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Excoecaria agallocha: a potential bioindicator of heavy metal pollution

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Abstract: We analyzed the concentrations of zinc, copper and lead in the root, stem and leaf of *Excoecaria agallocha* collected from 12 stations in the north east coast of Bay of Bengal during April, 2013. The region is extremely polluted due to presence of industries, tourism units, fish landing stations and trawler repairing units. In all the selected stations, the metals accumulated in the vegetative parts as per the order root > stem > leaf. In the root region of *E. agallocha*, the concentration of zinc ranged from 17.57 ppm dry wt. (at Bagmara) to 91.84 ppm dry wt. (at Nayachar island). In the stem region, the values ranged from10.05 ppm dry wt. (at Bagmara) to 74.61 ppm dry wt. (at Nayachar island), whereas in the leaf region the values ranged from 8.80 ppm dry wt. (at Bagmara) to 35.04 ppm dry wt. (at Nayachar island). In case of copper, the values in the root region ranged from 11.01 ppm dry wt. (at Bagmara) to 35.25 ppm dry wt. (at Nayachar island). In the stem region, the values ranged from 8.99 ppm dry wt. (at Bagmara) to 30.23 ppm dry wt. (at Nayachar island), and in the leaf region the values ranged from 7.46 ppm dry wt. (at Bagmara) to 19.85 ppm dry wt. (at Nayachar island). The concentrations of lead were lowest in all the vegetative parts and also in all the stations. The values ranged from 3.33 ppm dry wt. (at Bagmara) to 15.61 ppm dry wt. (at Nayachar island) in root, 2.56 ppm dry wt. (at Bagmara) to 8.92 ppm dry wt. (at Nayachar island) in stem and 1.98 ppm dry wt. (at Bagmara) to 6.54 ppm dry wt. (at Nayachar island) in leaf. Simultaneous analyses of dissolved heavy metals in the surface water of the selected stations revealed highest values of zinc followed by copper and lead. Among the 12 selected stations, highest concentrations of dissolved zinc, copper and lead were observed in Nayachar island (540.2 ppb, 159.8 ppb and 42.04 ppb respectively). Station 12 (at Bagmara) exhibited lowest concentrations of dissolved heavy metals viz. 299.47 ppb for zinc, 98.59 ppb for copper and 15.75 ppb for lead.

Keywords: *Excoecaria agallocha*, zinc, copper, lead, bioindicator.

Introduction

Rapid urbanization and industrialization near the coastal mangrove areas within numerous parts of the world have posed a threat to wetland ecosystems. Bioaccumulation of anthropogenic chemicals and non-essential nutrients through the food chain has recently become a matter of concern for several researchers (Alberic et al., 2006). Mangrove ecosystems act as sinks or buffer and they tend to remove or immobilize metals. Numerous studies have utilized mangrove species and their sediments as reliable bio-indicators for heavy metal pollution and contamination (Burchett et al., 2003). Due to close proximity to urban development, mangroves are exposed to significant direct contaminant input, including heavy metals (MacFarlane, 2002). The value of mangrove communities and particularly of mangrove forest sediments, as a buffer between potential sources of metalliferous pollutants and marine ecosystems has been noted previously (Harbison, 1986; Saenger et al., 1991). Several literatures on the response of mangrove species to heavy metal exposure (Chakraborty et al., 2014; Montgomery and Price, 1979; Peterson et al., 1979; Walsh et al., 1979; Thomas and Ong, 1984; Chiu et al., 1995; Chen et al., 1995; Wong et al., 1997) have been published, but detailed studies of heavy metals in mangrove forest of Indian Sundarbans are rare.

