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ORIGINAL ARTICLE

Physico-Chemical Properties of Raphia Fruit (*Raphia hookeri*) Pulp from Cameroon

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Abstract

The pulp of *Raphia hookeri* fruit is gaining lot of interest from Cameroonian consumers. Oil crops are the second most valuable commodity in the world trade, hence the study of its quality is important for human safety and health. Thus, this study was carried out to evaluate the physico-chemical properties of Cameroonian *Raphia hookeri* pulp as well as its antioxidant potential and nutritional properties. The oil was extracted from the dried pulp using hexane via maceration method. The color, iodine value, acid value, peroxide value and the thiobarbituric acid value which characterize physically and chemically the oil were analyzed respectively using the AOAC (Association of Official Analytical Chemists) method which was also used for the acid value, the IDF method and the 2-thiobarbituric acid (TBA) method. From this investigation, as result, the value red color, 84.38 g/100g, 1.36 %, 50.01meq O2/ kg and 7.08 ppm were obtained respectively. A total of three antioxidant tests which are the 2, 2-diphenyl-1-picrylhydrazyl test (DPPH test), the Ferric reducing antioxidant power and the Hydroxyl radical scavenging activity were done on the extract in other to determine the antioxidant activity. The results showed that the pulp of the fruit has good radical scavenging activity thus has a powerful antioxidant potential. Also, *Raphia hookeri* fruit pulp is nutritionally rich in fiber with 53.57% of fiber, 20.96% of lipid, contain 13.77% of carbohydrate, 8% of protein, 3.70% of minerals predominated with potassium and calcium. Therefore, the pulp of the fruit cannot only be recommended as food supplement because of it rich nutrient content, but also as source of antioxidant for pharmaceutical products and source of unsaturated oil to be used for human consumption to prevent against diseases.

Practical application

Raphia hookeri pulp from Cameroon has very good nutritional and antioxidant properties, thus can be used in food formulation as supplement as well as source for natural antioxidant.

Keywords: *Raphia hookeri*, oil, antioxidant, physico-chemical properties, nutritional, proximate analysis.

1. Introduction

Raphia palm is a monocotyledonous plant belonging to the family Palmaceae. It has a trunk covered with attractive unusual coils, usually reproduces through seeds and grows up to 10 m tall and 60 cm in trunk diameter (Hutchinson *et al.*, 1993). From scientific reports and investigations, it has been shown that the origin of Raphia palms is traceable to West Africa, particularly along swampy and semi swampy area of tropical and equatorial rain forest or derived savannas (Moore, 1973; Ndon, 2003).

Endemic to Africa, its distribution covered many countries of the tropical area like Cameroon, Burkina Fasso, Nigeria, Madagascar, Gambia, Ghana, Guinea, Ivory Coast, Kenya. About 30 species are known (Ugwu & Igboeli, 2009) among cited are *Raphia farinifera*, *Raphia sudanica*, *Raphia vinifera*, *Raphia regalis* and *Raphia hookeri* which is commonly distributed in Cameroon. Raphia palm produces fruits that are oblong-ellipsoid in a scaly cone comprised of rhombus triangular reddish-brown scales (Keay & Hepper, 1953). The fruits contain an important part called pulp or mesocarp which is considered inedible in some parts of the country. In addition, it is used as a bitter flavouring or occasionally as food, particularly when fresh. Due to its stomachic and laxative properties, it is used as medicine (Liu, 2004 & Altiok, 2010).

Every part of Raphia palm tree is useful economically, both in the food industry sector and the art sector. In the food industry sector, the mesocarp of the ripe raphia fruit pulp which is rich in many nutrients such as lipid (40-52%), protein (6.1%), carbohydrate (61.4%), vitamins such as niacin (0.27 mg), vitamin A (0.15 mg) and minerals (3%), as reported by Edem *et al.* (1984) and Esiegbuya *et al.* (2013) cannot only be used as food supplement. It can also be a main source of lipid since it yields edible oil, which can be use and exploit as a cheap and local product which lead to a decrease of resource wasting and environmental pollution (Ndon, 2003).

