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ORIGINAL PAPER

Evaluation of seed quality and oil parameters in native Iranian almond (*Prunus* L. spp.) species

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Abstract: We assessed chemical composition and variation in oil content and seed weight of 40 wild-growing almonds (*Prunus* L. spp.) accessions collected from different parts of Iran. There were significant differences in kernel weight and oil parameters. Accessions ranged from 0.20 to 1.5 g in kernel weight, 0.2–3.0 mm in shell thickness, and 16% to 55% in oil content. The predominant vegetable oil components of kernels were 4.6–9.5% palmitic acid, 0.4–0.8% palmitoleic acid, 1.0–3.4% stearic acid, 48.8–88.4% oleic acid and 11.3–33.2% linoleic acid. Linolenic acid was detected in 15 accessions. High heritability was recorded for all studied traits and was maximum for shell thickness (98.5%) and minimum for oil content (97.1%). Maximum and minimum 'Euclidean' pair wise dissimilarities were 17.9 and 0.5, respectively. All 40 accessions were grouped into two major clusters.

Keywords: genetic diversity, kernel quality, oil parameters, wild almond (*Prunus* L. spp.)

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Introduction

Almond (*Prunus dulcis* Miller (D. A Webb) syn. *P. amygdalus* Batsch) is one of the most important nut crops and today represents the largest production of any commercial tree nut product (Woodroof 1978; Kester et al. 1991). Almond species grow in subtropical Mediterranean climate, with mild, wet winters and warm, dry summers (Kester and Gradziel 1996). They originated from Central Asia and represent divergent evolution under xerophytic environments (Sorkheh et al. 2009; Szikriszt et al. 2011). Species of *Prunus* that are related to those used commercially grow wild from eastern China to mountainous areas and deserts of western China, Kurdistan, Turkistan, Afghanistan and Iran (Kester and Gradziel 1996). Iran, with a total land area of 1,648,195 square kilometers, lies between 25º and 39º N and 44º and 63º E and is primarily subtropical in the southern half of the country, temperate in the northern half, and mostly desert in the middle. The resultant variability in environment and climate has made possible extensive diversity in plant germplasm (Sorkheh et al. 2009).

The limited gene pool in cultivated almonds limits its cultivation to specific areas with a Mediterranean climate. The related species grow in more diverse climates therefore could have traits that enable them to survive in these climates and germplasm with these traits could be a valuable resource for breeding (Gradziel et al. 2001; Sorkheh et al. 2009). The size and chemical composition of the almonds is also very important. The quality of seeds is defined in particular by moisture content, lipid content, oil composition and oil ultraviolet absorption coefficients (Kumar et al. 1994; Martin-Carralata et al. 1998; Conelle et al. 2000). These characters are influenced by ecological conditions, location and technical and cultural practices used to extract the oil (Askin et al. 2007). Among agronomical operations, irrigation may be the most important factor affecting almond kernel weight, yield and quality, and sugar composition. Irrigation has no remarkable influence on lipid content or fatty acid composition (Soler et al. 1988; Schirra and Agabbio 1989; Farhoosh and Moosavi 2006;

Piscopo et al. 2010; Szikriszt et al. 2011).

Wild relatives of almond have not been subjected to any genetic improvement through domestication and are considered to possess greater natural variability compared to crop plants (Quebedeaux et al. 1990; Kaushik et al. 2007; Szikriszt et al. 2011). For rational tree-breeding programmes, knowledge of genetic variability of species is essential. This study was undertaken to assess wild almond germplasm collections based on multivariate analysis of seed quality traits and to quantify oil content and describe fatty acid composition of some native Iranian wild almonds and to evaluate relationships among fatty acid contents, kernel weight and shell thickness. In the search for new oilseed plants for nutritional, pharmaceutical, industrial and other uses, potential yield of oil or fat is of paramount importance (Lawson et al. 1995). The effectiveness of a tree improvement programme depends upon the nature and magnitude of existing genetic variability and also on the heritability of the economically important traits being pursued (Zobel e al. 1984; Yadav et al. 2011).

This study was undertaken to assess the genetic diversity among germplasm collections of wild almond based on multivariate analysis of seed quality traits. The objectives of this work were to identify oil content and fatty acid composition of some native Iranian wild almonds and to evaluate relationships among fatty acid content, kernel weight and shell thickness.

