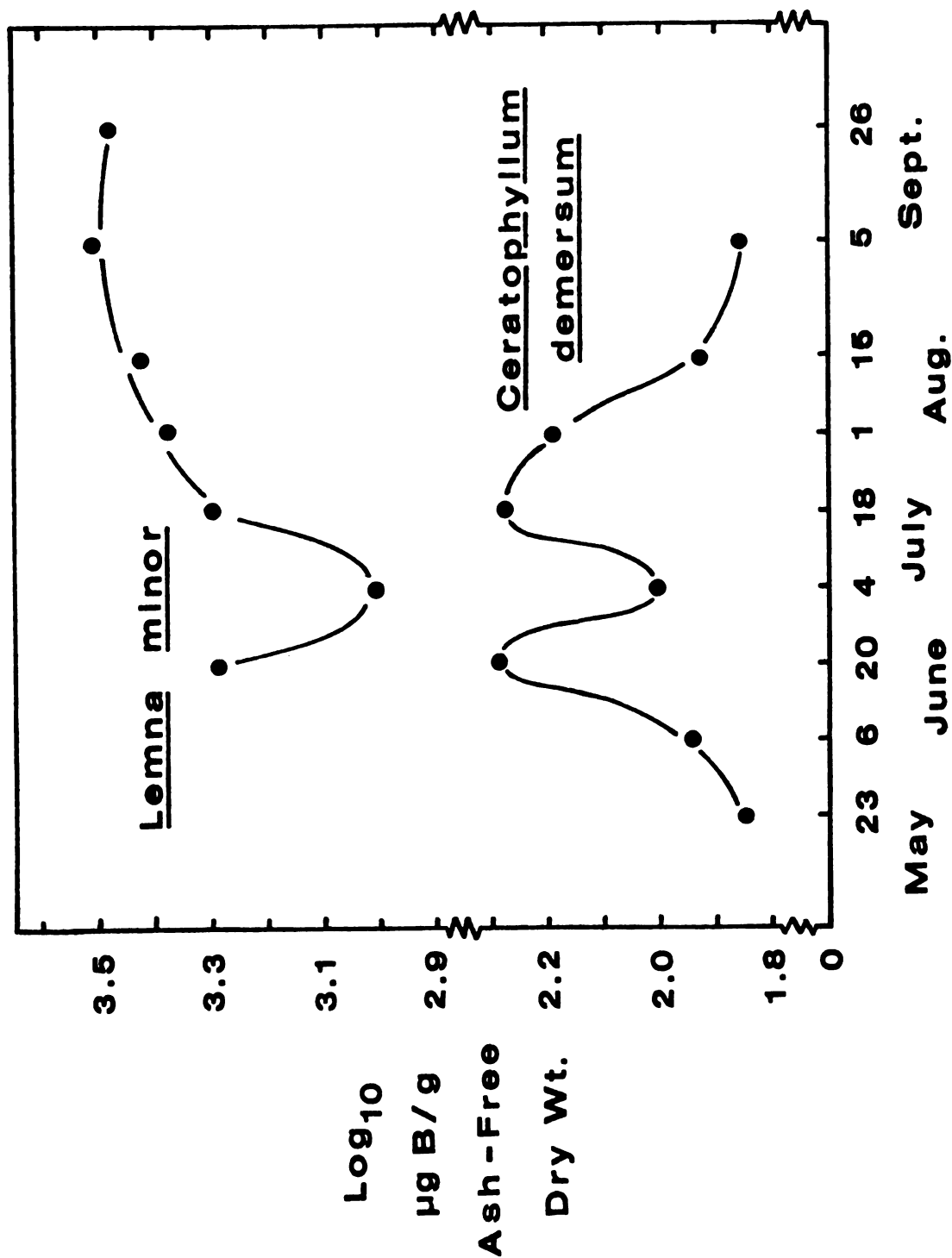


Figure 2. Changes in boron content of *Lemna minor* and *Ceratophyllum demersum* over the growing season of 1973 at Belding, Michigan.

in tissue of *L. minor* increased while that in *C. demersum* decreased. As a result, in the first week of September the concentration of *L. minor* was about 45 times that in *C. demersum*.

