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Exploring Nutritional and Nutraceutical Potential of Wild *Moringa peregrina* (Forssk) Fiori. of Oman

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Abstract

Since time immemorial medicinal tree plants have been harvested for use in traditional remedies for treating not only common ailments but also serious diseases like diabetes and cancer besides heart diseases. In advanced countries, however, plant-based medicines are purified and modified to use with increased awareness of their medicinal value. Of 65 tree species of medicinal importance reported in Oman, Moringa peregrina (Forssk) Fiori. is often found in either hilly or mountainous areas of Oman on rocky slopes amongst stones and wadi beds and is known for its medicinal use among ethnomedicine practitioners. The present investigations explore indigenous *Moringa peregrina* of Oman in terms of physical characteristics of seed and kernel, proximate, mineral, and fatty acid composition for plausible nutritional and nutraceutical applications. The soil and plant features, physical characteristics of mature seeds, and proximate and fatty acid composition of mature and immature seeds were recorded following standard procedures. The results indicated that indigenous wild Moringa peregrina trees were 6.8m tall occupying an area of 59.16 m^2 with satisfactory chlorophyll content (57.5 at LEAF value). Mature seeds were trigonous and grayish brown and able to give 68.91% of the milky white kernel. Seeds were highest in carbohydrates (36.05%), followed by total fat (33.52%) and proteins (22.21%), and contain more than 10% each of Oleic acid, Palmitic acid, Behenic acid, Eicisenoic acid, Palmitoleic acid, and Stearic acid. *Moringa peregrina* has been suggested for use for human consumption with caution until after the exclusion of the detrimental effects of phytotoxins possibly present in its products.

Keywords: Plant features; Seeds, Chlorophyll, Physical characteristics, Proximate composition, Fatty acid composition, Moringa peregrina.

INTRODUCTION

The people are still dependent on traditional medicine and dependent on plant-based medicines in several countries in Asia, Africa, and South and Central America. On the other hand, in advanced countries, traditional plant-based medicines are integrated through purification and modification for their use in line with the changing lifestyle of the people with increased awareness of the medicinal value (Suárez *et al*., 2012, WHO 2014, Velázquez-Rosas *et al*., 2018, García-Flores *et al*., 2019). For millennia, medicinal tree plants have been harvested and used for traditional remedies to help fight some of humankind's biggest killers, such as heart disease and cancer (WHO, 2023). As medicinal knowledge of such trees is advanced, pharmaceutical industries began to cultivate these trees to extract plant-based medicines from wood, bark, roots, leaves, flowers, fruits, or seeds for the welfare of millions of people (Petrvoska, 2012; Abera, 2014; Hidayat *et al*., 2021).

The Sultanate of Oman has physiographic and climatic features of both components of Asia and Africa that harbor all the types of species from typical temperate and subtropical to tropical crops, herbs, grasses, shrubs, and tree plants, most of which are known for their ethnomedicine properties (Krippner and Staples, 2003; Miller and Morris, 1988; Neelam *et al*., 2023). Of 65 tree species of medicinal importance reported in Oman, *Moringa peregrina* (Forssk) Fiori. is often known for its medicinal use among ethnomedicine practitioners in the country, and is found in either hilly or mountainous areas of Oman on rocky slopes amongst stones and wadi beds (Miller and Moris, 1988; Ghazanfar, 2003; Oliver, 2014). It is native to the Horn of Africa, Sudan, Egypt, the Arabian Peninsula, and as far north as Syria (Dadamouny *et al*., 2016; Anonymous, 2023). The plant's seed oil is recently reported to be valuable for industrial application as a bio-coagulant for water purification and lubricant (Elsergany, 2023) and cosmetic application (Kleiman *et al*., 2008). Recently, traditional uses, pharmacological efficacy, photochemistry, and nutritional importance of Moringa peregrina have been reviewed in detail by Senthilkumar *et al*., (2018) while Moringa peregrina has been reviewed as a natural medicine for increasing immunity against COVID-19 considering its nomenclature and systematic position, morphology, ecology, and chemistry by Moustafa and Monsour (2020). Hence, considering its diverse applications, our present investigations attempt to explore indigenous Moringa peregrina of Oman in terms of physical characteristics of seed and kernel, proximate, mineral, and fatty acid composition particularly for plausible nutritional and nutraceutical applications.

MATERIALS AND METHODS

Plant Material

The seeds of Moringa peregrina were randomly collected from different trees of the mountainous habitat of Saham of Wilayat Sohar of North Batinah governorate (56.541459º E; 23.888229º N) at the elevation of 604 m above mean sea level) (Figure 1; Plate1 (a-f); Table 1). At the same time, the relevant edaphic features of locations, such as soil type, structure, critical chemical contents, soil pH and salinity, and characters of 10 randomly selected trees, were recorded. The voucher specimens were collected and submitted to the Life Science Unit of Sultan Qaboos University, where the identification of specimens of Moringa peregrina was confirmed by the taxonomist at the Sultan Qaboos University Herbarium (SPUH). The voucher material was deposited in the herbarium collection under barcode SQUH00006514. The seeds were found to be of two types, grayish brown colored, confirmed as mature with 86.67 % mean germination ranging from 80 to 85% germination in three replicates of the germination test conducted in March- April 2022 and greyish white colored, confirmed as immature with no germination (0%). They were dried in the shade before their use for the extraction of essential oil and chemical analyses.

