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The Study of Phenotypic Variation of 'Shahrood1' × 'Shahrood12' Population and their Comparison with the Parents using Morphological Markers

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Abstract

The plant genetic resources are important, because they have most valuable national basis resources in the each country. In this study, to grouping the 92 almond progenies from 'Shahrood1' × 'Shahrood12' and comparing them in terms of heritability with their parents, measured on the basis of almond universal descriptor. Principal component analysis (PCA) was used to compare 15 morphological and phonological traits among study progenies. The important traits frequency such as flowering time, tree size, resistance to frost, growth habit, flower density and leaving time in all hybrid population and compare them with their parents showed their asymmetric distribution among them. The simple correlation coefficients traits showed that among some measured traits was a significant positive correlation. In other hand, the eigenvalue variance percentage and cumulative variance showed the among measure traits, three independent factors that their eigenvalue were more than seven, they could justified 60% of total variance. Also the results of cluster analysis all cultivars were divided into three main groups. some of progenies likes 'Shahrood1' in third group, some other likes 'Shahrood12'were in second group and some other that expression of phenotypic traits were additive mode, incomplete dominance or over dominance in first group.

Keywords: Cluster analysis, Correlation matrix, Morphological trait, Principal Component Analysis (PCA), Progenies, *Prunus dulcis*.

Introduction

Today, the most important and economic way to improve yield and quality is using desirable varieties with adaptable to limiting conditions such as water stress, heat, cold, salinity, nutrient-poor soil and resistant to diseases and pests. Currently there are different breeding programs in around the world for transferring desirable genes from wild species of plants to various crops progress (Baninasab and Rahemi, 2007). Phenotypic and genotypic variation studies are important for identifiable superior genotypes to maintain, evaluate, and use in breeding programs (De Giorgio and Polignano, 2001).

The almond (*Prunus amygdalus*, syn. *Prunus dulcis, Amygdalus communis, Amygdalus dulcis*) is a species of native tree of the Middle East and South Asia. Most of botanists believe that the origin of almond is Iran (Asma *et al.*, 2007; Rasouli *et al.* 2013; Sarkhosh *et al.*, 2006). Twenty-six almond species form a distinct and easily identified taxonomic group in the world (Gulcan, 1985). In Iran 21 almond species and 6 naturalprogenies have been described (De Giorgio *et al.*, 2007). Almond cultivation in Iran has a long historical background, and because of its self-incompatibility nature, there are many genotypes growing in different regions of the country. These genotypes include a vast range of diversity in many characteristics such as blooming time. One of the major concerns of modern agriculture is the conservation and utilization of valuable genetic resources of crop plants (Chalak *et al.*, 2007).

Today, for evaluate the cultivars and genotypes based on qualitative and quantitative characteristics and their relationships need to use multivariate statistics. This method can be an index between the independent and dependent traits (Mohammadi and Parmasa, 2003).

Reported of Talhook *et al.* (2000) on wild species *A. commonis* and *A. orientalis* showed the fruit weight with fruit size, shell thickness and kernel weight correlated significantly. Also kernel size is correlated with thickness of shell.

Factor analysis is a powerful method of multivariate statistical techniques that can group studied traits effectively. In way that, Lansari *et al.*, (1994) for diversity evaluation of almond cultivars and clones morphologically used Factor analysis. Their results showed that the kernel and

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fruit properties in variation of almond cultivars and clones are more important than leaving properties. Therefore, to classification of progenies and analysis the genetic relationships between progenies populations, one of the best ways, are using the multivariate statically methods. Among these methods, principal component analysis (PCA) technique has more application than the other available methods. Factor analysis is one of the other multivariate statically methods that reduces the number of studied traits and places them into the effective groups. This method has been used by Lansari *et al.* (1994), De Giorgio and Polidnano (2001), De Giorgio *et al.* (2007) and Chalak *et al.* (2007) in order to grouping and separating of almonds genotypes and cultivars. Also, Rasouli *et al.* (2013) in phenotype evolution some of 72 almond cultivars and genotypes using morphological markers showed that the 30 traits studied in 11 factors based on PCA were data reduction and classified with cluster analysis. In this study, 72 different varieties of almonds that collected in all over Iran were compared and correlated morphological traits and analyzed together. The 30 traits studied in 11 factors based on PCA were data reduction and classified with cluster analysis.

