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PROXIMATE, MINERAL, AND BIOACTIVE COMPOSITION OF EDIBLE *PIPER CUBEBA* LIANAS (PACOBIER) IN SOUTHERN CÔTE D'IVOIRE

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ABSTRACT

Piper cubeba (Piperaceae) is a plant with nutritional and therapeutic properties, often used in food and in alternative medicine for preventive purposes. Although the *Piper cubeba* lianas are widely consumed as a spice and condiment by populations of the 'La Mé' region in southern Côte d'Ivoire, there is a lack of studies on their nutritional potential. The Objective of This study was assessed the nutritional values of fresh and traditionally dried *Piper cubeba* lianas harvested. Fresh lianas and those dried for 10 days were analysed using standard biochemical methods. Nutritive parameters were determined according to standard methods and the energy content was calculated. Food plants mineral analysis was performed by atomic absorption spectrophotometric methods. The data were statistically analysed. The results revealed a significant effect ($P < 0.05$) of traditional sun-drying of *Piper cubeba* lianas on biochemical parameters. In the sample of dried lianas, moisture content decreased (86.76 - 7.53%), the pH increased (5.68 - 6.07), carbohydrates (45.87 to 49.67%) and total sugars increased (72.94 - 75.09%), protein decreased (6.33 to 5.13%) and fibre increased (5.80 - 10.10%). The dried lianas extract of *Piper cubeba* had a significantly ($P < 0.05$), with an increase in total polyphenols (540.25 - 2615.28 mg GAE/100 g). Potassium was the main element (2.3 – 2.7 mg/100 g), followed by calcium (268.9–289.3), the levels of which were higher in the dried lianas. As conclusion These results showed that consuming dried *Piper cubeba* lianas as a spice and condiment provides more nutrients and bioactive compounds, which could be beneficial to consumers' health.

Keywords: *Piper cubeba* lianas, Nutritive; Consumer, Health, Benefit

1 INTRODUCTION

In the context of global diets, edible vegetables are recognised for their culinary flexibility and nutritional value, which can be consumed either raw or cooked/processed. In the contemporary era, the significance of edible vegetables in addressing pressing issues, including but not limited to health, the environment, and food security, is paramount. In accordance with the recommendations set forth by the Food and Agriculture Organization (FAO), green leafy vegetables have been identified as significant sources of vitamins, minerals, fibre, and phytochemicals. The aforementioned nutrients have been demonstrated to function as antioxidants, thereby contributing to the retardation of the ageing process and the prevention of the development of certain diseases [1]. The indigenous flora of the geographical region of Sub-Saharan Africa is characterised by a high level of biodiversity, encompassing a substantial number of edible plants. It is important to note that a considerable proportion of these plants remain unidentified or underutilised, despite their recognised nutritional and therapeutic value. According to Ehilé *et al.*, 207 leafy vegetables are widely consumed in tropical Africa, with approximately 20 leafy vegetable species belonging to six botanical families being consumed and cultivated by people [1]. It is evident that the *Piper cubeba* lianas, which are members of the Piperaceae family, have a long history of utilisation as a spice or condiment by Ivorian populations [1]. The preceding ethnonutritional study of *Piper cubeba* lianas also referred to by consumers as 'pacobier', a natural seasoning which constitute one of the spices and condiments, was conducted among the population of four localities in the department of Alépé, in the La Mé region of southern Côte d'Ivoire. However, there is a paucity of information with regard to their nutritional composition. In consideration of the culinary significance of *Piper cubeba* lianas within the population's habitual dietary intake, it is imperative to undertake an assessment of its nutritional composition. The present study therefore sought to assess the phytochemical, proximate, and mineral composition of fresh and traditionally prepared foodstuffs. The aim of this study is therefore to assess the phytochemical composition, proximate and mineral content of fresh and traditionally dried *Piper cubeba* lianas consumed by indigenous populations, with a view to making the best possible use of them.