DISCUSSION

Investigations have shown that factors affecting boron uptake are complex. Temperature, light intensity, pH, evapotranspiration rate, growth stage of the plant, and levels of other nutrients have all been shown to affect the rate of boron accumulation in terrestrial plants (Bingham *et al.*, 1970; Reisenauer, 1971). In the laboratory portion of this study, the variable tested was the ambient boron concentration with other variables held constant.

Based on the laboratory data, the hypothesis that accumulation of boron by *Lemna minor* is related to environmental concentrations, is not rejected. The response is of the curvilinear nature described in Figure 1. The data imply that ambient boron was a rate determining factor in the incorporation of boron per unit of plant tissue. In a series of short term experiments, each totaling ten hours in duration, Thellier and LeGuier (1967) observed that the rate of boron uptake per unit biomass of *Lemna minor* increased with increasing boron levels in the nutrient medium up to 5 mg l⁻¹ B. Beyond that concentration uptake rates decreased, apparently in response to toxic effects of high ambient levels. In the experiments of this study, ambient concentrations of boron determined the total amounts of boron taken up over one week intervals. The tissue concentrations at the end of the experiments reflected the different uptake rates.

Boron uptake rates, as used here, are mass specific and are defined as the rate of change of boron fixed in *L. minor* tissue per unit average biomass over the time interval in question. Specific uptake rates for boron can be calculated according to the general formula:

$$\text{Specific uptake rate for boron} = \frac{\frac{[B_{t_1}] M_{t_1} - [B_{t_0}] M_{t_0}}{M_{t_1} + M_{t_0}}}{2} \bigg/ \Delta t, \quad (1)$$

where $[B_{t_0}]$ is the tissue boron concentration at time t_0 , M_{t_0} is the mass at time t_0 , Δt is the time interval in question, and $\frac{M_{t_1} + M_{t_0}}{2}$

is an approximation of the average standing-crop biomass over the interval (cf. Young and King, 1973). The estimation of the average standing crop biomass over an interval becomes more accurate as the interval becomes shorter. Calculations, using the laboratory data, show that the mean specific uptake rates for boron and their standard deviations at one week intervals ($\Delta t = 1$ week) are 159 ± 54 , 1022 ± 189 , and 1601 ± 260 $\mu\text{g B per gram ash-free dry weight}$ from the lowest to highest ambient levels respectively.

In order to more fully interpret the tissue boron response presented in Figure 1, the effect of rapid growth in the cultures must be considered. Growth rate, discussed here in terms of doubling time of plant biomass, reflects attendant internal physiological and external environmental conditions that can affect the specific rate of boron uptake. The biomass doubling time here was at or near the maximum for the species as indicated by the literature. The exclusive

consideration of growth in this discussion is the kinetic effect of growth acting to offset the effect of the specific uptake rate on the observed tissue concentration of boron. Some insight can be gained into the data presented here from a general discussion of the interaction of these two factors.

In the experiments of Figure 1, the specific uptake rate for boron was maintained at each experimental concentration by providing a constant supply of available ambient boron in flow-through cultures. Further, the tissue and ambient boron correlation was developed under conditions of rapid organic matter accrual. A stylized representation of the relationship between these variables is presented in Figure 3, where the ambient boron concentrations cause the specific uptake rates for boron to be constant. The figure implies that the tissue boron concentration of plants doubling in biomass at a given rate will approach a level reflecting the ambient boron level. Plants doubling at a faster rate at that ambient concentration will yield a proportionally lower tissue boron content. The figure suggests that the relationship between tissue and ambient boron is dependent upon the amount of growth over the interval in question.

The basis of the relationships illustrated in Figure 3 can be shown mathematically using an algebraic transformation of equation (1):

$$\left[B_{t_1} \right] = \frac{\left[B_{t_0} \right] M_{t_0} + \left(\frac{M_{t_1} + M_{t_0}}{2} \right) \left(\begin{array}{c} \text{specific uptake} \\ \text{rate for boron} \end{array} \right)}{M_{t_1}}, \quad (2)$$

where $\Delta t = 1$.

