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Evaluation of the sensory properties, volatile aroma compounds and functional food potentials of cold-press produced mahaleb (*Prunus mahaleb* L.) seed oil☆

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Abstract – The aims of this study were to cold-press mahaleb seed, and then fully characterize the oil to extent its food and functional food applications. The novelty of this study relies upon the first data provided by the thermal analysis, sensory analysis, and volatile aroma compounds composition. The seeds were pressed with a screw-type single-head press with a maximum oil exit temperature of 40 °C. Most common physico-chemical properties, composition analyses, volatile aroma compounds profile, sensory descriptive analysis and consumer tests were completed. The main properties were appropriate and the composition (fatty acids, phytosterols and tocopherols) data concurred with the available literature. Thermal data were provided, and the oil peak crystallization and melting temperatures were −44.45 °C and −8.41 °C, respectively. There were 38 volatile aroma compounds quantified mostly with almond, green, vanillin, woody, and fermented aroma definitions. The panel described the oil with 5 sensory descriptive (almond, vanillin, dough, green, cooling) terms. Consumers liked appearance the most (4.49) with a general acceptance score of 3.70 on a 5-point hedonic scale. Overall, the mahaleb seed oil is a conjugated linolenic, oleic and linoleic fatty acids, β-sitosterol and γ-tocopherol rich, very aromatic, and consumer-liked sample. Further studies with various food applications are foreseen.

Keywords: mahaleb seed / cold press oil / aromas / sensory / composition

Résumé – **Évaluation des propriétés sensorielles, des composés d'arômes volatils et du potentiel en tant qu'aliment fonctionnel de l'huile de graines de mahaleb (*Prunus mahaleb* L.) extraite par pression à froid.** Les objectifs de cette étude étaient de presser à froid les graines de mahaleb, puis de caractériser complètement l'huile afin de déterminer ses applications alimentaires et fonctionnelles. La nouveauté de cette étude repose sur les premières données fournies par l'analyse thermique, l'analyse sensorielle et la composition en composés d'arômes volatils. Les graines ont été pressées à l'aide d'une presse à vis à une tête avec une température maximale de sortie de l'huile de 40 °C. Les propriétés physico-chimiques les plus courantes, les analyses de composition, la détermination des composés d'arômes volatils, l'analyse sensorielle descriptive et des tests consommateurs ont été réalisés. Les principales propriétés se sont avérées appropriées aux applications visées et les données relatives à la composition (acides gras, phytostérols et tocophérols) concordantes avec la littérature disponible. Les données thermiques ont été fournies ainsi que les températures de cristallisation et de fusion maximales de l'huile respectivement de −44,45 °C et de −8,41 °C. Trente-huit composés volatiles aromatiques ont été quantifiés, principalement des arômes d'amande, de verdure, de vanilline, de bois et de fermentation. Le panel a décrit l'huile avec 5 termes descriptifs sensoriels (amande, vanilline, pâte à pain, vert, rafraîchissant). Les consommateurs ont le plus apprécié l'aspect (4,49) avec une note d'acceptation générale de 3,70 sur une échelle hédonique de 5 points. Dans l'ensemble, l'huile de graine de mahaleb s'avère riche en acides gras linolénique, oléique et linoléiques conjugués, β-sitostérol et γ-tocophérol, très aromatique et apprécié des consommateurs. D'autres études avec diverses applications alimentaires sont prévues.

Mots clés : graines de mahaleb / huile pressée à froid / arômes / sensorialité / composition

☆ Contribution to the Topical Issue "Minor oils from atypical plant sources / Huiles mineures de sources végétales atypiques".

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1 Introduction

Mahaleb cherry (*Prunus mahaleb* L.) is a tree classified under the Rosaceae family and Prunoidea subfamily. The tree grows wild and is also cultivated in the Mediterranean, West Asia, and Southeastern Europe regions. It has small and underutilized fruits, but the fruit seeds and seed flours are essential flavoring agents in many baked goods, foods, tonics, and medicines. The seeds have a typical almond-like and vanillin-like flavor, and consequently, its name (mahaleb) comes from the Arabic meaning "sweet aromatic or perfume king" (Özbey *et al.*, 2011; Alma *et al.*, 2012).

The mahaleb seeds have been shown to include 24.8–31.92% protein, 16.11–32.47% lipid, 15.07–31.80% carbohydrates, 2.1–4.5% ash, and 3.66–6.20% moisture (Mariod *et al.*, 2009; Alshehri, 2014). Further, the presence of coumarins, dihydro-o-coumaric acid 2-o-glucoside, and o-coumaric acid 2-o-glucoside in the seeds was published (Ieri *et al.*, 2012). Also, small quantities of cyano-genic glycosides, herniarin (7-methoxy coumarin), dihydro coumarin, and amygdaline (mandelonitrile-β-gentiobioside) were reported (Öztürk *et al.*, 2014).

Mahaleb seeds were extracted with petroleum ether (30.95% total oil), and the main physico-chemical properties (specific gravity, refractive index, free fatty acidity, acid number, peroxide number, saponification number, and unsaponifiable matter content) were published (Mariod *et al.*, 2009). In another similar study (Sbihi *et al.*, 2014), hexane-extracted Syrian mahaleb oil (31.26% total oil) was analyzed for the same properties, and also the thermal degradation temperature (520 °C) was provided. The fatty acid compositions of 11 selected seeds including mahaleb were published. Mahaleb oil contained around 4.6%, 27.6%, 28.5%, and 35.4% of palmitic, conjugated linolenic (CLNA), linoleic, and oleic acids, respectively (Özgül-Yücel, 2005). The main fatty acids of differently colored mahaleb seeds were reported as around 7.38–8.66% of palmitic, 22.48–24.01% of CLNA, 29.38–33.67% of oleic, and 29.11–34.13% of linoleic acids (Ercisli and Orhan, 2008). Sbihi *et al.* (2014) reported white mahaleb seed oil fatty acid compositions as α-eleostearic (CLNA) (38.32%), oleic (31.29%), and linoleic (22.96%) acids. In a later study (Erdogan Eliuz *et al.*, 2022), 31.76% oleic, 25.54% linoleic, 34.39% CLNA, and 3.67% palmitic acid were reported in mahaleb oil.

