

## Original Research Article

## Biochemical Characterization of Four Leafy Vegetables Collected in the Northern Zone of Brazzaville (Republic of Congo)

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**Abstract:** The present study involved the biochemical characterization of four leafy vegetables (*Basella alba*, *Hibiscus sabdariffa*, *Solanum macrocarpon* and *Lagenaria siceraria*) selected and collected from two markets (Texaco and Thomas Sankara) and two market gardening sites (Jardins Talangaï and Nkombo) in the northern part of the city of Brazzaville, in Congo. The water content and pH of the selected leafy vegetables in the four sites were determined from the fresh material, while their protein, lipid, ash and mineral contents were established from the dry material using standard methods. The results reveal that the studied leafy vegetables have a very high-water content ranging from 84.49±0.36-94.63±0.68%, a pH ranging from 2.7±0.6 to 7.3±0.6, a lipid content ranging from 2.09±0.74 to 5.02±0.42%, a protein content within the range of 14.87±0.81% and 35.81±1.85%, and an ash content varying between 6.86±0.53 and 41.59±1.62%. Furthermore, these results show a high content of mineral elements, with values ranging from 3.94±0.95 to 83.57±3.7mg/100g, 920.01±0.45 to 4677.99±2.24mg/100, 101.25±1.02 to 372.80±1.56mg/100g, and 192.28±1.73 to 1452.57±4.71 mg/100g, respectively for iron, calcium, phosphorus and magnesium. In accordance to these results the studied leafy vegetables available and accessible, are rich sources of protein and micronutrients. Thus, their consumption in sufficient quantities could contribute to the improvement of nutritional status and an adequate protection against malnutrition-related diseases.

**Keyword:** Biochemical characterization, Republic of Congo, leafy vegetables, *Basella alba*, *Solanum macrocarpum*, *Hibiscus sabdariffa*, *Lagenaria siceraria*, mineral elements.

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## INTRODUCTION

Unbalanced diet is one of the primary causes of multi-calorie malnutrition, i.e.. protein-calorie malnutrition or micronutrients deficiencies, also known as hidden hunger and increased the risk of non-communicable diseases (NCDs) such as high blood pressure, cardiovascular disease and diabetes [1]. Micronutrient deficiency is caused by poor dietary intake of calcium, iron, zinc, potassium, magnesium, Iodine, copper and selenium [2]. This situation constitutes a public health problem in the world and particularly in developing countries, resulting in high morbidity and mortality [3]. For example, according to the World Health Organization [4], iron, vitamin A and zinc deficiencies are among the top ten causes of death in developing countries. They more often affect

vulnerable people such as children, pregnant women and the elderly [5].

In the Republic of Congo, there is also the problem of multi-carbass malnutrition among its population. Indeed, taking into account national surveys, nearly a third of the population shows signs of chronic malnutrition, and the country's food and nutrition situation remain worrying despite its natural assets: 26% of children under 5 years of age suffer from chronic malnutrition, while 39% of households are unable to cover their food needs and they do not have access to the minimum calorie intake of 2.400 calories per day [6]. Their consumption ensures intake of various essential vitamins and mineral elements thus avoiding the problem of malnutrition.

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To remedy this situation, it therefore seems essential to encourage the consumption of food resources such as vegetables, known to be the main sources of micronutrients such as minerals, vitamins and antioxidants [7-9]. Due to their composition, the consumption of vegetables is recommended to ensure a balanced diet for children and adults as well as for the elderly [10-12]. Indeed, the mineral elements in vegetables are essential for the growth of children and the strengthening of bone mass in the elderly. In addition, the World Health Organization recommends a minimum of 400 g of fruit and vegetables for the prevention of chronic diseases such as heart disease, cancer, diabetes and obesity [4]. Unfortunately, nowadays, attention and resources are focused on a limited number of species of commercial interest, including the most 'sophisticated' Western vegetables [13], which are still inaccessible to the middle class due to their high cost. However, Sub-Saharan Africa in general, and the Republic of Congo in particular, are endowed with several species of traditional leafy vegetables that could be exploited in the diet and help solve certain problems of malnutrition or micro and macro-nutrient deficiencies, but regrettably these vegetables are generally neglected by most urban populations because of their rural origin and status of "food of the poor". They provide 10 to 100 times more micronutrients than salad, cabbage or leek [13]. These traditional leafy vegetables are generally a source of micronutrients [1, 14], addressing challenges related to nutritional deficiencies. They play an important role in the diets of local populations, particularly in Africa, by providing the essential part of nutritional and medicinal needs [15, 16]. Their crucial role as indispensable ingredients in traditional sauces has been demonstrated through the success of the World Health Organization's

(WHO) Global Fruit and Vegetable Initiative project [17]. Traditional leafy vegetables play a vital role in combatting hunger, food insecurity, and malnutrition and most are suitable for food intervention programs [2].

Despite the advantages of these traditional leafy vegetables, very few studies on their chemical characterization have been carried out up now in the Republic of Congo. It is with the aim of providing information on their chemical composition that we have conducted this study. We were interested in four (04) leafy vegetables: *Basella alba* (*B.alba*) or spinach, *Solanum macrocarpon* (*S. macrocarpon*), *Hibiscus sabdariffa* (*H. sabdariffa*) and *Lagenaria siceraria* (*L. siceraria*), produced and sold in the northern area of Brazzaville, in order to determine the contents of some macro- and microelements.



## MATERIALS AND METHODS



### Material

#### Biological material

The plant material used for this present study consists of leafy vegetables of *Basella alba* L., *Hibiscus sabdariffa* L., *Solanum macrocarpon* L. and *Lagenaria siceraria* (Molina) standl presented in Table 1. They were, on the one hand, harvested from two market garden sites (Talangai and Nkombo) and, on the other hand, purchased at the two markets (Texaco and Thomas Sankara) in the northern part of Brazzaville. The identical varieties were put together to form a pool for the site or market. The samples were put in airtight containers before being taken to the laboratory for analysis.

