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Refrigeration Storage Effect on Nutritive Value and Antioxidant Properties of Five Leafy Vegetables Consumed in Northern Côte d'Ivoire (Ivory Coast)

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Abstract: Storage and processing technologies have been utilized for centuries to prevent post harvest losses of vegetables and to transform them into safe, delicious and stable products. The aim of this study was to evaluate the effect of refrigeration storage (4°C) on the nutritional value of five leafy vegetables (*Hibiscus sabdariffa, Amaranthus hybridus, Andasonia digitata, Vigna unguiculata* and *Ceiba patendra*) that are consumed in Northern Côte d'Ivoire. The selected samples were subjected to storage at 4 °C for 5, 10 and 15 days and the physicochemical properties were determined using standard methods. Fresh selected samples were used as the control. The result of the study revealed that increased storage time affected the nutritional value of the selected leafy vegetables. The registered losses at 15 days of refrigeration storage were as follow: ash (2.41- 9.70%), proteins (5.66- 18.97%), vitamin C (11.43- 41.29%), carotenoids (25.44- 41.29%), oxalates (12.43- 14.55%), phytates (35.03- 44.10%). It was observed an increase (0.17- 2.41%) in total phenolic content during storage. In addition, a small increase (0.88- 2.66%) of fibres content was also observed in the studied leafy vegetables stored under refrigeration at 4°C. Furthermore, after 15 days of storage at 4°C the residual contents of minerals were: calcium (308.30- 858.75mg/100g). Magnesium (213.64- 610.14 mg/100g), potassium (565.32-1844.08 mg/100g), iron (49.29- 160.53 mg/100g) and zinc (17.95- 34.06 mg/100g). The result of the present study suggests that vegetables should be stored in cold storage for a short time (less to five days) in order to avoid loss of essential nutrients.

Keywords: Leafy Vegetables, Nutritive Value, Refrigeration Storage, Antioxidant Properties

1. Introduction

Green leafy vegetables hold an important place in well-balanced diets. Vegetables include leaves, stems, roots, flowers, seed, fruits, bulbs, tubers and fungi [1,2] than can be eaten. These plants have been reported to be particularly rich in precursor of vitamin A and iron, two nutrients that are currently believed to be deficient in the diet of people in many countries [3]. The vegetables are also rich sources of vitamin C, proteins, fibres and minerals (potassium, phosphorous, calcium and zinc) [4]. In addition to antioxidant and vitamins, vegetables also contain high contents of phytochemical such as phenolic compounds including flavonoids, which have been implicated in the prevention of aging related diseases such as cancer, arteriosclerosis and diabete. African leafy vegetables play a significant role in food security of the underprivileged in both urban and rural setting and are also vital for the income generation [5]. Among the twenty hundred and seven (207) leafy vegetables widely consumed in tropical Africa, about twenty (20) species of leafy vegetables belonging to 6 botanical families, are widely consumed and cultivated by Ivorian population [6]. Furthermore, the consumption of these leafy vegetables is linked to the region and ethno-botanical studies have stated that most people in Northern Côte d'Ivoire consume indigenous green leafy vegetables such as *Amaranthus hybridus* "boronbrou", *Andasonia digitata* "baobab", *Ceiba patendra* "fromager", *Hibiscus sabdariffa* "dah" and *Vigna unguiculata* "haricot" [7, 8]. Many fresh leafy vegetables have a shelf life of only few days before they are unsafe or undesirable for consumption. In fact, Vegetables are highly perishable foods subject to rapid deterioration by microorganisms, enzymes, or oxidation reactions. Storage and processing technologies have been utilized for centuries to transform these perishable vegetables into safe, delicious and stable products [9]. Refrigeration slows down the chemical and biological processes in food and the accompanying deterioration and the loss of quality. Also, refrigeration down the respiration of fruits and vegetables and allows for longer shelf lives [10]. Owing to substantial loss of unused fresh produce, use of postharvest preservation methods such as refrigeration and freezing might extend their shelf life but may also affect the nutritional content of these crops [11]. A literature review [10] also concluded that very few studies have monitored changes in nutritional parameters in leafy vegetables commodity from harvest through storage and domestic cooking. The aim of this study was to evaluate the impact of refrigeration storage at 4 °C on the nutritive value of selected fresh green leafy vegetables.