Many studies have been done on raphia palm, some of which include: the study of vitamin, phytochemicals and toxic elements in the pulp and seed of raphia palm fruit (Ogbuagu, 2008); the qualitative and quantitative evaluation of the phytochemicals of *Raphia hookeri* and *Raphia farinifera* fruits (Oluwaniyi *et al.*, 2014); some morphological and chemical characteristics of developing fruit of *Raphia hookeri* (Bassey, 1985); the evaluation of the chemical composition of *Dacryode edulis* and *Raphia hookeri* Mann and Wendl exudats used in herbal medicine in south eastern Nigeria (Okwu & Nnamdi, 2008). Although, studies have been done on *Raphia hookeri*'s fruit, very few have interested on it pulp in terms of physicochemical properties, antioxidant potential and nutritional composition which can be a source of rich knowledge and information for the health and nutrition sector.

Moreover, the exploitation of raphia palm tree is increasing over years in Cameroon, due to the high demand of its derived products such as bamboo, raphia fiber, piassava (Ndon, 2003) in the art sector but awareness on the potential nutritive value, antioxidant activity and physicochemical properties of the pulp which are beneficial to the human body is still lacking. Therefore, the aim of this study is to evaluate the physicochemical properties, the antioxidant potential and the nutritional composition of the Cameroonian raphia (*Raphia hookeri*) pulp which will be a great benefit for the population as it can be used as nutritive food to fight against many diseases, malnutrition and enhance the economy of the population as whole as the country.

2. Materials and Methods

2.1. Material

The fresh "Tchakoum" fruits with different dimensions (figure 1a) were harvested in Dschang (Latitude: 5° 26' 38.29'' N, Longitude: 10° 03' 11.95'' E), West Region Cameroon, in November 2018. All chemicals and reagents used were of analytical reagent grade.

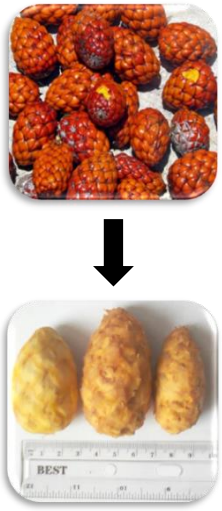


Figure 1: *Raphia hookeri* fruit (a), peeled *Raphia hookeri* fruit (b)

2.2. Methods

2.2.1. Sample preparation and processing

The fruits were peeled (figure 1b) and the flesh isolated and dried for 10 h under sunlight to constant weight. The dried flesh was used for further analyses.

2.2.2. Oil extraction

Oil was extracted from the dried pulp using the maceration method as described by Womeni *et al.* (2016). The pulp was separately grinded to pass 1 mm sieve. 80 g of each powder were separately macerated in 400 mL of hexane at room temperature for 24 h with constant shaking. After that, the mixture was filtered using the wathman paper N°1, and the filtrate was concentrated on a rotatory evaporator at 40 °C. The extracted oils were stored in the refrigerator at 4 °C for further analysis. The remaining solid fractions were dried in the oven at 50 °C for the determination of their proximate composition.

2.2.3. Extraction of "Tchakoum" polyphenols

100 g of "Tchakoum" powder was extracted with 400 mL of methanol for 48 h at room temperature. The mixture was regularly subjected to shaking during the extraction. The extract was filtered with a Whatman N° 1 filter paper, and residue was again extracted with 200 mL of methanol to ensure maximum extraction of phenolic compounds. The combined filtrates were subjected to rotary evaporation at 40 °C under reduced pressure for the removal of the solvent. The dried extract was used for further analysis.

