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Research Article



Phytochemical screening and proximate composition of bitter kola (*Garcinia kola*) and oil palm (*Elaeis guineensis*) syrup

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Abstract

The rising cases of diet-related diseases such as obesity, atherosclerotic cardiovascular disease, inflammatory diseases, diabetes mellitus and cancer have stimulated interest in the use of plant-based food as an alternative to synthetic food. *G. kola* and palm syrup are plant-based food with proven nutritional and health benefits. In the present study, phytochemical and proximate analysis of (*Elaeis guineensis*) syrup, bitter kola (*Garcinia kola*). Preliminary phytochemical screening, proximate analysis and mineral analysis were conducted. The aqueous extracts of palm syrup, cola seeds and their infusion were further subjected to high-performance liquid chromatography analysis to determine the phenolic compounds, organic acids and sugar composition of the extracts. Result of preliminary phytochemical screening, mineral composition, as well as phenolic compound and organic acid indicate that *Garcinia kola*, and palm syrup are nutritionally suitable. Results show that the proximate analysis of oil palm syrup and bitter kola shows high concentrations of crude protein (9.52), ash (4.02), crude fibre (12.23), and fat (15.01) compare to palm syrup ($P < 0.05$). While palm syrup showed high concentrations of Moisture content (34.50) and Carbohydrates (61.67). while The Oil palm syrup contained saponins, glycoside and flavonoids. While Bitter *kola* had a wider range of phytochemicals including the above-mentioned alkaloids, tannins and terpenoids.

Keywords: Palm Syrup, *Garcinia kola*, *Elaeis guineensis*, Antioxidant, Proximate Analysis, Phenolic Compounds, Phytochemicals.

Introduction

Plants are of importance to human existence due to their nutritional and therapeutic benefits, they also provide food and produce the oxygen we breathe. Plants have been extensively explored in pharmacology and nutrition to promote health and manage human diseases. The plant also provides raw materials for our building and in the manufacturing of biofuels, dyes, pesticides and drugs. The use of plants for traditional and medicinal practice has been for decades. The healing ability of plants is due to the abundance of phytochemicals and nutraceuticals in them (Wadood et al., 2013).

In the past, there has been increasing public interest in natural therapies in both developing and developed countries due to poverty and lack of access to modern medicines. The *Elaeis guineensis* is an African oil palm. The stem is stout

and erect. It could attain a height of 30m when fully grown. The plant is monoecious (bears both the male and female flowers). For palm sugar manufacture, fermentation of the sap during collection must be prevented by cooling to a low temperature and/or the addition of preservatives. The juice is filtered and concentrated in pans on open fires and then filled into suitable containers, where after cooling it hardens to a solid golden-brown product with a pleasant taste and smell (Udom et al., 2017; Frank and Beck, 1990). Palm syrup is a natural health product that is gaining global attention and as such been used traditionally as a sweetener (Francisco-Ortega et al., 2013). According to (Ranilla et al., 2008) reduced blood sugar can slow the activities of α -amylase, α -glucosidase, and ACE (angiotensin I-converting enzyme) in vitro. Hence could be considered a cheap and efficacious means of managing type 2 diabetes mellitus.

Garcinia kola (bitter kola) is a dicotyledonous plant belonging to the family of plants called Guttiferaeand. It is found mainly in the tropical forest region of Central and West Africa (Farombi et al., 2002). It is predominant in the rainforest belt of southern Nigeria (Okunji et al., 2002). The crude extract of *Garcinia kola* has been demonstrated to possess antimicrobial activity against a broad spectrum of gram-positive and gram-negative bacteria (Adegboye, et al., 2006). This study focused on identifying the phytochemical and proximate analysis in bitter kola and palm syrup.

Materials and methods

Preparation Of Plant Materials

Oil Palm Sap

The Nigerian Institute for Oil Palm Research in Edo State, Nigeria, provided the oil palm sap.

