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Phenology and self-(in)compatibility of some apricot (*Prunus armeniaca* L.) promising genotypes

K. Oroji Salmasi[1], R. Gharesheikhbayat[2], S. M. Miri[3*], M. Pirkhezri[4] and D. Davoodi[5]

[1 and 3] Department of Horticulture, Karaj Branch, Islamic Azad University, Karaj, Iran.
[2 and 4] Temperate Fruits Research Center, Horticultural Science Research Institute, Agricultural Research, Education and Extension Organization, Karaj, Iran.
[5] Department of Nanotechnology, Agricultural Biotechnology Research Institute of Iran, Karaj, Iran.
[*]Corresponding author's Email address: smmiri@kiau.ac.ir

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ABSTRACT

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Self-incompatibility and late spring frosts are the most important limiting factors in Asian apricot (*Prunus armeniaca* L.) production. Therefore, it is important to develop late-blooming and self-compatible genotypes in apricot breeding programs in order to increase yield, and in some cases to eliminate the need for pollinizers. This study was carried-out to evaluate the phenology and self-(in)compatibility in cv. Shahroudi and twenty-five apricot promising genotypes under Karaj environmental conditions during 2019-2020 growing seasons. Apricot genotypes flowered from early to late March (3rd to 26th March), and fruits harvested from early to late June. Genotype 499 was identified as a late blooming genotype. *In vitro* pollen germination varied from 0.0 to 74.0%, and showed no correlation with fruit set. Fruit set studies showed 17 genotypes were self-compatible (fruit set of 5.2 to 44.2%). The fluorescent microscopic examinations of the pollen tube growth in the style confirmed 11 genotypes as self-compatible, with at least one pollen tube reaching the ovary. Combining the results of the three primers designed from the first and second *Prunus S-RNase* introns, 11 *S*-alleles were identified, among which S_6, S_c and S_2 were the most frequent. Two late-bloom and self-compatible genotypes 447 and 534 were selected, and several late-bloom or self-compatible genotypes were identified which can be used in future apricot breeding programs.

Keywords: apricot, fluorescence microscopy, fruit set, pollen germination, S-allele

INTRODUCTION

Apricot originates from the north-eastern regions of China (Ham, 2009). Iran is also considered the other center of cultivated apricot (Bourguiba *et al*., 2020). The apricot (*Prunus armeniaca* L.) germplasm in Iran is genetically rich with a wide range of pollen and fruit diversity (Arzani *et al*., 2005).

Considering its wide adaptability, the cultivated apricots are divided into the Central Asian, the Iran-Caucasian, the European, and the Dzhungar-Zailij eco-geographical groups (Zhebentyayeva *et al*., 2012). Turkey, Uzbekistan, Iran, Italy, and Algeria are the top five apricot-producing countries in the world (FAO, 2021).

There are many apricot cultivars in the world, however, new cultivars are to be developed to adapt to the changing climate as well as the requirements of producers and consumers (Zhebentyayeva *et al*., 2012). The main purposes of apricot breeding programs are tree vigor, productivity, late-blooming, self-compatibility, fruit quality, disease resistance, and adaptability to changing climate (Ham, 2009; Zhebentyayeva *et al*., 2012). Hybridization is the most common method used for development of apricot cultivar.. Crosses among apricots belonging to different

eco-geographical groups have been proposed to develop and select cultivars with high fruit quality and better adaptation to different environmental conditions (Zhebentyayeva *et al*., 2012; Shamsolshoara *et al*., 2021).

In the genus *Prunus*, yield is reduced by gametophytic self-incompatibility (GSI). This system is controlled by a multiallelic *S*-locus including a pistil-expressed *S-RNase*, and the pollen-expressed *F-box* (*SFB*) which arrests pollen tube growth in the style (Muñoz-Sanzetal, 2017; Herrera *et al*., 2018; Gordillo-Romero *et al*., 2020). Self-incompatible cultivars are unsuitable due to their requirement for pollinizers with synchronized blooming times (Burgos *et al*., 2004; Shamsolshoara *et al*., 2022a).

