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Mineral Composition, Antioxidant Properties, Phytochemical and Anti-nutrient Composition of African Palmyra Palm (*Borassus aethiopum)* Fruit Flour

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Abstract African palmyra palm (*Borassus aethiopum*) grows widely across Africa. Previous studies indicated its fruit flour has a great potential in food applications. The objective of this work was to investigate the effect of different drying methods on the mineral composition, antioxidant properties, anti-nutrient composition and phytochemical composition of the African palmyra palm (APP) flour. The fresh fruit pulp was obtained, freeze dried, oven dried, solar dried and milled into flour. Phytochemical screening, mineral analysis, anti-nutrient analysis and antioxidant analysis were conducted on the flour obtained according to standard methods. The flour had high total phenols (1518.00 - 3896.71 mg GAE/100g), potassium (237.00 - 276.73 mg/100g), magnesium (211.61 - 293.62 mg/100g) and saponin (36.10 – 55.62 g/100g). The flour samples also had considerable free radical scavenging activities. Phytochemical screening indicated the presence of several phytochemicals including glycosides. Drying had a significant effect on the analysed composition of APP flour.

Keywords: *Borassus aethiopum, polyphenol, DPPH scavenging activity, drying*

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1. Introduction

Borassus aethiopum, a member of the *Arecaceae* family is known in Africa as the mother of trees or as the Savannah's guard [1]. The common names of *Borassus aethiopum* include African fan palm, African palmyra palm or deleb palm [2]. The African palmyra palm (*Borassus aethiopum*) is synonymously referred to as the Asian palmyra palm (*Borassus flabellifer*) in some publications. However, a clear observation indicates a distinction between these palms. The African palmyra palm is distinguished from the Asian palmyra palm by its rigid, numerous, less deeply folded leaves and a ventricose stem according to Dransfield (1986) as cited in Sambou *et al*. [3].

Borassus aethiopum is documented as being native to countries including Benin, Burkina Faso, Congo, Cote d'Ivoire, Ethiopia, Gambia, Ghana, Guinea, Kenya, Liberia, Nigeria and Sierra Leone [2]. In Ghana, *Borassus aethiopum* is dominant in some parts of the Volta, Eastern, Ashanti and Brong Ahafo regions. A survey conducted by Siaw *et al*. [4] in the Mampong Forest District of Ghana indicated the population density of *Borassus aethiopum* was about 18-61 trees per hectare.

The African fan palm has varied usefulness on the ecological, nutritional and therapeutic front. The leaves are useful in the basket and mat industries whereas the trunk is useful in the construction of bridges, telegraphic poles due to the toughness and termite resistant nature of the wood [5]. According to Sambou *et al*. [3], upon ripening, the mesocarp is fleshy and can be prepared and consumed by grilling, boiling or mixing with sugar or honey. The ripened fresh pulp of *Borassus aethiopum* are also separated, mashed and eaten boiled with corn or porridge in the rural areas of Ghana.

Borassus aethiopum fruits have been reported to contain 79 – 81% moisture, 107.61-108.25 mg/100g calcium, 107.61-108.25 mg/100g phosphorus, 20.61-21.01 mg/100g magnesium and 29.75% crude fibre according to Ali *et al*. [1] and Djibrilla [6] respectively. Another study by Waziri *et al*. [7] indicated the fruit contains relatively high levels of potassium (205.00-275.00 mg/100g) and sodium (202.40 – 220.80 mg/100g). *Borassus aethiopum* fruit flours produced in the study of Abe-Inge *et al*. [8] had high crude fibre (14.04 – 19.52 %), water absorption (307.18 – 517.66 %), oil absorption (83.48 – 164.38 %) and swelling (433.00 – 556.92 %) capacities. These findings indicated *Borassus aethiopum* fruit flour has great potential applications in the food industry. However, the mineral content, anti-nutrient content, phytochemical

and antioxidant properties of the flour produced from the African Palmyra palm fruit was not determined. Information on these properties of the flour may help increase and diversify the utilization of the *Borassus aethiopum* fruit flour in the food sector of Ghana as well as reduce wastage of *Borassus aethiopum* fruits.

In the present study, the mineral content, anti-nutrient composition, phytochemical and antioxidant properties of *Borassus aethiopum* fruit flour produced from oven, freeze and solar drying methods were investigated.

2. Materials and Methods

2.1. Source of Sample and Flour Preparation

Fresh African palmyra fruits were obtained from Congo 3 in the Ejura in the Sekyere Central District of Ghana. The fruits were decalyxed, washed, peeled and pulped manually using a knife. The obtained pulp was further cut into smaller sizes and divided into three portions with each portion weighing about 600 g. The three portions were dried differently using solar drying at 37–39 °C for 7 h, hot air oven drying (Binder Heating and Drying Oven, Tuttingen, Germany) at 60 °C for 4 h and vacuum freeze drying (vacuum freeze dryer, model: YK-118-50, Taiwan) for 72 h (at -47 °C to -55 °C, 0.002 to 2.7 Torr). Drying was continued until moisture content was reduced to levels below 10 %. The dried samples were milled using the kitchen blender at speed 2 whilst pausing after every 30 s for a total time of 3 minutes. The milled samples (≤ 450 microns) were packaged in zip-loc bags and stored in a freezer (Protech PRCF-500, China) for analysis.

