

Examining the Effect of Different Solvents on Fatty acid Profile and Nutritional Properties of Oil Extracted from TWO Wild almond Species of *Amygdalus Scoparia* and *Amygdalus Lycioides*

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Abstract

In the present study, we examined the effect of changing different solvents in the process of extracting oil from two species of wild almond: *Amygdalus Lycioides* (A.Ly) and *Amygdalus Scoparia* (A.Sc). The effect of these changes on the profile of fatty acids and their nutritional value was further investigated. These almonds were collected from some regions of Kohgiluyeh Province, Iran. The level of total unsaturated fatty acids (TUFA) in the extracted oils was more than 89%, which included oleic acid and linoleic acid. The highest level of unsaturated fatty acids belonged to palmitic acid and stearic acid. In the present study, three types of solvents, including hexane, diethyl ether, and petroleum ether, were used in the process of oil extraction from almond kernels. The effect of these solvents alone and the interaction effect on the extraction process of fatty acid profiles were examined. The nutritional quality indices such as atherogenic (AI), thrombogenic (TI), and hypocholesterolemic:hypercholesterolemic FA ratio (HH) were also calculated in different solvent extraction procedures. The process of extracting oil from almond A.Ly with the help of petroleum ether (100%) solvent showed the lowest amount of AI (0.05) and TI (0.15). In this regard, the highest amount of HH (17.32) was observed in the same extraction process. In the present study, the best solvent for the extraction of saturated and unsaturated fatty acids was identified and the iodine value was determined for each process. Selecting a suitable solvent for the selective extraction of saturated and unsaturated fatty acids and specifying indices for assessing the nutritional value of oils may be conducive to producing high-quality almond oil in the future.

Keywords; Fatty acids, *Amygdalus*, Oil extraction, Nutritional Properties

Introduction

Almond oil has long been used as a supplement in medicine owing to its numerous benefits. It is also used to prevent cardiovascular diseases (CVD), inflammation, clots, and rheumatism [1-3]. Almond oil was primarily utilized in the ancient civilizations of India, China, and Greece thanks to its many advantages for health and beauty [4-6].

Extensive studies have been conducted on almonds in different parts of world. Their levels of fatty acids, phenolic content, and their general physicochemical properties have also been studied. By investigating and comparing different properties of almonds, we can specify the geographical areas and climatic conditions suitable for the growth of specific species of almonds. [7]. Čolić et al. in a study on almonds grown in Serbia, their physicochemical data and oil extraction percentage and phenolic content demonstrated these regions were suitable for almond growth compared to standard regional conditions. Since almond oil is rich in essential fatty acids of oleic acid and linoleic acid, it has long been seriously considered in aromatherapy. Also, its moisturizing, penetrating, and restorative properties, it has been widely applied in the cosmetics industry. Moreover, the obtained results in many studies on uses and properties of almond oil as medication revealed markedly which have gigantic benefit in terms of minimizing scars and improving skin color [8].

Almond oil is mainly extracted from oilseeds by use of solvents. In a study carried out on A.SC kernel oil, it was found that the ratio of unsaturated fatty acids to saturated fatty acids was significantly higher (7.5). It was also found that the percentage of saturated fatty acids in A.SC almond oil was lower than that in olive oil. The iodine value (IV), which indicates the unsaturation degree of oil, was higher for the olive oil extracted from A.SC kernel compared with typical olive oil [9]. The percentage of fatty acids extracted from different oils varies depending

on the type of technology, extraction process, use of different solvents, fermentation, and debittering processes. For example, in a study conducted on *L.albus*, major changes were observed in nutrients, including fatty acids, after the debittering process. MUFA (mono unsaturated fatty acid) and PUFA (poly unsaturated fatty acid) levels significantly changed before and after the debittering process [10].

In line with these studies, regarding the effect of fermentation process on the level of fatty acids extracted from different oils, a study reported that the samples of white and brown rice were affected by the fermentation process with *Lactobacillus paracasei*. The level of fatty acids in the samples of fermented rice samples was compared with control rice samples, and it was observed that the levels of oleic acid and linoleic acid decreased during the fermentation process. In all samples, the level of linoleic acid was higher than that of oleic acid [11]. Solvents are commonly used in the extraction process. The use of subcritical fluids in the oil extraction process may be a useful technology and may lead to the production of higher quality almond oils in the future [12].

