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Short communication

Preliminary Studies on Trace Metal Accumulation by Aquatic Weed *Salvinia molesta* and its Implication in Phytoremediation

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ABSTRACT
Water, sediment sample and specimens of free-floating aquatic macrophyte, *Salvinia molesta* were collected from a Lake situated in North Goa to assess trace metal (Mn, Ni, Zn and Pb) accumulation by selected aquatic weed. The concentration of trace metals from water, sediment and *S. molesta* was determined using atomic absorption spectroscopy. Studies revealed that concentration of metals in water was less as compared to sediments. Brown fronds of *Salvinia* accumulated more concentration of trace metal as compared to green shoots. Metal accumulation in brown fronds in pre monsoon was as (Mn 3.32.ppm; Ni-0.04 ppm; Zn - 3.62ppm; Pb-BDL) while in green shoots (Mn 1.20ppm; Ni-BDL; Zn - 1.02ppm; Pb-BDL); in monsoon season the metal concentration in brown frond was (Mn 3.04.ppm; Ni-0.07 ppm; Zn – 1.93ppm; Pb-BDL) and in green shoot (Mn 0.62ppm; Ni-BDL; Zn - 0.30 ppm; Pb-BDL); whereas in post monsoon metal concentration in brown frond was recorded as (Mn 4.75ppm; Ni-0.55 ppm; Zn - 0.25ppm; Pb-0.39ppm) and in green shoot- (Mn 1.10.ppm; Ni-0.12 ppm; Zn - 0.10ppm ; Pb-0.10ppm). Bioaccumulation and Translocation factors for metal accumulation and translocation were above 1. Study revealed that *S. molesta* is hyper accumulator of above trace metals therefore suitable for phytoremediation.

Key words: *Bioaccumulation factor; Translocation factor; hyperaccumulator*

INTRODUCTION

Trace metal concentration in freshwater bodies is a growing concern. Mining activities, burning of fossil fuels, discharge of agricultural, industrial and domestic waste, and pesticides containing compounds of trace metals are responsible for trace metal contamination of aquatic environment (Hutchinson et al. 1993). Metals after entering the water, precipitate and get adsorbed on solid surfaces, remain suspended or are accumulated by aquatic organisms (Eralagere and Bhadravathi 2008). Degradation of water bodies due to trace metal pollution depends upon the degree of exposure and size of the water body to anthropogenic activities (Tu et al. 2004). Aquatic macrophytes act as biological filters as they absorb and accumulate elements from water and sediments, hence can be used as a biomonitoring tool of polluted waters (Zurayak et al. 2001). It is important to identify the hyperaccumulators that can be effectively used in phytoremediation. The success of trace metal accumulation by aquatic plants depends on proper selection of plant species that have high growth rate in the contaminated environment, large surface area of the portion in contact with water, and high translocation potential (Hadad et al. 2011). The bioaccumulation factor (BAF) and translocation factor (TF) are most important plant features in phytoremediation with respect to uptake of metals, their mobilization into plant tissues, and storage in the aerial plant biomass (McGrath and Zhao 2003). Present study aimed to know whether the selected Lake was contaminated with trace metals and to understand the metal accumulation and translocation ability of *Salvinia molesta* Mitchell.

MATERIALS AND METHODS

Present investigation has been carried by selecting a Lake from North Goa district located on the banks of Cumbarjua canal situated between 15.5° N Latitude 73.94° E Longitude. The study was carried out in three different season's viz., pre-monsoon, monsoon and post-monsoon season. The trace metals viz., Mn, Ni, Zn and Pb were analysed from water, sediment samples and aquatic plant using standard protocols.

Sample collection, preparation and analysis

Water sample was collected in 1 lit. sterile plastic can; acidified by adding few drops of concentrated HNO_3 which was later on filtered in lab using 0.45 microns pore (Millipore) filter. In a 1000 ml separating funnel, around 500 ml of water sample was taken and pH was adjusted to 4 - 5 with dilute NH3. Trace metals from water were extracted using APDC (Ammonium Pyrorolidene dithio Carbamate) and MIBK (Methyl Isobutyl Ketone) (APHA 2012). To this sample 10 ml of APDC and 15 ml MIBK was added. The mixture was shaken for two minutes. The two phases were allowed to separate after 15 - 30 minutes. Upper organic layer was drained into 100ml separating funnel (ensuring that MIBK extract was free from water sample). The procedure was continued by adding 5 ml APDC and 10 ml MIBK to water sample and the contents were transferred in a separating funnel. The contents were shaken again for 2 minutes and both the extracts were combined. Core of sediment was collected in triplicate from same sampling site using plastic corer from upper 10 cm layer of the bottom sediments. Each core was packed separately in acid soaked zip-lock plastic bag and stored in ice box until transported to laboratory. Sediment samples were oven-dried at 105 ºC and sieved through a 2 mm mesh screen to remove coarse materials like small stones, wood, and detrital materials. Other visible impurities were removed prior to grinding and the sample was made into a homogenous mixture using mortar and pestle. Digestion of sediment for total metal analysis was carried out as per the following protocol (APHA 2012; Mudroch et al. 1997). Finely ground sediment sample (0.2 g) was transferred into clean chromic acid washed teflon beaker. To this, a mixture of 10 ml Hydrofluoric acid, Nitric acid and Perchloric acid in the proportion of (7:3:1) was added slowly to avoid excessive frothing and was completely dried on the hot plate at 150ºC. After drying, again 5ml of the above mixture was added and dried on the hot plate for 1h and then 2ml conc. HCl was added and dried completely. Final residue was dissolved in the 10 ml of $HClO_4$ and HNO3. (1:1). After ensuring complete digestion (clear solution) of sediment sample, the content from teflon beakers was transferred into the acid washed polypropylene volumetric flask and the solution was made up to 50 ml with milique water.

