

*Full Length Research Paper*

# Determination and evaluation of superior self-compatible almond genotypes resulting from crosses between “Touno” and “Ferragnes” in Iran

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Self-incompatibility is one of the most considerable difficulties in almond production, which reducing fruit set dramatically and making orchard management difficult. Therefore, breeding almond to produce self-compatible genotypes is very important. The purpose of this investigation was determination and evaluation of self-compatible and late flowering cultivars with good kernel characteristics in 38 almond genotypes resulting from crosses between self-compatible cultivar, "Touno" (male parent) and self-incompatible cultivar, "Fragnees" (female parent). Determination of self-compatible genotypes was carried out by investigation of fruit set levels, microscopic study of pollen tube after self pollination and PCR method. In addition, flowering and fruit characteristics were studied according to Gulcan descriptor. Genotypes number Tf23 and Tf6 were identified as very late flowering and self-compatible genotypes, in which their flowering time is coincidental with the very late flowering commercial cultivar, "Tardi nonpariel". However, fruits of both genotypes were rather small. The highest fruit dry weight (4.24 and 4.06 g) was for self-incompatible genotype Tf28 and self-compatible genotype Tf15, respectively. These two genotypes had also the highest kernel weight being 1.16 and 1.13 g, respectively. Genotypes number Tf24 and Tf31 also had high dry weight fruit and kernel, which Tf24 had been identified as self-compatible, and Tf31 as self-incompatible genotypes. In genotypes Tf15, Tf31 and Tf24 fruit, nut and kernel had good qualities. The colour of nut and kernel of these genotypes were bright. Their kernels were uniform, smooth and sweet with low shrivelling. All fruits were full, with single kernel, which were in the same line as market demands. Investigations on fruit set level in field conditions revealed genotype number Tf23, with 18.23% on fruit set, as the most self-compatible genotype.

**Key words:** Self-compatibility, self-incompatibility, fruit set, microscopic study.

## INTRODUCTION

Almond is one of the temperate zone fruit trees being cultivated in different countries for its nutritional value. Late blooming has been one of the most important

objectives of almond breeding programs (Kester, 1965; Grasselly, 1972; Vargas et al., 1984; Socias i Company and Filipe, 1999), and its transmission has been studied (Kester et al., 1977a; Dicenta and Garcia, 1993a). Blooming density and productivity are also two important traits studied by Grasselly (1972), Kester and Asay (1975), Grasselly and Crossa-Raynaud (1980), Vargas et al. (1984) and Dicenta and Garcia (1993a). Some studies

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were also performed regarding the time of maturity of fruits (Grasselly, 1972; Kester and Asay, 1975; Dicenta and Garcia, 1993a). Furthermore, there are studies on transmission of some fruits and kernel traits (Grasselly, 1972; Spiegel-Roy and Kochba, 1974; Kester et al., 1977b; Vargas et al., 1984; Dicenta and Garcia, 1993b). Moradi (2006) studied several quantitative traits in some almond genotypes. Kodak and Socias i company (2006) in a breeding program performed crossing between the two varieties of "Felisia" and "Bertina". They introduced G-1-1, G-1-41, G-2-25, G-6-14 genotypes as self-compatible and very late flowering genotypes.

To evaluate almond genotypes, some research work were carried out in different parts of Iran, including Tehran province (Vezvaei, 1985), Khorasan province (Ghassir and Abadi, 1995) and Mianeh (Imani, 1997). It was reported that from 30 almond cultivars grown in different areas of Iran, 24 cultivars were introduced by choosing superior genotypes existing in endemic germplasm (Vezvaei, 2003). Almond encounters the problem of self-incompatibility classified as a gametophytic (Socias I Company et al., 1976), resulting in the arrest of pollen tube growth in the mid-third of the style (De Nettancourt, 1977). This problem appeared to be controlled by the multi-allelic *S* locus (Gagnard, 1954), and self-compatible allele (*Sf*) being dominant (Dicenta and García, 1993a).

Until now, 30 self-incompatibility (SI) RNase alleles in North American and Mediterranean accessions and one allele, *Sf* that allows self compatibility have been identified (Lo'pez et al., 2006; Ortega et al., 2006; Bos'kovic' et al., 2007). Application of DNA based molecular markers provides the opportunity for early selection of the common *S*-alleles (Mart'inez-Go'mez et al., 2007). PCR-based strategies have been developed for the identification of *S*-RNase alleles using genomic DNA (Ushijima et al., 1998; Tamura et al., 2000), and primers, which were designed from conserved regions (Channuntapipat et al., 2001; Tamura et al., 2000), resulting in the confirmation of the identity of many *S*-alleles and the identification of new ones (Channuntapipat et al., 2003; Ortega et al., 2006; Kodak et al., 2008a).

The purpose of this investigation was identification of late flowering self-compatible progenies with good kernel quality resulting from crossing between "Touno" as male parent and "Ferragnes" as female parent.

## MATERIALS AND METHODS

### Plant material

In this research work, 38 almond genotypes (six years old) obtained from crossing between "Touno" (male parent) and "Ferragnes" (female parent) were evaluated in Karaj, Iran. In year 2010, cold temperature occurred and lasted for three days (20 to 22 March 2010) in which temperature reached below zero (-3°C, -2°C, -1°C). Therefore, some genotypes had no fruit set at all and qualitative

and quantitative traits of fruits were studied only in 23 genotypes, which could produce fruit.