The recent methods for determining the level of fat and fatty acid composition in plant and animal tissues mostly require solvent extraction. In a study conducted by Klaus in 1997, fat extraction and fatty acid profiles changed in different extraction conditions. It has been reported that ASE (accelerated solvent extraction) is a promising method for extracting fat from all plant and animal tissues [13]. Another study showed that green solvents such as ethanol and methanol or alkanes such as heptane and hexane were more commonly used in the extraction process. Due to environmental hazards, these solvents are preferred over dioxane, acetonitrile, acids, formaldehyde, and tetrahydrofuran. It has further been observed that in pressurized liquid extraction (PLE), a wide range of organic solvents are used to extract bioactive substances, such as compounds found in foods and plants [14]. A growing number of studies are conducted in the area of pathologies such as CVD, obesity, and diabetes due to the alert status of diet in our community. Therefore, there is a great interest in healthy natural products that can prevent such diseases. In 2018, Pazotero used the (PLE) method to obtain fatty acids from the brown alga *Laminaria*. This extraction process was performed using four solvents with different polarities (hexane, ethyl acetate, ethanol, and an equal combination of water and ethanol). Nutritional quality of lipids in all extracts extracted by PLE method was assessed with regards to the ratio of fatty acids $\omega 6/\omega 3$. The best (lowest) index for the extract extracted with ethanol (4.4) was $\omega 6/\omega 3$ [15].

The aim of the present study was to examine the changes in the level of fatty acids in different extraction conditions with solvents (hexane, petroleum ether and diethyl ether) Also, by changing the percentage of solvents in binary ratios, the changes in the level of saturated and unsaturated fatty acids were further investigated. Studying nutritional indices can also determine the most suitable solvent in the extraction process.

The results reported here may be considered as a guide in the production of high-quality almond oil and a specific level of fatty acids with high nutritional values. Given the importance of these micronutrients in the diet and the quality of food, our data can provide useful information concerning the nutritional value and health of the selected oils and their optimal production conditions.

1. Material and Methods

1.1. Chemicals and reagents

Two types of almonds related to Kohgiluyeh Province, Iran, were collected. These two species grow naturally in these regions and cover a significant part of the natural areas. The level of humidity in the samples (almond kernels) collected for testing was 4-5% and the percentage of extracted fat was 35-37%. Hexane (HE), petroleum ether (PE), and diethyl ether (DEE) were prepared from chromatographic grade with a mass fraction of higher than 0.995. All chemicals were prepared by the German Company of Merck. The purity of chemicals was controlled by gas chromatography (GC), and the results confirmed that the mass fraction for all materials was greater than 0.99 [16]. The standard mixture of methyl ester fatty acids was prepared by Supelco (FAME Mix).

1.2. Extraction procedures

To extract oil from almond kernels, we used Soxhlet extractor apparatus (Buchi, B-811, extraction System) and solvents of hexane, petroleum ether, and diethyl ether and binary combinations of these solvents with different percentages. The solvent used in Soxhlet apparatus for the extraction process was specified by weighing the solvents in certain proportions using the scale (Sartorius-BL-210S) and mixed well to obtain a uniform compound [17].

About 10 g of the crushed kernels separated by a sieve (size of 850 μm) was weighed and the oil was extracted with 140 ml of solvent for six hours in a Soxhlet apparatus. To measure the humidity content, a psychrometer (Sartorius - MA 100) was used. The extraction process of primary oil from almond species was performed in three replications.

3.2. The analytical procedure for the fatty acid profile of almond kernel oils

The extracted fatty acids were determined by the method described by ZHOU QI in 2019. The analysis process was done by GC-FID using 7890A system (Agilent Technologies) with CP-Ci188 column (length=100m, 0.23mm inner diameter and Film thickness=0.2 μm). The standard reference fatty acid (supelco 37 component FAME Mix) was analyzed simultaneously by the GC apparatus for qualitative comparison. Through comparing the retention time (RT) of the sample and the reference standard, the final results were reported as the final percentage of the level below the peak [12].

4.2. Characterization of fatty acids

Based on the following formulas, the ratio of oleic acid to linoleic acid (O / L) and iodine value (IV) was calculated and reported in the relevant tables [18].

$O/L = \% \text{ Oleic Acid} / \text{Linoleic Acid}$

$IV = (C16:1 \times 0.95) + (C18:1 \times 0.86) + (C18:2 \times 1.732) + (C20:1 \times 0.785)$

5.2. Nutritional quality indexes

Using data on the composition of fatty acids, three important indices were calculated as follows:

1- Atherogenic index (AI) indicates the relationship between saturated fatty acids and unsaturated fatty acids [19].

$AI = [C12:0 + (C14:0 \times 4) + C16:0] / (\sum MUFA + \sum W - 6PUFA + \sum W - 3PUFA)$

In this ratio, with the increase in the level of unsaturated fatty acids, the anti-atherogenic properties increase, thereby preventing the accumulation of plaque and reducing the level of esterified fatty acids, cholesterol, and phospholipids [20].

2-Thrombogenic index (TI) indicates the tendency to create clot in blood vessels and also represents a relationship between prothrombogenic fatty acids (saturated) and anti-thrombogenic fatty acids (unsaturated) [19, 20].

$TI = [C14:0 + C16:0 + C18:0] / [(0.5 \times \sum MUFA) + (0.5 \times \sum W - 6PUFA) + (3 \times \sum W - 3PUFA) + (W - 3PUFA / W - 6PUFA)]$

3-The ratio of hypocholesterolemic to hypercholesterolemic fatty acids was calculated by the method presented by Santos and Bessa [21].

