

The role of heavy metals and toxic materials in the physiological ecology of submersed macrophytes

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ABSTRACT

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An overview of, and information from, a number of publications dealing with the availability, uptake rates and accumulation of metals (e.g. Fe, Cu, Cr, Cd, Pb) and organic substances (e.g. polychlorinated biphenyls (PCB) and pesticides) by submersed and floating macrophytes are provided.

Differences in metal enrichment are reported to be dependent mainly on the plant species, the seasonal growth rate changes, the tissue age, and metal or compound type. A brief review of the possible sites of absorption and accumulation of substances by the macrophytes is given and the role played by the roots and shoots is discussed. A higher concentration of metals in the plants usually, but not always, indicates a proportional increase in element levels in the water and/or sediments. Consequently, the implications for the use of macrophytes as phytogeochemical indicators for metals and other toxic materials in water bodies are also evaluated. Competition between elements and factors, indirectly influencing metal uptake such as, for example, light conditions and eutrophication, seem to limit in some cases the utility of using macrophytes as bio-indicators.

Further, the effects of metals, particularly Cu and Cr, and a few other organic compounds on growth and photosynthesis of some species are described.

In considering all these aspects, the importance of several environmental factors (e.g. pH, water hardness, nutrient concentration) in influencing the macrophyte–toxic compound cycle occurring in aquatic systems has been emphasized. It has been noted how contradictory results often emerge because of the large differences existing both in sampling time and in the methods used by different working groups.

As a general rule, it seems extremely difficult to extrapolate field results from one species to another.

INTRODUCTION

Recently there has been an enormous growth of interest in the significance of trace elements in aquatic environments. The reasons for this are as follows.

(1) The potential hazard for aquatic organisms from the continuous release of large amounts of heavy metals into water systems is great. Heavy

metals pose a serious water pollution problem because of their extreme toxicity. To ascertain the extent of the pollution, macrophytes have been widely used as bio-indicators, as they are able to integrate and rapidly monitor variations in the concentrations of the elements in water.

(2) The role played by aquatic plants in the biogeochemical cycling of elements is important. The many questions about the processes of metal uptake, the amount, forms, and sites of element release, and their toxic effect on plant metabolism are of growing interest to researchers.

(3) The availability of new analytical instruments utilizing techniques such as atomic absorption spectroscopy, inductively coupled plasma, X-ray fluorescence, pulse polarography, etc. increases the precision for studying heavy metal and toxic material cycling.

Most recently, scientists have devoted more attention to toxic compounds such as pentachlorophenol (PCP), a fungicide and bactericide; 2-chloro-4-[ethylamino]-6[-isopropylamino-]-s-triazine (atrazine), a powerful herbicide used for agricultural weed control; polychlorinated biphenyl (PCB); and some pesticides.

To test the effects of toxic materials, a number of organisms are often employed in laboratory experiments from the most simple and sensitive, such as bacteria and algae, to the more complex, such as bivalves, zooplankton, fish and macrophytes. Although relatively neglected, macrophytes are useful for biomonitoring because they do not migrate and they quickly reach equilibrium with their water environment (Muntau, 1981). Currently, heavy metal pollution is often described in terms of the tolerance and sensitivity of some of the macrophytes used as bio-indicators (Haslam, 1978; Haslam and Wolseley, 1981; Kovács and Podani, 1986).

Among the classical works in this field both laboratory studies and field research have been conducted (Boyd, 1970; Gerloff, 1973 and 1975; Cowgill, 1974; de Marte and Hartman, 1974; Hutchinson, 1975; Denny, 1980). Most of the in situ studies were descriptive and the data on element accumulation not always easily comparable between different situations.

In addition, studies have been conducted to verify the role played by aquatic plants in the fate of elements after their release into the littoral zone as well as the relative importance of macrophytes acting as a filter or as pollution source for the plankton in open waters (Howard-Williams and Lenton, 1975; Rawlence and Whitton, 1977; Beeton and Sikes, 1978; Prentki et al., 1979; Ravera et al., 1983).

This paper summarizes the relationships between macrophytes and toxic substances. Heavy metals have received greater attention because more is known about them. Nevertheless, some results regarding other pollutants are included.

AVAILABILITY, MOBILITY AND TOXICITY OF SOME SUBSTANCES

Heavy metals

The availability of trace metals for plant metabolism is related to their chemical forms in superficial and pore water, and to their availability in particulate matter. For this reason, material availability in both water and sediment is considered.

Most of the research performed on heavy metals has assessed the total concentration present in a water body. Although it is quite useful for establishing a pollution status, total concentration does not tell us much about the actual availability and the level of toxicity for aquatic organisms. It is clear that the environmental impact of a particular chemical species is more important than the total metal concentration (Leppard, 1983). Unfortunately, our knowledge of the interaction between trace metals, water, sediment and aquatic plants is still incomplete. This lack of information is due to the complexity of studying all the environmental factors which affect the availability of metals. These include the chemical species, the plant uptake and accumulation, and finally the metabolic activity of macrophytes.

What are the ecological factors affecting specific metal configuration and the presence of these more or less toxic chemical forms? The answer to these questions is important for interpreting results derived from ecotoxicological studies.

The behaviour of metals in aquatic ecosystems is connected to the role of organic matter in processes such as sorption and/or chelation/complexation of metals. Because of the large number of functional groups (e.g. humic and fulvic acids), dissolved organic matter plays an important role in the bonding of heavy metals. Complexation is the single most important abiotic factor in reducing metal toxicity (Salomons and Förstner, 1984).

In the aquatic system, trace elements also form highly stable complexes with clay and silt particles (Förstner and Prosi, 1978). Once the metals are bound with organic and inorganic compounds they can more or less rapidly settle to the sediments. In shallow waters, turbulence can resuspend the sediments making (under favourable conditions) the elements available to macrophytes and algae.

