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# Phytochemical and Trace Metal Analyses of Selected Local Spices Plants Consumed in Uyo, Akwa Ibom State, Nigeria

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**Abstract:** *Phytochemical and trace metal analyses of selected local spices plants: curry (Hyptis suaveolens), tea bush (Ocimum bassillicum), betel vine (Gongronnema latifolium) and climbing black pepper (Piper guineense) were performed using standard analytical methods in order to ascertain their curative properties in terms of Phytochemical contents and suitability for human consumption in terms of trace metal contents. From the results, phlobatatannins, saponins, reducing sugars, tannins, alkaloids, steroids and flavonoids were either absent or present in high, moderate or trace amount in all the spices analysed for phytochemicals. A total of eight trace metals (Zn, Mn, Cd, Cu, Fe, Pb, Ni and As) were analysed. The results indicated that the levels of trace metals analysed in Hyptis suaveolens ranged from 0.59 ± 0.10mg/kg As to 119.4± 0.25 mg/kg Zn. In Ocimum bassillicum, the trace metal levels ranged from 0.93± 0.02mg/kg As to 129.50± 0.15mg/kg Zn. In Gongronnema latifolium, the trace metal levels ranged from 0.68± 0.02mg/kg As to 116.30 ± 0.20 mg/kg Zn, while in Piper guineense, the trace metal levels ranged from 0.48± 0.02mg/kg As to 101.70 ± 0.20 mg/kg Zn. It was observed that most of the trace metal levels in the spices plants analysed in this study were within the WHO acceptable levels of such trace metals in plants. Going by the entire results, it could be concluded that these spices plants contain many useful phytochemicals which have preventive or disease protective properties. Hence the plants could be used locally for curing or prevention of various diseases as well as the production of new drugs for curing of such diseases and their consumption would not pose health hazards in terms of trace metal contents.*

**Keywords:** *Phytochemicals, Trace metals, Spices, Consumption, Curative, Hazards.*

## 1. INTRODUCTION

Phytochemicals are bioactive non-nutritive plant chemicals that have protective or disease preventive properties. They are non-essential nutrients, i.e., they are not required by the human body for sustaining life. These chemicals are produced naturally by plants to protect themselves, i.e., they are a part of plant natural defense system and they can also protect humans against diseases. They are formed during the plant's normal metabolic processes [1]. They confer plants with odour (terpenoids), pigmentation (tannins and quinines) and flavour (capsacin) [2]. Most plants are useful for healing and curing of human diseases because of the presence of photochemicals [3]. The use of plants as source of remedies for the treatment of diseases can be traced back to the prehistoric times [4]. Phytochemicals consist of primary and secondary components or metabolites. The primary components include chlorophyll, proteins and common sugars, while the secondary components are terpenoids, alkaloids, flavonoids, coumarins, glycosides, gums, tannins, steroids, terpenes, polysaccharides and phenols [5, 6]. Terpenoids exhibit various important pharmacological activities such as anti-inflammatory, anti-cancer, anti-malarial, inhibition of cholesterol synthesis, anti-viral and anti-bacterial activities [7]. Alkaloids are used as an aesthetic agents and are found in medicinal plants [8].

In this part of the world, many local plants such as curry (Hyptis suaveolens), tea bush (Ocimum bassillicum), betel vine (Gongronnema latifolium) and climbing black pepper (Piper guineense) are used as spices. Spices are chemicals which stimulate our taste buds. They are dried seed, fruit, root, bark, or vegetable substances used primarily for flavouring, colouring or preserving of foods. They are commonly used as chicken broth, oyster sauce, fish sauce, soup bases for instant noodles, many fast foods and restaurant dishes [9]. Many spices have antimicrobial properties. This may explain why spices are more commonly used in warmer climates, which have more infectious diseases, and why the use of spices is prominent in meat, which is particularly susceptible to spoiling [10]. Spices may have other uses such as medicinal, religious ritual, cosmetics or perfumes production and as vegetables [10].