2.2. Mineral Determination

Samples were digested according to AOAC [9] with modifications and mineral (Na, Ca, Fe, Mg, K, Zn, Cu, Mn, Pb, Cd) content determined using Atomic Absorption Spectroscopy (AAS). About 0.25 g of each flour sample was weighed into Kjedahl digestion tubes and 7.5mL of concentrated H_2SO_4 was added, followed by the addition of 2.5 mL of concentrated HNO_3. The samples were digested at 300 °C for 4 h until the initial colour of the solution cleared (initial dark color changed to colourless). The clear solution was cooled to room temperature and diluted with 50 mL distilled water. The diluted solution was warmed to vaporize and atomize the mineral components prior to their quantification with the Atomic Absorption Spectrophotometer (AAS).

2.3 Determination of Antioxidant Properties and Phytochemical Screening

2.3.1. Extract Preparation

Methanolic extracts of the flours were prepared by weighing 20 g of each flour sample into a conical flask and 50 mL methanol was added. The mixture was allowed to stand at room temperature for 48 hours with periodic manual shaking. The liquid extract was first filtered using Whatmann filter paper No.42, followed by filtration using cotton. The residue obtained was dried to obtain a powdered extract using a rotary evaporator with a water bath set at 40 °C. The powdered extract was used for both the phytochemical screening, determination of antioxidant activity and total phenol content.

2.3.2. Determination of DPPH Scavenging Activity

DPPH scavenging activity was determined according to the modified DPPH assay method by Larbie *et al*. [10]. Stock solution of the extract was prepared by dissolving 10 mg of the dried sample in 1.0 mL of methanol. Also, stock solutions of 10 mM of standard (Ascorbic acid) and 0.5 mM of DPPH were prepared by dissolving 0.176 mg of Ascorbic acid and 3 mg of DPPH in 1.0 mL of water and 15 mL absolute methanol respectively. The solutions were then vortexed until complete dissolution was achieved. The DPPH solution was immediately kept in the dark as it photo-bleaches in light.

In a 1.5 mL eppendorf tube, the extract was serially diluted in water to obtain a concentration range of 0.156-10 mg/mL. Hundred microliters of each concentration of the test sample was transferred into a 96 well plate. This was followed by the addition of 100 μL of 0.5 mM 2, 2-diphenyl-1-picrylhydrazyl radical (DPPH). For positive control or standard, ascorbic acid was used at a concentration range of 0.156–10 mg/mL in distilled water. Distilled water was used as blanks. Triplicate experiments were performed. The plates were covered with aluminum foil, shaken gently and kept in the dark for 20 min after which the absorbance was read on a Synergy H1 plate reader at the absorbance wavelength of 517 nm. Percentage scavenging activity was determined by;

$$\%\ Scavenging = \frac{\left[\begin{array}{l} Absorbance\ of\ blank\ (OD\mathbf{0}) \\ -\ Absorbance\ of\ test\ (OD\mathbf{1}) \end{array}\right]}{Absorbance\ of\ blank\ (OD\mathbf{0})} \times 100.$$

The inhibitory concentration at 50 % (IC50) values, which is the amount of antioxidant necessary to decrease the initial DPPH concentration by 50 %, were determined by nonlinear regression analysis.

2.3.3. Determination Total Phenol Content

The total phenol content of *B. aethiopum* fruit flour extracts was determined according to the modified Folin Ciocalteau method by Larbie *et al*. [10]. Stock solution of the extract was prepared by dissolving 10 mg of each of the dried samples in 1 mL methanol. A stock solution of 5 mg/mL of standard (gallic acid) was prepared by dissolving 50 mg of it in 1 mL absolute ethanol. This was then diluted in 9 mL distilled water to obtain the 5 mg/mL stock solution.

A two fold serial dilution was carried out on the gallic acid standard to obtain six different concentrations 5, 2.5, 1.25, 0.625, 0.3125 and 0.15625 mg/mL. A water blank, that is, water without gallic acid, was also prepared. A two fold serial dilution was also carried out on the extract to obtain three different concentrations (10, 5, 2.5 mg/mL). Water without extracts, was also prepared as blank.

A volume of 10 µL of the sample and gallic acid dilutions were aliquoted into a 2.0 mL eppendorf tube. Aliquots of 790 µL of distilled water were then added and

this was followed by the addition of 50 µL of Folin-Ciocalteau reagent. The mixture was mixed thoroughly by vortexing for five seconds. This was followed by incubation of the tubes in darkness at room temperature for eight minutes. Afterwards, a volume of 150 µL of 7 % sodium carbonate solution was added to each tube, mixed thoroughly by vortexing for five seconds and further incubation of the tubes in darkness at room temperature was done for two hours. After the two hour incubation, a volume of 200 µL of the extract and gallic acid standard dilutions was aliquoted into wells on a 96-well plate in triplicate and absorbance read at 750 nm using microplate spectrophotometer (Synergy H1). A graph of absorbance against concentration was plotted for the gallic acid standard. The concentration of phenolics in the extract was determined using the gallic acid standard plot.