In general, the availability of most metals is dependent on the following factors.

pH. pH values between 3.7 and 4.5 favour the dissolution of carbonate and hydroxide minerals, freeing the associated metals. Most trace metals are more toxic to plants at lower pH levels (Lepp, 1981).

Nutrient concentration. Oligotrophic alpine lakes usually have a higher heavy

metal concentration than unpolluted lowland lakes (Williams et al., 1974). In alpine lakes there are greater quantities of humic substances to solubilize the metals, and they lack significant quantities of dissolved salts to react with and precipitate metal ions. In eutrophic lakes, very high local concentrations of metals often occur as a result of the strong reducing environment coupled with industrial and municipal discharges containing metals (Williams et al., 1974).

Redox potential. An anoxic sediment–water interface typically exhibits a negative potential and easily releases metals (e.g. Fe, Cu, Zn, Cd).

Salinity. In estuarine environments the decrease in heavy metals concentration may be partially due to the competition between dissolved cations and adsorbed heavy metal ions (Förstner and Prosi, 1978).

Water hardness. Some metals (e.g. Cd) are more toxic to plant species in soft water (approximately $20 \text{ mg CaCO}_3 \text{ l}^{-1}$) than in hard water (approximately $300 \text{ mg CaCO}_3 \text{ l}^{-1}$) (Munovar et al., 1988). In general, the toxicity of metals is reduced in high carbonate waters because inorganic metal carbonates are less toxic than other metal species (Salomons and Förstner, 1984).

Light. Laboratory experiments with algae (no data are available on macrophytes in this respect) show that light enhances the toxicity of metals (Munovar et al., 1988).

Microbial activity. In general, microbial activity enhances the release of metals and is responsible for the methylation of mercury.

Physical factors such as resuspension through wind action, or from human activities (dredging, dredge spoil, navigational activities, etc.) and biological factors such as bioturbation, can mobilize trace metals (Munovar et al., 1988).

Organic herbicides, anionic detergents and the growing use of synthetic complexing agents such as nitrilacetic acid (NTA) in detergents can influence metal solubility and thus affect metal uptake by plants (Sutton et al., 1971; Muramoto and Oki, 1984).

Rubin (1974), Förstner and Prosi (1978), Förstner and Wittman (1979), Bowen (1979), Waldron (1980) and Salomons and Förstner (1984) provide detailed descriptions of these processes.

The above-mentioned factors are very important in metal distribution in water and sediments and thus their availability to aquatic macrophytes. It is equally important to realize that toxic elements (naturally occurring or synthesized industrially) are classified into three groups according to their pollution potential (Wood, 1974); (i) non-critical (Fe, Rb, Sr, Al); (ii) toxic

but very insoluble or very rare (Ti, Hf, W, Zr, Ta, Nb, Re, Ga, La, Os, Rh, Ir, Ru, Ba); (iii) very toxic and relatively accessible (Be, Co, Ni, Cu, Zn, Sn, As, Se, Te, Pd, Ag, Cd, Pt, Au, Hg, Tl, Pb, Sb, Bi).

Other elements such as Mn, Cr, etc. fit into more than one category depending on their specific mobility or chemical form.

These metals may be incorporated in the trophic food net and then partially accumulated in the sediments. The concentration level depends on the sources (e.g. industrial waste stream or mineralogy of rocks present in the watershed) and element mobility (e.g. Fe and Mn dissolve and migrate in pore water much more quickly than Cd, Pb, Zn, Cr, Hg). Mobility is closely connected with solubility. For example, the Pb compounds and their organic complex are mostly concentrated in the sediments because they are generally insoluble in natural waters (Knowlton et al., 1983).

Other substances – herbicides, pesticides and PCB

The wide use of pesticides and herbicides also poses a potential pollution threat to inland waters. The degree of danger depends on the amount of these compounds in water, but the problem is much more complex.

The effect of pollution by organic substances depends on their physiochemical properties (solubility in water, residence time, form in which they are employed, half-life of products, break-down of products, etc.) and the hydrological characteristics of the receiving water bodies. Organochlorine pesticides such as DDT, chlorinated phenol and PCB are persistent and relatively insoluble in water (Vrochinskiy et al., 1970; Mouvet et al., 1985).

The triazine herbicides such as terbutryn, simazine and atrazine have a half-life in water of between 1 and 100 days (Muir et al., 1981). In estuarine environments, concentrations of atrazine higher than $10 \mu\text{g l}^{-1}$ generally persist in the water column for less than 1 day. The negative effect on plant metabolism under such conditions would be minimal and reversible (Jones et al., 1986). As shown in an experiment in ponds with and without aquatic macrophytes, the plant biomass is responsible for about a 35% reduction in levels of added atrazine and simazine. The persistence of the three compounds was similar and much greater than several other herbicides such as fluridone, diquat, endothal and dichlobenil (Muir et al., 1981).

Falkner and Simonis (1982) concluded that “the simple and popularly accepted scheme of chlorinated hydrocarbons (PCB) magnification in food chains by feeding relations is invalid”. The uptake of PCB by organisms is complex with partitioning equilibria between the abiotic environment and the more or less lipid-rich organisms playing a dominating role. Waldrom (1974), Audus (1976) and Schmidtke (1988) provide an exhaustive revision of the fate of these substances in the aquatic environment.

