



PHYTOCHEMICAL AND PROXIMATE ANALYSIS OF DRY SEEDS AFRICAN NUTMEG (*Monodora myristica*)

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ABSTRACT

*A phytochemical and proximate analysis of dry seeds of African Nutmeg sold in Watt market in Calabar, Cross River State, Nigeria, was studied between May and June 2023. Ethanolic and aqueous (hot and cold water) extracts of dried seeds of *Monodora myristica* were evaluated for their phytochemical components using standard physicochemical methods. The extracts' phytochemical screening showed alkaloids, glycosides, tannins, saponins, flavonoids, reducing sugars, polyphenols, and phlobatanins. However, anthraquinones, hydroxymethyl anthraquinones, steroids, and terpenoids were absent. In the quantitative phytochemical composition of the dry seeds *Monodora myristica* (test plant); alkaloids, saponins, flavonoids, glycosides, and reducing sugars (compound) contents were found to be $0.6 \pm 0.02\%$, $4.20 \pm 0.1\%$, $6.40 \pm 0.1\%$, $6.43 \pm 0.01\%$, and $6.62 \pm 0.01\%$ respectively. The proximate analysis of the test plant showed moisture ($9.50 \pm 0.1\%$), ash ($4.40 \pm 0.1\%$), protein ($22.87 \pm 0.01\%$), fat ($45.60 \pm 0.1\%$), fibre ($4.85 \pm 0.01\%$) and carbohydrates ($13.04 \pm 0.02\%$). The different solvents used in this study have been reported to have the capacity to extract different bioactive constituents depending on their solubility or polarity in the solvent. The ethanolic solvent in this study had higher solubility for more phytochemicals. Therefore, *Monodora myristica* dry seeds can contribute significantly to man's health management and should be recommended for our daily nutritional needs.*

KEYWORDS

Monodora myristica, proximate analysis, phytochemicals, phytomedicine, organoleptic properties

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INTRODUCTION

Phytochemicals are naturally occurring chemical compounds found in plants. They are responsible for the colour and organoleptic properties of the plant (Hussian *et al.*, 2006; Agbo & Mbotto, 2012). The term phytochemical is generally used to refer to those chemicals that have biological significance but are not established as essential nutrients (Agbo & Mbotto, 2012). Plants are also the source of many drugs (*Pamplona-Roger, 2004*). It is estimated that approximately one-quarter of drugs or active ingredients are obtained from plant substances. Nature has been a source of medicinal agents since time immemorial (Kigigh & Charlie, 2012). *Monodora myristica*, called African or Jamaican nutmeg, is a perennial, edible plant that grows wild in the African forest (Burubai *et al.*, 2007). *Monodora myristica* belongs to the family Annonaceae (Kigigh & Charlie, 2012). Prominent members of this family include fruit trees like custard, apple, and sour chop and plants whose dried fruits and seeds are used as spices and condiments, such as Ethiopian pepper and the flower *Cananga odorata* (Kigigh & Charlie, 2012). It is most prevalent in the southern region of Nigeria (Ekeanyanwu & Etienajirheywe, 2012).

The generic name *Monodora myristica* was derived from the Greek word meaning Single gift, which refers to the usual solitary flower of the plant (Irvine, 2000). It is a wide-spread and attractive small tree with decorative flowers appearing just before the leaves. The tree is native to the West, Central and East African forest belts extending from Sierra Leone to Uganda (Kigigh & Charlie, 2012). The flowers and fruits of all members of this genus (*Monodora*) are very distinctive. The fruit is suspended long on a green stalk with numerous seeds embedded in the whitish-smelling pulp. Almost all tree parts have economic and medical importance (Ekeanyanwu *et al.*, 2010). However, the seeds are the most economically important parts (Burubai *et al.*, 2007). The seed production is seasonal, occurring between April and September (Burubai *et al.*, 2007).

After harvesting, a series of unit operations (fermentation, washing, drying and cracking) are carried out. The Kernel is obtained by cracking the nuts, which is quickly done by heating (Ekeanyanwu *et al.*, 2010). *Monodora myristica* is one of the medical plants in African countries, including Nigeria (Ekeanyanwu & Etienajirheywe, 2012). The documented biological or pharmacology activities are cardiovascular, neuromuscular, antidiarrhoeal, antisickle, anti-inflammatory, stimulants, stomachic, headaches, cough, fever, and skin diseases (Kigigh &

Charlie, 2012). Germicidal or antiseptic agents can be used to control postpartum haemorrhage and as an insect repellent and snuff. Also, when ground into powder, the seed can relieve constipation (Kigigh & Charlie, 2012). It can also be sprinkled on sores, especially those caused by guinea worm (Ekeanyanwu & Etienajirheywe, 2012). It can also treat pneumonia, tuberculosis and upper respiratory tract infections (Cimanga *et al.*, 2002). Several species of plants have been used for centuries as remedies for human disease (Kaushik *et al.*, 2000). Plants contain bioactive components, making them essential in both traditional and modern healthcare systems and have also made Scientists vigorously focus their identification of medical plants in indigenous pharmacopoeia (Adehola & Oladimeji, 2005). Therefore, this study aims to determine the phytochemical composition and approximate composition of African nutmeg (*Monodora myristica*) sold in the Watt market in Calabar, Nigeria, between May and June 2023.

