Study of the Genetic Diversity of Almond Seedling Populations in Morocco: Application of a Chemometric Approach

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Abstract

Almond (*Prunus amygdalus* Batsch) in Morocco is still propagated by farmers mostly from seed, generating a large genetic diversity. Thus, in order to evaluate the almond diversity in Morocco from the point of view of kernel quality, oil and protein contents and major fatty acid composition were determined. Principal component analysis (PCA) was used to compare the kernel components among 46 genotypes selected from different production regions, as well as five introduced cultivars. Oil and protein contents were highly variable between genotypes, ranging respectively from 48.29 to 65.19% and from 14.07 to 36.48% of the total kernel dry weight. Fatty acid composition of the oil fraction ranged between 5.6-8.34% for palmitic acid, 0.37-0.87% for palmitoleic acid, 1.3-3.3% for stearic acid, 57.95-81.97% for oleic acid, and 9.69-29.98% for linoleic acid. Clustering of genotypes from similar regions suggested the existence of parental relationship among these genotypes and, as a consequence, a common ancestral origin. The Moroccan genotypes did not cluster separately from the introduced varieties. Two genotypes had very high oil contents whereas four genotypes had exceptionally high protein contents (> 30%). One genotype had a very high oleic content (82%). The large variability observed for oil and fatty acid composition and the presence of genotypes with higher oil and fatty acid contents than the foreign cultivars, represent a very promising base to obtain new Moroccan almond cultivars with oil of higher quality.

INTRODUCTION

Almonds are grown in Morocco in several regions under different environmental conditions. About 55% of the almond trees grown in Morocco are seedlings, located primarily in the north and the south (Lansari et al., 1994). The genetic variation in local Moroccan almond populations is assumed to be widespread because of the large geographic distribution, different environmental conditions, dominance of seed propagation, and the presence of peach × almond natural hybrids (Barbeau and El Bouami, 1980; Lansari et al., 1994). Several studies were undertaken to evaluate the genetic variation in these populations, showing the presence of a great variability between genotypes of the same population (Lansari et al., 1994; Oukabli et al., 2006) and between populations (Lansari et al., 1998). Selection of local almond genotypes for late-bloom, frost resistance, and diseases have been carried out since 1975 (Barbeau and El Bouami, 1979; Laghezali, 1985). Other studies allowed the selection of materials with a high

physical kernel quality (Lansari et al., 1994; Oukabli et al., 2006) and others with high yield potential due to a high spur density (Lansari et al., 1994).

However, no studies have been carried out to evaluate the chemical quality of the kernels in these local almonds population. In other countries, such as in Spain (Romojaro et al., 1988), USA (Abdallah et al., 1998), Australia (Vezvaei and Jackson, 1996) and Turkey (Askin et al., 2008), the evaluation of the chemical components of the commercial and local almond genotypes allowed a better characterisation of genotypes, opening the possibilities of incorporating the best genotypes in breeding programs for improving kernel quality. The chemical composition of the kernel is essential when considering the different industrial applications and the high diversity of almond confectioneries (Socias i Company et al., 2008). Thus, our objective was to determine the oil content and composition for a set of selected almond genotypes from local populations of Morocco.

MATERIALS AND METHODS

Plant Material

This study was carried out in three populations from different regions with wealthy almond genetic resources: the Rif mountains (north of Morocco), the Atlas mountains and the valley of Tadla (central-south). Almond genotypes were selected from different zones of each region in 2008. Trees representing a selected genotype were marked in their native population. A total of 46 local genotypes and six commercials cultivars (‘Marcona’, ‘Desmayo Largueta’, ‘Ferragnès’, ‘Fournat de Brézenaud’, ‘Ferraduel’ and ‘Khoukhi’) were included in this study.

