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# Physicochemical Composition and Oil Characterization of *Dioclea reflexa* and *Monodora myristica*Seeds

**Akinyede, A.I.[1], Fagbemi, T.N.[1]*, Osundahunsi, O.F.[1] and Aluko, R.E.[2]**

[1]Department of Food Science and Technology, Federal University of Technology, P.M.B. 704 Akure, Nigeria
[2]Department of Human Nutritional Sciences University of Manitoba, Winnipeg, MB, R3T 2N2, Canada
*Corresponding author: tnfagbemi@futa.edu.ng

## ABSTRACT

*Potentials of oilseeds for commercial application is based on the knowledge of their nutritional composition and oil characterization. The proximate composition and oil characteristics of two selected seeds were determined using standard methods. The whole seeds were sorted, de-shelled, dried, milled, sieved and defatted using n – hexane. The proximate composition of Marble vine (Dioclea reflexa) and Calabash nutmeg (Monodora myristica) showed that the seeds contained total ash 2.51% and 3.51%; moisture, 7.2% and 9.4 %; crude fibre, 2.0% and 12.06 %; crude fat, 7.46% and 38.5%; crude protein 13.00% and 23.21%; carbohydrate 28.02% and 54.31% and energy value 370.02 and 510.58 Kcal respectively. The seeds contained 965.0mg/kg- 1020 mg/kg (potassium); 768.7mg/kg - 963.1 mg/kg (Calcium) and 639.1mg/kg - 650.90 mg/kg (Phosphorus). The oils were found to have: Saponification value: 196.0 – 212.0 mgKOH/g; peroxide value: 5.85 – 6.30 mEq OH/g; acid value: 5.60 – 6.30 mg KOH/g; iodine value: 105 – 112.6 g /100g (Wijs); specific gravity: 0.94 – 0.97; refractive index: 1.42 – 1.45; free fatty acid: 2.80 - 3.15 %. The fatty acid profile showed that the seed oils were high in unsaturated fatty acids (55.05 – 83.83%) of which linoleic (28.95 – 44.51%), oleic (22.79 – 37.71%), linolenic (1.16 – 2.44%) acids were dominant. Brassicasterol was not detected in both seed oils. The seed oils are good sources of essential fatty acids and may have potentials for both domestic and industrial applications.*

**Key words**: *Dioclea reflexa, Monodora myristica*, fatty acid, sterols, marble vine, calabash nutmeg, physicochemical

## INTRODUCTION

Oils and proteins derived from plant seed sources remain important for their nutritional, industrial and pharmaceutical applications (Chouhan *et al.,* 2011). Fats and oils are non-volatile substances insoluble in water but soluble in organic solvents which belong to a larger group of naturally-occurring compounds called lipids. Lipids serve as a convenient means of rapid heat transfer that have found increasing use in commercial frying operations (Andrew *et al.,* 2012). Fats and oils, and their several lipid components can extensively be used in the food, cosmetics, pharmaceuticals and biodiesel paints (Aremu *et al.,* 2015). Nut and seed oils are receiving growing interest due to their high concentration of bioactive lipid components which have shown various health benefits.

*Monodora myristica*, commonly known as 'calabash nutmeg', 'Ariwo'; 'Lakoshe' in Yoruba, 'Ebonoyeba' in Edo, and 'Ehuru' in Igbo is a tropical shrub of the Annonaceae or custard apple family of flowering plants (Akinwunmi and Oyedapo, 2014; Okonkwo and Ogu 2014).The Calabash nutmeg tree grows naturally in evergreen forests from Liberia to Nigeria and Cameroon, Angola, Uganda and west Kenya and almost every part of the tree have economic importance and medicinal values (Weiss, 2002; Ekeanyanwu *et al.,* 2010; Akinwunmi and Oyedapo, 2014). The *Monodora myristica* tree can reach a height of 35 m and 2 m in diameter at breast height (Osuagwu and Onwuegbuchulam, 2015). It has a clear trunk and branches horizontally. The fruits are collected from wild trees and the seeds are dried and sold whole or ground to be used in stews, soups, cakes and desserts (Weiss, 2002; Ekeanyanwu *et al.,* 2010 and Celtnet Recipes, 2011). The seed is reported to be used in the treatment of hermorrhoids, stomach ache and febrile pains (Ekeanyanwu *et al.,* 2010).

