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MORPHOLOGICAL CHARACTERIZATION, POLLEN GRAIN FERTILITY AND SOME CHEMICAL CHARACTERS OF SELECTED MANDARIN (*Citrus* Spp.) VARIETIES

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ABSTRACT

The present research aimed to characterize physical and morphological features of thirteen varieties of mandarin as one of the National Gene Bank and Genetic Resources (NGBGR) objectives in Egypt which include also, collection, conservation, characterization, and evaluation of agricultural genetic resources. The present study investigates the morphological characterization, pollen grain fertility and sterility as well as fruit chemical characterization (vitamin C, PH, T.S.S and acidity) of 13 mandarin varieties (*Citrus sp*.).Including Balady, Malawy, Chine mandarin, Clementine, Celiopatra, Centra, Satsuma, Tangerine Dancy, El-Shorbagee, Sonbol, Sayed Marri, Abd El-Razik and Aswan. The Physical and morphological characterization descriptors revealed differences among varieties. Characterization of tree shape resulted in eight obloid varieties and five were spheroids. Leaf lamina shape morphological characteristic studies showed one elliptic variety, three ovate and 9 lanceolate. Fruit shape studies indicated that five varieties were pyriforum, five obloid and three were spheroid. Fruit apex shape demonstrated that seven varieties were depressed, while six were truncate. Differences in flavedo (skin) color among varieties revealed that seven were orange; two were dark orange and one variety for each of (light orange, green yellow, dark yellow and yellow). As to the color of the pulp (flesh), results indicated that two varieties were yellow, 10 were orange and one was orange-red. The average number of carples per fruit ranged 10-14 carple in 9 varieties and from 5-9 carple in four varieties. The average number of seeds per fruit revealed that five varieties had ten to nineteen seeds; three varieties hold 20-50 seeds and two varieties contained five to nine seeds, one variety included one to four seeds, while two varieties had no seeds. Other morphological studies are investigated and will be presented. Fruit chemical characterization disclosed that Malawy had the highest vitamin C concentration (44.82 mg/100 ml) whereas both of Celiopatra and Satsuma showed the lowest concentration (15.0 and 14.0 mg/100 ml, respectively).The greatest pH was determined in Abd El-Razik (pH 3.91) variety in contrast to the more acidity (pH 3.12-3.77) found in El-Shorbagee and Chine mandarin juice, respectively. The best total soluble solids were determined in the Abd El-Razik (15.1%), whereas the least ones were found in Satsuma (9.13%). Chine mandarin recorded the highest titratable acidity of 3.88% in contrast to 0.67 % for Sonbol. Studies of pollen grain fertility showed that Celiopatra and Sayed Marri varieties gave the highest pollen fertility (99.84%and 99.44% respectively). Contrarily, the highest pollen sterility was found in Clementine variety (8.41%).

INTRODUCTION

Mandarins (*Citrus spp*.) are the second most important group of citrus plants in the worldwide and the mandarin group is comprised of the numerous species as well as intergeneric and inter

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specific hybrids which made them the most phenotypically heterogeneous of Citrus **(Moore, 2001)**, also it is one of the most important subtropical fruit crops grown in Egypt for their unique flavor and nutritional values. The area harvested (ha) during 2005 year was 38000 ha. The average productivity per hectare during the same year 17, 500, 00 kg/ ha **(FAO, 2007)**. The mandarin culture and its commercialization are restricted to a few varieties, offered in a small period during the year, causes the needs to import the product. For these reasons, the breeders should be investigate the available genetic resources (species and varieties), and supply the market demands.

Conservation, characterization and evaluation were one of the National Gene Bank and Genetic Resources (NGBGR) objectives in Egypt. Correct classification and identification of accessions in a germplasm bank allows solving management problems i.e., to avoid duplication in the exchange and in the conservation of the germplasm within the bank specially, indigenous and locally adapted varieties because of their genetic variation and their rapid disappearing rate through replacement by high yielding varieties.

The characterization of cultivar is an essential stage in the certification program, improvement and conservation of germplasm, and monitoring of the genetic quality **(IPGRI, 1999)**. In this regard, characterization descriptors are comprised of highly heritable qualitative traits that can be equally expressed in all conditions **(IPGRI, 1999)**. It may also include a limited number of additional traits thought desirable by a consensus of users. This type of analysis is simple and of less cost **(Ballve *et al* 1997)**, even so it presents limitation related to the characters that present inheritance additive, which highly are influenced by the environment, and to cultivating them with great phenotypic similarity **(Olivera *et al* 2000).**

The percentages of viable and aborted pollen were estimated by testing their stain ability in acetocarmine staining. Testing for pollen grain viability considered to be pollen fertility and pollen sterility. The cultivars with normal fertility have higher proline concentrations than those with partial or complete sterility. The proline concentrations of anther is positively correlated with the pollen germination percentage. Vitamin C is the most important nutrient in citrus fruit juice; it should be present in great concentrations in the form of ascorbic acid. Vitamin C is a natural antioxidant that may inhibit the development of major oxidative human conditions **(Omaye and Zhang, 1998).**

Citrus fruit are one of dietary sources of carotenoids, pro-vitamin A and vitamin C **(Gardner *et a*l 2000 and Gil-Izquierdo *et al* 2001**).

