

Biochemical Characteristic, Amino and Fatty Acids Contents of Pistachio *Citrullus mucospermus* Fursa Seeds from Abidjan (Côte d'Ivoire)

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Abstract The seeds of pistachio *Citrullus mucospermus* Fursa much used in the diet in Côte d'Ivoire, were studied to determine their chemical composition and their nutritional potential. The biochemical parameters of the seeds of *Citrullus mucospermus* Fursa characterized are dry matter, ash content, moisture content, carbohydrates, fiber, protein, amino and fatty acids. The proximate analysis of the seeds showed dry matter contents 93.6 ± 2.82 %, moisture 6.40 ± 0.14 %, ash 5.18 ± 0.06 %, carbohydrates 24.94 ± 0.18 %, fibre $9.93 \pm$ % and crude protein 28.46 ± 0.04 %. These proteins have two of the essential amino acids with a high level of lysine whereas serine was the limiting essential amino acid. The extraction yield of the lipids (crude fat) of the seeds was 31.49 ± 0.62 %. Unsaturated fatty acids: oleic acid (1.041 ± 0.002 %), linoleic acid (0.998 ± 0.001 %) and arachidonic acid (0.881 ± 0.001) were measured. Results revealed the seeds of pistachio *Citrullus mucospermus* to have great nutritional potentials with regard to their chemical composition therefore, their consumption could contribute to fulfill some food deficiencies in Côte d'Ivoire.

Keywords: nutritional value, amino acid, fatty acid, pistachio seeds, *Citrullus mucospermus*, Côte d'ivoire

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1. Introduction

Cucurbits are among the economically most important vegetable crops worldwide and are grown in both temperate and tropical regions [1,2]. Thus, cucurbits (pistachios) have a diverse range of species and are unconventional oil plants. The seeds are widely used around the world in combination with other foods to meet human protein and fat needs. *Citrullus mucospermus* Fursa is highly prized for its transformation into a peanut-like paste used to thicken sauces or extract oil [3]. The seeds of *Citrullus mucospermus* Fursa have an important social and cultural aspect for the populations which consume them [4]. These seeds are very important from a nutritional point of view given their richness in mineral salts (sulfur, calcium, potassium, phosphorus, iron, magnesium and zinc) [5] and in macronutrients. They can contain up to 36 % protein compared to peanuts which only contain 24 % [6]. They also have a high content of unsaturated and essential fatty acids, in particular linoleic acid [7,8].

But despite all of this dietary potential, pistachios are still considered marginal crops for nutritional interventions in Africa and are absent from major research and development programs [9]. These seeds of great potential in nutrients of important use by the ivorian populations deserve to be analyzed from a biochemical point of view to promote an efficient use in the approach to solving nutritional problems in the country. This justifies this study, the general objective of which is to evaluate the biochemical parameters of the seeds of *Citrullus mucospermus* Fursa with a view to their valorization in food.

2. Material and Methods

2.1. Biological Material

In this study, the biological (plant) material used was oilseeds of the species *Citrullus mucospermus* Fursa. These pistachio seeds were purchased in Abidjan (South, Côte d'Ivoire) and transported in plastic bags to Daloa (Central-West, Côte d'Ivoire) (Figure 1).



Figure 1. Seeds of *Citrullus mucospermus* Fursa

2.2. Preparation of *Citrullus mucospermus* Fursa Flour

For the production of the flour, a quantity of one hundred (100) g of *Citrullus mucospermus* Fursa seeds were dried in oven at 60°C for 72h and crushed using a blender type MOULINEX (Optibind 2000). The sieved grind obtained constituted the flour of *Citrullus mucospermus* Fursa. This flour was stored in pre-dried jars for further analysis according to the production diagram in Figure 2.