2 MATERIAL AND METHODS

2.1 Sampling

The survey was conducted in the southern region of Côte d'Ivoire, specifically in the La Mé area, which corresponds to the distribution area of the pepper *Piper cubeba*. The investigation was conducted in the localities of Dzeudji, Memni, Grand Alépé, and Montézo, which are located in the sub-prefecture of Alépé. The selection of these four localities was informed by studies conducted by the Fonds Interprofessionnel pour la Recherche et le Conseil Agricoles (FIRCA) [2]. *Piper cubeba*, lianas were harvested from the forests of the investigated localities.

2.2 Sample preparation and drying

The samples were collected and stored in clean, dry containers before being promptly transported to the laboratory for powder preparation. The *Piper cubeba* lianas (Figure 1) were washed under running cold water to remove any dust, dirt or parasites. They were then washed again with deionised water. These lianas were divided into two batches; one was oven-dried while still fresh and the other was left to 10 days air-dry in the laboratory for ten days before being oven-dried. The dehydrated samples were ground with an electric grinder (IKA M20). The powder was sieved to 100 µm for homogeneity and then stored in sterile, airtight glass containers [3].

Figure 1: Fresh Piper cubeba liana samples

2.3 Determination of pH

The hydrogen ion concentration (pH) of *Piper cubeba* samples was measured according to the AOAC method using a digital pH meter (PHS-3D). The pH meter was standardized with standard buffers of pH 4 and pH 7. The pH was determined by making 10% w/v suspension of sample in distilled water. The suspension was mixed thoroughly and a pH meter probe calibrated with pH buffer seven was introduced into each sample, and read when values were stable [4].

2.4 Macronutrient analysis

The proximate analysis of *Piper cubeba* liana samples was determined using standard methods. Moisture content was determined by gravimetry, heating in an oven at 105 ± 1 °C until a constant mass was achieved [5]. Protein content was determined using the Kjeldahl method. The sample was mineralised at 400°C with a catalyst and sulphuric acid, and then the nitrogen converted into ammonia was released by alkaline distillation (40% sodium hydroxide) and trapped in an acid-base solution with a colour indicator. The trapped ammonia is then titrated with a 0.1 N sulphuric acid solution, with a blank test carried out in parallel for correction. Total nitrogen was converted into protein content using a factor of 6.25 [6]. Lipid content was determined using n-hexane in a Soxhlet extraction apparatus, while crude fibre content was determined by dilute acid and alkali hydrolysis [6]. Total ash content was quantified by incinerating 1 g of the samples at 550 ± 1°C for 23 hours and weighing the residue in the crucible [7]. The total carbohydrate content was calculated using the following equation: 100 - (% moisture + % protein + % fat + % ash + % fibre) [8]. The energy value was calculated using the Atwater–Benedict conversion factors: 9 kcal/g of lipid, 4 kcal/g of carbohydrate, and 4 kcal/g of protein [9].

2.5 Determination of phytochemical compounds

2.5.1 Extraction of Phenolic Compounds

The extraction of phenolic compounds was performed according to the following protocol: 1 g of lianas powder was homogenized in 10 mL of 70% ethanol (v/v), then centrifuged at 1000 rpm for 10 min. The pellet was re-extracted with 10 mL of 70% ethanol and centrifuged again. The supernatants were combined in a 50 mL volumetric flask and brought to volume with distilled water [10].

2.5.2 Total Phenol Assay

Total phenol content was determined according to the method of Singleton *et al.* [10]: 0.5 mL of ethanolic extract was mixed with 0.5 mL of Folin-Ciocalteu reagent. After 3 min, 0.5 mL of Na_2CO_3 (20%) was added, and the volume adjusted to 3.5 mL with distilled water. The tubes were incubated for 30 min in the dark, then absorbance was read at 725 nm. A calibration curve with gallic acid (0–1 mg/mL) allowed results to be expressed in mg gallic acid equivalent (GAE)/100 g dry matter (DM).

2.5.3 Flavonoid Assay

To 0.5 mL of ethanolic extract were successively added 0.5 mL of distilled water, 0.5 mL of $AlCl_3$ (10% w/v), 0.5 mL of sodium acetate (1 M), and 2 mL of distilled water. After 20 min in the dark, the absorbance was read at 415 nm [11]. A quercetin calibration curve (0–0.1 mg/mL) allowed the results to be expressed in mg quercetin equivalents (QE)/100 g DM.

