DISTRIBUTION OF COPPER IN THE SEPANG MANGROVE RESERVE FOREST ENVIRONMENT, MALAYSIA

Mahmood Hossain*,

Forestry and Wood Technology Discipline, School of Life Science, Khulna University, Khulna 9100, Bangladesh

Saberi Othman, Japar Sidik Bujang

Faculty of Science and Environmental Studies, Universiti Putra Malaysia, 43400 Serdang, Selangor, Malaysia

&

Lim Meng Tsai

Faculty of Forestry, Universiti Putra Malaysia, 43400 Serdang, Malaysia

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MAHMOOD, H., SABERI, O., JAPAR SIDIK, B. & LIM, M.T. 2001. Distribution of copper in the Sepang mangrove reserve forest environment, Malaysia. The Sepang mangrove reserve forest is being polluted by waste disposal from different sources such as pig farms, oil palm industries and households. Of the discharges untreated waste from pig farms has an important role in copper pollution in this forest area. Soil is the main reservoir of copper in this mangrove forest. Total and available copper concentrations in soil were highest in the stations nearest to the waste discharge point and decreased towards the stations near to the sea. The mean total and available copper concentrations in the soil were 145 $\mu g\ g^{-1}$ and 36 $\mu g\ g^{-1}$ respectively, whereas the mean values of copper concentration in the river and infiltration water were 0.07 mg L^{-1} and 0.04 mg L^{-1} respectively. The highest mean copper concentration in *Rhizophora mucronata* seedlings was in the roots (4.54 $\mu g\ g^{-1}$), followed by the leaves (2.58 $\mu g\ g^{-1}$) and stems (2.19 $\mu g\ g^{-1}$).

Key word: Copper - mangrove - micronutrient - heavy metal - organic waste - pig farms

MAHMOOD, H., SABERI, O., JAPAR SIDIK, B. & LIM, M. T. 2001. Taburan kuprum di persekitaran hutan simpan bakau di Sepang, Malaysia. Hutan simpan bakau Sepang telah dicemari oleh sisa buangan daripada sumber-sumber yang berbeza seperti ladang ternakan khinzir, industri kelapa sawit dan isi rumah. Pembuangan sisa yang tidak dirawat dari ladang ternakan khinzir memainkan peranan yang penting dalam pencemaran kuprum di kawasan hutan ini. Tanah merupakan takungan kuprum yang utama di hutan bakau ini. Jumlah dan ketersediaan kepekatan kuprum di dalam tanah

**Author for correspondence.*

adalah tertinggi di stesen yang berdekatan dengan laut. Min kepekatan jumlah dan kepekatan ketersediaan kuprum di dalam tanah masing-masing ialah 145 $\mu g\ g^{-1}$ dan 36 $\mu g\ g^{-1}$, manakala nilai min kepekatan kuprum di dalam sungai dan penyusupan air masing-masing ialah 0.07 mg L^{-1} dan 0.04 mg L^{-1}. Min kepekatan kuprum yang tertinggi di dalam anak benih *Rhizophora mucronata* ialah di dalam akar (4.54 $\mu g\ g^{-1}$), diikuti oleh daun (2.58 $\mu g\ g^{-1}$) dan batang (2.19 $\mu g\ g^{-1}$).

Introduction

Pollution by heavy metals to the terrestrial environment is not a recent occurrence. It has appeared from the beginning of human civilisation, but nowadays, has become alarming with increasing industrialisation. Metals are ubiquitous in trace concentrations in soil and vegetation; in fact some of them are required by plants as micronutrients. Large concentrations of these trace elements/ metals can be toxic to plants. Copper is the most extensively used metal by men for different activities (Adriono 1986).

Slurries of pig wastes contain as much as 750 $\mu g\ g^{-1}$ of copper (Robinson *et al.* 1971) and copper concentrations up to 800 $\mu g\ g^{-1}$ (dry weight basis) are not unusual which are similar to those in the sewage sludge from industrial areas (Purves 1977).

Gambrell *et al.* (1991) reported that in brackish marsh sediment, soluble copper level increases with increasing salinity, redox potential, etc. Copper concentration of soil at Ranong mangrove forest, Thailand, varied with depth and also the soil type. It generally ranged from 0.4 to 1.8 $\mu g\ g^{-1}$ (Macintosh *et al.* 1990). Copper concentration can reached as high as 500 $\mu g\ g^{-1}$ in an area adjacent to the forest area which is heavily polluted by animal discharge from pig farms (Saberi 1997).