MATERIALS AND METHODS

Sample preparation and extraction

The dry seeds of *Monodora myristica* were bought from Watt Market, Calabar. Botanists identified and authenticated them in the Department of Botany, University of Calabar, Cross River State, Nigeria.

The sample was prepared based on the method described by Adeshina *et al.* (2010). The dry seeds (sample) of *Monodora myristica* were cracked and dried 25°C for three days. The dried sample was then pounded and reduced to a fine paste using a sterilised mortar and pestle.

Aqueous extraction

Aqueous plant material extraction was carried out in cold and hot water. Exactly 100g of the paste sample was weighed and soaked in 500ml of distilled water in a 1L capacity beaker for 24 hours at room temperature, with occasional stirring (Awofisayo *et al.*, 2010). After that, the extract was filtered using Whatmann's No.1 filter paper to obtain a solution free from solids. The exact amount of paste sample (100g) was weighed and soaked in 500ml of hot water in a 1L capacity beaker. After that, the extraction was filtered using Whatmann's No.1 filter paper to obtain a solution. Both filtrates were evaporated to dryness in a 100°C water bath (Agbo & Mboto, 2012) to obtain

a solid mass, which was weighed and appropriately stored in a tightly closed bottle in the refrigerator.

Ethanolic extraction

Extract 100g of paste sample was weighed and soaked in 500 ml of 95% ethanol in a 1L capacity flask for 24 hours. Then, it was filtered using Whatmann's No.1 filter paper and evaporated to dryness in a 100°C-water bath (Adeshina *et al.*, 2010) to remove the ethanol. The dried extract was weighed and stored in a tight bottle in the refrigerator.

Determination of extract yield

The percentage yield of each extraction was determined gravimetrically (Ekeanyanwu & Etienajirheywe, 2012) using the formula below;

$$\text{Extract yield (\%)} = \frac{W_2 - W_1}{W_s} \times \frac{100}{1}$$

Where:

W_1 = Weight of empty beaker

W_2 = Weight of beaker + extract after evaporation of the filtrate

W_s = Weight of the sample

Phytochemical analysis of the extract

The different extracts were analysed both qualitatively and quantitatively for the presence of alkaloids, cyanogenic glycosides, saponins, flavonoids, steroids, tannins, terpenoids, phlobatannins, polyphenols, reducing sugars and anthraquinones, hydroxymethyl anthraquinones using the standard methods described by (AOAC, 2005), (Egwaikhide & Gimba, 2007).

Qualitative analysis

Test for alkaloids

Two (2.0) ml of the extract was diluted in 5 ml of 1% sulphuric acid and warmed in a water bath for five minutes. An equal volume of the mixture was distributed into two different test tubes. Two drops of Dragendorff's reagent were added to the first tube, and the other Mayer's reagent was

added. An orange-red precipitate from either of the tubes indicated the presence of alkaloids (Egwaikhide & Gimba, 2007)

Test for cyanogenic glycosides

Approximately 2.0ml of each extract was diluted in 2.0ml of glacial acetic acid, followed by adding two drops of FeCl₂ solution to the tubes. After that, 2.0ml of 1% Sulphuric acid was carefully added to the mixture to form a layer or ring. A reddish-brown colour at the interface indicates the presence of cyanogenic glycosides (AOAC, 2005)

Test for saponins (Frothing)

Exactly 2.0ml of each extract was diluted in 10ml of distilled water in test tubes and placed in a water bath for five minutes. After that, tubes were removed, vigorously shaken and observed for visible change. The formation of stable froth (Foam or bubble) indicated the presence of saponins (AOAC, 2005)

Test for tannins

2.0ml of each extract was added to 10ml of distilled water contained in test tubes and heated in a water bath for a few minutes, followed by the addition of 1.0ml of 1% of ferric chloride to the mixture and observed for dark-green precipitate (Ekwenge & Okorie, 2010).

Test for flavonoids

Exactly 2.0ml of the extracts was diluted in 10ml of distilled water, after which a few pieces of aluminium metal were inserted into each tube to the level of the mixtures. After that, concentrated hydrochloric acid (HCl) was added to each and allowed to stand for about 10 minutes. Each tube was observed at the end of the reaction for the formation of an orange-red colour (AOAC, 2005).

Test for steroids

2.0ml of acetic anhydride was added to the test tubes containing 2.0ml of extract in 2.0ml of 1% sulphuric acids and observed for a visible change. A change from violet colour to blue or green in tubes indicated the presence of steroids (Gafar *et al.*, 2010).

Test for terpenoids

2.0ml of the extracts was mixed with 2.0ml of chloroform. Therefore, 2.0ml of 1% sulphuric acids was carefully added to form a layer and observed for visible change. A reddish-brown colouration of the interface indicated the presence of terpenoids (Ekeanyanwu & Etienajirheywe, 2012).

