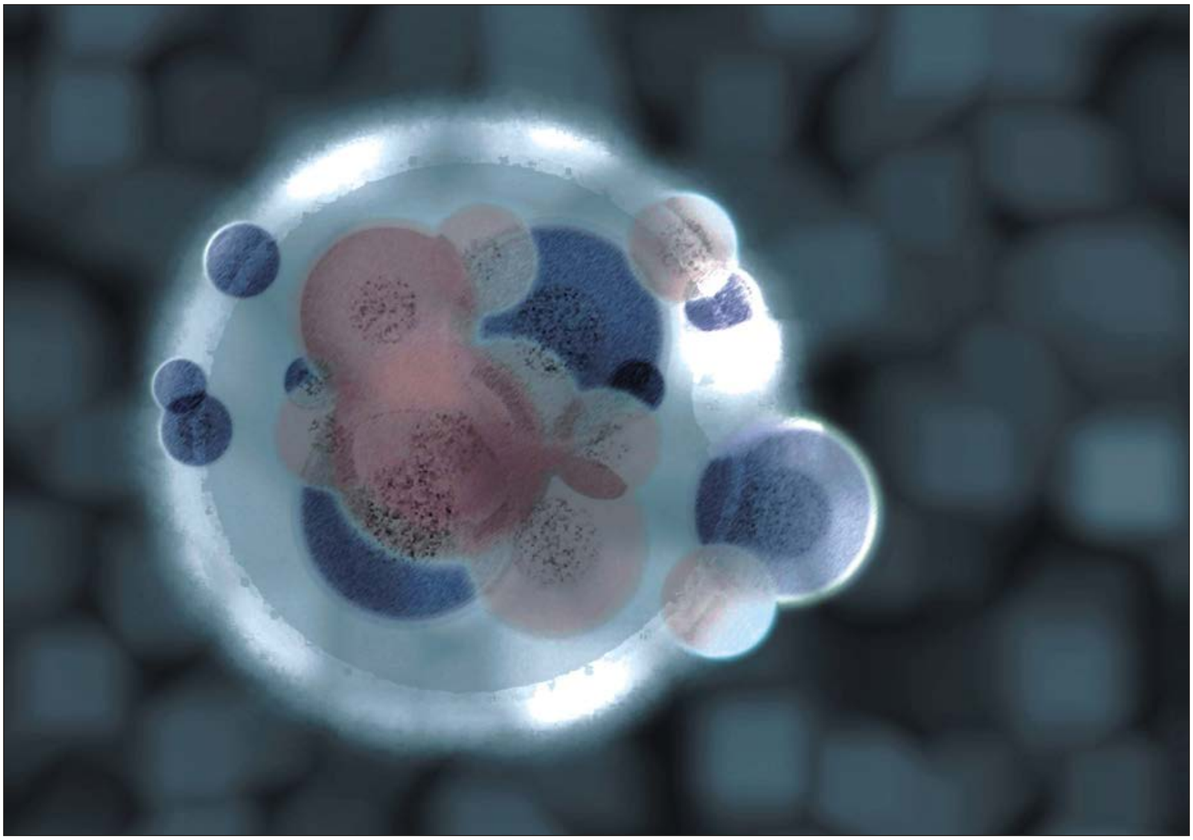

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Heavy Metal Accumulation in a Medicinal Plant *Centella asiatica* from Peninsular Malaysia

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Abstract: The objective of this research was to determine the metal accumulation of *C. asiatica* collected from 13 sampling sites from Peninsular Malaysia and its safety for consumption by the public in Malaysia. *Centella asiatica* plants were collected (or bought) from 13 sampling sites (9 from the wild and 4 from markets) between May and June of 2010. The leaves, stems and roots of *C. asiatica* were determined for Cadmium, Copper, Iron, Nickel, Lead and Zinc. Generally, Iron accumulation was the highest followed by Zinc, Lead, Copper, Nickel and Cadmium. For all metal accumulations, roots showed the highest level, followed by leaves and stems. Plants from sampling sites in Seremban, PPauh and Butterworth were higher in metal accumulation due to nearby activities such as industrial area and highway. When compared to the reference values from Recommended Dietary Allowance, Daily Dietary Intake, Tolerable Upper Intake Level and maximum level intake without detriment to health, all samples from the 13 sampling sites had metal levels which were within or lower than the safety levels or maximum permissible level for human consumption. Therefore, our results showed that *Centella asiatica* was safe to be consumed for all purposes especially for medical treatment of various illnesses.

Key words: *Centella asiatica*, correlation coefficient, dietary value, heavy metals accumulation, safety consume

INTRODUCTION

Centella asiatica (family: Umbelliferae), commonly known as pennywort has been widely used in folk medicine for hundreds of years to treat a wide range of illness (Brinkhaus *et al*., 2000). It is used in different continents by diverse ancient cultures and tribal groups. It is distributed in many Asian countries such as Japan, India, China, Indonesia and Sri Lanka (Brinkhaus *et al*., 2000). The areal parts of the plant are used for medicinal purposes and they can be harvested throughout the year (Zainol *et al*., 2003). This plant species has been used as a wound-healing agent and a constituent of a brain tonic for the mentally challenged (Veerendra and Gupta, 2002). It has also been used traditionally for central nervous system ailments including failing memory, insomnia, depression, stress and epilepsy (Ganachari *et al*., 2004). In South Africa, it is used to treat leprosy, wounds, cancer, fever and syphyllis, while in Europe; the extract has been used for many years to treat wounds (Oyedeji and Afolayan, 2005). In Malaysia, it has been used to aid fast healing of small wounds, chaps and scratches, surgical wounds and so on. Of the entire genus *Centella*, only the *asiatica* species is found in commercial drugs today (Zainol *et al*., 2003). The harvested plants are dried under the sun and later stored in closed containers under cool and dry conditions prior to further commercial processing (Brinkhaus *et al*., 2000). Nowadays, *C. asiatica* is commercially available in the markets and pharmacies in the form of edible products such as *C. asiatica* juice, can drinks, capsule form and cosmetic products such as shampoo, soap and shower foam (Schaneberg *et al*., 2003).