*Dioclea reflexa*, called marble vine or sea purse sea bean in English, 'agbarin', 'capenter', and 'Ukpo' (local names) is a legume belonging to the sub-family papillionoideae (Akinyede *et al.,* 2005; Ajayi and Adefioye, 2012; Akinyede *et al.,* 2016). The plant is a climber and it is usually propagated using 10-12 seeds. The desired spice element is the seed and the powdered cotyledon is used in preparing a special soup which is valued because of its

delicious aroma and sharp taste that increases appetite (Akinyede *et al.,* 2005; Oladosu *et al.,* 2010; and Akinyede *et al.,* 2016). There have been folk uses of the seeds as a remedy for treating rheumatism, itching and other infections caused by pathogens (Faleye, 2012). There are variations on the results of the nutritional composition of *Dioclea reflexa and Monodora myristica* seeds (Akinyede *et al.,* 2005; Yusuf and Lasisi, 2006; Faleye, 2010; Ekeanyanwu *et al.,* 2010; Oyeleke *et al.,* 2012; Ajayi and Adefioye, 2012; Okonkwo and Ogu, 2014) which may be as a result of geographical location, climate, genetic factor, cultural conditions, environment and nutritional status of the plant but there is dearth of information available on the characteristics of the seed oils. This study therefore is aimed at determining the chemical composition, physicochemical properties, sterols and fatty acid profile of *Dioclea reflexa* and *Monodora myristica* seeds and oils with a view to contribute to the existing knowledge of the potentials of the seeds.

## MATERIALS AND METHODS

### *Source of Materials*

The raw materials used in this work are the seeds of Marble vine (*Dioclea reflexa*) and Calabash nutmeg (*Monodora myristica*). The seeds were obtained from market depots at Ogbese, Ondo state and were identified at Forest Research Institute of Nigeria, Ibadan, Oyo state, Nigeria. The seeds were cleaned and milled into seed flours A portion of the seed flour was defatted continuously for 8 hours using n-hexane as solvent, thereafter dried and ground. The solvent was removed and the crude oil extract was made free of water by filtering through the anhydrous sodium sulphate. All determinations were run in triplicates. All chemicals used were of analytical grade and were procured from Fisher Scientific (Oakville, ON, Canada).

### Analyses

### *Determination of Proximate composition of Dioclea reflexa and Monodora myristica flours*

The contents of moisture, crude fat, crude fibre and total ash in the samples were assayed by the standard methods of AOAC (2005). The crude protein was determined using Lowry's method Lowry *et al.,* (1951) as modified by Markwell *et al.,* (1978). Carbohydrate content was determined by difference.

### *Determination of Mineral elements of Dioclea reflexa and Monodora myristica flours*

The mineral contents of the samples under study was carried out using Atomic absorption spectrophotometry while the phosphorus was determined by using the phospho vanado molybdate method, Vanodo molybdate reagent was used at 470 nm AOAC (2005).

### *Determination of Physicochemical properties of the seed oils*

The specific gravity (using a 10ml pycnometer 25$^{o}$C), refractive index (using Abbey refractometer Model TM 1600, Gibertini, Italy), iodine value, peroxide value, saponification number unsaponifiable matter, free fatty acid and acid value were determined as described by AOAC (2005).

### *Fatty acid analysis*

Fatty acid methyl esters (FAME) were prepared after alkaline hydrolysis, followed by methylation in methanol plus BF3 (14% boron trifluoride). Analysis of FAME was carried out by Gas Liquid Chromatography (GLC) using a Perkin Elmer Autosystem XL Gas Chromatograph. Column: SGE BPX70 (Phenomenex Cat No. CGO-5512), 30m length, 0.25mm interior diameter, 0.25µm film thickness. Carrier gas: Helium at 20Psi (1.85ml/min) Detector: Flame ionization (FID) at 265$^{o}$C. Hydrogen flow (45ml.min) and air (450ml/min), Injector: Split, packed at 265$^{o}$C, Split flow ratio 0:1 (76.9ml/min). Temperature Program: 60$^{o}$C for 20min, increase to 180$^{o}$C at 10$^{o}$C/min, increase to 235$^{o}$C at 4$^{o}$C/min, Total runtime was 27.7min (Ali *et al.,* 2010).

### *Sterol composition analysis*

The method reported by Dutta (1997) was used in the analysis of sterols in the seed oils. Each oil sample (1 g) was weighed into 50 ml screw topped glass test tube. Ten milliliter (10 ml) of 3% alcoholic KOH solution was added, the tube was capped and vortex to mix and allowed to stand overnight in the dark to saponify. Distilled water (10 ml) was added to the tube, capped and vortex to mix completely. Ten milliliter (10 ml) of methylene chloride was added, recapped, vortex for at least 1 minute to extract. The tubes were allowed to stand at room temperature until layers were formed; the top layer was removed by vacuum suction. Distilled water 10 ml was added to the test tube to wash the solvent layer, recapped and vortex until it was thoroughly mixed and centrifuged at 1000 rpm for 10 min. The top layer was later removed by suction. The solvent (1ml) was added to a clean tube, dried under nitrogen stream and the residue obtained was dissolved in 0.9 ml of n-hexane. Cholestane internal standard solution (100 ml) was added to the tube and transferred to GC vial; pyridine (100 ml) and Regisil® (100 ml) were added to the vial, capped tightly and vortex mix. The vial was heated for one hour using a heating block at 50$^{o}$C and later injected into the GC with standard

solution (Plant Sterol Standard Cat No. 1119, Matreya). GC Conditions:

Column: RTX-5, 30 m length, 0.32 mm interior diameter, 0.25 µm film thickness (50 % metylpolysiloxane cross bonded to 50 % phenyl polysiloxane). Temperature Program: 100$^{o}$C held for 0.2min. Increased at 10$^{o}$C per min to a final temperature of 290$^{o}$C held for 8min. Injector Temperature: 230$^{o}$C Detection: by flame ionization at 290$^{o}$C Carrier Gas: Helium at 2.1 ml/min Flame gases: Hydrogen at 45 ml/min and air at 450 ml/min. GC used: Perkin Elmer Autosystem XL.

### *Statistical Analysis*

Determinations were carried out in triplicates; errors were recorded as standard deviation from the mean. Data were subjected to analysis of variance using SPSS 17 computer programme while means were separated using New Duncan Multiple Range Test (NDMRT). Significance was accepted at 5 % level of probability. Statistically significant differences between groups were compared using analysis of independent of t – test at probability level of 95 %.

## RESULTS AND DISCUSSION

### Proximate composition of *Dioclea reflexa* and *Monodora myristica* flours

The proximate composition of full fat and defatted *Dioclea reflexa* and *Monodora myristica* seed flours are shown in Table 1. The moisture content of the seed flours ranged between 7.20 and 9.40 % and these values are comparable to the 7.68 – 8.41 % reported for pea, chickpea and lentil (Ladjal and Chibane, 2015); 9.14 – 9.49 % reported for guinea peanut and African oak seed flours (Ogunlade *et al.,* 2011). However, the values are low when compared with 10.83 - 11.2 % reported for *Monodora myristica* by Okonkwo and Ogu (2014) and higher than 6.52 and 6.73 % obtained for brown and golden flax, 6.02 and 6.40 for brown and white perilla (Sargi *et al.,* 2013). The low moisture content of these seeds may indicate that they can be stored for long time without spoilage except for oxidative rancidity in food. *Dioclea reflexa*and *Monodora myristica* seed flours recorded total ash content between 2.15 – 3.15%. The result obtained from these seed flours are comparable to 2.13% observed in the pra seeds (Anchan, 2010); 2.45 – 3.24% reported for pea, chickpea and lentil (Ladjal and Chibane, 2015). Sample with high ash contents is expected to have high concentration of various mineral elements, which are expected to speed up metabolic processes, improve growth and development (Elinge *et al.,* 2012). Crude fat content ranged from 16.23 – 38.54 % and are comparable to 21.69 – 42.27 % reported in chia, golden and brown flax seeds, white and brown perilla seeds (Sargi *et al.,* 2013); 15.29 – 16.35 % in guinea peanut and African oak seed flours (Ogunlade *et al.,* 2011) and 27.48% in pumpkin seed (Elinge *et al.,* 2012).

**Table 1:** Proximate composition of full fat and defatted *Dioclea reflexa and Monodora myristica* seed flours (g/100g dry weight basis)

| Parameters | *Dioclea reflexa* | | *Monodora myristica* | |
|---|---|---|---|---|
| | DR | DDR | MM | DMM |
| Moisture | $9.40^{a} \pm 0.20$ | $8.90^{b} \pm 0.1$ | $7.80^{c} \pm 0.1$ | $7.20^{d} \pm 0.1$ |
| Crude Protein | $15.11^{c} \pm 0.1$ | $21.41^{b} \pm 0.1$ | $13.00^{d} \pm 0.10$ | $23.21^{a} \pm 0.1$ |
| Crude Fat | $16.21^{b} \pm 0.1$ | $7.46^{d} \pm 0.1$ | $38.5^{a} \pm 0.1$ | $8.99^{c} \pm 0.1$ |
| Total Ash | $3.20^{b} \pm 0.46$ | $3.51^{a} \pm 0.20$ | $2.15^{d} \pm 0.10$ | $2.91^{c} \pm 0.10$ |
| Crude Fibre | $2.00^{d} \pm 0.2$ | $4.41^{c} \pm 0.3$ | $10.53^{b} \pm 0.2$ | $12.06^{a} \pm 0.3$ |
| Carbohydrate[k] | $54.08^{a} \pm 0.4$ | $54.31^{a} \pm 1.02$ | $28.02^{c} \pm 0.2$ | $45.63^{b} \pm 0.2$ |
| Energy Kcal | $422.13^{b} \pm 0.20$ | $370.02^{c} \pm 0.50$ | $510.58^{a} \pm 0.153$ | $356.27^{d} \pm 0.5$ |