$HH = [C18:1 + C18:2 + C18:3 + C18:4 + C20:4] / [C14:0 + C16:0]$

6.2. Statistical analysis

Data analysis was performed using SPSS 23 software. To analyze the data, their normality was first examined by Kolmogorov-Smirnov test. One-way analysis of ANOVA and Kruskal-Wallis test were used to compare the means of the data. Tukey test was used in cases of significant differences ($P < 0.05$) between the data.

2. Results

2.1. Fatty acid composition and lipid content of almond oils

Humidity content were 4.39% and 4.78% for A.SC and A.LY, respectively. The impact of various solvents and their different compositions and the fatty acid profile of the two types of almond oil were studied and reported. Tables 1 and 2 show the fatty acid contents of the two species of almonds, namely A.Sc and A.Ly. As shown, the solvents were used in pure (single) form and in combination with solvents at different percentages. The simultaneous comparison of SFA, MUFA, PUFA, and TUFA in A.Sc and A.Ly almond oil is reported in Figures 1 and 2 as bar graphs. In Figure 3, the iodine value is reported simultaneously for two almond types A.LY and A.SC, and the effect of extraction with different solvents in this form is comparable. When the solvents were used in pure form, the structure of the fatty acids was as follows:

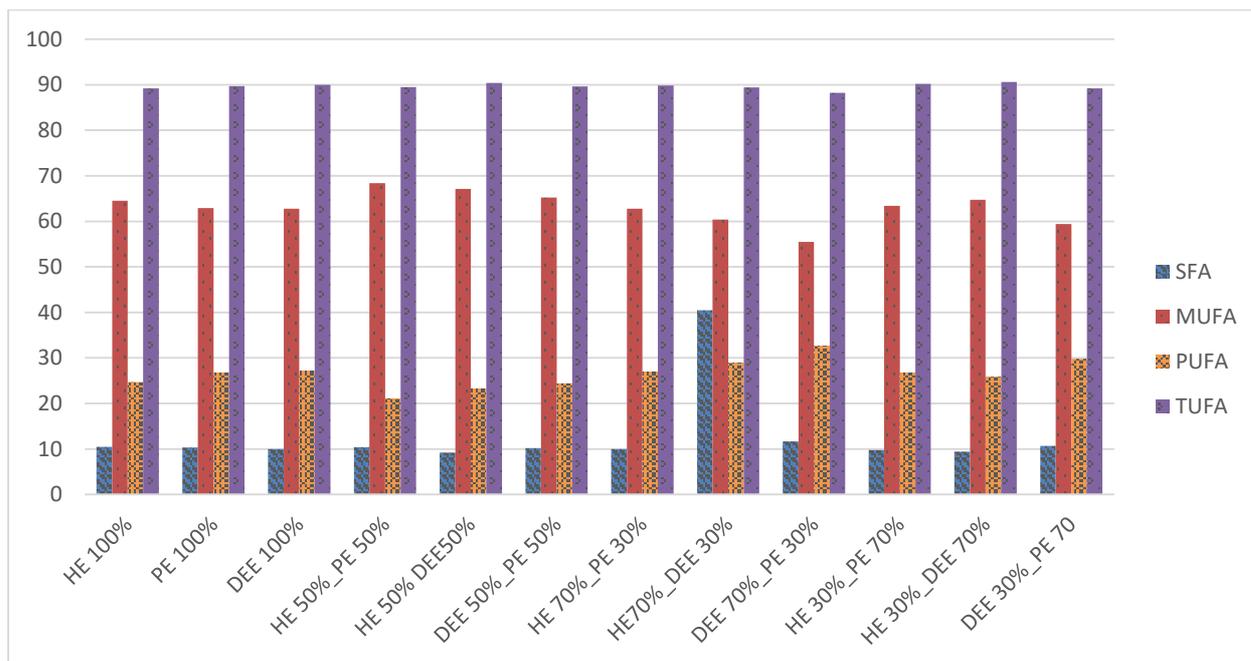


Figure 1. Simultaneous comparison of SFA, MUFA, PUFA, and TUFA for *Amygdalus Scoparia*

The highest level of saturated fatty acid (SFA) belonged to A.Ly in the extraction with hexane solvent (10.7). The lowest level of SFA belonged to A.Ly in extraction using petroleum ether solvent (7.5). Since unsaturated fatty acids (TUFA) are very important in food industry, the highest level of unsaturated fatty acids belonged to A.Ly in the extraction with petroleum ether solvent (92.3). The lowest level of TUFA belonged to the use of hexane solvent in both species (89.2). Moreover, in calculating the iodine value for oil extraction with the help of single solvents, it was found that the highest calculated iodine value belonged to A.Sc almond oil extraction with petroleum ether (100.4), and the lowest value belonged to A.Ly almond oil extraction where petroleum ether solvent was used (96.3).

