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REGULAR ARTICLE

SELECTION OF NEW PROMISING SEEDLESS MANDARINS TRIPLOID HYBRIDS FROM CROSSES BETWEEN MONOEMBRYONIC DIPLOID FEMALE AND DIPLOID MALE PARENTS

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ABSTRACT

Morocco is one of the major exporters of small citrus fruits, such as mandarin and Clementine. Seedlessness is a major criterion for this horticultural group. The present study focused on the selection of the best triploid mandarin hybrids (2n=3x=27) characterized by seedless fruits. A series of crosses between 'Sidi Aissa' clementine (female parent) and seven mandarin varieties ('Lee', 'Wilking', 'Osceola', 'Carvalhal', 'Satsuma Frost', 'Satsuma Owari' and 'Chienka') was performed by the National Institute for Agricultural Research. Forty triploid mandarins were obtained and planted since 2002 in an experimental field at El Menzeh. Varietal evaluation was focusing on fruit quality traits during seven years. Statistical analyzes showed that there is a significant difference for all studied characters and between hybrids. The number of seeds per fruit is the main criterion which differentiates between triploids mandarin's hybrids and their diploid parent "clementine Sidi Aissa". The best hybrids selected were: HT11, HT13, HT27, HT43, HT44, and HT49. The best crosses are *C1* ('Sidi Aissa' × 'Wilking') and*C2* ('Sidi Aissa' ×'Osceola'). These promising triploid hybrids of mandarin have been multiplied on several rootstock trials and are in the process of quantitative evaluation and multi-site testing.

Keywords: Morocco; *Citrus* spp.; mandarins; breeding program; triploidy; fruit quality

INTRODUCTION

The citrus sector is the largest fruit's production in both value and volume in the world economy. In recent years, the consumption of small citrus fruits, particularly clementines and mandarins, has grown rapidly in the international market. Fruit quality is the determining factor in the commercial value of Citrus crop. A number of variable criteria define the quality requirements of citrus fruits. Citrus quality standards depend on their variety, just as their region of production and market is expressed [1, 2].

Seedlessness major economic characteristic of mandarins which determines the market value, and mandarin triploid hybrids bears this important trait [3]. Triploid citrus cultivars (2n = 3 x = 27) have great commercial potential [4]. Triploid embryos will rarely germinate and found in smaller seeds [5]. Otherwise, these triploid plants can be obtained from the crosses of two diploid genotypes by fertilization of 2n mega gametophyte [6-13]. Spontaneous triploids also occur in about 5% of the seeds obtained from diploid parents. They are found in the small seed, weighing less than 0.1 g. In addition, triploid hybrids can be produced by crossing a diploid parent and a tetraploid parent. If mono-embryonic zygotic diploid genotypes are used as the female parent and tetraploid genotypes as the male parent, many poorly developed seeds are produced [14-16,3]. These triploids can be derived either from the union of unresolved female gametes with reduced male gametes (18+9), or from the fusion of reduced female gametes with unreduced male gametes (9+18) [17-19]. Hybridization and embryos rescue are a promising method for production of triploid hybrids yielding seedless fruit. The present study focused on the evaluation of 40 triploid new mandarin's hybrids from crosses between diploid parents.

MATERIALS AND METHODS

Plant material

Forty new triploid hybrids of mandarin trees were obtained after a series of directed crosses and shared common female parent (half sibling). Thus, seven combinations have been highlighted and each one of them was constituted by full siblings. The Clementine 'Sidi Aissa' was used as a female parent since it is mono-embryonic, self-incompatibility and very appreciated by high production and good quality. Seven mandarin's cultivars were therefore, used as male parents (table 1). These hybrids were regenerated from abortive seeds. These were sterilized and cultured on Murashige and Skoog (MS) basal medium supplemented with 500 mg L^{-1}malt extract (ME)

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and 1 mg L^{-1}gibberellic acid (GA_3), and kept at 26±1 °C with 16-h photoperiod. Seedlings at the five-leaf stage were transferred into plastic pots containing garden soil and kept in a greenhouse. Triploid seedlings were identified by flow cytometry (Sysmexpartec/Cyflow Space) (*fig. 1*). They were grafted on 'Troyer' citrange rootstock and planted at a spacing of 6 × 4m in April 2002.

