



---

## Physico-chemical properties and fatty acid composition of *Lagneraria siceraria* seed oil

T. O. Magu<sup>1,\*</sup>, E. A. Sunday<sup>2,\*</sup>, H. Louis<sup>1</sup>, P. U. Upla<sup>3</sup>,  
M. A. Ukpanah<sup>4</sup> and B. E. Nyong<sup>5</sup>

<sup>1</sup>Department of Pure and Applied Chemistry, University of Calabar, Nigeria

<sup>2</sup>Department of Chemistry, University of Uyo, Akwa Ibom State, Nigeria

<sup>3</sup>Department of Microbiology, Federal University Lafia, Nasarawa State, Nigeria

<sup>4</sup>Department of Microbiology, University of Uyo, Akwa Ibom State, Nigeria

<sup>5</sup>Department of Chemistry, Cross River University of Technology, Calabar, Nigeria

\*E-mail address: [esthersunday403@gmail.com](mailto:esthersunday403@gmail.com)

### ABSTRACT

Oil was extracted from the dehulled seeds of *Lagneraria siceraria* (bottle gourd) and analysed for physico-chemical properties, as well a fatty acid composition. Standard procedures were employed in all analysis. The seed oil was liquid at room temperature with percentage yield (23.65%). The oil was characterized in terms of specific gravity (0.918 g/cm<sup>3</sup>), refractive index (1.34), viscosity (26.46 X 10<sup>3</sup> poise), melting point (11-14.5 °C), moisture content (0.18%), saponification value (203.36 mg KOH/g), unsaponifiable matter (7.13%), iodine value (46.1 g/100g), peroxide value (7.5 meq/kg), free fatty acid value (18.42%), acid value (60.02 mg KOH/g) and ester value (143.34 mg KOH/g). It was also classified as non- drying (iodine value <115 g/100 g). The peroxide value indicates that the oil is less prone to rancidity with iodine value less than 30meq/kg. The high saponification value qualifies it for use in the manufacture of soaps and shampoos. Four classes of fatty acid were identified in the oil: palmitic acid (C16:1) (13.5 ± 0.21), stearic acid (C18:1) (6.5 ± 0.96), oleic acid (C18:1) (11.6 ± 0.62) and linoleic acid (C18:2) (68.4 ± 0.13). Linoleic acid was the most abundant fatty acid in the oil. The fatty acid content of the oil reveals that *L. Siceraria* seed oil could be a rich source of oil for domestic and industrial purposes if exploited.

**Keywords:** Cucurbitaceae, *Lagenaria siceraria*, seed oil, fatty acid, Linoleic acid

## 1. INTRODUCTION

Plant oils represent one of the key materials that can be obtained cheaply from biomass and processed readily to supply the appropriate raw material for chemical industries (Akintayo, 2002). Plants oils have both edible and non-edible applications such as: lubricants, soap production, cosmetics, insulating materials and biodiesel (Okpako *et al*, 2017; Akakuru *et al*, 2017; Sunday *et al*, 2016).

*Lagenaria siceraria* (Figure 1) is a vine grown for its fruit, which can either be harvested young and used as a vegetable, or harvested mature, dried, and used as a bottle, utensil, or pipe. It is also known by many other names that include: long melon, calabash plant, bottle gourd and Ayan-ukpok (Ibibio, Nigeria). The fresh fruit has a light-green smooth skin and a white flesh. The plant grows in a variety of shapes: they can be huge and rounded, small and bottle shaped, or slim and serpentine, more than a metre long (Akintayo, 2002). The gourd was one of the first cultivated plants in the world, grown not primarily for food, but for use as water containers (Akakuru *et al*, 2017).