2.3.4. Phytochemical Screening

Phytochemical screening on flour extracts were carried out according to the methods described in the works of Trease and Evans [11], Onike [12] and Tiwari *et al*. [13].

2.3.4.1. Test for Tannins

Two hundred milligrams of each flour extract was weighed and warmed with 20 mL distilled water in a water bath for 5 minutes and filtered. To 1 mL of the extract solution, 10 mL distilled water was added and 3 drops of 1 % lead acetate solution was added. The presence of colored precipitates the presence of tannins.

2.3.4.2. Test for Alkaloids

Two hundred milligrams of each flour extract was weighed, extracted with 50 mL ammoniacal alcohol (1 part concentrated ammonia: 9 parts of 95 % ethanol) and filtered. The filtrate was evaporated on a water bath to dryness. The scum obtained was extracted with 1 % H_2SO_4 and filtered. Dilute ammonia solution was added to the filtrate obtained to make it alkaline, then mixed with chloroform and shaken in a separating funnel. The chloroformic layer was separated and evaporated to dryness in a water bath. Then dried residue was dissolved in 1 % H_2SO_4 and 3 drops of Dragendorff's reagent (potassium bismuth iodide solution) were added. An orange-red precipitate indicates the presence of alkaloids.

2.3.4.3. Test for Terpenoids

Using Salkowski's test, a liquid chloroformic extract of each sample was prepared by adding 0.5g of the flour extracts into 2 mL chloroform. Concentrated H_2SO_4 (3 mL) was added gently down the side of the test tube to form a layer. A reddish-brown ring at the interface indicates the presence of triterpenoids.

2.3.4.4. Test for Flavonoids

Two hundred milligrams of each flour extract were macerated with 20 mL distilled water and filtered. A strip of filter paper was dipped into the extract and allowed to dry. It was then exposed to concentrated ammonia solution, an intense yellow color which disappears when exposed to fumes of concentrated HCl indicates the presence of flavonoids.

2.3.4.5. Test for Sterols

Using the Liebermann-Buchard test, a chloroformic extract was prepared for the powdered flour extract samples and filtered. To 5 mL of the filtrate, acetic anhydride was added followed by concentrated H_2SO_4 carefully down the side of the test tube to form a lower layer. A bluish color at the interface indicates the presence of a steroidal ring.

2.3.4.6. Test for Phenols

Two hundreds milligrams of the powdered flour extracts was dissolved in 1 mL of distilled water in a test tube. Two drops of $FeCl_3$ solution was added. A blue, green, red or purple color indicated the presence of phenols.

2.3.4.7. Test for Glycosides

An aqueous solution of the powdered flour extracts were prepared by dissolving about 200 mg of each sample in 1 mL distilled water in a test tube. Few (2) drops of aqueous NaOH were added. The observation of a yellow coloration indicated a positive response for glycosides.

2.3.4.8. Test for Cardiac Glycosides

To 0.5 g of extract diluted to 5 mL in water was added 2 mL of glacial acetic acid containing one drop of ferric chloride solution. This was underlayed with 1 mL of concentrated sulphuric acid. A brown ring at the interface indicated the presence of a deoxysugar characteristic of cardenolides.

2.3.4.9. Test for Saponins

To 0.5 g of the flour extracts in a test tube, 5 mL of distilled water was added. The solution was shaken vigorously and observed for a stable persistent froth which indicates the presence of saponins.

2.3.5. Determination of Antinutrients

2.3.5.1. Determination of Oxalate

Oxalate content of the flour samples were determined according to Day and Underwood [14]. About 0.2 gram of each sample was weighed and 40 mL of 1.5N H2SO4 was added. The mixture was stirred intermittently for 1 h and then filtered with Whatman No 1 filter paper. The filtrate obtained was titrated against while hot against 0.02 M KMnO4 until a faint pink colour was observed which indicated the end point. The oxalate content was calculated as follows;

$$\%\ Oxalate\ Content = \frac{0.004 \times 0.006303 \times \text{titre value}}{0.02 \times \text{Sample weight}} \times 100.$$

2.3.5.2. Determination of Alkaloids Content

Alkaloid content of the flour samples were determined using the method Harborne [15]. Two grams of each sample was weighed into conical flask. Then 50 mL of 10 % acetic acid in ethanol was added. The mixture was shaken and allowed to stand for 4 h and filtered using a Whatman No. 2 filter paper. The filtrate was then evaporated for 3min over a hot plate to about ¼ of its original volume. About 8 mL of (1+1) NH4OH was added dropwise to precipitate the alkaloids. The precipitate was

then filtered off with a preheated and weighed Whatman No. 1 filter paper and washed with 5 mL of 1 % NH_4OH solution. The precipitate in the filter paper was dried in an air oven at 60 °C for 30 min and reweighed. The alkaloid content was calculated as follows;

$$\%\ Alkaloid\ Content = \frac{\text{Difference in weight of filter paper}}{\text{Sample weight}} \times 100.$$

2.3.5.3. Determination of Tannin Content

Tannin content was determined according to Fagbemi *et al.* [16]. About 0.2 g of each sample was weighed and 10 mL of 70 % acetone was added. The mixture was shaken for 15 min to extract the tannins and then filtered. About 2.0 mL of 20 % Na_2CO_3 solution was added followed by the addition of 2.5 mL Folin-Ciocalteau and topped up with 70 % acetone. The mixture was incubated at room temperature for 40 minutes. The absorbance readings were taken at 700 nm using a spectrophotometer. A calibration curve was drawn with standard tannin acid solutions with concentrations 1-5 ppm. The standard calibration curve was used for calculating the tannin content in each sample.

