

## **‘HARLAYNE’ X ‘HARCOT’ – PERSPECTIVE CROSSBREED FOR COMBINING GOOD FRUIT QUALITY AND RESISTANCE TO PLUM POX VIRUS**

**Marieta NESHEVA, Valentina BOZHKOVA, Snezhana MILUSHEVA**

Fruit Growing Institute, 12 Ostromila Street, Plovdiv, Bulgaria

Corresponding author email: marieta.nesheva@abv.bg

### **Abstract**

*Most apricot breeding programs aim to obtain genotypes combining good fruit qualities and resistance to Plum pox virus (PPV). Using a combination between field observations and Marker Assisted Selection improves the efficiency of the breeding process. The study was conducted at a breeding orchard in Fruit Growing Institute - Plovdiv, Bulgaria and describes progenies from the hybrid family ‘Harlayne’ x ‘Harcot’, genotyped for resistance to PPV. Fruit ripening period, fruit biometry, total soluble solids (TSS), sensory analyses, fruit ground and over color were observed. The fruit ripening period of the most of the hybrids is after Harcot, the fruits were classified from small to large size and 82% of them surpass the TSS content of the parental cultivars. According to the sensory evaluation the hybrid’s fruits were highly scored (5.07-7.67). All of them were with orange ground color of varying intensity. The fruit over color was from 10 to 80 %. Two of the studied hybrids were selected and grafted on rootstock for final evaluation. A lot of the other hybrids had desirable traits and the breeding process with them will continue in F2 for improving their fruit quality.*

**Key words:** fruit breeding; hybrids; Plum pox virus; pomological traits; *Prunus armeniaca* L.

### **INTRODUCTION**

Most apricot breeding programs point at genotypes combining good fruit qualities and resistance to different pests. The commercial value of apricots depends entirely on the fruit quality. It is determined by the combination of physical and chemical fruit characteristics such as appearance, firmness, taste and aroma (Velisek and Cejpek, 2007). These characteristics are genetically highly variable and their phenotypic expression is influenced by the environmental conditions in the year of cultivation (Dirlewanger et al., 1999). The knowledge of the ways of inheritance of fruit quality traits would result in a higher efficiency of the breeding process and would help the choice of genitors. These traits can be improved by the breeding process and there is a great diversity of genetic resources for them (Krška et al., 2006a). Most of the fruit quality traits are under polygenic control and are quantitatively inherited (Salazar et al., 2013). The conventional breeding is successful in developing cultivars with improved fruit quality but the same cannot be said for the disease resistance. *Plum pox virus* (PPV) is the

most devastating disease and a major limiting factor for the apricot cultivation. The selection of resistant genotypes requires a lot of time and significantly slows down the whole breeding process. Therefore, a number of scientists have focused their efforts on developing molecular markers associated with genes of PPV resistance (Abernathy et al., 2004). The development of Marker Assisted Selection (MAS) effectively complements plant breeding process (Singh B.D., Singh A.K., 2015) and gives us an opportunity to make it shorter by doing a proper selection at the very early stages of it. Our study describes field observations of genotyped for PPV resistance hybrid family ‘Harlayne’ x ‘Harcot’ and its aim is to investigate the way of inheritance of the fruit quality traits and evaluate the progeny.

### **MATERIALS AND METHODS**

This research was conducted at a breeding orchard of Fruit Growing Institute - Plovdiv, Bulgaria. The hybrids were obtained by the methods of conventional fruit breeding and planted in 2011. In 2014 the progeny was genotyped for PPV resistance within the work

on project MARS (7 FP- Collaborative project nr. 613654). This hybrid family consists 153 seedlings. In the present study, the hybrids that fruited consistently over the three years (2015, 2016 and 2017) are described. An average sample of fruits was taken and biometric data was measured with Mitutoyo 500-196-30 Digimatic Absolute Caliper 150 mm. Total soluble solid content (Brix<sup>0</sup>) in juice using a handheld Sper Scientific 300019 Digital Refractometer was determined. For the descriptive characteristics of the fruits, UPOV (2007) and IBPGR (1984) descriptors were used. Sensory evaluation was done by a group of trained consumers. For statistical data processing, Duncan's multiple range test at  $P \leq 0.05$  through IBM SPSS Statistics 19 was used.

