

# Morphological, Molecular, and Self-(In) Compatibility Characteristics of New Promising Apricot Genotypes

H. Pinar<sup>1\*</sup>, S. Ercisli<sup>2</sup>, M. Bircan<sup>3</sup>, M. Unlu<sup>3</sup>, A. Uzun<sup>1</sup>, K. U. Yilmaz<sup>1</sup>, and M. Yaman<sup>1</sup>

## ABSTRACT

World apricot (*Prunus armeniaca* L.) production is increasing steadily due to breeding of new high yielding cultivars in different countries. More recently, breeding programs have been modified according to consumers' demands and also improvement in resistance to diseases (Sharka, Monilinia etc.), frost damages, and determination of self-(in) compatibility. In this study, fourteen apricot breeding progenies and six of their parents were evaluated by using both morphological and molecular markers. As morphological markers, fruit weight, width, length, height, total soluble solids, acidity, and fruit firmness were used. In molecular analysis, to determine genetic relationships, Sequence-Related Amplified Polymorphism (SRAP), Inter-Simple Sequence Repeat (ISSR) and Damage-Associated Molecular Patterns (DAMP) markers were used. In addition, SRc-F/R markers were used to determine S allele profile. The results showed that, although there were no earlier genotypes than Ninfa and Priana, Ay×P3 cross was a promising genotype with regard to earliness and fruit characteristics. A total of 224 scorable bands obtained with 8 SRAP primer combinations (25 bands), 8 DAMP primers (81 band) and 16 ISSR primers (118 bands) showing high diversity among crosses and cultivars. A total of 4 S-RNase alleles (S<sub>C</sub>, S<sub>2</sub>, S<sub>3</sub>, S<sub>6</sub>) were identified in this study and the most widely identified alleles were S<sub>C</sub> and S<sub>3</sub> alleles.

**Keywords:** Cross breeding, Hybridization, New variety, *Prunus armeniaca*.

## INTRODUCTION

Turkey dominates world apricot production with 795,000 tons yearly production, followed by Iran (465,000 tons) and Italy (247,000 tons) (FAO, 2012). Apricot production in the world is increasing steadily year-by-year and such increase is closely related to breeding activities carried out in different apricot producing countries. Apricot breeding programs are mostly modified to meet consumers' demands and preferences and also based on certain traits such as resistance to diseases (Sharka, Monilinia etc.), frost damages, and self-compatibility (Hormaza *et al.*, 2007; Ercisli, 2009; Yilmaz and Gurcan, 2012).

The primary goal in fruit tree breeding programs is to develop new cultivars with desired characteristics in an economical way. Despite the rich diversity in common apricot germplasm, high degree of heterozygosis within the species, most of the time, slow down the breeding processes. Production in many countries comes from chance seedlings and local cultivars (Bassi, 1999). Limited market value of local cultivars and poor adaptability of cultivars out of their native area are considered among the primary factors limiting the expansion of apricot growing sites in the world (Badenes *et al.*, 1998). Although the objectives in apricot breeding programs differ based on the country and on intent of

<sup>1</sup> Department of Horticulture, Faculty of Agriculture, Erciyes University, 38039, Kayseri, Turkey.

\* Corresponding author; email: hasanpinar@erciyes.edu.tr

<sup>2</sup> Department of Horticulture, Faculty of Agriculture, Ataturk University, 25240, Erzurum, Turkey.

<sup>3</sup> Alata Horticultural Research Institute, 33740, Erdemli/Mersin, Turkey.



use (dry, fresh, or canned), there are certain selection criteria commonly applied to most apricot breeding programs (Hormaza *et al.*, 2007).

More recently, climatic adaptation is considered one of the basic targets in the majority of apricot breeding programs. Apricot cultivars have highly specific ecological requirements. Thus, commercial production is commonly limited to certain locations where generally a couple of cultivars account for large portion of the production. Then, there is a need to evaluate apricot cultivars for high and reliable yield in each production site. Adaptation works are performed based on location and such adaptation works commonly involve breeding for late blooming cultivars to avoid frost damages, breeding for early blooming (low chilling) in frost-free regions, breeding to develop early ripening cultivars, or breeding for greater mid-winter cold hardiness in colder regions. Local adaptations are mostly expressed in terms of productivity and regularity of production and such adaptations are directly related to specific environmental conditions (Hormaza *et al.*, 2007).

Apricot cultivars are mostly evaluated by morphological, molecular, and self-(in) compatibility characteristics (Ercisli, 2009). Morphological characteristics are among the most significant quality attributes affecting consumers' preferences. For apricots, large size (more than 60 g), attractive appearance (a bright blush over bright orange or cream), firmness, freestone, uniform ripening, and resistance to skin cracking are the primary quality attributes. Good orange skin and flesh colors, as well as uniform medium size, regular shape, good texture, high sugar content, small pit, and a good balance of acid and sugar are preferred for canned apricots. On the other hand, high soluble solids, medium-large size and good texture are required for dried apricots (Ercisli, 2009).

The molecular marker technology, with a rapid development during the last 20 years, offered various new approaches in

identification of genetic diversity among the cultivars and, today, molecular markers are efficiently used in plant systematics, breeding, and assessment of gene sources (Kaczmarek *et al.*, 2015; Mishra *et al.*, 2015; Nemli *et al.*, 2015; Wojnicka-Poltorak *et al.*, 2015). So far, different DNA-based marker techniques such as Restriction Fragment Length Polymorphism (RFLP), Random Amplification of Polymorphic DNA (RAPD), Amplified Fragment Length Polymorphism (AFLP), Simple Sequence Repeats (SSR) have been applied to assess the genetic diversity and relationships between apricot cultivars.

Species of the family Rosaceae show Gametophytic Self-Incompatibility (GSI) to prevent inbreeding depression. This intercellular reaction is controlled by a single multi-allelic locus, the *S*-locus (de Nettancourt, 1977). The *S*-gene product in styles is a Ribonuclease enzyme (*S*-RNase) (McClure *et al.*, 1989), while the pollen product is an *F*-box protein (Romero *et al.*, 2004). Knowledge of the self-compatibility of commercial cultivars and selections from breeding programs is imperative for any apricot breeder hoping to design compatible combinations (Burgos *et al.*, 1998). In apricot, 7 *S*-alleles (*S*<sub>1</sub>-*S*<sub>7</sub>) were described in North American and Spanish apricot cultivars for self-incompatibility, while the *SC*-allele was identified as responsible for self-compatibility (Albuquerque *et al.*, 2002). Later, 9 additional alleles (*S*<sub>8</sub>-*S*<sub>16</sub>) were found using Non-Equilibrium pH Gradient Electro focusing (NEpHGE) and Polymerase Chain Reaction (PCR) analyses (Halász *et al.*, 2005). From Chinese cultivars, more than 50 *S*-alleles have been identified based on *S*-RNase activity staining, PCR, and sequencing (Jie *et al.*, 2005; Zhang *et al.*, 2008; Wu *et al.*, 2009; Halász *et al.*, 2012). In total, 17 Cross Incompatibility Groups (CIG) are known, including North American, European, Turkish, and Tunisian apricot cultivars (Szabó and Nyéki, 1991; Egea and Burgos, 1996; Halász *et al.*, 2010; Milatovic *et al.*, 2010; Lachkar *et al.*, 2013).