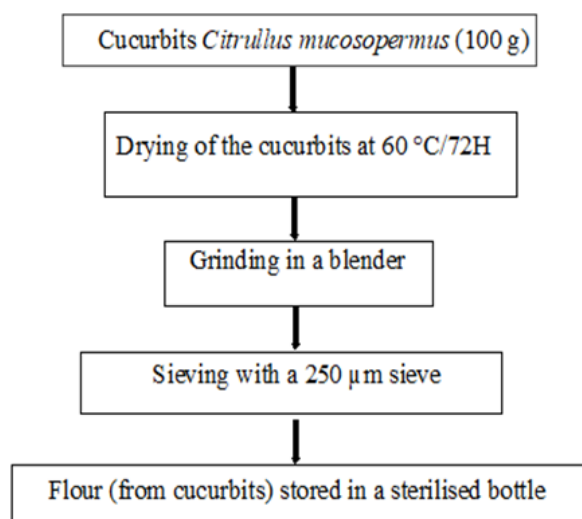


Figure 2. Flow diagram of the process for producing cucurbits *Citrullus mucospermus* flour

2.3. Proximate Analysis

The standard analytical procedures for food analysis were adopted for the determination of the moisture content, crude protein, crude fiber, percentage lipids, carbohydrate, ash and calorific value

2.3.1. Determination of Moisture Content

The method used is that of AOAC [10] whose principle is based on the dehydration by drying in the oven of the samples until obtaining a constant mass. A quantity of five (5) grams of the pistachio flour sample was weighed using a balance (Denver, model ABT 320 - 4M) in a crucible of known mass (M_0). The crucible + flour of mass M_1 was placed in an oven (MEMMERT 854 SCHWABACH, Germany) at 105°C ($\pm 2^\circ\text{C}$) for 24 hours. After cooling in a desiccator (GLASWERK WERTHEIM, 2 bars), the crucible was weighed again (M_2). Three tests were

performed and the dry matter content was determined from the following formula:

$$\text{Dry Matter Content (\%)} = \frac{M_1 - M_2}{ME_0} \times 100 \quad (1)$$

M_1 : mass of crucible + 5g of sample

M_2 : mass of crucible + 5g of sample placed in an oven (105 \pm 2°C) for 24h

ME_0 : mass of sample (5 g).

2.3.2. Determination of Ash Content

The ash content was determined according to the AOAC [10] method. A quantity of five (5) grams of the flour sample was weighed with a balance (Denver, model ABT 320 - 4M) into a porcelain crucible of known mass (M_0). The whole assembly (crucible + product) was heated in a furnace (VELP Scientifica, Spain) at 550 °C ($\pm 2^\circ\text{C}$) for 24 hours. After cooling in a desiccator (GLASWERK WERTHEIM, 2 bars), the crucible containing the ashes was weighed and noted (M_1). The tests were carried out in triplicate. The ash content was determined from the formula :

$$\text{Ash Content (\%)} = \frac{M_1 - M_0}{ME} \times 100 \quad (2)$$

M_1 : mass of crucible + ash (g) of sample placed in a furnace (550 \pm 2°C) for 24h

M_0 : mass of empty crucible

ME : mass of sample (5 g)

2.3.3. Crude Lipid and Fatty Acids Content Determination

From the pulverized sample, 3.00 g was used for determining the crude lipid by extracting the lipid from it for 5 h with (60 to 80°C) petroleum ether in a soxhlet extractor [11]. Triplicate samples were extracted to obtain triplicate values that were later averaged.

To analyze fatty acids of samples, the HPLC chromatography method was used. The HPLC WATERS ALLIANCE Chromatography equipped with column C18 or equivalent, wavelength of 262 nm, mobile phase (Methanol/Water (50:50)) and flow rate of 1.2 ml min⁻¹.