UPTAKE OF MATERIALS BY AQUATIC PLANTS

Mechanism of uptake – the role of roots and leaves

The uptake of trace metals by submersed macrophytes has been studied little. In particular, the relative contribution of roots vs. leaf uptake is not yet understood. For most elements and plant species, we only make hypotheses and assumptions, mainly based on the works of Sculthorpe (1967), Hutchinson (1975), Welsh (1977), Denny (1980), Muntau (1981), Gommès and Muntau (1981a) and Everard and Denny (1985). It is quite obvious that uptake depends on the chemical forms present in the system (Fig. 1) and on the life form of the macrophyte (floating-leaved, free-floating, well-rooted or rootless species). There is no question that free-floating plants such as *Lemna*, *Eichhornia* and *Pistia* take up elements from the water by roots and/or leaves (Sculthorpe, 1967; Hutchinson, 1975). Likewise, the rootless *Ceratophyllum* takes up elements mainly through its finely divided leaves. The situation of those species which have a well-developed root–rhizome system and totally submersed foliage such as *Myriophyllum*, *Potamogeton* and *Vallisneria* species is much more complex. Modern as well as classical experiments have demonstrated that these aquatic angiosperms extract nutrients and heavy metals mostly from the sediments via the root hairs with subsequent translocation to the upper parts (Bristow and Whitcombe, 1971; Welsh and Denny, 1979). However, uptake by leaves becomes important when the metal concentrations in the surroundings are high or when the metals are chemically bound in not readily available compounds (Denny, 1972; Nichols and Keeney, 1976).

The 'division of labour' between uptake by roots and shoots is probably associated with the anatomy and morphology of the different taxa (cuticle thickness, leaf cell layers), as well as by the sorptive capacities characteristic of species differing in growth rate, surface to volume ratio and physiological condition of individual plants (Denny, 1980; Franzin and McFarlane, 1980; Knowlton et al., 1983; Everard and Denny, 1985). For example, in senescing macrophyte tissues, there is evidence of increasing surface area and number of exposed Pb binding sites which increase uptake capacity for Pb (Sharpe and Denny, 1976; Odum and Drifmeyer, 1978). This metal (but also Cu) presumably becomes complexed to the anionic sites associated with pectic substances within the cell wall (the so-called Donnan-free space; Sharpe and Denny, 1976).

Moreover, it is likely that the controversy as to whether trace element absorption occurs primarily through the foliage from the water or through the roots from the sediments, is related to how essential the element is for plant growth. Stanley (1974) suggests preferential root uptake for essential ele-

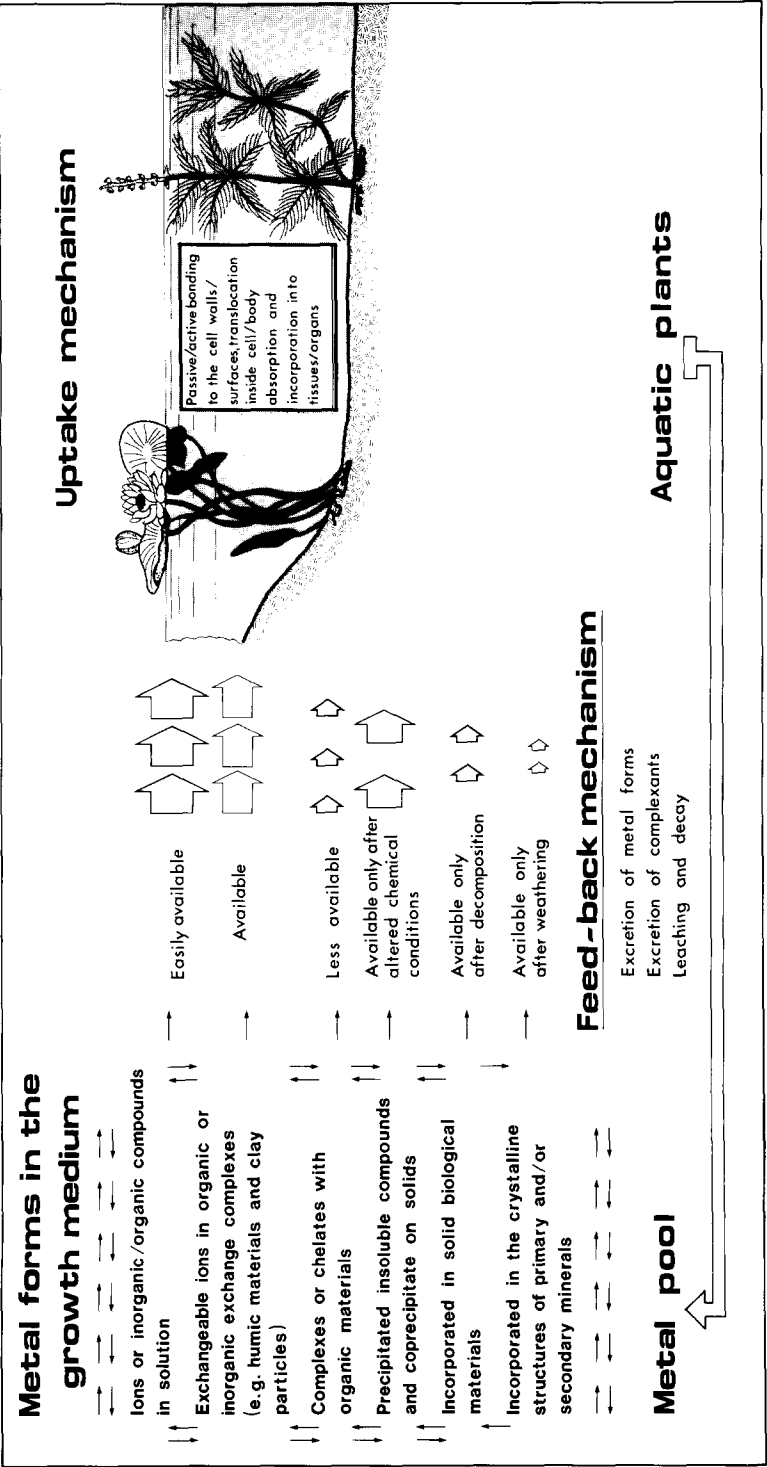


Fig. 1. Metal cycling between aquatic medium and submersed macrophytes (redrawn from Baudo, 1985).

ments (even when applied to toxic levels) but not preferential uptake for non-essential elements.