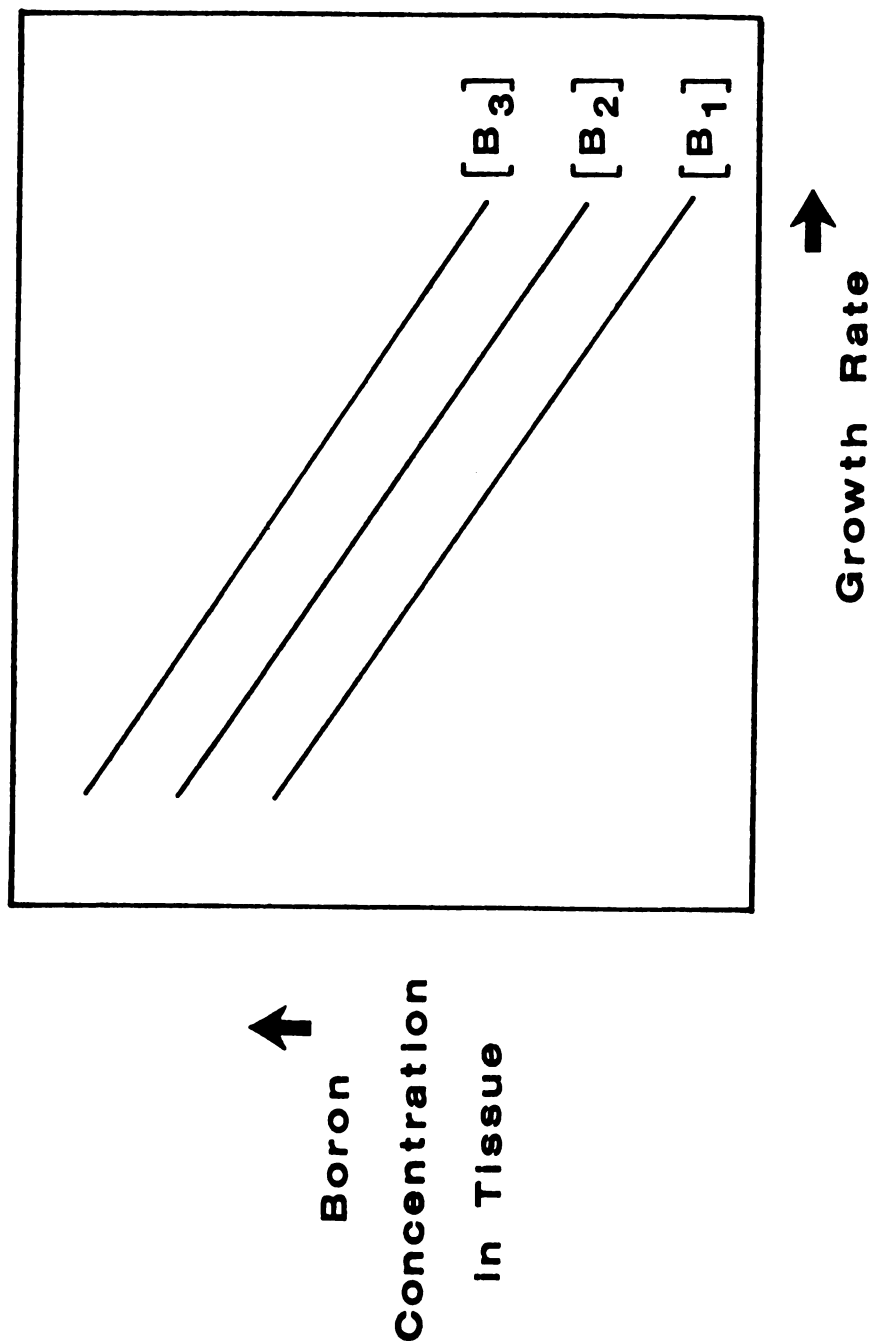


Figure 3. Stylized representation of the relationship between tissue concentration and growth rate at ambient boron concentrations where $[B_1] < [B_2] < [B_3] < [B_{\text{toxic}}]$.

