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Heavy Metal Contamination of Ready-to-use Herbal Remedies in South Eastern Nigeria

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Abstract: The Zn, Cd and Pb content of selected ready-to-use herbal remedies in Southeast Nigeria were determined by Atomic Absorption spectrophotometer (Model 2380 Perkine Elmer Inc., Norwalk, CT, USA) after acid digestion with 5:1:1 HNO_3: $HClO_4$: H_2SO_4. The concentration levels of Pb, Cd were generally high and above the safe limits set by WHO/FAO. Only Tunya B.fil, Virgy virgy worm expeller and Sekin powder had Zn concentrations above international safe limits representing 20% of the tested herbal remedies. The consumers of these herbal products are inadvertently exposed to heavy metal poisoning. It is important that regulators should intensify efforts to minimize human exposure risk.

Key words: Herbal remedies, heavy metals, Nigeria, contamination

INTRODUCTION

The World Health Organization (WHO) estimates that 80% of the world population use herbal medicines as their primary health care intervention (Kroll and Shaw, 2003; Caldas and Machado, 2004). This is prevalent in the developing countries and has been attributed to better cultural acceptability, better compatibility with human body and lesser side effects (Kamboj, 1999). The use of herbal and traditional medicines raises concerns in relation to their safety and there is a wide misconception that 'natural' means 'safe' (WHO, 2002). The contamination of herbal remedies with heavy metals due to soil and atmospheric contamination poses a threat to its quality and safety. Health implications of heavy metal poisoning are abound in many literatures (Trichopoulos, 1997; Harbison, 1998; Smitherman and Harber, 1991). Apart from occupational exposure, heavy metals gain access to human body through diets and inhalation of suspended heavy metal particles. Regulation of heavy metal emission from several industrial sources has not been effective in many parts of the world especially in the developing countries.

The uptake and bioaccumulation of heavy metals in herbs/plants are influenced by a number of factors such as climate, atmospheric depositions, the concentrations of heavy metals in soil, the nature of soil on which the herbs are grown and the degree of maturity of the plant at the time of harvest (Lake *et al*., 1984; Scott *et al*., 1996; Voutsa *et al*., 1996). Elevated levels of heavy metals in plants are reported from the areas having long-term uses of treated or untreated wastewater (Sinha *et al*., 2005; Sharma *et al*., 2006), plants growing along heavy traffic ways (Remon *et al*., 2005, Liu *et al*., 2007) and previous dumpsites (Nwachukwu *et al*., 2010).

Other anthropogenic sources of heavy metals include the addition of manures, sewage sludge, fertilizers and pesticides, which may affect the uptake of heavy metals by modifying the physico-chemical properties of the soil such as pH, organic matter and bioavailability of heavy metals in the soil.

Farm lands near heavy traffic high ways are exposed to atmospheric pollution in the form of metal containing aerosols. These aerosols can be deposited on soil and are absorbed by plants and/or deposited on leaves, barks and fruits. Voutsa *et al*. (1996) have reported high accumulation of Pb, Cr and Cd in leafy vegetables due to atmospheric depositions. In Nigeria, most Trado-medicine practitioners' sell their products along busy traffic urban centres. Herbs, barks and roots used for various ailments are displayed outside their stores thereby exposing them to air-borne heavy metals contamination. Machine blended herbs are also exposed to heavy metal contamination. Table 1 shows some of the Nigerian herbal preparations sold within and around southeastern states. Field surveys have found positive relationships between atmospheric metal deposition and elevated concentrations of heavy metals in plants and top soil (Larsen *et al*., 1992; Sanchez-Camazano *et al*., 1994). Urban activity may significantly contribute to elevated heavy metal loads in atmospheric deposits and consequently contaminate ready-to-use herbal products. This present work was conducted (a) to quantify the levels of Pb, Cd and Zn contamination on ready-to-use herbal products. (b) To determine the heavy metal pollution index of the total heavy metal load at different sampling locations in Southeastern Nigeria.

MATERIALS AND METHODS

The present study was carried out from March 2008 to February 2009 in the urban areas of southeastern Nigeria comprising Aba, Owerri, Onitsha, Abakiliki and Enugu. The climate of the area is tropical with two distinct seasons ie dry and rainy seasons.

