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An Analysis of the Aquatic Macrophyte, *Myriophyllum exalbescens*, as an Indicator of Metal Contamination of Aquatic Ecosystems near a Base Metal Smelter

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Aquatic macrophytes are known to concentrate metals in their tissues. Many researchers have suggested using aquatic vascular plants as metal pollution indicators (e.g. SCULTHORPE 1967, ADAMS *et al.* 1973, RAY and WHITE 1979). RAY and WHITE (1979) suggested the following criteria for use of plants as biological indicators of metal contamination. The plant should: 1) be representative of the area, 2) be ubiquitous and easily collected, 3) be easily identified unequivocally and 4) have a high tolerance of metals and a high concentration factor.

The purpose of this study was the selection of an aquatic plant suitable for assessing the metal burden, particularly Zn, Cu and Cd, of aquatic ecosystems in the vicinity of a base metal smelter located at Flin Flon, Manitoba, Canada. Following selection of a potential indicator plant from among five species collected from a contaminated and an uncontaminated lake, its suitability as an indicator was tested by analysing the metal concentrations of samples of the species' populations in six lakes having a range in metal contamination of water and sediment.

MATERIALS AND METHODS

Study Area

The Flin Flon area is situated on the Manitoba-Saskatchewan border at approximately 55°N, 102°W. Detailed descriptions of the study area may be found in McFARLANE and FRANZIN (1978) or FRANZIN *et al.* (1979). The six lakes chosen for this study are situated on the Precambrian Shield near a base metal smelter at Flin Flon (Figure 1) and receive varying amounts of metal deposition (FRANZIN *et al.* 1979). Physical and chemical descriptions of the lakes are presented in Table 1. Five aquatic plants - Bur Reed (*Sparganium* sp.), Bladderwort (*Utricularia vulgaris*), Water Milfoil (*Myriophyllum exalbescens*), Water lily (*Nuphar variegatum*) and Water Arum (*Calla palustris*) - were considered as potential indicators of metal contamination of aquatic ecosystems in the area.

Field

All water sample methods are described in McFARLANE *et al.* (1979).

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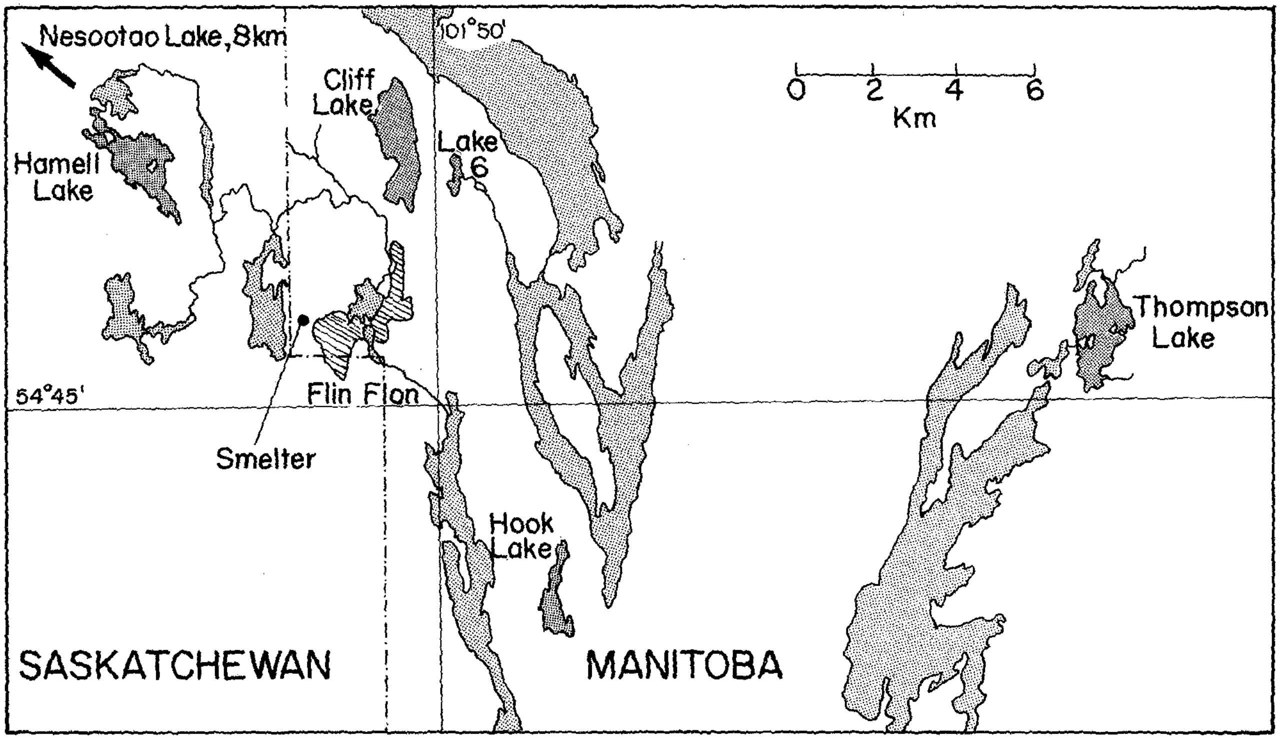