Test for phlobatanins

2.0ml of extract was diluted in 5ml of 1% hydrochloric acid in test tubes. The mixture was heated in a water bath and observed for colour change. The formation of red precipitate shows the presence of phlobatanins (AOAC, 2005).

Test for polyphenol

2.0ml of the extracts were treated with 5.0ml of distilled water and heated in a water bath for 30 minutes. After that, 1 ml of ferric chloride was added to the mixture, followed by 1% potassium ferrocyanide solution, and a colour change was observed. The formation of a green-blue colour indicated the presence of polyphenols (AOAC, 2005).

Test for reducing sugar

2.0ml of the extracts were diluted in distilled water and shaken gently for a few minutes. The mixture is then boiled with 5.0ml of Fehling's solution for a few minutes and observed for a colour change. A red precipitate formation indicates the presence of reducing sugar (Gafar *et al.*, 2010).

Test for anthraquinones

2.0ml of the extracts were mixed with 10ml of benzene in test tubes, then 5.0ml of 10% ammonia was added to the mixture and shaken. The formation of a rose-pink colour indicates the presence of anthraquinones (Egwaikhide & Gimba, 2007).

Test for hydroxymethyl anthraquinones

2.0ml of the extracts was diluted in 5.0ml of 5% ammonia solution and observed for change. The formation of a red colour precipitate indicates the presence of hydroxymethyl anthraquinones (AOAC, 2005).

Quantitative analysis

After screening the different extracts for the presence of the various phytochemical substances, the processed sample was subjected to quantitative analysis to determine the amount of each of the phytochemical present in the test sample according to the methods described by AOAC (2005) (Ekeanyanwu & Etienajirheywe, 2012).

Determination of alkaloids

The quantity of alkaloids in the sample was determined gravimetrically following the procedures described by Ekeanyanwu and Etienajirheywe (2012). 5g of the processed sample was weighed into a 250 ml conical flask, followed by adding 100 ml of 10% acetic acid in ethyl alcohol to the sample in the mixture. This was allowed to stand for 4 hours at room temperature with occasional stirring and filtered through Whatmann's No. 1 filter paper. After that, the filtrate was concentrated to one-quarter of its initial Volume in a steam bath and treated with a dropwise addition of concentrated aqueous ammonia until precipitation was obtained. The filtrate was removed using a pre-weighted filter paper by washing the precipitate over the filter paper with 1% ammonium hydroxide solution. The filter paper was then dried in the oven at 100°C for 30 minutes and cooled in a desiccator, and the final Weight of the paper was measured. The amount of alkaloids present in the test sample was then determined and expressed as a percentage of the sample weight and analysed according to the formula described by Ekeanyanwu and Etienajirheywe (2012) as presented below:

$$\% \text{ Alkaloids} = \frac{W_2 - W_1}{W_t} \times \frac{100}{1}$$

Where:

W_1 = Weight of blank filter paper

W_2 = Weight of paper alkaloids precipitation after drying

W_t = Weight of sample analysed

Determination of saponins:

Twenty (20) grams of the defatted sample were diluted in 200 ml of 80% aqueous ethanol and occasionally for 12 hours at 60°C in a water bath. Thereafter, the mixture was filtered through Whatman No.1 filter paper, and the residue was re-extracted in a fresh 200ml of 30% aqueous

ethanol. The ethanol was removed from the combined filtrate by evaporation using a laboratory hot plate to a volume of 4ml and then treated with 20 ml portions of diethyl ether to extract the solution, which was done until a colourless solution was obtained. The resultant solution was treated with 4g of NaCl and HCl to give a pH of 4.0- 5.0. The solution was then treated with 60ml and 30ml portions of n-butanol, respectively and shaken vigorously. After that, both mixtures were combined and washed twice with 10ml of 5% aqueous NaCl and evaporated to dryness. The extract after evaporation was then weighed to give the amount of saponins in the sample, according to the formula below:

$$\% \text{ Saponins} = \frac{W_2 - W_1}{W_t} \times \frac{100}{1}$$

Where:

W_1 = Weight of empty beaker

W_2 = Weight of beaker + Saponins

W_t = Weight of sample analysed

Determination of reducing sugars:

Benedict's quantitative test was used to determine sugars. Precisely 10 ml of the plant extract was diluted in 90 ml of distilled water. This solution was then transferred to a burette and titrated against 20ml of standard Benedict's reagent in a 100ml conical flask placed on an electric hot plate with anti-bump chips inside the conical flask. Titration was continued until the blue colour of the benedict's reagent was changed entirely and the endpoint recorded. The process was repeated three times, and the average Volume of the titre was calculated. The result obtained was then computed against that of a glucose standard and calculated using the formula described by AOAC (2005), as presented below:

$$\frac{18.mg \times \text{Standard Glucose} \times \text{Average Volume of Titre}}{10ml \text{ Benedict's Reagent}}$$

Determination of glycosides:

Ten grams (10g) of the ground dried sample was weighed and dissolved in 200ml water in an in250ml flask and allowed to stand for 2 hours. Complete distillation was then carried out, and 150-170ml of distillate was collected in a 250ml conical flash containing 20ml of 2.5% NaOH. An

antifoaming agent (tannic acid) was added before distillation. Then, 100ml of distillate was measured into a fresh 200ml flask and 8.0ml of 6N ammonium hydroxide, followed by the addition of 5% potassium iodide, mixed and titrated with 0.02N silver nitrate using a micro-burette against a black background. Permanent turbidity indicated the endpoint. The process is repeated, and the average titre volume is calculated. The glycoside content of the sample was then calculated using the formula below, as described by AOAC (2005).

$$\text{Glycosides (mg/g)} = \frac{\text{Titre Volume (ml)} \times 1.08(\text{g})}{\text{Weigh of Sample (g)}}$$

Determination of flavonoids:

This is carried out following the gravimetric method of ethyl lactate precipitation described by Ekeanyanwu and Etienajirheywe (2012). In this method, 5.0g of the grounded sample was weighed, mixed with 100 ml of 2% hydrogen chloride solution, and boiled for 30 minutes. The mixture was allowed to cool and then filtered through the Whatman No.1 filter paper, after which the filtrate was treated with a dropwise addition of ethyl lactate until a full precipitate was obtained. The mixture was filtered using a pre-weighed filter paper, dried in an oven at 100°C for 30 minutes, and cooled in a desiccator. The final Weight of the paper was measured after drying, and the amount of flavonoids in the test sample was determined using the formula below:

$$\% \text{ Flavonoids} = \frac{W_2 - W_1}{W_t} \times \frac{100}{1}$$

Where:

W_1 = Weight of empty filter paper

W_2 = Weight of filter paper + content

W_t = Weight of sample analysed

Proximate analyses

Proximate analyses were carried out using the standard method described by (Abii & Eligalam, 2007). The proximate composition of moisture, ash, crude lipid/fat, crude fibre, crude protein and carbohydrate was determined.

Moisture content:

This was done by weighing 2g of the ground small bit sample into a porcelain crucible and drying it in an oven at 105°C for 24 hours. After that, the crucible and its content were re-weighted, and the difference in Weight gave the percentage moisture content, using the formula below, as described by Abii and Eligalam (2007)

$$\% \text{ Moisture content (\% wet weight)} = \frac{W_b - W_a}{W_s} \times \frac{100}{1}$$

Where:

W_b = Weight of crucible + sample before oven drying

W_a = Weight of crucible + sample after oven drying

W_s = Weight of sample analysed

Ash content: This was carried out by incineration of 2g of the oven-dried sample placed in a muffle furnace at a temperature of 500°C for 8 hours and allowed to cool at room temperature in desiccators. The ash content was calculated and presented in percentage using the formula below:

$$\% \text{ Ash} = \frac{W_2 - W_c}{W_1 - W_c} \times \frac{100}{1}$$

Where:

W₁ = Weight of crucible + Sample after oven drying at 105°C

W₂ = Weight of crucible + Sample after drying at 500°C for 8 hours

W_c = Weight of the crucible

Crude protein content:

The modified micro-Kjeldahl method determined the crude protein content. This method weighed 2.0g of the ground sample and transferred it into the digestion flask, followed by a catalyst mixture (95% anhydrous sodium sulphate and 3.5% copper sulphate). Then, 20ml of concentrated sulphuric acid was added to the Kjeldahl digester at a minimum temperature of 400°C for 30 minutes, which helped to digest the sample. After cooling, the digest was transferred, and 10 ml of distilled water with 15 ml of 45% sodium hydroxide was added. This distillation was continued until the Volume in the recording flask reached 20. The compound formed ammonium borate

complex was diluted to 50ml and titrated with 0.002N HCl to a pink endpoint. The crude protein of the sample was therefore calculated by multiplying the percentage nitrogen content by a conversion factor of 6.25, using the formula below:

$$\% \text{ Crude Protein Content} = \frac{M \times 14mg \times D \times T \times 6.25}{W_s} \times \frac{100}{1}$$

Where:

M = Molarity of acid used (HCl)

14mg = Nitrogen constant per 100ml (i.e W_1 of N_2 in gram = 0.014 and Mg = 14)

D = Volume after dilution (100ml)

T = Titre value of sample

6.25 = Protein conversion factor (1mg of N_2 = 6.25mg of protein)

100 = Percentage conversion factor

Crude lipid/fat content:

2.0g of the test sample was extracted entirely with n-hexane in a soxhlet apparatus at 50°C for 6 hours. An anti-bombing agent was added to the n-hexane in the round bottom before extraction. The percentage crude lipid/fat content of the sample after evaporation of the n-hexane of the flask was calculated using the formula below:

$$\% \text{ Crude Lipid/Fat content} = \frac{W_2 - W_1}{W_s} \times \frac{100}{1}$$

Where:

W_1 = Weight of empty beaker

W_2 = Weight of beaker + content after evaporation

W_s = Weight of sample extract

100 = Percentage conversion factor

Crude fibre content:

Two grams (2.0g) of the ground sample were weighed into a conical flask, and 1.25% sulphuric acid solution was added and heated for about 30 minutes. After that, the mixture was filtered and

washed with distilled water using pH paper until traces of acid were undetected. The acid-free residue was then transferred into another 250ml conical flask, and 1.25% NaOH solution was added and heated again for 30 minutes and, after that, filtered and washed with distilled water until the base was undetected. The sample was transferred into a crucible and dried at 105°C for 24 hours. After that, the crucible was placed in a muffle furnace at 600°C for 8 hours, and its Weight was recorded. The ash was weighed after, and the difference in Weight gave the amount of the crude fibre in the sample, using the formula below:

$$\% \text{ Crude Fibre Content} = \frac{W_1 - W_2}{W_s} \times \frac{100}{1}$$

Where:

W_1 = Weight of crucible + residue after oven drying

W_2 = weight of crucible + ash after drying at 600°C for 8 hours

W_s = Weight of the sample.