2. Material and Methods

2.1. Samples Collection

Leafy vegetables (*Amaranthus hybridus, Andasonia digitata, Ceiba patendra, Hibiscus sabdariffa* and *Vigna unguiculata*) were collected fresh and at maturity from cultivated farmlands located at Dabou (latitude: 5° 19′ 14″ North; longitude: 4° 22′ 59″ West) (Abidjan District). The samples were harvested at the early stage (between one and two weeks of the appearance of the leaves). These plants were previously authenticated by the National Floristic Center (University Felix Houphouët-Boigny, Abidjan-Côte d'Ivoire). After harvesting, the fresh leafy vegetables were immediately transported in icebox (4°C) to the laboratory for further treatment.

2.2. Samples Processing

The fresh leafy vegetables were destalked, washed with deionized water and edible portions were separated from the inedible portion. The edible portions were chopped into small pieces (500g) and allowed to drain at ambient temperature. Each sample was divided into two lots. The first lot (raw, 250g) used as control was dried in an oven (Memmert, Germany) at 60°C for 72 h [12]. The dried leaves were ground with a laboratory crusher (Culatti, France) and were immediately analyzed. The second lot (250g) was stored in polyethylene bags and then placed at refrigerator (4°C) for 5, 10 and 15 days. After refrigeration storage times, the samples were dried and treated as the control.

2.3. Chemicals

All solvents (n-hexane, petroleum ether, acetone, ethanol and methanol) were purchased from Merck. Standards used (gallic acid, β-carotene) and reagents (metaphosphoric acid, Folin-Ciocalteu, DPPH) were purchased from Sigma-Aldrich. All chemicals used in the study were of analytical grade.

2.4. Nutritive Properties

2.4.1. Proximate Analysis

Ash, proteins and lipids were determined using official methods [12]. For crude fibres, 2 g of dried powdered sample were digested with 0.25 M sulphuric acid and 0.3 M sodium hydroxide solution. The insoluble residue obtained was washed with hot water and dried in an oven (Memmert, Germany) at 100 °C until constant weight. The dried residue was then incinerated, and weighed for the determination of crude fibres content. Carbohydrates and calorific value were calculated using the following formulas [13]:

Carbohydrates (dry matter basis):

$$100 - (\%\ \text{proteins} + \%\ \text{lipids} + \%\ \text{ash} + \%\ \text{fibres}) \quad (1)$$

Calorific value (dry matter basis):

$$(\%\ \text{proteins} \times 2.44) + (\%\ \text{carbohydrates} \times 3.57) + (\%\ \text{lipids} \times 8.37) \quad (2)$$

The results of ash, fibres, proteins, lipids and carbohydrates contents were expressed on dry matter basis.

2.4.2. Anti-Nutritional Factors Determination

Oxalates content was performed using by titration method [14]. One (1) g of dried powdered sample was weighed into 100 mL conical flask. A quantity of 75 mL of sulphuric acid (3 M) was added and stirred for 1 h with a magnetic stirrer. The mixture was filtered and 25 mL of the filtrate was titrated while hot against KMnO4 solution (0.05 M) to the end point.

Phytates contents were determined using the Wade's reagent colorimetric method [15]. A quantity (1 g) of dried powdered sample was mixed with 20 mL of hydrochloric acid (0.65 N) and stirred for 12 h with a magnetic. The mixture was centrifuged at 12000 rpm for 40 min. An aliquot (0.5 mL) of supernatant was added with 3 mL of Wade's reagent. The reaction mixture was incubated for 15 min and absorbance was measured at 490 nm by using a spectrophotometer (PG Instruments, England). Phytates content was estimated using a calibration curve of sodium phytate (10 mg/mL) as standard.

2.4.3. Mineral Analysis

Minerals contents were determined by the ICP-MS (inductively coupled argon plasma mass spectrometer) method [16].The dried powdered samples (5 g) were burned to ashes in a muffle furnace (Pyrolabo, France). The ashes obtained were dissolved in 10 mL of HCl/HNO_3 and transferred into 100 mL flasks and the volume was made up using deionized water. The mineral composition of each sample was determined using an Agilent 7500c argon plasma mass spectrometer. Calibrations were performed using external standards prepared from a 1000 ppm single stock solution made up with 2% nitric acid.