2.3.4. Protein and Amino Acids Content Determination

Total protein was determined by the Kjeldahl method. 0.5 g of the sample was weighed in triplicate into a filter paper and put into a Kjeldahl flask, 8 to 10 cm³ of concentrated H₂SO₄ were added and then digested in a fume cupboard until the solution became colourless. Distillation was carried out with about 10 cm³ of 40 % NaOH solution. The condenser tip was dipped into a conical flask containing 5 cm³ of 4 % boric acid in a mixed indicator till the boric acid solution turned green. Titration was done in the receiver flask with 0.01 M HCl until the solution turned red [11]. The crude protein was calculated by multiplying the estimated nitrogen by 6.25 and the amount of protein in each sample was obtained. To identify the amino acids after preparation of pistachio and standard solutions prepared, the samples were injected into HPLC WATERS ALLIANCE. The conditions of HPLC was determined as follows: column: C18 or equivalent, Wavelength of 254 nanometer, mobile phase (Methanol/Water (50:50)) and Flow Rate: 1.2 ml min⁻¹.

2.3.5. Determination of Crude Fiber

From the pounded sample, 3.00 g were used in triplicates for estimating the crude fiber by acid and alkaline digestion methods using 20 % H₂SO₄ and 20 % NaOH solutions [11].

2.3.6. Carbohydrate Determination

The carbohydrate content was calculated using the following formula: Available carbohydrate (%), = 100 – [protein (%) + Ash (%) + Fibre (%) + Crude Fat (%)] [12].

2.3.7. Caloric Value

The caloric value was calculated in kilocalories per 100 g (kcal/100 g) by multiplying the crude fat, protein and carbohydrate values by Atwater factors of 9, 4 and 4 respectively.

2.3.8. pH Measurement

The pH values of the samples were determined by suspending 10 % W/V of the sample in distilled water in each case. It was then thoroughly mixed in a 100 cm³ beaker, stirred and the pH was taken. This was repeated three times and the average calculated [13].

2.4. Statistical Analysis

All measurements were performed in triplicate. Statistical analyzes of the data were performed using STATISTICA 7 software (Statsoft Inc, Tulsa-USA Headquarters). This software was also used to calculate mean values and standard deviations of the trials.

3. Results and Discussion

3.1. Results

3.1.1. Biochemical Proximate Composition

Table 1 shows the proximate biochemical composition of pistachio *Citrullus mucospermus* seeds which showed that the seeds are good sources of crude fat (31.49 ± 0.62 % dw), protein (28.46 ± 0.04 % dw), fiber (9.93 ± 0.08 % dw) and carbohydrate (24.94 ± 0.18 % dw).

From the same Table 1, the moisture content of the seed was found to be 6.40 ± 0.14 % dw and the ash content was 5.18 ± 0.06 % dw. The pH of *Citrullus mucospermus* seed was 6.15 ± 0.07.

Table 1. Biochemical properties of pistachio *Citrullus mucospermus* Fursa seeds

Composition	Values (% DW)
Dry matter	93.60 ± 2.82
Moisture	6.40 ± 0.14
Ash	5.18 ± 0.06
Total sugar	6.50 ± 0.40
Carbohydrates	24.94 ± 0.18
Crude Fat	31.49 ± 0.62
Crude fiber	9.93 ± 0.08
Reducing sugar	1.95 ± 0.09
Crude Protein	28.46 ± 0.04
Energy Value (Kcal)	497.01 ± 2.75
pH	6.15 ± 0.07

Values are mean ± standard deviation (n=3).

3.1.2. Pistachio Amino Acids Content

Table 2 reveals the amino acid profile (%) of *C. mucospermus*. Of the total 4 types of amino acids estimated, the alanine was 0.506 ± 0.004 %, leucine was 0.686 ± 0.005 %, lysine was 9.607 ± 0.012 % and serine 0.189 ± 0.004 % are in the least quantity. Among the four amino acids determined, leucine and lysine are essential aminoacids compare to alanine and serine.

Table 2. Amino Acids of pistachio *Citrullus mucospermus* Fursa seeds

Parameters	(Values % DW)
Alanine	0.506 ± 0.004
Leucine*	0.686 ± 0.005
Lysine*	9.607 ± 0.012
Serine	0.189 ± 0.004

Values are mean ± standard deviation (n=3)

*Essential aminoacids.