The north east coast of Bay of Bengal is exposed to heavy metal pollution from several point and non-point sources (Mitra, 1998; Mitra et al. 2011; Banerjee et al. 2012, Mitra and Ghosh, 2014). Ample studies have not been conducted in India, particularly in the Sundarban mangroves, regarding the incidence of heavy metal status and pollution in these mangrove settings (Untawale et al., 1980; Seralathan, 1987). *Excoecaria agallocha*, a dominant mangrove species in Indian Sundarbans, can withstand a wide range of salinity ranging from 4 psu to 28 psu (Mitra et al., 2010). Several heavy metals like zinc, copper, iron manganese, cobalt, nickel and lead have been reported in the environment of north east coast of Bay of Bengal, but from the viewpoint of abundance the levels of zinc, copper and lead are the highest (Mitra, 1998). Hence, the present study focused on the bioaccumulation pattern of these three heavy metals in the selected mangrove species with the aim to evaluate the efficiency of the species as indicator of selected dissolved heavy metals to which this region is exposed.

Materials and methods

Sampling of *E. agallocha*

Twelve stations were selected in the north east coast of Bay of Bengal in and around Indian Sundarbans mangrove ecosystem (Table 1). *E. agallocha* was collected at ebb within 500 meter coastal stretch (from the low tide line) of the selected stations during 5 - 15th April, 2013. The collected samples were segregated into root, stem and leaf, washed with ambient sea water and brought to laboratory. The segregated samples were washed with double distilled water, dried with tissue paper and stored at -20^{0}C for further analysis.

Analysis of dissolved Zn, Cu and Pb

Surface water samples were collected using 10-l Teflon-lined Go-Flo bottles, fitted with Teflon taps and deployed on a rosette or on Kevlar line, with additional surface sampling carried out by hand. Shortly after collection, samples were filtered through Nuclepore filters (0.4 µm pore diameter) and aliquots of the filters were acidified with sub-boiling distilled nitric acid to a pH of about 2 and stored in cleaned low-density polyethylene bottles. Dissolved heavy metals were separated and pre-concentrated from the seawater using dithiocarbamate complexation and subsequent extraction into Freon TF, followed by back extraction into HNO_3 as per the procedure of Danielsson et al (1978). Extracts were analyzed for Zn, Cu and Pb by Atomic Absorption Spectrophotometer (Perkin Elmer: Model 3030). The accuracy of the dissolved heavy metal determination is indicated by good agreement between our values and reported for certified reference seawater materials (CASS 2) (Table 2).

Analysis of tissue Zn, Cu and Pb

Inductively Coupled Plasma – Mass Spectrometry (ICP-MS) is now - a - day accepted as a fast, reliable means of multi-elemental analysis for a wide variety of biological sample types. A Perkin-Elmer Sciex ELAN 5000 ICP mass spectrometer was used for analysis of selected heavy metals in the root, stem and leaf tissues of *E. agallocha*. A standard torch for this instrument was used with an outer argon gas flow rate of 15 L/min and an intermediate gas flow of 0.9 L/min. The applied power was 1.0 kW. The ion settings were standard settings recommended, when a conventional nebulizer/spray is used with a liquid sample uptake rate of 1.0 mL/min. A Moulinex Super Crousty microwave oven of 2450 MHz frequency magnetron and 1100 Watt maximum power Polytetrafluoroethylene (PTFE) reactor of 115 ml volume, 1 cm wall thickness with hermetic screw caps, were used for the digestion of the root, stem and leaf samples. All reagents used were of high purity available and of analytical reagent grade. High purity water was obtained with a Barnstead Nanopure II water-purification system. All glasswares were soaked in 10% (v/v) nitric acid for 24 h and washed with deionised water prior to use.

Data analysis

Statistical software SPSS 14.0 was used to determine the inter-relationships of heavy metal concentrations in the vegetative parts of *E. agallocha* and dissolved heavy metals in aquatic environment through Pearson correlation coefficient analysis.

Results

Dissolved heavy metal

The dissolved heavy metals were observed to follow the trend Zn > Cu > Pb irrespective of all stations. Dissolved Zn ranged from 299.47 ppb (at Bagmara) to 540.2 ppb (at Nayachar island). Dissolved Cu ranged from 98.59 ppb (at Bagmara) to 159.8 ppb (at Nayachar island), whereas, dissolved Pb ranged from 15.75 ppb (at Bagmara) to 42.04 ppb (at Nayachar island) (Figure 1).