### Evaluation of agronomic traits

During year 2010, the following tree traits were studied according to the protocols given by Gulcan (1985): Date of initial blooming (when 10% of flowers were opened), Date of full bloom (when 90% of flowers were opened), Pollen Pistil compatibility and its level, Coloration of shoot tip, Tree Habit of growth, Status of stigma compared to anther, Status of style junction to ovary, Location of flower buds, Blooming density, Double flower percentage in buds, Double budding percentage in branches and Ripening date (when 95% of fruits had their mesocarp opened).

At maturity stage, a sample of 20 fruits was taken from each genotype and then the fruit and kernel traits were studied: Ease of hulling, Shape of fruit, nut and kernel, Shell hardness, Fruit, Nut and Kernel weight (g), In-shell/kernel ratio (%), Empty nuts, Double kernels (%), Sound Kernels (%), Fruit, Nut and Kernel thickness (mm), Fruit, Nut and kernel length (mm), Fruit, Nut and kernel width (mm), Shell and kernel colour intensity, Suture opening of the shell, Kernel Pubescence, and Kernel bitterness.

### Fruit set determination

Fruit set percentages were evaluated according to Filipe (1977) and Imani (2005). In addition, one branch was left for open pollination to be compared with self-pollination.

### Pollen tube growth

Flower samples were harvested at 24, 72 and 120 h after hand pollination. The pistils were prepared according to Socias i Company (1976). Pollen tube growth was assessed by observation in a Leitz Ortholux II microscope with UV illumination of a mercury lamp Osram HBO 200 W/4, (Linskens and Esser, 1957).

The percentages of pistils, having self-pollen tube at the style base, were recorded in each cultivar during 2010. When more than 50% of pistils showed self-pollen tubes at the style base, the plant was considered to be self-compatible. When more than 75% of pistils had self pollen tube at the style base, the plant was categorized in the highly self-compatible group, and finally, when percentage of pistils with self-pollen tubes at the style base was less than 25%, the plant was considered to be self-incompatible. On the other hand, genotypes in which 25 to 50% of pistils showed self-pollen tubes at the style base were classified as doubtful phenotypes (Alonso and Socias i Company, 2005).

### PCR method

Genomic DNA was extracted from leaves following the CTAB extraction method based on Doyle and Doyle (1987). PCR reaction was carried out using the consensus primers *SfF* (forward) and *SfR* (reverse) (Channuntapipat et al., 2003) to confirm the presence of the *Sf* allele. PCR reaction contained 10 µl DNA of Stock concentration of 10 ng/L, 7.5 µl PCR kit (Sigma) of Stock concentration 2X, 0.625 µl of primers of stock concentration of 10 µM/L and 6.25 µl of water for ultimate reaction volume of 25 µl.

PCR was programmed for 3 min at 95°C, 34 cycles of 30 s at 95°C, 45 s at 53°C, and 1 min at 72°C, followed by 10 min at 72°C. The PCR products were separated on 1.5% (w/v) agarose gel containing 0.1% (v/v) ethidium bromide in a 0.25 1X TBE at 90 V for 1.5 h. DNA were visualized under UV light and the images were captured by a Kodak camera.

### Statistical analysis

The experiment was conducted in a Completely Randomized Design (CRD). All statistical analyses were performed using the SAS program (SAS Institute, 2000). The mean separation was conducted through the Duncan multiple range test.

## RESULTS

### Evaluation of flowering traits

Among 23 genotypes, Tf34 and Tf6 were the first (7/3/2010) and last (18/3/2010) genotypes, which started to bloom (Table 1). In this area, some almond cultivars had started blooming since 19/2/2010 (Cv. 'White'). However, cultivar "Tardi non pariel" was the last one to initiate blooming in 18/3/2010. Considering flowering date of started to bloom cultivar "Tardi non pareil" (control), Tf 10 and Tf 13 genotypes, which started blooming two days earlier and Tf 23 and Tf 6 genotypes, which began blooming one day sooner and synchronous with cultivar Tardy nonpareil (The control), were identified as very late flowering genotypes. Regarding stigma and anther positions, stigma was lower than anthers in genotypes Tf1, Tf22, Tf24, Tf28 and Tf37. Therefore, their pollination can be carried out better. According to the results (Table 2), percent of double flower in buds was between 0 to 75% (Tf32).

Genotype Tf24 had the highest double budding percentage on each branch with 60.1%. With the increase in double budding in almond tree branches, the number of flowers increases, which resulted in higher yields. The highest blooming density on each branch was for genotype Tf21 with 134 flowers per 20 cm length of a one-year-old branch. However, the lowest blooming density was observed in genotypes Tf12 and Tf13 with 50 and 53 flowers per 20 cm length of a one-year-old branch, respectively. High flower density is a good advantage that resulted in higher yields (Mohan Jain and Priyadarshan, 2009).