The majority of the European apricots group are considered self-compatible, while other eco-geographical groups are mostly self-incompatible (Gharesheikhbayat, 2010). GSI is mainly determined by assessing fruit set or pollen tube growth following controlled self-pollination, and PCR analysis by *S*-alleles (Imani *et al*., 2014; Najafi *et al*., 2015; Jamshidi *et al*., 2021).

The main objective of this study is to evaluate the phenology and determine the self-(in)compatibility in several apricot promising genotypes selected based on fruit yield and fruit quality.

MATERIALS AND METHODS

Plant material

The plant material consisted of twenty-five apricot progenies derived from open pollination of Iranian self-incompatible (cv. Shahroudi and cv.Shams as maternal parents) and Italian self-compatible (cv. San castrese, cv. Vitillo, Cafona and cv. Palumella as parental parents) apricot cultivars in the experimental orchard of Horticultural Science Research Institute, Karaj, Iran. The preliminary selection of genotypes was carried out based on their fruit yield and fruit quality (Oroji Salmasi *et al*., 2023). These 5-year-old promising genotypes were studied in two consecutive years (2019-2020). Cv. Shahroudi was also used as control. The mean monthly temperature from March to June is presented in Table 1.

Table 1. Mean daily temperature during blooming to fruit ripening of apricot genotypes (March to June) in 2019 and 2020 growing seasons

	Temperature (˚C)			
Year	March	April	May	June
2019	8.1	12.5	20.6	27.6
2020	10.6	12.4	21.0	26.1

Phenological characteristics

The main phenological stages were defined according to the BBCH General Scale (Meier *et al*., 1994). The defined phenological stages were as follows: flower bud swelling, beginning of flowering (10% of flowers open), full bloom (50% of flowers open), end of flowering (all petals fallen), fruit growth and development, and fruit ripening. To evaluate and record the phenological stages, three healthy trees (replications) were selected. For each tree, four branches were tagged from each compass direction.

In vitro pollen grain germination

Fresh pollen grains were collected and distributed on the culture medium containing 0.02 $g.l^{-1}$ boric acid, 0.3 $g.l^{-1}$ calcium nitrate, 10 $g.l^{-1}$ agar and 150 $g.l^{-1}$ sucrose. After 24 h, pollen grain germination percentage was determined under a light microscope from five areas of petri dish. Pollen grains were identified as germinated when: pollen tube length ≥ pollen grain diameter (Gharesheikhbayat, 2010).

Field-controlled self-pollination

Two branches with flowers at the balloon stage in the north and south directions of the tree were protected using a cloth bag. The self-pollination was carried out two days later using a small brush. The initial and final fruit sets were counted on two and eight weeks after self-pollination, respectively (Oroji Salmasi *et al*., 2022). Self-incompatibility was evaluated following Audergon *et al*. (1999). Genotypes which had less than 5% of fruit set derived from self-pollination were considered self-incompatible (Audergon *et al*., 1999).

Fluorescent microscopic observation of pollen tube growth

Shoots with flower buds at the balloon stage were isolated in the orchard and taken to the laboratory. They were kept in 5% (w/v) sucrose solution at room temperature (20 ± 2 ˚C). The opened flowers were hand self-pollinated using a fine brush. After 96 hours, the self-pollinated pistils were fixed in FAA (70% ethanol: acetic acid: formaldehyde (18: 1: 1, v/v/v) and stored at 4 ˚C, until required. The fixed pistils were washed several times with distilled water and left in 8N NaOH for 8 hours, then stained with 0.1% (v/v) aniline blue (Jamshidi *et al*., 2021). Pollen tube growth in the style was tracked under a fluorescent microscope (Eclipse TE300, Nikon).

S-genotyping

Genomic DNA was extracted from young leaves using the protocol described by Saghai-Maroof *et al*. (1984). DNA quality and

quantity were assessed using electrophoresis and NanoDrop ND-1000 spectrophotometer (Bio-Science, Hungary).