Salvinia molesta, was handpicked from the habitat and washed with lake water carefully. Brown fronds and shoots were separated, washed in distilled water and dried at 70ºC in hot air oven for 48 hours. Dried samples were homogenized and ground to yield fine powder. Nitric acid digestion method was followed for extraction of trace metals from plants (Zheljazkov and Nielson 1996). One gram of powdered sample was taken to which 10 ml of concentrated HNO_3 was added. The sample was heated for 45 minutes at 90ºC, and then the temperature was increased to 150ºC at which the sample was boiled for at least 8 hours until a clear solution was obtained. Concentrated HNO_3 (5 ml) was added thrice to the sample. Digestion was carried out until the volume was reduced to 1 ml. After cooling, 5 ml of 1% HNO_3 was added to the sample. The solution was filtered using Whatman No. 42 filter paper. It was then transferred to a 25 ml volumetric flask by adding milique water. The digested sample solutions were aspirated for trace metals using Atomic Absorption Spectrophotometer after ensuring the technicalities as per standard procedures (APHA 2012). Average values of three replicates were taken for all detections.

Bioaccumulation factor (BAF) and Translocation factor (TF) were calculated as follows:

BAF = Metal concentration in plant tissue / Metal concentration in water (Klavins et al. 1998).

TF = Metal concentration in root / Metal concentration in shoot (Wu and Sun 1998).

RESULTS AND DISCUSSION

Concentration of trace metals in water, sediments and *Salvinia molesta* of Syngenta Lake are presented in Table 1. The variations in metal concentration of water were compared with standard values of (WHO 2008) for drinking water and in sediments were compared with provisional sediment quality guidelines for metals MoE 1993 Ontario (Table 2). Results of Bioaccumulation factor and Translocation factor are depicted in Table 3.

It was observed that the concentration of trace metals in sediment was higher than water. Manganese is an essential trace element in physiological

Table 1. Trace metal concentration (ppm) in Syngenta Lake and phytoaccumulation by *S. molesta*

Metal	**Pre-monsoon**				**Monsoon**				**Post-monsoon**			
	Water	**Sediment**	**SBF**	**SS**	**Water**	**Sediment**	**SBF**	**SS**	**Water**	**sediment**	**SBF**	**SS**
Fe	4.60	8.10	4.49	0.78	0.38	7.75	1.93	0.30	0.80	9.56	4.29	1.42
Mn	BDL	4.40	3.22	1.20	0.42	10.07	3.04	0.62	0.01	7.76	4.75	1.10
Cu	BDL	0.31	0.10	0.06	0.40	5.50	0.16	0.10	0.04	7.36	1.70	0.20
Ni	0.019	0.45	0.04	BDL	0.30	1.75	0.07	BDL	BDL	2.49	0.55	0.12
Zn	0.45	8.47	3.62	1.02	4.45	7.00	1.93	0.30	1.27	0.95	0.25	0.10
Pb	0.16	2.71	BDL	BDL	BDL	2.50	BDL	BDL	BDL	3.32	0.39	0.10

BDL = below detectable level; Fe - Iron, Mn - Manganese, Cu - Copper, Ni - Nickel, Zn - Zinc, Pb - Lead; SBF - *Salvinia* (BrownFrond), SS - *Salvinia* (Shoot). All values are mean of three readings.

