Chemistry Research Journal, 2017, 2(6):197-204

Available online www.chemrj.org

Research Article

ISSN: 2455-8990
CODEN(USA): CRJHA5

Physico-Chemical Analysis of Oil Extracted from *Rothmannia longiflora* Seeds

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Abstract *Rothmannia longiflora* are usually shrubs or small trees which are widely available in Nigeria and known with different native names such as Katanbiri, Okukin, Asuri, Asogbodu among others. The aim of this study was to investigate the physico-chemical properties of oil yield from *Rothmannial longiflora* seeds using acetone and n-Hexane solvent extracts. Oil was extracted using saturation method. 25g of the powdered sample was accurately weighed twice and poured into 250ml conical flask separately. 50ml of n-hexane and acetone was added separately to the content of the two well labeled conical flasks respectively and covered with aluminum foil. Mixture was decanted and filtered using filtration method in order to separate the oil. The results showed that n-Hexane extract has higher acidic value (6.1±0.31 Mg/KOH/g) than acetone extract which had (1.6 ±0.22 Mg/KOH/g). Acetone extract had a higher percentage oil yield of 36.5% while n-Hexane extract had 32.7% oil yield. Both acetone and n-Hexane extracts have similar moisture and ash contents. From this study, it can be deduced that acetone extract of *Rothmannia longiflora* seeds has a higher oil yield than n-Hexane extract of the same seeds. This is an indication that *Rothmannia longiflora* seeds contain useful amount of oil that could be used for industrial and/or pharmaceutical purposes.

Keywords *Rothmannia longiflora*, physico-chemical, properties, acetone, n-Hexane

Introduction

Rothmannia longiflora (Family of Rubiaceae) are usually shrubs or small trees widely distributed in the forest of tropical Africa and occasionally herbs or climbers which grow in old farms of secondary forest [1]. It is found in the undergrowth of primary as well as secondary forest from sea- level up to 170m altitude.

The plant is widely distributed in tropical and sub-tropical regions of the globe and occasionally found in cold regions. They very much available in Ghana, Nigeria and Congo. They are also found from east of Gambia to Sudan to Kenya through south Tanzania and Angola [2]. The fruits of *Rothmannia longiflora* are ingredients of common anti-inflammatory remedies used locally [3] and have also been reported in the treatment of malaria, measles, wound [2], dysentery, fever and cancer [3,5].

The stem of these plants are used in making shifts of long- handled chisels in Sierra Leone, used in harvesting bunches of palm oil. The stems are also used in making spear handles and as chewing stick in Ghana [6]. The stem, fruits as well as seeds of *Rothmannia longiflora* have been reported to be useful in the making of blue- black markings on the hands, face and body sometimes to imitate tattooing in some parts of Africa [7].

Various oils are obtained from different sources which include the common seed/vegetable oil [8]. Seed oils gotten from plant sources have been shown to have nutritive and calorific values which make them important in diet. They are also good sources of edible oils [8,9]. The yield composition, physical and chemical properties of the oils determines its utilization in various applications [10].

Figure 1: The seeds of Rothmannia longiflora

Several factors such as agronomic technique, seasonal conditions, different analytical method, duration of storage, pressing technology etc affect the quality of oils obtained [11].

Valuable plants products with large industrial and domestic potential remain unexploited and *Rothmannia longiflora* is one of such plants. However, the increase demand of useful oils which has led to increase in the importation of cooking oils has further support the need to source for local oils bearing seed which can be use in the production of oils for consumption and industrial application [10].

The result of this study can be used to determine the suitability of these oil seed as substitute for more conventional oil seeds like soya bean, palm, groundnut, and coconut oils.

The aim of this study was to carryout physicochemical analysis of oil extracted from the seed of *Rothmannia longiflora* using acetone and n-Hexane.

Materials and Methods

Sample Collection, Identification and Treatment

The materials used in this research work were the routine laboratory apparatus and chemicals, which were of analytical grade.

The sample of *Rothmannia longiflora* seed, were obtained from Lungu Village 18km, away from Shagari Town of Shagari Local Government, Sokoto State, Nigeria. The sample was collected in November, 2015. The sample was taken to the Department of Botany of the Usmanu Danfodiyo University Sokoto, Nigeria for proper identification.

Sample Treatment

The sample obtained was washed with water but not excessively to remove dust, and other impurities. The sample were then dried under the shade for two weeks and then grounded into fine powder sieved using 22 mm mesh, the fine powder was stored in an air tight polyethene bag for further analysis.