In Turkey, the first planned apricot-breeding program was initiated in 1989 to develop new table apricot cultivars; and with this program, 5 apricot cultivars (Dr. Kaska, Çagataybey, Çagribey, Sahinbey and Alata Yildizi) were released (Bircan *et al.*, 2010). In addition, a cross breeding study between Turkish (Alata Yildizi, Çagribey, Çagataybey and Sakit-6) and foreign (Priana, Feriana and Precoc de Colomer) parents were initiated. Thus, in the present study, the aim was to select 14 crossbreed genotypes with regard to earliness, tree structure, fruit yield, fruit characteristics and development status and, for the first time, assess them with regard to pomological characteristics, self-compatibility, and genetic similarity.

## MATERIALS AND METHODS

In this study, a total of 20 apricot genotypes (14 of them are new hybrid and 6 of them are their Turkish and foreign parents) were used. The 14 new hybrids were obtained in cross breeding study within the table apricot breeding program initiated at Alata Horticultural Research Institute. This breeding program was started in 1989 by using Alata Yildizi, Çagribey and Çagataybey as Turkish parents and Priana, Feriana and Precoc de Colomer as foreign parents. The 14 crossbreed genotypes were selected with regard to earliness, tree structure, fruit yield, fruit characteristics, and development status. For pomological characteristics, fruit analyses on crosses were performed in 2013, 2014, and 2015 and average of 3 years were provided for all traits, except for harvest dates. Average fruit weight, width, length, height, seed weight, fruit firmness, Total Soluble Solids (TSS%), titratable acidity (%), TSS/acid ratio, and pH were determined. Samples were taken from 3 trees in 3 replications with 25 fruits in each replication. Data gathered through quality analyses on fruit samples were subjected to statistical analyses with JMP software and means were compared with

Tukey test. Sequence-Related Amplified Polymorphism (SRAP), Inter-Simple Sequence Repeat (ISSR) and Damage-Associated Molecular Patterns (DAMP) marker systems were used to identify genetic similarities among the crosses and parents. S allele analysis was also performed on crosses and parents. In molecular study, total genomic DNA was extracted from fresh leaf tissues by using the CTAB method described by Doyle and Doyle (1990). A microplate spectrophotometer (BioTek Instruments Inc. Winooski, USA) was used to measure DNA concentrations and 10 ng  $\mu\text{L}^{-1}$  DNA templates were prepared by using TE (10 mM Tris-HCl, 0.1 mM EDTA, pH 8.0). Eight DAMP primers (URP1F, URP2F, URP2R, URP4R, URP6R, URP9F, URP13R and URP17R), eight combinations of 14 forward and 16 reverse SRAP primers and sixteen ISSR primers (CA8R, VHVGTG7, GACA4, DBDACA7, GT8YA, AGC6G, HVHCA7T, AG7YC, CT8T6, TCC5RY, BDBCA7C, HVHTCC7, GA8YG, CAA6, AG8T, GT6GG) were used to target the amplification of marker sequences from genomic DNA of genotypes. For SRAP (Li and Quiros, 2001), ISSR (Uzun *et al.* 2015) and DAMP (Karaca and Ince, 2008), PCR conditions were used. Polymerase chain reaction (PCR) products were separated on 2.5% agarose gel at 90V for 4 or 5 hours. Amplified DNA bands were visualized with ethidium bromide staining. The fragment patterns were photographed under UV light for further analysis. A 100 bp standard DNA ladder was used as the molecular standard in order to confirm the appropriate markers.

Each band was scored as present (1) or absent (0) and data were analyzed with Numerical Taxonomy Multivariate Analysis System (NTSYS-pc) software (Rohlf, 2000). A similarity matrix was constructed by using SRAP, DAMP and ISSR data based on Simple Matching (SM) coefficient. Then, the similarity matrix was used to construct a dendrogram with UPGMA (Unweighted-Pair Group Method Arithmetic average) to determine genetic relationships among the cultivars studied. The genetic similarity



matrix and ultrametric distance matrix produced from UPGMA-based dendrogram with CPH module nested in the same software was compared using Mantel's matrix correspondence test (Mantel, 1967).

*SRc-R* (5'-GGC CATTGT TGC ACA AAT TG-3') (Vilanova *et al.*, 2005) and *SRc-F* (5'-CTC GCT TTC CTT GTT CTT GC-3') (Romero *et al.*, 2004) primer pairs were used to determine *S* allele composition of apricot genotypes (Vilanova *et al.*, 2005). The PCR products were electrophoresed in 2% (w/v) metaphor agarose stained with ethidium bromide ( $0.5 \text{ } \mu\text{g mL}^{-1}$ ) using 1X TAE buffer at 110V for 2 hours and visualized under UV light. Molecular size of the amplified fragment was estimated using a 100-bp ladder (Thermo).

## RESULTS AND DISCUSSION

New apricot cultivars are mostly characterized with their high fruit quality attributes satisfying consumers' preferences (Ruiz and Egea, 2008). The pomological characteristics of new apricot crosses and parents are shown in Table 1. The differences in fruit quality attributes of the crosses and cultivars were found to be significant at 5% level (Table 1).