According to this expression, the tissue concentration of plants taking up boron at a given rate, and doubling in biomass at a particular rate (*i.e.*, $\frac{M_t}{M_{t-1}} = \text{a constant}$), will approach a level set by these rates. This can be seen by assigning values for the uptake rate and biomass doubling time and solving for $\left[B_{t_1} \right]$ over successive equivalent time intervals. Furthermore, plants doubling at a slower rate (*i.e.*, doubling time is greater) at a given ambient boron concentration will yield a greater tissue boron content. These relationships assume that the supply of boron in a form available for uptake is constant.

The above considerations are useful in the interpretation of the mid-summer decline in tissue boron concentration in *L. minor*, and the strikingly high late-summer levels observed in this species from the Belding Pond. The data presented in Table 1 show that the decline in boron concentration in *L. minor* of approximately 50% corresponded to the period of its most rapid growth. The population of the pond increased from 1 to 126 kg organic weight during the interval of boron decline. The biomass doubling time for that two week period was calculated to be 2.00 days. As the summer progressed, the *L. minor* growth rate was reduced to the range of 24 days doubling time and after two months the tissue concentration rose to near $3200 \mu\text{g g}^{-1}$ organic dry weight.

The drop in tissue boron observed in *Ceratophyllum demersum* during the mid-summer interval does not correspond to the period of its most rapid growth. This observation points to a second consideration regarding the interpretation of the mid-summer change in tissue levels of that species. The customary formation of a mat of vegetation

by *C. demersum* has the effect of reducing wind driven circulation of the ponds, compartmentalizing the water strata and reducing the rate of supply of available nutrients to the canopy of the vegetation (cf. Wetzel, 1975). The boron stripping of an expanding biomass of *L. minor* apparently reduced the rate of supply of available boron to *C. demersum*. Ensuing production of the latter with a lowered ambient supply appears to have caused the drop in tissue boron concentration in *C. demersum*. It is hypothesized that in the absence of the hovering *L. minor* population the summer curve of *C. demersum* tissue boron would have bridged the mid-June and mid-July points on the curve of Figure 2.

A most conspicuous feature of this study is the disclosure of the boron accumulating ability of *L. minor*. The late summer concentrations in the material from the pond at Belding, Michigan provide especially dramatic contrasts to literature values.

Caution must be used when making comparisons between species because all species do not draw on the same boron supplies. Emergent plants likely absorb most of their boron from the hydrosol. Floating-leaved species probably absorb a large proportion of their total uptake from the sediments, but also absorb boron from the water. Relatively high evapotranspiration rates in both types may influence the concentrations observed in leaf or stem tissue. Submersed plants which lack or have greatly reduced root systems obtain most of their boron from the water (Hutchinson, 1975). Comparisons between specimens taken from different localities at different times of the year should also be made with reservation. The general trends observed have been dicotyledons usually contained more boron than monocotyledons and that

floating-leafed species contained more boron than submersed or emergent plants (Boyd and Walley, 1972; Cowgill, 1974).

Boyd and Walley (1972) working in Par Pond examined 22 species of macrophytes and found a range of $1.2\text{--}11\ \mu\text{g g}^{-1}$ B by dry weight in tissues. There was $0.005\ \text{mg l}^{-1}$ B in the water. Comparisons can be made with data of this paper by assuming an ash content of about 20% of dry material. Rooted, floating leafed species, *Nelumbo lutea* and *Nymphaea odorata*, contained the highest amount of boron with 10.9 and $11.3\ \mu\text{g g}^{-1}$ respectively. Submerged species, *Ceratophyllum demersum*, *Utricularia inflata*, and *Myriophyllum heterophyllum*, contained 4.3, 7.6, and $10.6\ \mu\text{g g}^{-1}$ B respectively. Cowgill (1974) analyzed 5 species from Cedar Lake ($0.395\ \text{mg l}^{-1}$ B in the water) and 7 species from Linsley Pond ($0.485\ \text{mg l}^{-1}$ B in the water). The range of boron in all plants examined was $27\text{--}65\ \mu\text{g g}^{-1}$ B by dry weight, with the floating-leafed species containing the highest amount at both sites. Ahl and Jönsson (1972) examined 6 emergent species taken from a "nutrient rich" bay of Lake Mälaren in Sweden. They reported a range of $4\text{--}19\ \mu\text{g g}^{-1}$ B by dry weight in tissue. Analyses made during this study of species of submerged plants collected in the fall from sites in central lower Michigan revealed a range of $38\ \mu\text{g g}^{-1}$ B by dry weight for *Myriophyllum spicatum* to $61\ \mu\text{g g}^{-1}$ B dry weight for *Elodea canadensis*. All of these low values highlight the efficacy of *L. minor* with respect to boron accumulation.