Some of medicinal plants have the ability to accumulate these heavy metals but the rate of uptake differs in each species based on their genetic features (Peris *et al*., 2007); therefore the toxic effects of heavy metals in different plants may also differ significantly (Leon *et al*., 2002). Some plant species are hyper-accumulator where it can accumulate 10-100 times than normal plants (Baker and Whiting, 2002). However, *C. asiatica* as a heavy metal accumulator has not yet been reported in the literature; hence it will be interesting to find out its level of heavy metal accumulation. In medical

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treatment of various illnesses by using herbs should be awared that apart from the pharmacological effect, they shall also take note of the toxic effect of the medication due to the presence of heavy metals and other impurities. Nowadays, there are estimated around 80% of the world population are using medicinal plants (WHO, 2008). For this reason, it is essential to control the levels of contaminants in medicinal raw materials. Foreseeing its potential, it has been chosen as the main interest in this study. Hence, the objective of this research was to determine the metal accumulation of *C. asiatica* collected from 13 sampling sites from Peninsular Malaysia and its safety for consumption by the public in Malaysia.

MATERIALS AND METHODS

Sample collection: Plants from a total of 13 sampling sites were collected between May and June of 2010. The samples collected were of commercial maturity (2-4 months) and the sampling sites included Permatang Pauh (PPauh), Karangan, Kluang, Butterworth, Universiti Putra Malaysia (UPM), Kapar, Seremban, Malacca, Cameron Highland (C. Highland), Bukit Mertajam (BM), Kampung Simpang Renggam (KSR), Kuala Lumpur (KL) and Pontian (Fig. 1). Whole plants were collected from the sampling sites and put into plastic bag (Table 1).

Heavy metal analysis: The harvested plants were separated into three different parts namely shoots, stems and roots. The separated plant tissues were then dried in the oven for 72 h at 60°C to constant dry weights. About 0.5 g of dried plant tissue parts were weighed using an analytical balance. Ten millilitre of concentrated nitric acid (AnalaR grade, BDH 69%) was added to a digestion tube to digest the plant tissues. Three replicates were done for each treatment and each plant part. Then, the digestion tube was placed in hot block digester at 40°C for 1 h and at 140°C for at least 3 h (Yap *et al*., 2007). After that, the digested samples were left to be cooled down and were topped up (diluted) to 40 mL with double de-ionized water. The solution was filtered through Whatman No. 1 filter paper into an acid-washed pill box and stored in a safe place until metal concentrations determination.

All the samples stored in acid-washed pill boxes were analyzed using an air-acetylene flame atomic absorption spectrophotometer Perkin-Elmer™ model AAnalyst 800 (Ahmad and Shuhaimi-Othman, 2010). Blank determination was carried out for calibration of the instrument. Standard solutions for Cd, Cu, Fe, Ni, Pb and Zn were prepared from 1000 ppm stock solution provided by MERCK Titrisol. All data obtained from the ASS were presented in $\mu g\ g^{-1}$ dry weight basis. To avoid possible contamination, all equipment and glassware were first acid-washed in 10% nitric acid solution.

Statistical analysis: The Analysis of Variance (ANOVA), Student-Newman-Keuls (SNK) and Post hoc test were

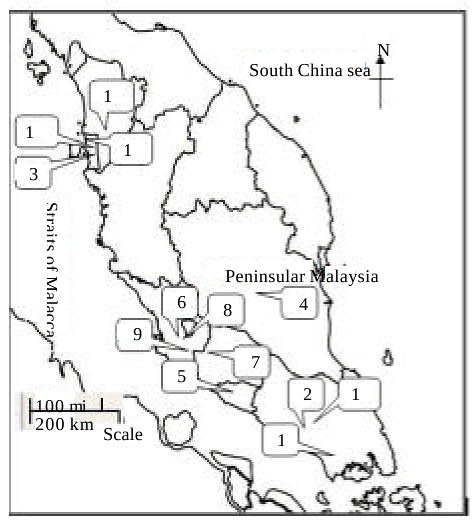

Fig. 1: Map showing the sampling sites for *Centella asiatica* in Peninsular Malaysia

Table 1: Sampling sites, sampling dates and site descriptions of *Centella asiatica* in Peninsular Malaysia

Sampling sites	Sampling dates	Site description
Pontian, Johore	9 May 2010	Near a plant agriculture area
Kampung Simpang Renggam (KSR), Johore	9 May 2010	Near a housing area
Bukit Mertajam (BM) , Penang	15 May 2010	Not clear
Cameron Highland (C. Highland), Pahang	22 May 2010	Not clear
Malacca	23 May 2010	Not clear
Kuala Lumpur (KL), Selangor	23 May 2010	Not clear
Seremban, Sembilan	4 May 2010	Near shop lots and road sides
Kapar, Selangor	5 June 2010	Small scale housing area
Universiti Putra Malaysia (UPM), Selangor	5 June 2010	Near an agriculture are
Butterworth, Penang	12 June 2010	Near an industrial and highway
Kluang, Johore	19 June 2010	Near a paddy fields
Karangan, Kedah	12 June 2010	Near an oil palm plantation
Permatang Pauh (PPauh), Penang	12 June 2010	Near a housing area and highway