2.3.5.4. Determination of Saponin Content

Saponin content was determined according to Nwosu [17]. Two grams (2 g) of each sample was weighed and defatted with petroleum ether for 8 hours using the Soxhlet extractor. The defatted samples were further extracted with methanol for 8 h using pre-heated, cooled and weighed flasks. The extract obtained were dried in the flask on a hot plate, allowed to cool and weighed. The saponin content was calculated as follows;

$$\%\ Saponin\ Content = \frac{\text{Difference in weight of flask}}{\text{Weight of sample}} \times 100.$$

2.4. Beta-Carotene Determination

Carotene was extracted from the *B. aethiopum* fruit flour samples according to the method by Khalil and Varananis (1996) as described by Ahamad *et al.* [18]. About 2.5 g of each flour sample was weighed and homogenized in 50 mL of acetone. The resulting extract was filtered through Buchner's funnel. The residue was washed twice with acetone until it became colorless. The residue was discarded and the filtrate was combined with 20g of anhydrous sodium sulphate. The anhydrous sodium sulphate was removed through filtration and the filtrate (extract) obtained was concentrated to reduce the initial volume 50 % using the rotatory evaporator. The extract was transferred quantitatively to 100 mL volumetric flask and diluted to up to the mark with acetone and distilled water, so that the final extract solution was 80% of acetone.

Beta carotene and standard calibration curves were done according to Ahamad *et al.* [18]. Five (5) beta carotene standard solutions with concentrations 0.015625, 0.03125, 0.0625, 0.125, and 0.25 µg/g were prepared. Each working standard solution was injected into HPLC system (Agilent 1100 series K15 A306 HPLC 12). Peak identification and quantification was made by "CSW 32 software" for HPLC system. HPLC was calibrated by running mobile phase (hexane and acetonitrile in the ratio 95:5 with 2 drops of triethylamine) at the rate of 2 mL per minute. The wave length was fixed at 452 nm. The pressure of the column was kept 1800-2000 PSI. Twenty microliters (20 µL) of each beta carotene standard solution was injected when the injector was in load mode. Concentrations of the beta carotene standards were plotted against the obtained peak areas to obtain a standard calibration curve. Twenty microliters (20 µL) of beta carotene extract of each flour in 80 % acetone was used for HPLC analysis just as the standard. The peaks of the samples were automatically identified and quantified by comparing their retention times with the standard retention time.

2.5. Statistical Analysis

The means and standard deviations of all replicated quantifications for each test parameter were calculated using Excel 2013. Using the Statistical Package for Social Sciences (SPSS, IBM SPSS Statistics v20), one-way analysis of variance (ANOVA) was employed to compare the means of all determined parameters. Multiple comparison of all test parameters was carried out using Tukey's test. All statistical tests were carried out 5 % significance level.

3. Results and Discussion

3.1. Mineral Composition

Drying had a significant effect on all the determined minerals of the flour samples except sodium. The concentrations of cadmium, copper, lead, manganese, and zinc were below the level of detection (0.002 mg/100g). The solar dried samples recorded higher values for all the minerals except for iron and sodium. The calcium and iron contents of *B. aethiopum* fruit flours in this study as shown in Table 1 are lower than the 107.61-108.25 mg/100g and 2.05-2.15 mg/100g iron, respectively in the study of Ali *et al.* [1] for the fresh *B. aethiopum* fruit pulp. However, the magnesium content in this study was higher than the 20.61-21.01 mg/100g for the fresh fruit pulp [1]. These discrepancies could be attributed differences in climatic factors including soils on which *B. aethiopum* fruit grows. Soils at different geographical regions have different mineral composition hence influence the mineral composition of crops grown on them.

The potassium content of the *B. aethiopum* fruit flour ranged from 237.00 to 276.73 mg/100g. There was a significant difference among the different drying methods with freeze-drying recording the least value and solar drying recording the highest value. These values were found to be lower than that of *A. altilis* flour reported as 673.5 mg/100g by Appiah *et al.* [19] but higher than a composite flour made of tigernut flour and HQCF (45.98 to 121.1 mg/100g) and white maize flour (32.80 to 34.90 mg/100g) [20]. The potassium content of the freeze dried sample is comparable to that (236.7 mg/100g DW) of *B. aethiopum* shoots [21]. The results obtained indicated *B. aethiopum* fruit flour is a relatively good source of potassium and may help in maintaining electrolyte balance in humans [22].

