



Inductively Coupled Plasma Optical Electro Spectroscopic Characterisation of Some Indigenous Nigerian Leafy Vegetables as Prospective Mineral Sources In Human Diets

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ABSTRACT

Minerals are nutrients that are needed in macro and micro amounts to guarantee proper body function. They are obtainable from plant and natural sources. There has been continuous change in climatic conditions and this has resulted to low yield, pest infestation, loss of farm land, and concomitant low income generated from vegetables that are consumed regularly. The regular cuisines are becoming scarce and expensive with many households not aware of possible sources of suitable substitute to complement and supplement mineral requirements. This study examined the mineral composition of some indigenous leafy vegetables that can be consumed by humans as an alternate source of mineral. *Colocasia esculenta* (red Cocoyam), *Laportea aestuans* (L) (Tropical nettle weed), *Daucus carota* L (Carrot leaves), *Manihot esculenta*, Crantz (Cassava leaves) and *Sesamum indicum* (Sesame leaves) were collected and authenticated. Inductively coupled Plasma-Optical Emission Spectrometry was used to analyse the mineral composition of each of the leaves. Microminerals like Boron, Copper, Iron, Manganese, Silicon, and Zinc were present in amount sufficient to fulfill the daily required intake when 100 g of the plant was consumed. Macrominerals like Calcium, Magnesium, Potassium, and Sodium were also observed to be in quantities able to supply the recommended daily allowance on consumption of 100 g of the plant. The plants are rich in micro and macro minerals and therefore should be explored further as prospective sources of minerals for humans.

Keywords: Minerals, Microminerals, Macrominerals, Oxidative stress, Vegetables.

Introduction

In the tropics, particularly in Nigeria, several plant organs are extensively utilized for dietetic and culinary purposes as authentic sources of nutrient (Delgado *et al.*, 2023). Hence, they can be utilized for the management and treatment of variety of physiological disorders in addition to serving as nutritious food constituent (Yedjou *et al.*, 2023). Plant parts, especially the leaves, have been

commonly used as sources of minerals due to its succulent nature (Jameel *et al.*, 2023). Minerals perform various biological roles in such processes like bone formation and Deoxyribonucleic acid (DNA) metabolism (Alghamdi, 2023; Yao *et al.*, 2023). Recommended Dietary Allowance (RDA) varies across minerals Hence, the classification of minerals into those that are needed in less than 100mg per day (microminerals) and those needed

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above 100mg per day (macrominerals) (Hamza *et al.*, 2023). Multiple deficiencies have been linked to insufficient consumption of these minerals. Depending on how severe the concurrent physiological impact is, the effects could be either reversible or irreversible (Stein, 2010; Saini *et al.*, 2016; Dhawan *et al.*, 2022). These elements are available naturally as salts and can be found also in plants and animals (Nemeth *et al.*, 2023). Meals rich in diverse vegetables can increase the chances of supplying the requisite measure of minerals for the various stages of the life cycle. Nigerian vegetation is characterised by tropical rainforest and Sahel grassland in Southern and Northern parts respectively. Challenges of seasonal variations have affected the cultivation of preferred vegetables used for various local cuisines. Therefore, there is a need to explore healthy alternatives outside of the traditionally preferred vegetables as dietary mineral sources.

Materials and Methods

Sample Collection and Identification

Fresh leaves samples of *Colocasia esculenta* (red Cocoyam), *Laportea aestuans* (L) (Tropical nettle weed), *Daucus carota* L (Carrot), *Manihot esculenta*, *Crantz* (Cassava) and *Sesamum indicum* (Sesame) were obtained from a local farm in Western Nigeria. The samples were identified and authenticated at a regional herbarium in Nigeria and voucher specimen numbers (UIH-23141, UIH-23142, UIH-23143, UIH-23144 and UIH-23149 respectively) were assigned. The voucher specimens were deposited at the herbarium for reference.

Preliminary Sample Preparation

The leaves were sorted to remove debris and extraneous matters, rinsed in plentiful water to remove debris and dried under convectional current to constant weight at room temperature. Each sample was then, pulverised and packed in zip lock polyethylene bag placed in air-tight lidded container and refrigerated (-4°C) until required for

analyses. Samples were stored for a maximum of two months in the refrigerator.

