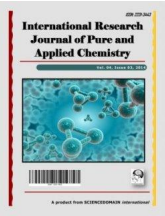

International Research Journal of Pure & Applied Chemistry

18(1): 1-5, 2019; Article no.IRJPAC.46340
ISSN: 2231-3443, NLM ID: 101647669

Physicochemical Analysis of *Moringa oleifera* Seeds

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Authors' contributions

This work was carried out in collaboration between all authors. Authors COI and MCU designed the study, performed the statistical analysis, wrote the protocol and wrote the first draft of the manuscript. Author COI managed the analyses of the study. Authors NMO, GUO, PIU and HON managed the literature searches. All authors read and approved the final manuscript.

Article Information

DOI: 10.9734/IRJPAC/2019/v18i130081
Editor(s):
(1) Dr. Wolfgang Linert, Professor, Institute of Applied Synthetic Chemistry, Vienna University of Technology Getreidemarkt, Austria.
Reviewers:
(1) P. Saravana Kumari, Rathnavel Subramaniam College of Arts and Science, India.
(2) Valdir Florencio da Veiga Junior, Military Institute of Engineering, Brazil.
(3) Tsado Daniel Babanma, Federal university of Technology Minna, Nigeria.
Complete Peer review History: http://www.sdiarticle3.com/review-history/46340

Original Research Article

Received 27 October 2018
Accepted 22 January 2019
Published 19 February 2019

ABSTRACT

The soxhlet extraction of Moringa seed oil was used to determine the proximate and physicochemical screening. The parameters obtained for the proximate screening were 7.64% moisture content, 4.05% ash content, 29.65% crude fat, 34.92% crude protein and 52.30% carbohydrate while the values obtained for the physico-chemical screening were 62.45% for Iodide, 1.1% for specific gravity, 9.84 for free fatty acid, 162.84% for saponification value, 4.10% for peroxide value, 1.46% for refractive index, 10.50% for viscosity and 5.95% for acid value. The results showed that *Moringa oleifera* seeds and seed oil could be employed for edible and commercial purposes.

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Keywords: Physico-chemical; Moringa oleifera; soxhlet extraction; medicinal.

1. INTRODUCTION

Moringa has long been cultivated and its parts consumed and used for a variety of purposes across the tropics [1]. This is because of its impressive range of nutritional and medicinal value [2]. All part of the moringa tree (leaves, seeds, root and flowers) are not only suitable for human consumption but also for animal consumption [3]. The leaves which are rich in protein, mineral, B- carotene and antioxidant compound are used not only for human, animal nutrition but also in traditional medicine [4]. The seeds contain a significant amount of oil (up to 40%) with a high quality fatty acid composition (oleic acid > 70%) after refining a notable resistance to oxidative degradation [5].

Moringa oleifera has received a great amount of attention as "natural nutrition of the tropics" [6]. The leaves, fruits, and immature pods and flowers of this tree are locally used as vegetable [7]. *Moringa oleifera* seeds have antimicrobial activity and are utilized for waste water treatment [8]. In Sudan, dry *Moringa oleifera* seeds are used in place of alum by rural woman to treat highly turbid Nile water [9].The seeds of *Moringa oleifera* are considered to antipyretic, acrid, and bitter [10].

It has also been found that extract obtained from the leaves of Moringa is 80% ethanol and contains growth enhancing principles for higher plants [11]. In ethno medicine, *Moringa oliefera* leaves have been used by local traditional healers in treatment of various ailments such as gastric discomfort, stomach ulcers, diarrhea, dysentery and skin infection [12].

Ghasi et al. [13] have reported that administration of crude leaf extract of *Moringa oleifera* along with a high fat diet decreased the high fat diet induced increases in serum, liver and kidney cholesterol level by 14.4, 6.4 and 11.1% respectively. The leaves have also been found to posses antitumor, antipyretic, antiepileptic, antihypertensive and antioxidant properties [14]. In certain case of diabetes, moringa can also be used to stabilize sugar level [15].

The seed of *Moringa oleifera* has been good antioxidants, able to reduce oxidative damage associated with aging and cancer [16].

2. MATERIALS AND METHODS

2.1 Sample Collection

Moringa oleifera seeds were collected from the agricultural farm of ESUT Agbani, Enugu state. They were authenticated by Prof. Olive Ngwu of Department of Agronomy and Crop Science, Enugu State University of Science and Technology, Enugu.

2.2 Sample Preparation

The collected fruits were opened to release seeds embedded inside the pods and were conveyed using a black polythene bag to the laboratory. Moringa seeds were cleaned, sun dried for seven days so as to minimize the moisture content. The dry seeds were grounded into powdery form using a grinding mill and packaged in an air tight plastic container until ready for n-hexane extraction [17].

2.3 Extraction Procedure

About 7 g of the sample were poured into soxhlet extraction apparatus fitted with a1-L round bottom flask and a condenser. The extraction was executed using 0.6 L of n-hexane at 70°C for 4-5 hours until a desired amount was achieved. After which the oil was obtained by evaporating the solvent using a water bath at 60°C. The sample was weighed and the difference was calculated as the weight of the sample before extraction – the weight of the sample after extraction multiply by 100, divided by the initial weight of the sample to give the percentage oil yield. The oil was stored in a cooled place for further analysis without further treatment.

2.4 Proximate Analysis

Proximate composition of seed samples were analyzed according to method described by AOAC [18]. The proximate analyses carried out involves moisture content, ash content, crude fiber, protein content, fats and oil, carbohydrate.