Earliness is the main desired attribute in fresh apricots grown in the Mediterranean region of Turkey (Polat and Caliskan, 2013). Feriana was the earliest harvested cultivar in 2013 (May 9) followed by Ninfa (May 14), AyxP3 (May 20) and FerxCol9 (May 25), respectively. The latest harvest was performed on June 7 in FerxCol12 genotype. In 2014, the earliest harvest was performed on May 8 in Feriana cultivar, on May 9 in Ninfa cultivar, on May 17 in AyxP3, FerxCol12 and FerxCol15 genotypes. In 2015, the earliest harvest was performed on May 5 in Feriana cultivar, on May 9 in Ninfa cultivar, on May 16 in FerxCol12 genotype and May 12 in FerxCol15 genotype. Polat and Caliskan (2013) reported Beliana and Feriana (May 20) as the earliest and Precoc de Colomer as the

latest (June 7) ripening apricot cultivars in eastern Mediterranean in Turkey. In previous researches, ripening dates of apricot cultivars were reported as between May 14-June 26 in Spain (Ruiz and Egea, 2008), between June 11-September 10 in Hungary (Hegedus *et al.*, 2010) and between May 26 - June 25 in Italy (Lo Bianco *et al.*, 2010).

Fruit size is another significant quality attribute in apricot effecting consumers' preferences. The greatest fruit weight was observed in AyxP3 genotype (65.10 g) and the lowest value was seen in FerxCol5 (25.52 g) genotype and Priana (24.95 g) cultivar. The genotypes AyxP5 (64.98 g) and AyxP7 (64.17 g) had the greatest fruit weights after AyxP3 genotype. The greatest fruit width was again observed in AyxP3 (47.76 mm) genotype, and it was followed by AyxP5 (47.27 mm) and AyxP7 (46.85 mm) genotypes. The lowest fruit width was observed in Priana (32.87 mm) cultivar. Fruit length was the highest in AyxP3 (50.13 mm) genotype and the lowest in Priana (34.41 mm) cultivar. With regard to fruit height, the greatest value was observed in AyxP5 (50.13 mm) genotype and the lowest in FerxCol10 (33.26 mm) genotype. With regard to seed weight, the highest value was seen in FerxCol7 (5.03 g) genotype and the lowest in FerxCol10 (2.52 g) genotype. Fruit flesh firmness is an important quality attribute for export and shelf life in local markets. Although Ninfa is an early cultivar, it is negatively affected in export because of low flesh firmness. The greatest flesh firmness was observed in CgrxCol1 ( $3.44 \text{ kg cm}^{-2}$ ) and the lowest in FerxCol5 ( $0.87 \text{ kg cm}^{-2}$ ). The TSS content is an important quality parameter with significant impacts on fruit taste (Polat and Caliskan, 2013). Total soluble solid (TSS) content was the highest in CgrxCol1 (15.11%) and the lowest in FerxCol9 (9.58%) genotype. The greatest titratable acidity was observed in FerxCol10 (3.18%) genotype and the lowest in the Ninfa (1.22%) cultivar. The highest TSS/acid ratio was seen in the Cagataybey (11.08%)

Table 1. Harvest dates and some fruit quality parameters of apricot genotypes and their parents from 2013, 2014, and 2015 growing seasons.

No	Genotype name	Harvest Dates			Fruit Characteristics																	
		2013	2014	2015	Fw <sup>a</sup>	Fwe <sup>b</sup>	Fh <sup>c</sup>	Fs <sup>d</sup>	Sw <sup>e</sup>	Ff <sup>f</sup>	Fa <sup>g</sup>	TSS (%) <sup>h</sup>	TSS/Fa ratio									
1	AY x P-5	June 05	May27	June 03	64,98	a	47,27	a	48,50	a	50,13	a	3,20	d-j	2,07	c-h	1,42	d-g	10,43	f-h	7,44	b-e
2	AY x P-7	June 06	May26	June 02	64,17	a	46,85	ab	47,53	a-c	48,66	ab	2,95	f-k	2,45	b-e	1,49	c-g	11,57	d-g	7,79	b-d
3	AY x Pr-3	May20	May17	May20	65,10	a	47,76	a	47,96	ab	48,68	ab	4,53	ab	1,74	d-i	1,73	b-f	12,55	b-e	7,34	b-e
4	Cgr x Col-1	May28	May19	May20	41,17	d-h	36,00	d-g	40,13	d-j	41,13	e-g	3,47	c-i	3,44	a	1,87	b-e	15,11	a	8,11	bc
5	Cgty x Col-8	May30	May25	May20	31,17	f-i	36,33	d-g	37,12	i-k	36,61	h-k	2,52	i-k	2,45	b-e	1,92	b-d	14,54	a	7,60	b-d
6	Fer x Col-10	May30	May28	May25	47,39	c-e	41,89	bc	44,15	a-e	45,05	b-e	3,71	b-g	2,70	a-c	3,19	a	10,53	f-h	3,41	ij
7	Fer x Col-12	June 07	May17	May16	30,78	f-i	33,16	g	34,58	jk	33,26	k	2,63	i-k	1,56	e-j	1,84	b-f	11,28	e-h	6,17	d-g
8	Fer x Col-14	May29	May20	May16	48,84	b-d	39,87	c-e	43,62	a-f	43,71	c-e	4,34	a-c	2,14	c-h	1,59	c-g	11,04	e-h	7,19	b-f
9	Fer x Col-15	May28	May17	May12	44,24	d-f	35,56	d-g	38,35	g-k	39,02	f-i	3,94	b-e	1,31	h-j	1,51	c-g	11,64	d-g	7,72	b-d
10	Fer x Col-21	June 04	May21	May22	44,83	d-f	42,22	bc	42,69	c-h	42,35	d-f	2,88	f-k	2,24	b-f	1,40	e-g	11,98	c-f	8,62	b
11	Fer x Col-4	June 05	May30	May27	61,40	a-c	46,45	ab	47,51	a-c	47,06	a-c	3,69	b-h	0,87	j	2,20	b	10,83	e-h	5,04	g-i
12	Fer x Col-5	May27	May25	May22	25,52	i	32,89	g	36,17	jk	37,68	g-j	2,81	g-k	1,54	f-j	3,69	a	11,22	e-h	3,09	j
13	Fer x Col-7	May27	May25	May27	42,71	d-g	40,45	cd	42,97	b-g	42,22	d-f	5,03	a	2,18	c-g	1,37	e-g	9,58	h	7,01	b-f
14	Fer x Col-9	May25	May22	May25	26,59	hi	34,61	g	35,17	jk	35,31	i-k	2,99	e-k	1,75	d-i	3,48	a	13,47	a-d	3,91	h-j
15	Alata Yildizi	May25	May27	May21	46,97	c-e	39,89	c-e	44,62	a-d	44,17	c-e	2,99	e-k	2,41	b-e	1,78	b-f	14,18	ab	7,99	b-d
16	Cagataybey	May25	May24	May22	36,43	d-i	33,57	g	39,45	d-k	42,23	d-f	2,60	i-k	1,87	c-i	1,35	fg	14,84	a	11,08	a
17	Cagribey	May25	May23	May22	33,79	e-i	34,85	e-g	38,59	f-k	39,27	f-i	3,18	d-j	1,79	d-i	1,93	bc	11,54	d-g	6,42	c-g
18	Feriana	May09	May08	May05	28,94	g-i	32,09	g	34,94	jk	34,55	jk	2,74	h-k	1,08	ij	1,96	bc	10,96	e-h	5,61	f-h
19	P. de Colomer	May17	May14	May12	39,40	d-i	42,00	bc	39,25	e-k	37,75	g-j	2,90	f-k	1,74	d-i	1,62	c-g	9,98	gh	6,34	d-g
20	Priana	May09	May09	May07	24,95	i	32,87	g	34,41	k	34,54	jk	2,43	jk	1,34	g-j	1,50	c-g	10,46	f-h	7,11	b-f
LSD					14,60		5,05		5,21		4,02		0,97		0,84		0,51		1,94			1,72

