

Horticultural Evaluation of New *Citrus latipes* Hybrids as Rootstocks for Citrus

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Abstract. In 1968, the CRA-Research Center for Citriculture and Mediterranean Crops (CRA-ACM) started a research program aimed at breeding citrus rootstocks. The monoembryonic species *C. latipes* (Swing.) Tan. was used as the female parent; trifoliolate orange [*Poncirus trifoliata* (L.) Raf.], sour orange, and volkamer lemon (*C. volkameriana* Pasq.) were used as male parents. The behavior of some of these hybrids tested with other standard rootstocks in Sicily and Sardinia was evaluated. The cultivars under comparison included ‘Washington’ navel orange and ‘SRA 92’ clementine in Sardinia and ‘Tarocco’ orange in Sicily. Our results showed the dramatic influence of rootstock on plant growth and yield; only minor effects on fruit quality were observed. Among the standard rootstocks tested, Swingle citrumelo provided the highest yield. Some of the tested hybrids (F5 P12, F6 P12, and F6 P13) may improve plant yield, thus maintaining good fruit quality. Encouraging data obtained with these hybrids may justify the use of monoembryonic species of the *Papeda* subgenus for breeding citrus rootstocks.

As a result of its excellent agronomic qualities, sour orange continues to be used for citrus in the regions where citrus tristeza virus (CTV) is rare or absent. In Italy, sour orange was the most widely used rootstock until recently. However, the recent discovery of CTV infections in some citrus groves made it necessary to exchange sour orange for a rootstock tolerant to the virus (Caruso et al., 2003; Zurrú et al., 2004). Soil sickness and insufficient adaptability to particular environments and/or cultivars reinforced the need for alternative rootstocks to replace sour orange.

Using controlled hybrids as candidate rootstocks is possible if the hybrids meet the following conditions: 1) selection propagation through nucellar embryony; 2) tolerance or resistance to primary biotic and abiotic stresses; 3) grafting compatibility with most citrus cultivars; and 4) adaptability to different pedoclimatic conditions.

Long periods of research are required to verify the presence of these characteristics. Rootstock alternatives to sour orange include ‘Troyer’ and ‘Carrizo’ citranges yielded by a cross performed in 1909 (Webber, 1948); the rootstock C 35 selected in 1951 (Cameron

and Soost, 1986); and the citrumelo ‘Swingle’ that resulted from a cross performed in 1907 (Hutchison, 1974; Wutsher, 1974). The widespread use of seed propagation for citrus rootstock led to neglected consideration of the monoembryonic species, particularly those of the subgenus *Papeda*, as well as rootstocks as parents in breeding.

In 1968, the CRA-Research Center for Citriculture and Mediterranean Crops (CRA-ACM) started a research program aimed at breeding citrus rootstocks. The monoembryonic species *C. latipes* (Swing.) Tan. was used as the female parent; trifoliolate orange [*Poncirus trifoliata* (L.) Raf.], sour orange, and volkamer lemon (*C. volkameriana* Pasq.) were used as male parents. In previous observations, *C. latipes* showed high vigor and tolerance to “mal secco.” Progenies were planted in the CRA-ACM experimental fields located at Fonti Ciane in Siracusa (Sicily, Italy). Seedling selection was performed for nucellar embryony, plant vigor, tolerance to “mal secco,” and tolerance to *Phytophthora* spp. (De Simone et al., 1998; Reforgiato Recupero et al., 1997; Reforgiato Recupero and Russo, 1992).

In this article, we report on the behavior of some of these hybrids tested with other citrus rootstocks in Sicily and Sardinia. The cultivars grafted on the rootstock under comparison included ‘Washington’ navel orange and ‘SRA 92’ clementine in Sardinia and ‘Tarocco’ orange in Sicily.

