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The Cross-Breeding Performances of Some Peach Varieties

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Abstract: In Turkey, peach breeding activities are quite inadequate. Only one breeding program to develop new peach varieties for fresh market and processing by controlled hybridization has been carried out in Ataturk Central Horticultural Research Institute. The present study was performed to evaluate fruit set rate, and a number of harvested fruit and stratified seed and germinated seed and seedling for each combinations and to determine the best parents to use in future crosses. Forty-two controlled crosses between ten peach varieties were made during 2008 and 2009 and total 32,922 flowers were pollinated. The full ripening fruits were harvested and hybrid seeds extracted. These seeds were stratified without endocarp at 4-5 °C for 40-90 days. The resulted seedlings were planted in an experimental orchard. Some problems including embryo and seed abortions, rosetting, multiple pistil and fungal infection occurred on fruits, trees and seedlings in the different developmental stages. The rates of fruit set were determined to be 78.27% and 73.10% for two years, respectively. Total 3,461 fruits were harvested and 2,730 seeds were stratified. The number of germinated seeds and resulted seedlings were 2,451 and 2,144, respectively. Our results showed that hybrid seeds should be stratified immediately after harvest to shorten breeding period and also stratification without endocarp can be lead the rosetting seedlings. Moreover, fluctuating temperature, not even frost, can cause to embryo abortions. Bayramic Tüysüzü, Sarı Papa, Vivian and Fortuna should be suggested as parent to obtain a large number of hybrid plants in a peach breeding program.

Key words: Abortion, Controlled hybridization, Germination, Peach, Rosetting, Seedling

Bazı Şeftali Çeşitlerinin Melezleme Performansları

Özet: Türkiye'de şeftali ıslahı konusundaki çalışmalar oldukça yetersizdir. Atatürk Bahçe Kültürleri Merkez Araştırma Enstitüsü'nde kontrollü melezleme yoluyla yeni sofralık ve sanayilik şeftali çeşitlerinin geliştirmek için devam eden bir çalışma bulunmaktadır. Bu çalışmada, farklı melezleme kombinasyonlarında meyve tutumu, hasat edilen meyve sayısı, katlanan tohum sayısı ile çimlenme oranı ve melez bitki sayılarının tespiti ve en iyi ebeveyn kombinasyonunun belirlenmesi amaçlanmıştır. 2008 ve 2009 yıllarında ebeveyn olarak on şeftali/nektarin çeşidi kullanılarak 42 kombinasyon oluşturulmuş ve 32,922 adet çiçek tozlanmıştır. Meyve tutum oranları her iki yılda sırasıyla %78.27 ve %73.10 olarak belirlenmiştir. Toplam olarak 3461 adet meyve elde edilmiş ve çekirdekler kırıldıktan sonra 2730 adet tohum katlamaya alınmıştır. Bu tohumlardan 2451 adedi çimlenmiş ve 2144 melez bitkinin araziye dikimi gerçekleştirilmiştir. Araştırma sonuçlarına göre tohumların hasattan hemen sonra katlamaya alınarak bitkilerin yetiştirilmesi, ıslah süresinin kısaltılmasında faydalıdır ancak çekirdek yerine tohum katlaması yapılması melez bireylerde rozetlenmeye neden olabilmektedir. Ayrıca don olayı olmasa bile meyve tutumundan sonraki gece/gündüz sıcaklıkları arasındaki düzensizlikler, embriyo aborsiyonlarına sebep olabilmektedir. Yapılan değerlendirmelere göre Bayramiç Tüysüzü, Sarı Papa, Vivian ve Fortuna tip/çeşitleri, melezleme çalışmalarında çok sayıda melez bireyin sorunsuz şekilde edilmesinde kullanılabilecek ebeveynler olarak belirlenmiştir.

Anahtar kelimeler: Aborsiyon, Kontrollü melezleme, Melez bitki, Şeftali, Rozetleşme, Çimlenme

Introduction

The cultivation of peach originated in China dates back to 3000 BC (Huang et al., 2008). First peach cultivars were usually developed by selecting the best plants from nature. Controlled peach breeding works began in the USA in the early 1900's and some cultivars such as J.H.Hale, Elberta and Redhaven that have been still used in many breeding programs through world were developed (Dosba, 2003; Okie *et al*., 2008).