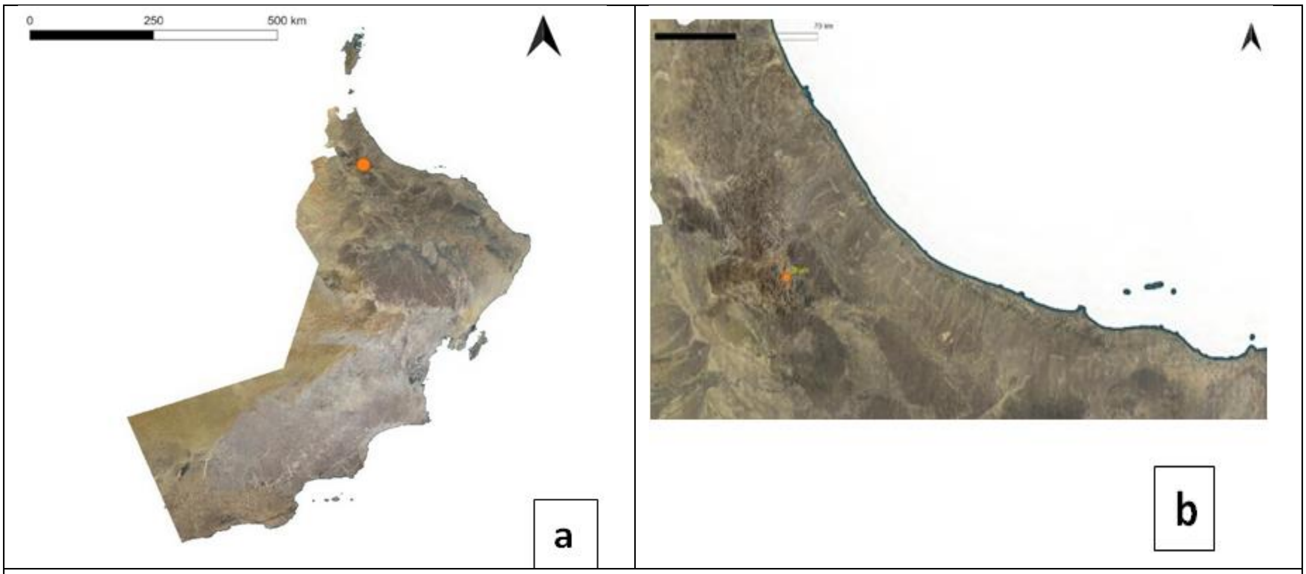


Fig 1 a & b: General map of Oman with the location, Sham, Wilayat Sohar of North Al-Batinah Governorate as green dot (a) and its zoomed image (b), from where *Moringa peregrina* seeds were collected.

Plant measurements such as plant height (m) (from the base of the plant to the highest tip of the plant) and two plant width measurements (m), one from north to south direction (N-S) (W1) and the other from east to west (E-W) (W2) were taken by the ruler. The chlorophyll contents of leaf samples were also recorded with the atLEAF CHL PLUS chlorophyll meter as atLEAF value from 0 to 99.9, showing the health status of plants; for instance, values of 35 or above refer to grades of good or better health as applied by researchers recently in different plant species (Zhu *et al*., 2012 in crop plants; Novichonok *et al*., 2016 in *Calamus diocus* and *Cleistosanthus*; Cahyo *et al*., 2020 in rubber plant *Hevea brasiliensis* Mull. Arg; Nadaf *et al*., 2023 in medicinal plants). The approximate plant area (m^2) was computed by applying a formula of L x B by multiplying plant height (H) with the mean of two plant widths (W1 and W2). In contrast, approximate plant volume was calculated with the formula H x L x B, by multiplying plant height (H), and the two plant widths (W1 and W2).

The edaphic characteristics of habitat and soil were briefed for the wadi habitat of Moringa peregrina plants and several chemical contents of soil were recorded with S1 Titan/Tracer 5/CTX equipment of Bruker (Stamatis *et al*., 2022; Nadaf *et al*., 2023). Only selected macro and microelements, relevant to plant nutrition, are presented here.

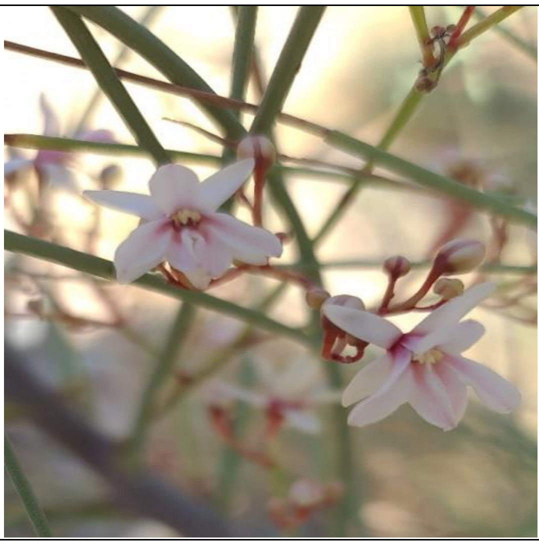
<table>
<tr><td></td><td></td></tr>
<tr><td>Plate 1a. General view of the plant in wadi habitat</td><td>Plate 1b. The branches and stems with transformed thin leaves</td></tr>
<tr><td></td><td></td></tr>
<tr><td>Plate 1c. The tree with the flush of flowers</td><td>Plate 1d. inflorescences (closer view)</td></tr>
<tr><td></td><td></td></tr>
<tr><td>Plate 1e. The tree with the flush of pods</td><td>Plate 1f. Pods with seeds (closer view)</td></tr>
</table>

Plate 1 g. The split pod with seeds

Plates 1(a-g): Images of *Moringa peregrina* plant found in the wadi habitat of the Sultanate of Oman

Physical Characteristics of mature seed and kernel

The color of the mature seed was assessed as grayish brown (2.5 Y -5/2 to 6/2) by following Munsell color charts for plant tissues (Munsell color, 1977) while that of the kernel was milky white on a visual basis. The length and width of the seed and kernel were measured with battery operated absolute digimatic Mitutoyo vernier caliper (300 mm) following operating instructions (ISO9001:2000). Weights of 50 seeds and their kernels after removing the seed coat were measured by electronic analytical weighing balance (1.000g to 100.000g). The shelling percent of kernel was computed as ((50 kernel weight /50 seed weight) x100).