2.2.4. Evaluation of the antioxidant activity

The ability of each extract to scavenge the DPPH radical was determined according to the method of Braca *et al.* (2002). A total of 4.5 mL of 0.002% alcoholic solution of DPPH was added to 0.5 mL of different concentrations (250, 500, 1000, and 2000 µg/mL) of samples and standard solutions separately, in order to have final concentrations of products of 25-200 µg/mL. The samples were kept at room temperature in the dark and after 30 min and the absorbance of the resulting solution was measured at 517 nm. The absorbance of the samples, control, and blank was measured in comparison with methanol. The antioxidant activity (AA) was calculated according to the formula:

$$AA\% = [(Abs_{control} - Abs_{sample}) \times 100 / Abs_{control}]$$

AA = Antioxidant activity, Abscontrol = Absorbance of the DPPH solution, Abs= Absorbance of the sample.

2.2.5. Oil characterization

The determination of the peroxide value of "Tchakoum" seed samples was made following the spectrophotometrical IDF standard method, 74A:1991 (IDF, 1991). Its iodine and acid values were determined according to the procedure of AOCS Official Method CD 1-25 and CD 3d-63 (AOCS, 2003) respectively. Finally, its thiobarbituric acid value was evaluated as described by Draper & Hadley (1990).

2.2.6. Analysis of the proximate composition of "Tchakoum"

The fat, ash and protein contents of Raphia hookeri powder was determined using standard analytical methods described by AOAC (1990) procedures. Ash content was determined by incineration of the dried pulps at 550 °C according to the AOAC procedures 942.05. Nitrogen (N) content was determined using micro-Kjeldahl method, according to AOAC procedures 984.13, the protein content was calculated as N x 6.25. Lipid content was determined using Soxhlet apparatus with hexane, following AOAC 963.15 methodology. The total percentage carbohydrate content was determined by the difference method as reported by Onyeike *et al.* (2015). This method involved adding the total values of crude protein, crude fat and ash constituents of the sample and subtracting it from 100. All samples were analyzed in triplicate.

2.2.7. Determination of the Mineral content of "Tchakoum"

For the determination of minerals, *Raphia hookeri* pulps were ashed at 550 °C and dissolved with 10 mL of 20% HCl in a beaker and then filtered into a 100 mL standard flask to determine the mineral content. Calcium (Ca), magnesium (Mg), sodium (Na), potassium (K) and iron (Fe) were determined by atomic absorption spectrometer (Varian 220FS Spectra AA, Les Ulis, France). Phosphorus (P) was determined colorimetrically using the vanadomolybdate, according to AOAC procedure 965.17 (AOAC, 1999). Mineral contents of the samples were determined from calibration curves of standards minerals. All samples were analyzed in triplicate.

2.3. Statistical analysis

Results obtained in the present study were subjected to one-way analysis of variance (ANOVA) with Student-Newman-Keuls tests using Graphpad-InStat version 3.05, to evaluate the statistical significance of the data. A probability value at $p<0.05$ was considered statistically significant.

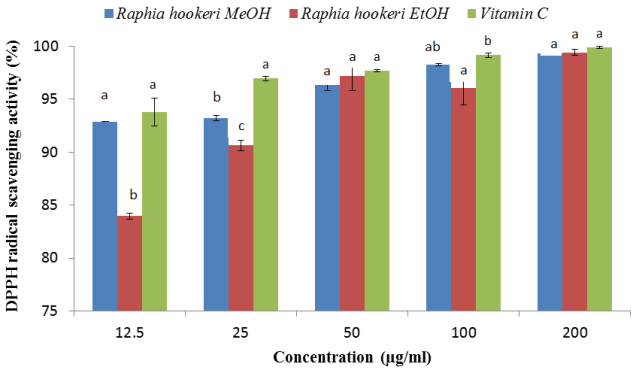
3. Results and Discussions

3.1.1. DDPH Radical Scavenging Activity

The DPPH radical scavenging activity of *Raphia hookeri* extract compared to Vitamin C is presented in Figure 2. It can be observed that, at almost all concentrations, the activity of methanolic extract was close to that of Vitamin C. Similar results were registered with the ethanolic extract but at concentrations 50, 100 and 200 µg/mL. However, at concentration 12.5 and 25 µg/mL, its activity was significantly lower ($p <0.01$) compared to that of the methanolic extract and Vitamin C. Generally, the activity of the extracts and Vitamin C was increasing with the concentration.