Table 2. Maximum permissible limit of metals in water (WHO 2008) and Provisional sediment protection guidelines for metals (MoE 1993 Ontario)

Metal	**WHO permissible limit (mg/L)**	**Sediment protection guidelines (ppm)**
Fe	0.30	2
Mn	0.10	460
Cu	0.05	16
Ni	0.05	16
Zn	5.00	120
Pb	0.05	31

Table 3. Bioaccumulation and translocation factor of *Salvinia molesta*

Metal	**BAF**	**TF**
Fe	2.28	3.02
Mn	32.32	2.12
Cu	1.60	1.66
Ni	2.51	4.58
Zn	1.07	2.5
Pb	3.06	3.9

processes, in plants (Doyle et al. 2003). Manganese concentration in water at Syngenta Lake ranged from 0.01 to 0.42 ppm. The concentration was higher than the WHO drinking water limit. In sediments Mn concentration varied from 4.40 to 10.07 ppm. The study revealed that the Mn concentration lower than the MoE of Ontario sediment protection guidelines. In brown fronds of *S. molesta* Mn concentration ranged from 3.04 to 4.75 ppm, while in shoots it ranged from 0.62 to 1.20 ppm. Nickel is released into the environment by various industrial processes. Nickel concentration in water varied from 0.019 to 0.30 ppm at Syngenta Lake, these levels were above drinking water standards by WHO. In Sediment, concentration showed the variations as 0.45 to 2.49 ppm at Syngenta Lake, which was lower than sediment protection guidelines of MoE, Ontario Canada. Nickel concentration ranged from 0.04 to 0.55 ppm in brown fronds and it ranged from BDL to 0.12 ppm, in the shoots of *S. molesta*. The largest natural emission of Zn to water results from erosion, waste disposal, incineration, use of Zn-containing fertilizers, sewage effluent, and agrochemical runoff land fill leachates (Boxall et al. 2000). In present study Zinc concentration in water ranged from 0.09 to 4.45 ppm at Syngenta Lake. The values were nearer to WHO drinking water standards. Sediments of Syngenta Lake showed Zn concentration ranging from 0.95 to 8.47 ppm. It appears that Zn have originated from agrochemical sewage from the vicinity of this Lake. However the concentration was low compared to sediment protection guidelines of MoE, Ontario Canada. Zinc concentration ranged from 0.25 to 3.62 ppm in brown fronds and 0.10 to 1.02 ppm, in shoots of *S. molesta*. According to George and Gabriel (2017) *S. molesta* has efficiency in reducing the Nickel and Zinc concentration from contaminated water. Lead and its compounds enter aquatic ecosystems from industrial effluents, sewage sludge, domestic wastes and combustion of fossil fuels (George and Gabriel 2017). Lead concentration in water of Syngenta Lake ranged from BDL to 0.16 ppm. The total concentration of Pb in water exceeded the WHO limit. In sediment the Pb concentration varied from 2.71 to 3.32 ppm which was lower than the sediment protection guidelines of MoE, Ontario Canada. Lead concentration ranged from BDL to 0.39 ppm in brown fronds and from BDL to 0.10 ppm in shoots of *S. molesta,* respectively. Studies by Jeffery et al. (1976) Mathew et al. 2003 reported that *S. molesta* as hyperaccumulator of Pb. Studies by Kipriyanova (1997) stated that *S. molesta* being a floating hydrophyte, has the highest trace metal accumulating and translocating capabilities.

Bioaccumulation and Translocation Factor

The ratio between trace metal concentration in plant and that of the media (water/sediment) expresses the BAF (Abd-Elmoniem 2003). Translocation is the movement of metal-containing sap from root to shoot of aquatic macrophytes which is primarily controlled by processes like root pressure and leaf transpiration. This reflects the affinity of an aquatic macrophyte to a specific metal element or pollutant. The ambient metal concentration in water can be the major factor influencing the metal uptake efficiency. BAF for trace metals like Mn, Ni, Zn in was highest in *S. molesta* proving it as hyperaccumulator of metals. BAF for analysed metals was in the following order: Mn >Pb> Ni >Zn. According to Yanqun et al. (2005) when the TF value is greater than 1, the plants are considered as ‘accumulator species, whereas when TF value is less than 1 the plants are considered as excluder species. TF values greater than 1 indicate that there is transport of metal from root to leaf and sequestration in leaf vacuoles and apoplast (Zhao et al. 2003) TF for analysed metals was in following order: Ni >Pb> Zn>Mn. Translocation of metals by macrophytes varies with species, presence of metal transporters and availability of binding sites, energy, environmental conditions like pH, photosynthesis, temperature *etc.* metabolic levels and regulatory proteins present in plants (Ghosh and Singh 2005). Trace metal concentration was below detectable level many a times in water, continuous results could not be obtained.

CONCLUSION

The present study revealed that the Lake under study is contaminated with trace metals. It can be concluded that *S. molesta* is suitable for phytoremediation as it is potential scavenger of trace metals and best suited candidate for ‘Green Revolution’ in the clean-up technologies for polluted Lakes. With the advancement in the field of genetic recombination technology, genetically engineered plants can be instrumental in the phytoremediation approaches towards cleaning of aquatic environment.

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