**Figure 1.** Calabash gourd (*Lagenaria siceraria*)

There is a high demand of vegetable oils for domestic, industrial and medicinal use as a result of increase in population. Due to urbanization and alternative polymer products, bottle gourds which were used as containers for palm wine, ornament, bowls and basins are becoming obsolete. The cultivation of gourds is no longer common (Akintayo, 2009). They are being replaced by plastic and glass receptacles (Akintayo, 2009). Despite the vast nutritional and medicinal importance of the plant, little is known on the physico-chemical properties as well as fatty acid and mineral composition of the plant. Hence, the scanty report

on the medicinal, nutritional and industrial uses of *Lagenaria siceraria* seeds. Therefore, this research will provide information on the extraction of oil from *Lagenaria siceraria* seed, physical and chemical properties of the oil extract, fatty acid composition and possible uses of the oil.

## 2. MATERIALS AND METHOD

### 2. 1. Sample collection and treatment

The matured fruits of *Lagenaria siceraria* were collected from an abandoned farmland at Ikot Andem Itam, Itu Local Government Area, Akwa Ibom State. The fruits were manually broken with a sharp object and the seeds were washed to separate them from the spongy part of the fruit. The seeds were then sun-dried for six days. The dried seeds were packed in an airtight container prior to analysis. The analysis was carried out according to the standard method described by A.O. A. C. (1984).

### 2. 2. Lipid extraction

The seeds were handled carefully for cleanliness. The seed (140 g) was poured on the receiving funnel of the cold presser. The oil which dripped from the outlet was collected in a container which was previously weighed. The weight of the oil obtained was calculated by the formula below:

$$\text{Weight of oil (g)} = W2 - W1 \quad \text{---} \quad [1]$$

W1 = weight of empty container

W2 = weight of container + oil

### 2. 3. Determination of physical and chemical properties of the oil

#### Melting point

The *Lagenaria siceraria* seed extract (1g) was weighed into a test tube and kept in a refrigerator for 3 days to obtain a solid. Thereafter, the temperature of which the oil changed from liquid to solid was monitored and recorded.

#### Refractive index

A concave mirror was placed on the base a retort stand and a pin was clamped approximately to enable adjustment of its position until it coincided with the image at  $C_0$ . The distance  $C_0$  was measured and sufficient oil sample was poured in the mirror. The position of the pin was adjusted again until it coincided with its image at position  $C_1$ . The distance  $C_1$  was measured.

#### Specific gravity and Viscosity

Oil sample was filled into a density bottle and weighed at 60 °C, heated for 20 mins and allowed to cool. On cooling, the bottle was reweighed and the difference in weighed was

recorded as the specific gravity of the oil sample. The viscosity of the oil sample was determined using the Ostwald viscometer.

### Moisture contents

Weight of empty beaker and beaker with oil sample was obtained. The beaker with oil sample was placed in an oven for 6 hours at 105 °C. The beaker was then cooled in a dessicator and reweighed on cooling. The moisture content was calculated according to the formula below:

$$\text{Moisture content (\%)} = \frac{(B-A)-(C-A)}{(B-A)} \times 100 \quad \text{---} \quad [2]$$

### Acid value

The *Lagenaria siceraria* seed extract (1 g) was weighed and added to carbon tetrachloromethane (10 ml). Phenolphthalein indicator (2 ml) was added to the mixture. A blank determination was also carried out in the same manner. Titration was done with 0.1 M alcoholic potassium hydroxide until a color change was observed.

### Saponification value

The oil sample (2 g) was weighed into a conical flask. Ethanoic potash (25 ml) was added into the conical flask and the flask was put into a water bath to boil. On cooling, Phenolphthalein indicator (3 ml) was added to the flask and the solution was titrated against 0.5 M HCl until the end point was reached.

### Iodine value (HANUS Method)

The oil sample (1 g) was weighed into a conical flask and HANUS solution (25 ml) and 10ml of chloroform was added to the flask. The flask was kept in the dark for 6 hours. Next, potassium iodide (10ml) and starch indicator was added to the flask. A blank sample containing starch indicator was also prepared. Both oil sample and blank sample was titrated against 0.1 M Na<sub>2</sub>S<sub>2</sub>O<sub>3</sub> until an end point was reached.

