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Article in *Journal of the American Oil Chemists' Society* · May 2018

DOI: 10.1002/aocs.12059

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# Physicochemical Characterization, Fatty Acid And Tocopherol Content of *Moringa ovalifolia* (African *Moringa*) Oil From Namibia

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Received: 20 April 2017 / Revised: 15 February 2018 / Accepted: 3 March 2018  
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**Abstract** *Moringa ovalifolia* tree is indigenous to Namibia and is a multipurpose tree whose leaves, pods, fruits, and flowers are edible. The tree is well adapted to the harsh climatic conditions of the region and easily cultivated. In this paper, we investigate the physicochemical characteristics of the oil extracted from *M. ovalifolia* seeds collected from Remhoogte Farm, Khomas region, Namibia. The oil yield was  $34.86 \pm 2.47\%$ . The oil contained a high level of mono-unsaturated fatty acids, with oleic acid (18:1) being the dominant one. The oil had high levels of tocopherol, with a total of  $44.56 \text{ mg } 100 \text{ g}^{-1}$  of oil, comprising  $\alpha$ -tocopherol ( $33.94 \text{ mg } 100 \text{ g}^{-1}$ ),  $\beta$ -tocopherol ( $6.64 \text{ mg } 100 \text{ g}^{-1}$ ), and

$\delta$ -tocopherol ( $3.98 \text{ mg } 100 \text{ g}^{-1}$ ). High levels of stigmasterol ( $142.41 \text{ mg } 100 \text{ g}^{-1}$ ) and  $\beta$ -sitosterol ( $330.70 \text{ mg } 100 \text{ g}^{-1}$ ) were also detected. The seed oil exhibited good-quality characteristics, making it a useful new seed oil to be considered for food and nonfood applications.

**Keywords** Namibia · *Moringa ovalifolia* · Seed oil · Physicochemical characterization · Fatty acids · Oleic acid

*J Am Oil Chem Soc* (2018) 95: 1163–1170.

**Electronic supplementary material** The online version of this article (doi:10.1002/aocs.12059) contains supplementary material, which is available to authorized users.

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## Introduction

*Moringa ovalifolia* Dinter & A. Berger (African *Moringa*) is a species from the family of Moringaceae, the *Moringa* family, and is a succulent, deciduous tree about 7 m in height, with a characteristic swollen stem (Fig. 1a, b). The *Moringa* tree bark is recognized by its pale whitish-gray and shiny appearance, with its large light green leaves reaching a length of up to 60 cm (Palgrave, 1983). The flowers, growing up to 3 mm in size, appear from November to February, with the edible, pod-like, three-sided fruits appearing from May to November (Palgrave, 1983). The ripe fruits split open into three valves, releasing the characteristic winged, membranous seeds (Curtis & Mannheimer, 2005; Palgrave, 1983). The *Moringa* tree is endemic to the semidesert areas of Namibia and southwestern Angola (Makita et al., 2017) and is widespread in western Namibia, growing more commonly on hill slopes and rocky outcrops. The *Moringa* tree can easily be cultivated from its seeds (Curtis & Mannheimer, 2005).

In Namibia, the tree is generally known as the “phantom tree” and locally known as *Omuhete* (Oshiwambo

language), *Omutindi* (Otjiherero language), and *Khaos* (Khoekhoegowab language) (Curtis & Mannheimer, 2005). *Moringa* trees are multipurpose crops (Bennett et al., 2003) that have nutritional, industrial, and medicinal potential (Makita, Chimuka, Steenkamp, Cukrowska, & Madala, 2016). The leaves, pods, fruits, and flowers are edible, making *Moringa* a useful food source (Manzoor, Anwar, Iqbal, & Bhangar, 2007). The most widely studied species of the Moringaceae family is *Moringa oleifera*, which is commercially cultivated and for which the composition of possible health-promoting secondary metabolites has been investigated (Bennett et al., 2003; Förster et al., 2015; Tumer, Rojas-Silva, Poulev, Raskin, & Waterman, 2015). Other species of *Moringa* that have been investigated, although to a lesser extent, are species such as *M. stenopetala* (Bennett et al., 2003), *M. drouhardii* (Kouteu, Baréa, Barouh, Blinand, & Villeneuve, 2016), and *M. peregrina* (Abu-Tarboush, 1998). Various plant parts of the *M. ovalifolia* species have also been reported to contain a wide array of flavonoid compounds exhibiting nutraceutical properties (Makita et al., 2016); in particular, the leaf extracts have been shown to have pharmacological activities (Makita et al., 2017).