Absorption rate of Cu by plant roots is lowest among the essential elements and a linear relationship is shown between absorption rate and external Cu concentration, or a curvilinear relationship in the high concentration range (Graham 1981). Once copper is absorbed, it accumulates in different parts of the plant and high accumulation in roots causes root damage by copper toxicity (Andrew & Thorne 1962, Dykeman & de Sousa 1966, Brams & Fiskell 1971).

Silva *et al.* (1990) stated that the mangrove plant usually takes up a small proportion of metals from the soil which contains a high level of metal concentration and therefore with very low cycling. He also noted that copper concentration in the roots of *Rhizophora mangle* was higher than in the other parts. Lacerda *et al.* (1991) noted that metal concentration in mangrove plant leaves varied little although the soil is enriched with a high metal level. The present study therefore aims firstly to determine the total and available copper concentrations in mangrove soil and the total copper in the water environment, and secondly to determine the copper accumulation and distribution in different parts of *Rhizophora mucronata* seedlings.

Materials and methods

Site description

This study was conducted at the Sepang mangrove reserve forest in Selangor, Malaysia. The Sepang mangrove reserve is situated at both sides of the Sepang Besar River which is at the state boundary between Selangor and Negri Sembilan. This mangrove forest is located between latitudes 2° 36'–2° 41' N and longitudes 101° 42'–101° 46' E (Figure 1). There are very few plant species found in this forest area of which *Rhizophora mucronata, R. apiculata, Avicennia alba, Sonneratia alba, Bruguiera gymnorrhiz, Ceriops tagal* and *Xylocarpus granatum* are most common. Of these species *R. mucronata* has a wide distribution and the highest density (350 trees per ha) (Saberi 1993).

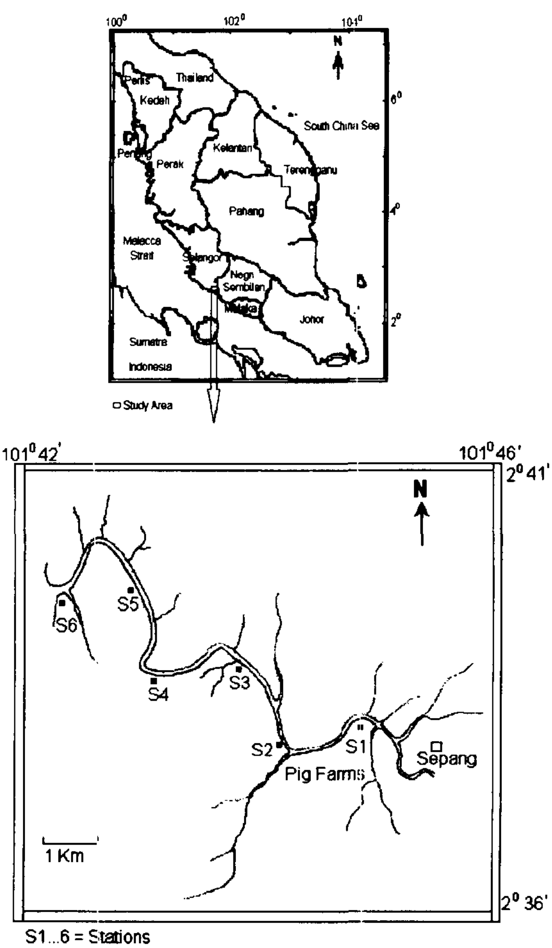

Figure 1. Map of the Sepang mangrove reserve forest area showing the location of Sepang Besar River with sample stations

Sampling procedure

Six stations were chosen starting from the waste discharge point of a pig farm at every 2 km interval towards the sea (Figure 1). Two plots (3 × 3 m) were taken from each station and each divided into three sub-plots of 3 × 1 m size. Total and available soil copper and copper concentrations in infiltration water were measured for each station. *Rhizophora mucronata* seedlings were not found at stations 1, 2 and 3, hence copper concentration was also measured in river water adjacent to each station. Copper concentrations in leaves, stems and roots of *R. mucronata* seedling were measured in the last three stations towards the sea.