Characteristics of the experimental site

The experiment was carried out at El Menzeh experimental area (INRA Kenitra, Morocco). The climate is of the Mediterranean type belonging to the sub humid area. The soil is of the sandy type at the surface (98%) and sandy-clayey at depth, with a pH of 6, a retention capacity of 3 to 6%, a cation exchange capacity of 5 meq $100g^{-1}$ with no limestone. The quantities of water needed to properly irrigate the plot are defined according to the climatic zone. The citrus water requirements are estimated at around 1,200 mm per year, spread over the whole year. The experimental period is characterized by an average annual rainfall of 511 mm. The 589 to 689 mm deficit was filled by irrigation from May to September-October (micro-jet) with modulated intakes based on potential evapo-transpiration values. Over seven years of observations, the average annual temperature was 16 °C with an average maximum of 22 °C and an average of the minimum of 10 °C. For the conduct of the test, nitrogen fertilization (amonitrate 33.5%) was carried out on the basis of three annual intakes distributed according to the following proportions: 50% before flowering, 25% after flowering and 25% after physiological fall. Contributions of potassium sulfate and triple superphosphate were respectively made in September and December.

Characters to be evaluated

The fruit ripeness of the new triploid hybrids started from the first week of October. Thus, samples of 30 fruits per replicate were randomly collected at harvest date for determining physical and chemical characteristics. Juice analysis was performed at precise dates during seven years: *D1*(22/10/2007); D2*(16/10/2008)*; D3(07/10/2009); D4(29/10/2010); *D5*(26/10/2011); D6*(29/10/2012)* and D7(17/10/2013). To obtain representative samples, a fruit collection was done on two trees per hybrid.

Juice content

The juice extraction for analysis was performed in the laboratory of INRA El Menzeh according to the standards [20]. The production of a blended juice per accession was carried out by a rotary extractor. The collected juice was filtered through a plastic filter and weighed. The juice content was expressed as a percentage offresh weight following the formula: Juice content

$$(\%) = \frac{\text{Weight of juice extracted from 30 fruits}}{\text{Total weight of 30 fruits}} \times 100$$

Total soluble solids

The sugar content was estimated by the total soluble solids (TSS) determined by digital benchtop refractometer (BellinghamandStanley RFM700-M). A drop of each sample fruit juice was deposited on the prism and the reading of the soluble solids (in °Brix) was made at room temperature.

Table 1: List of new mandarin hybrids from crosses between clementine 'Sidi Aissa' and eight male parents of mandarin (HT: triploid hybrid)

Crosses	Mandarins (♂)	Clementine 'Sidi Aissa' (♀)	Hybrids number
C1	Wilking	HT10 HT11 HT12 HT13 HT14 HT15 HT16 HT17 HT19 HT20 HT21 HT22 HT23 HT24 HT25 HT27 HT28 HT29 HT48 HT50 HT53	*21*
C2	Osceola	HT5 HT43 HT44 HT46 HT51 HT52 HT56	*7*
C3	Chienka	HT30 HT32 HT33 HT34 HT36 HT61	*6*
C4	Satsuma Frost	HT7 HT8	*2*
C5	Carvalhal	HT49	*1*
C6	Lee	HT6	*1*
C7	Satsuma Owari	HT47 HT59	*2*
Total	7	*40*	

