

CHEMICAL CHARACTERISTICS OF THE SEED OF IRANIAN ENDEMIC PLANT *Kelussia odoratissima*

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Iran is considered one of the major centers of plant biodiversity in the world. Iran's natural environment is very diverse, ranging from tropical to cold temperate. This ecological diversity has contributed not only to a high genetic diversity, but has also allowed the successful introduction and cultivation of a great number of plant taxa [1]. *Kelussia odoratissima* Mozaff. belonging to the *Apiaceae* family, is a robust, erect, glabrous, perennial aromatic herb, which grows to a height of 120 to 200 cm; the flowers are 1–2 mm in diameter, all hermaphrodites, and the petals are yellow and produced in compound umbels. It is native to Zagros (central region of Iran). Because of the large-scale use of the herb of this wild plant, it is widely cultivated. It is a sweet-smelling, self propagating plant, which is traditionally consumed in Iran as an anti-inflammatory, sedative, anti-tissue, and garnish. The seeds have long been used as a remedy for various ailments, particularly to treat head cold and stomachache [2, 3]. Some investigators have reported the essential oil composition of leaves [4] and antioxidant activity [5] of *K. odoratissima*.

K. odoratissima is an Iranian endemic plant; there is no report on the plant seed characteristics. The aim of this investigation was to study the characteristics and fatty acid, essential oil, total phenolic content, and minerals of the seed of *K. odoratissima* Mozaff. growing in Iran.

The total phenolic content of the *K. odoratissima* seeds was 218/15 mg GAE/g DW (Table 1). Results of fatty acid (yield 25%) composition of unsaturated (91/44%) and saturated fatty acid (5.55%) and total phenolics clearly showed that *K. odoratissima* seeds could be rich sources of phenolics. The essential oil isolated by hydrodistillation from the seeds of *K. odoratissima* was found to be a pale yellow liquid and was obtained in a yield of 2.1% w/w based on dry weight (Table 1).

The fatty oil yield of the samples was found in this investigation to be 25%. Results showed the presence of five dominant fatty acids, including: palmitic (6.65%), stearic (1.9), petroselinic (72.35), linoleic (19.14), and linolenic acids (0.95%). It is well known that petroselinic and linoleic acids are two essential fatty acids that humans require.

Table 1 presents the chemical composition of the essential oil. Out of 33 identified compounds, the major compounds were: Z-ligustilide (50.99%), δ -terpinen-7-al (10.28%), δ -terpinene (5.29%), cuminaldehyde (5.17%), 3Z-butylidene phthalide (4.94%), germacrene D (3.95%), β -pinene (3.03%), and bicyclogermacrene (2.05%). The other constituents present in more than 1% were: α -terpinen-7-al (1.85%), p -cymene (1.63%), β -phellandrene (1.43%), E-ligustilide (1.21%), and δ -amorphene (1.09%). It is possible to use *K. odoratissima* seed as a potential source of Z-ligustilide.

The mineral content of *K. odoratissima* is as follows: N, P, and K values of the seeds were 5.81%, 0.37%, and 0.33%, respectively. According to our data, Ca, Mg, and S were 1.86%, 0.56%, and 0.28%, respectively; Fe, Mn, Zn, and Na contents were 81 ppm, 28 ppm, 32 ppm, and 11 ppm, respectively.

In conclusion, the present study showed that *K. odoratissima* seeds were an extremely rich source, in particular, of phenolics, fatty acids, essential oil, and some minerals.

Plant Material. Seeds of the plant were collected in August, during the ripening stage, from Zagros Mountain of Feraidon Shahr in Isfahan province.

Extraction of Seed Fatty Oil. Fatty oil was extracted from 10 g of seeds using hexane solvent in a Soxhlet apparatus for 6–7 h, and removal of the solvent by a rotary evaporator. Oil content was determined by calculating the difference in dry weight of seed samples before and after the extraction [6].

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TABLE 1. Chemical Composition of the Seed of *K. odoratissima* Essential Oil

Compound	RI	%	Compound	RI	%
Sabinene	969	0.18	α -Guaiene	1478	0.4
β -pinene	977	3.03	Germacrene D	1486	3.95
Myrcene	981	0.21	β -Selinene	1493	0.27
α -Phellandrene	1001	0.16	α -Selinene	1501	0.33
ρ -Cymene	1015	1.63	Cuparene	1505	0.17
β -Phellandrene	1025	1.43	Sibirene	1508	0.28
δ -Terpinene	1052	5.29	δ -Cadinene	1515	0.43
Pentylcyclohexa-1,3-diene	1153	0.32	δ -Amorphene	1521	1.09
Cumin aldehyde	1218	5.17	Bicyclogermacrene	1536	2.05
α -Terpinen-7-al	1264	1.85	Germacrene B	1564	0.41
δ -Terpinen-7-al	1271	10.28	3-Butylphthalide	1619	0.5
Citronellyl acetate	1332	0.17	3Z-Butylidene phthalide	1647	4.94
α -Copaene	1384	0.42	Sedanolide	1693	0.8
β -Elemene	1394	0.75	Z-Ligustilide	1718	50.99
<i>E</i> -Caryophyllene	1429	0.13	<i>E</i> -Ligustilide	1772	1.21
δ -Elemene	1434	0.85	Folic acid	1864	0.2
α -Humulene	1461	0.13			

Determination of Fatty Acid Composition of the Seed Oil by Gas Chromatography. The extracted seed oil was methylated using the common procedure by refluxing with methanolic sodium hydroxide solution, boron trifluoride reagent, and heptane [6]. The fatty acid composition of the esterified oil was characterized and quantified using a Unicam 6400 Chromatograph. For this purpose a BPX 70 (30 m $\times$ 0.25 mm) column was used. The injection volume was 10 μ L. The injector and detector temperatures were 300°C and 350°C, respectively.

Determination of Total Phenolic Content. The phenolic content of seeds was determined using the method described previously by [7]. Briefly, aliquots of 0.1 g of lyophilized powder of fruit samples were dissolved in 1 mL of deionized water. The solution (0.1 mL) was mixed with 2.8 mL of deionized water, 2 mL of 2% sodium carbonate (Na₂CO₃), and 0.1 mL of 50% Folin-Ciocalteu reagent. After incubation at room temperature for 30 min, the absorbance of the mixture was measured at 750 nm against a deionized water blank on a spectrophotometer (Jenway, 6505). Gallic acid (GA) was chosen as a standard. Using a curve derived from six points (0, 40, 80, 120, 160, and 200 mg/L) of standard, total phenolic content in the seeds was determined and the results expressed as mg gallic acid equivalents (GAE) g⁻¹ dry weight (DW).

Isolation of the Essential Oils. Shade-dried seeds of *K. odoratissima* (50 g three times) were subjected to hydrodistillation for 4 h using a Clevenger-type apparatus to produce oil according to the method recommended by the European pharmacopoeia [8]. The oil was dried over anhydrous sodium sulfate and stored in sealed vials at low temperatures before analysis.

Determination of Nitrogen and Mineral Elements. Total N was determined by the Kjeldahl method. To determine the mineral composition of the fruits, samples were burned with a nitric acid solution, on a hot plate, at 200°C. Then, the absorbance of the extract was measured by an atomic absorbance spectrophotometer (Shimadzu, aa-670). The amounts of minerals were calculated by comparing with the standard curve of each element. The phosphorus content of the extract, however, was analyzed by determining the absorbance of the yellow solution obtained from the Barton reaction, using a spectrophotometer (Jenway, 6505) at 680 nm, and comparing the results with the standard curve [9].

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