Values with different superscripts on the same row are significantly different ($P \leq 0.05$). k – by difference. Errors are standard deviation from the mean, n=3, Cal. Metabolisable energy Kcal (Protein × 4 + fat × 9 + carbohydrate × 4)DR- *Dioclea reflexa* (fullfat); MM- *Monodora myristica* (fullfat); DDR – Defatted *Dioclea reflexa*; DMM- Defatted *Monodora myristica*

**Table 2:** Mineral composition of full fat and defatted *Dioclea reflexa and Monodora myristica* seed flours (mg/kg).

| Mineral element | DR | DDR | MM | DMM |
|---|---|---|---|---|
| Fe | 135.00 [b] ± 0.2 | 135.02 [b] ± 0.23 | 137.53 [a] ± 0.3 | 135.60 [b] ± 0.30 |
| Mg | 572.30 [b] ± 0.30 | 602.60 [a] ± 0.30 | 395.40 [c] ± 0.40 | 389.30 [d] ± 0.30 |
| Pb | 2.54 [c] ± 0.4 | 2.53 [c] ± 0.3 | 6.72 [a] ± 0.2 | 5.59 [b] ± 0.3 |
| Zn | 12.00 [a] ± 0.10 | 9.00 [c] ± 0.10 | 11.10 [b] ± 0.10 | 12.00 [a] ± 0.10 |
| Cu | 6.10 [a] ± 0.2 | 2.20 [b] ± 0.20 | 1.30 [c] ± 0.20 | 6.30 [a] ± 0.20 |
| Ni | 75.30 [c] ± 0.30 | 75.50 [c] ± 0.20 | 125.80 [a] ± 0.20 | 100.90 [b] ± 0.252 |
| P | 650.00 [a] ± 1.00 | 639.10 [c] ± 0.55 | 650.90 [a] ± 0.20 | 642.50 [b] ± 0.20 |
| Ca | 937.10 [c] ± 0.10 | 963.10 [a] ± 0.10 | 768.70 [d] ± 0.20 | 960.80 [b] ± 0.20 |
| K | 983.70 [c] ± 0.20 | 965.10 [d] ± 0.10 | 1020.00 [a] ± 0.50 | 998.70 [b] ± 0.10 |
| Na | 129.80 [c] ± 0.20 | 132.50 [b] ± 0.50 | 109.70 [d] ± 0.20 | 135.10 [a] ± 0.10 |
| Mn | 25.40 [d] ± 0.20 | 32.20 [c] ± 0.6 | 54.00 [b] ± 0.50 | 65.40 [a] ± 0.20 |
| Cd | Nd | Nd | Nd | Nd |
| Se | Nd | Nd | Nd | Nd |
| Na/K | 0.13 [a] ±0.2 | 0.14 [a] ±0.1 | 0.11 [a] ±0.1 | 0.14 [a] ±0.01 |
| Ca/P | 1.44 [b] ±0.1 | 1.51 [a] ±0.1 | 1.18 [c] ±0.1 | 1.18 [c] ±0.1 |
| Ca/Mg | 1.64 [b] ±0.1 | 1.60 [c] ±0.2 | 1.94 [a] ±0.1 | 1.05 [d] ±0.2 |

Values with different superscripts on the same row are significantly different ($P \leq 0.05$). Nd – not detected. Errors are standard deviation from the mean, n=3, DR- *Dioclea reflexa*; MM- *Monodora myristica*; DDR – Defatted *Dioclea reflexa* and DMM- Defatted *Monodora myristica*.

The crude protein content of the seed flours ranged between 13.1 and 15.12% for the fullfat and 21.21 - 23.21% for the defatted samples. The crude protein content of the seeds of *Dioclea reflexa* and *Monodora myristica* are comparable to those reported for guinea peanut (10.38%), African oak (16.52%) and almond 21% (Anchan, 2010; Ogunlade *et al.,* 2011).The protein of the seed of *Dioclea reflexa* was lower than the previous work reported by (Akinyede *et al.,* 2005; Yusuf and Lasisi 2006; Ajayi and Adefioye 2012). Also *Monodora myristica*, there were variations in the values reported by Ekeanyanwu *et al.* (2010); Okonkwo and Ogu, (2014); Enabulele *et al.* (2014). Defatting increased the crude protein concentration of the seed flours. The relatively high crude protein values reported for these seeds suggest that they may find use in food formulations; however, the availability and quality of the amino acids present in flour has to be determined.