**Table 1: The different vegetables used for analyses.**

Commun name	Popular name	Systematic name	Organs used	Pictures of vegetables
Spinach	UDJINI	<i>Basella alba</i> L.	Leaves	
Oseille	NGAI-NGAI	<i>Hibiscus sabdariffa</i> L.	Leaves	

African eggplant leaves	NKENKA	<i>Solanum macrocarpon</i> L.	Leaves	
<i>Lagenaria</i> leaves	NTIYA	<i>Lagenaria siceraria</i> (Molina) standl.	Leaves	

**Sample processing**

The different vegetables collected were sorted, carefully washed with tap water, drained, weighed using a precision balance and then oven dried at a temperature of 70°C for 72 hours until their mass had stabilized. At the end of the oven drying, the dried vegetables were crushed employing a porcelain mortar, and the powder obtained was stored before the various chemical analyses.

**Chemical analyses**

**Determination of pH**

The pH of each leafy vegetable sample was determined with a digital pH meter. Ten (10) g of vegetable sample were crushed in 100 mL of distilled water, the ground crushed material was centrifuged for 30 min. the mixture was filtered, and then filtrate was collected in an Erlenmeyer. Direct pH reading was taken on the filtrate with pH paper.

**Determination of moisture content**

The moisture content of the different samples was determined using the standard AOAC methods [18, 19], which is based on the principle of drying the samples by steaming. Thirty (30) grams of each leafy vegetables sample collected were weighed using a precision balance (OHAUS) in a petri dish of known mass (M0) in three repetitions. The whole (petri dish and leafy vegetables) was placed in an oven (SELECTA) at a temperature of 60°C for 72 hours until the mass stabilized. After cooling, the petri dish containing the dried sample was weighed and the mass of the dried sample was determined (Md), the moisture content was determined from the following formula:

$$\text{Moisture content (\%)} = \frac{Mf - Md}{Mf} * 100$$

Mf: mass of the fresh sample (g), Md: mass of dried sample (g)

**Determination of protein content**

The protein content was determined from the determination of total nitrogen in the dry matter, according to the Kjeldhal method [18, 19]. In a Matra, 0.5 (g) gram of previously dried and ground sample (M) was introduced, and then in turn 10 mL of concentrated sulfuric acid, a tip of mineralization a selenium catalyst and some glass beads were added. The mixture was mineralized cold for 30 minutes, and then hot for 2 hours at a temperature of 420 °C. After cooling, 20 mL of distilled water and 30 mL of sodium hydroxide were added. The vapor was drawn off by collecting the distillate in a 150 mL Erlenmeyer flask containing 20 mL boric acid and a few drops of color indicator. Titration of the nitrogen was carried out with N/20 sulfuric acid until the indicator turned from green to pink. The nitrogen percent was determined by the following formula:

$$\% N = \frac{VH_2SO_4 \times 0.07}{M}$$

VH<sub>2</sub>SO<sub>4</sub>: sulfuric acid volume, M: test sample (0.5g)

The percentage of crude proteins was calculated by multiplying the percent nitrogen with conversion factor 6.25.

**Determination of lipid content**

The lipid content was determined using a Soxhlet extractor [18, 19]. Into the cleaned WHATMAN cartridge, 30g (M) of dried sample wrapped in filter paper was introduced. This cartridge was placed in a Soxhlet type extractor and connected to the refrigerator. In a previously weighed 250 mL flask (Mo), 200 mL of n-hexane was placed and, on this flask, the Soxhlet extractor with a cooler was positioned and the whole was placed on a flask heater. After several continuous siphoning, a mixture of oil and solvent was obtained. The extracted oil from each sample was separated from the solvent and traces of the solvent were removed by drying in an oven at 60 °C for

10 minutes. After cooling, the flask containing the oil was weighed (M1). The lipid percentage was determined by the following formula:

$$\% \text{Lipid} = \frac{M1 - M0}{M} \times 100$$

M0: mass of the empty balloon, M1: mass of the balloon with oil, M: mass of the dried sample

#### Determination of ash content

The ash content of the different samples was determined using the standard AOAC methods [18, 19]. 5g of each sample were placed of pre-weighed crucibles (M1). The whole set was incinerated in an electrically heated muffle furnace at a temperature of 550°C until a constant mass was obtained for 8 hours. After removing the crucibles from the muffle furnace and cooling, it's were weighed again (M2). The ash content was determined by the following formula:

$$\text{Ash (\%)} = \frac{M2 - M0}{M1 - M0} \times 100$$

M0: mass in grams of the empty crucible, M1: mass in grams of the incineration crucible containing the test sample, M2: mass in grams of the incineration crucible charged with the ash.

#### Determination of mineral content

After mineralization of the different samples in the oven at 450°C, the ash was recovered, moistened with water and concentrated hydrochloric acid, and then minerals elements were determined using standard methods [20].

#### Determination Phosphorus (P) content

Phosphorus was assayed by the cold colorimetric method using Murphy and Riley's reagent (Martin-Prevel *et al.*, 1987) [20].

Murphy and Riley's reagent was obtained by adding solution A and B. Solution A was prepared by mixing 50 mL of water and 10 mL of concentrated sulfuric acid in a 100 mL vial. After cooling, 0.6 g of ammonium molybdate and 0.014 g of potassium antimonyl tartrate were added and the flask was then rounded up to the mark. As for solution B, 2 g of ascorbic acid, 50 ml of distilled water and 5 ml of concentrated hydrochloric acid were mixed, and the mixture was supplemented up to the mark of gauge. In a 200 mL vial, A and B solutions were mixed to obtain Murphy and Riley's reagent. In a plastic pill box, 0.5 mL of mineralized sample, 10 mL of distilled water and

3 mL of Riley reagent were mixed. After 30 minutes of incubation, the absorbance of the mixture was read at the wavelength ( $\lambda$ ) of 660 nm, the result of the sample is obtained by calculation taking into account a phosphorus range curve.

#### Determination iron (Fe) content

The iron content was determined using colorimetric Spectro colorimeter [20]. Iron was determined after mixing in a plastic pillbox, 5 mL of mineralized sample, 5 mL of 1% hydroxylamine chloride, 2 mL of 3% sodium citrate, 2 mL of sodium acetate buffer solution, pH 3.5 and 2 mL of 0.2% orthophenantroline. Under the same conditions, an iron standard was prepared. After 30 minutes of incubation at room temperature, the absorbance of the reaction medium was measured utilizing the spectrophotometer at 490 nm.