<sup>a</sup> Fruit weight; <sup>b</sup> Fruit weight; <sup>c</sup> Fruit height; <sup>d</sup> Fruit size; <sup>e</sup> Seed weight; <sup>f</sup> Fruit firmness; <sup>g</sup> Fruit acidity; <sup>h</sup> Total Soluble Solids.

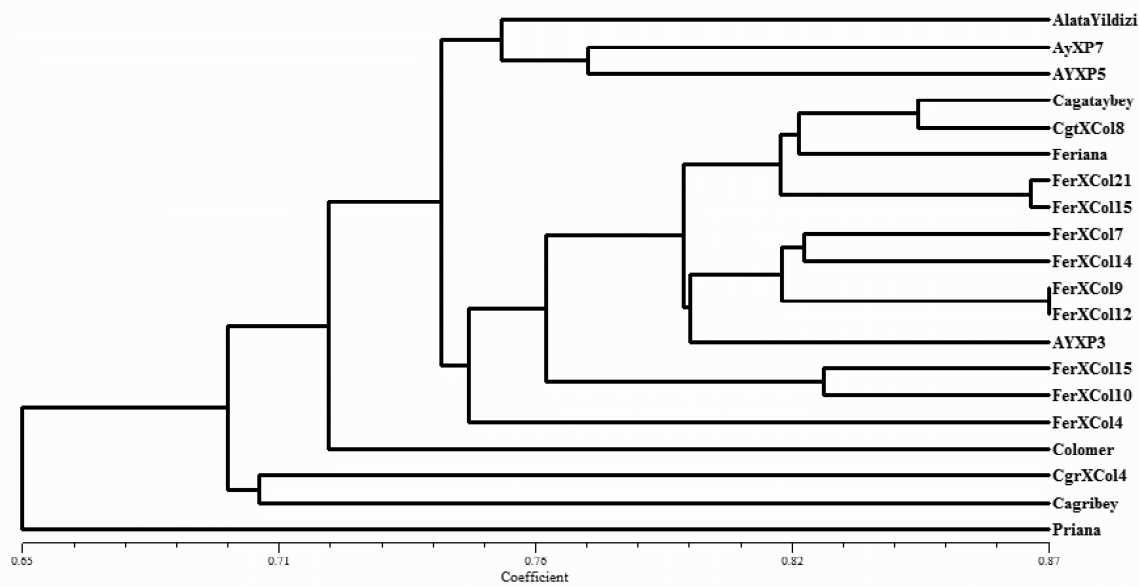


cultivar, the lowest TSS/acid ratio was observed in Ay×P5 (2.95%) genotype. Polat and Caliskan (2013) assessed the phenological and pomological characteristics (flowering period, average fruit weight, yield, fruit dimensions, flesh/seed ratio, acidity, and total soluble solids) of seven apricot cultivars (Beliana, Canino, Precoce de Colomer, Feriana, Rouge de Sernhac, Tokaloğlu, and Macar) in Hatay, Turkey. In that study, the highest average fruit weight was obtained from Rouge de Sernhac (37.9 g), while Feriana had the lowest (30.9 g) value; flesh/seed ratio was the highest in the Canino (16.8), which had the highest TSS content with 14.5%, whereas Beliana had the lowest TSS value with 10.6% and researchers recommended Beliana and Feriana cultivars for early production in Hatay. In the present study, Cgr×Col1 hybrid was prominent with TSS value.

In a previous breeding program, Bircan *et al.* (2010) used Alyanak, Sakit-1, Sakit-2, Sakit-6, 07-K-11 as local and Cafona, Canino, Fracasso, Joubert Foulon and Precoce de Colomer as foreign apricot cultivars and reported that 370 of 4,173 crosses contained desirable traits. Among them, 5 new apricot cultivars were registered as Dr. Kaska, Çagataybey, Çagribey, Sahinbey and Alatayildizi. Dr. Kaska has juicy and freestone fruits. Its fruits are sweet and aromatic. Çagataybey is freestone and fruits are juicy, aromatic, and more colored. Çagribey is freestone, juicy, and aromatic. Alata Yildizi is also freestone and has good fruit quality. In that study, Alata Yildizi had the greatest fruit weight and Çagataybey had the highest TSS (%) value. In the present study, the greatest fruit weight was observed in AY×Pr-3 crosses and the highest TSS content was observed in Cgr×Col-1 crosses. Pinar *et al.* (2008) determined that 2-89 genotype had the highest yield, Harcot had the largest fruit and Bebeco had the firmest fruits. While Ninfa, Priana, Precoce de Tyrinthe was found as the earliest cultivars, 15-90, Fracasso, Sahinbey were determined

as the latest genotypes in the Turkey from the same breeding program.