Soluble solids content (SSC) is a major characteristic used for assessing citrus fruit quality. Sweetness has high correlation with SSC, pH, and sugars (fructose, and glucose) **(Fernandez *et al* 2004). Caro and Joas (2005)** suggested that acid concentration passively increases as a result of water loss during storage. The increases of soluble solids content and titratable acidity may relate to fresh weight loss during storage.

The objectives of the current work were to study the morphological characteristics of 13 mandarin varieties, chemical analysis and their pollen grain viability.

MATERIALS AND METHODS

1. Morphological Characterizatio*n*

Morphological characterization and pollen grain viability testing of the 13 mandarin varieties, under the present investigation, were carried out on 18-20 years old mandarin trees grafted on sour orange grown at the Faculty of Agriculture Research Farm, Zagazig University during 2004 and 2005 years. Trees were spaced at 3x5 meters apart.

The morphological characteristics used to characterize and discriminate the 13 mandarin varieties were based on those previously prescribed for citrus by the International Plant Genetic Resources Institute **(IPGRI, 1999)** taking into consideration all the precautions reported. In this respect, 15 quantitative and 23 qualitative morphological characteristics were selected for the present investigation.

The study was performed using three trees for each variety; each tree was considered a replicate. In this respect, tree shape and growth habit were characterized in the natural state and immediately after harvest.

Thirty mature and fully developed leaves per tree (mature leaves from one year old branches) were collected and characterized for intensity of green color of leaf blade, leaf lamina attachment, leaf lamina length and width, ratio of leaf lamina length/width, leaf lamina shape, leaf lamina margin, leaf apex, and absence or presence of petiole wings **(Fig. 1)**.

Data were recorded for flower pedicel length, length of anthers relative to stigma, number of petals per flower, petal length and width.

All observations on the fruit and its related parts were made at the optimum ripening stage. Fruit characteristics were observed on 10 variety typical fruits per each tree of the three replication trees. Data were documented for fruit weight, diameter, length, and shape. Records also included shape of fruit (base and fruit apex), fruit flavido (skin) color, texture of epicarp surface, adherence of mesocarp to endocarp, density of oil gland on fruit surface and fruit rind thickness. The study comprised also number of carples per fruit, thickness of carple walls, color of fruit pulp and juice content in endocarp.

Fully developed seeds were extracted from 10 fully ripped fruits taken from each tree of the three replications. In this respect, average number of seeds per fruit, seed shape, seed surface, seed color, seed length, seed width and seed weight, **(Fig. 1)**.

2. Pollen Grain Viability

Ten randomly selected whole flowers replicate of the studied varieties were collected at an appropriate stage. Flowers were immediately fixed in a 3 alcohol: 1 acetic acid solution for 24 hours. Then flowers were washed several times with distilled water followed by storage in 70 % ethanol. Squash preparations of pollen mother cells (PMC's) were made in aceto-carmen as described by **Fayed *et al* (1984)**. Twenty slides were prepared using the previously randomly selected 10 flowers for each variety.

The percentage of viable and aborted pollen grains was estimated by testing their satiability in the aceto-carmen staining. Round and dark stained pollen grains were considered viable and functional, while non-viable ones were the shriveled or the light stained pollen grains as described by **Fayed (1990).**

All data were arranged in a randomized complete block design and were statistically analyzed according to **Snedecor and Cochran (1982).** The Fishers protected least significant difference (LSD) at $P \leq 0.05$ was employed to separate the treatment means.

Chemical analysis

1. pH

10 gm. of mandarin juice was blended with 20 ml deionized water. The pH was measured with a cyberscan pH meter **(Sanchez-Moreno *et al* 2003).**

2. Titratable acidity

After determining pH, the solution was titrated with 0.1 M NaOH to pH 8.1, monitoring with an electrode as grams of citric acid per Kilogram fresh weight (fw) **(Sanchez-Moreno *et al* 2003).**

3. Soluble solids contents (SSC)

Soluble solids were measured from mandarin juice samples with an Atago digital refractometer PAL-1 (Tokyo, Japan). Results are reported as %.

4. SSC/Acid ratio

It was calculated by dividing of soluble solids contents on acidity

5. Vitamin C

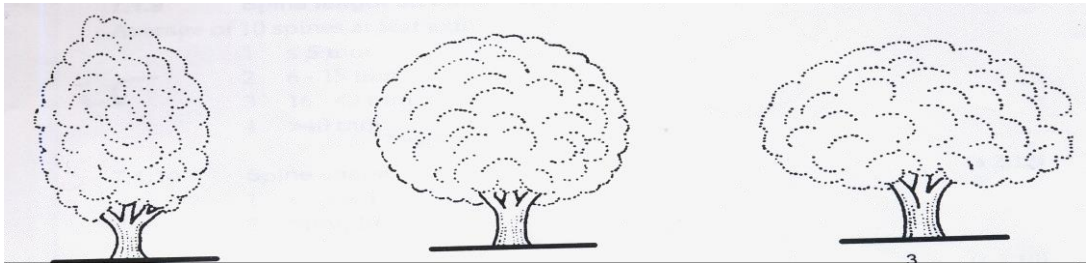
Vitamin C was determined by HPLC. A volume of 50 ml of each mandarin juice was homogenized with 40 ml of an extraction solution (30 g 1^{-1} meta-phosphoric acid + 80 g 1^{-1} % acetic acid). The resulting mixture was filtered under suction and adjusted up to 100 ml with distilled water. Samples were filtered through a 0.45 µm membrane filter and duplicates of 20µm for each extract were analysed by HPLC. Results are expressed as milligrams of ascorbic acid per 100 ml juice.