#### Determination calcium (Ca) and magnesium (Mg) content

The calcium and magnesium content were determined using the complexometric method [20].

Calcium and magnesium form soluble Ca-EDTA and Mg-EDTA complexes with EDTA (ethylene diamine tetraacetic acid). 5 mL of mineralized sample was introduced into a 200 mL Erlenmeyer flask, and then 45 mL of distilled water and 4 mL of buffer solution at pH 10 were added to the mixture. The titration was done with EDTA and NET as color indicators until the solution turned from pink to blue, Ca+Mg were thus determined simultaneously.

In addition, the calcium was determined by placing 5 mL of mineralized sample in a 200 mL Erlenmeyer flask, and then 45 mL of distilled water, 1 mL of KCN, 5 mL of triethanolamine hydrochloride, and 5 mL 2.5N of NaOH were added to the solution. The pH of the solution was increased to 12 to precipitate the Magnesium, and calcein (calcon) was used as a color indicator for the titration. Magnesium is calculated by difference of the results of the (Ca+Mg) and Ca.

#### Statistical Analysis

The analysis of variance (ANOVA) was carried out to compare the means, using SPSS Statistics 22.0 software. In the event of a significant difference, the Newman-Keuls and Tukey tests were used to identify the means responsible for the observed difference at the 5% threshold.

## RESULTS AND DISCUSSION

#### Hydrogen potential (pH)

The table 2 illustrates the values for pH of four leafy vegetables collected from four sites.

**Table 2: pH of leafy-vegetables in four sites**

vegetables	Sites			
	Talangai garden	Texaco market	Nkombo garden	Thomas Sankara market
<i>Basella alba L.</i>	6.7±0.6 <sup>ab</sup>	7.3±0.6 <sup>ab</sup>	6.7±0.6 <sup>ab</sup>	7.3±0.6 <sup>ab</sup>
<i>Solanum macrocarpum L.</i>	6.7±0.6 <sup>ab</sup>	7.0±0.0 <sup>ab</sup>	7.0±0.0 <sup>ab</sup>	6.7±0.6 <sup>ab</sup>
<i>Hibiscus sabdariffa L.</i>	2.7±0.6 <sup>a</sup>	3.0±0.0 <sup>a</sup>	2.7±0.6 <sup>a</sup>	3.0±0.0 <sup>a</sup>
<i>Lagenaria siceraria</i>	7.0±0.0 <sup>ab</sup>	7.0±0.0 <sup>ab</sup>	7.0±0.0 <sup>ab</sup>	6.7±0.6 <sup>ab</sup>

When, for the same lines, the superscript letters are identical, the averages are identical (at the 5% threshold). Otherwise, the averages are significantly different (at the 5% threshold).

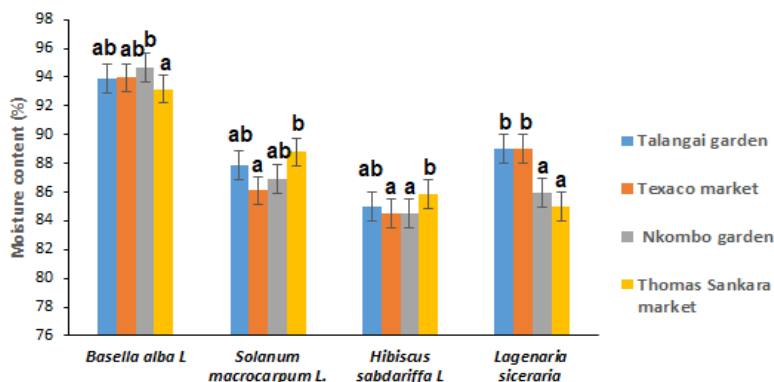
These results show that *Hibiscus sabdariffa L* leaves had statistically identical acidic pH (P-value>0.05) in all sites. which ranged from 2.7±0.6 to 3±0.0. On the other hand. the pH of the leaves of *Basella alba*. *Solanum macrocarpon* and *Lagenaria siceraria* (Molina) standl is close to neutrality with respective values of 6.7±0.6 to 7.3±0.6; 6.7±0.6 to 7±0.6 and 6.7±0.6 to 7±0.6; statistically the same (P-value>0.05) from one site to another. The pH is a determining factor for the development of microorganisms. Thus, the pH of *Basella alba*, *Solanum macrocarpon* and *Lagenaria siceraria*, close to neutrality could favor the growth of microorganisms. This would limit the shelf life or storage of these leafy vegetables. Indeed, most microorganisms grow at neutral Ph [21]. On the other hand, the acidic pH of the leaves of *Hibiscus sabdariffa L* could be a brake on their multiplication and extend their shelf life. The results obtained in this study, on *Basella alba*. *Solanum macrocarpon L* and *Lagenaria siceraria* (Molina) standl are almost similar to those on *Lactuca sativa* (6.64 ± 0.78 to 6.89 ± 0.98), on spinach (6.22), *Ipomea batatas* (6.56) and *cassava leaves* (6.37), on *Solanum macrocarpon L.* (6-8). and on four leafy vegetables consumed in northern Côte d'Ivoire (5.98 ± 0.02- 6.53 ± 0.06) [22-25]. The pH value of *Hibiscus sabdariffa* (2.7±0.6) in the present study is close to the value obtained by [25] for the same species, i.e., 2.45 ± 0.02.

**Moisture of content**

Mean values for moisture content of the four selected fresh leafy vegetables are presented on

Figure1. The analysis of this figure indicates that the investigated leafy vegetables have high moisture contents at all sites and the values between 84.49± 0.35 (*Hibiscus sabdariffa L.* from Nkombo garden) and 94.63±0.68% (*Basella alba L.* from Nkombo garden), which is usual for leafy vegetables [26]. The high moisture levels indicate that the studied leafy vegetables require appropriate storage as they would be susceptible to deterioration [27]. Indeed, high moisture content may induce a greater activity, on the one hand, of soluble active substances like enzymes and. on the other hand, of microorganisms, which make the vegetables perishable leading to postharvest losses [28].