**Mineral elements of *Dioclea reflexa* and *Monodora myristica* seed flours**

Results obtained for the mineral element determined showed that the seeds are rich in potassium (983.7 – 1020 mg/kg); calcium (768.70 – 937 mg/kg) and magnesium (395.40 – 572.30 mg/kg) (Table 2). Potassium was the most abundant mineral element in the seed flours and these results are in agreement with the observation of Olaofe and Sanni, (1988) that potassium is the most predominant element in Nigerian agricultural products. Similar observations have been reported for African nutmeg (Ekeanyanwu *et al.,* 2010); water lily (Musa *et al.,* 2012) and pumpkin (Elinge, 2012). The high concentration of potassium will help to regulate acid-base balance and normal metabolism. The toxic element detected was lead (2.53 – 6.72 mg/kg) while mercury, Cadmium and Arsenic were not detected. Sodium/Potassium ratio (Na/K) of 0.11 and 0.13 is less than the recommended 1.0 (Atasie *et al.,* 2009; Musa *et al.,* 2012) and thus suggesting the seeds may probably reduce high blood pressure and may not negatively affect the consumer. Ca/P ratios are indices of bone formation and the recommended ratio of Ca/P is 1.0. The Ca/P weight ratio ranged from 1.18 – 1.44 which is higher than the recommended ratio, hence the seeds may be considered to be of nutritional benefit, particularly for children and the aged who need higher intakes of calcium and phosphorus for bone formation and maintenance. The Ca/P ratio >1is considered 'good' and 'poor' if the ratio is < 0.5 while Ca/P ratio >2.0 helps to increase the absorption of calcium in the small intestine. The Ca/Mg serve as representation of homeostatic balances and predictive of future metabolic dysfunctions (ARL, 2012).The Ca/Mg weight ratio ranged from 1.64 – 1.94 and lower than the recommended 2.2. The ratio of Cd: Zn is nil hence the seed flours may be free from cadmium toxicity.

**Physicochemical Properties *Dioclea reflexa* and *Monodora myristica* Seed Oils**

Physical examination of *Dioclea reflexa* and *Monodora myristica* seed oils shown in (Table 3) revealed that the oils have golden brown appearance. The specific gravity of the oils (at 25$^{o}$C) ranged between 0.94 – 0.97. The values are in agreement with those reported for crude sunflower oil (0.934); cashew nut seed oil (0.96) (Ren, 2010; Evbuowman *et al.,* 2013) and higher than the values reported for groundnut seed oil 0.91; melon seed oil 0.92 and rapeseed oil 0.91 (Motojesi *et al.,*2011; Musa *et al.,* 2012; Olaofe *et al.,* 2012). The high specific gravity could be as an indication of high molecular weight and unsaturated fatty acid (Otunola *et al.,* 2009). The refractive index of the seed oils ranged between 1.42 – 1.45 and comparable to values reported for some conventional oil including cashew nut seed oil, 1.42; pumpkin seed oil, 1.46; *Citrulluslunatus* seed, 1.45; for two types of palm kernel oil and almond seed oil, 1.46 (Muibat *et al.,* 2011; Motojesi *et al.,* 2011; Akinyeye *et al.,* 2011; Ogunsuyi and Daramola, 2013; Aremu and Akinwumi, 2014). It is however lower than the values reported for pumpkin seed oil1.47 (Nwabanne, 2012; Aremu and Akinwumi, 2014). Refrative index is the ratio of the speed of light in a vacuum to that in the oil which is related to the degree of saturation and the ratio of double bonds and may provide information on the oxidative damage (Aremu *et al.,* 2015). There is no significant difference in the values of refractive index of the oils (Table 3) though a slight difference in the value occur which may suggest difference in the degree of flow or thickness of the oils at room temperature (Nkafamiya *et al.,*2010).

The saponification value of the seed oils ranged between 196 and 212 mg KOH/g. The values compares with those reported for cotton seed (185 – 198 mg KOH/g); melon seed oil (197 - 220 mg KOH/g) (Akinyeye *et al.,* 2011; Olaofe *et al.,* 2012; Edidiong and Ubong, 2013; Duduyemi *et al.,* 2013 and Egbebi, 2014). It is however lower than the values reported for some vegetable oils such as groundnut seed oil (227mg KOH/g) and *Jatropha curcas* seed oil (240 mg KOH/g), (Belewu *et al.,*2010; Amos-Tauta and Onigbinde, 2013). The saponification value of oil gives an indication of application or usefulness of the oil. Oil with high saponification value is more suitable for application such as the manufacture of soaps while high saponification in refined oil is an indication of contamination (Pomeranz and Meloan, 2000; Nkafamiya *et al.,* 2010). The saponification values of oil samples in the study may be an indication that the oils may contain high proportion of higher molecular weight fatty acids which may make it find application in food systems for both domestic and industrial applications.

