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## **NUTRACEUTICAL AND ANTINUTRITIONAL PROPERTIES OF WILD EDIBLE PLANTS CONSUMED BY PREGNANT WOMEN AND SCHOOL-AGE CHILDREN (6-12 YEARS) IN NAJJEMBE SUB-COUNTY, BUIKWE DISTRICT, UGANDA**

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## ABSTRACT

Wild edible plants (WEPs) immensely contribute to the diets and livelihoods of rural and marginalised communities. In rural Uganda, they play an important role as break tea packages for school-age children and due to their perceived nutraceutical significance, they are always added to the diets of pregnant women. In this study, it was hypothesised that *Basella alba* L. (Malabar spinach, Ceylon spinach, Indian spinach, Vine spinach, Malabar nightshade), *Termitomyces microcarpus* (Berk. & Broome) R. Heim (*Cendawan Tali; Cendawan Taugeh*) *Cucurbita pepo* L. (Courgette, Marrow, Pumpkin, Summer squash, Table queen squash, Vegetable Marrow, Zucchini) and *Solanum anguivi* L. (Forest bitter berry, African eggplant) have high nutraceutical significance and could contribute to the diets of school-age children and pregnant women. Using the standard Association of Official Analytical Chemists (AOAC) methods (method 935.14 and 992.24), the nutraceuticals and antinutrients in *B. alba*, *T. microcarpus*, *C. pepo*, and *S. anguivi* were investigated. All analyses and measurements were done in triplicate to increase the validity and reliability of the results. *Solanum anguivi* had the highest crude fibre ( $25.22 \pm 0.08$  %), fat ( $6.55 \pm 0.05$  %), and energy ( $371.37 \pm 1.17$  Kcal) content. *Termitomyces microcarpus* had the highest protein ( $21.99 \pm 0.16$  %) and moisture ( $10.71 \pm 0.35$  %) content. *Basella alba* and *C. pepo* had the highest ash ( $19.39 \pm 0.01$  %) and total carbohydrate ( $69.15 \pm 0.62$  %) content. In comparison with the recommended dietary daily intakes for pregnant mothers and school-going children, all the plants had high Potassium, Phosphorous contents (except for *T. Microcarpus* which had low K contents), iron, copper, zinc, and manganese., but with selenium content being below the detection limit. All the analysed species were good in antioxidants, *C. Pepo* being the best with an  $IC_{50}$  of  $11.78 \pm 0.47$  mg/ mL followed by *B. alba* ( $14.69 \pm 0.74$  mg/ mL). *Solanum anguivi* and *T. microcarpus* had the highest amount of phenols ( $14.48 \pm 0.02$  mg/g GAE) and tannins ( $0.42 \pm 0.00$  mg/g GAE). *Basella alba* had the highest amounts of flavonoids ( $2.45 \pm 0.01$  mg/g RAE). All the plant species had low contents of phytates ranging from ( $0.40 \pm 0.01$  for *S. Anguivi* to  $0.72 \pm 0.08$  for *B. Alba*). Cadmium and lead concentrations were below the permissible limit recommended by FAO/WHO Expert Committee in Food Additives. Thus, the analysed WEPs are a rich source of important nutrients and can be used to complement staple foods to ensure balanced nutrition for pregnant women and school-age children. Nonetheless, further comprehensive antinutrients and heavy metals analyses should be conducted to ensure that they do not affect the bioavailability of the nutrients. For value addition, nutrient-dense cocktails / phytonutraceuticals should be developed to alleviate malnutrition-related illnesses in vulnerable groups of people.

**Key words:** Wild edible plants, Nutraceutical, Antinutrients, Heavy metals, Pregnant women, School-age children

## INTRODUCTION

Pregnant women and school-age children in rural Uganda are facing malnutrition [1]. Thirty-one percent of pregnant women in Uganda suffer from anaemia while 10% of all births are low birth weight [1]. Uganda's maternal mortality rate is 336 deaths per 100,000 live births [2], and half of these deaths are attributable to maternal malnutrition [2]. Additionally, 2.2 million (29%) children under 5 years suffer from stunting and almost 49% suffer from anaemia [3]. Over 5.2 million (42.8 %) school-age children drop out of school [4] due to hunger among other causes. The government of Uganda recognised this problem and put in place directives to encourage pregnant women to go for antenatal visits and parents to pack food for school-age children. Unfortunately, over 5 million homesteads are under a peasantry system with poverty levels at 27.7% [5], and are unable to afford 3 meals a day. Finding alternatives for availing the dietary nutrient requirements for pregnant women and school-age children that are available, sustainable, reliable, and of quality is urgent.

Plants are considered excellent sources of essential nutrients that form the basic source of diet for the human community. Wild edible plants (WEPs) are food plants that are neither cultivated nor domesticated but are harvested from agricultural areas, uncultivated areas, or forestland [6]. They have high nutraceutical value and are especially important for the poorest members of user communities, rural populations, children, and women, particularly during critical food shortages. The nutritional quality of WEPs is comparable to and sometimes superior to domesticated varieties [7]. Fortunately, WEPs are available, affordable, and lucrative sources of nutrients for many economically deprived communities.

Despite the nutraceutical significance of WEPs, the correlation between their elemental content and nutraceutical ability is not yet well understood. This is due to the presence of antinutrients (oxalates, phytates, tannins, alkaloids, saponins) [7]. Antinutrients are chemicals that have been evolved by plants for their defense. They reduce the maximum utilization of nutrients by the body thereby decreasing the nutritive value of food [8]. Furthermore, antinutrients are deleterious to both human and animal health when consumed in large quantities but can also be advantageous if consumed in appropriate amounts. Therefore, bimolecular analyses of WEPs would help raise awareness about their nutraceutical significance to further scientifically prove their food and medicinal roles.

Following an ethnobotanical survey on WEPs consumed by pregnant women and school-age children in Najjembe sub-county, Buikwe district, Uganda, *Basella alba*,

*Termitomyces microcarpus*, *Cucurbita pepo*, and *Solanum anguivi* were prioritised by the local community as species of high nutraceutical significance which influenced our choice to consider them for nutritional analyses.

## METHODS

### Sample Collection and Preparation

A cross-sectional ethnobotanical study was conducted in Najjembe sub-county, Buikwe district, Uganda to identify the wild edible plants consumed by pregnant women and school-age children. Through purposive sampling, pregnant women and school-age children were identified in Najjembe sub-county, Buikwe district. To these, semi-structured questionnaires on the wild edible plants added to their diets were administered. More study participants were acquired through snowball sampling. Najjembe sub-county was selected for the ethnobotanical survey because it houses the largest natural forest in Uganda that is the Mabira central forest reserve. Mabira forest has a high diversity of WEPs which the local population relies on for their livelihood. Following questionnaire administration, focus group discussions with key informants mainly the elderly were conducted and using pairwise ranking, the most preferred species were noted. These are species that are perceived by the locals of Najjembe to have a high nutraceutical value based on long-term use within the community.