The moisture contents of *Basella alba L* from Nkombo garden (94.63±0.68%). Texaco market (93.95±0.14) and Talangai garden (93.92±0.33%) were higher and statistically identical (P-value>0.05). These values were similar to the ones found by [29] with *Basella alba* leaves (91.26%) from Burundi and by [30] with *Spinacia oleracea* leaves (92.1%). As for *Solanum macrocarpon L* leaves, the moisture contents of the samples from the Thomas Sankara market (88.78%), the market garden sites of Talangai (87.86%) and Nkombo (86.93%) were higher and they showed no significant difference (P-value>0.05). These values were close to these obtained by [28] with *Solanum macrocarpon* leaves consumed in the Republic of Benin, which ranged from 86.66±1.21% to 87.52±1.21%.



**Figure 1: Moisture content of four leafy vegetables from four different sites (Talangai garden. Texaco market. Nkombo garden and Thomas Sankara market)**

For the same vegetable. When the letters above the bars are identical, the averages are identical (at the 5% threshold). Otherwise, the averages are significantly different (at the 5% threshold).

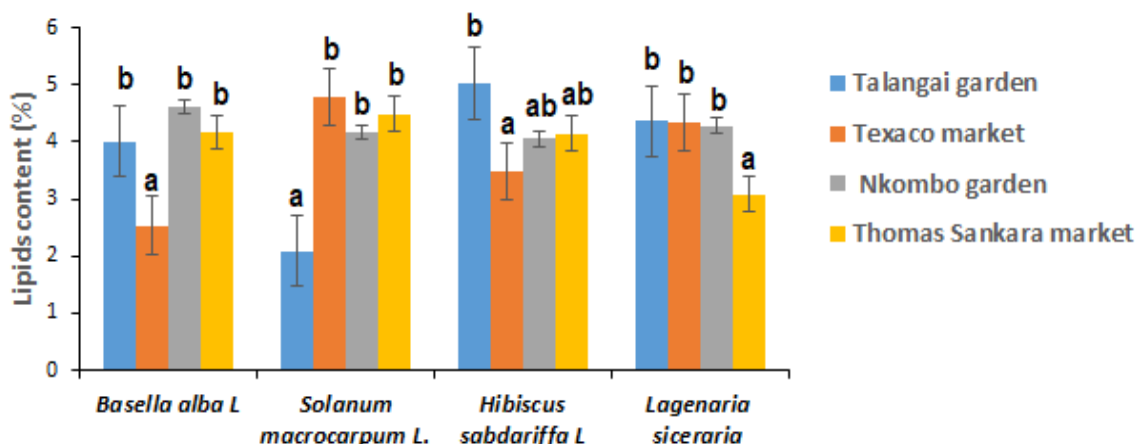
The moisture content of *Hibiscus sabdariffa* leaves collected at the Thomas Sankara market (85.89±0.78%) was higher than those of the samples from the Talangai garden. Texaco market and Nkombo garden, with respective values of 84.97±0.46; 84.49±0.36 and 84.52±0.27%, with no significant difference at the α=0.05 threshold. These values are close to those of the leaves of the red and green varieties of *H. sabdariffa*, which are respectively 85 ± 0.5% and 82.5 ± 1.2% obtained by [31]. The moisture contents of *Lagenaria siceraria* leaves from Talangai garden and Texaco market were higher, with the same values (88.98%). These values are statistically different (P-value < 0.05) from the results obtained with samples from the Nkombo garden (85.97%) and the Thomas Sankara market (84.98%). These values are very close to the work by [32] on *Lagenaria siceraria* leaves (87.07 ± 0.52%). All the results of this study are in the same order as in the investigations of on twelve (12) wild leafy vegetables used in Thailand with values comprised between 62.9 ± 2.6 and 95.3 ± 1.0% [33].

### Lipids content

Figure 2 shows the lipid contents of four leafy vegetables. The analysis of this figure reveals that the four vegetables collected in four sites have a significant average lipid content, which varies from 2.09±0.74%

(*Solanum macrocarpum* L. from the Talangai garden) to 5.02±0.42% (*Hibiscus sabdariffa* L. from the Talangai garden), with different contents from one site to another.

The lipid contents of *Basella alba* leaves collected from Talangai (4.01± 0.71%). Nkombo (4.62± 0.42%) and Thomas Sankara market (4.17±0.71%) gardens are high and statistically identical (Pvalue > 0.05). These values fall within the range of 3.10 - 11.04% reported by [34] on *Basella alba*. While the Texaco market sample (2.54±0.43%) had the lowest lipid content and statistically different (P-value < 0.05) from the other three sites. The lipid contents of *Basella alba* from the present study are lower than the value of 11.04% found by [35] on *Basella alba* from Nigeria. The *Solanum macrocarpum* leaves from Texaco market, Nkombo garden and Thomas Sankara market sites, showed high lipid contents with values of 4.80±0.05%. 4.18±0.38% and 4.49±0.71% respectively. and no significant difference (P-value > 0.05). However, the sample from Talangai garden site (2.09±0.74%) displayed low lipid content, with significant difference (P-value < 0.05). These values are higher than those found by [36] on *Solanum macrocarpum* from Nigeria (0.96%).



**Figure 2: Lipid content of four leafy vegetables from four different sites (Talangai garden. Texaco market. Nkombo garden and Thomas Sankara market)**

For the same vegetable, when the letters above the bars are identical, the averages are identical (at the 5% threshold). Otherwise, the averages are significantly different (at the 5% threshold).