The 20th century was the golden age of peach breeding and in that century a lot of peach cultivars were developed. The majority (43-61%) of the new peach cultivars were developed through hybridization. Today, significant peach breeding efforts have been undertaken by both public and private breeders in many countries like the USA, Spain, Italy, China, France, Australia and South Africa (Giovannini et al. 2006; Krewer et al. 1997; Todorovic et al. 1998; Bellini et al. 2000; Jiang et al. 2002; Hagillih et al. 2003; Sherman and Lyrene, 2003; Sansavini et al. 2006; Okie et al. 2008; Cantin et al. 2010). Although Turkey is one of the major peach producer in the world, recent peach breeding activities by controlled hybridization is only one. The firs breeding program began at Cukurova University in 1995 year, but it ended without releasing a new cultivar (Tanriver and Küden, 2004).

As a result of the USA peach breeding programs, improved USA cultivars spread around the world and some of these cultivars were used as parent in many peach breeding programs that has led to reduction in the genetic diversity of peach germplasm (Okie et al. 2008). Therefore, using the local germplasm in breeding programs is very important to add new traits in the genetic pool. Besides, some new progeny which would be ensured for future breeding programs is also important.

Hybridization is one of the methods of fruit tree breeding and usually made by hand after emasculation to ensure crossing between cultivars. Emasculation consists of removing, with fingernails or other tools, the petals, sepals, and stamens before anther dehiscence at balloon stage of flowers. Emasculated flowers of peach do not attract pollinators, so branches are not covered for cultivar development crosses (Arbeloa et al. 2006; Hancock et al. 2008).

Overcoming dormancy and promoting germination, seeds of peach are needed cold-humid stratification at 0-10 °C (opt. 4-5 °C) for 90-120 days (Malcolm et al. 2003a; Malcolm et al. 2003b; Arbeloa et al. 2006). Indeed, there is a relationship between the time of seed germinate and chilling request of seed parent. Many observations indicate that seeds of low-chill genotypes require less time in stratification to germinate than seeds of high-chill genotypes do. This information may be practised when genotypes are wanted to classify for flower bud chilling requirement at the time of seed germination (Topp et al. 2008).
Abnormally high temperatures during flower bud initiation may lead to double or more pistils, which in turn may give more fruits than one. Furthermore, water stress in late summer also interferes with flower bud development and can cause formulation of more pistils in the following year. (Handley and Johnson, 2000; Engin and Ünal 2004; Bassi and Monet 2008). Multiple pistil formation is usually associated with flower drop immediately after flowering if adequate pollination does not happen or double/triple fruits that have not commercial value occur. In this case, the number of seed that can germinate will decrease.
In Turkey, established peach breeding program at Ataturk Central Horticultural Research Institute (ACHRI) in 2008 is focused on development of late-ripening varieties possessing high fruit quality for fresh consumption and non-melting varieties for processing industry using classical breeding method. Another objective of ACHRI breeding program is to add novel progeny into peach germplasm that can be used in the future breeding work. To obtain enough number of seedlings and the desired results, it is necessary to select the best parents for hybridization. The present study aimed to determine hybridization performance of the parents and to select the best parent varieties. Another aim of this study was to identify local peach parents likely to produce a large amount of seedling and to determine the suitability to use in future breeding work.

Materials and Methods

Progenitors and cross-breeding combinations

Five foreign-commercial peach (Monroe, Rio Oso Gem, Fortuna, Jungerman and Vivian) and five local peach/nectarine (Alyanak Hulu, Bayramiç Tüysüzü Sapanca, Takunyacı-I and Sarı Papa) varieties were used as parents in the controlled crosses. Progenitors with different fruit types were used in this work are: Yellow-fleshed freestone peach (Monroe, Rio Oso Gem, Takunyacı-I); white-fleshed, clingstone, melting peach (Alyanak Hulu); white-fleshed, freestone, melting peach (Sapanca); yellow-fleshed, clingstone, non-melting peach (Fortuna, Jungerman, Vivian, Sarı Papa), and white-fleshed clingstone, melting nectarine (Bayramiç Tüysüzü). Forty-two crossing combinations created to develop fresh and processing market peaches in 2008 and 2009 were shown in Tables 1 and 2.