### Fruit set

Percentages of fruit set at 60 days after self-pollination are shown in Table 3. In the second year (2010), cold temperature occurred and lasted for three days (20-22 March 2010) reaching below zero (-3, -2 and -1°C). Therefore, some genotypes had no fruit set at all (Table 3). In these conditions, genotype Tf23 was identified as self-compatible with fruit set of 18.23%. Fruit set comparison between open pollination and self pollination in self-compatible genotypes showed that open pollination increases the fruit set of self-compatible genotypes. As observed in some species, fruit drop is sometimes associated with embryo abortion (Crane and Iwakiri, 1980; Furukawa and Bukovac, 1989), which has

been attributed to deficiencies in endosperm development (Bradley and Crane, 1975).

In the self-compatible almond cultivar 'Tuono', Oukabli et al. (2000) observed the degeneration of endosperm, which subsequently led to embryo abortion due to self fertilization. These results were explained as a consequence of inbreeding effects at post-zygotic stage. However, in prezygotic stage, differences were not observed for pollen tube growth along the pistil following self-pollination and cross pollination.

### Pollen tube growth

The microscopic observation of self-pollen tube growth showed high variability among progenies and allowed phenotypic classification of genotypes. Among 38 genotypes (according to Alonso and Socias i Company, 2005), 39.47% of genotypes were recognized as self-incompatible, 23.68% as doubtful, 26.32% as self-compatible and 10.53% as highly self compatible at 120 h after pollination (Table 4). Sampling times of 24 and 72 h after flower opening were recognized as inadequate periods for discriminating genotypes in order to select self compatible progenies (Table 4). As noted, the incompatibility system is controlled by the multi-allelic locus S (Gagnard, 1954), and self-compatible allele ( $S_i$ ) being the dominant (Dicenta and García, 1993b). Results of microscopic observation were similar to fruit set study in the field. Numbers of self-compatible genotypes were less in the investigation of fruit set level in orchard compared to that investigation under florescence microscope, which it may due to the occurrence of late-spring cold.

### PCR

Results of PCR reaction is shown in Figure 1. In this study, 19 genotypes, (Tf3, Tf5, Tf6, Tf7, Tf8, Tf10, Tf13, Tf15, Tf16, Tf17, Tf23, Tf24, Tf27, Tf29, Tf30, Tf32, Tf 33, Tf 34 and Tf 38) band with length of 450 bp, which were identified as self-compatible genotypes. Results indicated that "Touno" (male parent) and "Ferragnes" (female parent) had no common alleles, which resulted in ratio of 1:1 between self-compatible and self-incompatible genotypes being in the same line with others (Channuntapipat et al., 2003). PCR method could also discriminate the status of nine genotypes, which were realized as doubtful genotypes in microscopic study, in such a way that five genotypes were recognized as self-compatible and another four identified as self incompatible. It can be concluded that all employed methods in this study proved to be efficient in discriminating self-compatible genotypes from self-incompatible ones. However, combination of all three methods is more accurate in determining the level of self-

**Table 1.** Evaluation of qualitative vegetative and generative traits in 38 almond genotypes as well as cultivar Tardi non pareil as a control.

Genotypes	Incompatible or Compatible	Date of initial bloom ( when 10% of flowers were opened)	Date of full bloom (when 90% of flowers were opened)	Tree Habit of branches	Location of flower buds	Anthocyanin coloration on one-year old shoots	Status of style junction to ovary	Position of stigma compared to anther
Tf 1	Incompatible	13/3.2010	2010/15/3	Spreading	On one-year old shoots and spurs	No anthocyanin coloration	Style is one branches and limber	Below
Tf 2	Incompatible	15/3/2010	2010/17/3	Spreading	On one-year old shoots and spurs	Strong	Style is one branches and limber	Above
Tf 3	Compatible	15/3/2010	2010/16/3	Spreading	On one-year old shoots and spurs	Strong	Style is one branches and standing	Above
Tf 4	Incompatible	14/3/2010	2010/16/3	Upright	On one-year old shoots	Intermediate	Style is one branches and standing	Surface
Tf 5	Compatible	15/3/2010	2010/17/3	Extremely upright	On one-year old shoots and spurs	Strong	Style is one branches and standing	Surface
Tf 6	Compatible	18/3/2010	2010/21/3	Spreading	On one-year old shoots and spurs	Intermediate	Style is one branches and standing	Above
Tf 7	Compatible	15/3/2010	2010/16/3	Upright	On one-year old shoots and spurs	No anthocyanin coloration	Style is one branches and limber	Above
Tf 8	Compatible	14/3/2010	2010/16/3	Spreading	On one-year old shoots and spurs	Intermediate	Style is one branches and standing and 5% of Style and stigma are multi branches and joined together in junction	Above
Tf 9	Incompatible	15/3/2010	2010/17/3	Spreading	On one-year old shoots	Intermediate	Style is one branches and standing	Below

Table 1. Contd.