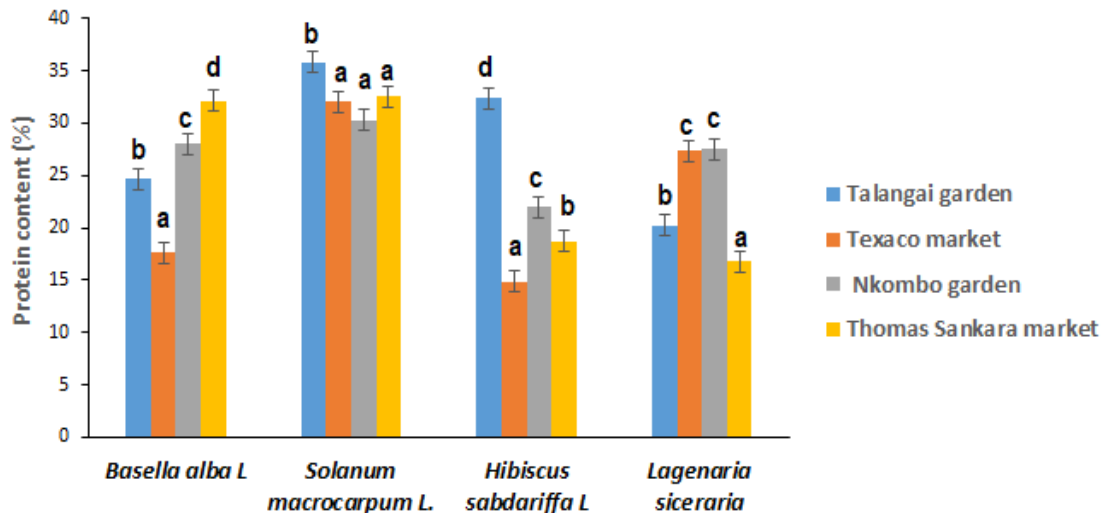
The *Hibiscus sabdariffa* leaf samples from Talangai (5.02 ± 0.42%) and Nkombo (4.053 ± 0.15%) gardens and Thomas Sankara market (4.14 ± 0.80%) showed high lipid contents, which were statistically identical (Pvalue > 0.05). The *Hibiscus sabdariffa* sample from the Texaco market (3.47± 0.46%) was low in lipid content, with no significant difference (Pvalue > 0.05) from those of the kombo garden and the Thomas Sankara market. These values are close to that found by [25] on *Hibiscus sabdariffa* leaves consumed in northern Côte d'Ivoire, which is 4.75 ± 0.15%. and are lower than the results on *Hibiscus sabdariffa* leaves

(6.30%) consumed in Nigeria [35]. Lipid contents of *Lagenaria siceraria* leaves varied between 3.08±0.28 and 4.36±1.32%. Samples from the Talangai garden (4.36 ± 1.32%), Texaco market (4.35 ± 0.21%) and Nkombo garden (4.29 ± 0.47%), showed high lipid contents, with no significant difference (Pvalue > 0.05). However, the sample from the Thomas Sankara market (3.08 ± 0.28%) had the lowest lipid content, significantly different (Pvalue < 0.05) from the three sites. These values are higher than that found by [32] on *Lagenaria siceraria* leaves (1.93± 0.15%). The lipid contents in the present study are close to the results of

the work by [37] on six green leafy vegetables consumed in Sri Lanka: *Premna latifolia* (3.38±0.23%), *Cardiospermum halicacabum* (4.75±0.22%), *Mollugo pentaphylla* (2.18±0.09%), *Delonix elata* (4.48±0.45%), *Argyrea pomacea* (3.88±0.26%) and *Pisonia grandis* (4.09±0.32%) and by [35] on *Gongronema latifolium* (3.51%), *Ocimum gratissimum* L (4.02%) and *Telfaria occidentalis* (5.50%), three leafy vegetables consumed in Nigeria.

**Protein content**

The mean protein contents of the studied leafy vegetables are presented in Figure 3. From the figure, the protein contents of the four studied vegetables at the four sites are high. ranged from 14.87±0.81% (*Hibiscus sabdariffa* L from Texaco market) to 35.81±1.85% (*Solanum macrocarpum* L from Nkomo garden). This high protein content could be related to the use of nitrogen fertilizer (NPK) during the production of leafy vegetables by vegetable farmers. This is confirmed by stating that the protein content would be influenced by the use of nitrogen fertilizer (NPK) during the production of leafy vegetables [38].



**Figure 3: Protein content of four leafy vegetables from four different sites (Talangai garden. Texaco market. Nkomo garden and Thomas Sankara market)**

For the same vegetable, when the letters above the bars are identical, the averages are identical (at the 5% threshold). Otherwise, the averages are significantly different (at the 5% threshold).

Protein contents of *Basella alba* leaves were significantly different (P-value < 0.05) from one site to another and varied between 17.58±0.11 and 32.13±2.23%. The highest protein contents were found in Talangai garden (24.62±1.47%), Nkomo garden (27.99±1%) and Thomas Sankara Market (32.13±3.16%). and they are higher than those of *Basella alba* leaves (22.1%) collected in the north-west of Burundi as found by [29] and are in the range of 27.70 - 58.80% reported by [34]. The lowest content was in the Texaco market sample (17.58 ± 0.15%). The leaves of *Solanum macrocarpum*, showed the protein contents ranging from 30.25±0.62 to 35.81±1.85%. The samples from Texaco market (32±1.18%). Nkomo garden (30.25±0.88%) and Thomas Sankara market (32.49±0.45%) sites showed protein contents statistically identical (Pvalue > 0.05). They are higher than the value found by [36] in Nigeria, which is 8.44%. The sample from Talangai garden (35.81 ± 2.61%) showed the highest protein content. with a significant difference (P-value < 0.05) from the other sites. The mean protein contents of *Hibiscus sabdariffa* leaves were significantly different (P-value > 0.05) between sites, ranging from 14.87±0.81 to 32.40±0.05%. The sample from Talangai garden

(32.34±0.07%) had the highest protein content and the samples from Texaco market, Nkomo garden and Thomas Sankara market had protein contents of 14.87±1.15%, 21.98±0.21 and 18.69±2.29%, respectively. These values are lower than the value found by [35], i.e. 46.56% and higher than that found on *Hibiscus sabdariffa* leaves (14.47 ± 0.10%) consumed in the north of Côte d'Ivoire [25]. Protein contents of *Lagenaria siceraria* leaves from the four sites ranged from 16.77±0.75 to 27.47±0.77%. The highest protein contents were observed in the samples from the Texaco market (27.35±1.79%) and the Nkomo garden (27.47±1.08%), which were statistically identical (Pvalue > 0.05). The samples from the Talangai garden and the Thomas Sankara market had protein contents of 20.25±0.7% and 16.78±1.05% respectively and statistically different at the 5% threshold. These results are almost identical to the result on *Lagenaria siceraria* leaves (24.5 ± 0.00%) [32].