Components of Proximate Analysis

The dry matter % of Moringa seeds was assessed by drying the seed samples in an oven at 70 °C, as documented in the (AOAC, 1990) guidelines. Moisture % was assessed by drying the sample in an oven at 105 °C until a constant weight was reached (AOAC, 1990). To determine the total ash, the samples were subjected to an oven at 550 °C for about 12 hours and weighed subsequently. The protein content was assessed using the micro-Kjeldahl method, however, the protein value was calculated using a nitrogen conversion factor of 6.25 according to the standards of AOAC (1990). In the crude fat analysis, approximately 5 grams of the sample were exhaustively extracted in a Soxhlet apparatus using n-hexane as the extractant, following the (AOAC, 1990) guidelines. The Soxhlet extraction method involved subjecting the sample to repeated extraction with the solvent to obtain the fat content. The percentage of the total fat content was subsequently calculated based on the weight of the extracted fat.

The carbohydrate content was determined by subtracting the percentages of moisture, ash, protein, and fats from 100% using the formula where the carbohydrate content =100%-(% moisture + % ash % + % protein + % fat). The total dietary fiber contents were assessed utilizing methodology suggested by Savage (2001) which includes specific protocols for precisely measuring the fiber components in plant samples.

A wet digestion method was followed to analyze the mineral composition of moringa seeds, both viable and non-viable ones. This procedure included the precise weighing and transfer of 1 gram of the powdered seed to a digestion vessel, where it was mixed with the concentrated nitric acid to guarantee complete mineral breakdown. After cooling, the digested solution was analyzed using spectrophotometry. Calibration curves were applied to accurately determine the concentration of minerals (Total N, Total P, Total K, and Ca) in the sample, in which the results are stated in suitable units (mg/g or %) (Kabas *et al*., 2007).

Fatty acid composition

Extraction of fatty acids using Hexane

The fatty acid composition was assessed by the method outlined in Savage *et al.*, (1999). After homogenization of 250.00 g each of viable and non-viable moringa seeds in liquid nitrogen, their contents were transferred to 50 ml graduated centrifuge tubes, subsequently to initiate a solvent extraction process. The conversion of lipids to fatty acid methyl esters (FAMEs) included trans-methylation, where the total lipid was dissolved in methanolic NaOH and then heated using a water bath. Methanolic HCl was added later, and the solution was heated again. The hexane was used as a solvent for obtaining the extracted FAMEs.

Extraction of fatty acids using Chloroform: Methanol

Here, after homogenization of 250.00 g each of viable and non-viable moringa seed in liquid nitrogen, the content was transferred to another tube to start a solvent reaction process using a mixture of chloroform and methanol in a ratio of 2:1 (chloroform: methanol). Phase separation was done by adding saline solution (NaCl) and then the mixture was vortexed resulting in the formation of two layers- an upper aqueous layer and a lower organic layer which contain the dissolved lipids.

The lower organic layer is evaporated to remove the solvents leaving the lipids contents.

The analysis of the FAME samples was conducted using gas chromatography coupled with a mass spectrometer (GC-MS QP-2010, Shimadzu, Japan), featuring an auto-sampler (AOC-5000) and flame ionization detection (FID). This analysis utilized an RTx-5MS capillary column (60 meters, 0.25 mm ID, and 0.5 µm df) from Rastek, USA. The carrier gas, helium (99.9% purity), was injected at a rate of 1 ml min^{-1}, with the pre-column pressure maintained at 112.9 kPa. The temperature of the column was initially set at 40°C for 3 minutes, with a subsequent gradual increase of 5°C per minute up to 230 °C, held for 40 minutes. The injection volume was 1µl over 67 minutes at a temperature of 230 °C. The mass spectrometer was operated in the electron compact mode with an electron energy of 70 eV while the ion sources and quadrupole were maintained at 200 °C.

Statistical Analysis

The standard errors (S.E. ±) were calculated as a part of the basic statistics from n observations wherever necessary in the study. The sample means of 6 chemical compounds commonly present in the essential oils of two seed types and other contents were compared for differences by applying the paired Student's t-test at a p<0.05 using the data analysis feature of the Excel 16 version.

RESULTS

Soil Characters

The mountainous location where Moringa peregrina seeds were collected has typical wadi habitat features down with a slope of 60^0. The site is at a higher elevation (604 m) in the mountain ranges. The plain at the foothill is composed of sandy soil with particles coarser with differential colors from black, brownish to yellowish, and at some places very dark reddish colored granite-like materials. The rocks and boulders are scattered below along with the length of the mountain range from East to West and are spread throughout with gravels and pebbles with some soil that seems covered with ephemerals of small creeping herbs, smaller herbs, and medium to larger shrubs, which are mainly restricted to inner crevices or small depressions with fine sediments. The soil of both sites is formed of more than 90% coarse sandy particles.

The soil material had 7.41 pH with an EC of 1.136 dSm^{-1} indicating the influence of dryland salinity. It has fairly higher contents of silicon (14352.6 ppm), iron (5815.1 ppm), calcium (2441.9 ppm) and magnesium (1807.3 ppm) and low contents of nitrate (140 ppm), manganese (63.8 ppm), and potassium (13.0 ppm) (Table 1).