Mineral analysis of samples

Preparation of digest

One gram of each sample of leaves was weighed into previously demineralized and desiccated porcelain crucible of known weight and ashed in muffle furnace at 550 °C (H-1081 , Medical Instrument Cooperative, Rakoczi). The ash was made into solution of digest and the minerals (such as B, Co, Cr, Fe, Mn, Mo, Ni, Se, Si and Zn) analyzed by Inductively Coupled Plasma-Optical Emission Spectrometry.

Inductively Coupled Plasma-Optical Emission Spectrometry of Sample

Agilent 720 Inductively Coupled Plasma-Optical Emission Spectrometer (ICP-OES) (Agilent 720-ICP-OES1, Agilent Technologies, Santa Clara, United States) was used for the analysis using SPS3 auto-sampler for sample introduction. The Instrument was equipped with Agilent 720 ES megapixel CCD detector, which provided simultaneous measurement. Data acquisition and instrument control was driven by Agilent Expert II Software.

Calibration and Quality Control (QC) solutions were prepared from the following reference material; Accu standard QCSTD-27 multi-element solution AG-QCS27-ASC-1, Agilent Calibration Mix Majors 6610030700, Agilent Calibration Mix 2 6610030600 and Agilent ICMS Internal Standard Solution 6610030400. Ultrapure Merck Lichrosolv water was used to dilute standards and QC solutions. All the solutions used for analyses were stabilized in high purity 2% v/v concentrated HNO₃ solution and the instrument set to the parameters detailed in Table 1 according to the manufactures protocol.

Table 1: Icpoes instrumentation parameters according to Manufacturer protocol

Parameter	Setting
Power	1 kW
Plasma gas flow	15 L/min
Auxiliary gas flow	1.5 L/min
Spray chamber type	Glass cyclonic
Torch	Standard axial torch
Nebulizer type	Sea Spray
Nebulizer gas pressure	220 kPa
Pump speed	15 rpm
Sample uptake	30 s
Replicate read time	30 s
Number of replicates	2
Sample delay time	20 s
Stabilization time	15 s
Rinse time	10 s
Fast pump	On

The calibration curves were drawn using the values obtained from the standards and the samples analyzed according to the specification in the sequence parameters of the newly created worksheet.

Result

Micro minerals

The microminerals (mg/100g) detected in the leaves sample of carrot, red cocoyam, cassava, sesame and nettle weed were B (8.18 ± 0.03 to 16.96 ± 0.00), Cu (5.98 ± 0.03 to 7.15 ± 0.04), Fe (27.60 ± 0.06 to 128.81 ± 0.00), Mn (4.40 ± 0.00 to 48.32 ± 0.01), Si (33.99 ± 0.16 to 131.36 ± 0.00), and Zn (1.35 ± 0.00 to 8.66 ± 0.01); whereas Co, Cr, Ni and Se were less than their respective limits of detection (LOD) . Sesame leaves had the highest levels of Boron (B). Copper was not seen in the leaves of red cocoyam, cassava, and sesame. Cassava leaves had the highest Fe content whereas, red cocoyam leaves had the highest level of Mn. The Zn content for red cocoyam was lower than LOD of the instrument (Table 2).

Table 2: Concentrations of micro minerals (mg/100g wet weight) of plant leaves

Leaves	B	Co	Cr	Cu	Fe	Mn	Mo	Ni	Se	Si	Zn
Carrot(mg/100g)	8.18 ± 0.03^a	<LOD	<LOD	5.98 ± 0.03^a	27.60 ± 0.06^a	18.43 ± 0.03^b	<LOD	<LOD	<LOD	33.99 ± 0.16^a	3.31 ± 0.04^b
Red cocoyam (mg/100g)	11.33 ± 0.01^c	<LOD	<LOD	<LOD	74.11 ± 0.02^c	48.32 ± 0.01^c	<LOD	<LOD	<LOD	122.05 ± 0.13^d	<LOD
Cassava (mg/100g)	13.60 ± 0.00^d	<LOD	<LOD	<LOD	128.81 ± 0.00^e	33.46 ± 0.00^d	<LOD	<LOD	<LOD	87.32 ± 0.00^c	8.66 ± 0.01^d
Sesame (mg/100g)	11.02 ± 0.01^b	<LOD	<LOD	<LOD	34.00 ± 0.03^b	28.40 ± 0.02^c	<LOD	<LOD	<LOD	50.83 ± 0.38^b	6.16 ± 0.06^c
Nettle weed (mg/100g)	16.96 ± 0.00^e	<LOD	<LOD	7.15 ± 0.04^b	44.10 ± 0.003^c	4.40 ± 0.00^a	<LOD	<LOD	<LOD	131.36 ± 0.00^e	1.35 ± 0.00^a