Table 1. Mineral Content (mg/100g) of *Borassus aethiopum* Fruit Flour

Mineral	Freeze Dried Flour	Oven Dried Flour	Solar Dried Flour
Potassium	237.00 ± 0.39^a	268.44 ± 1.27^b	276.73 ± 2.17^c
Magnesium	211.61 ± 11.29^a	211.76 ± 7.66^a	293.62 ± 4.56^b
Iron	0.60 ± 0.00^a	4.45 ± 0.02^b	0.35 ± 0.00^c
Calcium	68.21 ± 1.35^a	80.84 ± 0.84^b	95.92 ± 0.36^c
Sodium	28.92 ± 1.46^a	33.38 ± 1.18^a	31.75 ± 2.47^a

Values are means of three replicates. Values in same row with same superscripts are not significantly different (p>0.05). Zinc, Copper, Manganese, Lead (heavy metal) and Cadmium (heavy metal) were all below the level of detection (0.002mg/100g).

The magnesium content of the solar dried flour was significantly different from the oven dried flour and freeze dried flour. The *B. aethiopum* fruit flour irrespective of the different drying methods has higher magnesium content compared to the 90.63 mg/100g recorded for *A. altilis* flour [19], 36.58-37.71 mg/100 g recorded for cassava flour [23] and 9.40 to 10.58 mg/100 g reported for tigernut flour-HQCF blends [24]. Magnesium has been reported to be part of an essential enzyme system in the body and helps maintain electrical potential in nerves [25], therefore *B. aethiopum* fruit flour is a very good source of magnesium and can be composited with flours with low magnesium contents due to its high magnesium content.

The iron content of the oven dried flour was significantly higher than the freeze dried and solar dried flour. The highest iron content exhibited by the oven dried sample could be as a result of the higher temperature effect of the oven drier thereby increasing the concentration of the iron present in the *B. aethiopum* fruit flour. This is corroborated by a study done by Lahav and Turner [26] where increase in temperature resulted in three-fold increase in iron concentration. The iron content (0.35-4.45 mg/100g) of the *B. aethiopum* fruit flour was lower than 32 mg/100 g cassava flour [27] and is also below the recommended daily allowance of iron which is between 8-18 mg /100g as stated by National Academy of Science [28]. Iron is required for the synthesis of hemoglobin and myoglobin, which are oxygen carriers in the blood and muscle respectively. Although the iron content was relatively low, the oven dried flour could help meet the recommended daily intake of iron in the body when consumed.

The calcium content ranged from 95.92 to 68.21 mg/100g with solar dried flour recording the highest value and freeze drying recoding the least. The calcium level in the flour was higher than that of *A. altilis* flour (52.50-60.83 mg/100 g) reported by Appiah *et al. [19]* but lower the cassava-tigernut flour blends (138 to 214.3 mg/100g) in a study conducted by Adebowale *et al.* [24]. The relatively high calcium content of the flours in this study indicated the flours are a good source of calcium and its consumption could help reduce the risk of osteoporosis and provide stronger bones in humans.

The sodium content of the flour was not affected by any of the drying methods thus there was no significance difference among the sodium contents of *B. aethiopum* fruit flours. The sodium content of the flour in this study is comparable to cassava flour (36-50 mg/100g) as reported by Charles *et al*. [29] but lower than sweet potato (54 mg/100g) [30] and *A. altilis* flour (69.00 mg/100g). Higher sodium intake is associated with increased risk of hypertension in humans. Also Morgan [31] stated that reduced intake of high sodium diets reduces the risk of developing hypertension. Therefore the low sodium content indicates that *B. aethiopum* fruit flour is a low-sodium food and is good for individuals with hypertension.

3.2. Anti-nutrient Composition of Borassus aethiopum Fruit Flour

There was no significant difference between the oxalate contents of the *B. aethiopum* fruit flours. However, the freeze dried flour recorded the highest concentration of 6.47 ± 0.52 g/100g while the oven dried had the least concentration of oxalate of 6.25 ± 0.15 g/100g. The oxalate content of the flour from hypocotyl axes of *B. aethiopum* by Ahmed *et al*. (2010) was relatively lower (0.98 ± 0.05 mg/100g) than the values recorded for *B. aethiopum* fruit flours in this work. The difference in value can be attributed to the fact that different thermal conditions were employed in drying the samples and also the botanical source of the fruit. According to Oguchi *et al*. [32], the toxicity level of oxalates is 250 mg/100g but the minimum dose of oxalates that can cause death in adults is 4 to 5 g [33]. To meet the required intake of oxalates, *B. aethiopum* fruit flours should be incorporated into food products at smaller quantities to ensure consumers consume below 3.86 g of the flour at a time. Oxalates are considered as anti-nutrients due to their ability to chelate and bind to essential minerals such as calcium, magnesium, iron and zinc to form insoluble oxalate compounds that are attached to the gastrointestinal tract and reduces the absorption of these nutrients to cause mineral deficiency. It has been recommended that foods that contain high oxalate content should be consumed accompanied with calcium-rich foods such as shellfish and dairy products. According to Morrison and Savage [34], dairy products such as yoghurt contain probiotics which have the ability to digest insoluble oxalates that are deposited in the gut.