This study aimed to characterize cold-press produced mahaleb seed oil and the extent of its possible food and functional food applications. Further, to the best of our knowledge, the thermal properties, volatile aroma compounds composition, sensory descriptive properties, and consumer preferences of the oil were reported for the first time in the literature. Overall, these data provide the novelty of this study.

2 Materials and methods

2.1 Materials

The mahaleb seeds were purchased from Bora Baharat Co. (Adana, Türkiye). According to the company statement, the seeds were harvested in November–December 2019 and cultivated in orchards of Tokat Province. The seeds were purchased in June 2020 and stored until the end of the year 2020 in a deep freezer. The seeds were cold-pressed in January 2021, and immediately the analyses were started. All analyses of both replicate oil samples were completed by around June 2021. Consequently, it recommended that the seeds were one year stored samples before cold-pressing. The two replicates of cold-pressing were performed by one weak apart after dividing the seeds into two equal portions at the beginning. The seeds can be observed in Figure 1. All chemicals, solvents, standards, and utensils used were of analytical grade and purchased from Sigma Chem. Co. (St. Louis, US), Merck (Darmstadt, Germany), and local stores.

2.2 Seed analyses

The color of the seeds was measured with a Minolta CR-400 Reflectance Colorimeter (Minolta Camera Co., Osaka, Japan) randomly over several points of seed surfaces, and the values of L* (brightness/darkness), a* (redness/greenness) and b* (yellowness/blueness) were recorded. Seed dimensions were measured with a digital caliper (CD-15CP, Mitutoyo Ltd., Andover, UK) to record seed length, width, and thickness. Randomly selected 10 seeds were weighed with a Sartorius ED2245 scale (Sartorius, Germany), and then multiplied by 10 to get 100-seed weight for several times.

After grinding the seeds into fine flour, some compositional analyses were also completed. Total moisture content was determined with an Ohaus MB45 moisture analyzer (Ohaus, Pine Brook, US). Total crude oil content was measured with the Soxhlet technique according to AOAC 920.39 method (AOAC, 2002). Similarly, total crude protein by Kjeldahl according to AOCS method Aa 5-35, and ash content by oven burning following AOCS method Ba 5a-49 were measured (AOCS, 1998).

2.3 Cold press oil production

The mahaleb seeds had 5.20% moisture, and before cold-pressing, the moisture level of the seeds was adjusted to 10% by the water equilibration process. The amount of added water was calculated by the equation below. The tab water was added to the seeds in a closed vessel and stayed for 24 h at room temperature for moisture penetration and equilibration.

$$\text{Added water (g)} = [(A/B \times C)] - C,$$

where A is the measured total solids of the seeds (%), B is the targeted total solid of the seeds (%), and C is the total amounts of the seeds (g).

After moisture adjustment, the seeds were cold-pressed with a laboratory scale machine (single head, 1.5 kW, 12 kg seed/h capacity; Koçmaksan ESM3710 model, İzmir, Turkey) with 20 rpm screw rotation speed, 14 mm exit die, and 40 °C oil exit temperature. Then, the oils were centrifuged (Sigma 2-16 K, Postfach, Germany) at 6797 xg to remove suspended materials and weighed. Finally, the oil was placed into brown-colored and capped glass bottles after flushing with nitrogen gas. The oil samples were stored in a refrigerator at 4 °C during the analyses. The oil yield of the cold-pressing technique was calculated following the equation below:

Fig. 1. The mahalep seeds and cold-press produced mahaleb seed oil.

$$\text{Oil yield } (\%) = (A/B) * 100,$$

where A is the total amount of oil obtained by cold pressing (g), and B is the total oil content (g) of the seeds determined by the Soxhlet technique.

2.4 Physico-chemical properties of the oil samples

Oil refractive index with Abbe 5 (Bellingham and Stanley, UK) refractometer at 25 °C, specific extinctions with a spectrophotometer (Shimadzu UV-1800, Shimadzu Co., Japan) by AOCS method Ch 5-91 (AOCS, 1998), and viscosity with Brookfield Viscometer (model DV II+Pro with Rheocalc software, Brookfield Eng. Lab., Inc., MA, USA) donated with LV-SC4-18 spindle at 30 rpm at 25 °C were measured. Oil color values (L*, a* and b*) were measured with a Minolta Colorimeter CR-400 (Minolta Camera Co., Osaka, Japan).

The AOCS methods of Ca 5a-40 and Cd 3d-63 were followed to determine oil-free fatty acids (FFA) and acid values (AOCS, 1998). Similarly, oil peroxide value and iodine number were measured by the AOCS Cd 8-53 and Cd 1-25 methods, respectively (AOCS, 1998). The ISO 3596 method was followed to measure oil unsaponifiable matters content (ISO, 1991).

2.5 Thermal properties of the oil samples

Melting and crystallization temperatures and enthalpies of the oil samples were assessed with a Perkin-Elmer 4000 Series Differential Scanning Calorimeter-DSC (Groningen, The Netherlands). Around 8 mg sample was weighed and sealed in aluminum pans, and analyzed under 50 mL/min nitrogen flow rate with the following temperature program; cool from 20 °C to −60 °C by 5 °C/min rate, keep oil at that temperature for 3 min, and then heat oil to 60 °C by 5 °C/min rate. The Pyris 1 manager software was used to calculate the thermal parameters.