### Peroxide value

A solution containing 2 g of the sample, 6ml of chloroform, 9 ml of ethanoic acid was added to 2ml of saturated potassium iodide and allowed to stand for 2 mins. Distilled water (3 ml) and starch indicator (2 ml) was added to the solution. The resulting solution was titrated against 0.1 M Na<sub>2</sub>SO<sub>4</sub> until end point was reached.

### Fatty acid

The seed extract (1.5 g) was added to 25 ml of ethanol in a conical flask. The mixture was brought to boil. The warm mixture was titrated against 0.1 M NaOH until a purple colouration was observed. Phenolphthalein was used as the indicator.

### Determination of unsaponifiable matter

Alcoholic potash (25 ml) was added to 1g of the oil extract in a flask and refluxed for 1 hour at 30 °C. Content of the flask was soaked with 50 ml of distilled water in a 250 ml separating funnel and allowed to stand until the formation of two layers was observed. The water layer was drained off while the remaining layer was transferred to a flask and evaporated to dryness using ether as solvent. The ether extract was pre-weighed in a beaker and heated to a constant weight in at 100 °C.

### Analysis of the fatty acid of methyl ester

The fatty acid composition was determined by GC- Flame Ionization Detection (FID). Methyl esters were obtained by hydrolysing the triglycerides of the oil with potassium-methanol. GC-FID analysis were performed using a Hewlet Packard 6890 series GC system equipped with a HP- 5 (30 m × 0.22 mm id, 5% dimethyl (siloxane, film thickness, 0.25) capillary column with a FID. Helium was used as the carrier gas (1ml/ min. flow rate). The column was maintained at an initial temperature of 220 °C for 5 mins and was then programmed at 2 °C/min. to final temperature of 220 °C and was maintained for 18 minutes.

## 3. RESULTS AND DISCUSSION

The physico-chemical properties of *L. siceraria* seed oil are presented in Table 1. The golden yellow oil extract was liquid at room temperature.

**Table 1.** Physical properties of *Lagenaria siceraria* seed oil

Analysis	Results
Colour	Golden yellow
Odour	Mild
State at 30 °C (room temperature)	Liquid
Melting point	11-13.5 °C
Percentage yield (%)	23.62
Specific gravity (g/cm <sup>3</sup> )	0.9181
Viscosity (poise)	26.46 × 10 <sup>3</sup>
Moisture content (%)	0.18
Refractive index	1.34

The saponification value, unsaponifiable matter, iodine value, peroxide value, free fatty acid value, acid value and ester value is presented in Table 2.

**Table 2.** Chemical properties of *Lagenaria siceraria* seed oil

Analysis	Results
Saponification value (mg KOH/g)	203.36
Unsaponifiable matter (%)	7.31
Iodine value (g/100g)	46.1
Peroxide value (meq/kg)	7.5
Free fatty acid (%)	18.42
Acid value (mg KOH/g)	60.02
Ester value (mg KOH/g)	143.34

### Fatty acid composition

The fatty acid composition of *Lagenaria siceraria* seed oil is shown in Table 3. The oil has four main fatty acids namely: palmitic (13.5%), stearic (6.55), oleic (11.6%) and linoleic acid (68.4%).

**Table 3.** Oil content and fatty acid profile of *Lagenaria siceraria* seed oil

Parameter	Value
Oil (%)	23.65
Palmitic acid (C16:0) (%)	13.5±02.1
Stearic acid (C18:0) (%)	6.5 ± 0.96
Oleic acid (C18:1) (%)	11.6 ± 0.62
Linoleic (C18:2) (%)	68.4 ± 0.13
Unsaturated compound	80
Saturated compound	20
Mono saturated	11.6
Poly saturated	68.4

The oil of *L. siceraria* has a yellow colour. It has no objectionable odour and remained liquid at room temperature.