2.4.4. Antioxidant Properties

(i). Vitamin C and Carotenoids Determination

Vitamin C contained in analyzed samples was determined by titration [17]. About 10 g of ground fresh leaves were soaked for 10 min in 40 mL metaphosphoric acid-acetic acid (2%, w/v). The mixture was centrifuged at 3000 rpm for 20 min and the supernatant obtained was diluted and adjusted with 50 mL of bi-distilled water. Ten (10) mL of this mixture was titrated to the end point with dichlorophenol-indophenol (DCPIP) 0.5 g/L.

Carotenoids were extracted and quantified by using a spectrophotometric method [18]. Two (2) g of ground fresh leaves were mixed three times with 50 mL of acetone until loss of pigmentation. The mixture obtained was filtered and total carotenoids were extracted with 100 mL of petroleum ether. Absorbance of extracted fraction was then read at 450 nm by using a spectrophotometer (PG Instruments, England). Total carotenoids content was subsequently estimated using a calibration curve of β-carotene (1 mg/mL) as standard.

(ii). Polyphenols Determination

Polyphenols were extracted and determined using Folin–Ciocalteu's reagent [19]. A quantity (1 g) of dried powdered sample was soaked in 10 mL of methanol 70% (v/v) and centrifuged at 1000 rpm for 10 min. An aliquot (1 mL) of supernatant was oxidized with 1 mL of Folin–Ciocalteu's reagent and neutralized by 1 mL of 20% (w/v) sodium carbonate. The reaction mixture was incubated for 30 min at ambient temperature and absorbance was measured at 745 nm by using a spectrophotometer (PG Instruments, England). The polyphenols content was obtained using a calibration curve of gallic acid (1 mg/mL) as standard.

(iii). Antioxidant Activity

Antioxidant activity assay was carried out using the 2,2-diphenyl-1-pycrilhydrazyl (DPPH) spectrophotometric method [20]. About 1 mL of 0.3 mM DPPH solution in ethanol was added to 2.5 mL of sample solution (1 g of dried powdered sample mixed in 10 mL of methanol and filtered through Whatman No. 4 filter paper) and was allowed to react for 30 min at room temperature. Absorbance values were measured with a spectrophotometer (PG Instruments, England) set at 415 nm. The average absorbance values were converted to percentage antioxidant activity using the following formula:

$$\text{Antioxidant activity (\%)} = 100 - [(\text{Abs of sample} - \text{Abs of blank}) \times 100/\text{Abs positive control}] \quad (3)$$

2.4.5. Statistical Analysis

In the experiment, each test for the samples was analyzed in triplicate. The data were expressed as means ± standard deviation (SD). Differences between means were analyzed by analysis of variance (one way ANOVA) coupled with Duncan's test using STATISTICA 7.1 (StatSoft) software. Statistical significance was measured at $p < 0.05$.

