Fruit Growing Research, Vol. XXXV, 2019 DOI 10.33045/fgr.v35.2019.01
http://publications.icdp.ro/index.php

DIVERSITATEA GENETICĂ A SOIURILOR DE MAR CREATE IN ROMANIA ÎN ULTIMII 20 DE ANI

GENETIC DIVERSITY OF ROMANIAN APPLE CULTIVARS RELEASED IN THE LAST 20 YEARS

Militaru Mădălina[1], Sestraș Adriana[2*], Calinescu Mirela[1], Mareși Eugenia[1], Mihăescu Cristina[3]
[1]Research Institute for Fruit Growing Pitesti, Romania
[2]University of Agricultural Sciences and Veterinary Medicine Cluj-Napoca, Romania
[3]University of Pitesti, Faculty of Sciences, Romania
*Corresponding author: Sestraș Adriana[2] ; e-mail: asestras@gmail.com.

Abstract

An analysis of the pedigrees of apple cultivars developed in Romania in the last 20 years shows that the most are descended from 'Golden Delicious', 'McIntosh', 'Jonathan', 'Cox's Orange Pippin' and 'Red Delicious', called 'ancestors'. That means they have at least one of these apples in their family tree, as a parent, grandparent or great-grandparent. For those 41 apple cultivars registered in the last two decades an increased number of crosses with these 'ancestors' has led to what can only called 'inbreeding'. Thus, 'Bistritean' cv. (registered by Research Station for Fruit Growing Bistrita, 2002) has as ancestors 'McIntosh' (twice), 'Golden Delicious' and 'Red Delicious'; 'Precoce de Ardeal' cv. (registered by Research Station for Fruit Growing Cluj, 2005) has 'Golden Delicious' (three times), 'McIntosh' (twice) and 'Jonathan'; 'Luca' cv. (registered by Research Station for Fruit Growing Voinesti, 2006) has 'Golden Delicious' (twice), 'McIntosh' (twice) and 'Cox's Orange Pippin'; 'Rustic' cv. (registered by Research Institute for Fruit Growing Pitesti, 2008) has 'Golden Delicious' (twice), 'McIntosh' (twice) and 'Jonathan' (twice). Almost all cultivars studied are descendents from only five 'ancestors': 'Golden Delicious' (35 cultivars), 'McIntosh' (31 cultivars), 'Jonathan' (12 cultivars), 'Red Delicious' (6 cultivars). Coefficients of coancestry calculated were 0.105 for 'Golden Delicious', 0.051 for 'McIntosh', 0.046 for 'Jonathan', 0.039 for 'Red Delicious' and 0.003 for 'Cox's Orange Pippin'. This indicates that apple breeders has worked with populations of greatly reduced genetic diversity and this strategy becomes a problem because it leads to genetic impoverishment, and, also, the loss of the genetic resistance to scab because it was monogenic (mostly concentrated on *Vf* gene from *M. floribunda* clone 821).

Cuvinte cheie: ameliorare, consangvinizare, strămoși.
Key words: breeding, inbreeding, ancestors.

1. Introduction

Apple (*Malus* x *domestica* Borkh.) is one of the world's most ancient and widespread fruit crops in temperate regions. The cultivated apple is the result of a long evolutionary process extended over thousands of years. Several species have contributed to apple gene pool, but the wild Asian species *M. sieversii* M. Roem is considered the main contributor and the Tian Shan Mountains (Central Asia) the centre of origin. Hybridizations with other wild apple species along the Silk Route, such as *M. baccata* (L.) Borkh. in Siberia, *M. orientalis* Uglitz. in Caucasus and *M. silvestris* (L.) Miller in Europe, have produced the diversity of domestically apple (Cornille et al., 2014).

Today, more than 10,000 cultivars have been reported, but modern commercial apple production is dominated by only a few cultivars, and four of them, namely 'Golden Delicious', 'Gala', 'Red Delicious' and 'Idared', cover 48% of global production.