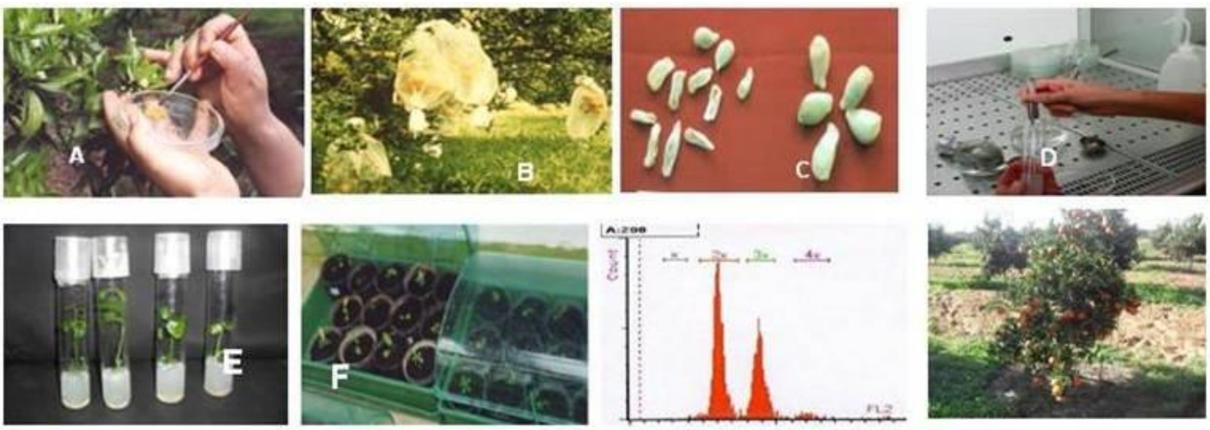

Fig. 1: Production phases of mandarin triploids hybrids. A: Pollination; B: Fruits from crosses; C: Abnormal seeds on the left; D and E: *In vitro* germination of immature embryos; F: Acclimatization phase in greenhouse; G: Histogram of flow cytometry for DNA amount counting; and H: Grafting and production of triploid hybrids

Juice acidity

Juice acidity was measured by titration. 10 ml of the decanted juice were used and three drops of phenolphthalein (1g100 ml^{-1} ethanol) were added. The titratable acidity (TA) is obtained according to the following formula: TA = $\frac{Vs}{10}$. Where *Vs* is the volume of NaOH (ml) used for the titration and 10 (mL) is the volume of juice used.

Maturity index

The citrus fruit behaves like a non-climacteric organ. If harvested before maturity, it will never acquire suitable organoleptic qualities [21] The maturity index (MI) is a criterion widely used in the determination of the maturity or ripening date (for harvesting) citrus fruit. It is determined by the ratio: MI = $\frac{TSS}{TA}$, where *TSS* is the sugar content estimate (°Brix) and *TA* is the Titratable acidity of the juice.

Average weight of a fruit

This average is determined by measuring total weight of 30 fruits per accession. It was appreciated using an electric scale (Platform scale/EBW-60).

Fruits morphological description

The selected hybrids were characterized by the following pomological criteria recorded on 10 fruits per accession: fruit diameter and length (mm), diameter/length ratio, fruit shape (ellipsoid, pyriform, obloid, round), fruit skin (orange, orange yellow, yellow), thickness of segment wall (thin (<2 mm), medium (2 mm<<6 mm) or thick (>6 mm), and adherence of segment walls (weak, medium, strong).

Statistical analysis

Quantitative data were analyzed using SAS (Statistical Analysis System version 9.1 and version 5.5) and were subjected to analysis of variance (ANOVA), and means were compared with Duncan test at 5% level of significance.

RESULTS AND DISCUSSION

Quality of the triploid fruits

A highly significant difference was observed between the genotypes for all the studied criteria while the year effect was not significant for the variables number of seeds per fruit and the maturity index (E/A) (*tables 2, 3, 4*). Likewise, the [year × genotype] interaction was not statistically significant. The data expressed a rather large variability in the profile of each hybrid compared to that of their female parent the Clementine 'Sidi Aissa'. There was a highly significant difference between the mandarin hybrids for all the criteria studied. The number of seeds ranged from 0 to 3 according to the new mandarin's hybrids, with 75% of the hybrids having 0 or 1 seed per fruit (*fig. 2*), compared to 7 seeds per fruit for the control Clementine 'Sidi Aissa'. Generally, the triploid causes male and female sterility, preventing cross-pollination on incompatible cultivars [5,11]. Seedlessness is an important criterion for distinguishing and selecting new mandarin varieties [23]. The average fruit weight of mandarin hybrids was 77g, in a range of 48g to 125g. The triploid hybrids HT19 and HT24 yielded the maximum fruit weight (98g and 108g, respectively) whereas the hybrids HT10 and HT21 had small sized fruits (47g and 50g, respectively). Furthermore, 18% of the hybrids had a higher average fruit weight than that of the control 'Sidi Aissa' (88g). This criterion is important in marketing but influenced by climatic factors, the date of harvest and the position of fruit in the tree [24, 26]. In addition, the juice content varied from 24% to 56% with an average of 44%. The triploid hybrids HT17, HT10 are the juiciest whereas HT32 and HT12 hybrids have the least juicy fruit. In addition, 28% of the hybrids studied have a percentage of juice higher than that of the control 'Sidi Aissa' (44%). This parameter increases at maturity and is used as a standard marketing index. It depends on the species and the destination market. Indeed, are the most requested and the most appreciated by consumers. As the others traits, the juice content can be improvement because it varies according to the varieties, the stage of maturity, the exposure and the positioning of the fruits in the tree, the contribution of fertilizers and the climatic conditions [27-34]. Also, variance analyze showed that acidity is highly variable across the 40 hybrids and as a result, several statistical groups were highlighted. The acidity ranged from 0.36 (HT5) to 1.87 (HT10) while the control has an acidity of 0.87. Similarly, 88% of hybrids have higher acidity than the control.

