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Phytochemicals and Fatty Acids Composition of *Sakkoty* Date Seeds Related To Their Antioxidant and Antibacterial Activity As a promising Functional Food

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Abstract

In this study, the seeds of the *Sakkoty* cultivar of date palm (*Phoenix dactylifera L.*) underwent analysis for their physical properties and chemical composition. Gas chromatography/mass spectrum (GC-MS) analysis of extracts of defatted normal (unroasted) and roasted seeds revealed the presence of 30 and 29 different phyto-compounds in the n-hexane fraction respectively. These bioactive compounds suggested potential applications of *Sakkoty* date seeds in future drug discovery systems for various medicinal activities, including anti-inflammatory, antioxidant, cardiovascular, anticancer, and immunosuppressant properties. Additionally, investigations were conducted on the properties of the oil extracted from these seeds and their fatty acid composition. On average, the seeds contained 5.78% protein, 9.74% fat, and 73.6% carbohydrates. Total antioxidant activity of unroasted and roasted extracts seeds, ranging from 67.8 to 137.8mg AAE/100g. The primary fatty acids present in the seed oils were oleic acid (42%), lauric acid (13.5%), and palmitic acid (12%). It was observed that unroasted date seed powder samples at a concentration of 10 mg/ml demonstrated effectiveness against all tested bacterial species, except *Proteus sp.* (Gram-negative). Conversely, the roasted sample exhibited lower effectiveness against ***Escherichia coli, Staphylococcus, Proteus, Klebsiella pneumoniae, and Staphylococcus epidermidis***. It was concluded that, unroasted *Sakkoty* date seed components is more effective than roasted ones and promising for identification a new antimicrobial and antioxidant agent in managing different clinical conditions.

Keywords: Phoenix dactylifera L.; Sakkoty date seeds, bioactive compounds, antibacterial activity

1. Introduction

Date palm fruit is highly esteemed and thrives in arid and semi-arid regions globally. Cultivation of date palms (*Phoenix dactylifera L.*) is a primary livelihood in these areas, particularly in Middle Eastern cultures where dates are revered for their nutritional richness, containing high sugar, dietary fibre, and an array of macro-and micronutrients [1-3]. Among the various cultivars, *Sakkoty* dates hold a unique position, are predominantly grown in Egypt, and are commonly chosen for breaking fasts during religious observances. Despite the escalating exportation of pitted dates, surplus generation of date seeds presents a challenge for manufacturers. Currently, these seeds are either discarded [4] or used as animal feed [5]. However, they hold significant importance beyond economics and are deeply ingrained in the social fabric of these regions.

However, this by-product possesses valuable chemical components, prominently featuring oil as a noteworthy constituent. It was reported that, date seeds contain primarily lauric and oleic acids as main oil [6], tocopherols, tocotrienols, phytosterols, and phenolic compounds as bioactive ingredients [7]. For this reason, date seed can be used in many products including cosmetics, and pharmaceuticals and are promising in developing a novel therapeutic agent.

Dates were reported as functional foods due to their high energetic and nutritional value. They were used to help lactating women, enhance breast milk quality, and give birth immunity against diseases and infections [8]. In addition, date seeds showed potential antiviral, antioxidant, and anti-inflammatory properties. thus, seeds represent a valuable resource from the date processing industries for health benefits [9-10].

The rational of current study was uses the wastes of date fruit as a source of natural antimicrobial and antioxidant agent. One of major palm trees in the ASWAN region is the Sakkoty date palm. This is due to the dry air condition in this region. It is characterized by high biological nutational value.

In Egypt, a commercially widely distributed date is known as Sakkoty. This study investigated the phyto-constituents of *Sakkoty* date seed (unroasted and roasted) and fatty acid compositions using GC-MS. In Addition, examination the activity of the extract against various bacterial pathogens and antioxidant activity were examined which is promising for developing a novel therapeutic agent in many diseases.

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2. Materials and Methods

2.1. Collection of seed material

The *Sakkoty* date in this study was procured from the local Egyptian markets (Aswan). The seeds removed , washed , dried in the sun and weighed. All samples were refrigerated at approximately 5°C to maintain integrity and freshness before analysis.