Table 2. Anti-nutrient Content of *Borassus aethiopum* Fruit Flour

Anti-nutrient	Freeze Dried Flour	Oven Dried Flour	Solar Dried Flour
Oxalate (g/100g)	6.47 ± 0.52^a	6.25 ± 0.15^a	6.25 ± 0.60^a
Alkaloid (g/100g)	0.59 ± 0.01^a	0.22 ± 0.00^b	0.29 ± 0.01^c
Saponin (g/100g)	55.62 ± 0.00^a	36.10 ± 0.00^b	42.24 ± 0.00^c
Tannin (mg/100g)	23.78 ± 1.52^a	25.28 ± 2.06^{ab}	31.21 ± 0.30^b

Values are means of three replicates. Values in same row with same superscripts are not significantly different (p>0.05).

Results obtained for the phytochemical screening as shown in Table 4 indicated the absence of alkaloids in *B. aethiopum* fruit flour. However, Table 2 shows the alkaloids content of the flours ranged from 0.22 ± 0.00 g/100g for the oven dried flour to 0.59 ± 0.01 g/100g in the freeze dried flour. These values were significantly different from each other at p = 0.05. Although, alkaloids concentrations were the least among all determined anti-nutrients content of *B. aethiopum* fruit flour, its concentrations exceeded the 20 mg/100g reported by Inuwa *et al*. [35]. Alkaloids are considered to be anti-nutrients because of their actions and how they affect the nervous system, disrupting and increasing of electrochemical transmission. At high concentration of alkaloids, it can cause rapid heartbeat, paralysis and in lethal dose can result in death. Alkaloids also cause the disruption of the cell membrane found in the gastrointestinal tract [36,37].

Drying method had a significant effect on the saponin content of *B. aethiopum* fruit flours. Saponins were the highest in value among the determined anti-nutrients. Oven dried flour had the least saponin content (36.10 ± 0.00 g/100g) while freeze dried flour had the highest value of saponins (55.62 ± 0.00 g/100g). All the saponin values recorded are significantly different from each other. It has been reported that saponins have a list of biological activities such as being anti-obesity, antioxidant, antiparasitic, antiulcer and diuretic among others. According to Toyin *et al*. [38], saponin has anti-diarrhoeal effects hence its presence in *B. aethiopum* fruit flour gives it some medicinal benefits. Adversely, saponins at high concentrations above the tolerable limits have negative effects on the absorption of minerals and vitamins, cognitive behaviour, ethanol induced amnesia and the inhibition of active nutrient, such as protein transportation in the body [39,40,41]. Saponins are regarded as anti-nutrients because they decrease the bioavailability of nutrients, decrease enzyme activity and affect protein digestibility by inhibiting some digestive enzymes such as trypsin and chymotrypsin which can lead to protein deficiency [42]. However, the toxicity of saponin to warm blooded mammals like humans depends on the manner at which it is administrated, its source, concentration and composition of the saponin content [43,44]. At tolerable concentrations, saponins possess hypocholesterolemic, antidiarrhoeal, immunostimulatory and anticarcinogenic properties [13,42,44].

Tannin is an astringent, bitter plant polyphenolic compound that either binds or precipitates proteins and various other organic compounds including amino acids and alkaloids [45]. Table 4 showed that tannins were present in the *Borassus aethiopum* fruit flours. There was a significant difference between the tannin contents of the freeze dried and solar dried flours. However, the concentration of tannins in the solar dried flour recorded the highest value of 31.21 ± 0.30 mg/100g while tannins concentration in the freeze dried flour was the least with the value of 23.78 ± 1.52 mg/100g. Although the tannin content of *B. aethiopum* fruit flours were higher than the 3mg/100g lethal dose stated in the work of Inuwa *et al*. [35], the tannin contents of the fruit flours are lower than the 239.76 mg/100g reported by Ahmed *et al*. [46] for the hypocotyl axes flour. According to Ali *et al*. [47] and Gbesso *et al*. [48], the hypocotyl axes have been used as food supplement for some rural Africans in Togo, Cameroon and Nigeria. Medicinally, tannins are used as haemostatic, antidiarrheoal, antiheamorrhoidal and anti-inflammatory agents [13,49]. The high tannin content of *B. aethiopum* fruit flour makes it applicable in the pharmaceutical industry as Amabeoku [50] suggested for plant materials that contained appreciable tannin content.

3.3. Antioxidant Properties

The total phenolic content (TPC) of African palmyra palm fruit flour ranged from 1518.00 mg GAE/100g in solar dried flour to 3896.71 mg GAE/100g in the freeze dried flour samples as shown in Table 3. There was a significant difference between total phenolic content of freeze dried flour samples and solar dried flour samples. However, there was no significant difference between oven dried flour samples and both freeze dried and solar dried flour samples.

Drying at lower temperatures retains more heat labile phytochemicals including phenols than drying at higher temperatures [51]. Hence the higher total phenol content in the freeze dried flour. The finding in this study is contrary to that of Dossou *et al*. [52] who reported that freeze dried ackee fruit arils retained less phenols compared to oven dried ackee fruit arils. This could be attributed to differences in the types of phenols in the various plant materials. The lower total phenol content of solar dried flour could be due to increased oxidative reactions during the drying period [53,54,55] which may lead to the loss of phenols and other antioxidants via reaction with oxidants.