Figure 1. Study area, near Flin Flon [illegible]

TABLE 1

Physical descriptions and selected water chemistry parameters† of the study lakes.

Lake	Hamell	Cliff	Hook	Nesootao	Thompson	Lake 6
Surface area (ha)	233	246	90	40	220	19
Distance from smelter (km)	4.2	4.0	9.5	12.5	20.2	5.2
Maximum depth (m)	6	30*	12*	10*	12	5
pH	7.8	8.1	8.4	8.2	7.9	8.0
SO_4 (mg/L)	13.6	14.4	15.6	12.2	5.6	11.8
Cl (mg/L)	3.2	2.0	3.2	0.8	1.4	1.4
Ca (mg/L)	14.9	16.2	38.8	10.0	14.7	15.0
Mg (mg/L)	2.98	3.97	7.73	2.37	4.30	3.64
Na (mg/L)	1.30	1.78	2.35	1.67	1.93	0.97
K (mg/L)	1.48	1.51	1.48	1.31	1.34	0.91

† All chemical parameters measured August, 1976.
* Estimated.

Sediment cores were taken by a SCUBA diver using 4.5 x 50 cm plexiglass tubes which were thrust by hand into the sediments until either the tube was nearly full or hard pan was contacted. Then, while still submerged, the diver capped both ends of the tubes. Cores were maintained in an upright position in a sample box, taken to a field camp and frozen prior to their transport to the Freshwater Institute. Three core samples were taken in each of Hamell and Thompson lakes and two cores each from the other four lakes. All were taken within a 3-day period in August 1976.

Only leaves and stems of submergent plants or submerged portions of plants were collected. Plant samples were cleaned as well as possible in lake water then put into clean polyethylene bags. At the field camp, the plants were washed in Flin Flon tap water, rinsed twice with distilled water, then frozen for transport to the Freshwater Institute. All plant samples were obtained during one week in August of 1975 and 1976.

Laboratory

All methods of water preparation and analyses are described in McFARLANE *et al.* (1979).

Sediment cores were removed from the collection tubes while partially frozen, cut into 5 cm lengths from sediment surface to a depth of 20 cm, air dried for 24 hr at 80 C and pulverized in a porcelain mortar. One gram portions of dried, ground and sieved sediment were weighed into 125 mL Erlenmeyer flasks. Ten mL of aqua regia (HNO_3/HCl = 1/3) was added to each flask and heated to boiling. Samples were boiled for 2-3 minutes, cooled and made to 50 mL with distilled water.

Plants were air dried for 24 hr at 80 C, then pulverized in a Wiley micromill. Duplicate 0.5 g portions were weighed into 25 x 200 mm digestion tubes to which were added 4 mL concentrated HNO_3 and 2 mL concentrated $HClO_4$. The tubes were heated in an aluminum block to $HClO_4$ fumes, cooled and made to 25 mL with distilled water.

Sample Analyses

Aliquots or diluted aliquots of the above digests were analysed for Fe, Mn, Pb, Cu, Cd and Zn by flame atomic absorption spectrophotometry using a Varian AA-5 atomic absorption spectrophotometer equipped with a BC-6 simultaneous background corrector. U.S. National Bureau of Standards Reference Material Bovine Liver 1577 was analyzed along with the samples and the results were within the specific tolerances for each metal. The table below indicates the metals and their practical detection limits by these methods.

Element	Cd	Cu	Zn	Fe	Mn	Pb
Wavelength (μm)	228.8	234.7	213.9	248.3	279.5	217.0
Detection limit (μg/g)*	0.08	0.25	0.2	0.8	0.2	1.0

* Detection limit is defined as the concentration of an element in a sample which will give a signal equal to twice the standard deviation of a series of measurements near to the blank level.