Material and methods

Plant materials

The wild almond species used in the study belong to the genus *Prunus*, subgenus *Amygdalus*, and include the following: *P. communis* (L.) Archang, *P. eleagnifolia* (Spach) Fritsch, *P. orientalis* Mill. (syn. *P. argentea* Lam.) in section Euamygdalus Spach; *P. lycioides* Spach, *P. reuteri* Boiss. et Bushe in section Lycioides Spach; and *P. arabica* (Olivier) Neikle, *P. glauca* (Browicz) A.E. Murray, *P. scoparia* Spach in section Spartioides Spach. The number of accessions sampled per site ranged from one to five, depending on habitat diversity and availability at collection time. The accession numbers for all samples and geographical locations of wild almond species populations collected in Iran was reported by Sorkheh et al. (2009). Wild almond trees representing in this study were marked in the native population in 2008. Field expeditions were carried out in 2009 and 2010 on the basis of recent literature (Sorkheh et al. 2009), indigenous information, or conspicuous presence. Collections came from both wild and cultivated habitats concentrated in two regions in Iran. The first region (Azerbaijan and Kurdistan, 36°00′ to 38°28′ N, 44°51′ to 45°46′ E, mean elevation of 1473 m a.s.l.) is characterized by relatively lush environment, mean annual rainfall of 507 mm, high biological diversity, and relatively low agricultural development. The second region (Shahrekord and Shiraz, 32° 17′ to 50°51′ N, 28°58′ to 53°41′ E, mean elevation of 2030 m a.s.l.), with mean annual rainfall of 436 mm, is in a more xerophytic area with widespread agriculture.

Fruit traits and evaluation of seed quality

The fruit samples of *Prunus* L. spp. were collected from two regions in Iran, and were stored at -20ºC prior to analysis. Almond fruits from each species, five per species, were harvested on August 10, and nut samples were collected for two years (2009–2010). Forty nuts were randomly chosen for fruit analyses and biochemical traits. In addition, kernel weight (g) and shell thickness (mm) were recorded for each accession of wild almond species. Three replications were used for analyses of nut quality traits according to Askin et al. (2007).

We analyzed total kernel oil content and fatty acid composition. The seeds were ground and extracted with hexane by agitation in a dark place at ambient temperature for 48 h following Farhoosh and Tavakoli (2008). The fatty acid profile of the oils was determined by gas-liquid chromatography and reported in relative area percentages. Fatty acids were transesterified into their corresponding fatty acid methyl esters (FAMEs) by vigorous shaking of a solution of oil in hexane (0.3 g in 7 mL) with 2 mL of 7 M methanolic potassium hydroxide at 50ºC for 10 min. The FAMEs were analyzed using a gas liquid chromatograph (GLC, Model-hp6890) equipped with a capillary column (30 mm × 0.25 mm, 0.25 mm film thickness), and a flame ionization detector. Temperature of both the injector and detector were 240ºC. The flow rate was 35 mL/min. The column temperature was 190ºC according to Askin et al. (2007).

Statistical analysis

The experimental design was completely randomized with three replications. The statistical package SAS (SAS Institute 2000) was utilized for analysis of variance (ANOVA). LSD values were computed for multiple comparisons of means. Significant differences were recorded at $p < 0.01$ and $p < 0.05$. Correlation analyses were performed to examine relationships among fatty acid content, kernel weight and shell thickness.

The analysis of variance, both phenotypic and genotypic, was calculated using software PBSTAT 1.2 (http://www.pbstat.com/) following Yadav et al. (2011). Euclidean coefficients of dissimilarity (Euclidean distance) among 40 *Prunus* accessions were calculated in pairwise combinations. The dissimilarity matrix was used to construct a dendrogram by the unweighted pair group method for arithmetic mean (UPGMA) based Sequential Agglomerative Hierarchical and Nested (SAHN) clustering. The receptivity to fit of the dendrogram/cluster analysis to the matrix, on which it was based, was checked by calculating cophenetic correlation using the cophenetic ultrametric values (COPH) program. The significance of cophenetic correlation was tested by computing the standardized Mantel statistics that test whether one set of relationships among a set of objects is independent of another using the MXCOMP procedure. We undertook Principle Coordinate Analysis (PCoA) based on the Euclidean coefficients of dissimilarity. All these calculations were made using NTSYS-pc version 2.11 (USA) (Rohlf et al. 2000).

Results

Estimation of genetic variability

ANOVA reflected significant genetic variation among wild almond accessions at p <0.01 (results not shown). All characters differed significantly by accession indicating abundant variability in the germplasm.