Therefore, aim of at present study was to investigate the Phenotypic variation of 'Shahrood1' × 'Shahrood12' population and their comparison them with the parents using morphological markers. The main purpose of this present study was the identification and analysis of Phenotypic special characteristics of 'Shahrood1' × 'Shahrood12' population and their comparison them with the parents using morphological markers in Karaj region almond collection, in order to reach to the promising progenies with special features of performance.

Materials and Methods

In this study 92 progenies from 'Shahrood1' × 'Shahrood12' and their parents evaluated using 19 morphological features during 2012 and 2013. The characters coded according to Gulcan (1985) descriptor (Tabal 1). The study morphological traits such as tree size (TS), Growth habbit (GH), Bearing habbit (BH), Branches Number (BN), Leaving time (LET), Flowering time (FT), Flower size (FLS), Flower color (FC), Resistance to Frost (RF), Flower density (FD), Position of Pistil to stamen (SP), Fruit stage (FS), Color fruit (CF), Fruit fuzz (FF), Fruit shape (FSH), Kernel Color (KC) and Fruit Maturity (FM) were determined on the basis of the International almond descriptor (Gulcan, 1985). Flowering time was figured by calculating days from the onset to the end of flowering. Fruit Maturity was the harvest date. For statistical analysis, Fruit Maturity was represented as the number of days from 1 August.

Statistical analysis

Data analysis was performed using SPSS (Version 21.0) such as trait frequency, descriptive statistics, simple correlation, principal component analysis and cluster analysis. Factors separation was done using the Principal component analysis (PCA) and maximum of variance and in each Principal and independent factor, factor coefficient was considered 0.5 or higher as significant.

Results

Traits Frequency

Evaluated progenies in some of the traits had distribution of normal relatively. Frequency in flowering time based on Gulcan (1985) descriptor indicates about 23% hybrid population from 'Shahrood1' × 'Shahrood12' close to one parent ('Shahrood1') and other hand about 8% for flowering time trait similar to other parent ('Shahrood12') and was late flowering. The leaving time, tree size, resistance to frost, growth habit and flower density were presented in (Fig.1).

Traits simple correlation coefficients

At present study, there were significant correlations among some traits. For example resistance to frost has with flowering time (r =0.5) correlated positively. There is a positive correlation for the branches number with fruit fuzz (r=0.52), tree size (r=0.66), bearing habit (r=0.50), leaving time (r=0.52), flower size (r=0.50), flower density (r=0.58) and fruit color (r=0.66). Furthermore flower density have positive correlated with leaving time (r =0.54), branches number (r =0.58), tree size (r =0.58), bearing habit (r =0.52), fruit fuzz (r =0.52) and fruit color(r =0.54). On the other hand, tree size correlated with fruit fuzz, (r=0.54), bearing habit (r=0.57), branches number (r = 0.66) flower size (r = 0.72), flower density (r=0.58), fruit size (r=0.50) and fruit color (r=0.65). Also flower size with fruit color (r= 0.58), branches number (r=0.50), bearing habit (r=0.50) and tree size (r= 0.72) has correlated positively. Also fruit size has correlated with flower color (r = 0.55), leaving time (r = 0.50), and tree size (r=0.50) (Table 2). These findings corresponded with Talhouk *et al.* (2000), Rasouli *et al.* (2013) and Baninasab and Rahemi, (2007).

Factor analysis

The table 6 shows eigenvalue variance percentage and cumulative variance of 15 traits factor analysis among which factors 1, 2 and 3 implement most portions in variance justification.

The eigenvalue indicates percentage of variance and cumulative variance that (pc1) (pc2) and (pc3) represent the largest variance (Table 3).

In other hand, the eigenvalue, variance percentage and cumulative variance showed the among measured traits three independent factors

that their eigenvalue were more than seven, they could justified 60% of total variance.