3. Results and Discussion

3.1. Proximate composition

The proximate composition of leafy vegetables stored at 4°C is presented in Table 1. During refrigeration storage at 4 °C, small fluctuation ($p > 0.05$) in moisture, ash, fibres, proteins, lipids, carbohydrates contents were observed compared to freshly samples. The slight decrease in moisture contents (70.45 – 86.05% to 66.62 – 84.18%) during refrigeration storage was due to the respiration and others senescence-related metabolic processes [10]. The stable values obtained may be due to refrigeration temperature (4°C) which slows down the respiration and transpiration rates. The ash content after 5 days of refrigeration ranged from 8.51 ± 0.33% (*A. hybridus*) to 24.81 ± 0.42% (*C. patendra*). These values were closed to 7.89 ± 0.47% and 23.18 ± 0.04% after 15 days of refrigeration. The reduced ash contents could be attributed to the loss water carrying off the minerals during transpiration stage [10]. Besides the decrease rate from (0.26-3.35%) to (2.41-10.75%) at 5 and 15 days of storage respectively, the studied leafy vegetables may be considered as a good source of minerals when compared to values (2 – 10 %) obtained for cereals and tubers [21]. Storage in refrigerator at 4 °C of the selected leafy vegetables resulted in slight increase (0.09-0.61%) to (0.88-2.66%) in their crude fibres contents ($p > 0.05$) after 5 and 15 days of storage, respectively. The increase in total fibres may be due to textural changes such as wilting and shriveling observed during refrigeration of vegetables [22]. Indeed, the high level of crude fibres in these leafy vegetables would be advantageous for their active role in the regulation of intestinal transit, increasing dietary bulk due to their ability to absorb water [23]. As concern protein contents, storage at 4°C caused 0.13 to 5.98% reduction after 5 days. It's worth noting that plant foods which provide more than 12 % of their calorific value from proteins have been shown to be good source of proteins [24]. This suggests that all the leafy vegetables investigated may be good sources of proteins and could play a significant role in providing cheap and available proteins for rural communities. The relatively low values of lipids contents after 5 days of refrigeration, corroborates the findings of many authors which showed that leafy vegetables are poor sources of fat [25]. However, it's important to note that diet providing 1 – 2 % of its caloric energy as fat is said to be sufficient to human beings, as excess fat consumption yields to cardiovascular disorders such as atherosclerosis, cancer and aging [26]. Therefore, the consumption of these leafy vegetables in large amount may be recommended to individuals suffering from obesity. The estimated caloric values agree with general observation that vegetables have low energy values due to their low crude fat and relatively high level of moisture [27].

3.2. Mineral Composition

Mineral content is an essential component of the nutritive value of fresh green leafy vegetable. Table 2 showed the mineral composition of leafy vegetables stored in refrigerator at 4 °C. The residual contents of minerals showed statistical difference ($p < 0.05$) after 5 days of storage at 4 °C. These contents were: calcium (396.70-920.90 mg/100g), magnesium (228.19-661.01 mg/100g), potassium (702.32-1967.63 mg/100g), iron (57.66-168.09 mg/100g) and zinc (21.81-39.74 mg/100g). These observed reductions may be due to the small losses of ashes by transpiration phenomena during refrigeration. There was a significant difference ($p < 0.05$) in mineral contents between the refrigerated leafy vegetables and this variability was expected since the green leafy vegetables belonged to different botanic families. Considering the recommended dietary allowance (RDA) for minerals [28], consumption of 5-days refrigerated leafy vegetables could cover at least 50% RDA. Therefore, they could contribute substantially for improving human diet. Calcium and phosphorus are associated for growth and maintenance of bones, teeth and muscles [29]. As concern magnesium, this mineral is known to prevent cardiomyopathy, muscle degeneration, growth retardation, congenital malformations and bleeding disorders [30]. Iron is important in the diet of pregnant, infants and the elderly to reduce cases of deficiency associated with disease such as anemia [31]. Zinc is important for vitamin A and vitamin E metabolism [28, 32]. To predict the bioavailability of calcium and iron, anti-nutrients to nutrients ratios of were calculated (Table 3). It was observed a small decrease of anti-nutritional factors (oxalates and phytates) in the refrigerated samples compared to the fresh green leafy vegetables (Figure 1). The calculated [oxalates]/[Ca] and [phytates]/[Ca] ratios in all the species, excepted *H. sabdariffa*, were below the critical level of 2.5 known to impair calcium bioavailability [33]. It was also observed that the calculated [phytates]/[Fe] ratios of *H. sabdariffa* were above the critical level of 0.4. This implies that the phytates of these leafy vegetables may hinder iron bioavailability [34]. However, the [phytates]/[Fe] ratios could be considerably reduced after processing such as soaking, boiling or frying [35].

