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## Assessment of nutrient accumulation capability of two economically important aquatic plants

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#### Abstract

The concentrations of eight plant nutrient elements (N, P, K, Fe, Mn, Cu, and Zn) were measured in the leaves and roots of the emergent macrophytes, *Euryale ferox* (Salisb.), and *Trapa bispinosa* (Roxb.) taken from experimental farm ponds of ICAR-RCER, Research Centre for Makhana, Darbhanga. *E. ferox* leaves showed higher concentrations of C, N, P, and Zn than roots while the roots exhibited significantly higher concentrations of K, Fe, Mn and Cu. *T. bispinosa* roots also contained higher concentrations of Fe, Mn and Cu than its leaves, but the differences were significant only for P. Fe was the only nutrient that showed the highest concentrations in roots of both the plants. *T. bispinosa* roots had higher values of N, P, K, Cu and Zn than *E. ferox* roots. By contrast, C, Fe and Mn concentrations were higher in *E. ferox* leaves than in *T. bispinosa* leaves. *E. ferox* and *T. bispinosa* showed high capability to accumulate Fe and Mn in their leaves as well as in roots, appeared a good biomagnification plants.

Keywords: Euryale ferox Salisb., Trapa bispinosa Roxb., nutrient uptake, vegetative tissues, bio magnification

#### Introduction

Macronutrients, such as N, P and K, have more often been the focus of nutritional studies because of their potential role in limiting plant growth. As water column concentrations of P are often considered to be potentially limiting through most of the growing season in temperate zone lakes, the sediments represent a large potential reserve of P which may supply nutritional demands of rooted macrophyte growth.

Early attempts to identify the relative importance of roots and shoots in element uptake were conducted under experimental conditions (Bristow and Whitcombe, 1971)<sup>[4]</sup>. These studies showed that macrophytes could take up elements (primarily P) through roots and translocate them to the shoots. In situ studies demonstrated more convincingly that rooted submerged macrophytes obtained virtually all their N and P from the sediments (Barko and Smart, 1981)<sup>[2]</sup> compromised the general ability of macrophytes to function as monitors of water column pollution. Potassium was also sediment derived, although the sediment contribution to the plants was lower and more variable than that of N and P. The majority of rooted freshwater macrophyte nutrition is generally believed to be sediment derived.

In aquatic ecosystem, the availability of micronutrients in the plant tissues depends on chemical composition and type of soil and irrigation water. Moreover, their concentration also depends on capability of accumulating nutrients from soil and water (Dutta *et al.*, 1986a) <sup>[8]</sup>. Aquatic plants can take up large quantities of nutrients (Singh, 2014) <sup>[23]</sup> and metals (Jha and Dutta, 2003 and Singh *et al.*, 2014) <sup>[13, 25]</sup> from the environment, releasing them when they decay. Aquatic macrophytes differ both in their capacity to take up metals in root tissues and in the proportion of metals transferred to above ground parts (Pip and Stepaniuk, 1992) <sup>[18]</sup>. Nutrient concentrations have been studied in some aquatic plant species, but data on their concentrations in the different parts of the plants are scanty, especially in the commercially important aquatic plants.

In this study, we tried to investigate the concentrations of C, N, P, K, Fe, Mn, Cu and Zn in the leaves/shoots and roots of *E. ferox* and *T. bispinosa* collected from research farm ponds. Our aim was to define which species and which plant organs exhibit the greatest accumulation and to evaluate whether these species could be usefully employed in biomonitoring studies. The use of these species for phytoremediation has also been considered.

#### **Materials and Methods**

Two native aquatics rooted emergent vascular macrophytes plant species were selected from four different ponds situated in close proximity on the same soil with the same source of water and similar age at experimental farm of ICAR-Research Complex for Eastern Region, Research Centre for Makhana, Darbhanga for the study so as to serve as a suitable replicated trial. The level of water depth ranging from 1.20 m-1.50 m in the ponds was maintained throughout the growth period of Euryale ferox Salisb and Trapa bispinosa. Both the species are the most dominant economical aquatic plants of the subtropical region of North Bihar (India). E. ferox has gigantic circular leaves of diameter ranging from 0.50 m-1.50 m. In contrast to this Trapa bispinosa has numerous small leaves size ranging from 5.0 cm-6.0 cm. Both the plant species are capable of taking-up the elements from the soil sediments as well as the water medium through their aerenchymatous tissues present in the roots, stems and leaves part of the plant.