Oil Extraction

Liquid-liquid extraction, also known as solvent extraction and portioning, is a method to separate compounds based on their relative solubility's in two different immiscible liquids, usually water and an organic solvent. It is an

extraction of a substance from one liquid phase into another liquid phase. Liquid-liquid extraction is a basic technique in chemical laboratories, where it is performed using a separator funnel. This type of process is commonly performed after a chemical reaction as part of the work-up.

In other words, this is the separation of a substance from a mixture by preferentially dissolving that substance in a suitable solvent. By this process a soluble compound is usually separated from an insoluble one [12].

In this present study, the oil was extracted using saturation method which consists of solvent and sample. Twenty-five grams (25g) of the powdered sample was accurately weighed twice and poured into 250ml conical flask separately and then 50ml of n-hexane and acetone was added to the content of the conical flask respectively and covered with aluminum foil and conical flask and covered with aluminum foil and label as n-hexane and acetone extracts, both were kept for 48 hours in other to obtain maximum oil yield and make a homogenous mixture. The mixture was then decanted and filtered using filtration method in order to separate the oil, the mixtures were transferred into to two different beakers leveled as acetone & n-hexane.it was then placed into oven at 60^{o}c for one hour in order to allow the remaining solvent to evaporate. The raw oil obtained was pale yellow color with high percentage in acetone than n- hexane and transferred into sample bottles for analysis: this method was carried out twice in order to have more percentage yield of oil from the seed. The mean and standard deviation was calculated as described by Yunusa *et al*., [13].

Figure 2: *Oil extracted from the seeds of Rothmannia longiflora*

Percentage Yield of Oil

The solvent free-oil was placed into a pre-weighed beaker, the beaker was place on the electrical weighing balance and the weight was measured using balance in the laboratory and calibrate into zero before pouring the oil into the beaker to take the readings. The percentage yield of the oil was calculated using equation 3 below [13]

Percentage yield of oil $= \frac{W_1}{W_O} \times 100$ ……. Equation (3)

Where:

W_o = Percentage powdered sample in gram (g)

W_1 = Percentage oil after extraction in gram (g)

Determination of Acid Value

The acid content is referring to the number of Potassium hydroxide (KOH) required to neutralize 1g of oil and it measures used to estimate the storage quality of the oil [14].

Two gram (2g) each of *Rothmannia longiflora* seed oil was poured separately into three different beakers, each beaker was placed into water bath and heated at 60 ^{o}C for 15 minutes, and 1 drop of phenolphthalein was added to give pink color, 0.1M solutions of KOH was titrated into the beaker till the color changed to colorless and forms some crystal.

The procedure was repeated 3 times for both the samples and blank. Acid value of KOH of the sample can be calculated using the equation (4) below [14]

$$= \frac{(A-B)\times M \times 56.1}{W}$$

.....Equation (4)

Where: A = volume, ml of standard alkali used in the titration

B = volume, ml of standard alkali used in the titrating the blank

M = normality of standard alkali

W = mass, grams of sample [14]

Determination of Saponification Value

The saponification value is the amount of alkali necessary to dissolve a definite quantity of the sample. It is expressed as the number of potassium hydroxide (KOH) required to saponify (dissolve) 1g of oil [14]

Procedure:

Two grams (2g) of *Rothmannia longiflora* seed oil was poured into a beaker and the content of the beaker was heated at 60^{o}C for 2 minutes on a water bath in order to have homogenous mixture, 1 drop of phenolphthalein was added and the mixture was titrated with 0.1M KOH solution until the solution turns pink color and produced some soap bubbles [14]

This was calculated using the equation (5).

$$SV = \frac{N(A-B)\times 56.1}{W}$$ …… equation (5)

Where

B = volume of titrate, ml of blank

A = volume of titrate, ml of sample

N = normality of standard alkali

Determination of Iodine Value

Oil sample of 0.5m was dissolved in 20cm^3 of chloroform in 500cm^3 conical flask and 25% Wij's solution was added. Then was shaken vigorously to ensure that both oil sample and reagent were mixed properly and then the mixture was allowed to cool in the dark place for one hour (1H). Also 20cm^3 of 10% potassium iodide solution was added followed by 100 cm^3 distilled water, and using starch solution as an indicator. The solution was then titrated with standard 0.1M sodium thiosulphate ($Na_2S_2O_3$) solution until the blue color disappeared. A blank titration was also conducted, without the oil sample [14]

The iodine value of a sample is calculated using the equation (6) below:

$$IV = \frac{(TB-TV)\times N \times 12.69}{W}$$ …… equation (6)

TB = titer blank

TV = titer value

W = weigh of oil

N = 0.1

Determination of Density

Density of a substance is the relationship between the mass of the substance and how much space it takes up volume [15].