Table 1: GPS data and main soil element contents (ppm) of the wadi location Sham, Wilayat Sohar of North Batinah governorate, from where *Moringa peregrina* seeds were collected

Element	Sham, Wilayat Sohar
Longitude (E)	56.541459
Latitude (N)	23.888229
Altitude (m)	604
	ppm
Nitrate (NO3)	140.0
Phosphorus (P)	13.6
Potassium (K)	13.0
Magnesium (Mg)	1807.3
Calcium (Ca)	2441.9
Sulphur (S)	Traces
Iron (Fe)	5815.1
Manganese (Mn)	63.8
Copper (Cu)	3.4
Silicon (Si)	143352.8
PH	7.41
EC(Electrical Conductivity) (dS^{-1})	1.136

Plant Morphological Characteristics

The mean plant characteristics of *Moringa peregrina* indicated that they were tall to the extent of 6.8 m with an N-S canopy width of 9.4 m and an E-W canopy width of 8.0 m (Table 2). With respect of the canopy area and volume, in general, in this wadi, the plant of *Moringa peregrina had* a higher plant area of 59.16 m^2 with a plant volume of 511.36 m^3. The data on chlorophyll content in terms of atLEAF value indicated the satisfactory health status of Moringa plants (57.5) (Table 2).

Table 2: Morphological characteristics of *Moringa peregrina* at the location of collection of seed samples, Sham, Wilayat Sohar, North Batinah Governorate, Oman

Sl. No	Characters	Sham	S.Em. (±)
1.	Plant Height (m)	6.8	0.017
2.	*Plant length (N-S) (m)	9.4	0.019
3.	*Plant length (E-W) (m)	8.0	0.013
4	Approximate plant area (m^2)	59.16	1.53
5	Approximate plant volume(m^3)	511.36	3.74
6	Chlorophyll (atLEAF value)	57.5	0.019

Physical characteristics of mature seed and kernel:

The matured viable seeds, which were grayish brown according to the Munsell color chart and their kernels being milky white, were used for measuring physical characteristics (Table 3). The shelling percentage of kernels was found to be 68.91 in terms of weight with a 50-seed weight of 80.979 g and a 50-kernel weight of 55.803 g (Table 3). The seeds and kernels are trigonous in shape and hence, they were found to be of three dimensions with a length and three distinct widths. Seed length varied from 17.10 mm to 18.66 mm with a mean length of 17.88 mm whereas kernel length ranged from 12.02 mm to 16.06 mm with a mean length of 14.04 mm. However, among the three widths, the maximum varied from 11.11 mm to 11.32 mm whereas the means were around 10.5 mm and in the case of kernel, the maximum ranged from 8.16 mm to 9.07 mm with means, about 8 mm.

Table 3: Physical characteristics of grayish brown mature seed and kernel of indigenous *Moringa peregrina*

Character		Seed	Kernel
Color		Grayish Brown*	Milky white
Color Description		2.5Y (5/2 to 6/2)*	Bright milky white
Length (mm)	Max.	18.66	16.06
	Min.	17.10	12.02
	Mean	17.88	14.04
	SE	1.103	2.856
Width 1 (mm)	Max.	11.32	9.07
	Min.	10.04	8.23
	Mean	10.68	8.65
	SE	0.905	0.593
Width 2 (mm)	Max.	11.14	8.82
	Min.	9.91	7.34
	Mean	10.53	8.08
	SE	0.870	1.0465
Width 3 (mm)	Max.	11.11	8.16
	Min.	9.72	7.31
	Mean	10.42	7.735
	SE	0.983	0.601
Mean seed coat weight (50 seed) (g)		25.176 ± 0.018	
Mean kernel weight (50 kernels) (g)		55.803 ± 0.011	
Mean seed weight (50 seed)		80.979 ± 0.007	
Mean Shelling % of Kernel		68.91	

* MUNSELL color charts for plant tissues (Second edition - Revised 1977)

Proximate composition and mineral contents:

The proximate analyses for moisture %, total ash, total fat, protein, carbohydrates, and crude fiber, were done both for mature and immature seeds to know any differences as mass harvest includes both kinds of seeds on a commercial scale (Table 4). The results of the paired-t test for proximate components and mineral contents in mature and immature seeds indicated no significant differences between their values ($p>0.05$) (Table 5).

Table 4: Proximate composition and mineral contents of Omani *Moringa peregrina* seed

	Mature Seed		Immature Seed		Mean
Sl. No	Mean	S.E.	Mean	S.E.	
Moisture % (w/w)	5.59	0.04	5.78	0.05	5.69
Total Ash (%)	2.26	0.07	2.79	0.07	2.53
Total Fat (%)	31.41	0.22	35.63	0.09	33.52

	Mature Seed		**Immature Seed**		**Mean**
Sl. No	**Mean**	**S.E.**	**Mean**	**S.E.**	
Protein (%)	21.00	0.11	23.43	0.26	22.22
Carbohydrates (%)	39.74	0.24	32.37	0.28	36.06
Crude Fiber (%)	25.65	0.13	22.69	0.45	24.17
Total Nitrogen (N) % (w/w))	3.36	0.02	3.75	0.04	3.56
Total Phosphorus (P) (ppm)	3929.39	61.08	3992.38	47.88	3960.89
Total Potassium (K) (ppm)	4259.71	66.54	5211.01	29.87	4735.36
Calcium (Ca) (ppm)	1747.57	23.82	1601.75	7.47	1674.66

Table 5: Results of paired t-test performed for proximate components and mineral contents of mature and immature seed of *Moringa peregrina*.

Seed type	Mature seed	Immature seed
Mean	1006.58	1093.16
Variance	2945684.11	3745868.732
Observations	10	10
Pearson Correlation	0.9929	
Hypothesized Mean Difference	0	
df	9	
t Stat	-0.888232667	
P(T<=t) one-tail	0.198766351	
t Critical one-tail	1.833112933	
P(T<=t) two-tail	0.397532703	
t Critical two-tail	2.262157163	

Among the proximate components in mature and immature seeds, carbohydrates were highest in percent (39.74 % and 32.37 %) followed by total fat (31.41 % and 35.63%), crude fiber (25.65% and 22.69%) and protein content (21.00% and 23.43%). Moisture percentage (5.59% and 5.78%) and total ash (2.26% and 2.79%) were less in contents (Table 5). With respect to the three mineral contents studied, Moringa peregrina had the highest total potassium (4259.71 ppm and 5211 ppm) followed by total phosphorus (3929.39 ppm and 3992.38 ppm) and total calcium (1747.57 ppm and 1601.75 ppm) in its mature and immature seeds, respectively (Table 5).