Separation of ascorbic acid was performed by HPLC using a Hypersil BDS C8 (5 µm) stainless steel column (250 mm x 4.6 mm) (Thermo Electron, United Kingdom).

The solvent system used was an isocratic gradient of a solution 70% Buffer (0.85 % v/v H_2SO_4 in 17.5 mM KH_2PO_4, pH 1.8) and 30% Methanol. The flow rate was fixed at 1.5 ml/min. A UV- vis detector was set at 245 nm; chromatographic data and UV-vis spectra were collected, stored and integrated using a chromostar light software. The calibration curve was built with one concentration level an ascorbic acid standard solution (100mg ml^{-1} in a solution 30 g 1-1 meta-phosphoric acid + 80 g 1-1 % acetic acid) **(Sanchez-Moreno *et al* 2003)**.

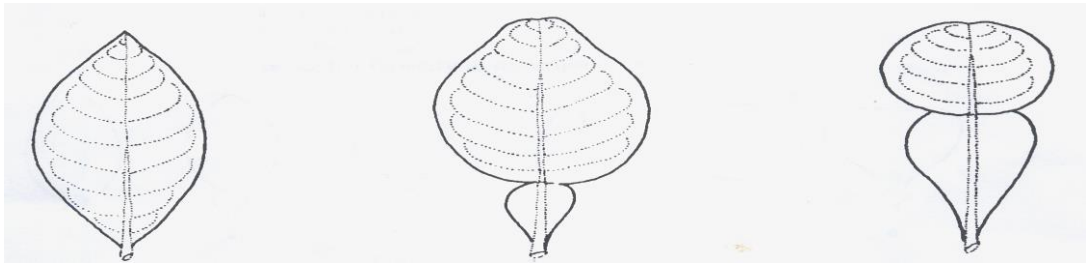
Figure1. Some characters utilized in morphological characterization of thirteen mandarin accession

Tree shape



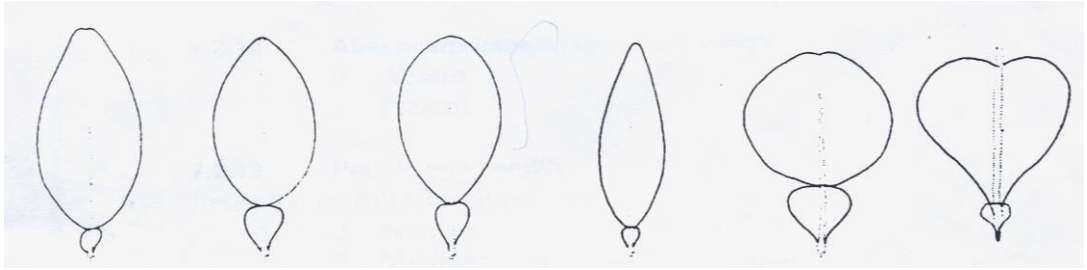
Erect Spreading Drooping

Leaf lamina attachment



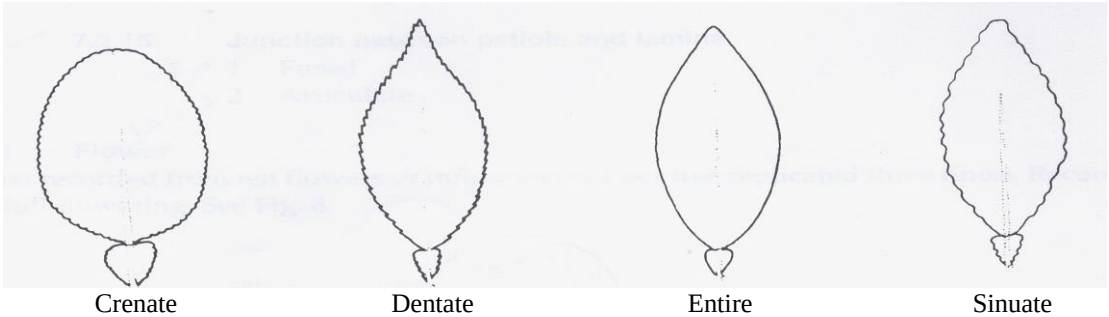
Sessile Brevipetiolate Longipetiolate

Leaf lamina shape



Elliptic Ovate Obovate Lanceolate Orbicular Obcordate

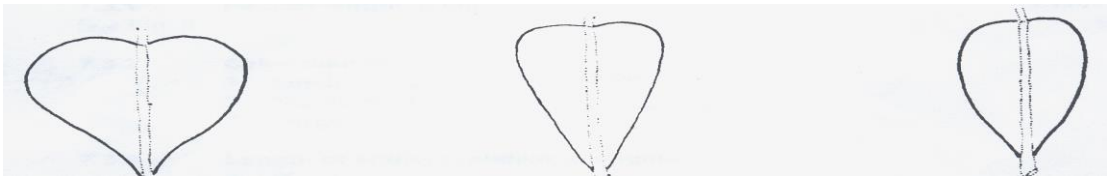
Leaf lamina margin



Crenate Dentate Entire Sinuate

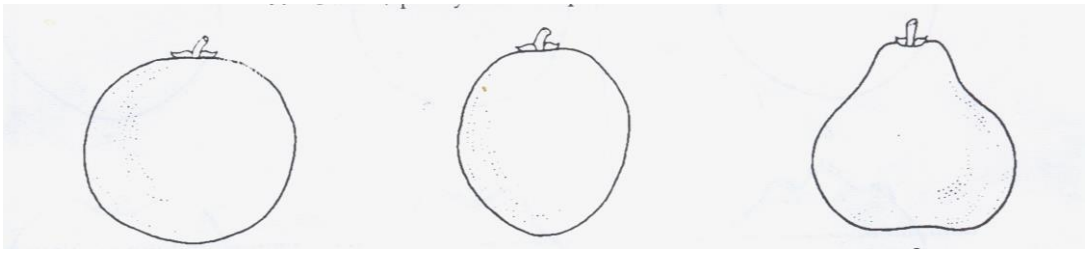
Continued Fig. 1.