### Percentage yield

The percentage yield of *L. siceraria* is 23.6%. This is comparable to those of cotton seed (18-28%), soya bean (11-25%), rubber (21-25%) (Kirschenbauer, 1965; Norris, 1965) and higher than *A. breviflorus* seed oil (22.9%) (Umerie *et al*, 2009). This value is lower than those obtained for various seed oils by other workers. Idourain *et al* (1996) reported a yield of 34.5-45.5% for *C. pepo* seeds and Martin (1998), reported a yield of 50% for melons, squashes and pumpkins. FAO. (1982) reported for sun flower (45.6%) and peanut (47.5%) but less than that for melon (*C. lanatus*) 59% by cherry, (1998), it had a melting point of 11-13 °C. The British standard stipulated that the lower the melting point of fruit oil, the better is the oil for making creams. It also had a specific gravity of 0.9181 g/cm<sup>3</sup> indicating that it is less dense than water.

The result of the refractive index 1.34, is slightly lower than the value of 1.46 obtained for *B. sapida* (Akintayo *et al*. 2002) and 1.45 obtained for *C. lanatus* (Oluba *et al*, 2008). This shows that the oil is not as thick as most drying oils whose refractive index were between 1.48 and 1.49 (Duel, 1951).

### Saponification value

High saponification values indicate high proportion of lower fatty acid. The saponification value of *L. siceraria* seed oil is 203.36 mg KOH/g. This high value indicates that the oil could be used in the manufacture of soap (Kirschenbauer, 1965). However, the saponification value was much lower than 242mg KOH/g in *L. siceraria* reported by Fokou *et al*, (2009). Nevertheless, the saponification value of the oil (203.36 mhKOH/g) was higher than 192.0 mg KOH/g in *L. siceraria* as presented by Chinyere *et al.*, (2009).

### Iodine value

*L. siceraria* seed oil can be classified as a non-drying oil as a result its iodine value (46.1 g / 100g ) which indicates that the oil contain high level of unsaturated fatty acid and is responsible for the liquid state of the oil at room temperature. The iodine value obtained in this work (46.1 g / 100g) agrees with 41.05 g / 100 g in *L. siceraria* reported by Chinyere *et al*, (2009). Higher values were found in peanut oil (86.0 g / 100 g) and soya bean oil (124.0 g / 100 g) (Aremuet *al* (2006).

### Acid value and free fatty acid value

The acid value and free fatty acid value obtained from *L. siceraria* seed oil were 60.02 mg KOH/g and 18.42% respectively. The acid value of *L. siceraria* (60.02 mg KOH/g) was relatively higher than 22.3 mg KOH/g reported by Fokou *et al* (2009). High acid values can result from high amounts of free fatty acid due to the method adopted in the seed processing. Duration of storage and drying of the seeds can increase the acid index. According to Aremu *et al* (2006), acid index can also be increased as a result of increase in temperature.

### **Peroxide value**

Peroxide value depends on a number of factors such as state of oxidation (quantity of oxygen consumed), method of extraction and type of fatty acid present in the oil. *L. siceraria* seed oil has a peroxide value of 7.5 meq/kg. This value is lower than the values recorded for *Citrullu slanatus* (8.34 meq/kg) and higher than *L. siceraria* (4.83 meq/kg) as reported by Chniyere *et al*, (2000).

### **Ester value and unsaponifiable matter**

The ester value and the unsaponifiable matter of *L. siceraria* seed oil is 143.34 mg KOH/g and 7.31% respectively. The ester value was obtained by subtracting the acid value from the saponification value (Aremu *et al* (2006). This value is lower than 172.2 mg KOH/g recorded for *A. breviflorus* seed oil reported by Umerie *et al* (2009).