As shown in tables 1 and 2, our results indicate extraction with the use of binary solvents (in equal ratios), the highest level of saturated fatty acids belonged to A.Ly almond oil extraction through the use of binary solvent (hexane and petroleum ether) (10.6) whereas the lowest level belonged to the extraction of A.Ly oil using binary solvent (hexane-diethyl ether) at the same ratio (8.9). Concerning unsaturated fatty acids (TUFA), the highest level was related to the extraction of A.Ly oil by use of binary solvent (hexane-diethyl ether) (90.8). The lowest level of TUFA observed in A.Ly oil extraction using binary solvent with the same ratios of hexane and petroleum ether (89).

Table 1. Fatty acids composition of plant oils (*A.Scoparia*). Lipids were extracted using the different solvent extraction methods. Results are mean \pm SD (n=3)

| Parameter | Area % <i>Amygdalus scoparia</i> | | | | | | | | | | | | P-Value |
|-----------------|----------------------------------|-------------------------------|-------------------------------|-----------------------------|-----------------------------|-----------------------------|-----------------------------|------------------------------|-----------------------------|--------------------------------|-------------------------------|-----------------------------|--------------|
| Fatty acids (%) | HE 100 | PE 100 | DEE 100 | HE 50 -PE 50 | HE50-DEE 50 | DEE 50- PE 50 | HE 70- PE 30 | HE70-DEE 30 | DEE70-PE 30 | HE 30 -PE 70 | HE30-DEE70 | DEE30- PE 70 | |
| C 6:0 | 1.3 \pm 0.1 ^a | 0.0 \pm 0 ^b | 0.0 \pm 0 ^b | 2.0 \pm 0.2 ^d | 0.4 \pm 0.2 ^c | 0.6 \pm 0.05 ^c | 0.0 \pm 0 ^b | 0.0 \pm 0 ^b | 0.0 \pm 0 ^b | 0.0 \pm 0 ^b | 0.0 \pm 0 ^b | 0.0 \pm 0 ^b | 0.000 |
| C 16:0 | 6.7 \pm 0.05 ^{def} | 7.6 \pm 0.706 ^{ij} | 7.4 \pm 0.1 ^j | 6.1 \pm 0.25 ^c | 6.3 \pm 0.8 ^{ce} | 7.0 \pm 0.2 ^{gh} | 7.4 \pm 0.4 ^{ij} | 7.8 \pm 0.2 ^{hik} | 8.7 \pm 0.1 ^c | 7.3 \pm 0.3 ^{ij} | 7.0 \pm 0.3 ^{ij} | 8.0 \pm 0.3 ^{ik} | 0.000 |
| C 16:1 | 0.4 \pm 0.05 ^a | 0.3 \pm 0.07 ^a | 0.3 \pm 0.09 ^a | 0.2 \pm 0.06 ^a | 0.3 \pm 0.09 ^a | 0.5 \pm 0.04 ^a | 0.3 \pm 0.09 ^a | 0.3 \pm 0.09 ^a | 0.4 \pm 0.1 ^a | 0.3 \pm 0.02 ^a | 0.4 \pm 0.05 ^a | 0.3 \pm 0.12 ^a | 0.011 |
| C 17:1 | 0.2 \pm 0.03 ^a | 0.1 \pm 0.01 ^b | 0.1 \pm 0.01 ^b | 0.0 \pm 0 ^c | 0.0 \pm 0 ^c | 0.0 \pm 0 ^c | 0.2 \pm 0.05 ^a | 0.0 \pm 0 ^c | 0.1 \pm 0.02 ^b | 0.1 \pm 0.02 ^b | 0.2 \pm 0.05 ^a | 0.2 \pm 0.05 ^a | 0.001 |
| C 18:0 | 2.3 \pm 0.2 | 2.5 \pm 0.07 | 2.4 \pm 0.09 | 2.1 \pm 0.09 | 2.2 \pm 0.4 | 2.4 \pm 0.2 | 2.4 \pm 0.4 | 2.5 \pm 0.4 | 2.8 \pm 0.2 | 2.3 \pm 0.2 | 2.2 \pm 0.3 | 2.5 \pm 0.15 | 0.167 |
| C 18:1 n9c | 63.5 \pm 1.04 ^{adth} | 62.4 \pm 0.6 ^{adg} | 62.3 \pm 0.3 ^{adg} | 68.1 \pm 0.6 ^c | 66.6 \pm 0.4 ^c | 64.6 \pm 0.6 ^f | 62.2 \pm 0.2 ^e | 60.0 \pm 0.5 ^b | 54.9 \pm 0.1 ⁱ | 62.9 \pm 0.35 ^{agj} | 64.0 \pm 0.29 ^{hf} | 58.7 \pm 0.3 ^b | 0.000 |