The fact that the antioxidant activity of the methanolic extract was close to that of vitamin C at almost all concentration is a proof of its

efficiency. Similar observations were made with the ethanolic extract but at concentration 50 to 200 µg/mL. The lowest activity registered with



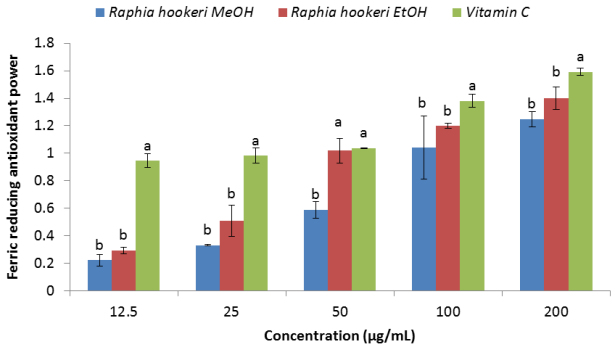
Values are presented as Mean ± SD. a-cValues of the same concentration with different superscripts are significantly differents (p<0.05)

Figure 2: DPPH Radical scavenging activity of *Raphia hookeri* extracts

The ethanolic extract compare to the methanolic extract at concentration 12.5 and 25 µg/mL can be explained by the nature of the solvent used. It has been demonstrated that methanol has strong extraction strength than other solvent. It has the power to extract more molecules from substrates. That is the reason why it is usually recommended for the extraction of antioxidant compound (Iqbal *et al.*, 2005). Bukhari *et al.* (2009), showed that the antioxidant activity of polar solvents extract is greater than that of less or non-polar solvent extract. Results obtained in this study showing that the methanolic and ethanolic extract of *Raphia hookeri* have good radical scavenging activity are in accordance with the report of Oluyori *et al.* (2018), who reported that the ethanolic extract of leaves and epicarp methanolic extract of *Raphia hookeri* from Nigeria range from 0 to 90%. They also pointed out that the epicarp and leave of the ethanolic extract of the same plant are rich in phenolic compound with a total phenolic content of 459.917 and 457.805 (mg/g GAE) and total flavonoid content of 97.660 and 351.170 (mg/g RE) for the epicarp and leave respectively. These molecules can be suspected as responsible of the activity obtained with the pulp of the same plant in this study.

2.1.2. Ferric Reducing Antioxidant Power

The ferric reducing antioxidant power of *Raphia hookeri* methanolic and ethanolic extracts compared to vitamin C is presented in Figure 3. Generally, the activity of all analyzed samples increases with the concentration. The lowest activity was registered with the methanolic extract of *Raphia hookeri*. However, at all concentrations (except 50 µg/mL), its activity was similar (p>0.05) to that of the ethanolic extract. The highest activity was registered with the synthetic antioxidant (vitamin C).



Values are presented as Mean ± SD. a-bValues of the same concentration with different superscripts are significantly differents (p<0.05)

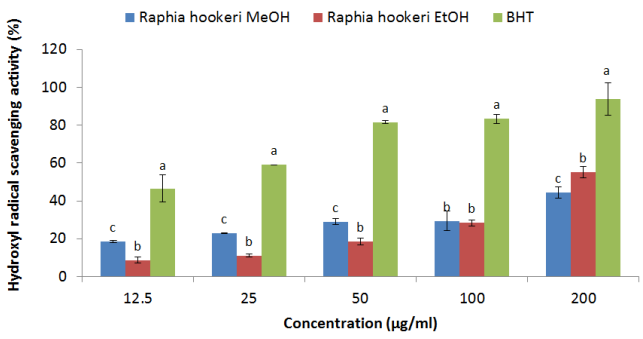
Figure 3: Ferric reducing antioxidant power of *Raphia hookeri* extracts