Yilmaz and Gurcan (2012) indicated that a precise characterization and discrimination of the cultivars were pre request for breeding of promising apricot cultivars. Different markers including morphological, molecular, and biochemical markers were employed in apricot. ISSR markers were used by Chenjing *et al.* (2005) and Yilmaz *et al.* (2012) and SRAP markers were used by Uzun *et al.* (2010). In the current study, a total of 224 scorable bands were obtained with 8 SRAP primer combinations (25 bands), 8 DAMP primers (81 band) and 16 ISSR primers (118 bands). A dendrogram was constructed by using the UPGMA method. There was a high cophenetic correlation ( $r= 0.86$ ;  $P<0.01$ ) between ultrametric similarities of tree and similarity matrix. Such a correlation indicated that constructed dendrogram strongly represented the similarity matrix. Current accessions had similarity values ranging from 0.65 to 0.87, indicating a high level of variation (Figure 1). Priana was identified as the most distinct cultivar with a similarity value of 0.65. Cagribey and Cgr×Col4 were identified as the second distinct ones with a similarity value of 0.69. Cagribey and Cgr×Col4 nested at the same cluster that Cagribey is progeny of Cgr×Col4. Also, Ay×P7 and Ay×P5 were placed at the same cluster with Alata Yildizi. Notably, Ay×P3 nested away from Ay×P5, while and Ay×P7 nested close to Priana because Ay×P3 progenies were obtained from Alata Yildizi x Priana crossing. There were nine Fer×Col progenies obtained from Feriana×Colomer crossing and they nested among Feriana and Colomer. There was one Çagataybey×Colomer progeny (Cgt×Col18) nested in the same cluster with Çagataybey cultivar. Progeny of Cagribey (Cgr×Col14) also nested in the same cluster with Cagribey. Uzun *et al.* (2010) carried out a study and determined genetic diversity and relationships among Turkish and some foreign apricot cultivars by using SRAP markers. The researchers obtained 87 bands



**Figure 1.** Dendrogram of 20 apricot genotypes (14 of them are new hybrid and 6 of them are their parents) obtained from SRAP, ISSR and DAMP markers.

and 63 of them (73%) were found to be polymorphic. Similarity values among the apricot cultivars were identified as between 0.77-0.97.

According to *S* allele results, some genotypes have only amplification of just one allele-*S* (Table 2). This could be a problem with the primers used or some alleles have few differences in molecular weight and also with the methods used. Vilanova *et al.* (2005) determined *S*-genotypes of total 20 apricot genotypes by using *SRc-F* and *SRc-R* consensus primers and compared the sizes of PCR products with previously published ones. A total of 4 *S-RNase* alleles (*S<sub>C</sub>*, *S<sub>2</sub>*, *S<sub>3</sub>*, *S<sub>6</sub>*) were identified in this study (Table 2). The most widely identified alleles were *S<sub>C</sub>* and *S<sub>3</sub>* (occurred in 10) and they were followed by, respectively, *S<sub>6</sub>* (9) and *S<sub>2</sub>* (3) alleles. Group of Fer×Col progenies had mostly *S<sub>C</sub>* allele like Colomer, except Fer×Col4 which carried *S<sub>3</sub>S<sub>6</sub>* coming from Colomer and Feriana. Besides, Ay×P5 and Ay×P7 had the same alleles (*S<sub>2</sub>S<sub>3</sub>*), but Ay×P3 had *S<sub>6</sub>* allele. Cagataybey yielded 355 bp bands and had *S<sub>C</sub>* allele. Also, Cgt×Col8 had *S<sub>C</sub>* and *S<sub>6</sub>* alleles coming from Cagataybey and Colomer. Cagribey had *S<sub>3</sub>* and *S<sub>6</sub>* alleles like Cgr×Col14. There were

no inconsistent results among parents and their progenies. Burgos *et al.* (1997) identified self-incompatibility to Priana carrying *S<sub>2</sub>* and *S<sub>7</sub>* alleles. In the present study, *S<sub>2</sub>* allele was identified in Priana and its two crosses (Ay×P5 and Ay×P7). Similar to other fruits, self-incompatibility is also a significant problem in apricots. As in other *Prunus* species, there is a gametophytic incompatibility in apricots usually controlled by a pair of *S* alleles (de Nettancourt, 2001). Similarly, Sonneveld *et al.* (2001) indicated that self-incompatibility in Rosaceae family was gametophytically observed with multi-allelic *S* loci. Faust (1989) carried out a study with inbred apricot genotypes under field conditions and indicated that “the individuals with a fruit set ratio below 5%” could be defined as self-incompatible; “the ones with 5-10% fruit set” could be defined as semi-compatible, and “the ones with more than 10% fruit set” could be defined as self-compatible. In previous molecular studies carried out until 2007, 20 different *S*-incompatibility alleles and a Self-Compatibility (*S<sub>C</sub>*) allele were identified in European apricots (Burgos *et al.*, 1998; Halasz *et al.*, 2005; Halasz *et al.*, 2007a; Halasz *et al.*, 2007b). Gulcan *et al.* (2006)


**Table 2.** S Allele profiles of 20 apricot genotypes.

		SrcF-R		1 <sup>st</sup> Allele	2 <sup>nd</sup> Allele
1	AlataYildizi	270	420	S <sub>3</sub>	S <sub>6</sub>
2	Ay×P3	420	-	S <sub>6</sub>	-
3	Ay×P5	270	332	S <sub>3</sub>	S <sub>2</sub>
4	Cagataybey	355	-	Sc	-
5	Cgt×Col8	355	420	Sc	S <sub>6</sub>
6	Fer ×Col5	355	-	Sc	-
7	Ay×P7	270	332	S <sub>3</sub>	S <sub>2</sub>
8	Fer×Col7	270	355	S <sub>3</sub>	Sc
9	Colomer	355	420	Sc	S <sub>6</sub>
10	Fer×Col14	355	420	Sc	S <sub>6</sub>
11	Fer×Col12	355	-	Sc	-
12	Fer×Col21	355	420	Sc	S <sub>6</sub>
13	Fer×Col15	270	355	S <sub>3</sub>	Sc
14	Fer×Col10	270	355	S <sub>3</sub>	Sc
15	Fer×Col4	270	420	S <sub>3</sub>	S <sub>6</sub>
16	Fer×Col9	355	-	Sc	-
17	Cagribey	270	420	S <sub>3</sub>	S <sub>6</sub>
18	Cgr×Col4	270	420	S <sub>3</sub>	S <sub>6</sub>
19	Priana	332	-	S <sub>2</sub>	-
20	Feriana	270	-	S <sub>3</sub>	-

evaluated local apricot genotypes in “National Apricot Genetic Sources Plot” of Malaya Apricot Research Institute of Turkey and indicated 32 of 64 local genotypes as self-compatible. In the present study, presence of S<sub>C</sub> allele was identified in promising genotypes and such alternative cultivar candidates for early table apricot culture may significantly increase production.