| | | | | | | | | | | | | | |
|------------|-------------------------|-------------------------|--------------------------|------------------------|------------------------|--------------------------|--------------------------|-------------------------|------------------------|-------------------------|------------------------|-------------------------|--------------|
| C 18:2 n6c | 24.7±0.15 ^a | 26.8±0.07 ^b | 27.2±0.2 ^b | 21.1±0.1 ^c | 23.3±0.3 ^d | 24.4±0.1 ^a | 27.0±0.3 ^b | 29.0±0.6 ^e | 32.7±0.4 ^f | 26.8±0.2 ^b | 25.9±0.1 ^s | 29.7±0.2 ^c | 0.000 |
| C 20:0 | 0.2±0.03 | 0.2±0.03 | 0.2±0.03 | 0.2±0.02 | 0.3±0.08 | 0.2±0.03 | 0.2±0.06 | 0.2±0.06 | 0.2±0.05 | 0.2±0 | 0.2±0.06 | 0.2±0.4 | 0.929 |
| C 20:1 | 0.4±0.1 | 0.1±0.2 | 0.1±0.2 | 0.1±0.0 | 0.2±0.09 | 0.1±0.01 | 0.1±0 | 0.1±0.02 | 0.1±0.01 | 0.1±0.02 | 0.1±0.01 | 0.2±0.06 | 0.080 |
| C 20:2 | 0.0 ^a | 0.0 ^a | 0.0 ^a | 0.0 ^a | 0.0 ^a | 0.0 ^a | 0.0 ^a | 0.0 ^a | 0.0 ^a | 0.0 ^a | 0.0 ^a | 0.1 ^b | 0.000 |
| SFA | 10.5±0.02 ^{af} | 10.3±0.15 ^{af} | 10.0±0.15 ^{abf} | 10.4±0.4 ^{af} | 9.2±0.4 ^{bc} | 10.2±0.20 ^{adf} | 10.0±0.15 ^{abf} | 10.5±0.5 ^{af} | 11.7±0.3 ^c | 9.8±0.2 ^{bc} | 9.4±0.2 ^{db} | 10.7±0.3 ^f | 0.000 |
| MUF A | 64.5±0.7 ^{afg} | 62.9±0.55 ^{ag} | 62.8±0.4 ^a | 68.4±0.6 ^{bc} | 67.1±0.27 ^c | 65.2±0.4 ^f | 62.8±0.2 ^{af} | 60.4±0.4 ^{bc} | 55.5±0.3 ^d | 63.4±0.4 ^{af} | 64.7±0.3 ^{af} | 59.4±0.6 ^c | 0.001 |
| PUF A | 24.7±0.2 ^a | 26.8±0.2 ^{bb} | 27.2±0.3 ^b | 21.1±0.25 ^c | 23.3±0.3 ^d | 24.4±0.15 ^a | 27.0±0.5 ^b | 29.0±0.4 ^{es} | 32.7±0.3 ^f | 26.8±0.2 ^{bb} | 25.9±0.1 ^h | 29.8±0.2 ^s | 0.000 |
| TUF A | 89.2±0.2 ^a | 89.7±0.3 ^{ab} | 90.0±0.7 ^{ab} | 89.5±0.5 ^{ab} | 90.4±0.6 ^b | 89.6±0.4 ^{ab} | 89.8±0.4 ^{ab} | 89.4±0.4 ^{ab} | 88.2±0.4 ^a | 90.2±0.5 ^{ab} | 90.6±0.4 ^{ab} | 89.2±0.46 ^{ab} | 0.008 |
| TUF A/SF A | 8.4±0.1 ^{ac} | 8.7±0.35 ^{ac} | 9.0±0.3 ^{abde} | 8.6±0.4 ^{ac} | 9.8±0.2 ^b | 8.8±0.2 ^{ac} | 9.0±0.5 ^{abe} | 8.5±0.3 ^{ac} | 7.5±0.3 ^{cc} | 9.2±0.2 ^{ab} | 9.6±0.25 ^{db} | 8.3±0.3 ^{cc} | 0.000 |
| O/L | 2.5±0.1 ^{abcd} | 2.3±0.3 ^{ad} | 2.3±0.3 ^{ad} | 3.2±1.5 ^{bcd} | 2.8±0.1 ^c | 2.6±0.05 ^{dc} | 2.3±0.3 ^{ac} | 2.0±0.1 ^{ad} | 1.7±0.4 ^a | 2.3±0.3 ^{ac} | 2.4±0.1 ^{ac} | 2.0±0.6 ^{ad} | 0.000 |
| IV | 98.0±0.26 ^a | 100.4±0.4 ^b | 101.0±0.6 ^b | 95.3±0.3 ^d | 98.0±0.6 ^a | 98.3±0.7 ^a | 100.6±0.6 ^{cb} | 102.2±0.4 ^{cd} | 104.3±0.3 ^h | 100.9±0.4 ^{bk} | 100.3±0.7 ^b | 102.3±0.8 ^{bc} | 0.000 |

