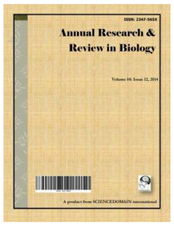

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Effect of Gamma Radiation on the Variability of Some Quality Criteria in Sidi Aissa Clementine

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Authors' contributions

This work was carried out in collaboration between all authors. Authors HB and NH designed the study. Author TA performed the statistical analysis, managed the analyses in the laboratory with the help of the author NA and wrote the manuscript. Author MM managed the analyses (irradiation) of the study. Author NB managed the literature searches and author BAEA performed the English corrections. All authors read and approved the final manuscript.

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ABSTRACT

Citrus fruits are an excellent source of vitamin C, a nutrient that strengthens the immune system and keeps the skin smooth, elastic and has a great socio-economic importance at the national level. Seedlessness is a desirable characteristic in citrus fruit sold for fresh consumption.

This work is part of a genetic diversification program of citrus. The aim of this research is to study the variability of selection criteria related to the quality of the fruit. Ninety-six clones were obtained by bud irradiation from Sidi aissa clementine with two different ionizing radiation doses (30 and 50 Gy). The buds were grafted onto *macrophylla* and planted with a spacing of 3x5 m^2. The evaluation focused on the organoleptic and pomological criteria. Statistical analysis showed that fruit quality was affected differently by gamma irradiation. Some clones presented no changes compared to the control *'Sidi Aissa'* clementine, while other clones showed significant differences. They showed

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either higher or lower levels of the researched characteristics. From the 96 clones, 60 showed a juice content higher than the average of 52.12%, in the sugar levels, 29 clones showed a higher level than the control Sidi Aissa with 9.09 %, 70 clones resulted in a lower number of acidity with a percentage of 0.79%, in weight, 72 clones were heavier with an average of 85,38g and lastly 34 clones were too late with a mean of 10,14 for the maturity index. Two irradiated clones of clementine 50 M85 and 30 M68 were selected because of their highest fruit quality for all criteria and seedlessness compared to the control Sidi Aissa that has 7 seeds per fruit. Gamma-irradiated varieties were slightly or significantly preferable to that of unirradiated Sidi Aissa.

Keywords: Citrus; Clementine; irradiation; genetic diversification; quality; clone.

1. INTRODUCTION

Moroccan citrus production reached 2.3 million metric tons in 2016/17, which corresponds to an increase of 15 percent compared to the previous year. Orange production has risen 4 percent over the previous year registering 962,250 MT, while tangerine/mandarin production has increased 24 percent and reached 1,325,246MT [1]. This significant rise of production is due to the good condition in the harvested area. Orange export has reached around 120,500 MT, while tangerine/mandarin export about 510,350 MT. The rise in export has been optimized due to more fruit being produced to satisfy the growing demand from Russia and the EU coupled with Morocco [1]. Sidi Aissa is an early clementine variety originated in the village of Sidi Aissa, which is located in the surroundings of the city of Beni-Mellal (Morocco). Sidi Aissa is known for the globular shape and its beautiful orange color. Although the fruit are quite acid, easy to peel, seedless, very juicy and tasty [2]. The trees are very productive and vigorous naturally, additionally they vary in size depending on the rootstock used. In the Gharb region, for example, the production of Sidi Aissa on Troyer citrange rootstock reaches 60 t/ha. The fruit are firm and have a good quality juice content [3].

Sidi Aissa was considered as a quick solution to small-scale problems of Moroccan citriculture [4]. Effectively, it has given satisfactory results in some localities with wet microclimate (Gharb and the coastal region between Rabat and Larache) [5] However, clementine varieties require an isolated planting from other pollinating varieties, hence the need to develop sterile varieties. Improvement methods are numerous and their choice depends on the means available and breeding objectives. Ionizing radiation (gamma rays) of seeds or buds can induce structural mutations leading to reduced number of seeds like Clementine Monreal or to gain totally aspermic mutants [6,7]. Seedlessness is an important economic trait relating to fruit quality. Actually, gamma irradiation mutagenesis of bud wood is the most common method used by citrus breeders worldwide to obtain seedless clones from commercial seeded varieties [8,9].