After the focus group discussions, walks led by local chiefs were made to the wild edible plants' collection sites, and samples of *T. microcarpus* (whole) (Fig. 1), *S. anguivi* (fruits) (Fig. 2), *B. alba* (leaves) (Fig. 3), and *C. pepo* (pulp) collected fresh in 100 kg sacks each. The plants were locally identified using the Luganda dialect (Local language used in Central Uganda) as *Obutiko obubaala*, *Katunkuma*, *Enderema*, and *Ensujju*, respectively. Voucher specimens were also collected and pressed then deposited at the Makerere University Herbarium, which doubles as a National Herbarium for proper scientific identification.

Priority wild edible plants collected from Najjembe sub-county, Buikwe district in January 2020, and the parts consumed. *T. microcarpus* (whole) (Fig. 1), *S. anguivi* (fruits) (Fig. 2), *B. alba* (leaves) (Fig. 3), and *C. pepo* (pulp) (Fig. 4).



Figure 1: *Termitomyces microcarpus* (Berk. & Broome) R. Heim



Figure 2: *Solanum anguivi* L.



Figure 3: *Basella alba* L.



Figure 4: *Cucurbita pepo* L.

Sacks of fresh samples were then taken to the laboratory, and cleaned of any extraneous material using distilled water to avoid any contamination. The leaves of *B. alba* were air-dried for 14 days whereas the fruits of *S. anguivi* fruits *C. Pepo* pulp and flesh *T. Microcarpus* were sliced into small pieces and dried in an oven (GallenKamp Hotbox oven, UK) at 40° – 90°C for 7 days. After drying, the samples were packed in airtight analytical-grade bags. Before grinding them to powder, the previously dried samples were placed in an oven again at 40° C for 24 hours to dry to constant weight. Using a grinder (Brook Crompton, Series 2000, UK), they were ground to a fine powder. The powders were stored in air-tight bottles at room temperature ready for analysis. All analyses followed standard AOAC methods using 100 g of each sample and were performed in triplicate and the average readings were determined.

### Proximate analysis

The content of moisture, ash, crude fibre, crude protein, lipid, and carbohydrates was determined following the standard methods of the AOAC [9].

**Moisture content:** This was determined by drying the samples at 105°C in an oven for 72 hours until a constant weight was obtained. The samples were then cooled in a desiccator and weighed. The difference between the original weight and the dried weight expressed as a percentage of the original weight gave the moisture content.

**Ash:** Ash content was determined by incineration in a muffle furnace at a temperature of 550°C until it turned grey.

**Crude fibre:** Crude fibre was obtained from the loss in weight on ignition of dried residue following the digestion of fat-free samples with 1.25% each of sulfuric acid and sodium hydroxide solutions.

**Crude Protein:** Nitrogen content was determined according to the Kjeldahl method and nitrogen value was multiplied by 6.25 as a conversion factor.

**Crude fat:** Total fat was determined by extracting the material in hexane using a Soxhlet extractor, distilling off the hexane, and drying for 5 days for the remaining solvent to evaporate. The weight of oil left was then expressed as a percentage of the dry weight of the sample.

**Carbohydrate:** Total carbohydrate was calculated as “crude by the difference”  
 $\% \text{ Total carbohydrate} = 100\% - (\% \text{ Crude protein} + \% \text{ Crude ash} + \% \text{ Crude lipid} + \% \text{ Moisture})$

**Energy content:** Energy content was estimated in kcal/100g using the Atwater general factors. The percentage available crude protein, crude fat, and crude carbohydrate were multiplied by 4, 9, and 4 respectively. Thus, energy values were calculated as:

$$(\text{Crude protein} \times 4 \text{ kcal}) + (\text{Crude fat} \times 9 \text{ kcal}) + (\text{Total carbohydrate} \times 4 \text{ kcal}) = \text{k cal}$$

**Atwater factors:** 17 kJ/g (4.0 kcal/g) for protein, 37 kJ/g (9.0 kcal/g) for fat, 17 kJ/g (4.0 kcal/g) for carbohydrates.

### Mineral Profile Analysis

One hundred grams of dried plant materials, in triplicate, were digested using wet digestion procedure with a mixture of HNO<sub>3</sub>/HCl<sub>4</sub>O/H<sub>2</sub>SO<sub>4</sub> in the ratio 9:2:1 v/v, respectively, allowed to cool and filtered through No. 42 Whatman filter paper. The samples were each made of up to 25 mL with deionised water and each sample aliquot was then used to determine the elemental content. The mineral ions sodium, potassium, calcium, magnesium, iron, zinc, manganese, lead, selenium, cadmium, and copper were measured after wet-ashing by Atomic Absorption Spectrophotometer (Perkin Elmer, model A Analyst 700). Phosphorus was

estimated calorimetrically (UV-visible spectrophotometer), using potassium dihydrogen phosphate standard [9].

## Phytochemical analysis

### Vitamin C Analysis

Vitamin C was determined by the dye reduction method [10]. Samples were extracted with metaphosphoric acid (1%, 10 mL) for 45 minutes at room temperature and filtered. The filtrate (1 mL) was mixed with 2,6-dichloroindophenol (9 mL) and the absorbance was measured spectrophotometrically at 515 nm against a blank. The content of vitamin C was calculated based on the calibration curve of authentic L-ascorbic acid.

### DPPH (2, 2-diphenyl-1-picryl hydrazyl) Radical Scavenging Assay

The antioxidant activity was determined by using the 2, 2-diphenyl-1-picryl hydrazyl (DPPH) radical scavenging assay as described by Choi [11]. The DPPH (1 mL, 0.3 mM) solution in methanol was added to 2.5 mL (100 mg mL<sup>-1</sup>) of extract and the mixture was allowed to react for 30 minutes at room temperature. The reduction of the DPPH radical was measured using a UV visible spectrophotometer at 517 nm. Ascorbic acid was used as a positive control, methanol as the negative control, and extract without DPPH as blank. The experiment was repeated in triplicate, and the average absorbance values were recorded. The radical scavenging activity (RSA) was calculated as a percentage of DPPH discoloration using the following equation:

$$RSA(\%) = \left[ \frac{\text{Absorbance of control} - \text{Absorbance of test sample}}{\text{Absorbance of control}} \right] \times 100$$

The scavenging activity was plotted against concentration and IC<sub>50</sub> (the extract concentration providing 50% of radicals scavenging activity) value was calculated from the graph by linear regression analysis and expressed as mg/mL.