3.1.3. Pistachio Fatty Acids Content

Table 3 highlights the fatty acid profile (%) of *C. mucospermus* seeds. Thus, of the total 5 types of fatty acids estimated, the oleic acid (1.041 ± 0.002 %), palmitic acid (1.167 ± 0.012) and stearic acid (1.369 ± 0.011) are the values that are greater than or equal to 1% whereas arachidonic acid (0.881 ± 0.001) and linoleic acid (0.998 ± 0.001) are less than 1 %. It is important to note that arachidonic, linoleic and oleic fatty acids are essential unsaturated fatty acids.

The result obtained from the biochemical analysis carried out on the pistachio *C. mucospermus* in this study confirmed that the biochemical, amino acid and fatty acid contents were found to be represented.

Table 3. Fatty Acids of pistachio *Citrullus mucospermus* Fursa seeds

Parameters	Values (% DW)
Arachidonic acid*	0.881 ± 0.001
Linoleic acid*	0.998 ± 0.001
Oleic acid*	1.041 ± 0.002
Palmitic acid	1.167 ± 0.012
Stearic acid	1.369 ± 0.011

Values are mean ± standard deviation (n=3)

*Unsaturated fatty acids.

3.2. Discussion

In the present study biochemical properties of seeds from fruits of pistachio were analyzed. The crude fat, protein, fiber, carbohydrates and ash contents of seeds were found to be 31.49 %, 28.46 %, 9.93 %, 24.94 %, 5.18 % respectively. The present work showed some what similar data for protein and fat content to those reported by Anwar et al. [14].

The crude fat was considerably high (31.49 %), a value higher than the 15.48 % reported for *Parkia biglobosa* seeds [15], than the 16.01 % reported for *Casiatora* seeds by Adamu et al. [16] and than the results reported in *Acacia nilotica* fruit as reported by Bwai et al. [17]. The high fat content suggests that *Citrullus mucospermus* seeds can be classified as an oil-rich seed. Thus, the main fatty acid component found in pistachio kernels was stearic acid, followed by palmitic acid, oleic acid, linoleic acid and arachidonic acid. In addition, the three:

arachidonic, oleic and linoleic are unsaturated fatty acids which are considered to have a good nutritional value because it is more reactive and naturally antioxidants in the body [18]. This result is agree with those reported by Atri et al. [19] and Maftoun et al. [20] in their studies. This gives a nutritional and medicinal importance for these plant seeds. Indeed, linoleic acid has a beneficial role for the human body [21]. The major saturated fatty acid is the stearic acid. Results show that *C. mucospermus* seeds are rich with unsaturated fatty acids. Moreover the monounsaturated fatty acid has an important physiological role [22]. These results indicate that *C. mucospermus* seed oils could be a good source of table oil. The content of linoleic acid is of particular interest especially in the fight against atherosclerosis [23]. Present results are also similar to that of previous study on *Citrullus* and *Cucurbita pepo* seed oil which was found to contain mostly palmitic, stearic, oleic and linoleic acids, with linoleic acid as the most abundant [23,24,25].

The protein content was 28.46 %, which was higher than 24.69 and 20 % obtained for unfermented groundnut and sesame seed by Ojokoh and Lawal [26] and Nzikou et al. [27], respectively. This value was also higher than 27.9 % crude protein reported for *Parkia biglobosa* seeds by Elemo et al. [15] and the 21.0 % obtained for *Pigeon pea* [28]. This protein value obtained from this study indicates that it can contribute to the daily human protein requirements based on 23 -56g as stipulated [29].

The amino acid profiles of *C. mucospermus seeds* flour are presented in Tables 2. The sample contains. The essential and non-essential amino acids for *C. mucospermus* determined in this study were 50 % and 50 %, respectively. The *C. mucospermus* contained two essential amino acids, with the highest value of lysine (9.607 %) whereas the leucine was in low quantity (0.686 %). The non-essential amino acids include : alanine and serine. The amino acid profile of the studied sample suggests that its protein is of high nutritive value which can be exploited by the feeds industries for feeds formulations.