This study focuses on the evaluation of new variants obtained from the irradiation of Sidi Aissa clementine with two doses of gamma radiation 30 and 50 Gray.

2. MATERIALS AND METHODS

2.1 Plant Material

The buds of Sidi Aissa clementine were irradiated in 2009 at the radioactivity station of INRA (Tanger), using two doses of gamma radiation (30Gy and 50Gy) and then grafted on macrophylla rootstock. After six months the 97 (96 clones + 1 control) associations obtained were planted in the fields of Abbes Kabbage (Belkciri, Morocco) with 3x5-m² spacing. Clone variants were numbered as 30Mx or 50Mx (30/50 correspond to the irradiation doses and x/y stand for the number of clones).

2.2 Tree Maintenance and Irrigation

The experiment was set up in 2009 at the fields of Abbes Kabbage in BelKciri (Morocco). Trees grafted on macrophylla were irrigated by a drip irrigation system. The amounts of water needed to properly irrigate the land were defined depending on the climate in that area. For citrus fruit the need in water ranges from 900 to 1200 mm per year. Temperature was measured regularly at the station during the experimental period (Table 2) and represents a critical factor for high internal quality and good coloring of fruit. Over a period of three years of observations, the fertigation system was most suitable in El Kabbage fields. The trees were maintained annually using standard cultural techniques practiced in citriculture.

Table 1. Clementine clones irradiated with Gamma ray

Dose (Gy)	Number of buds	Buds code
30	46	30M22; 30M23; 30M24; 3017; 30M1; 30M103; 30M11; 30M13; 30M15; 30M2; 30M25; 30M26; 30M27; 30M28; 30M3; 30M30; 30M32; 30M33; 30M36; 30M38; 30M41; 30M43; 30M46; 30M48; 30M49; 30M5; 30M50; 30M54; 30M58; 30M6; 30M60; 30M61; 30M62; 30M63; 30M66; 30M6830M7; 30M72; 30M77; 30M78; 30M8; 30M81; 30M87; 30M88; 30M89; 30M9
50	49	50M10; 50M100; 50M1; 50M12; 50M13; 50M15; 50M16; 50M17; 50M25; 50M26; 50M28; 50M31; 50M33; 50M34; 50M35; 50M37; 50M38; 50M4; 50M41; 50M43; 50M45; 50M47; 50M48; 50M50; 50M52; 50M53; 50M55; 50M57; 50M58; 50M6; 50M63; 50M65; 50M66; 50M69; 50M70; 50M72; 50M73; 50M75; 50M76; 50M77; 50M79; 50M8; 50M80; 50M81; 50M82; 50M85; 50M95; 50M96 50M98
0 control	1	SIDI AISSA CONTROL
Total		97

Mx: Irradiated clone

Table 2. Climate data in Belkciri station (2012/2013/2014)

Year	Pluv.(mm)	T.A max (C°)	T.A min (C°)
2012	344,5	22,57	11,64
2013	781,2	22,15	12,82
2014	570	24,63	12,7
Average	565,23	23,11	12,38

Pluv. (Pluviometry); T. (Temperature); A. (Average)

2.3 Fruit Measures

Fruit weight was measured using a semi-analytical balance (testut NH1200), while fruit height and diameter were measured using millimetric calipers (absolute digimztic).

2.4 Number of Seeds

The number of seeds per fruit was evaluated (visual counting) after cutting the fruits in half and extracting the juice with a hand extractor. Presented data are means of 10 replications (fruits) and 3 seasons (2012/2013, 2013/2014 and 2014/2015).

2.5 Total Soluble Solids (TSS), Acidity

Maturity index and juice percentage were estimated according to the methods described by Bermejo (2011) [10]. °Brix was analysed using a refractometer (Atago Co. Ltd., Japan) and acidity was determined by titration with a 0.1 N NOAH solution using phenolphthalein as indicator [11].