Tf 10	Compatible	16/3/2010	2010/18/3	Upright	On one-year old shoots and spurs	Intermediate	Style is one branches and standing	Above
Tf 11	Incompatible	15/3/2010	2010/18/3	Spreading	On one-year old shoots and spurs	Low	Style is one branches and standing and 5% of Style and stigma are multi branches and joined together in junction	Above
Tf 12	Incompatible	15/3/2010	2010/16/3	Upright	On one-year old shoots	Strong	Style is one branches and standing	Surface
Tf 13	Compatible	16/3/2010	2010/18/3	Dropping	On one-year old shoots and spurs	Intermediate	Style is one branches and standing	Surface
Tf 14	Incompatible	13/3/2010	2010/15/3	Spreading	On one-year old shoots and spurs	Strong	Style is one branches and limber	Surface
Tf 15	Compatible	13/3/2010	2010/15/3	Weeping	On one-year old shoots and spurs	Low	Style is one branches and limber	Above
Tf 16	Compatible	14/3/2010	2010/16/3	Upright	On one-year old shoots and spurs	Intermediate	Style is one branches and limber	Surface
Tf 17	Compatible	15/3/2010	2010/16/3	Spreading	On one-year old shoots and spurs	Low	Style is one branches and standing	Above
Tf 18	Incompatible	15/3/2010	2010/16/3	Upright	On one-year old shoots and spurs	Low	Style is one branches and standing	Surface
Tf 19	Incompatible	13/3/2010	2010/15/3	Upright	On one-year old shoots and spurs	Intermediate	Style and stigma are multi branches and joined together in junction	Surface

Table 1. Contd.

Tf 20	Incompatible	14/3/2010	2010/16/3	Spreading	On one-year old shoots and spurs	Low	Style is one branches and standing	Above
Tf 21	Incompatible	14/3/2010	2010/16/3	Dropping	On one-year old shoots and spurs	Low	Style is one branches and standing	Above
Tf 22	Incompatible	13/3/2010	2010/15/3	Spreading	On one-year old shoots and spurs	Intermediate	Style is one branches and standing	Below
Tf 23	Compatible	17/3/2010	2010/20/3	Dropping	On one-year old shoots and spurs	Low	Style is one branches and limber	Surface
Tf 24	Compatible	15/3/2010	2010/17/3	Upright	On one-year old shoots and spurs	Strong	Style is one branches and standing	Below
Tf 25	Incompatible	13/3/2010	2010/15/3	Spreading	On one-year old shoots and spurs	Strong	Style is one branches and limber	Above
Tf 26	Incompatible	13/3/2010	2010/15/3	Dropping	On one-year old shoots and spurs	Low	Style is one branches and limber	Above
Tf 27	Compatible	14/3/2010	2010/16/3	Upright	On one-year old shoots and spurs	Strong	Style is one branches and limber	Surface
Tf 28	Incompatible	14/3/2010	2010/16/3	Spreading	On spurs	Low	Style is one branches and standing	Below
Tf 29	Compatible	15/3/2010	2010/17/3	Spreading	On one-year old shoots and spurs	Strong	Style is one branches and standing	Above

Table 1. Contd.

Tf 30	Compatible	15/3/2010	2010/17/3	Extremely upright	On one-year old shoots and spurs	Low	Style is one branches and standing	Above
Tf 31	Incompatible	10/3/2010	2010/12/3	Spreading	On one-year old shoots and spurs	Strong	Style is one branches and standing	Surface
Tf 32	Compatible	14/3/2010	2010/17/3	Upright	On one-year old shoots and spurs	Strong	Style is one branches and standing	Above
Tf 33	Compatible	12/3/2010	2010/15/3	Spreading	On one-year old shoots and spurs	Low	Style is one branches and standing	Surface
Tf 34	Compatible	7/3/2010	2010/11/3	Spreading	On one-year old shoots and spurs	Strong	Style is one branches and standing	Surface
Tf 35	Incompatible	15/3/2010	2010/17/3	Spreading	On one-year old shoots and spurs	Strong	Style is one branches and standing	Above
Tf 36	Incompatible	16/3/2010	2010/17/3	Upright	On one-year old shoots and spurs	Low	Style is one branches and standing	Surface
Tf 37	Incompatible	16/3/2010	2010/17/3	Upright	On one-year old shoots and spurs	Intermediate	Style is one branches and standing	Below
Tf 38	Compatible	15/3/2010	2010/16/3	Upright	On one-year old shoots	Low	Style is one branches and limber	Surface
Tardy nonpareil	Incompatible	18/3/2010	2010/22/3	Upright	On one-year old shoots and spurs	Low	Style is one branches and standing	Surface

**Table 2.** Evaluation of quantitative generative traits in some almond genotypes as well as cultivar Tardy non pareil as a control.