KSR, BM, C. Highland and KL are not clear because bought from wet market

Table 2: Overall mean values (Mean±SD, μg g^{-1} dry weight) of heavy metals in different parts of *Centella asiatica* collected from 13 sampling sites from Peninsular Malaysia (N = 13)

Parts	Cd	Cu	Fe	Ni	Pb	Zn
Leaves						
Mean±SD	0.97±0.65	10.25±2.78	431.21±329.97	6.50±3.72	31.72±18.45	175.25±70.84
Min-max	0.32-1.62	7.47-13.03	101.24-761.18	2.78-10.22	13.27-50.17	104.41-246.09
Stems						
Mean±SD	0.50±0.41	7.22±2.76	217.75±168.22	4.7±3.11	24.40±16.81	129.96±26.10
Min-max	0.09-0.91	4.46-9.98	49.53-385.97	1.59-7.81	7.59-41.21	103.86-156.06
Roots						
Mean±SD	1.43±1.01	13.99±3.77	670.31±534.33	7.67±4.18	40.82±22.18	203.75±69.80
Min-max	0.42-2.44	10.22-17.76	135.98-1204.64	3.49-11.85	18.64-63.00	133.95-273.55

applied by using SPSS software version 17.0 for Windows to find the difference between the mean of heavy metal concentrations in the different parts of the plants from different sites and different treatment. Besides that, the STATISTICA version 8 software was also used to determine the correlation coefficient and for hierarchical cluster analysis (Zar, 1996).

RESULTS

The mean values for all the heavy metals (Cd, Cu, Fe, Ni, Pb and Zn) in leaves, stems and roots of *C. asiatica* are presented in Table 2. For all the metals, accumulation in roots were the highest followed by leaves and lastly in stems. Among the six metals, Fe accumulation was highest followed by Zn, Pb, Cu, Ni and Cd in all parts.

From Table 3, the correlation between different parts of the plant agrees with the expected results. For correlation between leaves and stems, Cu (R = 0.96), Cd (R = 0.623), Zn (R = 0.606), Ni (R = 0.872), Pb (R = 0.972) and Fe (R = 0.898) while for correlation between leaves and roots showed Cu (R = 0.827), Cd (R = 0.948), Zn (R = 0.962), Ni (R = 0.974), Pb (R = 0.982) and Fe(R = 0.891). The correlation between stems and roots showed similar results which were Cu (R = 0.741), Zn (R = 0.68), Ni (R = 0.834), Pb (R = 0.958) and Fe (R = 0.791). When compared among these three parts, the correlation between leaves and roots was more significant followed by the correlation between leaves and stems. The correlation between stems and roots was the least. It can be concluded that high accumulation in roots will subsequently cause a higher accumulation in leaves but not necessary causing a higher accumulation in stems.

Figure 2 shows the concentrations of heavy metals in leaves of *C. asiatica*. For Cd, only Butterwoth showed significantly ($p<0.05$) high accumulation while for Cu, Pontian, C. Highland, Seremban and Butterworth showed significant ($p<0.05$) difference when compared to the other sampling sites. For Fe, Butterworth was found to have the highest level among the 13 sites. Plants from the sampling sites in PPauh, Seremban and KL were significantly ($p<0.05$) higher in Ni accumulation. Malacca is the site that showed the highest accumulation of Pb in leaves. For Zn, PPauh and Butterworth showed significant ($p<0.05$) metal accumulation.

Table 3: The correlation coefficients of heavy metals between different parts of *Centella asiatica* based on the mean concentrations ($Log_{10}\mu g/g+1$) of 13 sampling sites

Parameter	Cu	Cd	Zn	Ni	Pb	Fe
Leaves-Stems						
Cu	**0.960**	0.030	0.435	-0.140	0.360	0.325
Cd	0.091	**0.623**	**0.678**	**0.611**	-0.071	0.392
Zn	0.294	0.397	**0.606**	0.418	0.028	0.380
Ni	-0.119	0.391	0.384	**0.872**	-0.108	0.296
Pb	0.347	-0.106	0.084	-0.164	**0.972**	**0.705**
Fe	0.380	0.285	**0.610**	0.236	0.496	**0.898**
Leaves-Roots						
Cu	**0.827**	0.239	0.356	0.065	0.288	0.452
Cd	-0.193	**0.948**	**0.715**	**0.553**	-0.039	**0.685**
Zn	0.094	**0.694**	**0.962**	**0.619**	0.056	0.433
Ni	0.032	0.445	**0.589**	**0.974**	-0.001	0.277
Pb	0.240	-0.080	-0.141	-0.028	**0.982**	0.481
Fe	0.095	**0.638**	0.466	0.200	**0.554**	**0.891**
Stems-Roots						
Cu	**0.741**	0.145	0.326	-0.006	0.333	0.361
Cd	-0.359	0.484	0.414	0.421	-0.125	0.228
Zn	0.174	**0.562**	**0.680**	0.409	0.047	**0.557**
Ni	-0.075	0.461	0.445	**0.834**	-0.138	0.314
Pb	0.336	-0.065	-0.105	-0.082	**0.958**	0.517
Fe	0.222	0.370	0.288	0.296	**0.754**	**0.791**

Values in bold are significant at $p<0.05$ (two-tailed)

Figure 3 shows the heavy metal accumulation in stems. For Ni, Cd and Fe, Seremban showed the highest accumulation when compared to the other 13 sampling sites. Cu accumulation in Seremban and Pontian were significantly higher ($p<0.05$) while Seremban, Butterworth and Pontian were significantly higher ($p<0.05$) for Zn accumulation. For Pb, samples from Malacca were the highest accumulator.