Table 1. *Proximate composition of refrigerated (4°C) leafy vegetables consumed in Northern Côte d'Ivoire*

	Moisture (%)	Ash* (%)	Fibres* (%)	Proteins* (%)	Lipids* (%)	Carbohydrates* (%)	Energy* (kcal /100g)
H. sabdariffa							
0 days	86.05 ± 1.35a	10.30 ±0.10a	14.27 ±1.70a	14.47 ±0.10a	4.75± 0.15a	56.21 ±1.78a	275.71± 0.55a
5 days	85.73 ±0.53a	10.13±0.02a	14.30±0.35a	14.01±0.03b	4.61±0.01a	56.95±0.41a	276.09±1.28a
10 days	84.43± 0.46a	9.97± 0.38a	14.50±1.41a	13.78±0.06b	4.44±0.03b	57.31±0.95a	275.39±3.77a
15 days	84.18±0.22a	9.31±0.27b	14.65±1.96a	13.15±0.08b	3.75±0.05c	60.13±3.83a	275.75±3.03a
A. hybridus							
0 days	72.98 ±0.10a	8.59 ±1.34a	17.80±0.30a	13.25 ±0.13a	2.15 ±0.01a	58.21± 1.78a	305.19 ±7.73a
5 days	72.28± 0.05a	8.51± 0.33a	17.05±2.47a	13.12±0.10a	1.93±0.03a	59.39±2.10a	260.20±7.77b
10 days	71.81±0.58a	8.15±1.43a	17.20±0.35a	12.91±0.04a	1.53±0.04b	60.21±1.70a	259.27±6.54 b
15 days	70.57±0.10a	7.89±0.47a	17.34±1.41a	12.50±0.01b	1.00±0.06c	61.11±1.84a	257.04±7.00b
A. digitata							
0 days	77.63± 0.15a	10.97± 0.4a	12.56± 0.45a	18.08± 0.10a	2.18± 0.03a	56.23± 1.25c	267.03± 4.00b
5 days	77.28±0.05a	10.87±0.05a	12.60±1.36a	17.65±0.14b	1.71±1.01b	57.75±1.57c	263.56±3.00b
10 days	76.78±0.40a	10.23±0.10b	12.78±0.35a	16.91±0.01c	1.22±1.03c	60.84±0.21b	289.59±1.03a
15 days	76.18±0.04a	9.79±0.20b	12.80±1.83a	14.65±0.04d	0.58±1.04d	64.23±2.55a	269.89±9.55b
V. unguiculata							
0 days	80.04± 0.56a	11.17± 0.25a	18.00 ±0.92a	21.96 ±0.30a	4.23± 0.25a	44.64 ±1.72b	248.35± 1.33b
5 days	79.80±0.14a	11.14±0.16a	18.11±1.95a	21.93 ±0.04a	3.92±0.03b	44.90±4.71b	246.62±1.17b
10 days	78.92±0.39a	11.07±0.93a	18.25±2.47a	20.88±0.07b	3.20±0.04c	46.60±3.52 b	244.08±2.03b
15 days	77.85±0.39a	10.90±0.89a	18.37±3.54a	19.65±0.06c	2.34±0.06d	52.24±4.54a	254.04±1.58a
C. patendra							
0 days	70.45 ±0.52a	25.67 ±1.12a	31.50± 1.50a	15.20 ±0.05a	1.39± 0.22a	26.30 ±0.11b	142.61 ±7.74a
5 days	70.22±1.10 a	24.81±0.42 a	31.53±1.06a	14.29±0.01b	1.15±1.03b	28.22±1.56b	145.24±4.96a
10 days	69.97±0.17a	23.67±0.05a	31.66±1.06a	13.46±0.08c	0.97±1.03c	30.24±1.14a	148.90±3.51a
15 days	66.62±3.01a	23.18±0.04a	31.78±1.62a	12.48± 0.11d	0.32±1.05d	33.14±1.52a	151.44±5.48a

Data are represented as Means ± SD (n = 3). Means in the column with no common letter differ significantly ($p<0.05$) for each leafy vegetable.
*: values given on dry matter basis.