2.2. Proximate Analysis

The moisture content was determined using the air oven method, whereas the protein content was assessed using the Kjeldahl method. The total nitrogen content was multiplied by a factor of 6.25 to estimate the crude protein percentage following the method described by Akasha *et al*. [11]. The ash content analysis involved incinerating approximately 2 g of date seed powder in a furnace set at 550°C for 8 h, by the protocol outlined by Rambabu *et al*. [12]. The carbohydrate content was calculated using the following formula:

100 - (%moisture + % ash + % protein + % oil) (Nehdi *et al*.) [6].

2.3. Sample preparation and phytochemical screening

Date seeds were carefully extracted, thoroughly cleaned with distilled water-dampened paper towels to remove any remaining flesh, and then dried using the same method. Half of seeds were roasted, then the roasted and unroasted seeds fragmented using a hammer mill equipped with a cooling jacket (Glen Creston Ltd, Stanmore, UK) and finely ground into a powder using a household coffee grinder. The resulting powdered seeds were washed with distilled water and dried in a vacuum oven at 70°C under a pressure of 660 mm Hg until a consistent weight was achieved. Once dried, samples were hermetically sealed in polyethylene bags and stored in a desiccator for preservation.

Initially, 50 g of powdered seeds was mixed with 120 ml of methanol (MeOH) and left at 25 ± 2°C for 72 h with occasional stirring. Using a separating funnel. The resulting extract was filtered using Whatman filter paper and fractionated with 100 ml of n-hexane (Merck, Darmstadt, Germany). The n-hexane layer was separated, and the solvent was evaporated using a rotary evaporator under reduced pressure at 40°C and 50 rpm, resulting in a concentrated extract for phytochemical analysis.

To extract fatty acids from these powders, a saponification process employing methanolic sodium hydroxide followed by methylation using a boron trifluoride/methanol reagent was utilized. This extraction method was performed by the protocol described by Czerwonka *et al*. [13].

2.4. Identification seed bioactive components by GC-MS.

One mg of n-hexane extarct was dissolved in 1 ml acetonitril and subjected to The GC-MS analysis using Agelint Technologi 5977A-M50. The capillary column, dimension: 30 m, ID: 0.25 mm, film: 0.25 mm was used ; Helium rate flow (mobile phase) was 1 ml min^{-1} . The oven temperature was vaired from 100 °C–250 °C . 5 µL was injected by Hamilton syring . The compound obtained were identified by GC libariry supplied by the macahine according to retemtion time.

2.5. Determination of Total Antioxidant Capacity.

Determination of total antioxidant activity is based on the reduction of molybdenum (VI) to molybdenum (V) by the date extracts, which produces a green phosphomolybdenum (V) in acidic conditions [14]. An 0.1 mL of (100 mg/mL) of each extract was combined with 1 mL of reagent solution (0.6 M sulfuric acid, 28 mM sodium phosphate, and 4 mM ammonium molybdate). The tubes were incubated in a thermal block at 95ºC for 90 minutes. After the mixture had cooled to room temperature, the absorbance of each solution was measured at 695 nm against a blank. Methanol (0.1 ml) in the place of extract was used as the blank. The antioxidant activity is expressed as the number of gram equivalent of ascorbic acid. The standard curve was prepared by different concentrations of ascorbic (10-100 µg/ml) with methanol. The antioxidant activity is expressed as the number of equivalents of ascorbic acid. Total antioxidant activity is expressed as mg of ascorbic acid equivalent AAE/100 g extract.

2.6. Antibacterial activity of extracts.

10 mg of each Extract (unroasted and roasted) was dissolved in 1ml Dimethyl Sulfoxide (DMSO). Six wells of 6 mm were bored in the inoculated media with the help of sterile cork-borer (6 mm). Each well was filled with 50-µl extract. The positive control (amikacin 20 µg) and negative control was DMSO. The plates were placed in the incubator at 37°C for 24 hours. After the incubation period, plates were taken out and check the microbial growth by measuring inhibition zone (mm).

Strains used for the Antibacterial activity were obtained from American Tpe Culture Colliction (ATCC) or German collectiobn of Microorganisms and cell cultures (DSMZ). Strains were subcultued and preserved using appropriate media.