Garcinia kola

Fresh seeds of *Garcinia kola* (Bitter kola) was bought from New Benin Market, Benin City, Nigeria. It was taken to the University of Benin, Edo state, (Botany Department) for proper identification before analysis. The bitter kola seeds were dried at a constant weight 40°C in a ventilated oven (UNISCOPE SM 9053 Laboratory oven).

Preparation of Palm Sugar

The freshly tapped oil palm sap was collected in a plastic container and suspended in ice block to minimize fermentation and was immediately brought to the laboratory for preparation and analysis. The Palm sap was filtered with a cheese cloth and the filtrate boiled in an open pan at 100°C until it turned viscous brown.

Preparation of Dried seed of *Garcinia kola*

The dried seeds of *G. kola* (70 g) were crushed in a mortar after which the pieces were grinded into powder using a laboratory blender.

Preliminary Phytochemical Screening

Preliminary phytochemical screening was carried out using the methods as described below:

Test for flavonoids

NaOH (1mL10%w/v aqueous solution) was added to 3mL of the extract in different test tubes. A yellow coloration indicates the presence of flavonoids.

Test for alkaloid

Mayer's reagent (2mL) was added to 2mL of plant extract along the side of the tube. A white creamy precipitate appeared, which indicated the presence of alkaloids (Evan, 2010).

Test for amino acids

Ninhydrin solution (2 drops) was added to 2mL of aqueous filtrate. The purple colour developed which indicated the presence of amino acids (Yasuma and Ichikawa, 2011).

Test for carbohydrates

Molish's test

Two drops of an ethanolic solution of α -naphthol were added to 2 mL of plant extract. The mixture was shaken vigorously in a test tube. To this mixture, a few drops of H₂SO₄ was added slowly along the side of the test tube. A violet ring appeared indicating the presence of carbohydrates.

Test for steroids

Two drops of concentrated H₂SO₄ were added to 1mL of the extract. The red colour indicated the presence of steroids.

Phylobatannins

An aliquot (1mL) of the extract was added to 1% Hcl. The presence of red precipitate indicates phylobatannins.

Test for fixed oils and fats

Saponification test

To a 3ml of extract, a few drops of 0.5N alcoholic KOH solution were added. Phenolphthalein was added. The mixture was allowed to boil in a water bath for 2 hours. The formation of foam indicated the presence of oils and fats (Kokate, 2008).

Test for phenolic compounds

Ferric Chloride test

Two drops of 5% ferric chloride solution were added to 50 mg of the plant extract. The formation of dark green colour indicated the presence of phenolic compounds (Mace, 2007).

Test for tannins

The powdered sample (3.0 g) was weighed and boiled in 50 mL of distilled water for 3 min on a hot plate. The mixture was filtered and a portion of the filtrate was diluted with distilled water; 3 drops of 10% FeCl₂ were added. There was a formation of green colour, which indicated the presence of tannins.

Test for phytosterols

Libermann-Burchard's test

The extract (50 mg) was dissolved in 2 mL acetic anhydride. 2 drops of concentrated H₂SO₄ were added slowly along the sides of the test tube. An array of colour changes showed the presence of phytosterols (Finar, 2011).

Test for proteins

The plant extract (100 mg) was dissolved in 10mL of distilled water and filtered through Whatmann No. 1 filter paper and the filtrate was subjected to million's test for proteins.

Millon's Test

Three drops of Millon's reagent were added to 2mL of the filtrate. White precipitate shows the presence of proteins (Rasch and Swift, 2010).

Test for saponins

The plant extract (50mg) was diluted with distilled water and made up to 20 mL. The suspension was shaken in a graduated cylinder for 15 minutes. A 2 cm layer of foam indicated the presence of saponins.

Proximate analysis of palm syrup and *Garcinia kola*

The Association of Official Analytical Chemists (AOAC, 2000) official procedures were used to determine the proximate composition of palm syrup and *G. kola*.