The rootstocks used in the three tests are listed in Table 1. The *C. latipes* × trifoliolate orange genotypes (labeled as 68-IG) used in these tests were selected from a population of 83 hybrids obtained by cross in 1968. Genotypes of *C. latipes* × sour orange (labeled as LTXAM) were selected from a population of 257 hybrids obtained in 1969. All the tested hybrids were selected for their potential to generate nucellar embryos. In a previous paper (Reforgiato Recupero and Russo, 1992) is reported the percent of hybrids having high polyembryony in the various crosses. The seeds used to generate seedlings for the experiment were harvested from plants growing in the Fonti Ciane experimental field at the CRA-ACM.

The orange cultivar Tarocco TDV was used in the experiment carried out in Sicily. Seedlings grafted in the greenhouse were planted the next year (May 1998) on medium-texture soil in the experimental orchard of the CRA-ACM located at Palazzelli (PA) (Sicily, Italy). The previous crop was orange grafted on sour orange rootstock. After the orchard was uprooted, the soil was left uncultivated for 2 years.

‘Tarocco TDV’ is a nucellar selection obtained in 1976 from a plant exhibiting juice vesicle degeneration (Reforgiato Recupero and Russo, 1976–1977). The nucellar selection does not present this unfavorable characteristic, probably because the mutation was chimerical and present only in the first layer (L1). The defining characteristics of this ‘Tarocco’ selection are: orange-colored peel with a light anthocyanic pigmentation; fruit of ovoid shape; equatorial mean diameter of 77 mm and longitudinal mean diameter of 79 mm; fruit mean weight of 240 g; deep red pulp color with a high anthocyan content; and early or midrange ripening time. Blood orange cultivars are the mainstay of Italian orange production. The red color of fruit is an important factor influencing consumer appeal and marketability of both fruit and juices. In addition, the antioxidant activity of anthocyanins promotes health when they are consumed in the diet, including prevention of cancer, inflammation, and arteriosclerosis.

For the first test carried out in Sardinia, we used a ‘Washington’ navel nucellar selection introduced from Riverside, CA, in the 1960s. The ‘SRA 92’ clementine selection was used for the second test. This cultivar was introduced from the INRA-CIRAD (Center INRA de Corse, San Giuliano, Italy) in the late 1970s. Seedlings grafted in the spring of the previous year were planted in 1996 on an experimental orchard at the AGRIS Sardegna (Department of Wood and Fruit Tree Research) in Uta (UT) (southern Sardinia, Italy).

The same planting distances were used in all experiments (6 × 4 m) following a completely randomized block design with 15 replications and a single tree plot for ‘Tarocco TDV’, 11 for ‘Washington’ navel, and 10 for ‘SRA 92’.

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Table 1. List of common, botanical names and code of rootstocks tested for 'Tarocco' orange at Palazzelli and for 'Washington' navel orange and 'SRA 92' clementine at UTA (CA).