### Total Flavonoid Content

The total flavonoid content was determined by the Aluminium chloride method [12] using quercetin as a standard. One millilitre of the test sample and 4 mL of water were added to a 10 mL volumetric flask. Zero-point three millilitres of 5 % Sodium nitrite and 0.3 mL of 10% aluminium chloride were added to the same 10 mL volumetric flask after 5 minutes. After 6 minutes of incubation at room temperature, 1 mL of 1 M Sodium hydroxide was added to the reaction mixture. Immediately the final volume was made up to 10 mL with distilled water. The absorbance of the sample was measured against the blank at 510 nm using a spectrophotometer.

The experiment was repeated three times for precision and values were expressed in mean  $\pm$  standard deviation of total flavonoid in Quercetin equivalent, (QE) per gram of dry weight.

## Antinutrients Analysis

### Total Phenol Content

The total phenol content was determined with the Folin- Ciocalteu's assay [13] using gallic acid as standard. In the procedure, 0.5 mL of plant extracts were mixed with 1.5 mL Folin- Ciocalteu's reagent (FCR) diluted 1:10 v/v then after 5 minutes 1.5 mL of 7% sodium carbonate solution was added. The final volume of the tubes was made up to 10 mL with distilled water and allowed to stand for 90 minutes at room temperature. The absorbance of the sample was measured against the blank at 750 nm using a spectrophotometer. The experiment was repeated three times for precision and values were expressed in mean  $\pm$  standard deviation in terms of phenol content (Gallic acid equivalent, GAE) per g of dry weight.

### Total Tannin Content

The tannin content was determined according to Broadhurst *et al.* [14] with slight modification, using catechin as a reference compound. Four hundred microlitres of extract were added to 3 mL of a solution of vanillin (4% in methanol) and 1.5 mL of concentrated hydrochloric acid. After 15 minutes of incubation, the absorbance was read at 500 nm. The tannin content was expressed as g E.Catechin.100g<sup>-1</sup>DM.

### Phytate Content

Phytate was determined using Reddy and Love [15] method. Four grams of the ground sample were soaked in 100 mL of 2% HCl for 5 hours and filtered. To 25 mL of the filtrate, 5 mL of 0.3% ammonium thiocyanate solution was added. The mixture was then titrated with Iron (III) chloride solution until a brownish-yellow colour that persisted for 5 minutes was obtained.

### Mineral and Vitamin scorings

Mineral and vitamin scores were computed as described by Kabasa *et al.* [16] to establish the species recommended daily intake (RDI) percentage contributions per 100 g of dry sample. The mineral and vitamin contents were transformed into mineral and vitamin scores, namely: individual mineral score (IMS) and individual vitamin score (IVS). Individual mineral/ vitamin score is defined as the score of a specific mineral/vitamin in each plant species, measured at the RDI. Therefore, the analyzed species' mineral and vitamin content was transformed into mineral/

vitamin scores based on the following formula to ease the interpretation of mineral/vitamin composition:

$$\text{IMS/IVS} = \frac{\text{Individual mineral/ Individual vitamin content}}{\text{RDI}} \times 100$$

Where:

IMS = Individual mineral score;

IVS = Individual vitamin score;

RDI = Recommended Daily Intake

### Minerals Bioavailability

Molar ratios of antinutrient/minerals were used to predict the bioavailability of the minerals [17]. The proposed critical values used to predict the mineral bioavailability were: Phytate/Zinc > 15, Phytate/Calcium >0.24, Phytate/Iron >1 and Phytate/Calcium/Zinc >200 [17].

### Data Analysis

Nutritional values were entered in Excel programme (Microsoft Excel v. 2010) and cleaned. The SPSS Version 16 was used to generate means, standard deviations, and standard mean errors. One-way ANOVA was used to determine the differences between the means of variables of the plant species at a confidence interval of 95%,  $p \leq 0.05$ . Results were given as mean  $\pm$  standard deviation.

## RESULTS AND DISCUSSION

Pregnant women and school-age children (6–12 years) are vulnerable groups of persons whose growth and development can easily be compromised by nutritional inadequacies. This situation is exacerbated by poverty and marginalisation, especially in the rural communities of Uganda and other developing countries [1]. Wild edible plants still play an important role when food crops are scarce, ensuring food sovereignty and food security, and they potentially contribute to well-being in vulnerable households [18]. Additionally, wild edible plants have for a long time been recognized as a cheap and abundant source of food and medicine. Thus, due to their nutraceutical significance, communities in and around Mabira central forest reserve have always added them to the diets of school-age children, pregnant women, and the elderly.

## Proximate composition of *Basella alba*, *T. microcarpus*, *C. pepo*, and *S. anguivi*

The proximate composition of *B. alba*, *T. microcarpus*, *C. pepo*, and *S. anguivi* is presented in Table 1. The proximate composition results of the species analysed in this study were above but with a similar trend to wild edible plants in other studies elsewhere [19, 20].

Moisture content is an important parameter for consideration during food processing, preservation, storage, labeling, microbial stability, and food quality [21]. It affects the physical and chemical content of food which relates to freshness and stability. Inappropriate moisture amounts are damaging to the shelf life of food. Thus, much effort is put into reducing the water content of dry foods to prevent microbial attacks and for prolonged shelf life. Most fruits and vegetables have very high water content (80-95%) at harvest and the rate of water loss is affected by the shape and structure of the produce, the plant factors as well as the environmental conditions such as temperature and relative humidity [22]. All the species in this study had low moisture content which ranged from  $8.10 \pm 0.19$  to  $10.71 \pm 0.35$  % for *Termitomyces microcarpus* and *S. anguivi* respectively. This may be attributed to the nature of the samples (dry). Nonetheless, the low moisture content in these species increases their shelf life thereby helping in improving household nutrition, poverty alleviation, and income generation [15] and are also used as planting materials, especially *S. anguivi* (fruits) and *C. pepo* (pulp) which when they dry up most content left behind are the dry fruits which can be planted and immediately germinate. Furthermore, because the samples' moisture contents are low, they can help obese people to manage their weight.