Petiole wing shape

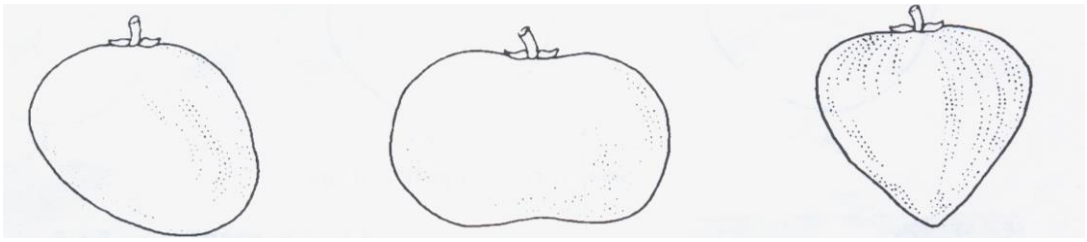


Obcordate Obdeltate Obovate

Fruit shape

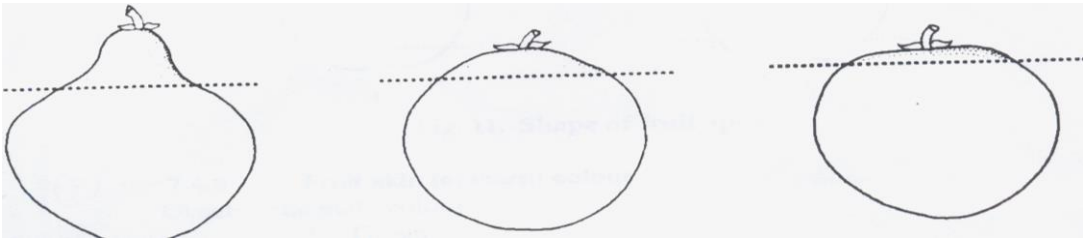


Spheroid Ellipsoid Pyriform

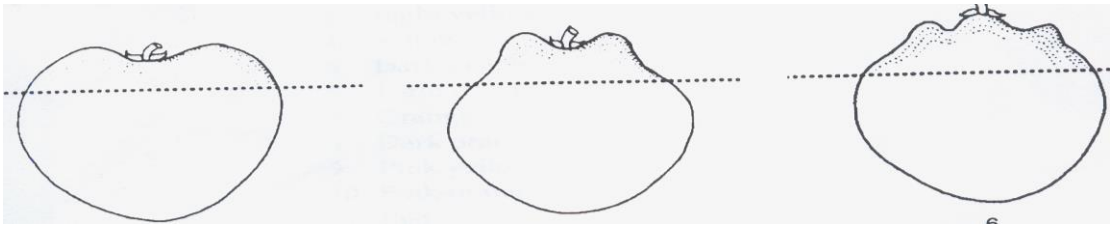


Oblique Obloid Ovoid

Shape of fruit base



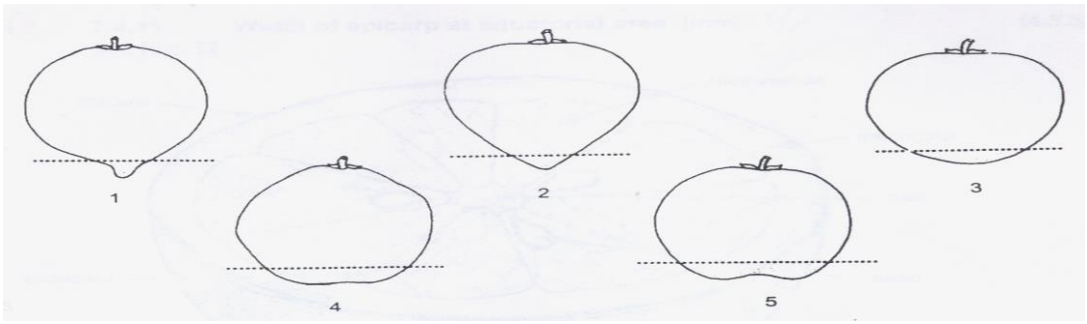
Mammiform Acute Rounded



Concave Concave collared Collared with neck

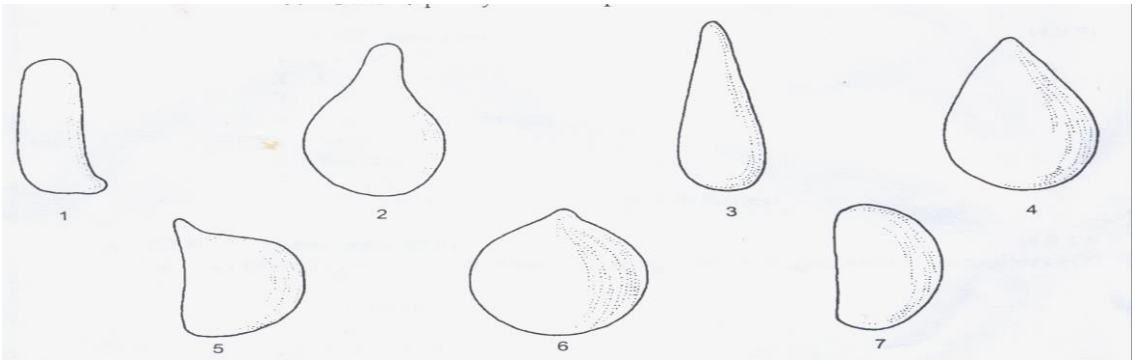
Continued Fig. 1.