Table 2. *Mineral composition (mg/100g dry matter) of refrigerated (4°C) leafy vegetables consumed in Northern Côte d'Ivoire.*

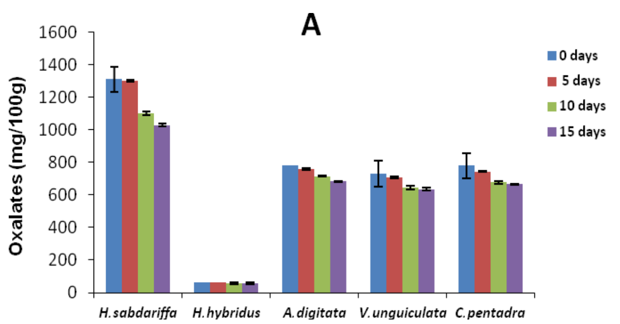
	Ca	Mg	P	K	Fe	Na	Zn
H.sabdariffa							
0 days	402.21±0.55a	295.93±0.41a	407.59±0.00a	816.19±1.12a	102.08±0.14a	23.46 ±0.03a	26.06 ±0.04a
5 days	396.70±0.54b	252.80±0.32b	393.39±1.93b	726.15 ±4.45b	57.66±0.12b	23.13 ±0.16a	24.96±0.74b
10 days	388.30±4.33c	233.06±5.10c	386.30±5.05c	647.52 ±0.95c	53.04 ±2.03c	22.93 ±2.88a	23.27± 0.03b
15 days	308.30±1.32d	229.52±6.02c	341.82±4.81d	625.97±8.58d	49.88 ±2.32d	22.44± 1.70a	22.42± 0.17b
A.hybridus							
0 days	932.6 ±0.55a	497.75±0.49a	368.69±0.00a	1989.32±2.12a	77.88± 0.05a	94.39 ±0.04a	31.73 ±0.04a
5 days	920.90±2.77b	486.69±2.77b	367.33±3.99a	1967.63±4.28b	73.58 ±2.88b	94.35±1.72a	31.27± 0.31a
10 days	911.49± 4.9c	480.40±5.54c	362.53±5.36a	1954.53 ±7.75c	69.33±3.47b	94.21±2.71a	30.69 ±1.53a
15 days	858.75±1.94d	451.19±7.57d	323.14±3.74b	1844.08±3.13d	49.29±8.51c	93.66±1.28a	27.07±0.7b

	Ca	Mg	P	K	Fe	Na	Zn
A.digitata							
0 days	496.26±2.20a	264.36±1.17a	761.63±0.00a	1856.90±8.23a	106.27±0.47a	37.13± 0.12a	22.61± 0.10a
5 days	481.33 ±1.91b	228.19±1.49b	573.72±6.37b	1783.33±6.43b	73.35 ±1.77b	37.10±0.78a	21.81 ±0.08b
10 days	474.82 ±3.83c	219.89±5.32c	502.48±4.11c	1777.52 ±1.12c	72.34± 0.47b	36.42 ±0.96a	18.48 ±0.07c
15 days	415.05±7.62d	213.64±3.34d	448.25±2.51d	1672.90±3.77d	71.50 ±1.12b	34.53 ±1.90a	17.95± 0.12c
V.unguiculata							
0 days	439.54±0.56a	341.34±0.18a	309.04±0.00a	718.11± 0.91a	91.45± 0.12a	33.32 ±0.02a	40.83 ±0.04a
5 days	430.81± 6.26b	323.33±2.93b	306.30±1.94a	702.32±6.88b	88.03±0.88b	32.35±1.09a	39.74 ±0.40a
10 days	428.05±2.09c	318.26±9.31b	301.58±8.09a	653.67±3.69c	85.09±3.10b	31.49±2.99a	37.94 ±1.30b
15 days	423.54±4.01c	310.35±3.19c	298.94±1.75a	565.32±3.71d	82.70±4.51b	31.26±1.37a	34.06±0.84c
C.patendra							
0 days	997.02±0.55a	773.55±0.43a	570.85±2.11a	1585.58±0.87a	219.84±0.12a	42.69± 0.02a	35.68 ±0.02a
5 days	823.39±6.31b	661.01±1.76b	532.46±6.18b	1278.19±9.52b	168.09± 0.59b	37.94±1.76b	34.67±0.78a
10 days	819.21±1.91b	637.24±2.07c	523.41±1.17c	1211.88 ±2.11c	160.53± 0.41c	36.78 ±0.70b	33.17± 0.08b
15 days	744.50±1.65c	610.14±5.34d	502.17±4.53d	1084.69±2.48d	152.36± 3.82d	33.14 ±0.39c	27.10± 0.02c

Data are represented as Means ± SD (n = 3). Means in the column with no common letter differ significantly ($p<0.05$) for each leafy vegetable.