Mean kernel weight and shell thickness in 40 wild almond accessions differed statistically (p <0.01). Kernel weight ranged from 0.2 to 1.5 g. Shell thickness ranged from 0.2–3.0 mm, with genotypes C1 (2.2 mm), C2 (3.0 mm), C3 (2.6 mm), C4 (2.5 mm), C5 (2.8 mm), E1 (2.1 mm), E2 (1.3 mm), E4 (1.5 mm), O5 (1.3 mm), L4 (1.2 mm), R4 (1.2 mm) and S5 (1.2 mm) having the thickest shells (Table 1). Oil content (%), and the percentage fraction of palmitic, palmitoleic, stearic, oleic and linoleic acids in the kernels also differed statistically (p <0.01). Kernels of E5, O5, L3 and L5 contained the highest oleic acid content.

Table 1: Kernel weight, shell thickness, oil content and fatty acid composition of various wild almond species from Iran.

Species	Kernel weight (g)	Shell thickness (mm)	Oil content (%)	Fatty acid					
				Palmitic acid (C16:0)	Palmitoleic acid (C16:1)	Stearic acid (C18:0)	Oleic acid (C18:1)	Linoleic acid (18:2)	Linolenic acid (C18:3)
P. communis									
C1	1.0 ± 0.35	2.2 ± 0.45	42.32 ± 1.25	6.35 ± 0.49	0.44 ± 0.25	1.18 ± 0.22	69.45 ± 2.34	22.05 ± 2.85	-
C2	1.1 ± 0.32	3.0 ± 0.47	33.21 ± 1.38	6.48 ± 1.33	0.48 ± 0.65	1.65 ± 0.48	68.25 ± 2.77	21.35 ± 1.95	-
C3	1.2 ± 0.29	2.6 ± 0.36	22.56 ± 1.02	5.48 ± 0.98	0.64 ± 0.28	1.32 ± 0.33	72.35 ± 2.25	18.45 ± 1.22	-
C4	1.5 ± 0.33	2.5 ± 0.42	26.58 ± 1.58	8.31 ± 0.87	0.70 ± 0.45	1.00 ± 0.12	74.88 ± 3.12	17.65 ± 1.35	-
C5	1.3 ± 0.28	2.8 ± 0.34	20.19 ± 1.01	7.21 ± 0.94	0.74 ± 0.74	1.89 ± 0.19	77.48 ± 3.74	14.12 ± 0.98	-
P. eleagnifolia									
E1	0.4 ± 0.21	2.1 ± 1.24	20.18 ± 1.25	5.61 ± 0.88	0.69 ± 0.25	1.68 ± 0.15	75.49 ±2.15	17.84 ± 0.28	-
E2	0.3 ± 0.18	1.3 ± 0.95	19.84 ± 2.33	6.25 ± 0.95	0.48 ± 0.18	1.34 ± 0.45	75.36 ±1.89	20.19 ± 0.38	-
E3	0.6 ± 0.11	1.2 ± 0.84	22.34 ± 1.48	7.25 ± 1.11	0.83 ± 0.28	1.47 ± 0.18	68.12 ±1.02	19.28 ± 0.46	-
E4	0.5 ± 0.09	1.5 ± 0.65	38.42 ± 1.25	6.35 ± 0.84	0.53 ± 0.38	1.82 ± 0.49	77.15 ±0.98	20.88 ± 0.45	-
E5	0.7 ± 0.05	0.7 ± 0.48	19.46 ± 1.18	5.28 ± 1.02	0.76 ± 0.19	1.39 ± 0.38	78.65 ±2.15	15.36 ± 0.19	-
P. orientalis									
O1	0.5 ± 0.05	0.3 ± 0.15	18.45 ± 1.52	6.04 ± 1.25	0.51± 0.02	1.42 ± 0.25	77.68 ±1.20	15.38 ± 0.87	-
O2	0.3 ± 0.02	0.4 ± 0.14	22.78 ± 2.35	4.58 ± 0.98	0.48 ±0.08	1.18 ± 0.39	74.35± 0.98	14.12 ±0.39	-
O3	0.2 ± 0.03	0.7 ± 0.08	32.77 ± 1.48	6.54 ± 0.74	0.44 ± 0.06	1.58 ±0.68	73.85 ±1.45	13.54 ± 0.31	-
O4	0.6 ± 0.02	0.2 ± 0.03	24.65 ± 2.11	5.48 ± 0.84	0.65 ± 0.45	1.68 ± 0.42	69.70 ±0.98	11.65 ± 0.12	-
O5	0.4 ± 0.01	1.3 ± 1.08	23.11 ±1.78	6.34 ± 1.89	0.74 ± 0.12	1.77 ± 0.34	78.22 ±2.35	12.19 ± 0.46	-
P. lycioides									
L1	0.5 ± 0.03	0.2 ± 0.24	16.45 ± 1.25	7.69 ± 1.52	0.40 ± 0.12	1.37 ± 0.88	68.35 ±1.36	15.25 ± 0.65	-