2.6 Juice Content

Juice was obtained from the whole fruit using an electrical machine (Somatic-AMD, Spain). Juice content, as expressed in percentage, was estimated from samples of 10 fruits per clone and per harvest as following:

Juice content = (Juice weight /total weight of 10 fruit sample) x 100

2.7 Maturity Factor

Sweet oranges, mandarins, grapefruits, and pummelos are considered mature when their juice content and total soluble solids to acidity ratio have reached certain minimum limits for palatability. Total soluble solids constitute about 80 % sugars, 10 % acids, and 10 % nitrogenous compounds. Thus, an increase in total soluble solids (TSS) is generally accompanied by an increase in sugars and is strongly correlated to acidity [10].

Maturity index=total soluble solids (E) / titratable acidity (A)

2.8 Statistical Analysis

Collected data were subjected to one-way ANOVA analyses using SAS SOFTWARE, and Duncan’s multiple range test was applied at *P<0.05* to examine significant differences among means.

3. RESULTS

Fruit weight, size, acidity, maturity index and harvest time, as well as chemical and nutritional composition are all important qualities. Different fruit quality features have been evaluated in the irradiated varieties. They presented significantly different values compared to the control (see Table 3).

3.1 Average Weight of Fruit (g)

Fruit weight in the irradiated varieties ranged from 60.83 g to 103.75 g with an average of 82.09 g compared to 75,83 g in the control. Statistically there were significant differences between the 96 clones and the control. Mean separation enabled to classify these into 10 different groups with the most important one including 30M48 and showing higher average value than the control.

3.2 Seed Number per Fruit

The average number of seeds varied between 0,8 and 7 seeds per fruit. Similar to fruit weight there were significant differences between the irradiated clones. The statistical results showed four distinct groups, among which group D was the most important. This included two clones 50 M85 and 30 M68 with an average value ranging from 0.8 to 0.9 seeds per fruit (Fig. 2).

3.3 Juice Content

Juice content ranged from 37.64 % to 59.76% with an average percentage of 48%. Group A, including the clone 30M3, had a significantly higher juice content than the control, whereas the other groups had equal or lower values than the latter.

3.4 Acidity

Acidity varied from 0.65 to 0.7 with an average value of 0.81.Statistical analysis revealed significant differences among irradiated clones. According to acidity means they were separated into fourteen groups (Table 3). The leading group was H, which included the clone 30M24 and showed minimum values compared to the control.

3.5 Sugar Content

Sugar content oscillated between 6.96 and 8.9 showing an average value of 8.71.Based on the results of Duncan's multiple range test we distinguished eight groups. The groups A and AB, including the clones 50M11 and 50M50, resulted in maximum values relatively to control.

3.6 Maturity Index

Maturity index (E / A) ranged from 12.73 to 8.22 with an average of 11. There were significant differences between irradiated clones. Indeed, 10 different groups resulted from mean separation of collected data. The lowest values were recorded in the clone 50M98.

3.7 Seasonal Effects on the Variability of Fruit Quality

The irradiated clones grafted on macrophylla and planted in 2009 began to be productive in 2012. The results obtained from this experiment are presented in Table 4.

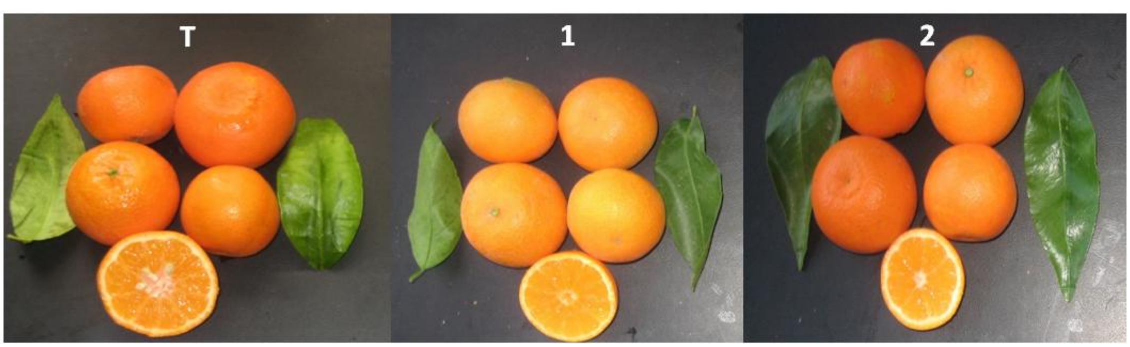

Fig. 1. Differences in aspect between seedless fruits of irradiated clones (1 and 2) and Sidi Aissa Control cultivar (T)
(T): Sidi Aissa Control; (1): 50 M85; (2): 30 M68