From Fig. 4, PPauh and Butterworth were significant highest ($p<0.05$) for Zn while Seremban and PPauh were significant highest ($p<0.05$) for Ni accumulation in roots. As for BM, it had the highest accumulator for Cu in roots. For Fe and Cd, Butterworth was significant highest ($p<0.05$) in roots. For Pb, Malacca again showed the highest accumulation in roots.

Table 4 shows a comparison of the Daily Dietary Intake (DDI) of *C. asiatica* (3 g dry weight per day) with the recommended metal intake values from several references. It was found that the overall metal

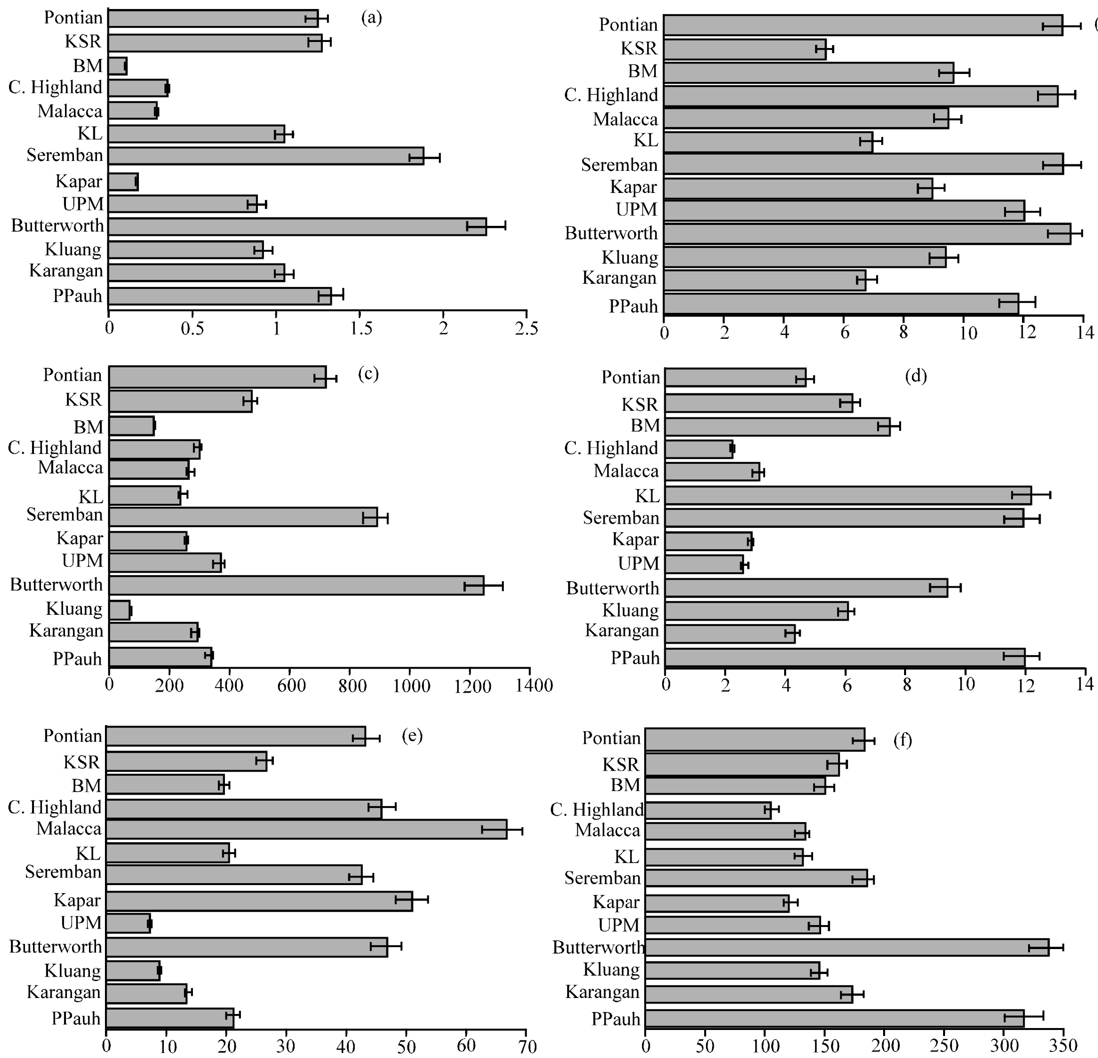

Fig. 2(a-f): Concentrations (Mean±SD, μg g^{-1} dry weight) of heavy metals (a) Cd, (b)Cu, (c) Fe, (d) Ni, (e) Pb and (f) Zn in leaves of *Centella asiatica* collected from 13 sampling sites in Peninsular Malaysia. Note: Sampling site names follow those in Table 1

Table 4: Dietary value of the analyzed *Centella asiatica* daily dietary intake (μg day^{-1}) in comparison with the recommended metal intake values (μg day^{-1})