L2	0.4 ± 0.12	0.3 ± 0.15	16.07 ± 1.44	8.44 ± 1.88	0.36 ± 0.35	1.84 ± 0.45	48.78 ±2.15	11.32 ± 0.37	-
L3	1.1 ± 0.05	0.4 ± 0.36	18.47 ± 1.01	6.25 ± 2.15	0.49 ± 0.18	1.95 ± 0.20	88.35 ±2.42	12.95 ± 1.15	-
L4	1.3 ± 0.87	1.2 ± 0.29	20.11 ± 2.01	7.44 ± 1.42	0.58 ± 0.08	1.66 ± 1.25	74.15 ±1.26	12.67 ± 0.87	-
L5	0.8 ± 0.44	0.6 ± 0.15	19.88 ± 2.48	5.15 ± 0.98	0.51 ± 0.04	1.44 ± 0.48	79.65 ±1.47	14.75 ± 1.12	-
P. reuteri									
R1	0.3 ±0.04	0.2 ± 0.62	20.14 ± 1.22	7.39 ± 0.68	0.42 ± 0.02	1.18 ± 0.12	54.87 ±2.35	15.00 ±1.05	-
R2	0.2 ± 0.02	0.3 ± 0.32	19.36 ± 1.02	6.55 ± 0.35	0.74 ± 0.03	1.87 ± 0.19	65.48 ±1.48	21.14 ±0.96	-
R3	0.4 ± 0.36	0.2 ± 0.15	22.14 ± 0.89	8.21 ± 0.25	0.68 ± 0.05	1.65 ± 0.84	66.98 ±1.98	15.36 ± 0.65	-
R4	0.4 ± 0.15	1.2 ± 0.80	18.56 ± 0.68	5.44 ± 0.14	0.49 ± 0.14	1.55 ± 0.35	77.12 ±0.98	18.35 ±1.47	-
R5	0.3 ± 0.06	0.2 ± 0.13	17.44 ± 1.05	6.77 ± 0.78	0.64 ± 0.18	1.68 ± 0.74	76.48 ±0.46	14.68 ± 0.68	-
P. scoparia									
S1	0.4 ± 0.47	0.2 ± 0.16	45.14 ± 0.94	8.87 ± 1.14	0.64 ± 0.10	2.76 ± 1.15	62.82 ±1.45	23.55 ± 1.28	0.80 ± 0.48
S2	0.3 ± 0.06	0.3 ± 0.64	44.21 ± 0.87	7.65 ± 0.98	0.58 ± 1.01	2.17 ± 1.02	74.35 ±2.15	24.36 ± 1.77	0.74 ± 0.05
S3	0.5 ± 0.03	0.4 ± 0.12	30.88 ± 1.04	9.48 ± 1.17	0.62 ± 0.87	2.34 ± 0.85	70.25 ±1.02	28.14 ± 0.46	0.75 ± 0.32
S4	0.6 ± 0.45	0.6 ± 0.10	28.12 ± 1.44	7.35 ± 0.35	0.40 ± 0.03	1.98 ± 0.67	64.35 ±1.09	20.44 ± 0.98	0.81 ± 0.48
S5	0.5 ± 0.16	1.2 ± 1.05	24.15 ± 0.48	8.78 ± 0.84	0.49 ± 0.15	2.85 ± 1.75	72.18 ±1.87	25.47 ± 1.28	0.65 ± 0.45
P. glaucea									
G1	0.3 ± 0.21	0.2 ± 0.04	27.24 ± 1.02	7.65 ± 2.03	0.59 ± 0.05	2.35 ±0.52	65.88 ±1.85	19.36±1.26	0.69±0.21
G2	0.4 ± 0.03	0.3 ± 0.25	25.33 ± 1.45	8.55 ± 1.25	0.60 ± 0.08	2.66 ± 1.02	64.97 ±2.36	21.44±1.02	0.64 ± 0.36
G3	0.4 ± 0.48	0.6 ± 0.12	31.25 ± 1.36	6.08 ± 1.78	0.48 ± 0.15	1.98 ± 0.36	69.33 ± 1.56	25.35±1.06	0.71±0.58
G4	0.6 ± 0.85	0.7 ± 0.68	55.44 ± 1.45	9.05 ± 0.25	0.62 ± 0.14	2.00 ± 0.54	70.18 ±1.87	30.22±0.87	0.56 ± 0.25
G5	0.7 ± 0.12	0.8 ± 0.96	45.95 ± 1.65	8.44 ± 0.36	0.58 ± 0.20	2.14 ± 1.38	50.64 ± 0.98	33.14±0.85	0.58 ± 0.34
P. arabica									
A1	1.2 ± 0.85	0.5 ± 0.48	24.35 ± 1.48	9.08 ± 1.08	0.65 ± 1.16	3.02 ± 0.85	68.48 ± 0.68	29.32±1.48	0.84 ± 0.48
A2	0.7 ± 0.38	0.3 ± 0.32	41.22 ± 0.98	8.15 ± 1.25	0.63 ± 0.12	2.85 ±0.54	70.48 ± 2.26	33.15±0.58	0.75 ± 0.12
A3	0.5 ± 0.02	0.4 ± 0.38	31.46 ± 0.64	7.45 ± 0.36	0.52 ± 0.65	2.67 ± 0.68	56.39 ±1.54	20.14±0.36	0.68 ± 0.11
A4	0.4 ± 0.09	0.2 ± 0.12	21.45 ± 1.02	6.56 ± 1.52	0.48 ± 0.15	3.44 ± 0.15	64.32 ± 2.38	19.78±1.65	0.58 ± 0.65
A5	0.3 ± 0.45	0.5 ± 0.39	24.57 ± 1.04	9.08 ± 1.45	0.38 ± 0.23	2.61 ± 0.36	50.48 ± 1.87	22.45±2.15	0.47 ± 0.46