In addition, the sugar content ranged from 8.20 to 11.55 with an average of 9.89. Triploid HT17 and HT16 hybrids have been characterized by the highest value while HT5 and HT8 hybrids are the least sweet. 15% of triploid hybrids have sweeter fruits than those of the control. Otherwise, the maturity index (E/A), an indicator of fruit ripeness, ranged from 6 to 23. The hybrids HT5, HT17 are the earliest, while the hybrids HT8, HT10, HT11 and HT50 are relatively late. So, 13% of the hybrids *ripen* three weeks later *than* its parental "Sidi Aissa". Usually, this variable is low in early-picking fruits compared to late-picking fruits. Generally, all fruit quality criteria are variables with the exception of seedlessness which is a central issue in the development of Citrus sector. Although, researchers continue to search for the right combination of factors that improve citrus fresh fruits quality, because it's influenced by many factors, including, rootstock, production techniques, environmental conditions [25, 26, 31, 35-39]. The seedless mandarin hybrids selected will be grafted on some rootstocks and tested in several areas.

Effect of the male parent on the variability of the fruit quality

The triploid hybrids of mandarin tree obtained from the cross (C4) [clementine 'Sidi Aissa' ×mandarin 'Satsuma Frost'] were characterized by their capacity to develop seeds (2 to 4 seeds per fruit). While, the hybrids from the others crosses produce seedless fruits (0 or 1 seed per fruit) (table 5). The average weight fruit was relatively low in hybrids from the combination [Sidi Aissa × Satsuma Owari]. However, the combination [Sidi Aissa × Carvalhal] was identified by the highest values for both the sugar content and the average fruit weight. The combination [Sidi Aissa × Chienka] was characterized by a high value of the juice percentage. While, the combination [Sidi Aissa × Wilking] was distinguished by a high value of acidity. Previous studies reported that in triploid progeny, the codified characteristic of many genes (fruit size and flavor) seemed to be strongly influenced by the tetraploid parent [15]. The same authors obtained triploid hybrids resembled the male parent tetraploid in fruit size, easy of peeling, and varying degrees of anthocyanin pigmentation that increased as with the maturity of the fruits. Likewise, [40] showed that the male parent has a large influence on the offspring and male parents with a deeper orange bark color, larger fruit size, better fruit shape and higher internal quality have resulted to families with an improvement of the population. Therefore, only male parents of the highest quality should be used in breeding programs to increase the chances of obtaining a new high quality cultivar. While, the female parent would be preferable self incompatible and monoembryony as the case of clementine. The best combinations are Sidi Aissa X Wilking (HT11, HT13, T25, HT27); Sidi Aissa X Osceola (HT43, HT44, HT51, HT52); Sidi Aissa X Chienka (HT33) and Sidi Aissa X Caravalhal (HT49). Also, Ray [1] showed that high variability among F1 hybrids is due to the high heterozygosity that occurs in Citrus.