Table 3. Statistical results of variability in fruit quality criteria of irradiated Sidi Aissa clementine clones

	Fruit mean weight (g)	Fruit number seed	Juice content (%)	Sugar content (E)	Acidity (A)	E/A
Aissa (T)	75,833 cdefghijk	7 a	48,648 abcdefgh	8,900 abcd	0,846 abcdefgh	10,907 abcdef
30M22	83,333 abcdefghijk	3,383 abcd	49,760 abcdefgh	9,066 abc	0,833 abcdefgh	11,133 abcde
30M23	91,250 abcdefg	2,733 abcd	47,987 abcdefgh	8,666 abcd	0,820 abcdefgh	10,753 abcdef
30M24	85,833 abcdefghij	3,950 abcd	52,990 abcdefg	8,633 abcd	0,780 abcdefgh	11,303 abcde
30M17	66,250 hijk	2,683 abcd	55,660 abcdef	8,900 abcd	0,826 abcdefgh	11,207 abcde
30M1	90,417 abcdefg	1,533 bcd	49,563 abcdefgh	8,333 abcd	0,906 ab	9,430 cdef
30M103	76,667 cdefghijk	1,617 bcd	56,283 abcde	8,900 abcd	0,830 abcdefgh	10,950 abcdef
30M11	92,083 abcdefg	3,733 abcd	44,843 cdefgh	8,666 abcd	0,876 abcdefg	10,280 abcdef
30M13	87,917 abcdefghi	2,883 abcd	47,343 abcdefgh	8,800 abcd	0,806 abcdefgh	11,157 abcde
30M15	65,833 ijk	3,617 abcd	56,150 adcde	8,333 abcd	0,783 abcdefgh	11,190 abcde
30M2	81,667 abcdefghijk	2,350 abcd	47,080 abcdefgh	8,833 abcd	0,826 abcdefgh	10,987 abcdef
30M25	74,583 efghijk	1,567 bcd	51,657 abcdefg	9,000 abcd	0,813 abcdefgh	11,340 abcde
30M26	92,917abcdef	3,383 abcd	45,160 cdefghi	8,866 abcd	0,813 abcdefgh	11,143 abcde
30M27	74,583 efghijk	4,267 abcd	47,415 abcdefgh	8,833 abcd	0,828 abcdefgh	11,020 abcdef
30M28	83,333 abcdefghijk	1,500 bcd	50,177 abcdefgh	8,866 abcd	0,810 abcdefgh	11,430 abcde
30M3	79,583 bcdefghijk	4,917 ab	51,477 abcdefg	8,466 abcd	0,833 abcdefgh	10,547 abcdef
30M30	80,000 bcdefghijk	3,083 abcd	48,730 abcdefgh	9,066 abc	0,806 abcdefgh	11,380 abcde
30M32	72,917 efghijk	1,267 cd	51,790 abcdefg	8,933 abcd	0,796 abcdefgh	11,503 abcde
30M33	93,333 abcdef	3,383 abcd	45,727 cdefgh	8,500 abcd	0,836 abcdefgh	10,257 abcdef
30M36	88,333 abcdefghi	2,033 abcd	49,973 abcdefgh	9,133 abc	0,843 abcdefgh	11,273 abcde
30M38	98,333 abc	2,767 abcd	52,130 abcdefg	9,000 abcd	0,786 abcdefgh	11,550 abc
30M41	83,750 abcdefghij	1,267 cd	48,743 abcdefgh	8,600 abcd	0,796 abcdefgh	11,043 abcde
30M43	89,167 abcdefg	1,800 abcd	46,287 cdefgh	8,700 abcd	0,846 abcdefgh	10,340 abcdef
30M46	81,667 abcdefghijk	4,983 ab	41,110 gh	9,266 abc	0,773 bcdefgh	12,570 a
30M48	103,750 a	3,150 abcd	49,827 abcdefgh	9,166 abc	0,773 bcdefgh	12,043 abc
30M49	86,250 abcdefghij	1,983 abcd	44,673 cdefgh	9,033 abcd	0,726 h	12,653 a
30M5	83,333 abcdefghijk	3,700 abcd	45,383 cdefgh	8,900 abcd	0,788 abcdefgh	11,753 abc
30M50	85,833 abcdefghij	2,583 abcd	49,430 abcdefgh	8,933 abcd	0,790 abcdefgh	11,343 abcde
30M54	83,333 abcdefghijk	1,850 abcd	42,987 fgh	9,000 abcd	0,876 abcdefg	10,573 abcdef
30M58	82,500 abcdefghijk	3,133 abcd	54,270 abcdef	8,500 abcd	0,743 fgh	11,780 abc
30M6	79,583 bcdefghijk	2,800 abcd	49,337 abcdefgh	8,866 abcd	0,753 defgh	12,037 abc
30M60	73,750 efghijk	4,217 abcd	54,400 abcdef	9,000 abcd	0,870 abcdefgh	10,710 abcdef