Theses strains are:

Bacillus subtilis (ATCC 6051)
Enterobacter aerogenes (ATCC 13048)
Escherichia coli (ATCC 4157)
Klebsiella pneumonia subsp. pneumonia (ATCC 10031)
Proteus sp (DSMZ 46265)
Pseudomonas. aeruginosa (ATCC 19429)
Staphylococcus aureus (ATCC 6538)
Staphylococcus. Epidermidis (ATCC 12228)

3. Results and discussion

3.1. *Proximate chemical composition of date seed from Sakkoty*

Table 1 presents the Comparative Chemical Composition of Date Seeds from Sakkoty compared with previous studies carried out on *Kentichi* and *Ruzeiz*. Date seeds contained 9.95% moisture. Concerning the crude lipid contents, this value was 9.74 %. The total protein content was 5.78% (Nx 6.25) and 73.6% (by difference) on dry matter basis. The results are in agreement with the data of Herchi *et al.* [14] , except for the protein content. On the other hand, seeds of Ruzeiz cultivar contained higher carbohydrates and lower crude fat content.

3.2. GC-MS analysis of date seed powder and phytochemical screening

The GC-MS data of date seed revelead identification of multiple compounds, which were detected . GC chromatogram of two samples (roasted and non roasted date seed). Table 3 presents the corresponding retention times, peak areas, and comprehensive details of the identified compounds. The relative percentage of each component was calculated by comparing the average peak area to the total area observed in the chromatogram.

The results in Table 2 showed the presence of 30 different phytocompounds. Phenolic compounds, fatty acids, hydrocarbons and steroids 9,19-Cyclolanost-23-ene-3β-25-diol were predominant in this fraction. According to Zhou *et al*., the presence of sulfurous acid and 2-ethylhexylisohexyl ester indicates that palm trees are subjected to saline stress [008]. Among the phytochemical compounds, we identified those with antifungal and antioxidant activities, such as 2,4 DTBP, and Indan-1,3-diol monopropionate [17,18]. The roasted date seed sample (fig 2) showed that, the extract contain 29 compounds that matching in NIST library. They include aromatic and aliphatic alkenes, alkanes, terpenoids, and antifungal agents. In addition, 5-Hydroxymethylfurfural (HMF) originates from the thermal decomposition of sugars during roasting [19]. Furthermore, an increase in the HMF content was observed upon adding an increased amount of oil to defatted crushed hazelnuts [20] . An increase in HMF content from 0.9 mg/kg to 8.5 mg/kg was found in roasted (140 C for 30 min) hazelnuts [21].

Table 1: Comparative Chemical Composition of Date Seeds from Sakkoty, Kentichi, and Ruzeiz.

Assay	**Date Seeds**		
Proximate components (%)	**Kentichi[15]**	**Ruzeiz [16]**	**Sakkoty (This study)**
Moisture	9.23±0.40	1.66	9.95±0.32
Fat	7.10±0.54	0.66	9.74±0.45
Protein	2.80±0.18	5.07	5.78±0.20
Ash	1.64±0.13	1.01	1.20±0.11
Total Carbohydrate	78.69±0.60	82.97	73.6±0.40

Table 2: Total. Antioxidant activity of roasted and unroasted seed date

Extract	T. Antioxidant activity (mg AAE/g extract)
Unroasted	137.9
Roasted	67.8

Table 3: Compounds present in the hexane fraction of Sakkoty date seeds using GC-MS analysis.

S.n0	RT	Peak area %	Name of the compound	MF	MW	Compound nature
1	6.454	1.424	2,5,9-Trimethyldecane	$C_{13}H_{28}$	184	Hydrocarbon
2	6.949	0.749	2,3,6-Trimethylheptane	$C_{10}H_{22}$	142	Hydrocarbon
3	7.214	0.730	2,3,4-Trimethylhexane	C_9H_{20}	128	Hydrocarbon
4	9.245	0.639	2,3,3-Trimethylpentane	C_8H_{18}	114	Hydrocarbon
5	9.805	0.520	β-Cymene	$C_{10}H_{14}$	134	Aromatic
6	9.920	0.637	1,3,8-Menthatriene	$C_{10}H_{14}$	135	menthane monoterpenoids.
7	11.065	0.542	Nerol	$C_{10}H_{18}O$	154	monoterpene
8	11.971	0.442	3-Octanol	$C_8H_{18}O$	130	Fatty alcohol