Oil and Fatty Acid Determination

After blanching, the kernels were ground in an electrical grinder. Oil was extracted from 4-5 g of ground almond kernel in the commercial fat-extractor Soxtec for 2 hr using petroleum ether as solvent and keeping the heating source at 135 ºC. The oil sample was utilized to prepare the methyl esters of the corresponding fatty acids (FAMEs) by trans-etherification according to the official method UNE-EN ISO 5509:2000 (February 2001). These methyl esters were separated using a flame ionization detector (FID) gas chromatograph. The major fatty acids detected were: palmitic (16:0), palmitoleic (16:1), stearic (18:0), oleic (18:1), and linoleic (18:2).

RESULTS AND DISCUSSION

Oil content varied significantly between genotypes, ranging from 48.29 to 65.19% of kernel dry matter, similar to previous reported data (Abdallah et al., 1998; Kodad et al., 2004a). Protein content was highly variable among genotypes, ranging from 14.07 to 34.3% of the total kernel dry weight. This range of variability is wider than those reported on others studies (Kodad et al., 2004b; Askin et al., 2008). Fatty acid composition of the oil kernels ranged between 5.6-8.34% for palmitic acid, 0.37-0.87% for palmitoleic acid, 1.3-3.3% for stearic acid, 57.95-81.97% for oleic acid, and 9.69-29.98% for linoleic acid. Kernels of Ag6 contained the highest oleic acid content (81.97%) and the lowest linoleic acid (9.69%). This genotype could be introduced in almond breeding programmes to improve the kernel quality, since a high oleic content in kernel oil is considered an index of good quality (Kester et al., 1993).

The first 3 principal components accounted for 36%; 31%, and 19% of the variance, respectively. The oleic and linoleic acid contents are the variables that explained

the largest portion of the variance, indicating that these components were more variable than oil and protein contents and those of the other major fatty acids.

The cluster obtained with the components studied shows a dendrogram (Fig 1) of similarity among the local genotypes and between these genotypes and the foreign cultivars grown in Morocco. According to this algorithm, two groups can be established at a scaled distance of 1, further separating two local genotypes from the rest. The first individualised genotype was AT8, characterised by the lowest values of oil and oleic acid contents, but with the highest values of linoleic acid and protein contents (Table 1). The second individualised genotype was Ag6 from the Rif mountains, with the highest value of oleic acid and the lowest of linoleic acid, and intermediate values of oil and protein contents (Table 1).

The first group contained 15 genotypes from the Rif mountains, one from the Atlas mountains and one from the Valley of Sfasif, and the foreign cultivars 'Fournat de Brézenaud', 'Desmayo Largueta' and 'Khoukhi'. This group shows low to intermediate values for oleic acid and for the ratio of oleic to linoleic acids, as well as high values for linoleic acid. Thus, the kernels of this group may be susceptible to rancidity and show lower oil stability. The genotypes DH8, DH3 and DH7, and the cultivars 'Fournat de Brézenaud' and 'Desmayo Largueta' show a similar chemical composition.

The second group is formed by eight genotype from the Atlas mountains, seven from the Rif mountains and the foreign cultivars 'Ferragnès', 'Ferraduel' and 'Marcona' (Fig 1). This group was characterised by an intermediate to high content of oleic acid and a low content of palmitic, palmitoleic and linoleic acids. This group shows a high ratio of oleic to linoleic acid, which is correlated with lower tendency to rancidity and greater oil stability (Kester et al., 1993). The genotypes Ik2, Ik4, Ik8 and Ak3 showed the highest values of oil, protein and oleic acid content than the foreign cultivars. This result must be evaluated over several years in order to consider their releas in their native area as new cultivars, as well as their incorporation in almond breeding programmes as parents to improve kernel quality. The high similarity between 'Marcona', appreciated in the fabrication of nougat (Socias i Company et al., 2008), and the genotype Tg2 indicates the possibility selecting genotypes with a high kernel quality aimed at producing nougat of high quality in Morocco.

The high similarity in the profile composition between some genotypes originating from a same region (Sa1 and Sa2; Ik5 and Ik6; DH4 and DH9; DH3 and DH7; DH1 and DH5) could be due to the paternity relationships existing in the local almond cultivars (Lansari and Lakhal, 2001), since in Morocco seed propagation was the main technique of almond propagation used in rural areas. Kodad et al. (2004a), applying a similar statistical analysis in an almond population obtained from different crosses, observed that the genotypes obtained from the same cross clustered in the same group.