	Fruit mean weight (g)	Fruit number seed	Juice content (%)	Sugar content (E)	Acidity (A)	E/A
30M61	75,833 cdefghijk	2,600 abcd	53,483 abcdefg	7,966 cde	0,923 a	8,687 ef
30M62	82,083 abcdefghijk	3,600 abcd	50,067 abcdefgh	9,033 abcd	0,850 abcdefgh	10,797 abcdef
30M63	85,000 abcdefghij	1,683 bcd	46,867 bcdefgh	8,633 abcd	0,783 abcdefgh	11,363 abcde
30M66	77,083 cdefghijk	2,467 abcd	54,053 abcdef	8,600 abcd	0,773 bcdefgh	11,270 abcde
30M68	72,083 efghijk	0,800 d	52,997 abcdefg	8,866 abcd	0,790 abcdefgh	11,420 abcde
30M7	69,583 ghijk	2,500 abcd	49,170 abcdefgh	8,333 abcd	0,886 abcdef	9,557 bcdef
30M72	78,333 bcdefghijk	4,583 abc	52,397 abcdefg	8,233 abcd	0,820 abcdefgh	10,200 abcdef
30M77	91,667 abcdefg	3,667 abcd	52,810 abcdefg	8,966 abcd	0,783 abcdefgh	11,797 abc
30M78	73,750 efghijk	2,917 abcd	50,527 abcdefg	8,633 abcd	0,823 qbcdefgh	10,670 abcdef
30M8	74,167 efghijk	2,833 abcd	47,863 abcdefgh	8,233 abcd	0,766 bcdefgh	11,097 abcde
30M81	78,333 bcdefghijk	3,533 abcd	51,403 abcdefg	7,7000 ed	0,890 abcde	8,697 def
30M87	77,917 bcdefghijk	4,500 abc	53,260 abcdefg	8,900 abcd	0,826 abcdefgh	11,173 abcde
30M88	73,333 efghijk	3,133 abcd	48,403 abcdefgh	9,066 abc	0,793 abcdefgh	11,680 abc
30M89	79,167 bcdefghijk	2,983 abcd	52,767 abcdefg	8,566 abcd	0,870 abcdefgh	10,247 abcadef
30M9	82,917 abcdefghijk	2,383 abcd	46,447 bcdefgh	9,266 abc	0,860 abcdefgh	10,840 abcdef
50M10	75,417 defghijk	2,517 abcd	59,767 a	9,066 abc	0,796 abcdefgh	11,563 abc
50M100	65,000 jk	3,800 abcd	45,293 cdefgh	8,800 abcd	0,840 abcdefgh	10,697 abcdef
50M11	81,250 abcdefghijk	2,467 abcd	49,793 abcdefgh	9,400 a	0,813 abcdefgh	12,737 a
50M12	88,750 abcdefgh	4,983 ab	49,663 abcdefgh	9,166 abc	0,830 abcdefgh	11,270 abcde
50M13	76,250 cdefghijk	2,833 abcd	50,510 abcdefg	8,766 abcd	0,793 abcdefgh	11,197 abcde
50M15	78,750 bcdefghijk	2,400 abcd	55,867 abcde	8,900 abcd	0,770 bcdefgh	11,850 abc
50M16	88,750 abcdefgh	3,800 abcd	50,713 abcdefg	8,500 abcd	0,786 abcdefgh	11,093 abcde
50M17	74,583 efghijk	2,000 abcd	45,917 cdefgh	8,766 abcd	0,766 bcdefgh	11,600 abc
50M25	79,167 bcdefghijk	1,467 bcd	49,827 abcdefgh	8,366 abcd	0,830 abcdefgh	10,187 abcdef
50M26	85,417 abcdefghij	2,783 abcd	51,750 abcdefg	8,033 bcde	0,840 abcdefgh	9,650 bcdef
50M28	79,583 bcdefghijk	1,933 abcd	49,500 abcdefgh	8,300 abcd	0,863 abcdefgh	9,940 abcdef
50M31	93,667 abcdef	1,250 cd	52,273 abcdefg	9,100 abc	0,830 abcdefgh	10,970 abcdef
50M33	90,833 abcdefg	1,500 bcd	46,587 bcdefgh	8,033 bcde	0,840 abcdefgh	9,913 abcdef
50M34	79,167 bcdefghijk	2,850 abcd	45,977 cdefgh	9,266 abc	0,880 abcdef	10,913 abcdef
50M35	85,000 abcdefghij	4,517 abc	55,690 abcdef	8,933 abcd	0,773 bcdefgh	11,557 abc
50M37	84,167 abcdefghij	2,100 abcd	37,647 h	8,233 abcd	0,886 abcdef	9,670 bcdef
50M38	83,333 abcdefghijk	3,050 abcd	46,727 bcdefgh	8,633 abcd	0,800 abcdefgh	11,123 abcde
50M4	77,083 cdefghijk	2,267 abcd	57,360 abc	9,166 abc	0,896 abcd	10,253 abcdef
50M41	80,000 bcdefghijk	3,683 abcd	49,123 abcdefgh	8,200 abcd	0,753 defgh	11,090 abcde