*Hyptis Suaveolens,* commonly known as curry and called igorh in Ibibio, belongs to the family lamiaceae and is an important medicinal plant. The plant is considered as an obnoxious weed and is distributed throughout the tropics and sub tropics. Almost all parts of the plant are being used in traditional medicine to treat various diseases [11]. Fumes of dried leaves of Hyptis suaveolens are used to repel mosquitoes and control of insect pests of stored grains [12].

*Ocimum Bassillicum,* commonly known as tea bus or basil leaves and called ntong in Ibibio is a perennial plant that is woody at the base. It is widely distributed in the tropical and warm temperature regions and belongs to the family lamiaceae. It is a scented shrub with lime green fuzzy leaves and produces a stain odour. It is a well known plant in traditional medicine for the treatment of various diseases and plays effective roles in pharmaceutical science in the production of drugs, essential oil and insecticides [13].

*Gongronnema Latifolium,* commonly referred to as betel vine and called *utasi* in Ibibio belongs to the class of vegetables called the green leafy vegetables which are regarded as nourishing foodstuff because they contain a full complement of the basic nutrients needed by man. It belongs to the same group of climbing vegetable like the fluted pumpkin (*Telferia Occidentalis*) and belongs to the family asclepiadaceae. *Utasi* is found in the tropical rain forest and it is used as spice and vegetable in traditional medicine [14].

*Piper Guineense*, commonly known as climbing black pepper and called *odusa* in Ibibio belongs to the family piperaceae. It is an important medicinal plant and commonly used as spices.

Although several works reporting compositional evaluation and functional properties of various types of edible plants have been carried out, much still need to be done considering the importance of these plants. This study is aimed at analysing the phytochemical and trace metal contents of selected local spices plants: curry (*Hyptis suaveolens*), tea bush (*Ocimum bassillicum*), betel vine (*Gongronnema latifolium*) and climbing black pepper (*Piper guineense*) obtained in Uyo, Akwa Ibom State with a view to establishing their curing or prevention properties of various diseases and production of new drugs for curing of such diseases as well as their suitability for human consumption.

## 2. MATERIALS AND METHODS

### 2.1. Samples Collection and Preparation

Plants samples (Hyptis suaveolens, Ocimum bassillicum, Gongronnema latifolium and Piper guineense) were purchased from Akpan Adem market, Uyo, Akwa Ibom State. The plant samples were identified and authenticated by a Taxonomist in the Department of Botany and Ecological studies, University of Uyo, Uyo. The leaves of each plant samples were carefully removed from the stems, washed with distilled water and air dried for one week. The dried leaves were ground into powder using pestle and mortar and kept in properly labeled plastic bottles in freezer prior analyses.

### 2.2. Extraction of the Plant Samples

Ethanolic extract of the plant samples was prepared by soaking 100 g of each of the dried powdered plant leaves in 1000 cm3 of absolute ethanol at room temperature for 48 hours. The extracts were then filtered first through a Whatmann filter paper. The extracts were thereafter concentrated using a rotary evaporator with the water bath set at 60oC to one-tenth its original volume and then dried. The dried residues (crude extracts) were properly labeled and stored in a refrigerator prior phytochemical screening.

### 2.3. Methods for Phytochemical Screening

Phytochemical screening was performed using standard procedures described by [15, 16, 17].

*2.3.1. Test for Alkaloids*

Extracts (2.0g) were dissolved individually in 2 $cm^3$ of 10% hydrochloric acid and filtered. Filtrates were: (a) treated with 1 $cm^3$ Mayer`s reagent (potassium mercuric iodide) or 1 $cm^3$ Hager's reagent (saturated picric acid solution). Formation of a yellow colored precipitate indicates the presence of alkaloids and (b) treated with Dragendroff`s reagent (solution of potassium bismuth iodide).

Formation of red precipitate indicates the presence of alkaloid.

*2.3.2. Test for Flavonoids*

Test for flavonoids was done using two methods: (a) a portion (0.2g) of each of the extract was heated with 10 cm3 of ethyl acetate over a steam bath for 3 minutes, the mixture was filtered and 4 cm3 of

the filtrate was shaken with 1 cm3 of dilute ammonia. A yellow colouration indicates the presence of flavonoids and (b) dilute ammonia solutions (5 cm3) were added to a portion of an aqueous filtrate of each extract. Then, concentrated sulphuric acid (1 cm3) was added. A yellow colouration indicates the presence of flavonoids.