Procedure: Measuring cylinder was place on weighing balance, and weighing balance was calibrated into zero, oil was pour inside the Measuring cylinder up to Mark (volume) and the gram of the oil was recorded as mass. The density of oil can be calculated using the following equation [15].

Density $(D) = \frac{M}{V}$ …….. equation (9)

Where

$M =$ Mass of the oil (gram);

$V =$ Volume of oil in (ml)

Determination of Moisture Content of the Powdered Sample

This method measures the amount of samples loss due to drying at a temperature of 105^{o}C. Two gram (2g) of powdered sample of *Rothmannia longiflora* was weighed, the crucible containing the sample was placed into an oven at temperature of 105^{o}c for six hours (6 hrs).

The clean watch glass containing the dry sample was then weighed.

The moisture of sample was calculated using equation (2) below [16]

Moisture content $= \frac{W_1 - W_2}{W_1 - W_O} \times 100$ ……. Equation (2)

Where W_o = weight of empty clean watch glass

W_1 = weight of sample in clean watch glass

W_2 = weight of dry sample in clean watch glass.

Determination of Ash Content of the Powdered Sample

Two gram **(**2g) powder of *Rothmannia longiflora* was weighed, the crucible containing the samples was placed into the Lenton furnace thermostat at 600 ^{o}C and allowed to burn the powder sample for 3 hours until the content become ash. The crucible containing the ash sample was weighted using electrical weighing balance, the ash content was determined using the equation 1 [17].

Ash content (%) $= \frac{W_2 - W_O}{W_1 - W_O} \times 100$ …….. equation (1)

Where W_O = weight of empty crucible

W_1 = weight of sample before ashes + weight of the crucible.

W_2 = weight of ash sample + weight of crucible after ashes.

Results

The chemical characteristics of oil extracted from the seed of *Rothmannia longiflora* are shown in Table 1.

Table 1

Parameter	**Values% Acetone**	**Values % n-hexane**
Moisture content	36.5 ±0.10	36.5 ± 0.10
Ash content	2.5 ±0.22	2.5 ±0.22
Acid value (Mg/KOH/g)	1.6 ±0.22	6.1±0.31
Iodine value (Kg/100 g oil)	23.27±0.20	15.44±0.10
Density (g/cm^3)	1.56 ± 0.70	0.96 ± 0.11
Percentage yield	12.5 ± 0.15	14.5 ± 0.27
Saponification (Mg KOH/g)	57.94±0.88	27.11±0.43

Discussion

Percentage Yield

The percentage yield of the extracted oil from *Rothmannia longiflora* seed was 12.5% in acetone and 14.5% in n-Hexane. The result indicated that the seed is a less potential source of lipid oil compared to tiger nut (35.56± 2.81%) and ground nut (47.3±7.5%) as reported by Linsen *et al.*, [18].

Acid Value

The acid value or acid number gives the amount of free acid present per gram of the sample which is determined by titration with standard solution of potassium hydroxide [19]. The acid value of the oil extracted from the seed of *Rothmannia longiflora* was found to be 1.6± 0.22 and 6.1± 0.31 for acetone and n-Hexane respectively. These results obtained were found to be very low compared to that of tiger nut 193. 44 and melon seed 204 ± 0.58 as reported in previous works [20] and [21]. Acid content (acidity value) is a measure of the extent to which hydrolysis has liberated fatty acid from their ester linkage with the parent triglyceride molecule, thus it is one of the most frequently determined quality used during oil production, storage and evaluation.

Saponification Value

The saponification value of oil is a measure of molecule weight of its acid present in the fat. Saponification process is the hydrolysis of triglyceride into glycerol and potassium salt of the fatty acid and using a KOH in alcohol. Therefore, the saponification value of *Rothmannia longiflora* seed oil is 57.97 ±0.88 in acetone and 27.11±0.43 in n- hexane. Hence, the saponification value shows that the oil is potential for soap making. In comparison with tiger nut and olive oil, *Rothmannia longiflora* seed oil has high saponification value in acetone which has 57.97± 0.88 and lower in n- hexane i.e. 27.11± 0.43 and also the saponification values are quite higher than tiger nut and olive oil 186.07±1.95.as reported by [18].