Soil analysis

Topsoil was collected by digging a hole of (10 × 10 × 10 cm) in 3 sub-plots of each main plot. One soil sample was taken from each sub-plot. Samples were air-dried at room temperature and thoroughly mixed before crushing and sieving. Hesse's (1971) method was followed for total soil copper extraction using concentrated HNO_3 and 60% $HClO_4$ at proportion 2:1. Wear and Sommer's (1948) method was employed for determining available soil copper where 0.1M HCl was used for extraction.

Water analysis

After taking a soil sample, accumulation of infiltration water can usually be observed at the bottom of the hole. Once the column reached equilibrium, the water in the hole was rapidly displaced with suction bomb and transferred to the bottle. Infiltration water samples were collected from all sub-plots of all stations. River surface water was collected at mid-width of the river adjacent to the plots of each station. Two river water samples were collected from each station. Total (dissolved and suspended) substances of river and infiltration water were brought into solution using concentrated H_2SO_4, 60% $HClO_4$ and HNO_3 at a proportion of 0.5:1:5 (Allen 1974).

Plant analysis

Three seedlings of *R. mucronata* having four leaves were collected from sub-plots of stations 4, 5 and 6. Samples were separated into parts (i.e. leaves, stems and roots) and then oven-dried at 80 °C. The dried material was then ground and sieved. Concentrated HNO_3 and 60% $HClO_4$ at proportion 1:1 were used for plant sample extraction (Jones *et al.* 1991). Atomic absorption spectrophotometer (AAS) Perkin Elmer 4100 was used for measuring copper ion concentration in the extract. Analysis was performed in duplicate.

Soil and water parameters other than copper

Texture of soil samples was measured by the pipette method (Black 1965). Fresh moisture content was measured by the weight loss method and cation exchange capacity of soil was measured by the distillation and titration method (Allen 1974). Soil organic matter content was determined by the ignition method (Allen 1974) where oven-dried (at 105 °C) soil sample was ignited at 450 °C for burning out the organic matter. A pH meter with automatic temperature compensator (Ciba-Corning Diagnostic Ltd.) whose accuracy was ± 0.05 was used to measure soil pH *in situ* from slurries of the sample in water (Peech 1964). The pH of river water samples was measured in the field following the same principle as described for soil sample. A conductivity and TDS cell (Ciba-Corning Diagnostic Ltd.) was used to determine the conductivity and total dissolved solids of the river water in the field. River water salinity was measured directly in the field using a salinity meter (YSI model 33 manufactured by Yellow Springs Instrument Co. USA). Dissolved oxygen was measured by (HI 9142) oxygen meter (HANNA Instrument, USA) which gave the reading in mg L^{-1}.

Results and discussion

Copper in soil

The mean values of the measured soil parameters at the six different stations are presented in Table 1. Very high copper concentration was found in soils of stations near to the discharge point. In stations 1, 2 and 3 (near to the discharge point) total soil copper concentration ranged from 517.63 to 95.25 µg g^{-1} (Table 1). These values are about 517 to 95 times higher than the concentration allowed by the Department of Environment, Malaysia (Saberi 1997). Available forms of copper in soil were also very high all over the study area. Stations 1, 2 and 3 contained 91.22 to 30.47 µg g^{-1} of copper in plant available form.

Total and available soil copper were positively correlated (r = 0.72 and 0.8) with soil organic matter. Higher copper level was observed in stations possessing high organic matter and high percentage of clay content. Copper could have higher affinity for soil organic matter and with clay content. Indigenous soil organic matter and added organic waste to the soil bind copper in soils (Adriano 1986) and copper is strongly held in soil with organic matter (Kabata-Pendias 1963). Copper content in soil increases with increasing clay content (Neelakanta & Mehta 1961) and decreases in coarse textured soils (Jarvis 1981). Total soil copper showed positive correlation (r = 0.51) with *in situ* soil pH. The availability, solubility and mobility of soil copper decrease at a soil pH above 7 and this copper becomes more available at pH below 5 (Lucas & Kenzek 1972).