*2.3.3. Test for Phlobatannins*

A portion (0.5g) of each extract was mixed with 5 cm3 of water and boiled with 1% aqueous solution of hydrochloric acid for 2 minutes. Deposition of a red precipitate indicates a positive result.

*2.3.4. Test for Saponins*

A portion (0.5g) of each of the extract was added to 5 cm3 of distilled water in a test tube and the resulting solution shaken vigorously and observed for a stable persistent froth. The frothing was mixed with 3 drops of olive oil and shaken vigorously. Formation of an emulsion indicates the presence of saponins.

*2.3.5. Test for Tannins*

Two methods were adopted in the test for tannins: (a) ten (10) cm3 of freshly prepared 10% potassium hydroxide was added to 0.5g of each of the extract in a beaker and shaken to dissolve. A dirty precipitate indicates the presence of tannins and (b) a portion (0.5) g of the extract was boiled in 10 cm3 of water in a test tube and then filtered. A few drops of 0.1% ferric chloride were then added to the filtrates and the formation of brownish green or a blue-black colouration indicates the presence of tannins.

*2.3.6. Test for Reducing Sugars (Fehling's Test)*

A portion (0.5g) of the extracts was dissolved in 5 cm3 distilled water and filtered. The filtrates were hydrolysed with dilute hydrochloric acid, neutralized with sodium hydroxide solution and heated with Ferling`s A and B solutions. Formation of red precipitates indicated the presence of reducing sugars.

*2.3.7. Test for Steroids*

A portion (0.5g) of the extracts was dissolved in 10 cm3 of chloroform and equal volume of concentrated sulphuric acid added along the sides of the test tubes. Reddish upper layer and yellowish sulphuric acid layer with green fluorescence indicate the presence of steroids.

### 2.4. Digestion of Samples for Trace Metal Analysis

Digestion of the plant samples was performed using standard procedures described by [18, 19]. A portion (1.0g) of each of the dried ground powdered plant samples were placed separately in 50 cm3 Teflon beakers and digested with 10 cm3 of perchloric acid and 20 cm3 of nitric acid at 130oC on a hot plate until they appeared colourless. On cooling, the digests were filtered into 50 cm3 volumetric flasks using Whatman No. 42 filter paper.

### 2.5. Trace Metal Analysis

Trace metals (Zn, Mn, Cd, Cu, Fe, Pb, Ni and As) levels in the plant samples were analysed using an SP 1900 Pye Unicam Atomic Absorption Spectrophotometer (AAS) equipped with an air – acetylene burner. The mean values of three analyses per sample were recorded.

### 2.6. Statistical Analysis

The analyses were performed in triplicate and data were analyzed using Excel and Graph Pad in Stat 3.1.

## 3. RESULTS AND DISCUSSION

The results obtained in this study are presented in Tables 1 and 2. Table 1 shows the results of the qualitative phytochemical analysis in each of the spices plant. The results indicated that the phytochemicals analysed for, were either absent or present in trace, moderate or high concentration. Flavonoids were absent in all the four plants. Phlobatannins were absent in Hyptis suaveolens and Ocimum bassillicum, present in trace amount in Gongronnema latifolium and in moderate amount in Piper guineense. Steroids were only absent in Hyptis suaveolens. They were present in moderate concentration in Ocimum bassillicum and Piper guineense and in high concentration in Gongronnema

latifolium. Reducing sugars were present in trace amount in Hyptis suaveolens, Gongronnema latifolium and Piper guineense and in moderate amount in Ocimum bassillicum. Saponins and tannins were present in high concentration in all the four spices plants, except that tannins were present in moderate concentration in Gongronnema latifolium. Alkaloids were present in trace amount in Ocimum bassillicum and Gongronnema latifolium, moderate concentration in Piper guineense and high concentration in Hyptis suaveolens. Indeed, this study has revealed the presence of phytochemicals considered as active medicinal chemical constituents in all the four spices plants investigated.