RESULTS

Qualitative Composition of Extracts

The results of the phytochemicals screening or composition of the extracts (Table 1) revealed that ethanolic extracts contained glycosides and reducing sugars heavily present (+++), flavonoids, tannins and polyphenols present (++), alkaloids and phlobatanins lightly present (+). At the same time, saponins, anthraquinones, hydroxymethyl anthraquinones, steroids, and terpenoids were absent (-). In hot water extract, polyphenols were heavily present (+++), reducing sugars and tannins were present (++), and alkaloids, glycosides, saponins, and flavonoids were lightly present (+). At the same time, phlobatanins, anthraquinones, hydroxymethyl anthraquinones, steroids and terpenoids were absent (-). In cold water extract, glycosides were present (++), alkaloids, tannins, saponins, polyphenols, and reducing sugars were lightly present (+), while flavonoids, phlobatanins, anthraquinones, hydroxymethyl anthraquinones, steroids, and terpenoids were absent (-).

Quantitative Composition of the Dry Seeds *Monodora myristica*

In the quantitative phytochemical composition of the dry seeds *Monodora myristica* (test plant), alkaloids, saponins, flavonoids, glycosides, and reducing sugars (compound) contents were found

to be $0.6 \pm 0.02\%$, $4.20 \pm 0.1\%$, $6.40 \pm 0.1\%$, $6.43 \pm 0.01\%$, and $6.62 \pm 0.01\%$ respectively (Figure 1).

Proximate composition of the Dry Seeds *Monodora myristica* (test plant)

The proximate composition of the dry seeds *M. myristica* (test plant) revealed moisture ($9.50 \pm 0.1\%$), ash ($4.40 \pm 0.1\%$), protein ($22.87 \pm 0.01\%$), fat ($45.60 \pm 0.1\%$), fibre ($4.85 \pm 0.01\%$), and carbohydrates ($13.04 \pm 0.02\%$). (Figure 2).

TABLE 1: Qualitative Phytochemical Composition of Extracts

S/N	CHEMICAL CONSTITUENTS	ETHANOL EXTRACT	HOT WATER EXTRACT	COLD WATER EXTRACT
1	Alkaloids	+	+	+
2	Glycosides	+++	+	++
3	Tannins	++	++	+
4	Saponins	-	+	+
5	Flavonoids	++	+	-
6	Reducing Sugars	+++	++	+
7	Polyphenols	++	+++	+
8	Phlobatanins	+	-	-
9	Anthraquinones	-	-	-
10	Hydroxymethyl anthraquinones	-	-	-
11	Steroids	-	-	-
12	Terpenoids	-	-	-

Keys: + = Lightly present
++ = Present
+++ = Heavily Present
- = Absent

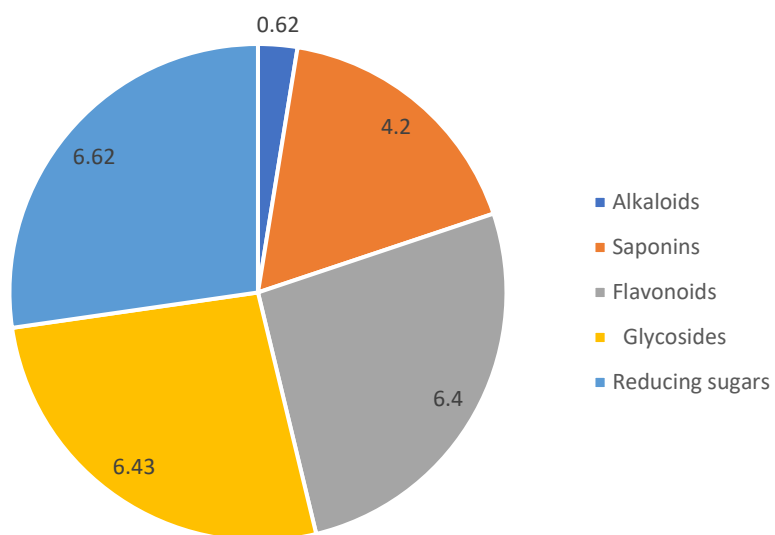


Figure 1: Quantitative analysis of the phytochemical constituents of *Monodora myristica* seeds (%/100g)