The percent oil content of the wild almond kernel ranged from 16% to 55% with a mean of 27%. The highest oil content was recorded for accessions G4 (55%), followed by G5 (46%), S1 (45%), S2 (44%), A2 (41%), E4 (38%), O3 (33%), A3 (36%), G3 (31%) and S3 (31%). Of the 40 accessions, 10 had more than 30% kernel oil content, which is a desirable trait for industrial use. These accessions can be used as a source of fat for industrial purposes.

Seed oil quality and its utility are determined mainly by fatty acid composition. Oil from wild *Prunus* accessions sampled in this study contained palmitic, stearic, oleic and linoleic acid as the major fatty acids. The highest content of linoleic acid was detected in A2 (33.15%), G5 (33.14%) and G4 (30.22%). The content of palmitic acid was highest (9.48%) in S3. The highest content of palmitoleic acid was recorded for E3 (0.83%) followed by E5 (0.76%), O5 (0.74%) and R2 (0.74%). Kernels of A4 had the highest content of stearic acid (3.44%). Linolenic acid was detected in accessions from 'Spartioides' section (*P. scoparia*, *P. glaucea*, *P. arabica*) at levels on par with palmitoleic acid, but was undetectable in accessions from 'Euamygdalus' and 'Lycioides' sections (Table 1).

Wild almond species evaluated in this study were low in saturated fatty acids (SFA) (palmitic and stearic) and high in mono unsaturated fatty acid (MUFA) (oleic acid), the latter ranging from 48.78% to 88.35%. Almonds had low poly unsaturated fatty acid (PUFA), especially linoleic acid, which ranged from 11.32% to 33.15%. In all cases, there was considerable variability in fatty acid profiles between the accessions under study.

Oleic acid is the main determinant of oil quality and high content is favored. The accessions having high oleic acid, were L3 (88.35%), L5 (79.65%), E2 (78.65%) and O5 (78.22%).

Small differences between phenotypic and genotypic coefficients of variation and high estimates of heritability (broad sense) for all characters indicate that the differences between accessions for these oil quality parameters are genetic in nature. The genotypic coefficients of variation were less than the phenotypic coefficients of variation for all characters, indicating the influence of non-additive gene action. Because the coefficient of variation measures the magnitude of variability present in the population, selection from populations with such coefficients of variation is likely to be effective in improvement of the studied traits.

