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Original Article

Fatty Acid Composition of *Lycium* (Solanaceae) Species

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Abstract

Background and Aim: There are more than one hundred species of *Lycium* (Solanaceae) in the arid and semi-arid regions of the world. These drought-resistant medicinal plants have several applications in traditional medicine and modern pharmacy. The fruits of these plants contain vitamins A, E, and C, flavonoids, and other valuable biological compounds. In the present study, the amounts and types of fatty acids in the fruit pulp of *Lycium* species have been determined.

Materials and Methods: The samples were collected from natural habitats in Iran. Cold methylation and gas chromatography were used to extract fatty acids. By examining the drawn curves, fatty acids were identified. The significance of the quantitative results was assessed by a one-way analysis of variance.

Results: A total of 22 major fatty acids were observed in the fruit pulps of *Lycium* species. The highest diversity of fatty acids was observed in *L. kopetdaghi*. In 4 species of *L. shawii*, *L. edgeworthii*, *L. ruthenicum*, and *L. depressum*, cis-linoleic is the major and abundant fatty acid, and only in *L. kopetdaghi*, the palmitic acid is more abundant.

Conclusion: The findings of the present study are in agreement with previous research. Due to the composition of valuable fatty acids in *Lycium* species, further research is recommended for its application in the pharmaceutical and cosmetic industries.

Keywords: *Lycium,* Box-thorn, Fatty Acid content, Iran

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Introduction

The *Lycium* L. (Solanaceae) with more than a hundred species is mainly distributed in tropical and subtropical parts of the world. It is a fast-growing and drought-resistant shrub. It has proper flowering in well-drained and moderately textured soils and is resistant to temperatures up to -15 ºC. These medicinal plants have several uses in traditional Chinese medicine in the treatment of heart and eye disorders, and strengthening the kidneys and liver (1). The leaves of *L. depressum* stocks have several healing properties. It balances the parasympathetic nervous system. Moreover, it has antispasmodic properties, and is used in the treatment of spasmodic coughs, throat irritations, severe gastric contractions, tuberculosis-induced transpiration, painful menstruation and prostate discomfort. The fruits of *Lycium* contain vitamins C, A, E, flavonoids and biological compounds such as polysaccharides and proteins, and are a good source of fatty acids, and if included in the diet, can prevent cancer or stop the growth of cancer (2-4). *L. barbarum*

L. has various biological effects in traditional Chinese medicine and is known to increase immunity, protect against liver damage, and reduce the side effects of chemotherapy and radiation therapy (5). In some parts of the world, *Lycium* fruit is used to make medicinal products, food supplements, beverages, and decoctions.

Many valuable compounds, including polysaccharides (6), glycosides (7), amino acids (8), phenolic compounds (9), and immunologically active pectins (10) have been extracted from different *Lycium* species. Many of them show antioxidant properties. Antioxidant and antimicrobial properties have been reported for Lycium shawii (11).

The fruit of *Lycium barbarum* is commonly used to reduce fat and blood sugar. Yu *et al*. (2005) pointed to the anti-aging effects of *Lycium barbarum* to treat aging diseases such as Alzheimer's by protecting neurons (12). The study of phytochemistry and nutritional factors of *Lycium ruthenicum* showed that the fruit has the highest percentage of protein and crude fat, and also the highest antioxidant properties compared to roots and leaves (13).

Some studies have recently investigated the chemical composition and the bioactive substances of different *Lycium* species. *Lycium barbarum* is rich in antioxidant content and its extracts have biological activities, including anti-aging, anti-tumor, immune-stimulatory, and cytoprotecting activities. Endes *et al*. (2015) investigated the fatty acid contents of *L. barbarum* fruits and found that palmitic, oleic, linoleic acids were major fatty acids (14). Boulila and Bejaoui (2015) pointed to an unexploited and rich source of unsaturated fatty acids in some *Lycium* species (15). Montesano *et al*., (2018) studied the chemical features of *L. barbarum*. They found high levels of unsaturated fatty acids in certain oleic and linoleic acids (1).

As there is no available information on fatty acids of Iranian *Lycium*, the present study was conducted to to extract and determine the type and composition of fatty acids of fruit pulp in *Lycium* species to expand the available knowledge on their nutraceutical and medicinal values.

Materials and Methods

Sampling

To provide proper samples in order to study the fatty acid composition and type, fruits of different species were gathered from nature in the growth period from five natural habitats (Table 1).