## RESULTS AND DISCUSSIONS

Seventeen of the hybrids fruited in the three consistently years. Their ripening period started in the second half of June and it's duration in 2015 was 18, in 2016 - 12 and in 2017 - 20 days. In 2016 the ripening period was shorter than in the other two years probably because this trait is strongly influenced by the climatic factor (Milošević et al, 2010). During the three years, the same trend was observed - fruit ripening time is genetically variable trait and in the progeny, there are hybrids which fruits ripen earlier or later than both parental cultivars. According to Audergon et al. (2011), this is due to the genetic background of the parents which has a strong influence on the inheritance of the fruit ripening time.

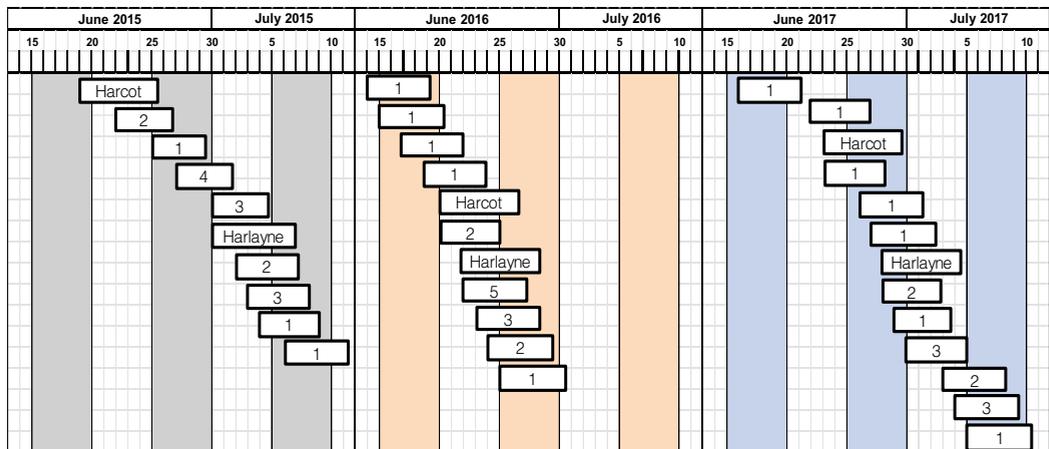


Figure 1. Fruit ripening date in 2015, 2016 and 2017  
Different digits show the number of hybrids ripened on that date

On figure 1 it is noticeable that most of the hybrids ripened later than 'Harcot' and 'Harlayne'. In our previous study with the crossbreed 'Modesto' x 'Harcot', the biggest group of hybrids were with intermediate ripening time (Bozhkova and Nesheva, 2016). In both cases, most of the hybrid fruits ripened later than 'Harcot' cv.

This might mean that 'Modesto' and 'Harlayne' later ripening time is the dominant trait.

These results are in accordance with Nyujtó and Banai (1986) proposition for who the late ripening period is dominant. Bassi and Negri (1991) assume it is under oligogenic control

and probably for that reason, there is such diversity in the hybrid family.

Table 1. Fruit size categories according to IBPGR descriptor

Fruit weight (g)	Fruit size (IBPGR)	Number of hybrids
< 20	Extremely small	0
20 - 30	Very small	0
31 - 40	Small	8
41 - 45	Small/medium	Harlayne + 3
46 - 55	Medium	3
56 - 60	Medium/large	Harcot + 2
61 - 70	Large	1
71 - 85	Very large	0
>85	Extremely large	0

Fruit weight is a trait of which depend the fruit quality and often the yield. According to the IBPGR descriptor, 47% of the fruits of the studied progeny were classified as small size (table 1). Attractive and medium-sized fruits are preferred by the producers and consumers and also are desired trait in apricot breeding (Guerriero et al.; 2005). Almost half of the studied hybrids (53 %) had small/medium to large sized fruits. Many well-known cultivars are classified as small to medium-sized fruits with an average weight up to 50-55 grams (Bozhkova and Todorova, 2012). Two of the hybrids had medium/large fruits as Harcot and one surpassed them with its large sized fruits. There is a high correlation between the main fruit physical characteristics - fruit height, fruit width, and thickness and all three of them are highly correlated with the fruit weight (Mratinic et al., 2011). Fruit biometry is important when the fruits are intended for processing, especially for their mechanical

sorting (Mohsenin, 1980). All three fruit dimensions depend on the cultivar.