The most common used 'ancestor variety' for apple breeding is 'Golden Delicious' (347 times crossed into a total of 255 of the examined 500 varieties), followed by 'McIntosh' (252 times cross-bred into 174 varieties), 'Jonathan' (167 times crossed into 154 varieties), 'Cox's Orange' (157 times crossed into 150 varieties), 'Red Delicious' (95 crosses in 90 varieties) and 'James Grieve' (101 crosses in 75 varieties). 'McIntosh' and 'Red Delicious' dominate American breeding programmes, and 'McIntosh' has a central role in developing columnar varieties. 'Cox's Orange Pippin' and 'James Grieve' are more commonly used in the European breeding programmes (Noiton and Alspach, 1996; Bannier, 2011).

During the last 20 years, in Romania, breeding objectives have mainly focused on eating quality and disease resistance. The apple breeding programs for resistance to scab (*Venturia inaequalis* Cke.) have mostly concentrated on *Vf* gene, originated from a cross between two selections of *M. floribunda* 821 x 'Rome Beauty', which carried the *Vf* gene (Crosby et al., 1992). Unfortunately, many other apple breeding programs have been using the same parents.

This study attempts to measure genetic diversity presently use in Romanian apple breeding. Pedigrees available in the literature and breeding records were used to study the genetic contributions of 'ancestor variety' for those 41 apple cultivars registered in the last 20 years.

2. Material and methods

Pedigrees of apple cultivars registered in Romania in the last 20 years were collected from breeding records and published sources (Braniste et al., 2007, Stefan et al., 2018) (Table 1). All open pollinated were assumed to be non-inbred and unrelated to the pollen parents, due to apple self-incompatibility. The degree of relationship of those cultivars with the five 'ancestor' ('Golden Delicious', 'McIntosh', 'Jonathan', 'Cox's Orange' and 'Red Delicious') was investigated by calculation of the individual coefficient of coancestry of each of them with the 41 Romanian cultivars. The degree of relationship by descent of the two parents is their coefficient of co ancestry, f, which is identical with the inbreeding coefficient, F, of their progeny (Alspach, 1976). All parents were treated as diploid and parents with unknown origin were assumed to be noninbred. Apples are mostly self-incompatible, and it was assumed that cultivars without known pedigree originated from outcrossed open-pollination, under estimating possible inbreeding. All mutants were regarded as the same as the original cultivar. Since only few genes are expected to be different between such mutants and the original, this simplification can lead to minor overestimation of inbreeding coefficients. Allelic contributions from both parents were assumed to be equal and unaltered by breeders' selection (Noiton and Alspach, 1996).

3. Results and discussions

An increase of inbreeding

For the apple cultivars registered in Romania in the last two decades an increase number of crosses of five ('Golden Delicious', 'McIntosh', 'Jonathan', 'Cox's Orange Pippin', 'Red Delicious') from six 'ancestors' has led to what can only called 'inbreeding'. Thus, 'Bistritean' cv. (registered by Research Station for Fruit Growing Bistrita, 2002) has as ancestors in its family tree 'McIntosh' (twice), 'Golden Delicious' and 'Red Delicious'; 'Rustic' cv. (registered by Research Institute for Fruit Growing Pitesti, 2008) has 'Golden Delicious' (twice), 'McIntosh' (twice) and 'Jonathan' (twice) - Figure 1; 'Precoce de Ardeal' cv. (registered by Research Station for Fruit Growing Cluj, 2005) has 'Golden Delicious' (three times), 'McIntosh' (twice) and 'Jonathan' (Figure 2); 'Luca' cv. (registered by Research Station for Fruit Growing Voinesti, 2006) has 'Golden Delicious' (twice), 'McIntosh' (twice) and 'Cox's Orange Pippin'.