*Solanum anguivi* had the highest crude fibre ( $25.22 \pm 0.08$  %) content. Fibre is an important part of the diet and the consumption of dietary fibre is important for optimal health. Crude fibre is made up largely of cellulose together with a little lignin which is indigestible in humans. Although crude fibre enhances digestibility, its presence at high level can cause serious health problems. Foods with high fibre content are considered good for diabetic patients and also reduce blood cholesterol, obesity, and diabetes [23]. Therefore, *B. alba*, *T. microcarpus*, *C. pepo*, and *S. anguivi* can help overcome gestational diabetes and preeclampsia while avoiding obesity in school-age children. Unlike foods of animal origin that contain all of the essential amino acids and are called complete proteins, plant foods do not have complete proteins. Complete proteins are obtained by eating combinations of plant and animal foods. About 20% of the human body is made up of protein. Protein plays many functions in the human body-building material for skin, bones, muscles, and other tissues in the body. It is a regulator of fluid

balance and acid-base balance; a major component of enzymes, antibodies, and hormones; acts as transporters in the body, and is used for energy. The protein content of the species under study ranged from  $11.85 \pm 0.40\%$  to  $21.99 \pm 0.16\%$ . *Termitomyces microcarpus* had the highest crude protein content and *C. pepo* had the lowest respectively. Crude protein values of all the species under study exceed values documented in other studies [24]. Proteins from plant sources have lower quality but their combination with many other sources of protein such as animal protein results in adequate nutritional value [25]. Therefore, the species under study should be eaten in combination with other vegetables or should be supplemented with animal protein to ensure maximum value and to help avert protein-energy malnutrition [21] common in school-age children and pregnant women.

The total ash represents the total mineral content in foods [9]. Since certain foods are higher than others in particular minerals, total ash content becomes important [10, 26]. The total ash content of most fresh foods is rarely greater than 5% [11]. In this study, the ash content of the species ranged from  $7.24 \pm 0.04$  to  $19.39 \pm 0.01\%$ . *Basella alba* and *T. Microcarpus* had the highest ash values  $19.39 \pm 0.01$  and  $17.08 \pm 0.11\%$ , respectively. Low ash values as depicted in *C. Pepo* ( $9.56 \pm 0.27\%$ ) and *S. anguivi* ( $7.24 \pm 0.04\%$ ). The high ash contents in *Basella alba* and *T. Microcarpus* present high concentrations of minerals that catalyse metabolic processes and improve growth and development. Nonetheless, low total ash content values in *C. Pepo* ( $9.56 \pm 0.27\%$ ) and *S. anguivi* ( $7.24 \pm 0.04\%$ ) are desirable because of their effect on biomass energy value; the higher the total ash content the lower the energy value [27]. The total ash content values of the species in this study exceed values documented in other studies [24].

Lipids provide very good sources of energy and aid in the transportation of fat-soluble vitamins (A, D, E, K), insulate and protect internal tissues and contribute to important cell processes [28]. The species under study had low-fat content ( $0.84 \pm 0.03$  to  $6.55 \pm 0.05$ ) which was in line with a study by Brahma *et al.* [29] which investigated the nutrients of selected wild edible species in Assam and reported crude fat ranging from 0.54-2.37% dry weight, hence, a need to supplement WEPs. Fat moderation in both pregnant women and school-age children is essential to avoid the undesired weight gain that may result in dystocias and obesity, respectively. During the prenatal period, lipids are also essential for the formation of cell membranes and hormones and are necessary for proper eye and brain development [30]. All species had very good amounts of carbohydrates and exceptionally high energy content. During pregnancy, the highest energy requirements are experienced during the third trimester [24]. These extra energy

requirements facilitate the growth of the fetus, placenta, and various maternal tissues in the uterus, breasts, and fat stores, as well as for changes in maternal metabolism [31]. School-age children require sufficient energy to meet their growth requirements, which is rapid during that time.

### **Micronutrients in *Basella alba*, *T. microcarpus*, *C. pepo* and *S. anguivi***

Micronutrients are critical for optimal pregnancy outcomes and proper metabolic activities that support tissue growth and function in the developing fetus. As such, deficiencies result in a vast array of adverse health outcomes which establishes a trajectory for chronic illness and other diseases in adolescence and adulthood [32]. In school-age children, micronutrient deficiencies are associated with stunting that can be sustained into adulthood, chronic diseases relating to nutrition, and lower educational attainment [33]. *Basella alba*, *T. microcarpus*, *C. pepo*, and *S. anguivi* contained major minerals Na, K, Ca, P, Mg (Table 2) and trace minerals Cu, Fe, Zn, Mn (Table 3). Mineral elements varied widely depending on the species. All the analysed species were high in K, P, Fe, Mn, Cu, and Zn contents and low in Na, Ca, and Mg. The Se content of all the species was below the detection limit (Table 3). This is contrary to other research carried out on wild edible plants in Uganda [19] where low levels of major minerals were reported as compared to the high contents of trace minerals. K and P were the most predominant minerals which are in line with other studies done elsewhere [19]. K contents ranged from  $111.48 \pm 8.49$  to  $3944.99 \pm 15.25$  mg/100g and P ranged  $334.33 \pm 2.75$  to  $857.50 \pm 9.54$  mg/100 g.

The Na content ranged from  $1.15 \pm 0.04$  to  $36.30 \pm 0.64$  mg/100g. High K content in relation to the low levels of Na in these WEPs is an important attribute nutritionally because they are the main electrolyte and major cations inside the cell. K and Na are essential for osmotic pressure and blood volume maintenance [34]. Regulation of K and Na is required to prevent hypertension both in pregnancy and school-age children. *Solanum anguivi* could meet the K RDI of pregnant women with a 90.88% score when 100g of plant material is consumed, although the K score was too high in the case of *B. alba* and *C. pepo*. A too low K score was observed for *T. microcarpus* (15.93%). For school-age children, the K IMS was low for *T. microcarpus* and *S. anguivi*. *Cucurbita pepo* K score was good (89.68 %) although *B. alba* score was too high (179.32 %). The P content of *B. alba*, *C. pepo*, and *S. anguivi* could meet the pregnancy RDI with 49.07, 66.95, and 47.76 % scores respectively. Nonetheless, *T. microcarpus* P score was way too high (122.50%). The P IMS for school-age children was low for *B. alba* (27.48%), *S. anguivi* (26.75%), and *C. pepo* (37.49%). Nonetheless, the *T. microcarpus* P IMS was good (68.60%). During pregnancy, P is needed for the growth, maintenance, and

repair of all tissues and cells, and the production of deoxyribonucleic acid and ribonucleic acid. Phosphorous is very important to school-age children due to its role in proper brain functioning, including the ability to concentrate, learn, problem-solve and remember information [35]. As most foods contain P, dietary deficiency of P is rare.