Samples of both the plants were collected four times throughout the year (From June 2017 to August 2019) every

year from four different ponds. Monthly biomass yields of the crop were recorded by the random-quadrant sampling method as suggested by Odum and Heald (1972) <sup>[17]</sup>. Samples were analysed for macro and micronutrients. Total nitrogen in the plant was determined by micro-Kjeldahl method as per AOAC (1975) <sup>[1]</sup>. The phosphorus and potassium were estimated from tri-acid mixture (9:4:1 HNO<sub>3</sub>:H<sub>2</sub>SO<sub>4</sub>:HClO<sub>4</sub>) digested plant samples as outlined by Jackson (1973) <sup>[10]</sup>. Fe, Mn, Cu and Zn were determined using atomic absorption spectrophotometer (Lindsay and Norvel 1978) <sup>[15]</sup>. All nutrient concentrations in plant and soil samples were expressed on a dry weight basis. The Paired Student t-test on the normalized data set by logarithmic transformations were used to evaluate the significance of the differences between roots and green parts for each species. The ranges of total and available element concentrations (ug g-1 d.w.) in the sediments of ponds, collected from four ponds during May 2017. The ranges of surface water concentrations (ug  $l^{-1}$ ) measured six times (from July 2017 to August, 2019) in 4 sites of pond are also reported in the table 1.

Table 1: Ranges of total and available element concentrations (µg g-1 d.w.) in the sediments of ponds

Elements	Sediment total concentrations (ug g <sup>-1</sup> d.w.)	Sediment available concentrations (ug g <sup>-1</sup> d.w.)	Water concentrations (ug l <sup>-1</sup> )
Ν	3000-4000	90.5-251.5	2205-2500
Р	350-870	8.2-12.4	5000-7000
K	4200-5000	180-200	0.0005-0.001
Fe	28000-35000	20.1-40.5	10-1400
Mn	400-700	6.51-13.33	0.02-130
Cu	2.21-23.0	0.75-2.25	0.2-30
Zn	10-40	0.12-0.56	0.2-100

#### **Results and Discussion**

#### E. ferox

The comparison between leaf and root concentrations of E. ferox (Figs. 1-3 & 8) showed that C, N, P, and Zn are 40%, 0.36%, 0.48%, 0.04%, respectively higher in leaves than in roots. The differences were significant (Table 2) for N (p < 0.01). By contrast, the concentrations of K and of the trace elements measured (Fe, Mn and Cu) were higher in roots than in leaves (Figs. 4-7) and the differences were statistically significant (p < 0.01) for C and Fe. The low content of K in leaf tissues might be attributed to the fact that root oxygen deficiency decreases the selectivity of K<sup>+</sup>/Na<sup>+</sup> uptake by roots in favour of Na<sup>+</sup> and retards the transport of K<sup>+</sup> to shoots (Thompson *et al.* 1989) <sup>[26]</sup>. This also corroborated the findings of Jat *et al* (2014) <sup>[12]</sup>. C, N and P concentrations were found to be higher in the leaves than in roots of E. ferox and T. bispinosa. On the other hand, for N and P several authors had measured concentrations in the leaves of aquatic macrophytes, but not in the roots. In particular, for nitrogen (Jha and Dutta, 2003)<sup>[13]</sup> they recorded

concentrations ranging from 0.192 to 0.196% and 0.170 to 0.172% in leaves and roots, respectively of *E. ferox*. There were no such reports regarding concentrations of N in roots of *T. bispinosa*. In the case of P, Jha and Dutta (2003) <sup>[13]</sup> had found concentrations ranging from 0.24 to 0.28% and 0.16 to 0.18% in leaves and roots, respectively of *E. ferox*. N and P concentrations reported by the cited authors was comparable to those measured in *E ferox* leaves collected from old traditional ponds. The higher concentration of P in tissues of T. *bispinosa* as compared to tissues of *E. ferox* was might be attributed to the fact that plant species differ in their requirement for P, ability to extract P from the soil on account of numbers of fine and prolific root hair formation, and