PCR amplifications were carried out in 25 µl reaction volume, containing 12.5 µl Master Mix 2x (Sinaclon, Iran), 1.5 µM of each forward and reverse primers, 2 µl (100 ng) of genomic DNA, and 7.5 µl ddH_2O. Three primers designed from *Prunus S-RNase* regions were used to amplify the first (SRc-F/SRc-R and PaConsI-F/PaConsI-R) and second (EM-PC2consFD/EM-PC3consRD) introns (Sonneveld *et al*., 2003; Romero *et al*., 2004; Sutherland *et al*., 2004; Vilanova *et al*., 2005).

PCR was performed on an Eppendorf's Master cycler Gradient. Cycling conditions for SRc-F/SRc-R primers consisted: initial denaturation at 94 °C for 60 s, followed by 37 cycles of denaturation at 94 °C for 20 s, annealing at 60 °C for 50 s, and extension at 68 °C for 60 °C; and a final extension at 68 °C for 10 min. The temperature profiles used for PaConsI-F/PaConsI-R and EM-PC2consFD/EM-PC3consRD primers were as described by Sonneveld *et al*. (2003) and Sutherland *et al*. (2004), respectively. The amplified bands were separated by gel electrophoresis in 1% (w/v) agarose gels at 95 V for 45-60 min using TAE 1x as running buffer.

The DNA bands were visualized using DNA Safe Stain EP5082 (Sinaclon, Iran). Amplified band sizes were estimated using a 100 bp DNA ladder (Sinaclon, Iran), and compared with those previously published by Vilanova *et al*. (2005), Sonneveld *et al*. (2003), Zhang *et al*. (2008), Halász *et al*. (2010) and Pinar *et al*. (2017).

Statistical analysis

The phenology and pollen grain germination data were subjected to an analysis of variance (ANOVA) using SPSS software ver. 22 and means were compared by Duncan's Multiple Range Test (DMRT) at $p < 0.01$. The relationship between flowering and fruit ripening dates as well as pollen grain germination and fruit set were estimated using Pearson's correlation coefficient test.

RESULTS

Phenological stages

Combined analysis of variance revealed significant differences among apricot genotypes for date of blooming, and fruit ripening affected by year, genotype and their interactions (Table 2).

Table 2. Combined analysis of variance for date of blooming and fruit ripening, and blooming period of apricot genotypes

		Mean square		
S.O.V.	d.f.	Date of blooming	Blooming period	Date of fruit ripening
Year	1	6.942*	3.769	42.481**
Error 1	4	1.936	1.503	1.972
Genotype	25	20.210**	1.797	35.481**
Year × Genotype	25	10.858**	1.548	20.167**
Error 2	100	1.865	1.308	5.442
C.V. (%)		24.20	3.30	1.90

* and **: significant at the 5% and 1% probability levels, respectively.

On average, blooming and fruit ripening in 2019 occurred 1.3 and 2.9 days later than in 2020, respectively. The apricot genotypes bloomed from early to late March (Fig. 1). Flowering began first in genotypes 450 and 553 on 5th March in 2019 and genotype 450 on 3rd March in 2020, while genotypes 447, 462, 484, 499, 533, 534 and 548 were the last to began flowering on 16th March 2019, and genotype 499 on 14th March in 2020. Cv. Shahroudi began flowering on 3rd March 2019 and 5th March in 2020 (Fig. 1).

Full bloom was at 4-10 and 4-8 days after the begining of flowering in 2019 and 2020, respectively. It was earliest on 12th March in 2019 (two genotypes) and 13th March in 2020 (10 genotypes), and latest on 22nd March 2019 (12 genotypes) and 21st March in 2020 (10 genotypes) (Fig. 1). The end of flowering was earliest in genotype 450 (17th March in 2019 and 15th March 2020) and latest in genotypes 447, 462, 484 and 499 on 27th March 2019 and in genotypes 413, 447, 462, 464, 484, 499, 533, 534 and 548 on 26th March in 2020. The flowering period ranged from 9-10 days (genotype 552 in 2020 and 2019, respectively) to 14 days (genotypes 509, 553 and 571 in 2019, and genotypes 413, 447, 462, 464, 484, 533, 534, 548, 588 and 592 in 2020) (Fig. 1).