The individual mineral scores of Na, Ca, and Mg for pregnant women and school-age children were too low, thus the need to complement their diets with other foods rich in Na, Ca, and Mg to avoid deficiencies. These may include other leafy vegetables like spinach which is rich in Ca and Mg, then beetroot and smoked fish which are rich in Na. Ca is important for normal heart and muscle functions, blood clotting and pressure, and immune defences [26]. Although Ca deficiency is rare, it appears in cases of hypoparathyroidism and in individuals who are unable to eat a diet rich in dairy products. Hypertensive disorders during pregnancy are a result of low Ca and Mg concentrations. Ca inadequacy in school-age children leads to osteoporosis later in life. Mg which is a co-factor in over 300 enzymes [37] is necessary for the conversion of vitamin D into its active form. This in turn supports calcium absorption, metabolism, and parathyroid hormone function required for bone growth and development. Dietary Mg inadequacy in pregnant women causes preeclampsia, miscarriages, fetal growth retardation, and pre-term delivery. School-age children require sufficient amounts of Mg as it is a natural N-methyl-D-aspartate (NMDA) antagonist and a gamma-aminobutyric acid (GABA) agonist, both of which have a calming and relaxing effect, as well as facilitating sleep [37]. Therefore, a deficiency in Mg can increase agitation, anxiety, irritability, hyperexcitability, attention, and aggression problems in children.

Pregnancy comes with an increase in the mother's blood volume thus higher demand for Fe being the vital trace element for blood formation [36]. All the species under study had good Fe levels, *T. microcarpus* having the highest Fe content of  $517.87 \pm 11.64$  mg. The Fe IMS for pregnant women was too high for *T. microcarpus* (119 %), *C. pepo* (184.19 %), and *B. alba* (103.44 %). The Fe IMS for school-age children was too high for *B. alba* (188.72 %), *T. microcarpus* (3499.12 %), and *C. pepo* (336.01 %) yet low for *S. anguivi* (41.22 %). Iron is most important to the fetus during the third trimester because, during the last three months, the fetus is accumulating Fe for use during the first six months of life [22]. Iron deficiency in school-age children results in poor school performance. The human body has a higher absorption of heme Fe as compared to non-heme Fe obtained from plant sources, especially green leafy vegetables and fruits. Fortunately, *T. microcarpus*, *S. anguivi*, *B. alba*, and *C. pepo* contained very little amounts of

phytates which have an insignificant impact on non-heme Fe absorption in the mother's body [22].

*Cucurbita pepo* had the highest Zn content ( $10.64 \pm 0.32$  mg). The Zn IMS for pregnant women and school-age children was too low for *S. anguivi* with scores of 16.73 % and 20.44 %, respectively. Nonetheless, the Zn IMS for school-age children was too high (118.22 %) for *C. pepo*. The Zn IMS for school-age children was exceptionally high for *S. anguivi* (213.75 %), too high for *B. alba* (120.63 %) and *T. microcarpus* (118.13 %), and too low for *C. pepo* (14.38 %). Zn intake makes a good effect on the growth of stunted children and protects children from diarrhoea [37]. During pregnancy, low plasma Zn concentrations cause congenital abnormalities, abortions, intrauterine growth retardation, premature birth, and preeclampsia [37].

The IMS derived from the pregnancy RDI mg per 100 g of dry plant material showed that the Cu score was low (44 %) for *C. pepo*. Nonetheless, the Cu score for school-age children was too high (119 %) for *T. microcarpus*. Copper is a trace mineral whose role is free radical eradication, energy production, connective tissue formation, oxygen metabolism, and iron maturation of extracellular matrix [38]. Foetuses and neonates chronically deprived of Cu during gestation and early life are characterized by severe connective tissue abnormalities, skeletal defects, and lung abnormalities [38]. School-age children need sufficient Cu to promote their growth and mental health. Severe Cu deficiency results in growth and mental retardation, teratogenesis, and fetal death.

The Mn IMS for pregnant women was too high for *S. anguivi* (171 %) and too low for *C. pepo* (11.50 %). Manganese is an important co-factor for many enzymes, including the antioxidant manganese superoxide dismutase (Mn-SOD) which protects the placenta from oxidative stress by detoxifying superoxide anions. It is also necessary for normal brain and nerve function.

Mineral elements in plants have their origin mainly from soil water and root uptake. This uptake depends on the environment of the root system. When the pH level of acid soils has been raised by the addition of limestone, the concentrations of minerals related to Cation Exchange Capacity (CEC) level; Ca, Mg, K, and Na increase, while Mn decreases [39]. The use of fertilizers with high P concentrations may also depress the Mn uptake [40]. The uptake by vegetables, of elements like N, is different at low and high pH levels, respectively. Thus, N is taken up predominantly as  $\text{NO}_3^-$  ions at high soil pH and predominantly as  $\text{NH}_4^+$  in acid soils. Ammonium ions formed by the decomposition of organic matter are oxidised

to nitrate by special bacteria, forming nitric acid (nitrification). In alkaline soil, nitric acid assists the release of mineral ions, which can then be assimilated by plants. This may partly explain higher mineral values in *B. alba*, *T. microcarpus*, *C. pepo*, and *S. anguivi*.  $\text{NH}_4$  fertilisers may depress the concentrations of Ca, Mn, K, and Na in plants, since the positively charged  $\text{NH}_4$ -ions increase the positive charges inside the cells of plants, increasing the in-flow of negatively charged ions, such as Cl,  $\text{SO}_4$ ,  $\text{NO}_3$ , and  $\text{PO}_4$ .

### ***Basella alba*, *T. microcarpus*, *C. pepo* and *S. anguivi* Contain Antioxidant**

*Basella alba*, *T. microcarpus*, *C. pepo*, and *S. anguivi* contained antioxidants as presented in Table 4. Cucurbita pepo was the best antioxidant with an  $\text{IC}_{50}$  of  $11.78 \pm 0.47$  mg/ mL followed by *B. alba* ( $14.69 \pm 0.74$  mg/ mL) and least with *S. Anguivi* ( $27.94 \pm 0.52$  mg/mL). Nonetheless, no species compared with the standard ascorbic acid (0.028 mg/ mL). In another study, in the case of Cucurbita pepo, the highest antioxidant activity was  $\text{IC}_{50} = 0.12$  mg/mL [41], which was low compared to the current study. Antioxidant compounds scavenge free radicals, chelate free catalytic metals, and are electron donors [42]. The vitamin C content of the analysed species was low hanging. Thus, the individual vitamin scores for all the species were too low to meet the pregnancy RDI neither could they meet the RDI for school-age children. Vitamin C enhances Fe absorption and carnitine synthesis, and helps the body to use Ca and other nutrients to build bones and blood vessel walls, assists with collagen synthesis, enhances immune function, assists in the synthesis of hormones, neurotransmitters, and DNA [42].