Table 3. *Anti-nutritional factors/mineral ratios of refrigerated (4°C) leafy vegetables consumed in Northern Côte d' Ivoire.*

	Phytates/Ca	Phytates/Fe	Oxalates/Ca
H.sabdariffa			
0 days	0.21	0.85	3.26
5 days	0.19	1.34	3.28
10 days	0.16	1.20	2.83
15 days	0.16	1.00	3.33
A.hybridus			
0 days	0.03	0.41	0.07
5 days	0.03	0.33	0.07
10 days	0.02	0.30	0.06
15 days	0.02	0.36	0.06
A.digitata			
0 days	0.04	0.19	1.57
5 days	0.04	0.26	1.57
10 days	0.03	0.23	1.51
15 days	0.03	0.18	1.64
V.unguiculata			
0 days	0.04	0.19	1.66
5 days	0.04	0.19	1.64
10 days	0.03	0.16	1.51
15 days	0.02	0.13	1.50
C.patendra			
0 days	0.04	0.17	0.78
5 days	0.04	0.19	0.90
10 days	0.03	0.17	0.83
15 days	0.03	0.15	0.89



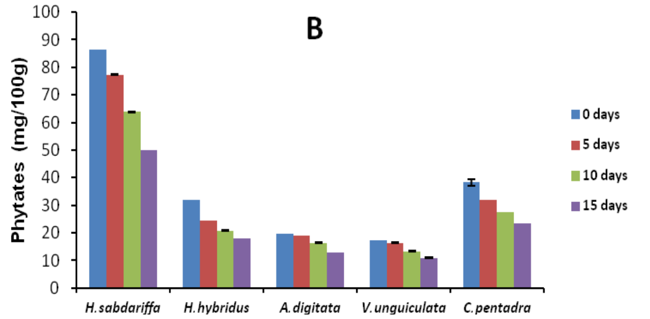


Figure 1. *Oxalates (A) and phytates (B) contents of refrigerated (4°C) leafy vegetables consumed in Northern Côte d'Ivoire.*

3.3. Antioxidant Properties

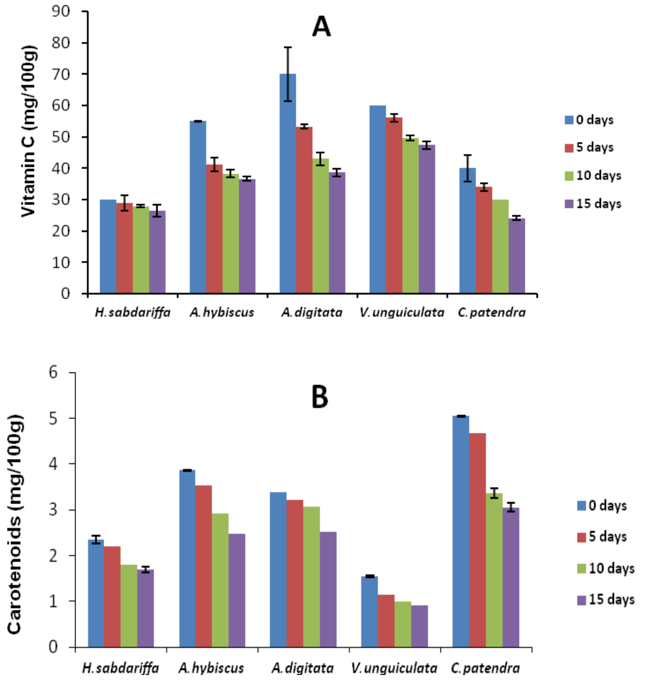


Figure 2. *Vitamin C (A) and carotenoids (B) contents of refrigerated (4°C) leafy vegetables consumed in Northern Côte d'Ivoire*