Bioaccumulation pattern

In *E. agallocha* samples, the heavy metals varied as per the order Zn > Cu > Pb. This sequence is uniform in all the twelve selected stations during the study period. In the present study, the concentration of Zn in root ranged from 17.57 ppm (at Bagmara) to 91.84 ppm (at Nayachar island). Cu ranged from 11.01 ppm (at Bagmara) to 35.25 ppm (at Nayachar island) and Pb ranged from 3.33 ppm (at Bagmara) to 15.61 ppm (at Nayachar island) (Figure 2).

The concentration of Zn in stem ranged from 10.05 ppm (at Bagmara) to 74.61 ppm (at Nayachar island). Cu ranged from 8.99 ppm (at Bagmara) to 30.23 ppm (at Nayachar island) and Pb ranged from 2.56 ppm (at Bagmara) to 8.92 ppm (at Nayachar island) (Figure 3).

The concentration of Zn in leaf ranged from 8.80 ppm (at Bagmara) to 35.04 ppm (at Nayachar island) and Cu ranged from 7.46 ppm (at Bagmara) to 19.85 ppm (at Nayachar island). Concentration of Pb ranged from 1.98 ppm (at Bagmara) to 6.54 ppm (at Nayachar island) (Figure 4).

Discussion

Heavy metal pollution in the estuarine sector is the result of land run-off, mining, activities like shipping and dredging and anthropogenic inputs (Panigrahy et al., 1997). The main sources of zinc in the present geographical locale are the galvanization units, paint manufacturing units and pharmaceutical processes, whereas the main sources of copper in the coastal waters are antifouling paints, particular type of algaecides used in different aquaculture farms, paint manufacturing units, pipe line corrosion and oil sludges (32 to 120 ppm). Ship bottom paint has been found to produce very high concentration of Cu is sea water and sediment in harbors of Great Britain and southern California (Bellinger and Benham 1978; Young et al. 1979). The most toxic of these three heavy metals is lead, which finds its way in coastal waters through the discharge of industrial waste waters, such as from painting, dyeing, battery manufacturing units and oil refineries *etc*. Antifouling paints used to prevent growth of marine organisms at the bottom of the boats and designed to constantly leach toxic metals into the water to kill organisms that may attach to bottom of the boats, ultimately get transported to the sediment and aquatic compartments. The study area is exposed to all these activities being proximal to the highly urbanized city of Kolkata, Howrah and the newly emerging Haldia port-cum- industrial complex (Mitra and Choudhury, 1993; Mitra, 1998; Mitra et al, 2011; Mitra et al., 2012).

The bioaccumulation pattern in plant parts like leaves, bark and roots may vary depending on the concentration of heavy metals in the sediment, the types of heavy metals and also the tolerance of the species and its parts towards the heavy metals (Baker and Walker, 1990; De Lacerda and Abrao, 1986). Heavy metal concentration in plant tissues is influenced by the metabolic requirements for essential micronutrients such as Cu and Zn, while non-essential metals like Pb tend to be excluded or compartmentalized (Baker and Walker,1990). Zinc is an essential micro-nutrient of numerous enzyme systems, respiration enzyme activators, and the biosynthesis of plant growth hormones (Ernst et al., 1992). Copper is an essential micronutrient required in mitochondria and chloroplast reactions, enzyme systems related to photosystem II electron transport, cell wall lignification, carbohydrate metabolism, and protein synthesis (Verkleij and Schat, 1990). The mobility of metals (in the order of Zn > Cu > Pb) is thus justified given the role played by essential metals like Zn and Cu in plant metabolism while the non essential metal like Pb was rather restricted in translocation to the upper plant parts. The order of the heavy metals in the vegetative parts of *E. agallocha* reflects that of the dissolved heavy metals, which clearly confirms the use of the species as effective indicator of dissolved Zn, Cu and Pb in the northeast coast of Indian subcontinent particularly in the inshore region of Bay of Bengal. The heavy metals from the ambient environment enter the coastal vegetation very frequently as they are inundated during high tide twice daily. The roots are exposed to water and sediment during most of the period, whereas the leaves come in contact with water during the high tide phase. This may be one of the reason of the maximum concentrations of heavy metals in the root followed by stem and leaf. Several researchers also observed variation in metal level in the vegetative parts of coastal vegetation particularly mangroves.