Table 2: Synthesis of statistical results of the quality criteria of all triploid hybrids of mandarin trees (El Menzeh, INRA, 2007-2013)

Crosses	Hybrids	Fruit weight (g)	Fruit seed number	Juice content (%)	Acidity (TA)	Soluble solids (°Brix) (TSS)	Maturity index (TSS/TA)
C1= Sidi Aissa x Wilking	HT10	47.9l	0.4cd	49.3ab	1.8a	10.9abcd	6.0f
	HT11	89.2bcdef	0.4cd	43.3bcdef	1.7ab	10.5abcdefg	6.4ef
	HT12	78.8cdefghij	3.6b	23.6i	0.9cdefgh	8.9ijk	10.2bcdef
	HT13	80.7cdefghi	0.9cd	38.4bcdefgh	1.3abcdef	10.3abcdefgh	8.5bcdef
	HT14	66.7fghijkl	0.7cd	48.9abc	1.0cdefg	10.1bcdefghi	10.1bcdef
	HT15	60.0ijkl	0.5cd	43.5bcdef	0.9cdefg	10.4abcdefg	11.4bcdef
	HT16	81.8cdefghi	0.4cd	38.8bcdefgh	1.1cdefg	11.2ab	11.7bcdef
	HT17	65.0fghijkl	3.1b	56.0a	0.8efgh	11.5a	13.9b
	HT19	97.7bc	0.1d	44.4abcde	1.0cdefg	10.2bcdefghi	11.1bcdef
	HT20	78.3cdefghij	0.2d	44.9abcde	1.1cdefg	9.9bcdefghij	9.2bcdef
	HT21	50.5kl	1.2cd	46.3abcde	1.3abcdefg	10.4abcdefgh	8.7bcdef
	HT22	74.1cdefghijk	0.5cd	37.7bcdefgh	1.1cdefg	9.4ghijk	8.9bcdef
	HT23	56.7jkl	0.4cd	44.6abcde	0.8efgh	9.7cdefghij	9.6bcdef
	HT24	108.2ab	0.6cd	36.8cdefgh	1.1cdefg	9.5efghij	10.4bcdef
	HT25	95.3bcd	1.3cd	37.7bcdefgh	0.7gh	9.9bcdefghij	11.6bcdef
	HT27	92.9bcde	0.9cd	34.5efghi	1.0cdefg	10.3abcdefgh	10.3bcdef
	HT28	72.6defghijk	0.1d	40.8bcdefg	1.1cdefg	10.1bcdefghi	10.0bcdef
	HT29	66.5fghijkl	2.5bc	36.5defgh	1.4abcde	9.8cdefghij	9.3bcdef
	HT48	76.3cdefghij	0.4cd	38.8bcdefgh	1.3abcdefg	10.2bcdefghi	8.6bcdef
	HT50	92.5bcde	0.8cd	39.9bcdefgh	1.5abc	10.0bcdefghi	6.6def
	HT53	93.4bcde	0.4cd	47.2abcd	0.9defgh	11.0abc	12.5bcd
C2	HT5	80.0cdefghij	0.0d	31.2ghi	0.3h	8.2k	22.8a
	HT43	84.9cdefgh	0.7cd	45.8abcde	1.0cdefg	10.1bcdefghi	10.1bcdef
	HT44	82.6cdefghi	0.7cd	43.4bcdef	1.0cdefg	10.7abcdef	10.5bcdef
	HT46	66.1fghijkl	0.3cd	48.2abcd	1.2bcdefg	9.5efghij	7.9cdef
	HT51	57.4jkl	0.9cd	43.7bcdef	1.0cdefg	10.5abcdefg	10.2bcdef
	HT52	69.5efghijkl	0.6cd	48.2abcd	0.9defgh	9.7defghij	11.7bcdef
	HT56	63.7ghijkl	0.7cd	42.3bcdefg	1.5abcd	9.9bcdefghij	7.1cdef
C3	HT30	83.6cdefghi	1.2cd	49.2ab	1.2bcdefg	9.6efghij	8.8bcdef
	HT32	76.6cdefghij	1.1cd	28.2hi	1.1cdefg	8.9ijk	9.5bcdef
	HT33	86.9bcdefgh	0.2d	32.4fghi	0.8fgh	8.6jk	12.4bcde
	HT34	77.2cdefghij	0.2d	43.1bcdefg	1.0cdefg	10.8abcde	11.1bcdef
	HT36	52.2kl	0.3cd	32.2fghi	1.1cdefg	9.1hijk	8.2bcdef
	HT61	62.7hijkl	1.0cd	45.4abcde	1.1cdefg	9.2ghijk	8.5bcdef
C4	HT7	59.8ijkl	3.9b	38.7bcdefgh	1.2bcdefg	9.4fghijk	8.5bcdef
	HT8	82.4cdefghi	3.6b	44.9abcde	1.4abcde	8.6jk	6.5def
C5	HT49	80.3cdefghij	0.5cd	40.4bcdefg	1.4abcdef	10.7abcdef	7.3cdef
C6	HT6	82.2cdefghi	0.3d	42.2bcdefg	1.1cdefg	9.2ghijk	8.4bcdef
C7	HT47	64.7ghijkl	0.4cd	37.8bcdefgh	1.3abcdefg	8.9ijk	7.5cdef
	HT59	125.0a	0.0d	32.0fghi	0.7gh	9.2ghijk	12.8bc
Control	'Sidi Aissa'	87.7bcdefg	7.2a	43.7bcdef	0.9defgh	10.0bcdefghi	11.6bcdef