The values in the present study indicate that these vegetables leaves may be considered as good sources of protein when compared to the values found on six green leafy vegetables consumed in Sri Lanka: *Premna latifolia* (1.35±0.07%), *Cardiospermum*

*halicacabum* (1.17±0.07%), *Mollugo pentaphylla* (0.53±0.04%), *Delonix elata* (3.52±0.06%) *Argyreia pomacea* (1.00±0.05%) and *Pisonia grandis* (1.67±0.04%) [37]. Thus, they can contribute to the food and nutritional security of the population.

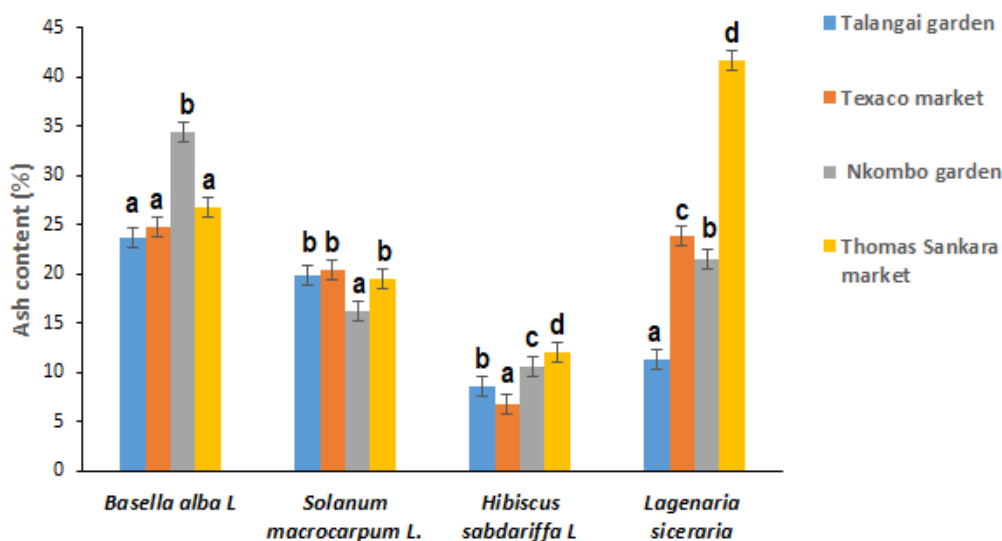
**Ash content**

Figure 4 shows the mean ash contents of the four studied leafy vegetables (*Basella alba*, *Solanum macrocarpum*, *Hibiscus sabdariffa* and *Lagenaria siceraria*). The average ash content values are high and they vary between 6.86±0.53 (*Hibiscus sabdariffa* L from Texaco market) and 41.59±1.62% DM (*Lagenaria siceraria* (Molina) standl from Thomas Sankara market).

The mean ash contents of *Basella alba* leaves are statistically identical (Pvalue > 0.05) in the Talangai garden. Texaco market and Thomas Sankara market at 23.65±2.24%, 24.84% and 26.83% respectively. The market gardener's sample from the Nkombo garden had the highest ash content of 34.3±0.17%. These values are higher than those reported by [34] which ranged from 5.02 -15.90% and on *Basella alba* leaves from Nigeria, 5.02% [35]. Regarding the ash contents of *Solanum macrocarpum* leaves, the mean values obtained at the market garden site of Talangai garden (19.86%),

Texaco (20.35%) and Thomas Sankara markets (19.50%) are higher and have no significant difference ((Pvalue > 0.05). While the content of the market garden site of Nkombo garden is the lowest at 16.31%. These values are higher than the one found on *Solanum macrocarpum* leaves (12.36%) from Nigeria [36].

Concerning *Hibiscus sabdariffa* L leaves, the ash contents showed a significant difference (P-value < 0.05) between sites. These contents are 8.58% in the Talangai garden, 6.86% in the Texaco market, 10.66% in the Nkombo garden and 12.12% in the Thomas Sankara market. These values are close to the results of on *Hibiscus sabdariffa* leaves (10.30 ± 0.10%) consumed in the North of Côte d'Ivoire [25] and on *Hibiscus sabdariffa* leaves (7.50%) in Nigeria [35]. As for *Lagenaria siceraria* leaves, the ash contents of the four sites are significantly different (P-value< 0.05). The highest content was obtained at the Thomas Sankara market, with a value of 41.59%. The ash contents in the Talangai garden, Texaco market and Nkombo garden were 11.31; 23.92 and 21.51% respectively. These values are far higher than the result on *Lagenaria siceraria* (Molina) standl (10.35± 0.55%), except for the one from the market garden site of Talangai garden that is slightly higher [39].



**Figure 4: Ash content of four leafy vegetables from four different sites (Talangai garden. Texaco market. Nkombo garden and Thomas Sankara market)**

For the same vegetable. When the letters above the bars are identical, the averages are identical (at the 5% threshold). Otherwise, the averages are significantly different (at the 5% threshold).

Overall, the ash contents of the four studied vegetables are higher than those of six green leafy vegetables consumed in Sri Lanka: *Premna latifolia* (9.12±0.14%), *Cardiospermum halicacabum* (7.33±0.38%), *Mollugo pentaphylla* (12.49±0.22%), *Delonix elata* (10.30±0.13%), *Argyreia pomacea* (11.86±0.26%) and *Pisonia grandis* (9.37±0.28%). according to work by [37]. Compared to other leafy

vegetables, these results show that, the studied leafy vegetables are rich and cheaper sources of ash. Indeed, ash is a good index of the mineral concentration in a sample. Since the ash content is high, these leafy vegetables are therefore rich in mineral elements and constitute a significant source of mineral elements in the human body.



**Mineral content**

The mineral content (iron, phosphorus, calcium and magnesium) of the leafy vegetables.

*Basella alba*, *Solanum macrocarpum*, *Hibiscus sabdariffa* and *Lagenaria siceraria*, collected in four sites are presented in Table 3.