Emasculation and pollination

The flowers of the female parents were emasculated at balloon stage by fingers. The emasculated flowers were pollinated with pollen collected from the anthers of the male parents in the same day or the next day. Pollinated flowers were not protected, because they do not attract insects (Monet and Bassi, 2008). In one family, about 400 and 2000 flowers were pollinated depending on the current number of flowers (Tables 1 and 2) and the pollinated flowers were counted for each family after pollination.

Raising seedlings

The pits (with endocarp) of peach fruits harvested in the soft full ripe stage were cleaned and cracked. The seeds extracted from endocarp were placed into plastic pots of 10 cm diameter with moist perlite and stored in a refrigerator at 3-5 °C for 40-90 days for seed germination. The germinated seeds having appeared out a young root were transplanted in the greenhouse. At the end of the dormant season, seedlings were planted (one tree per genotype) at a spacing of 3.5 m x 1 m in an experimental orchard in two consecutive years (2009-2010).

Evaluations

Initial fruit set (IFS) was determined by counting the number of fruitlets between 30th and 35th days after pollination and percent initial fruit set was calculated using the following equation: IFS% = Total number of fruit set for each combination / total number of pollinated flower for each combination x 100.

Final fruit set, stratified and germinated seed and seedling number and percent value of these were determined by the formula similar to the above one. The seedling number did not include the losses of seedling in the greenhouse. The number of rosette seedling was recorded for each family. In addition, physiological disorders and infectious diseases during the seedling growth cycle were determined.

Results and Discussion

Pollinated flowers, initial and final fruit set, stratified and germinated seeds and seedling numbers were shown at Table 2 for the year 2008 and at Table 3 for the year 2009 for each hybridization combination. Twenty controlled crosses were made in 2008 and 18,146 flowers were pollinated. Total initial fruit set number was 14,203 (78.27%) and 2,938 (15.99%) fruit was harvested (Table 1). In the second year, 14,776 flowers were pollinated by making twenty-two controlled crosses. Total initial fruit set number was 10,801 (73.10%) and the average percent harvested fruit rate was lower in 2009 (3.54%) than the rate of the first year (Table 2). According to Hesse (1975), initial fruit set of peach can vary from 10% to 90% and this information is consistent with the data of the present work.

No fruit could be obtained from combinations of Sapanca x Monroe (Table 1) and Rio Oso Gem x Sapanca (Table 2). In both combinations, the initial fruit set occurred, but all the fruitlets abscised nearly 1.5 months after the following pollination. This condition could be caused by several factors including fertilization deficient, nutrient deficiencies, climatic conditions, early seed abortion or some diseases such as powdery mildew and leaf curl.

Approximately from 5% to 50% of peach flowers were developed into fruit and it is considered that this rate (final fruit set) should be between 15% and 20% in commercial growing to obtain marketable fruits. It was reported that the final fruit set for a successful controlled pollination can be between 10%-20% (Hesse, 1975; Topp et al. 2008). Milatovic et al. (2010) found that the rates of the average initial and final fruit set by open-pollination in forty peach cultivars were 73% and 42% respectively. The fruit set by self-pollination in peach is usually between 15% and 20% (Nyeki and Szabo, 1996) and the highest set is obtained from non-melting-flesh peach cultivars. The fruit set is lower in nectarines than the one in melting-flesh peaches (Nyeki et al. 1998; Szabo et al. 2000). In the present study, the number of combinations that were more than 10% of the harvested fruit rate was thirteen in 2008 and two in 2009 (Tables 1 and 2). It is accepted that 10%-20% final fruit set in hybridization is a successful pollination (Hesse, 1975; Topp et al. 2008) which is corroborated by our data.

It is known that some factors like the climatic condition during blooming time, pollen production, pollen quality, the presence of pollinators, nutrition of tree, diseases, seed abortions or the need of chilling and heat accumulation of cultivar are effective to initial and final fruit set (Hesse, 1975; Bassi and Monet, 2008). Additionally, the degeneration and abortion of the ovule may be under environmental control and different fruit set rates among cultivars or years for same cultivars can occur due to environmental factors such as rainfall, humidity, temperature, etc. (Nava et al. 2009).