2.5.4 Tannin Assay

The tannin content was determined using the method described by Bainbridge *et al.* [12]. A volume of 1 mL of each methanolic extract was collected and mixed with 5 mL of reaction solution (vanillin 0.1 mg/mL in 70% sulphuric acid (V/V)). The mixture was allowed to stand at room temperature in the dark for 20 minutes. Absorbance was measured at 500 nm against a blank (without extract). Tannic acid was used for the calibration curve with a concentration range of 0–100 µg/mL. The results were expressed as mg of tannic acid equivalents (TAE)/100 g DW.

2.6 Determination of anti-nutritional compounds

2.6.1 Phytate Assay

The phytate content was determined using the Wade reagent. Two gram (2 g) of sample was homogenized in 20 mL of 0.65 N HCl, agitated for 12 hours at room temperature, and then centrifuged. After adding 0.5 mL of the supernatant to 3 mL of Wade reagent, the absorbance was measured at 490 nm following 20 minutes of incubation in the dark [13]. The results were expressed as mg of phytic acid equivalents (PAE)/100 g DW.

2.6.2 Oxalate Assay

Oxalates in samples content was determined by using the method described by Day and Underwood [14]. Two gram (2 g) of lianas powder was homogenised in 75 mL of H_2SO_4 (3M) under magnetic stirring for one hour. The resulting mixture was filtered through Whatman No. 1 filter paper. The filtrate was titrated under heat with a solution of KMnO4 (0.05 M) until the colour changed to a persistent pink. The oxalate content of the sample is given in mg/100 g by the following expression. The results were expressed as mg of oxalic acid equivalents (OAE)/100 g DW.

2.7 Determination of Ethanol-soluble Sugars

2.7.1 Total Sugar Content

The determination of total soluble sugars was carried out according to the method of Dubois *et al.* [15]. After extraction with 80° G.L ethanol (Gay Lussac) of the free sugars contained in a quantity of 5 g from each sample of the vegetable powder, purification by solutions of lead acetate (10%) and oxalic acid (10%) was carried out. The resulting solution was evaporated in a sand bath and supplemented with 25 mL with distilled water. A quantity of one millilitre (1 mL) of the previous sugar solution was taken, followed by 1 mL of phenol (5%) and 5 mL of concentrated sulphuric acid (94-97%). After cooling to room temperature, the reading was made at the wavelength of 490 nm on the spectrophotometer, against a standard range of 0 to 80 ụg glucose. The tests were done in triplicate.

2.7.2 Reducing Sugar Content

The quantification of reducing sugars was carried out according to the method of Bernfeld [16]. A volume of one millilitre (1 mL) of an ethanolic extract from Alchornea cordifolia powder was introduced into a test tube and successively a volume of 0.5 mL of distilled water and a volume of 0.5 mL of dinitrosalycilic acid (DNS) were added. The mixture was heated in a boiling water bath for 5 minutes and after cooling, a volume of 5 mL of distilled water was added. The optical density was thus read at 540 nm against a blank test. A calibration range using a 0.1 mg/mL glucose solution allowed the quantification of reducing sugars and the tests were carried out in triplicate.

2.8 Statistical Analysis

All analyses were performed in triplicate. Means and standard deviations were calculated with Excel 2021. Statistical analyses were performed with Statistica 7.1. An ANOVA was conducted to compare variables. In case of significant difference, the Duncan test was used to classify means into homogeneous groups. The significance threshold was set at $p < 0.05$.