The use of seed oils from natural sources in the pharmaceutical, food, and cosmetic industry is becoming a growing trend (Kleiman, Ashley, & Brown, 2008). This is due to the production of seed oils that has been increasing steadily over the last few decades to keep up with the demand for food and nonfood applications (Mitei, Ngila, Yeboah, Wessjohann, & Schmidt, 2008). The ever-growing human population and its increasing need for food supply necessitate the search for and promotion of other sources of edible oils (Nadeem & Imran, 2016) as part of the endeavor to ensure food security for the world's population.

The *Moringaceae* family has been reported to produce good-quality seed oils with oil compositions ranging between 35% and 45% (Bhutada, Jadhav, Pinjari, Nemade, & Jain, 2016; Kleiman et al., 2008; Lalas & Tsaknis, 2002; Tsaknis, Lalas, Gergis, Dourtoglou, & Spiliotis, 1999). The seed oils from various species of the *Moringa* genus such as *M. oleifera* from Kenya (Lalas & Tsaknis, 2002) and India (Bhutada et al., 2016), *M. peregrina* from Saudi Arabia (Tsaknis, 1998), and *M. pterygosperma* (Kleiman et al., 2008), among others, have been studied and characterized. The *M. oleifera* seed oil, also called "Ben oil" (Bhutada et al., 2016) because of its relatively high behenic acid (22:0) content (6–7%) (Bhutada et al., 2016; Lalas & Tsaknis, 2002; Tsaknis et al., 1999), has been the most widely studied and has been reported to possess various nutritional and medicinal properties (Fahey, 2005; Nadeem & Imran, 2016). The seed oil of *Moringa* species has been shown to exhibit high stability toward oxidation and is a good emollient (Kleiman et al., 2008;

LePoole, 1996), being composed of a relatively high number of long-chain fatty acids and monounsaturated fatty acids. The demand for monounsaturated oils is increasing globally because these oils have excellent oxidative stability and are beneficial to health (Corbett, 2003).

To our knowledge, this is the first detailed physicochemical characterization of *M. ovalifolia* seed oil, which has been sourced from the Khomas region of Namibia. In the search for different sources and varieties of unique seed oils, which could potentially contribute to the economy of the natural products sector of Namibia, we investigated the physicochemical characteristics of *M. ovalifolia* seed oil as a potential contributor to the natural products sector.

## Materials and Methods

### Oil Extraction From *Moringa ovalifolia* Seeds

Ripe seeds were collected during September 2016 from *M. ovalifolia* trees on Remhoogte Farm based in the Khomas region of southwestern Namibia. The seeds of *M. ovalifolia* used for oil extraction were collected from several trees within the same collection site. The species was identified by a botanist at the Department of Biological Sciences, University of Namibia, and verification of the scientific name was done by comparing specimens with those reported in the literature (Palgrave, 1983). The seed coat was carefully removed manually from the *M. ovalifolia* seeds (Fig. 1c) to obtain the oil-bearing seed kernel (Fig. 1d). The dehulled seeds were then ground on "fine setting" (typically 0.20–0.38 mm particle size) in a coffee grinder (Krupps Burr Grinder GVX2, KRUPS, Solingen, Germany). The finely ground seeds were then stored at  $-20\text{ }^{\circ}\text{C}$  until further use. The ground seeds (20 g) were added into a cellulose thimble and extracted with 250 mL of *n*-hexane (Merck, Darmstadt, Germany) for 6 hours at  $60\text{ }^{\circ}\text{C}$  in a Soxhlet apparatus (Merck KGaA, Darmstadt, Germany). The solvent was removed under vacuum at  $40\text{ }^{\circ}\text{C}$  using a rotary evaporator (Heidolph, Schwabach, Germany). The samples were stored in the dark at  $4\text{ }^{\circ}\text{C}$  until further analysis. The oil yield was calculated gravimetrically on dry weight basis as an average of three extractions using the following formula:

$$\text{Yield}(\%) = \left[ \frac{\text{Weight of the extracted oil(g)}}{\text{Weight of seed sample(g)}} \right] \times 100\%$$

### Determination of Physicochemical Parameters

The chemical characteristics such as saponification value, acid value, and iodine value of the *M. ovalifolia* seed oil were determined according to the Association of Official Analytical Chemists (AOAC) official methods (1998). The *p*-anisidine value was determined according to the American Oil

Chemists Society (AOCS) official method Cd 18-90 (1993). The ferric thiocyanate method according to Uluata and Özdemir (2012) was used to determine the peroxide values. These values were expressed in milliequivalent O<sub>2</sub> per kg of oil. The refractive indices of the oil samples were determined using an Abbe refractometer (K7135; MRC, Holon, Israel) at 25 °C, and the specific gravity was determined according to the AOAC official method No. 40.1.08 (1990). All analyses were carried out in triplicate with the means and SD reported.