Table 1. The mean values with standard errors of soil and water parameters measured at different stations

Parameter	Mean value for station 1	Mean value for station 2	Mean value for station 3	Mean value for station 4	Mean value for station 5	Mean value for station 6
<u>Soil parameter</u>						
*pH *in situ*	7.08 ± 0.17	6.98 ± 0.07	6.92 ± 0.02	6.36 ± 0.02	6.44 ± 0.06	5.60 ± 0.28
*pH after treatment with H_2O_2	4.24 ± 0.07	3.73 ± 0.03	3.72 ± 0.05	3.59 ± 0.15	2.63 ± 0.07	1.77 ± 0.04
*Fresh moisture content (%)	71.91 ± 0.64	69.01 ± 0.93	63.03 ± 1.07	48.82 ± 0.91	34.33 ± 1.43	27.94 ± 0.58
*Organic matter content (%)	20.46 ± 0.32	15.21 ± 0.52	14.49 ± 0.18	11.17 ± 0.40	6.22 ± 0.35	4.71 ± 0.36
*CEC (m.e./100 g)	21.13 ± 0.12	20.04 ± 0.18	18.85 ± 0.15	18.12 ± 0.02	11.17 ± 0.26	4.03 ± 0.08
*Total soil copper ($\mu g\ g^{-1}$)	517.63 ± 47.46	212.50 ± 12.29	95.25 ± 3.76	28.87 ± 2.13	13.64 ± 0.61	2.08 ± 0.21
*Available copper ($\mu g\ g^{-1}$)	91.22 ± 4.57	76.19 ± 3.00	30.47 ± 1.41	10.58 ± 1.84	6.53 ± 0.65	1.01 ± 0.08
<u>Water parameter</u>						
**pH	5.78 ± 0.02	6.23 ± 0.07	6.20 -	6.71 -	6.92 ± 0.02	6.94 -
**Salinity (ppt.)	21.50 ± 0.50	23.00 -	24.00 -	26.00 -	26.75 ± 0.75	32.50 ± 0.50
**Dissolved oxygen (mg L^{-1})	1.45 ± 0.05	2.25 ± 0.15	3.90 -	5.80 -	6.65 ± 0.35	7.10 ± 0.10
**Copper content in river water (mg L^{-1})	0.09 -	0.08 -	0.08 -	0.06 -	0.05 -	0.05 -
*Copper content in infiltration water (mg L^{-1})	0.05 ± 0.01	0.05 ± 0.01	0.04 -	0.04 -	0.03	0.02 -

* Results are means of 6 samples ± S.E.
**Results are means of 2 samples ± S.E.

Total and available soil copper showed positive correlation (r = 0.6 and 0.72) with cation exchange capacity of soil, and soil copper content increased with increasing CEC. It was found that both of the copper contents in soil exceeded 5% of the CEC value. According to Mathur and Levesque (1983), copper level in soil becomes phytotoxic when the copper content exceeds 5% of the soil CEC value. Mangrove soil may have the characteristics to accumulate high copper content. This opinion agrees with the observations of Harbinson (1981), Lacerda and Abrao (1984), and Silva *et al.* (1990). Total soil copper content at the study area was 19 to 103 times higher than those at the Kapar mangrove forest, Malaysia (Saberi 1997), Ranon mangrove reserve, Thailand (Macintosh *et al.* 1990), and Red Mangrove Forest, Brazil (Silva *et al.* 1990). These mangrove forests were reported as unpolluted from copper. On the other hand, the soil of the study area contained 5 to 32 times higher copper content than the soils at Sai Keng and Shenzhen mangrove forests at Hong Kong which were reported to be polluted by heavy metals (Tam & Wong 1996).

Copper in water

River water was black in colour particularly at low tide and its darkness increased towards the waste discharge point. Station 1 contained the highest copper in river and infiltration water samples and the copper concentrations decreased towards station 6. At station 1 copper concentrations in river and infiltration water were 0.09 mg L^{-1} and 0.05 mg L^{-1} respectively (Table 1). A high positive correlation (r = 0.93) was found between infiltration water and river water copper concentrations. During high tide, river water enters into the forest floor and percolates through the soil profile; this could be the reason for the positive correlation with river water copper content. Acidic pH was observed in the river water at the study area ranging from 5.78 to 6.94 (Table 1). Acidic water pH at the study area may promote the release of copper from the water body and hence absorption by the sediment.

Dissolved oxygen of the river water at stations 1, 2 and 3 was very low (Table 1) compared to normal level (5 to 7 mg L^{-1}) (Pryde 1973, Smith 1990). Copper complexes with organic waste and enters the water environment through the discharge of the pig farm. Thus, high copper concentrations in river water at the stations near to the discharge point may be detected in low dissolved oxygen condition in river water. The results show that the river water at stations 1, 2 and 3 contained much higher amounts of organic waste than stations 4, 5 and 6.