The high total phenol content of the flour could be responsible for its relatively high free DPPH scavenging activity (IC50 from 2.05 – 4.14 mg/mL). Phenols exhibit free radical scavenging activities thus have the ability to minimize the oxidative breakdown of biomolecules [56,57,58]. Scalbert and Williamson [59] recommended consumption of 1000 mg GAE/day of phenols hence consuming 100g of *B. aethiopum* a day could meet this nutritional requirement.

The results for DPPH scavenging activity of the *B. aethiopum* flour are as shown in Figure 1. The IC50 values ranged from 2.05 mg/mL in freeze dried flour to 4.14 mg/mL in solar dried flour (Table 3). The IC50 for African palmyra palm fruit flour in this study were higher than the 1.20 mg/mL reported for the fresh pulp by Amoateng *et al*. (2010). There was no significant difference between the IC50 values for both the freeze dried and oven dried flours. IC50 values indicate antioxidant activity which also depends the total phenols and other antioxidants present. As shown in Figure 2 and Figure 3, strong positive correlations existed between the total phenol contents and IC50 and also between the beta-carotene content and the IC50 values of the flours.

B. aethiopum fruit flour showed a concentration dependent DPPH inhibition activity as shown in Figure 1. The higher the concentration of the flour extract, the higher its free radical scavenging ability.

Table 3. Total Phenolic Content, IC50 and Beta-Carotene Content of *B. aethiopum* Fruit Flour

Parameter	Freeze Dried Flour	Oven Dried Flour	Solar Dried Flour
TPC (mg GAE/100g)	3896.71 ± 774.61^{a}	2550.86 ± 66.40^{ab}	1518.00 ± 509.03^{b}
IC50 (mg/mL)	2.05 ± 0.00^{a}	2.55 ± 0.29^{a}	4.14 ± 0.05^{b}
Beta-Carotene (µg/100g)	3.36 ± 0.00^{a}	4.19 ± 0.79^{a}	2.06 ± 0.89^{a}

Values are means of three replicates. Values in same row with same superscripts are not significantly different (p>0.05).

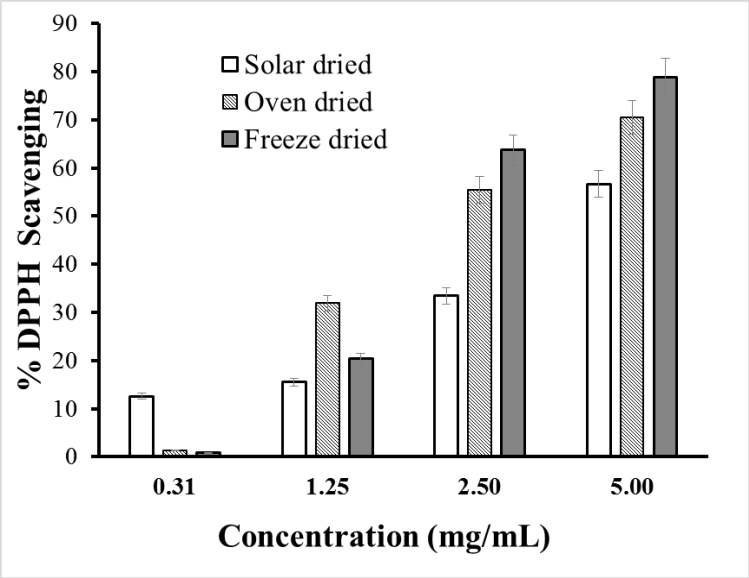

Figure 1. % DPPH scavenging at different flour extract concentrations

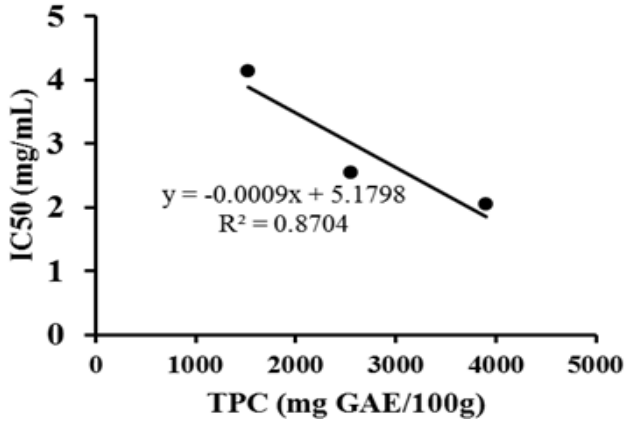

Figure 2. Correlation between total phenol content and IC50 of *B. aethiopum* fruit flour