Table 1. According to the hybridization combinations the pollinated flower, initial and final fruit set, stratified and germinated seed, and seedling numbers in the year 2008

Hybridization combinations	Pollinated flower number	Initial fruit set n	%	Final fruit set n	%	Stratified seed n	%	Germinated seed n	%	Seedling n	%
Combinations for fresh market											
Monroe x Bayramiç Tüysüzü	782	569	72.76	59	7.54	46	5.88	38	4.86	35	4.48
Monroe x Alyanak Hulu	722	600	83.10	79	10.94	62	8.59	47	6.51	39	5.40
Alyanak Hulu x Monroe	626	418	66.77	213	34.03	213	34.03	212	33.87	197	31.47
Monroe x Sapanca	844	742	87.91	51	6.04	41	4.86	35	4.15	33	3.91
Sapanca x Monroe	760	391	51.45	0	0.00	0	0.00	0	0.00	0	0.00
Monroe x Takunyacı-I	1696	1493	88.03	189	11.14	152	8.96	137	8.08	122	7.19
Takunyacı-I x Monroe	1039	638	61.41	90	8.66	58	5.58	54	5.20	45	4.33
Rio Oso Gem x B. Tüysüzü	1656	1610	97.22	543	32.79	487	29.41	461	27.84	448	27.05
Rio Oso Gem x Alyanak Hulu	1466	1259	85.88	289	19.71	169	11.53	149	10.16	121	8.25
Alyanak Hulu x Rio Oso Gem	496	446	89.92	264	53.23	261	52.62	256	51.61	228	45.97
Rio Oso Gem x Sapanca	1192	1170	98.15	240	20.13	111	9.31	90	7.55	79	6.63
Rio Oso Gem x Takunyacı-I	1922	1670	86.89	398	20.71	277	14.41	257	13.37	174	9.05
Takunyacı-I x Rio Oso Gem	646	410	63.47	77	11.92	58	8.98	56	8.67	53	8.20
Combinations for processing market											
Fortuna x Sarı Papa	394	259	65.74	79	20.05	63	15.99	41	10.41	16	4.06
Fortuna x Jungerman	449	400	89.09	59	13.14	37	8.24	24	5.35	8	1.78
Vivian x Sarı Papa	879	808	91.92	141	16.04	123	13.99	103	11.72	73	8.30
Sarı Papa x Vivian	1206	433	35.90	37	3.07	36	2.99	24	1.99	26	2.16
Vivian x Fortuna	569	471	82.78	113	19.86	112	19.68	99	17.40	80	14.06
Jungerman x Sarı Papa	369	315	85.37	15	4.07	9	2.44	7	1.90	6	1.63
Sarı Papa x Jungerman	433	101	23.33	2	0.46	1	0.23	1	0.23	1	0.23
Total (n) /mean (%)	**18 146**	**14203**	**78.27**	**2938**	**15.99**	**2316**	**12.61**	**2091**	**11.38**	**1 784**	**9.83**

Fruit doubling can be shown in some years and this has been associated with water stress due to drought or deficit irrigation and high temperature during bud formation (Handley and Johnson, 2000; Engin and Ünal, 2004; Bassi and Monet, 2008). Some cultivars are more prone to formation of double or triple fruit. In this work, multiple pistil formation of all foreign parents was more remarkable, in some cultivars (i.e. Rio Oso Gem) up to eight pistils in one flower. The local peach parents had much less multiple pistils (eight or ten in a tree) while Sarı Papa had none. This disorder caused to the loss of some fruits and to non- germinating of the seeds.

Table 2. According to the hybridization combinations the pollinated flower, initial and final fruit set, stratified and germinated seed, and seedling numbers in the year 2009