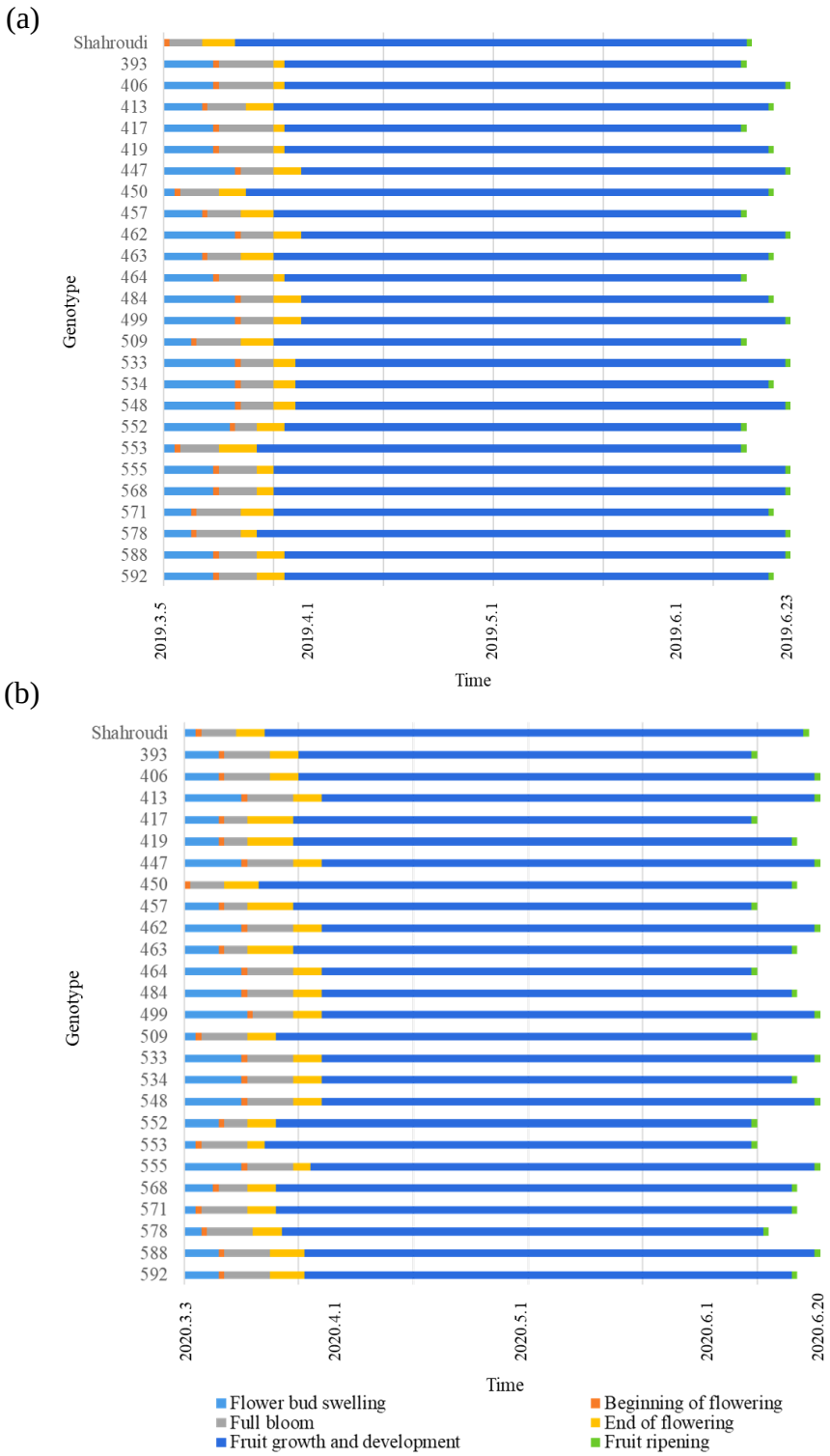

Fig. 1. Phenology of apricot genotypes in 2019 (a) and 2020 (b)

Fruit ripening time was earliest on 15th June 2019 and 9th June in 2020 (genotypes 393, 417, 457, 464, 509, 552 and 553), and latest on 23rd June in 2019 (genotypes 406, 499, 533, 548, 555, 568, 578 and 588) and 20th June in 2020 (genotypes 406, 413, 447, 462, 499, 533, 548, 555, 578 and 588). The fruits of cv. Shahroudi ripened on 25th June in 2019 and 20th June in 2020.