Apricot (*Prunus armeniaca* L.) is particularly prone to erratic productions and species are adapted to narrow spaces (Layne *et al.*, 1996). Therefore, the majority of apricot cultivars have highly specific ecological requirements and relatively lower yields are often experienced when the cultivars are grown in other regions. Climatic adaptation has become the subject matter of several apricot breeding programs (Hormaza *et al.*, 2007). However, the reasons for such low adaptability levels have

not been clarified, yet (Julian *et al.*, 2007). In recent years, apricot breeding programs were carried out in several countries and various new cultivars were developed. Bellini *et al.* (2008) implemented long-term breeding works since 2000 and selected 13 genotypes with ripening dates seven days before and three days after Aurora. Ruiz *et al.* (2010) indicated that the works to develop new apricot cultivars along with consumers’ preferences have been initiated since the early periods of 1900s. The researchers reported that they had developed 5 new cultivars, namely, Toni, Estrella, Sublime, Maravilla, and Rosa. Topor *et al.* (2010) initiated apricot breeding programs in Romania for late-ripening cultivars and developed three new apricot cultivars (Euxin, Histria and Augustin). Pennone *et al.* (2010) carried out cultivar development researches to meet the needs of producers and processing industry and registered two



early-ripening cultivars (Ischia and Procida). Bircan *et al.* (2010) initiated apricot breeding works in 1989 to develop new table apricot cultivars through hybridizations of local and foreign cultivars and developed and registered 5 new apricot cultivars (Dr. Kaska, Çagataybey, Çagribey, Sahinbey, and Alata Yildizi). Egea *et al.* (2010) developed 9 new apricot cultivars (Rojo Pasion, Selene, Murciana, Dorada, Toni, Estrella, Sublime, Maravilla and Rosa) complying with market demands. Demirtas *et al.* (2010) carried out an apricot breeding program to develop new apricot cultivars resistant to spring early freezes. In another multi-purpose apricot breeding carried out in Malatya (Turkey), an early-ripening apricot cultivar “Dilbay” was developed and it was indicated that this cultivar ripened 5 days later than the earliest-ripening Ninfa cultivar, but the cultivar was found to be superior with regard to other fruit characteristics (Asma, 2013).

In conclusion, the Mediterranean region of Turkey has an especially great potential for table apricot. Currently, productions are performed with Ninfa, P. de Tyrinthe and Aurora cultivars. Although these cultivars are early ones, quality problems experienced in Ninfa and P. de Tyrinthe and self-incompatibility problem in Aurora limit the productions. The promising cultivar candidates developed in the present breeding program may prevent such problems and may significantly improve apricot culture both in Turkey and in other Mediterranean countries.

## REFERENCES

- Albuquerque, N., Egea, J., Pérez-Tornero, O. and Burgos, L. 2002. Genotyping Apricot Cultivars for Self-(in) Compatibility by Means of *RNases* Associated with *S* Alleles. *Plant Breed.*, **121**: 343-347.
- Asma, B. M. 2013. A New Early-ripening Apricot, ‘Dilbay’. *HortSci.*, **47**:1367
- Badenes, L. M., Martinez-Calvo, J. and Lácer, G. 1998. Analysis of Apricot Germplasm from the European Ecogeographical Group. *Euphytica*, **102**: 93-99
- Bassi, D. 1999. Apricot Culture: Present and Future. *Acta Hort.*, **488**: 35-40.
- Bellini, E., Nencetti, V., Calderoni, F. and Morelli, D. 2008. First Early Ripening Selections of Apricot Obtained at Florence. *Acta Hort.* **488**: 35-40.
- Bircan, M., Pinar, H., Yilmaz, C., Yildiz, A., Paydaş S. and Kaşka, N. 2010. The Apricot Breeding Program among Some Turkish and Foreign Cultivars. *Acta Hort.*, **862**:103-108.
- Burgos, L., Egea, J., Guerriero, R., Viti, R., Monteleone, P. and Audergon, J. M. 1997. The Self-compatibility Trait of the Main Apricot Cultivars and New Selections from Breeding Programs. *J. Hortic. Sci.*, **72**:147-154.
- Burgos, L., Pérez-Tornero, O., Ballester, J. and Olmos, E. 1998. Detection and Inheritance of Stylar Ribonucleases Associated with Incompatibility Alleles in Apricot. *Sex. Plant Reprod.*, **11**:153-158.
- Chenjing, F., Yuanhui, Z. and Xiuying, X. 2005. Genetic Diversity Revealed by ISSR Marker in Apricot. *J. Agric. Univ. Hebei*, **28**:52-55.
- De Nettancourt, D. (1 977). Incompatibility in Angiosperms. In Monographs on Theoretical and Applied Genetics, Vol. 3, R. Frankel, G.A.E. Gall, M. Grossman, and H.F. Linskens, eds (Berlin: Springer-Verlag).
- De Nettancourt, D. 2001. *Incompatibility and Incongruity in Wild and Cultivated Plants*. Springer, Berlin.
- Demirtas, M. N., Atay, S., Gokalp, K., Sahin, M., Olmez, H. A., Yilmaz, K. U., Paydas Kargi, S., Altindag M. and Ercisli, S., 2010. Improvement of Late Blooming Drying Apricot Varieties by Cross Breeding. *Acta Hort.*, **862**: 219-224.
- Doyle, J. J. and Doyle, J. L. 1990. Isolation of Plant DNA from Fresh Tissue. *Focus*, **12**: 13-15
- Egea, J. L., Burgos, P., Martinez-Gomez, F. and Dicenta, F. 1999. Apricot Breeding for Sharka Resistance at CEBAS-CSIC, Murcia-Spain. *Acta Hort.*, **488**: 153-158;
- Egea, J., Dicenta, F., Burgos, L., Martinez-Gomez, P., Rubio, M., Campoy, J. A., Ortega, E., Patino, J. L., Nortes, L., Molina, A. and Ruiz, D. 2010. New Apricot Cultivars from Cebas-Csic (Murcia, Spain)