Iodine Value

The iodine value of an oil sample is a measure of unsaturation found present in the oil fat. It is the percentage of halogen absorbed and this is calculated in term of iodine. The reaction includes the addition of halogen to unsaturated bonds [19]. The iodine value for *Rothmannia longiflora* seed oil was 23.27±0.20 in acetone and 15.44± 0.10 in N-hexane. When compared with tiger nut (*Cyperus esculentus*) seed oil iodine value of 67.69±2.29 and bean seed with iodine value of 98.20±1.75 respectively, the iodine value of *Rothmannia longiflora* is lower. Hence the iodine value showed that *Rothmannia longiflora* seed is a non-drying oil and as such it is unsaturated thus making it suitable for utilization in industries especially in soap manufacturing [22].

Density (g/cm^3)

The density (g/cm^3) of *Rothmannia longiflora seed* is found to be 1.56% in acetone and 0.96% in n-Hexane which is higher compared to that of tiger nut seed (*Cyperus esculatus*) which is 0.78±03/g/cm^3 [19].

Moisture Content

This is the amount of moisture in seed sample. It may be water or liquid which tends to increase the density floral seed sample. *Rothmannia longiflora* seeds exhibits relatively percentage moisture of 36.50±0.10% in both acetone and N-hexane. Thus the seed contains fat and vitamins as well as some other mineral elements. The moisture contents of *Rothmannia longiflora* is less than that of tiger nut and olive oil 24±0.30 and 27±1.24 respectively [18].

Ash Content

The ash content of seed is the measure of its mineral content. The ash content of *Rothmannia longiflora was* found to be 2.5±0.22% in both acetone and n- hexane. The ashing show that some mineral elements are volatile. The ashing of the powdered sample of *Rothmannia longiflora* indicates the high volatility of the organic compound present in the seed. In comparison with tiger nut seed *(Cyperus esculatus*) and olive oil, *Rothmannia longiflora* has

lower ash content as tiger nut seed (C*yperus esculatus*) and olive oil has 16± 0.54 and 12.63±1.84 respectively [23]. Low ashing values of *Rothmannia longiflora* as observed in this study shows that the sample is rich in organic compounds.

Conclusion

The result obtained in the present study showed that *Rothmannia longiflora* seed is a rich oil source. Acetone extract of *Rothmannia longiflora* seeds has a higher oil yield than n-hexane extract of the same seeds. This is an indication that *Rothmannia longiflora* seeds contain useful amount of oil that could be used for industrial purposes. The fact that it leaves can be used in the treatment of diabetes, fever various disease conditions such as gastrointestinal problems, further suggests that its oil could be useful and valuable for industrial and pharmaceutical purposes.

Further research need to be carried out in order to isolate and identify more constituents of the plant (*Rothmannia longiflora*) using various extraction solvent. We also recommend further study of the plant parts such as phytochemical analysis to evaluate its medicinal values.