Almost all 41 cultivars studied were found to be descended from only five 'ancestors': 'Golden Delicious' (35 cultivars), 'McIntosh' (31 cultivars), 'Jonathan' (12 cultivars), 'Red Delicious' (6 cultivars). Among these, 32 cultivars had two or more 'ancestors' in their parentage (Table 1). These results support Brown's concern (1973) about the trend in excessive use 'Golden Delicious', 'McIntosh', 'Jonathan' and 'Red Delicious' as parents. The problem of restricted number of 'ancestors' in apple breeding is common to many fruit crops, such as raspberry (Dale et al., 1993), blueberry (Hancock and Siefker, 1982) and peach (Scorza et al., 1988). The predominance of only five founding clones in modern apple cultivars may be explained by the lack of information on the breeding value of apple germplasm, which make breeders to use untested parents. Cultivars such as 'Golden Delicious', 'McIntosh', 'Jonathan', 'Red Delicious' and 'Cox's Orange Pippin' have been reported to be generally valuable parents (Davis et al., 1954; Son et al., 2012). The traits which make the breeders to use the 'ancestors' varieties, especially 'Golden Delicious', as genitors for so long time are: regular and high-flower set, with some fruit on new wood even if incorrectly pruned, biennial tendency, moderate to low growth habit, uniform fruit shape and size, attractive fruit skin colour, sweet and aromatic taste, good shelf life, less preharvest fruit fall, good fruit firmness for transportation (Bannier, 2011).

Coancestry of apples

The mean coefficients of co ancestry of 41 apple cultivars included in this study are 0.105 with 'Golden Delicious', 0.051 with 'McIntosh', 0.046 with 'Jonathan', 0.003 with 'Cox's Orange Pippin' and 0.039 with 'Red Delicious' (Table 2). Coefficients of coancestry ranged between 0 to 0.250, and high levels of coancestry found indicate that further use of those five 'ancestors' or their descendents will increase the risk of inbreeding in future generations. Our results are comparable with coefficients of coancestry of the 77 modern apple cultivars: 0.101 with "Golden Delicious", 0.044 with "McIntosh", 0.055 with "Jonathan", 0.022 with "Cox's Orange Pippin" and 0.058 with "Red Delicious" (Noiton and Alspach, 1996).

Genetic narrowing in cultivars with monogenic scab-resistance

The *Vf* resistance from *M. floribunda* 821 is convenient to use in breeding because the gene can be identified using molecular markers, but it is quite unlike the polygenic resistance of older varieties. In Romanian breeding program, the most use *Vf* resistance source in different cross combination (as mother or father) were the following cultivars: 'Prima' (in 26 crosses), 'Florina' (in 4 crosses) and 'Pionier' (in 3 crosses). The inbreeding coefficients for these *Vf* cultivars range from 0 for 'Florina' and 'Pionier' to 0.031 for 'Prima' (Table 3). Coefficients of coancestry of *Vf*-carrier group are 0.083 with 'Golden Delicious', 0.031 with 'McIntosh', 0.375 with 'Jonathan', 0.041 with 'Red Delicious' (Noiton and Alspach, 1996). These results indicate that pedigrees should be carefully examined before selecting parents. In addition, report of a new race of apple scab virulent to all *Vf* gene cultivars reinforces the need to introduce new sources of scab resistance into breeding programs (Crosby et al.,1992; Militaru et al., 2017).

4. Conclusions

The massive use of limited cultivars in Romanian breeding program has dramatically reduced apple genetic diversity. The use of dominant 'ancestors' has introduced some vitality problems into bred Romanian cultivars. A severe problem of inbred apple cultivars is their susceptibility to apple scab, because the single-gene *Vf*-resistance has been overcome by fungal mutation.