response to insoluble forms of P (Mandal *et al.* 2009 and Guilizzoni, 1991) <sup>[16, 9]</sup>. Further, P has long been known to have an impact on the eutrophication of lakes and ponds those that are oligotrophic (i.e., those lakes where the concentration of P and/or other nutrients is very low and thus the growth of algae and other aquatic plants is limited). Pertaining to K, the roots of *E. ferox* showed higher concentrations than leaves while contrary to this the green organs i.e., leaves contained higher K concentrations than the roots of *T. bispinosa* which resulted due to luxury absorption of potassium by the root tissues of any plants. Thus, plant root system plays an important role in nutrient recycling (Singh *et al.* 2012) <sup>[22]</sup> and (Jat *et al.* 2014) <sup>[12]</sup>.

Our data concerning the higher concentrations of trace metals in the roots of *E. ferox* than in leaves, agreed with the findings of Dutta *et al.* (1986a and 1986b)<sup>[8, 7]</sup> who found values equal to 1947 and 2236 mg/kg for Fe, 840 and 1120 mg/kg for Mn and 8.3 and 8.5 mg/kg for Cu, respectively in the leaves and roots of E. ferox taken from the natural ponds in northern Bihar, India. Similarly, in rooted macrophyte, T. bispinosa, grown in field condition showed higher concentrations of Fe and Cu in roots than in leaves, and the concentration ratios between roots/leaves for Fe and Cu were of the almost same order of magnitude as in E. ferox. Cu concentrations were equal in the roots and in the leaves of both E. ferox and T. bispinosa, however, its concentration in T. bispinosa was 2fold higher than those found in E. ferox roots. This may be attributed to the factors like root release organic caids (Ryan et al. 2001)<sup>[19]</sup> and change in rhizosphere redox potential for micronutrient acquisition (Shiferaw et al.1992) <sup>[21]</sup> also play the important role in accumulation of nutrients to plants. Cu and Mn concentrations were almost equal in the leaves and roots of *E. ferox*.



Fig 1: Carbon © content in leaves and roots of E. ferox and T. bispinosa



Fig 2: Nitrogen (N) content in leaves and roots of E. ferox and T. bispinosa



Fig 3: Phosphorus (P) content in leaves and roots of *E. ferox* and *T. bispinosa*  $\sim$  2500  $\sim$ 

Micronutrient concentrations in aquatic plants varied considerably according to the part of the plant as well as to the chemical characteristics of the elements (Singh, 2017)<sup>[24]</sup>. There was usually very less mobility of Mn from the roots to the leaves (Kumar *et al*, 2017)<sup>[14]</sup>; the reason for the low mobility of this element from the roots to the green parts might be due to barriers or to immobility in plant system (Shewry and Peterson, 1974)<sup>[20]</sup>. However, in this study it was found that there was no regular pattern pertaining to absorption of micronutrients in the roots of both the aquatic plants, especially in case of Cu. Mn registered higher concentrations in roots than in green organs in *T. bispinosa*. However, in case of *E. ferox*, this fact does not hold true.

The findings showed the highest value of Fe and Mn in the roots of *E. ferox*, the emergent macrophyte. Similarly, the contents of Fe and Mn were found to be higher in the roots than in the shoots of *T. bispinosa*. However, Mn concentrations were even two orders of magnitude higher than shoots in *T. bispinosa*. Roots of both *E. ferox* and *T. bispinosa*.

showed high accumulation of trace elements, suggesting considerable availability in the sediments (Singh, 2014) <sup>[23]</sup> and marked root uptake for the emergent plant. The roots of *T. bispinosa* was observed to contain almost two-fold higher concentration of Mn than their leaf tissues which might be attributed not only to the fact of species difference but another fact is that Mn is the only micronutrient whose content varies in different tissues of the plant than any micronutrient. The capability of *E. ferox* roots and *T. bispinosa* shoots and roots to accumulate heavy metals, depending on the element concentrations in the sediments and/or water, might allow their use in heavy metals biomonitoring in pond/lake ecosystem.