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Table 1: Some Nigeria herbal remedies, traditional use and where purchased

Herbal remedies	Traditional use	Where purchased
Man power sexual energy	Low sperm count, diabetes	Owerri
U and D sweet bitter	Typhoid fever, malaria, stomach pain, scratches, weakness of organ	Owerri
Natural power stone	Malaria, hepatitis, internal heat, painful menstruation, rheumatism	Enugu
Dorasine powder	Typhoid fever, malaria, chest pain , stomach ulcer	Onitsha
Virgy-virgy computer worm-expeller	Worm expeller	Onitsha
B-success 28 plant	Antibiotic with aloe vera	Onitsha
H-Nal	Hormonal imbalance	Abakiliki
M-Reg	Hormonal regulation	Abakiliki
Diabor	Diabetes	Enugu
Infecta	Infection	Aba
Firas	Fibroid	Aba
Rinbacin forte	Bacterial and gastro-intestinal disease	Aba
C-candi	*Candida albieans*	Owerri
C-Cystra	Cyst	Owerri
Aloe Vera cure	Malaria fever, fever, typhoid fever, piles	Abakiliki
Omega roots gun sergy	Staphylococcus	Enugu
Tunya B. fil	Watery sperm count, convulsion	Enugu
Almocin powder	Malaria rheumatism	Abakiliki
Koso herbal blended powder	Low sperm count	Aba
Sekin powder	Ulcer pile vomiting	Onitsha

A reconnaissance survey was conducted to evaluate the land use, types of market, traffic flow and consumption of herbal remedies. The cities were divided into 10 grids each of about 3 km^2 and then 4 sampling locations were selected where herbal remedies were sold. These sampling locations were categorized into two groups based on the types of market i.e. wholesale, retailer. The magnitude of urban activities such as industrial, residential and commercial and vehicular density also varied between the sampling locations. The characteristics and relative position of sampling locations are shown in Table 2.

Sampling and pre-treatment: Samples of ready to use herbal remedies were obtained from each of the five southeastern states from three randomly selected different sale outlets. The samples were collected once in a month between March 2008 and February 2009 from selected sample sites (Table 2). Roots and plant parts samples were chopped into small pieces and blended into flour.

Analytical procedure for heavy metal analysis: For Heavy metal analysis of herbal remedies, 1 g of blended sample was taken into a 100 ml acid washed beaker and 15 ml of tri-acid mixture (70% high purity HNO_3, 65% $HClO_4$ and 70% H_2SO_4; 5:1:1) was added (Allen *et al*., 1986). The mixture was then digested at 80°C till transparent solution was achieved. After cooling, the digested samples were filtered using Whatman no. 42 filter paper and the filtrate was diluted to 50 ml with deionised water. Determination of the heavy metals such as Zn, Cd and Pb in the filtrate of herbal remedies was achieved by atomic absorption spectrophotometer (Model 2380 Perkine Elmer Inc., Norwalk, CT, USA). The instrument was calibrated using manually prepared standard solution of respective heavy metals. General purpose reagent cadmium nitrate of a minimum purity of 99% was used in the preparation of the solution used to spike the samples for Cd. An analytical grade of a nitrate salt of Pb and granules of Zn were used in the preparation of solutions used in the spiking of samples for Pb and Zn. Acetylene gas was used as the fuel and air as the support. An oxidizing flame was used in all cases. Table 3 shows different wavelength at which the instrument was set for each metal.

Table 2: Type of market, sampling sites and characteristics of the study area in the south east states

Sample site	Type of market	Land uses
AB1	Wholesale	DPRA, HT, NIA
AB2	Wholesale	CA, HT
AB3	Retailer	SPRA, NIA
AB4	Wholesale	DPRA, NIA
AN1	Retailer	LT, NIA
AN2	Retailer	IIA, HT
AN3	Wholesale	DPRA, HT
AN4	Wholesale	LT, IIA
EN1	Retailer	CA, HT
EN2	Retailer	DPRA, CA, HT
EN3	Wholesale	LT, IIA, SPRA
EN4	Retailer	NIA, HT
IM1	Retailer	DPRA, CA
IM2	Wholesale	HT, CA
IM3	Retailer	NIA, SPRA
IM4	Wholesale	IIA, LT
EB1	Retailer	DPRA, HT,
EB2	Retailer	SPRA, LT
EB3	Wholesale	NIA, LT, CA
EB4	Wholesale	IIA, LT