Briefly, not all epidermal cells are equally permeable to dissolved salts. In strictly submersed plants, a group of cells called the hydropoten are easily crossed by salts (Sculthorpe, 1967).

Evidence suggests that the uptake of ions by submersed leaves may be linked to the photosynthetic utilization of bicarbonates, and two different steps (bonding to the cell membrane and transfer inside) are involved.

In the light (but in some species also in the dark) cations generally enter the abaxial epidermis, although in several *Potamogeton* species the uptake of ions occurs in the adaxial surface (Sculthorpe, 1967). A multiple mechanism is involved: passive (simple diffusion) and active. The active mechanism for ion transport involves a kind of electrogenic pump supported by energy deriving from a metabolic process (e.g. NADH_2). The process can be described by Michaelis–Menten kinetics and the Nernst equation (Hutchinson, 1975). Passive uptake depends on pH, solid phase concentration, complex-forming ligands, and ionic strength of the water. Both passive and active uptake are based on diffusion throughout free space (e.g. pores) and by dissolution into a lipid layer (Baudo, 1985). At least for Pb, a vast proportion of the metal taken up by some *Potamogeton* species is simply adsorbed to sites in the cell walls according to the uronic acid theory (Knight et al., 1961; Welsh, 1977).

It is also likely that metal ion uptake is temperature dependent, as was noted for K^+ and Cl^- (Raven, 1967).

The role of Ca in uptake of metals also seems important. Franzin and MacFarlane (1980) found a significant negative correlation between plant metal accumulation and dissolved Ca, suggesting that Ca in solution might modify plant uptake of metals.

Indirect evidence of the mechanism involved in uptake of metals comes from in situ correlation between plant tissue concentration and corresponding environmental levels. Negative correlations are interpreted as the presence of active regulation of element uptake. However, this analysis should be treated with caution as the lack of significant correlations between plant, water and sediment levels may result from low concentrations of metals in the environment as well as from the different availability of metals (Muntau, 1981).

Uptake rates and distribution in plant tissue

Heavy metals

Uptake rates vary greatly according to the relative concentration of the element in the environment, the growth form of the plant, cuticle thickness, type of absorption mechanism, affinity of metals for the adsorption sites, metal speciation, metal stability and constants with ligands. Metal flux can also be affected by some external factors such as turbulence and quantity of 'Auf-

wuchs' present (Mayer et al., 1977). In general, at the beginning of the growth period, macrophytes show a rapid uptake of all the metals considered, with the exception of Cr.

From the many studies published on trace metal distribution and accumulation in plant organs, it appears that no clear correlation exists between heavy metal variability in water and in plants. Sampling intensity must be increased considerably to establish some clear relationships between element concentrations in water, sediments and macrophytes, and to define the actual rates of transfer from one compartment to the other.

A detailed in situ study on Cu, Ni, Pb, Zn, Cr, and Mn uptake in several submersed plant species was made by Gommès and Muntau (1981b) in Lake Maggiore (Italy) (Table 1). Uptake rates for Cu, Ni and Pb were uniformly low for most of the species investigated. The uptake rates of Zn, Cr and Mn were considerably higher, reaching exceptional values of $12.05 \mu\text{g g}^{-1} \text{ day}^{-1}$ for Mn in *Lagarosiphon major* (Ridl.) Moss and $66.52 \mu\text{g g}^{-1} \text{ day}^{-1}$ in *Egeria densa* Planch. Uptake rates of Pb as high as $314 \mu\text{g g}^{-1} \text{ h}^{-1}$, depending on Pb concentration and species were reported by Gommès and Muntau (1975a). In 6 h, the concentration of Cu increased from about $10 \mu\text{g g}^{-1}$ to about $40 \mu\text{g g}^{-1}$ dry weight in *Najas tenuifolia* R.Br. (Hart et al., 1983).

Welsh (1977) made a detailed study of Cu and Pb uptake rates using *Potamogeton crispus* L. and *Potamogeton pectinatus* L. The results show different uptake rates for the different species, plant tissue and metals. However, it was not possible to demonstrate different uptake mechanisms for the two metals; perhaps they became complexed to the anionic sites associated with pectic substances within the cell wall (Sharpe and Denny, 1976). As Pb and Cu have similar stability constants with ligands (Sillén and Martell, 1964), their absorption processes should be similar.

TABLE 1

Heavy metal uptake rates ($\mu\text{g g}^{-1} \text{ day}^{-1}$) in green tissue of some macrophytes of Lake Maggiore (Italy). Data from Gommès and Muntau (1981b)

	<i>Potamogeton crispus</i>	<i>Egeria densa</i>	<i>Lagarosiphon major</i> ¹	<i>Polygonum amphibium</i>	<i>Nymphoides peltata</i>	<i>Trapa natans</i>
Cu	0.08	0.21	0.23; 0.16	0.15	0.21	0.16–0.28
Ni	0.19	0.52	0.39; 0.52	0.55	0.53	0.38–0.53
Pb	0.10	0.43	0.11; 0.20	0.10	0.36	0.16–0.24
Zn	1.11	5.49	0.02; 4.92	0.72	1.22	0.56–0.93
Cr	0.35	0.64	0.38; 1.06	3.88	2.17	0.57–4.65
Mn	2.07	66.52	7.55; 12.05	1.44	1.88	3.27–3.75
Period	15/2–30/6	30/6–26/8	27/6–28/8	15/4–27/6	1/5–27/6	15/5–29/8
Days	135	57	60	73	58	

¹ Data from two different sampling stations.