The unsaponifiable matter of *Dioclea reflexa* and *Monodora myristica* seed oil ranged from 0.56 to 0.70. The unsaponifiable matter indicates the level of contamination of oil with high alcohol and hydrocarbons such as sterols, fatty alcohol and vitamins which have basic biological functions such as hormone synthesis. The low unsaponifiable matter of the seed oils is an indication of its safety and low levels of minor constituents. Hence, the oil can be used as component of drugs and as antioxidant agents. The maximum allowed is 2% (Pomeranz and Meloan, 2000). The acid value of *Dioclea reflexa*and *Monodora myristica* seed oils ranged between 5.60 – 6.30 mg KOH/g. Acid value is used as a check for freshness and edibility of oils. The acid value of the seed oils is within the range reported for kernel oil 3 - 7 mg KOH/g. The values are lower than the range of values 6.05 – 32.83 mg KOH/g reported for different types of palm oil (Akinyeye *et al.,*2011). The acid values of the seed oils are higher than the range of 3.13 – 4.22 mg KOH/g reported for some varieties of melon seeds oils (Abiodun and Adeleke, 2010). The acid value of *M. myristica* seed oil is lower than *D. reflexa* seed oil which may signify lower degree of susceptibility of the oil to rancidity and a higher degree of stability.

The iodine value of *Dioclea reflexa* and *Monodora myristica* seed oil ranged between 105.1 and112.60 g/100g (Wijs). These values compares favourably with those reported for groundnut seed oil (110 g/100g (Wijs), sesame seed oil (106 g/100g (Wijs)), pumpkin seed oil (101 g/100g (Wijs)) (Ayoola and Adeyeye, 2010; Bello *et al.,* 2012; Ogbonna and Ukaan, 2013) but lower than 122.56 g/100g reported for soyabean, (Nehdi, 2010) and 123 g/100g for pumpkin seed oil, (Nwabanne, 2012) . Iodine value gives an indication of the degree of unsaturation in the oil and thus gives a measure of susceptibility of oil to rancidity by oxidation. Studies have shown that the greater the degree of un-saturation the higher the iodine value and the greater the liability of the oil to become rancid by oxidation (Chabiri *et al.,*2009). *Dioclea reflexa* seed oil may be more stable than *Monodora myristica* seed oil. The peroxide value of *Dioclea reflexa* and *Monodora myristica* ranged between 5.85 and 6.3Meq/g oil. The range of peroxide value compares with *Dalbergia odorifera* seed oil (5.07 Meq/g oil), *Albizia julibrissin* (6.61 Meq/g oil) and higher than 4.13 Meq/g oil reported for *Monodora myristica* but lower than 8.85 reported for *Dioclea reflexa* (Yusuf and Lasisi, 2006; Nehdi, 2010; Ekeanyanwu *et al.,* 2010).

**Table 3:** Physicochemical properties of *Dioclea reflexa and Monodora myristica* seed oils

| **Parameter** | ***Dioclea reflexa*** | ***Monodora myristica*** |
|---|---|---|
| Colour | Golden yellow | Golden brown |
| Refractive index | 1.45±0.5 | 1.42±0.2 |
| Specific gravity | 0.97±0.2 | 0.94±0.2 |
| Acid value (mgKOH/g) | 6.30±0.30 | 5.60±0.20 |
| Free fatty acid (mgKOH/g) | 3.15±0.5 | 2.80±0.10 |
| Saponification value (mgKOH/g) | 212.00±0.20 | 196.00±0.20 |
| Unsaponifiable Matter (%) | 0.70±0.20 | 0.56±0.2 |
| Iodine value g/100g (Wijs) | 105.10±0.10 | 112.60±0.20 |
| Peroxide value (Meq KOH/g) | 5.85±0.5 | 6.30±0.10 |

Statistically significant differences between groups were compared using analysis of independent of t – test at probability level of 95%. Errors are standard deviation from the mean, n=3

**Table 4:** Fatty acid composition (%) of *Dioclea reflexa (DR) and Monodora myristica* (MM) seed oils.

| Fatty acid | | *Dioclea reflexa* | *Monodora myristica* |
|---|---|---|---|
| | C4:0 | 0 | 0.70±2.43 |
| | C6:0 | 0 | 1.15±0.05 |
| Caprylic acid | C8:0 | 0 | 0.14±0.58 |
| Capric acid | C10:0 | 0 | 0.19±0.02 |
| Lauric acid | C12:0 | 0 | 0.76 ±0.02 |
| Myristic acid | C14:0 | 0.34±0.02 | 0 |
| Pentadecanoic | C15:0 | 0.44 ±0.02 | 0.02±0.01 |
| Palmitic acid | C16:0 | 12.88±0.01 | 5.51±0.01 |
| Heptadecanoic | C17:0 | 0.04±0.02 | 0.10±0.02 |
| Stearic acid | C18:0 | 7.45±0.05 | 3.81±0.01 |
| Arachidic acid | C20:0 | 1.74±0.04 | 0.74±0.02 |
| Behenic acid | C22:0 | 4.29±0.02 | 0.33±0.03 |
| Lignoceric acid | C24:0 | 2.69±0.02 | 0.27±0.03 |
| Hexacosanoic | C26:0 | 0.11±0.01 | 0.04±0.02 |
| | C14:1 | 0.34±0.02 | 0 |
| Palmitoleic | C16:1 trans | 0 | 0.12±0.02 |
| Palmitoleic | C16:1 cis | 0.14±0.02 | 0.19±0.02 |
| | C17:1 cis | 0.03±0.02 | 0.13±0.03 |
| Oleic | C18:1 cis | 22.79±0.08 | 37.71±0.01 |
| Gadoleic | C20:1 cis | 0.36±0.02 | 0.22±0.02 |
| Linoleic | C18:2 trans | 0.20±0.10 | 0 |
| Linoleic | C18:2 cis | 28.95±0.05 | 44.51±0.01 |
| Linolenic | C18:3 | 2.44±0.04 | 1.16±0.04 |
| Unidentified | | 14.97±0.07 | 2.41±0.01 |
| Oleic/linolenic | | 0.79±0.02 | 0.85±0.05 |
| S: M:P(1:1:1) | | 1.27:1:1.31 | 1: 2.78: 3.29 |