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REFERENCES:

1. Alberic P, Baillif P, Baltzer F, Cossa , Lallier Verges, E. 2006. Heavy metals distribution in mangrove sediments along the mobile coastline of French Guiana, *Marine Chemistry*, **98**, 1-17.

2. Baker AJ, and Walker PI. 1990. Ecophysiology of metal uptake by tolerant plants, In: Shaw AJ (ed.) Heavy metal tolerance in plants; evolutionary aspects, CRC Press, Florida, 155–178.

3. Banerjee K, Roy Chowdhury M, Sengupta, K, Sett, S and Mitra, A. 2012. Influence of anthropogenic and natural factors on the mangrove soil of Indian Sundarban Wetland, *Architectural Environmental Science*, **6**, 80-91.

4. Bellinger E and Benhem B. 1978. The levels of metals in Dockyard sediments with particular reference to the contributions from ship bottom paints. *Environmental Pollution Assessment*, **15(1)**, 71-81.

5. Burchett MD, MacFarlane GR and Pulkownik A. 2003. Accumulation and distribution of heavy metals in the grey mangrove *Avicennia marina* (Forsk.) Vierh.: biological indication potential, *Environmental Pollution*, **123**, 139-151.

6. Chakraborty S, Trivedi S, Fazli P, Zaman S and Mitra M. 2014. *Avicennia alba*: an indicator of heavy metal pollution in Indian Sundarban estuaries, *Journal of Environmental Science, Computer Science and Engineering & Technology*, **3(4):** 1796-1807.

7. Chen GZ, Miao SY, Tam N, Wong FYS, Li SH and Lan CY. 1995. Effects of synthetic wastewater on young *Kandelia candel* plants growing under greenhouse conditions. *Hydrobiologia*, **295**,263-273.

8. Chiu CY, Hsiu, FS, Chen SS and Chou, CH. 1995. Reduced toxicity of Cu and Zn to mangrove seedlings in saline environments. *Botanical Bulletin Academy*, Singapore, **36**, 19-24.

9. Danielsson LG, Magnusson B and Westerlund S. 1978. An improved metal extraction procedure for the determination of trace metals in seawater by atomic absorption spectrometry with electrothermal atomization. *Analytical Chemistry Acta*, **98**, 45 – 57.

10. De Lacerda LD and Abrao JJ. 1986. Heavy metal accumulation by mangrove and saltmarsh intertidal sediments. *Marine Pollution Bulletin*, **17**, 246-250.

11. Ernst WHO, Verkleij JAC and Schat H. 1992. Metal tolerance in plants, *Acta Botanica Neerlandica*, **41**. 229–248.

11. Harbison P. 1986. Mangrove muds, A sink and a source for trace metals, *Marine Pollution Bulletin*, **17**, 246-250.

12. MacFarlane GR. 2002. Leaf biochemical parameters in Avicennia marina (Forsk.) Vierh as potential biomarkers of heavy metal stress in estuarine ecosystems, *Marine Pollution Bulletin*, **44**, 244–256.

13. Mitra A. 1998. Status of coastal pollution in West Bengal with special reference to heavy metals, *Journal of Indian Ocean Studies*, **5(2)**, 135-138.

14. Mitra A, Chakraborty R, Sengupta K and Banerjee K. 2011. Effects of various cooking processes on the concentrations of heavy metals in common finfish and shrimps of the River Ganga, *National Academy of Science Letters*, **34 (3 & 4)**, 161 – 168.

15. Mitra A and Choudhury A. 1993. Heavy metal concentrations in oyster *Crassostrea cucullata* of Sagar Island, India. *Indian Journal of Environmental Health*, NEERI, **35 (2)**, 139-141.

16. Mitra A, Chowdhury R, Sengupta K and Banerjee K. 2010. Impact of salinity on mangroves of Indian Sundarbans, *Journal. of Coastal Environment*, **1(1)**, 71-82.