Determination of percentage (%) moisture content

Two grams (2g) of palm syrup and *G. kola* were precisely weighed into a moisture dish (in triplicate), after which the sample was dried at 105°C for 24 hours. The samples were removed from the oven after drying and placed in a desiccator. The process was repeated for 6 hours till the weight became constant.

$$\% \text{ Moisture} = \frac{(Y - Z) \times 100}{X}$$

Where X = show the weight of the sample

Y = show the weight of the dish and sample, respectively before drying,

Z = the weight of the dish and sample after drying.

Determination ash content

Ash content was determined by the incineration of 5.0 g sample placed in a muffle furnace maintained at 500 °C for 2 hrs

Determination of % Crude Protein Content

0.3g of each dried sample was weighed and put in a conical flask. Then 4ml of concentrated sulphuric acid and Kjeldahl tablet was added to it. The sample was subjected to heat till it became colourless. After which reading was taken with a spectrophotometer at 630nm.

$$\text{Nitrogen content} = \frac{\text{Abs} \times \text{dilution volume (digest)}}{\text{Weight of sample} \times \text{colour developer}}$$

Determination of % Crude Fat Content

5g of the dried sample was placed in a thimble lined with a circle of filter paper. The thimble with its contents was placed in a 50 ml beaker and dried in an oven for 6 hours at 105 °C. The beaker was rinsed three times with Hexane and emptied into the Soxhlet extraction flask. The sample contained in the thimble was extracted with hexane for 6 to 8 hours at a condensation rate of 3 - 6 drops per second. At the completion of the extraction, the fat extract was transferred from the extraction flask into a pre-weighed evaporating dish with several rinsing of hexane. The evaporating dish was placed in a fume hood with the fan on, to evaporate the hexane until no odour was detectable. The dish with its contents was dried in an oven for 30 minutes at 105 °C, removed from the oven, cooled in a desiccator to constant weight and weighed. The percentage of crude fat was calculated as follows:

$$\% \text{ Crude fat (ether extract)} = \frac{W2 - W1 \times 100}{S}$$

Where:

W1 = weight of the empty evaporating dish

W2 = weight of evaporating dish + content after drying.

S = sample weight before drying.

Determination of % Crude Fibre Content

The crude fibre was obtained by digesting 5g of dried sample with H₂SO₄ and NaOH and incinerating the residue in a muffle furnace maintained at 500 °C for two hours.

Determination of Titratable Acidity

Titrate acidity of palm syrup and *G. kola* was determined by the titration of 5 mL samples with 0.1 N NaOH, using phenolphthalein as an indicator, to a definite pink end point. Oil palm syrup (20 g) was made up to 100 mL with previously neutralised distilled water. Aliquots (5 mL) were then titrated with 0.1 N NaOH using phenolphthalein as an indicator. Titrate acidity was calculated as % lactic acid using the formula below

$$\text{T.A} = \frac{\text{Titre value} \times \text{Normality} \times 0.9}{\text{Volume of sample used}}$$

Determination of pH

The pH values of palm sugar, and *G. kola* were measured using a Jenway Model 3505 pH meter (Camlab, Over, Cambridge, UK). 10ml of Oil Palm syrup, *G. kola* was read in triplicate. 2.0g of oil Palm syrup, *G. kola* were individually dissolved in 10.0 mL of distilled, deionized water and the pH of the solution was read in triplicate.

Determination of Total Carbohydrate by Proximate Analysis

The percentage carbohydrate content of the sample was determined by adding up the percentages of moisture content, ash, crude protein, fat and subtracting them from 100 (McDonald et al., 1973). The difference in value was taken as the percentage total carbohydrate content of the leaf sample.

Results and discussion

The phytochemical composition of syrup derived from the sap of the African oil palm (*Elaeis guineensis*) tree and *G. kola* seeds (bitter kola) are shown in Table 1.