Once the elements have been taken up by the macrophytes they are translocated and accumulated in different plant parts. Most aquatic species have a well-developed vascular system (Arber, 1963). Consequently, acropetal water flow very likely assists the upward transport of ions in the xylem. The reverse movements (basipetal) are also described in many cases and are probably dominant (Denny, 1980). For example, it is known that Hg is absorbed from water and transported to the root within *Elodea* (Eriksson and Mortimer, 1975). In *Vallisneria spiralis* L. the transport of ^{59}Fe occurred in both directions but was greater basipetally (Gentner, 1977) and Heisey and Damman (1982) reported evidence of translocation of small amounts of Pb in macrophytes.

Table 2 summarizes data on the rate of translocation of substances in aquatic plants. The marked variability of the results is due to species differences, age of the plant tissue, and nutrient characteristics of the environment (Welsh, 1977). In general, rates of metal absorption are lower in senescent than in actively growing plants (Mortimer, 1985). There are some exceptions in which

TABLE 2

Some data on the movement of substances in aquatic submersed macrophytes

Species	Translocation	Ref.
<i>Ranunculus trichophyllus</i>	3 cm h ⁻¹ (Li)	Pond, 1905
<i>Elodea</i>	4 cm h ⁻¹ (water + solution)	Forde and Steer, 1976
<i>Vallisneria spiralis</i>	0.5–4 cm h ⁻¹ (Cl)	Arisz, 1953, 1964
<i>Potamogeton crispus</i>	1 mm h ⁻¹ (Pb)	Sharpe, 1976
<i>Potamogeton foliosus</i>	No translocation of Pb ¹	Welsh and Denny, 1979, 1980;
<i>Najas guadalupensis</i> , <i>P. crispus</i> and <i>Potamogeton pectinatus</i>		Knowlton et al., 1983
<i>P. crispus</i> , <i>P. pectinatus</i> , 8 submersed species and Characeae	Translocation of Cu	Welsh and Denny, 1980
<i>Elodea canadensis</i>	Translocation of Cd and Pb	Mayer et al., 1977
<i>Heteranthera dubia</i>	Translocation of simazine	Funderburk and Lawrence, 1963
<i>Potamogeton nodosus</i> and <i>P. crispus</i>	No translocation of simazine and atrazine	Frank and Hodgson, 1964; Sutton and Bingham, 1968; Thomas and Seamon, 1968
<i>P. nodosus</i>	Translocation of endothal to phloem	Frank and Hodgson, 1964
<i>P. pectinatus</i>	Acropetal translation of fenac	Frank and Hodgson, 1964
<i>P. perfoliatus</i>	No translation of atrazine	Jones et al., 1986

¹Welsh and Denny (1976) and Mayer et al. (1977) reported evidence for some translocation of Pb in macrophytes. Movement of Pb from sediment to the foliage of macrophytes may occur through the water, perhaps through the desorption of Pb ions from sediment and their subsequent resorption in plant tissue (Sharpe and Denny, 1976).

rates of Cu and Pb absorption were higher in old and dead than in young leaves (Welsh, 1977).

Differences are clearly shown in element accumulation between various macrophyte organs. It appears that "the metabolic sink for essential ions is rhizomes or tuber for a mature to senescent plant, and apical growing tips for a young plant" (Welsh, 1977). Inflorescences are also rich in trace metals, particularly Cu (Guilizzoni, 1975; Baudo et al., 1981a). Lead is mobilized in the cell wall free space (Sharpe, 1976; Welsh and Denny, 1980). Stems are usually poor in metals although Bidwell and Foreman (1957) found for *Myriophyllum spicatum* L. that ^{90}Sr was accumulated in stem tissue rather than in leaves.

To summarize, the heavy metals present in an aquatic plant are generally determined by the following.

(1) The availability of the trace metals, which is defined by chemical species caused by abiotic parameters.

(2) The intrinsic biology of the organism which regulates the net uptake of the metals. In macroalgae the accumulation rates of Fe, Mn, Cr, Ti, Cu, Zn, Ni, Co and Mo are thought to be determined by genotypical specificity (Koryakov and Saento, 1981).

(3) The biological behaviour of the plant, which is expressed in its life cycle and life history, and by the trophic situation in which the plant grows.

In the ecosystem, macrophytes can act as either a source or sink for materials reaching the water body. In Lake Maggiore (Italy), a sink action rather than a source for plankton in open water was observed (Muntau, 1981). An example of this was given by the Mn-collecting floating-leaved species *Trapa natans* L. (concentration up to about 1% dry weight). Rankings for heavy metal accumulation are often proposed for the various environments, but the meaning of these rankings should be analyzed with respect to the following key question: "were the species showing the highest abundances favored by high water concentrations, or are they capable of concentrating high amounts in their tissues from low concentration in the surrounding environments?" (Muntau, 1981).

For Lake Maggiore, taking into account all variable factors, *Ceratophyllum demersum* L., *Myriophyllum spicatum* and *Littorella uniflora* (L.) Aschers. seem to be the best heavy metal indicators. According to their specific mechanisms of adsorption, translocation, utilization and excretion, the differing abilities of submersed macrophytes to accumulate metals are well documented in many papers (Cowgill, 1974; Hutchinson, 1975 and papers quoted therein; Gommès and Muntau 1981a). The water moss *Fontinalis* shows high accumulation capacity (Hutchinson, 1975) and the water chestnut, *Trapa natans*, is an accumulator plant for Mn and Fe (Gommès and Muntau, 1975b, 1981a).

It is well established that the concentration of trace metals in plants undergoes substantial changes according to the season. As a rule, heavy metal con-

centration in the plant population is highest in the cold season, decreasing rapidly during the period of maximal growth (Gommes and Muntau, 1981c).