2.5 Physicochemical Analysis

Specific gravity: An empty specific gravity bottle was weighed and recorded as w_1.Then another specific gravity bottle was filled with distilled H_2O and kept in a water bath at 500°c for 40 minutes,

then weight was taken and recorded as w_2. After drying, the bottle was filled with the extracted oil and the weight was recorded as w_3. The process was repeated to get the final weight.

$$\text{SG of oil} = \frac{w_2 - w_1}{w_3 - w_1}$$

w_1= weight of empty SG bottle
w_2 = weight of SG bottle + water
w_3 = weight of SG bottles + oil

Free Fatty Acid (FFA) value: 0.5 ml of sample was weighed into a 250 ml conical flask using a pipette. 20 ml of ethanol was added into the conical flask containing the sample with constant stirring. Then three drops of phenolphthalein indicator was added and titrated with 0.1N NaOH solution for 20s until it changed faint pink with thorough shaking.

$$\text{Free Fatty Acid value} = \frac{TV \times N \times 56.1}{\text{Weight of sample}}$$

Where TV = Titre value N= normality of titrant, 56.1 = acid constant

Acid value: 0.5 ml of oil was weighed using a pipette into a conical flask. Three drops of phenolphthalein indicator and 20 ml of ethanol were added into the conical flask. The mixtures were titrated with 0.1N NaOH solution until a pink coloration was obtained.

$$\text{Acid value} = \frac{TV \times 0.0282 \times 100}{\text{Weight of sample}}$$

Saponification value: 3 g of the sample was weighed into 200 ml conical flask. 40 ml of alcoholic potassium hydroxide was added into the container containing the sample with constant stirring. The resulting mixture was refluxed for an hour thirty minutes until the entire oil dissolved. Two drops of indicator was added and titrated against 0.5 N HCl with continuous shaking until the pink color changes to colorless.

$$\textbf{Saponification value} = \frac{N \times M \times (tv_2 - tv_1)}{\text{Weight of sample}}$$

Where $tv_2 - tv_1$ = difference in titre value of sample and blank
N = Normality of HCL, M = molecular weight of KOH

Iodine value: 0.5 g of sample was weighed into a conical flask and 15 ml of chloroform was added. 25 ml of wiji's reagent was added and the mixture was stirred thoroughly using a glass rod. The flask was covered tightly and placed in the dark corner for 1 hour. 40 ml of 15 % potassium iodide and 100 ml of distilled water was added and shaked vigorously. The mixtures were titrated against 0.1N solution of sodium thiosulphate until the reddish solution almost disappears. Small amount of starch indicator was added and titrated until the blue black colouration completely disappeared after vigorous shaking.

Iodine value = $\frac{TV_2 - TV_1 \times N \times 12.69}{\text{weight of sample}}$ Where 12.69 = constant for iodine value, N = Normality of titre, TV_2 = titre value of blank, TV_1 = titre value of the sample

Peroxide value: 0.5 g of oil sample was weighed into a conical flask. 1 g of potassium iodide and 20 ml of mixture of DMSO and acetic acid was added into the conical flask containing the oil sample. It was heated for 4 min. 15 ml of 3% potassium iodide was added and was titrated with 0.02 sodium thiosulphate until yellow color almost disappeared. 0.5 ml starch indicator was added and shaked vigorously and was titrated carefully until blue color disappears.

$$\text{Peroxide value} = \frac{S \times N \times 1000}{\text{Weight of sample}}$$

Where S = titre value, N = normality of titrant

Viscosity: The viscosity was determined using Brookfield viscometer (LVII, Brookfield Inc., USA) using spindle no.5 with shear rate 100 rev/ min.

Refractive index: The Refractive Index of oil was determined using an Abbe's refractometer. Two-three drops of sample applied and reading was recorded.

3. RESULTS AND DISCUSSION

The 38.8% oil percentage yield agrees with 35-40% yield reported by Solade [19]. The value of the 62.84% for saponification and 62.45% for iodine value was in agreement with Orhevba et al. [20]. The crude fat value of 29.65% and 10.50% for viscosity is contrary to what Nzikou et al. [21] and Olaleye et al. [22] reported. The 4.05% for ash and 52.30% for carbohydrate obtained agrees with 4.2% and 56.42% reported by Nzikou et al. [21] and Orhevba et al. [20] respectively. 7.64% value for moisture content, 1.1% for specific gravity, 4.10% for peroxide value, 5.95% for acid value and 1.46% for refractive index was in agreement with 7.51% for

moisture, 0.896 for specific gravity, 5.00 for peroxide, 6.35 for acid value and 1.457 for refractive index as reported by Olaleye et al. [2]. Orhevba et al. [20] reported 8.27% free fatty acid and Nzikou et al. [21] reported 37.6% for crude protein.

Table 1. Shows the result of proximate analysis of *Moringa oleiferia* oil Proximate analysis result

Parameters	Values in (%)
Moisture	7.64
Crude Protein	34.92
Oil	38.84
Crude fat	29.65
Ash	4.05
Carbohydrate	52.30

Table 2. Shows the result of physico-chemical analysis of moringa oleiferia oil Physic-chemical analysis result

Parameters	Values in (%)
Iodine value (I_2/100g)	62.45
Specific gravity	1.00
Free fatty acid (MgKOH/g)	9.84
Saponification value (MgKOH/g)	162.84
Peroxide value (MgEq/Kg)	4.10
Refractive index	1.46
Viscosity (MM^2/S)	10.50
Acid Value (mg/g)	5.95

4. CONCLUSION

The extracted oil from *moringa oleifera* seed could be utilized successfully as a source of edible oil for human consumption based on its high saturated oil and also be used for other industrial applications.

ACKNOWLEDGEMENTS

Our profound gratitude goes all those who contributed to the success of this research and presentation.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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Peer-review history:
The peer review history for this paper can be accessed here:
http://www.sdiarticle3.com/review-history/46340