According to the eigenvalue for different traits can be said some traits such as tree size, bearing habit, branches density, and immature fruit color and leaving time have an important role in distinguishing the progenies. While all of these traits have been at the first group principle factor, it creates 43% of the total variance. In other hand, fruit color, the stamen to pistil relative and flowering time in second group principle factor indicate 9% of the total variance (Table 4).

Cluster analysis

In this study, cluster analysis based on all traits measured (Table 1) done Ward method (Fig.2). The cultivars were divided into two main groups at distance of 10, so one parent (‘Shahrood12’) was at a cluster and other parent (‘Shahrood1’) was in other. From important factors to separate varieties in this traits distance were fruit shape, fruit color, flower color, flower time, resistance to frost, leaving time, growth habit, branches number, and leaving time. The cultivars were divided into three main groups at the 5 distance. In other words, the progenies were teammate with ‘Shahrood12’, in this distance divided into two distance groups. The important traits for separating this clusters were fruit size, position of pistil to stamen, flower size, flower time, growth habit. The ‘Shahrood12’ in group 2 was teammate with 10, 30, 55, 64, 73, 74, 83 and 91. Also the ‘Shahrood1’ in group 3 was teammate with 1, 4, 11, 43, 74, 83 and 86. Therefore progenies teammate with ‘Shahrood1’ and ‘Shahrood12’ to be used in breeding programs as parents in hybridization crosses. At the first group with 34 progenies were variations in studied traits. So the progenies in some traits were like ‘Shahrood1’ and for some other traits like ‘Shahrood12’, or the expression of phenotypic traits were additive mode, incomplete dominance or over dominance. For example, in number of 84 progenies for fruits shape trait was like one parent (‘Shahrood1’), for resistance to frost was like other parent (‘Shahrood12’) and for leaving time was like additive mode and in flower size was like over dominance.

Table1. The traits evaluation methods based on almond descriptor (Gulcan 1985).

Trait	obrivation	Measure method
Tree size	TS	3=Weak،5=Intermediate ،7=Strong
Growth habbit	GH	1= Extremely upright ،3= upright ،5= Spreading ،7= Dropping ،9= Weeping
Bearing habbit	BH	1= Most flower buds on one year old shoots ،2= Most flower ،3= Mixed
Branches Number	BN	0= without ،3= low ،5= medium ، 7= large ،9= very large
Leaving time	LT	1= extra early ،2= very early ،3= early ،4= early medium ،5= medium ،6= medium late ،7= late ، 8= very late ، 9= super late
Flowering time	FT	1= extra early ،2= very early ،3= early ،4= early medium ،5= medium ،6= medium late ،7= late ، 8= very late ، 9= super late
Flower size	FS	3= small ،5= medium ،7= large
Flower color	FC	1= white ،2= white pink ، 3= pink
Resistance to Frost	RF	1= sensitive ، 3= medium sensitive، 5= medium ، 7= medium resistance ،9= resistance
Flower density	FD	1= low ، 3= low medium ، 5= medium large, 7= large
Position of Pistil to stamen	SP	1= Pistil equal with stamen ، 3= Pistil to stamen ، 5= Pistil taller and equal with stamen ، 7= Pistil shorter and equal with stamen ، 9= Pistil shorterl with stamen
Fruit stage	FS	1= extra early ،3= early ،5= medium ، 7= late ، 9= super late
Color fruit	CF	3= white ، 5= white green ،7= green
Fruit fuzz	FF	1= low ، 3= low medium ، 5= medium large, 7= large
Fruit shape	FS	1= round ، 2= ovate ، 3= oblong ، 4= cordate ، 5= extremely narrow

Table 2. The simple double correlation of 15 measured almond progenies traits in this study.