DPRA = Densely Populated Residential Area. NIA = Near Industrial Area. HT = Heavy Traffic. CA = Commercial Area. SPRA = Sparsely Populated Residential Area. LT = Low Traffic. IIA = in Industrial Area. AB (Abia 1-4), AN (Anambra (1-4), IM (Imo 1-4), EB (Ebony 1-4), EN (Enugu 1-4)

Table 3: Wavelength and detection limits of each heavy metal measured by Atomic Absorption Spectrophotometers (Model 2380 PerkineElmer Inc., Norwalk, CT, USA)

Element	Wavelength (cm)	Detection limit (µg/ml)
Cadmium	288.8	0.0005
Lead	213.3	0.01
Zinc	213.9	0.0008

Heavy Metal Pollution Index (HPI): To compare the total heavy metal content in the herbal remedies at different sampling locations, Heavy Metal Pollution Index (HPI) was calculated using the equation given by Usero *et al.* (1997).

$$HPI = (\mu f_1 \times \mu f_2 \times \mu f_3 \times ..... \times \mu f_n)^{1/n}$$

Where µf is the concentration of n heavy metals in herbal preparation samples.

Statistical analyses: Mean, standard deviation and range were calculated using coupled Microsoft Excel + Analyse-it® v2.2 (2010).

RESULTS

The study revealed that some of the places herbal remedies where sold were industrial, commercial and residential areas with either high or low traffic situations (Table 2). Table 4-8 show the heavy metal content and Heavy Metal Pollution Index (HPI) of herbal remedies from the cities. In Owerri, mean heavy metal content of herbal remedies were; 34.75 µg Zn/g range (23-47 µg/g), 5.3 µg Cd/g range (0.83-6.5 µg/g) and 6.1 µg Pb/g range (2.6-10.3 µg/g) Table 4. C-candi recorded the highest Zn and Pb concentrations while C-cystra had highest Cd concentration. In Abakaliki, M-Reg had highest Zn content of 60 µg/g while H-Nal recorded highest of 10.6 µg Cd/g and 46 µg Pb/g (Table 5). Also Tunya B.fil had highest Zn concentration of 65 µg/g, 9.7 µg Cd/g and 34 µg Pb/g in Enugu (Table 6). In Aba, Infecta recorded highest Zn concentration of 56 µg/g, Rinbacin 7.3 µg Cd/g and Koso herbal 23 µg Pb/g (Table 7). Sekin powder recorded the highest Zn and Pb concentrations of 67 µg/g and 48 µg/g, respectively in Onitsha (Table 8), while Virgy-virgy had 8.4 µg Cd/g concentrations. H-Nal had the highest HPI of 30.3 followed by Virgy-virgy worm expeller of 26.4 and sekin powder; 24 and closely followed by Dorasine powder (23.3). All the herbal remedies sampled except Man power sexual and U and D sweet bitter had Pb concentrations exceeded FAO/WHO safe limits for consumable products. Similarly, Cd concentration exceeded safe limits for all the herbal remedies.

Table 4: Heavy metal content of the herbal remedies obtained from Owerri, Imo state

Herbal remedies[a]	Zn (µg/g)	Cd (µg/g)	Pb (µg/g)	HPI
Man power sexual	23±7.8	1.1±0.03	2.6±0.05	4.0
U and D sweet bitter	32±16.2	0.83±0.5	3.9±0.12	4.6
C-candi	47±12.4	2.9±0.04	10.3±2.5	11.1
C-cysta	37±2.6	6.5±1.3	7.9±0.8	12.3
Range	23-47	0.83-6.5	2.6-10.3	
Mean	34.75	5.3	6.1	
Safe limit[b]	60	0.3	5.0	

[a]Number of sample (n = 10), [b]FAO/WHO standard (2002)

Table 5: Heavy metal content of the herbal remedies obtained from Abakaliki, Ebonyi state