Accumulation rates of different metals vary by the sampling period, the physiological status of macrophytes, their tissue age, and species. In laboratory conditions, *Elodea canadensis* Michx. accumulates about four times as much Hg as *Egeria densa* (Mortimer, 1985).

Any comparative study with the enormous amount of data now available must examine the data carefully. Results from different sources are not indicative of the 'mean' content of the trace metals in aquatic macrophytes (Bergman and Mueller-Hoberg, 1978).

Two other relevant questions have to be stated (Guilizzoni et al., 1989): (i) are there specific metal collector plant species present in the water bodies studied?; (ii) which of the present plant species behave as unspecific collector species? The answers are contradictory. In Lake Maggiore, results did not support the hypothesis of metal-specific species (Guilizzoni et al., 1989). On the other hand, in some Austrian water courses there was a pronounced element specificity (Fe, Mn, Zn, Cu, Ni and Cd) and species specificity (Ledl et al., 1981); among the 20 macrophyte species investigated, *Fontinalis antipyretica* L., *Elodea canadensis*, *Ranunculus aquatilis* L. and *Myriophyllum verticillatum* L. have particularly high enrichment factors between water and plant.

As a general rule, the degree of enrichment depends both on the kind of metal and its chemical form (Low et al., 1984). For instance, the higher atomic weight ions (e.g. Hg and Pb) were concentrated by *Elodea* much more effectively than lower atomic weight ions such as Cd and Ni (Mortimer, 1985). Mortimer also showed that an internal fractionation of metallic ions may occur within the plant. The proportion of methylmercury to total Hg was about 31% for the shoots and about 10% for the roots.

Other substances

Few data are available on the uptake, translocation and accumulation of pesticides, PCBs, hydrocarbons, and other common toxic organic substances in submersed aquatic plants (Bartell et al., 1983). A fair amount of information on aquatic herbicides is available because of their use as weed control.

Most herbicides are taken up within 1 h by roots and shoots (Forney and Davis, 1981; Jones et al., 1986). They are probably translocated in the phloem stream with a generally greater basipetal transport than acropetal (Welsh, 1977). Simazine, however, is known to be a 'xylem-mobile' herbicide and results reported by Thomas and Seamon (1968) suggest basipetal translocation of this compound. On the other hand, diquat and paraquat are not translocated in either direction, perhaps because of strong adsorption and abnormal physiological responses (Thomas and Seamon, 1968). These authors also indicate movements to the growing tips of *Potamogeton nodosus* Poir. (via

the phloem transport) of the 'phloem-mobile' herbicide endothal. There is no evidence of acropetal movement by the 'xylem-mobile' herbicides atrazine and diuron. The reduced movement within the plants of these substances is probably explained by the absence of a transpiration stream in submersed species (unlike emergent and floating-leaved ones).

Little or no movement has been demonstrated for the organochlorine insecticides (e.g. DDT and hexachlorocyclohexane, HCCH) in *Lemna minor* L., *Spirodela polyrhiza* (L.) Schleid., *Nymphaea alba* L. and *Potamogeton pectinatus* (Vrochinskiy, 1970; Vrochinskiy et al., 1970). An important feature of these substances is that the highest concentration (up to about 10 mg DDT l⁻¹) was found in species with the highest lipids content. Lipids are abundant in the stems and root of *N. alba* (Vrochinskiy et al., 1970).

Limited work has been done on PCBs in aquatic macrophytes. The only data relate to general environmental studies in which some macrophytes species are analyzed among the organisms occupying different trophic levels (Monod, 1983; McCrea and Wickware, 1986). As these halogenated aromatic hydrocarbons are hydrophobic and resistant to environmental degradation, aquatic plants accumulate them from the surrounding water and sediment (Gooch and Hamby, 1982).

EFFECT ON GROWTH AND PHOTOSYNTHESIS

Heavy metals

The effects of trace elements and other toxic substances in an aquatic ecosystem can be assessed by changes in the community structure, physiological activity, and ultrastructural components of macrophytes.

To date, ecotoxicologists have emphasized the effects of the toxicant at the species level (Stanley, 1974; Hutchinson and Czyrska, 1975; Labus et al., 1977; Filbin and Hough, 1979; Baudo et al., 1981b; Guilizzoni et al., 1984). Field studies that have taken into account the variations in plant communities or populations as a result of pollutants are very rare (Sortkjaer, 1984; Clarkson, 1985).

Although laboratory experiments are of great practical use in assessing the responses of macrophytes to chemical elements, the effect of a single pollutant on a single species will be of limited relevance for estimating the well-being of the ecosystem (Butler, 1984). Investigations have been of two types (Wetzel, 1976): (a) attempts to correlate fluctuations in plant growth with changes in water chemistry; (b) work with pure or axenic cultures of plants in relation to nutritional requirements, limitations to growth and photosynthesis. Both approaches provide an essential basis for interpreting the responses of species to environmental variables.

It appears that Cu has been the most studied element. This is because it is essential for plant growth and is also a highly effective herbicide.

The effect of Cu on the photosynthetic rate of *Potamogeton perfoliatus* L. was studied in a small industrially polluted lake in northern Italy (Baudo et al., 1981b). This study revealed an inhibitory effect on photosynthesis of 23% at tissue concentrations ranging from 5 to 10 ppm dry weight.

The effect of Cr on *Myriophyllum spicatum* has been studied in laboratory experiments using algae-free culture media (Guilizzoni et al., 1984). The maximum increase in shoot length occurred at a concentration of Cr in the nutrient media of $50 \mu\text{g l}^{-1}$ (shoot concentration of about 60 ppm dry weight). Higher concentrations of up to $1000 \mu\text{g l}^{-1}$ caused an almost linear reduction both in shoot weight and length. Photosynthetic rates exhibited reductions beginning at a concentration as low as $50 \mu\text{g l}^{-1}$. From 63 to about 465 ppm dry weight, photosynthetic rates were reduced by 6.4–80% (Fig. 2).