Statistically significant differences between groups were compared using analysis of independent of t – test at probability level of 95%. Errors are standard deviation from the mean, n=3

Peroxide value depends on a number of factors such as state of oxidation (quantity of oxygen consumed), the method of extraction used and the type of fatty acids present in the oil. The range of values are lower than the Codex Alimetarius Commission (1982) stipulated permitted maximum peroxide levels of 10 Meq/g oil peroxide for unrefined olive oil (FAO/WHO, 1993).

**Fatty acid composition of *Dioclea reflexa* and *Monodora myristica* Seed Oils**

Fatty acid composition is one of the most valuable feature of oil, commonly employed to determine the identity and purity of the oil. The fatty acid composition of *Dioclea reflexa*and *Monodora myristica* seed oils is shown in Table 4. The predominant fatty acid is the unsaturated fatty acid, the percentage unsaturated fatty acid in the seed oils ranged from 55.05% – 83.83% consisting oleic, linoleic, linolenic and palmitoleic acids. The percentage saturated fatty acid in the seed oils ranged from 13.74 – 29.98% consisting palmitic acid, stearic acid, behenic acid, ligonoceric acid, arachidic acid, pentadecanoic acid, hexacosanoic, lauric acid, myristic acid, capric acid and caprylic acid. According to White (2000), there are basically three parameters to adjudge any oil as healthy cooking oil, that is, the ratio of saturated / monounsaturated / polyunsaturated fatty acids SMP (1: 1: 1), the ratio of essential fatty acids (omega 6 / Omega 3) and the presence of natural antioxidant. The oil of *Dioclea reflexa* contained 55.05% of unsaturated of which mono-unsaturated and polyunsaturated fatty acids represented 23.66% and 31.59% respectively.

The total unsaturated fatty acid of the oil was similar to cotton seed oil (69.42%) but lower than sesame (82.93%) and maize seed oils 84.90% – 86.61 % , (Ali *et al.,*2010; Mariod *et al.,*2011; Nehdi, 2011). The oil of *Monodora myristica* contained 83.85% unsaturated fatty acid of which mono unsaturation and poly unsaturation represent 38.15% and 45.67% respectively. The total unsaturated fatty acids are similar to that of groundnut 78.09% and soybean oils (80.96%), lower than grape seed oil (88.6%) but higher than 69.42% for cotton seed oil (Mariod *et al.,* 2011; Nehdi, 2011; Ali *et al.,* 2011). Unsaturated fatty acids are more prone to oxidation. In contrast, dietary intake of certain unsaturated fatty acids in particular, conjugated linoleic and fat - soluble antioxidants (α–tocopherol and carotenoids) has been linked to potential health benefits (Gillian *et al.,*2008). Linoleic acid was observed to be the most abundant fatty acid in the seed oils followed by oleic acid, palmitic acid and stearic acid and *Monodora myristica* is significantly higher in oleic and linoleic than *Dioclea reflexa* seed oil. Linoleic acid is an essential fatty acid which is indispensable for the healthy growth of human skin. It can be transformed by the organisms into series of long fatty acid chains, which are the precursors of eicosanoids (Nasri *et al.,*2005). Oleic acid is very important in nervous cell construction and has fundamental role in cardiovascular disease prevention. Trans fatty acids resulting from partial hydrogenation are implicated in incidences of certain chronic diseases. Studies have suggested that trans - fat consumption elevates low density lipoprotein cholesterol (bad cholesterol) and decreases the high density lipoprotein cholesterol (good cholesterol). There is no trans - fatty acid detected in *Monodora myristica* but very low amount were found in *Dioclea reflexa* (0.2%) which may not constitute health risk. The total unsaturated fatty acid (%) in seed oil varied with *Dioclea reflexa* having a lower percentage, which corroborates the deduction from the results of iodine value of the oil sample that the seed contain less degree of unsaturated fatty acids. The oleic/linoleic (O/L) ratio obtained in the seed oils of *Dioclea reflexa* and *Monodora myristica* were between 0.79 and 0.85 respectively. This ratio may be used to check for the level of purity and adulteration in seed oils. The values are lower than the values 1.13 - 4.22 reported for fluted pumpkin seed oils (Fagbemi, 2007). Oil stability has been associated with high oleic/linoleic (O/L) value; hence, both *Dioclea reflexa* and *Monodora myristica* may not be very stable. The Saturated/Monounsaturated/Polyunsaturated (SMP) ratio of the seed oils are 1.27: 1: 1.31 (*Dioclea reflexa*) and 1: 2.78: 3.29 (*Monodora myristica*) as compared to the ideal ratio of 1:1:1 may suggest that *Dioclea reflexa* seed oil will make more important contribution to human diet. The ideal intake ratio of omega-6/omega-3 is between 1:1 and 4:1. The value of omega-6/omega-3 ratio for both seed oils are far higher than the recommended range so the seed oils may not contribute antioxidant benefit to consumers since it has been adjudged that omega-6 and omega-3 imbalance is linked with serious health condition. The polyunsaturated to saturated (P/S) fatty acid ratio of *Dioclea reflexa* (1.06) and *Monodora myristica* (3.39) are lower than the value 3.92 reported for soybean oil. A high ratio of P/S is regarded favourable for the reduction of serum cholesterol and atherosclerosis and prevention of heart diseases (Nehdi 2011).