	Fruit mean weight (g)	Fruit number seed	Juice content (%)	Sugar content (E)	Acidity (A)	E/A
50M43	77,083 cdefghijk	2,433 abcd	47,853 abcdefgh	8,700 abcd	0,836 abcdefgh	10,537 abcdef
50M45	71,250 fghijk	2,000 abcd	53,907 abcdef	8,600 abcd	0,773 bcdefgh	11,193 abcde
50M47	73,750 efghijk	2,000 abcd	56,590 abcd	8,733 abcd	0,860 abcdefgh	10,330 abcdef
50M48	82,500 abcdefghijk	2,117 abcd	52,047 abcdefg	8,733 abcd	0,760 cdefgh	11,730 abc
50M50	97,917 abcd	5,300 ab	46,403 bcdefgh	9,333 ab	0,746 efgh	12,720 a
50M52	75,417 defghijk	1,583 bcd	59,130 ab	8,433 abcd	0,783 abcdefgh	10,983 abcdef
50M53	86,667 abcdefghij	4,183 abcd	49,533 abcdefgh	8,933 abcd	0,766 bcdefgh	11,767 abc
50M55	73,333 efghijk	3,500 abcd	47,977 abcdefgh	8,933 abcd	0,820 abcdefgh	11,130 abcde
50M57	78,750 bcdefghijk	1,667 bcd	49,923 abcdefgh	8,766 abcd	0,763 bcdefgh	11,530 abcd
50M58	75,417defghijk	1,700 abcd	47,990 abcdefgh	8,766 abcd	0,793 abcdefgh	11,157 abcde
50M6	82,500 abcdefghijk	3,100 abcd	50,457 abcdefg	8,466 abcd	0,766 bcdefgh	11,313 abcde
50M63	77,083 cdefghijk	2,233 abcd	50,330 abcdefg	8,866 abcd	0,763 bcdefgh	11,627 abc
50M65	85,833 abcdefghij	1,283 cd	49,920 abcdefgh	8,500 abcd	0,733 gh	11,673 abc
50M66	89,583 abcdefg	2,350 abcd	46,687 bcdefgh	8,633 abcd	0,796 abcdefgh	10,833 abcdef
50M69	94,443 abcde	3,557 abcd	48,897 abcdefgh	8,633 abcd	0,750 efgh	11,623 abc
50M70	100,000 a	1,733 abcd	43,590 efgh	9,300 abc	0,763 bcdefgh	12,403 ab
50M72	91,667 abcdefg	4,333 abcd	53,190 abcdefg	8,866 abcd	0,843 abcdefgh	10,600 abcdef
50M73	89,167 abcdefg	2,900 abcd	51,540 abcdefg	8,200 bcde	0,833 abcdefgh	10,320 abcdef
50M75	97,917 abcd	2,217 abcd	46,017 cdefgh	9,066 abc	0,790 abcdefgh	11,477 abcde
50M76	90,833 abcdefg	2,883 abcd	49,700 abcdefgh	8,766 abcd	0,763 bcdefgh	11,603 abc
50M77	85,833 abcdefghij	2,100 abcd	52,797 abcdefg	8,800 abcd	0,770 bcdefgh	11,550 abc
50M79	84,583 abcdefghij	1,917 abcd	47,507 abcdefgh	8,533 abcd	0,796 abcdefgh	11,013 abcdef
50M8	83,750 abcdefghij	1,717 abcd	57,113 abc	8,033 bcde	0,846 abcdefgh	9,587 bcdef
50M80	90,833 abcdefg	2,500 abcd	45,087 cdefgh	6,9667 e	0,896 abcd	8,223 f
50M81	73,750 efghijk	1,683 bcd	45,163 cdefgh	8,366 abcd	0,756 cdefgh	11,333 abcde
50M82	89,583 abcdefg	1,550 bcd	50,427 abcdefg	9,066 abc	0,903 abc	10,200 abcdef
50M85	72,500 efghijk	0,883 d	55,030 abcdef	8,700 abcd	0,810 abcdefgh	10,720 abcdef
50M95	80,000 bcdefghijk	1,517 bcd	43,873 defgh	8,366 abcd	0,823 abcdefgh	10,437 abcdef
50M96	83,333 abcdefghijk	3,633 abcd	45,607 cdefgh	8,866 abcd	0,753 defgh	11,943 abc
50M98	60,833 k	2,767 abcd	47,183 abcdefgh	8,600 abcd	0,863 abcdefgh	10,183 abcdef