2.6 Fatty acid, phytosterol and tocopherol composition of the oil samples

The fatty acid composition (%) of mahaleb seed oil was determined by following AOCS methods Ce 2-66 and Ce 1h-05 (AOCS, 1998). An Agilent 7890B gas chromatograph (Agilent Technologies, Palo Alto, CA, USA) equipped with flame ionization detector (FID) (Agilent Technologies, Palo Alto, CA, USA), and HP 88 capillary column (100 m × 0.25 mmID × 0.2 µm film thickness, J&W Scientific Co, CA, USA) were used. The thermal oven program was heating at 120 °C for 1 min, heating to 175 °C (10 °C/min) for 10 min, heating to 210 °C (5 °C/min) for 5 min, and heating to 230 °C (5 °C/min) for 5 min. The injection volume was 1 µL, the injector split ratio was 1:50, the flow rate was 2 mL/min, hydrogen was the carrier gas, and the injector and the detector temperatures were 250 °C and 280 °C, respectively. Fatty acid methyl esters were quantified by co-chromatography with FAME mixture standards (37-components, C4-C24, Supelco, Bellefonte, PA, USA). A sample chromatogram can be observed in Figure 2. The peak present at around 28 min was

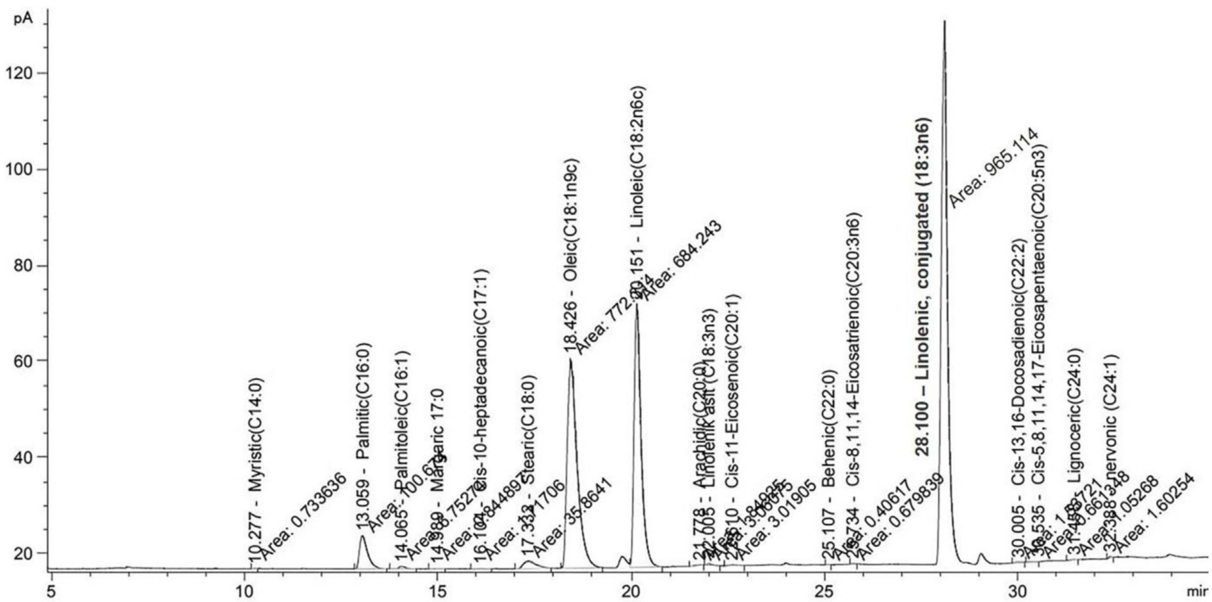

Fig. 2. Fatty acid GC chromatogram of a cold-press produced mahaleb seed oil sample.

not determined with 37-component standard, but after close comparison with available literature (Özgül-Yücel, 2005; Erdogan Eliuz *et al.*, 2022) providing the peak retention times run in a very similar GC system, it was decided that the peak corresponds to conjugated linolenic acid (CLNA), respectively.

The phytosterol composition (%) of the oils was measured by the ISO 12228 method (ISO, 1999). The unsaponifiable matter was separated previously according to ISO 3596 method (ISO, 1991). After separating the phytosterol fraction from other unsaponifiable matters on TLC, the phytosterol analysis was completed with the same GC and DB5 capillary column (30 m × 0.25 mm ID × 0.1 µm film thickness, J&W Scientific Co, CA, USA). The oven program was as follows: keep at 60 °C for 2 min, heat to 220 °C (40 °C/min) for 1 min, and heat to 310 °C (5 °C/min) for 30 min. The injection volume of 1 µL, injector split ratio of 1:100, and flow rate of 0.8 mL/min, with a hydrogen carrier gas (30 mL/min) was applied. The injector and the detector temperatures were 290 °C and 300 °C, respectively. Commercially available standards (cholesterol, brassicasterol, stigmasterol, β-sitosterol) were used under the same conditions to identify and quantify the sterols in the samples.

The tocopherol composition (mg/kg oil) of the samples was determined following the method of Grilo *et al.* (2014) with a reverse-phase HPLC (Shimadzu Corporation, Kyoto, Japan) equipped with an Inertsil ODS-3 column (250 mm × 4.6 mm × 5 µm, GL Sciences Inc., Japan), LC-20AT HPLC pump, DGU-20A5R degasser, CTQ-10ASVP column oven, and RF-20A diode array detector. Briefly, 0.15 g oil samples were dissolved in 3 mL dichloromethane, and from this stock, 20 µL were injected by an autosampler (SIL-20AHT) into the HPLC. The methanol: water (98:2 v/v) mobile phase was eluted with a 1.6 mL/min isocratic flow rate. The detector excitation and emission wavelengths were 290 and 330 nm, respectively. Quantification was performed using commercial tocopherol standards (Merck, Darmstadt, Germany).

