Journal of Environmental Science and Public Health
doi: 10.26502/jesph.96120053

Volume 3, Issue 2 Research Article

Unwholesome Herbal Medicines Marketed in Enugu Metropolis, Enugu State, South Eastern Nigeria: Public Health Implications

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Received: 18 December 2018; **Accepted:** 31 December 2018; **Published:** 24 January 2019

Abstract

Background: The public health implications associated with consumption of unwholesome herbal medicines, food substances and water are well documented. This study brought forth the case of heavy metal and microbial contamination as well as phyto-chemical composition of some herbal medicines marketed in Enugu Metropolis, Enugu State, Nigeria.

Methods: Ten herbal medicines were randomly purchased from herbal medicine vendors in the streets and markets in Enugu, Nigeria. The samples were labeled A-J, and were in triplicates. The heavy metal contents (Pb, Cu, Cd, Zn, Ni, Cr, Fe, Hg, As, Se) were determined using Atomic Absorption Spectrophotometer. The bacterial load was determined by the agar dilution technique and the phyto-chemical constituents of the medicines were determined following standard methods.

Results: Lead and cadmium were present in all the medicine samples at concentrations significantly higher ($p<0.05$) than the recommended standards of American Herbal Products Association, AHPA/WHO standards. The Cr, Fe, and Ni concentrations in 70%, 50% and 40% of the samples respectively, were significantly higher ($p<0.05$) than the AHPA/WHO standards. Bacteria and fungi were present in the herbal medicines in varying concentrations. The phyto-chemical constituents of the herbal medicines were carbohydrates, alkaloids, tannins, flavonoids, saponnins, steroids, terpernoids, proteins, and glycosides in varying quantitative compositions across the herbal medicines.

Interpretation: The phyto-constituents identified have medicinal values to justify for the ethno-medicinal uses claimed by the manufacturers. However, the heavy metals and microbial contaminants of all the medicines pose serious public health challenges and require the attention of the national drug regulatory authorities.

Keywords: Herbal medicine; Public health; Enugu state

1. Introduction

In Nigeria, the practice of herbal medicine is an age long profession cutting across ethnic and religious boundaries [1, 2] and recently, the practice has assumed a dramatic dimension as a result of paradigm shift from orthodox to alternative medicines. This could be traceable to recent awareness of the phyto-potency of indigenous plants in the management of a cock-tail of diseases and the high economic burden associated with orthodox medical practice. Besides, many individuals in the remote communities have little or no access to orthodox medical services partly because of poverty, illiteracy or complete absence of health facilities within their location. African scientists, herbalists and individuals have awoken to the reality of the numerous ethno-medical advantages of many medicinal plants in our environment and have beamed their research light to local plants for the management of tropical diseases. These have necessitated the proliferation of herbal medicines which are hawked around in urban and rural communities with claims of efficacy against many disease-causing germs and functional diseases. Although many of these claims are yet to be authenticated by appropriate scientific approaches, one outstanding problem is the purity of these medicines in terms of metal and microbial contamination. Recent trends in urbanization and industrialization have contributed to anthropogenic-induced environmental pollution, with the effects that different plant parts from where herbal medicines are made are exposed to environmental pollutants [3, 4]. Previous studies on herbal medicines and plants in some parts of Nigeria showed that plants from where herbal medicines are produced are contaminated by heavy metals [4, 5]. Secondly, heavy metal contamination of plants occur from soil by the processes of phytoextraction, phytoaccumulation, translocation and from the air, dust particles through deposition on the foliar structure [6], especially when these plants are exposed to anthropogenic pollutants of domestic and industrial origins. Thirdly, the phyto-chemical compositions of the herbal medicines are lacking in the labels placed on the medicine containers, raising doubts on the assumed claims of phyto-potency of the medicines. Bearing in mind that dying from disease is sometimes unavoidable but dying from a medicine is unacceptable [7], the current study seeks to evaluate the heavy metal and microbial contamination as well as the phyto-chemical composition of the herbal medicines with the aim of ascertaining the wholesomeness of the medicines for continuous use.

2. Methods

2.1 Study area

Enugu Metropolis is the administrative capital of Enugu State, South-Eastern Nigeria. It has the population of 722,000 [8] and density estimate of 6,400/km^2. The study area is located between 6° 21′N and 6° 30′N and 7° 26′E and 7° 30′E (Figure 1). It has a tropical humid climate. Mean annual rainfall ranges from 1600 mm and 2500 mm with three to four months of dry season, the dry season months having 29 mm of rainfall [9]. Mean monthly temperature ranges between 27°C and 29°C.