Fruit of fruit apex



1. Mammiform 2.Acute 3.Rounded 4.Truncate 5. Depressed

Seed shape



1. Fusiform 2.Clavate 3. Cuneiform 4.Ovoid
5. Semi-deltoid 6.Spheroid 7. Semi-spheroid

RESULTS AND DISCUSSION

1. Morphological Characterization.

1.1 Quantitative traits

The leaves and flowers characters of the mandarin varieties were presented in **Table (1)**. Results illustrated that Sonbol variety showed the highest leaf lamina length of (9.4) cm while the lowest value was presented by Sayed Marri variety (5.7 cm). Leaf lamina width exhibited high variable among the varieties, Chine mandarin variety displayed the greatest Leaf lamina width (3.65 cm) followed by Sonbol (1.39 cm.). The lowest Leaf lamina width value was obtained by Sayed Marri (1.6 cm) .The rest of varieties demonstrated midway values ranged 2.25-1.83 cm. The highest ratio of leaf lamina length/width was showed by variety Sayed Marri (3.56 cm) followed by Malawy and Balady (3.3) varieties. The lowest ratio was characterized in Sonbol (1.3 cm**) Table (1).**

The results showed that Malawy variety revealed the highest flower pedicel length (0.9 cm) followed by Satsuma, Sonbol and Tangerine

Table1. Quantitative characteristics of leaves and flowers of 13 mandarin varieties

Variety	Leaf lamina length (cm)	Leaf lamina width (cm)	Ratio of leaf lamina length/width	Flower pedicel length (cm)	Number of petals per flower	Petal length	Petal width.
Clementine	7.38 D	2.70 G	2.73 D	0.3 E	5 A	0.9 K	0.36 G
Celiopatra	7.2 E	3.05 E	2.36 F	0.4 D	5 A	1.15 I	0.44 E
Centra	6.6 H	2.98 F	2.21 I	0.4 D	5 A	1.10 J	0.40 F
Satsuma	7.2 E	2.25 C	2.25 H	0.6 B	5 A	1.76 B	0.62 B
Abd El- Razik	5.77 K	2.6 H	2.22 I	0.3 E	4 B	1.22 G	0.48 D
Sonbol	9.4 A	1.39 B	1.3 J	0.6 B	5 A	1.7 C	0.56 C
Tangerine Dancy	7.1 F	2.29 D	2.29 G	0.6 B	5 A	1.18 H	0.48 D
Malawy	8.1C	2.4 I	3.3 B	0.9A	5 A	1.8 A	0.94 A
Chine mandarin cv	8.6 B	3.65 A	2.36 G	0.20 G	5 A	1.30 F	0.50 D
Aswan	6.35 I	2.4 I	2.64 E	0.4 D	5 A	1.45 D	0.55 C
Sayed Marri	5.70 L	1.6 L	3.56 A	0.4 D	5 A	1.40 E	0.50 D
El-Shorbagee	7.07 G	2.3 J	3.03 C	0.3 F	5 A	1.35 F	0.45 E
Balady	6.07 J	1.83 K	3.3 B	0.30 F	5.0 A	1.3 F	0.50 D

Means followed by the same letter within the same column are not significantly different (P≤0.05; LSD test)

Dancy varieties (0.6 cm). The lowest one was exhibited by the Chine mandarin (0.20 cm) variety. Whereas the rest of the varieties illustrated intermediate values.

There was no significant differences among the investigated varieties for number of petals per flower (5 petals per flower) except, Abd El-Razik which showed four petals per flower. The petal length was different among the varieties where the Malawy variety presented the highest measurement (1.8 cm). On the contrary, the Clementine, Celiopatra and the Centra varieties had the lowest petal length (0.9, 1.15 and 1.10 cm) respectively.

The varieties Malawy and Satsuma presented the highest petal width (0.94 and 0.62cm) respectively. On the other hand, the respective lowest values of petal width 0.36 and 0.40 cm were showed by Clementine and Centra varieties.

Table (2) presents the quantitative characteristics of fruit and seed. Malawy variety showed the highest significant fruit weight (440.8 g) followed by Abd El-Razik (184.7g) variety. On the other hand, Clementine variety had the lowest fruit weight (81.1 g). The rest of the varieties gave intermediate fruit weights.

Aswan, Satsuma and Malawy exhibited the greatest fruit diameter (7.90, 7.28 and 7.10 cm) respectively. Whilst, Sonbol, Clementine and Tangarin Dancy varieties displayed the smallest fruit diameter (5.44, 5.50 and 5.9 cm) respectively. The highest fruit length (7.40cm) was demonstrated by Malawy variety.The lowest fruit length value (4.60cm) was achieved by Celiopatra variety. The rest of varieties had midway values ranged from 7.0 to 4.83 cm.