After Duncan's multiple range test at  $P \leq 0.05$ , it can be said that: according to the data for all fruit dimensions - length (FL), width (FW) and thickness (FT) the groups are overlapping and there are hybrids with intermediate phenotype and hybrids that resemble the parental cultivars (table 2).

Although the clear trend that the fruits in the progeny are getting smaller in size a few hybrids were found to surpass both parental cultivars: by FL – HH 12-42 and HH 12-26, by FW – HH 12-19 and HH 12-42, by FT – HH 12-26, HH 12-42 and HH 12-19 but the statistical difference with 'Harcot' is non-significant. The biggest fruit weight was recorded for HH 12-42 which is close to the one measured for 'Harcot'. The larger fruits had stones with higher weight. However, HH 12-42, HH 12-26, and 'Harcot' had very good stone relative share- less than 6.

Table 2. Fruit biometric analysis

Genotype	Fruit Length	Fruit Width	Fruit Thickness	Avarage Fruit Weight	Stone weight	Relative share %
HH 13-3	45.03 bcdef	41.39 abcdef	45.36 abcde	48.43 abc	3.06 abcd	6.32 abcd
HH 13-54	43.26 cdef	37.93 cdef	43.64 abcdef	41.89 bcd	3.27 abc	8.10 abc
HH 13-43	41.13 def	37.98 cdef	39.51 ef	35.53 cd	2.27 de	6.36 abcd
HH 12-63	45.74 bcde	40.97 abcdef	45.34 abcde	47.27 abcd	3.07 abcd	6.51 abcd
HH 13-14	38.58 f	37.39 def	39.72 ef	34.60 cd	2.07 e	6.04 bcd
HH 12-26	52.80 a	41.63 abcd	47.72 abc	55.87 ab	2.90 bcde	5.23 d
HH 12-67	47.49 abcd	39.60 abcdef	45.36 abcde	46.49 abcd	2.80 bcde	6.04 bcd
HH 12-42	50.65 ab	45.74 a	49.08 ab	63.09 a	3.57 ab	5.59 cd
HH 12-19	48.08 abc	44.46 ab	49.83 a	59.02 ab	3.87 a	6.56 abcd
HH 13-4	41.06 def	34.54 f	38.58 ef	32.64 cd	2.77 bcde	8.58 ab
HH 13-15	41.24 def	38.44 abcdef	40.88 def	36.45 cd	2.28 de	6.42 abcd
HH 12-62	44.56 bcdef	38.77 abcdef	42.50 abcdef	42.12 bcd	2.77 bcde	6.85 abcd
HH 12-41	43.45 cdef	36.55 def	40.31 def	36.14 cd	2.46 cde	7.30 abcd
HH 12-9	49.63 abc	38.84 abcdef	42.02 cdef	44.40 bcd	2.69 cde	6.08 bcd
HH 12-59	38.46 f	38.40 abcdef	39.60 ef	37.29 cd	2.49 cde	7.14 abcd
HH 12-60	38.80 f	34.86 f	39.49 ef	30.22 d	2.63 cde	8.82 a
HH 12-22	38.99 ef	34.96 ef	38.34 f	32.73 cd	2.53 cde	8.56 ab
Harlayne	43.57 cdef	39.72 abcdef	43.30 abcdef	43.29 bcd	2.67 cde	6.23 abcd
Harcot	49.83 abc	44.12 abc	46.58 abcd	58.44 ab	3.04 abcd	5.19 d

\* Mean values followed by different letters within a column are significantly different by Duncan's multiple range test at  $P \leq 0.05$ .