Arsenic Analysis

One half gram portions of pulverized plant material or sediment were digested with a mixture of $H_2SO_4/HNO_3/HClO_3$ (2/5/0.05) in an aluminum block to SO_3 fumes. The solutions were cooled, made to 25 mL and aliquots placed in sample cups. The analysis was by a modification of the semi-automated borohydride reduction method of VIJAN and WOOD (1974). The modification was a heated quartz cell with a quartz window and an inlet tube at the same end. The detection limit was 0.02 μg/g.

RESULTS AND DISCUSSION

Selection of an Indicator Plant

Table 2 shows metal concentrations of five plant species obtained from a contaminated lake (Lake 6) and a relatively uncontaminated lake (Thompson Lake). Zinc is the major metal found in fallout on the lakes but Cu, Cd, Pb and As in the fallout also originate from the smelter

TABLE 2

Metal concentrations of samples of five species of plants from two Flin Flon area lakes.

Lake	Plant sp.	Metals (μg/g dry wt.)						
		Zn	Cu	Cd	Pb	As	Mn	Fe
Lake 6	*Sparganium* sp.	1460	63	5	27	25	194	3010
Thompson		54	38	<0.2	<0.9	-	591	2390
Lake 6	*Utricularia vulgaris*	1330	36	14	17	20	136	1340
Thompson		134	20	0.7	<0.6	-	1050	188
Lake 6	*Myriophyllum exalbescens*	1640	66	6	27	40	203	1460
Thompson		78	32	<0.1	<0.6	-	92	1390
Lake 6	*Nuphar variegatum*	274	7	0.6	2	6	69	174
Thompson		11	4	<0.1	<0.6	0.9	73	264
Lake 6	*Calla pallustris*	389	9	4	2	5	46	1040
Thompson		190	11	<0.1	<0.6	1	190	1010

(FRANZIN *et al.* 1979). Zinc provided the best indication of the presence of smelter fallout contamination in lakes but a summation of the concentrations of all the smelter-related metals in the plant tissues was used for an integrated evaluation of the plants as potential indicator species. This criterion plus the others detailed in the introduction led to the selection of *Myriophyllum exalbescens* as our indicator species. It was apparent that the surface area to volume relationship of these aquatic plants was an important determinant of metal uptake (Table 2). Fleshy, heavily rooted plants such as *Nuphar* and *Calla* had about one fifth the metal concentrations in submerged leaves and stems as the ribbon-leaved or profusely-foliated plants such as *Sparganium*, *Utricularia* and *Myriophyllum*.

Myriophyllum exalbescens as an Indicator Species

Table 3 shows the estimated deposition of metals (FRANZIN *et al.* 1979) on the six study lakes and the resultant contamination of waters and sediments. Since Zn was the major metal in the deposition, the lakes were ranked in order of decreasing amounts of Zn in deposition and by Zn concentrations in waters and sediments. Pb and As in grab samples of lake waters were generally <5 µg/L, well below concentrations of toxicological importance, so these metals were not routinely monitored. Cd, although also in low concentrations in the lakes, is known to be extremely toxic in aquatic systems and was expected to have a potential effect on aquatic organisms in the lakes. Fe and Mn data are included as indices of similarity of the lake environments and were not associated with smelter fallout. Table 4 shows metal concentrations, ranked in order of decreasing Zn concentration, of leaves and stems of submerged portions of *Myriophyllum exalbescens* samples collected from the six study lakes in 1976, and, for comparison, samples collected from Lake 6 and Thompson Lake in 1975. It contains two interesting results. Firstly, there were large inter-year differences in metal concentrations of *Myriophyllum* samples collected from Lake 6 and Thompson Lake in 1975 and 1976. The only variable with which these differences could be associated was a nearly two-fold difference in rainfall during the growing periods of the two years (179 mm in 1975 vs 321 mm in 1976). Increased rainfall in 1976 may have increased movement of metals from drainage basins into the lakes and thereby increased the concentrations in aquatic plants. The difference in the magnitude of the response between the two lakes may be due to differences in vegetative cover of the drainage basins and in lake volume. Thompson Lake is much larger than Lake 6 (Table 1) and has a well-vegetated drainage basin. The drainage basin of Lake 6 was largely burned-off many years ago and now consists of small bogs with large areas of bare bedrock so that transport of materials into the lake by precipitation run-off may have been enhanced. Secondly, comparison of plant metal concentrations with the metal concentrations in lake waters and sediments in Table 3 shows that a fair correspondence exists only for Zn in water. This suggests that this plant may indicate qualitatively the degree of Zn contamination of aquatic environments if the contamination is severe. However this is not necessarily so for most trace metals;

TABLE 3

Metal deposition on six Flin Flon area lakes and metal concentrations in water and sediments of the lakes.