Correlation

Kernel weight was positively correlated with content of oleic acid ($r = 0.59$**), stearic acid ($r = 0.74$**) and palmitic acid ($r = 0.44$**) ($p < 0.05$ in all cases). The correlation between kernel weight and linoleic acid content was significant ($r = 0.34$**, $p < 0.05$). Correlations between palmitoleic acid and oil content with kernel weight were not significant. Shell thickness correlated positively ($r = 0.56$**) with oleic acid content. Shell thickness was negatively correlated with content of palmitic acid and stearic acid. Correlation coefficients for shell thickness–palmitic content and shell thickness–stearic acid content were $r = -0.32$** and -0.46**. Relationships between shell thickness and palmitoleic acid content, linoleic acid content and oil content were not significant (data not shown). Correlations between fatty acid contents were also recorded. The correlation between content of stearic and palmitic acid was positive and significant ($r = 0.46$, $p < 0.05$). Oleic acid content was correlated with palmitic acid content ($r = -0.33$**) and stearic acid content ($r = -0.46$**). Correlations between palmitic acid, palmitoleic acid, stearic acid and linoleic acid content were not significant. However, there was a high negative correlation coefficient ($r = -0.82$**) between content of linoleic and oleic acid.

Genetic distance and clustering of wild almond species

The highest genetic distance (17.9) was reported between genotypes C1 and A5, while the lowest genetic distance (0.5) was between genotypes O1 and E5. The accession-wise mean Euclidean genetic distance varied from a maximum genetic distance of 10.1 in R5 to a minimum genetic distance of 3.9 in R3. UPGMA cluster analysis was computed based on a pairwise Euclidean genetic distance matrix (Fig. 1). Using the MANOVA method, where the optimal number of clusters or groups occurs when the F value is highest; our cutting point was 0.42.

At a genetic distance of 7.93, the 40 accessions were grouped in two main clusters; cluster I with seven accessions (L1, L2, L3, L4, L5, R5 and C1), and cluster II with 33 accessions, which at a genetic distance of 6.69 divided into two subgroups. Subgroup I had 29 accessions, including accessions from 'Euamygdalus' and 'Spartioides' sections; subgroup II with four accessions was from the 'Lyscioides' section and included R1, R2, R3 and R4. Three accessions S3, G3 and A2 in Cluster II had high linoleic and oleic acid content and low palmitic acid content. Cluster I had seven accessions with the following characteristics: L1, L2, L3, L4, L5, R5 and C1 had low linoleic acid; G4, C1, S1, S2, G5 had high total saturated fatty acid percentages; A1 and A4 had the highest stearic acid content; L2 had the lowest linoleic acid content; L1 and L2 had the lowest oil content; C4 had highest kernel weight and low oil content, and A4 had highest stearic acid content and relative low oleic acid content.

Mantel's correlation was 0.89. The pairs of genotypes with large Euclidean distances could be used as parents in a hybridization program for combination breeding or for generating variability for selection of superior pure lines in advanced generations.

Principal coordinate analysis

Principal coordinate analysis (PCoA) was computed by considering all nine variables simultaneously and three of the principal coordinates accounted for more than 80.8% of the total variation encountered (Table 2). The first principal component accounted for more than 40.3% of the total variation due to kernel weight, palmetoleic acid, stearic acid and oleic acid, which all had relatively high and positive weight on this axis.