3 RESULTS AND DISCUSSION

3.1 Macronutrient Composition of the *Piper cubeba* lianas

Proximate analysis (Table 1) shows a significant variation ($p < 0.05$) in the physicochemical parameters of fresh *Piper cubeba* lianas and those dried at room temperature for ten days. The fresh lianas exhibit high moisture content characteristic of hydrated plant tissues, whereas the dried samples reach 7.53%, reflecting the removal of approximately 90% of the water. This dehydration concentrates the solids, limits oxidative enzymatic activity, and enhances chemical stability without notable alteration of bioactive compounds [1]. These results are consistent with those 65–69.55% from different regions as reported by Coulibaly *et al.* [1] for *Piper nigrum* fresh seed and 11.92% after drying. Moisture level below 10–14% is optimal to ensure the microbiological and biochemical stability of plant-based matrices intended for storage or processing into powder. The protein content in lianas decreases (6.33 - 5.13 %) significantly after drying. This decrease could be due to denaturation and partial degradation of proteins due to heat and oxidation during drying in dried tropical plants. Protein content is lower than those reported for *Piper nigrum* seeds (12.99- 14.56 %) founded by imo *et al* [17] in Nigeria and Coulibaly *et al.* [1] for different *Piper nigrum* grown regions in Côte d'Ivoire. This difference could be due to fact liana consists mainly of conductive and supportive tissue (rich in parietal compounds) [18], is physiologically less rich in protein nitrogen than leaves or seeds, which are organs with high metabolic or storage activity. The lipid content was low in both lianas' samples (2.77 - 2.87%). Lipids in lianas were inferior compared to *Piper guineense* seed (4.06 %) reported by Imo *et al* [17], probably because seed accumulate reserve lipids typical of reproductive organs and oilseeds. This difference supports the idea that the lianas are non-lipidogenic vegetative organ which could constitute a low-fat matrix advantageous in terms of its use as a dietary supplement with a low lipid-calorie density. Fibre content in lianas increased (5.8 - 10.1 %) significantly after drying. This increase could be mainly due to concentration effect after water removal, as the parietal fibres such as cellulose, hemicellulose, lignin, pectin minimally degraded by drying. Values are inferiors for dried leaves of *Piper guineense* (20.99 %) but significantly higher than those of its seeds (6.95 %) as reported by Imo *et al* [17]. However, Ndefo *et al.* [19] found low fibre content (2.90 %) in *Piper guineense* seeds. High fibre content is a valuable source of dietary fibre, the beneficial effects of which on bowel function and blood sugar regulation are well established in the literature on wild lianas traditionally consumed in West Africa [20]. The total carbohydrate content increases (45.87 - 49.67 %)

significantly after drying, as does the total sugar content (72.94 - 75.09 %). These carbohydrates constitute the major fraction of the lianas dry matters, which is consistent with the proportions reported for other organs of the genus Piper, where carbohydrates also represent the predominant fraction: 43.86–46.17% in the leaves of *Piper nigrum*, 51.37–53.74% in its fruits, and up to 64.54% in the seeds of *Piper guineense* [19].

Table 1 : Macronutrient composition of the *Piper cubeba* lianas

Parameters	Fresh lianas	Dried lianas
Moisture (%)	86.76 ±5.40*	7.53 ± 0.26
pH	5.68 ± 0.51	6.07 ± 0.70*
proteins (%)	6.33 ± 0.45*	5.13 ± 0.25
lipids (%)	2.77 ± 0.06	2.87 ± 0.40*
Fibres (%)	5.8 ± 1.10	10.1 ± 1.00*
Carbohydrates (%)	45.87 ± 1.03	49.67 ± 8.47*
Total sugars (%)	72.94 ± 1.99	75.09 ± 1.29*
Reducing sugars (%)	11.21 ± 6.30*	9.43 ± 1.26
Ash (%)	7.44 ± 0.60	7.43 ± 0.58
Energy (Kcal/100g MS)	202.39 ± 2.45	213.86 ± 1.30*

In the same line, means marked with an asterisk (*) are significantly different (p > 0.05) according to Duncan's test

The total sugars (72.94 – 75.09 %) were found high in dried lianas. Conversely, reducing sugars (11.21 –9.43 %) decreased and was low in dried lianas. This suggests that some of these simple sugars are consumed through non-enzymatic browning reactions by the Maillard reaction or through hydrolysis/interconversion to non-reducing forms during heat treatment, a phenomenon commonly reported during the drying of sugar-rich plant materials.