Measuring the TSS content is a method that can quickly give us an idea of the fruit biological value. For 128 apricot cultivars cultivated in Malatya, Turkey TSS range is from 11-26.50 °Brix (Asma and Ozturk, 2005). For the

cultivars 'Berge cot', 'Flavor cot', 'Lady cot', 'Tom cot', 'Perle cot', 'Jenny cot' and 'Sweet cot' TSS content ranges from 13.40 to 23.30 °Brix (Bozhkova and Nesheva, 2016). Total Soluble Solids (TSS) content is important

especially for the dried apricot production. High TSS content is associated with high amount of sugars in the apricot fruits and increases their quality and the yield of dried product (Akin et al., 2008). TSS content grows with the fruit ripening, i.e. the highest value (Brix<sup>0</sup>) is found in fully ripened fruits (Xi et al., 2016). This chemical characteristic is strongly influenced by the environmental factors (Bartolini et al.; 2015).

In the present study the lowest TSS content was recorded for HH 12-42 (15.5 °Brix) and the highest - HH 12-22 (21.9 °Brix). Here the trend is reversed - TSS content increases in the progeny and 82% of the hybrids outperform both the parental cultivars (table 3).

The differences between ‘Harcot’ and all hybrids are non-significant. There is a statistically significant difference between the other parental cultivar ‘Harlayne’ and HH 12-22.

Table 3. Fruit appearance and taste qualities

Genotype	Total Soluble Solids (°Brix) (TSS)	Fruit ground color	Relative area of fruit over color (%)	Sensory score	Sensory evaluation
HH 13-3	20.07 abcd	med. orange	10-30	6.22	Good
HH 13-54	21.47 ab	med. orange	80	5.71	Good
HH 13-43	18.6 abcd	med. orange	40-60	5.90	Good
HH 12-63	18.8 abcd	light to med. orange	10	5.91	Good
HH 13-14	19.97 abcd	med. to dark orange	30-40	5.26	Good
HH 12-26	18.2 abcd	light to med. orange	50-60	7.67	Very Good
HH 12-67	19.83 abcd	light to med. orange	20-50	5.91	Good
HH 12-42	15.5 d	med. orange	40-60	6.66	Good
HH 12-19	20 abcd	med. orange	60-70	6.33	Good
HH 13-4	19.83 abcd	med. orange	40-50	5.82	Good
HH 13-15	21.00 abc	med. to dark orange	50	5.71	Good
HH 12-62	15.8 cd	med. to dark orange	30	5.53	Good
HH 12-41	16.77 abcd	med. orange	almost missing	5.11	Good
HH 12-9	18.33 abcd	light to med. orange	40-50	5.31	Good
HH 12-59	18.67 abcd	med. to dark orange	40-60	5.07	Good
HH 12-60	20.97 abc	light to med. orange	10-30	5.74	Good
HH 12-22	21.9 a	med. orange	40	5.93	Good
Harlayne	16.47 bcd	med. to dark orange	60	6.62	Good
Harcot	17.37 abcd	med. to dark orange	10	6.03	Good

Like most of the fruit characteristics, the ground color is genetically determined trait which expression is highly influenced by the environment. Consumers in our country prefer large fruits, with dark orange ground color and bright red over color (Bozhkova and Nesheva, 2016). All of the fruits of the studied hybrids were with orange ground color which shade varies from light to medium and from medium to dark orange. The red blush is the most attractive feature of apricots and the larger the area it occupies is the more seductive the fruits are. It has a great commercial impact and it is much-desired trait in the breeding programs

(Mazza and Miniati, 1993). The intensity and relative area of fruit over color strongly depend on the light, radiation, irrigation and nutrition of the trees. The relative area of fruit over color for the studied hybrids ranged from 10 to 80 %. More than half of them (65%) have an over color above 40% which gives them very attractive appearance. After sensory evaluation, all the 17 hybrids were highly scored (5.07-7.67).

Their taste was evaluated as good as both parental cultivars with already proven qualities. One of the hybrids HH 12-26 is evaluated as better than the others with very good taste and

score 7.67. Usually, well-informed consumers prefer fruits with good taste and when they are valued by sensory analyzes, taste and aroma are of greater importance (Bozhkova and Nesheva, 2016).