17. Mitra A, Choudhury R and Banerjee K. 2012. Concentrations of some heavy metals in commercially important finfish and shellfish of the River Ganga. *Environmental Monitoring and Assessment*, **184**, 221 – 223 (SPRINGER DOI 10.1007/s10661-011-2111-x).

18. Mitra A and Ghosh R. 2014. Bioaccumulation Pattern of Heavy Metals in Commercially Important Fishes in and Around Indian Sundarbans, *Global Journal of Animal Science* Research, **2(1).**

19. Montgomery JR. and Price MT. 1979. Release of trace metals by sewage sludge and the

subsequent uptake by members of a turtle grass mangrove ecosystem. *Environmental Science & Technology*, **13**, 546-549.

20. Panigrahy PK, Nayak BB, Acharya BC, Das SN, Basu SC & Sahoo RK. 1997. Evaluation of heavy metal accumulation in coastal sediments of northern Bay of Bengal. 139-146. *In*: C.S.P. Iyer (ed.), *Advances in Enironmental Science*, Educational Publishers and Distributors, New Delhi.

21. Peterson PJ, Burton MA, Gregson M, Nye SM and Porter EK. 1979. Accumulation of tin by mangrove species in West Malaysia, *Science of the Total Environment*, **11**, 213-221.

22. Saenger P. 2002. Mangrove ecology, silviculture and conservation. Kluwer Academic Publishers, 360.

23. Seralathan P. 1987. Trace element geochemistry of modern deltaic sediments of the Cauvery River, east coast of India, Indian *Journal of Marine Science*, **16**, 235–239.

24. Thomas C and Ong JE. 1984. Effect of heavy metals zinc and lead on *Rhizophora mucronata* Lam. and *Avicennia alba* Bl. seedlings. In: Soepadmo, E., A.N. Rao and D.J.McIntosh (Eds.), Proceedings of the Asian Symposium Mangrove Environments - Research and Management, UNESCO, 568-574.

25. Untawale A.G., Wafar S., and Bhosle N.B., 1980. Seasonal variation in heavy metal concentration in mangrove foliage, *Mahasagar-Bulletin of the National Institute of Oceaography*, **13(3)**, 215-223.

26. Verkleij JAC and Schat H. 1990. Mechanisms of metal tolerance in plants. In: Shaw AJ (ed) Heavy metal tolerance in plants-evolutionary aspects, CRC Press,
Florida, 179–193.

27. Walsh GER, Ainsworth KA and Rigby, R. 1979. Resistance of red mangrove (*Rhizophora*
mangle L.) seedlings to lead, cadmium and mercury. *Biotropica*, **11**, 22-27.

28. Wong YS, Tam NFY and Lan CY. 1997. Mangrove wetlands as wastewater treatment facility: a field trial, *Hydrobiologia*, **352**, 49-59.

29. Young DR, Alexander GV, McDermott-Ehrlich D. 1979. Vessel related contamination of southern California harbours by copper and other metals. *Marine Pollution Bulletin*, **10**, 50-56.

Table 1. Coordinates of selected stations

Stations	Latitude	Longitude
Canning	22°18′37"N	88°40′36"E
Gosaba	22° 15' 45" N	88° 39' 46" E
Diamond Harbour	22° 11' 30" N	88° 11' 4" E
Nayachar island	21° 45' 24" N	88° 15' 24" E
Kakdwip	21°52′06″N	88°11′12″E
Chemaguri	21°38'25.86"N	88°08'53.55" E
Sagar South	21° 38' 51.55" N	88° 02' 20.97" E
Jambu island	21°35'42.03"N	88°10'22.76"E
Frasergunge	21° 33′ 47.76″ N	88° 15′ 33.98″ E
Digha	21° 42' 50.03" N	87° 05' 20.12" E
Bali	22º04'35.17"N	88º44'55.70"E
Bagmara	21°39' 4.45"N	89°04' 40.59" E

Table 2. Analysis of reference material for near shore seawater (CASS 2)