Fatty Acids (FA) Composition:

FA analyses of the seeds of both mature and immature seeds of Moringa peregrina were performed by two methods- using Hexane and Chloform: Methanol to compare and contrast their results in terms of the extraction ability of different fatty acids. The fatty acid (FA) components extracted by the two methods for mature and immature seed as well those extracted from mature and immature seed by the two methods viz. extraction using hexane (hexane method) and extraction using Chloroform: Methanol (chloroform: Methanol method)) were tested by using paired t-tests to know if any statistical differences exist. The results indicated that there were no significant differences (p>0.05) either among the values of FA components extracted from different methods (Tables 6 a & b) or among values obtained from mature (Table 6c) and immature seeds (Table 6d) by each method. This indicated that the two methods used have equal potential in extracting the fatty acids to accuracy and consistency as evidenced by several t-tests performed for paired values.

Table 6 a-d: Results of paired t-tests performed for all fatty acid components of wild *Moringa peregrina* (a) in mature seed from two methods of fatty acid analysis, (b) in immature seed from two methods of fatty acid analysis, (c) in mature and immature seed obtained from Hexane method of analysis and (d) in mature and immature seed obtained from Chloroform: Methanol method of analysis

Seed type	**Table 6a**		**Table 6b**		**Table 6c**		**Table 6d**	
	Hexane Method	**Chloroform: Methanol Method**	**Hexane Method**	**Chloroform: Methanol Method**	**Mature seed**	**Immature seed**	**Mature seed**	**Immature seed**
Mean	5.263	5.263	5.263	5.263	5.263	5.263	5.263	5.263
Variance	40.67	96.68	46.84	49.61	40.673	46.844	632.554	547.908
Observations	19	19	19	19	19	19	19	19
Pearson Correlation	0.712		0.976		0.996		0.799	
Hypothesized Mean Difference	0		0		0		0	
df	18		18		18		18	

Seed type	Table 6a		Table 6b		Table 6c		Table 6d	
	Hexane Method	**Chloroform: Methanol Method**	**Hexane Method**	**Chloroform: Methanol Method**	**Mature seed**	**Immature seed**	**Mature seed**	**Immature seed**
t Stat	2.42E-16		-4.36E-16		-1.43E-15		7.55E-17	
P(T<=t) one-tail	0.5		0.5		0.5		0.5	
t Critical one-tail	1.734		1.734		1.734		1.734	
P(T<=t) two-tail	1		1		1		1	
t Critical two-tail	2.101		2.101		2.101		2.101	

The results indicated that a total of 19 FA formed the visual outputs in chromatograms obtained by both the methods from mature seed (Table 7; Fig 2a and Fig 2b) as well as from immature seed (Table 8; Fig 3a and 3b) of indigenous Moringa peregrina tree.

Table 7: Fatty acid composition of samples of mature seed of *Moringa peregrina* collected from Sham, Wilayat Sohar, North Batinah Governorate, Oman[I] determined by two methods of analysis

Sl. No	Name	Hexane Method		Chloroform: Methanol Method	
		[I] RT (min)	% of Essential Oil	[I] RT (min))	% of Essential Oil
1	Lauric acid, methyl ester	15.337	1.75	15.345	0.04
2	Myristic acid, methyl ester	19.093	1.57	19.122	0.25
3	Pentadecanoic acid, methyl ester	21.178	0.08	21.183	0.05
4	Palmitic acid, methyl ester	23.381	22.16	23.38	14.74
5	Palmitoleic acid, methyl ester	24.764	10.08	24.764	7.34
6	Heptadecanoic acid, methyl ester	25.492	0.39	25.498	0.28
7	cis-10-Heptadecenoic acid, methyl ester	26.765	0.35	26.764	0.25
8	Stearic acid, methyl ester	27.847	6.47	27.779	11.10
9	Oleic acid, methyl ester	28.848	15.10	28.857	41.69
10	Linoleic acid, methyl ester	30.661	4.78	30.651	2.15
11	Arachidic acid methyl ester	31.694	8.71	31.68	5.56
12	cis-11-Eicosenoic acid, methyl ester	32.739	10.60	32.738	6.57
13	Heneicosanoic acid, methyl ester	33.586	0.24	33.593	0.09
14	Behenic acid, methyl ester	35.48	12.80	35.48	7.18
15	Erucic acid methyl ester	36.418	0.35	36.42	0.34
16	Tricosanoic acid, methyl ester	37.258	0.38	37.264	0.19
17	Lignoceric acid methyl ester	39.006	3.88	39.008	2.00
18	Pentacosanoic acid, methyl ester	40.842	0.10	40.843	0.09
19	Hexacosanoic acid, methyl ester	42.736	0.22	42.74	0.10
	Total		100.00		100.00