Apart from these two specific studies, only a few others have set up concentration limits for toxicity (Stanley, 1974; Hutchinson and Czyrska, 1975; Labus et al., 1977). Studies of this type are much more common with terrestrial and emergent marsh plants and with algae (Bowen, 1979; Lepp, 1981; Shkolnik, 1984).

The threshold of toxicity of some metal salts can be set at about 2 mg l^{-1} , according to Stanley (1974). He conducted an extensive study on the toxicity

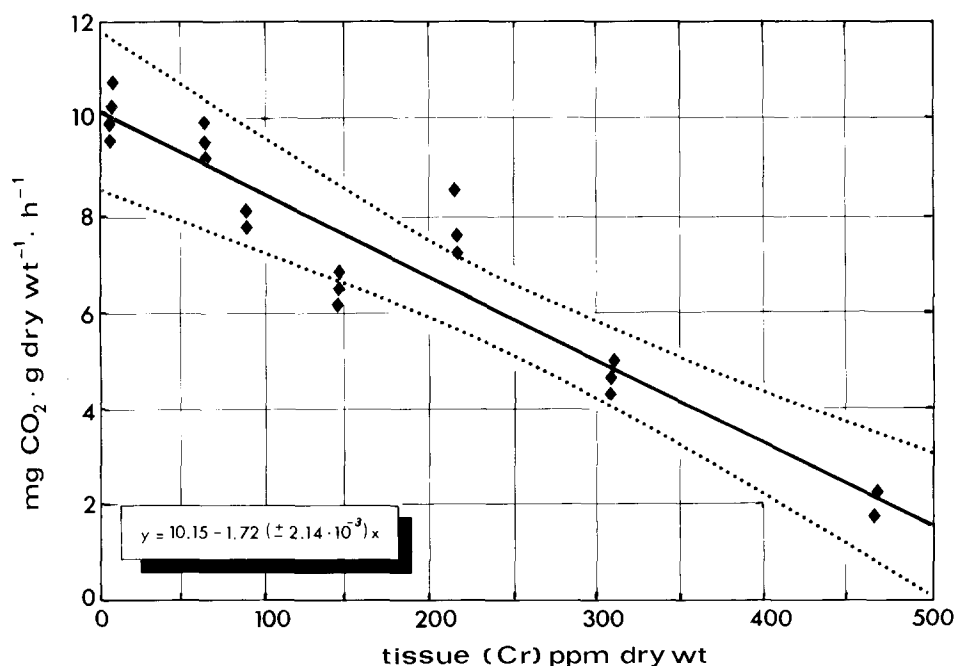


Fig. 2. Relationship between Cr accumulation in *Myriophyllum spicatum* tissues and its photosynthetic rates. Confidence limits ($P < 0.05$) are also indicated (Guilizzoni et al., 1984).

of various heavy metals and salts to *Myriophyllum spicatum*. He found that a 50% inhibition of root weight occurred at concentrations of 0.25 ppm Cu^{2+} , 1.9 ppm $\text{Cr}_2\text{O}_7^{2-}$, 3.4 ppm Hg^{2+} , 2.9 ppm AsO_4^{3-} , 7.4 ppm Cd^{2+} , 2.5 ppm Al^{3+} , 9.9 ppm Cr^{3+} , 21.6 ppm Zn^{2+} , 363 ppm Pb^{2+} . Moreover, it was found that the roots of *M. spicatum* were more sensitive to toxicants than were the shoots, and that weight increase was inhibited more than length growth. This final finding was confirmed by Guilizzoni et al. (1984).

The effect of Cu and Cd on the rates of photosynthesis and respiration of the aquatic moss *Fontinalis antipyretica* was studied by von Sommer and Winkler (1982). It was reported that an aqueous solution of copper and cadmium nitrate reduced the net photosynthesis of the moss. At low concentrations (0.64 mg l^{-1}), Cu caused a greater reduction than Cd, but at high concentrations (635 mg l^{-1}), the reverse was true. Interestingly, the damage to *Fontinalis* after addition of copper nitrate occurred during the first 15 min, after which there was a decided recovery. The rate of dark respiration increases after incubation with Cu and decreases with Cd. When Ca was added to the Cu and Cd solutions, their adverse effects on photosynthesis were reduced.

The latter result is an important one. In any ecotoxicological work, differential responses of natural stands of macrophytes to heavy metal toxicity are to be expected. Different physical and chemical environmental variables, as well as tolerance phenomena to toxic levels (Antonovics et al., 1971; Taylor and Crowder, 1984) might produce differential responses in plant metabolism.

In a study of three submersed species (*Potamogeton pectinatus*, *Vallisneria spiralis* and *Hydrilla verticillata* (L.f.) Royle), Hg, Pb, Cd and Cu hastened senescence of the macrophytes, and Cu was the most effective metal in inducing deleterious effects in *Potamogeton* and *Vallisneria* (Jana and Choudhuri, 1982). Of the three species, *Hydrilla verticillata*, known to be resistant, was the most tolerant to the heavy metals applied.

The toxic effect of Cu ($5 \mu\text{mol l}^{-1}$) was also observed on *Elodea nuttallii* (Planch.) St. John (van der Werff and Pruyt, 1982), but no differentiation in growth or mortality was detected for Zn, Pb and Cd in *Callitriche platycarpa* Kütz., *Spirodela polyrhiza* and *Lemna gibba* L.