In addition to data presented for *C. demersum* as results of this study, plants collected from the series of wastewater ponds at Belding in September, 1972 were analyzed for boron. These data are presented in Table 2 for comparison of the relative absorbtive

Table 2. Boron concentrations expressed as $\mu\text{g g}^{-1}$ ash-free dry weight¹ of macrophytes collected from the Belding system in September, 1972.

Species	Location	Average % Ash	$\mu\text{g B per gram}$ Ash-Free Dry Weight
<i>Ceratophyllum demersum</i>	Pond 3	19.0	84
	Pond 4	19.5	52
	Pond 5	18.2	51
<i>Lemna minor</i>	Pond 3	19.9	1692 ²
	Pond 4	21.6	1521 ²
	Pond 5	17.7	1332 ²
<i>Lemna trisulca</i>	Pond 4	26.4	2904 ²
	Pond 5	20.8	2600 ²
<i>Potamogeton foliosus</i>	Pond 4	20.9	37
<i>Potamogeton berchtoldi</i>	Pond 5	13.0	57
<i>Potamogeton pectinatus</i>	Pond 5	14.4	327 ²

¹Boron analyses made by emission spectrograph.

²Values above the standardized range of analysis.

capacities of plants. Albeit, certain of these values are beyond the sensitive range of the analytical method used, the species of *Lemna* are outstanding as boron accumulators. It is particularly important to note that the submerged-growing species, *L. trisulca*, contained boron concentrations of the same order of magnitude as the pleustonic *L. minor*. Ability to concentrate boron appears to be a feature of species within the genus *Lemna*, irrespective of differences in the portion of the environment occupied during growth.

Proficiency in boron stripping coupled with its high growth rate distinguishes *Lemna minor* as an important species with respect to boron cycling in a freshwater macrophyte community. The effectiveness of this species in consuming boron may be a potent force in lowering the concentrations of this essential element in aquatic systems. The potential of *L. minor* to modify the environment with regard to boron so as to make it suitable for sensitive species that would otherwise be unable to tolerate high boron levels also has possible ecological significance. These considerations make the uptake of boron by *Lemna minor* worthy of further study.

CONCLUSIONS

The hypothesis that the tissue boron concentration of *Lemna minor* L. is relative to the ambient boron concentration was examined in the laboratory. The results of repeatable laboratory experiments showed that ambient boron concentrations of 0.01, 0.11, and 1.01 mg l⁻¹ B were significantly different in their effects on the tissue boron concentration of *L. minor*. The data indicate that ambient boron concentration was a determining factor in rate of boron accumulation in plant tissue.

Field samples of *L. minor* and *Ceratophyllum demersum* L., collected over the growing season from a wastewater pond in Belding, Michigan showed that the tissue boron concentration of *L. minor* was tenfold that of *C. demersum* throughout the season. Further, the data indicate that the kinetic effect of growth acts to offset the effect of boron uptake rate on the observed tissue boron concentration.

The most conspicuous feature of this study is the characterization of *L. minor* as a bio-accumulator of boron concentrating it to levels an order of magnitude above those reported for other species of aquatic macrophytes. The results suggest that *Lemna minor* is an important species with respect to boron cycling in a freshwater macrophyte community.

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