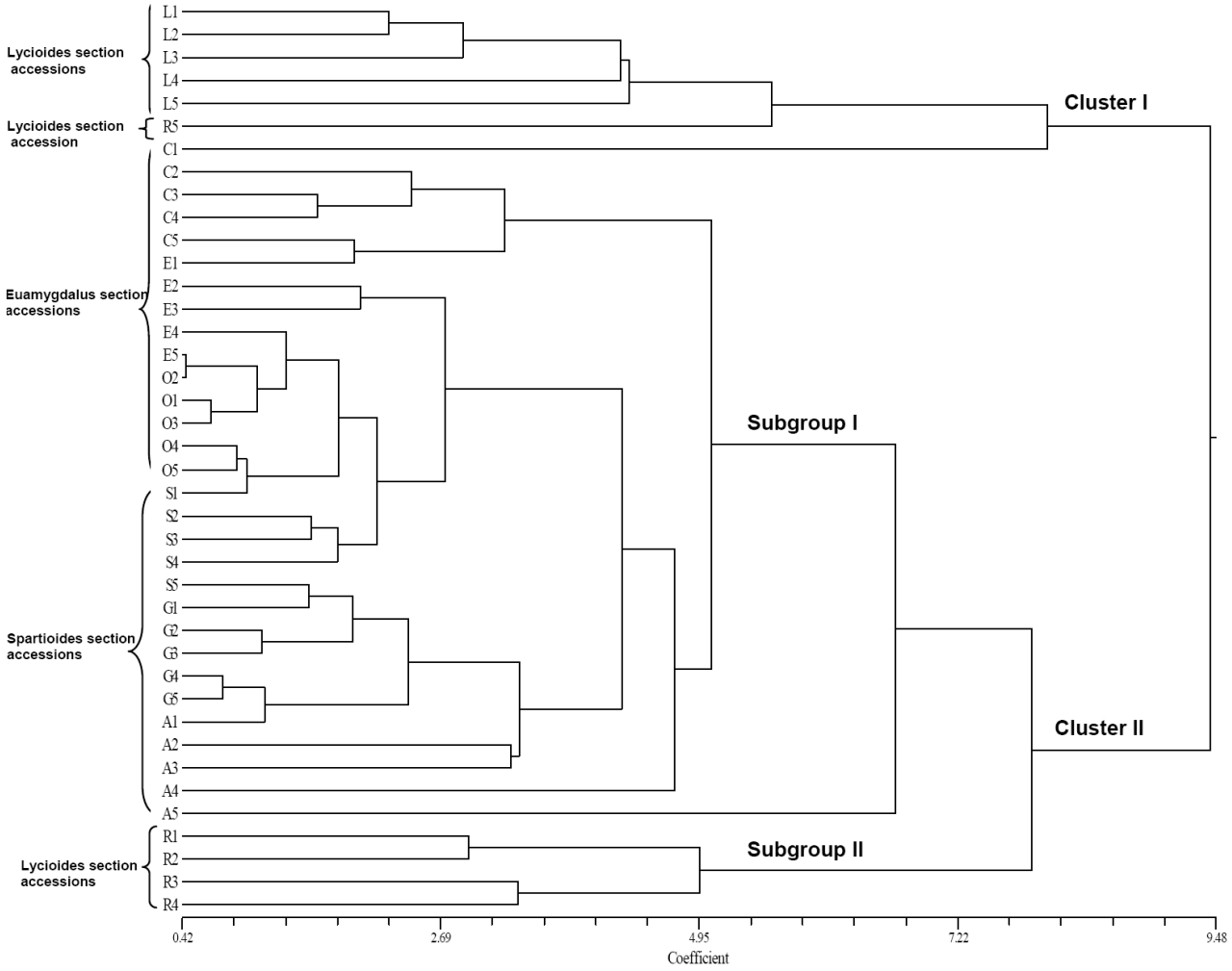

Fig. 1: Dendrogram obtained with the dissimilarity matrix of the biochemical characteristics and unweighted pair group method with arithmetic average clustering algorithm for 40 wild almond (*Prunus*) accessions. The value on the cut point on dendrogram gives the optimal clustering estimated with a MANOVA procedure.

Table 2: Correlation coefficients among the quantitative fruit traits, oil content and fatty acid composition and the first 3 principal components in the wild almond species evaluated.

Traits	Principal coordinate				
	PC1	PC2	PC3	Genotypic variance	Phenotypic variance
Kernel weight (g)	0.982	-0.062	0.079	2613.8	2711.5
Shell thickness (mm)	0.283	0.807	0.044	2741.7	2784.5
Oil content (%)	0.123	0.102	0.223	10.1	10.4
Palmitic acid (C16:0)	0.433	0.736	0.847	8.7	9.5
Palmitoleic acid (C16:1)	0.824	0.144	0.177	7.6	9.2
Stearic acid (C18:0)	0.936	0.785	0.322	7.4	8.2
Oleic acid (C18:1)	0.625	0.474	0.645	7.9	8.6
Linoleic acid (18:2)	0.274	0.364	0.135	2.5	2.7
Linolenic acid (C18:3)	0.342	0.188	0.447	2.3	2.5
component	40.3	25.3	20.2		
Cumulative% of total variance	40.3	65.6	80.8		

The second component accounted for an additional 25.3% of the total variation, depicting primarily the pattern of variation in oil content, which increased at the expense of palmitic acid, stearic acid and shell thickness. The third component accounted for

an additional 20.2% of the total variation, depicting primarily the content of palmitic acid and oleic acid, which were positively correlated.

The pattern of variations illustrated by PCoA was substantiated by the genotypic correlation coefficients determined for pairwise association of the oil quality traits. Consistent with the outputs of PCoA, the traits that contributed most for the 1st principal component were positively correlated with stearic acid.