Figure 2: Proximate composition of dry seeds *Monodora myristica*

DISCUSSIONS

Phytochemical composition of extracts

All three dry seed extracts contained at least five medicinal plants' twelve biological active constituents. These include alkaloids, glycosides, flavonoids, reducing sugars, tannins, polyphenols, and phlobatanins for ethanol extract. Hot water extract revealed the presence of alkaloids, glycosides, saponins, flavonoids, reducing sugars, tannins and polyphenols, while cold water showed alkaloids, glycosides, saponins, reducing sugars, tannins and polyphenols. These results agreed with that of Udoh *et al.* (2004) and Kigigh and Charlie (2012), who documented saponins, alkaloids, glycosides polyphenols and flavonoids present in the oily extract and aqueous extract, methanol and ethanol extracts of the fresh seeds and dried fruits of *Monodora myristica*. However, none possessed anthraquinones, hydroxymethyl anthraquinones, steroids, and trepenoids. On the contrary, this study shows the presence of phlobotanins in the dry seeds extract, which is absent (Udoh *et al.*, 2004). This could be attributed to cultivation, conditions in terms of soil constituents and environmental factors.

Quantitative Composition of the Dry Seeds *Monodora myristica*

The research results indicate the quantitative phytochemical composition of the dry seeds of *Monodora myristica*, a test plant. The study found that the seeds contain various bioactive compounds, including alkaloids, saponins, flavonoids, glycosides, and reducing sugars. The content of each of these compounds was measured as a percentage of the total weight of the dry seeds.

Alkaloids are nitrogen-containing compounds known for their pharmacological activities, including analgesic and anti-inflammatory effects (Pereira *et al.*, 2023). Saponins are glycosides with soap-like properties and are associated with various biological activities, such as antioxidant and anticancer effects (Timilsena *et al.*, 2023). Flavonoids are plant pigments with antioxidant properties that are believed to promote health (Hassanpour & Doroudi, 2023). Glycosides are compounds that contain a sugar molecule and are often associated with medicinal properties. Reducing sugars are carbohydrates that can act as reducing agents in chemical reactions (Bartnik & Facey, 2024).

Proximate composition of the test plant

The proximate studies of the test plant showed the presence of moisture $9.59 \pm 0.1\%$, ash $4.40 \pm 0.1\%$, protein $22.87 \pm 0.01\%$, fat $45.60 \pm 0.1\%$, fibre $48 \pm 0.01\%$, and carbohydrates $13.04 \pm 0.02\%$. The moisture mean value at $9.50 \pm 0.1\%$ dry weight is comparable with legumes' mean moisture value ranging between 7.0 and 11.0%, 13.15 ± 2.73 for African Nutmeg, and 5.0% for fluted pumpkin seeds reported by Ekeanyanwu *et al.* (2010). Thus, the seeds cannot be stored long without deterioration.

The ash content mean value for African Nutmeg seed in this present study was $4.40 \pm 0.01\%$, comparable with that of (Ekeanyanwu *et al.*, 2010), who reported an ash content value of $3.96 \pm 1.05\%$ in African Nutmeg. It has been documented that ash contents of nuts, seeds and tubers should fall within the range of $1.5 \pm 2.5\%$ to be suitable for animal feeds (Ekeanyanwu & Etienajirheywe, 2012); therefore, the seeds cannot be recommended for animal feed.

The crude protein content means value ($22.87 \pm 0.01\%$) of the test plant in this study is high compared to that reported by Ekeanyanwu *et al.* (2010) for African Nutmeg at $10.13 \pm 1.95\%$, but comparable with those of soybeans, cowpeas, melon, pigeon peas and gourd seeds that range between 23.1 and 33.0% (Ekeanyanwu *et al.*, 2010); chick beans and lima beans had (19.4%) and (19.8%) respectively (Adeyeye & Otokit, 1999). *M. myristica* can be recommended as an alternative source of protein in diet/ protein supplements, especially in underdeveloped countries such as Nigeria, where most of the populace live on starchy food and cereals.

The crude fat with a mean value of $45.60 \pm 0.1\%$ was very high compared to the mean value for African Nutmeg and soybeans at $27.77 \pm 2.59\%$ and 23.5%, respectively, but comparable with the mean value for pumpkin seeds, melon seeds, cashew nut at 49.2%, 47.9 -51.1% and 36.7% respectively (Ekeanyanwu *et al.*, 2010). Fat is vital because it enhances fat-soluble vitamin absorption and contains high calories.

The fibre of $45.60 \pm 0.1\%$ in this study is very high, comparable to legumes mean values that range between 5 and 6% (Ekeanyanwu *et al.*, 2010). It has been documented that a diet low in fibre is undesirable as it could cause constipation and similar diseases. The crude fibre of African Nutmeg is high; hence, it is essential in the diet. Fibre is also crucial in reducing the risk of certain types of cancer and heart diseases (Adeyeye & Otokit, 1999). This study's carbohydrate mean value of

13.04 ± 0.02% is comparable to that of legumes and African Nutmeg at 20-60% and 21.2%, respectively (Ekeanyanwu & Etienajirheywe, 2012). Thus, it is a good source of dietary energy.