Common name	Latin name	Code
68 IG 26-C1 F1-P6 ^z	<i>C. latipes</i> (Swing.) Tan. × <i>Poncirus trifoliata</i> (L.) Raf.	F1 P6
68 IG 26-C1 F2-P12 ^z	<i>C. latipes</i> × <i>P. trifoliata</i>	F2 P12
68 IG 26-C1 F3-P7 ^z	<i>C. latipes</i> × <i>P. trifoliata</i>	F3 P7
68 IG 26-C1 F4-P2 ^y	<i>C. latipes</i> × <i>P. trifoliata</i>	F4 P2
68 IG 26-C1 F4-P6 ^{z,y,x}	<i>C. latipes</i> × <i>P. trifoliata</i>	F4 P6
68 IG 26-C1 F5-P12 ^{z,y,x}	<i>C. latipes</i> × <i>P. trifoliata</i>	F5 P12
68 IG 26-C1 F5-P3 ^z	<i>C. latipes</i> × <i>P. trifoliata</i>	F 5 P3
68 IG 26-C1 F6-P2 ^z	<i>C. latipes</i> × <i>P. trifoliata</i>	F6 P2
68 IG 26-C1 F6-P12 ^z	<i>C. latipes</i> × <i>P. trifoliata</i>	F6 P12
68 IG 26-C1 F6-P13 ^{z,y,x}	<i>C. latipes</i> × <i>P. trifoliata</i>	F6 P13
68 IG 26-C1 1 F6-P17 ^z	<i>C. latipes</i> × <i>P. trifoliata</i>	F6 P17
68 IG 26-C1 1 F6-P20 ^x	<i>C. latipes</i> × <i>P. trifoliata</i>	F 6P20
69 LTXAM-C1 F8-P3 ^x	<i>C. latipes</i> × <i>C. aurantium</i> L.	F8 P3
69 LTXAM-C1 F13-P23 ^{z,y}	<i>C. latipes</i> × <i>C. aurantium</i>	F13 P23
69 LTXAM-C1 F14-P37 ^z	<i>C. latipes</i> × <i>C. aurantium</i>	F14 P37
Sour orange ^{z,y,x}	<i>C. aurantium</i>	SO
Christian trifoliolate orange ^z	<i>P. trifoliata</i>	CPT
Large Flowered trifoliolate orange ^z	<i>P. trifoliata</i>	LPT
Swingle citrumelo ^{z,y,x}	<i>C. paradisi</i> Macf. × <i>P. trifoliata</i>	SC
Carrizo citrange ^{z,y,x}	<i>C. sinensis</i> × <i>P. trifoliata</i>	CC
Troyer citrange ^z	<i>C. sinensis</i> × <i>P. trifoliata</i>	TC
Cleopatra mandarin × trifoliolate orange 30573 ^z	<i>C. reshni</i> Hort. ex Tan. × <i>P. trifoliata</i>	CLEHY

Rootstocks used for 'Tarocco' orange^z, 'Washington' navel^y, and 'SRA 92' clementine^x.

Table 2. Yield, tree size, and efficiency of 'Tarocco' TDV orange on 19 rootstocks planted at Palazzelli (SR).

Rootstocks	Cumulative yield (2003–2008) (kg/tree)		Canopy volume (m ³)		Yield efficiency (kg·m ⁻³)	
F1 P6	82.9	a ^z	9.9	ab	9.5	a-e
F5 P3	96.2	ab	19.7	c-e	4.6	a
F6 P2	115.2	a-c	13.7	a-c	9.0	a-e
F3P7	119.6	a-c	13.5	a-c	9.1	a-e
CLEHY	123.8	a-c	7.4	a	17.6	f
F6 P17	133.5	a-c	22.4	d-f	6.0	ab
TC	145.0	a-d	15.6	b-d	9.8	b-e
F4 P6	147.3	b-d	15.6	b-d	11.2	c-e
SO	147.5	b-d	19.1	c-e	8.2	a-d
LPT	151.2	b-d	15.6	b-d	10.7	b-e
F13 P23	154.1	b-d	24.3	ef	6.7	a-c
CPT	166.7	cd	16.9	b-d	10.0	b-e
F2 P12	169.3	c-e	13.4	a-c	13.7	ef
CC	172.6	c-e	14.1	a-c	13.1	d-f
F14 P37	176.6	c-e	29.2	f	6.2	a-c
F6 P13	202.7	d-f	24.5	ef	8.8	a-e
F5 P12	230.6	ef	24.4	ef	10.3	b-e
F6 P12	231.3	ef	21.3	de	12.1	de
SC	265.7	f	21.5	de	12.6	d-f

^zMean separation within columns by Tukey's multiple range test at $P \leq 0.05$.

Table 3. Yield, tree size, and efficiency of 'Washington' navel Frost orange on eight rootstocks planted at UTA (CA).

Rootstocks	Cumulative yield (1998–2007) (kg/tree)		Canopy volume (m ³)		Yield efficiency (kg·m ⁻³)	
SO	137.8	a ^z	15.7	ab	8.7	a
CC	141.8	a	14.2	a	10.1	ab
F4 P6	182.5	ab	13.5	a	13.3	bc
F4 P2	219.0	bc	14.5	a	15.2	c
F13 P23	227.1	bc	19.4	bc	12.4	a-c
SC	236.7	bc	19.9	bc	12.1	a-c
F6 P13	258.8	c	21.9	c	12.0	a-c
F5 P12	278.3	c	28.7	d	9.7	ab

^zMean separation within columns by Tukey's multiple range test at $P \leq 0.05$.