Table 2. Fatty acids composition of plant oils(*A.Lycioides*).Lipids were extracted using the different solvent

| Parameter | Area % <i>Amygdalus lycioides</i> | | | | | | | | | | | | P-Value |
|------------|--------------------------------------|------------------------|------------------------|------------------------|------------------------|-------------------------|------------------------|------------------------|------------------------|------------------------|------------------------|------------------------|--------------|
| | HE 100 | PE 100 | DEE 100 | HE 50 -PE 50 | HE50- DEE 50 | DEE 50- PE 50 | HE 70- PE 30 | HE70- DEE 30 | DEE70- PE 30 | HE 30 -PE 70 | HE30- DEE70 | DEE30- PE 70 | |
| C 6:0 | 1.8±0.1 ^a | 0.0±0 ^b | 0.0±0 ^b | 1.7±0.2 ^a | 0.3±0.1 ^c | 1.3±0.2 ^d | 0.0±0 ^b | 0.000 |
| C 16:0 | 6.4±0.15 ^{ac} | 5.3±0.1 ^b | 6.1±0.2 ^a | 6.4±0.1 ^{ac} | 6.2±0.2 ^{ac} | 6.6±0.2 ^{cd} | 7.6±0.3 ^d | 9.3±0.2 ^e | 9.0±0.1 ^e | 8.4±0.1 ^f | 8.2±0.2 ^f | 8.3±0.2 ^f | 0.000 |
| C 16:1 | 0.3±0.1 | 0.0±0.05 | 0.2±0.1 | 0.3±0.2 | 0.4±0.2 | 0.4±0.1 | 0.2±0.1 | 0.4±0.1 | 0.4±0.2 | 0.4±0.1 | 0.3±0 | 0.3±0.1 | 0.109 |
| C 17:1 | 0.0±0 ^a | 0.4±0.1 ^b | 0.2±0 ^c | 0.0±0 ^a | 0.0±0 ^a | 0.1±0.05 ^c | 0.1±0 ^c | 0.1±0 ^c | 0.1±0 | 0.0±0 ^a | 0.1±0 ^c | 0.1±0 ^c | 0.000 |
| C 18:0 | 2.3±0.2 ^{ab} | 2.0±0.3 ^a | 2.3±0.1 ^{ab} | 2.3±0.05 ^{ab} | 2.2±0.2 ^a | 2.3±0.1 ^{ab} | 2.8±0.2 ^b | 3.2±0.3 ^d | 3.4±0.1 ^c | 2.9±0.1 ^{dc} | 3.0±0.3 ^{dc} | 2.9±0.1 ^{dc} | 0.002 |
| C 18:1 n9c | 65.5±0.4 ^a | 71.9±0.6 ^b | 68.4±0.4 ^c | 65.2±0.2 ^a | 67.8±0.5 ^c | 65.0±0.3 ^a | 61.5±0.1 ^d | 57.5±0.1 ^e | 52.4±0.1 ^f | 62.6±0.3 ^s | 59.8±0.1 ^h | 58.4±0.3 ^c | 0.000 |
| C 18:2 n6c | 23.2±0.2 ^a | 19.9±0.3 ^b | 22.4±0.2 ^c | 23.1±0.1 ^a | 22.5±0.15 ^c | 23.7±0.3 ^a | 27.3±0.1 ^d | 28.8±0.2 ^e | 34.2±0.1 ^f | 25.2±0.2 ^s | 28.2±0.2 ^b | 29.4±0.1 ⁱ | 0.000 |
| C 20:0 | 0.2±0.1 | 0.2±0.1 | 0.2±0.1 | 0.2±0.05 | 0.2±0.1 | 0.2±0.1 | 0.2±0.1 | 0.3±0 | 0.3±0.1 | 0.3±0.1 | 0.3±0.1 | 0.3±0 | 0.308 |
| C 20:1 | 0.2±0 ^{ab} | 0.1±0 ^a | 0.1±0 ^a | 0.4±0.1 ^b | 0.1±0 ^a | 0.0±0 ^a | 0.2±0.2 ^{ab} | 0.2±0.1 ^{ab} | 0.2±0.1 ^{ab} | 0.1±0 ^a | 0.1±0 ^a | 0.2±0.1 ^{ab} | 0.013 |
| C 20:2 | 0.0±0 ^a | 0.0±0 ^a | 0.0±0 ^a | 0.0±0 ^a | 0.0±0 ^a | 0.0±0 ^a | 0.0±0 ^a | 0.2±0.1 ^b | 0.0±0 ^a | 0.1±0 ^c | 0.0±0 ^a | 0.1±0 ^{cc} | 0.000 |
| SFA | 10.7±0.2 ^a | 7.5±0.1 ^b | 8.6±0.1 ^c | 10.6±0.4 ^a | 8.9±0.1 ^c | 10.4±0.3 ^a | 10.6±0.2 ^a | 12.8±0.2 ^d | 12.7±0.2 ^d | 11.6±0.2 ^c | 11.5±0.1 ^c | 11.5±0.3 ^c | 0.000 |
| MUFA | 66.0±0.1 ^a | 72.4±0.5 ^b | 68.9±0.1 ^c | 65.9±0.1 ^a | 68.3±0.5 ^c | 65.5±0.5 ^a | 62.0±0.6 ^d | 58.2±0.3 ^e | 53.1±0.1 ^f | 63.1±0.3 ^s | 60.3±0.3 ^b | 59.0±0.2 ^f | 0.000 |
| PUFA | 23.2±0.2 ^{ad} | 19.9±0.2 ^b | 22.4±0.2 ^c | 23.1±0.2 ^a | 22.5±0.1 ^c | 23.7±0.2 ^d | 27.3±0.1 ^e | 29±0.3 ^f | 34.2±0.2 ^s | 25.3±0.1 ^h | 28.2±0.1 ⁱ | 29.5±0.1 ^f | 0.000 |
| TUFA | 89.2±0.1 ^a | 92.3±0.3 ^b | 91.3±0.2 ^c | 89.0±0.2 ^{bc} | 90.8±0.2 ^c | 89.2±0.3 ^{ai} | 89.3±0.1 ^{ac} | 87.2±0.2 ^f | 87.3±0.2 ^f | 88.4±0.4 ^d | 88.5±0.4 ^{di} | 88.5±0.2 ^{di} | 0.000 |
| TUFA/SFA | 8.3±0.1 ^a | 12.3±0.15 ^b | 10.6±0.1 ^c | 9.4±0.2 ^d | 10.2±0.2 ^c | 8.6±0.3 ^a | 8.4±0.2 ^a | 6.8±0.1 ^c | 6.8±0.1 ^c | 7.6±0.3 ^f | 7.7±0.1 ^f | 7.7±0.1 ^f | 0.000 |
| O/L | 2.8±0.3 ^{ac} | 3.6±0.1 ^b | 3.0±0.15 ^{ab} | 2.8±0.1 ^{ac} | 3.0±0.3 ^{ab} | 2.7±0.1 ^{acd} | 2.2±0.2 ^{cc} | 2.0±0.1 ^{cdg} | 1.5±0.3 ^s | 2.5±0.25 ^{ac} | 2.1±0.2 ^{cdg} | 2.0±0.3 ^{cd} | 0.000 |
| IV | 96.9±0.5 ^{ab} | 96.3±0.2 ^b | 97.8±0.2 ^c | 96.6±0.1 ^{ab} | 97.7±0.2 ^{ac} | 97.3±0.4 ^{acd} | 100.5±0.5 ^c | 99.9±0.1 ^c | 105.0±0.2 ^f | 97.9±0.1 ^c | 100.6±0.2 ^c | 101.6±0.3 ^s | 0.000 |