The WEPs under study contained phenols and flavonoids (Table 4). *Basella alba* and *Solanum anguvi* had the highest amount of flavonoids ( $2.45 \pm 0.01$  mg/g RAE) and phenols ( $14.48 \pm 0.02$  mg/g GAE), respectively. Phenols are powerful chain-breaking antioxidants due to their hydroxyl group thus concomitant with a high reducing power [27]. Phenols work with flavonoids to reduce oxidative damage to cell membrane lipid, protein, and nucleic acid thereby protecting the mother and fetus from cardiovascular diseases, immune diseases, and brain dysfunctions. Flavonoids are anti-inflammatory, hepatoprotective, antithrombotic, antiviral, and anticarcinogenic. In pregnancy, flavonoids reduce blood lipid/obesity thereby preventing preeclampsia and gestational diabetes [43].

### **Heavy metal contamination**

Plants easily uptake and accumulate heavy metals such as Pd, Cd, and Hg which are widely distributed throughout the environment due to soil erosion, intensive industrial and agricultural processes [44]. These elements are potent metabolic poisons to plants, animals, and humans, due to the poor potential of natural

detoxification and their tendency to accumulate within the organism [45]. Higher doses of heavy metals may cause metabolic disorders, and growth inhibition for most plants, thus reducing food quality [46]. *Basella alba*, *T. microcarpus*, *C. pepo*, and *S. anguivi* contained heavy metals Pb and Cd in low quantities (Table 4). The concentration of both metals was below FAO/WHO permissible limits of 0.21mg/100g and 0.43mg/100g in edibles, respectively, and for most foods (0.1-2 mg/kg) [28], which shows that the Pb content in the analysed species was below tolerable limits. *Termitomyces microcarpus* had the highest Cd content ( $0.12 \pm 0.01$  mg/100 g) and *B. alba* higher in Pb content ( $1.26 \pm 0.34$  mg/100 g). Cd is the seventh most toxic heavy metal which may be absorbed from the environment. Cd concentration increases 3000-fold when it binds to metallothionein. In the liver, the cystein-metallothionein complex causes hepatotoxicity, and then it circulates to the kidney and gets accumulated in the renal tissue causing nephrotoxicity. Acute Pb poisoning causes loss of appetite, headache, hypertension, renal dysfunction, fatigue, sleeplessness, arthritis, hallucinations and vertigo in both children and adults [36]. Chronic Pb exposure results in mental retardation, cancer, birth defects, psychosis, autism, allergies, dyslexia, weight loss, hyperactivity, paralysis, muscular weakness, brain damage, kidney damage, and may even cause death [36]. Although Pb poisoning is preventable it remains a dangerous disease that can affect most organs.

Accumulation of heavy metals in plants is a consequence of irrigation with contaminated water, the addition of fertilizers and metal-based pesticides, Industrial emissions, transportation, harvesting process, and activities during storage and/or sale [45]. The low levels of heavy metals in the analysed species may be explained by the nature of their habitats. These species were picked from the forest which may contain low levels of these elements in the soil due to low levels of contamination by human activities. High contents of these elements in soil are supposed to increase the risk of uptake by plants, considering the positive correlation between the metal content in soils and plants [47]. Therefore, soil quality standards are important instruments for risk assessment of polluted soils and their impact on human health, water resources, and other environmental impacts [48].

### **Antinutrients in *Basella alba*, *T. microcarpus*, *C. pepo*, and *S. anguivi***

All the analysed species contained antinutrients tannins and phytates.

*Termitomyces microcarpus* had the highest tannin content ( $0.42 \pm 0.00$  (mg/g GAE) and *B. alba* had the highest phytate content ( $0.72 \pm 0.08$  g/100g). However, the levels of tannins and phytates were found to be low in some wild edible plants (flowers, fruits, and leaves), ranging from  $0.002 \pm 0.0005$  to  $0.1110 \pm 0.006$  and

$0.02 \pm 0.005$  to  $0.12 \pm 0.02$ , respectively [49]. Antinutrients are widely distributed in WEPs and are responsible for several health complications. Nonetheless, some antinutrients are widely applied in nutrition and as pharmacologically active agents [49]. Phytates affect the bioavailability of minerals (Ca, Zn, Mn, Fe, and Mg) and have a strong effect on infants, pregnant and lactating women. Due to their chelating property, phytates are considered the most effective anti-nutrients in foods, and a cause of rickets and osteomalacia in children and adults, respectively. A cross-sectional study on the effect of dietary phytate on calcium, iron, and zinc bioavailability from the diets of economically vulnerable pregnant women [50] revealed the severe adverse effect of phytates on the bioavailability of the metals in question. Tannins form tannin-protein complexes which cause the inactivation of digestive enzymes and elicit 'Kwashiorkor'. Therefore, reduction in the antinutrients of foods is of great interest. Dietary diversification together with the different traditional household methods and technological processing ways such as cooking, soaking, milling, fermentation, roasting, and germination may reduce the antinutrients in foods, thereby significantly improving the mineral bioavailability to some degree improve while relating to your work.

### Mineral bioavailability and molar ratios

Table 5 shows the molar ratios of Phytates to minerals. Phytates: Zn, Phytates: Ca, Phytates: Fe and Phytates, Ca: Zn in the studied WEPs. Phytate: Ca molar ratios ranged from 0.001 to 0.004. The molar ratio of phytate:Ca less than 0.24 suggests adequate calcium bioavailability in food [51]. It is low for other foods studied elsewhere [52] that gave a ratio of phytate:Ca of oat as  $4.00 \pm 0.60$  (3.24–5.06). The molar ratio of phytate: Zn is the strong predictor of zinc bioavailability when compared to total phytate. The molar ratio of Phytate:Zn varied from 0.01 to 0.004, lower than the critical value of  $>15$ , indicating high bioavailability of zinc in these WEPs [51]. The ratio of phytate: Zn in this study is much lower than the molar ratio phytate: Zn of cereals such as barley yellow corn (14.6), (24) and oats (82.4) [52]. The phytate:Fe molar ratios ranged from 0 to 0.006. This value is below 1, which suggests a decrease in inhibition of Fe absorption by the phytates [51]. This ratio of phytates: Fe is much lower than those reported for cereals ranging from 2.6 for barley to oats (49.2) [52]. The Ca levels have a significant impact on the absorption of zinc when phytates are consumed in excess. Therefore, the  $\{\text{phytates}\}\{\text{Ca}\}/\{\text{Zn}\}$  molar ratios may be a more reliable predictor for the Zn bioavailability than the phytate: Zn molar ratio alone [51]. The  $\{\text{phytates}\}\{\text{Ca}\}/\{\text{Zn}\}$  molar ratio ranged from 0 to 0.011, a ratio lower than 0.5 mol/kg required for the high levels of Ca in foods to decrease the zinc bioavailability induced by high phytates [52]. The  $\{\text{phytates}\}\{\text{Ca}\}/\{\text{Zn}\}$  ratios in this study are much lower than that reported for cereals, corn (18.4), barley (140), and oats (820) [52]. Therefore, the phytate

contents of studied WEPs do not affect the bioavailability of Fe, Zn and Ca, and Zn diet. This is in agreement with other studies undertaken elsewhere that support the very low levels of phytate content of tubers and roots, fruits, and vegetables compared to cereals [53].