Element	Certified value ($\mu g\ l^{-1}$)	Laboratory results ($\mu g\ l^{-1}$)
Zn	1.97 ± 0.12	2.01 ± 0.14
Cu	0.675 ± 0.039	0.786 ± 0.058
Pb	0.019 ± 0.006	0.029 ± 0.009

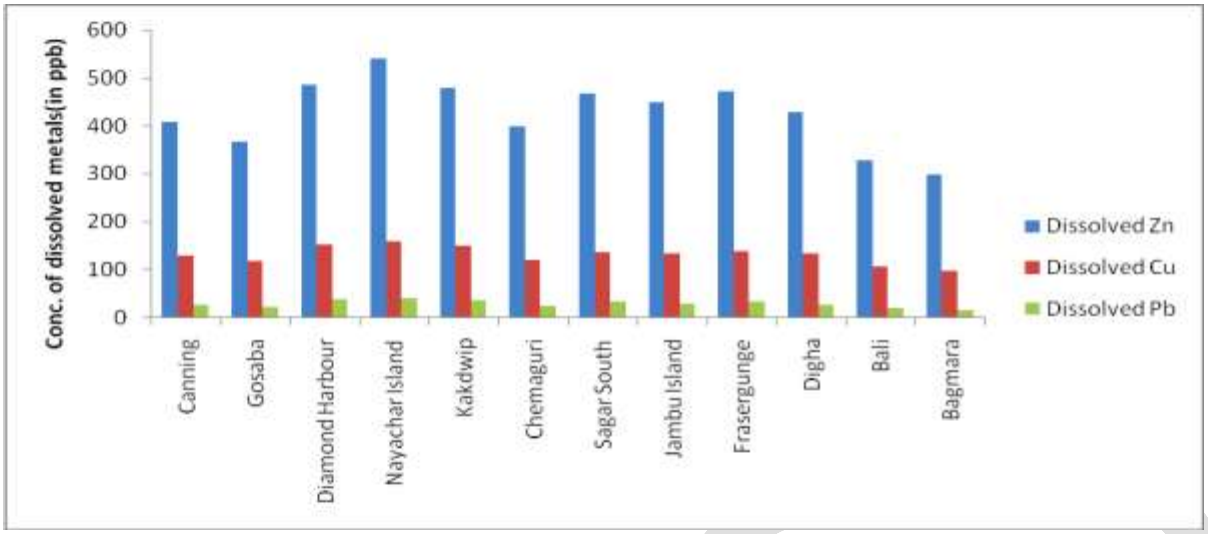

Figure 1. Spatial variations of dissolved heavy metal concentrations(in ppb)