Fatty Acid Extraction

We followed the procedure developed by Folch *et al*. (1957) (16). Totally, 20 g of fruit pulp samples were weighted into screw-tap glass bottles then chopped, and 50 ml of methanol was added and stirred for 30 minutes. About 40 ml of hexane was added and thoroughly vortexed for 5 min. By using centrifugation at 1100 rpm for 5 min, the upper phase was separated and used for the methylation process.

The standard blend of fatty acids (2, 5, 10, 25 and 50 mg/ml) was prepared by methylation similar to the sample preparation. To extract fatty acids, a cold methylation method with 2 M alcohol potassium solution was used. Two drops of oil were mixed with 5 ml of normal heptane and then 3 ml of 2 M potassium alcohol (methanol) was added to the mixture and stirred for 5 minutes.

The resulting mixture was kept stationary for a while until the upper phase became clear. Subsequently, 0.5 μl of the supernatant was injected into the GC model (6100) Young Lin. The same condition was used for the injection of standards. Analysis of fatty acids parameters was carried out by gas chromatography with SGE-BPX70 capillary column (60 cm * 0.25 mm. * 0.25 μm).

Temperature gradient ranged from 150 to 190 degree centigrade with a 5-degree incretuin per minute and preservation till the ende of the process (total analysis time was equal to 40 minutes). The type of carrier gas was hydrogen with a flow rate of 1ml per minute.

The conditions of the GC device were as follows:

Injector temperature: 280°C

Detector temperature: 300°C

SGE column temperature (BPX70): 150°C for 5 minutes and then 190°C for 30 minutes

Hydrogen transport gas flow rate: 1 ml/min

Fatty Acid Detection

After injecting the tested samples into the device, comparing the retention time of each sample, and comparing the curves drawn for standard fatty acids with the resulting curves, the fatty acids of the samples were identified. Each fatty acid was determined as the peak number, and its percentage in total fatty acids, and

Table 1: Details of the Samples Studied in the Present Project (Vouchers are deposited at the herbarium of Alzahra University (ALUH)).

Taxon	Locality Detail	Herbarium no.
L. depressum Stocks	Khorassan, east of Rashtkhar, between Saadat Abad and Yahoo old-Grmb	23485
L. ruthenicum Murray	Khorassan, south of Sabzevar, between Daulatabad and Hares Abad	38881
L. edgeworthii Miers	Hormozgan, between Minab and Bandare Abbas, Hasan langi-ye Bala	15
L. shawii Roem. & Schult.	Khuzestan, Mollasani	1
L. kopetdaghi Pojark.	Khorassan, Daregaz, Tandoureh Protected Area, Alibolagh forest	45284

the time of leaving the column were also determined. Based on the type and number of peaks, the type of each fatty acid was determined. Figure 1 indicates the chromatogram of standard fatty acids, and Figure 2 shows the chromatogram of fatty acids extracted from fruit pulp of the investigated species. The significance of the quantitative results was assessed by one-way analysis of variance test by SPSS 20 software.

Results and Discussion

The amount of oil obtained from the fruit pulp of 5 species of *Lycium* has been reported in Table 2. Based on the results of fatty acid analysis by gas chromatography, 22 major fatty acids were observed in *Lycium* fruit pulp, of which 12 were unsaturated and the rest were saturated fatty acids. Among the studied species, *L. kopetdaghi* had the highest amount of many fatty acids (Table 2), but for example, *L. ruthenicum* had the highest amount of linoleic acid, and *L. edgeworthii* had the highest amount of only three fatty acids compared to other species (arachidonic acid, nerolic and lignoceric acid).

In general, the number of fatty acids in the studied species of *Lycium* showed a difference, and the amount of unsaturated fatty acids was significantly different. A comparison of the profiles of the investigated species revealed that in at least 4 species of *L. shawii*, *L.*