References

1. Joffe, P. (2005). Creative gandeny with indigenous plants. A South Africa Guide. Briza Publications, Pretoria.
2. Coates Palgrave, K. (2002). Trees of southern Africa. New edition revised and updated by Meg Coates Palgrave. *Cape Town: Struik*, 1212, 118.
3. Abubakar, M. S., Musa, A. M., Ahmed, A & Hussaini, I. M. (2007). The perception and practice of traditional medicine in the treatment of cancers and inflammations by the Hausa and Fulani tribes of Northern Nigeria. *Journal of Ethnopharmacology*, 111(3), 625-629
4. Ikpi, D. E., Obembe, A. O. & Nku, C. O. (2009). Aqueous Leaf Extract of Rothmannia Longiflora Improves Basal Metabolic Rate and Electrolyte Parameters in Alloxan Induced Diabetic Rats. *Nigerian Journal of Physiological Sciences*, 24(1)
5. Ezugwu, A. C., Njoku, O. U., Ozoemena, N. F., Joshua, P. E., Nwachi, U. & Nwodo, O. F. C. (2010). Anti-Inflammatory Effects of *Rothmannia longiflora* Fruit Extract. *Research Journal of Pharmacognosy and Phytochemistry*, *2*(4), 327-330.
6. Hussain, J., Bahader, A., Ullah, F., Rehman, N., Khan, A. L., Ullah, W. & Shinwari, Z. K. (2009). Proximate and nutrient analysis of the locally manufactured herbal medicines and its raw material. *J. Am. Sci*, 5(6), 1-5
7. Jansen, P. C. M. (2005). *Rothmennis longiflora* Salish. In: Jansen, P. C. 19 and Cardon, D. (Edns). PROTA 3: Dyes and tannins/Colorants et tannins. PROTA, Wageningen, Netherlands.
8. Akubugwo, I. E., Chinyere, G. C. & Ugbogu, A. E. (2008). Comparative studies on oils from some common plant seeds in Nigeria. *Pak. J. Nutr*, 7(4), 570-573.
9. Knothe, G. & Steidley, K. R. (2005). Kinematic viscosity of biodiesel fuel components and related compounds. Influence of compound structure and comparison to petrodiesel fuel components. *Fuel*, 84(9), 1059-1065
10. Aluyor, E. O. & Ori-Jesu, M. (2008). The use of antioxidants in vegetable oils–A review. *African Journal of Biotechnology*, 7(25).
11. Bringmann, G., Hamm, A., Kraus, J., Ochse, M., Noureldeen, A., & Jumbam, D. N. (2001). Gardenamide A from *Rothmannia urcelliformis* (Rubiaceae)− Isolation, Absolute Stereostructure, and Biomimetic Synthesis from Genipine. *European Journal of Organic Chemistry*, 10; 1983-1987.
12. Kolthoff, I. M., Sandell, E. B., Meehan, E. J. & Bruckenstein, S. *Quantitative Chemical Analysis*, Macmillan: New York, 1969, p. 353.
13. Yunus, M.M., Zuru, A.A., Faruq, U.Z. & Aliero, A.A. (2013). Assessment of Physicochemical Properties of Biodiesel from *African Grapes* (*Lannea microcarpa* Engl. & K. Krause). *Nigerian Journal of Basic and Applied Sciences*, 21(2), pp.127-130.

14. Adejumo B. A., Olorunsogo S. T. & Njoku C. B. (2015). Qualities of Watermelon Seed Oil Extracted at Different Moisture Content. *International Journal of Advanced Engineering Reasearch and Technology.* 3 (3): 2348-8190.
15. Mukhtar, M. & Dabai, M.U. (2016). Production and Fuel Properties of Biodiesel from Gingerbread Plum (*Parinari macrophylla*) Seed Oil Using MgO/Al_2O_3 Catalyst. *American Journal of Environmental Protection*, 5 (5); 128-133.
16. Sokoto, M. A., Hassan, L. G., Dangoggo, S. M., Ahmad, H. G. & Uba, A. (2011). Influence of fatty acid methyl esters on fuel properties of biodiesel produced from the seeds oil of Curcubita pepo. *Nigerian Journal of Basic and Applied Sciences*, *19*(1).
17. Ceirwyn, S.J. (1998). Analytical Chemistry of Food. Chapman and Hall Publisher, London, pp. 75-77.
18. Linssen, J.P.H., Kielman, G.M., Cozijnsen, J.L. & Pilnik, W. (1988) Composition of chufa and olive oils. *Food Chem*., 28:279–285.
19. Abdulkarim, S. M., Long, K., Lai, O. M., Muhammad, S. K. S. & Ghazali, H. M. (2005). Some physico-chemical properties of *Moringa oleifera* seed oil extracted using solvent and aqueous enzymatic methods. *Food Chemistry*, *93*(2), 253-263
20. Zhang, H. Y., Hanna, M. A., Ali, Y. & Nan, L. (1996). Yellow nut-sedge (*Cyperus esculentus* L.) tuber oil as a fuel. *Industrial crops and products*, *5*(3), 177-181
21. Barminas, J. T., Maina, H. M., Tahir, S., Kubmarawa, D. & Tsware, K. (2001). A preliminary investigation into the biofuel characteristics of tigernut (*Cyperus esculentus*) oil. *Bioresource technology*, *79*(1), 87-89.
22. Linssen, J.P.H., Cozijnsen, J.L. & Pilnik, W. (1989) Chufa (*Cyperus esculentus*): a new source of dietary fiber. *J Sci Food Agric*., 49:291–296.
23. Belewu, M.A. & Belewu, K.Y. (2007). Comparative physico-chemical evaluation of Tigernut, soybean and coconut milk sources. International Journal of Agriculture and Biology, 9(5):785-787.