Pollen grain germination

The *in vitro* pollen grain germination rate of apricot genotypes varied significantly and was generally high (Table 3).

Table 3. Analysis of variance for pollen grain germination percentage of apricot genotypes

S.O.V.	d.f.	Mean square
Genotype	25	1821.255**
Error	104	35.360
C.V. (%)		14.7

**: significant at the 1% probability level.

The highest pollen grain germination (73.0, 74.0 and 69.4%) observed for genotypes 413, 457 and 463, respectively (Table 4). This was followed by the 548, 464, 450, 484, 578, 393, 417 genotypes and cv. Shahroudi (46.2 to 61.8%). The pollen germination rate of other genotypes was lower than cv. Shahroudi.

Table 4. Mean comparison of pollen grain germination in apricot genotypes (Mean of 2019 and 2020 growing seasons)

Genotype	Pollen grain germination (%)	Genotype	Pollen grain germination (%)
457	74.0a	533	37.0e-g
413	73.0a	568	36.6e-g
463	69.4a	509	33.6e-h
548	61.8b	552	33.2f-i
464	61.2b	588	29.6g-j
450	51.2c	571	29.4g-j
484	49.0cd	534	29.0g-k
578	48.4cd	447	26.8h-k
393	47.8cd	553	25.0i-k
417	47.2cd	406	24.4jk
Shahroudi	46.2cd	555	20.8k
462	41.8de	592	4.6l
499	41.4d-f	419	0.0l

Means followed by at least one letter in common are not significant different at the 1% probability level-using Duncan's Multiple Range Test.

Field fruit set

The highest fruit-set after self-pollination in the orchard was 44.2% (genotype 406) followed by genotypes 447 (22.6%), 413 (19.5%) and 534 (19.4%), respectively (Table 5). However, cv. Shahroudi and genotypes 450, 499, 509, 548, 568, and 578 showed no fruit set, and genotypes 417, 462 and 484 had less than 5% fruit set, indicating that these genotypes were self-incompatible (Table 5). There was no significant correlation between pollen grain germination and fruit set, but significant positive correlation (r = 0.91**) between the initial and final fruit set was observed (Table 6).

Table 5. Initial and final fruit set and self-(in)compatibility of apricot genotypes derived from field self-pollination and cv. Shahroudi (control) in 2019 and 2020 growing seasons

	2019		2020			
Genotype	Initial fruit set (%)	Final fruit set (%)	Initial fruit set (%)	Final fruit set (%)	Mean of final fruit set (%)	SC/SI
Shahroudi	0.0	0.0	0.0	0.0	0.0	SI
393	11.1	11.0	18.5	7.0	9.0	SC
406	62.1	51.0	41.3	37.5	44.2	SC
413	25.5	17.8	22.5	21.1	19.5	SC
417	7.7	1.5	0.0	0.0	0.7	SI
419	8.4	8.4	8.7	5.2	6.8	SC
447	22.2	22.2	28.2	23.0	22.6	SC
450	0.0	0.0	0.0	0.0	0.0	SI
457	20.0	12.5	14.3	8.5	10.5	SC
462	24.2	3.0	9.7	2.4	2.7	SI
463	14.3	5.7	17.4	6.5	6.1	SC
464	8.0	8.0	9.6	8.0	8.0	SC
484	3.5	3.5	4.7	3.1	3.3	SI
499	0.0	0.0	0.0	0.0	0.0	SI
509	0.0	0.0	0.0	0.0	0.0	SI
533	17.9	6.8	27.2	4.2	5.5	SC
534	19.7	15.8	28.0	23.1	19.4	SC
548	0.0	0.0	0.0	0.0	0.0	SI
552	5.6	5.6	7.9	6.3	5.9	SC
553	10.9	5.4	4.9	4.9	5.2	SC
555	16.9	4.6	25.3	7.1	5.8	SC
568	0.0	0.0	0.0	0.0	0.0	SI
571	9.6	6.8	9.2	6.1	6.5	SC
578	0.0	0.0	0.0	0.0	0.0	SI
588	23.0	11.0	12.5	10.4	10.7	SC
592	21.8	14.9	26.8	19.6	17.3	SC
Mean	17.5	11.3	16.6	10.8	11.0	