Herbal remedies[a]	Zn (µg/g)	Cd (µg/g)	Pb (µg/g)	HPI
Aloe vera cure	34±5.8	0.89±0.07	45±10.3	11.09
Almoci powder	39±8.9	2.8±0.5	34±3.7	15.40
M-Reg	60±12.6	4.9±1.7	18±4.7	17.40
H-Nal	57±4.6	10.6±1.5	46±16.3	30.30
Range	34-60	0.89-10.6	18-46	
Mean	47.5	4.8	35.7	
Safe limit[b]	60	0.3	5	

[a]Number of sample (n = 10), [b]FAO/WHO standard (2002)

Table 6: Heavy metal content of the herbal remedies obtained from Enugu , Enugu

Herbal remedies[a]	Zn (µg/g)	Cd (µg/g)	Pb (µg/g)	HPI
Tunya B.fil	65±23.3	9.7±1.3	34.7±1.4	28.0
Omega roots gun sergy	54±12.1	1.3±0.06	12.7±2.3	9.6
Diabor	49.5±11.2	1.1±0.03	23±5.3	10.8
Natural powder stone	23±2.3	4.5±0.06	22±4.3	13.1
Range	23-65	1.1-9.7	12.7-34.7	
Mean	47.7	4.2	23.1	
Safe limit[b]	60	0.3	5	

[a]Number of sample (n = 10), [b]FAO/WHO standard (2002)

Table 7: Heavy metal content of the herbal remedies obtained from Aba, Abia state

Herbal remedies[a]	Zn (µg/g)	Cd (µg/g)	Pb (µg/g)	HPI
Infecta	56±4.5	0.97±0.12	10.8±1.6	8.37
Firas	45±6.7	2.3±0.65	12.1±1.7	10.80
Rinbacin	34±4.7	7.3±0.43	18.9±4.6	16.70
Koso herbal powder	21±1.3	1.4±0.04	23.5±1.4	8.84
Range	21-56	0.97-1.4	10.8-23.5	
Mean	39	3.0	16.3	
Safe limit[b]	60	0.3	5.0	

[a]Number of sample (n = 10), [b]FAO/WHO standard (2002)

Table 8: Heavy metal content of the herbal remedies obtained from Onitsha Anambra state

Herbal remedies[a]	Zn (µg/g)	Cd (µg/g)	Pb (µg/g)	HPI
Dorasine powder	55.6±12.3	6.5±0.9	35±13.3	23.3
Virgy virgy worm expeller	65±14.7	8.4±1.1	34±11.3	26.4
B-success 28 plant	45±11.4	1.0±0.07	45±312.3	12.6
Sekin powder	67±23.3	4.3±0.08	48±17.8	24.0
Range	45-67	1.0-8.4	34-48	
Mean	58.2	5.1	40.5	
Safe limit[b]	60	0.3	5	

[a]Number of sample (n = 10), [b]FAO/WHO standard (2002)

DISCUSSION

The study showed that more than 98% of the Herbal Remedies (HR) analyzed did not comply with international safe limits for herbal products. Similar studies on herbal medicines are well documented in literature (Obi *et al.*, 2006; Saper *et al.*, 2004; Smolinske, 2005; Ang, 2008).

Several possibilities may be responsible for the presence of heavy metals in herbal remedies. Firstly, the presence of heavy metals may be the result of accidental contamination during the manufacturing process, such as grinding weights, lead-releasing containers or other manufacturing utensils (Koh and Woo, 2000; Yee *et al*., 2005). Secondly, these medicinal plants may have grown in seriously polluted soil (Schilcher, 1983; Kabelitz, 1998; McLaughlin *et al*., 1999), water or air (McLaughlin *et al*., 1999). It was reported that the contamination of plants with lead depends heavily on several factors such as traffic densities, distance from the road (Bosque *et al*., 1990) and proximity to roadways or metal mining and smelting operations (Pip, 1991). High levels of toxic metals can be found when fertilizers, organic mercury, or lead-based pesticides or fumigants contaminate irrigation water (Abou-Arab *et al*., 1999; Yee *et al*., 2005). Thirdly, some of the combined medicinal herbal remedies may also contain animal and mineral, or any other excipients may also be contaminated with heavy metals (Chuang *et al*., 2000). Poisonings associated with the presence of heavy toxic metals in medicinal plants were reported in Asia, Europe and the United States (Dunbabin *et al*., 1992).