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Table 1. Kernel oil and fatty acid composition for local almond genotypes and cultivars.

Genotype	Origin	Oil	Protein	C16:0	C16:1	C18:0	C18:1	C18:2	R1
Ag1	Rif mountains	55.1	28.6	6.2	0.6	2.0	74.5	16.0	4.7
Ag2		57.3	26.2	7.3	0.6	1.7	70.0	19.9	3.5
Ag3		57.1	24.6	6.6	0.6	2.2	74.4	15.8	4.7
Ag4		54.8	33.0	6.6	0.5	2.1	71.2	19.2	3.7
Ag6		58.2	21.3	5.6	0.4	1.9	82.0	9.7	8.5
Ak1		56.2	28.1	7.3	0.6	1.7	69.9	19.9	3.5
Ak3		60.3	21.0	5.6	0.4	2.3	73.3	18.1	4.1
Sa1		60.9	19.9	7.1	0.5	1.9	65.9	23.9	2.8
Sa2		60.0	18.2	6.8	0.4	1.9	62.6	27.5	2.3
Sa5		59.9	21.1	7.7	0.5	2.3	65.8	23.2	2.8
Sa6		57.3	25.1	7.0	0.8	1.7	74.0	16.6	4.5
DH 1		52.4	29.1	6.5	0.5	1.9	67.3	23.1	2.9
DH 2		58.8	27.3	6.2	0.5	1.9	74.2	16.7	4.7
DH 3		53.4	26.9	6.7	0.5	2.2	66.3	23.8	2.8
DH 4		48.3	34.3	6.6	0.4	1.6	67.2	23.8	2.8
DH 5		53.9	28.3	6.4	0.5	2.2	67.4	22.8	3.0
DH 7		51.5	21.6	6.2	0.5	1.8	65.5	25.3	2.6
DH 8		57.9	23.4	7.8	0.6	1.8	68.3	21.1	3.2
DH 9		49.0	31.0	6.2	0.4	2.0	69.1	21.6	3.2
DH 10		57.8	26.9	6.3	0.5	2.9	67.6	22.4	3.0
Ik1	Atlas mountains	65.2	14.1	6.9	0.5	3.1	67.3	21.4	3.1
Ik2		62.8	18.2	7.2	0.5	1.7	71.1	19.0	3.7
Ik4		59.2	22.2	7.5	0.8	1.3	75.3	14.6	5.2
Ik5		54.9	22.5	7.7	0.8	2.2	65.2	23.6	2.8
Ik6		56.8	22.2	7.4	0.6	1.9	65.3	23.9	2.7
Ik7		53.5	29.3	7.2	0.6	2.0	72.9	16.8	4.4
Ik8		62.1	18.7	7.9	0.9	1.6	71.0	18.0	4.0
AT8		49.8	30.9	7.7	0.5	3.3	58.0	30.0	1.9
Sf1	Sais valley	54.5	30.3	6.3	0.5	2.8	68.9	21.9	3.2
Sf2		57.6	26.0	6.6	0.5	1.6	67.8	22.8	3.0
Sf3		55.7	26.8	6.6	0.6	1.5	66.9	23.8	2.8
Marcona	Spain	54.6	27.7	6.6	0.6	2.1	71.9	18.1	4.0
D. Largueta		56.4	27.0	6.9	0.5	2.1	64.8	25.0	2.6
Ferragnès	France	57.9	22.6	6.4	0.5	2.0	74.5	16.1	4.6
Ferraduel		62.5	17.8	5.5	0.5	2.2	75.1	16.1	4.7
Fournat		58.1	20.9	6.8	0.6	2.1	69.1	20.6	3.4
Khoukhi	Tunisia	53.9	28.9	7.1	0.6	1.6	66.0	24.1	2.7