References

1. Alspach P.A., 1976. Computation of inbreeding coefficients from stored pedigree data using Malecot's method. MS thesis, Univ. of York, England.
2. Bannier H. J., 2011. Inbreeding in modern apple cultivation. Erwerbs-Obstbau 52(3-4): 85-110, DOI: 10.1007/s10341-010-0113-4.
3. Braniste N., Budan S., Butac M., Militaru M., 2007. Soiuri de pomi, arbuști fructiferi și căpșuni create în România, Ed. Paralela 45, pp. 51-103.
4. Brown A.G., 1973. The effect of inbreeding on vigour and length of juvenile period in apples, Proc. Eucarpia fruit section Symp. V. Top fruit breeding, Canterbury, England, 11-14 sept. 1973, pp. 30-39.
5. Cornille A., Giraud T., Smulders M. J., Roldan-Ruiz I., Gladieux P., 2014. The domestication and evolutionary ecology of apples. Trends Genet. 30, pp. 57–65.
6. Crosby J.A., Janick J., Pecknold P.C., Korban S.S., O'Connor P.A., Ries S.M., Goffreda J., Voordeckers A., 1992. Breeding apples for scab resistance: 1945-1990. Fruit Varieties Journal 46(3), pp. 145-166.
7. Dale A., Moore P.P., McNicol R.J., Sjulin T.M., Burmistrov L.A., 1993. Genetic diversity of red raspberry varieties throughout the world, Journal of the American Society for Horticultural Science 118(1), pp. 119-129.
8. Davis M.B., Blair D.S., Spangelo L.P.S., 1954. Apple breeding at the Central Experimental Farm, Ottawa, Canada, 1920-1951. Proc. Amer. Soc. Hort. Sci. 62, pp. 243-250.
9. Hancock J.F., Siefker J.H., 1982. Levels of inbreeding in highbush blueberry cultivars. HortScience 17, pp. 363-366.
10. Militaru M., Calinescu M.F., Butac M., 2017. Emerge of virulences on agrosystems: *Venturia inaequalis* at *Vf* apple cultivars grown in Romania as a case study. Fruit Growing Research, Vol. XXXIII, pp. 60-66.
11. Noiton D.A.M., Alspach P.A., 1996. Founding clones, inbreeding, coancestry and status number of modern apple cultivars, Journal of the American Society for Horticultural Science 121(5), pp. 773-782.
12. Scorza R., Sherman W.B., Lightner, 1988. Inbreeding and coancestry of low short fruit development period freestone peaches and nectarines produced by the University of Florida breeding program. Fruit Var. Journal 42, pp. 79-85.
13. Stefan N. and colab., 2018, Romania's Pomology, Ed. Ceres, pp. 47-193.
14. Son K.M., Kwon S. I., Choi C., 2012. Inbreeding, coancestry and founding clones of apple cultivars released from Korea. Horticulture, Environment, and Biotechnology, 53(5), pp. 404-409.

Tables and figures

Table 1. Parentage of apple cultivars released in Romania in the last 20 years

No.	Cultivar	Reported parentage	Year released
1	Romus 4	Romus 3 x Prima	1999
2	Aura	Prima x BN 33/39[a]	1999
3	Salva	Golden Delicious x Prima	2002
4	Starkprim	Starkrimson[b] x Prima	2000
5	Jonaprim	Jonathan x Prima	2000
6	Bistriţean	Starkrimson x Prima	2002
7	Goldprim	Golden Delicious x Prima	2003
8	Romus 5	Romus 3 x Prima	2003
9	Rebra	Prima x Florina	2003
10	Dany	Jonathan[c] irradiated (P32) x Prima	2005
11	Doina	Jonathan x Prima	2005
12	Redix	Goldspur x gamma radiation (2000 ϒ) of Prima polen	2004
13	Auriu de Cluj	CjX-5-52 [(III-VI-5-26 x NJ 46) x (Parmain d'or o.p.)] x Mutsu	2005
14	Estival	NJR 55 x Sir Prize	2005
15	Precoce de Ardeal	X-5-71 (Feleac x Sir Prize) x Prima	2005
16	Productiv de Cluj	Cluj III-VI-5-26 (Parmain d'or o.p.) x NJ 46 [a]	2005
17	Iris	gamma radiation (8000 ϒ) of Prima seeds (o.p.)	2005
18	Alex	Golden Delicious x BN 33/39[a]	2005
19	Nicol	Wijcik[d] X Pionier	2005
20	Voinicel	gamma radiation (5000 ϒ) of Prima seeds (o.p.)	2009
21	Luca	Champion[e] x Prima	2006
22	Irisem	gamma radiation (5000 ϒ) of Prima seeds (o.p.)	2006
23	Colmar	Wijcik[d] x Florina	2006
24	Colonade	Pionier x Wijcik[d]	2007
25	Real	gamma radiation (8000 ϒ) of Prima seeds (o.p.)	2007
26	Saruman	Cluj III-VI-5-26 (Parmain d'or - o.p.) x NJ 46 [a]	2007
27	Sauron	Cluj 3/83 [III-VI-5-26 (Parmain d'or o.p) x NJ 46 [a]] o.p.	2007
28	Chindia	Prima x Discovery[f]	2008
29	Discoprim	Prima x Discovery[f]	2009
30	Pomona	Starkrimson[b]x Prima	2008
31	Remar	gamma radiation (5000 ϒ) of Prima seeds (o.p.)	2008
32	Rustic	Pionier x Florina	2008
33	Dacian	Incomplete parentage available	2009
34	Inedit	gamma radiation (5000 ϒ) of Prima seeds (o.p)	2009
35	Silvan	White Transparent x Richared[g]	2010
36	Someşan	White Transparent x Richared[g]	2010
37	Ştefano	London Pepping (o.p.)	2010
38	Andrei	Prima x Generos	2012
39	Viorin	Prima x Generos	2012
40	Cezar	Prima x Starkrimson[b]	2016
41	Revidar	Prima (o.p.)	2016
42	Valery	Goldspur[h] x Florina	2016
43	Amalia	Incomplete parentage available	2018