	FS	FF	CF	FS	SP	FD	RF	FC	FS	FT	LT	BN	BH	GH	TS
Fruit shape	1														
Fruit fuzz	0.09	1													
Fruit color	0.35*	0.45*	1												
Fruit stage	0.35*	0.38*	0.55**	1											
Position of Pistil to stamen	0.24*	0.10	0.3*	0.16	1										
Flower density	0.27*	0.52**	0.54**	0.34*	0.32*	1									
Resistance to frost	0.14	0.36*	0.40*	0.35*	0.01	0.27*	1								
Flower color	0.12	0.30*	0.41*	0.17	0.08	0.17	0.29*	1							
Flower size	0.27*	0.44*	0.58**	0.33*	0.15	0.37*	0.25*	0.46*	1						
Flower time	0.38*	0.30*	0.34*	0.30*	0.33*	0.34*	0.29*	0.10	0.10	1					
Leaving time	0.34*	0.52**	0.66**	0.50**	0.30*	0.54**	0.50**	0.37*	0.45*	0.58**	1				
Branches number	0.26*	0.52**	0.64**	0.44*	0.21*	0.58**	0.35*	0.35*	0.50**	0.20*	0.52**	1			
Bearing habit	0.31*	0.53**	0.64**	0.40*	0.20*	0.52**	0.37*	0.31**	0.50**	0.32**	0.66**	0.50**	1		
Growth habit	0.12	0.30*	0.31*	0.12	0.10	0.23*	0.17	0.20*	0.37*	0.23*	0.22*	0.42*	0.36*	1	
Tree size	0.43*	0.54**	0.65**	0.50**	0.24*	0.58**	0.21*	0.44*	0.72**	0.32**	0.59**	0.66**	0.57**	0.36*	1

Table 3. Eigenvalue, percentage of variance and cumulative variance for the 3 prime factors.

Factors	eigenvalue	percentage of variance	cumulative variance
1	60.423	420.823	420.823
2	10.400	90.330	520.153
3	10.074	70.163	590.316

Table 4. Eigenvalue of the different traits in the 3 main factors.

Traits	PC1	PC2	PC3
Fruit shape	0.466	0.452	0.256
Fruit fuzz	0.685	-0.199	-0.224
Color Fruit	0.834	-0.013	0.030
Fruit size	0.625	0.168	-0.212
Position of Pistil to stamen	0.356	0.533	0.420
Flower density	0.707	0.126	0.068
Resistance to frost	0.520	-0.074	-0.635
Flower color	0.495	-0.417	0.032
Flower size	0.695	-0.367	0.305
Flower time	0.512	0.592	-0.198
Leaving time	0.819	0.194	-0.244
Branches	0.769	-0.200	0.108
Bearing habit	0.770	-0.044	-0.064
Growth habit	0.457	-0.260	0.256
Tree size	0.838	-0.123	0.234