Table 4. Variations of fruit quality parameters over the years of experimentation

Season	Fruit weight (g)	Seeds number per fruit	Juice content (%)	Sugar content	Acidity	E/A
2012/2013	116,258 a	4,9487 a	57,7837 a	8,00606 c	0,707879 c	11,4046 b
2013/2014	57,273 c	4,3869 b	36,2447 c	8,49293 a	0,962222 a	8,903 c
2014/2015	72,384 b	3,898 c	55,1352 b	9,66667 b	0,768182 b	12,7158 a
Mean	81,97	2,758	49,72	8,72	0,81	11,007
P>F	<,0001	<,0001	<,0001	<,0001	<,0001	<,0001

Statistical results showed a great variation among the clones studied over the years which was reflected by a significant effect (*P*< 0,001) of the factor 'season' on all studied criteria. On one hand, we observed a decrease in average fruit weight from 116 (g) in 2013 to 57 (g) in 2015, this was concomitant with a reduction in the number of seeds per fruit (from 5 to 0, 9) and in juice content (from58 to 36). On the other hand, an increasing tendency in sugar content was observed for the same period (between 2013 and 2015). Concerning the percentage of citrate (acidity) and the maturity index minimum values (values of interest) were recorded in 2015.

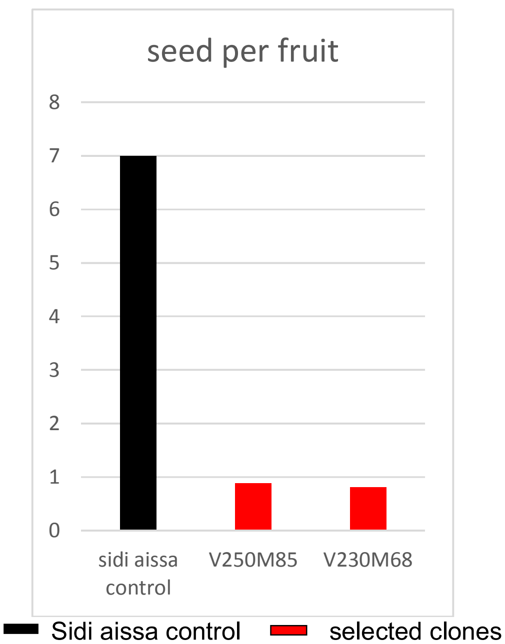

Fig. 2. Reduction of seed number per fruit between irradiated clones and sidi aissa control

4. DISCUSSION

Seedlessness is an important economic trait relating to fruit quality and gamma irradiation is a common technique used to obtain this criteria in citrus fruit. In the present study, we compared 96 irradiated clones and the control with respect to many fruit quality parameters. An interesting discovery emerging from the present study is that we observed that irradiation is a valuable tool to obtain seedless cultivars from seedy ones. Our first observations indicated a reduced number of seeds in some clones compared to the control. Moreover, the clones 50M85 and 30M68 were characterized by aspermic fruits. Similar observations were reported by several authors [12,13,14]. Regarding the possible effects of gamma mutagenesis irradiation on pomological fruit quality, our results are in agreement with those of previous studies [10,13]. Trees subjected to gamma radiation at 50 Gy dose, 50M11, 50M50 and 50M70 resulted in a higher sugar content than the control, similarly to the findings of Goldenberg et al. [15]. Concerning fruit acidity, several clones showed a lower acidity level than Sidi Aissa control. In the same line, Yen [16] showed that irradiated buds of Foster grapefruits gave less acidic fruit mutants. Varietal improvement in apples was achieved through physical mutagenesis leading to reduced acidity and bitterness [17]. Juice content of the fruits is an important selection criterion for marketing