Krška et al., (2006b) prove that Harleyne's resistance is controlled by three independent complementary dominant genes, and after 10 years of research, Polak and Kominek (2012) report it as immune to 6 strains of *Plum pox virus*. In this breeding program, this cultivar was used as a donor of resistance. Harcot is partially resistant to PPV - resistant to strain PPV - D but it is susceptible to PPV - M (Rankovic et al., 1997). Its fruits are large with excellent taste and in this breeding program and much more is used as a donor of good fruit quality (Karayiannis, 2005). After genotyping within the work on project MARS (7 FP-Collaborative project nr. 613654) 'Harlayne' was found to be resistant. Partial resistance was detected in 'Harcot'. In the progeny in 47% of the hybrids were genotyped as resistant, 33% as partially resistant and 20% - sensitive (table 4).

Table 4. Resistance to PPV virus

Genotype	PPV resistance MAS	Phenotype/PPV symptoms 2017
HH 13-3	resistant	-
HH 13-54	Missing Data	-
HH 13-43	sensitive	+
HH 12-63	sensitive	-
HH 13-14	resistant	-
HH 12-26	Missing Data	+
HH 12-67	partially resistant	-
HH 12-42	resistant	-
HH 12-19	resistant	-
HH 13-4	resistant	-
HH 13-15	partially resistant	-
HH 12-62	partially resistant	-
HH 12-41	partially resistant	-
HH 12-9	partially resistant	-
HH 12-59	sensitive	+
HH 12-60	resistant	-
HH 12-22	resistant	-
Harlayne	resistant	-
Harcot	partially resistant	-

For two of the hybrids, the data is missing. Until 2017 symptoms of PPV were observed on

the stones and fruits of three of the hybrids. Two of them were genotyped as sensitive and for one of them the data is missing.

## CONCLUSIONS

As a result of the hybrid analyses, for their good fruit qualities and resistance to PPV, HH 12-42 and HH 12-19 were grafted on *P.cerasifera* rootstock and continue to the next stage of the breeding process. These two hybrids were the best ones in the studied progeny. The others also have good qualities and the work with them will continue to F2 for improving their disadvantages. HH 12-26 also has good fruit qualities but on the field, it showed symptoms of PPV. The fruit weight of the resistant genotypes - HH 13-3 and HH12-22 should be improved. Because of the great heterozygosity and the big number of traits under polygenic control, picking up two hybrids out of only 17 is a considerable success for the breeding program. Usually, such a result is obtained by observing hundreds of hybrids. Very high percent of the progeny is resistant or partially resistant to the PPV. That gives us a reason to believe that the crossbreed 'Harlayne' x 'Harcot' is very perspective for the apricot breeding programs.

## REFERENCES

- Abernathy D., Zhebentyayeva T., Abbott A.G., Vilanova S., Badenes M.L., Salava J., Polak J., Damsteegt V.D., Vilanova S., Badenes M.L., Krška B., 2004. Molecular genetic mapping of the Plum pox virus resistance genes in apricot. *Acta horticulturae*, 657, 283-288.
- Akin E.B., Karabulut I., Topcu A., 2008. Some compositional properties of main Malatya apricot (*Prunus armeniaca* L.) varieties. *Food Chemistry*, 107 (2), 939-948.
- Asma B. M., Ozturk K., 2005. Analysis of morphological, pomological and yield characteristics of some apricot germplasm in Turkey. *Genetic Resources and Crop Evolution*, 52 (3), 305-313.
- Bartolini S., Leccese A., Viti R., 2015. Quality and antioxidant properties of apricot fruits at ready-to-eat: influence of the weather conditions under Mediterranean coastal area. *J Food Process Technol*, 7 (538), 2.
- Bassi D., Negri P., 1991. Ripening date and fruit traits in apricot progenies. *Acta Hort* 293.12
- Audergon J.M., Clauzel G., Blanc A., Roch G., Lambert P., Ruiz D., Campoy J.A., Salazar J.A., Martínez-Gómez P., Egea J., Bureau S., Gouble B., Bogé M.,