Values in a column with different superscripts are significantly different at $p < 0.05$ for $n=3$. LOD stands for limit of detection.

Macrominerals

Table 3 shows the different levels of Ca, Mg, K and Na (all in mg/100g) present in the leaves studied. The values for Ca ranged from 2225.80 ± 2.36 to 8109.31 ± 0.15 (mg/100g) with Nettle weed having the highest value. K ranged from 105.25 ± 0.62 to 15035.12 ± 11.91 (mg/100g) in nettle weed

leaves and sesame leaves, respectively; whereas Mg content of the leaves ranged from 476.21 ± 0.34 to 1305.31 ± 0.01 (mg/100g), with carrot leaves and nettle weed leaves having the lowest and highest values, respectively. The Na content of the leaves ranged from 22.66 ± 0.11 to 576.34 ± 0.02 , with red cocoyam leaves and cassava leaves having the lowest and highest value, respectively.

Table 3: Concentrations of macro minerals (mg/100g wet weight) in studied plant leaves

Leaves	Ca	K	Mg	Na
Carrot (mg/100g)	2225.80 ± 2.36^a	3433.73 ± 3.36^b	476.21 ± 0.34^a	89.80 ± 0.14^b
Red cocoyam (mg/100g)	3976.855 ± 0.92^c	7247.73 ± 2.79^c	890.38 ± 0.11^c	22.66 ± 0.11^a
Cassava (mg/100g)	6335.15 ± 0.15^d	10855.18 ± 0.52^d	1153.02 ± 0.04^d	576.34 ± 0.02^c
Sesame (mg/100g)	3660.99 ± 0.94^b	15035.12 ± 11.91^e	881.73 ± 0.15^b	206.56 ± 0.29^c
Nettle weed (mg/100g)	8109.31 ± 0.15^e	105.25 ± 0.62^a	1305.31 ± 0.01^e	255.73 ± 0.00^d

Values in a column with different superscripts are significantly different at $p < 0.05$ with three replicates ($n=3$) (LOD- limit of detection)

Discussion

Microminerals

Microminerals are needed in trace quantity (less than 100mg per day) for metabolic functioning, osmotic regulation, redox balancing, and structural development, among other biological roles (Maradi *et al.*, 2022). Copper, which plays catalytic roles as cofactor (National Research Council Committee on Copper in drinking water, 2000) predominantly, among its diverse biological roles (Wang *et al.*, 2021); has recommended daily intake for women and men as 1.0 to 1.1 mg and 1.2 to 1.6 mg, respectively (Ito *et al.*, 2022). Carrot and Nettle weed leaves contained 6.64 times and 7.94 times, respectively of the RDA respectively (see Table 2). Values observed in the vegetables studied are in the range of those reported by Moyo *et al.* (2018) for copper content of *Cleome gynandra*, *Brassica oleracea* and *Beta vulgaris*. Thus, Carrot leaves and Nettle weed leaves are excellent sources of copper that could be further

explored for nutraceutical and functional food purposes. As such, they could be adopted in managing biomedical conditions that require optimization of copper levels, including copper deficiency.

Iron has been observed to up-regulate erythropoiesis, enhanced oxidative processes, and optimize immune surveillance in biological systems (Chang *et al.*, 2019; Kumar *et al.*, 2019). Tropical nettle weed, red cocoyam, and cassava leaves were abundant in iron; implying that informed consumption of these leaves may help individuals meet the average dietary intake of 16 to 18 mg per day and 12 mg per day for men and women respectively (Stoffel *et al.*, 2020). Tropical nettle weed, red cocoyam, and cassava leaves could also be considered for possible inclusion in composite formulations aimed at addressing related conditions, such as iron-deficiency anaemia. The values of iron observed in the vegetable studied were higher than the 30.6 mg/100 g to 38.4 mg/100 g observed by Barminas

et al (1998) and 1.17 mg/100 g to 38.6 mg/100 g reported by Opazo-Navarrete *et al* (2021) in related study.