**Table 3: Mineral contents (mg/100gDW) of four (4) leafy vegetables collected in four sites**

	Minerals	Talangai garden	Texaco market	Nkombo garden	Thomas Sankara market
<i>Basella alba</i>	Iron	28.10±1.71 <sup>b</sup>	18.80±0.5 <sup>a</sup>	58.70±3.66 <sup>c</sup>	72.90±2.88 <sup>d</sup>
	Phosphorus	239.17±1.22 <sup>b</sup>	299.18±1.72 <sup>c</sup>	229.18±1.82 <sup>a</sup>	372.80±1.56 <sup>d</sup>
	Calcium	4677.99±2.24 <sup>d</sup>	2922.59±1.79 <sup>c</sup>	2441.83±1.83 <sup>b</sup>	1118.05±2.51 <sup>a</sup>
	Magnesium	554.49±3.29 <sup>b</sup>	482.95±2.81 <sup>a</sup>	1452.57±4.71 <sup>d</sup>	1265.82±4.11 <sup>c</sup>
<i>Solanum macrocarpum L</i>	Iron	29.51±1.12 <sup>b</sup>	29.34±1.38 <sup>b</sup>	22.13±3.83 <sup>a</sup>	48.95±3.83 <sup>c</sup>
	Phosphorus	250.83±1.57 <sup>d</sup>	181.64±1.19 <sup>a</sup>	193.39±2.71 <sup>b</sup>	219.37±1.39 <sup>c</sup>
	Calcium	2438.98±3.46 <sup>a</sup>	3994±4 <sup>d</sup>	3724.03±4.03 <sup>c</sup>	3122.64±2.64 <sup>b</sup>
	Magnesium	460.02±1.22 <sup>b</sup>	704.5±1.55 <sup>c</sup>	192.75±1.27 <sup>a</sup>	192.28±1.73 <sup>a</sup>
<i>Hibiscus sabdariffa</i>	Iron	37.46±3.83 <sup>a</sup>	28.84±4.79 <sup>a</sup>	29.55±0.83 <sup>a</sup>	39.38±5.74 <sup>a</sup>
	Phosphorus	226.22±1.15 <sup>c</sup>	147.84±1.33 <sup>a</sup>	169.51±1.57 <sup>b</sup>	170.69±1.38 <sup>b</sup>
	Calcium	920.01±0.45 <sup>a</sup>	960.49±0.95 <sup>b</sup>	1876.92±3.08 <sup>d</sup>	1680.93±1.59 <sup>c</sup>
	Magnesium	899.43±1.68 <sup>c</sup>	480.61±1.14 <sup>b</sup>	996.21±1.90 <sup>d</sup>	338.62±1.41 <sup>a</sup>
<i>Lagenaria siceraria</i>	Iron	3.94±0.95 <sup>a</sup>	83.57±3.71 <sup>d</sup>	77.69±1.92 <sup>c</sup>	62.36±3.83 <sup>b</sup>
	Phosphorus	101.25±1.02 <sup>a</sup>	162.74±2.08 <sup>b</sup>	183.14±2.69 <sup>c</sup>	217.73±2.64 <sup>d</sup>
	Calcium	1961.32±1.32 <sup>a</sup>	2988.51±1.49 <sup>d</sup>	2602.7±2.70 <sup>c</sup>	2443.20±0.76 <sup>b</sup>
	Magnesium	531.81±2.53 <sup>b</sup>	215.92±2.24 <sup>a</sup>	627.86±2.69 <sup>c</sup>	1284.07±2.54 <sup>d</sup>

When, for the same lines, the superscript letters are identical, the averages are identical (at the 5% threshold). Otherwise, the averages are significantly different (at the 5% threshold).

**Iron**

Iron content ranged from 3.94±0.95 mg/100 g DM (*Lagenaria siceraria* Talangai garden) to 83.57±3.71 mg/100 g DM (*Lagenaria siceraria* Texaco market) (Table 3). The results indicate that the mean iron content of *Basella alba*, *Solanum macrocarpum* and *Lagenaria siceraria* shows a significant difference at the  $\alpha = 0.05$  threshold between sites. There is no significant difference in the iron content of *Solanum macrocarpum* from the Talangai garden and the Texaco market. Regarding *Hibiscus sabdariffa*, there was no significant difference in the iron content between the four sites (Pvalue > 0.05). The iron contents of *Basella alba* L (18.80±0.5-72.90±2.88mg/100 g DM) from the four collection sites were higher than those obtained with the same vegetable species collected in Burundi, i.e., a value of 16.10 mg/100 g DM [29]. The iron content of the Texaco market (18.80±0.5mg/100gDM) was similar to the value reported by [23] in Côte d'Ivoire (19.37 ± 0.65mg/100g). However, these values were lower than that of *Spinacia oleracea* (83.29 ± 8.18 mg/100g DM) [39]. The iron value of *Solanum macrocarpum* (22.13±3.83-48.95±3.8388mg/100g DM) from all sites were higher than the value found by [28], which is 15.98 mg/100g DM. The iron contents of *Hibiscus sabdariffa* (28.84±4.79-39.38±5.74 mg/100 g DM) were higher than those reported by [35, 36], of values 21.84 and 11.285 mg/100 g DM respectively; and similar to that obtained by [25], with a value of 30.87 ± 0.16 mg/100 g DM, for the same vegetable species. These results (Table 3) show that the studied leafy vegetables are a good source of iron that is an essential mineral for human health, playing a role in

immune function, cardiovascular health and cognitive development [40]. Thus, these vegetables could be recommended for iron-deficient individuals, such as pregnant and lactating women [31]. In addition, their consumption could reduce the consequences associated with iron deficiency, such as anemia [7], impaired cognitive development, increased susceptibility to infection, increased risk of morbidity in children, unfavorable pregnancy outcomes and decreased work productivity in adults [41].

**Phosphorus**

The results in Table 3 indicate that the leafy vegetables collected at the four sites have phosphorus contents that vary from 101.25±1.02 mg/100 g DM (*Lagenaria siceraria* Talangai garden) to 372.80±1.56 mg/100 g DM (*Basella alba* Thomas Sankara market). These values show significant differences between the collection sites at the  $\alpha = 0.05$  threshold, except for the values of *Hibiscus sabdariffa* obtained at the Nkombo garden and Thomas Sankara market which are statistically identical (P>0.05).