SC: Self-compatible, SI: Self-incompatible

Table 6. Pearson's correlation coefficients between pollen grain germination and fruit set of apricot genotypes

Trait	Pollen grain germination	Initial fruit set
Initial fruit set	-0.16	
Final fruit set	-0.18	0.91**

**: significant at the 1% probability level.

Evaluation of pollen tube growth under fluorescent microscopy

Determination of the self-(in)compatibility of 26 self-pollinated apricot genotypes was performed by the observation of pollen tube growth in pistils under the fluorescent microscope. Germination of pollen grains was observed in the stigma of all genotypes except cv. Shahroudi and genotype 509 (Table 7). Nearly 42% of genotypes (11 genotypes) identified as self-compatible, when at least one pollen tube reached the ovary. In cv. Shahroudi and other remaining genotypes, pollen tube growth stopped in the middle- and lower- third sections of the style, therefore, were considered as self-incompatible.

Table 7. Pollen tube growth in the style of apricot genotypes 96 hours after self-pollination

Genotype	No. of evaluated pistils	Germinated pollen grains	Pollen tube growth				SC/SI
			Upper-third of the style	Middle-third of the style	Lower-third of the style	Into ovary	
Shahroudi	12	N					SI
393	5	H				+	SC
406	5	H				+	SC
413	5	H				+	SC
417	4	L	+				SI
419	13	M				+	SC
447	5	H				+	SC
450	4	M			+		SI
457	7	H				+	SC
462	7	M-H		+			SI
463	6	H		+			SI
464	5	H				+	SC
484	4	H			+		SI
499	4	M	+				SI
509	4	N					SI
533	4	H		+			SI
534	4	M-H				+	SC
548	5	H			+		SI
552	7	M			+		SI
553	4	H		+			SI
555	4	M		+			SI
568	4	M			+		SI
571	8	M-H				+	SC
578	7	H		+			SI
588	11	M				+	SC
592	7	H				+	SC

SI: Self-incompatible; SC: Self-compatible; N: No germination; L: Low germination (5-10%); M: Medium germination (20-40%); H: High germination (>50%).

Identification of *S-RNase* alleles

Three pairs of primer pairs were used to identify the *S*-alleles in 25 apricot genotypes (Table 8). EM-PC2consFD/EM-PC3consRD primers showed high degree of length polymorphism, so four bands were amplified ranging from 657 to 1300 bp. On the other hand, the13 bands yielded by the first intron (SRc-F/SRc-R and PaConsI-F/PaConsI-R) ranged from 222 to 458 bp. By combining the results of the three primer pairs used, 11 *S*-alleles were identified (Table 8).

Rc-F/SRc-R and PaConsI-F/PaConsI-R were the most useful primer combinations as they amplified six apricot *S*-alleles (S_c, S_1, S_2, S_5, S_6, S_{20} and S_1, S_5, S_6, S_{13}, S_{14}, S_{16}, respectively). The primer combination EM-PC2consFD/EM-PC3consRD was less effective as could amplify only four alleles (S_2, S_4, S_6 and S_9). Designed primers amplified two bands in 11 genotypes, while for 14 genotypes only one band was yielded (Table 8). S_6 was the most frequent *S*-allele (22.2%), followed by S_c, S_2 (16.6%), S_1, S_{16} (11.1%), and S_5 (8.3%), while S_4, S_9, S_{13}, S_{14}, and S_{20} alleles were the only found in genotype 588 (Table 8).