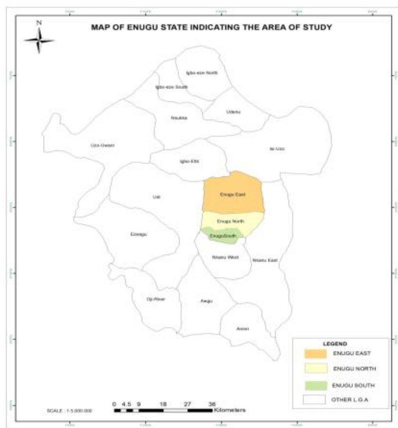

Figure 1: Map of Enugu State indicating the area of study.

In Chima et al. [10] vegetation type of the study area is rainforest savanna. The main river system (Nyaba, Ekulu, Idaw, Aria, Ogboete and Asata) that drains the city originates westward from the base of the Udi escarpment and flows eastwards into the cross river. Asata River, a third order stream is a tributary of the Ekulu River and has an area of about 40 sqkm. The Asata river basin falls within the large Abonie basin. The Ekulu River is the largest body of water in Enugu urban, and its reservoir contributes to part of the city's domestic water supply.

2.2 Collection of herbal medicines

Ten herbal medicines from different producers, marketed in Enugu metropolis were randomly purchased from herbal vendors in the streets and markets. The samples were labeled A-J and were in triplicates. The herbal medicines used in this study were chosen only if they were listed by the national drug regulatory body, National Agency for Food and Drug Administration and Control (NAFDAC). The medicines were collected from three different sampling locations; Ogboete market in Enugu East, Garki market in Enugu South and Obiagu Road in Enugu Urban Local Government Areas. Enugu metropolis having once housed the capital of the old Eastern region, serves as a home state for Igbos at home and in Diaspora. Consequently, the Ogboete and Garki markets in Enugu attract a wide variety of local and foreign products, including herbal medicines. The streets of Enugu are also inundated with herbal medicines produced by herbalists from different states in the South Eastern geopolitical zone. Different types of medicines preparations ranging from solid, semi-solid, and liquid are marketed in the Enugu Urban. This study focused on liquid preparations bottled in plastic containers. Some of the herbal medicines on display at Ogbete main market are shown in Figure 2.

Figure 2: Herbal medicines on display at Ogboete Main Market Enugu.

2.3 Heavy metal analysis

The heavy metal contents were determined using Atomic Absorption Spectrophotometer (AAS). The reagents used (HNO_3 and HCL) were of analytical grade, a product of British Drug House (BDH). Double-distilled-deionized water was used in the preparation of all the reagents. Stock standard solutions of each metal containing 1000 ppm were prepared and calibration standards were obtained by appropriate dilution of the stock solution with deionized water. Each herbal medicine was digested with HNO_3 and HCL in the ratio of 1:3 in a muffle furnace at 650°C and the digest was extracted with deionized water. The AAS was switched on and allowed 40 minutes to equilibrate, after which the hollow cathode lamp for each metal of interest was fitted and aligned properly with the in-built Deuterium lamp. The gas was opened and the hoses checked to ensure that there was no leakage. The "set up optics" button was clicked for the optical properties to adjust to suit the selected metal. The "Optimise" button and "Auto zero" button were clicked. The sequence pull down menu was checked to ensure that the sample identities were correctly entered. The calibration page was clicked to ensure that the right concentrations were entered and extract of each medicine was aspirated into the AAS.

3. Microbial Analysis

3.1 Bacteria isolates

Freshly prepared well dried MacConkey Agar and Blood Agar were incubated with standard wire loop and streaked well. Samples were thoroughly mixed prior to culturing. Culture plates were incubated at 27°C aerobically for 24 hrs and examined. Suspect colonies in pure cultures were counted and multiplied with the volume of the loop according to standard microbiological methods. Each organism was then identified by Gram characteristic, indole, methyl red, vogues prausker and other standard methods.

3.2 Fungi isolates

Tubes of sterile potato dextrose agar and Sabouraud dextrose agar were inoculated with the samples each and incubated at room temperature (28°C) for up to 21 days. They were examined at intervals and significant growth were identified by their pigmentation if any, elevation, texture, and also microscopically after teasing out in lacto phenol cotton blue. They were superimposed with cover slips and examined under × 10 and × 40 objectives and reported.