### **Oleic acid**

The C18:1 level in *L. siceraria* seed oil (11.6%) was significantly lower than the oleic acid levels in egusi seed oil (14.6) reported by Oluba *et al* (2008) and higher than 9.4% in *L. siceraria* seed oil reported by Hassan *et al* (2008). Low levels of C18:1 is indicative of low level of Mono-unsaturated Fatty Acid (MUFA) in oils. MUFA is known as an agent that increases HDL (good cholesterol) and decreases LDL (bad cholesterol) in oils. This was observed in a study by Umerie *et al* (2009) where rats were fed with experimental chees containing MUFA; this resulted in a significant reduction of LDL (31%) in the rats.

### **Linoleic acid**

The results presented in Table 3 reveal that linoleic acid is the most abundant fatty acid present in the oil extract with a value of 68.4%. This result competes favourable with 70% in *L. siceraria* reported by Chimsole and Hopkins (1964). *L. siceraria* contains high amount of linoleic fatty acid and could be a good raw material for collector preparation, especially in the phosphate flotation where linoleic acid has been reported to have a better recovery and yield in comparison to other fatty acids (Aremu *et al*, 2006).

### **Unsaturated fatty acid**

Unsaturated fatty acid content in the oil extract is 80%. This value is higher than 57.4% present in *C. lanatus* (Olubaet *al*, 2008) and 78.8% present in *L. siceraria* (Aremu *et al*, 2006). High levels of unsaturated fatty acids in *L. siceraria* is probably due to high level of linoleic acid. This shows that the oil is a good source of poly unsaturated fatty acid. Lower percentage of linoleic acid in oils improves flavour, quality and oxidation stability of the oil.

The higher the linoleic content of an oil, the more suitable it is for frying. Linoleic acid and oleic acids are primary products of plant polyunsaturated fatty acids (PUFAs) synthesis. The presence of these essential fatty acids in the oil is an indication that the oil is good for human consumption. These fatty acids when present in diet can control heart diseases, diabetes and cancers (Aremu *et al*, 2006).



#### 4. CONCLUSION AND RECOMMENDATION

*Lagneraria siceraria* seed oil has a good yield which can be processed for commercial purposes. The physico-chemical composition of the seed oil revealed that the oil could be used for industrial applications such as soap and lubricants production due to its high saponification value. The acid and the peroxide values were high due to long duration of drying, increase in temperature and method of processing of the seeds but these values are within recommended limits. *Lagneraria siceraria* seed oil is rich in linoleic acid suggesting that it could be good for cooking and frying. The abundance of linoleic acid followed by oleic acid makes the oil a good agent for fight against cardiovascular diseases. *Lagneraria siceraria* seed oil has the potential to substitute several materials used to manufacture oil in the chemical industry. In order to extend usage, this oil should be refined to improve the taste and colour.

#### References

- [1] Akakuru, O. U., Magu, T. O., Louis, H., Maitera, O. N., Fidelis, T. T. and Bisong, E. A. (2017). comparative study of some heavy metal concentrations in Coconut Fluid and Milk obtained from Taraba, Imo and Adamawa States, Nigeria. *International Journal of Scientific and Research Publications*, 7(3), 138-140
- [2] Akintayo, E. T., Adebayo, E. A. and Arogundade, L. A. (2002). Chemical composition, phytochemical and functional properties of Akee (*Bsapida*) pulp and seed flows. *Food Chem.* 77, 333-336
- [3] Aremu, M. O., Olonisakin, A., Bako, D. A. and Madu, P. C. (2006). Compositional studies and physicochemical characteristics of Cashew nut flour. *Pak. J. Nutr.* 5, 328-333
- [4] Cherry J. (1998). Egusi seeds are high in oil and protein. *Oil Crops Processing* 1, 4-8
- [5] Chineye, G. C., Akugwo, E. J., Chineye, N. I. and Ugbogu, A. E. (2009). Nutritional value of *Lagenariasphaerica* seed (wild bottle gour) from South- Eastern Nigeria. *Parkistan Journal of Nutrition* 8 (3), 284-287
- [6] Fokou, E., Achu, M. B. and Tchouanguiep, M. F. (2004). Preliminary nutritional evaluation of five species of egusi seeds in Cameroun. *Afr. J. Food Agric. Nur. Dev.* 4, 1-11
- [7] Idiourain, A., Kohlhepp, E. A., Weber, C. W., Warid, W. A. and Martinez T. J. (1996). Nutrient constituents from eight lines of Naked Squash (*Cucurbita pepo*). *J. Agric. Food Chem.* 44, 731-734
- [8] Kirschenbauer, H. G. (1965). *Fats and Oils: An outline of their chemistry and Technology*. Reinhold Publishing and Co., New York.
- [9] Martin, F. (1998). Cucurbit seeds as possible oil and protein sources for small scale household use in the hot humid tropics. *Oil Crops Processing.* 1, 1-4