Phlobatannins have been reported for its wound healing properties, these are anti-inflammatory and analgesic [20] and antioxidant [21]. Alkaloids have antibacterial and analgesic properties and plants that contain alkaloids are used in medicines for reducing headache and fever [22]. In addition, it is known that alkaloids have been used as central nervous system stimulants, topical anesthetics in ophthalmology, powerful pain relievers and anti-puretic action [23], so the use of these spices plants can serve these purposes. As noted by [24], the presence of tannins in the spices plants indicated that these plants have astringent properties that quicken the healing of wound and inflamed mucous membrane, contain antimicrobial properties and capable of protecting the kidneys from inflammation. Saponins in foods can be astringent [25]. Saponins in plants have been known to protect the plants from fungal and insect attacks [25]. Other studies have revealed that saponins have the ability to reduce the cholesterol levels in man and animals by forming complexes with cholesterol and bile acids which prevent them from being absorbed through the small intestine hence lowers the cholesterol level in the blood and liver [26]. Saponins are also known to serve as antioxidants as they prevent degeneration of DNA and also help to reduce colon damage and risk of cancer [23]. According to [27], saponins are used as adjuvants in vaccines and then oral intake has been used to help in managing retroviral infections. Also saponins stimulate antibody production, inhibit viruses and induce the response of lymphocytes which are white blood cells that fight infection [27]. As noted by [23], the presence of saponins in plants makes the plants good busters of the immune system. Therefore, the local spices plants investigated in this study can be regarded as good immune busters.

**Table1.** *Qualitative Phytochemical Analysis of the Spices Plants*

| **Phytochemicals** | **Local Spices Plants** | | | |
|---|---|---|---|---|
| | ***H. suaveolens*** | ***O. bassillicum*** | ***G. latifolium*** | ***P. guineense*** |
| **Flavonoids** | - | - | - | - |
| **Phlobatannins** | - | - | + | ++ |
| **Saponins** | +++ | +++ | +++ | +++ |
| **Tannins** | +++ | +++ | ++ | +++ |
| **Reducing Sugars** | + | ++ | + | + |
| **Steroids** | - | ++ | +++ | ++ |
| **Alkaloids** | +++ | + | + | ++ |

*- = Absent, + = Present in trace concentration, ++ = Present in moderate concentration, +++ = Present in high concentration*

The results of the trace metal analysis in spices plants are presented in Table 2. From the results, Zn levels in the plants ranged from 101.70 ± 0.20 mg/kg in Piper guineense to 129.50 ± 0.15 mg/kg in Ocimum bassillicum. Mn levels ranged from 44.36 ± 0.03 mg/kg in Gongronnema latifolium to 66.90 ± 0. 07 mg/kg in Piper guineense. The levels of Cd in the plants ranged from 1.68 ± 0.02 mg/kg in Gongronnema latifolium to 3.11 ± 0.01 mg/kg in Piper guineense. Cu levels in the plants ranged from 3.21 ± 0.01mg/kg in Gongronnema latifolium to 4.16± 0.02mg/kg in Piper guineense. Fe levels in the spices plants ranged from 289.00 ± 1.00mg/kg in Hyptis suaveolens to 549.00 ± 1.00mg/kg in Ocimum bassillicum. Pb ranged from 2.52 ± 0.02mg/kg in Piper guineense to 5.22± 0.03mg/kg in Gongronnema latifolium. Ni levels in plants ranged from 9.35± 0.02mg/kg in Gongronnema latifolium to Piper guineense and As ranged from 0.48± 0.02mg/kg in Piper guineense to 0.93± 0.02mg/kg in Ocimum bassillicum.