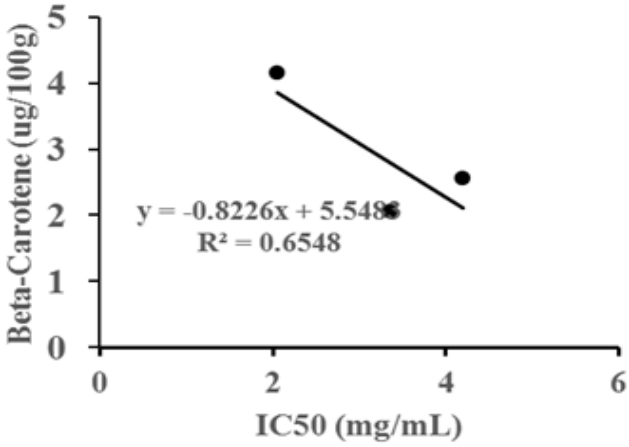

Figure 3. Correlation between beta-carotene content and IC50 of *B. aethiopum* fruit flour

The beta-carotene content of *B. aethiopum* fruit flour ranged from 2.06 µg/100g in the solar dried flour to 4.19 µg/100g in the oven dried flour. However, there difference among the beta-carotene content for all the flours was not significant. The low beta-carotene content could be influenced by the drying conditions. According to Ali *et al*. [60], oven drying at 40°C for 48 h decreased the total carotenoid content of B. aethiopum fruit pulp by 37 – 40 %. Drying is said to decrease the concentration of phytochemicals including antioxidants via browning reactions which are increased in the presence of moisture, heat and air [53,55,61]. The double bonds present in the chemical structure of beta-carotene makes it susceptible to thermal degradation. Beta-carotene is the precursor for vitamin A hence its presence in food materials from plant sources is desirable.

3.4. Phytochemical Screening of *Borassus aethiopum* Fruit Flour

As shown in Table 4, the phytochemical tests indicated the presence of tannins, saponins, glycosides, phenols, sterols, terpenoids, cardiac glycosides and the absence of flavonoids and alkaloids in the methanolic extracts of all the flour samples. However, Table 2 above indicated the flour samples had 0.22 – 0.59 % alkaloids. The negative test response for alkaloids as well as flavonoids could be due to their insolubility in methanol which was used in this study for the flour extract preparation. According to Cowan [62] alkaloids and flavonoids are soluble in ethanol and chloroform respectively hence could only be extracted with these solvents. This was confirmed for alkaloids in the study of Sarkodie *et al*. [5] who reported the presence of alkaloids as well as tannins, phenols,

saponins, and triterpenoids in an ethanolic extract of the fresh B. aethiopum fruit pulp.

Alkaloids, terpenoids, glycosides, saponins, sterols, and tannins have been reported to have anti-diarrhoeal effects due to their anti-enteropooling effects via the inhibition of the release of autocoids and prostaglandins [13,38,63,64,65].

The presence of cardiac glycosides in *B. aethiopum* fruit flours is desired. Cardiac glycosides have been reported to be important in the treatment of congestive of heart failure [66]. Cardiac glycosides have also been found present in sweetsop and soursop fruits which are also commonly consumed tropical fruits [67]. This finding confirmed the research report of Dembitsky *et al*. [68] who associated the use of sweetsop and soursop in the treatment of cardiac-related diseases to their cardiac glycoside composition. Furthermore, the presence of sterols in *B. aethiopum* fruit flours indicates their potential in preventing cancer. Previous findings showed an increased consumption of plant sterols decreased the occurrence of stomach cancer [69].

Table 4. Phytochemical Screening of *Borassus aethiopum* Fruit Flour Extract

Test	Solar Dried Flour	Freeze Dried Flour	Oven dried Flour
Tannins	+	+	+
Saponins	+	+	+
Alkaloids	-	-	-
Glycosides	+	+	+
Phenols	+	+	+
Sterols	+	+	+
Terpenoids	+	+	+
Flavonoids	-	-	-
Cardiac Glycosides	+	+	+

+Present; -Absent.

Phytochemicals including those present in the *B. aethiopum* fruit flour extracts have antioxidant properties, anti-inflammatory and antibacterial effects hence their use in treatment of diseases and bacterial infections. Sarkodie *et al*. [5] reported that fresh ethanolic extract of *B. aethiopum* fruit pulp had anti-inflammatory and anti-microbial effects which were attributed to the phytochemical composition of the extract. Therefore, the phytochemical composition of *B. aethiopum* fruit flour shows its potential medicinal benefits. Although saponins and tannins are anti-nutrients, they also function as antioxidants in the body when consumed at their acceptable levels.

4. Conclusion

African palmyra palm (*Borassus aethiopum)* fruit flour had relatively high levels of potassium, magnesium; considerable amount of sodium, calcium and very low iron content. However, lead, cadmium and zinc concentrations *B. aethiopum* fruit flour were below detection limits.

The flour also had high levels of total phenols as well as considerable free radical scavenging activities. *B. aethiopum* fruit flour however had low levels of beta-carotene and tannins but relatively high levels of saponins and oxalates. The presence of tannins, phenols, sterols, terpenoids, glycosides and saponins gives *B. aethiopum* fruit flour potential medicinal applications as anti-diarrhoeal, anti-microbial and anti-inflammatory agents.

Drying had a significant effect on all the detected minerals except sodium, total phenol content, DPPH scavenging activity, cardiac glycosides, saponin content, tannin content and alkaloid content of *B. aethiopum* fruit flour. However, there was no significant variation among the oxalate and beta-carotene contents of the flour.

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