[I]RT –Retention Time (minutes); KI- Distribution Constant

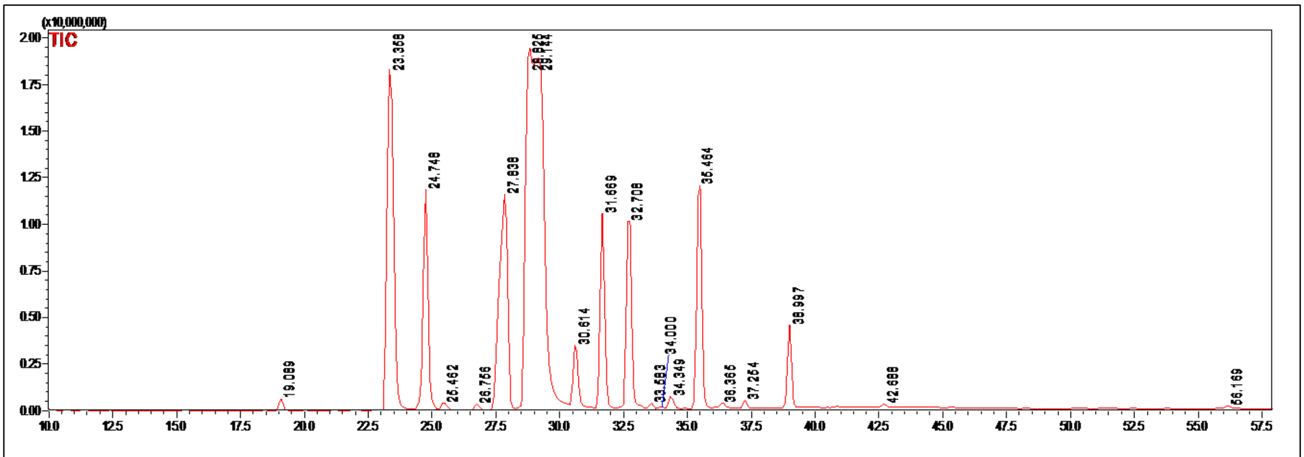


Figure 2a: Chromatograms of the volatile fractions of fatty acid determined using GC/MS for mature seed samples collected from Sham, Wilayat Sohar, North Batinah Governorate, Oman, from the Hexane method (i.e., most abundant and best separated peaks are labeled according to the numbering attributed to them in Table 7)

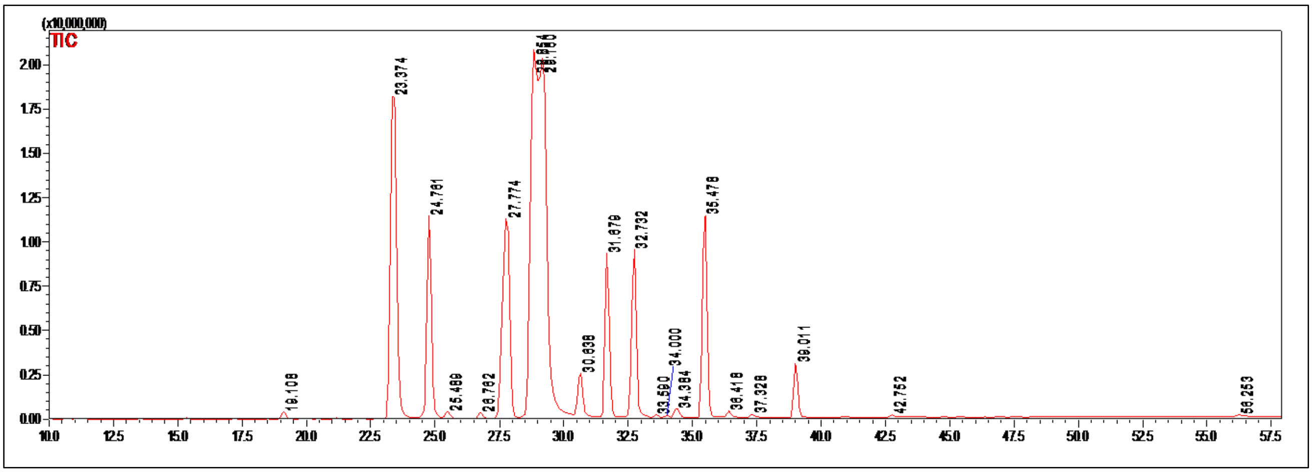


Figure 2b: Chromatograms of the volatile fractions of fatty acid determined using GC/MS for mature seed samples collected from Sham, Wilayat Sohar, North Batinah Governorate, Oman from Chloroform: Methanol Method (i.e., most abundant and best-separated peaks are labeled according to the numbering attributed to them in Table 7)

In the case of mature seed, in the Hexane method, Palmitic acid was the highest in all the FA recovered (22.16%) followed by Oleic acid (15.10%), Behenic acid (12.80%), Eicosenoic acid (10.08%) and Palmitoleic acid (10.60%) (Table 7) whereas in Chloroform: Methanol method, Oleic acid was the highest to the extent of 41.69% followed by Palmitic acid (14.74%) and Stearic acid (11.10%) (Table 7). In the case of immature seed, in both methods, Palmitic acid was the highest (22.82 % and 22.64%) followed by Oleic acid (16.82% and 18.66%) and Behenic acid (13.37% and 12.25%) (Table 8). However, in the Hexane method, Eicosenoic acid (11.16%) and Palmitoleic acid (10.41%) whereas in Chloroform: Methanol method, only Stearic acid (11.93%) exceeded 10% (Table 8).

Table 8: Fatty acid composition of samples of the immature seed of *Moringa peregrina* collected from Sham, Wilayat Sohar, North Batinah Governorate, Oman [I] determined by two methods of analysis

Sl. No	Name	Hexane Method		Chloroform: Methanol Method	
		[I] RT (min)	% of Essential Oil	[I] RT (min)	% of Essential Oil
1	Lauric acid, methyl ester	15.336	0.05	15.341	0.05
2	Myristic acid, methyl ester	19.089	0.55	19.108	0.36
3	Pentadecanoic acid, methyl ester	21.172	0.05	21.179	0.06
4	Palmitic acid, methyl ester	23.358	22.82	23.374	22.64
5	Palmitoleic acid, methyl ester	24.748	10.41	24.761	9.68
6	Heptadecanoic acid, methyl ester	25.462	0.39	25.489	0.36
7	cis-10-Heptadecenoic acid, methyl ester	26.756	0.27	26.762	0.30
8	Stearic acid, methyl ester	27.838	6.39	27.774	11.93
9	Oleic acid, methyl ester	28.825	16.82	28.854	18.66
10	Linoleic acid, methyl ester	30.614	3.42	30.638	2.61
11	Arachidic acid methyl ester	31.67	9.22	31.679	7.97
12	cis-11-Eicosenoic acid, methyl ester	32.708	11.16	32.732	9.74
13	Heneicosanoic acid, methyl ester	33.581	0.23	33.59	0.14
14	Behenic acid, methyl ester	35.464	13.37	35.479	12.25
15	Erucic acid methyl ester	36.365	0.28	36.418	0.31
16	Tricosanoic acid, methyl ester	37.254	0.38	37.328	0.11
17	Lignoceric acid methyl ester	38.997	3.94	39.011	2.67
18	Pentacosanoic acid, methyl ester	40.844	0.06	40.852	0.04
19	Hexacosanoic acid, methyl ester	42.688	0.17	42.752	0.13
			100.00		100.00