The most studied fraction of the unsaponifiable matter is that of sterols, which is frequently analyzed for tracking frauds (Rubio-Rodríguez *et al.,* 2010). This fraction has been considered as the major unsaponifiable fraction in many oils. Five sterols were determined in the seed oils of *Dioclea reflexa* and *Monodora myristica*. The seeds oils differ significantly at 95% confidence interval in the sterol composition (Table 5). Brassicasterol was not detected in both *Dioclea reflexa* and *Monodora myristica*. Cholesterol was not detected in *Monodora myristica*. Campesterol content of *Monodora myristica* is 20.68 mg/100g while that of *Dioclea reflexa* is 29.60 mg/100g. The values are close to 27.7 mg/100g reported for

Cephalocrotoncordofanus oil and 29.90 mg/100g for Phoenix canariensis (Nehdi *et al.,* 2010; Mariod *et al.,* 2011). It is however lower than the value of 42.9 mg/100g reported for groundnut oil, 43.9 mg/100g for cotton seed oil but higher than 13.0 mg/100g from sesame oil (Nehdi *et al.,* 2011). Campersterol has been reported to be involved in controlling cholesterol and lowering the risk of heart diseases. It is sometimes used to treat some specific prostate conditions (Awad *et al*., 2001). Stigmasterol content of *Monodora myristica* is 52.24 mg/100g while that of *Dioclea reflexa* is 125.43 mg/100g. *Dioclea reflexa* has a higher value than *Monodora myristica.* The value observed for *Monodora myristica* is close to 48.1 mg/100g reported for sesame oil but much higher than 5.3 mg/100g cotton seed oil, 24.3 mg/100g groundnut oil and lower than 72.8 mg/100g *Cephalocrotencordofanus* (Nehdi, 2011). Beta-sitosterols contents of *Monodora myristica* is 49.49 while that of *Dioclea reflexa*is 251.25 mg/100g. The values are close to 255 mg/100g reported for *Phoenix canarensis*. *Dioclea reflexa* has a significantly higher value of beta-sitosterols. Beta-sitosterols are known to be the principal sterol found in many seeds and oil seeds. Studies have shown that people with a diet containing 60 – 130 mg/day of plant β-sitosterol have lower incidence of prostate cancer (Awad *et al.,* 2001). Cholesterol was only present in the *Dioclea reflexa*seed oils which is 8.02 mg/100g and much higher than 1.42 mg/100g reported for *Phoenix canariensis*, (Nehdi *et al.,* 2010).

Cholesterol plays a major role in human heart health depending on the type, high cholesterol in serum is a leading risk factor for human cardiovascular disease (Ma, 2006), this is absent in *Monodora myristica* seed oil and this makes it a heart friendly oil.

## CONCLUSIONS

*Dioclea reflexa* and *Monodora myristica* seed flours are good sources of crude fat, protein, potassium, calcium, phosphorus and magnesium. Its choice of consumption to consumers in the diet would reduce the occurrence of nutritional deficiency and its associated health problem. This study has revealed that *Dioclea reflexa* and *Monodora myristica* seed oils are good sources of essential fatty acids. Brassicasterol was not present in both seed oils; hence they may have potentials for both domestic and industrial applications.

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