Aswan Variety exhibited the most significant diameter of fruit axis (3.10 mm). On the contrary Chine mandarin variety exhibited the least diameter of fruit axis (0.70 mm). However, no significant differences were obtained among Satsuma, Abd El-Razik and Tangarine Dancy (2.0 mm). Satsuma, Abd El-Razik and Sayed Marri varieties exhibited the greatest width of epicarp at equatorial area (0.40 mm). Conversely, varieties of Celiopatra, Centra, Sonbol and Tangarine Dancy revealed the lowest value (0.10 mm).The quantity of juice content in fruit was characterized with variability among varieties, Malawy variety displayed the highest one (75.0 ml/Fr), while Clementine was the least juice content (25.0

ml/Fr). The other varieties showed intermediate juice content ranged from 71.6 to 33.75 ml/Fr**. Table (2).**

Satsuma variety fruits were distinguished by tree seeds. On the other hand, the highest seed length value (1.48 cm) was obtained by Abd El-Razik variety, whereas both of Malawy and Balady varieties showed the least seed length (1.13 cm). Also, varieties Abd El-Razik and Balady showed the highest seed width (0.70 cm), whereas the least value was observed by Malawy variety. Also, at the same trend the largest average seed weight (0.22 g) was detected in the variety Abd El-Razik, while the Chine mandarin variety showed the least seed weight (0.10 g).

1.2 Qualitative traits

Table (3) presents the qualitative traits of tree, leaf and flower of the 13 mandarin varieties under the study. Tree shape were classified as obloid in the varieties Clementine, Celiopatra, Satsuma, Abd El-Razik, Chine mandarin, Aswan and Balady, whereas the rest of the varieties showed spheroid tree shape. The characterization degrees of tree growth habit included erect, spreading or drooping growth habit. None of the varieties under the study demonstrated erect growth habit. The tree growth habit of the varieties Clementine, Celiopatra, Sonbol, Malawy, Sayed Marri and El-Shorbagee was spreading and the others were drooping.

The intensity of green color of leaf blade was recorded on fully developed leaves. All varieties showed green color of leaf blade except for the Abd El-Razik, Sonbol and Malawy varieties which showed dark color **Table (3)**.The leaf lamina attachment (length of petiole relative to length of leaf lamina) appeared that, the most frequent of the varieties showed sessile leaf lamina attachment except for the varieties Centra, Satsuma, Sonbol, Malawy and Chine mandarin were Brevipetiolate.

Concerning leaf lamina shape, most of the studied varieties exhibited lanceolate except for the varieties of Satsuma, Tangarine Dancy, and Chine mandarin that showed ovate leaf, while Centra was elliptic lamina shape. None of the varieties had crenate leaf lamina margin. The varieties Celiopatra, Centra, Abd El-Razik, Chine mandarin, Aswan and El- Shorbagee showed dentate leaf lamina margin, whereas the varieties Satsuma, Sonbol, Tangarine Dancy and Malawy demonstrated sinuate. The rest of the studied varieties exhibited entire leaf lamina margin. The categories under which leaf apex of the different varieties was evaluated were attenuate, acuminate, acute obtuse, rounded or emarginated leaf apex. None of the studied varieties showed attenuate, emarginated or rounded leaf apex. The varieties Centra, Satsuma, Abd El-Razik and Tangarine Dancy proved obtuse for leaf apex, while Clementine showed acute leaf apex. However, the rest of the varieties, revealed acuminate leaf apex. Most varieties illustrated the absence of petiole wings excluding, Centra, Sonbol, Tangarine Dancy, Malawy and Chine mandarin varieties which characterize with the presence of petiol wings **Table (3).**

The length of anthers relative to stigma was estimated for the different varieties. The Centra, Sonbol, Malawy, Chine mandarin, Aswan, Sayed Marri and El-Shorbagee exhibited medium anther than stigma. On the other hand Celiopatra, Satsuma,Abd El-Razik and Tangarine Dancy showed short anther relative to stigma. While Clementine and Balady displayed anthers longer than stigma.

The fruit qualitative characteristics of the 13 mandarin varieties were illustrated in **Table (4)**. In this regard, most of the varieties under the study showed either pyriform, obloid fruit shape. While the varieties Clementine, Abd El-Razik and Sonbol showed sheroid fruit shape. None of the varieties proved, oblique, or ovoid fruit shape.

The categories under which shape of fruit base was evaluated and showed a wide range of shape of fruit base, only the Centra and Malawy showed collard with neck and concave collared, respectively. While the shape of fruit base of Clementine and Abd El-Razik was convex. Truncate was characteristic of Sonbol, Chine mandarine, Aswan, Sayed Marri and El-Shorbagee varieties. The varieties Celiopatra, Tangarine Dancy and Balady varieties revealed necked fruit base shape. Regarding shape of fruit apex, none of the varieties showed mammiform, acute and rounded. All the variety under the study showed both truncate and depressed shape of fruit apex.

The fruit flavido color included 12 colors ranged from green to red orange. Celiopatra and Tangarine Dancy had dark orange epicarp color while that of the Abd El-Razik, Sonbol, Aswan and El-Shorbagee varieties were dark yellow, light orange, green yellow and yellow, respectively **Table (4).**The rest of the varieties exhibited orange fruit epicarp color.