## CONCLUSION

Like in many rural areas of the world, wild edible plants have a major contribution to the dietary intake of pregnant women and school-age children in rural Uganda. They have the potential to greatly improve food security by providing alternative sources of affordable and nutritious food with the added advantage of being available all year round and being able to grow in water-stressed areas and diverse environmental conditions. Biochemical knowledge of the nutraceutical significance of wild edible plants is of crucial importance to evaluate the health benefits and physiological effects of these species to develop clinical investigations concerning their mechanisms of action, safety, and efficacy. In this study, *B. alba*, *T. microcarpus*, *C. pepo*, and *S. anguivi* were envisaged as rich sources of macronutrients, trace and major minerals, and antioxidant micronutrients. The phytochemical and nutritional profiles of these species can therefore constitute basic knowledge for food pairing with other ingredients to improve nutritional and/or sensory quality and to find innovative cooking methods, allowing key molecules responsible for functional properties to be enhanced. Fortunately, *B. alba*, *T. microcarpus*, *C. pepo*, and *S. anguivi* had low quantities of antinutrients and heavy metals. Therefore, these minimal amounts can be eliminated during processing and preparation making the species antinutrient free. The limitations of this study revolved around limited funds. For future studies, there is a need to conduct analyses on other important mineral elements, the bioavailability of the nutrients in vivo, and the effect of the different processing methods on the nutrients. Additionally, the nutraceutical significance of more wild edible plants ought to be investigated.

## FUNDING STATEMENT

This work was funded by the Government of Uganda through the Makerere University Research and Innovation Fund grant number RIF1/COVAB/013.

## ACKNOWLEDGEMENTS

Authors are thankful to the Government of Uganda through the Makerere University Research and Innovation Fund for funding the research (Grant number RIF1/COVAB/013). Authors also express their heartfelt gratitude to all the persons who made this work a success: Chairman Magomu of Mubango village, Najjembe



sub-county, Buikwe district, Research assistants who helped in collecting field data, Mr. Fred who drove the study team to and fro to Buikwe district and Mr. Mukamanasasira Godman who helped in the daily running of project activities.

**Table 1: Proximate composition of *B. alba*, *T. microcarpus*, *C. pepo* and *S. anguivi***

Nutritional Parameters	Proximate composition (%)			
	<i>B. alba</i>	<i>T. microcarpus</i>	<i>C. pepo</i>	<i>S. anguivi</i>
Crude fibre	7.19±0.21 <sup>c</sup>	4.07±0.28 <sup>d</sup>	13.72±0.86 <sup>b</sup>	25.22±0.08 <sup>a</sup>
Crude Protein	20.77±0.17 <sup>b</sup>	21.99±0.16 <sup>a</sup>	11.85±0.40 <sup>b</sup>	14.27±0.46 <sup>c</sup>
Moisture Content	9.08±0.08 <sup>a</sup>	10.71±0.35 <sup>b</sup>	8.61±0.02 <sup>c</sup>	8.10±0.19 <sup>d</sup>
Total Fat	2.19±0.03 <sup>a</sup>	2.29±0.04 <sup>b</sup>	0.84±0.03 <sup>c</sup>	6.55±0.05 <sup>d</sup>
Ash	19.39±0.01 <sup>d</sup>	17.08±0.11 <sup>a</sup>	9.56±0.27 <sup>c</sup>	7.24±0.04 <sup>b</sup>
Total carbohydrates	48.57±0.23 <sup>d</sup>	47.94±0.65 <sup>b</sup>	69.15±0.62 <sup>c</sup>	63.86±0.64 <sup>a</sup>
Energy (Kcal/ 100 g)	297.10±0.53 <sup>a</sup>	300.28±1.67 <sup>b</sup>	331.55±1.12 <sup>c</sup>	371.37±1.17 <sup>d</sup>

All values are means of three determinations ± Standard deviation (SD). Values with different alphabetical letters/superscripts in a row are significantly different (p>0.05)

**Table 2: Major minerals and Individual Mineral Scores of *B. alba*, *T. microcarpus*, *C. pepo* and *S. anguivi***

Species	Mineral content (mg/100 g) and Individual Mineral Scores (%)														
	Sodium	IMSP	IMSC	Potassium	IMSP	IMSC	Calcium	IMSP	IMSC	Phosphorous	IMSP	IMSC	Magnesium	IMSP	IMSC
<i>B. alba</i>	5.59±0.11 <sup>a</sup>	0.24	0.41	3944.99±15.25 <sup>a</sup>	563.57	179.32	47.09±2.22 <sup>a</sup>	4.71	3.62	343.50±3.00 <sup>a</sup>	49.07	27.48	4.93±0.05 <sup>a</sup>	1.37	2.05
<i>T. microcarpus</i>	36.30±0.64 <sup>a</sup>	1.58	2.63	111.48±8.49 <sup>a</sup>	15.93	5.07	0.92±0.10 <sup>b</sup>	0.09	0.07	857.50±9.54 <sup>b</sup>	122.50	68.60	4.29±0.05 <sup>a</sup>	1.19	1.79
<i>C. pepo</i>	1.15±0.04 <sup>b</sup>	0.05	0.08	1973.05±11.47 <sup>a</sup>	281.86	89.68	0.79±0.11 <sup>b</sup>	0.08	0.06	468.67±6.53 <sup>a</sup>	66.95	37.49	4.38±0.02 <sup>a</sup>	1.22	1.83
<i>S. anguivi</i>	5.85±0.14 <sup>a</sup>	0.25	0.42	636.14±2.70 <sup>b</sup>	90.88	28.92	10.35±0.85 <sup>b</sup>	1.04	0.80	334.33±2.75 <sup>a</sup>	47.76	26.75	3.92±3.92 <sup>a</sup>	1.09	1.63