[I] RT –Retention Time (minutes); KI- Distribution Constant

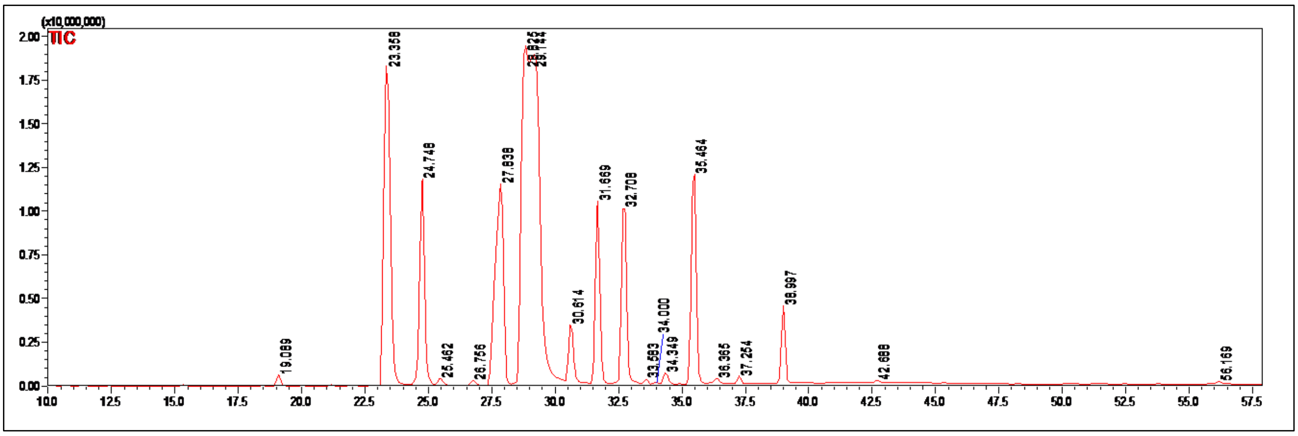


Figure 3a: Chromatograms of the volatile fractions of fatty acid determined using GC/MS for immature seed samples collected from Sham, Wilayat Sohar, North Batinah Governorate, Oman from the Hexane method (i.e., most abundant and best separated peaks are labeled according to the numbering attributed to them in Table 8

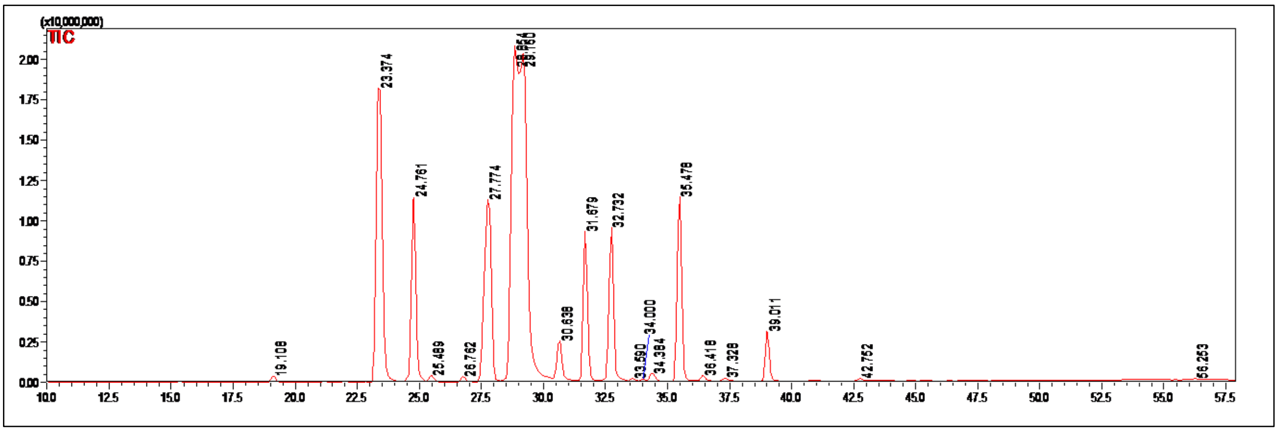


Figure 3b: Chromatograms of the volatile fractions of fatty acid determined using GC/MS for immature seed samples collected from Sham, Wilayat Sohar, North Batinah Governorate, Oman from Chloroform: Methanol Method (i.e., most abundant and best separated peaks are labeled according to the numbering attributed to them in Table 8)

DISCUSSION

There are several studies conducted on *Moringa peregrina* on its medicinal aspects since the report of Kaer *et al*., (1979) concerning its pharmacological activity as antibacterial and anthelmintic (Senthilkumar *et al*., 2018; Moustafaa and Mansour, 2018; Ma *et al*., 2020; Alkhudhayri *et al*., 2021; Wu *et al*., 2021) although healing potential of Moringa was documented as a folk medicine 5000 years ago (Patawardhan, 2000). However, physicochemical characteristics and nutritional and nutraceutical aspects of Moringa peregrina have been explored by few researchers. The results of the present investigation explored the possibility of using seeds of *Moringa* for nutritional and nutraceutical applications. Considering the output of harvest on a commercial scale which includes both matured (grayish brown) and immature (greenish white) seeds, our research examined whether or not immature seed affects the quality in terms of proximate, mineral, and fatty acid composition.