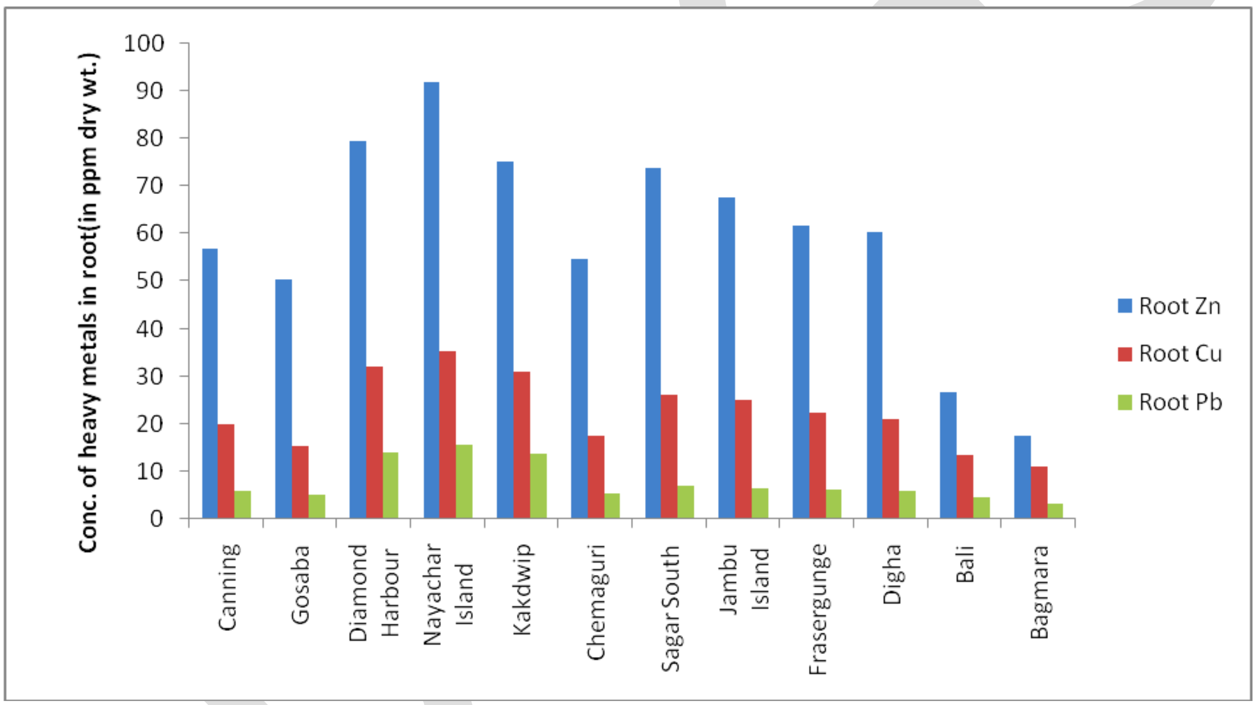

Figure 2. Spatial variations of heavy metal concentrations in root (ppm dry wt.) of *E. agallocha*

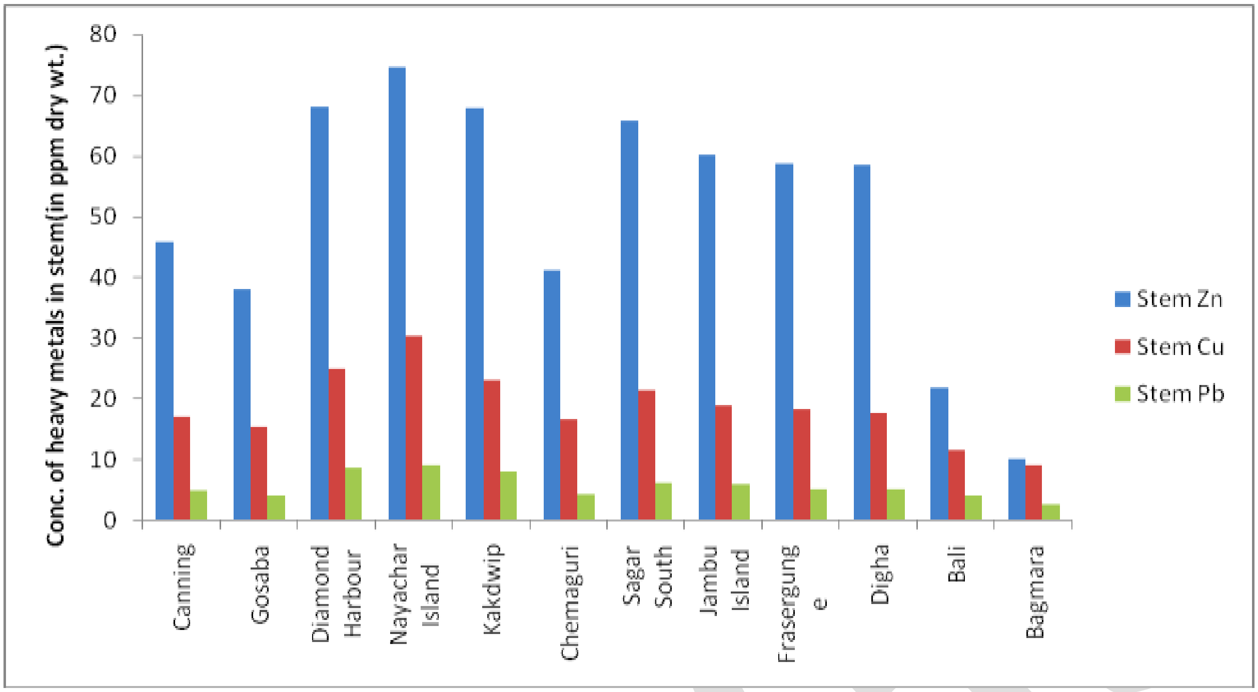

Figure 3. Spatial variations of heavy metal concentrations in stem (ppm dry wt.) of *E. agallocha*