### Nuclear Magnetic Resonance Spectral Analysis

<sup>1</sup>H and <sup>13</sup>C nuclear magnetic resonance (NMR) spectral analysis of the seed oil was performed using a Bruker Avance 400 MHz spectrometer (Bruker Biospin, Rheinstetten, Germany). Deuterated chloroform (CDCl<sub>3</sub>) (Merck, Darmstadt, Germany) was used as solvent at 25 °C. CDCl<sub>3</sub> was used as the reference with signals at 77.0 ppm for <sup>13</sup>C NMR and at 7.24 ppm for <sup>1</sup>H NMR. Data from the NMR spectral analysis were reported as chemical shifts ( $\delta$ ) in ppm.

### Composition of Fatty Acids

The fatty acids were converted to their fatty acid methyl esters (FAME) according to Yang, Pan, Zeng, Shupe, and Hse (2013). About 50 mg of oil was dissolved in 5 mL of hexane, and then 0.5 mL of 2 M potassium hydroxide in methanol solution was added. The tube was then agitated vigorously for 5 min and then centrifuged at 3000 rpm (Eppendorf Centrifuge 5810R; Hamburg, Germany) for 15 min. The top hexane layer containing the FAME was then removed into a capped vial and briefly stored at -20 °C until further analysis (Yang et al., 2013). The FAME were then analyzed in triplicate using a gas chromatograph, model 7820A, coupled with a mass spectrometry unit, model 5977E MSD (Agilent Technologies, Palo Alto, CA, USA). The FAME were separated using an Agilent capillary column HP5 MS (30 m  $\times$  0.25 mm, 0.25  $\mu$ m). Helium carrier gas flow rate of 1.50 mL min<sup>-1</sup> was used with a split ratio of 20:1. The injection temperature was 250 °C. The initial temperature was 40 °C, held for 8 min, ramp 1: 10–220 °C for 5 min and ramp 2: 20–300 °C for 10 min. The composition of FAME was determined as percentages of the total peak areas of methyl esters contained in the sample.

### Tocopherols and Major Sterols Analysis

The oil samples were derivatized and analyzed according to Du and Ahn (2002) using about 100 mg of seed oil. About 100 mg of oil was extracted with 10 mL of the saponification reagent (ethanol 33%:KOH [w/v] 20%:ascorbic acid—94:6:0.5). One hundred microliters of the internal standard, 5 $\alpha$ -cholestane, at a concentration of 10 ppm was added. The

mixture was then briefly vortexed and incubated for 60 min at 50 °C and then cooled for 10 min. Deionized water (5 mL) and hexane (5 mL) were added. To allow the sample to separate into phases, it was vortexed and left for 15 hours. One thousand microliters of the supernatant was then dried and reconstituted with 200  $\mu$ L pyridine and 100  $\mu$ L BSTFA (bis(trimethylsilyl) trifluoroacetamide) with 1% TMCS (trimethylsilyl chloride). The mixture was vortexed and then derivatized by incubating for 1 hour at 50 °C. The tocopherols and major sterols, with an injection volume of 1  $\mu$ L, were separated and identified using an Agilent 6890 N gas chromatograph coupled to an Agilent 5975 MS detector with a Zebron™ ZB-MultiResidue (30 m, 0.25 mm ID, 0.25  $\mu$ m) column. Helium carrier gas flow rate was 1.2 mL min<sup>-1</sup> and the injector temperature maintained at 200 °C in a splitless mode. The oven temperature was maintained at 100 °C for 2 min, ramped at 15 °C min<sup>-1</sup> to 180 °C, held for 0 min, ramped at 5 °C min<sup>-1</sup> to 250 °C, held for 3 min, and finally at 20 °C min<sup>-1</sup> to 320 °C, and held for 12 min. The mass spectral data was recorded on a mass-selective detector (MSD) operated in full-scan mode (35–600 *m/z*), with both the ion source and quadrupole temperatures maintained at 240 and 150 °C, respectively.

### Statistical Analysis

All experiments were carried out in triplicate unless otherwise stated with means and SD calculated using the IBM® SPSS® Statistics Version 24 Software (IBM Corporation, New York, United States).