In particular, the root/leaf ratio was ranged from 1.66 for K (Table 4). For K, Fe, Mn and Cu the root concentrations were slightly higher than the leaf concentrations (Table 4). The concentrations of Fe in root were found to be significantly higher than leaves of *E. ferox* species.

**Table 2:** Significance of differences (P) between the roots and leaves of E. ferox according to the paired Student – t – test (t). The degree of freedom (d.f.) is also reported in the table; n.s.: not significant.

Elements	t	р	d.f.
С	-7.427	< 0.001	8
Ν	-1.69	n.s.	8
Р	-12.24	< 0.001	8
K	63.99	< 0.001	8
Fe	37.32	< 0.001	8
Mn	2.33	n.s.	8
Cu	4.79	< 0.01	8
Zn	-5.07	< 0.001	8

#### Trapa bispinosa

Differences in element concentrations between the leaves and roots of *T. bispinosa* are almost similar to *E. ferox* species. Cu concentration is similar in the leaves and in roots. P and K

have higher concentrations in roots than in leaves (Figs. 3-4). The concentration ratio between the values of the elements in roots and in leaves of *T. bispinosa* ranged 0.57 for Mn.



Fig 4: Potassium (K) content in leaves and roots of E. ferox and T. bispinosa

The different trace metal concentrations in the organs of the two species analyzed might be reflected different pathways for the elements' uptake. Root uptake with subsequent translocations to above ground tissues in the *T. bispinosa* was reported as the main rout (Jackson, 1998) <sup>[11]</sup>. Guilizzoni

(1991) <sup>[9]</sup>, on the other hand, had stated that some rooted submerged plants might absorb metals directly from water when they were not readily available in sediments and/or in high concentrations in the surroundings.

**Table 3.** Significance of differences (P) between the roots andshoots/leaves of *T. bispinosa* according to the paired Student -t - test (t). The degree of freedom are also reported (d.f.) in the table;n.s.: not significant.

Elements	t	р	d.f
С	-6.07	< 0.001	8
Ν	-9.40	< 0.001	8
Р	-4.49	< 0.01	8
K	-6.83	< 0.001	8
Fe	1.85	n.s.	8
Mn	75.15	< 0.001	8
Cu	0.80	n.s.	8
Zn	-4.32	< 0.01	8

#### Comparison between E. ferox and T. bispinosa

The leaves of *T. bipinosa* species (Figs. 1, 2, 5 & 8) exhibited significantly higher values of C, N, Fe and Zn than the roots. By contrast the roots of *E. ferox* species (Figs. 1, 4, 5, 6 & 7) exhibited higher concentrations of C, K, Fe, Mn and Cu than leaves. *E. ferox* roots showed higher values of organic carbon,

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Fe and Mn than T. bispinosa roots (Figs. 5 & 6). The concentration ratios of these elements between E. ferox and T. bispinosa roots ranged from 1.09 for Fe to 1.60 for Mn (Table 4). The other elements exhibited low differences between the roots of the two species. Unlike the roots, the green parts showed higher concentrations of N, P, K, Cu and Zn in T. bispinosa leaves than in E. ferox leaves. The ratios between the concentrations of the leaves of T. bispinosa and E. ferox ranged from 1.11 (for N) to 3.06 (for K) (Figs. 2 and 4; Table 5). Both the plant species showed differential uptake pattern of nutrients in their leaves and shoot parts. E. ferox was observed to have higher concentrations of C, N and P in their leaves compared to its root parts. However, C, N, P, K and Zn were found to be contained in appreciably higher amounts in the tissues of leaves as compared to root tissues of T. bispinosa. While root parts of E. ferox were noted to be more efficient in uptake of Fe and Mn than the roots of T. bispinosa. Uptake trend of nutrients in both the species revealed that T. bispinosa were more efficient in absorbing the nutrients particularly N, P, K, Cu and Zn than E. ferox.