- Reling P., Renard C.M.G.C., Dondini L., Tartarini S., 2011. Inheritance of phenological traits in apricot progenies. In XV International Symposium on Apricot Breeding and Culture 966, 27-35.
- Bozhkova V., Todorova L., 2012. Some results of apricot cultivars testing grown in the region of Plovdiv. Journal of Mountain Agriculture on the Balkans, vol. 15,1, 193-204.
- Bozhkova V., Nesheva M., 2016. Some results of evaluation of new-introduced apricot cultivars under conditions of Plovdiv region. Agricultural Science and Technology, 8 (3), 262-265.
- Dirlwanger E., Moing A., Rothan C., Svanella L., Pronier V., Guye A., Plomion C., Monet R., 1999. Mapping QTLs controlling fruit quality in peach (*Prunus persica* L. Batsch). TAG Theoretical and Applied Genetics, 98 (1), 18-31
- Guerriero R., Lomi F., D'Onofrio C., 2005. Influence of some agronomic and ecological factors on the constancy of expression of some descriptive characters included in the UPOV apricot descriptor list. In XIII International Symposium on Apricot Breeding and Culture 717 (pp. 51-54).
- Karayannis I., 2005. Progress in apricot breeding for resistance to sharka disease (Plum pox virus, PPV) in Greece. XIII International Symposium on Apricot Breeding and Culture 717.
- IBPGR, 1984: Revised descriptor list for apricot (*Prunus armeniaca* L.). Guerrero, R. & Watkins, R.(Eds) Publ. by International board for plant genetic resources, Commission of European communities: Committee on disease resistance breeding and use of genebanks, IBPGR Secretariat, Rome & CEC Secretariat, Brussels.
- Krška B., Vachůn Z., Nečas T., 2006a. The apricot breeding programme at the Horticulture Faculty in Lednice. Acta Horticulturae, 717, 145-148.
- Krška B., Salava J., Polák J., 2006b. Breeding for resistance: breeding for Plum pox virus resistant apricots (*Prunus armeniaca* L.) in the Czech Republic. EPPO Bulletin, 36 (2), 330-331.
- Mazza G., Miniati E., 1993. Anthocyanins in fruits, vegetables, and grains. Boca Raton, FL: CRC Press. p. 362.
- Milošević T., Milošević N., Glišić I., Krška B., 2010. Characteristics of promising apricot (*Prunus armeniaca* L.) genetic resources in Central Serbia based on blossoming period and fruit quality. Horticult. Sci 37: 46-55.
- Mohsenin N.N., 1980. Thermal properties of foods and agricultural materials. New York. USA.
- Mratinic E., Popovski B., Miloshevic T., Popovska M., 2011. Postharvest chemical, sensorial and physical-mechanical properties of wild apricot (*Prunus armeniaca* L.), Notulae Scientia Biologicae, 3(4), 105.
- Nyujtó F., Banai Mrs M., 1986. Informative remarks on our breeding experiences with apricots. Acta Horticult. 192, 307-312
- Polak J., Kominek P., 2012. Biological Evidence for Practical Immunity of Apricot Cultivar Harlayne to Plum Pox Virus. Plant Protection Science, 48(4).
- Rankovic M., Dulic-Markovic I., Paunovic S., 1997. Sharka virus in apricot and its diagnosis. In XI International Symposium on Apricot Culture 488, 783-786.
- Salazar J.A., Ruiz D., Egea J., Martínez-Gómez P., 2013. Transmission of fruit quality traits in apricot (*Prunus armeniaca* L.) and analysis of linked quantitative trait loci (QTLs) using simple sequence repeat (SSR) markers. Plant molecular biology reporter, 31(6), 1506-1517.
- SPSS S., 2010. 19.0. 0 for Windows Software. SPSS Inc., IBM Company, USA.
- Singh B.D., Singh A.K., 2015. Marker-assisted plant breeding: principles and practices. New Delhi, India: Springer, 259-293
- UPOV 2007. Guidelines for the Conduct of Tests for Distinctness, Uniformity and Stability, TG/70/4, Apricot (*Prunus armeniaca* L.)
- Velisek J., Cejpek K., 2007. Biosynthesis of food constituents: Vitamins. 1. Fat-soluble vitamins-a review. Czech Journal of Food Sciences-UZPI (Czech Republic).
- Xi W., Zheng H., Zhang Q., Li W., 2016. Profiling taste and aroma compound metabolism during apricot fruit development and ripening. International journal of molecular sciences, 17 (7), 998.