## Results and Discussion

The Remhoogte Farm is situated in the southwestern part of Namibia and is part of the Khomas region. The farm is home to a number of *M. ovalifolia* trees growing among the rocky outcrops and valleys. The average annual rainfall for this area, received during the summer months (Namibia Ministry of Health and Social Services, 2013), is between 100 and 200 mm, which can be extremely varying from year to year (Namibia Statistics Agency, 2013). January and February are the hottest months of the year with temperatures ranging between 9 and 30 °C and above (Namibia Statistics Agency, 2013). Between May and September, temperatures can fluctuate between -6 and 10 °C at night and 20 °C in the day, with the occasional frost occurring during nights (Namibia Ministry of Health and Social Services, 2013).

### Physicochemical Parameters

The physical and chemical properties of *M. ovalifolia* seed oil from Namibia are presented in Table 1. The oil yield

**Table 1** Physicochemical characteristics of *M. ovalifolia* (African *Moringa*) seed oil

Parameter	Quantity
Oil yield (%)	34.86 ± 2.47
Saponification value (mg of KOH g <sup>-1</sup> of oil)	169.69 ± 2.38
Acid value (mg KOH g <sup>-1</sup> of oil)	2.12 ± 0.15
Peroxide value (mequiv kg <sup>-1</sup> )	2.55 ± 0.28
<i>p</i> -Anisidine value	0.56 ± 0.22
Iodine value (g of I <sub>2</sub> 100 g <sup>-1</sup> of oil)	62.30 ± 0.46
Specific gravity (20 °C)	0.905 ± 0.00
Refractive index (25 °C)	1.4630 ± 0.001
α-Tocopherol (mg 100 g <sup>-1</sup> of oil)	33.94
β-Tocopherol (mg 100 g <sup>-1</sup> of oil)	6.64
γ-Tocopherol (mg 100 g <sup>-1</sup> of oil)	nd
δ-Tocopherol (mg 100 g <sup>-1</sup> of oil)	3.98
Total tocopherol (mg 100 g <sup>-1</sup> of oil)	44.56
Stigmasterol (mg 100 g <sup>-1</sup> of oil)	142.41
β-Sitosterol (mg 100 g <sup>-1</sup> of oil)	330.70

nd, not detected.

obtained after Soxhlet (*n*-hexane) extraction of *M. ovalifolia* seeds was 34.86 ± 2.47%. The hexane extraction yielded a golden yellow oil that was liquid at room temperature (Fig. S2). The oil content of *M. ovalifolia* was low compared to the seed oil obtained from *M. concanensis* (38.82%) from Pakistan (Manzoor et al., 2007), *M. peregrina* (49.8%) from Saudi Arabia (Tsaknis, 1998), and *M. oleifera* seeds variety Periyakulam 1 (38.3%) from India (Lalas & Tsaknis, 2002), but similar to the oil content of *M. oleifera* (35.70%) from Kenya (Tsaknis et al., 1999) and *M. oleifera* (35.30%) from Malawi (Tsaknis, Lalas, Gergis, & Spiliotis, 1998). Variations in geological and environmental conditions may affect the oil yield and physicochemical parameters among different *Moringa* species (Manzoor et al., 2007; Ogunsina et al., 1994).

The saponification value (169.69 ± 2.38 mg of KOH g<sup>-1</sup> of oil) of the *M. ovalifolia* seed oil was lower than that of the *M. concanensis* seed oil (179 mg of KOH g<sup>-1</sup> of oil) from Pakistan (Manzoor et al., 2007), *M. peregrina* seed oil (185 mg of KOH g<sup>-1</sup> of oil) from Saudi Arabia (Tsaknis, 1998), *M. oleifera* var. Periyakulam (188.36 mg of KOH g<sup>-1</sup> of oil) from India (Lalas & Tsaknis, 2002), and olive oil (184–196 mg of KOH g<sup>-1</sup> of oil) (Gunstone, Harwood, & Dijkstra, 2007). However, the saponification value was found to be higher than that in the seed oil from *M. oleifera* (160.62 mg of KOH g<sup>-1</sup> of oil) from Mexico (Sánchez-Machado et al., 2015). The saponification value (169.69 ± 2.38 mg of KOH g<sup>-1</sup> of oil) of the *M. ovalifolia* seed oil was found to be in the range of that of the high erucic rapeseed oil (168–181 mg of KOH g<sup>-1</sup> of oil) (Rossell, 1991) extracted from the *Brassica napus* plant.