Within each column, the values bearing the same letters are significantly indifferent at$P$>5%.

Table 3: Descriptive statistics, analysis of variance (Y=year, G = genotypes, [Y × G] = their interaction) and coefficient of variation for some quality fruit characters in mandarin hybrids with mother control

Sources	Mean squares					
	Mean fruit weight (g)	Fruit seed number	Juice content (%)	Soluble solids(TSS)	Acidity (TA)	TSS/TA
Year (Y)	9,043.33 **	0.55 ns	364.73 **	12.61 **	0.12 ns	1.92 ns
Genotype (G)	684.07 **	10.22 **	125.87 **	1.72 **	0.25**	17.14 *
(Y) X (G)	318.32 ns	1.10 ns	49.76 ns	0.71 ns	0.17 ns	14.33 ns
Error	235.23	1.24	36.42	0.70	0.09	12.11
CV (%)	19.82	94.54	14.50	8.38	27.27	35.39
Average	77.40	1.18	41.61	9.99	1.11	9.829

NS: no significant, * and **: significant at 5% and 0.1%, respectively.

Table 4: Physico-chemical traits of mandarin hybrids according to year

Years	Variables					
	Mean fruit weight (g)	Fruit seed number	Juice content (%)	Soluble solids(TSS)	Acidity (TA)	TSS/TA
2007	84.66ab	1.29a	37.16b	10.29b	0.82c	13.15a
2008	67.85c	1.07a	37.65b	9.83bc	0.96c	10.78bc
2009	59.55c	0.95a	44.38a	9.56c	1.37a	6.59d
2010	80.75b	1.61a	40.97ab	9.46c	1.00 bc	9.28c
2011	84.86ab	1.09a	41.87ab	10.07bc	1.06bc	11.02bc
2012	89.37ab	1.16a	42.71ab	10.18b	1.22ab	9.31c
2013	96.36a	1.52a	44.57a	11.73a	1.00bc	11.97ab
Range						
2007	50-125	0-9	20-50	8-12	0.36-1.38	9-23
2008	45-90	0-5	23-47	8-11	0.60-1.43	7-17
2009	29-85	0-4	23-62	8-11	0.61-2.20	5-13
2010	55-110	0-10	25-50	8-11	0.77-1.62	6-14
2011	50-140	0-6	27-67	9-11	0.55-1.70	4-23
2012	70-115	0-12	26-50	8-12	0.54-1.70	5-23
2013	55-125	0-8	35-54	10-14	0.70-1.26	8-16

Within each column. the values bearing the same letters are significantly indifferent with *P*>5%. Juice analysis dates: D1: 22/10/2007; D2: 16/10/2008; D3: 07/10/2009; D4: 29/10/2010; D5: 26/10/2011; D6: 29/10/2012; D7:17/10/2013.

The final rate of selection was 15% (table ***6***). Currently, special attention is paid to the risk of pollination and the isolation of clementine orchards and other seedless varieties [41]. As a result, the cultivation of mandarin Wilking has been officially banned in Morocco to avoid the production of clementines with too many seeds. But, this mandarin has a great scientific interest in the program of creation of triploid mandarins. It has been used as a male parent in directed crosses between diploid mandarins and has resulted in relatively high numbers of immature embryos and triploid hybrids [9]. Therefore, it can be seen that the families with male parents Wilking and Osceola having showed a greater improvement in the population when crossed with female parent Sidi Aissa. The true seedlessness can be obtained by selection among triploid genotypes [42]. Another study [43] revealed the female parent will determine the success rate of hybridization. Many citrus triploid hybrids have originated from sexual hybridizations [6, 44, 45, 3].