Element	DDI[a,b,c,d]	RDA[e,f]	TUI[f]	MLI[g]	RDA[h,i]	DDI Leaves [j]	DDI Stems [j]	DDI Roots [j]	DDI 3 parts [j]
Cd	23-120	70	-	18-200	57	3	2	4	9
Cu	1000-4800	900	10000	3200	2500	30	22	42	94
Fe	8100-22700	8000-18000	45000	15000	15000	1294	653	2011	3958
Ni	50-799	-	1000	450	-	20	14	23	57
Pb	34-440	250	-	300	415	95	73	123	291
Zn	6800-22500	8000-11000	40000	17000	15000	525	390	611	1526

DDI: Daily dietary Intake, RDA: Recommended dietary allowance, TUI: Tolerable upper intake level, MLI: Maximum level of intake without detriment to health. [a]Alberti-Fidanza *et al*., 2003, [b]Biego *et al*. (1998), [c]Iyengar, 1998, [d]Jorhem *et al* (1998), [e]WHO (1996), [f]Food and Nutrition Board (2001), [g]Senczuk (1999), [h]Mahan and Escott-Stump (1996), [i]Powell *et al*. (1998) and [j]Based on the consumption of 3 g dry weight per day

accumulation in the whole plant for each of the metals were Fe (3958 μg day^{-1}), Pb (291 μg day^{-1}), Cu (94 μg day^{-1}), Zn (1526 μg day^{-1}), Cd (9 μg day^{-1}) and (57 μg day^{-1}).

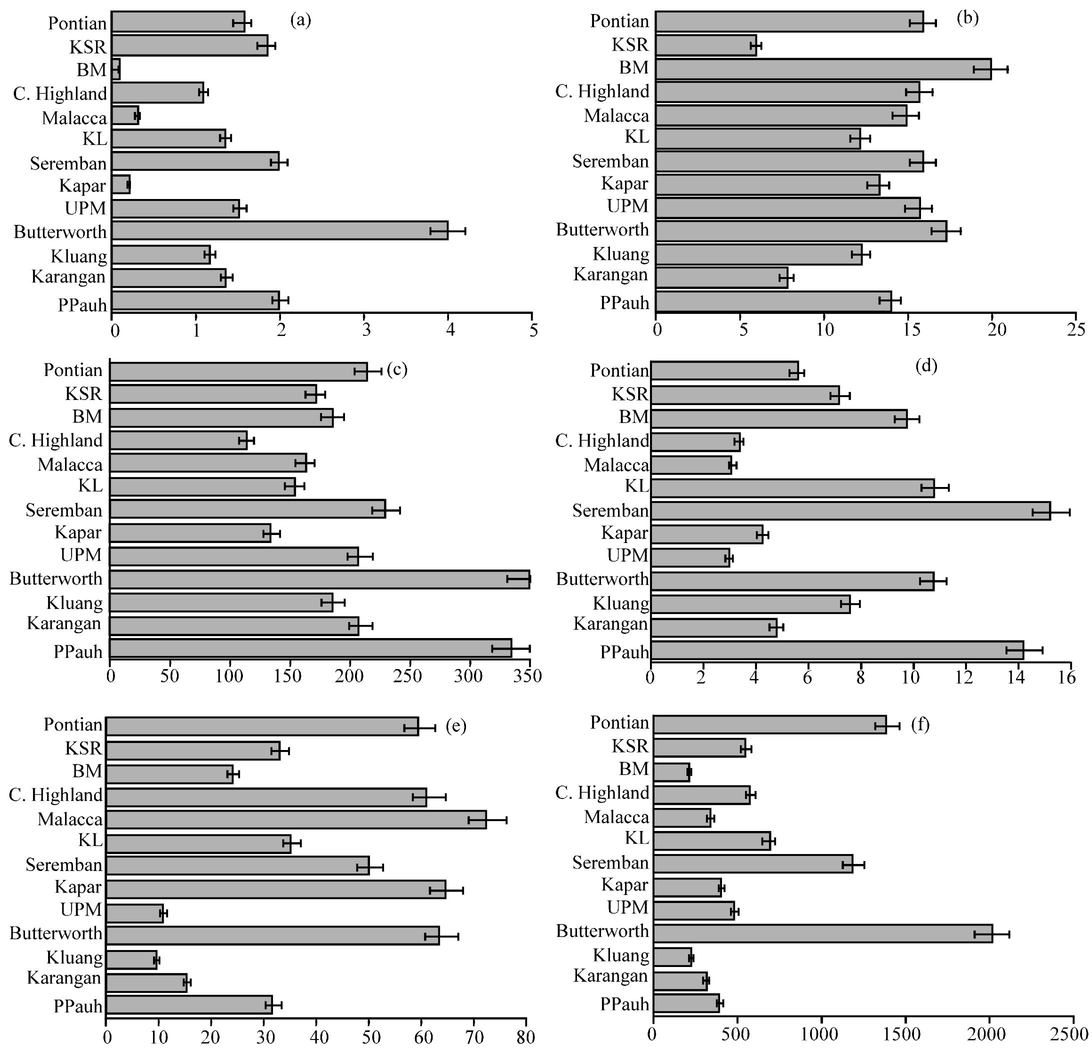

Fig. 3(a-f): Concentrations (Mean±SD, μg g^{-1} dry weight) of heavy metals (a) Cd, (b) Cu, (c) Fe, (d) Ni, (e) Pb and (f) Zn in stems of *Centella asiatica* collected from 13 sampling sites in Peninsular Malaysia. Note: Sampling site names follow those in Table 1