- Breeding Programme. *Acta Hort.*, **862**:113-118.
16. Ercisli, S. 2009. Apricot Culture in Turkey. *Sci. Res. Essays*, **4**: 715-719.
17. FAO. 2012. [Http://Faostat.Fao.Org/Site/567/Desktopdefault.aspx?Pageid=567#Anchor](http://Faostat.Fao.Org/Site/567/Desktopdefault.aspx?Pageid=567#Anchor)
18. Faust, M. 1989. *Physiology of Temperate Zone Fruit Trees*. John Willey and Sons, New York.
19. Gulcan, R., Misirli, A., Saglam, H., Olmez, H. A., Paydas, S., Derin, K., Eti, S. and Demir, T. 2006. Properties of Turkish Apricot Land Races. *Acta Hort.*, **701**:191-197.
20. Halasz, J., Hegedus, A., Herman, R., Stefanovits-Banyai, E. and Pedryc, A. 2005. New Self-incompatibility Alleles in Apricot (*Prunus armeniaca* L.) Revealed by Styler Ribonuclease Assay and S-PCR Analysis. *Euphytica*, **145**: 57-66.
21. Halasz, J., Pedryc, A. and Hegedus, A. 2007a. Origin and Dissemination of The Pollen-Part Mutated Sc-Haplotype That Confers Self-Compatibility in Apricot (*Prunus armeniaca*). *New Phytol.*, **176**:793-803.
22. Halasz, J., Hegedus, A., Szabo, Z., Nyeki, J. and Pedryc, A. 2007b. DNA-Based S-Genotyping of Japanese Plum and Plum Cultivars to Clarify Incompatibility Relationships. *HortSci.*, **42**:46-50.
23. Halász, J., Pedryc, A., Ercisli, S., Yilmaz, K. U. and Hegedüs, A. 2010. S-Genotyping Supports the Genetic Relationships between Turkish and Hungarian Apricot. *J. Am. Soc. Hortic. Sci.*, **135**: 410-417.
24. Halász, J., Pedryc, A., Ercisli, S., Yilmaz, K. U. and Hegedüs, A. 2012. Apricot Self-incompatibility Shows More Complex Picture than Believed: An Urge for Harmonization. *Acta Hort.*, **966**:193-197.
25. Hegedüs, A., Engel, R., Abrankó, L., Balogh, E., Blázkovics, A., Hermán, R., Halász, J., Ercisli S., Pedryc, A. and Stefanovits-Bányai, E. 2010. Antioxidant and Antiradical Capacities in Apricot (*Prunus armeniaca* L.) Fruits: Variations from Genotypes, Years, and Analytical Methods. *J. Food Sci.*, **75**: 722-730.
26. Hormaza, J. I., Yamane, H. and Rodrigo, J. 2007. Apricot. In: "Genome Mapping and Molecular Breeding in Plants (Fruits and Nuts)", (Ed.): Kole, C. Springer-Verlag, Berlin, PP. 171-187.
27. Jie, Q., Shupeng, G., Jixiang, Z., Manru, G. and Huairui, S. 2005. Identification of Self-incompatibility Genotypes of Apricot (*Prunus armeniaca* L.) by S-Allele-specific PCR Analysis. *Biotechnol. Lett.*, **27**:1205-1209.
28. Julian, C., Herrero, M. and Rodrigo J. 2007. Flower Bud Drop and Pre-blossom Frost Damage in Apricot (*Prunus armeniaca* L.). *J Appl. Bot. Food Qual.*, **81**: 21-25.
29. Kaczmarzka, E., Gawronski, J., Dyduch-Sieminska, M., Najda, A., Marecki, W. and Zebrowska, J. 2015. Genetic Diversity and Chemical Characterization of Selected Polish and Russian Cultivars and Clones of Blue Honeysuckle (*Lonicera caerulea*). *Turk. J. Agric. For.*, **39**: 394-402.
30. Karaca M. and Ince A. G. 2008. Minisatellites as DNA Markers to Classify Bermuda grasses (*Cynodon* Spp.): Confirmation of Minisatellite in Amplified Products. *J. Genet.*, **87**: 83-86.
31. Lachkar, A., Fattouch, S., Ghazouani, T., Halász, J., Pedryc, A., Hegedüs, A. Mars, M. 2013. Identification of Self-(in) Compatibility S-Alleles and New Cross-incompatibility Groups in Tunisian Apricot (*Prunus armeniaca* L.) Cultivars. *J. Hortic. Sci. Biotech.*, **88**: 497-501.
32. Layne, R.E.C., Bailey, C.H. and Hough, L.F., 1996. Apricots. 1. Tree and Tropical Fruits. In: "Fruit Breeding", (Eds.): Janick, J. and Moore, J. M. John Willey And Sons, New York, PP. 79-111,
33. Li, G. and Quiros, C. F. 2001. Sequence-Related Amplified Polymorphism (SRAP), a New Marker System Based on a Simple PCR Reaction: Its Application to Mapping and Gene Tagging in Brassica. *Theor. Appl. Genet.*, **103**:455-461
34. Lo Bianco, R., Farina, V., Indelicato, S. G., Filizzola, F. and Agozzino, P. 2010. Fruit Physical, Chemical and Aromatic Attributes Of Early, Intermediate and Late Apricot Cultivars. *J. Sci. Food Agric.*, **90**: 1008-1019.
35. Mantel, N. 1967. The Detection of Disease Clustering and a Generalized Regression Approach. *Cancer Res.*, **27**:209-220.
36. McClure, B. A., Haring, V., Ebert, P. R., Anderson, M. A., Simpson, R. J., Sakiyama, F. and Clarke, A. E. 1989. Style Self-incompatibility Gene Products of *Nicotiana glauca* Are Ribonucleases. *Nature*, **342**: 955-957.