At PA, the soil was a sandy-loam (62% sand, 20% silt, 18% clay), 8.0% calcium carbonate, and 4.5% active calcium carbonate; pH was 8.6. At UT, the soil was a sandy-loam (68% sand, 15% silt, 17% clay) with traces of calcium carbonate; pH was 6.8.

Standard cultural practices were used for all trials with mechanical weed control between rows and chemical control between trees. In the PA field, drip irrigation was provided by four suppliers (functioning at 4 L/h) per tree. In the UT field, irrigation was

provided by one microjet supplier (functioning at 80 L/h) per tree.

The cumulative yield from the 'Tarocco TDV' trees was measured over 6 years (2003 to 2008) (Table 2); the cumulative yield from the 'Washington' navel (Table 3) and 'SRA 92' trees (Table 4) was recorded over 10 years (1998 to 2007). Yield effectiveness of the canopy was calculated as the ratio of cumulative yield to canopy volume for the last year, calculated as $V = 0.5238 \times h \times d^2$ (h = plant height; d = plant lateral mean diameter).

To measure the yield, the crop of each tree was harvested and weighed. Fruit quality parameters were determined as the mean of 4 years (Tables 5–7). Samples were obtained during the third week of January for 'Tarocco TDV' and 'Washington' navel and during the second week of December for the 'SRA 92' clementine. After the harvest, 20 fruit samples were obtained from each tree and then weighed. Fruit diameter was measured and then the fruit was halved and rind thickness was determined with a digital caliper. Juice was extracted with an electric squeezer. Total soluble solids (TSS) were determined by an Atago digital temperature–refractometer, Model DBX-55 (Atago Co., LTD, Tokyo, Japan). Total acids were determined by titration with 0.1 N NaOH. Peel color index was determined before cutting the fruits with a Minolta colorimeter model CR-300 (Konica Minolta Sensing, Inc., Osaka, Japan) taking four measurements per fruit at the point of widest diameter.

Data were processed using analysis of variance and mean separation was performed with Tukey's test.

Results and Discussion

Our results showed the dramatic influence of rootstock on plant growth and yield; only minor effects on fruit quality were observed. Among the standard rootstocks tested, Swingle citrumelo provided the highest yield. Only the graft combination with 'SRA 92' did not significantly increase total yield in comparison with sour orange and 'Carrizo' citrange, thus confirming the results of previous experiments (Reforgiato Recupero et al., 1992; Reforgiato Recupero and Russo, 1988; Russo and Reforgiato Recupero, 1984).

The hybrid candidate rootstocks present a more variable range of responses in comparison with standard rootstocks. The 'Tarocco TDV' graft resulted in good yields when combined with the hybrids F5 P12, F6 P12, and F6 13 (Table 2); these yields were not significantly different from those obtained for 'Swingle' citrumelo. Candidate rootstocks F5 P12 and F6 P13 performed similarly in two other experiments (Tables 3 and 4). Moreover, F5 P12 showed a higher yield than 'Swingle' citrumelo when grafted to 'SRA 92' clementine.

Canopy size was increased by grafting F14 P37 to 'Tarocco TDV'; the slowest growth was exhibited by the Clehy selection. The mother plant of selection F14 P37 in the experimental orchard of Fonti Ciane

Table 4. Yield, tree size, and efficiency of ‘SRA 92’ clementine on eight rootstocks planted at UTA (CA).