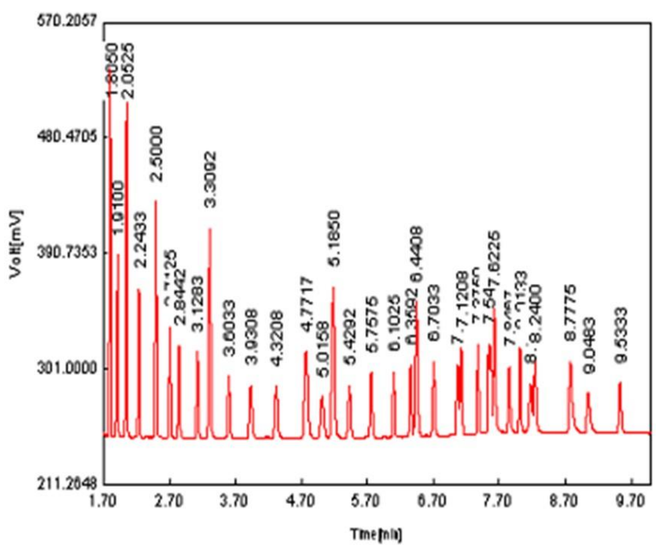

Figure 1. Gas Chromatographs of Standard Fatty Acids Used in the Present Study.

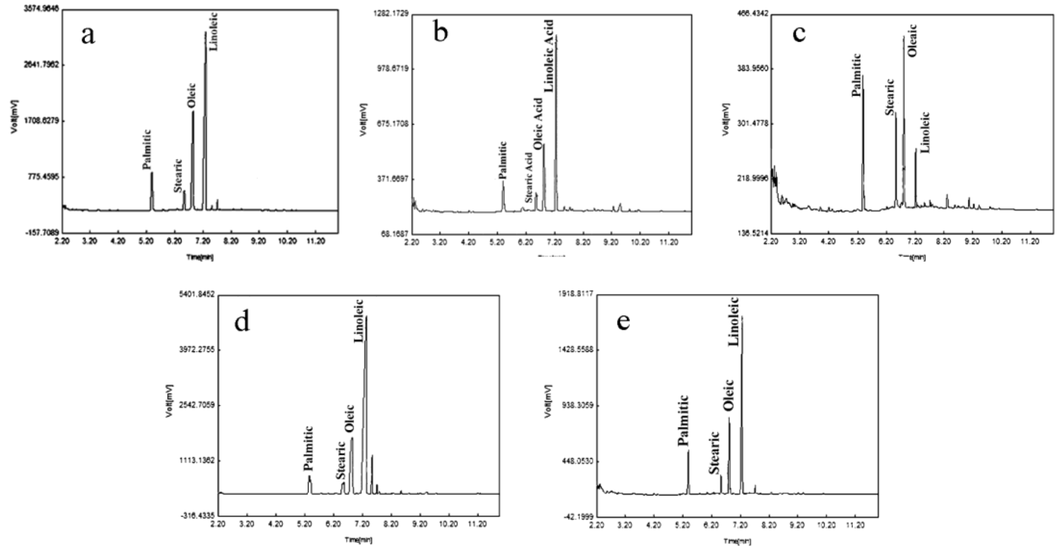

Figure 2. Gas Chromatographs of Fruit Pulp Fatty Acids Contents in a) *Lycium depressum,* b) *L. edgeworthii,* c) *L. kopetdaghi,* d) *L. ruthenicum,* e) *L. shawii* (mV stands for millivolts, Time in minutes)

edgeworthii, *L. ruthenicum*, and *L. depressum,* cis-linoleic was the major and abundant fatty acid, and only in *L. kopetdaghi* species, the palmitic fatty acid was more abundant.

Fatty acids are long chains of carboxylic acids found in nature in both saturated and unsaturated forms. The nutritional value of unsaturated fatty acids is higher and carries fewer risks (17). Providing proper information about the fatty acid's composition is very important in the specification of the health benefits of food. In this study, the fatty acids of *Lycium* medicinal plants were studied, and the results revealed that *L. kopetdaghi* had a higher percentage in most fatty acids than other species. Most species except *L. kopetdaghi* have a high percentage (about 50%) of cis-linoleic acid. This finding is in accordance with that of Nzeuwa *et al*. 2020; Boulila & Bejaoui (2015), Endes *et al*., (2015) and Montesano *et al*., (2018). *L. shawii* is the only species among the studied ones with omega3-trans fatty acid. It is evident that linoleic acid as a significant fatty acid is the precursor of arachidonic acid biosynthesis, the substrate for eicosanoid synthesis (15). This fatty acid is used in many cosmetics products. The same fatty acid is a valuable industrial and pharmaceutical one in human nutrition.