- [10] Oluba, O. M., Ogunlowo, Y. R., Ojieh, G. C., Adebisi, K. E., Eidangle, G. O. and Isiosio, I. O. (2008). Physicochemical properties and fatty acid composition of *Citrullus lanatus* (Egusi melon) seed oil. *Journal of Biological Sciences*, 8 (4), 814-817
- [11] Okpako, E. C., Louis, H., Magu, T. O., Akwo, J. K., Akakuru, O. U. and Bisong, E. A. (2017). Phytochemical screening and proximate nutritional analysis of brown leaves of Indian almond (*Terminalia catappa* L.). *International Journal of Scientific Research Publications*, 7 (3), 141-144
- [12] Umeri, S. C., Onuagha, M. C. and Nwobi, S. C. (2009). Characterization and utilization of *Adenopus breviflorus* Benth (Curcubitacae) seed oil. *Natural and Applied Sciences Journal* 10 (1), 104-111
- [13] Å. Frostegård, A. Tunlid and E. Bååth. Phospholipid Fatty Acid Composition, Biomass, and Activity of Microbial Communities from Two Soil Types Experimentally Exposed to Different Heavy Metals. *Appl. Environ. Microbiol.* November 1993 vol. 59 no. 11 3605-3617
- [14] Christopher D. Stubbs, Anthony D. Smith, The modification of mammalian membrane polyunsaturated fatty acid composition in relation to membrane fluidity and function. *Biochimica et Biophysica Acta (BBA) - Reviews on Biomembranes* Volume 779, Issue 1, 27 January 1984, Pages 89-137 [https://doi.org/10.1016/0304-4157\(84\)90005-4](https://doi.org/10.1016/0304-4157(84)90005-4)
- [15] van Dijk, P.W.M., de Kruijff, B., van Deenen, L.L.M., de Jier, J., and Demel, R.M. (1976). *Biochim. Biophys. Acta* 455, 576–587
- [16] Valtersson, C., van Duyn, G., Verkleij, A.J., Chojnacki, T., de Kruijff, B., and Dallner, G. (1985). *J. Biol. Chem.* 260, 2742–2751
- [17] Stokes, C.E., and Hawthorne, J.N. (1987). *J. Neurochem.* 48, 1018–1021.
- [18] Ellison, D.W., Beal, M.F., and Martin, J.B. (1987). *Brain Research* 417, 389–392.
- [19] Bowen, D.M., Smith, C.B., and Davison, A.N. (1973). *Brain* 96, 849–856.
- [20] Brookshank, B.W.L., and Martinez, M. (1989). *Mol. Chem. Neuropathol.* 11, 157–185.
- [21] Khachaturian, Z.S. (1985). *Arch. Neurol.* 42, 1097–1104.
- [22] Morrison, W.R., and Smith, L.M. (1964). *J. Lipid Res.* 5, 600–608.
- [23] Vanderkooi, G. (1974). *Biochim. Biophys. Acta* 344, 307–345.
- [24] Stier, A., and Sackmann, E. (1973). *Biochim. Biophys. Acta* 311, 400–408.
- [25] Valtersson, C., and Dallner, G. (1982). *J. Lipid Res.* 23, 868–876

( Received 10 April 2017; accepted 27 April 2017 )