In respect to morphological traits, the plant stature /height of Moringa peregrina grown wildly in the wadi habitat, studied, was about 6.8 m which was lesser than about 8 m (PFAF, 2023) or 10m (PROTA, 2023) reported elsewhere from Arabian Peninsula or African arid or semiarid habitats. However, plant height and width found in the present study were comparable to the results of Osman and Abohassan (2012). The area (59.16 m^2) and volume (511.36 m^3) of the plant approximated in the present study along with chlorophyll estimate (57.5 atLEAF value) indicating very satisfactory health was the first in the research. Such results on growth and yield-related traits obtained through agronomic experiments assist scientists in deciding on inter-plant spacing by adopting good agriculture practices (GAP) (Navarro *et al*., 2020 in cocoa and coffee; Saha and Basak, 2020, Zhang *et al*., 2021, Leung and Cheng, 2021 and Nadaf *et al*., 2023 a, b & c, in medicinal and aromatic plants) for maximizing the seed yield to meet the demands of both industries for economic products.

Among the physical characteristics, seed color, shape, and size represented by shape, length, width, and weight of indigenous wild Moringa peregrina observed in the present investigations were comparable with the results of available two researches attempted so far (Gharibzahedi et al., 2013; Abhary, 2022) attempted so far. We described the seed color as grayish-brown described as 2.5Y (5/2 to 6/2) (Munsell, color, 2009) in comparison with brown color (PROTA, 2023) and seed shape as ovoid to trigonous as described by Osman and Abohassan (2012) and PROTA (2023) in contrast to Abhary (2022) who described as oval and circular. Our results in respect of seed length (17.88 mm) and widths (10 mm) of seed samples are comparable to that of Gharibzahedi *et al.*, (2013) and Abhary (2022) whereas that in respect of seed weight (25.173 g/50 seeds) was comparable to those of Osman and Abohassan (2012) and Gharibzahedi *et al.*, (2013). The mean shelling % of mature kernels (68.91%) has been reported for the first time considering its commercial viewpoint by the nutraceutical or food industries. The parameters of proximate composition and the contents of total phosphorous, potassium, and calcium quantified in our studies are comparable with the results of previous studies (Osman and Abohassan, 2012; Ghazibzahedi *et al.*, 2013; Abdalla *et al*., 2023).

Regarding fatty acid composition, palmitic acid (14.74% to 22.82%) followed by oleic acid (15.10% to 18.66%) and Behenic acid (12.80% to 13.37%), Eicosenoic acid (10.08% to 11.16 %) and Palmitoleic acid (10.41% to 10.60%) and Stearic acid (11.10% to 11.93%) were found in the seed of Moringa peregrina irrespective of type of seed and method of fatty acid analysis. A similar range of contents was observed in the studies of Abdulla *et al*. (2022). However, in other studies, Oleic acid was found in higher proportions (Al-Dabbas *et al*., 2010- 74.81%; Gharibzahedi *et al*., 2013- 77.9%, Abhary, 2022- 79.55%; PFAF, 2023- 71%) in comparison with other compounds (<10%) ranging between 0.06% to 9.3% % in the seed sample of Moringa peregrina, studied. In our studies also, Oleic acid was obtained to the extent of 41.69% in mature seed from the Chloroform: Methanol method (Table 8). Among the major fatty acid compounds found in the present study, oleic acid is known to replace saturated fats in the diet towards improving heart conditions by lowering cholesterol and reducing inflammation while other fatty acids like Palmitic acid, Behenic acid, Palmitoleic acid, Eicosenoic acid, and Stearic acid have cosmetic and antiaging properties for skin treatment. Unlike Moringa oleifera, which is known for its versatile consumption as food, the use of parts/ products of Moringa peregrina as a part of food should be considered cautionary although its seeds are found to contain satisfactory proximate and nutrient composition until we find certain pretreatments to remove phytotoxins to safeguard from side effects.

The promotion of nutritional or nutraceutical products of *Moringa peregrina* is commercially feasible on a large scale by producing fruits or required leaf or stem parts of the plants by growing in the field with Good Agriculture Practice (GAP) formulated by using plant characteristics like approximated plant area (m^2) and plant volume (m^3) for optimum plant density with irrigation scheduling and fertilizer management techniques. It is possible to enhance low levels of essential oil as observed in the present study through elicitation treatments by using elicitors such as Salicylic acid (Alvarenga *et al.*, 2021).

CONCLUSION

Indigenous wild Moringa peregrina was 6.8m tall occupying an area of 59.16 m^2 with satisfactory chlorophyll content (57.5 atLEAF value). Seeds were trigonous and grayish brown and able to give 68.91% of the milky white kernel. Seeds had the highest % of carbohydrates (36.05%), followed by total fat (33.52%) and proteins (22.21%). The essential oil of the seeds was found to contain more than 10% each of Oleic acid, Palmitic acid, Behenic acid, Eicisenoic acid, Palmitoleic acid, and Stearic acid. Both nutritional and nutraceutical application of Moringa peregrina has been discussed with caution for human consumption of seed and its products until the removal of their detrimental effects due to phytotoxins.

Author contributions

SKN planned the investigation, statistically analyzed the data wrote the original draft, and modified/revised based on inputs from all other authors. JNA devised, and HKA and ASA carried out the extraction of essential oil and its chemical analysis. ARA, AAA, and KAA collected seed and plant samples and recorded plant characteristics. FAA and MKA helped in soil analysis. AF confirmed the specimens as that of *Moringa peregrina*.

Conflict of Interest: The authors have no conflict of interest.

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