The Centra and Sayed Marri varieties were the only ones showed rough texture of flavido surface**, Table (4)** Clementine, Satsuma, and Balady

proved papillate epicarp surface texture, whereas Abd El-Razik, Malawy and Chine mandarin showed pitted. The rest of the varieties had smooth surface texture of epicarp surface. Only the variety Abd El-Razik presented strong adherence of mesocarp to endocarp, whereas the Clementine, Tangarine Dancy, Chine mandarin and El-Shorbagee demonstrated medium one. The remaining varieties exhibited weak adherence of mesocarp to endocarp. All the varieties showed an intermediate density (45-65/ cm^2) of oil glands on fruit surface. The variety Clementine only proved low density (<40.cm^{-2}). In contrast, the varieties Celiopatra, Aswan and Balady demonstrated high density (>70.cm^{-2}) of oil glands on fruit surface. The majority of the varieties exhibited an orange color of fruit pulp while, both of Abd El-Razik and Sonbol showed Yellow. Red orange of fruit pulp was in Tangerine Dancy variety only.

Table (5) illustrate the qualitative characteristics of fruit carples and seed. As to number of segments per fruit, the common of the varieties showed 10-14 segments per fruit While the Clementine, Centra, Sonbol and Balady varieties demonstrated only 5-9 segments per fruit. Both of Clementine and Balady displayed thick and medium segment thick wall respectively. Whereas the rest of the varieties revealed thin thickness of segment walls **Table (5).**

The Clementine, Centra, Abd El Razik, Aswan and El-Shorbagee demonstrated 10-19 seeds per fruit. The highest counted number of seeds per fruit (20-50) was marked in the varieties Celiopatra and Sayed Marri. In contrast, Satsuma and Malawy illustrated with fruit free seeds. The varieties Tangerine Dancy, Chine mandarine, and Balady showed 5-9 seeds per fruit. The remaining of the variety (Sonbol) presented 1-4 seeds per fruit.

The seed shape was estimated for the different varieties, the Clementine, Chine mandarin, Aswan and El-Shorbagee varieties were the only ones presented ovoid seed shapes. Celiopatra, Centra, Tangerine Dancy and Sayed Marri showed a clavate for seed shape. Only three varieties namely the Abd El-Razik, Sonbol and Malawy showed Semi deltoid seed shape **Table (5).** Also, one variety (Balady) display fusiform seed shape.None of the varieties showed semi-spheroid and spheroid, cuneiform or ovoid seed shape.

All the varieties showed a smooth seed shape except Centra and Abd El-Razik demonstrated wrinkled seed surface.

Seed color for the investigated varieties revealed that the Balady and Malawy varieties proved brown and yellowish, respectively. Whereas the Aswan and El-Shorbagee had white seed. The residues varieties were cream seed color.

Sonbol, Malawy and Satsuma were the only varities to present a negligible number of seeds (average of 1-4 per fruit), one of characteristics mostly required for consumption. Another trait to be explored in directed crossings is the slight adherence of the epicarp to mesocarp, this trait is present in many of the varieties.

2. Pollen grain viability

The percentage of pollen grain fertility and sterility of the 13 mandarin varieties are presented in **Table (6)**.and **Figures (2 and 3)** demonstrates the external appearance of mandarin fertile and sterile pollen grain. Number of pollen grain examined ranged from 1245-3589 for all the studied varieties.

Table (6) showed that variety genotypes exhibited varied frequencies of pollen sterility and fertility. The highest pollen abortion was found in; Clementine, chine mandarin and Aswan, whereas the variety Celiopatra, Sayed Marri, El-Shorbagee and Sonbol showed the highest pollen fertility and lowest pollen aboration. Pollen sterility in variety Centra, Satsoma and Malawy was close to highest score found in variety Abd El-Razik, Tangerine Dancy and Balady mandarin.

The data in **Table (6)** showed that there were significant differences in mean frequency of pollen fertility and sterility between genotypes in almost cases. The reduced percentage of pollen fertility is some genotypes could be attributed to the effect the disturbance in chromosome during meiosis I, leading to genic imbalance and subsequently high frequency of pollen sterility.

Physico-chemical characteristics

Table (7) shows some physical and physico-chemical parameters of Mandarin accessions. pH in the studied varieties ranged between 3.12 in El-Shorbagee to 3.91 in the Abd El-Razek. Titratable acidity in the studied varieties ranged between 0.67 % with Sonbol to 3.88% in Chine mandarin. Generally, the highest acidity of Mandarin juice was reflected in the low pH of juice. In this work, the highest soluble solid content (SSC) (15.1°Brix) was found in the Abd El-Razek.

Table 5. Qualitative characteristics of fruit segment and seed of 13 mandarin varieties

Variety	Number of carpels per fruit	Thickness of carpel walls	Number of seeds per fruit	Seed shape	Seed surface	Seed color.
Clementine	5-9	Thick	10-19	Ovoid	Smooth	Cream
Celiopatra	10-14	Thin	20-50	Clavate	Smooth	Cream
Centra	5-9	Thin	10-19	Clavate	Wrinkled	Cream
Satsuma	10-14	Thin	None	None	None	None
Abd El-Razik	10-14	Thin	10-19	Semi deltoid	Wrinkled	Cream
Sonbol	5-9	Thin	1-4	Semi deltoid	Smooth	Cream
Tangerine Dancy	10-14	Thin	5-9	Clavate	Smooth	Cream
Malawy	10-14	Thin	1-4	Semi spheroid	Smooth	Yellowish
Chine mandarin cv.	10-14	Thin	5-9	Ovoid	Smooth	Cream
Aswan	10-14	Thin	10-19	Ovoid	Smooth	White
Sayed Marri	10-14	Thin	20-50	Clavate	Smooth	Cream
El-Shorbagee	10-14	Thin	10-19	Ovoid	Smooth	White
Balady	5-9	Medium	5-9	Fusiform	Smooth	Brown

Characteristics were described for citrus by the International Plant Genetic Resources Institute (1999).