Table 8. Allele sizes (bp) amplified by three primer pairs used for *S*-alleles identification in apricot genotypes

Genotype	Primer			*S*-genotype
	SRc-F/SRc-R	PaConsI-F/PaConsI-R	EM-PC2consFD/EM-PC3consRD	
393	353			S_c
406	353	413		S_cS_{16}
413	353		1300	S_cS_6
417	420	380		S_1S_6
419			1200	S_4
447		380		S_1
450	332		900	S_2
457	327		657, 900	S_2S_9
462	353			S_c
463		443		S_6
464		334		S_{14}
484		443		S_6
499	332			S_2
509	400	458		S_1S_{13}
533	420	413		S_6S_{16}
534	353	413		S_cS_{16}
548	327	443		S_2S_6
552	353			S_c
553		393		S_5
555	327			S_2
568	375		1300	S_5S_6
571	400			S_1
578			900, 1300	S_2S_6
588	222			S_{20}
592	375	413		S_5S_{16}

DISCUSSION

The phenology of the apricot genotypes is affected by environmental conditions (especially temperature) and genotype. The blooming begins follows the end of the endo- and eco-dormancy (Milošević *et al*., 2010). Low or high temperatures during dormancy are important factors in determining the blooming time (Kitamura *et al*., 2017). The higher mean temperature in March 2020 caused earlier flower buds blooming.

On the other hand, one of the main limiting factors of apricot production in temperate regions is frost blossoms due to late spring frosts (Milošević *et al*., 2010). Late blossoming is a critical factor to prevent spring frost damage (Mashhadi and Khadivi, 2022). Therefore, identifying or developing of new late-blooming apricot cultivars is one of the main objectives of apricot breeders (Oprita and Gavat, 2019).

Demirtas *et al*. (2010) crossed Turkish and foreign apricot varieties reciprocally, but none of the obtained hybrids were promising. Oprita and Gavat (2019) used cross-, self- and open-pollination breeding methods, and obtained two late-blooming apricot genotypes with reasonable yield and high fruit quality. In our study, the genotype 499 was late blooming in both years (6-7 days after cv. Shahroudi), which could be a suitable candidate for introduction in areas at late spring frost risk. Mashhadi and Khadivi (2022) selected 48 late-blooming apricot genotypes from 278 seedlings that could be useful as parents in devloping late blooming cultivars.

In *Prunus* genus, the fruit set is dependent on pollen grain germination, pollen tube growth and fertilization (Fotirić Akšić *et al*., 2022). The viability and germination percentage of pollen grains of most apricot cultivars are high (Burgos *et al*., 2004). The acceptable threshold of pollen grain germination in the plum is 25% (Wertheim, 1996). Similarly, it can be stated that in our study the pollen grain germination of most of apricot genotypes was beyond the threshold and acceptable.

Male sterility has been reported in apricot (Burgos *et al*., 2004). Since several genotypes with poor pollen grain germination had normal-shaped anthers (swollen and yellow appearance), and pollen germination on the stigma was medium or high, male sterility does not appear to have caused poor pollen grain germination in our study. The non-significance correlation between pollen grain germination and fruit set could also confirm that factors other than male sterility (such as self-incompatibility) affected fruit set following self-pollination. Initial and final fruit set percentages were slightly higher in

2019 than in 2020 (0.9 and 0.5%, respectively). Kodad *et al*. (2013) have also reported annual variations in fruit sets in apricot.

The most fruit drop occurred during the first two weeks after pollination, and thereafter about 6% of fruits dropped until fruit ripening. Flower and fruit drop at the initial stages of fruit set is the main problem in reducing the productivity of apricot trees (Zarrinbal *et al*., 2018). The major fruitlets drop occurred from 12-35 days after blossoming (Lichou *et al*., 1995). Fikret Balta *et al*. (2007) found that the highest fruitlet drop in apricot varieties occurred 3-4 weeks after full bloom. The fruit drop is then remarkably reduced, and is not related to lack of pollination and fertilization (Lichou *et al*., 1995).