This suggests that, the molecules able to reduce ferric irons are present in both extracts at almost all levels. The fact that their activity is lower than that of vitamin C can be attributed to impurities presence. The activities observed can

be attributed to the presence in these extract of phenolic antioxidant, with ferric reducing ability. This activity can be attributed to the presence of flavonoids which were found to be present in the epicarp and leaf extract from the same plant in Nigeria by Oluyori *et al.* (2018), who found that the flavonoids content of this plant parts extract is respectively 97.66 and 351.17 (mg/g RE). Similar results showing that plant extract can be good ferric reducer have already been reported (Womeni *et al.*, 2016; Kingne *et al.*, 2018; Silenou *et al.*, 2018).

2.1.3. *Hydroxyl Radical Scavenging Activity of Raphia hookeri extracts*

The hydroxyl radical scavenging activity of *Raphia hookeri* methanolic and ethanolic extracts compared to butylated hydroxytoluene is presented in Figure 4. The trend of the activity of all analyzed samples was significantly increasing ($p < 0.05$) with the concentration. The highest hydroxyl radical scavenging power was recorded with the synthetic antioxidant (BHT) followed by the methanolic extract of *Raphia hookeri*. The ethanolic extract exhibits the lowest activity.



Values are presented as Mean ± SD. a-cValues of the same concentration with different superscripts are significantly differents ($p<0.05$)

Figure 4: Hydroxyl radical scavenging activity of *Raphia hookeri* extracts

Generally, the activity of both extracts was low compared to that of the synthetic antioxidant. This can be explained by the classes of antioxidant present, as their mechanism of action may be different from that of molecules similar to BHT. Even though the hydroxyl radical scavenging activity is low, the potential of extracts obtained from the same plant in other countries in scavenging free radical has already been proven (Oluyori *et al.*, 2018).

3.1.4. *Initial characteristics of Raphia hookeri fruit pulp oil*

The initial quality parameters of *Raphia hookeri* pulp oil are presented in Table 1. Results showed that, the oil extracted from *Raphia hookeri* fruit pulp has a peroxide value of 50.01meq O_2/ Kg, a thiobarbituric acid value of 7.08 ppm, an iodine value of 84.38 g /100 g, an acid value of 1.36% and was red in color.

The color obtained in this study, is similar to that reported in *Raphia vinifera* seed pulp oil by Igwenyi *et al.* (2007) (Red). The acid value obtained in this study (1.36%) was significantly lower than that reported by Igwenyi *et al.* (2007), who obtained an acid value of 5.05% with *Raphia vinifera* seed pulp oil. The acid value obtained in this study was significantly lower than 4.0% which is the recommended acid value for cooled pressed and virgin oils as stated by (Codex Alimentarius, 1999). The iodine value shows that *Raphia hookeri* pulp oil is rich in unsaturated fatty acid which justify its liquid attitude at room temperature. The iodine value obtained in this study was significantly higher than that reported by Igwenyi *et al.* (2007), who found a value of 69.41 (g/100g) using similar method. These same authors in 2008 showed that the iodine value of *Raphia vinifera* seed pulp oil is ranged between 69.41 and 76.52 which is not

too far from 84.38 (g/100g) obtained in this study. And oil with such an iodine value can be good source of essential fatty acid. The peroxide value obtained in this study was almost 3 times the value recommended by the norm (Codex Alimentarius, 1999) for cooled press and virgin oils which should not be more than 15 (meq O2/ kg) of oil. The significant differences observed can be attributed to the method used to determine the PV of *Raphia hookeri* oil. The method used here was the spectrophotometrical technic of the IDF (1991). In this method, peroxide oxidizes the Fe^{2+}, which forms a red Fe^{3+} complex with Ammonium Thiocyanate. It is important to mention that the color of the *Raphia hookeri* pulp oil is red, which can easily interfere in the absorbance and lead to an over estimation of the hydroperoxide present. The thiobarbituric acid values generally quantify and give an idea of the secondary oxidation state of oil and precisely the amount of malondialdehyde present. From our knowledge there is no standard with the thiobarbituric acid value as far as vegetable oil are concerned. However, TBA value of 5 ppm in fish oil is a sign of its freshness (Ukekpe *et al.*, 2014).