Genotypes	Double budding percentage in branches	Double flower percentage in buds	Blooming density	Genotypes	Double budding percentage in branches	Double flower percentage in buds	Blooming density	Genotypes	Double budding percentage in branches	Double flower percentage in buds	Blooming density
Tf 1	0.5 rs	0 j	60 k	Tf 14	8 on	0 j	95 e	Tf 27	14.78 kj	0 j	57 kl
Tf 2	35 e	20 e	65 j	Tf 15	12.3 ml	55 a	65 j	Tf 28	23.55 h	0 j	95 e
Tf 3	18.5 i	0 j	85 f	Tf 16	50.5 b	15 f	102 d	Tf 29	31.25 f	0 j	80 g
Tf 4	2.2 r	0 j	55 l	Tf 17	6.67 oqp	2 ij	75 hi	Tf 30	30.66 f	0 j	75 hi
Tf 5	8 on	0 j	75 hi	Tf 18	37.5 d	0 j	80 g	Tf 31	0 s	3 i	80 g
Tf 6	8.75 n	0 j	80 g	Tf 19	28.58 g	40 b	65 j	Tf 32	50.2 b	75 a	73 i
Tf 7	5.72 pq	0 j	75 hi	Tf 20	0 s	0 j	115 c	Tf 33	5.5 q	2 ij	111 c
Tf 8	16.25 j	12 g	80 g	Tf 21	15.85 j	0 j	134 a	Tf 34	2.25 r	0 j	66 j
Tf 9	23.35 h	0 j	60 k	Tf 22	16.24 j	14 f	125 b	Tf 35	33.75 e	0 j	80 g
Tf 10	11.76 ml	0 j	85 f	Tf 23	7.5 onp	0 j	75 hi	Tf 36	15 j	0 j	55 l
Tf 11	41.54 c	5 h	65 j	Tf 24	60.1 a	0 j	85 f	Tf 37	11 m	0 j	65 j
Tf 12	2 r	0 j	50 m	Tf 25	8 on	0 j	78 hg	Tf 38	20.1 i	35 d	68 j
Tf 13	50 b	0 j	53 ml	Tf 26	13.02 kl	0 j	77 hgi	Tardy nonpareil	0 s	0 j	53 ml

Means, in each column and for each factor, followed by similar letter(s) are not significantly different at the 5% probability level- using Duncan's multiple range test.

**Table 3.** Percentage of fruit set of 38 genotypes obtained from breeding program after self and open pollination in 2010.

Genotypes	Fruit set in 60 day after self pollination (%)	Fruit set in 60 day after open pollination (%)	Genotypes	Fruit set in 60 day after self pollination (%)	Fruit set in 60 day after open pollination (%)	Genotypes	Fruit set in 60 day after self pollination (%)	Fruit set in 60 day after open pollination (%)
Tf 1	0	0	Tf 14	0	1.27	Tf 27	2.5	0
Tf 2	0.7	4	Tf 15	1.11	4.07	Tf 28	1.34	0.48
Tf 3	4.54	11.30	Tf 16	7.67	40.16	Tf 29	7.73	6.67
Tf 4	2.34	2.5	Tf 17	2.72	15.42	Tf 30	0	0.47
Tf 5	0	1.94	Tf 18	0	3.24	Tf 31	0	0.30
Tf 6	7.73	13.13	Tf 19	0	7.17	Tf 32	10	31.82
Tf 7	5.66	24.12	Tf 20	0	12.86	Tf 33	0	0
Tf 8	0.27	4	Tf 21	0	2.63	Tf 34	0	0
Tf 9	0	5	Tf 22	0	0	Tf 35	4.16	18.82
Tf 10	10.32	23.89	Tf 23	18.23	27.5	Tf 36	-	-

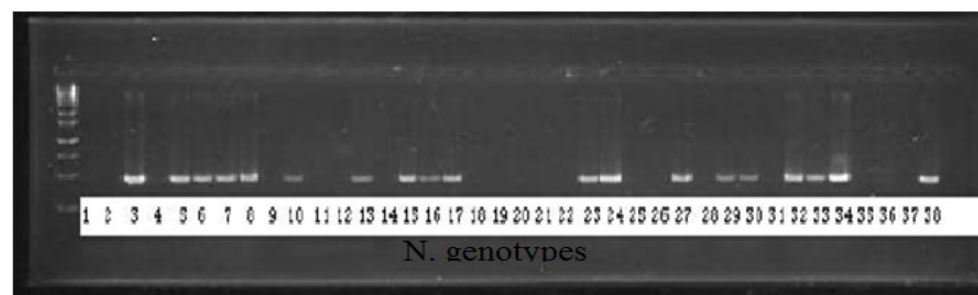


**Table 3.** Contd.

Tf 11	2.75	14.17	Tf 24	0	6.29	Tf 37	-	-
Tf 12	0	7.85	Tf 25	0	0	Tf 38	6.45	7.28
Tf 13	7.14	26	Tf 26	0.80	3.18	-	-	-

**Table 4.** Percent progeny with pollen tube at the end of their style at 24, 72 and, 120 h after self pollination in 2010.

Cross combination	No. of total genotypes	Time (Hour after self-pollination)	NO. of self-incompatible genotypes	Percent of self-incompatible genotypes	NO. of doubtful genotypes	Percent of doubtful genotypes	NO. of self-compatible genotypes	Percent of self-compatible genotypes	NO. of highly self-compatible genotypes	Percent of highly self-compatible genotypes
Touno * Ferragnes	38	24	38	100	0	0	0	0	0	0
Touno * Ferragnes	38	72	35	92.11	1	2.63	1	2.63	1	2.63
Touno * Ferragnes	38	120	15	39.47	9	23.68	10	26.32	4	10.53

**Figure 1.** Situation of progenies from the point of pollen-pistil compatibility studied by primers of SfF and SfR.

compatibility.

### Fruit quantitative and qualitative characteristics

Among 23 genotypes capable of producing fruits, genotypes Tf19, Tf28, and Tf29 were early ripening, (2/8/2010). However, genotypes Tf3 and tf24 had their fruit ripen as late as (12/8/2010). In

this area, fruits of Cv. "Shokofe" were the first ones to get ripen (30/7/2010), whereas Cv. "Shahrood 17" was the last to have its fruits ripen as late as 28/8/2010.