Summary and Conclusion

The different solvents used in this study have been reported to have the capacity to extract different bioactive constituents depending on their solubility or polarity in the solvent. The ethanolic solvent in this study had higher solubility for more phytochemicals. The dried seeds of *Monodora myristica* contained alkaloids, glycosides, tannins, saponins, flavonoids, reducing sugars, polyphenols and phlobatanins. However, anthraquinones, hydroxymethyl anthraquinones, steroids, and terpenoids were absent. The dried seeds of *Monodora myristica* contained moisture (9.50 ± 0.1%), ash (4.40 ± 0.1%), protein (22.87 ± 0.01%), fibre (4.85 ± 0.01%), and carbohydrates (13.04 ± 0.02%). Therefore, *Monodora myristica* dry seeds can contribute significantly to man's health management and should be recommended for our daily nutritional needs.

The findings of this research provide valuable information about the chemical composition of *Monodora myristica* seeds, which could be useful for further studies on its pharmacological properties and potential medicinal uses. However, it is important to note that the study provides only a snapshot of the chemical composition of the seeds and further research is needed to fully understand the bioactive properties of these compounds in *Monodora myristica* seeds.

REFERENCES

- Aqeel, U., Aftab, T., Khan, M. M. A., & Naeem, M. (2023). Regulation of essential oil in aromatic plants under changing environment. *Journal of Applied Research on Medicinal and Aromatic Plants*, 32, 100441.
- Abha, S., Swati, V., & Shukla, R. K. (2015). Phytochemical Screening, Proximate Analysis and Antioxidant Activity of *Dracaena reflexa* Lam. Leaves. *Indian Journal of Pharmaceutical Sciences*, 77(5): 640-644.
- Abii, T. A., & Eligalam, A. (2007). Investigation into the chemical composition of the dry Fruit of *Tetrapleura tetraplera* (Ugukirihu). *Journal of Food Technology*, 5, 229-232.
- Abiodun, B. A., Adewale A., & Abiodun. O. O. (2017). Phytochemical and Proximate analysis of some medicinal leaves. *Journal of Science Publishing group*, 6(6), 209-214.
- Adehola, T. T., & Oladimeji, S. A. (2005). Antimicrobial activity of leaf extracts of *Ocimum gratissimum* on Selected diarrhoea-causing bacteria in South-West, Nigeria. *African Journal of Biotechnology*, 4(7), 682-684.
- Adeshina, G. O., Noma, S. T., Onadapo, J. A., & Ehinmidu, J. O. (2010). Preliminary In-vitro antibacterial activities of ethanolic and aqueous extracts of *Rauwolfia caffra*. *International Journal of Pharmacological Research and Development*, pp. 2, 1-8.
- Adeyeye, E. I., & Otokit, M. K. (1999). Proximate composition and some nutritionally valuable minerals of two varieties of *Capsicum annum* (Bell cherry pepper). *Discovery and Innovation*, 11, 75-81.
- Agbo, B. E., & Mbotto, C. I. (2012). Phytochemical and antimicrobial evaluation of selected locally Produced herbal medicines sold in Calabar, Nigeria. *Archives of Applied Science Research*, 4(5), 1974-1990. m
- AOAC. (2005). *Official Methods of Analysis*. Association of Official Analytical Chemists, Washington D.C., pp. 45-46.
- Awofisayo, S. O., Udoh, I. E., & Mbagwu, H. O. C. (2010). Antimicrobial effects of the aqueous and ethanolic extracts of *Tetrapleura tetraptera* pods on the pathogens in nosocomial wound infection. *Journal of Biotechnology and Biotherapeutic*, 1, 18-23.
- Bartnik, M., & Facey, P. (2024). Glycosides. In *Pharmacognosy* (pp. 103-165). Academic Press.
- Bitwell, C., Indra, S. S., Luke, C., & Kakoma, M. K. (2023). A review of modern and conventional extraction techniques and their applications for extracting phytochemicals from plants. *Scientific African*, 19, e01585.
- Brglez-Mojzer, E., Knez Hrnčič, M., Škerget, M., Knez, Ž., & Bren, U. (2016). Polyphenols: Extraction methods, antioxidative action, bioavailability and anticarcinogenic effects. *Molecules*, 21(7), 901.