Rootstocks	Cumulative yield (1998–2007) (kg/tree)		Canopy volume (m ³)		Yield efficiency (kg/m ³)	
F4 P6	54.3	a ^z	11.7	a	4.5	a
SO	106.2	ab	14.8	ab	7.6	ab
Carrizo citrange	128.0	b	17.0	ab	7.5	ab
F8 P3	142.9	b	15.5	ab	9.0	bc
F6 P20	145.5	bc	20.8	bc	7.7	bc
SC	161.8	bc	16.2	ab	10.2	bc
F6 P13	223.3	cd	19.1	bc	11.9	c
F5 P12	250.1	d	24.6	c	10.5	bc

^zMean separation within columns by Tukey’s multiple range test at $P \leq 0.05$.

Table 5. Fruit quality of ‘Tarocco’ TDV orange on 19 rootstocks planted at Palazzelli (SR).

Rootstocks	Fruit size (g)		Juice (%)		Total soluble solids (TSS; %)		Total acidity (TA; %)		TSS/TA		Rind color index (a*/b*)	
F1 P6	207.0	a ^z	44.6	b	11.05	g	1.06	bc	10.53	d-f	0.44	cd
CLEHY	215.2	ab	41.6	ab	11.8	h	1.05	a-c	11.31	g	0.43	b-d
F13 P23	224.6	a-c	40.8	a	10.22	a-d	0.97	a	10.61	fg	0.42	a-d
F3 P7	231.0	a-c	41.6	ab	10.97	fg	1.04	a-c	10.69	fg	0.42	a-d
CPT	229.8	a-c	42.3	ab	10.72	d-g	1.09	cd	9.99	b-f	0.4	a-c
F14 P37	232.5	a-c	41.9	ab	10.31	a-d	0.99	ab	10.45	d-f	0.43	b-d
F5 P3	235.6	bc	41.9	ab	9.75	a	1.04	a-c	9.55	bc	0.42	a-d
F6 P12	232.9	a-c	42.3	ab	10.56	c-g	1.11	cd	9.59	bc	0.42	a-d
CC	238.4	bc	41.5	ab	10.39	b-f	1.12	cd	9.39	ab	0.42	a-d
F2 P12	239.5	bc	42.0	ab	11.03	g	1.06	bc	10.54	e-g	0.44	cd
SO	238.7	bc	41.0	a	10.49	b-g	1.09	cd	9.75	b-d	0.42	a-d
SC	246.0	c	42.9	ab	10.1	a-c	1.16	d	8.76	a	0.39	ab
F4 P6	239.8	bc	41.5	ab	10.93	e-g	1.05	a-c	10.49	d-f	0.41	a-d
TC	245.2	c	42.0	ab	10.35	b-e	1.1	cd	9.49	a-c	0.42	a-d
F6 P13	241.7	c	41.9	ab	9.94	ab	1.07	bc	9.32	ab	0.4	a-c
F5 P12	242.6	c	42.7	ab	10.36	b-e	1.07	bc	9.75	b-e	0.45	d
LPT	243.6	c	42.0	ab	10.74	d-g	1.08	b-d	10.04	b-f	0.43	a-d
F6 P2	244.6	c	39.9	a	10.75	d-g	1.09	cd	9.95	b-f	0.38	a
F6 P17	248.8	c	42.0	ab	10.09	a-c	1	ab	10.22	c-f	0.4	a-c

^zMean separation within columns by Tukey’s multiple range test at $P \leq 0.05$.

Table 6. Fruit quality of ‘Washington’ navel Frost orange on eight rootstocks planted at UTA (CA).

Rootstocks	Fruit size (g)		Juice (%)		Total Soluble Solids (TSS; %)		Total Acidity (TA; %)		TSS/TA ratio		Rind color index (a*/b*)	
F4 P2	313.3	d ^z	44.7 ^y		10.00 ^y		0.92	a	11.10 ^y		1.03 ^y	
F4 P6	319.2	d	44.0		10.39		0.99	ab	10.66		1.09	
F6 P13	278.5	a	44.0		10.15		0.96	ab	10.78		1.87	
F5 P12	283.8	ab	44.5		9.98		0.95	ab	10.70		2.14	
F13 P23	280.7	ab	43.6		9.98		0.97	ab	10.55		1.72	
SO	294.1	a-c	42.7		9.95		0.93	a	11.00		1.15	
CC	304.8	cd	44.0		10.33		0.96	ab	11.01		1.00	
SC	276.5	a	44.9		10.18		1.03	b	10.06		1.09	

^zMean separation within columns by Tukey’s multiple range test at $P \leq 0.05$.