Fruits of Cv. "Touno" were ripened at 14/8/2010. Therefore, comparing with "Touno", all genotypes of this population were considered as early or mid ripening.

Period for ripening (time between flowers

pollination to fruit harvesting) was between 137 days in Tf6 and 150 days in Tf3. However, it was about 139 days in "Tardi nonpareil" (Table 5). From the point of shell hardness, genotypes were classified in different groups (Table 5). In this population, there were no paper shell genotypes, due to the easy shell breaking of paper shell considered as an advantage. Genotype Tf6 and Tf19 had soft shell.

**Table 5.** Evaluation of qualitative traits of fruit, nut and kernel in the 23 almond genotypes as well as cultivar Tardi non pareil as a control.

Genotypes	Ripening date	Period for ripening	Fruit shape	Ease of hulling	Suture opening of the shell	Shell color intensity
Tf 2	8/8/2010	145	Ovate	High	Open (about 2 mm)	light brown
Tf 3	12/8/2010	150	Sharp oval	High	Open (about 2 mm)	light brown
Tf 4	4/8/2010	140	Sharp oval	High	Open (about 2 mm)	light brown
Tf 5	6/8/2010	142	Ovate	High	Open (about 2 mm)	light brown
Tf 6	6/8/2010	137	Sharp oval	High	Open (about 2 mm)	light brown
Tf 7	6/8/2010	144	Ovate	High	Excellent seal (no openings)	light brown
Tf 8	6/8/2010	144	Ovate	High	Excellent seal (no openings)	light brown
Tf 11	3/8/2010	139	Ovate	High	Open (about 2 mm)	intermediate
Tf 15	6/8/2010	147	Ovate	High	Excellent seal (no openings)	light brown
Tf 16	3/8/2010	141	Sharp oval	High	Open (about 2 mm)	intermediate
Tf 17	4/8/2010	142	Ovate	High	Open (about 2 mm)	intermediate
Tf 18	7/8/2010	145	Ovate	High	Open (about 2 mm)	light brown
Tf 19	2/8/2010	141	Ovate	High	Excellent seal (no openings)	light brown
Tf 20	11/8/2010	149	Sharp oval	High	Open (about 2 mm)	light brown
Tf 23	10/8/2010	144	Ovate	High	Open (about 2 mm)	light brown
Tf 24	12/8/2010	149	Ovate	High	Open (about 2 mm)	light brown
Tf 26	9/8/2010	148	Ovate	High	Open (about 2 mm)	light brown
Tf 27	5/8/2010	143	Ovate	High	Excellent seal (no openings)	intermediate
Tf 28	2/8/2010	141	Ovate	High	Open (about 2 mm)	intermediate
Tf 29	2/8/2010	140	Sharp oval	High	Excellent seal (no openings)	intermediate
Tf 31	6/8/2010	148	Ovate	High	Excellent seal (no openings)	light brown
Tf 32	5/8/2010	142	Ovate	High	Open (about 2 mm)	light brown
Tf 35	6/8/2010	143	Ovate	High	Excellent seal (no openings)	light brown
Tardy nonpareil	6/8/2010	139	Ovate	High	Excellent seal (no openings)	light brown
<b>Shell hardness</b>	<b>Nut Shape</b>	<b>Kernel shape</b>	<b>Kernel shriveling</b>	<b>kernel color intensity</b>	<b>Kernel Pubescence</b>	<b>Kernel bitterness</b>
Hard	Ovate	Ovate	wrinkle	light brown	High	slightly bitter
intermediate	Sharp oval	Ovate	Slightly wrinkle	light brown	Low	sweet
intermediate	Sharp oval	Ovate	Slightly wrinkle	light brown	Low	sweet
Hard	Ovate	Ovate	Slightly wrinkle	light brown	intermediate	sweet
Soft	Sharp oval	Ovate	Slightly wrinkle	light brown	Low	sweet
very hard	Sharp oval	Ovate	smooth	light brown	Low	sweet
very hard	Ovate	Ovate	smooth	light brown	Low	sweet
very hard	Ovate	Ovate	wrinkle	dark brown	intermediate	slightly bitter
Hard	Ovate	Ovate	smooth	light brown	Low	sweet
Soft	Sharp oval	Ovate	intermediate	intermediate	Low	sweet

Table 5. Contd.

intermediate	Ovate	Ovate	intermediate	dark brown	intermediate	slightly bitter
intermediate	Ovate	Ovate	Slightly wrinkle	light brown	intermediate	sweet
Hard	Ovate	Ovate	Slightly wrinkle	light brown	Low	sweet
Hard	Sharp oval	Ovate	Slightly wrinkle	intermediate	Low	sweet
Hard	Ovate	Ovate	Slightly wrinkle	light brown	Low	sweet
Hard	Ovate	Ovate	Slightly wrinkle	light brown	Low	sweet
Hard	Ovate	Ovate	Slightly wrinkle	light brown	Low	sweet
very hard	Ovate	Ovate	Slightly wrinkle	intermediate	intermediate	sweet
Hard	Ovate	Ovate	Slightly wrinkle	intermediate	intermediate	sweet
intermediate	Sharp oval	Ovate	Slightly wrinkle	intermediate	Low	sweet
very hard	Ovate	Ovate	Slightly wrinkle	light brown	intermediate	sweet
very hard	Ovate	Ovate	Slightly wrinkle	light brown	intermediate	sweet
Hard	Ovate	Ovate	Slightly wrinkle	light brown	Low	sweet
Soft	Ovate	Ovate	Slightly wrinkle	light brown	Low	sweet

All genotypes had sweet taste and there was no genotype with bitter taste (Table 5).