Lake	Metal						
	Zn	Cu	Cd	Pb	As	Mn	Fe
				Deposition $mg.m^{-2}.y^{-1}$			
Cliff	1400	62	3	32	6	-	-
Hamell	1290	58	3	31	6	8	241
Lake 6	889	42	2	24	4	-	-
Hook	313	27	2	27	3	4	74
Nesootao	195	11	0.7	9	0.9	-	-
Thompson	87	5	0.4	5	0.4	5	36
				Water $\mu g.L^{-1}$			
Lake 6	223	10	0.3			11	-
Hamell	220	14	0.7			26	69
Cliff	124	11	0.4			5	-
Nesootao	80	7	0.3			14	-
Hook	45	4	0.6			11	-
Thompson	15	20	<0.1			41	-
				Sediment $\mu g.g^{-1}$ dry wt.*			
Lake 6	3770	902	28	575	206	889	13500
Hamell	1990	442	16	274	72	175	8150
Hook	987	242	6	131	39	342	17800
Cliff	803	174	5	72	22	291	22300
Thompson	133	188	0.8	225	5	348	16400
Nesootao	110	29	0.8	24	9	113	4640

* Sediment concentrations are mean values of 0-10 cm for all cores collected from each lake.

in this instance Cd, Cu, Pb and As. This observation coupled with our observed high year to year variance in plant metal concentrations (apparently linked with climatological factors) suggests that analyses of aquatic plants cannot be relied upon to distinguish grades of metal contamination in their habitats which can be seen in water, sediment or fallout analyses.

Our data indicated plant metal concentrations which were mainly contrary to expected concentrations given our knowledge of the degree

TABLE 4

Metal concentrations in submerged leaves and stems of *Myriophyllum exalbescens* from six Flin Flon area lakes.

Lake	Metal $\mu g.g^{-1}$ dry wt.						
	Zn	Cu	Cd	Pb	As	Mn	Fe
				1975			
Lake 6	1640	66	6	27	40	203	1460
Thompson	78	32	<0.1	<0.6	-	92	1390
				1976			
Lake 6	12300	346	31	242	263	6270	8070
Hamell	1790	54	7	44	40	3150	1810
Nesootao	1470	75	9	24	32	4680	6040
Cliff	1380	90	10	20	48	1000	7980
Hook	848	21	4	15	13	2670	1490
Thompson	185	80	1	4	4	864	1510

of contamination of different lakes (e.g. compare Hook and Nesootao lakes in Tables 3 and 4). Therefore we attempted to determine causal relationships between environmental variables (Tables 1, 3) and plant metal concentrations. No correlations of plant metal concentrations and any environmental variables were apparent. However, it seemed that two unrelated conditions affected plant metal concentrations, namely the amount of deposition of smelter metals and Ca concentration of the lake waters. For example, Nesootao Lake was relatively uncontaminated, had a Ca concentration of 10 mg/L and high plant metal concentrations (as high as the most contaminated lakes) whereas Hook Lake was highly contaminated by deposition and sediment concentrations, had a Ca concentration of 39 mg/L and low plant metal concentrations. The effect of different levels of contamination at similar Ca concentration is clearly shown by comparison of Hamell and Thompson lakes (Tables 1, 3, 4). These observations, while not amenable to statistical validation suggest that the Ca concentration of lake waters may have a modifying role on plant uptake of all metals (see also a similar effect on Mn) whether essential or non-essential for survival and growth. Laboratory studies (CARROLL *et al.* 1979, KINKADE and ERDMAN 1975) have shown that uptake and toxicity of Cd in fishes was modified by Ca ion alone rather than the $CaCO_3$ system suggesting that this effect may be biological rather than the result of water chemistry. Our study on plants reported here, along with some observations on Cd uptake in two species of fish in these lakes and metal concentrations of plankton samples (FRANZIN and McFARLANE, unpublished data) suggest that this effect of Ca may be a general one. The mechanism is poorly understood and requires further research.

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