The acid value (2.12 ± 0.15 mg of KOH g<sup>-1</sup> of oil), reflecting the total acidity, and the peroxide value (2.55 ± 0.28 mequiv kg<sup>-1</sup> of oil), reflecting the presence of primary oxidation compounds, were below the maximum acceptable levels of the standards described for edible oils of the CODEX STAN 210-1999 (CODEX STAN, 2011), whereby an acceptable acid value level is to be <4.0 mg of KOH g<sup>-1</sup> of oil and an acceptable peroxide value is to be <15 mequiv kg<sup>-1</sup> of oil (CODEX STAN, 2011). The *p*-anisidine value (0.56 ± 0.22), reflecting the presence of secondary oxidation compounds, and the low acid and peroxide values of the *M. ovalifolia* seed oil indicated that this oil is of good quality.

The iodine value (62.30 ± 0.46 g of I<sub>2</sub> 100 g<sup>-1</sup> of oil), reflecting the level of unsaturation in the oil (Wrolstad et al., 2005), was lower than that (67.00 g of I<sub>2</sub> 100 g<sup>-1</sup> of oil) reported for *M. concanensis* from Pakistan (Manzoor et al., 2007), *M. peregrina* (69.6 g of I<sub>2</sub> 100 g<sup>-1</sup> of oil) from Saudi Arabia (Tsaknis, 1998), and *M. oleifera* var. Periyakulam (65.58 of I<sub>2</sub> 100 g<sup>-1</sup> of oil) from India (Lalas & Tsaknis, 2002).

The specific gravity (0.905) of the *M. ovalifolia* seed oil was comparable to those of *M. peregrina* seed oil (0.9095) from Saudi Arabia (Somali, Bajneid, & Al-Fhaimani, 1984) and *M. oleifera* (0.90) from India (Ogunsina et al., 1994). The refractive index (1.4630) of the *M. ovalifolia* seed oil was comparable to those of *M. concanensis* seed oil (1.4648) from Pakistan (6) and *M. peregrina* seed oil (1.460) from Saudi Arabia (Tsaknis, 1998), but was different from that of *M. oleifera* var. Periyakulam (1.4507) from India (Lalas & Tsaknis, 2002). The refractive index of *M. ovalifolia* seed oil was within the range of those of major vegetable oils such as groundnut (1.460–1.465), mustard seed (1.461–1.469), and almond (1.462–1.465) oil (Rossell, 1991).

### Tocopherols and Major Sterols Compositions

The compositions of tocopherol and major sterols (stigmasterol and β-sitosterol) of *M. ovalifolia* seed oil from Namibia are presented in Table 1. The total tocopherol content of the *M. ovalifolia* seed oil was 44.56 mg 100 g<sup>-1</sup> of oil, comprising α-tocopherol (33.94 mg 100 g<sup>-1</sup> of oil), β-tocopherol (6.64 mg 100 g<sup>-1</sup> of oil), and δ-tocopherol (3.98 mg 100 g<sup>-1</sup> of oil). γ-Tocopherol was not detected in *M. ovalifolia* seed oil, but was detected in the seed oils from *M. concanensis* from Pakistan (Manzoor et al., 2007) and *M. oleifera* var. Periyakulam from India (Lalas & Tsaknis, 2002), whereas the β-tocopherol was not detected in these seed oils. The total tocopherol composition for *M. ovalifolia* seed oil was higher than that reported for *M. concanensis* seed oil (115.24 mg kg<sup>-1</sup> of oil) from Pakistan (Manzoor et al., 2007), *M. oleifera* var.