Kernel color was light brown, mid brown or dark brown depending on the genotypes. Tf11 and Tf17 had dark brown kernel and Tf16, Tf20, Tf27, Tf28 and Tf29 had mid brown kernel colour. However, others had light brown kernel. This is an important characteristic because the market prefers light brown kernel for almond.

Genotypes Tf15, Tf24 and Tf28 had the longest shells (33.10, 32.40 and 32.38 mm, respectively), However, the shortest shell belonged to Tf16 (24.10 mm), (Table 6). The highest shell width, thickness and weight (4.24 g) belonged to Tf15, while this figure was 3.11 g for "Tardi nonpareil" at the same condition. On the other hand, Tf29 had the lowest shell weight, length, width and thickness (Table 6).

The longest kernel (23.98 and 23.44 mm) was observed in genotypes Tf15 and Tf28; however, the shortest kernel belonged to Tf20. Genotypes Tf15 and Tf28 had also the highest width, thickness, as well as weight of kernel (1.16 and

1.13 gr, respectively). The highest kernel/ shell weight was observed in Tf23, Tf16, Tf4, Tf6 and Tf18 (0.33, 0.33, 0.32, 0.32 and 0.32%, respectively). This ratio was 0.25 in "Tardi nonpareil" (control). There was no double kernel in studied genotypes, whereas, it was 5% in "Tardi nonpareil".

### General conclusion

The main objective of this study was to obtain late blooming self-compatible genotypes with good fruit and kernel quality. Late spring frost is one of the main problems in almond production in temperate climate and improving almond to produce late blooming cultivars is very important (kester, 1965; kester and asi, 1975; kester et al., 1977; Sosias i compony et al., 1999; Sanchez prez et al., 2007). Among all studied genotypes, Tf6 and Tf23 were recognized as very late blooming self-compatible genotypes. Their fruits and kernels were smaller than "Tardi nonpareil"

(Control). The kernel of these two genotypes were light brown, uniform, smooth, sweet, with no double kernel. In addition, the shell of Tf6 was soft. In addition, time of fruit ripening, absence of double kernel, the high ratio of kernel/ shell and kernel quality characteristics such as size, weight and color are also very important (kester, 1965; Kester and Asi, 1975; kester et al., 1977; Sosias i company et al., 1999; Sanchez-prez et al., 2007).

The highest dry fruit weight (4.24 and 4.06 g) and kernel weight (1.16 and 1.13 g) belonged to Tf15 and Tf28. Tf15 was recognized as self-compatible, but Tf28 was self-incompatible. Tf28 and Tf15 were classified as late blooming, because they started to bloom four and five days earlier than "Tardi nonpareil", respectively.

Fruits, shell and kernel of Tf15, Tf24 and Tf31 had also good qualities. Finally, self-compatible genotypes Tf6 and Tf23 are recommended for areas with the possibility of late spring frost. However, self-compatible genotypes Tf15 and Tf24 are recommended for areas with the possible fewer late spring frosts.

**Table 6.** Evaluation of quantitative traits of fruit and nut and kernel of the 23 almond genotypes as well as cultivar Tardi non pareil as a control.