The highest iodine value calculated in this type of extraction using binary solvents with the same ratios belonged to the extraction of A.Sc oil through the use of diethyl ether-petroleum ether solvents (98.3), and the lowest level was related to A.Sc oil extraction with the use of binary solvent of hexane-petroleum ether (95.3). The data obtained from Tables 1 and 2 for different solvents with different percentage combinations (ratio of 70 to 30) also showed that the highest level of SFA was related to extraction with the use of binary solvents of 70% hexane and 30% diethyl ether in A.Ly species (12.8) while the lowest SFA belonged to the use of binary solvents (70% hexane - 30% petroleum ether) in A.Sc species (10.0). The highest level of unsaturated fatty acids (TUFA) was observed in the extraction using binary solvent (70% hexane-30% petroleum ether) in A.Sc species (89.8), and the lowest was detected in the extraction where the solvent of 70% hexane - 30% diethyl ether was used in A.Ly species (87.2%). Furthermore, the most iodine value (IV) was observed in A.Ly species in which oil was extracted by use of binary solvent (70% diethyl ether - 30% petroleum ether) (10.5%). The least iodine value was found in A.Ly species where oil was extracted with the use of binary solvent (hexane 70% - 30% diethyl ether) (9.9%).

The results of Tables 1 and 2 (ratio of 30 to 70) show that the highest level of SFA was in the oil extracted from A.Ly species with the use of binary solvent (30% hexane and 70% petroleum ether) (11.6). The lowest level of SFA belonged to the oil extracted from A.Sc species with the use of binary solvent (30% hexane and 70% diethyl ether) (9.4). In determining the level of TUFA, it was observed that the oil extracted from A.Sc with the use of binary solvent (30% hexane and 70% diethyl ether) had the highest level (90.6%). The oil extracted from A.Ly species using binary solvent (30% hexane and 70% petroleum ether) showed the lowest level of TUFA (88.4%). Based on the calculations of iodine value for binary solvents at the ratio of 30 to 70, the oil extracted from A.Sc species using binary solvent (30% diethyl ether - 70% petroleum ether) had the highest Iodine value (102.3%), and the oil extracted from A.Ly species by use of binary solvent (30% hexane and 70% petroleum ether) had the least iodine value (97%).