[a] Unknown parentage;
[b]Starkrimson = sport of “Red Delicious“;
[c] irradiated with P32;
[d]Wijcik = sport of “McIntosh“;
[e]Champion = Golden Delicious x Cox's Orange Pippin;
[f]Discovery = Worcester Pearmain x Beauty of Bath;
[g]Richared = sport of “Red Delicious“;
[h]Goldspur = sport of “Golden Delicious“;
o.p. = open-pollinated.

Table 2. Coancestry coefficients with 'Golden Delicious', 'McIntosh', 'Jonathan', 'Cox's Orange Pippin' and 'Red Delicious' of 41 Romanian apple cultivars

Cultivar	Coefficient of coancestry with				
	Golden Delicious	McIntosh	Jonathan	Cox's Orange Pippin	Red Delicious
Romus 4	0.125	0.063	0.000	0.000	0.000
Aura	0.125	0.063	0.000	0.000	0.000
Salva	0.031	0.063	0.000	0.000	0.000
Starkprim	0.125	0.063	0.000	0.000	0.250
Jonaprim	0.125	0.063	0.250	0.000	0.000
Bistriţean	0.125	0.063	0.000	0.000	0.250
Goldprim	0.031	0.063	0.000	0.000	0.000
Romus 5	0.125	0.063	0.000	0.000	0.000
Rebra	0.003	0.001	0.125	0.000	0.000
Dany	0.125	0.063	0.000	0.000	0.000
Doina	0.125	0.063	0.250	0.000	0.000
Redix	0.000	0.000	0.000	0.000	0.000
Auriu de Cluj	0.000	0.000	0.000	0.000	0.000
Estival	0.375	0.000	0.000	0.000	0.000
Precoce de Ardeal	0.250	0.063	0.250	0.000	0.000
Productiv de Cluj	0.000	0.000	0.000	0.000	0.000
Iris	0.125	0.063	0.000	0.000	0.000
Alex	0.250	0.000	0.000	0.000	0.000
Nicol	0.063	0.031	0.125	0.000	0.000
Voinicel	0.125	0.063	0.000	0.000	0.000
Luca	0.250	0.063	0.000	0.125	0.000
Irisem	0.125	0.063	0.000	0.000	0.000
Colmar	0.063	0.250	0.250	0.000	0.000
Colonade	0.063	0.250	0.125	0.000	0.000
Real	0.125	0.063	0.000	0.000	0.000
Saruman	0.000	0.000	0.000	0.000	0.000
Sauron	0.000	0.000	0.000	0.000	0.000
Chindia	0.125	0.063	0.000	0.000	0.000
Discoprim	0.125	0.063	0.000	0.000	0.000
Pomona	0.125	0.063	0.000	0.000	0.250
Remar	0.125	0.063	0.000	0.000	0.000
Rustic	0.063	0.000	0.031	0.000	0.000
Inedit	0.125	0.063	0.000	0.000	0.000
Silvan	0.000	0.000	0.000	0.000	0.250
Someşan	0.000	0.000	0.000	0.000	0.250
Ştefano	0.000	0.000	0.000	0.000	0.000
Andrei	0.125	0.063	0.125	0.000	0.000
Viorin	0.125	0.063	0.125	0.000	0.000
Cezar	0.125	0.063	0.000	0.000	0.250
Revidar	0.125	0.063	0.000	0.000	0.000
Valery	0.250	0.000	0.250	0.000	0.125
Average	**0.105**	**0.051**	**0.046**	**0.003**	**0.039**