DISCUSSION

Differences of metal accumulation in different parts of *Centella asiatica*: Overall, Fe accumulation was the highest followed by Zn, Pb, Cu, Ni and Cd in leaves, stems and roots. Fe is considered a key metal in energy transformation and is needed for biosynthesis and other life processes of the cells. Hence, the accumulation of Fe was highest among the six metals (Edem *et al*., 2009). This finding was supported by Prasad (2003). After Fe, Zn had a higher level of accumulation when compared to the other metals because Zn plays an essential metabolic role in the plant. It is a component of different enzyme, such as dehydrogenase, proteinases, peptidases and phospohydrolases (Marschner, 1995). In plant, Ni activates several enzymes that are involved in a variety of processes. It can substitute for Zn and Fe as a cofactor in some enzymes (Barker and Pilbeam, 2007). Owing to the relative immobility of Cu and Pb, the plant will accumulate more of Cu and Pb (Zheljazkov *et al*., 2006) even though the Cu requirement is just as a micro-nutrient while Pb is considered as a non-essential metal. Cd which is a non-essential metal for plants showed accumulation due to its high mobility (Vanek *et al*., 2004). Other heavy metals such as Fe, Zn and Cu are essential for the growth of higher plants while Ni is known as beneficial because it is essential for some plant groups (Marschner, 1995). Figure 5 shows the hierarchical cluster analysis for metals

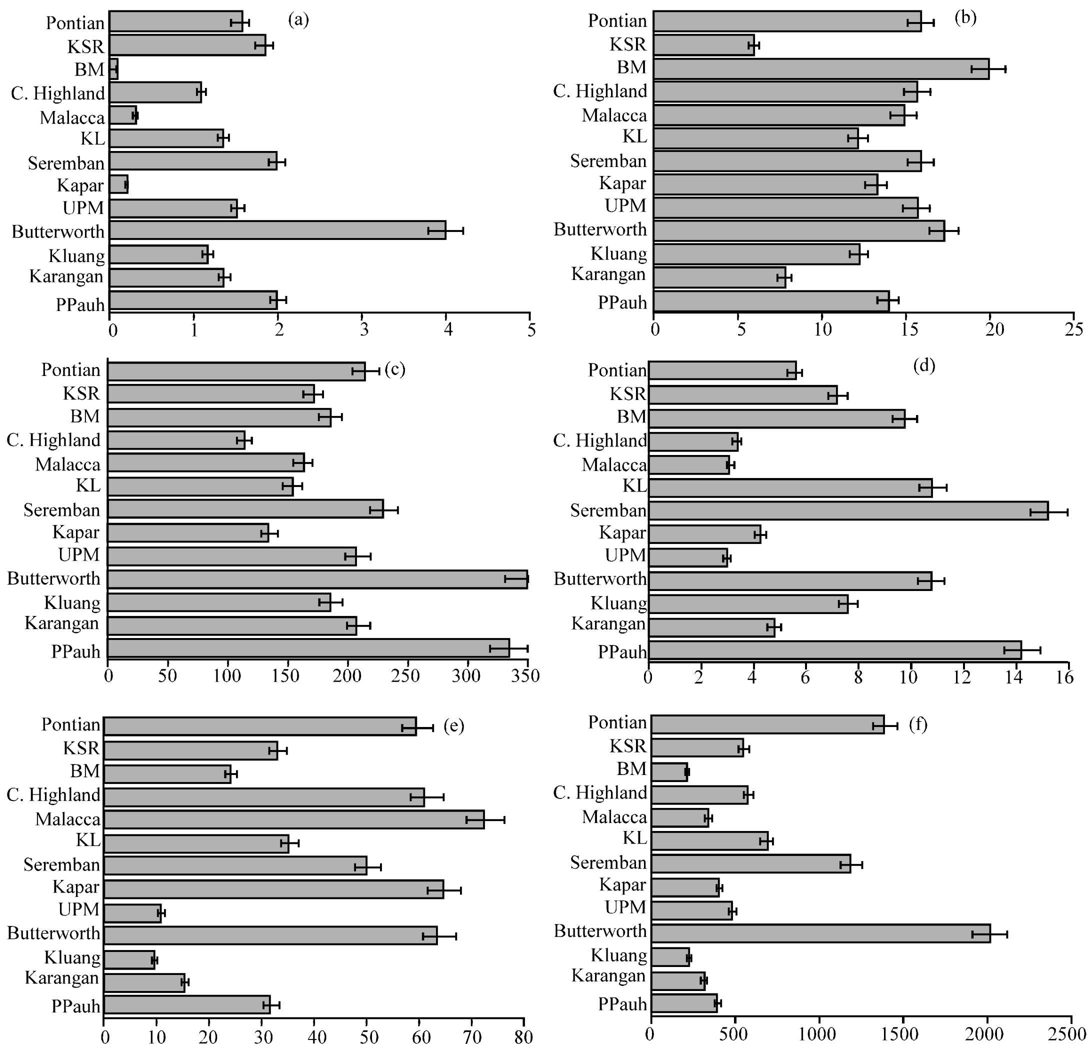

Fig. 4(a-f): Concentrations (Mean±SD, μg g^{-1} dry weight) of heavy metals (a) Cd, (b) Cu, (c) Fe, (d)Ni, (e) Pb and (f) Zn in roots of *Centella asiatica* collected from 13 sampling sites in Peninsular Malaysia. Note: Sampling site names follow those in Table 1

accumulation in leaves, stems and roots. Among the 13 sampling sites, the metals accumulation based on leaves showed that Kapar, Malacca and C. Highland had the least metal levels. For metal accumulation in stems, Kluang showed the least. However in roots; BM showed the lowest metal accumulation. Overall, the sampling sites at Kluang had the lowest metal accumulation in all the parts of the plant.