Table 6. Characteristic of pollen fertility and sterility of 13mandarin varieties in Egypt

Variety	No. of pollen grains examined	No. of viable pollen grains	Mean % of pollen fertility	Mean % of pollen sterility
Clementine	1249	1144	91.59 M	8.41 A
Celiopatra	1520	1513	99.54 A	0.46 M
Centra	1871	1830	97.81 I	2.19 E
Satsuma	1202	1175	97.75 J	2.25 D
Abd El-Razik	1720	1689	98.20 G	1.80 G
Sonbol	3589	3550	98.91 D	
Tangerine Dancy	1245	1224	98.31 F	1.69 H
Malawy	2415	2368	98.05 H	1.95 F
Chine mandarin cv.	1639	1584	96.64 L	3.63 B
Aswan	2019	1957	96.93 K	3.07 C
Sayed Marri	1797	1787	99.44 B	0.56 L
El-Shorbagee	1620	1603	98.85 C	1.05 K
Balady	1409	1392	98.79 E	1.21 I

*Values within each column fallowed by the same letter are not statistically different at 5% level.

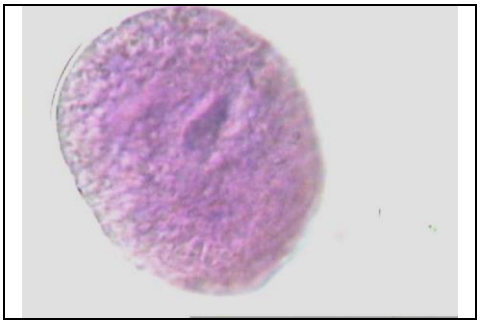


Figure 2.Fertile pollen grain in mandarin

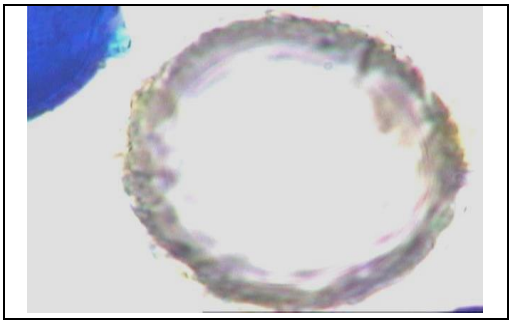


Figure 3. Sterile pollen grain in mandarin.

Table 7. Some Physical and physicochemical characteristics of mandarin varieties

Variety	pH	SSC °Brix	Acidity %	SSC/Acidity	Vit.C mg /100 ml
Clementine	3.77 ab	10.73 cde	0.75 de	14.3	22.04 cd
Celiopatra	3.74 ab	12.2 b	0.79 de	15.44	15.01 d
Centra	3.26 ef	11.5 bcd	1.23 bcde	9.34	29.3 bc
Satsuma	3.59 bcd	9.13 f	0.68 de	13.4	14.0 d
Abd-El-Razek	3.91 a	15.1 a	1.01 bcde	14.95	22.67 cd
Sonbol	3.66 bc	9.93 ef	0.67 e	14.82	38.61 ab
Tangerine Dancy	3.51 cd	12.3 b	1.23 bcde	10	28.56 c
Malawy	3.74 ab	12.66 b	1.45 b	8.73	44.82 a
Chine mandarine	3.23 f	11.8 bcd	3.88 a	3.04	15.99 d
Aswan	3.67 bc	10.75 cde	0.88 cde	12.2	22.76 cd
Sayed marri	3.26 ef	10.55 de	1.25 bcd	8.44	30.38 bc
El-Shorbagee	3.12 f	9.95 ef	1.4 bc	7.1	38.64 ab
Balady	3.42 de	11.86 bc	1.05 bcde	11.3	28.1

*Values have the same letter(s) in the same column, are not significantly different at 5% level using Duncan's Multiple Range Test.

The lowest soluble solids content (9.13°Brix) was found in Satsuma. Ascorbic acid was determined by HPLC. In the present survey, ascorbic acid content varied from 14.0 to 44.82 mg/100ml **Table (7).**

It had been observed an important influence of the genotype on the ascorbic acid content. In this work, the highest Ascorbic acid content (~38.61 mg/100ml) was found in the Sonbol, El-Shorbagee and Malawy. There were no significant differences found between these three different kinds of varieties. The lowest Ascorbic acid content (14.0 mg/100ml) was found in Satsuma.

Vitamin C being the major contributor to the antioxidant activity of citrus fruits, whereas sweetness has high correlation with T.S.S. and pH **(Fernandez *et al* 2004)**. Vitamin C is a natural antioxidant that may inhibit the development of major oxidative human conditions. In conclusion, the HPLC methods allow a fast, quantitative and reproducible determination of important bioactive compounds in Mandarin.

Conclusions

1- Despite the fact that the morphologic and agronomic variability is not extensive, good background material exists for breeding programs.
2- Morphological variations do not coincide, therefore complicating attempts of selective improvement.

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