3.3 Phyto-chemical analysis

Qualitative and quantitative phytochemical analyses were done by standard method as reported in Harbourne [11], Trease and Evans [12].

3.4 Statistical analysis

SPSS Version 20 was used to analyze data generated and some of the results presented in means. Statistical values were considered significant at $p<0.05$.

4. Results

The results of the heavy metal concentrations of the medicines are presented in Table 1. The values were compared with the recommended standards of American Herbal Products Association (AHPA) in collaboration with World Health Organization [11].

Heavy metal load (mg/l)										
Herbal	**Cu**	**Pb**	**Cd**	**Zn**	**Ni**	**Cr**	**Fe**	**Hg**	**As**	**Se**
WHO Standard Limit	1.000	0.005	0.005	5.000	0.002	0.050	0.030	0.002	0.010	0.050
A	<0.001+	0.019*	0.116*	0.027+	<0.001+	0.033	0.386*	<0.001	<0.001+	<0.001+
B	<0.001+	0.063*	0.201*	0.009+	<0.001+	0.149*	0.042	<0.001	<0.001+	<0.001+
C	<0.001+	0.166*	0.153*	<0.001+	<0.001+	0.424*	0.048	<0.001	<0.001+	<0.001+
D	<0.001+	0.088*	0.003+	<0.001+	<0.001+	0.186*	0.053	<0.001	<0.001+	<0.001+
E	<0.001+	<0.001+	0.215*	0.039+	<0.001+	0.052	0.458*	<0.001	<0.001+	<0.001+
F	<0.001+	0.074*	0.455*	<0.001+	0.107*	0.161*	0.055	<0.001	<0.001+	<0.001+
G	<0.001+	0.162*	0.273*	0.019+	0.014*	<0.001+	0.058	<0.001	<0.001+	<0.001+
H	<0.001+	0.223*	0.285*	0.049+	0.090*	0.453*	0.153*	<0.001	<0.001+	<0.001+
I	<0.001+	0.053*	0.472*	0.055+	0.780*	0.122*	1.151*	<0.001	<0.001+	<0.001+
J	<0.001+	0.551*	0.056*	0.032+	<0.001+	1.102*	0.477*	<0.001	<0.001+	<0.001+

*Significantly higher ($p<0.05$) than the WHO standard limit; +Significantly lower ($p<0.05$) than the WHO standard limit.

Table 1: Heavy metal analysis of ten herbal drugs marketed in Enugu Metropolis.

Figure 3 represents the proportion of the drugs significantly contaminated by various heavy metals. Pb and Cd gave the highest contaminations (90% each), followed by Cr (70%) and Fe (50%).

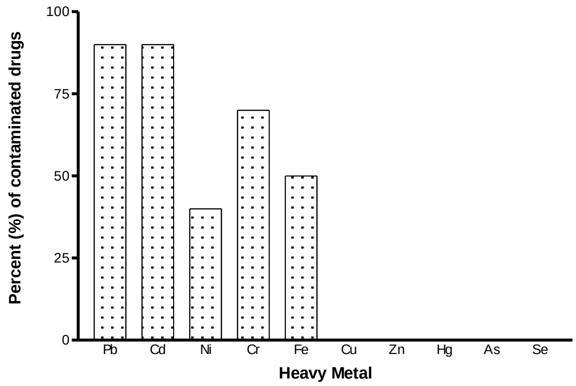

Figure 3: Percentage of medicines with significant metal contamination.

The microbial loads of the samples are shown in Table 2. The bacterial and fungal contaminations were recorded in coliform forming units (cfu), (A). *E. coli*, 10 [1]; (B). *P. aeruginosa*, 10 [2], fungi, *C. tropicalis*; (C). *E. coli*, 10 [3], fungi, *A. flavus*; (D). *C. freundii*, 10 [3]; (E). *P. aeruginosa* and *S. aureus*, 10 [1]; (F). *S. epidermidis*, 10 [1], fungi, *Saccharomyces species*; (G). *S. aureus*, 10 [4]; (H). *P. aeruginosa*, 10 [1], fungi, *A. flavus* and *Rhizopus species*; (I). *P. aeruginosa*, 10 [5], fungi, *C. albicans*, and (J). fungi, *Saccharomyces specie*.