^yNo significance.

Table 7. Fruit quality of ‘SRA 92’ clementine on eight rootstocks planted at UTA (CA).

Rootstocks	Fruit size (g)		Juice (%)		Total Soluble Solids (TSS; %)		Total acidity (TA; %)		TSS/TA		Rind color index (a*/b*)	
F4 P6	106.0	b ^z	35.4	a-c	10.13	a	0.73	ab	14.17	a	0.56 ^y	
F6 P13	96.9	ab	38.7	cd	10.59	ab	0.76	b	14.20	a	0.68	
F5 P12	99.0	ab	39.7	d	10.13	a	0.72	ab	14.33	ab	0.65	
SO	93.0	ab	37.7	b-d	10.59	ab	0.75	ab	14.55	ab	0.62	
CC	89.1	a	33.6	a	10.15	a	0.73	ab	14.37	ab	0.61	
SC	99.0	ab	38.9	d	10.84	b	0.77	b	14.51	ab	0.64	
F6 P20	103.6	b	34.6	ab	10.32	ab	0.7	a	15.30	b	0.62	
F8 P3	101.9	ab	39.1	d	10.28	a	0.75	ab	13.94	a	0.61	

^zMean separation within columns by Tukey’s multiple range test at $P \leq 0.05$.

^yNo significance.

displayed upright growth habit; the same canopy shape was transmitted to ‘Tarocco TDV’. F5 P12 and F6 P13 induced a canopy size not significantly different from those obtained for F14 P37. In the two experiments

carried out in Sardinia, the hybrid F4 P6 yielded the smallest canopy size.

No difference among standard rootstocks was observed in effectiveness in controlling yield efficiency (Tables 2–4). For ‘Tarocco

TDV’, the Clehy selection showed the best yield efficiency resulting from reduced canopy size. The hybrid F2 P12, ‘Carrizo’ citrange, and ‘Swingle’ citrumelo also provided high yield efficiency. As a result of low yield, the sour orange showed the lowest yield efficiency effectiveness in the experiment with ‘Washington’ navel (Table 3). The same trend was recorded for the F5 P12 hybrid as a consequence of large canopy size; the highest values for effective control of yield efficiency were observed for F4 P2, resulting from reduced canopy volume. In the experiment with the ‘SRA 92’ clementine (Table 4), F4 P6, ‘Carrizo’ citrange, sour orange, and F6 P20 induced low canopy-yield effectiveness, whereas the highest values were recorded for F6 P13.

We observed less influence of rootstock on fruit quality characteristics. In the experiment with ‘Tarocco TDV’ (Table 5), a small fruit size was obtained with the F1 P6 hybrid; this hybrid reverted also among those less productive. The F5 P12, F6 P12, and F6 P13 selections showed high total yields. Their fruit size is not significantly different from that obtained for other rootstocks. Fruit juice content was slightly influenced by rootstock; F1 P6 showed the highest value and F6 P2, sour orange, and F13 P23 displayed the lowest values, but not significantly different from those obtained for other rootstocks. Clehy, F1 P6, and F2 P12 selections yielded the highest fruit TSS content. The yield from plants grafted on Clehy rootstock also showed the highest maturity index, whereas F5 P3, F6 P13, and F6 P17 had the lowest TSS content. ‘Swingle’ citrumelo produced the juice with the highest acidity content. The highest values of TSS and a/b flesh were encountered in Clehy followed by F1 P6, F3 P7, and F4 P6 (Table 8). The correlation between TSS content and anthocyanin pigmentation of 2006 sampling was measured by Pearson coefficient and found to be positive ($r = 0.89$).

Table 8. Correlation between total soluble solids (TSS) and a/b flesh in 2006 (by Pearson coefficient^z) for ‘Tarocco’ TDV orange.