For all the accumulation of metals, roots showed the highest accumulation followed by the leaves and the least was the stems. This result was supported by those of Soares *et al*. (2001), Singh and Sinha (2005) and Tang *et al*. (2009). This is because the roots are the first organ to be in contact with the metals and roots adhere to the soil all the time. Given this, the exposure of roots towards metals in soil is higher; thus increasing the chances of metal accumulation in roots. Moreover, the large surface area of roots dues to the root hairs elevate the adsorption and absorption of metals and facilitate nutrient uptake (Yap *et al*., 2010). Roots also function as the site of water and nutrient uptake of plants by osmosis, therefore all the metals uptake of the plant must pass through the roots before reaching the other parts (Clemens *et al*., 2002). Hence, the excess metals that are not further transported upwards by the plant will be accumulated in the roots. The high accumulation of heavy metals in root can also be a result of complexation of heavy metals with sulphydryl groups causing lesser

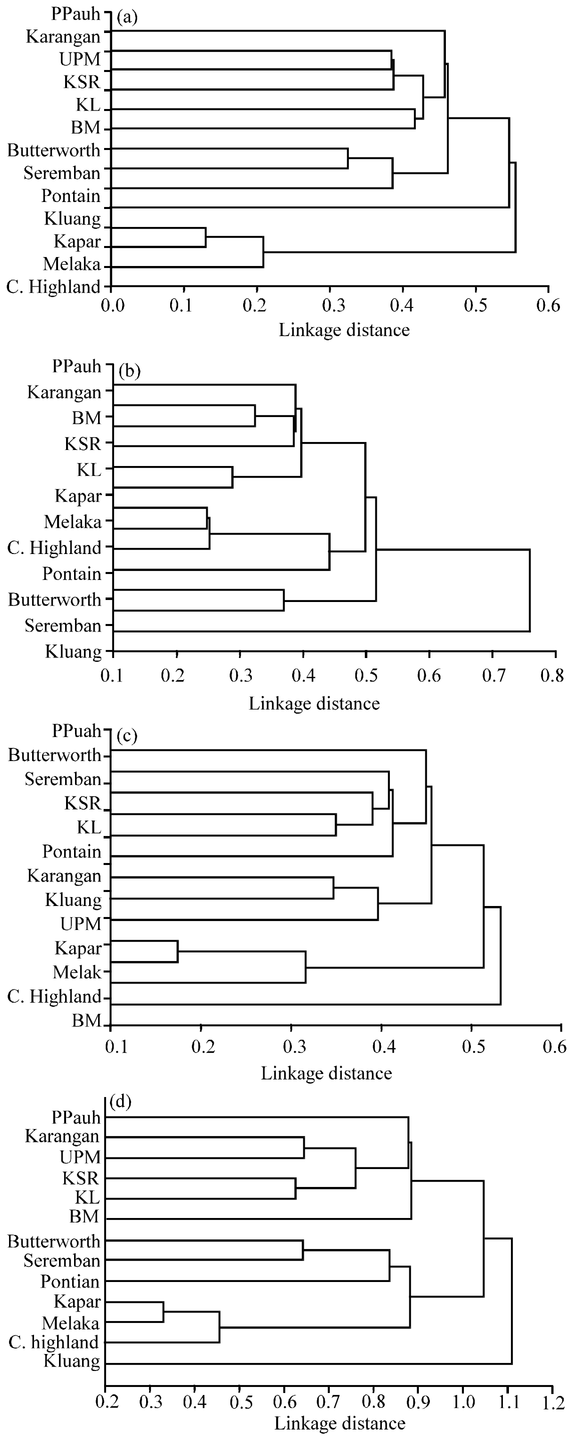

Fig. 5: Hierarchical cluster analysis of *Centella asiatica* based on heavy metal concentrations (Log_{10}mean+1) in (a) leaves, (b) stems,(c) roots and (d) all parts for 13 sampling sites in Peninsular Malaysia. Note: Sampling site names follow those in Table 1

translocation of metals to the shoots (Singh and Sinha, 2005). The translocation processes in plant are regulated by two processes namely root pressure and leaf transpiration (Lasat, 2000). Translocation into leaves enables metals to be reabsorbed from sap into the leaf cells (Lasat, 2000). Overall, the uptake mechanism is selective, plant prefers to acquire a certain ion more than others and this also depends on the structure and properties of the membrane transporter.

The accumulation of metal was the lowest in stems because the stems only function as a channel for the passage of metals to other deposit on parts of the plants such as leaves and flowers. Other than that, it provides support to the plant (Raven *et al*., 1981). Leaves showed the second highest accumulation of metal due to its property as a storage site (Bell and Bryan, 1993). Normally the nutrient uptake of the plant is transported to leaves and stored in it for further metabolic activities. The carbon metabolic pathways in leaves which are known as the photosynthetic carbon reduction cycle and trehalose biosynthesis, have provided insights into the linkage between metabolism and development (Raines and Paul, 2006). The metals were concentrated in the leaves, as the elements Zn and Cu play important roles in photosynthesis (Sawidis *et al*., 1995). The results of this deduction were supported by Street *et al.* (2009) and Rout *et al.* (2001) that roots accumulate more metals than shoots in plants. Based on the results of Arslan *et al.* (2010), the heavy metals content (Cd^{2+}, Cr^{3+}, Cu^{2+}, Fe^{3+}, Ni^{2+}, Pb^{2+} and Zn^{2+}) showed that roots were the highest compared to leaves and stems in *Verbascum bombyciferum.*

Spatial variation of metals accumulation: The accumulation of Cd in both leaves and roots were the highest for plants from Butterworth. This shows the development of Butterworth has caused pollution to the soil of the area resulting in the accumulation of Cd in the plants. The usage of Cd in industries such as smelting and refining of metals can cause the release of Cd into the air. In addition to this, the production of batteries, alloys and plastic will also contribute to the release of Cd into the environment (Forstner and Wittmann, 1981).