The Food and Agriculture Organization/World Health Organization (FAO/WHO) Joint Expert Committee on Food Additives (JECFA) in their 59th meeting set the provisional tolerable intake for Cd, Pb and Zn at 0.3 µg/g, 5 µg/g and 60 µg/g respectively. In this study, the lowest Cd, Pb and Zn concentrations were 0.83, 2.6 and 21 µg/g, respectively and the highest Cd, Pb and Zn concentrations were 6.5, 9.7 and 4.8 µg/g, all HR exceeded safe standards for heavy metal analyzed except Zn which did not fall within safe limits especially for Tunya B.fil (65 µg/g), Virgy-virgy (65 µg/g), Sekin powder (67 µg/g).

The manifestations of cadmium nephrotoxicity, aminoaciduria, glycosuria and tubular necrosis have been detected at renal cadmium concentration of less than 50 µg/g tissue. The effect of cadmium on the kidney takes the form of renal tubular dysfunction and subsequent pathological changes. However, cadmium-linked kidney toxicity occurred in higher than expected frequencies in human populations whose intakes were well within the current permissible limits, raising doubts that the current permissible limits is sufficient to protect the general population (Satarug and Moore, 2004).

Lead is found everywhere and is a toxicant (Konat and Clausen, 1974). Lead poisoning is an insidioural disease that can result in developmental delays, behavioral disorder and irreversible brain damage (Hanchette, 2007). The major signs and symptoms of lead poisoning are pallor, gingivial lead line, gastrointestinal disorder, anaemia, renal and neurological symptoms (peripheral neuropathy, ataxia and memory) (Ernst and Coon, 2001). The target organs for lead are kidney and nervous system. Decrement in kidney function and abnormalities of nervous system function should prompt, in the absence of more obvious profound causes, suspicions about the use of alternative therapies.

The Food and Agricultural Organization/World Health Organization (1993) has established a 'Provisional Tolerable Weekly Intake' (PTWI) of 25 µg lead/kg body weight for humans. In Canada, the dietary intake of lead has been reported to be 168 µg/week (Anon, 1995). The mean blood lead level intake of adults is in the range of 20-514 µg/day (NRC, 1980). Results obtained from the study showed higher lead levels from the HR than the PTWI established limits. This finding portrays danger to the users of HR in Nigeria.

From the study, 20% of the tested HR gave Zn concentration in excess of 60 µg/g. Few instances of acute zinc poisoning have been reported; its manifestations include nausea, vomiting, diarrhoea, fever and lethargy. Long-term exposure to high zinc intakes substantially in excess of requirements has been shown to result in interference with the metabolism of other trace elements (Fischer *et al*., 1984) Zinc is an element find virtually in every cell in the human body and plays important role in growth and has a recognized action on more than 300 enzymes by participating in their structure or in their catalytic and regulatory action.

One major problem with traditional remedies is lack of standardization. Consistency in composition and biologic activity are essential requirements for safe and effective use of therapeutic agents. However, botanical preparations rarely meet this standard, because of problems in identifying plants, genetic variability, variable growing conditions, differences in harvesting procedures and processing of extracts and above all, the lack of information about active pharmacologic principles (Marcus and Grollman, 2002). In most developing countries, there are hardly any legislation controlling the safety and efficacy in the manufacture/preparation of herbal remedies nor the submission or documentation of products testing, quality control or approval before these products enter the market. Most developing nations have national regulation on herbal medicines but legislative control of medicinal plants has not evolved around a structured model like in most countries (WHO, 2002). Only recently National Agency of Food and Drug Administration and Control (NAFDAC) in Nigeria has made efforts in this direction. The therapeutic/toxic components of plants vary depending on part of the plant used, stage of ripeness, geographic area where the plant is grown, and storage conditions. This implies that significant variation in herbal medicine should be expected. This is worsened in developing countries by lack of standardization resulting from the crude methods of preparing herbal remedies.

Conclusion: The study showed that herbal remedies where sold in industrial, commercial and residential areas with either high or low traffic situations. More than 98% of the Herbal Remedies (HR) analyzed did not comply with international safe limits for herbal products. Except Zn element that had concentrations below the safe international limits in some herbal remedies, all other heavy metals concentration well exceeded safe limits. It is evident that users of these herbal products are inadvertently exposed to heavy metal poisoning and very important that regulators should intensify efforts to minimize human exposure risk.

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