Periyakulam (35.36 mg kg<sup>-1</sup> of oil) from India (Lalas & Tsaknis, 2002), *M. oleifera* (255.44 mg kg<sup>-1</sup> of oil) from Malawi (Tsaknis et al., 1998), and some common vegetable oils such as groundnut (37 mg 100 g<sup>-1</sup> of oil), coconut (1 mg 100 g<sup>-1</sup> of oil), and olive (22 mg 100 g<sup>-1</sup> of oil) oil (Gunstone et al., 2007).  $\alpha$ -Tocopherol was the dominant tocopherol, which is considered the most active ingredient with regard to its vitamin E potency (O'Brian, 2009). Tocopherols are potent natural antioxidants and efficiently prevent lipid peroxidation (Nasri et al., 2012) by imparting stability on free radicals and improving the quality of the oil (O'Brian, 2009). The comparatively high content of tocopherol in *M. ovalifolia* seed oil among the species of *Moringa* would suggest that this oil has excellent stability toward oxidation (Manzoor et al., 2007). The stigmaterol content of *M. ovalifolia* seed oil (142.41 mg 100 g<sup>-1</sup> of oil) was higher than that reported for soybean (57.7 mg 100 g<sup>-1</sup> of oil), sunflower (33.7 mg 100 g<sup>-1</sup> of oil), corn (67.7 mg 100 g<sup>-1</sup> of oil) oil (31), and *M. oleifera* (23.10 mg 100 g<sup>-1</sup> of oil) seed oil from Malawi (Tsaknis et al., 1998). The  $\beta$ -sitosterol content of *M. ovalifolia* seed oil (330.70 mg 100 g<sup>-1</sup> of oil) was higher than that of sunflower (265.3 mg 100 g<sup>-1</sup> of oil), olive (130.3 mg 100 g<sup>-1</sup> of oil), and *M. oleifera* (45.58 mg 100 g<sup>-1</sup> of oil) seed oil from Malawi (Tsaknis et al., 1998), but was lower than that of cotton (401.8 mg 100 g<sup>-1</sup> of oil) and rapeseed (419.8 mg 100 g<sup>-1</sup> of oil) oil (Gunstone et al., 2007). Stigmaterol has been shown to have antiosteoarthritic properties (Gabay et al., 2010). The presence of  $\beta$ -sitosterol, a main dietary phytosterol (National Center for Biotechnology Information, 2017), has been shown to impart antifungal, antiinflammatory, antiviral (Malini & Vanithakumari, 1990), anticarcinogenic, and antiatherogenic properties (National Center for Biotechnology Information, 2017).

### NMR Spectral Analysis

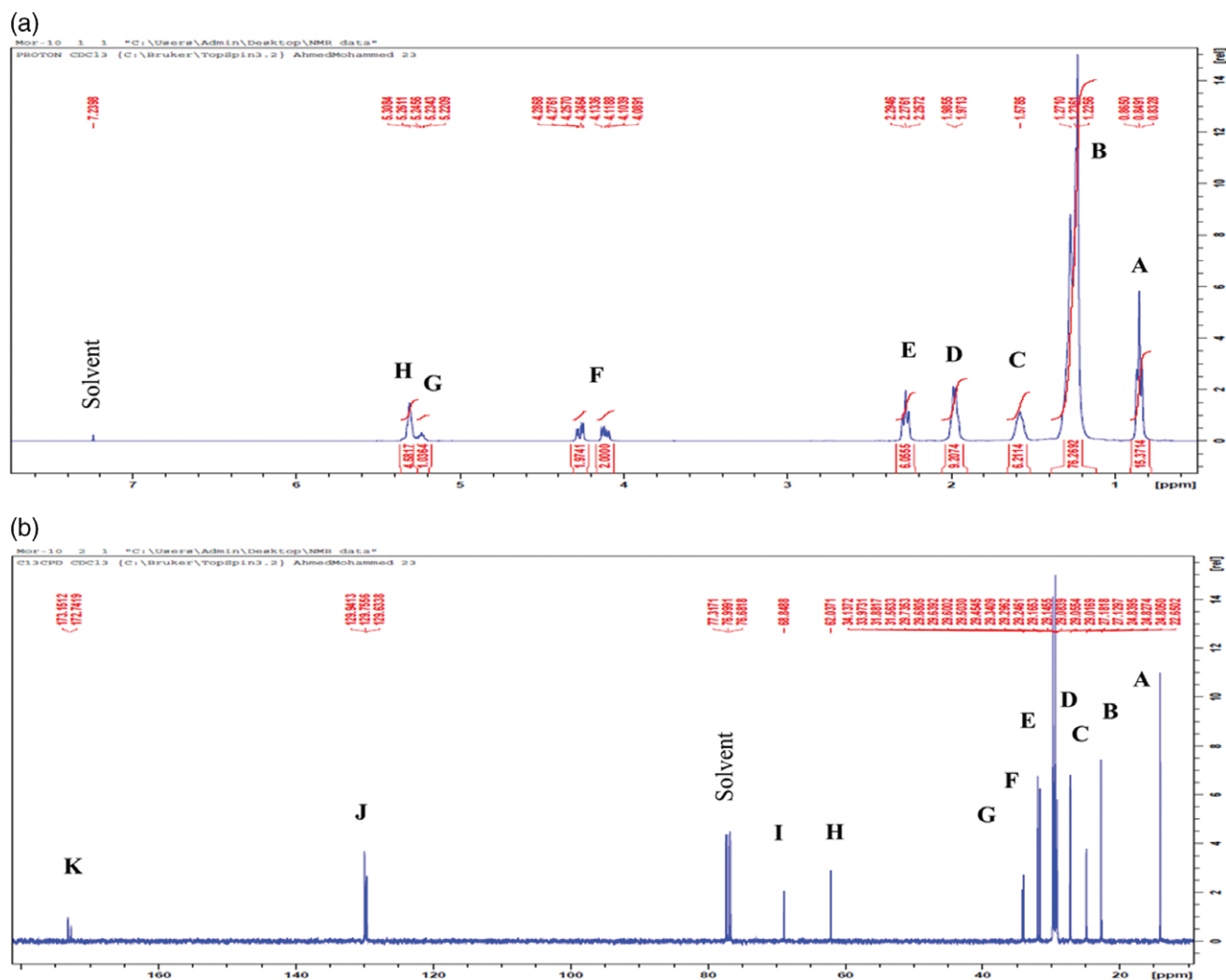
The <sup>1</sup>H and <sup>13</sup>C NMR spectra are presented in Fig. 1. The major regions, indicated by letters on the spectra, were assigned according to Sacchi, Addeo, and Paolillo (1997), Siyanbola et al. (2015), and Timilsena, Vongsvivut, Adhikari, and Adhikari (2017). The <sup>1</sup>H NMR spectra (Fig. 1a) of *M. ovalifolia* seed oil show signals at 0.83 ppm (A), indicating the presence of terminal methyl protons ( $-CH_3$ ). The signals in the region 1.23–1.27 ppm (B) account for the presence of internal protons ( $-CH_2$ ). The presence of unsaturated fatty acids in the seed oil is confirmed by the signals observed at 1.97–1.99 ppm (D) and 5.31 ppm (H). The presence of oleyl acyl (and other fatty acids) chains is confirmed by the signals arising from the methylene groups,  $-OCO-CH_2-CH_2-$  at 1.58 ppm (C) and  $-OCO-CH_2-$  at 2.26–2.29 ppm (E). The signal observed at 4.09 and 4.29 ppm (F) arise from the 1- and 3-glycerol