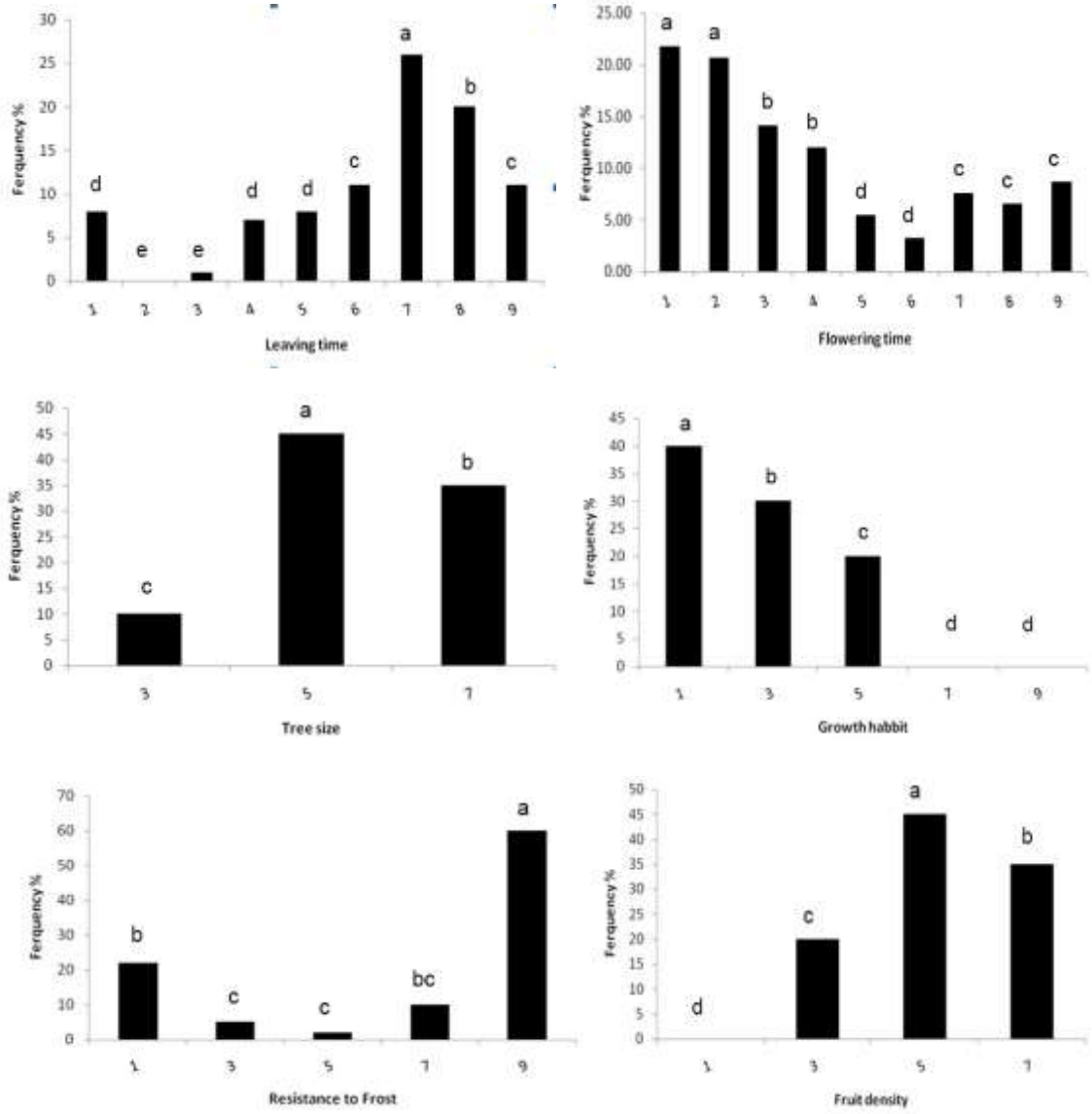

Fig. 1. Frequency of leaving time, growth habit, flowering time, fruit density, resistance to frost and tree size in study progenies based on Gulcan descriptor.