purposes. According to the values determined by OECD (Organisation for economic co-operation and development), this parameter is set at 40% for clementine varieties [18]. In our study we detected 14 irradiated clones that revealed a juice rate much higher than the control, unlike the preliminary studies [12]. Knowing that maturity, evaluated by the coefficient E / A, is a parameter that responds to genetic variations between varieties, the irradiated buds of Kutdiken lemon gave mature mutants earlier or later than the control [19]. *Nero*, a clementine Nules mutant obtained by irradiation, was distinguished by its late maturity compared to its control (mid-November vs. mid-January) [20] In our study, three Sidi Aissa clones among the 96 studied (50M80, 30M61 and 30M81) had a lower maturity index than the control. Similar results were reported by Bermejo 2012 [12].

Finally, we should note that the effect of irradiation dose could increase or decrease the frequency of character variability. The optimal doses that can be envisaged for a slow seed irradiation are very much superior to the doses commonly used for bud mutagenesis [21,22]. In this regard, author of several publications reported that buds taken from irradiated trees by 60Co gamma ray at a dose of 80Gy at 2.5 Gy / min are re-irradiated with the same dose, confirming that recurrent irradiation could enhance the frequency of mutation.

5. CONCLUSION

Gamma ray irradiation of buds is a classical method for varietal creation. Using this method in our study, we intended to improve or diversify one or some of the fruit qualities. The results obtained showed that all irradiated clones presented a lower number of seeds per fruit than Sidi Aissa control. In short, fruit quality was affected differently, some clones showed no differences from the control, while others showed great and interesting variances such as 30M68 and 50M85. Mutations may lead to the creation of an increasing number of new variants. Not only do these mutants give birth to new varieties that may enrich germplasm collections, but they can also be included as parents in cross-breeding programs. In this sense gamma irradiation may be a promising tool for the induction of genetic variability with regards to certain criteria related to fruit quality. Our results have particularly demonstrated that it has a considerable effect on reducing the number of seeds and favoring aspermy. However, to confirm and support these findings, obtained clones should be evaluated for another two years before being registered to the official catalog.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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