$-CH_2-$  protons. The methylene proton signal of the glycerol backbone is observed at 5.22–5.26 ppm (G), referring to the CHOCOR group.

The <sup>13</sup>C NMR spectra (Fig. 1b) of *M. ovalifolia* seed oil show signals at 22.7 ppm (A), 24.8 ppm (B), 27.1–27.2 ppm (C), and around 29.0 ppm (D), indicating the presence of the CH<sub>2</sub>,  $\omega$ 2; C3; C8–C11; and the (CH<sub>2</sub>)<sub>n</sub> (acyl chains) in the *M. ovalifolia* seed oil, respectively. The presence of  $\omega$ 3 (saturated, *n*-9 and *n*-6 acids); C2, *sn*-1,3 (acyl chains); and C2, *sn*-2 (acyl chains) was observed at 31.9 ppm (E), 33.9 ppm (F), and 34.1 ppm (G), respectively. The presence of the CH<sub>2</sub>O-, *sn*-1,3 and the CHO-, *sn*-2 of the triacylglycerol was observed at 62.0 ppm (H) and 68.9 ppm (I), respectively. The signals observed in the region 129.6–129.9 ppm (J) indicated the presence of the cis double bond of the acyl side chain, C9, *sn*-2 and C10, *sn*-1,3. The signals observed at 172.7 and 173.2 ppm (K) indicated the presence of carboxylic group of the fatty acid esters C1, *sn*-2 and C1, *sn*-1,3.

### Fatty Acid Composition

The fatty acid composition of *M. ovalifolia* seed oil from Namibia is presented in Table 2. The saturated fatty acids with a total of 39% were palmitic acid (9.58%), stearic acid (12.17%), arachidic acid (7.46%), behenic acid (7.75%), and lignoceric acid (2.22%). The unsaturated fatty acids with a total of 60% were palmitoleic acid (0.61%), oleic acid (55.22%, the dominant fatty acid), and gondoic acid (4.02%). The *M. ovalifolia* seed oil is unique in that it contains a number of long-chain fatty acids, namely arachidic (20:0), behenic (22:0), and lignoceric (24:0). This characteristic is common among the seed oils of the *Moringa* species (Kleiman et al., 2008; Manzoor et al., 2007), with the oleic acid, a monounsaturated omega-9 fatty acid, being the dominant fatty acid. Seed oils containing between 50% and 80% oleic acid (18:1) are regarded as “high-oleic-acid-containing” seed oils (Corbett, 2003) and are known to have cholesterol-lowering abilities (Lokuruka, 2007). Behenic acid (22:0) was found in high concentration and was within the reported values for the common *M. oleifera* (Bhutada et al., 2016; Lalas & Tsaknis, 2002; Tsaknis et al., 1999), making it comparatively useful for various industrial applications. The stearic acid (18:0) contents of *Moringa* species have been reported to vary between 3% and 9% (Kleiman et al., 2008; Manzoor et al., 2007). The stearic acid (18:0) content of *M. ovalifolia* compares more closely with that of *M. stenopetala* (Kleiman et al., 2008) at 9.9%. Oils with a relatively high content of stearic acid content compared to conventional oils are recommended for industrial uses such as cosmetic and biodiesel applications. The total composition of unsaturated fatty acids as