When considering the toxicity of heavy metals, a distinction should be made between elements essential to plants and metals which have no proven beneficial biochemical effect. In some cases, increased levels of toxic metals (e.g. Cr) may actually stimulate growth without being essential for any metabolic process (Gardner, 1980; Guilizzoni et al., 1984).

Other toxic substances

Several recent studies have examined the effect of atrazine on various macrophytes (see references quoted in Jones and Winchel, 1984 and Jones et al.,

1986; Delistraty and Hershner, 1984). This substance caused photosynthetic inhibition of 1% (at $20 \mu\text{g l}^{-1}$) and 50% (at $95 \mu\text{g l}^{-1}$) in *Potamogeton perfoliatus*, *Ruppia maritima* L., *Myriophyllum spicatum* and *Zannichellia palustris* L. (Jones and Winchel, 1984).

Experiments conducted with *Potamogeton perfoliatus* and *Elodea canadensis* demonstrated that inhibition of photosynthesis with increasing atrazine concentration follows Michaelis–Menten kinetics (Forney and Davis, 1981; Jones et al., 1986). The maximum effect observed (about 90% inhibition) occurs at concentrations of about $600 \mu\text{g l}^{-1}$.

In general, a 50% inhibition of photosynthesis and growth probably occurs at atrazine concentrations of between 50 and $100 \mu\text{g l}^{-1}$ in many macrophytes (Jones et al., 1986). Fully grown plants are probably more sensitive to atrazine than dormant plants.

Among the pesticides, organochlorines have the greatest inhibitory effect on photosynthesis and respiration of macrophytes (Ramachandran et al., 1984). A study conducted on the effect of six commonly used pesticides on some marine macrophytes demonstrated that DDT at a concentration of $50 \mu\text{g l}^{-1}$ negatively affects both net photosynthesis and dark respiration. This compound together with lindane exerts a more toxic effect on plants than other pesticides such as endosulphan, methyl paration, dimethoate and sevin.

Potamogeton crispus, *Potamogeton lucens* L., *Elodea canadensis*, *Groenlandia densa* (L.) Fourr., *Ranunculus penicillatus* (Dum.) Bab. and *Myriophyllum alterniflorum* DC exposed to 10 mg l^{-1} of phenol, showed reduced photosynthetic activity after 28 days (Labus et al., 1977). On the other hand, much lower concentrations ($< 0.5 \text{ mg l}^{-1}$) of the anionic detergent Marlon A (alkalbenzosulphonate) are required to significantly reduce the photosynthetic values of the above macrophytes (Labus et al., 1977).

CONCLUSIONS

The interactions between aquatic plants and some toxic substances such as heavy metals are amply described in many scientific articles. Unfortunately, the complexity of these studies, and in many cases the vagueness of their objectives, particularly those carried out in the field, have made it difficult to obtain unambiguous results or to describe general trends. Many papers have ranked macrophytes according to their affinity with heavy metals, but the meaning of these rankings is not always clear. Answers to the basic question of whether a high heavy metal content in plants is the result of high external metal levels or high efficiency uptake from environments that are actually low in concentrations, are not forthcoming (Muntau, 1981). In other words, the high concentrations of certain metals found in macrophytes may not reflect high pollution levels, but rather the efficiency of a plant at concentrating sub-

stances (Wells et al., 1980). This could cause problems in using some species as bio-indicators.

I have cited how several environmental factors are important in the macrophyte-toxic compound cycle occurring in aquatic systems and how contradictory results often emerge. For many years there has been a debate concerning root and shoot uptake of elements by submersed rooted macrophytes. Both are important assimilatory organs, but some very important questions have not received enough attention (Denny, 1980). One of these questions, which is of particular relevance, is: what conditions govern root or shoot absorption, and by what mechanism(s) does translocation occur?

Laboratory studies have shown that competition between metals for absorption sites leads to antagonistic and synergistic effects (Low et al., 1984). Thus, both in ecotoxicology studies and field work this aspect should be considered; in one case, to set up realistic water quality criteria, and in the other, to compare data from different environments.

Comparison of metal content in macrophytes is often difficult because of differences in sampling time (aging of plants), tissues analyzed (growing tips, whole plant), presence of pollution sources, etc. Hence, metal data cannot be extrapolated from one species to another or even within the same species. Because of the presence of metal-binding proteins, Henning (1984) suggests that "future metal pollution studies spend less time on baseline surveys and unrealistic accumulation experiments, but rather concentrate on bioassay of these metal complexes".

There could be a difference of two orders of magnitude between the effect observed on the enzyme system and the visible effect on leaf morphology when plants are exposed to the same toxic substance (Sortkjaer, 1984). Therefore, I suggest planning laboratory experiments with approximately the actual field level of the pollutants investigated and efforts directed at finding out the response to photosynthesis and respiration of a target species.

Two more problems connected with methodology should be considered carefully. The first is to define the role of 'Aufwuchs'. They are agents in the rapid uptake of trace elements, and their presence reduces the concentration in the plant (Patrick and Loutit, 1977). Epiphytes are often abundant on stems and leaves of submersed macrophytes and are difficult to wash away completely in the cleaning process. Consequently, it is not clear whether metals are absorbed by the plant or linked to microflora and fauna attached to the plant surface.

The second problem, the variability in the concentrations reported, could be due to different or poor laboratory techniques such as: (i) different digestion procedures (wet and dry ashing); (ii) matrix interferences (e.g. Ca, Na, K, SO_4^{2-} , Cl^-); (iii) the use of different standard solution (e.g. acid strength); (iv) different analytical equipment; (v) contamination of the plant material

by the use of unsuitable mills in the grinding operation (Baudo et al., 1979; Brix et al., 1983).

Workers studying heavy metals in biological material should, as far as possible, adopt standardized procedures and methods which are continually checked against certified reference materials.

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