The ash content remains statistically stable between fresh and dried lianas (7.44 – 7.43%), the absence of a significant difference reflecting the refractory nature of the minerals with regard to thermal degradation and water loss: drying results in neither an apparent gain nor loss of mineral matter. This content is relatively high to that reported for the leaves of *Piper nigrum* (3.54 – 5.33 %) founded by Coulibaly *et al.* [1] for various *Piper nigrum* seeds, whilst remaining lower than that of *Piper guineense* leaves (11.98 %) found by Imo *et al* [17], which highlights the mineral richness of the *Piper cubeba* lianas, contributing to the supply minerals for electrolyte balance and bone health. This liana high ash consistent with its role as sap-conducting tissue and the primary conduit for mineral elements absorbed by the roots. The energy value increases significantly after drying, rising (202.39 – 213.86 kcal/100), consistent with the parallel increase in carbohydrates and, to a lesser extent, lipids, these two fractions being the main contributors to the calorie content of the matrix. These values remain moderate compared with those reported for the leaves (273.68–295.67 kcal/100 g) and seeds (361.65–384.18 kcal/100 g) of *Piper nigrum* previously found by Nwofia *et al.* [21]. This difference is directly linked to the lianas lower lipid and protein content compared with these organs. This moderate energy value, combined with a high fibre content, is typical of wild edible lianas from West Africa, which are generally described as food sources providing a moderate calorie intake, derived mainly from carbohydrates, with a low contribution from protein and fat.

3.2 Mineral Composition of the *Piper cubeba* lianas

Table 2 presents the mineral composition of fresh and dried extracts of *Piper cubeba*. Potassium is the most abundant macroelement, with contents of 23.01 g/100 g DM (fresh) and 27.07 g/100 g DM (dried), followed by calcium (2.69 and 2.89 g/100 g DM respectively). Potassium increased after drying, reflecting a concentration effect. The high K/Na ratios (79.87 in fresh; 95.65 in dried) suggest an antihypertensive effect, while the Ca/P ratios (1.57 and 1.37) and Mg/Ca ratios (0.26 and 0.25) are favourable for bone absorption [22]. Our potassium contents exceed those reported by Borquaye *et al.* [23] for Nigella sativa (~1.2–1.5 g/100 g DM). Similarly, the calcium and phosphorus values surpass those observed by Okiki *et al.* [24] for nutmeg (*Myristica fragrans*). The Ca/P ratios fall within the range described by Janet *et al.* [25] for wild Nigerian vegetables (1.89–2.75). Regarding trace elements, iron dominated with 0.18 mg/100 g DM in both extract types, followed by copper and zinc (0.04 mg/100 g DM each). Iron content remained stable in both fresh and dried extracts, indicating good resistance to the drying process. This stability is explained by the inorganic nature of iron, which is relatively insensitive to thermal treatments [26]. Zinc contents (0.04 mg/100 g DM) were also stable between fresh and dried matrices, suggesting the absence of losses associated with the drying process [27].

Similarly, copper contents remained stable (0.04 mg/100 g DM) in both forms, a characteristic attributable to copper existing predominantly in bound forms integrated into organic complexes such as metalloproteins [28].

Table 2: Mineral composition of the *Piper cubeba* lianas

Parameters	Mineral	Fresh lianas	Dried lianas
Macroelements (g/100 g)	Sodium (Na)	0.29	0.28
	Phosphorus (P)	2.10	2.11
	Magnesium (Mg)	0.70	0.73
	Calcium (Ca)	2.69	2.89
	Potassium (K)	23.01	27.07
Trace elements (mg/100 g)	Iron (Fe)	0.18	0.18
	Iodine (I)	0.02	0.01
	Copper (Cu)	0.04	0.04
	Zinc (Zn)	0.04	0.04
Ratios	K/Na	79.87	95.65
	Ca/P	1.57	1.37
	Mg/Ca	0.26	0.25

3.3 Phytochemical compounds of the *Piper cubeba* lianas

The analysis of phytochemical and antinutritional properties of liana extracts of *Piper cubeba* (Table 3) reveals notable differences between fresh and dry samples. Phytates are absent from both types of extracts. Oxalates are detectable only in dry extracts (220 mg OAE/100 g DM), while absent in fresh extracts. This presence after drying can be explained by the concentration of organic constituents resulting from water removal, a classic phenomenon in plant matrices where dehydration increases the concentration of non-volatile metabolites [29]. These values remain substantially lower than those reported by Borquaye *et al.* [23] for five Ghanaian spices (1500–4270 mg/100 g for phytates and 80–760 mg/100 g for oxalates), suggesting a more favourable antinutritional profile for *Piper cubeba*.