Table 1: Quality parameters of *Raphia hookeri* fruit oil

Quality parameter	
Peroxide value (meq O_2/Kg)	50.01±0.14
Thiobarbituric acid value (ppm)	7.08±0,96
Iodine value (g/100 g)	84.38±3.84
Acid value (%)	1.36±0.36
Color	Red

3.1.5. Proximate composition of Raphia hookeri fruit pulp oil

The proximate composition of *Raphia hookeri* fruit pulp shows that it contains 20.96 % of lipid, 8 % of protein, 53.57% of fiber, 3.70% of ash and 13.77% of carbohydrate (Table 2). The energy value provided by this sample is 275.72 Kcal.

Table 2: Proximate composition of *Raphia hookeri* fruit pulp

Parameter	
Lipids (%)	20.96±0.21
Proteins	8.00±0.06
Fibers (%)	53.57±1.44
Ash (%)	3.70±0.02
Carbohydrates	13.77±0.12

The amount of ash obtained in this study, was closed to that obtained by Edem *et al.* (1984) and Esiegbuya *et al.* (2013) who respectively obtained a value of 3.0% and 4.33 % with *Raphia hookeri*. However, the lipid and protein content were significantly higher than that reported by these authors. Edem *et al.* (1984) obtained 11.8 % and 6.1% respectively for the lipid and protein content; while Esiegbuya *et al.* (2013) obtained 8.2% and 0.95 ppm. the crude fiber content was however similar to 56.15 as reported by Esiegbuya *et al.* (2013) but significantly higher than 17.7 obtained by Edem *et al.* (1984). Edem *et al.* (1984) has also shown that, the carbohydrate content of fruit of Raphia palm is 61.4% which is significantly higher than 13.77% obtained in this study. Similar observation was made with the total energy provided which was 215.72 Kcal compared to

that reported by Edem *et al.* (1984) (380.5 Kcal). The differences observed in the proximate composition of *Raphia hookeri* fruit compared to the data from the literature can be related to the origin of the fruit.

3.1.6. Mineral composition of Raphia hookeri fruit pulp

The mineral composition of *Raphia hookeri* fruit pulp is presented in Table 3. From that table, *Raphia hookeri* contains 0.08 mg/100 g of iron, 50 mg/100g of phosphorus, 376 mg/100g of calcium, 34 mg/100 g of magnesium, 1562 mg/100 g of potassium and 137 mg/100 g of sodium.

Table 3: Mineral composition of *Raphia hookeri* fruit pulp

Mineral	
Iron (mg/100 g)	0.08±0.00
Phosphorus (mg/100 g)	50±3.22
Calcium (mg/100 g)	376±7.10
Magnesium (mg/100 g)	34±2.13
Potassium (mg/100 g)	1562±2.06
Sodium (mg/100 g)	137±1.88

The mineral content shows that *Raphia hookeri* pulp is very poor in iron. This result is in accordance with that reported by Edem *et al.* (1984) who obtained a value of 0 with the same plant part in Nigeria. The potassium content was significantly higher than that reported by these authors but significantly lower than the amount recommended which is 4700 mg/day (WHO, 1982). As far as the phosphorus is concerned its concentration was significantly lower than 77.8% reported by Edem *et al.* (1984). Similar observation was made with calcium and magnesium, where these authors obtained 875% of calcium and 315% of magnesium. However, the sodium content obtained in this study was significantly higher than 16% obtained by these same authors. These minerals are very important as they have several biological functions. A deficiency in them generally lead to nutritional disorders.