Genotypes	Fruit length (mm)	Fruit width (mm)	Fruit thickness (mm)	Fruit weight (g)	Nut length (mm)	Nut width (mm)	Nut thickness (mm)	Nut weight (g)
Tf 2	37.60 de	26.14 egdf	21.18 gh	9.75 hg	31.84 bcd	19.40 hgi	13.82 gf	2.85 g
Tf 3	32.16 ijk	23.38 ij	18.78 mlk	8.56 k	24.78 hi	19.78 hg	13.44 gh	1.95 l
Tf 4	30.66 ljk	25/00 igh	19.86 ij	8.08 l	26.12 g	19.96 fg	13.36 ghi	2.38 hj
Tf 5	36.12 feg	25.62 eghf	19.82 ijk	8.65 k	28.90 e	18.38 hg	12.98 jhi	2.14 k
Tf 6	32.26 ij	26.62 egdf	21.26 gh	9.25 i	24.62 hi	18.86 kj	13.36 ghi	1.90 lm
Tf 7	34.76 fhg	27.58 d	21.06 gh	9.83 g	28.84 e	22.36 d	14.66 e	3.25 f
Tf 8	35.05 fhg	27.50 d	21.10 gh	9.88 g	28.82 e	23.00 cd	14.44 ef	3.30 f
Tf 11	36.66 fe	27.60 d	21.60 gf	10.82 d	31.10 cd	21.10 e	14.80 de	3.22 f
Tf 15	39.96 c	30.15 b	25.16 b	15.96 b	32.38 ab	26.44 a	17.71 a	4.24 a
Tf 16	29.94 l	23.22 kj	19.10 mljk	6.60 op	24.10 i	18.74 kji	14.32 ef	1.88 lm
Tf 17	36.00 feg	25.00 igh	19.08 mljk	7.80 m	25.50 hg	19.00 hji	12.50 j	1.86 lm
Tf 18	34.48 hg	26.36 egdf	20.36 ih	8.92 j	29.00 e	18.44 kj	12.98 jhi	2.18 k
Tf 19	40.18 c	27.22 ed	22.42 ef	10.50 f	30.94 d	23.88 bc	14.94 cde	3.36 e
Tf 20	30.38 l	21.66 k	17.68 n	6.46 p	24.12 i	17.10 l	12.80 jhi	1.71 n
Tf 23	29.24 l	21.62 k	18.52 mn	6.806 o	24.78 hi	17.26 l	12.74 jhi	1.85 m
Tf 24	44.84 a	33.94 a	27.46 a	16.54 a	32.40 ab	24.94 b	15.98 b	3.95 b
Tf 26	37.78 de	26.33 egdf	20.48 ih	10.62 ef	31.50 bcd	21.90 d	14.94 cde	3.42 d
Tf 27	40.24 c	27.74 d	23.00 ed	12.60 dc	31.26 cd	23.54 bc	15.06 cde	3.75 c
Tf 28	42.00 b	32.08 b	24.24 bc	16.06 b	33.10 a	24.16 b	16.28 b	4.06 ab
Tf 29	33.30 ih	25.12 ighf	19.72 iljk	7.60 n	24.96 hi	17.10 l	11.66 l	1.55 o
Tf 31	38.76 dc	29.45 c	23.60 cd	13.75c	32.10 bc	24.40 b	15.65 bc	4.01 b
Tf 32	37.42 de	26.88 edf	21.42 gh	9.71 h	32.50 ab	20.75 ef	15.50 cd	3.18 fg
Tf 35	34.24 hg	24.24 ihg	18.68 ml	7.64nm	27.28 f	18.26 kj	12.68 ji	2.59 h
Tardy nonpareil	41.85 b	26.34 degf	21.10 gh	9.80 g	33.10 a	22.02 d	14.02 f	3.11 fg
<b>kernel length (mm)</b>	<b>kernel width (mm)</b>	<b>Kernel thickness (mm)</b>	<b>Kernel weight (g)</b>	<b>In-shell/kernel ratio (%)</b>	<b>Sound Kernels (%)</b>	<b>Empty nuts (%)</b>	<b>Double kernels (%)</b>	
22.80 bc	11.08 h	6.35 defgh	0.75 g	0.264 ij	90 c	10 b	0 b	
18.82 ih	12.00 f	6.48 cdefg	0.65 j	0.333 a	95 b	5 c	0 b	
20.16 f	12.12 ef	6.86 bc	0.78 f	0.327 ab	100 a	0 d	0 b	
21.42 e	11.06 h	6.18 fgghi	0.68 i	0.317 bcd	100 a	0 d	0 b	
18.28 i	11.52 g	6.80 bc	0.64 kj	0.326 abc	100 a	0 d	0 b	
19.64 gf	12.87 d	6.13 ghi	0.72 h	0.221 l	100 a	0 d	0 b	
19.72 gf	12.96 dc	6.20 fgghi	0.74 gh	0.224 l	100 a	0 d	0 b	
23.05 b	12.50 de	5.62 j	0.70 ih	0.217 l	85 d	15 a	0 b	

Table 5. contd.

23.44 ab	14.71 a	7.08 b	1.13 ab	0.266 ij	100 a	0 d	0 b
18.56 ih	11.78 fg	6.26 fg	0.62 kj	0.330 a	90 c	10 b	0 b
20.20 f	11.86 fg	5.90 ji	0.52 m	0.279 fgh	85 d	15 a	0 b
21.46 e	11.08 h	6.20 fg	0.71 h	0.325 abc	95 b	5 c	0 b
21.84 ed	13.36 cd	7.06 b	1.06 c	0.315 cd	95 b	5 c	0 b
17.42 j	10.24 ij	6.14 ghi	0.48 n	0.281 fgh	100 a	0 d	0 b
18.40 i	10.52 i	5.88 ji	0.50 mn	0.270 ih	100 a	0 d	0 b
23.303 b	13.92 b	7.10 b	1.10 b	0.278 fgh	100 a	0 d	0 b
21.87 ed	12.59 de	6.75 bcd	0.99 d	0.289 f	90 c	10 b	0 b
22.25 d	13.40 cd	6.66 cde	0.96 d	0.256 j	90 c	10 b	0 b
23.98 a	13.84 bc	7.60 a	1.16 a	0.286 gf	100 a	0 d	0 b
19.28 gh	9.78 j	6.13 ghi	0.47 n	0.303 e	100 a	0 d	0 b
23.10 b	13.95 b	6.70 bcde	1.10 b	0.274 igh	100 a	0 d	0 b
23.00 b	12.25 ef	6.75 bcd	0.98 d	0.311 de	100 a	0 d	0 b
19.94 gf	11.00 hi	5.92 ji	0.62 kj	0.241 k	95 b	5 c	0 b
23.00 b	12.00 f	6.40 defgh	0.79 f	0.254 j	95 b	0 d	5 a

To produce self-compatible genotypes is also very important because it results in the production of uniform nuts and reduces costs of orchard management (Rahemi, 2002).

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