In this study, for the accumulation of Cu in leaves, Butterworth showed the highest accumulation followed by Seremban and Pontian. Meanwhile, BM was significant highest in the accumulation of Cu in roots. The accumulation of Cu in the leaves was due to the industrial environment (DOE, 2009) where the plants were grown. Anthropogenic emissions such as from smelters, iron foundries and combustion sources caused the deposition of metal on the leaves. The build ups in the roots were cause by the usage of products containing copper in agricultural areas. These products were normally fertilizers, bactericides, fungicides and algicides; the excessive usage of which would cause it to leach into the ground since according to reports by WHO (2001) 2% of copper was released into the soil in agriculture sites.

The accumulation of Fe not discussed further because in nature, the concentration of Fe in soil was high when compared to other metals and it rarely causes any toxic effects to plants. For Pb, Malacca showed significantly highest accumulation for all the parts of the plant (leaves, stems and roots). This was due to the increase of the traffic density by the booming of the tourism industry causing residues from lead-containing gasoline to be deposited onto the leaves and soil the plantations (Woolf *et al*., 2007). Furthermore, the distance of the plantation from the roadside could also influence the metals accumulation (Sofyan and Murjaya, 1997).

For Ni, the accumulation in leaves was higher for K.L, Seremban and PPauh while for roots the accumulation was higher in Seremban and PPauh. The accumulation in leaves was highest in K.L because of the large numbers of cars in the city causing incomplete combustion of the fuel releasing nickel into the environment (DOE, 2009). This was followed by Seremban and PPauh, both developing industrial areas with factories manufacturing stainless steel items and alloys. Plant species grown with high intensive industrial activities accumulated higher concentrations light industrial activities (Al-Khateeb and Leilah, 2005).

In Butterworth and PPauh, the accumulation of Zn was significantly ($p<0.05$) higher in leaves and roots when compared to the other sites. Butterworth was a well developed industrial area with massive usage of electronics. This has been reported by Seng *et al*. (1987) since the 1980s. The types of industries in operation in this were electronics, textiles, food processing and rubber based industry. Therefore the release of gasses containing Zn due to waste combustion and steel processing was unavoidable (Alkarkhi *et al*., 2009). For PPauh, the accumulation of Zn was high because of the agricultural activities in this area. The use of fungicides and fertilizers containing organo-zinc could have caused the excess to leach into the soil (WHO, 2001).

Metals can be deported into the environment by several means such as air, water and natural deposits, thereby increasing the metal accumulation in the environment (Greger, 2004). Gravel *et al*. (1994) and Januz *et al*. (1994) reported that the plants grown in an industrialized region had higher contents of heavy metals than plants growing in a less industrialized region.

Safety for human consumption: Fe, Cu, Zn and Cd were below the range of the DDI references whereas Pb and Ni were within the range (Biego *et al*., 1998; Iyengar, 1998; Jorhem *et al*., 1998; Alberti-Fidanza *et al*., 2003). Furthermore, for the comparison with the Recommended Dietary Allowance (RDA) all the metals except for Pb were lower than the recommended level (Food and Nutrition Board, 2001). The overall value of Pb in the plant was excessive but this would not negatively impact the well being of human because generally the consumption of these plants were more towards the leaf and stem. Moreover, the result shows the metals accumulation in these two parts does not exceed the safety level. When compared to RDA (Mahan and Escott-Stump, 1996; Powell *et al*., 1998), Tolerable Upper Intake Level (TUI) (Food and Nutrition Board, 2001) and Maximum Level of Intake without Detriment to Health (MLI) all the metals level were within the safety baseline for human consumption (Senczuk, 1999). In addition by using individual parts of the plants for comparisons to the standards, all were found to be lower than the suggested guidelines values.

From a first glance, a relatively large dietary intake of *C. asiatica* is needed to exceed the standard levels of the heavy metals but the results shown are based on the current research and it is not known if this is sufficient to mitigate lifelong dietary exposure to the heavy metals. Thus, values that are considered as being low risk to consumers and values that are close to the current limits should be taken seriously.

CONCLUSION

For all the metal accumulations in *C. asiatica*, roots showed the highest and it was followed by the leaves and the least was the stems. Fe accumulation was the highest followed by Zn, Pb, Cu, Ni and Cd in leaves, stems and roots. The samples sites in Seremban, PPauh and Butterworth showed the highest metal accumulations due to nearby site activities than from plants of the other sites. Even though certain sites showed significant differences in metal accumulations in *C. asiatica,* all of them were within the safety levels for human consumption based on DDI, RDA, TUI and MLI. As a conclusion, these results indicated that *C. asiatica* from Peninsular Malaysia is safe to be consumed. In future research, the extracts or products of *C. asiatica* should be checked for heavy metal levels because there may be contamination during storage, transportation or the manufacturing process beside accumulation of metals from where they were planted. These precautions are indispensable when large amounts of the products are consumed.

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