3.1.7.Fourier-Transformed-Infrared Spectroscopy of Raphia hookeri fruit pulp oil

The Fourier-Transformed-Infrared Spectrum (FT-IS) of *Raphia hookeri* fruit pulp oil was done and the result is presented in figure 5.

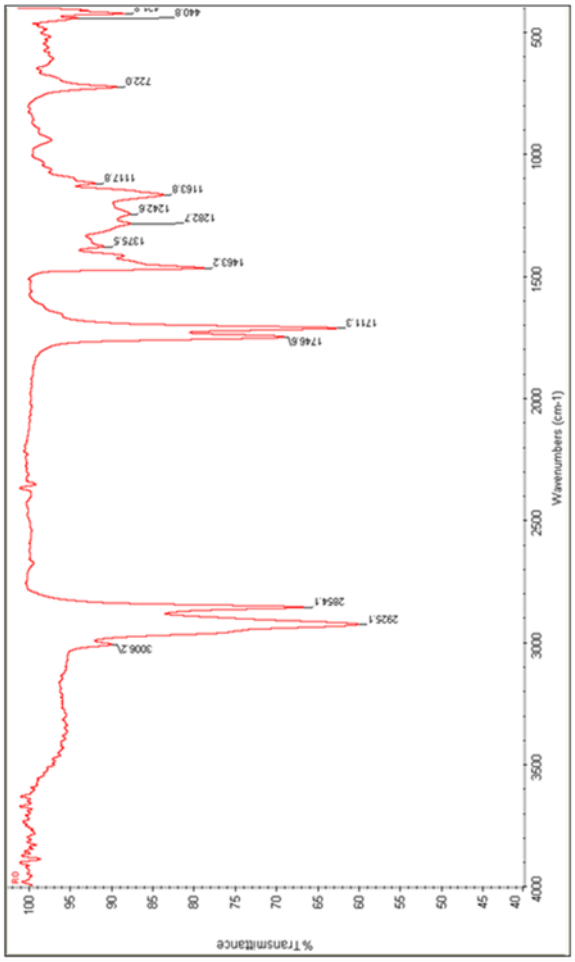


Figure 5: FT-IR spectrum of *Raphia hookeri* fruit pulp oil

Bands at 2925.1 and 1711.3-1746.5 cm^{-1} are respectively due to the vibration of =C-H (cis) and -C=C- (cis) (Vlachos *et al.*, 2006). These bands suggested of the presence of unsaturated fatty acids which can justify the high peroxide and iodine values obtained for the *Raphia hookeri* fruit pulp oil. The peaks appearing in the range of 1100-1500 cm^{-1} (1117.8, 1163.8, 1242.6, 1282.7, 175.5 and 1463.2 cm^{-1}) mark the presence of stretching vibrations of the C-O group in esters and asymmetric coupled vibrations C-C(=O)-O and O-C-C of triglyceride and fatty acid (Silverstein *et al.*, 1974, Vlachos *et al.*, 2006). Finally, the band pointing out at 722 cm^{-1} indicate the overlapping of the methylene (CH_2) rocking vibration and the bending vibration of cis-disubstituted olefins (Silverstein *et al.*, 1974).

4. Conclusion

The objective of this study was to evaluate the nutritional composition, the physico-chemical properties and the antioxidant potential of Cameroonian *Raphia hookeri* pulp. From the results, it can be concluded that the methanolic and ethanolic extract of *Raphia hookeri* have good antioxidant can therefore be used naturally to fight against oxidative stress. The Cameroonian *Raphia hookeri* pulp content a considerable amount of oil with high iodine value and its Fourier-Transformed-Infrared Spectrum shows the presence of double bonds carbon-carbon, which encouraged that further research should be carried out about its essential fatty acid content.

Conflict of interest

The authors declare that there are not conflicts of interest.

Ethics

This Study does not involve Human or Animal Testing.

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