- Burubai, W., Akor, A. J., Igoni, A. H., & Puyate, Y. T. (2007). Some physical properties of African Nutmeg. *International Journal of Society of Industrial Agrophysics*, 21, 123-126.
- Cheng, H., Wang, J., & Xie, J. (2023). Progress on odor deterioration of aquatic products: Characteristic volatile compounds, analysis methods, and formation mechanisms. *Food Bioscience*, 102666.
- Cimanga, K., Kanbu, K., Toma, L., Apars, S., De Buryne, T., Homan, N., Totte, J., Pietre, L., & Vlietnick, A. J. (2002). Correlation between chemical composition and antimicrobial activity of essential oils of some aromatic medicinal plants growing in the Democratic Republic of Congo. *Journal of Ethnopharmacology*, 79, 213-220.
- Duta-Cornescu, G., Constantin, N., Pojoga, D. M., Nicuta, D., & Simon-Gruita, A. (2023). Somaclonal variation—Advantage or disadvantage in micropropagation of the medicinal plants. *International Journal of Molecular Sciences*, 24(1), 838.
- Egwaikhide, P. A., & Gimba, C. E. (2007). Analysis of the phytochemical content and antimicrobial activities of *Plectranthus glandulosus* Whole Plant. *Middle-East Journal of Scientific Research*, 2(4), 135-138.
- Ekeanyanwu, C., Okigilo, R., Gideon, B., & Nwachukwu, U. (2010). Biochemical characteristics of the African Nutmeg, *Monodora myristica*. *Medwell Journal of Agriculture*, 5(5), 303-308.
- Ekeanyanwu, R. C., & Etienajirheywe, O. F. (2012). In-vitro antihelminthic potentials of *Xylopiya aethiopica* and *Monodora myristica* from Nigeria. *African Journal of Biochemistry Research*, 6(9), 115-120.
- Ekwenge, N., & Okorie, F. (2010). Antimicrobial activities of *Tetrapleura tetraptera* TAUB., Pod Extracts. *International Journal of Pharmacy and Bioscience*, 1, 734-741.
- Gafar, M. K., Hassan, L. G., Dangoggo, S. M., & Hod, A. U. (2010). Amino acid estimation and phytochemical screening of *Indigofera astragalina* leaves. *Journal of Chemical and Pharmaceutical Research*, 2, 277-285.
- Hassanpour, S. H., & Doroudi, A. (2023). Review of the antioxidant potential of flavonoids as a subgroup of polyphenols and partial substitute for synthetic antioxidants. *Avicenna Journal of Phytomedicine*, 13(4), 354.
- Hussian, I., Saleem, M., Igbal, Y., & Khali, S. J. (2006). Comparison of vitamin C contents in commercial tea brands and fresh tea leaves. *Journal of Chemical Society Pakistan*, 28(5), 421-425.
- Imafiodon, K. E., Abu, O. D., Obayuwana, H. O., & Okuopu, E. D. (2018). Phytochemical, proximate, and metal content analysis of *Citrullus lanatus* (watermelon) seed. *FUDMA Journal of Sciences*, 2(2).
- Irvine, F. R. (2000). *Woody plants of Ghana with particular reference to their uses*. Oxford University Press, London, 13–23.

- Kaushik, P., Upadhyay, V., & Singh, P. (2000). Antibacterial potential of Bryophytes. *The Botanica*, 50, 132–136.
- Kigigh, L. T., & Charlie, E. J. (2012). Effects *Monodora myristica* of pepper-soup cooking on antibacterial activity of. *Continental Journal of Food Science and Technology*, 6(1), 8-11.
- Ma, M., Li, X., Liang, H., Jiang, H., & Cui, H. (2023). Chemical constituents variation of seed oil of the Chinese tallow tree (*Sapium sebiferum* (L.) Roxb) at different harvesting time. *Industrial Crops and Products*, 202, 117061.
- Nwachujor, I. U., Ayanda, I. S., & Lawal, I. O. (2019). Phytochemical analysis, proximate composition and mineral contents of the seed of *Annona muricata*. *Journal of Medical Pharmaceutical and Allied Sciences*, 8(12) 2096-2103.
- Pamplona-Roger, G. D. (2004). *Encyclopaedia of foods and their healing power*. Review and Herald Publishing, Maryland, USA, 412.
- Pereira, A. G., Cassani, L., Garcia-Oliveira, P., Otero, P., Mansoor, S., Echave, J., ... & Prieto, M. A. (2023). Plant Alkaloids: Production, Extraction, and Potential Therapeutic Properties. In *Natural Secondary Metabolites: From Nature, Through Science, to Industry* (pp. 157-200). Cham: Springer International Publishing.
- Timilsena, Y. P., Phosanam, A., & Stockmann, R. (2023). Perspectives on saponins: Food functionality and applications. *International Journal of Molecular Sciences*, 24(17), 13538.
- Udoh, U. E., Ezugwu, C. O., & Agali, I. U. (2004). Antimicrobial activity of *Monodora myristica* seed oil. *Journal of Pharmacy and Allied Science*, 2(2).
- Zhang, L., Guan, Q., Jiang, J., & Khan, M. S. (2023). Tannin complexation with metal ions and its implication on human health, environment and industry: An overview. *International Journal of Biological Macromolecules*, 253, 127485.

APPENDICES

Appendix 1: Quantitative composition of the dry seeds *Monodora myristica* (test plant) Chemical Constituents (g/100g)

Alkaloids	Saponins	Flavonoids	Glycosides	Reducing sugars
0.62 ± 0.02	4.20 ± 0.1	6.40 ± 0.1	6.43 ± 0.01	6.62 ± 0.01

Each value represents the Mean of three (3) determinations ± standard deviation ($X \pm SD$).

Appendix 2: Proximate composition of dry seeds *Monodora myristica* (test plant) Nutrient compositions

Moisture	Ash	Protein	Fat	Fibre	CHO
9.50 ± 0.1%	4.40 ± 0.1%	22.87 ± 0.01%	45.60 ± 0.01%	4.85 ± 0.01%	13.04 ± 0.02%

Each value represents the mean of three (3) determinations ± Standard deviation ($X \pm SD$)

CHO = Carbohydrates.