Table 3. Information about *Vf* cultivars used as genitors in Romanian breeding program (source Noiton and Alspach, 1996)

Cultivar	Origin / Year	Reported parentage	Inbreeding coefficients	Coefficient of coancestry with			
				Golden Delicious	McIntosh	Jonathan	Red Delicious
Prima	USA, 1970	(Golden Delicious x F2 26829-2-2[a]) x [(Melba[b] x (Wealthy x Star)] x (Red Rome Beauty[c] x Melba[b])	0.031	0.125	0.063	0.000	0.000
Florina	France, 1977	(Jonathan x (Starking Delicious[d] x (Golden Delicious x F2 26829-2-2[a]))	0.000	0.063	0.000	0.250	0.125
Pionier	Romania, 1982	(Verzisoare[e] x Jonathan) x Prima	0.000	0.063	0.031	0.125	0.000
Mean	**x**	**x**	**0.010**	**0.083**	**0.031**	**0.375**	**0.041**

[a] F2 26829-2-2 = (Rome Beauty x *M. floribunda* 821) x (Rome Beauty x *M. floribunda* 821);
[b]Melba = McIntosh open-pollinated;
[c]Red Rome Beauty = sport of Rome Beauty;
[d]Starking Delicious = sport of Red Delicious;
[e]Verzisoare = Romanian local variety.