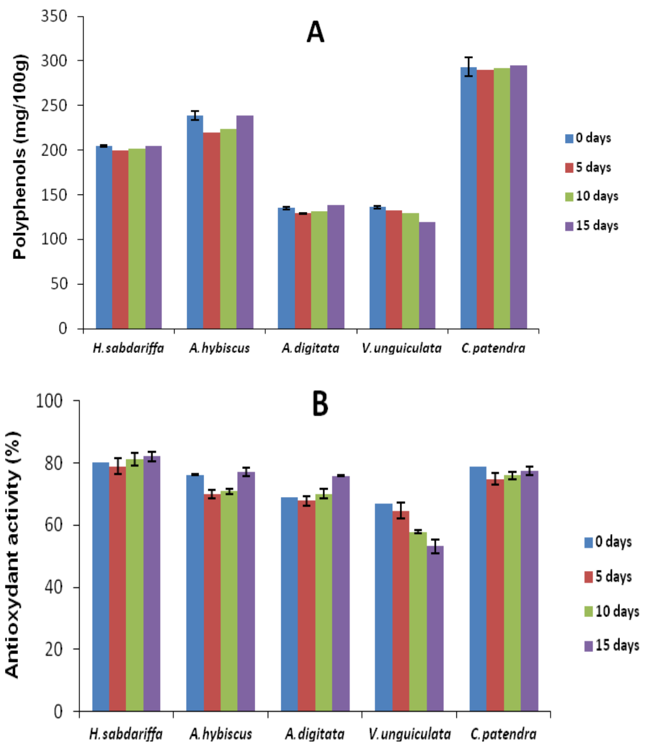


Figure 3. Polyphenols content (A) and antioxidant activity (B) of refrigerated (4°C) leafy vegetables consumed in Northern Côte d'Ivoire

Refrigeration at 4°C resulted in a small decrease of carotenoids and vitamin C contents in the studied leafy vegetables (Figure 2). Carotenoids play an important potential role in human health by acting as biological antioxidants [36]. Carotenoids, losses at 5 days were estimated from 5.02 to 25.80%. Loss of carotenoid contents during storage has been reported in numerous studies [37, 38]. The decrease in the concentration of total carotenoids could be attributed to the degradation of pigments during refrigeration [39]. Vitamin C is vulnerable to enzymatic oxidation; therefore, it is sensitive and appropriate marker for monitoring quality change during, transportation, processing and storage [40, 41]. Losses in vitamin C ranged from 3.56 to 25% and 11.43 to 44.68% at 5 and 15 days, respectively of storage at 4°C (Figure 2).This result corroborates previous work [42] that reported only 20% retention of ascorbic acid in spinach stored under refrigerated temperature (4°C) for 7 days. Also, the decrease of vitamin C content is in agreement with other studies [43, 44]. Vitamin C degradation is due to auto-oxidation and also enzymatic degradation. Indeed, vitamin C losses continue through post harvest handling, processing and storage of fruit and vegetables [45]. The effect of refrigeration on polyphenols content and antioxidant activity of the selected leafy vegetables is shown in figure 3. Phenolic compounds also seem to be more affected by storage factors such as temperature, atmosphere and light, than either vitamin C or carotenoids [46]. At the beginning (0-day), the total phenolic content of the five fresh leafy vegetables ranged from 135.21 to 293.08 mg/100g. At the end of the storage (15 days) total phenolic content increased except for *Vigna unguiculata*. This increase in total phenols agrees with some reports [47, 44]. During storage at 4°C, the species showed variation of total phenolic; this trend was portrayed as an initial decrease followed by an increase in phenolic content. The decrease and increase in phenolic content during refrigeration may be due to degradation in phenolic compounds by enzyme action.

4. Conclusion

From the above investigation, it can be concluded that leafy vegetables consumed in Northern Côte d'Ivoire contain significant levels of nutrients that are essential for human health. However, refrigeration storage minimizes nutritive losses in the studied leafy vegetables. During refrigeration at 4°C, small fluctuation in ash, fibres, proteins and fat were observed after 5, 10 and 15 days of storage. It was also observed a decrease of vitamin C and carotenoids and a cyclic variation in phenolic compounds. For best storage, the result of the present study recommends that leafy vegetables should be harvested fresh, washed free of soil and kept in perforated plastic bags within a few days (less than 5 days) at cleaned refrigerator since longer storage results in loss of freshness and flavor. In addition, each group of leafy vegetables must be separated (use one drawer for each group) in order to minimize the detrimental effects of ethylene produced during respiration phenomenon.

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