The high levels of Zn, Fe and Mn obtained in all the spices plant samples analysed in this study indicated that the plants may be good sources of these very essential elements in the body. Zn is known to be useful in almost all biochemical reactions in the human systems. It is a constituent of plasma or serum of the erythrocytes and leucocytes. It is an essential component of a large number (>300) of enzymes participating in the synthesis and degradation of carbohydrates, lipids, proteins,

and nucleic acids as well as in the metabolism of other micronutrients [28]. Zn stabilises the molecular structure of cellular components and membranes and contributes to the maintenance of cells and organs. It is known that Zn has an essential role in polynucleotide transcription as well as in the process of genetic expression. Its involvement in such fundamental activities probably accounts for the essentiality of Zn for all life forms [28]. It is a constituent of plasma or serum of the erythrocytes and leucocytes. Zn plays a central role in the immune system, affecting a number of aspects of cellular and humoral immunity [29].

The clinical features of severe Zn deficiency in humans are growth retardation, sexual infertility, delayed bone maturation, skin lesions, diarrhoea, alopecia, impaired appetite, increased susceptibility to infections mediated via defects in the immune system, and the appearance of behavioural changes [30]. [31] noted that Zn deficiency in humans is also associated with beri-beri, nephorosis, hepatic porphyria and post alcoholic cirrhosis of the liver. Fe has several vital functions in the body. It serves as a carrier of oxygen from the lungs to the tissues by the red blood cell haemoglobin, as a transport medium for electrons within the cells, and as an integrated part of important enzyme systems in various tissues [28]. It is known that Fe nutrition is of great importance for the adequate development of the brain and other tissues such as muscles, which are finally differentiated early in life. Mn is essential in nutrition. It serves as essential element for growth and metabolism in the human system.

**Table2.** *Trace Metal Levels (mg/kg) of the Spices Plants*

| Trace Metals | Local Spices Plants | | | |
|---|---|---|---|---|
| | ***H. suaveolens*** | ***O. bassillicum*** | ***G. latifolium*** | ***P. guineense*** |
| **Zn** | 119.40 ± 0.25 | 129.50 ± 0.15 | 116.30 ± 0.20 | 101.70 ± 0.20 |
| **Mn** | 63.29 ± 0.03 | 58.16 ± 0.02 | 44.36 ± 0.03 | 66.90 ± 0.07 |
| **Cd** | 3.01 ± 0.02 | 2.25 ± 0.02 | 1.68 ± 0.02 | 3.11 ± 0.01 |
| **Cu** | 3.48 ± 0.02 | 3.28 ± 0.02 | 3.21 ± 0.01 | 4.16 ± 0.02 |
| **Fe** | 289.00 ± 1.00 | 549.00 ± 1.00 | 316.00 ± 3.00 | 479.00 ± 3.00 |
| **Pb** | 3.08 ± 0.02 | 4.48 ± 0.02 | 5.22 ± 0.03 | 2.52 ± 0.02 |
| **Ni** | 11.03 ± 0.02 | 9.91 ± 0.02 | 9.35 ± 0.02 | 12.52 ± 0.02 |
| **As** | 0.59 ± 0.10 | 0.93 ± 0.02 | 0.68 ± 0.02 | 0.48 ± 0.02 |

*Above values are Means ± SD of triplicate analyses.*

Appreciable levels of Ni and Cu were recorded in all the spices plant samples analysed in this study, indicating that the plants may be good sources of the trace metals. Ni is believed to play a role in

Physiological processes as a co-factor in the absorption of iron from the intestine while Cu is known to play an important role in proteins synthesis [32].

The low levels of Cd, As and Pb in the studied spices plant samples suggest that the plants are nontoxic with respect to the metals. It is known that Cd, As and Pb are best known for their toxicological properties [33].

## 4. CONCLUSION

Going by the entire analyses and the results, it could be concluded that the spices plants: curry (Hyptis suaveolens), tea bush (Ocimum bassillicum), betel vine (Gongronnema latifolium) and climbing black pepper (Piper guineense) analyzed in this study contain many useful phytochemicals which have preventive or disease protective properties. Hence the plants could be used locally for curing or prevention of various diseases as well as the production of new drugs for curing of such diseases. Accordingly, the consumption of the spices plants would not pose possible health hazards in terms of trace metal contents since the levels of most of the trace metals analyzed in the plants were within the WHO acceptable levels of such trace metals in plant

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