37. Milatović, D., Nikolić, D., Rakonjac, V. and Fotirić-Akšić, M. 2010. Cross-(in) Compatibility in apricot (*Prunus armeniaca* L.). *J. Hortic. Sci. Biotech.*, **85**: 394-398.
38. Mishra, P. K., Ram, R. B. and Kumar, N. 2015. Genetic Variability, Heritability, and Genetic Advance in Strawberry (*Fragaria×ananassa* Duch.). *Turk. J. Agric. For.*, **39**: 451-458.
39. Nemli, S., Kianoosh, T. and Tanyolac, M. B. 2015. Genetic Diversity and Population Structure of Common Bean (*Phaseolus vulgaris* L.) Accessions through Retrotransposon-based Interprimer Binding Sites (iPBSs) Markers. *Turk. J. Agric. For.*, **39**: 940-948.
40. Pennone, F., Abbate, V., Carbone, A. and Scarpato, L. 2010. Apricot Breeding in Caserta: Result and Perspectives. *Acta Hort.*, **862**: 67-72.
41. Pinar, H., Bircan, M., Yilmaz, C., Kargi, S. P., Kaska, N. and Yildiz, A. 2008. The Performance of Some Apricot Cultivars in the Mersin Ecological Conditions. *14<sup>th</sup> International Symposium on Apricot Breeding and Culture*, 16-20 Haziran 2008, Matera, Italy, 862: 109-112
42. Polat, A. and Caliskan, O. 2013. Yield and Fruit Characteristics of Various Apricot Cultivars under Subtropical Climate Conditions of the Mediterranean Region in Turkey. *Int. J. Agron.*, Article ID 687345.
43. Rohlf, F. J. 2000. NTSYS-PC: Numerical Taxonomy and Multivariate Analysis System, Version 2.2. Exeter Software, Setauket, New York.
44. Romero, C., Vilanova, S., Burgos, L., Martínez-Calvo, J., Vicente, M., Llácer, G. and Badenes, M. L. 2004. Analysis of the S-Locus Structure in *Prunus Armeniaca* L. Identification of S-Haplotype Specific S-RNase and F-Box Genes. *Plant Mol. Biol.*, **56**: 145-157.
45. Ruiz, D. and Egea, J. 2008. Phenotypic Diversity and Relationships of Fruit Quality Traits in Apricot (*Prunus armeniaca* L.) Germplasm. *Euphytica*, **163**: 143-158.
46. Ruiz, D., Dicenta, F., Burgos, L., Martinez-Gomez, P., Rubio, M., Campoy, J. A., Ortega, E., Patino, J. L., Molina, A. and Egea, J. 2010. New Apricot Cultivars from CEBAS\_CSIC (Murcia, Spain) Breeding Programme. *Acta Hort.*, **862**: 113-118.
47. Sonneveld, T., Robbins, T. P., Boskovic, R. and Tobutt, K. R. 2001. Cloning of Six Cherry Self-incompatibility Alleles and Development of Allele-specific PCR Detection. *Theor. Appl. Genet.*, **102**: 1046-1055.
48. Szabó, Z. and Nyéki, J. 1991. Blossoming, Fructification and Combination of Apricot Varieties. *Acta Hort.*, **293**: 295-302.
49. Topor, E., Vasilescu, R., Balan, V. and Tudor, V. 2010. Apricot Breeding Programme for Late and Very Late Ripening Period in Romania. *Acta Hort.*, **862**: 137-142.
50. Uzun, A., Gulsen, O., Seday, U., Bircan, M. and Yilmaz, K. U. 2010. SRAP Based Genetic Analysis of Some Apricot Cultivars. *Rom. Biotech. Lett.*, **15**: 5396-5404.
51. Uzun, A., Celik, B., Karadeniz, T., Yilmaz, K. U. and Altintas, C. 2015. Assessment of Fruit Characteristics and Genetic Variation among Naturally Growing wild Fruit *Elaeagnus angustifolia* Accessions. *Turk. J. Agric. For.*, **39**: 286-294.
52. Vilanova, S., Romero, C., Burgos, L., Llácer, G. and Badenes, M. L. 2005. Identification of Self-(in) Compatibility Alleles in Apricot (*Prunus armeniaca* L.) By PCR and Sequence Analysis. *J. Am. Soc. Hortic. Sci.*, **130**: 893-898.
53. Wojnicka-Poltorak, A., Celinski, K., Chudzinska, E., Prus-Glowacki, W. and Niemtur, S. 2015. Genetic resources of *Pinus cembra* L. marginal populations from the Tatra Mountains: Implications for Conservation. *Biochem. Genet.*, **53**: 49-61.
54. Wu, J., Gu, C., Zhang, S. L., Zhang, S. J., Wu, H. Q. and Heng, W. 2009. Identification of S-Haplotype-specific S-RNase and SFB Alleles in Native Chinese Apricot (*Prunus armeniaca* L.). *J. Hortic. Sci. Biotech.*, **160**: 241-248.
55. Yilmaz, K. U. and Gurcan, K. 2012. "Genetic Diversity in Apricot". In: "Genetic Diversity in Plants", (Ed.): Caliskan, M. Intech , Rijeka, PP. 249-270.
56. Yilmaz, K. U., Paydas-Kargi, S., Doğan, Y. and Kafkas, S. 2012. Genetic Diversity Analysis Based on ISSR, RAPD and SSR among Turkish Apricot Germplasms in Iran Caucasian Eco-Geographical Group. *Sci. Hortic.*, **138**: 138-143.
57. Zhang, L., Chen, X., Chen, X. L., Zhang, C., Liu, X., Ci, Z. J., Zhang, H., Wu, C. and Liu, C. 2008. Identification of Self-incompatibility (S-) Genotypes of Chinese Apricot Cultivars. *Euphytica*, **160**: 241-248.



## ویژگی های مورفولوژیکی، مولکولی، و خود (نا) سازگاری ژنوتیپ های جدید و امید بخش زرد آلو

ه. پینار، س. ارسیسلی، م. بیرکن، م. اولنو، ا. ازون، ک. و. ایلماز، و م. یامان

### چکیده

تولید زرد آلو در جهان به طور پیوسته ای رو به افزایش است و علت آن اصلاح نژاد کولتیوارها ی جدید پر محصول است. اخیراً، برنامه های اصلاح نژاد برای مطابقت با خواسته های مصرف کنندگان و بهبود مقاومت به امراض (مانند کولتیوارهای Sharka و Monilinia)، صدمات یخزدگی، و تعیین خود (نا) سازگاری تغییر کرده است. در این پژوهش، ۱۴ فرزند (نتاج) از زرد آلو و ۶ عدد از والدین آن ها با استفاده از نشانگر های مورفولوژیکی و مولکولی ارزیابی شدند. به عنوان نشانگر های مورفولوژیکی از وزن میوه، طول و عرض و ارتفاع آن، کل جامدات محلول، اسیدیته، و سفتی میوه استفاده شد. در تحلیل مولکولی، به منظور تعیین روابط ژنتیکی، از نشانگر های پلی مورفسم (چند شکلی) تکثیر شده مبتنی بر توالی (SRAP)، تکرار توالی بین ساده (ISSR)، و الگوهای مولکولی همراه خسارت (DAMP) استفاده شد. افزون بر این، برای تعیین مشخصات آلل S، نشانگر های SRc-F/R به کار گرفته شد. نتایج نشان داد که هر چند هیچ ژنوتیپی زودرس تر از Ninfa و Priana وجود نداشت، حاصل تلاقی Ay x P3 ژنوتیپ امید بخشی در مورد زودرسی و ویژگی های میوه بود. در کل، ۲۲۴ باند قابل امتیازدهی به دست آمد با ۸ ترکیب آغازگر SRAP (۲۵ باند)، ۸ آغازگر DAMP (۸۱ باند) و ۱۸ باند ISSR (۱۱۸ باند) که نشان دهنده تنوع زیاد بین دورگه ها و کولتیوارها بود. همچنین، در این پژوهش، کلا ۴ آلل S- RNase (SC, S2, S3, S6) شناسایی شد که در میان آن ها آلل های S<sub>C</sub> و S<sub>3</sub> به طور گسترده ای حضور داشتند.