9	13.536	0.424	2,3,5,8-Tetramethyl-decane	$C_{14}H_{30}$	198	Hydrocarbon
10	13.792	1.378	Tridecane	$C_{13}H_{28}$	184	Hydrocarbon
11	14.687	0.446	2,4,6-Trimethyl-octane	$C_{11}H_{24}$	156	Hydrocarbon
12	16.263	0.440	Hexadecane	$C_{16}H_{34}$	226	Hydrocarbon
13	16.408	1.906	Anthranilic acid	$C_9H_{11}NO_2$	165	Aromatic acid
14	16.858	0.682	O-Decylhydroxylamine	$C_{10}H_{23}NO$	173	Alkyhydroxylamine
15	17.338	0.536	3,3-Dimethylhexane	C_8H_{18}	114	Hydrocarbon
16	17.678	0.582	3,8-Dimethylundecane	$C_{13}H_{28}$	184	Hydrocarbon
17	17.753	2.552	2,6,11-Trimethyldodecane	$C_{15}H_{32}$	212	Hydrocarbon
18	17.858	0.463	2-Methyltridecane	$C_{14}H_{30}$	198	Hydrocarbon
19	17.903	0.627	2-ethylhexyl 4-methylpentyl sulfite	$C_{14}H_{30}O_3S$	278.	Alkylsulfates
20	18.053	2.463	2,4-di-t-Butylphenol	$C_{14}H_{22}O$	206	Phenolic
21	18.248	0.669	Indan-1,3-diol monopropionate	$C_{12}H_{14}O_3$	206	alkanediol dialkanoates
22	18.438	1.255	2,3,5,8-Tetramethyl-decane	$C_{14}H_{30}$	198	Hydrocarbon
23	18.824	0.725	Dodecanoic acid	$C_{12}H_{24}O_2$	200	Fatty acid
24	19.384	0.474	Octadecane	$C_{18}H_{38}$	254	Hydrocarbon
25	19.474	0.553	Myristic acid	$C_{14}H_{28}O_2$	228	Fatty acid
26	19.736	0.517	Nonadecane	$C_{19}H_{40}$	268	Hydrocarbon
27	20.614	0.527	Palmitic acid	$C_{16}H_{32}O_2$	256	Fatty acid
28	20.674	2.971	9,19-Cyclolanost-23-ene-3β-25-diol	$C_{30}H_{50}O$	426	Sterol
29	20.724	0.434	9,19-Cyclolanost-24-ene-3β-ol	$C_{31}H_{50}O$	438	Sterol
30	21.024	0.420	9,19-Cyclolanost-3-ol,24-methylene, 3β-acetate	$C_{31}H_{52}O$	440	Sterol

Table 4: Compounds present in the hexane fraction of roasted Sakkoty date seeds using GC-MS analysis.