2.7 Volatile aroma compounds composition of the oil samples

Volatile aroma compounds of the oil samples were analyzed following Yılmaz *et al.* (2015). First, mahaleb seed oil (2 g), salt (0.5 g), and internal standard (20 µL; 1 µL of 2-methyl-3-heptanone dissolved in 10 mL methanol) were placed into an amber-colored vial and vortexed for 1 min. Then, the vial was kept at 60 °C for 15 min in a water bath before immersing an SPME fiber (2 cm, 50/30 µm DVB/Carboxen/PDMS, Supelco, Bellafonte) into the vial headspace. The inserted vial was kept for 45 minutes to adsorb the volatiles. Finally, fiber collected volatiles were analyzed with a GC/MS (Agilent 6890HB/Agilent 5875C mass spectrometer; Agilent Technologies, Wilmington, DE, USA) equipped with HP5 MS column (30 m × 0.25 mm, i.d. 0.25-µm film thickness, J & W Scientific, Folsom, CA, USA). The helium was the carrier gas with a 1.61 mL/min flow rate and 1:2 split ratio. The oven temperature program was waiting at 40 °C for 2 min, heating to 200 °C (4 °C/min), heating to 240 °C (7 °C/min), and waiting at that temperature for 15 min. The working conditions of MS detector were 280 °C capillary direct interface temperature, 70 eV ionization energy, 35–350 amu mass range, 4.45 scans/s scanning rate. A C7-C30 saturated alkane standard was used to calculate the retention indices. Finally, the volatiles tentatively identified using the mass spectra libraries of Wiley, Nist, Tutor, and FFNSC. The data presented as % area values and retention times of the measured volatile compounds.

2.8 Sensory analyses of the oil samples

The Quantitative Descriptive Analysis (QDA) method was used to describe the oil samples with a trained panel (Meilgaard *et al.*, 1991). There were 10 panelists (6 female and 4 male, aged between 21–51) and trained for at least 16 h in

Table 1. Some physical properties and proximate composition of the mahalep seeds used (Mean ± SD).

	Mahalep seed		Mahalep seed
Color L* value	49.10 ± 3.30	Moisture (%)	5.20 ± 0.12
Color a* value	9.96 ± 2.19	Crude Oil (% Dry content, Soxhelet)	32.12 ± 0.14
Color b* value	25.67 ± 1.05	Protein (% Dry content)	32.65 ± 0.21
Length (mm)	5.73 ± 0.23	Ash (%)	3.87 ± 0.08
Width (mm)	3.43 ± 0.17		
Thickness (mm)	4.03 ± 0.39		
100-seed weight (gr)	3.38 ± 0.39		

different sessions under the moderation of the panel leader. Panel training was accomplished with oil samples and some food and chemical solution standards to familiarize the panelist with the sensory attribute terms extensively. After training, some pre-tests were completed to measure the panel response ranges, and training continued until all members get trained, and had quite similar test scores. The panel used some commercial cold-pressed oil samples and actual oils produced in this study to determine the sensory terms. The panel defined and described 5 sensory definition terms for this oil. The term "almond" was described as the flavor and aromas associated with almonds, and fresh almonds and benzaldehyde were the standards for 10 point, and absence of the almond was decided with a zero score. "Vanillin" was defined as a sweet-vanilla-like aroma with commercial vanillin as the calibration standard for full score. The term "dough" was defined with the total aromas associated from freshly prepared wheat dough. Fresh wheat flour dough was used as the standard to calibrate the panelist for 10 score. "Green" was defined as aromas associated with cut grass and *cis*-3-hexanol, and cut grass was used as the standard for a total score. Finally, "cooling" was defined as the cool sensation felt in the mouth space after oil tasting, and menthol candy was the standard (10 score) for it. Line scales (10 cm) were used for quantification, anchored with 1 at the left end for minimum intensity and 10 at the right end for maximum intensity. The samples were coded with 3-digit numbers and served in glasses to the panelists with water, unsalted crackers, and expectoration cups. A signed consent form was prepared indicating that the oil sample was food grade and safe, but panelists were suggested not to ingest it during the test. The analyses were conducted under daylight at room temperature in different sessions in a randomized order.

The appearance, smell/aroma, taste/flavor, and general acceptance of the oil samples were evaluated with 171 volunteer consumers (109 men, 62 women, 19–60 years old) using a 5-point hedonic scale (1 = dislike extremely, 5 = like extremely). The samples were coded with 3-digit numbers and served in glasses to the consumers with water, unsalted crackers, and expectoration cups in different sessions. The same consent form was also provided to the consumers (Meilgaard *et al.*, 1991).

2.9 Statistical analyses

Cold pressing of the mahaleb seeds was replicated twice. For each replicate sample, the listed analyses were done in triplicate. The collected data were provided as mean values with standard deviations.

3 Results and discussion

3.1 Properties of the mahaleb seeds

Some physical properties and proximate composition of the mahaleb seeds used to produce the cold-pressed oils were measured, and the data are summarized in Table 1. The seeds could also be observed in Figure 1. It would be valuable to acquaint the source of the oil. The seeds seem similar to almonds with much smaller dimensions, and their color is brown-yellow with 25.67 b* value and 9.96 a* value. The seed cold-pressed in this study had around 7.73 mm length, 3.43 mm width, and 4.03 mm thickness. The length, width, and thickness of 9.5% and 23% moisture-containing mahaleb seeds were reported as 7.6–8.62 mm, 6.11–7.03 mm and 5.59–7.03 mm, respectively (Yesiloğlu and Pınar, 2006). Clearly, seed dimensions changes by seed total moisture level, but the findings concur with literature. The measured 100-seed weight (3.38 g) is in good agreement with the literature reporting a 3.7 g value (Özgül-Yücel, 2005). The measured moisture content (5.20%) indicates that the seeds were dried, and the moisture was within the reported ranges (Mariod *et al.*, 2009; Yesiloğlu and Pınar, 2006). Likewise, around 32.12% of total crude oil and 32.65% of crude protein on dry matter basis and 3.87% of ash seem in good agreement with previous studies (Mariod *et al.*, 2009; Alshehri, 2014). Lately, mahaleb seed composition was reported as 4.30–4.69% moisture, 3.21–3.56% ash, 35.01–35.67% fat, 27.52–27.98% protein, and 28.57–29.85% carbohydrate (Dadalı, 2022). The mahaleb seeds used in this study to produce the cold-pressed oil had quite similar composition data to those reported in the literature.