Hybridization combinations	Pollinated flower number	Initial fruit set		Final fruit set		Stratified seed		Germinated seed		Seedling	
		n	%	n	%	n	%	n	%	n	%
Combinations for fresh market											
Monroe x Bayramiç Tüysüzü	816	684	83.82	17	2.08	16	1.96	16	1.96	15	1.84
Monroe x Alyanak Hulu	757	578	76.35	5	0.66	3	0.40	3	0.40	3	0.40
Alyanak Hulu x Monroe	633	469	74.09	139	21.96	133	21.01	130	20.54	126	19.91
Monroe x Sapanca	561	348	62.03	13	2.32	9	1.60	9	1.60	8	1.43
Sapanca x Monroe	327	179	54.74	3	0.92	1	0.31	1	0.31	1	0.31
Monroe x Takunyacı-I	1293	1121	86.70	10	0.77	9	0.70	9	0.70	8	0.62
Takunyacı-I x Monroe	322	195	60.56	4	1.24	4	1.24	4	1.24	3	0.93
Rio Oso Gem x B. Tüysüzü	532	398	74.81	7	1.32	6	1.13	6	1.13	6	1.13
Rio Oso Gem x Alyanak Hulu	1195	987	82.59	6	0.50	4	0.33	3	0.25	3	0.25
Alyanak Hulu x Rio Oso Gem	445	297	66.74	59	13.26	55	12.36	52	11.69	49	11.01
Rio Oso Gem x Sapanca	466	225	48.28	0	0.00	0	0.00	0	0.00	0	0.00
Rio Oso Gem x Takunyacı-I	1362	1129	82.89	11	0.81	11	0.81	10	0.73	10	0.73
Takunyacı-I x Rio Oso Gem	371	213	57.41	4	1.08	4	1.08	4	1.08	4	1.08
Combinations for processing market											
Fortuna x Vivian	652	498	76.38	45	6.90	25	3.83	24	3.68	20	3.07
Fortuna x Jungerman	564	452	80.14	41	7.27	20	3.55	15	2.66	11	1.95
Jungerman x Fortuna	166	73	43.98	7	4.22	1	0.60	1	0.60	1	0.60
Vivian x Sarı Papa	669	487	72.80	36	5.38	24	3.59	16	2.39	13	1.94
Sarı Papa x Vivian	2231	1618	72.52	85	3.81	60	2.69	60	2.69	50	2.24
Vivian x Jungerman	581	396	68.16	19	3.27	17	2.93	17	2.93	17	2.93
Jungerman x Vivian	148	67	45.27	4	2.70	4	2.70	4	2.70	4	2.70
Sarı Papa x Fortuna	414	265	64.01	5	1.21	5	1.21	5	1.21	5	1.21
Sarı Papa x Jungerman	271	122	45.02	3	1.11	3	1.11	3	1.11	3	1.11
Total (n) /mean (%)	**14776**	**10801**	**73.10**	**523**	**3.54**	**414**	**2.80**	**392**	**2.65**	**360**	**2.44**

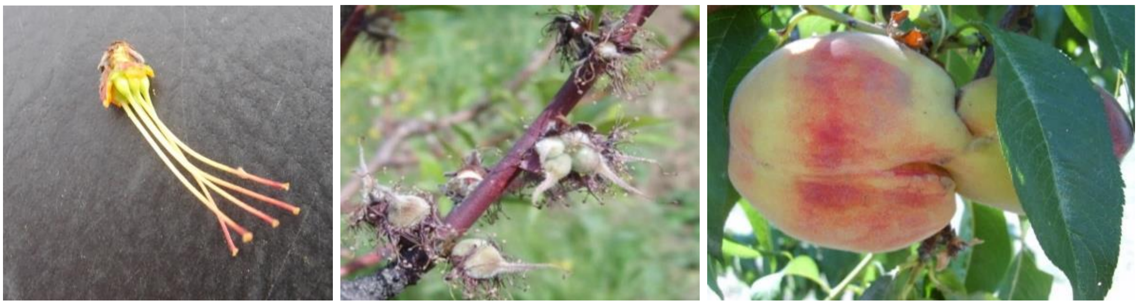

Figure 1. Multiple pistil of peach in different periods

The number of stratified seed was less than the number of harvested fruit for each year (Tables 1 and 2). It is likely that several physiological disorders and diseases (e.g. split pit, brown rot or the late seed abortion) could cause to reduction in value.

In early-ripening peach cultivars, inadequate development of many seeds are common and the embryos of these seeds can have little reserves. Therefore, they are too weak to germinate. The embryo of peach cultivars with a longer fruit development period (FDP; days from flowering to harvest) can develop fully.

Also, if the amount dry matter of embryo increases, the seed germination rate enhances. It has reported that the rates of seed germination and the resulting seedling survival in the seeds which they are obtained from genotypes with a fruit development period more than 105 days can be increased up to 85% (Bacon and Byrne, 1995). Furthermore, high temperatures during the spring also increase the seed dry matter accumulation (Sherman and Beckman, 2003). Although, in this study, early-maturing varieties were not used as parents, some seeds could have accumulated less reserves and not achieved maximum dry weight. These seeds were considered to have aborted after pit hardening since the embryos could not fill the seed coat completely (type I, Fig. 2).