**Fig. 1** (a) <sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>) spectra of *M. ovalifolia* seed oil, (A)  $-\text{CH}_3$  (methyl proton), (B)  $(-\text{CH}_2)_n$  (acyl groups), (C)  $-\text{OCO}-\text{CH}_2-\text{CH}_2-$  (acyl groups), (D)  $-\text{CH}_2-\text{CH}=\text{CH}-$  (allylic protons), (E)  $-\text{OCO}-\text{CH}_2$  (acyl groups), (F)  $-\text{CH}_2\text{OCOR}$  (glycerol group), (G)  $>\text{CHOCOR}$  (glycerol group), (H)  $-\text{CH}=\text{CH}-$  (olefinic proton). (b) <sup>13</sup>C NMR (100 MHz, CDCl<sub>3</sub>) spectra of *M. ovalifolia* seed oil, (A)  $\text{CH}_2$ ,  $\omega_2$ ; (B) C3; (C) allylic (C8–C11); (D)  $(\text{CH}_2)_n$  (acyl chains); (E)  $\omega_3$ ; (F) C2, *sn*-1,3; (G) C2, *sn*-2 (acyl chains); (H)  $\text{CH}_2\text{O}-$ , *sn*-1,3; (I)  $\text{CHO}-$ , *sn*-2; (J) C9, *sn*-2; C10, *sn*-1,3; (K) C1, *sn*-2; C1, *sn*-1,3

**Table 2** Fatty acid composition of *M. ovalifolia* (African *Moringa*) seed oil

Fatty acid	Quantity (%)
Palmitoleic acid (16:1)	0.61 ± 0.10
Palmitic acid (16:0)	9.58 ± 0.21
Stearic acid (18:0)	12.17 ± 0.94
Arachidic acid (20:0)	7.46 ± 0.23
Behenic acid (22:0)	7.75 ± 0.21
Lignoceric acid (24:0)	2.22 ± 0.25
Oleic acid (18:1)	55.22 ± 2.00
Gondoic acid (11–20:1)	4.02 ± 0.25

monounsaturated fatty acids for *M. ovalifolia* was 60%, whereas the total composition of saturated fatty acid was 39%. The total composition of saturated fatty acids in the *M. ovalifolia* seed oil is higher than that of the *M. concanensis* seed oil (25.15%) from Pakistan (Manzoor et al., 2007), *M. oleifera* (19.67%) from Kenya (Tsaknis et al., 1999), and *M. oleifera* (18.3%) from India (Ogunsina et al., 1994). The content of behenic acid in *M. ovalifolia* seed oil is comparable to that of *M. concanensis* (7.09%) seed oil from Pakistan (Manzoor et al., 2007), but it is higher than that reported for *M. oleifera* (4.5%) from India (Ogunsina et al., 1994).

## Conclusion

In this study, the seed oil of *M. ovalifolia*, indigenous to Namibia, was characterized. The seeds of the *M. ovalifolia* tree can be easily cultivated, which ensures a sustainable resource for commercialization of end products such as the *M. ovalifolia* seed oil. The oil contained significant levels of monounsaturated fatty acids (omega-9) and saturated fatty acids such as behenic acid and arachidic acid, allowing this oil to be used in various food and nonfood applications. The tocopherol and major sterols content have sufficient potential for this oil to be further investigated and developed as a nutritious and health-promoting oil with downstream value-added products.

**Acknowledgments** The authors would like to thank the National Commission for Research, Science and Technology, Namibia, for providing funding under project no. Inc/0814/0018, inclusive of a PhD Fellowship Program. The authors also thank Timothy Lesch from the Chemistry Department of the University of the Western Cape (fatty acid analysis) and the Central Analytical Facility, University of Stellenbosch (tocopherol analysis) for the technical assistance provided. The authors are grateful to Dr Ezekeil Kwembeya and Dr Ndafuda Shiponeni for species identification and verification.

**Conflict of Interest** The authors declare that they have no conflicts of interest.

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