3.2 Physico-chemical properties of the cold-pressed mahaleb seed oil

The oil yield was around 24.90% with the cold-pressing of the mahaleb seeds in this study (Tab. 2). This is an expected result for the cold-pressing technique. According to the literature, the cold-pressing technique yield lower quantity oils in expense of higher quality (Yılmaz *et al.*, 2015, 2020; Ok and Yılmaz, 2019). The main physico-chemical properties of the cold-pressed mahaleb seed oil are presented in Table 2. The oil refractive index (1.495) seems to be within the range (1.4–1.5)

Table 2. Some physical and chemical properties of the cold-pressed mahalep seed oil (Mean ± SD).

	Mahalep oil		Mahalep oil
Refractive Index (25 °C)	1.495 ± 0.00	Free Fatty Acidity (oleic %)	0.65 ± 0.04
Specific Extinction-E232	2.723 ± 0.138	Acid Number (mg KOH/g yağ)	1.31 ± 0.09
Specific Extinction-E270	0.290 ± 0.003	Peroxide Value (meq active O_2/kg oil)	4.29 ± 0.15
Viscosity (25 °C, cP)	113.5 ± 0.7	Iodine Value (g I/100 g oil)	158.15 ± 6.51
Color L* value	27.18 ± 0.01	Unsaponifiable Matter (%)	0.72 ± 0.04
Color a* value	−0.3 ± 0.03		cold-pressing
Color b* value	5.03 ± 0.00	Oil Yield (%)	24.90 ± 0.55

defined for vegetable oils and mahaleb seed oil (Mariod *et al.*, 2009; O'Brien, 2004). The specific extinctions K_{232} (2.723) and K_{270} (0.290) values indicate the level of the primary (hydroperoxides and conjugated dienes) and secondary (carbonyl compounds and conjugated triens) oxidation compounds, and oxidation status of edible oils. The seeds and the oil contain higher quantities of CLNA, and it is known that CLNA is reactive and might interfere with specific extinction measurements, and this situation must be taken into consideration (O'Brien, 2004). During seed storage, some oil oxidation reactions may have also taken place. There is no data in the literature for mahaleb seed oil, but for virgin olive oil, a limit K_{232} value of 2.5–2.6, and K_{270} value of 0.22–0.25 were defined in the codex standard (Codex, 2017). There is no data about mahaleb seed oil viscosity in the literature, but it was measured as 113.5 cP at 25 °C, and this is within the range of similar vegetable oils (O'Brien, 2004). The literature lacks the color values of the mahaleb seed oil, but it seems to be a yellow-greenish oil according to the measured L*, a*, and b* values (Fig. 1 and Tab. 2).

Free fatty acidity of 0.65% oleate and an acid number of 1.31 mg KOH/g were measured (Tab. 2). Mariod *et al.* (2009) found 1.21–7.86 mg KOH/g acid numbers for the mahaleb seed oil. Turkish food codex for vegetable oils indicates a limit acid value for cold-pressed oils as 4.0 mg KOH/g oil (Codex, 2012). Clearly, our sample is fairly good for these quality parameters. Similarly, the codex limits the peroxide value of cold-pressed oils to a max of 15 meqO_2/kg oil (Codex, 2012). The measured 4.29 meqO_2/kg oil peroxide value is far lower than the limit, indicating that the oxidation status of the oil was acceptable. The iodine number of 158.15 gI/100 g oil and unsaponifiable content of 0.72% was measured (Tab. 2). Sbihi *et al.* (2014) reported Syrian mahaleb seed oil iodine number as 169.2 gI/100 g oil, and unsaponifiable content as 1.3%, and these data concur with this study. Clearly, the mahaleb seed oil is an unsaturated drying oil according to the classification based on iodine numbers (O'Brien, 2004). The chemical quality indices of the cold-pressed mahaleb seed oil were within the ranges reported in the available literature for solvent-extracted mahaleb seed oils, and were within the acceptability limits of the related codex standards. This situation could be accepted as an advantage to exploring the oil as an edible source.

3.3 Fatty acid, phytosterol and tocopherol compositions of the cold-pressed mahaleb seed oil

Table 3 shows the main fatty acid, phytosterol and tocopherol compositions of the cold-pressed mahaleb seed oil. Sixteen different fatty acids were quantified (Fig. 2 and Tab. 3), but the majority occurred as CLNA (37.29%), oleic (29.84%), linoleic (26.44%) and palmitic (3.78%) acids. The oil contained a total of 5.42% saturated (SFA), 30.34% mono-unsaturated (MUFA), and 64.24% of poly-unsaturated fatty acids (PUFA). This data concurs with most of the literature (Özgül-Yücel, 2005; Ercisli and Orhan, 2008; Mariod *et al.*, 2009; Erdogan Eliuz *et al.*, 2022). The major fatty acids of mahaled seed oil were reported as 39.76, 31.33, and 23.01% of CLNA, oleic, and linoleic acids, respectively (Öztürk *et al.*, 2014). In another study (Erdogan Eliuz *et al.*, 2022), 34.39% of CLNA, 31.76% of oleic acid, 25.54% of linoleic acid, and 3.67% of palmitic acid were reported. Sbihi *et al.* (2015) even reported presence of 40.71% of CLNA in mahaleb seed oil. Clearly, fatty acid data of the present study agrees with literature. The CLNA has been linked with many health benefits including body fat mobilization, cytotoxic activity, and anticancer properties. The edible oils containing CLNA were mostly reported as functional oils (O'Brien, 2004; Ercisli and Orhan 2008; Erdogan Eliuz *et al.*, 2022). CLNA has also classified as reactive fatty acids and prone to oil oxidation, and care during storage and distribution was suggested for those oils containing higher quantities of this fatty acid (O'Brien, 2004).