Rootstocks	TSS	a/b flesh
F5-P3	9.78	0.77
F6-P13	9.78	0.82
F6-P17	9.89	0.73
SC	9.96	0.70
F14-P37	10.14	0.99
F13-P23	10.31	1.02
F5-P12	10.42	0.83
CC	10.45	1.12
LPT	10.54	1.03
SO	10.57	0.98
TC	10.61	1.06
F6-P2	10.78	0.98
CPT	10.81	1.15
F6-P12	10.82	1.03
F2-P12	10.91	1.11
F4-P6	11.2	1.21
F3-P7	11.31	1.33
F1-P6	11.34	1.33
CLEHY	12.24	1.32

^z $r = 0.89$; $P = 0$; $n = 19$.

In the experiment with 'Washington' navel (Table 6), the SW rootstock resulted in the highest juice acidity levels significantly different from those obtained for SO and F4 P2. In the experiment with 'SRA 92' clementine (Table 7), the lowest acidity content was obtained for F6 P20.

No influence of rootstock was observed on peel color, TSS (with the exception of the 'Swingle' citrumelo for clementine), or maturity ratio (with the exception of F6 P20 for clementine).

Our data, however, suggest that 'Tarocco' orange may exhibit more intense red color flesh and related high sugar content when grafted on some of the tested hybrids as opposed to trifoliolate orange (Table 8). The anthocyan flesh color of 'Tarocco 57/1E/1' orange (a poorly pigmented selection) was increased by the trifoliolate orange rootstock 'Flying Dragon' as observed in a previous trial carried out in an environment unfavorable to anthocyan biosynthesis (Reforgiato Recupero et al., 1999). These observations were confirmed by recent data from the new 'Tarocco' plantation in China experimenting with the trifoliolate orange as a rootstock (Reforgiato Recupero, personal communication). In Castellarin et al. (2007), the authors showed that the onset of anthocyanin accumulation was closely coordinated with sugar accumulation, consistent with a sugar-dependent trigger as suggested by Solfanelli et al. (2006). Carbohydrate accumulation, locally or at a whole plant level, is a common response to all the main environmental inducers of anthocyanin synthesis (Steyn et al., 2002).

Regarding iron chlorosis, 'Swingle' citrumelo sensitivity remains a limiting factor in widespread use of this rootstock. In our experiment carried out at PA, the active calcium carbonate was 4.5%, sufficient to cause iron chlorosis symptoms according to previous reports (Castle and Stover, 2001). However, certain factors may increase the natural sensitivity of the 'Swingle' citrumelo to active calcium carbonate in soil; consequently, it is not easy to determine the limit for the recommended use of this rootstock in calcareous soils. In a previous experiment with the orange cultivar Navelina ISA 315, iron chlorosis symptoms were observed within 10 years of the planting date (Reforgiato Recupero et al., 1992). F6 P12 showed tolerance; F5 P12 displayed intermediate sensitivity; and F6 P13 demonstrated sensitivity

similar to that of the 'Swingle' citrumelo. More experiments in different citrus-growing areas are necessary to determine the effective limit of their active calcium carbonate tolerance.

Conclusions

Some of the tested hybrids (F5 P12, F6 P12, and F6 P13) may improve plant yield, thus maintaining good fruit quality. However, the sensitivity of these hybrids was not studied with relation to the tristeza sensitivity; further research is necessary to determine whether these genotypes display male parent resistance (Asins et al., 2004; Brnnet et al., 2004; Gmitter et al., 1996).

Encouraging data obtained with F5 P12, F6 P12, and F6 P13 may justify the use of monoembryonic species of the *Papeda* subgenus for breeding citrus rootstocks. Selection for nucellar embryony eliminated many hybrids (Reforgiato Recupero and Russo, 1992); however, the few polyembryonic hybrids obtained revealed the potential of unexplored germplasm. This selection's success highlights the likelihood of additional candidate citrus rootstocks in the near future.

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