The carbohydrate content obtained was 24.94 % which was lower compared to 29.72 % reported for *Acacia nilotica* seeds [30]. But this value of carbohydrate was considerably higher than the 8.2 ± 0.32 % reported for *Acacia sieberina* seeds by Abubakar et al. [31]. This value of carbohydrate content with protein and fat content obtained make the seeds of *Citrullus mucospermus* a better source of energy (497.01 ± 2.75 kCal) than the most of the report seeds. The amount of carbohydrate in pistachios, as in other nuts, is moderate, but pistachios are rich in fiber.

Fiber content is important because epidemiological and clinical studies have consistently demonstrated that fiber intake is associated with weight loss [32], diabetes and some types of cancer [33]. Moreover, pistachios have a low glycaemic index, which contributes to maintaining satiety longer and lowering postprandial blood glucose concentrations [34,35]. As for fibers, which are recognised for their crucial role in the proper functioning of the intestinal transit, they constitute an important fraction of *C. mucospermus*. Its consumption represents an asset in a healthy and balanced diet. Kouamé et al. [36] reported that a high-fiber diet reduces the daily insulin requirement of

diabetic patients and stabilises their glycaemic profile. Thus the fibre contained in *C. mucospermus* could be advised to diabetic patients by decreasing the rate of glucose absorption and/or delaying gastric emptying. It could also contribute to the prevention of colon cancer by binding to the cancer-causing chemicals, driving them away from from the cells lining the colon [37].

The physicochemical composition of the seeds of *Citrullus mucospermus* Fursa indicated that the seeds have a low moisture (6.40 ± 0.14 %) content and a high dry matter content (93.60 ± 2.82 %). This moisture value was low in comparison with that of Casiatora seeds (11.50 %) reported by Adamu et al. [16]. The low moisture content will give long shelf life to *C. mucospermus* seed flour. The high dry matter content of *C. mucospermus* Fursa seeds could be explained by the fact that they are oilseed cucurbites. Oilseed cucurbites are very rich in fat and protein, but they have a low moisture content in the dried state. Low moisture content does not appear to influence the enzymatic and microbial degradation process. Therefore low moisture content as saw in the sample is an evidence that this specimen may not be more inclined to decay, since nourishments with high dampness substance are more inclined to perishability [38]. According to Soro et al. [39], a low moisture content of less than 12 % would allow a better conservation of flours. Therefore, it might be profitable in perspective of the specimen timeframe of realistic usability. Indeed, according to Siddiqui and Chowdhury [40] moisture is an important parameter that significantly affects the shelf life and development of microbial contaminants in flour.

The ash content is 5.18 %. This assumes that these seeds have a good source of important minerals. This ash level is lower than those obtained by N'Dri [41] in his research on akpi almonds which is 7.32g/100g dw and those of 7.45 % reported for *Cucurbita* species [42]. This value was higher than the 2.48 ± 0.18 % reported for *Cumis melo* variant by Adekunle and Oluwo [43]. Ash is a residue of mineral compounds that persists after the incineration of organic substances of animal or vegetable origin.

The pH is a very important factor. Indeed, the pH of the *C. mucospermus* Fursa seeds obtained was 6.15 and is higher compared to those reported *Citrullus lanatus* (5.18) and *Cucumis sativus* (5.75) by Olayinka and Etejere [44]. This pH is favourable to the enzymatic activity of these seeds. The slightly acidic pH value showed clear indications that the seeds used for the study were matured.

4. Conclusion

The results of the present study showed that *C. mucospermus* Fursa seeds contained appreciable amounts of fiber, protein and crude fat and thus can be consumed for dietary purposes. The oil, extracted from the *C. mucospermus* Fursa seeds, revealed the presence of high amount of oleic, palmitic, linoleic and stearic acid as well as it exhibited low levels of saturated fatty acids, suggesting that *C. mucospermus* Fursa oil can be used as a diluent with other edible oils to enhance their essential fatty acid content i.e. linoleic acid and make its potential

food uses for health benefits. Additional research on the detailed physicochemical and bioactive properties of *C. mucospermus* Fursa seed and seed oil is crucial to explore their commercial and functional foods applications.

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