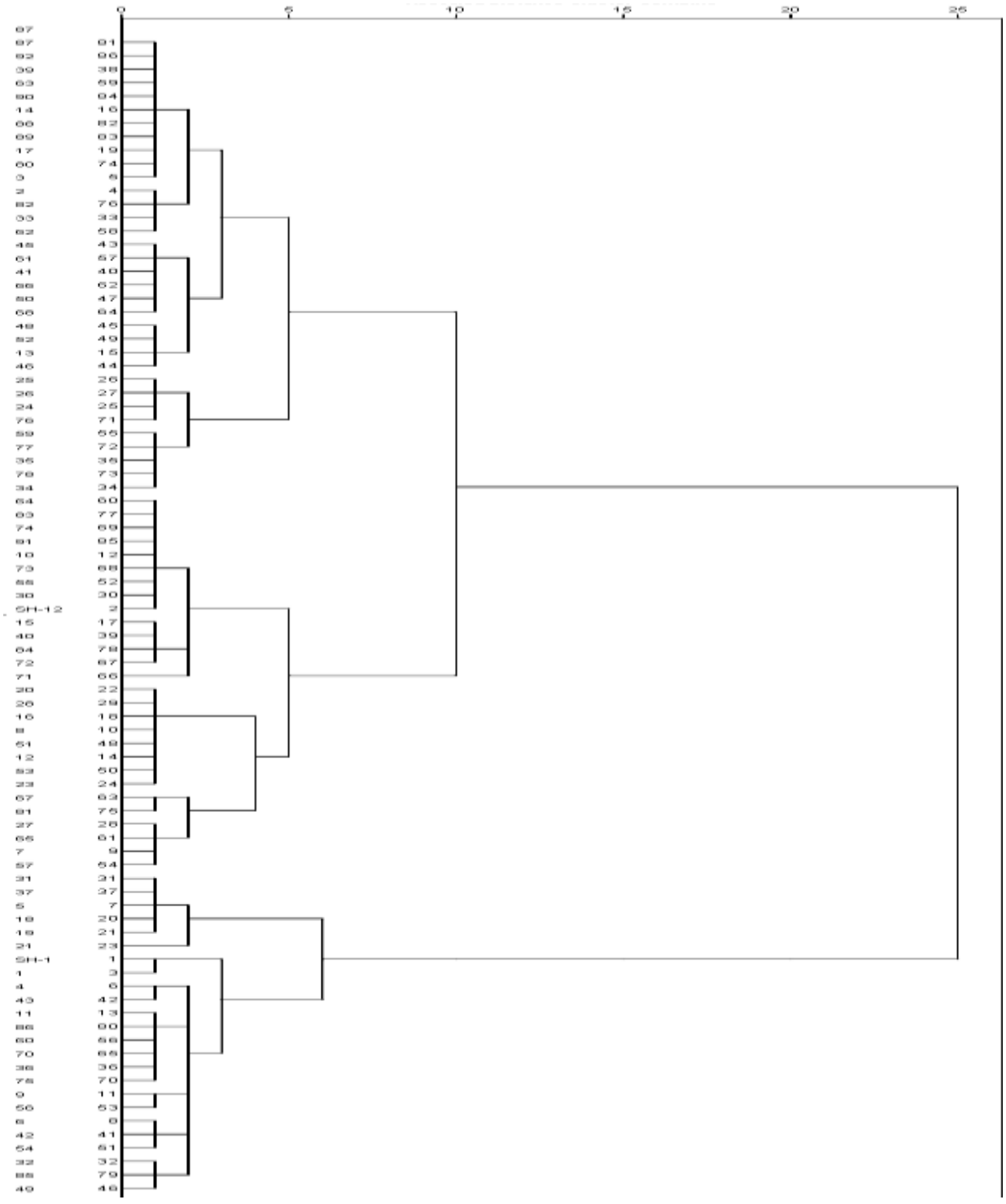

Fig. 2. The Dendrogram of Cluster analysis in 92 study progenies ($Sh_1 \times Sh_{12}$) with their parents using Ward's method.

Discussion

The success of breeding programs is dependent on the availability of diversity genetic resources, and using hybridization can be generated variation based on scientific requirements (Kester and Gradziel, 1996). Therefore, studies on morphology of almond hybrids and their comparison by their parents are important valuable genes for using heterosis and hybridization (Garcia *et al.*, 1996). Lansari *et al.* (1994) for diversity evaluation of almond cultivars and clones morphologically used factor analysis. Their results showed that the

kernel and fruit properties in variation of almond cultivars and clones are more important than leaving properties. These results are consistent with this study results.

Almond cultivation in Iran has a long historical background, and because of its self-incompatibility nature there are many genotypes growing in different regions of the country.

The results of pomological traits indicated that tree habit growth, buds, leaf, flowers and fruit attributes were from a high diversity among studied progenies. Also time of flowering among almond progenies varied widely and as early flowering, middle flowering and late. Performances of almond progenies based on their quantity and quality characteristics were different. Similar results have been reported by Ledbetter and Shonnard (1992); Lansari *et al.* (1994); Karl *et al.* (1998); Talhouk (2000), De Giorgio and Polidnano, (2001); Fatahi *et al.* (2004); Sarkhosh *et al.* (2006); De Giorgio *et al.* (2007); Asma *et al.* (2007) and Chalak *et al.* (2007) in order to grouping and separating of almonds genotypes and cultivars. This study needs to be completed by a DNA analysis, in order to highlight the influence of environmental conditions on the variability in the progenies studied and to confirm the genetic distances among accessions.

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