IMSP = Individual mineral score of a species measured at recommended daily intake for physiological needs of pregnant women; IMSC = Individual mineral score of a species measured at recommended daily intake for physiological needs of school-age children (6-12 years). All values are means of three determinations ± Standard deviation (SD). Values with different alphabetical numbers in a column are significantly different (p>0.05)



**Table 3: Trace minerals, Vitamin C, and Individual Mineral/ Vitamin scores of *B. alba*, *T. microcarpus*, *C. pepo*, and *S. anguivi***

Species	Mineral and Vitamin C content (mg/100 g) and Individual Mineral Scores (%)															
	Copper	IMS <sup>P</sup>	IMS <sup>C</sup>	Iron	IMS <sup>P</sup>	IMS <sup>C</sup>	Zinc	IMS <sup>P</sup>	IMS <sup>C</sup>	Manganese	IMS <sup>P</sup>	IMS <sup>C</sup>	Selenium	Vitamin C	IVS <sup>P</sup>	IVS <sup>C</sup>
<i>B. alba</i>	0.83±0.01 <sup>a</sup>	55.33	83.00	27.93±2.32 <sup>a</sup>	103.44	188.72	7.33±0.08 <sup>b</sup>	66.64	81.44	1.93±0.08 <sup>a</sup>	96.50	120.63	ND	5.38±0.18 <sup>a</sup>	6.33	17.93
<i>T. microcarpus</i>	1.19±0.01 <sup>a</sup>	79.33	119.00	517.87±11.64 <sup>b</sup>	1918.04	3 499.12	5.87±0.03 <sup>c</sup>	53.36	65.22	1.89±0.08 <sup>a</sup>	94.50	118.13	ND	7.38±0.53 <sup>a</sup>	8.68	24.60
<i>C. pepo</i>	0.66±0.03 <sup>a</sup>	44.00	66.00	49.73±0.99 <sup>a</sup>	184.19	336.01	10.64±0.32 <sup>a</sup>	96.73	118.22	0.23±0.03 <sup>b</sup>	11.50	14.38	ND	5.00±0.35 <sup>a</sup>	5.88	16.67
<i>S. anguivi</i>	1.00±0.02 <sup>a</sup>	66.67	100.00	6.10±0.07 <sup>a</sup>	22.59	41.22	1.84±0.02 <sup>c</sup>	16.73	20.44	3.42±0.18 <sup>c</sup>	171.00	213.75	ND	5.75±0.00 <sup>a</sup>	6.76	19.17

**IMS<sup>P</sup>** = Individual mineral score of a species measured at recommended daily intake for physiological needs of pregnant women: **IMS<sup>C</sup>** = Individual mineral score of a species measured at recommended daily intake for physiological needs of school-age children 6-12 years. **IVS<sup>P</sup>** = Individual vitamin score of a species measured at recommended daily intake for physiological needs of pregnant women: **IVS<sup>C</sup>** = Individual vitamin score of a species (measured at recommended daily intake for physiological needs of school-age children 6-12 years. All values are means of three determinations ± Standard deviation (SD). Values with different alphabetical numbers in a column, are significantly different (p>0.05). Values with different alphabetical numbers in a column, are significantly different (p>0.05)

**Table 4: Antioxidant Micronutrients, Antinutrients and Heavy metals in *B. alba*, *T. microcarpus*, *C. pepo* and *S. anguivi***

**Antioxidant Micronutrients, Antinutrients and Heavy metals**

Species	Total antioxidants IC <sub>50</sub> (mg/ mL)	Total Flavonoids (mg/g RAE)	Total Phenols (mg/g GAE)	Total tannins (mg/g GAE)	Phytate content (g/100g)	Cadmium (mg/100 g)	Lead (mg/100 g)
<i>B. alba</i>	14.69±0.74 <sup>a</sup>	2.45±0.01 <sup>b</sup>	6.30±0.07 <sup>c</sup>	0.08±0.01 <sup>d</sup>	0.72±0.08 <sup>e</sup>	0.07±0.02 <sup>f</sup>	1.26±0.34 <sup>g</sup>
<i>T. microcarpus</i>	22.59±10.44 <sup>a</sup>	1.76±0.02 <sup>b</sup>	8.95±0.11 <sup>c</sup>	0.42±0.00 <sup>d</sup>	0.55±0.01 <sup>e</sup>	0.12±0.01 <sup>f</sup>	0.12±0.03 <sup>h</sup>
<i>C. pepo</i>	11.78±0.47 <sup>a</sup>	0.27±0.02 <sup>b</sup>	8.29±0.30 <sup>c</sup>	0.14±0.01 <sup>d</sup>	0.42±0.04 <sup>e</sup>	0.05±0.00 <sup>f</sup>	0.05±0.01 <sup>h</sup>
<i>S. anguivi</i>	27.94±0.52 <sup>a</sup>	0.30±0.01 <sup>b</sup>	14.48±0.02 <sup>c</sup>	0.40±0.02 <sup>d</sup>	0.40±0.01 <sup>e</sup>	0.06±0.00 <sup>f</sup>	0.10±0.08 <sup>h</sup>

All values are means of two determinations ± Standard deviation (SD) (n=2); Standard antioxidant - Ascorbic acid (0.028 mg/ mL); mg/g RAE = milligrams of retinol activity equivalents; mg/g GAE = milligrams of gallic acid equivalents per 100 g. Values with different alphabetical numbers in a column, are significantly different (p>0.05)

**Table 5 Calculated mean molar ratios of phytates to minerals (Ca, Fe, Zn) of *B. alba*, *T. microcarpus*, *C. pepo* and *S. anguivi***

Species	{Phy}/{Zn} <sup>1</sup>	{Phy}/{Ca} <sup>2</sup>	{Phy}/{Fe} <sup>3</sup>	{Phy: Ca}/{Zn} <sup>4</sup>
<i>B. alba</i>	0.01	0.001	0.002	0.01
<i>T. microcarpus</i>	0.01	0.004	.000	.000
<i>C. pepo</i>	0.004	0.005	0.004	0.005
<i>S. anguivi</i>	0.03	0.002	0.006	0.007

Notes: <sup>1</sup>mg of phytates/MW of phytates:mg of zinc/MW of zinc <sup>2</sup>mg of phytates/MW of phytates:mg of calcium/MW of calcium; <sup>3</sup>mg of phytates/MW of phytates:mg of iron/MW of iron; <sup>4</sup>(mg of calcium/MW of calcium)\*(mg of phytates/MW of phytates)/(mg of zinc/MW of zinc)



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