2.2. Nutritional impact of lipid fraction using lipid quality indices

Due to the interest in the content of healthy fats, many studies have been conducted on different species of almonds. Since these fatty acids are known as important sources of FAs with therapeutic effects, there has been an increase in the research in this area. Despite being a source of healthy lipids, almond species have SFA levels (palmitic acid) that are unfavorable for human consumption. Therefore, it is important to evaluate the nutritional impact of these fats using fat quality indices. Nutritional indices that are mostly related to cardiovascular diseases such as AI (Atherogenicity index), TI (Thrombogenicity index), and HH (Hypocholesterolemic: Hypercholesterolemic) have a great importance in human health.

Table 3 shows the quality indices of the oil extracted by use of various solvents. As observed, the lowest index belonged to *Amygdalus Lycioides* and the use of petroleum ether solvent (100%) (TI=0.15 and AI=0.05). The highest level of HH was also obtained at this extraction (17.32). The results obtained for *Amygdalus Scoparia* showed that the best (lowest) index was related to the use of hexane and petroleum ether combined solvent at the same ratio of 0.06 (AI) and 0.18 (TI). In this extraction, the calculations showed the highest level of HH (14.62). In general, the rate of AI and TI changes in the two studied almond species was: A.Ly [AI (0.05 - 0.1), TI (0.15 - 0.28)] and A.Sc [AI 0.06 - 0.09, TI (0.18 - 0.26)]. Comparison of quality indices of oil extracted from two types of almonds A.Ly and A.Sc with quality indices of oil extracted from algae *Laminaria Ochroleuca* [AI (0.7 - 1.2), TI (1.1 - 1.9)] shows that the oils of these two types of almonds are much more useful in terms of quality and nutritional value[15]. Also, the comparison of these indices with the results of Fish Indices [AI (0.33 - 1.47), TI (0.16 - 1.15)] indicate the importance and very high quality of these two species of almond oil [22].

Table 3. Nutritional quality indices of the oil extracted by use of various solvents

| | | HE 100 | PE 100 | DEE 100 | HE 50 -PE 50 | HE50- DEE 50 | DEE 50- PE 50 | HE 70- PE 30 | HE70- DEE 30 | DEE70- PE 30 | HE 30 - PE 70 | HE30- DEE70 | DEE30- PE 70 |
|------|----|--------|--------|---------|-----------------|-----------------|------------------|-----------------|-----------------|-----------------|------------------|----------------|-----------------|
| | AI | 0.07 | 0.08 | 0.08 | 0.06 | 0.06 | 0.07 | 0.08 | 0.08 | 0.09 | 0.08 | 0.07 | 0.08 |
| A.SC | TI | 0.20 | 0.22 | 0.21 | 0.18 | 0.18 | 0.20 | 0.21 | 0.23 | 0.26 | 0.21 | 0.20 | 0.23 |
| | HH | 13.16 | 11.37 | 12.09 | 14.62 | 14.26 | 12.71 | 12.05 | 11.41 | 10.06 | 12.28 | 12.84 | 11.05 |

| | | | | | | | | | | | | | |
|------|----|------|-------|-------|-------|-------|-------|-------|------|------|-------|-------|-------|
| | | | | | | | | | | | | | |
| | AI | 0.07 | 0.05 | 0.06 | 0.07 | 0.06 | 0.07 | 0.08 | 0.10 | 0.10 | 0.09 | 0.09 | 0.09 |
| A.LY | TI | 0.19 | 0.15 | 0.18 | 0.19 | 0.18 | 0.19 | 0.23 | 0.28 | 0.28 | 0.25 | 0.25 | 0.25 |
| | HH | 13.8 | 17.32 | 14.88 | 13.79 | 14.56 | 13.43 | 11.68 | 9.27 | 9.62 | 10.45 | 10.73 | 10.57 |

3. Conclusion

The present study investigated the oils obtained from extraction with different solvent and binary combination solvents. The effects of solvents on the level of saturated, unsaturated fatty acids, nutritional properties, and iodine value of these oils were further investigated. Based on the results, the highest level of saturated fatty acids belonged to the extraction of A.Ly species using hexane 70% and diethyl ether 30% solvents (12.8%) while the lowest level of SFA was observed in the extraction of A. Ly species with the use of the pure solvent of petroleum ether (7.5%). Results also showed that the highest TUFA was related to the extraction of A.LY species using the pure solvent of petroleum ether. The lowest TUFA belonged to the extraction of A.LY species (92.3%) using the binary solvent of hexane 70% and 30% diethyl ether (87.2%).

The general results observed for iodine value showed that the highest iodine value was related to the extraction of A.Ly species with the use of the binary solvent of diethyl ether 70% and petroleum ether 30% (105%) whereas the least iodine value belonged to the extraction of A.Sc species through the use of the binary solvent of hexane and petroleum ether with equal ratios (50-50) (95.3%). In this study, the best method for the optimal extraction of unsaturated fatty acids was the use of the pure solvent of petroleum ether in A.Ly species (92.3%). Also, the most optimal TUFA / SFA value was related to the use of petroleum ether

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