Eight different phytosterols were quantified (Tab. 3), with the majority of β-sitosterol (88.6%) and campesterol (3.2%), respectively. The major portion of the tocopherols in the mahaleb seed oil (Tab. 3) was γ-tocopherol (400.34 mg/kg) and β-tocopherol (348.18 mg/kg), respectively. In one study, γ-, α-, and δ-tocopherols were quantified in the units of mg/100 g oil as 20.7, 6.4, and 1.4, respectively (Mariod *et al.*, 2009). In another study, 192.52 mg/100 g γ-tocopherol, 43.22 mg/100 g δ-tocopherol, and 25.84 mg/100 g α-tocopherol were determined (Sbihi *et al.*, 2014). It seems that the tocopherol content in this study is higher than Mariod *et al.* (2009) and lower than Sbihi *et al.* (2014). Also, β-tocopherol was present in our sample. These differences could be attributed to the material, processing, and analysis differences.

Table 3. The fatty acid composition (%), phystosterol composition (%), and tocopherol composition (mg/kg oil) of the cold-pressed mahalep seed oil (Mean ± SD).

	Mahalep oil		Mahalep oil
Myristic acid (C14:0)	0.02 ± 0.0	Beta-sitosterol	88.6 ± 3.5
Palmitic acid (C16:0)	3.78 ± 0.1	Brassicasterol	0.1 ± 0.0
Palmitoleic acid (C16:1)	0.26 ± 0.0	Delta-5-avenasterol	2.3 ± 0.1
Margaric acid (C17:0)	0.02 ± 0.0	Delta-7-stigmasterol	2.4 ± 0.2
Heptadecanoic acid (C17:1)	0.03 ± 0.0	Campesterol	3.2 ± 0.1
Stearic acid (C18:0)	1.39 ± 0.0	Cholesterol	0.1 ± 0.0
Oleic acid (C18:1)	29.84 ± 0.9	Stigmasterol	0.1 ± 0.0
Linoleic acid (C18:2)	26.44 ± 1.0	Delta-7-Avenasterol	0.7 ± 0.0
Arashidic acid (C20:0)	0.10 ± 0.0	Total	97.8
α-Linolenic acid (C18:3)	0.12 ± 0.0		
Eicosenoic acid (C20:1)	0.11 ± 0.0		
Conjugated Linolenic acid (18:3)	37.29 ± 0.8	δ-Tocopherol	77.27 ± 1.58
Behenic acid (C22:0)	0.02 ± 0.0	γ-Tocopherol	400.34 ± 13.86
Docosadienoic acid (C22:2)	0.10 ± 0.0	α-Tocopherol	87.64 ± 1.47
Lignoceric acid (C24:0)	0.09 ± 0.0	β-Tocopherol	348.18 ± 4.5
Nervonic acid (C24:1)	0.10 ± 0.0	Total	913.43
Docosahexaenoic acid (C22:6)	0.29 ± 0.0		
ΣSFA	5.42		
ΣMUFA	30.34		
ΣPUFA	64.24		

3.4 Thermal properties of the cold-pressed mahaleb seed oil

The DSC-determined thermal properties of the mahaleb seed oil sample are listed in Table 4. Fully melted oil starts crystallization at around −44.45 °C, and crystallization completes at around −46.62 °C with an enthalpy value of −39.46 J/g. Similarly, mahaleb seed oil's peak melting temperature was determined at −8.41 °C (Tab. 4). Since this oil is an oleic-linoleic type oil (Tab. 4), its melting temperature is quite low. It seems to be liquid at refrigerator temperature. There is no thermal data in the literature for mahaleb seed oil, and hence data presented in this study would be important. Sbihi *et al.* (2014) reported only thermal degradation data for this oil, indicating a degradation temperature of around 350–525 °C. Similar thermal behavior for other cold-pressed seed oils in our laboratory was published previously (Yılmaz *et al.*, 2015; Ok and Yılmaz, 2019). The thermal data reported here would be interesting to those who are interested to use this oil.

Table 4. Thermal properties of the cold-pressed mahalep seed oil (Mean ± SD).

		Mahalep oil
Crystallization	Onset (°C)	−42.09 ± 0.54
	Peak (°C)	−44.45 ± 0.26
	End (°C)	−46.62 ± 0.15
	ΔH (J/g)	−39.46 ± 0.31
Melting	Onset (°C)	−20.91 ± 0.46
	Peak (°C)	−8.41 ± 0.04
	End (°C)	−4.21 ± 0.10
	ΔH (J/g)	61.78 ± 0.15

3.5 Volatile aroma compounds composition of the cold-pressed mahaleb seed oil

The volatile aroma compounds quantified in the cold-press produced mahaleb seed oil samples are presented in Table 5. There were 38 different volatile aroma compounds. The highest mean peak area% values were determined for 1-pentanol (9.69%), n-pentanal (9.30%), limonene (3.01%), 2-methylnonane (2.75%), coumarin (2.46%), E-hexenal (2.36%), and 4-methyl nonane (2.15%), respectively. The most common aroma descriptors of these compounds were fermented, almond, fruity, citrus, sweet, hay, terpene, green, and balsamic. Since, in aroma science, the concentration of a compound itself has no meaning, higher concentrations do not mean that they contribute most to the perceived aroma. In fact, the odor (aroma) threshold value indicates how an individual compound contributes to the perceived aroma of a sample. The odor threshold is a minimum concentration value in a specific matrix, indicating that at that lowest concentration, the aroma of the compound could be perceived by human subjects. From the aroma science literature, it is well-known that some compounds produce very intense aroma sensations at low concentrations and some cannot be perceived at even higher concentrations or vice versa. Unfortunately, only a limited number of odor threshold values in specified matrices (aqueous or lipid) are known. Consequently, the perceived characteristic aroma of a food sample is rather a cumulative effect of all volatile aroma compounds present (Meilgaard *et al.*, 1991;

Table 5. The volatile aroma compounds composition of the cold-pressed mahalep seed oil (Mean ± SE).