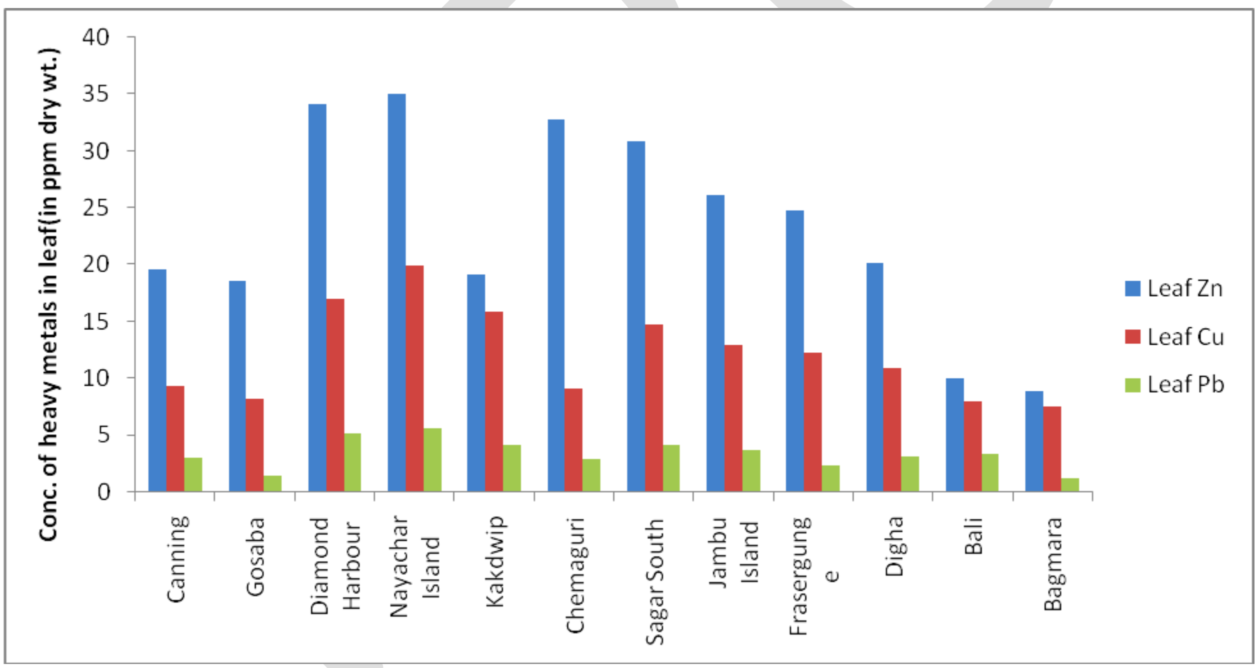

Figure 4. Spatial variations of heavy metal concentrations in leaf (ppm dry wt.) of *E. agallocha*

Table 3

Inter-relationships between heavy metal concentrations in *E. agallocha* and ambient environment

Study period	Combination	r-value	p-value
April, 2013	Root Zn x Dissolved Zn	0.960899	<0.01
	Stem Zn x Dissolved Zn	0.976413	<0.01
	Leaf Zn x Dissolved Zn	0.793961	<0.01
	Root Cu x Dissolved Cu	0.968539	<0.01
	Stem Cu x Dissolved Cu	0.962121	<0.01
	Leaf Cu x Dissolved Cu	0.922790	<0.01
	Root Pb x Disosolved Pb	0.880778	<0.01
	Stem Pb x Dissolved Pb	0.947862	<0.01
	Leaf Pb x Dissolved Pb	0.961542	<0.01