S/N	Sample	Bacterial Isolates	Bio-load	Fungi Isolates	Bio-load
1	A	Escherichia coli	10	No growth	-
2	B	Pseudomonas aeruginosa	102	Candida tropicalis	-
3	C	Escherichia coli	103	Aspergillus flavus	-
4	D	Citrobacter freundii	103	No growth	-
5	E	Pseudomonas aeruginosa and Staphylococcus aureus	10	No growth	-
6	F	Staphylococcus epidermidis	10	Saccharomyces species	-
7	G	Staphylococcus aureus	104	No growth	-
8	H	Pseudomonas aeruginosa	10	Aspergillus flavus and Rhizopus species	-
9	I	Pseudomonas aeruginosa	105	Candida albicans	-
10	J	No growth	-	Saccharomyces species	-

Table 2: Microbial load (cfu) of the ten (10) herbal medicines marketed in Enugu metropolis.

The phyto-chemical constituents identified are presented in Table 3. They were carbohydrates, reducing sugar and alkaloids in all the samples; tannins, flavonoids, saponnins, steroids, terpernoids and acidic pH in 90% of the samples; proteins in 60% of the samples; and glycosides in 50% of the samples.

S/N	Parameters	Sample									
		A	B	C	D	E	F	G	H	I	J
1	Tannins	+	++	++	++	++	++	+++	++	+++	+
2	Resin	-	-	-	-	-	-	-	-	-	-
3	Proteins	+	++	-	+	-	++	-	+	++	-
4	Flavonoids	+	+	+	+	+	+	+	+	+	-
5	Acid	+	+	-	+	+	+	+	+	+	+
6	carbohydrates	+	++	++	++	++	++	++	+	+	++
7	Reducing sugars	+++	+++	+++	+++	+++	+++	+++	+++	+++	+++
8	Saponnins	++	+	-	+	+	++	++	++	++	++
9	Alkaloids	+	+	+	+	+	+	+	+	+	+
10	Glycosides	-	+	-	-	-	+	+	-	+	+
11	Steroids	+	+	+	-	+	+	+	+	+	+
12	Terpernoids	+	+	+	+	+	+	+	+	-	++

+=slightly present; ++=moderately present; +++=abundantly present; -=absent.

Table 3: Qualitative phyto-chemical composition of ten (10) herbal medicines marketed in Enugu metropolis.

Quantitative phyto-chemical analysis in Table 4 showed the composition of the identified phyto-constituents in varying concentrations. Tannins ranged from 1.67% in samples A and J to 22.50% in sample I with an average of 10.17%. Proteins ranged from 1.86% in sample A to 6.15% in sample B with an average of (4.04%). Flavonoids ranged from 0.10% in samples D and I to 0.50% in sample F with an average of (0.25%). The pH ranged from 4.4% in sample F to 7.1% in sample C with an average of (5.48%). Carbohydrates ranged from 2.90% in sample A to 7.12% in sample F with an average of (5.04%). Reducing sugar ranged from 11.80% in sample E to 15.80% in sample I with an average of (14.0%). Saponnins ranged from 1.36% in sample E to 4.05% in sample I with an average of (2.53%). Alkaloids ranged from 0.1% in samples D, F and G to 0.4% in sample A with an average of (0.21%). Glycoside ranged from 0.45% in sample I to 0.70% in sample J with an average of (0.55%). Steroids ranged from 0.25% in sample C to 0.66% in J with an average of (0.47%). Terpernoids ranged from 0.87% in H to 2.56% in E with an average of (1.48%).

4.1 Interpretation

The study evaluated the heavy metal and microbial contamination of herbal medicines marketed in Enugu Urban. The phyto-chemical compositions of the herbs were also analyzed. The Pb and Cd were present in all the medicine samples at concentrations significantly higher ($p<0.05$) than the recommended standards of American Herbal Products Association (AHPA)/WHO standards) [13]. The Cr, Fe, and Ni concentrations in 70%, 50% and 40% of the samples respectively, were significantly higher ($p<0.05$) than the AHPA/WHO standards. Bacteria and fungi were present in the herbal medicines in varying concentrations. The phyto-chemical constituents of the herbal medicines identified were carbohydrates, alkaloids, tannins, flavonoids, saponnins, steroids, terpernoids, proteins, and glycosides in varying quantitative compositions across the herbal medicines.