RI[†] (min)	Volatile compound	Aroma definition[††]	Peak area (Mean ± SE)	Peak value (%)
1.382	Ethyl alcohol	Alcohol, medicine	86 605 ± 3300	0.15
1.476	Ethyl ether	Etheral	352 355 ± 13 076	0.60
1.539	Methyl acetate	Etheral	218 698 ± 27 500	0.38
1.805	n-Butanal	Pungent, green	837 570 ± 55 050	1.44
1.833	Acetic acid	Vinegar	526 116 ± 48 932	0.90
2.345	n-Butanol	Fruit, wine	465 191 ± 33 700	0.80
2.727	n-Pentanal	Almond, malt, pungent	5 421 768 ± 10 300	9.30
3.345	3-Methyl-1-butanol	Fermented, fruity	161 485 ± 8750	0.28
3.412	1-Butanol, 2-methyl	Ethereal, alcoholic, fatty, greasy, cocoa	128 682 ± 56 700	0.22
3.964	1-Pentanol	Pungent, fermented, yeasty	5 651 374 ± 49 927	9.69
4.690	E-2-Hexenal	Green, banana, aldehydic	1 378 654 ± 12 300	2.36
5.446	Formic acid, pentyl ester	Sweet fruity, fresh, cognac	120 974 ± 5505	0.21
5.636	2-Ethyl Hexanol	Citrus, fresh, floral, oily	95 722 ± 3872	0.16
6.743	n-Hexanol	Pungent, etherial, fusel oil, green	726 270 ± 6005	1.25
7.400	2-Butylfuran	Faint, fruity, wine-like	195 724 ± 17 540	0.34
7.550	n-Hexanoic acid	Sour, fatty, cheese	226 044 ± 7450	0.39
8.256	Isoamyl acetate	Sweet fruity, banana, solvent	253 879 ± 14 000	0.44
8.875	*trans*-Sabinene hydrate	Woody, balsam	71 493 ± 580	0.12
9.995	4-Methylnonane	Sweet, balsamic	1 251 562 ± 1050	2.15
10.367	3-Methylnonane	Fruity, almond	1 604 384 ± 1809	2.75
10.511	2-beta-Pinene	Woody, pine	369 740 ± 12 900	0.63
11.215	n-Hexanoic acid	Cheesy, sour	85 824 ± 3476	0.15
11.385	Cyclopentane, isopentyl-	Floral	79 310 ± 1987	0.14
11.641	alpha-Thujene	Woody, green, herb	506 032 ± 21 040	0.87
11.741	Delta-3-Carene	Citrus, terpenic, herbal, pine	933 126 ± 18 780	1.60
12.350	*p*-Cymene	Harsh chemical, woody and terpy-like	1 305 242 ± 52 000	2.24
12.543	Limonene	Citrus, lemon, green	1 756 408 ± 1020	3.01
12.740	Benzyl alcohol	Sweet, floral	561 409 ± 1201	0.96
12.827	n-Tetradecane	Mild waxy	636 865 ± 875	1.09
12.975	2-Hexyl-1-octanol	Waxy	399 764 ± 1735	0.69
13.374	2(3H)-Furanone, 5-ethyldihydro-	Sweet, creamy, lactonic	64 923 ± 19 700	0.11
15.535	n-Hexanal	Grass, fatty	100 532 ± 11 070	0.17
15.985	Heptane, 1,1’-oxybis-	Sweet, etheric	59 254 ± 5300	0.10
17.876	Docosane, 5-butyl-	Waxy	58 075 ± 4761	0.10
19.734	*Trans, trans*-Nona-2,4-Dienal	Fatty, melon, waxy, green	71 344 ± 9850	0.12
20.680	Cuminic aldehyde	Spicy, green, cumin-like	74 179 ± 9500	0.13
20.761	2-Methyl-5-isopropenyl-2-cyclohexenone	Minty licorice	91 806 ± 3815	0.16
27.377	Coumarin	Sweet, hay, tonka	1 431 389 ± 1370	2.46

[†]RI (Kovat Index) on HP 5 MS column.
[††]Aromatic definitions of the volatile compounds are found from the web pages of http://www.thegoodscentscompany.com/index.html#

Ok and Yılmaz, 2019). From the most frequently occurring aroma definitions of the volatile compounds listed in Table 5, it would be claimed that mahaleb seed oil is something like pungent, almond, green, fruity, citrus, and woody. Clearly, it is an aromatic-rich oil. Further, these aromatics definitions are usually in agreement with the panel-determined sensory descriptive terms (Tab. 6) for this oil.

As far as we reached, there is no study in the literature on the volatile aroma compounds composition of mahaleb seed oil. The aroma, sugar, and anthocyanin profile of black mahaleb fruit and seed were published (Öztürk *et al.*, 2014). After comparing the aromatic profiles, it was observed that 3-methyl-1-butanol, n-hexanol, benzyl alcohol, and coumarin were common in their black mahaleb fruit and in our oil sample. Similarly, common volatile aroma compounds of this study (Tab. 5) and their seeds were 1-hexanol, benzyl alcohol and coumarin. In a recent study (Dadalı, 2022), the volatiles of the mahaleb kernel, sour cherry, and apricot kernel were analyzed and compared to determine the adulteration of mahaleb kernel with the other kernels. Mahaleb kernel contained 25 volatiles, including 1 acid, 2 alcohols, 3 aldehydes, 1 furan, 2 ketones, and 16 terpenes. Coumarin was

Table 6. Sensory descriptive properties of of the cold-pressed mahalep seed oil (Mean ± SD).