Copper in Rhizophora mucronata

Seedlings of *R. mucronata* were not found in stations 1, 2 and 3. Copper concentrations in seedling parts at stations 4, 5 and 6 are presented in Table 2. Seedling roots at station 4 contained the highest copper (9.24 $\mu g\ g^{-1}$) and the lowest was at station 6 (1.54 $\mu g\ g^{-1}$). The copper concentration in seedlings at stations 4 and 5 was highest in the roots followed by stems and leaves. But at station 6 copper concentration was highest in leaves followed by stems and roots (Table 2).

Table 2. Copper concentration in different parts of seedlings at individual stations

Station	Soil texture	Mean ± S.E. value for leaves ($\mu g\ g^{-1}$)	Mean ± S.E. value for stems ($\mu g\ g^{-1}$)	Mean ± S.E. value for roots ($\mu g\ g^{-1}$)
4	Silty clay	3.1 ± 0.21	3.12 ± 0.26	9.24 ± 0.36
5	Sandy clay loam	2.39 ± 0.1	1.82 ± 0.14	2.84 ± 0.24
6	Sandy	2.25 ± 0.1	1.64 ± 0.14	1.54 ± 0.08

Results are means of 18 samples ± S.E.

Not only the seedlings of *R. mucronata* but also the seedlings of other mangrove plants were absent at stations 1, 2 and 3. These stations contained high amounts of total and available soil copper ranging from 517.63 to 95.25 $\mu g\ g^{-1}$ and from 30.47 to 91.22 $\mu g\ g^{-1}$ respectively (Table 1). This high copper level in soil could be the reason for the unavailability of seedlings.

The low copper detected in seedlings at station 6 may be due to the lower level of soil copper in station 6 compared to stations 4 and 5. Plant copper content showed positive correlation with total ($r = 0.88$) and available ($r = 0.7$) forms of soil copper. The availability of copper and uptake by plants related to the copper concentration in soil (Fleming 1965) and copper concentration in plant tissue significantly correlate with the total copper concentration in soil (Sloan *et al.* 1997).

Soil moisture content was also high in stations 4 and 5 compared to station 6 (Table 1). Soil moisture has an influence on copper availability to plants where a positive correlation ($r = 0.93$) was found between plant copper content and soil moisture. Soil moisture is reported as an important factor that is able to bring the insoluble form of copper into soluble form and made available for plant uptake (Mitchell 1964, Caldwell 1971). Plant copper content also showed a positive correlation ($r = 0.9$) with soil organic matter content. Organic matter in soil helps to bind as well as enhance affinity of copper to soil (Adriano 1986) and most of the soil copper is associated with organic matter (Jarvis 1981). So it was not surprising to get positive correlation between plant copper content and soil organic matter content in the study area. The above discussion illustrates the relationship of plant copper content with soil moisture and organic matter content.

Roots absorbed higher copper compared to leaves and stem except at station 6 (Table 2). Copper enters into the plant through the root system and its distribution in roots and shoots varies widely with plant species and the level of copper supply. The concentration of copper in roots of most species increases much more rapidly than in shoots with increasing copper level in soil. But higher copper concentration in leaves than roots was found in station 6 (Table 2).

Seedling parts of station 6 contained larger copper concentrations than soil level (Table 1 and 2) though this station contained less soil copper than stations 4 and 5. More acidic soil pH of 5.60 was observed at station 6 compared to stations 4 and 5 (Table 1). Soil pH may be an important factor for the availability of copper to the seedlings (Lucas & Knezek 1972, Mulchi *et al.* 1992). The solubility, mobility and availability of copper to plant are largely dependent on soil pH and its availability is drastically reduced at a soil pH above 7 and is most readily available below pH 6 (Adriano 1986) and especially at pH below 5 (Lucas & Knezek 1972).

Rhizophora mucronata may have the ability to tolerate high copper level in soil and has the characteristic of selective ion transport (Saberi 1997). Due to the release of oxygen by mangrove roots, an oxidant geochemical microenvironment is created (Silva *et al.* 1990) which helps to oxidise soluble Fe^{2+} and Mn^{2+} to insoluble $Fe(OH)_3$ and MnO_2. After oxidisation, these oxide-hydroxides of Fe and Mn strongly coprecipitate other metals (Barlett 1961). The presence of an 'ion plate' is frequent in salt marsh plants and is responsible for the precipitation of various metals at the root's surface (Otte *et al.* 1987). High copper concentrations in roots may be due to the process involving the release of oxygen by mangrove plant roots.

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