The significantly high concentrations of Pb and Cd have serious health implications as they have been associated with hepatic and renal problems. This observation is in line with previous studies by Onyemelukwe et al. [14] where evidences of necrosized liver and atrophied kidney were observed in albino rats exposed to herbal medicines. Heavy metal contaminations of the drugs may be attributed to heavy metal content of plants from where they were produced. Nwachukwu et al. [15] assessed heavy metal pollution in soil and their implication within and around mechanic villages and Ogbonna et al. [6, 16] observed significant Pb contamination of road side trees in Enugu Urban and heavy metal contamination of edible plants within the vicinity of automobile services and repair station in Okigwe, Imo State, Nigeria. Although lead-zinc induces physiological, biochemical and behavioral disturbances in humans, exposure to this xenobiotic through food chain is unavoidable because of its accumulation in the environment and use in industrial applications [17, 18]. Being present in the contaminated water, air, food and herbs, [19] lead is mostly absorbed by the lungs and gastrointestinal tract. Because lead also inhibits ferrochelatase, it impairs the chain reaction that leads to the formation of haem and this impairment results in anemia and the accumulation of delta-aminolevulinic acid (ALA) and zinc protoporphyrin (ZPP) in erythrocytes [20, 21] Furthermore, lead interacts with some essential metals [17]. One of them is selenium (Se), which plays an important role as an antioxidant [22]. Se is a cofactor of glutathione peroxidase, decreases the amount of lipid peroxidation and protects DNA, RNA and proteins from oxidative damage. Additionally, Se forms inactive selenium-lead ions in the body [23]. Some other metals identified in herbal medicines such as Zn, Fe, Cu, Cd, As, Hg and Ni, are also implicated as causative factors in the etiology of many diseases, including disorders of the hematopoietic system.

The presence of microorganisms (bacteria and fungi) in the herbal medicine samples contradicts some claims on the labels. *E.coli, S.aureus, P.aeruginosa* are enteric organisms associated with gastroenteritis while some fungi spp are associated with candidiasis. Microbial contamination may be as a result of poor processing and packaging procedures or through contaminated water. Majority of the herbal practitioners are ill-equipped with hygiene education and operate in environment of poor water and sanitation quality. It is surprising that medicines credited to have activities against microorganisms would have such microbial contaminants as recorded. This calls for further investigations as to whether the organisms identified were new strains developed from the effects of the active constituents of the herbal medicines.

Physical examination of the medicine labels revealed that plant names were listed as the common active ingredients. These plants have previously been reported to have activities against common tropical diseases [1, 2, 24-29]. The manufacturers also claimed on the labels that the drugs have activities against various diseases. These claims may be justified by the phyto-chemical constituents identified. For instance, Akerele et al. [27] reported that the presence of flavonoids, tannins, saponins and alkaloid were responsible for many activities of N.laevis against gastroenteritis and other bacteria infections. Anti-malaria, anti-asthmatic, anti-cancer, cholinomimetric, anti-lipidemic and anti-hyperglycemic activities are also associated with the secondary metabolites of the above plant [30-35]. Pharmaceutical important alkaloids include quinine used in malaria endemic zones to reduce mortality due to malaria caused by Plasmodium falciparum, hyoscine and hyoscamine that are useful in post-operative procedures, and nicotine as well as cocaine whose addictive property is of public health importance [36]. Many of these alleged effects of flavonoids are reported to be linked to their strong antioxidants, free radical scavenging and metal chelating properties [37]. The coexistence of heavy metals and flavonoids in the herbal medicines will attenuate the anti-oxidative potentials of the flavonoids and hence, mask the metal chelating properties of the herbal medicines against metal-induced cellular peroxidation.

5. Conclusion

Given the above findings, the medicines have potentials for phyto-therapeutic management of tropical diseases as indicated on the labels. However, the extraneous contaminants such as heavy metals and microbial particles may pose public health problems. Further studies on removal of metal and microbial contaminants of herbal medicines using natural adsorbents are important steps towards improvement of the herbal medicines for better ethno-medicinal uses in Nigeria.

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Citation: Elvis N Shu, Ngozi F Onyemelukwe, Emmanuel S Nwodo, Fred C Otuu, Lawrence Ilouno. Unwholesome Herbal Medicines Marketed in Enugu Metropolis, Enugu State, South Eastern Nigeria: Public Health Implications. Journal of Environmental Science and Public Health 3 (2019): 122-132.

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