	Mahalep oil
Almond	5.5 ± 0.5
Vanillin	1.8 ± 0.2
Dough	3.0 ± 0.7
Green	2.2 ± 0.8
Cooling	1.8 ± 0.5

Table 7. Consumer test results of of the cold-pressed mahalep seed oil (Mean ± SD).

	Mahalep oil
Appearance	4.49 ± 0.13
Smell/Aroma	3.56 ± 0.10
Taste/Flavor	3.50 ± 0.11
General acceptance	3.70 ± 0.11

determined as the main volatile, and it was also present in our sample (Tab. 5) with 10 other common volatiles. In another recent study (Dadalı and Elmacı, 2022), the headspace solid-phase microextraction technique was optimized, and 21 volatile aroma compounds from terpene, aldehyde, ketone, and alcohol chemical classes were determined from mahaleb kernels. The common volatile aroma compounds in our oil sample and their kernel sample were β-pinene, sabinene, 3-carene, limonene, β-thujene, 2,4-nonadienal, benzyl alcohol, cumin aldehyde, and coumarin. It must be kept in mind that the volatile aroma compounds in this study are from the cold-pressed seed oil, while theirs were from fruits and/or seeds (kernels). During seed pressing, cell disruption might have caused some reactions to change the volatiles present. Still, the oil and seed had quite similar distinct volatile aroma compounds. These distinct aromas make this oil special gourmet edible oil. Overall, the volatile aroma compound composition data provided in this study for the cold-press produced mahaleb seed oil could be an important contribution to the literature.

3.6 Sensory descriptions of the cold-pressed mahaleb seed oil

The panel defined the mahaleb oil sample with 5 sensory descriptor terms and the mean data generated by the QDA test is summarized in Table 6. Obviously, the most dominant attribute was "almond" with 5.5 mean score on 10 max score indicating the highest intensity. In previous sections, the oil had volatile aroma compounds (Tab. 5) with almond or similar definitions (n-pentanal, 3-methylnonane, coumarin). "Dough" was defined for this oil by the panel with 3.0 score, the second highest value among all. It could also be related with sour and/or fermented aroma notes. The volatiles (Tab. 5) ethyl alcohol, 3-methyl-1-butanol, 1-pentanol, and 2-butylfuran could be related with the dough descriptor. Similar to most cold-pressed and virgin oils, mahaleb seed oil also had the "green" descriptor (Yılmaz *et al.*, 2015; Ok and Yılmaz, 2019). The volatiles n-butanal, E-2-hexenal, n-hexanol, alpha-thujene, and n-hexanal were defined with green or grassy aroma notes, and these compounds were well perceived by the panel. The panel also defined "vanillin" and "cooling" descriptors with a 1.8 score each. The interaction and combination of the 38 volatile aroma compounds could have created a vanillin like sensation. "Cooling" is the sensation felt in the mouth space, mostly related to mint-like or rapid evaporating compounds. The 2-Methyl-5-isopropenyl-2-cyclohexenone compound was defined as minty licorice could be responsible for the cooling sensation.

There is no sensory study in the literature for mahaleb seed oil. These results could provide important additions to the knowledge about it. In one of our previous studies (Yılmaz *et al.*, 2020), sensory evaluations were determined for the cold-press generated cherry and sour-cherry seed oils. Since the plants are from the same subfamily, there would be some similarities. The sensory definition terms common in both studies were almond and cooling (menthol). Overall, cold-pressed mahaleb seed oil was an aromatic-rich oil to be used in various food formulations.

3.7 Consumer tests of the cold-pressed mahaleb seed oil

The results of the consumer tests are shown in Table 7. With the 5-point hedonic scale, the highest score for this oil was for appearance (4.49), followed by general acceptance (3.70). Since all attributes (appearance, smell/aroma, taste/flavor, general acceptance) got scores above the neutrality point (neither like-nor dislike of 3.0 score), it could be claimed that this oil would be accepted by the consumers. Some further processes (pre-roasting, pre-steaming, microwave treatment of seeds, etc.) could be applied to enhance the consumer hedonic scores of the cold-press produced mahaleb oil. Mariod *et al.* (2009) evaluated solvent-extracted white mahaleb seed oil with 10-point hedonic scale and found 8.7, 8.5, and 8.0 scores for color, odor, and taste, respectively. These findings concur with ours that the appearance and/or color of the oil was liked most.

4 Conclusions

In this study, cold-press produced mahaleb seed oil was characterized to extend the food and functional food applications of this special oil. Although literature reports some data about solvent-extracted mahaleb seed oils, this study provides the first data about cold-pressed samples. Further, some physico-chemical data (color, viscosity, thermal properties etc.), sensory data, and volatile aroma compounds composition data were reported for the first time in the literature for this oil. The oil has been consumed as an edible oil in some local regions of Turkey, and it is known to be safe and food-grade. Especially, cold-pressing would provide beneficial oil samples as functional foods or functional ingredients for processed foods since they contain many bioactive and volatile compounds intact. It is expected that edible applications of this oil will be extended and/or new studies about food applications will be studied. The oil seems rich in unsaturated CLNA, oleic and linoleic fatty acids, β-sitosterol, and tocopherols. The distinct almond-like and fermented-like vanillin aroma notes would be the main reason to prefer this oil for some food formulations. In addition, the typical aroma and flavor of the oil could be well preferred for some bakery products, confectionaries, and others. Consumers liked this oil, but care (neutral gas atmosphere or vacuum) must be given to control oil oxidation during production, storage, and distribution. More studies about food applications will be anticipated.

Conflicts of interest

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Data availability

All data generated or analyzed during this study are included in this published article and its supplementary information files.

Code availability

Not applicable.

Authors' contributions

Emin Yilmaz: Conceptualization, Data curation, Investigation, Methodology, Project administration, Resources, Supervision, Validation, Writing–original draft preparation, Writing–review and editing. Burak Karatas: Formal analysis, Investigation, Methodology, Project administration, Software, Validation and Visualization.

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