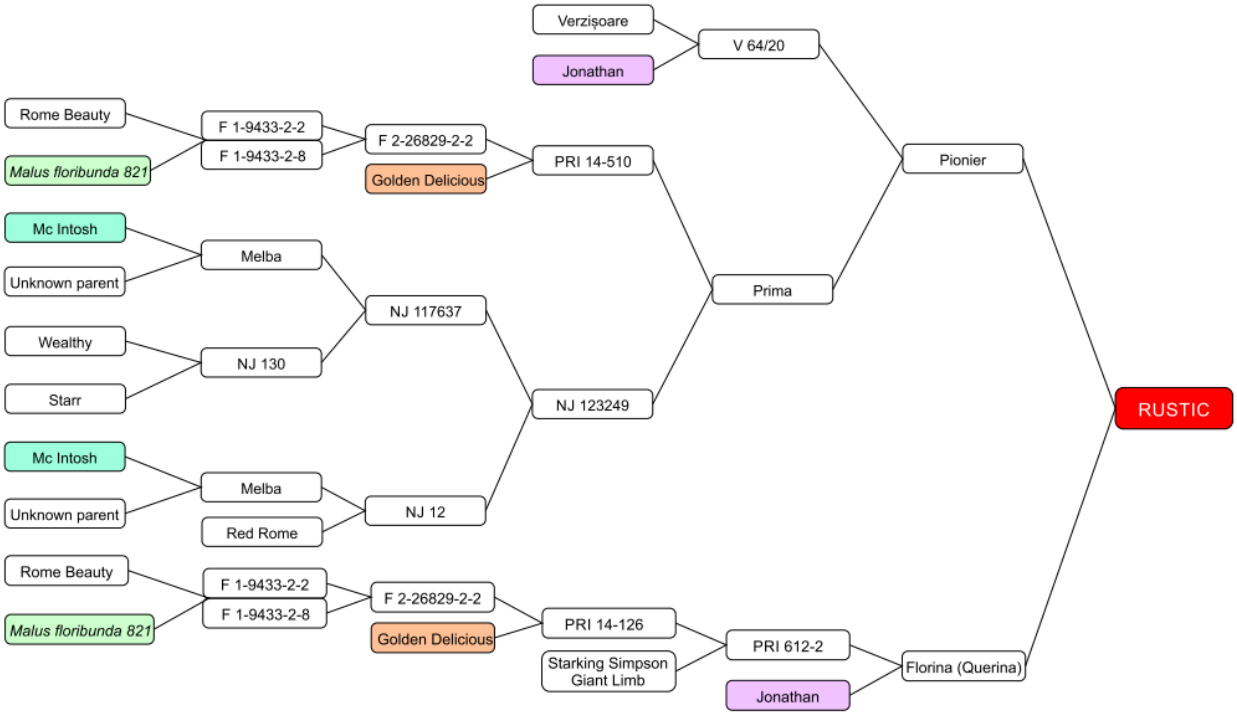

Fig. 1. 'Rustic' cv. with gene accumulation from three 'ancestors': twice 'Golden Delicious', twice 'McIntosh', twice 'Jonathan'

DOI 10.33045/fgr.v35.2019.01

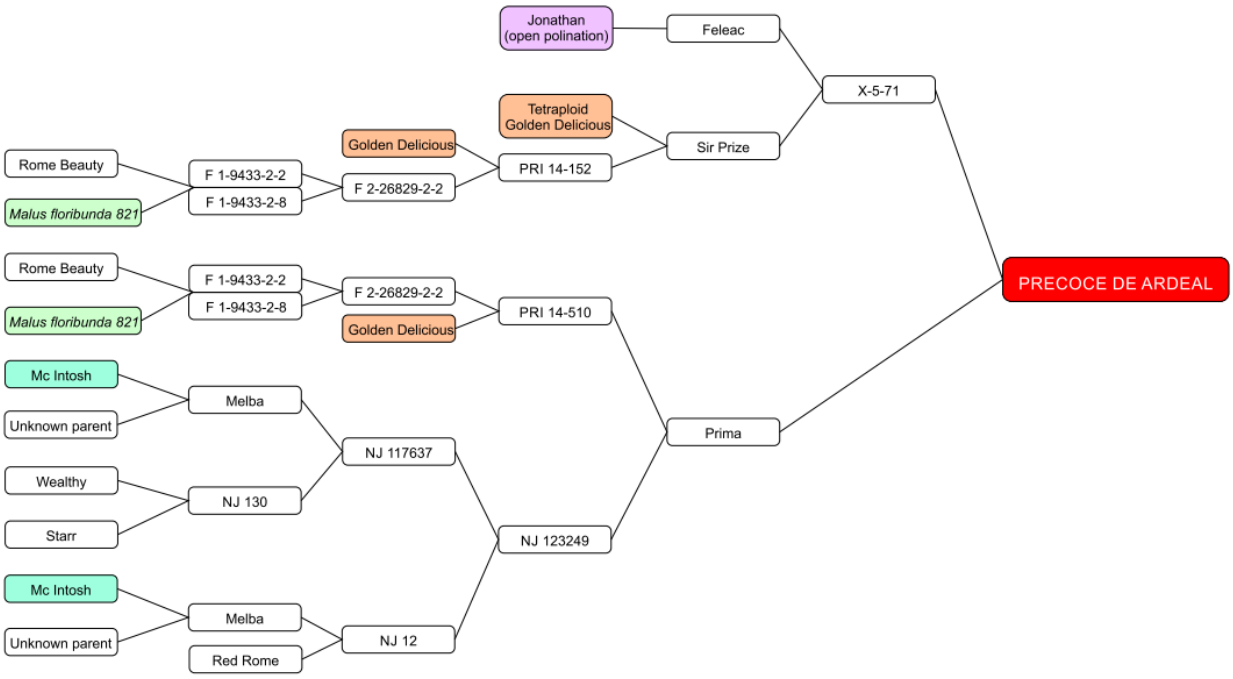

Fig. 2. ‘Precoce de Ardeal’ cv. with gene accumulation from three 'ancestors': three times ‘Golden Delicious’, twice ‘McIntosh’, once ‘Jonathan’