S.n0	RT	Peak area %	Name of the compound	MF	MW	Compound nature
1	4.358	1.099	Cumene	C_9H_{12}	120	Aromatic
2	4.434	0.445				
3	4.549	0.475	1,3,4-Trimethylbenzene	C_9H_{12}	128	Aromatic
4	5.054	1.182	2,2,4,6,6-Pentamethylheptane	$C_{12}H_{26}$	170	Hydrocarbon
5	5.199	1.699	2,4,6-Trimethylbenzene	C_9H_{12}	134	Aromatic
6	6.229	0.708	2,5-Dimethylheptane	C_9H_{20}	128	Hydrocarbon
7	6.429	0.429	3,7-Dimethylnonane	$C_{11}H_{24}$	156	Hydrocarbon
8	6.489	1.170	2,5,9-Trimethyldecane	$C_{13}H_{28}$	184	Hydrocarbon
9	7.010	0.746	2,2,3,3-Tetramethylpentane	C_9H_{20}	128	Hydrocarbon
10	7.190	0.446	2,2,3,4-Tetramethylpentane	C_9H_{20}	128	Hydrocarbon
11	7.280	0.611	Hexadecane	$C_{16}H_{34}$	226	Hydrocarbon
12	8.640	0.439	m-Xylene	$C_{10}H_{14}$	134	Aromatic
13	9.286	0.506	6-ethyl-2-methyldecane	$C_{13}H_{28}$	184	Hydrocarbon
14	9.681	0.433	maltol	$C_6H_6O_3$	126	Oxygenated furans
15	9.961	0.612	1,3,8-p-Menthatriene	$C_{10}H_{14}$	134	menthane monoterpenoids
16	10.686	1.101	Pyranone	$C_6H_8O_4$	144	Unsaturated cyclic esters
17	12.797	1.487	Hydroxymethylfurfural	$C_6H_6O_3$	126	furandicarboxylic acid
18	13.807	1.083	2-Methyltridecane	$C_{14}H_{30}$	198	Hydrocarbon
19	16.213	0.446	L-Glutamine	$C_5H_{10}N_2O_3$	146	amino acid
20	16.273	0.439	n-Hexadecane	$C_{16}H_{34}$	226	paraffin
21	16.423	0.871	Anthranilic acid	$C_9H_{11}NO_2$	165	Aromatic acid
22	17.354	0.420	2,2,3,3-Tetramethylhexane	$C_{10}H_{22}$	142	Hydrocarbon
23	17.689	0.414	Dodecanoic acid	$C_{12}H_{24}O_2$	200	Fatty acid
24	17.764	1.808	2,6,11-Trimethyldodecane	$C_{15}H_{32}$	212	Hydrocarbon
25	17.939	0.400	***Oxalic acid*** dimethyl ***ester***	$C_{24}H_{46}O_4$	398	Ester
26	18.064	1.130	2,4-Bis(1,1-dimethylethyl)phenol	$C_{14}H_{22}O$	206	Phenolic
27	18.454	1.008	2,9-dimethyldecane	$C_{12}H_{26}$	170	Hydrocarbon
28	19.390	0.428	Tetradecane	$C_{14}H_{30}$	198	parafin
29	19.500	0.446	2-methyl-5-propylnonane	$C_{13}H_{28}$	184	Hydrocarbon

Table 5: Fatty acid profiles of date seed oil expressed as percentage of total fatty acid quantified.

	Seeds				
Compounds	This study Sakkoty (no roasted)	This study Sakkoty (roasted)	Mazafati and Kalutah [[29]	Barhi, Khalas, Manifi, Rezeiz, Sulaj, and Sukkari [30]	Arechti, Degla-Baida, Deglet Nour, Ghars, Haloua, Itima, Mech-Degla, Tentbouchet [31]
Capric acid (10:0)	3.0	4.0	0.2–0.5	0.71	0.25–0.61
Lauric acid (12:0)	8.0	13.5	14–15.8	10.36	19.76–22.25
Myristic acid (14:0)	7.0	15.0	10.6–10.9	10.44	9.57-10.17
Palmitic acid (16:0)	12.0	15.0	10.8–11.8	12.83	9.17–10.37
Heptadecanoic acid (17:0)	nd	14.0	nd	nd	0.07-0.13
Stearic acid (18:0)	8.0	10.0	3–3.4	5.56	3.07-3.52
Palmitoleic acid (16:1)	nd	nd	0.2–0.4	nd	0.12-0.16
Oleic acid (18:1)	42	15	48.1–50.5	51.45	42.74–48.14
Gadoleic acid (20:1)	nd	nd	0.2–0.4	0.37–0.52	0.01-0.13
Linoleic acid (18.2)	12	nd	7.7–8.2	7.2	6.58–8.12
Linolenic acid (18:3)	nd	nd	0.4–0.7	nd	nd
Archidonic acid (20:4)	3.0	nd	nd	nd	nd

[1] nd: not detected.

Table 6: Antibacterial activity against different microbes (Inhibition zone, mm) at zero time.

Bacterial species	Unroasted	Roasted
Escherichia coli (Grame –Ve)	10 ±1.9	6 ±0.65
Pseudomonas. aeruginosa (Grame –Ve)	8±0.74	Resistant
Proteus spp (Grame –Ve)	Resistant	8±0.92
Klebsiella pneumonia (Grame –Ve)	9±0.9	7±1.00
Enterobacter aerogenes (Grame –Ve)	10±0.34	Resistant
Staphylococcus. Epidermidis (Grame +Ve)	7±1.11	8±0.44
Staphylococcus aureus (Grame +Ve)	9±0.98	Resistant
Bacillus subtilis (Grame +Ve)	6±0.11	Resistant

3.3. Total Antioxidant activity

Results of total antioxidant activity of Unroasted and roasted extracts are shown in Table 2. The ability of a seed date extracts to scavenge the reactive metabolites would inhibit the formation of primary and secondary amines oxidation products. In this investigation, unroasted showed highest free radical scavenging activity (137.9 mg AAE/g) compared to roasted seed date (67.8 mg AAE/g).