The average intake of Mn is estimated at 2.3 and 1.8 mg/day for men and women respectively (Choi & Bae, 2020). Red cocoyam leaves, cassava leaves, sesame leaves and carrot leaves are very rich sources of Mn that could be considered for inclusion in functional food and nutraceutical formulations targeted at addressing Mn under-nutrition. Inadequate intake of Mn has been associated with aberrant biological conditions such as stunted growth, temporary dermatitis, diminished fertility and birth abnormalities, hypocholesterolemia, poor glucose tolerance, nausea, and vomiting (Martins *et al.*, 2020).

Zinc, with Recommended Dietary Allowance of 8mg and 11 mg per day for male and female adults respectively (Horváth & Babinszky, 2018), was the only antioxidant mineral present in all the leaves except red cocoyam. Moyo *et al.* (2018) reported similar trend in certain non-conventional vegetables studied. The positive effect of zinc in biological redox system, catalytic activity, structural functions, immunological modulation and neurological effectiveness among others (Maret, 2013; Roohani *et al.*, 2013), can also be maintained by consuming cassava leaves, sesame leaves and carrot leaves objectively.

Macroelements

Healthy nutrition requires that all the microminerals be consumed in adequate amount (Adenegan-Alakinde & Ojo, 2022), furthermore, other nutrients required for optimum growth and development (Granato *et al.*, 2010). Macrominerals are just as vital as microminerals and are needed in comparatively larger quantities. They have been found to be involved in nearly every metabolic activity in biological systems (Ali *et al.*, 2023).

Calcium plays a plethora of metabolic and physiological roles, including bone formation,

enamel reinforcement, homeostasis and signal transduction pathway (Cormick & Belizán, 2019; Li *et al.*, 2018; Suherman *et al.*, 2022). The recommended dietary allowance of calcium is 1000 to 1300 mg per (National Institute of Health, 2022). All the leaves (100 g) contained about 2-4 times the RDA of calcium and therefore, may be applied in the management of calcium related conditions, such as osteoporosis (Elizabeth & Olawumi, 2018).

Potassium (K) is involved in trans-membrane electrochemical potential modulation, Sodium homeostatis, and cardiac function (Natale *et al.*, 2021). All samples (100 g portion each) contained abundant potassium exceeding the RDA of 2600-3400 mg/day (Eggleston *et al.*, 2022). Cassava leaves and sesame leaves are excellent sources that could be adopted when the need to boost level of potassium in biological systems arises.

Sodium is essential for maintaining proper hydration and regulating blood pressure, much like potassium (Jhee *et al.*, 2022). It co-regulates ATP with potassium and results in hypertension when in excess. The upper tolerable level is 2300mg/day while its recommended daily allowance is 1500mg/day. Eggleston *et al.* (2022) observed that foods that are low in sodium but relatively high in potassium are naturally veritable in moderating vascular volume (see table 3). Therefore, vascular volume moderation can be enhanced by copious consumption of sesame, tropical nettle weed and cassava because they have low sodium content but high potassium levels.

Conclusion

Carrot leaves, red cocoyam leaves, cassava leaves, sesame leaves and nettle weed leaves are widely available vegetables that can serve as excellent sources of micro- and macrominerals that could be considered for dietary inclusion in healthy nutrition. They can also serve as both functional food components and sources of mineral nutraceuticals for management of related malnutrition and downstream impacts. These

vegetables should be further analyzed for safety as they have great potentials as possible sources of attaining recommended daily intakes of micro- and macrominerals.

Recommendation

The toxicological characteristics and bioactivities of the secondary metabolites of carrot leaves, red cocoyam leaves, cassava leaves, sesame leaves and nettle weed leaves should be investigated alongside their nutraceutical potentials. This would further generate data that could form broader bases for informed choices in the utilisation of these non-conventional vegetables for diverse applications.

Conflicts of Interest

The authors declare that there is no conflict of interest.

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