Discussion

In almond breeding, the most desirable varieties or genotypes have kernel weight over 1.0 g (Askin et al. 2007), although commercial almond varieties exist with kernel weight <1.0 g (Kester et al. 1991; Askin et al. 2007). In native species of *Prunus*, kernel weights varied from 0.6 to 1.0 g (Moradi 2005; Sorkheh et al. 2009). In USA, Spain, Italy and France, most almond varieties or genotypes were reported to have kernel weight of > 1.0 g (Okie 2000). Shell thickness in almond genotypes ranges from 0.2 to 3.0 mm. Thus, it can be said that fruits have hard or semi-hard shells. It has been reported that most native Iranian *Prunus* species have hard-shelled fruits (Sorkheh et al. 2009). Kester et al. (1991) recorded that the varieties from California and Spain have hard-shelled nuts. In addition, hard-shelled almonds are the most preferred varieties in Mediterranean countries.

We recorded oil content at percentages between 16.1% (L2) and 55.4% (G4). In C1, C2, E4, O3, S1, S2, S3, G3, G4, A2 and A3, oil contents exceeded 30%. In various almond varieties and genotypes, oil contents were reported as 52.08–57.49% by Agar et al. (1997), 54% by Gradziel et al. (2000), 53% by Calixto et al. (1981) and 52–56% by Barbera et al. (1994). Martins et al. (2000) reported that 12 almond varieties from Portugal contained 30.1–51.0% oil. Askin et al. (2007) reported that 26 almond varieties from Turkey contained oil at 25.19–60.77%. In this study, although the genotypes showed oil content in the range of the above reports, some genotypes contained more oil than reported for other recognized almond varieties or genotypes. Oleic acid content was between 48.78% (L2) and 88.35% (L3). Linoleic acid content ranged from 11.32% (L2) to 33.15% (A2). Palmitic acid content ranged from 4.58% (O2) to 9.48% (S3). Stearic acid content was between 1.0% (C4) and 3.44% (A4). Palmitoleic acid content ranged from 0.36% (L2) to 0.83% (E3). Gradziel et al. (2000) noted that 'Wood Colony', 'Monterey', 'Padre', 'Butte', 'Nonpareil', 'Carmel', 'LeGrand', 'Fritz', 'Mission', 'Price', 'Sonora', 'Ne Plus Ultra' and 'Aldrich' almond varieties contained 5.0–6.4% palmitic acid, 64.7–76.0% oleic acid and 16.3–26.9% linoleic acid. Askin et al. (2007) reported that rich almond genetic resources were located in the Eastern Anatolia region of Turkey, and contained 50.41–81.2% oleic acid, 6.21–37.13% linoleic acid, 5.46–15.78% palmitic acid, 0.80–3.83% stearic acid, and 0.36–2.52% palmitoleic acid.

Results for fatty acid composition in this study showed that some wild almond species contained higher oleic, stearic, linoleic and palmitic acids than some other *Prunus* species (Kodad et al 2004; Askin et al. 2007). The use of genotypes with higher fatty acid composition might contribute to almond improvements directed at increasing nutritional value.

Linoleic acid content was negatively correlated with oleic acid content (r = - 0.79**). Kodad et al. (2004) and Askin et al. (2007) reported similar results but our correlation was lower than that reported by Askin et al. (2007).

Kernel weight was positively correlated with the content of palmitic, stearic and oleic acids. This indicates that almond genotypes with high kernel weight have higher content of oleic, stearic and palmitic acids, but lower content of linoleic acid. Shell thickness was negatively correlated with content of palmitic and stearic acids, but positively correlated with content of oleic acid: thicker-shelled almond genotypes had higher oleic acid content. Our results are supported by those of Askin et al. (2007).

This study suggests that compositional improvements of almond could be achieved through the use of wild almond species in hybridization programs. In addition, wild almond species investigated here are rich in oleic acid and can be considered as potential vegetable oils in the human diet. Plant sterols or phytosterols resemble cholesterol and account for approximately 0.3–2% of plant oil but more than 10% in certain plants (Stuchlik and Zak 2002). Phytosterols have attracted the interest of food chemists because they are of great importance for food labeling and nutritional purposes. They are also characteristics used in testing the authenticity of vegetable oils (Crane et al. 2005).

Our study will aid characterization of almonds and their close relatives, and provides a knowledge base for almond breeders who formulate breeding programs to develop the potential oil almond production and its commercial exploitation in Asia.

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