Total polyphenols are significantly more concentrated in dry extracts (2615.28 mg GAE/100 g DM) than in fresh ones (540.25 mg GAE/100 g DM), revealing a strong concentration effect after drying. This increase is explained by the removal of water and improved release of compounds bound to the cellular matrix, despite possible minor oxidative losses [30]. These values far exceed those reported for caraway (3.99 mg GAE/100 g DM) and nutmeg (0.63 mg GAE/100 g DM) by Vallverdú-Queralt *et al.* [31], as well as those measured in certain Ghanaian spices (100–500 mg GAE/100 g) by Borquaye *et al.* [23]. Polyphenols, secondary metabolites widely distributed in the plant kingdom, exert a protective function against environmental stresses owing to their antioxidant properties [32].

Flavonoid contents measured in the fresh and dried extracts of *Piper cubeba* were 185.30 mg QE/100 g DM and 176.60 mg QE/100 g DM respectively, revealing a statistically significant decrease ($p < 0.05$) after drying. This reduction may result from thermal or oxidative degradation, as water-soluble flavonoids are sensitive to dehydration processes [30]. The observed levels confirm the richness in flavonols and flavones characteristic of the Piper (genus), and far exceed those reported for strawberries (20–50 mg QE/100 g DM) [33].

Tannin content decreased significantly after drying, from 57.53 mg TAE/100 g DM in the fresh lianas to 44.53 mg TAE/100 g DM in the dried lianas. This reduction may result from thermal degradation and/or oxidative polymerization of tannins during dehydration, converting them into higher-molecular-weight, poorly soluble complexes that are less detectable by the Folin-Ciocalteu-based colorimetric assay, and/or from residual polyphenol oxidase activity acting on these compounds before complete enzyme inactivation. Comparable reductions in tannin content after drying have been reported for pomegranate (*Punica granatum*) peel, where oven-drying produced significantly lower total tannin concentrations compared with fresh or freeze-dried material [34]. This pattern is consistent with the broader literature on the thermal instability of condensed and hydrolysable tannins under elevated processing temperatures.

Table 3: Phytochemical and antinutritional properties of the *Piper cubeba* lianas

Phytochemical compound (100 g DM)	Fresh lianas	Dried lianas
Oxalates (mg OAE/100 g)	ND	220 ± 00
Phytates (mg PAE/100 g)	ND	ND
Total polyphenols (mg GAE/100 g DM)	540.25 ± 18.84	2615.28 ± 139*
Flavonoids (mg QE/100 g DM)	185.30 ± 3.51*	176.60 ± 2.58
Tannins (mg TAE/100 g DM)	57.53 ± 2.60*	44.53 ± 1.86

In the same line, means marked with an asterisk (*) are significantly different ($p > 0.05$) according to Duncan's test

4 CONCLUSION

The results confirm that drying is a major transformative process that, beyond the simple reduction of moisture (from 86.76% to 7.53%), induces a significant concentration and qualitative modification of all biochemical constituents of this plant. Phytochemical compounds, particularly polyphenols and tannins, concentrate considerably in dry extracts, giving them significantly superior antioxidant and therapeutic potential. On the mineral level, the exceptional richness in potassium, combined with a very favourable K/Na ratio, positions *Piper cubeba* as a plant of great nutritional interest for the prevention of cardiovascular diseases. These results valorise the lianas of *Piper cubeba* not only as a simple traditional condiment, but as a plant resource with high functional, nutritional, and nutraceutical potential. Complementary studies on its in vivo biological activities and bioavailability are necessary to confirm and extend these promising perspectives.

STATEMENTS ON COMPLIANCE WITH ETHICAL STANDARDS

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Disclosure of conflict of interest

No conflict of interest to be disclosed.

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