Figure 2. Late-seed abortion of peach (type I)
(Usual (left) and aborted (right) seeds.)

Between seed abortions during late-period and embryo abortions (type II, Fig. 3) significant differences occurred. Accordingly, the seeds in type I wrinkled and did not germinate. In type II, some of the fruits dropped subsequently, while the fruits remained as buttons on the tree could reach approximately 1/4 of full size and ripened for 20 to 30 days from harvest time later. Furthermore, their seeds were completely brown and did not germinate because of the nonviable seed. It was considered that this situation could be due to the effect of fluctuating temperature at day/night, despite temperature did not fall below zero.

Peach seeds have dormancy of seed coat and embryo. To overcome the dormancy and to promote germination, a cold treatment under humid conditions is required (Gomez and Dicenta, 2001). Tukey and Carlson (1945) indicated that stratifying seed removed endocarp has increased the germination rate and shorten the length of germination duration. In this work, since the seeds without endocarp were stratified, the germination duration decreased from 90-120 days (Hesse, 1975) to 40-90 days. It is clear that stratifying only seed without endocarp will contribute to shorten the breeding duration. It is also known that duration of chilling and germination is positively correlated. In many works, it was shown that seeds of genotypes with low chilling are need less time for stratification duration than seeds from high-chill genotypes (Topp *et al.*, 2008). This information should also be used to classify hybrid plants during the seed germination time.

Figure 3. Embryo abortion of peach (type II) (Fruits with aborted embryos did not attain full size because of the nonviable seed.)

The seedling rates in the first year except one breeding combination (Sapanca x Monroe with 0.0%) varied between 0.23% (Sarı Papa x Jungerman) and 45.97 % (Alyanak Hulu x Rio Oso Gem) (Table 1). In the second year, the percent of the resulted seedling excluding Rio Oso Gem x Sapanca (0.0%) was the lowest in Rio Oso Gem x Alyanak Hulu (0.25%) and the highest in Alyanak Hulu x Monroe (19.91%) (Table 2). Almost all germinated seeds turned to seedlings. However, some of seedlings in the greenhouse were lost as a result of fungal infection and rosetting of plant terminal bud until transplanting into the experimental orchard. Although fungicide was used for the control of this disease, fungal pathogen/s attacked the root crown of plants (the base of the trunk) and caused them to rot and to die. It was though that high temperature and humidity conditions in the greenhouse led to emergence of the disease.

The rosetted terminal bud occurred at some of seedlings (Fig. 4a) was pinched out to develop the normal shoots (Fig. 4b); however, some of these seedlings were unable to do so. The numbers of the lost plants due to the rosetting were fifty in 2008 and twenty-two in 2009. The rosette seedlings in the combinations of Rio Oso Gem and Takunyacı-I used as female parents were not detected for each year, but, the other combinations were observed from 1 to 25 at the rosette plants. Sherman and Beckman (2003) indicated that the rosetting can be seen in a lot of stone fruits. The rosetting can occur when seed without endocarp is stratified and it is also variety-dependent (Topp *et al*., 2008). Bacon and Byrne (1995), and Byrne *et al*., (2000) reported that seeds of genotypes with less than 110 days FDP can produce at high rate of rosetted seedlings but this rate drops rapidly when the length of FDP increases. In this work, because early-ripening female parent was not use, the number of rosetting seedling was very low. So, the stratifying without endocarp of hybrid seeds could be considered to the reason of the rosetting in this study.

(a) (b)

Figure 4. Rosetting of peach seedling (left plate; a) and new shoots developing from below the rosetted terminal pinching out (right plate; b)

Conclusions

It is recommended that the hybrid seeds should be stratified without delay after each harvest and to allow germinating in a short period such as approximately 40-90 days. So, it is possible that hybrid plant population can be obtained 5-7 months earlier than usual and this is important in terms of shortening the breeding period. In this work, Fortuna and Vivian as female parents, and Bayramiç Tüysüzü and Sarı Papa as pollen parents were the best ones according to pollen quality and the rates of the fruit set, the fruit harvesting and the seed germination. Alyanak Hulu, Monroe and Rio Oso Gem had shown a good performance; however, progenies from these varieties have had low fruit quality (not shown data) and also the latter two cultivars can occur a lot of multiple pistils in dry and high temperatures condition which it is not desired in breeding.

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