Fig 5: Iron (Fe) content in leaves and roots of E. ferox and T. bispinosa



Fig 6: Manganese (Mn) content in leaves and roots of *E. ferox* and *T. bispinosa*  $\sim _{2502} \sim$ 



Fig 7: Copper (Cu) content in leaves and roots of E. ferox and T. bispinosa



Fig 8: Zinc (Zn) content in leaves and roots of E. ferox and T. bispinosa

The extent of mobility of Mn, Cu and Zn in plants varied between species, from mobile in some plants (Bosserman, 1981)<sup>[3]</sup> to immobile in others (Guilizzoni, 1991)<sup>[9]</sup>. In case of absorptions of Cu, their distribution in roots and shoots of both the plants was found equal. This type of uptake pattern confirmed the direct absorption from the water into the shoots or confirmed the contribution of water pertaining to translocation of nutrients to the shoot parts of the plants.

Zn was found to be more mobile from roots to leaves than other micronutrients such as Fe and Mn (Kumar *et al.* 2017) <sup>[14]</sup>. Both in *E. ferox* and in *T. bispinosa* Zn appeared more mobile than Fe and Mn, since its concentrations ratios in roots/leaves 0.86 and 0.63 that was lower than those for Fe (1.12 and 1.05) and Mn (1.00 and 1.75). Zn concentrations were higher in shoots than in roots in both the emergent macrophyte. This kind of translocation of Zn in aquatic macrophytes was generally found to be true in case of shallow

wetlands (Collins *et al.*, 2005) <sup>[6]</sup>. Campbell *et al.* (1988) <sup>[5]</sup> reiterated that metals that preferentially bind to organic ligands, like Cu and Zn, should be less available to rooted macrophytes when such ligands are available for binding.

Table 4: Nutrient content in leaf and root tissues of E. ferox

Elements	Leaf	Root	Ratio of Root/Leaves	Ratio of roots of <i>E. ferox</i> to roots of <i>T. bispinosa</i>
C%	40	30	0.75	1.20
N%	0.36	0.26	0.72	0.93
P%	0.48	0.38	0.79	0.54
K%	0.30	0.50	1.66	0.62
Fe (mg/kg)	2060	2310	1.12	1.09
Mn (mg/kg)	942	945	1.00	1.60
Cu (mg/kg)	6	7	1.16	0.42
Zn (mg/kg)	106	92	0.86	0.48

**Table 5:** Nutrient content in leaf and root tissues of *T. bispinosa*

Elements	Leaf	Root	Ratio of Root/Leaves	Ratio of leaves of T. bispinosa to leaves of E. ferox
C%	35	25	0.71	0.87
N%	0.40	0.28	0.70	1.11
P%	0.80	0.70	0.87	1.66
K%	0.92	0.80	0.87	3.06
Fe (mg/kg)	2013	2113	1.05	0.97
Mn (mg/kg)	337	591	1.75	0.35
Cu (mg/kg)	16.00	16.60	1.04	2.66
Zn (mg/kg)	300	191	0.63	2.83

It was difficult to establish whether, and in what proportion an element was taken up from sediments by the roots; it might be dependent on the chemical behavior of elements and also on sediment geochemistry (Jackson, 1998)<sup>[11]</sup>. Moreover, it was also difficult to establish the amount of an element that transferred from the roots to the shoots as different plant species and their different parts exhibited varied concentration of micronutrients.

On the basis of this study, it may be concluded that both *E. ferox* and *T. bispinosa* have shown differential uptake pattern of nutrients in their leaves and shoot parts. *E. ferox* had shown higher concentrations of C, N and P in their leaves compared to its root parts. However, C, N, P, K and Zn were found to be contained in appreciably higher amounts in the tissues of leaves as compared to root tissues of *T. bispinosa*. While root parts of *E. ferox* were noted to be more efficient in uptake of Fe and Mn than the roots of *T. bispinosa*. Uptake trend of nutrients in both the species revealed that *T. bispinosa* were more efficient in absorbing the nutrients particularly N, P, K, Cu and Zn than *E. ferox*.

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