Cantarelli et al. (1993) reported a high level of palmitic acid and stearic acid as well as linoleic acid and oleic acid unsaturated fatty acids in tomato seed oil (18). Feki et al. (2013) studied the seed oils of *Solanum elaegnifolium* (Solanaceae) and found unsaturated fatty acids, including linoleic acid, oleic acid, and saturated fats such as palmitic acid and stearic acid (19). The results of this study are in agreement with the records of these researchers. The high content of unsaturated fatty acids in species studied proposes their probable use as good sources of omega 6 with antiallergic and anti-inflammatory properties.

Different values of fatty acid profiles in the investigated species may be the result of different enzymatic activities according to the geographical conditions. Hence, it is necessary to evaluate the degree of stability of these factors in the species that were studied. Among the fatty acids extracted from these plants, valuable types were obtained, especially unsaturated ones, which can be considered in future pharmaceutical and nutritional research. Due to the extensive use of this plant in traditional medicine and the presence of useful and valuable fatty acids, plant cultivation is recommended to extract fats for medicinal and cosmetic uses such as shampoo and soap.

Table 2: Fatty Acid Composition (% mol) of Seed Oil in *Lycium* the investigated Species (mean values ± SD, *n*=5).

Fatty Acid	Sat (S)/ Unsat (U)	*L. depressum*	*L. edgeworthii*	*L. kopetdaghi*	*L. ruthenicum*	*L. shawii*
Myristic Acid	U	0.11±0.6	0.06±0.1	0.87±0.2	0.03±0.4	0.21±0.5
Palmitic Acid	S	9.31±0.8	11.25±0.1	30.39±0.3	5.49±0.6	12.43±0.2
Palmitoleic	U	0.22±0.1	0.53±0.3	1.19±0.5	0.23±0.1	0.38±0.2
Margaric Acid	S	0.33±0.2	0.17±0.6	0.38±0.2	0.16±0.1	0.69±0.4
Heptadecanoic Acid	S	0.39±0.1	0.11±0.2	0.62±0.1	0.02±0.4	0.33±0.3
Stearic Acid	S	5.06±0.5	6.90±0.4	17.62±0.6	3.97±0.1	3.67±0.2
Oleic Acid-Trans	U	0.09±0.2	0.11±0.3	1.49±0.4		0.40±0.1
Cis-Oleic Acid	U	25.18±0.1	23.15±0.3	29.67±0.4	19.6±0.3	18.24±0.2
Linoleic Acid-Trans	U	0.40±0.3	0.27±0.1	0.22±0.1	0.05±0.2	0.39±0.1
Cis-Linoleic Acid	U	55.35±0.1	49.39±0.2	8.82±0.2	68.3±0.1	58.07±0.2
Omega3-Trans	U					0.10±0.1
Omega3-cis	U	1.13±0.2	1.15±0.3	0.62±0.2	0.28±0.2	1.45±0.4
Arachidic Acid	S	0.48±0.2	1.33±0.2	0.74±0.4	0.74±0.3	0.31±0.2
Gondoic Acid	U	0.24±0.1	0.36±0.1	0.39±0.2	0.03±0.1	0.18±0.3
Behenic Acid	S	0.24±0.1	0.51±0.1	0.94±0.2	0.30±0.2	0.13±0.2
Erucic Acid	U	0.13±0.1	0.03±0.1		0.07±0.1	0.38±0.2
Cis-13,16-Docosadienoic Acid	S	0.14±0.2				0.51±0.1
Lignoceric Acid	S	0.12±0.1	0.54±0.2	0.50±0.2	0.13±0.1	0.14±0.3
Capric Acid	S	0.13±0.2		1.97±0.4	0.01±0.1	
Lauric Acid	S	0.08±0.2	0.28±0.1		0.02±0.1	
Nervonic Acid	U	0.06±0.3	0.75±0.1	0.21±0.1		
Docosahexaenoic (DHA)	U				0.09±0.1	

Conclusion

The predominant fatty acids in the fruits were oleic acid and linoleic acid, which are very similar to other solanaceae members. High levels of unsaturated fatty acids may indicate the potential value of these compounds for pharmaceutical and health purposes. This research provides valuable information in the determination of the potential of the *Lycium* species berries to be commercially applied for nutraceutical uses and formulations of edible materials for human health. The fatty acid capacity of these native plants in the country can be used in food after control tests.

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Conflict of Interest

The authors declare that they have no conflict of interest.

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