Table 5: Study male parent effect on fruit quality variation according to seven series of crosses

Crosses	Variables					
	Mean fruit weight (g)	Fruit seed number	Juice content (%)	Soluble solids (TSS)	Acidity (TA)	TSS/TA
C1	78.0 c	0.91b	41.35 b	*1.17 a*	*10.20 a*	9.58 b
C2	72.0 c	0.57 b	*43.26 a*	1.02 ab	*9.81a*	*11.46 a*
C3	73.2 c	0.66 b	38.41 c	1.06 ab	9.39 ab	9.74 b
C4	71.1 c	*3.75 a*	41.84 b	1.32 a	9.05b	7.53 c
C5	80.3 b	0.47 b	40.42 b	1.37 a	*10.72 a*	7.26c
C6	73.4 c	0.33 b	39.97 c	1.20 a	9.07 b	7.97 c
C7	*106.4 a*	*3.61a*	37.69 c	0.79 b	9.61 ab	12.20 a
Means	79.2	1.47	40.42	1.13	9.69	9.39
SD	*12.45*	*1.52*	*1.94*	*0.20*	*0.61*	*1.93*

Within each column, the values bearing the same letters are significantly indifferent with *P*>5%.

Table 6: Selection rate of triploid hybrids according to seven crosses mandarins

	Parents (♂ X ♀)	Total number	Selection number	Selection rate(%)	Triploids hybrids seleted
C1	*Sidi Aissa X Wilking*	21	3	14.29	HT11, HT13, HT27
C2	*Sidi Aissa X Osceola*	7	2	28.57	HT43, HT44
C3	*Sidi Aissa X Chienka*	6	-	-	-
C4	*Sidi Aissa XSatsuma Frost*	2	-	-	-
C5	*Sidi Aissa X Caravalhal*	1	1	100	HT49
C6	*Sidi Aissa X Lee*	1	-	-	-
C7	*Sidi Aissa X SastumaOwari*	2	-	-	-
	Total	40	6	15.00	

C1: Croisement (Sidi Aissa X Wilking) C2: Croisement (Sidi Aissa X Osceola);C3: Croisement (Sidi Aissa X Chienka) C4: Croisement (Sidi Aissa X Satsuma Frost)C5: Croisement (Sidi Aissa X Caravalhal) C6: Croisement (Sidi Aissa X Lee) C7: Croisement (Sidi Aissa X Satsuma Owari)

Table 7: Diameter and shape of fruits of promising mandarin hybrids

Crosses	Selected hybrids	Diameter (D) (mm)	length (P) (mm)	(D/P)	Shape
C1	HT11	61,14	60,00	1,02	Pyriform and round
C1	HT13	61,14	60,00	1,02	Pyriform and round
C1	HT27	68,11	53,11	1,28	flattened
C2	HT43	66,61	54,60	1,21	flattened
C2	HT44	63,91	50,41	1,27	flattened
C5	HT49	61,14	60,00	1,02	Pyriform and round
Control	Sidi Aissa	62,86	51,52	1,22	flattened

(D/P indicator of fruit shape if D/P=1, the fruit is round)

Pomological study of promising mandarin hybrids

The coloring of fruit bark is among the most important quality criteria that influence the perception and appreciation of consumers. Generally, it depends on the climatic conditions [41]. The selected hybrids HT43, HT 44 and HT49 have an attractive dark orange rind, while the others are yellowish (fig. 3). All triploids hybrids selected were characterized by sweet flavor, medium fruits, easily to peel, firm fruits with thin epicarp (<2 mm) and weak adherence of segment walls. In addition, they have a flattened shape except for the hybrid HT11, HT13 and HT49 which have a pyriform and round shape (table 7).

CONCLUSION

Triploid mandarin hybrids selected were grafted on several rootstocks in order to complete the present study by multifocal varietal trials. The hybrids selected will be registered in the Moroccan official catalog, multiplied and then put at the disposal of the nurserymen. The seedlessness illustrates this interest of the triploids. Thus, spontaneous triploidy is a promising way for the selection of seedless mandarin fruit varieties, but, still remains the frequency of relatively low triploid compared to crosses (2x X 4x).

Fig. 3: New promising mandarin seedless triploid hybrids compared to mother plant Clementine *Sidi Aissa*

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