The phosphorus content of *Basella alba* and *Lagenaria siceraria* is high at the Thomas Sankara market with values of 372.80±1.56 mg/100g DW and 217.73±2.64 mg/100g DW, respectively. Concerning *Solanum macrocarpum* and *Hibiscus sabdariffa*, the phosphorus content is high at the Talangai garden, with values of 250.83±1.57 mg/100g DW and 226.22±1.15 mg/100g DW, respectively. However, the values in the present study are lower than those found by [25] with the traditional leafy vegetables, namely *Amaranthus*

*hybridus* ( $353.23 \pm 9.70$  mg/100g), *Andansonia digitata* ( $462.80 \pm 10.00$  mg/100g), *Ceiba patendra* ( $1290 \pm 30.00$  mg/100g), *Hibiscus sabdariffa* ( $472.5 \pm 12.00$  mg/100g) and *Vigna unguiculata* ( $682.86 \pm 50.00$  mg/100g), as well as those by [30] with spinach ( $513$  mg /100g) and kale ( $519$  mg/100g). These results show that the studied vegetables are a significant source of phosphorus which is an essential element for the body, as it is involved in many cellular reactions, particularly in glycolysis and oxidative phosphorylation, which are the main sources of ATP. Phosphorus is also important for metabolism as well as the synthesis of DNA and RNA [42].

### Calcium

The average calcium content of the analyzed leafy vegetables (Table 3) is very high, with values ranged from  $920.01$  mg/100 g DM (*Hibiscus Sabdariffa* Talangai garden) to  $4677.99$  mg/100 g DM (*Baselle alba* Talangai garden). They are significantly different ( $P < 0.05$ ) between the collection sites for the same studied vegetable.

The calcium contents of *Solanum macrocarpon* L from the four sites, ranging from  $3122.64 \pm 2.64$  to  $3994 \pm 4$  mg/100 g DM, are higher than the results obtained by [28] with the same vegetable species from Porto Novo in Benin, i.e., values ranging from  $351.52$  to  $539.96$  mg/100 g DM. The calcium contents of *Baselle alba* from Talangai garden ( $4677.99 \pm 2.24$  mg/100 g DM). Texaco market ( $2922.59 \pm 1.79$  mg/100 g DM) and Nkombo garden ( $2441.83 \pm 1.83$  mg/100 g DM) are higher than that obtained by [29] with *Baselle* from Ivory Coast. i.e.,  $1329.51$  mg/100g DM that reported by [30] with spinach ( $1036$  mg /100g) and that obtained by [23] with spinach consumed in Ivory Coast. i.e.,  $111.02 \pm 0.29$  mg/100g. In addition, the values of the present study are higher than the ones found by [39] with *Spinacia oleracea* ( $110 \pm 0.01$  mg/100g DM) and *Phytolacca dodecandra* ( $40 \pm 0.01$  mg/100g DM), two edible leafy vegetables in the Republic of Congo, and are close to the data of [25] with five leafy vegetables consumed in the north of Côte d'Ivoire whose values vary between  $1331.15$  mg/100g and  $4680$  mg/100g. These results show that the investigated vegetables are undeniable sources of calcium. which is involved as a constituent of bones and teeth, 9% of calcium in the human body is used for this function [43, 28]. The calcium is also involved in the regulation of nerve and muscle functions [28].

### Magnesium

The mean magnesium content of the analyzed leafy vegetables was non-negligible for the four study sites and ranged from  $192.28 \pm 1.73$  mg/100 g DM (*Solanum macrocarpon* L from Thomas Sankara market) to  $1452.57 \pm 4.71$  mg/100 g DM (*Baselle alba* L from Talangai garden). The results obtained showed a significant difference ( $P$ -value  $< 0.05$ ) between sites for

the four studied vegetables. Nevertheless, there was no significant difference ( $P$ -value  $> 0.05$ ) between the values of Nkombo garden ( $192.75 \pm 1.27$  mg/100 g DM) and Thomas Sankara market ( $192.28 \pm 1.73$  mg/100 g DM) for *Solanum macrocarpon* L. The magnesium values of *Solanum macrocarpon* L from the four sites ( $192.28 \pm 1.73$ - $704.5 \pm 1.55$  mg/100 g) are higher than those obtained by [28] on the same vegetable species consumed in Porto Novo, Republic of Benin, i.e.,  $112.97$ - $142.12$  mg/100 g DM. and lower than the value of  $1390$  mg/100 g, obtained by [36] on the same vegetable species used in Nigeria. The magnesium contents of the four sites are significantly higher than the value obtained by [29] which is  $390.4$  mg/100 g DM. These results show that the studied vegetables are an appreciable source of magnesium, a mineral known to prevent cardiomyopathy, muscle degeneration, growth retardation, alopecia, dermatitis, immunologic dysfunction, gonadal atrophy, impaired spermatogenesis, congenital malformations and bleeding disorders [37]. It is also involved in energy metabolism, in protein synthesis and as a cofactor for more than 300 enzymatic reactions [44].

## CONCLUSION

The present study focused on the biochemical characterization of the leaves of *Basella alba* L., *Hibiscus sabdariffa* L., *Solanum macrocarpon* L. and *Lagenaria siceraria* (Molina) Standl). four leafy vegetables. selected and collected in two sales sites (Texaco and Thomas Sankara markets) and market gardening sites (Talangai and Nkombo gardens) in the northern part of Brazzaville city.

The results obtained show that all the studied leafy vegetables are rich in water. which indicates that they cannot be stored for a long period at room temperature. The leaves of *Basella alba* L., *Solanum macrocarpon* and *Lagenaria siceraria* have a pH close to neutrality; while those of *Hibiscus sabdariffa* have an acid pH. Furthermore, the investigated vegetables are good sources of protein, ash and minerals (Ca, P, Fe and Mg). however, they are poor in lipids. These results suggest that the consumption of sufficient quantities of these locally available and accessible vegetables could contribute to the improvement of nutritional status and adequate protection against malnutrition-related diseases.

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