Table 1. Phytochemical Composition of Palm Syrup, and *Garcinia Kola*

Phytochemical	Palm syrup	<i>Garcinia kola</i>
Alkaloid	-	+
Tannins	-	+
Saponins	+	+
Glycosides	+	+
Flavonoids	++	+
Terpenoids	-	+

Key + =Presence, - = Absence

The Oil palm syrup contained saponins, glycoside and flavonoids. While Bitter *kola* had a wider range of phytochemicals including the above-mentioned alkaloids, tannins and terpenoids.

Table 2. Proximate Composition of Palm syrup and *Garcinia kola*

Composition (%)	Palm syrup	<i>G. kola</i>
Moisture Content	34.50±4.24 ^a	2.54±0.06 ^b
Crude protein	0.94±0.01 ^b	9.52±0.03 ^a
Crude fibre	0.01±0.01 ^a	12.23±0.01 ^a
Crude fat	2.42±0.08 ^b	15.01±0.015 ^a
Ash	0.46±0.2 ^b	4.02±0.07 ^a
Carbohydrate (NFE)	61.67±0.06 ^a	56.68±0.05 ^b

Values are mean +- STD

Table 3. Sugar Composition of Oil Palm Syrup

Name	Amount (mg/100g)	Percentage Composition
HMF	0.00096134	0.0019 ^t
Ribose	0.00000946567	0.0000195 ^t
Xylose	0.000063218	0.000130 ^t
Arabinose	0.0000979034	0.000202 ^t
Rhamnose	0.00000845505	0.000017 ^t
Fructose	8.39166	17.296 ^a
Glucose	36.82062	75.889 ^a
Manitol	0.0000910465	0.000188 ^t
Maltose	0.119798	0.247 ^t
Galactose	0.000161382	0.00033262 ^t
Sorbitol	0.0000545655	0.00011246 ^t
Sucrose	3.18490	6.564 ^b
Total	48.51848	100

KEY: t = Trace Concentration, a = High Concentration, b = Moderate Concentration.

Discussion

In the study, the phytochemical of components revealed the presence of alkaloids, tannins, saponins, terpenoids, glycosides and flavonoids in bitter kola, and flavonoids, saponins and glycosides in palm syrup. The latter agreed with the finding of Oboh and Imafidon (2018) who studied the performance of oil palm syrup in beverage formulations containing alligator citric acid. The potential benefits of flavonoids are well established by several scientific reports. The wide spectrum of pharmacological advantages of flavonoids includes anti-oxidant, anti-inflammatory, anti-microbial, anti-diabetic, anti-ulcer, ant atherosclerotic, anti-thrombotic, anti-neoplastic, anti-antigenic, anti-tumorigenic activities (references). Flavonoids also protect the cardiovascular, hepatic, renal, and gastric systems. The presence of luteolin in both palm syrup and *G. kola* justifies the use of these plants in the management of various degenerative diseases. Luteolin is an active flavonoid with a wide range of biological activities. Its well-known pharmacological activities, such as anti-oxidant, anti-inflammatory, anti-estrogenic, anti-tumorigenic, anti-mutagenic, anti-apoptotic, anti-allergic and anti-diabetic, have been established scientifically (Strikaeo and Tonga, 2015). Luteolin exerts beneficial immunomodulatory effects and thus assists in the treatment of neurodegenerative diseases; it mediates its pharmacological activities through the modification of signaling systems.

The proximate analysis of oil palm syrup and bitter kola shows high concentrations of crude protein (9.52), ash (4.02), crude fibre (12.23), and fat (15.01) compare to palm syrup (P< 0.05). While palm syrup showed high concentrations of Moisture content (34.50) and Carbohydrates (61.67). This finding agrees with the work of Edet et al. (2017). This justifies the nutritional value of these plant products.

Conclusion

Base on the findings of the present study, *Garcinia kola* seeds and oil palm syrup could be considered important functional food. The combination of these plant products is demonstrated to be nutritionally suitable with optimized physicochemical stability. The results further suggest that *G. kola* and palm syrup could be considered in beverage formulation.

Conflict of Interest

The author hereby declares no conflict of interest.

Consent for publication

The author declares that the work has consent for publication.

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