3.4. Fatty Acid Composition

Data obtained in Table 4 showed the fatty acid composition of *Sakkoty* date seeds. In normal seed (non roasted) , it was found that, five major fatty acids, (oleic acid (C18:1), linoleic acid (C18:2), palmitic acid (C16:0), lauric acid (C12:0) , and myristic acid (C14:0) with difftrnt concentartions. However, minor fatty acids were detected in lower amounts, including capric (C10:0), palmitoleic (C16:1), linolenic (C18:3), and gadoleic (C20:1) acids . In addition, the roasted sample fatty acids content is different from normal sample by presence of monounsaturated fatty acid: Myristioleic (C14:1) and Heptadecanoic (C17:0).However, the higher percent of saturated relative to unsaturated (28.5% unsaturated and 71.5 saturated fatty acids). In general, date seed oil comprises a range of fatty acids: saturated (lauric, myristic, and palmitic acids), mono-unsaturated (palmitoleic and oleic acids), and polyunsaturated (linoleic and linolenic acids). According to Nehdi *et al.* [22], among six Saudi Arabian cultivars, the highest percentages of unsaturated and saturated fatty acids are oleic, palmitic, and lauric acids, respectively. Harkat *et al.* [23] reported that Algerian date seed oils were primarily composed of oleic acid (42.74-48.14%) and lauric acid (19.76-22.25%). Another study classified Iranian date seed oils as oleic-lauric [24]. Oils abundant in oleic acid have received significant attention owing to their exceptional stability and nutritional benefits. Oleic acid (OA), a crucial unsaturated fatty acid, is of vital importance for human nutrition. Its properties include the ability to combat specific heart and vascular issues, low saturation levels, potential for reducing cholesterol in the bloodstream, and impressive resistance to oxidation [25]. There is widespread recognition that incorporating oils rich in unsaturated fatty acids into one's diet prevents cardiovascular and inflammatory conditions [26]. Moreover, numerous studies have underscored the role of lauric acid in inhibiting the development of prostatic hyperplasia [27], its superior characteristics compared to trans-fatty acids [28], and its antimicrobial properties, effectively restraining microbial growth and toxin production [27, 28].

3.5. Antimicrobial activity of extracts

The effectiveness of the extracts in tested bacterial strains was determined by measuring the minimum inhibitory concentration (MIC). MIC was performed for only those organisms which showed a zone of inhibition and were sensitive to the extracts.

It was found that, normal date seed powder sample (10 mg/ml) are effective on all tested bacteria species except Proteus sp. (Grame –Ve). The highest effective on *Escherichia coli* (Grame –Ve), *Enterobacter aerogenes* (Grame –Ve), *Klebsiella pneumonia* subsp. *pneumonia* (Grame –Ve), *Staphylococcus aureus* (Grame +Ve) and *Pseudomonas. aeruginosa* (Grame –Ve). However it showed low activity toward *Staphylococcus. Epidermidis* (Grame +Ve) and *Bacillus subtilis* (Grame +Ve)

Testing roasted sample showed effective on Staphylococcus. Epidermidis (Grame +Ve), Proteus sp. (Grame –Ve), *Klebsiella pneumonia* subsp. pneumonia (Grame –Ve), *Escherichia coli* (Grame –Ve) w, while *Pseudomonas. aeruginosa* (Grame –Ve), *Enterobacter aerogenes* (Grame –Ve), *Staphylococcus aureus* (Grame +Ve) and *Bacillus subtilis* (Grame +Ve) showed resistance to the roasted extract.

5. Conclusions

Phytochemical and GC-MS analyses of Egyptain commerical *Sakkoty date* seed showed promising pharmacological potential, including antioxidant , analgesic, and antimicriobial agent. Notably, the predominant fatty acids in date seed oils were oleic acid and palmitic acid, followed by linoleic acid. Remarkably, these oils displayed lower levels of polyunsaturated fatty acids (approximately 15 %), which are crucial for ensuring stability during storage. This hints at the potential for an extended shelf life. Consequently, these date seeds hold promise as a valuable source of edible oils for human consumption, pending comprehensive safety evaluation.

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