The fruit set percentages recorded of most self-compatible genotypes were less than commercial product of apricot [20-25% (Westwood, 1993)]. There are many environmental, physiological and genotype-dependent factors such as temperature, pollen source, bee activity, floral biology, and fertility, which influence fruit set and yield (McLaren *et al*., 1996; Fikret Balta *et al*., 2007; Ruiza and Egea, 2008; Shamsolshoara *et al*., 2022b). In agreement with our results, Nekonam *et al*. (2010) stated that high temperature inside the bag during controlled pollination may cause more fruit drop.

Apricot cultivars are considered self-compatible if the fruit set after self-pollination in the field is more than 3% (Burgos *et al*., 1997) or 5% (Audergon *et al*., 1999). Our fluorescent microscope observations showed that in genotypes with mean fruit set percentage of less than 6.5%, the pollen tube did not reach the ovary, and considered self-incompatible. Therefore, a fruit set of $\geq 6.5\%$ could be considered as a more accurate threshold for self-compatibility in apricot genotypes.

Complete *S*-genotypes were determined for 11 apricot genotypes (nine different *S*-genotypes). For other 14 genotypes, only partial *S*-genotypes were identified, which could be due to the mismatching of primers with specific *S*-alleles sequences or homozygosity of the genotypes in the case of the *S*-allele (Herrera *et al*., 2018). Only four *S*-alleles were identified by primer EM-PC2consFD/EMPC3consR, while each of primers SRc-F/SRc-R and PaConsI-F/PaConsI-R were able to amplify six *S*-alleles. These results are inconsistent with the report by Zhang *et al*. (2008) who stated that primer EM-PC2consFD/EMPC3consR was able to identify all Chinese apricot *S*-alleles. The reason for this inconsistency may be sought in different origin of the studied apricots.

The S_c-allele has been identified for self-compatibility in apricots, as most self-compatible apricot cultivars show the S_c-allele (Muñoz-Sanz *et al*., 2017). However, in our study, seven self-compatible genotypes did not carry the S_c allele, which is to some extent similar to the results of Lora *et al*. (2019). A study of natural self-compatible mutants of apricots has shown that pollen grains carrying other *S*-haplotypes such as S_1- and S_2-haplotypes or two *S*-loci unlinked pollen-part mutations (PPMs) were able to breakdown the incompatibility system in apricot (Vilanova *et al*., 2006; Zuriaga *et al*., 2013; Muñoz-Sanz *et al*., 2017).

A pollen-part mutation in the S_8 haplotype resulted in S_c allele in apricot. The S_c- and S_8-*RNase* alleles produced equally sized fragments, and the sequences of their first intron regions was identical (Halász *et al*., 2007). Therefore, it is possible that allele S_c- in self-incompatible genotypes is actually allele S_8, which requires band sequencing to be accurately identified.

In the present study, allele S_c was observed in six genotypes. Since maternal parents are self-incompatible, so the S_c allele must have originated from the self-compatible parental parents. Fruit set and fluorescent microscope analyses, respectively, confirmed the self-compatibility of five (genotypes 393, 406, 413, 534 and 552) and four (genotypes 393, 406, 413 and 534) of these genotypes. Halász *et al*. (2010) determined seven self-compatible apricot cultivars out of 55 Turkish and Hungarian apricot cultivars, and self-compatibility of six cultivars were confirmed by open-field fruit set analysis. They found that if the results of several years of fruit set analysis after self-pollination were consistent, it could be considered a reliable method.

CONCLUSION

The evaluation of apricot promising genotypes derived from open pollination of Iranian and Italian apricot cultivars led to the identification of two late-bloom and self-compatible genotypes. Several late-bloom or self-compatible genotypes were also identified which can be considered as valuable germplasm for being used in apricot breeding programs. In addition, *S*-allele of some genotypes was identified which can be used by researchers in choosing the correct pollinizers

in establishment of new apricot orchard.

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