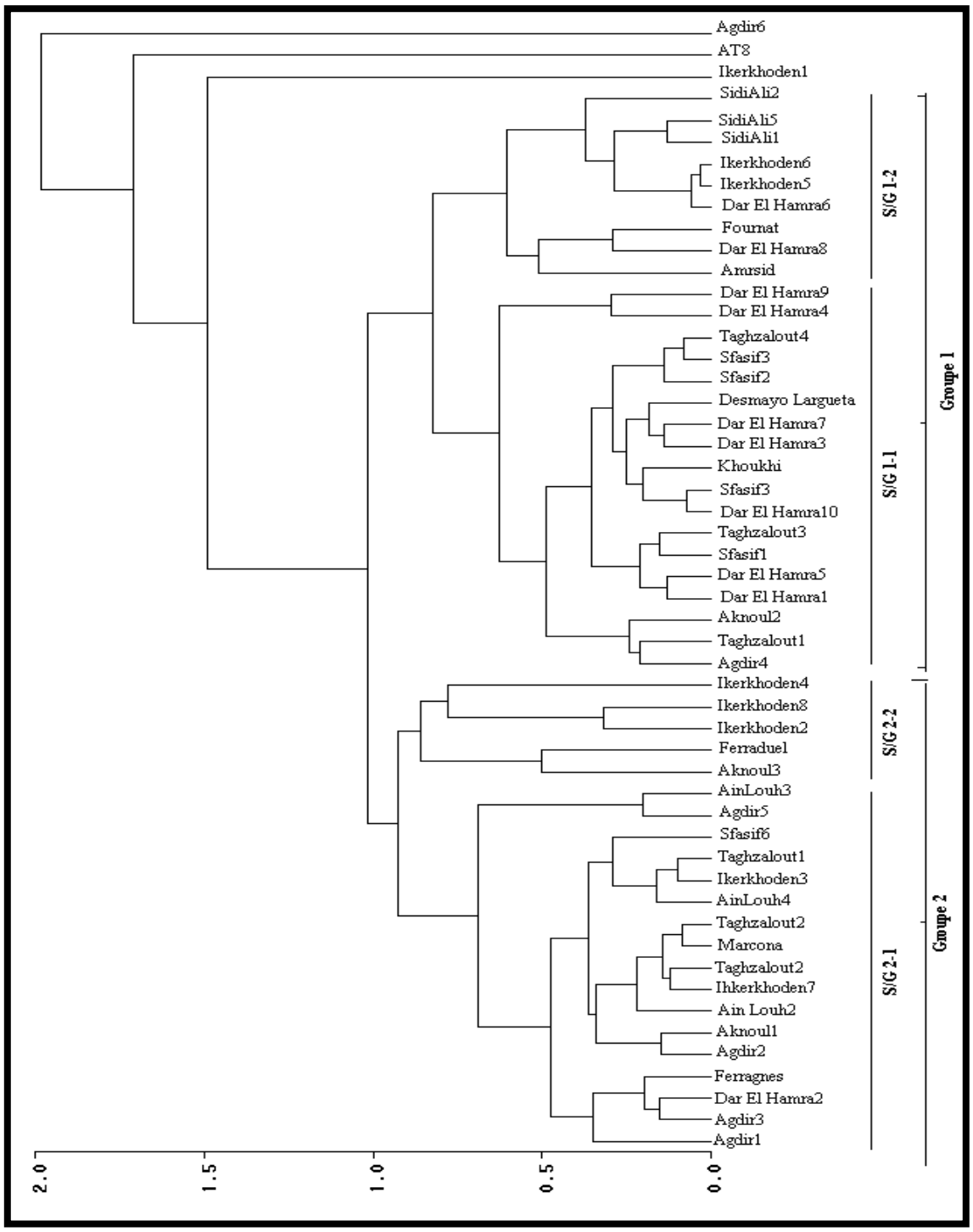

Figure 1. Dendrogram showing the results of the cluster analysis for fatty acid composition.